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Effects of the ratio of carbon to nitrogen concentration on lipid production by bacterial consortium of sewage sludge using food wastewater as a carbon source

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Abstract−Food wastewater (FWW) and sewage sludge (SS) were used to control the C : N ratio in cultures as a method to increase lipid production by microbial species in SS. FWW and SS were mixed in volumetric ratios (FWW : SS) of 5 : 0 (F5), 4 : 1 (F4), or 3 : 2 (F3). Compared to raw SS, total lipid content production was increased by 263% in F5, 142% in F4, and 111% in F3. These results were caused by increases in the concentrations of triglycerides (TAGs) during lipid enhancement. The fatty acid methyl ester content of TAGs (wt% of extract) was 25.3 in F5, 20.2 in F4 and 13.25 in F3; these were significant improvements over biodiesel production using raw SS. C16:0 fatty acid was mostly converted to C18:1 fatty acid; this is an important result because the proportion of C18:1 strongly influences the quality of biodiesel. This is the first effort to produce biodiesel using FWW instead of synthetic medium as a carbon source. Hence, this study provides a useful solution for treating organic wastes (SS and FWW) simultaneously; this strategy may be an economically viable method for producing biodiesel from organic wastes.

Keywords: Sewage Sludge, Food Wastewater, Lipid, Triacylglycerol (TAG), Fatty Acid Methyl Esters (FAMEs), Biodiesel

INTRODUCTION

Biodiesel is produced by trans-esterification of lipid sources from a feedstock. However, biodiesel is currently an uneconomic energy source because of the high price of feedstock (70-80% of total production cost) [3]. Hence, identification of a low-priced feedstock for biodiesel production is necessary; sewage sludge (SS) is one possibility.

SS is an abundant biomass that is generated by wastewater treatment plants (WWTPs) and generally categorized as either primary sludge or secondary sludge [4]. Primary sludge comes from the primary clarifier and is composed of several types of organic matter (cellulose, lipids, proteins) and a minor inert fraction. Lipids from primary sludge contain a composite organic matrix (greases, fats, oils) that originates from domestic and industrial waste. Secondary sludge, also known as activated sludge, comes from the secondary clarifier and is mostly composed of microorganisms, primarily bacteria [5]. Thus, lipids from secondary sludge contain usually lipidic compounds that originate in the cell membranes of microorganisms. These differences in lipid characteristics between primary and secondary sludge can influence the homogeneity of biodiesel, so this study will focus on secondary sludge to ensure consistent lipid constitution.

According to economic analysis, biodiesel production using SS is only practical if the yield is >10% (wt/wt on the basis of dry sludge) [6]. Several studies related to biodiesel production from sewage sludge have obtained yields near 10% when extracting lipids and producing fatty acid methyl esters (FAMEs) by changing the solvents or the catalysts. Using methanol and hexane as independent solvents, 10.04% of lipid content (on the basis of total lipid in dry sludge) was obtained using methanol and 3.04% of lipid content was obtained using hexane from the secondary sludge [7]. The estimated FAME yields from the solvents were 26.89% and 30.28% from the secondary sludge, respectively. Use of mixed polar and non-polar solvents obtained a maximum lipid yield of 27.43% (on the basis of total lipid in dry sludge) using Hexane : Methanol : Acetone in 3 : 1 : 1 volumetric ratio; and use of hexane, methanol, and supercritical CO_2 as solvents obtained maximum yields of 1.94%, 19.39%, 3.55% (on the basis of total lipid in dry sludge), respectively [8]. However, these studies have focused on extraction of pre-existing lipids from the sludge. Efficient methods to increase the lipid fraction are desirable.

Bacteria can synthesize a lipid storage compound (i.e., acylglycerol) under physiologically stressful conditions [9], especially nitrogen limitation [10]. Thus, recent research has attempted to increase lipid accumulation in sludge by controlling the C : N ratio in a synthetic medium [11]. The quantities of saponifiable lipids [i.e., free fatty acids (FFAs), triglycerides (TAGs), diglycerides (DAGs), phospholipids (PLs)] were dramatically increased (59-222% and 150-250% from conventional activated sludge and oxidation ditch processes, respectively) and the yields of FAMEs originated from TAGs also showed significant improvements from 6% to 10% (conventional activated sludge) and 4% to 8% (oxidation ditch). However,

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this research used synthetic medium, and this strategy can increase the cost of lipid production. To increase the economic viability of the process, the cost of the growth medium must be reduced, but it should still have high concentrations of organic carbon. In this study, food wastewater (FWW) was adapted as a carbon source to control C : N ratio. FWW has high concentration of carbon and relatively low concentration of nitrogen; these traits mean that it may be an optimum medium for growth of bacteria, to increase lipid concentration in the sludge. Also, approximately 9,398 tons/d of FWW is released as a secondary pollutant from the food waste recycling process generated in Korea, 2012 [12] and this rate has increased annually, so use of FWW may contribute to increase the economic viability of biodiesel production.

