Modeling transport and biodegradation of PCE in sandy soil

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Abstract

Transport of microorganism activating tetrachloroethylene (PCE) dechlorination was well described by mobile-immobile two-region model from laboratory column tests. Biological reaction was identified as first-order kinetics from batch tests where the bacterium strain was mixed with halogenated aliphatic compounds of PCE or TCE (trichloroethylene) as initial pollutant. PCE dechloroeneation to dichloroethylene (DCE) via TCE was mathematically expressed as Michaelis-Menten equation while microbial growth rate did not show good performance due to Monod function.

1 Introduction

It has been reported more than one thousands of sites were polluted by halogenated aliphatic compounds, such as tetrachloroethylene (PCE), trichloroethylene (TCE), dichloroethylene (cis-1,2-DCE, trans-1,2-DCE and 1,1-DCE), vinyl chloride(VC), etc., in the environment white paper of Japanese Government. These chemicals were commonly used for industrial and domestic purposes until the use was prohibited by the regulation of water quality protection in 1989. Halogenated aliphatic compounds are characterized by its large amount of unit weight, small viscosity and volatility. It easily infiltrates into ground when its being spilled away on the surface. Several rehabilitations have been reported as success manner for remediation of soil and groundwater contaminated by PCE or TCE.
Bioremediation is one of inexpensive rehabilitation practices to be available for removal of pollution at low concentration close to the Japanese environment standard, however, with spreading large area. There are several barriers to be overcome in realization of biological techniques as in-situ remediation. We have to have know-how about distribute manner of bacteria and/or nutrient into wide area of ground, and to know microorganism and nutrient transport characteristics through complicated soil pores. There is still lack of well findings on behavior of bacteria and nutrient in actual flow region.

The purpose of this study is to stimulate basic understandings of microorganism transport and fundamental description of biodegradation of PCE via TCE to DCE for setting up bioremediation practices. One of the authors has succeeded in isolating PCE-degrading bacterium from mixed culture under anaerobic condition. In the present study, bacterial transport and biological dehalogenation was investigated from laboratory tests using the bacterium identified as *Clostridium bifermentans* DPH-1 [1].
2 Bacterial Transport through Saturated Sand

Transport of the bacteria was characterized from laboratory column tests with steady state flow condition in fully saturated Toyoura sand of which dry density is 1.55g/cm³. Constant discharge rate was maintained at 0.1 or 3.5 mL/min by using a double plunger pump during the tests. The column size is 10cm in length and 2 cm in inner diameter as shown in Figure 1. Bacterium solution was supplied to the bottom and flow out the top through the sand column. The test conditions were shown in Table 1.

Breakthrough curve in Run-7 was shown in Figure 2 as relationship between relative concentration \( C/C_0 \) describing the ratio of concentration to the initial and pore volume (PV) describing the ratio of total discharge to water contained within soil pore. Measurements are open circles. The knowledge of miscible displacement tells that breakthrough curve passes through \( C/C_0=0.5 \) at PV=1.0. The bacterial transport consumes more PV to reach at \( C/C_0=0.5 \). Fluctuation at large amount of PV may take place due to pores clogging by microorganism. The size of
Clostridium bifermentans DPH-1 is about 2.5 μm in long which is larger than average pore diameter of the sand. The bacteria may choke up small pore where water does not smoothly move. Change of C/C₀ is caused from buildup and breakdown of bacterial clogs in small pore.

The conventional transport models were fitted to the measures to check the applicability to assessment of bioremediation in actual subsurface. Dashed line was the first-order kinetic model (FKM) considering the flow in the column as complete mixing flow. Real line was advection-dispersion model (ADM) with chemical reaction where pore-water flows in the whole area of soil pore. Dotted line was two-region model (TRM) originally proposed for solute transport through unsaturated particulate soil [2]. The measures in Run-3 were not successfully fitted by ADM and TRM. Fitting results in Fig.2 indicate that ADM is difficult to simulate bacterial transport while TRM shows good performance. TRM is featured by four parameters, Peclet number Pe, Retardation factor R, fraction of mobile water β and Stanton number ω. The parameters were identified from the best fitting to the measures by the least square method as shown in Table 2.

