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Citation: AIP Conference Proceedings **1868**, 020013 (2017); doi: 10.1063/1.4995099
View online: http://dx.doi.org/10.1063/1.4995099
View Table of Contents: http://aip.scitation.org/toc/apc/1868/1
Published by the American Institute of Physics

Effect of Dilution and Ash Supplement on the Bio-methane Potential of Palm Oil Mill Effluent (POME)

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Abstract. This study aimed to evaluate the bio-methane potential of POME at different dilutions (100, 80, 60, 40, and 20 percent of initial POME) and different pH dues to different levels of ash supplement. Five different amounts of ash were added to digesters (0, 2, 4, 6, and 8 grams of ash were added to 170 ml of POME respectively). The digesters were operated in batch anaerobic digestion systems at room temperature (28-30 °C) and the experiments were performed in duplicate manner. The results showed that POME without dilution gave highest cumulative biogas (950 ml). However, 80% dilution from original POME gave the highest methane yield (45.83 mL CH_4/ $gCOD_{added}$ or 103.13 mL CH_4/ $gCOD_{removed}$). Finally, the results of experiment 2, this adding ash into POME increased pH as well as enhanced the biogas production. It was found that adding ash at the ash:POME ratio of 2 g: 170 ml gave the highest both the cumulative biogas and methane yield (1,520 mL and 218.79 mL CH_4/ $gCOD_{removed}$ respectively). The addition of ash in the raw waste of POME gave the pH in the range of criteria and highest bio-methane potential. The modified Gompertz equation, Schnute as well as Monod kinetic models were used to compare the data from the experiments. It was found that the factors that affected included, the bio-methane production and the kinetic parameters (the maximum specific methane production rates (R_m ml/day) and the methane production potential (P, mL)), initial COD, nutrients, levels of dilution, and initial pH (by adding different level of ash). However, λ (lag phase period) was not affected by initial COD and other factors. While Monod kinetics provides valuable insight in explaining what could happen behind the systematic trends.

***Keywords*:** Anaerobic digestion, Bio-methane, POME, Modified Gompertz

INTRODUCTION

Palm oil industry is the biggest agro-industry in the southern Thailand. The industry produces very large amount of high-COD effluent (called palm oil mill effluent or, in short, POME). If not sufficiently utilized or treated, it will create unbearable, hazardous environmental problems thus requires appropriate and comprehensive management approach [1]. Fortunately, POME has become very valuable as an abundant source of biogas for electrical generation by anaerobic digestion. The digestion process has many advantages and is applicable for a wide range of material including agriculture and industrial wastes [2]. This process produces energy instead of consuming energy unlike the aerobic wastewater treatment counterpart [3]. Furthermore, besides being renewable, biogas energy has some advantages over wind energy and solar energy as it provides energy source uninterruptedly by day-night cycle. The anaerobic digestion, thus, is potentially a main source of clean and renewable energy which

The 4th International Conference on Research, Implementation, and Education of Mathematics and Science (4th ICRIEMS)
AIP Conf. Proc. 1868, 020013-1–020013-10; doi: 10.1063/1.4995099
Published by AIP Publishing. 978-0-7354-1548-5/$30.00

can significantly substitute conventional energy sources such as fossil, fuels, and oil. In the anaerobic digestion, organic waste is converted into biogas and other products by microorganisms through four steps: hydrolysis, acidogenesis, acetogenesis and methanogenesis [4, 5]. In normal anaerobic condition, biogas generated comprises of methane (CH_4), carbon dioxide (CO_2), and some trace of gasses in a variable amount [6]. Recently, many researchers have paid a lot attention to the use of the wastewater from agro-industrial for biogas production. In their investigations, biomethane potential (BMP) is now widely used to compare substrates for different sources and conditions for biogas production [7-11]. One of the problems of the wastewater from palm oil industry is its low pH value (pH ~ 4.7) and nutrient imbalance such as too high C/N ratio or incomplete range of mineral source [12]. Adding chemical to improve initial pH of the wastewater is normally not cost effective. In searching for cheaper alternatives to alkali chemicals, it is observed that in the same region there are many power plants which generate electricity by burning biomass (mainly residues from rubber plantation and oil palm residues) which produce large amount of solid waste in the form of ash to be disposed. Instead of disposing this ash by dumping or use as soil improvement, it may be used to enhance the anaerobic digestion of agro-industrial wastewater such as POME. Thus, the scope of this study is to improve the biogas production by adding the ash waste which will increase the POME pH value and could also provide some minerals, thus promoting the microbial growth and enhancing the biogas production. This could result in cost saving, more profit by increasing biogas productivity as well as better satisfying the holistic waste management for both POME and the waste from biomass power plants.

