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To cite this article: Ankita Trivedi , Ranjan Bhattacharyya , D. R. Biswas , S. Das , T. K. Das , P. Mahapatra , D. K. Shahi & C. Sharma (2020) Long-term impacts of integrated nutrient management with equivalent nutrient doses to mineral fertilization on soil organic carbon sequestration in a sub-tropical Alfisol of India, Carbon Management, 11:5, 483-497, DOI: [10.1080/17583004.2020.1808766](https://doi.org/10.1080/17583004.2020.1808766)

To link to this article: <https://doi.org/10.1080/17583004.2020.1808766>



Published online: 26 Aug 2020.



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## Long-term impacts of integrated nutrient management with equivalent nutrient doses to mineral fertilization on soil organic carbon sequestration in a sub-tropical Alfisol of India

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### ABSTRACT

Limited evidence is available on carbon sequestration potential of long-term integrated nutrient management (INM) versus mineral fertilization, when equivalent amounts of nutrients were added. Hence, this study was carried out to understand the impact of 60 years of INM with adjusted nutrient doses and mineral fertilization in an Alfisol in a maize (*Zea mays* L.)-wheat (*Triticum aestivum* L.) system on soil organic carbon (SOC) sequestration in surface (0–30 cm) and deep (30–60 cm) soil layers. Conventional tillage was done twice before sowing of both maize and wheat using a spade. In farmyard manure (FYM) and lime treated plots (FYMP'K'L: plots with nitrogen (N) applied in terms of FYM; additional dose of phosphorus (P) and potassium (K) and lime) total SOC concentration was nearly 83% higher than unfertilized control plots. The FYMP'K'L plots had ~11% more total SOC concentration than plots treated with mineral fertilizer and lime (NPKL: recommended dose of N, P, K and lime) in the 0–30 cm soil layer. Labile C, including KMnO<sub>4</sub>-C, was more in plots with FYM than NPKL plots, whereas the recalcitrant C stock was more in NPKL than FYM treated plots. In the 0–60 cm soil layer, the labile C stock was highest in FYMP'K'L plots, but the recalcitrant C stock was highest in NPKL. Total SOC accumulation rate (over unfertilized control plots) was highest for FYMP'K'L plots (0.38 Mg ha<sup>-1</sup> year<sup>-1</sup>) in the surface soil layer, whereas SOC sequestration rate was highest in NPKL plots (0.18 Mg ha<sup>-1</sup> year<sup>-1</sup>) in the deep layer and in the 0–60 cm layer. Overall, although NPKL management practice had the highest C sequestration in the 0–60 cm layer, FYMP'K'L had the best CMI and labile C pools. Thus, resource poor farmers need not to use full doses of NPK and FYM for soil C management in the region.

### KEYWORDS

Labile and recalcitrant C pools; liming; farmyard manure application; carbon management index; alfisol

## 1. Introduction

Soil organic carbon (SOC) is a key parameter for nutrient management in tropical cropping systems [1]. Increased SOC stock is needed to decrease risks of erosion and degradation, hold water and nutrients, provide energy to soil microorganisms and improve soil structure and tilth. The SOC sequestration is also a major sink for atmospheric CH<sub>4</sub> and CO<sub>2</sub>. Researchers have found over the years that addition of organic matter with mineral fertilizers (integrated nutrient management, INM) has become an effective practice to sequester SOC and enhance its health and quality [2, 3]. So, SOC sequestration is a strategy to achieve food security through improvement in soil quality. Since, wheat (*Triticum aestivum*) and maize (*Zea mays*) are staple

crops, which are widely cultivated throughout the world, 60 years of long-term nutrient management practices can give an insight about the C sequestration rate in tropics and the alternative management that could be followed over the farmers' practice (mainly mineral fertilization).

Long-term balance between SOC addition and losses are reflected by SOC stocks. Ghosh *et al.* [4] found that in Inceptisols, SOC stock increased in all fertilized and unfertilized plots, with a maximum increase in 100% NPK with farmyard manure (NPKF) after 44 years of cropping and similar pattern was observed in KMnO<sub>4</sub> oxidizable C and particulate organic matter carbon. However, other researchers reported that under same soil and climatic conditions in a pearl millet (*Pennisetum*

*glaucum*)-wheat cropping system, only farmyard manure (FYM) delineated equal response to NPKF with regards to total SOC and its labile pools after 10 years of cropping [5]. In a rice (*Oryza sativa*)–wheat system, there was 17% higher SOC concentration under FYM addition at 20 t ha<sup>-1</sup> than mineral fertilizers (NPK) in the 0–15 cm layer after 32 years in an Inceptisol of the western Indo-Gangetic Plains [6].

Soil organic C is the combination of labile and recalcitrant C pools. The labile carbon pool is the fraction of total SOC with the most rapid turnover rates. The labile pool consists of living microbes and their products besides soil organic matter (SOM). At the same time, this pool fuels the soil food web and therefore greatly influences nutrient cycling for maintaining soil quality and its productivity [7]. Labile-C is mainly responsible for supplying nutrients to plants through mineralization processes, as well as providing energy and carbon to soil microorganisms [8]. The loss of C from a soil with a large carbon pool is of less consequence than the loss of the same amount of C from a soil already depleted of C or which started with a smaller total C pool. Similarly, the more a soil has been depleted of carbon, the more difficult it is to rehabilitate [9]. To account for this a carbon pool index (CPI) is calculated.

The SOC pool and the C lability directly influence soil physical, chemical and biological attributes and the self-organization capacity of soils [10]. Therefore, the integration of both SOC pool and C lability into the C management index (CMI), originally proposed by Blair *et al.* [9], can provide a useful parameter to assess the capacity of management systems in promoting soil quality [9]. The CMI is derived from the total SOC pool and C lability. The C lability index (LI) is the ratio of labile C to non-labile C. The index (CMI) provides a sensitive measure of the rate of change in soil C dynamics of systems relative to a more stable reference soil.

Recalcitrant carbon pools take more time to decompose and are not readily available to microorganisms [11]. Recalcitrant carbon pools show high variability in their chemical composition, stage of decomposition, and play role in soil functioning and health [12]. Humic substances represent 60%–80% of the total SOC with the highest concentration of humin, followed by fulvic acid or humic acid [13]. Among them, humin presents the greatest concentration in soil and strong resistance against microbial degradation due to higher

proportions of aromatic functions and bonds to mineral components [12]. Therefore, both recalcitrant and labile C pools can elucidate how the soil has been used and which management is adequate to increase carbon stocks, mainly in tropical conditions.

In the eastern Indo-Gangetic Plains, Mandal *et al.* [14] observed that the proportion of added C retention in soils was 6.9% of each additional t C input ha<sup>-1</sup> when organic manures were present, but only 4.2% in their absence. Thus, integrated nutrient management (INM) has been described as one of the effective means to sequester C in soils. INM is the way to improve soil physical structure and chemical fertility as well [2]. Liming in acid soils neutralizes acidity and it increases crop productivity. Liming improves soil aggregation as it contains Ca, increases microbial activity [15].

Despite many efforts in the past on these aspects, SOC sequestration in surface versus deep soil layers as affected by long-term application of manure and mineral fertilizers only compared with manure application in combination with mineral fertilizers (NPK) is poorly understood in tropical and subtropical climates. This is more so, if resource poor farmers could use equivalent amounts of nutrients (NPK) under INM. Researchers [16] have reported that carbon management index (CMI) as an early indicator of soil quality changes due to management practices. So, the specific objectives of the study were: (i) to assess SOC pools and CMI of INM versus mineral fertilization, when equivalent amounts of nutrients were added, and (ii) to evaluate the impacts of aforesaid management practices on SOC accumulation and sequestration rates in surface versus deep soil layers. The hypotheses were: (i) long-term INM (FYM application based on equivalent mineral N fertilization + adjusted doses of P and K) would increase SOC stock more than mineral fertilization especially in surface soils due to continuous FYM addition and (ii) CMI would be more under INM than mineral fertilization in an acidic subtropical Alfisol.

