1. Introduction
   1. Biodegradable contaminants

The main contaminants which are nowadays considered biodegradable are the following:

1) Petroleum hydrocarbons,

2) Polyaromatic hydrocarbons,

3) Non-chlorinated solvents,

4) Chlorinated solvents.

Chlorinated solvents comprise several widespread groundwater contaminants, one of which will be the subject of this exercise. Particularly, we will investigate the potential for biological remediation of a site contaminated by trichloroethene (TCE).

* 1. In situ biological remediation of contaminated groundwater

In situ biological remediation of contaminated groundwater is a remediation technology which relies on biological processes for the transformation of contaminants into less toxic or non-toxic substances. Biological treatment of contaminated groundwater can occur spontaneously and naturally (natural attenuation) or it may necessitate human intervention (enhanced biological remediation). These alternative in situ remediation schemes are briefly described in the following sections.

* + 1. Natural attenuation

During natural attenuation, achievement of remedial objectives relies on naturally occurring processes and human intervention is not a requisite. Detoxification of the contaminant will result from the growth of indigenous microbial populations, which thrive under the prevailing biochemical conditions (e.g. concentration of nutrients and contaminants, pH, temperature etc.) and metabolize target contaminants. Practically, the contaminants function as substrates for the growth of the microbial populations.

* + 1. Enhanced biological remediation

If the biochemical conditions do not favor the growth of the appropriate microbial populations, natural attenuation may result in slow or even incomplete removal of the target contaminant. Hence, human intervention is a required for the detoxification of the contaminant. To this end, several techniques have been developed for the injection of nutrients or even microorganisms in order to optimize the enhancement of biological remediation.

1. Mathematical description of the problem

The consumption rate, *rs* (MsT-1), of a contaminant by a microbial population is usually described by Monod kinetics, which is an ordinary differential equation described as follows:

 (1)

where *S* (MsL-3) is the contaminant concentration, *Xa* (MxL-3) is the concentration of the microbial population, *μmax* (T-1) is the maximum specific growth rate, *Κs* (MsL-3) is the half-velocity coefficient which depends on the type of the substrate and the microorganism, and *Y* (MxMs-1) is the growth yield.

According to the Monod equation, the consumption rate is affected by (a) the concentrations of the contaminant and the microbial population, and (b) by the growth parameters of the microorganism, that is to say the parameters *μmax*, *Ks* και *Υ*. Monod kinetic equations are quantitative measures for assessing contaminant removal attributed to microbial processes. Therefore, a possible application of Monod kinetics is related to the biological remediation technologies of contaminated groundwater.

1. Numerical application

Presumably a quantity of TCE has been released in the subsurface of a given site. During the characterization phase of this contaminated site, we estimated that the initial concentration of TCE in the groundwater was equal to *So*=250 mg/l. We aim to clean-up the site and as a remedial objective we consider the maximum contamination level (MCL) for drinking water, *SMCL* = 5x10-3 mg/l.

For the biological removal of TCE, the presence of specific bacteria is required, i.e. bacteria belonging to the genus *Dehalococcoides*. *Dehalococcoides* use chlorinated ethenes as electron acceptors and molecular hydrogen (Η2) as electron donor in a step-wise redox reaction, which produces the requisite energy for their growth. If *Dehalococcoides* are present in sufficient numbers and specific biochemical conditions prevail (anaerobic conditions, sufficient Η2 concentrations, a specific range of pH etc.), then TCE can be completely detoxified and ultimately result in the production of the innocuous ethene.

At the contaminated site of our interest, we detected the presence of *Dehalococcoides*, with a concentration equal to *Xa*=6x106 cells/l. Then, we searched in the literature for the growth parameters of *Dehalococcoides* utilized in Monod kinetics. According to Cupples et al. (2004) the maximum specific growth rate of *Dehalococcoides* is equal to *μmax*=0.40d-1. In addition, Holmes et al. (2006) estimated a growth yield equal to *Υ*=1.07x108 cells/mg TCE. Finally, we selected a half-velocity coefficient equal to *Ks*=0.70 mg/l, which lies within the range of values reported by Cupples et al. (2004) for TCE biodegradation.

There are two options to consider regarding the remediation strategy of the contaminated site: (a) to rely on the natural attenuation of TCE, and (b) to inject molecular Η2 in the groundwater and, hence, enhance the consumption rate of TCE by creating a stronger community of *Dehalococcoides*. The first question to consider regards the timeframe for natural attenuation:

1. How much time, *Τna,* will it take for the indigenous *Dehalococcoides* population to reduce TCE concentration to the maximum contamination level, *SMCL,* without our intervention?

Monod equation can be analytically solved with the help of the Symbolic Math Toolbox of MATLAB® (the complete code is included in the M-file ‘*MonodAnalytical.m*’):

 (2)\*

where *W* is the Lambert function, *W(x)*. Eq. (2) can be used to calculate the time, *Τna,* required for the reduction of the initial TCE concentration, *So*, to the MCL concentration, *SMCL* (for the complete code refer to the M-file *‘TimeNaturalAttenuation.m’*). With the existing microbial community, approximately *Τna*=31.5 years are needed for the accomplishment of the remedial goal. This is not considered as a reasonable timeframe for remediation and, hence, we decide to intervene by injecting Η2, and, consequently, by favoring the growth of *Dehalococcoides*. A second question of applied interest derives:

1. Which is the minimum required Dehalococcoides concentration, *Xan*, in order to accomplish the remedial goal (i.e. TCE concentration equal to *SMCL)* in two years?

Again, from Eq. (2), we can estimate that a concentration of *Xan*=9.4x107 cells/l is needed for the remediation of the aquifer within two years (the complete command sequence for this question is included in the M-file ‘*EnhancedBioremediation.m*’).

**Note**

\* This is a simplified representation of the equation resulting from the implementation of the Symbolic Math Toolbox of MATLAB®.

**References**

Cupples AM, Spormann AM and McCarty PL (2004) Vinyl chloride and cis-dichloroethene dechlorination kinetics and microorganism growth under substrate limiting conditions. Environ Sci Technol 38:1102-1107

Holmes VF, He J, Lee PKH and Alvarez-Cohen L (2006) Discrimination of Multiple *Dehalococcoides* Strains in a Trichloroethene Enrichment by Quantification of Their Reductive Dehalogenase Genes, App Environ Microbiol 72(9): 5877-5883.