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Conservation tillage enhances both organic and inorganic carbon in dryland: Insights from a 20-year field experiment and meta-analysis

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ABSTRACT

Conservation tillage is widely recognized as a promising practice for sequestering soil organic carbon (SOC). However, its impact on soil inorganic carbon (SIC) remains less understood and has seldom been quantified. This study aimed to examine the effects of conservation tillage on soil carbon pools, focusing on SIC, by combining a 20-year field experiment in an arid-calcareous cropland of China with a meta-analysis of 76 pairwise data from 7 studies. The field experiment confirms that conservation tillage significantly increases carbon stock, with reduced tillage (RT) increasing SOC (25.09 %, 0–40 cm) and no-tillage (NT) increasing SIC (10.67 %, 0–20 cm). SOC and SIC exhibit a complementary relationship, whereby an increase in SOC effectively compensates for reduced SIC within RT. Notably, the proliferation of calcifying bacteria (e.g., *Bacillus*) and reduced urease activity suggest that microbial-induced carbonate precipitation, a process known to be facilitated by these bacteria, may contribute significantly to SIC formation under NT. Agronomic practices, as well as soil abiotic and biotic factors, collectively influence SIC. The relative importance of these factors varies with soil depth: biotic variables effects weaken with depth, while abiotic variables increase. Furthermore, our meta-analysis reveals that the response of SIC to conservation tillage varies with climatic, edaphic, and agronomic factors. Arid regions benefit the most from NT in enhancing SIC stock (3.27 %). These findings provide valuable insights into how conservation tillage influences soil carbon, particularly SIC, and enhance our understanding of carbon dynamics in arid systems.

1. Introduction

Soil constitutes the largest carbon pool in terrestrial ecosystems (Jobbágy and Jackson, 2000), consisting of two distinct components: soil organic carbon (SOC) and soil inorganic carbon (SIC), and plays a crucial role in the global carbon cycle (Lal, 2004). In efforts to mitigate the increasing atmospheric CO_2 concentrations, previous studies have predominantly concentrated on SOC due to its association with various ecosystem functions or services and its rapid response to agronomic practices such as tillage, straw return, and fertilization (Lin et al., 2023; Tian et al., 2024; Ding et al., 2025a). In contrast to SOC, SIC is often overlooked as a carbon stock because it is perceived as relatively stable and less responsive to agronomic practices over the mankind lifetime (Schlesinger, 1985; Zamanian et al., 2016). However, as evidence of accelerated SIC dynamics continues to ascend, this perspective is shifting (Kim et al., 2020; Huang et al., 2024). Recent findings have revealed that agronomic practices can fundamentally alter SIC cycling and lead to SIC sinks or SIC sources (Bughio et al., 2016; Zamanian et al., 2018; Raza et al., 2020; Song et al., 2022). SIC comprises most of the soil carbon pool, especially in drylands, where it accounts for 90 % of total carbon (TC) stock (Filippi et al., 2020). The presence of SIC affects the soil properties, acidity-buffering capacity, and nutrient availability

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(Sharififar et al., 2023; Tao and Houlton, 2024). Therefore, the maintenance of SIC holds crucial significance for reducing carbon loss while preserving soil health and productivity (Zamanian et al., 2021).

Among various land use types, cropland undergoes tremendous disturbance, and farmland soil is subject to degradation, typically acting as a carbon source (Chappell et al., 2016). Recommended best management practices, like conservation tillage (e.g., no-tillage (NT) and reduced tillage (RT)), have emerged as potential solutions (Van Kessel et al., 2013; Cambron et al., 2024). Historically, carbon research regarding conservation tillage has been focused on SOC (Ashworth et al., 2014; Powlson et al., 2014; Duval et al., 2016; VandenBygaart, 2016; Du et al., 2017; Bai et al., 2019; Li et al., 2020, 2024a; Xiao et al., 2021). Only a few studies have examined SIC, demonstrating that in certain arid and semi-arid conditions, the conversion from conventional tillage (CT) to conservation tillage, along with the retention or application of crop residues and fertilizers, can either decrease (Jacinthe et al., 2011) or increase (Sainju et al., 2007; Dey et al., 2020) SIC. Changes in SIC stock induced by conservation tillage are observed to be variable and highly dependent on experimental design (e.g., residue management, nitrogen fertilization, duration, or irrigation) and site-specific conditions (e.g., soil properties and climate) (Du et al., 2017; Li et al., 2020). However, to date, a comprehensive quantitative analysis of the influence of conservation tillage on SIC stock at the national scale of China remains absent.

Furthermore, SIC has traditionally been considered to be primarily driven by abiotic processes (Vicca et al., 2022). However, recent studies have found that, similar to SOC, the dynamic variations in SIC are intricately associated with numerous biotic and abiotic processes (Liu et al., 2018; McKenna et al., 2022; Ball et al., 2023). Tillage and straw return affect the soil microbial community directly by changing substrate quantity or quality, or indirectly by altering soil conditions that influence microbial structure, function, and stability (Hao et al., 2019). This, in turn, enhances the role of biological processes in pedogenic carbonate formation (Fernández-Ugalde et al., 2011). For instance, elevated pCO_2 within the soil matrix resulting from root and microbial respiration would affect secondary carbonate formation (Gocke and Kuzyakov, 2011). Recent studies have reported another metabolic approach of microbial-induced carbonate precipitation (MICP), which can directly promote secondary carbonate formation (Görgen et al., 2021; Liu et al., 2023). MICP is a widespread biomineralization process in which carbonates undergo precipitation due to the intricate workings of microbial growth and metabolism (Wang et al., 2024). Native ureolytic microorganisms in soil catalyze the decomposition of urea into ammonium and carbonate ions, which subsequently precipitate as calcium carbonate minerals in the presence of Ca^{2+} (Comadran-Casas et al., 2022). Considering this, it is likely that MICP plays a crucial role in SIC pools (Renforth et al., 2009), particularly within agronomic practices that facilitate the development of microbial communities in calcium-rich soil, such as the arid and calcareous soil in northern China (Ennaciri et al., 2022). Despite its importance, the contribution of MICP to SIC stock in agricultural soils remains virtually unknown.

In this study, a 20-year field experiment—one of the longest empirical evaluations of conservation tillage effects in arid-calcareous soils—was conducted in China to (1) assess whether tillage practices differentially affect the accumulation of SOC, SIC, and TC, as well as whether these effects are mediated by microbial factors. Additionally, a meta-analysis was performed using historical data from other long-term experiments to (2) determine the broader response pattern of SIC. We hypothesized that (1) RT would enhance SOC levels due to reduced tillage combined with straw incorporation, (2) while NT would result in higher SIC levels through increased carbonate precipitation; (3) NT-induced changes in SIC stock vary significantly among different moderators (e.g., climate, soil properties, and agronomic management).

2. Materials and methods

2.1. 20 years' field experiment

2.1.1. Site description

A continuous field experiment was initiated in 2003 at the National Dryland Farming Experimental Station, located on the Loess Plateau of China (Shouyang, Shanxi Province, China, 37° 58′ N, 113° 10′ E, 1100 m above sea level) (Fig. 1**a**). The site exhibits a typical continental monsoon climate, characterized by a mean annual air temperature (MAT) of 7.4 °C, a mean annual precipitation (MAP) of 483 mm, a mean annual evaporation of 1750 mm, a mean sunshine duration of 2858 h, and a mean annual frost-free period of 130 days. Soil type is sandy loam, specifically identified as a Calcaric-Fluvic Cambisol (Wang et al., 2011). Prior to the initiation of the experiment, soil exhibited an organic matter content of 25.7 g kg^{-1}, total nitrogen (TN) of 1.04 g kg^{-1}, available nitrogen of 54 mg kg^{-1}, available phosphorus (AP) of 7.3 mg kg^{-1}, available potassium (AK) of 84 mg kg^{-1}, pH of 7.87, and a bulk density of 1.13 g cm^{-3} within the depth range of 0–20 cm.

2.1.2. Experimental design

The field experiment employs a randomized complete block design. The three treatments are as follows (Fig. 1**b**): (1) CT with maize straw removal, annual plow twice after harvest in October and before sowing in April with a depth of about 25 cm, and fertilizer is applied before sowing; (2) NT with maize straw mulch retention, no annual plowing, and the seed and fertilizers are applied with a no-till seed drill in April; and (3) RT with fertilizers and maize straw incorporation and annual plow once with a depth of about 25 cm after harvest. Each treatment is conducted with three replicates. The cropping system is a continuous rain-fed monoculture maize crop, which is fallowed from November to March each year. Each plot receives the same amounts of fertilizer: N 105 kg ha^{-1} y^{-1} (urea), P_2O_5 105 kg ha^{-1} y^{-1} (calcium superphosphate). The tested maize varieties are local dominant varieties, sown in April at a density of 30 kg ha^{-1}. The growth period is from April (sowing) to October (harvest). More detailed field operations are summarized in Table S1.

