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**RESEARCH ARTICLE** 



# Assessment of long-term Site-Specific Nutrient Management (SSNM) on soil organic carbon dynamics and sequestration in a rice-rice cropping system

A Chitra Devi<sup>1</sup>, S Maragatham<sup>1</sup>\*, K Sathiyabama<sup>1</sup>, D Balachandar<sup>2</sup>, V Davamani<sup>3</sup>, M Gopalakrishnan<sup>1</sup> R Rajeswari<sup>1</sup> & P Malathi<sup>1</sup>

<sup>1</sup>Department of Soil Science and Agricultural Chemistry, Tamil Nadu Agricultural University, Coimbatore 641003, Tamil Nadu, India <sup>2</sup>Department of Agricultural Microbiology, Tamil Nadu Agricultural University, Coimbatore 641003, Tamil Nadu, India <sup>3</sup>Department of Environmental Sciences, Tamil Nadu Agricultural University, Coimbatore 641003, Tamil Nadu, India

\*Email: maragatham.s@tnau.ac.in

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## Abstract

This study examined the long-term effects of the Site-Specific Nutrient Management Integrated Plant Nutrient System (SSNM-IPNS) on carbon sequestration, stock, loss and mineralization in soil within a 26-year-old ricerice cropping system at wetland of Tamil Nadu Agricultural University, Coimbatore. The research focused on the impact of integrated organic and inorganic nutrient application on carbon fractions compared to conventional fertilization methods under wetland ecosystems. Results indicated that the SSNM-IPNS approach significantly enhances soil total organic carbon (TOC), with levels reaching 1.56% compared to just 0.78% in control treatments. Furthermore, labile carbon fractions such as permanganate-oxidizable carbon (POx-C), microbial biomass carbon (MBC), water-soluble carbon (WSC), and particulate organic carbon (POC) were found to be greater in the SSNM-IPNS management. Increased rates of carbon mineralization and basal respiration also reflected a more active and efficient microbial community in these soils. Soil microbial indices, including the microbial quotient (qMic) and the metabolic quotient (qCO<sub>2</sub>), further emphasized the benefits of the SSNM-IPNS approach in enhancing soil health. Overall, the findings demonstrate that SSNM -IPNS significantly improves carbon sequestration, nutrient cycling, and soil fertility, promoting sustainable agricultural practices vital for long-term productivity in intensive cropping systems. This research underscores the importance of adopting innovative nutrient management practices for sustainable agriculture.

### **Keywords**

active pools; carbon stock; organic carbon pools; passive pools; SSNM-IPNS

## Introduction

Rice (*Oryza sativa L*.) cultivation is the most important agricultural operation in India, not only in terms of food security but also as an assurance of livelihoods. It is a stable food crop. To achieve sustainable agriculture without harming soil fertility, the integration of fertilizers and organic manure is followed. However, intensive rice cultivation can lead to soil degradation, nutrient depletion, and increased greenhouse gas emissions. The concept of a soil test based nutrient management approach has been found to be most effective in developing recommendations for potential productivity of crops and maintaining soil health (1). Site-Specific Nutrient Management Integrated Plant Nutrient System (SSNM-IPNS) studies are based on inductive cum targeted yield model which is efficient and profitable for site-specific nutrient management. The research focuses on providing balanced, efficient and profitable nutrient application rates for preset yield targets considering the basic fertility status of soil. The use of SSNM-IPNS based fertilization has been useful for prescribing fertilizer doses to rice, to achieve the desired productivity and improve soil health and quality. The longterm fertilization of rice influences the soil quality parameters and fertility status.

Soil organic matter (SOM) can be categorized into labile, slow, and recalcitrant organic matter on the basis of turnover rates (2). Labile organic carbon (LOC), a collection of reactive chemical compounds, plays a crucial role in global carbon cycling because of its rapid turnover time and sensitivity to environmental changes (3). Soil microbial biomass is considered a sensitive indicator of changes in total soil organic matter (SOM) because it quickly responds to modifications in plant vegetation or land use (4). Soil respiration is crucial for the carbon cycle in terrestrial ecosystems, as small changes can significantly impact carbon sequestration. Water-soluble carbon accounts for a small part of SOM, but serves as the primary energy and substrate source for soil microorganisms. The carbon management index (CMI), which is based on the total soil organic carbon pool and labile carbon, is widely used as a sensitive indicator of SOC variation due to changes in soil management (5),(6).

Carbon mineralization is the process by which organic carbon compounds in soil are broken down into inorganic forms, such as carbon dioxide (CO<sub>2</sub>) or carbonate ions (CO<sub>3</sub><sup>2-</sup>), through microbial activity and chemical reactions. This process is a key part of the carbon cycle and influences soil health and greenhouse gas emissions. The potential of soil to sequester carbon, as well as its dynamics and turnover, may depend on the soil type and climate. It is important to determine C dynamics under different nutrient management practices. Very few studies have been conducted to understand the organic carbon dynamics in SSNM-IPNS based fertilized soil in India. Therefore, an attempt is made to investigate the carbon sequestration potential and dynamics of the labile pool of SOC under long-term SSNM based fertilization in a rice-rice cropping system.

