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Enzymatic production of biodiesel from waste cooking oil in a packed-bed reactor: An engineering approach to separation of hydrophilic impurities

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HIGHLIGHTS

- An engineering approach to produce biodiesel enzymatically from waste cooking oil.
- The yields of methyl esters and glycerol reached 94.3% and 99.7%, respectively.
- The reactor system was modified to improve the separation of hydrophilic impurities.
- Water generated during esterification of free fatty acids was removed efficiently.
- Long-term operation was possible with less than 0.2% residual triglyceride.

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ABSTRACT

An engineering approach was applied to an efficient biodiesel production from waste cooking oil. In this work, an enzymatic packed-bed reactor (PBR) was integrated with a glycerol-separating system and used successfully for methanolysis, yielding a methyl ester content of 94.3% and glycerol removal of 99.7%. In the glycerol-separating system with enhanced retention time, the effluent contained lesser amounts of glycerol and methanol than those in the unmodified system, suggesting its promising ability to remove hydrophilic impurities from the oil layer. The PBR system was also applied to oils with high acid values, in which fatty acids could be esterified and the large amount of water was extracted using the glycerol-separating system. The long-term operation demonstrated the high lipase stability affording less than 0.2% residual triglyceride in 22 batches. Therefore, the PBR system, which facilitates the separation of hydrophilic impurities, is applicable to the enzymatic biodiesel production from waste cooking oil.

1. Introduction

The enzymatic process using lipase offers considerable advantages in biodiesel fuel production through plant oil transesterification. These advantages, namely, low energy consumption and easy recovery of glycerol byproduct, make the lipase-catalyzed process a promising alternative to the conventional process using a homogeneous alkaline catalyst, which suffers from complicated downstream processes (Fukuda et al., 2001; Ranganathan et al., 2008). Moreover, lipase can convert fatty acids into their corresponding methyl esters owing to the esterification activity, whereas soaps are formed when an alkaline catalyst is used for oils containing free fatty acids of more than 0.5% (Talukder et al., 2010).

The cost of raw materials currently constitutes a large percentage of the total production cost of biodiesel (Robles-Medina et al., 2009), which means that the use of refined edible oil is not economically feasible. Waste cooking oil is thus a better alternative as a raw material for biodiesel production (Zhang et al., 2003a, b). Compared with refined oils, waste cooking oils have different chemical and physical properties; for example, oils undergo degradation by hydrolytic reactions during frying, resulting in increased acidity (Felizardo et al., 2006). Such chemical and physical changes in oils require process considerations during biodiesel production. Nonetheless, the huge abundance and low cost of waste cooking oil are attractive for ensuring the economic viability of the process as well as for preventing environmental pollution. Therefore, an investigation of low-cost feedstocks including waste oils has recently been of considerable interest in related research fields.

Using waste cooking oil for enzymatic biodiesel production was investigated previously (Charpe and Rathod 2011; Chen et al., 2009; Dizge et al., 2009; Halim et al., 2009; Jian-Xun et al., 2007; Watanabe et al., 2001; Yagiz et al., 2007), where free fatty acids present in acidified oils could be esterified by lipase catalysis.

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Despite numerous studies focusing on various reaction parameters, published information on reactor configurations is relatively scarce. To minimize the technological limitations for biodiesel production by the enzymatic method, further studies on bioreactor design and scale up of the process are necessary (Bajaj et al., 2010; Fjerbaek et al., 2009).

PBR, one of the most commonly used reactor types in biotechnology, is applicable to continuous production using heterogeneous catalysts (Wang et al., 2011). However, the main disadvantage is that the resulting glycerol remains at the bottom of the reactor and adsorbs on the surface of the immobilized lipase, thus decreasing the catalytic efficiency (Gog et al., 2012). A previous study by Hama et al. (2011a) described a basic model of an enzymatic PBR, in which the effluent was continuously collected and recycled into the same column with the stepwise addition of methanol. The glycerol byproduct can be separated quantitatively in the glycerol-separating tank connected to the column as described in a previous paper using refined vegetable oil (Hama et al., 2011a). In this study, to adjust the PBR system to process waste cooking oil, the glycerol-separating tank was modified. An engineering approach using the modified system improved the removal of hydrophilic impurities from the methyl ester layer. The effect of increasing the acidity of oil and the long-term operation of the PBR system were also presented.

2. Methods

2.1. Materials

Novozym® 435 (Novozymes, Bagsvaerd, Denmark), commercial *Candida antarctica* lipase B immobilized on a macroporous acrylic resin (Lewatit VP OC 1600; Lanxess, Leverkusen, Germany), was used as an immobilized lipase. Waste cooking oil, obtained from Hamada Kagaku Corporation (Amagasaki, Japan), contained 780 ppm water (determined by Karl Fischer titration) and its acid value was 1.9 (mg-KOH/g). Methanol was purchased from Wako Pure Chemical Industries Ltd. (Osaka, Japan).

