Scientific and Engineering Considerations for Cost-Effective In Situ Bioremediation of Large, Deep Plumes

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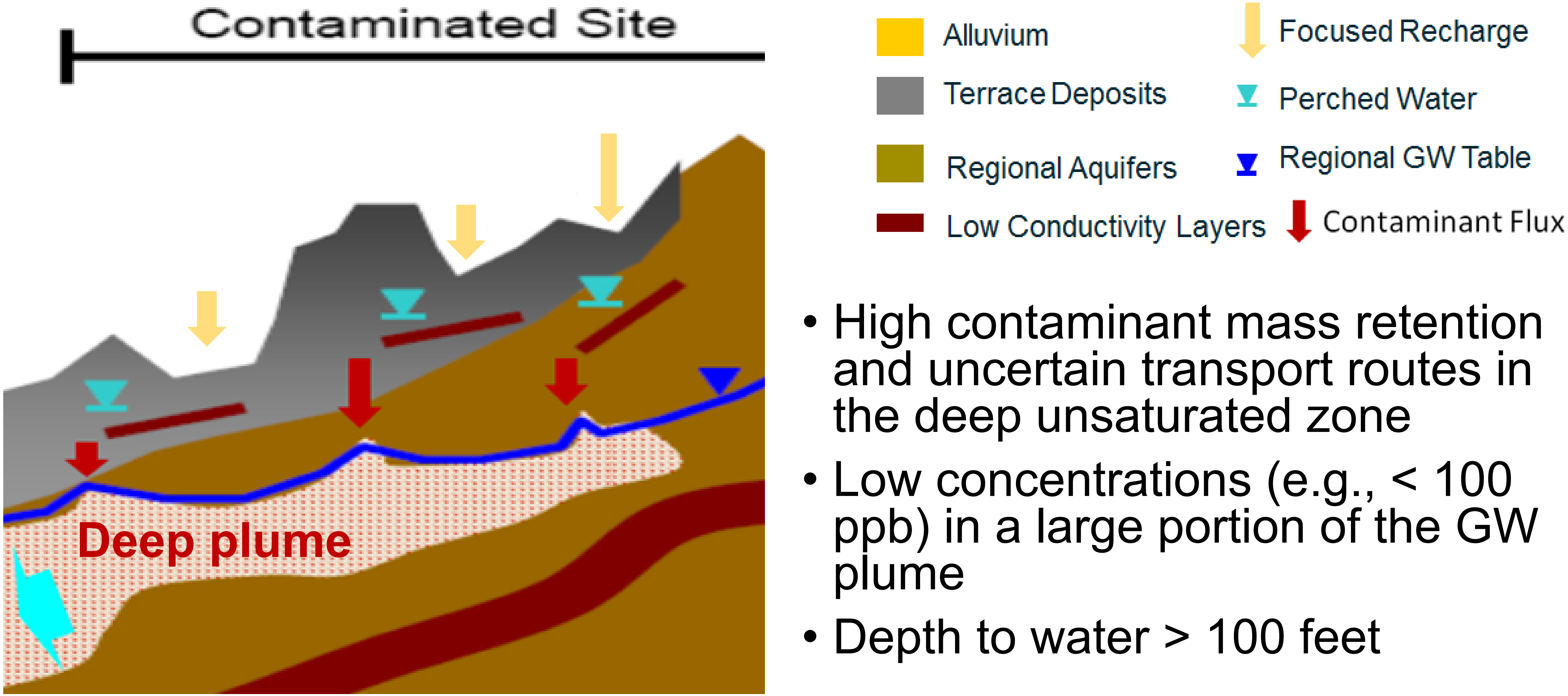
Example Settings for a Large, Deep Plume



- High contaminant mass retention and uncertain transport routes in the deep unsaturated zone
- Low concentrations (e.g., < 100 ppb) in a large portion of the GW plume
- Depth to water > 100 feet

Challenges in Treating Large, Deep Plumes

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STRATEGIC ENVIRONMENTAL RESEARCH AND DEVELOPMENT PROGRAM (SERDP)
ENVIRONMENTAL RESTORATION (ER) FOCUS AREA

FY 2009 STATEMENT OF NEED

REDUCED UNCERTAINTY AND COSTS FOR MANAGING LARGE, DILUTE CONTAMINANT GROUNDWATER PLUMES

1. Develop more cost-effective techniques to remediate large, dilute groundwater plumes, including techniques to enhance natural attenuation processes. The focus is on deep plumes where traditional reactive barriers are ineffective.

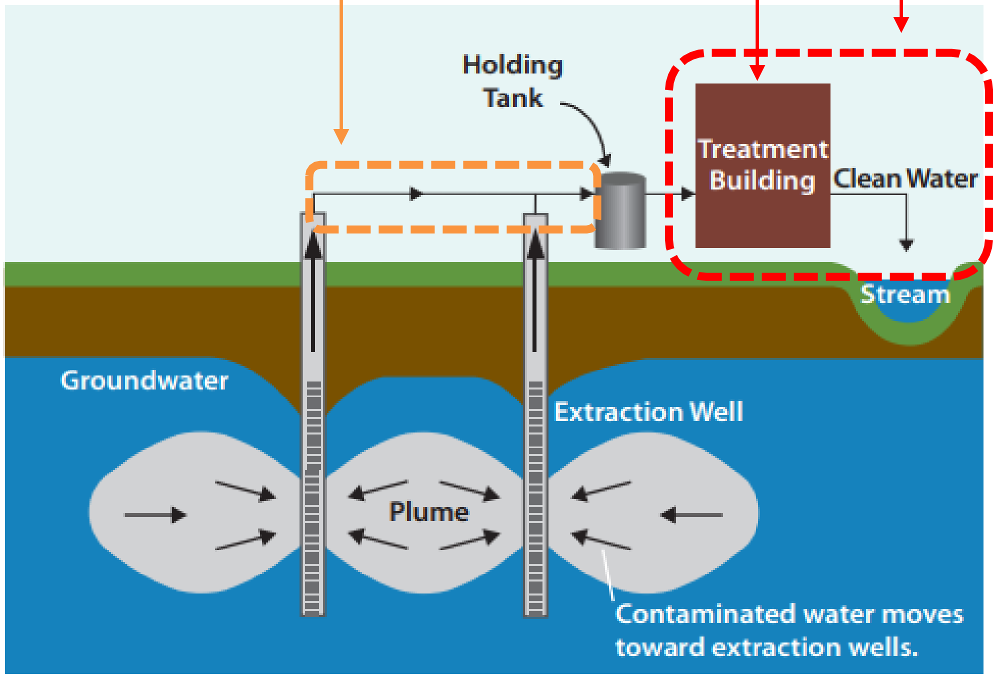
Alternative to Pump-and-Treat (P&T)?

A pump-and-treat (P&T) system is often a remedial component for the large, deep plume management

- Quick, robust plume containment
- Many tools to deal w/ a suite of contaminants (treatment trains)
- Easier maintenance for an above ground system

Potential challenges:

- Large above-ground footprint for treatment complex
- Short reaction time needed for reactor
- High cost for conveyance piping



Example of a pump and treat system with two extraction wells.

https://semspub.epa.gov/work/HQ/401617.pdf

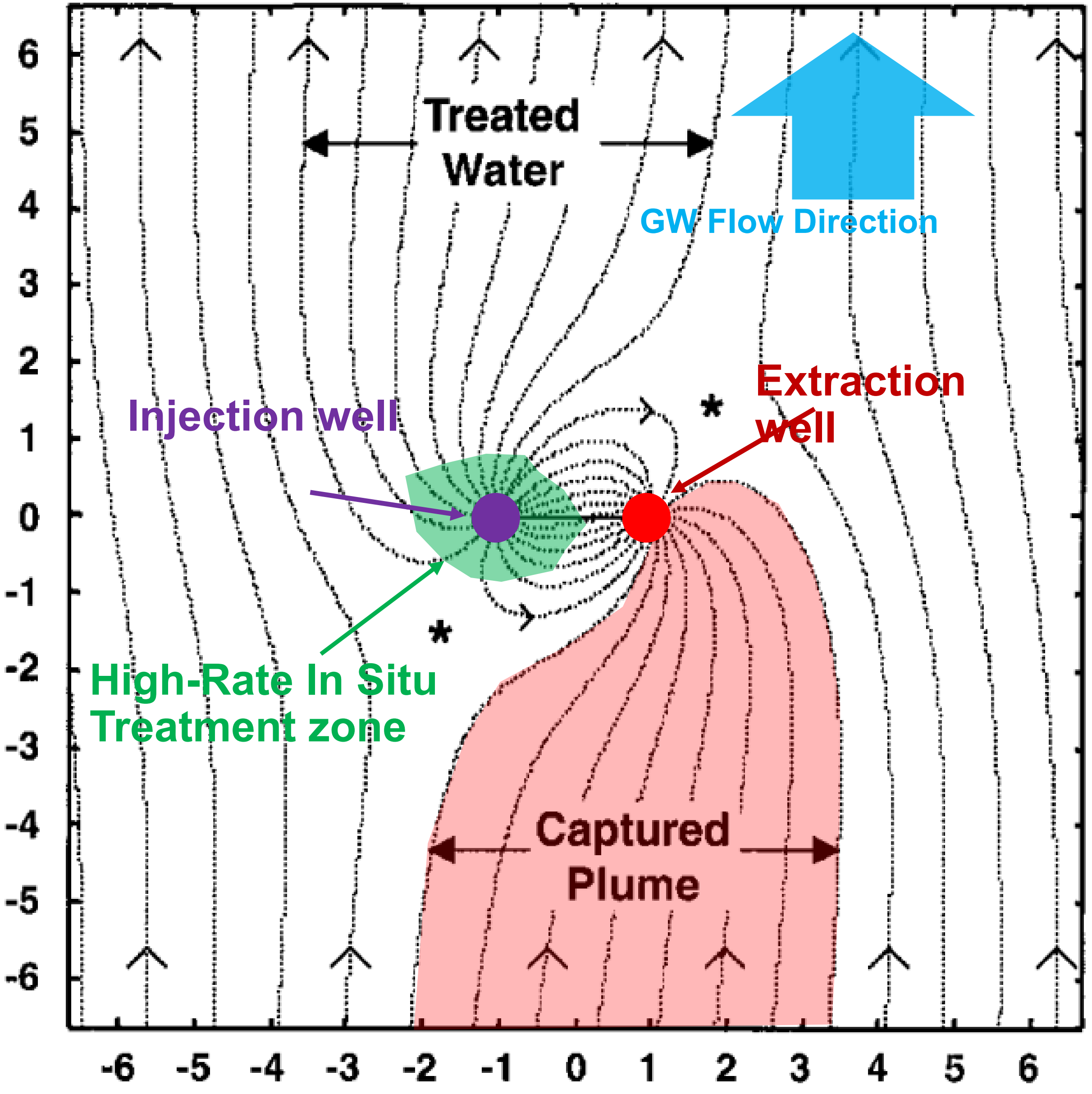
Can a hydraulic reactive barrier be a cost-effective alternative for P&T?

