

Article

Black Soil Quality Under Contrasting Straw Management–Tillage Systems: Associations with Aggregate Stability and Labile Carbon Dynamics

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Abstract

The degradation of soil structure and organic carbon in the black soil region of Northeast China threatens sustainable crop production, and straw return combined with appropriate tillage may offer an effective strategy for soil restoration. However, the depth-dependent mechanisms by which straw management–tillage systems regulate aggregate stability, labile carbon distribution, and soil quality under long-term straw return remain unclear. Based on a seven-year field experiment established in 2018 in eastern Inner Mongolia, this study compared conventional tillage with straw removal (CT) and four straw-return practices: deep tillage with straw return (DTS), deep harrowing with straw return (DHS), rotary tillage with straw return (RTS), and no-tillage with straw mulching and direct seeding (NTS). Soil aggregate stability, soil organic carbon (SOC), SOC stock, labile organic carbon fractions, microbial biomass carbon, carbon-acquiring enzyme activity, available nutrients, and soil quality index (SQI) were evaluated across the 0–60 cm soil profile. Straw return combined with tillage improved soil structure, carbon accumulation, nutrient availability, and SQI, with stronger responses observed under NTS and DTS. NTS mainly increased macroaggregate formation and the accumulation of SOC, light fraction organic carbon (LFOC), and particulate organic carbon (POC) in the 0–20 cm layer and was associated with the highest SOC stock in the whole profile. In contrast, DTS showed greater increases in SOC stock, dissolved organic carbon (DOC), readily oxidizable organic carbon (ROC), available nutrients, and SQI in the 20–40 cm layer, likely related to deeper straw incorporation. Partial least squares (PLS) path modeling indicated that labile carbon pools were closely associated with the relationship between aggregate stability and soil quality improvement. These results suggest that NTS appears promising for surface soil conservation, whereas DTS may be beneficial for improving subsoil fertility and carbon sequestration in black soil farmland.

Keywords: straw management–tillage systems; aggregate stability; labile organic carbon; soil quality; carbon sequestration



Academic Editor: Pardon Muchaonyerwa

Received: 26 May 2026

Revised: 23 June 2026

Accepted: 26 June 2026

Published: 28 June 2026

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1. Introduction

Soil quality reflects the capacity of soil to sustain crop productivity, regulate nutrient cycling, and maintain the long-term sustainability of agroecosystems [1,2]. It is determined by the combined effects of physical, chemical, and biological properties, which regulate soil structure, nutrient supply, water retention, and biological processes. Accordingly, soil organic matter, pH, nutrient availability, bulk density, aggregate stability, microbial biomass, and enzyme activities are commonly used to evaluate changes in soil fertility and ecosystem functioning under different land-use and management practices [3,4]. Black soil is characterized by high natural fertility and organic carbon stocks, and the black soil region of Northeast China is one of the most important grain-producing areas in China. However, long-term intensive cultivation, particularly conventional tillage combined with straw removal, has caused pronounced declines in soil organic matter, aggregate stability, and nutrient-retention capacity [5]. These changes can accelerate aggregate breakdown and organic carbon mineralization, thereby weakening soil fertility, cropland quality, and carbon sequestration capacity in the black soil region [6].

Straw return, as an important pathway for external organic carbon input, can increase soil organic matter inputs and improve soil physicochemical properties [7]. However, the transformation of straw-derived carbon into soil organic carbon (SOC) and stable carbon pools depends on soil disturbance intensity, straw distribution within the cultivated layer, and microbial utilization. This process involves the enzyme-mediated decomposition of cellulose, hemicellulose, and lignin by fungi, actinomycetes, and other decomposer bacteria [8,9]. Soil aggregates are fundamental structural units that regulate the physical protection of organic carbon, nutrient retention, and soil ecological functions [10,11]. Stable macroaggregates can improve pore structure and water-holding capacity and reduce contact among organic carbon, microorganisms, and extracellular enzymes through physical protection, thereby influencing SOC turnover and stabilization [12,13]. No-tillage can reduce mechanical disturbance and maintain surface straw mulch, which is conducive to the formation of surface macroaggregates and SOC accumulation [14]. In contrast, deep plowing may break the plow pan, promote straw incorporation into the subsurface layer, and potentially improve soil structure and nutrient distribution in the 20–40 cm soil layer [15]. Therefore, different straw-return practices may regulate structural stability and SOC stocks in black soil by altering the distribution of straw within the cultivated layer and its pathways of influence on soil structure.

However, total SOC mainly reflects the dynamic balance between carbon inputs and decomposition, whereas SOC content alone is insufficient to characterize carbon turnover activity and changes in carbon pool quality [16]. Labile carbon fractions, such as dissolved organic carbon (DOC), readily oxidizable organic carbon (ROC), light fraction organic carbon (LFOC), particulate organic carbon (POC), and microbial biomass carbon (MBC), are sensitive to management practices and serve as important indicators for identifying changes in soil carbon dynamics and evaluating changes in soil quality [17]. These labile organic carbon fractions not only provide substrates for microbial metabolism but also play important roles in regulating extracellular enzyme activity and nutrient mineralization processes [18,19]. Previous studies have shown that straw return may increase organic carbon inputs and promote soil organic carbon (SOC) accumulation in croplands [20,21]. After entering the soil, straw-derived carbon can be mineralized as CO₂, retained in labile carbon fractions such as particulate organic carbon, or stabilized through microbial processing, organo-mineral association, and aggregate protection [22,23]. Thus, the effect of straw return on SOC accumulation depends not only on carbon input but also on how straw-derived carbon is transformed and stabilized [24].

Although previous studies have shown that straw return may improve soil structure, increase soil organic carbon stock, and promote the accumulation of labile organic carbon fractions, it remains unclear whether different straw-return practices differentially regulate aggregate stability, labile carbon transformation, and SOC accumulation by altering the distribution of straw within the cultivated layer of black soil farmland. Therefore, based on a straw-return field experiment in the black soil region of eastern Inner Mongolia, this study investigated the effects of different straw-return practices on aggregate stability, SOC accumulation, and the distribution of labile organic carbon fractions in black soil. By clarifying these relationships, this study aims to provide a practical basis for selecting straw-return practices that improve soil structure, enhance SOC sequestration potential, and sustain maize productivity and cropland quality in black soil regions.

2. Materials and Methods

2.1. Study Areas

The experiment was initiated in 2018 as a field trial in Najitun Town, Arun Banner, Hulunbuir City, located on the eastern foot of the Greater Khingan Mountains (48°14' N, 123°47' E). The region has a temperate continental semi-humid climate, and the soil type is typical black soil. The mean annual temperature is 1.7 °C. The average annual precipitation is 458.4 mm. Before the experiment, the basic physicochemical properties of the 0–60 cm soil layer, including soil bulk density (SBD), soil organic carbon (SOC), total nitrogen (TN), available phosphorus (AP), available potassium (AK), and soil pH, were determined. The detailed values are provided in Table S1.

2.2. Experimental Design and Management

The field experiment adopted a single-factor randomized complete block design and included four straw-return treatments. (1) Deep tillage with straw return (DTS): After maize harvest, all straw was chopped and returned to the field, followed by deep plowing to a depth of 30–35 cm; maize was sown after deep harrowing in spring. (2) Deep harrowing with straw return (DHS): After maize harvest, all straw was chopped and returned to the field, followed by deep harrowing to a depth of 10–15 cm; maize was sown after deep harrowing in spring. (3) Rotary tillage with straw return (RTS): After maize harvest, all straw was chopped and returned to the field, followed by rotary tillage to a depth of 10–15 cm; maize was sown after deep harrowing in spring. (4) No-tillage with straw mulching and direct seeding (NTS): After maize harvest, all straw was chopped and uniformly mulched on the soil surface; in spring, maize sowing, fertilization, and soil compaction were performed directly using a no-tillage planter. Conventional tillage with straw removal (CT) was used as the control. In this treatment, all straw was removed after maize harvest, followed by shallow plowing to a depth of 15–20 cm; maize was sown after deep harrowing in spring.

During the experimental period, the straw-return method for each treatment remained unchanged to ensure consistency across treatments and facilitate comparative analysis. Each straw-return treatment was replicated three times, with each experimental plot measuring 30 m × 30 m, resulting in a total of 15 experimental plots. Maize was used as the experimental crop, with the cultivar Fengyu 8 planted at a uniform density of 75,000 plants ha^{−1}. The experimental site was under rainfed farming conditions, and no irrigation was applied throughout the maize growing season. The fertilizers consisted of an NPK compound fertilizer (N–P₂O₅–K₂O: 19–19–19) and urea (N ≥ 46%). The former was applied at sowing as starter fertilizer at 450 kg ha^{−1}, whereas the latter was top-dressed at the jointing stage at 225 kg ha^{−1}.