The main objective of this study was to provide practical information about the conditions of biodiesel production from activated sludge using not synthetic medium but a natural organic wastewater, FWW. Particularly, FWW and SS were mixed in reactors at different ratios to control the C : N ratio, and purified air was supplied to each individual reactor with complete mixing to maintain aerobic conditions. Another objective of this study was to quantify the effect of lipid enhancement on the speciation of lipids in the sewage sludge. Hence, this study provides the information about period of reaction and ultimate profile of lipid species.

METHODS

1. Sample Preparation

1-1. Food Wastewater

FWW (Table 1) was collected from a food waste recycling center in Pohang, Korea. This plant treats 180 ton/d of food waste and generates 50 ton/d of FWW. The FWW also was filtered through a 1.0-mm sieve to remove inert matter, then distributed to 4-L bottles. FWW was used as a substrate in 3 : 2 (F3) and 4 : 1 (F4) and the activity of microbial species in FWW is also important because of its role as an inoculum at high C : N ratio in 5 : 0 (F5). Before the experiment, FWW was filtered through a GF/C glass microfiber filter (47-mm diameter, 2.7 μm pore size, Whatman) to remove extra solid matter that could interfere with biomass measurement. Samples were frozen (~ −25 °C) until use.

1-2. Activated Sludge

Activated SS (Table 1) was collected from the municipal WWTP in Daegu, Korea. This WWTP treats 520,000 m^3/d of domestic wastewater and generates 220 m^3/d of activated sludge. Before use in the experiment, the sample was filtered through a 1.0-mm sieve to remove inert matter, and was transferred to 4-L bottles. Samples were refrigerated (~4 °C) until use to prevent decomposition [13].

2. Lipid Enhancement of FWW and SS

2-1. Mixing Ratio

Initial mixing ratio of SS and FWW was chosen by referring to a previous study [14]. FWW and SS were mixed in volumetric ratios 5 : 0 (F5), 4 : 1 (F4), or 3 : 2 (F3) to control the C : N ratios in the cultures. Each condition was operated in duplicate.

2-2. Operating Conditions

Overall lipid enhancement processes (Fig. 1) were performed according to previous research [14]. In this study, 5-L fermentation reactors were designed to control the temperature during the batch experiments; the initial working volume was 3 L per reactor. The pH was not controlled, but was monitored by using a built-in pH probe (405-DPAS-SC-K85, METTLER TOLEDO, Switzerland). Temperature was maintained at 25±1 °C using a refrigerating bath (RW-0525G, JEIO TECH, Korea) and monitored with a thermometer (AL568M101ISO, Daehan Science, Korea) during the whole process. Lipid synthesis requires aerobic conditions, so purified air was supplied after filtration through a 0.45-μm HEPA vent filter (Whatman, Kent, UK) and injected into the culture at 1 vvm (volume of air per volume of media per minute). The reactors were initially agitated at 400 rpm; the rate was gradually increased to 800 rpm in increments of 100 rpm. Foaming was prevented by using Antifoam 204 concentrate (Sigma-Aldrich, St. Louis, MO). Reactions were continued for 7 d; each experiment was performed in duplicate.

Table 1. Characteristics of secondary sludge and food wastewater (average±standard deviation)

Parameter	Secondary sludge	Food wastewater
pH	6.84±0.03	4.68±0.15
TS (g/L)	49.28±1.29	135.46±1.81
VS (g/L)	38.51±1.88	120.54±2.33
TSS (g/L)	43.28±4.38	50.45±0.5
VSS (g/L)	32.12±2.31	3.27±0.54
TCOD (g/L)	42.96±0.42	114.24±1.55
SCOD (g/L)	1.23±0.14	104.12±0.4
Carbohydrate (g/L)	4.44±0.27	12.05±0.32
Protein (g/L)	1.22±0.11	5.15±0.32
Lipid (g/L)	3.41±0.08	0.32±0.01
TAGs wt% of extract	3.64±0.1	0.28±0.02
FFAs, DAGs, MAGs wt% of extract	2.83±0.1	24.87±1.03
Wes and SEs wt% of extract	5.21±1.1	10.62±0.38
PLs wt% of extract	6.8±0.2	0.81±0.03
Nitrogen		
TN (g/L)	3.88±0.55	3.36±0.2
STN (g/L)	1.18±0.13	2.92±0.2
NH_4^+-N (g/L)	0.84±0.04	0.63±0.33
NO_2^- (g/L)	N.D.	N.D.
NO_3^- (g/L)	0.13±0.14	0.18±0.01
Phosphorus		
TP (g/L)	1.71±0.12	3.53±0.33
STP (g/L)	0.53±0.11	2.33±0.28
PO_4^{3-} (g/L)	0.41±0.04	1.97±0.02
Total carbon sources (g/L)	1.14±0.08	62.84±0.81
Glucose (g/L)	N.D.	20.99±0.40
Acetic acid (g/L)	1.14±0.08	8.23±0.31
Propionic acid (g/L)	N.D.	N.D.
Lactic acid (g/L)	N.D.	20.9±0.53
Mannitol (g/L)	N.D.	10.86±0.02

