Sequential Production of Lignin, Fatty Acid Methyl Esters and Biogas from Spent Coffee Grounds via an Integrated Physicochemical and Biological Process

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Abstract: Spent coffee grounds (SCG) are one of the lignocellulosic biomasses that have gained much attention due to their high potential both in valorization and biomethane production. Previous studies have reported single processes that extract either fatty acids/lignin or biogas. In this study, an integrated physicochemical and biological process was investigated, which sequentially recovers lignin, fatty acid methyl esters (FAME) and biogas from the residue of SCG. The determination of optimal conditions for sequential separation was based on central composite design (CCD) and response surface methodology (RSM). Independent variables adopted in this study were reaction temperature (86.1–203.9 °C), concentration of sulfuric acid (0.0–6.4%v/v) and methanol to SCG ratio (1.3–4.7 mL/g). Under determined optimal conditions of 161.0 °C, 3.6% and 4.7 mL/g, lignin and FAME yields were estimated to be 55.5% and 62.4%, respectively. FAME extracted from SCG consisted of 41.7% C16 and 48.16% C18, which makes the extractives appropriate materials to convert into biodiesel. Results from Fourier transform infrared spectroscopy (FT-IR) further support that lignin and FAME extracted from SCG have structures similar to previously reported extractives from other lignocellulosic biomasses. The solid residue remaining after lignin and FAME extraction was anaerobically digested under mesophilic conditions, resulting in a methane yield of 36.0 mL-CH_4/g-VS_{added}. This study is the first to introduce an integrated resource recovery platform capable of valorization of a municipal solid waste stream.

Keywords: spent coffee grounds; lignin; fatty acid methyl esters; organosolv; response surface methodology; anaerobic digestion

1. Introduction

Coffee is one of the most widely consumed beverages in the world and is second only to petroleum as a globally traded commodity [1]. According to the International Coffee Organization (ICO), the global coffee industry processed approximately 9.6 million tons in 2017. Following the manufacturing steps of coffee including processing, roasting and brewing, more than 90% of initial dry mass is generated as coffee by-products in the form of so-called spent coffee grounds (SCG) [2]. Most of the SCG found in municipal solid waste is being incinerated, dumped in landfills or composted, raising

environmental concerns of the bio-toxicity of caffeine, tannins and polyphenols in SCG [3]. However, this biowaste SCG are rich in a variety of organic compounds in the form of cellulose, hemicellulose, lignin, lipids and antioxidant compounds [4], representing a valuable energy source.

Currently, several studies on the valorization potential of SCG both for reducing solid waste and applying to alternative resources have been reported employing either physicochemical or biotechnological strategies. Panusa et al. [5] studied the recovery of antioxidants from SCG using pure water and 60% aqueous ethanol. Bio-syngas production from SCG by steam gasification has been shown to be feasible [6]. Also, SCG can be used as additives via improving properties in its formed composites [7]. In addition, extraction of fatty acids from SCG has been investigated using various solvent-based techniques for biodiesel production [8]. Due to the organic richness of SCG, they have been subjects of anaerobic digestion (AD) in the quest to produce biogas using a sustainable biological approach [9]. SCG are especially rich in sugars, proteins and lipids, comprising 45.3% [10], 13.6% [11] and 9.3–16.2% [12], respectively, of its total weight, which makes SCG attractive subjects of AD research [13]. AD is one of the most favored biological processes that can retrieve energy from solid wastes under anaerobic conditions. During AD processes, organic wastes are broken down from polymers to monomers and eventually to methane, a viable source of energy [13]. Since AD processes are mostly operated under the mesophilic temperature region (30–40 °C), the resource recovery platform needs to be established primarily under mesophilic conditions.

Although many chemical components of SCG have been investigated for their valorization, utilizing lignin in SCG via a physiochemical or biological process has been ignored to date as a potential biofuel or resource. Lignin have several functional groups including aliphatic hydroxyl, carboxyl and phenol groups which can be applied in various fields yielding products such as emulsifiers, adhesives, resins, dispersants, paints and fuels [14]. Using these functional groups, lignin can serve as a precursor of carbon fiber, vanilla, phenol and ethylene [15]. Practically, separating lignin from various lignocellulosic biomasses in biorefineries has been carried out in high-value applications [16]. To separate lignin with higher purity and quality from lignocellulosic biomasses, organosolv treatments using an organic solvent and a catalyst at high temperature have been developed as a promising method [17]. After an organosolv treatment, the remaining SCG mass may still be characterized as high lipids and proteins, leading to further reuse as sources for production of biodiesel and biogas. To acquire higher amounts of fatty acid methyl esters (FAME) for converting lipids to biodiesel, an esterification step of SCG with sulfuric acid is required, followed by transesterification of triglycerides [18]. During the organosolv treatment with sulfuric acid, SCG mass will become esterified, eventually simplifying the pretreatment step for lipid extraction of SCG. Additionally, SCG residues after sequential separation of lignin and lipid might be subsequently exploited as a substrate in an AD process for producing biogas.