From 2018 to 2024, all aboveground straw was returned to the field after autumn harvest each year using the different straw-return practices. To quantify the amount of straw returned under each treatment, after physiological maturity of the previous crop, aboveground plants were sampled from a 5 m × 2 row area for each straw-return practice, with three replicates. The samples were oven-dried to constant weight and weighed to determine the straw-return amount. The straw-return amounts during the experimental period are shown in Table S2.

2.3. Soil Sampling and Analysis

Soil samples were collected after maize harvest in October of 2018, 2021, and 2024. In each plot, composite soil samples were collected from the 0–20 cm, 20–40 cm, and 40–60 cm soil layers using the five-point sampling method. Plant residues and stones were removed, and the samples were passed through a 2 mm sieve. Fresh soil samples were divided into two subsamples: one was stored at 4 °C for the determination of microbial biomass carbon (MBC) and carbon-acquiring enzyme activity (C_{acq}), while the other was air-dried for the determination of soil organic carbon (SOC), available nitrogen (AN), available phosphorus (AP), available potassium (AK), labile organic carbon fractions, including light fraction organic carbon (LFOC), readily oxidizable organic carbon (ROC), particulate organic carbon (POC), and dissolved organic carbon (DOC).

SOC was determined using the combustion analysis method [25]. LFOC was measured after separation by the sodium iodide dispersion method [26]. ROC was determined using the potassium permanganate oxidation method [27]. POC was measured after separation by sodium hexametaphosphate dispersion [2]. DOC was determined according to the method of Jones and Willett (2006) [28]. MBC was measured using the chloroform fumigation–extraction method [29]. AN was determined by the alkaline hydrolysis diffusion method, AK was measured using flame photometry (Inesa, Shanghai, China) [30], and AP was determined by the molybdenum–antimony colorimetric method (Shimadzu, Tokyo, Japan) [31]. All indicators were calculated based on air-dried soil weight, while indicators measured using fresh soil were converted according to soil moisture content.

2.4. Soil Organic Carbon Stock and Carbon Sequestration Rate

Soil organic carbon stock (SOC_{stock}) was calculated based on soil organic carbon content and soil bulk density (SBD) in three soil layers within the 0–60 cm profile. The soil bulk density data are provided in Table S3.

$$SOC_{stock} \left(\text{Mg} \cdot \text{ha}^{-1} \right) = SOC_i \times h \times SBD \times 104 \cdot \text{m}^{-2} \cdot \text{hm}^{-2} \times 10$$

where SOC_{stock} is the soil organic carbon stock, SOC_i is the soil organic carbon content of the soil layer, h is the soil layer thickness, and SBD is the soil bulk density.

$$\Delta SOC_{stock} \left(\text{Mg} \cdot \text{ha}^{-1} \right) = SOC_{initial} - SOC_{finish}$$

$$RSOC_{stock} \left(\text{Mg} \cdot \text{ha}^{-1} \cdot \text{y}^{-1} \right) = \frac{SOC_{initial} - SOC_{finish}}{Y}$$

where ΔSOC_{stock} is the amount of soil organic carbon sequestered, $RSOC_{stock}$ is the soil organic carbon sequestration rate, $SOC_{initial}$ is the soil organic carbon stock at the beginning of the experiment, SOC_{finish} is the soil organic carbon stock at the end of the experiment, and Y is the experimental duration.

2.5. Soil Aggregate Determination

After six years of field experimentation, intact soil samples were obtained from the 0–20, 20–40, and 40–60 cm depths following maize harvest in October 2023 and 2024. After air-drying, visible roots, plant fragments, and stones were manually removed. Soil aggregate stability was determined using the wet-sieving method [32]. A 100 g subsample of air-dried soil was transferred to a nest of sieves with apertures of 2, 1, 0.5, 0.25, and 0.053 mm. The samples were presoaked for 10 min and then vertically oscillated for another 10 min. Aggregates retained on each sieve were collected, dried at low temperature, and weighed. The mean weight diameter (MWD) and macroaggregate percentage ($R > 0.25$ mm) were then calculated using the following equations:

$$MWD \text{ (mm)} = \sum_{i=1}^n (X_i W_i)$$

where MWD denotes the mean weight diameter (mm), X_i represents the mean diameter of the aggregate size class of variable i , and W_i is its corresponding mass fraction.

$$R > 0.25 \text{ (mm)} = \frac{M_{r>0.25}}{M_t} \times 100\%$$

where $R > 0.25$ mm represents the macroaggregate proportion, $M_{r > 0.25}$ is the mass of aggregates retained in the >0.25 mm fractions, and M_t is the total aggregate mass.

2.6. Soil Enzyme Measurement

Fresh soil samples sieved after maize harvest in October 2023 and 2024 were used to determine carbon-acquiring enzyme activity (C_{acq}). The activities of soil β -glucosidase (BG), β -xylosidase (BX), and cellobiohydrolase (CBH) were measured using the microplate fluorometric method [33,34]. C_{acq} represents the mean activity of the three carbon-acquiring enzymes and was used to characterize the potential capacity of microorganisms to acquire carbon substrates. The activities of individual enzymes, including BG, BX, and CBH, are shown in Figures S1–S3. C_{acq} was calculated using the following equation:

$$C_{acq} \text{ (nmol g}^{-1} \text{ h}^{-1}) = \frac{BG + BX + CBH}{3}$$

2.7. Soil Quality Index (SQI) Evaluation

Mean weight diameter (MWD), available nitrogen (AN), available phosphorus (AP), available potassium (AK), soil organic carbon (SOC), microbial biomass carbon (MBC), and carbon-acquiring enzyme activity (C_{acq}) were selected as candidate indicators for soil quality assessment. Each indicator was transformed into a dimensionless value ranging from 0 to 1 using nonlinear scoring functions. Correlation analysis (Pearson correlation coefficient) and principal component analysis (PCA) were then used to determine the minimum data set (MDS) of soil property indicators. After scoring and assigning weights to the MDS indicators, the SQI was calculated.

The soil properties were obtained from each experimental plot. These properties were analyzed with PCA and correlation analysis to identify MDS. Principal components (PC) with eigenvalues >1 were retained. Within each retained PC, only indicators whose absolute loading values were within 10% of the maximum loading were included in the MDS. When multiple indicators were retained in a PC, highly correlated indicators ($r > 0.60$) with lower weighted loadings were excluded [35].

Each MDS indicator was transformed to a dimensionless score ranging from 0 to 1 using a direction-specific, piecewise linear scoring function. Within the agronomically relevant range observed in this study, all retained indicators were treated as “more is better,”

as higher values indicated improved soil aggregation, nutrient availability, organic carbon status, or microbial activity [36]. The score of indicator i was calculated as follows:

$$S = \begin{cases} 0 & x \leq b \\ \frac{x-b}{a-b} & b < x < a \\ 1 & x \geq a \end{cases}$$

where S is the soil property score, x is the measured value of the soil property, and a and b represent the maximum and minimum measured values, respectively.

After scoring and assigning weights to the MDS indicators, the soil quality index was calculated [37]. The weight values of soil quality indicators were calculated using the following equation:

$$Wi = \frac{Fi}{\sum_{i=1}^n Fi}$$

where n is the number of soil property indicators, Wi is the weighting coefficient of the soil property indicator of variable i , and Fi represents the communality of the soil property indicator of variable i .

$$SQI = \sum_{i=0}^n Wi \times Si$$

where Wi is the weight value of the indicator of variable i , and Si represents the soil property score of variable i .

2.8. Statistical Analysis

Statistical analyses were performed using SPSS 25.0 software (IBM Inc., Armonk, NY, USA). Linear mixed-effects models were used to evaluate the fixed effects of treatment, year, soil depth, and their interactions on soil properties, with block and plot included as random effects to account for repeated observations. Significant effects were further examined using Tukey-adjusted estimated marginal means. Statistical significance was set at $p < 0.05$. Principal component analysis (PCA) was conducted using Origin 2024 (OriginLab, Northampton, MA, USA). Partial least squares path modeling (PLS-PM) was performed using SmartPLS 4.0 to evaluate the relationships among aggregate stability, carbon fractions, nutrient availability, and soil quality. The goodness-of-fit (GoF) index was used to assess the overall model performance.

3. Results

3.1. Effects of Straw Management–Tillage Systems on Soil Aggregate Stability

As shown in Figure 1, the mean weight diameter (MWD) of soil aggregates differed significantly among DTS, DHS, RTS, and NTS across the 0–60 cm soil profile. Compared with CT, all straw-return treatments significantly increased MWD. On average, MWD in the 0–60 cm soil layer under the straw-return treatments was 27.8% higher than that under CT. Among them, NTS showed the greatest increase, with a two-year average increase of 42.6% compared with CT, followed by DTS, which increased by 30.4%. DHS and RTS did not differ significantly. The effects of different straw-return practices on MWD were mainly concentrated in the 0–40 cm cultivated layer. Specifically, NTS had the highest MWD in both the 0–20 cm and 20–40 cm soil layers, increasing by 41.0% and 44.2% compared with CT, respectively. However, in the 20–40 cm subsoil layer, MWD under DTS did not differ significantly from that under NTS; it was 35.8% higher than that under CT and 20.4% and 10.5% higher than those under DHS and RTS, respectively.