## 2. Materials and methods

### 2.1. Site

The study site is Kanke (Ranchi), Jharkhand, India at the Birsa Agricultural University, India (at 85°32' east longitude, 23°44' north latitude and 625 m altitude above the mean sea level). A permanent manorial trial (PMT) was initiated in 1956. The

**Table 1. Initial soil (0–15 cm) properties.**

Physical properties	Physico-chemical properties	Chemical properties
Mechanical composition	pH = 5.5±0.4	Organic C (%) = 0.52±0.05
i. Sand = 45.5±4.4%	C.E.C. (c mol (p+) kg <sup>-1</sup> ) = 10.5±0.9	Total N (%) = 0.067±0.006
i. Silt = 15.3±1.6%		
i. Clay = 36.4±3.7%		
Textural class : Clay loam		
Bulk density (Mg m <sup>-3</sup> ) = 1.45±0.02		

**Table 2. Treatment details.**

Treatments	Denoted as	Source of nutrient			Nutrient dose		
		N	P	K	Lime	FYM (Mg ha <sup>-1</sup> )	N:P <sub>2</sub> O <sub>5</sub> :K <sub>2</sub> O (kg ha <sup>-1</sup> )
Control	Control	–	–	–	–	–	0:0:0
N	N	Urea	–	–	–	–	110:0:0
FYM	FYM	FYM	–	–	–	22.0	0:0:0
NPK	NPK	Urea	SSP	MOP	–	–	110:90:70
FYM + P <sub>(A-X)</sub> + K <sub>(B-Y)</sub>	FYMP'K'	FYM	SSP	MOP	–	22.0	0:21.2:15.8
Lime + NPK	NPKL	Urea	SSP	MOP	LR	–	110:90:70
Lime + FYM + P <sub>(A-X)</sub> + K <sub>(B-Y)</sub>	FYMP'K'L	FYM	SSP	MOP	LR	22.0	0:21.2:15.8
Lime + N	NL	Urea	–	–	LR	–	110:0:0

Where, the subscripts A and B stand for full dose of P and K i.e. 90 kg P<sub>2</sub>O<sub>5</sub> and 70 kg K<sub>2</sub>O ha<sup>-1</sup>, respectively. X and Y represent the amount of P and K present in full dose of FYM applied on N basis to meet 110 kg N ha<sup>-1</sup>. LR denotes lime applied after calculating lime requirement.

climate is sub-tropical, with summer temperatures ranges from 20° C to 42° C. The winter temperature is mild. Average annual temperature of the area is around 23.1° C. December and January are the coolest months. The annual rainfall is about 1450 mm with >90% of it occurs between June to September. The soil (Paleustalf) of the experimental field is acidic red clay loam with available nutrients are in the range of low to medium. Kaolinite and illite are the dominant clay minerals. Some initial soil properties are given in Table 1.

## 2.2. Management of crops

A fixed crop rotation of maize in *kharif* and wheat in *rabi* was followed since 1956, but the varieties of both crops were changed for increased productivity. In 2015–16, the varieties grown were 'Suwan' and 'K 9107' for maize and wheat, respectively. Pesticides were applied as and when required to all plots for disease and pest management. Nutrients were applied as per the treatments in both crops. Residues of both crops were removed during harvesting. Grain yields of maize and wheat were recorded. Mean yield of both crops were not recorded during 1956–1959. Wheat yield data were not obtained during 1978, 1989 and 1990 due to hailstorm. Yield data of maize were not obtained during 1990 and 1995 due to severe pest and disease infestation.

## 2.3. Experimental design

There were eight treatments, each replicated thrice in a randomized block design (RBD) having

individual plot size of 4 m × 2.5 m area (Table 2). The levels of fertilizer applied were of 44 kg ha<sup>-1</sup> each of N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O through ammonium sulphate, single super phosphate (SSP) and muriate of potash (MOP), respectively. In the year 1969, during *Kharif* (rainy) season, the fertilizer dose was changed to 100:60:40 kg ha<sup>-1</sup> for N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O ha<sup>-1</sup>, respectively, and ultimately to 110, 90 and 70 kg ha<sup>-1</sup> from 1976 (Table 3). The recommended doses of mineral fertilizers for maize and wheat were 110:90:70 kg N:P<sub>2</sub>O<sub>5</sub>:K<sub>2</sub>O ha<sup>-1</sup>. Each year, these were applied to both crops. Nitrogen was applied in three splits through ammonium sulphate up to 1992 and thereafter through urea. Full doses of P and K were applied at wheat sowing as single superphosphate and muriate of potash, respectively. Farmyard manure (FYM) was applied at equivalent to 110 kg N ha<sup>-1</sup>. The FYM was added 15 days before sowing of maize and wheat. The amounts of P and K contained in FYM were taken into consideration in adjusting the final P and K doses, when applied in conjunction with mineral fertilizers. Lime was applied as per lime requirement following Shoemaker buffer method [17]. Amount varies from 3–4 Mg ha<sup>-1</sup> lime application once in every four years. Other details of the experiment can be found in Singh *et al.* [18] and Kumari *et al.* [19]. All agricultural operations were performed manually. Conventional tillage (up to 15 cm soil depth) was done twice before sowing of both maize and wheat using a spade. After manual harvesting, stubble heights were about 5 cm for maize and about 15 cm for wheat.

## 2.4. Soil sampling and processing

Triplicate soil samples were collected at wheat harvest (on 29 April 2016) using a core sampler (15 cm high and 7.6 cm diameter) from 0–15, 15–30, 30–45 and 45–60 cm depths from all plots. There was no gravel content in all depths. Collection of soils from deeper depths (>60 cm) was not possible due to presence of a hard pan. Soil bulk density values of all soil layers were computed. Treatment-wise triplicate samples were bulked to obtain one bulk samples per plot. There were 24 such bulk samples for one soil depth. Samples were air-dried under shade after collection, ground, and sieved to pass through a 4.75-mm sieve and preserved for further chemical analysis.

## 2.5. Soil analysis

The initial soil (0–15 cm) was collected in 1956 and analysed for bulk density using a core sampler. Walkley and Black [20] procedure was followed for organic C determination. The samples were archived until 1970. Cation exchange capacity (C.E.C.), pH and total N was measured following Jackson [21]. Soil texture was determined using the international pipette method.

Total SOC (of the air-dried samples taken in 2016) was determined using an isotopic ratio mass spectrometer (IRMS) (Isoprime; òsoprime UK) coupled with an Elemental Analyzer [22]. As the soils were acidic, there was no inorganic C present in soils. Organic carbon pools in the soil were estimated using the procedure of Chan *et al.* [23]. Total SOC stock was calculated using the given formula:

$$\begin{aligned} \text{Total SOC stock (Mg ha}^{-1}\text{)} \\ &= [\text{Total SOC (\%)/100}] \\ &\quad * \text{bulk density (Mg m}^{-3}\text{)} * \text{depth (m)} \\ &\quad * 10,000 \text{ (m}^2\text{ha}^{-1}\text{)} \end{aligned} \quad (1)$$

Total SOC accumulation rate was calculated using the following formula:

$$\begin{aligned} \text{Total SOC accumulation rate (Mg C ha}^{-1}\text{yr}^{-1}\text{)} \\ &= (\text{SOC}_{\text{treatment}} - \text{SOC}_{\text{UC}})/60 \end{aligned} \quad (2)$$

where,  $\text{SOC}_{\text{treatment}}$  and  $\text{SOC}_{\text{UC}}$  indicate the SOC stocks ( $\text{Mg ha}^{-1}$ ) of a given treatment and unfertilized control (UC) plots, respectively, in 2016 [24] and 60 indicates number of experiment years. Carbon sequestration rate was calculated using the given formula:

$$\begin{aligned} \text{C sequestration rate (Mg C ha}^{-1}\text{yr}^{-1}\text{)} \\ &= (\text{Recalcitrant C}_{\text{treatment}} - \text{Recalcitrant C}_{\text{UC}})/60 \end{aligned} \quad (3)$$

where,  $\text{Recalcitrant C}_{\text{treatment}}$  and  $\text{Recalcitrant C}_{\text{UC}}$  indicate the recalcitrant C stocks ( $\text{Mg ha}^{-1}$ ) of a given treatment and unfertilized control (UC) plots, respectively, in 2016 and 60 is the number of experimental years.

We also calculated treatment impacts on C accumulation in soils based on values expressed in terms of equivalent initial soil mass, to take into full consideration the impact of soil mass on C storage [25]. The equivalent soil mass was calculated to reach approximately 0–15, 15–30, 30–45 and 45–60 cm depth layers based on the bulk density of the samples taken before the start of the experiment for 0–15 cm layer (in 1956). For other layers, present bulk density of the unfertilized control plots was taken. Thus, for 0–15 cm soil layer, an equivalent mass of soil was 2175 Mg. For 15–30, 30–45 and 45–60 cm layers, equivalent soil masses were considered as 2370, 2415 and 2505 Mg, respectively. Briefly, equivalent mass of C was determined using the following equation [26].