Sequential energy recovery from SCG via various processes could offset the cost of separation steps. These sequential methods can lead to more sustainable and efficient recovery requiring no complex steps without incurring additional costs. Thus, integrated resource recovery from SCG could present a feasible platform for the valorization of a municipal solid waste stream. To test this premise, organosolv treatment of SCG was carried out to sequentially recover lignin and esterified lipids under identical conditions. To determine the optimal recovery of lignin and FAME, three independent variables (temperature, sulfuric acid concentration and ratio of methanol to SCG) were selected. Experimental sets were designed using a rotatable central composite design (CCD), and modeling for optimization of the variables maximizing recovery of lignin and FAME was performed using response surface methodology (RSM). The lignin isolated via the organosolv treatment was evaluated for physicochemical characteristics and FAME was analyzed to investigate esterified lipids. To measure the biogas production of AD from SCG residues, batch type AD reactors were operated under mesophilic conditions. As SCG are fast becoming one of the main organic fractions of municipal waste, a reliable process needs to be developed to properly treat it or to recover resources from SCG. In this study, an integrated resource recovery platform was investigated which enables sequential separation of lignin, FAME and biogas.

2. Materials and Methods

2.1. Feedstock

Fresh SCG were collected periodically from a local coffee shop located on the campus of Gwangju Institute Science and Technology, Republic of Korea. The sampled SCG had an average 61.0% wt. of water and all samples were immediately used to the organosolv treatment without additional drying to reduce energy costs. To quantify the extractable portion of SCG, the composition of SCG was determined based on the type of analyzed material following protocols provided by the National Renewable Energy Laboratory: (i) ash content (NREL/TP-510-42622); (ii) extractives (NREL/TP-510-42619); (iii) acid insoluble lignin (T222 Om-02); and (iv) acid soluble lignin and monosaccharides (NREL/TP-510-42618). The lipid content was determined by the Soxhlet extraction method using 300 mL of n-hexane (Samcheon Chemical, Republic of Korea) as the solvent for 8 h. Those constituents were measured on a dry weight basis. All samples were analyzed in duplicate.

2.2. Organosolv Pretreatment

Organosolv pretreatment was performed to disintegrate the lignocellulosic structure of SCG with the aid of an acidic catalyst and eventually to dissolute methanol-soluble materials and n-hexane-soluble contents into individual solvents. Organosolv pretreatment of SCG was performed in a 1 L batch reactor (CHEMRE SYSTEM Inc., Anyang, Korea) which was continuously mixed with a magnetic stirrer [19]. The reactor consisted of a stainless steel vessel, electric band heater, magnetic stirrer, safety valve, pressure gauge, temperature sensor and temperature controller box. Pretreatment was performed using 50 g of wet SCG with 61.0 wt.% by varying the temperatures and the concentrations of sulfuric acid and solvent but with a fixed reaction time of 10 min. Two organic solvents having different polarity were chosen as the pretreatment solvent: identical volumes of extra pure n-hexane (Samchun Chemical, Seoul, Korea) or 99% methanol (OCI, Seoul, Korea) were mixed together to facilitate both lignin decomposition and esterification reaction using sulfuric acid. Overall experimental procedures were presented as a schematic diagram (Figure 1).
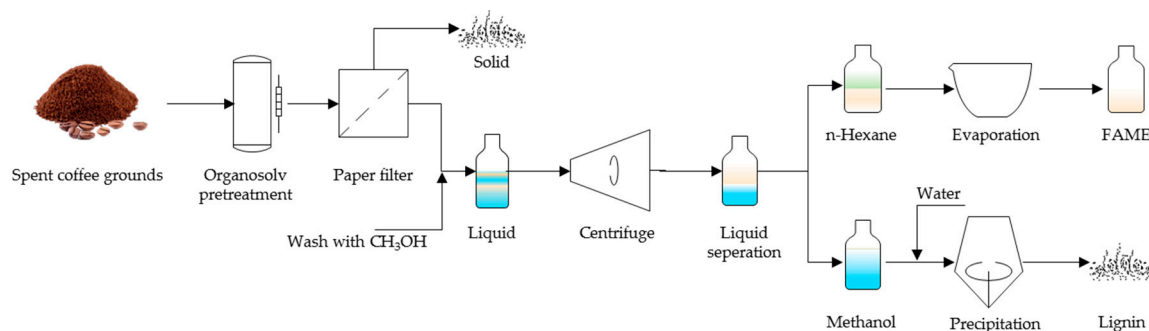

Figure 1. A schematic diagram of sequential separation procedure.