2.1.3. Soil sampling and preparation

In October 2023 (20 years after the experiment was established), soil samples were collected randomly from 0–100 cm layer within each plot using a soil auger. Three undisturbed soil cores were collected from each plot using a steel ring from depths of 0–20, 20–40, 40–60, 60–80, and 80–100 cm, respectively. Subsequently, soil samples were stored at 4 ℃. Prior to analysis, fresh soil samples underwent sieving through a 2 mm stainless steel sieve. Soil samples with a particle size smaller than 2 mm were separated into three parts: one portion was preserved at –80 ℃ for microbial analyses, a second portion was maintained under field-moist conditions at 4 ℃ to assess soil moisture content (SMC), soil respiration intensity, ammonium nitrogen (NH_4^+-N), and nitrate nitrogen (NO_3^--N) content, while the third portion was air-dried for the measurement of soil carbon, TN, AP, AK, exchangeable calcium (Ca) and magnesium (Mg), urease activity (UA), soil clay content, soil electrical conductivity (SEC), and pH.

2.1.4. Soil physiochemical analysis

SMC of field-moist soil was quantified gravimetrically after being dried at 105 °C for a duration of 24 h. Soil pH was determined using pH meters (1:2.5 w/v). SEC was measured using conductivity meter (1:5 w/v). Soil texture analysis was performed with a laser particle size analyzer. TC and TN were analyzed using a Vario Macro C/N analyzer (Elementar, Germany). Calcium carbonate ($CaCO_3$) was quantified by manometrically collecting the evolved CO_2 after the addition of hydrochloric acid (4 mol L^{-1}) (Wu et al., 2009). SIC is primarily composed of $CaCO_3$, with other carbonate types present in negligible amounts. Therefore, the conversion from $CaCO_3$ values to SIC used a constant

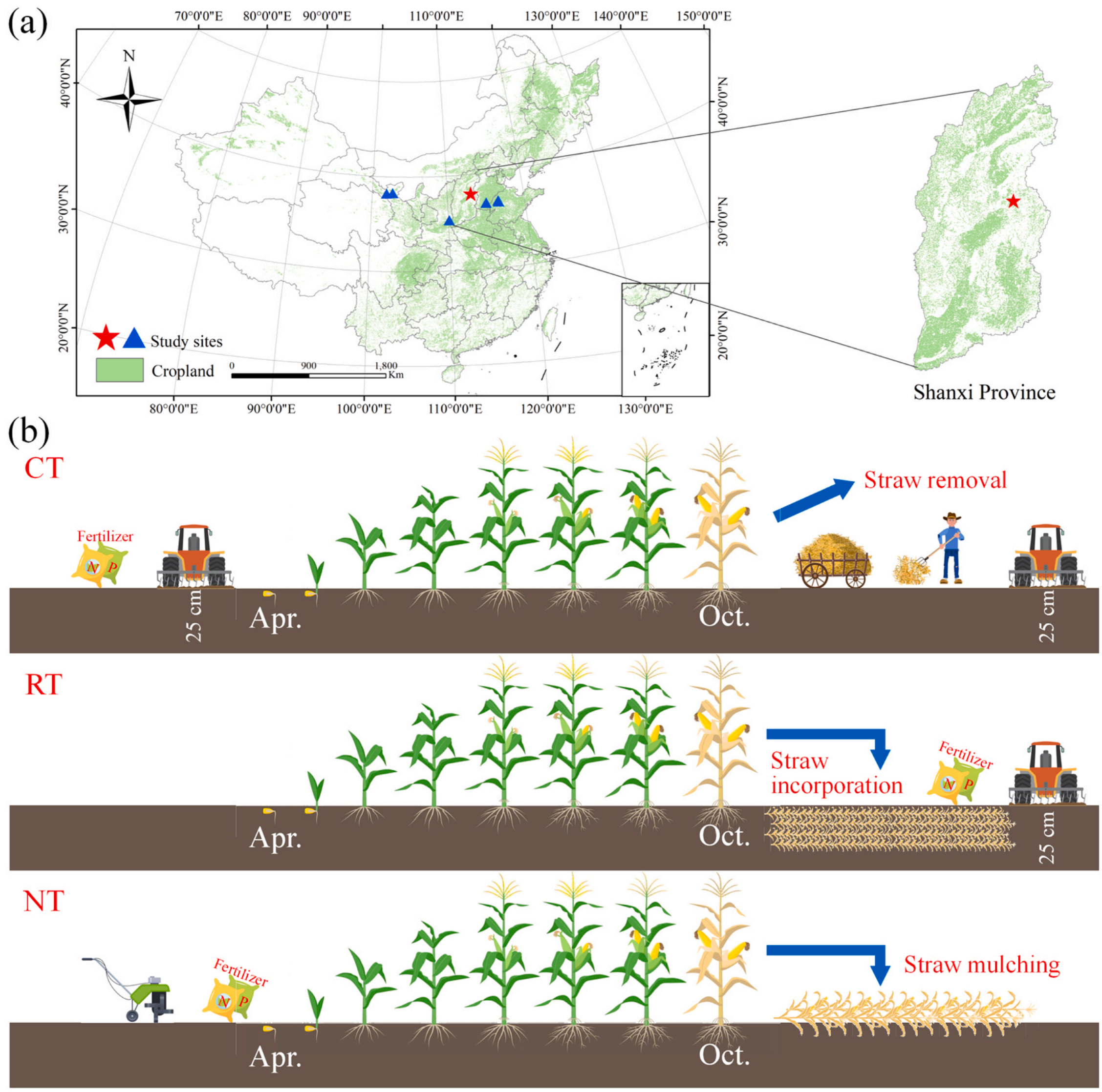

Fig. 1. Location of the study and sampling sites in China. The five-pointed star indicate the sampling points of the 20-year field experiment, whereas the triangular and five-pointed star jointly represent the sampling points included in the meta-analysis (a) and the long-term field experimental design (b). Abbreviations: CT, conventional tillage; RT, reduced tillage; NT, no-tillage.

factor of 0.12 (Yang et al., 2012). SOC was then determined by subtracting SIC from TC. The equation for calculating carbon stocks can be found in the Supplementary materials. Dissolved organic carbon (DOC) was extracted with ultrapure water and air-dried samples (1:5 w/v). The extract was filtered using a 0.45 μm filter and then analyzed with an Elementar TOC analyzer (Vario TOC, Elementar, Germany). NH_4^+-N and NO_3^--N were extracted (1:5 w/v) using 2 M KCl and were quantified using a continuous flow auto-analyzer (Autoanalyzer 3, SEAL, Germany). AP was extracted utilizing a 0.5 M $NaHCO_3$ and was quantified employing molybdenum blue colorimetry (Truog, 1930). AK was quantified via a flame photometry-based extraction process utilizing 1 M NH_4OAC. Exchangeable calcium (Ca^{2+}) and magnesium (Mg^{2+}) were extracted with ammonium acetate and subsequently analyzed via flame photometry (Shaw and Veal, 1956).

2.1.5. *Soil microbial properties analysis*

UA, which intuitively reflects MICP process (Li et al., 2014), was quantified by the sodium phenolate-sodium hypochlorite colorimetric method (mg g^{-1} 24 h^{-1}) (Tabatabai and Bremner, 1972). Soil respiration (CO_2) was measured using the alkali absorption titration method (Yim et al., 2002). The 0.5 g fresh soil were used to extract total DNA using a MagBind soil DNA kit. The quality of the extracted DNA was examined via 1.2 % agarose gel electrophoresis. The extracted DNA was subsequently utilized for quantitative analysis as well as 16S rRNA and ITS gene amplicon sequencing. The bacteria 16S rRNA gene and fungi 18S rRNA gene were amplified using primer pair 338 F/806 R (Derakhshani et al., 2016) and ITS1F/ITS2R (Luan et al., 2015), respectively.

2.1.6. *Statistical analyses*

IBM SPSS 25 was used to compare significant differences through one-way ANOVA (Duncan test, $p < 0.05$) in soil properties (Table S2). Prior to statistical analysis, we tested the assumption of homogeneity of variance using Levene's test and confirmed that the data for each variable satisfied this assumption. OTUs with an average relative abundance greater than 0.01 % were included in the soil microbial co-occurrence network analysis, and the network was visualized in Gephi (version 0.10.1) using the Fruchterman Reingold layout algorithm. We conducted variable partitioning analysis (VPA) to quantify the contributions of agronomic practices, soil biotic and abiotic factors, as well as their interactions, to the variation in SIC. VPA was performed using the R software (version 3.4.2) with the 'vegan' package. Partial least squares path model (PLS-PM) analysis was employed to evaluate the relationships among soil properties (abiotic factors), agronomic practices, soil microorganisms (biotic factors), and SOC and SIC. The performance of the PLS-PM model was assessed using the goodness of fit (GoF) index. With a good model fit, we could explain the model's path coefficients, which describe both the magnitude and direction of relationships between variables. PLS-PM was carried out using the 'plspm' package (999 bootstraps) in R (version 4.2.2).

