

E-ISSN: 2278-4136 P-ISSN: 2349-8234 JPP 2019; 8(2): 1179-1186 Received: 06-01-2019 Accepted: 09-02-2019

#### Saurabh Tyagi

Faculty of Agricuture Rudra Institute of Technology Mawana Khurd, Chaudhary Charan Singh University, Meerut, Uttar Pradesh, India

#### Shivani Rana

Division of Biotechnology, Meerut Institute of Engineering & Technology, Meerut, Uttar Pradesh, India

#### NC Mahajan

Institute of Agricultural Sciences: Department of Agronomy, Banaras Hindu University, Varanasi, Uttar Pradesh, India

#### Shivi Tyagi

Department of Botany, Punjab University, Chandigarh, Punjab, India

#### **RK Naresh**

Department of Agronomy, Sardar Vallabhbhai Patel University of Agriculture & Technology, Meerut, Uttar Pradesh, India

Correspondence Saurabh Tyagi Faculty of Agricuture Rudra Institute of Technology Mawana Khurd, Chaudhary Charan Singh University, Meerut, Uttar Pradesh, India

# Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com



Journal of Pharmacognosy and

Phytochemistry

# Saurabh Tyagi, Shivani Rana, NC Mahajan, Shivi Tyagi and RK Naresh

#### Abstract

To improve C sequestration in soils and mitigate climate change, it is essential to understand how nutrient management strategies impact on soil organic carbon (SOC) stocks and labile fractions. Compared to conventional tillage, conservation tillage (no-tillage coupled with straw return) increased water-stable large macro-aggregates (>2 mm) by 35.18%, small macro-aggregates (2–0.25 mm) by 33.52% and micro-aggregates by 25.10% in the topsoil (0–20 cm). The subsoil (20–40 cm) also showed the same trend. Compared to conventional tillage without straw return, large and, small macro-aggregates and micro-aggregates in conservation tillage were increased by 24.52%, 28.48% and 18.12%, respectively. Straw return also caused a significant increase in aggregate-associated C within all the aggregate fractions in the topsoil. But the different is that conventional tillage with straw return resulted in more aggregate-associated C than conservation tillage in the subsoil. A considerable proportion of the SOC was found to be stocked in the small macro-aggregates under both topsoil (74.56%) and subsoil (67.09%).

Tillage systems can changes in soil organic carbon dynamics and soil microbial biomass by changing aggregate formation and C distribution within the aggregate. Wheat straw ditch-buried returning (WD) had significantly higher total organic carbon than did wheat straw returning with ploughing (WP) and no straw returning (CK) in wheat season. Soil dissolved organic carbon and easily oxidizable carbon contents were significantly increased by 21.3%, 24.3%, 38.6%, and 43.5% under wheat straw returning with rotary tillage (WR) than that under CK in rice and wheat seasons, respectively. Soil microbial biomass carbon (MBC) content was highest under WP in rice season, but in wheat season, WR had significantly higher MBC than WP and WD. The content of SOC in WSAma increased on average in the following order:  $T < G < G + NPK_1 < G + NPK_3 < T + FYM$ . Intensive soil cultivation in the T treatment resulted in a statistically significant build-up of SOC in WSAma at an average rate of 1.33, 1.18, 0.97, 1.22 and 0.76 g/kg/y across the size fractions > 5 mm, 5–3 mm, 2–1 mm, 1–0.5 mm and 0.5–0.25 mm, respectively. The content of non-labile carbon reflected the contents of SOC in WSA. The highest labile carbon (CL) in WSAma, as compared to others, was found in T+FYM. Overall, application of higher NPK doses resulted in higher content of C<sub>L</sub> in WSA<sub>ma</sub> compared with the lower applications of NPK. On the other hand, lower applications of NPK to soil increased the content of CL in WSAmi, as compared to G+NPK<sub>3</sub>. However, higher SOC content of 8.14 gkg<sup>-1</sup> of soil was found in reduced tilled residue retained plots followed by 10.34 g kg<sup>-1</sup>in furrow irrigated raised beds with residue retained plots. Whereas, the lowest level of SOC content of 5.49 gkg<sup>-1</sup>of soil were found in puddled transplanted rice followed by wheat planted under conventionally tilled plots. Average SOC concentration of the control treatment was 0.54%, which increased to 0.65% in the RDF treatment and 0.82% in the RDF+FYM treatment. Compared to F<sub>1</sub>control treatment the RDF+FYM treatment sequestered 0.33 Mg C  $ha^{-1}yr^{-1}$  whereas the NPK treatment sequestered 0.16 Mg Cha<sup>-1</sup>yr<sup>-1</sup>. Long term conservation tillage significantly increased 66.1%, 57.9%, 50.9%, 38.3%, 37.3% and 32% LFOC, SOC, PON, TN, LFON, DOC and POC, over conventional tillage (T7) treatment and WSC 39.6% in surface soil and 37.4% in subsurface soil. Under RWCS, TOC contents were similar in 50% RDN as CF+50% RDN as GM/SPM (F<sub>6</sub>) and 1/3<sup>rd</sup>N as CF+1/3rdN as FYM+1/3rdN as GM/SPM (F7) and 75% RDN as CF+25% RDN as FYM (F3) plots and significantly higher than those in control (no manure and fertilizer) ( $F_1$ ) plots (by 50.4% 48.3%, and 43.3% respectively).