ground water

Injection–Extraction Treatment Well Pairs: An Alternative to Permeable Reactive Barriers

by Jeffrey A. Cunningham[1] and Martin Reinhard[2]

Vol. 40, No. 6—GROUND WATER—November–December 2002 (pages 599–607



The high-rate in Situ treatment zone receives constant flow of impacted groundwater

Benefits of a high-rate in situ bioreactor (HR-ISB)

- Less energy consumption than advanced oxidation treatment
- Often a lower operation & maintenance cost than an advanced oxidation process (AOP)
- No size restriction for an underground reactor (i.e., no residence time constraint)

Challenges/Limitations?

- May not handle a sudden increase in contaminant loading
- No quick way to do ***backflush*** (like an above ground fix-bed bioreactor)
- Bioclogging
  - Making injection impossible eventually
  - Reduce treatment efficiency due to short circuiting

Bioclogging in a pump

What's next

- Use of high-rate in situ bioreactors (**HR-ISB**) as a cost-effective alternative for P&T
  - Examples 1 & 2: Metabolic anaerobic biotransformation
  - Example 3: Cometabolic chloroethenes and 1,4-dioxane biodegradation
- **Bioclogging**, a common nuisance for **HR-ISB**, which reduces treatment performance and system uptime
- **Chemical toxicity**: a double-edge sword!
- **Deficiency in native consortia**: Intrinsic limiting factor

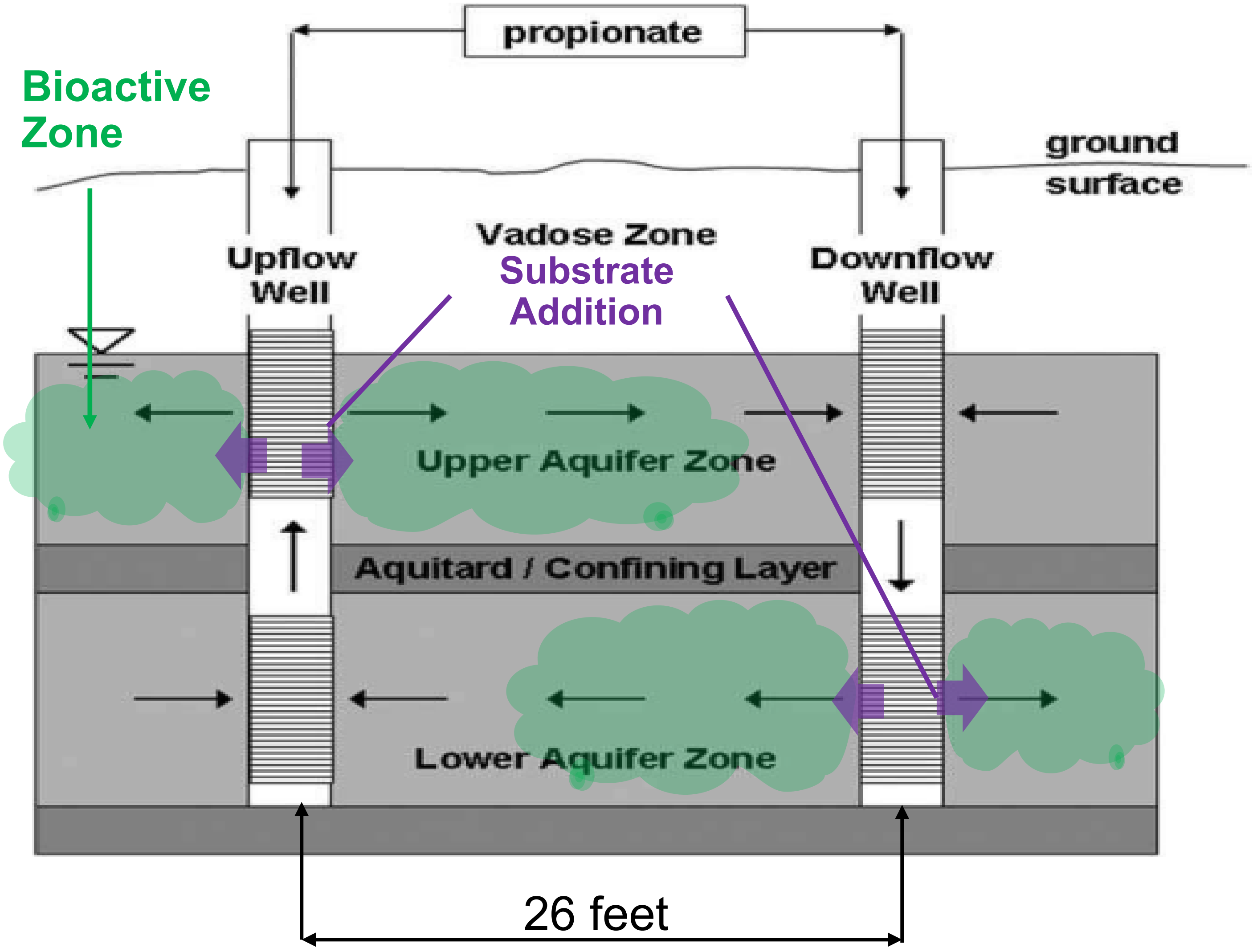
Example 1: HR-ISB for dehalogenation of cDCE

- Well spacing = 26 feet; **constant recirculation (mark spacing underneath the figure)**
- Propionate **recirculated** between well pairs **constantly (0.37 gpm)**
- Take ~ 8 weeks to reach stable, optimal performance
- ~ 95% treatment efficiency for cDCE over one week in the bioactive zone (t1/2= 1.6 day)
- Near-complete removal of ~250 mg/L sulfate accompanied the rapid dechlorination, but no methanogenesis was observed
- Water head increased 4.6 ft over ~ 8 weeks

Ground Water Monitoring&Remediation
Ground Water Monitoring & Remediation 26, no. 3/ Summer 2006/pages 82–91

Bioremediation of cis-DCE at a Sulfidogenic Site by Amendment with Propionate

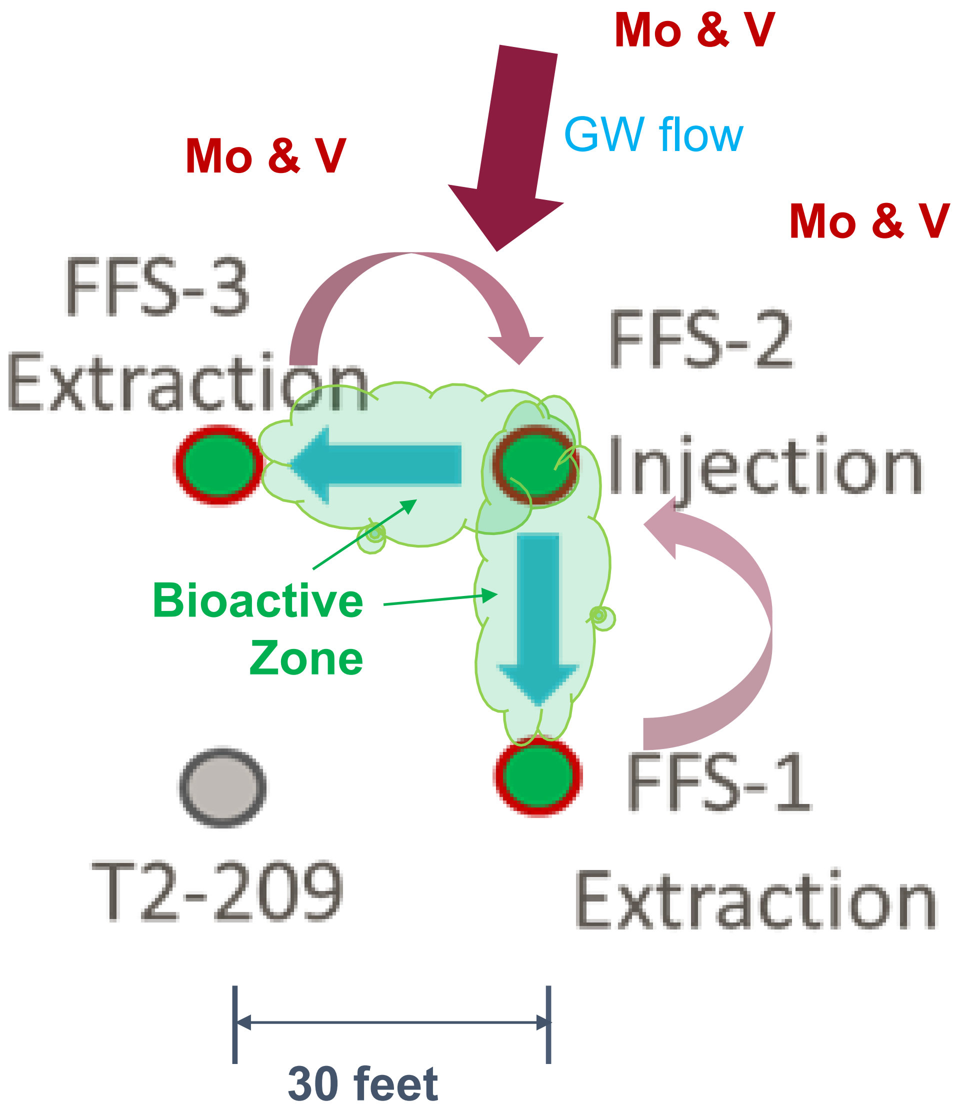
by T.P. Hoelen, J.A. Cunningham, G.D. Hopkins, C.A. Lebron, and M. Reinhard