2.3. Recovery of Lignin and Esterified Lipid

Lignin content and esterified lipids were recovered for further analysis to determine the optimal reaction conditions for the proposed resource recovery platform. After pretreatment, the mixture was separated into a solid phase and two liquid phases. The mixed n-hexane and methanol solvents were separated into two layers. To obtain dissolved lignin and biodiesel in the two different solvent layers, the solid particles were separated using a vacuum filtration with Whatman grade 1 paper-filter. After washing the solid phase with the same amount of methanol used for the reaction, the filtrate was centrifuged at 4000 rpm for 10 min with the washing agent for complete separation. The upper layer of the solution was the n-hexane layer with its esterified lipids could be collected by solvent evaporation. The lower layer was methanol with lignin. The solid state of lignin was collected by

adding water equaling three times the volume of total methanol solution and then drying the vacuum filtered precipitates. All samples were stored at −20 °C. The recovery of obtained lignin and FAME was calculated by the following equations [19]:

$$\text{Lignin recovery (\%)} = \frac{\text{Separated lignin}}{\text{Total lignin in initial SCG}} \times 100 \tag{1}$$

$$\text{FAME recovery (\%)} = \frac{\text{Quantified FAME}}{\text{Lipid content of initial SCG}} \times 100 \tag{2}$$

2.4. Experimental Design and Statistical Analysis

Determination of the optimal reaction conditions is the primary concern in developing a process. Rotatable CCD was adopted in this study since it is an experimental design method that can determine the optimal value of the independent variables by using less experiments. Sixteen experimental sets were designed based on a rotatable CCD using three parameters [19]: reaction temperature (86.1–203.9 °C), concentration of sulfuric acid (0.0–6.4 %v/v) and ratio of methanol to SCG (1.3–4.7 mL/g) as independent variables (Table 1). Five levels of varying ranges of independent variables were coded as −1.68, −1, 0, +1 and +1.68 (Table 1). Multiple regression analysis to predict recovery of both lignin and FAME was employed based on the experimental results using RSM (Table 2). A general second-order polynomial equation was used, as expressed in Equation (3).

$$Y = \beta_0 + \sum_{i=1}^{n} \beta_i X_i + \sum_{i=1}^{n} \beta_{ii} X_i^2 + \sum \sum_{i<j-1}^{n} \beta_{ij} X_i X_j + \varepsilon \tag{3}$$

where y is the dependent variable (recovery of lignin and FAME, %); β_0, β_i. β_{ii}, β_{ij} are the coefficients of Equation (3); and X_i and X_j are independent variables.

Table 1. Levels of independent variables.

Level		Independent Variables		
		(X_1) Reaction Temperature (°C)	(X_2) Concentration of Sulfuric Acid (%v/v)	(X_3) Ratio of Methanol to Spent Coffee Grounds (SCG) (mL/g)
+Alpha	+1.68	203.9	6.4	4.7
Max	+1	180.0	5.0	4.0
Central level	0	145.0	3.0	3.0
Min	−1	110.0	1.0	2.0
−Alpha	−1.68	86.1	0	1.3

Table 2. Experimental sets with three independent variables and results of recoveries of lignin and fatty acid methyl esters (FAME).

Serial Number	Independent Variables						Dependent Variables	
	Reaction Temperature (°C)		Concentration of Sulfuric Acid (%v/v)		Ratio of Methanol to SCG (mL/g)		Recovery of Lignin (%)	Recovery of FAME (%)
	X_1		X_2		X_3		Y_1	Y_2
1	110.0	−1	1.0	−1	2.0	−1	38.18	4.76
2	180.0	1	1.0	−1	2.0	−1	29.01	40.46
3	110.0	−1	5.0	1	2.0	−1	60.47	19.66
4	180.0	1	5.0	1	2.0	−1	45.51	41.27
5	110.0	−1	1.0	−1	4.0	1	28.71	13.52
6	180.0	1	1.0	−1	4.0	1	34.21	39.34
7	110.0	−1	5.0	1	4.0	1	62.30	40.46
8	180.0	1	5.0	1	4.0	1	46.42	53.40
9	86.1	−1.68	3.0	0	3.0	0	38.48	4.03

