HYDROGEN PRODUCTION THROUGH ANAEROBIC FERMENTATION OF CRUDE GLYCEROLAND ITS USE FOR FUEL CELL POWER GENERATION

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ABSTRACT

A way of processing and utilising crude glycerol, a byproduct from biodiesel fuel (BDF) production, is described. Anaerobic fermentation of crude glycerol as substrate with an inoculum from easily obtainable cattle excrement led to the suppression of methane production and enhanced hydrogen fermentation. Biohydrogen production with H_2/glycerol (in crude glycerol) molar ratio of 0.17 was achieved. Characteristics of this hydrogen fermentation were investigated, and factors contributing to this phenomenon were identified as synergy among the impurities present in BDF byproduct glycerol, viz., methanol and KOH, with pure glycerol for the production of H_2. The usability of this biohydrogen for direct power generation without further purification or processing through a polymer electrode fuel cell (PEFC) was demonstrated, with a possible power generation of 117 J/g of crude glycerol.

Keywords: *change to: anaerobic fermentation, crude glycerol, hydrogen, fuel cell*

1 INTRODUCTION

The utilisation of byproduct biomass often poses difficulties, some of which are due to the characteristics added to its intrinsic nature through the process it went through. Crude glycerol is one such byproduct, discharged from the biodiesel fuel (BDF) production process through transesterification of vegetable oil with methanol. The biodiesel fuel production process necessarily produces crude glycerol byproduct, which amounts to ca. 10% (w/w) of BDF produced. Worldwide production of BDF as of 2020 amounts to 46.8 billion L/yr [1], which gives a simple estimate of 5.0 billion L/yr crude glycerol production. The byproduct crude glycerol from the BDF production process contains unreacted methanol, catalyst KOH, free fatty acids, soap and/or oil cake (or oil meal). These impurities make using the byproduct crude glycerol for any purpose difficult. Instead of separating the impurities and purifying glycerol, we considered utilising it as an energy source by converting it to gas fuel by anaerobic fermentation (refer to Refs. [2],[3] for the preliminary results).

There exists a variety of research work on processing and converting crude glycerol by anaerobic digestion [2]–[16], some with a significant amount of hydrogen production. However, the digestion of crude glycerol with easily obtainable Inoculum from cattle excrement has not been reported except in our study [2]-[3], and the details of the fermentation, including the factors affecting the hydrogen production with this Inoculum is not well known.

Anaerobic methane fermentation (or digestion) is characterised by its possibility of energy recovery from waste biomass. It has already become a common renewable energy production process from municipal wastes, livestock excrement and sewage sludge, with the produced methane gas (biogas) being utilised as fuel. Anaerobic digestion is a biotic process collection composed of the following steps: hydrolysis, acidogenesis, acetogenesis and methanogenesis. In the first step, biomass macromolecules are hydrolysed into simpler water-soluble molecules. In the second step, products from the first step are further converted to volatile fatty acids, ammonia, CO_2 and H_2S. The main products of the third step are acetic acid, CO_2 and H_2. In the last step, methanogen produces a mixture of CH_4 and CO_2 (commonly called biogas) from the products of previous steps and H_2. The culture (or flora) that causes the entire anaerobic digestion may differ depending on its source. It may also change during fermentation, adapting to the substrate, as well as digestion conditions such as temperature.

In this paper, we have attempted anaerobic fermentation of crude glycerol as a substrate for biohydrogen production using an easily obtainable cattle excrement as an inoculum. Preliminary results of this experiment series have already been reported [2]-[3], and the present paper expands and concludes those results reported, with fuel cell power generation demonstration using the obtained hydrogen. Characteristics of the hydrogen fermentation process with crude glycerol were investigated further in detail, and factors contributing to biohydrogen production were identified and confirmed. The usability of this biohydrogen for direct power generation through a polymer electrode fuel cell (PEFC) was demonstrated without further biogas processing.

2 MATERIALS AND METHODS

Due to the serial nature of this work, a major part of the fermentation materials and fermentation procedures closely follow our previous publication [2]-[3]. Nevertheless, for the sake of readers, the essential part is reproduced below, with some additions in particular to the present paper.