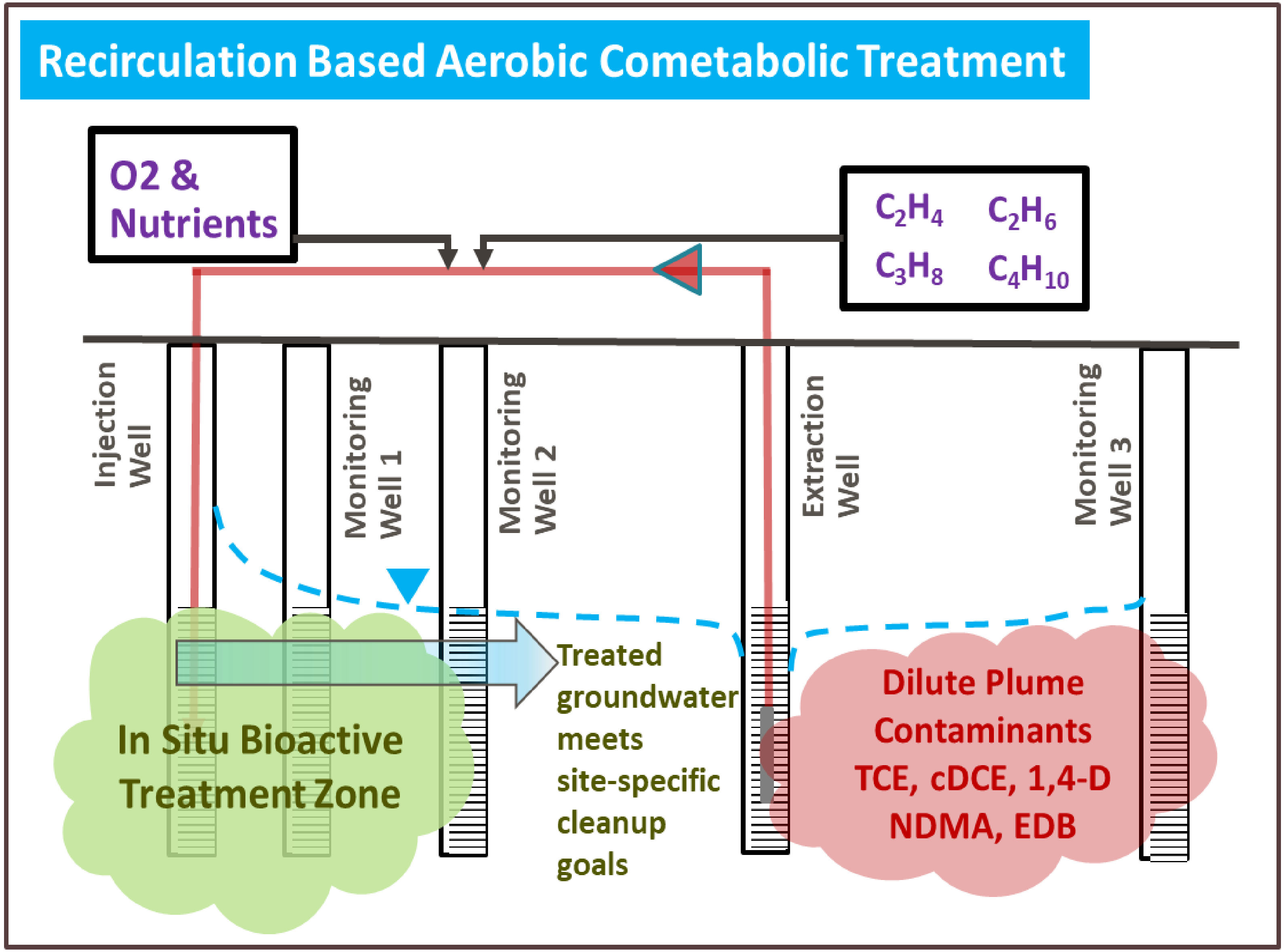
Example 2: HR-ISB for anaerobic metal precipitation

- Anaerobic precipitation of **molybdenum (Mo) and vanadium (V)** under low redox conditions
- Fast flow basalt aquifer (flow velocity > 100 ft/d)
- **Emulsified vegetable oil (EVO) recirculated** between well pairs for < 1 day; no recirculation afterwards
- Travel time in the bioactive zone **~ 8 hours**
- **~ 90% and ~ 95%** treatment efficiency for **Mo** and **V** in the bioactive zone **(Mo $t_{1/2}$= = 2.4 hrs. & V $t_{1/2}$= 1.8 hrs.)**
- Concurrent sulfate reducing activity **SO_4^{2-} $t_{1/2}$= = 2.4 hrs.**
- **No bioclogging** observed
- ~850 gallons of EVO lasts for 5-6 months

Recirculation only for substrate distribution



Example 3: HR-ISB for aerobic cometabolic biodegradation



McCarty et al. 1995

McCarty et al . 1998

Chu et al. 2018

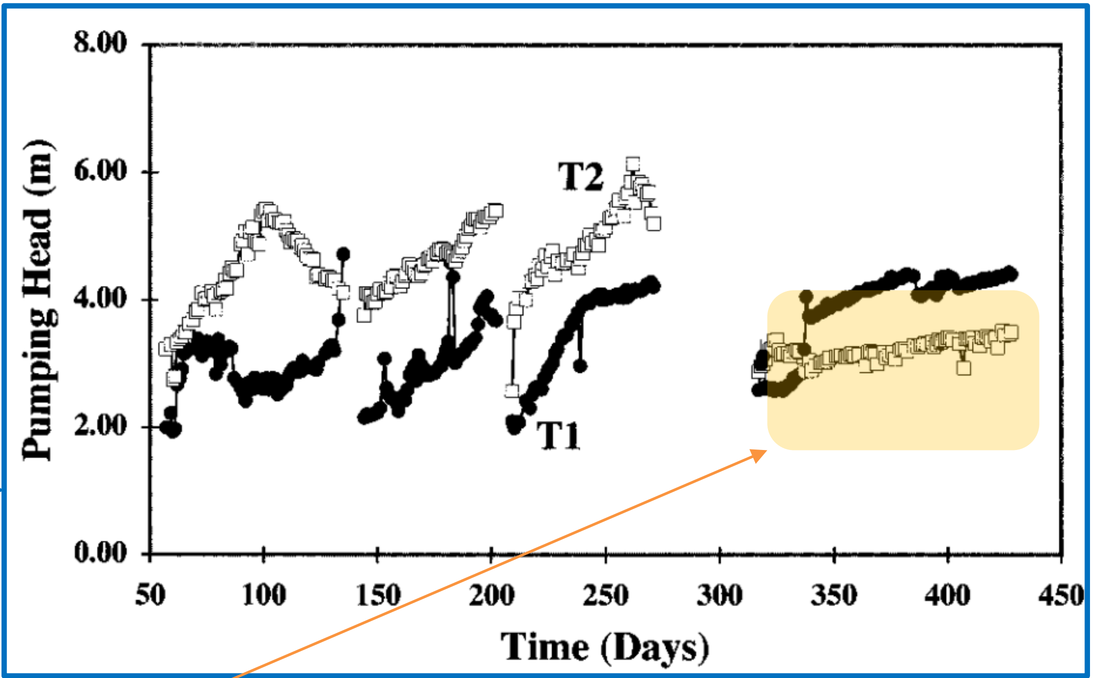
Hatzinger et al. 2018

Bioclogging

- More pronounced in an aerobic system
- Bioclogging hindering remediation performance

  (McCarty et al 1998)

