Summary

Title; Molecular and physiological characterization of dehalorespiring microbial communities

(脱ハロゲン呼吸微生物群集の分子生物学的および生理学的特性)

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In this study, two dechlorinating cultures were obtained, YN3 culture (which dechlorinates the suspected carcinogen tetrachloroethene (PCE) completely into non-toxic ethene (ETH)) and KJ-TCA culture (which dechlorinates the suspected carcinogen 1,1,2-trichloroethane (112-TCA) completely into vinyl chloride (VC)). YN3 culture dechlorinates up to 800 μM of PCE dechlorinated completely into ETH within only two weeks. Thus, YN3 culture is a potential candidate for bioaugmentation technologies because of its marked chloroethenes (CEs) dechlorination activities. For KJ-TCA culture, the product of 112-TCA dechlorination is the proven carcinogen VC, this problem can be overcome, however, by combining both YN3 culture with KJ-TCA culture. Thus, impact of this thesis in environmental cleanup is not restricted to PCE–contaminated sites but also can be extended to 112-TCA contaminated ones, or sites contaminated with both.

Also in this study, the identification of the microbes comprising the YN3 cultures were characterized using metagenome analysis. The microbes comprising YN3 culture were found to be belonging to phyla *Actinobacteria*, *Bacteroidetes*, *Firmicutes* and *Dehalococcoides*. Eighteen *rdhA* genes, which encode the catalytic subunit of reductive dehalogenases, were identified in the *Dehalococcoides*-metagenome assembled in this study. In addition and using reverse transcription-quantitative polymerase chain reaction (RT-qPCR), four *rdhA* genes were functionally characterized to be involved in the PCE to ETH dechlorination. Two of these four *rdhA* genes, were newly introduced as a genes suggested to be involved in the CEs dechlorination and can serve as a markers for the predict VC to ETH dechlorination in a given contaminated site. A further analysis at protein level is required to confirm the integration of these two genes in the dechlorination process.

Finally, the experiments conducted in this study proved the enhancement of the dechlorination of *cis*-dichloroethene (*cis*-DCE) to ETH by co-culturing of strain YN3PY1 (a novel species of the genus *Bacteroides* isolated from YN3 culture) beside *Dehalococcoides* enriched culture obtained from YN3 culture, C4C4 culture. However, even with this enhancement the overall dechlorination speed was observed to be slow if compared to that of YN3 culture. This indicated that other experiments are required to judge the role of the other bacteria coexist in YN3 culture. Collectively, this study would contribute the development of bioremediation technology using

Dehalococcoides as a dehalogenator with the enhancement by the coexisting bacteria such as *Bacteroides*. This study provides potential candidates for *in situ* bioaugmentation for remediation of sites contaminated with PCE (using YN3 culture) and 112-TCA using (using YN3 and KJ-TCA cultures).