

Supplementary Information

Title: Soil organic carbon thresholds control fertilizer effects on carbon accrual in croplands worldwide

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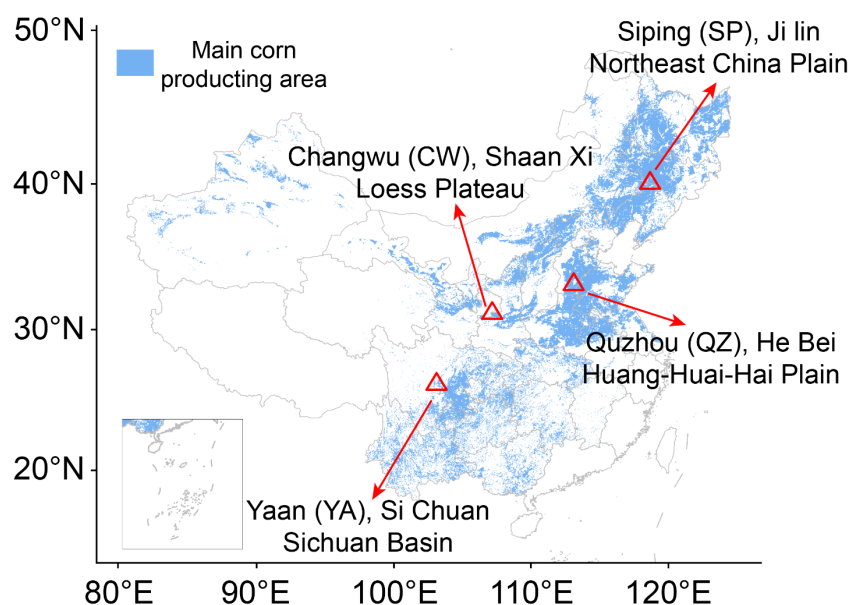
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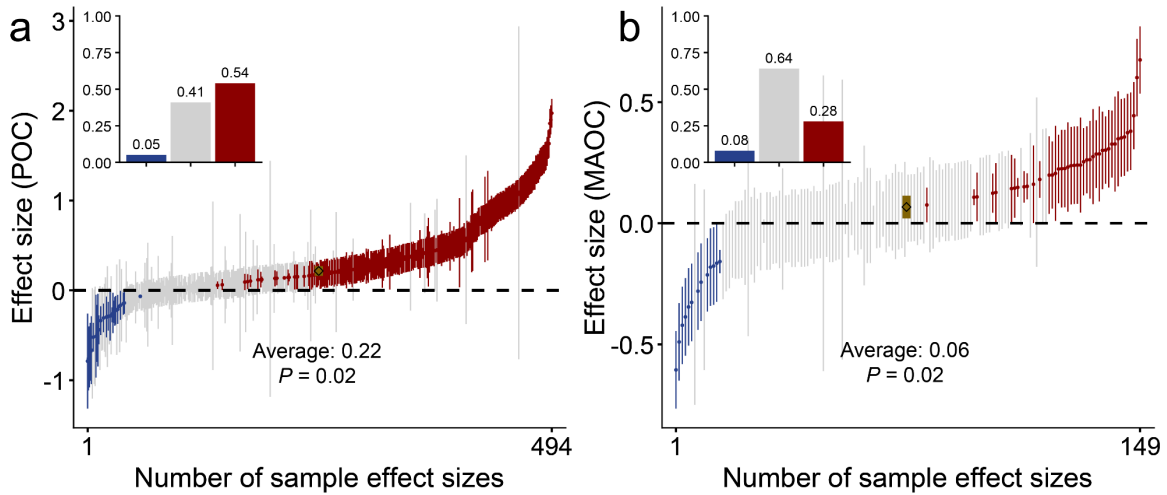
Supplementary Figure 1 to 18

Supplementary Table 1 to 8



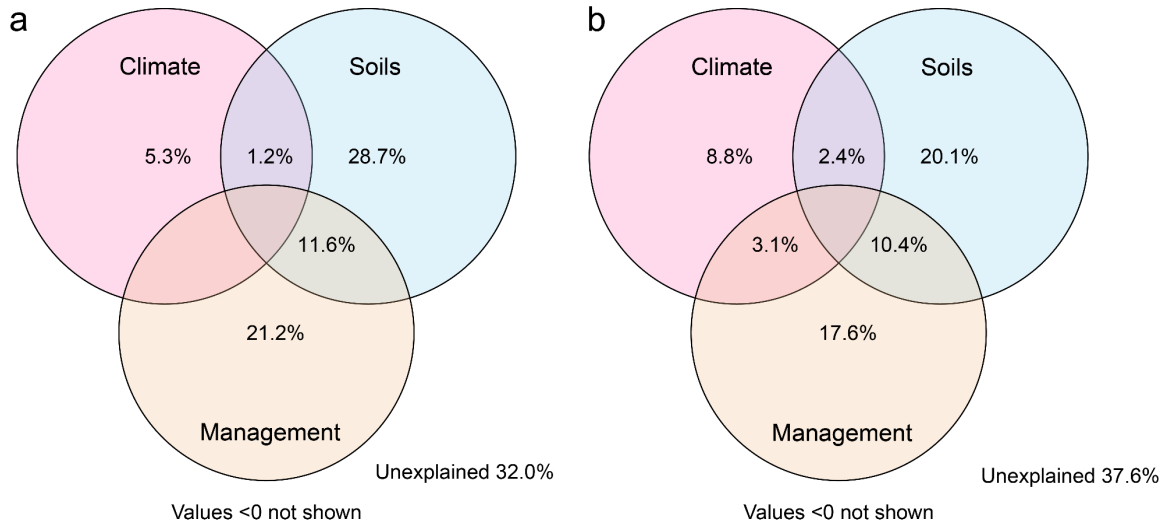
Supplementary Figure 1. Detailed information and enlarged distribution of four nitrogen (N) fertilizer network field experiments across China.

Base map data adapted from GS(2020)4619, <http://bzdt.ch.mnr.gov.cn/>. The maize distribution in China from <https://doi.org/10.57760/sciencedb.08490>¹. Map created using QGIS software.



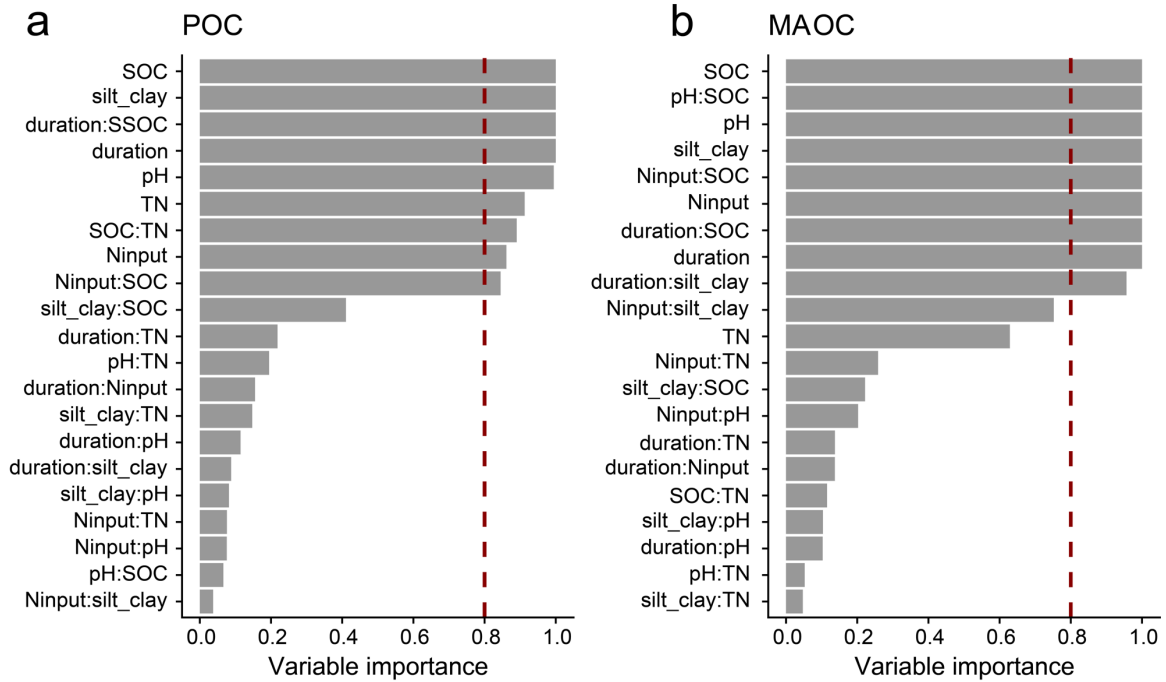
Supplementary Figure 2. Sample effect sizes of POC (a) and MAOC (b) in responding to N addition.

The heights of bar plots are relative proportions of negative (blue), neutral (grey) and positive (red) (CIs crossing zero) effect sizes, and were standardized, ranging from 0 to 1. POC: particulate organic carbon; MAOC: mineral-associated organic carbon. Circles and error bars represent average parameter estimates and 95% confidence intervals (CIs) in the linear mixed effects models (two-sided). The vertical dashed line represents the warming effect size = 0. If the 95% CI did not overlap zero, the effect of warming was statistically significant ($P < 0.05$).



