### Integrated BioRefinery: BioMass to BioRenewables

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The Technology/The Market: 1,3-Propanediol

- Anaerobic fermentation of 1,3-Propanediol from waste glycerol using bacteria found in local soil.
- Previous research done to help optimize production:
  - Different feed mediums for bacteria growth.
  - Effect of pH control or not on production.
  - Design of Experiment of batch fermentations to determine optimal feed composition and conditions.
- Current annual production of Waste Glycerol in the USA consists of over 2.5 billion pounds.
- 1,3-Propanediol is a specialty chemical used in the making of polyesters, adhesives, laminates, medicines, solvents, lubricants, and antifreezes.

**Objective:** To understand the effect that acetate and butyrate have on the anaerobic fermentation of waste glycerol to 1,3-Propanediol and its yield.

### Methods:

#### Culture

- Using incubator shaker, grow bacteria from local soil in distinct medium until post exponential phase.
- Inoculate bacteria into the same medium to remove soil particulate and continue growth.

#### Ferment

- Re-inoculate bacteria into fermentation feed once post-exponential growth is achieved.
- Store in incubator shaker.
- Take periodic samples for analysis.

#### Analyze

- Measure biomass using optical density readings from UV-Vis Spectrophotometer.
- Observe composition of fermentation products through Gas Chromatography.

### Results:

#### Biomass Growth

<table>
<thead>
<tr>
<th>Acetate Initial (g/L)</th>
<th>Biomass (g/L)</th>
<th>Lag Time (h)</th>
<th>Butyrate (g/L)</th>
<th>Acetate (g/L)</th>
<th>1,3-PD Yield (mol/mol glycerol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5.0</td>
<td>0.811</td>
<td>2</td>
<td>2.01</td>
<td>3.72</td>
<td>0.113</td>
</tr>
<tr>
<td>7.5</td>
<td>0.647</td>
<td>4</td>
<td>2.01</td>
<td>3.87</td>
<td>0.096</td>
</tr>
<tr>
<td>10.0</td>
<td>0.426</td>
<td>5</td>
<td>3.71</td>
<td>4.99</td>
<td>0.120</td>
</tr>
<tr>
<td>12.5</td>
<td>0.411</td>
<td>6</td>
<td>2.73</td>
<td>8.00</td>
<td>0.114</td>
</tr>
</tbody>
</table>

#### Waste Glycerol

No increase in biomass from butyrate to acetate was found. Could be due to experimental error or the fact that the absence of pH control during fermentation killed the bacteria.

### Conclusion:

- Acetate feeds show reduced lag time over butyrate feeds.
- Butyrate feeds show higher yield of 1,3-Propanediol.
- Optimal concentration of acetate and butyrate feeds for 1,3-Propanediol production is 5 g/L, found by taking into account the lag time, biomass, and 1,3-Propanediol yield.
- No increase in biomass from butyrate to acetate was found. Could be due to experimental error or the fact that the absence of pH control during fermentation killed the bacteria.

### Ongoing Research:

- CSTR construction using optimal conditions found in previous research.
- Membrane development for 1,3-Propanediol separation.
- Generative growth of bacteria and its effect on 1,3-Propanediol production.
- CSTR cycling of normal, acetate, and butyrate laced feeds.