Received: 17 October 2023 | Revised: 6 February 2024 | Accepted: 5 March 2024

DOI: 10.1111/gcb.17247

RESEARCH ARTICLE

Global Change Biology WILEY

Depth-dependent responses of soil organic carbon under nitrogen deposition

Yuanliu Hu[1,2,3,4] | Qi Deng[1,2,3] | Thomas Kätterer[5] | Jørgen Eivind Olesen[4,6] | Samantha C. Ying[7] | Raúl Ochoa-Hueso[8,9] | Carsten W. Mueller[10,11] | Michael N. Weintraub[12] | Ji Chen[4,13,14]

[1]Key Laboratory of Vegetation Restoration and Management of Degraded Ecosystems/Guangdong Provincial Key Laboratory of Applied Botany, South China Botanical Garden, Chinese Academy of Sciences, Guangzhou, China

[2]South China National Botanical Garden, Guangzhou, China

[3]University of Chinese Academy of Sciences, Beijing, China

[4]Department of Agroecology, Aarhus University, Tjele, Denmark

[5]Department of Ecology, Swedish University of Agricultural Sciences, Uppsala, Sweden

[6]Aarhus University Centre for Circular Bioeconomy, Aarhus University, Tjele, Denmark

[7]Department of Environmental Sciences, University of California, Riverside, California, USA

[8]Department of Biology, IVAGRO, University of Cádiz, Campus de Excelencia Internacional Agroalimentario (CeiA3), Cádiz, Spain

[9]Department of Terrestrial Ecology, Netherlands Institute of Ecology (NIOO-KNAW), Wageningen, The Netherlands

[10]Institute of Ecology, Chair of Soil Science, Technische Universitaet Berlin, Berlin, Germany

[11]Department of Geosciences and Natural Resource Management, University of Copenhagen, Copenhagen, Denmark

[12]Department of Environmental Sciences, University of Toledo, Toledo, Ohio, USA

[13]State Key Laboratory of Loess and Quaternary Geology, Institute of Earth Environment, Chinese Academy of Sciences, Xi'an, China

[14]Institute of Global Environmental Change, Department of Earth and Environmental Science, School of Human Settlements and Civil Engineering, Xi'an Jiaotong University, Xi'an, Shaanxi Province, China

Correspondence

Ji Chen, State Key Laboratory of Loess and Quaternary Geology, Institute of Earth Environment, Chinese Academy of Sciences, Xi'an 710001, China.
Email: ji.chen@agro.au.dk

Qi Deng, Key Laboratory of Vegetation Restoration and Management of Degraded Ecosystems, South China Botanical Garden, Chinese Academy of Sciences, Guangzhou, Guangdong 510650, China.
Email: dengqi@scbg.ac.cn

Funding information

Shaanxi Province Natural Science Foundation for Distinguished Young Scholar, Grant/Award Number: 2024JC-JCQN-32; EU H2020 Marie Skłodowska-Curie Actions, Grant/Award Number: 839806; Aarhus University iClimate; National Natural Science Foundation of China, Grant/Award Number: 32371675

Abstract

Emerging evidence points out that the responses of soil organic carbon (SOC) to nitrogen (N) addition differ along the soil profile, highlighting the importance of synthesizing results from different soil layers. Here, using a global meta-analysis, we found that N addition significantly enhanced topsoil (0–30 cm) SOC by 3.7% (±1.4%) in forests and grasslands. In contrast, SOC in the subsoil (30–100 cm) initially increased with N addition but decreased over time. The model selection analysis revealed that experimental duration and vegetation type are among the most important predictors across a wide range of climatic, environmental, and edaphic variables. The contrasting responses of SOC to N addition indicate the importance of considering deep soil layers, particularly for long-term continuous N deposition. Finally, the lack of depth-dependent SOC responses to N addition in experimental and modeling frameworks has likely resulted in the overestimation of changes in SOC storage under enhanced N deposition.

and 42361144886; Nordic Committee of Agriculture and Food Research; "Young Top-notch Talent" in Pearl River talent plan of Guangdong Province, Grant/Award Number: 2019QN01L763; Danish Independent Research Foundation, Grant/Award Number: 1127-00015B; Aarhus University Research Foundation, Grant/Award Number: AUFF-E-2019-7-1

KEYWORDS
carbon cycle–climate feedbacks, carbon losses, nitrogen addition duration, soil carbon sequestration, soil profiles

1 | INTRODUCTION

Excessive nitrogen (N) deposition from human activities has continuously increased over the past decades (Fowler et al., 2013; Gruber & Galloway, 2008; IPCC, 2021), with substantial consequences on soil organic carbon (SOC) dynamics (Liu & Greaver, 2010; Pregitzer et al., 2008; Xu et al., 2021). Despite the rapid increase in the number of studies reporting the effects of N addition on SOC, no consensus has been reached on the response along the soil profile. For example, Bowden et al. (2019) found that N addition significantly increased SOC in the topsoil in boreal forests (0–30 cm), whereas N addition significantly decreased SOC in temperate grasslands when the whole soil profile (0–100 cm) was considered (Li et al., 2014; Poeplau et al., 2018). Thus, a better understanding of N deposition impacts on SOC at different depths is urgently needed to accurately predict carbon (C) cycle–climate feedbacks.

The topsoil typically receives the majority of plant-derived C inputs, and it thus has the highest SOC and microbial biomass content (Cochran et al., 1989). It is proposed that topsoil SOC displays a relatively high decomposition rate and a rapid exchange of C with the atmosphere (Hartemink et al., 2020; Rumpel & Kögel-Knabner, 2011). In contrast, SOC buried in the subsoil is generally assumed to be more persistent, with a longer turnover time (Hicks Pries et al., 2017). Based on this assumption, the Intergovernmental Panel on Climate Change has primarily emphasized the 0–30 cm soil layer in the greenhouse gas inventory (IPCC, 2006, 2021). Indeed, this sampling depth has been adopted by many experiments and models when investigating SOC dynamics under various conditions. However, the assumption of the persistence of SOC in the subsoil has recently been challenged (Chen et al., 2023; Li et al., 2022; Luo et al., 2019). Several studies have demonstrated the depth-dependent responses of SOC to nutrient addition (Fierer et al., 2003), climate warming (Wang et al., 2022), and land use change (Chen et al., 2022; Mobley et al., 2015). Furthermore, long-term regional and global studies have revealed different SOC accumulation rates and driving forces when comparing topsoil to subsoil (Balesdent et al., 2018; He et al., 2022; Li et al., 2022; Sun et al., 2023). These results imply that a lack of exploration for subsoil SOC may hamper our ability to assess the potential of soil C sequestration under future climate change scenarios.

Despite the growing recognition of the importance in subsoil SOC, only a limited number of studies have investigated the effects of N addition on SOC in subsoil across the globe. Several recent meta-analyses indicated that the effects of N addition on SOC were highly dependent on soil and microbial properties (Ni et al., 2022; Niu et al., 2021), with both types of variables showing substantial variations along soil profiles (Eilers et al., 2012; Federle et al., 1986; Mathieu et al., 2015). Other studies further suggested that the response of SOC to N addition may not always be aligned in the topsoil and subsoil, and in some cases, responses may even be contradictory. For example, Canary et al. (2000) and Li et al. (2014) found that N addition significantly increased SOC in the topsoil, whereas SOC decreased in the subsoil, leading to neutral or even negative responses of SOC to N addition when the whole soil profile was considered. These results demonstrate that neglecting the effects of N addition on subsoil SOC may lead to biased interpretations, or even misleading conclusions about the potential consequences of increased N deposition on SOC sequestration.

In this study, we conducted a comprehensive meta-analysis to examine the effects of N addition on SOC in both topsoil and subsoil. We compiled data from 177 N addition experiments conducted worldwide, regarding 0–30 cm as topsoil and 30–100 cm as subsoil according to the recommendations from IPCC (IPCC, 2021; Minasny et al., 2017). We also recorded information on environmental factors (mean annual temperature and precipitation) and experimental protocols (N addition form, rate, frequency, and duration) that may be relevant in determining the responses of SOC to N addition in both topsoil and subsoil. Given that N addition effects on SOC may be cumulative, we also distinguished the responses of SOC to N addition between short-term (<5 years) and long-term (≥5 years) studies. Overall, we addressed two specific questions in this study: (1) How did SOC in topsoil and subsoil respond to N addition at the global scale? (2) What were the important modulators of the responses of SOC to N addition in topsoil and subsoil?

2 | MATERIALS AND METHODS

2.1 | Data collection

We searched for peer-reviewed articles published before December 2023 from Web of Science (http://apps.webofknowledge.com/), Google Scholar (http://scholar.google.com/), and China National

Knowledge Infrastructure (http://www.cnki.net/). In order to find articles reporting the effects of N addition on SOC from different soil depths, the used keywords or combinations of keywords were: (a) "nitrogen addition" or "nitrogen enrichment" or "nitrogen fertilizer" or "nitrogen elevated" or "nitrogen deposition" and (b) "soil organic carbon" or "soil organic matter" and (c) "soil depth" or "soil layer" and (d) "terrestrial" or "soil" or "land."

To explore the N-induced changes in SOC across different soil layers, observations were removed according to the following criteria (Figure S1): (1) Measurements from croplands and other managed ecosystems, because disturbance through agricultural practices may alter the response of SOC to N addition along soil depth; (2) studies did not clearly specify soil sampling depth; (3) studies where other nutrients were added (e.g., P, K, Ca, compost, or slurry additions) or other treatments were imposed (e.g., CO_2, warming, or precipitation change); (4) missing details on N addition methods (fertilization rate, frequency, form, and duration); (5) N addition studies without directly applying N to the soil. Following the common soil sampling method, we did not consider the response of SOC to N addition in the litter layer in this study. Finally, we synthesized data (overview is provided in Data S1) on the responses of SOC to N addition from 177 published articles (Figure S2).

We recorded SOC concentration (percentage of SOC per soil mass) in the corresponding treatments with and without N addition of each study involved in this dataset, respectively. Overall, 8.5% of the observations in this dataset reported SOC storage representing mass C per volume, which could not be converted to SOC concentration on a mass basis because of unreported soil bulk density. To identify the influence of changes in soil bulk density on the responses of SOC to long-term N addition (Chen et al., 2019), we also recorded soil bulk density with and without N addition. The soil sampling depth in this dataset ranged from 5 to 100 cm, with an average depth of 25 cm. To be consistent with the IPCC's soil layer classification method, data from different soil layers were categorized into 0–30 cm (topsoil) and 30–100 cm (subsoil).

Furthermore, to explore the underlying mechanisms related to the responses of SOC to N addition, we also recorded vegetation type (grassland and forest), mean annual temperature (MAT), mean annual precipitation (MAP), longitude, latitude, background N deposition rate (BND), and N addition method (experimental duration, added N rate, the ratio of BND to added N rate and added N frequency) for each experimental site. The experimental duration of N addition studies included in our analysis ranged from 1 to 58 years. The cutoff of 5 years for short-term versus Longer term studies was chosen based on a large survey of long-term research in ecology and evolution, indicating that experiments lasting less than 5 years often quantified only the immediate and transient effects of perturbation (Kuebbing et al., 2018). Most studies added N in each year along the experimental duration, except for the study conducted by Canary et al. (2000), in which N was added intermittently every few years. In this dataset, the N addition rate ranged from 0.5 to 64 $g\,m^{-2}\,year^{-1}$.

As N application forms, we considered inorganic N (NH_4, NO_3, or NH_4NO_3), organic N (urea), and the mixture of inorganic and organic N (NH_4, NO_3, and urea). Besides, we obtained unreported MAT and MAP from the WorldClim database (Fick & Hijmans, 2017), which resulted in the inclusion of a broad range of MAT (−4.6 to 26°C) and MAP (69–4300 mm $year^{-1}$) in our dataset. Unreported background N deposition rate was collected from the Global N deposition database (ORNL DAAC, 2017). The missing standard deviation was imputed using Rubin and Schenker's resampling approach from studies with similar means (Rubin & Schenker, 1991). When results were presented graphically, we used GetData Graph Digitizer v.2.24 (http://getdata-graph-digitizer.com/) to digitize the data.

2.2 | Data analysis

We used the natural log response ratio to assess the effect of N addition on each variable. Each individual observation was weighted by the inverse of the mixed model variance, ensuring that meta-analysis data statistics have simpler standard sampling distributions while avoiding the possibility of unequal variance effects on statistical results (Hedges et al., 1999).

The natural log response ratio (ln *R*) was calculated as:

$$\ln R = \ln\left(X_N / X_C\right)$$

where X_C and X_N are the mean values of the studied variables in the control and N addition treatments, respectively.

The variance (*V*) of the logarithmic effect size was calculated as:

$$V = S_N^2 / \left(n_N X_N^2\right) + S_C^2 / \left(n_C X_C^2\right)$$

where S_C and S_N are the standard deviation in the control and N addition treatments, respectively, and n_C and n_N are the number of replicates in the control and N addition treatments, respectively.

