

专论与综述

油藏微生物群落研究的方法学

谷峻¹, 石成芳², 吴晓磊^{1,*}, 赵俊义³

1.清华大学环境科学与工程系 北京100084 2.大庆油田勘探开发研究院, 大庆163414 3.大庆油田采油二厂, 大庆163414

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摘要 油藏微生物群落的解析和认知是开发和应用微生物采油技术的基础。利用各种提高油藏微生物可培养性的方法和非培养技术解析不同油藏微生物的群落结构、功能和多样性, 对定向调控油藏微生物群落、开发和应用有效微生物驱油技术具有重要的指导意义。通过调查新近发展的提高微生物可培养性的方法和措施以及不依赖于培养的分子微生物生态学技术, 总结了油藏微生物群落研究方法学的最新进展。提高微生物可培养性的方法和措施主要通过模拟微生物的生存环境, 减少富营养的毒害作用、添加信号分子维持微生物细胞间的作用和提供新型电子供体和受体等手段采用稀释法、高通量培养法等方法得以实现; 不依赖于培养的分子微生物生态学技术主要包括荧光原位杂交、末端限制性片段长度多态性分析、变性梯度凝胶电泳和构建克隆文库等技术。这些方法学的进展为更有效的获得各种油藏微生物资源、调控油藏微生物群落以提高石油采收率提供理论指导。

关键词 [微生物群落](#); [非培养技术](#); [微生物采油](#); [提高微生物可培养性](#)

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Progress in methodological research of microbial community in oil fields

Gu Jun¹, Shi Chengfang², Wu Xiaolei^{1,*}, Zhao Junyi³

1 *Department of Environmental Science and Engineering, Tsinghua University, Beijing 100084, China*

2 *Exploitation and Development Research Institute of Daqing Oil Fields Co. Ltd., Daqing 163414, China*

3 *Oil Product No.2 of Daqing Oilfield Co. Ltd., Daqing 163414, China*

Abstract

Understanding microbial community in oil reservoirs is crucial to application of microbial enhanced oil recovery (MEOR) technology. Much effort has been directed toward more insights into the microbial community in oil field, resulting in the development of culture dependent and independent techniques. Culture-dependent methods for enhancement of microbial culturability and culture-independent methods for analyzing microbial communities are reviewed in an attempt to better understand the recent progress in methodological researches on the oil field microbial communities.

Microbial community is analyzed conventionally by culture-dependent approaches which cultivate, enrich and isolate microbial cells, identify and count them, then figure out the microbial structure from the data of the cultivated microbial cells. However, the cultivating nutrients and conditions afforded in laboratory are too simplified and somehow far different from those in the environments where microorganisms live. Consequently, majority of microorganisms can not grow and be culturable. In addition, microorganisms grow at different speeds on the same media and under the same conditions, leading to different enrichment and increase of the microbial population. The relative amounts of the cultivated microorganisms are therefore different from the real ratio of the microorganisms in the environments. As a consequence, the pattern of microbial community structure obtained by the conventional culture-dependent methods is not true and not helpful for directing the

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e development and application of MEOR.

Two strategies, the increasing microbial culturability and culture-independent methods, are therefore developed and applied for getting more culturable microbial isolates and more insights into the real microbial community structure and functions. Increasing microbial culturability is to cultivate microorganisms with novel methods and under novel conditions. These novel cultivating conditions include: "oligotrophic" media instead of "rich" nutrients, adding signal molecules, novel electronic donating and accepting chemicals to media, etc.. The novel cultivating methods include dilution culture, high-through culturing, diffusion-growth chamber, cell encapsulation, sequence-guiding isolation techniques, etc. which can mimic environmental conditions and lead to more microorganisms cultivated. Although the novel cultivating techniques and conditions can make much more microorganisms culturable, they can not figure out the overall pattern of microbial community structure, therefore culture-independent approaches are therefore absolutely necessary for analyzing oil reservoir microbial community. Culture-independent approaches are based on the analyses of the functional genes, such as *mcrA* gene, and 16S rRNA and its gene (16S rDNA). The analyzing techniques contain FISH, T-RFLP, DGGE, clone library and sequencing etc.. Many researches with the culture-independent methods have showed the power of these methods and got many new ideas on the structure and functions of the microbial community in oil reservoirs. Moreover, it is a trend to apply both culture-dependent and culture-independent approaches to understand the functions of microorganisms for MEOR and structure of microbial community. With the microbial strains obtained from oil reservoirs and the knowledge of oil reservoir microbial community, MEOR can be developed and applied feasibly and reliably by manipulating the microbial community in the oil reservoirs.

Key words [microbial community](#) [culture-independent methods](#) [microbial enhanced oil recovery](#) [increasing microbial culturability](#)

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通讯作者 吴晓磊 xiaolei_wu@tsinghua.edu.cn