## TREATABILITY STUDY FOR

## HILL AFB'S OPERABLE UNIT-1:

# ENHANCED MICROAEROBIC DECHLORINATION

## USING VARIOUS ELECTRON DONORS

by

Peter G. Breed

A report submitted in partial fulfillment of the requirements for the degree

of

MASTER OF SCIENCE

in

**Environmental Engineering** 

Approved:

Michael J. McFarland, Major Professor

Sorensen, Committee Member

Daniel Stone, Committee Member

UTAH STATE UNIVERSITY Logan, Utah

1999

## **DISTRIBUTION STATEMENT A** Approved for Public Release

Distribution Unlimited

DTIC QUALITY INSPECTED 4

REPORT	Form Approved OMB No. 0704-0188		
Public reporting burden for this collection of im gathering and maintaining the data needed, an collection of information, including suggestions Davis Highway, Suite 1204, Arlington, VA 222	formation is estimated to average 1 hour pe d completing and reviewing the collection or for reducing this burden, to Washington He 202-4302, and to the Office of Management	r response, including the time for re f information. Send comments reg eadquarters Services, Directorate fo and Budget, Paperwork Reduction	eviewing instructions, searching existing data sources, arding this burden estimate or any other aspect of this or Information Operations and Reports, 1215 Jeffersor Project (0704-0188), Washington, DC 20503.
1. AGENCY USE ONLY (Leave blai		3. REPORT TYPE AN	DATES COVERED
	13.May.99		MAJOR REPORT
4. TITLE AND SUBTITLE TREATABILITY STUDY FOR MICROAEROBIC DECHLORI			5. FUNDING NUMBERS
6. AUTHOR(S) CAPT BREED PETER G			
7. PERFORMING ORGANIZATION UTAH STATE UNIVERSITY	NAME(S) AND ADDRESS(ES)		8. PERFORMING ORGANIZATION REPORT NUMBER
9. SPONSORING/MONITORING AC THE DEPARTMENT OF THE		ES)	10. SPONSORING/MONITORING AGENCY REPORT NUMBER
AFIT/CIA, BLDG 125			FY99-103
2950 P STREET			1.139-105
WPAFB OH 45433			
11. SUPPLEMENTARY NOTES			
12a. DISTRIBUTION AVAILABILITY	STATEMENT		12b. DISTRIBUTION CODE
Unlimited distribution			
In Accordance With AFI 35-205	/AFIT Sup 1		
13. ABSTRACT (Maximum 200 wo	rds)		
14. SUBJECT TERMS		· · · · · · · · · · · · · · · · · · ·	15. NUMBER OF PAGES
14. SUDJECT TERMIS			96
		_	16. PRICE CODE
17. SECURITY CLASSIFICATION OF REPORT	18. SECURITY CLASSIFICATION OF THIS PAGE	19. SECURITY CLASSIF OF ABSTRACT	ICATION 20. LIMITATION OF ABSTRACT
	· · · · · · · · · · · · · · · · · · ·		Standard Form 298 (Rev. 2-89) (EG

Prescribed by ANSI Std. 239.18 Designed using Perform Pro, WHS/DIOR, Oct 94

#### ACKNOWLEDGMENTS

This project and all its parts are the result of many people working toward a common goal. If not for the efforts of a wide range of people, I would not have been able to complete this research and this report. Though there are too many to individually thank for all the bits of help and guidance, I would like to express my thanks to some of the more critical team members.

My sincere gratitude goes to the United States Air Force Institute of Technology for providing me the opportunity and the time to pursue this challenge and see it through to completion. Through their support and resources, I have developed an understanding of technical issues essential to remediating some of the Air Force's current environmental problems and minimizing future environmental impacts as we move into the 21<sup>st</sup> century.

If not for the time, effort, and guidance of my committee, this project would never have gotten off the ground, let alone completed. To Dr. Michael McFarland, Dr. Darwin Sorensen, and Dr. Daniel Stone, thank you very much for sharing your wisdom and appreciating the time constraints of an academic schedule established by the United States Air Force. Special thanks are also due to Dr. Daniel Smith for the conception, development, and initiation of this project; without his help in establishing fundamental building blocks, this project would have been far beyond my grasp.

Thank you also to the Hill AFB Environmental Restoration Division without who's financial support, this project would not have been completed. Individual thanks are due to Mr. Robert Elliot for providing the equipment and laboratory support and Dr. Jon Ginn for technical guidance as well as facilitating many of the issues associated with Operable Unit 1 research. To the team at Hill AFB's analytical laboratory, I also extend my thanks. Ms. Diane Luke, Ms. Linda Garner, and Ms. Jody Roper were always a pleasure to work with and consistently offered much needed assistance.

To other faculty members that I pestered with questions throughout my course of study I extend my gratitude, particularly Ms. Joan E. McLean and Dr. William Doucette. A special thanks to my fellow students without whose camaraderie, I may have lost my mind.

The largest individual piece of credit goes to Jenny for helping maintain my focus while offering never ending support. Her encouragement, patience, and support particularly during the most frustrating times cannot go unrecognized. Thanks you to my parents and family for making me who I am and instilling the desire to tackle each challenge and persevere to its completion.

A special thanks is owed to GOD for guiding me through the challenges associated with this project and simply that it is finished.

ACKOWLEDGEMENTS	
LIST OF TABLES	vi
LIST OF FIGURES	vii
ABSTRACT	viii
CHAPTER	
I. INTRODUCTION	1
II. OBJECTIVES	4
III. LITERATURE REVIEW	5
Chloroethene Reduction Dechlorinating Microorganisms Electron Donor Studies	7
Reduction of Tetrachloroethylene to Dichloroethene Reduction of Tetrachloroethylene/Dichloroethene to Ethene and Ethane .	10 12
IV. EXPERIMENTAL APPROACH	19
Site History and Characteristics Experimental Design Materials and Methods	22
Soil Collection Aquifer Sampling Donor Selection Microcosm Configuration and Assembly Analytical Methods	24 25 26
V. RESULTS AND DISCUSSION	33
Chloroethene Dechlorination Efficiencies in Laboratory Columns	33
Chloroethene Degradation in the Absence of Additives Chloroethene Degradation in the Presence of Vitamins and Yeast Columns Amended with Electron Donors Chloroethene Degradation in the Presence of n-Butyric Acid Chloroethene Degradation in the Presence of Benzoic Acid	. 35 . 35 . 36

## TABLE OF CONTENTS

Chloroethene Degradation in the Presence of Lactic Acid	37
Chloroethene Degradation in the Presence of Propionic Acid	37
Chloroethene Degradation in the Presence of n-Propanol	38
Chloroethene Degradation in the Presence of Toluene	38
Comparison of Dechlorination Efficiencies	39
Comparison of Background Columns; Unamended versus Amended	39
Comparison of Different Donor Dechlorination Efficiencies	40
Comparison of Donor Dechlorination Efficiencies to Background	
Columns	41
Discussion of Dechlorination Efficiencies	42
Prediction of Field Dechlorination Values	43
Other Reactions of Interest	44
VI. CONCLUSIONS	45
Conclusions Directly Related To Study Objectives	45
Supplemental Study Conclusions	46
VII. RECOMMENDATIONS	. 48
REFERENCES	51
	57
APPENDIXES	, 51
Appendix A. Soil Respirometry Test Procedures and Results	58
Appendix B. Electron Donor Equations and Hydrogen Release Calculations	. 64
Appendix D. Electron Donor Delivery Calculations	. 65
Appendix D. Hydrogen Demand Calculations	. 66
Appendix E. Electron Donor Properties and Actual Delivery Calculations	. 67
Appendix F. Material and Chemical Inventory	. 69
Appendix G. Column Flow Calculations and Microcosm HRTs	. 71
Appendix H. Analytical Results Spreadsheet, Removal Efficiencies and Rates	
Appendix I. Chloride Calculations and Results	. 95
· PP · · · · · · · · · · · · · · · · ·	

## LIST OF TABLES

Table		Page
1.1	Chloroethene Properties and MCLs	2
3.1	Electron Acceptor Reduction Half Reactions	
3.2	Past Research Identifying Anaerobic Dechlorinating Microorganisms	9
3.3	Studies Demonstrating Reduction of PCE to DCE	11
3.4	Studies Demonstrating Reduction of PCE or DCE to Ethene	15
4.1	Intrinsic Bioremediation Indicators for Hill AFB, OU-1	20
4.2	Electron Donor Oxidation Half Reactions	25
4.3	Vitamin Concentrations Used in Microcosm Study	27
4.4	Electron Donor Supply Values	
4.5	Analytical Parameters	
5.1	cis-Dichloroethene Percentage Removal Efficiencies	
5.2	Vinyl Chloride Percentage Apparent Removal Efficiencies	

## LIST OF FIGURES

Figure	I	Page
3.1	Reductive Dechlorination Model	5
4.1	Hill AFB Operable Unit 1 Site Map	21
4.2	Single Microcosm Schematic	
4.3	Full Microcosm System Schematic	
5.1	Chloroethene Removal Efficiencies for the Unamended Column	
5.2	Chloroethene Removal Efficiencies for the Amended Column	35
5.3	Chloroethene Removal Efficiencies for the n-Butyric Acid/Amended Column	36
5.4	Chloroethene Removal Efficiencies for the Benzoic Acid/Amended Column	36
5.5	Chloroethene Removal Efficiencies for the Lactic Acid/Amended Column	37
5.6	Chloroethene Removal Efficiencies for the Propionic Acid/Amended Column	37
5.7	Chloroethene Removal Efficiencies for the n-Propanol/Amended Column	38
5.8	Chloroethene Removal Efficiencies for the Toluene/Amended Column	38

#### ABSTRACT

A treatability study of the microaerobic biodegradation of *cis*-dichloroethene (c-DCE) was completed using a series of eight continuously operated columns filled with contaminated soils from Hill Air Force Base's Operable Unit 1. Columns were supplied groundwater from the site, vitamins and yeast, and an electron donor solution containing one of the following donors: n-butyric acid, benzoic acid, lactic acid, propionic acid, n-propanol, or toluene. Concentrations of c-DCE varied over six months and ranged from 2736  $\mu$ g/L to 30  $\mu$ g/L. Though attempted as an anaerobic study, the ability to continuously eliminate oxygen from an active system proved difficult and columns operated as microaerobic systems.

In all columns the degradation of c-DCE was observed, however, the removal efficiencies determined by comparing the influent and effluent concentrations were highly inconsistent throughout the experiment. By comparing the background columns to the columns supplied electron donors, it does not appear the addition of vitamins or electron donors enhance the indigenous microorganism's ability to remove c-DCE. While c-DCE removal within the background column averaged 17%, the vitamin amended control column averaged only 7% c-DCE removal within the column and the electron donor supplied columns averaged between 7% removal and 5% apparent production. Of the electron donors supporting c-DCE removal, benzoic acid demonstrated 7% removal followed closely by propionic acid and n-propanol, both showing 5% c-DCE removal.

The accumulation of vinyl chloride (VC) was initially noted in all columns, but rapidly declined until typical operating conditions showed persistent and complete removal of VC. Ethene removal appeared in all columns and was typically an order of magnitude greater in columns provided with an electron donor. Methanogenesis was apparent in all columns with methane production in the vitamin and electron donor columns being two to five times greater that the unamended control column.

This research identified the critical need to determine in situ limitations before enhanced bioremediation is attempted. The lower threshold concentration of the contaminant of concern and the acclimation period for indigenous microorganisms must be adequately defined before remediation predictions or field applications can be accomplished.

#### CHAPTER I

#### INTRODUCTION

The contamination of soils and groundwater by waste solvents such as the chlorinated ethenes tetrachloroethene, trichloroethene, dichloroethene, and vinyl chloride (PCE, TCE, DCE, and VC respectively) is of significant environmental concern. These chlorinated aliphatic hydrocarbons (CAHs) have been widely used as solvents in many industries including the aerospace industry. Their subsequent release and disposal has commonly resulted in contamination of the groundwater and soil. Due to the toxicity of these compounds and their known or potential carcinogenic affects, the U.S. Environmental Protection Agency has listed many of them as priority pollutants (40CFR 141). As a result of the persistence and toxicity of some CAHs, natural attenuation, which occurs at some sites, may not be adequate to protect human health and the environment. At many contaminated sites, some form of active or enhanced remediation should be considered as a more rapid option for site clean up.

The area known as Operable Unit 1 (OU-1) located on Hill Air Force Base, Utah, contains significant levels of CAHs and is characterized by high levels of PCE and TCE daughter compounds, particularly the *cis* isomer of DCE (c-DCE). Past site investigations completed by the Air Force show worst-case groundwater contaminant levels of PCE at 58 micrograms per liter (µg/L), TCE at 2,300 µg/L, total DCE dominated by the *cis* isomer but including the *trans* isomer (t-DCE) at 42,000 µg/L and VC at 2,400 µg/L (Montgomery Watson, 1995). These levels represent source area concentrations and are much higher than average concentrations encountered in OU-1 groundwater. Any clean-up strategy for OU-1 groundwater will be dominated by the treatment of DCE and VC with very little PCE and TCE present. The average concentration of c-DCE to be treated would likely be 1,000 µg/L or less. As a result of known or suspected health affects from these compounds, the EPA has established drinking water Maximum Contaminant Levels (MCLs) for each of them. These MCLs and some other physical and chemical properties of concern are listed in Table 1.1.

Reductive dehalogenation of CAHs has become widely recognized as a technology with great potential for in situ remediation. Current work by the Department of Defense shows that Reductive Anaerobic Biological In Situ Treatment Technology (RABITT) is one of the most promising in situ treatment technologies for chloroethenes (Morse *et al.* 1997). The more highly chlorinated ethenes PCE and TCE resist aerobic degradation while the lesserchlorinated compounds of DCE and VC are readily degraded by a variety of aerobic microorganisms. It has been demonstrated, however, that all chloroethenes can be degraded under anaerobic conditions (Freedman and Gossett 1989, DiStefano *et al.* 1991 and 1992, deBruin *et al.* 1992, Carter and Jewell 1992, Beeman *et al.* 1994, Fennell and Gossett 1987, Smatlak 1996, and Yager 1997). Research has discovered that if the proper electron donor is available, anaerobic or microaerobic microbes, possibly indigenous to a contamination site, can completely dechlorinate these compounds into innocuous byproducts. However, if the site does not have an adequate microbial population or an adequate source of electron donors, PCE and TCE may persist or reductive dechlorination may proceed only as far as the intermediate daughter products, specifically the DCEs and VC. For remediation to be considered complete, these intermediates must also be dechlorinated to ethene (ETH) or ethane.

Table 1.1. Chlo	oroethene P	roperties and	I MCLs			
Contaminant	MW <sup>(a)</sup> (g/mole)	Vapor Pressure (mm Hg) @25°C	Density (g/mL)	Solubility (mg/L) @25°C	OU-1 <sup>(b)</sup> Maximum Concentration (µg/L)	MCL <sup>(c)</sup> (µg/L)
PCE	165.82	19	1.623	150	52	5.0
TCE	131.38	77	1.465	1100	2300	5.0
1,1-DCE	96.94	591	1.213	2250	N/D	7.0
cis-DCE*	96.94	206	1.282	3500	42000 (Total)	70.0
trans-DCE*	96.94	331	1.255	6300	9.3 Estimated	100.0
VC	62.50	760(STP)	0.911	2700	2400 μg/L	2.0
Ethene	28.05	760(STP)	1.260	131		-

(a) Perry's Chemical Engineer's Handbook, 7th Edition

(b) Montgomery-Watson, 1995

(c) EPA Drinking Water Regulations and Health Advisories, October 1996

\* The cis and trans isomers of 1,2 DCE can be analytically distinguished, however, sample results show only trace levels of trans therefore, they are discussed as a portion of the Total DCE

The goal of this research project was to show that enhanced anaerobic or microaerobic bioremediation, facilitated by electron donor addition, is a viable alternative to the pump and treat methods currently being proposed for Hill AFB's OU-1 groundwater contamination. This project successfully demonstrated partial removal of the primary contaminant, c-DCE,

without the accumulation of VC, however the best removal efficiencies were in the background column that was not provided an electron donor. Though this experiment did not clearly demonstrate that electron-donor-enhanced in situ bioremediation is a more cost-effective alternative than the proposed pump and treat methods, further research should be completed before this technology is disregarded.

#### CHAPTER II

#### **RESEARCH OBJECTIVES**

The overall objective of this study was to determine if in-situ enhanced reductive dechlorination is a viable alternative to pump-and-treat technologies currently proposed for the remediation of Operable Unit 1. The focus of this research was on stimulating indigenous microorganisms to reductively dechlorinate the relatively high levels of dichloroethene contamination. Stimulation of these organisms was attempted by supplying nutrients and additional substrate in the form of electron donors. To be completely successful, enhanced reductive dechlorination had to lower the level of all chlorinated ethenes to below regulatory standards. The specific research objectives were to:

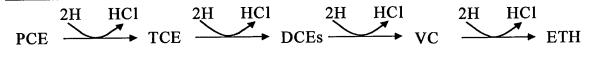
- 1. Demonstrate that complete reductive dechlorination of *cis*-dichloroethene without an accumulation of vinyl chloride is possible under anaerobic or microaerobic conditions using OU-1 soil and groundwater.
- 2. Compare the suitability of various electron donors and determine the most promising electron donor(s) for maintaining reductive dechlorination under OU-1 site conditions.
- 3. Determine if vitamin and yeast amendments are necessary for complete reductive dechlorination in OU-1.
- 4. Demonstrate a cost-effective alternative for remediation of chlorinated ethenes at OU-1.

#### CHAPTER III

#### TREATMENT STUDIES

Past studies have demonstrated that chlorinated solvents can be reductively dechlorinated to ethene and ethane under anaerobic conditions. Successfully accomplishing complete reduction to these innocuous compounds is dependent on at least two key parameters: an adequate microbial population capable of completing this reduction and a sufficient supply of electron donors. Various researchers have focused on these two key variables and have successfully identified microorganisms and electron donors that support reductive dechlorination. (See Tables 3.2-3.4).

The reductive dechlorination model shown in Figure 3.1 is accepted as the general anaerobic transformation pattern for chloroethenes. In the previous studies reviewed, either a portion of the reductive dechlorination sequence model (e.g. TCE  $\rightarrow$  DCE) or the complete



### Figure 3.1. Reductive Dechlorination Model

transformation model (e.g. PCE  $\rightarrow$  ETH) is demonstrated. Most studies and typical field sites demonstrate that PCE and TCE are the initial sources of contamination while DCE and VC are shown to be daughter products of their reduction. In general DCE, dominated by the *cis* isomer, is the most persistent daughter product found at field sites. This may be the result of the much slower kinetics for the DCE  $\rightarrow$  VC step than the TCE  $\rightarrow$  DCE step, but may also be due to the exhaustion of available electron donor supply. Historically, microcosm studies examined c-DCE reduction as an intermediate step in the complete reduction of PCE or TCE to ETH. More recently, studies began to target c-DCE reduction specifically and the observations support this project and suggest a great potential for stimulating c-DCE reduction by supplying adequate electron donor supplements (Yang and McCarty, 1998 and Windfuhr 1998).

#### CHLOROETHENE REDUCTION

Organic compounds have been biotransformed through three identified processes: (1) as an electron donor in energy metabolism, (2) cometabolism, and (3) as an electron acceptor in energy metabolism (Adriaens and Vogel, 1995, Wackett 1995, and McCarty 1998). Under anaerobic conditions, reductive dehalogenation is the dominant mechanism for halogen removal (Mohn and Tiedji, 1992) and until recently, it was believed that all dechlorination of chloroethenes was a cometabolic process occurring as a beneficial result of other dominant electron receptor reactions. In these fortuitous reactions, the chloroethene is reduced, but the microorganisms receive no energy from the reaction. Recently, research has elucidated the microbial process called halorespiration (Hollinger and Schumacher, 1994) in which chloroethenes are used as respiratory electron acceptors and support metabolism which provides organisms with energy for growth and maintenance. As metabolism proceeds, electrons are transferred from donors to the chloroethenes in a manner that substitutes a hydrogen atom for a chlorine atom. In short, if energy is obtained directly from the dechlorination the process it is called halorespiration and if no energy is obtained it is reductive dehalogenation (McCarty, 1998). Synthesis of new cells from the carbon available in chloroethenes apparently does not occur in either cometabolism or halorespiration reactions. The carbon source for synthesis is not well understood.

The reduction half reactions defined in Table 3.1 show the relative energetic favorability of common environmental electron acceptors and chloroethenes. The Gibb's free energy

-	
Half Reaction	$\Delta G^{o}$
	(kJ/e equiv)
Oxygen	
$0.25 O_2 + H^+ + e^- = 0.5 H_2 O$	-78.14
Nitrate	
$0.2 \text{ NO}_3^- + 1.2 \text{ H}^+ + \text{e}^- = 0.1 \text{ N}_2 + 0.6 \text{ H}_2\text{O}$	-71.67
РСЕ	
$0.5 \text{ CCl}_2\text{CCl}_2 + 0.5 \text{ H}^+ + \text{e}^- = 0.5 \text{ CHClCCl}_2 + 0.5 \text{ Cl}^-$	-53.31
ТСЕ	
$0.5 \text{ CHClCCl}_2 + 0.5 \text{ H}^+ + \text{e}^- = 0.5 \text{ CHClCHCl} + 0.5 \text{ Cl}^-$	-52.11
DCE	
$0.5 \text{ CHClCHCl} + 0.5 \text{ H}^{+} + e^{-} = 0.5 \text{ CH}_2 \text{CHCl} + 0.5 \text{ CI}^{-}$	-42.12
VC	
$0.5 \text{ CH}_2\text{CHCl} + 0.5 \text{ H}^+ + \text{e}^- = 0.5 \text{ CH}_2\text{CH}_2 + 0.5 \text{ CI}^-$	-45.22
Sulfate	
$0.125 \text{ SO}_4^{2-} + 1.188 \text{ H}^+ + \text{e}^- = 0.063 \text{ H}_2\text{S} + 0.063 \text{ HS}^- + 0.5 \text{ H}_2\text{O}$	21.27
Methane Fermentation	
$0.125 \text{ CO}_2 + \text{H}^+ + \text{e}^- = 0.125 \text{ CH}_4 + 0.25 \text{ H}_2\text{O}$	24.11

Table 3.1. Electron Acceptor Reduction Half Reactions

 $(\Delta G^{\circ})$  values in Table 3.1 show that oxygen and nitrate reductions are more energetically favorable than chloroethene reduction. The table also shows chloroethene reduction is more favorable than sulfate reduction or methane fermentation and that VC reduction to ETH is energetically more favorable than c-DCE reduction to VC. Therefore, based on energetics we would expect c-DCE to be the rate limiting step in anaerobic reductive dechlorination and dechlorination would be expected to be inhibited where oxygen or nitrate are in abundance or possibly where sulfate concentrations are high.

### DECHLORINATING MICROORGANISMS

Since 1981, we have known that the potential for the biological transformation of chlorinated aliphatic hydrocarbons (CAHs) is possible under anaerobic conditions (Bouwer et al. 1981). Since that time a great deal of work has been completed to isolate microorganisms capable of degrading these CAHs. Though it has often been found that a consortium of microorganisms is required for complete dehalogenation, in some cases specific microorganisms have been identified.

Microcosm studies have shown that organisms frequently found in natural soils have the ability to dechlorinate solvents. In 1987, Fatherpure *et al.* completed a study comparing nine pure cultures of anaerobes and identified *Methanosarcina* sp., *Methanosarcina mazei*, and dechlorinating bacterium DCB-1 as significant dechlorinators. In batch studies, these organisms reduced 1 mg/L of PCE to TCE within one week. The DCB-1 demonstrated the highest rate of reduction and was approximately three times faster than *Methanosarcina* sp. and five times faster than *Methanosarcina mazei*. A subsequent study (Fatherpure and Boyd, 1988) showed that *Methanosarcina* sp. strain DCM could also reduce 1-3 mg/L of PCE to TCE within one week under methanogenic conditions. A direct relationship between microbial concentration and the transformation of PCE was identified. In this test some samples were autoclaved to demonstrate the critical nature of microbial activity in the reduction of the contaminant. No dechlorination occurred in the autoclaved microcosms. Also demonstrated in this study was the dependence of microorganisms on an adequate supply of electron donors.

Hollinger *et al.* (1993) demonstrated the reductive capacity of an anaerobic bacterium by reducing 200  $\mu$ M (3.32mg/L) of PCE using an organism called PER-K23. PER-K23

completely reduced the PCE within 33 days. Of particular interest was the production of ethene, previously thought to require aerobic conditions. Also of interest was the PER-K23's dependence on PCE without which the bacterium ceased to grow. This marks the first documented case of microorganisms using PCE as electron acceptors in energy metabolism i.e., halorespiration.

In 1996 Gossett and Zinder presented a list of direct dechlorinators capable of dechlorinating PCE and TCE to c-DCE. This list included *Dehalobacter restrictus, Dehalospirillium multivorans*, Strain TT4B, *Enterobacter agglomerans*, and *Desulfobacterium* sp strain PCE1 (Hollinger 1992, Neumann *et al.*, 1994, Krumholz 1995, Sharma and McCarty 1996 and Gerritse *et al.*, 1996). Expanding the menu of microbial direct dechlorinators, Sharma and McCarty (1996) demonstrated the ability of a facultative aerobic bacterium, *Enterobacteriaceae* strain MS-1, to transform 1mM (165 mg/L) of PCE to c-DCE in less than 12 days. Due to the nature of this organism, the energetically preferred oxygen was depleted before the PCE was used as an electron acceptor and for energy metabolism. Column studies containing this organism and PCE contaminated aquifer soils showed complete reduction to ethene when benzoate and sulfate were added. The MS-1 did not use the sulfate and benzoate directly, but used the benzoate oxidation products, acetate and formate, for PCE dehalogenation. Later work with this microorganism demonstrated that it could be successfully used for bioaugmentation in a fixed film reactor and successfully dechlorinate PCE and TCE to ethene (Newberg *et al.*, 1997).

Reported in 1997, an eubacterium had been isolated from an enrichment culture and shown to be capable of completely dechlorinating PCE to ethene (Maymo-Gatell *et al.*, 1997). This microorganism, *Dehalococcus ethenogenes*, strain 195, was capable of sustaining dechlorination while using hydrogen as the sole electron donor. Methanol, ethanol, pyruvate, glucose, formate, acetate, lactate and yeast extract were also tested as electron donors, but none were utilized by the microorganism. PCE, TCE, c-DCE, 1,1-DCE, 1,2-DCA and 1,2-dibromomethane (DBM) were all shown to support growth of the microorganism while t-DCE and VC did not support growth but were also converted to ethene. This work is remarkable in that it identifies an isolated organism capable of completely dechlorinating PCE to ethene and it demonstrates that in many steps of the process the CAHs serve as the electron acceptors and support microbial growth. Though we

have made progress in identifying specific dechlorinators, the complete destruction of CAHs under microaerobic or anaerobic in-situ conditions likely still requires a consortia of microorganisms working together (McCarty, 1998).

