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Straw incorporation and nitrogen fertilization regulate soil quality, enzyme activities and maize crop productivity in dual maize cropping system

Li Yang^{1†}, Teng Yan Chen^{1†}, Zhong Yi Li^{2†}, Ihsan Muhammad¹, Yu Xin Chi¹ and Xun Bo Zhou^{1*}

Abstract

Background Straw incorporation serves as an effective strategy to enhance soil fertility and soil microbial biomass carbon (SMBC), which in turn improves maize yield and agricultural sustainability. However, our understanding of nitrogen (N) fertilization and straw incorporation into soil microenvironment is still evolving. This study explored the impact of six N fertilization rates (N0, N100, N150, N200, N250, and N300) with and without straw incorporation on soil fertility, SMBC, enzyme activities, and maize yield.

Results Results showed that both straw management and N fertilization significantly affected soil organic carbon (SOC), total N, SMBC, soil enzyme activities, and maize yield. Specifically, the N250 treatment combined with straw incorporation significantly increased SOC, total N, and SMBC compared to lower fertilization rates. Additionally, enzyme activities such as urease, cellulase, sucrose, catalase, and acid phosphatase reached their peak during the V6 growth stage in the N200 treatment under for both straw management conditions. Compared to N250 and N300 treatments of traditional planting, the N200 treatment with residue incorporation significantly increased yield by 8.30 and 4.22%, respectively. All measured parameters, except for cellulase activity, were significantly higher in spring than in the autumn across both study years, with notable increases observed in 2021.

Conclusions These findings suggest that optimal levels of SOC, soil total N (STN), and SMBC, along with increased soil enzyme activities, is crucial for sustaining soil fertility and enhancing maize grain yield under straw incorporation and N200 treatments.

Keywords Straw management, Soil fertility, Soil enzyme, Nitrogen fertilization, Maize

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Introduction

Maize (*Zea mays* L.), a major food crop, has seen a significant increase in grain yield, rising by 460% from 1961 to 2019. This growth is crucial for sustaining global food security and economic stability [1, 2]. According to a United Nations estimate, this is anticipated to increase maize grain yield by 40% till 2050 [3]. Research has shown that straw management, including incorporation and mulching, enhances soil fertility and crop productivity, while also mitigating environmental pollution by reducing N fertilization requirements [4–6]. Such practices not only improve soil fertility but also increase SOC, other soil nutrients, soil structure, and overall crop production [7–10]. Furthermore, appropriate C/N ratio accelerates straw decomposition, releasing nutrients essential for crop growth, thus optimizing N fertilizer utilization [11]. X Meng, et al. [12] reported that straw incorporation with N fertilization (150–225 kg ha⁻¹) dramatically improved plant N uptake and increased grain and biomass yield.

Nitrogen fertilization greatly increased soil nutrient availability, boosted SOC, and resulted in a higher soil carbon pool [13]. However, long-term excessive N fertilization leads to increased losses of N through ammonia volatilization [14], NO₃⁻ leaching [15], and N₂O emissions [6]. Conversely, combining straw incorporation with N fertilization not only improves soil fertility and agriculture sustainability but also reduced environmental pollution [6]. The gradual release of nutrients from decomposing straw mitigates N losses, thereby enhancing soil quality and promoting sustainable crop productivity [16]. Research indicates that maize grain yield increases with N fertilization rate up to 200 kg N ha⁻¹, but higher rates, such as 300 kg N ha⁻¹, substantially decrease grain yield [17, 18]. The yield reduction could stem from soil acidification, which accelerates the decomposition of soil organic and inorganic carbon and is particularly noted in southern China [19, 20]. Such acidification adversely affects the growth of soil microorganisms and their contribution to SOC pool [21].

China, a leading producer of crop residues [22], generated 598 million tons of crop straw in 2017. Straw incorporation, a prevalent and beneficial practice, enhances soil fertility, especially SOC, through decomposition and reduced reliance on chemical N fertilization [23, 24]. It also increased soil enzymatic activities, which are a primary source and key indicator of soil quality [16, 25]. Soil organic carbon is crucial in defining soil's physical, chemical, and biological soil properties [26], improving soil structure, nutrient availability, and soil microbial biomass and activity [27]. The faster decomposition of residues and SOC turnover mainly depends on the structure and activity of soil microbial biomass [27, 28]. Straw incorporation, which facilitates close contact with

soil, enhances soil microbial biomass and activity, making it highly responsive to changes in soil quality [28]. The response of SOC to N fertilization is greatly affected by environmental and agriculture management practices like crop residue incorporation, fertilizer application, and tillage management [29, 30]. Straw incorporation directly increases SOC, SMBC, and N thereby improving soil fertility [31, 32] and mitigate the negative impacts of excessive chemical N fertilization [15, 33]. Moreover, it reduces evaporation, increases soil moisture content and water use efficiency, leading to enhanced plant growth and grain yield [28, 34]. Changes in soil enzymes or soil degradation affect soil fertility, maintaining environmental stability and soil quality [15].

Incorporating straw into soil notably enhances the interaction between straw and soil microbes [31], concurrently boosting and activating soil enzyme activities which elevate nutrient mobilization [35]. Urease, an extracellular enzyme in soil, aids in converting organic N into ammonium N, thus making it more readily available for plants adsorption by soil particles [27]. Found adsorbed on clay particles or within humic complexes, urease released from both living and decomposed microbial organisms. Phosphatase enzymes, potentially originating from the plant roots associated with mycorrhiza, other fungi, or soil bacteria, catalyze the conversion of organic phosphate esters and anhydrides to inorganic phosphorus [27, 36, 37]. Additionally, straw incorporation not only enhances soil organic matter and significantly impacts soil cellulase activity but also markedly increases SOC and active C fractions [8]; these outcomes demonstrate a substantial and positive correlation between SOC and soil sucrase activity.

Organic and inorganic fertilization in combination significantly improves soil physical, chemical, and biological properties [38, 39]. S Huang, et al. [40] reported that straw incorporation increased crop yield in subtropical areas because warm weather accelerates the breakdown of incorporated straw, thus increasing the availability of soil nutrients [41]. Additionally, the open burning of agricultural straw emits dangerous air pollution and is prohibited in China [42]. Few studies have investigated the comparative effects of straw incorporation combined with N fertilization on SOC, STN, SMBC, and soil enzyme activities in a double cropping system. However, no study has reported the effect of straw incorporation with and without N fertilization on soil fertility, SMBC, and soil enzymes across different seasons in South China. Therefore the aims of the study were: (1) to investigate the effect of straw incorporation with N fertilization on soil fertility and soil enzyme activities under a double-cropping system; (2) to determine the effect of growth stages and seasons on straw decomposition and crop yield; and (3) to understand the seasonal differences with

and without straw incorporation on soil fertility, enzyme activities, and maize grain yield in subtropical monsoon humid region, South China. We hypothesized that straw incorporation with N200 treatment would economically improve STN, SOC, and SMBC, urease, cellulase, sucrose, catalase, acid phosphatase activities, maize grain yield. We also hypothesized that V6 growth stage would increase soil urease, sucrose, catalase, and acid phosphatase activities under both season.

Materials and methods

Experimental site

The field experiment was performed at the Agronomy Research Farm of Guangxi University, Nanning, Guangxi, China (22°50' N, 108°17' E). This region is characterized by a subtropical climate with a mean annual precipitation of 1298.0 mm and a mean annual temperature of 21.7 °C [15]. Generally, this region has two distinct maize growing seasons: spring growing season from March to July and autumn from August to November. According to Chinese Soil Taxonomy, the soil was classified as clay loam, with a pH of 6.5, SOC of 14.6 g kg⁻¹, total N, available phosphorus, and potassium of 0.80 g kg⁻¹, 0.43 g kg⁻¹, and 0.89 g kg⁻¹, respectively.

