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Effective Lipid Extraction from Fat Balls Using Liquefied Dimethyl Ether: Process Optimization with a Box−Behnken Design

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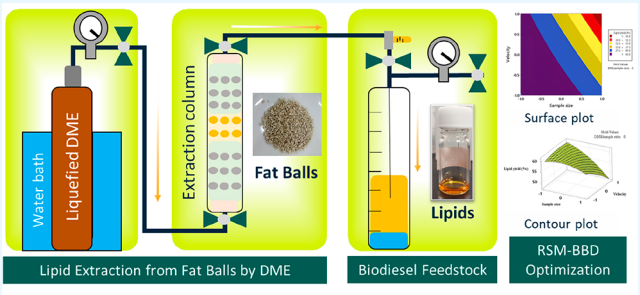
ABSTRACT: In recent years, lipids reused from urban wastewater materials have come to prominence as possible raw materials for biodiesel production. The present work investigated liquefied dimethyl ether (DME) for the lipid extraction of fat balls from sewage pumping stations. A response surface methodology (RSM) based on a Box−Behnken design (BBD) was utilized to optimize DME extraction parameters (sample size, velocity of liquefied DME, and DME/sample ratio). The maximum lipid yield was 65.2% under optimal DME extraction conditions (sample size 1 mm, velocity of liquefied DME 3.3 m/h, and DME/sample ratio 80 mL/g). Under the optimum conditions, the DME technique exhibited higher lipid recovery than that of mechanical shaking extraction (49.0%) or Soxhlet extraction (62.0%). The extracted lipids were converted into biodiesel, resulting in an approximately 35.2−46.2% biodiesel yield. Furthermore, the fatty acid methyl ester composition of the extracted lipids was characterized. These significant findings highlight the promising potential of fat balls as sustainable biodiesel feedstocks and provide valuable insight that will aid the development of better technology for lipid extraction.



1. INTRODUCTION

Global concerns over the depletion of fossil fuels and the environmental pollution problem have led to the development of carbon-neutral energy sources.[1] Biodiesel, as one of the most promising biofuels, has attracted attention due to lower emissions, renewability, and biodegradability.[2,3] Biodiesel consists of fatty acid methyl esters (FAME) that can be produced from the transesterification reaction of vegetable oil or animal fat.[4] The current biodiesel feedstock relies mainly on edible vegetable oil sources, which account for 70−80% of the total production cost, thus limiting the growth of biodiesel commercialization.[5−7] Therefore, there is an urgent need for cost-effective and sustainable biodiesel feedstocks. In this context, the utilization of fat balls from wastewater treatment plants has emerged as a viable alternative for biodiesel production, promoting the valorization of waste sources.

The term "fat ball" refers to deposits of fat, oil, and grease (FOG) that accumulate on the water surface during the sewage treatment process. This waste is considered to be a nuisance and is disposed of by incinerating or landfilling.[8] Due to their high lipid content and availability, fat balls have the potential to serve as an alternative lipid feedstock. The raw fat balls consist primarily of FOG, along with water, solids, and other impurities. Researchers have examined FOG waste from different locations in sewage treatment systems and discovered that fat balls from the pumping station had a moisture content of 44% and a significant proportion of extractable oils (181 mg/g).[9] In another study, fat balls were obtained from a pumping station and inlet of a sewage treatment plant in London, U.K., with a 93−94% (dry base) lipid content.[10]

The various conventional methods used for lipid extraction from wastewater residual include Soxhlet extraction, the Bligh−Dyer method, and liquid−liquid extraction.[11] These methods employ various organic solvents such as hexane, chloroform, isopropyl alcohol, dichloromethane, and methanol.[12−14] However, these processes are affected by solvent characteristics and are less effective for raw materials with a high moisture content; in addition, the postextraction process consumes high amounts of energy, which is not sustainable.[15,16] Although a pretreatment step can be employed to enhance the extraction performance, it tends to be energy-intensive. Consequently, finding an alternative extraction method would be preferable.

Dimethyl ether (DME) is a synthetic polar gas (at room temperature) that has gained prominence as an eco-friendly and nontoxic extraction agent.[17] It possesses the ability to be liquefied under pressure and vaporizes at standard temperatures.[18] In its liquefied state (0.51−0.59 MPa at room

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Table 1. Characteristics of Fat Balls[a]

			ultimate analysis, wt% DB			
total solids (TS), wt%, DB	higher heating value (HHV), MJ/kg, DB	lower heating value (LHV), MJ/kg, WB	C	H	N	O
55.0	39.2	20.1	68.4	11.4	0.20	12.5

[a]Oxygen content was calculated by the difference. DB: dry basis. WB: wet basis.

temperature), DME has a strong affinity for oil-based substances, with a solubility in water of about 7.8%.[19,20] Consequently, it has proven to be effective in the extraction of neutral and complex lipids from wet feedstock.[21,22] Several studies have reported lipid recovery by DME from various feedstocks, including microalgae, sewage sludge, and biomass.[23−28] It is reported that extraction using liquefied DME can achieve comparable lipid yield and properties close to those of conventional extraction methods.[19,22,26,28] Furthermore, organic matter can be easily recovered after extraction, and DME can be recycled, reducing the energy required. While numerous researchers have performed lipid extraction procedures using DME, the impact of the processing parameters, the possible interactions among parameters, and extraction optimization have not been adequately explored. Processing parameters can affect the properties and enhance extraction efficiency.[15]

In this study, we investigated the performance of lipid extraction from fat balls using the DME technique and optimized the process parameters (sample size, velocity of liquefied DME, and DME/sample ratio) using response surface methodology (RSM) through a Box−Behnken design (BBD). The performance of DME was compared to mechanical shaking and Soxhlet extraction in terms of the lipid and biodiesel yields and FAME profiles. The results of this study have significance for the development of lipid extraction technology and the better use of wastewater residuals for sustainable biodiesel feedstock.