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2.1 Raw Materials

Crude glycerol is obtained from a biodiesel production plant (Nanko Co. Ltd., Japan, model ME-100, 100 L/day) installed at the Faculty of Engineering, University of Yamanashi, Japan. The characteristic of this BDF plant is that it does not wash BDF to separate glycerol: instead, it uses a separation-by-sedimentation technique and settled crude glycerol is withdrawn from the bottom of the reactor tank. The catalyst for transesterification is potassium hydroxide, and as a result, crude glycerol contains a high concentration of potassium hydroxide and is highly alkaline. The pH and the composition of the crude glycerol employed here are listed in Table 1. Cattle excrement is used as starting Inoculum for the present work was obtained from Yamanashi Prefectural Dairy Experimental Station, Hokuto City, Japan.

Table 1 Characteristics and composition of crude glycerol employed in the present study

pH	12.7
Compositions	
glycerol	43.0 *wt%*
methanol	13.0 *wt%*
water	2.5 *wt%*
palmitate	4.1 *wt%*
oleate	20.0 *wt%*
steroids	0.3 *wt%*
ash	8.6 *wt%*
potassium	4.2 *wt%*
sodium	72 *wt ppm*
phosphorous	10 *wt ppm*

In order to determine the effect of individual components contained in the employed crude glycerol, pure chemical counterparts were purchased and used: glycerol (purity 99.0%, Kanto Chemicals), potassium hydroxide (purity 99. 5%, Wako Pure Chemicals), oleic acid (purity 99.5%, Wako Pure Chemicals) and methanol (purity 99.8%, Wako Pure Chemicals).

2.2 Inoculum Preparation

The cattle excrement was first sieved with a screen with a 1.7 mm opening in order to remove straw mulching and other debris. Two (2) kg of the sieved cattle excrement was mixed with water of the same weight, agitated, and its pH was measured. This is named Inoculum A. Inoculum A prepared two types of inoculums, as shown in Table 2.

For Inoculum B preparation, Inoculum A was thrown into a 10 L polyethylene tank. The air in the headspace was replaced with nitrogen, a Tedlar Bag was connected, and shaking the entire assembly commenced with a temperature-controlled water bath shaker at 37°C. The amount of gas produced was measured once a day by the water displacement method, until when the inoculum substrate was almost exhausted, i.e., gas production had levelled off. Inoculum C was prepared by thermally treating Inoculum B at 80°C for 2 h.

2.3 Fermentation Procedures

Each Inoculum prepared as above was then divided into smaller batches, and crude glycerol or other chemicals were added as substrate. Again, the air in the headspace of each reactor (flask) was replaced with nitrogen, a Tedlar Bag was connected, and shaking the entire assembly commenced with a temperature-controlled water bath shaker. A mesophilic digestion temperature of 35°C or 37°C was employed throughout the present experiment. The water displacement method measured the amount of gas produced with appropriate intervals. Gas composition analysis was done using a Shimadzu GC-8A gas chromatograph equipped with a thermal conductivity detector.

2.4 Power Generation with Fuel Cell

For power generation demonstration, a miniature PEFC model HPFCRD0830 (13 cells, 8 V, 30 W output) manufactured by FC-R&D Co. Ltd. Was employed, with a diaphragm pump THOMAS 3003 VD BLDC manufactured by Gardner Denver Medical, U.S.A. Generated power was expended to run an N-gauge model railroad in KATO, Japan.

3 RESULTS AND DISCUSSION

3.1 Biogas Production Characteristics of Crude Glycerol as Substrate with Inoculum A

Firstly, the crude glycerol characteristics as a substrate were examined. Inoculum A (100 g) fermentation characteristics were examined in the first paper of this series [2]. It is used as a starting culture, and various amounts of crude glycerol (Run A(1) through A(7) with none, 0.2 g, 1.0 g, 2.0 g, 6.0 g, 10.0 g and 15.0 g of crude glycerol) was added to each reactor. The initial pH of each reactor varied slightly due to the alkalinity of the crude glycerol, from 7.6 (Run A(1) through A(4) to 7.8 (Run A(6) and A(7). Fermentation was performed at 35°C for 188 h with a constant-temperature water bath shaker. At the end of the fermentation, the pH of all

Table 2 Three types of Inoculum prepared

	Method of preparation	Total solid(TS)/wt%	Volatile solid(VS)/wt%
Inoculum A	Cattle excrement is sieved (2 kg after sieving) and mixed with 2 L of water	5.99	5.07
Inoculum B	Inoculum A has anaerobically fermented at 37°C for ca. 150 h until the substrate in the Inoculum is almost exhausted, i.e., biogas production has levelled off	5.33	4.54
Inoculum C	Inoculum B is thermally treated at 80°C for 2 h	NA*	NA*

the reactors was 6.4. Figure 1 shows the time course of biogas accumulation, and Figure 2, the overall biogas composition for each run. The details of each run are as follows:

1. Run A(1) with no substrate addition,
2. Run A(2) with 0.2 g of crude glycerol,
3. Run A(3) with 1.0 g of the same,
4. Run A(4) with 2.0 g,
5. Run A(5) with 6.0 g
6. Run A(6) with 10.0 g, and
7. Run A(7) with 15.0 g of crude glycerol.