Test	Dechlorination	g Anaerobic Dechlorinating M Organism; Metabolic	Conclusions	Reference
	Range	Group (Conditions)		
Test Method	1 mg/L PCE	-DCB-1; Methanogen	Dechlorination	Fatherpure
not specified	for each	(0.2% Pyruvate, 1mM 3-	for DCB-1 was	et al.,
but assumed	different test	Chloro-benzoate)	3-5 times faster	1987
to be 50 mL		-Methanosarcina sp. or	than the others;	
of medium in		-Methanosarcina mazei;	Complete	
160 mL		Methanogen (25mM	reduction to TCE	
bottles		Methanol, PREM)	in 1 week	
Microcosms:	1-3 mg/L PCE	Methanosarcina sp. Strain	PCE to TCE/CH4	Fatherpure
50 mL of	C	DCM; Methanogen	in 1 week;	and Boyd,
medium in		(25mM Methanol, PREM,	Controls showed	1988
160 mL		50mM Sodium Acetate)	no reduction	
bottles				
Microcosms:	Up to 200 µM	PER-K23; Unknown	Degradation	Hollinger
200 mL of	of PCE	(H <sub>2</sub> or formate in Rhine	products	et al.,
medium in		River Sediment with	including ETH in	1993
500 mL		anaerobic sludge, yeast	33 days; No	
bottles		extract, vitamin solution)	growth in the	
			absence of PCE	
Test Method	1 mM PCE	Enterobacteriaceae Strain	Dechlorination of	Sharma
not specified	1 1111	MS-1; Facultative Aerobe	PCE to c-DCE in	and
notopounce		(glucose, lactate, pyruvate,	4-12 days at room	McCarty,
		yeast, formate, amino acid	temp	1996
		or acetate in basal and		
		vitamin solutions)		
12-L	PCE	Enterobacteriaceae Strain	Dechlorination of	Newberg
anaerobic	unspecified	MS-1; Facultative Aerobe	>95% TCE to c-	<i>et al.</i> ,
fixed film	1-2 mg/L TCE	(yeast extract and sodium benzoate)	DCE in 5 days	1997
reactor				
Test Method	Not Specified	Dehalococcus	Dechlorination of	Maymo- Gatell, <i>et</i>
not specified		<i>ethenogenes</i> strain 195 Eubacterium	PCE and all intermediates to	<i>al.</i> , 1997
		(hydrogen, acetate, B12,	Ethene	u., 1771
		anaerobic supernatant)		

These studies demonstrate the critical role microorganisms play in the reduction of chloroethenes. They also show that under anaerobic conditions, all species of chloroethenes can ultimately be reduced to ethene and subsequently ethane. The results of these studies support the potential use of anaerobic, in situ reductive dechlorination at CAH contaminated sites, but they also demonstrate the demand for an adequate supply of electron donors and perhaps other nutrients if the microbial population is to effectively dechlorinate the contaminants.

#### ELECTRON DONOR STUDIES

Site characteristics and indigenous microbial populations vary from site to site; therefore, the identification of an appropriate electron donor and needed nutrients is critical for successful in situ reductive dechlorination. A substantial amount of research has been conducted to define the effectiveness of various electron donors. It has been understood for some time that dechlorination of PCE to TCE proceeds under strictly anaerobic conditions while the dechlorination of TCE to DCE will proceed under anaerobic conditions or aerobic conditions. A review of anaerobic studies demonstrating the degradation of PCE to DCE was completed to provide some insight to what has occurred historically at Hill AFB OU-1. A brief summary of some of these studies is presented in Table 3.3. More recently, research has shown that the continued dechlorination of DCE to VC and ethene can also progress under anaerobic conditions and is not limited to the energetically favorable aerobic conditions as once believed (McCarty, 1998). Table 3.4 focuses on some of these studies and shows complete anaerobic dechlorination in a variety of experiments. Results of these studies and shows complete anaerobic dechlorination in a variety of experiments. Results of these studies and shows complete anaerobic dechlorination in a variety of experiments.

#### Reduction of Tetrachloroethene to Dichloroethene

Sealed microcosm studies have been the principal method of research used to identify electron donors that facilitate dechlorination. In a comparative study of eight electron donors (Gibson & Sewell, 1992), microcosms were prepared using 10 grams of contaminated soils from a Coast Guard Station and inoculated with 30  $\mu$ M of PCE. Lactate and ethanol supported production of TCE within six days and DCE after ten days. Butyrate, crotonate,

10

and propionate had lag times of ten to fifteen days but also showed production of TCE and DCE. Acetate, methanol, and isopropanol did not support dechlorination at a rate any higher than the unamended controls. These observations indicate there can be significant differences in dechlorination results depending on the electron donor selected.

Table 3.3. Studies De		action of PCE to I	DCE.	D.C.
Test	Source	Successful	Conclusions	Reference
	Concentrations	Donors		<u> </u>
Microcosms: 10 g sediment in 20 mL bottles, headspace filled with amended solutions	Soil from USCG site with 30 µM PCE	Lactate, Propionate, Crotonate, Butyrate, Ethanol	PCE reduction started within 1 week under methanogenic conditions	Gibson and Sewell, 1992
Microcosms: 50 mL of reactor biomass culture in 100 mL bottles	2.8 mM PCE	Benzoate with protein from biofilm reactor	Conversion to c- DCE completed w/o methanogenesis	Scholz- Muramatsu, en al, 1989
Microcosms: 100 mL of digester medium in 160 mL bottles	600 μg/L PCE in anaerobic digester sludge	Lactate with vitamin solution	92% PCE reduction to c-DCE in 13 days under sulfate reducing conditions	Bagley and Gossett, 1990
Microcosms: 25 mL bottles filled with contaminated soil, voids filled with amended solution	Soils from photocopier refurbishing facility: concentrations not specified	Ethanol, Acetate, or Lactate with nitrate, sulfate, or yeast amendments	99% PCE reduction to c-DCE in 200 days under nitrate and sulfate reducing and methanogenic conditions: anoxic and anaerobic	Pavlostathis and Zhuang, 1993
Microcosms: 6 g sediment and 60 mL of medium in 124 mL bottles	Soil from Tinker AFB and Victoria TX spiked to 9 µM PCE	Methanol, formate, lactate, acetate, or sucrose	Up to 70% conversion to TCE and c-DCE to 200 days under various metabolic conditions	Gao, <i>et al</i> , 1997

Scholz-Muramatsu *et al*, (1989) used 50 mL of biomass culture from a biofilm reactor fed with benzoate. Benzoate was added to the 100 mL microcosms as the sole energy source and 2.8 mM PCE was added as the inoculum. Methanogenesis was selectively inhibited with bromoethane-sulfonic acid. c-DCE was the only transformation product measured in this test and formed in nearly the same concentrations in cultures with and without methanogenesis. The uptake of benzoate was directly proportional to the c-DCE formed. In a similar study using anaerobic sludge (Bagley and Gossett, 1990), 600 µg/L of PCE was reduced to c-DCE in thirteen days. Lactate was used as a donor and methanogenic inhibitors were applied. Results showed reductive dechlorination of PCE proceeds under sulfate reducing conditions. Reductive dechlorination under both sulfate reducing and methanogenic conditions was also demonstrated in a study using anoxic/anaerobic field-contaminated soils containing PCE, TCE, and DCE (Pavostathis and Zhuang, 1993). Without electron donors the reductive dechlorination proceeded. Within 200 days, 99% of the PCE and TCE in the soils was reduced to c-DCE. Nitrate reducing conditions were also evaluated and did support dechlorination but at a much lower rate.

Multiple donor experiments were conducted and demonstrated PCE reduction while various levels of sulfate reducing, acetogenic, fermentative, and methanogenic activity was observed (Gao *et al*, 1997). Subsurface soils were collected from contaminated sites and tested with methanol, formate, lactate, acetate, and sucrose as donors. All substrates supported dechlorination to TCE and c-DCE, however, the lactate amended microcosm showed the most significant reduction of PCE. Though the results of this study do not demonstrate consistent dehalogenation rates, they do demonstrate the ability of indigenous microorganisms to degrade chloroethenes using a variety of anaerobic metabolisms. Collectively, these studies demonstrate the ability of microorganisms to anaerobically reduce PCE under a variety of metabolic conditions but all demonstrate the demand for an appropriate donor.

## Reduction of Tetrachloroethene/Dichloroethene to Ethene and Ethane

Many researchers have demonstrated complete anaerobic reduction of PCE to ethene and ethane using microbial dechlorinators. More recently the problem of c-DCE accumulation at sites where the more rapid PCE and TCE reductions are complete, has resulted in greater attention being focused on the anaerobic reduction from the intermediate c-DCE. In the research reviewed (See Table 3.4), the selection of electron donors was critical to the success of many of the experiments. The following discussion only lists the donors that contributed to successful reduction. For further details on the other donors and nutrient amendments, see the appropriate reference. This research predicts and in some cases actually demonstrates (Beeman *et al.*, 1994, Yager *et al.*, 1997, Becvar *et al.*, 1998) that complete in-situ dechlorination of contaminants without accumulation of toxic daughter products is possible.

Using standard 160 mL or 120 mL batch microcosm studies, complete reduction of PCE has taken from two to forty days (DiStefano et al., 1991, 1992, Freedman et al., 1989, Fennell and Gossett, 1997, Smatlak et al., 1996, and Lorah et al., 1997) and proceeded using a variety of electron donors. As early as 1989, anaerobic studies (Freedman et al, 1989) showed that by using methanol, hydrogen, formate, acetate or glucose as an electron donor, low concentrations of PCE and TCE could undergo 100% conversion to VC and ethene in less than three days under methanogenic conditions. Similar studies by DiStefano et al., (1991, 1992) have also shown that by providing an adequate electron donor, high concentrations of PCE could be quickly reduced. In 1991 they demonstrated that by supplying methanol, 55 mg/L of PCE was 100% reduced to ethene in four days without methanogenesis. In 1992, they demonstrated 91 mg/L of PCE supplied at two day intervals could be completely reduced to VC and ethene within 14-40 days once a microbial population was established. In this study, researchers also demonstrated that it is the available hydrogen that is key to the reduction of chloroethenes and the metabolism of more complex donors serves to regulate the delivery of hydrogen. More recently, microcosm studies have also shown that PCE and TCE can be dechlorinated to ethene in 20 days and kinetics are directly affected by substate concentration (Nielsen and Keasling, 1998). By using groundwater from a PCE/TCE contaminated site and providing glucose as a substrate, complete reduction without accumulation of vinyl chloride was demonstrated. It was determined that for high PCE concentrations (>1 mg/L) degradation follow zero order kinetics while for low concentrations, degradation follows first order kinetics.

Fennell and Gossett, (1997) showed reductive dechlorination is dependant on the level of available hydrogen and that ethanol, lactate, propionate, and butyrate all served as effective hydrogen sources. The rate at which these donors provide hydrogen directly affects the conditions of dechlorination. If relatively high levels of hydrogen are produced, methanogens dominate while if the available hydrogen levels are kept low, reductive dechlorinators dominate without the production of methane. This competition for hydrogen by methanogens and reductive dechlorinators was also witnessed in a study using hydrogen and formate as donors to successfully reduce 12  $\mu$ M of PCE to ethene in only two days (Smatlak *et al*, 1996).

At an Aberdeen Proving Grounds site, sediments and groundwater were tested in microcosms to confirm in-situ dechlorination and to evaluate the potential for natural attenuation (Lorah *et al.*, 1997). With no additional donor added, complete removal of TCE and all daughter products was accomplished under methanogenic conditions. This indicated adequate donor supplies and capable microorganisms existed in-situ and natural attenuation is occurring.

Yang and McCarty (1998) demonstrated that dechlorination could be initiated on c-DCE and continued until it was completely reduced to ethene. Using benzoate and propionate it was shown that dehalogenators could use hydrogen at lower concentrations than methanogens or acetogens. The slower degradation of the propionate substrate provided hydrogen at a slow steady rate that favored greater dehalogenation than the benzoate that delivered adequate hydrogen to promote the competitive methanogens. When formate and acetate were provided in a mixed culture study (Windfuhr, 1998), microorganisms demonstrated the ability to completely dechlorinate c-DCE. Though successful dechlorination was observed, many inhibitors were also identified. These studies illuminate a promising outlook for the reduction of persistent c-DCE plumes, yet clearly establish the need to better understand the organisms involved in c-DCE reduction.

In an attempt to more closely mimic in-situ conditions, column microcosms have also been used to show complete reductive dechlorination is enhanced with the use of an electron donor (Carter and Jewell, 1992, DeBruin *et al*, 1992, Lee 1997, and Isalou *et al.*, 1998). In an expanded bed column with recycle (Carter and Jewell, 1992), it was shown that under methanogenic conditions, up to 12 mg/L of PCE could undergo 98% conversion to VC and ethene within three days when sucrose was supplied as a donor. Column operating temperatures in this study were maintained at 15 °C to simulate groundwater conditions. Using a fixed bed column to simulate passing groundwater through subsurface soils (DeBruin *et al*, 1992), it was shown that with lactate as a donor, 9  $\mu$ M of PCE could undergo 100% conversion to ethane. PCE was no longer detected in the column effluent after two weeks and after 240 days no ethenes were detected in the column effluent. To further simulate groundwater conditions, the operating temperature of this column was reduced from 20 °C to 10 °C and operated in the dark. Complete conversion of PCE continued under these conditions. While exploring the potential for bioaugmentation (Lee et al., 1997), 30mg/L of PCE were shown to be completely dechlorinated, under methanogenic conditions, in less than eight days during column studies. During long term column testing, it was also demonstrated that 600 µM of PCE could be completely reduced in 17 hours (Isalou et al., 1998). In a column that was operated for two and a half years under acetogenic conditions, PCE concentrations were raised from 12  $\mu$ M to 600  $\mu$ M while being supplied methanol as a substrate. For the first 21 months VC was the terminal endpoint. As acetogenesis became the primary metabolic pathway for methanol, ethene production began and continued through the remainder of the study.

As ultimate proof for complete in situ reductive dechlorination, a review of field studies was accomplished. In a field test in Victoria, Texas, PCE, TCE and DCE at 1700  $\mu$ g/L, 535  $\mu$ g/L, and 385  $\mu$ g/L, respectively, were reduced to below detection limits in less than two years (Beeman et al., 1994). Groundwater from a 450 square meter plot of land was continuously extracted from the down gradient side, augmented with benzoate and sulfate and injected up gradient under anaerobic conditions. Sulfate reducing conditions were allowed to dominate to control the production of VC, which is produced under methanogenic conditions, but not sulfate reducing conditions.

In-situ reductive dechlorination has also been observed at a New York site heavily contaminated with TCE, (Yager et al., 1997). Though no donor has been added, it is believed there is an adequate supply of subsurface donors from co-contaminants to facilitate the reduction of TCE to ethene. Differing subsurface soil zones and groundwater migrations are currently retarding the complete dechlorination of all TCE, but enhanced bioremediation is being considered.

Test	Source Concentrations	Successful Donors See ref. for nutrients	Conclusions	Reference
Microcosms: 100mL suspensions in 160 mL bottles	PCE 0.5 mg/L TCE 1.0 mg/L repeated as depletion occurred	Methanol, Hydrogen, Formate, Acetate, or Glucose in digester sludge	100% conversion to VC, Partial to ETH in 3 days; methanogenic conditions	Freedman and Gossett, 1989

CDOD 1DOD ( D.1 1 D.1

Test	Source	Successful Donors	Conclusions	Reference
Microcosms: 100mL suspensions in 160 mL bottles	Concentrations PCE 55 mg/L at 2 day intervals	See ref. for nutrients Methanol and yeast	100% Reduced to Ethene in 4 days w/o methanogenesis	DiStefano et al., 1991
Microcosms: 100mL suspensions in 160 mL bottles	PCE 91 mg/L at 2 day intervals	Hydrogen or Methanol with yeast	Complete Reduction to VC and ETH within 14- 40 days;acetogenic conditions.	DiStefano et al., 1992
Microcosms: 100 mL of medium in 160 mL bottles	12 μM PCE	Hydrogen and Formate with yeast and butyrate	Complete reduction to ETH in 2 days; Demonstrated methanogen/ dehalogenator competition	Smatlak <i>et</i> <i>al.</i> , 1996
Microcosms: 100mL suspensions in 160 mL bottles	PCE 110 μM at 2 day intervals	Ethanol, Lactate, Propionate, or Butyrate with yeast	Comparable conversion to ETH with 4 different donors; methanogenic conditions	Fennell and Gossett, 1997
Microcosms: 162 mL bottles with ground-water and sediment	7.6 μM TCE	None: No donor supplied; field conditions applied	Complete reduction to ETH in 34 days; methanogenic conditions	Lorah <i>et</i> <i>al</i> ., 1997
Microcosms: 100 mL of medium in 160 mL bottles	5 μM <i>c</i> -DCE incrementally	Benzoate or propionate	Complete reduction to ETH; Dehalogenator advantage at lower hydrogen levels	Yang and McCarty, 1998
Microcosms: 120 mL vials	100 μM c-DCE	Formate, acetate and yeast	Complete reduction to ETH	Windfuhr, et al., 1998
Up-flow, 900 mL Continuous Flow Reactor	PCE 8-12 mg/L	Sucrose and yeast extract	98% Conversion to VC and ETH in 3 days at 15°C under methanogenic conditions	Carter and Jewell, 1992

Test	Source Concentrations	Successful Donors See ref. for nutrients	Conclusions	Reference
Fixed Bed Columns	PCE 9 µM	Lactate in Rhine River sediment with anaerobic sludge	100% Conversion to ETH in 240 days	DeBruin et al., 1992
Column Studies	180 μM PCE	Not Specified	100% Conversion to ETH in 8 days under methanogenic conditions	Lee, et al., 1997
Columns: 16L Up-flow Continuous Feed; Long Term (2.5 yrs)	600 µM PCE	Methanol	Complete reduction within 17 hrs; Some residual 1,1 DCE under acetogenic conditions	Isalou, et al., 1998
In Situ Field Test, 3 feed wells, 3 extraction wells	PCE 1700 μg/L TCE 535 μg/L DCE 385 μg/L	Sodium Benzoate	All chlorinated ethenes reduced to BDL in 2 years under sulfate- reducing conditions	Beeman <i>et</i> <i>al.</i> , 1994
In Situ Biotrans- formation	TCE up to 20 mg/L	In-situ donor not identified; none added	Complete conversion to ETH in 6 months predicted through site modeling	Yager <i>et</i> al., 1997
In Situ Treatability NAS Fallon	PCE <2130μg/L TCE <675μg/L DCE<2130μg/L VC<3.8μg/L	Lactate or Ethanol and benzoate; with yeast and vitamins	Initial results suggest reductive dechlorination and	Becvar <i>et</i> <i>al.</i> , 1998

Enhanced bioremediation is currently being field tested at a site in Nevada with encouraging indicators of in-situ reductive dechlorination (Becvar *et al.*, 1998). Five parallel test beds have been isolated in a former fire training pit. The beds are supplied with yeast, vitamins and either lactate or ethanol and benzoate. Though it is too early to show a direct correlation between decreasing parent chloroethenes and increased daughter products, initial indicators suggest an increasing anaerobic environment with overall chloroethene reduction.

Taken collectively, these studies clearly show that complete reductive in situ anaerobic dechlorination of chlorinated ethenes is possible. Results show that anaerobic reduction is

not limited to only the more highly chlorinated compounds, but will continue through all daughter products to produce ethene and ethane as the final products. Successful dechlorination at an acceptable rate for contaminated site remediation appears to be dependent on enhancement with an electron donor. Results show there is not one specific donor that works in all situations. The cumulative evidence suggests however, that the most promising choices of electron donors are short carbon chain alcohols such as ethanol and methanol or weak organic acids such as lactic, butyric, and propionic acids. The evidence also suggests that slow release hydrogenic substrates may be preferable to enhance dehalogenators and sustain in situ remediation. Studies also examined the effects of temperature and show that enhanced reductive dechlorination can be successful at typical groundwater temperatures of 10 to 15 °C. There is ample evidence from these studies to show the key to enhanced reductive dechlorination is identifying the most successful electron donor and possibly nutrient limitations for each site's conditions.

#### CHAPTER IV

#### EXPERIMENTAL APPROACH

#### SITE HISTORY AND CHARACTERISTICS

Operable Unit 1 is located on the eastern side of Hill Air Force Base in Northern Utah and contains groundwater that is heavily contaminated with chlorinated solvents from past disposal practices. Historically this piece of property has had many uses and contains a number of sites that functioned as on-Base disposal sites for fuels, oils and solvents. This list includes:

- Chemical Disposal Pits 1 and 2 used for industrial liquid waste disposal,
- Landfill 3 used for industrial liquid and solid waste disposal (dump and burn),
- Landfill 4 used for sanitary refuse disposal,
- Fire Training Areas 1 and 2 used to practice extinguishing aircraft fires,
- The Waste Phenol/Oil Pit used to dispose and burn waste oils and phenols, and

- A Waste Oil Storage Tank Site used to store waste fuels, fuel oil and hydraulic fluids. These sites were in operation at various times from 1940 through the mid 1960s, and in some cases to the early 1970s at which time disposal and waste management practices were changed.

As a result of past practices, the contamination at this site is very complex and includes partially weathered or degraded fuels and solvents. Soil contamination includes PCE (up to 9,100 micrograms per kilogram [ $\mu$ g/kg]), TCE (up to 40,000  $\mu$ g/kg), DCE (up to 14,000  $\mu$ g/kg), and jet fuel (up to 42,100  $\mu$ g/kg). Contamination also affects approximately seven acres of groundwater that are characterized by a floating layer of non-aqueous phase liquid (NAPL). This NAPL contains high concentrations of solubilized chlorinated solvents particularly TCE (up to 2,300 micrograms per liter ( $\mu$ g/L), DCE (up to 42,000 mL), and VC (up to 2,400  $\mu$ g/L) (Montgomery Watson, 1995). PCE and TCE have been identified as wastes that were disposed of on-site. The DCE and VC were not identified as past waste and are presumed to be biodegradation products of PCE and TCE. Their presence indicates intrinsic bioremediation is occurring. Further review of data available in the 1995 RI/FS supports this conclusion and shows conditions are appropriate for intrinsic bioremediation in the Chemical Disposal Pit areas (CDP) of OU-1 (See Table 4.1). The decrease in dissolved oxygen, nitrate, and sulfate with a corresponding increase in dissolved iron and manganese in the source area and immediately adjacent to the source area support the conclusion that the subsurface environment is highly reducing and capable of supporting reductive dehalogenation.

	Intrinsic Bioremediation Indicators								
CDP Area Well U1-072 Well U1-088:									
	Bookground:		Well U1-067:		Downgradient				
	<b>v</b>	<b>v</b>	Source Area	CDPs	from CDPs				
	Upgradient		(1986-94)	(1986-94)	(1990-94)				
	from CDPs	RI & 1/4ly	(1900-94)	(1900-94)	(1990-04)				
Redox Potential (mV)	>-225		(-75100)	(-125150)	(-75100)				
Dissolved Oxygen (mg/L)	>7		<1	1-2	1-2				
Anions (mg/L)									
Bicarbonate	70-500	70-820	470	480	380-410				
Alkalinity, Bicarbonate	255-849			485-642	260-303				
Sulfate	0.96-58		ND	0.3-11	0.37-8.4				
Sulfide	ND-1	ND-1	1	0.1	ND				
Chloride	39-99.6	35-370	56	43.2-79.5	43.8-51.6				
Nitrate	0.88-5.8	ND-50	30	0.11-10	ND				
Cations (µg/L)									
Dissolved Iron	ND-874	ND-43000	36000	17100-32300	465-3000				
Dissolved Manganese	ND-251	ND-1810	820	582-960	59.6-960				
VOCs (µg/L)									
Tetrachloroethene			ND						
Trichloroethene			ND	•					
cis-Dichloroethene		ND-42000		2700-9700	ND				
Vinyl Chloride		ND-24000	2400	1	ND				
Ethene/Ethane/Methane	5	N/A	N/A	N/A	N/A				

Table 4.1. Intrinsic Bioremediation Indicators

Past studies conducted by the United States Air Force provide a great deal of information on physical characteristics of OU-1. Surface soils consist of moderate to excessively well drained sand-silt mixtures imbedded with gravel and possessing moderately high to high permeability. The horizontal hydraulic conductivity of the site has been estimated to range from 0.0002 to 0.0004 centimeters per second while the calculated horizontal interstitial velocity ranges from 0.85 to 12.76 feet per day. Vertical hydraulic conductivity of the site ranges form  $10^{-5}$  to  $10^{-8}$  centimeters per second while vertical velocity ranges from 0.014 to 0.240 feet per year. Monitoring data show the ground water is moving primarily horizontally across OU-1 and down the escarpment at the edge of the Base property with only minor vertical migration. (Montgomery Watson, 1995).

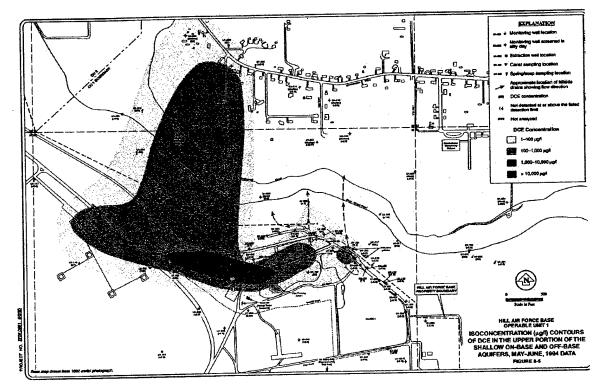


Figure 4.1. Hill AFB Operable Unit 1 Site Map

The stated worst-case contaminant levels are the maximum concentrations that have been found in the source area identified as the Chemical Disposal Pits. Any proposed remediation technology or implementation of the RABITT approach would involve treatment of much lower contaminant levels. Degradation and dissolution within the source area as well as in the area between the source area and treatment area will result in lower, more manageable concentrations that could be treated by an in situ bioremediation system. A review of chlorinated solvent plumes at Hill AFB in 1997 (Graves, *et. al.*) concluded that natural attenuation mechanisms at OU-1 are not limited to reductive dehalogenation. Other mechanisms include volatilization through the vadose zone, evapotranspiration occurring by capture of the groundwater on the hill slope by the root zone, and discharge of the shallow groundwater through seeps and springs.

The dichloroethene is of particular concern at OU-1 not only because it significantly exceeds the drinking water standards near the source of contamination, but also because it is

detectable in the groundwater over an area that covers 193 acres as shown in Figure 4.1 (Montgomery Watson, 1995). Incomplete dechlorination of this compound could result in the accumulation of significant levels of the more toxic vinyl chloride, a known carcinogen. Therefore, complete and rapid dechlorination of all chloroethene compounds is essential to successful remediation of OU-1. The 1998 Proposed Plan of action for remediation of OU-1 clearly demonstrates that partially due to past corrective actions, the c-DCE plume is shrinking, however, even with the proposed additional corrective action the estimated restoration timeframe for the source area is 50+ years and 12 years for the non-source area. Enhancing the bioremediation occurring on-site could significantly reduce these timeframes and greatly reduce any potential future health concerns.

#### EXPERIMENTAL DESIGN

By understanding the stoichiometry of chemical and biological reactions, we can anticipate changes in microbial environments. Microorganisms obtain energy for growth and maintenance by removing electrons from donors and transferring them to electron acceptors. When the donor or acceptor concentration is deficient, microbial activity is limited. Under anaerobic or microaerobic conditions and when an adequate donor concentration is available, chloroethenes can act as microbial substrates while serving as respiratory electron acceptors. As these microorganisms grow, the rate of substrate utilization is directly proportional to the mass of the microbial population mediating the reaction:

$$\mathbf{r}_{su} = -\mathbf{k}\mathbf{X}\left(\mathbf{S}/(\mathbf{K}_s + \mathbf{S})\right) \tag{1}$$

$$\mathbf{r}_{g} = \mathbf{Y}\mathbf{k}\mathbf{X}\left(\mathbf{S}/(\mathbf{K}_{s}+\mathbf{S})-\mathbf{B}\mathbf{x}\right)$$
(2)

$$\mu = Yk (S/(K_s + S) - b$$
 (Monod, 1942, van Uden, 1967) (3)

where:  $r_{su}$  = rate of substrate utilization, gS/l-day

- k = maximum substrate utilization rate, gS/gX-day
- X = biomass concentration, g/l

S = rate-limiting substrate concentration, mg/L

- $K_s =$  Monod constant; half-velocity coefficient, mg/L
- $r_g = rate of microorganism growth$
- Y = Yield coefficient, gX/gS
- $b = decay coefficient, day^{-1}$
- $\mu$  = specific growth rate, day<sup>-1</sup>

According to these equations, the concentration of microorganisms will increase if the substrate concentration is in excess of the rate of decay. By providing excess donor in the form of organic compounds, naturally occurring electron acceptors will be depleted and microorganisms capable of discharging electrons to other available acceptors gain a selective advantage. As this proceeds, biologically active zones can be established which accelerate the rate of reductive dechlorination and growth of halorespiring microorganisms. With excess electron donor, the chloroethene electron acceptors become the limiting substrates and conditions are optimized for their effective biotransformation.

