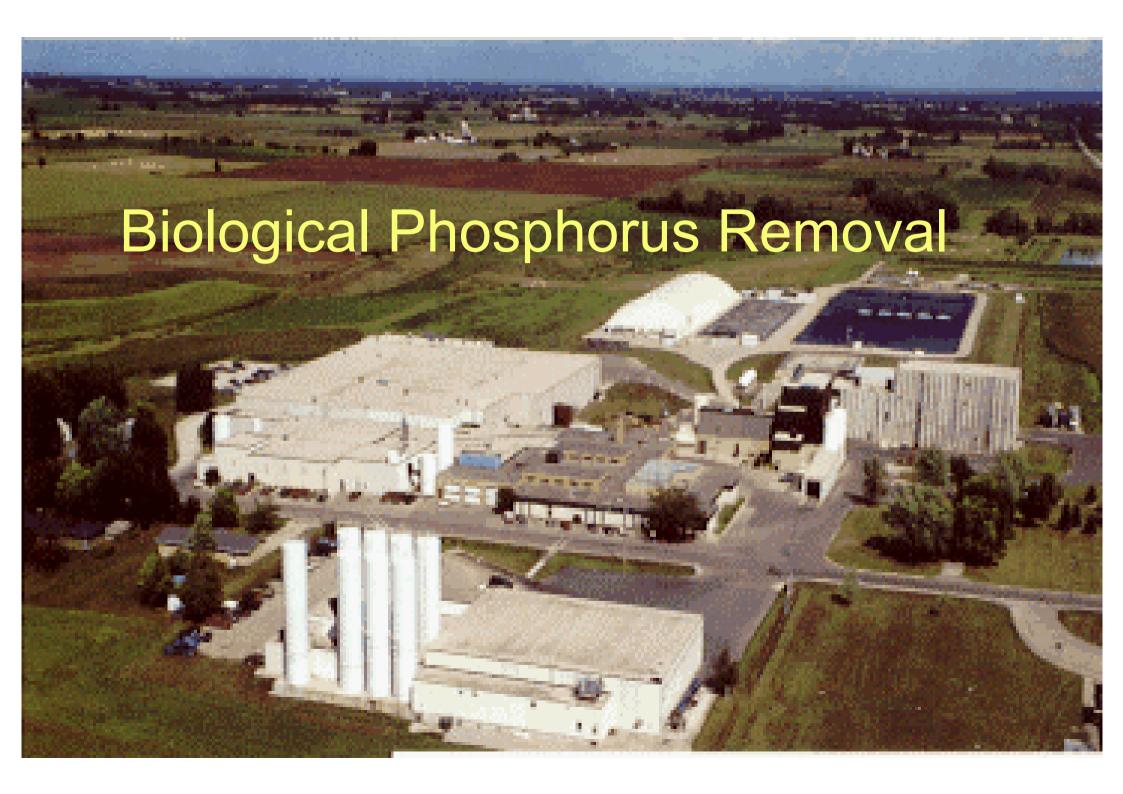
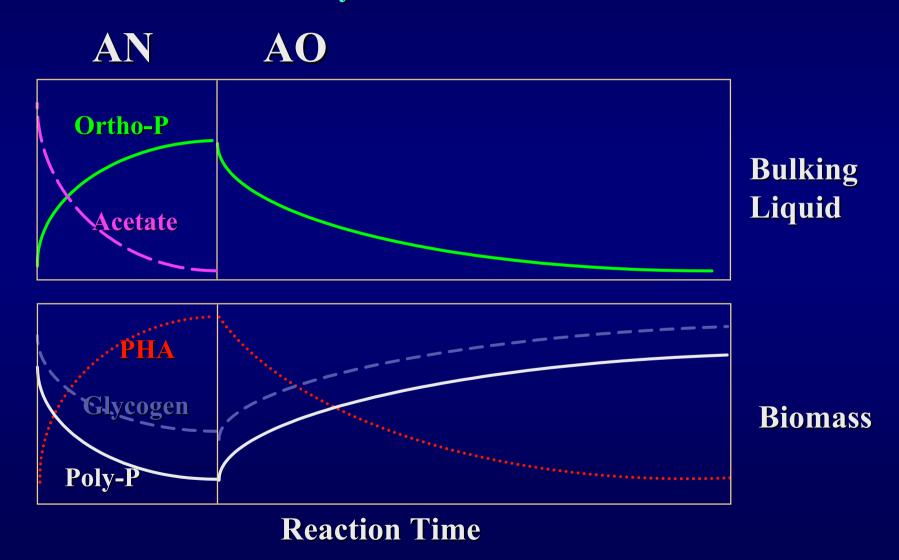
Research Activities

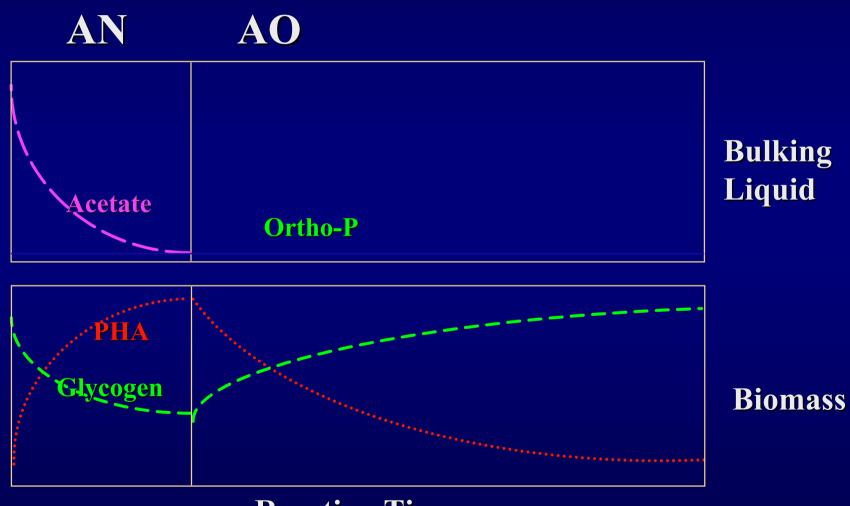
Jim K. Park
University of Wisconsin-Madison
Department of Civil & Environmental Engineering



