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Soil organic carbon pools and enzymatic activity in aggregate size fractions attributed by tillage and Fertilization in a rice-wheat system: A review

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Abstract

Labile organic carbon (LOC) fractions are considered as sensitive indicators of change in soil quality and can serve as proxies for soil organic carbon (SOC). Although the impact of tillage, fertilization and crop residue management on soil quality is well known, less is known about LOC and SOC dynamics in the rice-wheat production systems in North West India. Tillage was the main factor to influence SOC and LOC fractions under the rice-wheat cropping system in North West India. NT increased SOC and all LOC fractions compared to CT. An increase in residue retention led to an increase in microbial biomass carbon (MBC). Plots under NT-NT had about 10% higher coarse (250–2000 μm) intra-aggregate particulate organic matter-C (iPOM-C) within >2000 μm sand free aggregates in the 0- to 5-cm soil layer compared with CT-CT plots. The fine (53–250 μm) iPOM-C within the 250- to 2000- μm aggregates was also higher in the continuous NT plots compared with CT within both >2000 and 250 to 2000 μm sand free aggregate size classes in that soil layer. Macro-aggregates (>0.25 mm) constituted 32.5–54.5% of total water stable aggregates (WSA) and were linearly related ($R^2 = 0.69$) to soil organic carbon content. The addition of rice straw and FYM significantly improved the formation of macro-aggregates with a concomitant decrease in the proportion of micro-aggregates at all the three sampling depths (0–5, 5–10 and 10–15 cm). Macro-aggregates had higher C and N density as compared to micro-aggregates. Application of rice straw and FYM improved C and N density in different aggregate sizes and the improvement was greatest in plots that received both rice straw and FYM each year. Application of FYM along with inorganic fertilizer resulted in a net C sequestration of 0.44 t ha⁻¹ in the plough layer of rice-wheat cropping.

Soil C pools (very labile, labile, less-labile and non-labile) were significantly higher under no-till dry-seeded rice (NTDSR)-NTW+RR cycle than conventional-till puddled transplanted rice-CTW. Macro-aggregates (>0.25 mm) had higher labile C pools and enzyme activities than micro-aggregates. NTW+RR significantly increased soil C pools within both macro- and micro-aggregates. Compared with CTW, NTW+RR increased soil dehydrogenase, cellulase and alkaline phosphatase activities by 23%, 34% and 14%, and water-soluble organic C by 31%, and increased water-stable aggregates and mean-weight-diameter. NTDSR-NTW+RR increased SOC, enzyme activity, and aggregate stability. Soil labile-C pools across aggregate fractions were the most sensitive indicators of soil quality when determining the effects of changes in management practices. Total water stable aggregates (WSA) ranged between 69.8-91.2% in which 0.1-0.053 mm aggregate fraction contributed (2.11-3.87%), whereas 0.25-0.5 mm aggregate fraction was having the highest (27.3-32.6%) contribution. The activities of enzymes in whole soil as well in aggregate fractions were lowest in control and highest in FYM+NPK.

Keywords: Total organic carbon, enzymatic activity, soil aggregate

Introduction

Soil organic carbon (SOC) is one of the central carbon pools (Yu *et al.*, 2017) [77] regarded as the centre of soil quality and its functions and a leading indicator of soil health (Lal, 2007; Zhao *et al.*, 2016) [31, 78]. The depletion of SOC leads to poor soil aggregation and stability, loss in water holding capacity, fertility, enzymatic activities and soil biology (Ghani *et al.*, 2003) [22]. Labile organic matter in soil mainly originates from the decomposition of plant and faunal biomass, root exudates, and deceased microbial biomass (Bolan *et al.*, 2011) [9]. Labile carbon is the SOC pool which is directly available for microbial activity and, hence, is considered to be the primary energy source for microorganisms (Haynes, 2005) [27]. Addition of organic matter as fertilizer (Gattinger *et al.*, 2012) [21] and reduced tillage will likely increase labile organic carbon (Cooper *et al.*, 2016) [13]. In addition, these practices have the potential to enhance carbon and nitrogen cycling as well as soil aggregation, which are one of the primary

mechanisms through which organic carbon is sequestered in soil (Panettieri *et al.*, 2015) [47]. Therefore, labile carbon has potential as an indicator of soil functions, in particular: nutrient cycling, soil aggregate formation, carbon sequestration (typically derived from changes in total organic carbon content) and habitat provision for biodiversity.

The effects of chemical fertilizers and organic amendments on soil microorganisms have been given particular attention. A meta-analysis of long-term inorganic fertilizer trials revealed a 15.1% increase of the microbial biomass after mineral fertilizers application compared to unfertilized treatments (Geisseler and Scow, 2014) [23]. Eo and Park, (2016) [19] also found that inputs of nitrogen (N) and phosphorus (P) fertilizers had considerable effects on specific bacterial groups. Furthermore, it is generally accepted that organic fertilizers have more significant effect on abundances of microorganisms in soils compared with mineral fertilizers (Wu *et al.*, 2011; Li *et al.*, 2015) [73, 32]. Ngosong *et al.* (2010) [46] observed that organic manure increased fungal abundance especially that of arbuscular mycorrhizal fungi (AMF). Moreover, Elfstrand *et al.* (2007) [18] also found higher fungi/bacteria ratios (F/B) in soils receiving green manure. Changes in microbial community structures in turn have important implications for the SOC mineralization. For example, Lipson *et al.* (2009) [34] stated that bacteria had higher growth rates and lower yields than fungi, suggesting a more important role for bacteria in determining soil heterotrophic respiration. However, Dai *et al.* (2016) [14] reported that alterations in soil microbial abundance and community composition did not significantly influence the C mineralization under long-term fertilization in paddy soils. The relationships between soil microbial community composition and function are not always straightforward because of the existence of several microbial groups that carry out similar functions and the complexity of soil system (Nannipieri *et al.* 2003) [41]. Thus, uncertainties still remain about the impacts of mineral and organic fertilization on soil microbial communities and their roles in SOC mineralization. Extensive tillage and removal of crop residue either as feedstock or as most prevalent practice of in-situ burning after mechanical harvesting of crops in the Indian IGPs (Beri *et al.*, 2003) [7], has raised soil health and environment threatening concerns due to large emissions of greenhouse gases (GHGs) (Singh *et al.*, 2020) [60]. The TOC is the most important component in maintaining soil quality due to its role in improving physical, chemical and biological properties of the soil (Sharma *et al.*, 2019a) [52] and crop productivity (Thind *et al.*, 2019) [67]. Because of the historic losses of organic C from the croplands (Singh and Benbi, 2018b) [57], most of the cultivated soils are exhausted of TOC and are far away from saturation (Vaccari *et al.*, 2011) [69]. But the capacity of soils to sequester C depends on amount of C input into the soil from plant productivity, relative to its export that is controlled by microbial decomposition (Singh and Benbi, 2020a) [58]. The management practices that ensure greater amounts of C returned to the soil is expected to cause a net build-up of TOC stock (Singh and Benbi, 2020b) [59]. Maintaining TOC stocks is important for sustainable agricultural development and the mitigation of global warming by reducing C emissions (Singh and Benbi, 2020a) [58].

Soil microbial biomass and enzymatic activity are the potential indicators of soil quality that rapidly responds to management and environmental induced changes

(Mohammadi, 2011) [40]. All crop residues' C passes through soil microbial biomass at least once, and thereby transferred from one C pool to another and intern lost as carbon dioxide (CO₂) (Ryan and Aravena, 1994) [50]. The un-decomposed crop residue remains in soil contributes toward C sequestration (Majumder and Kuzyakov, 2010) [37]. In fact decomposition pattern of rice residues is related to its quality, with non-cellulosic polysaccharides and hemi-celluloses being fast degradable rates are decomposed during the initial phase (Thevenot *et al.*, 2010) [66], while the stable/resistant C in lignin and cellulose are decomposed later in second phase (Majumder and Kuzyakov, 2010) [37]. Although, degradation and transformation of cellulose and lignin are highly complex, extra-cellular enzymatic activity of β -glucosidases, cellobiohydrolases and endo-glucanases enzymes had the high cellulose degradation abilities (Baldrian and Valášková, 2008) [3], while lignin degradation is caused by laccases, lignin peroxidases and Mn-peroxidases enzymes (Higuchi, 2004) [28]. Soil organic C is a heterogeneous mixture of organic materials comprising labile and stabilized (i.e., recalcitrant) organic C pools based on variable turnover times (Singh and Benbi, 2018a) [56]. The dynamics of TOC is described by partitioning the soil organic matter (SOM) into physical, chemical and biological pools (Banger *et al.*, 2010) [4]. The change in C input to soil can rapidly persuade TOC pools *viz.* potassium permanganate oxidizable C, water extractable organic C (Benbi *et al.*, 2016), hot water C (Singh and Benbi 2018b) [57], and microbial biomass C (Chan *et al.*, 2001). Labile C fractions act as sensitive indicators to soil management induced changes in TOC pool under short to medium term effects (Benbi *et al.*, 2016) [55]. These pools have shorter turnover times and are more sensitive indicators to soil management practices compared with TOC (Chen *et al.*, 2016) [10]. Conversely, the recalcitrant pools are difficult for soil microorganisms to decompose and have relatively long turnover times (Yan *et al.*, 2007) [74]. The review study was therefore; investigate the effect of fertilizer application and tillage practices on (1) C input through above-and below-ground plant biomass (2) change in soil enzymatic activity and TOC pools, and (3) C sequestration potential of tillage and fertilization under RWCS. We aimed at identifying organic C pools influenced due to changed soil enzymatic activities to assess ability of rice residue management/ tillage options to sequester C and suggest long-term best agricultural sustainability options.

Soil Organic Carbon Pools

Mamta Kumari *et al.* (2011) [38] revealed that macro-aggregates increased under a ZT rice and wheat rotation with the 2- to 4-mm fraction greater than that of the 0.25- to 2-mm fraction. Bulk and aggregate associated C increased in ZT systems with greater accumulation in macro-aggregates. The fine (0.053–0.25 mm) intra-aggregate particulate organic C (iPOM-C), in 0.25- to 2-mm aggregates, was also higher in ZT than conventional tillage. A higher amount of macro-aggregates along with greater accumulation of particulate organic C indicates the potential of ZT for improving soil C over the long-term in rice-wheat rotation. The aggregate-associated SOC concentration in different soil layers was influenced by tillage systems. In the 0.00-0.05 m layer, SOC concentration in macro-aggregates showed the order of NT+S>MP+S = NT-S>MP-S, whereas the NT system was superior to the MP system. However, the NT system

significantly reduced the SOC concentration in the 2.00-0.25 mm fraction in the 0.05-0.20 m layer. A similar trend was observed in the 0.25-0.053 mm fraction in the 0.20-0.30 m layer. Across all the soil layers, there was no difference in the <0.053 mm fraction between NT-S and MP-S, as well as between NT+S and MP+S, indicating that the NT system did not affect the SOC concentration in the silt + clay fraction. In average across the soil layers, the SOC concentration in the macro-aggregate was increased by 13.5% in MP+S, 4.4% in NT-S and 19.3% in NT+S, and those in the micro-aggregate (<0.25 mm) were increased by 6.1% in MP+S and 7.0% in NT+S compared to MP-S. For all the soil layers, the SOC concentration in all the aggregate size classes was increased with straw incorporation, by 20.0, 3.8 and 5.7% under the MP system, and 20.2, 6.3 and 8.8% under the NT system. The higher proportion of >2 mm aggregates and lower proportion of <0.053 mm aggregates under NT systems might be the result of the higher soil hydrophobicity, low intensity of wetting and drying cycles, higher soil C concentration or the physical and chemical characteristics of large macro-aggregates making them more resistant to breaking up (Vogelmann *et al.*, 2013)^[70].