Table 2. *Cont.*

Serial Number	Independent Variables						Dependent Variables	
	Reaction Temperature (°C)		Concentration of Sulfuric Acid (%v/v)		Ratio of Methanol to SCG (mL/g)		Recovery of Lignin (%)	Recovery of FAME (%)
	X_1		X_2		X_3		Y_1	Y_2
10	203.9	1.68	3.0	0	3.0	0	56.20	23.15
11	145.0	0	0.0	−1.68	3.0	0	4.89	0.60
12	145.0	0	6.4	1.68	3.0	0	50.39	23.88
13	145.0	0	3.0	0	1.3	−1.68	61.08	24.28
14	145.0	0	3.0	0	4.7	1.68	59.25	51.04
15	145.0	0	3.0	0	3.0	0	51.31	55.46
16	145.0	0	3.0	0	3.0	0	52.22	53.89

RSM, analysis of variance (ANOVA) and creation of 2D and 3D graphs were performed using Design-Expert (Version 7.0.0, Stat-Ease Inc., Minneapolis, MN, USA).

2.5. Lignin Characterization and FAME Quantification

FT-IR spectra analyses were conducted to ascertain if the extractives obtained from SCG were lignin and FAME as had been expected. FT-IR spectra analyses of the lignin samples isolated from SCG were performed using a Nicolet 6700 FT-IR spectrophotometer (Thermo, Waltham, MA, USA). The lignin samples were recorded with a resolution of 8 cm^{-1} in the region of 4000–650 cm^{-1}. Conversion of triglycerides and free fatty acids (FFAs) to methyl esters was determined by measuring the methyl esters content of biodiesel using gas chromatography (GC). FAME of SCG was separated using GC (Model YL6500 GC, Young Lin Instrument Co., Anyang-si, Korea) equipped with a flame ionization detector (FID), a HP-1 column (30 m × 0.25 mm i.d., 0.20 μm film thickness) and helium (0.7 mL/min) as a carrier gas. The temperatures of the injector and detector were set at 210 °C and 240 °C, respectively. Peaks were identified by comparison to reference standards [20,21].

2.6. Biochemical Methane Potential Test

As an indicator of the amount of energy that can be obtained from wastes, the biochemical methane potential (BMP) needs to be estimated. To determine the anaerobic digestibility of the solid fraction residues of SCG from the organosolv pretreatment, BMP tests were performed for 16 days in 5 L glass reactors (PYREX). Each reactor was filled with 4.5 L of inoculum and mixed with 45 g of the solid fraction residues of SCG. The inoculum to substrate ratio was 10 g-substrate/L-inoculum, which can be converted to 2.43 gVS-substrate/gVS-inoculum. The inoculum was taken from an AD effluent pipeline in a mesophilic AD plant treating sewage sludge located at Gwangju, Republic of Korea. Total solids (TS) concentration of inoculum was 18.87 g/L and volatile solids (VS) was 12.51 g/L. Reactors were incubated at 37 °C and the initial pH was adjusted to 7.00 ± 0.05 with 1 M HCl and NaOH. Biogas produced was collected through a gas sampling line to a gas pack (Tedlar) for further analysis. Specific BMP (mL-CH$_4$/g-VS$_{added}$) was calculated by dividing total volume of methane by the measured total VS$_{added}$. The experiment was conducted following Jang et al. [22]. Biogas production was measured by gas chromatography (Carboxen-1000 column, Model YL6500 GC, Young Lin Instrument Co., Korea) equipped with a thermal conductivity detector and helium as a carrier gas. The temperature of the inlet, oven and detector was maintained at 150 °C [23]. All analysis was carried out in duplicate.

3. Results and Discussion

3.1. Feedstock Characterization

The main constituents of SCG were hemicellulose (representing xylose, mannose, galactose and arabinose, 38.3%), total lignin (acid soluble and insoluble lignin, 28.0%) and lipids (14.9%). Cellulose (glucose, 9.0%) and ash (1.8%) were present as minor constituents and water content was 61.0%.

Chemical compositions of SCG in previous studies [11,24,25] showed similar fraction of hemicellulose (30%–40%), total lignin (25%–33%), lignin (10%–20%) and water (60%–66%), compared to that in our study. Unlike most other lignocellulosic biomasses, SCG had many hemicelluloses, particularly mannans and galactans with only a minor portion of arabinans. A high portion of lignin, one of the essential constituents, contributed to a high C/N ratio, allowing a high calorific value. Considering its lignin content, SCG were seen as a valuable source of lignin, which could be extracted with lipids by subsequent organosolv treatment.

