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Sustainable Biohydrogen Production from Palm Oil Mill Effluent: Effect of Hydraulic Retention Time in a Hybrid Anaerobic System

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Abstract

Currently, the world is facing two crises: a shortage of fossil fuels and global climate change. Climate change is linked to increased environmental damage from fossil fuel use and the effects of greenhouse gases. Therefore, it is important to achieve breakthroughs to develop alternative energy sources that can replace fossil fuels. One of these is biohydrogen, which plays an important role in future energy because it is environmentally friendly, renewable, and sustainable. In addition, Indonesia is the world's largest producer of palm oil, which naturally generates liquid waste. Using palm oil mill liquid waste to produce biohydrogen via an anaerobic hybrid bioreactor during the acidogenesis phase is the best solution to address environmental impacts while simultaneously providing a clean energy source. This research aims to produce biohydrogen from palm oil mill liquid waste. This was done using an anaerobic hybrid bioreactor during the acidogenesis phase, with hydraulic retention times of 6, 12, and 18 hours. The research results show that the best hydraulic retention time is 18 hours, with a VSS removal efficiency of 98% and biogas production of 5.0 L/day, yielding 64% biohydrogen.



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1. Introduction

The global energy crisis has become a major challenge to sustainable development, driven by the rapid depletion of fossil fuel reserves and growing concerns over energy security [1, 2]. Consequently, efforts have increased to reduce dependence on fossil fuels and promote renewable energy sources such as solar, wind, geothermal, biogas [3], ethanol, and biodiesel [4]. Among these alternatives, hydrogen has attracted significant attention as a clean energy carrier due to its high energy density and environmental friendliness, with an annual production of approximately 55 million tons [5–7]. The hydrogen market is expanding rapidly, with an estimated annual growth rate of up to 10% and a projected value

exceeding \$191.80 billion in 2024 [6, 8]. Despite this potential, the widespread adoption of hydrogen as a fuel remains constrained by techno-economic challenges, reliance on fossil-based production pathways, and issues related to storage and transportation [9–12].

Palm oil mill effluent (POME) is one of the most environmentally problematic agro-industrial wastewaters due to its extremely high organic load, acidity, and large discharge volumes. With chemical oxygen demand (COD) values ranging from 30,000 to 60,000 mg/L, untreated POME can cause severe water pollution, oxygen depletion in aquatic ecosystems, and substantial greenhouse gas emissions. Consequently, there is a critical need for sustainable treatment

technologies that not only mitigate environmental impacts but also enable resource recovery.

In Indonesia, efforts to enhance energy security and environmental sustainability have encouraged the development of renewable energy sources; however, their utilization remains suboptimal. Among emerging alternatives, biohydrogen has gained attention as a promising non-carbon energy carrier with the potential to replace conventional fossil fuels [13]. It can be produced through anaerobic processes, particularly during the acidogenesis phase, using substrates rich in carbohydrates, proteins, and lipids. In this context, palm oil mill effluent (POME) is an abundant, readily available feedstock in Indonesia, offering significant potential for sustainable biohydrogen production [14].

Although anaerobic digestion of POME has been widely studied, most existing research focuses on biomethane production, while biohydrogen generation remains relatively underexplored. In particular, few studies have investigated the performance of hybrid anaerobic systems under varying hydraulic retention times (HRT) to enhance hydrogen production. The optimization of HRT is especially important in continuous systems, where shorter retention times may increase productivity but increase the risk of biomass washout. In comparison, longer retention times improve stability at the expense of efficiency.

Currently, Indonesia is the world's largest producer of crude palm oil, accounting for 40% of global production, followed by Malaysia at 37.3%. In 2024, crude palm oil production reached 48.16 million tons. For every ton of crude palm oil produced, 2.5 m³ of liquid waste is generated. Meanwhile, Riau Province is the largest exporter of crude palm oil (CPO) in Indonesia, accounting for 38% of the country's total export volume, with 225 palm oil mills (POMEs) producing 6.3 million tons of CPO. This results in 15.75 million m³ of liquid waste with a COD (Chemical Oxygen Demand) content ranging from 30,000 to 60,000 mg COD/L. One method of utilizing this liquid waste is through the anaerobic process [14]. The anaerobic process has dual advantages: it reduces COD concentration, allows the liquid to be used as liquid organic fertilizer, and produces fuel and electricity.

Biohydrogen from palm biomass offers various advantages, including zero carbon emissions, and can be used as a fuel to generate heat, with pure water as a byproduct [15]. In addition, biohydrogen is considered a future energy carrier that can be used for various purposes as a fuel. The development of biohydrogen production through anaerobic fermentation using

bacterial communities derived from animal manure to generate new, renewable energy that is sustainable in the future by reviewing various aspects to improve the process of biohydrogen production by anaerobic bacteria in the acidogenesis phase [16]. Biohydrogen plays an important role in future energy because it is environmentally friendly, renewable, sustainable, and uses low energy [17]. In addition, fossil fuel energy sources are a significant contributor to greenhouse gases.

