US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT

MtBE Biodegradation in High Biomass Reactors

Albert D. Venosa
Land Remediation and Pollution Control Division
NRMRL-Cincinnati, OH

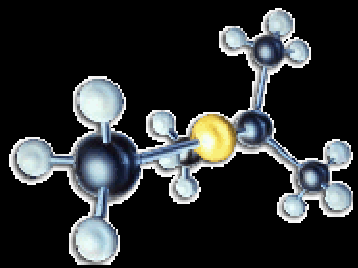
US EPA ARCHIVE DOCUMENT

Topics To Be Discussed

- Brief background
- Initial bench top studies with continuous flow porous pot reactors
  - MtBE as sole carbon source
  - MtBE in combination with other carbon sources
  - Kinetics of MtBE biodegradation
- Bench top MBR performance
- Microbiology (culture ID)
- Pilot-scale gravity-flow reactor
- Porous pot study of Millville, NJ Superfund Site
- Economic Analysis

US EPA ARCHIVE DOCUMENT

Background: MtBE Properties



- **Low odor and taste threshold**
  - **53 μg/L odor threshold**
  - **20-40 μg/L taste threshold**
- **Highly soluble in water (55 g/L) with limited sorptivity to aquifer solids**
- **Low yields of MtBE-degrading cultures**
  - **0.20-0.28 g cells/g MTBE (Salanitro *et al.*, 1994)**
  - **0.09-0.12 g cells/g MTBE (Fortin & Deshusses, 1999)**
  - **0.083 g VSS/g MTBE (G. Wilson *et al.*, 2001)**

US EPA ARCHIVE DOCUMENT

Consequences of Low Yield

- Expected biomass levels in a typical, conventional reactor if influent MtBE is 5 mg/L:

  5 mg MtBE/L x 0.14 mg VSS/mg MtBE = 0.7 mg VSS/L

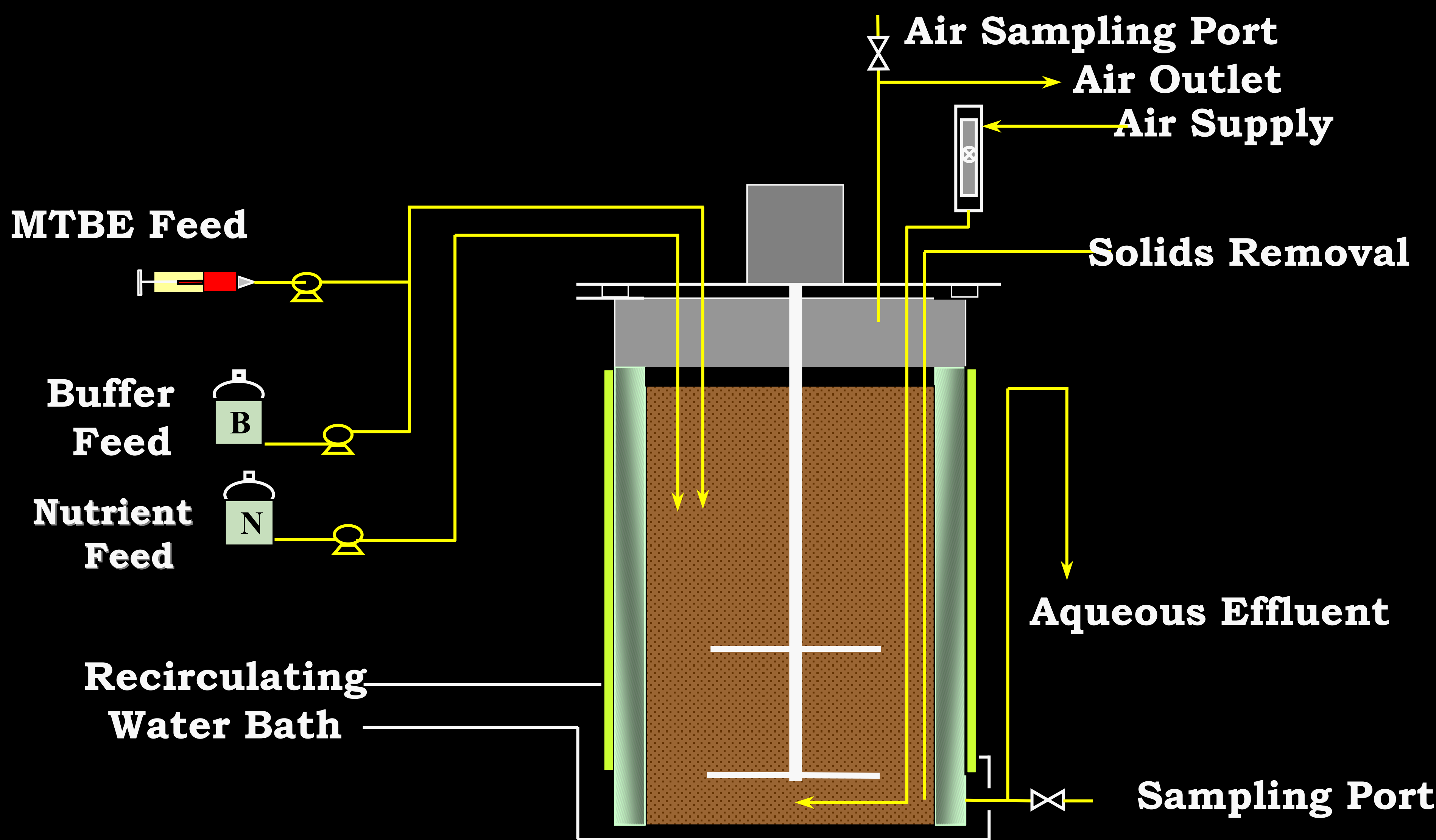
- Need to retain as much biomass as possible to achieve treatment goals

US EPA ARCHIVE DOCUMENT

Initial Bench Top Porous Pot Reactor Studies

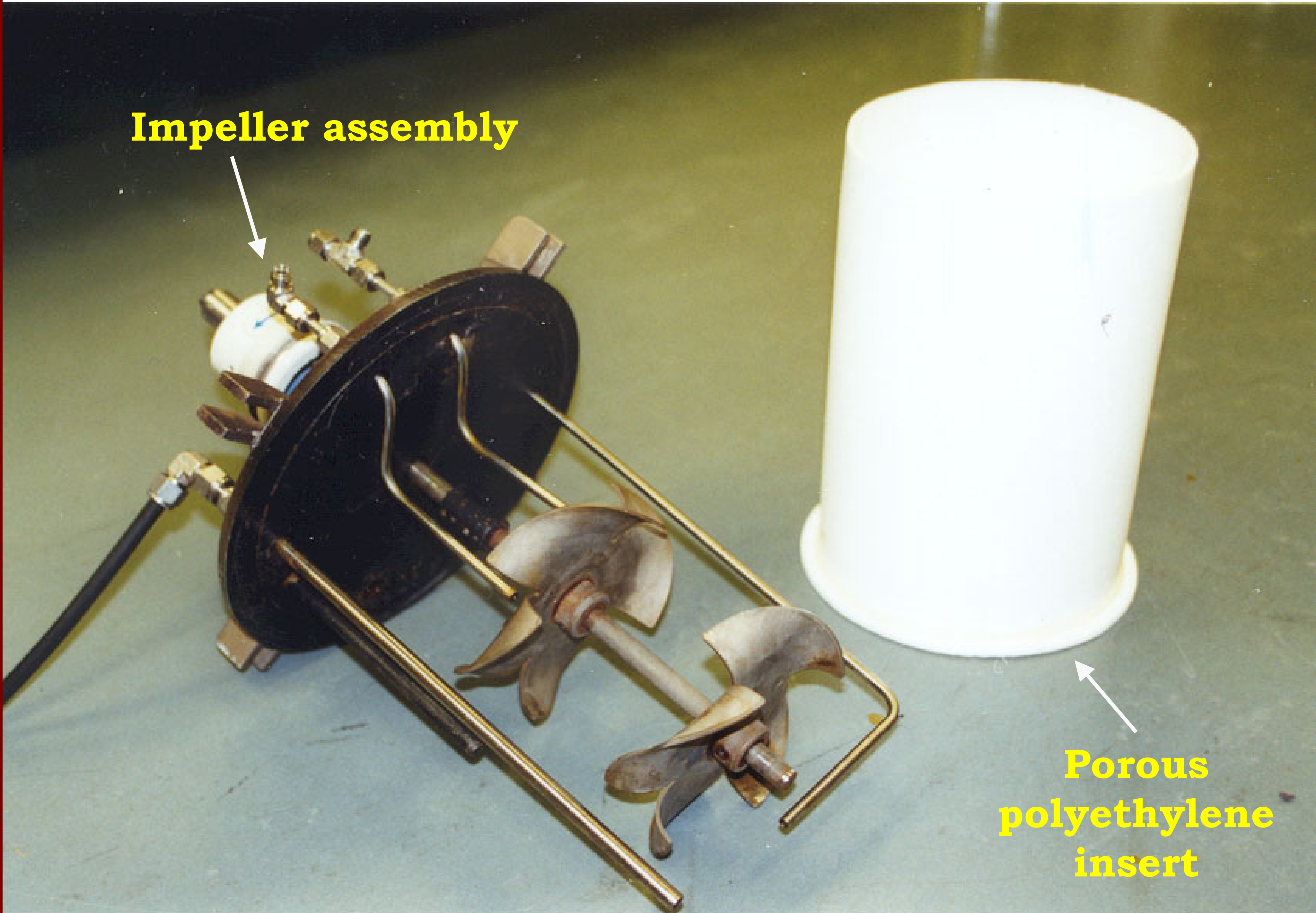
US EPA ARCHIVE DOCUMENT

Schematic of Porous Pot Bioreactor

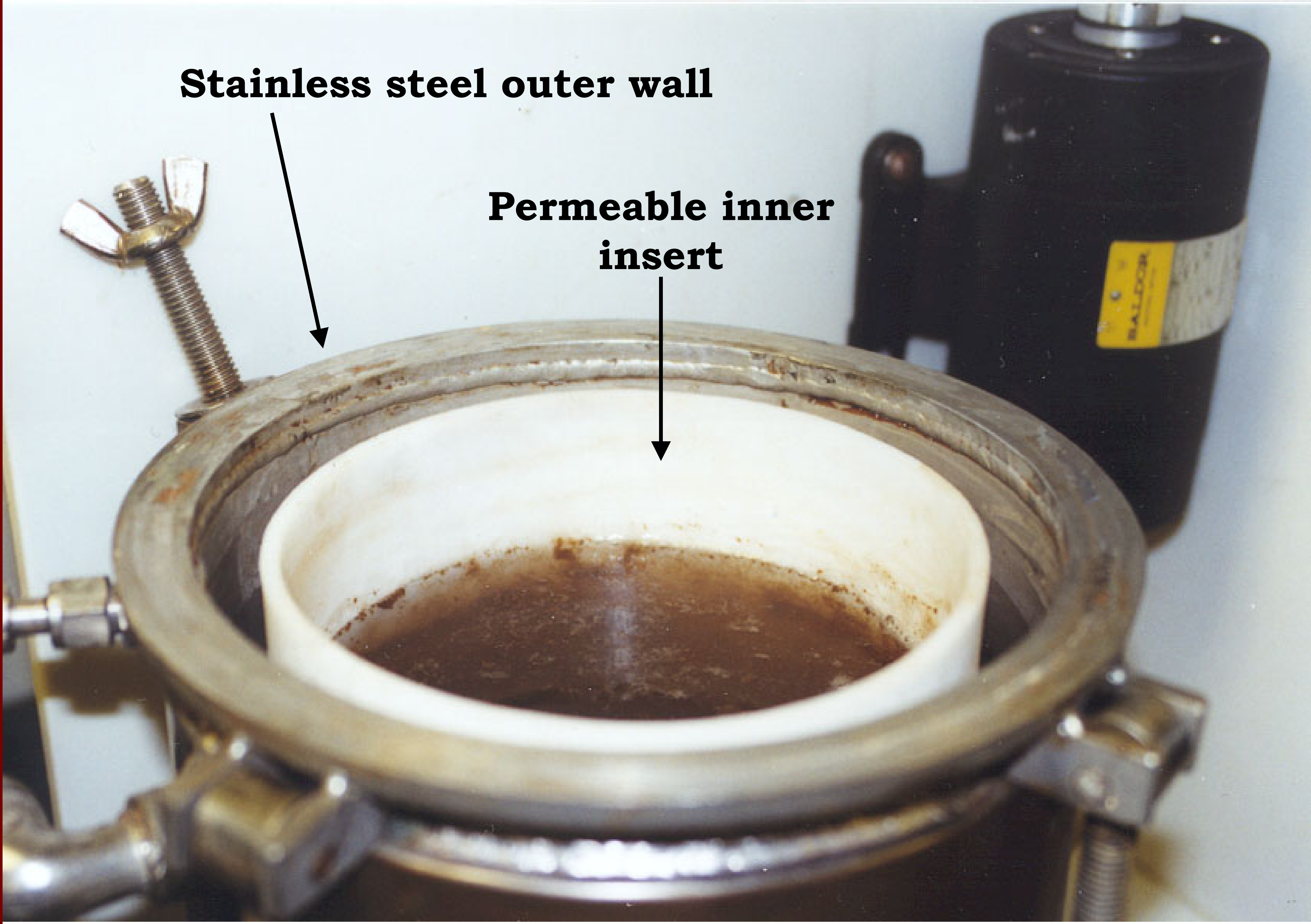


US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT



US EPA ARCHIVE DOCUMENT



US EPA ARCHIVE DOCUMENT

Operating Conditions of Porous Pot

- 6-L aeration volume in 12-L capacity vessel
  - Porous pot insert made of polyethylene
- Feed flow rate: 2.4 L/day
  - HRT = 2.5 days
- Initial solids wastage rate: 5%/day
  - SRT = 20 days
- Wastage rate substantially reduced after ~115 days (solids wasted only due to sampling)