Keywords: Microbial biomass, conservation tillage, total organic carbon, labile organic C fraction

#### Introduction

Soil organic carbon (SOC) is recognized as the largest terrestrial carbon (C) reservoir and has gained much attention because of its importance to soil fertility, crop productivity, and climate change mitigation (Sollins *et al.*, 2007; Lal, 2004) <sup>[42, 25]</sup>.

Fertilization is an important determinant of quantities of SOC in croplands since it can change the equilibrium between primary C inputs and C decomposition (Dou et al., 2016; Neff et al., 2002) <sup>[13, 38]</sup>. Soil microorganisms are the main decomposers of SOC and key drivers of soil nutrient cycling in agricultural eco-systems (LuÈtzow et al., 2006)<sup>[29]</sup>. A better understanding of the mechanisms of SOC decomposition via microorganisms is critical for identifying fertilization strategies that maintain and improve soil C accumulation and soil fertility. The changes of soil microbial communities under different fertilization regimes may be contributed to changes in environmental characteristics, such as soil water content (Demoling et al., 2007)<sup>[9]</sup>, temperature (Fang *et al.*, 2016) <sup>[14]</sup>, pH (Dong *et al.*, 2014) <sup>[11]</sup> and substrate availability (Li et al., 2015)<sup>[27]</sup>. The content and quality of SOC are considered key factors that affect soil microbial communities (Li et al., 2015) [27]. However, increases in SOC content following the addition of fertilizers may take considerable time. Consequently, changes in SOC cannot fully and quickly reflect the influence that the complexity of the organic compounds may have on the microbiological processes controlling nutrient availability. Soil labile C fractions are a series of small, but sensitive, proportions of SOC with turnover times of a few days to months. It was revealed that soil labile C fractions, like dissolved organic C (DOC) and microbial biomass C (MBC) were major determinants for the preservation of soil microbial diversity in long-term fertilization trials (Li et al., 2015)<sup>[27]</sup>.

The slight positive effect of tillage reduction and the fact that fertilization had little effect on organic C sequestration in that study suggest that the physical environment, characterized by hot and relatively dry conditions constitutes a great challenge to increasing soil organic matter content. Another mechanism by which soil organic matter is retained in conservationtillage systems may be due to reduced oxygen availability below the surface of no-till systems, which affects decomposition rates (Wershaw, 1993)<sup>[46]</sup> and the distribution of aerobic and anaerobic microbes and microbial processes (Doran, 1980) <sup>[12]</sup>. Slower subsurface decomposition rates would lower oxidative losses of organic C. Tillage also affects the distribution of the soil microbial biomass, being displaced toward the soil surface with no-tillage, and toward lower depths with plow tillage. Specific microbial population distributions may also be affected by tillage (Doran, 1980)<sup>[12]</sup>. Holland and Coleman (1987) <sup>[17]</sup> reported an increase in the fungal to bacterial ratio with no-tillage, which may have implications for C and N cycling in the soil. Tillage has been shown to disproportionately affect the more labile forms of organic carbon in soil (Cambardella and Elliott, 1992)<sup>[6]</sup> including the fraction that accounts for most of the simpler in macro-aggregate polymers involved formation. Stratification of soil properties is a natural consequence of soil development that can become accentuated in soils subjected to reduced tillage. Unger (1991)<sup>[44]</sup> and Bruce et al. (1995)<sup>[5]</sup> reported that soil nutrients become stratified when no-till management is employed. There is a marked stratification of soil organic matter with soil depth under notillage (Blevins et al., 1984)<sup>[4]</sup>.