Biohydrogen production involves several process stages, namely hydrolysis and acidogenesis. The performance of the acidogenesis phase depends on the hydrolysis process, which breaks down complex organic compounds into simpler ones, which are then converted into acetic acid, H₂, and CO₂ [18]. One of the raw materials for biohydrogen production is POME, which is a suitable substrate in Indonesia because the country is a palm oil producer and the world's largest contributor of POME.

A hybrid anaerobic bioreactor with an acidogenesis phase was developed as a biohydrogen generator by optimizing the growth of anaerobic bacteria during acidogenesis, thereby maximizing biohydrogen production from palm oil mill effluent (POME). In addition to reducing COD values, it can produce biohydrogen that can be used as a new, renewable fuel and as a substitute for fossil fuels, and generate combustion energy. Previous research by Ahmad has used a hybrid anaerobic bioreactor for biomethane production, combining the acidogenesis and methanogenic phases [19].

The novelty of this biohydrogen production approach lies in the use of an acidogenesis-phase hybrid anaerobic bioreactor that integrates suspended- and attached-growth systems, in which solid media are employed as a cell-immobilization support to enhance biomass retention and minimize washout during the acidogenesis phase. Furthermore, selective inhibition of methanogenic activity enables the enrichment of acidogenic bacteria, thereby promoting efficient hydrogen production under reduced hydraulic retention time (HRT) conditions [18]. In addition to improving process performance, this system offers environmental benefits by facilitating the treatment of high-strength POME and recovering clean energy, thereby reducing organic pollution and greenhouse gas emissions. Accordingly, this study investigates the influence of hydraulic retention time on biohydrogen production from POME in a hybrid anaerobic bioreactor, with particular emphasis on both energy recovery and wastewater treatment performance.

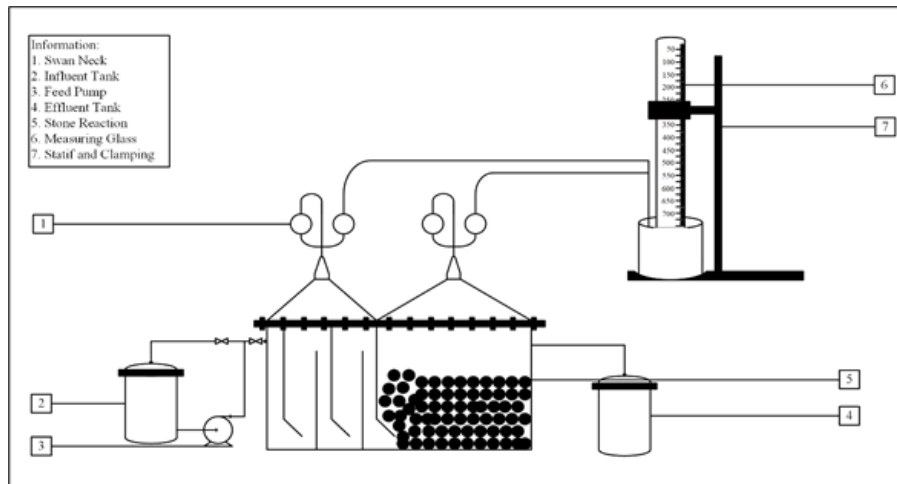


Figure 1. Continuous anaerobic hybrid bioreactor acidogenesis setup.

Table 1. Characteristics of palm oil mill liquid waste.

Parameter	Unit	Value
pH	-	6
Acetic Acid	mg/L	48,250
VSS	mg/L	7600
COD	mg/L	52,500
Environmental Quality Standards		
pH	-	6.0 – 9.0
TSS	mg/L	300
COD	mg/L	500
BOD	mg/L	250

2. Materials and Methods

2.1. Liquid Waste Sources

Palm oil mill effluent (POME) was collected from a palm oil mill in Riau Province, Indonesia, and its COD, pH, volatile fatty acids (VFAs), alkalinity, and volatile suspended solids (VSS) levels were measured.

2.2. Source of Anaerobic Bacteria

The anaerobic bacterial community was developed from cow dung acclimated to POME [19]. Meanwhile, the start-up bioreactor process lasted 14 days at pH 5,6–6,8 under mesophilic conditions until it reached steady-state conditions.

2.3. Anaerobic Hybrid Bioreactor

Design of an Anaerobic Hybrid Bioreactor. The anaerobic hybrid bioreactor used has a volume of 12.5 L and is equipped with stone media as a cell immobilization medium. The continuous acidogenesis phase setup of the anaerobic hybrid bioreactor is shown in Figure 1.

2.4. Research Variables

The variation was achieved by changing the hydraulic retention time to 6, 12, and 18 hours. Samples were taken

daily for analysis of pH, COD, VFA (acetic acid), alkalinity, VSS concentration, and biohydrogen volume and concentration using Biogas 5000 Geotech. The analysis was conducted in accordance with the Standard Methods for the Examination of Water and Wastewater [20].

