Enhanced Aerobic bio-remediation of Chlorinated Solvents at a Strip Mall in York, PA

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1 Enhanced Aerobic Bio-remediation Report

Introduction

In this report, we present data collected on *aerobic* bio-remediation of soil or soil-gas locations contaminated with Perchloroethene (PCE), Trichloroethene (TCE), cis-1,2-Dichloroethene (cis12DCE) and Vinyl Chloride (VC) using VaporRemed.

VaporRemed is included in the (National Contingency Planning Product Schedule) NCP Product Schedule (Products Available for Use on Oil Spills) as a bio-remediation agent.
VaporRemed (aka SpillRemed (Industrial)) is also (accepted) by the Florida Department of Environment Protection as a product for bio-remediation of petroleum and other suitable contaminants in groundwater.
VaporRemed is mentioned as part of (Heating Oil Release Information) along with OdorGone. The section on OdorGone and VaporRemed tries to distinguish masking agents from bio-remediation agents and we are happy to clarify that VaporRemed breaksdown petroleum hydrocarbons as well as chlorinated solvents.

Enhanced bio-remediation of Chlorinated Hydrocarbons

National Research Council. 1993. In Situ Bio-remediation: When Does it Work? [1] lists three forms of evidence for demonstrating that *in-situ bio-remediation* is effective for a given project:

1. documented loss of contaminants from the site due to bio-remediation,
2. laboratory assays showing that the microorganisms in site samples have the *potential* to transform the contaminants under expected site condition, and
3. one or more pieces of evidence showing that the bio-remediation potential is *actually realized* in the field.

1.1 Enhanced Aerobic Bio-remediation of Chlorinated Hydrocarbons using VaporRemed

1 - Documented loss of contaminants from the site

Aerobic Degradation - It has long been thought that TCE is resistant to degradation under aerobic conditions due to its already oxidized state. Recently, a number of monooxygenases produced under aerobic conditions have been shown to degrade TCE (Nelson et al., 1987; Harker and Young, 1990). This report corroborates our assertion that TCE can be degraded under aerobic conditions: Link. We believe that this paper provides us with sufficient to test the hypothesis on the field.

2 - Laboratory assays showing that microorganisms in the site samples have the potential

The figure 1 displays a modified version of the Hazen table, link here, highlighting that the following compounds can be broken down by cometabolic bioremediation.

VaporRemed is a bio-remediation agent comprising of a consortium of bacteria listed here

- *Pseudomonas alkaligenes*
- *Phenylobacterium immobile*
- *Stentrophomonas maltophilia*
- *Gluconobacte cerinus*
- *Agrobacter radiobacter*

Most of the literature supports the hypothesis that *Pseudomonas* can remediate chlorinated hydrocarbons in anaerobic conditions, however, bio-remediation of chlorinated hydrocarbons under aerobic conditions has not been documented before.

3 - One or more pieces of evidence showing that the bio-remediation potential is actually being realized at the site.

This section outlines the effectiveness of VaporRemed as an agent accomplishing enhanced aerobic bio-remediation of chlorinated solvents by collecting data after adding VaporRemed to various locations at the site. We started with soil-bore locations, identified as SB-121, then proceeded to sub-slab areas VP-003, followed by near-source soil gas locations, identified as SG-101 and SG-106, while at all times measuring the indoor air levels in the rooms in the lower levels including IA-001.

1. Soil-bore analysis

   The table below shows the degradation observed after introducing AgroRemed to upto 10' bgs at SB 121 cluster of locations.