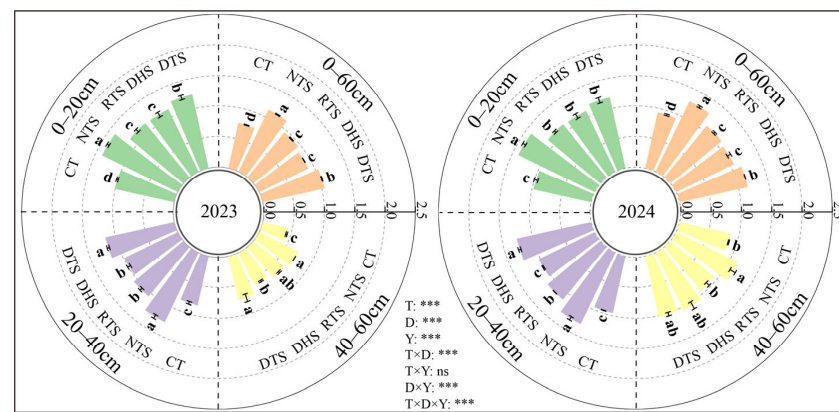


Figure 1. Changes in the mean weight diameter (MWD) of soil aggregates. Data are expressed as the mean $\pm$ SE ($n = 3$). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal. T, D, and Y denote treatment, soil depth, and year, respectively; $\times$ represent their interactions. The symbols in the figure indicate statistical significance: *** represents $p < 0.001$; ns denotes not significant.

Combined straw-return and tillage systems significantly altered soil aggregate size distribution by promoting macroaggregate formation ($R > 0.25$ mm) and reducing the proportion of microaggregates ($R < 0.25$ mm) (Figure 2). Compared with CT, straw-return treatments increased the proportion of $R > 0.25$ mm aggregates in the 0–60 cm soil profile by an average of 32.5%. NTS showed the highest macroaggregate proportion, averaging 69.9% over two years, which was 42.2% higher than CT. NTS had the highest proportion of macroaggregates in the 0–20 cm layer, exceeding those under DHS and RTS by 11.2% and 9.7%, respectively. In the 20–40 cm layer, DTS significantly increased the macroaggregate proportion compared with DHS, RTS, and CT, but did not differ significantly from NTS.

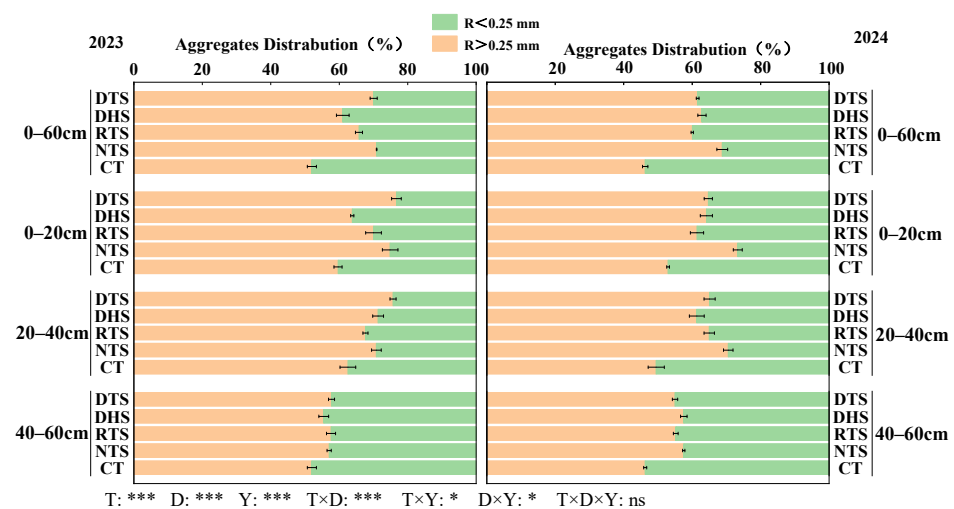


Figure 2. Proportions of soil macroaggregates ($R > 0.25$ mm) and microaggregates ($R < 0.25$ mm). Data are expressed as the mean $\pm$ SE ($n = 3$). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. The solid lines represent the error among the aggregate proportions. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal. T, D, and Y denote treatment, soil depth, and year, respectively; $\times$ represent their interactions. The symbols in the figure indicate statistical significance: *** represents $p < 0.001$; * represents $p < 0.05$; ns denotes not significant.

3.2. Effects of Straw Management–Tillage Systems on Soil Organic Carbon and Labile Carbon Fractions

Combined straw-return and tillage systems significantly affected soil organic carbon (SOC) content in the 0–40 cm soil layer, showing a gradual accumulation trend with increasing years of straw return (Figure 3). At the initial stage of the experiment in 2018, SOC did not differ significantly among straw-return treatments in the 0–20 cm soil layer. By 2021, SOC contents under the straw-return treatments (DTS, DHS, RTS, and NTS) were significantly higher than those under CT, with average increases of 22.0% and 34.6% in the 0–20 cm and 20–40 cm soil layers, respectively. Among them, DTS and NTS showed marked increases in SOC, rising by 50.1% and 42.6%, respectively, compared with 2018. By 2024, SOC contents under DTS and NTS were significantly increased by 38.2% and 35.7%, respectively, compared with CT. In the 20–40 cm soil layer, SOC under DTS and NTS was 39.9% and 30.7% higher than that under CT, respectively.

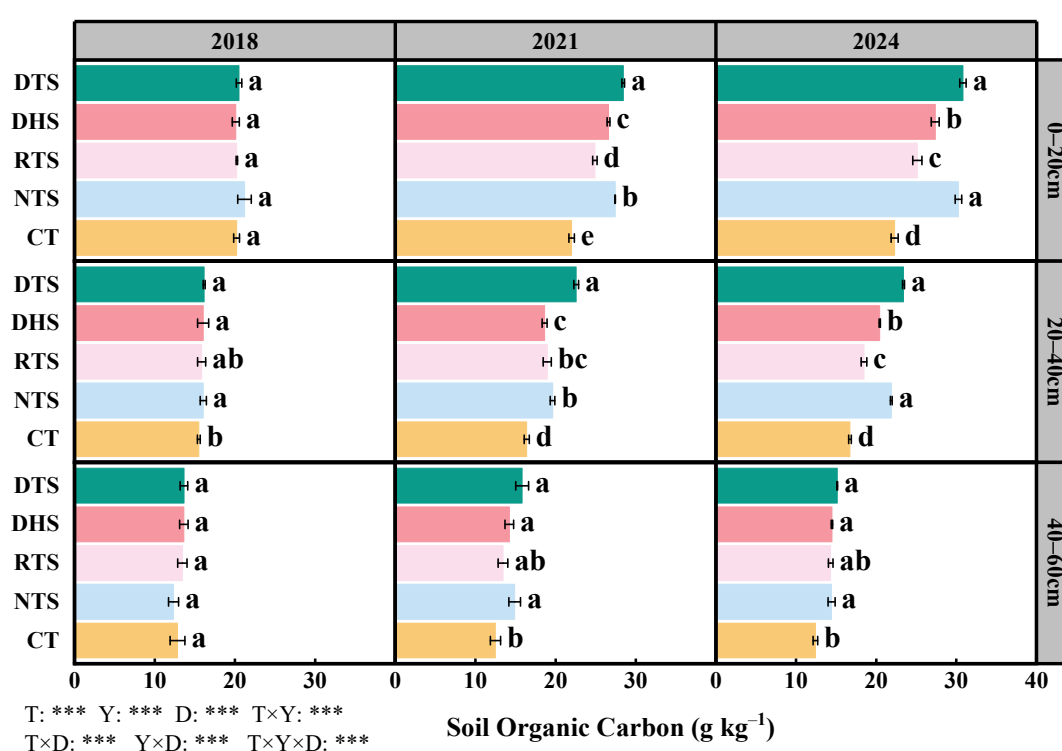


Figure 3. Changes in soil organic carbon content. Data are expressed as the mean $\pm$ SE ($n = 3$). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal. T, D, and Y denote treatment, soil depth, and year, respectively; $\times$ represent their interactions. The symbols in the figure indicate statistical significance: *** represents $p < 0.001$.