### **Materials and Methods**

Long-term SSNM-IPNS experiments (26 years) have been conducted since kharif 1998 in the wetland farm of Tamil Nadu Agricultural University, Coimbatore and two crops were grown annually during kharif and rabi seasons. The experimental field is located in the agroclimatic zone of southern plateau and hills at 11.003139° N latitude and 76.924889° E longitude, with an elevation of 411 m above mean sea level (MSL). This technique has been maintained as a long-term experiment with the objective of monitoring the changes in soil fertility affected by the continuous adoption of SSNM-IPNS technology. The soil of the field is classified as Typic Haplustalf and the experimental soil belongs to the Noyyal series. It is clay loam in texture, has a neutral pH, is slightly calcareous and non-saline. Rice variety CO55 was grown. Five treatments were technically replicated:  $T_1$ : Blanket recommendation,  $T_2$ : SSNM Target I,  $T_3$ : SSNM Target II,  $T_4$ : SSNM-IPNS Target II,  $T_5$ : Absolute control. Where SSNM Target I and Target II are the soil test crop response for a yield target of 5 and 6 t ha<sup>-1</sup>, respectively.

The STCR-IPNS based fertilizer prescription equation was given in Table 1. For the IPNS plots, FYM @ 12.5 t ha<sup>-1</sup> and Azospirillum and Phosphobacteria each @ 2 kg ha<sup>-1</sup> were applied and the fertilizer doses were adjusted accordingly. Full dose of  $P_2O_5$  and 25% N and K<sub>2</sub>O were applied basally during transplanting. The remaining N and K<sub>2</sub>O were applied in three equal splits during active tillering, panicle initiation, heading stages and routine agronomic practices were carried out periodically.

### Soil sample collection

Surface soil samples were collected from each treatment plot to a depth of 0 -15 cm after the harvest of Kharif rice 2023. The soil samples were homogenized and processed by removing the stones and stubbles. The samples were then passed through a 2 mm sieve before being analyzed for the soil carbon fractions. The samples were stored in air-tight plastic bags.

 Table 1. STCR- IPNS based fertilizer prescription for rice during rabi season (2023).

FN	4.63 T – 0.52 SN – 0.80 ON
$FP_2O_5$	1.98 T – 3.18 SP – 0.99 OP
FK <sub>2</sub> O	2.57 T – 0.42 SK – 0.67 OK

Where FN, FP2O5, and FK2O are fertilizer N, P2O5, K2O in kg ha-1 respectively. T is the grain yield target in q ha-1; SN, SP, and SK are alkaline KMnO4- N, Olsen -P and NH4OAc-K in kg ha-1 and ON, OP, and OK are the quantities of N, P and K in kg ha-1 supplied through the FYM

### **Chemicals and reagents**

Chemicals used for experimental analysis includes sulphuric acid ( $H_2SO_4$ ), potassium permanganate ( $KMnO_4$ ), Orthophosphoric acid, Diphenyl amine, potassium sulphate ( $K_2SO_4$ ), ferrous ammonim sulphate (FAS), Potassium dichromate, chloroform, sodium hydroxide (NaOH), hydrochloric acid (HCl), Barium chloride (BaCl<sub>2</sub>) and phenolphthalein were procured from LABTECH laboratory products and PALLAV chemicals and solvents PVT LTD.

## Soil analysis

Soil organic carbon (SOC) was measured using the wet combustion method by Walkley and Black (1934). SOC fractions were estimated using a modified Walkley and Black method with 5, 10, and 20 ml of concentrated  $H_2SO_4$ corresponding to 12.0 N, 18.0 N, and 24.0 N  $H_2SO_4$ , respectively (7). Total organic carbon (TOC) was classified into four fractions with decreasing oxidizability, as shown in Fig. 1. The microbial biomass carbon in the soil samples was estimated using the chloroform fumigation and incubation method(8). Fumigated and non-fumigated samples were extracted with 0.5 M  $K_2SO_4$ , and the difference in the carbon content between the two samples represented the microbial carbon. This value was converted to microbial biomass carbon by dividing the carbon by 0.41 (9). Particulate organic carbon (POC) was measured by separating the organic and Very labile (VL or C pool I) organic C oxidisable under 12.0 N H<sub>2</sub>SO<sub>4</sub> Labile (L or C pool II) The difference in SOC extracted between 18.0 N and 12.0 N H<sub>2</sub>SO<sub>4</sub> Less labile (LL or C pool III) The difference in SOC extracted between 24.0 N and 18.0 N H<sub>2</sub>SO<sub>4</sub> Non labile (NL or C pool IV) The difference in SOC extracted between 24N N  $H_2SO_4$  and TOC .