2.2. Reactor setup

A schematic of the reactor setup used in the current study is described in a previous paper (Hama et al., 2011a). Briefly, a stainless steel pipe (length, 1.5 m; inner diameter, 15.7 mm; and volume, 290.2 ml) was packed with immobilized lipase at a volumetric packing ratio of 60%, maintained at 30 °C by circulating water, and used as a reaction column. By using a diaphragm metering pump, the reaction mixture was supplied to the upper portion of the column and overflowed through a glycerol-separating tank placed under the reaction column. As illustrated in Fig. 1, the reaction mixture was retained in a given space in the glycerol-separating tank. During the retention period, the glycerol droplets accumulated at the bottom of the separation tank because of their high density and hydrophilicity (Type A). In the modified system (Type B), the outlet of the column was lengthened to submerge it in the glycerol layer. Thus, the reaction mixture was first allowed to contact with the glycerol layer during the retention period, and then overflowed as in Type A. In both types, the outflowing liquid consisting mainly of fatty acid methyl esters and residual glycerides was collected as the effluent, into which methanol was further added until the completion of the reaction.

2.3. Enzymatic reaction in PBR

For methanolysis using one reaction column, 1000 g of waste cooking oil was mixed with 0.5 mol equivalent of methanol. The reaction mixture was supplied to the reaction column at a flow rate of 600 ml/h, and the effluent was collected until the reaction mixture tank became empty. After each pass, a small amount of the effluent without glycerol was collected for analysis, and the removed glycerol was collected by opening the valve in the glycerol-separating tank (Fig. 1). The effluent was mixed with 0.5 mol equivalent of methanol, and supplied again to the same reaction column.

To prepare oils with different acid values, waste cooking oil was mixed with oleic acid. The amount of methanol added at each pass was 0.5 M equivalent to a mixture of waste oil and oleic acid. Because only a small amount of glycerol is generated from waste oil with high acid values, 100 g of glycerol was initially added to the glycerol-separating tank.

2.4. Analytical methods

Methyl ester, oleic acid, and triglyceride contents (wt.%) of the PBR effluent were determined using a gas chromatograph connected to a ZB-5HT capillary column (0.25 mm × 15 m; Phenomenex, USA). The contents were calculated as the ratio of each material present in the reaction mixture without purification. Glycerol contents in the methyl ester and crude glycerol layers were also determined using a gas chromatograph connected to a ZB-WAXplus capillary column (0.25 mm × 15 m; Phenomenex, USA). The detailed procedures for gas chromatography are found in a previous paper (Hama et al., 2011a).

The methanol content in the methyl ester was measured by heating at 70 °C until a constant weight was achieved. The water content in the methyl ester was determined by Karl Fischer titration (Hiranuma Sangyo Co., Ltd., Ibaraki, Japan). The acid value of oil was determined according to an official titration method (JIS K 0070) using potassium hydroxide (KOH).

3. Results and discussion

3.1. Methanolysis of waste cooking oil in PBR

Methanolysis of waste cooking oil was carried out using PBR integrated with a glycerol-separating system (Fig. 1, Type A). Fig. 2a shows the methyl ester content of the PBR effluent and total amounts of glycerol collected in the glycerol-separating tank. The methyl ester content in the effluent increased with increasing pass number, reaching 94.3% in the oils including methyl esters, methanol, residual glycerides, and other unknown compounds (10th pass). As methanolysis proceeds, the sum of removed glycerol also increased more significantly between the 5th and 7th passes. The final glycerol amount of 103.7 g corresponds to 99.7% of the theoretical value, indicating the successful removal of glycerol from the column.

Compared with previous studies using refined vegetable oil (Hama et al., 2011a,b), methanolysis of waste cooking oil required an excess amount of methanol to reach a high conversion. Namely, the total amount of methanol added to waste cooking oil in this study was 5 M equivalents, whereas 4 M equivalents of methanol were sufficient for refined vegetable oil. This is probably because the reaction rate of methanolysis is relatively higher with refined oil than with using waste cooking oil, which is subjected to unfavorable reactions such as thermolytic, oxidative, and hydrolytic reactions during frying (Lam et al., 2010). The location of the unreacted methanol is one of the major issues in terms of its toxicity to lipase. To confirm the contribution of glycerol removal to the prevention of the accumulation of an excess amount of methanol inside the column, the glycerol and methanol contents in crude glycerol were investigated. As shown in Fig. 2b, the glycerol