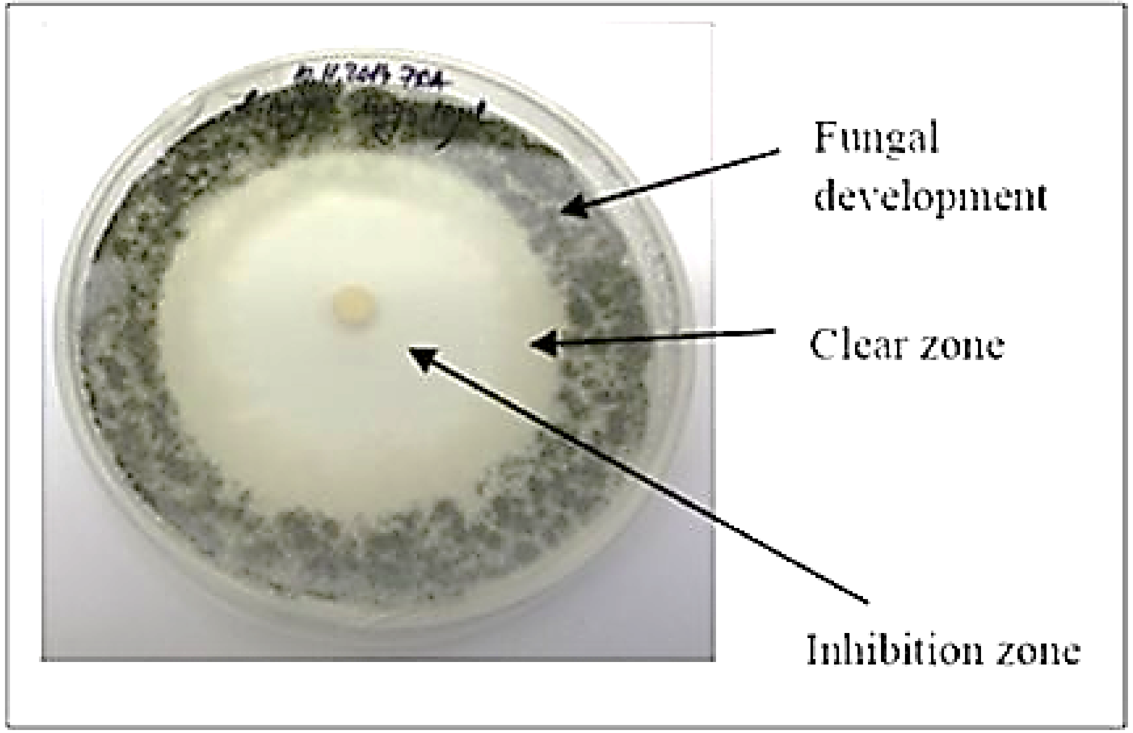
- Established bioclogging mitigation measures
  - Reducing primary substrate concentration to minimum needed to sustain remediation
  - Alternated substrate pulsing
  - General biocide addition (e.g., hydrogen peroxide)
  - Enzyme-specific chemical toxicity



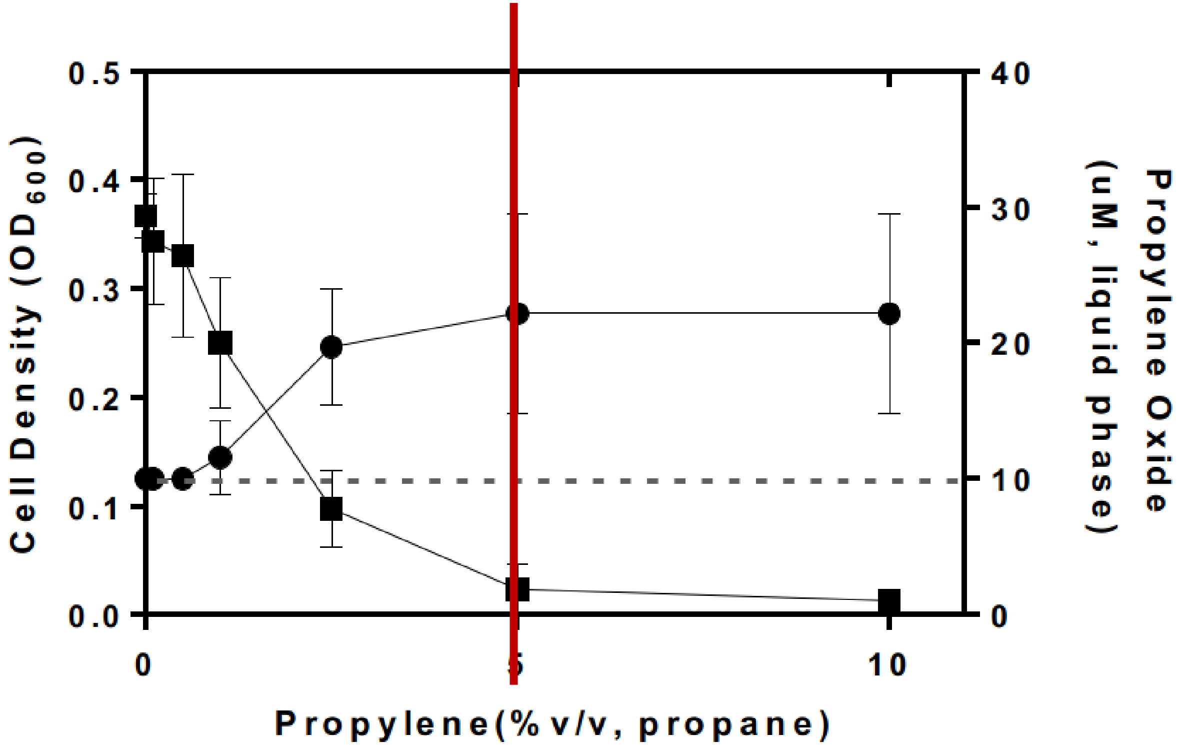
Inhibitor Type	Chemical	Mode of Action
General Oxidizing Biocide	Chlorine oxide	Oxidizing biomass
	Ozone	
	Hydrogen peroxide	
Oxidating Biocide Precursor	Propylene (precursor of propylene oxide, a general biocide)	Aerobic degradation of propylene produces propylene oxide, which is a general biocide
Inhibition to Hydrocarbons Uptake	Acetylene	Bonding to oxygenase or monooxygenase enzymes

Cox 2005 – A review of biofouling controls for enhance in situ bioremediation of groundwater

Chemical toxicity to mitigate bioclogging-beneficial



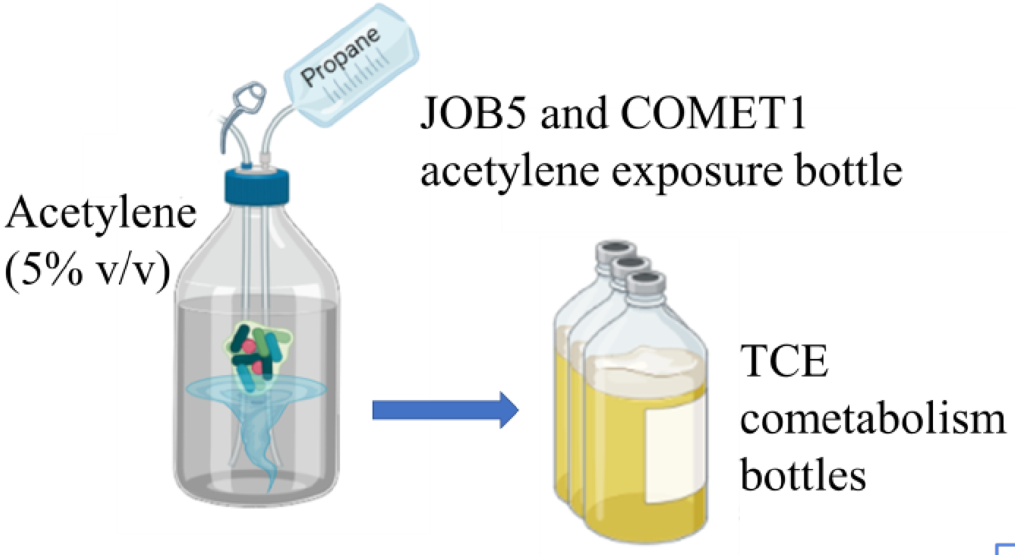
Mihai, A.L. and Popa, M.E., 2015. In vitro activity of natural antimicrobial compounds against Aspergillus strains. Agriculture and Agricultural Science Procedia, 6, pp.585-592.



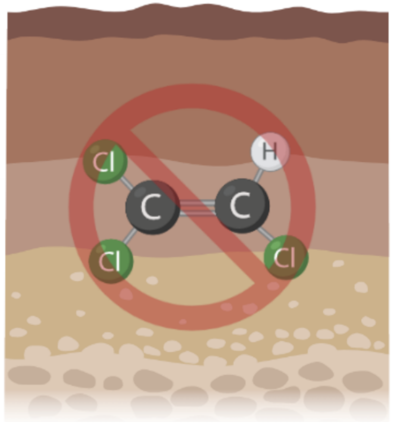
SALAS MEZA, LINDA. Effect of Propane Grade on Growth and Activity of Gaseous Hydrocarbon-Oxidizing Bacteria (Under the direction of Dr. Michael R. Hyman).

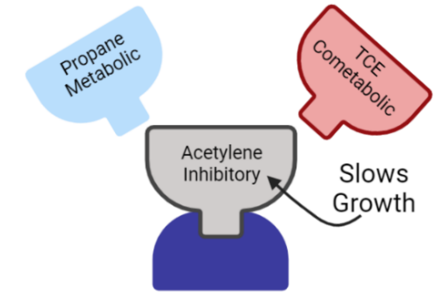
Understanding of contaminants effects on biostimulated/augmented culture is key for optimal performance.