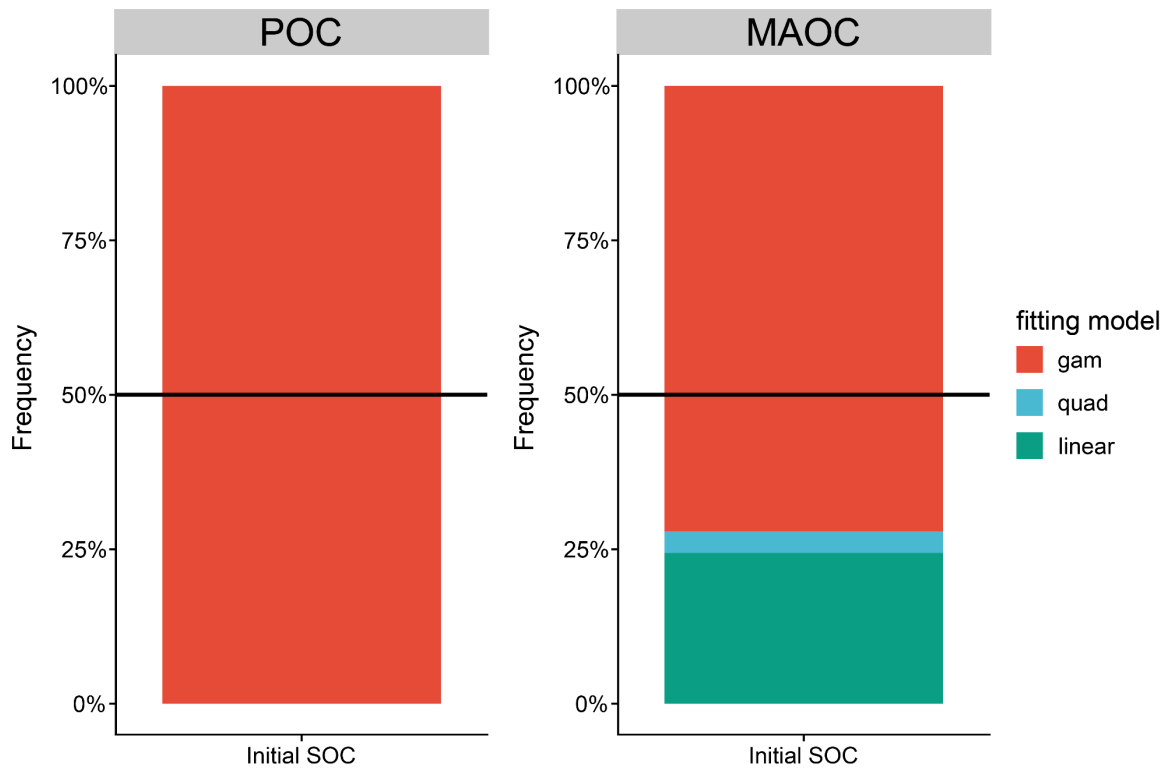
Supplementary Figure 3. Variation partitioning analysis (VPA) of the POC (a) and MAOC (b) contents explained by climatic, soils variables and management practices and their interactions.

Climatic factors: mean annual temperature and mean annual precipitation; Soils variables: initial soil organic carbon, initial soil total nitrogen, initial soil pH and initial silt and clay. Management: nitrogen fertilizer uses and experimental duration. Unexplained represents the variance of changes that are not captured by the Variance Partitioning Analysis.



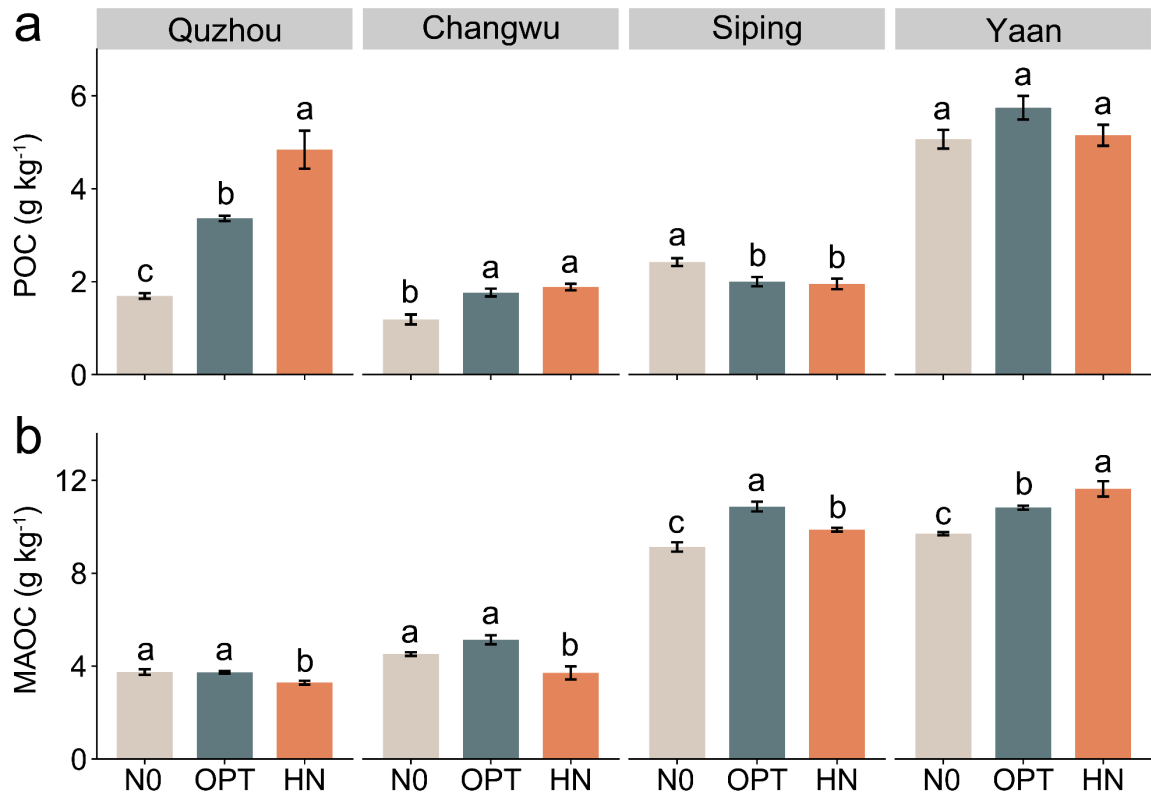
Supplementary Figure 4. Model-averaged importance of the predictors of the effects N addition on POC (a) and MAOC (b) contents.

The importance value is based on the sum of Akaike weights derived from model selection using corrected Akaike's information criteria. Cutoff is set at 0.8 to differentiate between essential and nonessential predictors. POC: particulate organic carbon; MAOC: mineral-associated organic carbon. SOC: soil organic carbon; TN: total nitrogen; N_{input}: N fertilizer use. All soil properties were the initial soil conditions before experiment. The ':' denotes the interaction between factors.



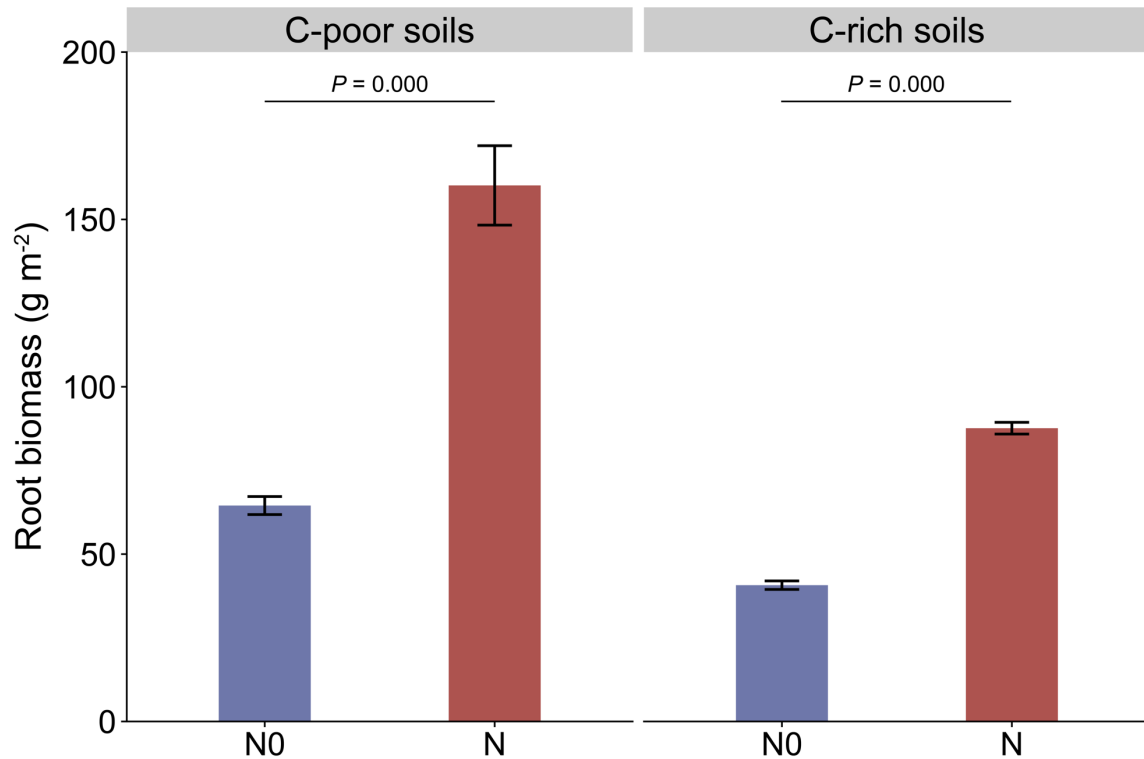
Supplementary Figure 5. Evaluation of linear and nonlinear responses of POC and MAOC contents to initial SOC based on Meta-analysis data.

