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Full-Scale Evaluation of *In Situ* Cometabolic Degradation of Trichloroethylene in Groundwater through Toluene Injection

PERRY L. MCCARTY,* MARK N. GOLTZ,[†]
GARY D. HOPKINS, MARK E. DOLAN,
JASON P. ALLAN,
BRETT T. KAWAKAMI, AND
T. J. CARROTHERS[‡]
Department of Civil and Environmental Engineering, Stanford University, Stanford, California 94305-4020

Groundwater contaminated with 500−1200 μg/L trichloroethylene (TCE) was treated *in situ* over a 410-day period by cometabolic biodegradation through injection of 7−13.4 mg/L toluene, oxygen, and hydrogen peroxide in groundwater circulated between two contaminated aquifers through two treatment wells located 10 m apart. One well pumped contaminated groundwater from the 8 m thick upper aquifer to the 5 m thick lower aquifer, while the other pumped contaminated water from the lower to the upper aquifers using flow rates of 25−38 L/min, effecting groundwater circulation between them. Following 18 days of periodic toluene injection to develop an active biological population, continuous pulses of toluene were added. Over 312 days, an average 87 ± 8% TCE removal was obtained in the upper aquifer with each pass through the treatment well. In the lower aquifer, removals were 83 ± 16% over the last 79 days when peroxide addition was reduced. Treatment reduced TCE in the regional groundwater plumes from about 1000 μg/L in new water entering the 480 m^2 monitored treatment zone to an average of 18−24 μg/L in groundwater leaving the treatment zone, indicating total TCE removal of 97−98%. Pumping heads for groundwater recirculation were less than 6 m. Toluene was removed by 99.98% through biodegradation to an average of 1.1 ± 1.6 μg/L at the 22 m × 22 m boundaries of the study zone, well below the goal of 20 μg/L maximum.

Introduction

The aerobic cometabolic biodegradation of trichloroethylene (TCE) and other chlorinated alkenes in groundwater was demonstrated in pilot scale at the Moffett Federal Air Field using methane, phenol, and toluene as primary substrates (*1*). TCE removal efficiency was much greater with either phenol or toluene than with methane (>85% versus 15%). The next step in the potential implementation of this technology was evaluation at a scale representative of full-scale operation at a TCE-contaminated site. The results of such an implementation at site 19, Edwards Air Force Base, CA, are reported here. The purpose of this study was to obtain sufficient documentation and time of operation to permit evaluation of the advantages and limitations of this process for full-scale aquifer remediation.

Site 19 was selected as it presented near-ideal conditions for this evaluation. It has a TCE-contaminated groundwater plume containing on the order of 500−1500 μg/L TCE; a groundwater table that is not too deep, thus permitting the more economical installation of monitoring wells and the two treatment wells desired; a relative freedom from excessive geological heterogeneities that could otherwise make results ambiguous; sufficient hydraulic conductivity so that groundwater could be pumped at a significant flow rate; site security to protect the overall treatment and monitoring system when no one was in attendance; and support for the demonstration by the owner of the site and local, regional, and federal regulatory agencies.

Edwards AFB is located on the western portion of the Mojave Desert, about 60 mi north of Los Angeles. Site 19 is an area of about 53 acres on the west side of Rogers Dry Lake. From 1958 through 1967, engines for the X-15 rocket plane were maintained in facilities at the site. Approximately one 55-gal drum of TCE was used each month to clean the engines. Disposal of the TCE into the nearby desert created a large groundwater contaminant plume. As shown in Figure 1, the area of the plume selected for the evaluation lies about 400 m east of the contamination source. On the basis of investigatory work previously completed, site 19 appeared to possess many of the desirable characteristics sought. The groundwater surface lies about 9 m below ground surface. Hydraulic conductivities ranged from 1.5 to 5.5 × 10^{-3} cm/s, with an average of 3.4 × 10^{-3} cm/s. TCE concentrations were in the desired range. No 1,1-DCE, which could potentially harm aerobic cometabolism (*1*), nor indeed any other contaminants of significance were present as co-contaminants (*2*). Edwards AFB remedial project managers and federal, state, and local officials charged with overseeing the remediation activities at site 19 were all supportive of the proposed evaluation. Such support came largely because of the extensive laboratory and pilot-scale field studies conducted over the past decade on the proposed process with peer review of the results and also because of the extensive near real-time monitoring system proposed and the presence of a contingency plan for toluene removal from groundwater if this became necessary.

Site Characterization

Physical. The site selected for the evaluation contained two aquifers (*2*). The upper aquifer is unconfined and about 8 m thick. It is separated from the lower aquifer by a 2 m thick aquitard. The lower aquifer, which is confined, is about 5 m thick and lies above weathered bedrock. The aquifer sediments are of alluvial origin interfingered with varying amounts of lacustrine deposits. There is approximately a 0.23 m head difference in the downward direction between the upper and lower aquifers. A natural hydraulic gradient exists toward the east southeast of 0.007 at site 19 (*2*).

Short-term slug and pump tests indicated that hydraulic conductivities at the site were comparable to those obtained in other areas of site 19 (i.e., 1−10 × 10^{-3} cm/s). Using a hydraulic conductivity of 3.4 × 10^{-3} cm/s and assuming a porosity of 0.30 for the sandy aquifer material, groundwater velocity is estimated at 6.9 cm/d to the east southeast. This is in reasonable agreement with a groundwater velocity of 7.7 cm/d estimated from the 700 m the TCE plume has

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* Corresponding author fax: 650-725-9474; e-mail: mccarty@ce.stanford.edu.

[†] Present address: Air Force Institute of Technology, Wright-Patterson Air Force Base, OH 45433-7765.

[‡] Present address: Harvard University, Cambridge, MA.

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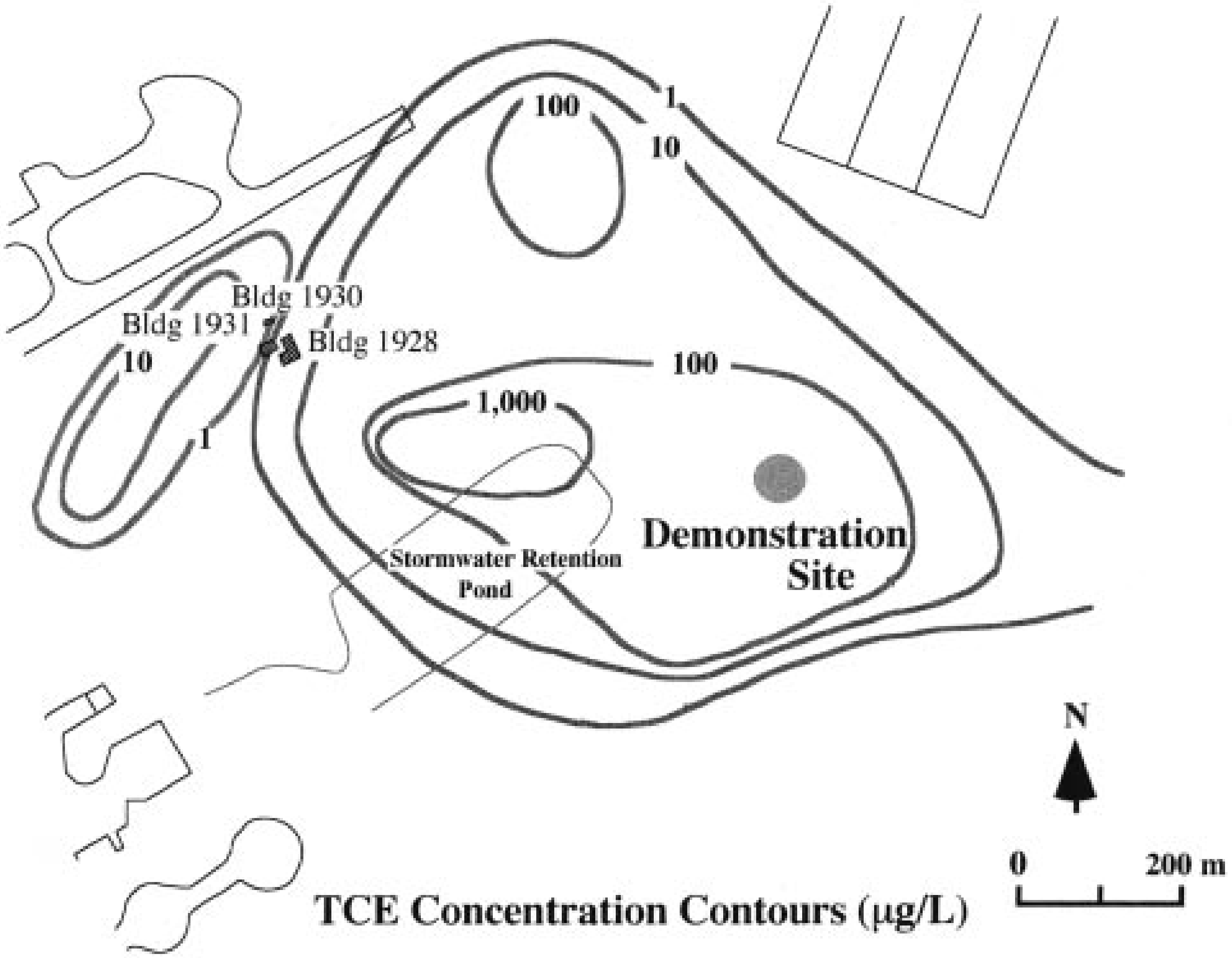

FIGURE 1. Edwards site 19 plan view showing TCE isoconcentration contours.

apparently traveled since its origin nearly 40 years ago and assuming a retardation factor of 1.6 for TCE movement (see below).

The aquifer material consists of fine to medium size sand with some silt. Sieve analysis of aquifer material from borings at the evaluation site at four depths indicates that the lower aquifer material is somewhat larger [Suter mean diameter (d_{sm}) = 0.4 mm] and better graded [coefficient of uniformity (C_u) = 7, coefficient of gradation (C_g) = 1] than upper aquifer material (d_{sm} = 0.2 mm, C_u = 4, C_g = 1). The fraction of organic carbon (f_{oc}) for both the upper and lower aquifer material was found to be relatively low (f_{oc} = 0.0001−0.0004).