As shown in Figure 4, all combined straw-return and tillage systems significantly increased SOC_{stock} and net carbon sequestration in the 0–60 cm soil profile. By 2024, NTS had the highest SOC_{stock} in the 0–60 cm layer, reaching 178.9 Mg ha^{-1} , 32.6% higher than CT and significantly higher than DTS, DHS, and RTS. The SOC_{stock} of DTS was 169.2 Mg ha^{-1} , an increase of 25.4% compared to CT. In the 0–20 cm layer, NTS showed the highest SOC_{stock} , reaching 76.8 Mg ha^{-1} . In the 20–40 cm layer, SOC_{stock} under DTS was 58.9 Mg ha^{-1} , showing no significant difference from NTS but increasing by 28.7% compared with CT. In contrast, SOC_{stock} under CT declined with time, decreasing from 39.9 Mg ha^{-1} in 2018 to 34.3 Mg ha^{-1} in 2024. From 2018 to 2024, NTS achieved

a net SOC_{stock} increase of 42.6 Mg ha⁻¹, with an average annual sequestration rate of 6.1 Mg ha⁻¹ y⁻¹, while DTS showed a comparable rate of 5.7 Mg ha⁻¹ y⁻¹, with no significant difference from NTS (Figure 5).

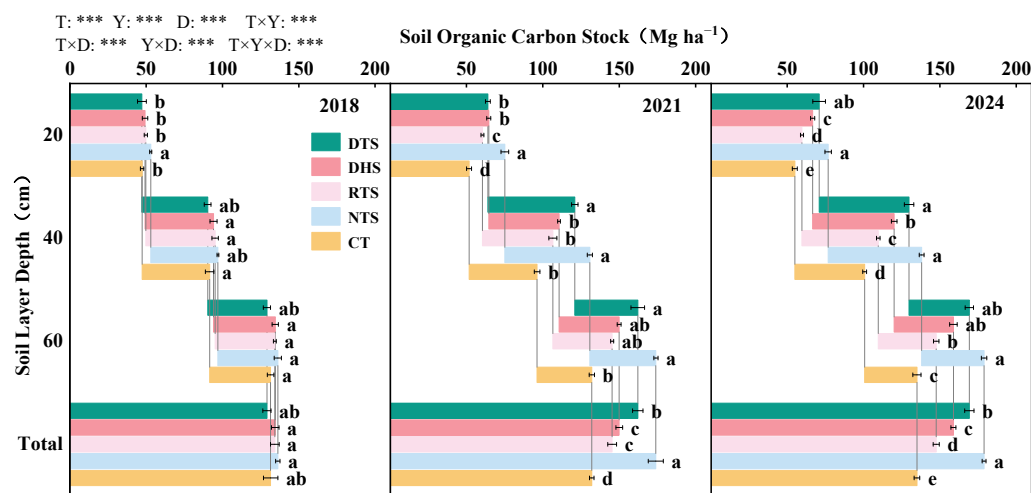


Figure 4. Changes in soil organic carbon stock. Data are expressed as the mean $\pm$ SE (n = 3). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal. T, D, and Y denote treatment, soil depth, and year, respectively; × represent their interactions. The symbols in the figure indicate statistical significance: *** represents $p < 0.001$.

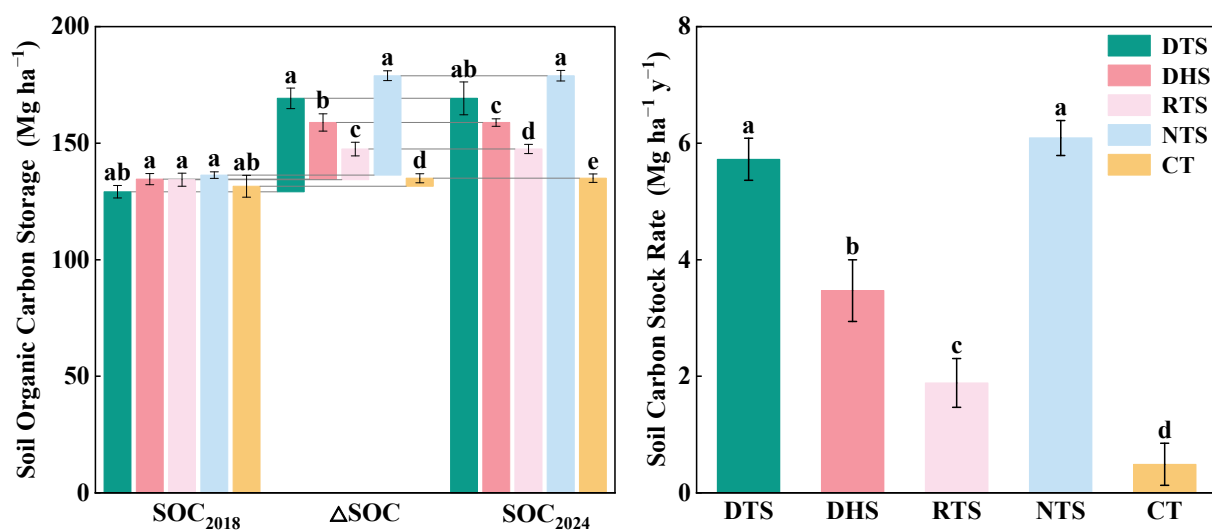


Figure 5. Changes in soil carbon sequestration amount and sequestration rate. Data are expressed as the mean $\pm$ SE (n = 3). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal.

Combined straw-return and tillage systems enhanced the accumulation of CAFs, with the strongest responses occurring in the 0–20 cm soil layer. No significant differences among treatments were observed in 2018. By 2021, NTS mainly increased CAF contents in the surface layer, whereas DTS showed greater effects in the 20–40 cm layer, especially for ROC, LFOC, and DOC (Table S4). By 2024, NTS and DTS had significantly higher CAF contents than DHS and RTS, particularly in the 0–20 cm layer. In this layer, DTS showed the

highest ROC and DOC contents, exceeding CT by 3.1 g kg^{-1} and 83.7 mg kg^{-1} , respectively, whereas NTS had the highest LFOC and POC contents, exceeding CT by 3.0 and 4.1 g kg^{-1} , respectively. In the 20–40 cm layer, POC under NTS was 46.9% higher than that under CT and significantly higher than those under DTS, DHS, and RTS.

3.3. Effects of Straw Management–Tillage Systems on Soil Microbial Biomass Carbon and Carbon-Acquiring Enzymes

Straw management–tillage systems enhanced soil microbial biomass carbon (MBC), and its positive effect generally weakened with increasing soil depth (Figure 6). In 2018, MBC showed no clear treatment-related variation across the three soil layers. After three years, the response of MBC was mainly found in the 0–40 cm layer, where straw-return treatments increased MBC by an average of 28.3% relative to CT. Among these treatments, DTS produced the greatest improvement, with MBC being 42.5% higher than that under CT. By 2024, the influence of straw return on MBC became more evident throughout the 0–60 cm profile. DTS and NTS showed comparable MBC levels, and both were higher than DHS and RTS, with increases of 60.1% and 54.7%, respectively, relative to CT. In the surface layer, DTS had the highest MBC value, reaching 409.3 mg kg^{-1} . For the 20–40 cm layers, NTS and DTS showed similar effects, with MBC increasing by approximately 54.0% compared with CT.