Akaike information criterion (AIC) was used to decide the model that provided the best fit in each case, and the most likely model has the lowest AIC value. We chose quadratic to synthesize the simplest case of nonlinear trend, and GAM to summarize more complex trends. This comparison was conducted 1000 times by bootstrapping samples and a linear regression was a better fit over the nonlinear relationship only when the linear part ranked above half frequency spotted by horizontal black lines in each facet. SOC: soil organic carbon; POC: particulate organic carbon; MAOC: mineral-associated organic carbon.



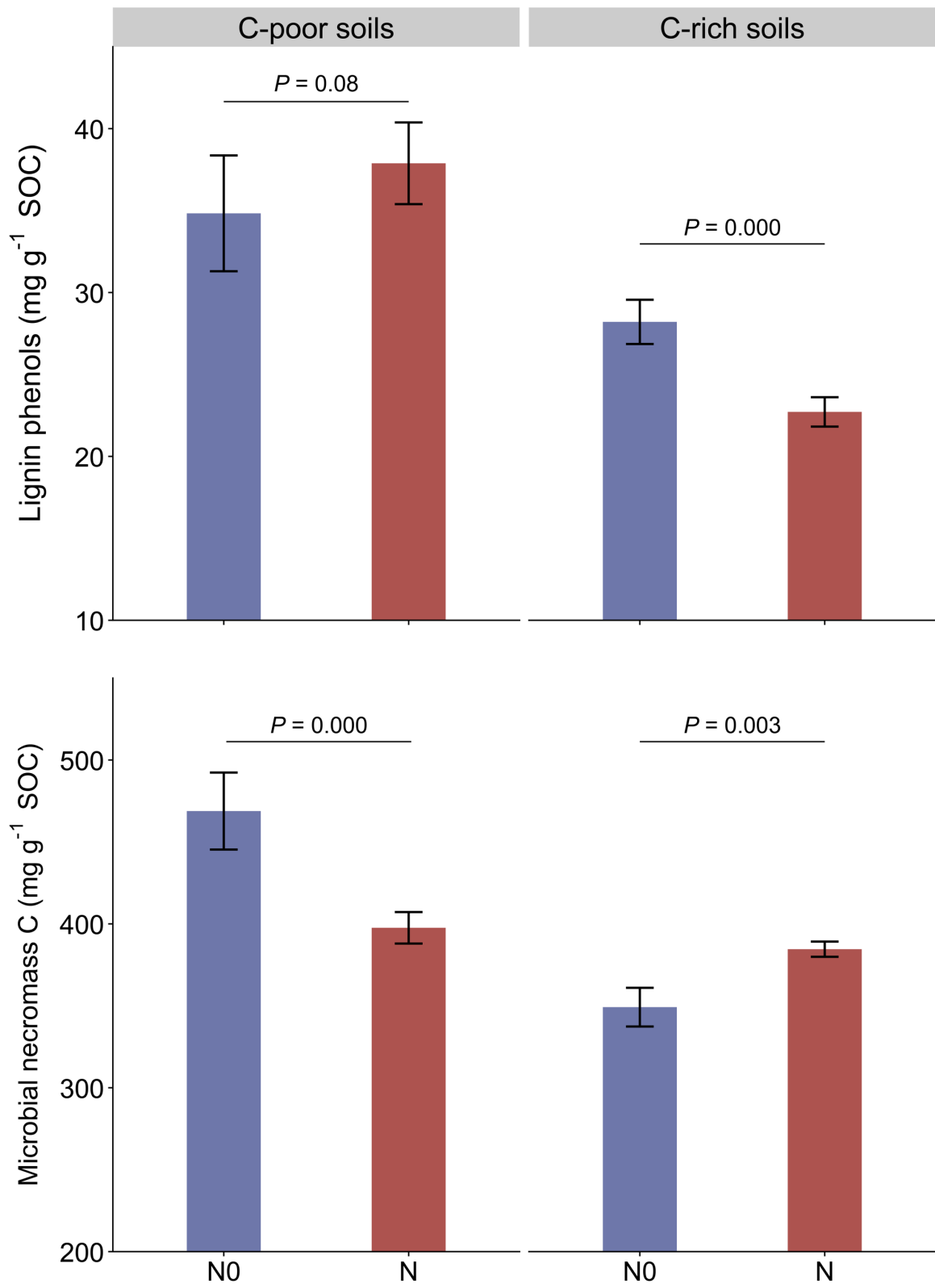
Supplementary Figure 6. Effects of N fertilizer on POC (a) and MAOC contents (b) across four sites.

Data show mean \pm standard error ($n = 4$). Different lowercase letters indicate significant difference among treatments ($P < 0.05$) based on two-sided tests for multiple comparisons (Tukey-post-hoc test). N0: no N fertilizer; OPT: optimized N fertilizer; HN: high nitrogen fertilizer. POC: particulate organic carbon; MAOC: mineral-associated organic carbon.



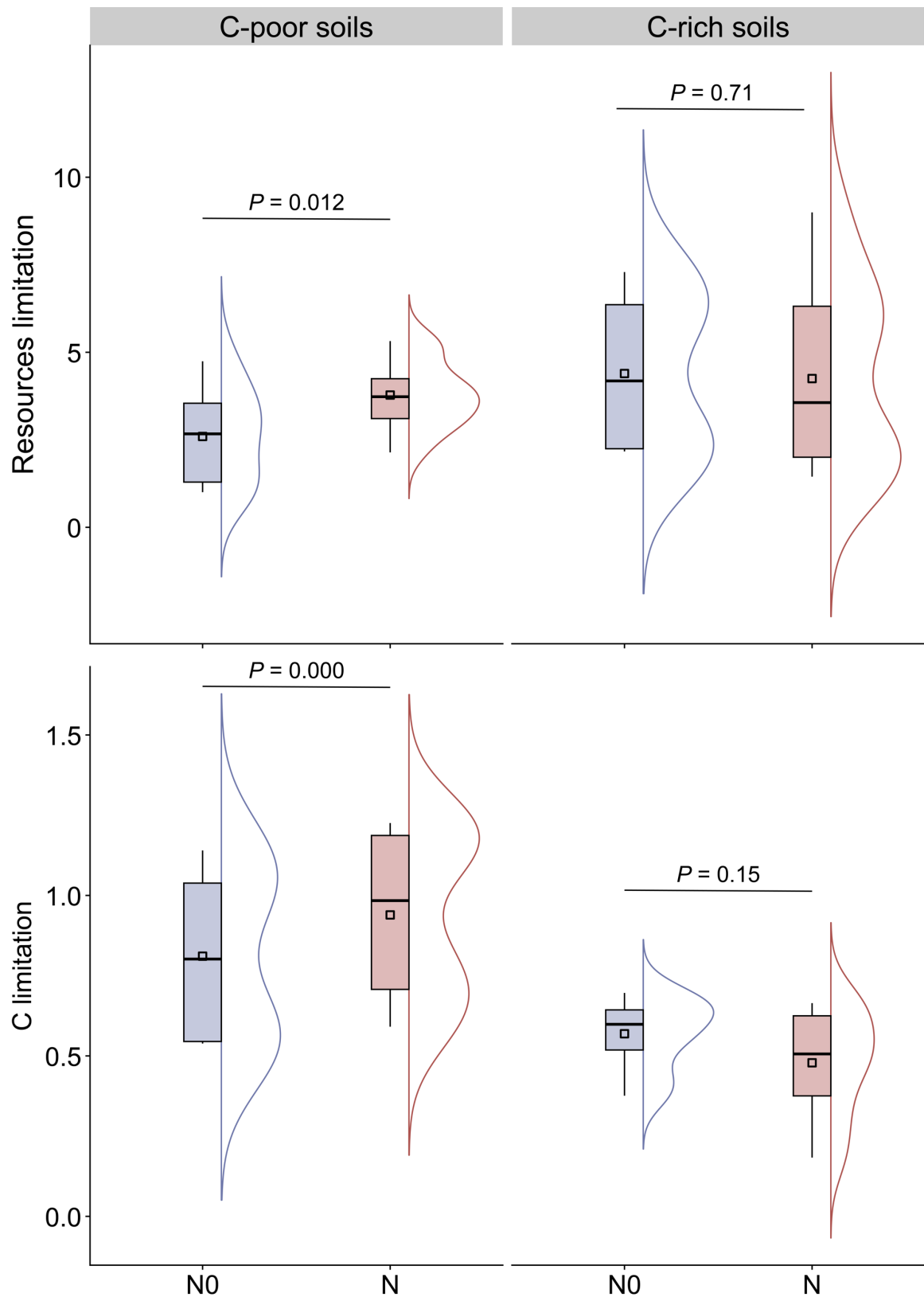
Supplementary Figure 7. Changes of root biomass under different N fertilizer treatments between C-poor and C-rich soils.