Chemical. TCE desorption isotherms were determined for site aquifer material following the method of Farrell and Reinhard (*3, 4*). These studies indicated that 98% of the sorbed TCE mass would desorb in less than 1 min. A linear distribution coefficient (K_d) for TCE of 0.1 mL/g was found for material from both aquifers. Assuming aquifer material with a porosity of 0.30 and bulk density of 1.75 g/mL, this corresponds to a retardation factor of 1.6 (*5*).

Groundwater quality at the evaluation site is summarized in Table 1. Also, TCE concentrations in both aquifers were about the same, between 500 and 1200 µg/L, with an average concentration of 680 and 750 µg/L in the upper and lower aquifers, respectively.

Microbiological. A laboratory semicontinuous slurry microcosm method, which mimicked cometabolic biodegradation field results at Moffett Federal Airfield (*6*), was applied to aquifer material from Edwards AFB (*7*). Phenol and toluene were added to separate microcosms in concentrations of 13.4 and 9.7 mg/L, respectively, along with 0.66 mg/L TCE and 32 mg/L dissolved oxygen (DO). Between 87 and 99% TCE removal was obtained as a function of sample depth for both phenol- and toluene-fed microcosms (*7*). The dominant effective enzyme system was toluene ortho-monooxygenase, as it had been at the Moffett site. Rate coefficients of importance for modeling were evaluated. For toluene, the primary substrate selected for full-scale evaluation, the estimated growth yield (*Y*) was 0.77 g of cells/g of

TABLE 1. Groundwater Chemistry at the Treatment Evaluation Site[a]

parameter	value
total organic carbon (mg/L)	6.7
boron (mg/L)	3.4
calcium (mg/L)	180
chemical oxygen demand (mg/L)	60
chloride[b] (mg/L)	720
iron (mg/L)	<0.1
magnesium (mg/L)	60
manganese (mg/L)	0.02
nitrate[b] (mg/L)	26
dissolved oxygen (mg/L)	<0.5
potassium (mg/L)	1.7
sodium (mg/L)	560
total dissolved solids (mg/L)	2500
sulfate[b] (mg/L)	710
total phosphates (as P) (mg/L)	<0.05
pH	7.36
total alkalinity (as $CaCO_3$) (mg/L)	340

[a] Measurements, except as indicated, from PACE Environmental Laboratories (Novato, CA), report dated November 30, 1994. [b] Measurements from on-site ion chromatography analysis using automated sampling and analysis platform.

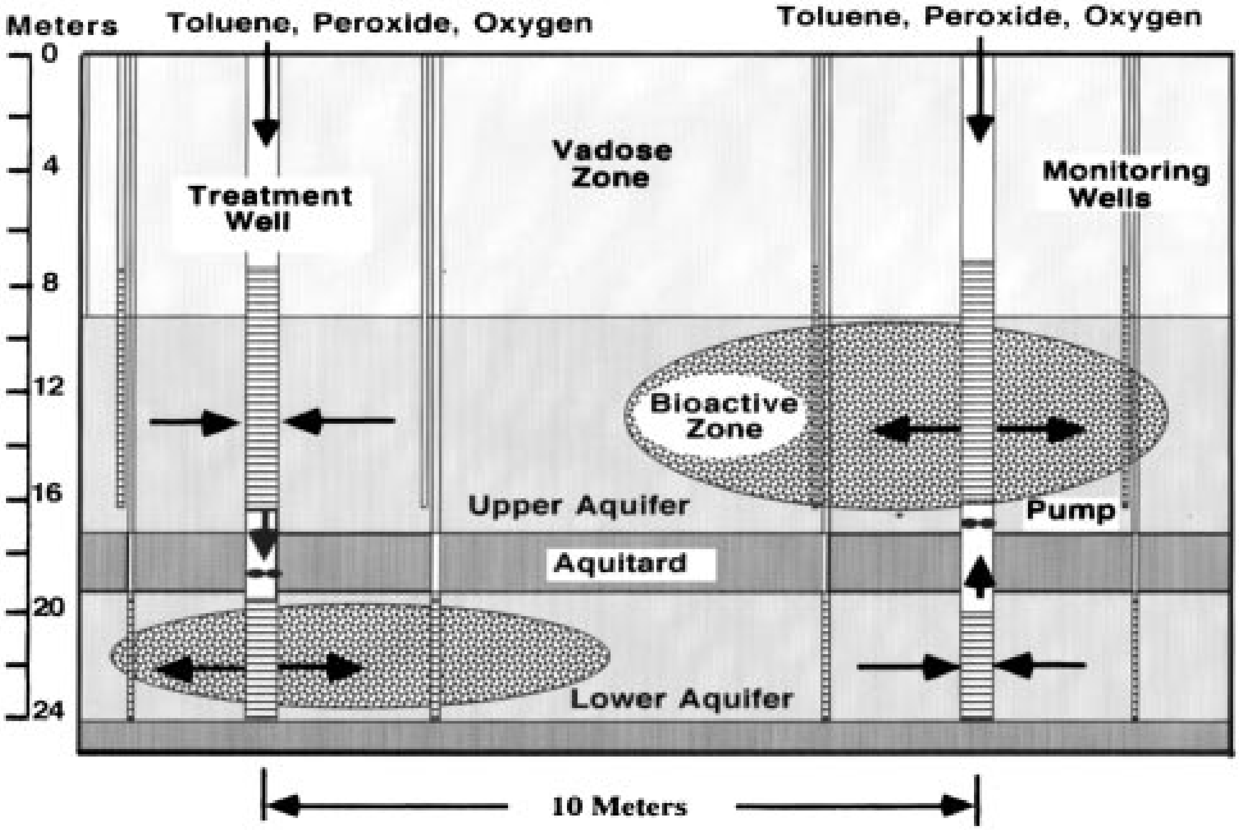

FIGURE 2. Cross-section of two-well cometabolic TCE biodegradation treatment system spanning two separate aquifers.

toluene, the organism decay coefficient (b) was 0.15/d, the maximum primary substrate utilization rate (k_1) was 1.5 g of toluene d^{-1} (g of cells)$^{-1}$, and the second-order rate coefficient for TCE biodegradation (k_2/K_{S2}) was 0.07 L mg^{-1} d^{-1}. These values were used in the modeling studies reported below.

System Design and Construction

The subsurface recirculation system, consisting of two treatment wells (Figure 2), was constructed after the circulation wells described by Herrling et al. (*8*) and McCarty and Semprini (*9*). Each treatment well was screened at two depths. On the basis of the results of the aquifer testing and model studies, a flow rate of 38 L/min (10 gal/min) at each treatment well was initially selected because this should be obtainable without excessive drawdown in the upper aquifer or pressure change in the lower aquifer (no more than 5 m total hydraulic head change). A submersible pump that could deliver this flow rate was installed between the two screens of each well to draw TCE-contaminated water into the well at one of the screened intervals. The primary substrate and oxygen source are introduced into the well through feed lines and mixed into the TCE-contaminated water using two 2.54 cm diameter Model 100-812 static mixers placed in series in the treatment well (TAH Industries, Inc., Robinsville, NJ). The groundwater, containing a mixture of TCE, primary substrate, and oxygen, is discharged into the aquifer from the second screened interval. An *in situ* bioactive treatment zone is thus created in the aquifer around the discharge screen of each treatment well. Treatment well 1 (T1) withdraws groundwater from the upper aquifer and discharges it into the lower aquifer, while treatment well 2 (T2) does the reverse. Thus, water is caused to circulate between the two aquifers. Water is never brought to the surface except for sampling, with the attendant savings in pumping cost and treatment and disposal requirements.

The choice of primary substrate to use depended upon a number of factors. The prior studies (*1, 7*) showed that efficient TCE cometabolism could be obtained with either phenol or toluene. Both phenol and toluene are relatively inexpensive chemicals. In addition to cost, toluene was selected for several reasons. Toluene, a component of gasoline and a naturally produced organic chemical, is a common groundwater contaminant and already is present as a contaminant at Edwards AFB. Based on toxicological data, a maximum contaminant level (MCL) for toluene in drinking water of 14.3 mg/L was recommended (*10*). The U.S. EPA, taking a more conservative approach, has promulgated both a health-based MCL and a maximum contaminant level goal (MCLG) for toluene of 1 mg/L (*11*). Phenol, which is rarely found in water at levels where health effects would be anticipated, has had no MCLG established, although toxicological data suggest that to avoid health risks drinking water concentrations should not exceed 3.5 mg/L (*10*). Considering aesthetics, toluene has an odor threshold of 24 μg/L and a taste threshold of 120−160 μg/L (*12*). Phenol has odor and taste thresholds of 1000 and 100 μg/L, respectively (*13*). However, phenol is a particular problem since it reacts during chlorination to form chlorophenols that have extremely low taste and odor thresholds below 1 μg/L. In addition, pentachlorophenol and 2,4,6-trichlorophenol are probable human carcinogens (*14*). Because of this, the World Health Organization recommends that the total phenol content of water to be chlorinated be below 1 μg/L, and the European Drinking Water Standard for phenol is set at 0.5 μg/L (*13, 15*). Thus, although phenol and toluene are similar from a health effects perspective, the fact that toluene has an established MCLG, does not create taste and odor problems at concentrations below 24 μg/L, and is not a precursor to probable carcinogens when chlorinated led to its selection as the primary substrate for use in this evaluation. Another factor of importance is that toluene can

TABLE 2. Parameter Values Used in the Modeling Study

parameter	value
distance between treatment wells (m)	10
flow rate through each treatment well (L/min)	38 (10.0 gal/min)
regional flow rate (cm/d)	6.9
treatment well screen length (m)	5
concn of oxygen source (hydrogen peroxide) added (mg/L)	100 (as O_2)
time-averaged concn of primary substrate (toluene) added (mg/L)	9
toluene pulse length	30 min once every 8 h
initial dissolved TCE concn in aquifer (mg/L)	1.0
TCE sorption distribution coeff (mL/g)	0.10
TCE sorption/desorption rate coeff	0.10/day
TCE retardation factor	1.6
cometabolism rate constant (k_2)	0.7 mg of TCE (mg of cells)$^{-1}$ day^{-1}
TCE half-saturation coeff (K_{S2})	10 mg of TCE/L
organism yield coeff (Y)	0.77 g of cells/g of toluene
organism decay coeff (b)	0.15/day
max toluene utilization rate (k_1)	1.5 mg of toluene (mg of cells)$^{-1}$ day^{-1}
toluene half-saturation coeff (K_{S1})	1.0 mg of toluene/L

be air stripped from groundwater while phenol cannot. This property could be important if it were found after adding the primary substrate to the groundwater that its degradation was inadequate and had to be removed. Additionally, toluene is a liquid that can be pump-fed neat to the treatment system. Both field and laboratory studies indicated that, as long as adequate dissolved oxygen was present, toluene concentrations near 1 μg/L could be obtained from toluene biodegradation in the treatment system (*1*). This concentration is more than an order of magnitude below the taste and odor threshold and several orders of magnitude below the drinking water MCLG.

