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# 窒素・リンの飢餓状態が浮遊性微生物の好気的メタン生成に及ぼす影響
# Aerobic methane production by planktonic microbes under nitrogen and phosphorus starved conditions in a lake

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Lake ecosystems are now recognized as an important source of atmospheric methane ($CH_4$), which account for about 6-16% of global methane emission from natural sources. In lake ecosystems, it has long been believed that $CH_4$ is produced only in anoxic environments (e.g., lake sediments and anoxic hypolimnion) by anaerobic methanogens. However, recent empirical and experimental works have revealed that planktonic microbes can produce methane in aerobic water columns of oligotrophic lakes through the use of methylphosphonic acid (MPn) by C-P lyase enzyme under P-limited conditions. But, there is no study examining the effects of cell nutritional conditions (N starved or P starved) on aerobic methane production by bacterioplankton.

We performed the batch-culture experiments to identify the effects of cell starvation on the rate of aerobic methane production under nitrogen and phosphorus limited conditions. Planktonic microbes collected from well-oxygenated water of Lake Saiko (Yamanashi Prefecture) were incubated with a growth medium (BG-11) for several months and used for the starvation experiment to make their cells N-starved or P-starved conditions by removing either element from the BG-11 medium. Then, we added MPn and/or inorganic nitrogen ($N_i$) and inorganic phosphorus ($P_i$) to confirm the response of N-starved or P-starved microbes to such experimental additions.

The results showed that although the cell nutritional conditions did not affect the production of $CH_4$, nutrient balance of lake water (N excess or P excess) greatly influenced the aerobic methane production. First, we confirmed aerobic $CH_4$ production in the MPn addition treatment, suggesting the active C-P lyase catalysis that converts MPn to methane and inorganic phosphate. Moreover, we found that MPn + $N_i$ addition accelerated the aerobic $CH_4$ production. This is due probably to the fact that $N_i$ addition promoted the biosynthesis of C-P lyase and/or made lake water more P-limited condition (increase of N/P ratio); both may contribute to increasing the MPn utilization by microbes. However, MPn + $P_i$ addition did not increase the $CH_4$ production, indicating the opportunistic utilization of MPn alternative to $P_i$ under phosphorus limited conditions. The present results suggest that the input of excess N into lake ecosystems promotes the metabolism of MPn by planktonic microorganisms, which leads to increase of aerobic methane production in phosphorus-limited oligotrophic lakes.

Keywords: Aerobic methane production, C-P lyase, methylphosphonic acid, phosphorus and nitrogen starvation, planktonic microbes