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Environmental Meta-Genome Biotechnology

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Introduction

Environmental meta-genome biotechnology is a new field in which genetic material is extracted directly from environmental samples such as soil, sludge, marine, deep sea, freshwater, wastewater, sewage water, food, human gut, insect gut, animals, etc. to understand their microbial community structures by passing the need for isolation and laboratory cultivation of individual microbial species. This new field of advanced molecular genetics research enables studies the vast majority of microbes on earth, of which more than 99% can not be cultured in the laboratory as well as provides the extensive information on the predicted gene functions and metabolic pathways of diverse environmental microbial communities. for example meta-genome biotechnology technique can be apply for identifying genes from yet-uncultured microbes which have very low or no similarity to known genes in the GenBank .

Advances in the field of genomics and meta-genomics have led to rapid and accurate strategies for the monitoring of microbial biodiversity and have revealed its potential for biotechnological applications. Some of the Meta-genomics techniques based on PCR amplification such as denaturing gradient gel electrophoresis (DGGE), terminal restriction fragment length polymorphism analysis (T-RFLP), rRNA gene clone library, etc., have been widely used for analyzing microbial community structures in various environments. These molecular biological methods avoid the limitation of cultivation and thus reflect the microbes in their environmental community more accurately. Such techniques are based on the detection of nucleic acids, as every microorganism holds unique sequences, which allows the differentiation of microorganisms within complex microbial communities.

PCR-DGGE

This meta-genomic technique examine microbial diversity based upon electrophoresis of small PCR-amplified DNA fragments (200-500 bp) on an acrylamide gel having a low to high denaturant gradient. In this meta-genomic technique DNA fragments of similar length but with different sequences can be separated according to their melting properties. Individual band can be excised from the gel and identified by sequencing. The main advantages of DGGE are that; it enables the monitoring of the spatial/temporal changes in microbial community structure and provides a simple view of the dominant microbial species within a sample. The DGGE technique has been used to examine the community of bacteria [1-4], yeast [5-8], and fungi [9] and also gene clusters [10].

T-RFLP meta-genome

T-RFLP analysis is one of PCR-based community profiling method that is commonly used for comparative microbial community analysis based on the position of a restriction site closest to a labeled end of an amplified gene. Briefly, marker genes are amplified by PCR using fluorescently labeled primers at their 5' end, followed by digestion using one or more restriction enzymes. Following the restriction digestion, the mixture of fragments is separated using either capillary or polyacrylamide electrophoresis in an automated DNA sequencer and the sizes of the different terminal fragments are determined by the fluorescence detector. Only labeled terminal restriction fragments are detected and their length heterogeneity indicates the complexity of the community visualized by an electropherogram. The major advantage of T-RFLP is the use of an automated sequencer which gives highly reproducible results for repeated samples. T-RFLP was used for the first time to the study of 16S rDNA diversity in environmental samples by Liu et al., [11], and has been adopted for the study of bacterial [12], and archaeal [13], communities using rRNA gene systems, and in the study of functional gene diversity like *amoA* genes [14].

rRNA clone library

This meta-genome technique is also based on PCR. The methodology implies the extraction of DNA from an environmental sample, amplification and cloning of the rRNA genes into a suitable vector, followed by sequencing and finally identification and affiliation of the isolated clone with the aid of phylogenetic software. The quality of DNA in terms of size, amount and presence of contaminants should be considered before the library is made as poor quality DNA can lead to low coverage of environmental genomes in the library therefore, purity and recovery of open reading frames from meta-genomic nucleic acids is necessary. Phylogenetic analysis of meta-genomic clone libraries bearing rRNA genes has led to the discovery of many new bacterial and archaeal phylotypes in the environmental samples . The method was applied to study the structure of microbial communities such as bacteria and archaeal [15], and yeast and fungi [16,17].

In conclusion, Meta-genome biotechnology is a new field which used to understand the microbial community structures by passing the need for isolation and laboratory cultivation of individual microbial species. Meta-genomics is also one of novel approach for engendering novel genes.

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