Fig.1 Standard operating procedure of oxidizable organic carbon fractions

mineral fractions of SOC ranging from 53-200 microns and analysing them via a CHNS analyser. POC differs from active soil carbon as it is often associated with the slow pool of organic carbon (10).

Total organic carbon (TOC) in soil and organic samples was analysed via a CHNS analyser. Soil carbon sequestration was estimated by difference between the initial and final SOC concentrations over the experimental years (11)

Carbon sequestration (Mg/ha soil) = <u>Current SOC – Initial SOC</u> 26

Organic carbon stock in soils was calculated using given formula (12).

Carbon stock = TOC × Depth × Bulk Density

Where the C content is given in g C kg<sup>-1</sup>, BD in Mg m<sup>-3</sup>, depth in m and C stock in Mg ha<sup>-1</sup>.

## **Incubation experiment**

Incubation experiments were carried out to study the carbon mineralization pattern and decomposition rate under the influence of SSNM-IPNS based fertilizer prescription. Under controlled conditions, an incubation experiment was conducted in the laboratory. Fifty grams of soil was weighed, placed in a 500 ml conical flask, and moistened to 50% waterfilled pore space. The vials were placed inside conical flask containing 10 ml of 0.1 M NaOH to trap the CO<sub>2</sub> evolved and were incubated for 48 days. To determine the evolved  $CO_2$ , the NaOH traps were exchanged regularly (in two days intervals for first one week and four days interval for second one week and weekly interval up-to incubation period) with new traps and then determined by titrating the alkali (NaOH) in the traps with 0.1N H<sub>2</sub>SO<sub>4</sub> using phenolphthalein as an indicator. The CO<sub>2</sub> that evolved after 48 days of incubation was considered as mineralizable C ( $CO_2$ -Ccum) (13).

#### **Carbon management index**

The carbon management index (CMI) was calculated by mathematical procedures developed previously (14).

CMI=CPI × LI × 100

Where,

CPI is the carbon pool index and LI is the lability index. The CPI and LI are calculated as

## Carbon pool index (CPI) =

<u>Total organic carbon in fertilized treatment</u> Total organic carbon in control treatment Lability index (LI) =

Lability of carbon in fertilized treatment Lability of carbon in control treatment

#### **Statistical analysis**

The data were analysed using a randomized block design (RBD). Statistical analysis was performed by using SPSS version 17.0 on a DOS-based system. The analysis of variance (ANOVA) procedure in SPSS was employed to assess the statistical significance of the treatments. A significance level of 5% (p < 0.05) was considered statistically significant.

## **Results and Discussion**

### **Total organic carbon**

The intensive cultivation of rice with SSNM-IPNS based fertilization led to an increase in total organic carbon (TOC) in the soil. The soil under recommended and no fertilization conditions showed a decrease in the TOC levels. Similar findings were reported previously (15). Specifically, the TOC content in the SSNM-IPNS treatment was 1.56%, which was significantly (P=0.05) higher than the TOC content in the control (0.78%). The results indicate that the SSNM-IPNS Target II treatment is the most effective in enhancing TOC accumulation, followed by SSNM - Target II (1.35%) and the SSNM-Target I (9.3) treatments. The amount of carbon found in organic compounds within a given sample indicates good soil health and fertility. The optimized nutrient levels in the SSNM-IPNS approach increases microbial activity, facilitating the decomposition of organic matter and resulting in increased accumulation of TOC.

## **Organic Carbon pools**

The concentration of oxidizable organic carbon in surface soils (0-15 cm) increased significantly (P=0.05) with the application of inorganic fertilizers with organic amendments, compared to the control treatment. The carbon pool, extracted under varying oxidizing conditions, was recorded in the following order: very labile ( $C_{VL}$ ) > Labile ( $C_L$ ) > Less labile ( $C_{LLC}$ ) (Table 2). The contributions of the VL, L, and LC pools to the total organic carbon were 21.6%, 13.9%, and 6.73%, respectively. The application of manure in conjunction with NPK fertilizer increases the recalcitrant and labile C in the subsoil which results in a substantial increase in SOC (16). The active carbon pools ( $C_{VL} + C_L$ ) account for approximately 35.8% of the TOC, the passive pools ( $C_{LL} + C_{RC}$ ) made up 64.4% of the TOC. This indicates that the active carbon pools, which are more readily decomposable, represent a smaller portion