US EPA ARCHIVE DOCUMENT

Operating Conditions

- Influent carbon source: MtBE alone or in combination with DEE, DIPE, EtOH, BTEX
- Substrate concentrations (COD = ~420 mg/L):
  - 150 mg/L when MtBE alone
  - 75 mg/L when MtBE combined with other carbon sources
- Temperature = 20°C
- HRT = 2.5 days
- pH = 7.5 to 8.1
- Dissolved Oxygen > 3 mg/L

US EPA ARCHIVE DOCUMENT

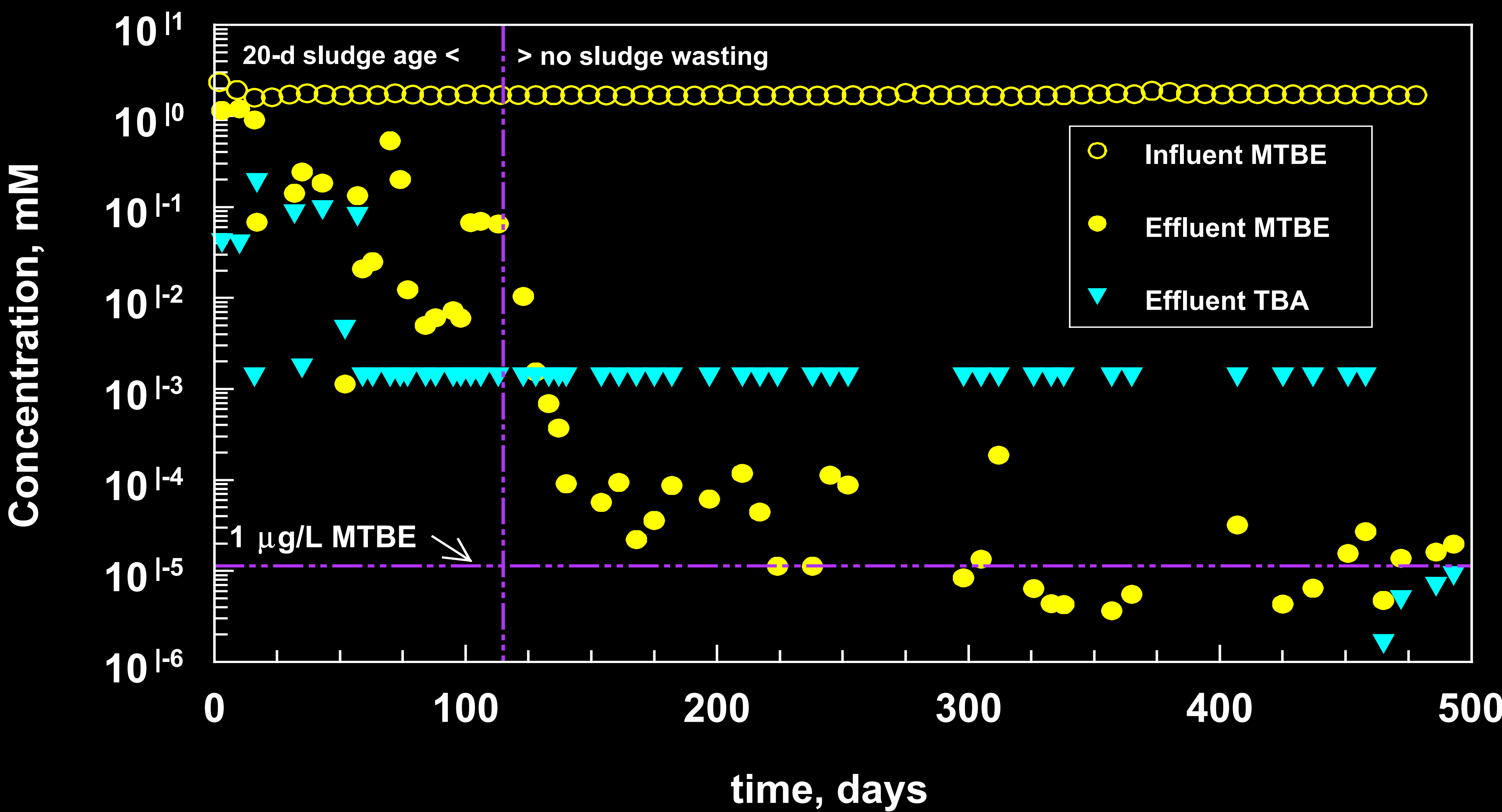
System Startup: Seed Culture

- 2 L mixed liquor from Mill Creek Sewage Treatment Plant, Cincinnati, OH
- 600 mL of mixed liquor from Shell Dev't. Corp. Refinery, Houston, TX
- 140 mL of aquifer material wash water, Port Hueneme, CA

US EPA ARCHIVE DOCUMENT

Performance Results from Initial Porous Pot Studies

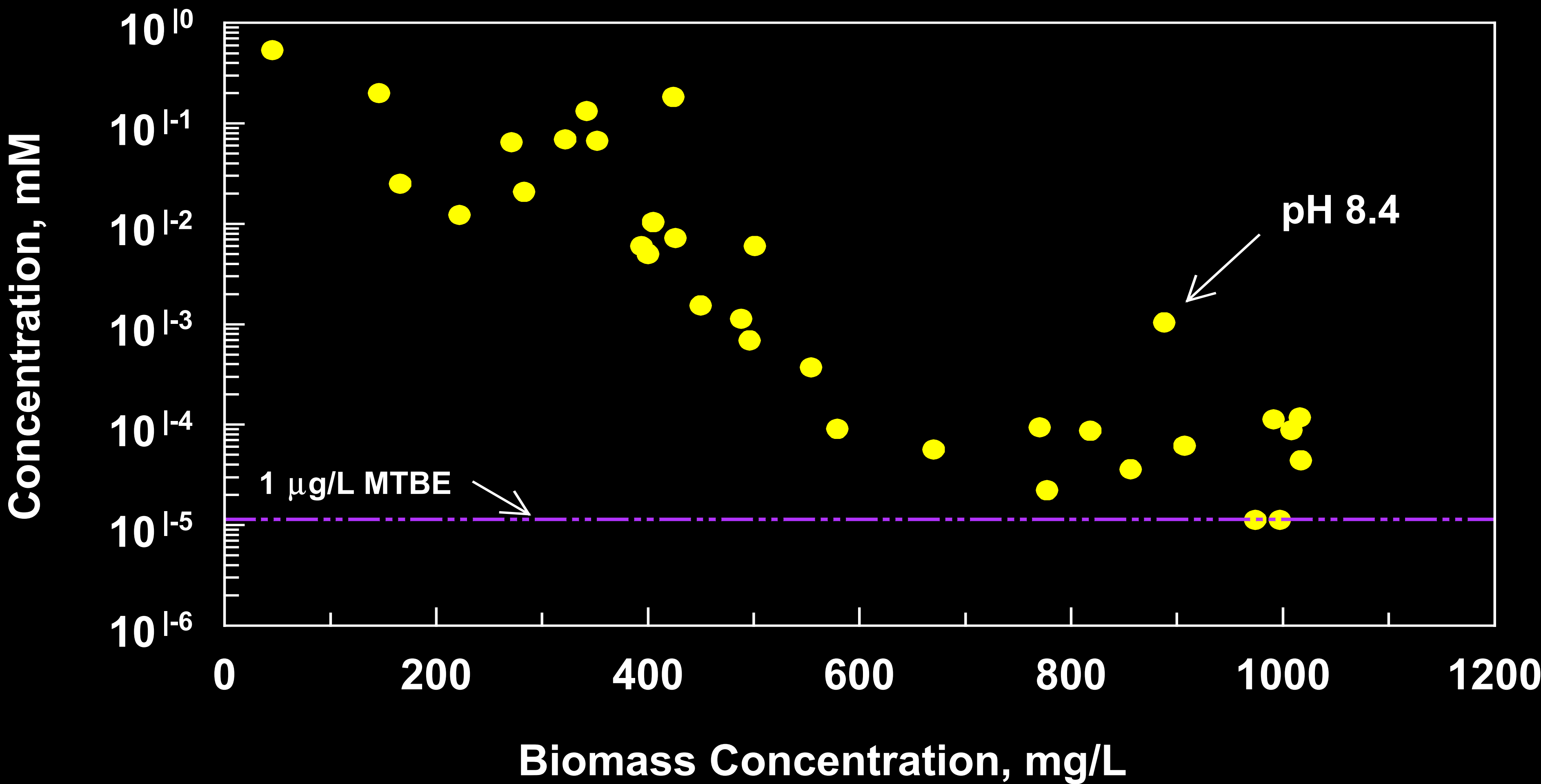
MTBE-Fed Reactor



US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT

MTBE Biodegradation as a Function of Biomass Concentration

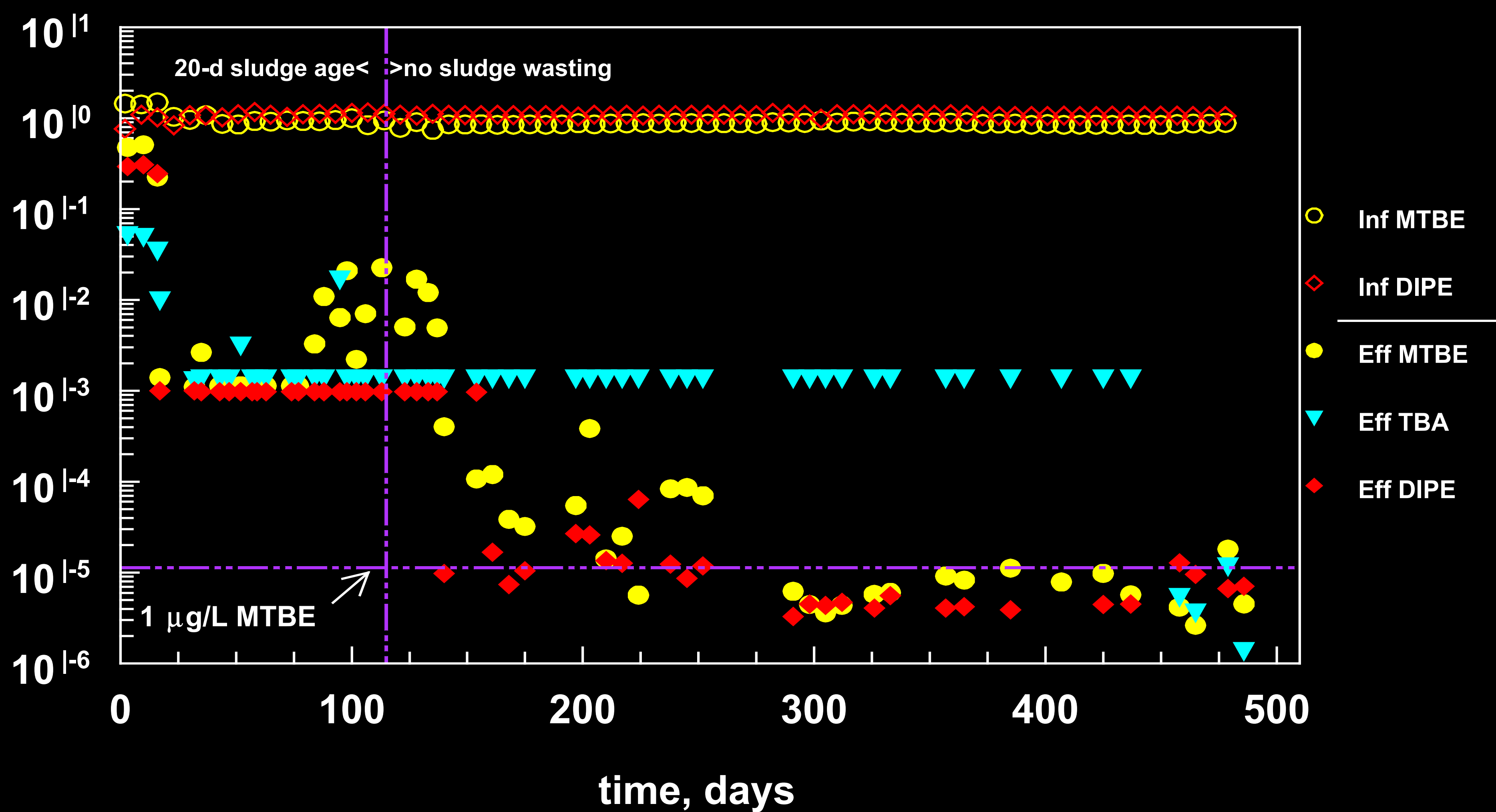


US EPA ARCHIVE DOCUMENT

MTBE/DIPE-Fed Reactor

US EPA ARCHIVE DOCUMENT

MTBE/DIPE-Fed Reactor



Summary of System Performance: All Active Reactors

	Influent MTBE, mg/d	MTBE in Effluent, mg/d	MTBE in Exhaust Air, mg/d	Percent Removed
MTBE Alone	355.50	0.013	0.002	99.99
MTBE and DEE	177.75	0.033	0.003	99.98
MTBE and DIPE	177.75	0.018	0.002	99.99
MTBE and Ethanol	177.75	0.010	0.002	99.99
MTBE and BTEX	202.35	0.060	0.002	99.97