Experimental design

The experiment was carried out in a randomized complete block design (RCBD) with three replications in a split-plot arrangement whereas the straw management was allotted to the main plot, and N fertilization treatments were allotted to the sub-plot. The two straw management were straw incorporation (maize straw incorporated) and traditional planting (maize straw removed) during spring and autumn seasons in 2020 and 2021. The N fertilization treatments were N0 (0 kg N ha⁻¹), N100 (100 kg N ha⁻¹), N150 (150 kg N ha⁻¹), N200 (200 kg N ha⁻¹), N250 (250 kg N ha⁻¹), and N300 (300 kg N ha⁻¹).

The “Zhengda-619” maize variety was planted twice in the spring and autumn seasons. During 2020 the maize was sown on 11th March and 2nd August, and harvested on 9th July and 30th November in spring and autumn seasons, respectively. Likewise, during 2021, the maize was sown on 4th March and 8th August, and harvested on 5th July and 26th November in spring and autumn seasons, respectively. The sub-plot size was 4.2×4.2 m² having a planting density of 55,556 plants ha⁻¹ with row-to-row spacing of 60 cm and plant-to-plant spacing of 30 cm. Calcium magnesium phosphate (P₂O₅ 18%) and potassium chloride (K₂O 60%) were incorporated at the rate of 100 kg ha⁻¹ into the soil before sowing as a basal recommended fertilizer. The field management followed standard agricultural practices, with two-thirds of the N

fertilizer applied before sowing and the remaining one-third applied during the 12-leaf stage.

Soil sampling

Before maize harvest, at the jointing stage (V6) and maturity stage (R6), five random soil samples were taken from each sub-plot at a depth of 0–20 cm soil depth. The collected soil samples were completely mixed, sieved through a mesh of 2 mm to remove the stones, roots and gravel. The sieved soils were then divided into two sections, one immediately kept at 4 °C in the refrigerator for soil microbial carbon analysis. The remaining portion of the soil samples was air-dried at room temperature and sieved through 0.069 mm mesh for determining enzyme activities and/or 0.15 mm mesh for determining STN and SOC.

Soil fertility and enzyme activities analysis

Soil microbial biomass carbon was measured by the chloroform fumigation culture method [43]. Soil organic carbon was quantified using volumetric potassium dichromate and an external heating method previously used by [44], whereas the semi-micro kelvin method was used to measure STN content [45]. Soil enzyme activities were analyzed using Solarbio Science & Technology Co. (Beijing, China). The activities of soil urease (BC0125; BC0245), sucrose (BC0155S), cellulase (BC0105), catalase (BC0145), and acid phosphatase (S-ACP) were determined using the soil enzymatic kits.

Grain yield

At the maturity growth stage of maize, an area of 2 m² was randomly selected in each plot, and the grain yield was determined after maize threshing and drying.

$$\text{Grain yield (kg ha}^{-1}\text{)} = \text{maize grain weight (kg m}^{-2}\text{)} \times 10000 \text{ (m}^2\text{)} \quad (1)$$

Statistical analysis

The analysis of variance (ANOVA) was used to analyze STN, SOC, SMBC, soil enzymatic activities, and grain yield using an *f*-test (SPSS Inc., Chicago, IL, USA). Straw management and N fertilization rates were fixed effects. However, seasons and years were repetitive measure factors and fixed effects, whereas all the interactions were taken as fixed effects except treatment × replication interaction, which was taken as a random effect. Duncan's multiple range test (Duncan, *P*<0.05) was used to compare the mean value of the three replications within treatments. Data are expressed as the mean and ± standard deviation (SD). All graphs were drawn with Origin Pro, Version 2021 (Origin Lab Corporation, Northampton, MA, USA). The GGally package was used in R v.3.63

to evaluate the relationship between the N fertilization rates and plant enzyme activities.

Results

Soil organic carbon

The incorporation of maize straw, N fertilization, various growth stages, seasons, and annual variation positively affected SOC (Fig. 1). In both years, the highest SOC content was observed at the V6 growth stage under straw incorporated plot, while the lowest was observed at R6 under traditional planting (Fig. 1). Averaged across seasons, stages, and straw management, 2021 resulted in significantly higher SOC content compared to 2020. Similarly, the spring season had significantly higher SOC compared to the autumn season in both years. The lower SOC in autumn season might be due to higher temperatures, lower solar radiation, and a shorter growing season for autumn crops. Moreover, SOC significantly increased with increasing N fertilization in both growth stages and seasons. Compared to N0, the SOC content was significantly higher for N300 treatment under both years and seasons. There were no significant differences between N200 and N250 in straw management and

years, while there were differences among N200, N250, and N300 treatments in 2020 under traditional planting. The interaction between N fertilization and straw management was not significant, except in spring season of 2020 (Fig. 1A). Our results showed that the N200 treatment coupled with straw returning, is the best choice to reduced the N fertilization use and increase SOC content, with increases of 0.74% and 0.61% compared to N250 and N300 under traditional planting (Fig. 1).

Soil total nitrogen

Soil total N content under different straw management and N fertilization across two seasons is shown in Fig. 2. The effect of straw incorporation with N fertilization on STN was statistically higher at V6 than at the R6 growth stage in both seasons and years. Similarly, under traditional planting, the STN content at the V6 growth stage was 6.42 and 3.98% higher in the spring and autumn seasons during 2020 and 9.46 and 9.56% higher in spring and autumn seasons during 2021, respectively (Fig. 2A-C). On average, the V6 growth stage had 6.55 and 10.13% higher STN content compared to R6 growth stage in 2020 and 2021. Compared to the autumn season, the spring season

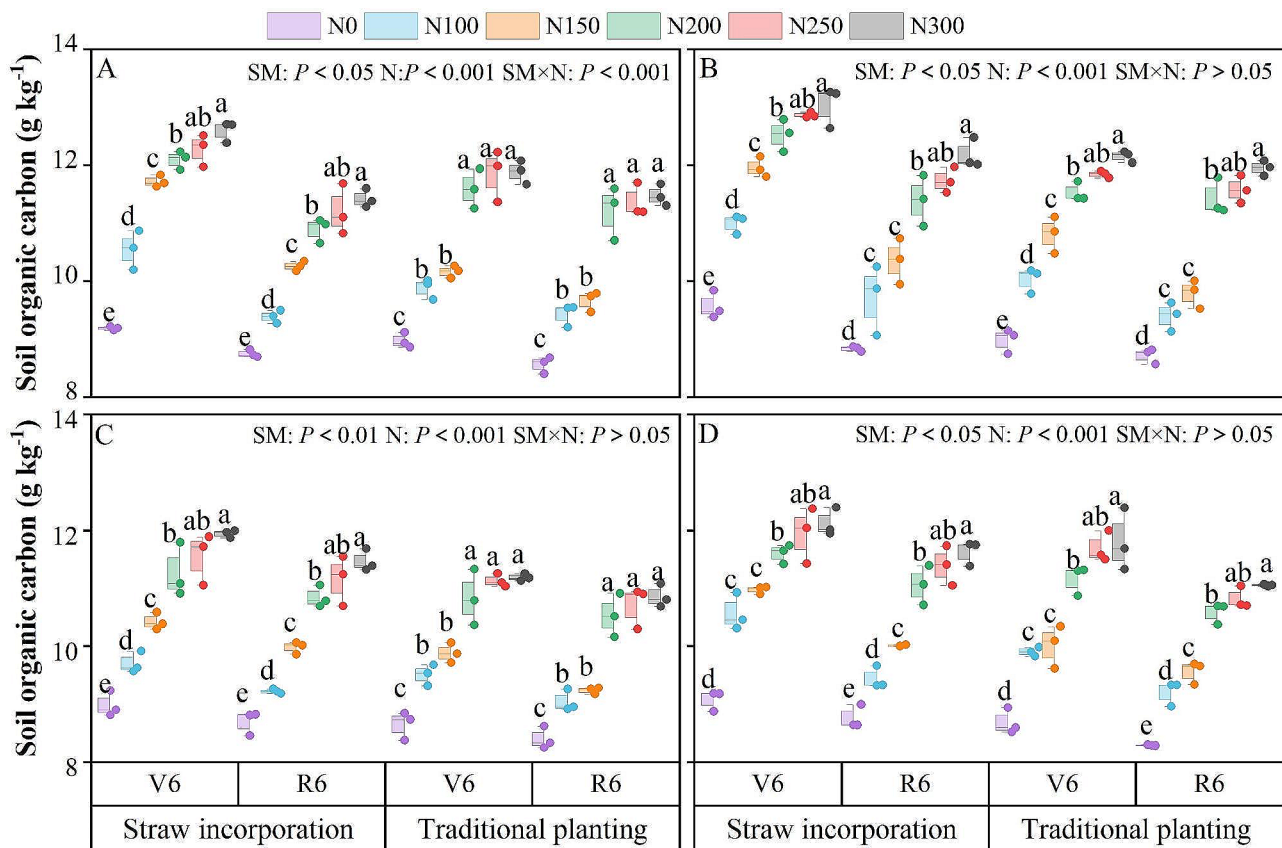


Fig. 1 Effect of straw incorporation and nitrogen fertilization on soil organic carbon (SOC) under dual-cropping system during spring in 2020 (A) and 2021 (B), and autumn seasons in 2020 (C) and 2021 (D). SM: straw management, N: nitrogen fertilization. Different letters indicate significant differences between samples ($P < 0.05$)