Table 2. Effect of long-term SSNM approach on soil carbon fractions

Treatments	CVLC	CLC	CLLC	C <sub>RC</sub>	C <sub>TOC</sub>	AP	PP
T1: Blanket	2.25	1.95	0.82	6.67	11.7	4.2	7.5
T <sub>2</sub> : SSNM- Target I	2.77	1.35	0.82	4.35	9.3	4.1	5.18
T₃: SSNM- Target II	2.85	1.87	0.9	7.87	13.5	4.7	8.78
T₄: SSNM-IPNS Target II	3.37	2.17	1.05	9	15.6	5.6	10.05
T <sub>5:</sub> Absolute control	1.87	1.2	0.45	4.27	7.8	3.1	4.73
SE d	0.14	0.32	NS	0.23	0.1	0.29	0.254
CD (P < 0.05)	0.44	0.7	NS	0.72	0.33	0.894	0.781

(very labile, labile, less labile and nonlabile) in soils (g kg-1) under different treatment (CVLC – very labile carbon, CL- Labile carbon, CLLC- less labile carbon CRC - Recalcitrant carbon, CTOC- total organic carbon, AP- Active pools, PP- Passive pools)

of the TOC compared to the more stable, passive carbon pools. Among the treatments, highest  $C_{VL}$  (3.37),  $C_L$  (2.17),  $C_{LLC}$ (1.05) and Recalcitrant Carbon (RC) (9.0) were recorded in SSNM-IPNS Target II treatment, followed by the SSNM- Target II treatments. The SSNM-IPNS- Target II treatment resulted in the highest levels of both active (5.6) and passive carbon pools (10.05) than absolute control. The increase in oxidizable carbon in the SSNM-IPNS Target II is primarily due to the high concentration of mineral nutrients and enhanced microbial activity, that stimulated root activity and secretion, leading to increased carbon fractions in rhizosphere soil. Similar findings were also reported (17).

## **Labile Carbon fractions**

Permanganate oxidizable carbon (POx-C) represents 5.6% of TOC. The SSNM-IPNS treatment resulted in significantly greater POx-C. POx-C was highly sensitive to farming practices as it varied significantly in all the treatments tested. Intensive rice cultivation without any fertilization (control) significantly (P= 0.05) decreased the permanganate oxidizable carbon (POx-C) content. Biomass carbon of microbes (MBC) followed the same trend as oxidizable organic carbon. Higher amount of MBC was exhibited by the SSNM-IPNS Target II (432.2 mg/kg) treatment followed by SSNM-Target II (376.2 mg/kg); lower range was observed in control (191 mg/kg). Water soluble carbon (WSC) concentrations were highest in T<sub>4</sub> (SSNM-IPNS Target II) at 42.0 mg kg<sup>-1</sup>, followed by  $T_3$  (SSNM- Target II at 37.8 mg kg<sup>-1</sup> and the lowest value was recorded in the control (23.1 mg kg<sup>-1</sup>). The particulate organic carbon was higher in the SSNM-IPNS Target II treatment than in the absolute control (Table 3 and Fig. 2.) SSNM-IPNS Target II (Soil test based fertilizer with FYM) enhances carbon availability and microbial activity due to high organic matter decomposition which increases the

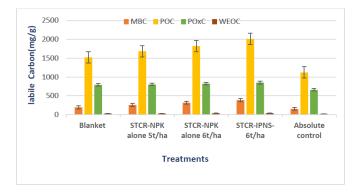


Fig.2 Effect of SSNM approach on organic carbon pools

POx-C, MBC, WSC, and POC levels. The absence of fertilization in the control resulted in lower carbon and microbial activity.

## Mineralizable carbon and basal respiration

Mineralizable Carbon (Cmin) and Basal Respiration (BSR) represent the performance of the organic C decomposer. The effect of soil test crop response fertilization on C min ranged from 299.3 mg CO<sub>2</sub>-C kg<sup>-1</sup> in the control treatment to 561.7 mg CO<sub>2</sub>-C kg<sup>-1</sup> in the SSNM-IPNS Target II treatment. The C <sub>min</sub> of the different treatments was in the following the order: SSNM -IPNS Target II > SSNM- Target II > SSNM- Target I > Blanket > Absolute control. The higher carbon mineralization (C min) observed in treatments combining NPK fertilizers with organic amendments is attributed to the sufficient supply of labile carbon substrates, as noted by previous studies (18); (13). The long-term integrated addition of synthetic chemical fertilizers and farmyard manures not only produces a favourable environment for microbial growth and activity but also provides the necessary substrates for mineralization processes (19). The surface soil has abundant organic material and extensive root proliferation, which enhances microbial activity. BSR is a sensitive indicator for soil quality and a valid biomarker for identifying changes in soil microbial activity due to soil management, agronomic practices, and climate variations (20). In surface soil (0-15 cm) the BSR ranged from 0.99 in the control to 1.48 mg CO<sub>2</sub>-C kg <sup>-1</sup> day<sup>-1</sup> in SSNM-IPNS Target II. The presence of easily decomposable organic matter and readily available nutrients creates a favourable environment for microbial activity, resulting in a relatively higher rate of respiration.

