

# A molecular dawn for biogeochemistry

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**Biogeochemistry is at the dawn of an era in which molecular advances enable the discovery of novel microorganisms having unforeseen metabolic capabilities, revealing new insight into the underlying processes regulating elemental cycles at local to global scales. Traditionally, biogeochemical inquiry began by studying a process of interest, and then focusing downward to uncover the microorganisms and metabolic pathways mediating that process. With the ability to sequence functional genes from the environment, molecular approaches now enable the flow of inquiry in the opposite direction. Here, we argue that a focus on functional genes, the microorganisms in which they reside, and the interaction of those organisms with the broader microbial community could transform our understanding of many globally important biogeochemical processes.**

## Introduction

Over the past 20 years, the field of biogeochemistry has substantially advanced our understanding of the processes controlling the cycling and distribution of elements on Earth. Key to accomplishing this task has been determining the major pools of biologically important elements, understanding the processes controlling flow among those pools, and using this information to model and predict patterns of elemental cycling within and among ecosystems. By doing so, we have compiled a relatively thorough biogeochemical inventory for many ecosystems as well as for the entire Earth. This has enabled scientists and society to understand the extent to which human activity has manipulated biogeochemistry at local, regional and global scales [1,2], knowledge that lies at the heart of efforts to reduce greenhouse gas emissions and coastal eutrophication [3].

## Moving beyond boxes and arrows

Most current perceptions and conceptions of biogeochemistry draw to mind box-and-arrow diagrams in which quantified pools of elements (boxes) are connected to one another by biological or physical processes (arrows) controlling flow among the pools (Figure 1). Hidden beneath these diagrams are the genetic and physiological attributes of the individual organisms that give rise to population dynamics and community-level interactions, all of which drive biogeochemical processes. However,

mechanisms operating at molecular, population and community levels can be absent from some conceptualizations of biogeochemical dynamics. This situation is particularly acute for microbial communities composed of archaea, bacteria and fungi, many of which mediate the biogeochemical cycling of carbon, nitrogen and sulfur across a range of spatial scales [4]. Microorganisms mediate key steps in biogeochemical cycles through the production of particular enzymes, which are encoded by functional genes (Table 1). Understanding the presence, abundance and expression of particular genes, as well as the identity of organisms in which they occur, could reveal the manner and extent to which molecular mechanisms regulate biogeochemical dynamics (Figure 2).

Prior to the advent of molecular microbial ecology, studies of microbial communities were limited to *in vitro* growth or by methods that treated microbial communities more or less as a homogeneous group. Microbial phylogenetics was at a standstill [5] and there had been little progress in the study of *in situ* microbial communities, much less relating physiological and community ecology to biogeochemical processes [6]. Hence, box-and-arrow conceptualizations of biogeochemistry were left unchallenged by microbial ecologists. Although we will continue to learn much from *in vitro* studies, molecular phylogenetics and functional genomics have revolutionized our ability to link microbial ecology to biogeochemical processes. Here, we provide evidence that molecular approaches make it possible to gain novel insight into the diversity of microorganisms mediating biogeochemical processes. We present a general approach and identify the tools necessary to link the function of microbial genes to community dynamics and biogeochemical processes. We then discuss how a molecular approach could provide new insight into the process of plant-litter decay, a globally important biogeochemical process that is mediated largely by heterotrophic microorganisms inhabiting soils and sediments. In doing so, we contend that biogeochemistry is awakening to a molecular dawn.

