



Batch Mode Biomethane Production from Anaerobic Digestion of Palm Oil Mill Effluent (POME) with Cassava Peels and Cabbage Waste

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

This study aimed to investigate anaerobic co-digestion of palm oil mill effluent (POME) with cassava peels (CP) and cabbage waste (CW) for biomethane production. The anaerobic digestion (AD) in 10L capacity bioreactors loaded separately with three different ratios (3L/ 520g, 3L/ 600g and 3L/ 680g) of POME/CP, POME/CW and POME only (control) was operated under ambient temperature (25 - 36^o C) and pH range of 6.5 - 8.5 for 45 days. Standard methods were adopted in the characterization of the bioreactor feeds and microbiological study. The biomethane content of the biogas was determined using Gas Chromatography (GC). The results showed the presence of *Escherichia coli*, *Staphylococcus*, *Pseudomonas sp*, *Bacillus sp* *Salmonella sp* among others. Fungal isolates identified include *Saccharomyces*, *Aspergillus*, *Rhizopus*, *Penicillium*, and *Geotrichum* species. The mean cumulative biogas yield recorded in bioreactors charged with POME/ CP 520g, POME/ CP 600g, POME/ CP 680g and POME/CW 520g, POME/CW 600g, POME/CW 680g were 7.08, 5.18, 9.06, 9.13, 9.28 and 8.33 dm³, respectively, whereas POME alone (control) was 4.64 dm³. The best performance in biogas yield was exhibited by POME/CW 600g (9.28dm³), and the highest percentage biomethane content (68.80%). Analysis of variance

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(ANOVA) revealed a significant difference ($P \leq 0.05$) in biogas yield in all the treatments compared to control (POME alone) except in POME/CP 3L: 520g and POME/CP 3L: 600g. The results have shown that biogas production and biomethane content could be efficiently improved via co-digestion process, depending on the substrates used as feedstock.

Keywords: POME; kitchen wastes; co-digestion; biomethane; gas chromatography.

1. INTRODUCTION

World population increase has led to the depletion of fossil fuel which is the major source of energy supply. Greenpeace Southeast Asia and the Center for Research on Energy and Clean Air (CREA) in their recent research observed that air pollution from burning of fossil fuels caused the death so many people yearly in the world as well as global economic losses as 230,000 deaths were recorded in United States alone [1]. Also the use of fossil fuel and wastes generated from industrial, commercial and agricultural activities without proper management practices cause climate change, environmental pollution and health hazards due to greenhouse gas emission [2,3]. The recent policy now is to minimize fossil fuel use which is non - renewable and shift to clean renewable energy, as energy has always been a driving force for a successful farming system [4].

Palm oil tree (*Elaeis guineensis*) is one of the crops in the tropics and the most productive oil producing plant worldwide as one hectare of it produces between 10 and 35 tons of fresh fruit bunch (FFB) per year [5,6,7]. During the production of palm oil from the fresh fruit, large quantities of wastewater known as palm oil mill effluent (POME) is generated [8]. POME is made up of carbohydrate and oil with very high values of chemical oxygen demand (COD) and biochemical oxygen demand (BOD) of about 80,000 mg/l [9]. It causes water pollution when channeled into local rivers or lakes without treatment, also causes environmental pollution if not properly disposed.

Cassava (*Manihot esculenta*) is another important crop produced in Nigeria and many other sub-Saharan African countries. Its carbohydrate content is higher than other staple crops and can be used in place of maize as a source of energy in the production of animal feeds [10]. In Africa, cassava tuber can be processed traditionally into products such as "garri" and "fufu" which are fermented products or flour, and about 42.5 million metric tons production of cassava is processed for garri in

Nigeria alone per annum [11]. During the processing of cassava, the tubers are usually peeled to get rid of two outer coverings and these peels which are regarded as wastes are generated in heaps which pose a disposal problem and may even be more problematic in future, as there is currently increase in industrial production of cassava products in Nigeria [12]. Small proportion of this waste is used in the production of animal feed. This is because of the harmful effect of the hydrocyanic acid which is one of the contents of cassava, on the growth and development of non - ruminant animals [13]. Piles of the rest of the wastes generated are usually disposed of indiscriminately or allowed to rot in the environment with offensive odour emanating from it as a result of microbial degradation as the demand increases causing a lot of environmental pollution and health hazards [14,15].

Cabbage (*Brassica oleracea*) is a vegetable crop, leafy green and biennial plant with high nutritional value. The world production of cabbage and other brassica for 2014 was reported to be about 71.8 million metric tons with approximately 45% of the 60 million tons deposited as waste during harvest and when these wastes accumulate without proper disposal, they cause environmental pollution and health hazards [11].