In the case of column reactors, macroscopic transport of chloroethenes can be characterized by a 1-D advection/dispersion/sorption/reaction equation:

$$\varepsilon (dS/dt) = D_{H}(d^{2}S/dX^{2}) - V (dS/dX) - aJ + QS$$

$$= dispersion - advection - reaction + the source$$
(4)

where:  $\varepsilon = \text{porosity}$ 

S = substrate concentration t = time  $D_{H} = Diffusion coefficient$  X = Biofilm thickness V = Specific Discharge at Darcy Velocity a = surface area J = Flux in the biofilm layerQ = Flowrate

By solving these coupled equations for each chloroethylene, key kinetic parameters for the reduction of chloroethenes can be predicted and a process model can be developed for field application.

#### MATERIALS AND METHODS

#### Soil Collection

Soil samples were collected in the Hill Air Force Base source area identified as Chemical Disposal Pits number 1 and 2. All soils were collected using a hollow stem auger drill with split-spoon samplers. Samples were collected at depths ranging from 29 to 33 feet below ground surface. This depth is characterized by a sandy/gravel formation that comprises the saturated zone just above a clay layer. To minimize exchange with the atmosphere, cores were transferred from the split spoons in the field and immediately placed in sterile 1 quart

glass Mason<sup>TM</sup> jars containing groundwater from the same hole used to collect soil samples. Groundwater in the jars was allowed to overflow as the soil was added and all jars were capped with Teflon<sup>TM</sup> lined lids as described in the RABITT protocol (Morse *et al.*, 1998). Soil jars were maintained under anaerobic conditions at 4° C for approximately six-months prior to assembling microcosms. Soils were characterized by a black oily appearance and strong hydrocarbon odor.

Preliminary soil respirometry testing was completed to verify the viability of these soils. (See Appendix A for Respirometry Protocol). Respirometers were assembled in an anaerobic glovebox with a 95%  $N_2/5$ %  $H_2$  atmosphere and purged with nitrogen prior to being removed from the glovebox. Regardless of efforts to limit oxygen introduction, respirometers were not maintained anaerobically. Respirometry tests proceeded under microaerobic conditions. Results of testing showed carbon dioxide production in nearly all respirometers and methane production in n-propanol amended respirometers. Results of this testing suggested an active, indigenous microbial population and viability of these soils for the electron donor study.

#### Aquifer Sampling

Groundwater supplied to all the microcosms was collected from OU-1 Dewatering Well # U1-201, immediately adjacent to the Chemical Disposal Pits. This well is 31 feet deep and screened from 20-30 feet below ground surface and located approximately 50 feet to the East of the site used for soil collection. Well #U1-201 has a submerged pump to deliver groundwater to the leachate collection system that transports water to the Industrial Waste Treatment Plant (IWTP). The well head is equipped with a faucet that is pressurized due to backpressure in the leachate collection system. Water was collected from this faucet using a Tygon<sup>TM</sup> hose dedicated for this purpose. To minimize mixing with the atmosphere, the water flow rate was kept very low and the hose outfall was maintained at the bottom of the twenty-liter collection bottle until it overflowed. The bottle was sealed with no headspace using a rubber stopper and Parafilm® and immediately transported to the laboratory. In the laboratory the water was stored at room temperature (19°C) in the dark until it was separated into unamended control water and nutrient amended water bottles which were installed inline in the microcosm apparatus. Water was collected from Well #U1-201 approximately every seven days. The water was slightly greenish/yellowish in appearance with a noticeable hydrocarbon sheen. Occasionally globs of brown oily materials were present in the water.

#### **Donor Selection**

Electron donors were selected based on the literature review and each chemical's ability to release diatomic hydrogen (See Appendix B). Fatty and aromatic acids, an alcohol and an aromatic hydrocarbon were chosen to allow a broad examination of the potential to enhance dechlorination. In specific, lactic acid, benzoic acid, n-butyric acid, and toluene were obtained from Fisher Scientific Co. (Fairlawn, NJ) while n-propanol and propionic acid were obtained from Mallinckrodt Baker Inc. (Paris KY). These donors supply electrons according to the oxidation half reactions defined in Table 4.2. The candidate donors were selected based on their ability to supply some of their H<sub>2</sub> equivalents in low-energy, low-rate biochemical reactions. The moles H<sub>2</sub> per electron equivalent of the selected donors and final donor concentrations are shown in Appendix B and C.

Table 4.2. Electron Donor Oxidation Half Reactions	
Electron Donors Selected	$\Delta G^{o}$
	(kJ/eequiv)
n-Propanol	-31.42
$1/18 \text{ CH}_3 \text{CH}_2 \text{CH}_2 \text{OH} + 5/18 \text{ H}_2 \text{O} = 1/6 \text{ CO}_2 + \text{H}^+ + \text{e}^-$	
Propionic Acid	-27.88
$1/14 \text{ CH}_3\text{CH}_2\text{COO}^- + 5/14 \text{ H}_2\text{O} = 1/14 \text{ HCO}_3^- + 1/7 \text{ CO}_2 + \text{H}^+ + \text{e}^-$	
Lactic Acid	-32.94
$1/12 \text{ CH}_3\text{CHOHCOO}^- + 1/3 \text{ H}_2\text{O} = 1/12 \text{ HCO}_3^- + 1/6 \text{ CO}_2 + \text{H}^+ + \text{e}^-$	
Butyric Acid	-17.63
$1/20 \text{ CH}_3\text{CH}_2\text{CH}_2\text{COO}^- + 7/20 \text{ H}_2\text{O} = 1/20 \text{ HCO}_3^- + 3/20 \text{ CO}_2 + \text{H}^+ + \text{e}^-$	
Benzoic Acid	-28.84
$1/30 C_6 H_5 COO^- + 13/30 H_2 O = 1/30 H CO_3^- + 1/5 CO_2 + H^+ + e^-$	
Toluene	-28.15
$1/36 C_7 H_8 + 14/36 H_2 O = 7/36 CO_2 + H^+ + e^-$	

As previously stated the reduction of the chloroethenes in microcosm studies and field evaluations has been dependent upon an adequate supply of substrate. Operable Unit 1 soils and groundwater are characterized by very high levels of dichloroethene. To provide complete remediation, the calculated electron equivalent demand was based on the premise that all existing species of chloroethenes would need to be completely reduced to ethene. To ensure an adequate supply of the electron donors, the predicted electron equivalents demand was based on worst case chloroethene concentrations at OU-1. The calculated electron equivalent demand was then increased by a safety factor of four (See Appendix D). To stimulate microbial dehalogenation in the continuous flow microcosms, butyric acid, lactic acid, propionic acid, and n-propanol solutions were supplied at approximately 2% of the total flow while benzoic acid and toluene solutions were supplied at approximately 8% of the total flow. Differences in supply rates were based on solubility limits of benzoic acid and toluene.

#### Microcosm Configuration and Assembly

Continuous upflow soil columns were designed to simulate subsurface conditions characteristic of OU-1. A schematic of a single column reactor is shown in Figure 4.2, however, the full system included eight parallel columns and is shown in Figure 4.3. Each column contained a lower layer of soil and an upper layer of OU-1 groundwater. All columns were operated in the dark at 19° C throughout the experiment and received continuous flow of the same OU-1 source groundwater, amended differently for each microcosm. The groundwater supply was amended with Resazurin® (Sigma Chemical Co, St Louis, MO) as an indicator of low redox potential (<1 mg/L to avoid toxicity). With the exception of one column receiving only groundwater, the columns also received yeast extract (Sigma Chemical Co, St Louis, MO), NaHCO<sub>3</sub> (Sigma Chemical Co, St Louis, MO) as a buffer, and vitamins. The yeast extract was supplied in low doses (20 mg/L) while the water was buffered with 1g/L to maintain an alkalinity of 300-1500 mg/L as CaCO<sub>3</sub>. Improper buffering between days 59 and 166 did not affect alkalinity, however, during this period the pH in all vitamin amended columns ranged from 8.5 to 9.0. Vitamins were supplied according to the draft RABITT protocol recipe (Morse *et al.*, 1997) and listed in Table 4.3.

On experiment day 134, spiking of the groundwater supply began to compensate for near non-existent levels of c-DCE in the water collected from Well #UI-201. After start up of all columns, the water from Well #U1-201 showed a progressive and drastic reduction in c-DCE concentrations. Spiking to approximately 1000  $\mu$ g/L c-DCE (Supelco, Bellefonte, PA) was attempted based on average concentrations of c-DCE found in monitoring wells in and around Chemical Disposal Pits 1 and 2 (Montgomery-Watson, 1995). Difficulties in mixing c-DCE into the water in the feed reservoirs resulted in actual c-DCE concentrations ranging from 150-663  $\mu$ g/L. Spiking with c-DCE continued throughout the remainder of the study.

26

Table 4.3. Vitamin Concentrations Used in Microcosm Study						
Quantity	Chemical Supplier					
(mg/L)						
20	Sigma Chemical Co, St Louis, MO					
20	Fisher Scientific Co. Fairlawn, NJ					
100	Fisher Scientific Co. Fairlawn, NJ					
50	Sigma Chemical Co, St Louis, MO					
50	Eastman Kodak Co, Rochester, NY					
50	Aldrich Chemical Co, Milwaukee, WI					
50	Sigma Chemical Co, St Louis, MO					
10	Sigma Chemical Co, St Louis, MO					
50	Sigma Chemical Co, St Louis, MO					
50	Fisher Scientific Co. Fairlawn, NJ					
	Quantity (mg/L) 20 20 100 50 50 50 50 50 10 50					

. ... <u> .</u> 1

Other than the c-DCE spiking, columns were supplied with three combinations of ingredients. One column received only unamended groundwater and served as a background reactor simulating current groundwater conditions. One column was supplied with OU-1 groundwater containing Resazurin®, buffer, vitamins and yeast extract but no electron donors. This column served as both the background reactor to evaluate the effects of nutrients and as a control for electron-donor-augmented reactors. The remaining columns were supplied groundwater, Resazurin®, buffer, vitamins, yeast extract, and an electron donor solution provided according to values shown in Table 4.4. See Appendix E for donor delivery calculations based on actual Total Organic Carbon (TOC) analysis.

Table 4.4. Electron Donor Supply Values								
				Total	Donor %	Donor		Molar
			Rate	Flow	of Total	Supply	Donor	Concentration
Electron Donor	M.W.	(mm)	(ml/min)	(ml/min)	Flow	(mg/L)	(mg/L)	(mM)
n-Butyric Acid	88.10	0.19	0.0044	0.235	1.87	8536.63	159.8	1.814
Benzoic Acid	122.12			0.233	8.15	2180.71	177.7	1.455
Lactic Acid	90.08			1	1.80	7882.00	142.1	1.578
Propionic Acid	74.08	1		0.223	1.97	5658.89	111.7	1.507
n-propanol	60.09				1.90	3672.17	69.9	1.164
Toluene	92.13		-		8.11	72.55	5.9	0.064

By examining the chemical oxygen demand (COD) associated with c-DCE, we also confirmed these TOC levels represent an excess of electron equivalents. Eight milligrams of COD is equivalent to 1.0 milliequivalent (mequiv) of electrons and four mequiv of electrons are required for the reduction of 1 mM of c-DCE to ethene. Therefore, a minimum of 32 mg/L of COD is required for the reduction of 1 mM or 97 mg/L of c-DCE to ethene. Assuming only 10% of the electrons are available for reductive dehalogenation (McCarty,

1998), 320 mg/L would be required. The COD to organic carbon ratio for organic material is typically 2.5 to 3.5; the amount of TOC equivalent to achieve the reduction of 1 mM of c-DCE would be on the order of 90 to 130 mg/L COD. For the 1000 ppb c-DCE spiking goal, these values would range from 0.93 mg/L to 1.34 mg/L. Analytical results show that all donors were supplied well in excess of these values (See Appendix E).

Groundwater collected as previously described was mixed and added to the system approximately every seven days. Unamended and amended feed reservoirs were drained and refilled each time water was needed to ensure consistency in the water provided to all columns. Fresh donor supplies were mixed and installed at intervals not exceeding 30 days.

The system used in this study had three primary components: a feed assembly, the columns, and an effluent assembly. With the exception of the manifold tubing in the peristaltic pump, donor feed line, and sampling septums, all components were stainless steel, glass, or Teflon<sup>TM</sup> to avoid incompatibilities with chlorinated compounds and to limit sorption and volatilization losses (See Appendix F for Materials Inventory).

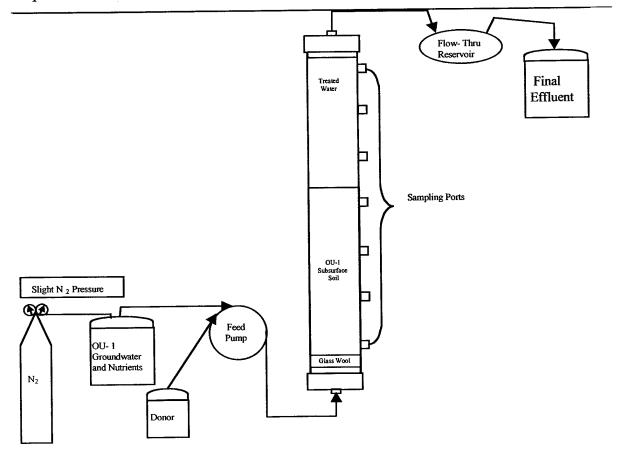
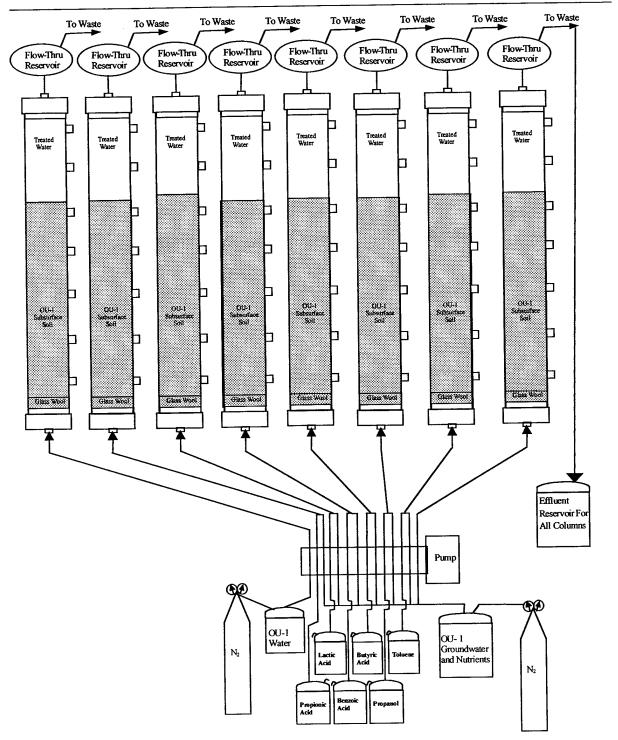
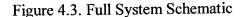


Figure 4.2. Single Column Schematic

The feed assembly consisted of the groundwater reservoirs and 1L-donor solution bottles, a nitrogen headspace system for the influent reservoirs, the pump and the associated fittings. Nitrogen was supplied to the groundwater reservoirs at just above atmospheric pressure to





provide an inert head on the feed bottles and compensate for the vacuum created as the water was pumped from the bottles. Donor bottles were fitted with pressure compensation lines to allow stabilization with atmospheric pressure while controlling the potential entry of foreign materials into the bottles. A multichanneled (Watson-Marlow Model 205U) peristaltic pump, operated at 3 rpm was used as the sole driver for all groundwater and donor solutions and provided flow (0.233+/-0.011 mL/min) at a rate reflective of the hydraulic conductivity encountered in OU-1 (0.045cm/min (0.221mL/min)) (Montgomery -Watson, 1995). See Appendix G for predicted and measured HRTs and flow calculations. Groundwater supplied through 1.42 mm ID manifold tubing (Watson-Marlow #978.0142.000) and donor solutions were combined prior to entering the columns. Butyrate, lactate, propionate, and propanol were supplied through 0.19 mm ID manifold tubes (Watson-Marlow #984.0019.000) while benzoate and toluene were supplied through 0.38 mm ID manifold tubes (Watson-Marlow #984.0038.000) to compensate for lower solubility levels. Immediately after combining, all solutions were pumped through the base of the vertical columns.

The microcosm columns were glass liquid chromatography columns (Ace Glass, Inc #5820-37) measuring 25 millimeters ID and having a useable length of 600 millimeters. All columns were fitted with a vertical series of  $\frac{1}{4}$  inch glass sampling ports with rubber septa to facilitate profile sampling. Each column contained a lower layer of grossly contaminated soil supported by 1 centimeter of glass wool (Alltech Associates, Inc) and an upper layer consisting of OU-1 groundwater percolated through the soil. The soil layer was 46 centimeters  $\frac{1}{-2.5}$  centimeters in depth. The slight difference in soil depths has been attributed to a variance in settling after column assembly. Soil homogenization and column assembly took place in an anaerobic glovebox ( $95\%N_2/5\%H_2$ ) to avoid adding oxygen to the soils. After assembly, microcosms were sealed and removed from the glovebox. Seals were interrupted only long enough to allow connection within the full microcosm system.

The effluent from each column passed through an in-line effluent reservoir prior to disposal. The 40-mL reservoir served a dual purpose: this reservoir bottle filled with effluent water provided an air tight seal to the effluent end of each reactor and provided fluid for temporary back flow to compensate for the sample volume removed during sampling. Final effluent from these reservoirs was collected in a single waste reservoir that was returned to Hill AFB and disposed of through the Industrial Wastewater Treatment Plant.

### Analytical Method

The parameters of concern for this project are defined in Table 4.5 along with their respective analytical method of measurement. Each microcosm was sampled for volatile organics, pH, dissolved oxygen content, and alkalinity after a thirty-day acclimation period. Influent samples were collected directly from the unamended and amended groundwater reservoirs using a 25-milliliter pipette. Effluent samples for all columns were collected from the upper most sampling port on the microcosm using 50-mL syringes equipped with 18 gauge needles. Samples were slowly transferred to beakers or sample vials in a manner minimizing possible aerobic mixing. Dissolved oxygen, pH and alkalinity were then sampled on a weekly basis throughout the remainder of the experiment. Volatile organic compounds (VOC) were sampled once every three weeks for the first 15 weeks of the experiment and then every week for the remaining nine weeks in which DCE spiking occurred. All VOC samples were collected in 44mL VOA vials and preserved for transportation to the laboratory using 0.4 mLs of a solution containing 0.1 mg/L sodium azide (J.T. Baker Chemical Co, Phillipsburg, N.J.).

Parameter	Analytical Methods	Performed By:
РСЕ	SW846 Method 8260B	Hill AFB Environmental
TCE		Chemistry Laboratory (TIEL)
c-1,2-DCE, t-1,2-DCE		
1,1-DCE		
VC		
Ethene, Ethane, Methane	Kampbell et al., 1998	Hill AFB TIEL
Chloride	EPA Method 300	In-House; UWRL
TOC	EPA Method 5310C	American Analytical
Alkalinity	AWWA Method 2320B	In-House
pH	Ion Selective Electrode	In-House
Dissolved Oxygen	DO Probe	In-House

Samples were collected on a weekly basis and analyzed for pH, dissolved oxygen and alkalinity. The pH and dissolved oxygen were analyzed using a direct reading Accumet AR50 (Cat#13636AR50) Dual Channel pH/Ion Conductivity meter with a AccTupH probe (Cat# 13-620-181) and a dissolved oxygen probe (Cat# 970899). Alkalinity was determined using the titration method described in Standard Methods, 17th edition.

Concentrations of chlorinated organics were determined by their chromatographic mobility and their mass spectral fragmentation using EPA Method SW8260 with purge and trap. Prior to analysis, the samples were purged with helium for 12 minutes at 30°C while headspace gases were collected on a 25 cm x 0.267 cm I.D. Tekmar #3 diphenyl oximer polymer with silica gel and coconut charcoal trap. After four minutes desorbing at 225°C, a 5 mL aliquot was introduced through a splitless injection port into a Finnigan Mat -Incos 50 XL gas chromatograph/mass spectrometer (GC/MS). A 75m x 0.53mm ID Mega Bore capillary column coated with DB 624 (J&W Scientific) was installed on the GC. The system was temperature programmed as follows: hold at 5°C for 10 minutes and ramped to 145°C at 8°C per minute. The system was then elevated to 225°C for 6 minutes to drive off water vapor and heavier analytes.

Concentration of ethene, ethane, and methane were determined using a headspace equilibrium method described by Kampbell and Vandegrift (1998). Prior to analysis the samples are allowed to equilibrate to room temperature and inverted. A 10-mL aliquot was removed by inserting a needle attached to a gas tight syringe through the septa. A second needle connected to a supply of helium at ambient pressure was inserted through the septa. As the sample water was removed, helium was allowed to fill the 10 mL headspace and a 100 ul aliquot of headspace was drawn into a gastight syringe and injected into a Perkin-Elmer Model 8400 capillary gas chromatograph (GC) equipped with a flame ionization detector (FID). A 25m x 0.53mm ID plot fused silica Pora Plot Q column was installed on the GC. The system was temperature programmed as follows: hold at 30°C for 4 minutes and ramped to 150°C at 30°C per minute to drive off water vapor and heavier analytes. Methane, ethene and ethane eluted at 0.7, 1.5, and 2.0 minutes respectively. Analyte peaks were integrated and concentrations were calculated by comparing to standard curves.

# CHAPTER V

## **RESULTS AND DISCUSSION**

The c-DCE removal efficiencies and related discussion of this experiment are presented in this chapter. The discussion is organized to address the project objectives including: 1.) demonstrating complete reductive dechlorination of *cis*-DCE without accumulation of VC is possible under microaerobic conditions using OU-1 soil and groundwater, 2.) comparing various electron donors and determining the most promising donor(s) for maintaining dechlorination under OU-1 site conditions, 3.) determining if vitamin and yeast amendments are necessary for complete dechlorination in OU-1, and 4.) demonstrating a cost-effective alternative for remediation of chloroethenes at OU-1. To address objectives, examination of c-DCE removal efficiencies is followed by a comparison of removal in unamended and amended columns as well as the columns supplied with donors. By examining the data (See Appendix H for analytical results) some interesting reactions other than those involving chloroethenes were identified. These are discussed at the end of this chapter.

# CHLOROETHENE DECHLORINATION EFFICIENCIES IN LABORATORY COLUMNS

Removal efficiencies of c-DCE in all columns encompassed a wide range and were very inconsistent. By reviewing Table 5.1, it is apparent that not only is the c-DCE removal efficiency highly variable it appears that at times c-DCE is produced within the columns.

Table 5.1.	cis-Dichlor	oethene Percer	ntage Rer	noval Effi	ciencies	(Influent	vs Effluent	.)
	Unamended	Vitamin/Yeast						
	Groundwater	Amended	n-Butyric	Benzoic	Lactic	Propionic		
Experiment	Effluent	Groundwater	Acid	Acid	Acid	Acid	n-Propanol	Toluene
Day	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
31	11.41	4.35	6.76	8.40	7.35		6.05	
52	-10.04	2.11	5.74	11.25	3.28	8.36		10.13
72	11.84	10.26	13.57	19.12		9.07	12.90	
93	15.04	1.14	-2.24	8.08	15.10		5.80	
115	61.69	9.01	12.76	16.84	-81.72			
136	29.78	22.50	21.86	34.49	20.77	33.22		
143	33.48	14.92	19.71	23.49				
150	-4.02	25.33	19.95	33.74				
158	49.82	-68.44	-151.79	-123.05	-146.55			
165	51.58	14.33	14.73	21.80	22.39			
171	-1.45	3.05	-0.16	0.02	0.69	37.76		
178	-0.44	-0.06	0.18	10.14	3.39	2.92		
185	-11.95	-0.47	5.19	21.30	3.33	10.59		
192	13.76	69.35	3.47	13.78	9.47	3.47		
199	3	4.19	6.39					
Range	-10/62%	-68/69%	-152/22%	-123/34%	-147/22%	-134/38%	-93/28%	-179/25%

For VC and other compounds discussed, sample results that showed concentrations below detection limits were assigned "apparent" removal efficiencies by assuming the sample concentration was zero. In sample sets with a detectable influent concentration and an effluent concentration below detectable limits, an apparent removal efficiency of 100% was assigned. In sampling events where the influent value was below detectable limits, an apparent removal efficiency of zero has been assigned. In some cases, assuming a zero concentration may hide production of some compounds during a sampling event, however, even by applying production values greater than 200%, the relationship between average removal efficiencies of any compound examined does not change.

In Table 5.2, vinyl chloride apparent removal efficiencies are presented to address the issue of accumulation resulting from the dehalogenation of c-DCE. In most cases, VC accumulation was demonstrated at decreasing levels until day 115 at which time all columns began showing zero accumulation. Zero accumulation continued through the remainder of the experiments. All removal efficiency values of zero in Table 5.2 reflect the "apparent" removal efficiencies and demonstrate no accumulation or reduction of VC.

1 20 001 1

Table 5.2.	v myr Chlori	de Apparent R	cilloval L	/IIICICIIC)	03			
	Unamended	Nutrient/Yeast						
	Groundwater	Amended	n-Butyric	Benzoic	Lactic	Propionic		
Experiment	Effluent	Groundwater	Acid	Acid	Acid	Acid	n-Propanol	Toluene
Day	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
31	8.44	-7.42	-6.96	-10.78	-5.07	-8.75	-10.99	-14.69
52	-109.20	-95.38	-95.38	-72.88	-89.48	-61.56	-36.50	-92.58
72	0.00	-17.24	-43.02	-15.80	-16.20	-31.22	-28.22	
93	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
115	0.00	100.00	-21.45	-9.54	-96.54	-4.17	-8.46	
136	0.00	0.00	0.00	0.00	0.00			
143	0.00	0.00	0.00	0.00	0.00	0.00		
150	0.00	0.00	0.00	0.00	0.00			
158	0.00	0.00	0.00	0.00	0.00	0.00		
165	0.00	0.00	0.00	0.00	0.00	0.00		
171	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
178	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
185	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
192	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
199	0.00	0.00	0.00	0.00	0.00	0.00	0.00	
Range	-109/8%	-95/100%	-95/0	-72/0%	-97/0%	-62/0%	-37/0%	-93/0%

Table 5.2.	Vinvl	Chloride .	Apparent	Removal	Efficiencies

-

. . . . . . . . . . . . . . .

Chloroethene-DCE Degradation in the Absence of Additives

Removal of c-DCE and VC in the unamended column varied widely and did not demonstrate a consistent pattern (See Figure 5.1). Removal efficiencies for c-DCE ranged from -10% to 62% including five sampling events that appeared to demonstrate accumulation or production of c-DCE, five sampling events that demonstrated removal at greater than 20%, and five sampling events that demonstrated removal between zero and 20%. Removal efficiencies for VC ranged from -109% to 8%, however, most sampling events did not identify VC at detectable concentrations.

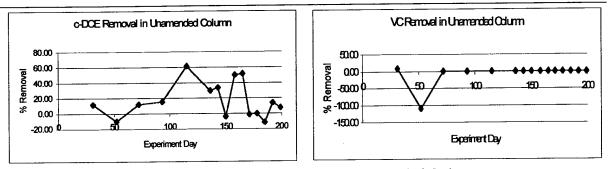
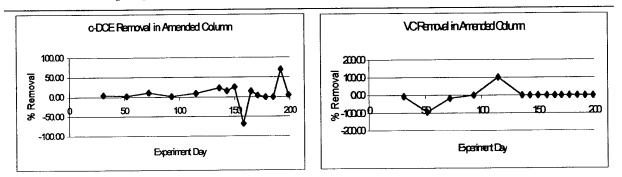
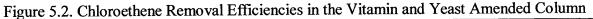


Figure 5.1. Chloroethene Removal Efficiencies in the Unamended Column

### Chloroethene Degradation in the Presence of Vitamins and Yeast

Removal efficiencies for c-DCE and VC in the vitamin and yeast amended column covered a wide range and did not present a consistent trend (See Figure 5.2). The c-DCE removal efficiencies ranged from -68% to 69% including three sampling events that demonstrated the accumulation or production of c-DCE, three sampling events that represented greater than 20% removal, and nine sampling events that represented removal efficiencies between zero and 20%. VC removal efficiencies ranged from -95% to 100% and included 11 sampling events that had less than detectable levels of VC.