2.2. Meta-analysis for the effect of no-tillage on SIC stock in Chinese farmland

2.2.1. Data collection

To assess the general response of SIC stock to no-till in Chinese farmlands, we performed an exhaustive literature search based on two databases: China National Knowledge Infrastructure (https://www.cnki.net) and Web of Science (https://www.webofscience.com). The query sets were "conservation tillage" OR "no-till" AND "soil inorganic carbon". Each retrieved paper was carefully read to ensure it contained field observations of SIC. To avoid potential bias, we established the following screening criteria: (1) experiments were conducted under field conditions; (2) laboratory incubation experiments and greenhouse studies were excluded; (3) the experimental design involves pairwise comparisons between NT and CT with at least three repetitions; (4) each study include at least one CT control and one NT treatment; (5) mean value and standard deviation (SD) or standard error for treatment and control groups were clearly reported. This meta-analysis included a total of 7 studies, comprising 76 observations (Fig. 1**a** and Table S3).

In addition, we incorporated a range of environmental variables associated with the conservation tillage experiments, including longitude, latitude, altitude, MAP, MAT, initial soil physicochemical properties (e.g., SOC, SIC, TN, and pH), soil depth, experimental duration, and nitrogen fertilizer input rate. The above-mentioned eight variables, including MAT, MAP, duration, N input, initial SOC, SIC, TN, and soil depth, were further categorized into sub-groups (Table S4), based on previous meta-analyses (Xiang et al., 2022; Lin et al., 2023; Ding et al., 2024b, 2024c, 2025b) and our dataset characteristics.

2.2.2. Meta-analysis

The meta-analysis was conducted to estimate the effect size of different studies and categorical variables (Hedges et al., 1999). The natural logarithm of the response ratio (ln*RR*) was determined to evaluate the variations between NT and CT.

$$\ln RR = \ln \frac{\overline{x}_{NT}}{\overline{x}_{CT}} \tag{1}$$

where $\overline{x}_{NT}$ and $\overline{x}_{CT}$ are the arithmetic mean values of the NT and CT, respectively. The corresponding variance (v) of ln*RR* was estimated as:

$$v = SD_{NT}^2/(n_{NT} \times x_{NT}^2) + SD_{CT}^2/(n_{CT} \times x_{CT}^2) \tag{2}$$

The weighting factor (w_i) of each response ratio was computed as:

$$w_i = 1/v_i \tag{3}$$

where w_i and v_i are the weight and variance of *i*th study, respectively.

$$\ln RR_{++} = \frac{\sum_i (\ln RR_i \times w_i)}{\sum_i w_i} \tag{4}$$

The chi-square test was employed to evaluate heterogeneity. Additionally, the I^2 statistic was used to quantify the proportion of heterogeneity in the overall variation of $\ln RR_{++}$. An I^2 value greater than 75 % suggests substantial heterogeneity (Senior et al., 2016). The 95 % confidence intervals (CIs) were calculated using a bootstrapping procedure with 4999 iterations (Zhao et al., 2020). The effects of NT were considered significant if the 95 % CIs excluded zero ($p < 0.05$). The frequency distribution of the ln*RR* of SIC fitted by a Gaussian function (Zheng et al., 2023; Ding et al., 2024a). The meta-analysis was performed using MetaWin 2.1. Random forest analysis was performed utilizing the "randomForest" package (Breiman, 2001) in R software (version 3.4.2) to identify the relative importance of environmental factors.

3. Results

3.1. Changes in soil biotic and abiotic properties

20 years' conservation tillage significantly altered soil properties (Fig. 2), with effects varying by depth and tillage type. NT and RT significantly improve soil quality compared with CT, as indicated by the largest shaded area regardless of soil depth. Specifically, NT increases average SMC by 25.9 %, SOC by 4.1 %, SIC by 11.2 %, TN by 10.3 %, pH by 0.14 units, SEC by 11.2 %, NO_{-3}-N by 21.8 %, NH_4^+-N by 30.1 %, AK by 20.0 %, AP by 108.8 %, Shannon-B by 9.3 %, and Shannon-F by 2.6 % at 0–20 cm depth. However, these increases diminished with depth for both NT and RT (Table S5).

3.2. Soil carbon stock and depth distribution

To examine the effects of tillage on soil carbon pool, variations in SIC (Fig. 3**a**), SOC (Fig. 3**b**), TC stock (Fig. 3**c**), and SIC/SOC ratios (Fig. 3**d**) were compared using the Duncan test. Results show that conservation tillage, particularly RT, significantly increases SOC stock by 25.1 % (0–40 cm, $p < 0.05$). NT showed the highest SIC stock (0–20 cm, $p < 0.05$), followed by CT, while RT records the lowest SIC stock (0–40 cm, $p < 0.05$). Both RT and NT significantly increase TC stock (0–40 cm) by 11.7 % and 5.7 %, respectively. No significant differences in SIC, SOC, and TC stock are observed at 40–100 cm across treatments. SIC/SOC ratio ranges from 0.3 to 5.3, with values below 1 in the 0–40 cm and above 1 in the 40–100 cm.

3.3. Effect of tillage practices on soil pH, Ca^{2+}, Mg^{2+}, and UA and their correlations with SIC and SOC

The impact of tillage practices varies for soil pH (Fig. 3**e**), Ca^{2+} (Fig. 3**f**), Mg^{2+} (Fig. 3**g**), and UA (Fig. 3**h**). NT consistently increases soil pH compared to RT and CT across all depths ($p < 0.05$), while RT decreases pH by −0.2 units compared to CT ($p < 0.05$). At 0–20 cm, RT and NT show higher concentrations of Ca^{2+} and Mg^{2+}. Compared to CT and RT, NT reduces UA by 7.6 % and 13.6 %, respectively. SOC correlates negatively with pH, Ca, and Mg, while SIC correlates positively with these variables (Fig. 3**i–k and m–o**). UA shows a positive correlation with SOC and a negative correlation with SIC (Fig. 3**l and p**).

3.4. Bacterial and fungal composition and co-occurrence networks

We found significant effects of tillage practices on microbial composition (Fig. 4**A**). At the phylum level, dominant bacterial communities include *Actinobacteriota* (20.6 %), *Acidobacteriota* (20.1 %), *Proteobacteria* (18.3 %), *Gemmatimonadota* (8.2 %), and *Chloroflexi* (8.2 %). The relative abundance of *Proteobacteria* is greater under CT (18.8 %), followed by RT (18.0 %) and NT (17.9 %). *Acidobacteriota* has the highest proportion in NT (21.0 %), followed by RT (19.8 %) and CT (19.4 %). The abundance of *Acidobacteriota*, *Myxococcota*, *Bacteroidota*, and *Planctomycetota* increases, whereas the relative abundance of *Proteobacteria* decreases under NT and RT. *Firmicutes*, which consist mostly of ureolytic microorganisms, show particularly enhanced growth in the topsoil under NT. The abundance of *Firmicutes* increases by 117.2 % and 221.1 % compared to CT and RT, respectively (Fig. 4Aa). Further investigation at the genus level reveals that the abundance of *Bacillus* (a genus within *Firmicutes*) in the topsoil under NT increases by 129.1 % and 196.7 % compared to CT and RT, respectively (Fig. 4Ac). This suggests that NT stimulates the proliferation of urease-producing

