FINAL REPORT

Remediation of DNAPL through Sequential In Situ Chemical Oxidation and Bioaugmentation

ESTCP Project ER-0116

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LIST OF ABBREVIATIONS

ASTM	American Standard for Testing and Materials
C-C	carbon-carbon
C_2Cl_3H	tetrachloroethene
C_2Cl_4	trichloroethene
Cl^-	chloride
CMT	continuous multichannel tubing
CO_2	carbon dioxide
cDCE	cis-1,2 dichloroethene
Dhc	*Dehalococcoides*
DNAPL	dense non-aqueous phase liquid
DoD	Department of Defense
DOE	Department of Energy
DQI	data quality indicators
DQO	data quality objectives
ESB	Engineering Support Building
ESTCP	Environmental Security Technology Certification Program
EVO	emulsified vegetable oil
EZVI	emulsified zero-valent iron
ft	feet
GAC	granular activated carbon
Geosyntec	Geosyntec Consultants, Incorporated
H^+	hydrogen
H_2O_2	hydrogen peroxide
ID	inside diameter
ISB	enhanced in situ bioremediation
ISCO	in situ chemical oxidation
KSC	Kennedy Space Center
LC-34	Launch Complex 34
LSU	lower sand unit
MCL	maximum contaminant level
MDL	method detection limit
MFGU	middle fine-grained unit
mg/L	milligrams per liter
mM	millimolar
MNA	monitored natural attenuation
MnO_2	manganese dioxide

MnO_4^-	permanganate
MS/MSD	matrix spike/matrix spike duplicate
NASA	National Aeronautics and Space Administration
OD	outside diameter
O&M	operations and maintenance
ORP	oxidation-reduction potential
PCE	tetrachloroethene
PLC	programmable logic controller
PLFA	phospholipid fatty acid
ppb	parts per billion
PTA	pilot test area
PVC	polyvinyl chloride
QAPP	quality assurance project plan
QA/QC	quality assurance/quality control
QPCR	quantitative polymerase chain reaction
RCRA	Resource Conservation and Recovery Act
RFI	RCRA Facility Investigation
SAP	Sampling and Analysis Plan
SPH	six phase heating
TCE	trichloroethene
TOC	total organic compounds
USEPA	U.S. Environmental Protection Agency
USU	upper sand unit
UT	University of Toronto
VC	vinyl chloride
VFA	volatile fatty acid
VOC	volatile organic compounds

EXECUTIVE SUMMARY

The principal benefit of in situ chemical oxidation (ISCO) using permanganate (MnO_4^-) is that it aggressively enhances dissolution and destruction of the target contaminants within a relatively short period of time (i.e., months to years); however, the cost-benefit of this technology diminishes as the mass of target chemicals decreases. The most effective application of ISCO consists of rapid destruction of the readily accessible target chemical mass within the source area, although it can also be coupled with a less costly, in situ remediation mass removal technology such as in situ bioremediation (ISB).

The main objectives of this project was to assess the technical feasibility of sequential application of these technologies and to identify the optimal timing of the transition from ISCO to ISB.

The principal results of the project include:

- Electron donor addition (ISB) after ISCO resulted in partial biodegradation of trichloroethene (TCE), with complete biodegradation observed after bioaugmentation;
- At the field-scale, ISB did not increase the mass flux of chloroethenes after ISCO;
- The precipitated manganese dioxide produced by MnO_4^- reduction, which can oxidize some organic compounds, did not abiotically degrade any of the chloroethenes or ethene;
- Manganese dioxide (MnO_2) greatly increases the electron donor demand above that typically required to reduced the dissolved constituents (e.g., oxygen, nitrate, sulfate, and the target chloroethenes) during ISB;
- MnO_2 can be dissolved by the activity of Mn(IV)- reducing bacteria, that appear to preferentially utilize hydrogen and inhibit the activity of dechlorinating microorganisms (i.e., *Dehalococcoides*, which use hydrogen as their sole electron donor); and
- The limited cost assessment indicated that there was a significant cost and schedule advantage for the sequential treatment strategy over using pump and treat or ISCO alone.

1.0 INTRODUCTION

1.1 Background

Chlorinated solvents such as tetrachloroethene (PCE) and trichloroethene (TCE) are present in groundwater as dense non-aqueous phase liquids (DNAPLs) at many Department of Defense (DoD), Department of Energy (DOE), and related contractor facilities. DNAPLs have very low aqueous solubility's that may exceed regulatory criteria by as much as five orders of magnitude (Pankow and Cherry, 1996); as a result, these compounds only slowly dissolve in groundwater and act as long-term sources of groundwater contamination.

The physico-chemical properties of PCE and TCE make these contaminants particularly difficult to remove from groundwater systems. It is now widely recognized that removal using groundwater extraction and above-ground treatment (pump-and-treat) is only effective as a containment approach due to the slow dissolution of solvents from residual or pooled DNAPL sources (U.S. Environmental Protection Agency [USEPA], 1992; National Research Council, 1994). These systems will require operation over indefinite periods of time (i.e., decades to centuries) incurring continuing annual operations and maintenance (O&M) costs over that period. Accordingly, treatment technologies that enhance the dissolution rate of a DNAPL will decrease the remediation time, which ultimately reduces total lifecycle costs of remediation. The difficulty in removing PCE and TCE DNAPL from contaminated aquifers has emphasized the need for effective in situ treatment technologies that target DNAPL source zones. In situ treatment technologies capable of treating DNAPL source zones are listed in Table 1-1. Those technologies offering mass destruction are advantageous in that the DNAPL mass is not simply transferred into a second matrix but destroyed in situ.

Laboratory experimentation and field applications have demonstrated that in situ chemical oxidation (ISCO) with permanganate (MnO_4^-) is an effective technique for degrading chlorinated solvents (e.g., Schnarr *et al.*, 1998; Hood and Thomson, 2000; Thomson *al.*, 2000). ISCO typically involves injection and/or recirculation of a concentrated oxidant solution to promote rapid oxidation of the target chemicals. MnO_4^- attacks the carbon-carbon (C-C) double bonds in chlorinated ethenes (e.g., TCE) mineralizing the target compound to inorganic products such as carbon dioxide (CO_2), water, and chloride (Cl^-).

Table 1-1. Summary of DNAPL Remediation Technologies

Focus	*Technology Class*	*Remediation Technology*	*Physico-chemical Remediation Process*
Plume Management	Reactive Barriers	Zero-valent Iron	-minimizes the migration of contaminated groundwater by intercepting and degrading the dissolved phase contaminants
	Containment	Impermeable Walls Pump and Treat	-minimizes the migration of contaminated groundwater by either preventing groundwater flow or hydraulically containing the contaminated groundwater
	Bioremediation	Monitored Natural Attenuation	-minimizes migration of contaminated groundwater by degrading the dissolved phase contaminant
Source Management	Flushing	Alcohol Surfactant	-removes DNAPL by either mobilizing pure phase or increasing the solubility of the contaminant
		Oxidant	-removes DNAPL by rapidly degrading the dissolved phase contaminant
	Volatilization	Soil Vapour Extraction Air Sparging In-well Stripping	-removes vapor phase contaminant from either the vadose or saturated zones by enhancing partitioning into the vapor phase
	Thermal	Steam Flushing Electrical Heating In Situ Vitrification	-removes DNAPL by enhancing volatilization and/or mobilizing the pure phase
	Enhanced Bioremediation	Biostimulation	-removes DNAPL mass by enhancing the rate of biodegradation within the source zone
		Bioaugmentation	-minimizes migration of contaminated groundwater (increases degradation rate and promotes complete dechlorination to ethene) by increasing the activity of dechlorinating microorganisms

Note

[1] After Fountain (1998)
DNAPL - Dense Non-aqueous Phase Liquid

The principal benefit of the ISCO technology is that it aggressively enhances dissolution and destruction of the target contaminants within a relatively short period of time (i.e., months to years) in comparison to conventional treatment technologies. However, the cost-benefit of ISCO diminishes as the mass of target chemicals decreases, particularly at sites where low permeability zones limit mass transfer. Results of a technology status review of in situ oxidation technology demonstrations indicated that rebound of volatile organic compounds (VOC) concentrations was observed at many ISCO sites, and that re-application of the oxidant or implementation of a secondary polishing technology was required (Environmental Security Technology Certification Program [ESTCP], 1999). Based on the current status of the technology, it appears that the most effective application of ISCO consists of rapid destruction of the readily accessible target chemical mass within the source area coupled with a less costly and more passive in situ remediation approach to control the remaining mass (e.g., in situ bioremediation [ISB] or natural attenuation).

Like ISCO, in situ bioremediation technologies have rapidly evolved in recent years to the point where demonstrations are being conducted to evaluate the technical and economic feasibility of

DNAPL source zone bioremediation. Like ISCO, rapid biological destruction of dissolved-phase chlorinated solvents can enhance dissolution of the chlorinated solvent DNAPLs, to reduce the duration and cost of remediation. However, while TCE half-lives are on the order of minutes with ISCO, biodegradation rates are typically on the order of hours to days, suggesting that the rate of DNAPL removal using ISB is likely to be less than that achieved during ISCO, but may still be high enough so that ISB can be used as a secondary source treatment technology. This is contingent upon increasing microbial activity in the vicinity of the DNAPL and overcoming mass transfer limitations. In the event that mass transfer enhancements by ISB are negligible the enhanced biodegradation rates provide significant benefit through biological containment of the remaining VOC in groundwater.

Unfortunately, little is known regarding the impact of ISCO on groundwater geochemistry and microbiology. Specifically, the application of an aggressive oxidant such as MnO_4^- may have adverse impacts on the indigenous microbial community such that bioremediation of the chlorinated solvents cannot be stimulated through electron donor addition alone. Re-seeding (bioaugmentation) of the ISCO treatment area with microorganisms capable of degrading chlorinated solvents (e.g., dehalorespirers) may be required to permit successful implementation of in situ bioremediation as a polishing technology. To achieve this objective, several dehalogenating microbial cultures are available for field application, including the Pinellas (Ellis *et al.*, 2000; Harkness *et al.*, 1999) and KB-1™ (Duhamel *et al.*, 2002; Major *et al.*, 2002) cultures. Both cultures contain *Dehalococcoides* bacteria, which are the only dehalorespiring bacteria capable of completely dechlorinating TCE to ethene (Maymo-Gatell *et al.*, 1997), and have been used in laboratory and/or field trials to successfully promote rapid and complete dechlorination of PCE and TCE to ethene.

Coupling the ISCO primary source treatment technology, to rapidly remove accessible DNAPL mass, with semi-passive in situ bioremediation via biostimulation (if possible) or bioaugmentation (likely to be required) as a secondary source treatment or plume containment technology is an attractive remediation approach. The combined treatment approach is expected to reduce the duration and cost of remediation at chlorinated solvent sites (relative to application of either technology alone or in conjunction with other technologies), which will in turn reduce the financial drain of these sites on DoD funds and programs.

1.2 Objectives of the Demonstration

The primary objectives of the demonstration were:

- Determine the impacts of ISCO application on the natural microbial community (biomass, diversity) and specifically on the presence of dehalorespiring bacteria, and determine whether the post-ISCO indigenous microbial community can be stimulated

to biodegrade remaining chlorinated solvents, or whether bioaugmentation is required to re-seed the treatment area to promote in situ bioremediation;

- Assess the impacts of ISCO on the groundwater chemistry and microbiology, and identify aquifer conditioning requirements for application of in situ bioremediation;
- Identify the appropriate switchover point from chemical oxidation to enhanced bioremediation;
- Demonstrate in situ bioremediation of VOC remaining in groundwater following ISCO treatment using either biostimulation (addition of electron donors only) or bioaugmentation (addition of dehalorespiring bacteria and electron donors), as required; and
- Evaluate whether ISB will act as a secondary mass removal technology or mass containment technology following ISCO.

The study approach consisted of a field trial to demonstrate that biostimulation and/or bioaugmentation can stimulate complete dechlorination of a non-toxic product (i.e., providing a mass containment) and whether the mass flux from a source zone increases when biological dehalorespiration activity is enhanced through nutrient addition and bioaugmentation (i.e., providing a secondary source removal technology post-ISCO). The field demonstration was conducted at Launch Complex 34 (LC-34), an unused launch facility at the Kennedy Space Center (KSC), Florida, where an extensive TCE DNAPL source is present in groundwater in the area adjacent to the Engineering Support Building (ESB). Historical records suggest that chlorinated organic solvents, including TCE, were used to clean rocket engines on the launch pad and on outdoor racks along the west side of and inside the ESB. During cleaning operations, solvents evaporated, infiltrated directly into the subsurface, or migrated as runoff into drainage pits. The National Aeronautics and Space Agency (NASA) is currently in the process of developing remedial alternatives as part of the ongoing Resource Conservation and Recovery Act (RCRA) response actions at LC-34. The results of this technology demonstration were incorporated into the process for selecting a final source zone remediation technology.

To date, a number of remediation technology demonstrations have been conducted at LC-34. In 1998, three test plots measuring 50 feet (ft) by 75 ft were established on the north side of the ESB and subsequently used for demonstrations of ISCO using MnO_4^- (completed 2000), six phase heating (completed 2001), and steam flushing (completed 2002). In addition, Geosyntec Consultants, Incorporated (Geosyntec) has completed demonstrations of enhanced bioremediation using bioaugmentation and DNAPL removal with emulsified zero-valent iron in smaller test plots located within the ESB. Of particular interest for this demonstration project is the ISCO test plot, which was used to contain the pilot test area (PTA) for this technology

demonstration. The selection of LC-34 as the demonstration site reduced the requirement for ISCO as part of this technology demonstration.

During the demonstration, groundwater was recirculated through the PTA at a constant groundwater velocity. A number of treatment phases were used to evaluate the rate of DNAPL removal and the extent of VOC treatment. Each phase was operated for sufficient duration to establish a near "steady-state" rate of TCE removal under each of the different operating conditions (i.e., baseline groundwater recirculation only, electron donor addition, electron donor addition, plus bioaugmentation).

1.3 Regulatory Drivers

Since 1976, both PCE and TCE have been designated by the USEPA as priority pollutants. The Safe Drinking Water Act Amendments of 1986 strictly regulate these compounds; each has a maximum contaminant level (MCL) in drinking water of 5 parts per billion (ppb; USEPA, 1996). When concentrations of these compounds at a contaminated site exceed these criteria, remedial action is required to lower these concentrations and reduce the risk to human health and the environment.

Additionally, the DoD lists the following directives as high priority requirements:

- Navy: 1.I.1.g. *Improved remediation of groundwater contaminated with chlorinated hydrocarbons and other organics.*
- Army: A(1.2.c) *Enhanced Alternative and In-Situ Treatment Technologies for Solvents and Halogenated Organics in Groundwater (96-97)*
- Air Force: 2008: *Methods and Remedial Techniques are Needed to More Effectively Treat Groundwater Contaminated with Chlorinated Solvents Such as TCE, TCA, and PCE*

1.4 Stakeholder/End-User Issues

The demonstration helped to assess the applicability of sequencing enhanced bioremediation with chemical oxidation. It has provided the fundamental technology components (e.g., level of monitoring, monitoring parameters, sampling frequency, distribution or mixing of nutrients/microorganisms, nutrients loading) to guide the application of this technology at other sites where the technology application may be evaluated. Secondary impacts of both the ISCO and ISB technologies were monitored and evaluated over the demonstration. These included impacts such as the possible generation of methane and hydrogen sulfide during ISB and

potential secondary water quality impacts (e.g., increased biochemical oxygen demand during ISB, enhanced solubility of metals such as iron and manganese during ISCO and ISB).

The demonstration also provided an evaluation of the extent of the enhancement in the DNAPL removal rate and the anticipated decrease in treatment time to justify the selection of sequential chemical oxidation/bioremediation as an effective source remediation alternative. In addition, the demonstration provided operational and performance data that will allow informed evaluation of the technology at other facilities.

2.0 Technology Description

The following sections provide: an overview of the ISCO and ISB technologies (Section 2.1), a summary of the theoretical impacts of ISCO on ISB (Section 2.2), an overview of previous studies evaluating the sequential ISCO/ISB approach (Section 2.3), key technology factors impacting cost and performance (Section 2.4), and a description of the potential advantages and limitations of the sequential technology approach (Section 2.5).

2.1 Technology Development and Application

Conventional groundwater remediation technologies have emphasized treatment of contaminants present in the dissolved phase plume migrating downgradient of the DNAPL source area. While a number of plume management technologies, including pump-and-treat, air sparging, and permeable reactive barriers, have proven effective in containing plume migration, the low solute flux from many DNAPL source zones implies that O&M of plume remediation technologies will be required for an indefinite duration ranging from decades to centuries (Johnson and Pankow, 1992). The rate of removal of DNAPL mass from the subsurface, identified as one of the principal impediments to the effectiveness of groundwater remediation efforts (National Research Council, 1994), is limited by low aqueous solubility and weak mixing effects. Accordingly, research in the last decade has emphasized the development of treatment technologies, such as those described in Table 1-1, which aggressively remove and/or degrade DNAPL. These technologies provide the benefit of reducing the time required for clean-up by increasing the mass flux from the source zone; however, the applicability of these technologies may be limited by cost, regulatory acceptance, and uncertain performance.

The performance of remediation technologies can be expressed in terms of the enhancement in the rate of DNAPL removal during treatment relative to the rate of removal under conditions of ambient groundwater flow (i.e., no applied treatment). In general, the mass transfer enhancement provided by a technology under treatment conditions is proportional to operating cost. For example, bioremediation, generally thought to be of limited effectiveness in DNAPL source areas (Pankow and Cherry, 1996), results in a lower mass transfer enhancement than ISCO but requires only the addition of a dilute nutrient solution (i.e., minimal operating cost) while ISCO, which can result in a relatively large mass transfer enhancement, requires the addition of concentrated MnO_4^- solution (i.e., high operating cost). Strategically coupling these technologies to match the rate of DNAPL mass removal from the source may be used to minimize the lifetime cost of source area remediation.

To minimize the cost of source zone remediation, technology selection must be consistent with conditions present in the source area; however, multiple laboratory and field studies have

demonstrated that conditions in the source zone change over time. As a result of the depletion of DNAPL mass and the accompanying decrease in DNAPL:water interfacial surface area, the rate of mass transfer gradually decreases, implying that different technologies are required to cost-effectively treat the source zone at different times. Over time as DNAPL mass is removed, the enhancement in the rate of mass transfer will diminish, eventually reaching a point where the presence of the treatment reagent does not increase the rate of mass removal. Figure 2-1 presents a scenario where the primary mass removal technology (i.e., ISCO), which is capable of rapid removal rates, efficiently removes DNAPL mass until this point of diminishing returns occurs; however, as the rate of mass removal decreases, the cost of per unit DNAPL mass removed will rapidly increase. Accordingly, a secondary, lower operating cost mass removal technology (i.e., ISB) with a lower maximum mass removal rate capability, is better suited to the mass transfer conditions at this point in time.

As previously discussed, the sequential application of ISCO using MnO_4^- and ISB can potentially achieve the objective of reducing technology operating costs while removing DNAPL mass at the maximum possible rate. Although ISCO involves the comparatively high operating cost of continuous oxidant addition, this technology can result in up to 40-fold enhancements of the DNAPL removal rate (Schnarr *et al.*, 1998). In contrast, the operating cost of ISB is much lower (requiring the addition of relatively inexpensive electron donors) while a maximum of 16-fold enhancements in the rate of DNAPL removal may be achievable (Cope and Hughes, 2001). As previously discussed (Sections 1.1 and 1.2), DNAPL removal using ISB is contingent on mass transfer rate limitations imposed by the deposition of MnO_2, and in the event that there is no rate enhancement, this technology may be used to simply contain the remaining VOC in groundwater. The following sections provide a description of ISCO (Section 2.1.1), a rationale for the switchover point between the ISCO and ISB (Section 2.1.2), and a description of ISB (Section 2.1.3).

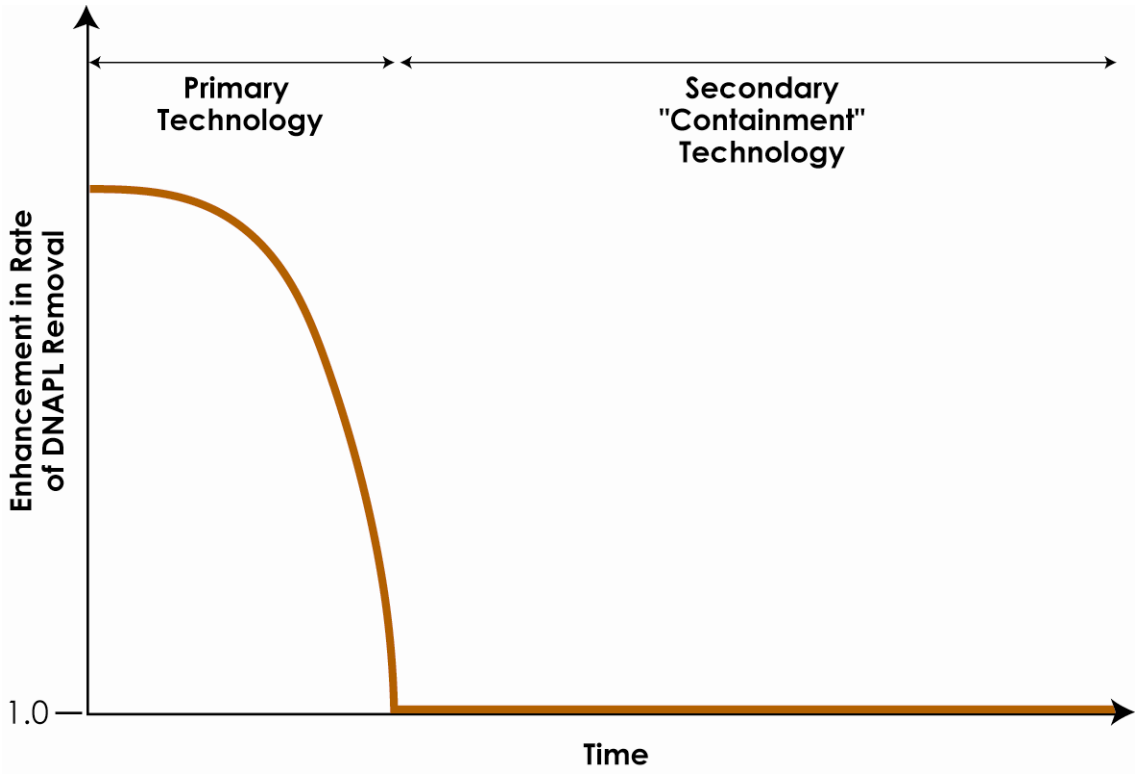


Figure 2-1. Sequential Treatment of DNAPL Using Complementary Technologies

2.1.1 In Situ Chemical Oxidation Using Permanganate

Various oxidants have been used in laboratory and field applications to aggressively destroy PCE and TCE DNAPL, including MnO_4^- and Fenton's reagent (hydrogen peroxide [H_2O_2] and a ferrous iron catalyst). Of these, MnO_4^- offers significant advantages because it: i) is less reactive with aquifer solids, resulting in improved oxidant delivery to the target contaminants; ii) is typically more stable and safer to handle than Fenton's reagent; iii) does not require pH adjustment with concentrated acid; and iv) produces less heat and insoluble gas in the treatment zone.