	Cometabolic Bioremediation Conditions				
	Aerobic	**Aerobic**	**Aerobic**	**Anaerobic**	**Anaerobic**
Contaminants	• TCE • DCE • VC • PAHs • PCBs • MTBE • Creosote • >300 other compounds	• TCE • DCE • VC • TNT	• TCE • DCE • VC • 1,1-DCE • 1,1,1-TCA • MTBE	• PCE • TCE • DCE • VC • Hexachlorocyclohexane	• BTEX • PCE • PAHs • Atrazine • TNT
Substrates	• Methane • Methanol • Propane • Propylene	• Ammonia • Nitrate	• Toluene • Butane • Phenol • Citral • Cumin Aldehyde • Cumene • Limonene	• Methanol	• Glucose • Acetate • Lactate • Sulfate • Pyruvate
Microoganism(s)	• *Methylosinus*	• *Nitrosomonas* • *Nitrobacter*	• *Rhodococcus* • *Pseudomonas* • *Arthrobacter*	• *Pseudomonas* • *Streptomyces* • *Corynebacterium*	• *Dehalococcoides* • *Methanogens* • *Desulfovibrio* • *Clostridium* • *Geobacter* • *Clavibacter*
Enzyme(s) produced	• Methane monooxygenase • Methanol dehydrogenase • Alkene mono-oxygenase • Catechol dioxygenase	• Ammonia monooxygenase	• Toluene monooxygenase • Toluene dioxygenase	• Alcohol dehydrogenases	• Dehalogenase • AtzA • Dichloromethane Dehalogenase

Modified from Hazen (2010)

Figure 1: Modified table from Hazen (2010)

Table 1: Contamination levels at SB 121

UTCTime	PCE
06/28/2017 00:00	2680000.00
09/11/2017 00:00	73.00

Table 1 shows that in 70 days both PCE and TCE reduced by more than 90 % in soil. AgroRemed is commercially available to help reduce TPH levels in soil, groundwater by 90 % for over a decade. However, this is the *first* project where AgroRemed is being applied to reduction of PCE/TCE levels in soil.

This reduction is significant and also provides the first clear evidence of aerobic bio-remediation of PCE/TCE at the site. Further, a significant in-

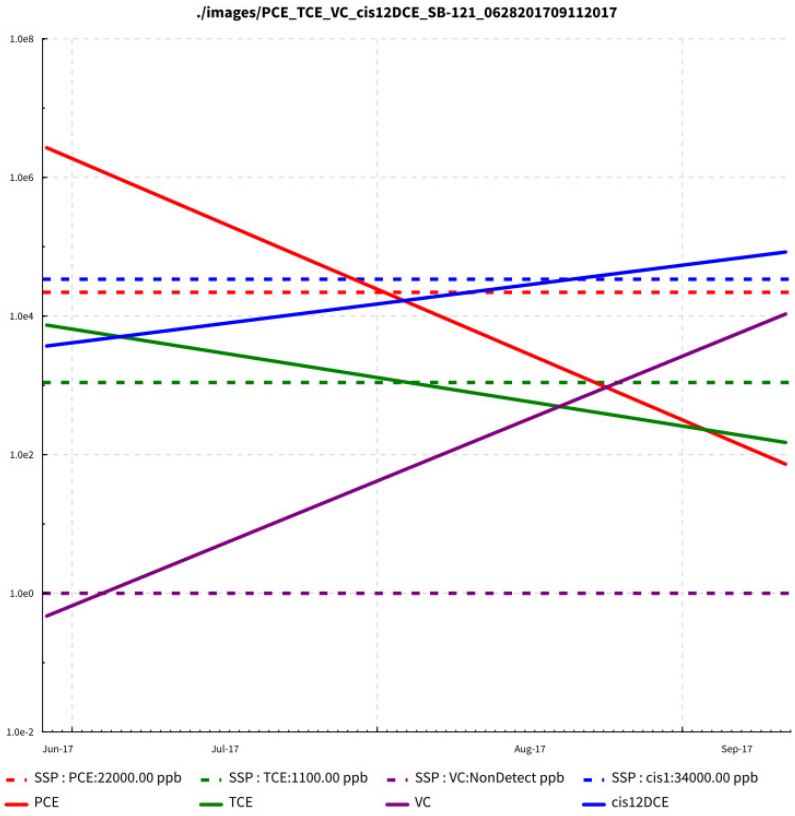

Figure 2: Contamination levels at SB 121, results in ppb*

crease in the values of cis-1,2-Dichloroethene (cis12DCE) and Vinyl Chloride (VC) by 2167 % and 2127 % respectively confirms the evidence of active aerobic bio-remediation as reported in the literature. The significant reduction in the values of PCE and TCE supports our hypothesis that remediation of both these contaminants is being accomplished under aerobic conditions.