A strategy of pulsing in pure toluene for 30 min every 8 h was used in the Moffett studies. This strategy helped distribute the toluene more uniformly through the aquifer, thereby reducing microbial clogging potential at the well screens. It also helped reduce the effects of competitive inhibition, where TCE degradation rate is reduced when primary substrate and TCE are simultaneously present. The design for the Edwards evaluation incorporated the toluene concentration and pulsing strategies used at Moffett, with allowance for changes to compensate for site-specific conditions.

On the basis of the Moffett results (*1*) and microcosm studies (*7*), a toluene concentration of 7−15 mg/L was proposed to achieve on the order of 90% TCE destruction with each pass through a circulation well. Theoretically, 9 mol of oxygen is required for complete oxidation of 1 mole of toluene, with actual oxygen requirements being about 6 mol/mol as a portion of the toluene is synthesized into bacterial cells. This translates into a minimum requirement of about 2.1 g of oxygen/g of toluene or up to 31 mg/L dissolved oxygen for 15 mg/L toluene. Some additional oxygen is needed to ensure that aerobic conditions are maintained and to satisfy potential background oxygen-demanding materials in the aquifer. Thus, 30−40 mg/L of dissolved oxygen would be required to support the planned aerobic cometabolism (*1*), and this was substantiated through the microcosm studies (*7*). Either oxygen gas or hydrogen peroxide had been used as an oxygen source at Moffett. Initially at Edwards, it was decided to add only oxygen gas because of the potential bactericidal properties of hydrogen peroxide. Once an active population was developed, hydrogen peroxide might then be used as an additional source of oxygen, which could also later help to suppress potential microbial clogging near the well screens due to these same bactericidal properties (*1*). Additionally, due to hydrogen peroxide's solubility in water, higher dissolved concentrations could be achieved than through the use of pure oxygen alone.

In order to determine appropriate spacings for the treatment and monitoring wells, preliminary numerical modeling studies were conducted. The following criteria were used to help design the site:

(1) The direct path flow time from injection to extraction well should be about 3−5 days.

(2) After several months of operation, at least 80% of the water treated in one well should have arrived at the second treatment well, so results could be demonstrated within a reasonable time frame.

(3) During several months of operation, TCE concentrations should be demonstrably reduced at the site.

These criteria were selected due to financial constraints, which limited the study time, and the desire to obtain adequate information for a good evaluation of treatment effectiveness.

A two-dimensional code, RESSQ, was used to simulate steady-state pumping-induced groundwater flow (*16, 17*). This code helped establish the flow patterns for the system in the form of stream tubes. These stream tubes were then used in conjunction with a one-dimensional transport code that incorporated the effects of pertinent chemical and biological fate and transport processes along each stream tube. Together, these codes helped to obtain a clear picture of how bioremediation might occur at the site and helped to determine the optimal design to achieve the above criteria.

Table 2 lists the modeling parameters used. The biological parameters match those obtained from the microbiological studies. A much slower desorption rate was chosen for modeling than obtained in the chemical study, because at the larger field scale such factors as diffusion from low permeability lenses are likely to come into play. The value chosen (0.1/d) was similar to that found appropriate from results at Moffett Field (*18*). The toluene and oxygen delivery parameters and toluene pulsing strategy have already been discussed.

On the basis of the modeling studies, a 10 m spacing between the wells appeared appropriate in conjunction with the previously selected pumping rate of 38 L/min to meet the selected design criteria. Figure 3 illustrates flow model results for the upper aquifer that are similar to that for the lower aquifer (not shown). The flow was divided into 20 stream tubes, each of which represents a flow rate equal to 5% of the pumped flow or 1.9 L/min. Thus, 20 stream tubes enter or leave each treatment well. For the upper aquifer illustrated in Figure 3, five stream tubes enter the downflow well (T1) from the incoming captured regional flow. This captures an 80 m wide portion of the contaminated plume. The other 15 stream tubes entering T1 come from flow

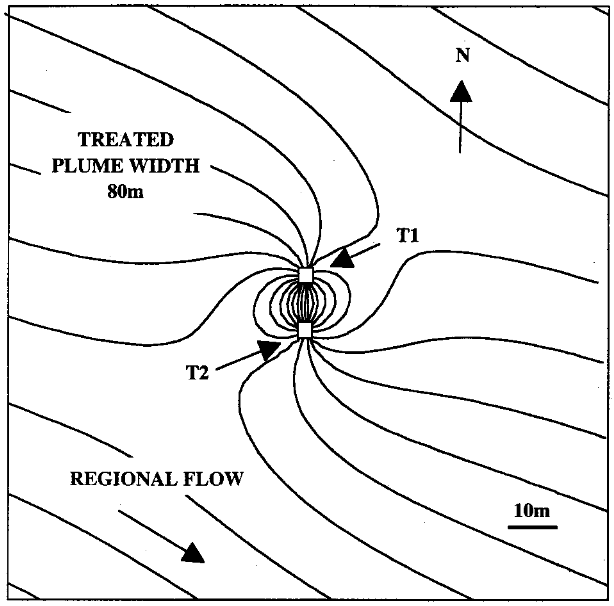

FIGURE 3. Model simulations of upper aquifer stream tubes entering the treatment zone, recirculating between the two treatment wells, and then leaving the treatment zone with balanced pumping rates of 38 L/min.

recirculated back from the upward flow at T2. In the lower aquifer, which is thinner, only 2.5 stream tubes of flow entering T2 come from the regional flow and 17.5 come from flow recirculated from T1. The width of plume captured here is 66 m. Since each stream tube represents equal flow, the wider stream tubes indicate slower fluid velocities. Travel times were calculated for the path directly between the wells and for the outermost recirculated stream tube illustrated in Figure 3. The modeled 5-day flow time for the direct path between the wells satisfies the selected criteria. A travel time of 90 days for the outermost recycled stream tube means that at least 100% of the flow treated at T2 will reach T1 within the originally proposed 120-day timeframe for the evaluation.

The model code used to estimate remediation efficacy was the one-dimensional transport code developed by Semprini and McCarty (*18*) and modified by Lang (*17*). The code includes the microbial processes of bacterial growth, toluene and oxygen utilization, and cometabolic transformation of TCE coupled with transport processes of advection, dispersion, and rate-limited sorption onto aquifer solids. Figure 4 depicts model output for TCE concentration at selected locations over time. The upper horizontal line in the figure represents the constant TCE concentration in the influent or incoming regional flow of 1 mg/L. The TCE concentrations at T1 and T2 represent that of the combined regional flow and recirculated flows entering and mixing within each treatment well. The TCE concentration here decreases with time and approaches steady-state values of about 200 μg/L. The concentrations in the effluents or exiting flows from the two aquifers represent the results of treatment of the water exiting from each of the treatment wells. As biomass increases in the aquifer with time, the percentage removal of TCE with each pass through a treatment well increases to a steady-state value of about 83%. After 120 days, the resulting TCE concentrations are about 29 μg/L in the upper aquifer and 47 μg/L in the lower aquifer in water exiting the treatment zones to join the downgradient regional flow. Because of the recirculation and thus repeated treatment of a portion of the aquifer water, the overall TCE removal of regional water entering the two-well treatment system is estimated to be about 95–97%. The model results

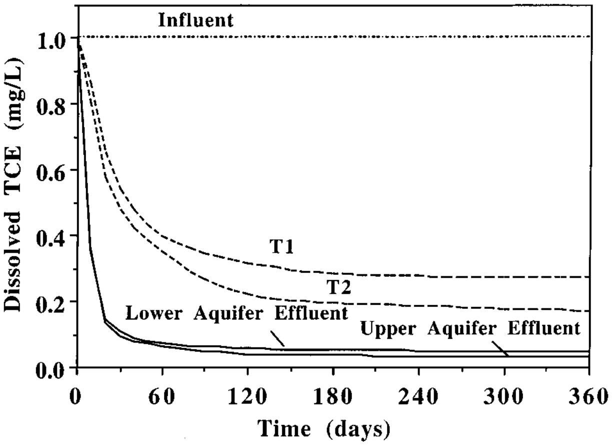

FIGURE 4. Model simulations of TCE concentrations in aquifer water entering the treatment zone, passing through the two treatment wells, and exiting the treatment zone as functions of time of treatment for balanced pumping rates of 38 L/min.