A mixed-effects model was used to determine the effect of N addition on the selected variables through "rma.mv" function from the R package "metafor" (Viechtbauer, 2010). We considered the "Publication" as random effect in the mixed-effects model, which would ensure the independence of each observation (Chen et al., 2018). The effects of N addition were considered significant if the 95% confidence intervals did not overlap with zero. The results were reported as mass percentage change with N addition to ease interpretation.

We carried out a model selection analysis to determine which were the main environmental and procedural variables driving the response of SOC to N across depths. Model selection was based on the corrected Akaike information criterion (AIC corrected for small samples). A predictor including models with large Akaike weights was assigned with a high importance value. Thus, the cumulative Akaike weight of the models containing the specific predictor was used to indicate the relative importance value for this predictor. A cutoff value of 0.8 was set to identify the critical and non-critical predictors (Terrer et al., 2016; van Groenigen et al., 2011). This analysis was conducted through the "glmulti" package in R (Calcagno & De Mazancourt, 2010).

3 | RESULTS

Averaged across all studies, N addition significantly increased SOC in the topsoil by 3.7% (95% CIs, 2.2–5.1%, $p<.001$). However, N addition did not affect subsoil SOC (Figure 1a). The depth-dependent responses of SOC to N addition were consistent regardless of vegetation type (forest and grassland; Figure 1b). Based on the corrected Akaike information criterion, the responses of SOC to N addition were best predicted by soil layer and experimental duration (Figure S3). The results from mixed linear regression model showed a significant interactive effect of soil layer and experimental duration on the responses of SOC to N addition (Table S1).

In the topsoil, experimental duration and vegetation type were the important predictors of the response of SOC to N addition (Figure 2a). Short-term (<5 years) N addition did not change topsoil SOC (−0.6% to 3.3%, $p=.18$; Figure 3a), whereas long-term (≥5 years) N addition significantly increased topsoil SOC by 5.6% (3.7%–7.5%, $p<.001$). The time-dependent responses of topsoil SOC to N addition were consistent regardless of vegetation type (Figure S4).

Regarding the subsoil, the responses of SOC to N addition were best predicted by experimental duration (Figure 2b). Short-term (<5 years) N addition significantly increased SOC by 4.3% (1.1%–7.6%, $p<.05$; Figure 3b), whereas long-term (≥5 years) N addition decreased SOC by −3.9% (−1.4% to −6.3%, $p<.01$). The significant decreases in subsoil SOC under long-term N addition were only observed in forests; however, there was a lack of significant interactive effects of vegetation type and experimental duration on the responses of subsoil SOC to N addition (Table S1; Figure S4). Additionally, there was a positive relationship between the responses of topsoil and subsoil SOC to N addition in the short-term N addition studies, whereas no significant relationship was found between the responses of topsoil and subsoil SOC to N addition in the long-term studies (Figure 3c).

4 | DISCUSSION

4.1 | Nitrogen addition increased topsoil organic carbon over time

We found an increase in topsoil SOC under N addition (Figure 1a). Increased N availability under N addition likely stimulates plant growth and litter inputs (Greaver et al., 2016; Reay et al., 2008). The additional litter inputs can be partly incorporated into soils, contributing to enhanced topsoil SOC, as demonstrated in several N addition studies (Canary et al., 2000; Hyvönen et al., 2008; Liao et al., 2023). Nitrogen addition may also enhance topsoil SOC by repressing soil C losses (Janssens et al., 2010; Lu et al., 2021). For example, N addition often causes soil acidification or an increase in osmotic stress, potentially suppressing microbial growth and activity, and consequently reducing C losses from microbial respiration (Treseder, 2008; Zhou et al., 2014). The N-induced plant growth may also lead to increased uptake of base cations, thereby accelerating soil acidification and inhibiting microbial activity (Duan et al., 2004). Similarly, N addition may inhibit the phenol oxidase activity of white-rod basidiomycetous fungi in the floor of hardwood forests, leading to a greater accumulation of under-decomposed litter (Chen et al., 2018; Waldrop et al., 2004). In addition, the potential reduction in soil C losses under N addition may also be related to the enhanced stabilization of SOC compounds that are typically subject to physical and chemical shielding (Figure S5; Jilling et al., 2021; Treseder, 2004).

Our study indicated that the increased topsoil SOC only manifested with long-term N addition (Figure 3a), which was supported by recent meta-analyses (Lu et al., 2023; Xu et al., 2021). First, this

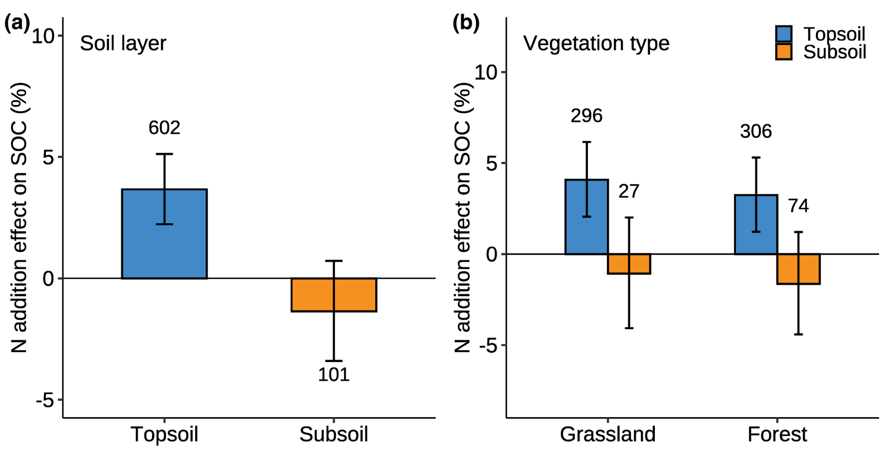

FIGURE 1 Nitrogen (N) addition only increased soil organic carbon (SOC) in the topsoil but not in the subsoil. (a) Meta-analysis of the effects of N addition on SOC when grouped by soil layers. (b) Meta-analysis of the effects of N addition on SOC when grouped by soil layers and vegetation types. Error bars represented bootstrap 95% confidence intervals (CIs). The effects of N addition were considered significant if the 95% CIs did not overlap with zero; the numbers above or below the error bars indicated sample sizes.

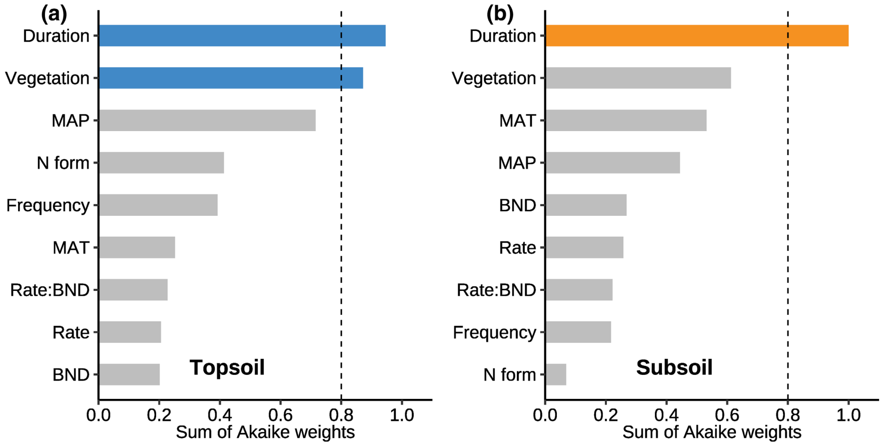

FIGURE 2 Experimental duration was the most important predictor in the responses of soil organic carbon (SOC) to nitrogen (N) addition in both topsoil and subsoil. (a) Model selection analysis identified that experimental duration and vegetation type were the important predictors of the response of SOC to N addition in the topsoil; the dashed line indicated the cutoff to distinguish important predictors that exceeded the 0.8 sum-of-Akaike-weights threshold. (b) Model selection analysis identified that experimental duration was the important predictor of the response of SOC to N addition in the subsoil. BND, background N deposition rates; duration, experimental duration; frequency, N addition frequency; MAP, mean annual precipitation; MAT, mean annual temperature; rate, N addition rate.

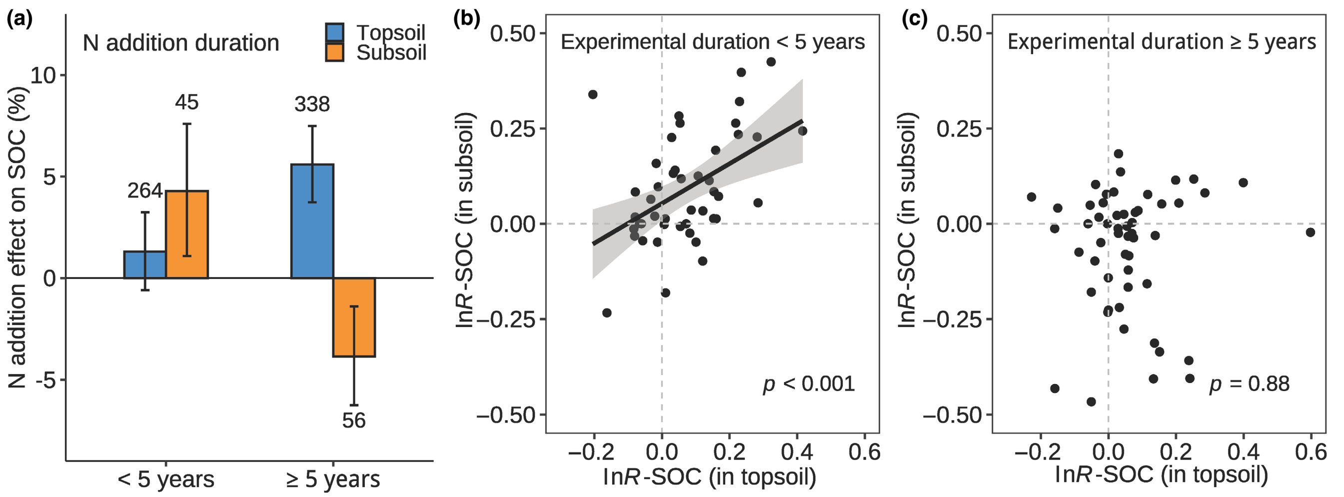

FIGURE 3 The effects of nitrogen (N) addition on soil organic carbon (SOC) depend on experimental duration. (a) In the topsoil, short-term N addition (<5 years) had nonsignificant effect on SOC, whereas long-term N addition (≥5 years) significantly increased SOC. In the subsoil, short-term N addition significantly increased SOC, whereas long-term N addition decreased SOC; error bars represented bootstrap 95% confidence intervals (CIs); the effects of N addition were considered significant if the 95% CIs did not overlap with zero; the numbers above or below the error bars indicated sample sizes. (b) In the short-term N addition studies, there was a significant positive relationship between the responses of SOC (ln*R*-SOC) in the topsoil and in the subsoil [coefficient of determination (r^2) = .23, $p < .001$]; the light gray area indicated the confidence interval around the regression line. (c) In the long-term studies, the relationship between the responses of SOC in the topsoil and the subsoil was not significant ($r^2 = .01$, $p = .88$).

result may suggest that it takes time for SOC accumulation to be observable. For example, Xu et al. (2021) suggested that significant increases in plant C inputs and topsoil SOC were typically observed in N addition studies lasting more than 3 years. Some studies also indicated that the plant C inputs at the initial stage of N addition may not cause as rapid SOC storage as previously assumed (Lu et al., 2023; Vourlitis & Hentz, 2016). However, a recent study based on global grasslands showed that SOC increased significantly under short-term N addition (<5 years), probably due to the inclusion of both topsoil and subsoil SOC in the database (Liu et al., 2023). Alternatively, the time-dependent response of topsoil SOC to N addition may be linked to the balance between plant C inputs and soil C losses (Janssens et al., 2010; Pregitzer et al., 2008; Zhou et al., 2014). For example, in the initial stage of N addition, alleviation of microbial N limitation may be associated with stimulated microbial respiration and C losses, which may offset the positive effects of plant C inputs on topsoil SOC, especially in N-poor ecosystems (Figure S6; Ågren et al., 2001; Lu et al., 2011). In the later stage of N addition, microbial activities may be repressed by N-induced soil acidification or increased osmotic stress (Zhou et al., 2014). Suppressed microbial activities and associated C losses have indeed been found to be significant under long-term N addition (Ning et al., 2021; Treseder, 2008).

4.2 | Long-term N addition decreased subsoil organic carbon

In contrast to the N-induced increases in topsoil SOC, we found that subsoil SOC significantly increased in short-term N addition studies (<5 years) but decreased in the long-term studies (≥5 years; Figure 3a). One possible reason for the increased subsoil SOC under short-term N addition may be attributed to the increased C leaching from aboveground litter decomposition (Li et al., 2021; Sinsabaugh et al., 2004). For example, the increased C leaching was likely associated with accelerated decomposition of N-induced additional litter inputs in the topsoil under short-term N addition (Wang et al., 2019). In the early stages of N addition, aboveground C input is typically associated with less efficient decomposition and lower C stabilization, which may stimulate the leaching of dissolved C downward to the subsoil, resulting in increased subsoil SOC (Pregitzer et al., 2004). In contrast to the topsoil, increased N availability within short-term N addition may not stimulate subsoil SOC mineralization and associated C losses due to the lack of fresh C and energy supply in this soil layer (Figure S6; Wang et al., 2014). Additionally, the positive effects of increased plant C inputs on subsoil SOC in the short term may also be related to the inherently lower SOC in this layer (Balesdent et al., 2018).