Sapkota *et al.* (2017)^[51] revealed that the effects on SOC stock were significant at 0–0.05 and 0.05–0.15 m soil depths only. At 0–0.05 m, ZTDSR-ZTW+R and PBDSR-PBW+R, on an average, had significantly higher SOC stocks, that is 2.4 t/ha more than CTR-CTW. ZTDSR-ZTW, ZTDSR-ZTW+R and PBDSR-PBW+R had a similar improvement in total SOC at 0.05–0.15 m, which was significantly higher (by about 2.0 t/ha) than for CTR-CTW. All the treatments had similar SOC stocks at 0.15–0.3 m and 0.3–0.6 m soil depths. Calculations for the whole 0–0.6 m depth showed that ZTDSR-ZTW+R and PBDSR-PBW+R contained 5.6 t and 3.9 t/ha more SOC than CTR-CTW, respectively.

Samal *et al.* (2017)^[63] reported that full CA recorded significantly higher TOC stock ($47.71 \pm 2.46 \text{ Mg C ha}^{-1}$ soil) as compared to other scenarios in the total depth of soil studied. On the contrary, S4 (diversified cropping system with high cropping intensity) showed significantly lower C stock ($39.33 \pm 2.40 \text{ Mg C ha}^{-1}$) than all other scenarios. On an average, TOC stock in different scenarios follows the order: S3 (47.71 ± 2.46) > S2 (43.91 ± 0.84) > S1 (41.65 ± 0.13) > S4 ($39.33 \pm 2.40 \text{ Mg C ha}^{-1}$ soil). Maximum accumulation of SOC ($19.41 \pm 1.84 \text{ Mg C ha}^{-1}$) in top depth of soil was observed under S3 followed by S4 ($16.56 \pm 1.71 \text{ Mg C ha}^{-1}$), S2 ($16.53 \pm 0.78 \text{ Mg C ha}^{-1}$) and S1 ($16.22 \pm 0.60 \text{ Mg C ha}^{-1}$) and SOC accumulation reduced in lower depths. In 10–20 cm depth significantly low SOC was observed in S4 ($12.61 \pm 0.10 \text{ Mg C ha}^{-1}$) and statistically at par values of SOC were obtained in rest scenarios (S1–S3). In 20–30 cm soil depth significantly greater SOC accumulation was recorded in S2 ($12.82 \pm 1.10 \text{ Mg C ha}^{-1}$) and S3 ($13.10 \pm 0.21 \text{ Mg C ha}^{-1}$) in comparison to S1 ($10.36 \pm 1.07 \text{ Mg C ha}^{-1}$) and S4 ($10.16 \pm 0.80 \text{ Mg C ha}^{-1}$).

Li *et al.* (2018)^[33] reported that after nine years application of organic fertilizers, straw with manure (NPSM) or straw only (NPS) substantially increased SOC content by 143% and 71%, respectively, while application of chemical fertilizers

alone did not affect SOC level compared with that of CK (Fig. 1a). The effects of fertilization on soil labile organic C showed a similar trend to total SOC. The contents of DOC, LFOC, and MBC were respectively 264%, 108%, and 102% higher after NPSM application, and respectively 57%, 82% and 38% higher after NPS application than compared with those of CK. The C/N ratio of bulk soil was constant across all fertilization treatments, but C/N ratio of labile organic C fractions had differential responses to the different treatments (Fig. 1a). Ratios of DOC/DON and LFOC/LFN were lower in treatments with additions of exogenous organic amendment and chemical fertilizers than in the control. In contrast, the MBC/MBN was 19% higher under NPSM application than compared to that of CK.

Changes in labile organic C fractions can respond to soil management practices more quickly than total SOC content (Gong *et al.*, 2009)^[24]. It has been widely accepted that application of organic manure markedly increases labile organic C fractions (Ding *et al.* 2012; Wang *et al.*, 2015)^[15, 72]. The DOC is mobile within the soil solution and is thus considered to be the most bioavailable source of C substrates for microbial populations (Cookson *et al.*, 2005)^[12]. However, Li *et al.* (2018)^[33] suggested that application of chemical fertilizers alone had no significant effect on DOC, confirming that the primary source of DOC was organic amendments. Moreover, the NPSM treatment resulted in a sharp elevation of DOC compared to that of the NPS, which could be explained possibly by the presence of a considerable amount of soluble materials in manure amendments (Long *et al.*, 2015)^[35]. The LFOC represents the slightly decomposed plant litter and functions as nucleation sites for fungi and other soil microbes (Dorodnikov *et al.*, 2011)^[16].

Benbi and Senapati, (2010)^[5] also found that across fertilizer N rates, incorporation of rice straw increased the occurrence of macro-aggregates by 100% in the >2 mm size, by 49% in 1–2 mm size, by 60% in the 0.5–1 mm size and by 15% in the 0.25–0.50 mm size over control at the 0–5 cm soil depth (Fig. 1b). The effect of FYM was slightly more and its application increased the amount of macro-aggregates of different sizes by 18–105%. The formation of macro-aggregates was maximum in plots where both rice straw and FYM were applied and the increase over control ranged between 23 and 120% for different aggregate sizes. Application of rice straw and FYM decreased the occurrence of micro-aggregates in the top 5 cm soil by 5–10%. In all the organic matter treatments the macro-aggregate density decreased with soil depth (Fig. 1b). In the 5–10 cm soil depth, application of rice straw improved the macro-aggregate density over unfertilized control by 15–84%. The increase in the macro-aggregate density ranged between 17 and 95% in FYM amended plots and between 30 and 100% in rice straw + FYM amended plots. The effect of addition of organic sources on macro-aggregation in the 10–15 cm soil depth was small and ranged between nil and 35% for the three organic matter treatments. For the data pooled across treatments and sampling depths, the amount of macro-aggregates was linearly related to SOC content (Fig. 1b).

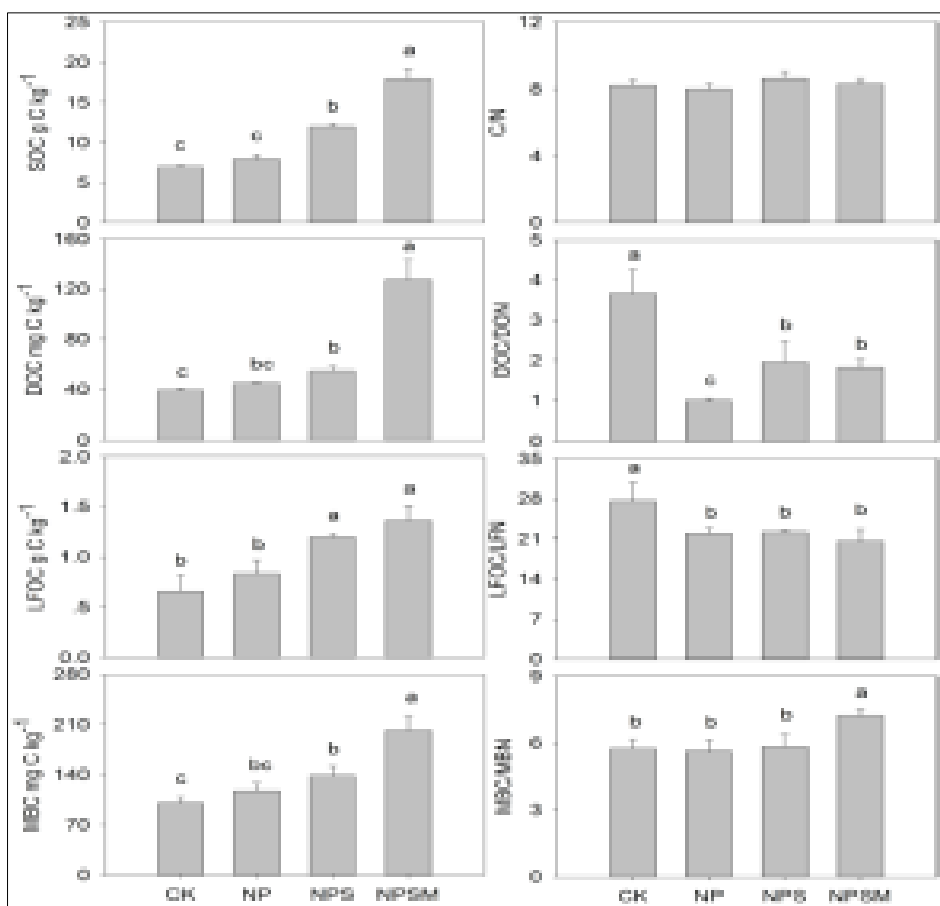


Fig 1a: Organic C contents and C/N ratios of bulk soil and labile fractions under different fertilization regimes [Source: Li *et al.*, 2018] ^[33]