#### Columns Amended with Electron Donors

Six columns received continuous feed of a specified electron donor in addition to the vitamin and yeast amendments and are individually discussed below. A review of c-DCE results shows a wide range of removal efficiencies with a notable event occurring on day

158. Results for that sampling event show all columns receiving amendments had considerably higher c-DCE levels in the effluent than in the influent water (See Appendix H for c-DCE removal rates and average rates excluding day 158 results). VC removal efficiencies also varied greatly during periods of VC accumulation, which were followed by less than detectable VC concentrations in all columns.

### Chloroethene Degradation in the Presence of n-Butyric Acid

The n-butyric acid column removal and apparent removal efficiencies for c-DCE and VC ranged from -152% to 22% and -95% to zero respectively (See Figure 5.3). Removal of c-DCE included three sampling events that demonstrated apparent production, nine sampling events that demonstrated between zero and 20% removal, and one sampling event that demonstrated higher than 20% removal. The VC removal efficiencies varied during the initial period of VC accumulation and eleven on the sampling events showed no detectable VC in the influent or effluent.

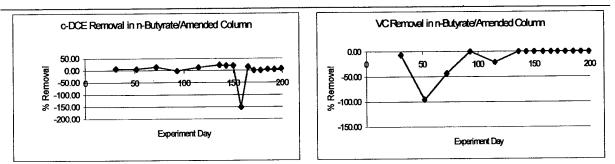


Figure 5.3. Chloroethene Removal Efficiencies in the n-Butyric Acid/Amended Column

## Chloroethene Degradation in the Presence of Benzoic Acid

Removal and apparent removal efficiencies for the column supplied with benzoic acid were -123% to 34% and -72% to zero for c-DCE and VC respectively (See Figure 5.4). The

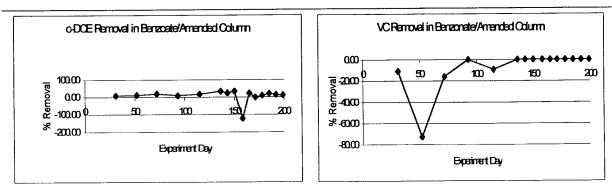


Figure 5.4. Chloroethene Removal Efficiencies in the Benzoic Acid/Amended Column

only sampling event that demonstrated an apparent production of c-DCE was on day 158. Nine sampling events demonstrated zero to 20% c-DCE removal while five sampling events demonstrated greater than 20% c-DCE removal. Four sampling events demonstrated the accumulation of VC while eleven sampling events showed no detectable VC.

## Chloroethene Degradation in the Presence of Lactic Acid

Chloroethene removal and apparent removal efficiencies in the column supplied with lactic acid were also widely varied (See Figure 5.5). Removal efficiencies for c-DCE ranged from -147% to 22% while the apparent removal efficiencies for VC ranged from -97% to zero. Two sampling events showed production of c-DCE. Removal efficiencies of zero to 20% were identified during eleven sampling events and twice the removal efficiencies of c-DCE exceeded 20%. VC apparent removal efficiencies demonstrated accumulation during four sampling events while the remaining eleven events had no detectable VC.

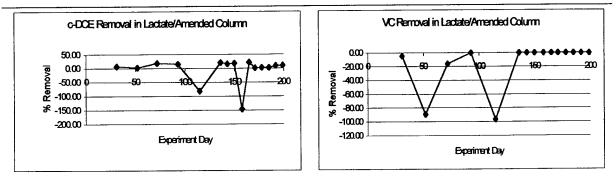
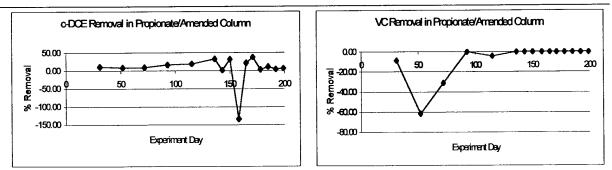
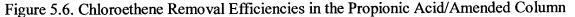


Figure 5.5. Chloroethene Removal Efficiencies in the Lactic Acid/Amended Column

### Chloroethene Degradation in the Presence of Propionic Acid

Removal efficiencies of c-DCE and apparent removal efficiencies of VC in the propionic acid column ranged from -134% to 38% and -62% to zero respectively (See Figure 5.6). Removal efficiencies for c-DCE suggest the production of c-DCE only once (day 158) while

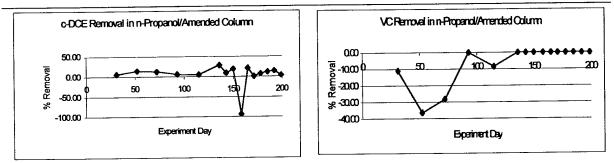


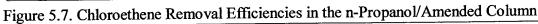


removal efficiencies were between zero and 20% ten time and exceeded 20% four times. The apparent removal efficiencies of VC demonstrated the accumulation of VC during four sampling events while the remaining eleven events showed no detectable VC.

## Chloroethene Degradation in the Presence of n-Propanol

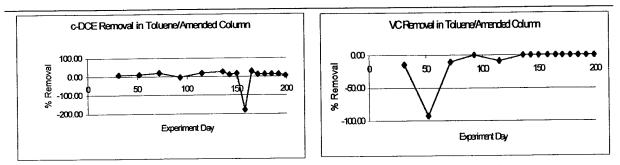
In the column supplied with n-propanol, the removal efficiencies for c-DCE ranged from -93% to 28% while the apparent removal efficiencies for VC ranged from -37% to zero (See Figure 5.7). Removal efficiencies of c-DCE indicate the production of c-DCE only once (day 158) while 12 sampling events demonstrated removal efficiencies between zero and 20% and two sampling events demonstrated removal efficiencies exceeding 20%. The VC apparent removal efficiencies for the propanol column also demonstrated four events that show VC accumulation while the other eleven events showed no detectable VC.

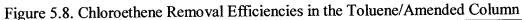




## Chloroethene Degradation in the Presence of Toluene

Removal and apparent removal efficiencies in the toluene column ranged from -179% to 25% for c-DCE and -93% to zero for the VC (See Figure 5.8). Two sampling events demonstrated the apparent production of c-DCE while eleven sampling events demonstrated removal efficiencies between zero and 20% and two sampling events demonstrated removal





efficiencies greater than twenty percent. VC accumulation was demonstrated during four sampling events while the remaining eleven sampling events showed no detectable VC.

## COMPARISON OF DECHLORINATION EFFICIENCIES

The wide range of dechlorination efficiencies and the lack of obvious trends make comparison of the various removal efficiencies difficult. However, to address the stated objectives, comparisons between unamended, amended, and electron donor supplied columns must be accomplished. The following paragraphs document these comparisons in an effort to identify the best conditions for in-situ reductive dehalogenation at OU-1.

# Comparison of Background Columns; Unamended versus Amended

By comparing the unamended column with the vitamin and yeast-amended column removal efficiencies, we see that both columns have inconsistent efficiencies and show both the production or accumulation of c-DCE and high levels of c-DCE removal. The unamended column shows c-DCE production more frequently than the amended column, however, the amended column shows a much higher level of c-DCE production (day 158). The number of sampling events that demonstrate c-DCE removal in these columns is very similar, but the unamended column removal efficiencies tend to be larger in magnitude. Average removal efficiencies for the unamended and amended columns are 17% and 7% respectively. From the data available, the unamended column demonstrated greater c-DCE removal efficiencies than that of the vitamin and yeast-amended column. By comparing these two columns, there is no apparent advantage to adding vitamins and yeast to stimulate microbial growth, however, more consistent c-DCE removal efficiencies are needed to substantiate this conclusion.

The unamended and amended columns both demonstrate a VC accumulation phase that may represent biological acclimation. This is followed by a series of samples with no detectable VC. These data show that when c-DCE dechlorination is occurring in these columns, VC is also dechlorinated within the columns and VC accumulation does not persist. In each column there is only one event that clearly demonstrates actual VC removal. In the unamended column removal is shown at 8% in samples collected on day 31 while the amended column showed complete removal in the samples collected on day 158. Based on the available apparent removal efficiencies, the performance of these columns is similar and there is no apparent advantage to supplying vitamins and yeast.

## Comparison of Different Donor Dechlorination Efficiencies

In each column supplied vitamins, yeast, and an electron donor, removal efficiencies cover a wide range but all follow a similar pattern. In all columns some c-DCE production is demonstrated and all columns showed their highest production in the day 158 samples. The butyrate column had the highest frequency of c-DCE production with three sampling events showing production. The toluene column had the highest single-event c-DCE production represented by a removal efficiency of -179% (day 158 samples). During dechlorination, all columns showed similar removal efficiencies with the highest single-event removal efficiency of 38% demonstrated in the propionate column. Strongly influenced by the results of day 158, the average removal efficiencies for the columns supplied electron donors ranged from -5% to 7%. The n-butyric and lactic acid columns both had negative average removal values at -2% and -5% respectively. The benzoic acid column had the highest average removal of 7% followed closely by the propionic acid and n-propanol columns which both averaged 5% c-DCE removal. A comparison of the data suggests that of the electron donors examined, benzoic acid is the best source of electrons needed to support reduction of c-DCE. However, data from the propionic acid and n-propanol columns also suggest they supply adequate electrons to support sustained reduction of c-DCE. Further and more consistent removal data is needed before one preferred electron donor can be specified.

The pattern of apparent VC removal is very similar for all columns supplied electron donors. All columns began with low levels of VC accumulation (5-14%) followed by an increase in accumulation. This accumulation then dropped to undetectable levels of VC only to be followed by accumulation in all columns on day 115. After day 115 all columns showed zero apparent VC removal. The worst single VC accumulation event occurred in the lactic acid column (-97%) followed closely by the n-butyric acid column (-95%). Average apparent removal efficiencies showed n-propanol had the lowest VC accumulation (14%) during acclimation, however, benzoic and propionic acids supported only slightly higher VC accumulation during acclimation. Further data is needed to accurately define the electron donor that best supports VC removal. By comparing the six columns that received electron donors it is difficult to select the single donor that best supported complete reductive dehalogenation of c-DCE. Benzoic acid had the highest average removal efficiency of c-DCE while n-propanol had the lowest accumulation for VC. Propionic acid had similar removal efficiencies to both benzoic acid and n- propanol for both these reductive steps while benzoic acid and n-propanol were similar to each other for both steps. The removal efficiencies for n-butyric and lactic acids indicated they would not be as efficient at supporting reductive dechlorination of c-DCE or VC. The removal efficiency of toluene placed it between these two groups with regard to supporting complete reductive dechlorination.

# Comparison of Donor Column Dechlorination Efficiencies to Background Columns

A comparison of c-DCE removal efficiencies in columns provided an electron donor and the background columns suggests the electron donors did not offer an advantage to dechlorinators in this experiment. The vitamin and yeast amended column demonstrated the highest single-event removal at 69%, followed closely by the unamended column at 62%. These efficiencies were much higher than the highest event in the donor supplied columns which was 38%. Average removal efficiencies for all columns receiving the vitamin and yeast amendment were strongly affected by the high c-DCE production values noted on day 158. The amended column with no electron donor had the same average c-DCE removal efficiency as the benzoic acid supplied column while it had a better average removal than the other electron donor supplied columns. The average removal efficiency for the amended column was 7% while the electron donor supplied columns averaged 7% for benzoic acid, 5% for propionic acid and n-propanol, 0.6% for toluene, -2% for n-butyric acid, and -5% for lactic acid. In comparison, the unamended column with no donor supplied had an average removal efficiency of 17%. Based on the average removal efficiencies, neither the vitamins, yeast, nor electron donors appear to offer an advantage for accelerating reductive dechlorination under conditions present in this experiment. It must, however, be noted that, while all columns demonstrated a reduction of dissolved oxygen (DO) between the influent and effluent samples, the unamended column maintained higher DO levels. Effluent DO values for the unamended column ranged from 1 to 2 part per million (ppm) while the effluent from all amended columns was consistently below 0.5 ppm. This may have given an advantage to the oxidative dechlorination of c-DCE in the unamended column while the

amended columns did not have excess DO and were limited to the energetically more demanding reduction of c-DCE.

Apparent VC removal efficiencies suggest the unamended column acclimated more rapidly than the amended and donor supplied columns. The preponderance of nondetectable levels of VC make it difficult to conclude vitamin amendments or electron donors offer an advantage in avoiding the accumulation of VC during the dechlorination of c-DCE.

### DISCUSSION OF DECHLORINATION EFFICIENCIES

During most sampling events the concentration of c-DCE appeared to be decreasing, however, the explanation for this decrease is not clear and may be attributed to a variety of processes. Typical c-DCE removal efficiencies throughout the experiment ranged from 2% to 30% and results from c-DCE samples do not indicate a microbial acclimation period occurred. Occasional unexplained negative removal efficiencies were noted. Of particular interest are the samples collected on day 158 that suggest high levels of c-DCE production, however, these results are suspected to be the product of poor sampling technique. With electron donors supplied well in excess of the calculated demand, results do not indicate a population of microbial dechlorinators able to utilize the donors and establish a robust population capable of degrading the c-DCE according to Monod kinetics. Degradation that did occur may be attributed to an existing population of dechlorinators that were not able to multiply significantly, cometabolism within the columns, or aerobic removal resulting from sample collection and handling.

Analytical results for VC offer a more promising outlook for dechlorinators. After an initial acclimation period (three months), nearly all sample results for VC were below detectable limits. Column influent samples showed less than detectable levels of VC, however, degradation of c-DCE should have lead to some production of VC within the columns. All production of VC within the columns was not detectable at the effluent of the columns in nearly all sampling events. Again, the overall database of results is not consistent enough to conclude this to be the results of dechlorinator activity, but VC results certainly suggest it.

A review of chloride levels was completed to evaluate the complete reduction of c-DCE. Due to financial constraints, only one set of chloride samples was analyzed. In all columns, except the unamended column, a slight increase in chloride concentration was observed.

42

Though initially encouraging, the levels of chloride production could easily be attributed to the chloride available from the vitamin amendments (See Appendix I for chloride calculations and results). With the vitamins serving as a possible source of chloride, it was not possible to demonstrate c-DCE reduction by examining chloride production.

Ethene and methane were also examined as indicators of dechlorination and results were inconsistent but showed a decrease in average ethene concentrations and an increase in average methane concentrations. Ethene results intermittently showed the production of ethene, possibly from the reduction of VC, in all columns except the unamended column. The average removal efficiencies in all these columns indicate a loss of ethene and may contraindicate reductive dechlorination. A review of ethane results indicates that rapid conversion of the produced ethene to ethane did not occur. Alternatively these results may also indicate complete mineralization of chloroethenes. The unamended column showed no ethene production while removal efficiencies were lower than in any of the donor supplied columns. The highest level of ethene removal was seen in the column supplied propionic acid while the worst removal was in the column supplied only vitamins and yeast. All columns showed periodic methane removal, however, average removal efficiencies for all columns indicate methanogenic conditions. Energetics suggest this indicates the depletion of available electron acceptors including oxygen, nitrate, chloroethenes, and sulfate, however, in this experiment c-DCE clearly persisted while methane was produced. This production of methane demonstrated the competition for reducing equivalents between dechlorinators and methanogenic microorganisms. Methane production was lowest (45%) in the unamended column and of the donor supplied columns methane production was lowest (74%) in the propionic acid column while it was highest (-232%) in the column supplied with lactic acid. The remaining columns showed methane production ranging from 99% to 125%.

#### PREDICTION OF FIELD DECHLORINATION VALUES

Having established experimental conditions similar to those within OU-1, predictions can be made regarding the level of dechlorination per distance the groundwater travels. The unamended column demonstrated the highest c-DCE removal efficiencies and provides the best basis for predictions within OU-1, assuming enhanced dechlorination is not attempted. Experimental results show that the highest influent c-DCE concentration also has the highest removal per distance traveled though the aquifer solids. At a groundwater concentrations of 2736  $\mu$ g/L experimental conditions demonstrated 7.33  $\mu$ g/L of c-DCE was lost per cm of aquifer solids traversed. The average c-DCE removal per cm of aquifer solids used throughout this experiment was calculated to be 1.33  $\mu$ g/L/cm of aquifer solids and reflects the predicted removal for current conditions within OU-1 (See Appendix H). A predicted loss of 1.33  $\mu$ g/L/cm assumes the same in situ removal efficiencies as those seen in the laboratory and is likely an optimistic prediction, however, this level of removal has been demonstrated and could be used in future decisions regarding clean up proposals for OU-1.

#### OTHER REACTIONS OF INTEREST

The complete reduction of c-DCE without the accumulation of VC was the primary focus of this experiment, however, a review of analytical data shows some other beneficial reactions consistently occurred. Though changes in groundwater may have had an influence on chemical concentrations, analytical results indicated decreasing concentrations of 1,1,1-trichloroethane (1,1,1 TCA), 1,4-dichlorobenzene (1,4 DCB), and chlorobenzene (CB) as water passed through the columns. For all these compounds the last three sampling events suggest a change in trends, however, not enough data are available to pursue this change.

Nearly complete removal of 1,1,1 TCA was observed in all amended columns. The unamended column showed varying degrees of 1,1,1 TCA production and removal and had an average removal of 28%. In contrast, all columns that received the vitamin and yeast amendment had typical removal values of 100% with average removal efficiencies ranging from 71% to 80% in spite of three sampling events with no influent 1,1,1 TCA and therefore assigned apparent removal efficiencies of zero.

Chlorobenzene and 1,4 DCB had between 25% and 39% average removal efficiencies in all amended columns. CB results showed limited periods of production in all vitamin and yeast amended columns, however, removal efficiencies were consistent during periods of removal. All amended columns showed CB removal approximating twice that of the unamended column. The 1,4 DCB results showed a pattern similar to the removal of VC. After an initial period of 1,4 DCB accumulation (three months), removal efficiencies became consistent between 30% and 60%. The average removal efficiency for each of the amended columns was more than an order of magnitude greater than the average removal efficiency in the unamended column.

# CHAPTER VI CONCLUSIONS

After a review of all data and procedures associated with this research project, conclusions have been drawn to support the stated objectives as well as address issues associated with future work similar to this effort. The high degree of variability in analytical results makes conclusions regarding reductive dechlorination difficult, however there are sufficient data regarding removal efficiencies to address some of the objectives. Though it cannot be conclusively stated that the physical setup or operations of this project contributed to incomplete dechlorination, some operational limitations are clear.

## CONCLUSIONS DIRECTLY RELATED TO STUDY OBJECTIVES

Degradation of c-DCE did occur without the accumulation of VC in unamended, amended, and donor supplied columns. The removal of c-DCE was initially demonstrated in the first sample set collected on day 31. The VC removal demonstrated an acclimation phase which was followed by compete removal of VC regardless of the c-DCE concentration or c-DCE removal efficiencies during the same sampling event.

Of the electron donors supplied, benzoic acid supported the highest c-DCE removal. Propionic acid and n-propanol had similar c-DCE removal efficiencies while n-butyric and lactic acid had the lowest c-DCE removal efficiencies. In all cases the data are not consistent enough to predict success or failure if applied in-situ.

The addition of vitamins and yeast did not improve reductive dechlorination of c-DCE or VC. For the c-DCE to VC step, the unamended column had better removal efficiencies than any column receiving vitamins and yeast. Similarly for the VC-ETH step, the unamended column more rapidly acclimated and demonstrated complete removal of VC faster than any column receiving vitamins and yeast. These data showed that the microorganisms involved in these two steps are not nutrient limited and vitamin amendments are not necessary for reductive dechlorination to proceed.

This experiment has failed to demonstrate a cost-effective treatment alternative to the currently proposed pump-and-treat system intended for use at OU-1. Regardless of the abundance of indicators suggesting the application of an electron donor to stimulate the

reductive dechlorination of c-DCE in OU-1, results from this experiment do not offer conclusive evidence that in-situ reductive dechlorination can be enhanced and field testing should not be attempted at this time. Experimental results under different operating conditions could prove more successful and ultimately offer a treatment solution.

#### SUPPLEMENTAL STUDY CONCLUSIONS

Based on removal efficiencies, the column that showed the best reduction of c-DCE was the unamended column. Though oxygen levels of 1-2 ppm may have facilitated the higher removal values, this can not be concretely determined.

An inadequate number of analytical parameters were regularly examined. The analysis of chloride, dissolved hydrogen and other electron acceptors including nitrate, manganese (IV), iron (III), and sulfate are needed to determine the removal pathways and establish a balance for the reducing equivalents.

The c-DCE lower threshold concentration for the microorganisms in this soil and groundwater is not known, therefore we do not know if experimental c-DCE concentrations ever exceeded this threshold. Past research has typically shown 100 to 300  $\mu$ M (9,694-29,082 ppb), and in some cases 10  $\mu$ M (969 ppb) (Yang and McCarty, 1998 and Beeman *et al*, 1994) to be above threshold limits. The highest concentration of c-DCE recorded during this experiment was 2,736 ppb while typical concentrations were less than 300 ppb. Even after the c-DCE available in the groundwater decreased to negligible levels, the attempted spiking failed to raise the delivered concentrations of c-DCE to 1,000 ppb. As a results typical OU-1 site levels were not demonstrated to be above the lower threshold concentration for indigenous microorganisms.

The acclimation period of indigenous c-DCE dechlorinating microorganisms is not known and therefore we do not know if experimental conditions allowed acclimation to occur. Site characteristics indicate that dechlorinators are present in-situ, however once soils and groundwater are taken from the site, the delicate conditions necessary to support these microorganisms are altered. Even though great effort was extended to keep the soils and groundwater as close to in-situ conditions as possible, variations did occur. Column operating temperatures for this experiment were 19°C which is much higher than in-situ conditions. During water collection, sampling, and transferring to the feed assemble the water was mixed causing the system to function as a series of microaerobic columns (< 2.0 ppm oxygen with oxygen utilizing microbial activity) instead of anaerobic (zero oxygen and no oxygen utilizing microbial activity) columns. The addition of amendments alters the water chemistry and may offer advantages to microorganisms other than dechlorinators.

The addition of vitamin and yeast facilitated the removal of 1,1,1 TCA, 1,4 DCB and CB. In all columns receiving the amendments, the removal of these compounds was substantially greater than that seen in the unamended column. In direct conflict with the reduction of chloroethenes, these data show nutrient limitations for the organisms facilitating these reductions.

Samples collected from all vitamin and yeast amended columns on day 158 demonstrated results far removed from the predominant values in all amended columns. These results demonstrated a high level of c-DCE production or accumulation and affected much lower average removal efficiencies for all these columns. To further address the effect of this sampling event, removal rates based on time between sampling events and average removal rates excluding this sample were calculated and can be found in Appendix H. These sample results did not, however, affect the comparative relationships between column performance and the same conclusions still apply.

The effects of running microaerobic microcosms as opposed to anaerobic microcosms are not known. A majority of the literature reviewed addressed similar studies as anaerobic, however, very few offered any data regarding oxygen content. This research demonstrated the difficulty in maintaining anaerobic conditions and showed the greatest dechlorination in the column with the highest oxygen content.

# CHAPTER VII RECOMMENDATIONS

The construction and operation of this system, as well as the review of analytical results, illuminated some problem areas encountered during this experiment and promoted the generation of recommendations to improve further research. Clearly the demonstrated results do not show complete manifestation of the theories given in Chapter IV, however, further research exploring different operating parameters should be conducted before enhanced insitu biological treatment is eliminated as a treatment technology for OU-1.

Benzoic acid, propionic acid, and n-propanol should be included in future studies. These compounds were the top performers in this study and may demonstrate greater success in future studies incorporating recommendations listed below.

The delivered concentration of c-DCE should be better regulated to provide microbial populations a stable supply of electron acceptors on which to acclimate and grow. The varied concentration of c-DCE may have made it difficult for a microbial population to grow to a level sufficient to utilize and reduce the contaminants to below the MCL of 70 ppb. Stabilizing concentrations may allow the microbes to acclimate, grow and utilize the c-DCE in a manner that demonstrates Monod kinetics. Sufficient groundwater needed for the duration of any future research should be collected and spiked as a single batch. Continued mixing should ensure consistent concentrations throughout the project.

Concentrations of 1,000 ppb and higher should be evaluated to identify the c-DCE threshold concentrations associated with OU-1 indigenous microorganisms. The spiking goal of 1,000 ppb ( $10.3\mu$ M) was established based on average groundwater monitoring data near the Chemical Disposal Pits. Past research (Yang and McCarty, 1998 and Beeman *et al*, 1994) has shown this level to be above threshold concentration for various microbial populations, however the population in OU-1 may be quite different than populations in other research. Spiking never achieved the goal of 1,000 ppb; therefore it was never determined that this level is above threshold concentrations for OU-1. The identification of the threshold concentration for microorganisms in OU-1 is critical for site treatment applications. If the lower threshold concentration exceeds the clean-up goal, in this case an MCL of 70 ppb, the technology is not appropriate for the site.

Nutrient limitations should be further explored, however the vitamin recipe used in this research should be simplified. The most recent draft of the RABITT protocol (Morse *et al*, 1998) suggests the use of only vitamin  $B_{12}$ . The vitamin recipe used in this experiment was followed faithfully, yet the mixing of these vitamins adds one more opportunity for slight variation and operator error.

Further evaluations attempting to mimic site conditions should operate at lower temperature. The dominant theory is that elevating the temperature will increase microbial growth and the probability of successfully promoting dechlorinator growth, however this theory has not always proved correct. To truly mimic OU-1 conditions, columns should be operated at 10°C (Montgomery Watson 1995).

Routine analysis of chemical parameters should be expanded in any future study to identify appropriate electron donors for OU-1. Chloride, dissolved hydrogen, and additional electron acceptors should be included in the list of analytes. Reductive dechlorination of chloroethenes results in an increase in the concentration of chloride ions and analytical results could provide conclusive evidence that this process is occurring. Chloride ion results would also provide the missing data needed to calculate a mass balance on the chlorine within the system. Each terminal electron accepting process has a characteristic hydrogen concentration associated with it and analytical data could be used to indicate the dominant redox processes. Analysis of nitrate, manganese (IV), iron (III) and sulfate, at least initially, could provide evidence of the depletion of other electron acceptors. Failure to demonstrate depletion of these competing electron acceptors, particularly the nitrate, could indicate the microbial population is unable to utilize the chloroethenes as electron acceptors.

Future column studies should include a longer experimental run time. The RABITT protocol (Morse *et al*, 1998) indicates studies should include a minimum of six months. It is likely that indigenous microbial populations would not require this long to reacclimate to soils and water in the assembled columns, however, this can not be concretely demonstrated. The time needed to deplete other available electron acceptor, acclimatize a healthy population of dechlorinators, and allow the dechlorination process to proceed is influenced by site conditions and may be greater than the 199 days allowed in this experiment.

A microbial examination using most probable number (MPN) assays should be considered. Of interest would be anaerobic heterotrophs, sulfate reducers, hydrogen and

> , ,

49

acetate using methanogens and dechlorinators. Procedures for these MPN assays are described by Maymo-Gatell *et al.* (1995). Results of these tests could be compared to chemical measurements from the study and provide some indication of the types of microorganisms indigenous to these soils and water.

#### REFERENCES

Adriaens, P. and Vogel, T. M. 1995. "Biological Treatment of Chlorinated Organics." p. 435-486. In Young, L. Y. and Cerniglia, C. E., (Eds). Microbial Transformation and Degradation of Toxic Organic Chemicals. Wiley-Liss, New York, New York.

Alvarez, P. J. J., Cronkhite, L. A., and Hunt, C. S. 1998. "Use of Benzoate to Establish Reactive Buffer Zones for Enhanced Attenuation of BTX Migration: Aquifer Column Experiments." Environmental Science & Technology. 32(4): 509-515.

Ballapragada, B. S., Puhakka, J. A., Stensel, H. D., and Ferguson, J. F. 1995. Development of Tetrachloroethene Transforming Anaerobic Cultures from Municipal Digestor Sludge, p. 91-97. <u>In</u> Hinchee, R. E., Leeson, A., and Semprini, L. (Eds). Bioremediation of Chlorinated Solvents. Battelle Press, Columbus, Ohio.

Beeman, R. E., Howell, J. E., Shoemaker, S. H., Salazar, E. A., and Buttram, J. R. 1994. "A Field Evaluation of In Situ Microbial Reductive Dehalogenation by the Biotransformation of Chlorinated Ethenes." <u>In Bioremediation of Chlorinated and</u> Polycyclic Aromatic Hydrocarbon Compounds: 14-27 Lewis Publishers, Ann Arbor, MI.