2. MATERIALS AND METHODS

2.1. Sample Preparation. Fat balls were collected from the water surface layer of a pumping station in Kobe City, Japan. The fat balls appeared as floating, yellowish, spherical substances. The samples were stored at 4 °C immediately after collection. Table 1 presents the characteristics of the fat balls.

Before being used in lipid extraction, the fat balls were homogenized and sieved to similar sizes (1 and 3.3 mm). To prepare a larger sample (5.6 mm), fat balls were rubbed by hand into a spherical form and the diameter was checked using a Vernier scale. The prepared fat balls were dried in a fume hood for 24 h at room temperature, and the moisture level was reduced to below 30%.

2.2. DME Extraction Method. Figure 1 shows a schematic diagram of the DME apparatus. The experimental device was a fixed bed extraction system consisting of three main parts: the DME supply tank/vessel 1 (TVS-1-100; volume: 100 cm^3, Taiatsu Techno Corp., Japan), the extraction column/vessel 2 (HPG-10-5; volume:10 cm^3, Φ 11.6 mm × 190 mm, Taiatsu Techno Corp., Japan), and the recovery tank/vessel 3 (HPG-96-3, volume: 96 cm^3, Taiatsu Techno Corp., Japan). A needle valve was installed to control the liquefied DME flow rate.

Fat balls and glass beads were packed into vessel 2. A filter (polytetrafluoroethylene; pore size: 0.8 μm; Advantec Toyo

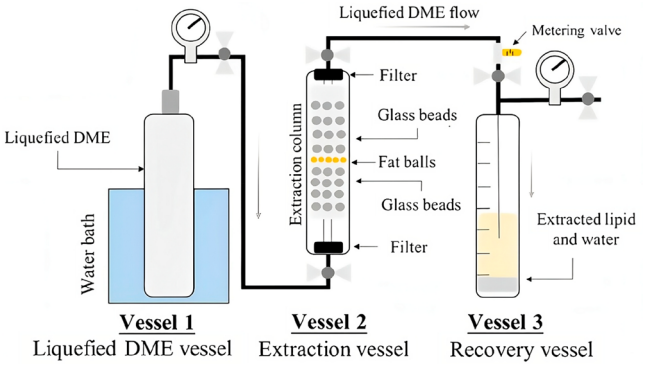

Figure 1. Schematic diagram of the lipid extraction process using liquefied dimethyl ether (DME).

Kaisha Corp., Japan) was placed at the outlet of vessel 2. Liquefied DME was produced by cooling pure gaseous DME (Tamiya Ltd., Japan) to −12 °C with ethanol (Guaranteed Reagent; Wako Pure Chemicals Ltd., Japan). The required amount of Liquefied DME (according to the experimental design) was stored in vessel 1. The volume of DME was determined by measuring the weight of the supply vessel.

Vessel 1 was then placed in a water bath, and the temperature was maintained at 37 °C. Liquefied DME was transferred from vessel 1 to vessel 2 under pressurization at 0.7 MPa at room temperature. The flow rate was adjusted by a needle valve, and the liquefied DME passed through vessel 2 at different velocities. A color change in vessel 2 indicated the presence of a lipid extract. The total consumed liquefied DME was set at around 150 mL per batch.

After the extraction process, the lipids were recovered by reducing the pressure of vessel 3, allowing for complete gasification of the DME. The adhered raw lipid in vessel 3 was flushed with hexane and dried to determine the lipid yield. The lipid extraction yield is calculated by the following equation:

$$\text{lipid yield}(\%) = \frac{W}{W_0} \times 100\% \quad (1)$$

where W is the weight of the lipid extracted (g), and W_0 is the weight of the initial fat balls (g).

The mass balance for DME extraction was calculated by measuring the weight variation of vessels 1−3 before and after the experiments.

2.2.1. Optimization of DME Extraction Using RSM. RSM through the BBD was used for the optimization of lipid extraction of fat balls by DME. Three operational parameters, sample size, velocity, and DME/sample ratio, were chosen to study the independent and interactive effects of the variables on the lipid extraction yield. Table 2 illustrates the factors and levels for lipid extraction by DME.

Minitab software version 21 (Minitab Inc., State College PA, USA) was utilized to conduct the statistical analysis. The experimental outcomes were developed with a second-order

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ACS Omega 2024, 9, 34859−34868

Table 2. Independent Variables: Units and Range of Actual Values

parameter		unit	level		
			−1	0	1
X_1	sample size	mm	1	3.3	5.6
X_2	velocity	m/h	2.8	5.7	8.5
X_3	DME/sample ratio	mL/g	10	45	80

polynomial equation using response surface regression analysis, as given by eq 2:

$$Y = b_0 + \sum_{i=1}^{3} b_i X_i + \sum_{i=1}^{3} b_{ii} X_i^2 + \sum_{i=1}^{2} \sum_{j=i+1}^{3} b_{ij} X_i X_j \quad (2)$$

where Y is the response factor (lipid yield); b_0 is the constant coefficient; X_i is the independent variable; and b_i, b_{ii}, and b_{ij} are the coefficients of linear, quadratic, and interaction terms, respectively.