US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT

Summary of Continuous Flow Porous Pot Experiments

- At high biomass concentrations, MTBE biodegraded ~99.99% in *presence or absence* of other carbon sources
- COD and carbon analysis confirmed mineralization of MTBE and its intermediates, with the effluent carbon virtually all in inorganic form
- No significant loss of MTBE from the control abiotic reactor, indicating good system integrity

US EPA ARCHIVE DOCUMENT

Serum Bottle Batch Experiments

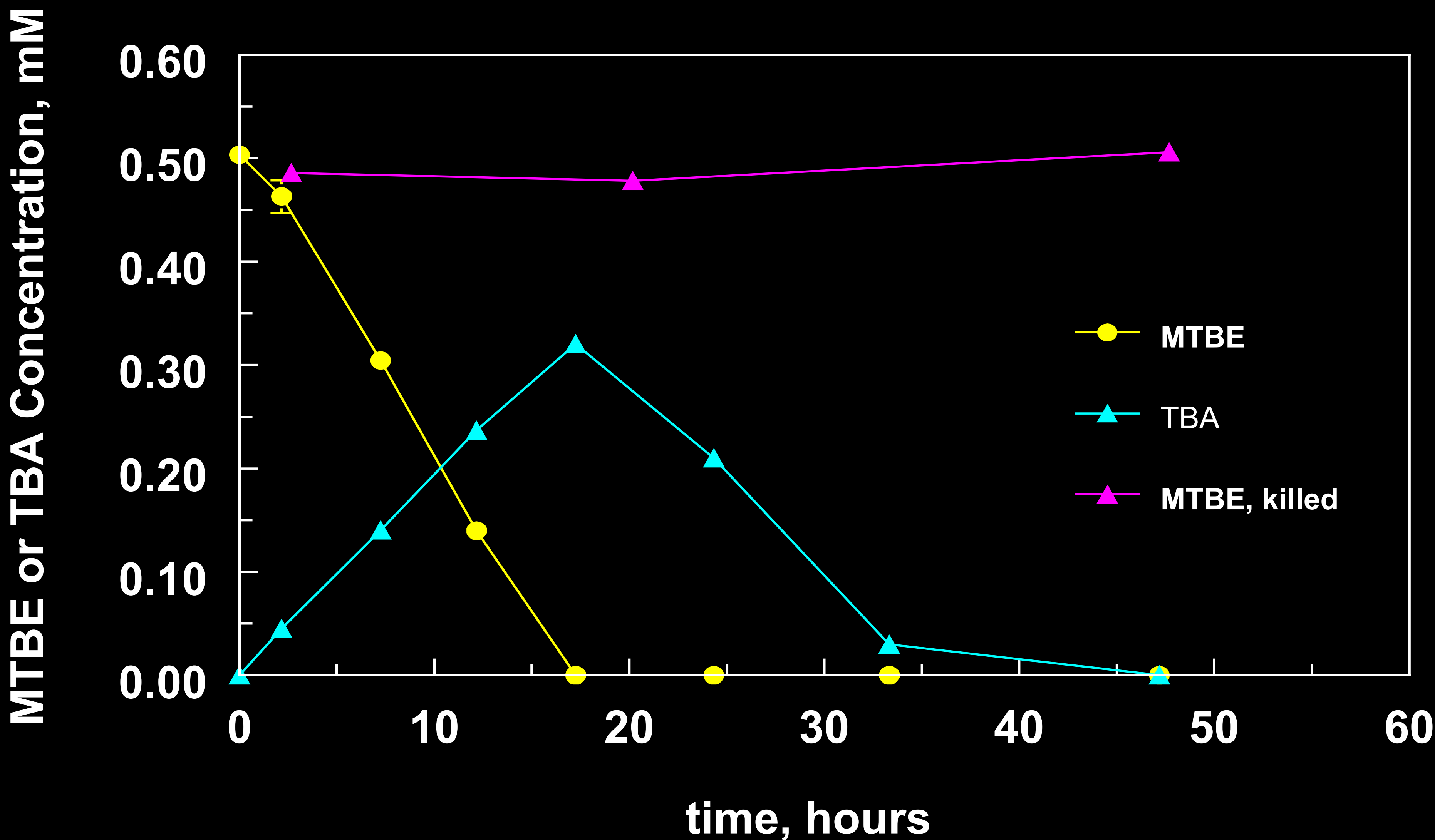
US EPA ARCHIVE DOCUMENT

Experimental Conditions

- 3 substrates fed to separate serum bottles in triplicate
  - MTBE alone
  - TBA alone
  - MTBE + TBA
- 3 Feed Concentrations
  - 5, 15, and 45 mg/L (0.057, 0.17, 0.51 mM)
- Analyze for substrates and intermediates during biodegradation

