Single-Well Natural Gradient Drift and Push-Pull Tests for Assessing In-Situ Aerobic Cometabolism of Trichlororethylene (TCE)

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ABSTRACT

This study focused on the evaluation of in situ aerobic cometabolism of trichloroethylene (TCE) using single-well natural gradient drift tests (SWNGDTs) and single-well push-pull tests (SWPPTs). A series of SWNGDTs and SWPPTs were conducted using a monitoring well at an industrial area where the aquifer is contaminated mainly with TCE (~ 1 mg/L). To evaluate transport characteristics of dissolved solutes [bromide (a tracer), toluene (a growth substrate), ethylene (a non-toxic surrogate substrate to probe for TCE transformation activity), and dissolved oxygen (DO, an electron acceptor)], one single-well push-pull transport test was performed by injecting ~280 L of groundwater containing the solutes into the aquifer (push), providing a rest period of 1.0 hour (resting), and then extracting the test solution/groundwater mixtures (pull). Mass balance showed approximately 100% of the injected bromide was recovered, and the recoveries of other solutes were comparable with bromide, suggesting that bromide be used as a conservative tracer for biological activity tests, and that little loss of the substrates occurred prior to biostimulation of the aquifer.

One biostimulation SWNGDT was performed by injecting 4830 L-groundwater containing toluene (7.8±0.8 mg/L), DO (37±2.9 mg/L) and bromide (85±4.3 mg/L) into

the aquifer. Temporal groundwater samples were obtained from the injection well under natural gradient "drift" conditions. The rates of toluene and DO disappearance were greater than that of bromide dilution due to natural groundwater drift, and the production of CO₂ was highly correlated with the disappearance of toluene and DO. These results demonstrated the method of injecting and monitoring under natural gradient drift conditions to assess biostimulation of toluene-oxidizing microorganisms. One surrogate SWNGDT was then performed using the same procedures as the biostimulation SWNGDT except that test solution contained 6.1±0.7 mg/L of ethylene. Simultaneous utilization of toluene, DO and ethylene was observed with the production of ethylene oxide, a cometabolic by-product of ethylene transformation by a tolueneoxidizing microorganism expressing toluene-2-monooxygenase. cometabolic TCE transformation was confirmed by the retardation of a breakthrough curve for background TCE, compared with that for bromide.

Four SWPP activity tests were conducted to quantify rates of toluene degradation and transformation of ethylene and to confirm the involvement of toluene-2-monooxygenase in the transformation of both toluene and ethylene. Estimated zero-order rates for toluene degradation and ethylene transformation were 4.6 and 29 µmol/L/hr, respectively. During the blocking test using Hexyne known as an inactivator of toluene-2-monooxygenase, toluene degradation rate was slightly decreased, while the rate of ethylene transformation significantly decreased by a factor of 10 compared to that in the absence of Hexyne. These results suggest that toluene-2-monooxygenase might be responsible for ethylene, and toluene degradation was carried out by not toluene-monooxygenase but also toluene-dioxygenase.

Evidences that toluene and oxygen additions in these field tests stimulated indigenous toluene-utilizers with having ability to aerobically cometabolize TCE are: (1) the observed simultaneous utilization of toluene and DO; (2) the transformation of ethylene to ethylene oxide; and (3) transformation of TCE. Both SWNGDT and SWPPT methods provide a simple, low-cost and effective means for conducting rapid field assessments and pilot testing of aerobic cometabolism, which has previously hindered application of this technology to groundwater remediation.

Key words: TCE, toluene, aerobic cometabolism, single-well natural gradient drift test (SWNGDT), single-well push-pull test (SWPPT), ethylene, ethylene oxide

Experiment	Parameters	Toluene	Ethylene	Bromide
Transport SWPPT	Recovery (%)	89	91	99.9
	Zero-order rate (µmol/L/hr)	1.4	0.49	_
Toluene activity SWPPT	Recovery (%)	10	0	74
	Zero-order rate (µmol/L/hr)	4.6	0	-
Ethylene activity SWPPT	Recovery (%)	0	15	78
	Zero-order rate (µmol/L/hr)	0	29	-
Toluene and ethylene activity SWPPT	Recovery (%)	19	30	71
	Zero-order rate (µmol/L/hr)	3.9	9.5	_
Blocking SWPPT	Recovery (%)	25	45	57
	Zero-order rate (µmol/L/hr)	3.0	3.3	-