Why did long-term N addition significantly decrease subsoil SOC? This unexpected result can be partly attributed to the potential reduction in the downward movement of C along the soil profile in the later stage of N addition. For example, long-term N addition may repress litter decomposition, which likely results in accumulation of fresh aboveground litter in the humus layer instead of being transported into deeper soil layers (Franklin et al., 2003; Treseder, 2004). A 10-year N addition study conducted in northern American broadleaved forests revealed that the accumulation of SOC derived from leaf litter was observed only in the topsoil (Pregitzer et al., 2008). Besides, in the long term, the alleviated microbial N limitation with N addition is generally associated with decreased oxidase activities and increased formation of stable SOC in the topsoil (Chen et al., 2018; Lu et al., 2021), thereby reducing the C leaching along the soil profile (Kaiser & Kalbitz, 2012). This explanation is supported by both increased particulate and mineral-associated organic carbon in the topsoil only observed in long-term N addition studies (Figure S5; Qi et al., 2023). Similarly, bioturbation contributes to mixing SOC from the topsoil to the subsoil (Wilkinson et al., 2009), whereas this process may be repressed in the later stage of N addition due to possible soil acidification or toxicity effects on soil organisms (Jansone et al., 2020).

There was a lack of a significant positive correlation between the responses of SOC in the topsoil and the subsoil to long-term (as opposed to short-term) N addition (Figure 3c). This is likely attributed to contrasting mechanisms driving the responses of SOC to N addition in shallow versus deeper layers. In contrast to the topsoil, the subsoil may become more susceptible to losing C through microbial-mediated SOC decomposition in the later stage of N addition (Fontaine et al., 2007; Karhu et al., 2016). For example, long-term N addition may stimulate the decomposition of mineral-associated SOC in the subsoil as indicated by increases in microbial biomass (Figure S6), while this process is generally suppressed by insufficient energy supply within this soil layer compared to the topsoil (Henneron et al., 2022; Jilling et al., 2021). This explanation could be attributed to more energy investment from plants in the form of root exudates for resource acquisition, such as phosphorus and water, as the demand of plants for these resources gradually increases with the continuous N addition (Li et al., 2016; Peñuelas et al., 2013). For example, N addition significantly increased fine root biomass in the subsoil after 5 years (Yan et al., 2017), but decreased it in the first year of N addition (Zhu et al., 2021). Similarly, more C investment from plants in root growth for resource uptake in the later stage of N addition may induce priming effects of SOC decomposition. This N-induced root exploration for resource acquisition may also increase the accessibility of subsoil SOC that is often persistent due to physical separation from decomposers (Henneron et al., 2022; Salomé et al., 2010). The C losses from the decomposition of recalcitrant SOC may outweigh the potential increases in root C inputs (Mobley et al., 2015; Shahzad et al., 2019). Additionally, a recent 20-year study even found that N addition could accelerate the decomposition of ancient SOC in the subsoil by mitigating the oxygen limitation of SOC mineralization through the accumulation of nitrate (Qin et al., 2023), as nitrate is an alternative electron accepter for microbial respiration (Sierra & Renault, 1998).

4.3 | Implications

Understanding the depth-dependent responses of SOC to N addition can help us reconcile the apparently conflicting results often reported from individual studies (Deng et al., 2020; Xu et al., 2021; Zheng et al., 2022). It is widely recognized that responses of SOC to N addition are often cumulative, which takes time to be statistically significant. This meta-analysis study underlines the importance of considering soil depth together with time, as we found the contrasting responses of topsoil and subsoil SOC to short- and long-term N additions. Our finds thus imply that both topsoil and subsoil layers need to be evaluated for a more realistic understanding of the responses of SOC to N addition, especially in the context of long-term field experiments. Importantly, the sharp decline in subsoil SOC in response to long-term N addition presented here may challenge the previously assumed role of soils as a global SOC sink under increased N deposition (Janssens et al., 2010; Lu et al., 2011; Nave et al., 2009; Xu et al., 2021). However, most current N addition studies have primarily focused on the responses of SOC in the topsoil (typically in 0–20 cm or 0–30 cm), which may lead to biased conclusions regarding SOC sequestration. In addition, to be consistent with the IPCC's soil layer classification method, some observations of SOC in the soil layers of 20–40 cm or 20–60 cm were considered as subsoil SOC in this study (Figure S7). Considering the crucial role of subsoils for SOC sequestration (Jobbágy & Jackson, 2000; Rumpel & Kögel-Knabner, 2011; Shi et al., 2020), our study highlights the necessity to distinguish the responses of SOC to increased N deposition across different soil layers, rather than typically extrapolating the responses of SOC to subsoil based on its responses in the topsoil. Overall, our findings suggest the overestimation of soil C sequestration under N addition if without considering the subsoil C dynamics, highlighting the necessity for explicit incorporation of depth-dependent responses of SOC into current global C cycle models.

AUTHOR CONTRIBUTIONS

Yuanliu Hu: Data curation; formal analysis; writing – original draft. **Qi Deng:** Conceptualization; data curation; writing – original draft. **Thomas Kätterer:** Data curation; writing – review and editing. **Jørgen Eivind Olesen:** Data curation; writing – review and editing. **Samantha C. Ying:** Data curation; writing – review and editing. **Raúl Ochoa-Hueso:** Data curation; writing – review and editing. **Carsten W. Mueller:** Data curation; writing – review and editing. **Michael N. Weintraub:** Data curation; writing – review and editing. **Ji Chen:** Conceptualization; data curation; writing – original draft.

ACKNOWLEDGMENTS

This study is granted by the National Natural Science Foundation of China (42361144886, 32371675), Shaanxi Province Natural Science Foundation for Distinguished Young Scholar (2024JC-JCQN-32), EU H2020 Marie Skłodowska-Curie Actions (no. 839806), Aarhus University Research Foundation (AUFF-E-2019-7-1), Danish Independent Research Foundation (1127-00015B), "Young Top-notch Talent" in Pearl River talent plan of Guangdong Province (2019QN01L763), Nordic Committee of Agriculture and Food Research, and Aarhus University iClimate.

CONFLICT OF INTEREST STATEMENT

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 article.

DATA AVAILABILITY STATEMENT

The data associated with this paper are available from figshare (https://doi.org/10.6084/m9.figshare.25358857.v1).

ORCID

Yuanliu Hu https://orcid.org/0000-0003-4137-225X
Qi Deng https://orcid.org/0000-0001-8208-9763
Thomas Kätterer https://orcid.org/0000-0002-1751-007X
Jørgen Eivind Olesen https://orcid.org/0000-0002-6639-1273
Samantha C. Ying https://orcid.org/0000-0002-1247-2529
Raúl Ochoa-Hueso https://orcid.org/0000-0002-1839-6926
Carsten W. Mueller https://orcid.org/0000-0003-4119-0544
Michael N. Weintraub https://orcid.org/0000-0002-9623-2855
Ji Chen https://orcid.org/0000-0001-7026-6312

REFERENCES

Ågren, G. I., Bosatta, E., & Magill, A. H. (2001). Combining theory and experiment to understand effects of inorganic nitrogen on litter decomposition. *Oecologia*, *128*, 94–98.

Balesdent, J., Basile-Doelsch, I., Chadoeuf, J., Cornu, S., Derrien, D., Fekiacova, Z., & Hatté, C. (2018). Atmosphere–soil carbon transfer as a function of soil depth. *Nature*, *559*, 599–602.

Bowden, R. D., Wurzbacher, S. J., Washko, S. E., Wind, L., Rice, A. M., Coble, A. E., Baldauf, N., Johnson, B., Wang, J. J., Simpson, M., & Lajtha, K. (2019). Long-term nitrogen addition decreases organic matter decomposition and increases forest soil carbon. *Soil Science Society of America Journal*, *83*, S82–S95.

Calcagno, V., & De Mazancourt, C. (2010). Glmulti: An R package for easy automated model selection with (generalized) linear models. *Journal of Statistical Software*, *34*, 1.

Canary, J. D., Harrison, R. B., Compton, J. E., & Chappell, H. N. (2000). Additional carbon sequestration following repeated urea fertilization of second-growth Douglas-fir stands in western Washington. *Forest Ecology and Management*, *138*, 225–232.

Chen, J., Luo, Y., Kätterer, T., & Olesen, J. E. (2022). Depth-dependent responses of soil organic carbon stock under annual and perennial cropping systems. *Proceedings of the National Academy of Sciences of the United States of America*, *119*, e2203486119.

Chen, J., Luo, Y., & Sinsabaugh, R. L. (2023). Subsoil carbon loss. *Nature Geoscience*, *16*, 284–285.

Chen, J., Luo, Y., Van Groenigen, K. J., Hungate, B. A., Cao, J., Zhou, X., & Wang, R. (2018). A keystone microbial enzyme for nitrogen control of soil carbon storage. *Science Advances*, *4*, eaaq1689.

Chen, X., Chen, H. Y. H., Chen, C., Ma, Z., Searle, E. B., Yu, Z., & Huang, Z. (2019). Effects of plant diversity on soil carbon in diverse ecosystems: A global meta-analysis. *Biological Reviews*, *95*, 167–183.

Cochran, V. L., Elliott, L. F., & Lewis, C. E. (1989). Soil microbial biomass and enzyme activity in subarctic agricultural and forest soils. *Biology and Fertility of Soils*, *7*, 283–288.

Deng, L., Huang, C., Kim, D. G., Shangguan, Z., Wang, K., Song, X., & Peng, C. (2020). Soil GHG fluxes are altered by N deposition: New data indicate lower N stimulation of the N_2O flux and greater stimulation of the calculated C pools. *Global Change Biology*, *26*, 2613–2629.

Duan, L., Huang, Y., Hao, J., Xie, S., & Hou, M. (2004). Vegetation uptake of nitrogen and base cations in China and its role in soil acidification. *Science of the Total Environment*, *330*, 187–198.

Eilers, K. G., Debenport, S., Anderson, S., & Fierer, N. (2012). Digging deeper to find unique microbial communities: The strong effect of depth on the structure of bacterial and archaeal communities in soil. *Soil Biology and Biochemistry*, *50*, 58–65.

Federle, T. W., Dobbins, D. C., Thornton-Manning, J. R., & Jones, D. D. (1986). Microbial biomass, activity, and community structure in subsurface soils. *Groundwater*, *24*, 365–374.

Fick, S. E., & Hijmans, R. J. (2017). WorldClim 2: New 1-km spatial resolution climate surfaces for global land areas. *International Journal of Climatology*, *37*, 4302–4315.

Fierer, N., Allen, A. S., Schimel, J. P., & Holden, P. A. (2003). Controls on microbial CO_2 production: A comparison of surface and subsurface soil horizons. *Global Change Biology*, *9*, 1322–1332.

Fontaine, S., Barot, S., Barré, P., Bdioui, N., Mary, B., & Rumpel, C. (2007). Stability of organic carbon in deep soil layers controlled by fresh carbon supply. *Nature*, *450*, 277–280.

Fowler, D., Coyle, M., Skiba, U., Sutton, M. A., Cape, J. N., Reis, S., Sheppard, L. J., Jenkins, A., Grizzetti, B., Galloway, J. N., Vitousek, P., Leach, A., Bouwman, A. F., Butterbach-Bahl, K., Dentener, F., Stevenson, D., Amann, M., & Voss, M. (2013). The global nitrogen cycle in the twenty-first century. *Philosophical Transactions of the Royal Society, B: Biological Sciences*, *368*, 20130164.

Franklin, O., Högberg, P., Ekblad, A., & Ågren, G. I. (2003). Pine forest floor carbon accumulation in response to N and PK additions:

Bomb ^{14}C modelling and respiration studies. *Ecosystems*, *6*, 644–658.

Greaver, T. L., Clark, C. M., Compton, J. E., Vallano, D., Talhelm, A. F., Weaver, C. P., Band, L. E., Baron, J. S., Davidson, E. A., Tague, C. L., Felker-Quinn, E., Lynch, J. A., Herrick, J. D., Liu, L., Goodale, C. L., Novak, K. J., & Haeuber, R. A. (2016). Key ecological responses to nitrogen are altered by climate change. *Nature Climate Change*, *6*, 836–843.

Gruber, N., & Galloway, J. N. (2008). An earth-system perspective of the global nitrogen cycle. *Nature*, *451*, 293–296.

