Biohythane from organic waste: An overview

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Abstract. Organic wastes considered as a common problem in Indonesia, especially from agricultural waste, such as fruits and vegetables waste. However, these organic wastes can also converted into biogas. Biohydrogen and biomethane play important roles for future economical energy sources due to clean, CO₂ neutral and environmentally friendly. Biohythane is a mixture of methane and hydrogen that produced in two separated bioreactor. The total energy recovered from two stages fermentation considered higher than single stage bioreactor. The short hydraulic retention time applied in the first stage is enough to separate acidogenesis from methanogenesis; however the pretreatment is also needed for lignocellulosic waste. Temperature application in the first or both reactors are effective to improve the yield of biohythane from the organic waste; meanwhile it needs more energy rather than mesophilic condition. Bioreactor utilization by considering types of fermentation should become attention in the case we have to choose between solid or liquid organic waste. The range results from two stages are methane 45-55 %, hydrogen 11-15 %, and carbon dioxide 30-35 %, respectively.

Keywords: Biohythane, dark fermentation, hidrolysis, acidogenesis, methanogenesis

1. Introduction

Bioenergy is a renewable energy from biomass, to use organic waste as energy resources. While several attempt to promote biogas as renewable resources, the critical analysis and comprehensive is very limited [1]. Biohythane is mixture of gas which consisted of approximately 10-30 % hydrogen and methane [2]. Biohythane can be produced from many different organic substrates using two different types of reactor for fermentative and anaerobic digestions which produce hydrogen and methane. A Two-phase bioreactor combined biohydrogen and biomethane which can be one of solution to improve the efficiency of anaerobic digestion [3]. During hydrogen fermentation, the loss of hydrogen by specific methane-producing microorganisms needs to be prevented [4,5], pH, and Hydraulic Retention Time (HRT) adjustments are most applicable methods to prevent methanogenesis in industrial scale [6,7,8]. However, short HRT cannot be applied for complex material such as lignocellulose and manure due to the poor hydrolysis of these materials. To solve the problem, pretreatment is needed for lignocellulosic materials prior to biohydrogen production at the first reactor [9]. The aims of this research are to reduce organic waste by converting into biogas as energy source. This review is also collecting the information and resume about biohythane production from previous studies.
2. Two Phase Anaerobic Digestion

The definition of biomass is the part of plant material that forms the process of contemporary photosynthesis and is stored as chemical energy [10]. Anaerobic digestion technology is used organic waste or biomass to produce biogas, in addition to the benefits of anaerobic digestion it can also be used for waste treatment, reduction of pollutants, odours and diseases [11]. In two stage anaerobic digestion, residual substrates from the first stage could be reduced continuously in the second stage with similar mechanism as the first reactor. Unlike the first terminology, two-phases of anaerobic digestion mainly to separate two different processes in anaerobic digestion: acidogenesis and methanogenesis. The separation can be done with different HRT. Liquefaction and acidification of the substrate is accomplished in the first reactor, while only methanogenesis takes place in the second reactor [12].

Two-phase anaerobic digestion has been reported by many researchers. Many of them used modified bioreactor to improve biohydrogen production. Chun-Feng chu et.al [13] used thermophilic acidogenesis reactor which followed by mesophilic reactor for methanogenesis. The focus of most researchers is on the first acidogenic reactor, because the first reactor accelerates the melting of organic material. This can happen in the first reactor, where the hydrolysis process converts large molecules into monomers, and the pH decreases to become acidic. The bacteria are difficult to survive under low pH conditions, methanogens can be automatically removed in the first phase, and biohydrogen produces more intensity in this phase [14]. To optimize biohydrogen production, some researchers use heat treatment to remove methanogens and induce endospore of biohydrogen-production bacteria. The process of maturity seems to determine the whole process of succession. Stable conditions have been obtained after three months of operation.

