Judging Adaptability of Anaerobic Biomass to Changed Feed Substrates through Methanogenic Activity Tests and its Verification

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Introduction

Anaerobic Processes investigated for

- Sudden/gradual change in operational/environmental conditions – short duration impacts (without any change in feed type)
  - Hydraulic shock
  - Organic shock
  - Other shock types

✓ Focused on temporal performance of processes
Feeds causing secondary operational problems

- Sludge bulking
- Foaming
- Difficulty in biomass settling

Identified sludge settleability as a serious problem
Reactor maintained on a typical substrate suddenly fed with altogether different substrate type?

Relevant for developing countries?
Objective

Assessing anaerobic biomass adaptability to sudden change in feed substrate through methanogenic activity tests
Experimental

Suspended growth batch reactors: R1, R2 and R3

- Aspirator Bottle
- Anaerobic Biomass
- Gas Outlet
- 11.2% (w/v) KOH Trap
- Gas Flow Meter
- Thermometer
- Rubber Tubes/Connectors

**Reactor Charging:** 1.5 L of anaerobic seed + 3.5 L of tap water

**Feeding:** Once in a day (250 mL of supernatant replaced with feed)
## Selected feed substrate types:

<table>
<thead>
<tr>
<th>Substrate</th>
<th>Characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Jaggery (Soluble)</td>
<td>per 100 g: Sucrose sugar=65-85 g, Reducing sugar=5-15 g, Protein=0.4 g, Fat=0.1 g, Calcium=8 mg, Phosphorous=3-4 mg, Total minerals=0.6-1 g, Moisture content=3-8 g, Carotene (Vitamin A)= 280 μg, Nicotinic acid=1 μg, Thiamine (Vitamin B)=20 μg, Colour=Golden yellow to brown, Energy=383 kcal. COD=957±22 mg/g Jaggery</td>
</tr>
<tr>
<td>Cerelac (Suspended)</td>
<td>per 100 g: Carbohydrate=67.9 g, Protein=15 g, Fat=9 g, Calcium=480 mg, Potassium=500 mg, Sodium=150 mg, Phosphorous=370 mg, Moisture content=2.5 g, Folic acid=25 μg, Thiamine (Vitamin B)=0.8 mg, Ash=3.2 g, Colour=White, Energy=413 kcal. COD=1055±49 mg/g Cerelac</td>
</tr>
<tr>
<td>Neutralized acetic acid (Acetate)</td>
<td>100 mL AR grade acetic acid + 50 mL distilled water + AR grade NaOH pellets to obtain a pH of 7 and final volume made up to 500 mL with distilled water to give a stock acetate solution. COD=161±2 mg/mL of stock solution</td>
</tr>
</tbody>
</table>
Methanogenic activity test set-up

- Serum Bottle
- Anaerobic Biomass
- Reaction Mixture
- CO₂ Scrubber (11.2% w/v KOH + thymol blue indicator)
- Rubber Tubing
- Pinch-Cock
- Hypodermic Needle
- Conical Funnel
- Displaced Liquid
- Liquid Displacement System Serum
Operating Conditions:

- Three different phases maintained
- OLR = 1.40 kg COD/m³ d
- Room temperature (18-26 °C)

Phase – I

Aim: To obtain similar steady responses
Feed: Model Feed (Jaggery) to R1, R2 & R3
Period: 65 days
Phase – II

Aim: To impose changed feed substrate types
Feed: Model feed (Jaggery) to R1
       Cerelac to R2 &
       Neutralized acetic acid to R3
Period: 64 days

Phase – III

Aim: To discontinue changed feeds & to restore
      Phase - I model feed
Feed: Model feed (Jaggery) to R1, R2 & R3
Period: 54 days
Performance Monitoring:

- COD (feed & effluent) – alternate day
- VFA & BA – every 2-3 days
- Room temp. & biogas or CH₄ production – daily
- Methanogenic activity tests – in Phase II
## Results and Discussion