Becvar, E. S., Fisher, A., Sewell, G., Magar, V., Gossett, J., and Vogel, C. M. 1998. "Enhanced In Situ Reductive Dechlorination." In Hinchee, R. E. (Ed). Bioremediation and Phytoremediation Chlorinated and Recalcitrant Componds: 121-127. Battelle Press, Columbus, OH.

Bagley, D. M., and Gossett, J. M. 1990. "Tetrachloroethene Transformation and cis-1,2-Dichloroethene by Sulfate-Reducing Enrichment Cultures." Applied and Environmental Microbiology. 56(8):2511-2516.

Carter, S. R., and Jewell, W. J. 1992. "Biotransformation of Tetrachloroethylene By Anaerobic Attached-Films at Low Temperatures." Water Resources Research. 27(4):607-615.

Clesceri, L.S., Greenburg, A.E., and Trussell, R.R. 1989. "Standard Methods For The Examination of Water and Wastewater, 17<sup>th</sup> Edition." American Public Health Association, Washington D.C.

Criddle, C. S., Alvarez, L. A., and McCarty, P. L. 1991. Microbial Processes In Porous Media, p. 639-691. In J. Bear and M. Y. Corapcioglu (Eds) Transport Processes in Porous Media. Kluwer Academic Publishers, Netherlands.

DeBruin, W. P., Kotterman, M. J. J., Posthumus, M. A., Schraa, G., and Zehnder, A. J. B. 1992. "Complete Biological Reductive Transformation of Tetrachloroethene to Ethane." Applied and Environmental Microbiology. 58(6):1996-2000.

DiStefano, T. D., Gossett, J. M., and Zinder, S.H. 1991. "Reductive Dechlorination of High Concentrations of Tetrachloroethene to Ethene by an Anaerobic Enrichment of Cultures in the Absence of Methanogenesis." Applied and Environmental Microbiology. 57(8):2287-2292.

DiStefano, T. D., Gossett, J. M., and Zinder, S.H. 1992. "Hydrogen as an Electron Donor for Dechlorination of Tetrachloroethene by an Anaerobic Mixed Culture." Applied and Environmental Microbiology. 58(11):3622-3629.

EPA-450/3-87-026. 1987 "Hazardous Waste Treatment, Storage and Disposal Facilities (TSDF) Air Emission Models

Fatherpure, B. Z., and Boyd, S. A. 1987. "Reductive Dechlorination of Perchloroethylene and the Role of Methanogens." FEMS Microbiology Letters. 49(1988):149-156.

Fatherpure, B. Z., Nengu, J. P., and Boyd, S. A. 1987. "Anaerobic Bacteria That Dechlorinate Perchloroethene." Applied and Environmental Microbiology. 53(11):2671-2674.

Fatherpure, B. Z., and Boyd, S. A. 1988. "Dependence of Tetrachloroethylene Dechlorination on Methanogenic Substrate Consumption by *Methanosarcina* sp. Strain DCM." Applied and Environmental Microbiology. 54(12):2976-2980.

Fennell, D. E., Gossett, J. M., and Zinder, S. H. 1997. "Comparison of Butyric Acid, Ethanol, Lactic Acid, and Propionic Acid as Hydrogen Donors for the Reductive Dechlorination of Tetrachloroethene." Environmental Science & Technology. 31(3):918-926.

Freedman, D. L., and Gossett, J. M. 1989. "Biological Reductive Dechlorination of Tetrachloroethylene and Trichloroethylene to Ethylene Under Methanogenic Conditions." Applied and Environmental Microbiology. 55(9):2144-2151.

Gao, J., Skeen, R. S., Hooker, B. S., and Quesenberry, R. D. 1997. "Effects of Several Electron Donors on Tetrachloroethylene Dechlorination in Anaerobic Soil Microcosms." Water Research. 32(10):2479-2486.

Gerritse, J., Renard, V., Pedro-Gomes, T. M., Lawson, P. A., Collins, M. D., and Gottschal, J. C., 1996. "*Desulfitobacterium* sp strain PCE1, an Anaerobic Bacterium that can Grow by Reductive Dechlorination of Tetracholoethen or *ortho*-chlorinated phenols." Arch. Microbiology. 165:132-140.

Gibson, S. A., and Sulfita, J. M. 1986. "Extrapolation of Biodegradation Results to Groundwater Aquifers: Reductive Dehalogenation of Aromatic Compounds." Applied and Environmental Microbiology. 52(4):681-688.

Gibson, S. A., and Sewell, G. W. 1992. "Stimulation of Reductive Dechlorination of Tetrachloroethene in Anaerobic Aquifer Microcosms by Addition of Short-Chain Organic Acids or Alcohols." Applied and Environmental Microbiology. 58(4):1392-1393.

Gossett, J. M., and Zinder, S.H. 1996. "Microbiological Aspects Relevant to Natural Attenuation of Chlorinated Ethenes" From the Proceedings of Symposium on Natural Attenuation of Chlorinated Organics in Ground Water. p. 10-13. EPA/540/R-96/509.

Graves, R. W., Hinchee, R. E., Jensen, T. M., Graves, A. E., Weidemeier, T., Wheeler, M., Elliot, R. 1997. Natural Attenuation of Chlorinated Solvents, p. 141-145. <u>In</u> Alleman B. C., Leeson, A. (Eds). In Situ and On-Site Bioremediation. Battelle Press, Columbus, Ohio.

Heath, M. S., Wirtel, S. A., and Rittman, B. E. 1990. "Simplified Design of Biofilm Processes Using Normalized Loading Curves." Research Journal. 62(2):185-192.

Hollinger, C. 1992. Reductive Dehalogenation by Anaerobic Bacteria. PhD. Dissertation. Agricultural University, Wageningen, the Netherlands.

Hollinger, C., Schraa, G., Stams, A. J. M., and Zehnder, A. J. B. 1993. "A Highly Purified Enrichment Culture Couples the Reductive Dechlorination of Tetrachloroethene to Growth." Applied and Environmental Microbiology. 59(9):2991-2997.

Hollinger, C. and Schumacher W. 1994. "Reductive Dehalogenation as a Respiratory Process." Antoine van Leeuwenhoek. 66:239-246.

Hutchins, S. R. 1997. "Column Study on Nitrate-Based Bioremediation." DRAFT, U.S. Environmental Protection Agency, Robert S. Kerr Environmental Research Laboratory. Ada, OK.

Isalou, M., Sleep, B. E., and Liss, S. N. 1998. "Biodegradation of High Concentrations of Tetrachloroethene in a Continuous Flow Column System." Environmental Science & Technology. 32:3579-3585.

Kampbell, D.H., and Vandergrift, S.A. 1998. "Anaylsis of Dissolved Methane, Ethane, and Ethylene in Ground Water by a Standard Gas Chromatographic Technique." Journal of Chromatographic Science, Vol 36: 253-256.

Krumholz, L. R. 1995. A New Anaerobe That Grows with Tetrachloroethylene as an Electron Acceptor. Abstract Presented at the 95<sup>th</sup> General Meeting of the American Society for Microbiology.

Lee, M. D., Bledsoe, S. A., Solek, S. M., Ellis, D. E., and Buchanen Jr., R. J. 1997. "Bioaugmentation with Anaerobic Enrichment Culture Completely Degrades Tetrachloroethene in Column Studies." p. 21. <u>In</u> Alleman B. C., Leeson, A. (Eds). In Situ and On-Site Bioremediation. Battelle Press, Columbus, Ohio.

Lorah, M. M., Olsen, L. D., Smith, B. L., Johnson, M. A., and Fleck, W. B. 1997. "Natural Attenuation of Chlorinated Volatile Organic Compounds in a Freshwater Tidal Wetland, Aberdeen Proving Ground, Maryland." USGS Water-Resources Investigatons Report 97-4171

Maymo-Gatell, X., Gossett, J. M., and Zinder, S. H. 1995. "Dehalococcus Ethenogenes" Strain 195: Ethene Production from Halogenated Aliphatics, p. 23. <u>In</u> Alleman B. C., Leeson, A. (Eds). In Situ and On-Site Bioremediation. Battelle Press, Columbus, Ohio.

Maymo-Gatell, X., Tandoi, V., Gossett, J. M., and Zinder, S. H. 1995. "Characterization of an H2-Utilizing Enrichment Culture That Reductively Dechlorinates Tetrachloroethene to Vinyl Chloride and Ethene in the Absence of Methanogenisis and Acetogenesis." Applied and Environmental Microbiology. 61(11):3928-3933.

McCarty, P. L. 1969. Stoichiometry of Biological Reactions. International Conference "Toward a Unified Concept of Biological Waste Treatment Design," October 6, 1972, Atlanta, Georgia.

McCarty, P. L. 1969. Energetics and Bacterial Growth. Fifth Rudolph research Conference, July 2, 1969 The State University, New Brunswick, New Jersey.

McCarty. P. L., 1998. "Remediation of Chlorinated Solvent Contamination." From Proceedings of the Workshop on Environmental Acceptable Endpoints: Chlorinated Organics, Energetics, and Heavy Metals. Strategic Environmental Research and Development Program, Arlington, VA.

Mohn, W. W., and Tiedje, J. M. 1992. "Microbial Reductive Dehalogenation." Microbiological Reviews. 56(3): 482-507.

Montgomery Watson. 1995. "Final Comprehensive Remedial Investigation Reports for Operable Unit 1, Hill Air Force Base, Utah."

Morse, J. J., Alleman, B. C., Gosset, J. M., Zinder, S. H., Sewell, G. W., and Vogel, C. M. 1997. Draft "A Treatability Test for Evaluating the Potential Applicability of the Reductive Anaerobic Biological In Situ Treatment Technology (RABITT) to Remediate Chloroethenes." Environmental Security Technology Certification Program; Battelle Memorial Institute, Columbus, Ohio.

Morse, J. J., Alleman, B. C., Gosset, J. M., Zinder, S. H., Fennell, D. E., Sewell, G. W., and Vogel, C. M. 1998. Draft "A Treatability Test for Evaluating the Potential Applicability of the Reductive Anaerobic Biological In Situ Treatment Technology (RABITT) to Remediate Chloroethenes." Environmental Security Technology Certification Program; Battelle Memorial Institute, Columbus, Ohio.

Neumann, A., Scholz-Muramatsu, and Diekert, G. 1994. Tetrachloroethene Metabolism of *Dehalospirillum multivorans*. Arch. Microbiology. 162:295-301.

Newberg, S, Warikoo, V., Sharma, P. K., and McCarty, P. L., 1997. Bioaugmentation with Strain MS-1 for Tetrachloroethene Anaerobic Biotransformation to *cis*-1,2-Dichloroethene, p. 1. <u>In</u> Alleman B. C., Leeson, A. (Eds). In Situ and On-Site Bioremediation. Battelle Press, Columbus, Ohio.

Nielsen, R. B., and Keasling, J. D. 1998. "Anaerobic Degradation of PCE and TCE DNAPLs by Groundwater Microorganisms." In Hinchee, R. E. (Ed). Bioremediation and Phytoremediation Chlorinated and Recalcitrant Componds: 97-101. Battelle Press, Columbus, OH.

OO-ALC/EMR. 1998. "Proposed Plan: Operable Unit 1, Hill Air Force Base, Utah."

Pavlostathis, S. G., and Zhuang, P. 1993. "Reductive Dechlorination of Chloroalkenes in Microcosms Developed with a Field Contaminated Soil." Chemosphere 27(4): 585-595.

Scholz-Muramatsu, H., Szewzyk, R., Szewzyk, U., and Gaiser, S. 1989. "Tetrachloroethylene as electron acceptor for the anaerobic degradation of benzoate." FEMS Microbiology Letters 66(1990): 81-86

Sharma, P. K., and McCarty, P. L. 1996. "Isolation and Characterization of a Facultatively Aerobic Bacterium That Reductively Dehalogenates Tetrachloroethene to *cis*-1,2-Dichloroethene." Applied and Environmental Microbiology. 62(3): 761-765.

Smatlak, C. R., Gossett, J. M., and Zinder, S. H. 1996. "Comparative Kinetics of Hydrogen Utilization for Reductive Dechlorination of Tetrachloroethene and Methanogenesis in an Anaerobic Enrichment Culture." Environmental Science & Technology. 30(9):2850-2858.

Test Methods for Evaluating Solid Wastes, Volume 1B: Laboratory Manual Physical/Chemical Methods SW-846; U. S. Environmental Protection Agency, Office of Solid Waste and Emergency Response, Washington D.C. 1986

Wackett, L. P. 1995. "Bacterial Co-Metabolism of Halogenatec Organic Compounds." p. 217-241. In Young, L. Y. and Cerniglian, C. E., (Eds). Microbial Transformation and Degradation of Toxic Organic Chemicals. Wiley-Liss, New York, New York.

Weidemeier, T. H., Swanson, M. A., Moutoux, D. E., Gordon E. K., Wilson, J. T., Wilson, B. H., Kampbell, D. H., Hansen, J. E., Hass P., and Chapelle, F. H. 1996. "Technical Protocol for Evaluating Natural Attenuation of Chlorinated Solvents in Groundwater." Air Force Center for Environmental Excellence; San Antonio, TX.

Windfuhr, C., Granzow, S., Scholz-Muramatsu, H., and Diekert, G. 1998. "Reductive Dechlorination of *cis*-1,2-Dichloroethene with an Enriched Mixed Culture." <u>In</u> Hinchee, R. E. (Ed). Bioremediation and Phytoremediation Chlorinated and Recalcitrant Componds: 155-159. Battelle Press, Columbus, OH.

Wu, W. M., Nye, J., Hickey, R. F., Jain, M. K., and Zeikus, J. G. 1995. Dechlorination of PCE and TCE to Ethene Using Anaerobic Microbial Consortium, p. 45-52. <u>In</u> Hinchee, R. E., Leeson, A., and Semprini, L. (Eds). Bioremediation of Chlorinated Solvents. Battelle Press, Columbus, Ohio.

Yager, R. M., Bilotta, S. E., Mann, C. L., and Madsen, E. L. 1997. "Metabolic Adaptation and in Situ Attenuation of Chlorinated Ethenes by Naturally Occurring Microorganisms in a Fractured Dolomite Aquifer near Niagara Falls, New York." Environmental Science & Technology. 31(11):3138-3147.

Yang, Y., and McCarty, P. L. 1998. "Competition for Hydrogen within a Chlorinated Solvent Dehalogenating Anaerobic Mixed Culture." Environmental Science & Technology. 32:3591-3597.

Appendices

#### Appendix A

#### Soil Respirometry Test Procedure OU-1 Electron Donor Study

		_
Equipn	nent:	Te
12	250mL E-flasks	3
24	glass tubes	3
12	2 hole stoppers	2
12	glass tube septum	2
12	manometer hoses	2
1	ruler	
12	syringes (5mL w/21 guage ne	edles)
300g	Silica sand	
1200g	OU-1 Soil	
1	Sterile Filter Apparatus	
2 ea	filters (sequential down to 0.2	um)

Tests:

Soil, Amended Water, + Acetate Soil, Amended Water, + Propanol Sand, + D.W. (Thermal Barometer) Soil, Amended Water, Acetate, + Sodium Azide Soil, Amended Water, Propanol, + Sodium Azide

Concentrations: Acetate = TBD based on solubility Propanol = 5.22mL/L

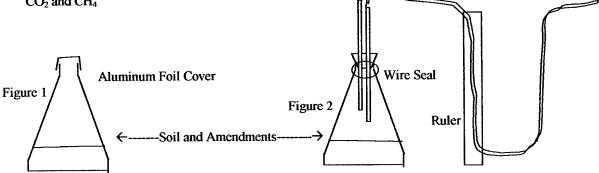
Procedure:

1

- Add 150 g aquifer soils to two 250 mL flasks, cover with aluminum foil and autoclave for 30 minutes
- Autoclave glassware, hoses, stoppers, filters for 30 minutes
- Filter the acetate or propanol amended groundwater to sterilize
- Filter the acetate or propanol amended groundwater and sodium azide to sterilize
- Prep the anaerobic glovebox

Balance

- Place all items in the glovebox and seal
- Fill and purge the glovebox with nitrogen gas twice
- Fill the glove box with 95% nitrogen/5% hydrogen
- Turn on oxygen scavengers in the glovebox
- In an anaerobic glovebox prepare test microcosms, See Figure 1
  - Allow soils to drain excess water onto sterilized worksurface
  - Place 150 g of OU-1 soil in the flask
  - Add 30 mL (or enough to fill soil pore space, 60 mL for the distilled water flasks) of the filtered water containing acetate or propanol to the appropriate flasks
  - Add 30 mL (or enough to fill soil pore space) of the filtered water containing acetate or propanol and sodium azide to the appropriate flasks
- Leave all microcosms covered with aluminum foil in the anaerobic glovebox for five days to acclimate
- After the acclimation period, while still in the anaerobic glove box, stopper the microcosms and add the sampling glass tube and the manometer tubing assembly
- Remove the microcosms from the anaerobic glovebox and secure the stopper and manometer lines to minimize air transfer and facilitate pressure readings, add 5mL salt water (<75% solubility limit of sodium chloride) to the manometer lines to make reading possible, See Figure 2
- Seal the stoppers into the flasks using copper wire
- Collect gas samples from each flask using the syringe, analyze using GC/TCD for CO<sub>2</sub> and CH<sub>4</sub>
- Take manometer readings every 4 hours for 48 hours or as needed based on preliminary observations
- At the end of 48 hours, collect gas samples from the sampling tube and analyze using GC/TCD for CO<sub>2</sub> and CH<sub>4</sub>



Soil Test Measurements 23 Nov 98

		Carbon Dioxide			Oxygen		Methane		
	Peak Area	Calculated %	Calculated	Peak Area	Calculated %	Calculated	Peak Area	Calculated %	Calculated
	t=0.95		ppm		Concentration	opm	t=4.95		ppm
DW1	0	-0.36	-3581.44	8800598	3.40	34002.99	0	0.08	798.08
DW2	õ	-0.36	-3581.44	5324939	1.66	16624.70	0	0.08	798.08
SWP1	•	-0.36	-3581.44	5912176	1.96	19560.88	0	0.08	
SWP2		-0.36	-3581.44	4519845	1.26	12599.23	0	0.08	
SWP3		-0.36	-3581.44	5356726	1.68	16783.63	0	0.08	
SWP NaAZ1		-0.36	-3581.44		2.09	20895.02	0	0.08	
SWP NaAZ2		-0.36	-3581.44		2.10	21001.05	0	0.08	798.08
SWA1	2690450		9870.82		1.36	13628.11	0	0.08	798.08
SWA2	3199318		12415.16		2.35	23524.46	0	0.08	<b>798.08</b>
SWA2 SWA3	2600449		9420.81		2.13	21344.34	0	0.08	798.08
SWA NaAZ1	1893040		5883.77		2.83	28250.39	0	0.08	798.08
SWA NaAZI	1878442		5810.78		1.59	15941.86	đ	0.08	798.08

Soil Test Measurements 25 Nov 98

	<b></b>	Carbon Dioxide			Oxygen			Methane	
	Peak Area	Calculated %	Calculated	Peak Area	Calculated %	Calculated		Calculated %	Calculated
	t=0.95	Concentration	ppm	t=1.86	Concentration	ppm			ppm
DW1	1913724	0.66	6589.89	14096789	6.66	66644.13		0.03	
DW2	0	-0.30	-2978.73	11773353	5.50	55026.95		0.02	
SWP1	-	-0.30	-2978.73	15348802	7.29	72904.20	3037	0.02	
SWP2		-0.30	-2978.73	7624430	3.43	34282.34		0.02	
SWP3		-0.30	-2978.73	9651589	4.44	44418.13	13651	0.02	203.90
SWP NaAZ1		-0.30	-2978.73		7.19	71898.29		0.02	169.77
SWP NaAZ2		-0.30	-2978.73		3.28	32811.55		0.02	169.77
SWA1	4458044	1.93	19311.49		2.74	27407.22		0.02	169.77
SWA1	4608720		20064.87		2.85	28535.67		0.02	169.77
<b>•</b> • • • • • •	3786258		15952.56		5.36	53621.50		0.02	169.77
SWA3	3396582			••••				0.02	169.77
SWA NaAZ1 SWA NaAZ2	2990252		11972.53					0.02	169.77

Soil Test Measurements 30 Nov 98

	Γ	Carbon Dioxide			Oxygen		Methane			
		00.00.010100								
	Peak Area	Calculated %	Calculated	Peak Area	Calculated %	Calculated	Peak Area	•••••	Calculated	
	t=0.95		ppm	t=1.86	Concentration	ppm	t=4.95		ppm	
DW1	1632316	0.65	6501.87	9530478	9.01	90120.32	32878	0.08	770.54	
DW2	1369356	0.52	5187.07	14266925	13.75	137484.79	0	0.07	688.35	
SWP1	4920965	2.29	22945.12	12341292	11.82	118228.46	19074	0.07	736.03	
SWP2	2495829	1.08	10819.44	931625	0.41	4131.79	56845	0.08		
SWP3	3720578		16943.18		6.23	62348.48	35701	0.08	777.60	
SWP NaAZ1	2566296		11171.77		2.34	23403.74		0.07	688.35	
SWP NaAZ2	3188980		14285.19			6388.87		0.07	688.35	
SWP Na-22 SWA1	5750165		27091.12			48727.66		0.07	688.35	
SWA1	6386175		30271.17					0.07	688.35	
	6120172		28941.15					0.07	688.35	
SWA3	5582297	2.63				-		0.07	688.35	
SWA NaAZ1 SWA NaAZ2	4517312				3.40			0.07	688.35	

t = sample run time to representative peak DW = Sand and Distilled Water

SWP = Soil, Amended Water, and n-Propanol SWPNaAZ = Soil, Amended Water, n-Propanol, and Sodium Azide

SWA = Soil, Amended Water, and Acetate

SWANaAZ = Soil, Amended Water, Acetate, and Sodium Azide

Comparison of Values Table

Day		DW1	DW2	SWP1	SWP2	SWP3	SWP NaAZ1	SWP NaAZ2	SWA1	SWA2	SWA3	SWA NaAZ1	SWA NaAZ2
Ali V	alues in p Carbon D	•											
1	23-Nov	-3581	-3581	-3581	-3581	-3581	-3581	-3581	9871	12415	9421	5884	5811
3	25-Nov	6590	-2979	-2979	-2979	-2979	-2979	-2979	19311	20065	15953		
8	30-Nov	6502	5187	22945	10819	16943	11172	14285	27091	30271	28941	26252	20927

Ovaen

1	23-Nov	34003	16625	19561	12599	16784	20895	21001	13628	23524	21344	28250	15942
3	25-Nov	66644	55027	72904	34282	44418	71898	32812	27407	28536	53621	57153	22320
8	30-Nov	90120	137485	118228	4132	62348	23404	6389	48728	51487	97215	96293	33986

 Methane
 1
 23-Nov
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 798
 79

798 170

688

798 170 688

Soil Test Calibration Measurements 23 Nov 98

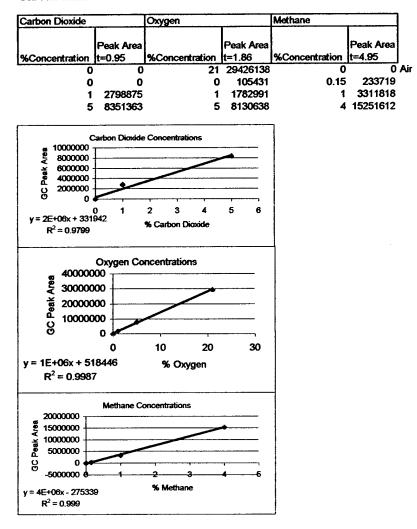
Carbon Dioxide		Oxygen	<u> </u>	Methane	r
N 0	Peak Area	%Concentration	Peak Area	%Concentration	Peak Are
%Concentration 0	t=0.95 0	21			1=4.95
0	0	21			34759
1	4986511	1	2655040		463109
5	12755675	5	12183233		2158442
c	arbon Dioxide	Concentrations		]	
g 1500000	T				
8 15000000 ¥ 10000000 5000000 0 0					
a 5000000					
8 0					
y = 2E+06x + 716	<del>9</del> 87 1	2 3 4	56	.†	
$R^2 = 0.9599$		% Carbon Dioxide			
	Oxygen Co	ncentrations			
<del>g</del> 400000	1		~		
₹ 200000 ₹ 200000 0 100000					
୍ଷି 200000					
ິບ 100000		·			
-	0	40 45	, ,		
y = 2E+06x +			20 25		
R <sup>2</sup> = 0.99	07	% Oxygen			
	Methane Co	oncentrations			
_ 30000000	<b>.</b>				
20000000			<b>.</b>		
₹ 20000000 ₹ 10000000 0 0	ł				
0 -10000000	T .	2 3	4		
y = 5E+06x - 3990		% Methane			
v - 3554	140			1	
$R^2 = 0.9988$					

.

