Operation optimization of the lipase-catalyzed biodiesel production

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[Abstract]

Biodiesel has significantly few emissions because it is made from biological resources. Lipase catalyzed transesterification of olive oil with ethanol to produce biodiesel was studied. The conversion was low because Lipase OF had low resistance for ethanol. To prevent deactivation, hydrolysis – esterification two stage system was employed. Ethanol was added in three ways, adding at once, dividing ethanol into 3 and using pump. Introducing ethanol by a pump had the highest conversion. Also diluted ethanol improved the conversion. Fermentation raw ethanol can produce bio-diesel almost same amount as diluted regent

[Introduction]

Biodiesel is made from renewable biological sources such as vegetable oils or animal fats. Biodiesel is biodegradable and non-toxic, and has significantly fewer emissions than petroleum-based diesel when burned. Pure or blended biodiesel can be used as diesel fuel. Bio diesel is made from transesterification of biological sources with alcohols such as methanol or ethanol. Acid or base was major catalyst for the transesterification[1,2]. But product should be washed or neutralized after reaction. Recently, in the view of the environmental safety, lipase was employed as catalyst by some researchers[3-5].

Because of the high cost of enzyme, researchers used many kinds of lipase and oils to identify the optimal conditions. For example, lipase from *Rhizomucor miehei* achieved conversion of 92.2 % with soybean oil and methanol[3]. Lipase from *Mucor miehei* achieved conversion of 98 % with tallow and ethanol[4]. Lipase from *Pseudomonas fluorescens* had higher activity than *Mucor javanicus*, *Candida rugosa and Rhizopus niveus* when triolein and 1-propanol were used. It achieved conversion of 100 % however the other lipase conversion was lower than 3 %[5]. Fukuda *et al.* reviewed lipase catalyzed biodiesel production[6]. Reaction temperature, kinds of alcohol, amount of water, reactant ratio, etc. were varied in the many researches.

Also ethanol can be made by fermentation of biomass. Combination of vegetable oil and fermentation ethanol does not increase carbon dioxide in the air when it is used as

diesel fuel. In this presentation olive oil and ethanol were used as reactant. Effect of ethanol addition way was mainly discussed. Finally, raw bio-alcohol was examined.

[Experiments]

A jacketed stirred tank reactor was used. Reaction temperature was 313 K in all experiment. Lipase OF (from *Candida cylindracea* (*C.rugosa*)) and Lipase QLC (from *Alcaligenes sp*.) were used as catalyst and were supported by Meito Sangyo Co., Ltd. Olive oil, oleic acid and ethanol (99.5 %) were purchased from Wako Pure Chemical Industries LTD. No further purification was carried out.

<Transesterification of olive oil>

4.0×10^{-5} m^3 of olive oil and 7.2×10^{-6} m^3 of ethanol were set in the reactor. Mol ratio of ethanol to olive oil was stoichiometric, 3. 300 unit/g-oil or 1100 unit/g-oil of lipase was used.

<Hydrolysis – esterification of olive oil>

At first 3.3×10^{-5} m^3 of olive oil, 6.0×10^{-6} m^3 of water and 300 unit/g-oil or 1100 unit/g-oil of Lipase OF was set in the reactor. When hydrolysis was finished, 6.0×10^{-6} m^3 of ethanol was introduced into the reactor. Three different methods were tested to feed ethanol. (1) Add the ethanol at once. (2) Add the 1/3 amount of ethanol by 3 times. (3) Add the ethanol continuously by a pump. The total amount of the ethanol was same in each operation.

<Esterification of oleic acid>

1.8×10^{-5} m^3 of ethanol was diluted with water to 89, 69, 53, 36 and 10 wt%. 8.1×10^{-5} m^3 of oleic acid and diluted ethanol were set in the reactor. Mol ratio of ethanol to oleic acid was stoichiometric, 1. 1150 unit/g-oil of Lipase OF was used. Japanese sake (Tengumai, Syata Syuzou Co. Ltd) and red wine (Stamp series red, Hardy Wine Company) were also used as bio-alcohol.

[Results and Discussion]

Transesterification was carried out. Figure 1 shows the experimental results. The conversion was 50 % at 30 h., when Lipase QLC was used. Lipase OF had lower activity than Lipase QLC. The conversion was 5 % at 30 h. Figure 2 shows the effect of lipase concentration. Lipase was increased from 300 unit/g-oil to 1100 unit/g-oil. The conversion increased to 80 % when Lipase QLC was used. But Lipase OF still shows low conversion. Generally enzyme was deactivated with ethanol. Lipase QLC had higher resistance for

ethanol than Lipase OF. So it can increase the conversion with increase of lipase concentration, but it could not attained 100 % even in high catalyst concentration. Lipase OF was suddenly deactivated by ethanol. Deactivation by ethanol was great problem in this reaction system.