Data show mean \pm standard error ($n = 4$ for N0, $n = 8$ for N). A mixed linear model was used to test the treatment effect. N0: no N addition; N: N addition. C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. C levels are categorized according to the organic carbon threshold (i.e., SOC = 15 g kg⁻¹) determined by the global meta-analysis. A mixed linear model was used to test the treatment effect at $P = 0.05$.



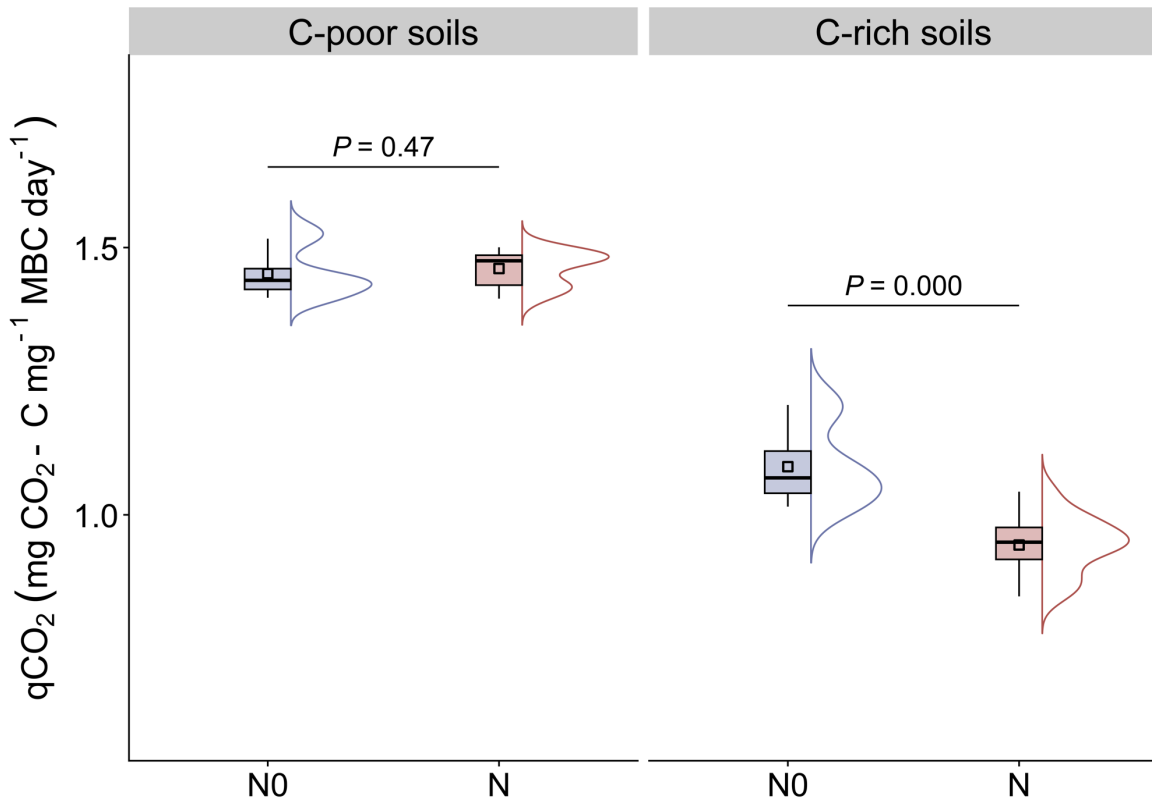
Supplementary Figure 8. Changes of microbial necromass C and lignin phenols under different N fertilizer treatments between C-poor and C-rich soils.

Data show mean \pm standard error ($n = 4$ for N0, $n = 8$ for N). A mixed linear model was used to test the treatment effect. N0: no N addition; N: N addition. C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. C levels are categorized according to the organic carbon threshold (i.e., SOC = 15 g kg⁻¹) determined by the global meta-analysis. A mixed linear model was used to test the treatment effect at $P = 0.05$.



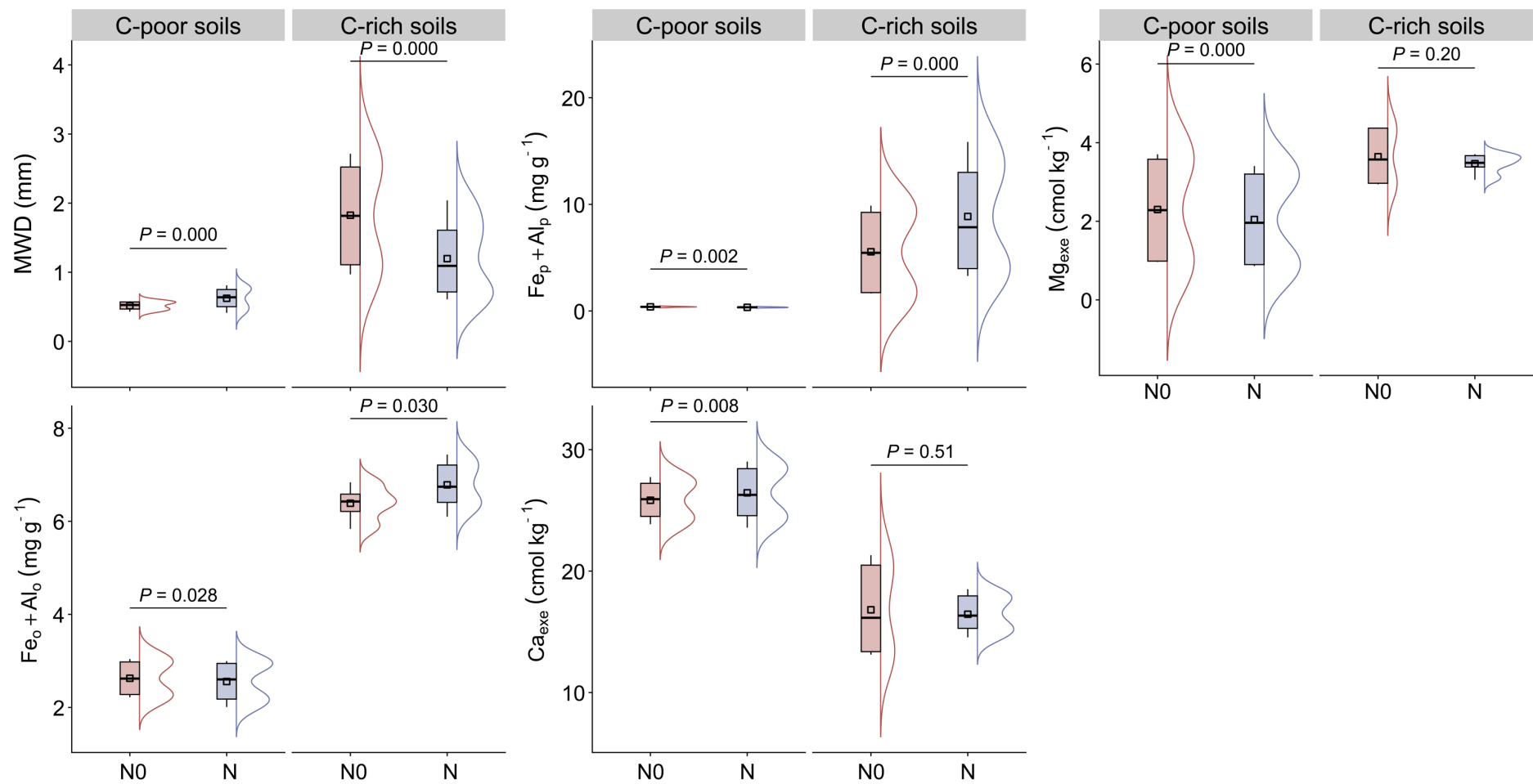
Supplementary Figure 9. Microbial resources and C limitation response to N addition between C-poor and C-rich soils.

The resources limitation is the ratio of the resource C:N (SOC:TN) ratios normalized to BC:N (MBC:MBN). The C limitation is the square root of the sum of x^2 and y^2 , where x represents the relative activity of C vs. P-acquiring enzymes, and y represents the relative activity of C vs. N-acquiring enzymes. In box plots, the solid lines and squares in the boxes, the lower and upper edges of the boxes and the lower and upper lines extending from the boxes represent median and mean values ($n = 4$ for N0, $n = 8$ for N), Q1 and Q3, $1.5 \times \text{IQR}$ above the box's Q1 and $1.5 \times \text{IQR}$ below the box's Q3 of all data, respectively. A mixed linear model was used to test the treatment effect at $P = 0.05$. SOC: soil organic carbon; TN: total nitrogen; MBC: microbial biomass C; MBN: microbial biomass N; C-acquiring enzymes: the sum of BG (β -1,4-glucosidase) and CBH (cellobiohydrolase); N-acquiring enzymes: the sum of NAG (β -1,4-N-acetylglucosaminidase) and LAP (L-leucine aminopeptidase); N-acquiring enzymes: ALP (alkaline phosphomonoesterase). Asterisks indicate significant differences between N0 and N. N0: no N addition; N: N addition. C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. C levels are categorized according to the organic carbon threshold (i.e., $\text{SOC} = 15 \text{ g kg}^{-1}$) determined by the global meta-analysis.