Hartemink, A. E., Zhang, Y., Bockheim, J. G., Curi, N., Silva, S. H. G., Grauer-Gray, J., Lowe, D. J., & Krasilnikov, P. (2020). Soil horizon variation: A review. *Advances in Agronomy*, *160*, 125–185.

He, M., Fang, K., Chen, L., Feng, X., Qin, S., Kou, D., He, H., Liang, C., & Yang, Y. (2022). Depth-dependent drivers of soil microbial necromass carbon across Tibetan alpine grasslands. *Global Change Biology*, *28*, 936–949.

Hedges, L. V., Gurevitch, J., & Curtis, P. S. (1999). The meta-analysis of response ratios in experimental ecology. *Ecology*, *80*, 1150–1156.

Henneron, L., Balesdent, J., Alvarez, G., Barré, P., Baudin, F., Basile-Doelsch, I., Cécillon, L., Fernandez-Martinez, A., Hatté, C., & Fontaine, S. (2022). Bioenergetic control of soil carbon dynamics across depth. *Nature Communications*, *13*, 7676.

Hicks Pries, C. E., Castanha, C., Porras, R. C., & Torn, M. S. (2017). The whole-soil carbon flux in response to warming. *Science*, *355*, 1420–1423.

Hyvönen, R., Persson, T., Andersson, S., Olsson, B., Ågren, G. I., & Linder, S. (2008). Impact of long-term nitrogen addition on carbon stocks in trees and soils in northern Europe. *Biogeochemistry*, *89*, 121–137.

IPCC (Intergovernmental Panel on Climate Change). (2006). Agriculture, forestry and other land use. In H. Eggleston, L. Buendia, K. Miwa, T. Ngara, & K. Tanabe (Eds.), *IPCC Guidelines for National Greenhouse Gas Inventories*. Kanagawa, Japan: Institute for Global Environmental Strategies.

IPCC (Intergovernmental Panel on Climate Change). (2021). Summary for Policymakers. In V. Masson-Delmotte, P. Zhai, A. Pirani, S. L. Connors, C. Péan, S. Berger, N. Caud, Y. Chen, & L. Goldfarb (Eds.), *Climate change 2021: The physical science basis. Contribution of working group I to the sixth assessment report of the Intergovernmental Panel on Climate Change*. Cambridge University Press.

Jansone, L., von Wilpert, K., & Hartmann, P. (2020). Natural recovery and liming effects in acidified forest soils in SW-Germany. *Soil Systems*, *4*, 38.

Janssens, I. A., Dieleman, W., Luyssaert, S., Subke, J. A., Reichstein, M., Ceulemans, R., Ciais, P., Dolman, A. J., Grace, J., Matteucci, G., Papale, D., Piao, S. L., Schulze, E. D., Tang, J., & Law, B. E. (2010). Reduction of forest soil respiration in response to nitrogen deposition. *Nature Geoscience*, *3*, 315–322.

Jilling, A., Keiluweit, M., Gutknecht, J. L. M., & Grandy, A. S. (2021). Priming mechanisms providing plants and microbes access to mineral-associated organic matter. *Soil Biology and Biochemistry*, *158*, 108265.

Jobbágy, E. G., & Jackson, R. B. (2000). The vertical distribution of soil organic carbon and its relation to climate and vegetation. *Ecological Applications*, *10*, 423–436.

Kaiser, K., & Kalbitz, K. (2012). Cycling downwards—Dissolved organic matter in soils. *Soil Biology and Biochemistry*, *52*, 29–32.

Karhu, K., Hilasvuori, E., Fritze, H., Biasi, C., Nykänen, H., Liski, J., Vanhala, P., Heinonsalo, J., & Pumpanen, J. (2016). Priming effect increases with depth in a boreal forest soil. *Soil Biology and Biochemistry*, *99*, 104–107.

Kuebbing, S. E., Reimer, A. P., Rosenthal, S. A., Feinberg, G., Leiserowitz, A., Lau, J. A., & Bradford, M. A. (2018). Long-term research in ecology and evolution: A survey of challenges and opportunities. *Ecological Monographs*, *88*, 245–258.

Li, H., Wu, Y., Liu, S., Xiao, J., Zhao, W., Chen, J., Alexandrov, G., & Cao, Y. (2022). Decipher soil organic carbon dynamics and driving forces across China using machine learning. *Global Change Biology*, *28*, 3394–3410.

Li, J. H., Yang, Y. J., Li, B. W., Li, W. J., Wang, G., & Knops, J. M. H. (2014). Effects of nitrogen and phosphorus fertilization on soil carbon fractions in alpine meadows on the Qinghai-Tibetan plateau. *PLoS ONE*, *9*, e103266.

Li, X., Zhang, C., Zhang, B., Wu, D., Zhu, D., Zhang, W., Ye, Q., Yan, J., Fu, J., Fang, C., Ha, D., & Fu, S. (2021). Nitrogen deposition and increased precipitation interact to affect fine root production and biomass in a temperate forest: Implications for carbon cycling. *Science of the Total Environment*, *765*, 144497.

Li, Y., Niu, S., & Yu, G. (2016). Aggravated phosphorus limitation on biomass production under increasing nitrogen loading: A meta-analysis. *Global Change Biology*, *22*, 934–943.

Liao, L., Wang, J., Lei, S., Zhang, L., Ye, Z., Liu, G., & Zhang, C. (2023). Differential effects of nitrogen addition on the organic carbon fractions of rhizosphere and bulk soil based on a pot experiment. *Journal of Soils and Sediments*, *23*, 103–117.

Liu, H. Y., Huang, N., Zhao, C. M., & Li, J. H. (2023). Responses of carbon cycling and soil organic carbon content to nitrogen addition in grasslands globally. *Soil Biology and Biochemistry*, *186*, 109164.

Liu, L., & Greaver, T. L. (2010). A global perspective on belowground carbon dynamics under nitrogen enrichment. *Ecology Letters*, *13*, 819–828.

Lu, M., Zhou, X., Luo, Y., Yang, Y., Fang, C., Chen, J., & Li, B. (2011). Minor stimulation of soil carbon storage by nitrogen addition: A meta-analysis. *Agriculture, Ecosystems & Environment*, *140*, 234–244.

Lu, X., Gilliam, F. S., Yue, X., Wang, B., & Kuang, Y. (2023). Shifts in above-versus below-ground carbon gains to terrestrial ecosystems carbon sinks under excess nitrogen inputs. *Global Biogeochemical Cycles*, *37*, e2022GB007638.

Lu, X., Hou, E., Guo, J., Gilliam, F. S., Li, J., Tang, S., & Kuang, Y. (2021). Nitrogen addition stimulates soil aggregation and enhances carbon storage in terrestrial ecosystems of China: A meta-analysis. *Global Change Biology*, *27*, 2780–2792.

Luo, Z., Wang, G., & Wang, E. (2019). Global subsoil organic carbon turnover times dominantly controlled by soil properties rather than climate. *Nature Communications*, *10*, 3688.

Mathieu, J. A., Hatté, C., Balesdent, J., & Parent, É. (2015). Deep soil carbon dynamics are driven more by soil type than by climate: A worldwide meta-analysis of radiocarbon profiles. *Global Change Biology*, *21*, 4278–4292.

Minasny, B., Malone, B. P., McBratney, A. B., Angers, D. A., Arrouays, D., Chambers, A., Chaplot, V., Chen, Z. S., Cheng, K., Das, B. S., Field, D. J., Gimona, A., Hedley, C. B., Young Hong, S., Mandal, B., Marchant, B. P., Martin, M., McConkey, B. G., Mulder, V. L., … Winowiecki, L. (2017). Soil carbon 4 per mille. *Geoderma*, *292*, 59–86.

Mobley, M. L., Lajtha, K., Kramer, M. G., Bacon, A. R., Heine, P. R., & Richter, D. D. (2015). Surficial gains and subsoil losses of soil carbon and nitrogen during secondary forest development. *Global Change Biology*, *21*, 986–996.

Nave, L. E., Vance, E. D., Swanston, C. W., & Curtis, P. S. (2009). Impacts of elevated N inputs on north temperate forest soil C storage, C/N, and net N-mineralization. *Geoderma*, *153*, 231–240.

Ni, H., Liu, C., Sun, B., & Liang, Y. (2022). Response of global farmland soil organic carbon to nitrogen application over time depends on soil type. *Geoderma*, *406*, 115542.

Ning, Q., Hättenschwiler, S., Lü, X., Kardol, P., Zhang, Y., Wei, C., Xu, C., Huang, J., Li, A., Yang, J., Wang, J., Peng, Y., Peñuelas, J., Sardans, J., He, J., Xu, Z., Gao, Y., & Han, X. (2021). Carbon limitation overrides acidification in mediating soil microbial activity to nitrogen enrichment in a temperate grassland. *Global Change Biology*, *27*, 5976–5988.

Niu, G., Hasi, M., Wang, R., Wang, Y., Geng, Q., Hu, S., Xu, X., Yang, J., Wang, C., Han, X., & Huang, J. (2021). Soil microbial community responses to long-term nitrogen addition at different soil depths in a typical steppe. *Applied Soil Ecology*, *167*, 104054.

ORNL DAAC. (2017). *Spatial data access tool (SDAT)*. ORNL DAAC.

Peñuelas, J., Poulter, B., Sardans, J., Ciais, P., van der Velde, M., Bopp, L., Boucher, O., Godderis, Y., Hinsinger, P., Llusia, J., Nardin, E., Vicca, S., Obersteiner, M., & Janssens, I. A. (2013). Human-induced nitrogen–phosphorus imbalances alter natural and managed ecosystems across the globe. *Nature Communications*, *4*, 2934.

Poeplau, C., Zopf, D., Greiner, B., Geerts, R., Korvaar, H., Thumm, U., Don, A., Heidkamp, A., & Flessa, H. (2018). Why does mineral fertilization increase soil carbon stocks in temperate grasslands? *Agriculture, Ecosystems & Environment*, *265*, 144–155.

Pregitzer, K. S., Burton, A. J., Zak, D. R., & Talhelm, A. F. (2008). Simulated chronic nitrogen deposition increases carbon storage in northern temperate forests. *Global Change Biology*, *14*, 142–153.

Pregitzer, K. S., Zak, D. R., Burton, A. J., Ashby, J. A., & MacDonald, N. W. (2004). Chronic nitrate additions dramatically increase the export of carbon and nitrogen from northern hardwood ecosystems. *Biogeochemistry*, *68*, 179–197.

Qi, P., Chen, J., Wang, X., Zhang, R., Cai, L., Jiao, Y., Li, Z., & Han, G. (2023). Changes in soil particulate and mineral-associated organic carbon concentrations under nitrogen addition in China—A meta-analysis. *Plant and Soil*, *489*, 439–452.

Qin, S., Yuan, H., Hu, C., Li, X., Wang, Y., Zhang, Y., Dong, W., Clough, T., Luo, J., Zhou, S., Wrage-Mönnig, N., Ma, L., & Oenema, O. (2023). Anthropogenic N input increases global warming potential by awakening the "sleeping" ancient C in deep critical zones. *Science Advances*, *9*, eadd0041.

Reay, D. S., Dentener, F., Smith, P., Grace, J., & Feely, R. A. (2008). Global nitrogen deposition and carbon sinks. *Nature Geoscience*, *1*, 430–437.

Rubin, D. B., & Schenker, N. (1991). Multiple imputation in health-are databases: An overview and some applications. *Statistics in Medicine*, *10*, 585–598.

Rumpel, C., & Kögel-Knabner, I. (2011). Deep soil organic matter—A key but poorly understood component of terrestrial C cycle. *Plant and Soil*, *338*, 143–158.

Salomé, C., Nunan, N., Pouteau, V., Lerch, T. Z., & Chenu, C. (2010). Carbon dynamics in topsoil and in subsoil may be controlled by different regulatory mechanisms. *Global Change Biology*, *16*, 416–426.

Shahzad, T., Anwar, F., Hussain, S., Mahmood, F., Arif, M. S., Sahar, A., Nawaz, M. F., Perveen, N., Sanaullah, M., Rehman, K., & Rashid, M. I. (2019). Carbon dynamics in surface and deep soil in response to increasing litter addition rates in an agro-ecosystem. *Geoderma*, *333*, 1–9.

Shi, Z., Allison, S. D., He, Y., Levine, P. A., Hoyt, A. M., Beem-Miller, J., Zhu, Q., Wieder, W. R., Trumbore, S., & Randerson, J. T. (2020). The age distribution of global soil carbon inferred from radiocarbon measurements. *Nature Geoscience*, *13*, 555–559.

Sierra, J., & Renault, P. (1998). Temporal pattern of oxygen concentration in a hydromorphic soil. *Soil Science Society of America Journal*, *62*, 1398–1405.

Sinsabaugh, R. L., Zak, D. R., Gallo, M., Lauber, C., & Amonette, R. (2004). Nitrogen deposition and dissolved organic carbon production in northern temperate forests. *Soil Biology and Biochemistry*, *36*, 1509–1515.