# Effects of long term SSNM fertilization and manuring on soil microbial indices

After 26 years of continuous cropping with a rice-rice sequence, the microbial indices in soil treated with various manures and fertilizers significantly differed among the different treatments. The results are shown in Table 4. Microbial quotients are crucial for evaluating the long-term effects of nutrient management practices on soil quality (21). A high microbial quotient indicates an abundant pool of readily available carbon that supports a large microbial community. qMic ranged from 2.3% (control) to 2.47% (SSNM -IPNS Target II). The highest qMic was observed with the integrated application of NPK and FYM (SSNM-IPNS Target II) which is attributed to an improved nutritional environment. A high qMic signifies the presence of a readily available carbon pool that supports a diverse microbial population, while a lower value suggests nutritional stress (22). The application of blanket recommended fertilizer had a lower microbial

#### Table 3. Impact of STCR- IPNS fertilization on Soil Organic Carbon Fractions.

Treatments	POx-C	МВС	WSC	POC
T1: Blanket	765.8 <sup>d</sup>	248.0 <sup>d</sup>	27.2	1520
T <sub>2</sub> : SSNM- Target I	816.8 <sup>c</sup>	301.7°	33.6	1689
<u>T<sub>3</sub>: SSNM- Target II</u>	<u>864.2<sup>b</sup></u>	376.2 <sup>b</sup>	37.8	1820
T <sub>4</sub> : SSNM-IPNS Target II	937.1ª	432.2ª	42	2009
T <sub>5:</sub> Absolute control	689.3°	191.0 <sup>e</sup>	23.1	1121
SE(d)	11.89	2.92	0.591	14.25
CD	25.91	6.36	1.287	31.05

(\*Values in same column followed by different treatment are significantly different at alpha =0.05 according to Latin square test (LSD) for separation of means)

Tractments	C <sub>min</sub>	BSR	qCO <sub>2</sub>	Qmin
Treatments	(mg CO <sub>2</sub> -C kg <sup>-1</sup> )	(mg CO <sub>2</sub> -C kg <sup>-1</sup> day <sup>-1</sup> )	(X 10 <sup>-3</sup> mg CO <sub>2</sub> - C mg <sup>-1</sup> )	(%)
T₁: Blanket	397.6 <sup>d</sup>	1.21 <sup>c</sup>	4.75 <sup>b</sup>	2.1
T <sub>2</sub> : SSNM- Target I	440.9 <sup>c</sup>	1.17 <sup>c</sup>	4.02 <sup>c</sup>	2.32
T₃: SSNM- Target II	490.5 <sup>b</sup>	1.35 <sup>b</sup>	3.62 <sup>d</sup>	2.42
T <sub>4</sub> : SSNM-IPNS Target II	561.7ª	1.48ª	<b>3.42</b> <sup>d</sup>	2.47
T <sub>5:</sub> Absolute control	299.3 <sup>e</sup>	0.99 <sup>d</sup>	5.17ª	2.3
SE(d)	6.4	0.02	0.1	0.12
CD	13.9	0.05	0.22	0.25

(qCO2 - metabolic quotient, Microbial quotient (qMic), CMin - cumulative carbon mineralized )

quotient (2.10%) than SSNM-IPNS Target II fertilization. A higher qMic value indicates superior stability of SOC under this treatment which indicates the effectiveness of soil microbes in degrading residues. A higher qCO<sub>2</sub>, associated with recalcitrant organic carbon fractions, indicates a less effective microbial population. In the soil,  $qCO_2$  (x 10<sup>-3</sup> mg CO<sub>2</sub>-C mg<sup>-1</sup>) ranged from 3.42 in SSNM-IPNS Target II to 5.17 in the control. The higher  $qCO_2$  in the control indicates reduced efficiency of carbon utilization by the microbes which suggests the presence of recalcitrant carbon not utilized for microbial growth (23),(24). In contrast, the lower qCO<sub>2</sub> (indicating greater metabolic efficiency of the microbial community) observed in the SSNM-IPNS Target II treatment showed the effectiveness in preserving soil organic carbon (25). The mineralization quotients showed significant variation across the different fertilization treatments.