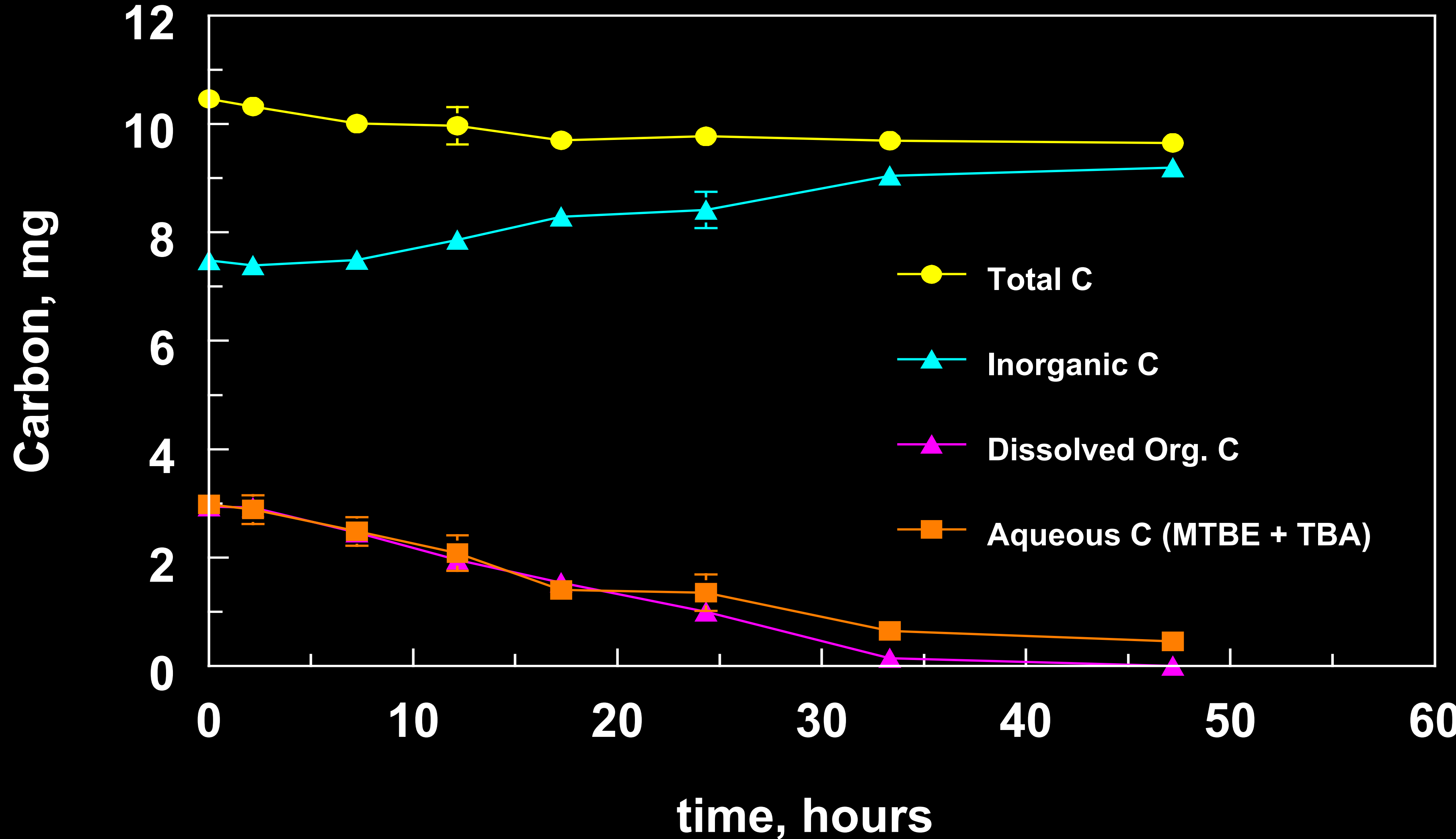
US EPA ARCHIVE DOCUMENT

MTBE-Spiked Serum Bottle
0.5 mM



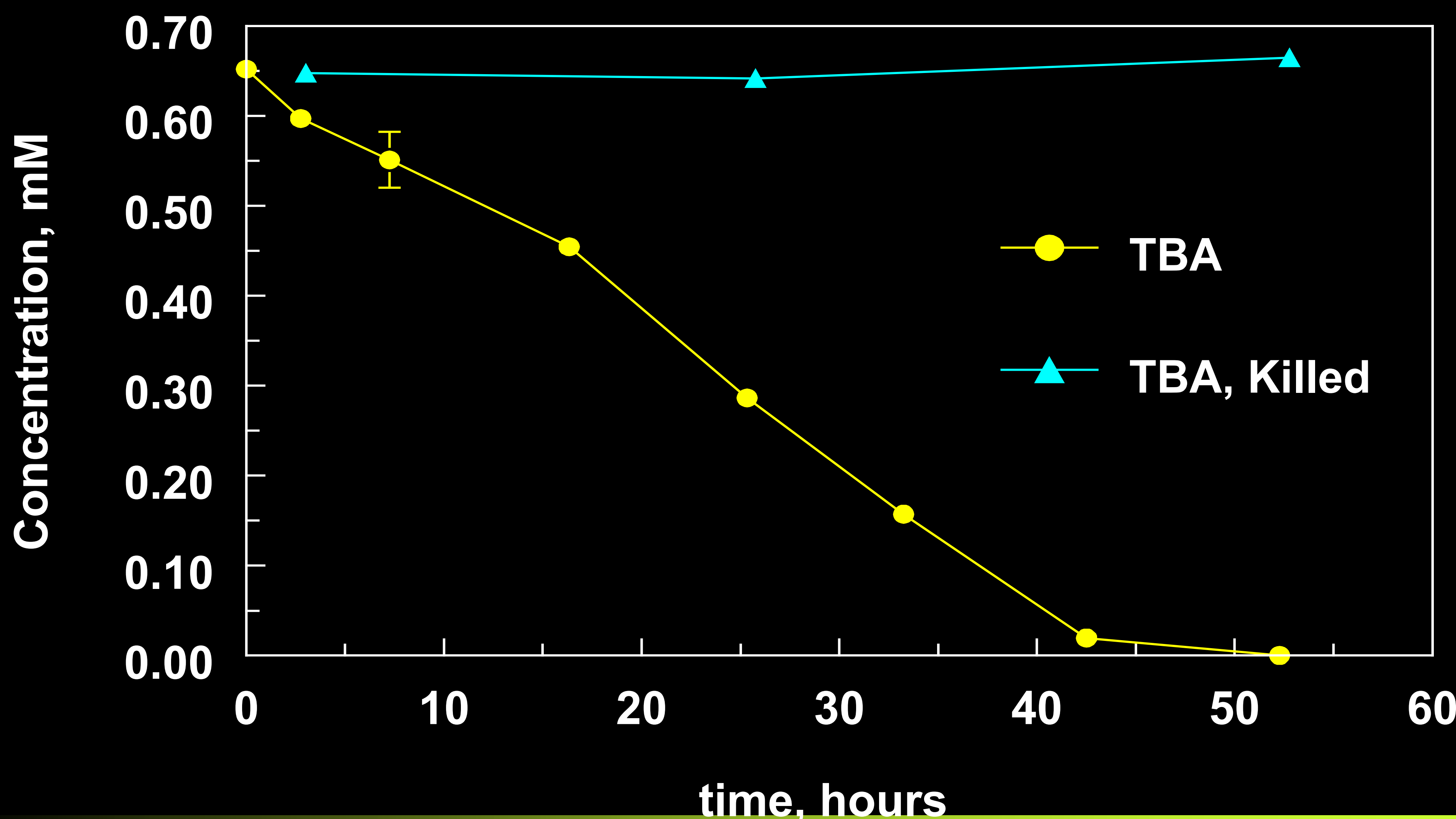
US EPA ARCHIVE DOCUMENT

Carbon Balance
0.5 mM



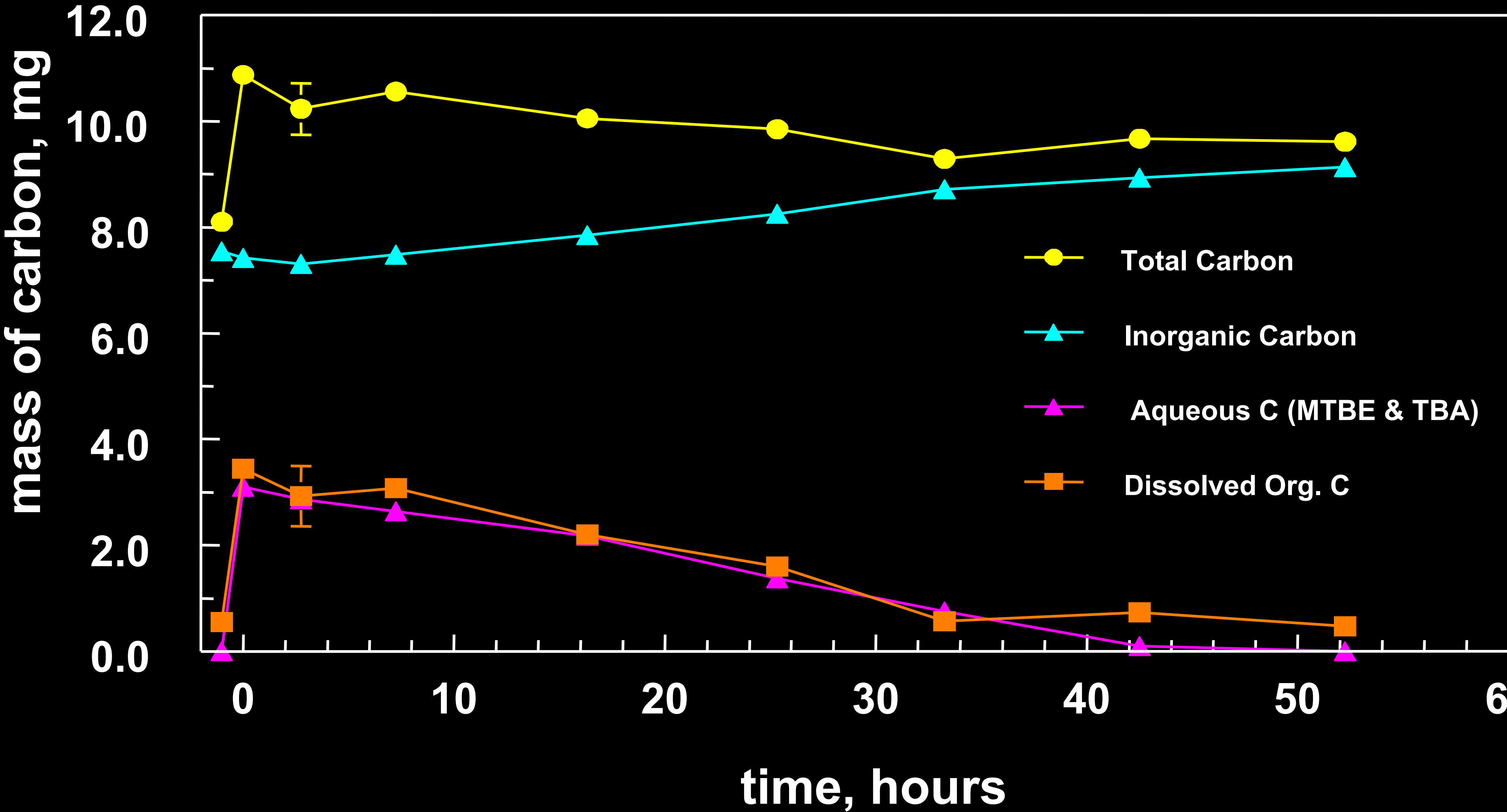
US EPA ARCHIVE DOCUMENT

TBA Spiked
0.65 mM



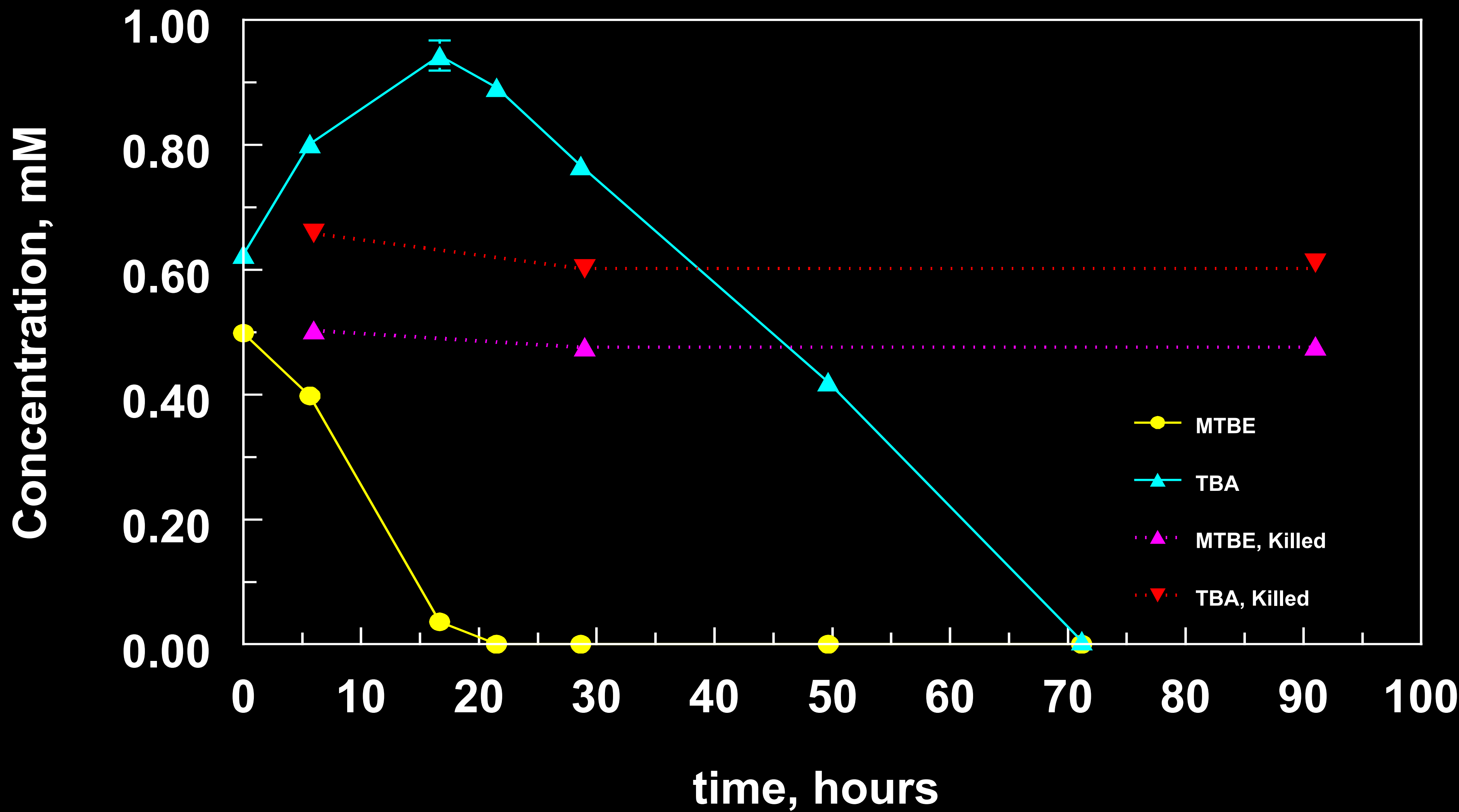
US EPA ARCHIVE DOCUMENT

TBA-Spiked Serum Bottle: 0.65 mM

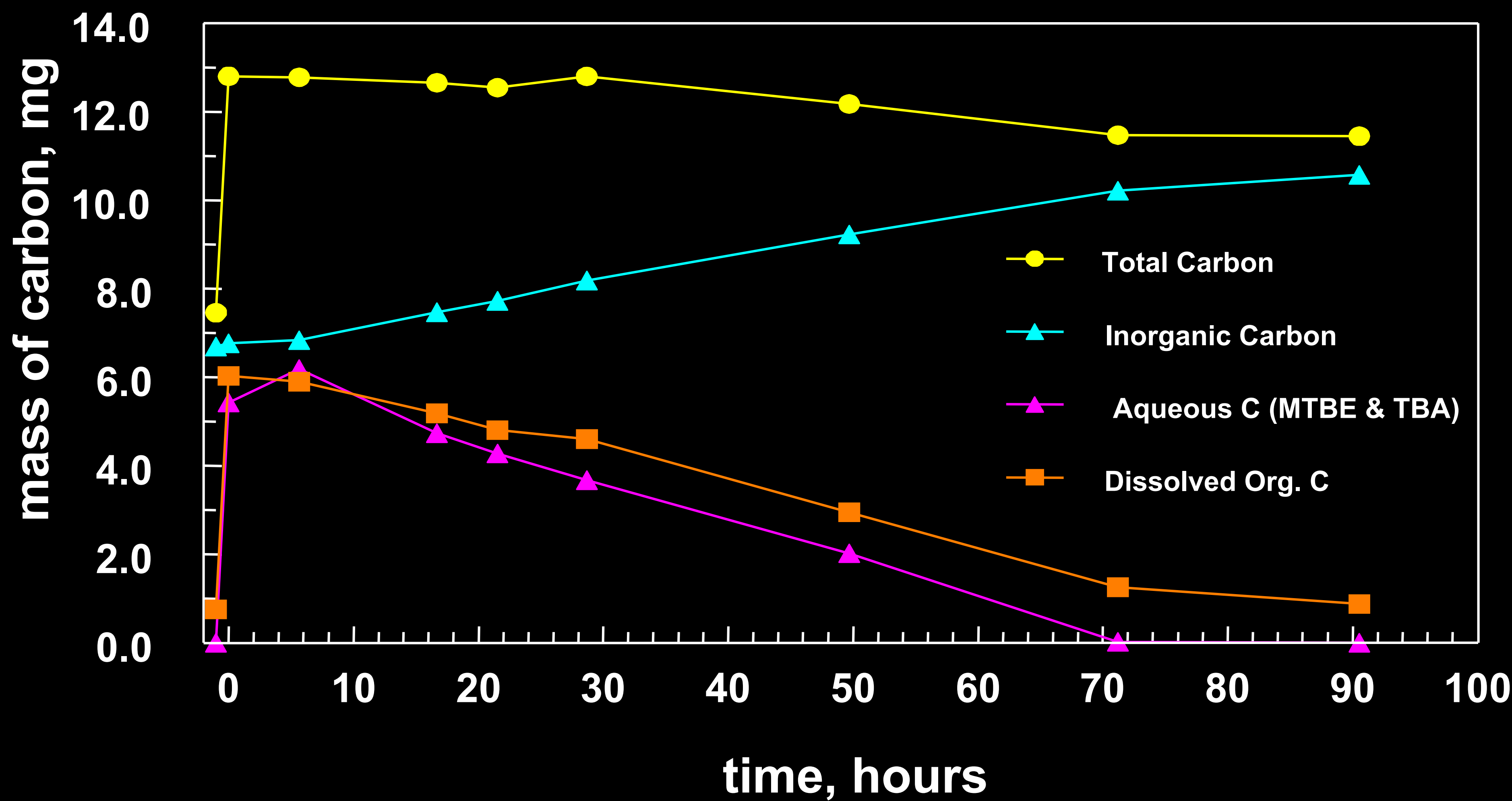


US EPA ARCHIVE DOCUMENT

MTBE & TBA Spiked Serum Bottle
0.5 mM MTBE, 0.60 mM TBA



MTBE/TBA Spiked Serum Bottle



US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT

Summary of Serum Bottle Experiments

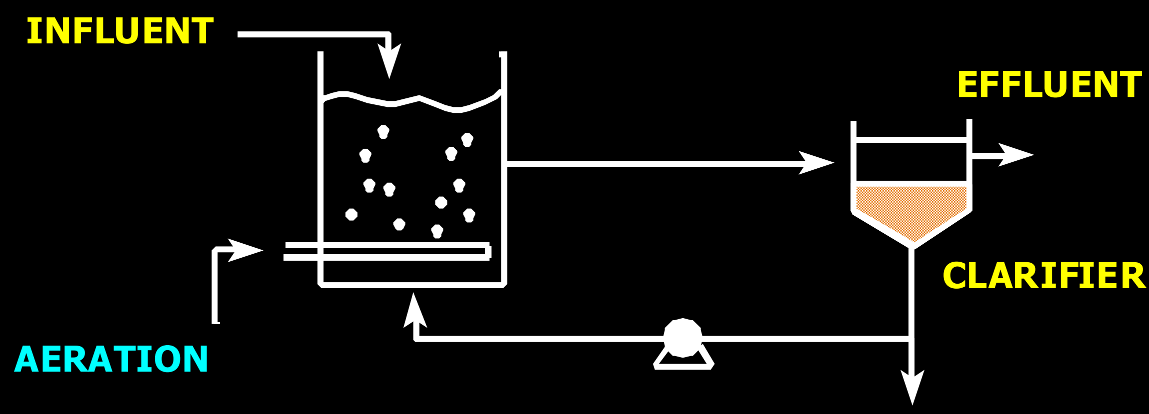
- Complete carbon balance obtained: all MTBE converted to CO_2 and H_2O with no buildup of intermediates
- As MTBE degrades, TBA first increases, then declines to undetectable levels
- Rate-limiting step is mineralization of TBA