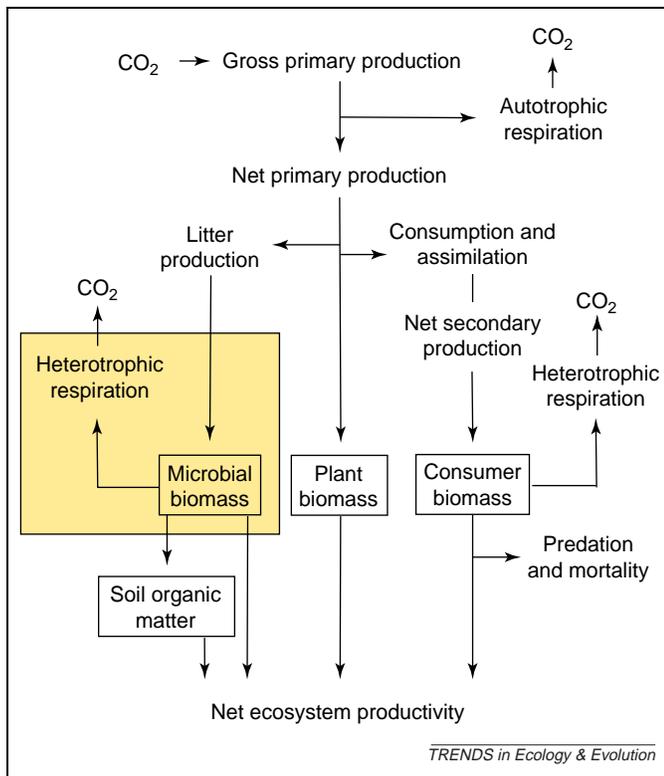
## Biogeochemical processes and the discovery of microbial diversity

### Discovery of microorganisms and genes

Whereas the natural history of plant and animal species has been studied for centuries, it took the development of molecular microbial ecology to uncover the diversity of microorganisms living literally beneath our feet [7]. The first realizations of the extent to which the microbial world had been unexplored came from ribosomal sequences (see

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**Figure 1.** The biogeochemical cycling of carbon in terrestrial ecosystems. The biogeochemical cycling of carbon is typically conceived of as a box (ecosystem pools) and arrow (processes controlling flow among pools) diagram. Using this approach, decomposer microorganisms (highlighted in yellow) have been conceptualized as a homogenous community catalyzing plant-litter decay, soil organic matter formation and the return of CO<sub>2</sub> to the atmosphere via microbial respiration. Hidden within this box-and-arrow conceptualization are diverse bacteria and fungi whose physiological attributes give rise to population and community dynamics, ultimately yielding a globally important biogeochemical process.

Online Supplementary Information, [8]), which rebuilt the tree of life and revealed novel microorganisms residing in most of the environments sampled. Entirely new microbial phyla without cultured representatives were found to be widespread in soil as well as in oceans, animal guts and other environments [9]. Recently, approaches have been developed that couple the discovery of microbial diversity directly to physiology and biogeochemistry. In a metagenomics study, Beja *et al.* [10] investigated the genetic

potential of an uncultured marine proteobacterium (SAR86) discovered through its ribosomal sequence. Their analysis revealed a gene for a rhodopsin-like protein (i.e. a photoreceptor) that had not been previously detected within bacteria. DNA sequences indicated that SAR86 was a globally distributed group, comprising up to 10% of marine bacteria and, moreover, it engaged in a previously unknown form of photolithotrophy or photo-heterotrophy [11]. The biogeochemical consequences of this finding are currently unknown, but they could alter our understanding of carbon cycling in the oceans [12].

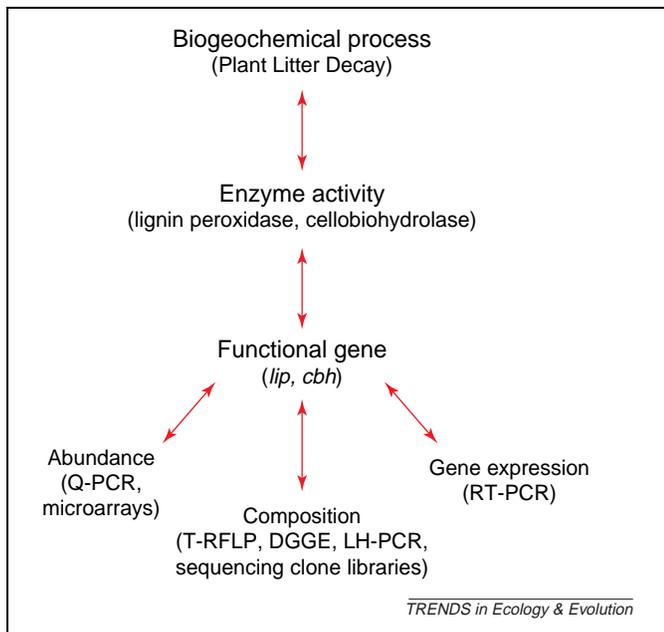
The approaches discussed here rely on targeting ribosomal and functional genes for analysis by using knowledge stored in molecular databases (Box 1). To avoid this bias, Venter *et al.* [13] used shotgun sequencing of a metagenome clone library from the Sargasso Sea. They demonstrated that bacterial rhodopsins were widespread and not restricted to the proteobacterial clade in which they were initially detected. The authors also identified 794 061 genes (i.e. 65% of the total) that could not be categorized according to their function [13]. Could these genes encode proteins that participate in important biogeochemical processes? Understanding the function of these uncategorized genes as well as other environmental sequences (e.g. ribosomal DNA sequences; see Online Supplementary Information) has reinvigorated attempts to isolate and study the diversity of microorganisms that we now know to exist in a wide range of environments [14]. Although we still have much to learn before the biochemical steps mediating any biogeochemical process are fully understood, molecular tools now make it possible to at least embark on this journey.

