

Soil Metagenomics: Way to Unravel Microbial Treasure

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ABSTRACT

Soil metagenomics, the study of the collective genomes of soil microorganisms, addresses limitations in traditional culture-dependent methods by using culture-independent sequencing technologies. This field directly investigates microbial diversity, functional activities, and community structure in soil settings using methods including shotgun sequencing and high-throughput sequencing. To learn more about the roles that the soil microbiome plays in nutrient cycle and soil health, the procedure includes DNA extraction, sequencing, data processing, taxonomic classification, and functional annotation. Its uses in agriculture have revolutionized the field, especially in improving biological nitrogen fixation by targeted microbial insights and increasing crop output by controlling biotic stress. These advances support sustainable farming practices and promote soil health.

INTRODUCTION

Soil metagenomics is the study of the collective genomes of microorganisms present in the soil environment. This field emerged to address the limitations of traditional culture-dependent methods, which are unable to provide clear estimates of

microbial diversity and fail to tap into many microbial resources. By using culture-independent approaches, metagenomics enables the examination of microbial diversity, community structure, genetic relationships, and functional activities within their natural

habitat. The process of soil metagenomics involves advanced techniques such as functional gene screening or sequencing analysis. Sequencing technologies have evolved from shotgun sequencing to high-throughput, next-generation sequencing (NGS), and third-generation sequencing (TGS), each offering enhanced capabilities for detecting pathogenic microorganisms rapidly (Zhang *et al.*, 2021). These technologies, combined with new algorithms, improve the accuracy of taxonomic profiling and gene prediction in microbial species. Functional metagenomics further aids in identifying new bioactive substances and functional genes from soil microorganisms and their metabolites. Understanding the microbial diversity in soil is crucial as it plays a significant role in nutrient recycling, soil structure formation, and humus creation, all of which are vital for maintaining soil health. Enhanced strategies to examine RNA or DNA from soil have advanced the field by providing insights that traditional methods could not achieve. This growing body of literature supports the implementation of soil metagenomics in promoting agricultural sustainability and improving soil survey initiatives. Despite its benefits, soil metagenomics faces challenges such as the co-extraction of humic acid with nucleic acids, which can hinder downstream processes (Sabale *et al.*, 2020). Moreover, a comprehensive catalogue of functional genes and microorganisms for any soil is yet to be established, indicating the ongoing need for further research and methodological refinement in this field (Kaushik *et al.*, 2021).

Steps involved in soil metagenomics

Metagenomic techniques offer a comprehensive approach to identify and classify microorganisms within soil samples, allowing for a better understanding of soil microbial diversity and functions. It involves four major approaches: **A. Shotgun**

metagenomics. B. Activity Driven Study C. Sequence driven study D. Direct determination of whole collection. One of the primary methodologies involves sequencing the total DNA extracted from soil samples to capture the entire microbial community present. This process includes several key steps:

- 1. DNA Extraction:** The initial step involves isolating DNA from soil, which can be challenging due to the presence of humic acids and other contaminants that can inhibit downstream processes.
- 2. Sequencing:** Next, high-throughput sequencing technologies, such as Illumina, PacBio, or Oxford Nanopore, are employed to sequence the extracted DNA. These technologies generate vast amounts of data, enabling a comprehensive analysis of the microbial diversity present in the soil sample.
- 3. Data Processing and Assembly:** The raw sequencing data undergoes quality control and assembly into longer sequences, often referred to as contigs. Bioinformatic tools, such as assemblers and error-correction software, play a crucial role in this stage to ensure accurate reconstruction of microbial genomes.
- 4. Taxonomic Classification:** One of the significant challenges in soil metagenomics is the classification of microorganisms due to the complexity and diversity of soil microbial communities. Classifiers rely on reference databases to match sequences to known microorganisms. However, the paucity of soil-specific reference databases hampers accurate classification. To address this, researchers are developing more comprehensive databases and utilizing in-vitro mock communities derived from soil strains for benchmarking the accuracy of taxonomic classification.

5. Functional Annotation: Beyond identifying microorganisms, metagenomics also allows for the functional annotation of genes to understand the potential metabolic capabilities of the soil microbiome. This step involves comparing sequences against functional databases like KEGG or COG to infer the roles of different microbial genes within the ecosystem

The methodologies described above represent a robust framework for exploring the microbial landscape of soils. However, continuous advancements in sequencing technologies and bioinformatics are essential to overcome the current limitations and improve the accuracy of microbial identification and classification in soil metagenomics.

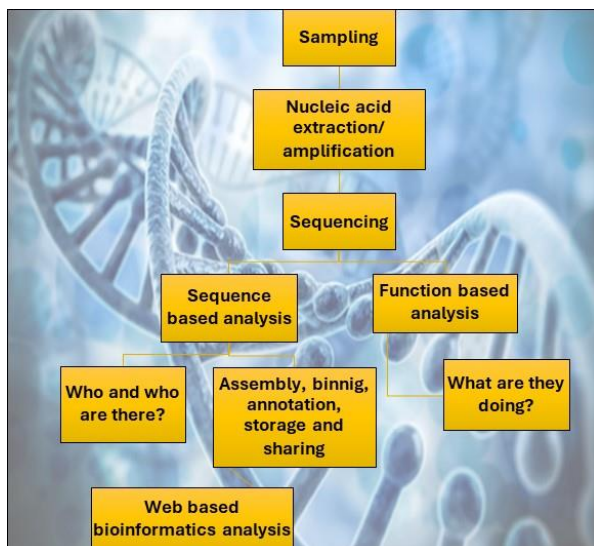


Fig. Steps involved in soil metagenomics

Applications in agriculture

Soil metagenomics has been successfully implemented in agriculture to enhance crop productivity and manage plant diseases. The use of soil shotgun metagenomes and expanded culture collections has allowed researchers to evaluate the global distribution and diversity of terrestrial diazotrophic microbiomes. This comprehensive understanding has facilitated the development of targeted agricultural practices aimed at

optimizing biological nitrogen fixation, a crucial process for plant growth and soil health. The evaluation of metagenomic classifiers specific to soil environments has also contributed to the advancement of soil metagenomics in agriculture. By developing soil-specific reference databases and in-vitro mock communities derived from soil strains, researchers have been able to improve the accuracy of taxonomic classification. This has led to better identification and utilization of beneficial soil microbes, thereby enhancing crop yields and reducing the incidence of soil contamination. Additionally, insights gained from both culturomics and metagenomics have been pivotal in the preservation and application of soil microbiomes for sustainable agriculture. These approaches have enabled the identification of microbial communities that support soil health and crop productivity. Implementing these microbial insights in agricultural practices has shown promising results in improving soil fertility and plant health, demonstrating the practical benefits of soil metagenomics in sustainable farming.

CONCLUSION:

Soil metagenomics overcomes the drawbacks of conventional techniques by offering a culture-independent means of comprehending the intricate microbial diversity and roles within soil ecosystems. Researchers can investigate the structure of microbial communities, genetic linkages, and functional genes essential to soil health using cutting-edge sequencing technologies like NGS and TGS. By discovering advantageous soil microbes, this field can improve crop productivity, control plant diseases, and advance sustainable farming, among other important agricultural uses. Ongoing developments in sequencing and bioinformatics continue to increase the precision of microbiological identification and classification in spite of obstacles including contamination problems and a lack of soil-

specific reference databases. All things considered, soil metagenomics has enormous potential to improve soil health and agricultural sustainability.

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