Life and death in the soil microbiome: how ecological processes influence biogeochemistry

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Abstract | Soil microorganisms shape global element cycles in life and death. Living soil microorganisms are a major engine of terrestrial biogeochemistry, driving the turnover of soil organic matter — Earth's largest terrestrial carbon pool and the primary source of plant nutrients. Their metabolic functions are influenced by ecological interactions with other soil microbial populations, soil fauna and plants, and the surrounding soil environment. Remnants of dead microbial cells serve as fuel for these biogeochemical engines because their chemical constituents persist as soil organic matter. This non-living microbial biomass accretes over time in soil, forming one of the largest pools of organic matter on the planet. In this Review, we discuss how the biogeochemical cycling of organic matter depends on both living and dead soil microorganisms, their functional traits, and their interactions with the soil matrix and other organisms. With recent omics advances, many of the traits that frame microbial population dynamics and their ecophysiological adaptations can be deciphered directly from assembled genomes or patterns of gene or protein expression. Thus, it is now possible to leverage a trait-based understanding of microbial life and death within improved biogeochemical models and to better predict ecosystem functioning under new climate regimes.

Microbial necromass
Dead cellular biomass (for example, cell envelopes) and extracellular products (for example, extracellular polymeric substances).

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https://doi.org/10.1038/s41579-022-00695-z

The soil microbiome is the most biologically diverse community in the biosphere, holding at least a quarter of Earth's total biodiversity[1]. Tens of millions of species of bacteria, archaea, fungi, viruses and microeukaryotes coexist below ground, although only a few hundred thousand have been characterized in detail[2]. A single gram of surface soil can contain more than 10^9 bacterial and archaeal cells[3], trillions of viruses[4], tens of thousands of protists[5] and 200 m of fungal hyphae[6]. In microbial ecology, the taxonomic diversity of a microbial assemblage and the abundance of its individual members is termed 'community structure'. Soil microbiome structure varies widely both across different ecosystems[7,8] and smaller-scale soil habitats[9], but bacteria and fungi typically dominate soil microbial biomass and diversity, with abundances several orders of magnitude higher than other microbial groups[10,11]. Across Earth's biomes, soil microbial diversity is positively related to a range of ecosystem functions such as nutrient cycling, decomposition and plant productivity[12].

Soil microorganisms strongly influence terrestrial biogeochemistry by forming and decomposing soil organic matter (SOM) — the planet's largest terrestrial stock of organic carbon and nitrogen, and a primary source of other crucial macronutrients and micronutrients[13]. By shaping the turnover of SOM, soil microorganisms influence atmospheric concentrations of CO_2 and global climate, and help provide crucial ecosystem services like soil fertility, carbon sequestration, and plant productivity and health. However, the soil microbiome's influence on biogeochemistry extends well beyond the metabolic activities of living organisms. After death, microbial necromass accretes in soil, constituting as much as 50% of the SOM pool[14–17]. Because soil microbial necromass represents one of the most globally significant pools of carbon and other nutrients, the mechanism and rate of microbial death likely impact terrestrial biogeochemical cycling.

An understanding of how microbial life and death shape soil biogeochemistry is now emerging, and

Ecological succession
A consistent, distinct trajectory of community change through time.

Rhizosphere
The zone of soil under direct influence of a living plant root.

Hyphosphere
The zone of soil under direct influence of fungal hyphae.

Detritusphere
The zone of soil under direct influence of decaying litter.

Bulk soil
Soil that is not in the direct influence of living or dead roots; characterized by lower levels of microbial density and activity relative to high-resource habitats.

Ecophysiological traits
Traits related to the physiology of a microorganism, as shaped by their biotic and abiotic ecological context.

emphasizes the role of microbial population dynamics, trophic relationships, microbial interactions with their soil environment, and the causes and consequences of microbial mortality. New evidence shows that soil microbiomes are shaped by: (1) microbial population-level processes, such as varied taxon-specific growth and death rates in response to growing roots, decaying litter and environmental perturbations[18–20]; (2) microbial community dynamics, such as distinct patterns of ecological succession in different soil habitats[21–23]; and (3) biotic interactions, such as how different types of competition and predation can influence microbial physiology[24] and necromass chemistry[25]. At the same time, a parallel body of research illuminates how different components of microbial necromass, such as cell walls, proteins, DNA and extracellular products, undergo decomposition[26,27] and form SOM[25,28–31].

In this Review, we illustrate how the ecological processes of living and decaying microorganisms can shape soil biogeochemistry. First, we summarize how the traits of living microorganisms interface with the properties of the soil mineral matrix to affect organic matter cycling and promote accrual of microbial necromass. Next, we describe how community-level processes (for example, succession) and biotic interactions (for example, competition and predation) influence how microbial activity and mortality impact SOM. In so doing, we illustrate how different mechanisms of microbial death may yield distinct effects on the formation and persistence of SOM. Finally, we discuss how new trait-based approaches provide a tractable means to incorporate processes of microbial life and death into models that predict soil biogeochemical dynamics.

Microbial traits and the mineral matrix

The mineral matrix is a complex and heterogeneous landscape where soil microorganisms interact and express traits that directly shape SOM cycling. It is also the graveyard that houses microbial products after death[32]. Far more than just a physical substrate for microbial colonies, the mineral matrix facilitates electron transfer and provides crucial elements as well as oxidants and reactive minerals that mediate the transformation of microbial necromass into SOM[33]. Different habitats within this mineral matrix host distinct microbial communities that vary in their density, activity and composition, with direct consequences for how organic matter is cycled (FIG. 1).

Habitats in the mineral matrix. The soil mineral matrix develops from physical and biochemical weathering as rocks are broken and chemically transformed into successively smaller particles, ranging in size from centimetre-scale stones to nanometre-scale clays. The heterogeneity of this three-dimensional porous architecture generates a wide range of habitats for microorganisms to colonize and inhabit. These habitats include living and dead roots, preferential paths along which water and resources flow, as well as the interior and exterior regions of porous soil aggregates. In much of this mineral matrix, microorganisms are minimally active[34] and sparsely distributed; less than 0.000001% of the total surface area in mineral soil may be occupied by living microorganisms[35]. However, in localized, dynamic, resource-rich habitats of the mineral matrix, such as the rhizosphere, hyphosphere and detritusphere, carbon and other nutrients are more abundant than in bulk soil[36]. These resource-rich habitats can constitute a significant portion of surface soil: between 8% and 26% of the total soil volume in the top 10 cm can be occupied by the rhizosphere alone[37]. Within habitats where roots and fungal hyphae are actively growing or decaying, microbial biomass and activity is 0.5–20 times greater than in the surrounding bulk soil[36,37], biotic interactions are more frequent[38,39], and microbially driven organic matter transformations are rapid[36,40] (FIG. 1).

Microbial ecophysiology and SOM cycling. Soil microorganisms possess a broad range of ecophysiological traits that can influence how organic matter persists within the mineral matrix[41], including cellular chemical composition, life history and biophysical characteristics, and adaptations to environmental and biotic stressors (TABLE 1). These traits vary across soil habitats as a function of compositional differences in the microbial community and their resource environment[20,36,42]. As previously described[43], the term 'trait' reflects the phenotypic character of an organism: "any morphological, physiological, or phenological heritable feature measurable at the individual level". Current microbial trait compilations are biased towards taxa that can be cultured in the laboratory. For instance, the 10 most prevalent soil-dwelling microbial taxa included in a recent trait compilation are bacteria from the genera *Burkholderia*, *Campylobacter* and *Bacillus*[44]. These 10 taxa comprise 14% of trait observations compiled from soil, whereas major soil bacterial phyla that are more difficult to culture represent a much smaller fraction (for example, Acidobacteria comprise only 0.5%)[44]. Promisingly, metagenome-assembled genomes and other omics approaches now enable trait predictions for microorganisms that have yet to be cultivated or visually observed[45]. Many of the traits described in TABLE 1 can