#### Uncovering novel biogeochemical function

Stable isotope probing (SIP) is an approach that can uncover novel microorganisms involved in a biogeochemical process, despite a lack of related DNA sequences in molecular databases. SIP avoids the need for *a priori* knowledge of functional genes that mediate a particular biogeochemical pathway, because an isotopically enriched substrate (e.g. <sup>13</sup>C or <sup>15</sup>N) is used to directly label the DNA of organisms participating in that pathway. SIP was

**Table 1. Examples of microbial genes and enzymes mediating biogeochemical processes**

Process	Enzyme	Genes	Refs
N <sub>2</sub> fixation	Nitrogenase	<i>nif</i>	[18]
Denitrification	Nitrite reductase	<i>nirK, nirS</i>	[38]
Nitrification			
NH <sub>4</sub> <sup>+</sup> oxidation	Ammonium monooxygenase	<i>amo</i>	[39]
	Hydroxylamine oxidoreductase	<i>hao</i>	[40]
CO <sub>2</sub> fixation	RUBP carboxylase-oxygenase	<i>cbbL, cbbM</i>	[41]
Methanogenesis	Methyl-coenzyme M reductase	<i>mcr</i>	[42,43]
Methane oxidation	Methane mono-oxygenase	<i>mmo</i>	[44]
Sulfate reduction	Dissimilatory sulfite reductase	<i>dsr</i>	[45]
Sulfur oxidation	Sulfite oxidoreductase	<i>sor</i>	[46]
Plant litter decay			
Lignin	Phenol oxidase	<i>lcc</i>	[47]
	Manganese peroxidase	<i>mnp</i>	[48]
	Lignin peroxidase	<i>lip</i>	[49]
	Glyoxyl oxidase	<i>glx</i>	[50]
Cellulose	β-Glucosidase	<i>bgl</i>	[36]
	Cellobiohydrolase	<i>cbh</i>	[36]
	Endoglucanase	<i>egl</i>	[36]



**Figure 2.** A conceptual model linking biogeochemical processes to functional genes via the activity of enzymes mediating key steps in biogeochemical pathways. Plant-litter decay is used as an example to illustrate how functional genes and enzymes control a particular biogeochemical process. Listed beneath the aspects of functional gene analysis are the molecular techniques that provide insight into the abundance, composition and activity of functional genes and the organisms in which those genes occur.

initially used to investigate microbial methane consumption in soil and suggested that methylotrophic activity occurs in Acidobacterium, a ubiquitous, but rarely-cultured bacterial phylum that was previously unknown to metabolize methane [15]. This approach has since been used in a variety of environments to demonstrate that metabolically active microorganisms are, in many cases, neither the most commonly studied nor even the most abundant [16,17]. In combination, metagenomics and SIP could yield important discoveries about active microorganisms and the diversity of biochemical pathways by which biogeochemical processes are carried out, although both have important limitations as well as advantages (see Table 2 and Online Supplementary Information). Although many biogeochemical investigations begin by studying a process (e.g. methane oxidation) and then focus downward to reveal the organisms, biochemical pathways and genes involved, molecular microbial ecology complements this approach by enabling movement in the opposite direction (Figure 2).

