

## **Opinion** Microbial Metagenomics Reveals Climate-Relevant Subsurface Biogeochemical Processes

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Microorganisms play key roles in terrestrial system processes, including the turnover of natural organic carbon, such as leaf litter and woody debris that accumulate in soils and subsurface sediments. What has emerged from a series of recent DNA sequencing-based studies is recognition of the enormous variety of little known and previously unknown microorganisms that mediate recycling of these vast stores of buried carbon in subsoil compartments of the terrestrial system. More importantly, the genome resolution achieved in these studies has enabled association of specific members of these microbial communities with carbon compound transformations and other linked biogeochemical processes-such as the nitrogen cycle-that can impact the quality of groundwater, surface water, and atmospheric trace gas concentrations. The emerging view also emphasizes the importance of organism interactions through exchange of metabolic byproducts (e.g., within the carbon, nitrogen, and sulfur cycles) and via symbioses since many novel organisms exhibit restricted metabolic capabilities and an associated extremely small cell size. New, genome-resolved information reshapes our view of subsurface microbial communities and provides critical new inputs for advanced reactive transport models. These inputs are needed for accurate prediction of feedbacks in watershed biogeochemical functioning and their influence on the climate via the fluxes of greenhouse gases, CO<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O.

## The Global Importance of Subsurface Microbial Communities

We stand on the cusp of a much deeper grasp of subsurface microbes as agents of biogeochemistry than has ever been achieved before. An understanding of subsurface microbial communities is important at this juncture because the subsurface remains poorly represented in current models of biogeochemical processes, while DNA sequencing and mass spectrometric capabilities now position us to explore this topic comprehensively via metagenomics, metatranscriptomics, and metaproteomics [1–6]. By 'subsurface' we mean that part of the earth below the surface of the terrestrial landscape. As a practical matter, this paper is focused on the shallow subsurface, which we arbitrarily define as <10 m below the terrestrial surface. The practical and scientific implications of understanding microbial community structure and interactions among species at a mechanistic level, including species present at <0.1% of the community, in this environment are enormous [7,8]. These include greatly improved ability to (i) estimate greenhouse gas fluxes to and from the subsurface via modeling of microbially driven

## Trends

Datasets from subsurface samples can now be resolved into collections of complete or near-complete microbial genomes, yielding information about biogeochemical roles and mechanisms by which surface- and groundwater quality and atmospheric compositions are impacted.

Deep sequencing reveals extremely high levels of diversity in both the vadose zone and groundwater.

Many novel organisms have an extremely small cell size and small genome size, with restricted metabolic capability. Their growth is likely tightly linked to that of other community members.

Genomic analyses suggest that subsurface geochemical processes reflect the functioning of complex communities as opposed to a few dominant species.

Newly discovered microorganisms catalyze transformations relevant to greenhouse gases and processing of biologically critical elements.

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CO<sub>2</sub> fixation and degradation of organic matter, (ii) environmental management of mining impacts from toxic metals via a complete understanding of microbial metal redox cycles, and (iii) improved design of waste handling from animal confinements via the ability to accurately model nitrogen cycling in the subsurface. A key scientific impact is that we now 'see' all of the species in an environmental sample, including those from previously unknown lineages, and this informs major revisions of the tree of life [9]. Newly acquired data that reveal what specific microbes can and cannot do also impact our understanding of relationships among microbial species. Other benefits are perhaps less predictable but no less real, for example, the discovery of new biocatalysts from genes in the vast array of new microbial genomes.

We posit that recently discovered microorganisms from the terrestrial subsurface, now known almost exclusively from metagenomics, play key roles in subsurface biogeochemical reactions that control fluxes of climate-relevant gases (CO<sub>2</sub>, CH<sub>4</sub>, N<sub>2</sub>O, http://cdiac.ornl.gov/pns/ current\_ghg.html) including breakdown of buried organic carbon (Figure 1). Moreover, to understand the role of subsurface microbial communities in environmental processes it is



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Figure 1. Organic Carbon Deposits and Naturally Bioreduced Zones. These are created when organic matter accumulations, typical of most floodplains, are buried in the shallow subsurface (<10 m below the terrestrial surface of the earth). Genomic information on microorganisms in such zones provides information about their metabolic capacities, enabling modeling of subsurface biogeochemical processes. This will allow accurate prediction of how subsurface processes impact the quality of water and nutrient cycling within watersheds, as well as the flux of greenhouse gases to the atmosphere. The term 'watershed' here refers to a region or area of the terrestrial surface that is bounded by a divide and drains to a particular river or water body, including all the streams or rivers that drain into it. Watersheds are a key functional unit of the earth, governing water resources and mediating biogeochemical processes that support all terrestrial life. For details of the microbially catalyzed biogeochemical reactions that occur in the subsurface, see Figure 4.

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