Note: N.D.: "not detected"

3. Analytical Method

3-1. Physico-chemical Analytical Method

Total solid (TS), volatile solid (VS), total suspended solid (TSS), volatile suspended solid (VSS), total nitrogen (TN), total phospho-

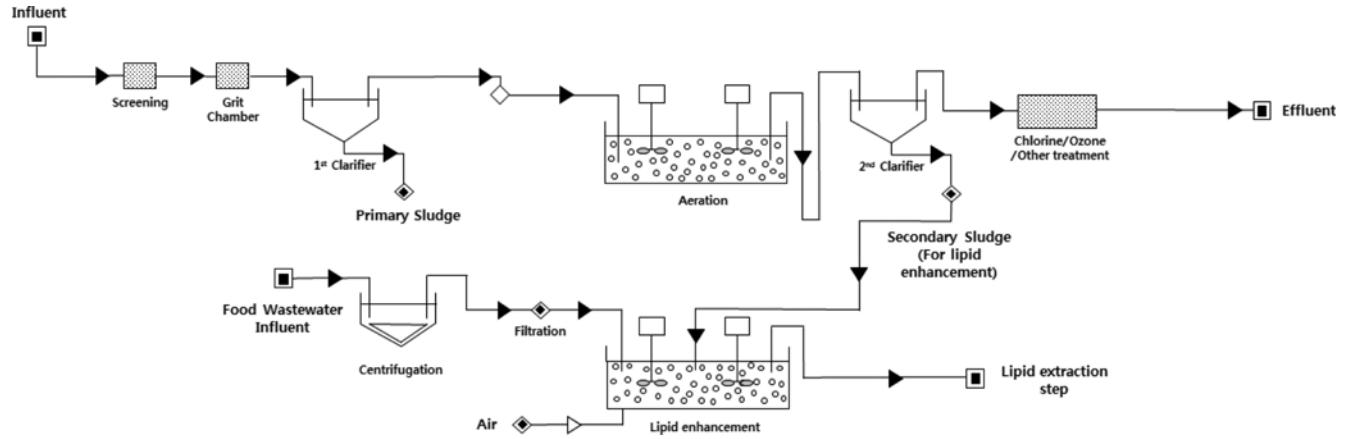

Fig. 1. Schematic diagram of lipid enhancement processes using activated sludge with FWW as a substrate. Components and processes are described in the text.

rus (TP) and chemical oxygen demand (COD) were measured according to the standard method [15]. VSS quantity was used as the estimate of biomass concentration during the process. Characteristics of soluble components were determined after insoluble particles were eliminated by using a 0.45-μm-pore polyethersulfone syringe filter (Millipore, USA). Carbohydrate quantity was measured by the phenol-sulfuric acid method [16], and protein quantity was measured by the Lowry-Folin method [17].

Glucose, mannitol and VFAs were analyzed through high-performance liquid chromatography (HPLC, Agilent Technology 1100 series) equipped with an Aminex HPX-87H column (BIORAD Inc., USA), a diode array detector and a refractive index detector. The eluent was 0.004 M H_2SO_4, and the flow rate was 0.6 mL/min. Temperature during analysis was maintained at 50 °C. Every sample was filtered by using a 0.22-μm syringe filter (Millipore, USA).

3-2. Determination of Initial C : N Ratio

Initial C : N ratio (Table 2) for each condition was estimated as the ratio of the proportion of total organic carbon (TOC) to the proportion of nitrogen (NH_4^+, NO_3^-) in the aqueous phase. All samples were centrifuged at 8,000 rpm to remove solid particles, then passed through a 0.22 μm syringe filter (Millipore, USA) before analysis. Total organic carbon was measured by using a total organic carbon analyzer (TOC-5000A, Shimadzu, Japan) and estimated as the difference between total carbon (TC) and inorganic carbon (IC).

Ammonium-nitrogen (NH_4^+-N) was measured using an ICS 1100 ion chromatograph (DIONEX Co., USA) and the anion concentration including nitrate-nitrogen (NO_3^--N) was determined using ion chromatography (ICS-1000, DIONEX Co., USA).

3-3. Lipid Extraction

Total lipid extraction was performed by a modification [19] of the method developed by Bligh and Dyer [18]. A freeze-dried sample was mixed with chloroform and methanol (2 : 1, v/v); the lipid dissolved in the chloroform. Then the chloroform was removed using a nitrogen evaporation system (Organomation Associates Inc., USA), and the weight of remaining solid was measured to determine gravimetric lipid content (%, weight of lipid/weight of biomass).