### Linking microbial genes to biogeochemical processes

Functional genes encode enzymes that catalyze key steps in biogeochemical pathways, and it is now possible to quantify their abundance, diversity and expression in the environment [18] (Table 1; Figure 2). This advance provides an opportunity to expand biogeochemical inquiry to genes encoding enzymes catalyzing key reactions in biogeochemical processes and to the community of microorganisms mediating those processes. As we have highlighted earlier, molecular advances also enable inquiry starting with genes (e.g. encoding bacteriorhodopsin) and ending with biogeochemical processes

### Box 1. Molecular databases

Many databases, and their associated search functions, have become standard tools in molecular biology and microbial ecology. Molecular databases are used by microbial ecologists to identify uncharacterized genes, or to develop assays targeting genes of a specific function or phylogeny. In the first case, sequence similarities can strongly suggest the function and phylogenetic history of the uncharacterized gene, or at least a set of probable scenarios. In the second, short oligonucleotide sequences can be used, through PCR or probing, to quantify the abundance and composition of organisms that are specifically targeted because they belong to a particular functional or taxonomic group.

The largest databases (e.g. GenBank) are warehouses where raw DNA or protein sequences of all types are deposited, and are comprehensive, but uncurated. Data obtained from such databases must be used with caution because of the potential for misinterpretation and annotation errors [52,53]. There are many smaller molecular databases created to provide summaries and interpretation of a variety of data and with varying levels of curation. The 2005 version of the Molecular Database Collection (a database of databases) includes 719 entries [54], but it is not a comprehensive list. Commonly used databases that are of relevance to microbial ecologists include:

- The National Center for Biotechnology Information (NCBI) website is home to GenBank and a variety of other databases and tools (<http://www.ncbi.nih.gov/>).
- The Ribosomal Database Project (RDP) maintains an aligned database of prokaryotic small-subunit (16S) ribosomal sequences (<http://rdp.cme.msu.edu/>).
- CAZy includes molecular sequences of carbohydrate-active enzymes cross-referenced by protein family and biochemical activity. (<http://afmb.cnrs-mrs.fr/CAZY/>).
- Pfam is a database of protein domains and domain profiles that can be used to categorize new sequences and examine protein structure (<http://www.sanger.ac.uk/Software/Pfam/>).
- BRENDA is a database of biological and biochemical data organized by enzyme function, with links to protein sequences (<http://www.brenda.uni-koeln.de/>).

(e.g. photolithotrophy or photoheterotrophy). We illustrate this conceptual flow with bi-directional arrows in Figure 2, and we contend that the ability to traverse levels of biological organization in both directions could transform our current approach to, and understanding of, biogeochemistry.

### Microbial functional groups

A key step in making this transformation is identifying functional groups within microbial communities, the enzymes of which mediate particular biogeochemical processes. The presence of a functional gene in a genome determines the membership of a microorganism in a functional group engaged in a particular biogeochemical process. Delineating functional groups using functional genes forces the researcher to reconceptualize the biogeochemical process in the context of the basic biological requirements of that process. This can reveal the complexity of some seemingly simple processes that in fact require the activity of a variety of functional groups (e.g. plant-litter decay). Although there is still unexplored diversity in microbial communities, functional genes performing the same task in disparate taxa are frequently related, falling into one enzyme family. This arises when functional genes are ancient evolutionary developments or have been the subject of horizontal gene transfer.