The issue of disposal of all these agricultural wastes and environmental problems that emanate from it, as well as the high cost of fossil fuel makes anaerobic digestion of waste for biogas production a better option because it is a low cost, renewable and sustainable source of alternative energy [16].

The use of fossil fuel and wood fuel is causing global warming in the world today but the use of biogas doesn't, neither does it have any effect on the atmosphere [17,18]. Also biogas technology being integrated into agriculture will improve the income levels of farmers [19]. Biogas technology as an eco-friendly technology can be of help to both animal husbandry and crop farmers. They are easy to manage, eco-friendly and generates

an end-product that is used as soil conditioner for improving soil fertility for more yield [20]. The complex biochemical reaction of anaerobic digestion for biogas production is been affected by so many factors like temperature, loading rate, C: N ratio, bioreactor design, inoculums, pretreatment methods etc. [21-24]. Also the proximate composition of the feedstock has a role to play in the quality of biogas and its cumulative yield, therefore the use of one agricultural waste in anaerobic digestion may not give a better biogas yield, hence co-digestion of two or more feedstocks is required to improve biogas yield [25]. The research work was then carried out to evaluate the anaerobic co-digestion of palm oil mill effluent with cassava peels and cabbage waste for biomethane production.

2. MATERIALS AND METHODS

The substrates used in this study were Palm oil mill effluent (POME); cassava peels (CP) and cabbage waste (CW). The POME was collected from an oil mill industry at Umuagwo in Ohaji-Egbema LGA, Imo State. The collected sample was kept for some hours to sediment and then filtered to remove the debris.

The cassava peels (CP) was collected from a cassava processing plant for 'garri' production and cabbage waste (CW) from a fruit market in Imo state, Nigeria. Unwanted materials were removed from the samples and sun-dried to a moisture content of 11.48% and 11.15%, respectively, milled to reduce the size of the particles, it was then sieved before use. The source of the inoculum used to stabilize the wastes was cow rumen liquor. It was processed by filtering in cheesecloth and stored air-tight to maintain the required anoxic condition so that the viability of the microorganisms (methanogens) for methane production could be sustained.

2.1 Proximate Analysis of the Bioreactors Feeds

Standard methods were adopted for the proximate analysis of Palm oil mill effluent (POME), Cassava peels (CP) and Cabbage waste (CW) to determine the Total Solid (TS), Volatile Solid (VS), Carbon to Nitrogen (C: N) ratio, Ash content, Moisture content etc. [26].

2.2 Design of Experiment

The experiment was designed and set-up as described by Oporum et al., [27] with some

modifications. Batch bioreactor systems of 10L capacity were used for the anaerobic digestion. Each bioreactor was installed with a thermometer for measuring the temperature and an outlet for gas passing to the gas collecting system. The hose from the bioreactor was connected to a 13L transparent bucket and 3L transparent bucket inverted in it which served as a gas collector. The bioreactors were separately loaded with POME/CP and POME/CW: 3L/ 520g, 600g and 680g, respectively and 3L of POME as control.

Cow rumen liquor was used as the inoculum which provided the source of methanogens. Digestion of the substrates was carried out at room temperature which was between 25°C-36°C. Each bioreactor was manually agitated daily to prevent sedimentation and for evenly distribution of substrates, enzymes and microorganisms and also to promote heat transfer and facilitates the release of produced biogas from the bioreactor contents [28,29]. The biogas yield from each bioreactor was recorded by adopting the downward water displacement method and the mean values recorded on daily basis every 24hrs for 45days hydraulic retention time [27]. The pH was adjusted at the range of 6.8 to 8.0 with Sodium hydroxide (NaOH) and Hydrochloric acid (HCl).

2.3 Microbiological Analysis

The samples used for microbiological analysis were collected in sterile bottles from the outlet hose of the digester immediately after the digesters were set up. The bacterial and fungal species in the digesting slurry were determined by the use of various culture media using the spread plate technique as described by Bergey's Manual of Determinative Bacteriology [30]. Each prepared medium was inoculated after 10 fold serial dilutions. An aliquot (0.1ml) of 10³ dilution of the sample was inoculated by spread plate technique onto these media: Nutrient agar (NA), Eosin Methylene Blue agar (EMBA), MacConkey agar (MCA), Salmonella-Shigella agar (SSA) and Potato Dextrose agar (PDA) prepared according to manufacturers' instructions. Plates for bacterial cultures were incubated at 30°C for 48hr whereas PDA plates were incubated at room temperature for 72 - 96hr. The microbial growth subjected to microscopic examination, biochemical tests and Gram staining were also carried out as described by Cappucino and Cherman [31].