The reaction between MnO_4^- and chlorinated ethenes involves an electrophilic attack on the ethene's C-C double bonds and the formation of a cyclic hypomanganate ester. Rapid hydrolysis of the cyclic ester results in the production of carbon dioxide. The stoichiometric reactions describing the oxidation of TCE and PCE by MnO_4^- is given by Yan and Schwartz (1999),

$$C_2Cl_3H + 2MnO_4^- \rightarrow 2CO_2(g) + 2MnO_2(s) + 3Cl^- + H^+ \qquad (1)$$

$$C_2Cl_3H + 2MnO_4^- \rightarrow 2CO_2(g) + 2MnO_2(s) + 3Cl^- + H^+ \qquad (2)$$

where TCE and PCE are presented by the chemical formulae C_2Cl_4 and C_2Cl_3H, respectively. These reactions indicate that oxidation by MnO_4^- is accompanied by the production of MnO_2 solid, carbon dioxide (CO_2), hydrogen (H^+) and chloride(Cl^-).

In groundwater, the rapid rate of VOC oxidation by MnO_4^- increases the concentration gradient between the DNAPL:water interface and the bulk groundwater, which increases the rate of remediation through enhanced dissolution of the DNAPL (Schnarr *et al.*, 1998). The effectiveness of a MnO_4^- flush in removing DNAPL source mass varies as a function of a number of factors including the design of the oxidant delivery system (including duration of operation, oxidant delivery efficiency and injected oxidant concentration) and the initial DNAPL distribution. While the design approach may be modified to optimize the remediation process, the limited understanding of the impact of DNAPL distribution on technology performance suggests that this factor is of critical importance. For example, in the portions of a DNAPL source zone containing only residual, nearly all DNAPL mass can be destroyed in situ with the expectation of a comparable level of mass flux reduction (Thomson *et al.*, 2000). In comparison, for source zone regions with large DNAPL accumulations that fill a significant fraction of the available pore volume, it is likely that a lower level of mass removal and mass flux reduction may be achieved (Thomson *et al.*, 2000). The differences in mass flux removal are primarily due to the difference in the DNAPL:water interfacial surface area. For a comparable DNAPL volume, the total surface area of a DNAPL pool is much smaller than that of residual DNAPL; therefore, the rate of DNAPL mass removal is lower.

During ISCO the rate of DNAPL removal, as inferred by the production of chloride, follows a typical progression as shown in Figure 2-2 (Schnarr *et al.*, 1998; Hood and Thomson, 2000; MacKinnon and Thomson, 2002; Lee *et al.*, 2003). At early times, as the oxidant migrates into zones containing readily accessible DNAPL, the oxidation reaction results in high chloride concentrations (corresponding to high rates of DNAPL removal); however, the chloride concentrations tend to decrease over time as the readily accessible DNAPL mass is depleted. Eventually, the presence of the MnO_4^- has only a minimal impact on the rate of DNAPL removal once diffusion limitations on the delivery of the oxidant to the DNAPL control the rate of removal (Hood and Thomson, 2000). As the rate of DNAPL removal by ISCO decreases, the unit cost of removing DNAPL mass using this technology increases proportionately.

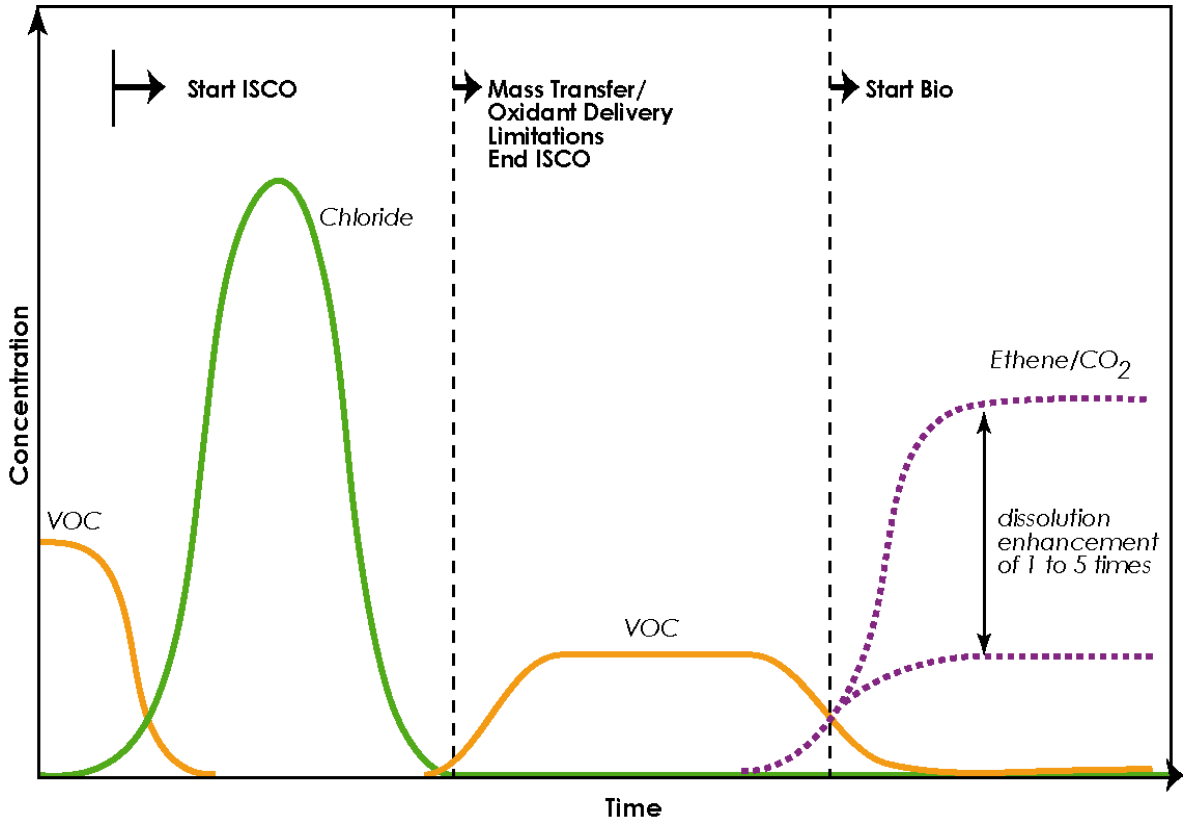


Figure 2-2. Typical Progression of Contaminant Mass Removal During ISCO.

2.1.2 Switching from In Situ Chemical Oxidation to Enhanced In Situ Bioremediation

After the initial phase of ISCO in which the rate of DNAPL removal is very high, typically the rate decreases to a point at which the presence of the oxidant in the treatment zone does not increase the rate of DNAPL removal over that rate expected by flushing the source area with only groundwater. Accordingly, the application of a lower cost remediation technology is appropriate. The switchover point may be determined based on either the total rate of DNAPL removal (as determined by either measurement of the parent compound or chloride, an oxidation reaction product) or by comparison to the background chloride concentration. Using the rate of DNAPL removal prior to oxidation treatment as a baseline condition, the corresponding rate during ISCO treatment may be calculated by stoichiometric conversion of the rate of chloride production. Since the point of source remediation is to increase the rate of source mass removal above that occurring under intrinsic conditions, the minimum condition for continuing the application of ISCO is that the rate of DNAPL removal during treatment (measured by chloride production) exceeds the rate of DNAPL removal prior to treatment. However, the presence of a high background concentration of chloride can complicate determination of the DNAPL mass removal rates. The removal rate of DNAPL must be sufficiently high that the concentration of

chloride produced by the oxidation reaction is distinguishable from the variability in the background chloride concentration.

While MnO_4^- treatment may be an effective means of treating dissolved phase TCE and PCE, continuous addition of a low MnO_4^- concentration to the source area would be required indefinitely, an unfavorable situation given the cost of MnO_4^- and the potential for adverse impacts to groundwater quality by dissolved manganese. Under conditions where the minimum criteria for the effectiveness of ISCO are not achieved (i.e., no measurable mass transfer enhancement) a complementary remediation technology with a lower operating cost may be more appropriately applied.

2.1.3 Enhanced In Situ Bioremediation

Of particular interest are biological remediation approaches for chlorinated solvent contamination that use anaerobic degradation processes. Aerobic processes require the addition of co-substrates and are often limited in the concentrations of VOC that can be treated because of the solubility constraints of oxygen in groundwater and possible toxicity effects of intermediate compounds on the microorganisms. Anaerobic reductive dechlorination does not share these limitations and is more commonly used to degrade chlorinated solvents. Under anaerobic conditions, reductive dechlorination is a well understood degradation mechanism for PCE and the lesser chlorinated alkenes that may result in complete dechlorination to ethene and ethane. Reductive dechlorination involves the stepwise replacement of individual chlorine atoms with hydrogen atoms (Figure 2-3) where the chlorinated ethene acts as an electron acceptor while an electron donor is required to provide energy for this process (McCarty, 1994). Hydrogen is generally considered the direct electron donor for reductive dechlorination, and is typically produced from the anaerobic oxidation of other carbon substrates, such as organic acids or alcohols (Maymo-Gatell *et al.*, 1997).

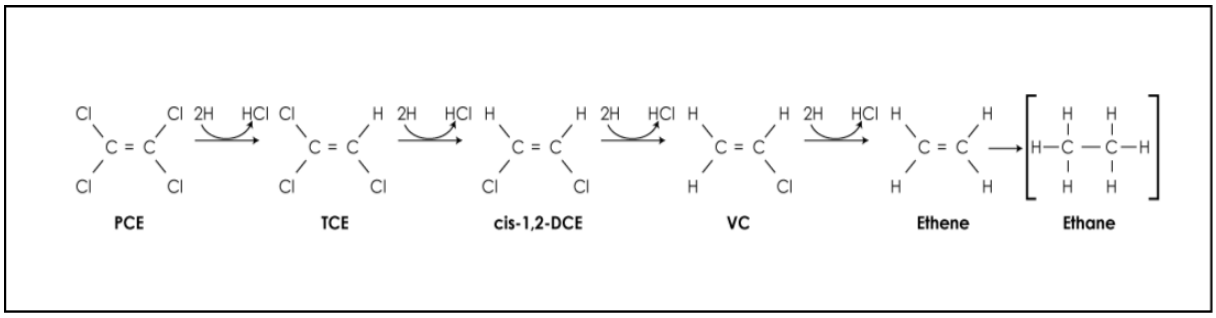


Figure 2-3. Reductive Dechlorination Reaction Sequence for Chlorinated Ethenes.

Recent research and field observations at several sites have demonstrated that both PCE and TCE may be reductively dechlorinated to ethene by indigenous microorganisms in groundwater (Ellis *et al.*, 2000; Major et al., 1995 and 2002; DiStefano *et al.*, 1991). Several indigenous bacteria have been identified, including *Dehalococcoides ethenogenes* (DHE), which directly use VOC such as PCE and TCE as terminal electron acceptors (i.e., respiration). While dehalorespiring bacteria have been identified at a number of sites, the relatively common occurrence of PCE or TCE dechlorination stalling at the formation of cis-1,2-dichloroethene (cDCE) and vinyl chloride (VC; Hendrickson *et al.*, 2001), suggests that these microorganisms are not ubiquitous in groundwater systems. A number of field and laboratory studies examining the use of several enriched indigenous microbial consortia containing these dehalorespiring bacteria have demonstrated that the activity of the dechlorinating microorganisms was not inhibited at high chlorinated ethene concentrations (Table 2-1). These results suggest that some dehalorespiring microorganisms are tolerant to high concentrations of chlorinated solvents and can be active in close proximity to DNAPL. Given sufficient microbial activity adjacent to the DNAPL, the dechlorination reaction may be able to significantly accelerate mass transfer from the DNAPL free phase surface and enhance the dissolution of the DNAPL phase.

At field sites where the background geochemistry is generally conducive to reductive dechlorination, several engineering approaches are now feasible that may significantly increase the applicability and effectiveness of bioremediation. The process of biostimulation involves the introduction of a suitable electron donor to increase the activity of indigenous microorganisms and promote complete dechlorination to ethene. However, if the appropriate dehalorespiring microorganisms are not present, the increase in activity may simply result in rapid degradation of the parent VOC and the accumulation of daughter products (typically either cDCE or VC). Accordingly, bioaugmenting the aquifer with a microbial consortium containing *Dehalococcoides*–like bacteria with the ability to completely dechlorinate chloroethenes in the presence of electron donors is required. A summary of various sites where biostimulation and bioaugmentation have been demonstrated at field scale is presented in Table 2-2. While only a few field demonstrations of bioaugmentation have been reported, each was successful at

Notes:

stimulating biodegradation of the target compound(s) to non-toxic end products such as ethene and CO_2.

Table 2-1. Reductive Dechlorination of High Concentrations of PCE and TCE.

VOC	*Scale*	*Source*	*Summary of Results*
PCE and TCE	Laboratory (batch and column)	Yang and McCarty (2000)	Anaerobic dehalogenation of PCE occurred at the solubility limit (>0.9 mM). TCE was dehalogenated at concentrations of up to 2.26 mM. Pentanol was used as the electron donor for dehalogentation by the Victoria TX culture. In the presence of DNAPL, the dominant product was 1, 2-*cis*- DCE Mass balances indicated that DNAPL dissolution was enhanced by a factor of ~five.
TCE	Field	Major et al. (1994)	Complete dechlorination of TCE to ethene occurred at a field site where TCE was at 80% of its aqueous solubility.
TCE	Laboratory (batch)	Sleep et al. (2006)	A microbial consortium (KB-1) isolated by GeoSyntec and the University of Toronto has been shown to dechlorinate TCE and 1,2-*cis* -DCE at concentrations as high as 100 mg/L. Methanol was used as an electron donor.
TCE	Laboratory (batch)	General Electric	Two anaerobic cultures were demonstrated to degrade TCE to ethene at TCE concentrations of 100 and 160 mg/L (corresponding to 8% and 13% of TCE's solubility limit). Lactate and methanol were used as electron donors.
PCE	Laboratory (batch)	**DiStefano et al. (1991)**	An anaerobic bacterial culture enriched from natural sources by researchers at Cornell has been shown to completely dehalogenate PCE to ethene at concentrations as high as 55 mg/L . The electron donor was methanol.
PCE	Laboratory (column)	Isalou et al. (1998)	Column experiments with PCE at 115 mg/L (0.07 mM) resulted in complete conversion to ethene. Anaerobic digestor sludge was used as an inoculum. The electron donor was methanol.

Notes
DNAPL - Dense Non-aqueous Phase Liquid
TCE - Trichloroethene
PCE - Tetrachloroethene

Table 2-2. Summary of Bioaugmentation Field-Scale Demonstrations

VOC	*Location*	*Source*	*Summary of Results*
TCE	Dover AFB, Delaware	Ellis et al. (2000)	Aquifer was bioaugmented with the Pinellas culture. Complete transformation of TCE to ethene was only observed after bioaugmentation. Lactate was used as an electron donor.
TCE	Pennsauken, New Jersey	Steffan et al. (1999)	Aquifer was bioaugmented with an aerobic TCE degrading culture (Burkholderia cepacia ENV435). TCE degradation was observed within several days. Cell densities remained high during the 30 day study.
TCE	Flemington, New Jersey	Walsh et al. (2000)	An aerobic TCE degrading culture (Burkholderia cepacia ENV435) was inoculated into pneumatically induced fractures where TCE concentrations as high as 30 mg/L were measured. TCE transformation to CO2 was observed.
TCE	Edison, New Jersey	Envirogen	An aerobic TCE degrading culture (Burkholderia cepacia ENV435) was inoculated into a sandy aquifer to treat TCE in the vadose zone. TCE (<1 mg/L) degradation was observed
TCE	Aerojet Superfund Site, California	GeoSyntec/ Aerojet	GeoSyntec Consultants and Aerojet initiated a bioaugmentation pilot test to enhance anaerobic in situ TCE and perchlorate remediation (on-going). The nutrients/electron donors include acetate and lactate.
PCE	Kelly AFB, Texas	Major et al. (2002)	Bioaugmentation of the test plot amended with methanol and acetate resulted in complete transformation of PCE to ethene after a lag period of ~70 days.
PCE	Bachman Road Residential Wells	Lendvay et al. (2003)	Bioaugmentation of test plot using a Dehalococcoides innoculum enriched from the same aquifer resulted in complete transformation of PCE to ethene in 6 weeks as compared to biostimulation where complete dechlorination was only observed after a 3 month lag period.
Carbon Tetrachloride	Schoolcraft, Michigan	Dybas et al. (1999)	Bioaugmentation of the aquifer with Pseudomonas stutzeri KC resulted in the biodegradation of carbon tetrachloride to carbon dioxide without production of chloroform. Acetate amendments and pH modifications were used to sustain microbial activity.

Note

* KB-1 is a natural stable consortium isolated by GeoSyntec and the University of Toronto

TCE - Trichloroethene

PCE - Tetrachloroethene

Field evidence exists to suggest that microbial populations can exist close to DNAPLs and enhance dissolution rates (Major *et al.*, 1995). As discussed earlier (Section 2.1) there is a growing body of laboratory evidence that suggests microbial populations can degrade high concentrations of PCE and TCE). These studies involved column and batch tests where dechlorinating cultures were exposed to saturated concentration of chlorinated solvents. In recent column studies, a 16-fold increase in PCE removal from a DNAPL was achieved through source zone bioremediation (Cope and Hughes, 2001). Yang and McCarty (2000) showed that PCE degrading microorganisms could completely dechlorinate PCE at concentrations up to the PCE solubility limit. The dissolution rate of the PCE DNAPL under these conditions was enhanced by 10 to 14 times over baseline conditions, suggesting that enhanced bioremediation of source zones is a feasible technology. Field tests specifically designed to monitor biologically mediated enhanced dissolution of a DNAPL are underway. If enhanced dissolution as a result of

bioremediation is possible following ISCO, ISB can be used as either a primary or a secondary source removal technology. As discussed earlier, if complete dechlorination is possible either via biostimulation alone or biostimulation with bioaugmentation but does not yield enhanced dissolution, the ISB technology can be used as a source containment technology.

2.2 Potential Impacts of In Situ Chemical Oxidation on Enhanced In Situ Bioremediation

The addition of MnO_4^- into groundwater can potentially result in both direct and indirect impacts on the subsequent application of ISB. Since oxidants like MnO_4^- have disinfection properties and impose an oxidizing redox potential in groundwater, the presence of residual MnO_4^- in the target treatment zone will directly impact ISB by inhibiting reductive dechlorination. In addition, if the electron donor used to stimulate biodegradation is reactive with MnO_4^- (e.g., ethanol) residual MnO_4^- in the treatment zone may preferentially react with the donor, requiring the addition of excess donor over that typically required for enhanced bioremediation. Through disinfection, ISCO will lower the indigenous microbial population present in the groundwater system. If insufficient biomass capable of supporting reductive dechlorination processes is present in the treatment zone following ISCO, longer periods of electron donor addition will be required to: 1) create reducing redox conditions in the target zone suitable for reductive dechlorination, and 2) increase the amount of biomass present in groundwater to a level capable of supporting a significant rate of reductive dechlorination of the target VOC. However, the removal of the indigenous biomass may enhance subsequent bioaugmentation of the treatment zone by decreasing the competition for electron donors, resulting in environmental conditions that favor the activity of the dechlorinating microorganisms over other species (e.g., methanogens, sulfate reducers).

In addition to directly impacting the biomass present in the treatment zone and reacting with electron donors, the deposition of manganese oxides (e.g., MnO_2, the dominant form of reduced manganese) in the treatment zone may have indirect impacts on the performance of enhanced bioremediation processes following ISCO. The dissolution of manganese oxides can result in a pH increase, potentially inhibiting microbial activity. Since these precipitates are themselves oxidants, electron donors added into the treatment zone may be consumed through either abiotic and/or biotic redox reactions mediated by the manganese oxides, resulting in an additional electron donor demand above that exerted by both organic (e.g., the target VOC) and inorganic (e.g., sulfate) solutes. Further, little is known about the impact of manganese oxides on reductive dechlorination processes, which may be intrinsically inhibited as a result of redox poising by the solid phase.

2.3 Previous Testing of the Technology

To date, a limited number of laboratory investigations have evaluated the impacts of ISCO using MnO_4^- on microbial populations and dechlorinating activity. As an oxidizing agent, contact with MnO_4^- will adversely impact microorganisms present in groundwater, although complete sterilization of a heterogeneous groundwater environment is generally considered unlikely to occur. In a study evaluating the impact of MnO_4^- addition on indigenous microorganisms, reductions in the populations of aerobic and anaerobic heterotrophs, nitrate, nitrite, and sulfate reducers, and methanogens following treatment ranged from 47% to 99.95% (Klens *et al.*, 2001). Replicate samples collected six months after treatment suggested that the population of heterotrophic aerobic microorganisms rebounded although enumeration of anaerobic heterotrophic microorganisms indicated that minimal regrowth of these microorganisms had occurred. At least one microcosm study of sequential ISCO and bioremediation (Rowland *et al.*, 2001) suggests that ISCO does not intrinsically inhibit the dechlorinating activity of the microbial population.

Several investigators have evaluated the impact of ISCO using MnO_4^- on microbial populations and dechlorinating activity after the completion of a field application of ISCO. Azadpour-Keeley *et al.* (2004) reported on microbial sampling conducted to evaluate the effects of ISCO testing at Launch Complex 34 in Cape Canaveral, Florida. Soil samples were collected from five locations within the test plot one month prior to and one, six and twelve months following the ISCO application. Phospholipid fatty acid (PLFA) analysis was used to evaluate microbial biomass in the test plot at these time points, and showed that biomass had increased markedly from pre-ISCO levels at one month post-ISCO, but then returned to pre-ISCO levels over the remainder of the monitoring period. Based upon the profile of the fatty acids which increased in concentration, the authors characterized the increase as largely due to *Proteobacteria*, and that other bacteria classifications were largely unaffected.

In a second study, Macbeth *et al.* (2005) used microbial community profiling and quantitative polymerase chain reaction (QPCR) testing to track *Dehalococcoides*-like species prior to and one year following MnO_4^- injection at the Savage Municipal Water Supply Well Site in Milford, New Hampshire. The data collected showed that the MnO_4^- treatment resulted in decreases in both biomass and diversity, but that these had partially recovered one year after residual MnO_4^- concentrations had decreased. *Dehalococcoides* was found to be present at low levels in the treatment area post-ISCO.

In summary, there are a number of laboratory and field studies which suggest that application of ISCO may have no long term impacts upon a follow-on ISB application. However, while these studies have shown that biomass returns post-ISCO, they have not clearly demonstrated a return

of microbial activity resulting in dechlorination of chlorinated VOC. Clearly additional work is necessary to fully evaluate the effectiveness of the sequential ISCO-ISB technology.