3.2. Effects of Treatment Conditions and Optimization

Based on the experimental results provided in Table 2, the relationship between reaction temperature (X_1), sulfuric acid concentration (X_2), ratio of methanol to SCG (X_3) and recovery of lignin and FAME (Y) were analyzed using RSM. The results obtained were then statistically tested by ANOVA (Table 3). The RSM and ANOVA tests convey that both the proposed models are not significantly different from actual values which is supported by the low values of lack of fit. The ANOVA of equations yielded *p*-values of less than 0.05 confirming the statistical significance of the proposed models. The values of the coefficient of determination (R^2) for recovery of lignin and FAME were 0.815 and 0.819, respectively, indicating that the sample variation in approximately 82% of lignin and FAME recovery can be attributed to the independent variables. The relatively lower values of R^2 for both lignin and FAME recovery might result from errors in the chemical analysis of lignin and FAME.

In the lignin recovery model, the probability of X_2 ($p < 0.001$) was found to have a significant effect on lignin recovery (Y_1) (Table 3). Therefore, the concentration of sulfuric acid exhibited a significant effect on promoting lignin recovery from SCG. An increase in acid concentration improved recovery of lignin in the liquid fraction. This result was consistent with a previous study which found that acid treatment could enhance solubilization of lignin in the aqueous phase through degradation of the linkage between lignin monomers [19]. However, neither applying the lowest nor highest reaction temperature nor altering the ratio of methanol to SCG produced optimal lignin recovery. As shown in Table 3, FAME recovery was positively related to the linear effect of all independent variables (reaction temperature, concentration of sulfuric acid and ratio of methanol to SCG). The quadratic effects of reaction temperature and concentration of sulfuric acid were found, but no interaction effect between independent variables was reported. Among the variables, reaction temperature (X_1) contributed most significantly to FAME recovery (Figure 2a). Increasing the temperature up to approximately 161 °C consistently led to an increase in FAME recovery, regardless of the amounts of sulfuric acid and solvents, but the recovery started to decrease after exceeding 161 °C. A previous study also suggested temperature as the most dominant parameter affecting the kinetics of esterification for biodiesel production [26]. Meanwhile, effects of sulfuric acid on FAME recovery were similar to lignin recovery, exhibiting almost the same optimal concentrations for recovering both lignin and FAME. The FAME recovery was proportional to the increase in the ratio of methanol to SCG within the levels of varying ranges in our study, without indicating an optimal point (Figure 2b).

According to the plots (Figure 2b), the simultaneous maximum recovery of both lignin and FAME at 161.0 °C, 3.6% H_2SO_4 and 4.7 mL/g (ratio of methanol to SCG) were estimated to be 55.50% and 62.4%, respectively. To validate our model, simultaneous extraction of lignin and FAME from SCG was conducted under the optimal conditions derived from the 2D contour graph. The results revealed that the developed model could closely estimate the actual amounts of extracting both lignin and FAME from SCG by organosolv treatment (data not shown). However, compared to other studies reporting higher FAME recovery from SCG [11,24–27], our maximum FAME recovery was slightly lower, probably resulting from a decrease in solubility of FAME by reduction in the dielectric constant of methanol under high temperature conditions [28]. On the other hand, lignin recovery was minimally affected by our proposed method for sequential extraction, considering lignin recovery ranging from 30% to 55% by organosolv treatment of lignocellulosic biomasses [19,29]. These results suggest that

simultaneous recovery of lignin and FAME from SCG via organosolv treatment might be feasible, raising the possibility of SCG utilization as an untapped source of lignin.

Table 3. ANOVA for the fitted model of recovery of lignin and FAME.