MATERIAL AND METHOD

Materials

The wastewater sample was collected from a palm oil industry and the granular sludge or inoculum was collected from the up-flow anaerobic sludge blanket (UASB) reactor of the seafood industry. The physical characteristic of granule was irregular spheres having size range 0.7 – 1.0 mm. The volatile suspended solid of the granule sludge was 9,278 mg/l. Ash was collected from the biomass power plants in Yala province. The characteristic of wastewater is shown in Table 1. The wastewater samples were kept at 0-4 °C until used in the experiment.

TABLE 1. Basic parameter of palm oil mill effluent (POME)

Parameter	pH	COD(g/l)	TKN(mg/l)	TP(mg/l)	TS(g/l)	VS(g/l)	SS(g/l)	VSS(g/l)
Value	4.2	88	984	249	81	62.2	47	41

Experimental set- up

The anaerobic digesters having a total volume of 300 ml. and a working volume of 200 ml. was used in all experiments. The BMP test was conducted using the method of Owen *et al.* (1979) [13]. Initial pH for all reactors was adjusted to 6.8-7.2 by the addition of NaOH 1 N. in experiment 1 (study effect of dilution). The digesters were sealed with the rubber plug and cover with aluminum cap. The experiment was conducted at the room temperature (28-30 °C) Biogas production was measured daily by water displacement method as used by other authors [14-15]. The methane content was measured using Gas Chromatograph (GC-8A Shimadzu). The experimental setup is shown in Figure 1.

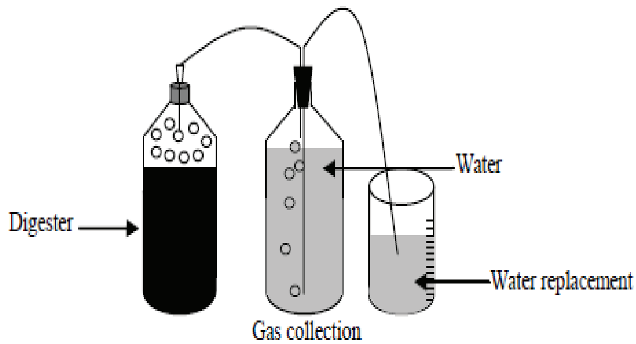

FIGURE 1. Schematic view of the experimental set-up

Experimental design

The All experiments were operated in batch mode in duplicate manner. The granules, 15% by volume, were used as inocula of methanogenic bacteria. The experimental designs for experiment 1 (effect of dilution) and experiment 2 (effect of pH) is shown in Table 2. The experiment 1 was aimed to study the effect of substrate concentration on the biogas production. The different amount of ash added into POME in experiment 2 represented the different pH and the effect of ash supplement on the biogas production.

Chemical analysis

In all experiments, we analyzed pH, chemical oxygen demand (COD), total Kjeldahl nitrogen (TKN), total phosphorus (TP), total solids (TS), volatile solids (VS), suspended solids (SS), volatile suspended solids (VSS), alkalinity and volatile fatty acids (VFA). All analytical procedures are performed in accordance with standard methods for the examination of water and wastewater [16].

TABLE 2. Experimental design for experiment 1 (study effect of dilution) and experiment 2 (study effect of pH)

Digester	POME (ml)	Granules (ml)	Total working volume (ml)	Experiment 1 POME (%)	Experiment 2 ash (g)
1	170	30	200	100	0
2	170	30	200	80	2
3	170	30	200	60	4
4	170	30	200	40	6
5	170	30	200	20	8

Kinetic model of biogas production

One of the most widely-used semi-empirical models for kinetic study the methane production is the modified Gompertz equation as shown in Eq. (1) [16-17]

$$P = P_{\infty} \cdot \exp\left\{-\exp\left[\frac{R_m \cdot e}{P_{\infty}}(\lambda - t) + 1\right]\right\} \tag{1}$$

where P is Cumulative methane production (ml), P_∞ is methane production potential (ml), R_m is the maximum specific methane production rates (ml/d), λ is lag phase period or minimum time to produce biogas (days) and e is mathematical constant (2.718282). This is equivalent to the original form of Gompertz equation.

$$P = P_\infty \exp\left(\frac{-r_0}{\alpha}\exp(-\alpha t)\right) \quad (2)$$

where r_0 and α are parameter in Gompertz which directly related to R_m and λ in Eq. (1)

The Schnute model is actually ageneralize from a class of growth model. Another, more generalized time-derivative Gompertz extension, the Schnute model which have the following time derivative of the specific growth rate and the modified form as shown in the Eg.(3) [17-18].

$$P = P_\infty \exp\left(\frac{-\alpha t}{\beta}\right)\left(\exp(\alpha t) - \frac{\beta r_0}{\alpha + \beta r_0}\right)^{1/\beta} \quad (3)$$

where P is biogas generated, r is the specific biogas production rate, and α, β are Schnute parameters respectively.