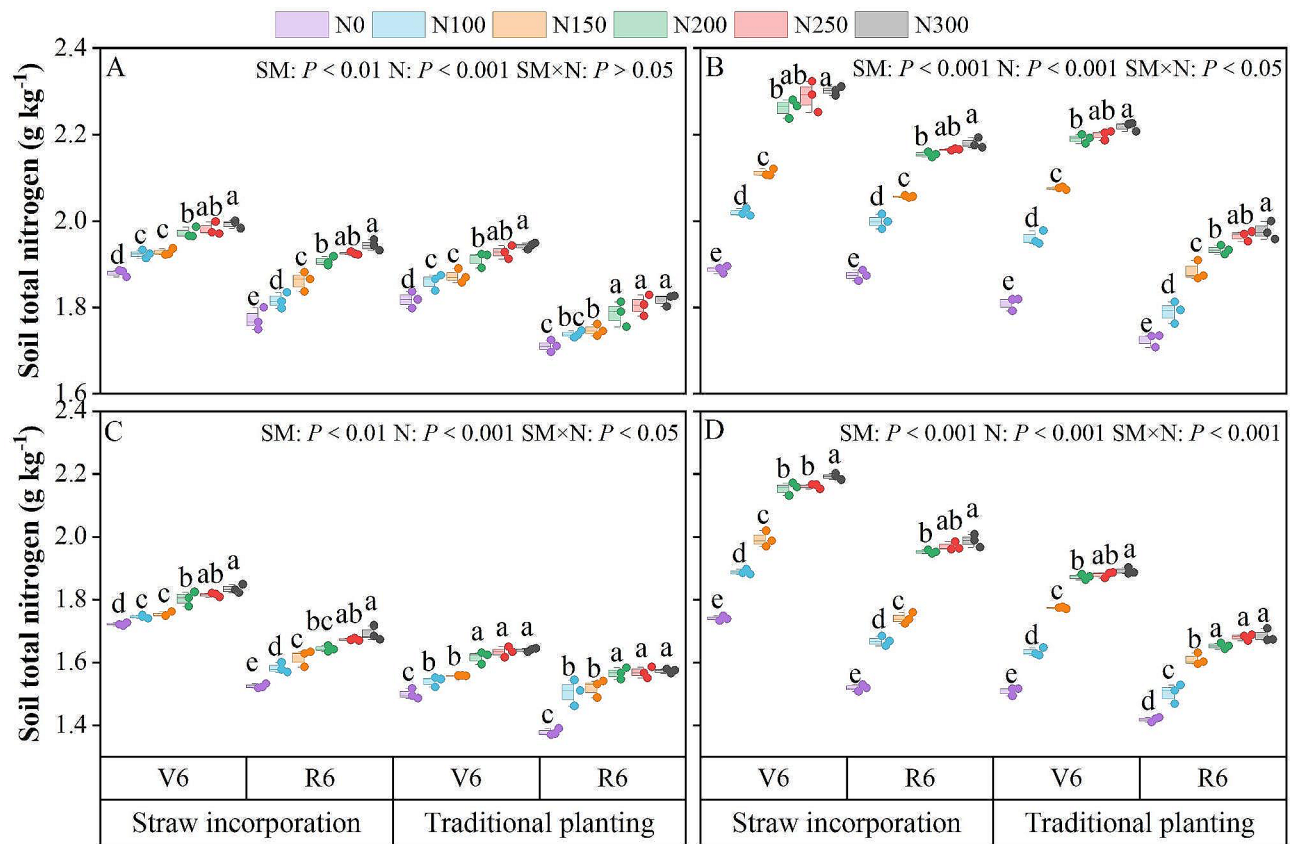


Fig. 2 Effect of straw incorporation and nitrogen fertilization on soil total nitrogen under dual-cropping system during spring in 2020 (A) and 2021 (B), and autumn seasons in 2020 (C) and 2021 (D). The names of the treatments are the same as in (Fig. 1). Different letters indicate significant differences between samples ($P < 0.05$)

increased STN content by 13.02 and 12.16% in 2020 and 2021, respectively. Additionally, the results demonstrated that the overall STN content was 1.75 g kg⁻¹ in 2020 and 1.92 g kg⁻¹ in 2021, suggesting that t STN in 2021 was 8.85% higher than in 2020. Our results showed that STN content increased with increasing N fertilization rates in both seasons, straw management, and years. Moreover, the STN content was significantly higher for N300 treatment compared to other N fertilization rates but not significantly different from N250 (Fig. 2A-D). These results suggest that straw incorporation significantly increased the STN content under the same N fertilization rates. The interaction between straw incorporation and N fertilization was significant in the autumn season of 2020, and in both seasons of 2021 ($P < 0.05$; Fig. 1B and D).

Soil microbial biomass carbon

Soil microbial biomass carbon significantly increased with N fertilization rates and maize straw incorporation into the field ($P < 0.05$; Fig. 3). Soil microbial biomass carbon was significantly higher at the V6 growth stage under straw incorporation, while the lowest was observed at R6 growth stage under traditional planting. On an average,

straw incorporation in spring and autumn seasons significantly increased the SMBC by 15.43 and 32.64% during 2020 (Fig. 3A and C) and by 17.30 and 22.87% during 2021 compared to traditional planting (Fig. 3B and D). The results showed that the spring season resulted in higher SMBC (178.98 mg C kg⁻¹ of soil) compared to autumn season (134.35 mg C kg⁻¹ of soil). Moreover, our results demonstrated that SMBC significantly increased with increasing N fertilization. The results also showed that the N300 treatment had significantly higher SMBC compared to the N0, N100, N150 treatments ($P < 0.05$), but but was not statistically different from the N200 and N250 treatments ($P > 0.05$). Soil microbial biomass carbon was significantly higher in straw incorporated plots compared to traditional planting with the same N fertilization rates, might be due to higher decomposition and SOC content. The interaction between straw incorporation and N fertilization rates was not significant, except in autumn 2021.

