Hydrogen Production from Palm Oil Mill Effluent by Fermentation

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Abstract

Hydrogen production by fermentation was examined by using palm oil mill effluent. *Clostridium butyricum* produced more than 2.2 NL of hydrogen from 1 L of raw POME at pH 5.0, and *Enterobacter aerogenes* produced ca. 1.9 NL at pH 6.0. While from the culture liquid added 1% of peptone on the raw POME, *C. butyricum* produced more than 3.3 NL and also *E. aerogenes* 3.4 NL at pH 6.0 and 5.0, respectively. In this manner, the addition of nitrogen source to the POME liquid exerted an influence on the volume of hydrogen production.

Since *Aspergillus niger* has ability to produce cellulase, co-cultivation of *C.butyricum* with *A. niger* was tried to utilize celluloses in the POME. Against our expectations, however, the results were lower productivities than pure cultivation's.

We analyzed the components of POME by liquid chromatography and capillary electrophoresis before and after cultivation. The main substrate for hydrogen production was found to be glycerol.

1. Introduction

In Malaysia, approximately 50% of agricultural land is the Oil Palm farm. The yield of Palm Oil in 1997 was 9.07milion tonns. The production is now the chief industries of the nation. Since palm oil mills also produce approximately 3.7 tonns of effluent to the 1 ton oil production, the suspended organic solid and dissolved organic matters have brought serious destruction of water resource. We are therefore studying digestive systems of the organic matters in the palm oil mill effluent (POME) to biogas energy especially to H_2 and CO_2.

The POME we obtained had a pH value of approximately 4.0 and contained 3.4 wt-% of nonvolatile organic matters. It is not clear that how much soluble component is contained, but nearly 30ml of H_2 was evolved from 100ml of POME when POME was fermented by micro flora from a POME digestion pond. Therefore, we can simply expect H_2 gas more than 11million Nm^3 from POME in Malaysia. Analysis of soluble components of POME by liquid chromatograph showed declines in peeks likely to be citrate and ascorbate.

2. Experimental method

Batch cultivation was executed by using apparatuses shown in Fig.1 to know the optimum conditions such as pH and nutrients of culture, co-cultivation with microorganisms which produce enzymes for hydrogen production from POME. Since it was known that both *Enterobacter aerogenes* and *Clostridium butyricum* can produce hydrogen considerable activity from glucose,

these two bacteria were used for hydrogen production from POME.

POME was obtained from Havys Oil Mill Sdn. Bhd. Malaysia and Malaysia Palm Oil Board (MPOB) Experimental Palm Oil Mill. Components of the POME was analyzed by liquid chromatography and capillary electrophoresis before and after cultivation. For the LC analysis, we used a packed column GL-C610H which was made by Hitach Chemical Co, Ltd.

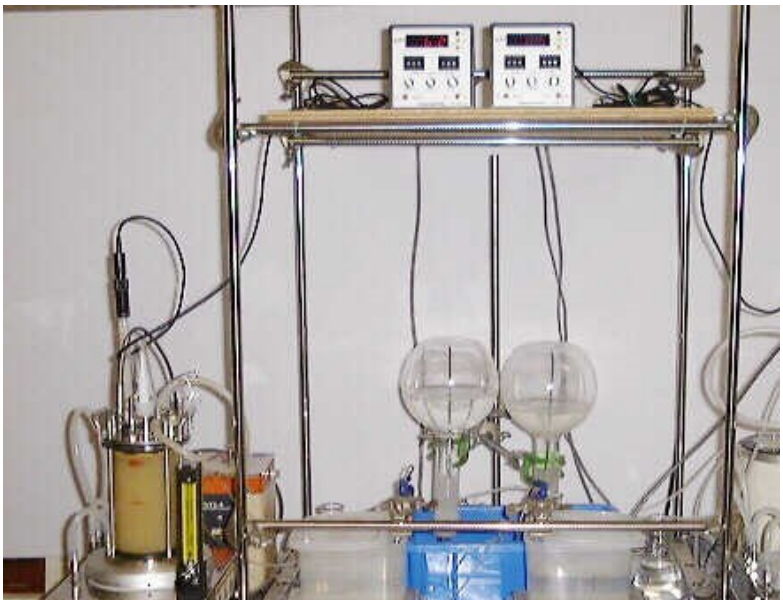

Fig. 1 Photograph of experimental apparatus

The amount of non-volatile component was measured by drying 30ml of POME under 105°C for enough hours.