2.3. Conventional Extraction of Lipids with Hexane. Mechanical shaking extraction and Soxhlet extraction were utilized to analyze the extraction performance of the DME technique. Hexane was selected due to its advantage in extracting organic substances.[13]

2.3.1. Mechanical Shaking Extraction. Liquid−liquid extraction of lipids was carried out using a mechanical shaker (SA300; Yamato Scientific Ltd., Japan) with hexane (Guaranteed Reagent; Wako Pure Chemicals Ltd., Japan).[28,29] The extraction process was conducted for 60 min at ambient temperature (200 rpm) in three consecutive experiments using 5 mL of hexane and 1 g of fat balls. After shaking, the tube was centrifuged at 3000 rpm for 15 min. The supernatant phase was collected, and the solvent was separated from the lipid extract by using a rotary evaporator. The lipid extract was then dried to a constant mass in a vacuum desiccator, and the lipid yield was calculated using eq 1.

2.3.2. Soxhlet Extraction. The extraction of lipids was carried out by using a Soxhlet apparatus. Fat balls were placed into a thimble filter (26 × 30 × 100 mm^3, No. 84; Advantec Toyo Kaisha Ltd.) and extracted using 50 mL of hexane per 1 g of fat balls. The extraction was performed at 80 °C for 8 h. After extraction, hexane was removed by a rotary evaporator. The lipid extract was then dried to a constant mass in a vacuum desiccator, and the lipid yield was calculated according to eq 1.

2.4. Characterization Techniques. The total solid (TS) and volatile solid (VS) contents of fat balls were determined according to the standard method 2540 G.[30] Carbon, hydrogen, and nitrogen (C/H/N) analysis was performed using an elemental analyzer (JM10; J-Science Lab Ltd., Japan). The calorific value was determined using a bomb calorimeter (CA-4J; Shimadzu Ltd., Japan). The lower heating value of the dry solids was calculated from the measured higher heating values.[31] The functional group compositions were measured via Fourier transform-infrared spectroscopy (FTIR; IRSpirit-T; Shimadzu Ltd.) in attenuated total reflectance mode. The detection range was 500−4000 cm^{-1}.

For fatty acid analysis, the extracted lipids were converted into FAMEs using a fatty acid methylation kit (06482-04; Nacalai Tesque Ltd., Japan); the mixtures were heated in a constant temperature bath (EYELA Ltd., Tokyo, Japan) at 37 °C. The analysis of methyl ester composition was carried out using a gas chromatography−mass spectrometry (GC-MS; QP2010 Plus; Shimadzu Ltd.) system equipped with a SP-2560 capillary column (100 m × 0.20 μm × 0.25 μm; Supelco/Sigma Aldrich, U.S.A.). The initial GC oven temperature of 100 °C was held for 5 min, increased to 180 °C at a rate of 4 °C/min and 240 °C at a rate of 2 °C/min, and then held at 240 °C for 15 min. The calibration curve was created using the fatty acid standard (FAME Mix 37 Components; Sigma-Aldrich). Quantitative analysis was conducted to measure the quantity of transesterifiable substances. The biodiesel yield (dry basis) was calculated based on the mass of lipids extracted and their transesterifiable material content.

Table 3. Box−Behnken Design: Coded and Actual Levels of the Main and Response Variables in the Model of Lipid Extraction from Fat Balls Using the DME Method[a]

	coded			uncoded			lipid yield, %	
run order	X_1	X_2	X_3	X_1	X_2	X_3	exptl	predicted
1	−1	−1	0	1	2.8	45	61.0	59.9
10	1	−1	0	5.6	2.8	45	59.0	58.3
7	−1	1	0	1	8.5	45	60.0	60.8
11	1	1	0	5.6	8.5	45	46.6	47.7
2	−1	0	−1	1	5.7	10	55.0	55.2
8	1	0	−1	5.6	5.7	10	53.0	52.8
4	−1	0	1	1	5.7	80	65.0	65.2
13	1	0	1	5.6	5.7	80	53.0	52.8
9	0	−1	−1	3.3	2.8	10	55.0	55.9
6	0	1	−1	3.3	8.5	10	55.0	54.1
5	0	−1	1	3.3	2.8	80	63.0	63.9
3	0	1	1	3.3	8.5	80	57.0	56.1
12	0	0	0	3.3	5.7	45	59.0	59.3
14	0	0	0	3.3	5.7	45	59.0	59.3
15	0	0	0	3.3	5.7	45	60.0	59.3

[a]Note: X_1 = sample size (mm), X_2 = velocity (m/h), X_3 = DME/sample ratio (mL/g).