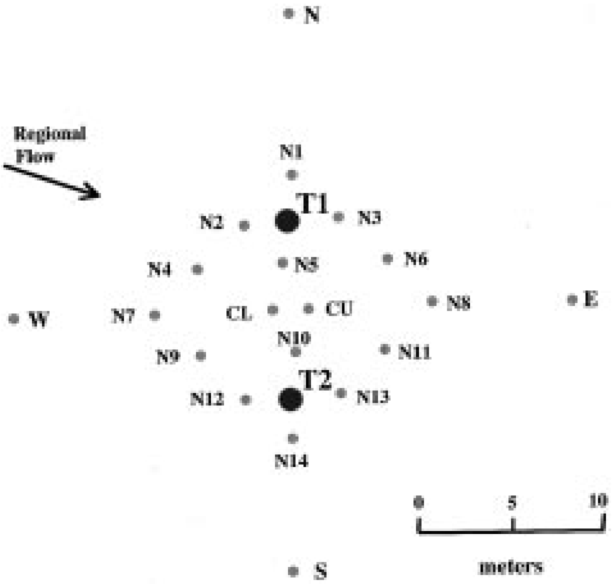

FIGURE 5. Plan view of evaluation site layout including placement of treatment and monitoring wells.

indicate that, with a well spacing of 10 m, significant TCE removal should be observed over the course of the proposed study.

The resulting treatment system (Figure 5) consists of two 20 cm diameter treatment wells (T1 and T2) located 10 m apart and screened in both the upper and lower aquifers. Each of the nested monitoring locations (N1–N14) has two 5 cm diameter monitoring wells, each screened in a different aquifer (Figure 2). At the center of the site, C-U is a 10 cm diameter monitoring well screened in the upper aquifer, while C-L is a 10-cm well screened in the lower aquifer. Surrounding the site are four 10 cm diameter “compass point” wells, located 15 m from the site center, approximately to the north, south, east, and west. The north, south, and west wells each allow groundwater samples to be taken from both the upper and lower aquifers, while the east well, which was the first well constructed at the site, can only be sampled in the upper aquifer. These compass point wells were installed to meet regulatory concerns and to provide further monitoring information for evaluating treatment effectiveness. It was proposed that toluene concentrations not exceed 20 μg/L at these locations, a concentration below the taste and odor threshold. Thus, altogether at the site, there were 41 sampling locations, including the two treatment wells, which can each be sampled in both the upper and lower aquifers.

After installation of the treatment and monitoring wells, groundwater heads at the evaluation site were measured. A hydraulic gradient of 0.004 in the upper aquifer and 0.010 in the lower aquifer, both to the east southeast direction, were measured. These gradients are in general agreement with the overall gradient for site 19 of approximately 0.007 to the east southeast (*2*).

Monitoring System

An automated sampling and analysis platform (ASAP, Analytical and Remedial Technology, Inc., Milpitas, CA) with 28 sample ports was connected directly to submergible Grundfos Rediflo-2 pumps placed in the treatment wells and in selected monitoring wells that were to be monitored most frequently. The remaining 13 monitoring well locations (the compass point wells, N1−N3 upper aquifer wells, and N12−N14 lower aquifer wells) were manually connected to the ASAP system with Grundfos pumps as needed. The ASAP incorporates an interface module used to control the Grundfos pumps for purging the sample wells (approximately 200 L) and providing a representative groundwater sample to the ASAP system. The excess extracted groundwater was filtered and returned to the subsurface through treatment well T1.

The ASAP allowed continuous, near real-time sampling of approximately 30 samples per day using modules to process aqueous sample aliquots for introduction into attached analytical instrumentation (*19*). Analytical results were automatically stored in a computer data base for both local and remote access and analysis including graphic display. The ASAP allowed remote control of sampling activities and provided automated calibrations and QA/QC analysis of known standards (*19*). Purgeable hydrocarbons (chlorinated aliphatic hydrocarbons and aromatic hydrocarbons) were analyzed by gas chromatography (GC), inorganic ions through single-column ion chromatography, and dissolved oxygen and pH with probes.

A multi-port sample loop valve provided samples for purge-and-trap GC analysis with selectable volumes from 0.2 to 10 mL. A 30 m thick film DB-5 mega bore GC column in series with a 15 m thick film DB-624 mega bore column (J&W Scientific, Folsom, CA) provided good resolution for the compounds of interest. The GC (Finnigan-Tremetrics, Model 9000, San Jose, CA) was equipped with tandem photoionization and flame ionization detectors for TCE and toluene analyses. Chromjet integrators (Thermal Separation Products, San Jose, CA) provided integration of the detector signals and communicated with the ASAP computer for data storage and retrieval.

Inorganic ions (bromide) were processed by the ASAP high-performance liquid chromatography (HPLC) module using fixed-loop injection, a standard anion column and conductivity detector (Model 350) (both from Alltech Associates, Deerfield, IL), and a binary gradient HPLC pump (Thermal Separations Products, Model P2000, San Jose, CA). The eluent was 4 mM potassium acid phthalate. The ASAP system also collected data for dissolved oxygen and pH using probes and associated meters (Orion Research, Inc., Models 860 and 520A, respectively, Beverly, MA).

Results

Operational details of the treatment system over the 444 days of total operation are summarized in Table 3 for treatment well T1 and in Table 4 for treatment well T2. Five phases of system evaluation are described: (1) pre-operational studies (days 0−33), (2) the establishment of a toluene-degrading consortium (days 34−55), (3) pre-steady-state operation (days 56−136), (4) steady-state operation (days 142−271), and (5) balanced flow operation (days 317−444). During the first 89 days of the overall study, the two treatment wells were operated similarly, except the net flow rate at T1 was lower than at T2, and the resulting chemical concentrations were higher. Noticeable differences in operation then led to a different mode of operation for each up through the fourth phase. Finally, in the fifth and last phase using lessons learned during the first four phases, chemical additions and flow rates for the two treatment wells were set the same and were maintained constant for the entire period.

TABLE 3. Operational Schedule for T1

days of operation	pumping rate (L/min)	toluene addition		DO added (mg/L)	H_2O_2 added (mg/L)
		time-av (mg/L)	pulses per day		
0−33		preoperational studies			
34−55	0−25	0−11.6	0−24	0−44	0
56−136	25	5.8−11.6	1−12	44	0−71
142−204	25	0−11.6	1	44	17−117
209−271	25	0−11.6	0.67−1	44	35−71
317−444	25	9.0	0.67	44	47

TABLE 4. Operational Schedule for T2

days of operation	pumping rate (L/min)	toluene addition		DO added (mg/L)	H_2O_2 added (mg/L)
		time-av (mg/L)	pulses per day		
0−33		preoperational studies			
34−55	0−38	0−7.6	0−24	0−29	0
56−136	38	3.8−13.4	1 −12	29	0−47
142−204	38	13.4	1	29	47−63
209−271	38	13.4	1	29	47
317−444	25	9.0	0.67	44	47

Pre-Operational Studies. After construction of the treatment system, 24-h pump tests were conducted at each treatment well using a perceived flow rate of 38 L/min. Under homogeneous conditions and equal flow rates, the lower (confined) aquifer drawdown response to the upflow test was anticipated to be identical to the mounding response due to the downflow test. However, the mounding at T1 during the downflow test (0.84 m) was about 70% of the drawdown at T2 during the upflow test (1.15 m). While this may be due to site hydrogeology, another possible explanation is that the net flow rate at T1 was less than that at T2 by about 30%. This may be due to suspected damage during initial testing to the bentonite slurry seal, which separates the lower and upper aquifers at T1 because of inadequate well development. This could have allowed a portion of the water injected into the lower aquifer to circulate directly back to the influent screen in the upper aquifer.

An additional verification of lower net flow at T1 comes from head responses at various monitoring wells during the T1 and T2 24-h pump tests. The head data were analyzed quantitatively using the following expression which, for two monitoring wells in a homogeneous confined aquifer, relates observed drawdown (or mounding) at the two wells (s_1 and s_2), the distance from each monitoring well to the extraction (or injection) well (r_1 and r_2), the aquifer transmissivity (T), and the pumping rate (Q) of the extraction (or injection) well (*21*):

$$s_1 - s_2 = \frac{2.3Q}{2\pi T}\log\frac{r_2}{r_1} \qquad (1)$$

Assuming that T is equal for each of the pump tests, a ratio of pumping rates for the upflow and downflow tests can be

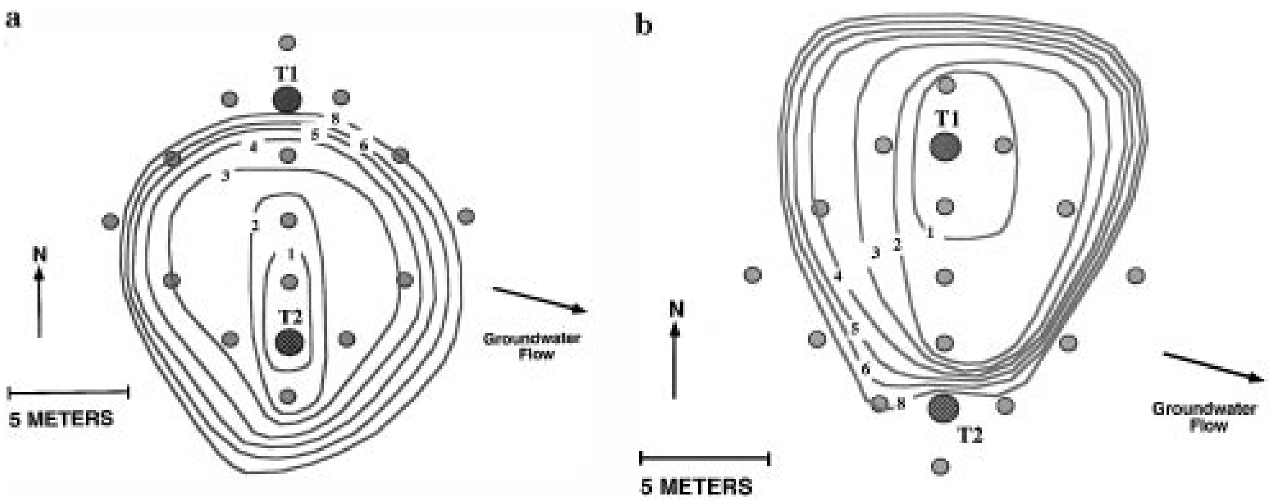

FIGURE 6. Methane 50% arrival time contours (in days) for (a) upper aquifer and (b) lower aquifer.

derived by comparing the responses of monitoring well pairs. Applying eq 1 to all pairs of the 15 lower aquifer monitoring wells (and discarding those pairs where $s_1 \approx s_2$ and/or $r_1 \approx r_2$) and averaging the results, the flow through T1 in the downflow test was calculated to be 62% of the flow through T2 in the upflow test or about 24 L/min.

