Agri Articles
(e-Magazine for Agricultural Articles)
Volume: 06, Issue: 04 (JULY-AUG, 2026)
Available online at http://www.agriarticles.com

Soil Biology & Biochemistry: An Assessment of Soil Microbial Biomass

*Ragini Verma, Ankul Kumar and Satyarath Sonkar
Chandra Shekhar Azad University of Agriculture and Technology, Kanpur-208002, Uttar Pradesh, India
*Corresponding Author's email: ragini1999verma@gmail.com

Soil microbial biomass (SMB), the living fraction of soil organic matter excluding plant roots and macrofauna acts as an essential eye of the needle through which all organic materials must pass during biogeochemical transformations. Accounting for 1% to 5% of total soil organic carbon (SOC) and 2% to 6% of organic nitrogen (N), SMB serves as a sensitive, rapid-response indicator of environmental perturbation and soil quality. This comprehensive treatise evaluates SMB within soil biology and biochemistry. It details SMB's biochemical functions, contrasts classic and modern analytical quantification techniques, analyzes environmental drivers, and examines its role as a diagnostic tool for sustainable land management.

Keywords: Soil Microbial Biomass, Carbon Use Efficiency, Chloroform Fumigation-Extraction, Substrate-Induced Respiration, Phospholipid Fatty Acids, Soil Quality Indicators.

Introduction & Conceptual Framework

Soil organic matter (SOM) represents the largest terrestrial carbon pool. However, the vast majority of SOM consists of recalcitrant or mineral-associated organic matter that changes slowly over decadal or centennial timescales. In contrast, **soil microbial biomass (SMB)** represents the living, highly active fraction of SOM, comprising bacteria, fungi, archaea, microalgae, and protozoa (Bargali, 2024).

The biomass pool acts simultaneously as a **sink** and a **source** for plant nutrients:

- **As a sink:** Microorganisms temporarily immobilize nutrients within cellular structures during growth, protecting elements like nitrogen, phosphorus, and sulfur from leaching or volatilization.
- **As a source:** Microscopic die-off and turn-over release bioavailable ions back into the soil solution, sustaining primary productivity.

Biochemical and Functional Roles in Terrestrial Ecosystems

Biogeochemical Cycling and Nutrient Fluxes

Microorganisms drive the key metabolic transformations of terrestrial biogeochemistry:

1. **Carbon Cycling:** Microbes decompose plant residues through extracellular enzymes (e.g., cellulases, chitinases, β-glucosidase), releasing CO_2 through respiration while assimilating carbon into microbial tissue.
2. **Nitrogen Mineralization & Immobilization:** The breakdown of organic nitrogen compounds (proteins, amino sugars) into inorganic ammonium (NH_4^+) and nitrate (NO_3^-).
3. **Phosphorus and Sulfur Turnover:** Enzymatic hydrolysis of organic phosphate esters via phosphatases mobilizes orthophosphate ($H_2PO_4^-$ / HPO_4^{2-}), making phosphorus available to plant roots (Sinegani & Hosseinpur, 2010).

Soil Structure and Physical Stabilization

Soil biology directly influences physical soil architecture. Microorganisms synthesize extracellular polymeric substances (EPS)—mucilaginous polysaccharides and glycoproteins such as glomalin—that bind microaggregates (<250µm) into stable macroaggregates (>250µm). Fungal hyphae further enmesh these soil particles, forming a network that improves soil aeration, water infiltration, and resistance to erosion.

Eco-Physiological Indices

To assess the physiological status of the microbial community, two key bioenergetic parameters are frequently evaluated:

- **The Metabolic Quotient (qCO2):** Calculated as the unit of respiration per unit of biomass carbon per hour:

Metabolic Quotient (qCO2)=Microbial Biomass Carbon (mg Cmic
kg−1)Basal Respiration (mg CO2-C kg−1 h−1)

A elevated qCO2 often indicates environmental stress, high energy expenditures for cellular maintenance, or an early stage of ecological succession.