| Regression	Source	Sum of Squares	Degrees of Freedom	Mean of Square	F-Value	Prob > F	Remarks
Lignin	Model	2921.48	2	1460.74	28.64	<0.001	Significant
	X_2	1901.08	1	1902.95	37.30	<0.001	Significant
	X_2^2	1020.40	1	1020.40	20.00	0.001	Significant
	Residual	663.14	13	51.01			
	Lack of fit	32.74	2	16.37	0.29	0.757	Not significant
	Pure Error	630.40	11	57.31			
	R^2	0.82					
FAME	Model	4348.03	5	869.61	9.05	0.002	Significant
	X_1	1203.93	1	1203.93	12.54	0.005	Significant
	X_2	672.89	1	672.89	7.01	0.024	Significant
	X_3	536.22	1	536.22	5.58	0.040	Significant
	X_1^2	624.71	1	1196.90	12.46	0.005	Significant
	X_2^2	1310.29	1	1310.29	13.64	0.004	Significant
	Residual	960.39	10	96.04			
	Lack of fit	959.16	9	106.57	86.47	0.083	Not significant
	Pure Error	1.23	1	1.23			
	R^2	0.82					
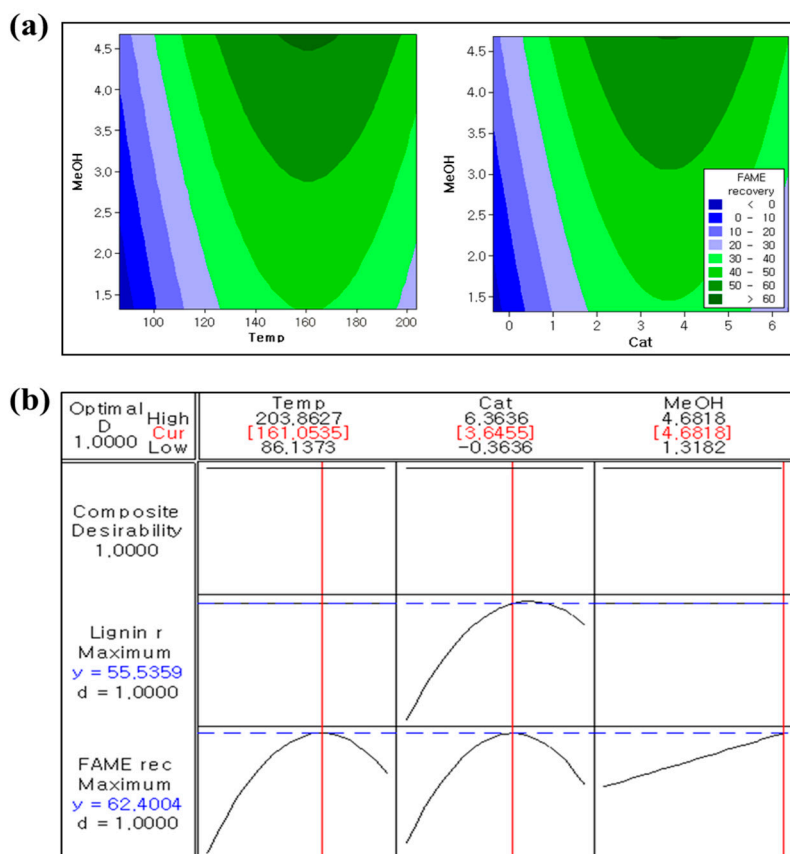
Figure 2. (**a**) 2D contour and (**b**) optimization plot describing the effects of temperature, sulfuric acid concentration and ratio of methanol to SCG on recovery of lignin and FAME from SCG via organosolv treatment.

3.3. Structural Characterization of Lignin and FAME

The FT-IR spectra of lignin from SCG were analyzed band assignments as shown in Table 4. SCG-lignin in Figure 3a shows that a broad band around 3200 cm^{-1} is related to phenolic and aliphatic O–H stretching; the peak at 2927 cm^{-1} represents the characteristics of C–H stretching in the methyl and methylene groups of side chains. The peak at 1703 cm^{-1} was assigned as non-conjugated carbonyl groups. Non-etherified phenolic OH groups in lignin corresponding to a 1366 cm^{-1} peak were found, which had been observed in the final structure of lignin isolated by the Kraft method. Despite using higher concentrations of H$_2$SO$_4$ catalyst in our proposed method than in general organosolv treatments, the lignin structure of SCG had characteristics similar to extracted lignin shown in other studies [30–32]. For organosolv treated FAME (Table 4), the esters were exhibited at two characteristically strong peaks arising from carbonyl around 1741 cm^{-1} and C–O stretching at 1300–1000 cm^{-1} [33]. The most prominent carbonyl peak (1741 cm^{-1}) (Figure 3b) might be attributed to a decrease in the aldehyde peak (1727 cm^{-1}) or the FFA peak (1712 cm^{-1}) via oxidation or esterification [34]. These responses might indicate successful conversion of FAME through an esterification step of SCG, but further analysis is required for better understanding of the removal efficiency of FFA. The compositions of FAME produced from sequential extraction of lignin and FAME under optimal conditions showed the highest contents (90%) of saturated C16 (41.7%) and C18 (48.16%) acids (Figure 4), which is similar to the result from wet in-situ transesterification of SCG [35]. These resultant fatty acids could be regarded as suitable for biodiesel production [36]. However, accomplishing complete transesterification required post-treatment with a base catalyst [37].
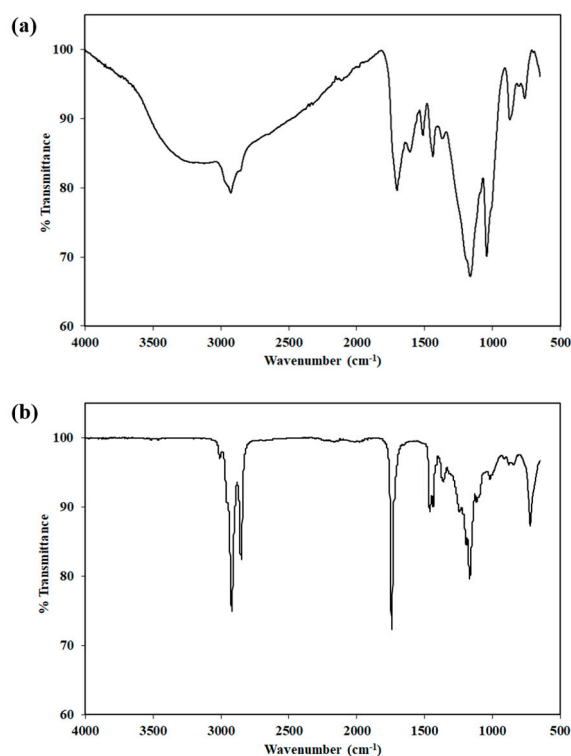