Soil C pools that promote microbial activity and nutrient cycling are primarily the labile pools (Kaye and Hart, 1997), a series of small, but variable, proportions of SOC with turnover times of a few days to months. These pools have been suggested as early sensitive indicators of soil quality which influence soil function in specific ways (Xu *et al.*, 2011; Blanco-Moure *et al.*, 2016) <sup>[3]</sup>. Triberti *et al.* (2008) <sup>[43]</sup>

found that continuous additions of organic material to the soil led to a SOC build up at rates 0.16-0.26 t C/ha/y over a 22 year period. Purakayastha et al. (2008) [39] reported the rates of SOC build-up in the soil up to a level of 1.0 Mg/ha/y due to NPK + farmyard application under a maize-wheat-cowpea cropping system. Abdollahi et al. (2014)<sup>[1]</sup> showed the rate of SOC due to added organic fertilisers to be between 220-240 kg C/ha/y. Zibilsk et al. (2002) [50] reported that the No-till resulted in significantly greater soil organic C in the top 4 cm of soil, where the organic C concentration was 58% greater than in the top 4 cm of the plow-till treatment. In the 4–8 cm depth, organic C was 15% greater than the plow-till control. The differences were relatively modest, but consistent with organic C gains observed in hot climates where conservation tillage has been adopted. Higher concentrations of total soil N occurred in the same treatments; however a significant reduction in N was detected below 12 cm in the ridge-till treatment. The relatively low amount of readily oxidizable C (ROC) in all tillage treatments suggests that much of the soil organic C gained is humic in nature which would be expected to improve C sequestration in this soil. Therefore, the review paper would be beneficial for determining how tillage and fertilizer practices influence soil organic carbon fractions and stocks and soil microbial biomass in sub-tropical ecosystems.

# Soil labile organic C fractions

Haynes (2005)<sup>[16]</sup> reported values of 20–45% for POC and 2– 18% for LFOC as a proportion of SOC in agricultural soils and Wander (2004)<sup>[45]</sup> reported that the proportion of POC and LFOC varied from 2 to 30%.

Chen et al. (2009)<sup>[7]</sup> revealed that the MWD and GMD of soil aggregates were significantly influenced by tillage. At 0-15 cm, MWDs and GMDs were significantly lower under CT than ST or NT, whereas the differences between ST and NT were not significant. At 15-30 cm, ST and NT had higher MWD and GMD than CT, but the differences were only significant between CT and NT. Both MWDs and GMDs decreased with increase in soil depth for all tillage treatments. Yang et al. (2005) <sup>[47]</sup> reported that soil PMOC, LFOC, and POC under water regime of continuous water-logging decreased by 30.6, 8.3, and 10.6% in wheat straw treatment, respectively, as compared to the water regime of alternative wetting and drying. This confirmed that the adoption of soil water regimes is an important factor to improve the transformation of soil organic carbon pools after the addition of rice straw. Yang et al. (2012) showed that LFOC, POC, and PMOC were improved by 2.25, 1.84, and 2.15 times after the addition of wheat straw or maize stalk in a silt clay loam soil. They also mentioned that PMOC was higher in wheat straw or maize stalk-amended soil than the control could be explained by the higher labile organic carbon inputs, which associated with the straw and stalk.

Chen *et al.* (2009) <sup>[7]</sup> stated that the content of large macroaggregates (>2 mm) was very low (around 1% of the soil weight). Small macro-aggregates (2–0.25 mm) represented the greatest portions (52–70% of whole soil) in all treatments at both 0–15 and 15– 30 cm. At 0–15 cm, CT contained significantly less small macro-aggregates (2– 0.25 mm) than ST or NT, which were not different from each other. In contrast, CT had significantly higher amounts of microaggregates (0.25–0.05 mm) than ST or NT, which were similar. At 15– 30 cm, ST and NT contained higher amounts of small macro-aggregates than CT; the difference was only significant between CT and NT. However, the <0.05 mm fraction was dominated by CT. Lewis *et al.*, (2011) <sup>[26]</sup> increasing tillage intensity could reduce DOC levels in soils as a result of destruction of soil macro-aggregates and elevated respiration. Lower amount of DOC, hence is likely under CT due to increased soil disturbances subjecting aggregated protected SOC fraction to rapid decomposition via oxidation.

Bhattacharya et al. (2013) [2] reported that tillage-induced changes in POM C were distinguishable only in the 0- to 5cm soil layer; the differences were insignificant in the 5- to 15-cm soil layer. Plots under ZT had about 14% higher POM C than CT plots (3.61 g kg<sup>-1</sup> bulk soil) in the surface soil layer. Liu *et al.* (2013) <sup>[28]</sup> revealed that the 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. Irrespective of soil depths, NP+FYM invariably showed higher content of DOC over all other treatments. The CK and N treatments showed lower content of DOC. The DOC concentrations in 0-20 cm, 20-40 cm and 40-60 cm depths were observed highest for NP+FYM followed by NP+S and FYM, and both of them were significant higher than NP. However, in the deeper layers (60-80 cm and 80-100 cm), the difference in DOC among the treatments was not significant.

