Improvement of Mesophilic Biohydrogen Production from Palm Oil Mill Effluent Using Ozonation Process

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Abstract

Biological fermentative production of hydrogen from the ozonated palm oil mill effluent (POME) was conducted in batch reactors using an anaerobic sludge as a microbial seed. Fermentation was setup at pH 4.0-6.0, varying POME concentration range of 5,000-37,000 mg L\textsuperscript{-1} under mesophilic condition (37°C). The results showed that pH 6.0 is an optimum pH and the maximum hydrogen yield of 28.3 mL g\textsuperscript{-1}COD was obtained. Comparative results of hydrogen production from the raw POME versus the ozonated POME indicated that the ozone pretreatment of POME (mg COD: mg ozone = 102.8) elevated the biodegradability of the POME constituents and significantly enhanced yield and rate of the hydrogen production. Hydrogen production using ozonated POME concentration of 30,000 mg L\textsuperscript{-1} displayed the maximum yield of 182.3 mL g\textsuperscript{-1} COD, which is 49\% higher than that from raw POME. Meanwhile the maximum production rate of 43.1 mL h\textsuperscript{-1} was observed at COD concentration of 25,000 mg L\textsuperscript{-1} ozonated POME. Maximum COD removal was 44\% at COD concentration of 15,000 mg L\textsuperscript{-1} ozonated POME. This work demonstrated ozonation of POME significant improved performance of hydrogen production.

Keywords: Hydrogen; ozonation; palm oil mill effluent; POME; fermentation; bioenergy

1. Introduction

Palm oil industry in Thailand has been rapidly expanded. Palm oil mill effluent (POME) is an important source of water pollution when released into local rivers or lakes without proper treatment. The untreated, viscous,
brown liquid POME contains high content of chemical oxygen demand (COD), biochemical oxygen demand (BOD), and suspended solids (SS). Since the viscous, brown liquid POME contains high chemical composition and concentrations of simple organic compounds (e.g. carbohydrate, protein, and lipids) and complex organic complex compounds (e.g. tannins and lignins) [1]. As a result high biodegradable organic content, anaerobic process for reduction of the pollution in the POME is advantageous before being released to the environment. Therefore, the current study is intrigue on the anaerobic process capable of reducing the pollution in the POME and generating the hydrogen at the same time. Hydrogen can also be biologically produced through the dark fermentation of renewable feedstocks (organic-rich biomass and wastewater) with no requirement of external energy input [2]. Partial ozone pretreatment is aimed to breaks down complex molecules of organic matters in POME to be more biodegradable. In addition, the impact of partial ozone pretreatment on the rate and yield of hydrogen production of POME was explored.

2. Materials and methods

2.1. Microbial seed

Microbial seed used throughout the study was obtained from a full-scale up-flow anaerobic sludge blanket (UASB) reactor (Malee Sampran Co., Thailand). Granular seeds with diameter >0.5 mm were washed with tap water and pre-treated at 100°C for 30 min [3] to inhibit the methane-producing bacteria prior to use into the hydrogen reactor.

2.2. Palm oil mill effluent (POME)

The raw POME was collected from the stabilization pond (Sooksomboon Palm Oil Co., Ltd. Chonburi, Thailand). Raw POME exhibits dark brown color, a temperature of 70-80°C, and pH 4.4-4.7. Suspended solids in the raw POME were allowed to settle down for one day (24h) before its storage in a cold room at 4°C to use. The ozonated POME was prepared by using the pre-settled POME was ozonated (Ozonator, model OZ-553, Thailand) with ozone loading rate of 300 mg h⁻¹ (mg COD: mg ozone = 102.78 from separated experiment to clarify the impact of ozonation on the improved hydrogen production yield).

2.3 Batch fermentation

The first set of batch experiment was varied pHs 4.0-6.0 to obtain an optimal pH for the hydrogen production and second batch were varied POME concentrations of 5,000-37,000 mg L⁻¹ under the optimal pH at 37°C. After placing POME and microbial seed in the reactor, it was capped tightly with the silicone stopper and flushed with nitrogen gas to create an anaerobic condition. All experiments were performed in duplicate. Gas sample was taken once in every 4 h, the volume of biogas was collected with measured by water displacement method. Mixed liquor samples were analyzed for pH, total chemical oxygen demand (COD₅), dissolved chemical oxygen demand (CODs), total solid (TS) and total volatile solids (TVS) according to standard methods [4].