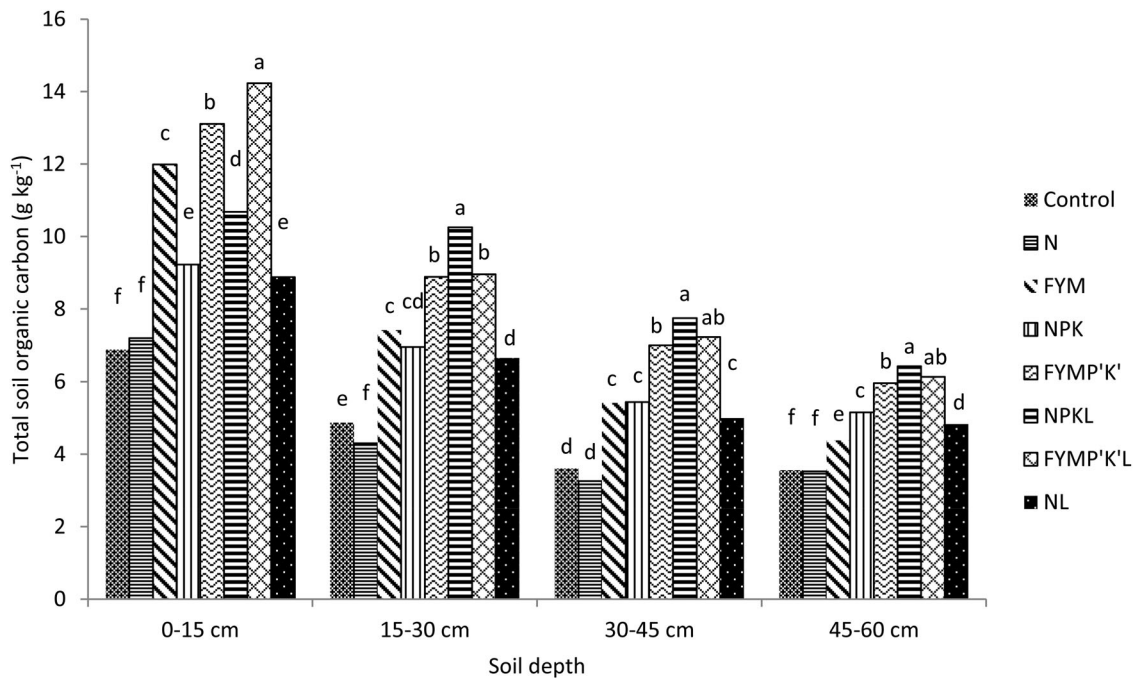
$$\begin{aligned} M_c &= [(\text{Conc}_c \times D_b \times \text{depth}) \\ &\quad - \text{Conc}_c (M_{\text{soil}} - M_{\text{soil, equiv}})] \times 10 \end{aligned} \quad (4)$$

where  $M_c$  equals equivalent SOC mass per unit area ( $\text{Mg ha}^{-1}$ );  $\text{Conc}_c$  equals SOC content ( $\text{kg Mg}^{-1}$ );  $D_b$  equals soil bulk density ( $\text{Mg m}^{-3}$ ); depth equals horizon depth (m);  $M_{\text{soil}}$  equals soil mass ( $\text{Mg m}^{-2}$ ) and  $M_{\text{soil, equiv}}$  equals equivalent soil mass ( $\text{Mg m}^{-2}$ ).

The permanganate-oxidisable organic carbon was determined following Tirol-Padre and Ladha [27]. Carbon management index (CMI) was calculated following Blair *et al.* [9]. Here the unfertilized control treatment was considered the reference soil.

## 2.6. Statistical analysis

All soil properties were analyzed using Analysis of Variance (ANOVA) for a randomized block design. Fisher's Least Significant Difference test was used as a *post hoc* mean separation test ( $p < 0.05$ ) using IASRI (Indian Agricultural Statistics Research Institute) portal. Correlation matrix was prepared using OPSTAT of Hisar Agricultural University [28]. All figures were drawn using Microsoft Office Excel (2007) of Microsoft, USA.



**Figure 1.** Total organic carbon concentration ( $\text{g kg}^{-1}$ ) in bulk soils at 0–15 cm, 15–30 cm, 30–45 cm and 45–60 cm depths as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol. Bars with similar lowercase letters within a soil depth are not significant at  $p < 0.05$  according to Tukey's HSD.

### 3. Results

#### 3.1. Total soil organic carbon concentration in bulk soils

In lime + FYM applied on  $110 \text{ kg N ha}^{-1}$  basis with adjusted doses of P and K (FYMP'K'L) treated plots in the 0–15 cm soil layer (soil surface), total SOC was nearly 107% more compared with the unfertilized control (UC) plots and 33% higher than full doses of NPK with lime (NPKL) after 60 years of maize-wheat cropping system (Figure 1). That total SOC value (under FYMP'K'L) was highest among all treatments. Organic manuring plots (FYM only) had 74 and 30% more total SOC concentration than UC and only minerally fertilized (NPK) plots, respectively, in the surface soil. In soil surface, all organic manure treated plots either alone or in combination with mineral fertilizers, and organic manure plots with lime alone or in combination with lime and mineral fertilizers contained larger total SOC than NPK, NPKL and UC plots due to major contribution of manure in the soil surface. In the sub-surface soil (15–30 cm depth) also the trend of total SOC concentration was similar, except for NPKL plots, where total SOC was the highest among all treatments (Figure 1). In the sub-surface layer, FYMP'K'L plots contained 16% less total SOC than NPKL plots. NPK and FYM treated plots had equal amount of total SOC concentrations in the 30–45 cm depth layer (Figure 1). In the 45–60 cm layer, total SOC distribution

followed almost same trend as that of 15–30 and 30–45 cm layers, with UC and N plots had equal total SOC concentrations (Figure 1). There was a clear-cut trend of decreasing total SOC concentrations in deeper soil depth layers. In plots with NL and NPKL, total SOC concentrations were significantly higher than respective plots without lime (N and NPK plots) in all soil layers. But in FYMP'K'L treated plots, liming played significant role in increasing SOC distribution only in the soil surface compared to FYMP'K' plots.

#### 3.2. Soil bulk density and total soil organic carbon stock

All organically manured plots had less soil bulk density compared with UC, NPK and NPKL plots (Table 4). In the soil surface, plots under FYMP'K'L had highest total SOC stock, which was ~30 and ~49% higher than NPKL and NPK treated plots, respectively (Table 4). Higher total SOC stock of FYMP'K'L plots in the surface soil was caused by addition of organic inputs over the years. Among all treatments, plots having FYM alone or in combination with mineral fertilizers and or lime showed greater total SOC stock compared with plots under NPK. But in the 15–30, 30–45 and 45–60 cm layers, NPKL treated plots contained highest total SOC content. In the 15–30 and 30–45 cm soil layers, total SOC contents were ~15 and 9% higher, respectively, than FYMP'K'L plots

**Table 3.** Details of the package of practices to raise the crops.

Practices	Maize Rainfed	Wheat Irrigated
Incorporation of farmyard manure	Last fortnight of May or 1 <sup>st</sup> week of June	Last fortnight of October or 1 <sup>st</sup> week of November
Field preparation, basal application of PK and 1 <sup>st</sup> split of N in rows, followed by line sowing of crops.	15–30 June Spacing (60 × 20 cm)	15–30 November Spacing (25 × 5 cm) continuous
Gap filling (if required to maintain optimum population)	1 <sup>st</sup> week of July	1st week of December One irrigation
Weeding, intercultural operations and irrigation	Generally not required	Generally not required
Earthing up	2 <sup>nd</sup> –3 <sup>rd</sup> week of July	Not applicable (N.A.)
Application of Furadon Granules	3 <sup>rd</sup> –4 <sup>th</sup> week of July	N.A.
Top dressing of Urea 2 <sup>nd</sup> split	Knee high stage (4 <sup>th</sup> week of July–1 <sup>st</sup> week of August)	Crown root initiation stage (4 <sup>th</sup> week of December–1 <sup>st</sup> week of January)
Top dressing of Urea 3 <sup>rd</sup> split	Tasseling stage (4 <sup>th</sup> week of August–1 <sup>st</sup> week of September)	Late tillering stage (4 <sup>th</sup> week of January–1 <sup>st</sup> week of February)
Plant protection measures	If required (insecticide)	If required (fungicide/insecticide)
Irrigation	N.A.	3–4
Harvesting	4 <sup>th</sup> week of September to 2 <sup>nd</sup> week of October	3–4 <sup>th</sup> week of April

**Table 4.** Bulk density and total soil organic carbon stock on equivalent depth basis in the 0–15, 15–30, 30–45 and 45–60 cm soil layers as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol.

