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Perspective

Microbial Ecology Research using Metagenomic Techniques

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PERSPECTIVE

The ability to sequence DNA directly from the environment had a long-term impact on microbial ecology. Here, we look at the novel insights into microbial life gained through the use of metagenomics, as well as the comprehensive collection of analytical methods that make exploring the diversity and function of complex microbial communities possible. While gene-centric and genome-centric approaches are altering our understanding of microbial functions in ecosystems, annotating functions, metagenome assembly, and binning in heterogeneous samples remains difficult. The development of new analysis and sequencing platforms that generate high-throughput long-read sequences and functional screening opportunities will aid in harnessing metagenomes to improve our understanding of microbial taxonomy, function, ecology, and evolution in the environment will aid in harnessing metagenomes to increase our understanding of microbial taxonomy, function, ecology, and evolution in the environment.

Over 20 years ago, the word "metagenomics" was coined, heralding a new era in the study of the intersection of biology and chemistry. Metagenomics has become a driving force for discoveries in microbial ecology and biotechnology, as well as a vital approach for investigating the microbial universe, since its first application in soil. Our ability to assess microbial diversity and functional potential in any ecosystem expanded tremendously as sequencing technology became cheaper, faster, easier to use, and greater throughput. The availability and competency of computational approaches are crucial for successful metagenomics applications. We emphasise the new knowledge gained from metagenomics applications in Earth's various ecosystems, as well as the analytical techniques that enable such findings, in this study.

Direct sequencing and analysis of DNA from microbial assemblages, known as metagenomics, has quickly become a standard tool for determining the functional potential of microbial communities. DNA is extracted, processed into libraries, and sequenced on short-read (Illumina, Roche 454, Ion Torrent) or long-read (PacBio, Oxford Nanopore)

systems in the most basic application. Quality control of sequence reads is the first step in any metagenomics analysis, and it seeks to reduce sequence bias and artefacts by deleting adapter sequences, low-quality bases calls, and contaminant sequences that aren't from the source environment. Stand-alone tools or web-based apps can be used to do gene-centric metagenome analysis. To infer functional gene abundances and distribution, read-based annotations require aligning predicted gene sequences to known genes. Users can quickly examine the sensitivity of different parameters and analytical methodologies to improve annotations by developing a stand-alone analysis capability, which requires local computational resources and bioinformatics expertise.

Web servers provide a user-friendly analysis platform for researchers of all levels of experience, but they are limited to modest data quantities, only deliver findings from a limited set of analysis tools, and analysis completion might take weeks to months (depending on the server load). Despite these constraints, systems like JGI IMG/M, MGRAST, CyVerse, and KBase facilitate the development and implementation of novel analytical tools for a large research community. Many academics now have access to a rising number of datasets and analysis tools thanks to metagenomics, which is already a well-established technology. Gene-centric metagenomics provides a better knowledge of microbial mechanisms driving biogeochemical cycles in ecosystems, especially when combined with RNA sequencing (metatranscriptomics) and protein identification (metaproteomics). For example, a metagenomic investigation of taxonomic and functional diversity in prairie soil microbiomes revealed how long-term agricultural practises can lead to the loss of keystone species, resulting in functional diversity loss.

The use of metagenomics, metatranscriptomics, and metaproteomics in combination allows for the discovery of microbial functions that control greenhouse gas emissions and distribution in climate-sensitive northern tundra soils. Furthermore, metagenomics was used to examine biochemical and environmental parameters influencing microbial activities in sandy sediments, revealing the

importance of temporal processes that result in frequent changes between H_2 -fermentation and H_2 -respiration processes. Soil metagenomes can be studied further to discover novel physiologically and ecologically significant enzymes. This approach, when combined with empirical testing, can be used to broaden the category of known enzymes in databases. Gene-centric investigations of seldom accessed natural and manmade habitats increase our knowledge of fundamental microbial activities beyond the highly complex and diverse soil and sediment microbiomes. Genes encoding proteins involved in repairing UV-induced DNA damage, as well as chemotaxis, germination, and heat-shock proteins, were found in abundance across diverse sample locations in North Africa, according to metagenomics of dust microbiomes. Patterns in zoonotic protist diversity in raw sewage were explored to better explain their dispersion in urban contexts, revealing that New York City sewers were home to a functionally comparable but phylogenetically heterogeneous protist community.

Plant production and health can be influenced by microbes found inside or on the surface of plant tissues (roots, stems, and leaves). Plant-associated microbes were found to have a large collection of carbohydrate metabolism functions

but fewer motility genes, implying that endophytic bacteria have access to a wide range of carbon substrates. Host DNA contamination can decrease the ability to sequence microbial readings in plant tissues, just as it can in other microbe-host systems. This can, however, be used to quantify microbial concentrations. Metagenomic investigations of Earth's ecosystems are difficult to perform, but they have the potential to reveal the whole range of microbial activity and ecology. While microbial ecologists delve into the novel information provided by metagenomes, metagenomic investigations of microbiomes will continue to evolve as DNA and RNA sequencing technology and accessibility improve. Long-read (>10 kb) sequencing technologies have a lot of promise for improving genome assembly and taxonomy and function assignment. These advantages, however, are limited by a high error rate (10–15%). Adapting current sequencing methods for absolute quantification of all chemicals within a microbial cell could help in the scaling of core and dynamic functionality complex microbiomes to broader biogeochemical and ecosystem level interactions that drive the Earth's material cycles. Overcoming methodological obstacles will help us learn more about microbial taxonomy, function, ecology, and evolution.