Dormancy and Recovery Testing for Biological Wastewater Processors

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Introduction

• Resource recovery and recycling waste streams to usable water via biological water processors is a plausible component of an integrated water purification system.

• Biological processing as a pretreatment can reduce the load of organic carbon and nitrogen compounds entering physiochemical systems downstream.

• Aerated hollow fiber membrane bioreactors, have been proposed and studied for a number of years as an approach for treating wastewater streams for space exploration.
Challenges

- **Transport**
  - of the system to its destination, whether that be the International Space Station or beyond

- **Storage**
  - the system for various durations of nonuse (e.g., if the ISS or a beyond Earth habitat was to be unmanned for a period of time) with rapid recovery of that system when operations are restarted. Few studies have been done to examine a intentional stop, store, and start operating regime as proposed, which could introduce stresses to the microbial communities essential for wastewater processing. However, numerous

- **Restoration**
  - how terrestrially established bioreactors could be transported to space and stored for periods of time between operations.

- **Studies**
  - have been done, some using small scale biological reactors, to address the impact of chemical waste streams, changes in feed composition and potential starvation conditions due to industrial process shut downs and less then optimal environmental conditions on wastewater treatment plants and the microbial communities involved in conversions of carbon and nitrogen compounds. Strategies to restore a functioning microbial community to a wastewater treatment process can be
Goals

• Explore several dormancy processes for established bioreactors/biofilms to determine optimal storage and recovery conditions.
  – storage temperature: ambient (25°C) and cold (4°C)
  – Wet or dry
• Isolation of the established biofilm microbial community from an operational standpoint. i.e., simply turning off power, pumps, feed, etc. without the need for extended storage preparations).
Experiments

• Storage conditions:
  – 1 liter reactors
  – 48 day old biofilm
  – 4 week storage period.

<table>
<thead>
<tr>
<th>Reactor</th>
<th>Storage Conditions</th>
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<tbody>
<tr>
<td>11</td>
<td>4°C, No Bulk Fluid</td>
</tr>
<tr>
<td>12</td>
<td>25°C, No Bulk Fluid</td>
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<tr>
<td>13</td>
<td>4°C, With Bulk Fluid</td>
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<tr>
<td>14</td>
<td>25°C, With Bulk Fluid</td>
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• Long Duration Storage Test
  – 3.8 liter reactor
  – 13 month old biofilm
  – 7 month storage at 4°C/no bulk fluid.
  – Compare biofilm composition to 69 day old biofilm; never stored
Recovery

• Recovery methods were chosen to represent a possible scenario for a reactor restart in microgravity or in a beyond-Earth mission application.
  – To recover the reactors stored with their bulk fluid full strength wastewater at a 3.79-day residence was immediately introduced to the reactors and allowed to slowly dilute into the bulk fluid over time.
  – To recover those stored without their bulk fluid, the same feed and residence time was used and reactors were allowed to slowly fill up the bulk fluid volume over time.
  – Successful recovery was evaluated based on each individual reactor’s ability to regain or surpass its predormancy performance metrics including TOC, urea and ammonia removal (nitrification/denitrification)
Microscopic Analysis of Biofilm

• To determine the ratio of live/dead bacteria before and after storage.
• To determine the presence and relative quantity of AOB/NOB before and after storage.
Results

C. Urea Hydrolysis

D. TOC Removal
After 29 days of operation, average performance metrics were as follows: 75.0 ± 3.7% ammonia removal, 62.1 ± 5.1% ammonia removal, 99.3 ± 0.4% urea hydrolysis, and 95.0 ± 0.5% TOC removal.
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Conclusion

• Dormancy-recovery cycles are an important feature of bioreactor systems, yet very little is known about the microbial response to such processes in this specific application.

• In the 1-L MABR studies, it did not appear that any dormancy condition (with/without bulk fluid, 4°C/25°C) was better at preserving reactor performance metrics.

• Reactors recovered and even experienced improved performance, the recovery period of 20+ days is not satisfactory.

• Several factors may have contributed to such a long recovery period;
  – the biofilms in these studies were not well established (48 days of reactor operation), and may not have been robust enough to handle full wastewater feed directly after a four-week dormancy period.
  – the amount of nitrifying and denitrifying communities may not have been well enough established in the biofilms to provide adequate conversion rates.
Conclusion

• In the long duration dormancy experiment, fully established biofilms (13 months old) were able to recover from a 7-month dormancy period to steady state operation within 4 days.

• The maturity of the biofilms and the addition of a “buffer” bulk fluid to dilute the introduction of the full wastewater feed likely helped to speed up the recovery period.

• Future studies should include further long duration dormancy cycles at room temperature along with studies to correlate of recovery times and biofilm age/initial biofilm health.

• These studies successfully demonstrated that biological systems can survive periods of nonuse for future microgravity application.
Questions?