2.4 Factors Affecting Cost and Performance

It is anticipated that a number of factors will influence the cost of the sequential application of ISCO and bioaugmentation at full scale field sites. Primary factor affecting the cost of the technology include the duration of remediation, which is a function of the performance of the remediation technologies, the DNAPL distribution, and the hydrogeologic characteristics of the treatment zone. In addition, the availability of useful infrastructure (e.g., storage buildings, water treatment/disposal facilities, services) can significantly impact technology cost. The DNAPL distribution is a significant cost factor since it impacts mass transfer rates and the volume of aquifer requiring treatment. ISCO and ISB systems rely upon the following: i) delivery of amendments (e.g., oxidant, electron donor, nutrients, biomass) through injection wells to promote contaminant degradation; ii) volume of the aquifer defined by the horizontal and vertical extent of the DNAPL will control the amendment flow rate and the size of the amendment dosing system; and iii) number of wells required to circulate the amendments through the treatment zone. In a sequential application of ISCO and ISB significant cost savings can be realized as much of the same infrastructure can be used for both technologies, including injection, extraction and monitoring wells, piping and electrical infrastructure; computer control systems and remote dial-in software; and process instrumentation.

While it is anticipated that a substantial enhancement in the removal rate of the DNAPL will be achieved during the demonstration, the rate of mass removal may still be small in comparison to the mass of DNAPL initially present, suggesting that at some sites the large mass of DNAPL present may limit the effectiveness of the technology. Because ISB requires the establishment of anaerobic and reducing conditions in the treatment zone, the ability of the background redox conditions to intrinsically support reductive dechlorination will also substantially improve the performance of this technology. Geological heterogeneity will strongly influence the performance of bioaugmentation by limiting the delivery of the amendments to the microorganisms adjacent to the DNAPL. In particular, the delivery of a sufficient concentration of electron donor to support the microbial activity immediately adjacent to the DNAPL: water interface may limit the maximum concentration of the target contaminant that can be degraded. This limitation will depend on the type and concentration of the electron donor added into the source zone, the utilization rate by the microorganism, and the design of the nutrient delivery system.

Another limitation of these technologies will be the cost of DNAPL source zone characterization. At many sites, it will not be feasible to characterize the DNAPL distribution; instead, the design

of the treatment system should be sufficiently large as to encompass the entire DNAPL source zone. This may increase the annual O&M costs of remediation; however, this increase is offset by the reduction in the cost of site characterization.

2.5 Advantages and Limitations of the Technology

The main advantages of the technology are:

- Enhancing the dissolution rate of a DNAPL will decrease clean-up times;
- Mass will be destroyed and not simply transferred to another medium;
- Expansion of a treatment area to include uncertainties related to the exact DNAPL distribution are unlikely to be difficult or significantly increase total cost;
- The enhanced bioremediation process provides a long-term, lower cost, polishing of VOC remaining after ISCO; and
- Lower expected capital and O&M costs than alternative technologies (see Table 2-1).

The main limitations of the technology are:

- The need to understand and identify the source extent and mass to minimize the volume of the zone requiring treatment (i.e., minimize cost);
- Inaccessible DNAPL mass;
- The cost of the amendments required (i.e., MnO_4^-, electron donor);
- Weak advective-dispersive solute transport processes which may limit the delivery of treatment reagents (e.g. MnO_4^-, electron donors) to the DNAPL;
- The occurrence of geochemical conditions (e.g., high sulfate) that may be inhibitory to biodegradation; and
- The presence of co-contaminants that may be inhibitory to biodegradation (e.g., chloroform, hydrogen sulfide).

In addition, there is the potential that ISCO will adversely impact the subsequent implementation of ISB. Adverse impacts could occur: 1) during ISCO through disinfection of the treatment zone resulting in a reduction in the dechlorinating microbial populations required for ISB; and/or 2) as a result of long-term changes in the groundwater geochemistry caused by oxidation of the soil and deposition of MnO_2. The latter consideration is particularly important given the low solubility of manganese under typical geochemical conditions and its expected long-term persistence in the treatment zone.

3.0 Demonstration Design

3.1 Performance Objectives

The performance objectives that will be used to meet the project objectives described in Section 1.2 and to evaluate the performance and cost of the demonstration are provided in Table 3-1.

Table 3-1. Performance Objectives.

Type of Performance Objective	*Primary Performance Criteria*	*Expected Performance*	*Actual Performance (Objective Met?)*
Qualitative	Activity of Microbial Community	Microbial activity present prior to the addition of nutrients and/or bioaugmentation; community activity increased after these additions	Microbial activity present prior to the addition of nutrients and/or bioaugmentation; community activity increased after these additions
	Increase Extent of Dehalogenation	Complete dehalogenation to ethene	Complete dehalogenation to ethene
	VOC Concentration Reduction	Some VOC concentration reduced in areas of high microbial activity	Some VOC concentration reduced in areas of high microbial activity
Quantitative	Increase in Microbial Biomass	Increase in microbial biomass above the base case treatment[1]	Increase in microbial biomass above the base case treatment[1]
	Increased Mass Flux from DNAPL During Treatment > after amendment with electron donor	Increase in mass flux above the base case treatment[1]	Increase in mass flux above the base case treatment[1]
	Reduce DNAPL Mass	Reduction in DNAPL present at start of base case treatment[1]	Reduction in DNAPL present at start of base case treatment[1]

Notes:
[1] Base case treatment - operation of pilot system post-oxidation without addition of electron donor/nutrients or bioaugmentation

3.2 Site Selection

As described previously (Geosyntec, 2002), efforts were made to identify field sites at which the demonstration could be conducted. In addition to sites identified by Geosyntec, a request for candidate sites was distributed to USACE Innovative Technology Advocates by the Project COR (Lance Hansen, USACE), resulting in a preliminary list consisting of six candidate sites. Monitoring data provided by the site contacts was reviewed, screened for the presence of target compounds (TCE) and dechlorinated daughter products (e.g., cDCE and VC), and assessed against a number of site selection criteria including:

- The presence or suspected presence of DNAPL;
- Background geochemistry favorable to reductive dechlorination;
- The feasibility of securing access to the area above the source zone;
- A shallow depth to groundwater to facilitate the installation of boreholes and monitoring wells; and
- DNAPL present in a region of relatively high permeability porous media;
- The extent to which the geologic stratigraphy was delineated.

The results of the site screening process are presented in Table 3-2. Based on review of the characterization data for each site, two sites (East Gate Disposal Yard site, Fort Lewis, Washington and LC-34, Cape Canaveral Air Force Station, Florida) potentially satisfied the selection criteria identified for this project. Both facilities were candidate sites based on shallow depth, high permeability, the existing evidence of incomplete dechlorination to *cDCE*, and the previous acceptance of technology demonstration projects at the facility. However, the point of contact for the Fort Lewis facility indicated that an ongoing technology demonstration at the Disposal Yard site precluded the use of the site for this demonstration (personal communication, Kira Lynch, USACE). LC-34 was available for a technology demonstration and, in addition, had previously been the site of an oxidation demonstration, thereby reducing the need to repeat that component of the demonstration project. A bioaugmentation demonstration had also been recently completed in a nearby area, providing a convenient basis for comparing the sequential technology to a standalone demonstration of bioremediation. Accordingly, based on the site selection criteria, LC-34 was selected as the demonstration site.

Table 3-2. Summary of Site Selection Evaluation.

Criteria	*Site ID*					
	1	*2*	*3*	*4*	*5*	*6*
Presence of TCE or PCE DNAPL	✓	✓	✓	✓	✓	?
Defined source area (extent/mass0	✓	x	x	x	✓	x
Incomplete dechlorination	✓	✓	✓	✓	✓	✓
Shallow groundwater	✓	✓	✓	✓	✓	✓
Source area well instrumented	✓	✓	✓	✓	✓	✓
Accessible power/infrastructure	✓	✓	✓	✓	✓	?
Suitable *K* and *i*	✓	x	x	x	✓	x
Low sulfate/chloride concentrations	x	?	?	?	✓	?
No/low chloroform/1,1,1-TCS concentrations	?	x	✓	✓	?	x
Enlightened regulatory environment	✓	?	?	?	✓	?

1- LC-24, Cape Canaveral Air Force Station, Florida:	Max TCE 630 mg/L; chloride and sulfate concentrations of 1730 mg/L and 535 mg/L (LSU), 582 mg/L and 1380 mg/L (MFGU) and mg/L and 483 mg/L (USU), respectively, Sand and silty sand aquifer with K ranging from 4 to 5.1 ft/day (USU), 1.4 to .4 ft/day in (MGFU) and 1.3 to 2.3 ft/day (LSU).
2-Bulding 348, Red River Army Depot, Texas:	TCE concentrations up to 623 mg/L in degreaser pit and 13 mg/L in gw – volume/mass of source not defined. Metals and BTEX contamination present; 1,1,1-TCA up to 440 µg/L. Clay and weathered shale to ~40 ft bgs and then shale (K= 7.7 x 10^{-6} cm/s)
3-Building 433, Red River Army Deport, Texas:	Max TCE in gw 336 mg/L, max TCE in soils 7490 µg/kg. Fractured rock and weathered clay system.
4-WWT Area, Red River Army Deport, Texas:	Max TCE in gw 3980 µg/L, max TCE in soils 40,400 mg/kg; little gw chem. Data available beyond VOC and metals. Clay, weathered shale, and fractured bedrock.
5-East Gate Disposal Yard, Fort Lewis, Washington	Max TCE in soil 3000 mg/kg; TPH present up to 13,500 mg/kg in soil (Same location as TCE hot spot); 1,1,1-TCA used as degreaser detected in available soils data. Glacial sediments (glacial outwash sands and gravels and tills).
6-Niagara Falls IAP-ARS Site 10 Fire Pit, Niagara Falls New York	Max TCE 5280 ppb, max cis-1,2-DCE 24,179 ppb at overburden/bedrock interface; metals (Zn, Cr, B and Pb) and benzene present VOC (chlorobenzene, chloroform, 1,4-dichlorobenzene, 1,1-DCE, tetrachloroethane, 1,1,2-TCA, acetone and chloromethane up to 36 total); groundwater extraction system in place to treat overburden. Most mass in overburden and a bedrock contact.

3.3 Test Site Description

The location of LC-34 is provided in Figure 3-1. Due to the relatively simple geology at the site and the known presence of DNAPL, a number of research-oriented technology demonstrations have been conducted at LC-34, including performance evaluations of ISCO using potassium MnO_4^-, six phase heating (SPH), steam (Battelle, 2001a), bioaugmentation (Battelle, 2004b) and emulsified zero-valent iron (Battelle, 2004a). The locations of these demonstrations and the estimated extent of their influence are presented in Figure 3-2.

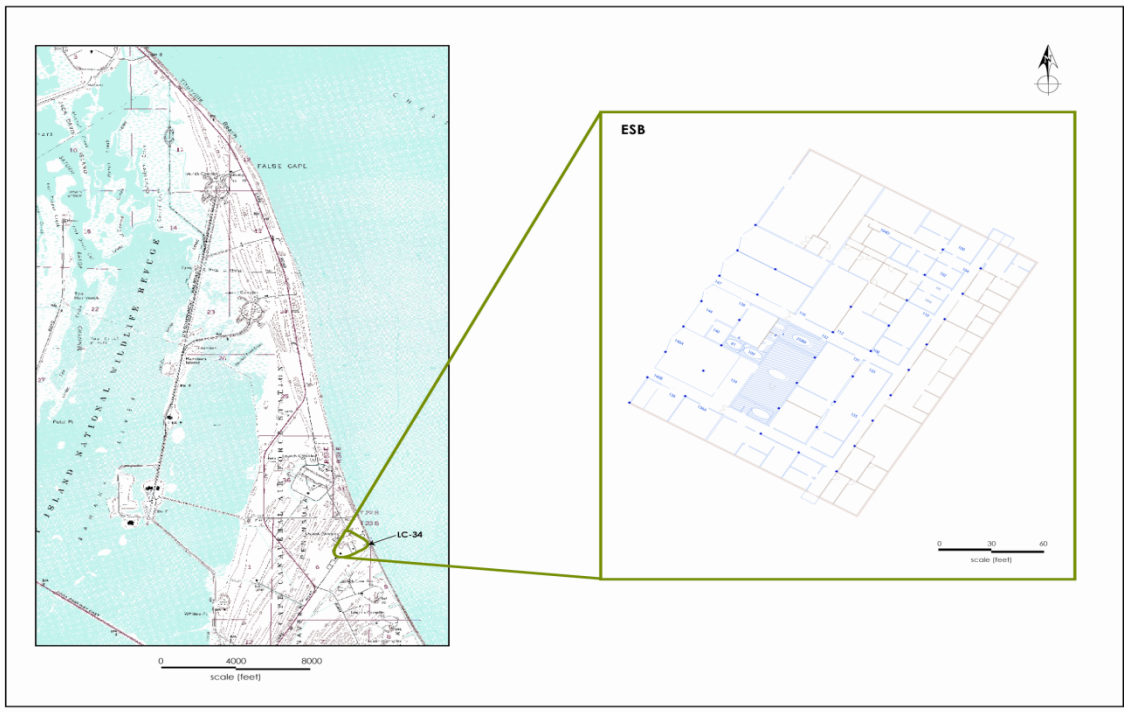


Figure 3-1. Location of Cape Canaveral Air Force Base, Cape Canaveral, Florida.

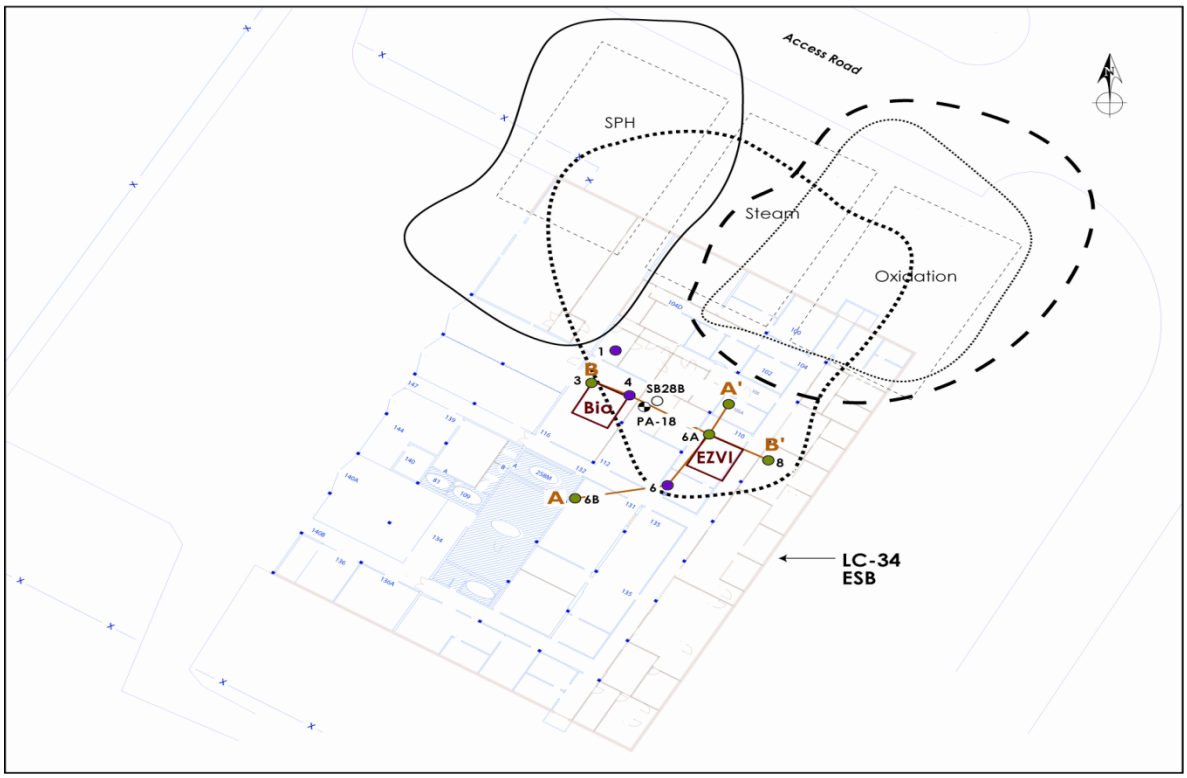


Figure 3-2. Site Plan and Technology Demonstration Locations at LC-34.

Historical records from LC-34, which was used as a launch facility from 1960-1968, suggest that rocket engines were cleaned on the launch pad with chlorinated organic solvents such as TCE. Other rocket parts were cleaned on outdoor racks along the west side of and inside the ESB. During cleaning operations the solvents evaporated, infiltrated directly into the subsurface, or migrated as runoff into drainage pits. The presence of DNAPL below the ESB was documented by Eddy-Dilek *et al.* (1998) and Battelle (1999). DNAPL source characterization efforts suggest that 20,600 kilograms (kg) (Battelle, 1999) to 40,000 kg (Eddy-Dilek *et al.*, 1998) of TCE DNAPL are present in the subsurface near the ESB.

In 1999, a demonstration of ISCO using MnO_4^- at LC-34 was completed in a 75 ft x 50 ft test plot adjacent to ESB. During the demonstration, 842,985 gallons (gal) of a potassium MnO_4^- solution (typical concentration of 1.4% to 2%) was injected into the ISCO test plot through a drive-point injection system. The total mass of MnO_4^- used during the demonstration was 68,479 kg.

3.3.1 Environmental Setting and Geology

Hydrogeological conditions at LC-34 are highly favorable to the implementation of a recirculation-based remediation technology. The aquifer consists of relatively homogeneous sand and silty sands, and is easily instrumented using low-cost, direct-push drilling technologies (i.e., GeoProbe). A surficial aquifer and a semi-confined aquifer beneath a clay unit comprise the major water bearing units at LC-34. The surficial aquifer extends from the water table to approximately 45 ft below ground surface (bgs). The clay confining unit ranges in thickness from 1 to 3 ft. The surficial aquifer is sub-divided into the upper sand unit (USU), the middle fine-grained unit (MFGU), and the lower sand unit (LSU) (Eddy-Dilek *et al.*, 1998). The USU is composed of medium to coarse grained sand and crushed shells and extends from ground surface to approximately 18 to 25 ft bgs. The MFGU, which varies in thickness from about 4 to 14 feet, is composed of gray, fine-grained silty/clayey sand and generally contains finer-grained sediment than the remainder of the aquifer unit. The MFGU is thicker to the north of the ESB and appears to thin towards the south and west of the ESB. The LSU, the deepest subunit of the surficial aquifer, consists of gray fine to medium-sized sand and shell fragments. In addition, the LSU contains some isolated fine-grained lenses of silt and/or clay. The thickness of the underlying confining unit is unknown since boreholes are typically completed at the top of clay unit to prevent drilling-induced migration from the LSU into the confined aquifer. The confining unit may act as a barrier to DNAPL migration into the confined aquifer.

The Atlantic Ocean is located immediately to the east of LC-34. To determine the effects of tidal influences on the groundwater system, water levels were monitored in 12 piezometers over a 50-hour period during RCRA Facility Investigation (RFI) activities (G&E Engineering, Inc., 1996).

All the piezometers used in the study were screened in the surficial aquifer. No detectable effect from the tidal cycles were identified in the subject area. However, the Atlantic Ocean and the Banana River (west of LC-34) are sufficiently close to the Site and appear to act as hydraulic barriers or sinks, as groundwater likely flows toward these surface water bodies and discharges into them. Other hydrologic influences at LC-34 include features such as paving, constructed drainage ditches, and topographical relief. Permeable soils exist from the ground surface to the water table and drainage is excellent. Water infiltrates directly to the water table.

Only limited data was available to characterize background geochemistry at LC-34 (Battelle, 1999; CRA, 1999) prior to the sequential technology demonstration. As may be expected, the salinity of groundwater in the surficial units (USU, MFGU, and LSU) increases with depth with concentrations of total dissolved solids as high as 1,200 mg/L in the LSU (predominantly Na, K, Mg, Ca, Al, Cl, and total SO_4/S). Groundwater pH is near neutral (7.3-8.0) with an alkalinity of up to 360 mg/L (as $CaCO_3$). Although no direct measurements of oxidation-reduction potential

are available, the high concentrations of dissolved iron and manganese indicate that the groundwater redox potential is generally reducing.

Following the ISCO demonstration at LC-34, the residual MnO_4^- remaining in the test plot likely continued to slowly react with soil and/or residual TCE present in the subsurface while slowly migrating down-gradient of the test plot. MnO_4^- was not observed during a groundwater monitoring event (October 2002) conducted using monitoring wells located in and adjacent to the test plot, suggesting that the residual MnO_4^- was depleted, which was an essential step prior to initiating treatment via bioremediation.

A preliminary site investigation was conducted by Geosyntec in December 2002 to facilitate selection of locations for the ISCO pilot demonstration. Five boreholes were drilled within the ISCO PTA adjacent to the ESB to characterize the geology, and soil and groundwater chemistry. (PID). Soil samples from five boreholes were submitted for laboratory analysis of VOC. The presence of DNAPL was inferred based on PID readings exceeding 9,999 ppmv and concentrations of TCE in soil exceeding 10,800 mg/kg. A detailed summary of the preliminary Site investigation is provided in Appendix A.

3.3.2 Test Plot Microbial Characterization

Only limited data was available characterizing the microbial population at LC-34 prior to the sequential technology demonstration. Prior to the oxidation demonstration, Eddy-Dilek *et al.* (1998) analyzed a limited number of soil and groundwater samples collected from the vicinity of ESB (in and outside of the DNAPL source zone) using heterotrophic plate and acridine orange enumeration techniques. While the limited number of samples precluded a definitive comparison, Eddy-Dilek *et al.* (1998) reported that the plate and acridine orange direct counts of samples collected from outside the source zone were consistent with a normal range; however, the single source zone sample was below the reliably enumerated range, suggesting that the presence of DNAPL may inhibit microbial growth. There is some evidence available to suggest that *D. ethenogenes* (DHE) are present in groundwater at LC-34. In May 2001, Geosyntec submitted groundwater samples from two monitoring wells to Dupont Laboratories, Delaware, for analysis using molecular genetic techniques to detect the presence these dechlorinating microorganisms and determined that *Dehalococcoides* bacteria are present in both background (IW-24D) and plume (IW-15D) samples. Subsequent samples submitted to SiREM Labs (ON) indicated that *Dehalococcoides*-like bacteria were present in five of six groundwater samples collected from the source area.