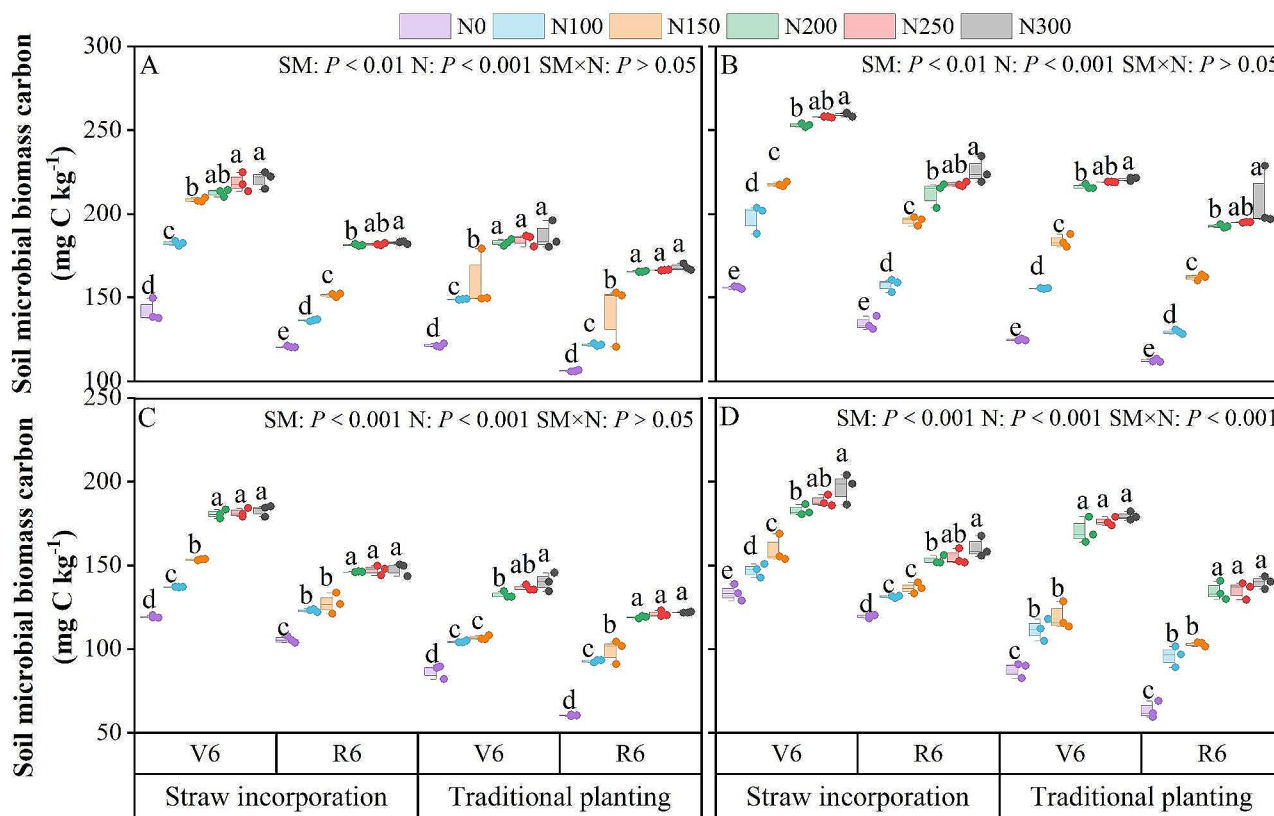


Fig. 3 Effect of straw incorporation and nitrogen fertilization on soil microbial biomass carbon under dual-cropping system during spring in 2020 (A) and 2021 (B), and autumn seasons in 2020 (C) and 2021 (D). The names of the treatments are the same as in (Fig. 1). Different letters indicate significant differences between samples ($P < 0.05$)

Soil enzyme activities

Soil urease

Soil urease activity increased significantly with higher N fertilization rates across both seasons, growth stages, and years (Fig. S1). Plots incorporated straw exhibited 5.64 and 9.56% higher soil urease activity in 2020 and 2021, respectively, compared to traditional planting (Table 1). The lowest urease activities were observed in the N0 treatment (218.13 $U\ g^{-1}$ and 228.11 $U\ g^{-1}$), while the highest were in the N300 treatment (284.23 $U\ g^{-1}$ and 305.60 $U\ g^{-1}$), which did not differ from the N250 treatment (276.67 $U\ g^{-1}$ and 303.39 $U\ g^{-1}$) in 2020 and 2021, respectively. These results showed that N250 treatment increased soil urease activity by 26.84, 16.90 and 10.54% in 2020 and by 33.00, 20.31 and 12.59% in 2021, compared to N0, N100, and N150 treatments, respectively (Table 1). The interaction between straw management and N fertilization was significant in 2021 ($P < 0.001$). In addition, the soil urease activity was significantly higher at the V6 growth stage in spring 2021 and lower at the R6 growth stage in autumn 2020 (Table 2). Regressions analysis showed that soil urease activity increased positively and polynomially with SOC ($R^2 = 0.94$; $P < 0.001$; Fig. 4A),

STN ($R^2 = 0.84$; $P < 0.001$; Fig. 4B), and increasing SMBC ($R^2 = 0.95$; $P < 0.001$; Fig. 4C).

Soil cellulase

Soil cellulase activity was significantly affected by N fertilization, growth stages, seasons, and years (Fig. S2). The N300 treatment significantly increased soil cellulase activity compared to other N fertilization rates in 2020, but it was not significantly different from N250 in 2021 (Table 1). Similarly, N250 treatment was statistically similar to N200 treatment, suggesting that N200 treatment with straw incorporation is a better choice for improved soil enzyme activity and reduced N fertilization. Compared to traditional planting, straw incorporation significantly increased soil cellulase by 9.43 and 11.25% in 2020 and 2021, respectively (Table 1). Moreover, soil cellulase activity was significantly higher at R6 during the autumn of 2021 compared to V6 during the spring of 2020 (Table 2). All interactions were significant except among years, seasons, and growth stages. Regressions analysis showed that soil cellulase activity increased positively and polynomially with increasing SOC ($R^2 = 0.90$; $P < 0.001$; Fig. 4D), STN ($R^2 = 0.85$; $P < 0.001$; Fig. 4E), and SMBC ($R^2 = 0.93$; $P < 0.001$; Fig. 4F). This increase in soil

Table 1 Effect of straw incorporation and nitrogen fertilization on soil enzyme activities under dual-cropping system in 2020–2021

Year	Straw management (SM)	Nitrogen (N)	S-UE U g ⁻¹	S-CL U g ⁻¹	S-SC U g ⁻¹	S-CAT U g ⁻¹	S-ACP U g ⁻¹
2020	Straw incorporation		262.22 ± 0.21a	11.09 ± 0.04a	29.78 ± 0.13a	18.74 ± 0.14a	19055.98 ± 67.06a
	Traditional planting		244.20 ± 1.20b	10.14 ± 0.02b	27.16 ± 0.04b	17.38 ± 0.02b	17181.53 ± 17.43b
		N0	218.14 ± 2.74e	8.61 ± 0.02f	22.18 ± 0.17f	15.39 ± 0.00f	15653.32 ± 91.43f
		N100	229.40 ± 0.89d	9.94 ± 0.02e	25.36 ± 0.33e	17.04 ± 0.18e	16996.61 ± 23.29e
		N150	245.45 ± 0.66c	10.54 ± 0.02d	27.19 ± 0.19d	17.65 ± 0.11d	17689.71 ± 62.81d
		N200	269.37 ± 3.66b	11.26 ± 0.10c	30.82 ± 0.24c	18.87 ± 0.14c	18998.86 ± 108.36c
		N250	275.89 ± 1.37a	11.55 ± 0.03b	32.26 ± 0.08b	19.38 ± 0.13b	19474.45 ± 85.00b
		N300	281.02 ± 1.25a	11.79 ± 0.13a	33.01 ± 0.28a	20.06 ± 0.23a	19899.60 ± 67.64a
ANOVA							
	SM		**	***	**	*	**
	N		***	***	***	***	***
	SM×N		ns	***	ns	ns	***
2021	Straw incorporation		297.50 ± 0.50a	11.70 ± 0.06a	35.04 ± 0.04a	24.68 ± 0.10a	25420.27 ± 119.01a
	Traditional planting		265.74 ± 0.07b	10.52 ± 0.03b	30.70 ± 0.05b	20.61 ± 0.04b	21471.37 ± 97.76b
		N0	230.51 ± 1.19e	9.16 ± 0.04e	26.97 ± 0.23e	19.96 ± 0.10e	18949.61 ± 201.56e
		N100	256.31 ± 0.14d	10.34 ± 0.01d	30.23 ± 0.25d	21.50 ± 0.01d	20727.29 ± 120.70d
		N150	273.59 ± 1.45c	10.78 ± 0.06c	32.39 ± 0.00c	22.30 ± 0.13c	23379.49 ± 97.97c
		N200	307.98 ± 0.73b	11.92 ± 0.14b	35.46 ± 0.06b	23.80 ± 0.08b	25609.98 ± 54.00b
		N250	309.76 ± 0.99ab	12.12 ± 0.06ab	35.87 ± 0.02ab	24.07 ± 0.13ab	25865.50 ± 112.51ab
		N300	311.57 ± 0.42a	12.33 ± 0.06a	36.29 ± 0.07a	24.26 ± 0.04a	26143.03 ± 59.38a
ANOVA							
	SM		***	***	***	**	**
	N		***	***	***	***	***
	SM×N		***	*	***	ns	***