2.3. Analytical methods

Gas composition in the batch reactor headspace was measured on a gas chromatography (Shimadzu GC 2014, Japan) equipped with thermal conductivity detectors (TCD) fitted with stainless steel column packed with Unibeads C (80/100 mesh). Helium was used as a carrier gas. The temperatures of the injection port, column and detector were 120, 70 and 150°C, respectively. Volatile fatty acids (VFAs) and ethanol were analyzed by a gas chromatography (Shimadzu GC 2010, Japan) equipped with a flame ionization detector (FID) fitted with Stabl wax DA capillary column (Restek, USA). Hydrogen, air, nitrogen and helium were used as a carrier gas. The temperature of the injection port, column and detector were set up at 230, 80 and 250°C, respectively.
2.4. Data analyses of batch experiment

Kinetics of hydrogen production was calculated from the cumulative hydrogen production versus time data of each batch experiment fitted with the modified Gompertz equation [5].

\[
H = H_{\text{max}} \cdot \exp\left\{ -\exp\left[ \frac{R_{\text{max}} \cdot e^{(\lambda - t)}}{H_{\text{max}}} + 1 \right] \right\}
\]

where, \( H \) is the cumulative volume of hydrogen production (mL), \( t \) is time of fermentation (h), \( H_{\text{max}} \) is the hydrogen production potential (mL), \( R_{\text{max}} \) is the maximum hydrogen production rate (mL h\(^{-1}\)), \( \lambda \) is lag phase (h) and \( e \) is a constant (2.71828).

3. Results and discussion

3.1. Optimum pH

The first experiment was performed under varied pHs ranging from 4.0-6.0, using the raw POME and the ozonated POME with the initial concentration of 10,000 mg L\(^{-1}\). The results showed that pH 6.0 was the optimum pH and gave the highest hydrogen production yield for both types of POME (Table 1). Hydrogen production tended to decrease when pH decreased and no hydrogen was produced at pH 4.0. pH is the most factors of hydrogen fermentation processes on the hydrogenase activity in the metabolic pathways [6]. If pH is not maintained, it may inhibit hydrogen production. Kinetics data \( H_{\text{max}} \), \( R_{\text{max}} \), \( \lambda \), and yield (Table 1) were clearly indicated the significant role of pH on the hydrogen fermentation, and could be strongly concluded that pH 6.0 is the optimum pH for the current study.

Table 1. Kinetic parameters of hydrogen production under varied initial pHs

<table>
<thead>
<tr>
<th>pH</th>
<th>( H_{\text{max}} ) (mL)</th>
<th>( R_{\text{max}} ) (mL h(^{-1}))</th>
<th>( \lambda ) (h)</th>
<th>Yield (mL g(^{-1})COD)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Raw POME</td>
<td>Ozonated POME</td>
<td>Raw POME</td>
<td>Ozonated POME</td>
</tr>
<tr>
<td>4.0</td>
<td>0.0±0.0</td>
<td>0.0±0.0</td>
<td>0.0±0.0</td>
<td>0.0±0.0</td>
</tr>
<tr>
<td>4.5</td>
<td>0.4±0.1</td>
<td>0.4±0.1</td>
<td>0.2±0.1</td>
<td>0.2±0.1</td>
</tr>
<tr>
<td>5.0</td>
<td>27.1±4.0</td>
<td>39.7±1.0</td>
<td>4.2±1.2</td>
<td>6.2±1.0</td>
</tr>
<tr>
<td>5.5</td>
<td>37.6±2.5</td>
<td>51.7±4.9</td>
<td>4.1±0.9</td>
<td>5.1±1.3</td>
</tr>
<tr>
<td>6.0</td>
<td>58.0±2.4</td>
<td>100.5±8.7</td>
<td>5.0±0.7</td>
<td>12.9±1.3</td>
</tr>
</tbody>
</table>

3.2. Impact of ozone pretreatment on hydrogen fermentation

The experiment was setup with varied POME concentrations and fixed pH 6.0. pH drop after 72 h incubation was observed in the narrow ranges of 5.38-5.97 for the raw POME and 5.46 to 5.98 for the ozonated POME. The result suggested that organic acid by-products from the hydrogen fermentation were accumulated in the reactor content. The biogas was free of methane at all tested POME concentrations and trend of hydrogen content was increased with increased POME concentration. The cumulative hydrogen production trends to increase with POME concentration from 5,000-30,000 mg L\(^{-1}\), and decreased with high concentration was 37,000 mg L\(^{-1}\) (Fig. 1). All cumulative hydrogen fermentation profile data were fitted to the modified Gompertz equation (\( r^2 > 0.99 \)).

Table 2 summarized the kinetic parameters for the hydrogen production with varied POME concentrations. The regression results showed that \( H_{\text{max}} \) and Yield were all dependent on the POME concentrations. The \( H_{\text{max}} \), \( R_{\text{max}} \), \( \lambda \) and Yield trends to increase as POME concentration. While \( R_{\text{max}} \) increased and then decreased at the POME concentration higher than 25,000 mg L\(^{-1}\). The highest \( R_{\text{max}} \) was 43.1 mL h\(^{-1}\) of 25,000 mg L\(^{-1}\) ozonated POME. At the POME concentration from 25,000-37,000 mg L\(^{-1}\), the \( \lambda \) ranged was increased because POME contained very...
Fig. 1. Cumulative hydrogen production of raw POME (a); ozonated POME (b) with the initial concentrations of 5,000 (○); 10,000 (●); 15,000 (●); 20,000 (○); 25,000 (□); 30,000 (▲); 35,000 (●); 40,000 (●); 45,000 (●); and 50,000 (●) mg L\(^{-1}\). The data represent mean values and I-bars represent standard deviation of duplicate experiments.