First-order decay formula used throughout the report
Definition : A quantity is subject to exponential decay if it decreases at a rate proportional to its current value. In this report and the attached source code, the following formula is being used.

$$rate = oneDay * (\ln r2 - \ln r1)/timeInSeconds$$

where

$$oneDay = 24 * 3600$$

seconds
where *timeInSeconds* is difference in time in seconds, *r1* is the contamination level at time T1, *r2* is the contamination level at time T2.
Further, as one is dealing with decay rates of PCE + TCE, the model accounts for the PCE decay by accumulating the decreased amount completely into the concentrations of TCE. The current model is not ideal because it doesn't account for "daughter" compounds such as cis12DCE and *trans* Trichloroethene. Further, the model is as approximation as the ppb values are being used instead gram molar concentrations.

Computed first order decay for SB-121

- PCE = -14.00 %
- TCE = -0.005 %

Some more notes about the plot
Assuming a first order decay of PCE in the plot above, one can estimate the PCE levels at any point between 6/28 and 9/11. For example, it can be seen that the site specific levels for PCE were reached at the site in the first week of August, 2017.

2. Sub-slab Soil Gas Analysis

   Historical data at VP 3 located in the mechanical room/pump room has shown high concentrations of the chemicals of concerns. Therefore, the effectiveness of VaporRemed was tested by introducing VaporRemed directly into VP-003 and monitoring the results over a period of *2 hours*. [1] [2]

Table 2: Contamination levels of COCs at VP-003.

UTCTime	PCE
02/21/2018 00:00	107000.00
04/06/2018 00:00	587.00
06/21/2018 00:00	32000.00
07/25/2018 00:00	55600.00
08/28/2018 00:00	15500.00
09/26/2018 00:00	99.20
05/22/2019 00:00	568.00
05/23/2019 00:00	445.00
07/01/2019 00:00	470.00

Computed first order decay rate for PCE and TCE on 7/25/2018

- PCE = -800 %
- TCE = -762 %

For example, based on the above rates, one can estimate that it will take about 6 to 8 hours to bring 55,000 ppm to close to site-specfic limits. What this implies is that a project can establish tighter controls over various locations and measure for exceedances.

In order to confirm whether VP 3 is the primary source of vapor intrusion, another sub-slab soil gas sampling location, identified as EPS 1 was selected just below the dock in the basement. The contaminations recorded in EPS 1 were not significant as shown below.

[1] VaporRemed was not injected under pressure; this is important as injection increases project costs and require the DEP's approval.

[2] Same day measurements were taken 2 hours after adding VaporRemed to the location. For example on 7/25/2018 VaporRemed was added at 9:00 am.

Table 3: Contamination levels measured at EPS-1

Date	PCE	TCE
2/21/2018	876	107
5/17/2018	412	84.6
8/28/2018	585	833

Bacterial counts in VP-003 between Mar 23rd and Mar 25th 2020
Bacterial counts are an important parameter for measuring the effectiveness of bio-remediation. The fact that the bacterial population is continuing and growing indicates that the conditions in the sub-slab soil-gas location are *not* anaerobic.

- : Bacterial count after 2 hours was **14,800,000**
- : Bacterial count after 72 hours was **15,300,000**

The sustained population count indicates that PCE and TCE contamination levels at VP-003 are not toxic to the bacteria in VaporRemed.

3. Monitoring Indoor Air Quality

   A total of 12 Indoor Air sampling locations were selected for monitoring indoor air quality. However, *focal point* was the sampling station IA 001 which is just outside the sub-slab monitoring point VP 3. Data collected at each of the sampling locations is presented below. The sampling points IA 001, IA 002 and IA 003 are in the basement and closer to VP 3, the source of vapor intrusion. The higher concentrations of PCE and TCE in these rooms is a direct result of vapor intrusion from VP 003. Application of VaporRemed has shown to bring the values closer to their respective site-specific levels.

   - IA 001: Basement: Just outside VP 3 sub-slab sampling point
     The location was measured at a greater frequency than the other locations as this location is closest to VP 3.