Fermentation experiments were performed without or with 1% peptone as nitrogen source while stirred under a temperature of 38°C. Culture pH was kept constant by an automatic controller during cultivation.

3. Results and discussions

3.1. pH of POME and contents of non-volatile substance

pH of POME varied in each lot of samples. The pH changed form 3.8 to 4.7 as shown in Fig.2. Since the cultural pH for both *E. aerogenes* and *C. butyricum* is preferable from 5.0 to 7.0, it was realized that the pH of POME should be adjusted to the suitable pH for the bacteria before cultivation.

The contents of non-volatile substances in POME were also changed from 2.32 to 5.61 % in weight. The average was 4.14 %. These facts mean that the volume of hydrogen from POME changes whenever lot is changed.

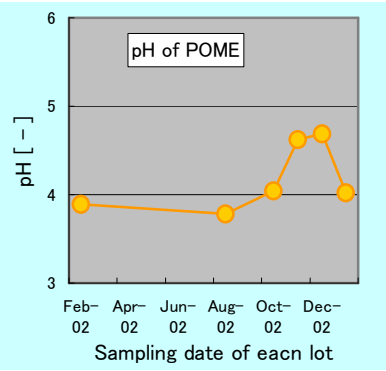

Fig.2 pH variation of POME

Table 1. Contents of non-volatile substance

lot	non-volatile solid [%-wt]
Feb-02	2.32
Aug-02	5.61
Nov-02	3.93
Jan-03	4.68
average	4.14

3.2. Effect of pH of culture liquid on hydrogen evolution

The effect of pH of culture liquid on hydrogen evolution were checked by changing the cultural pH of the culture while the culture pH were kept constant by automatic pH controller at batch cultivation. As seen in Fig.3, the optimum pHs for *E. aerogenes* and *C. butyricum* were 5.0 and 6.0, respectively. These results were very different from the results obtained from the experiments of synthetic culture using glucose and sucrose for the substrate. This might depend on the property of lot of POME, we should therefore continue the check experiment. However both bacterium produced hydrogen ca. 3.5 NL from 1 liter POME, it was clarified that we can get hydrogen gas at least 3.5 NL from the substrates in 1 liter raw POME.

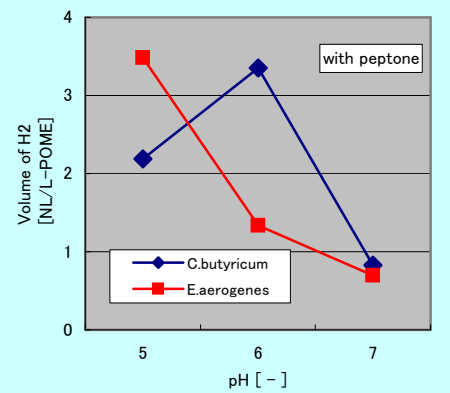

Fig.3 Effect of culture pH

3.3. Effect of nitrogen source addition to POME

We conjectured that POME might not contain enough amount of nitrogen source for the bacterial growth. Depending on this conjecture, we checked the effect of nitrogen source by adding peptone on raw POME. The results were shown in Figs. 4 and 5. In case of *C. butyricum*, the effect clearly appeared as shown in Fig. 4. The volume of hydrogen produced from 1 liter POME was ca. 1.5 NL at pH 6.0 when peptone was not added, while ca. 3.4NL when peptone was added. However, it was too short number of data to conclude the effect of peptone addition, we should check the nitrogen source effect also in the coming year.

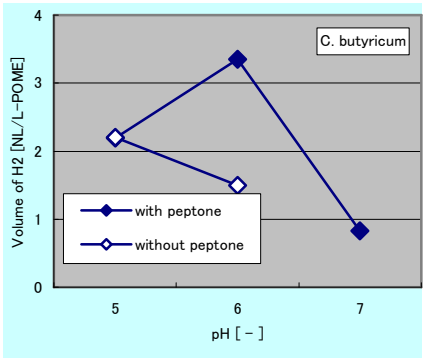

Fig.4 Effect of nitrogen source (in case of *C. butyricum*)

