Augmenting anaerobic digestion of microalgal biomass

Anna Doloman
PhD Candidate
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What is anaerobic digestion and why to care?

Organic waste
(plant/algae biomass, wastewater, animal manure)

Microbial buffet

Biogas

🌟 Methane potential of USA – 7.9 million tones/year
🌟 5% of natural gas in the electric power
🌟 56% of natural gas in the transportation
Problem: how to efficiently digest algae?

- “Good” algae that polish wastewater and needs to be processed afterwards;
- “Bad” algae that cover surface waters and needs to be processed.

I have never eaten this stuff before!
Augmentation is a solution

We have a very efficient anaerobic granular sludge

from Sekiguchi Y. et al., 1999
Solving the problem: finding algae-loving bacteria

Logan City Wastewater Lagoons
Solving the problem: finding algae-loving bacteria

Logan City Wastewater Lagoons

Isolated: *Citrobacter*, *Alcaligenes* and *Pseudomonas* spp..
Experimental set-up
Specific Methanogenic Activity (SMA) test

+ self-digestion controls

9.6 gVSS/L of algae
19 gVSS/L of granules
10% algalytic bacteria
1:2 substrate:inoculum

**Algae came from the surface of a trickling filter polishing municipal wastewater
Results: after 70 days, at 35±2°C, at 100 rpm...
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**Results:** after 70 days, at 35±2°C, at 100 rpm...

11% increase due to algalytic mix.
Algae already figured things out...

A “clean” experiment on augmentation would be to use an axenic culture of algae.

But large scale industries will never do it. They deal with waste. Waste is contaminated.
How can we help those industries who need augmentation?

We can make a computer model to help predict the success of augmentation!
A computer model for augmenting anaerobic granules

Model is based on:
- Kinetics of substrate consumption (Monod, Haldane, Simple inhibition)
- Bacterial attraction towards substrate

Input parameters:
- Growth characteristics ($K_s$, $K_i$, $\mu_{max}$, $\mu_g$)
- Diffusivity of substrates/products
- Strength of chemotactic attractance

Glucose $\rightarrow$ Acetate $\rightarrow$ Methane

Laboratory image (Sekiguchi, 1999)
Simulated image from our model (40 days)

A computer model for augmenting anaerobic granules

Day 0-17: cellobiose, 1.5 g/L
Day 17-60: cellobiose and oleate

1) Cellobiose and oleate at 1.5 g/L
2) Cellobiose and oleate at 0.5 g/L
3) Oleate at 1.5 g/L
Conclusions and food for thoughts

✓ Algae digestion successfully augmented in batch conditions;

✓ Trial with axenic algal biomass is needed;

✓ When augmenting, remember to feed the existing bacteria, **BUT**, do not overfeed them!
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- Utah Science and Technology Agency (USTAR)
- USU Graduate Research and Collaborative Opportunities Grant

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Results: what’s up with the microorganisms?

![Bar chart showing the number of sequences for different microbial communities.]

- **Algae + Granules**: The chart shows the distribution of various microbial categories including Synergistetes, Proteobacteria, Aegiribacteria, Armatimonadetes, Actinobacteria, Tenericutes, Acidobacteria, Spirochaetes, Chloroflexi, Firmicutes, and Bacteroidetes. The chart indicates the relative abundance of each category in the Algae + Granules treatment.

- **Algae + Granules + Bacteria**: Similar to Algae + Granules, but with additional bacteria included.

- **Granules + Bacteria**: The chart focuses on the granules and bacterial components, highlighting different microbial categories.

- **Granules C**: This category likely represents a control or a specific condition not detailed in the text but shown in the graph.
Results: what’s up with the microorganisms?

- **Algae+Granules**
  - Other (less than 1%)
  - Synergistetes
  - Proteobacteria
  - Aegiribacteria
  - Armatimonadetes
  - Actinobacteria
  - Tenericutes
  - Acidobacteria
  - Spirochaetes
  - Chloroflexi
  - Firmicutes
  - Bacteroidetes

- **Algae+Granules+Bacteria**
  - Other (less than 1%)
  - Synergistetes
  - Proteobacteria
  - Aegiribacteria
  - Armatimonadetes
  - Actinobacteria
  - Tenericutes
  - Acidobacteria
  - Spirochaetes
  - Chloroflexi
  - Firmicutes
  - Bacteroidetes

- **Granules+Bacteria**
  - Other (less than 1%)
  - Synergistetes
  - Proteobacteria
  - Aegiribacteria
  - Armatimonadetes
  - Actinobacteria
  - Tenericutes
  - Acidobacteria
  - Spirochaetes
  - Chloroflexi
  - Firmicutes
  - Bacteroidetes

- **GranulesC**
  - Other (less than 1%)
  - Synergistetes
  - Proteobacteria
  - Aegiribacteria
  - Armatimonadetes
  - Actinobacteria
  - Tenericutes
  - Acidobacteria
  - Spirochaetes
  - Chloroflexi
  - Firmicutes
  - Bacteroidetes
Results: what’s up with the microorganisms?

Augmented VS Non-augmented

- Increased number of polysaccharide and protein digesters (Bacteroidetes)
  More primary fermenters

- Number of Clostridia members shifted to those utilizing amino acids and sugars (Firmicutes)
  More secondary fermenters
<table>
<thead>
<tr>
<th>Algae+Granules</th>
<th>Algae+Granules+Bacteria</th>
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</thead>
<tbody>
<tr>
<td>Selenomonadales, Proteiniphilum <em>(Firmicutes)</em></td>
<td><strong>Hydrogenispora, Lutispora</strong> <em>(Firmicutes)</em></td>
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<tr>
<td>Syntrophomonas, Syntrophorhabdus <em>(Firmicutes)</em></td>
<td><strong>Syntrophobacter</strong> <em>(Firmicutes)</em></td>
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<td>Aminobacterium <em>(Firmicutes)</em></td>
<td><strong>Veillonellaceae</strong> <em>(Firmicutes)</em></td>
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<td>Christensenellaceae <em>(Firmicutes)</em></td>
<td><strong>Peptococcaceae</strong> <em>(Firmicutes)</em></td>
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<td>Sedimentibacter <em>(Firmicutes)</em></td>
<td><strong>Gracilibacteraceae</strong> <em>(Firmicutes)</em></td>
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<tr>
<td>Propionibacterales <em>(Actinobacteria)</em></td>
<td><strong>Cellulosimicrobium</strong> <em>(Actinobacteria)</em></td>
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<tr>
<td>Phycisphaerae <em>(Planctomycetes)</em></td>
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<tr>
<td>Geobacter <em>(Proteobacteria)</em></td>
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<td>Desulfovibrio <em>(Proteobacteria)</em></td>
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