A further evaluation of the flow rate at T1 was conducted during steady-state operation (phase 4) through a sodium bromide tracer study. Here, 50 g/L bromide was continuously pumped into the T1 well at a flow rate of 34 mL/min for 5 days, and bromide measurements were made at the lower aquifer monitoring wells nearest T1 (N1, N2, N3, and N5). The resulting increased bromide concentration at these wells was 65−68 mg/L. From mass balance on these values, the T1 flow rate is estimated in this manner to be 25−26 L/min, values that are similar to those estimated from the pumping tests.

To further characterize flow at the site as well as to assure the regulatory agencies and ourselves that the toluene that would be injected during the evaluation could be tracked by the monitoring network, an initial tracer test was conducted. As the aquifer was anoxic (Table 1), methane could serve here as a conservative tracer. The tracer test consisted of continuously adding methane to the circulating groundwater at both treatment wells at a concentration of 25−30 mg/L over a 14-day period. Methane was found to arrive at all the monitoring locations. Based on an analysis of 50% breakthrough times for methane at the monitoring wells, methane travel time contours were constructed for the upper and lower aquifers as shown in Figure 6, panels a and b, respectively. These results indicate the aquifers are relatively homogeneous, with groundwater flow from the treatment wells reaching all the monitoring wells. This homogeneity allowed evaluation of system efficiency based upon TCE reduction at the monitoring locations. Additionally, the methane tracer test results provided assurance that the fate and transport of the toluene to be injected into the upper and lower aquifers for the evaluation could be adequately monitored.

Establishment of Toluene-Degrading Consortium. In order to establish a toluene-degrading consortium in the two aquifers, the pumping rates were set, and on day 34 pure oxygen was added for 4 days in order to provide an aerobic environment for the toluene injection. Then, while oxygen addition was continued, sufficient neat toluene was added in pulses, one per hour, to provide time-averaged concentrations of 2.7 and 1.8 mg/L at T1 and T2, respectively. Such toluene injection was continued for two days (days 38−40) and then toluene and oxygen injection was stopped and the pumps were turned off. Samples from various monitoring wells were analyzed for toluene and oxygen decrease, which would provide evidence for the growth of an indigenous toluene-oxidizing population. Toluene reached near zero concentration within 10 days (Figure 7). Pumping and oxygen and toluene injection were re-instituted for a 2-day period, this time with about double the initial toluene concentration. As a toluene-degrading population had now been established, toluene utilization was quicker and reached near zero within 5 days. This procedure was repeated once again, but with another doubling of the toluene concentration. A subsequent rapid depletion of toluene by day 56 indicated that an adequate population had been established to proceed to the next stage.

Pre-Steady-State Operation. In this phase from day 56 to day 136, operation of the treatment system with pumping was continuous as was the introduction of DO. Toluene was added initially at 12 pulses per day, but this gradually decreased to 3 pulses per day by day 62 as a steady-state population became established. This pulsing strategy was designed to reduce organism growth near the treatment well and to reduce the effects of competitive inhibition (*20*). Initially, toluene was added to provide a continuous low time-averaged concentration. Then the concentration was slowly increased with time to a maximum of 11.6 on day 108 and 13.4 mg/L on day 115 at wells T1 and T2, respectively, as field evidence of adequate toluene degradation was obtained together with evidence that toluene did not exceed regulatory levels at the compass point wells. Results from TCE analyses at selected wells in the upper aquifer during this period are illustrated in Figure 8 (days 56−136). TCE concentration decreased as water passed from T2 to N10 and N5, which are located 2.5 and 7.5 m, respectively, from T2 on the path between T2 and T1. TCE removal through treatment is represented by the TCE concentration differences at any given time between T2 and N5. Similar results were found in the lower aquifer. TCE removal generally increased with time in line with the increase in injected toluene concentration and the resulting buildup in a toluene-consuming TCE-degrading population.

By the end of the pre-steady-state period, TCE removal evidenced in this manner was over 80% in both aquifers. However, the overall TCE removal performance differed markedly between the upper and lower aquifers. In the upper aquifer (Figure 8), TCE removal increased gradually with time. Up to about day 105, most of the removal occurred between T2 and N10 after that, most occurred between N10 and N5. This progression of the treatment zone away from T2 was desired and resulted from planned operational modifications. One modification involved increasing the time between

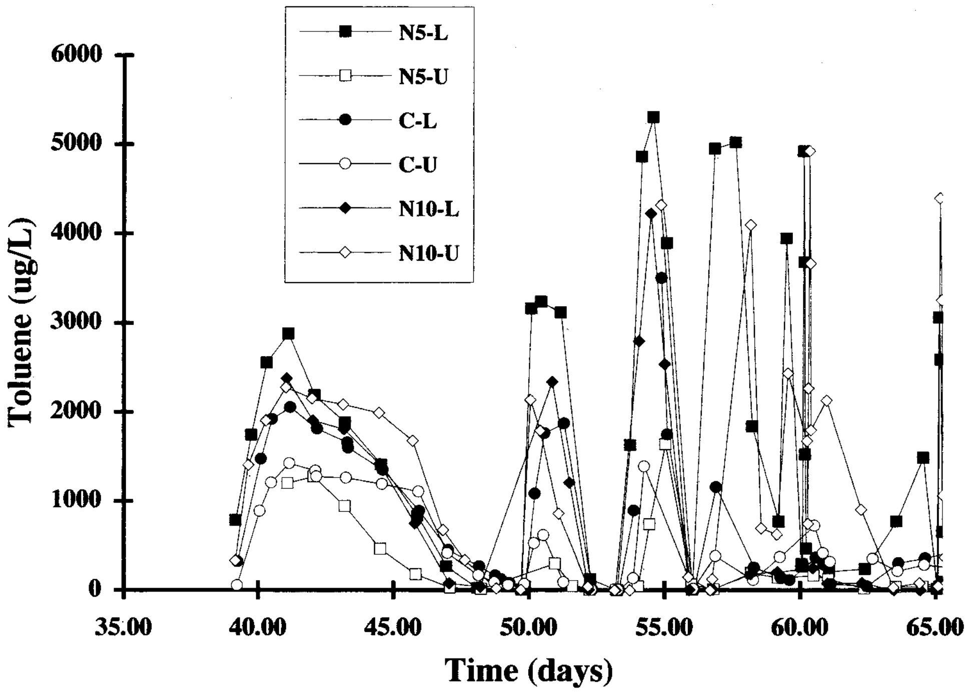

FIGURE 7. Concentrations of toluene at various centerline monitoring locations during the first weeks of toluene addition.

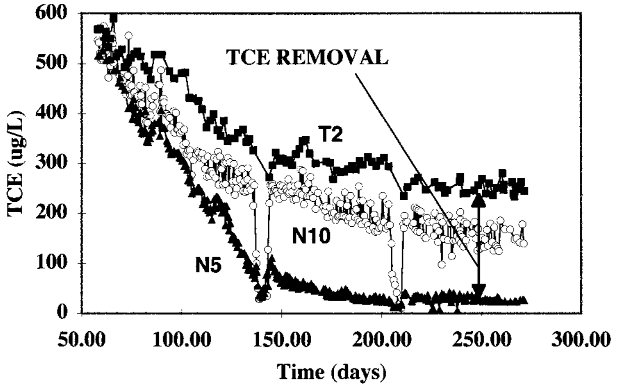

FIGURE 8. TCE concentration changes and removal along the upper aquifer centerline during pre-steady-state and steady-state operation following continuous toluene addition.

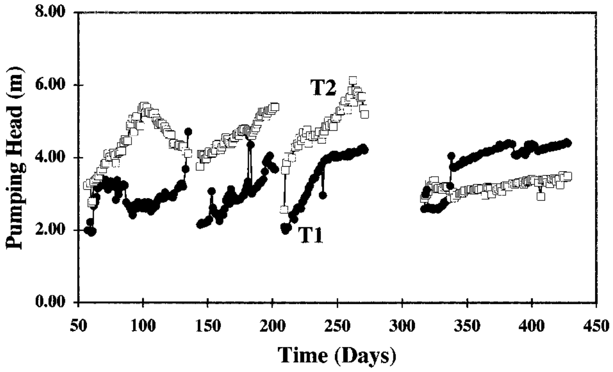

FIGURE 9. Pumping head changes at T1 and T2 throughout the study period following the beginning of toluene addition.

pulses, a strategy designed to move the toluene out further into the aquifer before complete degradation could occur. A second modification was the addition of hydrogen peroxide after day 80 to increase the DO supply as needed for degradation of higher toluene concentrations. This also tended to inhibit bacterial growth near the treatment well. The combination of these two strategies thus was successful.

The above strategies were not as successful for TCE removal in the lower aquifer. Although initially the mass rate of addition of toluene, oxygen, and hydrogen peroxide were the same at the two treatment wells, the concentrations at T1 were higher because the net flow rate was lower. Once this was recognized, the concentration of toluene was reduced. Perhaps hydrogen peroxide addition should also have been reduced because of the excess inhibition that it caused. In spite of this problem, TCE removal up to day 80 was better in the lower aquifer than in the upper aquifer (not shown), perhaps because the actual concentration of toluene then at T1 was 1.33 times that at T2. However, when hydrogen peroxide was added on day 81 (at a concentration 1.33 times higher than at T2 as the difference in net flow rates at the two wells was then not known), inhibition appeared to be very high and TCE removal stopped. On day 89, injected toluene concentration was dropped somewhat, and on day 99 hydrogen peroxide addition was stopped. By day 100, TCE removal began again. TCE removal increased to about 80% by day 120. However, a sudden increase in pumping head at T1 around day 130 suggested that clogging may become a problem (Figure 9). A small amount of hydrogen peroxide was added in an attempt to control this clogging, but this was insufficient, and the well field was shut down on day 136 to redevelop T1 as preventive maintenance. This ended the period of pre-steady-state operation.