Figure 3. FT-IR spectra of (**a**) SCG-lignin and (**b**) SCG-FAME.

Table 4. Assignment of FT-IR spectra of SCG-lignin and SCG-FAME from organosolv.

Peak (cm^{-1})		Assignment
SCG-lignin	SCG-FAME	
3200	-	Phenolic and aliphatic O–H stretch
-	3008	C–H stretching alkene
2927	-	C–H stretch
-	2922	C–H stretching of CH_2 and CH_3
-	2852	C–H stretching of CH_2 and CH_3
-	1741	C=O stretching of ester
1703	-	C=O stretch unconjugated
1608	-	C–C stretch (aromatic skeletal vibration)
1512	-	C–C stretch (aromatic skeletal vibration), G
-	1460	C–H deformation of methyl and methylene
1439	-	C–H deformation (aromatic skeletal vibration)
-	1436	Aromatic C–H deformation
1366	-	Phenolic O–H and aliphatic C–H stretch methyl groups
-	1362	O–CH_2 deformation in glycerol moiety
-	1242	Phenolic O–H deformation
-	1194	O–CH_3 stretching
1164	-	C–O stretch in ester group
-	1163	C–O stretching
-	1117	O–CH_2–C stretching in triglyceride
1044	-	Aromatic C–H deformation G + S
-	1016	C–O stretching of ester groups
874	-	C–H glycosidic linkage and G
-	844	C–O stretching of ester groups
763	-	CH alkene
-	722	aromatic compounds