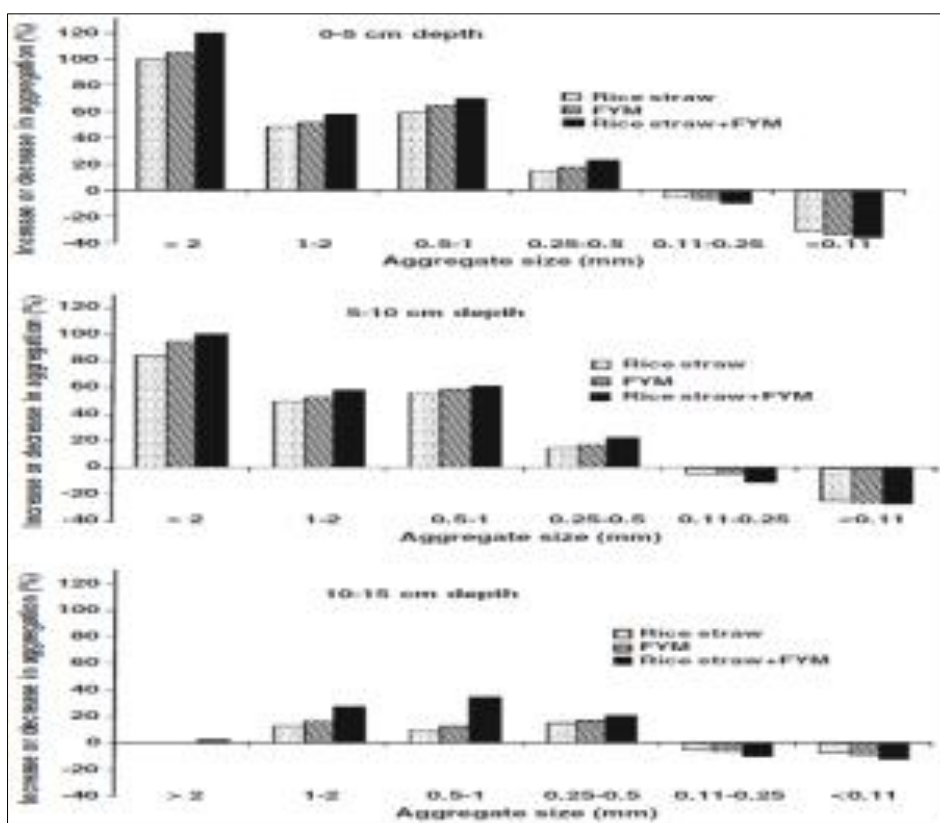


Fig 1b: Influence of rice straw and farm yard manure (FYM) application on improvement in the occurrence of macro-aggregates and decrease in the proportion of micro-aggregates over unfertilized control at three soil depths after 7 years of rice-wheat cropping in northwest India [Source: Benbi and Senapati, 2010] ^[5]

Naresh *et al.* (2016) [43] also found significantly higher POC content was probably also due to higher biomass C. PON content after 3-year showed that in 0-5 cm soil layer of CT system, T₁ and T₅ treatments increased PON content from 35.8 mgkg⁻¹ in CT (T₀) to 47.3 and 67.7 mg·kg⁻¹ without CR, and to 78.3, 92.4 and 103.8 mgkg⁻¹ with CR @ 2, 4 and 6 tha⁻¹, respectively. The corresponding increase of PON content under CA system was from 35.9 mgkg⁻¹ in CT systems to 49 and 69.6 mgkg⁻¹ without CR and 79.3, 93.0 and 104.3 mgkg⁻¹ with CR @ 2, 4 and 6tha⁻¹, respectively. Small improvement in PON content was observed after 4 years of the experiment. Singh *et al.* (2014) found that carbon stock of 18.75, 19.84 and 23.83Mg ha⁻¹ in the surface 0.4 m soil depth observed under CT was increased to 22.32, 26.73 and 33.07Mg ha⁻¹ in 15 years of ZT in sandy loam, loam and clay loam soil. This increase was highest in clay loam (38.8%) followed by loam (34.7%) and sandy loam (19.0%) soil. The carbon sequestration rate was found to be 0.24, 0.46 and 0.62 Mg ha⁻¹yr⁻¹ in sandy loam, loam and clay loam soil under ZT over CT.

Trivedi *et al.* (2021) [68] also found that the value was 33 and 92% greater than only minerally fertilized (NPK) and unfertilized control plots, respectively, whereas micro-aggregate-associated C was highest in plots with FYM + P'K' and lime (FYM + P'K' + L), which was 48 and 183% more than unfertilized control and NPK plots, respectively. Inside soil micro-aggregates, plots under FYM + P'K' had highest labile C, while NPK + L plots had highest recalcitrant C. Plots treated with FYM + P'K' had maximum intra-aggregate particulate organic matter within micro-aggregates inside macro-aggregates (iPOM_mM), which was 28 and 74% higher than NPK and unfertilized control plots, respectively. Total C stock inside the protected micro-aggregates within macro-aggregates was maximum for FYM + P'K' plots. It had 38, 67, and 171% higher C stock than NPK, FYM, and

unfertilized control plots, respectively (Fig. 2a).

Kubar *et al.* (2018) [30] reported that Tillage and straw returning treatments had a significantly higher SOC content than the other treatments in both soil depths (Fig. 2b). The greater SOC content in the 0-20 cm was observed under straw returning plots NTS and CTS treatments throughout 3 yr (2013-2015). In year wise comparison, NTS treatment had comparatively higher results as compared to CT, which increased from 15.7 to 18.8 g kg⁻¹ (2013), 15.8 to 19.0 g kg⁻¹ (2014), and 16.2 to 20.2 g kg⁻¹ (2015) in 0-20 cm depth (Fig. 2b). However, a variable trend was observed between the treatments in 20-40 cm depth, which were changed from 3.2 to 5.0 g kg⁻¹ (2013), 3.9 to 5.2 g kg⁻¹ (2014), and 4.8 to 6.1 g kg⁻¹ (Fig. 2b). The significant interactive effect of the straw returning and tillage treatments on the SOC content was obtained at the 0-20 cm for the whole soil profile (0-40 cm), during the 3 yr the trend was NTS > CTS > CT > NT. The average SOC content indicates that NTS significantly increased the SOC content the 0-20 cm depth in relative to the other three treatments. Singh *et al.* (2016) [55] discussed that crop establishment techniques and residue management significantly increased SOC after 5 yr at 0-15 and 15-30 cm depths. Higher SOC content under straw returning plots NTS and CTS treatment in the surface layer may be associated higher inputs of straw residue, which resulting the high retention of SOC content in surface soil (Gathala *et al.*, 2015; Naresh *et al.*, 2017) [20]. The higher SOC content may also be because of the interactive effect of tillage and straw returning, due to higher conversion efficiency of straw residue C into SOC (Al-Kaisi *et al.*, 2014; Naresh *et al.*, 2020) [1, 45]. The diverse studies also suggest that tillage influences on SOC can be depended on regional factors, i.e., soil type, residue management, crop rotation, region, type of clay and land management practices (Rabbi *et al.*, 2015) [49].

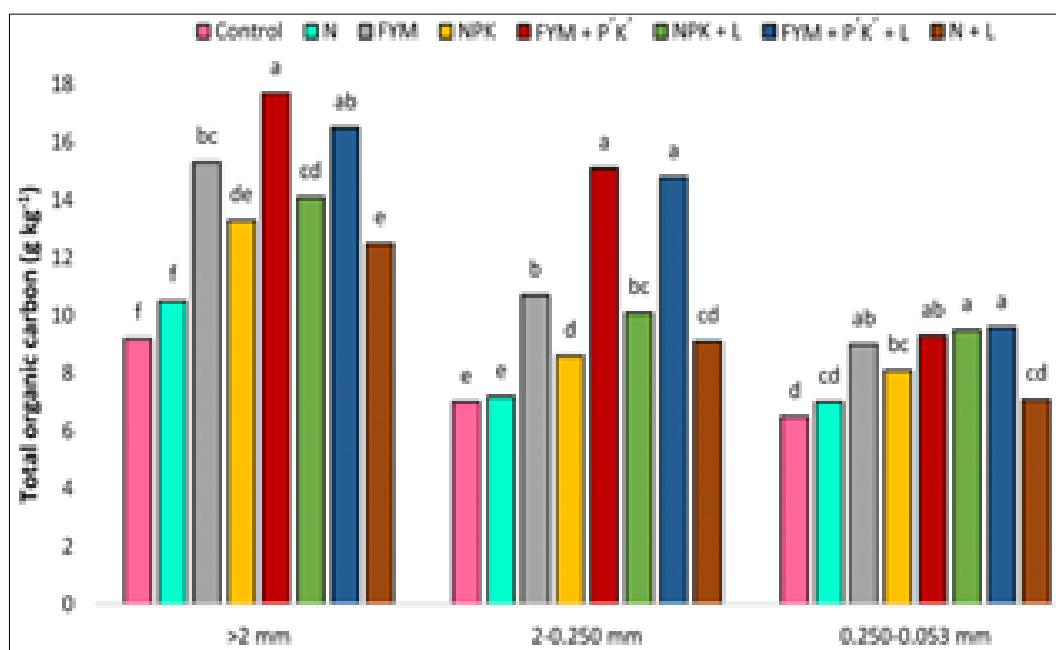


Fig 2a: Fertilization and liming impacts on soil organic carbon stabilization [Source: Trivedi *et al.*, 2021] [68]

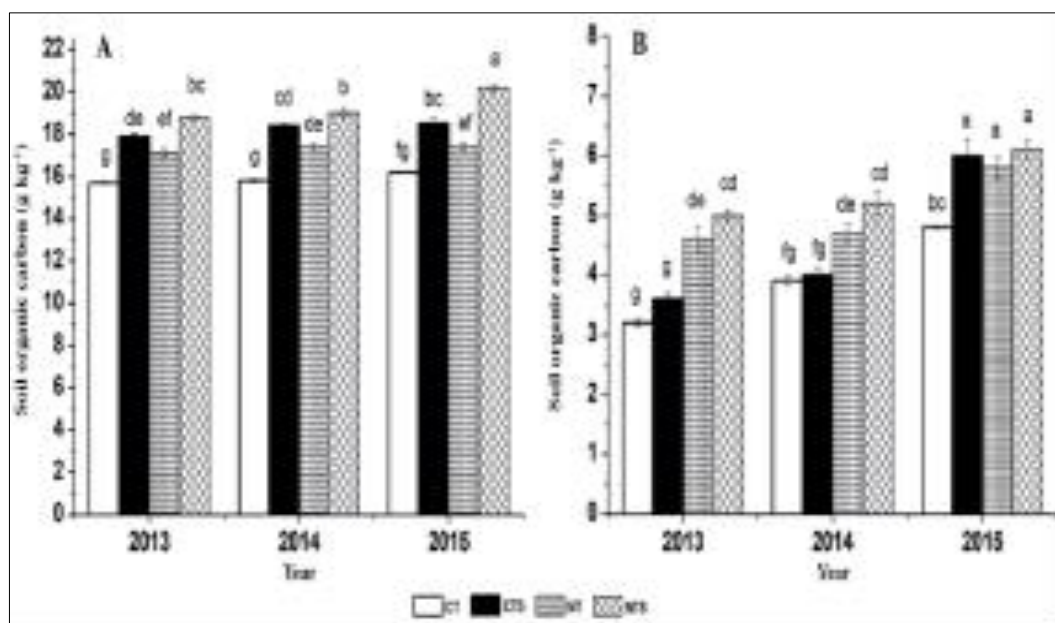


Fig 2b: Soil organic carbon content in different treatments. CT: Conventional tillage, CTS: conventional tillage with straw returning, NT: no-tillage, NTS: no-tillage with straw returning [Source: Kubar *et al.*, 2018]^[30]

Awanish, (2016)^[2] reported that the greater variations among carbon fractions were observed at surface layer (0-5 cm). F₁=very labile, F₂=labile, F₃=less labile and, F₄=non-labile. At this depth, C fraction in vertisols varied in this order: F₄>F₁>F₂=F₃. Below 5 cm, the carbon fraction was in the order: F₄>F₁>F₃>F₂. For 15-30 cm depth it was in the order F₄>F₁>F₂>F₃. At lower depth, almost similar trend was followed as that of 30-45 cm. Regardless of tillage system, contribution of different fractions of carbon (C) to the TOC varied from, 33 to 41%; 9.30 to 30.11%; 8.11 to 26%; 30.6 to 45.20% for very labile, labile, less labile and non-labile fractions, respectively at 0-5cm depth. For subsurface layer (5-15cm), contribution of different fractions to the TOC varied from 27.8 to 40%; 7.80 to 12.40%; 11.11 to 19.0% 38.0 to 50.0% for very labile, labile, less labile and non-labile fraction, respectively. In general, C contents decreased with increasing depth, mainly for very labile fraction (F₁) which was contributing around 40% or more in surface and surface layers (0-5 and 5-15 cm) as compared to deeper layers (15-30 and 30-45 cm).