**Table 2. Methods for molecular microbial ecology<sup>a</sup>**

Methodology	Description	Advantages	Limitations
DGGE	Profiles microbial community composition based on differences between taxa in denaturation of a PCR-amplified gene	Compares community composition and relative abundance of sequences within a targeted microbial group without random subsampling of sequences (e.g. cloning)	Inappropriate for measuring diversity because only dominant community members can be detected. DNA with different sequences can migrate to same point in a DGGE gel, making sequencing of individual bands necessary for identification
Meta-genomics	Involves cloning and analysis of large genomic DNA fragments isolated from a mixed community. A metagenomic clone library can be screened for functional or taxonomic genes of interest, or sequenced by shotgun sequencing <sup>a</sup>	Results in discovery of new enzymes, metabolic pathways and organisms with impacts on biogeochemical processes. Detected genes are linked to their genomic context, which reveals further information about the potential niche, activity and life history of the organism	In a complex community, it is necessary to analyze an enormous clone library to overcome random sampling of many genomes. This approach is currently used for biological discovery, rather than quantitative hypothesis testing
Microarrays	Assembly of hundreds to thousands of oligonucleotide probes distributed in spots on a microscope slide, enabling simultaneous measurement of abundance of functional genes as well as ribosomal genes, which can be used for phylogenetic determination	Provides quantitative data on abundance of a target group	No information is obtained about diversity or composition within the target groups, although many groups and sub-groups can be targeted. Only genes for which <i>a priori</i> information is available can be targeted, and the genes are not placed in a genomic context. Can also be difficult to optimize for complex communities because there are many probes with varying optimal hybridization conditions
PCR	Use of targeted primers and a DNA polymerase to amplify a particular gene or region of DNA, normally performed <i>in vitro</i> . The gene might be of interest for phylogenetic determination or functional gene analysis	Enables analyses to be conducted on a particularly well-understood gene, and limits the analysis to a manageable amount of genetic variability	Only genes for which <i>a priori</i> information is available can be targeted. Horizontal gene transfer and duplicate genes can confound efforts to infer phylogeny when analysis is focused on single genes. Methods that rely on PCR are prone to a variety of artifacts that can arise in this process [51]
Q-PCR	Used to measure gene abundance by detecting the cycle at which a PCR product starts to accumulate exponentially	Provides quantitative data on abundance of a target group	Information is obtained about diversity or composition within the target groups. Also see PCR
T-RFLP	Profiles microbial community composition based on differences among taxa in the lengths of terminal restriction fragments of a PCR-amplified gene. Such fragments are labeled using a fluorescent primer and then separated by electrophoresis	Compares community composition and relative abundance of sequences within a targeted microbial group without random subsampling	Inappropriate for measuring diversity because only dominant community members are detected. Taxa with different sequences can have the same sized terminal restriction fragments, making analysis of parallel clone libraries necessary for identification

<sup>a</sup>For further details, see Online Supplementary Information.

Hence, some functional groups, such as nitrogen-fixing bacteria, can be defined by the presence of a single functional gene occurring across disparate taxa (Table 1). Other groups, such as denitrifying bacteria, include microorganisms with a few functional genes that have unrelated sequences but that catalyze the same biochemical reaction (Table 1). These functional genes, as well as others, are mechanistically linked to biogeochemical processes via the enzymes that they produce, the activity of which can be assessed by the use of a range of physiological assays, such as assessing the activity of extracellular enzymes or the use of stable isotope tracers.

Biogeochemical processes could be influenced by the abundance of a particular functional gene, the taxonomic composition of a particular function group (e.g. owing to differences in gene expression), or interactions between the functional group and the broader microbial community. Clearly, a larger population of organisms has a greater capacity for enzyme production. The abundance of

a microbial functional group can be assessed using quantitative polymerase chain reactions (Q-PCR, or real-time PCR), or by probing functional genes using DNA microarrays [19] (Table 2). For example, Hawkes *et al.* [20] used Q-PCR to estimate the abundance of ammonia-oxidizing bacteria in soil beneath a variety of plant species and found that the abundance of these organisms accounted for 52% of the variability of in gross nitrification rates.

Members of a functional group are likely to differ in the rate at which they perform a particular process and in their responses to environmental conditions, providing a potential link between functional group composition and rates of biogeochemical processes. However, the effect of species composition, diversity and evenness on ecosystem function (i.e. biogeochemical processes) is the subject of ongoing debate [21]. Insight into the composition, diversity and evenness of microbial functional groups can now be obtained by community analysis of

PCR-amplified functional genes. Cloning and then sequencing yields the most detailed picture, because sequence information is obtained directly. Community profiling approaches, such as terminal restriction fragment length polymorphism (T-RFLP) and denaturing gradient gel electrophoresis (DGGE), can also be used to compare the composition of dominant functional group members [22] in a larger number of samples (Table 2). We expect that the aforementioned approaches will provide new insight into how biogeochemical processes are influenced by the composition and diversity of microbial functional groups as well as the broader community of microorganisms in which particular functional groups reside.