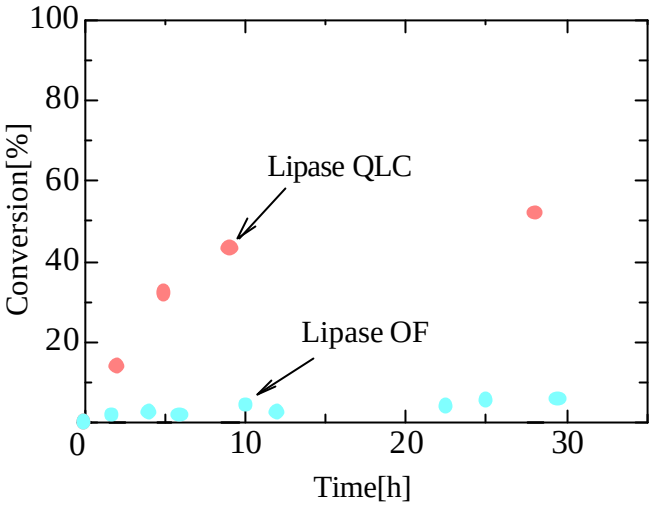

Fig.1 Transesterification

Lipase : 300 unit/g-oil

Catalyst cost is one of the important problem in the industry. Lipase QLC had a higher activity for transesterification than Lipase OF, but it is 40 times expensive (unit base). Lipase OF was used in the following experience. How to avoid deactivation of Lipase OF was mainly discussed.

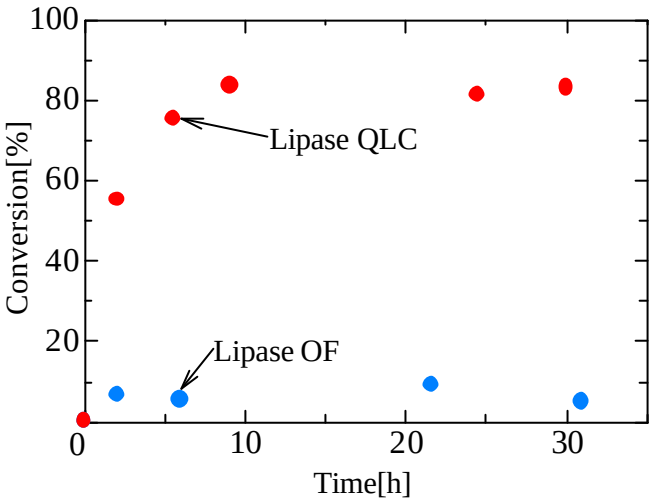

Fig.2 Transesterification

Lipase : 1100 unit/g-oil

Preliminary experimental results showed that Lipase OF has high activity in hydrolysis of fatty oil. Hydrolysis - esterification two stage system was examined instead of direct transesterification. Figure 3 shows the results of two stage system. Ethanol was added in the reactor at reaction time 0. Conversion was 80 % with 1100 unit/g-oil. It was large increase from transesterification and was same as the conversion of transesterification with Lipase QLC. Also the conversion of 300 unit/g-oil was increased to 30 %. Large excess of water was set in the reactor for hydrolysis, so water was remained during esterification. Lipase deactivation was prevented because ethanol was diluted with water.

To prevent the deactivation effect of ethanol, 1/3 amount of ethanol was added by 3 times. Initial ethanol concentration in the reactor was decreased in this way. As shown in Figure 4, conversion became 30 % at 7 h. Ethanol concentration was estimated to be 0

when conversion became 33%. Then next ethanol was introduced into the reactor at 11 h. But conversion was increased a little. Final ethanol was introduced into the reactor at 25 h. Final conversion was 40 %. Conversion was improved with changing addition way. Decrease of ethanol concentration in the reactor was effective in this reaction.

