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## INTRODUCTION

Biological process is one of the promising technique for remediation of chlorinated hydrocarbons contaminated soil and water. Some researchers have reported PCE and TCE dechlorination under methanogenic conditions via substitution of chlorine atoms by hydrogen ones. However, mostly resulted in the accumulation of vinyl chloride, which is even more toxic than PCE and TCE. In this study, PCE and TCE biotransformation pathways which were supported by  $H_2$  gas and/or organic hydrogen such as glucose, acetate and formate were investigated experimentally using methane fermentation mixed culture which had ability to dechlorination of PCE and TCE. The effect of metabolites such as vinyl chloride on PCE and TCE biodechlorination was also studied.

## MATERIALS AND METHODS

### Inoculum source

Mixture of anaerobic sludge obtained from a municipal wastewater treatment facility and anaerobic sediment from a lake near Ashikaga city, was used as inoculum source for the culture. Previous the experimentation, the methane fermentation mixed culture was cultivated under semi-batch operation for 4 months feeding organic substrate and PCE at 5 mg/l nominal concentration.

### Procedure

Dechlorination pathway was investigated using 22 ml suspensions culture in 32-ml serum vials sealed with teflon-lined septa, incubated with shaking in the dark at 35°C in an inverted position. Reduction conditions in the vials were maintained by the addition of  $Na_2S$ . Experimental conditions were shown in Table 1. The headspace gas was analyzed by gas chromatography equipped with ECD and FID to measure PCE, TCE and their conversion products. After initial dose of chlorinated compounds was recover as DCE, VC and/or ethylene, a part of the supernatant was removed and new solution containing organic substrate (RA, RB) or not (RC) was fed. The culture solution was purged via a cannula with  $N_2$  for 1 min to maintain anaerobic conditions, to remove chlorinated organic compounds and to prevent potential inhibition of the culture because of accumulation of DCE, VC and/or ETH. Before sealing the vials, hydrogen gas, PCE and/or TCE were fed.

Table 1. Experimental conditons  
 Effect of Hydrogen donor on PCE and TCE dechlorination

REACTOR	PCE (mg/l)	TCE (mg/l)	hydrogen donor	TOC (mg/l)
RA	5	4	$C_6H_{12}O_6$ , $CH_3COOH$ , $HCOOH$	70
RB	5	4	$C_6H_{12}O_6$ , $CH_3COOH$ , $HCOOH$ $H_2$ gas	35
RC	5	4	$H_2$ gas	0

\* Methane fermentation bacteria, MLVSS = 2000 ppm, temp. = 35°C

## RESULTS

### Effect of hydrogen donor on PCE and TCE bio-dechlorination.

Effect of hydrogen donor on PCE and TCE dechlorination was shown in Fig. 1. In reactor RA.III, 91.9%

of initial dose of chlorinated compounds was recovered as vinyl chloride within 10 days. However in reactor RC.III, 63.3% of the initial dose was recovered as DCE (mainly cis-DCE) and vinyl chloride was not recovered. This results suggested that hydrogen gas used as electron donor would support PCE and TCE dechlorination to DCE mainly. When hydrogen gas was used as electron donor, the reaction rates observed were smaller than those observed when organic compounds were used as energy source and electron donor.

### Effect of vinyl chloride on PCE and TCE biodechlorination.

Effect of vinyl chloride on PCE and TCE biodechlorination was shown in Fig. 2. Since vinyl chloride was accumulated as metabolite in the third addition batch experiments and the cultures were exposed to vinyl chloride rich condition for about 10 days (RA.III), the inhibition effect of vinyl chloride can be observed by comparing the results in the third and in the fourth batch test. In the reactor RA.III, at the day 10, neither PCE nor TCE remained. However in the reactor RA.IV, 20.8% and 2.2% of the initial doses remained as PCE and TCE respectively. It seems that vinyl chloride inhibits PCE degradation but not TCE.

### CONCLUSIONS:

1) PCE and TCE were transformed mainly to vinyl chloride when dechlorination was supported by hydrogen generated by oxidation of organic compounds fed as energy source and electron donor, whereas, they were transformed to DCE when hydrogen gas was used as sole electron donor.

2) PCE dechlorination was inhibited by vinyl chloride, but not TCE dechlorination

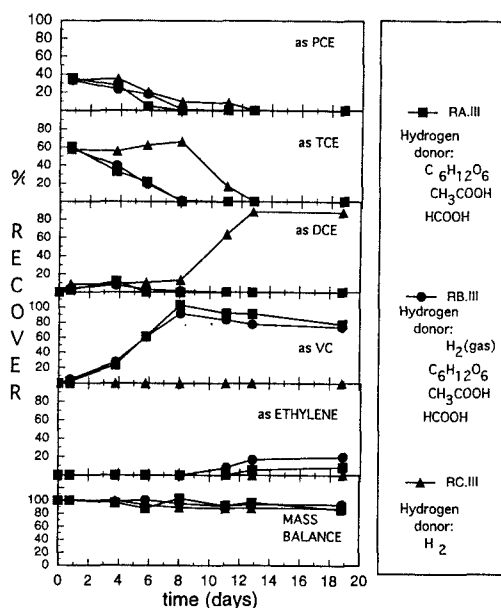


Fig. 1 Effect of hydrogen donor on PCE and TCE dechlorination

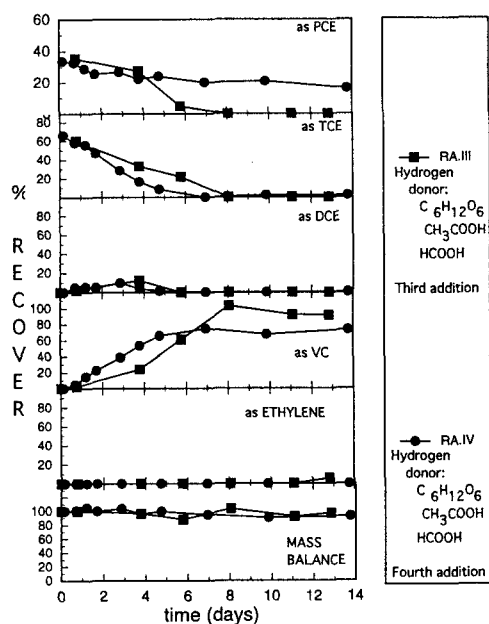


Fig. 2 Effect of vinyl chloride on PCE and TCE dechlorination supported by organic substrates