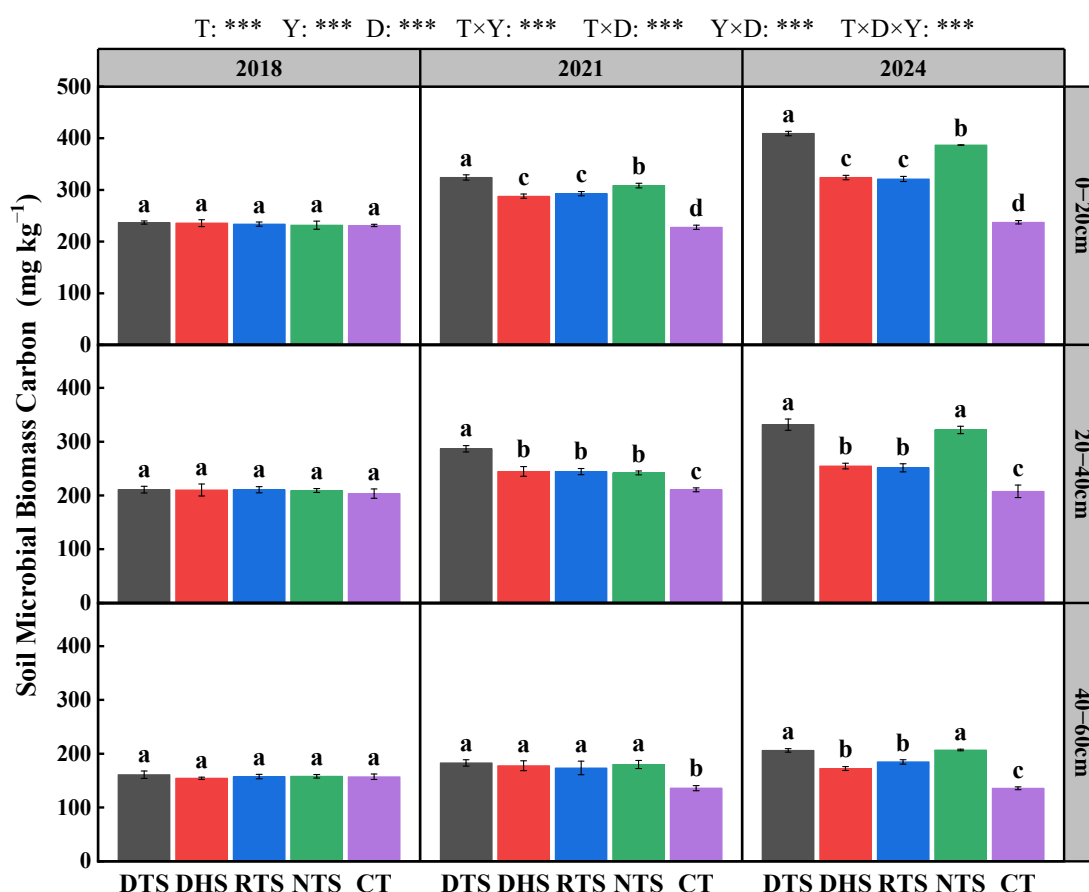


Figure 6. Changes in soil microbial biomass carbon content. Data are expressed as the mean $\pm$ SE ($n = 3$). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal. T, D, and Y denote treatment, soil depth, and year, respectively; $\times$ represent their interactions. The symbols in the figure indicate statistical significance: *** represents $p < 0.001$.

Straw management–tillage systems modified the mean activity of soil C-acquiring enzymes (C_{acq}), with an average increase of 38.2% across the 0–60 cm profile relative to CT (Figure 7). In the surface layer, DTS and NTS produced greater C_{acq} activity than DHS and RTS, showing increases of 42.7% and 38.7%, respectively, over CT. A similar pattern was observed at 20–40 cm, where C_{acq} activity under DTS and NTS was enhanced by 53.6% and 45.7%, respectively.

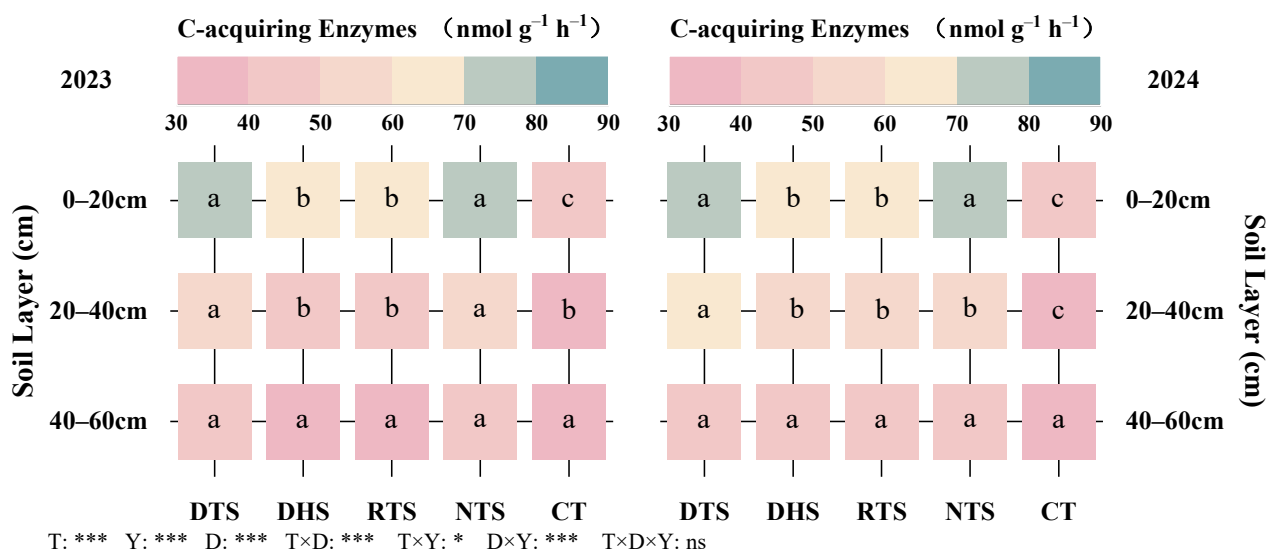


Figure 7. Changes in soil carbon-acquiring enzyme activity. Data are expressed as the mean $\pm$ SE ($n = 3$). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal. T, D, and Y denote treatment, soil depth, and year, respectively; $\times$ represent their interactions. The symbols in the figure indicate statistical significance: *** represents $p < 0.001$; * represents $p < 0.05$; ns denotes not significant.

3.4. Effects of Straw Management–Tillage Systems on Soil Available Nutrients

With increasing years of straw return, different straw-return practices significantly affected the contents of soil available nitrogen (AN), available phosphorus (AP), and available potassium (AK) in the 0–60 cm soil profile. The contents of AN, AP, and AK showed an increasing trend over time, with significantly greater increases under DTS, DHS, RTS, and NTS than under CT (Table S5). Among them, DTS and NTS produced significantly greater increases in AN, AP, and AK within the 0–40 cm cultivated layer than DHS and RTS.

At the initial stage of the experiment in 2018, AN, AP, and AK contents did not differ significantly between the straw-return treatments and CT. By 2024, AN, AP, and AK contents under DTS in the 0–40 cm soil layer reached 95.7 mg kg⁻¹, 20.8 mg kg⁻¹, and 81.7 mg kg⁻¹, respectively, representing increases of 79.0%, 32.6%, and 47.9% compared with CT. Under NTS, AN, AP, and AK contents were 95.7 mg kg⁻¹, 18.9 mg kg⁻¹, and 79.7 mg kg⁻¹, respectively, increasing by 79.0%, 20.4%, and 44.3% compared with CT. In comparison, DHS increased AN, AP, and AK by 59.0%, 21.8%, and 25.5%, respectively, while RTS increased them by 66.8%, 14.8%, and 28.7%, respectively, compared with CT.

In terms of the mean annual accumulation rate, the accumulation of AN, AP, and AK in the 0–40 cm soil layer did not differ significantly between DTS and NTS. AN increased annually by 6.13–6.26 mg kg⁻¹ yr⁻¹, which was 5.6 times higher than that under CT, while AK increased annually by 1.8–3.0 mg kg⁻¹ yr⁻¹, 3.8 times higher than that under CT. Although the annual increase rate of AP was relatively slow, it was still 33.0% higher than

that under CT. These results indicate that the interaction between straw return and different straw management–tillage systems significantly enhanced soil nutrient availability and its accumulation rate under both DTS and NTS, although the distribution of AN, AP, and AK availability varied markedly among soil layers.

3.5. Effects of Straw Management–Tillage Systems on Soil Quality in Black Soil Farmland

Straw return significantly increased the soil quality index (SQI) across the 0–60 cm soil profile (Figure 8). Compared with CT, straw-return treatments increased SQI by an average of approximately 0.2, and SQI decreased with increasing soil depth. NTS and DTS had the highest SQI in the 0–20 cm soil layer, with no significant difference between them, but both were significantly higher than DHS and RTS, increasing by approximately 0.3 compared with CT. In the 20–40 cm layer, DTS showed the highest SQI, reaching 0.6, which was significantly higher than those under DHS, RTS, and NTS and approximately 0.3 higher than CT.