Soil Test Calibration Measurements 25 Nov 98

Carbon Dioxide		Oxygen		Methane	
	ak Area		Peak Area	N O testion	Peak Area
%Concentration t=0		%Concentration	t=1.86 33549276	%Concentration 0	t=4.95 0
0	0	21 0		0.15	•
	043697	1	2382692	0.13	4726118
	090805	5		-	17797927
Carboo 5 15000000 4 10000000 5 500000 0 0 y = 2E+06x + 59574 $R^2 = 0.9558$		2 4 % Carbon Dioxide	6		
Oxy s 40000000 ₹ 30000000 a 20000000 C 10000000 C 10000000		ncentrations			
y = 2E+06x + 76796		10 20	30		
R <sup>2</sup> = 0.9985 M 20000000 2 1500000 3 1000000 3 1000000 4 1000000 5 5000000 0 500000 0 5000000 0 500000 0 500000 0 5000000 0 50000000 0 5000000 0 50000000 0 5000000 0 5000000 0 5000000 0 5000000 0 5000000 0 5000000 0 5000000 0 50000000000	ethane Co	% Oxygen Oncentrations	▲ 		

Soil Test Calibration Measurements 30 Nov 98



### Appendix B Electron Donor Equations and Hydrogen Release Calculations

			0		
Selected [	Donors				H eqs
Butyric Ac	id (Butyrate)	CH <sub>3</sub> CH <sub>2</sub> CH <sub>2</sub> COO <sup>-</sup> + 7H <sub>2</sub> 0> 3	3CO <sub>2</sub> + HCO <sub>3</sub> <sup>-</sup> + 20H <sup>+</sup>	+ 20e <sup>-</sup>	
M.W. 88.1	-				
(slow) fatty	y acid		2 H <sub>2</sub> /mole		
• • •	CH <sub>3</sub> CH <sub>2</sub> CH <sub>2</sub> COOH+	2H <sub>2</sub> O> 2CH <sub>3</sub> COOH+2H <sub>2</sub>	20 ee/mole		0.100 H <sub>2</sub> /ee
	•				
Benzoic A	cid (Benzonate)	$C_6H_5COO^- + 13H_20 -> 6CO_2^-$	+ HCO <sub>3</sub> ⁻ + 30H <sup>+</sup> + 30∉	e	
M.W. 122	.12 aromatic acid				
		_	3 H <sub>2</sub> /mole		
	C <sub>6</sub> H <sub>5</sub> COOH+6H <sub>2</sub> O -	$\rightarrow$ 3CH <sub>3</sub> COOH + 3H <sub>2</sub> + CO <sub>2</sub>	30 ee/mole		0.100 H <sub>2</sub> /ee
		•			
				10-	
	d (Lactate)	$CH_3CHOHCOO^- + 4H_20 \rightarrow 20$	$CO_2 + HCO_3 + 12H^{-1}$	+ 12e	
	)8 fatty acid				
(fast)	•	$H_2 \rightarrow CH_3CH_2COOH$	2 H <sub>2</sub> /mole		0.167 H <sub>2</sub> /ee
(słow)		$l_2O \rightarrow CH_3COOH + 3H_2$	12 ee/mole		
	CH <sub>3</sub> CHOHCOOH +	$2H_2O \rightarrow 2H_2 + CH_3COOH$	:	slow release	0.111 H <sub>2</sub> /ee
		CH3CH2COO <sup>-</sup> + 5H20> 2CC		10	
•	Acid (Propionate)	$CH_3CH_2COO + 5H_20 = 2CC$	<sup>2</sup> • 1100 <sub>3</sub> • 1 <del>4</del> 11 • 1		
M.VV. 74.	08 fatty acid		3 H <sub>2</sub> /mole		
(-1			14 ee/mole		0.214 H <sub>2</sub> /ee
(slow)			14 cc/more		
n-Propano	h	CH3CH2CH2OH + 5H20> 30	CO₂ +18H <sup>+</sup> + 18e <sup>-</sup>		
M.W. 60.0			-		
(fast)		$_{2}O \rightarrow CH_{3}CH_{2}COOH + 2H_{2}$	5 H <sub>2</sub> /mole		
(slow)	•	$H_2O \rightarrow CH_3COOH + 3H_2$	18 ee/mole	total release	0.278 H <sub>2</sub> /ee
(,		H <sub>2</sub> O> CH <sub>3</sub> COOH + 5H <sub>2</sub>		slow release	0.167 H <sub>2</sub> /ee
		2 2 2			· · · · · · · · · · · · · · · · · · ·
Toluene		C <sub>7</sub> H <sub>8</sub> + 14H <sub>2</sub> O> 7CO <sub>2</sub> + 36	H <sup>+</sup> + 36e <sup>-</sup>		
M.W.92.1	3				
(fast)	C <sub>7</sub> H <sub>8</sub> + 2H <sub>2</sub> O> C <sub>6</sub>	-	6 H <sub>2</sub> /mole		
(slow)	C <sub>6</sub> H <sub>5</sub> COOH+6H <sub>2</sub> O -	$\rightarrow$ 3CH <sub>3</sub> COOH + 3H <sub>2</sub> + CO <sub>2</sub>	36 ee/mole	total release	0.167 H <sub>2</sub> /ee
	C <sub>7</sub> H <sub>8</sub> + 8H <sub>2</sub> O> 3C	$H_3COOH + 6H_2 + CO_2$		slow release	0.083 H <sub>2</sub> /ee
Acetate	(Soil Test)	$CO_2$ + $HCO_3^-$ + $8H^+$ + $12e^ \rightarrow$			
M.W.60.0	)5	-	1 H <sub>2</sub> /mole		
	CH <sub>3</sub> COO <sup>-</sup> + H> C	$H_4 + CO_2$	8 ee/mole	total release	0.125 H <sub>2</sub> /ee

#### Appendix C Electron Donor Delivery Calculations

Molar Concentrations of Donors Based on  $H_2$  Demand and slow  $H_2$  fermentation values Worst Case Contaminat Concentrations from Remedial Investigation Report (Montgomery-Watson, 1995)

Demand = ((mol H<sub>2</sub>demand/I)/mol H<sub>2</sub> donor/ee)\* 1mol donor/ee

4.06E-03

Donor Concentration = X : Donor Flow Rate (X) = Total Flow (Conc Needed)

	mol H <sub>2</sub> /i	mol H <sub>2</sub> /ee	ee/1mol	(mol/l) donor	(mg/l) donor	Donor Bottle		Total Donor	Total Donor
	Demand	Donor	Donor	Needed	mol/l*mg/mol	Concentratio	n	Needed	Needed (ml)
	Dornario	2 0.10.				(mol/L)	(mg/L) <i>ml</i>	(mg)	mg/(SG*1000)
n-Butyric Acid (Butyrate)	4.06E-03	0.100	20	2.030E-03	178.88	1.051E-01	9261.292	13141.773	13.6325
S.G. : 0.964							9.6071		
Benzoic Acid (Benzonate)	4.06E-03	0.100	30	1.353E-03	165.27	7.007E-02	8556.430	12141.575	9.5905
S.G. : 1.266							6.7586		
Benzoate calculations due to s	solubility lim	its of crysta	benzoic ac	id. Donor 1/4 a	s concentrated	and supplied	at 4 times t	he normal don	or feed rate
Benzoic Acid (Benzonate)	4.06E-03			1.353E-03	165.27	1.853E-02		3211.281	2.5366
Barzore Alda (Barzondio)							1.7876		
Lactic Acid (Lactate)	4.06E-03	0.167	12	2.026E-03	182.50	1.049E-01	9448.388	13407.263	10.7258
S.G. : 1.250	4.002-00	0.101					7.5587		
Propionic Acid (Propionate)	4.06E-03	0.214	14	1.355E-03	100.39	7.016E-02	5197.401	7375.112	7.4271
S.G. : 0.993	4.002-00	0.211					5.2340		
n-propanol	4.06E-03	0.167	18	1.351E-03	81.16	6.993E-02	4201.848	5962.422	7.4067
S.G. : 0.805	1.002.00		1				5.2197		
Toluene	4.06E-03	0.083	36	1.359E-03	125.18	7.035E-02	6481.083	9196.656	10.6074
S.G. : 0.867	4.002-00	0.000					7.4753		
Solubility limits of toluene = 0.	<u>L</u> 577ml/I Г	l Jonor at sati	Ination limit	is supplied at 4	times the nor	nal donor fee	d rate but sti	II 4X <demand< td=""><td></td></demand<>	
4X Feed Rate	4 06E-03	0.083	36	1.359E-03	125.18	1.861E-02	1714.158	2432.390	2.8055
4/ 1000 1000	4.002.00	0.000				1	1.9771		
Actual Toluene		0.083	36				499.9989		
Actual Toldene	0.001184	Best Possi					0.5767		
Apotato	4.06E-03				367.17	3.166E-01	19009.558		
Acetate	4.002-00	0.000	<u> </u>	1			18.1389	1	
(Soil Testing) S.G.: 1.048	L	L		L			L	·	

Predicted Volume of Donor Needed:

Donor Solution Rate ml/min \* 10080 min/wk \* 32 wk/exp =

1419.26 ml/exp 1.42 L/exp

Predicted Total Fluid:

Total Solution Rate ml/min \* 10080 min/wk \* 32 wk/exp =

 wk/exp =
 73479.17 ml/exp

 73.48 L/exp
 Per Column

 X 8 Columns
 587.83 L/exp
 Total Experiment

Vitamins: For Vitamin Dema	and (ma/L):		Needed (mg)	(g/exp)
	10	587.832	5878.32	5.88
	20	587.832	11756.64	11.76
	50	587.832	29391.6	29.39
F	100	587.832	58783.2	58.78
Resazurin	1	587.832	587.832	0.59
Yeast	20	587.832	11756.64	11.76

#### Appendix D Hydrogen Demand Calculations

Concentrations from OU-1 Comprehensive RI Report Section 8

DCE Groundwater – up to 42,000 ug/L -Total DCE DCE Soil – up to 4.2 mg/kg -Total DCE

Worst Case V	Vell Data						1
	Molecular					eq H Demand	
Contaminant		Conc (ug/L)	Molar Conc (mol/L)	H equivalents	e equiv	mol H <sub>2</sub> /liter	ee/liter
PCE	165.82		3.50E-07	4	8	1.40E-06	2.80E-06
TCE	131.38		1.75E-05	3	6	5.25E-05	1
DCE	96.94		And the second se	2	4	8.67E-04	
VC	62.5			1	2	3.84E-05	7.68E-05
<b>V</b> C						9.59E-04	1.92E-03

TCA	133.4	2000	1.50E-05	3	6	4.50E-05	
DCA	98.96	520	5.25E-06	2	4	1.05E-05	2.10E-05

Iron Chlorinated Benzenes etc

	1.01E-03	2.03E-03
Multiply by Safety Factor	4	4
Equivalent Hydrogen Demand	4.06E-03	
Equivalent Electron Demand		8.11E-03

Flow Rates		
Feed Solution:	0.2234 ml/min	
Donor Solution:	0.0044 ml/min	
Total Feed Rate:	0.2278 ml/min	_
	mol H <sub>2</sub> /liter demand	X 1mol

mol H<sub>2</sub>/ee donor ee/mol donor

#### Appendix E Electron Donor Properties and Actual Delivery Calculations

Measured TOC Levels Based on Equivalent Hydrogen Demand Calculations

	Butyrate	Benzoate	Lactate	Propionate	Propanol	Toluene
TOC (mg/L)	9261.29	8556.43	9448.39	5197.40	4201.85	
M.W.	88.12	122.12	90.08	74.08		
Carbon Weight	48.00	84.00	36.00	36.00		
Equivalent Carbon	0.54	0.69	0.40			
As Carbon (mg/L)	5044.73	5885.52	3776.00	2525.73	2517.33	
As Carbon Based on Solubility (mg/L)		1556.60				455.88
Analytically Measured TOC						
25-Nov-98						
TOC (mg/L)	3400.00	1100.00	2600.00	2500.00	1800.00	2.30
% Recovery						
Based on Predicted Loading	67.40		68.86	98.98	71.50	
Based on Solubility Limits		70.67				0.50
Equivalent Donor Supplied (mg/L)	6241.83				3004.50	
% of Predicted Actually Supplied	67.40	18.69	68.86	98.98	71.50	0.04
2-Feb-99						
TOC (mg/L)	5900.00	1900.00	3700.00	3000.00	2600.00	130.00
% Recovery						
Based on Predicted Loading	116.95		the second s	118.78	103.28	
Based on Solubility Limits		122.06				28.52
Equivalent Donor Supplied (mg/L)	10831.42					
% of Predicted Actually Supplied	116.95	32.28	97.99	118.78	103.28	2.20
Average Equivalent Donor Supplied	8536.63	2180.71	7882.00	5658.89	3672.17	72.55

#### Appendix E Electron Donor Properties and Actual Delivery Calculations

	Molecular	Specific Grav	ity***	Solubility	
	Weight**	•	Reference		Reference
Donor Properties*				g donor/100g H2	0 Temperature
n-Butyric Acid (Butyrate)	88.1	0.964	20/4	miscible	
Benzoic Acid (Benzonate)	122.12	1.266	15/4	c	.2 17
Lactic Acid (Lactate)	90.08	1.249	15/4	miscible	
Propionic Acid (Propionate)	74.08	0.992	20/4	miscible	
n-propanol	60.09	0.804	20/4	miscible	
Toluene	92.13	0.866	20/4	0.	05 16
Spike		1.00	1514		35
cis-DCE****	96.94	1.29	15/4	0.	

Soil Test

60.05	1.049	20/4	In	niscible		
58.44	2.163	20/4				0 100
44.01	1.53	20/4				0 20
16.01	0.7491		0 18.7 184	C	.4	20
	44.01	58.44       2.163         44.01       1.53         16.01       0.5547         0.7491	58.44       2.163       20/4         44.01       1.53       20/4         16.01       0.5547       0.7491	58.44       2.163       20/4         44.01       1.53       20/4       1         16.01       0.5547       0         0.7491       18.7	58.44         2.163         20/4         35           44.01         1.53         20/4         179.7cc           16.01         0.5547         0         0	58.44       2.163       20/4       35.7         44.01       1.53       20/4       179.7cc         16.01       0.5547       0       0.4         0.7491       18.7       0       0.4

\* Data taken form Perry's Chemical Engineer's Handbook, 7th Edition,

\*\* Molecular Weights based on the 1941 Atomic Weight Values

\*\*\* Chemical density at given temperature referred to water at second temperature

## Appendix F Material and Chemical Inventory

	HAFB OU-1 Treatability	HAFB OU-1 Treatability Study Using Electon Donor Column Microcosms	or Column Microcosm						
	1 w/inst RW: 1 w/GW and Nutrients: 5 w/GW. Nutrients. and Donors	o Corumn System trients: 6 w/GW. Nutrients	. and Donors						
*	Item Description	Vendor	Part/Vendor #	Vendor Phone #	ğ	Projected Unit Cost	Total Cost	Purchaser	Total Cost to USAF
u	True Ground Reservoir	Fisher	02-887-1	1-800-766-7000	+	133.40	133.40	HIII AFB	133.40
T	Main Feed/Sample Col	Fisher	02-887-5	1-800-766-7000	2	170.70	341.40	HIII AFB	341.40
T	5di H20 Collection Carboys	Straw Ibis	Local		2	20.00	40.00	Self	Self Pald
T	Teflon Stopper Billets	Teftech	Custom	1-800-677-6854	Э	100.00	100.00	Self	Self Paid
	"O" Rings for Reservoirs	Ace Glass	7855-845	1-800-626-5381	2	20.89	41.78	HIII AFB	41.78
1	"O" Rings for Reservoirs	Ace Glass	7855-844	1-800-826-5381	-	20.00	20.00	Hill AFB	20.00
	1/4" SS Tube 50' Coll	Altech	30309	1-800-255-8324	2	190.00	380.00	HIII AFB	380.00
	Manifold 1/4" T	Salt Lake Valve&Fit	SS-400-3	1-801-266-3560	9	15.70	94.20	HIII AFB	94.20
	Manifold 1/4" Elbow	Salt Lake Valve&Fit	SS-400-9	1-801-266-3560	2	11.10	22.20	HIII AFB	22.20
a	Peristattic Pump	Watson-Marlow	020.5524.00A	1-800-282-8823	+	4450.00	4450.00	HIII AFB	4450.00
٩	Pump Mainfold Tubing	Wetson-Marlow	984.0019.000	1-800-282-8823	5	39.50	79.00	HIII AFB	79.00
a.	Pump Mainfold Tubing	Wetson-Marlow	984.0013.000	1-800-282-8823	1	39.50	39.50	Hill AFB	39.50
۵.	Pump Mainfold Tubing	Watson-Marlow	984.0038.000	1-800-282-8823	1	39.50	39.50	Hill AFB	39.50
٩	Pump Mainfold Tubing	Watson-Marlow	984.0142.000	1-800-282-8823	5	39.50	197.50	Hill AFB	197.50
u	Donor Feed Reservoir 12/cs	Fisher	06-421-13	1-800-766-7000		65.36	65.36	Hill AFB	65.36
Ľ	Donor/Feed Line T 1/4"	Saft Lake Valve&Fit	SS-400-3	1-801-266-3560	6	15.70	94.20	Hill AFB	94.20
. 0	Glass Columns	Ace Glass	5820-37	1-800-626-5381	8	34.86	278.88	Hill AFB	278.88
	Column End Fitting	Ace Gass	5802-37	1-800-626-5381	16	45.75	732.00	HIII AFB	732.00
0	Glass Wool	Altech	4034	1-800-255-8324	7	23.00	46.00	HIII AFB	46.00
0	Swage Column/Tube Adapter	Saft Lake Valve&Fit	SS-400-1-4	1-801-266-3560	16	4.90	78.40	HIII AFB	78.40
D/C	17 Guage Needles	Fisher	14815605	1-800-766-7000	9	24.00	72.00	HII AFB	72.00
D/C		Fisher	14815621	1-800-766-7000	3	40.00	120.00	HII AFB	120.00
D/C		Fisher	NC-9436352	1-800-766-7000	32	13.50	432.00	HIII AFB	432.00
D)d		Saft Lake Valve&Fit	SS-400-6	1-801-266-3560	5	7.60	15.20	Self	Self Pakd
0 V		Salt Lake Valve&Fit	SS-400-6	1-801-266-3560	õ	7.60	228.00	Hill AFB	228.00
0/4		Saft Lake Valve&Fit	SS-405-2	1-801-266-3560	7	1.50	3.00	Self	Self Paid
0)d	Swage Barbed Fittings	Salt Lake Valve&Fit	SS-405-2	1-801-266-3560	30	1.50	45.00	Hill AFB	45.00
ш	Effluent In-line/Sampler Btis	Fisher	05-719-121	1-800-766-7000	9	72.00	432.00	HII AFB	432.00
ш	Waste Reservoir	Fisher	02-961B	1-800-766-7000	-	56.00	56.00	HII AFB	56.00
S	Tube Cutter	Altech	13995	1-800-255-8324	-	49.00	49.00	Hill AFB	49.00
S	Tube Bender	Aittech	357090	1-800-255-8324	-	2.00	2.00	HII AFB	2.00
Ľ	Female 1/4-1/4Swage,N2 line	Salt Lake Valve&Fit	SS-400-7-4	1-801-266-3560	2	8.00	16.00	HIII AFB	16.00
L	Nitrogen Cylinder, 1 yr	Whitmore Gas		1-801-753-1111	-	70.00	15.00	USU	nsu -
u	Ntrogen Gas Refilis	Whitmore Gas		1-801-753-1111	4	7.00	28.00	Ser	Sert Paid
L	N2 Combined Regulator	Whitmore Gas		1-801-753-1111	-	70.00	70.00	USU 1	USU .
S	Mounting System	USU Shop			-	120.00	120.00	Self	Self Paid
ш	Sample Syringe, 60 ml	Fisher			80	5.00	40.00	Self	Self Paid
L	Chemicals (see separate pg)				-	720.89	720.89	USU/UWRL	USU/UWRL
	Glasswork				ω	35.00	280.00	Hill AFB	280.00
. u	Septum/Stopper	Chem Store			02	0.29	20.30	Self	Self Paid
. <b>u</b>	3/4" Tvaon Tubina	Chem Store			9	2.75	16.50	nsu	USU
·	Local Sampling Glassware	UWRL				200.00	200.00	USU/UWRL	USU/UWRL
	Soil Testing Setups	UWRL			12	30.00	360.00	UWRL	USSU/UWRL
	Total AF Allowed \$8955			TOTAL COST			10054.2		8865.32
				Page 69					

.

Bail

## Appendix F Material and Chemical Inventory

		I I I I I I I I I I I I I I I I I I I							
		8 Column System							
	1 w/just GW; 1 w/GW and Nutrients; 6	trients; 6 w/GW, Nutrients, and Donors	s, and Donors						
*	Item Description	Vendor	Part/Vendor #	Vendor Phone #	Qţ	Projected Unit Cost	Total Cost	Purchaser	
	Donors								
	n-Propanol	Mailinckrodt	7169 (500ml)		1	4.71	4.71	nsn	
	Propionic Acid	Mailinckrodt	7179 (500ml)		+	5.78	5.78	nsn	
		Fisher	A162-500 (500ml)	1-201-796-7100	ţ.	6.87	6.87	nsn	
	Butyric Acid	Acros	10800-100 (100ml)	1-800-227-6764	1	10.64	10.64	nsn	
	Benzoic Acid	Fisher	A-65 (100g)	1-201-796-7100	0	8.00	0.00	UWRL	
]		Fisher	T290-1 (1000ml)	1-201-796-7101	0	10.65	0.00	UWRL	
	Vitamin Solution								
1	d-biotin	Sigma	B 4501 (10g)	1-800-325-3010	+	264.80	264.80	nsu	
	folic acid	Acros	21663-0100 (10g)	1-800-227-6764	-	11.30	11.30	nsu	
1	pyridoxine hydrochloride	Acros	15077-0500 (50g)	1-800-227-6764	+	24.55	24.55	nsu	
	thiamin hydrochloride	Sigma	T 4625 (25g)	1-800-325-3010	0	10.30	0.00	UWRL	
	riboflavin	Kodak	5181 (100g)	1-800-325-3010	0		0.00	UWRL	
	nicotinic acid	Aldrich	N785-0 (100g)	1-800-325-3010	0		0.00	UWRL	
	1	Sigma	P 5710 (25g)	1-800-325-3010	2	21.40	42.80	usu	
	balamin)	Sigma	V 2876 (5g)	1-800-325-3010	-	10	162.25	usu	
	nzoic acid	Sigma	A 0129 (100g)	1-800-325-3010	-	20.96	20.96	USU	
		Acros	13872-0250 (25g)	1-800-227-6764	-	120.39	120.39	USU	
	Ammendments								
	resazurin	Sigma	R 2127 (1g)	1-800-325-3010	1	10.23	10.23	USU	
		Sigma	YSC-1 (100g)	1-800-325-3010	1	23.38	23.38	nsn	
	sodium bicarbonate	Sigma	S 6014 (500g)	1-800-325-3010	0	13.40	0.00	UWRL	
	Shibbing					12.23	12.23		
						779.74	720.89	USU Total	
	cis Dichloroethene	Supelco	48597 (LA-78817)(1g)	1-814-359-3441	+	44.91	44.91	Self	
						824.65	765.80	Grand Total	
	Note* Cty 0 denotes chemicals obtained	obtained from UWRL excess	55						

Appendix G Column Flow Calculations and Microcosm HRTs

	ter Horizontal	l Report for Ol Linear Velocity w/Deborah D	pg 5-18		atson, values in	clud	e porosity	
	ie velocity s Low	tated is (Q/A)/p High		Average				
becomes	310 ft/yr 0.85 ft/dy	4656 ft/ут 12.76 ft/dy		1950 ft/yr 5.3 ft/dy				
	0.018 cm/m	nir 0.27 cm/mir	Ì	0.113 cm/m	iin			Average
Assume P	orosity = 0.4							Darcy Velocity
	•	'm 0.108 cm/m	in	0.045 cm/m	in			0.045 cm/min
Column Di	imensions ID = 25 mn	n = D	Area = (r	$D^{2} / 4 =$	490.87 mm <sup>2</sup>		Col Area	4.91 cm <sup>2</sup>
	Length = $60$			=((pD <sup>2</sup> )/4)L			Col Vol	294.6 cm <sup>3</sup>
Primary Fl	(Interstitial	ng Average Vel = Q/A/e) a = 0.113 * 4.91		5400 om <sup>3</sup> (mi	_	=		0.550 ml/min
	Flow - Area	3 = 0.115 * 4.91	CHI = 0.3	5499 Citt /illi	1	-		Solution
	(Darcy) Flow * Area	a = 0.045 * 4.91	cm <sup>2</sup> = 0.2	221 cm <sup>3</sup> /min		æ		Flow Rate 0.221 ml/min
Flow per c	olumn per we	eek				=		2.23 L/wk
Donor Flo		'min * 5% = 0.0	11 ml/min			=		Donor Flow Rate 110.9 ml/wk 0.011 ml/min
1/4" 316 8		I Tubing Area		2				
	Area ≃ (pD Q/A = (0.2	<sup>2</sup> )/4 = 21 cm <sup>3</sup> /min)/0.3	817 cm <sup>2</sup> =	0.317 cm <sup>2</sup> 0.697 cm/r	nin			
HRT: Em	pty Bed Colu Flow =	mn, Ideal Flow 0.221 + .01	1 = 0.23	2			HRT in mins	HRT in day
	HRT =	Col Vol/Flov	v	294.6 cm <sup>3</sup>	/0.232 ml/min	=	1269.8	0.882
HRT: Actu	ual Microcosn Flow =	n Values Based 0.2234 + 0.		278				
0	HRT =	Col Vol/Flor assume poros	-		/0.2278 ml/min	=	1293.24	0.898

#### Appendix G Column Flow Calculations and Microcosm HRTs

HRT: Actual Microcosm Values

Column Cross Sectional Are	ea = 490.87mm <sup>2</sup>	
Column Height =	600 mm	
Assume Porosity =	0.4	
Col Vol =	294.6 c	:m3
HRT =	= Col Vol/Flow	

Corrected for porosity; assume porosity is 0.4 for the sandy soil HRT = (Col Vol\*porosity/Flow)

		Groundwater	Amended	n-Butyrate	Benzoate	Lactate	Propionate	n-Propanol	Toluene
	Column Area (cm <sup>2</sup> )	4.91	4.91	4.91	4.91	4.91	4.91	4.91	4.91
	Sediment Depth							-	
	(inches)	16.75	17.25	18.25	17.75	16.75	18.75	18.00	18.25
	Sediment Depth								
	(cm)	42.55	43.82	46.36	45.09	42.55	47.63	45.72	46.36
	Sediment Volume	000.04	245.07	227.54	224 24	208.84	233.78	224.43	227.54
	(cm3) Water Volume in	208.84	215.07	227.54	221.31	200.04	233.70	224.43	227.34
		47.00	47 50	40 54	18.03	17.02	19.05	18.29	18.54
	Sediment (cm <sup>3</sup> )	17.02	17.53	18.54					13.65
	Supernatant Depth Water Volumn in	17.46	16.19	13.65	14.92	17.46	12.38	14.28	13.03
		05.00	79.45	66.98	73.21	85.68	60.75	70,10	66.98
	Supernatant (cm <sup>3</sup> ) Total Water Volume	85.68	19.43	00.90	13.21	05.00	00.75	70.10	00.30
	(cm <sup>3</sup> )	102.70	96.97	85.52	91.25	102.70	79.80	88.38	85.52
<b>D</b> -4-		102.70	30.37	00.02	51.25	102.70	13.00	00.00	
Date	Measured Flow								
22_Nov	(ml/min)	0.250	0.260	0.242	0.232	0.235	0.229	0.257	0.250
22-1404	HRT (min)	410.797	372.974		393.307	437.019	348.450		342.085
	HRT(day)	0.285	0.259	· · · ·	0.273	0.303	0.242	0.239	0.238
		0.200							
	Measured Flow							<b> </b>	
11. ian	(ml/min)	0.215	0.223	0.234	0.226	0.266	0.215	0.212	0.212
11-Vall	HRT(min)	477.671	434,858						403.402
	HRT (day)	0.332	0.302	0.254	0.280	0.268	0.258	0.290	0.280
	1 1 1 1 1							[	ļ
4 5 - 1	Measured Flow (ml/min)	0.211	0.228	0.228	0.241	0.231	0.224	0.224	0.241
1-reb	· · · · · · · · · · · · · · · · · · ·							<u>.</u>	354.860
	HRT(min)	486.727	425.322						
	HRT (day)	0.338	0.295	0.260	0.263	0.309	0.247	0.274	
Ave Flow		0.225	0.237	0.235	0.233	0.244	0.223	0.231	0.234
Overall A	verage Flow								0.233
Deviation	from Average Flow	0.007	-0.004	-0.002	0.000	-0.011	0.010	0.002	-0.002
	· · · · · · · · · · · · · · · · · · ·	h		• • • • • • •	· · · · · · · · · · · · · · · · · · ·		· · · · · · · · · · · · · · · · · · ·		

ะโ	e		6.78 7.19	7.11 7.29	7.09	9.11 9.25	9.13	9.20	9.06 9.05	8.85 9.07	9.53	9.43	9.33 9.40	9.16 0.22	9.22 9.16	8.76	9.25	9.23	7.88	7.64	7.33	7.29	7.14	7.37
Don	Toluen (Col 8)																							
and Electro	n-Propanol Toluene (Col 7) (Col 8)		6.79 7.07	7.00	7.07	9.06 0.21	9.07	9.14	8.96	8.84 9.92 9.92	9.42	9.33	9.24	9.05	9. 10 0 06	8.72	9.23	9.41	7.82	7.41	6.99	7.05	6.92	7.21
rination Groundwater Amended with Nutrient∕Yeast and Electron Donor	Propionic Acid n (Col 6) ((		6.84 6.99	7.00 20 20	6.91	8.81	9.02	9.10	8.79	8.85 8 73	9.03 9.03	8.95	8.85	8.60	0.44 8 05	8.39	8.99	9.21	7.28	7.08	7.10	7.10	6.95	7.16
ed with Nut	Lactic F Acid A (Col 5) ((		6.66 6.90	6.96 7 12	7.01	9.03 0.17	9.17 8.57	8.53	7.95	7.73	8.82	8.79	8.67	8.54	0.8U 7.4	8.19 8.19			7.06				ġ.	6.89
ter Amende	Benzoic   Acid /		6.77 7.10	7.02	6.85		8.58 8.58			8.02					8.67									7.04
ination Groundwa	n-Butyric Acid (Col 3)		6.74 7.00	6.92 6.07	6.75	8.32	8.55 8.55	9.06		8.79					8.68 0 54				7.16	6.94		6.90	9	6.73
Appendix H pH Results nhance Dechlor	east iter Col 2)		6.84 7.19	7.14	7.07	9.08	9.10 9.02	9.03	8.88	8.88 9.88	9.33 9.33	9.31	9.19	9.08	9.17	9.00 8.78	9.15	9.34	7.81	7.39	7.20	7.33	7.21	7.35
n Donors to E	Unamended Vitamin/Y Groundwater Amended Effluent Groundwa (Coi 1) Effluent ((	6.97 7.44	7.05	7.15	6.99	7.28	7.29 7.10	7.38	7.16	7.15	7.46	7.38	7.39	7.38	7.30	7.20	7 28	7.26	7.12	7.44	7.24	7.54	7.15	7.22
Appendix H pH Results USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination Groung	Vitamin/Yeast L Amended G Groundwater E Influent		7.18 7.40	7.27	7.19	9.36	8.32 9.05	8.40	7.7	7.95	9.33 8.73	9.18	9.30	8.87	8.10	9.21	9.51 8.61	8.08	7.18	7.30	7.37	7.59	7.38	7.63
<b>Freatability Stu</b>	Experiment Groundwater C	6.94 7.03 7.40	7.07	7.07	7.26	7.46	7.29	7.26	7.38		7.65					7.38								
. B EM OU-1	Experiment		31 1		46 73		67				3 102						101 6							
USU/Hill AF	ete C	13-Jun-98 1-Jul-98 25-Jul-98	10-Aug-98 9-Sep-98	15-Sep-98	24-Sep-98	8-Oct-98	15-Oct-98	22-OCI-98	5-Nov-98	12-Nov-98	19-Nov-98	86-7901-02	10-Dec-98	17-Dec-98	24-Dec-98	31-Dec-98	7-Jan-99	04 -1211-99	25- 12n-00	28-190-02	4-Feh-99	12-Fah-99	17-Feh-99	24-Feb-99