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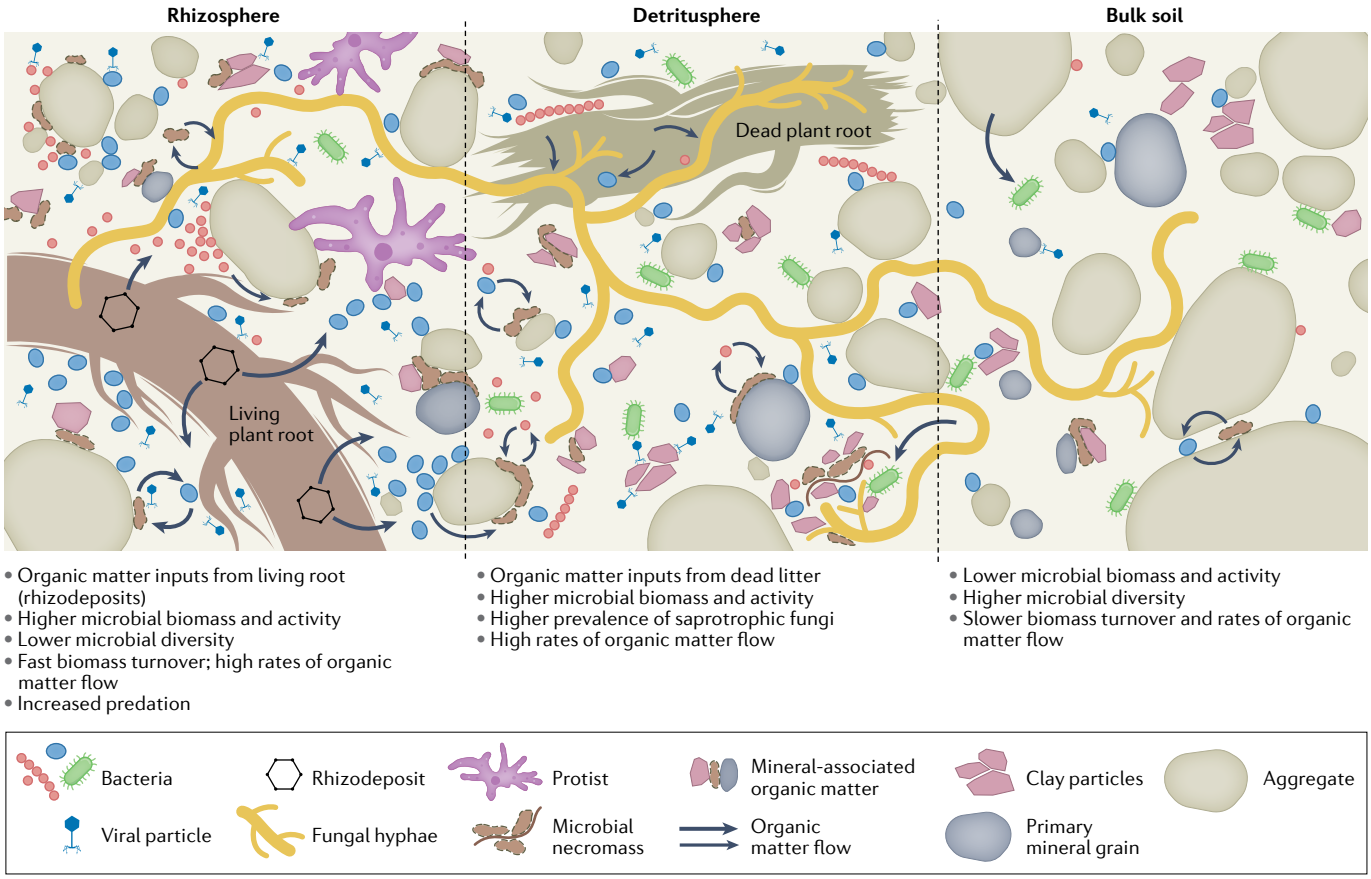


Fig. 1 | **Composition of the soil microbiome and its role in organic matter cycling in different soil habitats.** The biomass and activity of the soil microbiome is greater in the rhizosphere and detritusphere relative to the bulk soil. Organic matter transformations also occur at a faster rate in the rhizosphere and detritusphere relative to the bulk soil, leading to greater accumulation of soil organic matter (for example, mineral-associated organic matter derived from microbial necromass). Arrows indicate flow of organic matter; thicker arrows indicate greater rates of flow.

be detected in genomes, either as genome-inferred traits or via gene or protein expression[46–50].

Traits related to the composition of cell walls and cellular products may be key in the biogeochemistry of SOM because they affect the composition of microbial necromass. Cell envelope and exudate composition varies across microbial groups, with contrasting proportions of peptidoglycan, glycolipids, glycoproteins, compatible solutes, storage compounds, adhesion or stress-tolerance compounds like melanin, exopolysaccharides, and other extracellular products. The presence of different compounds can lead to variable decay rates of microbial necromass based on factors like necromass melanization, stoichiometry and cell morphology[27]. For example, in a boreal forest soil, fungal necromass with higher melanin and lower nitrogen content (*Meliniomyces bicolor*) decomposed more slowly than necromass with lower melanin and higher nitrogen content (*Mortierella elongata*)[51]. Reduced decomposition of melanized fungal necromass can lead to its accrual in boreal forests, driving an overall increase in SOM stocks[52]. Necromass composition can also influence its association with soil minerals and its longer-term persistence as SOM. In a soil chronosequence along a glacier forefield, SOM contained an increasing amount of microbial cell envelope fragments as soils developed[30] — particularly small-sized (100–500 nm) bacterial membrane and cell wall fragments that associate with mineral surfaces[32]. Interactions between mineral surfaces and the functional groups of cell envelope fragments, such as lipids[53], amino sugars[31], and proteins[54], can increase the persistence of SOM, such as by reducing the wettability of soil minerals[54].

Microbial traits also affect SOM turnover by mediating interactions between microorganisms and minerals[55]. For instance, traits related to mineral surface attachment are likely important to SOM formation. The surface-attached lifestyle of many soil microorganisms enables them to withstand severe limitations of water, nutrients and motility[56]. Often living in biofilms, soil microbial cells are embedded in a hydrated matrix of extracellular polymeric substances (EPS) comprised of polysaccharides, proteins, nucleic acids, lipids and other biopolymers, which together make up 80% of the dry mass of the biofilm[57]. EPS bind soil minerals together: for instance, microorganisms living in the rhizosphere of the perennial grass *Panicum virgatum* generate polysaccharides that have been linked to the formation of SOM in mineral aggregates[58]. Microorganisms also contribute to mineral dissolution and formation processes: laboratory experiments with soil microbial isolates

Table 1 | **Microbial ecophysiological traits involved in soil organic matter cycling**

	Traits	Prediction and validation[a]
Life history traits	Minimum generation time	Codon usage, rRNA copy number, microscopy, optical density
	Optimum growth temperature	Amino acid frequencies, microscopy, optical density
Biophysical	Genome	Assembled genome length, DNA yield per cell, GC content
	Cell size and shape	Genome size to cell size, SEM, light microscopy, FACS (isolates or Nycodenz)
	Adhesion and motility	Adhesins, holdfast genes; Pilli, flagella genes, microscopy, capillary assays
Cellular composition	Cell wall or envelope composition	Polysaccharide, lipid, glycoprotein, pigment or Gram-type genes, lipidomics, FTIR, NMR, HPLC, mass spectrometry
	EPS or other residues	EPSac genes, bulk EPS quantification, FTIR, mass spectrometry
Resource acquisition	Exoenzymes	Secreted enzyme genes, activity essays, protein-SIP
	Transport systems	Transporter genes
	Secretion systems	Secretion genes, SEM or TEM
	Metallophores	NRPS siderophore genes, siderophore assays, mass spectrometry
	Storage materials	Phosphoester, phospholipid, polyhydroxybutarate, microscopy, FTIR
Stress tolerance	Stress regulation	Regulatory genes (sigma factors, anti-sigmas, two-component)
	Spore formation	Sporulation genes, spore stains, bulk quantification, DNA-SIP–dormancy
	Osmotolerance	Osmotic response genes (osmolytes, efflux pumps), viral integrity experiments, mass spectrometry, protein-SIP
Antagonism or defence	Antibiotics, toxin–antitoxin systems	Biosynthetic clusters, toxin or antitoxin genes, mass spectrometry
Emergent traits	Realized growth rate	Genome inferred (iREP[46]), heavy water DNA-SIP[18]
	CUE	Genome predicted ranges, quantitative SIP[173], isotope tracing, bulk CUE
	Stoichiometric range	Genome predictions and allometric scaling, nanoSIMS, bulk measurements

CUE, carbon-use efficiency; EPS, extracellular polymeric substances; EPSac, extracellular polysaccharides; FACS, fluorescence-activated cell sorting; FTIR, Fourier-transform infrared spectroscopy; HPLC, high-performance liquid chromatography; iREP, Index of Replication; nanoSIMS, nanoscale secondary ion mass spectrometry; NMR, nuclear magnetic resonance; NRPS, non-ribosomal peptide synthetase; SEM, scanning electron microscopy; SIP, stable isotope probing; TEM, transmission electron microscopy. [a]Many of these traits can be directly measured in genomes and can also be corroborated through bulk characterization or taxon-specific measurements.

show that mineral-attached biofilms accelerate mineral dissolution[56], and imaging of weathered minerals in the field indicates that fungal hyphae can act as nucleation sites for clay minerals and metal-oxide nanoparticles[59]. The close associations between living microbial cells, extracellular products and reactive mineral surfaces create the conditions for microbial necromass to persist in soil as mineral-associated organic matter[30,31,40].

