

Kotelnikova S., 2002, "Microbial production and oxidation of methane in the deep subsurface," *Earth Science Review*, 58/3, pp. 367-395.

The goal of this review is to summarize present studies on microbial production and oxidation of methane in the deep subterranean environments. Methane is one of the most active gases causing the "greenhouse" effect in the planet's atmosphere. Earlier, the deep oligotrophic subsurface was not considered as a source of biogenic methane. Evidence of active methanogenesis and presence of viable methanogens including autotrophic organisms were obtained for some subsurface environments including water-flooded oil fields, deep basalt aquifers, deep-sea hydrothermal vents, the deep sediments and granitic groundwater at depths of 10 to 2,000 meters below sea level. As a rule, the deep subterranean microbial populations dwell at more or less oligotrophic conditions. Molecular hydrogen has been found in a variety of subsurface environments, where its concentrations were significantly higher than in the tested surface aquatic environments. Chemolithoautotrophic microorganisms from deep aquifers that could grow on hydrogen and carbon dioxide greatly outnumbered those that could grow on organic compounds. As potential hydrogen consumers, methanogens can act as primary producers of organic carbon, initiating heterotrophic food chains in the deep subterranean environments independent of photosynthesis.

Produced methane may be oxidized to carbon dioxide with sulfate or oxygen. Microbial methane oxidation is a biogeochemical process that limits the release of methane, a greenhouse gas from anaerobic environments. Anaerobic methane oxidation plays an important role in marine sediments. Similar processes may take place in deep subsurface and thus fuel the deep microbial community. Organisms or consortia responsible for anaerobic methane oxidation have not yet been cultured, although diverse aerobic methanotrophs have been isolated from a variety of underground niches. The presence of aerobic methanotrophs in the anoxic subsurface remains to be explained. When the impact of the subsurface on global methane emission is estimated, it is necessary to take into account the activity of methane-oxidizing biofilters in shallow groundwater layers. Some studies have shown the presence of oxygen in the deep groundwater. Groundwater carrying oxygen down to depth increases productivity of the subsurface biota. Methane is consumed by methanotrophs on the way of its evolution in oxidized environments and is transformed to organic form, available for further microbial processing.

The presence of methane in the deep subsurface have been shown all over the world. Microbial production and oxidation of methane is involved in the carbon cycle in the deep subsurface environments. The flux of gases between the deep subsurface and the atmosphere is driven by the concentration gradient from the depth to the atmosphere. However, it is modulated to a large extent by methane oxidation associated with biofilms and in the water phase and taking place in the oxic recharge and shallow groundwater. Granitic geological formations are widely distributed over the Northern hemisphere. Methane is one of the most active gases causing the "greenhouse" effect on the planet's atmosphere. When the impact of granitic environments is estimated it is necessary to take into account the activity of methane-oxidizing biofilters in shallow groundwater layers.

Most probable viable counts attested the presence of methanotrophs in the deep granitic ground waters from the depths of 68-460 m below surface in Sweden and 600 m in Finland. Methanotrophs constituted from 0.1 to 35% of the total cell population. Methane oxidation was responsible for 0.4-57% of microbial oxygen reduction in the studied deep granitic groundwaters. Short term methane oxidation experiments carried out in sealed bottles with $^{14}\text{CH}_4$ showed that the $^{14}\text{CH}_4$ was transformed to $^{14}\text{CO}_2$ and biomass in the groundwater. Methane oxidation was accelerated by addition of a mineral solid surfaces. The methanotrophic biofilms attached to a granite were more active than the free-living cells. PCR analysis

has showed presence of methanol dehydrogenase (mxaF), soluble methane monooxygenase (sMMO), particulate methane monooxygenase (pmoA) genes and 16S rDNA genes specific for *Methylobacterium*, *Methylomonas*, and *Methylococcus* directly in the deep groundwaters. Methanotrophs of gamma-subgroup of *Proteobacteria* were associated with the aquifers and with the groundwater from the tunnel ponds. In accordance with membrane cytology and biochemical properties the studied cultured microorganisms were affiliated to *Methylocystis* and *Methylomonas* genera. One strain A3500 possessed enzymes of ribulosemonophosphate pathway and GC/GOGAT system for NH_4^+ utilisation. It was psychrophilic, halotolerant and microaerophilic. Methane oxidation of this organism was inhibited by oxygen at concentration of 0.3 μM . *Methylomonas* strain A3500 is highly adapted to the subsurface habitat.