It was observed that after summer of 2019 the results for Vinyl Chloride were non-detect. This degradation can be attributed to the bio-remediation of Vinyl Chloride by VaporRemed. This confirms the results of an earlier project wherein biodegradation of poly vinyl chloride (PVC) was evaluated both under laboratory and semi-industrial scale and it was observed that the bacterial consortium in VaporRemed were effective in bio-degradation of PVC.

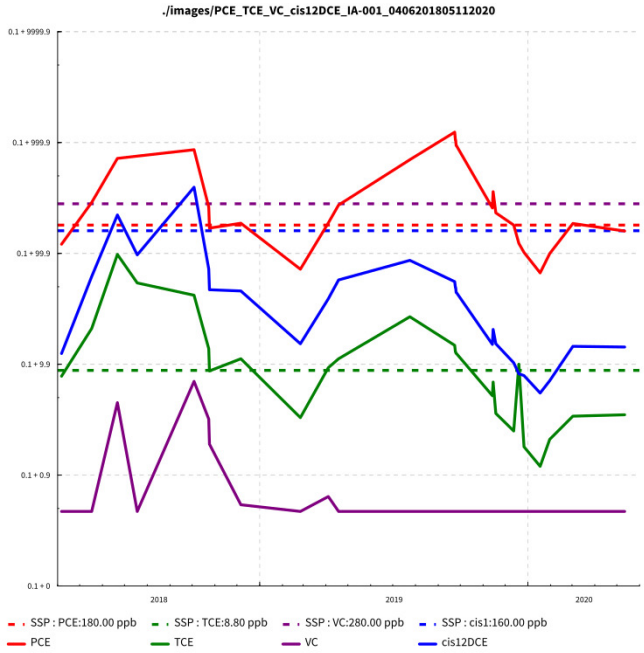

Figure 3: Values of PCE/TCE at IA-001

- IA 002: Basement: the Middle room in the basement away from VP 3
- IA 003: Basement: Just below the warehouse and the loading dock
- IA 004: H Block closer to the rear parking lot away from VP 3
- IA 005: Restroom away from VP 3
- IA 006: A room identified as a Vault away from VP 3
- IA 007: Yoga room In the line of VP 3
- IA 008: WIS office in the line of VP 3
- IA 009: Ambient air on top of the warehouse away from VP 3

Table 4: Documented reduction of TCE/PCE in IA-001

UTCTime	PCE
04/06/2018 00:00	121.00
05/17/2018 00:00	288.00
06/21/2018 00:00	719.00
07/18/2018 00:00	754.00
10/03/2018 00:00	861.00
10/23/2018 00:00	262.00
10/24/2018 00:00	169.00
12/06/2018 00:00	187.00
02/25/2019 00:00	72.00
04/04/2019 00:00	188.00
04/18/2019 00:00	273.00
07/24/2019 00:00	700.00
09/23/2019 00:00	1240.00
09/24/2019 00:00	1050.00
09/25/2019 00:00	939.00
11/13/2019 00:00	257.00
11/14/2019 00:00	360.00
11/18/2019 00:00	232.00
12/12/2019 00:00	180.00
12/19/2019 00:00	123.00
12/26/2019 00:00	102.00
01/17/2020 00:00	66.70
01/30/2020 00:00	99.50
03/02/2020 00:00	186.00

Table 5: Documented reduction of TCE/PCE in IA-002

UTCTime	PCE
07/18/2018 00:00	244.00
02/26/2019 00:00	102.00
04/04/2019 00:00	190.00
07/24/2019 00:00	603.00
01/20/2020 00:00	86.40

- IA 010: Store in the front D Block: Jewelry store away from VP 3
- IA 011: Store in the front C block away from VP 3
- IA 012: Store in Front C block away from VP 3

Table 6: Documented reduction of TCE/PCE in IA-003

UTCTime	PCE
02/27/2019 00:00	132.00
04/04/2019 00:00	188.00
07/24/2019 00:00	739.00

Table 7: Documented reduction of TCE/PCE in IA-004

UTCTime	PCE
02/28/2019 00:00	98.20
04/04/2019 00:00	160.00
07/24/2019 00:00	607.00
01/20/2020 00:00	101.00