3-4. Lipid Analysis

Lipids in the extracts were separated as described previously [20]. The procedure entails precipitation, sequential solid phase extraction, thin layer chromatography and gas chromatography. Each lipid component [i.e., triglycerides (TAGs), diglycerides (DAGs), monoglycerides (MAGs), wax esters (WEs), steryl esters (SEs), free fatty acids (FFAs), phospholipids (PLs)] was separated and analyzed by using a solid phase extraction procedure with a 1000-mg extra-clean silica column (Grace Davison Discovery Sciences, IL, USA) followed by gas chromatography. For separation of lipids, n-hexane/diethyl ether (94 : 6), n-hexane/diethyl ether/acetic acid (85 : 15 : 2) and methanol were used as elution solvents.

3-5. Fatty acid Methyl Esters

The composition of fatty acid was determined by transesterification of lipid extract followed by gas chromatography analysis. First, 1 mL of 2.5% (v/v) sulfuric acid in methanol was added to the lipid extract. The samples were mixed vigorously, then incubated at 90 °C for 45 min. Then 1 mL H_2O and 2 mL n-hexane were added and each sample was mixed using a vortex for 5 min. The samples were centrifuged at 2,000 rpm for 15 min, then the upper phase (n-hexane with FAMEs) was transferred into a glass vial. The FAMEs were identified with a gas chromatograph (6890N, Agilent, USA) equipped with a flame-ionized detector (FID) and an INNO-WAX capillary column (Agilent, USA, 30 m×0.32 mm×0.5 μm). The

Table 2. Experimental design used in this study (average±standard deviation)

Treatment no.	Condition	TOC (g/L)	Nitrogen (NH_4^+, NO_3^-) (g/L)	C : N ratio
1	F5 (F5 : S0)	61.23±1.31	0.82±0.01	74.67 : 1
2	F4 (F4 : S1)	49.42±0.81	0.93±0.01	53.14 : 1
3	F3 (F3 : S2)	39.95±0.09	1.19±0.02	33.57 : 1

oven temperature was set at 100 °C and held there for 5 min, then increased at 10 °C/min to 250 °C and held constant for 30 min. The temperatures of injector and detector were set at 250 °C. FAMEs analyzed were identified and quantified by comparing the peak areas and retention times with those of a standard FAME mixture containing C_8-C_{24} methyl esters (Sigma-Aldrich, St. Louis, MO).

RESULTS AND DISCUSSION

1. C : N Ratio Effects

Initial concentrations of organic acids, glucose and mannitol all increased with the ratio of FWW in the medium; the concentrations decreased over time logistically after a lag period (Fig. 2). Initially, all growth media contained sufficient carbon sources such as glucose, acetic acid and even mannitol in FWW, which can be used by the bacterial consortium in the sludge. In the F5 culture, glucose and acetic acid were almost totally consumed, but mannitol and lactic acid were not consumed within 7 days of reaction; in contrast, in the F4 and F3 cultures, all of the organic carbon sources were consumed within 7 days. The difference in consumption rates can be explained by the effect of high C : N ratio. The rate of carbon uptake is limited when the C : N ratio is high [14], because metabolic fluxes are channeled to lipid accumulation rather than to cell growth in this condition. Because the consumption rate of organic carbon is much slower during lipid accumulation than during cell growth, the consumption of carbon sources is much slower at high C : N ratio than at low C : N ratio [14]. This phenomenon also affects lipid content and biomass concentration.

Because the pH was not controlled, it increased from 4.5 to 6.5 due to the consumption of organic acids, but this increase was slower in F5 than in F4 and F3, in which it increased at nearly the same rate (Fig. 3(a)). The relatively slow increase in F5 is also due to the limited consumption of organic acids when C : N ratio is high.

Growth rate of biomass was significantly lower in F5 than in F4 and F3. Biomass concentration did not change until 1.5 days on average; then in F5 it increased slowly and linearly, but in F4 and F3 it increased exponentially then decreased starting at 3 days in F4 and at 2.5 days in F3 (Fig. 3(b)). The exponential increase was slower in F4 than in F3. The decrease after the exponential phase occurred because carbon sources that were available for bacterial consortium had become limiting. Little research has been performed concerning the effect of C : N effect on bacterial biomass; more has considered this effect on biomass production by oleaginous yeast. Increase in biomass of the Saccharomycete *Lipomyces*