Further characterization of the soil microbial community at LC-34 was carried out on samples from five soil cores collected in February 2003. Measurements of total phospholipid fatty acids (PLFA) (Microbial Insights, Rockford, Tennessee) indicated an average of 115 picomoles

PLFA/g dry weight, corresponding to an estimated cell density of 2.3 x 106 cells/gram of soil. Heterotrophic plate counts (GAP Enviromicrobial Services London, Ontario) were significantly lower with maximum values of 8700 colony forming units/ gram (CFU/g) and anaerobic plate counts with maximum values of 11,300 CFU/g. Most probable number analysis (GAP) indicated negligible concentrations of sulfate-reducing organisms while targeted PCR for the domain Archaea (SiREM, Guelph, Ontario) indicated the absence of DNA belonging to methanogens. Instead of sulfate reducers and methanogens the microbial community was dominated by members of the division Proteobacteria, specifically several *Pseudomonas* species, based on denaturing gradient gel electrophoresis (DGGE) analysis (SiREM). Furthermore targeted PCR indicated the presence of dechlorinating Dehalococcoides group organisms, with microcosm studies (SiREM) confirming the ability of the soil microorganisms to mediate complete dechlorination of TCE to ethene, suggesting that viable Dehalococcoides populations were present. A summary of these data is provided in Appendix M.

3.3.3 Contaminant Distribution in Pilot Test Area

Pre- and post-treatment soil sampling was performed by Battelle during the previous technology demonstrations (Battelle, 2001a). The results of post-treatment monitoring in the ISCO test plot indicate that there 844 kg of total TCE mass, including 637 kg of TCE DNAPL, remained in the LSU.

3.4 Pre-Demonstration Activities

Prior to initiating the demonstration, a number of pre-demonstration tasks were completed to collect essential data required to effectively implement this technology demonstration. As described in the following sections, these tasks include pre-design chemical and microbiological laboratory testing (Section 3.4.1), University of Toronto (UT) laboratory studies (Section 3.4.2), and preliminary site characterization (Section 3.4.3).

3.4.1 Pre-Design Treatability Studies

A series of pre-design treatability studies were performed to:

- Assess the effect of MnO_2 on the utilization of common electron donors by indigenous microorganisms;
- Determine if MnO_2 reacts via an abiotic pathway with common electron donors at significant rates;
- To evaluate the impact of MnO_4^- addition on enhanced biodegradation of TCE in groundwater; and
- Measure the natural oxidant demand of soil at the demonstration site

The design of these studies and the methods used are provided in Appendix B.

3.4.2 University of Toronto Column Studies

Column studies examining the sequential application of ISCO and ISB were completed at the UT. The design of these studies and the methods used are provided in Appendix C.

3.5 Testing and Evaluation Plan

3.5.1 Demonstration Installation and Start-Up

The treatment system includes injection and extraction wells, the above-ground treatment system, process instrumentation, and process controls. The locations of monitoring and recirculation wells are presented in Figure 3-3. The process flow diagram of the above-ground recirculation system is presented in Figure 3-4.

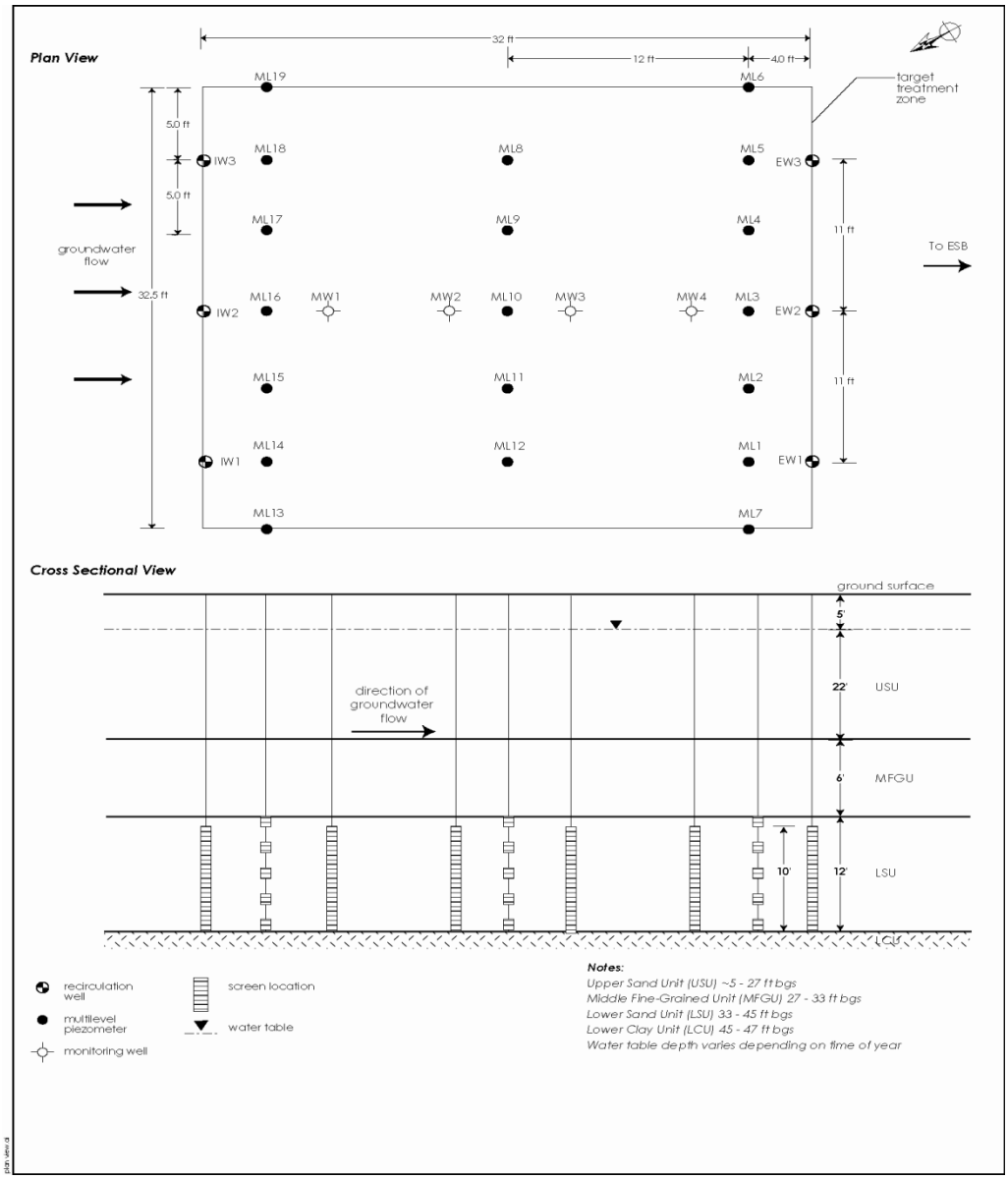
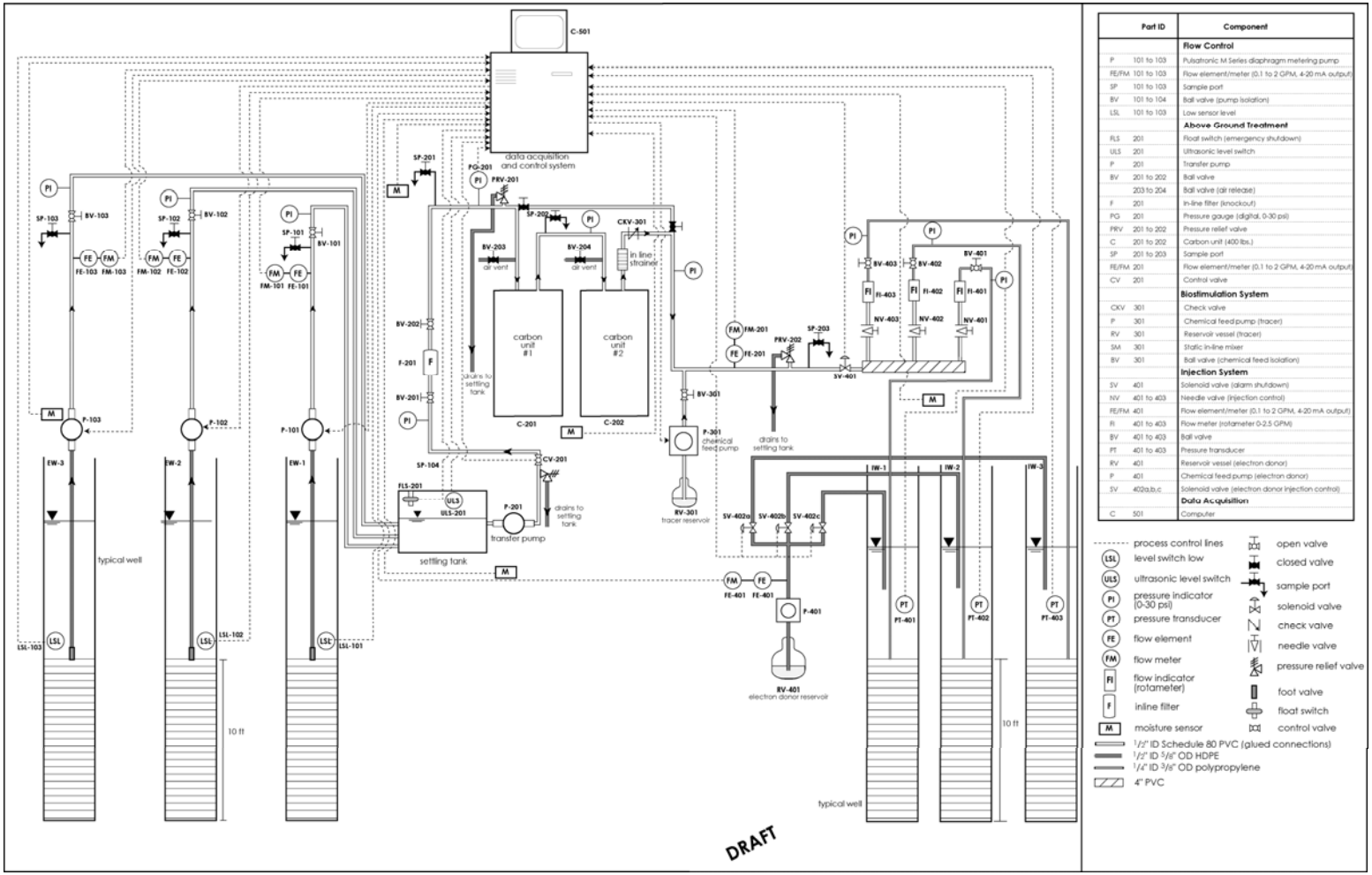


Figure 3-3. Instrumentation of PTA at LC-34.



Part ID		Component
		Flow Control
P	101 to 103	Pulsatronic M Series diaphragm metering pump
FE/FM	101 to 103	Flow element/meter (0.1 to 2 GPM, 4-20 mA output)
SP	101 to 103	Sample port
BV	101 to 104	Ball valve (pump isolation)
LSL	101 to 103	Low sensor level
		Above Ground Treatment
FLS	201	Float switch (emergency shutdown)
ULS	201	Ultrasonic level switch
P	201	Transfer pump
BV	201 to 202	Ball valve
	203 to 204	Ball valve (air release)
F	201	In-line filter (knockout)
PG	201	Pressure gauge (digital, 0-30 psi)
PRV	201 to 202	Pressure relief valve
C	201 to 202	Carbon unit (400 lbs.)
SP	201 to 203	Sample port
FE/FM	201	Flow element/meter (0.1 to 2 GPM, 4-20 mA output)
CV	201	Control valve
		Biostimulation System
CKV	301	Check valve
P	301	Chemical feed pump (tracer)
RV	301	Reservoir vessel (tracer)
SM	301	Static in-line mixer
BV	301	Ball valve (chemical feed isolation)
		Injection System
SV	401	Solenoid valve (alarm shutdown)
NV	401 to 403	Needle valve (injection control)
FE/FM	401	Flow element/meter (0.1 to 2 GPM, 4-20 mA output)
FI	401 to 403	Flow meter (rotameter 0-2.5 GPM)
BV	401 to 403	Ball valve
PT	401 to 403	Pressure transducer
RV	401	Reservoir vessel (electron donor)
P	401	Chemical feed pump (electron donor)
SV	402a,b,c	Solenoid valve (electron donor injection control)
		Data Acquisition
C	501	Computer

Figure 3-4. Process Flow Diagram.

Construction of the treatment system was initiated on 12 May 2003 and completed on 4 June 2003. Testing and modifications to the treatment system were completed between 6 June 2003 through 27 January 2004. Tracer testing of the recirculation system started 14 October 2003. During the tracer test, reinjected groundwater was amended for six days with a concentrated potassium bromide (KBr) to achieve an injected concentration of 75 mg/L (as Br^-). Programmable wastewater autosamplers were employed to collected groundwater samples from MW-1, MW-2, MW-3, and MW-4, initially at a frequency of two samples per day for MW-1 and -2, followed by a frequency of one sample per day for MW-2, -3, and -4. On Day 27 of the tracer test, the treatment system shut down due to a system malfunction. The tracer test was subsequently terminated and restarted on 01-March-2004. A detailed summary of the tracer test is provided in Appendix D.

Continuous groundwater recirculation began on 10 Jun 2002. Beginning on 8 March 2004, reinjected groundwater was amended with ethanol at a concentration equivalent to the stoichiometric demand exerted by all electron acceptors (including VOC). On 12-April-04, the PTA was bioaugmented by amending each of the three injection wells with 60 L of KB-1™.

Approximately one month after amending the recirculated groundwater with ethanol, indications of biofouling became present. Biofilm was accumulating on injection well screens and within the tubing and parts of the treatment system. In response to the indications of biofouling the following measures were implemented:

- Decreasing the duration of ethanol amendment, resulting in addition of ethanol in a concentrated (~10% v/v) daily pulse to inhibit microbial activity in the well screen
- Scrubbing, surging and purging each of the injection wells on a monthly basis to remove biofilm on screen and in the surrounding formation; and
- Amending the reinjected groundwater with sodium hypochlorite (time-weighted daily average of 0.1 mg/L) concurrently with ethanol amendment to inhibit microbial activity in the well.

3.5.2 Injection, Extraction, and Monitoring Well Construction

Borehole drilling and well installations were completed by Precision Drilling. The monitoring wells were completed using a direct-push (DPT) rig and the injection/extraction wells were completed using a Sonic drill rig. The boreholes were advanced to the target depth using a 3.5-inch (DPT) or 7-inch (Sonic) diameter casing with a disposable tip. All injection, and extraction wells were constructed with 10 ft screens completed at the bottom of the USU (44 ft bgs); monitoring wells were also completed with 10 ft screens. Monitoring well construction details and a typical well completion record are provided in Appendix E.

The injection and extraction wells were constructed from 2-inch inside diameter (ID) 304 stainless steel casing with flush-threaded joints. The well screens were constructed of 304 wire wrap screen with a 0.020 inch opening. All well casing and screens were certified clean by the manufacturer and delivered to the Site sealed in individual protective wrappings. To prevent entry of water into the well through joints, Buna-N/Nitrile™ O-rings were placed between casing sections. The bottoms of the screens were plugged with flush-threaded end caps. A filter pack consisting of washed and screened 6/20 silica sand was installed in the annulus between the well screen and the casing for all wells. The conventional monitoring wells were constructed in a similar manner as the injection and extraction wells, but with 0.010 inch slot size screens and 20/30 silica sand for the filter pack.

Wells were developed by purging 30 gallons (15 casing volumes) of water from each well using dedicated Waterra® tubing and foot valves. Each well was completed at surface with a steel, flush-mount protective casing set in concrete and equipped with dedicated Waterra® pump system consisting of a Delrin® foot-valve attached to stiff, $^{5}/_{8}$-inch outside diameter, high density, polyethylene tubing equal in length to the depth of the well. Following development, hydraulic testing was performed using slug tests.

Multilevel monitoring wells were installed using the same direct–push rig technique. Each multilevel was constructed of 1.5 inch outside diameter (OD) continuous multichannel tubing (CMT) with five 6-inch screened intervals spaced 3 ft apart. Sample locations are shown in Figure 3-5. Each sample interval was equipped with a dedicated Waterra® microflow pump system consisting of a stainless steel foot-valve attached to rigid 3/8-inch outside diameter HDPE tubing equal in length to the depth of the well.

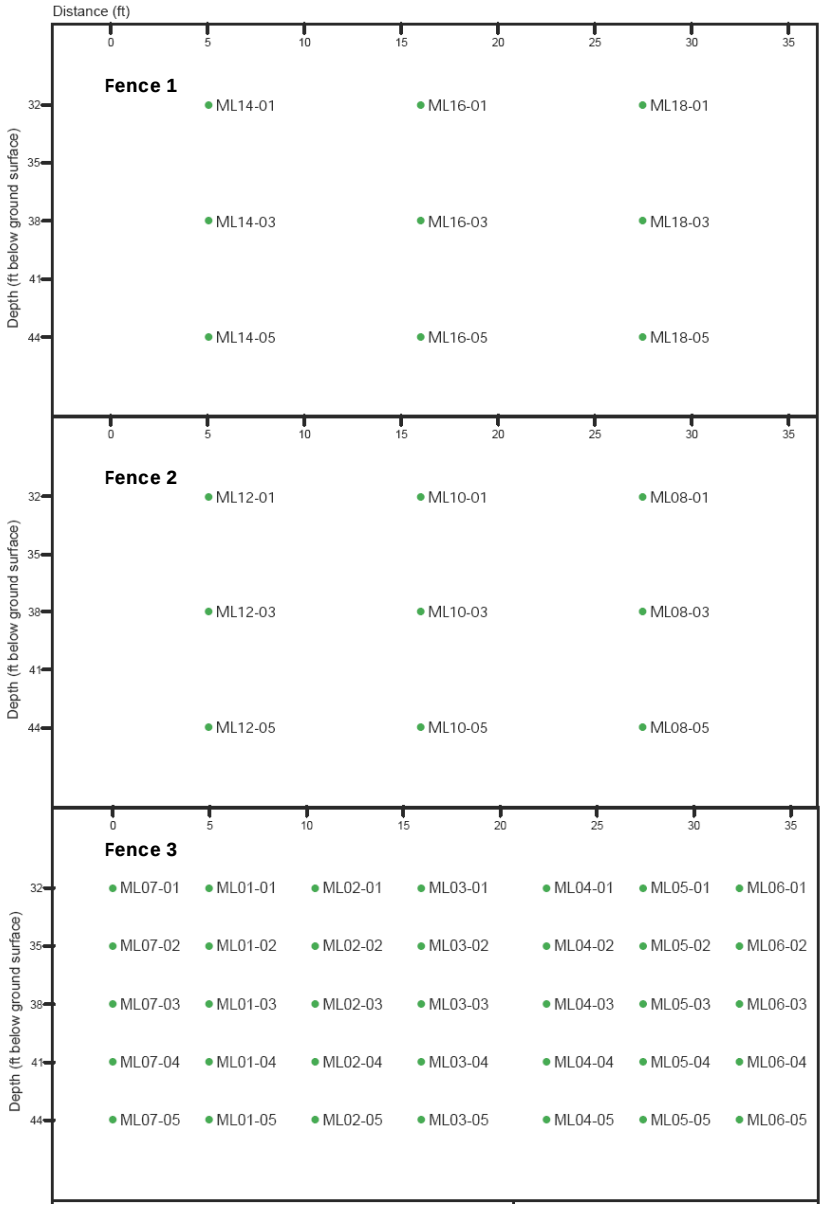


Figure 3-5. Locations of Multilevel Sampling Wells in Fences 1, 2, and 3.

3.5.3 Period of Operation

The system was operated between June 2003 and August 2004.

3.5.4 Amount/Treatment Rate of Material to be Treated

Detailed measurements of the mass of TCE in the demonstration plot were not performed. The volume of the demonstration plot was 12,500 ft^3. Based on the average bulk concentrations of TCE in soil samples (836 mg/kg) collected from the PTA during baseline characterization, the total mass of TCE is approximately 370 kg.

3.5.5 Residuals Handling

Spent granular activated carbon (GAC) from the treatment system was characterized and disposed of by the manufacturer. Solid waste (e.g., gloves, sampling tubing, drill cuttings, sludge from sedimentation) was stored in open-top Department of Transportation #1A2y1.5/100 or 300 solid waste drums and stored on site in a location designated by NASA. Liquid waste was stored in Department of Transportation #1A1y1.5/100 or 300 closed-top, two-bung drums and stored on site in a location designated by NASA. Groundwater extracted from the PTA during sampling and maintenance activities was treated using the on-site treatment system to remove VOC and discharged.

3.5.6 Operating Parameters for the Technology

The demonstration of the technology was designed to be completed in three operational phases. Baseline operation began started on 8 December 2003. Biostimulation (addition of electron donor to increase the activity of the indigenous microorganisms and stimulate dechlorination) began 1 March 2004. Ethanol was used as an electron donor at a time-weighted average concentration (added as a weekly pulse) was 1,384 mg/L (based on a four-fold excess of the stoichiometric electron donor demand exerted by the reduction sulfate and TCE). The ethanol concentration was re-calculated on a monthly basis and changed as necessary to correspond with the change in electron donor demand exerted by the electron acceptors in the groundwater. On 15 April 2004, the PTA was bioaugmented with KB-1™, a bacterial consortium containing *Dehalococcoides* species (Bioaugmentation). Ethanol amendment was continued during the Bioaugmentation phase.

The initial site investigation and the installation of the monitoring points and groundwater circulation system required the services of contractors to safely install soil boreholes, complete electrical wiring and set up the programmable logic controller (PLC). In general, a field technician checked the system on a weekly basis. The on-site PLC allowed for remote monitoring of the recirculation system flow rates, water levels, and alarm conditions. Data files were created on a daily basis by the PLC.

A number of operating parameters were maintained at constant set points. The groundwater circulation rate was maintained at approximately 1.5 gpm; however, system alarms were tripped due to storm damage and recurring biofouling of the injection wells, resulting in frequent system shutdowns.

The progress of the demonstration was primarily monitored by collecting groundwater samples from the PTA for analysis of VOC, DHG, and sulfate. Samples were periodically collected for metals, PLFA, and DHC analysis. Snapshot sampling events (i.e. complete rounds of samples collected from all multilevel sampling points) were completed at the end of each phase of the demonstration. The data from each snapshot was utilized to calculate mass discharge at the appropriate multilevel transect.

Following the completion of the demonstration, a final round of groundwater samples was collected from the centerline monitoring wells (August 2005). At this time, the system had been shut-down for twelve months.

3.5.7 Experimental Design

The approach used to meet the project objectives was to compare VOC/DHG concentrations and the mass discharge of VOC from the test plot during the Baseline, Biostimulation, and Bioaugmentation phases of the study. It was anticipated that amendment of the PTA would result in reductive dechlorination of TCE to ethene (i.e. decreasing TCE concentrations) and increase VOC mass discharge. Prior to the Baseline phase, tracer tests were performed to determine groundwater velocities.

3.5.8 Sampling Plan

Groundwater Sample Collection Protocols

Prior to collecting groundwater samples for chemical analysis, the stagnant water in the well casing was evacuated (purged) to allow sampling of groundwater that is representative of aquifer conditions. Purged water was passed through a flow-through cell containing the electrode of a YSI 556 multi-parameter meter for concurrent measurement of dissolved oxygen concentration, pH, ORP, temperature and specific conductance. Purging was continued for a minimum of three well volumes and until field parameters stabilized. Subsequently, groundwater samples were collected directly into the sample bottles using Waterra tubing at a slow flow rate to minimize sample agitation. Detailed groundwater sample collection protocols, including standard procedures for the measurement of field parameters, are provided in the Sampling and Analysis Plan (SAP; Appendix F). Procedures to ensuring data quality are summarized in the Quality Assurance Project Plan ([QAPP]; Appendix G).