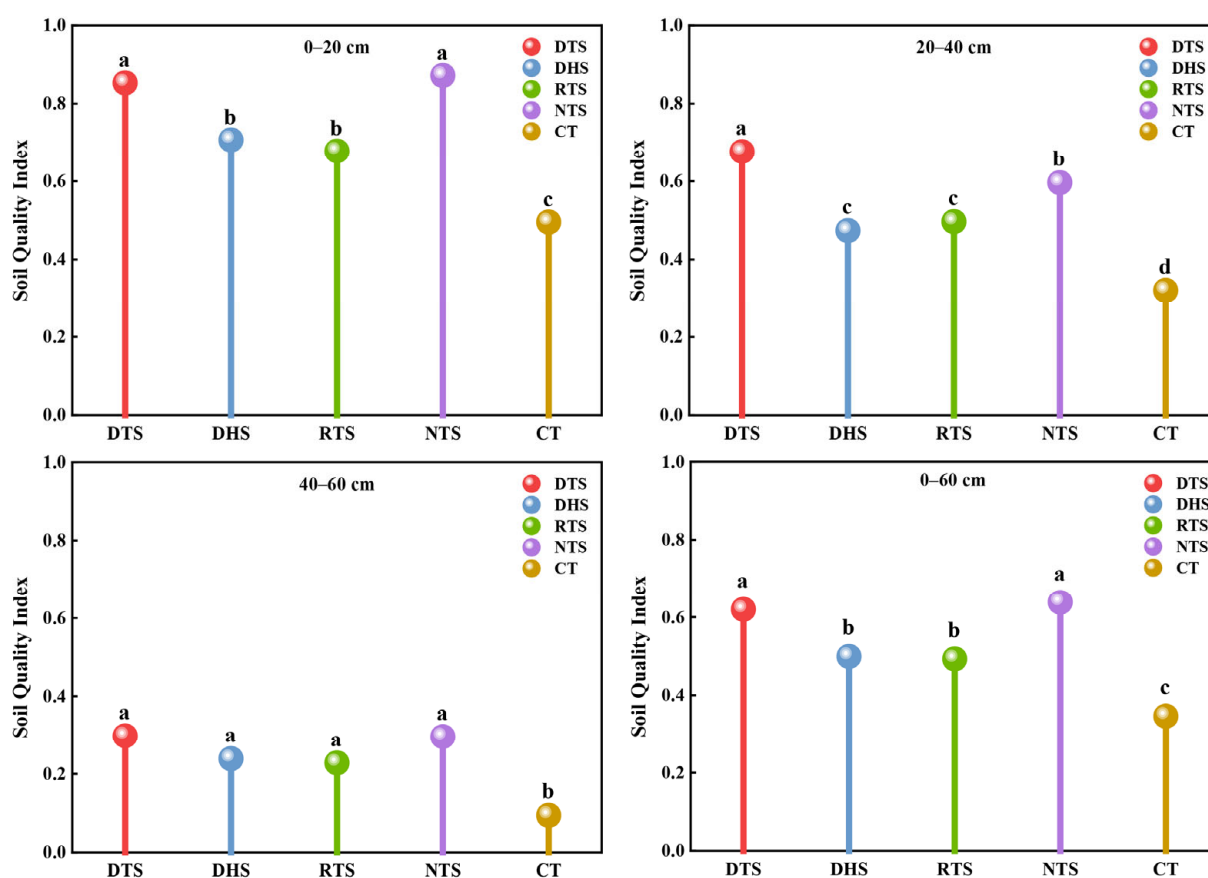


Figure 8. Changes in the soil quality index (SQI). Data are expressed as the mean $\pm$ SE ($n = 3$). Lowercase letters indicate statistical differences ($p < 0.05$) between different tillage methods in the 0–60 cm soil layer. DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal.

3.6. Synergistic Regulatory Mechanisms of Soil Structural Stability, Labile Carbon Pools, and Soil Quality Improvement

Correlation analysis showed clear synergistic changes among soil structure, labile organic carbon pools, and nutrients (Figure 9). MWD was significantly positively correlated with $R > 0.25$, as well as with SOC and labile carbon fractions, including DOC, ROC, LFOC, and POC. Meanwhile, SOC and labile carbon fractions were generally significantly positively correlated with AN, AP, and AK. MBC was significantly positively correlated

with SOC, labile carbon fractions, and available nutrients. SQI was significantly positively correlated with SOC, labile carbon fractions, AP, and C_{acq}.

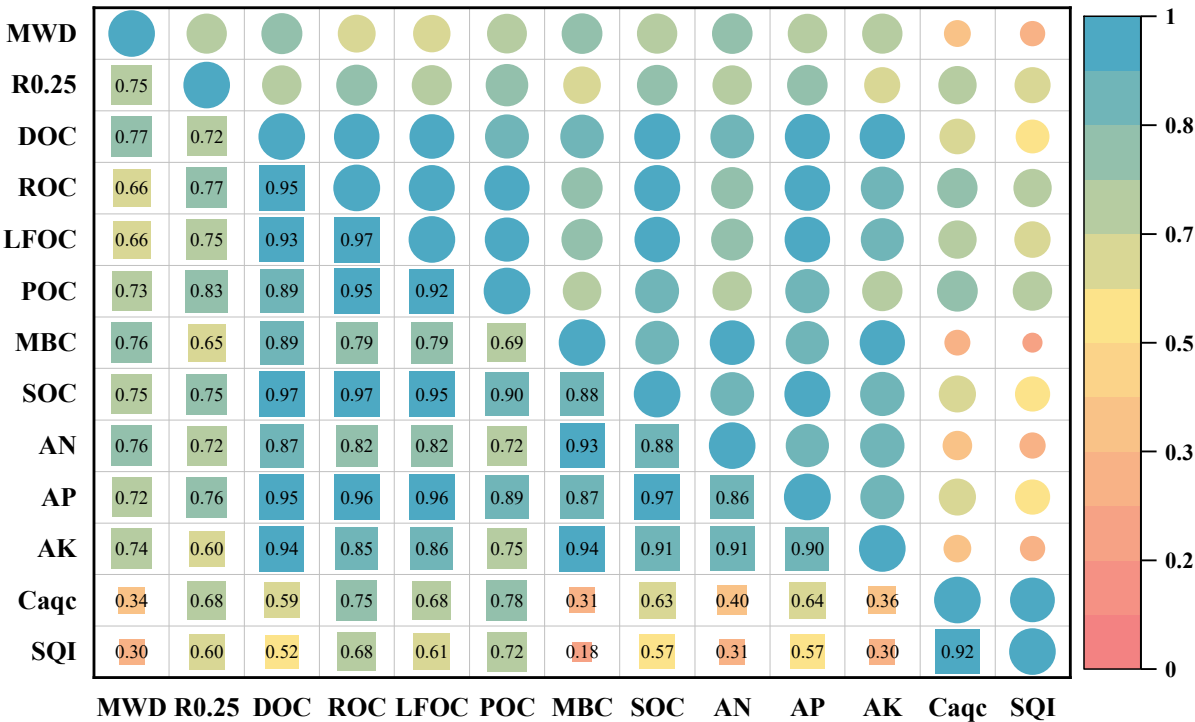


Figure 9. Correlation analysis among soil structural stability, labile carbon pools, and soil quality index. MWD: mean weight diameter; R0.25: aggregates with a particle size greater than 0.25 mm; DOC: dissolved organic carbon; ROC: readily oxidizable organic carbon; LFOC: light fraction organic carbon (LFOC); POC: particulate organic carbon (POC); MBC: microbial biomass carbon; SOC: soil organic carbon; AN: available nitrogen; AP: available phosphorus; AK: available potassium; C_{acq}: carbon-acquiring enzyme activity; SQI: soil quality index. The patterns and sizes of the squares represent the correlation coefficients: larger squares and darker patterns indicate stronger correlations. Numerical values are the correlation coefficients.

PLS-PM was used to evaluate the associations among straw-return and tillage systems, soil structural stability, labile organic carbon pools, nutrient availability, enzyme activity, and SQI (Figure 10). Straw-return and tillage systems were positively associated with soil structural stability ($\beta = 0.55$, $R^2 = 0.30$), which was strongly associated with the labile organic carbon pool ($\beta = 0.87$, $R^2 = 0.63$). The labile organic carbon pool was further associated with available nutrients ($\beta = 0.93$, $R^2 = 0.86$), enzyme activity ($\beta = 0.61$, $R^2 = 0.48$), and SQI. The model explained 77% of the variation in SQI. These results suggest that soil quality improvement was closely associated with aggregate stability, labile carbon accumulation, nutrient availability, and enzyme activity.

3.7. Effects of Straw Management–Tillage Systems on Maize Yield

In terms of annual average maize yield (Figure 11), DTS, DHS, RTS, and NTS significantly increased maize yield in the black soil region. Compared with CT, straw-return treatments increased average maize yield by 11.2%. DTS had a significantly higher annual average yield than DHS, RTS, and NTS, with an 18.5% increase relative to CT. The annual average yields of DHS, RTS, and NTS did not differ significantly from each other, but they were still significantly higher than that of CT, with increases of 11.8%, 11.1%, and 7.2%, respectively.