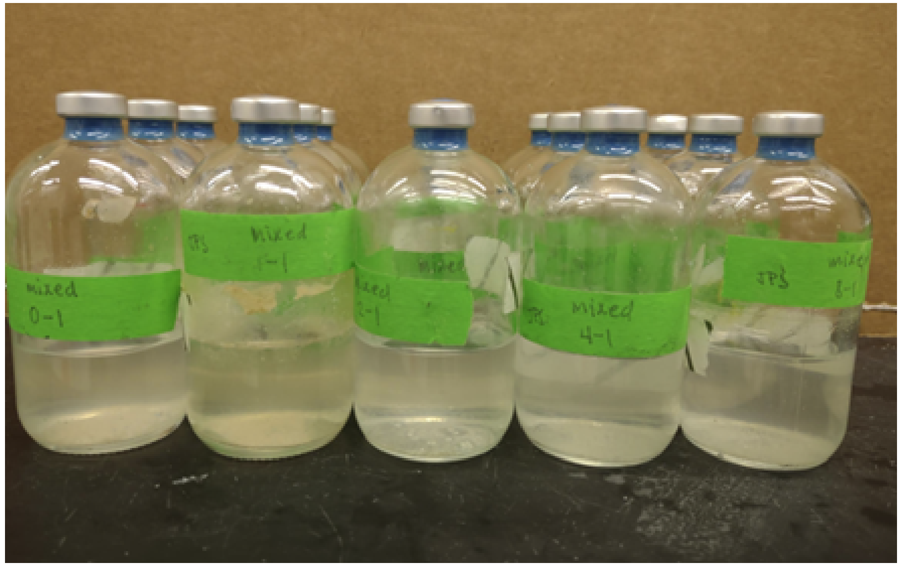
Enzyme-specific inhibitor - acetylene



Ultimate Objective:
Enhanced *In Situ*
Bioremediation of Dilute
TCE Plumes







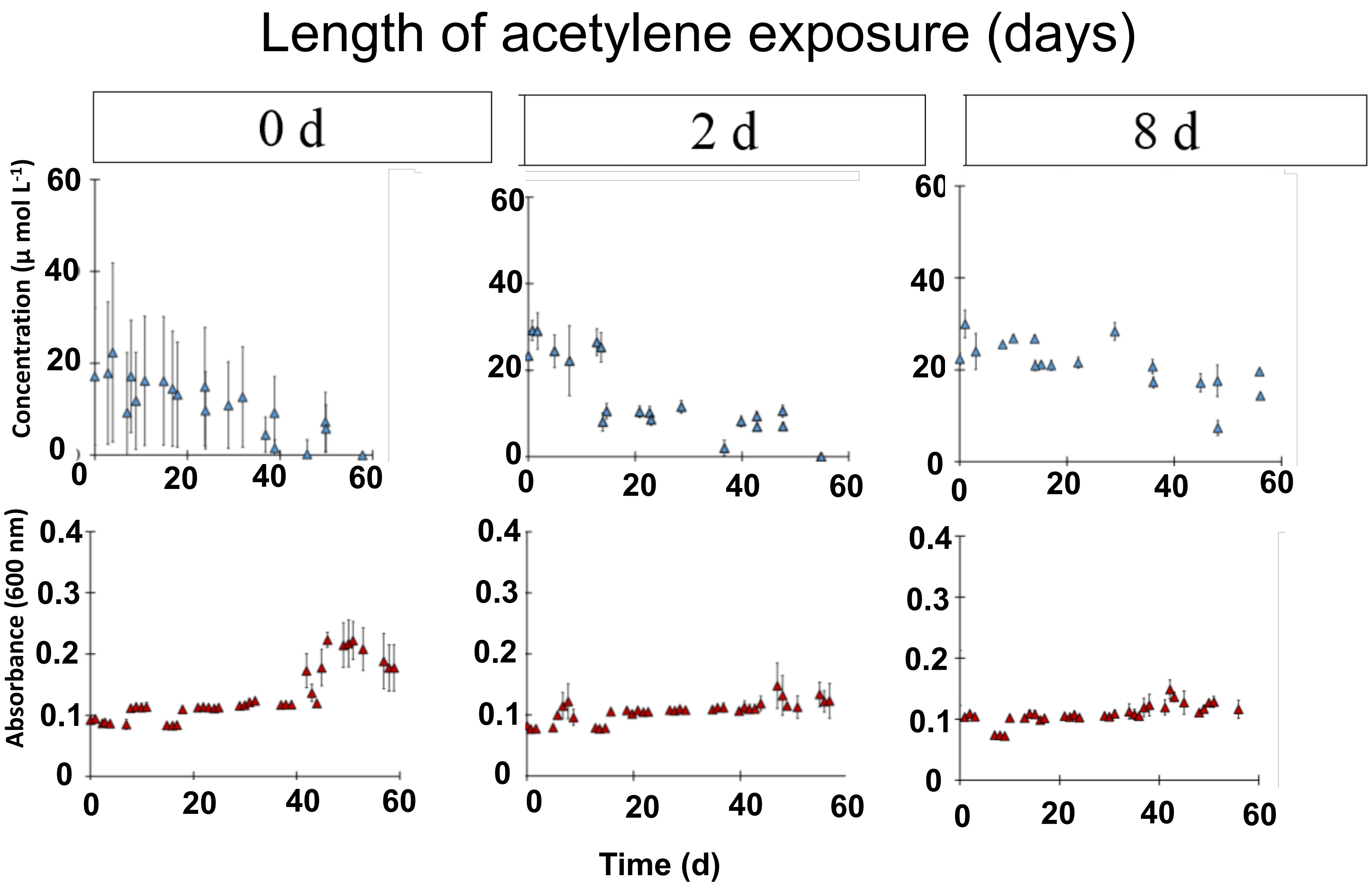
Acetylene exposure periods

0 days 1 day 2 days 4 days 8 days

Experimental setup of acetylene's effects on TCE cometabolism

COMET1 Experimental bottles

Acetylene’s effects on a TCE cometabolizing culture

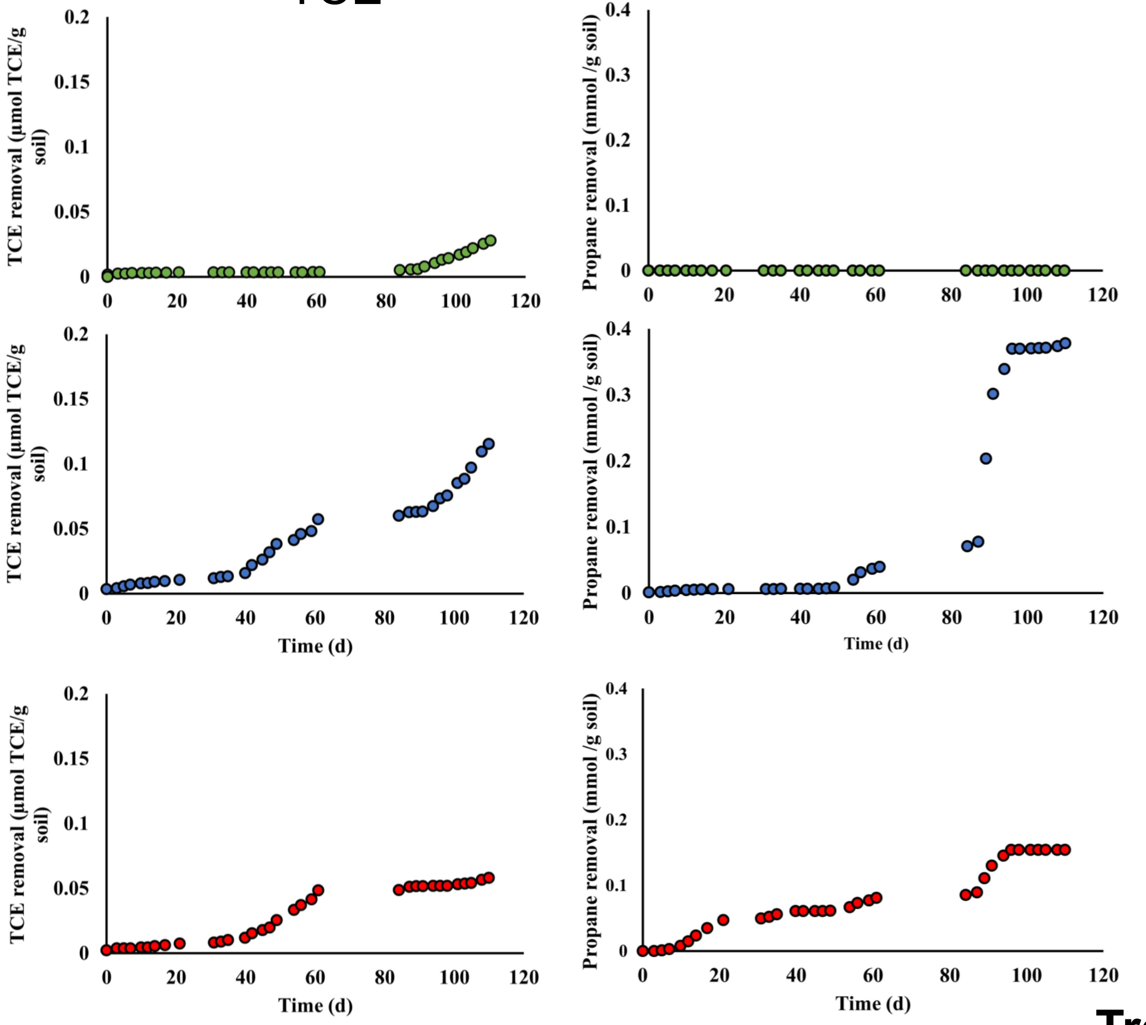


Longer acetylene exposures at 5% v/v cause progressive reductions in TCE degradation and biomass production rates.