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ACS Omega 2024, 9, 34859−34868

Table 4. Analysis of Variance Results for the Lipid Yield Using the DME Method[a]

source	DF	adjusted SS	adjusted MS	*F*-value	*p*-value
model	9	289.088	32.121	20.47	0.002
linear	3	205.578	68.526	43.67	0.001
X_1	1	108.339	108.339	69.05	0
X_2	1	47.239	47.239	30.11	0.003
X_3	1	50	50	31.87	0.002
square	3	16.791	5.597	3.57	0.102
$X_1 \times X_1$	1	12.591	12.591	8.03	0.037
$X_2 \times X_2$	1	2.647	2.647	1.69	0.251
$X_3 \times X_3$	1	3.595	3.595	2.29	0.191
two-way interaction	3	66.718	22.239	14.17	0.007
$X_1 \times X_2$	1	32.718	32.718	20.85	0.006
$X_1 \times X_3$	1	25	25	15.93	0.01
$X_2 \times X_3$	1	9	9	5.74	0.062
error	5	7.845	1.569		
lack-of-fit	3	7.178	2.393	7.18	0.125
pure error	2	0.667	0.333		
total	14	296.933			
S	1.2526	R^2	0.9736	R^2(adj)	0.9260

[a]Note: X_1 = sample size (mm), X_2 = velocity (m/h), and X_3 = DME/sample ratio (mL/g). DF: degrees of freedom, SS: sums of squares, MS: mean squares.

3. RESULTS AND DISCUSSION

3.1. Development of the Regression Model Equation for Lipid Extraction by DME. *3.1.1. BBD Experiments.* Lipid extraction using DME, consisting of 15 experimental runs, was performed using a BBD with three levels of three factors, as shown in Table 3. The lipid yield (response variable) is expressed in terms of the percent of dry base. The results show that the lipid yield varied from 46.6% to 65.0%.

The coefficient of the full model was evaluated by using second-order polynomial regression analysis. The regression equation for lipid yield (%) was as follows:

$$Y = 59.333 - 3.680X_1 - 2.430X_2 + 2.500X_3 - 1.847X_1 \times X_1 - 0.847X_2 \times X_2 - 0.987X_3 \times X_3 - 2.860X_1 \times X_2 - 2.500X_1 \times X_3 - 1.500X_2 \times X_3 \quad (3)$$

where X_1 = sample size (mm), X_2 = velocity (m/h), and X_3 = DME/sample ratio (mL/g).

According to Table S1 (Supporting Information), the linear effect of the three factors for lipid extraction, size (X_1), velocity (X_2), and DME/sample ratio (X_3), is important for lipid extraction via the DME method. The coefficients of X_1 (−3.68) and X_2 (−2.43) have negative signs (an antagonistic effect), while X_3 (2.5) has a positive sign (a synergistic effect). This means that using lower levels of X_1 and X_2 and a higher level of X_3 is necessary to obtain a high lipid yield. The interactions of $X_1 \times X_3$ (−2.5) and $X_1 \times X_2$ (−2.86) were found to be statistically significant factors in the model. Moreover, the quadratic parameters of X_1 (−1.84) were significant.

Figure S1 (Supporting Information) presents the perturbation plot of the operational variables against the lipid yield. The perturbation plot compares the effects of all the factors at a specific point within the range of design variables. Notably, X_2 and X_3 show equal effects on the lipid yield, whereas X_1 shows more curvature as the most sensitive factor regarding lipid yield.

3.1.2. Analysis of Variance for Lipid Extraction Yield. The experimental results were evaluated using analysis of variance (ANOVA) to determine the fitness and significance of the model, as shown in Table 4. The regression coefficient is significant when the probability of error (*p*-value) is < 0.05.[32] The ANOVA showed an *F*-value of 20.47 with a *p*-value < 0.005, indicating that the model was significant. The fit of the model was assessed using the coefficient of determination (R^2): the *R*-value and R^2 value (adjusted) were 0.973 and 0.926, respectively. This indicates that >97.3% of the variability in the response could be predicted by the model. The R^2 (adjusted) value indicates that the model accounted for 92.6% of the variance due to the addition of ineffectual predictor variables. A regression model is considered to show a good fit if the regression coefficient (R^2) is >80%.[33] The lack of fit test yielded an insignificant *p*-value; this suggests that the model adequately fits the data.[34]

The predicted and experimental plots of lipid yield are presented in Figure S2 (Supporting Information). The actual and predicted data showed no significant discrepancies, indicating that the model fits the observed data well, generating a good estimate of the response within the range studied.

3.2. Interactions between Process Variables. To illustrate the main and interactive effects of the process variables on the lipid yield, a two-dimensional contour plot and three-dimensional response surface plots were generated based on the developed model.

3.2.1. Interaction of Sample Size and Velocity. The sample size was varied (1, 3.3, and 5.6 mm) to study the influence of the fat ball size on lipid extraction. Figure 2 presents contour and surface plots of the combined effects of sample size and velocity on the lipid yield, while the DME/sample ratio was fixed at the center level (45 mL/g). A reduction in fat ball size and velocity of DME had a positive influence on lipid yield. However, a larger fat ball size and higher DME velocity contributed to the lower yield of lipids due to the limiting effect of mass transfer, which prevents contact between DME and the lipids.[35,36] It has been reported that the lipid yield increases almost linearly with a