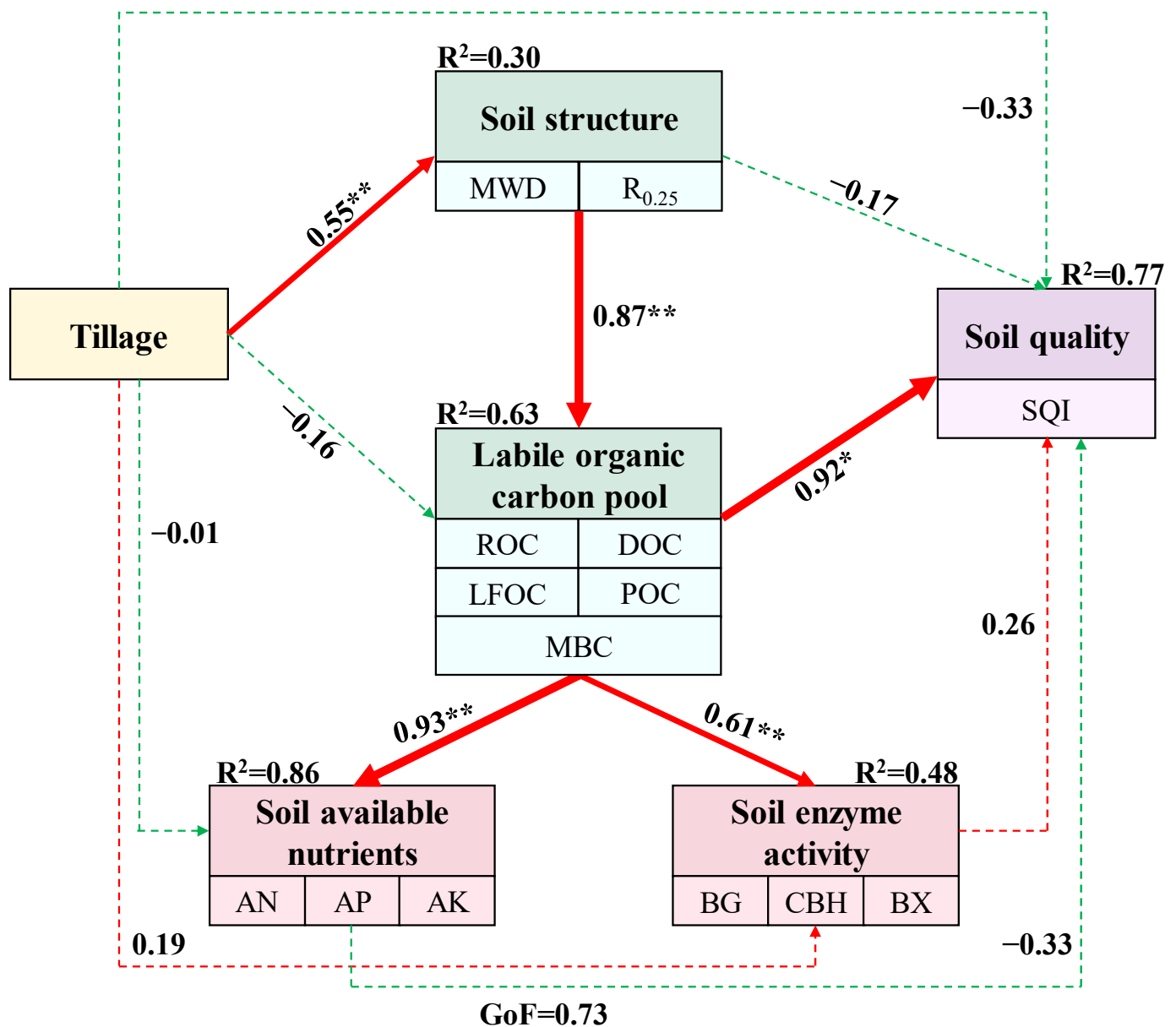


Figure 10. PLS-PM path analysis of soil structural stability, labile carbon pools, and soil quality index ($n = 30$). Soil structure was characterized by mean weight diameter (MWD) and the proportion of aggregates larger than 0.25 mm ($R_{0.25}$); the labile organic carbon pool was represented by dissolved organic carbon (DOC), readily oxidizable organic carbon (ROC), light fraction organic carbon (LFOC), particulate organic carbon (POC), and microbial biomass carbon (MBC); soil available nutrients were represented by available nitrogen (AN), available phosphorus (AP), and available potassium (AK); soil enzyme activity was represented by β -glucosidase (BG), cellobiohydrolase (CBH), and β -xylosidase (BX); and soil quality was represented by soil quality index (SQI). The values next to the arrows represent standardized path coefficients; * indicates significance: * $p < 0.05$, ** $p < 0.01$. Bootstrapping of 5000 resamples was used to check the precision of the PLS parameter estimates. GoF denotes the goodness-of-fit index, and R^2 represents the coefficient of determination for endogenous constructs. Red represents positive correlation, green represents negative correlation, solid lines indicate significant correlations, dashed lines indicate non-significant correlations, and the thickness of the line represents the magnitude of the path coefficient.

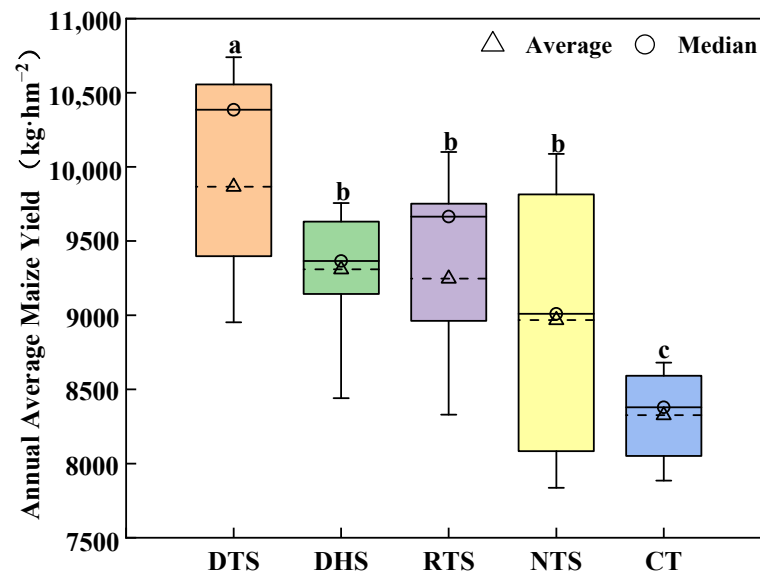


Figure 11. Changes in annual average maize yield. Lowercase letters indicate statistical differences ($p < 0.05$). DTS: deep tillage with straw return; DHS: deep harrowing with straw return; RTS: rotary tillage with straw return; NTS: no-tillage with straw mulching and direct seeding; CT: conventional tillage with straw removal.

4. Discussion

4.1. Effects of Straw Management–Tillage Systems on the Coupling Mechanism Between Soil Aggregate Structure and Organic Carbon Stability

Combined straw-return and tillage systems were associated with changes in SOC accumulation mainly through their effects on soil aggregation [38]. In this study, NTS and DTS improved aggregate stability, as reflected by higher mean weight diameter (MWD) values and a greater proportion of aggregates larger than 0.25 mm, especially within the upper 40 cm of cultivated soil. This finding is consistent with previous studies [39,40]. In contrast, intensive disturbance under conventional tillage may weaken aggregate integrity and increase the exposure of organic matter to microbial decomposition, which may partly explain the lower SOC and particulate organic carbon contents observed under CT [41,42].

No-tillage reduces mechanical disturbance and maintains crop residues on the soil surface, which may favor macroaggregate formation and surface SOC accumulation [43,44]. In the present study, NTS increased macroaggregate proportion and MWD in the 0–20 cm layer, suggesting that reduced disturbance and surface straw retention helped maintain surface soil structure. Similar results were reported by Ren et al. [45] in the black soil region of Northeast China, where no-tillage combined with straw mulching improved topsoil aggregate stability and promoted soil carbon retention. However, because aggregate-associated SOC fractions were not directly measured in this study, the role of aggregate protection in SOC stabilization should be interpreted as a potential mechanism rather than direct evidence.

In contrast, DTS may improve the 20–40 cm subsurface layer by breaking the plow pan and incorporating straw residues into deeper soil layers [46]. The results of this study showed that DTS increased aggregate stability and SOC accumulation in the 20–40 cm layer, indicating that deep tillage with straw return had a depth-specific advantage in improving subsurface soil structure. Stable aggregates may contribute to SOC retention by physically limiting the exposure of organic matter to microbial and enzymatic decomposition [47,48]. Nevertheless, the formation of aggregate-protected carbon or mineral-associated organic carbon was not directly quantified in this study. Therefore, the observed increases in SOC and aggregate stability under DTS should be interpreted as evidence of improved soil

structural condition and SOC accumulation, while the contribution of aggregate occlusion or mineral association to long-term carbon stabilization remains a hypothesis that requires further verification.

4.2. Effects of Straw Management–Tillage Systems on Labile Carbon Fractions, Microbial Biomass Carbon, and Carbon-Acquiring Enzymes

Labile organic carbon fractions are readily available carbon sources for soil microorganisms and are sensitive indicators of changes in soil carbon dynamics under different straw management–tillage systems [49,50]. In this study, continuous straw return significantly increased the contents of DOC, ROC, LFOC, and POC, with pronounced increases in the 0–20 cm surface layer, whereas changes in these fractions were relatively weak in the 40–60 cm deep soil layer. This indicates that the response of labile organic carbon pools to tillage and straw management practices exhibited a clear vertical gradient, particularly in the 0–20 cm surface layer, whereas the responses of these fractions were relatively weak in the 40–60 cm soil layer. These results indicate that the effects of straw return on labile organic carbon pools showed a clear vertical gradient, with stronger accumulation in the surface soil and weaker responses in deeper soil layers. Similar findings were reported by Zhang et al. [51], who found that straw return increased DOC, POC, and MBC in black soil.