## **Cumulative mineralization carbon**

The carbon mineralization rate varied significantly across the various treatments throughout the incubation experiment. The cumulative carbon mineralization ( $C_0$ ) for each treatment is shown in Table 4. and Fig. 3. The carbon mineralization during the incubation period ranges from 299.3 mg CO<sub>2</sub>–C kg<sup>-1</sup> to 561.7 mg CO<sub>2</sub>–C kg<sup>-1</sup> in which SSNM-IPNS Target II fertilization resulted in greater mineralization, followed by SSNM- Target II whereas, lesser carbon mineralization was observed in absolute control with value of 299.3 mg CO<sub>2</sub>–C kg<sup>-1</sup>. Carbon mineralization was rapid during the initial incubation period but gradually decreased over time. This

decline in the mineralization rate was attributed to the accumulation of structural carbohydrates (such as lignin and hemicelluloses) as other components (such as sugars and starches) in the detritus were depleted (26). The soil mineralization potential is influenced by microbial activity, total organic carbon, and various physical and chemical factors. The enhancement of microbial activity was facilitated by the combined application of organic and inorganic fertilizers for a long period which provided the substrates required for mineralization (27). The lowest mineralization rates in the control plots were due to nutrient depletion from continuous cropping, which negatively impacted the physical and fertility properties of the soil, creating an unfavourable environment for microbial activity.

## **Carbon management index**

The carbon management index (CMI) is an important factor for evaluating the efficacy of various fertilization practices for

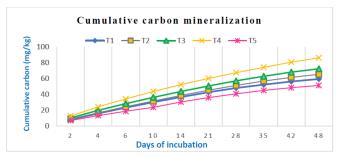


Fig.3 Cumulative CO2–C evolved from soil under long-term fertilizer experiment

soil productivity. Higher CMI values indicate that the management techniques were essential for maintaining SOC (14). The SSNM-IPNS Target II treatment shows a high CMI value of 383.2% indicating greater stable SOC (Table 5). This higher value was attributed to the application of external C inputs to soil and these practices may lead to stable SOC (28). The CMI was significantly lower value of 100% in absolute control followed by the treatment that received only inorganic fertilizer. Improved CMI values under integrated plant nutrient system (IPNS) system have also been reported (29). Higher CMI values may result from both increased organic matter formation due to greater annual carbon inputs and changes in organic matter quality, including changes in the C/N ratio and the contents of lignin, cellulose, hemicellulose, proteins, and carbohydrates, which affect the C lability towards oxidation (30). Based on these findings, we can conclude that the addition of organics had a higher rehabilitation rate.

soil properties (Fig. 5.). Specifically, TOC is strongly positively correlated with MBC, carbon mineralization (Cmin), and both POC and water-soluble carbon (WSC). This means that as TOC increases, other carbon-related metrics also increases suggesting that higher organic carbon enhances microbial activity and increases the availability of different forms of soil organic carbon. MBC shows strong positive correlations with TOC and carbon mineralization, reflecting that more organic carbon supports greater microbial growth and activity. MBC was also positively correlated with both soluble and particulate organic carbon. TOC has a strong negative correlation with bulk density (BD), indicating that soils with more organic carbon tend to have lower bulk density. A negative correlation between bulk density and SOC content has been previously reported (31), (32). This reduction in bulk density signifies improved soil structure and reduced compaction, which facilitates better root growth and soil aeration. BD is also negatively correlated with carbon

Table 5. Carbon sequestration in surface soil (0-15 cm) after	er 25 years of application of STCR- IPNS fertilization in rice -	<ul> <li>rice cropping system</li> </ul>
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Treatments	тос	Bulk density	C stock	СМІ	C sequestrated		
Treatments	(g/kg)	(mg/cm³)	(Mg/ha)	(%)			
T1: Blanket	9.30 <sup>d</sup>	1.25 <sup>ab</sup>	17.61	150.93°	0.06 <sup>b</sup>		
T <sub>2</sub> : SSNM- Target I	11.70 <sup>c</sup>	1.19 <sup>bc</sup>	20.69	263.03 <sup>b</sup>	0.05 <sup>b</sup>		
T₃: SSNM- Target II	13.50 <sup>b</sup>	1.16 <sup>bc</sup>	23.79	279.57 <sup>b</sup>	0.11 <sup>b</sup>		
T <sub>4</sub> : SSNM-IPNS Target II	15.60ª	1.11 <sup>c</sup>	25.48	383.20ª	0.196ª		
۲ <sub>5:</sub> Absolute control	7.80 <sup>e</sup>	1.308ª	15.28	100 <sup>c</sup>	-0.06 <sup>c</sup>		
SE(d)	0.15	0.046	0.79	46	0.03		
CD	0.33	0.101	1.73	100.2	0.06		