3. Results and Discussion

3.1. POME Characteristics

Table 1 shows that the liquid waste content of the palm oil mill has characteristic values for several parameters that are relatively higher compared to the environmental quality standards set by the Ministry of Environment of the Republic of Indonesia No. KEP-51/MENLH/10/95. This data indicates that it is important to treat palm oil mill liquid waste to reduce parameters considered to negatively impact the surrounding environment by converting it into biohydrogen using an anaerobic hybrid bioreactor during the acidogenesis phase.

3.2. Effect of Hydraulic Retention Time on Solids Concentration

The anaerobic hybrid bioreactor was operated with variations in hydraulic retention time (6 hours, 12 hours, and 18 hours). Figure 2 shows that hydraulic retention time influences the removal of volatile suspended solids (VSS) in palm oil mill wastewater. The VSS value at a hydraulic retention time of 6 hours was obtained as 0.16 g/L. At 12 hours, the VSS was 0.12 g/L, and at a hydraulic retention time of 18 hours, the VSS was 0.10 g/L. Overall, the concentration of VSS carried out in the effluent increased as the hydraulic retention time was shortened, indicating that VSS loss increased because when the hydraulic retention time was shortened, the feed flow rate increased, causing the flow pattern within the system to become turbulent, thus flushing biomass solids

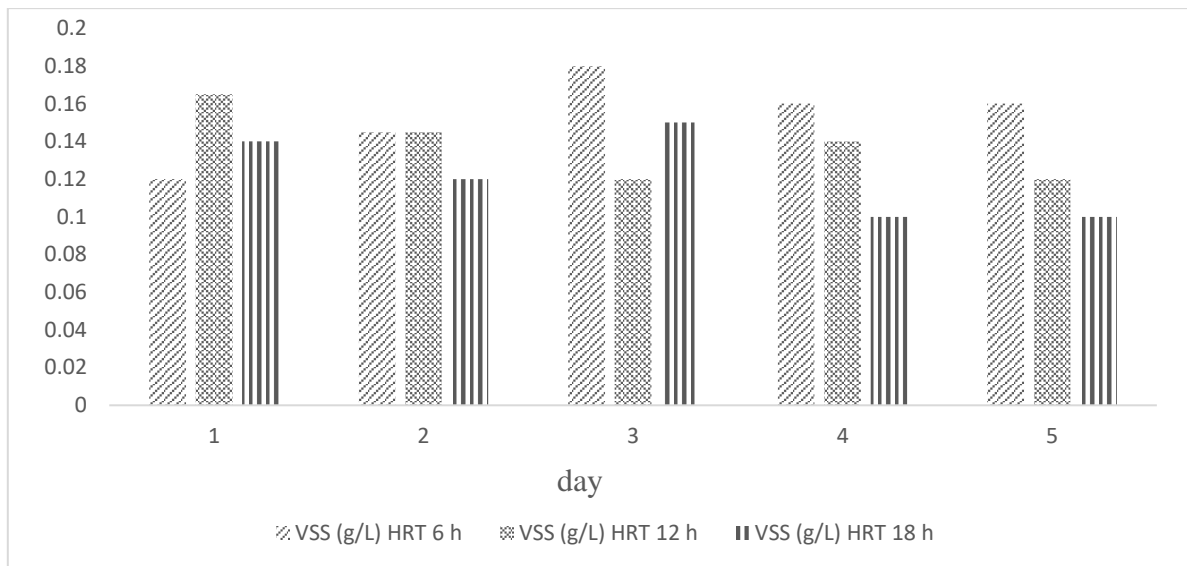


Figure 2. VSS profile at different hydraulic retention times.

out of the system [21]. The VSS concentration was 0.1 g/L, indicating that the washout of anaerobic bacterial biomass within the system was relatively low.

These results indicate that the reduction in VSS is significantly greater than in previous research using a two-phase anaerobic fluidized bed bioreactor with a hydraulic retention time of 48 hours, which yielded a VSS concentration of 7.8 g/L [18]. In a single-phase anaerobic baffled bioreactor with a hydraulic retention time of 7.5 hours, a VSS concentration of 0.85 g/L was obtained [22]. Meanwhile, using a two-phase polyether sulfone-based anaerobic membrane bioreactor with a hydraulic retention time of 20 hours, an MLSS concentration of 2.0-2.5 g/L was achieved [14], and using a two-phase polypropylene-based anaerobic membrane bioreactor with a capacity of 20 L and a hydraulic retention time of 24 hours, an MLSS concentration of 4.8 g/L was obtained [23]. If a single-phase anaerobic hybrid bioreactor equipped with stone media for cell immobilization, with a capacity of 10 L, capable of efficiently converting palm oil mill effluent, good performance, and a hydraulic retention time of 24 hours, was used, a VSS concentration of 4.2 g/L was obtained [24]. Subsequently, using a two-phase anaerobic hybrid bioreactor with a hydraulic retention time of 12 hours, a VSS concentration of 0.8-0.96 g/L was obtained [19, 24]. The two-phase hybrid reactor can reduce VSS levels in a shorter retention time.