Page 73

Appendix H Dissolved Oxygen Results

USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination

USU/Hill AFB EM ( Docute as nom Oc	B EM OU-1	Treatability St	USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Etimatice Decimination Documents as now O.	ron Lonois to I		Groundwat	ter Amend	ed with N	utrient/Yeas	Groundwater Amended with Nutrient/Yeast and Electron Donor	n Donor
			Vitamin/Yeast	Unamended	Vitamin/Yeast	a Dutharia	Benznin	l actic	Pronionic		
	Experiment	Unamended Experiment Groundwater		Groundwater Effluent	Groundwater	Acid		Acid Acid	Acid (Col 6)	n-Propanol Toluene (Col 7) (Col 8)	Toluene (Col 8)
	Day	Influent	Influent	(COI 1)			1	1000	10.00		
13-Jun-98		3.37	•	2.34							
25-Jul-98		1.10	~								
25-Jul-98		1.25	10	0.86							
10-Aug-98	-	-							0 02	0	0.03
10-Sep-98	32			-	0.17		0.11				0.34
15-Sep-98	37						0.32				
24-Sep-98	46						0.30				
1-Oct-98	53	3 1.24		•			0.12				
8-Oct-98	60	0 1.81					0.19			0.10	
15-Oct-98	67	7 2.05					0.17				
22-Oct-98		4 1.60		<b>~</b>							
29-Oct-98		1.53		-		0.08	0.0				
5-Nov-98				•							
12-Nov-98				<b>~</b>							
10-Nov-08	÷			-							
75-Nov-98											
2.000-008										0.16	
10-Dec-98			2 0.51	·							0.21
					3 0.33	0.26	0.22				
				Ţ							
21-Den-98			8 1.17								
7_ Ian_00				•		0.31					
	·	158 270				1 0.28	0.28	3 0.82			
14-141-55						0.27	0.30	0.34			
						0.34	0.35	5 0.32	2 0.36	6 0.34	
20-191-00						0.43	0.34	4 0.35	5 0.30	0 0.31	
20-191-22							0.20		1 0.27	7 0.41	
4-rep-vy							0.36		9 0.27	7 0.39	
	·						0.29	9 0.49	9 0.34	4 0.46	
17-rep-99		192 Z.U							9 0.52	2 0.50	0.60
24-Feb-99	·	<u> 9</u> 9 1.55			Ċ						

Page 74

Appendix H Dissolved Oxygen Results	
--	--

Groundwater Amended with Nutrient/Yeast and Electron Donor USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination Derrentane Removal Efficiencies

Percentage Removal E	moval Efficiencies	es		Groundwa	IEI AILIEIN	בח אווו ואח		Groundwater Annendeu With Indunend Teast and Erection Cond	
		Unamended	Unamended Nutrient/Yeast						
		Groundwater		n-Butvric Benzoic Lactic	Benzoic	Lactic	Propionic		
									Talinoon
	Experiment	Effluent	Groundwater Acid		Acid	Acid	Acia	n-Proparioi I oluelle	
	,	(Col 1)	Effluent (Col 2) (Col 3) (Col 4) (Col 5) (Col 6)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7) (Col 8)	(Col 8)
רמוט									

23.33	43.33	56.86	42.42	54.84	74.36	88.42	42.31	78.41	81.51	92.66	81.25	82.72	58.82	78.72	87.40	58.12	70.37	77.41	78.47	45.90	11.11	16.13	51.92	14.29	-33.33	56.07	
63.33	51.67	43.14	51.52	51.61	76.92	84.21	57.69	80.68	89.08	92.66	81.25	80.25	54.90	79.79	87.40	74.36	74.07	69.26	84.72	44.26	13.89	-32.26	25.00	17.86	-11.11	57.16	
23.33	30.00	5.88	51.52	48.39	71.79	91.58	53.85	78.41	86.55	92.66	78.75	81.48	54.90	77.66	86.61	51.28	73.15	82.59	79.86	40.98	16.67	12.90	48.08	39.29	-15.56	55.48	
20.00	33.33	3.92	60.61	51.61	64.10	87.37	53.85	78.41	88.24	92.09	78.75	79.01	52.94	76.60	85.83	84.62	75.00	69.63	76.39	47.54	2.78	-32.26	44.23	12.50	-8.89	53.01	
63.33	46.67	29.41	63.64	38.71	56.41	87.37	65.38	78.41	84.03	90.96	76.25	79.01	54.90	76.60	85.04	47.01	74.07	89.63	79.17	42.62	5.56	35.48	30.77	48.21	-2.22	58.71	age 75
26.67	38.33	11.76	60.61	22.58	71.79	84.21	69.23	75.00	85.71	89.83	72.50	74.07	50.98	72.34	84.25	82.05	71.30	89.63	81.25	44.26	-19.44	-6.45	53.85	33.93	-8.89	54.28	
43.33	30.00	29.41	66.67	12.90	48.72	80.00	26.92	71.59	82.35	86.44	71.25	67.90	33.33	64.89	81.10	76.92	70.37	80.00	77.78	45.90	13.89	-9.68	42.31	26.79	-24.44	49.87	
-31.65	-2.17	39.10	-16.13	27.62	38.05	15.00	14.38	31.05	-10.08	3.50	33.33	30.68	35.64	-10.45	45.69	35.29	18.52	48.89	51.09	37.72	-11.59	22.81	42.93	34.33	1.29	20.19	
- ç	37	46	53	60	67	74	81	88	95	102	108	116	123	130	137	44	151	158	165	169	172	179	187	192	199	Ave Removal	
10-Aug-98	15_Sen-98	24-Sen-98	1-Oct-98	8-Oct-98	15-Oct-98	22-Oct-98	29-Oct-98	5-Nov-98	12-Nov-98	19-Nov-98	25-Nov-98	3-Dec-98	10-Dec-98	17-Dec-98	24-Dec-98	31-Dec-98	7lan-99	14-Jan-99	21-Jan-99	25-Jan-99	28-Jan-99	4-Feb-99	12-Feb-99	17-Feb-99	24-Feh-99	· · · · ·	]

Imamended Immended         Vitamin/Yeast         Inamended Immended         Vitamin/Yeast         Inamended Immended         Propionic Immended         Propionic Acid         Propionicid Acid	Results as mg/L of CaCO <sub>3</sub> Ground	Results as mg/L of CaCU <sub>3</sub>	<b>(</b> )									
50         51         515         1081         566         1040         1071         1040         1081         1030           7         515         970         495         1030         1010         990         1081         1030         1010         990         1081         1030         1010         990         1030         1010         990         1030         1010         990         1030         1040         1031         1040         1031         1030         1010         990         1030         1040         1031         1030         1010         990         1040         1031         1030         1010         990         1040         1031         1030         1010         990         1040         1031         1030         1010         990         1040         1031         1030         1010         990         940	<u>⊢                                     </u>	Experiment	Unamended Groundwater Influent	Vitamin/Yeast Amended Groundwater Influent	Unamended Groundwater Effluent (Col 1)	Vitamin/Yeast Amended Groundwater Effluent (Col 2)	n-Butyric Acid (Col 3)	Benzoic Acid (Col 4)	Lactic Acid (Col 5)	Propionic Acid (Col 6)	n-Propanol (Col 7)	Toluene (Col 8)
1         23         515         1081         566         1040         1071         1081         566           37         515         970         495         1030         1071         1040         1071         1030           37         515         970         495         1030         1010         990         1030         1031           53         404         980         444         980         939         990         930         990         930         990         930         990         930         940         851         841         1037         920         940         851         840         1031         1031         930         1040         1071         1040         1071         1040         1071         1040         1071         1040         1031         930												
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	စ္ဆ		62		64							
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	ø	~									0101	10.4
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	œ	32				1040	1071	1040	• •		10401 10401	
46 $485$ $980$ $444$ $980$ $343$ $960$ $339$ $960$ $339$ $930$ <th< td=""><td>œ</td><td>37</td><td></td><td></td><td>-</td><td>1030</td><td>0101</td><td></td><td>-</td><td>-</td><td>-</td><td>55</td></th<>	œ	37			-	1030	0101		-	-	-	55
53         404         980         424         919         949         0.20         970 <td>ĝ</td> <td>46</td> <td>-</td> <td></td> <td>-</td> <td>980</td> <td>808 070</td> <td></td> <td></td> <td></td> <td>T</td> <td>S O</td>	ĝ	46	-		-	980	808 070				T	S O
67 $411$ $1380$ $401$ $386$ $431$ $1037$ $920$ $910$ $912$ $108$ $74$ $470$ $988$ $430$ $1116$ $1096$ $1008$ $100$ $972$ $1008$ $1007$ $1007$ $1007$ $1007$ $1007$ $1007$ $1007$ $1007$ $1007$ $1007$ $100$	80	2 2 2			-	8 8 0 10	040 FY8	0701 803				60
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	ĝ	ی م			-	949 1037	026	910 910		·		626
110 $110$ $110$ $110$ $110$ $110$ $100$ $110$ $110$ $88$ $421$ $587$ $401$ $1184$ $1057$ $988$ $1076$ $1096$ $1008$ $1008$ $1116$ $95$ $352$ $548$ $362$ $930$ $822$ $773$ $851$ $881$ $108$ $372$ $675$ $372$ $675$ $372$ $1067$ $988$ $1076$ $1096$ $1008$ $1076$ $1096$ $1007$ $1096$ $1007$ $1096$ $1007$ $1096$ $1007$ $1096$ $1007$ $1096$ $1007$ $1096$ $1007$	p g	õř				-	1008	910		•		
88         421         587         401         1184         1057         988         1076         1096           95         352         548         362         930         822         773         851         881           102         387         675         372         1067         959         872         873         881           108         372         675         372         1067         959         842         969         959           116         397         915         1076         1067         966         1017         1007           123         417         1129         397         1047         1007         966         1017         1007           130         376         814         397         1068         976         915         960         959           133         427         1129         386         1271         1077         986         1017         1007           151         427         1119         1271         1078         976         915         1007           151         427         1018         966         945         1017         1007           1	o a	- <del>6</del>					•	1008				
95         352         548         362         930         822         773         851         881           102         382         998         577         891         812         812         832         833           108         372         675         372         1067         959         842         969         959           116         397         925         509         1078         997         915         1007         1007           123         417         1129         397         1067         959         842         969         959         950         950         950         950         950         956         1007	ç œ	5 8						986				
	0.00	, G										
108         372         675         372         1067         959         842         969         959           116         397         925         509         1078         997         915         1007         1007           123         417         1129         397         1047         1007         966         1017         1007           130         376         814         397         1047         1007         966         1017         1007           137         427         610         386         1271         1078         976         915         986         1007         1007           151         427         610         386         1271         1078         956         1119         1159           144         427         1129         427         1088         1027         997         1067           158         407         814         390         1268         1305         1203         1223         1322           166         427         1078         366         945         1017         1007           167         427         1078         366         976         913         3222	2 2	102										
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	x œ	100										
$ \begin{array}{cccccccccccccccccccccccccccccccccccc$	8 8	116										
130       376       814       397       1068       976       915       986       1007         137       427       610       386       1271       1078       956       1119       1159         144       427       610       386       1271       1078       956       1119       1159         151       437       1078       397       1068       1027       997       1068       1007         151       437       1078       397       1058       1108       1027       997       1068       1017         165       427       1078       397       1058       1108       1027       997       1068       1017         169       393       953       383       1058       1305       1200       1119       1251         169       393       953       383       962       972       913       953       962         172       422       641       417       1363       1200       1119       1210       1251         169       363       953       982       972       913       953       962         1779       354       933       8	8	12:										
137       427       610       386       1271       1078       956       1119       1159         144       427       1129       427       1088       1027       997       1058       108         151       437       1078       397       1058       1108       1027       997       1058       108         151       437       1078       397       1058       1108       1027       997       1058       1017         158       407       814       390       1288       1305       1203       1322       1322         165       427       641       417       1363       1200       1119       1210       1251         169       393       953       383       962       972       913       953       962         172       422       894       383       982       972       913       953       962         172       353       913       383       984       835       884       903         179       354       923       383       894       874       835       894       903         179       354       923       384	80	13(										
144       427       1129       427       1088       1027       997       1058       1088         151       437       1078       397       1058       1108       966       945       1017         158       407       814       390       1288       1305       1203       1322       1322         165       427       641       417       1363       1200       1119       1210       1251         165       393       953       383       962       972       913       352       1322       1322         172       422       894       383       962       972       913       953       962         172       353       913       383       982       874       835       884       903         172       354       923       383       894       874       835       884       903         187       354       923       383       894       874       835       884       903         187       353       884       471       803       874       835       894       903         192       383       874       355 <td< td=""><td>80</td><td>137</td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td><td></td></td<>	80	137										
151       437       1078       397       1058       1108       966       945       1017         158       407       814       390       1288       1305       1203       1322       1322         165       427       641       417       1363       1206       1203       1322       1322         169       393       953       383       962       972       913       953       962         172       422       894       383       963       884       835       884       903         172       422       894       383       963       874       835       884       903         179       363       913       383       894       874       835       894       903         187       354       923       383       884       471       803       874       825       884       903         192       383       884       471       805       845       903       903         199       383       884       471       805       845       903       903         199       383       884       471       805       845 <td>86</td> <td>41</td> <td></td>	86	41										
158       407       814       390       1288       1305       1203       1322       1322         165       427       641       417       1363       1200       1119       1210       1251         169       393       953       383       962       972       913       953       962         172       422       894       383       962       972       913       953       962         172       422       894       383       962       972       913       953       962         179       363       913       383       962       972       913       953       962         187       354       923       384       874       835       894       903         187       354       923       344       903       874       835       894       903         187       353       884       471       805       845       903       845       903         192       383       884       471       805       845       903       845       903         199       383       884       471       805       845       903	66	15,										
165         427         641         417         1363         1200         1119         1210         1251           169         393         953         953         383         962         972         913         953         962           172         422         894         383         962         972         913         953         962           172         422         894         383         962         972         913         953         962           179         363         913         383         984         835         884         903           187         354         923         344         903         874         835         894         894           192         363         874         354         903         874         825         854         903           192         383         884         471         805         845         845         845           199         383         884         471         805         845         864	66	156						-	•			
169         393         953         383         962         972         913         953         962           172         422         894         383         963         933         884         903           172         422         894         383         933         884         835         884         903           179         363         913         383         933         894         874         835         894         894           187         354         923         344         903         874         825         854         903           192         363         874         354         903         874         825         845         903           192         383         884         471         805         845         845         845         845           199         383         884         471         805         845         845         864	66	16							•	~		
172         422         894         383         933         884         835         884         903           179         363         913         383         933         884         835         884         903           187         354         923         383         894         874         835         894         894           192         363         874         355         894         766         786         845         903           192         363         874         354         903         874         825         845         903           192         363         874         354         894         766         786         845         845           199         383         884         471         805         845         855         864	66	16										
179         363         913         383         894         874         835         894         894           187         354         923         344         903         874         825         854         903           187         353         874         903         874         825         854         903           192         363         874         354         903         874         825         845         903           192         363         874         354         894         766         786         845         845           199         383         884         471         805         845         825         864	6	17:										
187         354         923         344         903         874         825         854         903           192         363         874         354         903         874         825         854         903           192         363         874         354         894         766         786         845         845           199         383         884         471         805         845         825         835         864	66	17										87
192         363         874         354         894         766         786         845         845           199         383         884         471         805         845         825         835         864	66	18										80
199 383 884 471 805 845 825 835 864	6	19.				_						815
	6	10	_		•							

Appendix H Alkalinity Results

Page 76

Appendix H cis 1,2-Dichloroethene Results

USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination

USU/HIII AF		I reatability ou					Shore A read	od with Mu	triant/Vaset	Correction Amondod with Nutrient/Veast and Electron Donor	Donor
Results in ug/L	l/br					Groundwar					
			Vitamin/Yeast	Unamended	Vitamin/Yeast	n-Butvric	Benzoic	Lactic	Propionic		
	+		Amenueu Groundwater	Groundwaren	Groundwater			Acid	Acid	n-Propanol	Toluene
Date	Dav	Influent	Influent	(Col 1)	Effluent (Col 2) (Col 3)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
10-Aug-98											
0 Cen-08	31	2736.22	2363.56	2424.12	2260.68	2203.72	2164.96	2189.84	2124.44	2220.68	2109.48
a-deb-o		50 08		66.00	65.45	63.02	59.34	64.67	61.27	56.96	60.09
00-040-00				37.16	45.49	43.81	41.00	41.28	46.09	44.15	40.89
20-001-98					25.21	26.07	23.44	21.65	21.37	24.02	26.42
10-Nov-98			92.44		10.70	10.26	9.78	21.37	9.54	11.14	9.62
2-Dec-98	8 115					04.00	70.05	04 30	70 56	85.39	89.03
23-Dec-98	<b>v</b> -	150.93				80.10	0.00				00 101
20-Dec-08	Ţ	414.98			117.52	110.90	105.69	115.52	130.70		144.00
	•			228.93	190.59	204.31	169.11	208.43	173.34		211.78
	_					362.40		354.86	337.72	277.99	401.79
14-Jan-99						115 88	106.28				98.04
21-Jan-99	9 165										153 50
27- Jan-99	9 171	132.20		134.12	166.81	172.33		1/0.00			
	•			141.89	180.67	180.24	162.26	174.45			
00-LEU-90	·	-	018 OB			207.61	172.33	211.69		192.78	187.19
10-Feb-99	•						20 122	000			
17-Feb-99	9 192	2 486.31		419.39			00.1./0				
24-Feb-99		9 393.79	556.66	364.99	533.35	521.11	489.02	502.05			

	Results
Appendix H	cis 1,2-Dichloroethene

USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination

<b>USU/Hill AFB</b>	EM OU-1 1	Freatability Stu	USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Decilorination	Donors to	) Enhance L	Jechiorinari		i	ſ	acitalization	Field Prodiction from Column Ope
Dercentade Removal Efficiencies	emoval Effi	ciencies		Groundwa	ter Amende	d with Nutri	ient/Yeast a	Groundwater Amended with Nutrient/Yeast and Electron Donor	Lonor		
		here	Vitamin/Veast						<u></u>	Change in	
			1000	n-Butyric	Benzoic	Lactic	Propionic			Concentration,	Concentration, A Concentration/
<u>u</u>	Evnariment		ater		Acid	Acid	Acid	n-Propanol	Toluene	Col 1(Inf-Eff):	Distance I hrough
			5	<u>(</u>	4)	(Col 5)	(Col 6)		(Col 8)	(ng/L)	Soll: (ug/L/cm)
00 00	21	11 41	35	6.76	8.40	7.35	10.12	6.05	8.21	312.10	
4-0ep-40	- C		· c	5 74	11 25	3.28	8.36	14.81	10.13	-6.02	•
30-Sep-98	70	-10.04	i ç	10.1	10 10	18 56	9.07	12.90	19.33	4.99	
20-Oct-98	72		<u>.</u>	10.01	13.14	10.00	16.20	5.80	-3.61	4.48	0.11
10-Nov-98	63 03	15.04	-	-2.24	0.0			0000 E 27	10.00	15 73	0.37
2-Dec-98	115	61.69	σ	12.76	16.84	•	10.00	0000	04.01	14 DE	
23 Dar-08	136	29.78	22.50	21.86	34.49	20.77	33.22	28.33	17.07		
20-00-00	641		14	19.71	23.49	16.37	0.99	9.63	9.63	138.92	
30-DeC-90		_	25	19.95	33.74		32.09	18.99	17.03	-8.84	•
6-Jan-99			0 0	161 70	123.05	7	-134 64	-93.14	-179.16	122.38	2.88
14-Jan-99	158		ę : P	1			01 10	21 21	27 BG	173.80	4.08
21-Jan-99	165	51.58	4	-	Z1.6U	V	01.12		10.10	1 02	
27. Ian-00	171	-1.45	3.05	-0.16	0.02		37.76	1.12	10.78	-1.32	
	1 10		C		10.14	3.39	2.92	7.22	12.50	-0.62	
3-Feb-49							10.59	11.96	14.52	-45.34	•
10-Feb-99	185	•	<b>?</b> ;				2 47	14 80		66.92	1.57
17-Feb-99	192	2 13.76	69					00.11		28.80	0.68
24-Feh-99	199	9 7.31	4.19	6.39		9.81	_ L	20.0	11.0		101 20 2120 1
	Rande	-10/62%		-68/69% -152/22%		-123/34% -147/22%	-134/38%	-93/28%	-93/28%-11/9/25%	капде	
	Shan	72.00	137.00	174.00	157.00	169.00	172.00	121.00	20		
	Chair		1		7 44	-5 29	5.08	4.57	09.0	Ave	9 1.33
	Ave	17.19	t t. /								
Dence Mithout Day 158	uit Dav 158	-10/62%	0/69%	-2/22%	0/34%	-81/22%	1/38%	1/28%	-4/25%		
Ralige villin	Jut Lay 134				34.00	103.00	37.00	27.00	29.00		
	opail		10.00				15.06	11.55	13.44		
Ave Without Day 158	Day 158	14.86									

Page 78

## Appendix H cis 1,2-Dichloroethene Results

ded with Nutricot No USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination -

Percentade Removal Rate (%/dav)	Dercentage Removal Rate (%/dav)	ie (%/dav)		Groundwa	ter Amende	d with Nut	rient/Yeast	Groundwater Amended with Nutrient/Yeast and Electron Donor	Donor
		led	Vitamin/Yeast						
		Groundwater	Amended	n-Butyric	Benzoic	Lactic	Propionic		
	Experiment		Groundwater	Acid	Acid	Acid	Acid	n-Propanol	Toluene
Date	Day	(Col 1)	Effluent (Col 2) (Col 3)		(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
9-Sep-98	31						•		
30-Sep-98	52	0.03	0.15	0.30	0.47	0.25	0.44	0.50	0.44
20-Oct-98	72	0.05	0.31	0.48	0.76	0.55	0.44	0.69	0.74
10-Nov-98	93		0.27	0.27	0.65	0.80	0.60	0.45	0.37
2-Dec-98	115		0.23		0.57	-1.51	0.80	0.25	0.33
23-Dec-98	136		0.75		1.22	-1.45	1.24	0.80	1.04
30-Dec-98	143				4.14	2.65	2.44		2.49
6- lan-99	150	2.10		2.83	4.09	2.48	2.36	2.04	1.90
14lan-99	158		•		-5.58	•	-	-4.63	
21-Jan-99					-7.23		-8.08	-5.14	-10.81
27-Jan-99	·				1.82	1.92	4.94	1.86	3.22
3-Feh-99		1	-		0.73	0.29	2.91	0.60	1.66
10-Feh-99	•		1		2.25		0.96	-	1.93
17-Feh-99					2.51	0.91	1.00		1.90
24-Feh-99			5.25	0.70	1.85	1.38	0.65		1.30
	Range	-0.1/7	Ĺ	-3.9/5.3% -9.8/3.0%		-8.9/2.7%	-7.2/4.1% -8.9/2.7% -8.1/4.9%		-5.1/2.7% -10.8/3.2%
	Span	7.38	9.11	12.76	11.37	11.52	13.02		14.03
	Ave	1.87		9 -0.51	0.59	-0.58	0.31	0.34	-0.26
Ave Without Day 158	t Day 158	1.79	1.17	0.08	1.06	-0.01	0.82	0.72	0.50

Page 79

Appendix H Vinyl Chloride Results USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination Commuter Amended with Nitrient/Yeast and Flectron Donor

Results in ug/L	g/L					Groundwa	Iter Amend	ed with NU	Itrienv y easi		
			Vitamin/Yeast	Unamended	Vitamin/Yeast	Ċ		cito	Dincincia		
		Unamended Amended	Amended	Groundwater	Amended	n-Burync	Denzoic	Arid		n-Pronanol	Toluene
	Experiment	Experiment Groundwater			Groundwater					_	(Col 8)
Date	Day	Influent	Influent	(1. 10)		6 00		5 5 5 5			
10-Aua-98	-										
0_Cen_08	31	295.37	244.12	270.43			•••				
20 COD 08				68.66							
		5	46.00	36.81	53.93	65.79	53.27	53.45	60.36	58.98	50.90
DU-Nov-98											
2-Dec-98	•										
23-Der-98	•										
o-Jan-44											
14-Jan-99											
21-Jan-99											
27. lan-00											
2 Eah 00	178										
10-Feb-99											
17-Feb-99											
24-Fah-99											
	<u>}</u>										

Appendix H Vinyì Chloride Results

USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination Zero Value Listed instead of N/D

Results in ua/L	a/L					Groundwa	ter Amende	ed with Nu	trientYeast	Groundwater Amended with Nutrient/Yeast and Electron Donor	n Donor
	Experiment	Vitamin/V Unamended Amended Experiment Groundwater Groundwa	Vitamin/Yeast Amended Groundwater	Unamended Groundwater Effluent	Vitamin/Yeast Amended Groundwater	n-Butyric Acid	Benzoic Acid		Propionic Acid	n-Propanol	Toluene
Date	Day	Influent	Influent	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
10-Aug-98	1										
9-Sep-98	31	295.37	244.12	270.43	262.23		•••	•••		•••	2/9.99
30-Sep-98	52	32.82	33.56	68.66	65.57					45.81	64.63
20-00-98				36.81							50.90
10-Nov-98				0.00						<b>~</b>	
2-De0-98	-			00.00							
23-Dec-98	136			0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
30-Dec-98	-		0.00								
6-Jan-99			_								
14lan-99											
21-Jan-99											
27-Jan-99											
3-Feh-99											
10-Feh-99	185										
17-Feb-99	•										
24-Feb-99	•	0.00		25.56							

## Appendix H Vinyl Chloride Results

USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination

%Apparent Removal Efficiencies Groundwater Amended with Nutrient/Yeast and Electron Donor

				Groundwa	ater Amen			Groundwater Amended Will INULIERIN FEAST and Electron Point	
		Unamended		n Buthuric Benzoic		l artic	Pronionic		
		Groundwater	Groundwater	Acid		Acid	Acid	n-Propanol	Toluene
Date	Experiment Dav	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	<u></u>	<u>ບ</u>
O Can OR		31 8.44	-7.42	-96.96 -	-10.78	-5.07	-8.75	-10.99	•
a-dec-a		1	-95.38	1		-89.48	-61.56	-36.50	-92.58
50-0ep-ao			-17 24		-15.80	-16.20	-31.22	-28.22	-10.65
20-OCI-98					00.0	00.00	00.0		0.00
06-70N-01	T		100.001	•1	-9.54	-96.54	4.17	-8.46	-8.70
2-Dec-90					0.00	00.0	0.00	0.00	0.00
23-DeC-98					0.00	00.0	0.00	0.00	0.00
30-Dec-90				00.0	0.00	00.00	0.00	0.0	0.00
					0.00	0.00			
14-Jan-99	•		00.0			0.00			
21-Jan-99	-	0.00		00.0	0.00	00.0	0.00	0.00	0.00
2/-Jan-99						0.0			
3-Feb-99	-					0.00		0.00	
10-rep-88						000			
17-Feb-99			_						
24-Feb-99		199 0.00	0.00			- 1			ľ
-	Range	-109/8%		-95/100% -95100%	-72/0%	%0/26-	-62/0%	-37/0%	-93/0%
		52 <del>9</del> -					-7.05	-5.61	-8.44
Ave Accim	Avia Accilimitation (Eirst 5 davs)	-21.84	-2.52	2 -31.97	-19.64	40.44	-19.39	-14.64	
AVE JULIU									

