

研究论文

## PCR-DGGE技术用于湖泊沉积物中微生物群落结构多样性研究

赵兴青, 杨柳燕\*, 陈灿, 肖琳, 蒋丽娟, 马, 朱昊巍, 于振洋, 尹大强

污染控制与资源化研究国家重点实验室, 南京大学环境学院, 南京210093

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**摘要** 采用PCR-DGGE分子指纹图谱技术比较南京市玄武湖、莫愁湖和太湖不同位置的表层沉积物微生物群落结构, 研究表明, 三湖泊沉积物微生物的16S rDNA的PCR扩增结果约为 626 bp, 为 16S rDNA V3~V5 区特异性片段。玄武湖和莫愁湖表层沉积物中大约有20种优势菌群, 且同一湖泊不同采样点DGGE图谱的差异性不大, 细菌群落结构具有较高的相似性, 而太湖样品DGGE条带的数目和位置表现出明显差异, 且不同采样点图谱的差异性较大。三湖泊除具有特征性的微生物种属外, 还分布约5个相同的细菌种群, 可能与沉积物的理化性质和水生植被的影响相关。对DGGE图谱中7条主带进行回收、扩增和测序, 结果显示其优势菌群具有不同的序列组成, 其中5个序列与Genebank中已登录的细菌种群的同源性≥99%, 2个序列的同源性为96%和93%, 其中2个相似的细菌类群目前尚未获得纯培养。

**关键词** [沉积物](#); [微生物多样性](#); [变性梯度凝胶电泳 \(DGGE\)](#); [16S rDNA](#); [序列测定](#)

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## Study on the microbial diversity in lake sediments by the method of PCR-DGGE

ZHAO Xi ng-Qi ng, YANG Li u-Yan\*, CHEN Can, XIAO Lin, JIANG Li -Juan, M A Zhe, ZHU Hao-Wei, YU Zhen-Yang, YIN Da-Qi ang

State Key Laboratory of Pollution Control and Resource Reuse, School of t he Environment, Nanjing University, Nanjing 210093, Chi na

**Abstract** As a new technique for DNA fingerprinting, denaturing gradient gel electrophoresis (D GGE) can be used for microbial diversity analysis in environmental samples. In this study, PCR-D GGE was applied to analyze the microbial communities in lake sediments. Sediment samples fro m seven locations in three lakes were collected and their genomic DNA was directly extracted. T he DNA yield of sediments of Lake Xuanwu and Lake Mochou was high (10μg/g), while that o f sediments in Lake Taihu was relatively low. After DNA purification, the 16S rRNA genes (V3 t o V5 region) were amplified with two universal primers (F341GC and R907). These amplified D NA fragments were separated by parallel DGGE.

The DGGE profiles indicate different bands' patterns for different samples. There are five commo n bands in all lake sediment samples, which indicate that there are similarities among the populatio ns of microorganisms that exist in all lake sediments. But, the specific bands in sediment samples i ndicate that the microbial communities in different lakes are different. The DGGE profiles of Lak e Xuanwu and Lake Mochou are very similar and about twenty types of microorganisms are ident ified in the sediment samples of both lakes. These results suggest that the sediment samples of the se two city lakes (Xuanwu, Mochou) have similar microbial communities. However, the DGGE p rofiles of sediment samples in Lake Taihu are significant different from these two lakes. Furthermo re, the DGGE profiles of sediment samples in different locations in Lake Taihu are also very differ ent, which suggests that the microbial communities in Lake Taihu are more diversified than those i n Lake Xuanwu and Lake Mochou. The differences in microbial diversity may be caused by the d ifferent environmental conditions, such as redox potential, pH, and the concentrations of organi c matters, etc.

Seven major bands of 16S rDNA genes fragments from DGGE profiles of sediment samples wer e further eluted from gel, reamplified and sequenced. The sequences of these fragments were com

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pared with the database in GeneBank (NCBI). The results indicate that five sequences share 99% ~ 100% homology with known sequences (Bacillus and Brevibacillus, uncultured bacteria), while the other two sequences share 96% ~ 93% homology with known sequences (Acinetobacter and Bacillus).

These results indicate that the PCR-DGGE technique combined with sequences determination is a feasible and efficient method for microbial community analysis in sediment samples.

**Key words** sediment microbial diversity denaturing gradient gel electrophoresis (DGGE) 16S ribosome DNA (16S rDNA) sequence

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通讯作者 杨柳燕 [yangly@nju.edu.cn](mailto:yangly@nju.edu.cn)