Dou *et al.* (2016)^[17] observed that application of N and NPK fertilizers resulted in no remarkable increases in SOC storage across all aggregates compared with CK, except for 2000-250 μ m aggregates in N-treated soils (Fig.3a). Long-term MNPK fertilizer strongly increased the SOC storage by an average of 466.0 g C m² in all aggregates. The SNPK fertilizer increased SOC by an average of 191.1 g C m² in macro-aggregates (> 250 μ m) but decreased it by an average of 131.4 g C m² in

micro-aggregates (250-53 μ m) compared with CK (Fig 3a). Besides, the SOC storage showed a decrease in 250-53 μ m aggregates compared with other aggregate sizes in the fertilized soils except for MNPK treatment. Generally, the SOC storage in macro-aggregates (> 250 μ m) was greater than in micro-aggregates (< 250 μ m) across the fertilizer treatments (Fig. 3a). Long-term fertilization resulted in no significant changes in the C:N ratios across all the aggregate sizes (Fig. 3a). Moreover, the higher C: N ratios occurred in LF while the lower C: N ratios occurred in mSOM among soil density fractions across all fertilization treatments (Fig. 3b). Long-term fertilization led to less negative δ 13C values in LF, iPOM, and mSOM compared with CK, and the least negative δ 13C values were found in SNPK-treated soil across all the soil fractions.

Additionally, higher C: N ratios of LF reflected more recent litter inputs, while mSOM with much lower C: N ratios suggested decreasing C: N ratios in soil C fractions have been associated with increasing SOM decomposition and mineral association (John *et al.*, 2005). Long-term application of MNPK fertilizer may be better for future soil C sequestration compared with other fertilizers. Additionally, the LF of SOM, as an early and sensitive indicator of the response to the long-term effects of agricultural practices (Yague *et al.*, 2012)^[75] indicated that improvement of SOM in MNPK-treated soils may be first ascribed to a decline of C/N ratios in LF (Hai *et al.*, 2010)^[26].

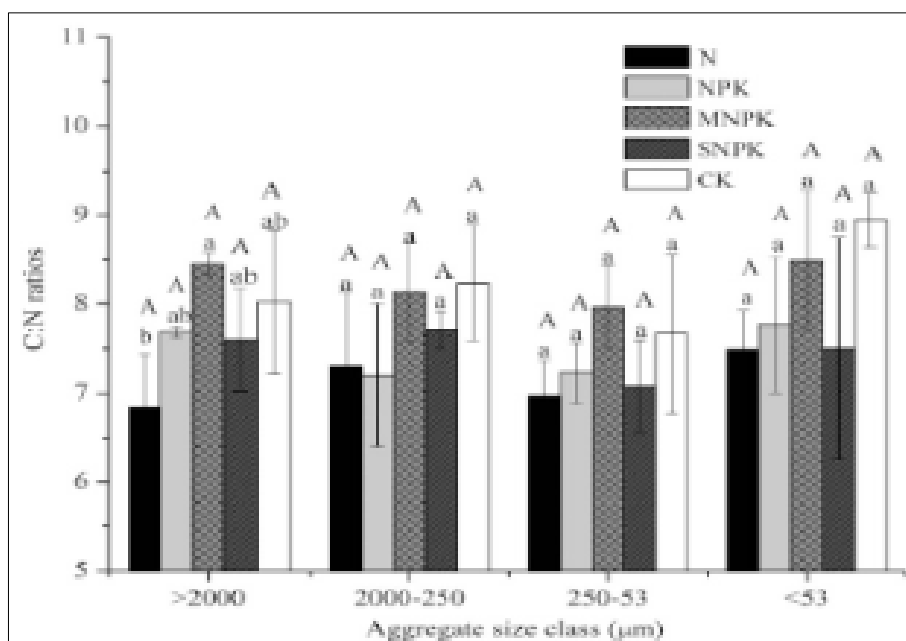


Fig 3a: C: N ratios of soil aggregate size classes separated from soils (0–20 cm) under long-term fertilization [Source: Dou *et al.*, 2016] [17]

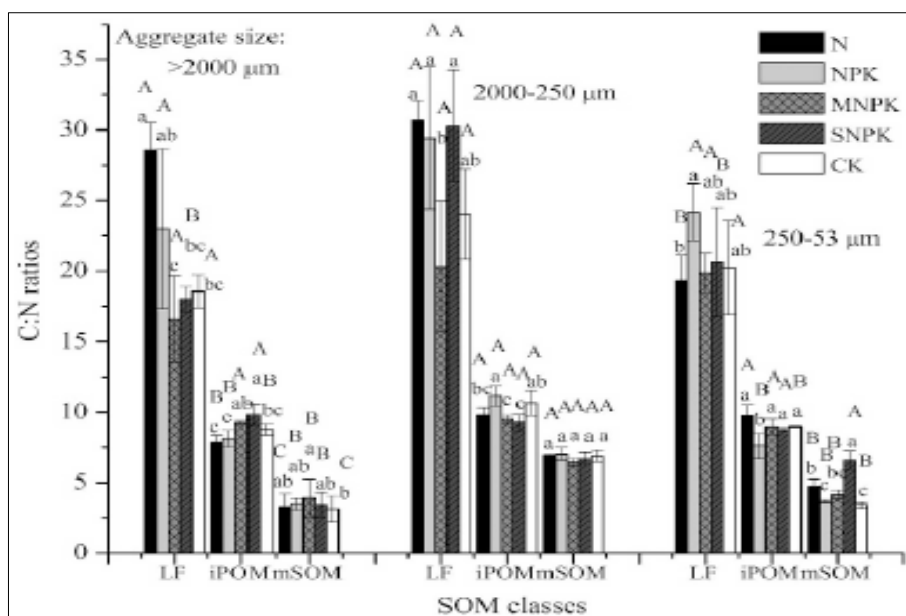


Fig 3b: C: N ratios of LF, iPOM and mSOM of aggregate size classes separated from the soils (0–20 cm) under long-term fertilization [Source: Dou *et al.*, 2016] [17]

Sheng *et al.* (2020) [54] also found that The OC concentrations of different density and humic fractions differed obviously among soil aggregate hierarchy and land use patterns. Specifically, within all aggregate fractions, the OC concentrations of all density and humic fractions were highest in grassland, followed by cropland and bareland. Also, the OC concentration showed no significant differences in cropland and bareland soils among aggregates, with the exception of higher OC of free light (fLF) and occluded light (oLF) fractions within micro-aggregate in cropland than those in bareland soil. The free light (fLF) and occluded light (oLF) fractions contained obviously higher OC concentrations than those in heavy fraction (HF) and humic fractions (Fig. 4a). Among all the field treatments, the OC concentrations of density fractions were highest within 0.25-2 mm aggregates, and then decreased with the size of the aggregates, with the

<0.053 mm silt-clay unit having the lowest OC concentrations (Fig. 4a). The OC concentrations of humic fractions within aggregates did not show the same changing trends as density fractions after 31-year land use changes. There were no significant differences of OC concentration of fulvic acids (FA) and humic acids (HA) among field soils, with the exception of higher OC concentrations of FA in >2 mm and 0.25–0.053 mm aggregate fractions in grassland than in cropland. The OC concentration of humin (HU) showed similar changes as HF, significantly higher OC concentrations were found within 0.25–2 mm and 0.053–0.25 mm aggregates in grassland (Fig.4a).

However, the proportions of density- and humic-associated OC in SOC differed obviously with OC concentration among land use patterns and soil aggregate hierarchy (Fig. 4b). Among all the field treatments, the HF and HU fractions,

containing lower OC concentrations, were the most abundant OC fractions relative to total SOC. The highest proportions were found within 0.25-2 mm aggregate in grassland and 0.053-0.25 mm aggregate in cropland and bareland (Fig. 4b). Specifically, within >2 mm and 0.25-2 mm macro-aggregate fractions, the proportions in SOC were significantly higher in all density and humic fractions in grassland than in the respective fractions of both cropland and bareland soils. The bareland soil had the lowest OC proportions in density and humic fractions, with the exception of higher occluded light fractions (oLF) within >2 mm macro-aggregate in bareland than that in cropland. Within 0.053-0.25 mm and <0.053 mm aggregates, higher OC proportions were found in bareland soil, followed by cropland, and grassland had the lowest proportion except for higher OC proportion of fLF in grassland than that of cropland and bareland (Fig. 4b). The proportions of OC in density and humic fractions to total

SOC were higher in large and small macro-aggregates in grassland while the proportions of OC in density and humic fractions in micro-aggregates and silt-clay units were lower in grassland than those in cropland and bareland soil. This suggested that not only plant residue were supposed to enhance C accumulation in macro-aggregates, but also did so for vegetation type reduction in tillage, or a combination. The proportions of OC in total SOC decreased in density and humic fractions within macro-aggregates, and increased within micro-aggregates and silt-clay units in bareland soil without any fresh OC amendments and in cropland soil with only limited root exudates. This indicated a predominant loss of labile and decomposable organic matter in the macro-aggregate fraction likely due to long-term substrate limitation and soil erosion (Simmons and Coleman, 2008). Also, the 0.25-0.053 mm micro-aggregates was considered as the “preferential aggregate” in cropland and bareland soils.