Table 8: Documented reduction of TCE/PCE in IA-005

UTCTime	PCE
04/04/2019 00:00	146.00
07/24/2019 00:00	818.00

Table 9: Documented reduction of TCE/PCE in IA-006

UTCTime	PCE
04/04/2019 00:00	117.00
07/24/2019 00:00	439.00

Table 10: Documented reduction of TCE/PCE in IA-007

UTCTime	PCE
01/17/2020 00:00	62.40

Table 11: Documented reduction of TCE/PCE in IA-008

UTCTime	PCE
04/04/2019 00:00	196.00
07/24/2019 00:00	524.00
01/17/2020 00:00	62.80

Table 12: Documented reduction of TCE/PCE in IA-009

UTCTime	PCE
04/18/2019 00:00	75.20
07/24/2019 00:00	1.10
01/20/2020 00:00	4.30

Table 13: Documented reduction of TCE/PCE in IA-010

UTCTime	PCE
07/18/2018 00:00	32.10
12/06/2018 00:00	34.30
04/18/2019 00:00	7.70

Table 14: Documented reduction of TCE/PCE in IA-011

UTCTime	PCE
01/20/2020 00:00	48.10

Table 15: Documented reduction of TCE/PCE in IA-012

UTCTime	PCE
04/18/2019 00:00	31.50

4. Bio-remediation of Soil Gas using VaporRemed

   We decided to introduce VaporRemed at three new injection points drilled near SG 101 and SG 106. The holes at SG 101 were shallow less than 1' as standing water was recorded at this depth. At SG 106, these injection points were drilled to 6' below surface.

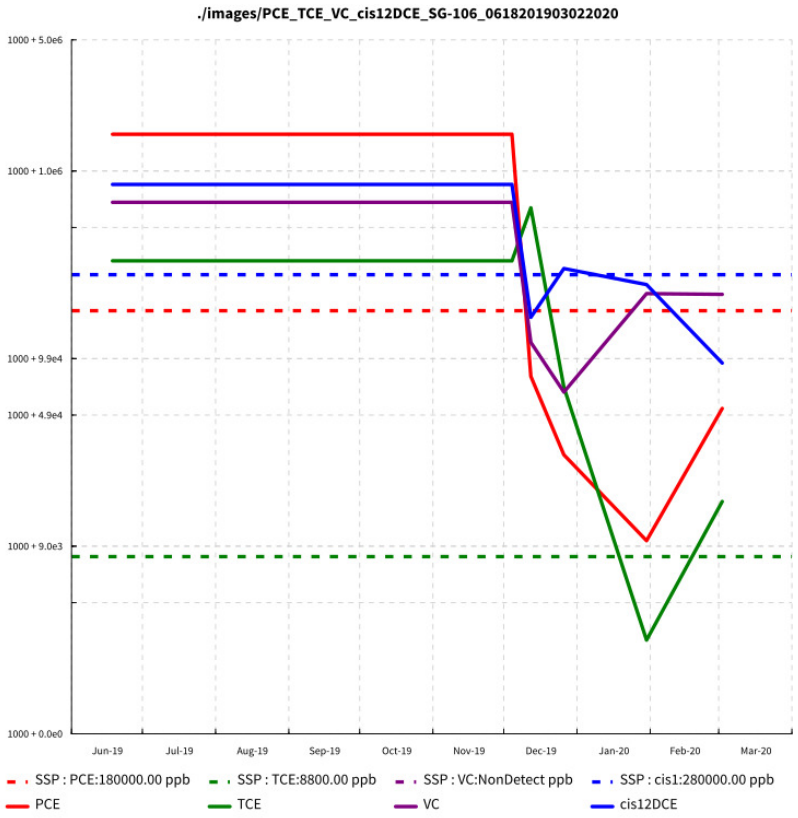

Figure 4: Values of COCs between 2019 - 2020

Table 16: Soil Gas Values in SG 106

UTCTime	PCE
06/18/2019 00:00	1570000.00
12/04/2019 00:00	1570000.00
12/12/2019 00:00	80400.00
12/26/2019 00:00	30700.00
01/30/2020 00:00	10700.00
03/02/2020 00:00	54200.00