Soil microorganisms use secretion systems, extracellular enzymes and membrane transporters to bring external resources into their cells — traits that influence growth, mortality and carbon-use efficiency (CUE). Realized growth rate (the actual net growth rate of a microorganism) and mortality rate determine the degree of microbial biomass turnover in soil, which affects the total standing stock of microbial necromass and SOM. For instance, in a long-term grassland biodiversity experiment, accelerated microbial growth and turnover was associated with increased microbial necromass and SOM stocks[60]. CUE describes the proportion of a cell's resources converted into microbial biomass relative to the total resources consumed, and is thought to be important for SOM cycling, influencing the amount of necromass produced per unit of substrate consumed[61,62]. In culture, CUE may be estimated directly from microbial populations undergoing exponential growth. However, in soil, a wide diversity of taxa may be experiencing population growth or decline at any given time, and carbon is recycled through the community via the predation and decomposition of necromass and extracellular products. Consequently, 'community-level' CUE estimates derived from soil are not equivalent to culture-based CUE estimates[63]. Nonetheless, community-level CUE is a highly informative index of SOM cycling efficiency: for instance, a direct positive relationship has been observed between CUE and SOM formation in artificial soils, where different carbon substrates were added to SOM-free minerals[41]. Fungal-dominated communities with greater CUE have been associated with particularly high SOM formation[41]. Positive relationships between CUE, necromass production and SOM have also been observed in field studies in both agricultural systems[64] and grassland soils[65]. CUE and realized growth rate are 'emergent' traits in that they depend on the interaction of many biochemical and ecological processes. For instance, genome-based metabolic models suggest that

Mineral-associated organic matter
Soil organic matter that exists in some degree of association with soil minerals.

Carbon-use efficiency
(CUE). Microbial biomass yield given a quantity of available substrate.

microorganisms with a larger genome size can produce the enzymes and transporters needed to access a wide variety of carbon substrates but demonstrate lower CUE than taxa with smaller genomes[66].

To cope with fluctuating resource availability and stress, many microorganisms produce compounds that influence cellular composition and can affect how necromass may persist as SOM[67]. These traits include the production of storage polymers used for energy, carbon, and nutrients such as starch, glycogen, inorganic polyphosphates, triacyl glycerides, trehalose, wax esters, cyanophycin and polyhydroxybuturate[68,69]. The chemistry of different microbial stress compounds may have contrasting effects on their persistence as SOM. For example, in response to grazing by soil isopods (small crustaceans), some saprotrophic soil fungi produce calcium oxalate crystals on their surfaces for physical protection such as the cord-forming basidiomycete fungi *Phanerochaete velutina* and *Resinicium bicolor*[25]. This stress response increases the recalcitrance of their biomass and affects the proportion retained as SOM[25]. Other stress and storage compounds, such as trehalose, are readily hydrolysed to simple sugars and mineralized. For instance, dry grassland soils in California can hold microbial biomass that is up to 20% trehalose by mass[70], and most of this trehalose can be converted into glucose, driving rapid CO_2 loss via mineralization but also potentially supporting microbial growth[71].

When soil conditions become particularly harsh, many microorganisms favour dormancy — broadly defined as a reversible state of reduced metabolic activity[72]. Entering dormancy, microorganisms allocate significant energy and resources towards the formation of resting structures such as spores. As much as 80% of microbial cells are estimated to be dormant at any given time[34], although this value likely varies temporally and spatially (for example, in the rhizosphere versus bulk soil). To the extent that entering dormancy imposes metabolic costs, it might reduce microbial CUE in the short term. However, over the long term, dormant cells both grow and respire less than active cells, so the overall effect of dormancy on CUE and microbial necromass production is unclear[63,73]. Dormant cells do not produce enzymes, and hence regional model simulations suggest that greater dormancy reduces overall SOM decomposition[74]. Most insights regarding the effects of microbial dormancy on SOM cycling are entirely theoretical, highlighting the need for new empirical approaches to study dormancy in situ.

Microbial necromass in the mineral matrix. In many ecosystems, the majority of SOM appears to be derived from microbial necromass[14,15]. Living microbial biomass typically accounts for less than 5% of total SOM[75], and only a small subset of this biomass is active at any given time[34]. Through iterative cycles of microbial growth, death and turnover, a massive stock of microbial necromass is generated in soil, which far exceeds living microbial biomass[62]. This stock accrues into a significant fraction of SOM, partially via interactions with reactive mineral surfaces, which promote its persistence in soil[30,40].

Mineral-associated microbial residues include cellular constituents (for example, cell envelopes)[30], microbially derived nitrogenous compounds (amino sugars and nucleic acids)[31] and extracellular products (for example, EPS, enzymes and glycoproteins)[76]. Traits such as peptidoglycan content, cell size and hydrophobicity also affect the association of cells with minerals[28,77]. For instance, small-celled, hydrophobic *Rhodococcus erythropolis* were retained within soil pores more readily than *Escherichia coli* after being added to soil columns, whereas hydrophilic *E. coli* were more readily leached from the mineral matrix[77]. Mineral identity also matters: distinct minerals associate with different types, sources and amounts of necromass, both because they host distinct living bacterial, fungal, and archaeal communities[23,78,79] and because more reactive minerals (for example, amorphous aluminium hydroxide) can sorb necromass more strongly[29]. Microbial residues may be particularly significant in mildly acidic to mildly alkaline pH soils that are typical of arid climates and grasslands[80]. Although it is clear that microbially derived SOM is created by complex interactions between microbial traits, soil mineralogy and climate, more research is needed to compare SOM formation and persistence in different ecosystem contexts.

In sum, soils contain highly diverse microbial communities distributed across a complex framework of pores, water films, mineral surfaces and organic matter-rich habitats. Ecophysiological traits of both living and dead microorganisms have a crucial role in shaping soil biogeochemical cycles. In adapting to life in a mineral matrix, soil microorganisms have evolved a surface-attached lifestyle and exhibit distinct life history, stress tolerance, cell composition, resource acquisition and defence traits. These traits control the nature and quantity of microbial cellular residues, generating microbially derived SOM that comprises much of the global soil organic carbon and nitrogen reservoir.

Microbial population and community processes

Community assembly, succession, and ecological interactions (for example, predation, competition and mutualism) dictate which microbial taxa are present and how they change through time. These population and community processes influence SOM dynamics by shaping the set of microbial traits present in the community and by triggering changes in necromass chemistry by inducing different mechanisms of microbial stress and death[18,20,25,81].

Microbial succession in a heterogeneous matrix. Soil microbial communities exhibit distinct and reproducible successional trajectories in response to changing conditions. In the rhizosphere, plant roots preferentially stimulate or inhibit specific taxa, resulting in a community that becomes increasingly distinct from bulk soil and is often less diverse[19–21] (FIG. 2). Succession in the rhizosphere is best documented for soil bacteria but also observed for protists[82], fungi[20] and RNA viruses[83]. In temperate grasslands, as the rhizosphere bacterial community develops along growing plant roots, Proteobacteria and Bacteroidetes typically increase in

Community assembly
Processes that shape the identity and abundance of species within a biological community.

Mutualism
A form of symbiosis where both partners benefit.