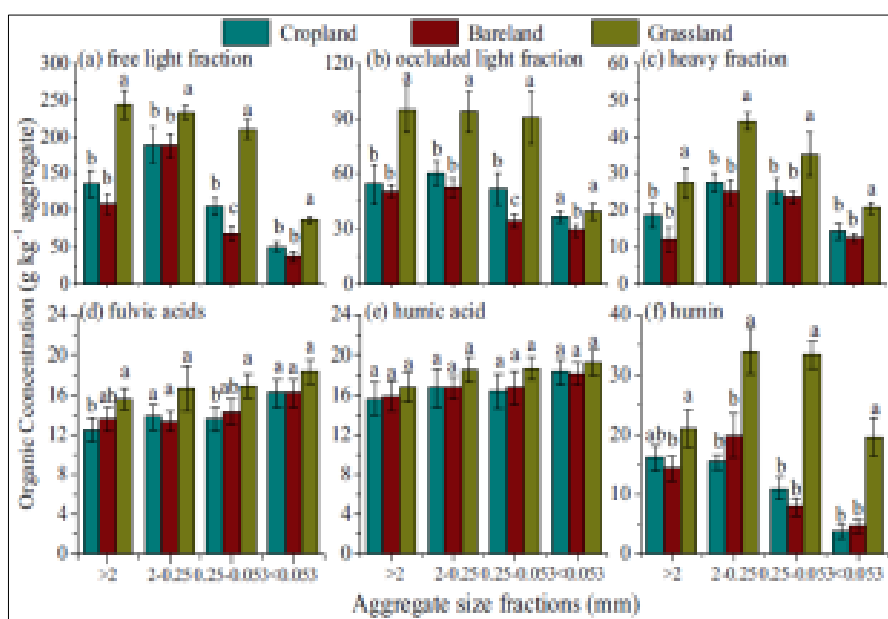


Fig 4a: Organic carbon concentrations of density (a–c) and humic fractions (d–f) within soil aggregate fractions after 31-year different land use patterns [Source: Sheng *et al.*, 2020] ^[54]

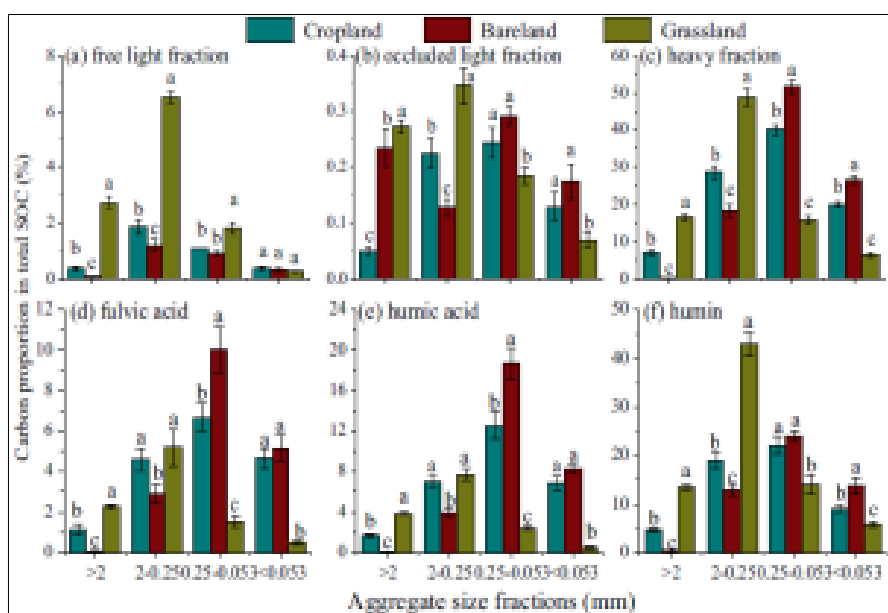


Fig 4b: Proportions of organic carbon of density (a–c) and humic fractions (d–f) to total SOC within different sized aggregates after 31-year different land use patterns [Source: Sheng *et al.*, 2020] ^[54]

Sharma *et al.* (2020) [53] observed that the TOC concentration varied between 4.37 and 6.44 g C kg⁻¹ in the surface (0–7.5 cm) and 3.50 and 5.98 g C kg⁻¹ in the sub-surface (7.5–15 cm) soil layers (Table 1). Alone fertilizer-N application (RS₀N₉₀, RS₀N₁₂₀, and RS₀N₁₅₀) did not significantly increase the TOC concentration in the surface soil layer, as compared with CK at both soil layers. Average across the treatments, TOC in the surface soil layer was ~18.4% higher, compared with sub-surface soil layer. The WEOC was the smallest

fraction constituted only ~0.6% of TOC in both the layers, although the concentration of WEOC was ~20.4% higher in the surface than the sub-surface soil layer. The WEOC concentration was significantly lower in CK, and increased by 14.9 and 14.6 mg C kg⁻¹ in the surface and sub-surface soil layers, respectively in RS_{10.0}N₁₅₀ treatment. The HWC constituting 4.9–5.2% of TOC pool was ~21% higher in the surface than the sub-surface soil layer.

Table 1: Effect of fertilizer-N application and rice straw (RS) incorporation on total organic carbon (TOC), water extractable organic carbon (WEOC) and hot water carbon (HWC) in surface (0–7.5 cm) and sub-surface (7.5–15 cm) soil depth under rice-wheat cropping system in north-western India [Source: Sharma *et al.*, 2020] [53]

Treatment	TOC (g kg ⁻¹)	WEOC (mg kg ⁻¹)	HWC (mg kg ⁻¹)	TOC (g kg ⁻¹)	WEOC (mg kg ⁻¹)	HWC (mg kg ⁻¹)
	Surface (0–7.5 cm) soil			Sub-surface (7.5–15 cm) soil		
CK (RS ₀ N ₀)	4.37a	24.8a	223.5a	3.50a	19.7a	177.6a
RS _{5.0}	4.69abc	27.0abcd	231.9a	3.59ab	20.4ab	177.4a
RS _{7.5}	4.88abc	27.5bcd	242.7ab	4.05abcd	22.5abc	202.7abc
RS _{10.0}	5.66cde	32.0e	294.6cde	4.56de	24.3bcd	227.8cd
RS ₀ N ₉₀	4.56a	25.9abc	225.3a	3.68abc	21.7abc	185.2ab
RS _{5.0} N ₉₀	4.65ab	25.5ab	232.1a	3.96abcd	22.3abc	201.5abc
RS _{7.5} N ₉₀	5.06abcd	28.1cd	257.3abc	4.05abcd	23.9abcd	214.1abcd
RS _{10.0} N ₉₀	5.94de	33.7ef	293.5cde	4.79def	26.1cd	241.0cd
RS ₀ N ₁₂₀	4.69abc	26.7abcd	230.7a	4.37bcd	24.9cd	216.5abcd
RS _{5.0} N ₁₂₀	5.61bcde	31.8fg	278.2bcd	4.42cde	25.2cd	222.2bcd
RS _{7.5} N ₁₂₀	5.94de	33.6ef	292.7cde	4.51de	25.7cd	222.4bcd
RS _{10.0} N ₁₂₀	6.44ef	36.2g	318.9de	5.20efg	28.2de	253.0d
RS ₀ N ₁₅₀	4.97abc	28.5d	249.2abc	4.60de	26.1cd	232.3cd
RS _{5.0} N ₁₅₀	6.26e	35.4e	293.0cde	5.38fgh	30.7ef	245.1cd
RS _{7.5} N ₁₅₀	6.58ef	36.5g	333.3ef	5.75gh	32.7f	290.9e
RS _{10.0} N ₁₅₀	6.37ef	39.7h	373.6f	5.96h	34.3f	297.3e

Values within a column, followed by different letters are significantly different at $p < 0.05$ by Duncan's multiple range test (DMRT).

Soil C mineralization

Yan *et al.* (2013) [76] observed that particulate organic C was found stratified along the soil depth. A higher POC was found in surface soil decreasing with depth. At the 0–20 cm, POC content under NP+FYM, NP+S and FYM were 103, 89 and 90% greater than under CK, respectively. In 20–40 cm and 40–60 cm soil layers, NP+FYM had maximum POC which was significantly higher than NP+S and FYM treatments. Even though POC below 60 cm depth was statistically similar among fertilization treatments, the general trend was for increased POC with farmyard manure or straw application down to 100 cm soil depth. Guo *et al.* (2016) [25] reported that in the 0–5 cm soil layer, NT treatments significantly increased SOC concentration by 5.8%, 6.8%, and 7.9% of bulk soil, >0.25 mm aggregate, and <0.25 mm aggregate, respectively, compared with CT treatments. NT treatments significantly increased MBC of bulk soil, >0.25 mm and <0.25 mm aggregates by 11.2%, 11.5% and 20.0%, respectively, compared with CT treatments. DOC concentrations of bulk soil, >0.25 mm aggregate, and <0.25 mm aggregate under NT treatments were 15.5%, 29.5%, and 14.1% higher than those under CT treatments, respectively. In comparison with NS treatments, S treatments significantly increased SOC concentrations of bulk soil by 12.8%, >0.25 mm aggregate by 11.3%, and <0.25 mm aggregate by 14.1%. In addition, MBC of bulk soil, >0.25 mm aggregate, and <0.25 mm aggregate under S treatments were 29.8%, 30.2%, and 24.1% higher than those of NS treatments, respectively. S treatments exhibited 25.0%, 37.5%, and 23.2% higher DOC

concentrations of bulk soil, >0.25 mm aggregate, and <0.25 mm aggregate compared with NS treatments, respectively. In the 0–5 cm soil layer, there were significant interactions of tillage and straw returning on SOC concentration of >0.25 mm and <0.25 mm aggregates, MBC of bulk soil and <0.25 mm aggregate, and DOC concentration of >0.25 mm aggregate. Conservation tillage significantly increased SOC concentration of bulk soil in the 0–5 cm soil layer.

Li *et al.* (2018) [33] observed that the cumulative CO₂-C emission over time tended to be higher in NPSM and NPS than in CK and NP treatments throughout the incubation period (Fig 5a). By the end of the incubation, the NPSM treatment had largest increase of the cumulative mineralization C (C_{min}) by 85%, and the NPS treatment also resulted in an increase of 53% compared to CK. Moreover, compared to CK, the NPSM and NPS treatments enlarged the size of the easily mineralizable C pool (C₀) by 103 and 78%, respectively. Likewise, mineralization rates of the resistant C pool (k_s) were higher in organic fertilization plots than in CK plots, though only significantly in NPSM fertilization plots. Mohanty *et al.* (2013) [39] reported that long-term application of farmyard manure significantly and positively affected C mineralization in a rice-rice system. The mineralization of SOC is directly governed by interactions between the effects of microbial biomass, microbial community structure, substrate quality and availability, and microclimates (Juarez *et al.*, 2013) [36]. The C contents of SOC and its labile fractions, and microbial community abundance and composition were crucial for C mineralization, however, the C/N ratios of total

soil and labile organic C fractions were less important for the C mineralization. These results were expected given that soil microbial community respiration is usually limited by the C substrates supply in the intensive agricultural systems. Witzgall *et al.* (2021)^[71] reported that particulate OM acts as an important precursor for the aggregate formation⁴¹ and parallel occlusion of litter-derived POM into aggregated soil structures (Fig. 5b). Regardless of soil texture, fresh litter surfaces serve as hotspots of microbial activity driving the

formation of organo-mineral associations in concert with comprising a nucleus for aggregate formation. Thus, the biogeochemical interfaces of decaying plant litter determine—via promoted microbial activity—the two most prominent mechanisms which increase the persistence of OC in soils; the (i) occlusion of POM in soil aggregates and (ii) the association of OM with mineral surfaces as simultaneous processes across soils of different structure (Fig. 5b).