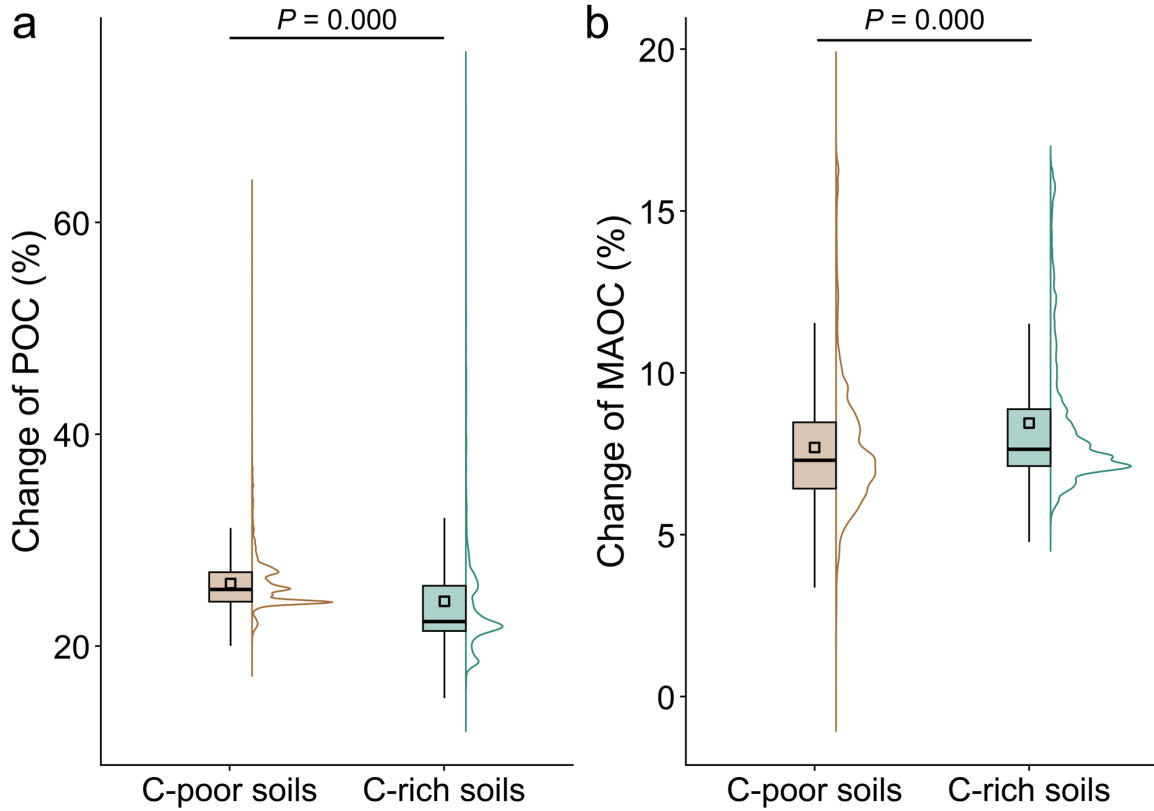
Supplementary Figure 10. Changes of qCO₂ (microbial metabolic quotient) under different N fertilizer treatments between C-poor and C-rich soils.

In box plots, the solid lines and squares in the boxes, the lower and upper edges of the boxes and the lower and upper lines extending from the boxes represent median and mean values ($n = 4$ for N0, $n = 8$ for N), Q1 and Q3, $1.5 \times \text{IQR}$ above the box's Q1 and $1.5 \times \text{IQR}$ below the box's Q3 of all data, respectively. A mixed linear model was used to test the treatment effect at $P = 0.05$. N0: no N addition; N: N addition. C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. C levels are categorized according to the organic carbon threshold (i.e., $\text{SOC} = 15 \text{ g kg}^{-1}$) determined by the global meta-analysis.



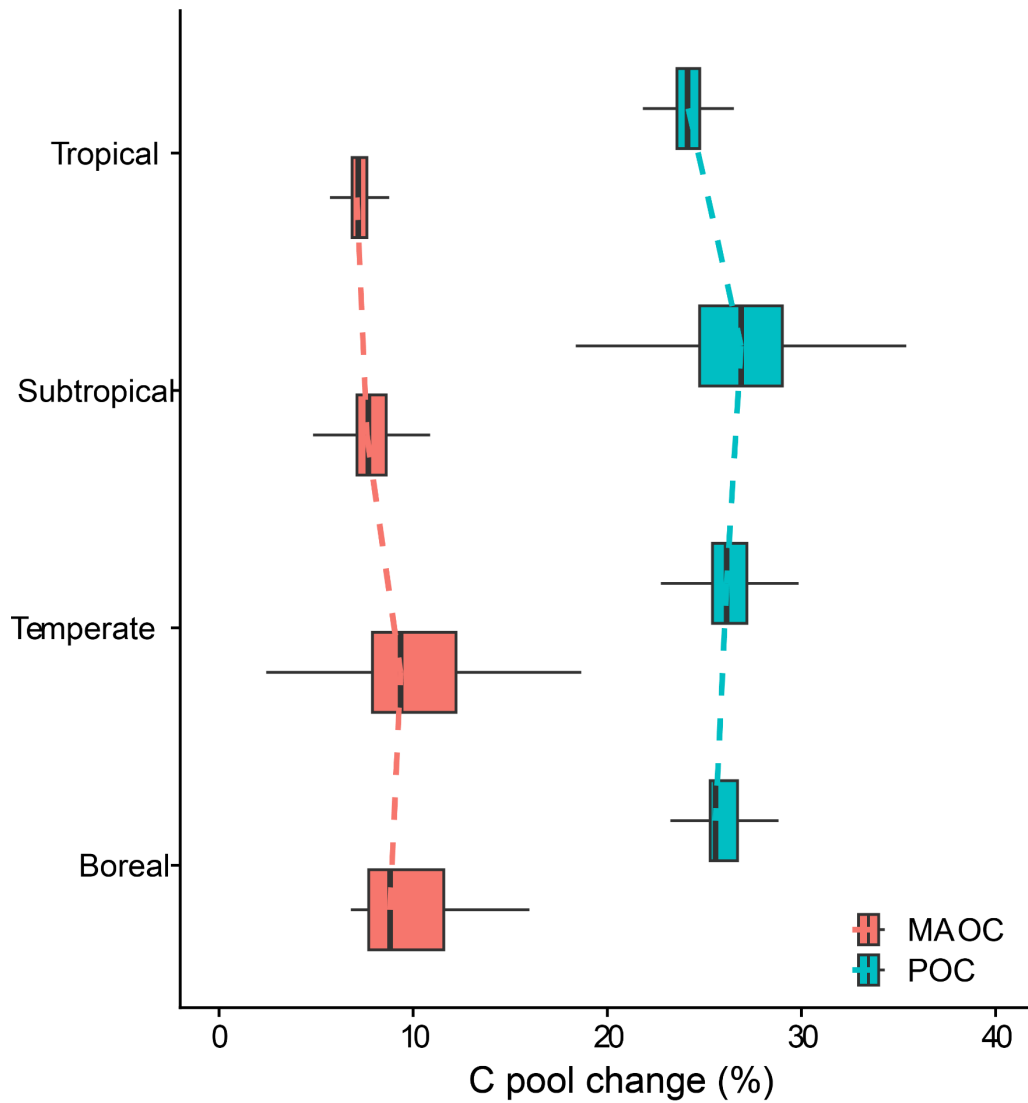
Supplementary Figure 11. Changes of mean weight diameter (MWD) and mineral protection ($\text{Fe}_o + \text{Al}_o$; $\text{Fe}_p + \text{Al}_p$) under different N fertilizer treatments between C-poor and C-rich soils.

In box plots, the solid lines and squares in the boxes, the lower and upper edges of the boxes and the lower and upper lines extending from the boxes represent median and mean values ($n = 4$ for N0, $n = 8$ for N), Q1 and Q3, $1.5 \times \text{IQR}$ above the box's Q1 and $1.5 \times \text{IQR}$ below the box's Q3 of all data, respectively. A mixed linear model was used to test the treatment effect at $P = 0.05$. MWD: mean weight diameter; $\text{Fe}_o + \text{Al}_o$: poorly crystalline Fe/Al oxides; $\text{Fe}_p + \text{Al}_p$: organically complexed Fe/Al oxides. Asterisks indicate significant differences between N0 and N. N0: no N addition; N: N addition. C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. C levels are categorized according to the organic carbon threshold (i.e., $\text{SOC} = 15 \text{ g kg}^{-1}$) determined by the global meta-analysis.