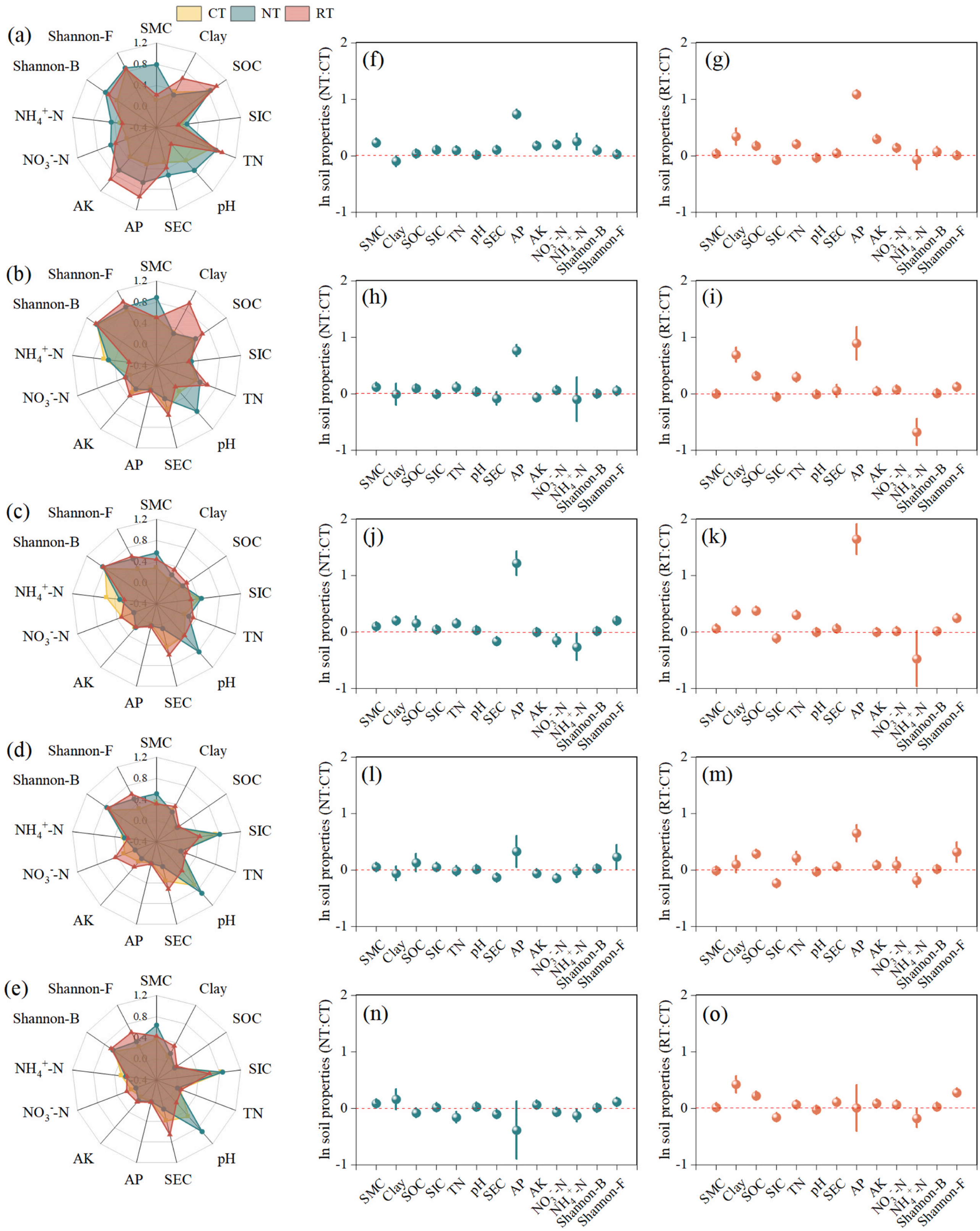

Fig. 2. The impact of tillage practices on soil properties across varying soil depths (a, 0–20 cm; b, 20–40 cm; c, 40–60 cm; d, 60–80 cm; e, 80–100 cm) and relative changes in soil characteristics between conservation tillage (RT and NT) and CT (f and g, 0–20 cm; h and i, 20–40 cm; j and k, 40–60 cm; l and m, 60–80 cm; n and o, 80–100 cm). Each soil variable is standardized into dimensionless units, facilitating the comparability of indicators across different treatments. A larger shaded area corresponds to a higher soil quality. Values represent the mean ± standard error. Abbreviations: CT, conventional tillage; RT, reduced tillage; NT, no-tillage; SMC, soil moisture content; SOC, soil organic carbon; SIC, soil inorganic carbon; TN, total nitrogen; SEC, soil electrical conductivity; AP, available phosphorus; AK, available potassium; Shannon-F: fungal diversity; Shannon-B: bacterial diversity.

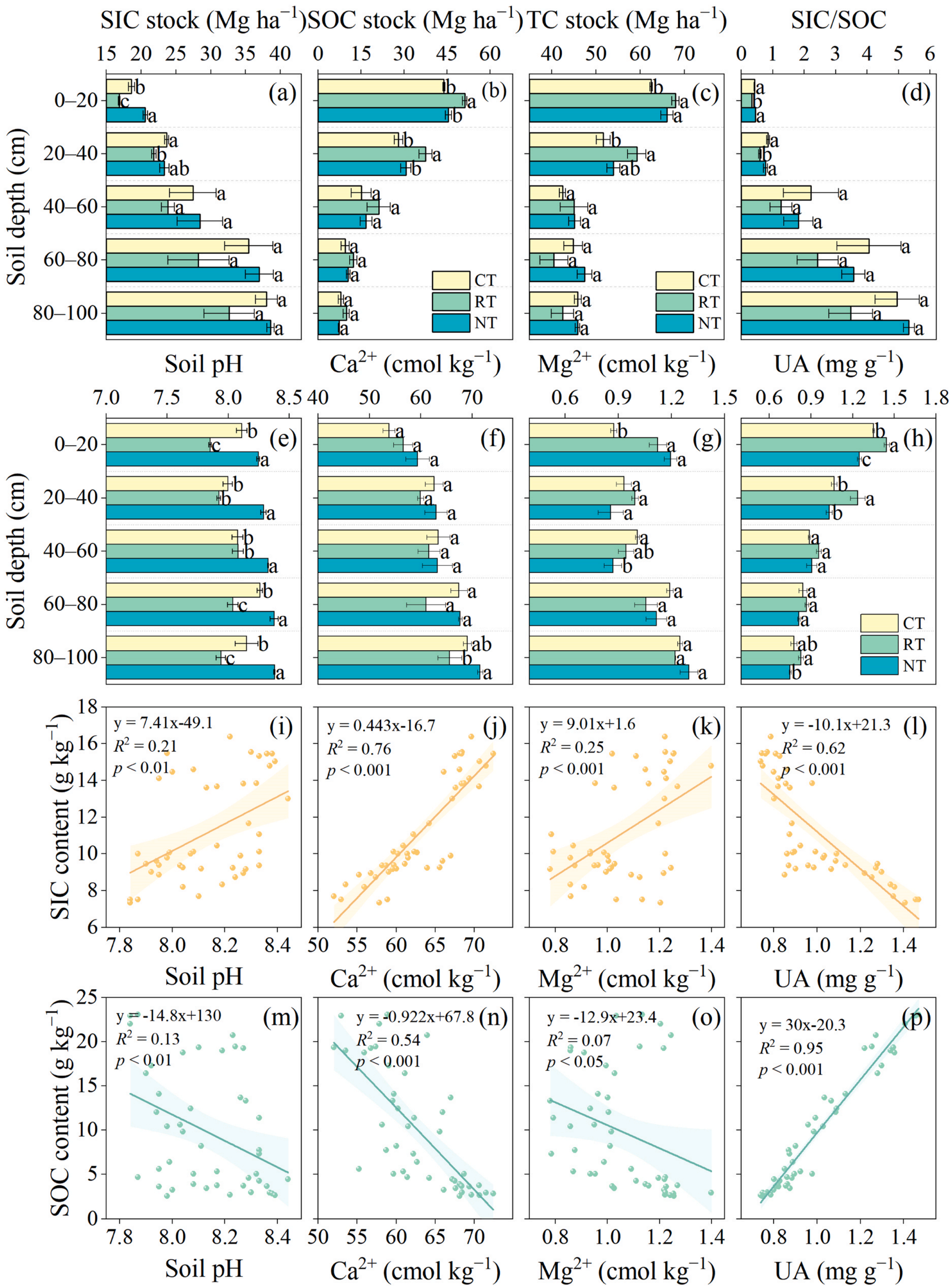

Fig. 3. Stocks of SIC (a), SOC (b), and TC (c), SIC/SOC ratio (d), soil pH (e), Ca^{2+} (f), Mg^{2+} concentration (g), and UA (h) are evaluated based on soil depth (0–20 cm; 20–40 cm; 40–60 cm; 60–80 cm; 80–100 cm) 20 years after the application of conservation tillage practices. SIC and SOC are related to soil pH, Ca^{2+}, Mg^{2+}, and UA (i–p) by tillage condition at the 0–100 cm depth. Values represent the mean ± standard error. Different lowercase letters indicate significant differences among treatments at the same soil depths ($p < 0.05$). Abbreviations: CT, conventional tillage; RT, reduced tillage; NT, no-tillage; SIC, soil inorganic carbon; SOC, soil organic carbon; TC, soil total carbon; SIC/SOC, the ratio of SIC to SOC; Ca^{2+}, exchangeable calcium; Mg^{2+}, exchangeable magnesium; UA, urease activity.