High particulates which required long hydrolysis time. The ozonated POME concentration of 30,000 mg L\(^{-1}\) gave the highest yield of 182.3 mL g\(^{-1}\)COD. Yield was directly related to the elevated POME concentrations, except that the yield was dropped at the highest concentration of 37,000 mg L\(^{-1}\) for both POMES. At the corresponding concentration, the yield from the ozonated POME was higher than that from the raw POME in most cases. The results strongly indicated that the partial ozonation of POME enhance the hydrogen fermentation of POME under mesophilic condition.
Table 2. Kinetic parameters of hydrogen production at pH 6.0 and varied POME concentrations

<table>
<thead>
<tr>
<th>[COD] (mg L⁻¹)</th>
<th>Hmax (mL)</th>
<th>Rmax (mL h⁻¹)</th>
<th>λ (h)</th>
<th>Yield (mL g⁻¹ COD)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Raw POME</td>
<td>Ozonated POME</td>
<td>Raw POME</td>
<td>Ozonated POME</td>
</tr>
<tr>
<td>5,000</td>
<td>9.2±7.3</td>
<td>22.3±22.5</td>
<td>2.0±1.6</td>
<td>3.3±3.7</td>
</tr>
<tr>
<td>10,000</td>
<td>65.5±5.8</td>
<td>102.0±7.9</td>
<td>14.7±0.3</td>
<td>13.7±1.1</td>
</tr>
<tr>
<td>15,000</td>
<td>121.7±4.3</td>
<td>207.4±0.4</td>
<td>17.1±3.9</td>
<td>19.6±8.8</td>
</tr>
<tr>
<td>20,000</td>
<td>188.5±29.6</td>
<td>291.6±0.8</td>
<td>24.5±5.3</td>
<td>33.3±7.2</td>
</tr>
<tr>
<td>25,000</td>
<td>228.6±22.5</td>
<td>317.2±45.9</td>
<td>30.1±4.9</td>
<td>43.1±2.5</td>
</tr>
<tr>
<td>30,000</td>
<td>391.3±16.1</td>
<td>349.7±13.9</td>
<td>20.0±1.0</td>
<td>23.4±1.2</td>
</tr>
<tr>
<td>37,000</td>
<td>346.5±9.2</td>
<td>243.9±7.0</td>
<td>16.1±1.1</td>
<td>15.1±3.0</td>
</tr>
</tbody>
</table>

3.3 COD reduction

The COD removal efficiencies trends to increase with the increased of POME concentration from 5,000-15,000 mg L⁻¹, and then was decreased at concentrations increased from 15,000 - 30,000 mg L⁻¹. The POME concentration of 30,000 mg L⁻¹ gave the lowest COD removal efficiencies in both studied and the raw POME showed higher COD reduction than the ozonated POME. The COD and COD₅ removal efficiencies for both POME exhibited similar trends but the COD₅ removal efficiencies were dropped at the concentration of 37,000 mg L⁻¹ for both types of POME. Higher COD₅ removal in ozonated POME in the corresponding concentrations suggested that partial ozonation of POME improved the biodegradability of soluble substrate in the POME.

3.4 Volatile fatty acids

Acetic and butyric acids were dominant soluble by-products of both types POME in all batches. The concentrations of acetic and butyric acids were increased as the increased of the POME concentration. Little amount of valeric and propionic acids were constant although the concentration of POME was increased. Ethanol accumulation was detected when the POME concentration of 25,000 mg L⁻¹ and then increased at the POME concentration higher than 25,000 mg L⁻¹ for both POME. The ozonated POME concentration of 30,000 mg L⁻¹ showed the highest hydrogen production yield, in which acetate (56.4 mM) and butyrate (37.6 mM) were dominant end products. Propionate and ethanol were negligible.

4. Conclusion

This study successfully demonstrated the optimum pH for the hydrogen production using the UASB granular seed reactor at mesophilic conditions was 6.0. The comparative kinetic parameters of hydrogen production from two types of POME, raw POME and ozonated POME (102.78 mg COD mg⁻¹ ozone) in the concentration ranging from 5,000-37,000 mg L⁻¹, the ozonated POME concentrations of 30,000 mg L⁻¹ gave the maximal hydrogen production potential of 349.8 mL and the maximal hydrogen production yield of 182.3 mL g⁻¹ COD. The maximal hydrogen production rate (Rmax) was 43.1 mL h⁻¹ was observed of 25,000 mg L⁻¹. The maximum COD₅ removal was 44% at POME concentration of 15,000 mg L⁻¹. Ozonation pretreatment of POME was significantly enhanced the biodegradability of POME constituents, hydrogen production yield, and rate.

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References