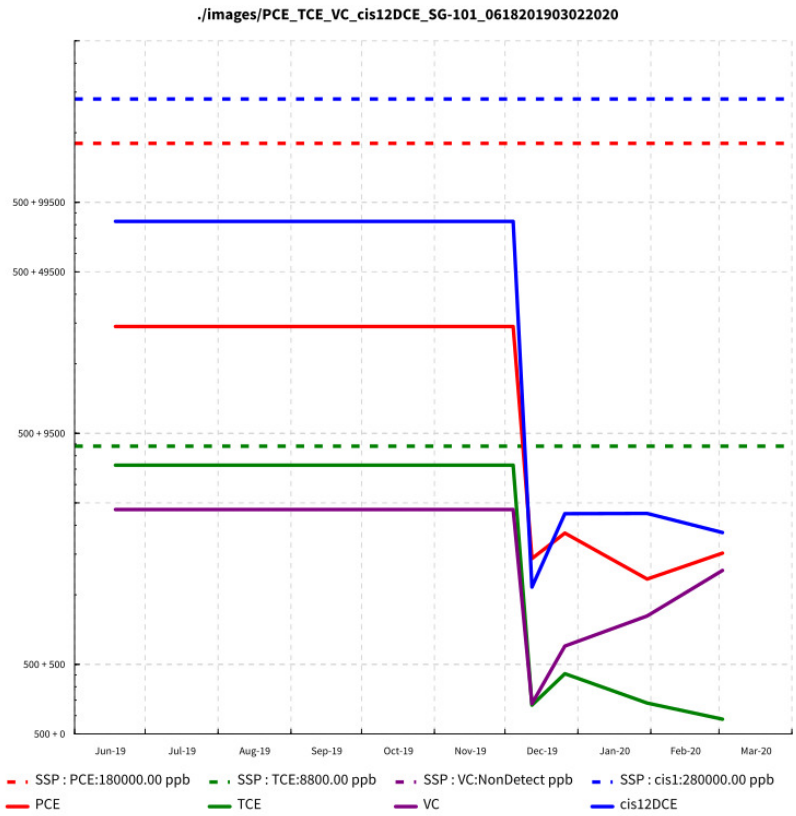

Figure 5: Values of COCs between 2019 - 2020 at SG-101

Table 17: Soil Gas Values in SG 101

UTCTime	PCE
06/18/2019 00:00	29000.00
12/04/2019 00:00	29000.00
12/12/2019 00:00	2870.00
12/26/2019 00:00	3700.00
01/30/2020 00:00	2340.00
03/02/2020 00:00	3030.00

2 Conclusions

The study was carried out for almost two years with close consultations with the PaDEP. Based on the data collected and presented above, we enumerate the following observations:

1. Enhanced aerobic bio-remediation of chlorinated hydrocarbons was demonstrated at various soil matrices.
2. SB 121 cluster showed enhanced bio-remediation in soil due to addition of VaporRemed.
3. VP 3 was identified as a primary source of Vapor intrusion in the basement; VaporRemed was added to VP 3 to control contaminant levels.
4. SG-101, SG-106 were important sources of TCE/PCE contamination and remediation of the rest of the site.

Finally, we conclude that aerobic bio-remediation of TCE and PCE was observed at site; thereby providing a field engineer with an additional set of tools to help cleanup a site contaminated with TCE/PCE.

3 Open Source Libraries and Software Used to Create this report

- Chart : A library for generating 2D charts and plots
- Haskell Programming Language
- Evaluating Vapor Intrusion Pathways
- Evualation of spatial and temporal variability in VOC concentrations at Vapor Intrusion Investigation Sites.

4 Source code and data

Source-code and collected data is committed on github.com and is available upon request.

5 Reviewers

- Steve Vedder - svedder@epsofvermont.com
- Satya Ganti - sales@sarvabioremed.com

6 Bibliography

[1] National Research Council. *In Situ Bioremediation: When Does it Work?* The National Academies Press, Washington, DC, 1993.

7 References

- TO-15
- DNR Guidance on Vapor Intrusion

Revision History

Revision	Date	Author(s)	Description
1.0	04.12.2020	DG	created
1.1	04.12.2020	DG	modified, changed voice, added more rows to vp3
1.2	04.29.2020	DG	More edits