A classical way of describing growth and product formation kinetics is due to Monod (1949) [19].

$$\frac{dx'}{dt} = \mu X = \frac{\mu_m S}{K_s + S} \quad (4)$$

$$\frac{dx}{dt} = (\mu_m - k_d)X = \left(\frac{\mu_m S}{K_s + S} - K_d\right)X \quad (5)$$

$$\frac{dx}{dt} = \left(\frac{\mu_m (S_0 Y_{ps} - P)}{\left[(K_s + S_0)Y_{ps} - P\right]} - K_d\right)X \quad (6)$$

Where X' the total accumulated microbial growth assuming no death, μ_m, μ are maximum and general specific growth rate, K_d is specific death rate and K_s is the saturation constant

Using the definitions $Y_{ps} = \Delta P/\Delta S, Y_{x's} = \Delta X'/\Delta S, Y_{px'} = \Delta p/\Delta X = Y_{ps}/Y_{x's}$ and noting that $P'_0/Y_{ps} = X'_0/Y_{x's}$. The rate change of substrate and product can be write in Eg.7-8.

$$\frac{dS}{dt} = -\left(\frac{1}{Y_{x's}}\right)\frac{dX'}{dt} = -\left(\frac{\mu}{Y_{x's}}\right)X = -\left(\frac{1}{Y_{x's}}\right)\mu_m \frac{SX}{(K_s + S)} \quad (7)$$

$$\frac{dP}{dt} = -Y_{PS}\frac{dS}{dt} = Y_{PX'}\mu X = Y_{PX'}\frac{\mu_m S}{K_S + S}X = \frac{Y_{PS}}{Y_{X'S}}\frac{\mu_m (P_\infty - P)}{K_S Y_{PS} + P_\infty - P}X \quad (8)$$

Monod-type kinetics with constant cell density

$$\frac{dS}{dt} = -\frac{1}{Y_{XS}}\mu X_0 = -\frac{\mu_m X_0}{Y_{XS}}\frac{S}{K_S + S} = -K_1 \frac{S}{K_S + S} \text{ where } K_1 = \frac{\mu_m X_0}{Y_{XS}} = \frac{\mu_m P'_0}{Y_{PS}} \quad (9)$$

$$t = (1/K_1)\left[K_S \ln(S_0/S) + S_0 - S\right] = (1/K_1)\left[K_S \ln\left(P_\infty/(P_\infty - P)\right) + P/Y_{PS}\right] \quad (10)$$

Model with constant yield coefficients and no microbial death [18]

$$X = X_0 + Y_{X'S}(S_0 - S) = X_0 + (Y_{X'S}/Y_{PS})P,\ S = S_0 - \frac{P}{Y_{PS}}, S_0 = \frac{P_\infty}{Y_{PS}},\ Y_{X'S} = Y_{XS} \tag{11}$$

$$P = Y_{PS}(S_0 - S),\ C = \frac{X_0}{Y_{X'S}} + S_0 = \frac{X_0}{Y_{X'S}} + \frac{P_\infty}{Y_{PS}} = \frac{P'_0 + P_\infty}{Y_{PS}} = \frac{P'_\infty}{Y_{PS}} \tag{12}$$

$$t = \frac{1}{\mu_m}\left[\frac{K_S}{C}\ln\left(\frac{S_0(C-S)}{S(C-S_0)}\right) + \ln\left(\frac{C-S}{C-S_0}\right)\right] = \frac{1}{\mu_m}\left[\frac{K_s Y_{PS}}{P'_\infty}\ln\left(\frac{P_\infty}{P_\infty - P}\frac{P'_0 + P}{P'_0}\right) + \ln\left(\frac{P'_0 + P}{P'_0}\right)\right] \tag{13}$$