US EPA ARCHIVE DOCUMENT

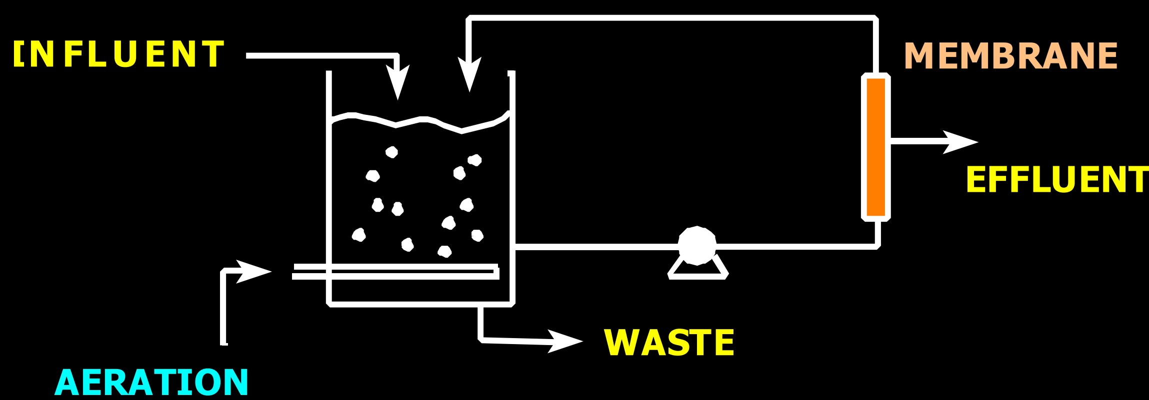
Performance of Membrane Bioreactor

US EPA ARCHIVE DOCUMENT

MBR vs. Activated Sludge



AS SYSTEM



MBR SYSTEM

US EPA ARCHIVE DOCUMENT

Characteristics of Membrane Bioreactors (MBR)

- **Advantages:**
  - Close control of solids
  - Extremely high effluent quality
  - Compact design
- **Disadvantages**
  - Fouling
  - Cost (both capital and operating)
  - High shear stress

US EPA ARCHIVE DOCUMENT

Treatment Approach Used

- Ultrafiltration membrane
  - Tech-Sep Kerasep™ (Rhone Poulenc, France)
  - External cross-flow, tubular ceramic membrane
  - Pore size = 0.02 μm (300 kDaltons)
  - Total surface area of 0.085 m^2
  - Volume of reactor = 6 L
  - Operated at 6 L/h or 1 h HRT
- Mixed culture from previous bench top porous pot chemostats fed MtBE
- Feed water = dechlorinated Cincinnati tap water

US EPA ARCHIVE DOCUMENT

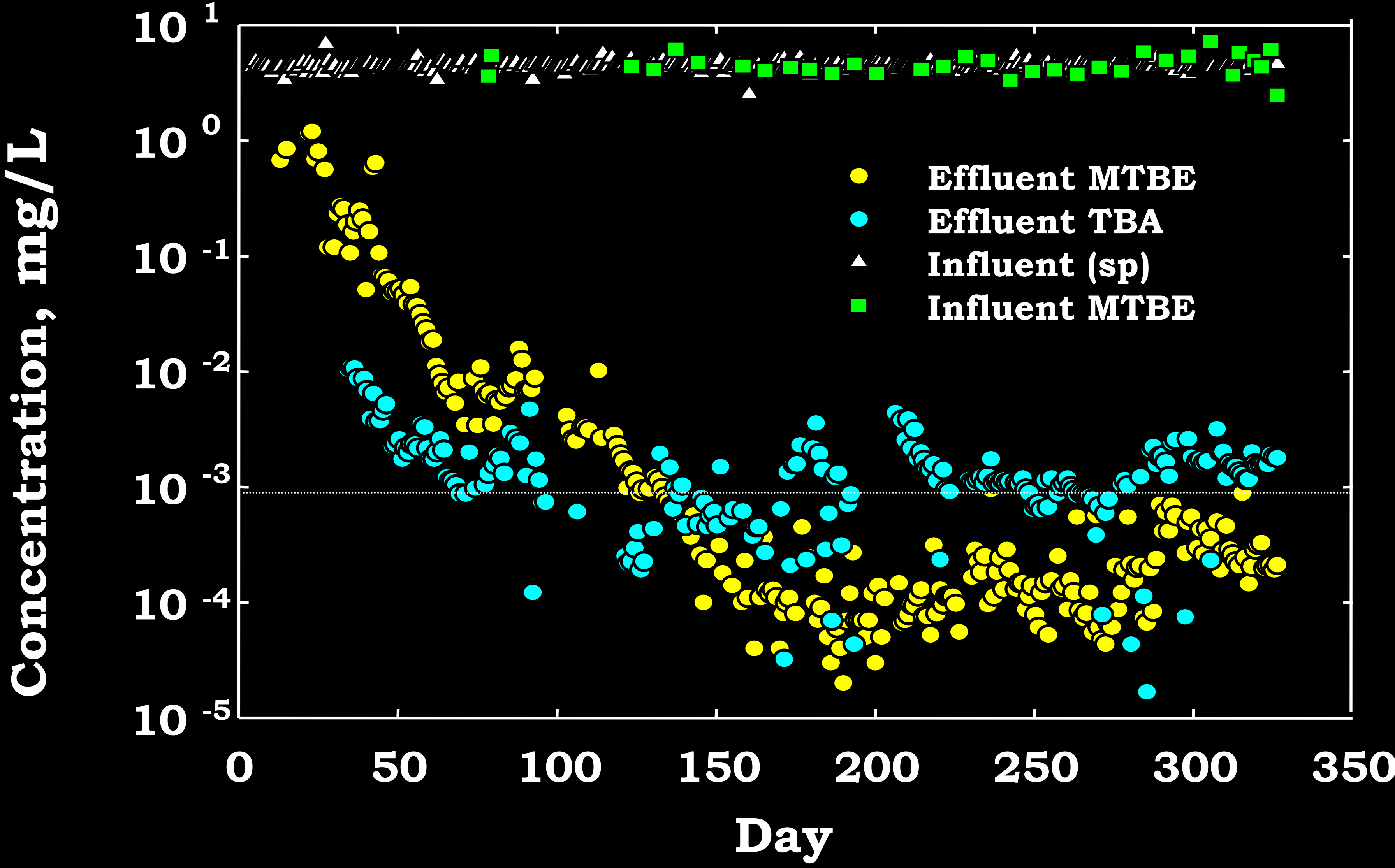
Summary of Operating Conditions

• Feed MtBE concentration	5 mg/L
• Hydraulic Retention Time	1 hour
• Temperature	18-20°C
• pH	7.2-7.8
• Dissolved oxygen	> 3 mg/L
• Solids retention time	>100 days