First of all, it is noted in Figure 1 that in the present experimental series A(1) through A(7), acclimation duration of 20 to 40 h is present at the beginning of fermentation, contrary to the fermentation of Inoculum A only [2]. A close examination of Figure 1 further indicates that biogas production behaviour at digestion time around 70 h seems to group the present seven runs into two: the lesser amount of biogas production below 50 mL (Runs A(1) through A(4), with none or a lesser amount of added crude glycerol), and a larger amount of biogas production above 80 mL (Runs A(5) through A(7), with a larger amount of added crude glycerol). The addition of a certain amount of crude glycerol (Runs A(5) through A(7) does increase total biogas production compared to unadded Run A(1) (navy diamonds), indicating that crude glycerol does work as a substrate for anaerobic fermentation.

The difference between these two groups becomes more prominent in their gas compositions (Figure 2): biogas from Runs A(1) through A(4) almost exclusively consist of CH_4 and CO_2 with a trace amount of H_2, while biogas from Runs A(5) through A(7) contain a larger amount of H_2. From Figure 2, it is read that: 1) crude glycerol up to 2.0 g/100 g inoculum progressively suppresses the overall methane fermentation without a substantial effect on biogas composition (Runs A(1) through A(4), 2) crude glycerol above 6.0 g/100 g inoculum gives a large amount of H_2, 3) within this range of crude glycerol concentration, CH_4 production is progressively suppressed while H_2 production is gradually increased (Runs A(5) and A(6), 4) H_2 production goes through a maximum, as in the case of Inoculum B reported earlier [2], with the amount of crude glycerol addition and 5) at the limit of 15 g/100 g inoculum of crude glycerol addition methane production is completely suppressed.

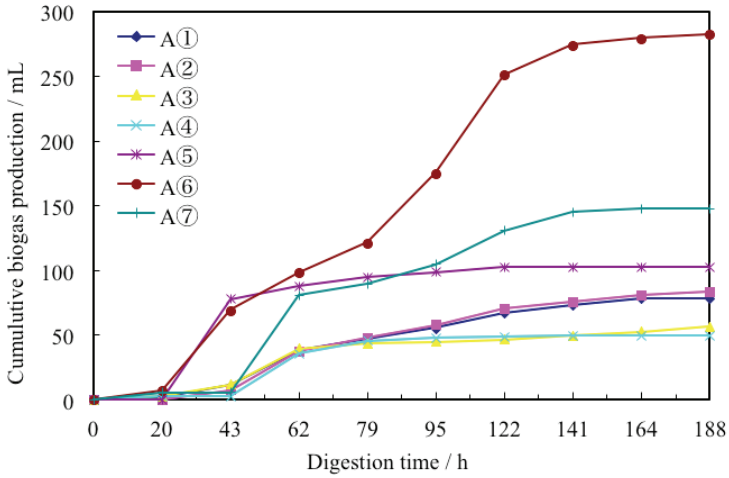


Figure 1 Time course of biogas accumulation with 100 g of Inoculum A and various amounts of crude glycerol as substrate

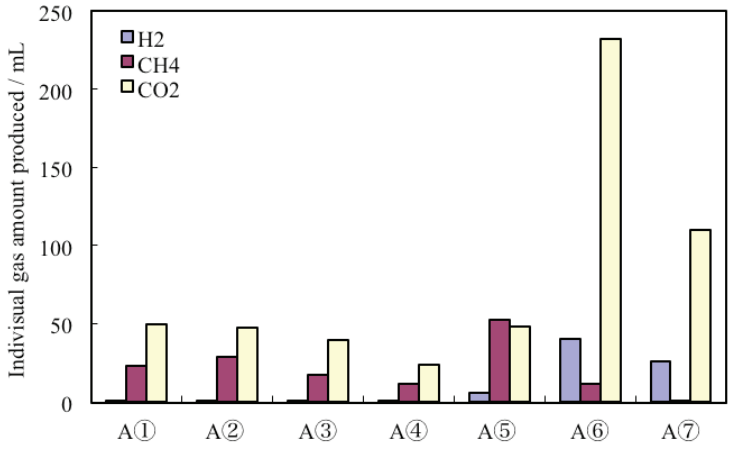


Figure 2 Overall biogas compositions produced with 100 g of Inoculum A and various amounts of crude glycerol as substrate

