Hydrogen Production From Peach Waste By Thermophilic Bacterium

Thermotoga neapolitana

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Hydrogen As Fuel

• Hydrogen is one of the answers to foreseeable energy crisis and environmental pollution.
• High conversion efficiency and non polluting nature.
• 50 million metric tons of hydrogen being traded worldwide with a growth rate of 10%.
  – Primarily from fossil fuels.
Hydrogen production by Dark Fermentation

• Hydrogen can be produced by anaerobic bacteria through dark fermentation.

Hydrogen production by Dark Fermentation

- Dark fermentation can use various renewable biomass including agriculture waste (Hussy et al., 2005; Logan et al., 2002), municipal waste (Wang et al., 2003), food processing waste (Van Ginkel et al., 2005)

- The amount of hydrogen production from glucose by bacterium is affected by metabolic pathway and end-products.
Stoichiometry

• \( \text{C}_6\text{H}_{12}\text{O}_6 + 6\text{H}_2\text{O} \rightarrow 12\text{H}_2 + 6\text{CO}_2 \)  
  \( \Delta G = -6.18 \text{ kcal/mol} \) (Thauer, 1976)

• \( \text{C}_6\text{H}_{12}\text{O}_6 + 2\text{H}_2\text{O} \rightarrow 2 \text{CH}_3\text{COOH} + 4\text{H}_2 + 2\text{CO}_2 \)  
  \( \Delta G = -51.6 \text{ kcal/mol} \) (Thauer, 1976)

• \( \text{C}_6\text{H}_{12}\text{O}_6 + 2\text{H}_2\text{O} \rightarrow 2 \text{CH}_3\text{CH}_2\text{COOH} + 2\text{H}_2 + 2\text{CO}_2 \)

• \( \text{C}_6\text{H}_{12}\text{O}_6 + 2\text{H}_2\text{O} \rightarrow \text{CH}_3\text{CH}_2\text{OH} + \text{CH}_3\text{COOH} + 2\text{H}_2 + 2\text{CO}_2 \)
Microorganisms used

- Hydrogen-producing microbes have been found in environments with a wide range of temperature, including mesophiles (25-40°C) (Kotay and Das, 2007; Shin et al., 2007), thermophiles (40-65°C), extreme thermophiles (65-80°C), or hyperthermophiles (>80°C) (Jannasch, 1988).
Thermotoga neapolitana

- Originally isolated around the bay of Naples, Italy (Belkin et al., 1986), *Thermotoga neapolitan* is a Gram-negative, rod-shaped, obligate anaerobic, fermentative extreme thermophile surrounded by a bag-shaped sheath-like outer structure called “toga” (Huber et al., 1986).

- *Thermotoga neapolitana* accumulated 25-30% hydrogen during its incubation (Schroder, 1994).
Prior Research at Clemson

- Carbon source affected the incubation time of *Thermotoga neapolitana* to produce hydrogen.
- *Thermotoga neapolitana* can utilize different carbon sources, to produce hydrogen, such as glucose, sucrose, xylan, rice flour, cellobiose, corn starch, starch.
Prior Research at Clemson

• pH and hydrogen partial pressure ($\text{pH}_2$) are two important factors that affect the hydrogen production and bacterial growth.

• When pH decreased to 5.0, it appears to inhibit hydrogen production.
  – (Huber and Hannig, 2006; Ravot et al., 1995; Van Ooteghem et al., 2002).

• Maximum hydrogen pH$_2$ in this study was 45 kPa.
  – Hydrogen production is inhibited at partial pressures greater than 38 kPa.
Effect of pH

- Jannasch et al reported that pH range for growth is between 5.5 and 9 (Jannasch et al., 1988). Van Ooteghem also reported that pH of medium dropped from 7.5 to 4.5 within 20hrs.

- The proton concentration affects the yield and rate of hydrogen production and the range of pH favorable to hydrogen production is narrow (Lay, 2000)
Agricultural Wastes- The Answer

• Fermentation of agricultural waste products provides a much cheaper option for production of hydrogen.

• South Carolina –second largest producer of peaches in US.

• 20 million pounds of rotten or spoiled peach waste dumped annually.
Peach Waste As Substrate

• This peach waste has high organic value with 4.6%-9.6% of sugars (Wills et. al., 1986) on wet weight basis.

• Major sugars include glucose, fructose, xylose.
Objectives of study

• Investigate the hydrogen production by *Thermotoga neapolitana* on peach media.
• Investigate unaucloaved peach media for hydrogen production.
• Investigate the effect of pH on hydrogen production using peach media.
Methods Used

• All experiments were done in triplicates using 500 ml serum bottles as batch reactors with 100 ml working volume.

• The media used consisted of 50gms/liter of meshed peaches (on wet weight basis) as carbon source along with nitrogen sources (ammonium chloride, trypticase and yeast extract) and other nutrients.

• The batch reactors were run at 77°C for 20 hrs.
Methods Used

• Hydrogen concentration in the headspace was analyzed through gas chromatography using TCD detector and Argon as the carrier gas on a silica column.
Results

• Using T-test, at 5% level of significance, there was no difference in the mean hydrogen concentration produced after 20 hrs for batch reactors with 5 g/L glucose and reactors with 5 g/L (dry weight basis) peaches as carbon source respectively.
Results

• For the autoclaved media the pH decreased to 4.88 after 20 hrs of incubation. Hydrogen concentration did not increase beyond 20 hrs.
## Unautoclaved vs. Autoclaved Media

<table>
<thead>
<tr>
<th>Media</th>
<th>[H2] (%) after 20 hrs</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unautoclaved</td>
<td>24.74</td>
<td>2.33</td>
</tr>
<tr>
<td>Autoclaved</td>
<td>24.33</td>
<td>0.70</td>
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</table>
Results

- The initial pH of 8 was found to be the best for hydrogen production using non autoclaved peach media. Initial pH of 8.5 was found to be the next best pH.

<table>
<thead>
<tr>
<th>Initial pH</th>
<th>[H2] (%) after 20 hrs</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>7.5</td>
<td>18.37</td>
<td>3.6</td>
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<tr>
<td>8.0</td>
<td>24.23</td>
<td>0.69</td>
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<tr>
<td>8.5</td>
<td>22.08</td>
<td>0.098</td>
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</tbody>
</table>
[H2] (%) vs. Initial pH
Results

• [H2] when pH was set back to pH of 7.5 and 6.5 respectively after 12hrs of incubation.
  a) There was an approximate increase of 21.2% in hydrogen gas concentration when pH was set back to 7.5 after 12 hrs of incubation.
  b) There was an approximate increase of 20.6% in hydrogen gas concentration when pH was set back to 6.5 after 12 hrs of incubation.
Conclusions

• Peach media using peaches as carbon source was found suitable for hydrogen production with hydrogen production of 24%-25% after 20 hrs of incubation at 77C.

• Unautoclaved peach media can be used for hydrogen production on peach media.

• An initial pH of 8 was found best for hydrogen production on unautoclaved peach media.

• pH maintenance is required for hydrogen production on peach media.
References


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