3.4. Digestibility of SCG Residues in a Batch Anaerobic Digester

Since SCG contain a high percentage of carbohydrates, proteins and lipids, they have been investigated as a source of biogas production [38]. Atabani et al. [39] suggested a research approach utilizing organic solvent extractives for biodiesel and glycerin production from SCG, while the remaining solids fraction was used as feedstock for ethanol fermentation or biogas production. However, specific attempts to produce biogas were not performed via sequentially exploiting solid fraction remaining after extracting both lignin and lipids by organosolv treatment. To investigate methane yields of the solid remaining SCG extracted from organosolv treatment in the AD, the values of cumulative methane production were analyzed in BMP tests (Figure 5). The result was estimated to be 36.0 mL-CH_4/g-VS_{added}, a relatively low value compared to conventional organic wastes used for methane production [13]. This result was inconsistent with a previous study, which reported that organosolv treatment could improve BMP of lignocellulosic substrate through degrading non-biodegradable lignocellulosic structures into easily digestible materials [40]. During the organosolv treatment in our study, the liquid fraction was simultaneously separated for further recovery of lignin and lipids, probably resulting in the remaining solid fraction having insufficient organic material available for fermentation by anaerobes. After the sequential organosolv-liquid/solid separation process, a decrease in concentrations of total chemical oxygen demand (tCOD) in the remaining SCG solid fraction could account for the relatively low BMP of the remaining SCG. A total COD of 75.7% in the initial SCG samples was detected in the liquid fraction after liquid/solid separation. As a result of the process, the organic residue in the solid fraction decreased, explaining the decrease in theoretical BMP to 16.1% of original SCG (theoretical BMP of SCG with 248.48 mL-CH_4/g-VS_{added}), which corresponds to 40.0 mL-CH_4/g-VS_{added} (Table 5). Thus, the remaining solid SCG cannot be recommended as the sole substrate for bio-methanation. It may require co-digestion with higher amounts of organic matter such as food waste.
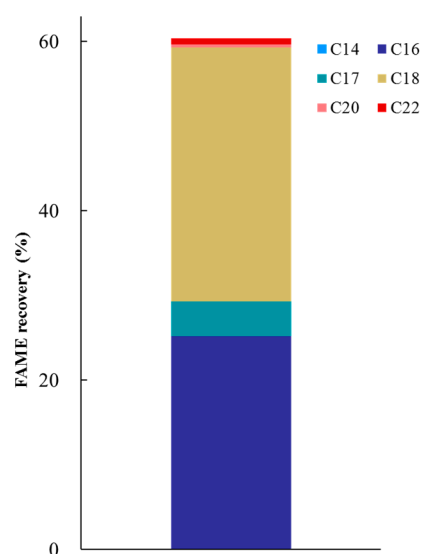
Figure 4. Compositions of FAME obtained under optimal conditions for sequential extraction.
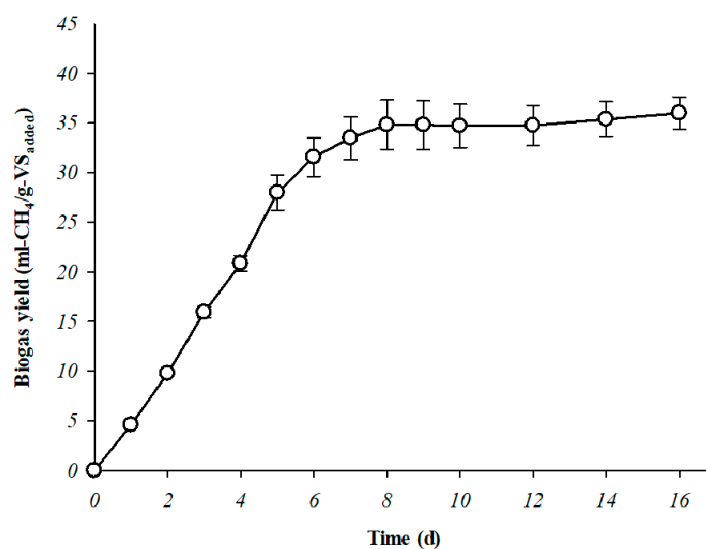

Figure 5. Cumulative methane production in batch type anaerobic digestion of the solid remaining SCG extracted using organosolv treatment.

Table 5. Chemical compositions of the solid fraction residues of SCG and theoretical biochemical methane potential (BMP) of the solid remaining SCG extracted from organosolv treatment.

Parameter	Unit	Value	
		Original SCG	**The Solid Remaining SCG**
Total solids (TS)	g-TS/g	0.977 ± 0.001	0.958 ± 0.010
Volatile solids (VS)	g-VS/g	0.960 ± 0.005	0.720 ± 0.010
Total COD (tCOD)	g-COD/g	1.030 ± 0.034	0.250 ± 0.014
Soluble COD (sCOD)	g-COD/g	0.038 ± 0.001	0.053 ± 0.002
Lignin fraction	% (w/w)	28.200 ± 0.300	36.500 ± 0.081
Theoretical BMP [a]	mL-CH$_4$/g-VS$_{added}$	248.480	40.000
Specific BMP	mL-CH$_4$/g-VS$_{added}$	-	36.000 ± 1.150

[a] Theoretical BMP (mL-CH$_4$/g-VS$_{added}$) = TCOD (g-COD/g) × 0.35 L-CH$_4$/g-COD × g-VS/g × (1-lignin fraction).

4. Conclusions

In this work, the possibility of achieving integrated biofuel production from SCG was evaluated sequentially using organosolv treatment and anaerobic digestion. Using the parameters of 161.0 °C (temperature), 3.6% (sulfuric acid concentration) and 4.7 mL/g (ratio of methanol to SCG), optimal recoveries of lignin and FAME were achieved of 55.5% and 62.4%, respectively, with short reaction times. The methane yield of the solid remaining SCG extracted from organosolv treatment was estimated to be 36.0 mL-CH_4/g-VS_{added}. Thus, our integrated resource recovery from SCG could present a feasible platform for a municipal solid waste stream. However, further study should be conducted for each process to identify the optimum conditions for cost-effective SCG valorization.

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