Zhao et al. (2014) [49] concluded that the contents of SOC, TN, POC and LOC responded differently as the change of soil depth. In all land use types, contents of SOC, TN, POC and LOC in top soil (0-10 cm) were 3.26-7.86 gkg<sup>-1</sup> 0.39-0.72 gkg<sup>-1</sup>, 0.65–1.31 gkg<sup>-1</sup> and 0.76–1.07 gkg<sup>-1</sup>, respectively, which were significantly higher than other soil layers. The contents of SOC, TN, POC and LOC decreased significantly in soil depth of 10-40 cm while the decreases trended to be flatter in subsoil (40-100 cm). Additionally, the differences in contents of SOC, TN, POC and LOC in deep subsoil (100-200 cm) were negligible. Vegetation can greatly influence soil quality, C and N cycling, and regional socioeconomic development (Fu et al., 2010)<sup>[15]</sup>. Sheng et al., (2015)<sup>[40]</sup> also found that the stocks associated with the different LOC fractions in topsoil and subsoil responded differently to land use changes. POC decreased by 15%, 38%, and 33% at 0-20 cm depth, and by 10%, 12%, and 18% at 20-100 cm depth following natural forest conversion to plantation, orchard, and sloping tillage, respectively. Regarding the different POC components, only fPOC stock in 0-20 cm topsoil decreased by 21%, 53%, and 51% after natural forest conversion to plantation, orchard, and sloping tillage, respectively. This implied that the reduction of POC stock after land use change mainly resulted from the loss of topsoil fPOC, which, consequently, could be used as a sensitive indicator to detect SOC changes. Noticeably, fPOC stock in subsoil below 40 cm increased by 11-74% following the land use change, indicating that changes in POC fractions in subsoil may follow the opposite direction to those in topsoil. Loss of LFOC occurred not only in topsoil, but also in subsoil below 20 cm following land use change. The topsoil showed a greater reduction in LFOC stock than did subsoil following the conversion of natural forest to orchard and sloping tillage. LFOC appeared to be more sensitive to land use changes than SOC both in top and subsoil. The decrease in ROC stock

through the soil depth profile following land use change was smaller than that of LFOC.

Naresh et al. (2016)<sup>[34]</sup> also found significantly higher POC content was probably also due to higher biomass C. Results on PON content after 3-year showed that in 0-5 cm soil layer of CT system,  $T_1$ , and  $T_5$  treatments increased PON content from 35.8 mgkg<sup>-1</sup> in CT ( $T_9$ ) to 47.3 and 67.7 mgkg<sup>-1</sup> without CR, and to 78.3, 92.4 and 103.8 mgkg<sup>-1</sup> with CR @ 2, 4and 6 tha-1, respectively. The corresponding increase of PON content under CA system was from 35.9 mgkg<sup>-1</sup> in CT system to 49 and 69.6  $mgkg^{-1}$  without CR and 79.3, 93.0 and 104.3mgkg<sup>-1</sup> with CR @ 2, 4 and 6 tha<sup>-1</sup>, respectively. Small improvement in PON content was observed after 4 years of the experiment. Naresh *et al.* (2017) <sup>[35]</sup> reported that the  $T_3$ treatment resulted in significantly increased 66.1%, 50.9%, 38.3% and 32% LFOC, PON, LFON and POC, over T<sub>7</sub> treatment and WSC 39.6% in surface soil and 37.4% in subsurface soil. LFOC were also significantly higher following the treatments including organic amendment than following applications solely of chemical fertilizers, except that the F<sub>5</sub>, F<sub>6</sub> and F<sub>7</sub> treatments resulted in similar LFOC contents. Application solely of chemical fertilizers had no significant effects on LFOC compared with unfertilized control plots. Nevertheless, application of F<sub>5</sub> or F<sub>6</sub> significantly increased contents of POC relative to  $F_1$  (by 49.6% and 63.4%, respectively).

Juan et al. (2018) <sup>[19]</sup> observed that the pure organic manure treatments (DMA and SMA) showed significantly higher concentrations of POC as compared to integrate (1/ 2SMF + 1/2SMA) and mineral-fertilized plots (DMF and SMF). POC constituted 10.20 to 23.65% of total SOC with a mean value of 16.43%. Highest proportion of POC was observed under DMA, followed by SMA, which was not significantly different from DMF; 1/ 2SMF + 1/2SMA and SMF had a lower proportion of POC and the lowest proportion was found in the CK treatment. In the surface soil (0-20 cm), the LFOC concentration was 60% higher under DMA than under CK. Other treatments showed no significant effects on LFOC concentrations relative to CK. At the same standard N input level, SMA contained 27% higher organic C in LFOC than SMF, however, there was no significant difference between 1/ 2SMF + 1/2SMA and SMF. Long term application of mineral fertilizers alone slightly decreased LFOC concentrations relative to CK, but this effect was not statistically significant. The fraction of SOC as LFOC ranged from 1.73 to 3.29% with an average value of 2.56% in SOC, and exhibited a pattern similar to MBC.