Expression of a functional gene is also a crucial aspect of how microbes regulate biogeochemical cycles. Many microorganisms can remain dormant for extended periods and can grow quickly under favorable conditions. All organisms alter gene expression to some degree under different environmental conditions. If a subset of the organisms in an environment is active, then how can we identify which organisms these are? SIP can reveal which microorganisms are actively participating in a particular process (i.e. those which metabolize an isotopically labeled substrate and produce labeled DNA). Active microorganisms can also be studied through mRNA extracted from the environment, which is then subjected to reverse transcription and subsequent amplification (RT-PCR) [23]. mRNA transcripts are a clear indication of gene expression by a particular microorganism; however, transcripts have a lifespan of only minutes to hours [24], and careful consideration of this fact is necessary when interpreting this information (Table 2).

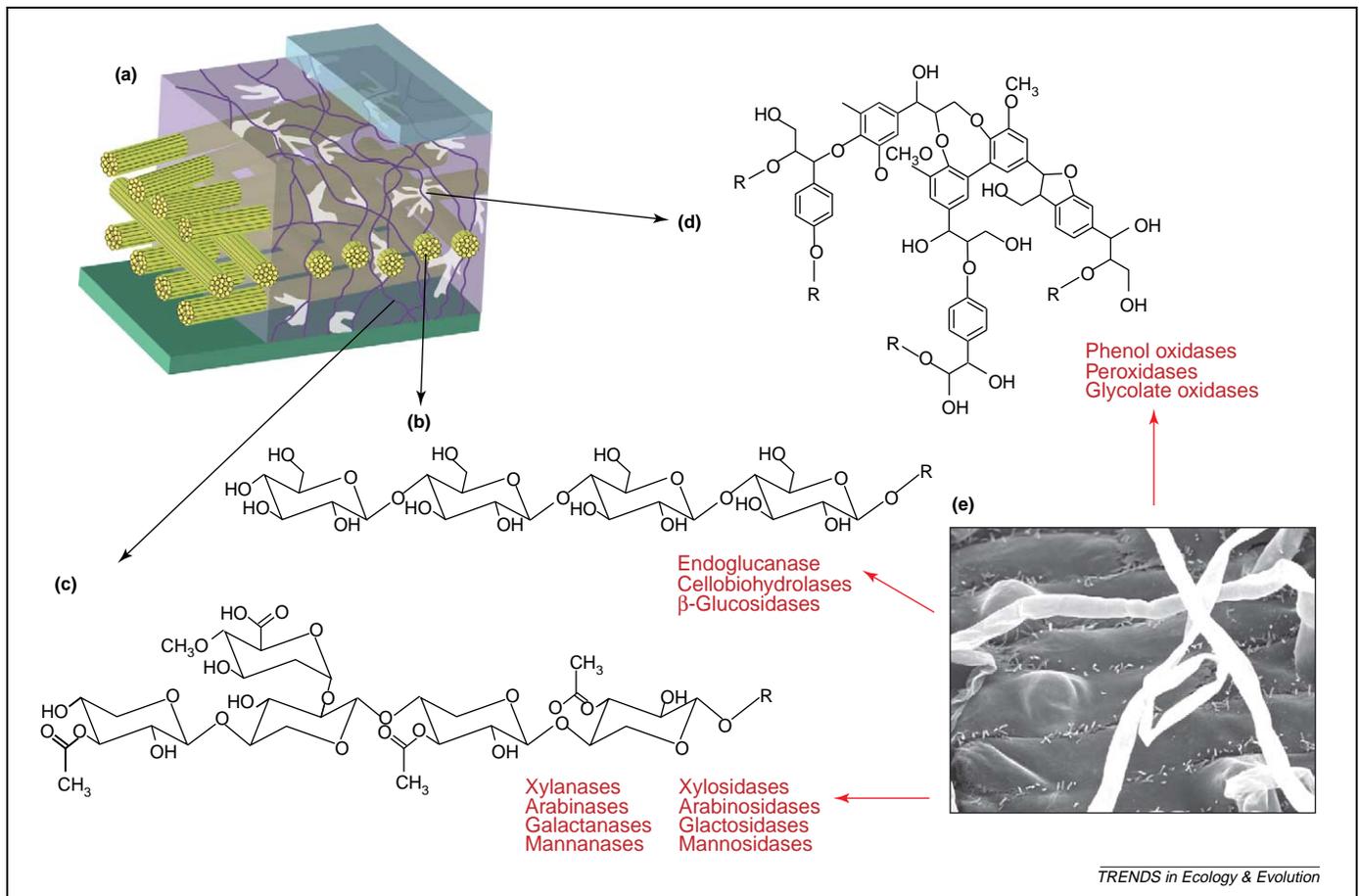
Functional genes exist in the context of genomes, which define the genetic potential of a microorganism and constrain its phenotypic traits. Species can be categorized into different functional groups depending on the process of interest, and it might be necessary to use a variety of species traits to understand fully the dynamics within a particular functional group [25,26]. We expect that functional gene assays will have the greatest power when genes are affiliated with their genomic context, including other functional genes, phylogenetic history, growth form, life history and habitat. Just as functional genes exist in the context of genomes, organisms in functional groups exist in the context of a broader microbial community. Members of a functional group undoubtedly interact with other microorganisms in a variety of ways, including as mutualists, competitors, parasites, or predators. Many of the approaches discussed here have been applied to ribosomal genes, which provide phylogenetic information that can be used to understand microbial community composition [22]. Although there is much to be learned by approaching biogeochemical processes from the perspective of functional genes, microbial functional group abundance and composition should be interpreted in a broader ecological context. Doing so will help unfold how genetic and physiological attributes of particular microorganisms give rise to population dynamics and community-level interactions, which ultimately drive biogeochemical processes.