2.4 Biogas Analysis using Gas Chromatography (GC)

Flammable biogas was tested on daily basis, and Gas Chromatography (GC), GC-TCN (M910) with helium as a carrier gas at 5psi, and a flow rate of 20ml per minute was used ascertain the composition of the biogas.

2.5 Analysis of Data

The cumulative biogas yield in all the treatments and control were statistically analyzed using analysis of variance (ANOVA) implemented via IBM SPSS version 20.0.

3. RESULTS AND DISCUSSION

3.1 Characteristics of the Bioreactor Feeds

The characteristics of the POME, CP and CW (Table 1) were carried out to determine the available digestible nutrients in the feedstocks that can be accessed by the anaerobic microorganisms during the digestion process. The carbon to nitrogen (C/N) ratio of the POME, CP and CW are 10:1, 46:1 and 17:1, respectively. It was observed that cassava peels has a higher C: N ratio than POME and cabbage waste. It is therefore necessary to co-digest the substrates for nutrient balance and enhance biogas production.

3.2 Anaerobic Digestion Process and Biogas Production

Shown in Table 2 is the mean cumulative biogas yield from the various treatments. It was observed that co-digestion of POME/CP and POME/CW at the ratios of 3L: 520g, 3L: 600g and 3L: 680g had higher cumulative biogas yield of (7.08dm³), (5.18dm³), (9.055dm³) and (9.13dm³), (9.28dm³), (8.33dm³) respectively than POME alone with cumulative biogas yield of 4.64 dm³. This shows that the co-digestion of the substrates was able to improve the efficiency of biogas production.

The bioreactors loaded with POME/CP 3L: 520g, 3L: 600g and 3L: 680g ratios biogas productions started on second day. Flammable biogas production in 3L: 680g started on 11th day, 3L:

520g ratio on 13th day and 3L: 600g on day 14th. POME/CP 3L: 680g started biogas production on the 2nd day and flammable gas on the 11th day, the peak of gas production was observed on the 13th day having biogas yield of 1.89 dm³ and its cumulative biogas yield of 9.055dm³ (Table 2). In POME/CW 3L: 520g, 3L: 600g and 3L: 680g the production of biogas started on the 2nd. In 3L: 600g ratio flammable biogas production started on the 5th while 3L: 520g and 3L: 680g flammable biogas started on 15th day.

POME/CW 600g had the highest biogas yield as well as the percentage biomethane content. The biogas production started on the 2nd day, the peak was observed on the 11th day and it had biogas yield of 1.55dm³ and cumulative biogas yield of 9.28dm³. POME alone which was the control, started its biogas production on the 2nd day, flammable biogas production on the 4th day and the peak was observed on the 5th day as it had biogas yield of 1.21dm³ and cumulative biogas yield of 4.64dm³ which was recorded as one of the lowest biogas yield had in the research work.

3.3 Gas Chromatography Result

POME/ CW (3L: 600g) yielded the highest percentage biomethane (Table 3). The percentage biomethane content recorded in the study was 68.80% for POME/CW (3L: 600g), 65.28% for POME/CP (3L: 680g) and 56.53% for POME (control). The co-digestion of POME/CW (3L: 600g) proved to be the most suitable combination to enhance production of biogas and biomethane content as well.

3.4 Microbiological Analysis

The microbial species identified include *Bacillus*, *Saphylococcus* *Escherichia coli*, *Pseudomonas*, *Enterococcus*, *Enterobacter*, *Salmonella* and *Micrococcus* species. Fungi isolated were *Saccharomyces*, *Aspergillus*, *Rhizopus*, *Penicillum*, and *Geotrichum* species.

3.5 Statistical Analysis

Post-Hoc Duncan test was used to determine the means of maximum cumulative biogas yield and it was observed that there is significant difference between POME alone and all other treatments except POME/CP 3L: 520g and 3L: 600g.