3.3. Effect of Hydraulic Retention Time on COD Concentration

Figure 3 shows that the COD value decreases until it reaches a steady state. Under steady-state conditions, the lowest COD concentration, 3000 mg/L, was observed on days 3 to 5 for each retention time. The greatest decrease in value was observed at hydraulic retention

times of 6 and 12 hours, specifically on day 3, whereas at a hydraulic retention time of 18 hours, it began on day 2. This decrease in COD concentration indicates that the longer the hydraulic retention time and the smaller the feed flow rate, the faster the COD value decreases. This is because, with a longer hydraulic retention time, the biodegradation of organic materials in the wastewater proceeds well due to the extended contact time between microorganisms and the wastewater as a substrate. The longer the hydraulic retention time, the better the biodegradation process of organic materials in the wastewater, because the contact between microorganisms and the wastewater is prolonged [25].

In previous research, Ahmad conducted an anaerobic process using a two-phase anaerobic fluidized bed bioreactor system, with the acidogenesis and methanogenesis phases kept separate. In the acidogenesis phase, using a hydraulic retention time of 48 hours with a COD removal efficiency of 57% [6]. Furthermore, Wong et al. reported a single-phase anaerobic baffled bioreactor system, namely the integrated acidogenesis and methanogenesis phases, with a capacity of 10 L, capable of converting palm oil mill liquid waste at a hydraulic retention time of 7.5 hours, achieving a COD removal efficiency of 75% [22].

In addition, Yusnitato et al. conducted acidogenesis using a two-phase polyether sulfone-based anaerobic membrane bioreactor system with a hydraulic retention time of 20 hours, achieving a COD removal efficiency of 59-61% and a COD concentration of 7.2 g/L [14]. Faisal et al. performed acidogenesis in a two-phase, polypropylene-based anaerobic membrane bioreactor system with a hydraulic retention time of 24 hours, achieving a COD removal efficiency of 60.7% [23].

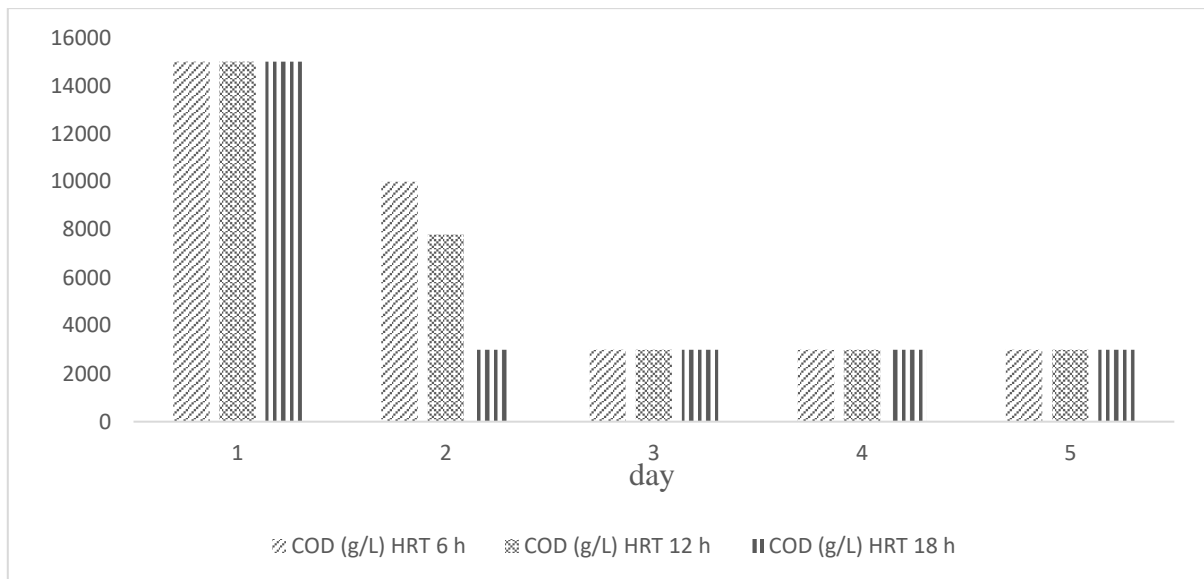


Figure 3. COD concentration profile at varying hydraulic retention times.

Furthermore, Maulina et al. developed a single-phase anaerobic hybrid bioreactor that integrates acidogenesis and methanogenesis. This system had a hydraulic retention time of 24 hours per day and a COD removal efficiency of 85%. The system was able to convert palm oil mill liquid waste with good performance [24].

Ahmad et al. reported a single-phase anaerobic hybrid bioreactor system with integrated acidogenesis and methanogenesis phases, with a capacity of 12.5 L. This system converted palm oil mill effluent with good performance at a high organic loading rate, achieving COD removal efficiencies of 84% for the palm frond media bioreactor and 88% for the palm empty fruit bunch media bioreactor, with a retention time of 24 hours [19].