### Steady Response in Phase – I:

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Reactor</th>
<th>Phase–I 50–65 d</th>
<th>Temp = 25±0 °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biogas Produced (L/d)</td>
<td>R1</td>
<td>2.09±0.07 (13)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R2</td>
<td>1.91±0.11 (14)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R3</td>
<td>2.15±0.14 (13)</td>
<td></td>
</tr>
<tr>
<td>Methane Produced (L/d)</td>
<td>R1</td>
<td>0.59±0.02 (2)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R2</td>
<td>0.73±0.01 (2)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R3</td>
<td>0.67±0.12 (2)</td>
<td></td>
</tr>
</tbody>
</table>

- **Phase – I:** Similar steady response for biogas/methane
Response in Phase – II (under shock conditions):

Variation in biogas and methane production
Steady Response in Phase – II:

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Reactor</th>
<th>Phase–II</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>44–66 d</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Temp = 19±1 °C</td>
</tr>
<tr>
<td>Biogas Produced (L/d)</td>
<td>R1</td>
<td>1.96±0.31 (9)</td>
</tr>
<tr>
<td></td>
<td>R2</td>
<td>1.27±0.30 (8)</td>
</tr>
<tr>
<td></td>
<td>R3</td>
<td>–</td>
</tr>
<tr>
<td>Methane Produced (L/d)</td>
<td>R1</td>
<td>0.60±0.11 (11)</td>
</tr>
<tr>
<td></td>
<td>R2</td>
<td>0.58±0.15 (11)</td>
</tr>
<tr>
<td></td>
<td>R3</td>
<td>–</td>
</tr>
</tbody>
</table>

- Reduced biogas production in reactor R2
- Complete cessation of biogas production in reactor R3
Methanogenic Activity Tests in Phase – II: Reactor R1

- Good adaptability of biomass from model feed to cerelac feed
- Non-adaptability of biomass from model feed to acetate feed

MA_{Jaggery} = 0.126 \text{ g CH}_4\text{-COD/g VSS d}  
MA_{Cerelac} = 0.126 \text{ g CH}_4\text{-COD/g VSS d}  
MA_{Acetate} = 0.033 \text{ g CH}_4\text{-COD/g VSS d}
Biomass Adaptability to Model Feed: Phase – II (Reactor R2)

$MA_{\text{Cerelac}} = 0.088 \text{ g CH}_4\text{-COD/g VSS d}$

$MA_{\text{Jaggery}} = 0.120 \text{ g CH}_4\text{-COD/g VSS d}$

- Indicates good adaptability of biomass from cerelac to model feed
Biomass Adaptability to Model Feed: Phase – II (Reactor R3)

Indicates good adaptability of biomass from acetate to model feed

MA_{Acetate} = 0.027 \text{ g CH}_4\text{-COD/g VSS d} 

MA_{Jaggery} = 0.136 \text{ g CH}_4\text{-COD/g VSS d}
Response in Phase – III (model feed again):

Variation in biogas and methane production
**Steady Response in Phase – III:**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Reactor</th>
<th>Phase–III 30–60 d</th>
<th>Temp = 25±1 °C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Biogas Produced (L/d)</td>
<td>R1</td>
<td>2.16±0.33 (18)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R2</td>
<td>2.11±0.10 (19)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R3</td>
<td>1.92±0.14 (19)</td>
<td></td>
</tr>
<tr>
<td>Methane Produced (L/d)</td>
<td>R1</td>
<td>0.60±0.10 (12)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R2</td>
<td>0.79±0.04 (12)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>R3</td>
<td>0.54±0.03 (12)</td>
<td></td>
</tr>
</tbody>
</table>

- Comparable performances as of Phase – I
- Complete recovery of reactor R3 in Phase – III
Summary

- **Impact of change in feed types (Phase – II)**
  - Steady biogas production for R2 goes down
  - Complete cessation in biogas production for R3
  - Biomass adaptability to cerelac but not to acetate
  - MA values for reactor R1 indicated:
    - Adaptability to cerelac feed but not to acetate feed
  - MA values for reactor R2 and R3 indicated:
    - Adaptability to model feed

- **Impact of restoration of model feed (Phase – III)**
  - Reactors recover steady responses of Phase – I
Adaptability of anaerobic biomass to newer feed substrate types may be assessed through methanogenic activity tests.
Thank You