### Plant litter decay: integration of molecular microbial ecology and biogeochemistry

The microbial breakdown of plant litter is a biogeochemical process of global importance and is largely mediated by microbial enzymes that disassemble the polymeric components of the plant cell wall (Figure 3). Inasmuch, decomposition provides an example where functional genes have direct implications for population-, community- and ecosystem-level dynamics. For example, it is well understood that a freshly fallen leaf harbors a microbial community broadly different from that occurring during the latter stages of decomposition [27]. But how do physiological attributes and life form influence microbial community composition as decay progresses? Do compositional shifts during the course of decay result in functional changes that, in turn, drive the biogeochemical cycling of carbon in the soil? Although these questions have been asked by microbial ecologists for decades, we can now address them in a manner that integrates across levels of biological organization by linking the occurrence and activity of individual populations to community dynamics and biogeochemical processes.

Plant litter is a chemically heterogeneous substrate for microbial growth, containing a range of organic compounds that serves as a substrate for microbial metabolism (Figure 3). To a large extent, heterotrophic bacteria and fungi enzymatically harvest energy from plant detritus by metabolizing carbohydrate polymers (e.g. cellulose and hemicellulose) in plant cell walls. These energy-rich compounds are often enmeshed by lignin (Figure 3), an aromatic polymer that conveys structural rigidity to cell walls, as well as protection against pathogenic attack. Once plant litter enters the soil, several classes of extracellular enzymes are deployed by soil fungi to depolymerize lignin, exposing hemicellulose and cellulose microfibrils from which energy can be derived by fungi and bacteria alike. Although many enzymes catalyzing the breakdown of plant litter occur across a range of microbial taxa, an increasing amount of genetic information makes it possible to assay the abundance of particular functional genes and organisms mediating key aspects of plant litter decay (Table 1).

White rot Basidiomycota and xylariaceous Ascomycota are the primary agents of lignin degradation, and these fungi synthesize several classes of extracellular enzymes that oxidatively depolymerize lignin by cleaving  $\beta$ -O-4 bonds [28] (Figure 3). Accordingly, lignin-degrading organisms comprise a well-defined microbial functional group; they conduct a key step during litter decomposition, and their genome contains functional genes that produce enzymes catalyzing a particular biochemical task. Lignolytic enzymes are well characterized, gene sequences are known for a variety of fungi [29] and primers have been developed to amplify genes encoding fungal phenol oxidase and manganese-peroxidase [30–32]. With this information, it becomes possible to quantify the abundance, composition and activity of these slow-growing fungi over the course of plant-litter decay, or in response to environmental factors. Furthermore, the ability to assess the activity of lignolytic enzymes in the environment [33], in combination with information about