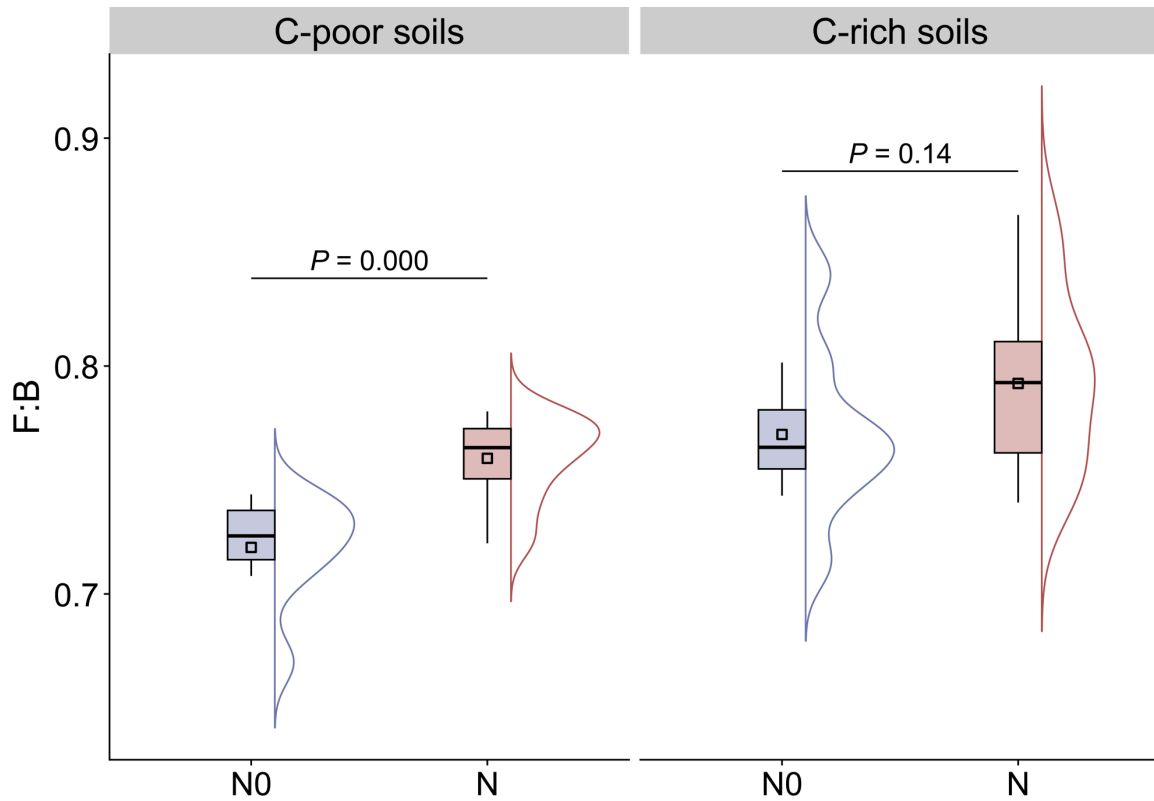
Supplementary Figure 12. Changes in POC (a) and MAOC (b) due to N fertilization in C-poor and C-rich soils at the global scale over the past decade.

In box plots, the solid lines and squares in the boxes, the lower and upper edges of the boxes and the lower and upper lines extending from the boxes represent median and mean values ($n = 4$ for N0, $n = 8$ for N), Q1 and Q3, $1.5 \times \text{IQR}$ above the box's Q1 and $1.5 \times \text{IQR}$ below the box's Q3 of all data, respectively. A mixed linear model was used to test the treatment effect at $P = 0.05$. C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. C levels are categorized according to the organic carbon threshold (i.e., $\text{SOC} = 15 \text{ g kg}^{-1}$) determined by the global meta-analysis.



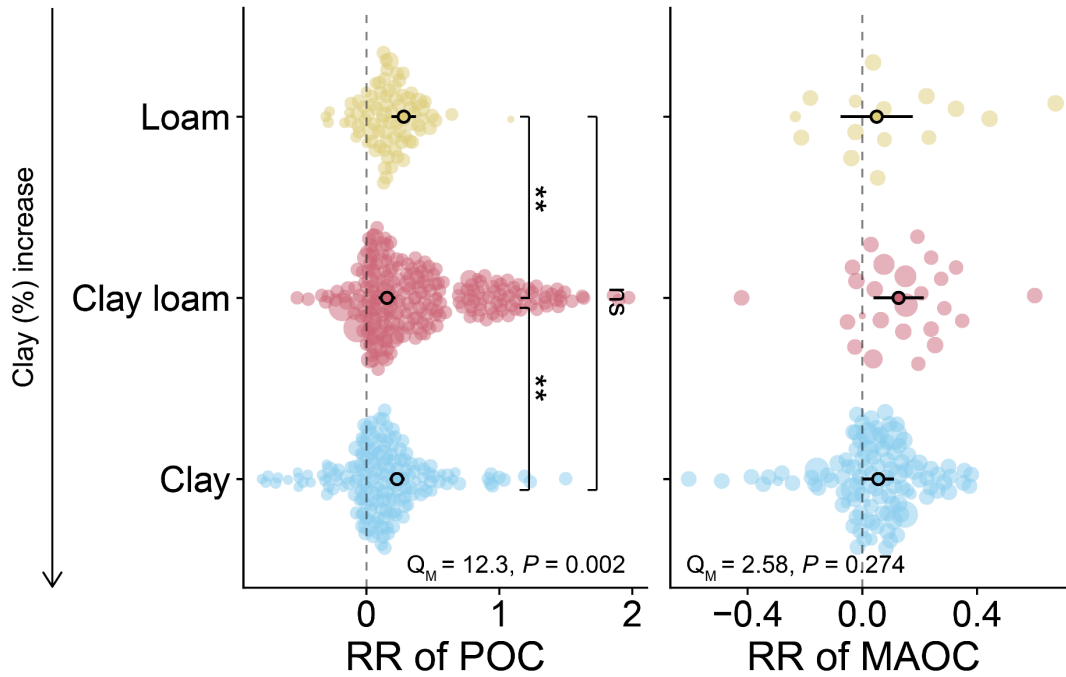
Supplementary Figure 13. POC and MAOC changes due to N fertilization by ecozones over the past decade.

Box plot shows the 25th and 75th percentiles (box borders), median (central black lines) and data ranges (whiskers). POC: particulate organic carbon; MAOC: mineral-associated organic carbon.



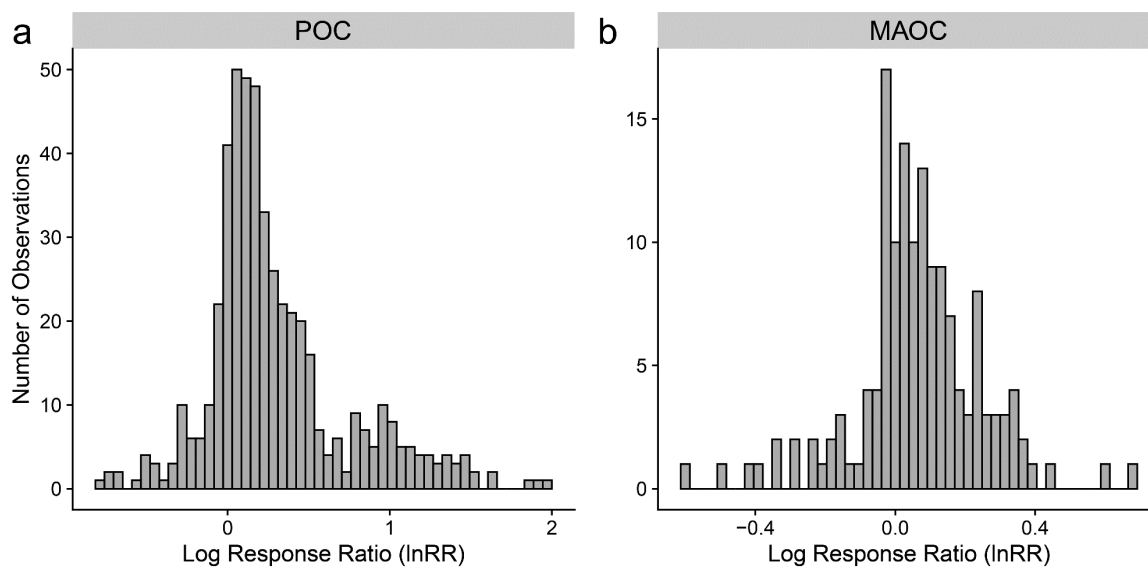
Supplementary Figure 14. The ratio of fungal (F) to bacterial (B) biomass under different N fertilizer treatments between C-poor and C-rich soils.

In box plots, the solid lines and squares in the boxes, the lower and upper edges of the boxes and the lower and upper lines extending from the boxes represent median and mean values ($n = 4$ for N0, $n = 8$ for N), Q1 and Q3, $1.5 \times \text{IQR}$ above the box's Q1 and $1.5 \times \text{IQR}$ below the box's Q3 of all data, respectively. A mixed linear model was used to test the treatment effect at $P = 0.05$. N0: no N addition; N: N addition.