Kumar *et al.* (2018)<sup>[24]</sup> also found that the ZTR (zero till with residue retention) (T<sub>1</sub>) and RTR (Reduced till with residue retention) (T<sub>3</sub>) showed significantly higher BC, WSOC, SOC and OC content of 24.5%, 21.9%, 19.37 and 18.34 gkg-1, respectively as compared to the other treatments. Irrespective of residue retention, wheat sown in zero till plots enhanced 22.7%, 15.7%, 36.9% and 28.8% of BC, WSOC, SOC and OC, respectively, in surface soil as compared to conventional tillage. Simultaneously, residue retention in zero tillage caused an increment of 22.3%, 14.0%, 24.1% and 19.4% in BC, WSOC, SOC and OC, respectively over the treatments with no residue management. Similar increasing trends of conservation practices on different forms of carbon under subsurface (15- 30 cm) soil were observed however, the magnitude was relatively lower. However, the 0-15 and 15-30 cm, POC, PON, LFOC and LFON content under ZT and RT with residue retention was greater than under without residue and conventional sown plots, respectively. The decrease in the

disruption of soil macro-aggregates under ZT plots permitted a greater accumulation of SOC between and within the aggregates. Thus, less soil disturbance is the major cause of higher POC in the ZT and RT plots compared with the CT plots in the 0-15cm and 15-30 cm soil layers. This phenomenon might lead to micro-aggregate formation within macro-aggregates formed around fine intra-aggregate POC and to a long-term stabilization of SOC occluded within these micro-aggregates. The sequestration rate of POC, PON, LFOC and LFON in all the treatments followed the order 200 kg Nha<sup>-1</sup> (F<sub>4</sub>) > 160 kg Nha<sup>-1</sup> (F<sub>3</sub>) > 120 kg Nha<sup>-1</sup> (F<sub>2</sub>) >800 kg Nha<sup>-1</sup> (F<sub>1</sub>) > control (unfertilized) (F<sub>0</sub>). Kashif *et al.* (2019) also found that the particulate organic carbon (POC), easily oxidizable carbon (EOC), dissolved organic carbon (DOC) contents of 0-20 cm depth were 80, 22 and 13%, respectively, higher under no-tillage with straw returning (NTS) treatment.

# Soil organic carbon stocks

Dikgwatlhe *et al.* (2014) compared with PTO and PT, significantly higher SOC and N concentrations were observed in surface layer (0-10 cm depth) under NT and RT. In 2004, the SOC stocks were lower under NT and RT than under PT and PTO, however, the opposite trend was observed in 2012. Compared with 2001, the net profile (0-30 cm) SOC sequestration rate was 10.60, 13.95, 13.65, and 14.92 Mg ha<sup>-1</sup>in 2012 under PTO, PT, RT, and NT, respectively. As for stocks in the 0-50 cm profile, no significant differences were observed among NT, RT, and PT. The trends in N stocks in profile (0-30, 0-50 cm depth) were NT>TR>PT>PTO in both the years. Compared with other treatments, SOC and N stocks were the lowest under PTO. Therefore, crop residues play an important role in SOC and N management, and improvement of soil quality.

Lu et al. (2015) <sup>[29]</sup> indicated that compared with straw removal (SR), SI significantly increased soil C storage by 12%. Moreover, incorporation of chopped straw with tillage treatment (ploughing and rotary tillage) increased C storage compared to un-chopped straw without tillage treatment. SI implementation with upland cropping, in the northwest and northeast resulted in higher C storage compared with rice cropping, and in the northern and southern regions. Changes in soil C were observed based on SI variables, including tillage and straw amounts in fine-textured soils, however straw amount rather than tillage treatment exhibited a greater influence on soil C in coarse-textured soils. Singh et al. (2015) <sup>[41]</sup> revealed that the total carbon stock in the topsoil (equivalent to 200 kg m<sup>-2</sup>) was slightly lower under reduced tillage (5.0 kg m<sup>-2</sup>) than under conventional tillage (5.2 kg m<sup>-2</sup>). Reduced tillage changed the soil composition by increasing the percentage of macro-aggregates and decreasing the percentage of micro-aggregates. However, due to the higher total amount of macro-aggregates in the soil, more carbon was bound to the macro-aggregate-occluded microaggregates in reduced tillage. Moreover, reduced tillage can improve clay soil structure, generally the chances to increase topsoil carbon sequestration by reduced tillage or straw management practices appear limited in cereal monoculture systems.