Note: Means followed by different lowercase letters within each column indicate significant differences ($p < 0.05$) using LSD test. Values are means ± SE ($n = 3$). Whereas, the nitrogen treatments N0, N100, N150, N200, N250, and N300 represent the application of nitrogen at the rate of 0, 100, 150, 200, 250, and 300 kg ha⁻¹. *Significant at $P < 0.05$; **Significant at $P < 0.01$; ***Significant at $P < 0.001$; ns, not significant

Table 2 Soil enzyme activities under dual-cropping system in V6 and R6 during spring and autumn seasons in 2020–2021

Year (Y)	Season (S)	stages (St)	S-UE U g ⁻¹	S-CL U g ⁻¹	S-SC U g ⁻¹	S-CAT U g ⁻¹	S-ACP U g ⁻¹
2020			253.21 ± 0.62b	10.61 ± 0.03b	28.47 ± 0.05b	18.06 ± 0.07b	18118.76 ± 36.98b
2021	Spring		281.62 ± 0.29a	11.11 ± 0.04a	32.87 ± 0.03a	22.65 ± 0.04a	23445.82 ± 89.52a
			290.12 ± 0.91a	8.99 ± 0.02b	32.67 ± 0.09a	19.11 ± 0.09a	19424.67 ± 39.12a
	Autumn		216.30 ± 0.33b	12.24 ± 0.04a	24.27 ± 0.05b	17.01 ± 0.06b	16812.84 ± 35.25b
			269.77 ± 1.50a	10.34 ± 0.04b	30.12 ± 0.05a	19.36 ± 0.10a	19437.24 ± 25.42a
		R6	236.65 ± 0.38b	10.90 ± 0.05a	26.83 ± 0.07b	16.76 ± 0.04b	16800.27 ± 70.95b
ANOVA							
	Y		***	**	***	***	***
	S		***	***	***	***	***
	St		***	***	***	***	***
	Y×S		***	***	***	***	ns
	Y×St		***	***	***	***	***
	S×St		***	*	***	**	***
	Y×S×St		**	ns	*	***	***

Note: Means followed by different lowercase letters within each column indicate significant differences ($p < 0.05$) using LSD test. Values are means ± SE ($n = 3$). Whereas, the nitrogen treatments N0, N100, N150, N200, N250, and N300 represent the application of nitrogen at the rate of 0, 100, 150, 200, 250, and 300 kg ha⁻¹. *Significant at $P < 0.05$; **Significant at $P < 0.01$; ***Significant at $P < 0.001$; ns, not significant

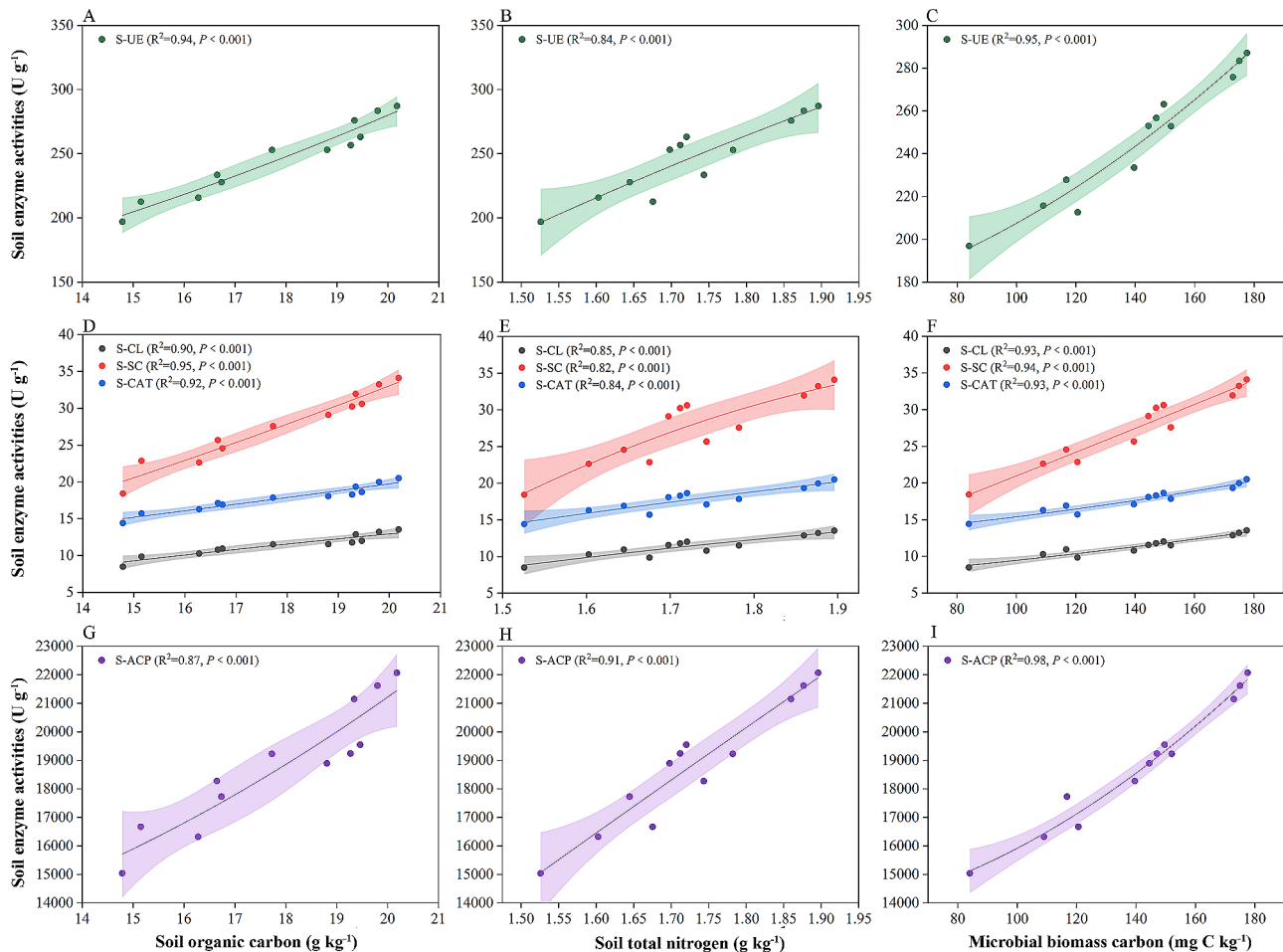


Fig. 4 Relationships of soil organic carbon (A, D, G), total nitrogen (B, E, H), microbial biomass carbon (C, F, I) with soil enzyme activities

cellulase activity could be attributed to higher SOC, STN, and SMBC due to optimum N fertilization rates and straw incorporation.

Soil sucrase

Similar to soil urease, sucrase activity was significantly higher in spring compared to autumn (Fig. S3). Soil sucrase activity was significantly higher in the V6 growth stage compared to R6 in both seasons and years (Fig. S3). Nitrogen fertilization with straw incorporation significantly increased soil sucrase activity. Additionally, soil sucrase activity increased with higher N fertilization rates, suggesting that N300 treatment increased soil sucrase activity by 48.79, 30.16, 21.39, 7.12, and 2.32% compared to N0, N100, N150, N200, and N250 in 2020 (Table 1). However, in 2021 the N300 and N250 treatments had a statistically similar soil sucrase activity, whereas the N250 and N200 were also statistically similar. Soil sucrase activity was significantly higher at the V6 growth stage in spring 2021 compared to the R6 growth stage in autumn 2020 (Table 2). Soil sucrase activity was 15.17% higher in 2021 compared to 2020,

with the highest activity observed in spring (35.36 U g⁻¹) compared to autumn (26.68 U g⁻¹). Regression analysis showed a positive relationship between soil sucrase activity and SOC, STN, and SMBC (Fig. 4D, E, and F). These results suggest that soil enzyme activities correlate with higher soil nutrient availability and SMBC. The higher soil sucrase activity in straw incorporated plots might be due to higher SOC, STN, and SMBC.