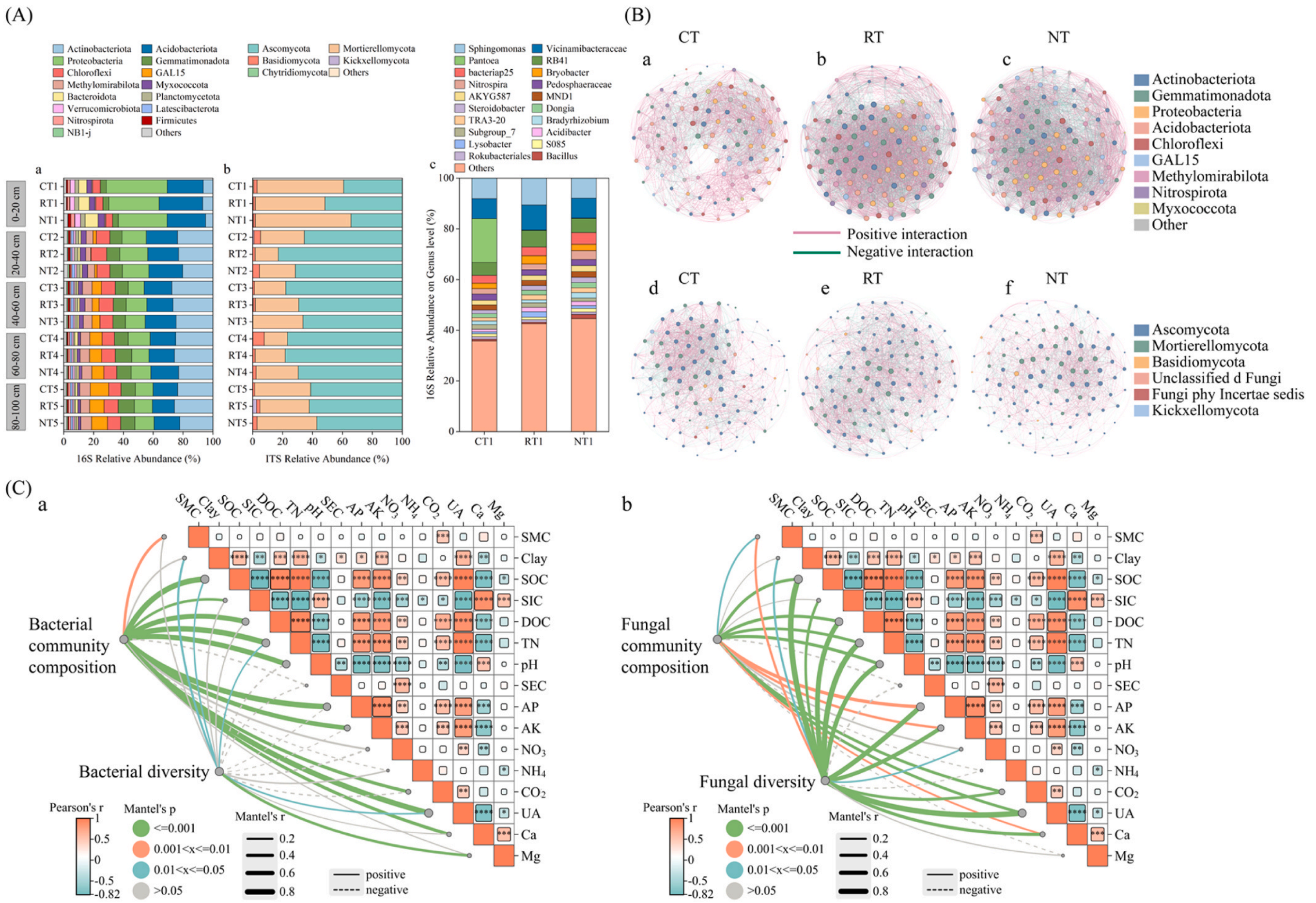

Fig. 4. The relative abundances of the top 15 bacterial (a) and top 5 fungal (b) community structures (0–20 cm; 20–40 cm; 40–60 cm; 60–80 cm; 80–100 cm) at the phylum level, together with top 10 bacterial (c) community structures (0–20 cm) at the genus level of three treatments (A). The microbial co-occurrence networks of the bacterial (a, b, and c) and fungal (d, e, and f) communities (0–100 cm) for three treatments (B). The node size is proportional to the relative abundance. The lines connecting each pair of nodes represent strong positive (red) or negative (green) interactions. Correlations of the environmental variables with bacterial (a) and fungal (b) community composition and diversity. *, $p < 0.05$, **, $p < 0.01$, ***, $p < 0.001$, ****, $p < 0.0001$ (C). Abbreviations: CT, conventional tillage; RT, reduced tillage; NT, no-tillage; SMC, soil moisture content; SOC, soil organic carbon; SIC, soil inorganic carbon; DOC, soil dissolved organic carbon; TN, total nitrogen; SEC, soil electrical conductivity; AP, available phosphorus; AK, available potassium; NO_3^--N, nitrate nitrogen; NH_4^+-N, ammonium nitrogen; CO_2, soil respiration intensity; UA, urease activity; Ca, exchangeable calcium; Mg, exchangeable magnesium.

microorganisms, which in turn enhances the MICP process. The dominant fungal phyla are composed of *Ascomycota* (64.4 %), *Mortierellomycota* (33.0 %), and *Basidiomycota* (2.2 %). *Ascomycota* has the highest proportion in RT (68.9 %), while conservation tillage decreases the relative abundance of *Basidiomycota*. The proportion of *Mortierellomycota* is lowest under RT (28.9 %) and highest under NT (38.0 %) (Fig. 4Ab).

Bacterial and fungal co-occurrence networks at the OTU level are constructed for three treatments (Fig. 4**B**). Different tillage practices significantly alter soil microbial co-occurrence patterns. Conservation tillage enhances the stability of the soil bacterial co-occurrence network (**Fig. 4Bb—c**). Compared with CT, the number of connections and average degree of the microbial network in RT and NT increase by more than 70 %, and the average clustering coefficient increases by 11 %. For the fungal network (**Fig. 4Bd—f**), RT enhances the stability of the soil fungal network, while NT reduces the average degree and average clustering coefficient of fungi, but the average path distance increases by 6.3 %.

The Mantel test is conducted to examine the environmental variables significantly correlated with the composition and diversity of soil bacterial and fungal communities (**Fig. 4Ca–b**). Microbial community composition and diversity are significantly and strongly correlated with soil variables.

3.5. The relative contribution of abiotic and biotic variables to SIC

The VPA revealed that agronomic practices, abiotic and biotic variables, and their interactions collectively explained over 90 % of SIC variability (Fig. 5). The relative importance of these factors varied with soil depth. In the 0–40 cm layer (Fig. 5**a**), the interaction between agronomic practices and biotic and abiotic factors contributed most (77 %), followed by abiotic factors (4.5 %) and biotic factors (3.1 %). In 40–100 cm layer (Fig. 5**b**), abiotic factors dominated (46 %), followed by biotic and abiotic factors interactions (25 %). Unlike agronomic practices and biotic factors, the influence of abiotic factors on SIC strengthened with depth, among which Ca being the strongest predictor.

3.6. Overall responses of SIC stock to NT

A total of 76 pairwise observations (NT *vs*. CT) of SIC stock were extracted. The ln*RR* of SIC stock follows a normal distribution ($R^2 = 0.952$, $p < 0.001$; Fig. 6**a**). The heterogeneity testing ($Q_T = 353.562$, $\tau^2 = 0.01$, $p < 0.001$; $I^2 = 90.9$ %; Table S6) showed that the effects of NT practices vary, with effect sizes ranging from —0.73–3.18. When pooling all data, we find that NT does not result in significant differences in SIC stock (mean: 1.21 %; CI: —0.73 %–3.18 %; Fig. 6**b**).