Khairul *et al.* (2016) <sup>[21]</sup> also found that after 4 years, ZT under WDT and WMT significantly increased soil organic matter (SOM) at 0–150 mm depth. Soil organic carbon (C) increased at a rate of 1.17 and 1.14 tha<sup>-1</sup>yr<sup>-1</sup>in ZT under WDT and WMT, respectively, while CT and DT under WFT were almost unchanged. After 4 years of tillage and rice–wheat systems, TOC ranged from 16-28 t/ha in CT under WFT to

21.47 t/ha in ZT under WDT, while in ZT under WMT it was 21.35 t/ha. In the surface 150 mm of soil, TOC increased by 28 and 27% in ZT under WDT and WMT cropping systems. Kuhn *et al.* (2016) <sup>[23]</sup> also found that the benefit of NT compared to CT on the changes of SOC stocks varied across different soil depths. In topsoil layers (above 20 cm), NT in general had greater SOC stocks than CT but the benefit tended to decline with soil depths, and even turned to be negative in soil layers deeper than 20 cm. Zhang *et al.* (2016) <sup>[48]</sup> reported that an application of inorganic fertilizers (NPK) plus animal manure over 20-30 years significantly increased SOC stocks to 20-cm depth by 32-87% whilst NPK plus wheat/maize straw application increased it by 26-38% compared to controls.

Das et al. (2017)<sup>[8]</sup> revealed that the total organic C increased significantly with the integrated use of fertilizers and organic sources (from 13 to 16.03 g kg<sup>-1</sup>) compared with unfertilized control (11.5 g kg<sup>-1</sup>) or sole fertilizer (NPKZn; 12.17 g kg<sup>-1</sup>) treatment at 0-7.5 cm soil depth. Naresh et al. (2017)<sup>[35]</sup> reported that the highest SOC concentration was obtained for 0-5 cm depth and decreased with sub surface depth for all treatments. The SOC concentration in 0-5 and 5-15 cm depths increased significantly by farmyard manure or GM/SPM application. At the 0-5 and 5-15 cm soil depths, SOC was highest in 50% RDN as CF+50% RDN as FYM (F<sub>5</sub>) followed by 50% RDN as CF+50% RDN as GM/SPM (F<sub>6</sub>) treatments and the least in Control (no manure and fertilizer) F1treatment. The total SOC stocks in the 0-15 cm layer was 35.17 Mgha<sup>-1</sup> for 50% RDN as CF+50% RDN as FYM-treated soils compared with 28.43 Mgha<sup>-1</sup> for 100% RDN as CFtreated plots and 26.45 Mg ha<sup>-1</sup> for unfertilized control plots. Soil organic C content in the 0–15 cm soil layer in the plots under 50% RDN as CF+50% RDN as FYM treatment was 16% higher than that under 75% RDN as CF+25% RDN as FYM treated plots. The TOC in surface soil were in the order of 50% RDN as CF+50% RDN as FYM (23.65 g kg<sup>-1</sup>) > 50% RDN as CF+50% RDN as GM/SPM (21.47 g  $kg^{-1}) > 1/3^{rd}\,N$ as CF+1/3<sup>rd</sup>N as FYM+1/3<sup>rd</sup>N as GM/ SPM (21.40 gkg<sup>-1</sup>) >75% RDN as CF+25% RDN as FYM (19.64 gkg-1) >unfertilized control (10.99 gkg<sup>-1</sup>).

Dhaliwal et al. (2018) <sup>[10]</sup> revealed that the mean SOC concentration decreased with the size of the dry stable aggregates (DSA) and water stable aggregates (WSA). In DSA, the mean SOC concentration was 58.06 and 24.2% higher in large and small macro-aggregates than in microaggregates respectively; in WSA it was 295.6 and 226.08% higher in large and small macro-aggregates than in microaggregates respectively in surface soil layer. The mean SOC concentration in surface soil was higher in DSA (0.79%) and WSA (0.63%) as compared to bulk soil (0.52%). Krishna et al., (2018) <sup>[22]</sup> reported that the total organic carbon (TOC) allocated into different pools in order of very labile > less labile > non labile >labile, constituting about 41.4, 20.6, 19.3 and 18.7%, respectively. In comparison with control, system receiving farmyard manure (FYM-10 Mg ha<sup>-1</sup> season1) alone showed greater C build up (40.5%) followed by 100% NPK+FYM (120:60:40 kg N, P, K ha<sup>-1</sup> + 5 Mg FYM ha<sup>-1</sup> season-1 ) (16.2%). In fact, a net depletion of carbon stock was observed with 50% NPK (-1.2 Mg ha<sup>-1</sup>) and control (-1.8 Mg ha<sup>-1</sup>) treatments. Only 28.9% of C applied through FYM was stabilized as SOC. A minimal input of 2.34 Mg C ha<sup>-1</sup> yr <sup>1</sup> is needed to maintain SOC level. The magnitude of carbon pools extracted under a gradient of oxidizing conditions was as follows:  $C_{VL} > C_{LL} > C_{NL} > C_L$  constituting about 41.4, 20.6, and 19.3 and 18.7%, respectively, of the TOC.