Steady-State Operation. Steady-state operation occurred between day 142 and day 271. TCE removal in the upper aquifer during this period is illustrated in Figure 8. TCE concentration at T2 remained relatively constant during this period and near 300 μg/L. The measured concentration at monitoring well N5 averaged about 30 μg/L. About two-thirds of the removal took place between the N10 and N5 monitoring wells or further out in the aquifer as desired. TCE concentration varied more markedly at the near monitoring well (N10) than at the distant monitoring well (N5), a phenomenon observed previously at Moffett Field (*1*). This is a direct result of the once per day pulsing of toluene and resulting competitive inhibition. As the pulse moved through the aquifer, TCE removal momentarily decreased due to competition between toluene and TCE for the oxygenase.

TABLE 5. TCE Removal during Steady-State Periods

steady-state period (days)	Upper Aquifer TCE (μg/L) T2-U n	T2-U av	N5-U n	N5-U av	TCE removal (%)
145−204	30	304 ± 17	96	46 ± 19	85 ± 9
212−271	32	254 ± 12	79	29 ± 7	89 ± 7
365−444	90	171 ± 12	88	24 ± 3	86 ± 9

steady-state period (days)	Lower Aquifer TCE (μg/L) T1-L n	T1-L av	N10-L n	N10-L av	TCE removal (%)
145−204	29	80 ± 26	96	17 ± 8	79 ± 42
215−271	49	63 ± 7	89	26 ± 11	59 ± 22
365−444	80	107 ± 13	84	18 ± 6	83 ± 16

Such TCE oscillations were attenuated by the time the circulating groundwater reached N5 due to the lower toluene concentration near there as well as TCE sorption/desorption effects. The only operational change here occurred from day 204 to day 209 when the system was shut down for routine redevelopment of wells T1 and T2 to reduce the pumping head, which was slowly increasing at both wells due to biomass buildup in the aquifer (Figure 9). Upon restarting, pumping pressures were as at the beginning of the study, and efficient TCE removal resumed almost immediately upon restarting.

The steady-state period TCE removal results for the lower aquifer had much greater variation than in the upper aquifer (not shown). A great deal of this variation was related to the operational changes made in toluene and peroxide additions. A major problem was the unreliability in the hydrogen peroxide feeding system to the lower aquifer, which resulted in excessive feeding of peroxide at times and underfeeding at others. This problem was finally resolved late in the study on day 242. After well redevelopment on days 204−209, periodic addition of toluene was made at T1 to re-establish a toluene-consuming population, and following day 237, a time-averaged toluene addition of 7.8 mg/L was maintained throughout the remaining 34 days of this study period. The problem with lower net flow rate at T1 became recognized late in the study, and the hydrogen peroxide concentration was reduced on day 256 to 35 mg/L. This resulted in dramatic improvement in removal efficiency over the last 15 days of this evaluation.

A summary of TCE removal efficiencies based upon the removals obtained between the two treatment wells and the monitoring wells located 7.5 m away for initial and final steady-state conditions is given in Table 5. The average TCE removal was 87% in the upper aquifer with 13.6 mg/L time-averaged toluene addition, with the highest removal of 89% obtained during the second half of the steady-state period. This is above the 83% removal predicted from site characterization and modeling studies conducted before this evaluation began (Figure 4), a result that may be partly attributed to the fact that the model study assumed a lower injected toluene concentration of 9 mg/L. The average TCE removal in the lower aquifer was much less and averaged only 69% over the steady-state period, a problem related to excessive peroxide addition as noted above. TCE removal efficiencies in the lower aquifer were periodically equivalent to that obtained in the upper aquifer during short periods when excessive peroxide addition did not occur. Excessive peroxide addition is obviously a problem that needs to be avoided.

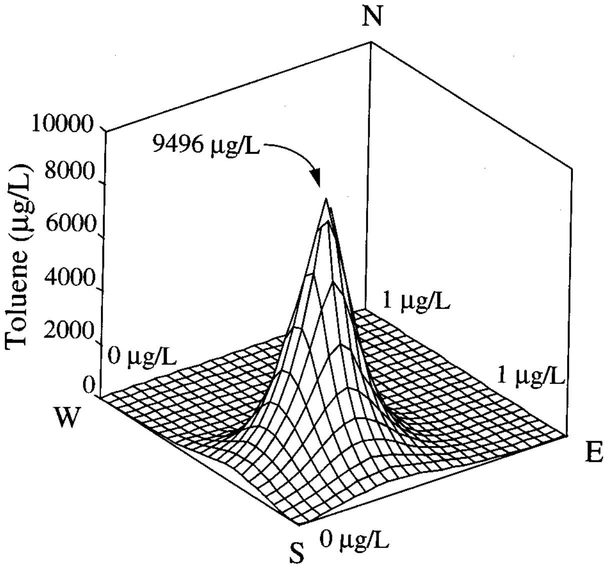

FIGURE 10. Three-dimensional view of toluene concentration in upper aquifer during steady-state operation along with average concentrations measured at four compass-point locations (averages for days 220−260).

Balanced Flow Operation. Following redevelopment of the two treatment wells and adjustment of chemical additions, the treatment system was restarted on day 317 using a balanced flow rate at each well of 25 L/min. The toluene addition was set at 9.0 mg/L, the originally planned concentration, an oxygen addition of 44 mg/L, and a hydrogen peroxide addition of 47 mg/L (the concentration found to work satisfactorily at T2 during steady-state operation). These conditions were maintained throughout the balanced flow operation period, which lasted through day 444. With this change in flow rate at T2, the system went through a period of adjustment before near steady-state conditions were reached. Operational results were averaged between day 365 and day 444, a time period when steady-state operation was being achieved and little significant concentration changes at the monitoring locations were observed.

The results of operation during this steady-state period are summarized in Table 5. The balanced flow conditions resulted in a somewhat decreased TCE concentration in T2 and a somewhat increased concentration at T1, as would be expected. Treatment at both T1 and T2 was satisfactory with no significant operational problems occurring over the course of the study. TCE removal was good in both aquifers, 86% in the upper aquifer and 83% in the lower aquifer. The removal in the upper aquifer was somewhat lower than during the earlier steady-state operation (89%), which can be attributed primarily to the lower toluene addition. The performance in the lower aquifer was much better than previously and just what was predicted from the modeling studies (83%). With the lower hydrogen peroxide addition, the previous operational problems were here avoided. Thus, it appears that with this mode of operation, the system could be operated for long periods of time without significant operational problems.

Toluene Removal. The majority of toluene removal occurred within the first 2.5 m of travel from each of the treatment wells. Figure 10 presents a three-dimensional view of measured toluene concentration in the upper aquifer during the steady-state operation. Results in the lower aquifer were similar. The toluene concentration surface shown was prepared using a 25 × 25 grid to represent the 480 m^2 study area with each of the sample locations lying at one of the grid nodes. Values at other nodes were estimated through Kriging

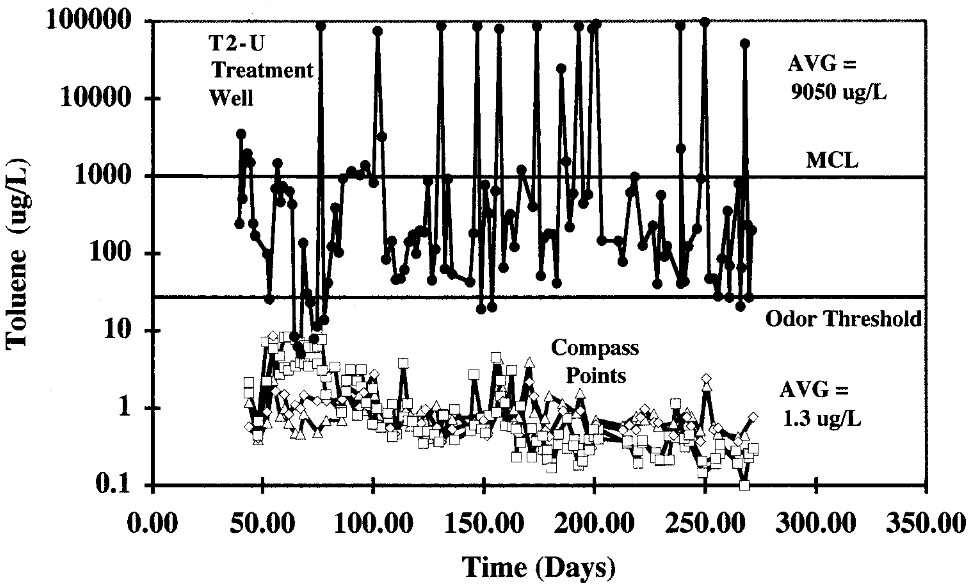

FIGURE 11. Toluene concentration changes for upper aquifer through the steady-state study period measured at treatment well T2 and compass points.

using variograms with assumed data transformations that resulted in a minimum mean square error while keeping residual variance near zero (*21*). The results were plotted using a computer program (Matlab, The Mathworks, Inc., Natick, MA). Because of the pulsing strategy used, toluene concentrations in the treatment wells and at the near monitoring wells normally varied considerably. Toluene concentrations in T1 or T2 were measured as high as 100 mg/L during pulses. In order to prepare Figure 10, measured concentrations at each monitoring location were averaged over the period from day 220 to day 260. A significant decrease in concentration as water moves from the treatment well to the monitoring wells results. Of particular interest is the toluene concentration arriving at the compass points, where the agreed upon goal was to maintain the concentration below 20 μg/L. This was always achieved in both aquifers.