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**Figure 3.** The plant cell wall (a) and the location and chemical composition of cellulose (b), hemicellulose (c) and lignin (d). Hemicellulose refers to a variety of carbohydrates and shown is *O*-acetyl-4-*O*-methyl-*D*-glucuronoxylan, which is common in angiosperms. Listed beneath each compound are the extracellular enzymes used by soil microorganisms to depolymerize the compounds during the process of plant-litter decay. The process of decomposition is mediated by a community of microorganisms (e), some of which are more or less active as plant detritus enters soil and particular compounds are successively depleted from that material. (e) Reproduced with permission from Frank Dazzo.

the abundance and occurrence of functional genes, provides an opportunity to discover how compositional shifts elicit functional responses that influence rates of decay. In short, it enables us to explore whether composition and function are linked in soil microbial communities and to better understand the implications of such a link on biogeochemical processes.

Although lignin degradation is mediated by a narrowly defined functional group, cellulose and hemicellulose are metabolized by a range of fungi and bacteria requiring an array of extracellular enzymes, each working synergistically on a particular chemical bond [34–36]. For example, the cellulase systems of fungi and bacteria include three well-described enzyme classes (i.e. endoglucanases, exoglucanases and glucosidases, [36]), each carrying out a particular biochemical task. Exoglucanases (cellobiohydrolases) cleave cellobiose from either the reducing or nonreducing end of the cellulose polymer, and these cellulolytic enzymes are predominantly produced by three gene families occurring across divergent microbial taxa. One cellobiohydrolase family resides in both bacterial and fungal taxa (GH6 family [37]) whereas others are exclusively bacterial (GH48 family) or fungal (GH7 family). The advent of molecular databases (Box 1) containing sequences for the aforementioned gene

families provides an opportunity to develop primers that selectively amplify these genes from genomic DNA extracted from litter or soil. By doing so, one can begin to answer questions regarding the occurrence and activity of particular cellulolytic fungi or bacteria as fresh litter passes through stages of decay, especially if the aforementioned approach is combined with physiological assays for cellobiohydrolase activity [33]. Moreover, phylogenetic markers for community-level analysis can also be amplified from the same genomic DNA. Conceptually, such an approach can integrate the dynamics of functional groups (i.e. lignin-degrading fungi or cellulolytic bacteria) within the context of the broader microbial community resident at a particular time during the decomposition of plant litter.

Different conceptual approaches have been used to understand plant-litter decay, ranging from mathematical models of mass loss, to critical ratios (carbon:nitrogen or lignin:nitrogen), to simulation models predicting the metabolism of operationally defined chemical classes. We have learned much from these approaches, but none are based on the underlying mechanisms within microbial communities that fundamentally control litter decay. Although it is parsimonious to simplify complex phenomenon such as plant-litter decay for heuristic purposes,

molecular approaches that we describe here provide the means to attain previously unforeseen insight into underlying physiological-, population- and community-level dynamics controlling a globally important biogeochemical process. Doing so should enable us to better understand the basic biology controlling litter decay and to evaluate more reliably the range of conceptual approaches that have been used to describe and predict it.

### Concluding remarks

Microorganisms in soil, as well as other environments, engage in an array of biochemical processes, interact with one another in a dynamic community and, in turn, mediate biogeochemical cycles that are of global importance. We have argued here that a focus on functional genes, the microorganisms in which they reside, and the interaction of these organisms with the broader microbial community are a necessary complement to more traditional forms of biogeochemical inquiry. Such an approach can reveal the underlying biological dynamics that are sometimes absent from box-and-arrow conceptualizations of biogeochemistry.

Previously, biogeochemical investigation began by observing a particular process, uncovering the enzymes mediating that process, and studying the microorganisms responsible, at least those that would grow in culture. The ability to sequence genes from the environment, identify their potential function and uncover previously unknown organisms having unanticipated metabolic capabilities could transform the manner in which we build our understanding of biogeochemical dynamics in many environments. These advances mark the molecular dawn for the field of biogeochemistry.

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### Supplementary data

Supplementary data associated with this article can be found at [doi:10.1016/j.tree.2006.04.003](https://doi.org/10.1016/j.tree.2006.04.003)

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