Furthermore, Anggamulia et al. identified the optimal conditions for a two-phase anaerobic hybrid bioreactor, comprising the acidogenesis and methanogenesis phases, with a capacity of 2.5 m³, in the bioconversion of palm oil mill liquid waste into biogas. The optimum conditions for the anaerobic hybrid bioreactor in the acidogenesis phase were a hydraulic retention time of 12 hours with a COD removal efficiency of 75% [27].

According to Anggamulia et al., the effluent from an anaerobic hybrid bioreactor can be used as liquid fertilizer because it meets the requirements of Indonesian government regulations, with a COD content below 10000 mg/L, which is equivalent to a BOD below 5000 mg/L. The output of this bioreactor can be used as liquid organic fertilizer for land application, thereby reducing the environmental pollution load from palm oil mill wastewater and simultaneously producing liquid fertilizer and biogas [26].

Furthermore, the optimal conditions for a two-phase anaerobic hybrid bioreactor with a capacity of 250 m³/day for the bioconversion of palm oil mill liquid waste into biogas have been identified. The optimal conditions for the acidogenesis phase of the anaerobic hybrid bioreactor are a hydraulic retention time of 24 hours with a COD removal efficiency of 78% [19].

Based on the research data, it can be concluded that hydraulic retention time significantly affects the biodegradation of palm oil mill liquid waste in the acidogenesis phase using an anaerobic hybrid bioreactor. This indicates that the longer the hydraulic retention time, the faster the COD reduction, resulting in a COD concentration of 3000 mg/L or 75% of the initial concentration, which meets the established quality standards. This value also shows that a two-phase acidogenesis-anaerobic hybrid reactor is far more effective at reducing COD levels than other reactor types.

3.4. Effect of Hydraulic Retention Time on Volatile Fatty Acid

Figure 4 shows that volatile fatty acids (VFAs) fluctuate at different hydraulic retention times. The highest VFA concentration at a hydraulic retention time of 12 hours was 6,860 mg/L, and the lowest VFA concentration at a hydraulic retention time of 6 hours was 3,180 mg/L. This occurs because the shorter the hydraulic retention time, the less substrate is broken down and converted into VFA. Therefore, to obtain optimal biohydrogen production, the composition of organic acids in the fermentation process must be balanced. According to Ng et al., the optimal conditions for acidogenesis are

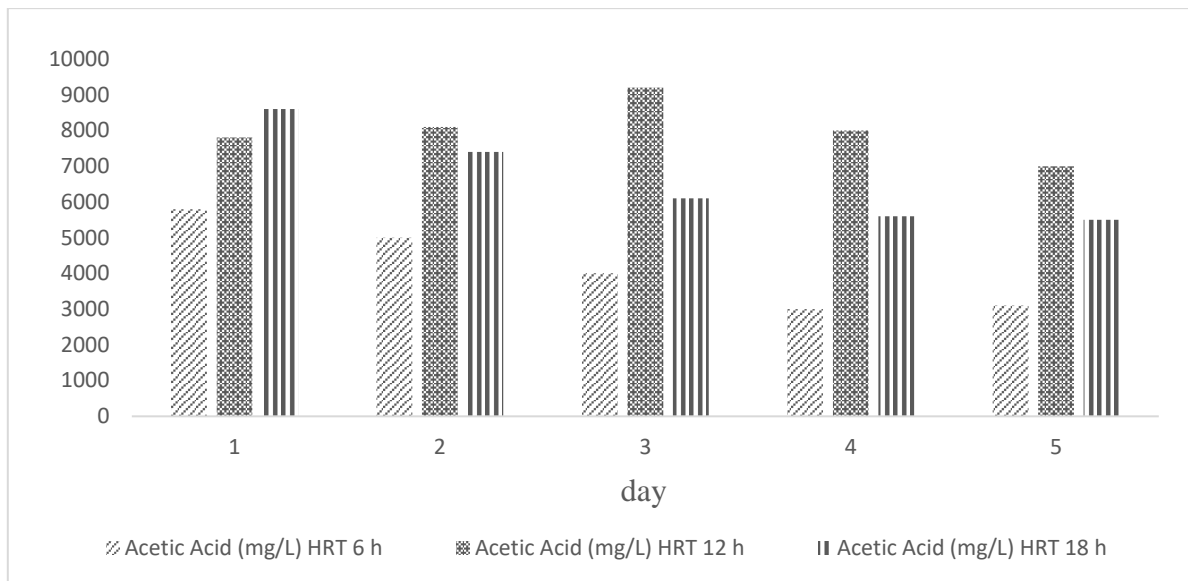


Figure 4. Effect of hydraulic retention time on VFA concentration.

achieved at a VFA concentration of 5,458 mg/L and a pH between 5.5 and 6.5 [27].

In previous research, Zusri et al. conducted acidogenesis using a two-phase anaerobic fluidized bed bioreactor system, with separate acidogenesis and methanogenesis phases. In the acidogenesis phase with a hydraulic retention time of 48 hours, a VFA concentration of 2026 mg/L was obtained [18]. Furthermore, Wong et al. reported a single-phase anaerobic baffled bioreactor system with a capacity of 10 L, integrating acidogenesis and methanogenesis and capable of converting palm oil mill wastewater. With a hydraulic retention time of 7.5 hours, a VFA concentration of 234 mg/L was obtained [22].