## **Carbon stock**

In the 0-15 cm soil layer, the Bulk Density (BD) was significantly lower in SSNM-IPNS Target II (1.11 Mg/cm<sup>3</sup>). The soil test based application of NPK reduced bulk density across the various soil depths. The absolute control had the highest bulk density (1.308 Mg/cm<sup>3</sup>). Soils with higher organic matter contents have better structures and lower densities. The highest carbon stock was recorded in the SSNM-IPNS Target II treatment at 10.77 t ha<sup>-1</sup>. This was followed by the SSNM- Target II and the blanket recommendation with values of 9.89 t ha<sup>-1</sup> and 9.38 t ha<sup>-1</sup> respectively. SSNM- Target I resulted in a slightly lower carbon stock of 8.87 t ha<sup>-1</sup>, and the absolute control had the lowest carbon stock at 6.92 t ha<sup>-1</sup>. These results indicate that nutrient management significantly enhances soil carbon sequestration. The higher carbon stock in the SSNM-IPNS Target II treatment was attributed to the combined application of inorganic fertilizers and organic inputs, which directly add organic carbon to the soil. OM improves the soil structure, promotes microbial activity, and leads to the formation of stable humus, all of which contribute to increased carbon sequestration in the soil. Hence, carbon sequestration was also highest in the SSNM-IPNS 6t ha<sup>-1</sup> treatment (5.15 Mg/ha), as shown in Fig. 4.

# Relationships between different organic carbon fractions

The correlation matrix shows the impact of TOC on various

mineralization, suggesting that less compacted soils with higher organic carbon content have higher microbial activity and carbon turnover. Overall, the data highlights the critical role of TOC in improving soil structure, microbial health, and carbon dynamics, leading to enhanced soil fertility and function.

## Effect of Long-term Fertilization on the Functional Groups of Humic Acid, Fulvic acid and Humin

The analysis of the FTIR spectra for different treatments reveals significant insights into the functional groups present in humic acids (HA) and their relationships with long-term fertilization (Fig. 6.). The FTIR spectra of humic acid extracted from various treatments revealed key functional groups, including aliphatic C-H, O-H deformation, C-O stretching of phenolic OH, and COO<sup>-</sup> antisymmetric stretching. These functional groups are critical in understanding the structural characteristics of soil organic matter and its turnover in soil.

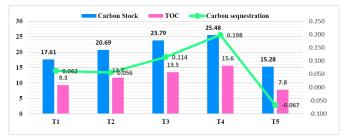


Fig.4 Impact of long-term SSNM Fertilization on TOC (g/kg), Carbon stock (Mg/ha), C sequestrated (Mg/ha).

	тос	BD	C_min	POxC	MBC	WSC	POC	BSR	VLC	LC	LLC	RC	AP	PP	qCO2	qmic
тос	1.00															
BD	-0.99	1.00														
C_min	0.98	-0.99	1.00													
POxC	0.99	-0.99	1.00	1.00												
MBC	1.00	-0.99	0.98	0.99	1.00											
WSC	1.00	-0.99	0.98	0.99	0.99	1.00										
POC	0.96	-0.99	0.99	0.98	0.97	0.97	1.00									
BSR	0.94	-0.94	0.97	0.96	0.96	0.93	0.95	1.00								
VLC	0.99	-0.99	0.98	0.99	0.90	0.99	0.97	0.92	1.00							
LC	0.67	-0.68	0.77	0.74	0.72	0.66	0.75	0.89	0.65	1.00						
LLC	0.89	-0.93	0.96	0.94	0.91	0.90	0.98	0.95	0.92	0.84	1.00					
RC	0.77	-0.75	0.82	0.81	0.82	0.75	0.77	0.93	0.71	0.95	0.81	1.00				
AP	0.85	-0.84	0.90	0.89	0.89	0.84	0.87	0.98	0.81	0.95	0.89	0.99	1.00			
PP	0.94	-0.94	0.98	0.97	0.95	0.93	0.96	0.99	0.93	0.88	0.96	0.90	0.96	1.00		
qCO2	-0.98	0.99	-0.97	-0.97	-0.98	-0.99	-0.97	-0.90	-0.97	-0.61	-0.89	-0.71	-0.80	-0.89	1.00	
qmic	0.77	-0.69	0.63	0.68	0.74	0.76	0.57	0.55	0.70	0.18	0.40	0.43	0.47	0.54	-0.74	1.00

Fig. 5. Pearson's correlation among the soil attributes as influenced by long-term SSNM-IPNS nutrient management. The values in the plot represents Pearson's correlation coefficients and insignificant values (p<0.05) were crossed. Green colours represent negative correlations. Red colours represent positive correlations. The intensity of the colour indicates correlation strength.

