Supplementary Materials

Deep carbon in agricultural soils: considerations for its treatment in soil carbon studies

Maxwell Locke, Adrian Unc

School of Science and the Environment, Memorial University of Newfoundland, Corner Brook A2H 5G4, Canada.

Correspondence to: Prof. Adrian Unc, School of Science and the Environment, Memorial University of Newfoundland, Corner Brook A2H 5G4, Canada. E-mail: aunc@mun.ca

ORCID: Maxwell Locke (0009-0002-8851-9933), Adrian Unc (0000-0002-7265-9758)

As an initial assessment of how research on deep SOC under agriculture has developed, on May 14, 2026, a Scopus search was conducted using the following search string in titles, abstracts and keywords:

> "deep soil*" AND ("carbon" OR "organic carbon" OR "organic matter" OR "soil organic carbon" OR "soil organic matter" OR "SOM" OR "SOC") AND ("agricultur*")

The search was filtered to include English-language research articles and excluded documents from 2026, yielding 334 results. A second search using the same string but without "agricultur*" and applying the same filters yielded 1,935 results.

Another Scopus search was conducted on June 10, 2026, to identify examples of deep soil warming experiments used in Figure 3. The titles, abstracts and keywords were searched using the following string:

> “warming” AND “soil” AND (“deep” OR “subsoil”) AND “carbon” AND ("respiration" OR "Q10" OR "temperature sensitivity") AND NOT ("river" OR "lake")

The search was filtered to include English-language research articles and excluded documents from 2026, yielding 65 results. Documents were subset to include or exclude “incubation,” yielding 24 and 41 results, respectively. Documents were scanned, and the following were excluded:

- Meta-analyses
- Ecosystem respiration or NPP studies not explicitly measuring soil heterotrophic respiration
- Modelling studies
- CH_4 and N_2O studies
- Studies not including deep or subsoils declared by the authors
- Studies where changes in canopy coverage from wildfire were used as a treatment for soil warming

Once filtered, this left a total of 11 for the incubation subset and 4 for the subset excluding incubation. Articles were scanned to extract information on climate, soil type, *In situ* warming and length, the inclusion of natural temperature gradients, baseline temperature and moisture regimes, incubation temperature and length (Table S1).

Table S1. Contrasting top versus subsoil microbial community characteristics associated with the age structure panel in Figure 2

Dimension	Topsoil — Generalists	Deep Soil — Specialists
Soil layer	Surface/topsoil, approximately 0–30 cm	Deep soil/subsoil, commonly >30 cm and extending beyond 1 m
Resource availability	High and variable	Low and relatively stable
Nutrient regime	Resource-rich; copiotrophic conditions	Resource-poor; oligotrophic conditions
Environmental variability	Dynamic; frequent changes in moisture, temperature, nutrients, and disturbance	More buffered; slower-changing physical and chemical conditions
Management influence	Strong surface-management effect	Management effect decreases with depth
Microbial diversity	Higher diversity	Lower diversity
Community structure	Diverse, interactive, and heterogeneous communities	Narrower, more filtered communities
Network complexity	High co-occurrence network complexity	Simpler co-occurrence networks
Dominant taxa favoured	Habitat generalists	Habitat specialists
Community assembly	Often more stochastic, especially under disturbance	More deterministic under stressed or nutrient-limited conditions
Response to disturbance	Generalists can outcompete specialists during disturbance	Specialists are favoured under persistent, stable, or stressful niche conditions

Information from Refs[1-5].

Table S2. Extracted information from the retained deep SOC warming experiment Scopus search articles