Observations from BPR Systems

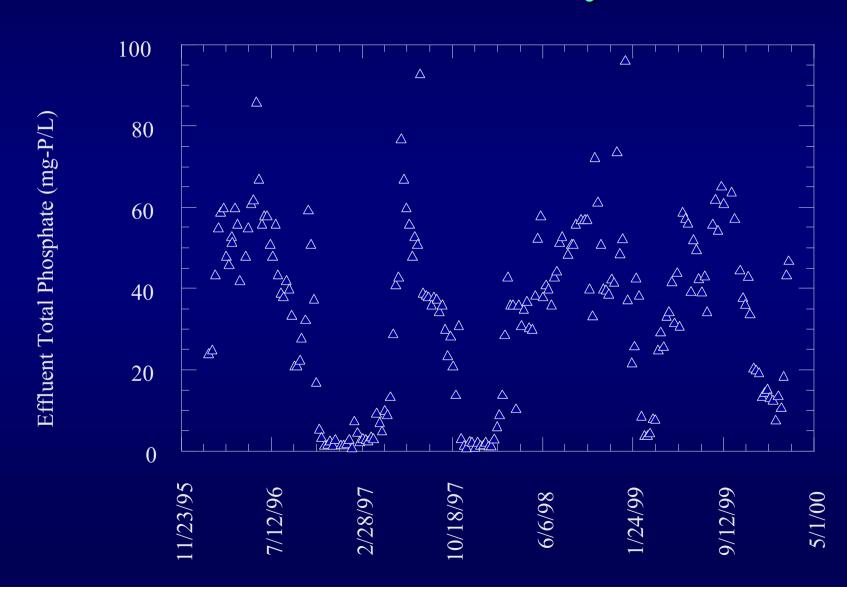


GAOs Metabolisms

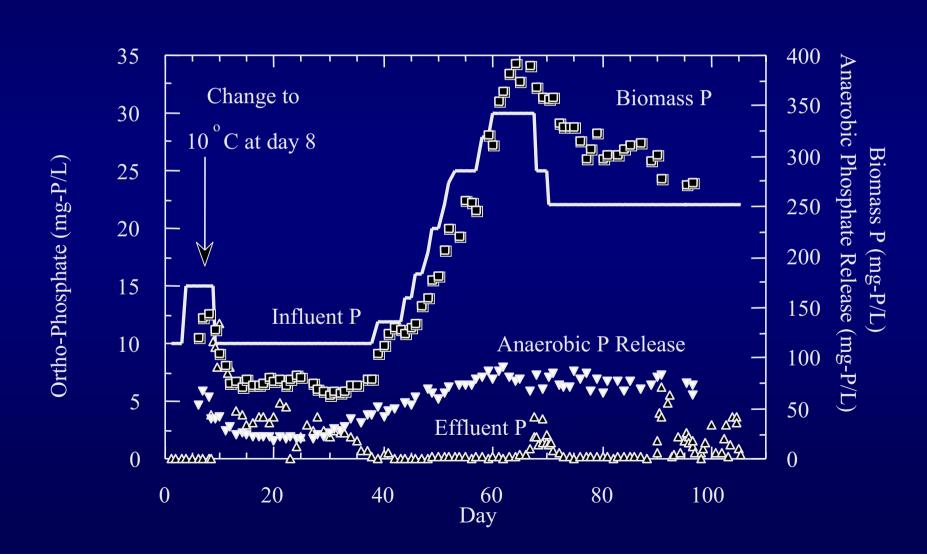


Reaction Time

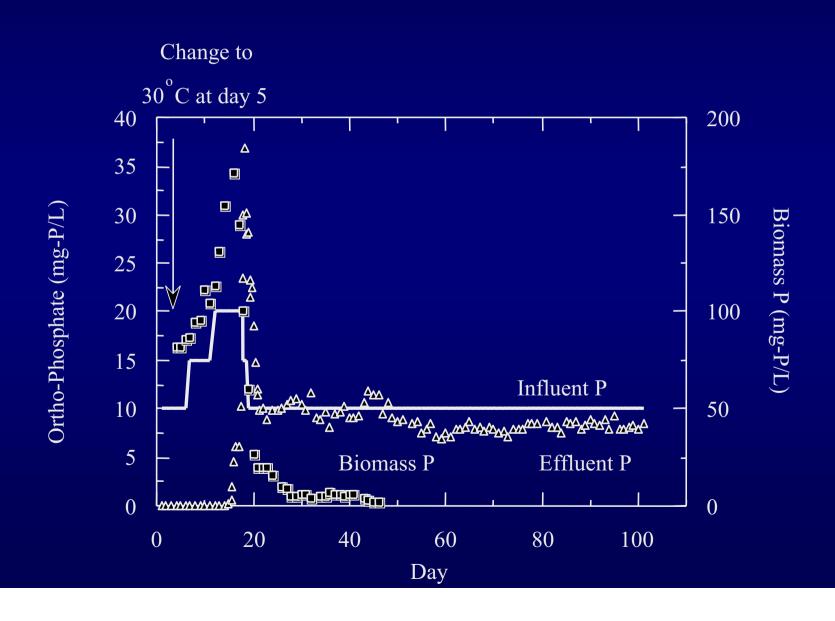
Problem in Full-Scale BPR System

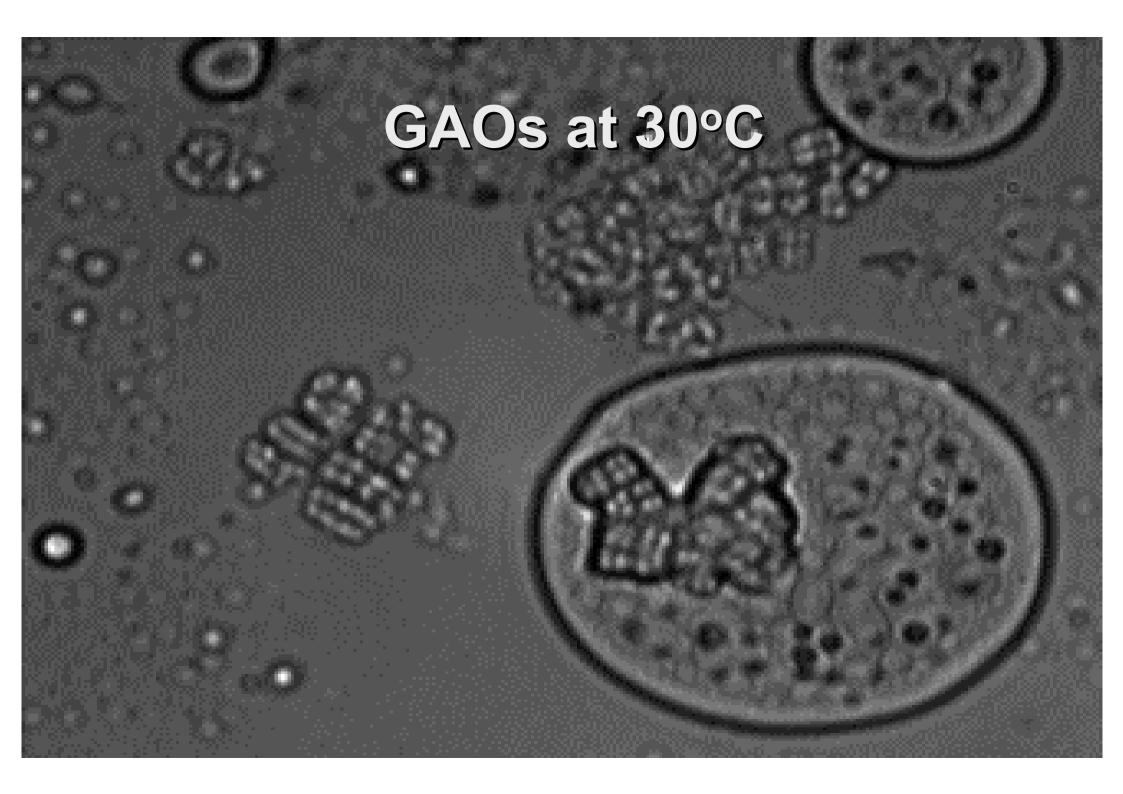


Daily Data of Reactor at 10°C and SRT 10 days

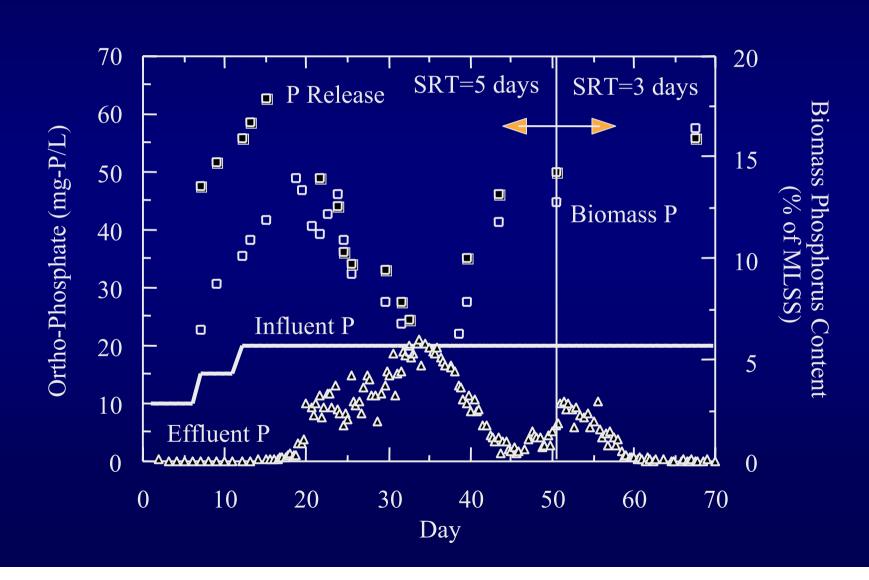


Daily Data of Reactor at 30°C and SRT 10 days

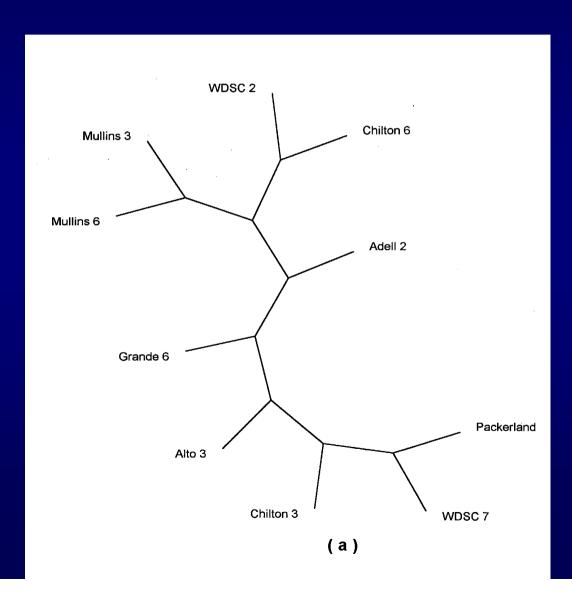




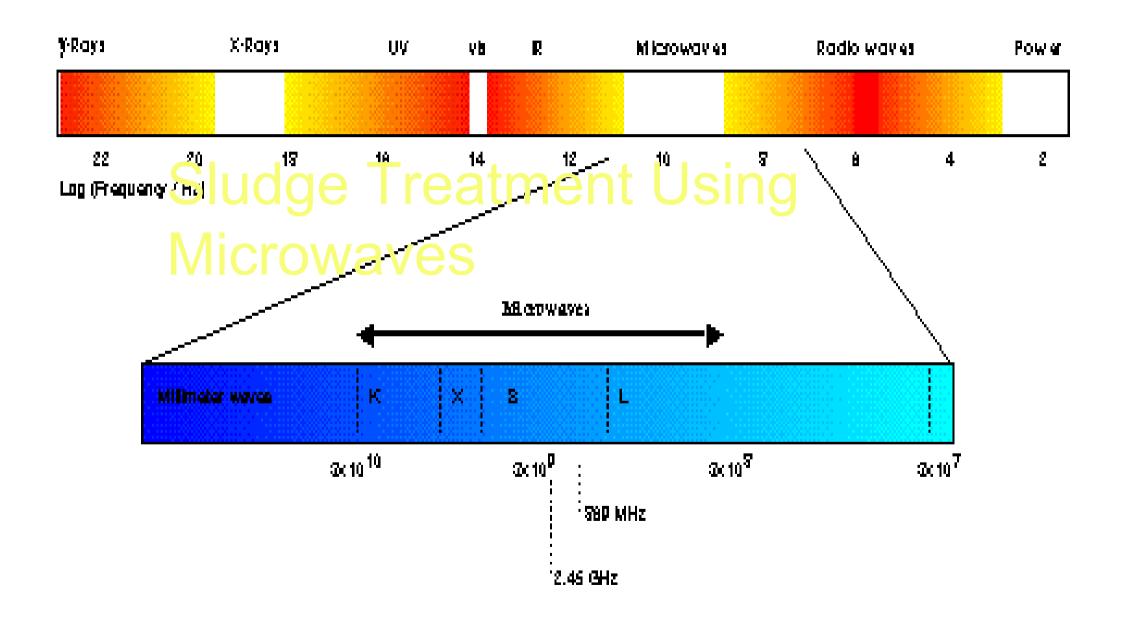
Effect of Sludge Age on GAO Growth



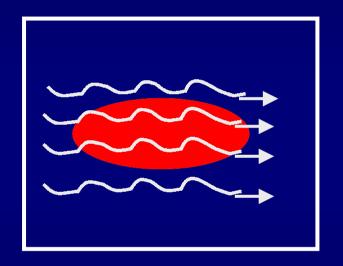
Terminal Restriction Fragment Length Polymorphism (t-RFLP) Analysis



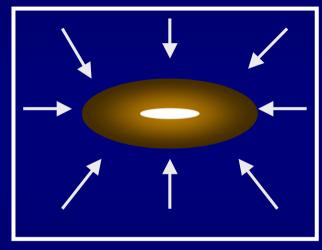
Unrooted tree of dairy wastewater sludge samples based on the presence-absence of each restriction fragment digested by *Alu*I



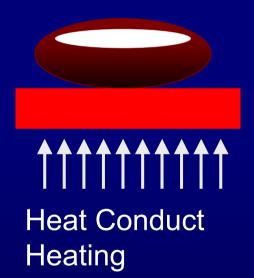
Heating Types



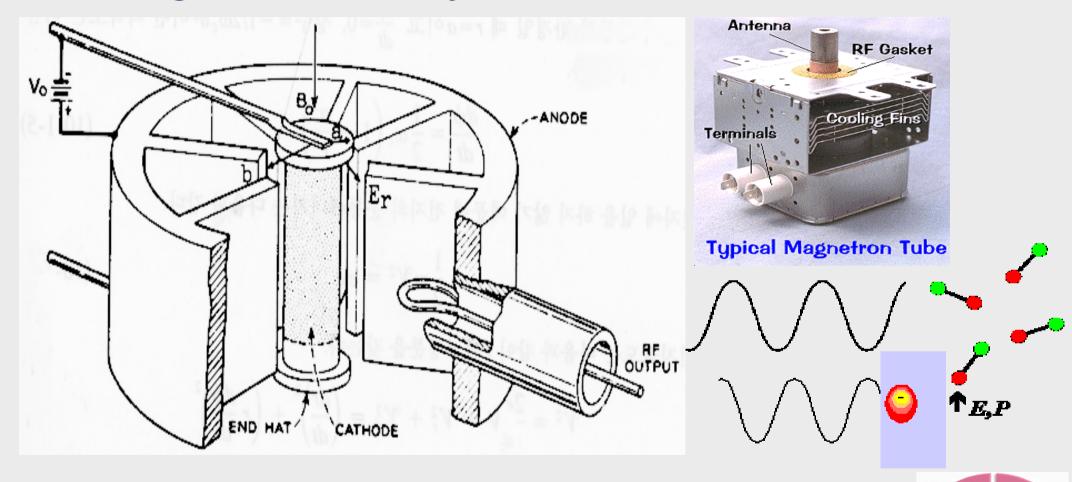
Electromagnetic Radiation Heating



Heat Radiation Heating



Magnetron Layout



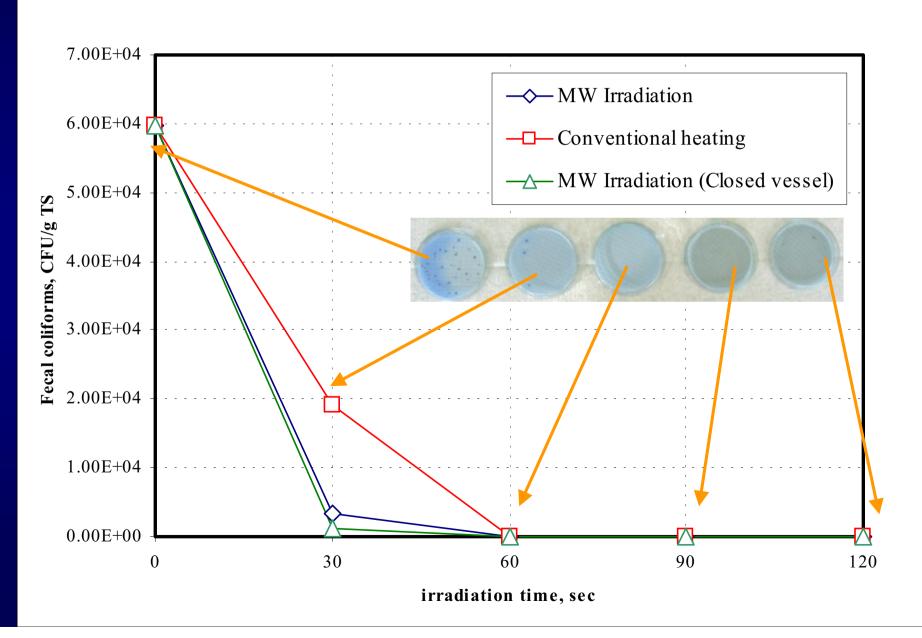
http://www.ed.ac.uk/~ah05/microwave.html