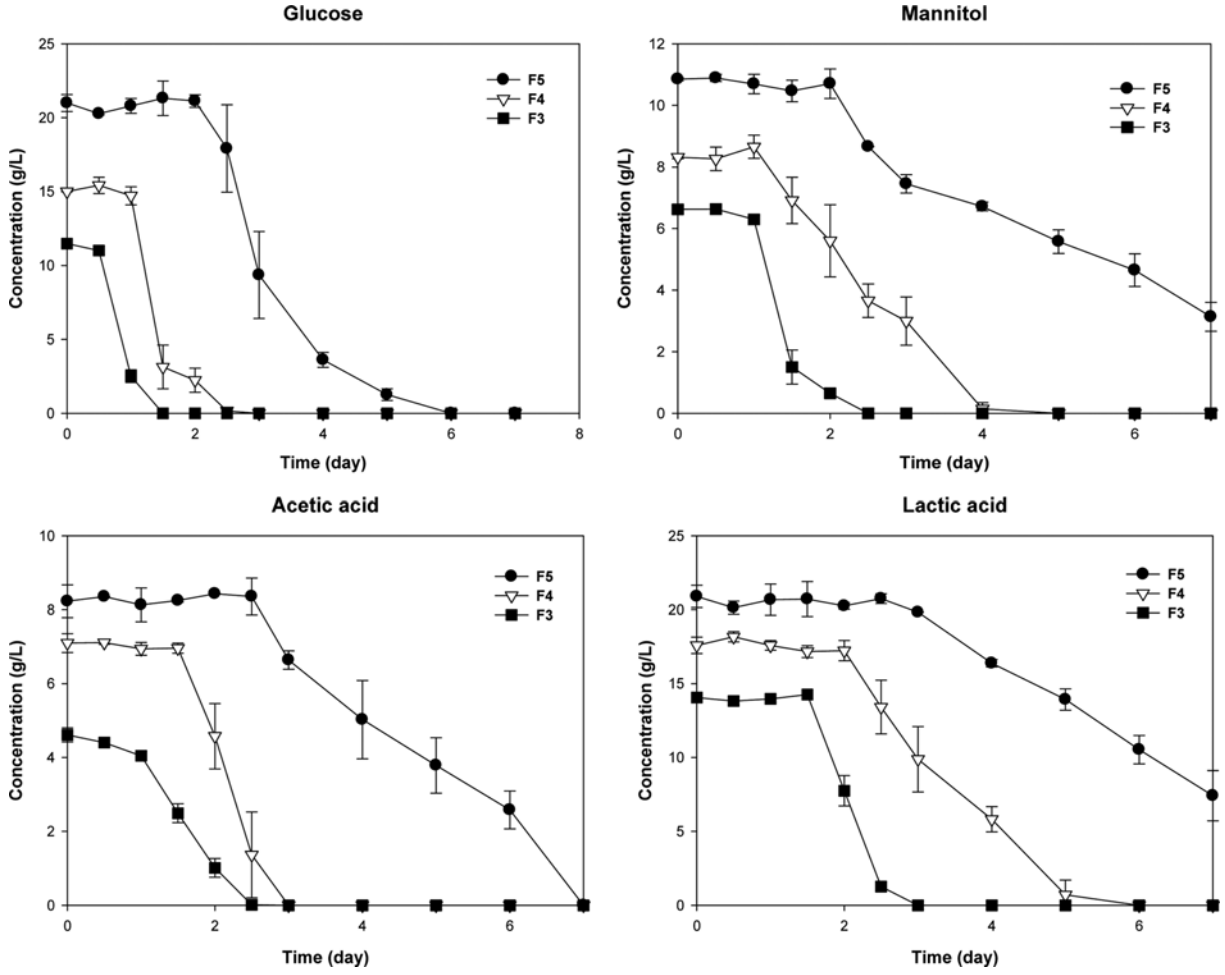

Fig. 2. Profile of various carbon sources during lipid enhancement.