In the Oil Palm Plantation, there is a huge amount of organic agricultural waste such as empty fruit bunches (EFB), oil palm fibre (PPF) and decanter cake (DC). Solid State Anaerobic Digestion (SS-AD) investigations using Palm Oil Mill Effluent (POME) as co-digestion have been reported [15,16,17]. Meanwhile Pamungkas et.al., [18] was investigated the effect of temperature and pH on sweet potato starch residue fermentation to produce biohydrogen through dark fermentation using yeast. The optimum temperature to produce biohydrogen was 27 °C and optimum pH was 5.00. Other researcher evaluate the use of thermophilic reactor 55 °C [19,20] and 60 °C [21]. Cavinato et.al [22], has investigated the optimization of a two-phase thermophilic in two stirred reactors (CSTRs), without any physical/chemical pretreatment of inoculum. Sung et.al [23] used heat repetition in acidogenic reactor. On the other hand, an operational due to the loading rate effect  of pH 5.5 was shown to be optimal for hydrogen production.

Wang et.al [24] has reported the effects of the stepwise increased organic loading rate (OLR) and solid retention time (SRT) on integrated two-stage process. Van Ginkel et.al [25] investigated biohydrogen production from food wastewater processing that considered potential and has the low range of COD reduction which only 5 – 11 %. Furthermore, C. Mamimin et.al [26] has investigated the effect of organic loading rate (OLR), hydraulic retention time (HRT) and temperature variation on process stability of hydrogen production from palm oil mill effluent (POME) under thermophilic condition. Similar research has been reported by Hendroko et.al [27] that used Jatropha curcas Linn capsule husk at two stages digestion. There are also interesting investigations in the thermophilic condition related to volatile fatty acid (VFA) pattern. C.Mamimin et.al [28] investigated the effects of volatile fatty acids in effluent of biohydrogen reactors biohythane production from palm oil mill effluent under thermophilic. It was found that in the biodeterioriation effluent contains a high concentration of volatile fatty acid.
Table 1. The Summaries of anaerobic digestion

| No. | Types of digestion and substrate utilization | Temperature | Results | Ref. |
|-----|---------------------------------------------|-------------|---------|------|
| 1.  | Two-Phase CSTR reactor with Municipal Solid Waste (MSW) | Thermophilic-Mesophilic | 205 ml H₂/g VS<sub>added</sub> and 464 ml CH₄/g VS<sub>added</sub>. | [13] |
| 2.  | Acidogenic CSTR reactor of POME | Mesophilic | HRT 4 day and pH 6 was optimum condition | [14] |
| 3.  | Solid State-AD of EFB, PPF and DC | Mesophilic | F/1 2:1 is the highest methane production. | [15] |
| 4.  | Two State SS-AD EFB and DC co-digestion with POME | Thermophilic-mesophilic | 10 % co-digestion of POME is optimum condition | [16] |
| 5.  | SS-AD of EFB and LS-AD of POME co-digestion with sludge waste. | Mesophilic | LS-AD 2-3 higher volumetric biogas productivity than SS-AD | [17] |
| 6.  | Co-digestion EFB with POME | Mesophilic | POME:EFB ratio of 4.5-7.5 and size EFB of 3.3-6 cm was more practical for biogas. | [18] |
| 7.  | Acidogenic reactor of sweet potatoes residues | Variation temperature and pH | 27 °C and pH 5.0 is optimum condition. Others at 55 °C, 55 °C, and 60 °C. | [19,20,21,22] |
| 8.  | Two-phase CSTR reactor | Thermophilic | The composition of mixture gas of 6.7 % H₂, 40.1 % CO₂ and 52.3 % CH₄, and with an overall biogas of 0.78 m<sup>3</sup>/kgVS<sub>fed</sub>. | [23] |
| 9.  | Acidogenic reactor with sucrose as substrate | Heat repetition | Conversion efficiency and hydrogen yield were 0.0892 L-H₂/g-COD and 1.5291 mole of H₂/mole of sucrose | [24] |
| 10  | Two-phase reactor | Mesophilic | Optimal OLR and SRT were found to be 22.65 kg VS/m<sup>3</sup> d (160 h) for hydrogen fermentation reactor and 4.61 kg VS/m<sup>3</sup> d (26.67 d) for methane | [25] |
| 11  | Acidogenic reactor | Thermophilic | High OLR (70 g COD/l/d) and low HRT (4 d) has significant affected on hydrogen productivity but not affected by variation of temperature. | [27] |
| 12  | Two-phase reactor | Thermophilic-Mesophilic | Preventing the high concentration of butyric acid, and propionic acid in the hydrogenic effluent could enhance methane production in two-stage anaerobic digestion for biohythane production | [28] |
| 13  | Two phase reactor | Mesophilic-mesophilic | The reduction of COD from organic waste was more than 80 % for all processes | [29] |
3. Organic Waste as Biohythane Substrate