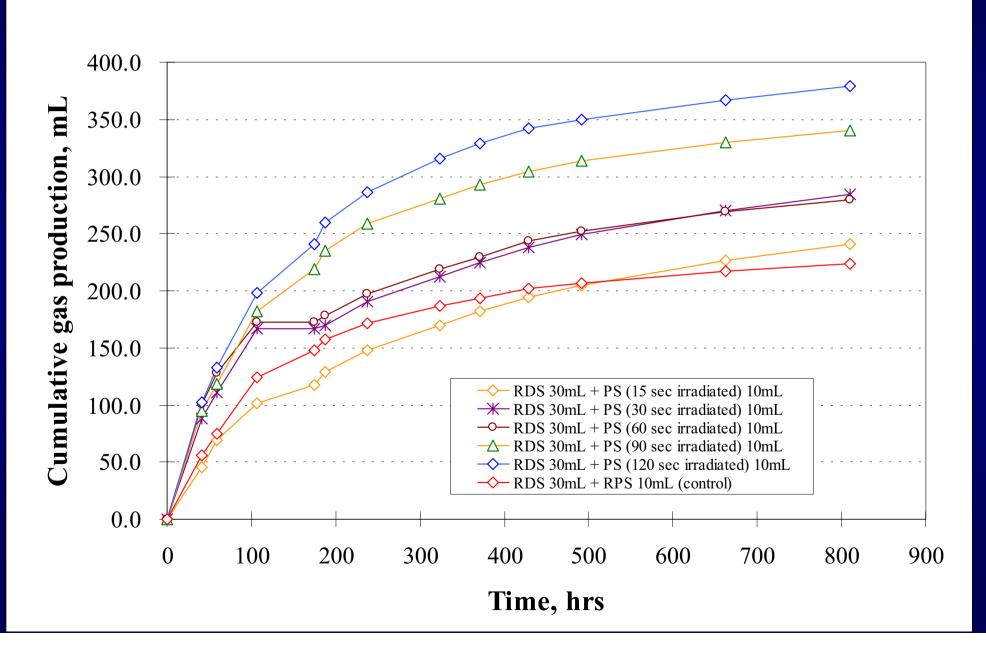


- Conveyor belt: 0~50 ft/min
- Weight: 1,180 kg
- Heating power: 12 kW, 2.45 GHz
- **•** \$15,000~45,000

Fecal Coliorms of ADS with MW irradiation and Conventional heating



Cumulative Gas Production of Primary Sludge Irradiated with Microwaves



Economic Analysis Microwave System vs. TPAD

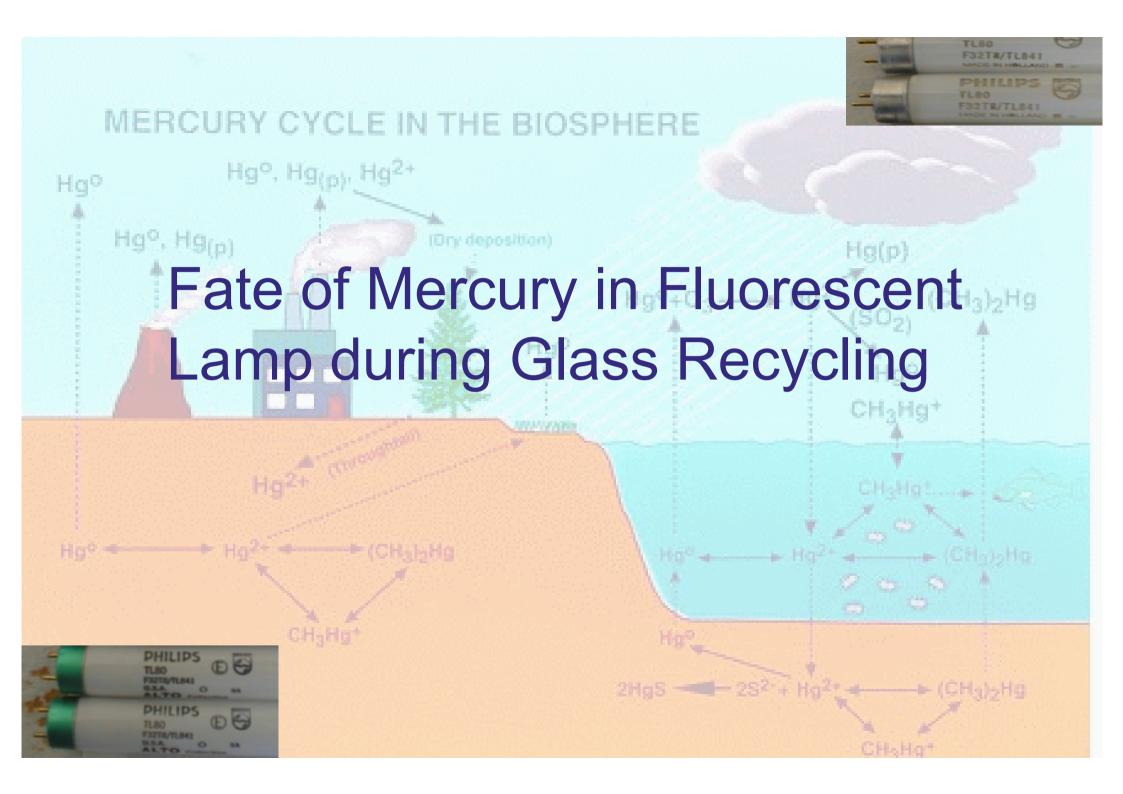
Post-pasteurization

	Proposed	Microwave system		
	100%	25%	50%	100%
Total cost	\$12,174,000	\$1,165,000	\$2,694,000	\$4,752,000
System	-	240 kW × 2 ea	240 kW × 4 ea	240 kW × 8 ea

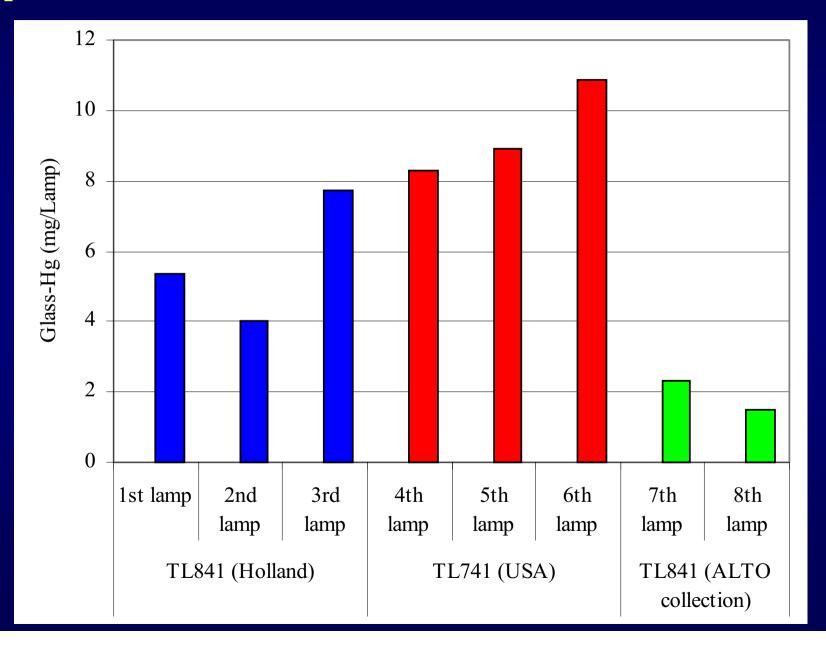
Pre-pasteurization

	Proposed	Microwave system		
	100%	25%	50%	100%
Total cost	\$12,174,000	\$2,207,000	\$3,779,000	\$6,922,000
System	-	240 kW × 3 ea	240 kW × 6 ea	240 kW × 12 ea

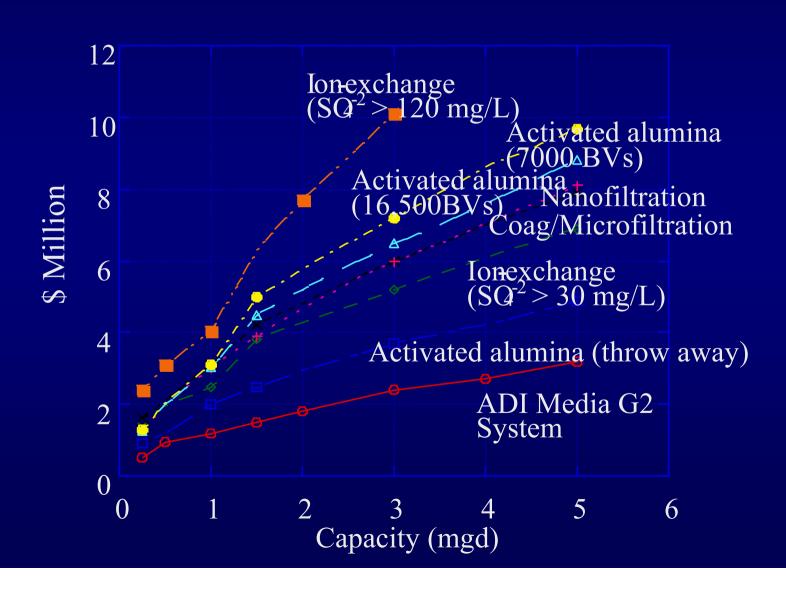
240 kW MW System: \$150,000; Total flow: 432,000 gpd