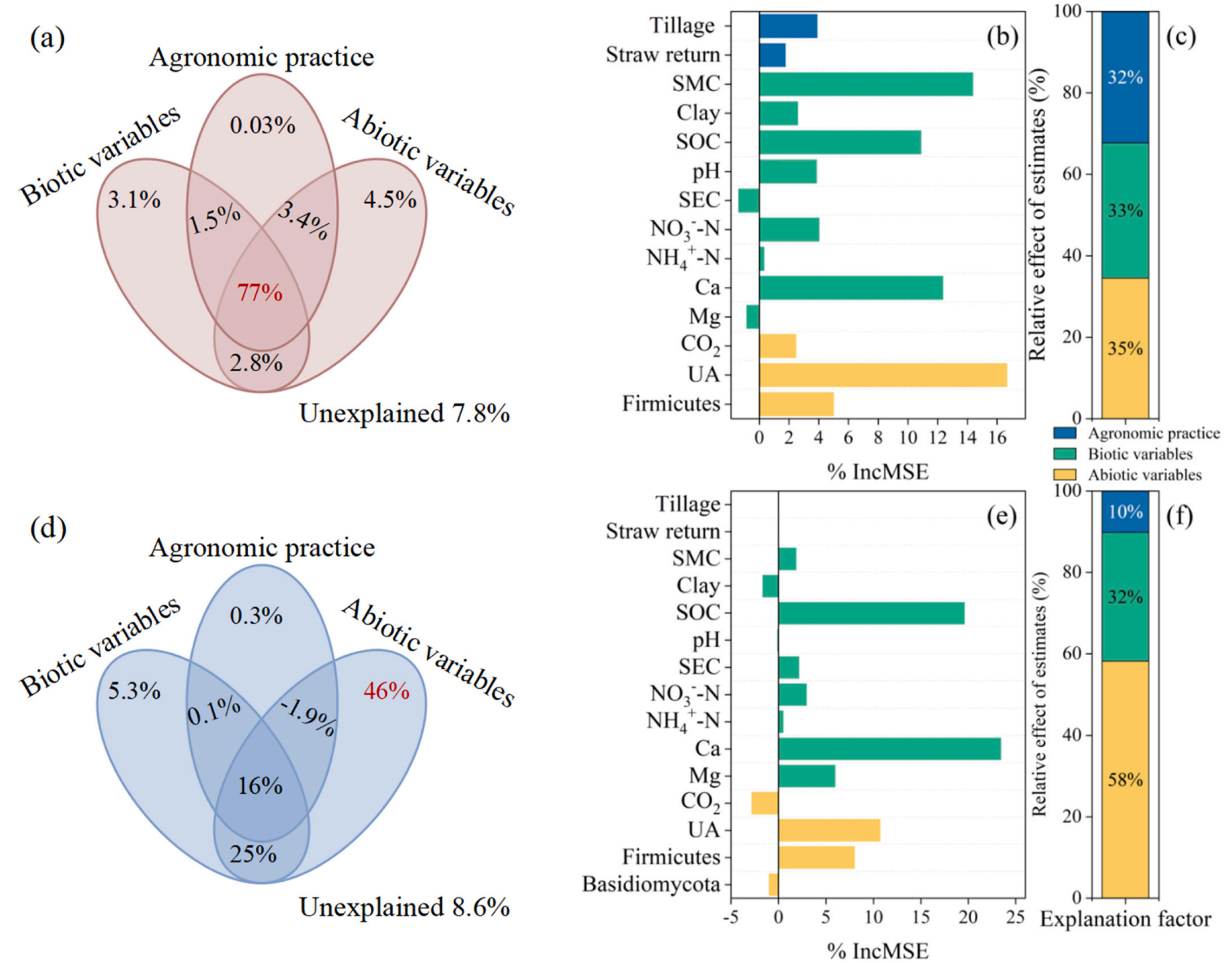

Fig. 5. The impact of environmental factors on SIC. The proportion of the explained variance of SIC changes by agronomic practices, soil biotic and abiotic factors and their interactions (a and d). The relative importance of multiple environmental factors influencing SIC changes (b and e). The relative contribution of three types of environmental factors related to SIC (c and f). (a)–(c) represent the 0–40 cm soil layer, and (d)–(f) represent the 40–100 cm. Abbreviations: SMC, soil moisture content; SOC, soil organic carbon; SIC, soil inorganic carbon; SEC, soil electrical conductivity; NO_3^--N, nitrate nitrogen; NH_4^+-N, ammonium nitrogen; CO_2, soil respiration intensity; UA, urease activity; Ca, exchangeable calcium; Mg, exchangeable magnesium.

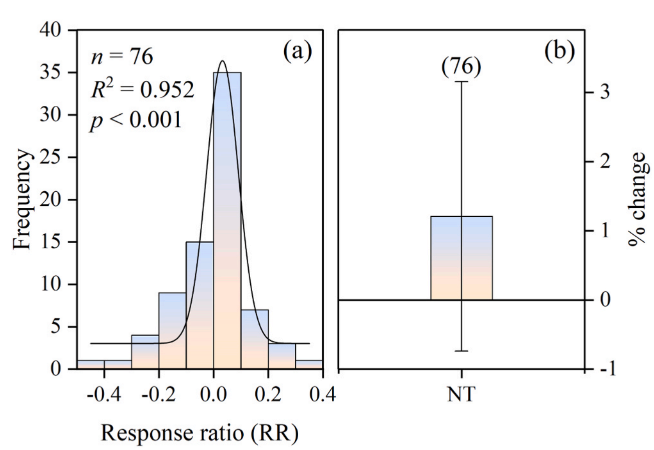

Fig. 6. Frequency distribution of ln*RR* of SIC stock (a) and overall effect of NT practice on SIC stock (b). Error bars represent 95 % confidence intervals (CIs). The 'n' and numbers in parentheses indicate sample sizes of observations. Abbreviations: NT, no tillage.

3.7. Effects of different moderators on SIC stock responses

NT-induced changes in SIC stock vary significantly among different moderators (e.g., climate, soil properties, and agronomic management) (Fig. 7 **and** Table S7). Compared to CT, NT increases SIC stock under the following conditions: (1) MAP < 400 mm (3.27 %, 95 % CIs: 1.47 %–5.47 %), (2) N input < 150 kg N ha^{-1} yr^{-1} (3.64 %, 95 % CIs: 0.04 %–8.05 %), and (3) soil depth at 0–60 cm (5.05 %, 95 % CIs: 0.71 %–8.98 %). However, NT significantly decreases SIC stock in the topsoil (0–10 cm, —7.99 %, 95 % CIs: −11.35 %–−4.51 %). No differences are detected when MAP > 400 mm, N input > 150 kg N ha^{-1}, soil depth (0–20, 20–40, 40–60, 60–80, and 80–100 cm), regardless of MAT, duration of NT, and initial SOC, SIC, and TN ($p > 0.05$).

3.8. Determining major influential factors for increased SIC stock

Random forest model was employed to compare the relative importance of factors influencing SIC stock under NT (Fig. 8**a**). Results show that edaphic, climatic, and experimental variables explain 82 % of SIC stock variation. The ln*RR* of SOC stock is identified as the most critical variable affecting NT-induced changes in SIC stock, followed by

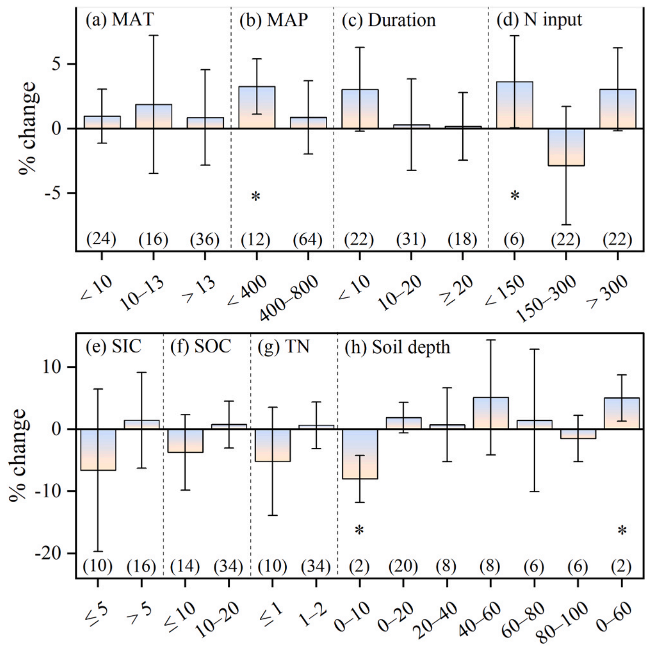

Fig. 7. Effect of NT on SIC stock under distinct soil properties (SIC, soil inorganic carbon; SOC, soil organic carbon; TN, total nitrogen; and soil depth), climatic factors (MAT, mean annual air temperature; MAP, mean annual precipitation), and experimental methods (experimental duration and nitrogen fertilizer input). Error bars represent 95 % confidence intervals (CIs). The numbers in parentheses indicate sample sizes of observations. * indicates that there are significant differences ($p < 0.05$) between conventional tillage (CT) and conservation tillage (NT).

soil depth and N input. Initial soil properties and irrigation have less significant impacts. Regression analyses show that NT effects on SIC stock peak at an ln*RR* of SOC stock value of ∼—0.1, a depth of ∼50—80 cm, and a fertilizer rate of 230 kg N ha^{-1} yr^{-1} (Fig. 8**b–d**).

4. Discussion

4.1. Effect of conservation tillage on the SOC sequestration

Our research findings offered compelling evidence supporting the beneficial effects of conservation tillage (RT) on SOC stock (Fig. 3**b**), a finding that aligns with multiple meta-analyses (Luo et al., 2010b; Du et al., 2017; Bai et al., 2019; Li et al., 2020) and long-term field experiments conducted across regions including the United States (Burke et al., 2019), Europe (Mäder and Berner, 2012), and China (Ma et al., 2024). RT generally led to significantly higher SOC levels relative to NT and CT (0–40 cm, $p < 0.05$) (Fig. 3**b**), supporting our first hypothesis that RT positively influences SOC. Varvel and Wilhelm (2010) reported that, after 20 years of continuous maize cropping, SOC content was the highest under RT. This is likely due to residue input throughout the soil profile by minimal tillage under RT, which enhances soil carbon and nitrogen stock (Zheng et al., 2022). Typically, the biomass returned to the soil after crop harvest exhibits a trend similar to grain yield (Fiorini et al., 2020). RT significantly increased corn grain yield compared to NT and CT, enhancing biomass return (Li et al., 2024a). Additionally, calcium-mediated SOC stabilization mechanisms likely drive the substantial increase in SOC content under RT (Figs. 3**b**, 3**f, and** 3**n and** Fig. 9). Under calcium-rich conditions, microbial SOC decomposition decreases (Rowley et al., 2020), while fungal necromass significantly contributes to recalcitrant SOC in calcareous soil (Yang et al., 2022; Huang et al., 2023). In RT treatment, SOC and SIC exhibit a complementary relationship (Fig. 3**a–c**). We hypothesize that higher autotrophic microorganism abundance in RT facilitates the conversion of SIC to SOC (Fig. 9). Autotrophic microorganisms assimilate carbonate as their primary or sole carbon source. Shao et al. (2022) confirmed through 94 field surveys that microbial residues are essential for the conversion of SIC to SOC.