MBR Performance Results

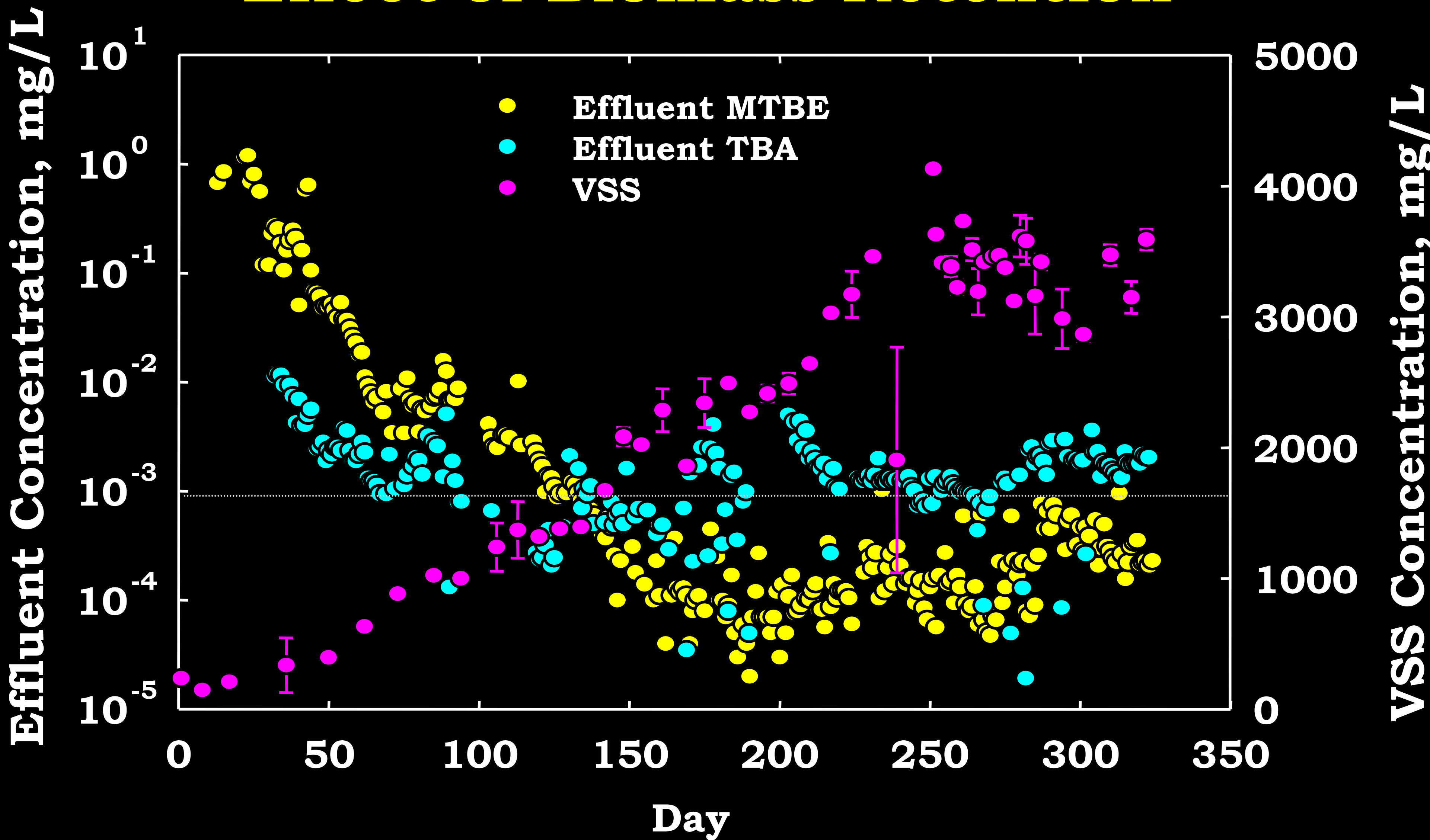
US EPA ARCHIVE DOCUMENT

MBR Performance, 5 mg/L MtBE in Feed



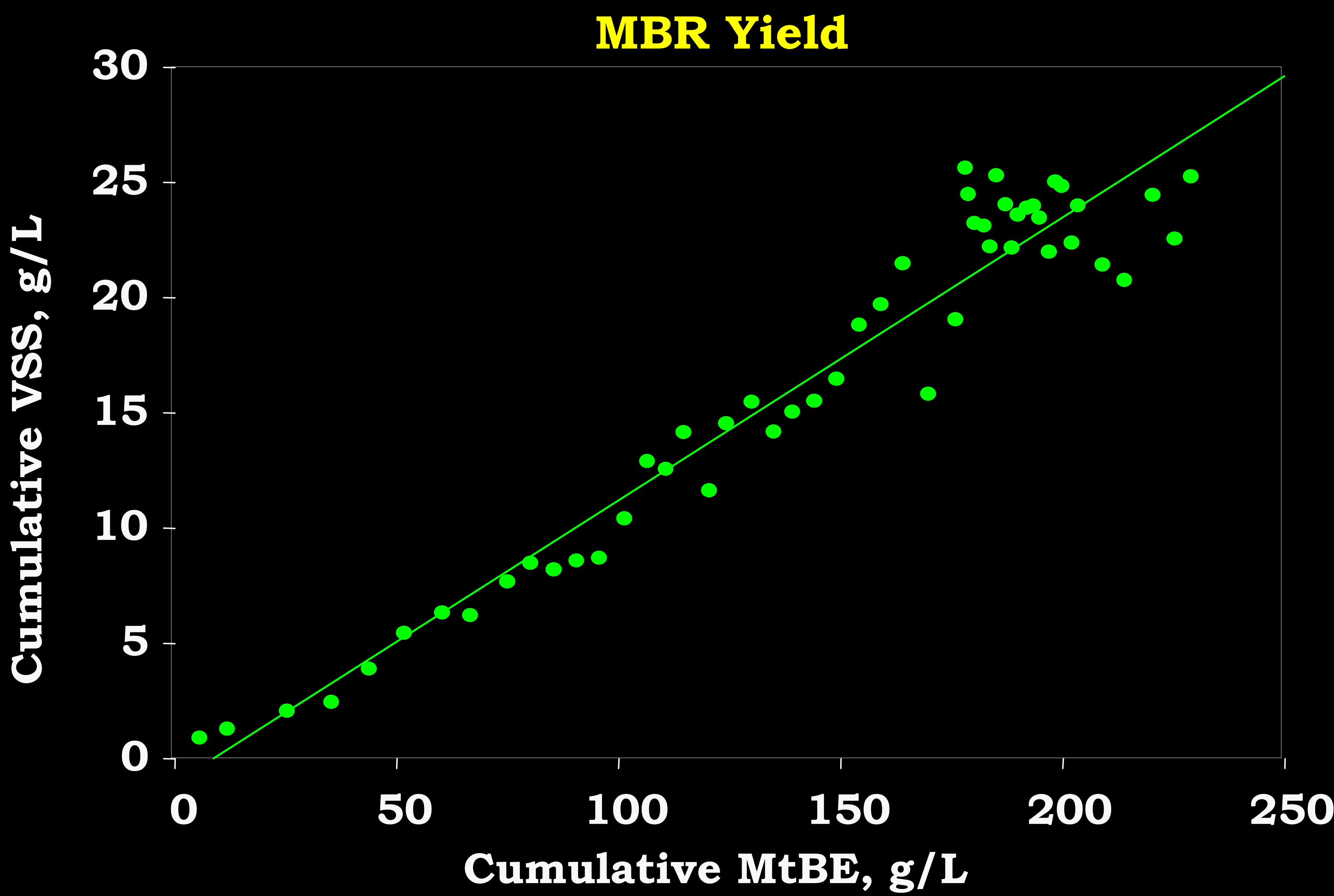
US EPA ARCHIVE DOCUMENT

Effect of Biomass Retention

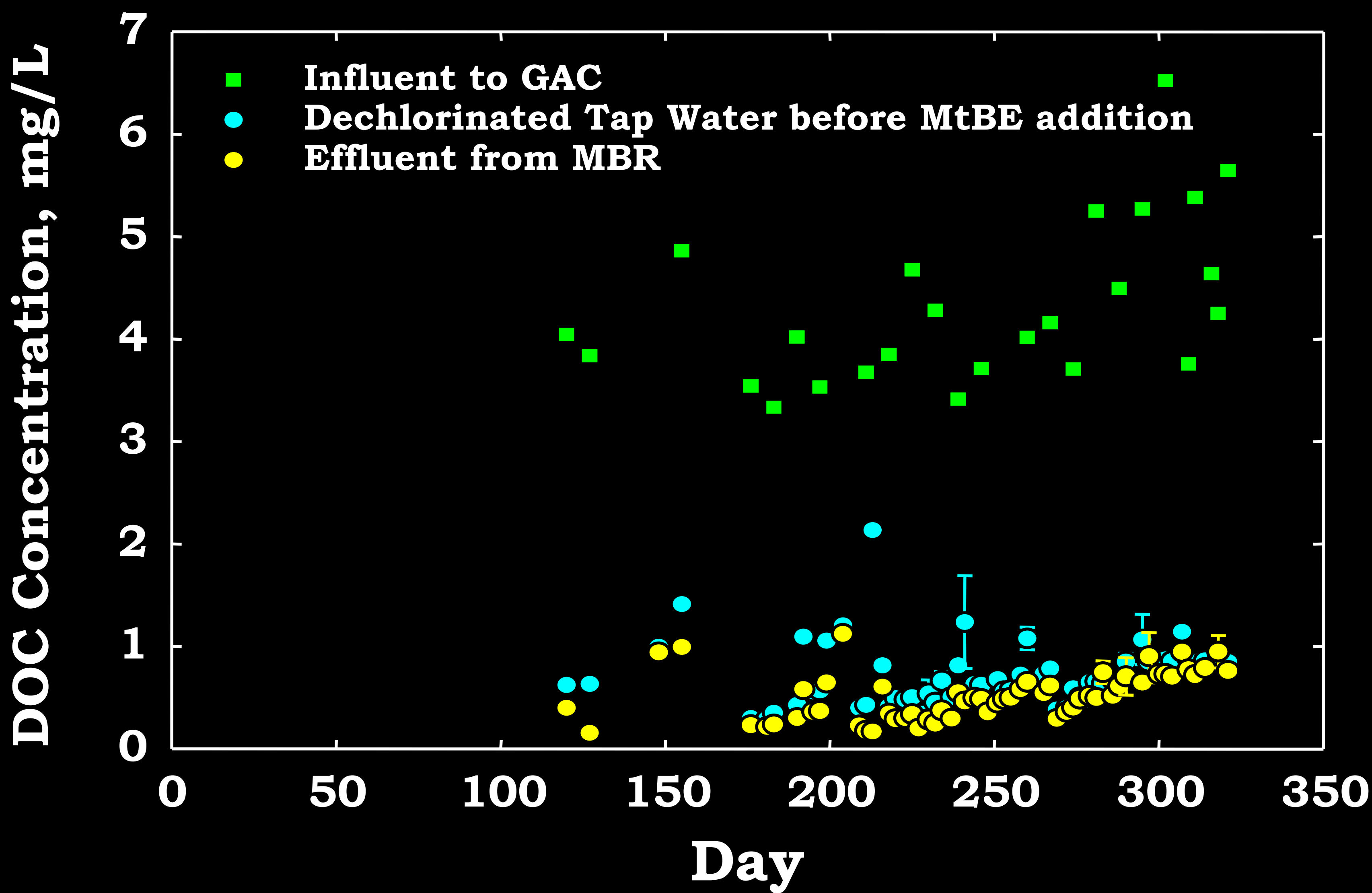


US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT



Dissolved Organic Carbon



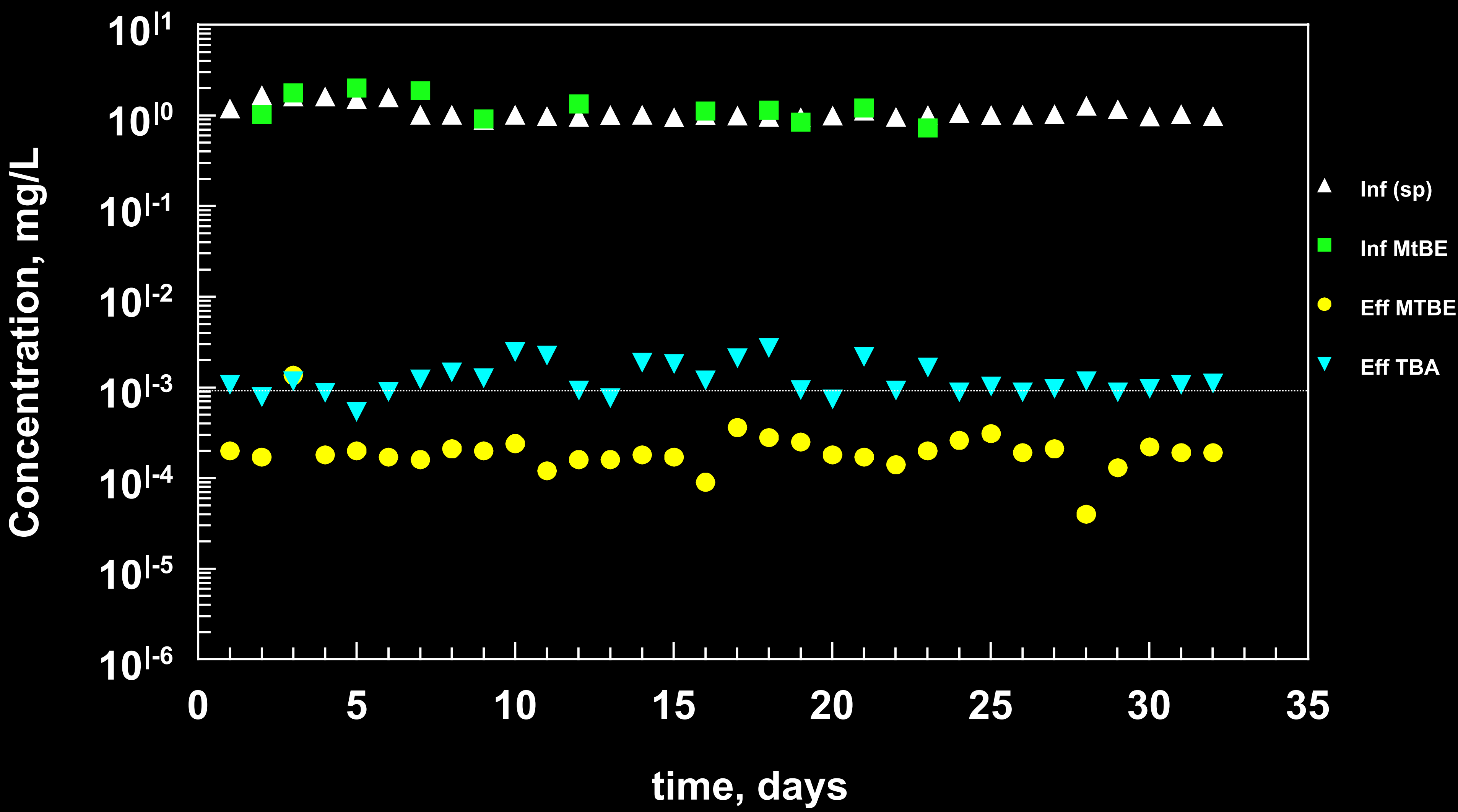
US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT