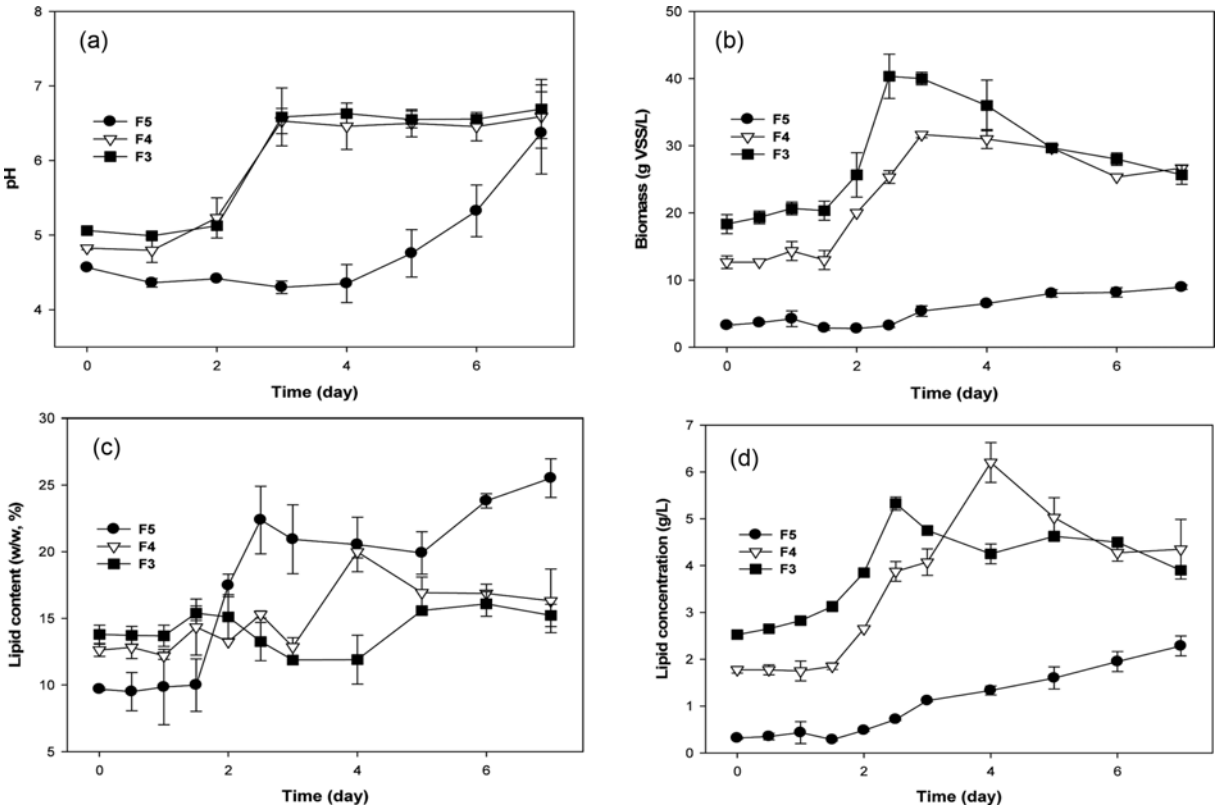

Fig. 3. Reaction profiles during lipid enhancement. (a) pH; (b) biomass concentration; (c) lipid content; (d) lipid concentration.

starkeyi is significantly inhibited when C : N ratio is high [21] because nutrients are diverted to lipid accumulation rather than to cell growth.

Time trends in total lipid contents which are complementary to those of biomass are shown in Fig. 3(c). Lipid content remained relatively constant for the first 1.5 days then increased, more quickly in F5 than in F4 and F3. The rates differed because lipid production was increased at high C : N ratio. In F5, neither biomass (Fig. 3(b)) nor lipid concentration (Fig. 3(d)) increased before 1.5 days, but thereafter biomass concentration started to increase more slowly than lipid concentration; hence total lipid content increased. This phenomenon continued until the end of lipid enhancement processes and caused a gradual increase in the lipid content in F5.

In F4, the proportional lipid content did not change much (Fig. 3(c)), because biomass concentration increased as lipid concentration increased [10,24]. This is a common phenomenon when C : N ratio is low. The total lipid concentration increased from 1.5 to 4 days until the carbon sources were mostly consumed, but lipid content remained constant until 3 days of reaction. The nitrogen was almost completely consumed by 3 days (Fig. 4), but carbon sources were still available (Fig. 2). Because of a momentary high C : N ratio, microorganism in the culture may synthesize large amount of lipids. Hence, lipid content stayed constantly from 0 to 3 days, then increased from 3 days to 4 days. After 4 days, both lipid content and concentration declined because carbon sources became limiting.

The result in F3 was similar to that in F4. All measured carbon sources were completely consumed within 3 days, which accords well with the ammonium concentration profile in Fig. 4. Lipid concentration was highest at 2.5 days, then decreased because the bacteria began to use their stored lipids as carbon sources due to the fact that external carbon availability had become limiting [22].

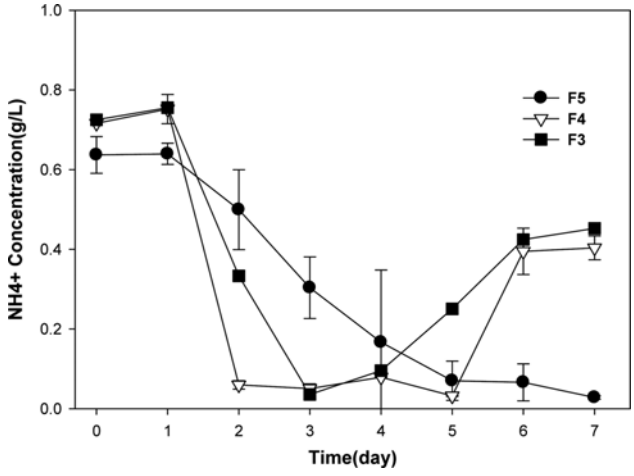

Fig. 4. Profile of ammonium-N during lipid enhancement.

High C : N ratio seems to provide a good environment for lipid accumulation but not for cell growth. Although gravimetric lipid content did not increase in F4 and F3, volumetric lipid yields increased much more than in F5. Thus, it was inferred that to maximize biodiesel production, cell growth may be more important than lipid accumulation.