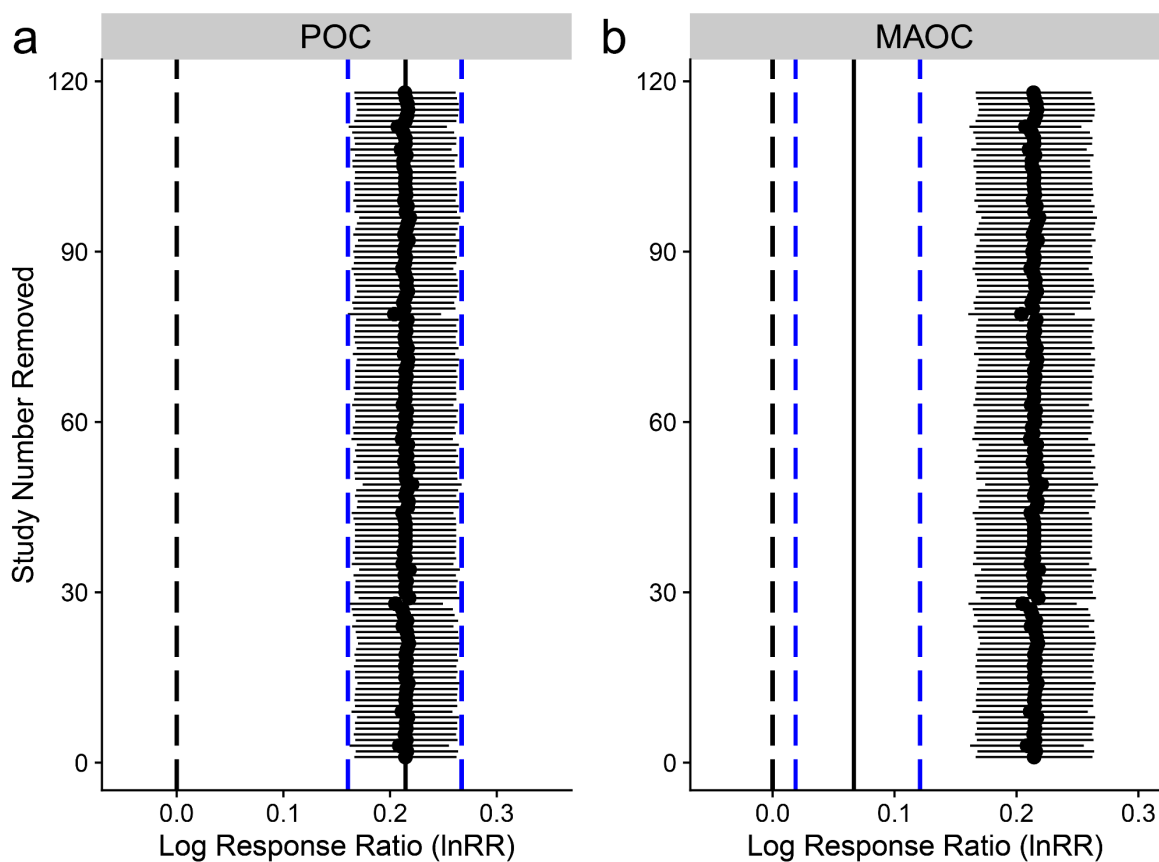
Supplementary Figure 15. Effects of N fertilization on POC and MAOC associated with soil texture.

The between-group heterogeneity (Q_M) statistical test was used to compare the differences in weighted effect sizes among groups divided by the soil texture. A significant Q_M value ($P < 0.05$) suggested that the weighted effect sizes of a given variable differed among groups. No samples were classified as sandy, resulting in only three observed soil texture categories. An asterisk means there is a significant difference between the two groups, and ns means no difference. * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$.



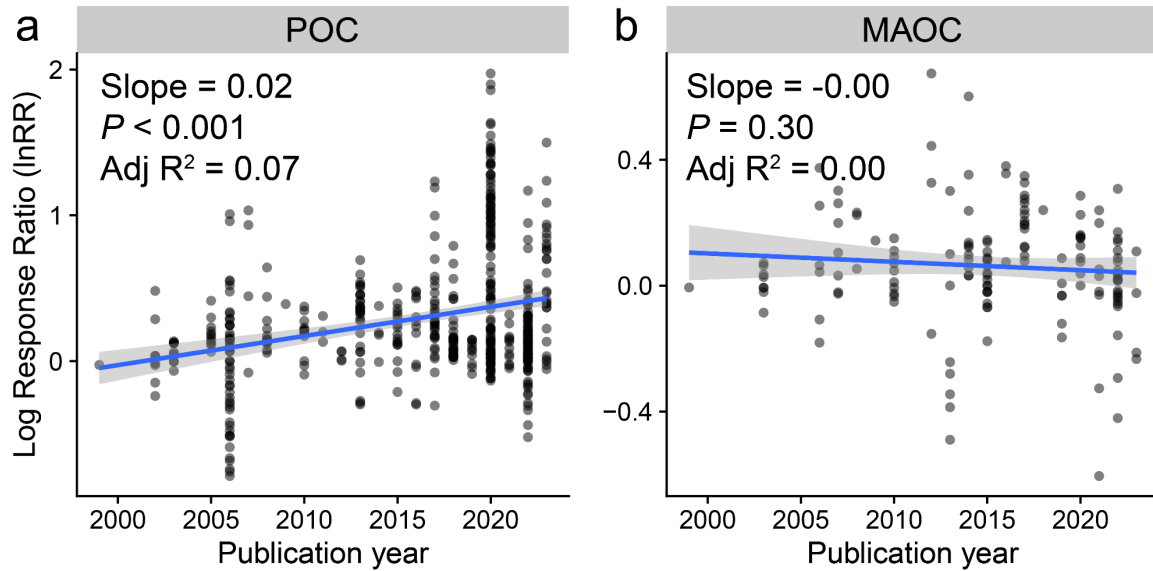
Supplementary Figure 16. Publication bias of POC (a) and MAOC (b) in the datasets.

The histogram showed that the observations were close to normal distribution, and that the meta-analysis was not subject to publication bias. POC: particulate organic carbon; MAOC: mineral-associated organic carbon.



Supplementary Figure 17. The results of the sensitivity analysis for POC (a) and MAOC (b), which was completed using a Jackknife technique.

The solid black line represents the overall lnRR and the lower and higher 95% confidence intervals are also provided as dashed blue lines. The removal of any single study had no effect on the results. POC: particulate organic carbon; MAOC: mineral-associated organic carbon.



Supplementary Figure 18. Temporal change in effect size of nitrogen fertilizer on POC (a) and MAOC (b) with publication year based on meta-regression analysis.

Linear mixed regression model with two-sided test was used for the statistical analysis, and adjusted R-squared was used. The solid line represents the linear regression, and the gray shading indicates the 95% confidence intervals. POC: particulate organic carbon; MAOC: mineral-associated organic carbon.

Supplementary Table 1. Results of the model selection procedure used to evaluate the response between effect sizes of POC/MAOC and initial SOC.

Variables	model	AICs	Δ AICs	BICs	Δ BICs
POC	Stegmented	491.79	0.00	519.01	0.00
	M22	522.92	31.13	554.34	35.33
	Step	545.40	53.61	564.21	45.20
	M12	558.60	66.81	585.81	66.80
	Segmented	564.19	72.40	587.20	68.19
	GAM	589.53	97.74	602.13	83.12
MAOC	Stegmented	-100.18	0.00	-82.28	0.00
	Step	-96.30	3.88	-80.15	2.13
	M22	-92.88	7.30	-76.77	5.51
	M12	-92.56	7.62	-75.48	6.80
	Segmented	-92.50	7.68	-72.54	9.74
	GAM	-85.78	14.4	-69.85	12.43

Bold fonts indicate the best model. The best fitting models showed the lowest AIC and BIC values within a Δ AIC and Δ BIC < 2. POC: particulate organic carbon; MAOC: mineral-associated organic carbon; SOC: soil organic carbon. GAM: generalized additive model; step: two intercept models, with no slope are fitted and the intercept is changed at the breakpoint; segmented: two linear models in which the slope is changed at the breakpoint; stegmented: two linear models in which both the slope and the intercept are changed at the breakpoint; M12: hinge model 12, one linear model is fitted for the left part of the breakpoint and a second-degree polynomial is fitted for the right part; M22: hinge model 22, two different second-degree polynomial models are fitted at both sides of the breakpoint. AIC: Akaike information criteria; BIC: Bayesian information criteria.

Supplementary Table 2. The independent and interactive effect (P values) of initial C conditions (C) and N fertilization input (N) on studied variables.

Variables	Initial SOC (C)		N input (N)		C×N	
	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
POC	0.491	0.555	10.8	< 0.001	15.1	< 0.001
MAOC	120	0.008	13.9	< 0.001	18.0	< 0.001
AGB	1.44	0.353	386	< 0.001	0.008	0.991
Lignin phenols	1.71	0.321	12.3	< 0.001	31.6	< 0.001
Necromass C	1.60	0.333	2.81	0.072	22.7	< 0.001
qCO ₂	111	0.008	15.5	< 0.001	19.9	< 0.001
Oxidase	6.81	0.121	30.3	< 0.001	14.8	< 0.001
F:B	4.15	0.179	7.10	0.002	0.88	0.422
Resource limitation	0.16	0.728	3.57	0.037	3.91	0.027
C limitation	2.68	0.243	0.16	0.849	6.28	0.004
MWD	1.94	0.299	38.2	< 0.001	56.1	< 0.001
Fe _o + Al _o	92.4	0.011	1.91	0.161	4.23	0.022
Fe _p + Fe _p	2.70	0.242	37.5	< 0.001	39.4	< 0.001
Ca _{exe}	12.5	0.071	0.58	0.561	1.41	0.256
Mg _{exe}	1.26	0.379	4.81	0.013	1.65	0.206
Aliphaticity	0.05	0.846	0.20	0.822	9.08	< 0.001
Recalcitrance	10.8	0.081	0.01	0.987	2.92	0.065

Bold values indicate $P < 0.05$. Linear mixed effect models used Satterthwaite approximation for degrees of freedom. AGB: Aboveground biomass; POC: particulate organic carbon; MAOC: mineral-associated organic carbon; qCO₂: microbial metabolic quotient (respiration-to-biomass ratio); F:B: the ratio of fungal to bacterial biomass; Resources limitation: the ratio of the resource C:N (SOC:TN) ratios normalized to B_{C:N} (MBC:MBN); C limitation: the square root of the sum of x^2 and y^2 , where x represents the relative activity of C vs. P-acquiring enzymes, and y represents the relative activity of C vs. N-acquiring enzymes; MWD: mean weight diameter; Fe_o + Al_o: poorly crystalline Fe/Al oxides; Fe_p + Al_p: organically complexed Fe/Al oxides; Aliphaticity: the ratio of the

alkyl region divided by the O-alkyl region; Recalcitrance: the ratio of (alkyl + aromatic C) to (O-alkyl + carboxyl C).