Bulk density (Mg m <sup>-3</sup> )	Total soil organic carbon stock (Mg ha <sup>-1</sup> )							
	0–15 cm	15–30 cm	30–45 cm	45–60 cm	0–15 cm	15–30 cm	30–45 cm	45–60 cm
Control	1.57 ± 0.03a	1.58 ± 0.03a	1.61 ± 0.04a	1.67 ± 0.03a	16.22 ± 1.67f	11.55 ± 1.17d	8.72 ± 0.81d	8.91 ± 0.85e
N	1.57 ± 0.02a	1.57 ± 0.03a	1.61 ± 0.04a	1.66 ± 0.04ab	16.96 ± 1.71f	10.13 ± 0.99e	7.87 ± 0.82d	8.78 ± 0.83e
FYM	1.43 ± 0.02c	1.49 ± 0.03bc	1.56 ± 0.03bcd	1.62 ± 0.03cd	25.73 ± 2.48c	16.59 ± 1.75c	12.68 ± 1.17c	10.63 ± 1.01d
NPK	1.48 ± 0.03b	1.52 ± 0.04b	1.57 ± 0.04bc	1.63 ± 0.03bcd	20.47 ± 2.11e	15.87 ± 1.55c	12.81 ± 1.25c	12.62 ± 1.22c
FYMP'K'	1.45 ± 0.03bc	1.50 ± 0.03b	1.54 ± 0.02de	1.62 ± 0.04cd	28.57 ± 2.91b	20.02 ± 1.91b	16.17 ± 1.57b	14.47 ± 1.37b
NPKL	1.47 ± 0.02b	1.51 ± 0.03b	1.55 ± 0.03cde	1.60 ± 0.04d	23.57 ± 2.45d	23.29 ± 2.33a	18.02 ± 1.91a	15.44 ± 1.61a
FYMP'K'L	1.43 ± 0.03c	1.50 ± 0.02b	1.53 ± 0.04e	1.61 ± 0.04d	30.55 ± 3.01a	20.21 ± 1.98b	16.59 ± 1.59b	14.84 ± 1.52ab
NL	1.55 ± 0.02ab	1.57 ± 0.03a	1.58 ± 0.03b	1.65 ± 0.03abc	20.62 ± 2.09e	15.70 ± 1.42c	11.85 ± 1.21c	11.96 ± 1.12c
LSD (P < 0.05)	0.02	0.03	0.02	0.03	1.46	1.15	1.39	0.77

Means (± SD) with similar lower-case letters within a column are not significantly different at  $p < 0.05$  according to Fisher's LSD test.  $n = 3$ .

(Table 4). In the 45–60 cm layer, plots under NPKL and FYMP'K'L had identical total SOC contents. In all soil layers, lime treated plots contained more total SOC stock compared with plots without lime, other nutrients be the same. Nonetheless, in the 0–60 cm layer, FYMP'K'L plots contained significantly more total SOC stock than rest of the treatments. The total SOC stock of FYMP'K'L plots was 33% greater compared with plots under NPK in the 0–60 cm layer.

Total soil organic C storage was also computed on equivalent soil mass basis (Table 5). For the equivalent mass of 2175 Mg soil (approximately 0–15 cm soil layer), plots under FYMP'K'L had significantly higher SOC storage than all other plots, except FYMP'K' plots (Table 5). However, for the equivalent mass of 2370, 2415 and 2505 Mg soil (approximately to reach 15–30, 30–45 and 45–60 cm soil layers, respectively), NPKL treated plots had higher SOC than other treatments, except FYMP'K' and FYMP'K'L plots for the equivalent mass of 2415 and 2505 Mg soil. Overall, the trends of SOC storages under different treatments on equivalent depth and equivalent mass basis were similar (Tables 4 and 5).

### 3.3. Permanganate oxidizable soil carbon

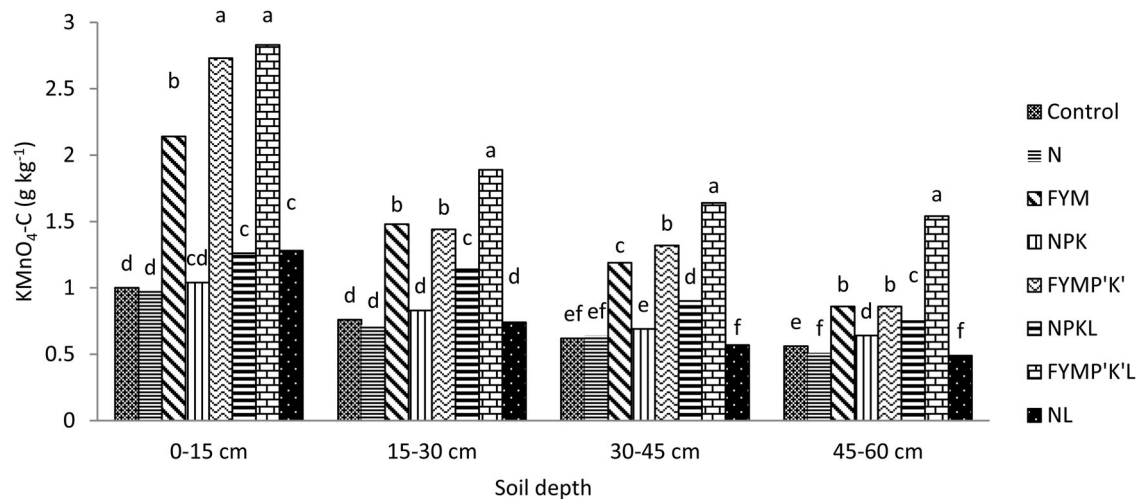
In the soil surface, FYMP'K'L plots contained 125 and 172% more permanganate oxidizable carbon (KMnO<sub>4</sub>-C) than NPKL and NPK plots, respectively (Figure 2). Plots containing organic manure showed higher KMnO<sub>4</sub>-C than UC and minerally fertilized plots, irrespective of lime integration in the treatments. In the surface soil, whenever lime was integrated with FYM and/or mineral fertilizer, it showed higher KMnO<sub>4</sub>-C than plots without lime, rest nutrients and their sources being the same (Figure 2). In the 15–30, 30–45 and 45–60 cm soil layers, the trends were comparable (Figure 2). In the lower soil depths, KMnO<sub>4</sub>-C showed a decreasing trend.

Lability index (LI) was maximum for FYMP'K' plots followed by the plots with FYM in the 0–15 cm layer (Table 6). Plots containing lime had greater LI than plots devoid of lime, although rest nutrients and their sources being same in the soil surface. The plots with FYMP'K'L had 200 and 146% higher CMI compared with plots under NPK and NPKL (Table 6), respectively, in the soil surface. For all deeper soil layers, LI and CMI were the highest in FYMP'K'L plots (Table 6). However,

**Table 5.** Total soil organic carbon stock on equivalent mass depth of approximately 0–15, 15–30, 30–45 and 45–60 cm soil layers as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol.

Treatments	Total soil organic carbon storage (Mg ha <sup>-1</sup> ) on equivalent mass of			
	2175 Mg soil	2370 Mg soil	2415 Mg soil	2505 Mg soil
Control	14.97 ± 1.56e	11.55 ± 1.12d	8.72 ± 0.82c	8.91 ± 0.95c
N	15.66 ± 1.49e	10.19 ± 1.08d	7.87 ± 0.79c	8.84 ± 0.89c
FYM	26.08 ± 2.74b	17.58 ± 1.61c	13.09 ± 1.29b	10.97 ± 1.05bc
NPK	20.07 ± 2.01d	16.47 ± 1.64c	13.14 ± 1.31b	12.93 ± 1.30b
FYMP'K'	28.51 ± 2.78ab	21.07 ± 1.99b	16.91 ± 1.75a	14.93 ± 1.55a
NPKL	23.24 ± 2.31c	24.32 ± 2.37a	18.72 ± 1.91a	16.11 ± 1.64a
FYMP'K'L	30.79 ± 3.02a	21.23 ± 2.08b	17.46 ± 1.72a	15.38 ± 1.49a
NL	19.65 ± 2.01d	15.78 ± 1.62c	12.07 ± 1.18b	12.10 ± 1.17b
LSD (P < 0.05)	2.33	1.97	1.66	2.01

Means (± SD) with similar lower-case letters within a column are not significantly different at  $p < 0.05$  according to Fisher's LSD test.  $n = 3$ .

**Figure 2.** Permanganate oxidizable soil organic carbon (g kg<sup>-1</sup>) in the bulk soils at 0–15 cm, 15–30 cm, 30–45 cm and 45–60 cm depths as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol. Bars with similar lowercase letters within a soil depth are not significant at  $p < 0.05$  according to Tukey's HSD.

carbon pool index (CPI) was the highest for NPKL plots in all deeper soil depth layers.

### 3.4. Oxidizable soil organic carbon fractions in bulk soils

In the surface soil, labile C was highest for FYMP'K' and FYMP'K'L treated plots and was least for N plots (Table 7). However, in the 15–30 cm layer, NPKL treated plots contained the highest recalcitrant C stock among all treatments (Table 8). All treatments had higher amounts of recalcitrant SOC stocks compared with labile C, except for FYM treated plots, where labile SOC stock was dominant in the 0–60 cm depth layer.