Experiments – Results



Removal of Arsenic Using Mesoporus Sorption Media



High Level Arsenic in Drinking Water Can Cause Blackfoot Disease

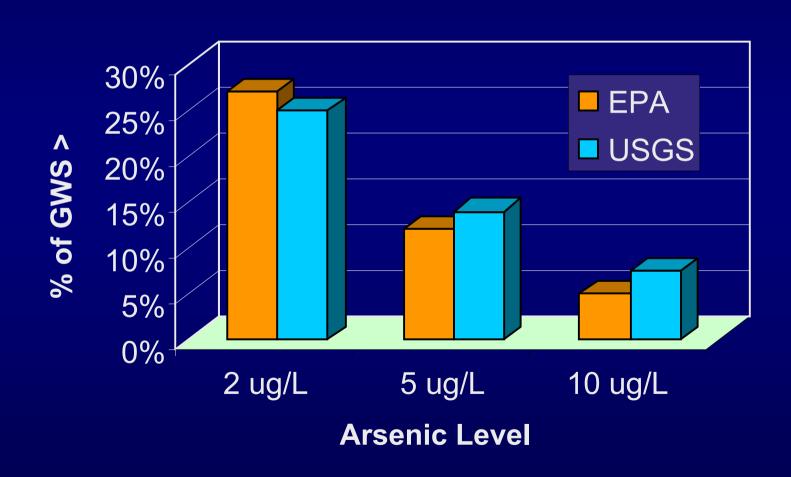




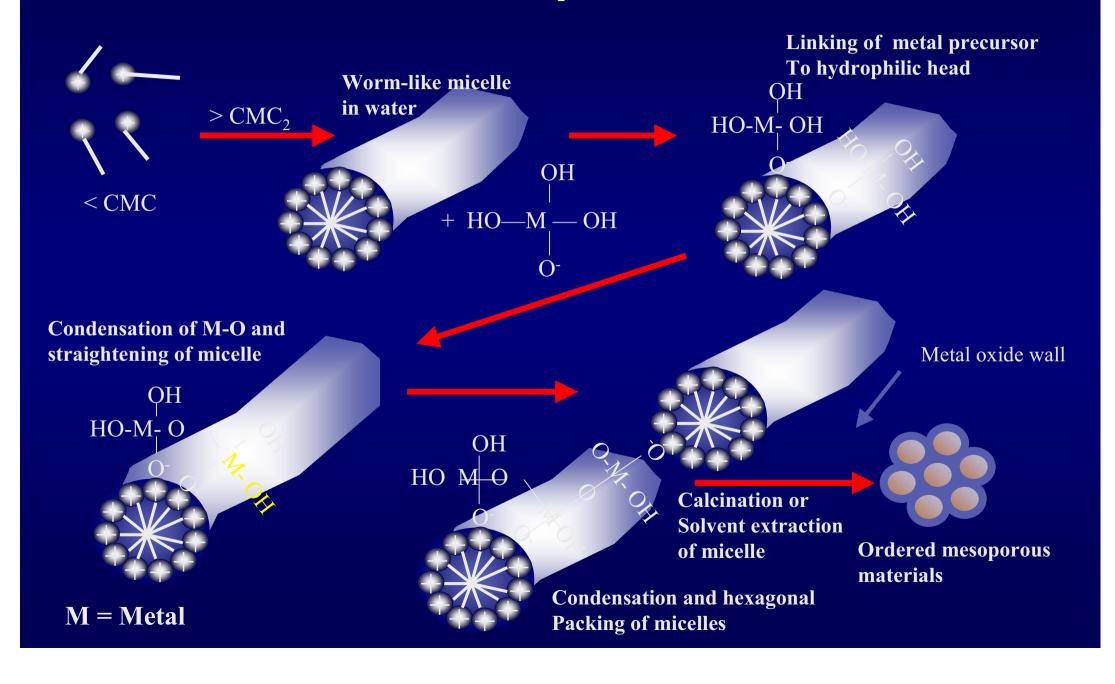


Drinking Water Arsenic Can Cause Skin Cancer

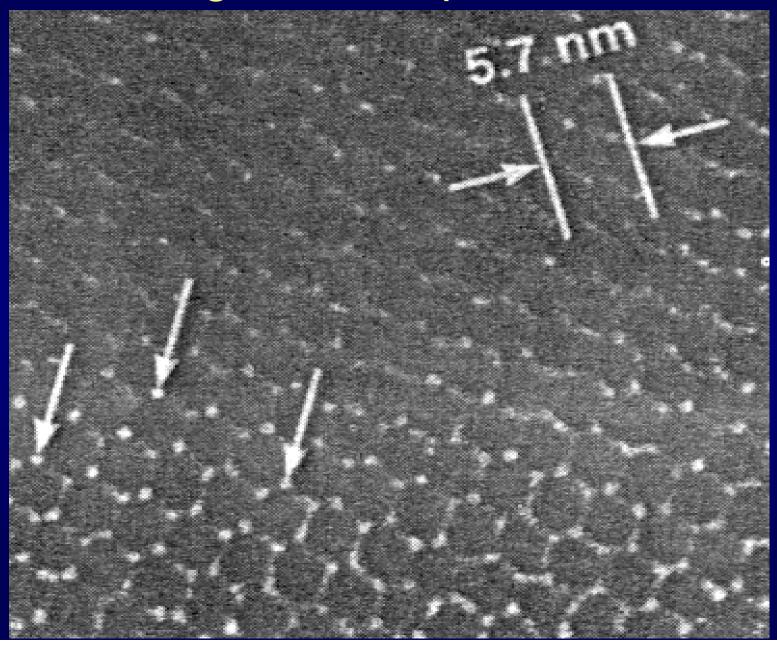
Estimates for Arsenic in U.S. Groundwaters

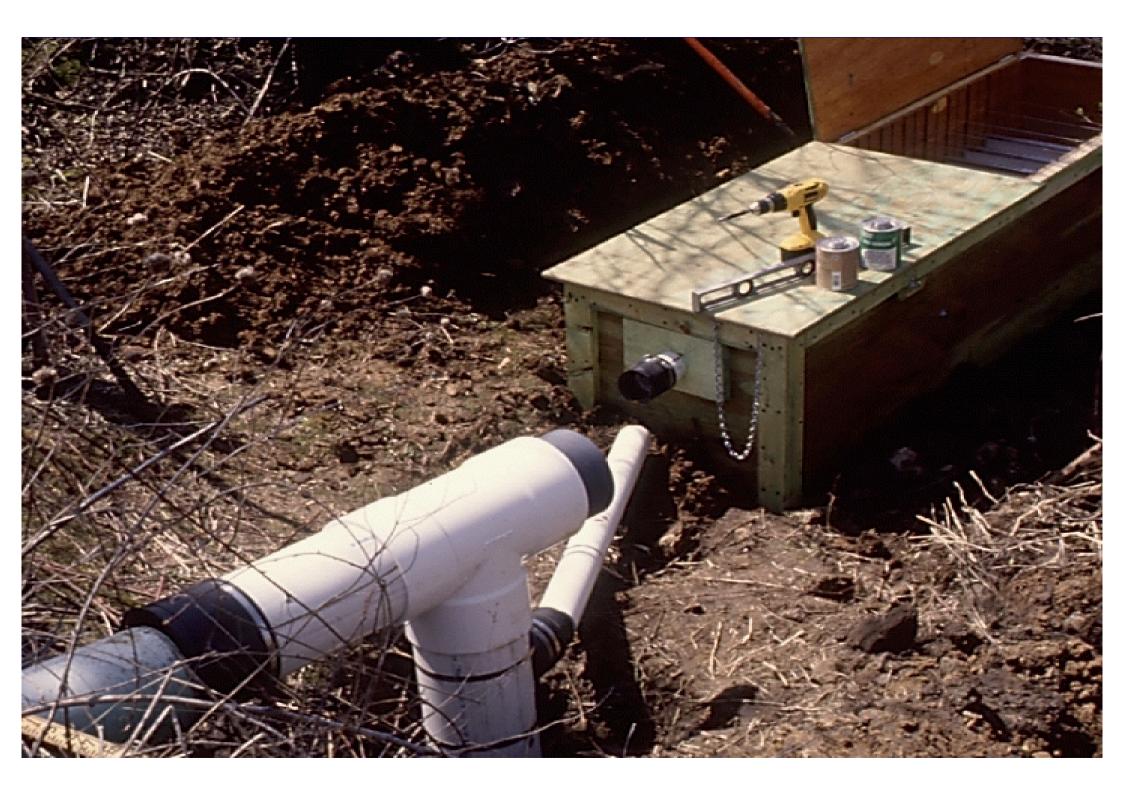


Schematics of Mesoporous Materials

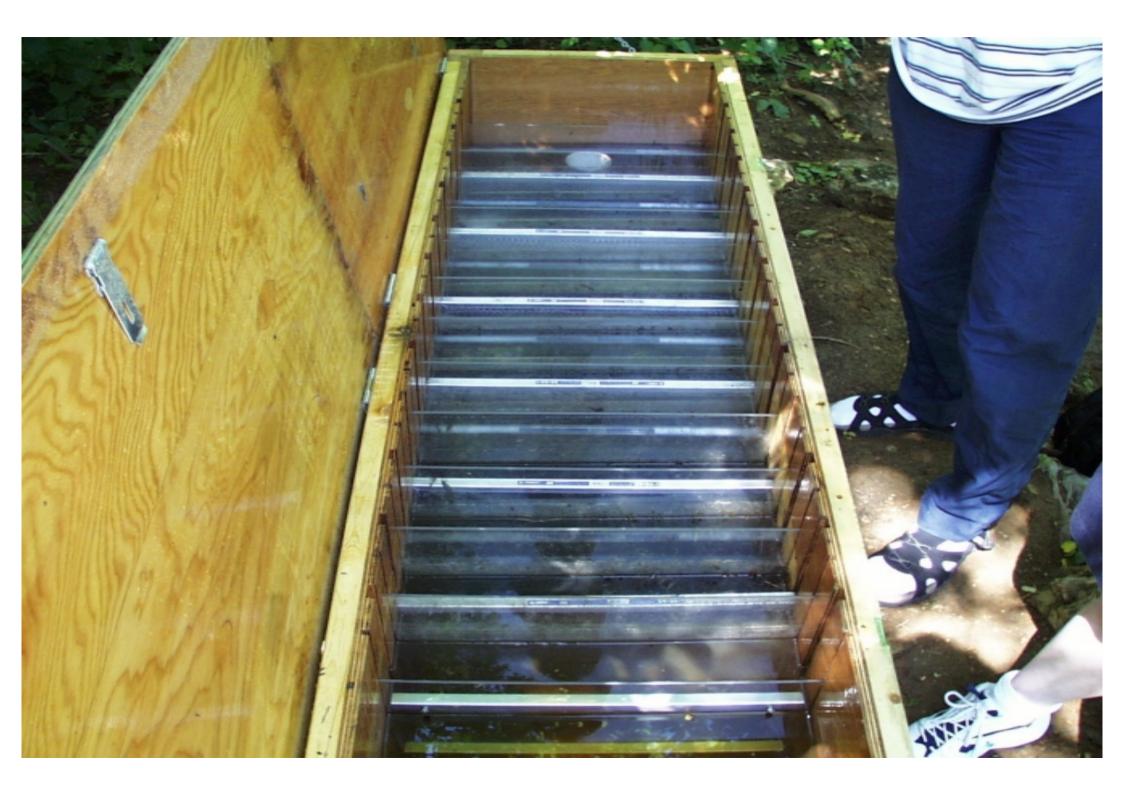


HRTEM Image of Mesoporous Media



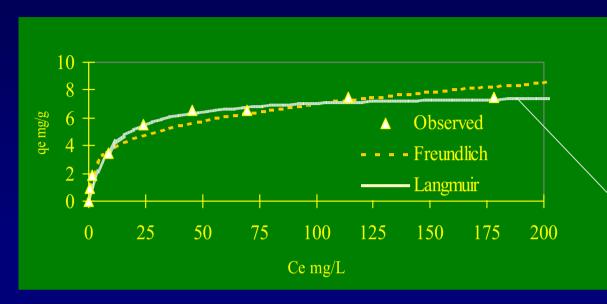






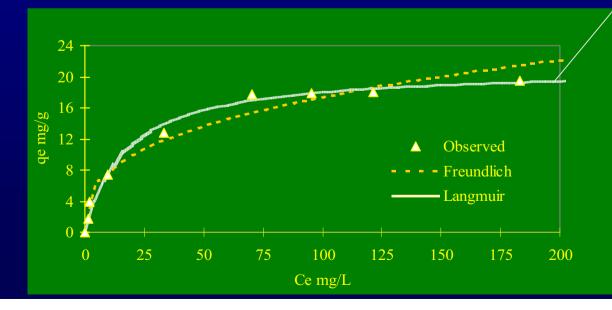


A. Isotherms for the sorption of Cd on to Juniper.



Ion Exchange Reaction

B. Isotherms for the sorption of Cd on to **BTJF**.



Reported Cd Sorption Capacities for Various Sorbents

Sorbent	Cd (mg/g)	Source	
BTJF	<u>21.1</u>	Min et al., (2001)	
Juniper	<u>7.75</u>	Min et al., (2001)	
Walnut shell	1.5	Orhan and Büyükgüngör (1993)	
Pinus pinaster bark	8.00	Teles de Vasconcelos and González Beça (1993)	
Chlorella minutissima	11.14	Roy et al., (1993)	
Activated carbon	11.1	Larsen and Schierup (1981)	
Geothite	3.08	Johnson (1990)	
Acid-treated bentonite	4.11	Pradas <i>et al.</i> , (1994)	
Heat-treated bentonite	16.50	Pradas <i>et al.</i> , (1994)	
Rastunsuo peat	5.058	Tummavuori and Aho, (1980)	
Sphagnum moss peat	5.8	Mclelland and Rock (1988)	
Ascophyllum andosum	<u>67</u>	Volesky and Prasetyo (1994)	
Chitosan powder	<u>420</u>	Pradas <i>et al.</i> , (1994)	

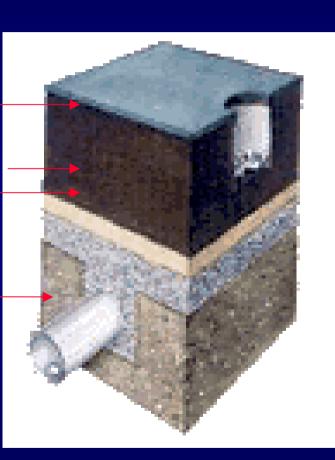


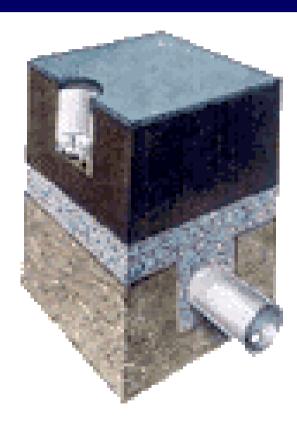
USGA Green Construction Profile

12-14" root zone consisting of an 80% sand/20% peat mix

2" coarse sand intermediate layer
4" pea gravel drainage layer

4" perforated drain tile





Crown III Crumb Rubber Topdressing

Granulated tires with non-rubber components removed.