Soil catalase

The potential activity of soil catalase was determined at two growth stages (V6 and R6) in the spring and autumn seasons of 2020 and 2021 (Fig. S4). Soil catalase activity was significantly higher at V6 growth stage compared to R6 in both seasons and years, with activity significantly higher in 2021 than in 2020 (Table 1; Fig. S4). The activity in straw incorporated plots was 5.70 and 19.74% higher in 2020 and 2021, respectively, compared to traditional planting. In 2020, the N300 treatment resulted in 29.91, 17.10, 13.07, 5.67 and 3.04% higher catalase activity compared to N0, N100, N150, N200, and N250 treatments, respectively (Table 1). There were no significant

differences between N300 and N250 treatments or N250 and N200 treatments in 2021, possibly due to higher straw decomposition and microbial activity compared to 2020. Furthermore, the activity in 2021 was 21.06% higher than in 2020 (Table 2). On average, the activity was significantly higher in spring (22.90 U g^{-1}) compared to autumn (17.62 U g^{-1}), suggesting that the spring season increased the catalase activity by 29.97% (Table 2). The results showed that activity was much higher at V6 than at the R6 growth stage, likely due to higher soil nutrients and SMBC at V6. Soil catalase was positively and significantly correlated with SOC ($R^2=0.92$; $P<0.001$; Fig. 4D), STN ($R^2=0.84$; $P<0.001$; Fig. 4E), and SMBC ($R^2=0.93$; $P<0.001$; Fig. 4F). Our results demonstrated that higher soil fertility and SMBC could be the possible reason for improved soil enzymatic activity.

Soil acidic phosphatase

Nitrogen fertilization with and without straw incorporation significantly increased soil acidic phosphatase activity in both years during the spring and autumn seasons (Fig. S5). The activity was significantly enhanced with straw incorporation in both seasons and years, with the highest activity observed in spring 2021 (Fig. S5; Table 2). Compared to traditional planting, straw incorporated plots enhanced acidic phosphatase activity by 10.91 and 18.39% in 2020 and 2021, respectively (Table 1). Acidic phosphatase activity significantly increased with increasing N fertilization rates, with N300 resulting in 27.13% higher activity than N0 treatment in 2020 (Table 1). However, in 2021, acidic phosphates activity was statistically similar for N300 and N250 treatments, and for N250 and N200 treatments. The interaction between straw management and N fertilization was significant ($P<0.001$). Moreover, soil acidic phosphatase activity was significantly higher at the V6 growth stage during the spring of 2021 and the lowest at R6 growth stage during the autumn of 2020 (Table 2). Regression analysis explained that soil acidic phosphatase activity was positively correlated with SOC, STN, and SMBC. These results suggest that soil acidic phosphatase activity significantly increased with increasing SOC ($R^2=0.87$; $P<0.001$; Fig. 4G), STN ($R^2=0.91$; $P<0.001$; Fig. 4H), and SMBC ($R^2=0.98$; $P<0.001$; Fig. 4I).

Grain yield

The maize grain yield was significantly affected by N fertilization, straw management, seasons, and their interactions (Table 3). Grain yield increased significantly with higher N fertilization during both seasons and years. In 2020, the N300 treatment significantly increased grain yield by 144.35 and 160.83% in the spring and by 155.26 and 236.19% in autumn compared to N0 treatment in straw incorporated and traditional planting, respectively

(Table 3). There were no significant differences between the N300 and N250 treatments and between N250 and N200 in both seasons and straw management. In the of 2021, the N300 increased grain yield by 137.14, 70.08, 35.62, 7.79 and 3.75% in straw incorporated plots, and by 167.61, 71.95, 35.71, 7.34 and 5.56% in traditional planting compared to N0, N100, N150, N200, and N250 treatments, respectively. The mean results showed that N300 treatment significantly increased grain yield compared to other N fertilization rates in both straw management and years. Our results suggest that N200 treatment in straw incorporated plots significantly increased maize yield by 8.30 and 4.22% compared to N250 and N300 treatments of traditional planting systems. Therefore, the N200 treatment with straw incorporation economically increased crop yield and agriculture sustainability while reducing the negative impact of N fertilization. Compared to traditional planting, straw incorporated plot increased grain yield by 10.79 and 12.06% in the spring and autumn of 2020 and 9.77 and 10.71% in the spring and autumn of 2021, respectively (Table 3). Average across the seasons and straw management, the highest grain yield was obtained in 2021 compared to 2020. On average, the N300 treatment resulted in significantly higher grain yield than other N fertilization rates. The interaction between straw management and N fertilization was significant in both season during 2020 but had no significant effect during 2021.

Correlation analysis

The correlation analysis revealed that soil sucrose, catalase, and acidic phosphatase activities were positively and strongly correlated with soil urease activity ($P<0.001$; Fig. 5). Soil cellulase activity was negatively correlated with urease activity under the N100 treatment ($P<0.05$). Soil catalase and acidic phosphatase activities were positively correlated with soil cellulase activity ($P<0.05$), while soil sucrose activity was negatively correlated under N150 treatment. Additionally, soil catalase and acidic phosphatase activities were positively correlated with soil sucrose activity. Acidic phosphatase was strongly and positively correlated with catalase ($P<0.001$; Fig. 5). Soil urease activity was strongly correlated with sucrose and catalase activities at all the N fertilization rates. Similarly, sucrose was highly correlated with catalase, and catalase with acidic phosphatase activity in all N fertilization rates.

Discussion

Overall, N fertilization and straw incorporation considerably improved maize growth performance. However, lower soil fertility and yield were found under the traditional planting system. The four factors such as SOC, STN, soil microbial biomass, and soil enzyme activities

Table 3 Effect of straw incorporation and nitrogen fertilization on grain yield under dual-cropping system during spring and autumn in 2020–2021

Straw management (SM)	Nitrogen (N)	2020 (kg ha ⁻¹)		2021 (kg ha ⁻¹)	
		Spring	Autumn	Spring	Autumn
Straw incorporation	N0	2728 ± 54e	2533 ± 33e	2917 ± 83e	2667 ± 83e
	N100	3967 ± 33d	3550 ± 50d	4067 ± 17d	3683 ± 44d
	N150	5033 ± 33c	4583 ± 44c	5100 ± 29c	4667 ± 17c
	N200	6483 ± 44b	6175 ± 75b	6417 ± 83b	6250 ± 144b
	N250	6600 ± 29ab	6333 ± 83ab	6667 ± 83ab	6417 ± 83ab
	N300	6667 ± 44a	6467 ± 33a	6917 ± 167a	6667 ± 83a
Traditional planting	N0	2332 ± 38e	1750 ± 144e	2367 ± 73e	1750 ± 0e
	N100	3667 ± 83d	3067 ± 34d	3683 ± 44d	3333 ± 83d
	N150	4367 ± 73c	4200 ± 50c	4667 ± 44c	4517 ± 17c
	N200	5750 ± 58b	5500 ± 144b	5900 ± 76b	5667 ± 83b
	N250	5883 ± 60ab	5667 ± 83ab	6000 ± 144b	5833 ± 83ab
	N300	6083 ± 83a	5883 ± 73a	6333 ± 83a	6000 ± 144a
Straw incorporation		5246 ± 7a	4940 ± 35a	5347 ± 34a	5058 ± 27a
Traditional planting		4680 ± 6b	4397 ± 59b	4825 ± 25b	4344 ± 67b
	N0	2531 ± 15f	2142 ± 58f	2642 ± 74e	2208 ± 42e
	N100	3817 ± 36e	3401 ± 35e	3875 ± 25d	3375 ± 38d
	N150	4700 ± 52d	4458 ± 60d	4883 ± 8c	4433 ± 17c
	N200	6117 ± 22c	5838 ± 44c	6158 ± 46b	5875 ± 125b
	N250	6242 ± 17b	6000 ± 0b	6333 ± 110b	6042 ± 42b
	N300	6375 ± 63a	6175 ± 38a	6625 ± 72a	6275 ± 76a
ANOVA	SM	***	*	***	**
	N	***	***	***	***
	SM×N	**	**	ns	ns