Peclet number expresses spatial distribution of the bacteria in the sand column. It takes almost constant independently on discharge rate and the bacteria concentration in the solution. This is identical to miscible displacement using non-reactive tracer. The amount of dispersion coefficient depends linearly on discharge rate. Peclet number relates to dispersion coefficient as follows;

\[ Pe = \frac{vL}{D} \]  

in which \( v \), \( L \) and \( D \) are flow velocity in mobile region, column length and dispersion coefficient, respectively. Peclet number is constant when dispersion coefficient linearly depends on velocity.

Retardation factor tends to increase as the initial bacteria concentration increases. On the contrary, fraction rate of mobile water decreases with increase of the initial concentration. These findings indicates retardation factor and fraction rate of mobile water strongly depends on mass of microorganism in pores. These are independent of pore water velocity. The fraction rate takes a constant value of 0.14 when the initial concentration of bacteria solution becomes more than 10 mg protein/L.

Stanton number is defined as;

\[ \omega = \frac{aL}{q} \]  

in which \( q \), \( a \) is discharge velocity (Darcy velocity), mass transfer coefficient, respectively. The equation (2) defines ratio of advection time scale to bacteria diffusion into micro-pore via mass transfer. There is no evidence enough to show dependence of Stanton number on discharge velocity. It linearly increases with the initial bacteria concentration. It takes long time in convection when initial bacteria concentration becomes large. Clogs of microorganism prevent smooth flow in mobile water and diffusion into micro-pore.
3 PCE and TCE Dechlorination

Performance of a PCE and TCE-degrading bacterium from the highly enriched culture was checked using batch procedure. *Clostridium bifermentans* DPH-1 isolated from ditch sludge [1] was used as the source of dechlorinating bacterium. Batch procedure was done by adding PCE or TCE solution to mix culture with growth medium, yeast extract and the bacterium. Cultivation was performed at 30°C. Measurements were made for identifying PCE or TCE dechlorinating speed and the bacteria growth rate. The tests were carried out at 5, 50 and 150 mg/L of initial concentration of PCE and 2, 20 and 60 mg/L of initial concentration of TCE. PCE degrading was shown in Figure 3 of which case is 50 mg/L of the initial PCE concentration. The figure indicates PCE degradation, via TCE to DCE and increase of cell protein. PCE concentration decreased at 0.022 mg/L in 54 hours. The Japanese environment standard is less than 0.01 mg/L for PCE. The test results will pass the standard when operating time is expanded.

For the case of 50 mg/L of PCE concentration, TCE decreased to zero in 54 hours while it increased at 1.1 mg/L in 24 hours later. Other cases showed that TCE concentration smoothly became to zero in 72 hours. TCE degrading speed is rapid rather than PCE and DCE. The mass balance was confirmed from decrease of PCE and increase of DCE. The amount of DCE concentration measured in 72 hours was 25.7 mg/L while that estimated from decrease of PCE concentration 29.2 mg/L. The difference comes from the measurement by ECD-GC for PCE in 72 hours and FID-GC for DCE.

The bacteria concentration was shown in Figure 4, which showed that *Clostridium bifermentans* DPH-1 does not increase with the increase of PCE concentration. The bacterial concentration was estimated from optical density measured by UV spectrophotometer. The concentration, however, gets to be larger than the control. Then, the figure showed the bacterial increase at least relates to PCE concentration.
PCE dechlorination kinetics to DCE via TCE was studied in the batch test series. Activity of PCE dechlorination was assumed to be effective in 60 minutes. The kinetic parameters for PCE degradation were estimated from the Lineweaver-Burk transformation of the Michaelis-Menten equation. Michaelis-Menten equation is given by

$$v = \frac{v_{\text{max}} [S]}{K_m + [S]}$$

in which \(v\) is degrading rate, \(v_{\text{max}}\) is maximum degrading rate, \(K_m\) is Michaelis constant and \([S]\) is substrate concentration.