Sun, S., Liu, X., Lu, S., Cao, P., Hui, D., Chen, J., Guo, J., & Yang, Y. (2023). Depth-dependent response of particulate and mineral-associated organic carbon to long-term throughfall reduction in a subtropical natural forest. *Catena*, *223*, 106904.

Terrer, C., Vicca, S., Hungate, B. A., Phillips, R. P., & Prentice, I. C. (2016). Mycorrhizal association as a primary control of the CO_2 fertilization effect. *Science*, *353*, 72–74.

Treseder, K. K. (2004). A meta-analysis of mycorrhizal responses to nitrogen, phosphorus, and atmospheric CO_2 in field studies. *New Phytologist*, *164*, 347–355.

Treseder, K. K. (2008). Nitrogen additions and microbial biomass: A meta-analysis of ecosystem studies. *Ecology Letters*, *11*, 1111–1120.

van Groenigen, K. J., Osenberg, C. W., & Hungate, B. A. (2011). Increased soil emissions of potent greenhouse gases under increased atmospheric CO_2. *Nature*, *475*, 214–216.

Viechtbauer, W. (2010). Conducting meta-analyses in R with the metafor package. *Journal of Statistical Software*, *36*, 1–48.

Vourlitis, G. L., & Hentz, C. S. (2016). Impacts of chronic N input on the carbon and nitrogen storage of a postfire Mediterranean-type shrubland. *Journal of Geophysical Research: Biogeosciences*, *121*, 385–398.

Waldrop, M. P., Zak, D. R., Sinsabaugh, R. L., Gallo, M., & Lauber, C. (2004). Nitrogen deposition modifies soil carbon storage through changes in microbial enzymatic activity. *Ecological Applications*, *14*, 1172–1177.

Wang, J. J., Bowden, R. D., Lajtha, K., Washko, S. E., Wurzbacher, S. J., & Simpson, M. J. (2019). Long-term nitrogen addition suppresses microbial degradation, enhances soil carbon storage, and alters the molecular composition of soil organic matter. *Biogeochemistry*, *142*, 299–313.

Wang, M., Guo, X., Zhang, S., Xiao, L., Mishra, U., Yang, Y., Zhu, B., Wang, G., Mao, X., Qian, T., Jiang, T., Shi, Z., & Luo, Z. (2022). Global soil profiles indicate depth-dependent soil carbon losses under a warmer climate. *Nature Communications*, *13*, 5514.

Wang, Q., Wang, Y., Wang, S., He, T., & Liu, L. (2014). Fresh carbon and nitrogen inputs alter organic carbon mineralization and microbial community in forest deep soil layers. *Soil Biology and Biochemistry*, *72*, 145–151.

Wilkinson, M. T., Richards, P. J., & Humphreys, G. S. (2009). Breaking ground: Pedological, geological, and ecological implications of soil bioturbation. *Earth-Science Reviews*, *97*, 257–272.

Xu, C., Xu, X., Ju, C., Chen, H. Y. H., Wilsey, B. J., Luo, Y., & Fan, W. (2021). Long-term, amplified responses of soil organic carbon to nitrogen addition worldwide. *Global Change Biology*, *27*, 1170–1180.

Yan, G., Chen, F., Zhang, X., Wang, J., Han, S., Xing, Y., & Wang, Q. (2017). Spatial and temporal effects of nitrogen addition on root morphology and growth in a boreal forest. *Geoderma*, *303*, 178–187.

Zheng, M., Zhang, T., Luo, Y., Liu, J., Lu, X., Ye, Q., Wang, S., Huang, J., Mao, Q., Mo, J., & Zhang, W. (2022). Temporal patterns of soil carbon emission in tropical forests under long-term nitrogen deposition. *Nature Geoscience*, *15*, 1002–1010.

Zhou, L., Zhou, X., Zhang, B., Lu, M., Luo, Y., Liu, L., & Li, B. (2014). Different responses of soil respiration and its components to nitrogen addition among biomes: A meta-analysis. *Global Change Biology*, *20*, 2332–2343.

Zhu, H., Zhao, J., & Gong, L. (2021). The morphological and chemical properties of fine roots respond to nitrogen addition in a temperate Schrenk's spruce (Picea schrenkiana) forest. *Scientific Reports*, *11*, 3839.

DATA SOURCES

Aarnio, T., Räty, M., & Martikainen, P. J. (2003). Long-term availability of nutients in forest soil derived from fast- and slow-release fertilizers. *Plant and Soil*, *252*, 227–239.

Ajwa, H. A., Dell, C. J., & Rice, C. W. (1999). Changes in enzyme activities and microbial biomass of tallgrass prairie soil as related to burning and nitrogen fertilization. *Soil Biology and Biochemistry*, *31*, 769–777.

Baer, S. G., & Blair, J. M. (2008). Grassland establishment under varying resource availability: A test of positive and negative feedback. *Ecology*, *89*, 1859–1871.

Bechtold, H. A., & Inouye, R. S. (2007). Distribution of carbon and nitrogen in sagebrush steppe after six years of nitrogen addition and shrub removal. *Journal of Arid Environments*, *71*, 122–132.

Bowden, R. D., Wurzbacher, S. J., Washko, S. E., Wind, L., Rice, A. M., Coble, A. E., Baldauf, N., Johnson, B., Wang, J.-J., Simpson, M., & Lajtha, K. (2019).

Long-term nitrogen addition decreases organic matter decomposition and increases forest soil carbon. *Soil Science Society of America Journal*, *83*, S82–S95.

Bradley, K., Drijber, R. A., & Knops, J. (2006). Increased N availability in grassland soils modifies their microbial communities and decreases the abundance of arbuscular mycorrhizal fungi. *Soil Biology and Biochemistry*, *38*, 1583–1595.

Brenner, R. E., Boone, R. D., & Ruess, R. W. (2005). Nitrogen additions to pristine, high-latitude, forest ecosystems: Consequences for soil nitrogen transformations and retention in mid and late succession. *Biogeochemistry*, *72*, 257–282.

Calvo-Fernández, J., Taboada, Á., Fichtner, A., Härdtle, W., Calvo, L., & Marcos, E. (2018). Time- and age-related effects of experimentally simulated nitrogen deposition on the functioning of montane heathland ecosystems. *Science of the Total Environment*, *613–614*, 149–159.

Canary, J. D., Harrison, R. B., Compton, J. E., & Chappell, H. N. (2000). Additional carbon sequestration following repeated urea fertilization of second-growth Douglas-fir stands in western Washington. *Forest Ecology and Management*, *138*, 225–232.

Cenini, V. L., Fornara, D. A., McMullan, G., Ternan, N., Lajtha, K., & Crawley, M. J. (2015). Chronic nitrogen fertilization and carbon sequestration in grassland soils: Evidence of a microbial enzyme link. *Biogeochemistry*, *126*, 301–313.

Chen, C., Xu, Z., & Hughes, J. (2002). Effects of nitrogen fertilization on soil nitrogen pools and microbial properties in a hoop pine (*Araucaria cunninghamii*) plantation in southeast Queensland, Australia. *Biology and Fertility of Soils*, *36*, 276–283.

Chen, H., Chen, M., Li, D., Mao, Q., Zhang, W., & Mo, J. (2018). Responses of soil phosphorus availability to nitrogen addition in a legume and a non-legume plantation. *Geoderma*, *322*, 12–18.

Chen, W., Su, F., Nie, Y., Zhong, B., Zheng, Y., Mo, J., Xiong, B., & Lu, X. (2022). Divergent responses of soil microbial functional groups to long-term high nitrogen presence in the tropical forests. *Science of the Total Environment*, *821*, 153251.

Chen, W., Zhou, H., Wu, Y., Wang, J., Zhao, Z., Li, Y., Qiao, L., Chen, K., Liu, G., & Xue, S. (2020). Direct and indirect influences of long-term fertilization on microbial carbon and nitrogen cycles in an alpine grassland. *Soil Biology and Biochemistry*, *149*, 107922.

Chen, X., Hao, B., Jing, X., He, J., Ma, W., & Zhu, B. (2019). Minor responses of soil microbial biomass, community structure and enzyme activities to nitrogen and phosphorus addition in three grassland ecosystems. *Plant and Soil*, *444*, 21–37.

Chen, X., Liu, J., Deng, Q., Yan, J., & Zhang, D. (2012). Effects of elevated CO_2 and nitrogen addition on soil organic carbon fractions in a subtropical forest. *Plant and Soil*, *357*, 25–34.

Chu, B., Li, P., Sun, M., Wang, J., Teng, Z., & Xu, X. (2021). Effects of nitrogen and phosphorus additions on soil carbon and nitrogen and their stable isotopes in a subtropical evergreen broad-leaved forest. *Journal of Central South University of Forestry & Technology*, *8*, 100–107.

Cui, H., Fan, M., Wang, Y., Zhang, Z., Xu, W., Li, Y., Song, W., Ma, J., & Sun, W. (2023). Impacts of mowing and N addition on soil organic phosphorus mineralization rates in a semi-natural grassland in Northeast China. *Plant and Soil*, *482*, 7–23.

Cui, H., Sun, W., Delgado-Baquerizo, M., Song, W., Ma, J., Wang, K., & Ling, X. (2022). Phosphorus addition regulates the responses of soil multifunctionality to nitrogen over-fertilization in a temperate grassland. *Plant and Soil*, *473*, 73–87.

Cusack, D. F., Torn, M. S., McDowell, W. H., & Silver, W. L. (2010). The response of heterotrophic activity and carbon cycling to nitrogen additions and warming in two tropical soils. *Global Change Biology*, *16*, 2555–2572.

Dijkstra, F. A., Hobbie, S. E., Reich, P. B., & Knops, J. M. H. (2005). Divergent effects of elevated CO_2, N fertilization, and plant diversity on soil C and N dynamics in a grassland field experiment. *Plant and Soil*, *272*, 41–52.

Dong, C., Wang, W., Liu, H., Xu, X., Chen, X., & Zeng, H. (2021). Comparison of soil microbial responses to nitrogen addition between ex-arable grassland and natural grassland. *Journal of Soils and Sediments*, *21*, 1371–1384.

Fan, Y., Lin, F., Yang, L., Zhong, X., Wang, M., Zhou, J., Chen, Y., & Yang, Y. (2018). Decreased soil organic P fraction associated with ectomycorrhizal fungal activity to meet increased P demand under N application in a subtropical forest ecosystem. *Biology and Fertility of Soils*, *54*, 149–161.

Fan, Y., Yang, L., Zhong, X., Yang, Z., Lin, Y., Guo, J., Chen, G., & Yang, Y. (2020). N addition increased microbial residual carbon by altering soil P availability and microbial composition in a subtropical *Castanopsis* forest. *Geoderma*, *375*, 114470.

Fang, X., Zhang, X., Chen, F., Zong, Y., Bu, W., Wan, S., Luo, Y., & Wang, H. (2019). Phosphorus addition alters the response of soil organic carbon decomposition to nitrogen deposition in a subtropical forest. *Soil Biology and Biochemistry*, *133*, 119–128.

Fenner, A. (2015). Effects of long-term atmospheric nitrogen deposition on extracellular enzyme activity in semi-arid shrubland soil.

Fornara, D. A., Banin, L., & Crawley, M. J. (2013). Multi-nutrient vs. nitrogen-only effects on carbon sequestration in grassland soils. *Global Change Biology*, *19*, 3848–3857.

Fornara, D. A., & Tilman, D. (2012). Soil carbon sequestration in prairie grasslands increased by chronic nitrogen addition. *Ecology*, *93*, 2030–2036.

Fox, T. R. (2004). Nitrogen mineralization following fertilization of Douglas-fir forests with urea in western Washington. *Soil Science Society of America Journal*, *68*, 1720–1728.

Freppaz, M., Williams, M. W., Seastedt, T., & Filippa, G. (2012). Response of soil organic and inorganic nutrients in alpine soils to a 16-year factorial snow and N-fertilization experiment, Colorado Front Range, USA. *Applied Soil Ecology*, *62*, 131–141.

Frey, S. D., Knorr, M., Parrent, J. L., & Simpson, R. T. (2004). Chronic nitrogen enrichment affects the structure and function of the soil microbial community in temperate hardwood and pine forests. *Forest Ecology and Management*, *196*, 159–171.

Gallo, M. E., Lauber, C. L., Cabaniss, S. E., Waldrop, M. P., Sinsabaugh, R. L., & Zak, D. R. (2005). Soil organic matter and litter chemistry response to experimental N deposition in northern temperate deciduous forest ecosystems. *Global Change Biology*, *11*, 1514–1521.

Geng, P., & Jin, G. (2022). Fine root morphology and chemical responses to N addition depend on root function and soil depth in a Korean pine plantation in Northeast China. *Forest Ecology and Management*, *520*, 120407.

Gong, S., Zhang, T., Guo, R., Cao, H., Shi, L., Guo, J., & Sun, W. (2015). Response of soil enzyme activity to warming and nitrogen addition in a meadow steppe. *Soil Research*, *53*, 242–252.