Additionally, Yusnitato et al. conducted acidogenesis using a two-phase polyether sulfone-based anaerobic membrane bioreactor system with a hydraulic retention time of 20 hours, achieving a VFA concentration of 1420 mg/L during acidogenesis [14]. Faisal et al. performed acidogenesis using a two-phase polypropylene-based anaerobic membrane bioreactor system with a hydraulic retention time of 24 hours, resulting in a VFA concentration of 520 mg/L [23].

Maulina et al. found a single-phase anaerobic hybrid bioreactor system equipped with stone media for cell immobilization, with a capacity of 10 L, capable of converting palm oil mill effluent with good performance at a hydraulic retention time of 24 hours, achieving a VFA concentration of 4952 mg/L [24]. Furthermore, Anggamulia et al. identified optimal conditions for a two-phase anaerobic hybrid bioreactor with a capacity of 2.5 m³/day for the bioconversion of palm oil mill effluent into biogas. The optimal conditions for the acidogenesis

phase of the anaerobic hybrid bioreactor with a hydraulic retention time of 12 hours resulted in a VFA concentration of 546 mg/L [26].

Furthermore, Ahmad et al. determined the optimal conditions for a two-phase anaerobic hybrid bioreactor with a capacity of 250 m³/day for the bioconversion of palm oil mill liquid waste into biogas. The optimum conditions for the acidogenesis phase of the anaerobic hybrid bioreactor, with a hydraulic retention time of 24 hours, resulted in a VFA concentration of 774-1180 mg/L [19].

Acidogenesis metabolism in the formation of VFA follows what was presented by Nurkholis et al. [28]. Thus, the effect of hydraulic retention time on the biodegradation of palm oil mill wastewater using an anaerobic hybrid bioreactor in the acidogenesis phase showed that the longer the hydraulic retention time, the lower the VFA concentration, which is influenced by the hydrogen concentration in the metabolic system, resulting in a VFA concentration of 5,630 mg/L. This indicates that VFA accumulated because methanogenic bacteria were inactive during the methanogenesis phase, which is responsible for converting VFA into biomethane.

3.5. Effect of Hydraulic Retention Time on Biogas Volume

Figure 5 shows that cumulative biogas production in the continuous stage at a hydraulic retention time of 18 hours is higher than at 6 or 12 hours. The highest biogas production, 5 L/day, was obtained at a hydraulic retention time of 18 hours. Biogas production increased at a hydraulic retention time of 18 hours because longer retention time increases contact between anaerobic

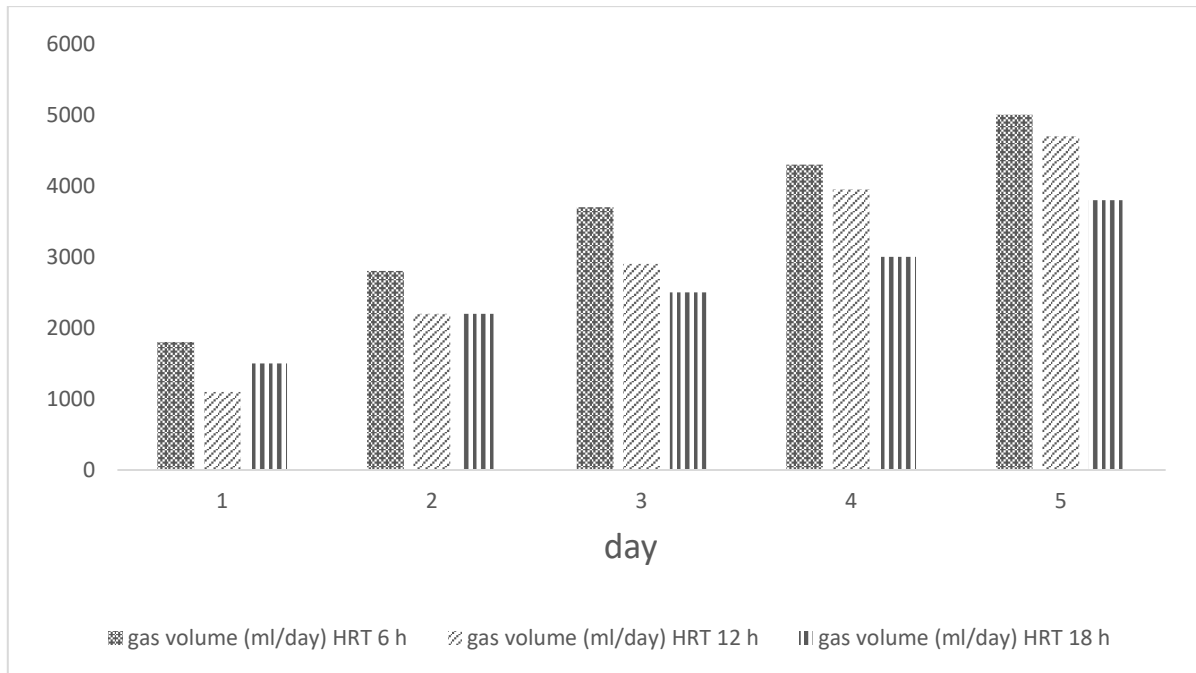


Figure 5. Biogas formation during the continuous stage at varying hydraulic retention times.

bacteria and the substrate, resulting in greater biogas formation.