For PCE, \(v_{\text{max}}, K_m\) was 0.591 mg/hr/mg protein, 242.12 mg/L, respectively. For TCE, \(v_{\text{max}}, K_m\) was 0.222 mg/hr/mg protein, 13.84 mg/L, respectively. The growth rate of the bacteria could not be described in Monod type function [3]. The empirical description was extracted from the batch tests as follows;

For PCE,

$$\mu_p = -0.0015[PCE] + 0.5064$$

and for TCE,

$$\mu_t = \frac{0.163}{[TCE]} + 0.435$$

in which \(\mu_p\) is bacteria increase rate in PCE-degrading, \(\mu_t\) is bacteria increase rate in TCE-degrading, \([PCE]\) is PCE concentration and \([TCE]\) is TCE concentration.

The bacterial increase rate in PCE-degrading was different from TCE-degrading in the present study.
4 Numerical Bio-assessment Applications

4.1 Modeling microorganism transport

Bacteria transport was modeled by TRM in accordance with considerations on laboratory column tests in the preceding section. For mobile zone,

\[
\theta_m \frac{\partial x}{\partial t} + \frac{\partial}{\partial z} \left( \theta_m v_m x_1 \right) = \frac{\partial}{\partial z} \left( \theta_m D_1 \frac{\partial x_i}{\partial z} \right) + \mu_1 x_i - \mu_1 x_i - \alpha(x_i - x_{i,m})
\]

(6)

and for immobile zone,

\[
\theta_{i,m} \frac{\partial x_{i,m}}{\partial t} + \rho_d \frac{\partial S_{i,m}}{\partial t} = \mu_2 x_{i,m} - \mu_2 x_{i,m} - \alpha(x_i - x_{i,m})
\]

(7)

in which \(\theta_m, \theta_{i,m}\) is volumetric water content of mobile zone, immobile zone, \(v_m\) is average pore water velocity of mobile zone, \(\mu_1, \mu_2\) is bacterial increase rate which is given by the preceding section, \(\mu_1, \mu_2\) is decrease rate by death, respectively.

4.2 PCE transport with biodegradation

PCE transport characteristics were determined from NaCl tracer test by the column used in bio-reactor. TRM was fitted to measured breakthrough curve to identify the parameters as shown in Figure 5.

For mobile zone,

\[
\theta_m \frac{\partial C_i}{\partial t} + \frac{\partial}{\partial z} \left( \theta_m v_m C_i \right) = \frac{\partial}{\partial z} \left( \theta_m D_z \frac{\partial C_i}{\partial z} \right) + V_{max} \frac{C_i}{K_{s_i} + C_i} x_i - \alpha_2 (C_i - C_{pce})
\]

(8)

For immobile zone,
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\[
\theta \frac{\partial C_{\text{PCE}}}{\partial t} + \rho d \frac{\partial S_2}{\partial t} = -V_{\text{max}12} \frac{C_{\text{PCE}}}{K_p + C_{\text{PCE}}} x_{\text{im}} + \alpha_2 (C_1 - C_{\text{PCE}}) \tag{9}
\]

In which \( V_{\text{max}1}, V_{\text{max}12} \) is maximum degradation rate for PCE which is given by preceding section, \( C_1, C_{\text{PCE}} \) is concentration of PCE in mobile, immobile zone.