Grandy, A. S., Sinsabaugh, R. L., Neff, J. C., Stursova, M., & Zak, D. R. (2008). Nitrogen deposition effects on soil organic matter chemistry are linked to variation in enzymes, ecosystems and size fractions. *Biogeochemistry*, *91*, 37–49.

Grulke, N. E., Dobrowolski, W., Mingus, P., & Fenn, M. E. (2005). California black oak response to nitrogen amendment at a high O_3, nitrogen-saturated site. *Environmental Pollution*, *137*, 536–545.

Guan, B., Xie, B., Yang, S., Hou, A., Chen, M., & Han, G. (2019). Effects of five years' nitrogen deposition on soil properties and plant growth in a salinized reed wetland of the Yellow River Delta. *Ecological Engineering*, *136*, 160–166.

Gulledge, J., Hrywna, Y., Cavanaugh, C., & Steudler, P. A. (2004). Effects of long-term nitrogen fertilization on the uptake kinetics of atmospheric methane in temperate forest soils. *FEMS Microbiology Ecology*, *49*, 389–400.

Haile-Mariam, S., Cheng, W., Johnson, D. W., Ball, J. T., & Paul, E. A. (2000). Use of carbon-13 and carbon-14 to measure the effects of carbon dioxide and nitrogen fertilization on carbon dynamics in ponderosa pine. *Soil Science Society of America Journal*, *64*, 1984–1993.

Han, B., Li, J., Liu, K., Zhang, H., Wei, X., & Shao, X. (2021). Variations in soil properties rather than functional gene abundances dominate soil phosphorus dynamics under short-term nitrogen input. *Plant and Soil*, *469*, 227–241.

He, L. (2018). Effects of nitrogen addition on soil microbial and soil ecological stoichiometry properties of *Pinus tabulaeformis* forest.

He, P., Li, Y., Jiang, M., Li, Y., Du, W., Zhang, J., & Jing, H. (2021). Effects of 14-year continuous nitrogen addition on soil carbon and nitrogen composition and physical structure at different depths in a typical temperate steppe. *Acta Ecologica Sinica*, *5*, 1808–1823.

He, R., Yan, B., Sun, Y., He, G., & Shi, L. (2021). Effects of plant species and nitrogen addition on soil organic carbon decomposition in the Yuanmou dry-hot valley grassland, China. *Chinese Journal of Applied & Environmental Biology*, *6*, 1547–1553.

Herbert, E. R., Schubauer-Berigan, J. P., & Craft, C. B. (2020). Effects of 10 yr of nitrogen and phosphorus fertilization on carbon and nutrient cycling in a tidal freshwater marsh. *Limnology and Oceanography*, *65*, 1669–1687.

Homann, P. S., Caldwell, B. A., Chappell, H. N., Sollins, P., & Swanston, C. W. (2001). Douglas-fir soil C and N properties a decade after termination of urea fertilization. *Canadian Journal of Forest Research*, *31*, 2225–2236.

Hu, Y., Cong, M., Chen, M., Hou, T., Yu, G., Maidimuer, A., Zhu, X., & Jia, H. (2022). Nitrogen addition significantly affected the soil inorganic phosphorus forms in Bayinbuluk alpine wetland. *Journal of Soil and Water Conservation*, *3*, 252–258.

Hu, Y., Jung, K., Zeng, D., & Chang, S. (2013). Nitrogen- and sulfur-deposition-altered soil microbial community functions and enzyme activities in a boreal mixedwood forest in western Canada. *Canadian Journal of Forest Research*, *43*, 777–784.

Hu, Y., Zeng, D., Liu, Y., Zhang, Y., Chen, Z., & Wang, Z. (2010). Responses of soil chemical and biological properties to nitrogen addition in a Dahurian larch plantation in Northeast China. *Plant and Soil*, *333*, 81–92.

Huang, W., Zhou, G., Liu, J., Duan, H., Liu, X., Fang, X., & Zhang, D. (2014). Shifts in soil phosphorus fractions under elevated CO_2 and N addition in model forest ecosystems in subtropical China. *Plant Ecology*, *215*, 1373–1384.

Hungate, B. A., Hart, S. C., Selmants, P. C., Boyle, S. I., & Gehring, C. A. (2007). Soil responses to management, increased precipitation, and added nitrogen in ponderosa pine forests. *Ecological Applications*, *17*, 1352–1365.

Iyyemperumal, K., & Shi, W. (2008). Soil enzyme activities in two forage systems following application of different rates of swine lagoon effluent or ammonium nitrate. *Applied Soil Ecology*, *38*, 128–136.

Jia, S., Liu, X., Lin, W., Zheng, Y., Li, J., Hui, D., & Guo, J. (2022). Decreased glomalin-related soil protein with nitrogen deposition in a 3-year-old Cunninghamia lanceolata plantation. *Journal of Soils and Sediments*, *22*, 931–941.

Jiang, X., Cao, L., & Zhang, R. (2014). Changes of labile and recalcitrant carbon pools under nitrogen addition in a city lawn soil. *Journal of Soils and Sediments*, *14*, 515–524.

Johnson, D. W., Cheng, W., & Ball, J. T. (2000). Effects of [CO_2] and nitrogen fertilization on soils planted with ponderosa pine. *Plant and Soil*, *224*, 99–113.

Jones, S. K., Rees, R. M., Kosmas, D., Ball, B. C., & Skiba, U. M. (2006). Carbon sequestration in a temperate grassland; management and climatic controls. *Soil Use and Management*, *22*, 132–142.

Jung, J. Y., Lal, R., & Ussiri, D. A. N. (2011). Changes in CO_2, ^{13}C abundance, inorganic nitrogen, β-glucosidase, and oxidative enzyme activities of soil during the decomposition of switchgrass root carbon as affected by inorganic nitrogen additions. *Biology and Fertility of Soils*, *47*, 801–813.

Keeler, B. L., Hobbie, S. E., & Kellogg, L. E. (2009). Effects of long-term nitrogen addition on microbial enzyme activity in eight forested and grassland sites: Implications for litter and soil organic matter decomposition. *Ecosystems*, *12*, 1–15.

Kritzler, U. H., & Johnson, D. (2010). Mineralisation of carbon and plant uptake of phosphorus from microbially-derived organic matter in response to 19 years simulated nitrogen deposition. *Plant and Soil*, *326*, 311–319.

Lee, K., & Jose, S. (2003). Soil respiration, fine root production, and microbial biomass in cottonwood and loblolly pine plantations along a nitrogen fertilization gradient. *Forest Ecology and Management*, *185*, 263–273.

Li, G., Cai, J., Song, X., Pan, X., Pan, D., Jiang, S., Sun, J., Zhang, M., & Wang, L. (2022). Herbivore grazing mitigates the negative effects of nitrogen deposition on soil organic carbon in low-diversity grassland. *Journal of Applied Ecology*, *59*, 483–491.

Li, J., Sang, C., Yang, J., Qu, L., Xia, Z., Sun, H., Jiang, P., Wang, X., He, H., & Wang, C. (2021). Stoichiometric imbalance and microbial community regulate microbial elements use efficiencies under nitrogen addition. *Soil Biology and Biochemistry*, *156*, 108207.

Li, J. H., Hou, Y. L., Zhang, S. X., Li, W. J., Xu, D. H., Knops, J. M. H., & Shi, X. M. (2018). Fertilization with nitrogen and/or phosphorus lowers soil organic carbon sequestration in alpine meadows. *Land Degradation & Development*, *29*, 1634–1641.

Li, J. H., Yang, Y. J., Li, B. W., Li, W. J., Wang, G., & Knops, J. M. H. (2014). Effects of nitrogen and phosphorus fertilization on soil carbon fractions in alpine meadows on the Qinghai-Tibetan plateau. *PLoS ONE*, *9*, e103266.

Li, M., Jie, Q., Yu, H., Dian, Y., Guang, Z., Yu, W., & Li, W. (2019). Effects of nitrogen addition on ecological stoichiometric characteristics of carbon, nitrogen and phosphorus in Stipa baicalensis grassland soil aggregates. *Acta Prataculturae Sinica*, *28*, 29–33.

Li, S., Cai, Q., Liu, W., & Wu, J. (2023). Responses of stocks and stoichiometric characteristics of soil carbon, nitrogen and phosphorus to nitrogen addition in a Cunninghamia lanceolata plantation. *Journal of Yunnan University(Natural Sciences Edition)*, *6*, 1349–1358.

Li, Y., He, T., & Wang, Q. (2016). Impact of fertilization on soil organic carbon and enzyme activities in a Cunninghamia lanceolata plantation. *Chinese Journal of Ecology*, *35*, 2722.

Li, Y., Nie, C., Liu, Y., Du, W., & He, P. (2019). Soil microbial community composition closely associates with specific enzyme activities and soil carbon chemistry in a long-term nitrogen fertilized grassland. *Science of the Total Environment*, *654*, 264–274.

Li, Z., Qiu, X., Sun, Y., Liu, S., Hu, H., Xie, J., Chen, G., Xiao, Y., Tang, Y., & Tu, L. (2021). C:N:P stoichiometry responses to 10 years of nitrogen addition differ across soil components and plant organs in a subtropical *Pleioblastus amarus* forest. *Science of the Total Environment*, *796*, 148925.

Liu, C., Wu, D., Liu, Y., Chen, H., Shen, B., Jiang, Z., & Liu, X. (2021). Effects of nitrogen deposition on soil organic carbon and soil microbial communities in a natural Castanopsis carlesii forest. *Forest Research*, *2*, 42–49.

Liu, J., Xu, Z., Zhang, D., Zhou, G., Deng, Q., Duan, H., Zhao, L., & Wang, C. (2011). Effects of carbon dioxide enrichment and nitrogen addition on inorganic carbon leaching in subtropical model forest ecosystems. *Ecosystems*, *14*, 683–697.

Liu, J., Zhan, W., Huang, X., Tang, D., Jin, S., Zhu, D., & Chen, H. (2023). Nitrogen addition increases topsoil carbon stock in an alpine meadow of the Qinghai-Tibet Plateau. *Science of the Total Environment*, *888*, 164071.

Liu, J. X., Zhou, G. Y., Zhang, D. Q., Xu, Z. H., Duan, H. L., Deng, Q., & Zhao, L. (2010). Carbon dynamics in subtropical forest soil: Effects of atmospheric carbon dioxide enrichment and nitrogen addition. *Journal of Soils and Sediments*, *10*, 730–738.

Liu, K., & Crowley, D. (2009). Nitrogen deposition effects on carbon storage and fungal:Bacterial ratios in coastal sage scrub soils of southern California. *Journal of Environmental Quality*, *38*, 2267–2272.

Liu, Q., Yin, H., Chen, J., Zhao, C., Cheng, X., Wei, Y., & Lin, B. (2011). Belowground responses of Picea asperata seedlings to warming and nitrogen fertilization in the eastern Tibetan Plateau. *Ecological Research*, *26*, 637–648.

Liu, W., Jiang, Y., Wang, G., Su, Y., Smoak, J. M., Liu, M., & Duan, B. (2021). Effects of N addition and clipping on above and belowground plant biomass, soil microbial community structure, and function in an alpine meadow on the Qinghai-Tibetan Plateau. *European Journal of Soil Biology*, *106*, 103344.

Liu, X., Wang, J., & Zhao, X. (2015). Effects of simulated nitrogen deposition on the soil enzyme activities in a *Pinus tabulaeformis* forest at the Taiyue Mountain. *Acta Ecologica Sinica*, *35*, 4613–4624.

Liu, Y., Zhang, J., Li, Y., He, P., & Dong, J. (2022). Do long-term N additions affect the soil organic carbon pool in temperate grasslands? *Science of the Total Environment*, *810*, 152227.

Lovett, G. M., Arthur, M. A., Weathers, K. C., Fitzhugh, R. D., & Templer, P. H. (2013). Nitrogen addition increases carbon storage in soils, but not in trees, in an eastern U.S. deciduous forest. *Ecosystems*, *16*, 980–1001.

Lü, F., Lü, X., Liu, W., Han, X., Zhang, G., Kong, D., & Han, X. (2011). Carbon and nitrogen storage in plant and soil as related to nitrogen and water amendment in a temperate steppe of northern China. *Biology and Fertility of Soils*, *47*, 187–196.

Lü, S., Song, S., Li, Y., Zhong, Q., Ma, W., & Tu, L. (2022). Effects of nitrogen addition and litter increase or decrease on soil aggregates and their C and N in evergreen broad-leaved forest in rain screen area of west China. *Journal of Soil and Water Conservation*, *36*, 277–287.

Lu, X., Gilliam, F. S., Yu, G., Li, L., Mao, Q., Chen, H., & Mo, J. (2013). Long-term nitrogen addition decreases carbon leaching in a nitrogen-rich forest ecosystem. *Biogeosciences*, *10*, 3931–3941.

Lu, X., Vitousek, P. M., Mao, Q., Gilliam, F. S., Luo, Y., Turner, B. L., Zhou, G., & Mo, J. (2021). Nitrogen deposition accelerates soil carbon sequestration in tropical forests. *Proceedings of the National Academy of Sciences of the United States of America*, *118*, e2020790118.