Ave Accumulation (First 5 days)

Appendix H Ethene Results

Results as mg/L	ng/L	•				Groundwa	ter Ameno	led with N	utrient/Yeas	Groundwater Amended with Nutrient/Yeast and Electron Donor	on Donor	
			Vitamin/Yeast	Unamended	Vitamin/Yeast							
		Unamended Amended	Amended	Groundwater	Amended	n-Butyric	Benzoic	Lactic	Propionic			
	Experiment	Experiment  Groundwater	Groundwater	Effluent	Groundwater	Acid	Acid	Acid	Acid	n-Propanol	Toluene	Detection
Date	Day	Influent	Influent	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)	Limit
10-Aug-98	1											
9-Sep-98	31	0.010					0.011					
30-Sep-98	52	0.010					0.013					
20-Oct-98	72	0.018					0.029					
10-Nov-98	93	0.0096	Ŭ	U	U	U	0.0096	Ŭ	Ű	Ŭ	-	0
2-Dec-98	115						0.010					
23-Dec-98	-						0.017					
30-Dec-98	-						0.031					
6-Jan-99	150	0.010	0.024	0.010	0.017	0.019	0.016	0.020	0.018	0.012	0.019	0.01
14-Jan-99	158						0.016					
21-Jan-99	-						0.010					
27-Jan-99	-						0.020					
3-Feb-99	178						0.014					
10-Feb-99	185						0.010					
17-Feb-99	192						0.010					
24-Feb-99	199						0.010					

### Appendix H Ethene Results

Percentage Rem	Removal Effi	oval Efficiencies	)	Groundwa	ter Amend	ed with Nu	trient/Yeas	Groundwater Amended with Nutrient/Yeast and Electron Donol	n Donor
2		Unamended	Vitamin/Yeast						
		Groundwater	Amended	n-Butyric	Benzoic II	Lactic	Propionic		
	Experiment	Effluent	Groundwater	Acid	Acid /	Acid	Acid	n-Propanol Toluene	Toluene
Date	Day	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
10-Aug-98	F								1
9-Sep-98	31	10.00	27.27	0.00	0.00	0.00	18.18		-9.09
30-Sep-98	52	0.00	-63.64	-72.73	-18.18	-18.18	-9.09	-27.27	-18.18
20-Oct-98	72	0.00	-69.57	-26.09	-26.09	13.04	56.52	8.70	56.52
10-Nov-98	93	0.00	-35.42	0.00	0.00	0.00	0.00	0.00	-4.17
2-Dec-98	<b>~</b>		0.00	50.00	50.00	45.00	50.00	25.00	25.00
23-Dec-98	-		58.33		29.17	-4.17	0.00	33,33	41.67
30-Dec-98	<b>~</b> -		-20.00	-12.50	22.50	30.00	17.50		15.00
6-Jan-99	Ţ		29.17	20.83	33.33	16.67	25.00		20.83
14-Jan-99	-		77.78	82.22	64.44	60.00	73.33		80.00
21-Jan-99	<b>T</b>	ص 		54.55	54.55	50.00	59.09	50.00	50.00
27-Jan-99	Ţ		-43.75	•	-25.00	12.50	12.50	6.25	-31.25
3-Feb-99	-			21.43	0.00	7.14	28.57	0.00	-50.00
10-Feb-99	185	0.00	0.00		0.00	0.00	0.00	0.00	00.00
17-Feb-99				00.0	0.00	0.00	0.00		00.00
24-Feb-99			0.00	0.00	0.00	0.00	0.00	0.00	00.00
	Range	0/33%	-70/78%	-73/82%	-26/64%	-18/60%	-9/73%	-27/78%	-50/80%
	AveRemova	1 2.89	0.03	5.76	12.31	14.13	22.11	18.68	11.76

### Appendix H Ethane Results

				0.004	10.0	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0000	0.008	0.008	0.008	
-	Dete			<b>.</b> .	~	~	~	~	~	~	~	~	~	~	~		n	ŝ	ŝ	,
n Donor	Toluene	COI 8)			0.010	0.008	0.008	0.008	0.008	0.00	0.00	0.00	0.008	0.00	0.00		0.00	0.00	00.00	
Groundwater Amended with Nutrient/Yeast and Electron Donor	anol	(Col /) [(		0.004	0.010	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008		0.008	0.008	0.008	
utrient/Yeast	Propionic	(Col 6)				_							0.008							
ed with Nu	Lactic Acid	(Col 5)											0.008							
ter Amend	Benzoic Acid	(Col 4)		0.004	0.010	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0 008		0.000	0.008	0.008	0,008	>>>>>
Broundwat	ic.	Col 3)		0.004	0.010	0.008	0.008	0.008	0.008	0.008	0.008	0 008	0.008	0.008		0000	0.008	0.008	000	0.000
ionors to Ennance Decinonnauor Groun	east ater	Effluent (Col 2) (Col 3)		0.004	0.010	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008	0.008		0,000	0.008	0.008		0.000
	Unamended Vitamin/V Groundwater Amended Effluent Groundwa	(Col 1) E		0.004	0.010	0.008	0.008	0.008	0.008	0,008	0008	0.000	0.000	0.008		0.000	0.008	0.008		0,000
JSU/Hill AFB EM OU-1 Treatability Study Using Electron L Results as mo/l	++	Influent (		0.004	0.010	0.008	0.008	0 008	0.008	0.008	0.008	0.000	0000		0.000	0.008	0.008	0.008		0,000
eatability Stud	Vitamin/Yeas Unamended Amended Experiment Groundwater	Influent Ir		0.004	0.010	0 008	0.008	0.008	0.008	0000	0.000	0,000	0,000	0.000	0.000	0.008	0.008	0.008		0.008
3 EM OU-1 Tr Id/l	Experiment (	Day	-	31	52	C1	1 6	115	136	00-7 770	- <del>1</del> 0	0.01	1001			178	185	191	101	199
USU/Hill AFB Ef Results as m0/l		Date	10-Aug-98	9-Sep-98	30-Sen-98	20-100-00	10-Nov-08		22 Doo 00	20-00-00	00-DeC-90		14-Jan-99	21-Jan-99	27-Jan-99	3-Feb-99	10-Feb-99	17.Fah.00		24-Feb-99

### Appendix H Ethane Results

USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination Percentage Removal Efficiencies Groundwater Amended with Nutrient/Yeast and Electron Donor

Remova	٣	Percentage Removal Efficiencies		Groundwa	Iter Amend	ed with N	utrient/Yea	Groundwater Amended with Nutrient/Yeast and Electron Donor	
Unamended	Unamen	ded	Vitamin/Yeast						
Groundwater	Groundw	ater	Amended	n-Butyric	Benzoic	Lactic	Propionic		
Experiment Effluent	_		Groundwater	Acid	Acid	Acid	Acid	n-Propanol	·
Day (Col 1)	(Col 1)		Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
ļ									
31 0.	ō	8			Ū	00.00	Ŧ		-
		8			-	0.00	-		-
72		8			-	0.00	-	-	-
		8				0.00			-
115		0			-	0.00	-		
		0				0.00			
143		0				0.00			-
150		0			-	0.00	_		
		0				0.00			
165		0	0.00	0.00	0.00	0.00	0.00	00.0	0.00
171		2				0.00			
		8			_	0.00			
185		8				0.00			
192		8				0.00			
199		2				0.00			

Appendix H Methane Results

		:	ction		004		0.004	0.004	0.004	0.004	0 004	100.0	0.004	0.004	0.004		0.004	0.004	0.004		0.004	0.004	0.004	
	Г		Detection					_	_															
n Donor				(0 10)	0 879		0.393	0.03	0.34	0.17	0 10		12.0	0.18	0.23		0.11	0.42	0.43		40.0	0.12	0 16	>>
and Electro			anol		0200	0.5.0	0.528	0.380	0.125	0.255	1100		0.559	0.170	0 221	- 44.0	0.115	0.275	0.271		10.0	0.161	0 248	2112
nation Groundwater Amended with Nutrient/Yeast and Electron Donor		ionic		(Col 0) 1((		0.410	0.333	0.223	0.100	0.181	105	0.400	0.510	0.303	0 234		0.103	0.228	0 194		0.035	0.079	0 225	01410
od with Nutr		actic P		Col 5)		0.588	0.411	0.244	0.152	0.142		0.420	0.479	0.361	0.423	001.0	0.132	0.243	0 282	202.0	0.118	0.287	0 2 5 0	0.000
er Amende		Benzoic L		(Col 4)		0.585	0.546	0.424	0.217	0 116		1.42.0	0.559	0.297	0000	0.028	0.078	0.347	0000	0.730	0.115	0.132	0110	U. 140
lation Eroundwet		n-Butyric		(Col 3) (		0.597	0.523	0.272	0.209	0.038	0.000	0.312	0.688	0.256		1.62.0	0.074	0.406	1100	4.1.0	0.037	0.062	0000	0.403
Donors to Enhance Dechlorination Group		east	ater	Effluent (Col 2) (		0.412	0.576	0.398	0.333	0.326	0.040	0.136	0.881	0.268	0100	0.2.0	0.098	0 409	277 C	0.01/	0.036	0.107		0.183
		Unamended Vitamin/Y Groundwater Amended	fluent	Col 1) E		0.470	0.211	0.048	0 169	0.030	0.038	0.146	0.059	0.038		0.170	0.020	0.173		0.094	0.011	0 011		0.019
Using Electro		east		Influent (		0.349	0.226	0 453	0.075		0.432	0.560	0 768	0.557	100.0	1.376	0.343	0 317	10.0	0.282	0.019	0.021	10.0	0.019
USU/Hill AFB EM OU-1 Treatability Study Using Electron		Vitamin/V	Experiment Groundwater Groundwater	Influent Infl		0.594	0.056	0311	0.055		0.18/	0.284	0 025		0.123	0.175	0.229		0.104	0.033	0.004			0.016
EM OU-1 Tres	۲		xperiment Gro	Day Infl	F	31	52	1 C 7	4 6	0 J	115	136	2113	7 C	150	158	165	1 0		178	185	2007	132	199
USU/Hill AFB	Results as mg/L		<u></u>	Date	10-Aug-98	9-Sen-98	20-Sen-08			28-70N-0L	2-Dec-98	23-Dec-98		20-DEC-80	6-Jan-99	14-Jan-99	01-lan_00	2	27-Jan-99	3-Feb-99	10 Cab 00		1/-FCD-49	24-Feh-99

### Appendix H Methane Results

Dercentage	USU/MIII AFD EIVI UU-L L Percentare Removal Effic	fficiencies	ficiencies Study Company Erection Solution Street	Groundwate	r Amended	Groundwater Amended with Nutrient/Yeast and Electron Donor	Meast and E	Electron Done	or
		Unamended	Unamended Vitamin/Yeast						
		Groundwater Amended	Amended	n-Butyric	Benzoic	_	ō		
	Experiment	Effluent	Groundwater	Acid	Acid	Lactic Acid //	Acid (Col	anol	Toluene
Date	Day	(Col 1)	Effluent (Col 2) (Col 3)	(Col 3)	(Col 4)	(Col 5) (6	6)	(Col 7)	(Col 8)
10-Aug-98	1								
9-Sep-98	31	20.875	-18.052	-71.060	-67.622	-68.481	-18.911	-6.017	-94.269
30-Sen-98	52	? -	-154.867	-131.416	-141.593	-81.858	-47.345	-133.628	-73.894
20-Oct-98			12.141	39.956	6.402	46.137	50.773	16.115	91.391
10-Nov-98	. 0	ې 	-344.000	-178.667	-189.333	-102.667	-33.333	-66.667	-364.000
2-Dec-98				91.204	73.148	67.130	58.102	40.972	60.185
23-Dec-98	13		75.714	44.286	48.036	11.964	27.679	56.964	77.143
30-Dec-98	4	- -	-14.714	10.417	27.214	37.630	33.594	27.214	72.396
6lan-99	15			54.039	46.679	35.189	45.601	69.479	66.427
14-Jan-99	10			81.759	76.090	68.532	82.994	83.939	82.631
21-Jan-99	16	0	71.429	78.426	77.259	61.516	69.971	66.472	66.472
27-Jan-99	17	-66.346	-29.022	-28.076	-9.464	23.344	28.076	13.249	-32.492
3-Feb-99	178	-184.848	-12.411	24.113	-2.837	0.000	31.206	3.901	-55.319
10-Feb-99	18	-175.000	-89.474	-94.737	-505.263	-521.053	-84.211	-168.421	-126.316
17-Feb-99	19	0.000	-409.524	-195.238	-528.571	-1266.667	-276.190	-666.667	-480.952
24-Feb-99	19	-18.750	-863.158	-1284.211	-678.947	-1789.474	-1084.211	-1205.263	-768.421
	Range	-277/91%	-863/85%	-1284/91%	-679/77%	-1789/69%	-1084/83%	-1205/84%	-768/91%
	AveRemoval	44.573	-107.652	-103.947	-117.920	-231.917	-74.414	-124.557	-98.601

	Results
Appendix H	1,1,1-Trichloroethane

4 USU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Dechlorination

USU/HII AFB		USU/HIII AFB EIVI UU-1 Treatability Study USII ig Docutio in 10/1	_			Groundwa	tter Amend	ed with Nu	trient/Yeast	Groundwater Amended with Nutrient/Yeast and Electron Donor	n Donor	
			Vitamin/Yeast	Unamended	Vitamin/Yeast							
		Unamended Amende	Amended	Groundwater	Amended	n-Butyric	Benzoic	Lactic	Propionic			
	Experiment	Experiment Groundwater Groundv	Groundwater	Effluent	Groundwater	Acid	Acid	Acid	Acid	n-Propanol	Toluene	
Date	Day	Influent	Influent	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)	
10-Aug-98	-											
9-Sep-98	31	160.77		135.46			Q/N					
30-Sep-98	52	16.67	58.49	19.45			31.11			~		
20-Oct-98			-	10.51			D/N					
10-Nov-98	- 86 - 86	11.22		12.05			QN					
2-Dec-98	<b>v</b>		6.90	3.72	0/N	QN	QN	1.70	QN	0/N	Q/X	
23-Dec-98	136		4.38	2.40			0/N					
30-Dec-98	``			2.57			<b>D/D</b>					
6-Jan-99	150			7.88			4.04					
14-Jan-99	•	3.55		1.49			QN					
21-Jan-99				D/N			QN					
27-Jan-99	-			1.80			ava					
3-Feb-99	•	2.04		1.21			QN					
10-Feb-99	185			<b>U/N</b>			QN					
17-Feb-99	-	2.26	4.44	1.11			QN					
24-Feb-99	-			1.22			ND					

## Appendix H 1,1,1-Trichloroethane Results

100.00 100.00 100.00 53.96 53.96 100.00 0.00 59.55 83.92 62.27 0.00 100.00 0.0 70.65 0/100% n-Propanol | Toluene Groundwater Amended with Nutrient/Yeast and Electron Donor (Col 8) 100.00 100.00 100.00 100.00 100.00 0.00 73.47 100.00 0.00 100.00 100.00 100.00 78.23 0.0 0/100% (Col 7) 100.00 61.36 100.00 100.00 100.00 100.00 0.0 100.00 100.00 100.00 0.00 100.00 0.0 0/100% 77.42 Propionic (Col 6) Acid 0.00 57.62 100.00 75.36 100.00 0/100% 75.53 100.00 100.00 100.00 100.00 100.00 100.00 0.00 100.00 0.0 (Col 5) Lactic Acid 0.00 46.81 100.00 100.00 100.00 100.00 41.53 100.00 100.00 100.00 0.0 100.00 0.00 0/100% 100.00 72.56 Benzoic (Col 4) Acid 0.00 100.00 n-Butyric 0.00 100.00 100.00 100.00 65.12 100.00 100.00 100.00 100.00 100.00 100.00 0.00 0/100% 77.67 (Col 3) Acid 0.00 100.00 100.00 100.00 100.00 100.00 100.00 100.00 100.00 Effluent (Col 2) 100.00 0.0 100.00 0.0 80.00 0/100% Unamended Vitamin/Yeast Groundwater Amended Groundwater 15.45 58.03 100.00 20.35 40.69 -7.40 56.12 17.36 -53.01 00,00 50.88 12.86 -16.68 14.97 28.36 15.74 -53/100% Effluent (Col 1) Percentage Apparent Removal Experiment E Day 32 52 33 15 136 143 150 150 158 165 171 171 85 92 99 Apparent Ave Removal Range 2-Dec-98 23-Dec-98 30-Dec-98 9-Sep-98 30-Sep-98 20-Oct-98 10-Nov-98 6-Jan-99 10-Aug-98 14-Jan-99 21-Jan-99 27-Jan-99 3-Feb-99 10-Feb-99 17-Feb-99 24-Feb-99 Date

Appendix H 1,4-Dichlorobenzene Results

USU/HIII AFB I Pesults in un/l	-BEM CC-1	USU/Hill AFB EM UU-1 Treatability Study Using Ele Desuits in un/l				Groundwa	ter Amend	ed with Nu	Itrient/Yeast	Groundwater Amended with Nutrient/Yeast and Electron Donor	n Donor
	1		Vitamin/Yeast Unamended Vitamin/Yeast	Unamended	Vitamin/Yeast						
		Unamended Amended	Amended	Groundwater Amended	Amended	n-Butyric	Benzoic	Lactic	Propionic		
	Experiment	Experiment Groundwater Groundwater	Groundwater	Effluent	Groundwater	Acid	Acid	Acid	Acid	n-Propanol	Toluene
Date	Day	Influent	Influent	(Col 1)	Effluent (Col 2) (Col 3)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
10-Aug-98	-						1		!	(	
9-Sep-98	31	220.49	21					aN		n/N	
30-Sep-98	52	12.41			23.94			24.48		26.80	36.29
20-Oct-98							12.35	14.44	12.90	16.94	16.02
10-Nov-98		2.44	5.99				8.82	8.78	6.36	6.41	5.27
2-Dec-98	<del></del>	5.99						6.36	4.20	3.39	
23-Dec-98	- 1-										
20-Dec-08	·										
6- Jan-00											
14- Ian-00	-				1.93	2.02	2.18	2.38	3.02	2.10	1.94
01. Jan-99	-										
27- Jan-00	`										
2-Fah-00								1.80			
10-Fah-99	185										
17-Feh-99											
24-Feh-99			2.35	<b>D/N</b>							1.55

## Appendix H 1,4-Dichlorobenzene Results

Percentage Annarent R		t iteataumy on temoval	emoval	Groundwater Amended with Nutrient/Yeast and Electron Donor	r Amended	with Nutri	ent/Yeast a	ind Electron	Donor
		lended	Nutrient/Yeast						
		Groundwater	Amended	n-Butyric	Benzoic	Lactic	Propionic		
	Experiment	nt Effluent	Groundwater	Acid	Acid /	Acid /	Acid	n-Propanol Toluene	Toluene
Date	Day	(Col 1)	Effluent (Col 2) (Col 3)	(Col 3)	(Col 4)	(Col 5) (	(Col 6)	(Col 7)	(Col 8)
10-Aug-98	-								
9-Sep-98	31	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
30-Sep-98	52	-124.66	0.00	0.00	0.00	0.00	0.00	0.00	0.00
20-Oct-98	2	-114.81	-29.57	-143.38	-42.12	-66.17	-48.45	-94.94	-84.35
10-Nov-98	о 	-148.36	-33.89	-49.75	-47.25	-46.58	-6.18	-7.01	12.02
2-Dec-98	<del>.</del>	49.75	34.82	40.43	12.05	-4.95	30.69	44.06	52.81
23-Dec-98	13		42.96	50.74	37.78	33.52	37.78	52.78	53.33
30-Dec-98	4		43.55	45.16	34.56	33.64	36.18	52.07	55.30
6-Jan-99	1	52.37	-0.20	52.76	50.92	51.12	40.70	59.92	60.33
14-Jan-99	15		57.40	55.41	51.88	47.46	33.33	53.64	57.17
21-Jan-99	<i>4</i>	63.96		54.80	57.86	54.37	51.97	60.92	63.97
27-Jan-99	17	-4.55	52.37	58.92	54.63	50.56	59.59	60.27	67.27
3-Feb-99	178	40.45	43.63	48.09		42.68	35.99	45.54	55.73
10-Feb-99	185	-51.95	1.67	12.08	25.83	5.83	8.75	20.00	32.92
17-Feb-99	192	13.64	7.77	15.03	19.17	11.92	-1.55	23.32	22.28
24-Feb-99		0.00	31.91	40.85	44.68	29.36	40.43	17.87	34.04
	Range	-148/100%	-34/100%	-143/100% -47/100% -66/100%	-47/100%	-66/100%	-48/100%	-94/100%	-84
Apparent A	Apparent Ave Removal	2.24	27.41	25.41	29.94	22.85	27.95	32.56	38.85

Appendix H Chiorobenzene Results

USU/Hill AF	-B EM OU-1	Treatability St	tuay using Electi		JSU/Hill AFB EM OU-1 Treatability Study Using Electron Donors to Enhance Decimanan Documents in train	Groundwat	ter Amende	ed with Nu	itrient/Yeast	Groundwater Amended with Nutrient/Yeast and Electron Donor	n Donor
			Vitamin/Yeast	Unamended	Vitamin/Yeast						
		I inamended Amended		Groundwater Amended	Amended	n-Butyric	Benzoic	Lactic	Propionic		;
	Experiment	Experiment Groundwater Groundwater	Groundwater	Effluent	Groundwater	Acid	Acid //	Acid	Acid	anol	Toluene
Date	Day	Influent	Influent	(Col 1)	Effluent (Col 2) (Col 3)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
10-Aug-98	-									00 10	76 67
9-Sep-98	31	107.96	-	42.17	-		38.64	48.33		30./3	00.00
30-Sen-98	52	Ω/N		12.66		13.18	13.93	13.58	-	12.05	14.00
		12 42					9.81	7.89		8.87	7.57
						-	10.68	10.53	<b>v</b>	12.54	12.76
	Ŧ		15.11	4.60	9.02	8.37		10.84	7.50		7.40
	- •	Ŧ					8.68			9.00	
23-DEC-90											
30-Dec-98											
6-Jan-99	•										
14-Jan-99	•	10.29									
21-Jan-99	165	13.90									
07-101-00		•									
								8.85			
10-Feb-99											
17-Feb-99	9 192	2.51									
24-Feb-99	·	3.03		2.27							

## Appendix H Chlorobenzene Results

Percentage Removal				Groundwa	ter Amende	ed with Nut	irient/Yeast	Groundwater Amended with Nutrient/Yeast and Electron Donor	n Donor
		Unamended	Nutrient/Yeast						
		Groundwater Amended	Amended	n-Butyric	Benzoic	Lactic	Propionic		
	Experiment Effluent	Effluent	Groundwater	Acid	Acid /	Acid /	Acid	n-Propanol	Toluene
Date	Day	(Col 1)	Effluent (Col 2)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7)	(Col 8)
10-Aug-98	1								
9-Sep-98	31	60.94	53.39	61.85	61.87	52.31	64.53	63.76	63.92
30-Sep-98	52	0.00	10.34	16.37	11.61	13.83	-3.74	23.54	11.17
20-Oct-98		84.14	73.35	65.04	61.33	68.90	62.24	65.04	70.16
10-Nov-98		46.57	-18.85	-58.85	-36.05	-34.14	-38.09	-59.75	-62.55
2-Dec-98	115	41.40	40.30	44.61	43.41	28.26	50.36	35.80	51.03
23-Dec-98	136	66.40	40.04	49.34	51.99	48.23	53.32	50.22	50.11
30-Dec-98	-	31.81	36.15	38.62	49.48	37.65	37.52	45.64	45.71
6-Jan-99		47.23	49.72		58.98	50.16	48.48		51.46
14-Jan-99	-	51.12		54.77	60.82	54.02	49.47	55.52	43.54
21-Jan-99	~	61.01	57.65	54.93	63.34	58.26	57.11	55.84	51.42
27-Jan-99		31.32	41.67	41.28	44.20	41.09	52.69	43.68	48.28
3-Feb-99	-	-191.15	13.09	16.70	20.11	16.03	13.95	15.46	17.46
10-Feb-99	185	-171.81	-32.48	-35.95	-65.33	-29.93	-35.04	-42.88	-24.64
17-Feb-99	192	44.73	-18.00	-2.67	5.56	7.11	-10.00	12.89	14.00
24-Feb-99	<b>·</b>	25.08	29.32	35.89	35.07	23.56	36.03	19.04	38.08
	Range	-191/84%	-32/73%	-59/65%	-65/62%	-34/69%	-38/65%	-60/65%	-63/70%
Apparent Ave Remov	ve Removal	15.25	28.92	28.81	31.09	29.02	29.25	29.25	31.28

### Appendix I Chloride Calculations and Results

					Potential
			Chlorine	Qty Added to Feed	
Amendment	Formula	M.W.	Fraction	Water (mg/L)	(mg/L)
d-Biotin	C <sub>10</sub> H <sub>16</sub> N <sub>2</sub> O <sub>3</sub> S	244.3	0.000	20	
Folic acid	C <sub>19</sub> H <sub>19</sub> N <sub>7</sub> O <sub>6</sub>	441.4	0.000	20	
Pyridoxine hydrochloride $(B_6)$	C <sub>8</sub> H <sub>11</sub> NO <sub>3</sub> .HCl	205.6	0.058	100	5.84
Thiamin hydrochloride(B <sub>1</sub> )	C <sub>12</sub> H <sub>17</sub> CIN₄OS.HCl	337.3	0.036	50	1.78
Riboflavin(B <sub>2</sub> )	C <sub>17</sub> H <sub>20</sub> N₄O <sub>6</sub>	376.4	0.000	50	1 1
Nicotinic acid	C <sub>6</sub> H₅NO₂	123.1	0.000	50	
DL-calcium pantothenate	C <sub>9</sub> H <sub>16</sub> NO₅.1/2Ca	238.3	0.000	50	0.00
Vitamin B <sub>12</sub> (cyanocobalamin)	C <sub>63</sub> H <sub>88</sub> CoN <sub>14</sub> O <sub>14</sub> P	1355.4	0.000	10	0.00
p-Aminobenzoic acid	C <sub>7</sub> H <sub>7</sub> NO₂	137.1	0.000	50	0.00
Lipoic acid	$C_8H_{16}O_2S_5$	208.3	0.000	50	0.00
Resazurin	C <sub>12</sub> H <sub>6</sub> NO₄Na	251.2	0.000	1	0.00
Yeast				20	0.00
Sodium Bicarbonate	NaHCO₃	84.01	0.000	1000	0.00
	Total Potential Chlo	oride From	Amendmer	nts =	7.62

Potential Chloride Loading from Amendment Decomposition

# Appendix I Chloride Calculations and Results

	1
~	1
de	-11-
<u>G</u>	
Ĕ	j
0	C

Results in ma/L	na/l-					Groundwat	er Amend	ed with Nu	trient/Yeast	Groundwater Amended with Nutrient/Yeast and Electron Donor	on Donor
	<u></u>		Vitamin/Yeast	Unamended	/itamin/Yeast  Unamended  Vitamin/Yeast						
	_	Unamended Amended	Amended	Groundwater Amended		n-Butyric Benzoic Lactic	Benzoic		Propionic		
	Experiment	Experiment Groundwater Groundwater	Groundwater	Effluent	Groundwater	Acid	Acid	Acid		n-Propanol Toluene	Toluene
Date	Dav	Influent	Influent	(Col 1)	Effluent (Col 2) (Col 3) (Col 4) (Col 5)	(Col 3)	(Col 4)	(Col 5)	(Col 6)	(Col 7) (Col 8)	(Col 8)
21-Jan-99				-		74.14 74.19 67.08	67.08	69.85	71.32	70.72	64.74

1.47 7.45 8.05 6.58 3.81 10.92 10.87 -5.65 mg/L Produced