

Development of methanotroph strains converting methane to succinic acid with high efficiency

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Recent development of mining technology enables to utilize CH<sub>4</sub> in shale gas as a cheap and abundant feedstock. Succinate, which is a precursor for various chemicals such as tetrahydrofuran,  $\gamma$ -butyrolactone, 1,4-butanediol, is of our primary interest in this study. *Methylobacterium* sp. DH-1 isolated from brewery plant sludge was suggested to have a strong potential for succinate production based on the existence of a full TCA cycle in the genome. In this study wild type and mutant strains were cultured in the fed batch reactor where mixture gas of methane and air was supplied continuously under either aerobic or O<sub>2</sub>-limited conditions. Under O<sub>2</sub>-limited condition the succinate production increased comparing to aerobic condition. When succinate dehydrogenase gene cluster (*sdhB*) was removed from the genome,  $\Delta$ *sdhB* strain showed specific productivity ~ 60% higher than wild type despite of decreased cell growth. To overcome the growth defect of  $\Delta$ *sdhB*, glyoxylate shunt genes were inserted into the genome. The mutant with glyoxylate shunt showed the highest succinate production, 176 mg/L. Further genetic manipulations are necessary to maximize succinate production of this strain.