MBR Performance When Influent MtBE Reduced to 1 mg/L

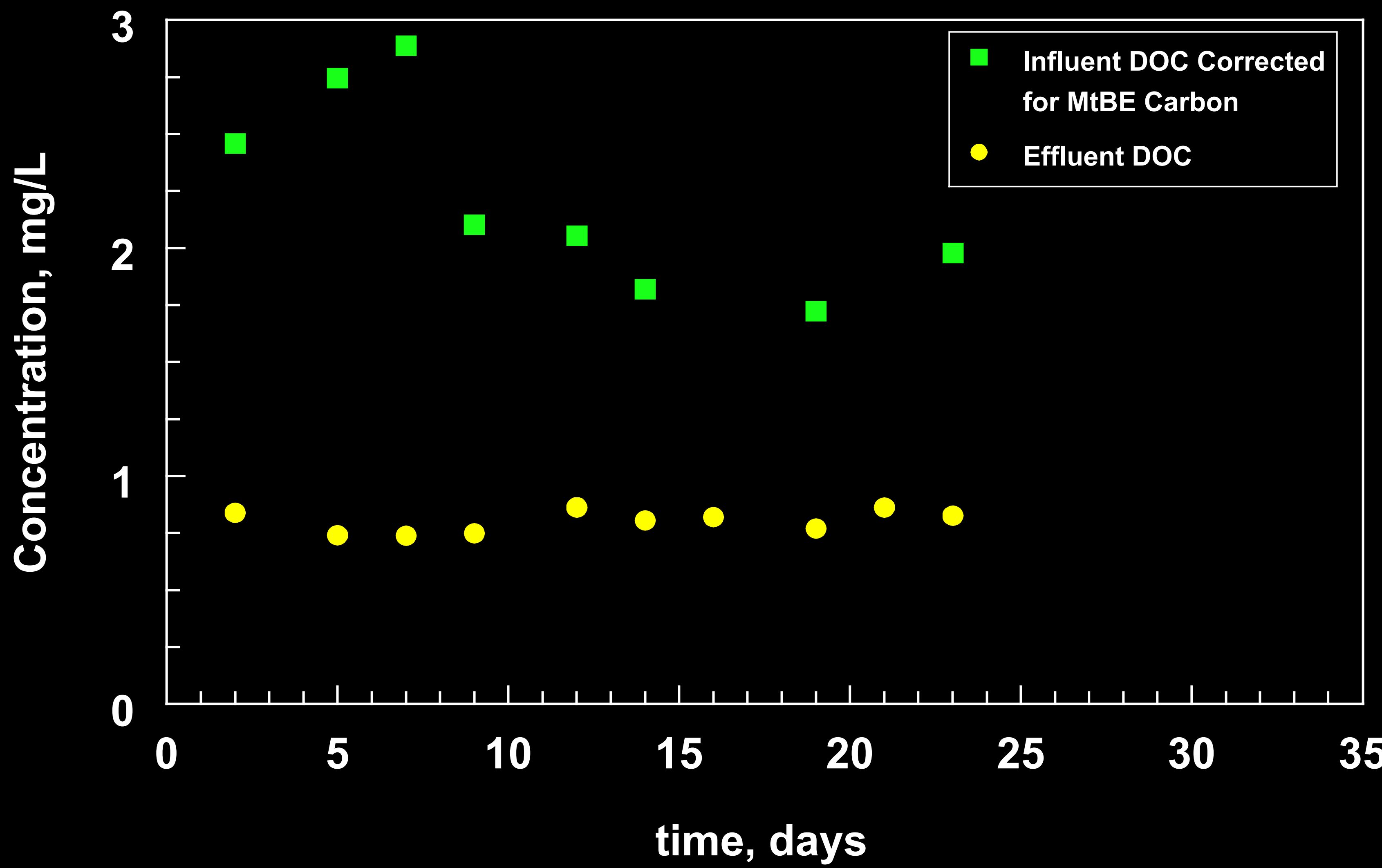
US EPA ARCHIVE DOCUMENT

MBR Performance, 1 mg/L MtBE in Feed



US EPA ARCHIVE DOCUMENT

DOC in Effluent Fed 1 mg/L MtBE



US EPA ARCHIVE DOCUMENT

Identification of Active Microbial Species in MBR

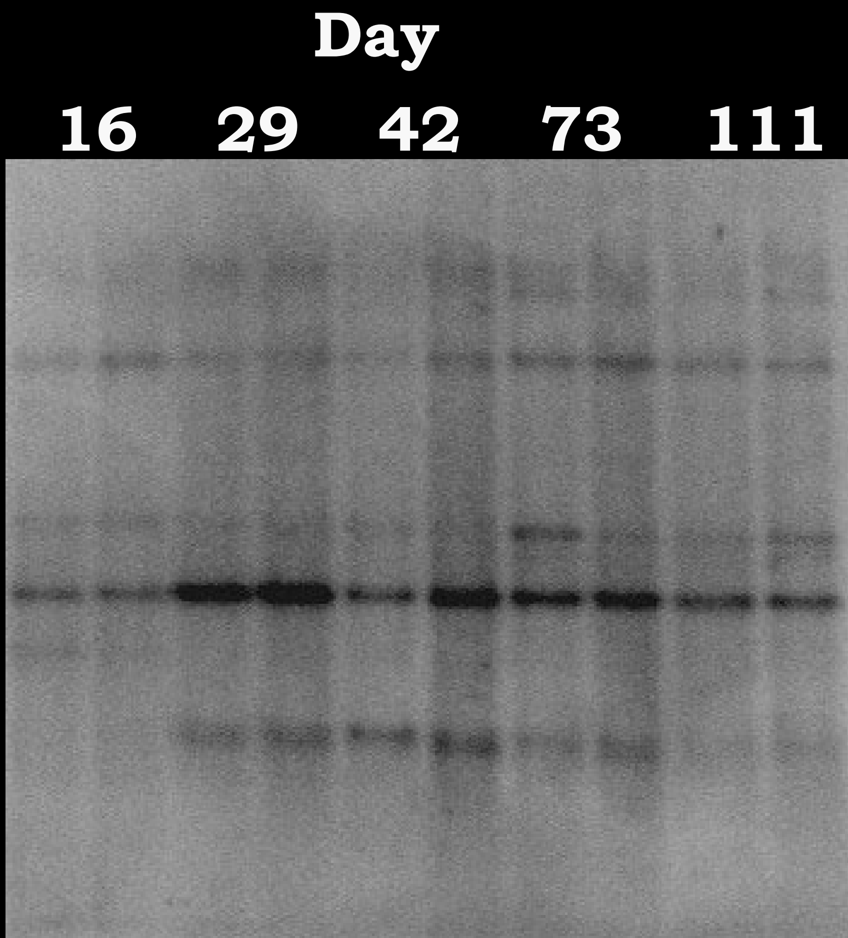
US EPA ARCHIVE DOCUMENT

Denaturing Gradient Gel Electrophoresis (DGGE)

- Molecular tool targeting 16S rDNA
- Resolves DNA from mixed culture based on melting and electrophoretic mobility behavior
- Banding pattern gives a snapshot of community structure
- One band ~ one microorganism

US EPA ARCHIVE DOCUMENT

Bacterial Identification by DGGE Pattern in MBR



DGGE Band	Microorganism
3	*Nitrospina*
5	*Sphingomonas*
6	*Sphingomonas*
8	*Sphingomonas*
9	*Sphingomonas*
Porous Pot Culture	*Flavobacteria-Cytophaga*

US EPA ARCHIVE DOCUMENT

Characteristics of *Cytophaga-Flexibacteria* Found in the Porous Pot Systems

- Dominant in activated sludge systems
  - Good floc-formers, settle readily
- Implicated in biodegradation of PAHs, phenols, and other substituted aromatics
- Abundant in organically rich, not oligotrophic, environments
  - Presence in groundwater not documented
    - Could partially explain why field studies have been so inconsistent in observing biodegradation of MTBE in aquifers

US EPA ARCHIVE DOCUMENT

Characteristics of *a-Proteobacteria*

- Predominant species in MBR
- *Sphingomonas* spp.
  - Known degraders of complex substrates like PCP
  - Often detected in subsurface samples
  - Can readily attach to surfaces
  - Highly hydrophobic, good oil degraders
  - Not subject to shear stress as are *Cytophaga*
- Since only present in porous pot reactors with dual substrates, most likely responsible for biodegradation of the alternate substrate

US EPA ARCHIVE DOCUMENT

Bench Top Study of Millville, NJ Superfund Site Groundwater

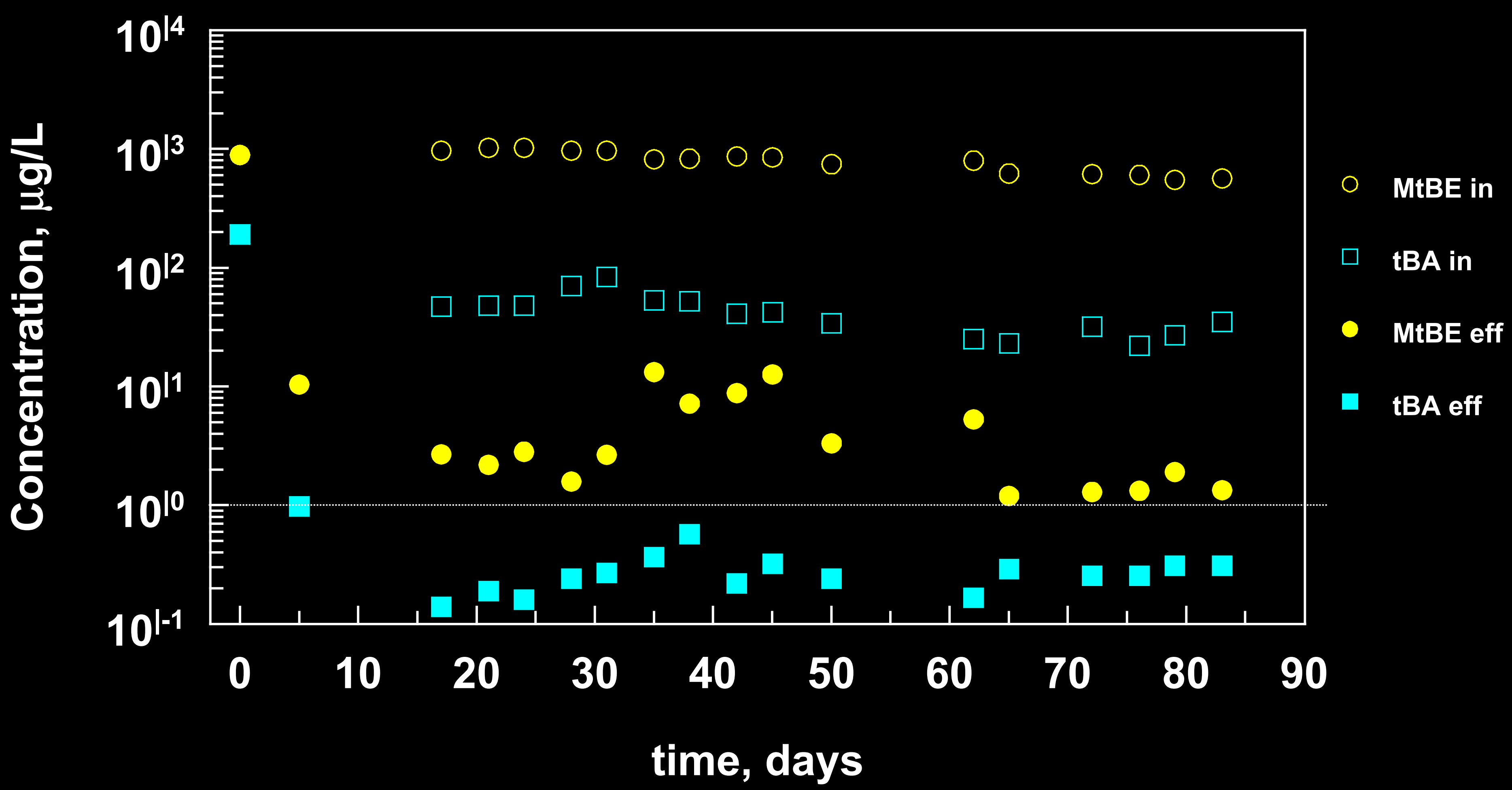
US EPA ARCHIVE DOCUMENT

Millville, NJ Study

- MGP site with co-mingled plume of PAHs and fuel hydrocarbons from gasoline spill
- Groundwater shipped every month as feed to porous pot chemostats
- Reactors inoculated with acclimated MtBE cultures and oil degraders
- Reactors operated at 2 different flow rates: 4.8 and 8.5 L/d