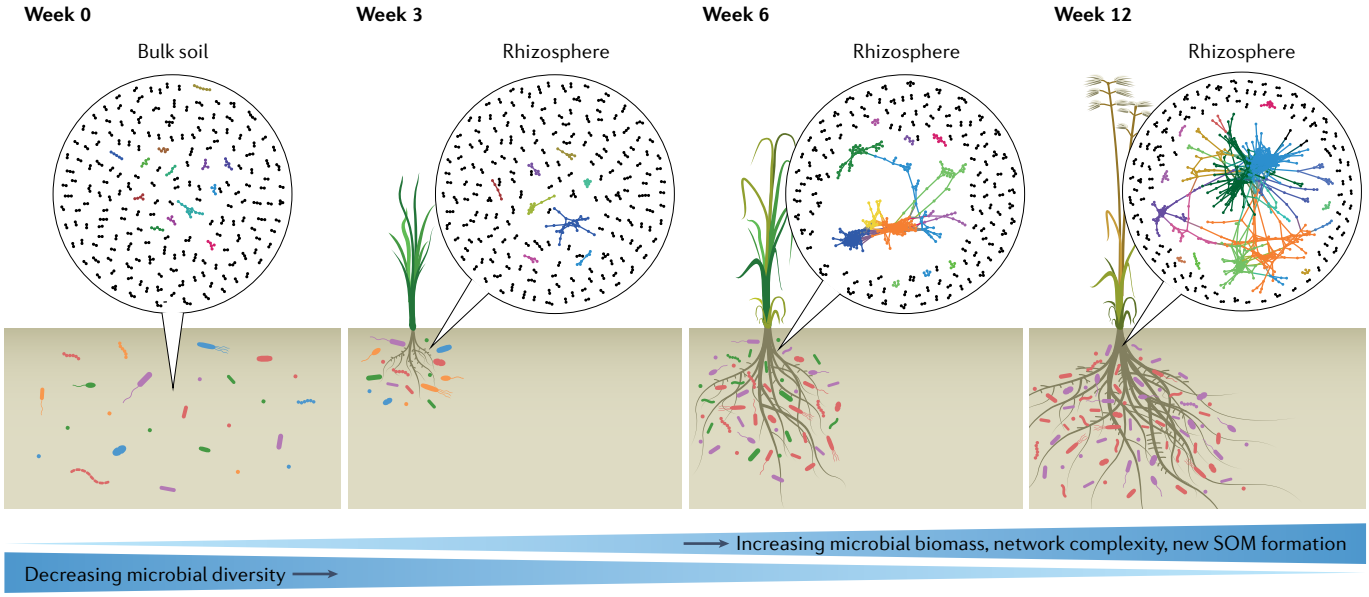


Fig. 2 | **Microbial succession and organic matter formation in the rhizosphere.** During the growing season of an annual plant, as a rhizosphere community develops, its diversity often decreases relative to the bulk soil, even while its biomass and network complexity increases. Simultaneously, new soil organic matter (SOM) is formed in the rhizosphere, although only a subset of it may persist. Adapted with permission from REF.[84], Wiley.

abundance, whereas Acidobacteria, Chloroflexi and Planctomycetes decline[19–21]. Rhizosphere community networks also become larger and more complex than those in bulk soil through time, suggesting that microbial interactions strongly influence community assembly[84,85].

Successional trajectories in the rhizosphere are associated with shifts in microbial functional traits. For example, in a sandy dune soil in the Netherlands, bacteria in the rhizosphere possessed a greater abundance of functional genes related to transporters, glycolysis and hydrogen metabolism relative to the bulk soil community[85]. Many of these functional traits may affect SOM cycling — in particular, the expression of traits driven by organic inputs from growing roots. In the developing rhizosphere of wild oat grass (*Avena* spp.), microorganisms capable of using low molecular weight compounds increase in abundance relative to bulk soil[86]. This shift in the functional potential of the rhizosphere community, evidenced by a higher abundance of genes for organic acid and amino acid transporters, occurred in response to the changing amount and type of carbon that flowed through the rhizosphere[19,20,86,87]. As this functional shift occurs over the growing season, new microbially derived SOM is formed in the rhizosphere, although only a subset of this SOM may persist through time[40,87].

Succession in the detritusphere begins with fast-growing microorganisms, such as taxa in the phyla Proteobacteria and Bacteroidetes, that rapidly consume water-soluble compounds and simple carbohydrates that are released into soil early in decomposition. This is followed by a second wave of slower-growing microorganisms that consume more complex compounds contained within litter such as Basidiomycota fungal taxa, Actinobacteria and Deltaproteobacteria[22]. In the detritusphere of rye (*Secale cereale*), these community shifts were associated with changes in the rate of SOM accumulation over several months. SOM formation was most pronounced early in decomposition and progressively slowed[88]. Similar successional shifts also occur on soil mineral surfaces — a microhabitat known as the 'mineralosphere'[23,78,79]. Fast-growing fungi and Betaproteobacteria that are the first colonizers of mineral surfaces can rapidly contribute necromass that forms mineral-associated SOM[23,79]. Secondary colonizers tend to be slower-growing and adapted to nutrient-poor conditions (for example, Chloroflexi, Verrucomicrobia and Gemmatimonadetes); these microorganisms may feed on the initial necromass, decelerating its accrual and altering the chemical composition of the resulting microbially derived SOM[79].

Biotic interactions: predation, competition and mutualisms. Biotic interactions, such as predation, mutualism and competition (FIG. 3a), are important forces that shape soil microbial community structure and SOM biogeochemistry[89]. The heterogeneity of the mineral matrix limits interactions between microorganisms by increasing physical separation, limiting motility and decreasing hydrological connectivity[3,35,90]. Limited competition may be one of the main forces maintaining the incredibly high diversity (for example, tens of thousands of taxa in a gram of soil[91]) but low density of microbial life in bulk mineral soils[3,92,93]. This is visually apparent in thin soil sections, where clusters of tens to hundreds of bacterial cells are widely dispersed, especially in bulk soils and deeper soil depths[93]. Where resource availability is higher (for example, the rhizosphere compared with the bulk soil, or surface soil compared with deep soil), microbial density increases[37,94] and the influence of density-dependent processes, such as competition[38,85,95]

Density-dependent
A process that regulates population size based on population density.

Viral shunt
The theory that viral lysis of microbial cells returns labile organic matter to an available pool.

and predation on microbial communities and SOM cycling, may be more apparent[89,96] (FIG. 3b).

Soil predators span scales, trophic levels and taxonomic domains, ranging in size from large microarthropods (for example, mites and springtails) to nematodes, protists, predatory bacteria (for example, *Bdellovibrio* spp.) and nanoscopic viruses. Predators can feed selectively or non-selectively, with distinct effects on microbial community structure and SOM cycling[97]. Many bacterivorous nematodes are generalists and consume bacteria taxa indiscriminately via filter feeding. By contrast, bacterivorous protists are more selective, favouring Gram-negative bacteria, which are easier to digest[97]. This selectivity can promote the abundance of Gram-positive bacteria[97], which preferentially consume SOM over plant inputs, potentially leading to enhanced SOM decomposition[98].

Predatory bacteria, although less well studied than other soil predators, cause widespread effects on food web structure[99] and carbon and nutrient flows[100]. Predatory bacteria are highly active in soil and their activity rates increase with resource supply to the base of the foodweb[100]. For example, experimental additions of carbon substrates to soil stimulated growth rates of the obligate predatory bacteria Bdellovibrionales and Vampirovibrionales, with growth responses more than 60% larger than those of non-predatory bacteria[75]. This suggests that, in soil microbiomes with higher productivity, there is increased predator control of lower trophic levels, influencing carbon flow through the belowground food web.

Soil viruses, which can be highly host specific, transform the cell biomass of their prey into extracellularized organic compounds in the process of producing virions (for example, nucleotides and proteins) and lysing the contents of their host cell. Highly diverse and abundant, soil viruses may outnumber microbial cells by as much as 100 to 1 (BOX 1). In oceans, viruses have a prominent role in organic matter cycling, killing up to ~40% of bacteria daily and sustaining up to 55% of bacterial production by continuously liberating and remineralizing dissolved organic matter — a phenomenon known as 'the viral shunt'[101]. Similarly, soil viruses may drive a terrestrial viral shunt, and consequently the cycling of SOM, by disseminating microbial necromass, altering host cell metabolism and CUE, and shaping the pools of organic matter that interact with soil minerals. Resolving how boom and bust cycles of viruses contribute to microbial succession and SOM cycling is a major ongoing area of research[83,102] (BOX 1).

Many soil mutualisms influence SOM dynamics, either by enhancing new SOM production or accelerating decomposition. Soil biofilms are known hotspots of microbial mutualisms and may be comprised of multiple soil bacteria living in highly productive consortia[103]. The synergistic effect of these multispecies interactions has been linked to enhanced syntrophy, stress resistance[104] and biomass production[103]. In a comparison of 35 combinations of multispecies biofilm consortia versus single-species biofilms, greater biomass production was observed in over 60% of the consortia compared with the single-species biofilms[103]. Greater biomass production may promote more SOM formation derived from biofilm EPS[58,105].

Mutualisms between plants and mycorrhizal fungi can also affect SOM production and decomposition[106].