- **Microbial Carbon Use Efficiency (CUE):** The proportion of ingested carbon allocated to growth versus respiration:

CUE=Total Carbon AssimilatedCarbon Allocated to Biomass Production

Quantitative Methodology for Estimating Soil Microbial Biomass

Accurately measuring SMB presents unique challenges due to the complex soil matrix, mineral interactions, and spatial heterogeneity (Broos et al., 2007). Several classical physiological methods and modern molecular techniques have been developed over recent decades.

Chloroform Fumigation Methods

Chloroform Fumigation-Incubation (CFI)

Introduced by Jenkinson and Powlson (1976), CFI relies on exposing moist soil samples to ethanol-free chloroform ($CHCl_3$) vapor inside a vacuum desiccator for 24 hours. The chloroform disrupts cell membranes, causing cell lysis. After removing the residual fumigant, the soil is inoculated with fresh, unfumigated soil and incubated for 10 days. The flush of CO_2 evolved during this period (attributable to the decomposition of killed microbial tissues by the fresh inoculum) is measured.

Chloroform Fumigation-Extraction (CFE)

To eliminate the time-consuming 10-day incubation requirement of CFI, Vance et al. (1987) introduced the Chloroform Fumigation-Extraction procedure. Moist soil samples are split into two subsets: one is fumigated with $CHCl_3$ for 24 hours, while the other serves as an unfumigated control. Both sets are extracted using 0.5M K_2SO_4 solution. Dissolved organic carbon (DOC) and extractable total nitrogen (ETN) in the extracts are analyzed.

Substrate-Induced Respiration (SIR)

Developed by Anderson and Domsch (1978), SIR measures the initial, physiological respiratory response of soil microbes following the addition of an easily degradable carbon source (typically D-glucose). Within the first 1–4 hours—before population growth occurs—the rate of CO2 production is directly proportional to the active microbial biomass.

Biochemical and Molecular Approaches

1. **Phospholipid Fatty Acid (PLFA) Profiling:** Lipids are extracted from cell membranes. Because phospholipids break down rapidly upon cell death, PLFA serves as a quantitative measure of living microbial biomass and provides insight into community composition (e.g., fungal-to-bacterial ratios, Gram-positive vs. Gram-negative bacteria).
2. **Adenosine Triphosphate (ATP) Extraction:** Measures cellular energy state. Although highly sensitive, ATP concentrations fluctuate significantly with metabolic state, requiring standardized assay conditions.
3. **Genomic DNA Extraction & Quantitative PCR (qPCR):** Quantifies ribosomal gene copy numbers (16S rRNA for bacteria/archaea, ITS regions for fungi) to estimate abundance and biomass across environmental gradients.

Environmental and Management Factors Regulating Microbial Dynamics

Microbial biomass dynamics are shaped by interactions between abiotic environmental drivers and human management practices.

Abiotic Controllers

- **Soil Texture and Structure:** Soils with higher clay content typically maintain larger microbial biomass reserves than sandy soils. Fine clay particles create microhabitats that protect microbial cells from predation by protozoa and nematodes.
- **Soil pH:** Near-neutral pH levels (6.0−7.5) generally support maximum microbial biomass and diversity. Highly acidic soils (pH<5.0) tend to inhibit bacterial populations while shifting dominance toward fungal taxa.
- **Moisture and Temperature:** Optimal microbial turnover occurs at field capacity (30–60 kPa suction) and ambient temperatures between 20∘C and 30∘C. Severe drying-rewetting cycles cause osmolytic lysis, releasing transient flushes of nutrients.

Anthropogenic Impacts and Land Management

Agricultural Intensification: Continuous conventional tillage physically disrupts fungal networks and exposes previously protected organic matter to rapid oxidation, leading to declines in SMB. Conversely, **conservation agriculture**—characterized by minimal soil disturbance, cover cropping, and organic amendments—restores SMB pools and enhances biological health (Semenov et al., 2025).

- **Exogenous Amendments (Compost, Manure, Biochar):** Adding organic residues fuels microbial proliferation. However, amendments like biochar can complicate standard analytical measurements due to sorption effects during chemical extraction (Lehmann et al., 2011; Paliaga et al., 2025).