Our study found no significant difference in SOC stock between NT and CT (Fig. 3**b**). Previous studies have also demonstrated that SOC sequestration potential of NT has been overestimated (Luo et al., 2010c; Du et al., 2017) and that SOC stock in the 0–30 cm layer has not increased compared to CT (Ogle et al., 2005; Zhang et al., 2018). However, numerous other studies have revealed that under NT, crop residues continuously accumulate on the soil surface, providing fresh organic matter and further enhancing SOC accumulation in the topsoil (0–10 cm) over time (Lou et al., 2012; Fiorini et al., 2020; Zheng et al., 2022). Meta-analyses have pointed out that this discrepancy might be ascribed to variations in climatic conditions, site-specific environmental factors, and management practices (Du et al., 2017; Sun et al., 2020; Xiao et al., 2021). On the other hand, while NT effectively inhibits soil carbon release by minimizing soil disturbance, it concurrently restricts the vertical translocation of carbon to deeper soil horizons. Additionally, increased soil compaction under NT can suppress microbial activity and root growth, further diminishes carbon supply to deeper soil layers (Kuhn et al., 2016). Thus, NT's contribution to SOC sequestration, particularly in deeper soil layers, should not to be overestimated (Powlson et al., 2014; Xiao et al., 2020).

Soil disturbance resulting from tillage practices and crop residues removal is widely accepted to be the primary factor leading to SOC loss (Du et al., 2015). Anderson-Teixeira et al. (2009) reported that straw removal (25 %–100 %) led to an average SOC loss of 3–8 Mg ha^{-1} (0–30 cm). Zhang et al. (2018) similarly found that 12-year of continuous residue removal under CT caused a significant SOC decline (9 Mg ha^{-1}, 0–30 cm). However, this study demonstrated that no significant carbon loss after 20 years of CT (Fig. S1). In CT systems, although crop straw is completely removed after harvest, root biomass remaining in the soil can partially compensate for carbon loss. Tillage relocates crop stubble and topsoil carbon to deeper soil layers, enhancing carbon protection. Furthermore, plowing improves soil loosening, promoting deeper root growth and increasing carbon input via root turnover (Luo et al., 2010a).

4.2. NT promotes SIC accumulation in arid croplands

The results of 20-year field experiment demonstrated that NT significantly increased SIC stock in continuous maize cropping systems (0–20 cm, $p < 0.05$, Fig. 3**a**), confirming our second hypothesis that NT had a positive effect on SIC. Typically, alkaline conditions are conducive to carbonate formation (Ferdush and Paul, 2021). NT significantly increased soil pH compared to CT and RT (Fig. 3**e**). Reduced tillage with stover mulching boosts soil respiration, elevating CO_2 concentration (Fig. S2). Higher pH and CO_2 under NT promote energy-favorable environments for $CaCO_3$ and $MgCO_3$ precipitation (Kuzyakov et al., 2006; Wang et al., 2023). Furthermore, sufficient Ca^{2+} and Mg^{2+} supply is another prerequisite for carbonates formation (Ball et al., 2023). NT increases soil Ca and Mg content through higher exogenic Ca inputs (Fig. 3**f–g**). Additionally, NT reduces soil calcium layer exposure and loss, decreases SIC dissolution in the tillage layer and re-precipitation in deeper layers, and lowers soil aeration and CO_2 emissions, supporting SIC formation and preservation (Niu et al., 2023).

Conventionally, soil carbonates formation is an extremely slow process, primarily governed by abiotic mechanisms (Zamanian et al., 2016). However, our findings suggest that biotic factors explain a considerable proportion (32 %, Fig. 5**c and f**) of SIC variation when agronomic practices and key environmental factors are considered. These findings align with recent studies highlighting the importance of biotic factors in SIC accumulation (Virto et al., 2011; Liu et al., 2018). In arid calcareous soils, increasing organic matter input might enhance biological processes influencing pedogenic carbonates formation

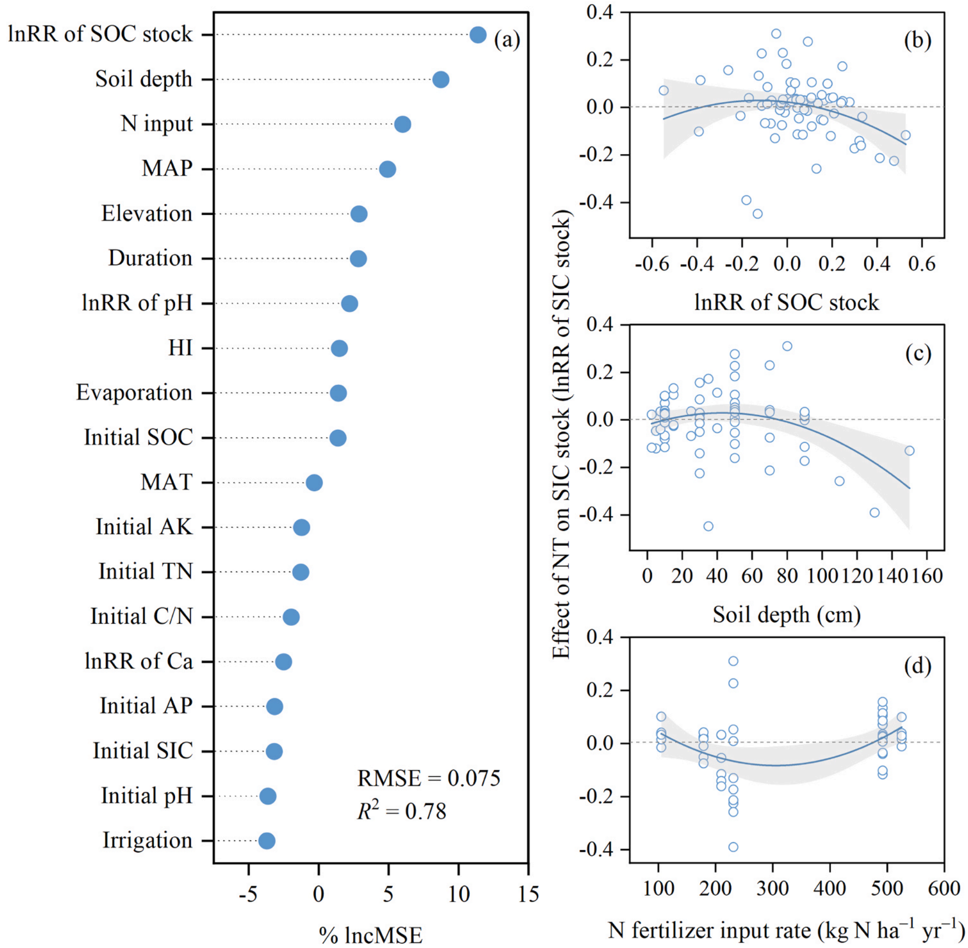

Fig. 8. Relative importance of explanatory variables on NT-induced changes in SIC stock (a). Meta-analytic scatterplots between the effects of NT on SIC stock and the most important predictors of these effects (ln*RR* of SOC stock, soil depth, and N input) (b, c, and d). The explanatory variables include the climatic factors (MAT, mean annual temperature; MAP, mean annual precipitation; HI: MAP/MAT; and evaporation), geographical factors (elevation), edaphic factors (initial SOC, soil organic carbon; SIC, soil inorganic carbon; pH; TN, total nitrogen; C/N, SOC to TN ratio; AP, available phosphorus; AK, available potassium; ln*RR* of SOC stock, pH, and Ca, and soil depth), and management factors (N input, nitrogen fertilizer input rate; duration of no-till; and irrigation).

(Fernández-Ugalde et al., 2011). Straw return provides a large quantity of organic matter for microorganisms (Ven et al., 2020), enhancing microbial abundance and activity. Plant roots and microorganisms respiration increase CO_2 concentration, promoting carbonate precipitate in the presence of Ca^{2+} (Gocke and Kuzyakov, 2011).

MICP plays a crucial role in SIC formation in arid ecosystems (Ennaciri et al., 2022). In arid and semi-arid calcareous soils, previous research has indicated that calcifying bacteria demonstrate high abundance and diversity (Párraga et al., 2004; Bibi, 2018). It was noted that NT enhanced the abundance of *Firmicutes* phylum (Fig. 4Aa). This phylum comprised a significant proportion of urease-producing genera, which was pivotal for the MICP (Rodriguez-Navarro et al., 2012; Li et al., 2024b). In arid calcareous soils characterized by low organic matter content, microorganisms may exhibit a preference for deriving energy from chemical synthesis or alternative metabolic pathways conducive to carbonate precipitation (Bindschedler et al., 2016). Ball et al. (2023) provided evidence that organic matter may not always be the main factor influencing microbial biomass size, while calcium could significantly contribute to bacterial biomass accumulation and SIC pool formation in dryland calcareous soils. Higher bacterial and fungal species were observed under NT, but it had lower SOC than RT (Fig. 3**b**), suggesting that microbial communities may not be limited by the availability of organic substrates.

Soil enzyme activity serves as a key indicator of soil quality, nutrient status, and microbial ecosystem health (Wu et al., 2023). In MICP systems, UA functions as a crucial metric for evaluating fixation efficiency (Li et al., 2024b). The reduction in UA under NT, coupled with its inverse relationship with SIC, likely attributable to UA consumption during the MICP process (Fig. 3**h and l**). SIC (or $CaCO_3$) content analysis revealed a pronounced calcium fixation effect in NT, necessitating a considerable amount of CO_3^{2-}, which generally results in significant urease consumption. Therefore, based on the aforementioned results, our second hypothesis is confirmed: NT increases SIC through the MICP process (Fig. 3**a and h,** Fig. 4**A,** Fig. 5 **and** Fig. 9). To our knowledge, this is the first systematic exploration of the contribution of the MICP to SIC formation in arid croplands within conservation tillage.