4.3 TCE transport with biodegradation

TCE and DCE transports were modeled as the same description of PCE. For mobile zone,

\[
\theta \frac{\partial C_{\text{TCE}}}{\partial t} + \frac{\partial}{\partial z} \left( \theta_n V_{\text{m}} C_{\text{TCE}} \right) = \left( \theta_n D_n \frac{\partial C_{\text{TCE}}}{\partial z} \right) + V_{\text{max}21} \frac{C_2}{K_2 + C_2} x_{\text{m}} + V_{\text{max}11} \frac{C_1}{K_1 + C_1} x_{\text{im}} - \alpha_2 (C_2 - C_{\text{TCE}}) \tag{10}
\]

For immobile zone,

\[
\theta \frac{\partial C_{\text{TCE}}}{\partial t} + \rho d \frac{\partial S_2}{\partial t} = -V_{\text{max}22} \frac{C_{\text{TCE}}}{K_p + C_{\text{TCE}}} x_{\text{im}} + V_{\text{max}12} \frac{C_{\text{PCE}}}{K_p + C_{\text{PCE}}} x_{\text{im}} + \alpha_2 (C_1 - C_{\text{TCE}}) \tag{11}
\]

In which \( V_{\text{max}21}, V_{\text{max}22} \) is maximum degradation rate of TCE, \( C_2, C_{\text{TCE}} \) is concentration of TCE in mobile, immobile zone.

4.4 Transport of DCE with biodegradation

For mobile zone,

\[
\theta \frac{\partial C_{\text{DCE}}}{\partial t} + \frac{\partial}{\partial z} \left( \theta_n V_{\text{m}} C_{\text{DCE}} \right) = \left( \theta_n D_n \frac{\partial C_{\text{DCE}}}{\partial z} \right) + V_{\text{max}21} \frac{C_2}{K_2 + C_2} x_{\text{m}} - \alpha_4 (C_3 - C_{\text{DCE}}) \tag{12}
\]

\[
\theta \frac{\partial C_{\text{DCE}}}{\partial t} + \rho d \frac{\partial S_4}{\partial t} = V_{\text{max}22} \frac{C_{\text{DCE}}}{K_p + C_{\text{DCE}}} x_{\text{im}} + \alpha_4 (C_3 - C_{\text{DCE}}) \tag{13}
\]

Dehalogenation of DCE to EC (ethylene) via VC(vinyl chloride) may take place in this biodegradation process but it was not detected in the present study. Then the modeling biodegradation is limited to PCE dehalogenation to DCE. The model will be improved in the future when DCE-degrading bacteria is isolated and equipment is developed for easily measuring concentration of VC and EC.

4.5 Application to reactor column

A continuously-fed up-flow, anaerobic column reactor was developed using a pure organism. The reactor employed a glass column of 22 cm in length and 4.5 cm in diameter filled with immobilized ceramic beads and MY medium (Fig.6). The reactor was percolated in an up-flow mode (ascending flow) under anaerobic
conditions. PCE solution with 5mg/L was supplied at the bottom of the column. The operation was conducted at 0.3mL/min of flow rate which is resident time within the column is about 12 hours. The measurement of optical density was done at 660nm using a UV spectrophotometer. PCE, TCE and DCE were identified and quantified by static-headspace analysis using a gas chromatograph. The reactor performance was simulated using the preceding model. There were no measures of microbial decrease factor by death. The model adopted 0.185 (1/hour) as $\mu_{d1}$ and $\mu_{d2}$. Simulation was described in Figure 7. PCE degradation to DCE via TCE was well computed. The measured TCE takes a small concentration during the tests series while the simulation gives a little large. The maximum DCE concentration in the simulation occurs at different time from the measures. The model, however, shows a good agreement with the reactor performance.

5 Concluding Remarks

The bacteria transport and tetrachloroethylene (PCE) and trichloroethylene (TCE) dehalogenation have been studied using laboratory column and batch test results. The numerical simulation was done for checking the applicability in bioremediation of brownfields polluted by PCE or TCE. The study concludes that; (1) Transport of bacteria was well described by two-region model while conventional advection-dispersion model did not show satisfactory
Figure 7: Simulation of PCE dechlorination in bioreactor.

Performance.
(2) PCE and TCE degrading characteristics can be accurately quantified by Michaelis-Menten equation.
(3) Bacteria growth rate was not described by Monod type function. It was mathematically expressed by proportion to PCE and by inverse proportion to TCE.
(4) Numerical model showed a good performance of PCE dehalogenation measured in fixed-bed reactor column.

References