



Optimization and microbial community analysis for production of biogas from solid waste residues of palm oil mill industry by solid-state anaerobic digestion



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HIGHLIGHTS

- Solid-state anaerobic digestion (SS-AD) of oil palm biomass was studied.
- Optimal conditions for methane production was 16% TS, C:N of 30:1 and F:I of 2:1.
- The highest methane production of 77.8 m³ CH₄ ton⁻¹ biomass was achieved from EFB.
- Bacteria community in SS-AD was dominated by *Ruminococcus* sp. and *Clostridium* sp.
- Archaea community in SS-AD was dominated by *Methanoculleus* sp.

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ABSTRACT

This study investigated the improvement of biogas production from solid-state anaerobic digestion (SS-AD) of oil palm biomass by optimizing of total solids (TS) contents, feedstock to inoculum (F:I) ratios and carbon to nitrogen (C:N) ratios. Highest methane yield from EFB, OPF and OPT of 358, 280 and 324 m³ CH₄ ton⁻¹ VS, respectively, was achieved at TS content of 16%, C:N ratio of 30:1 and F:I ratio of 2:1. The main contribution to methane from biomass was the degradation of cellulose and hemicellulose. The highest methane production of 72 m³ CH₄ ton⁻¹ biomass was achieved from EFB. Bacteria community structure in SS-AD process of oil palm biomass was dominated by *Ruminococcus* sp. and *Clostridium* sp., while archaea community was dominated by *Methanoculleus* sp. Oil palm biomass has great potential for methane production via SS-AD.

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1. Introduction

Solid-state anaerobic digestion (SS-AD) for lignocellulosic materials has gained increasing attention in recent years. SS-AD could handles feedstocks with high total solids content (>15% TS) (Motte et al., 2013). It also has lower energy demand for heating, smaller reactor volume, high volumetric biogas productivity, more effectively at high organic loading rates and less wastewater generation comparison to liquid anaerobic digestion (L-AD). The digestate of SS-AD can be further used as fertilizer due to lower

moisture content which is easy for transportation and land applications. A previous study also showed that biogas yield from SS-AD was comparable to that produced from L-AD for some lignocellulosic feedstocks such as switchgrass and corn stover (Brown et al., 2012). Brown et al. (2012) reported that the volumetric productivity of SS-AD system was 2 to 7 fold greater than L-AD. Liew et al. (2012) reported that high methane yield from corn stover, wheat straw, yard waste and leaves of 81.2, 66.9, 40.8 and 55.4 m³ CH₄ ton⁻¹ VS via SS-AD process. Biogas gas achieved from SS-AD could be potentially utilized for electricity production and convert to value added transportation fuels (Yan et al., 2015). This can proof the economic feasibility of SS-AD of lignocellulosic materials.

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Lignocellulosic biomass is an abundantly available as raw material from agricultural solid wastes. Palm oil industry generated huge amount of lignocellulosic biomass from oil extraction plant and plantation area (Mohammed et al., 2011). The process to extracting oil from palm fruit generated lignocellulosic biomass such as empty fruit bunch (EFB) (23%), mesocarp fiber (12%) decanter cake (6%) and shell (5%) for every ton of fresh fruit bunches (Najafpour et al., 2005). Plantation area also generated oil palm trunk (OPT) and oil palm fronds (OPF) as a lignocellulosic biomass (Mohammed et al., 2011). Lignocellulosic biomass from palm oil industry mainly composed of cellulose (30–40%), hemicelluloses (25–35%), and lignin (15–25%) (Mohammed et al., 2011; Kong et al., 2014). Since high moisture content (65–75%) in lignocellulosic biomass from palm oil industry such as EFB, OPF and OPT, it is therefore not technical economic to be used directly as fuel for steam production (Foo and Hameed, 2009). Combustion is feasible for biomass having moisture content less than 50%, otherwise the energy efficiency is low (10–30%) and the pollutant emissions are the by-products of combustion (Mohammed et al., 2011). Currently, a variety of oil palm biomass including EFB and decanter cake has been examined for biogas production. O-Thong et al. (2012) reported that the maximum methane potential of EFB was $202 \text{ m}^3 \text{ CH}_4 \text{ ton}^{-1}$ VS corresponding to $79.1 \text{ m}^3 \text{ CH}_4 \text{ ton}^{-1}$ EFB under the L-AD. Previous study usually employed L-AD to digest lignocellulosic biomass, but high solid content of oil palm biomass has favored researcher to consider SS-AD. Chaikitkaew et al. (2015) used SS-AD to digest EFB that contained 25% TS and achieved maximum methane production of $144 \text{ m}^3 \text{ CH}_4 \text{ ton}^{-1}$ VS EFB. SS-AD of solid wastes from palm oil industry in a single treatment step would simplify the technical and economical requirements for the transformation of solid wastes into biogas and compost (Yang et al., 2015).