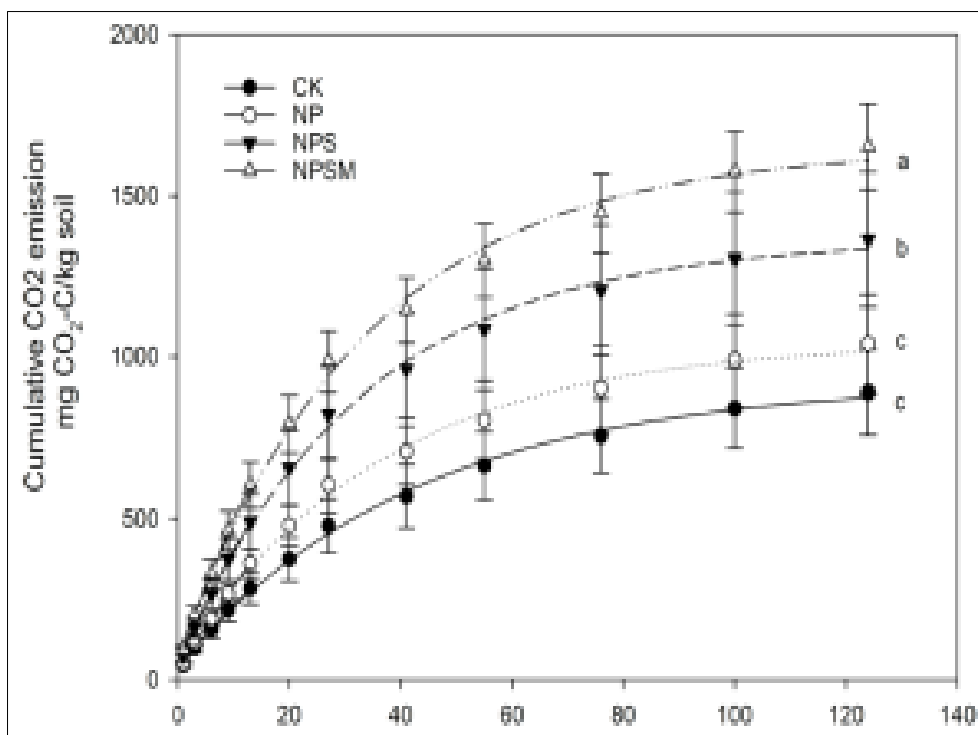


Fig 5a: Cumulative CO₂ emission over time under different fertilization regimes [Li *et al.*, 2018]^[33]

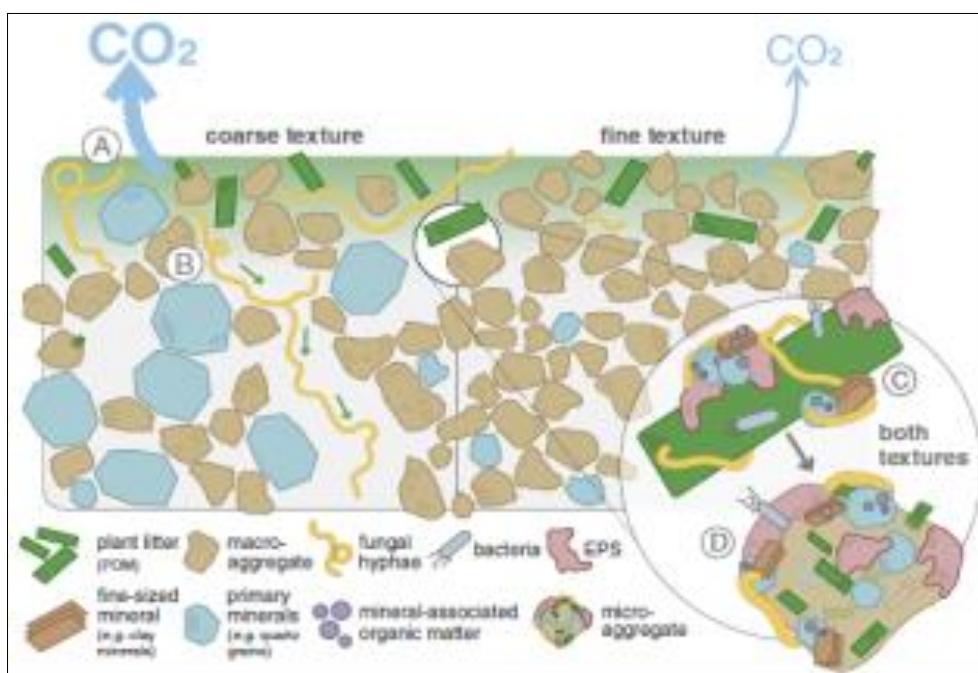


Fig 5b: Aggregate and mineral-associated organic matter formation in soils of different textures driven by interactions between litter, microorganisms, and soil matrix [Source: Witzgall *et al.*, 2021]^[71]

Priyadarshini *et al.* (2019)^[48] revealed that the contribution of 0.1-0.053 mm aggregate fraction was least (2.11-3.87%),

whereas 0.25-0.5 mm aggregate fraction was having the highest (27.3- 32.6%) contribution in total WSA at two

sampling depths (Table 2). The percentages of macro and meso-aggregates were significantly higher when FYM was applied either alone or in combination with inorganic fertilizers compared to unfertilized control at both soil depths. Application of FYM alone increased macro-aggregates (5-2 mm) by 162.6 per cent whereas meso-aggregates increased by 132 per cent in 2-1 mm fraction, by 283 per cent in 1-0.5 mm fraction over unfertilized control in 0-15 cm soil depth. The MWD was higher in FYM applied treatments than inorganic fertilizer alone and unfertilized control. Su *et al.* (2006) [65]

have also reported that long-term incorporation of FYM enhances the aggregate fractions (>2 mm and 250-2,000 μm) compared with the soils without FYM application. The increase in macro-aggregates (>2 mm) proportion by FYM application may be due to incorporation of additional organic residues and available C to the soils compared with inorganic fertilizer application alone. Reduction in micro-aggregates proportion with FYM application may be attributed to binding of the micro-aggregates to macro-aggregates due to secretion of mucilaginous substances (Sodhi *et al.*, 2009) [64].

Table 2: Distribution of water stable aggregates in different treatments of long-term fertilizer experiment analyzed by wet aggregate analysis [Source: Priyadarshini *et al.*, 2019] [48]

	5-2 mm	2-1 mm	% Water stable aggregates				% Total WSA	MWD
			1-0.5 mm	0.5-0.25 mm	0.25-0.1 mm	0.1-0.053 mm		
			0-15 cm					
Control	2.14±0.60 ^a	3.50±0.55 ^a	2.12±0.75 ^a	29.3±0.15 ^a	31.2±0.62 ^a	3.29±0.41 ^c	71.5	0.43
N	3.58±0.20 ^b	4.59±0.29 ^b	4.12±0.54 ^b	30.2±0.44 ^b	30.0±0.37 ^{bc}	2.62±0.32 ^{abc}	75.2	0.52
NP	4.41±0.20 ^c	4.55±0.44 ^c	6.10±0.37 ^c	32.1±0.40 ^{ab}	30.4±0.31 ^{ab}	2.98±0.28 ^{bc}	80.6	0.55
NK	4.61±0.68 ^{cd}	7.22±0.25 ^{cd}	7.33±0.06 ^d	32.6±0.39 ^{cd}	27.7±0.75 ^{bc}	2.49±0.38 ^{ab}	82.1	0.61
NPK	5.31±0.68 ^{de}	8.09±0.09 ^{de}	7.65±0.20 ^{de}	32.6±0.38 ^c	28.0±0.49 ^{bc}	2.44±0.10 ^{ab}	84.2	0.64
FYM	5.62±0.30 ^e	8.12±0.47 ^e	8.12±0.13 ^e	28.2±0.46 ^a	30.4±0.78 ^{bc}	2.32±0.83 ^a	82.8	0.65
N+FYM	5.51±0.40 ^e	8.19±0.40 ^e	8.28±0.41 ^e	30.1±0.29 ^b	30.0±0.38 ^{bc}	2.31±0.41 ^a	84.4	0.65
NP+FYM	5.93±0.31 ^e	8.40±0.42 ^e	9.98±0.68 ^f	30.2±0.41 ^b	30.1±0.20 ^c	2.21±0.48 ^a	86.8	0.66
NK+FYM	5.63±0.25 ^e	9.61±0.16 ^e	10.97±0.59 ^f	31.3±0.33 ^d	28.1±0.50 ^c	2.21±0.13 ^a	87.9	0.67
NPK+FYM	7.85±0.37 ^f	11.17±0.43 ^f	13.94±0.22 ^h	30.4±0.26 ^e	25.3±0.28 ^{bc}	2.42±0.33 ^a	91.2	0.78
			15-30 cm					
Control	1.54±0.35 ^a	3.32±0.27 ^a	2.03±0.41 ^a	27.3±0.11 ^a	31.7±0.10 ^a	3.87±0.35 ^a	69.8	0.40
N	2.96±0.32 ^b	4.41±0.71 ^b	4.01±0.32 ^b	28.7±0.26 ^b	30.6±0.18 ^b	2.50±0.50 ^b	73.2	0.50
NP	3.64±0.27 ^b	4.47±0.26 ^b	5.89±0.48 ^c	30.2±0.44 ^c	31.7±0.58 ^a	2.57±0.63 ^b	78.5	0.52
NK	3.63±0.41 ^b	4.78±0.63 ^b	5.98±0.49 ^c	31.1±0.27 ^d	30.1±0.28 ^{bc}	2.65±0.43 ^b	78.3	0.53
NPK	4.63±0.51 ^c	6.58±0.38 ^c	7.02±0.22 ^d	32.2±0.65 ^e	28.3±0.37 ^d	2.26±0.81 ^b	81.0	0.60
FYM	4.22±0.52 ^{bc}	7.28±0.25 ^d	8.12±0.27 ^e	28.1±0.71 ^b	30.1±0.24 ^{bc}	2.13±0.42 ^b	79.9	0.60
N+FYM	4.23±0.33 ^{bc}	7.34±0.54 ^d	8.20±0.44 ^e	28.1±0.54 ^b	30.1±0.45 ^{bc}	2.46±0.39 ^b	80.4	0.60
NP+FYM	4.38±0.78 ^{bc}	7.58±0.41 ^d	8.24±0.61 ^e	29.5±0.22 ^c	29.6±0.26 ^c	2.12±0.35 ^b	81.4	0.61
NK+FYM	4.34±0.22 ^{bc}	8.70±0.47 ^e	9.90±0.30 ^f	30.1±0.42 ^c	27.8±0.20 ^d	2.11±0.34 ^b	83.0	0.63
NPK+FYM	6.48±0.46 ^d	9.89±0.49 ^f	11.16±0.60 ^f	29.6±0.16 ^c	26.6±0.21 ^e	2.29±0.23 ^b	86.1	0.72

Value±SD is provided; within the same aggregate fraction different small letters indicate significant difference at $p < 0.05$

Witzgall *et al.* (2021) [71] also found that the changes caused by litter addition in microbial community structures between the textures were captured via the measurement of microbial-derived PLFA. The litter amendment led to a slight increase in the total PLFA content in the top layers of both the coarse-textured (61 $\text{nmol g}^{-1} \text{mg soil C}^{-1}$) and fine textured soil (76 $\text{nmol g}^{-1} \text{mg soil C}^{-1}$), whereas the total PLFA contents in the center and bottom layers were similar to those of the controls (Fig. 6a). While the differences in soil texture had no effect on the overall community structure, a strong response to litter addition was detected in fungal biomarkers. The increase in fungal markers was particularly pronounced in the top layer of the coarse-textured soil where fungal abundance increased by a factor of 5.4 compared to 2.6 in the fine-textured soil.

