Fiber Attachment Module Experiment (FAME): Using a Multiplexed Miniature Hollow Fiber Membrane Bioreactor Solution for Rapid Process Testing

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Introduction:

- Bioreactor research is mostly limited to continuous stirred-tank reactors (CSTRs) which are not an option for microgravity (μg) applications due to the lack of a gravity gradient to drive aeration as described by the Archimedes principle. Bioreactors and filtration systems for treating wastewater in μg could avoid the need for harsh pretreatment chemicals and improve overall water recovery.
- Solution: Membrane Aerated Bioreactors (MABRs) for μg applications, including possible use for wastewater treatment systems for the International Space Station (ISS).
- Small 1-L volume MABRs, do not lend themselves to rapid testing capacities. To address this, we designed 125-mL, rectangular reactors, to create the Fiber Attachment Module Experiment (FAME) system.
  - Each FAME module (i.e., reactor) is self-contained.
  - Easy to plumb with pumps for continuous recycling of fluids/feeding and sensors for parameter monitoring similar to their larger counterparts.
- Application: Rapid Biofilm Attachment Studies
  - Goal: Achieve rapid reactor startup and biofilm attachment (based on carbon oxidation and nitrification of wastewater).
  - Multi-factorial test with data collected over short to study and improve bioreactor performance.
  - Thirteen fiber surface treatments and three inoculation sources tested.

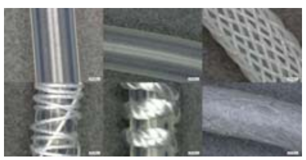

Figure 1: Various fiber treatments tested:
Top: PE (5 sec), 1500-Grit Sanding, Cable Sheath
Bottom: Cotton thread, Silk Thread, Spider Silk

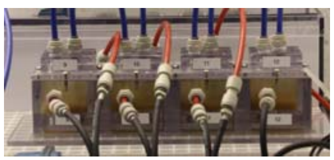

Figure 2: FAME rack containing four individual modules during fiber testing.

Materials and Methods:

Reactor Setup:

- Multiple polycarbonate FAME racks, each containing 4 modules
- Peristaltic pumps used for fluid recirculation (1025 reactor volumes per day)
- Standard breathing air used as reactor oxygen source
- Modules contained in a Controlled Environment Chamber (CEC) at 25°C in darkness

Fiber Treatments:

Table 1: Various fiber treatments studied

Etching	Polymer Coating	Mechanical	Scaffolding
3% HF	Polyacrylic Acid (PAA)	Abrasion via Sand Paper	Nylon Cable Sheaths
Fluoroetch® (FE)	Polystyrene (PS)	(Various Grits)	Silk Thread
Piranha Etch (PE)			Cotton Thread
			Spider Silk

Inoculation Sources:

- Effluent from a KSC single-stage carbon oxidation reactor (R3)
- Effluent from a Texas Tech University (TTU) single-stage MABR
- Activated sewage sludge from a local septic tank

Evaluation Parameters:

- Bulk fluid analysis weekly: NH_3, NO_2^-, NO_3^-, total cell counts
- Sample volumes replaced with reactor feed (real urine solution)
- End of experiment analysis: total cell counts for biofilms attached to fibers
- Oxygen permeation of modified fibers compared to controls

Biological Results:

- TTU inoculum showed significantly higher attached cell counts compared to R3 inoculum in all treatments except HF - 15 min.
- Highest cell counts overall were seen for TTU inoculum on FE – 18 sec & HF – 12 sec fibers.
- R3 counts were the lowest; no significant difference between TTU and Septic.
  - R3 inoculum was effluent from well-established bioreactor; selecting for planktonic cells in the effluent could explain lower attachment.
- Septic tank inoculum was compared to test a local and consistent source for future studies if comparable to TTU inoculum results.

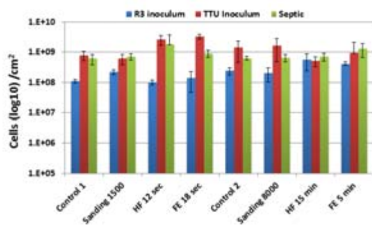

Figure 3: Attached cells on fibers with six surface treatments and three different inoculum sources. Error bars represent standard deviation (n=6).