Soil Sample Collection Protocols

Discrete soil samples were collected of the materials from the LSU using a modified direct-push rig technique. The soil cores were collected using a 4-ft long 2.5 inch-diameter core barrel that contained a 4-ft long, 1.5-inch diameter butyrate sleeve to collect the soil. Each core was separated into 2-ft intervals and subsampled for VOC, headspace screening, and metals. Detailed soil sample collection protocols, including VOC extraction procedures, are provided in the SAP. Procedures to ensuring data quality are summarized in the QAPP.

Sample Analysis

The monitoring program included bi-weekly collection of groundwater samples from the combined flow of the extraction wells, and the centerline monitoring wells for both VOC and DHG analysis. Periodic samples were also collected from the combined flow of the extraction wells and centerline monitoring wells for analysis of VOC, DHG, anions, dissolved iron, and manganese. Additionally, select samples from extraction wells and centerline monitoring wells were submitted for analysis of volatile fatty acid (VFA), total organic compound (TOC), and phospholipid fatty acids (PLFA). Groundwater samples were also collected from the multilevel wells during each phase of the pilot test, with the exact timing of these sample events based upon the results from the weekly and monthly monitoring of extraction and monitoring wells. The monitoring program for the pilot test activities is described in further detail in the SAP. Procedures to ensuring data quality are summarized in the QAPP.

The primary component of the performance monitoring approach consisted of biweekly monitoring of chlorinated ethene and ethene concentrations in the centerline monitoring wells and the total extracted groundwater flow to assess trends in chloroethene biodegradation and the rate of chloroethene and ethene mass removal from the PTA. During each of the three operational phases, up to three complete rounds of groundwater samples were collected from the multilevel transect sampling points for analysis of chloroethene and ethene concentrations, which were used to estimate VOC mass discharge/mass flux at the transect.

In addition, periodic samples were collected for analysis of PLFA analysis to characterize changes in biomass concentration, VFA and TOC to characterize the distribution of electron donors within the PTA, and inorganic analytes (dissolved iron and manganese, and sulfate) to characterize geochemical impacts on groundwater quality.

Field Quality Assurance/Quality Control (QA/QC) Controls

A description of the field QA/QC controls used during the demonstration is presented in Appendix F. Field QA/QC samples consisted of trip blanks, field blanks, equipment blanks,

matrix spike/matrix spike duplicate (MS/MSD) samples, and field replicate samples were collected to monitor sampling and laboratory analytical performance with respect to groundwater samples. The collection frequencies for these control samples are summarized in Appendix F.

Data Quality Parameters

Data quality objectives (DQO) are based on the need to monitor the primary data quality indicators (DQI): precision, bias, accuracy, representativeness, completeness, and comparability (often referred to as PARCC criteria). Measures to ensure data qualities are described in Appendix G.

Data Quality Indicators

Quantitative QA objectives for the demonstration are based on the analysis of samples collected and analyzed as outlined in Appendix G. Calculations of DQI and QA objectives for precision, accuracy, method detection limit (MDL) are provided in Appendix G.

3.5.9 Demobilization

All wells at the demonstration site were abandoned in accordance with CCAFS requirements. All ex situ infrastructure was removed from the site.

3.6 Selection of Analytical/Testing Method

A summary of the analytical methods used during the demonstration are presented in Table 3-3. Detailed descriptions of these methods are summarized in Appendix H. Field methods are summarized in Appendix I. Where possible, the methods chosen were standard methods promulgated by either the USEPA or American Standard for Testing and Materials (ASTM).

3.7 Selection of Analytical/Testing Laboratory

Commercial laboratories for analyses were selected on the criteria of lowest cost and demonstrated technical competence. VOC (groundwater), DHG, and anion analyses of groundwater samples were performed by SiREM (Guelph, Ontario) using published analytical protocols similar to those of USEPA. In addition, the microbial characterization tasks (DGGE, GeneTrac DHE assay, microcosms) were completed by SIREM. Dissolved metals and TOC analysis were completed by PSC (Mississauga, Ontairo) using US EPA Method 200.7 (Revision 4.4) and 5310 C, respectively. MPN and plating were conducted by GAP Environmental (London, Ontairo) using Standard Methods 9512C and 9221, respectively, while VFA concentrations were determined by Microseeps Laboratory (Pittsburgh, Pennsylvania) using USEPA method AM21G. Laboratory Analytical methods used during the demonstration are

summarized in Table 3-3 and detailed descriptions of the analytical methods employed for VOC, DHG, VFA, PLFA, anion, and metals analysis are provided in Appendix I.

Table 3-3. Summary of Analytical Methods.

Parameter	Analytical Method	Method Number	Analytical Laboratory	Quantitation Limit	Sample Container	Preservative	Holding
Field Parameters (pH, DO, ORP, specific conductance, temperature)	Ion Specific Electrode	Field	NA	Varies	NA	NA	NA
Permanganate	Colorimetric	Field	NA	Varies	NA	NA	NA
Volatile Organic Compounds-Groundwater	GC/FID	Colorimetric	SiREM	10 mg/L	40 mL VOA	HCl to pH<2 cool to 4°C	14 days
Volatile Organic Compounds - Soil	GC/MS	[1]8260B	PSC	0.002 ug/g	2-60mL Teflon lined jars	HCl, cool to 4°C	14 days
Dissolved Hydrocarbon Gases (ethene, ethane, methane)	GC/FID	-	SiREM	10 mg/L	40 mL VOA	HCl, cool to 4°C	7 days
Bromide (conservative tracer)	Ion Chromatography Ion Specific Electrode	- Field	SiREM	0.14 mg/L	20 mL plastic	cool to 4°C	28 days
Dissolved Iron/Manganese	Field-filtered, ICP	-	PSC	0.05 mg/L	125 mL plastic	nitric acid to pH<2 cool to 4°C	28 days
Total Organic Carbon	Ion Chromatography	[2]5310 C	PSC	0.2 mg/L	125 mL plastic	sulfuric acid cool to 4°C	10 days
Volatile Fatty Acids (acetate, lactate)	GC/FID	AM21G	Microseeps	varies mg/L	40 mL VOA	cool to 4°C	21 days
Inorganic Anions (nitrate, nitrite, phosphate, sulfate)	Ion Chromatography	-	SiREM	varies mg/L	15 mL plastic	cool to 4°C	2 to 28 days
DHE/DGGE - Groundwater	16SRNA	-	SiREM	Trace	2-1 L plastic	cool to 4°C	7 days
DHE/DGGE - Soil	16SRNA	-	SiREM	Trace	30 mL	cool to 4°C	7 days
PLFA	-	-	Microbial Insights	-	-	-	-
Plating	-	[2]9215C	GAP	-	-	-	-
MPN	-	[2]9221C	[2]GAP	-	-	-	-

Notes:

[1] United States Environmental Protection Agency Method Number
[2] Standard Methods Method Number
N/A - not Applicable
SiREM – Site Remediation Laboratory
PSC - Environmental & Analytical Services
Microseeps Inc. - Environmental Sampling & Analytical Services
Microbial Insights - Molecular & Microbial Analyses
GAP - EnviroMicrobial Services
GC - Gas Chromatograph
TOC - Total Organic Carbon
DO - Dissolved Oxygen
ORP - Oxidation Reduction Potential
VOA - Volatile Organic Analyte
PLFA - Phosopholipid Fatty Acid Analyses
MPN - Most Probable Number
DHE - Dehalococcoides Ethenogenes
DGGE - Denaturing Gradient Gel Electrophoresis
FID - Flame Ionization Detector
MS - Mass Spectrometry

4.0 Performance Assessment

4.1 Performance Criteria

The performance of the demonstration will be evaluated using the general performance criteria provided in Table 4-1. Qualitative and quantitative criteria are classed as either primary or secondary performance assessment criteria, respectively.

The primary criteria constitute the performance objectives of the technology demonstration. As stated in Section 1.2, the general objectives of the demonstration are to evaluate the impacts of ISCO on the native microbial community, determine if ISB is feasible, and whether bioaugmentation enhances VOC degradation post-ISCO. In general, the performance criteria will be used to evaluate these objectives by:

- Determining the ability of the native and bioaugmented microbial consortia to colonize the ISCO test area and remaining source zone;
- Quantifying the effect of the technology on the mass flux from the source zone;
- Quantifying the effect of the technology on VOC degradation rates;
- Assessing the potential benefits of bioaugmentation; and
- Evaluating the difficulty in implementing this technology at the field scale.

4.2 Performance Confirmation Methods

The success of the technology demonstration was evaluated using the performance expectations and confirmation methods presented in Table 4-2. Successful implementation of the technology will demonstrate that the technology results in significant post-ISCO microbial activity, a statistically significant increase in the degradation rate of aqueous TCE with rapid and complete degradation to ethene. As a consequence of the microbial activity and VOC degradation, the rate of TCE DNAPL removal will increase as compared to pump and treat, decreasing the duration of remediation required for complete restoration of the PTA.

Table 4-1. Performance Criteria.

	Performance Criteria	*Description of Criteria*
PRIMARY	Microbial Activity In Source Zone	The ability of the indigenous and inoculated consortia to colonize the source zone after oxidant treatment is essential for the coupling of oxidation and bioremediation technologies
	TCE Degradation Rate	Degradation of the parent compound (TCE) will enhance the rate of DNAPL removal; rapid DNAPL dissolution decreases length of remediation. Degradation rate may be impacted by microbial inhibition via post-oxidation geochemical conditions
	Extent of Dehalogenation	Dehalogenation of TCE will indicate activity of microorganisms capable of degradation. Complete degradation of TCE to ethene will limit the mobility of the chlorinated daughter products
	Mass Flux from DNAPL	Rate that mass is removed from DNAPL by remedial technology; presence of DNAPL mass requires remediation of the groundwater plume over a period of decades to centuries
SECONDARY	Duration of Remediation	Time required to remove the source zone using enhanced bioremediation/bioaugmentation relative to flushing with unamended groundwater (base case treatment).Estimated based upon a comparison of TCE concentration in initial boreholes and mass flux data
	Factors Affecting Performance > location and amount of biomass injected into PTA	Creating a zone of highly active dehalogenating biomass in the immediate vicinity of the DNAPL is of critical importance; colonization of dehalogenating microorganisms is influenced by specifications of innoculum, location of injection point, and concentration
	> location and concentration of electron donor injected into PTA	Electron donor is anaerobically fermented to produce hydrogen (the primary substrate) which can be utilized by non-dehalogenating microorganisms; need to ensure that electron donor is supplied to active dehalogenators in the source zone
	> geologic heterogeneity	The presence of low permeability zones may limit delivery of both the inoculum and electron donor to the source zone
	> post-oxidation geochemical conditions	Elevated pH, highly oxidizing conditions, and manganese species may inhibit microbial activity
	Implemenation Issues	
	>maintenance requirements	One operator with minimal additional training is required for occasional visits during the demonstration; weekly adjustments and maintenance will be needed in addition to sample collection
	>reliability	Operation of system expected to be highly reliable and capable of operating without the need for a full-time operator
	Appropriate pH & Redox Conditions	Near neutral pH (or near site background), low dissolved oxygen concentration and oxidation-reduction potential are required to permit an increase in the activity of the dehalogenating microorganisms

Notes:

PTA - Pilot Test Area

TCE - Trichloroethene

DNAPL - Dense Non-aqueous Phase Liquid

Table 4-2. Expected Performance and Performance Confirmation Methods.

	Performance Criteria	*Expected Performance Metric*	*Performance Confirmation Method*	*Actual*
PRIMARY CRITERIA			***Qualitative***	
	Microbial Activity In Source Zone	Increased in the concentration of biomass and the extent of colonization of source by indigenous and bioaugmented consortia	PLFA, Dhc, and DGGE analysis; aerobic and anaerobic plating; microcosms to confirm degradation rates	Significant microbial activity was present throughout the demonstration. Organisms present during the Baseline phase did not dechlorinate TCE; apparent inhibition of dechlorination in the presence of manganese dioxide during Biostimulation & Bioaugmentation phases (Appendix I)
	Extent of Dehalogenation	Complete dehalogenation to ethene	Analysis of groundwater samples for TCE and TCE daughter products	Minimal dechlorination during Baseline; dechlorination to cisDCE, VC and ethene during the Biostimulation & Bioaugmentation phases (Figures 4.3, 4.4 and 4.5)
			Quantitative	
	Mass Flux from DNAPL - after amendment with electron donor (biostimulation)	Increase in mass flux above the base case[1] treatment	Measurement of the concentrations of VOCS, ethene; calculation of mass flux	No significant increase in mass flux (Figure 4.4)
	- after bioaugmentation	Increase in mass flux above the relative to biostimulation[1]		No significant increase in mass flux (Figure 4.4)
SECONDARY CRITERIA	*Qualitative*			
	TCE Degradation Rate	Increase in degradation rate following bioaugmentation	Interpretation of trend and distribution of VOCs, ethene, in groundwater	Increases in the rate of cis-DCE production following electron donor addition
	Factors Affecting Performance - location and amount of biomass injected into test plot	Mobility of biomass may be limited in porous media; accumulation of biomass in the source zone preferred	Experience from operation of demonstration; collection of samples for microbial characterization	Not evaluated: a small increase in biomass density was observed.
	- location and concentration of electron donor injected into test plot	Electron donor may be preferentially consumed by biomass without stimulating dehalogenation of chlorinated ethenes	Experience from operation of demonstration; collection of groundwater samples and analysis of electron donor concentration	Evidence of significant sulfate reduction; minimal methanogenesis during the demonstration
	- geologic heterogeneity	Low permeability may limit the delivery of electron donor and biomass to the source	Experience from operation of demonstration; tracer testing and soil sampling	Not evaluated except at injection wells; permeability reductions during to biofouling
	- post-oxidation geochemical conditions	High pH, high manganese concentrations, oxidizing conditions may inhibit microbial activity	Measurement of the concentrations of Mn, field parameters (pH level, ORP, DO)	Some evidence (via microcosm studies) that high manganese concentrations (MnO2) inhbited reductive dechlorination
	Implementation Issues - maintenance requirements	Replacement of tubing in peristaltic pumps; adjustment of injection level control system; replenishment of amendments	Evaluation of maintenance records and daily field logs	Implementation of biofouling control measures were the only significant maintenance requirement
	- reliability	Fraction of time system is shut down (zero flow)	Evaluation of system operational records	Significant downtime due to biofouling and hurricane damage
			Quantitative	
	Mobility of Groundwater Plume	Decrease in the steady-state plume length	Calculated based on simulated steady-state plumes using degradation rates estimated from test plot	Not assessed due to small size of demonstration plot
	Achieve Appropriate Geochemical (pH, Redox, Mn, Fe) Conditions	Anaerobic and reducing groundwater in test cell; pH at neutral / background levels; minimize Mn & Fe dissolution (which would lead to fouling)	Field measurements of pH, dissolved oxygen, oxidation/reduction potential, Mn, Fe	Highly reducing conditions achieved (see Section 4.3)

Notes

DNAPL - Dense Non-aqueous Phase Liquid
TCE - Trichloroethene
PLFA - Phosopholipid Fatty Acid Analyses
Dhc - Dehalococcoides
DGGE - Denaturing Gradient Gel Electrophoresis
VFAs - Volatile Fatty Acids
Mn - Manganese
Fe - Iron

4.3 Pre-Design Laboratory Studies

The results of pre-design laboratory studies are summarized in Appendix B. These studies resulted in the following conclusions:

- MnO_4^- treatment did not significantly inhibit the utilization of electron donors by fermenting bacteria in microcosms;
- Neither ethanol, methanol, glucose, lactate, glycol or acetate were abiotically oxidized by MnO_2 at a significant rate; however, MnO_2 reacted rapidly with oxalic acid;
- Complete dechlorination occurred only in microcosms bioaugmented with KB-1. However, stoichiometric conversion of the amended TCE to ethene was slower in microcosms that were pretreated with MnO_4^-;
- The average oxidant demand of the LC-34 soil was 2.3 g-$KMnO_4$/kg over 72 days; and
- There was no evidence of abiotic chloroethene or ethene oxidation by MnO_2 at environmentally significant rates.

4.4 Column Studies

The results of column studies completed at the University of Toronto are summarized in Appendix C. These studies resulted in the following conclusions:

- Rebounding of TCE concentrations following oxidation indicates that a polishing technology (such as ISB) is required;
- The addition of bacteria, either through the ambient movement of site groundwater or bioaugmentation, may be required to restore microbial activity following oxidant treatment;
- The inoculation of dechlorinating cultures into oxidized conditions may impair the ability of the culture to subsequently degrade cDCE, even when reducing conditions are reestablished; and
- Columns bioaugmented prior to the onset of manganese-reducing conditions could only dechlorinate TCE to cDCE; however, complete dechlorination to ethene occurred in columns bioaugmented after the onset of manganese-reduction.

4.5 Field Demonstration

4.5.1 Tracer Testing

A groundwater tracer test was conducted on 14 October 2004 using a conservative solute (NaBr) to evaluate flow conditions within the PTA. A tracer pulse (75 mg/L) was metered into the PTA through the injection wells for a period of 6 days. Migration of the tracer pulse was monitored by collecting bromide samples from each of the multilevel well sampling ports. Prior to collecting groundwater samples for chemical analysis, the stagnant water in the well casing was purged using a peristaltic pump. Multilevel wells were purged at a flow rate of 0.5 ml/min for a minimum of 2L and until field parameters stabilized. Subsequently, groundwater samples were collected directly into the sample bottles using Waterra tubing at a slow flow rate to minimize sample agitation. The results of the tracer tests are provided in Appendix D.

4.5.2 Slug Testing

Hydraulic response tests of IW-2 and IW-3 were completed on 30 July 2003 to determine the hydraulic conductivity of the LSU. The results from these tests were interpreted using Aquifer Test 3.0 (Waterloo Hydrogeologic) and are summarized in Appendix E. The average hydraulic conductivity was $7x10^{-6}$ m/s (2 ft/day).

4.5.3 Baseline Electron Acceptor Concentrations & Calculated Electron Donor Demand

Baseline concentrations of electron acceptors in groundwater and electron donor demand calculations are summarized in Table 4-3, resulting in a stoichiometric electron donor demand of 107 mg/L (as ethanol). The majority of the donor demand was exerted by sulfate (74 mg/L as EtOH).

Table 4-3. Summary of Electron Donor Demand Calculations

Electron Acceptor	Concentration[1] (mg/L)	Molecular Weight (g/mol)	Molar Ethanol Consumption Ratios	Ethanol Demand
Oxygen (O_2)	0.01	32.0	1/3	0
Nitrate (NO_3^-)	-	62.0	5/12	-
Sulfate (SO_4^{2-})	232	96.1	2/3	74
Perchloroethene (PCE)	-	165.8	2/3	-
Trichloroethene (TCE)	175	131.4	1/2	31
Dichloroethene (DCE)	13	96.9	1/3	2
Vinyl Chloride (VC)	0.39	62.5	1/6	0
Electron Donor Demand				107
Electron Donor Demand X 4				428

Notes

[1] Electron acceptor concentrations are based on the average constituent concentrations in the test plot

g/mol - grams per mole

mg/L - milligrams per liter

Note that Table 4-3 only includes the electron donor demand exerted by soluble electron acceptors. Assuming a unit volume of soil (1 cubic meter, porosity 0.33), the total electron donor demand of the soluble acceptors corresponds to 35 g of ethanol. However, MnO_2 in soil (average concentration soil of 7,224 mg/kg, bulk density 2,000 kg/m^3) is also a significant electron acceptor: in a unit volume of soil, the electron donor demand of the insoluble MnO_2 corresponds to 853 g of ethanol. Accordingly, the presence of MnO_2 in the Test Plot results in a 25-fold increase in the electron donor dosing requirements

4.5.4 Field Parameters & Geochemical Indicators

Field parameters (temperature, pH, specific conductance, and ORP) for the centerline monitoring wells are provided in Appendix K and summarized in Figure 4-1. Key geochemical indicators (total VFA, manganese, and sulfate) are provided in Appendix L and summarized in Figure 4-2. Temperature in test plot monitoring wells ranged from 24°C (Mar 2004) to 29°C (Oct 2003), reflecting seasonal variation in surface temperatures. The specific conductance of test plot groundwater was ~3,500 µS/cm throughout the demonstration, which is considered a brackish groundwater (Freeze and Cherry, 1979). Initial environmental conditions in the test plot were anaerobic (ORP measurements ranging from 32 to -40 mV, characteristic of Mn-reducing conditions, Wiedemeier et al., 1999) and slightly alkaline (pH measurements ranging from 81. to 9.2). During the Biostimulation/Bioaugmentation phases, both pH (final pH ranging from 7.4 to 7.6) and ORP (final ORP ranging -265 to -295 mV, characteristic of sulfate-reducing or methanogenic conditions; Wiedemeier et al., 1999) decreased in the test plot.

Concurrent with these changes the average concentration of total VFA (representing a combined concentration of acetate and lactate) in these monitoring wells increased from 24 mg/L (Baseline) to 453 mg/L (20 Aug 2004) while the average concentration of sulfate decreased from 883 mg/L (Baseline) to 50 mg/L (20 Aug 2004). This change indicates that the available electron donor was initially utilized for sulfate-reduction until the sulfate reservoir was depleted. Addition fingerprinting of the VFA data was performed (data presented in Appendix L) indicating that acetate, butyrate, and propionate were the predominant VFA. Acetate is not utilized as an electron donor *Dehalococcoides* (Duhamel et al., 2002); however, it is readily used by sulfate-reducing, manganese reducing, and methanogenic organisms.

The maximum methane concentration observed during the Baseline treatment phase was 0.3 mg/L. During electron donor addition (Biostimulation/Bioaugmentation) only a small increase in methanogenesis were observed. The maximum methane concentration was only 0.7 mg/L (MW-3). The absence of a significant increase in methane concentrations suggests that methanogenesis may have been inhibited by the high VOC concentrations (DiStefano et al., 1991).