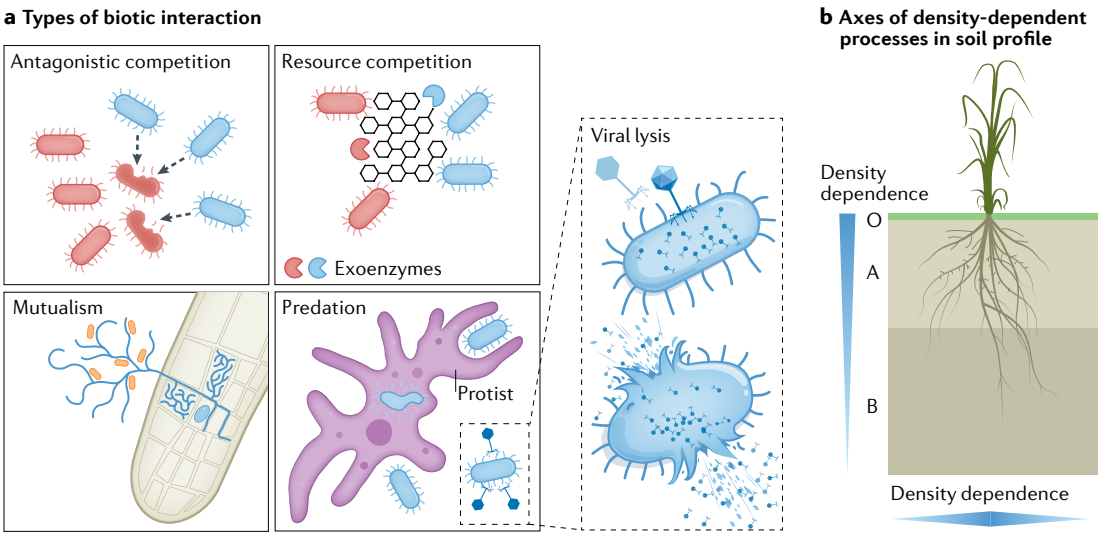


Fig. 3 | **Biotic interactions and density-dependent processes in the soil profile. a** | Primary types of biotic interaction in the soil microbiome, which shape microbial community structure and organic matter cycling. Interactions include antagonistic competition (combative interactions for resources), exploitative competition (indirect competition for resources), mutualisms (for example, interactions between mycorrhizal fungi and plant roots) and predation (for example, protists consuming bacteria or viral lysis). These interactions influence how organisms allocate carbon and can shape the chemical composition and flow of organic matter. **b** | As microbial density decreases with depth or with increasing distance from the rhizosphere (or other resource-rich area), there is also a decrease in density-dependent processes (for example, predation) along these same axes and their effects on organic matter cycling. Shown is a soil profile with an organic layer (O horizon), top layer of mineral soil (A horizon) and mineral subsoil (B horizon).

Box 1 | **The soil virome: composition and ecological functions**

Viral diversity and abundance are staggering, with an estimated ~10^{31} viruses globally and 10^6–10^{10} viral particles per gram of soil[4]. Reports characterizing soil viruses are rapidly expanding, including in forests, permafrost, agricultural soils, wetlands and deserts. These viruses range in size from ~10–100 nm to nearly bacteria-sized giant eukaryotic viruses[186] and include double-stranded DNA viruses that mostly infect bacteria (bacteriophages (phages)) but also archaea and eukaryotes as well as RNA viruses that predominantly infect eukaryotes, particularly plants and fungi.

Viral community composition can be characterized by several culture-independent approaches, including 'viromics', the sequencing of viral-like particles isolated from the soil matrix via filtration or centrifugation[187], or 'viral metagenomics', where nucleic acids of viruses are distinguished within a full microbiome sequence dataset by detecting marker genes, viral hallmark genes and viral motifs[188]. Soil RNA viruses may be detected and characterized by metatranscriptomic approaches that target total RNA[83]. These techniques indicate that soil viromes have an extraordinarily diverse composition but much remains unknown about the soil virome ecological role and function[189], and viruses represent a vast reservoir of hypothetical and uncharacterized proteins[190].

The heterogeneity and complexity of soil constrains the distribution and activity of soil viruses in unique ways. Both abiotic and biotic factors, including pH, water content, depth, bacterial abundance and soil type, strongly impact virus abundance, virion persistence, life cycles (that is, strictly lytic or temperate) and infectivity in soils[4]. Although phages often require cell lysis to propagate (that is, the lytic pathway), environmental conditions may induce lysogeny, where a phage genome integrates into its host chromosome or stably co-exists separately inside the cell. Growing evidence suggest lysogeny is prevalent in soil viruses[191], but to what extent phages may switch between lytic and lysogenic modalities in soil is unknown.

In soil, viruses likely impact microbial community structure and biogeochemistry via (1) cell lysis, which impacts microbial community composition and shapes the pools of labile organic carbon and nutrients that may interact with soil minerals; and (2) host metabolism takeover accompanied by host-mediated expression of virus-encoded auxiliary metabolic genes (AMGs)[192]. AMGs are host genes thought to boost virus replication by maximizing cell resource acquisition and may be acquired through horizontal gene transfer. During infections, viruses fundamentally reprogramme their host's metabolism, which can alter their host cell's biochemical composition as infection proceeds within a virus-infected cell[193,194]. In soils, AMGs are also known to have a role in polysaccharide degradation[102] and sporulation genes[195], sulfur metabolism, ammonia oxidation, dehalogenation, and chitin degradation.

By preferentially preying on certain microbial lineages, viruses may hone community composition and biogeochemical function. For example, a recent study in biological soil crusts showed a bloom in Firmicutes was followed by an increase in Firmicutes-targeting phages[195]. However, testing soil viral–host ecological hypotheses in situ remains a major challenge. Stable isotope probing (SIP) studies have directly implicated soil viruses in carbon cycling through linking isotopically enriched viruses to hosts using CRISPR spacers in plant pathogens and plant growth-promoting bacteria in the rhizosphere[175], in arctic peat soils[196], and with diverse methanotrophs involved in soil methane metabolism[197]. In addition to SIP, methods such as bioorthogonal non-canonical amino acid tagging (BONCAT), nanoscale secondary ion mass spectrometry (nanoSIMS), phageFISH and Virocell-FISH targeting suggest that it is possible to simultaneously link viruses and hosts to carbon flow[189].

Plants allocate up to 30% of photosynthetically fixed carbon to their mycorrhizal symbionts[107] and this transfer fuels hyphal biomass production and growth of other nearby microorganisms[108]. There are two dominant mycorrhizal fungal types with different abilities to scavenge nutrients within SOM: ectomycorrhizal (ECM) fungi produce enzymes that directly oxidize SOM[109], whereas arbuscular mycorrhizal (AM) fungi rely on saprotrophs to perform this function[110,111]. Relative to ECM-dominated ecosystems, AM-dominated ecosystems are associated with faster litter decomposition and nutrient cycling, which can lead to distinct effects on SOM[112]. Although there may be greater topsoil SOM in ECM-dominated ecosystems due to slower SOM cycling[113], the higher rates of microbial necromass production in AM-dominated ecosystems can lead to enhanced formation of more persistent, mineral-associated organic matter[114]. For instance, in a gradient of AM-dominated versus ECM-dominated temperate forest sites, there was more SOM in the upper 10 cm of the ECM-dominated soil but more mineral-associated organic matter and total SOM in the AM-dominated soil at a depth of 1 m (REF.[114]).

Fungal necromass — from mycorrhizae and other fungi that associate with plant roots — may be an important direct source of SOM because the high surface area and small diameter of hyphae allow them to access areas that roots cannot reach such as small soil pores and the interior of soil aggregates[106]. Although quantitative estimates are sparse, available data suggest that, in the top 30 cm of soil, there is an average of 102,000 cm of total fungal hyphae per cubic centimetre of soil, compared with an average of only 6.8 cm of fine roots per cubic centimetre of soil[115]. As hyphae grow and die, their necromass can form SOM as it associates with plant root tannins (that is, tannin–necromass complexes)[116], aggregate structures or mineral surfaces[15,117]. Indeed, the majority of microbially derived SOM may be from fungal necromass in some ecosystems[15], although the specific proportion contributed by mycorrhizal fungi versus saprotrophic fungi remains unquantified.

Two other types of biotic interaction in soil — exploitative competition and antagonistic competition — likely vary in importance in different soil habitats and soil depths, and have contrasting effects on SOM cycling[36,95,118]. Bacterial–fungal antagonistic interactions are pervasive in organic horizons and upper mineral soils, where antibiotic resistance genes are common among bacteria and are used as a strategy to counteract antimicrobial compounds produced by fungi[7]. Overall, antagonistic competition may be more common in habitats where high densities of microbial cells physically encounter one another (for example, litter layer, decaying wood and organic horizons), whereas exploitative competition is thought to be more prevalent in heterogeneous environments like the bulk mineral soil, where lower densities of microorganisms compete for resources and have lower probability of direct encounters[90,95,118].

