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Further Development and Applications of GeoChip 3.0 for Microbial Community Analysis

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Abstract

Microarrays constructed with the genes encoding key enzymes involved in various biological and geochemical processes are referred to as functional gene arrays (FGAs), or GeoChip. On the basis of GeoChip 2.0 (He et al. 2007), which contains 24,243 oligonucleotide (50mer) probes and covers >10,000 gene sequences in ~150 functional groups involved in nitrogen, carbon, sulfur and phosphorus cycling, metal reduction and resistance, and organic contaminant degradation, a new generation of GeoChip (GeoChip 3.0) has been developed. GeoChip 3.0 has several new features compared to GeoChip 2.0. First, GeoChip 3.0 cover about 47,000 gene sequences for 292 gene families, and such a coverage allows us to obtain more information about microbial communities and analyze more diverse environmental samples. Second, the homology of automatically retrieved sequences by key words is verified by HUMMER using seed sequences so that potential unrelated sequences can be removed. Third, GeoChip 3.0 includes phylogenetic markers, such as gyrB. Fourth, a software package (including databases) has been developed for sequence retrieval, probe and array design, probe verification, array construction, array data analysis, information storage, and automatic update, which greatly facilitate the management of such a complicated array, especially for future update. Fifth, universal standards have been implemented in GeoChip 3.0 so that data normalization and comparison of different samples can be conducted. Finally, genomic standards are also used to quantitatively analyze gene abundance. GeoChip was used to analyze effects of elevated CO₂ on belowground microbial communities at BioCON.

The results demonstrated that GeoChip 3.0 could provide more capability for studying biogeochemical processes and functional activities of microbial communities important to human health, agriculture, energy, global climate change, ecosystem management, and environmental cleanup and restoration. It is also particularly useful for providing direct linkages of microbial genes/populations to ecosystem processes and functions.

New features of GeoChip 3.0

- GeoChip 3.0 is more comprehensive, and it contains ~24,500 probes and covers about 47,000 (~10,000 for GeoChip 2.0) gene sequences of 292 gene families (~150 gene family on GeoChip2.0). Thus, GeoChip 3.0 will be more representative.
- The homology of automatically retrieved sequences by key words is verified by HUMMER using seed sequences so that potential unrelated sequences can be removed.
- A software package (including databases) has been developed for sequence retrieval, probe and array design, probe verification, array construction, array data analysis, information storage, and automatic update, which greatly facilitate the management of such a complicated array, especially for future updates.
- GeoChip has implemented a universal standard, which can compare different samples, and normalize data.
- GeoChip 3.0 implements a genomic control/standard, which can quantitatively analyze functional gene data.
- Automatic update greatly facilitates the management of such a complicated functional gene array.

Applications of GeoChip 3.0 to analyze effects of elevated CO₂ on belowground microbial communities at BioCON

24 soil samples were taken from 12 plots of high CO₂ (568 µmol/mol, red) rings (1, 3 and 5) and 12 plots of ambient CO₂ (384 µmol/mol, green) rings (2, 4 and 6) in July 2007 at the BioCON site as shown above.

CONCLUSIONS

1. GeoChip 3.0 has been constructed with more than 24,000 probes and covers about 47,000 gene sequences in 292 gene families. To our knowledge, this is the most comprehensive functional gene array currently available for environmental studies.
2. GeoChip 3.0 has many new features from rapid sequence retrieval and verification, probe design and validation, data comparison and normalization, quantitative analysis of functional gene abundance, to future updates.
3. GeoChip 3.0 has been used to analyze effects of elevated CO₂ on belowground microbial communities, and the results demonstrate that GeoChip is a powerful tool for analysis of biogeochemical, biological and environmental processes and associated microbial communities.