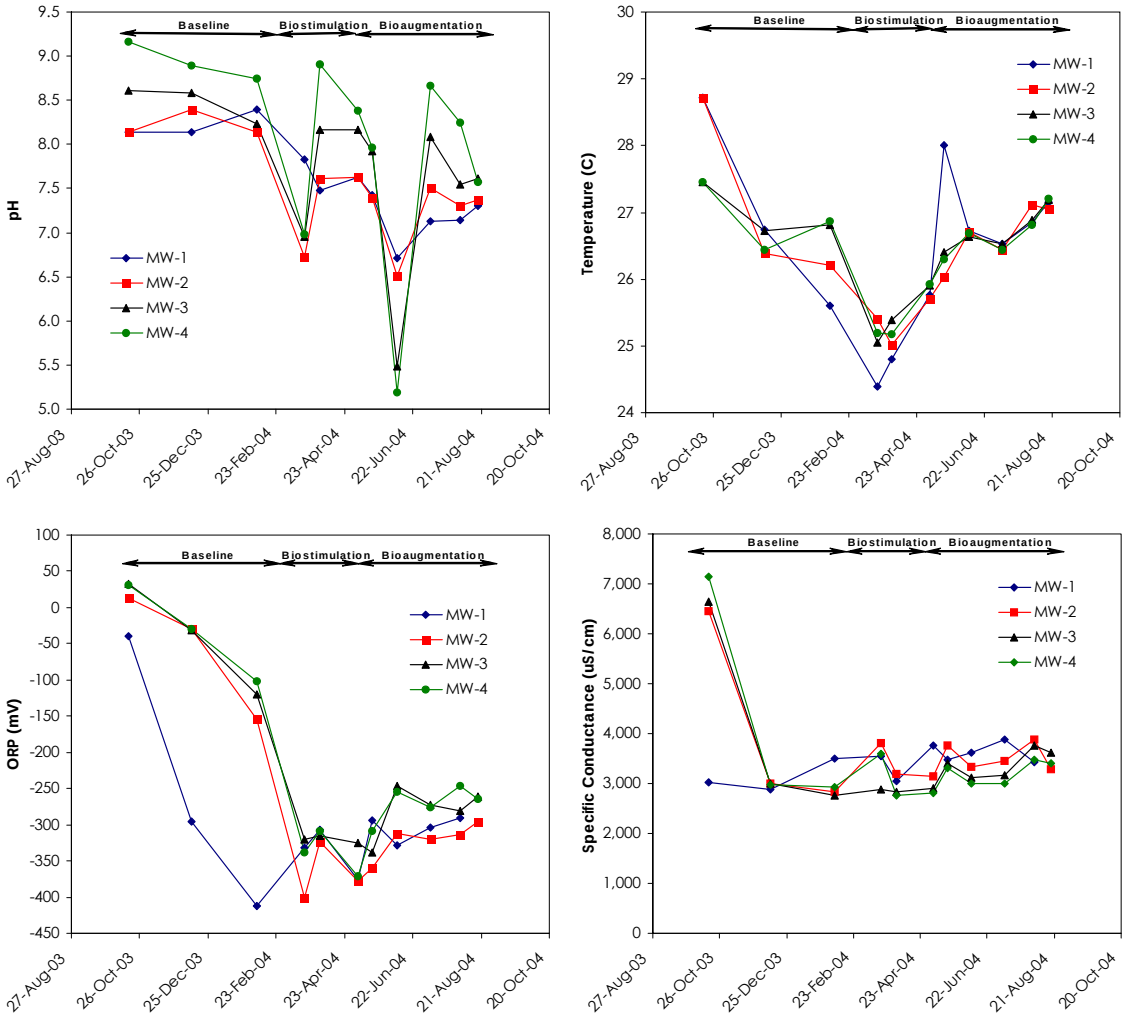


Figure 4-1. Field Parameters in Centerline Monitoring Wells.

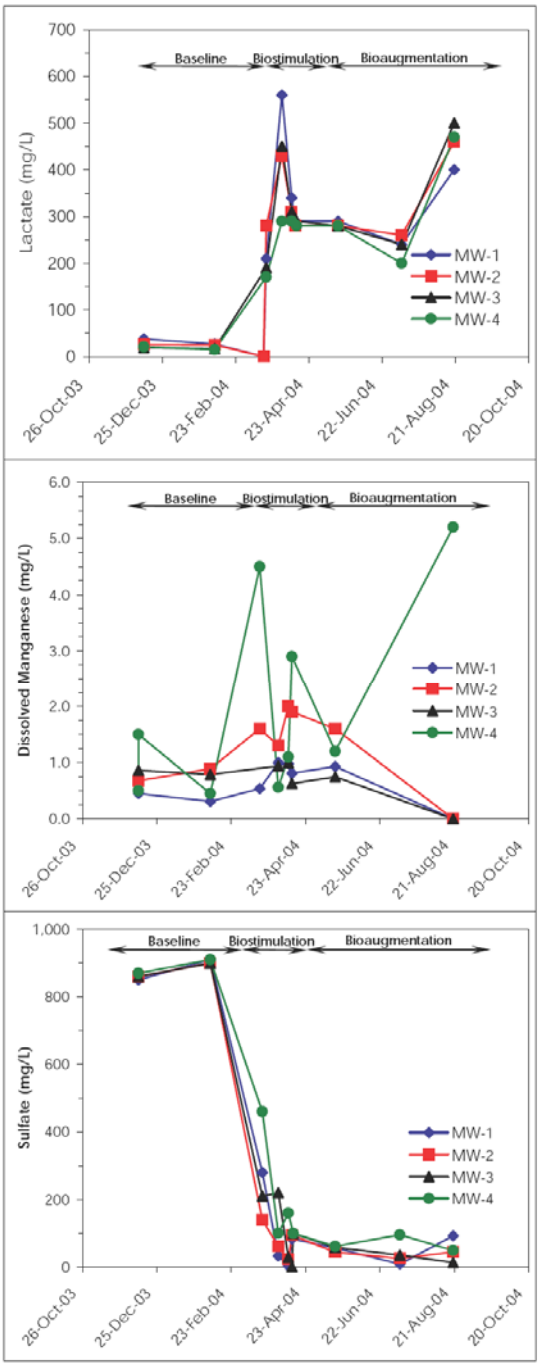


Figure 4-2. Key Geochemical Indicators in Centerline Monitoring Wells.

The average baseline sulfate concentration in groundwater was 285 mg/L; the maximum concentration observed following electron donor addition was 11 mg/L with several non-detects, indicative of increases in the activity of sulfate-reducing microorganisms in groundwater. The decrease in sulfate concentration in the Jan-03 sample event occurred as concentrations of butyric and propionic acid increased (see Section 4.5.5).

There was limited evidence that electron donor amendment resulted in either iron or manganese reduction. Although not measured during the Baseline phase of system operation, dissolved iron is present in background groundwater at an average concentration of 5.2 mg/L (CRA, 1999). In comparison, the maximum dissolved iron concentration observed in PTA monitoring wells following electron donor amendment was 10 mg/L. Background dissolved manganese concentrations range from 0.006 to 0.527 mg/L (CRA, 1999); dissolved manganese concentrations observed during the demonstration range from 0.056 to 0.85 mg/L, suggesting that electron donor addition did not result in significant manganese reduction. No significant increase in the concentration of dissolved manganese, expected to be a significant sink for electron donor under these environmental conditions, appeared to occur during the demonstration (Figure 4-2).

VOC and DHG

All VOC and DHG data are provided in Appendix L. Time-series plots presenting the concentrations of chloroethenes, ethene and methane in the centre-line monitoring wells are presented in Figure 4-3. Average VOC and DHG concentrations during each demonstration phase are summarized in Table 4-4. Under intrinsic conditions (Baseline), TCE, cDCE, and VC were detected in test plot groundwater samples; however the concentrations of cDCE (the dominant TCE degradation product in these samples) and VC represent only 16% of the total ethenes concentration. Ethene was not detected in any of the twelve Baseline groundwater samples (<0.1 mg/L). The limited extent of reductive dechlorination of TCE under intrinsic conditions was likely limited by the absence of sufficient electron donor to overcome the significant electron donor demand exerted by other reductants (e.g., manganese and sulfate).

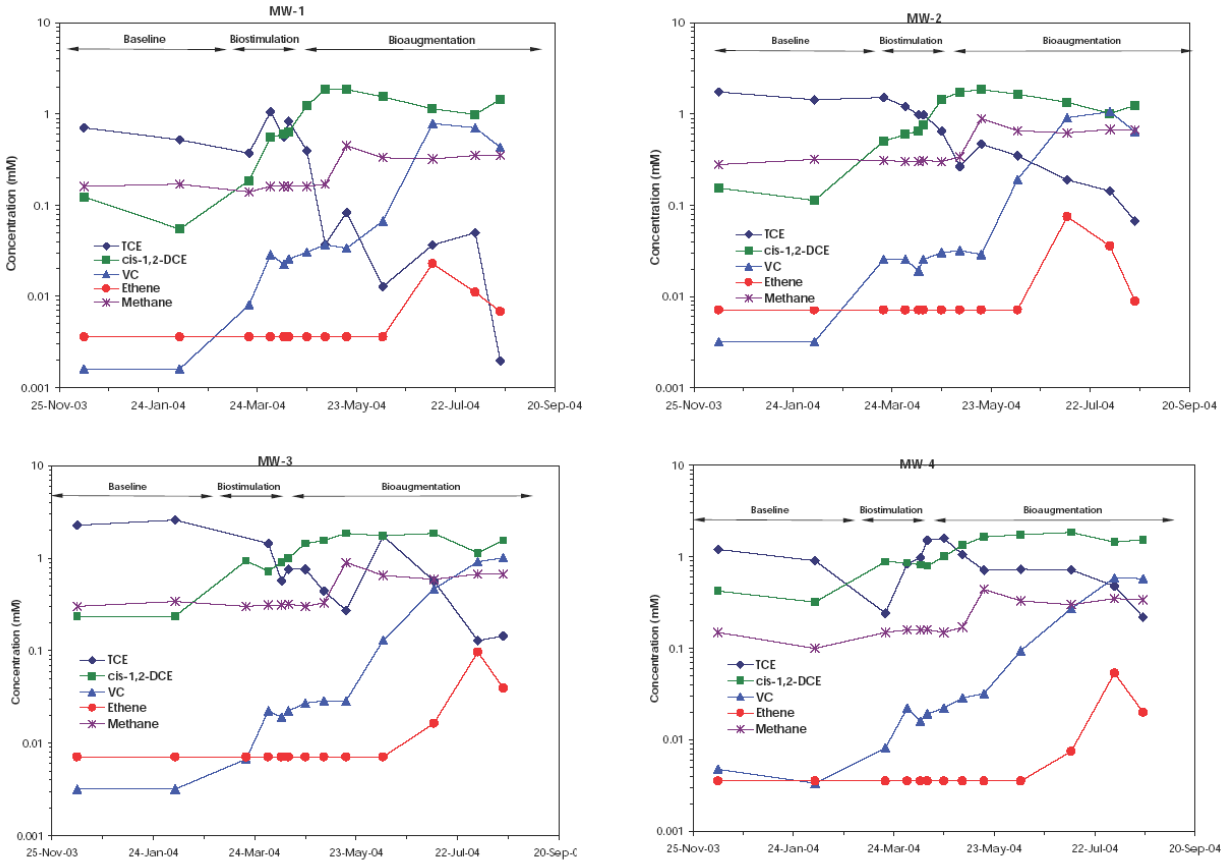


Figure 4-3. Chloroethene, Ethene, and Methane Concentrations in Centerline Monitoring Wells.

Table 4-4. Summary of PTA Geochemistry.

		Concentration (mg/L)					
Phase	**Date Collected**	**TCE**	**cis-DCE**	**VC**	**Ethene**	**Methane**	**Sulfate**
Baseline	19-Mar-04	94	61	0.8	0.2	0.2	273
Biostimulation	12-Apr-04	112	125	2	0.2	0.2	94
Bioaugmentation	18-Aug-04	14	140	42	0.5	0.5	50
Post-Demonstration	16-Aug-05	0.1	20	22	4	0.9	5

1. Post-demonstration groundwater samples were collected twelve months after system shut-down
2. Concentrations represent the average result of groundwater samples collected from the centreline monitoring wells at the end of each demonstration phase

Following the start of the Biostimulation phase increased dechlorination of TCE to 1,2-*cis*-dichloroethene (cDCE) was observed, suggesting that organisms mediating TCE dechlorination to cDCE were present in the PTA. The addition of electron donor did not appear to stimulate methanogenesis and concentrations of methane remained <1 mg/L throughout the demonstration. There was minimal dechlorination of cDCE to vinyl chloride (VC) and/or ethene. Further dechlorination, including the production of ethene at concentrations as high as 2.7 mg/L, occurred during the Bioaugmentation phase (Figure 4-3).

The mean chloroethene mass flux at the downgradient fence of multilevel monitoring wells (Fence 3) is summarized in Figure 4-4. Chloroethene mass fluxes at Fence 3 ranged from 39 to 53 mmole/ft^2/day, corresponding to TCE removal rates of 2.5 to 3.6 kg/day. A significant increase in chloroethene mass flux was not observed during the demonstration. The extent of dechlorinating activity (given by the dechlorination score, which represents the mole fraction of chlorine removed from the initial concentration of the parent compound) at Fence 3 is summarized in Figure 4-5 using box-and-whisker plots. The dechlorination score (N_D) is given by:

$$N_D = \frac{[cisDCE] + 2[VC] + 3[Ethene]}{3([TCE] + [cisDCE] + [VC] + [Ethene])}$$

where the values in parentheses represent molar concentration units and scores of 0.33 and 0.66 represent complete conversion to equivalent concentrations of cDCE and VC, respectively. A dechlorination score may be calculated for individual groundwater samples. Figure 4-5 represents the summary statistics (range, 25th and 75th percentiles, and median) for each complete round of samples collected from Fence 3. Although in each sample event there is a large range of scores, the dechlorination scores for the Baseline and Biostimulation sample events indicate

that there is relatively little dechlorination of TCE and that it results in the accumulation of cDCE. However the median dechlorination score of samples collected for the Bioaugmentation sample event (0.32)indicates that there was a shift in the extent of reductive dechlorination past cDCE by the end of the Bioaugmentation phase. This is evident in the distributions of dechlorination products shown in Figure 4-4, which illustrates that the Bioaugmentation sample event was unique in that it was the only sample event in which cDCE concentrations exceeded TCE concentrations and it had the highest concentrations of VC and ethene.

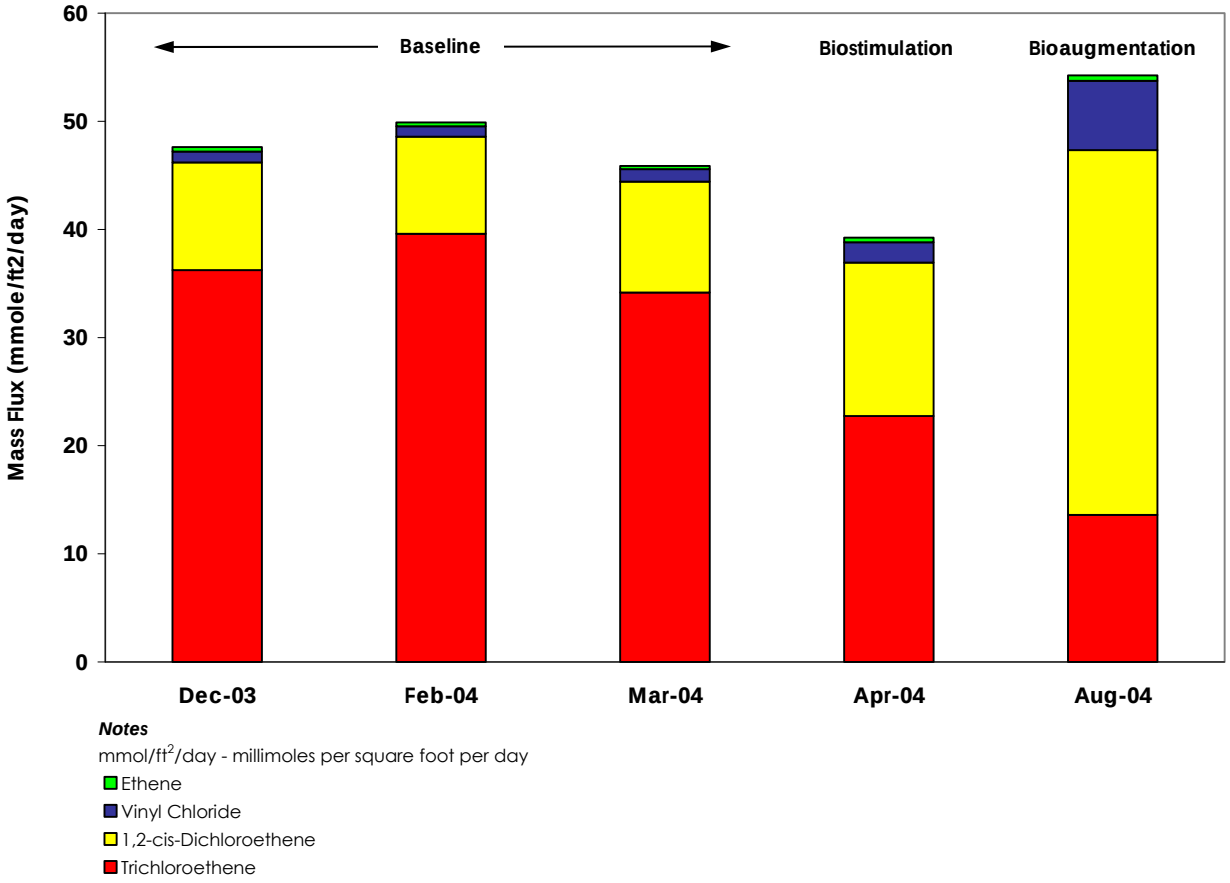
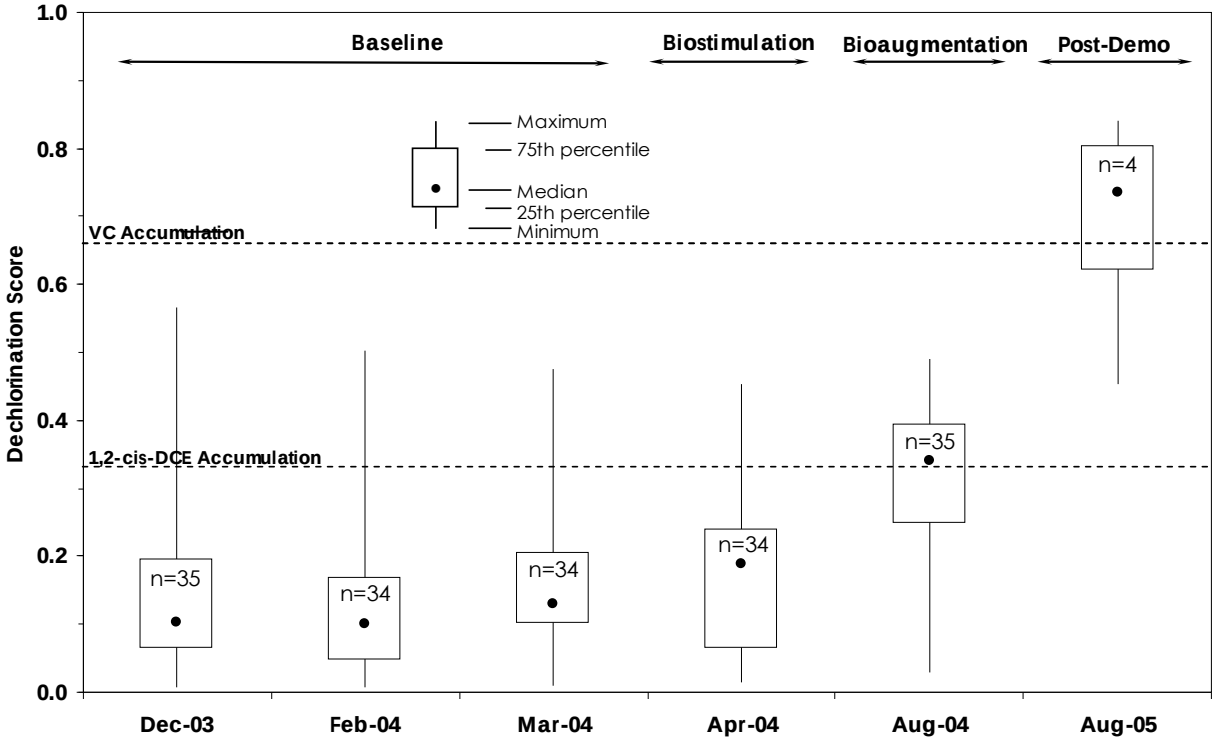


Figure 4.4. Mass Flux of Chloroethenes and Ethene in Groundwater at Fence 3.



Notes

1. Dechlorinating scores represent the molar fraction of chlorine removed from the initial concentration of the parent compound (i.e., TCE), which is equal to the total ethenes concentration,
2. Dechlorination scores calculated using data from all Fence 3 sampling locations, with the exception of the Aug-05 event (monitoring wells only).

Figure 4.5. The Extent of Dechlorination at Fence 3. Dashed Lines at 33% and 66% Represent Complete Conversion of the Parent TCE to Either cDCE or VC, Respectively.

This shift in the extent of dechlorination was confirmed in the Post-Demonstration sampling event (Table 4-4). The maximum ethene concentration in this sampling event was 10 mg/L (MW-1).

4.5.6 Chloroethene & Ethene Concentrations / Mass Discharge in Extracted Groundwater

Concentrations of chloroethenes and ethene in the extracted groundwater are summarized in Figure 4-6. These concentrations reflect mixing of groundwater containing TCE from both the PTA (i.e., groundwater impacted by electron donor addition and bioaugmentation) and from the surrounding aquifer (i.e., background groundwater). The concentrations of chloroethenes and ethene indicate that 20% of the parent TCE in the extracted groundwater was converted to cDCE and VC during the Baseline phase of the demonstration. No detectable concentrations of ethene were observed during Baseline.

During Biostimulation, changes in the proportions of the less-chlorinated degradation by-products observed in the extracted groundwater were consistent with the changes observed in the centerline monitoring wells. Following electron donor addition, increases in the concentration of cDCE were followed by increases in VC concentrations corresponding to 31% molar conversion of the parent TCE concentration to these less-chlorinated compounds. No detectable concentrations of ethene were observed during Biostimulation. During the Bioaugmentation phase of the demonstration, further increases in the concentration of cDCE, VC, and ethene occurred, corresponding to 56% molar conversion of the parent TCE concentration. The maximum concentration of ethene in the extracted groundwater was 0.3 mg/L.

The total chloroethene mass discharge in the extracted groundwater during the Baseline, Biostimulation and Bioaugmentation phases was 1.8, 1.9 and 2.0 kg/day (as TCE), respectively. A significant increase in the mass discharge did not occur during the demonstration.

4.5.7 Microbial Characterization

An extensive program of microbial characterization was completed using samples collected at the end of the Baseline, Biostimulation, and Bioaugmentation phases of the demonstration. Detailed reports for each of these three studies are included in Appendix M.