Illustrated in Figure 11 are the measured concentrations at the treatment and four monitored compass point wells for the upper aquifer for the first two steady-state periods. Results for the third steady-state period and the lower aquifer were similar. Only on three occasions were measured concentrations at the compass point wells greater than 10 μg/L, but 20 μg/L was never reached. The average toluene concentration in the 588 compass point samples analyzed over the course of this study was 1.1 ± 1.6 μg/L. Obtaining accurate concentration measurements at the compass points proved difficult as all samples, including those with concentrations ranging up to 100 000 μg/L, were measured with the same instrument. Some cross contamination between samples was unavoidable. While all compass point samples had concentrations well below the regulatory goals, the values are likely to have been even lower if possible cross contamination between samples could be completely avoided. In any event, toluene removal between the treatment wells and the bounds of the compass points was very efficient, generally exceeding 99.98%, which is consistent with the findings from Moffett pilot studies (*1*) and Edwards laboratory microcosm studies (*7*).

Regional TCE Removals. The initial and final TCE distributions throughout the study area in the upper aquifer were examined using the variogram approach described for the three-dimensional views of toluene distribution and are illustrated in Figure 12, panels a and b, respectively. Figure 12a represents average TCE concentration just before continuous toluene injection began (days 20−55). Groundwater flowed from the northwest to the southeast (Figure 5). At the north and west compass points, TCE concentrations were initially in the 1200−1400 μg/L range in both aquifers. The concentrations dropped rapidly downgradient toward the southeast to a more or less level concentration of about 600 μg/L throughout the rest of the sampling field. This leveling of concentration was the result of groundwater mixing during the first 50 days of system testing. Figure 12b illustrates the study zone TCE distribution near the end of the steady-state period (days 220−260) for comparison. A significant drop in TCE concentration had occurred throughout the study area over the 200 days of continuous toluene addition (compare Figure 12, panels a and b). The 27 μg/L shown at the south compass point represents TCE concentration in the treated water moving out of the study area and downgradient with the regional flow. This change from the TCE concentration of 1149 μg/L moving into the study area at the north compass point represents a TCE removal in passing through the study area of 97.6%, a result of several cycles of the water through the treatment wells.

Dissolved Oxygen Concentration. Figure 13 represent a three-dimensional view of the average DO distribution over days 220−260 at the end of the steady-state period in the upper aquifer. The DO in the native groundwater was zero. The plume water arrived by the north compass point and is near zero there (1 μg/L). The peak shown of 25 mg/L is at the T2 treatment well where oxygen was added. The DO in the water exiting the treatment zone at the southeast boundary was about 12 mg/L, representing the excess in DO added over and above that needed for toluene oxidation and organism respiration. DO was successfully maintained throughout the upper aquifer monitored areas as required for toluene oxidation and TCE removal. In the lower aquifer where channeling occurred, the DO distribution was not as uniform and approached zero near the north, west, and south compass points (not shown). Nevertheless, DO was present where toluene was present so that toluene could be effectively removed and TCE could be degraded.

Biomass Distribution. Biomass distribution throughout the treatment zone can be inferred from the areas where toluene consumption was the greatest, which was near the T1 and T2 treatment wells. However, distribution can also be estimated from DO uptake at the various monitoring locations following shutdown of pumping, which occurred

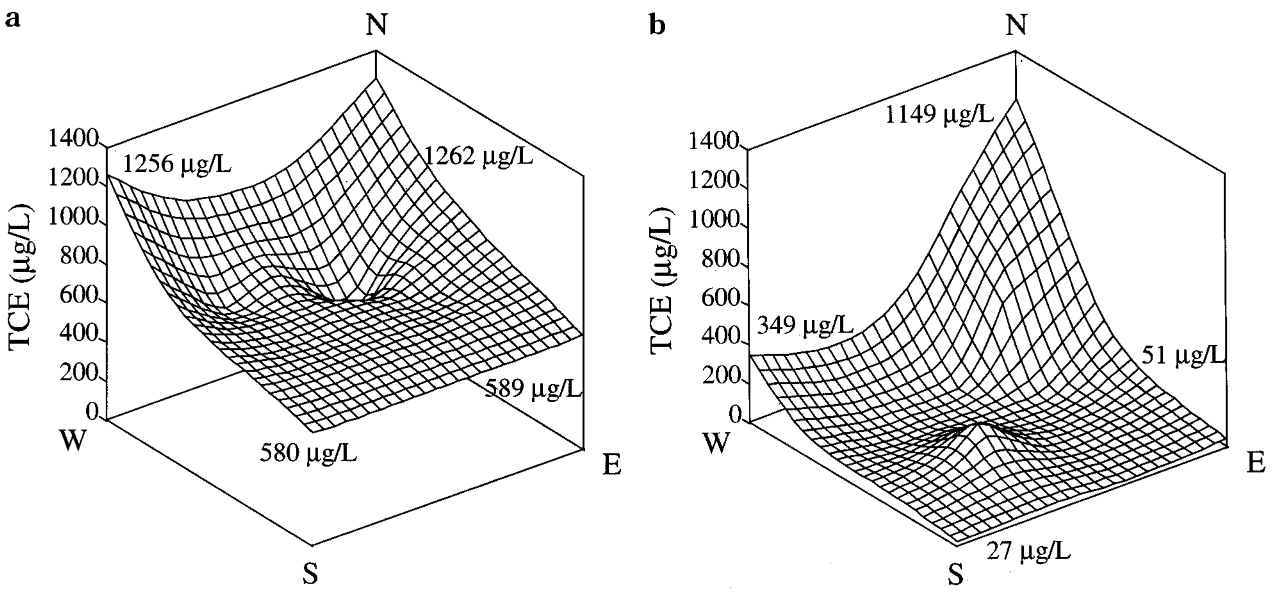

FIGURE 12. Three-dimensional view of TCE concentration in upper aquifer. (a) Over the study area prior to toluene addition (averages for days 20−55). (b) Near the end of the steady-state period (averages for days 220−260).

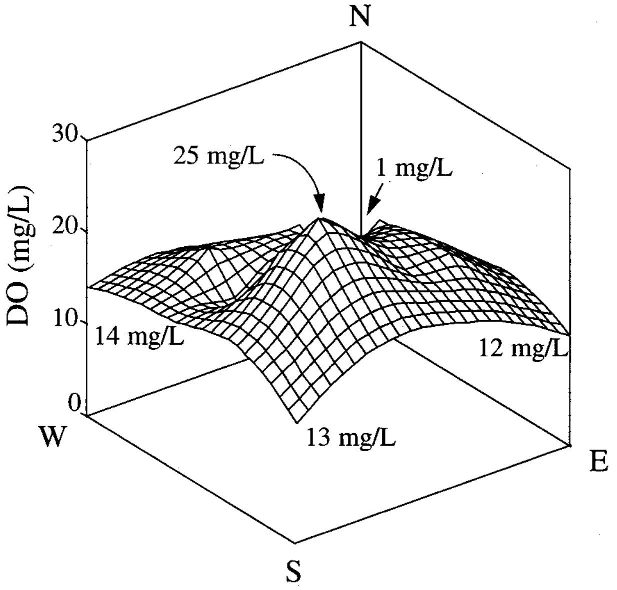

FIGURE 13. Three-dimensional view of DO concentration in the upper aquifer near the end of the steady-state period (averages for days 220−260).

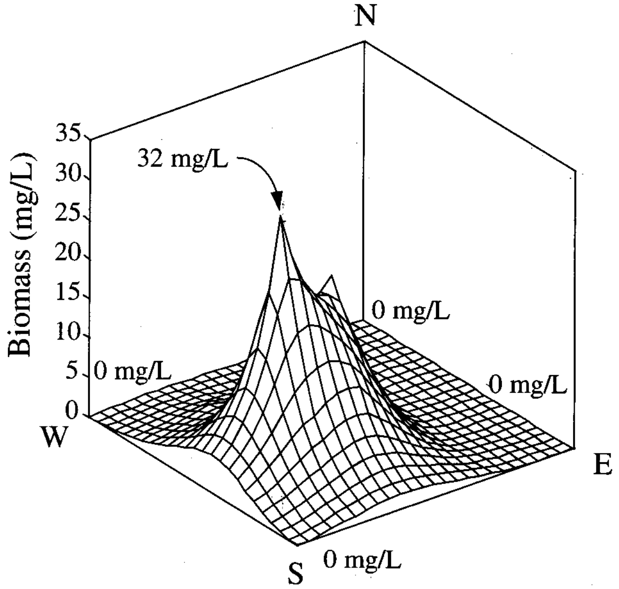

FIGURE 14. Three-dimensional view of biomass concentration (dry weight) estimated for the upper aquifer from DO uptake rates during shut-down periods between days 136−142 and days 204−209.

three times during the study period following toluene addition. Where biomass was present, the DO concentration decreased due to respiration by the organisms as well as due to oxidation of residual toluene. Rates of DO utilization following toluene depletion were determined for shutdown periods on days 136−142 and 204−209. A typical biomass decay rate (*b*) of 0.15/d and oxygen uptake from microbial decay of 1.42 of mg DO/mg of biomass decay was taken from the Edward microcosm results (*7*). With this procedure, the biomass concentration at a given point (X_i, mg/L) may be estimated from the following relationship:

$$X_i = \text{DO uptake rate at point } i \text{ (mg L}^{-1}\text{ d}^{-1})/(0.15 \text{ d}^{-1})(1.42) \quad (2)$$

Figure 14 represents three-dimensional views of biomass distribution in the upper aquifer based upon this analysis. The results for the lower aquifer were similar. As anticipated, most of the biomass appears to reside near the treatment wells and has an estimated maximum dry weight biomass concentration of 32 mg/L, while the average concentration over the 480 m^2 study area was 3.4 mg/L in the upper and 2.56 mg/L in the lower aquifer.