### 3.5. Carbon accumulation and carbon sequestration rate in bulk soils

In the 0–30 cm soil layer, organic manure treated plots had more labile C stock than UC and mineral fertilized plots. However, the highest labile C stock was found in FYMP'K' plots (Table 9). The FYMP'K' plots contained ~27 and 74% more labile

C stock compared with NPKL and NPK plots, respectively. Whereas in FYMP'K'L plots, labile C stock was 23 and 68% higher than NPKL and NPK plots, correspondingly, in the 0–30 cm soil depth layer. Liming in mineral fertilized plots had significant impact on improving labile C stock in that soil layer. The NPKL and NL plots contained ~37 and 68% higher labile C stock than NPK and N plots, respectively. Highest recalcitrant C stock was obtained in NPKL plots in the 0–30 cm soil layer (Table 9). The plots under NPK contained ~25% higher recalcitrant C stock compared with plots under FYM whereas, recalcitrant C stocks of NPKL and FYMP'K'L plots were comparable. However, total C stock in the 0–30 cm soil layer was the highest for FYMP'K'L plots and was 8 and 40% more than NPKL and NPK plots, respectively. Plots with NPKL contained greater total SOC stock in the 0–30 cm layer compared with FYM plots, but had lower total SOC stock than FYMP'K'L plots. Total SOC accumulation rate (as calculated over UC plots after 60 years) was the highest for FYMP'K'L plots (0.38 Mg ha<sup>-1</sup> yr<sup>-1</sup>) in the 0–30 cm layer, whereas C sequestration rate (as calculated over UC plots



**Table 6.** Lability index (LI), carbon pool index (CPI) and carbon management index (CMI) in 0–15, 15–30, 30–45 and 45–60 cm soil layers as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol.

Treatments	0–15 cm			15–30 cm			30–45 cm			45–60 cm		
	LI	CPI	CMI	LI	CPI	CMI	LI	CPI	CMI	LI	CPI	CMI
Control	1.00 ± 0c	1.00 ± 0f	100.0 ± 0c	1.00 ± 0b	1.00 ± 0e	100.0 ± 0d	1.00 ± 0d	1.00 ± 0d	100.0 ± 0de	1.00 ± 0c	1.00 ± 0f	100.0 ± 0de
N	0.92 ± 0.08cd	1.05 ± 0.10f	95.9 ± 10.1c	1.10 ± 0.12b	0.88 ± 0.09e	97.1 ± 10.1d	1.21 ± 0.11bc	0.91 ± 0.09d	106.3 ± 10.1d	0.91 ± 0.09c	0.99 ± 0.10f	89.8 ± 9.5ef
FYM	1.29 ± 0.12b	1.74 ± 0.16c	224.7 ± 23.5b	1.35 ± 0.14a	1.53 ± 0.14c	206.3 ± 20.7b	1.35 ± 0.13ab	1.50 ± 0.14c	203.4 ± 19.7b	1.29 ± 0.13b	1.23 ± 0.12e	159.1 ± 15.2b
NPK	0.75 ± 0.06d	1.34 ± 0.13e	100.6 ± 10.5c	0.74 ± 0.07c	1.43 ± 0.15cd	104.8 ± 10.8d	0.72 ± 0.07e	1.53 ± 0.14c	106.2 ± 9.9d	0.75 ± 0.07d	1.45 ± 0.15c	109.1 ± 10.2d
FYMP <sup>a</sup> K	1.55 ± 0.14a	1.91 ± 0.18b	296.1 ± 30.2a	1.05 ± 0.11b	1.83 ± 0.18b	191.8 ± 10.1b	1.13 ± 0.11cd	1.95 ± 0.20b	218.8 ± 22.7b	0.90 ± 0.08c	1.68 ± 0.17b	149.9 ± 15.7b
NPkL	0.79 ± 0.08cd	1.55 ± 0.16d	122.6 ± 11.9c	0.68 ± 0.07c	2.12 ± 0.22a	143.0 ± 13.9c	0.64 ± 0.06e	2.16 ± 0.22a	136.3 ± 13.1c	0.71 ± 0.07de	1.81 ± 0.18a	127.6 ± 12.2c
FYMP <sup>a</sup> KL	1.47 ± 0.15ab	2.08 ± 0.20a	302.2 ± 29.7a	1.45 ± 0.14a	1.84 ± 0.19b	267.9 ± 27.5a	1.41 ± 0.14a	2.01 ± 0.22ab	282.9 ± 30.1a	1.79 ± 0.17ae	1.73 ± 0.18b	308.2 ± 29.8a
NL	0.99 ± 0.09c	1.29 ± 0.13e	128.2 ± 14.1c	0.68 ± 0.07c	1.37 ± 0.13d	93.5 ± 9.8d	0.63 ± 0.06e	1.40 ± 0.13c	85.7 ± 8.9e	0.60 ± 0.06e	1.36 ± 0.14d	81.4 ± 8.5f
LSD (P < 0.05)	0.21	0.13	33.9	0.11	0.14	18.6	0.19	0.16	18.4	0.14	0.09	17.5

Means (± SD) with similar lower-case letters within a column are not significantly different at  $p < 0.05$  according to Fisher's LSD test.  $n = 3$ .

**Table 7.** Labile and recalcitrant carbon pools in bulk soils of 0–15, 15–30, 30–45 and 45–60 cm soil layers as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol.

Treatments	0–15 cm			15–30 cm			30–45 cm			45–60 cm		
	Labile C	Recalcitrant C	Recalcitrant C	Labile C	Recalcitrant C	Recalcitrant C	Labile C	Recalcitrant C	Recalcitrant C	Labile C	Recalcitrant C	
Control	3.81 ± 0.37e	3.07 ± 0.31e	3.02 ± 0.29e	1.85 ± 0.02e	3.02 ± 0.29e	3.02 ± 0.29e	1.73 ± 0.19e	1.88 ± 0.19e	1.88 ± 0.19e	1.02 ± 0.11g	2.53 ± 0.24d	
N	3.15 ± 0.33f	4.05 ± 0.44d	2.77 ± 0.28e	1.53 ± 0.16f	2.77 ± 0.28e	2.77 ± 0.28e	1.38 ± 0.14f	1.88 ± 0.18e	1.88 ± 0.18e	0.61 ± 0.06h	2.92 ± 0.28c	
FYM	6.23 ± 0.64b	5.76 ± 0.61a	3.55 ± 0.37cd	3.87 ± 0.39b	3.55 ± 0.37cd	3.55 ± 0.37cd	3.12 ± 0.33b	2.30 ± 0.25d	2.30 ± 0.25d	1.97 ± 0.21c	2.40 ± 0.24d	
NPK	4.06 ± 0.41de	5.16 ± 0.48b	4.07 ± 0.38c	2.89 ± 0.31d	4.07 ± 0.38c	4.07 ± 0.38c	1.94 ± 0.20de	3.50 ± 0.35b	3.50 ± 0.35b	1.22 ± 0.12f	3.94 ± 0.41a	
FYMP <sup>a</sup> K	8.13 ± 0.80a	4.98 ± 0.52b	4.69 ± 0.48b	4.20 ± 0.41a	4.69 ± 0.48b	4.69 ± 0.48b	3.30 ± 0.31ab	3.70 ± 0.35b	3.70 ± 0.35b	1.78 ± 0.18d	4.18 ± 0.41a	
NPkL	5.43 ± 0.55c	5.25 ± 0.51b	6.10 ± 0.60a	4.15 ± 0.39a	6.10 ± 0.60a	6.10 ± 0.60a	2.75 ± 0.29c	5.00 ± 0.47a	5.00 ± 0.47a	2.51 ± 0.26a	3.93 ± 0.39a	
FYMP <sup>a</sup> KL	8.10 ± 0.83a	6.13 ± 0.60a	5.02 ± 0.48b	3.94 ± 0.38b	5.02 ± 0.48b	5.02 ± 0.48b	3.54 ± 0.35a	3.69 ± 0.37b	3.69 ± 0.37b	2.22 ± 0.20b	3.93 ± 0.40a	
NL	4.32 ± 0.43d	4.56 ± 0.47c	3.05 ± 0.31de	3.62 ± 0.37c	3.05 ± 0.31de	3.05 ± 0.31de	2.10 ± 0.22d	2.90 ± 0.28c	2.90 ± 0.28c	1.60 ± 0.15e	3.23 ± 0.31b	
LSD (P < 0.05)	0.38	0.38	0.53	0.11	0.53	0.53	0.35	0.36	0.36	0.09	0.27	

Means (± SD) with similar lower-case letters within a column are not significantly different at  $p < 0.05$  according to Fisher's LSD test.  $n = 3$ .

