



## **Microbial diversity in methanogenic hydrocarbon-degrading enrichment cultures isolated from a water-flooded oil reservoir (Dagang oil field, China)**

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Microbial transformation of oil to methane is one of the main degradation processes taking place in oil reservoirs, and it has important consequences as it negatively affects the quality and economic value of the oil. Nevertheless, methane could constitute a recovery method of carbon from exhausted reservoirs. Previous studies combining geochemical and isotopic analysis with molecular methods showed evidence for in situ methanogenic oil degradation in the Dagang oil field, China (Jiménez et al., 2012). However, the main key microbial players and the underlying mechanisms are still relatively unknown.

In order to better characterize these processes and identify the main microorganisms involved, laboratory biodegradation experiments under methanogenic conditions were performed. Microcosms were inoculated with production and injection waters from the reservoir, and oil or  $^{13}\text{C}$ -labelled single hydrocarbons (e.g. n-hexadecane or 2-methylnaphthalene) were added as sole substrates.

Indigenous microbiota were able to extensively degrade oil within months, depleting most of the n-alkanes in 200 days, and producing methane at a rate of  $76 \pm 6 \mu\text{mol day}^{-1} \text{g}^{-1}$  oil added. They could also produce heavy methane from  $^{13}\text{C}$ -labeled 2-methylnaphthalene, suggesting that further methanogenesis may occur from the aromatic and polyaromatic fractions of Dagang reservoir fluids.

Microbial communities from oil and 2-methyl-naphthalene enrichment cultures were slightly different. Although, in both cases Deltaproteobacteria, mainly belonging to Syntrophobacterales (e.g. Syntrophobacter, Smithella or Syntrophus) and Clostridia, mostly Clostridiales, were among the most represented taxa, Gammaproteobacteria could be only identified in oil-degrading cultures. The proportion of Chloroflexi, exclusively belonging to Anaerolineales (e.g. Leptolinea, Bellilinea) was considerably higher in 2-methyl-naphthalene degrading cultures. Archaeal communities consisted almost exclusively of representatives of Methanomicrobia (mainly belonging to genera Methanosaeta and Methanoculleus).

As both syntrophic Bacteria and methanogenic Archaea are abundant in Dagang, the studied areas of this oil field may have a significant potential to test the in situ conversion of oil into methane as a possible way to increase total hydrocarbon recovery.