Applied to high traffic areas on golf courses and athletic fields to increase turfgrass traffic resistance.



Sustained TCE Degradation in Counter-Diffusional Membrane-Attached Methanotrophic Biofilms

Properties of TCE

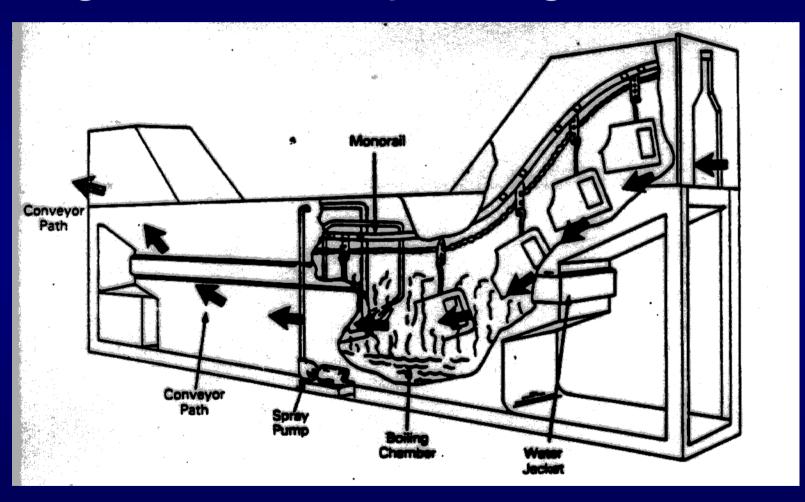
- Chlorinated aliphatic.
- * Xenobiotic.
- Non-flammable.
- Average carbon oxidation state of +1.
- Persistent in environment.
- Excellent solvent properties.

Industrial use of TCE

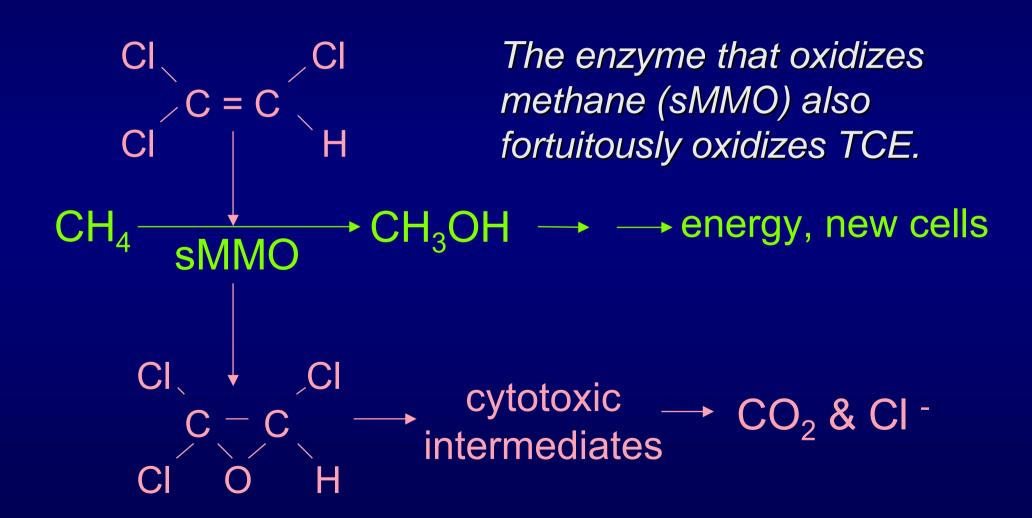
- U.S. demand = 290,000 tons ('92).
- 90% degreasing, 10% chemical intermediate.
- ~30,000 batch vapor degreasers.
- ~3000 large conveyorized vapor degreasers.
- Example:

Brunk industries used ~21 tons of TCE in 1997 in two small vapor degreasers. Waste sludge shipped to TSDF.

Large solvent vapor degreaser



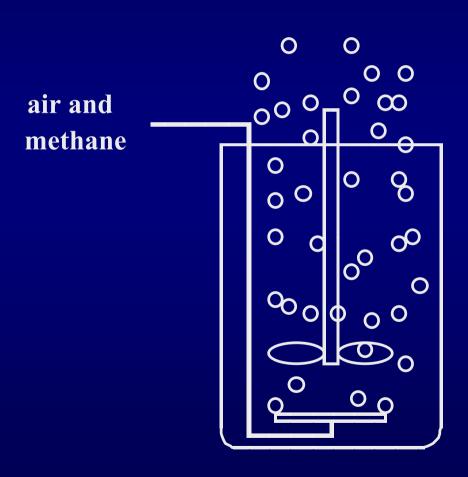
How is TCE cometabolized?



Problems associated with methanotrophic degradation of TCE

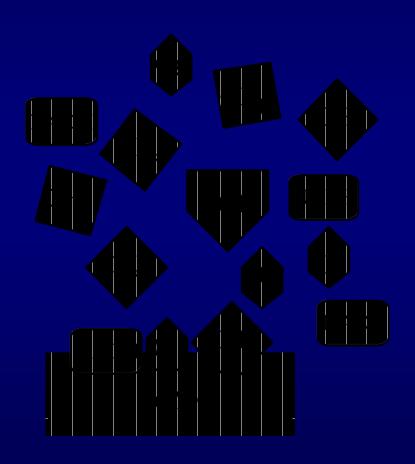
- CH₄ must be supplied as growth substrate.
- Need efficient CH₄ mass transfer.
- Must minimize competitive inhibition.
- Need to overcome TCE byproduct toxicity.
- Want optimal TCE transformation yield.

Mass transfer problems



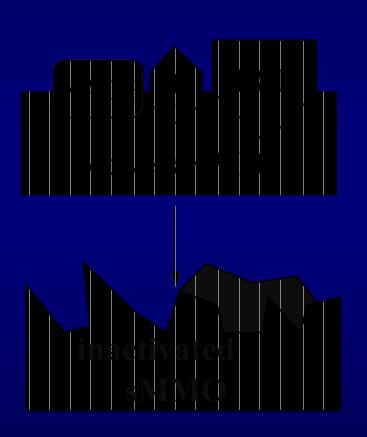
Conventional diffusers
 attain only ~ 30% transfer
 efficiency.

Competitive inhibition problems



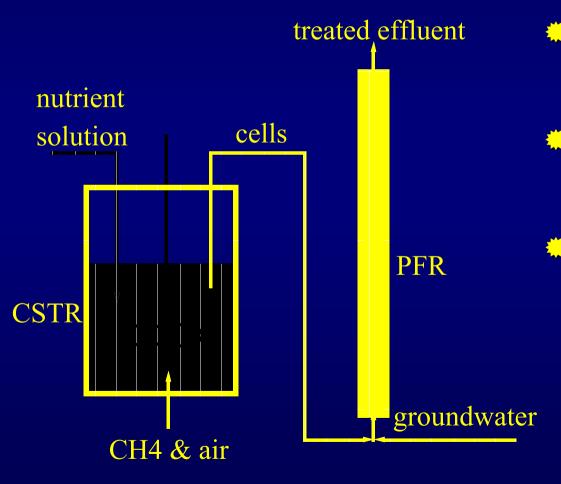
sMMO has a greater affinity for CH₄; thus TCE degradation is inhibited in presence of CH₄.

TCE byproduct toxicity problems



- * TCE transformation products will inactivate sMMO.
- Thus, enzymes have a limited "TCE transformation capacity."

Two-stage reactor concept



- separate growth & treatment reactors.
- continuous supply of new cells to treatment reactor.
- newer designs use membrane gas-transfer

Problems with two-stage designs

- (1) Poor CH₄ & O₂ transfer efficiency in growth reactor.
- (2) O₂ limitations in treatment reactor.
- (3) Dilution of cells in treatment reactor.
- (4) Incomplete expenditure of active cells in treatment reactor.

Inventing by Problem Recognition and Solution

- Most inventions are conceived by the following two-step procedure:
- 1. Recognizing a problem (P)
- 2. Fashioning a solution (S)

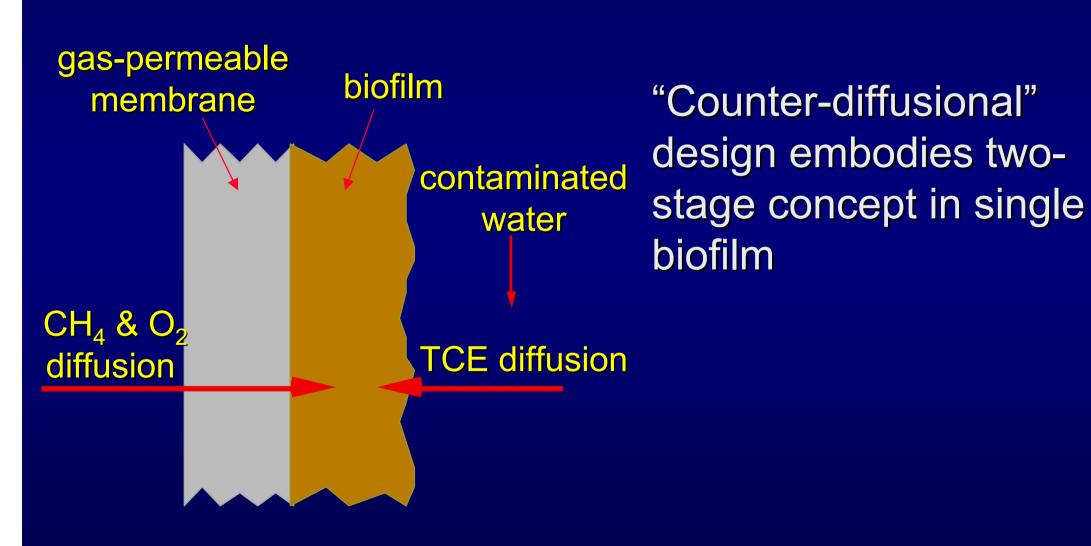
Examples:

- Buried plastic cable-locator strip.
- Magnetic safety lock for police pistols
- VCR plus
- Xerography
- Organic production of acetone
- Grocery shopping cart

Inventing by Magic (Accident and Flash of Genius)

- Goodyear invented rubber vulcanization when he accidentally added some sulfur to a rubber melt.
- A chemist accidentally left a crutcher (soap-making machine) on too long, causing air to be dispersed into the soap mixture. He found that the soap floated when it hardened, thus giving birth to floatable soap bars, such as Ivory® brand. Another chemist accidentally mixed some chemicals together and spilled them, finding they hardened to a flexible, transparent sheet (later known as "cellophane").