Table 1. Proximate characteristics of the substrates

Parameters (%)	Substrates		
	POME	CP	CW
Fat content	8.23	3.94	3.83
Fibre content	-	11.07	13.26
Ash content	0.54	6.77	23.81
Crude Protein	3.25	6.778	14.76
Organic carbon	48.90	49.87	39.54
C:N Ratio	10.13	46	16.90
Nitrogen	5.12	1.084	2.361
Moisture content (MC)	86.08	11.48	11.15
Carbohydrate	1.90	-	-
Volatile Solids (VS)	13.39	81.75	65.04
Total Solids (TS)	13.93	88.52	88.85
pH	4.06	5.91	6.84

Table 2. Mean cumulative biogas yield (dm³)

Treatment	3L:520g	3L:600g	3L:680g
POME/CP	7.08	5.18	9.06
POME/CW	9.13	9.28	8.33
POME control	4.64	4.64	4.64

Table 3. Percentage biogas composition of bioreactors with the highest biomethane yield

Substrates (g)	Percentage Composition (%)		
	Methane (CH ₄)	Carbon-dioxide (CO ₂)	Carbon Monoxide (CO)
POME/CP 3L/680	65.28	38.80	3.22
POME/CW 3L/600	68.80	28.50	2.09
POME control	56.54	30.92	0.45

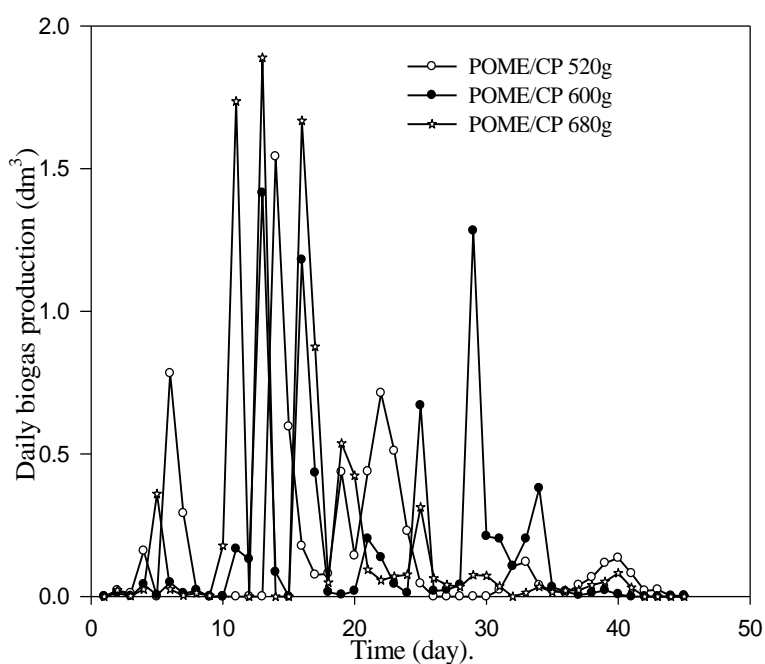


Fig. 1. Profile of daily biogas production from mixtures of POME/CP

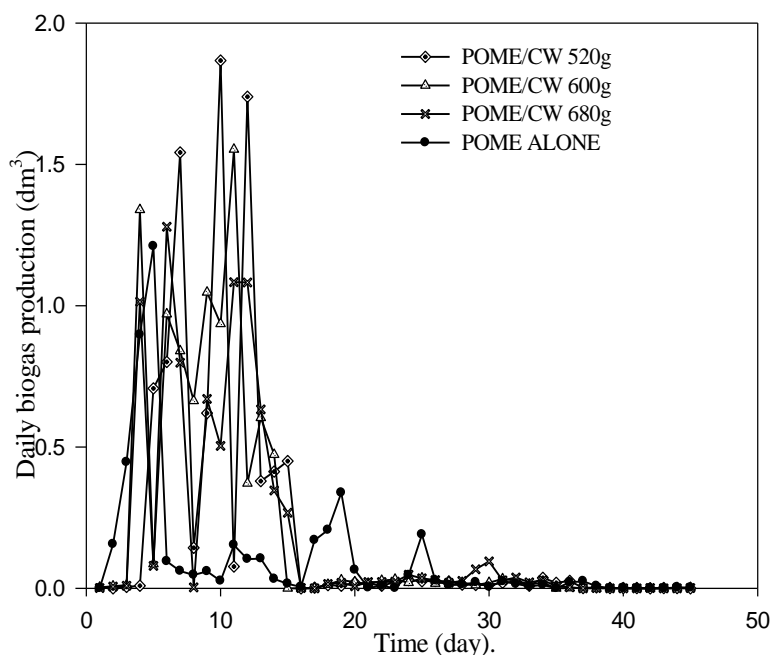


Fig. 2. Profile of daily biogas production from mixtures of POME/CW and POME alone

4. DISCUSSION

The characteristics of the substrates showed that cassava peel has a higher C: N ratio (46:1) than POME (10:1) and cabbage waste (17:1), hence it is necessary to co-digest it with other acceptable substrate to improve the Carbon: Nitrogen ratio. The C: N ratio of the POME (10.13) is in concurrent with that report of Adela et al., [32] with C/N ratio of 10.58%.