The total biomass in the upper and lower aquifer can then be estimated from the volume of the treatment zones assuming a typical aquifer porosity of 0.3. On this basis for the upper 8 m thick aquifer, the total biomass represented is 3.95 kg dry weight and for the 5 m thick lower aquifer is 1.86 kg dry weight. A second independent approach for estimating total biomass under steady-state operation was provided from the Edwards microcosm study (*7*) through an equation presented there:

$$\text{total steady-state biomass} = X_{a(ss)}V_p = \frac{YS^oQ}{b} \quad (3)$$

Y for Edwards microcosms was found to be 0.77 mg of cells/

mg of toluene (*7*). For the full-scale evaluation in the upper aquifer, S^o was 13.4 mg/L toluene and Q was 38 L/min, while for the lower aquifer, S^o was 11.6 mg/L and Q was 25 L/min. Substituting into eq 3 results in estimated biomass through this approach of 3.76 kg dry weight for the upper aquifer and 2.14 kg dry weight for the lower, values that are within 20% of the values estimated from field DO uptake measurements. Such close agreement was unexpected and undoubtedly involves some measure of coincidence. Nevertheless, the closeness of the two independent total biomass estimates provides some validation for the use of DO uptake measurements for estimating biomass distribution throughout the aquifers, estimates that would be very difficult and costly to obtain by most other approaches.

Pumping Head Changes. The Edwards site hydraulic conductivities of about 3.4×10^{-3} cm/s are sufficiently low that they limit the pumping rate possible in the subsurface treatment system. The finer aquifer material at Edwards than for the pilot field studies at Moffett Federal Airfield led to a closer evaluation of bioclogging potential. This can be characterized by the changes in pumping head required to maintain the constant flows at the two treatment wells. Water surface measurements for the upper aquifer were made manually at T1 and T2, and pressure measurements in the lower casings below the pumps were made through pressure transducer readings. The net changes in total head across the pumps during pumping obtained from this combination of measurements are illustrated in Figure 9. The pumping rate to the upper aquifer at T2 was about 50% higher than to the lower aquifer at T1 during unbalanced flow rate conditions (days 50−271), accounting for the greater average pumping head at T2. With balanced flow rates (days 317−444), the higher pumping head was at T1, although there was then little difference between the two.

The pumping heads generally increased with time, presumably as a result of biomass buildup within the aquifer near the treatment wells. However, some transport of fine materials from the aquifer being extracted may also have been involved. The addition of hydrogen peroxide at day 81 appeared beneficial for lowering the pumping head at T1, where a higher peroxide concentration of 71 mg/L was used, but not at T2, where only 47 mg/L was used. Well T1 was shut down for redevelopment due to a sudden head increase at day 134 and at day 204 as a routine procedure. Redevelopment of T1 resulted in the pumping head returning to near its initial value before toluene injection was begun. Both wells were shut down for routine redevelopment on days 204 and 272, and this brought the pumping head to near the initial values.

These pumping head increases following redevelopment indicate that some aquifer clogging did occur. The impact could be mitigated by periodic well redevelopment. Also, hydrogen peroxide addition and once per day toluene pulsing appear to be beneficial in reducing clogging potential. The pumping heads noted in Figure 9 are small in any event, much less than would be required simply to pump the water to the ground surface (about 10 m). Thus, excessive concern may have been paid to aquifer clogging potential in this study. Probably, well redevelopment could have been carried out on a less frequent basis, and the hydrogen peroxide addition could have been reduced.

Discussion

The major purpose of this study was to demonstrate at full scale that *in situ* cometabolic biodegradation of TCE through toluene injection is a viable process and that the scientific understanding of the process is satisfactory for predicting performance at a site. The microcosm studies that were used as a basis for predicting treatment performance (*7*) suggested that the potential for TCE removal differed with location in the aquifer and varied between 87 and 99%. The modeling effort using the relevant biodegradation coefficients from that study and hydrogeological characteristics of the aquifer itself indicated about 83% TCE removal should be obtained with each pass through the treatment system. The steady-state removal of 86% in the upper and 83% in the lower aquifer obtained during the balanced flow period with 9 mg/L toluene addition indicates that good agreement was obtained between predicted and actual results.

Aquifer capture zones resulting from the 25 L/min balanced flow conditions during the last study phase were modeled and compared with that for the 38 L/min illustrated in Figure 3. This indicated the width of the capture zones were 62 m and 53 m in the upper and lower aquifers, respectively. This simulation also indicated that the T1 flow had 71% recycle flow from T2 and that the T2 flow had 85% recycle flow from T1. If it is assumed that the TCE concentration in the regional flow entering the test zone is 1000 μg/L (near that measured), the concentration in the lower aquifer recycle flow is 18 μg/L (Table 5, N10-L), and the concentration in T2 is 171 μg/L (Table 5), a mass balance indicates that recycle flow is 84% at T2 (very close to the modeled value). However, a similar balance for the upper aquifer indicates the recycle flow in T1 is 91.5% rather than the modeled 71%. The difference here is quite significant and may be due to any one of a number of factors, such as inadequate time to reach a true steady-state or errors in modeling assumptions made concerning such factors as aquifer thickness, regional velocity, or homogeneity. A more detailed site characterization would be needed to determine the factor or factors involved.

The decrease with time in the concentration of TCE passing through each treatment well due to recirculation of treated water and mixing with untreated upgradient plume water was similar to that anticipated (compare Figures 4 and 8). During balanced flow conditions, treatment of the water entering the upper aquifer from T2 reduced the T2 TCE concentrations by 86% over the last 80 days of near steady-state operation to an average 24 μg/L. The reduction from 1000 to 24 μg/L as groundwater passed through the treatment system represents an overall TCE removal efficiency of 97−98%, about what was suggested from the modeling studies. With the lower 18 μg/L in water exiting the treatment zone in the lower aquifer, the percent removal is even higher.

Potential advantages of this *in situ* biological system over traditional pump-and-treat systems are (1) cost for pumping of groundwater to the surface is avoided, (2) no above ground treatment system is required to treat groundwater contaminants, (3) TCE is destroyed in the process and not simply concentrated in another medium for disposal, (4) disposal of treated groundwater is not an issue, and (5) uncontaminated groundwater is not wasted by being brought into the TCE contaminated zone as generally occurs in pump-and-treat systems.

The latter is a major issue in water shortage areas. Pump-and-treat systems withdraw contaminated water from an aquifer, but this causes a gradient by which uncontaminated groundwater is brought into the zone. Contaminants sorbed to the soil in that zone then desorb and contaminate this freshwater. Following pumpage to the surface, this water too generally requires treatment, and regulations may prohibit returning the treated water to the aquifer. Thus, water lost in this manner can represent a wastage of a valuable resource.

There are also disadvantages of the biological treatment system. Toluene is a regulated chemical, and there is a concern that it may not be biodegraded if added to groundwater. The experiences both at Moffett Field and Edwards Air Force Base indicate that as long as sufficient oxygen is present and the aquifer has sufficient nitrogen, phosphorus, and other nutrients for biological growth, concentrations well below levels of any regulatory concern can be obtained. Fortunately, toluene is also biodegradable anaerobically so that even if sufficient oxygen were not available due to some system failure the problem posed may not be severe.

The other major possible problem with this process, especially in low conductivity aquifers, is the potential clogging due to excessive biological growth near the treatment well or fines brought from the aquifer being extracted: Two strategies were used here to reduce bioclogging besides well redevelopment, one was pulsing of the primary substrate so that it can be distributed further out into the aquifer before biodegradation the other was to add an inhibitory material such as hydrogen peroxide. Peroxide is a good chemical for this purpose as it serves to inhibit growth near the treatment well but hydrolyzes out in the aquifer to produce oxygen, which is needed by the process. The disadvantage of hydrogen peroxide is its expense. Another problem is that it is inhibitory at higher concentration so that it adversely affects the treatment process as found here in the lower aquifer during the steady-state operation period. Well redevelopment was found to be effective for reducing pumping heads to near initial values at Edwards. Such redevelopment was used three times for the lower aquifer and twice in the upper aquifer, although it is questionable whether redevelopment was needed so often. It is also possible that the 47 mg/L hydrogen peroxide addition used during balanced flow conditions was in excess of that needed to control clogging. There is probably some trade off in costs between hydrogen peroxide addition and redevelopment. Both add significant cost to the treatment system. Such costs should be greatly reduced with treatment in an aquifer having larger sized materials than the fine to medium sand present at Edwards. In any event the increase in pumping head with time at the two wells was quite small.

The dual-well system spanning two aquifers is a unique approach that may not appear to be applicable to many contamination sites. However, the dual-well dual-screen system may also be applicable to single aquifer systems where low permeability layers separate lower and upper screen intervals, leading to hydraulic conductivity anisotropy, where vertical hydraulic conductivity is significantly lower than horizontal conductivity. The system itself has now been demonstrated through this study to be technically feasible and might be applied in other locations with suitable modifications. For example, with a relatively homogeneous single contaminated aquifer, groundwater might be pumped to the surface from one location and then reinjected at another location in the same aquifer with chemicals either added at the surface or downwell at the injection location (*22*). Such a standard approach in effect achieves in a single aquifer a groundwater treatment regime similar to that used here.

The major operational costs of the *in situ* treatment system are those associated with prevention of clogging. This includes the cost for chemicals such as hydrogen peroxide, used to inhibit growth, and well redevelopment after some clogging has occurred. In order to reduce such costs, other avenues are worthy of exploration, such as different designs around the injection screens. The degree of this problem will depend to some extent upon the characteristics of the aquifer itself; coarser materials will have even less tendency to clog. This study has demonstrated that engineered *in situ* biodegradation of TCE using aerobic cometabolism can work. It may have advantages over alternative technologies at some locations but not at others. The advantage for treating plumes containing other readily-cometabolized chlorinated aliphatic hydrocarbons, such as vinyl chloride, is obvious for primary substrates other than toluene can be highly effective. This demonstration and evaluation represent another step in the evolution of *in situ* bioremediation approaches.

Acknowledgments

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