3.2 Biogas Production Characteristics of Pure Glycerol as Substrate with Inoculum A

The above results of crude glycerol on anaerobic fermentation prompted us to examine the controlling factor of this effect, regarding the chemical species present in the crude glycerol [2]-[3]. The natural first candidate is the major component of crude glycerol, pure glycerol, since the crude glycerol comprises 43 wt% of glycerol (cf. Table 1).

Here Inoculum A (100 g) was employed as a starting inoculum, and various amounts of pure glycerol (Run B(1) through B(7) with none, 0.2 g, 1.0 g, 2.0 g, 6.0 g, 10.0 g and 15.0 g/100 g inoculum) was added to each reactor. The initial pH of each reactor varied slightly due to the addition of pure glycerol, from 7.6 (Run B(1) to 7.2 (Run B(7). Fermentation was performed at 35°C for 188 h in a constant-temperature water bath shaker. At the end of the fermentation, the pH of all the reactors was 6.4.

The time course of biogas production with pure glycerol was similar to the ones presented in the previous section with crude glycerol: a weak acclimation period was observed in the first 20–40 h of digestion, biogas production picked up after that and arrived at a plateau by the end of the experiment. The difference in the amount of produced biogas among the B series runs (with pure glycerol) was less pronounced than the previous A series runs (with crude glycerol). However, Run B(7), with the highest amount of pure glycerol, showed a notably low amount of biogas production compared to other runs in this series.

Figure 3 shows the overall biogas composition produced with 100 g of Inoculum A and various amounts of pure glycerol were added as substrate. The effect of pure glycerol addition to Inoculum A on biogas composition (Figure 3) is similar to that of crude glycerol addition in many respects. Similarities are: 1) pure glycerol up to 1.0 g/100 g inoculum progressively suppresses the overall methane production (Figure 3, Runs B(1) through B(3), 2) pure glycerol above 2.0 g/100 g inoculum produces H_2, and 3) H_2 production goes through maxima with crude or pure glycerol addition. The different aspects of pure glycerol in Inoculum A (Figure 3) from the crude glycerol (Figure 2) are: 1) H_2 and CH_4 production appears to occur concomitantly in the range of higher amounts of pure glycerol addition (2.0 g to 15.0 g/100 g inoculum, Runs B(4) through B(7), instead of hydrogen production taking over that of methane at the highest amount of crude glycerol addition (Run A (7) and 2), no complete suppression of CH_4 production was observed with an increasing amount of pure glycerol but at the highest end of hydrogen production is suppressed. The lower shift in glycerol amount for fermentation characteristics, such as the appearance of H_2, may be due to the fact that the actual amount of glycerol in crude glycerol is only about half of the added crude glycerol: for the same added weight, pure glycerol will have more than twice as much glycerol compared to crude glycerol.

The persistent appearance of CH_4 at higher glycerol concentration for pure glycerol addition (Figure 3), or its complete suppression at higher crude glycerol concentration (Figure 2), cannot be explained only by the glycerol concentration difference, and other possibilities need to be considered. In the next section, a search for crude glycerol components responsible for hydrogen production was attempted with Inoculum B.

3.3 Crude Glycerol Component Effective for Hydrogen Production with Inoculum B

In order to identify the chemical component in the crude glycerol that produces H2 and suppresses the CH4 production, the effect of each major component present in the present crude glycerol (Table 1) and their combinations are examined. For this purpose, Inoculum B, with the most substrate in Inoculum A depleted by anaerobic fermentation. It is used as an inoculum starter to separate and accentuate the crude glycerol characteristics and its constituents as the substrate for anaerobic fermentation from the substrate in the Inoculum itself.