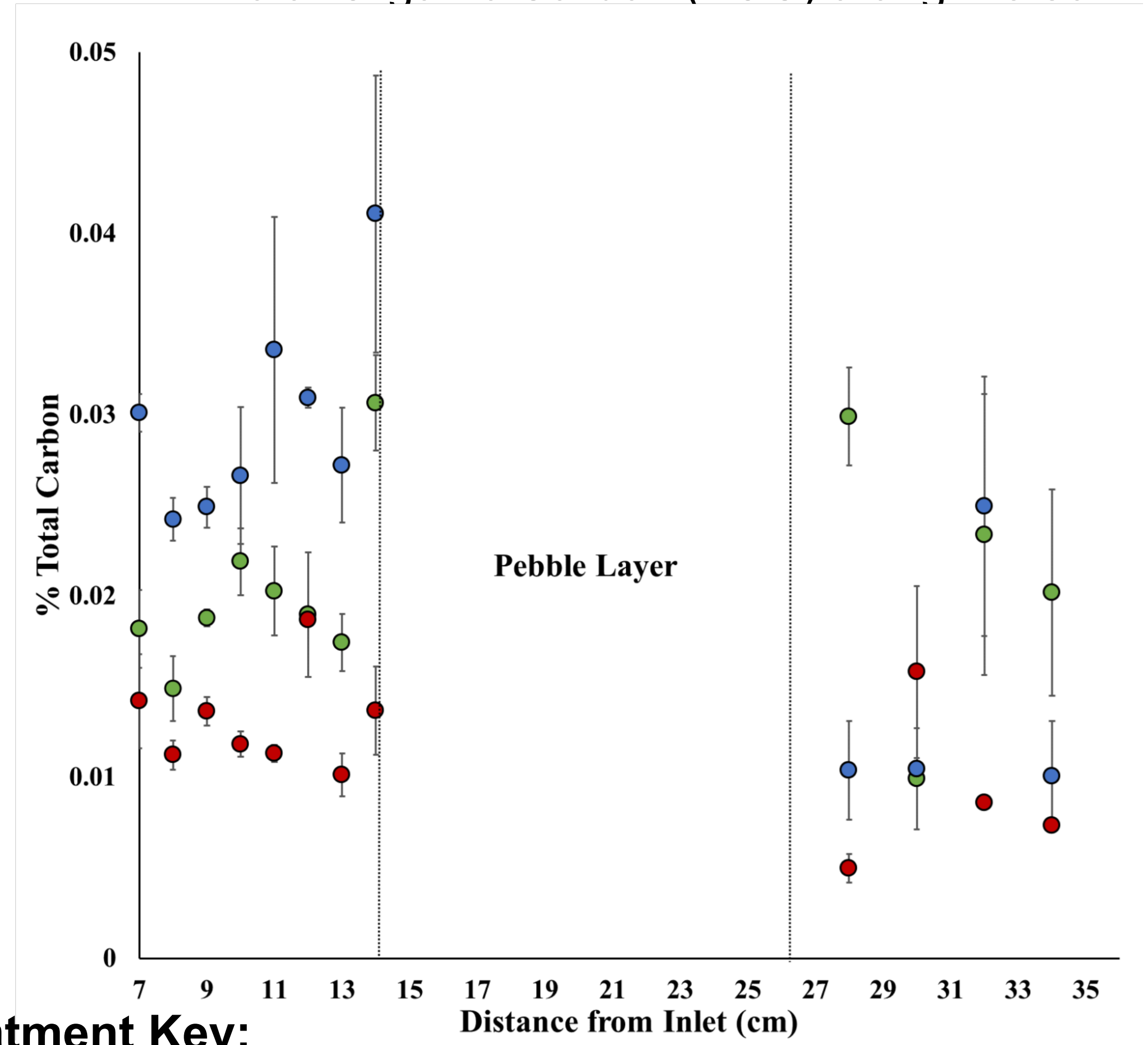
Acetylene's effects on TCE cometabolism soil columns

TCE

Propane



Total Organic Carbon (TOC) along the soil



Acetylene slows TCE and propane degradation

Treatment Key:

Green- Control

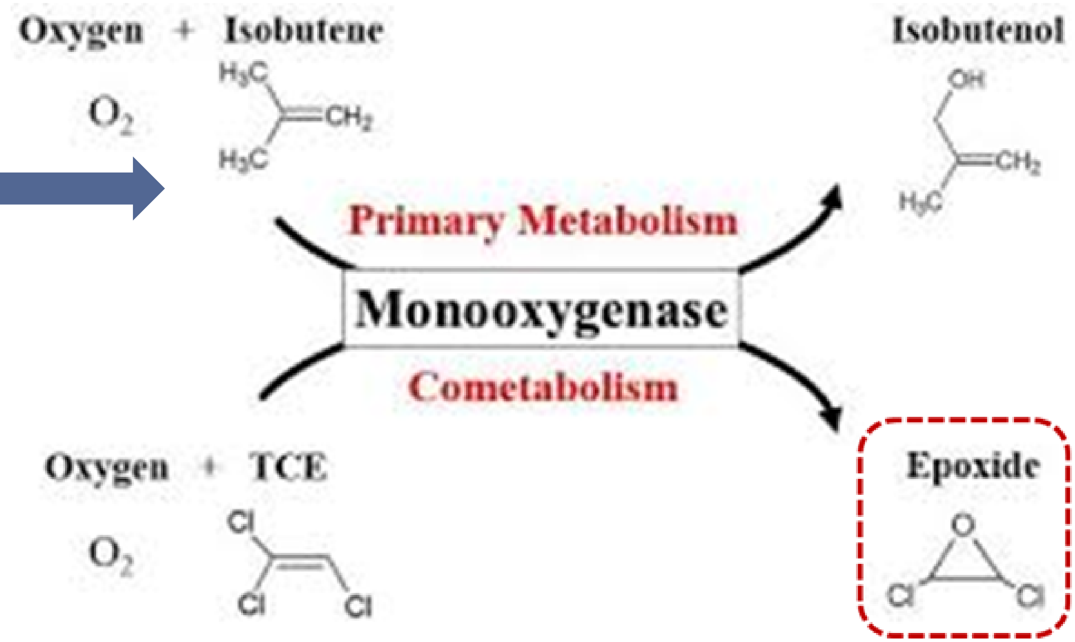
Blue- Propane

Red- Propane+ Acetylene

Acetylene exposed columns have the lowest TOC.

Chemical toxicity reducing treatment performance-detrimental

- Chloroethenes
- Very high toxic effects of 1,1,1-TCA & 1,1 DCE related epoxides



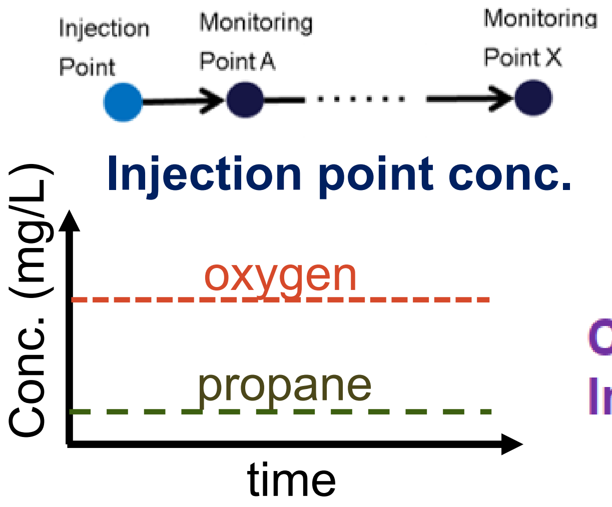
Microbial transformation capacities (T_c) for various contaminants

Contaminant	Culture	T_c (μmol Contaminant/ mg Cells)	Reference
1,1,1-TCA	Mixed culture CAC1	0.07	(H. L. Chang and Alvarez-Cohen 1996)
1,2-DCA		13	
TCE		1.2	
1,1-DCE		0.39	
cDCE		2.4	
tDCE		3.3	
VC		6.6	
1,1,1-TCA	Methylosinus trichosporium OB3b	0.05	
1,2-DCA		10	
TCE		2.0	
1,1-DCE		0.36	
cDCE		2.6	
tDCE		8.0	
VC		11	

Substrate delivery patterns (alternate substrate pulsing)

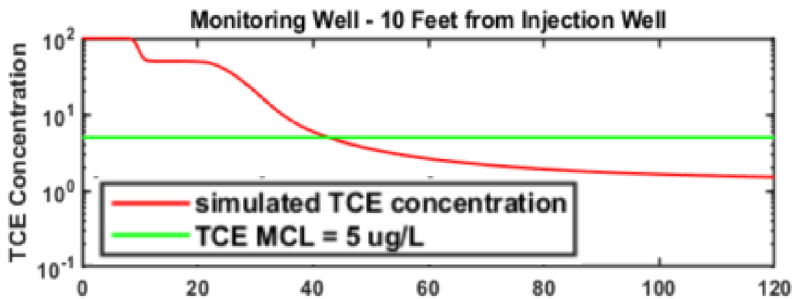
Key tuning parameter => 1. Primary loading rate & frequency