Antagonistic competition can reduce microbial CUE because carbon is allocated to combat or defence traits as opposed to growth. Combative, interspecific interactions among wood-decay Basidiomycete fungi in soil microcosms, for example, reduced community-level CUE by up to 25%[24]. As a result, in soil habitats where antagonistic interactions are dominant, reduced CUE may decrease the proportion of plant carbon that is transformed into microbially derived SOM. By contrast, in areas of the mineral soil where exploitative competition predominates and microbial diversity is greater, CUE may be relatively high[119,120], leading to more efficient formation of microbially derived SOM[61]. However, the relationship between different forms of competition and organic matter cycling remains a key area of inquiry. For example, competition between mycorrhizal fungi and saprotrophs may inhibit SOM decomposition (known as 'the Gadgil effect') but the relative importance

Exploitative competition
Indirect competition for resources.

Antagonistic competition
Direct competition involving combative interactions.

Trophic transfer
The transfer of energy between trophic levels.

Microbial loop
The flux of nutrients, energy and organic matter within microbial communities.

Birch effect
The ephemeral pulse of CO_2 following wetting of dry soil.

of different types of competition that underlie this phenomenon remain unresolved[121].

Additional biotic interactions in soil remain uncategorized such as those of candidate phyla radiation (CPR) bacteria — ultrasmall bacteria that may act as either bacterial parasites or episymbionts[122]. CPR bacteria, such as *Saccharibacteria* spp., which recycle DNA from their hosts, have incomplete pathways for amino acid and lipid biosynthesis and have been associated with bacterial hosts that live off plant root exudates in grassland soils[123]. In nutrient-depleted environments, CPR bacteria may have an essential role in recycling nutrients and microbial biomass from dead community members, potentially having an important but unexplored role in SOM recycling.

Mechanisms of microbial mortality and SOM cycling. Soil microorganisms die in many ways: via grazing, bacterial predation, viral lysis, osmolysis, desiccation and allelopathy. These mechanisms of microbial mortality may have distinct effects on the structure and function of soil microbiomes[124], cause compositional differences in microbial necromass, and have cascading effects on SOM cycling (FIG. 4). In the ocean, for example, microbial death by viral infection versus protozoan grazing has been linked to distinct changes to microbial community structure[125], necromass chemistry[126] and trophic pathways of carbon flux[101]. In soil, however, links between the modes of microbial mortality and biogeochemical cycling remain largely unexplored. Characterizing these links may be crucial for the accurate prediction of short-term and long-term SOM dynamics[18,81].

Death by predation versus viral lysis should cause notably different effects on the fate of microbial necromass. Microbial grazing by a larger predator, such as a protist, will shift microbial necromass into a higher trophic level, tying the fate of this organic matter to the ultimate fate of the predator. This may reduce SOM accumulation by mineralizing a portion of the organic carbon contained in the prey via the inefficiencies of trophic transfer[127]. Additionally, because protists and nematodes possess a wider C:N ratio than the prey they consume, they excrete excess nitrogen as a waste product, which is then used by plants and other microorganisms via the 'microbial loop'[128]. Bacterial epibiotic predators lyse cells and consume their cytoplasm[129], leaving behind necromass primarily composed of cell membranes and cell wall structures. By contrast, viral lysis may liberate more organic matter than bacterial predation, releasing not only cell membranes and cell walls but also cytoplasm and phosphate-rich phage particles[101]. Although much of this lysed material may be relatively accessible as dissolved organic matter, extracellular virions could constitute an important precursor of mineral-associated organic matter.

Abiotic mechanisms of death, such as osmolysis or desiccation, may drive a parallel set of contrasting effects to these biotic mechanisms. Abrupt environmental changes to the soil environment, such as rapid changes in soil water potential, cause massive microbial die-off and substantial releases of CO_2 — a phenomenon known as 'the Birch effect'. In Mediterranean grasslands, 50% of soil bacteria and 25% of fungi can die within hours following the first precipitation after the long summer dry season[81]. In temperate forests, up to 5–10% of the annual ecosystem carbon budget can be released as CO_2 after a single rainfall event[130]. A sizeable portion of the CO_2 pulse observed after wet-up in grasslands could be derived from osmolysis and the subsequent mineralization of the liberated organic matter within microbial cells[81]. Desiccation, by contrast, may affect soil SOM pools by elevating EPS production, which many microorganisms produce as a response to severe moisture limitation, potentially enhancing cellular

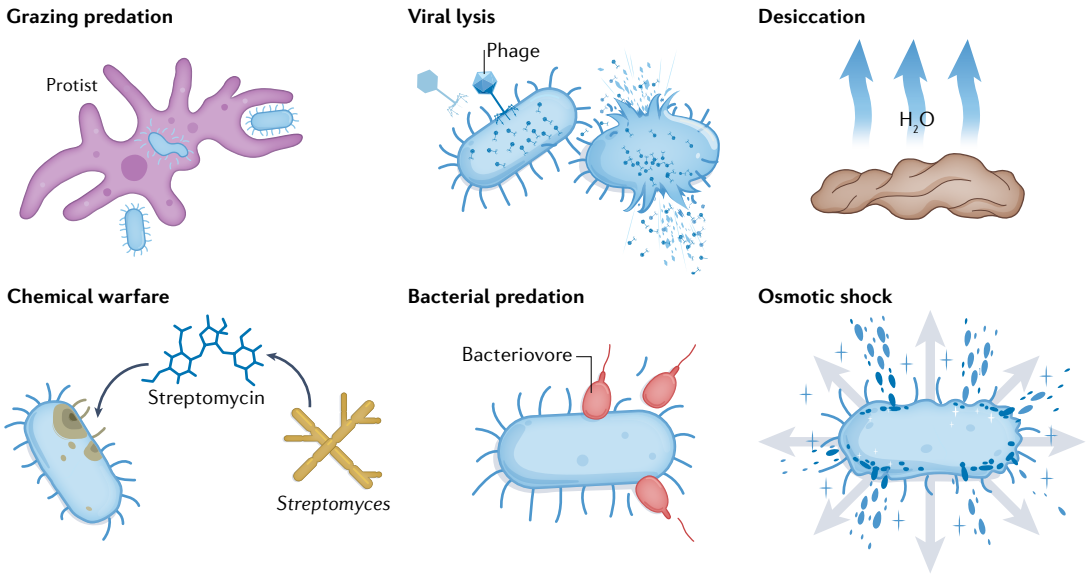


Fig. 4 | **Mechanisms of microbial mortality and theorized effects on the fate of microbial necromass.** There are different ways for a microorganism to die in soil, including grazing, bacterial predation, viral lysis, osmotic shock, desiccation and chemical warfare. The mechanism of death may affect the fate of its necromass, with direct consequences for organic matter cycling.

adhesion to mineral surfaces and increasing the formation of mineral-associated SOM[131]. Finally, chemical warfare is common among soil microorganisms[7]. The mechanisms of allelopathy are diverse, and its consequences may mimic other modes of death, including lysis, desiccation or altered biochemistry of cellular structures[132]. Future research should discern the relative importance of different mechanisms of mortality on the soil microbiome and SOM cycling, and how the rates and dominant mechanisms of death may vary within different soil habitats.

In sum, population and community processes in the soil microbiome (for example, succession, assembly and biotic interactions) can affect the dominant functional traits present in a community, with direct consequences for SOM cycling. Different mechanisms of microbial death may affect the composition and fate of microbial necromass, influencing its persistence in soil. It follows that microbial activity, ecological interactions, and the speed and mode of microbial death are potentially key variables for biogeochemical models that aim to predict SOM dynamics.

Incorporating life and death into biogeochemical models

Biogeochemical models can be used to study the fate of carbon and other elements at many scales. At the global scale, models are particularly crucial for simulating the carbon cycle in order to forecast climate and related Earth system processes[133]. Most global scale biogeochemical models do not resolve microbial population dynamics explicitly but instead simulate the flow of carbon between pools of SOM, where each pool is defined by an intrinsic empirically derived decomposition rate[134]. More complex models that explicitly resolve microbial population growth can incorporate results of field and laboratory incubations at the ecosystem scale[135] or mechanistically simulate interactions between physiologically distinct microbial functional types and their consequences for SOM formation[136]. Fine-scale biogeochemical models can test the relevance of microbial processes at smaller scales[137,138], link across scales and inform larger scale models via process representations that capture underlying microbial community dynamics[139].