Lugli, L. F., Rosa, J. S., Andersen, K. M., Di Ponzio, R., Almeida, R. V., Pires, M., Cordeiro, A. L., Cunha, H. F. V., Martins, N. P., Assis, R. L., et al. (2021). Rapid responses of root traits and productivity to phosphorus and cation additions in a tropical lowland forest in Amazonia. *New Phytologist*, *230*, 116–128.

Lv, J., Li, Q., Zhang, J., Li, Y., Peng, C., & Song, X. (2022). Nitrogen addition increases CO_2, CH_4, and N_2O concentrations of topsoils and subsoils in a subtropical Moso bamboo forest. *Catena*, *216*, 106397.

Ma, S., Chen, G., Tang, W., Xing, A., Chen, X., Xiao, W., Zhou, L., Zhu, J., Li, Y., Zhu, B., & Fang, J. (2021). Inconsistent responses of soil microbial community structure and enzyme activity to nitrogen and phosphorus additions in two tropical forests. *Plant and Soil*, *460*, 453–468.

Ma, X., Zhou, Z., Chen, J., Xu, H., Ma, S., Dippold, M. A., & Kuzyakov, Y. (2023). Long-term nitrogen and phosphorus fertilization reveals that phosphorus limitation shapes the microbial community composition and functions in tropical montane forest soil. *Science of the Total Environment*, *854*, 158709.

Mu, R., Jiao, T., Chen, X., Ma, S., Zhang, X., Qi, J., & Yu, X. (2022). Effects of nitrogen addition on soil fertility of elymus nutans grassland under alpine ecological conditions. *Chinese Journal of Grassland*, *5*, 50–57.

Nadelhoffer, K. J., Downs, M. R., & Fry, B. (1999). Sinks for 15N-enriched additions to an oak Forest and a red pine plantation. *Ecological Applications*, *9*, 72–86.

Neff, J. C., Townsend, A. R., Gleixner, G., Lehman, S. J., Turnbull, J., & Bowman, W. D. (2002). Variable effects of nitrogen additions on the stability and turnover of soil carbon. *Nature*, *419*, 915–917.

Ning, C., Mueller, G. M., Egerton-Warburton, L. M., Wilson, A. W., Yan, W., & Xiang, W. (2018). Diversity and enzyme activity of ectomycorrhizal fungal communities following nitrogen fertilization in an urban-adjacent pine plantation. *Forests*, *9*, 99.

Niu, G., Hasi, M., Wang, R., Wang, Y., Geng, Q., Hu, S., Xu, X., Yang, J., Wang, C., Han, X., & Huang, J. (2021). Soil microbial community responses to long-term nitrogen addition at different soil depths in a typical steppe. *Applied Soil Ecology*, *167*, 104054.

Ochoa-Hueso, R., Bell, M. D., & Manrique, E. (2014). Impacts of increased nitrogen deposition and altered precipitation regimes on soil fertility and functioning in semiarid Mediterranean shrublands. *Journal of Arid Environments*, *104*, 106–115.

Pastore, M. A., Hobbie, S. E., & Reich, P. B. (2021). Sensitivity of grassland carbon pools to plant diversity, elevated CO_2, and soil nitrogen addition over 19 years. *Proceedings of the National Academy of Sciences of the United States of America*, *118*, e2016965118.

Peng, C., Li, Q., Zhang, Z., Wu, Z., Song, X., Zhou, G., & Song, X. (2019). Biochar amendment changes the effects of nitrogen deposition on soil enzyme activities in a Moso bamboo plantation. *Journal of Forest Research*, *24*, 275–284.

Poeplau, C., Zopf, D., Greiner, B., Geerts, R., Korvaar, H., Thumm, U., Don, A., Heidkamp, A., & Flessa, H. (2018). Why does mineral fertilization increase soil carbon stocks in temperate grasslands? *Agriculture, Ecosystems & Environment*, *265*, 144–155.

Pregitzer, K. S., Burton, A. J., Zak, D. R., & Talhelm, A. F. (2008). Simulated chronic nitrogen deposition increases carbon storage in Northern Temperate forests. *Global Change Biology*, *14*, 142–153.

Reid, J. P., Adair, E. C., Hobbie, S. E., & Reich, P. B. (2012). Biodiversity, nitrogen deposition, and CO_2 affect grassland soil carbon cycling but not storage. *Ecosystems*, *15*, 580–590.

Riggs, C. E., Hobbie, S. E., Bach, E. M., Hofmockel, K. S., & Kazanski, C. E. (2015). Nitrogen addition changes grassland soil organic matter decomposition. *Biogeochemistry*, *125*, 203–219.

Sager, E. P., & Hutchinson, T. C. (2005). The effects of UV-B, nitrogen fertilization, and springtime warming on sugar maple seedlings and the soil chemistry of two central Ontario forests. *Canadian Journal of Forest Research*, *35*, 2432–2446.

Shen, F., Wu, J., Fan, H., Liu, W., Guo, X., Duan, H., Hu, L., Lei, X., & Wei, X. (2019). Soil N/P and C/P ratio regulate the responses of soil microbial community composition and enzyme activities in a long-term nitrogen loaded Chinese fir forest. *Plant and Soil*, *436*, 91–107.

Shi, Y., Sheng, L., Wang, Z., Zhang, X., He, N., & Yu, Q. (2016). Responses of soil enzyme activity and microbial community compositions to nitrogen addition in bulk and microaggregate soil in the temperate steppe of Inner Mongolia. *Eurasian Soil Science*, *49*, 1149–1160.

Silver, W. L., Thompson, A. W., Reich, A., Ewel, J. J., & Firestone, M. K. (2005). Nitrogen cycling in tropical plantation forests: Potential controls on nitrogen retention. *Ecological Applications*, *15*, 1604–1614.

Sirulnik, A. G., Allen, E. B., Meixner, T., Fenn, M. E., & Allen, M. F. (2007). Changes in N cycling and microbial N with elevated N in exotic annual grasslands of southern California. *Applied Soil Ecology*, *36*, 1–9.

Song, B., Li, Y., Yang, L., Shi, H., Li, L., Bai, W., & Zhao, Y. (2023). Soil acidification under long-term N addition decreases the diversity of soil bacteria and fungi and changes their community composition in a semiarid grassland. *Microbial Ecology*, *85*, 221–231.

Song, C., Liu, D., Song, Y., & Mao, R. (2013). Effect of nitrogen addition on soil organic carbon in freshwater marsh of Northeast China. *Environmental Earth Sciences*, *70*, 1653–1659.

Song, K., Wang, X., Xu, D., Li, Y., Sa, C., & Ma, S. (2022). Effects of short-term nitrogen addition on soil biological properties in desert steppe. *Journal of Soil and Water Conservation*, *3*, 303–310.

Song, Y., Song, C., Li, Y., Hou, C., Yang, G., & Zhu, X. (2013). Short-term effects of nitrogen addition and vegetation removal on soil chemical and biological properties in a freshwater marsh in Sanjiang Plain, Northeast China. *Catena*, *104*, 265–271.

Stursova, M., Crenshaw, C. L., & Sinsabaugh, R. L. (2006). Microbial responses to long-term N deposition in a semiarid grassland. *Microbial Ecology*, *51*, 90–98.

Su, J., Li, X., & Bao, J. (2014). Effects of nitrogen addition on soil physico-chemical properties and enzyme activities in desertified steppe. *The Journal of Applied Ecology*, *25*, 664–670.

Su, Z., Zeng, F., & Zheng, C. (2022). Effects of nitrogen addition on soil organic carbon and soil respiration in subtropical evergreen broad-leaved forest. *Beijing Da Xue Xue Bao. Yi Xue Ban*, *58*, 517–525.

Sun, X., Zhao, J., You, Y., & Jianxin, S. O. (2016). Soil microbial responses to forest floor litter manipulation and nitrogen addition in a mixed-wood forest of northern China. *Scientific Reports*, *6*, 19536.

Thirukkumaran, C. M., & Parkinson, D. (2002). Microbial activity, nutrient dynamics and litter decomposition in a Canadian Rocky Mountain pine forest as affected by N and P fertilizers. *Forest Ecology and Management*, *159*, 187–201.

Tian, D., Jiang, L., Ma, S., Fang, W., Schmid, B., Xu, L., Zhu, J., Li, P., Losapio, G., Jing, X., Zheng, C., Shen, H., Xu, X., Zhu, B., & Fang, J. (2017). Effects of nitrogen deposition on soil microbial communities in temperate and subtropical forests in China. *Science of the Total Environment*, *607–608*, 1367–1375.

Tian, X., Hu, H., Ding, Q., Song, M., Xu, X., Zheng, Y., & Guo, L. (2014). Influence of nitrogen fertilization on soil ammonia oxidizer and denitrifier abundance, microbial biomass, and enzyme activities in an alpine meadow. *Biology and Fertility of Soils*, *50*, 703–713.

Torn, M. S., Vitousek, P. M., & Trumbore, S. E. (2005). The influence of nutrient availability on soil organic matter turnover estimated by incubations and radiocarbon modeling. *Ecosystems*, *8*, 352–372.

Tu, L., Chen, G., Peng, Y., Hu, H., Hu, T., Zhang, J., Li, X., Liu, L., & Tang, Y. (2014). Soil biochemical responses to nitrogen addition in a bamboo forest. *PLoS ONE*, *9*, e102315.

Tu, L., Peng, Y., Chen, G., Hu, H., Xao, Y., Hu, T., Liu, L., & Tang, Y. (2015). Direct and indirect effects of nitrogen additions on fine root decomposition in a subtropical bamboo forest. *Plant and Soil*, *389*, 273–288.

Ulm, F., Gouveia, C., Dias, T., & Cruz, C. (2017). N fertilization in a Mediterranean ecosystem alters N and P turnover in soil, roots and the ectomycorrhizal community. *Soil Biology and Biochemistry*, *113*, 60–70.

Verma, P., & Sagar, R. (2022). Soil respiration response to nitrogen fertilization experiment in tropical grassland. *Ecological Research*, *37*, 390–405.

Vourlitis, G. L., Kirby, K., Vallejo, I., Asaeli, J., & Holloway, J. M. (2021). Potential soil extracellular enzyme activity is altered by long-term experimental nitrogen deposition in semiarid shrublands. *Applied Soil Ecology*, *158*, 103779.

Waldrop, M. P., Zak, D. R., & Sinsabaugh, R. L. (2004). Microbial community response to nitrogen deposition in northern forest ecosystems. *Soil Biology and Biochemistry*, *36*, 1443–1451.

Waldrop, M. P., Zak, D. R., Sinsabaugh, R. L., Gallo, M., & Lauber, C. (2004). Nitrogen deposition modifies soil carbon storage through changes in microbial enzymatic activity. *Ecological Applications*, *14*, 1172–1177.

Wallenstein, M. D., McNulty, S., Fernandez, I. J., Boggs, J., & Schlesinger, W. H. (2006). Nitrogen fertilization decreases forest soil fungal and bacterial biomass in three long-term experiments. *Forest Ecology and Management*, *222*, 459–468.

Wang, C., Lu, X., Mori, T., Mao, Q., Zhou, K., Zhou, G., Nie, Y., & Mo, J. (2018). Responses of soil microbial community to continuous experimental nitrogen additions for 13 years in a nitrogen-rich tropical forest. *Soil Biology and Biochemistry*, *121*, 103–112.

Wang, C., Shi, B., Sun, W., & Guan, Q. (2020). Different forms and rates of nitrogen addition show variable effects on the soil hydrolytic enzyme activities in a meadow steppe. *Soil Research*, *58*, 258–267.

Wang, D., He, H. L., Gao, Q., Zhao, C. Z., Zhao, W. Q., Yin, C. Y., Chen, X. L., Ma, Z. L., Li, D. D., Sun, D. D., Cheng, X. Y., & Liu, Q. (2017). Effects of short-term N addition on plant biomass allocation and C and N pools of the Sibiraea angustata scrub ecosystem. *European Journal of Soil Science*, *68*, 212–220.

Wang, F., Li, Z., Su, F., Guo, H., Wang, P., Guo, J., Zhu, W., Wang, Y., & Hu, S. (2022). Sensitive groups of bacteria dictate microbial functional responses to short-term warming and N input in a semiarid grassland. *Ecosystems*, *25*, 1346–1357.

Wang, H., Wang, J., Teng, Z., Fan, W., Deng, P., Wen, Z., Zhou, K., & Xu, X. (2022). Nitrogen and phosphorus additions impact stability of soil organic carbon and nitrogen in subtropical evergreen broad-leaved forest. *Eurasian Soil Science*, *55*, 425–436.

Wang, H., Wu, J., Li, G., Yan, L., Wei, X., & Ma, W. (2022). Effects of simulated nitrogen deposition on soil active carbon fractions in a wet meadow in the Qinghai-Tibet plateau. *Journal of Soil Science and Plant Nutrition*, *22*, 2943–2954.