**Table 8.** Labile and recalcitrant soil organic carbon stock (on equivalent depth basis) in the 0–15, 15–30, 30–45 and 45–60 cm soil layers as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol.

Treatments	Soil organic carbon stock (Mg ha <sup>-1</sup> )							
	0–15 cm		15–30 cm		30–45 cm		45–60 cm	
	Labile	Recalcitrant	Labile	Recalcitrant	Labile	Recalcitrant	Labile	Recalcitrant
Control	8.98 ± 0.91e	7.23 ± 0.66f	4.39 ± 0.41e	7.15 ± 0.73de	4.18 ± 0.42c	4.54 ± 0.44d	2.56 ± 0.27g	6.35 ± 0.66e
N	7.42 ± 0.71f	9.53 ± 0.91e	3.61 ± 0.35f	6.53 ± 0.66e	3.33 ± 0.31d	4.54 ± 0.46d	1.51 ± 0.14h	7.27 ± 0.72d
FYM	13.36 ± 1.28b	12.37 ± 1.17ab	8.65 ± 0.77bc	7.94 ± 0.76d	7.30 ± 0.77a	5.38 ± 0.51d	4.79 ± 0.48c	5.84 ± 0.59e
NPK	9.02 ± 0.92e	11.46 ± 1.13c	6.59 ± 0.64d	9.28 ± 0.88c	4.57 ± 0.45c	8.24 ± 0.85b	2.99 ± 0.33f	9.63 ± 1.01ab
FYMP'K'	17.72 ± 1.85a	10.85 ± 1.09cd	9.45 ± 0.91a	10.57 ± 1.02b	7.63 ± 0.74a	8.54 ± 0.81b	4.32 ± 0.45d	10.15 ± 1.07a
NPKL	11.99 ± 1.22c	11.58 ± 1.14bc	9.43 ± 0.97a	13.86 ± 1.34a	6.39 ± 0.62b	11.6 ± 1.19a	6.02 ± 0.57a	9.43 ± 0.92b
FYMP'K'L	17.39 ± 1.67a	13.16 ± 1.29a	8.89 ± 0.79b	11.3 ± 1.07b	8.12 ± 0.85a	8.47 ± 0.87b	5.35 ± 0.49b	9.49 ± 0.95ab
NL	10.04 ± 1.01d	10.58 ± 1.07d	8.52 ± 0.82c	7.18 ± 0.76de	4.98 ± 0.50c	6.87 ± 0.69c	3.96 ± 0.42e	8.00 ± 0.75c
LSD (P < 0.05)	0.85	0.83	0.24	1.19	0.83	0.84	0.23	0.71

Means with similar lower-case letters within a column are not significantly different at  $p < 0.05$  according to Fisher's LSD test.  $n = 3$ .

after 60 years) was highest for plots treated with NPKL (0.18 Mg ha<sup>-1</sup> yr<sup>-1</sup>) although C sequestration rate of FYMP'K'L plots was similar to NPKL plots in this soil layer.

In the 30–60 cm soil layer, recalcitrant C stock was substantially higher than labile C stock across all treatments. The FYMP'K'L plots had the highest labile C stock in that soil layer (Table 9). The labile C stock under FYMP'K'L plots was 100, 9 and 78% more than UC, NPKL and NPK plots, respectively. However, recalcitrant C stock was highest for NPKL (21.05 Mg ha<sup>-1</sup>) plots, followed by FYMP'K' and FYMP'K'L in the deep soil layer (30–60 cm depth).

Total SOC stock was also the highest under NPKL (33.46 Mg ha<sup>-1</sup>), followed by plots with FYMP'K'L and FYMP'K'. SOC accumulation rates (as calculated over UC plots after 60 years) were similar in FYMP'K'L, NPKL and FYMP'K' plots in the 0–60 cm soil depth layer (Figure 3). Plots treated with N only had negative SOC accumulation rate in the 0–60 cm layer. However, C sequestration rate was highest in NPKL plots, followed by plots under FYMP'K'L and FYMP'K' (Figure 3). In the 0–60 cm soil depth, plots with FYM only sequestered less C than NPK plots. Liming enhanced C sequestration rate of both mineral fertilized and organically manured plots (Figure 3).

### 3.6. Mean crop yields

The mean wheat yield data (for 53 years) and mean maize yield data (for 54 years) are presented in Table 10. Reasons due to non-availability of yield data for some years are mentioned in materials and methods section. Results reveal that NPKL treated plots had significantly higher mean maize and wheat yields than all other treatments (Table 10). Plots under NPKL had ~28 and 36% more mean maize yield than FYMP'K and FYMP'K'L treated plots, respectively. Similarly, NPKL treated plots had ~48 and 65% higher mean wheat yield

compared with FYMP'K and FYMP'K'L treated plots, respectively. The FYMP'K' and FYMP'K'L treatments yielded similarly in both crops (Table 10). Interestingly, plots under FYM had ~99 and 24% more mean maize and wheat yields, respectively, than NPK treated plots, and plots under NL yielded 62% more maize than NPK. However, only N treated plots recorded significantly lower wheat yield than the unfertilized control plots. This might be due to buildup of residual acidity because of repeated application of N fertilizer in already acidic soil.

## 4. Discussion

### 4.1. Effect of long-term fertilization and liming on bulk density

In the surface soil, bulk density in plots under FYMP'K'L and FYM was similar and least among all treatments. This could be due to better soil aggregation, increased porosity and presence of more SOM with all FYM-added plots than without FYM [4]. Furthermore, organic matter is comprised of mainly fibrous crop residues, which increase the soil volume and decrease soil bulk density [29]. Whereas, in all the mineral fertilized treatments bulk density increased in the soil surface, primarily due to reduced porosity and decreased SOM [2]. In the deep soil layers, bulk density values increased, irrespective of treatments, mainly due to overlying soil pressure, reduced SOM and increased compaction in deeper soil layers [30].

### 4.2. Long-term fertilization and liming effect on total soil organic carbon stock

Higher total SOC stock of FYMP'K'L plots in the surface soil was caused by addition of organic inputs over the years. Better aggregation and encapsulation of C in the soil aggregates under manure-amended plots might also be the reason of

**Table 9. Carbon accumulation and carbon sequestration rate in 0–30 and 30–60 cm depths as affected by 60 years of fertilization and liming under maize-wheat based cropping system in an Alfisol.**