Table 3. Composition of lipid extract from each fermentation conditions (average±standard deviation)

Parameter	F : S (5 : 0)		F : S (4 : 1)		F : S (3 : 2)	
	Raw	Enhanced	Raw	Enhanced	Raw	Enhanced
Initial biomass concentration (g VSS/L)	3.27±0.31		12.67±0.66		18.33±1.00	
Bligh & Dyer extraction yield, wt% of dry biomass	9.70±0.09	25.52±1.02	14.07±0.62	19.99±0.32	13.80±0.40	15.39±0.38
TAG wt% of extract	0.28±0.02	25.32±0.3	0.79±0.2	20.50±0.01	1.31±0.25	13.25±0.05
FFA, DAG, MAG wt% of extract	24.87±1.03	11.60±0.24	21.76±0.19	1.19±0.02	18.91±0.84	2.38±0.81
WE and SE wt% of extract	10.62±0.38	5.20±0.75	9.32±0.46	2.90±0.1	7.91±1.01	3.19±1.38
PL wt% of extract	0.81±0.03	0.52±0.18	1.78±0.6	0.83±0.72	2.79±0.2	1.81±0.35
FAME (from TAG), wt% of extract	0.26±0.03	25.30±0.5	0.80±0.2	20.20±0.01	1.30±0.1	13.25±0.2
FAME (from FFA, DAG, MAG), wt% of extract	15.59±0.15	8.59±0.06	13.6±0.09	9.24±0.2	9.55±0.36	5.47±1.08
FAMEs (from WE and SE), wt% of extract	4.41±0.13	2.21±0.07	3.26±0.31	1.00±0.06	2.40±0.03	1.66±0.19
FAMEs (from PL), wt% of extract	4.70±0.05	1.31±0.24	6.70±0.38	1.33±0.02	5.72±0.07	1.53±0.02
Total FAME yield, wt% of extract	24.96±0.19	37.41±0.49	24.36±0.62	31.77±0.18	18.97±0.24	21.91±0.73
Total FAME yield, wt% of biomass	4.85±0.28	7.44±0.07	2.55±0.2	5.37±0.04	2.32±0.11	3.27±0.07
Total FAME concentration (g/L)	0.17±0.01	0.56±0.01	0.32±0.01	1.49±0.01	0.42±0.01	1.15±0.01

2. Unsaponifiable Lipids

Concentrations of unsaponifiable wax esters (WEs) and steryl esters (SEs) decreased after lipid enhancement in all reaction conditions (Table 3). WEs in the sludge usually originate from detergent fatty alcohols; these fatty alcohols are bioavailable and microorganisms usually use them as resources in synthesis of WEs [23]. WEs and SEs are anthropogenic, and are mainly found in human excrement. The input of these contributions was limited during the enhancement processes, so total concentrations of these unsaponifiable lipids were decreased by the enhancement process. Microorganisms consume these lipids to their growth instead of consuming extra carbon sources in the medium [24]. These two reasons might explain the decrease of WEs and SEs after enhancement process.

3. Saponifiable Lipids

3-1. Raw Culture

The composition of saponifiable lipids was also affected by the enhancement process (Table 3). In the raw FWW (F5) saponifiable lipids were mainly FFAs, WEs and SEs; TAGs, DAGs, MAGs, PLs were relatively uncommon due to the low initial concentration of biomass. Hence, the portion of FAMEs produced by transesterification from TAGs was also very small (0.26 wt% from TAGs). In F4 and F3, addition of SS caused slight increase in the content of TAGs and PLs (0.79, 1.78 to 1.31, 2.79 wt%, respectively) because addition of SS increased the initial concentration of biomass. As a result, total FAMEs obtained from F5 (0.17 g/L) were lower than those from F4 (0.32 g/L) and F3 (0.42 g/L).

3-2. Increase in FAME Concentration

TAG content increased dramatically after the enhancement process in all reaction conditions (Table 3). TAGs are the most valuable feedstock for biodiesel production because they have high conversion potential to biodiesel. The quality of biodiesel is represented as the inverse of the free fatty acid content [25]; in all treatments, the concentration of TAGs significantly increased after the enhancement process, and the concentration of FFAs decreased (Table 3); these trends mean that the quality of the biodiesel increased. Due to the increase in the concentration of TAGs, the proportion of FAMEs from TAGs (wt% of extract) increased significantly from 0.26 to 25.30 in F5, from 0.80 to 20.20 in F4, and from 1.30 to 13.25 in F3. The yields of total FAMEs (wt% of extract) produced from lipid extract increased from 24.96 to 37.41 in F5, from 24.36 to 21,91 in F4 and from 18.97 to 21.91 in F3. The yields of total FAMEs (wt% of biomass) increased from 4.85 to 7.44 in F5, from 2.55 to 5.37 in F4 and from 2.32 to 3.27 in F3. Although the rate of increase seemed higher in F5 than in F4 and F3, the actual concentration of total FAMEs after enhancement process was much higher in F4 and F3 than in F5.