Note: Means followed by different lowercase letters within each column indicate significant differences ($p < 0.05$) using LSD test. Values are means ± SE ($n = 3$). Whereas, the nitrogen treatments N0, N100, N150, N200, N250, and N300 represent the application of nitrogen at the rate of 0, 100, 150, 200, 250, and 300 kg ha⁻¹. *Significant at $P < 0.05$; **Significant at $P < 0.01$; ***Significant at $P < 0.001$; ns, not significant

are responsible for the distinct impacts of N fertilization under straw incorporated and traditional planting systems. Straw incorporation is well documented to increase soil fertility by providing accessible C and N substrates for microbial populations [4, 6]. Soil organic carbon is an important and useful indicator of soil fertility, impacting crop development both directly and indirectly [46], and its fractions are sensitive to changes in C supply [47]. Similarly, J Wang, et al. [48] also reported that straw incorporation significantly increased SOC and total N compared to traditional planting, indicating improved soil fertility. Previous meta-analysis showed that residue incorporation with a high C/N ratio or lower N content dramatically increased straw decomposition through soil microorganisms, thereby improving SOC content [49]. In addition, straw incorporation boosted soil microbial activity and biomass, accelerating SOC decomposition and improving soil nutrient content [50, 51].

Straw incorporation positively correlated with soil nutrient content [52], and increased biomass incorporation and decomposition led to an increase in soil organic matter [53]. It also reduces SOC losses from the agroecosystem and increases crop productivity and

sustainability [54]. In the current study, straw incorporation significantly increased SOC content, especially in spring season, compared to traditional planting. In contrast, traditional planting drastically decreased SOC content by 3.87 and 5.09% during 2020 and 2021 compared to straw incorporation (Fig. 1). Compared to the R6 growth stage, the incremental changes in SOC content at the V6 growth stage were 7.31 and 8.48% higher in straw incorporated plots, and 4.18 and 5.10% higher in traditional planting during 2020 and 2021, respectively. These results suggested that straw incorporation into the soil is essential to maintaining SOC levels [32]. Additionally, our finding revealed that continuous straw incorporation significantly and sustainably boosted soil fertility in both years, independent of N fertilization in double-cropping system [6, 55]. Moderate N application increases SOC content and eventually increases crop yield [52, 56], while high amounts of N application encourage SOC mineralization, which decreases SOC content and crop yield [57]. Straw incorporation with the N200 treatment significantly increased SOC content compared to N0, N100, N150, N250, and N300 treatments of traditional planting. Consistent with our results, ME Duval, et al. [58] also

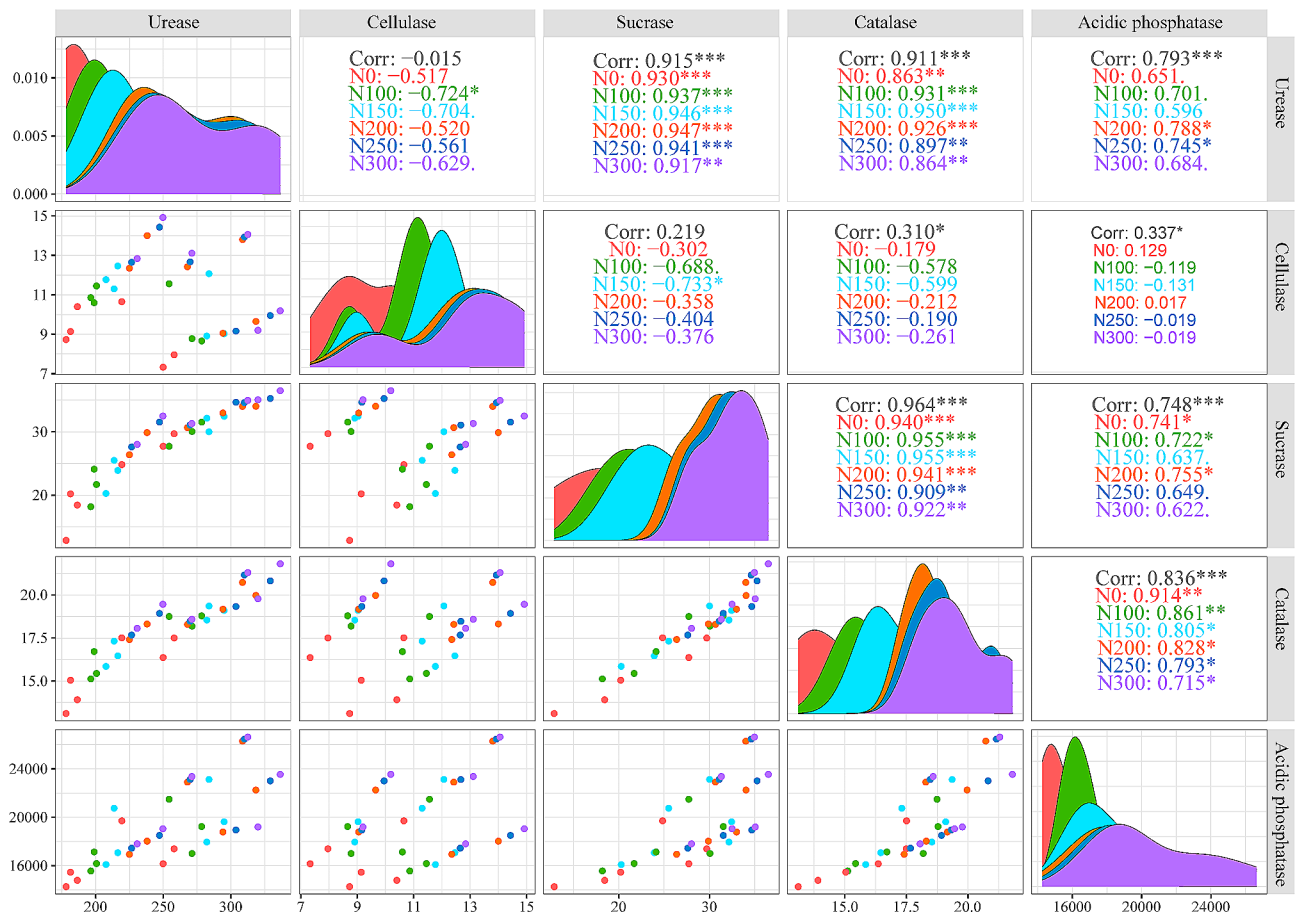


Fig. 5 Correlation analysis among soil enzyme activities and nitrogen fertilization rates that are significantly positively/negatively correlated with each other. ** $P \leq 0.05$, *** $P \leq 0.01$, and **** $P \leq 0.001$

reported a strong and positive relationship between carbon input and SOC storage.

Nitrogen is an essential macronutrient and is well understood as one of the key limiting nutrients for plant growth and development [59]. Exogenous application of N improves plant growth and increases crop yield [56]. However, the increase in soil N content with N fertilization might not be the main factor ameliorating maize growth under straw incorporated plots might be due to higher decomposition rates (Fig. 2). Similarly to N fertilization, straw incorporation also releases more N content into the soil and becomes available to plants via microbial mineralization [28]. The current results showed that straw incorporation with the N200 treatment resulted in higher STN content compared to the N250 and N300 treatments of traditional planting. These results suggest that straw incorporation drastically reduces the excessive use of N fertilization and slowly releases nutrients to the soil, reducing water pollution [56], soil acidification, and other adverse environmental effects [60]. Moreover, it facilitates the transition of soil N into a crop-appropriate form, thereby increasing N efficiency [61]. The variations

in STN contents are probable due to straw incorporation combined with different N fertilization rates [28].