Moreover, when considering the proportion of fatty acids with did not incorporate litter-derived ^{13}C within the observed groups in relation to the total amount of enriched FAs in the sample, texture nor depth had an effect (Fig. 6b). This corroborated the consistent community structures detected during the PLFA analysis. When considering the proportions of ^{13}C -enriched FAs to unlabeled FAs within each microbial group, the proportion was by far the highest in the fungal markers (92% in the coarser-textured and 82% in fine-textured soil (Fig. 6b). This distinction of fungi compared to other microbial groups was significant in top layers of both textures, as well as in the center layer of the coarse-textured soil (over 42% of FAs were enriched compared to 21% in the fine-textured soil).

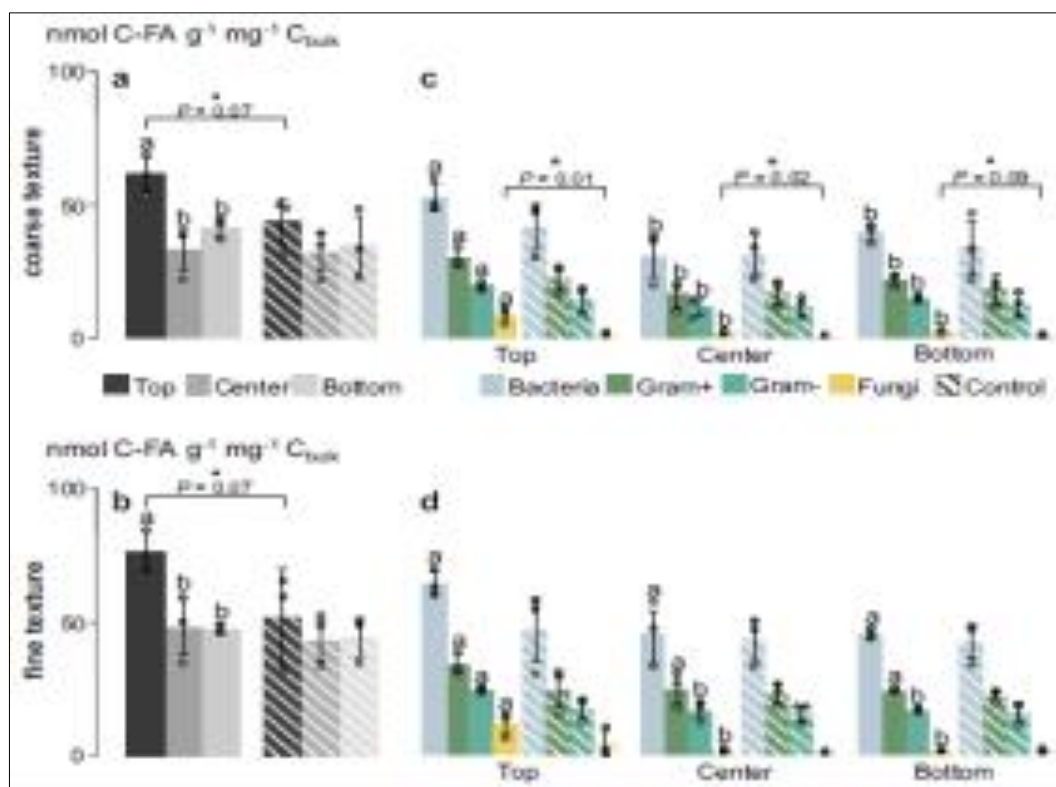


Fig 6a: Community structures and functionality of microorganisms [Source: Witzgall *et al.*, 2021] ^[71]

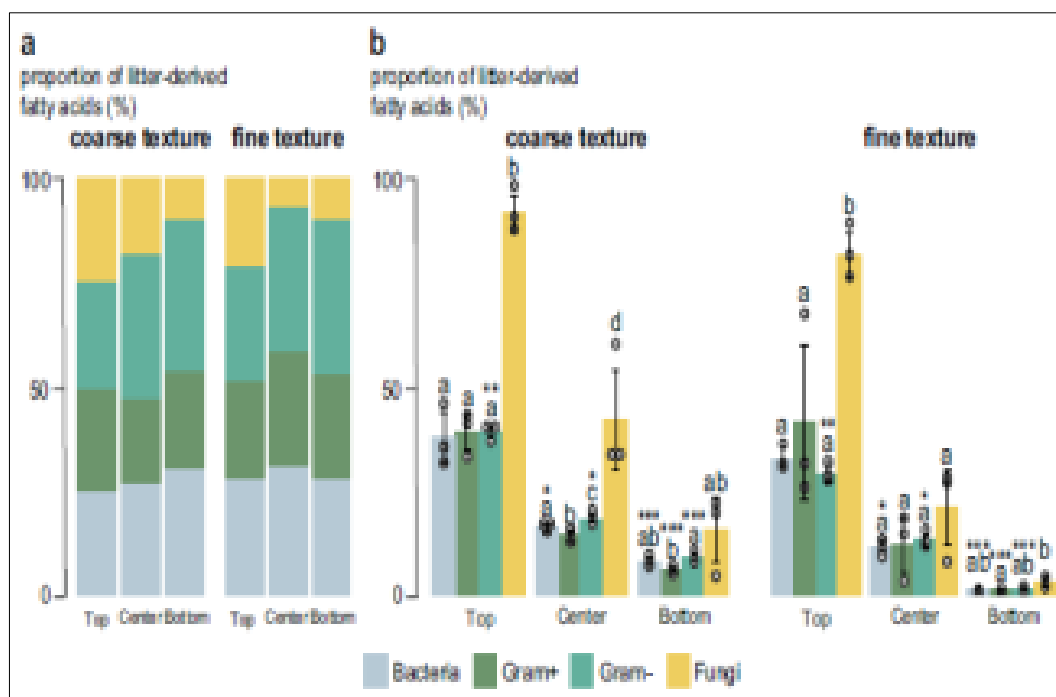


Fig 6b: Litter incorporation in microbial biomass [Source: Witzgall *et al.*, 2021] ^[71]

He *et al.* (2021) revealed that fertilization significantly affected the SOM content in bulk soils and in aggregates (Fig. 7a). Concentrations of SOM in bulk soil were manure > NPK + straw > NPK > CK. Compared to CK, SOM increased by 125%, 54.6% and 0.3% for manure, NPK + straw and NPK, respectively. In addition, after water-sieving, the SOM content was consistently higher in macro-aggregates (> 2 mm) than micro-aggregates (< 0.25 mm), irrespective of the fertilization type (Fig. 7a). For example, for CK, the SOM decreased almost 50% from macro-aggregates to micro-

aggregates. Compared to CK, the decline of SOM with aggregate size was less, with 7.6% and 39.1% decline for manure and NPK + straw, respectively. Similar SOM distribution patterns occurred among dry-sieved aggregate sizes. Moreover, the soil aggregate was dominated by size > 2 mm for manure and NPK + straw, while aggregates were dominated by size 2–0.25 mm for NPK and CK (Fig. 7b). The > 2 mm macro-aggregate portion displayed a slight decrease with the increase of concentration from 0 to aggregate portion with concentration depended on fertilization types. For

example, for aggregates (< 0.25 mm), manure and NPK + straw yielded in smaller percentage values and smaller degree of change with increased concentration compared to CK (Fig. 7b). When concentration increased from 0 to 0.001 mol L⁻¹, <

0.25 mm portion increased from 9.6 to 13.7% from 17.3 to 16.4% (-5%), and from 15.2 to 23.1% (52.4%) for manure, NPK + straw and CK, respectively.

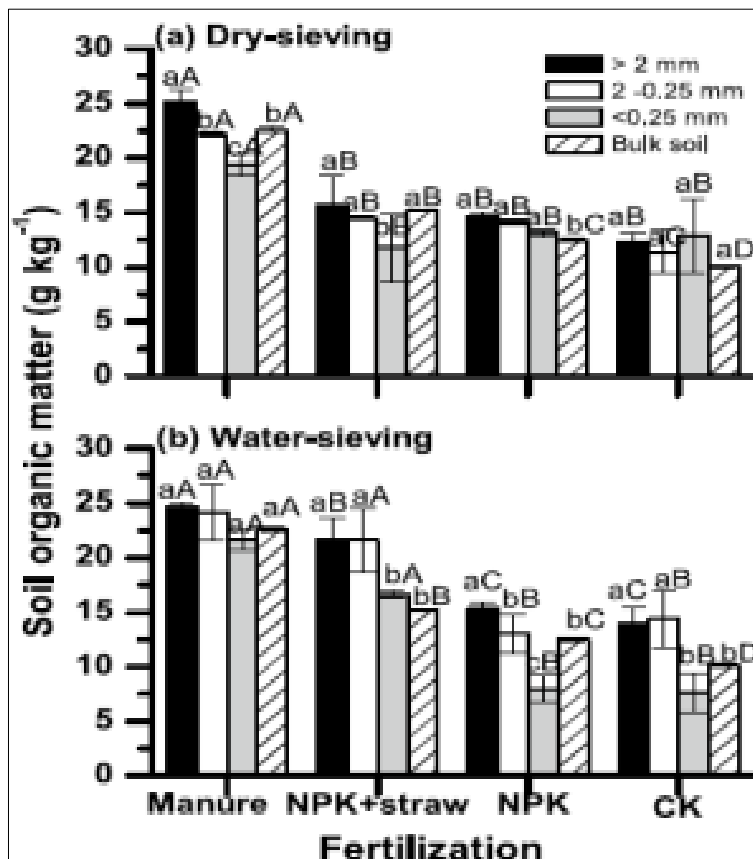


Fig 7a: Soil organic matter (SOM) distribution on different aggregate size across treatments at (a) dry sieving and (b) water sieving [Source: He *et al.*, 2021]