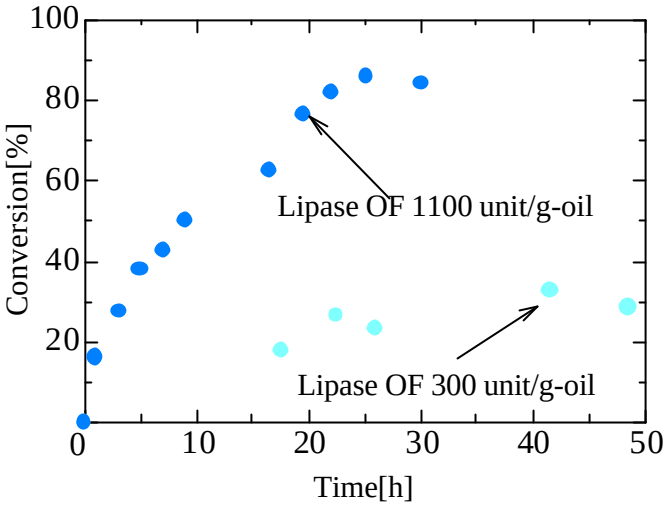

Fig.3 Hydrolysis - esterification system

Adding at once

Other addition way was tested. Ethanol was continuously added by a pump. Ethanol concentration in the reactor became lower than adding by 3 times. Figure 5 shows the results. Conversions became higher by using the pump. They were about 60 %. Lower ethanol flow rate gave a little higher conversion.

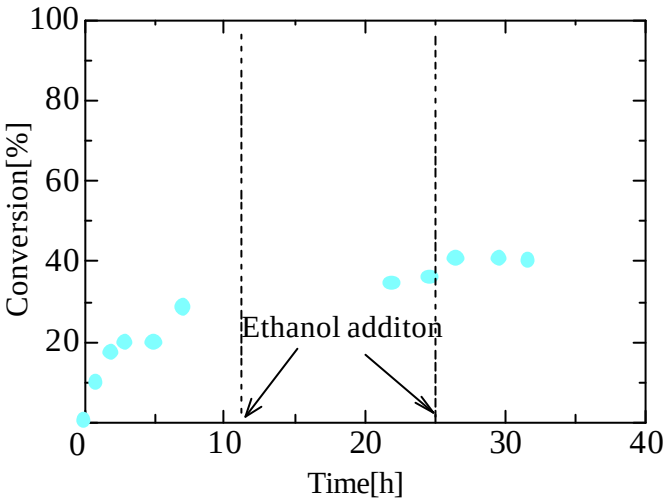

Fig.4 Hydrolysis - esterification system

Adding 1/3 amount by 3 times

Lipase OF 300 unit/g-oil

Effect of ethanol concentration was studied with diluted ethanol. Oleic acid was used as a reactant instead of hydrolysis of olive oil. Figure 6 shows the results. Ethanol concentration was estimated to be about 53 wt% after hydrolysis in fig.3. Comparing ethanol concentration of 53 wt% in Fig.6 with Fig .3, the conversion was largely decreased when oleic acid was used. The reason was not clear. Olive oil and oleic acid were not pure. There purity was around 60 %. Some impurities in the reactant may affect the reaction. Conversion was increased with decrease of ethanol concentration. But no significant improvement was observed under the concentration of 36 wt%. The highest conversion was 45 %.

Dilution of ethanol was effective for this reaction. Distilled bio-alcohol should not be

used for the reaction. Japanese sake (ethanol 15.5 %) and red wine (14 %) were used as representatives of raw bio-alcohol. Figure 7 shows the experimental results. The conversion was 40% and 50% with red wine and Japanese sake, respectively. Fermentation raw ethanol can produce bio-diesel almost same amount as diluted regent ethanol. Figure 8 was the reaction mixture after the reaction which Japanese sake was used. Bio-diesel was top phase and it was separated from water-glycerin phase. It was easy to separate. As we can use raw bio-ethanol without distillation, the energy for the distillation process can be saved.