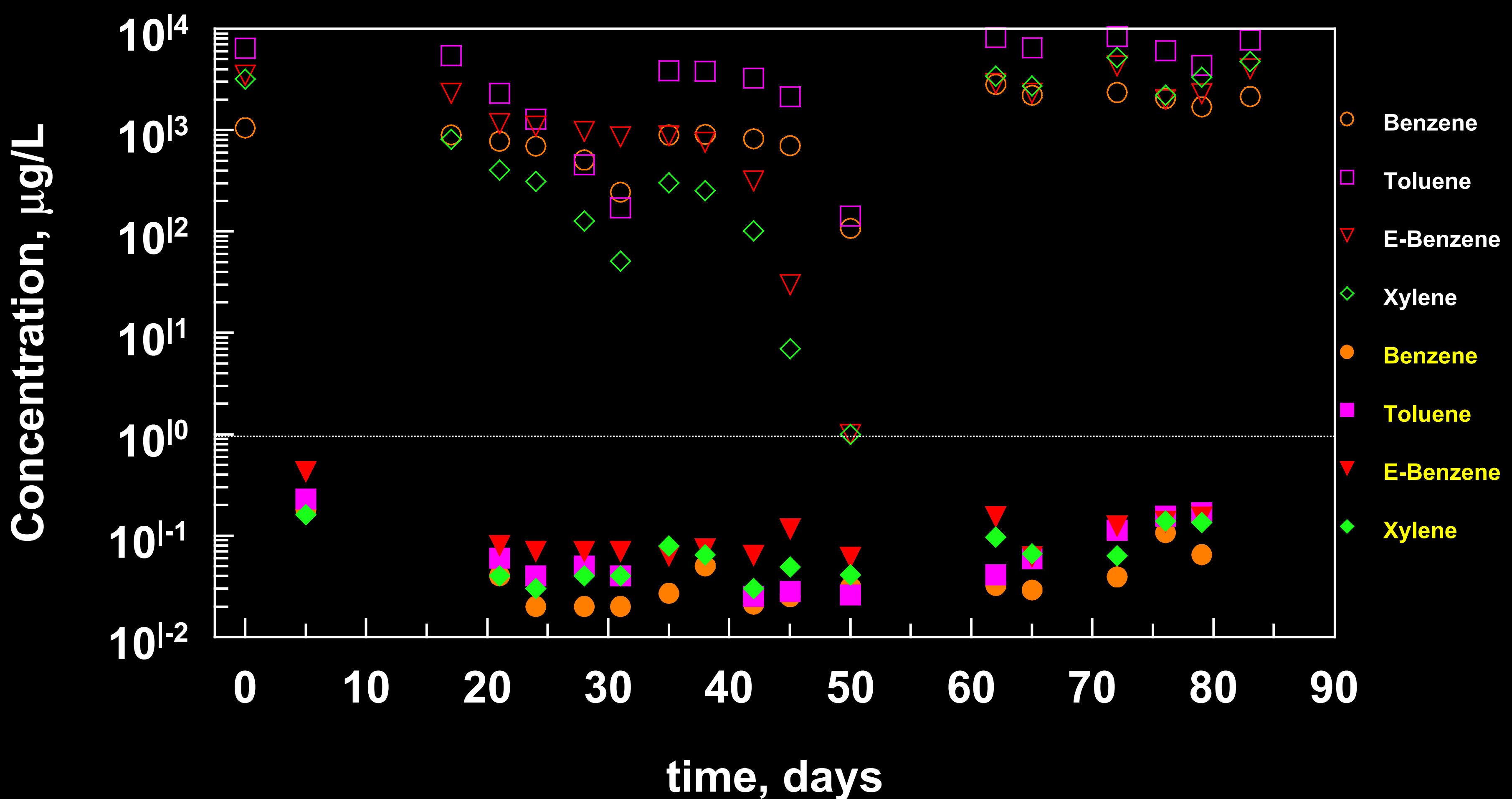
US EPA ARCHIVE DOCUMENT

MtBE and tBA Degradation



US EPA ARCHIVE DOCUMENT

BTEX Degradation



Pilot-Scale Biomass Concentrator Reactor (BCR) Performance

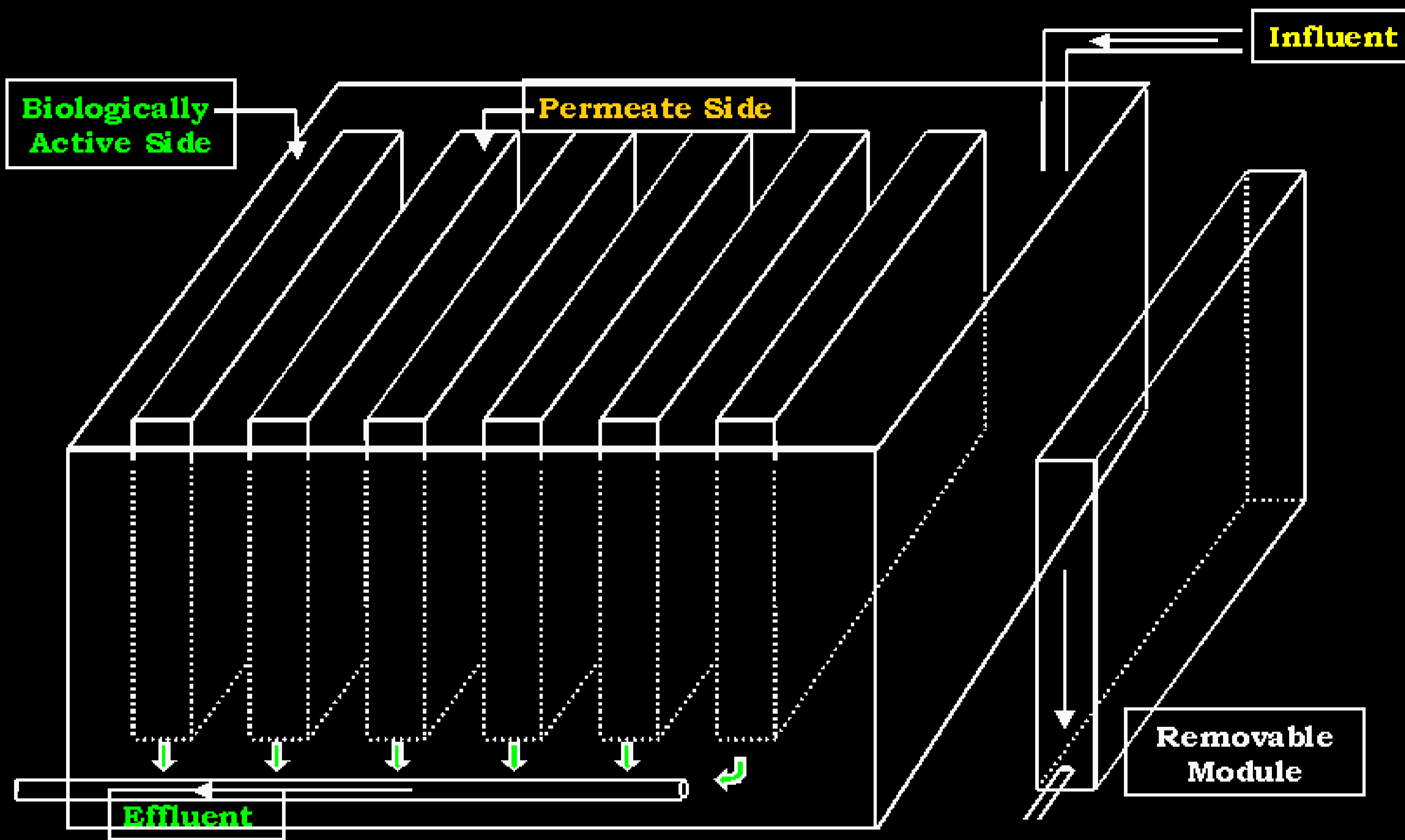
US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT

Description of BCR

- 0.5 m^3 capacity system built last year (1.0 m^3 total volume)
- Applied for joint UC/EPA patent
  - Design conceptually based on porous pot reactor
    - Much higher surface area, allowing for greater flow rates under gravity
    - Flow rate of 2,500 L/d (4.8 h HRT)
    - Started up in May, 2001
    - Producing effluent MtBE and tBA concentrations of ~1 μg/L since September, 2001 (5 mg/L in feed)
    - Will increase flow to 5,000 L/d (2.4 h HRT) and reduce MtBE in feed by half
    - Planned for use in a field study at Millville, NJ next year

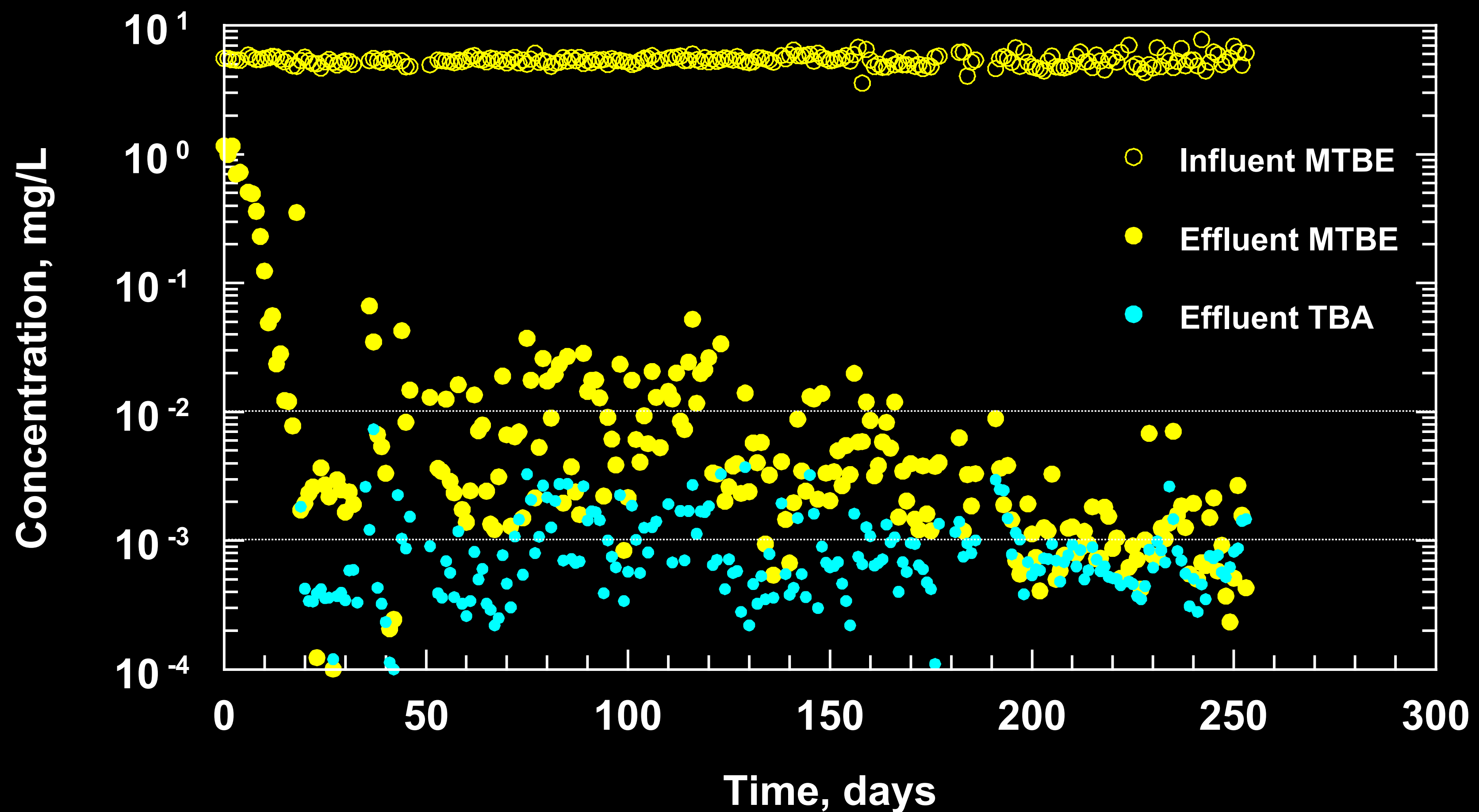
Schematic Diagram of Biomass Concentrator Reactor



US EPA ARCHIVE DOCUMENT

US EPA ARCHIVE DOCUMENT

Biodegradation of MtBE in BCR



US EPA ARCHIVE DOCUMENT

Economic Evaluation of *Ex-Situ* Reactors

- Comparison of MBR and BCR with Air Stripping
  - Assumptions:
    - 2 mg/L MtBE influent
    - 5 μg/L MtBE effluent
    - 3 GROUNDWATER flow rates (0.1, 0.3, and 1.0 mgd)
    - Air stripping equipped with GAC off-gas treatment

US EPA ARCHIVE DOCUMENT

Economic Evaluation of *Ex-Situ* Reactors

Cost of *ex-situ* treatments, $/1000 gal*			
Flow	Stripping	MBR	BCR
0.1 mgd	2.11	1.76	1.72
0.3 mgd	0.88	0.93	0.91
1.0 mgd	0.41	0.54	0.76

*Estimates by Richard Scharp

US EPA ARCHIVE DOCUMENT

Economic Comparison

- BCR had not been optimized when the cost analysis was done (costs based on prototype configuration only)
  - Preliminary studies indicate that the BCR can be operated at substantially lower HRTs, which will result in greatly reduced costs
- Thus, biotreatment appears to be cost competitive with air stripping

US EPA ARCHIVE DOCUMENT

Conclusions: Ex-Situ Biotreatment

- Porous Pot Chemostat and Batch Studies
  - Maintaining high biomass, aerobic conditions, and pH between 7.2 and 7.7 key for consistent performance
  - MtBE and tBA degradation is neither positively nor negatively affected by presence of BTEX
    - All compounds were degraded to << 1 μg/L whether MtBE, tBA, or BTEX were sole carbon sources or in combination

US EPA ARCHIVE DOCUMENT

Conclusions: MBR Study

- Effluent quality excellent and consistently below California advisory limits (5 μg/L)
  - Average 0.33 μg/L MtBE
  - Median 0.18 μg/L MtBE
- DOC effluent quality comparable to dechlorinated Cincinnati tap water
- Low cellular yield (0.12 g cells/g MTBE)
- Good performance at 5 and 1 mg/L in feed
- Several advantages over conventional treatment

US EPA ARCHIVE DOCUMENT

Collaborators at the University of Cincinnati

John R. Morrison

Maher Zein

Gregory J. Wilson

Amy Pruden

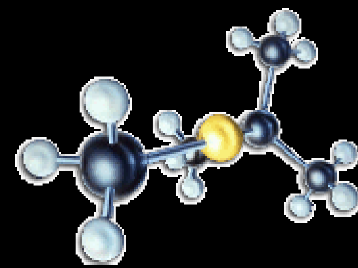
Marie A. Sedran

Makram T. Suidan

Department of Civil & Environmental Engineering

University of Cincinnati

US EPA ARCHIVE DOCUMENT



THANK YOU