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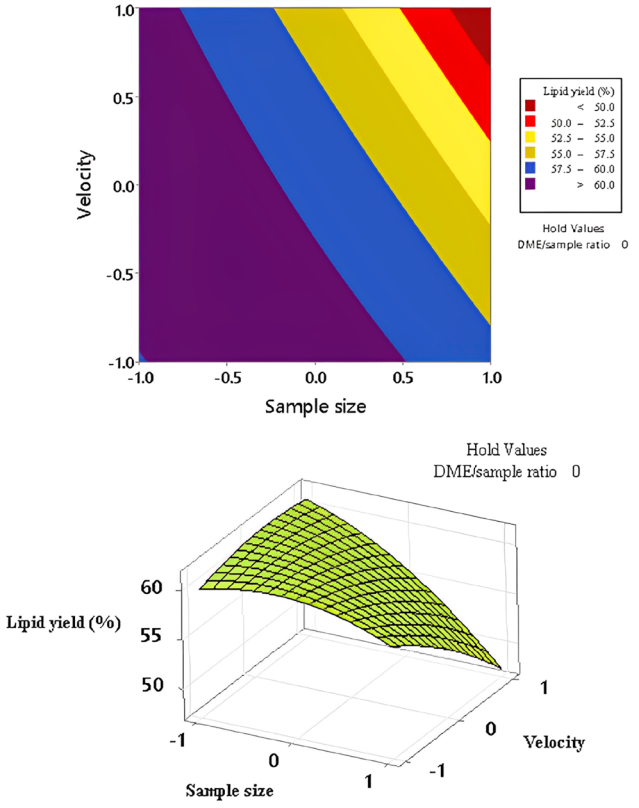

Figure 2. Contour plot and surface plot for the interaction effect of sample size and velocity.

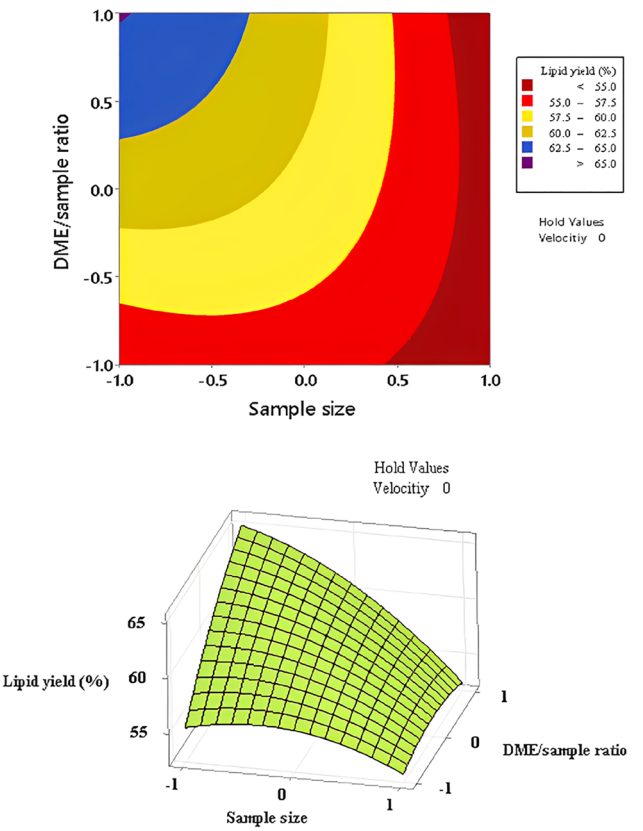

Figure 3. Contour plot and surface plot for the interaction effect of the DME/sample ratio and sample size.

decrease in sample diameter (<3.7 mm) due to the increased contact area between the DME and sludge ball sample.[23]

3.2.2. Interaction of DME/Sample Ratio and Sample Size. Figure 3 shows contour and surface plots of the interactive effect of the DME/sample ratio and sample size on the lipid yield when the velocity was fixed at 4.3 m/h. The DME/sample ratio was varied from 10 to 45 and 80 mL/g to observe the effects of the DME/sample ratio on the lipid yield. The lipid recovery increased gradually when the DME/sample ratio was >40 mL/g. This indicates that using a larger volume of solvent enhances the lipid extraction, leading to an increased yield of extracted lipids.[28,34] However, increasing the DME ratio would require a higher expense and energy; DME reusability could be implemented to compensate for this.

3.2.3. Interaction of Velocity and DME/Sample Ratio. Figure 4 presents contour and surface plots of the yield of lipids as an interactive function of velocity and the DME/sample ratio with the sample size maintained at 3.3 mm. The velocity varied from 2.8 to 4.3 and 5.7 m/h. The DME/sample ratio had a greater influence on the lipid content compared with velocity. Thus, the influence of low velocity was not substantial for a low DME/sample ratio, because an insufficient amount of DME resulted in incomplete lipid extraction. However, it resulted in better extraction performance with the increment of DME amount. Similar results are noted in the literature for process optimization of lipid extraction using the DME technique, in which the lipid content decreased at high velocity as there was less time for DME penetration, thus preventing extraction equilibrium.[36−38]