Ref.	Location/climate	Soil type	*In situ* (yes/no)	Baseline temp regime (°C)	*In situ* soil warming above baseline (°C)	*In situ* warming length	Baseline moisture regime	Incubation (yes/no)	Incubation temp (°C)	Depths	Incubation length
[6]	Jiangsu Province, east coast of China	Fluvisols	No	Not reported; MAT of 14.6			Not reported	Yes	15, 25, 35	0-10 and 40-60 cm	28 days
[7]	northern China temperate deserts	Not reported	No	Not reported; average temps -0.6 to 13.9			Not reported	Yes	5 to 35 in 5 degree intervals	0-10, 30-50 cm	2.5 to 20 hours
[8]	subtropical monsoon, Zhejiang province, China	Haplic Luvisols, Acric Plinthosols, Profondic Acrisols, Drystic Cambisols (WRB) each on different parent material	No	MAT of 17			Not reported	Yes	15 and 25	0-10, 10-20	90 days
[9]	Temperate continental, Hebei Province, China	Not reported	No	MAT of 12.9, inc temp of 15 was chosen because it's the mean of the wheat season air temp			Not reported	Yes	15 and 25	0-20, 20-40	90 days
[10]	cold temperate continental monsoon, Great Hing'an mountains, China	sparse island permafrost wetland	No	MAT is -1.5 ranging from -32 in Jan to 27 in Aug, active layer temp 4-7 in Aug			Not reported	Yes	5 and 15	50 cm in 10 cm increments	97 days
[11]	southern Tibet	Matti-Gelic Cambisol, Albic Umbri-Gelic Cambisol, Drystic Podzoluvisol, Eutric Cambisol	No	Air temp ranges from -0.27 to 11; soil temperature was measured but not reported			Not reported	Yes	5 and 15	100 cm	128 days
[12]	Central belt of Scotland, temperate oceanic climate	Lowland raised ombrotrophic peat bog	No	MAT of 9.3 C; not reported			Not reported	Yes	5, 10, 15, 20, 25	60 cm	5 days; 24 hours for each temperature
[13]	Loess Plateau, northwest China, temperate, arid and semiarid continental monsoon	Haplic Calcisols	No	MAT is 7 C with max summer temp of above 30 C; soil temp not reported			Not reported	Yes	25 and 35	100 cm	30 days
[14]	Subtropical mountain humid monsoon	Hapludalfs	No	Monthly temp ranges from 1.3 to 21.1; annual soil temps 1.3-21.1 for 0-10 cm, 1.8-			Not reported	Yes	20 and 25	0-10, 10-30, 30-60	150 days

				20.5 for 10-30, 3-19.1 for 30-60 cm							
[15]	Continental monsoon, Qinghai-Tibetan Plateau alpine grassland	Mat-Gryic Cambisol	Yes	MAT of -1.2 C, average warming plot surface temp increase by 2 C. Soil temp measured (5, 10, 20 cm) but not reported.	2	2 years	Not reported	Yes	25 C	0-10 and 30-40 cm	86 days
[16]	Alaska, permafrost	Poorly drained silt loam, no soil type	No	MAT is -2.5; soil temps not reported			Not reported but field moisture content of cores before incubation ranged from 19 to 77% by mass	Yes	4 and 20 C	30 cm intact cores extracted under ~80 cm permafrost active layer	100 days
[17]	western France, oceanic climate	Cambisol	Yes	MAT is 12 (-3.1 to 27.9 C)	4 C (uniform across the profile)	2 years	Recorded for each depth in control and warmed plots (warming decreased water content for most depths)	No		100 cm	
[18]	California, mediterranean	Alfisol	Yes	Only temp difference between warm and control plots reported; MAT of 12.5 C	4	4.5 years	Not reported; only volumetric moisture difference reported	No		90 cm	
[19]	southeast Tibet; temperate, frigid dark coniferous, alpine subfrigid shrub meadow, alpine frigid meadow and desert zones	luvisols, cambisols and cryosols	No	Not reported; MAT ranges from -1 to 2.6		60 days	Not reported; field moisture ranges from 29-47% for 0-20 cm and 15-38 for 40-60 cm	Yes	15 and 25	0-20, 40-60	60 days
[20]	California, mediterranean	Alfisols	Yes	Difference between warm and control plots reported; MAT of 12.5 C	4	2 years	Not reported; only VMC difference reported	No		100 cm	

REFERENCES

1. Kehlet-Delgado H, Montoya AP, Jensen KT, et al. The evolutionary genomics of adaptation to stress in wild rhizobium bacteria. *Proc Natl Acad Sci U S A*. 2024;121(13):e2311127121. doi:10.1073/PNAS.2311127121;WGROUP:STRING:PUBLICATION

2. Dragone NB, Hoffert M, Strickland MS, Fierer N. Taxonomic and genomic attributes of oligotrophic soil bacteria. *ISME Communications*. 2024;4(1). doi:10.1093/ismeco/ycae081

3. Hao J, Chai YN, Lopes LD, et al. The Effects of Soil Depth on the Structure of Microbial Communities in Agricultural Soils in Iowa (United States). *Appl Environ Microbiol*. 2021;87(4). doi:10.1128/AEM.02673-20