Almost agricultural organic waste have high contents of lignocellulose substrate that naturally recalcitrant. Biogas produced from anaerobic digestion cannot run effectively if the high lignocellulose content in agricultural biomass does not offer prior pretreatment. Biohythane by two phases of anaerobic digestion seems a promising initiative for the treatment of lignocellulosic waste [30]. Hydrothermal liquefaction can be applied as a pretreatment for lignocellulusic biomass waste, if integrated with two-phase anaerobic digestion for biohythane production can increase overall system effectiveness. Conversion of solid and liquid biomass by hydrothermal liquefaction can be a substrate for two-phase anaerobic digestion [31].

The diversity of microorganisms in biohydrogen production at the first phase was not only for biohydrogen production but also for degradation of inhibitor that might happen in the sequential phase, methanogenesis. The higher distribution of the detoxification family Clostridiaceae, Bacillaceae, and Pseudomonadaceae was found in the biohydrogen process. In addition, a higher distribution of acetate-oxidizing bacteria (Spirochaetaceae) was observed in the two-stage systems biomethane production, revealing improved acetogenesis accompanied with an efficient conversion of acetate. Biohythane production could be a promising process for the recovery of energy and degradation of organic compounds from hydrothermal liquefied biomass. The two-phase processes not only contributes to improve the quality of gas fuels but also strengthen the biotransformation process, which results from the detoxification function during biohydrogen production and increased acetogenesis during biomethane production [32].

POME was used for Biohydrogen production in CSTR reactor [33,34,35]. It was also investigated the anaerobic degradation process for POME in anaerobic bench scale reactor (ASBR) [36,37]. Z.Rasdi et.al [38] has investigated the kinetic models of biohydrogen production from anaerobically treated POME. Meanwhile Cheng Jun et.al [39] has investigated bio-hydrogen production from hyacinth by anaerobic fermentation of digested sludge. Maojin Cui et.al [40] was also investigated in batch experiments that carried out to convert pre-treated poplar leaves by different methods into hydrogen using anaerobic mixed bacteria at 35 °C. The effects of acid (HCl), alkaline (NaOH) and
enzymatic (Viscozyme L, a mixture of arabanase, cellulase, β-glucanase, hemicellulase and xylanase) pretreatments on the saccharification of poplar leaves were studied. The results show that enzymatic pretreatment is an effective method for enhancing the hydrogen yield from poplar leaves.

H. Jiang et al. [41] reported the two-stage hythane fermentation of cassava residue. Cassava residue is low in protein, rich in iron, and deficient in nickel and cobalt, resulted in failure after long-term operation, showing a radical decrease in methane production along with an increase of volatile fatty acids (VFAs) accumulation in the second stage. The proliferation of hydrolysis bacteria, acidogens, and hydrogen producing bacteria and methanogens were guaranteed by sufficient concentration of N (0.7 g/L), S (30 mg/L), Ni (1.0 mg/L), and Co (1.0 mg/L), and the metabolism of sustainable hythane fermentation was recovered. Otherwise, several researchers used other organic waste such as cornstalks [42], Potatoes waste [43], and with head shocked treatment [44] and banana peel [45]. Comparative performance and total energy recovery between two-phases processes (sequential hydrogen and methane fermentation) and one-phase process (methane fermentation) were evaluated in batch reactor under mesophilic incubation at various ratios of feedstock to microbial inoculum (F/M) ranging from 2.5 – 10. Yao-Ting Fan et al. [46] was investigated the conversion efficiency of wheat straw wastes into biohydrogen gas by cow manure compost, where the result is acceptable and recommended.