Notably, while our long-term field experiment demonstrated significant SIC gains under NT, the meta-analysis indicates that such effects are context-dependent, thereby underscoring the importance of adapting tillage practices to specific soil and climate conditions (Fig. 7 **and** Table S7). Arid regions benefit the most from NT in terms of enhancing SIC storage (3.27 %) (Fig. 7). Approximately 40 % of the global terrestrial area is categorized as arid and semi-arid regions (Reynolds et al., 2007), where SIC constitutes 90 % of total carbon stock (Filippi et al., 2020). These findings highlight the importance of conservation tillage for carbon sequestration in drylands, particularly through the often-overlooked SIC pool. In addition, according to the results of the field experiments, SIC is actively modulated by conservation tillage via biotic and abiotic mechanisms. The demonstrated role of microbial communities in carbonate precipitation suggests that microbial-driven

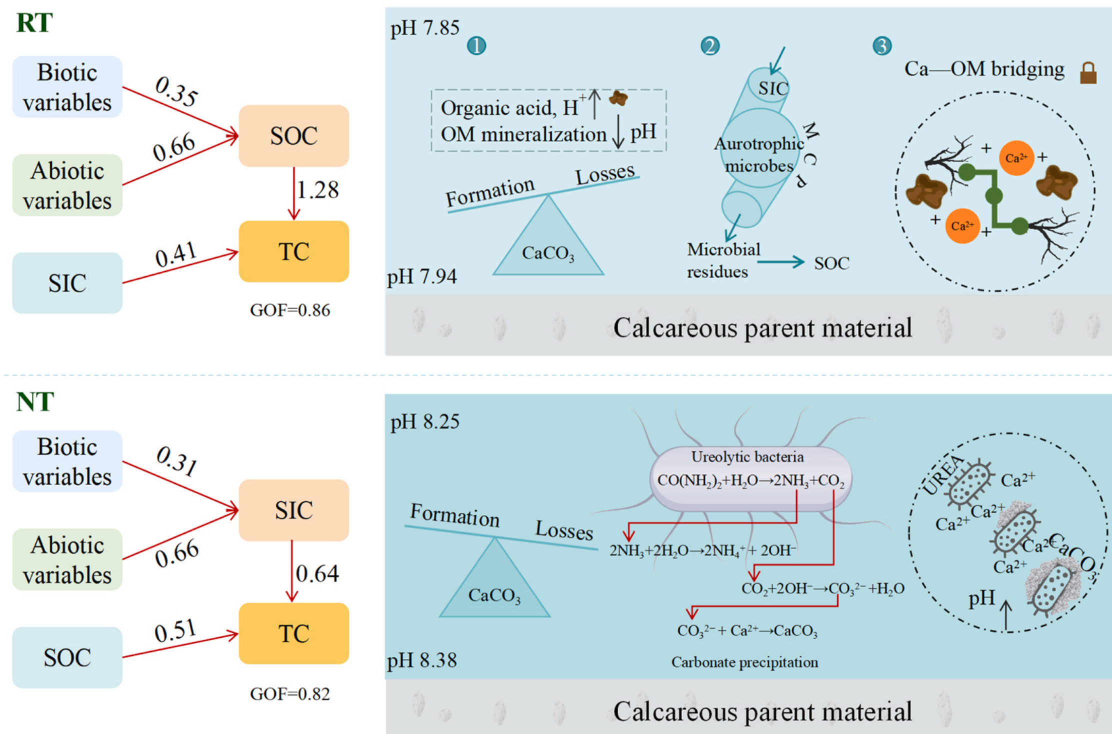

Fig. 9. The PLS-PM model on the left panel shows the contributions of soil biotic and abiotic factors, SOC, and SIC to TC under RT and NT. Numbers near arrows are standardized path coefficients. The right panel includes a conceptual diagram of SOC increase under RT and a schematic of MICP promoting SIC accumulation under NT. The depth of the background color on the right panel reflects the levels of soil moisture content. Abbreviations: RT, reduced tillage; NT, no-tillage; SOC, soil organic carbon; SIC, soil inorganic carbon; TC, total carbon content; MCP, microbial carbon pump; OM, organic matter.

SIC sequestration could be leveraged as a novel carbon management strategy in dryland agriculture.

4.3. Implications, limitations, and prospects

In this study, we quantitatively evaluated the effect of conservation tillage on soil carbon pools, with a particular focus on SIC stock, by integrating a 20-year field experiment, meta-analysis, and a multidisciplinary approach. Our long-term experiment provides vital evidence that NT (0–20 cm) and RT (0–40 cm) combined with straw mulching significantly enhance topsoil carbon sequestration, while subsoil carbon sequestration remains limited. The proliferation of calcifying bacteria (such as *Bacillus*, belonging to *Firmicutes*) and the depletion of urease suggest that MICP might be a significant contributing factor to the formation of SIC under NT. The meta-analysis reveals that arid regions benefit the most from NT in terms of enhancing SIC stock (3.27 %). These findings highlight the importance of conservation tillage for carbon sequestration in drylands, particularly through the often-overlooked SIC pool. Microbial-driven SIC sequestration could be exploited as a potential strategy and employed as a novel carbon management approach in arid agriculture.

Limitations, however, still exist in this study: (1) Geographical constraints: This field experiment was conducted in a single arid area in northern China, with specific climate and soil conditions may limit the generalizability of the findings. (2) Limited sample size and missing data: This meta-analysis encompasses 7 studies and 76 observations. Moreover, some previous studies only reported SIC content without providing soil bulk density and SD values, adding uncertainty. (3) Neglected synergies: This study focused on comparing NT and CT, but did not account for the interactive influences of management variables such as irrigation and organic amendment might mediate the response of SIC. (4) Insufficient microbial evidence: Although the potential involvement of ureolytic bacteria in SIC formation was observed, direct evidence such as isotope tracing to confirm the causal relationship is lacking. (5) Temporal heterogeneity of microbial data: Single-time-point sampling might not accurately reflect the long-term effects of tillage on the microbial community structure and function. (6) Absence of economic and yield trade-off analysis: The trade-off between NT, carbon sequestration, crop yields, and farmers' adoption willingness was not quantified, potentially undermining the practical applications of the research findings.

Future research should focus on: (1) Constructing cross-climate-zone and multi-factor field experiments to systematically reveal the biogeochemical mechanisms of SIC formation. (2) Employing stratified sampling and isotope techniques to quantify fungal-bacterial contributions and regulation mechanisms to carbonate precipitation. (3) Establishing a multi-objective optimization framework integrating economic assessment models to elucidate the coordinated regulation pathways and socio-economic benefits between carbon sequestration potential and agricultural productivity. (4) Coupling eddy covariance observations with process models for concurrent monitoring of soil carbon dynamics and greenhouse gas fluxes, providing multi-scale and high-precision data support for land carbon budget assessment. The potential of SIC sequestration through NT offers an untapped opportunity to enhance global soil carbon stock and mitigate CO_2 emissions, particularly in dryland systems.

5. Conclusion

This study found that NT is a promising practice for sequestering SIC in dryland agriculture, supported by a 20-year field experiment and a meta-analysis across China. The field experiment provides vital evidence that NT and RT, combined with straw mulching, significantly enhance the sequestration of SIC (10.67 %, 0–20 cm) and SOC (25.09 %, 0–40 cm), respectively, while subsoil carbon sequestration remains limited. Increased calcifying bacteria (e.g., *Bacillus*) and reduced urease activity suggest that MICP may contribute significantly to SIC formation under NT. Our meta-analysis further shows that NT-induced changes in SIC stock vary remarkably among different moderators (e.g., climate, soil, and management conditions). Specifically, optimal SIC stock is achieved in arid areas (MAP was < 400 mm), N input level < 150 kg N ha^{-1} yr^{-1}, and soil profiles with an average depth of 1 m. These findings highlight the importance of conservation tillage for carbon sequestration in drylands, particularly through the often-overlooked SIC pool. The study highlights that SIC, despite being historically considered stable, is actively modulated by conservation tillage via biotic and abiotic mechanisms. Microbial-driven SIC sequestration, namely MICP, could be leveraged as a novel carbon management strategy in dryland agriculture.

CRediT authorship contribution statement

Weiting Ding: Writing – original draft, Methodology, Data curation, Software, Investigation, Writing – review & editing, Visualization. **Liangjie Sun:** Writing – review & editing. **Zhidong Qi:** Writing – review & editing. **Shengping Li:** Writing – review & editing. **Vilim Filipović:** Writing – review & editing. **Xueping Wu:** Resources, Writing – review & editing. **Hailong He:** Supervision, Methodology, Formal analysis, Writing – review & editing, Project administration, Funding acquisition, Resources, Investigation, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.agee.2025.109845.

Data availability

Data will be made available on request.

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