However, the contribution of VL, L and LL pools to SOC was 51.2, 23.1 and 25.5%, respectively. While active pool ( $C_{VL}$  +  $C_L$ ) constituted about 60.1%, passive pool ( $C_{LL}$  +  $C_{NL}$ ) represented 39.9% of the TOC.

Naresh *et al.*, (2018) <sup>[36]</sup> estimated that the coarse fractions SA and POM and the liquid DOC fraction contributed only 4%, 2.3% and 1.7% respectively to the total SOC stock. Furthermore, 93% of the total surplus of SOC due to 40 years of residue incorporation was found in the SC fraction. Within the SC fraction, the greatest change in SOC stock occurred in the more active SC-rSOC fraction, while the more passive rSOC on average did not change at all.

# Microbial community composition

Mandal et al. (2013) [33] reported that averaged across fertilization and manure treatments, MBC varied significantly with soil depth, with mean values of 239, 189 and 127 mg kg-<sup>1</sup>at 0–7.5, 7.5–15 and 15–30 cm depths respectively. Surface soil had higher MBC than deeper soil layers, due primarily to the addition of leftover CRs and root biomass to the topsoil. When averaged across soil depths, the MBC content under the different treatments was in the order: NPK+GR +FYM>NPK+FYM=NPK +GR> NPK + SPM> NPK+CR> PKZnS >NPKZn = control. Incorporation of CR slows mineralization processes; hence, microbes take longer to decompose the residue and use the released nutrients. Conversely, incorporation of GR, with a narrow C: N ratio, hastened mineralization by enhancing microbial activity in the soil (Nayak et al. 2012) [37].

Guo et al. (2016) reported that as compared with CT treatments, NT treatments increased MBC by 11.2%, 11.5%, and 20%, and dissolved organic carbon (DOC) concentration by 15.5%, 29.5%, and 14.1% of bulk soil, >0.25 mm aggregate, and <0.25 mm aggregate in the 0-5 cm soil layer, respectively. Compared with NS treatments, S treatments significantly increased MBC by 29.8%, 30.2%, and 24.1%, and DOC concentration by 23.2%, 25.0%, and 37.5% of bulk soil, >0.25 mm aggregate, and <0.25 mm aggregate in the 0-5 cm soil layer, respectively. Conservation tillage (NT and S) increased microbial metabolic activities and Shannon index in >0.25 and <0.25 mm aggregates in the 0-5 cm soil layer. Naresh et al. (2018) [36] revealed that the tillage intensity increased there was a redistribution of SOC in the profile, but it occurred only between ZT and PRB since under CT, SOC stock decreased even below the plow layer. However, higher SOC content of 8.14 g kg<sup>-1</sup> of soil was found in reduced tilled residue retained plots followed by 10.34 g kg<sup>-1</sup>in furrow irrigated raised beds with residue retained plots. Whereas, the lowest level of SOC content of 5.49 g kg<sup>-1</sup> of soil were found in puddled transplanted rice followed by wheat planted under conventionally tilled plots. Average SOC concentration of the control treatment was 0.54%, which increased to 0.65% in the RDF treatment and 0.82% in the RDF+FYM treatment. Compared to F<sub>1</sub>control treatment the RDF+FYM treatment sequestered 0.33 Mg C ha<sup>-1</sup>yr<sup>-1</sup>whereas the NPK treatment sequestered 0.16 Mg Cha<sup>-1</sup>yr<sup>-1</sup>.

# Soil Microbial biomass carbon

SMB is defined as the small (0-4%) living component of soil organic matter excluding macro-fauna and plant roots (Dalal, 1998). Soil microbial biomass carbon ( $C_{mic}$ ) have been used as indicators of changes in soil organic matter status that will occur in response to alterations in land use, cropping system, tillage practice and soil pollution (Sparling *et al.*, 1992). Jiang *et al.* (2011) <sup>[18]</sup> observed that the highest levels of MBC were

associated with the 1.0– 2.0 mm aggregate size class (1025 and 805 mg C kg<sup>-1</sup> for RNT and CT, respectively) which may imply that RNT was the ideal enhancer of soil productivity for this subtropical rice ecosystem. However, the lowest in the <0.053 mm fraction (390 and 251 mg Ckg<sup>-1</sup> for RNT and CT respectively). It is interesting to note the sudden decrease of MBC values in 1–0.25 mm aggregates (511 and 353 mg C kg<sup>-1</sup> for RNT and CT, respectively). The highest values corresponded to the largest aggregates, N4.76 mm, (6.8 and 5.4% for RNT and CT, respectively) and the lowest to the aggregate size of 1.0–0.25 mm (1.6 and 1.7 for RNT and CT, respectively).