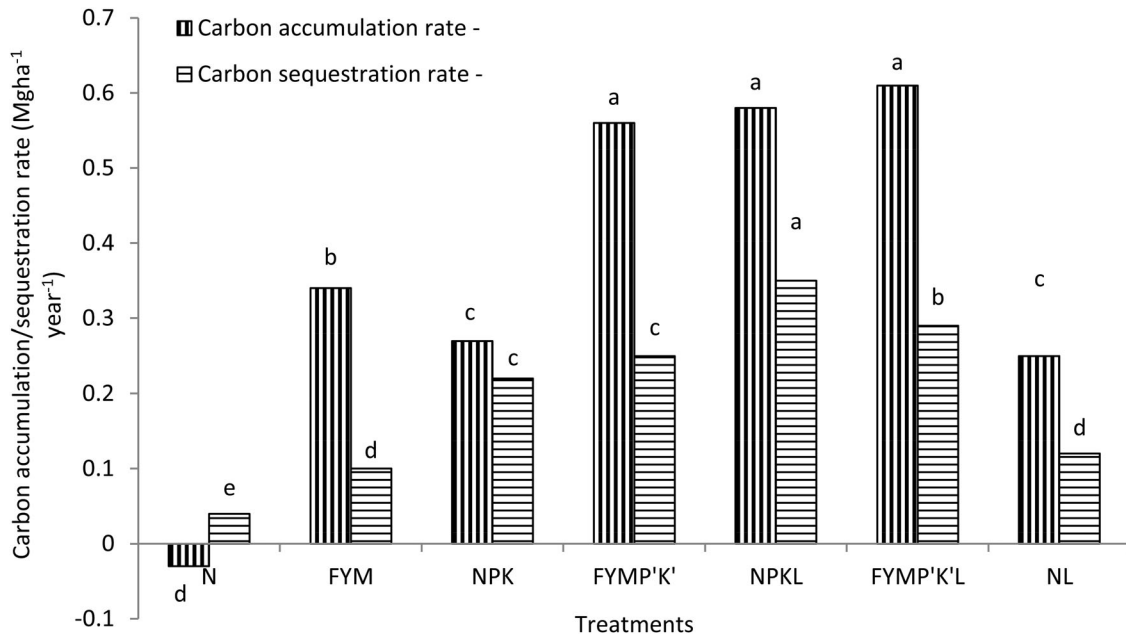
Treatments	0–30 cm						30–60 cm					
	Carbon stock (Mg ha <sup>-1</sup> )			Total SOC accumulation rate (Mg ha <sup>-1</sup> year <sup>-1</sup> )			Carbon stock (Mg ha <sup>-1</sup> )			Total SOC accumulation rate (Mg ha <sup>-1</sup> year <sup>-1</sup> )		
	Labile	Recalcitrant	Total	C sequestration rate (Mg ha <sup>-1</sup> year <sup>-1</sup> )	Total	Labile	Recalcitrant	Total	C sequestration rate (Mg ha <sup>-1</sup> year <sup>-1</sup> )	Total	C sequestration rate (Mg ha <sup>-1</sup> year <sup>-1</sup> )	
Control	13.38 ± 1.35f	14.39 ± 1.41e	27.76 ± 2.81e	–	–	6.74 ± 0.07d	10.89 ± 1.05d	17.63 ± 1.87e	–	–	–	
N	11.03 ± 1.07g	16.06 ± 1.58d	27.09 ± 2.67e	–0.01 ± 0.00e	–0.03 ± 0.00d	4.84 ± 0.04e	11.81 ± 1.11d	16.65 ± 1.58e	0.02 ± 0.00d	0.02 ± 0.00d	–0.03 ± 0.00d	
FYM	22.02 ± 2.17c	20.31 ± 1.97b	42.32 ± 4.12c	0.24 ± 0.02c	0.10 ± 0.01b	12.09 ± 1.11b	11.22 ± 1.07d	23.31 ± 2.44d	0.01 ± 0.00d	0.01 ± 0.00d	0.33 ± 0.03b	
NPK	15.60 ± 1.51e	20.74 ± 2.01b	36.34 ± 3.43d	0.14 ± 0.01d	0.11 ± 0.01b	7.56 ± 0.08d	17.87 ± 1.78b	25.43 ± 2.58c	0.12 ± 0.01b	0.12 ± 0.01b	0.27 ± 0.03bc	
FYMP'K'	27.17 ± 2.94a	21.42 ± 2.07b	48.59 ± 5.07b	0.35 ± 0.04b	0.12 ± 0.01b	11.95 ± 1.27b	18.69 ± 1.77b	30.64 ± 2.99b	0.13 ± 0.01b	0.13 ± 0.01b	0.57 ± 0.05a	
NPKL	21.42 ± 2.11c	25.44 ± 2.31a	46.86 ± 4.87b	0.32 ± 0.03b	0.18 ± 0.02a	12.41 ± 1.21b	21.05 ± 2.01a	33.46 ± 3.27a	0.17 ± 0.02a	0.17 ± 0.02a	0.58 ± 0.06a	
FYMP'K'L	26.28 ± 2.71b	24.47 ± 2.25a	50.76 ± 5.18a	0.38 ± 0.04a	0.17 ± 0.02a	13.48 ± 1.33a	17.95 ± 1.85b	31.43 ± 3.12b	0.23 ± 0.02b	0.12 ± 0.01b	0.61 ± 0.06a	
NL	18.56 ± 1.74d	17.76 ± 1.71c	36.33 ± 3.55d	0.14 ± 0.01d	0.06 ± 0.00c	8.94 ± 0.85c	14.87 ± 1.43c	23.81 ± 2.36cd	0.10 ± 0.01cd	0.07 ± 0.01c	0.24 ± 0.02c	
LSD (P < 0.05)	0.80	1.45	1.88	0.03	0.02	0.81	1.09	1.70	0.02	0.01	0.07	

Means (± SD) with similar lower-case letters within a column are not significantly different at  $p < 0.05$  according to Fisher's LSD test.  $n = 3$ .

enriched SOC stock [4, 24, 31]. But in the lower soil depths, increased SOC stock in NPKL plots might be associated with the more biomass production and turnover over the years [32]. In the entire 0–60 cm soil layer, highest total SOC stock of FYMP'K'L plots could be due to integrated effect of (i) increased C inputs (ii) increased biomass production and (iii) impact of liming on better soil aggregation [4]. It is to be mentioned that unlike other studies, full doses of NPK and recommended doses of FYM (which is 10 Mg ha<sup>-1</sup>) were not applied over the years in this experiment. Here, FYM was applied based on equivalent recommended N fertilization of both crops and then, P and K doses were adjusted in the FYMP'K' and FYMP'K'L plots.

### 4.3. Impact of long-term fertilization and liming on oxidizable soil organic fractions in bulk soils

Increased labile C in FYMP'K' and FYMP'K'L plots might be due to addition of well decomposed manure and high rate of organic matter decomposition in the surface layer in this sub-tropical climate [31]. In the 0–30 cm soil layer, labile C stocks of different treatments positively and significantly correlated with mean (of 54 years) maize yields ( $r = 0.862^{**}$ ) (Table 11), but not the mean (of 53 years) wheat yields of all treatments (yield data are given in Table 10). In the 0–30 cm soil layer, a positive and significant correlation of maize yield with labile C stock might be due to the reason that labile C pool was capable of supplying essential nutrients for crop growth. Lower labile C in NPK plots compared with FYM treated plots (FYM, FYMP'K' and FYMP'K'L) might be due to lower above-ground biomass returning to soils. The increased labile C in NPKL plots than NPK plots could be owing to selective preservation of recalcitrant compounds. Liming was assumed to contribute Ca<sup>2+</sup>, and organic matter complexation by Ca<sup>2+</sup> probably contributed to C stabilization in the labile pools. Thus, higher Ca<sup>2+</sup> content in the NPKL than NPK plots probably contributed to better SOC distribution in the labile pools. The additional OM input under integrated nutrient management could enhance POM–C [33], because FYM could increase the root biomass, root exudates (ligno-cellulose residues), and microbial biomass debris [34]. Same could be the reason for NPKL plots. Decreased POM–C with increased soil depth under integrated nutrient management might be due to slow and low translocation of leaf litter and



**Figure 3.** Total SOC accumulation rate ( $\text{Mg ha}^{-1} \text{ year}^{-1}$ ) and C sequestration rate ( $\text{Mg ha}^{-1} \text{ year}^{-1}$ ) in the 0–60 cm soil layer as affected by 60 years of fertilization and liming under maize-wheat cropping system in an Alfisol. Bars with similar lowercase letters are not significant at  $p < 0.05$  according to Tukey's HSD.

applied FYM. The increasing trend of  $\text{KMnO}_4\text{-C}$  content with fertilization could be explained by increased C inputs from stubble, root biomass and rhizodeposition [35].

Apart from manure, presence of partially-decomposed plant material might also lead to the increased recalcitrant C content under integrated nutrient management [36]. The higher recalcitrant C retention in integrated nutrient management over NPKL plots was probably because the manure was already partly decomposed and contained a lower proportion of chemically recalcitrant organic compounds [37]. Recalcitrant C in plots treated with N alone might be due to lower net primary productivity of these plots, having least biomass production and high level of soil acidity ( $\text{pH} = 4.48$ , data not shown). Lower acidity in soils might be the cause of reduced microbial activity. In the surface soil layer, increased recalcitrant C pool in plots treated with FYM might be caused due to the composition of FYM and presence of partially decomposed product of organic matter [38, 39]. Belay-Tedla *et al.* [40] also reported that FYM application resulted in an augmented lignin. However, positive correlations between recalcitrant C stocks and yield of both crops signified that recalcitrant C was also beneficial for crop growth in this agroecosystem [41]. Similarity, C lability in both FYM and NPKL treated plots in the sub-surface soil might be due to increased root biomass and increased C inputs in NPKL and FYM plots, respectively [42]. Additionally, increased mineralization of organic matter due to liming might have caused

increased lability of C in the NPKL plots. But increased recalcitrance of C in NPKL plots, among all treatments, was associated with presence of partially decomposed root biomass and lesser influence of FYM in contributing recalcitrant C in the sub-surface soil. A good correlation between recalcitrant and labile C stocks (Table 11) indicated their inter conversion [43], as some labile C may get incorporated into some stable C pools (as in case of humus synthesis) or recalcitrant pool may get mineralized to labile C through microbial action depending upon the climatic condition [12]. In the deeper soil layer, increased contribution of recalcitrant C to total SOC was due to lesser microbial activity and slower turnover of SOM.