Below, we review current approaches and challenges to incorporating microbial processes into biogeochemical models. We specifically highlight trait-based approaches that have gained traction in microbial ecology as a means to synthesize the complexity of microorganisms[140]. These trait-based representations of the soil microbiome are a promising approach to extend current modelling approaches across scales.

Including microbial processes in biogeochemical models. Microbially explicit biogeochemical models have widely different structures but nearly all track the flows and mass balance of carbon and other elements between compartments that represent SOM and microbial biomass[134]. The most coarse-scale microbially explicit models include total microbial biomass as a single compartment[141] but more detailed models simulate populations of functional groups[142] and the energy allocation of individual microbial cells[143]. Models also differ in their characteristic timescale, some capture short-term microbial physiological stress responses, such as during soil drying and rewetting events[144], whereas others predict temperature-dependent adaption of community CUE[145], drought-legacy effects on community function[146], or consequences for long-term changes in soil carbon stocks[147]. In addition to bulk soil representations, some models aim to capture carbon pathways in distinct spatial habitats such as in the litter layer[137,138], rhizosphere[148] and individual soil aggregates[139].

Irrespective of scale, microbially explicit models can be used to deepen our understanding of how microbial physiology and community composition shape SOM dynamics and responses to environmental change[149]. For instance, microbially explicit models can be used to explore the influence of abiotic variables on microbial processes. These include dispersal rates in the soil mineral matrix[150], soil moisture effects on diffusion-limited substrate uptake[151], soil moisture response functions for microbial dormancy[152], EPS and osmolyte production[153], or effects of temperature on carbon uptake rates[145]. Major research efforts have been devoted to constraining environmental response functions with data from soil incubations[154,155] but many uncertainties remain unresolved, including the coupling of combined temperature and moisture effects on microbial process rates[151,156]. There have also been efforts to consider variation in microbial community structure that may regulate ecosystem process rates, treating the microbial biomass as a set of interacting model compartments. These efforts have mostly been focused on exploitative competition for resources, simulating spatial interactions between resource availability and decomposer metabolic traits[157], or focusing on social dynamics within decomposer communities[158]. Only recently have microbially explicit models begun to address ecological processes at the population level[89,96]; relatively few existing models represent density-dependent resource losses such as mortality rates and predator-mediated competition[147,159]. In general, microbial models that represent community structure suggest that emergent carbon processing rates (that is, rates averaged across the community) are distinct from rates predicted for more homogeneous communities[129]. This suggests that the modelling of changes in microbial community structure is relevant for predicting changes in soil carbon stocks.

Incorporating microorganisms into global-scale models affects the magnitude of projected responses of soil carbon stocks to global warming, selectively improving model projections of SOM[160]. Yet, multi-model comparisons reveal that microbially explicit models yield drastically different predictions depending on which processes they represent[161]. In addition, key parameters, including microbial growth and mortality rates[135,161], are often essentially unknown.

The representation of soil microorganisms in SOM modelling is thus poised at a threshold: exploratory efforts have established the importance of microbial processes at a conceptual level, but the complexity of microbial ecology and the challenges of in situ measurements makes it difficult to design and parameterize

Microbially explicit biogeochemical models Models that represent the amount of microbial biomass as a dynamic variable that mediates biogeochemical transformation rates.

Trait inference
Indirect trait quantification based on genomic data, as opposed to direct observation of microbial trait.

biogeochemical models[162]. Trait-based models represent one promising path forward.

Trait-based approaches to biogeochemical modelling. Efforts to incorporate microbial community properties into soil biogeochemical models share a common approach: they group microbial taxa into functional groups based on traits or life history strategies. Similar to community-wide efforts for plants and other organisms, many trait records have been compiled for microorganisms that can be cultured in the laboratory[44]. However, many soil microorganisms are difficult to cultivate and thus trait inference is required. Using genomes and other omics approaches, trait predictions can theoretically be made for the majority of soil microorganisms that have yet to be cultivated[45]. Trait inference can range from detection of simple traits, such as genes that encode key enzymes in metabolic processes, to identification of complex traits that require the coordination of multiple metabolic and biophysical features[163]. Organisms that specialize in soils with distinct resource availability, pH or temperature regimes may have genome-wide signatures that can be used to infer both traits and the organisms' preferred ecological niche[20,86,164].

The task of parametrizing traits in biogeochemical models is aided by the fact that physiological trade-offs constrain microbial fitness and, hence, trait distributions[140]. Owing to fundamental thermodynamic limitations, trade-offs are found across all levels of biological organization from molecules to ecosystems. These trait trade-offs can be represented in terms of energy and resources: the benefit and cost to an organism of each trait in space and time. Models that account for biophysical constraints can represent trait relationships continuously rather than relying on fixed categories[165]. Trade-offs due to cell size are a key example: surface area and volume scale with a 2/3 exponent power law, meaning that the greater surface area-to-volume ratio of smaller cells allows for a comparatively higher substrate uptake affinity[166], but the proportionally lower cell volume limits protein translation[167,168]. As has been found in marine systems, this trade-off results in an ecological strategy where fitness of smaller cells is greater under lower resource concentrations[169]. Together with these biophysical traits, trade-offs related to substrate acquisition, energy generation, stress tolerance and defence define the ecological niche of soil microorganisms in life[11,170] and, ultimately, in death. The mortality rate of microorganisms in soil depends on traits such as growth rate, cell wall composition, production of osmolytes and storage compounds, antimicrobials, and the formation of biofilms and other sessile structures. Because the kinetics of microbial death translate into kinetics of substrate supply for neighbouring microorganisms or minerals, the rate of death of a microorganism in soil must be accurately represented in soil biogeochemical models.

Whereas major advances have been made in measuring microbial functional traits and generating large-scale datasets, data-model integration has lagged behind[140]. This is exemplified by current implementations of trait-based soil biogeochemical models that, at most, distinguish three functional groups based on a limited number of 'functional traits'. For instance, beyond copiotroph or oligotroph (r-K) representations of life history strategies[136], individual-based microbial models simulate interactions between plant polymer degraders, microbial necromass degraders and opportunists[138], or opportunists, decomposers and miners (lignin degraders)[142]. Trait distributions have been predominantly identified through physiological studies (for example, enzyme, substrate uptake or growth kinetics, cell surface area, biomass stoichiometry, composition of storage pools, etc.) or through trait combinations that are constrained by pre-imposed trade-offs[145]. Clearly, the complexity of the soil microbiome renders these approaches unscalable. However, a key recent advance is the ability to reconstruct whole genomes from complex communities. The genome-centric view allows inferred traits to be linked within organisms and patterns of trait-covariance to be assessed and generalized in order to reduce the dimensionality of trait diversity. This allows microbial functional traits to be represented probabilistically. Because such omics data represents the potential of organisms and not realized phenotypes, validating genotype to phenotype linkages in vitro and in situ is important. By using statistically grounded approaches to reduce dimensionality and quantify information loss as microbial traits are aggregated, it is possible to connect omics data with a hierarchy of model structural complexities (BOX 2).

In sum, efforts to incorporate the spectacular diversity of traits present in soil microbiomes into soil biogeochemical models and to represent microbiome variation among systems and in system-specific responses to changing environmental conditions, all require further development. A well-designed trait-based modelling approach may help to resolve the challenges of specifying and parametrizing microbially explicit models and to unlock their predictive power. Linking genome-scale data with biogeochemical process measurements can reveal the structural relationships that are hidden when only one type of data is considered, providing a data-driven foundation for building microbial ecology into biogeochemical models. This will need robust transdisciplinary science that coordinates model development, omics measurements and biogeochemical process measurements within a single iterative framework. We expect that this type of transdisciplinary coordination will ultimately accelerate model structural convergence and achieve more robust forecasts of global-scale soil biogeochemistry.