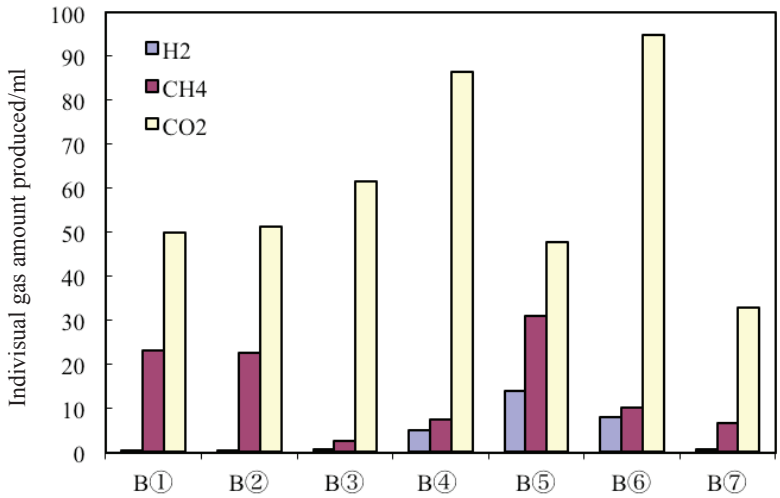


Figure 3 Overall biogas compositions produced with 100 *g* of Inoculum A and various amount of pure glycerol as substrate. Pure glycerol added: none, 0.2 g, 1.0 g, 2.0 g, 6.0 g, 10.0 g and 15.0 g for Run B(1) through B(7), respectively. Fermentation was performed at 35°C for 188 h.

The effect of the added crude glycerol amount to Inoculum B in the range of 0 to 14 wt% has been studied in detail [2]. Briefly, at the fermentation temperature of 37°C and the starting pH adjusted to 5.59, which is the inherent pH of Inoculum B, the study found that there exist 2 maxima for added crude glycerol amount in H_2 production: at ca. 3 wt% and at ca. 7 wt% [2], the latter giving higher H2 production than the former.

Thus, choosing the optimum crude glycerol concentration to be at the higher peak of 7 wt%, major components in the crude glycerol, viz., pure glycerol, methanol, KOH and oleic acid, and their combinations were tested for their effect on anaerobic hydrogen production as listed in Table 3 (preliminary experiments with a similar but narrower scope were carried out previously [3]). Duplicate experiments were performed for each component or combination, and the data were averaged for the two. Runs C(1) (with crude glycerol) and C(2) (with pure glycerol) are to be compared with the experiments run with Inoculum A, viz., Runs A(6) (Figure 2) and B(5) (Figure 3), respectively. Figure 4 summarises the biogas composition obtained.

Firstly, it is noted in Figure 4, Run C(1), that crude glycerol appears to give a similar effect on fermentation characteristics in Inoculum B, as it did in Inoculum A (Figure 2): it suppressed methane production and enhanced hydrogen production. With Inoculum B, however, the effect is more pronounced than in Inoculum A, probably because in Inoculum B, most of the substrate is already exhausted, and the impact of the new substrate becomes more conspicuous. Thus, with Inoculum B, CH4 production is almost entirely suppressed at the crude glycerol concentration of 7.0 wt%, while H_2 production was significantlyenhanced to the amount comparable to that of CO_2 (Run C(1). The effect of pure glycerol, run C(2) is similar to the one with Inoculum A (Figure 3): CH_4 production is suppressed and H_2 production is enhanced, but not to the extent found for crude glycerol.

Figure 4 also shows that among the four major components of crude glycerol (viz., pure glycerol, methanol, oleic acid and KOH (cf. Table 1), methanol (Run C(3), oleic acid (Run C(4), KOH (Run C(5) and methanol–KOH combination (Run C(6) per se do not contribute to H_2 production by anaerobic fermentation at all

Table 3 The effect of crude glycerol components on the anaerobic fermentation of Inoculum B

Run #	Amount of Inoculum B/g	Crude glycerol components added/g					pH after fermentation
		Crude glycerol	Pure glycerol	Methanol	Oleic acid	KOH	
C(1)	93.0	7.0	-	-	-	-	5.04
C(2)	93.0	-	3.0	-	-	-	4.59
C(3)	93.0	-	-	0.9	-	-	5.54
C(4)	93.0	-	-	-	1.4	-	5.27
C(5)	93.0	-	-	-	-	0.3	5.52
C(6)	93.0	-	-	0.9	-	0.3	5.49
C(7)	93.0	-	3.0	0.9	-	-	4.58
C(8)	93.0	-	3.0	-	1.4	-	4.60
C(9)	93.0	-	3.0	-	-	0.3	4.81
C(10)	93.0	-	3.0	0.9	-	0.3	4.85