Constant biomass

$$t = \frac{Y_{XS}}{\mu_m X_0}\left[K_S \ln\left(\frac{S_0}{S}\right) + S_0 - S\right] = \frac{Y_{PS}}{\mu_m P'_0}\left[K_S \ln\left(\frac{P_\infty}{P_\infty - P}\right) + \frac{P}{Y_{PS}}\right] \tag{14}$$

RESULTS AND DISCUSSION

The result of this study in experiment 1 is shown in Table 3 summarizes for studying the effect of substrate concentration on the biogas production. At the end of experiment period, the cumulative biogas production from all digesters reached the value of 115-950 ml. and methane content was in the range of 38.38-73.75%. The cumulative methane at the end of the experiment was in the ranged from 59-622 ml. It was observed that the digester used 80% of wastewater from original POME gave the highest methane production potential (45.83 ml CH_4/ $gCOD_{added}$ or 103.13 ml CH_4/ $gCOD_{removed}$). The results showed that the diluted wastewater (Used 80% from POME original) gave better results than that from the original wastewater. This could be attributed to better nutrient balance (COD/N) and environmental condition suitable for the microorganism in anaerobic digestion and balance between substrate: microorganism. Thus, it was clearly indicated that there was a weak substrate inhibition at high COD which negatively affected the methane production. The results suggest that the initial COD has a strong effect on methane yield, P, and R_m but not for λ lag phase period or minimum time to produce biogas. The parameters obtained using Eq. (1) to describe the methane production are shown in Figure 2 and Table 3. It was found that most experiments showed essentially very short time lag phase before the microorganisms started to function fully. This implies that the microorganism in the system is viable and the substrates were readily biodegradable, thus causing biogas production to occur immediately after inoculation. In other words, the microorganisms did not need to adapt themselves to the new environment, because the granules or inocula in this study were collected from the methanogenic fermentation stage of the Upflow Anaerobic Sludge Blanket (UASB). These results are similar to that of Rincon *et al.* (2010) [8].

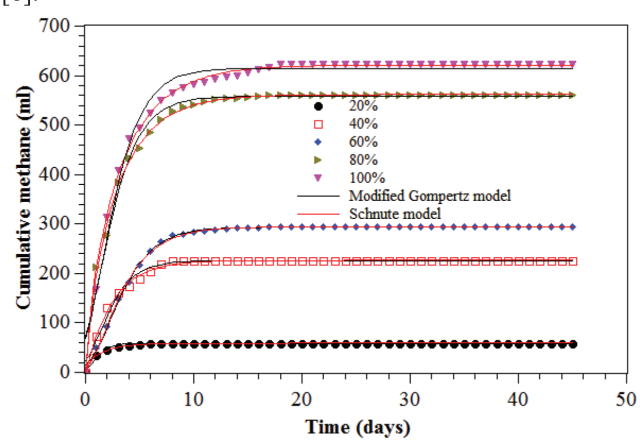

FIGURE 2. Comparison of experimental data and models

TABLE 3. Kinetic constant of methane production in experiment 1

Digester	POME (%)	COD (mg/l)	TKN (mg/l)	COD/N	Modified Gompertz model				Methane yield (ml CH_4/gCOD removed)
					P (ml)	R_m (ml/d)	λ (d)	R^2	
1	100	88,000	984	89.43	615.12	114.46	-0.41	0.9780	91.47
2	80	72,000	835	86.23	557.99	106.40	-0.55	0.9786	103.15
3	60	56,000	642	87.23	293.56	50.67	0.20	0.9980	72.06
4	40	36,000	511	70.45	225.71	50.69	-0.28	0.9843	55.39
5	20	16,000	348	45.98	58.68	26.59	-0.09	0.9732	43.38

The environmental condition, In case pH after digestion less than initial in all digesters, which would mean that all the substrate feed on to the process was not converted into methane. It is reasonable to assume that part of the particulate matter hydrolyzed and turned into volatile fatty acids (VFA), but not convert into methane. Our results were agreement with the study of Gomez *et al.* [20]. Regarding the effect of COD/N Sumardiono *et al*. [21] reported that the biogas production showed a satisfactory performance in the range of 71.4-85.7 of COD/N ratio. The results in all digester gave COD/N in the range of 45.98-89.43 thus, the decrease of percent dilution POME to decrease of the COD/N ratio. The results showed that the 80% from original POME dilution COD/N ratio was 86.23 gave the highest methane production potential.