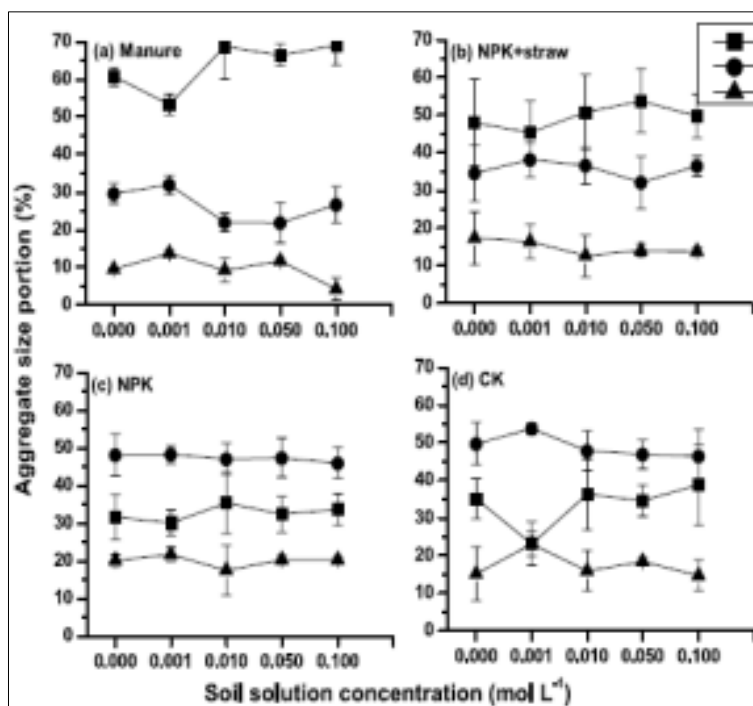


Fig 7b: Size distribution with concentration over four types of fertilizations, including (a) Manure, (b) NPK + straw, (c) NPK, (d) CK [Source: He *et al.*, 2021]

Soil Enzymatic Activity and Microbial Quotients

Priyadarshini *et al.* (2019) [48] observed that urease activity in surface soil (0- 15 cm) was least in unfertilized control (217.4 μg urea hydrolyzed $\text{g}^{-1} \text{h}^{-1}$) and highest in FYM+NPK (301.5 μg urea hydrolyzed $\text{g}^{-1} \text{h}^{-1}$) (Fig. 8a). Urease activity reduced at 15-30 cm soil depth irrespective of treatments which varies from 185.3 μg urea hydrolyzed $\text{g}^{-1} \text{h}^{-1}$ in control to 239.2 μg urea hydrolyzed $\text{g}^{-1} \text{h}^{-1}$ in NPK+FYM (Fig. 8a). Trend of urease activity at 15-30 cm depth was similar to surface soil. Urease activity was significantly higher in FYM alone compared to inorganic fertilizer alone treatments irrespective of soil depth. Urease activity was lowest in control and highest in FYM+NPK with 33.1-43.9 per cent increase over control across aggregate fractions (Fig. 8a). Macro-aggregates (5-2 mm) had highest urease activity and micro-aggregate fraction (0.1-0.053 mm) had least urease activity among aggregate fractions. Urease activity was lower at 15-30 cm depth compared to surface soil (0-15 cm) in all the aggregate fractions (Fig. 8a). The trend of urease activity at 15-30 cm depth in different aggregate fractions across treatments was

similar to surface soil. Moreover, combined application of FYM and inorganic fertilizers resulted in significantly higher protease activity against inorganic fertilizer alone or unfertilized control in all the aggregate fractions (Fig. 8b). In contrast to urease activity, protease activity was significantly higher in NPK treatments compared to FYM alone treatment with 21.4-131.0 per cent increase over FYM treatments across aggregate fractions in surface soil and 0.3-3.3 per cent increase in 15-30 cm soil depth (Fig. 8b). Protease activity decreased with soil depth in all the aggregate fractions. The amount of proteases may be indicative of the biological capacity indicated that the application of FYM along with inorganic fertilizer increased the protease activity in comparison to inorganic fertilizer alone treatment across aggregate fractions. This higher activity might be attributed to more substrate availability in these soils. This indicated that there is greater biological activity in FYM applied plots and extracellular enzymes stabilize by making complex with humic substances (Colvan *et al.*, 2001) [11].

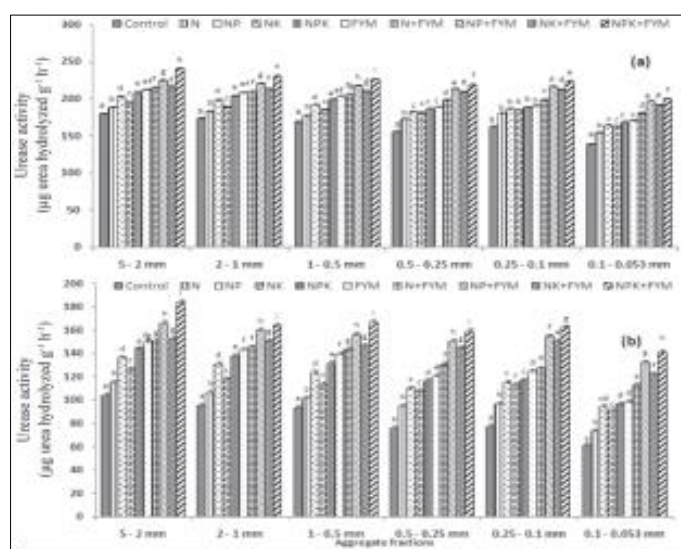


Fig 8a: Urease activity in different fractions of soil aggregates (a) 0-15 cm and (b) 15-30 cm in different treatments [Source: Priyadarshini *et al.*, 2019] [48]

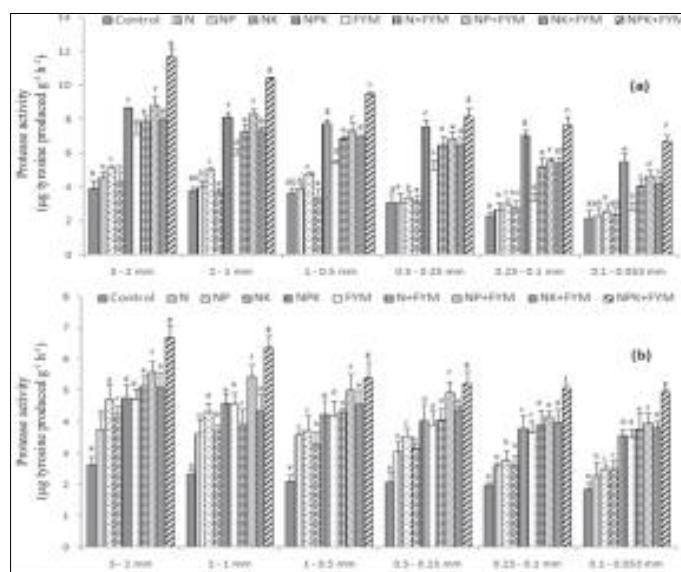


Fig 8b: Protease activity in different fractions of soil aggregates (a) 0-15 cm and (b) 15-30 cm in different treatments [Source: Priyadarshini *et al.*, 2019] [48]

Bharti *et al.* (2017) ^[8] reported that the urease activity increased by 4.99, 3.80 and 7.40 percent over control (102.6 mg of urea hydrolysed kg⁻¹ soil ha⁻¹) with the addition of compost, crop residue and compost + crop residue, respectively, whereas, inorganic treatments received 50, 100 and 150 percent NPK increased the total urease activity by 5.29, 12.62 and 16.90 percent over control (98.2 mg of urea hydrolysed kg⁻¹ soil ha⁻¹), respectively. Kumar *et al.* (2020) revealed that soil enzymatic activity (DHA, FDA and Alk-P) was significantly lower in CK and highest under RS_{10.0}N₁₅₀ treatment. DHA activity did not increase significantly in RS_{10.0}N₉₀ and RS_{10.0}N₁₂₀ treatments, but increased significantly with increased fertilizer-N application rate (RS_{10.0}N₁₅₀), compared with RS_{10.0}. Average across treatments, DHA in the surface soil layer was ~68.4% higher, than the sub-surface soil layer. The corresponding increase in FDA and Alk-P activity was ~10.5 and ~62.4%, respectively. The DHA, FDA, and Alk-P increased by ~49.4, 57.8, and 99.9%, ~45.1, 47.6, and 34.1% and ~65.2, 71.9, and 27.5%, respectively in RS₁₀N₉₀, RS₁₀N₁₂₀, and RS₁₀N₁₅₀ treatments as compared to CK.

Conclusions

This review study clearly indicated that applied organic fertilizers significantly increased total SOC contents and labile organic C fractions (DOC, LFOC and MBC) in agricultural soils. Moreover, the greatest increases were observed in treatment with the combined applications of manure, straw and mineral fertilizers. Organic fertilization also slightly altered the composition of microbial communities. Furthermore, the application of organic fertilizers resulted in 53%-85% greater cumulative mineralization of C. Soil labile C fractions and soil microbial communities predominantly determined the variance in C mineralization, while C/N ratios of labile fractions did not significantly influenced C mineralization in the current agricultural system. Fertilizer- application and conservation tillage significantly increased TOC, its labile C pools and soil enzymes compared with the CK. Among the labile pools; WEOc and BRS were the most sensitive indicators for assessing changes in TOC in the surface soil. The change in labile C pool in soil was related to change in soil enzymatic activity. The CMI was suitable for use as a sensitive indicator for assessing TOC changes in a RWCS. The TOC exhibited a linear increase with above-and below-ground plant biomass C.

No-tillage system promoted large macro-aggregation in both the 0.00-0.05 and 0.05-0.20 m layers. Accordingly, associated SOC concentration, and SOC stock within the >0.25 mm macro-aggregate fraction also significantly increased in the 0.00-0.05 m layer in the NT system, while those within the 2.00-0.25 mm aggregate fraction were significantly reduced in the 0.05-0.20 m layer under the NT system. It may be stated that the adoption of the NT system that increases SOC stock in the 0.00-0.05 m soil profile might be dependent on the macro-aggregate fractions and the high amount of associated SOC. Activities of all the enzymes reduced at 15-30 cm soil depth compared to surface soil in whole soil as well as in soil aggregate fractions. The reason for higher enzymatic activity in FYM applied plots may be attributed to higher organic matter content which provides a more positive environment for the accumulation of enzymes in the soil matrix.

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