To enhance biogas production from feedstock, the carbon to Nitrogen ratio is one of the important factors that must be taken into consideration in co-digestion of substrates as it influences production process [33]. The methanogenic bacteria use up the nitrogen content of the substrate to meet their protein needs when the C: N ratio is very high resulting to imbalance in C: N ratio which causes low biogas production. On the other hand if the C: N ratio is low, excess nitrogen will be released during microbial metabolism and this piles up in the form ammonia which increases the pH value of the feedstock above 8.5 and resultant effect being the exertion of toxic substances on the methanogens causing low biogas yield [34].

The total solids (TS) and volatile solids content of POME has low total solids (TS) and volatile solids content of 13.93 and 13.39%, respectively

and this may be attributed to the low biogas yield while CP and CB are sufficiently high (88.52; 81.75 and 88.85; 65.04 %), indicating that both substrates are suitable for biogas production. The increase in biogas yield from Cabbage co-digested with POME could be due to the synergistic effect of the nutrient contents of the feedstocks. Esposito et al., [35] reported that there is synergistic effect on co-digestion of different organic substrates as it was observed that the degradation level of the mixture increased more than that of the single substrates. The total solid (TS) content of the cassava peels used is 88.52% and moisture content of 11.48% and this supports the report of Nkodi et al., [36]. The proximate composition of substrates has great influence on the quality of biogas and its yield [37]. Bolaji and Adebayo [38] reported that hydrolysis of the lignocellulosic constituents of crop residues which are wastes from plant materials make it difficult for microorganisms to digest them easily like animal wastes.

Co-digestion of substrates has to be adopted to efficiently enhance biogas yield because the quality of biogas /biomethane content and its cumulative yield highly depends on the characteristics of the feedstock [39,40]. Similarly in this study, biogas yield from co-digested substrates were higher than that of POME alone, this could be attributed to the low volatile solid

(TS) content of 13.39% and low C: N ratio of 10:1%. Improvement of the characteristics of the substrates and other factors of the bioreactor is required for optimum microbial activities in anaerobic digestion and effective biogas production [41,42].

POME/CP and POME/CW, 3L: 520g, 3L: 600g and 3L: 680g had higher cumulative biogas yield of (7.08 dm³), (5.18 dm³), (9.055dm³) and (9.13dm³), (9.28dm³), (8.33dm³) respectively than POME alone with cumulative biogas yield of 4.63dm³ as shown in Table 2. The study proves that co-digestion of the bioreactor feedstock was capable of enhancing biogas production. Previously effluents improved biogas yield as well as the biomethane content while mono-substrate digestion didn't [43]. Similar result was observed by Sawyerr et al., [25]. Ibrahim et al., [44] also reported that the methanogenic process is affected when POME is anaerobically digested alone and this, being the final stage of biogas production affects the yield.

Similar microorganisms isolated in the course of the study were also reported by Asikong et al., [45]. POME/CW (3L: 600g) had the highest percentage methane content (Table 3). The high percentage biomethane content were 68.80% for POME/CW (3L: 600g), 65.28% for POME/CP (3L: 680g), and 56.53% for POME alone (control) while the remaining percentages are: Carbon-dioxide (CO₂), Carbon-monoxide (CO), and trace elements namely; hydrogen (H₂), oxygen (O₂), nitrogen (N₂) and water (H₂O). Findings from this investigation have shown that the co-digestion of POME/CW (3L: 600g) has prospect in biomethane production. Biomethane production through the combination of substrates improved yield as the percentage increased from 56.53% to 65.28 and 68.80%. Similar result was observed by Aragwa et al., [46] who reported that co - digestion of organic kitchen waste and dairy manure improved biogas yields by 24 to 47% over the control.

5. CONCLUSION

This study has demonstrated that anaerobic co-digestion of palm oil mill effluent with cassava peels and cabbage is capable of significantly enhancing biogas yield. The best performance in biogas production was recorded in bioreactor charged with POME/CW 600g, followed by POME/CP 680g. The feedstock characteristics indicated their potentials as suitable substrates in biogas production. Co-digestion of palm oil mill

effluent with cassava peels and cabbage could be adopted in large scale production of biogas as this could help in the production of renewable energy, proper management of agro-wastes and mitigation of greenhouse gas emission which causes global warming.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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