In previous research, Zusri et al. conducted acidogenesis using a two-phase anaerobic fluidized bed bioreactor system, with separate acidogenesis and methanogenesis phases and a hydraulic retention time of 48 hours, producing a biogas volume of 24000 mL/day [18]. Subsequently, Wong et al. reported a single-phase anaerobic baffled bioreactor system with a capacity of 10 L, capable of converting palm oil mill wastewater at a hydraulic retention time of 7.5 hours, yielding a biogas volume of 12600 mL/day [22]. Furthermore, Yusnitato et al. performed acidogenesis using a two-phase polyether sulfone-based anaerobic membrane bioreactor system with a hydraulic retention time of 20 hours, producing a biogas volume of 310 L/day. In Ahmad's study, acidogenesis was conducted using a two-phase polypropylene-based anaerobic membrane bioreactor system with a hydraulic retention time of 24 hours, producing 9.5 L/day of biogas [14]. Faisal et al. performed acidogenesis using a polyether sulfone-based anaerobic membrane bioreactor system with a hydraulic retention time of 48 hours, producing 134000 mL/day of biogas [23]. Wong et al. developed a single-phase anaerobic baffled bioreactor system with a capacity of 10 L that effectively converted palm oil mill effluent, achieving good performance at an organic loading rate of 3.2 g/L.day, a hydraulic retention time of 20 hours, and biogas production of 5600 mL/day [22].

Maulina et al. found a single-phase anaerobic hybrid bioreactor system equipped with stone media for cell

immobilization, with a capacity of 10 L, capable of converting palm oil mill effluent with good performance at a hydraulic retention time of 24 hours, yielding a biogas volume of 195 mL/day [24].

Furthermore, Anggamulia et al. determined the optimal conditions for a two-phase anaerobic hybrid bioreactor with a capacity of 2.5 m³/day for the bioconversion of palm oil mill effluent into bioethanol. The optimum conditions for the acidogenesis phase of the anaerobic hybrid bioreactor, with a hydraulic retention time of 12 hours, resulted in a specific methane production of 46 m³/kg COD [26].

Optimal conditions for a single-phase anaerobic hybrid bioreactor with a capacity of 250 m³/day for the bioconversion of palm oil mill liquid waste into biogas fuel have been identified. The optimal conditions for the acidogenesis phase of the anaerobic hybrid bioreactor with a hydraulic retention time of 24 hours resulted in biogas production of 5 L/minute [19]. Thus, the effect of hydraulic retention time on the biodegradation of palm oil mill liquid waste during the acidogenesis phase of the anaerobic hybrid bioreactor shows that the longer the hydraulic retention time, the higher the biogas production; the highest hydraulic retention time of 18 hours yielded a biogas volume of 5 L/day.

3.6. Effect of Hydraulic Retention Time on Biohydrogen Composition

Figure 6 shows that the highest biohydrogen concentration was obtained at a hydraulic retention time of 18 hours, reaching 64%. Biohydrogen concentration