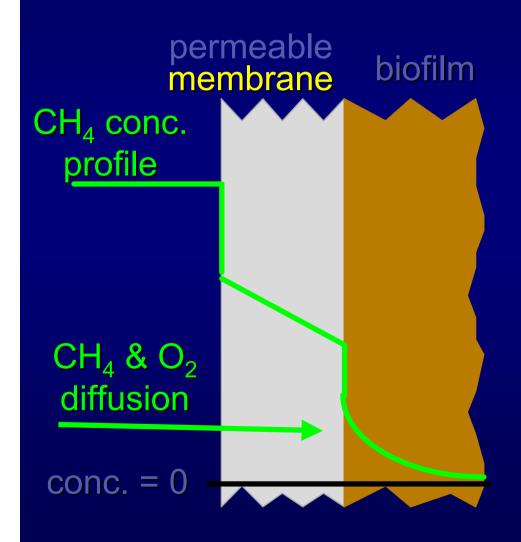
Research objective



Main hypotheses

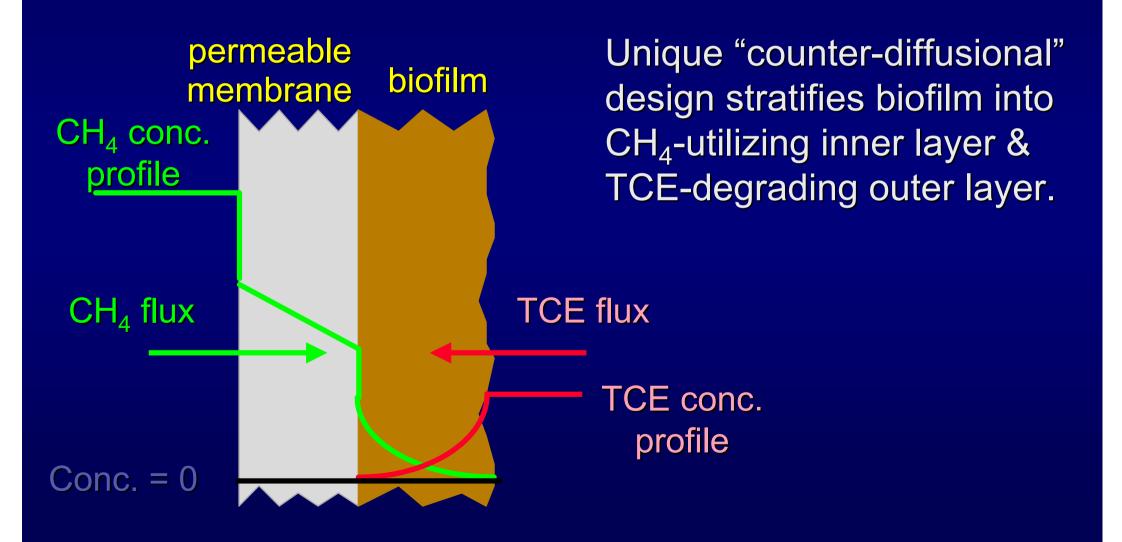
- * Attain of 100% CH₄ & O₂ transfer efficiencies.
- Minimize competitive inhibition between CH₄ & TCE.
- Optimize retention of TCE-active biomass.
- Minimize accumulation of TCE-inactive biomass.

How are 100% CH₄ & O₂ transfer efficiencies attained?

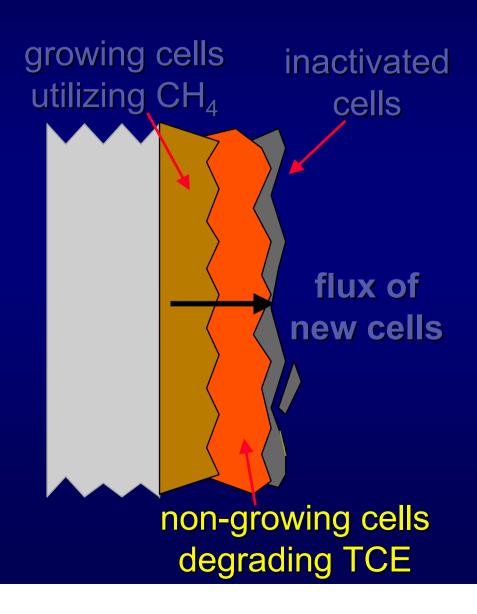


All CH₄ & O₂ diffusing through membrane is utilized in biofilm.

How is competitive inhibition minimized?



How is active biomass retained & inactive biomass removed?

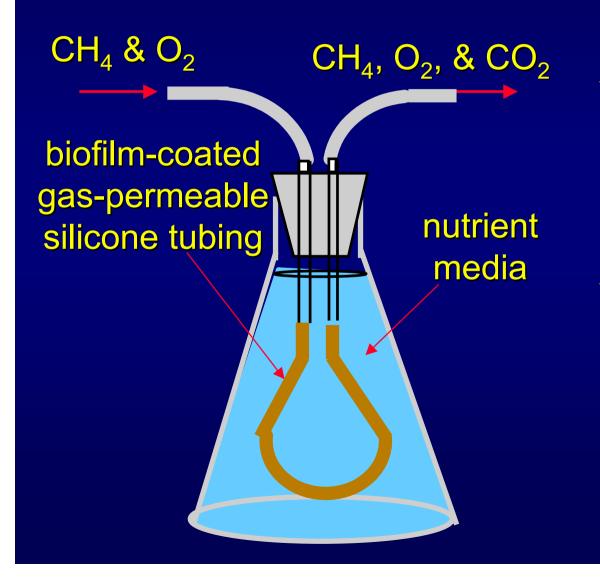


- Active cells at biofilm interior are retained.
- Inactived cells at biofilm exterior erode from surface.
- Flux of new cells from biofilm interior.

Research objectives

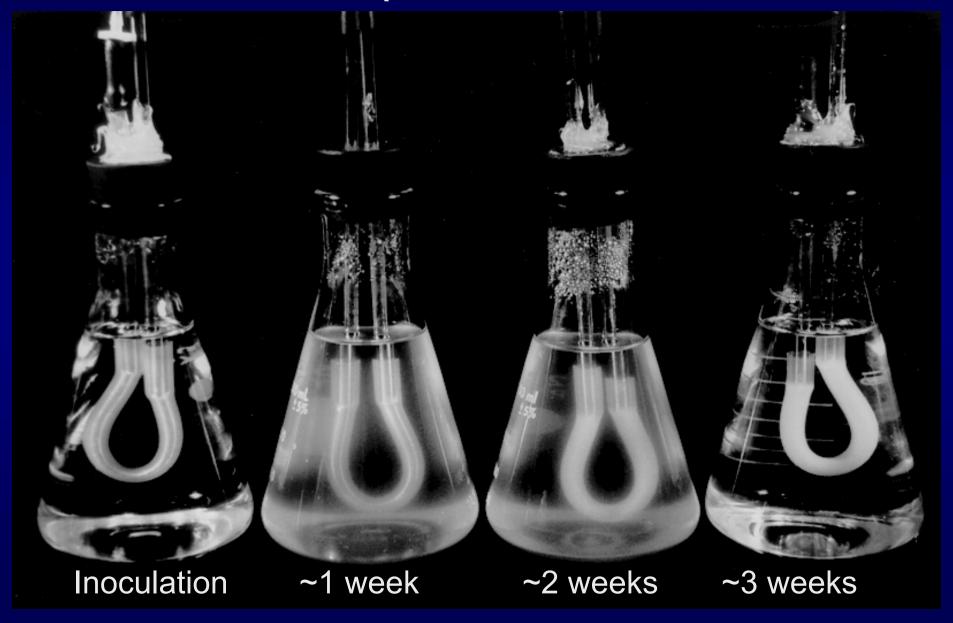
- Cultivate biofilms on gas-permeable membranes pressurized with CH₄ & O₂.
- Characterize biofilm structure.
- Quantify biofilm metabolic activity.
- Quantify TCE degradation rates.
- Quantify TCE transformation yields.

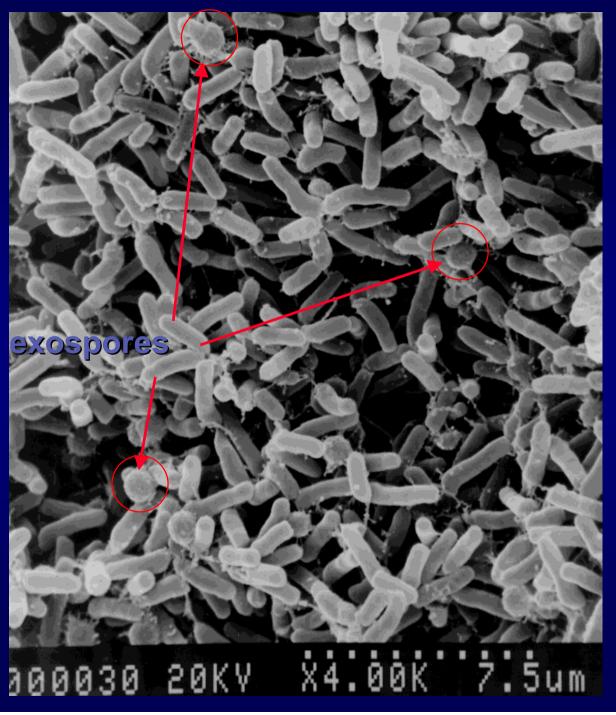
Batch experiments



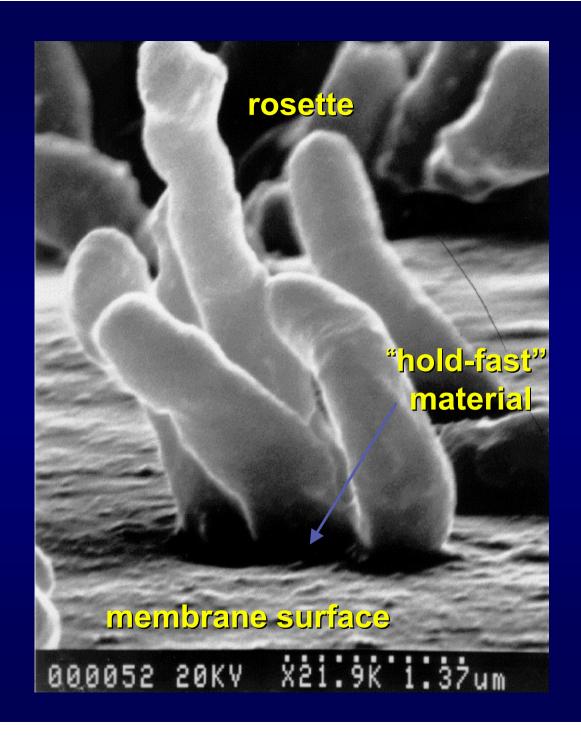
- CH₄ & O₂ pumped through silicone tubing immersed in sterile nutrient media.
- Media inoculated with pure Methylosinus trichosporium OB3b.

Biofilm Development Over Time

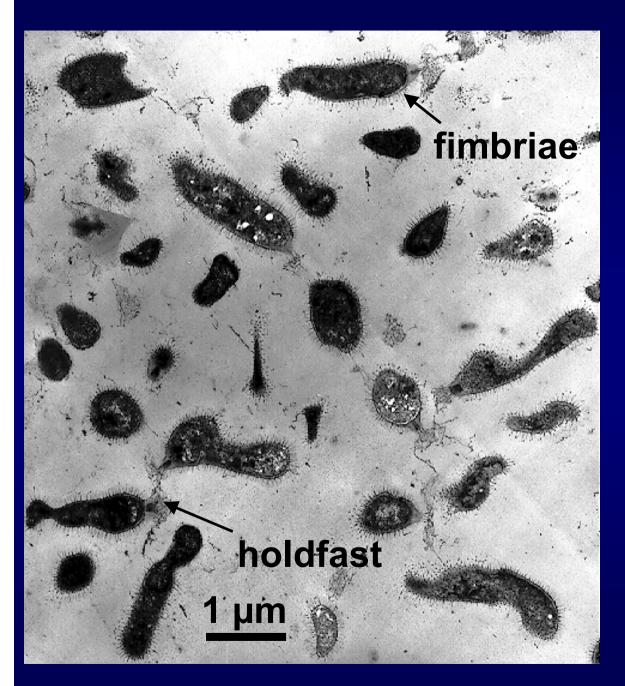




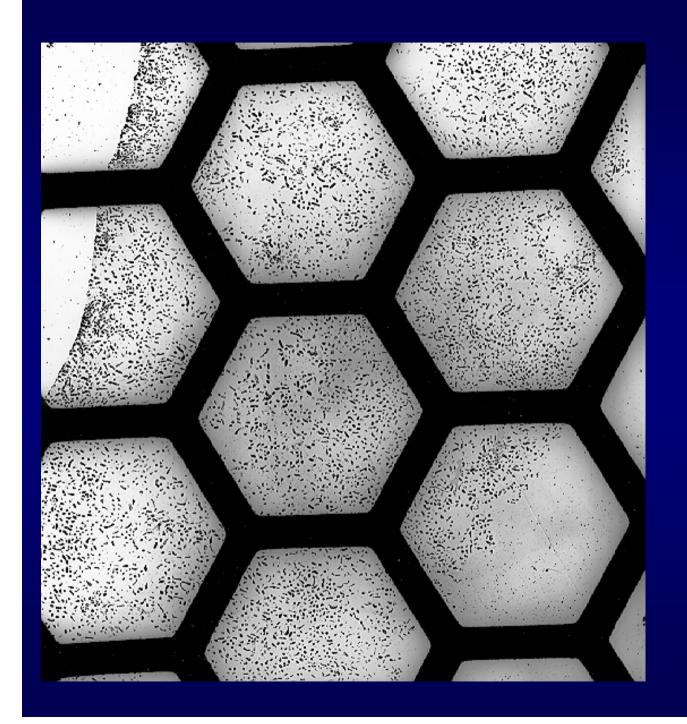
- Homogeneous cell distribution & high cell density.
- Uniform cell morphology verified pure culture.
- Scattered presence of exospores.