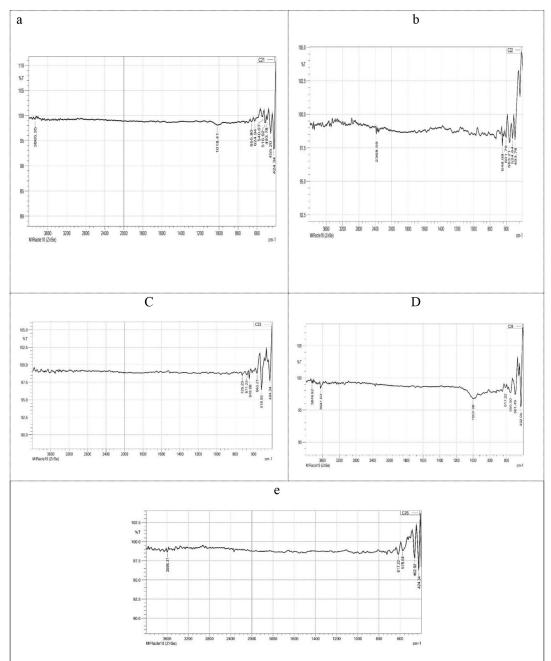


Fig. 6. Fourier transform infrared (FTIR) spectra of soil humic acid under (a) Blanket recommendations (b) SSNM- Target I (c) SSNM- Target II (d) SSNM- IPNS Target II and (e) Absolute control treatment of during long-term fertilizer experiments. % T= Transmittance; cm-1 = Wavenumber.

O-H and N-H stretching (3849.92 cm<sup>-1</sup>, 3641.60 cm<sup>-1</sup>) in the SSNM-IPNS (6t/ha) treatment indicates a higher concentration of hydroxyl groups, commonly found in fulvic and humic acids. The addition of organic inputs to the soil leads to increase in the HA content (33), (34), and (35). The band at 1018.41 cm<sup>-1</sup> is assigned to C=O stretch of carboxyl and carbonyl groups is the higher in SSNM-IPNS-Target II treatment, and SSNM-Target- I, which contribute to the chemical complexity and nutrient retention capacity of humic substances. Aromatic C-H bonding (655.80 cm<sup>-1</sup>, 624.94 cm<sup>-1</sup>) is present in treatments like SSNM- Target I, these bands suggest aromatic nature in the humic substances, particularly humic acid and Humin. Aromaticity in humic acids is often associated with more stable and recalcitrant organic matter. The presence of disulfide bonds S-S Stretch (432.05 cm<sup>-1</sup>) in the SSNM-IPNS Target II and SSNM- Target I treatments, indicates sulphur-containing functional groups, which are more prevalent in humic acid and contribute to soil fertility through enhanced sulphur content. Similar to previous studies, the NPK with FYM treatment shows additional functional groups beyond the basic structure of humic acids, indicating a more complex and chemically diverse organic matter profile.

The spectral intensity variations, particularly in the 1655 cm<sup>-1</sup> to 1560 cm<sup>-1</sup> range, suggest stronger aromatic C=C vibrations and symmetric stretching of COO<sup>-</sup> groups in these treatments. This implies that the combination of NPK with organic inputs like FYM enhances the stability and functionality of humic acids. The control treatment's spectra reveal a lower content of aliphatic and nitrogen-containing groups but more bands attributable to polysaccharides. This suggests that in the absence of additional fertilization, the humic acid structure is less complex, with fewer labile organic materials. SSNM- Target I and II treatments exhibit significant aromatic C-H bending and halogen stretches. However, the presence of halogens could suggest environmental contamination, which might not contribute positively to the natural humic substance profile. Long-term fertilization, especially with combinations of NPK and organic amendments such as FYM, leads to the improvement of more complex and stable humic acids. The SSNM-IPNS Target II treatment appears to enhance the presence of critical functional groups like hydroxyl and ether groups, which are essential for maintaining soil organic carbon and improving the soil structure (36). This treatment is considered superior in promoting the formation of high humic acids compared with other treatments, which show either less complex or potentially contaminated profiles.

## Conclusion

The long-term implementation of Site-Specific Nutrient Management (SSNM-IPNS) fertilization significantly impacts carbon dynamics and mineralization in rice-rice cropping systems. The SSNM-IPNS approach, which integrates inorganic with organic inputs based on the soil testing and crop needs, enhances soil carbon stock and promotes optimal carbon sequestration. The application of SSNM-IPNS has been shown to increase TOC and MBC, leading to improved soil structure and reduced bulk density. This results in more efficient carbon mineralization processes and a healthier soil ecosystem. The positive effects of SSNM-IPNS on soil carbon dynamics are attributed to the combined benefits of organic matter inputs, which enhance soil aggregation, increase microbial activity, and stabilize humus formation. Consequently, this approach not only improves soil fertility and productivity but also contributes to sustainable carbon management in rice-rice cropping systems. Overall, SSNM-IPNS fertilization offers a robust strategy for optimizing soil health and carbon sequestration, aligning with both agricultural productivity goals and environmental sustainability.

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# **Authors' contributions**

A carried out writing, and original draft preparation. S carried out conceptualization, data validation, methodology and editing. K participated in data curation, editing. D participated in the visualization, re-drafting and editing. V carried out reviewing and editing. M, R & P participated in correcting the final version of the manuscript. All authors read and approved the final manuscript.

## **Compliance with ethical standards**

**Conflict of interest:** Authors do not have any conflict of interests to declare.

Ethical issues: None

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