Tripathi et al., (2014) observed that the significant positive correlations were observed between TOC and organic C fractions (POC and SMBC), illustrating a close relationship between TOC and POC and TOC and SMBC and that SOC is a major determinant of POC and SMBC. The microbial biomass carbon includes living microbial bodies (bacteria, fungi, soil fauna and algae) (Divya et al., 2014); it is more sensitive to soil disturbance than TOC. The proportion of SMBC to TOC is evaluation of carbon availability indexes for agriculture soil, which is usually 0.5-4.6% (Marumoto and Domsch, 1982). Liu et al., (2012) showed that SMBC may provide a more sensitive appraisal and an indication of the effects of tillage and residue management practices on TOC concentrations. Ma et al., (2016) <sup>[32]</sup> reported that the differences in SMBC were limited to the surface layers (0-5 and 5-10 cm) in the PRB treatment. There was a significant reduction in SMBC content with depth in all treatments. SMBC in the PRB treatment increased by 19.8%, 26.2%, 10.3%, 27.7%, 10% and 9% at 0-5, 5-10, 10-20, 20-40, 40-60 and 60–90

cm depths, respectively, when compared with the TT treatment. The mean SMBC of the PRB treatment was 14% higher than that in the TT treatment.

Ma *et al.* (2016) <sup>[32]</sup> reported that the proportion of SMBC to TOC ranged from 1.02 to 4.49, indicating that TOC is relatively low, or due to sampling for the summer after spring harvest, when soil temperature is high, the microbial activity is relatively strong. The SMBC at all depths (0–90 cm) with a sharp decline in depth increased perhaps due to a higher microbial biomass and organic matter content. SMBC was significantly higher in PRB in the surface soil layer (0–10 cm) than in TT and FB, which showed that no-till and accumulation of crop residues enriches the topsoil with microbial biomass.

# Conclusion

Soil microbial biomass, the active fraction of soil organic matter which plays a central role in the flow of C and N in ecosystems responds rapidly to management practices, and serves as an index of soil fertility. The practices of crop residue retention and tillage reduction provided an increased supply of C and N which was reflected in terms of increased levels of microbial biomass, N-mineralization rate in soil. Residue retention and tillage reduction both increased the proportion of organic C and total N present in soil organic matter as microbial biomass. Microbial immobilization of available-N during the early phase of crops and its pulsed release later during the period of greater N demand of crops enhanced the degree of synchronization between crop demand and N supply. The maximum enhancement effects were recorded in the minimum tillage along with residue retained treatment. The conservation tillage (ST and NT) treatments effectively improved the soil structure and strengthened the

stability of water-stable soil aggregates. In addition, they increased the SOC content and storage in aggregates of different sizes with comparison of MP and CT. Furthermore, long-term adoption of conservation tillage methods significantly increased the content of water-stable macro-aggregates and of aggregate MWD, and increased the SOC content, ratio of, and storage in the macro-aggregates. In particular, the ST treatment increased the SOC content and enriched the newly formed C in macro-aggregates. In addition, correlation analysis suggested a significant correlation between SOC and aggregate- associated C in differently sized aggregates. The 0.25-1 and 1-2mm aggregates were the main sites of SOC storage and were also the important indices of the soil C pool saturation.

Conventional tillage in comparison with NT significantly reduced macro-aggregates with a significant redistribution of aggregates - into micro-aggregates. Aggregate protected labile C and N were significantly greater for macro-aggregates, (>2000 and 250-2000 µm) than - micro-aggregates (53-250 and 20-53 µm) and greater for M than F indicating physical protection of labile C within macro-aggregates. No -tillage and M a lone each significantly increased soil aggregation and aggregate-associated C and N; however, NT and M together further improved soil aggregation and aggregate-protected C and N. The distribution pattern of soil microbial biomass associated with aggregates was likely governed by the size of aggregates, whereas the tillage effect was not significant at the aggregate-size scale. Tillage regimes that contribute to greater soil aggregation also will improve soil microbial activity to aid in crop production. Heterogeneous distribution of OC and microbial biomass may lead to "hotspots" of aggregation, and suggests that microorganisms associated with 1.0-2.0 mm aggregates are the most biologically active in the ecosystem.

Conventional tillage (CT) significantly reduces macroaggregates to smaller ones, thus aggregate stability was reduced by 35% compared with conservation system (CS), further indicating that tillage practices led to soil structural damage. The concentrations of SOC and other nutrients are also significantly higher under CS than CT, implying that CS may be an ideal enhancer of soil productivity in this subtropical ecosystem through improving soil structure which leads to the protection of SOM and nutrients, and the maintenance of higher nutrient content. The average concentration of particulate organic carbon (POC), dissolved organic carbon (DOC) and microbial biomass carbon (MBC) in organic manure plus inorganic fertilizer treatments (NP+S and NP+FYM) in 0-60 cm depth were increased by 64.9-91.9%, 42.5–56.9%, and 74.7–99.4%, respectively, over the CK treatment.

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