Supplementary Table 3. Effects of N fertilizer application on activities of oxidative enzymes including polyphenol oxidase (PPO) and peroxidase (POD).

Fertility conditions	Treatments	PPO (nmol h⁻¹ g⁻¹)	POD (nmol h⁻¹ g⁻¹)	Oxidase (nmol h⁻¹ g⁻¹)
C-poor soils	N0	4334 (792)	2759 (350)	82.3 (6.76) a
	N	3976 (368)	1645 (176)	73.9 (3.33) b
C-rich soils	N0	1738 (210) a	800 (94.1) a	50.2 (1.58) a
	N	963 (92.3) b	435 (24.3) b	37.1 (1.24) b

Data are presented as mean and SE ($n = 4$ for N0, $n = 8$ for N). Different lower-case letters indicate significant differences between N0 and N within one site at $P = 0.05$ (Tukey-post-hoc test). The oxidase activity is calculated as the square root of the sum of PPO and POD. PPO: polyphenol activity. POD: Peroxidase activity. Fertility conditions: classified according to the soil organic carbon threshold obtained from the global meta-analysis (i.e., SOC = 15 g kg⁻¹). OPT: optimized N addition; HN: high N addition. C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. N0: no N addition; N: N addition.

Supplementary Table 4. Percentage (%) contribution of C functional groups of bulk soil based on integration of CPMAS ¹³C NMR spectra.

Fertility conditions	Treatments	Alkyl-C (0-45 ppm)	O-alkyl-C (45-110 ppm)	Aromatic-C (110-160 ppm)	Carbonyl-C (160-220 ppm)	Aliphaticity	Recalcitrance
C-poor soils	N0	23.61 (1.02)	46.70 (0.44) b	17.69 (0.32) b	11.53 (0.4)	0.51 (0.02) a	0.71 (0.02)
	N	22.12 (0.47)	48.67 (0.39) a	18.86 (0.16) a	11.32 (0.65)	0.45 (0.01) b	0.68 (0.01)
C-rich soils	N0	21.10 (0.94) b	46.15 (0.56)	20.78 (0.49)	11.25 (0.54)	0.46 (0.02) b	0.73 (0.01) b
	N	22.94 (0.75) a	45.85 (0.29)	20.16 (0.64)	10.96 (0.20)	0.50 (0.02) a	0.76 (0.01) a

Data are presented as mean and SE ($n = 4$ for N0, $n = 8$ for N). Different lower-case letters indicate significant differences between N0 and N within one site at $P = 0.05$ (Tukey-post-hoc test). The Aliphaticity index is the ratio of the alkyl region divided by the O-alkyl region. The Recalcitrance index is the ratio of (alkyl + aromatic C) to (O-alkyl + carboxyl C). Fertility conditions: classified according to the soil organic carbon threshold obtained from the global meta-analysis (i.e., SOC = 15 g kg⁻¹). C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA results. N0: no N addition; N: N addition. N0: no N addition; N: N addition.

Supplementary Table 5. Performance of the four machine-learning algorithms tested predicting POC and MAOC changes, including the gradient boosting decision tree model for POC and MAOC in the main text.

Machine-learning algorithms	Variables	MAE	MSE	R ²
RF	POC	0.17	0.05	0.51
	MAOC	0.11	0.02	0.16
GBDT	POC	0.16	0.05	0.56
	MAOC	0.08	0.02	0.28
XGBoost	POC	0.18	0.06	0.45
	MAOC	NA	NA	NA
LightGBM	POC	0.18	0.06	0.45
	MAOC	0.11	0.03	0.11

Bold indicates the model chosen for best performance and its R². RF: random forest; GBDT: gradient-boosting decision trees model; XGBoost: extreme gradient-boosting model; LightGBM: light gradient-boosting machine model. MAE: mean absolute error; MSE: mean squared error. NA indicates that such models are not suitable for modeling and predicting the current dataset, with performance worse than that of a mean model. The optimal model is characterized by the maximum R² and the minimum MAE and MSE.

Supplementary Table 6. Summary of covariates used in this study for upscaling our results to a global context.

Group	Covariates	Units	Resolution	Source
Climate	Mean annual temperature (MAT)	°C	30 s	WorldClim, ²
	Mean annual precipitation (MAP)	mm year ⁻¹	30 s	
	Soil organic carbon (SOC)	g kg ⁻¹	250 m	
Soil	Total nitrogen (TN)	g kg ⁻¹	250 m	SoilGrids, ³
	pH	/	250 m	
	Clay Fraction (Clay)	%	250 m	
	Silt Fraction (Silt)	%	250 m	
Fertilizer	Nitrogen fertilization rate (Ninput)	kg ha ⁻¹ year ⁻¹	5 min	PANGAEA, ⁴
/	Land use data	/	30 m	⁵

Soil properties from SoilGrids were converted into standardized units based on conversion relationships provided in the official manual to align with our dataset.

Supplementary Table 7. Site description and general description of the experimental design at four sites of nitrogen fertilization treatments.

Sites	Geography	N fertilization rate (Kg N ha ⁻¹ year ⁻¹)	Duration of N fertilization (years)	Altitude (m)	Fertility conditions
QuZhou station (QZ)	43.33 N, 124.00 W	N0: 0 OPT: 171 HN: 250	13	41	C-poor soils
Changwu station (CW)	35.22 N, 107.80 W	N0: 0 OPT: 200 HN: 400	12	1096	C-poor soils
Siping station (SP)	36.88 N, 115.20 W	N0: 0 OPT: 200 HN: 312	12	167	C-rich soils
Yaan station (YA)	29.98 N, 103.00 W	N0: 0 OPT: 190 HN: 381	13	560	C-rich soils

Fertility conditions: classified according to the soil organic carbon threshold obtained from the global meta-analysis (i.e., SOC = 15 g kg⁻¹). C-poor soils: combining QZ and CW results; C-rich soils: combining SP and YA result. OPT: optimized N addition; HN: high N addition.

Supplementary Table 8. A summary of the quality check of this meta-analysis.
Quality criteria are derived from Koricheva and Gurevitch.

Quality criteria	This study
1. Has formal meta-analysis been conducted (i.e. combination of effect sizes using standard meta-analytical methodology) or is it simply a vote count?	Yes, this study used natural log response ratio (lnRR) weighted by inverse variance to evaluate the effect size. Vote count was used to map the global distribution.
2. Are details of bibliographic search (electronic data bases used, keyword combinations, years) reported in sufficient detail to allow replication?	Yes, all electronic data (including extracted data and reference list) used in the study are provided, and a PRISMA flow diagram is also provided.
3. Are criteria for study inclusion/exclusion explicitly listed?	Yes, all criteria are listed in the Methods section.
4. Have standard metrics of effect size been used or, if nonstandard metrics have been employed, is the distribution of these parameters known and have the authors explained how they calculated variances for such metrics?	Yes, we used standard metrics of effect size, i.e., Ln(Response Ratio) weighted by inverse variance, and effect size is also calculated.
5. If more than one estimate of effect size per study was included in the analysis, has potential non-independence of these estimates been taken into account?	Yes, we performed a sensitivity analysis to assess the reliability of the results
6. Have effect sizes been weighted by study precision or has the rational for using unweighted approach been provided?	Yes, effect sizes were weighted by the inverse variance.
7. Have statistical model for meta-analysis and the software used been described?	Yes, we calculated the weighted mean of lnRR in a mixed-effects model using the “metafor” package in R version 4.2.1.
8. Has heterogeneity of effect sizes between studies been quantified?	Yes, confidence interval was given in all meta-analyses, and the accumulated percentage of effect sizes were shown in Fig. 5.
9. Have the causes of existent heterogeneity in effect sizes been explored by meta-regression?	Yes, grouping experiments were used.
10. If effects of multiple moderators have been tested, have potential non-independence of and interactions between moderators been taken into account?	Yes, both potential non-independence of and interactions between moderators were taken into account by the <i>glmulti</i> package in R.
11. If meta-analysis combined studies conducted on different species, has phylogenetic relatedness of species been taken into account?	Not applicable to this study.
12. Have tests for publication bias been conducted?	Yes, we carried out egger test and drew the histogram plot.
13. If meta-analysis combines studies published over considerable time span, have possible temporal changes in effect size been tested?	Yes, possible temporal changes in effect size were tested using a meta-regression method, as shown in Supplementary Fig. 7.
14. Have sensitivity analysis been performed to test the robustness of results?	Yes, the jackknife technique was run to test the robustness of results, as show in Supplementary Fig. 6.
15. Have full bibliographic details of primary studies included in a meta-analysis been provided?	Yes, the full bibliographic details of primary studies are deposited in a public repository.
16. Has the data set used for meta-analysis, including effect sizes and variances/sample sizes from individual primary studies and moderator variables, been provided as electronic appendix?	Yes, all the data are deposited in a public repository.

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