4. Chen YJ, Leung PM, Wood JL, et al. Metabolic flexibility allows bacterial habitat generalists to become dominant in a frequently disturbed ecosystem. *ISME J*. 2021;15(10):2986-3004. doi:10.1038/S41396-021-00988-W

5. Liu XJA, Han S, Frey SD, Melillo JM, Zhou J, Deangelis KM. Microbial responses to long-term warming differ across soil microenvironments. *ISME Communications*. 2024;4(1). doi:10.1093/ismeco/ycae051

6. Yang S, Xu X, Peng F, et al. Unexpected responses of SOC decomposition and its temperature sensitivity to plant invasion across soil layers: Implications for plantation understory management. *CATENA* 2025;256:109110. DOI:10.1016/j.catena.2025.109110

7. Huang G, Su Y. Microorganisms exert overriding impacts on the temperature sensitivity of soil C decomposition than substrate quality. *Soil Ecol Lett* 2025;7:303. DOI:10.1007/s42832-025-0303-5

8. Mao X, Sun T, Liu X, et al. Disentangle the drivers of soil organic carbon mineralization and their temperature sensitivity in both topsoil and subsoil: Implication of thermal stability and chemical composition. *Ecological Indicators* 2024;158:111399. DOI:10.1016/j.ecolind.2023.111399

9. Yan Z, Jia R, Zhou J, et al. Soybean inclusion reduces soil organic matter mineralization despite increasing its temperature sensitivity. *Sci Total Environ* 2024;922:171334. DOI:10.1016/j.scitotenv.2024.171334

10. Wang Y, Guo Y, Wang X, et al. Mineral protection controls soil organic carbon stability in permafrost wetlands. *Sci Total Environ* 2023;869:161864. DOI:10.1016/j.scitotenv.2023.161864

11. Mao X, Zheng J, Yu W, et al. Climate-induced shifts in composition and protection regulate temperature sensitivity of carbon decomposition through soil profile. *Soil Biology and Biochemistry* 2022;172:108743. DOI:10.1016/j.soilbio.2022.108743
12. Schimmel H, Braun M, Subke J, Amelung W, Bol R. Carbon stability in a Scottish lowland raised bog: potential legacy effects of historical land use and implications for global change. *Soil Biology and Biochemistry* 2021;154:108124. DOI:10.1016/j.soilbio.2020.108124
13. Bai T, Tao J, Li Z, et al. Different microbial responses in top- and sub-soils to elevated temperature and substrate addition in a semiarid grassland on the Loess Plateau. *European J Soil Science* 2019;70:1025-36. DOI:10.1111/ejss.12800
14. Tian Q, Wang X, Wang D, et al. Decoupled linkage between soil carbon and nitrogen mineralization among soil depths in a subtropical mixed forest. *Soil Biology and Biochemistry* 2017;109:135-44. DOI:10.1016/j.soilbio.2017.02.009
15. Jia J, Feng X, He J, He H, Lin L, Liu Z. Comparing microbial carbon sequestration and priming in the subsoil versus topsoil of a Qinghai-Tibetan alpine grassland. *Soil Biology and Biochemistry* 2017;104:141-51. DOI:10.1016/j.soilbio.2016.10.018
16. Bond-lamberty B, Smith AP, Bailey V. Temperature and moisture effects on greenhouse gas emissions from deep active-layer boreal soils. *Biogeosciences* 2016;13:6669-81. DOI:10.5194/bg-13-6669-2016
17. Khan MZ, Chabbi A, Hicks Pries CE, Torn MS, Rumpel C. Management impacts on whole soil warming responses of CO2 production and efflux in temperate climate. *Geoderma* 2023;440:116725. DOI:10.1016/j.geoderma.2023.116725
18. Zosso CU, Ofiti NOE, Soong JL, et al. Whole-soil warming decreases abundance and modifies the community structure of microorganisms in the subsoil but not in surface soil. *SOIL* 2021;7:477-94. DOI:10.5194/soil-7-477-2021
19. Xu M, Li X, Kuyper TW, Xu M, Li X, Zhang J. High microbial diversity stabilizes the responses of soil organic carbon decomposition to warming in the subsoil on the Tibetan Plateau. *Glob Chang Biol* 2021;27:2061-75. DOI:10.1111/gcb.15553
20. Hicks Pries CE, Castanha C, Porras RC, Torn MS. The whole-soil carbon flux in response to warming. *Science* 2017;355:1420-3. DOI:10.1126/science.aal1319