#### 4.4. Long-term fertilization and liming impacts on total organic carbon accumulation versus carbon sequestration rates in bulk soils

In the 0–30 cm soil layer, higher C accumulation rate in FYMP'K'L treated plots might have been due to combined impact of external organic C inputs, C inputs through root biomass and better soil physico-chemical condition responsible for accumulating more C in this soil layer [44, 45]. Greater C accumulation rates in FYMP'K'L, FYMP'K' plots than only FYM plots imply role of other nutrients that were complementary to FYM in terms of accumulating more C. Among minerally fertilized plots, NPKL plots were able to accumulate more C due to greater root biomass caused by higher biomass production [46]. Higher amount of

**Table 10. Mean grain yields of maize and wheat of the long-term experiment.**

Treatments	Mean (of 54 years) maize yield (Mg ha <sup>-1</sup> )	Mean (of 53 years) wheat yield (Mg ha <sup>-1</sup> )
Control	0.52 ± 0.06e	0.71 ± 0.06d
N	0.44 ± 0.05e	0.17 ± 0.05e
FYM	2.90 ± 0.32b	2.30 ± 0.25b
NPK	1.46 ± 0.14d	1.86 ± 0.19c
FYMP'K'	3.11 ± 0.32b	2.44 ± 0.25b
NPKL	3.97 ± 0.41a	3.61 ± 0.38a
FYMP'K'L	2.91 ± 0.28b	2.18 ± 0.21b
NL	2.36 ± 0.24c	1.67 ± 0.18c

Means (± SD) with similar lower-case letters within a column are not significantly different at  $p < 0.05$  according to Fisher's LSD test.  $n = 3$ .

**Table 11. Pearson's correlations matrix for mean (of 54 years) maize yield, mean (of 53 years) wheat yield, labile carbon stock, recalcitrant carbon stock, carbon management index (CMI), carbon accumulation rate and carbon sequestration rate in 0–30 cm soil depth.**

	Maize yield	Wheat yield	Labile C stock	Recalcitrant C stock	CMI	C accumulation rate	C sequestration rate
Maize yield	1.000						
Wheat yield	0.924**	1.000					
Labile C stock	0.862**	0.704NS	1.000				
Recalcitrant C stock	0.833**	0.797*	0.755*	1.000			
CMI	0.600NS	0.357NS	0.891**	0.590NS	1.000		
C accumulation rate	0.906**	0.790*	0.961**	0.907**	0.819*	1.000	
C sequestration rate	0.617NS	0.486NS	0.591NS	0.635NS	0.383NS	0.643NS	1.000

\*Significant at  $P < 0.05$ ; \*\*Significant at  $P < 0.01$ .

**Table 12. Pearson's correlations matrix for mean (of 54 years) maize yield, mean (of 53 years) wheat yield, labile carbon stock, recalcitrant carbon stock, carbon management index (CMI), carbon accumulation rate and carbon sequestration rate in 30–60 cm soil depth.**

	Maize yield	Wheat yield	Labile C stock	Recalcitrant C stock	CMI	C accumulation rate	C sequestration rate
Maize yield	1.000						
Wheat yield	0.924**	1.000					
Labile C stock	0.930**	0.799*	1.000				
Recalcitrant C stock	0.662NS	0.715*	0.560NS	1.000			
CMI	0.509NS	0.258NS	0.764*	0.309NS	1.000		
C accumulation rate	0.877**	0.844*	0.852*	0.910**	0.584NS	1.000	
C sequestration rate	0.661NS	0.714*	0.557NS	0.999**	0.309NS	0.908**	1.000

\*Significant at  $P < 0.05$ ; \*\*Significant at  $P < 0.01$ .

large macroaggregates in NPKL (as observed by us; data not shown) might be one of the major reasons for this, as macroaggregates provide better biophysical and chemical protection to SOC from getting degraded by microorganisms. Correlations of C accumulation rates with maize yield, wheat yield, labile C stocks, recalcitrant C stocks and CMI values were positive and significant, whereas C sequestration rates had non-significant correlations with all these parameters (Table 11) in the 0–30 cm soil layer, indicating greater role of C accumulation in the surface layer, but non-significant impact of C sequestration in this soil layer. Positive and significant correlations between crop yields and carbon accumulation rates [47] suggested that crop biomass played important roles in accumulating more C in the 0–30 cm soil layer. Non-significant correlation between C sequestration rate and other parameters might be due to the fact that in the 0–30 cm soil layer, due to continuous disturbance in the surface layer (caused due to various crop management practices), C stabilization was lower, even after longer period of time. In the deep soil

layer, NPKL treated plots could accumulate and sequester more C than organically amended plots. This could be due to the fact that, FYM had little influence in increasing C stock in lower soil depth [48]. In this soil depth, root biomass might be the major factor in binding soil aggregates [49] and increasing and sequestering more C than organically amended plots. In both soil layers, N treated plots had negative C accumulation rate. This could be due to lower soil reaction [50], less soil aggregation and least biomass production. The correlation of C accumulation rate with recalcitrant C stock was highly significant ( $r = 0.910^{**}$ ), indicating role of recalcitrant C stocks in C sequestration in the deep soil layer (30–60 cm depth) (Table 12). Carbon sequestration rate was well correlated with mean wheat yield and C accumulation rate in the deep soil layer. It is worth noting that C sequestration rate was highly correlated with recalcitrant C stock in the deep soil layer ( $r = 0.999^{**}$ ) (Table 12). This implied that in the deep soil layer, C stabilization was more due to fewer disturbances. Highest degree of correlation was found between labile C

**Table 13.** Pearson's correlations matrix for mean (of 54 years) maize yield, mean (of 53 years) wheat yield, labile carbon stock, recalcitrant carbon stock, carbon management index (CMI), carbon accumulation rate and carbon sequestration rate in 0–60 cm soil depth.

	Maize yield	Wheat yield	Labile C stock	Recalcitrant C stock	CMI	C accumulation rate	C sequestration rate
Maize yield	1.000						
Wheat yield	0.924**	1.000					
Labile C stock	0.897**	0.747*	1.000				
Recalcitrant C stock	0.779*	0.789*	0.715*	1.000			
CMI	0.564NS	0.313NS	0.844**	0.481NS	1.000		
C accumulation rate	0.914**	0.830*	0.941**	0.909**	0.732*	1.000	
C sequestration rate	0.777*	0.782*	0.722*	0.999**	0.494NS	0.913	1.000

\*\*Correlation is significant at  $p < 0.01$ ;

\*Correlation is significant at  $p < 0.05$ .

NS = Non-significant.

stock and C accumulation rate in the 0–60 cm soil layer ( $r = 0.941^{**}$ ) (Table 13).

#### 4.5. Long-term fertilization and liming effects on carbon management indices (CMI)

It is imperative to study the labile fractions of soil C, which greatly influence nutrient cycles. Thus, CMI is a good indicator which serves this very purpose. Increased amount of  $\text{KMnO}_4\text{-C}$  in the surface soil layer was attributed to the better physical environment, increased nutrient availability and total SOC in the surface layer [4]. This might have encouraged soil microbial activity, thereby increasing the lability of C. Significant and positive correlations of labile C stocks with CMI values ( $r = 0.819^{**}$ ) in surface layer is indicative of this (Table 11). Increased CMI of FYMP'K'L plots in all soil layers was due to addition of external source of organic inputs, which might have increased nutrient availability, improved soil physical property and facilitated biological activity in soils [45]. Furthermore, integration of lime increased pH and reduced toxicity of  $\text{Al}^{3+}$  and  $\text{Fe}^{3+}$  in these acid soils, and also ameliorated for deficiency of  $\text{Ca}^{2+}$  [4] Increased CMI of plots under NPKL plots in the sub-surface soil indicated it's potential of maintaining higher labile C for sustaining crop yield in the long-run.

## 5. Conclusions

Plots with FYMP'K'L and FYMP'K' had higher carbon accumulation rates in the 0–30 cm soil layer than NPKL plots. However, C sequestration rate in the 0–60 cm soil layer was highest ( $0.35 \text{ Mg ha}^{-1} \text{ yr}^{-1}$ ) for NPKL treated plots. It indicates optimum dose of mineral fertilization along with liming is the best management practice for sequestering SOC in an Alfisol. In this study, full doses of NPK and FYM were not applied under INM treatments, but the FYM was applied on equivalent N basis to both crops and P and K

doses were adjusted based on P and K contents in applied FYM over the years (FYMP'K'L). Thus, this study concludes that resource poor farmers need not to use full doses of NPK and FYM for soil C management. About  $0.17 \text{ Mg ha}^{-1} \text{ yr}^{-1}$  of C sequestration was observed in the deep soil layer (30–60 cm) of NPKL plots over the unfertilized control plots. So it indicates the need to alter soil sampling strategy (sampling in deeper soil layers as well) in future while studying C sequestration of soil. Increased CMI of NPKL plots in the sub-surface soil layer indicated it's potential of maintaining higher labile C for sustaining crop yield in the long-run. Positive and significant correlations between crop productivity and both labile and recalcitrant C pools in the surface as well as deep soil layers indicate that both these pools are beneficial for crop growth.

## Disclosure statement

No potential conflict of interest was reported by the author(s).

## Funding

For this study financial assistance was provided by the Indian Council of Agricultural Research (ICAR).

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