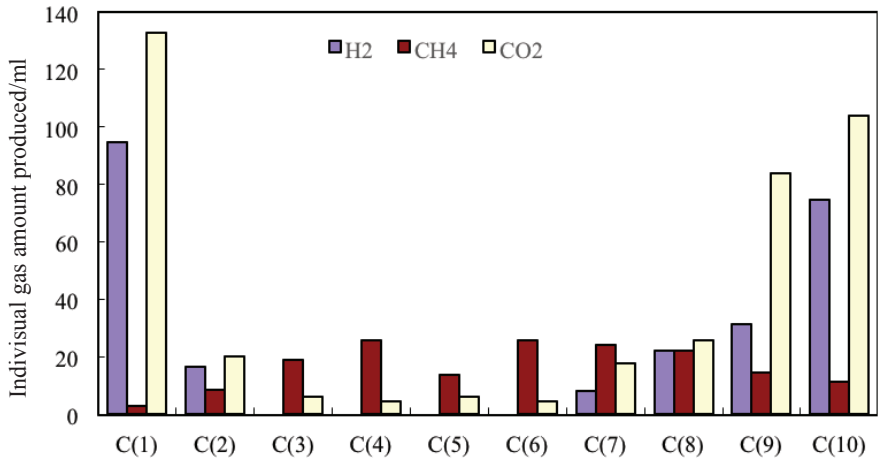


Figure 4 Biogas compositions produced with 93 g of Inoculum B and various amounts of crude glycerol components as substrate (for details see Table 3). Fermentation time is 144 h, except the Runs C(4) and C(8) which were run for 120 h

(previous preliminary experiments with Inoculum B also found that methanol, KOH, feed vegetable oil, and product biodiesel per se do not contribute to H_2 production [3]). Furthermore, the addition of methanol or oleic acid with pure glycerol (Runs C(7) and C(8) do not seem to have any obvious effects in terms of biogas composition (cf. Run C(2), negative or positive.

However, the addition of pure glycerol and KOH to Inoculum B (Run C(9) enhanced the H_2 production above the case of pure glycerol addition only (cf. Run C(2), and adding methanol to this KOH-pure glycerol combination further enhanced the H_2 production (Run C(10), almost to the level of crude glycerol addition (Run C(1). The effect of components present in crude glycerol toward anaerobic fermentation is complex, and methanol and KOH exert synergetic contribution to pure glycerol for the production of H_2.

3.4 Crude Glycerol Component Effective for Hydrogen Production with Inoculum C

It may be interesting to examine whether the similar effects of crude glycerol and its components would be observed for an inoculum tailored for hydrogen fermentation. For this purpose, Inoculum C is employed as starting Inoculum, which is Inoculum B heat-treated at 80°C for 2 h in order to sterilise methanogens while keeping other spore-forming flora.

The compositions of substrates tested are listed in Table 4, and the fermentation results are shown in Figure 5. Inoculum C showed slower fermentation progress, not reaching the gas production plateau even after 400 h of fermentation, compared to Inoculum A, which reached a plateau at ca. 180 h and Inoculum B at ca. 250 h. Nevertheless, its fermentation characteristic in comparison to Inoculums A and B are quite apparent in Figure 5: it gives almost no CH_4. The effects of substrate compositions are similar to that on Inoculum B: crude glycerol is effective in producing H_2 (Run E(1)), and it is further enhanced by the addition of KOH (Run E(4)) or methanol (Run E(5)) to crude glycerol. It is interesting to note that in combination with pure glycerol, the amount of added KOH is important (cf. Runs E(6) and E(7)), but the addition of methanol is not as effective (cf. Runs E(7) and E(8)).