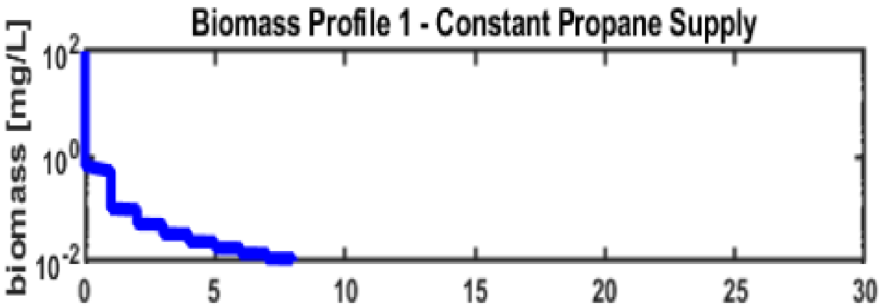
2. Oxygen loading rate ÷ primary substrate loading rate



120 day substrate addition

Constant Injection

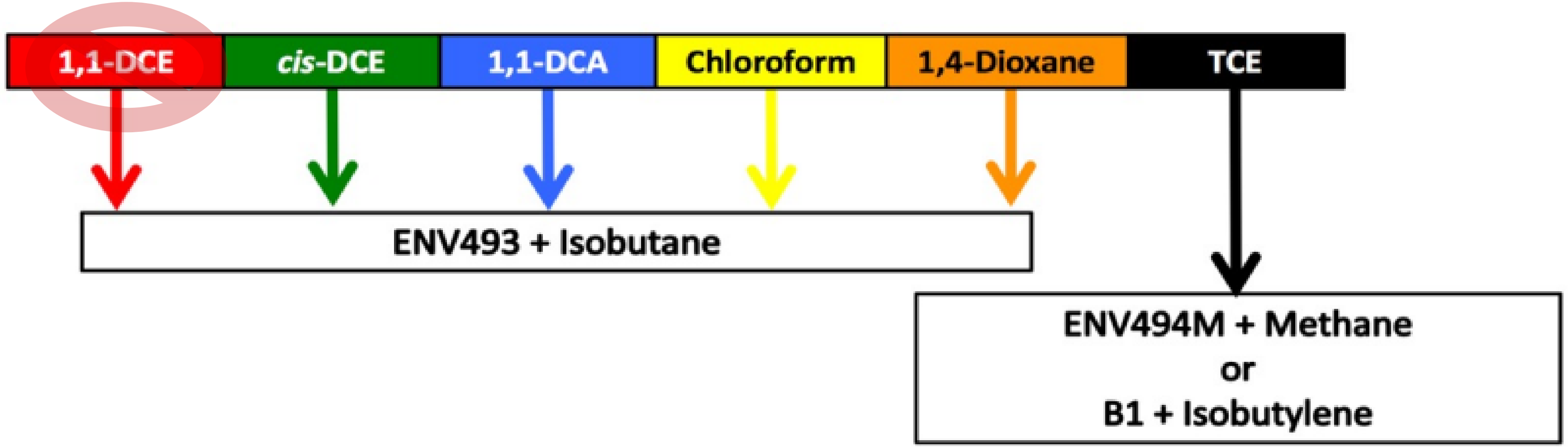




Substrate blends?

- Single substrate may not be sufficient to treat multiple contaminants concurrently

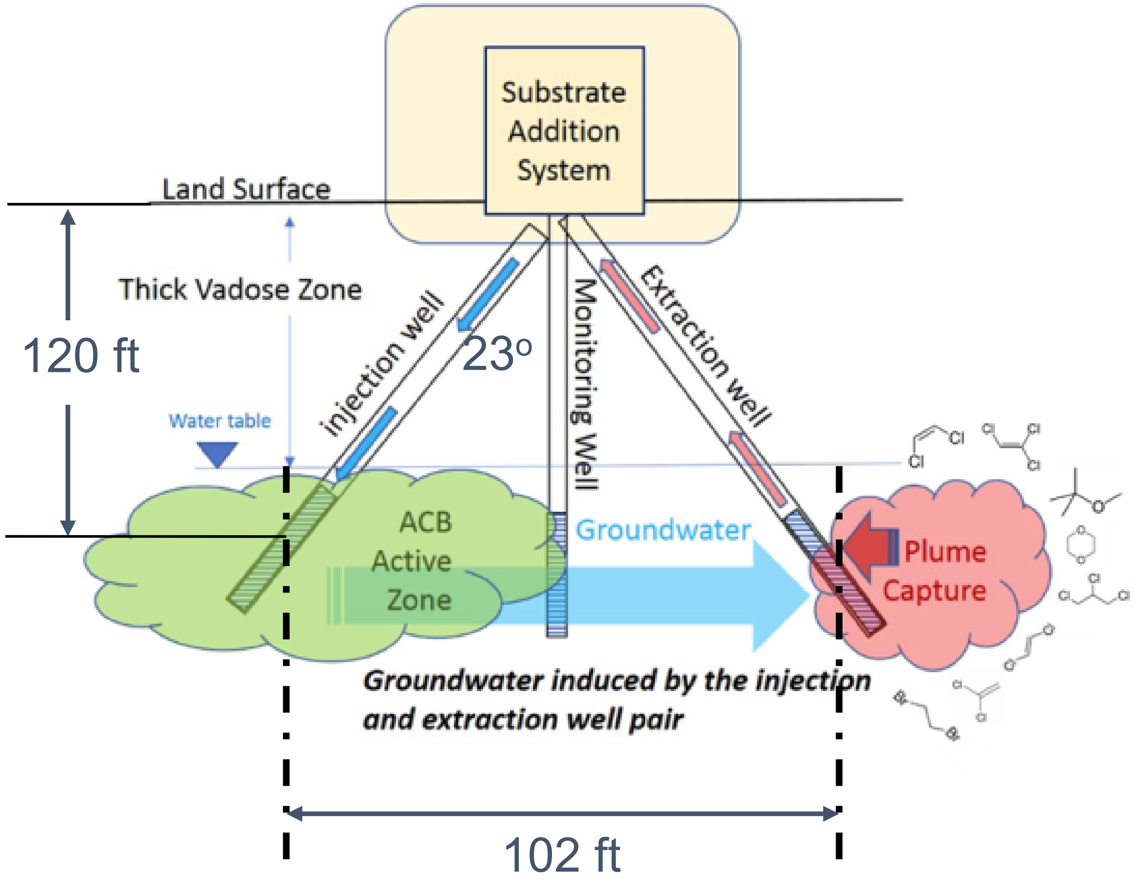
- Use multiple substrates may stimulate a broader spectrum of cometabolic activity



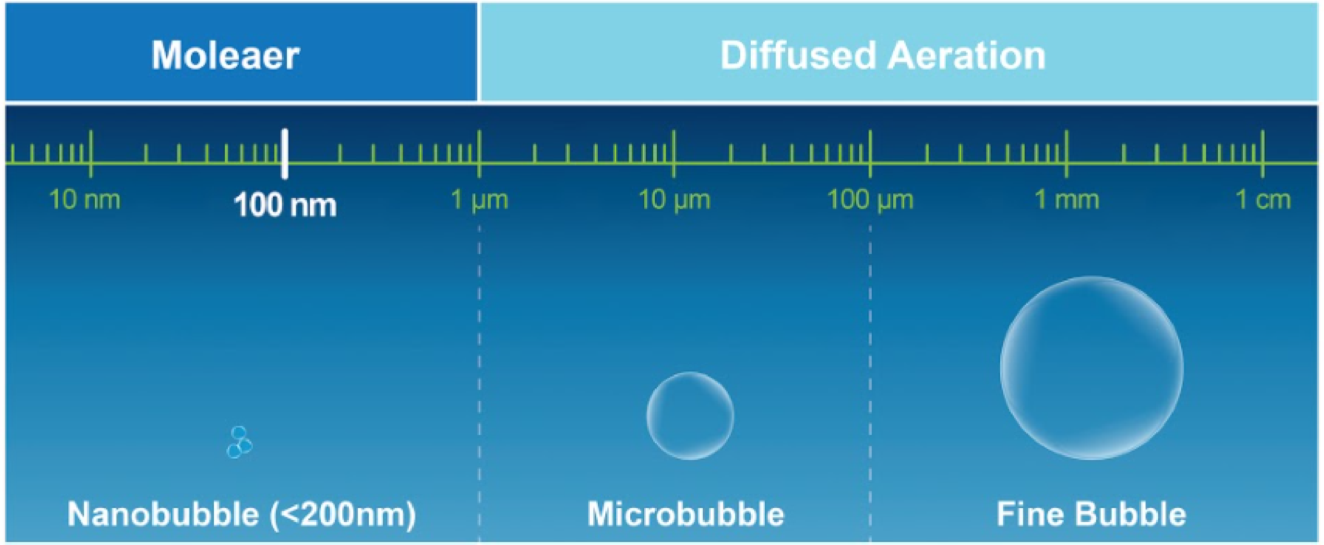
Tony Danko/ Hyman ER-201733

More efficient substrate distribution methods

Angled wells may reduce the system footprint and trenching for a recirculation treatment