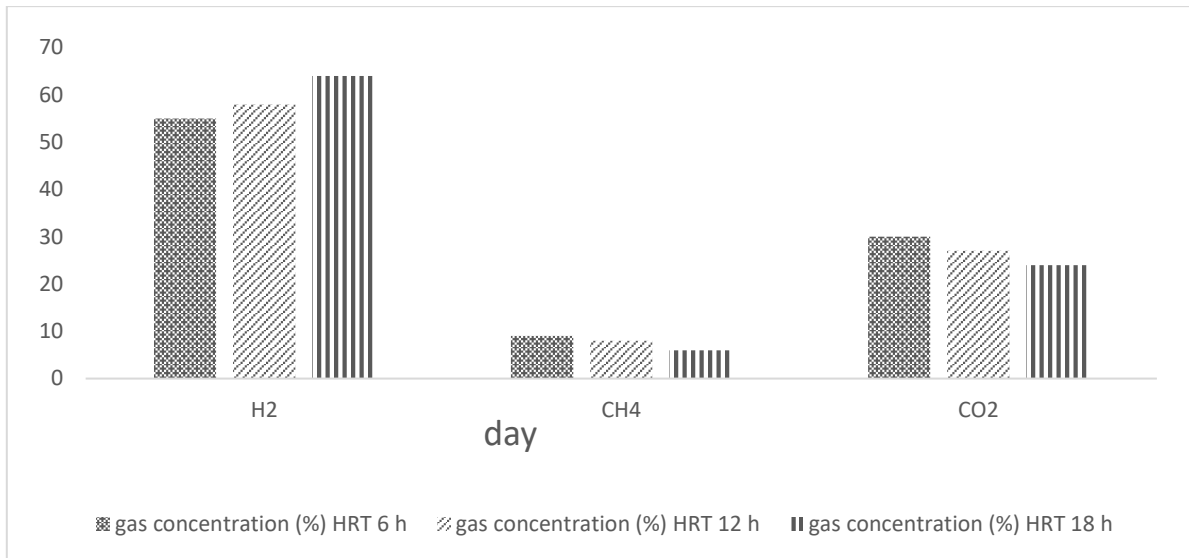


Figure 6. Concentration of H₂, CH₄, and CO₂ gasses during the continuous stage at varying hydraulic retention times.

Table 2. Comparative study of bioreactor systems in biogas production from POME.

No.	Bioreactor	System Phase	Biogas composition (%)			Ref.
			H ₂	CO ₂	CH ₄	
1.	Anaerobic Membrane Bioreactor POLIETER SULFON	Methanogenesis	Not detected	42.5	57.5	[14]
2.	Anaerobic Membrane Bioreactor POLIPROPILEN	Methanogenesis	Not detected	96.2	3.8	[23]
3.	Anaerobic Fluidized Bed Bioreactor	Methanogenesis	Not detected	28	72	[18]
4.	Anaerobic Hybrid Bioreactor	Methanogenesis	Not detected	54	6.6	[19]
5.	Anaerobic Hybrid Bioreactor	Acidogenesis	64	5.3	22	This result

indicates the amount of biohydrogen that can be produced per unit weight of volatile solids (VS), and the effectiveness of the substrate being converted into biohydrogen because the concentration and volume of biogas are related to the efficiency of microorganism activity in breaking down the substrate into biohydrogen, compared to the production of organic acids or cell maintenance [28]. Biohydrogen concentration is highly influenced by the biodegradation of VFA, which are converted into biomethane. The lowest biohydrogen concentration was 55% at a hydraulic retention time of 6 hours, and the highest was 64% at 18 hours. This is because the hydraulic retention time affects the formation of VFA and biohydrogen. Biohydrogen is formed during the acidogenesis phase, which converts POME substrate into H₂, CH₄, and CO₂.

In previous research, Wong et al. found a single-phase anaerobic baffled bioreactor system, with integrated acidogenesis and methanogenesis phases, having a capacity of 10 L, capable of converting palm oil mill effluent with a hydraulic retention time of 7.5 hours, yielding a biogas volume of 12600 mL/day with a methane gas concentration of 72.2% [22]. Subsequently, Yusnitato et al. conducted an acidification process using a two-phase polyether sulfone-based anaerobic membrane bioreactor system with a hydraulic retention

time of 20 hours, and observed no detectable biohydrogen concentration [14]. Faisal et al. conducted an acidification process using a two-phase polypropylene-based anaerobic membrane bioreactor system with a hydraulic retention time of 24 hours. They found that the biohydrogen concentration was undetectable [23].

Furthermore, Anggamulia et al. identified optimal conditions for a two-phase anaerobic hybrid bioreactor with a capacity of 2.5 m³/day for the bioconversion of palm oil mill liquid waste into biogas. The optimal conditions for the acidogenesis phase of the anaerobic hybrid bioreactor, with a hydraulic retention time of 12 hours, yielded a specific methane production of 46 m³/kg COD [26]. Additionally, optimal conditions were obtained for a single-phase anaerobic hybrid bioreactor with a capacity of 250 m³/day in the bioconversion process of palm oil mill liquid waste into biogas. This anaerobic hybrid bioreactor converted liquid waste with a hydraulic retention time of 24 hours, resulting in methane and CO₂ concentrations of 54% and 6.6%, respectively [19]. A comparison study of bioreactor systems is presented in Table 2.

The pathway for the anaerobic decomposition of complex organic matter into biohydrogen and CO₂

follows that presented by Pavlostathis and Giraldo-Gomez [29]. Hydraulic retention time positively influences the biodegradation of palm oil mill liquid waste in the acidogenesis phase of an anaerobic hybrid bioreactor, indicating that longer hydraulic retention time yields greater biohydrogen volume. The highest biohydrogen concentration achieved was 64%. This shows that the acidogenesis phase produces relatively more biohydrogen than biomethane, due to the inhibition of methanogenic bacteria during the methanogenesis phase.

4. Conclusions

The COD concentration at an 18-hour HRT was 3000 mg/L; the solid concentration (VSS) decreased with increasing HRT in the bioreactor, reaching 0.10 g/L at 18 hours. The highest VFA concentration at an 18-hour hydraulic retention time was 5500 mg VFA/L; biogas production in the continuous stage was 5 L/day. The highest H₂ gas concentration was obtained at an 18-hour hydraulic retention time, reaching 64%.

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Informed Consent Statement: Not applicable.

Data Availability Statement: The data that support the findings of this study are available from the corresponding author upon reasonable request.

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