M.t. OB3b cells appeared to adhere to conditioning film on membrane surface by means of "hold-fast" material responsible for rosette formation.



- Cells had pronounced fimbriae, which may aid biofilm formation.
- Cells also had polar "holdfast" structures.

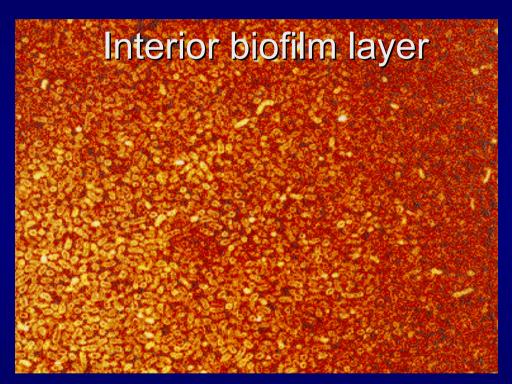


TEMs revealed uniform cell distribution, with biofilm thicknesses between 150-350 µm.

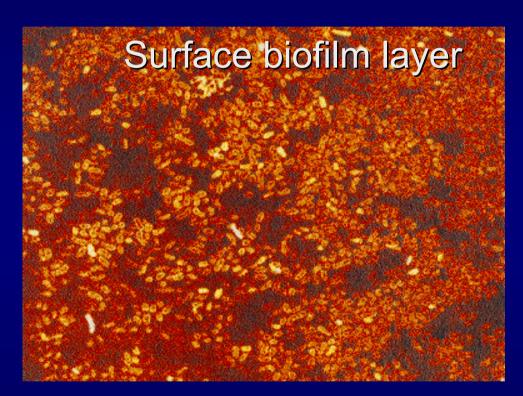
Treating chlorinated solvent contaminated groundwaters

- Air stripping.
- GAC adsorption.
- GAC & incineration.
- Anaerobic biological dehalogenation.
- Metallic iron dehalogenation.
- Cometabolic oxidation toluene degraders.
- Cometabolic oxidation methanotrophs.

Scanning confocal laser microscope (SCLM) optical-sections of biofilms

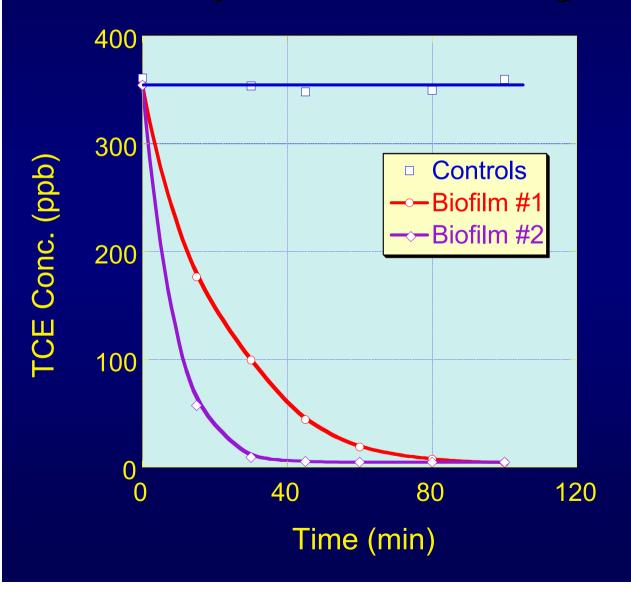


Cells near CH₄-permeable membrane were densely & homogeneously packed.



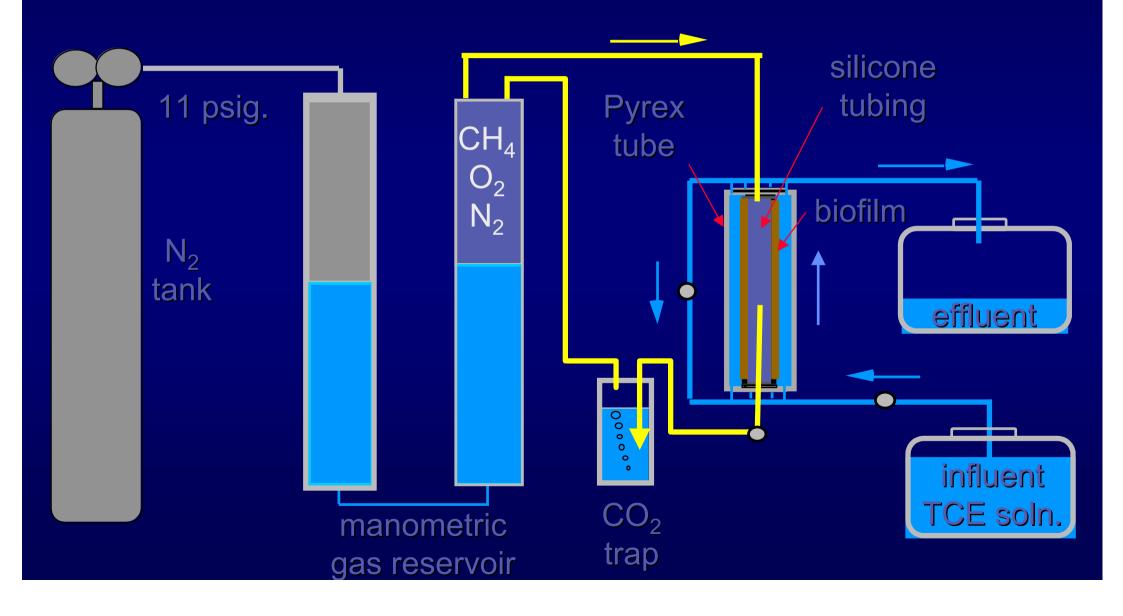
Cells near biofim surface were less densely packed & had a contoured texture.

Batch system TCE degradation results



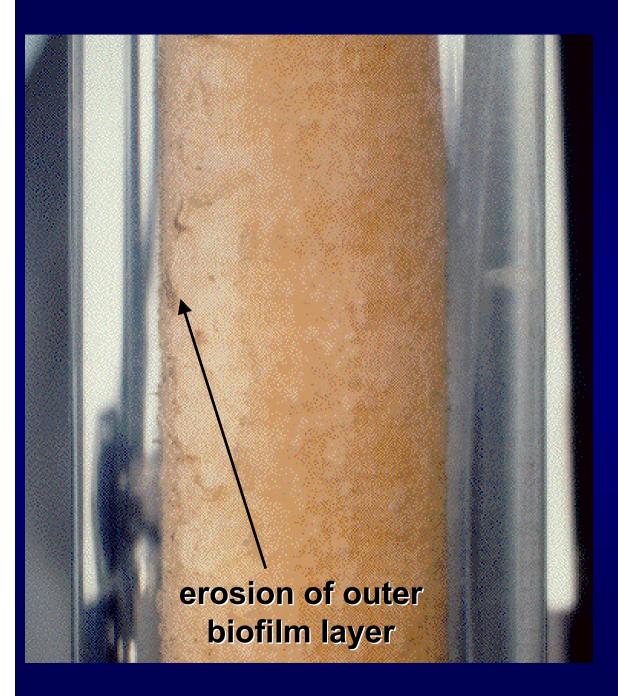
- Biomass scraped off of membrane & suspended in buffer could rapidly degrade TCE.
- 1st-order rate constants ranged from 0.13-0.33 mL/min/mg.

Bench-scale bioreactor studies

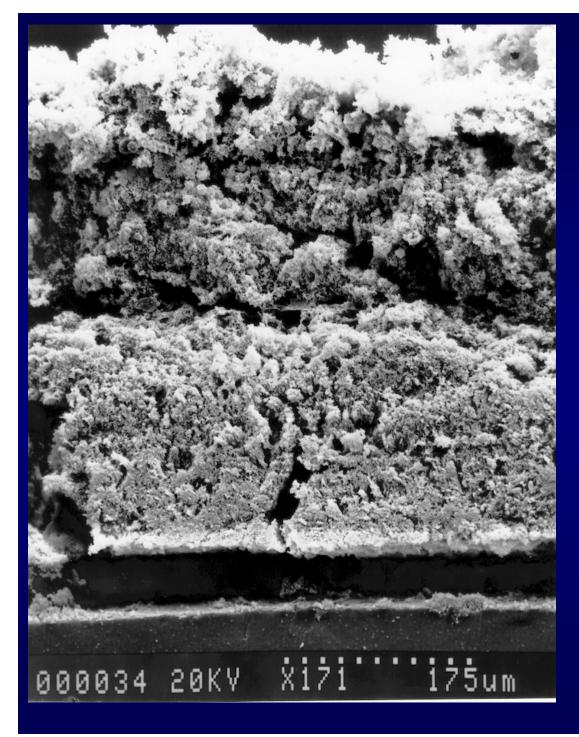


Methods for cultivating methanotrophic biofilm in reactor

- Pure M. t. OB3b culture circulated past tubing until biofilm formed.
- 35% CH₄ & 55% O₂ in gas reservoir at total pressure of 1.04 atm.
- Once seeded, non-sterile nutrient solution pumped through reactor.

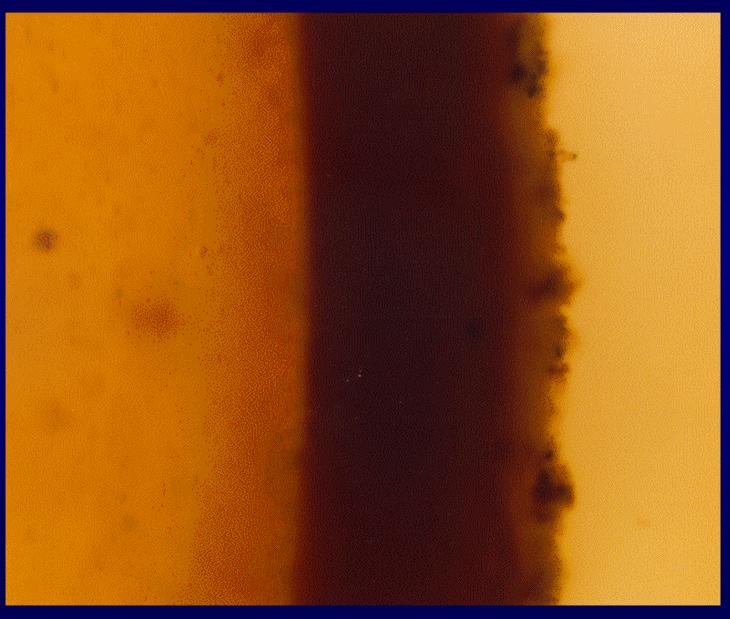


- Thick, robust biofilm formed after ~3 weeks.
- Evidence of outer layer eroding from biofilm.
- Bulk liquid remained very clear.



- Final "steady-state" biofilm thickness ~500 μm.
- Cell density appeared greater at biofilm interior.

Dense biofilms could be cross-sectioned while attached to the silicone membrane.