3.5 Continuous Production of Hydrogen by Anaerobic Fermentation

As the batch fermentation conditions optimised in Section 3.3, conditions for continuous production of hydrogen by anaerobic

Table 4 The effect of crude glycerol components on the anaerobic fermentation of Inoculum C

Run #	Amount of Inoculum C/g	Crude glycerol components added/g					pH after fermentation
		Crude glycerol	Pure glycerol	Methanol	Waste oil	KOH	
E(1)	100.0	10.0	-	-	-	-	7.4
E(2)	100.0	20.0	-	-	-	-	7.6
E(3)	100.0	10.0	5.8	-	-	-	7.3
E(4)	100.0	10.0	-	-	-	0.26	7.8
E(5)	100.0	10.0	5.8	-	-	0.26	7.8
E(6)	100.0	-	5.8	-	-	0.26	7.3
E(7)	100.0	-	5.8	-	-	0.52	8.0
E(8)	100.0	-	5.8	2.5	2.0	0.26	7.3

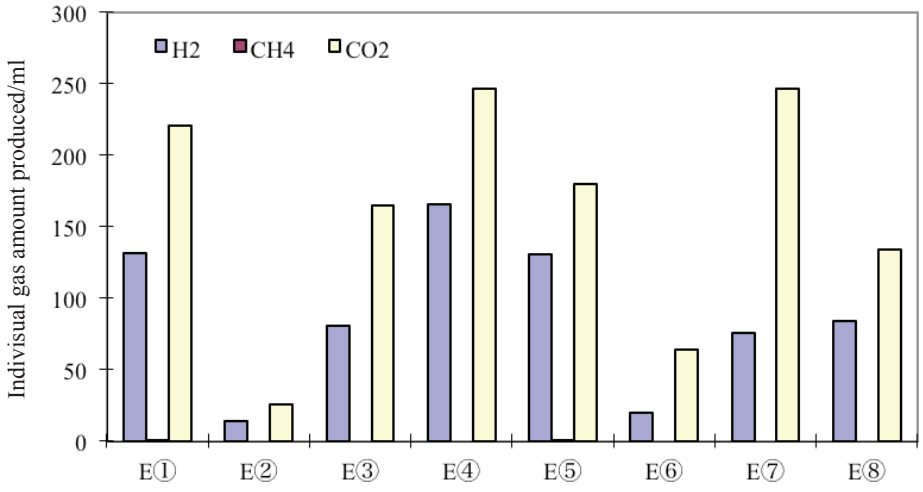


Figure 5 Biogas compositions produced with 100 g of Inoculum C and various amount of crude glycerol components as substrate (for details see Table 4). Fermentation time is 432 h

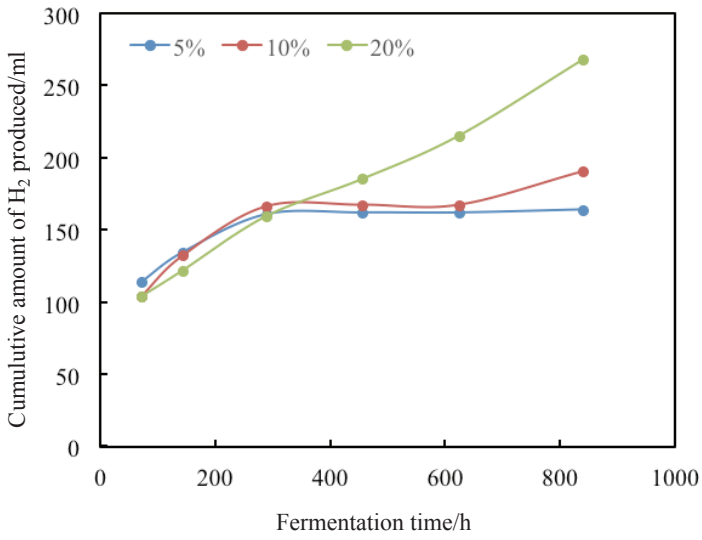


Figure 6 Time course of H_2 accumulation. Starting with 93 g of Inoculum B and 7 g of crude glycerol, the same substrate is added at different intervals

fermentation of crude glycerol was searched. For this purpose, Inoculum B was employed, and the following three sets of experiments were performed, two runs for each set with obtained data are averaging. Starting with 93 g of Inoculum B and 7 g of crude glycerol (total of 100 g), 5 wt% of the starting substrate (Inoculum 4.65 g and crude glycerol 0.35 g), 10 wt% (Inoculum 9.30 g and crude glycerol 0.70 g) or 20 wt% (Inoculum 18.60 g and crude glycerol 1.40 g) were thrown into the reactor at 72 h, 144 h, 288 h, 456 h and 624 h.

As may be found in Figure 6, the accumulated amount of hydrogen hit a plateau at ca. 300 h for 5 wt% and 10 wt% incremental additions. In contrast, with 20 wt% addition, H_2 was continuously produced with a steady rate of ca. 7.7 mL H_2/day. In terms of added crude glycerol, a total of 268.4 mL H_2 was produced out of the total of 14.0 g of added crude glycerol (6.0 g of glycerol), resulting in the H_2/glycerol molar ratio of 0.17.