Nano bubbles may help create a large bioactive zone due to its potential of long-term suspension in water



https://www.moleaer.com/nanobubbles

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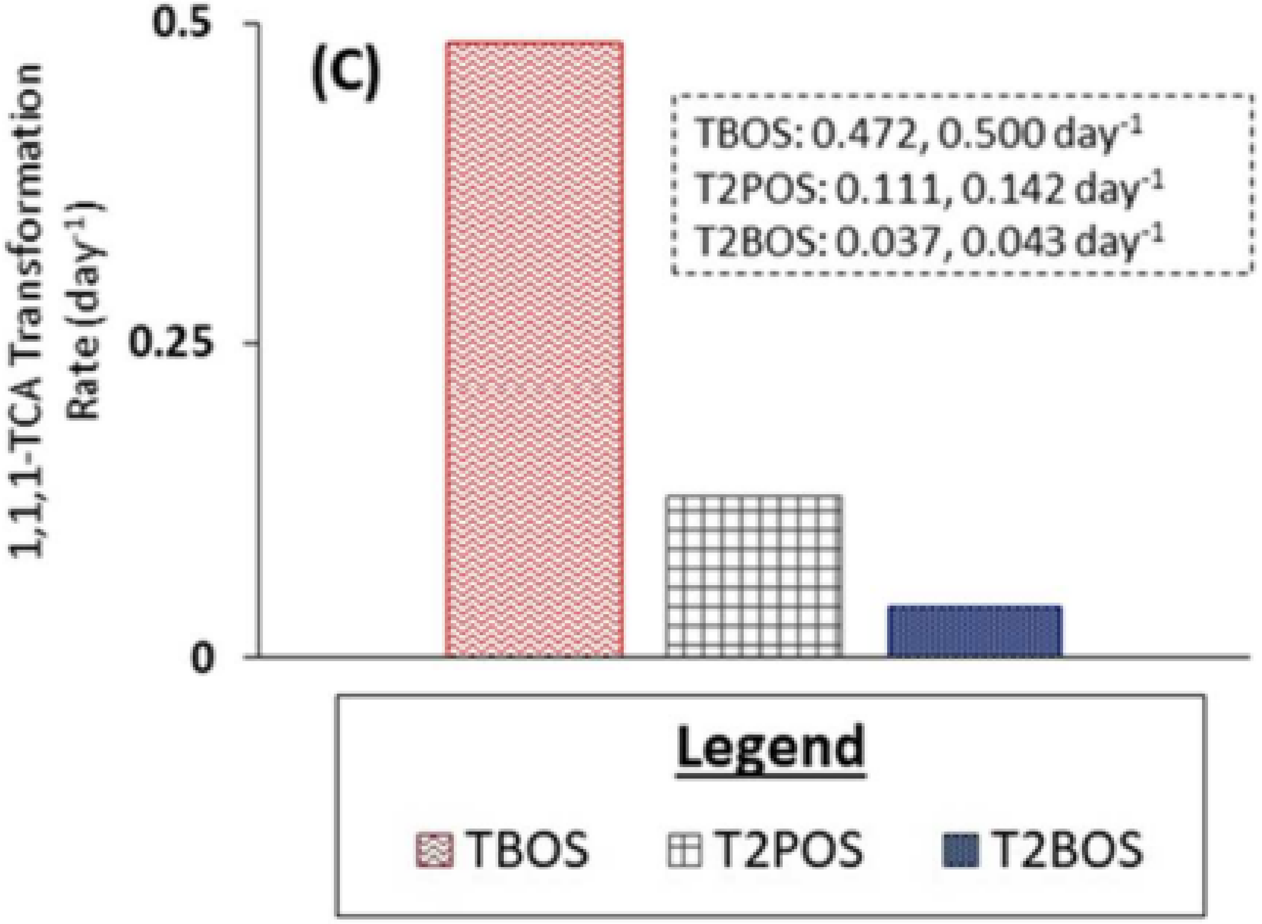
ADVANCING NANOBUBBLE TECHNOLOGY

Overcoming intrinsic limitation

- Some contaminants in dilute plumes are difficult to biodegraded
  - e.g., 1,1-DCE and 1,1,1-TCA
- *Rhodococcus rhodochrous* 21198 was found for 1,1,1-TCA treatment
- Find appropriate microbial strains for such recalcitrant contaminants
  - thrive along with the indigenous microbial community



Rapid 1,1,1 TCA degradation (Murnane et al. 2021)

ER2716 Semprini

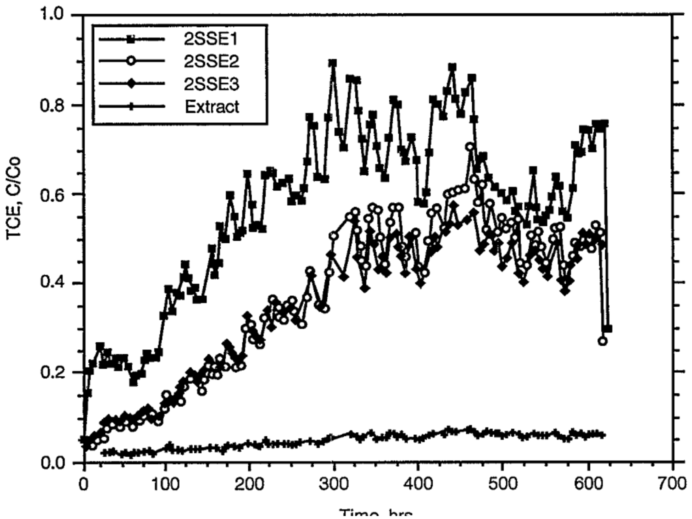
Long-term microbial community evolution

Problems

- Declining relative abundance of contaminant degraders
- Loss of contaminant degradation capacity

Diagnosis and response

- Aquifer material microcosms
- Transcriptomic/proteomic assays
- Contaminant degrading consortia with multiple contaminant degrading strains
- Substrate pulsing (feast and famine)
- Substrate blends



EPA ENVIRONMENTAL RESEARCH BRIEF

Bioaugmentation with *Burkholderia cepacia* $PR1_{301}$ for *In Situ* Bioremediation of Trichloroethylene Contaminated Groundwater

Perry L. McCarty[1], Gary D. Hopkins[1], Junko Munakata-Marr[1], V. Grace Matheson[2], Mark E. Dolan[1], Louise B. Dion[1], Malcolm Shields[3], Larry J. Forney[2], and James M. Tiedje[2]

Groundwater Monitoring & Remediation — NGWA

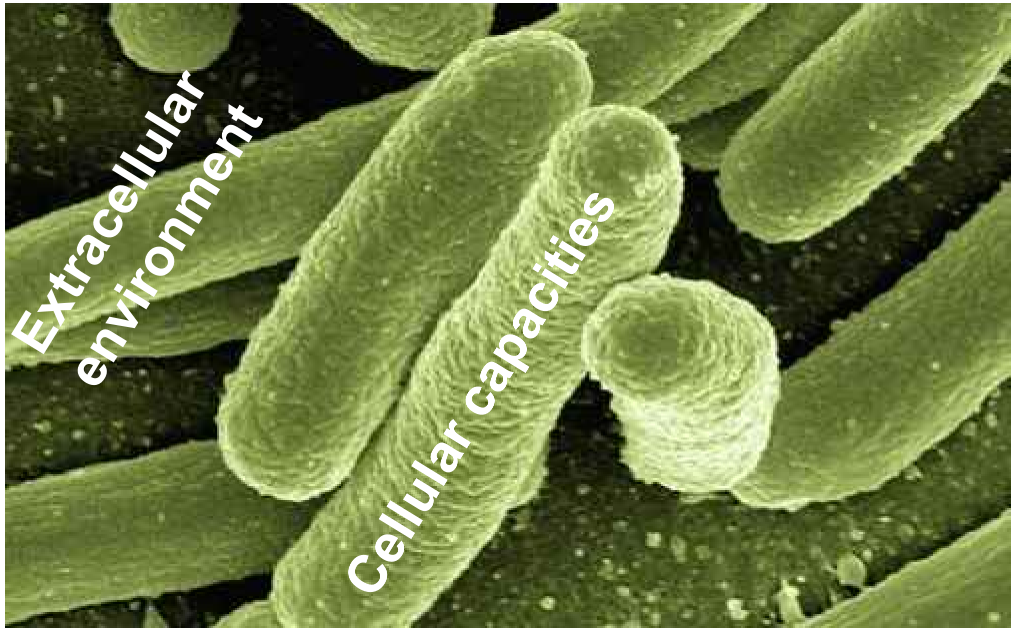
Cutting-Edge Technical Paper | Open Access

Using qPCR Assays to Predict Rates of Cometabolism of TCE in Aerobic Groundwater

by John T. Wilson, James C. Mills IV, Barbara H. Wilson, Mark L. Ferrey, David L. Freedman, Dora Taggart

Results/Lessons Learned

- Overcoming extrinsic limitations
  - More efficient substrate distribution methods
  - Substrate delivery patterns
  - Substrate blends
  - Chemical inhibitors



- Overcoming intrinsic limitations
  - Recalcitrant/toxic contaminant specific strains
  - Guarding strains from toxicity via precise slow release of substrate/bacteria from encapsulation.

Don’t add more substrate than needed.

Enabling in situ bioremediation of large, deep groundwater plumes

Thank you.

Questions and discussion

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