Straw incorporation replenishes soil nutrients and enhances the physio-chemical properties of the plow layer, benefiting sustainable crop growth, biomass, and grain yield [31, 39]. During 2020 and 2021, this practice significantly increased SOC, STN, and SMBC in double-cropping systems (Figs. 1 and 2, and 3). These fertility gains likely results from the enhanced decomposition of maize straw, driven by soil microbial activity [49, 62], which releases nutrients such as dissolved organic matter and stimulates microbial development [42, 59]. Similarly, N fertilization significantly elevated SMBC and improved soil carbon sequestration and microbial activity [28, 63]. Notably, the N200 treatment in straw incorporated plots considerably enhanced STN and SMBC compared to N250 and N300 treatments in traditional planting systems during both study years (Figs. 2 and 3). Benbi et al. [64] reported similar increases in soil nutrients and SMBC with straw incorporation due to greater decomposition and soil moisture content. The SOC levels were also higher under the N200 treatment with straw incorporation

than under the N250 treatment in traditional planting (Fig. 1). These results aligns with previous research indicating that straw incorporation combined with optimal N fertilizer application significantly improves soil fertility and SMBC by facilitating nutrient release from residues decomposition and maintaining sustainable C/N ratio [65]. In addition, the effects of environmental variables, such as precipitation patterns, should be considered. In this study, spring rainfall was significantly higher than autumn rainfall, potentially increasing maize straw decomposition and, consequently, soil fertility [28].

Soil microbial biomass and enzyme activities are more sensitive to changes in soil quality than SOC and STN content [66]. Straw incorporation significantly enhanced the physio-chemical characteristics of soil, providing a more suitable environment for soil microbial biomass and enzyme activities [67, 68]. In 2021, straw incorporation in the N300 treatment significantly increased the activities of soil enzymes such as urease, cellulase, sucrase, catalase, and soil acid phosphatase compared to traditional planting. These increases were statistically similar to those in the N250 and N200 treatments. However, in 2020, the N300 treatment showed significant increases in the activities of cellulase, sucrase, catalase, and soil acid phosphatase, surpassing the effects observed in the N250 treatment (Table 1). The increase in soil enzyme activities can be attributed to straw incorporation, which augments SMBC, N, and microbial population. This, in turn, provided organic matter that serves as a substrate for soil enzymes [24]. Furthermore, straw decomposition not only provides energy but also fosters a favorable environment for soil enzyme activity [24]. There is a positive correlation between soil enzyme activities and both soil organic matter and microbial population [69]. Similarly, activities of soil urease, cellulase, sucrase, and acidic phosphatase are strongly and positively correlated with soil organic matter [69].

Soil urease, an amide enzyme that uses urea as a substrate, is mainly affected by soil nutrients and microorganisms. It plays a crucial role in soil N cycling and indicates soil N supply capacity. Additionally, cellulase and sucrase activities are pivotal in regulating SOC decomposition [70]. Straw incorporation boosts the turnover of easily oxidized SOC and STN, enhancing their availability to soil microorganisms and thereby increasing soil enzyme activities [4]. Previous studies have shown that phosphatase activity significantly increased with straw incorporation, likely due to increased substrate availability [24, 36]. Our results also showed that soil acidic phosphatase activity was significantly higher in straw incorporated plots compared to traditional planting (Table 1). Notably, the highest activity was observed in 2021 for the N300 treatment, which was not significantly different from N250 treatment.

According to R Murugan, et al. [65], N fertilization improves soil enzyme activity by preferentially allocating N to enzyme production, under the influence of soil microorganisms. Additionally, N can stabilize the soil colloidal structure and prevent soil urease degradation. A previous study demonstrated that chemical and organic fertilizer increased soil N levels, subsequently boosting the activity of carbon cycling enzymes in the soil [71]. Similarly to Y Liu, et al. [19], our results also showed that the activities of soil urease, cellulase, sucrase, catalase, and acidic phosphatase enhanced by N fertilization and straw incorporation (Table 1). This increase in enzyme activities may result from higher carbon demand for microbial activity and development in high N conditions. Previous research has shown that straw decomposition accelerates initially after incorporation and increases with increasing temperature and perception [6, 28]. It is also influenced by the quality (C/N ratio) and quantity of biomass [31].

Straw incorporation and N fertilization have a varying effects on soil enzymes at different growth stages and across seasons. Our results showed that activities of urease, sucrase, catalase, and acidic phosphatase were significantly higher at V6 growth stage than at the R6 stage in both seasons. However, with the exception of cellulase, all soil enzyme activities were significantly higher during the spring than in the autumn (Table 2). The enhanced enzymatic activities in spring may result from more rapid straw decomposition at an appropriate temperature, which produce a huge amount of SOC and N for soil microbes as a substrate [28, 63]. Conversely, cellulase activity was significantly higher in than in spring (Table 2), potentially due to greater abundance of cellulose-decomposing fungi during the autumn [72].

Straw incorporation established a favourable chemical, physical, and biological soil environment that enhances enzymatic activities, increasing soil fertility and crop yield [73]. Similarly, our results also demonstrated that straw incorporation and N fertilization significantly increased maize grain yield in a dual cropping system (Table 3). Notably, the highest grain yield was observed in spring season compared to autumn season. This increase in straw incorporated plots could be due to improved SOC, along with enhanced biological and physical properties of soil [74]. A Khan, et al. [39] reported that N fertilization in combination with organic manure significantly increased soil fertility, N use efficiency, SOC, and crop yield. Additionally, straw incorporation has been shown to substantially increase soil nutrients, microbial biomass and activity as well as improve soil texture, resulting in higher nutrient uptake and maize grain yield [16, 28, 72].

The N300 treatment resulted in higher grain yield compared to N0 treatment, although it was not significantly

different from N250 treatment across both seasons and straw management practices in 2020. Similar results were observed in autumn season during 2021. However, in spring, the N300 significantly increased grain yield compared to other N treatments. S Li, et al. [75] reported that excessive N fertilization could increased plant nutrient uptake and affected crop reproductive growth, ultimately decreased the economic value of maize yield. Incorporating straw incorporation with the N200 treatment significantly increased crop yield, soil fertility, and SMBC compared to N250 and N300 treatments in traditional planting systems over both years. These results are in line with the previously published studies, and they demonstrated that N fertilization with straw incorporation resulted in higher grain yield [2, 28]. Similarly, A Khan, et al. [39] reported that the combine application of organic and inorganic N fertilization significantly increased crop yield due to the slow release of N improved soil fertility and soil microbial biomass (carbon and N) on a sustainable basis and thus increased both biological and grain yield [38].

Conclusions

Our study investigated the impact of straw incorporation and N fertilization rates on SOC, STN, SMBC, enzyme activities, and maize yield. In 2021, N fertilization particularly enhanced physio-chemical properties, enzyme activities, and grain yield in straw incorporated plots. Both straw incorporation and N fertilization significantly improved SOC, STN, SMBC, and soil enzyme activities at the V6 growth stage across all seasons and years compared to R6. The N200 treatment with straw incorporation has a significant higher maize grain yield than the N250 and N300 treatments of traditional planting. In spring season, across both years, increases in SOC, STN, SMBC, and activities of urease, sucrose, catalase, and acid phosphatase activities were notably higher, whereas soil cellulase activity peaked in autumn season during 2020. All the other parameters showed significant enhancements in 2021. We concluded that optimum N fertilization (N200) with straw incorporation is the most effective strategy for improved soil fertility, SMBC, and crop yield. Furthermore, this approach helps mitigate the environmental impacts of straw burning, reduced fertilizer cost, and increased agricultural sustainability.

Supplementary Information

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Supplementary Material 1

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Author contributions

Author contributions: Conceptualization: LY. Methodology: IM. Formal analysis: LY and ZYL. Investigation: YXC and TYC. Resources: XBZ. Data curation: LY and YXC. Writing-original draft preparation: LY. Writing-review and editing: IM. Supervision: XBZ.

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Data availability

Data is provided within the manuscript or supplementary information files.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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