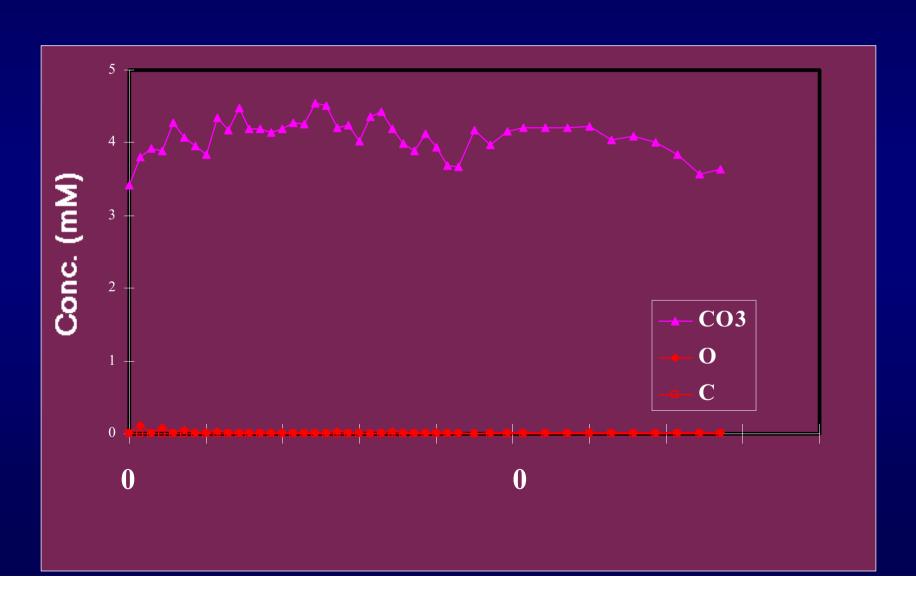


- SEMs confirmed a diverse bacterial population.
- Predominant cell morphology was consistent with *M.t.* OB3b.
- Numerous cells with morphology of *Hyphomicrobium* spp.

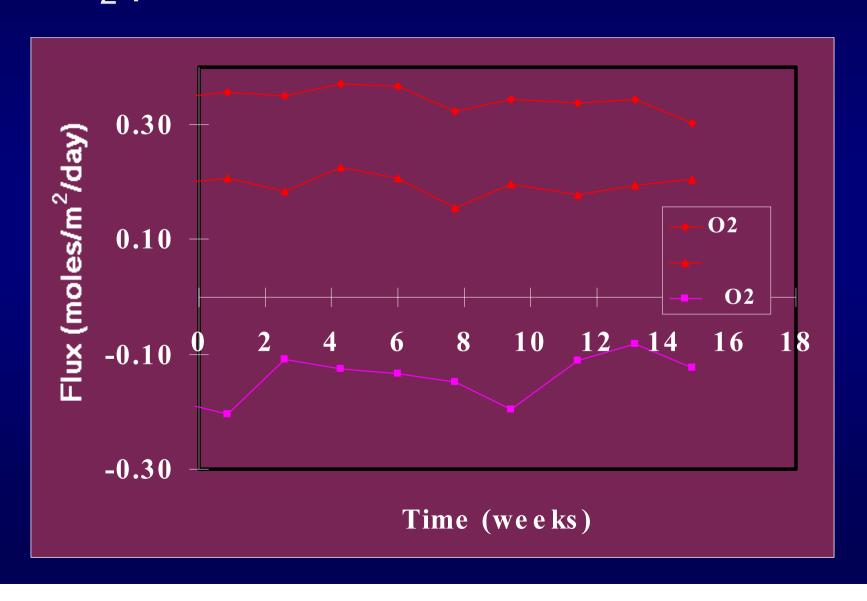
Reactor operating conditions

gas pressures		aqueous flows		aqueous concs.	
total	1.72 atm	feed	7.6 mL/hr	O2 & CO2	atm. equil
CH4	0.40 atm	recycle	300 mL/mii	CH4	0
02	0.68 atm	HRT	28 hr	CI-	0
N2	0.60 atm			TCE	0-50 킡
CO2	<0.01 atm				

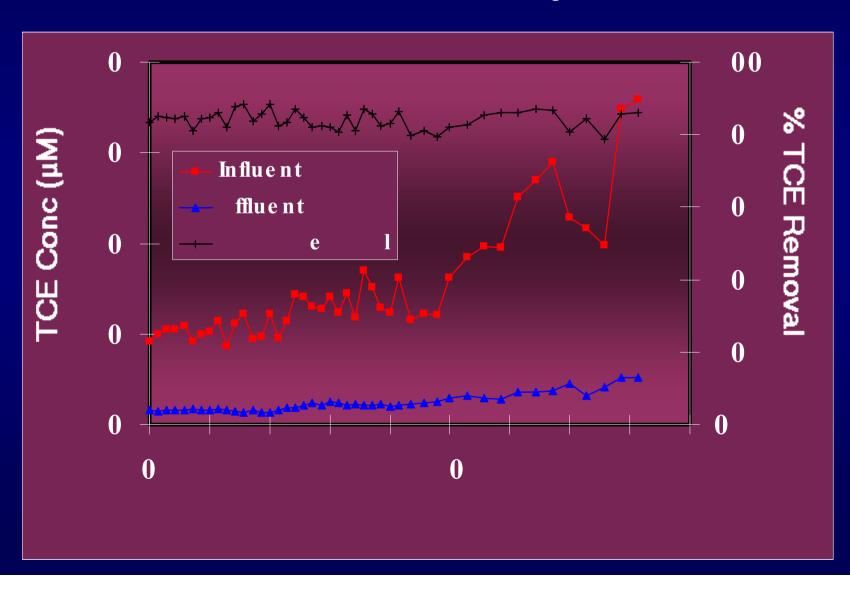
Effluent CO₃, O₂, & CH₄ concentrations



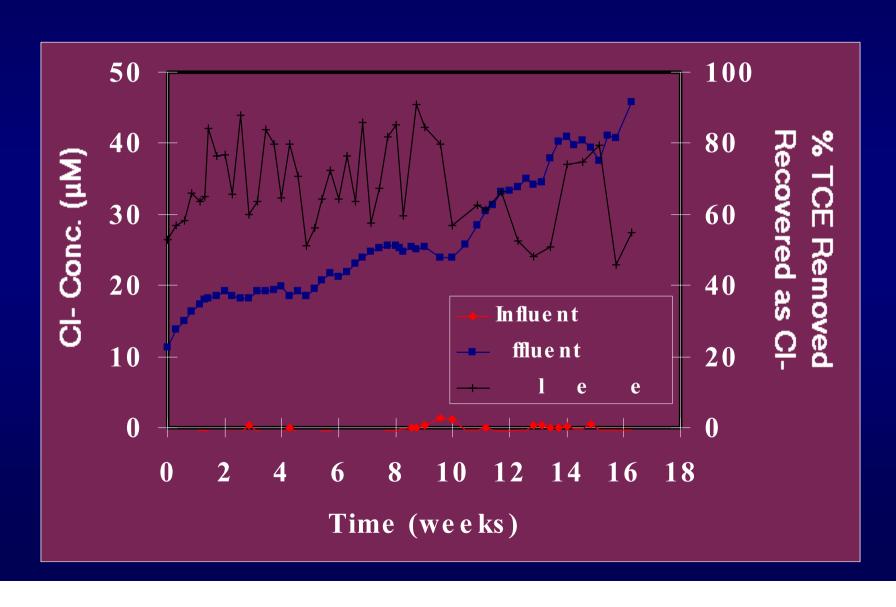
Rates of O₂ & CH₄ utilization and CO₂ production in biofilm



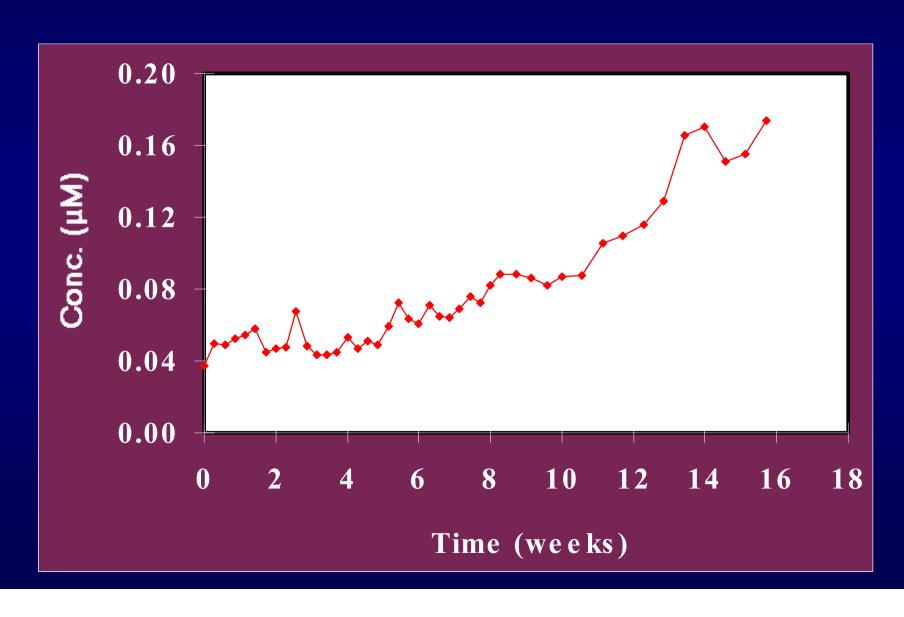
TCE removal efficiency



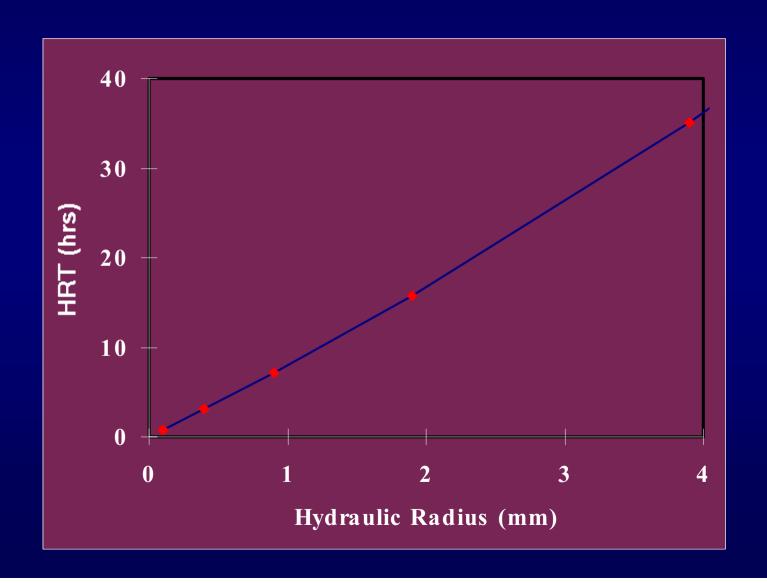
Percent of removed TCE recovered as Cl-ion

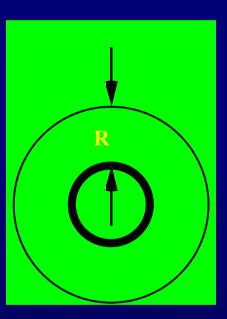


Effluent trichloroethanol concentrations



Theoretical HRT for 95% removal versus hydraulic radius





Conclusions

- High TCE degradation rates sustained.
- ♦ 100% CH₄ & O₂ transfer efficiencies.
- Competitive inhibition minimized.
- Active cells optimally retained in the reactor.
- Minimal accumulation of inactive cells?
- High overall TCE transformation yields.

Thanks to:

- Dr. Lee Clapp, Dr. Daniel Noguera, Dr. Bill Boyle, Dr. Brian Fox, Dr. Doug Cameron, Dr. Bill Hickey, Dr. Tim Donohue, Paul Fritschel.
- Jack Newman, John Regan, Ruddy Hartono, Firdaus Ali, Elizabeth Marshall, Anna Casasus.
- Wisconsin Groundwater Coordinating Council, U. W. Industrial Relations Board, NIH Biotechnology Training Program.