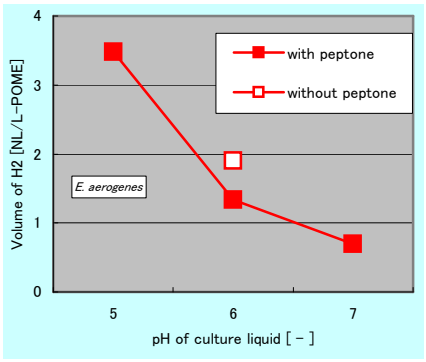

Fig.5 Effect of nitrogen source (in case of *E. aerogenes*)

3.4. Effect of co-cultivation with *Aspergillus niger*

To utilize cellulose in POME as the substrate for hydrogen production, we tried co-cultivation with *Aspergillus niger* and *C. butyricum* or *E. aerogenes*. The results were shown in Figs 6 and 7. Contrary to our expectation, in both case of *C. butyricum* and *E. aerogenes*, the results were that the co-cultivation did not bring good effect on hydrogen evolution. The reason has not yet been cleared, so we have to clarify in the coming fiscal year.

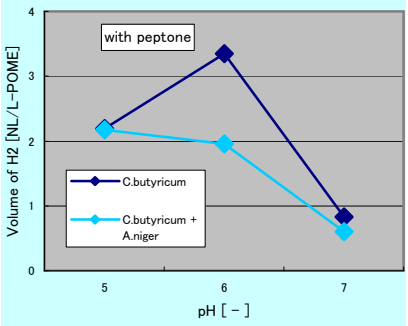

Fig.6 Effect of co-cultivation (*C.butvricum* and *A. niger*)

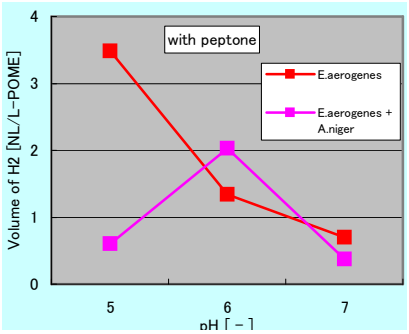

Fig.7 Effect of co-cultivation (*E.aerogenes* and *A. niger*)

3. Analysis of components of POME

We analyzed the components of POME by liquid chromatograph and capillary electrophoresis. We found several known peeks and also many unknown peeks. Main component of the POME was not good substrate for *C. butyricum* and *E. aerogens*, but microflora obtained from a POME digestion pond utilized many unknown organic matters as seen in Figs.8 and 9.

We have to determine these unknown organic matters in the coming year.

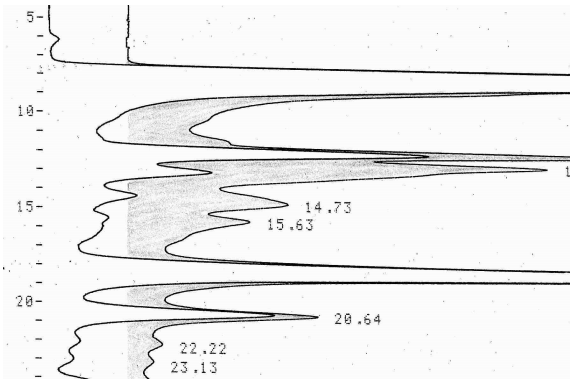

Fig.8 Results of LC analysis of POME before and after cultivation with microflora.

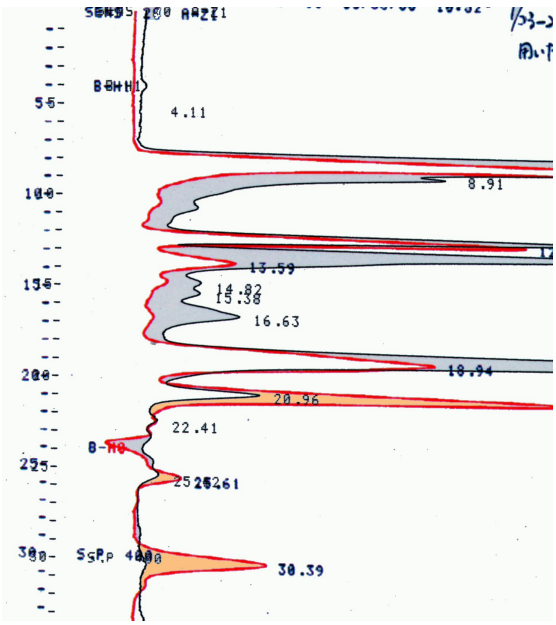

Fig.9 LC analysis of POME before and after cultivation with C. butvricum