The measured increases in MBC and carbon-acquiring enzyme activity suggest that straw return enhanced microbial biomass and the potential activity of enzymes involved in carbon substrate acquisition. All straw-return treatments increased MBC content and C-acquiring enzyme activity compared with CT, with relatively greater responses under DTS and NTS. These results are consistent with previous studies showing that straw return can increase microbial biomass and stimulate extracellular enzyme activity [52]. The increase in DOC, ROC, LFOC, and POC may have provided more available carbon substrates for microbial growth and enzymatic decomposition.

Notably, labile carbon pools are closely coupled with microbial metabolic processes. As one of the most sensitive labile carbon fractions, DOC directly supplies energy for microorganisms and can be selectively utilized by specific functional groups [53]. Increased DOC generally stimulates microbial growth and induces the production and secretion of carbon-acquiring hydrolases, such as β -glucosidase (BG) and cellobiohydrolase (CBH), through a substrate-induced effect, thereby enhancing C_{acq} activity and accelerating organic matter depolymerization [54]. In this study, DOC showed a similar trend to MBC and C_{acq} activity, indicating that straw return not only increased microbial biomass but also enhanced microbial acquisition and transformation of labile carbon substrates. This process further promoted the conversion of particulate carbon into soluble carbon [55,56].

However, higher enzyme activity does not necessarily indicate a net increase in SOC; rather, microbial carbon use efficiency determines whether substrate carbon is lost through respiration or transformed into microbial residues [57]. Cotrufo et al. [58] suggested that, with substantial inputs of readily decomposable organic matter, labile carbon can be assimilated by microorganisms and converted into microbial residues, which may then associate with aggregates and form more stable mineral-associated organic carbon, thereby expanding the stable SOC pool.

Overall, the results indicate that straw-return and tillage systems altered labile carbon distribution, microbial biomass carbon, and carbon-acquiring enzyme activity in a depth-dependent manner. NTS mainly promoted the accumulation of LFOC and POC in the surface layer, probably because surface straw mulching and reduced disturbance favored the retention of particulate organic matter. In contrast, DTS increased DOC, ROC, MBC, and enzyme activity in the 20–40 cm layer, likely because deeper straw incorporation supplied organic substrates to the subsurface soil. However, further studies combining microbial community analysis, microbial carbon-use efficiency, aggregate-associated carbon, mineral-

associated organic carbon, and straw-derived carbon tracing are needed to verify the mechanisms of carbon transformation and stabilization under different straw-return and tillage systems.

4.3. Comprehensive Effects of Optimized Straw Management–Tillage Systems on Soil Quality Improvement

Straw return under suitable tillage regimes is considered an effective strategy for mitigating organic matter depletion caused by continuous conventional plowing and for improving carbon stock in black soil croplands [59]. In the present study, both DTS and NTS increased SOC and SOC_{stock} within the 0–60 cm soil profile. However, their dominant soil layers differed: NTS was more favorable for SOC accumulation in the 0–20 cm surface layer, whereas DTS was more effective in increasing SOC in the 20–40 cm subsurface layer. These results indicate that DTS and NTS have depth-specific advantages in improving soil carbon accumulation rather than one practice being universally superior [46]. Similarly, both DTS and NTS increased AN, AP, and AK contents, but their effects differed with soil depth. DTS was more effective in improving nutrient storage and promoting a more uniform nutrient distribution within the 0–40 cm soil layer, whereas NTS mainly favored surface nutrient enrichment [60,61]. These differences suggest that DTS and NTS may improve soil functions through different pathways: DTS through “subsurface improvement” and NTS through “surface conservation.”

From the perspective of SQI, straw–return practices improved soil quality in black soil farmland, with NTS mainly increasing SQI in the 0–20 cm cultivated layer and DTS enhancing SQI in the 20–40 cm subsoil layer. The PLS-PM results indicated statistical associations among aggregate stability, labile carbon fractions, nutrient availability, enzyme activity, and SQI. Specifically, the model suggested that improved soil structural stability was associated with greater accumulation of labile organic carbon fractions, which were further associated with higher nutrient availability and carbon-acquiring enzyme activity, ultimately contributing to SQI improvement.

DTS mainly enhanced subsoil quality by improving aggregate structure, expanding carbon pools, and promoting the accumulation of ROC and DOC in the 20–40 cm layer. In contrast, NTS mainly improved surface soil quality by optimizing aggregate structure, increasing surface carbon storage, and enhancing LFOC and POC accumulation in the 0–20 cm layer. In terms of soil structural improvement, enhanced aggregate stability can strengthen the physical protection of organic carbon and promote its association and preservation within aggregates or on mineral surfaces [62,63]. Meanwhile, the increase in labile carbon pools provides sufficient metabolic substrates for microorganisms, promoting microbial biomass growth and extracellular enzyme activity, thereby strengthening the synergistic processes among organic residue decomposition, nutrient mineralization, and soil structure formation [53]. Future studies should further examine microbial communities, carbon-use efficiency, aggregate- and mineral-associated carbon, and straw-derived carbon dynamics to clarify the mechanisms of long-term SOC stabilization.

In summary, combined straw-return and tillage systems improved SOC, labile carbon fractions, and SQI in the 0–40 cm soil layer of black soil farmland, but their dominant response pathways differed among practices. NTS reduced mechanical disturbance and maintained surface straw mulch, creating a relatively stable and protected cultivated layer, thereby strongly promoting carbon accumulation and preservation in the 0–20 cm surface layer. In contrast, DTS directly incorporated straw residues into the 20–40 cm subsoil layer, thereby increasing organic substrate input at depth. It may also loosen the compacted subsoil, improve porosity, and promote deeper root growth [64]. Root residues and exudates can further provide carbon sources for microbial activity and aggregate formation [65]. These combined effects may explain why DTS was more effective in

enhancing SOC accumulation and SQI in the 20–40 cm layer. By comparison, DHS and RTS showed weaker effects on carbon sequestration and soil quality enhancement than DTS and NTS, mainly because of stronger soil disturbance and limited straw distribution depth and stabilization. Therefore, NTS is more suitable for surface soil conservation and carbon sequestration, whereas DTS has greater advantages in promoting carbon allocation to the subsoil and improving deeper cultivated-layer quality.

5. Conclusions

Straw return combined with tillage improved soil quality in the 0–60 cm black soil profile, with clear depth-dependent responses. NTS mainly enhanced surface soil quality by promoting aggregate stability and SOC accumulation in the 0–20 cm layer, whereas DTS showed greater potential for improving subsoil fertility and carbon storage in the 20–40 cm layer. Overall, NTS and DTS appeared more promising than DHS and RTS for improving soil quality and carbon sequestration. Nevertheless, the broader applicability of these findings requires further validation through long-term, multi-site studies across different climatic conditions, soil types, cropping systems, and productivity outcomes.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/agriculture16131407/s1>, Figure S1: Changes in soil β -glucosidase (BG) activity in the 0–60 cm soil layer; Figure S2: Changes in soil β -xylosidase (BX) activity in the 0–60 cm soil layer; Figure S3: Changes in soil β -1,4-cellobiohydrolase (CBH) activity in the 0–60 cm soil layer. Table S1: Physical and chemical properties of 0–60 cm soil layer before the test started in 2018; Table S2: Straw-return amount under different treatments after maize harvest from 2018 to 2024. Table S3: Changes in soil bulk density under different straw management–tillage systems. Table S4: Changes in the contents of soil labile organic carbon fractions. Table S5: Changes in soil available nutrient contents.

Author Contributions: Conceptualization, M.R., X.Z. and Z.W.; methodology, M.R. and Z.C.; software, M.R. and Z.C.; validation, L.B., Q.M. and X.G., Y.W.; formal analysis, M.R. and Z.C.; investigation, F.L. and H.W.; resources, T.M., J.S. and H.L.; data curation, M.R. and Z.C.; writing—original draft preparation, M.R. and Z.C.; writing—review and editing, X.Z. and Z.W.; visualization, M.R. and Z.C.; supervision, L.B., Q.M., X.G. and Y.W.; project administration, T.M., J.S. and H.L.; funding acquisition, X.Z. and Z.W. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by Natural Science Foundation of China, grant number 32460534; Major Demonstration of Technological Innovation in Inner Mongolia Autonomous Region, grant number 2025ZDSF0028; and Scientific Research Support Fund Project, grant number 2026KYBZN14.

Data Availability Statement: The original contributions presented in this study are included in the Supplementary Material. Further inquiries can be directed to the corresponding authors.

Conflicts of Interest: The authors declare no conflicts of interest.

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