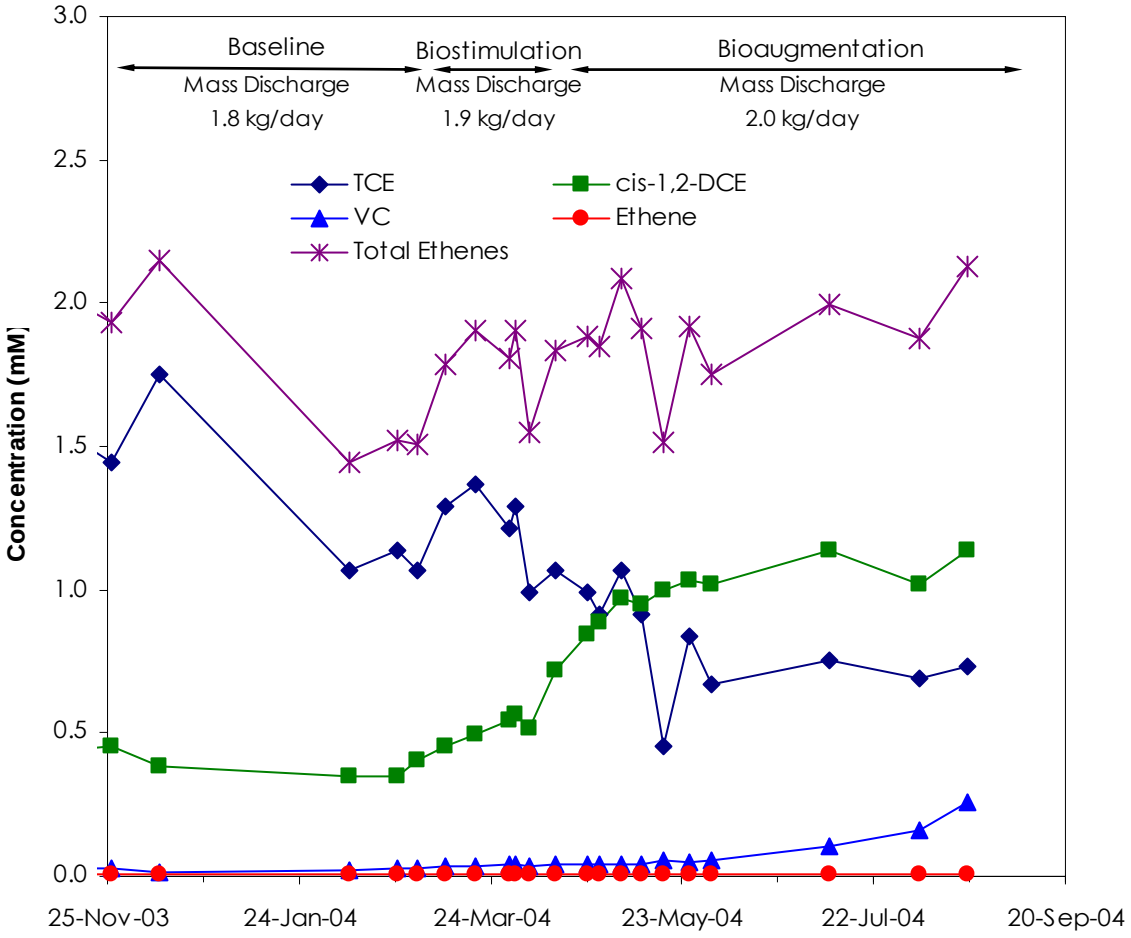


Figure 4-6. Chloroethene and Ethene Concentrations in Extracted Groundwater.

Key results of the Baseline microbial characterization study, using samples collected from the Test Plot and a background Control Plot, include:

- Biomass density in the Test Plot appears to be approximately two-fold higher than the biomass density in the Control Plot;
- There were significant differences in the microbial community structure between the Test and Control Plots. The Test Plot community includes members of the *Acinitobacteria*, *Acidovorax,* and *Symbiobacterium* which were not detected in the Control Plot. The microbial community in the Control Plot appeared to be dominated by members of the gamma subdivision of the Proteobacteria and, more specifically, Pseudomonas species, a number of which were not present in the Test Plot; and
- DNA from *Dehalococcoides* organisms is present in the Test Plot; however, only bacteria from Control Plot samples exhibited dechlorinating activity. Dechlorination activity in the ISCO Test Plot was strongly inhibited, even after over 276 days of incubation.

Key results of the Biostimulation microbial characterization study include:

- As anticipated, the shift in trophic conditions (e.g., substrate availability) resulted in a decrease in overall microbial diversity;
- The microbial population is dominated by Bacteria, with no evidence of methanogenic organisms;
- *Dehalococcoides* microorganisms were not detected; however, organisms at 2/3 sample locations completely dechlorinated TCE to ethene, indicating that these organisms were present at an initial cell density below the detection limit of the Dehalococcoides PCR method. Methanogenesis occurred concurrently in these samples with TCE dechlorination to ethene; and
- In a third sample, which contained visible MnO_2, dechlorination and methanogenesis were completely inhibited over 70 days of incubation.

Key results of the Bioaugmentation microbial characterization study include:

- There was no evidence of a further significant shift in the diversity of the microbial community. The total biomass density appeared to slightly increase;
- The microbial population is dominated by Bacteria. Targeted PCR assays for Archaea were negative, consistent with an ongoing absence of methane;
- Targeted PCR assays for *Dehalococcoides* indicate that this organism was present in 2/3 samples. Microorganisms at these samples completely dechlorinated TCE to

ethene. Methanogenesis occurred concurrently in these samples with TCE dechlorination to ethene; and

- In a third sample containing MnO_2, dechlorination and methanogenesis were completely inhibited over 84 days of incubation.

4.5.8 Field Demonstration Conclusions

The field demonstration resulted in the following conclusions:

- Ethanol fermentation occurred rapidly and fermentation products (such as acetate and lactate) were distributed throughout the Test Plot;
- Sulfate reduction occurred rapidly after the start of electron donor addition;
- Biodegradation of TCE to cDCE with much lower concentrations of vinyl chloride occurred following electron donor addition. Concentrations of TCE decreased significantly during the demonstration and were non-detect in the post-demonstration sampling event;
- Additional dechlorination occurred following Test Plot bioaugmentation, resulting in the formation of vinyl chloride and much lower concentrations of ethene; and
- A small increase in methane concentration occurred during following Test Plot bioaugmentation (average concentration 0.5 mg/L).

It was intended that the demonstration would continue longer to test the hypothesis that the ethene would eventually become the predominant dechlorination product; however, system operation was severely impacted by a series of hurricanes and the demonstration was terminated.

4.5.9. Key Geochemical Processes

In addition to the decreases in ORP measurements to levels characteristic of sulfate-reducing/methanogenic activity, there was strong evidence of anaerobic microbial processes usually associated with reductive dechlorination, including fermentation, manganese reduction, and sulfate reduction. Fermentation was evident by the rapid disappearance of ethanol and the appearance of typical fermentation products (including acetate, lactate, propionate, and butyrate). Although there were no significant increases in dissolved manganese concentrations (Figure 4-2), soil samples collected for the microbial characterization studies provided evidence of manganese reduction. While the color of Baseline soil samples (dark brown to black) was characteristic of MnO_2, subsequent soil samples from the same locations were light grey, indicating that the MnO_2 had been depleted by manganese reduction and that the manganese was now present in other mineral forms (e.g. reduced manganese mineral species, sorbed to other mineral surfaces, etc.). Declines in sulfate concentration were indicative of sulfate reduction by indigenous test plot microorganisms, although sulfate was not entirely depleted (Figure 4-2).

Interestingly, there was no evidence of methanogenesis, although ORP measurements (typically less than -240 mV during the Biostimulation/Bioaugmentation phases) indicated that the environmental conditions would support this process (Wiedemeier et al., 1999). The inhibition of methanogenesis may result from the presence of high chloroethene concentrations, competitive electron donor utilization by other reduction processes (esp. manganese reduction), and the absence of methanogenic bacteria (as determined by Archeae-specific molecular testing, see Section 4.3.3). In conjunction with the microbial diversity studies and consistent with data reported by Azadpour-Keely et al. (2004), it is evident that the PTA was rapidly recolonized in the three years following MnO_4^- amendment by a diverse microbial community.

Although the addition of electron donor resulted in environmental conditions suitable for manganese reduction, the absence of appreciable increases in dissolved manganese concentrations during the demonstration suggest that the transport of reduced manganese (Mn^{2+}) was limited. The results of geochemical modeling (Appendix J) suggest that the presence of sufficient sulfate favors the precipitation of reduced manganese (Mn^{2+}) as alabandite (MnS). Other significant controls on dissolved Mn transport in groundwater include cation exchange onto MnO_2 surfaces, precipitation as MnO_2, and precipitation as $MnCO_3$ (Stumm and Morgan, 1970).

These data suggest that the deposition of MnO_2 during ISCO has important consequences on the performance of subsequent efforts to promote reductive dechlorination. Manganese reduction is thermodynamically favored in comparison to dechlorination (Figure 4.7) and exerts an electron donor demand in significant excess of that required solely to support reductive dechlorination, increasing the quantity of electron donor required for source area treatment. In comparison to *Dehalococcoides* microorganisms that are necessary for cDCE and VC dechlorination to ethene, manganese reducing microorganisms rapidly utilize hydrogen, the sole electron donor for reduction of these chloroethenes. As shown in Figure 4-7, the reported hydrogen threshold for manganese-reduction is <0.1 nM; however, the reported thresholds for reductive dechlorination range from 2 to 11 nM (AFCEE, 2004). Accordingly, highly efficient hydrogen utilization by manganese-reducing microorganisms appears to inhibit dechlorination by maintaining the hydrogen concentration below the minimum threshold required to support reductive dechlorination of cDCE and VC.

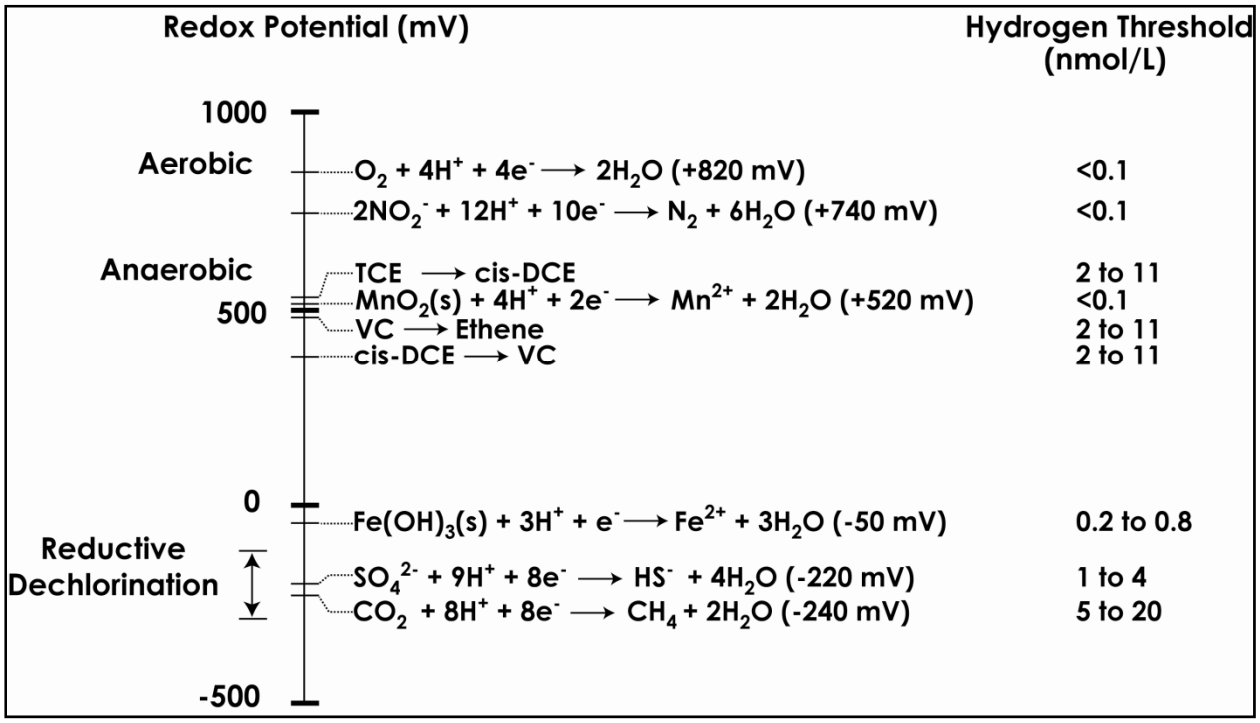


Figure 4-7. Oxidation-reduction Potentials and Hydrogen Thresholds for Common Electron Acceptors in Groundwater Environments (modified from Bouwer, 1994, includes data from AFCEE, 2004 and Yang and McCarty, 1998**).**

5.0 Cost Assessment

5.1 Cost Reporting

Costs were tracked by project milestones that were defined at the start of the demonstration in the on-line SERDP project financial tracking system (SEMS). The distribution of project funds by milestone is shown in Figure 5-1. The highest-cost milestone was the operation of the demonstration system (including monitoring) which comprised 30% of the total project cost.
The total cost of the demonstration was $843,000, resulting in the treatment of 12,500 ft^3 of soil containing approximately 370 kg of TCE. The corresponding unit costs of the demonstration are $2,381/$m^3$ and $2,280/kg-TCE. The unit costs incurred during the demonstration are much higher than those likely to be experienced during full-scale implement due to: 1) the small scale of the demonstration; 2) the extensive monitoring effort; and 3) the implementation of a groundwater recirculation system.

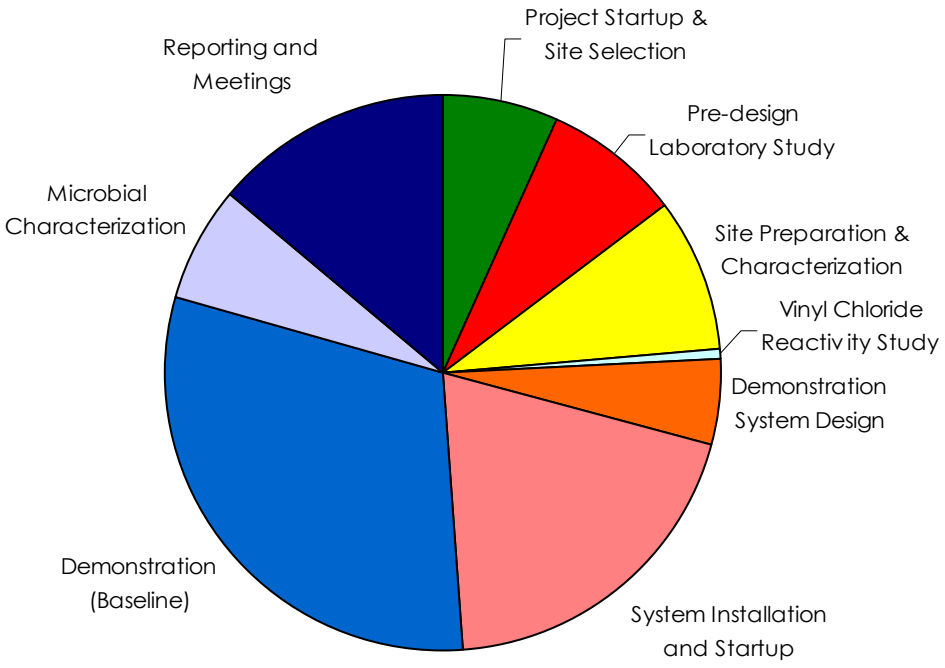


Figure 5-1. Distribution of Project Expenditures by Major Milestones.

5.2 Cost Analysis

5.2.1 Cost Comparison

The cost of full-scale source remediation was assessed by comparing the lifetime costs of sequential ISCO/ISB to the following technologies for a theoretical site:

- Pump-and-treat – contain groundwater in the source area using groundwater extraction wells and ex situ VOC treatment;
- ISCO – remove VOC mass from the source area using the injection of a concentrated solution of MnO_4^- followed by monitored natural attenuation (MNA); and
- ISB – contain groundwater in the source area and/or remove VOC mass using rapid biodegradation (ISB).

5.2.2 Cost Basis

Costing parameters are based upon a theoretical site with dimensions of 100 ft long by 100 ft wide. The corresponding source area is assumed to contain 1,500,000 gallons of TCE-impacted groundwater, with the TCE source zone present from 10 to 80 ft bgs. The geology in the source area includes a sand unit from 10 to 40 ft bgs, and a silty sand unit from 40 to 80 ft bgs. The corresponding mass of impacted soil is 35.7×10^6 kg (porosity 0.3, bulk density 1,800 kg/m^3). The total mass of TCE (dissolved, sorbed, and NAPL) in the source area is 12,500 pounds, and the average groundwater concentration exiting the source area is 175 mg/L. Additional details used in the cost assessment are provided in Table 5-1.

Capital and operating costs focus on those costs associated the implementation of the technology and do not include costs that may be site-specific and/or equal between technologies such as regulatory approvals. The operating period of each technology was evaluated by considering the time for the source zone to be removed via dissolution using the numerical solutions proposed by Falta et al. (Falta et al., 2005a and 2005b). The use of this approach for evaluating the operating period is further described in Section 5.2.4. All technologies were compared using the operating periods predicted based upon Falta's method at a real discount rate of 2.8% (Office of Management and Budget, 1992). A summary of the basis of the costs for each alternative is provided in Table 5-1. A brief description of the approach for each alternative is provided in the following sections.

Table 5-1: Basis of Cost Evaluation.

Parameter	Unit	Quantity
Porosity	v/v	0.3
Dimensions - Length	ft	100
Dimensions - Width	ft	100
Dimensions - Depth	ft	70
Pore Volume	Mgal	1.57
Bulk Volume	Mgal	5.23
Bulk Volume	m^3	19,810
Bulk Density (dry)	kg/m^3	1,800
Mass of Soil	kg	35,658,000
Groundwater Velocity	ft/day	0.02
Groundwater Flux	ft^3/day	133
Geochemistry		
Average Sulfate Concentration	mg/L	41
Average TCE Concentration Post-ISCO	mg/L	175
Average Mn Concentration Post-ISCO	mg/L	6
Sulfate Mass	kg	242
MnO_2 Mass	kg	40,320
TCE Mass	kg	5,600
In Situ Chemical Oxidation		
Natural Oxidant Demand	g $KMnO_4$/kg soil	1.7
Required Mass of $KMnO_4$ for NOD	kg	60,619
Required Mass of $KMnO_4$ for 100% TCE Removal	kg	13,458
Total Mass of $KMnO_4$ for 100% TCE Removal	kg	74,076
Equivalent Mass of MnO_2	kg	40,320
Treatment Parameters		
Discount Rate	%	2.8

Notes

Mgal - megagallon
m^3 - meter cubed
ft - foot
kg - kilogram
mg/L - milligram per Liter
L - Liter
v - volume
g - gram
% - percent

Alternative 1: Pump-and-treat

Two groundwater extraction wells screened in either the 10-40 ft bgs or the 40-80 ft bgs depth intervals (one shallow and one deep well) and equipped with electrically-operated submersible pumps. The total groundwater extraction rate is assumed to be ~5 gpm. Extracted groundwater will be treated using an air stripping tower and then recharged into the shallow aquifer via an infiltration gallery. The vapor stream from the air stripping tower will be treated using two granular activated carbon vessels connected in series. The duration of the pump and treat remedy to achieve a remedial goal of 5 µg/L is estimated to be 34 years using the approach described in Section 5.2.4.

Alternative 2: Enhanced In Situ Bioremediation

Shallow and deep permanent injection wells (35 total) will be installed in a grid across the source area. A solution of emulsified vegetable oil (EVO) will injected through these wells with the mass of EVO based on exceeding the electron donor demand (sulfate and TCE) by a factor of four in the first year of operation. Following the first year, the source area will be amended with EVO on an annual basis with a four-fold reduction in the mass of electron donor (1X stoichiometric excess). The frequency of EVO addition would be reduced at year 11 to once every 3 years and at year 21 to once every 5 years. The duration of the ISB source area treatment to achieve a remedial goal of 5 µg/L is estimated to be 55 years using the approach described in Section 5.2.4. However, it should be noted that substantial mass and concentration reductions may be observed in a shorter timespan, for example during an ISB study at LC34 achieved significant mass and concentration reductions within two years (Hood et al, 2008).

Alternative 3: In Situ Chemical Oxidation and Monitored Natural Attenuation

Six groundwater extraction wells and six injection wells screened in either the 10-40 ft bgs or the 40-80 ft bgs depth intervals (three shallow and three deep wells for each line of wells) and equipped with electrically-operated submersible pumps. The total groundwater extraction rate is assumed to be ~5 gpm. In the first two years of operation, potassium MnO_4^- will be recirculated through the source area. The total mass of MnO_4^-, which is based upon providing sufficient oxidant to meet the demand exerted by both uncontaminated soil (1.7 g $KMnO_4$/kg soil; IT, 2000) and TCE (2.4 mg $KMnO_4$/mg TCE), is 74,000 kg. The duration of the ISCO source area treatment to achieve a remedial goal of 5 µg/L is estimated to be 1.5 years using the approach described in Section 5.2.4. However, rebound post-ISCO is commonly observed at ISCO sites, with the cost efficiency of repeating oxidant injection decreasing with each injection event. For the purposes of this cost assessment it is assumed that ISCO results in removal of 85% of the TCE mass over the two years of operation. It would take an estimated additional 37 years following ISCO to meet remedial criteria of 5 µg/L through natural attenuation processes. The O&M costs during MNA would include long term groundwater monitoring and reporting.

Alternative 4: In Situ Chemical Oxidation and Enhanced In Situ Bioremediation

Six groundwater extraction wells and six injection wells screened in either the 10-40 ft bgs or the 40-80 ft bgs depth intervals (three shallow and three deep wells for each line of wells) and equipped with electrically-operated submersible pumps. The total groundwater extraction rate is assumed to be ~5 gpm. In the first two years of operation, potassium MnO_4^- will be recirculated through the source area. The total mass of MnO_4^-, which is based upon providing sufficient MnO_4^- to meet 100% of the demand exerted by both uncontaminated soil (1.7 g $KMnO_4$/kg soil; IT, 2000) and TCE (2.4 mg $KMnO_4$/mg TCE), is 74,000 kg. It is assumed that ISCO results in removal of 85% of the TCE mass over the two years of operation (i.e. lowers the electron donor demand of TCE by 85% during ISB). Given the shorter duration of the sequential approach (and correspondingly lower number of reinjection events), EVO injections would be completed using direct push wells in this alternative. In the third year of operation, shallow and deep injection wells (20 total) will be installed in a grid across the source area. A solution of EVO will injected through these wells with the mass of EVO based on electron donor demand of:

- Sulfate with a four-fold stoichiometric excess;
- The remaining TCE (i.e., 15% of the initial TCE) with a four-fold stoichiometric excess; and
- MnO_2 (corresponding to the mass of MnO_4^- injected in the previous two years) at the stoichiometric demand.

The contribution of these electron acceptors (i.e., sulfate, TCE, and MnO_2) to the total electron donor demand is shown in Figure 5-2. Note that MnO_2 exerted 77% of the total electron donor demand. In subsequent years, the source area will be amended with EVO every other year; however, the amount of electron donor required for sulfate and TCE reduction will be reduced by a factor of four and it is assumed that MnO_2 will not exert a further demand.

