



TCE BIODEGRADATION BY MIXED BACTERIAL CONSORTIUM ENRICHED FROM RIVER SEDIMENT AND FIRST STEP STUDY OF MOLECULAR IDENTIFICATION OF THE CATABOLIC REDUCTASE

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Introduction.

Trichloroethene (TCE) is one of the most chlorinated ethenes pollutants in most of under groundwater and soil. It causes critical problems for human health and the environment. It has strongly persistent chemical structure and therefore, elimination is very difficult. Despite of that, TCE can be degraded by microorganisms under anaerobic conditions. In this study, a mixture consortium of bacteria were isolated and demonstrated that they supported one another to dismiss TCE completely from 11ppm initial concentration by reductive dehalogenation to dichloroethene (DCE), vinyl chloride (VC), and ethene.

Methods.

The inoculum source taken from river sediment were inoculated to a reducing media (reference). Four different carbon sources: glucose, acetate + H₂, methanol, lactate were used to select the best condition for TCE degradation activity. Initial TCE 11ppm were injected by Aligent syringe to each bottle. Bacteria were grown in 165ml glass serum bottles crimp-sealed with Teflon-lined rubber stopper to avoid TCE leaking. Every step was carried out in the anaerobic chamber to maintain strictly anaerobic condition. Gas – Headspace method by GC/FID (Gas chromatography/ flame ionization detector)–6890N machine was used to analyze the concentration of TCE. To conduct enzyme assays, the mixture were grown in 4litter of reducing media to harvest enough enzyme fraction. The test was carried out by addition of gaseous TCE to the enzyme supernatant. Genomic DNA were also isolated and target DNA segment were amplified by PCR (Primers T7f and M13r were used), cloned by using pCR2.1-TOPO vector, transformed to One Shot TOP10 competent *E. coli* cells. Finally, the sequencing of the plasmid inserts were determined.

Results.

From initial concentration 11ppm, TCE were found completely degrade during 35 days in sample used acetate as the sole carbon source, meanwhile TCE removed efficiency in the others samples used Methanol, Lactate and glucose were 47.1%, 42.9%,30.8%

respectively.

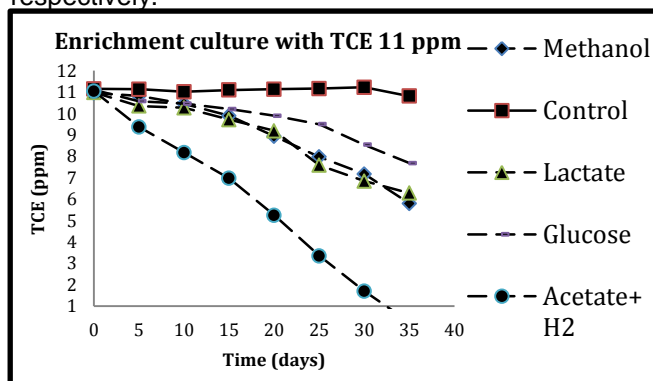


Fig 1. TCE degradation by using reducing media with 4 different carbon sources: Methanol, Lactate, Glucose and Acetate 10mM after 35 days, initial TCE concentration 11ppm were injected.

Conclusions.

The enriched mixed bacterial consortium showed the evidence that the bacteria can be able to degrade the TCE anaerobically. Acetate with H₂ gas as electron donor showed the good potential of TCE degradation. Enzyme TCE reductase has been considered as an essential factor to completely remove TCE from contaminated site.

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