Conclusions

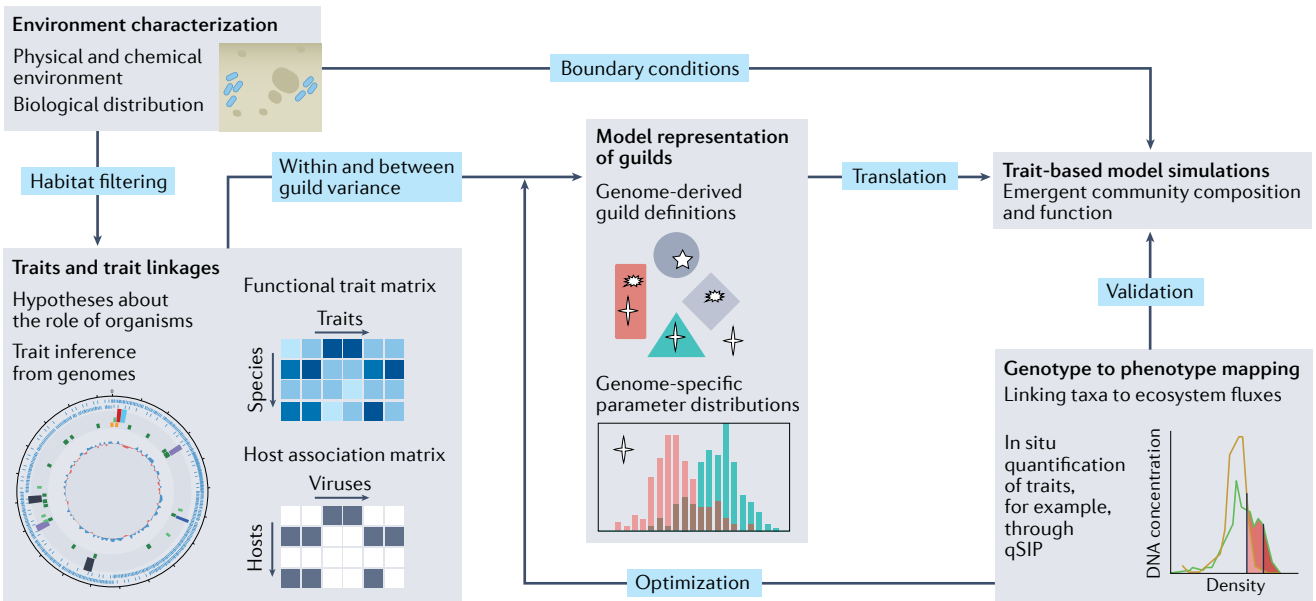
In recent decades, soil microbiology research has started to resolve the taxonomic and functional diversity of wild soil microbiomes[11]. A growing emphasis has now begun to unpack the ecological processes that shape this diversity. This maturing ecological understanding of the soil microbiome has the potential to advance biogeochemical predictions but needs to become fully quantitative. Soils are renowned for their complexity — thus, quantitative efforts are most useful when they can simplify and aggregate essential microbial traits into tractable model parameters. Some continuous trait parameters can be

Box 2 | **Iterative workflow for trait-based model development**

A key challenge to scaling from the diverse microorganisms in soils to ecosystem function is identifying microbial traits that underlie biogeochemical processes. Trait-based models can integrate trait information that is observed as a phenotype (for example, growth rates, microbial substrate uptake, substrate assimilation efficiency and maintenance rates), inferred from genomic proxies directly (for example, minimum generation times and optimal growth temperatures), or via model synthesis. For example, when combined with allometric scaling laws and biophysical modelling approaches, emergent processes like carbon-use efficiency, respiration and microbial biomass turnover can be predicted at the population and community levels. This integration of theory and observations via models is key to understanding the importance of traits for the fitness and activity of soil microorganisms. The variance in trait predictions within and between microorganisms can be used to assign organisms or genomes to 'guilds' (for example, groups of genomes with shared metabolic traits). Although these organisms might be expected to perform a similar range of functions, variance in other traits (such as cell size or generation times) may help to identify guild members with distinct life history strategies that occupy distinct niches. Trait-based model simulations allow us to explore these multi-variate strategies in terms of the shape of trade-offs and trait variation at population or community level, and understand how the fundamental niche becomes the realized niche in a dynamic physical and chemical environment.

Trait-based models rely on a functional trait matrix (genomes × traits) to represent an environment, which may include both binary and continuous microbial trait variables (example workflows to extract microbial fitness traits from genome sequences include microTrait and DRAM[45] (Distilled and Refined Annotation of Metabolism)). Traits related to biotic interactions, such as virus–host associations, may also be included in the functional trait matrix (for example, using the iVirus suite of viromic tools and datasets[198]). Once a matrix of genomes and inferred traits has been built, functional guilds are defined based on the percent of inter-genome trait variance explained. This process provides a statistically grounded approach to reduce the dimensionality of the trait space that is represented in the model. Through trait-based model simulations informed by environmental characterization, realized niches may be predicted, providing hypotheses that can be tested experimentally in an iterative manner that challenges and improves model accuracy.

To connect theoretical relationships between genes, genomes, traits, environment and biogeochemical processes, model benchmarking is a critical step. Models provide predictions at the population and community scale but these need to be benchmarked against observations. For example, taxon-specific microbial growth and mortality rates can now be estimated in situ using quantitative stable isotope probing (qSIP) for thousands of interacting populations within a soil sample and can be condensed into guild-level statistics to compare against model predictions. Similarly, observations of biogeochemical fluxes provide additional objectives with which to evaluate model accuracy. This process is iterative, where both the guild definitions (model structure) and the trait values (model parameters) can be varied until satisfactory agreement between model predictions and observations are achieved, providing additional model-derived hypotheses to confirm with appropriately designed experiments.



included directly in models (for example, maximum specific growth rate[171] and optimal growth temperature[172]), whereas others require an additional translation step before they may be represented in models (for example, acquisition of chemical classes of substrates via transporters or through extracellular breakdown, and binary trait parameters like the presence or absence of a functional capacity) (BOX 2). Key among emerging techniques are those that can measure microbial growth, death and trophic interactions within the complex soil environment[18,173–175].

Multiple techniques now enable a quantitative pathway for integrating genome-informed and omics-informed data into modelling and synthesis efforts. A range of different tracer approaches has been used to determine growth and turnover of the whole bacterial and fungal community in situ (for example, incorporation of radioactive thymidine into soil bacteria, or acetate into ergosterol)[176]. In addition, taxon-specific approaches can capture growth and mortality rates of individual taxa within complex soil environments. These techniques include both non-tracer approaches (for example, iRep[46]) and tracer approaches like quantitative stable isotope probing (qSIP) and bioorthogonal non-canonical amino acid tagging combined with fluorescently active cell sorting (BONCAT–FACS)[173,174]. By tracking the

uptake of elements directly into individual taxa through time, these tracer approaches not only provide insight into ecological processes (for example, succession and trophic interactions[100,175,177]) but can directly connect population dynamics with biogeochemical fluxes (for example, organic matter decomposition and mineralization)[18,178]. After death, techniques exploiting various microbial biomarkers, such as lipidomics[179], ^{13}C-labelled amino sugar analysis[180] and measurements of extracellular DNA[181], can track the fate and composition of microbial necromass in soil. Last, promising developments in methods and experimental design, such as microfluidics and 'transparent soil' microcosms[182], allow direct visualization of microorganism–mineral interactions within solid matrices. When combined with imaging tools (for example, confocal and fluorescence microscopy and nanoscale secondary ion mass spectrometry (nanoSIMS) isotopic imaging, stable isotope probing (SIP), and Raman microspectroscopy), these approaches can provide high-resolution insight into interactions between living and dead microorganisms and their effects on organic matter cycling within a mineral matrix[31,182,183].

As the primary agents of organic matter formation and decomposition, soil microorganisms are front-line managers of the global carbon balance. Climate change is already drastically altering the structure and functioning of the soil microbial communities yet the effects of microbial life cycles on the trajectory of the global climate remain unclear[184,185]. To better understand this complexity and develop predictive models of the soil microbiome's biogeochemical effects, an ecologically informed trait-based framework may provide the most fruitful path forward.

Published online 28 February 2022

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Acknowledgements
The authors thank the Lawrence Livermore National Laboratory (LLNL) Soil Microbiome Scientific Focus Area team for helpful discussions, and K. Georgiou and E. Whalen for providing comments on earlier drafts of the manuscript. This work was supported by the U.S. Department of Energy (DOE), Office of Biological and Environmental Research, Genomic Science Program (GSP) LLNL 'Microbes Persist' Soil Microbiome Scientific Focus Area SCW1632. Work at LLNL was performed under the auspices of the DOE, Contract DE-AC52-07NA27344. Part of this work was performed at Lawrence Berkeley National Laboratory funded under U.S. Department of Energy contract number DE-AC02-05CH11231.

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N.W.S., E.S., G.L.M., A.N., S.J.B., E.L.B., M.M.F., R.H., B.A.H., B.J.K., B.W.S., O.Z. and J.P.-R. wrote the article. All main authors helped contribute to discussions of the content and reviewed or edited the article before submission. The Consortium contributed to several ideas in the manuscript, particularly to Table 1.

Competing interests
The authors declare no competing interests.

Peer review information
Nature Reviews Microbiology thanks the anonymous reviewers for their contribution to the peer review of this work.

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