4. Fatty Acid Composition

The fatty acids produced from each cultures were mainly palmitic acid (C16:0), palmitoleic acid (C16:1), stearic acid (C18:0), oleic acid (C18:1) and linoleic acid (C18:2) (Fig. 5(a)); C8:0, C10:0, C20:0, C20:1, C20:2 and C24:0 were also present. Maximum FAMEs yield occurred at 2.5 d in F3 (Fig. 5(b)) and at 4 d in F4 (Fig. 5(c)). In F5, dominant fatty acids were saturated fatty acids (C16:0, C18:0) and changed to unsaturated fatty acids, mainly into C16:1 after the reaction. No significant changes were observed in the compositions of other fatty acids. In F4, the content of 18 : 1 increased significantly from 24.63% (initial) to 38.44% (4 d) and became the dominant fatty acyl residue. This is an important result in terms of quality of biodiesel typified by ignition quality (cetane number) and cold flow, which both improve with the content of oleic acid, which is an 18 : 1 fatty acid [14]. In F3, the proportion of C18:1 increased from 21.36% to 35.29% within 2.5 d, but decreased thereafter to 33.65% at 4 d.

The compositions of FAMEs proposed in this study were slightly different that observed in a previous study [24]. The difference can be attributed plausibly to the differences of sludge and media used, and is undesirable if the objective is to produce a consistent biodiesel mixture from SS. However, the compositions of FAMEs were homogenized after the enhancement process in this study; i.e., cultures that used the same medium, FWW, as a carbon source, produced a consistent bend of fatty acids.

CONCLUSIONS

Lipid enhancement using two organic wastes, food wastewater

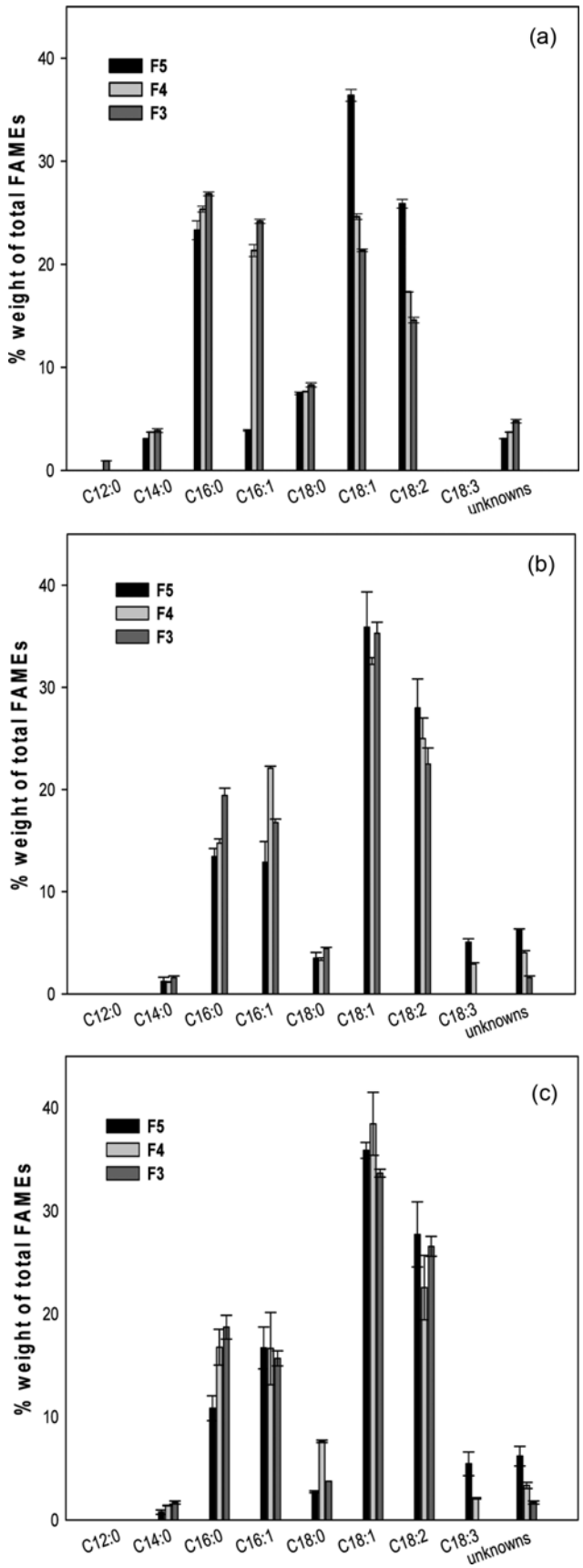

Fig. 5. The composition of fatty acids (a) initial; (b) at 2.5 d; (c) at 4 d.

(FWW) and sewage sludge (SS), was used to increase the amount of total lipids originating from saponifiable lipids. After lipid enhancement process, TAG content production was increased drastically compared to raw SS. The experimental results showed that increase in lipid content was mostly from TAGs, and the proportion of FAMEs from TAGs (wt% of extract) increased significantly from 0.26 to 25.30 in F5, from 0.80 to 20.20 in F4, and from 1.30 to 13.25 in F3. Particularly, C16:0 fatty acid was mostly converted to C18:1 fatty acid and became the dominant fatty acyl residue in F4. Taken together, this study shows the feasibility of biodiesel production using organic wastes (SS and FWW) which are troublesome to treat.

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