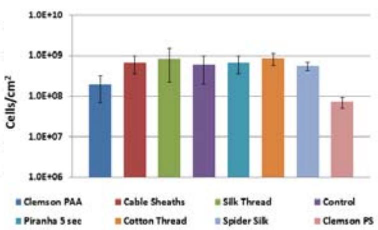

Figure 4: Attached cells on additional fiber treatments tested with septic tank inoculum. Error bars represent standard deviation (n=6).

- Bulk fluid counts lowest in TTU-inoculated modules and highest in septic-inoculated modules.
- Two fiber treatments with highest number of attached cells ***regardless*** of inoculum:
  - Fluoroeth (18 sec)-treated
  - HF (12-sec)-treated
- General trend: higher number of attached cells in a module correlated to lower bulk fluid cell counts.
- None of the additional treatments tested solely with septic tank inoculum exhibited increased biofilm attachment over the FE 18-sec and HF 12-sec treatments
- Polystyrene polymer coating treatment resulted in significantly ***lower*** fiber cell counts compared to other treatments.

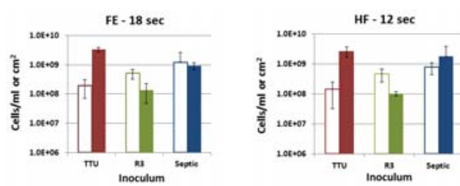

Figure 5: Cell counts in bulk fluid (outlined bar) and attached (solid bar) in modules with FE 18-sec and HF 12-sec treatments. Error bars represent standard deviation (n=6).

Figure 6: Biofilms present on samples after fiber harvest for TTU inoculum with control (Left) fibers and FE 18-sec-treated (Right) fibers showing drastic cell attachment differences.

Chemical Analysis Results:

- Limited chemical analysis performed due to nature of experiment; bulk fluid sam collected weekly for pH, NH_3, NO_2^-, and NO_3^-.
  - pH for reactor inocula and feed ranged between 6.3 and 6.5; as experim progressed, pH rose rapidly to between 8 and 9 due to urea hydrolys ammonia.
  - Buildup of NH_3 indicative of no nitrification; supported by absence of ni and nitrite species. Attempt made to lower pH to induce nitrification at 40; attempts were unsuccessful.
- Results show that formed biofilms were heterotrophs responsible for ca oxidation/urea hydrolysis. Further fiber development may be required to b attract nitrifying biofilms.

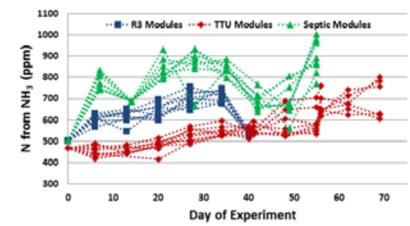

Figure 7: Ammonia trends over progression of experiment for all inocula.

Oxygen Permeation Results:

- PDMS fibers traditionally used for their high O_2 permeation, su mechanical/chemical resistance, and toleration of high intra-membrane pressure
- Modification of PDMS for better biofilm attachment must not sacrifice properties, especially O_2 permeation.
- O_2 permeation comparisons completed for five of the modification processes:
  - Mass transfer coefficient, K, derived for each membrane type.
  - Describes resistance between gas phase, membrane, and liquid boundary layers.
  - Increased K equates to increased O_2 transfer across the membrane.
- None of the modifications exhibited statistically different K values, showi permeation was not inhibited by the treatment.
- Further shows that differences in biofilm attachment are not due to changes permeation.

Table 2: Mass transfer coefficients for control and treated fibers

Treatment	Max K (cm s^{-1})	Average K (cm s^{-1})
Control	0.00171	0.00115
FE – 18 sec	0.00173	0.00107
PE – 5 sec	0.00148	0.00082
1500-Grit Sanding	0.00147	0.00105
8000-Grit Sanding	0.00146	0.00109
HF – 5 min	0.00178	0.00096

Future Work:

- Future experiments involving FAME hardware will include further testing of modifications, inoculum choice, reactor feed composition, reactor poisoning eff biofilm repulsion treatments, and more.
- Fiber modifications which have shown promise will also undergo further verifica testing.
- Modifications to current hardware include the addition of external probe tank continuous monitoring of pH and/or dissolved oxygen (DO) within the module allow for improved process controls in hopes of gaining nitrification in the reactor
- The value of this hardware is demonstrated for rapid testing of numerous param in parallel. FAME racks may also be utilized for future reactor hibernation biofilm-development time course studies.