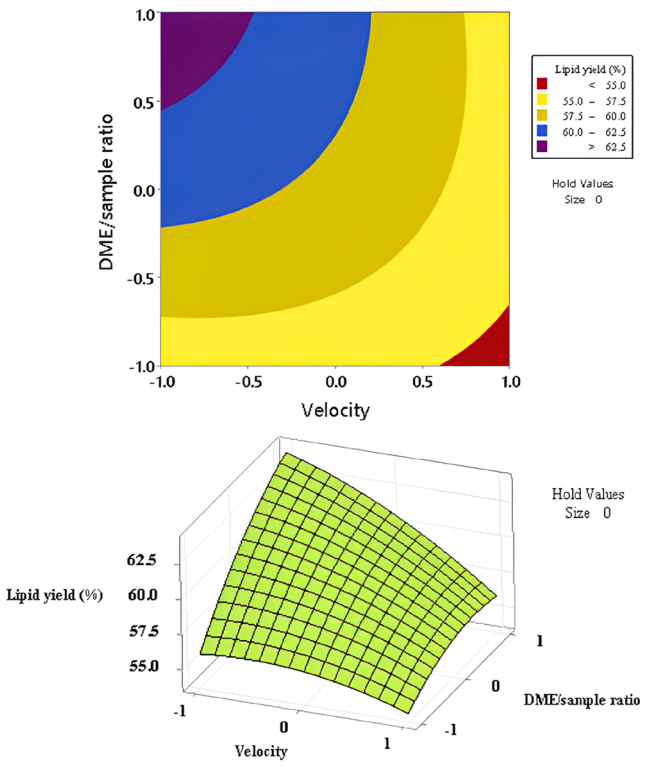

Figure 4. Contour plot and surface plot for the interaction effect of velocity and the DME/sample ratio.

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ACS Omega 2024, 9, 34859−34868

3.3. Optimization and Validation of the Reaction Parameters. The optimization of lipid extraction was determined by using the response optimizer in Minitab software to obtain the optimum combination within the specified range of variables. The optimal conditions for lipid extraction by the DME method were as follows: sample size, 1 (mm); velocity, 3.3 (m/h); and DME/sample ratio, 80 (mL/g). The predicted value under optimal conditions was validated by the experimental results. The observed value was 65.2%, which is in good agreement with the value estimated by the model (65.5%). The contact time (residence time) during optimal conditions is approximately 21.8 s. Detailed calculations are provided in the Supporting Information (Table S2).

3.4. Comparison of DME, Mechanical Shaking, and Soxhlet Methods. *3.4.1. Lipid and Biodiesel Yield.* To analyze the extraction performance and biodiesel yield of DME, the lipid extraction method was conducted by mechanical shaking and Soxhlet extraction with hexane (Figure 5). The maximum lipid yield for fat balls using

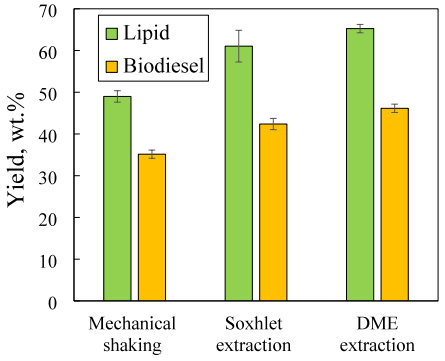

Figure 5. Comparison of the yield and purity of biodiesel produced by the three methods.

DME was higher compared with the yields obtained by mechanical shaking (49.0%) and Soxhlet extraction (62.0%). One possible reason for the superiority of DME extraction was the high solubility and low viscosity, allowing for better diffusion of the solvent into the solid phase of the fat balls.[39] DME is of medium polarity, which can be exploited to extract neutral and complex lipids.[40] In contrast, hexane can only extract nonpolar lipids;[13,41] thus, the difference in polarity contributes to their varied solubility properties and applications in different extraction processes. Previous studies have shown that using more polar organic solvents in the extraction process leads to a higher lipid yield, as these solvents efficiently extract a broader range of lipids, including the phospholipids and glycolipids.[13,42,43]

Although hexane was used in mechanical shaking and Soxhlet extraction, the latter was more efficient due to the use of a reflux condensation system.[14,35] Furthermore, the effectiveness of the solvent for extracting lipids through mechanical shaking decreases as the lipid concentration increases, limiting its potential. The lipids were converted into biodiesel. The transesterifiable matter content was about 69.0−72.0%. The overall biodiesel yields were 46.2%, 42.4%, and 35.2% for lipids derived by DME, mechanical shaking, and Soxhlet, respectively.

Figure 6 shows the mass composition (initial and after treatment) resulting from the three extraction methods. The

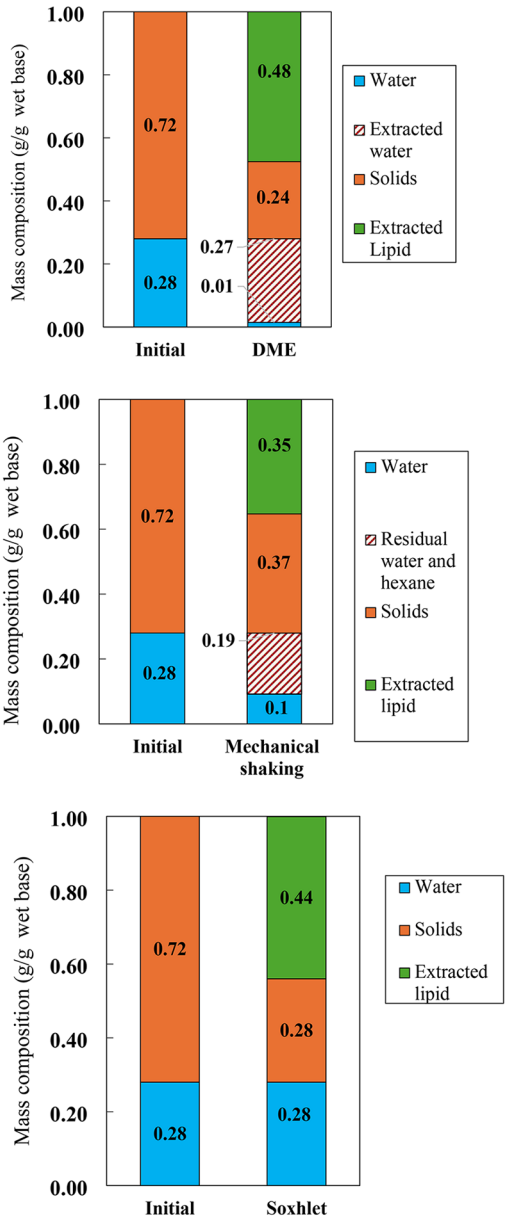

Figure 6. Mass balance (initial and after) treatment using different methods.