SS-AD of lignocellulosic biomass has recently been studied by several researchers, but methane yields and total solids (TS) degradation were generally low and potential instability (Liew et al., 2012; Yan et al., 2015; Yang et al., 2015). Low methane yield could be caused by the recalcitrance of lignocellulosic biomass or retarded mass transfer in SS-AD, while the imbalance of nutrients and accumulation of digestion intermediates, such as ammonia and volatile fatty acids (VFAs), may lead to system instability. Several process parameters can be optimized to operate SS-AD such as feedstock-to-inoculum ratio (F:I), initial total solids concentration (TS), carbon-to-nitrogen ratio (C:N) and type of feedstocks (Yan et al., 2015). Additionally, the F:I ratio and C:N ratio are two among the most important factors for onset of a balanced microbial population. Li et al. (2011) found that when operating at high F:I ratio (4.58 and 7.41) a significant decreasing amount of biogas was observed in the first few days and stop at day 10. High F:I ratio resulted high acidity in the digesters probably due to the over production of VFAs and methanogenic archaea were subsequently inhibited (Alzate et al., 2012). Furthermore, previous report showed methane yield in SS-AD decreased by about 60% when TS increased from 20% to 30% (Yang et al., 2015). Excessive TS loading could result in rapid hydrolysis as well as overproduction of VFAs and thus reduce the process stability of SS-AD digesters (Li et al., 2011). Nevertheless, proper TS loading were also necessary for SS-AD process, as TS overloading could delay the start-up of the digestion and thus affect the performance of the SS-AD process (Yan et al., 2015). Oil palm biomass generally has a C:N ratio higher than 50:1, which is improper C:N ratio for anaerobic digestion (Mohammed et al., 2011). The effect of F:I ratio, C:N ratio and TS on SS-AD performance for biogas production from oil palm biomass has not been investigated.

The overall objective of this study was to evaluate the feasibility of utilization of oil palm biomass for biogas production via SS-AD process. Effect of F:I ratio, TS content and C:N ratio on methane

production of solid wastes from palm oil industry via SS-AD was investigated. The stability of SS-AD process was evaluated by methane production, biodegradation and microbial community to better understand the SS-AD process.

2. Materials and methods

2.1. Feedstocks and inoculums

Oil palm biomass (EFB, OPF, OPT) and mesophilic methane production sludge inoculums were collected from United Palm Oil Industry Public Company Limited, Krabi province. Oil palm biomass was oven dried at 95°C in a convection oven until moisture content less than 10% moisture. Dry biomass was ground with a hammer mill equipped with an 5 mm screen and stored in airtight containers prior to use. Oil palm biomass was analyzed for total solids, volatile solids, total Kjeldahl nitrogen (TKN), C:N ratio, cellulose, hemicellulose, lignin and lipid content. The mesophilic methane production sludge was settled and decanted supernatant. The concentrated sludge was enriched with palm oil mill effluent adjusting pH with 0.025% (w/v) sodium hydrogen carbonate. Prior to use the sludge inoculums was incubated for 1 weeks to activate microorganism activities. The enriched sludge having a volatile solids (VS) concentration higher than 8.0 g L^{-1} was used in the SS-AD experiments. The composition of inoculums had TS, VS and C:N ratio of 11.45%, 8.3% and 2.5:1, respectively.

2.2. Effect of F:I ratios, TS and C:N ratios for methane production on SS-AD

The effect of F:I ratios (2:1, 3:1, 4:1, 5:1 base on dry VS) on the performance of methane production via SS-AD were investigated at a fixed initial TS content of 16% in batch assays under mesophilic condition (35°C). The effect of TS and C:N ratios on biogas production via SS-AD was investigated with optimum F:I ratio. Total solids concentrations of 16%, 25% and 35% with different C:N ratios of 20:1, 30:1 and 40:1 were designed to evaluate their effect on the biogas production via SS-AD. The urea was added to adjust the C:N ratios. Biogas potential was determined in batch assays under mesophilic conditions as described previously by Angelidaki et al. (2011). The inoculums and oil palm biomass feedstocks were mixed by a hand-mixer and flushed with nitrogen gas to generate anaerobic conditions. All tests were conducted in 0.5 L serum bottles with a working volume of 0.3 L. Afterwards the serum bottles were closed with butyl stoppers and placed in a 35°C incubator for 45–70 days and duplicate serum bottles were run for each condition. Biogas volume and composition were daily monitored by displacement method and gas chromatograph.

2.3. Analytical methods

Chemical and physical composition of EFB, OPF, OPT, inoculums and digestate after SS-AD were analyzed for TS, VS, pH, TKN total volatile fatty acid (TVFA) and alkalinity according to Standard Methods for the Examination of Water and Wastewater (APHA, 1998). The contents of lignin, cellulose, and hemicelluloses were determined according to the procedures proposed by Seot et al. (1991). The daily biogas production for each experiment was recorded using the water displacement method (Yan et al., 2015). The biogas composition was measured by gas chromatography equipped with thermal conductivity detectors (TCD). Methane, carbon dioxide, hydrogen and nitrogen were analyzed by GC-TCD fitted with 3.3 ft stainless steel column packed with Shin Carbon (60/80 mesh). Argon was used as a carrier gas at a flow rate of 14 ml min^{-1} . The temperatures of the injection port, oven and

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