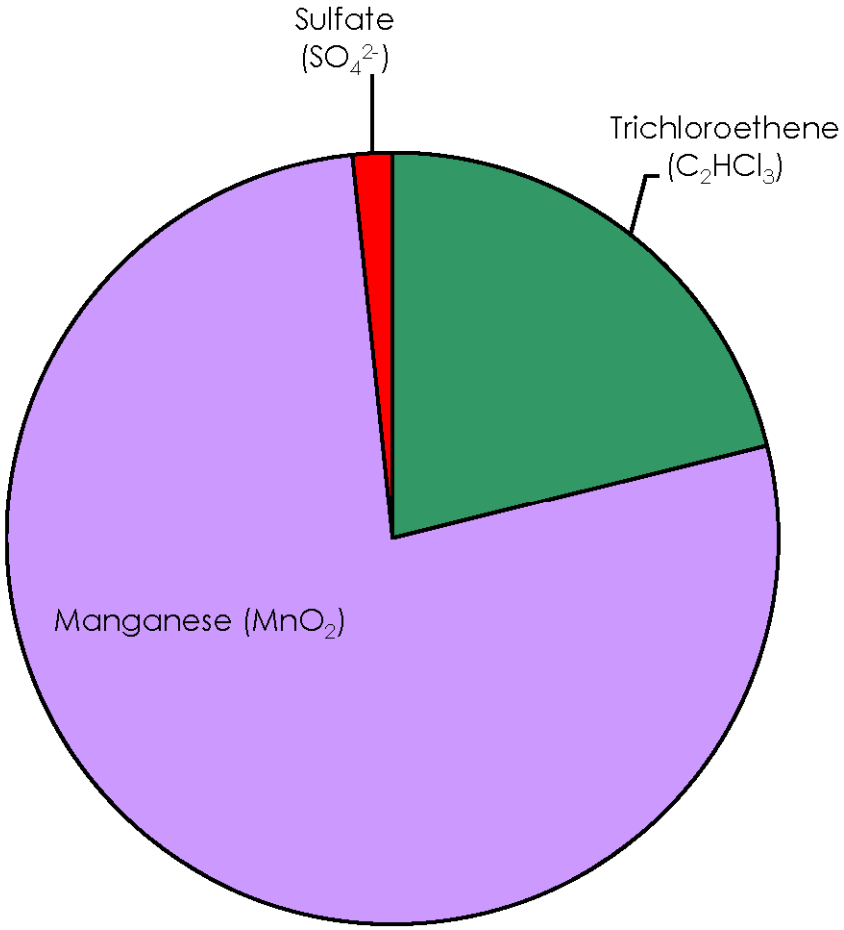


Figure 5-2. Contributions of the Principal Electron Acceptors on the Total Electron Donor Demand.

5.2.3 Cost Drivers

The principal cost drivers for the sequential technology include the costs of:

- Infrastructure – including injection well drilling and installation, above-ground piping and process instrumentation;
- O & M – including potassium MnO_4^- injection, electron donor injection, labor required for the annual injection events, performance monitoring, and reporting.

MnO_2 deposited during ISCO significantly contributes to the project cost by increasing the initial electron donor dosing requirements for ISB conducted in the ISCO treatment area. In Alternative 4, Mn-reduction, which was optimistically assumed to require only a stoichiometric electron donor dose to complete, exerted 77% of the electron donor demand. To some extent, this issue is specific to MnO_4^- since it is the only oxidant that results in the formation of a precipitate. However, the application of either persulfate or Fenton's reagent may also have an adverse impact on the subsequent application of ISB. The decomposition of persulfate results in the formation of sulfate which would be present at very high concentration and, although it is a soluble species and is more likely to attenuate over time in a source area through natural groundwater flow, could exert a significant electron donor demand and competitively inhibit reductive dechlorination. Similarly, the decomposition of peroxide in Fenton's reagent results in the formation of oxygen. At high peroxide concentration, there is the potential for exsolution of oxygen gas. Trapped oxygen gas within the formation could act as a long-term source of dissolved oxygen, which is toxic to some dechlorinating microorganisms (e.g., *Dehalococcoides*).

5.2.4 Life Cycle Costs

Summaries of the costs of all four alternatives (including both capital and annual operations and maintenance) are provided in Tables 5-2, 5-3, 5-4, 5-5, 5-6 and 5-7. The estimated life-cycle cost for the sequential ISCO/ISB technology is based on the capital cost of the infrastructure, plus operations and maintenance (including reagents, performance monitoring and reporting over the period of technology implementation. Total lifecycle costs of each alternative were calculated as the net present value over the estimated operating period at a real discount rate of 2.8% (Office of Management and Budget, 1992).

The operating period of each technology was evaluated by considering the time for the source zone to be removed via dissolution using the numerical solutions proposed by Falta et al. (Falta et al., 2005a and 2005b). This approach uses the following variables to evaluate source zone depletion and lifespan:

- Initial source mass;
- Groundwater flux through the source area;
- Mass discharge rate of the chemical;
- Target discharge rate of the chemical;
- Mass flux enhancement factor achieved by the technology;
- The relationship between remaining source mass and the contaminant mass discharge rate (what Falta et al. denote as Γ); and
- Fraction of the source remaining following the initial technology (i.e. ISCO).

The values used for each of these variables are presented in Table 5-2. The first four variables are based upon the physical and chemical properties assumed for the theoretical site (Table 5-1). The mass flux enhancement factor for each technology was assumed based upon values reported in literature. For ISB, a mass flux enhancement factor of 5 was considered based upon Christ et al. (Christ et. al, 2005). In addition, a second ISB scenario with a mass flux enhancement factor of 10, was considered to evaluate the sensitivity of the technology cost to that factor. For ISCO, a mass flux enhancement factor of 30 was considered based upon a survey of ISCO demonstration results performed by Krembs (Krembs, 2008). No mass flux enhancement (i.e. a value of 1) was used for pump and treat or natural attenuation. Γ, the relationship between remaining source mass and contaminant mass discharge rate, has been assumed to be 1 for the values presented in Table 5-2.

The predicted operating periods and total costs for each technology are summarized in Table 5-2. Based on this assessment, the P&T remedy would be expected to have an operating period of 34 years, ISB 55 years, ISCO/MNA 40 years and ISCO/ISB 10 years. Without any remedial actions, the source would take an estimated 145 years to be removed through natural attenuation processes. Should the mass flux enhancement of ISB be as high as a factor of 10, there is a predicted decrease in the operating period to 29 years. Clearly, for a site where schedule is the strongest driver for technology selection ISCO/ISB has a strong advantage over all other technologies.

Table 5-3 presents a sensitivity analysis of the operating periods for Alternatives 1 and 4 to the relationship between remaining source mass and contaminant mass discharge rate (Γ). It can be seen that for the theoretical site considered the predicted operating periods do not change substantially with changes in Γ.

In terms of capital costs (infrastructure only) ISB has the lowest capital costs. Pump and treat also has relatively low capital costs, primarily due to the low flow rate required to contain groundwater in the source area (5 gpm). The ISCO and ISCO/ISB options have the highest capital costs. Long-term annual O&M costs vary by alternative. ISCO is assumed to have low long-term O&M costs associated with MNA. The O&M costs for Alternatives 2 and 4, which include bioremediation, are higher since they include annual on-going electron donor addition with an aggressive dosing strategy intended to remove contaminant mass.

Overall, Alternative 2 (ISB) offers the smallest lifecycle costs, and the costs of implementing the sequential technology (Alternative 4 [ISCO/ISB]) are somewhat lower than that of implementing ISCO alone. However, the duration of the remedy is also a critical factor for most sites, and the

sequential technology (Alternative 4 [ISCO/ISB]) clearly offers advantages as compared to all other alternatives evaluated.

The cost analysis suggests that all three aggressive in situ alternatives have lower lifetime costs than pump-and-treat, providing that they have short operating durations, as predicted in the analysis presented herein. While ISCO/ISB option has a higher lifecycle cost than ISB alone the shorter lifetime of sequential approach may make it more advantageous than ISB alone.

Table 5-2. Summary of Mass Flux Parameters and Total Remedy Costs for Each Alternative.

TECHNOLOGY	Source Mass (kg)	Groundwater Flux through Source (m^3/year)	Initial Discharge Rate [b] (kg/year)	Target Discharge Rate [c] (kg/year)	Mass Flux Enhancement Factor	Γ	Remaining Fraction Source Mass for Secondary Technology	Remedy Duration (years)	Total Cost of Remedy ($)
Natural Attenuation	5,600	1422	249	0.01	1	1	na	235	na
Source Area Pump and Treat, 5 gpm	5,600	9948	1,741	0.05	1	1	na	34	$ 3,268,491
Source Area Bioremediation, Passive Approach	5,600	1422	249	0.01	5	1	na	55	$ 1,737,483
Source Area Bioremediation, Passive Approach	5,600	1422	249	0.01	10	1	na	29	$ 1,393,532
Source Area ISCO Recirculation, 85% removal / Followed by MNA	5,600	9,948 / 1,422 [a]	1,741 / 249	0.05 / 0.01	30 / 1	1	0.15	40	$ 2,801,206
Source Area ISCO Recirculation, 85% removal / Followed by Source Area Bioremediation	5,600	9,948 / 1,422	1,741 / 249	0.05 / 0.01	30 / 5	1	0.15	10	$ 2,613,724

Notes:

na - not applicable

[a] Where two values are shown the first value represents the primary technology, the second value the secondary technology.

[b] Based on Current Conditions, TCE 175 mg/L

[c] Based on TCE MCL, 5 µg/L

Table 5-3: Evaluation of the Sensitivity of Remedy Duration to Γ.

TECHNOLOGY	Γ	# Years to Meet MCL
Source Area Pump and Treat, 10 gpm	0.5	36
	1	34
	2	32
Source Area ISCO Recirculation / Followed by Source Area Bioremediation	0.5	11
	1	10
	2	9

Table 5-4: Cost Estimate for Alternative 1 (Source Area Pump & Treat**).**

TASK DESCRIPTION	UNIT	UNIT COST	QUANTITY	COST ($)	COST ($) WITH 20% CONTINGENCY
EXTRACTION WELL DRILLING					
Installation of two 6-inch wells (1 shallow/1 deep). Mobilization, development, per diem, decontamination, IDW management, drums included	LS			$ 13,200	$ 15,840
Drilling Oversight (Staff Professional)	day	$ 1,500	2	$ 3,000	$ 3,600
DRILLING SUBTOTAL				$ 16,200	**$ 19,440**
TREATMENT SYSTEM CONSTRUCTION AND STARTUP					
Trenching	LS			$ 50,000	$ 60,000
Air stripping tower	LS			$ 200,000	$ 240,000
Vapor phase activated carbon vessels (2)	each	$ 35,000	2	$ 70,000	$ 84,000
Piping, instrumentation and process control equipment	LS			$ 25,000	$ 30,000
Infiltration gallery	LS			$ 40,000	$ 48,000
Construction supervision/oversight (Technician and Staff Professional)	day	$ 2,600	40	$ 104,000	$ 124,800
Startup Testing	day	$ 1,500	10	$ 15,000	$ 18,000
TREATMENT SYSTEM CONSTRUCTION AND STARTUP SUBTOTAL				$ 504,000	**$ 604,800**
TOTAL CAPITAL COSTS (INCL. CONTINGENCY)					**$ 624,240**
ANNUAL OPERATIONS AND MAINTENANCE COST					
Activated carbon changeout	LS			$ 35,000	$ 42,000
Process Monitoring	day	$ 1,500	26	$ 39,000	$ 46,800
Performance Monitoring (including sampling and analysis)	sample	$ 250	52	$ 13,000	$ 15,600
Reporting	LS			$ 15,000	$ 18,000
ANNUAL OPERATIONS AND MAINTENANCE COST SUBTOTAL				$ 102,000	**$ 122,400**
ALTERNATIVE 1: COST FOR ALTERNATIVE 1 (SOURCE AREA PUMP & TREAT) TOTAL FOR FIRST YEAR					**$ 746,640**

Table 5-5: Cost Estimate for Alternative 2 (Source Area ISB**)**.

TASK DESCRIPTION	UNIT	UNIT COST	QUANTITY	COST ($)	COST ($) WITH 20% CONTINGENCY
INJECTION WELL SYSTEM DRILLING					
Installation of 35 injection well clusters, with 20 well clusters installed to 80 ft and 15 well clusters to 40 ft Mobilization, per diem, decontamination, IDW management, drums included	LS			$ 170,000	$ 204,000
Drilling Oversight (Staff Professional)	day	$ 1,500	30	$ 45,000	$ 54,000
DRILLING SUBTOTAL				$ 215,000	**$ 258,000**
FIRST YEAR ELECTRON DONOR AND KB-1 INJECTION					
EVO Electron Donor (1X)	lb	$ 1.30	8,000	$ 10,400	$ 12,480
KB-1 Culture	L	$ 300	240	$ 72,000	$ 86,400
Injection Field Activities - Electron Donor (Technician and Staff Professional)	day	$ 2,600	20	$ 52,000	$ 62,400
Electron Donor Injection Equipment	week	$ 2,500	3	$ 7,500	$ 9,000
KB-1 Culture Injection (Staff Proffessional)	day	$ 1,500	10	$ 15,000	$ 18,000
FIRST YEAR ELECTRON DONOR/KB-1 INJECTION SUBTOTAL				$ 156,900	**$ 188,280**
TOTAL CAPITAL COSTS (INCL. CONTINGENCY)					**$ 446,280**
ANNUAL OPERATIONS AND MAINTENANCE COST					
EVO Electron Donor (1X)	lb	$ 1.30	2,000	$ 2,600	$ 3,120
Injection Field Activities - Electron Donor (Technician and Staff Professional)	day	$ 2,600	7	$ 18,200	$ 21,840
Electron Donor Injection Equipment	week	$ 2,500	1	$ 2,500	$ 3,000
Performance Monitoring (including sampling and analysis)	sample	$ 300	40	$ 12,000	$ 14,400
Reporting	LS			$ 15,000	$ 18,000
ANNUAL OPERATIONS AND MAINTENANCE COST SUBTOTAL				$ 50,300	**$ 60,360**
ALTERNATIVE 2: COST FOR SOURCE AREA BIOAUGMENTATION TOTAL FOR FIRST YEAR					**$ 506,640**

Table 5-6: Cost Estimate for Alternative 3 (Source Area In Situ Chemical Oxidation**).**

TASK DESCRIPTION	UNIT	UNIT COST	QUANTITY	COST ($)	COST ($) WITH 20% CONTINGENCY
INJECTION/EXTRACTION WELL DRILLING					
Installation of six 6-inch wells (3 shallow/3 deep) for both the injection and extraction wells. Mobilization, development, per diem, decontamination, IDW management, drums included	LS			$ 79,200	$ 95,040
Drilling Oversight (Staff Professional)	day	$ 1,500	12	$ 18,000	$ 21,600
DRILLING SUBTOTAL				$ 97,200	**$ 116,640**
TREATMENT SYSTEM CONSTRUCTION AND STARTUP					
Trenching	LS			$ 50,000	$ 60,000
Piping, instrumentation and process control equipment	LS			$ 25,000	$ 30,000
Construction supervision/oversight (Technician and Staff Professional)	day	$ 2,600	40	$ 104,000	$ 124,800
Startup Testing	day	$ 1,500	10	$ 15,000	$ 18,000
TREATMENT SYSTEM CONSTRUCTION AND STARTUP SUBTOTAL				$ 194,000	**$ 232,800**
YEAR ONE PERMANGANATE INJECTION					
Potassium Permanganate	lbs	$ 2.10	82,000	$ 172,200	$ 206,640
Injection Field Activities (Technician and Staff Professional)	day	$ 2,600	180	$ 468,000	$ 561,600
Oxidant Mixing/Injection Equipment	months	$ 15,000	8	$ 120,000	$ 144,000
YEAR ONE PERMANGANATE INJECTION SUBTOTAL				$ 760,200	**$ 912,240**
TOTAL CAPITAL COSTS (INCL. CONTINGENCY)					**$ 1,261,680**
YEAR TWO PERMANGANATE INJECTION					
Potassium Permanganate	lbs	$ 2.10	82,000	$ 172,200	$ 206,640
Injection Field Activities (Technician and Staff Professional)	day	$ 2,600	160	$ 416,000	$ 499,200
Oxidant Mixing/Injection Equipment	months	$ 15,000	7	$ 105,000	$ 126,000
YEAR TWO PERMANGANATE INJECTION SUBTOTAL				$ 693,200	**$ 831,840**
YEAR THREE CONFIRMATORY MONITORING					
Performance Monitoring (including sampling and analysis)	sample	$ 300	80	$ 24,000	$ 28,800
Reporting	LS			$ 15,000	$ 18,000
YEAR THREE CONFIRMATORY MONITORING SUBTOTAL				$ 39,000	**$ 46,800**
ANNUAL OPERATIONS AND MAINTENANCE COST					
Performance Monitoring (including sampling and analysis)	sample	$ 300	40	$ 12,000	$ 14,400
Reporting	LS			$ 15,000	$ 18,000
ANNUAL OPERATIONS AND MAINTENANCE COST SUBTOTAL				$ 27,000	**$ 32,400**
ALTERNATIVE 3: COST FOR SOURCE AREA ISCO TOTAL FOR FIRST YEAR					**$ 1,294,080**

Table 5-7. Cost Estimate for Alternative 4 (Source Area ISCO/ISB**).**

TASK DESCRIPTION	UNIT	UNIT COST	QUANTITY	COST ($)	COST ($) WITH 20% CONTINGENCY
EXTRACTION WELL DRILLING					
Installation of six 6-inch wells (3 shallow/3 deep) for both the injection and extraction wells. Mobilization, development, per diem, decontamination, IDW management, drums included	LS			$ 79,200	$ 95,040
Drilling Oversight (Staff Professional)	day	$ 1,500	12	$ 18,000	$ 21,600
DRILLING SUBTOTAL				$ 97,200	$ **116,640**
TREATMENT SYSTEM CONSTRUCTION AND STARTUP					
Trenching	LS			$ 50,000	$ 60,000
Piping, instrumentation and process control equipment	LS			$ 25,000	$ 30,000
Construction supervision/oversight (Technician and Staff Professional)	day	$ 2,600	40	$ 104,000	$ 124,800
Startup Testing	day	$ 1,500	10	$ 15,000	$ 18,000
TREATMENT SYSTEM CONSTRUCTION AND STARTUP SUBTOTAL				$ 194,000	$ **232,800**
YEAR ONE PERMANGANATE INJECTION					
Potassium Permanganate	lbs	$ 2.10	82,000	$ 172,200	$ 206,640
Injection Field Activities (Technician and Staff Professional)	day	$ 2,600	180	$ 468,000	$ 561,600
Oxidant Mixing/Injection Equipment	months	$ 15,000	8	$ 120,000	$ 144,000
YEAR ONE PERMANGANATE INJECTION SUBTOTAL				$ 760,200	$ **912,240**
TOTAL CAPITAL COSTS (INCL. CONTINGENCY)					$ **1,261,680**
YEAR TWO PERMANGANATE INJECTION					
Potassium Permanganate	lbs	$ 2.10	82,000	$ 172,200	$ 206,640
Injection Field Activities (Technician and Staff Professional)	day	$ 2,600	160	$ 416,000	$ 499,200
Oxidant Mixing/Injection Equipment	months	$ 15,000	7	$ 105,000	$ 126,000
YEAR TWO PERMANGANATE INJECTION SUBTOTAL				$ 693,200	$ **831,840**
YEAR THREE ELECTRON DONOR AND KB-1 INJECTION					
Direct push drilling for 20 points installed to 80 feet	day	$ 1,500	15	$ 22,500	$ 27,000
Drilling Oversight (Staff Professional)	day	$ 1,500	15	$ 22,500	$ 27,000
EVO Electron Donor (4X demand exerted by sulfate and 15% of TCE, 1X Mn)	lb	$ 1.30	5,100	$ 6,630	$ 7,956
KB-1 Culture	L	$ 300	300	$ 90,000	$ 108,000
Injection Field Activities - Electron Donor (Technician and Staff Professional)	day	$ 2,600	15	$ 39,000	$ 46,800
Electron Donor Injection Equipment	week	$ 2,500	2	$ 5,000	$ 6,000
KB-1 Culture Injection (Staff Proffessional)	day	$ 1,500	12	$ 18,000	$ 21,600
YEAR THREE ELECTRON DONOR/KB-1 INJECTION SUBTOTAL				$ 203,630	$ **244,356**
ANNUAL OPERATIONS AND MAINTENANCE COST					
Direct push drilling for 10 points installed to 80 feet	day	$ 1,500	7	$ 10,500	$ 12,600
Drilling Oversight (Staff Professional)	day	$ 1,500	7	$ 10,500	$ 12,600
EVO Electron Donor (1X demand for sulfate and 15% of TCE)	lb	$ 1.30	253	$ 329	$ 395
Injection Field Activities - Electron Donor (Technician and Staff Professional)	day	$ 2,600	7	$ 18,200	$ 21,840
Electron Donor Injection Equipment	week	$ 2,500	1	$ 2,500	$ 3,000
Performance Monitoring (including sampling and analysis)	sample	$ 300	40	$ 12,000	$ 14,400
Reporting	LS			$ 15,000	$ 18,000
ANNUAL OPERATIONS AND MAINTENANCE COST SUBTOTAL				$ 69,029	$ **82,835**
ALTERNATIVE 4: COST FOR SOURCE AREA ISCO COUPLED WITH BIOAUGMENTATION					$ **1,344,515**

6.0 Implementation Issues

This section describes all applicable or relevant regulatory requirements related to the demonstration. These requirements include the acquisition of permits and the compliance with regulations. The necessary permitting and compliance issues are described below.

- Approval from Local and State Authorities to Release Microbial Consortium.
- CCAFB assisted in obtaining the necessary approvals for the release of a natural consortium of microorganisms into the PTA.
- Approval for the purchase and use of tax-free ethanol
- Geosyntec submitted a permit application from the US Bureau of Alcohol, Tobacco and Firearms for the use of denatured ethanol as an electron donor for the biostimulation and bioaugmentation phases of the demonstration.

Sequential application of ISCO and ISB is potentially widely applicable at chlorinated solvent sites throughout North America. However, several issues may potentially limit the widespread application of this technology. In the long term, ISCO application is likely to increase the concentration of manganese in groundwater, a potentially adverse geochemical impact. The capital cost associated with implementing two source control technologies (ISCO & ISB) may be a barrier to implementation. However, implementing these technologies sequentially may provide substantial schedule advantages over the implementation of either technology alone, off-setting the increased capital costs with reduced O&M costs. The uncertainty surrounding the performance of this technology is another barrier, particularly the performance of ISCO at full-scale. The completion of this demonstration and publication of the results in both peer-reviewed and other technical literature will provide site managers with an improved degree of certainty when assessing either the sequential technology or ISB as a stand-alone technology.

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8.0 Points of Contact

A summary of contact information for all personnel associated with this demonstration project is presented below.

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