DME treatment concurrently extracted lipids and water during the extraction process, generating dried residue that would likely undergo waste-to-energy incineration.[44] Figure S3 (Supporting Information) shows photographs of the raw fat balls and their residue after DME extraction. Conversely, the mechanical shaking and Soxhlet methods still retained a considerable amount of water, which means that further treatment would be required to reach a dried form.

3.4.2. FAME Profiles of the Recovered Lipids. The FAMEs from the fat ball lipids extracted using the three methods were compared to those of other biodiesel feedstocks, as presented in Figure 7. Irrespective of the extraction technique, palmitic acid (C16:0) was the most abundant (>50%), followed by oleic acid (C18:1) and linoleic acid (C18:2). The predominance of palmitic acid in sewage lipids has also been reported by other researchers.[13,45] In this study, the percentage of saturated fatty acids (SFAs) in the lipids

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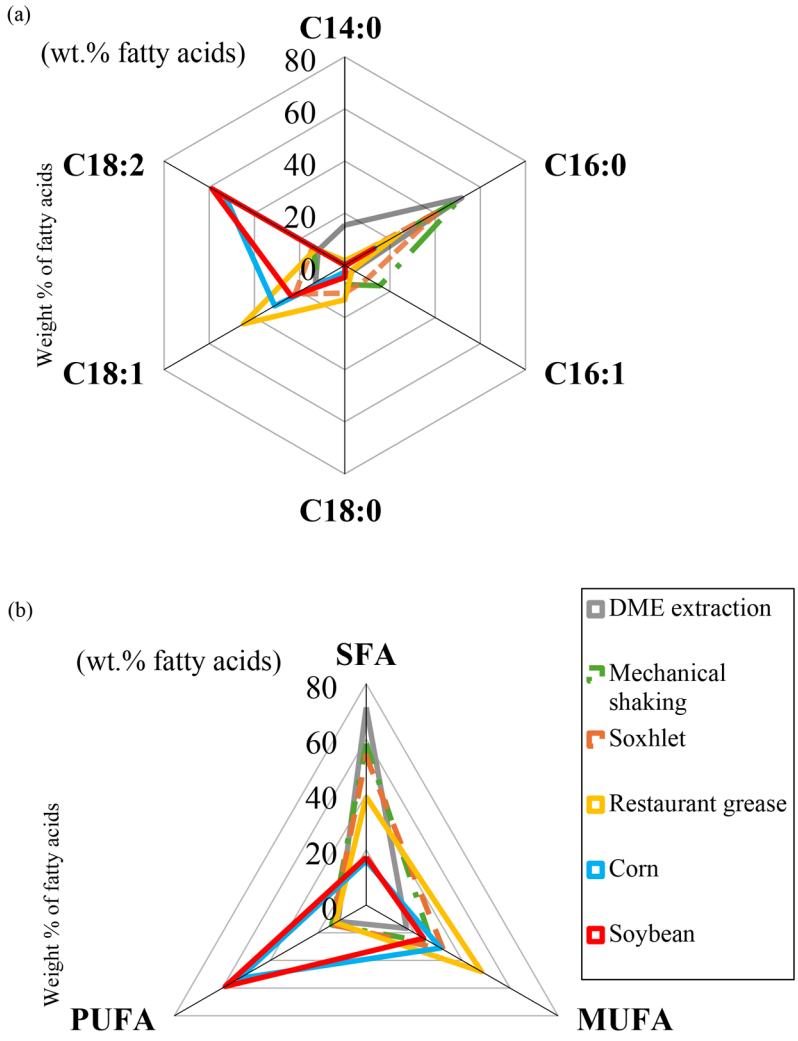

Figure 7. (a) Fatty acid methyl ester (FAME) profiles and (b) FAMEs based on saturated levels. PUFA: polyunsaturated fatty acid (PUFA); MUFA: monounsaturated fatty acid; SFA: saturated fatty acid.

obtained by DME extraction was higher (71%) compared to that from mechanical shaking and the Soxhlet process (53.7−58.3%). However, they showed similar fractions, in which the lipids predominantly consisted of SFA. Meanwhile, polyunsaturated fatty acid (PUFA) was dominant in edible oil feedstock (corn and soybean), followed by monounsaturated fatty acid (MUFA) and SFA. The saturation levels of fatty acids affect the biodiesel properties, including oxidative stability, cetane numbers, and cold-flow properties.[6,46] Biodiesel with a high amount of SFA has higher cloud points, cetane number, and oxidative degradation.[47] On the other hand, more PUFA reduces the cetane number and oxidation stability.[48,49]