3.6 Fuel Cell Power Generation

Using nine batches of 930 g Inoculum B and 70 g of crude glycerol, a large-scale batch of hydrogen production is attempted for the purpose of power generation through the fuel cell. Desulfurising agent GS-6 and zeolite as ammonia adsorbent were tucked inside the Tedlar Bag used for biogas collection in order to remove H_2S and NH_3 molecules that are harmful to fuel cell electrode catalysts. After 24 h, a total of 7 L of biogas was collected, which composition is listed in Table 5. Lesser H_2/glycerol molar ratio obtained is here (0.031) compared to the one found in Section 3.4 is due to a much shorter digestion time as well as batch operation with no addition of substrate employed here.

Table 5 Composition of biogas obtained by large-scale batch fermentation

	Produced gas amount/L
H_2	2.07
CH_4	0.07
CO_2	4.87

Figure 7 shows a setup for power generation with the present biohydrogen. The system is stand-alone and connected to no external power supply. In order to start power generation, biohydrogen in the Tedlar Bag must first be supplied to the fuel cell (PEFC) by pressing the Tedlar Bag by hand. Once power is generated, the diaphragm pump will provide biohydrogen from the bag to the PEFC unit until the biohydrogen in the bag is exhausted. The major part of generated power (except the one consumed at the diaphragm pump) will be supplied to the N-gauge model railroad to run the model bullet train.

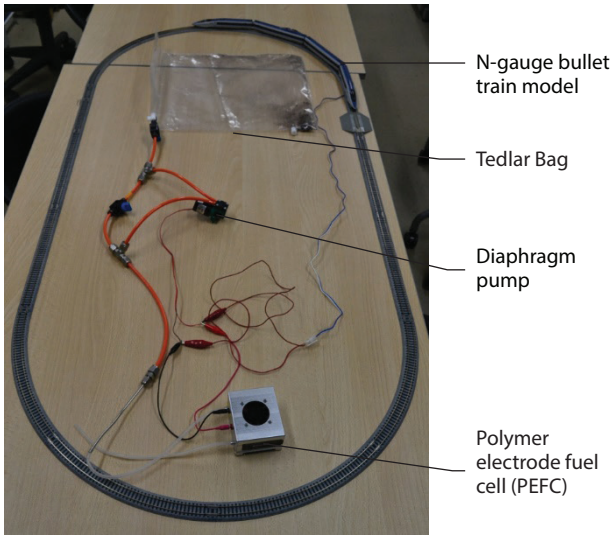


Figure 7 A setup for the power generation with the present biohydrogen. It consists of a Tedlar Bag that contain biohydrogen, a diaphragm pump, fuel cell (PEFC) and an N-gauge model railroad

The open circuit PEFC voltage with the present biohydrogen (for composition, see Table 4) was 8.0 V, and with the diaphragm pump on, 7.7 V. Seven litters of present biohydrogen were able to run the trained model for 7 minutes. After all, 630 g crude glycerol (271 g glycerol) can convert to the electricity of 12.6 kJ, or 46.5 J/g glycerol (in crude glycerol). Given the higher conversion efficiency of glycerol to hydrogen found in Section 3.4 (H2/glycerol molar ratio of 0.18), the present methodology would convert one g of crude glycerol into 117 J of electricity.

4 CONCLUSIONS

Energy conversion of crude glycerol, a byproduct from biodiesel production, was attempted. Anaerobic digestion with Inoculum from easily obtainable cattle excrement produced a 2:1 mixture of CO2 and biohydrogen with H2/glycerol (in crude glycerol) molar ratio of 0.18. The biohydrogen gas mixture obtained from a single batch fermentation was fed to a fuel cell without further purifications (except adsorptive removal of H2S and NH3). It could generate electric power at 46.5 J/g glycerol (in crude glycerol). With the highest hydrogen production ratio observed in the present work, the current methodology would convert 1 g of crude glycerol into 117 J of electricity.

In future work, it would be interesting to test with crude glycerol produced in a different region with different feed since that might have affected the composition of crude glycerol and consequently their hydrogen fermentation characteristics. In that case, sensitivity analysis of the effect of each crude glycerol component may be necessary. It would also be interesting to make the process a continuous flow and be able to run power generation continuously.

ACKNOWLEDGEMENTS

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