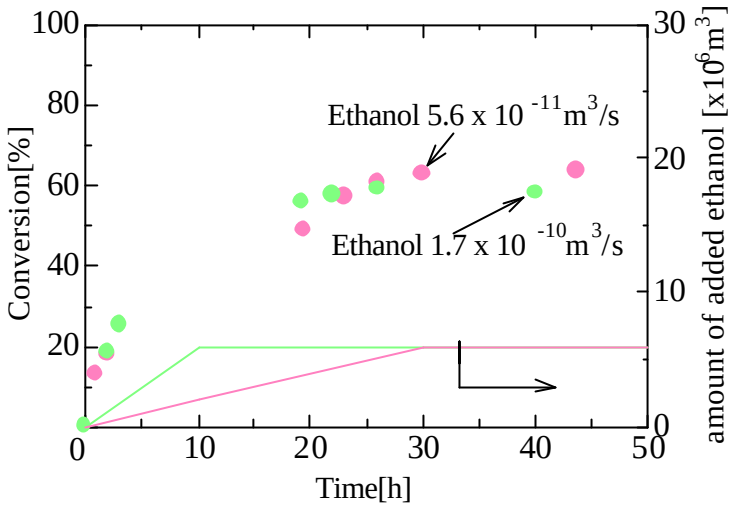

Fig.5 Hydrolysis - esterification system

Adding by pump

Lipase OF 300 unit/g-oil

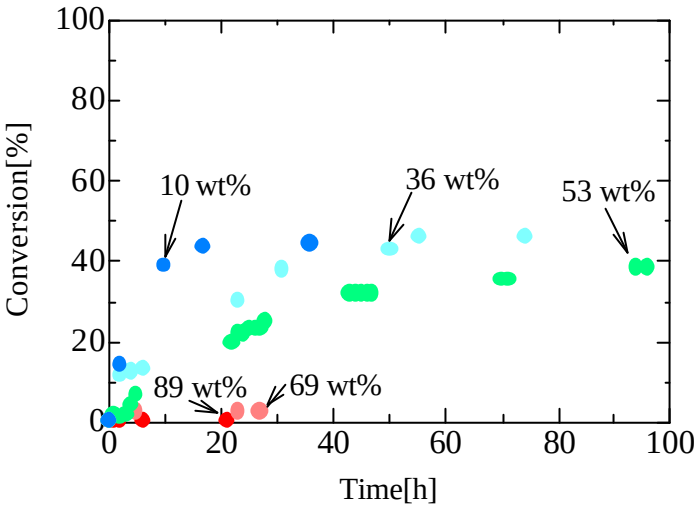

Fig.6 Effect of water

Lipase OF 1150 unit/g-oil

[Conclusion]

Lipase OF has low resistance for ethanol. Transesterification was not good for the Lipase OF. Hydrolysis - esterification method was employed. Make a ethanol concentration in the reactor being low was important for the reaction. Adding ethanol by the pump slowly was a good for the system. Raw bio alcohol was also good for the system. Because it was low concentration ethanol. We can use energy not for distillation of raw bio-alcohol but for production of bio-diesel.

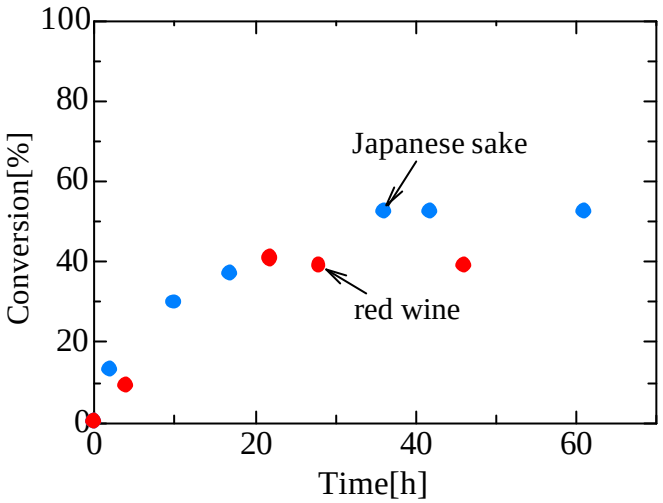

Fig.7 Raw bio-ethanol

Lipase OF 1150 unit/g-oil

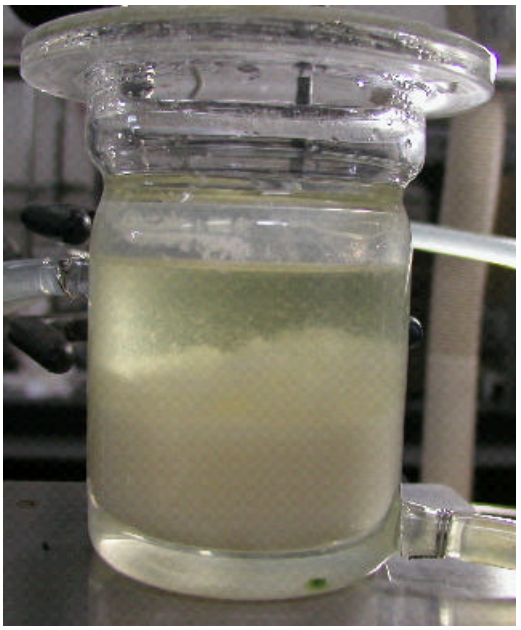

Fig.8 Phase separation

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