3.4.3. FTIR Analysis. The FTIR spectra of raw fat balls, recovered lipids, and fat ball residue are shown in Figure 8. The peak observed between 3100−3600 cm^{-1} and the strong, sharp band at 1720−1740 cm^{-1} were assigned to the stretching vibration of hydrogen bonds and the carboxyl group of esters, respectively.[50,51] Upon lipid extraction, these peaks were absent in the spectra of the fat ball residue, confirming the removal of water and lipids. The peaks detected at 2950 and 2800 cm^{-1} are attributed to C−H stretching absorptions of the methylene and methyl groups in fatty acids.[48,52] The bands of the carboxylate group between 1530−1630 cm^{-1} and 1300−1420 cm^{-1} were evident in the FTIR spectra.[53] The peak near 720 cm^{-1} is likely due to methylene vibration.[50] The FTIR spectra of lipids extracted via DME exhibited some similarities to those obtained through mechanical shaking and the Soxhlet extraction method, indicating the presence of similar components.

3.5. Future Work. In this study, the optimization of the DME process for lipid extraction from fat balls and its comparative efficiency with the existing lipid extraction methods have been demonstrated. However, more research is needed to understand the process's energy consumption. Other challenges remain, such as potential solvent loss during extraction, additional cost for water removal from the extract, and circulation cost for compressing and vaporizing DME. Addressing these challenges is crucial for improving the technical and economic feasibility of the process.

4. CONCLUSION

This work aimed to extract lipids from fat balls as a feedstock for biodiesel production in which RSM-BBD was applied to optimize the process parameters of DME extraction. Our results showed that the use of a smaller sample size, a lower

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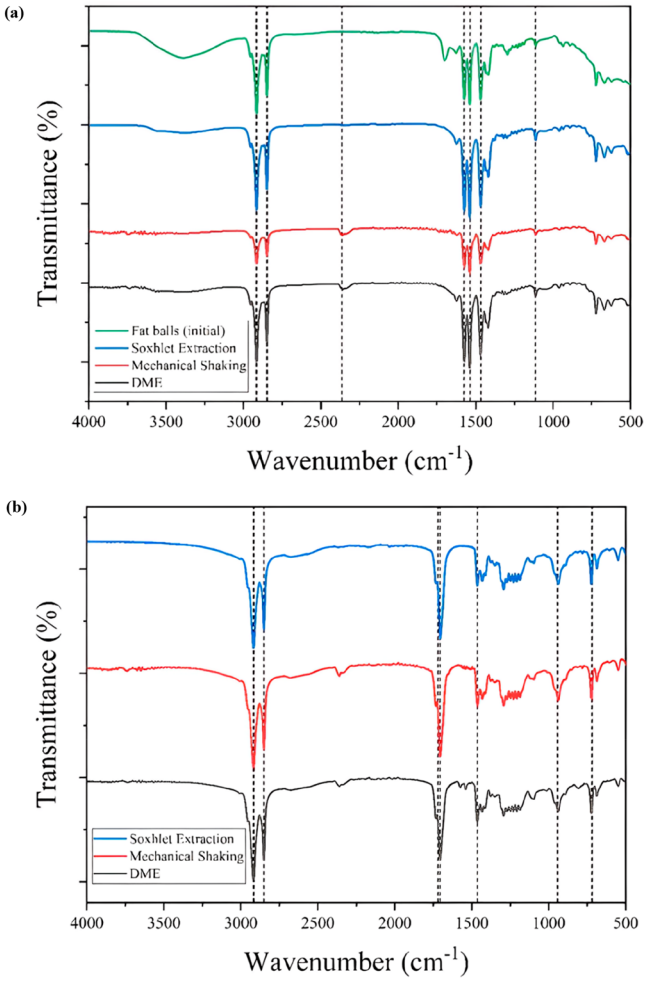

Figure 8. Fourier transform-infrared spectra: (a) raw fat balls and residue and (b) extracted lipids.

velocity of liquefied DME, and a higher DME/sample ratio enhances lipid recovery when using the DME technique. Through response surface optimization, the optimized DME extraction achieved higher lipid recovery (65.2%) than that of mechanical shaking and Soxhlet extraction. The methyl ester characteristics of biodiesel from the extracted lipids were investigated and found to be similar among the three methods. Thus, it can be postulated that the DME technique would enhance lipid recovery to promote the utilization of wastewater lipids as a sustainable source for biodiesel production. Further work needs to be performed to understand the reusability of DME, which can enhance the efficiency of this technology.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsomega.4c04005.

Estimated regression coefficients for the BBD model for lipid extraction by DME (Table S1); Estimated residence time for lipid extraction using DME method under optimum condition (Table S2); Perturbation plot for lipid extraction yield of fat balls by DME method (Figure S1); Actual vs predicted plots for lipid extraction from fat balls using the DME method (Figure S2); Photo images after DME extraction: (a) raw fat balls, (b) residue after DME extraction, (c) the extracted lipids (Figure S3) (PDF)

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The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript.

Notes

The authors declare no competing financial interest.

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■ ABBREVIATIONS

DME, Dimethyl ether; FOG, Fat, oil, and grease; FAME, Fatty acid methyl esters; RSM, Response surface methodology; BBD, Box−Behnken design; TS, Total solid; HHV, Higher heating values; LHV, Lower heating values; FTIR, Fourier transform-infrared spectroscopy; ATR, Attenuated total reflectance; SFA, Saturated fatty acids; MUFA, Monounsaturated fatty acids; PUFA, Polyunsaturated fatty acid.

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