Methodological Challenges and Research Horizons

Despite significant advances in soil biochemistry, several methodological challenges remain:

1. **Conversion Factor Uncertainties:** Standard conversion factors (kEC,kEN,kEP) assume uniform extraction efficiencies across diverse soil orders. However, varying clay mineralogy, organic matter content, and soil chemistry can distort these calculations.
2. **Matrix Interferences:** Materials with high surface reactivity—such as biochar, expanded clays, or high-iron oxisols—can re-adsorb lysed cellular contents during extraction, underestimating SMB (Paliaga et al., 2025).
3. **Multi-Omic Integration:** Bridging quantitative biomass measurements (Cmic, Nmic) with high-throughput multi-omics (metagenomics, metatranscriptomics, metaproteomics) remains a key goal. Integrating these approaches will help link overall biomass size with specific, active metabolic pathways in the soil.

Conclusion

Soil microbial biomass plays a pivotal role in maintaining soil functionality, nutrient availability, and ecosystem stability. Though accounting for only a small fraction of total soil organic matter, its rapid turnover makes it an irreplaceable engine for biogeochemical processes. Integrating standard chemical extraction methodologies with physiological metrics and molecular tools allows researchers to effectively assess soil biological health, guide sustainable land management, and monitor soil responses to global environmental change.

References

1. Bargali, S. S. (2024). Soil microbial biomass: A crucial indicator of soil health. *Current Agriculture Research Journal*, *12*(1), 01–06. https://doi.org/10.12944/carj.12.1.01
2. Broos, K., Macdonald, L. M., Warne, M. S. J., Heemsbergen, D. A., Barnes, M. B., Bell, M., & McLaughlin, M. J. (2007). Limitations of soil microbial biomass carbon as an indicator of soil pollution in the field. *Soil Biology and Biochemistry*, *39*(10), 2693–2695. https://doi.org/10.1016/j.soilbio.2007.05.014
3. Bünemann, E. K., Bongiorno, G., Bai, Z., Creamer, R. E., De Deyn, G., de Goede, R., Fleskens, L., Geissen, V., Kuyper, T. W., Mäder, P., Pulleman, M., Sukkel, W., van

Groenigen, J. W., & Brussaard, L. (2018). Soil quality – A critical review. *Soil Biology and Biochemistry*, *120*, 105–125. https://doi.org/10.1016/j.soilbio.2018.01.030

4. Chen, L., Xu, J., Feng, Y., Wang, J., Yu, Y., & Brookes, P. C. (2015). Responses of soil microeukaryotic communities to short-term fumigation-incubation revealed by MiSeq amplicon sequencing. *Frontiers in Microbiology*, *6*, Article 1149. https://doi.org/10.3390/fmicb.2015.01149
5. Lehmann, J., Rillig, M. C., Thies, J., Masiello, C. A., Hockaday, W. C., & Crowley, D. (2011). Biochar effects on soil biota – A review. *Soil Biology and Biochemistry*, *43*(9), 1812–1836. https://doi.org/10.1016/j.soilbio.2011.04.022
6. Paliaga, S., Laudicina, V. A., Muscarella, S. M., Said-Pullicino, D., & Badalucco, L. (2025). Comparison of different methods for estimating microbial biomass in biochar-amended soils. *Soil Biology and Biochemistry*, *203*, Article 109733. https://doi.org/10.1016/j.soilbio.2025.109733
7. Powlson, D., Xu, J., & Brookes, P. (2017). Through the eye of the needle — The story of the soil microbial biomass. In *Microbial Biomass* (pp. 1–40). World Scientific Publishing. https://doi.org/10.1142/9781786341310_0001
8. Semenov, M. V., Zhelezova, A. D., Ksenofontova, N. A., Ivanova, E. A., Nikitin, D. A., & Semenov, V. M. (2025). Microbiological indicators for assessing the effects of agricultural practices on soil health: A review. *Agronomy*, *15*(2), Article 335. https://doi.org/10.3390/agronomy15020335
9. Sinegani, A. A. S., & Hosseinpur, A. (2010). Evaluation of effect of different sterilization methods on soil biomass phosphorus extracted with NaHCO3. *Plant, Soil and Environment*, *56*(4), 156–162. https://doi.org/10.17221/86/2009-pse