### Table 2. Summaries of Substrate Utilization

| No | Substrate Utilization | Parameter | Results | Ref. |
|----|-----------------------|-----------|---------|------|
| 1  | Palm Oil Mill Efluent (POME) | Gas production in mesophilic temperature | The results showed that pH 6.0 is an optimum pH and the maximum H₂ yield of 28.3 mL g⁻¹COD was obtained. H₂ production using ozonised POME concentration of 30,000 mg L⁻¹ displayed the maximum yield of 182.3 mL g⁻¹ COD, which is 49 % higher than that from raw POME. | [33,34,35] |
| 2  | Palm Oil Mill Efluent (POME) | Gas production in thermophilic temperature | Hydrogen production using the ozonised POME achieved the maximum yield of 77.1 mL g⁻¹ COD at 35,000 mg COD L⁻¹, which higher than that from the raw POME by 20 %. | [36,37,38] |
| 3  | Hyacinth | Gas production | The highest hydrogen production of 122.3 mL/g is obtained when the initial pH value of fermentation solution is 5.5, the fermentation temperature is 55 ℃ and the weight ratio of hyacinth to microorganism is 1:1. | [39] |
| 4  | Poplar leaves | Gas production and pretreatment of substrate | A maximum cumulative hydrogen yield of 44.92 mL/g-dry poplar leaves was achieved from substrate pretreated with 2 % Viscozyme L | [40] |
| 5  | Cassava residue | Gas production in two-phase reactor | Trace element needs to sufficient growth, N (0.7 g/L), S (30 mg/L), Ni (1.0 mg/L), and Co (1.0 mg/L). | [41] |
| 6  | Cornstalks | Gas production in two or three phases | Hydrolysates from 1 kg of cornstalks could produce 2.61 mol (63.7 L) hydrogen by augmentation with C. paraputrificum and 4.69 mol (114.6 L) methane by anaerobic granular sludge, corresponding to 54.1 % energy recovery | [42] |
| No | Substrate Utilization | Parameter | Results | Ref. |
|----|----------------------|-----------|---------|------|
| 7  | Potatoes waste       | Gas production in two stage reactor | The hydrogen and methane yields were 30 L/kg TS (with a max of 68 L/kg) and 183 L/kg TS (with a max of 225 L/kg), respectively. | [43] |
| 8  | Potatoes waste       | Gas production in two phase reactor | The results showed that the energy efficiency increased from about 20 % (hydrogen production process) to about 60 % with enzyme pretreatment, which indicated the energy efficiency can be improved by combined hydrogen and methane production process. | [44] |
| 9  | Banana peel          | Gas production in two phase reactor | Hydrogen and methane yields of 209.9 and 284.1 mL g⁻¹ VS mL g⁻¹ VS were achieved at F/M of 5.0 in two stage process. | [45] |
| 11 | Wheat straw wastes   | Gas production in acidogenic stage | The maximum cumulative hydrogen yield of 68.1 mL H₂/g TVS was observed at 126.5 h, and the maximum hydrogen production rate of 10.14 mL H₂/g TVS/h with hydrogen content in the biogas was 52.0 % and there was no significant methane observed | [46] |

4. Conclusion
Anaerobic digestion in development is mature technology, therefore many journals or experimental studies have been reported for years. Bioenergy from biomass has provided a lot of energy for human activities, such as electricity, transportation, cooking. Though, the progress of bioenergy from organic matter is not as fast as fossil fuels. At this time energy consumption preferences are increasing rapidly out of balance with the availability of energy from fossil fuels. Hydrogen is a promising energy when used by methane because biogas has added value for combustion. The research emphasizes more on the laboratory scale. It is interesting to investigate the process and design in the pilot plant before implementation on a large scale.

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