Wang, J., Wu, L., Zhang, C., Zhao, X., Bu, W., & Gadow, K. V. (2016). Combined effects of nitrogen addition and organic matter manipulation on soil respiration in a Chinese pine forest. *Environmental Science and Pollution Research*, *23*, 22701–22710.

Wang, Q., Chen, L., Yang, Q., Sun, T., & Li, C. (2019). Different effects of single versus repeated additions of glucose on the soil organic carbon turnover in a temperate forest receiving long-term N addition. *Geoderma*, *341*, 59–67.

Wang, Q., Wang, S., & Liu, Y. (2008). Responses to N and P fertilization in a young *Eucalyptus dunnii* plantation: Microbial properties, enzyme activities and dissolved organic matter. *Applied Soil Ecology*, *40*, 484–490.

Wang, Q., Zheng, Y., Song, G., Jin, S., & He, J. (2021). Impacts of simulated nitrogen and phosphorus depositions on soil microbial biomass and soil nutrients along two secondary succession stages in a subtropical forest. *Acta Ecologica Sinica*, *41*, 6245–6256.

Wang, R., Dijkstra, F. A., Liu, H., Yin, J., Wang, X., Feng, X., Xu, Z., & Jiang, Y. (2019). Response of soil carbon to nitrogen and water addition differs between labile and recalcitrant fractions: Evidence from multi–year data and different soil depths in a semi-arid steppe. *Catena*, *172*, 857–865.

Wang, R., Filley, T. R., Xu, Z., Wang, X., Li, M.-H., Zhang, Y., Luo, W., & Jiang, Y. (2014). Coupled response of soil carbon and nitrogen pools and enzyme activities to nitrogen and water addition in a semi-arid grassland of Inner Mongolia. *Plant and Soil*, *381*, 323–336.

Wang, Y., Cheng, S., Yu, G., Fang, H., Mo, J., Xu, M., & Gao, W. (2015). Response of carbon utilization and enzymatic activities to nitrogen deposition in three forests of subtropical China. *Canadian Journal of Forest Research*, *45*, 394–401.

Wang, Y., Zheng, M., Wang, S., Mao, J., & Mo, J. (2021). Effects of long-term nitrogen and phosphorus additions on soil enzyme activities related N and P cycle in two plantations in south China. *Journal of Tropical and Subtropical Botany*, *29*, 244–250.

Wang, Z., Wang, Z., Li, T., Wang, C., Dang, N., Wang, R., Jiang, Y., Wang, H., & Li, H. (2021). N and P fertilization enhanced carbon decomposition function by shifting microbes towards an r-selected community in meadow grassland soils. *Ecological Indicators*, *132*, 108306.

Wang, Z., Yang, S., Wang, R., Xu, Z., Feng, K., Feng, X., Li, T., Liu, H., Ma, R., Li, H., & Jiang, Y. (2020). Compositional and functional responses of soil microbial communities to long-term nitrogen and phosphorus addition in a calcareous grassland. *Pedobiologia*, *78*, 150612.

Weber, C. F., Vilgalys, R., & Kuske, C. R. (2013). Changes in fungal community composition in response to elevated atmospheric CO_2 and nitrogen fertilization varies with soil horizon. *Frontiers in Microbiology*, *4*, 78.

Weng, X., Wang, M., Sui, X., Frey, B., Liu, Y., Zhang, R., Ni, H., & Li, M. (2023). High ammonium addition changes the diversity and structure of bacterial communities in temperate wetland soils of northeastern China. *Microorganisms*, *11*, 2033.

Widdig, M., Schleuss, P., Weig, A. R., Guhr, A., Biederman, L. A., Borer, E. T., Crawley, M. J., Kirkman, K. P., Seabloom, E. W., Wragg, P. D., et al. (2019). Nitrogen and phosphorus additions alter the abundance of phosphorus-solubilizing bacteria and phosphatase activity in grassland soils. *Frontiers in Environmental Science*, *7*, 785.

Widdig, M., Schleuss, P.-M., Biederman, L. A., Borer, E. T., Crawley, M. J., Kirkman, K. P., Seabloom, E. W., Wragg, P. D., & Spohn, M. (2020). Microbial carbon use efficiency in grassland soils subjected to nitrogen and phosphorus additions. *Soil Biology and Biochemistry*, *146*, 107815.

Wu, J., Liu, W., Fan, H., Huang, G., Wan, S., Yuan, Y., & Ji, C. (2013). Asynchronous responses of soil microbial community and understory plant community to simulated nitrogen deposition in a subtropical forest. *Ecology and Evolution*, *3*, 3895–3905.

Wu, T., Schoenau, J. J., Li, F., Qian, P., Malhi, S. S., & Shi, Y. (2005). Influence of fertilization and organic amendments on organic-carbon fractions in Heilu soil on the loess plateau of China. *Journal of Plant Nutrition and Soil Science*, *168*, 100–107.

Wu, Y., Sen, Y., Xin, W., Jun-Sheng, H., Bin, W., Liu, W.-X., & Liu, L.-L. (2021). Responses of soil nitrogen in different soil organic matter fractions to long-term nitrogen addition in a semi-arid grassland. *Chinese Journal of Plant Ecology*, *45*, 790–798.

Xiao, H., Yang, H., Zhao, M., Monaco, T. A., Rong, Y., Huang, D., Song, Q., Zhao, K., & Wang, D. (2021). Soil extracellular enzyme activities and the abundance of nitrogen-cycling functional genes responded more to N addition than P addition in an inner Mongolian meadow steppe. *Science of the Total Environment*, *759*, 143541.

Xiong, L., Deng, X., Ji, L., & Wang, J. (2022). Effects of nitrogen deposition on carbon storage of Alnus cremastogyne ecosystem was simulated with different frequency of nitrogen addition. *Ecological Science*, *5*, 28–34.

Xu, R., Wang, Z., & Zhu, J. (2023). Effects of climate change and nitrogen addition on carbon loss in alpine wetland of Qinghai–Tibet plateau. *Atmosphere*, *14*, 1342.

Xu, Z., Ren, H., Li, M.-H., Brunner, I., Yin, J., Liu, H., Kong, D., Lü, X., Sun, T., Cai, J., et al. (2017). Experimentally increased water and nitrogen affect root production and vertical allocation of an old-field grassland. *Plant and Soil*, *412*, 369–380.

Yan, G., Chen, F., Zhang, X., Wang, J., Han, S., Xing, Y., & Wang, Q. (2017). Spatial and temporal effects of nitrogen addition on root morphology and growth in a boreal forest. *Geoderma*, *303*, 178–187.

Yang, J., Wang, Y., Zhou, Z., & Shen, Y. (2023). Soil microbial nitrogen use efficiency and its influencing factors under inorganic nitrogen supplementation in *Pinus tabulaeformis* forest of Taiyue mountain, Shanxi province. *Journal of Northeast Forestry University*, *8*, 40–47.

Yang, N., Wang, B., Liu, D., Wang, X., Li, X., Zhang, Y., Xu, Y., Peng, S., Ge, Z., Mao, L., Ruan, H., & Pena, R. (2021). Long-term nitrogen deposition alters ectomycorrhizal community composition and function in a poplar plantation. *Journal of Fungi*, *7*, 791.

Yang, X., Duan, P., Wang, K., & Li, D. (2023). Topography modulates effects of nitrogen deposition on soil nitrogen transformations by impacting soil properties in a subtropical forest. *Geoderma*, *432*, 116381.

Ye, Y., Liu, Y., Han, Y., Shao, X., Tang, D., & Yang, K. (2017). Short-term effects of nitrogen deposition on soil physical and chemical properties of alpine shrub meadow in Tibet. *Acta Agrestia Sinica*, *25*, 973.

Yixiong, W., Huafeng, Z., Quan, L. I., Junbo, Z., Shaoliang, W., & Xinzhang, S. (2022). Effect of nitrogen addition on soil phosphorus fractions in the *Phyllostachys edulis* plantation. *Journal of Zhejiang A & F University*, *39*, 695–704.

Yuan, X., Cui, J., Lin, K., Liu, C., Zhou, J., Zhang, Q., Zeng, Q., Wu, L., Wu, Y., Mei, K., et al. (2022). Effects of nitrogen addition on the concentration and composition of soil-based dissolved organic matter in subtropical *Pinus taiwanensis* forests. *Journal of Soils and Sediments*, *22*, 1924–1937.

Yuan, X., Si, Y., Lin, W., Yang, J., Wang, Z., Zhang, Q., Qian, W., Chen, Y., & Yang, Y. (2018). Effects of short-term warming and nitrogen addition on the quantity and quality of dissolved organic matter in a subtropical Cunninghamia lanceolata plantation. *PLoS ONE*, *13*, e0191403.

Yue, P., Zhang, J., Zhu, G., Yin, X., Zhang, X., Wang, S., Müller, C., Misselbrook, T., & Zuo, X. (2023). Impact of nitrogen and phosphorus additions on soil gross nitrogen transformations in a temperate desert steppe. *European Journal of Soil Science*, *74*, e13416.

Zeglin, L. H., Stursova, M., Sinsabaugh, R. L., & Collins, S. L. (2007). Microbial responses to nitrogen addition in three contrasting grassland ecosystems. *Oecologia*, *154*, 349–359.

Zeng, Q., Zhang, Q., Lin, K., Zhou, J., Yuan, X., Mei, K., Wu, Y., Cui, J., Xu, J., & Chen, Y. (2021). Enzyme stoichiometry evidence revealed that five years nitrogen addition exacerbated the carbon and phosphorus limitation of soil microorganisms in a *Phyllostachys pubescens* forest. *Chinese Journal of Applied Ecology*, *2*, 521–528.

Zhang, C., Yang, Z., Shen, J., Sun, Y., Wang, J., Han, H., Wan, S., Zhang, L., & He, J. (2018). Impacts of long-term nitrogen addition, watering and mowing on ammonia oxidizers, denitrifiers and plant communities in a temperate steppe. *Applied Soil Ecology*, *130*, 241–250.

Zhang, J., Ai, Z., Liang, C., Wang, G., Liu, G., & Xue, S. (2019). How microbes cope with short-term N addition in a *Pinus tabuliformis* forest-ecological stoichiometry. *Geoderma*, *337*, 630–640.

Zhang, J., Ru, J., Song, J., Li, H., Li, X., Ma, Y., Li, Z., Hao, Y., Chi, Z., Hui, D., & Wan, S. (2022). Increased precipitation and nitrogen addition accelerate the temporal increase in soil respiration during 8-year old-field grassland succession. *Global Change Biology*, *28*, 3944–3959.

Zhang, J., Zhou, J., Sayer, E. J., Lambers, H., Liu, Z., Lu, X., Li, Y., Li, Y., Li, H., & Wang, F. (2023). Nitrogen deposition enhances soil organic carbon and microbial residual carbon in a tropical forest. *Plant and Soil*, *484*, 217–235.

Zhang, Q., Zhou, J., Li, X., Zheng, Y., Xie, L., Yang, Z., Liu, X., Xu, C., Lin, H., Yuan, X., Liu, C., Zhu, B., Chen, Y., & Yang, Y. (2022). Contrasting effects of warming and N deposition on soil microbial functional genes in a subtropical forest. *Geoderma*, *408*, 115588.

Zhang, R., Liu, Y., Zhong, H., Chen, X., & Sui, X. (2022). Effects of simulated nitrogen deposition on the soil microbial community diversity of a *Deyeuxia angustifolia* wetland in the Sanjiang Plain, Northeastern China. *Annals of Microbiology*, *72*, 1–13.

Zhao, G., Chen, Y., Zhang, Y., Cong, N., Zheng, Z., Zhu, J., & Chen, N. (2022). Decoupling of plant carbon and nitrogen under elevated CO_2 and nitrogen addition in a typical alpine ecosystem. *Plant and Soil*, *474*, 485–498.

Zhao, L., Zhang, L., Yi, H., Lan, S., Chen, L., Lu, F., & Han, G. (2021). The short-term effects of nitrogen addition on the growth of *Phragmites australis* and soil physical and chemical properties in a high marsh of the Yellow River Delta. *Ecological Science*, *2*, 18–25.

Zheng, L., Zhao, Q., Lin, G., Hong, X., & Zeng, D. (2023). Nitrogen addition impacts on soil phosphorus transformations depending upon its influences on soil organic carbon and microbial biomass in temperate larch forests across northern China. *Catena*, *230*, 107252.

Zhu, H., Zhao, J., & Gong, L. (2021). The morphological and chemical properties of fine roots respond to nitrogen addition in a temperate Schrenk's spruce (*Picea schrenkiana*) forest. *Scientific Reports*, *11*, 3839.

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Hu, Y., Deng, Q., Kätterer, T., Olesen, J. E., Ying, S. C., Ochoa-Hueso, R., Mueller, C. W., Weintraub, M. N., & Chen, J. (2024). Depth-dependent responses of soil organic carbon under nitrogen deposition. *Global Change Biology*, *30*, e17247. https://doi.org/10.1111/gcb.17247