In experiment 2, the results is shown in the Figure 3 and Table 4 selected 80% dilution of original POME from experiment 1 to study the effect of pH on the bio-methane potential by adding different amounts of ash from biomass power plant (0g, 2g, 4g, 6g, and 8g). The experiment design is shown in Table 2. At the end of the study, the cumulative biogas production from digester 2-5 was averaged and fell in the range of 924-1,520 ml which was higher than digester 1 (without ash supplement). Methane content was in the range of 62.0-73.75%. The cumulative methane at the end of the experiment were 561, 1116, 1025, 711, and 541 ml for 0g, 2g, 4g, 6g, and 8g respectively. In the initial phase of anaerobic digestion, the microorganism took up the readily consumable organic substance for growth and producing biogas simultaneously. Then the cumulative methane production was considerable delayed in methane production after 5 days because only slowly-digestible organic matter was left to be consumed. Methane production stopped after 10 days because of inhibition in wastewater or other unclear reasons.

The results suggested that the digester 2 (ash 2 g to POME 170 ml) gave highest methane production and bio-methane potential which were 1,116 ml and 218 ml CH_4 / $gCOD_{removed}$ respectively. All digesters with ash supplement (2-8 g) gave higher methane yield than the original POME (without ash supplement). The high performer was the digester 2 (added 2 g of ashs). This could be attributed to more suitable environmental condition including, the pH in the range (6.8-7.2 pH) and nutrient balance suitable for methane formation [22, 23]. Which clearly enhanced the anaerobic digestion process. And the digester 2 gave a high buffer capacity (VFA/ALK = 0.55) after the end of the digestion process. As a result, it is the environment condition of digester 2 was suitable than that other digesters for the microorganism in anaerobic digestion and thus the rate of biogas production was relatively higher. Our results were in agreement with the study of Gomez *et al* [20].

TABLE 4. pH, Alkalinity, VFA and methane yield in experiment 2

No.	Ash (g)	pH initial	pH final	ALK Before	VFA Before	VFA/ ALK Before	ALK After	VFA After	VFA/ ALK After	Methane yield (ml.CH_4/ $gCOD_{added}$)	Methane yield (ml.CH_4/ $gCOD_{removed}$)
1	0	7.0	5.0	4,833	3,833	0.79	5,000	7,033	1.41	45.84	103.15
2	2	7.0	5.7	7,166	3,167	0.44	14,575	7,975	0.55	91.16	218.79
3	4	7.1	5.2	7,083	3,283	0.46	11,200	9,667	0.86	83.74	215.34
4	6	7.2	5.3	7,333	3,300	0.45	11,125	10,167	0.91	58.08	149.34
5	8	7.4	5.5	7,500	3,333	0.44	11,650	10,833	0.93	44.17	159.01

Digester No.1 (pH = 7.0 Added NaOH 1 N)
ALK = Alkalinity (mg/l as$CaCO_3$)
VFA = Volatile fatty acid (mg/l asCH_3COOH)

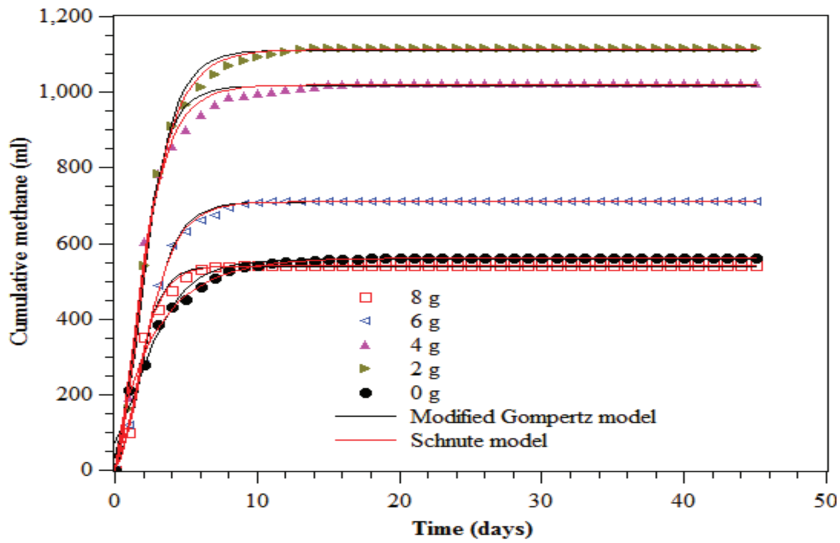

FIGURE 3. Comparison of experimental data and models

The results of the kinetic parameters from kinetic models of experiment 2 are shown in Table 5. One of the most widely used for the kinetic study of biogas production is the modified Gompertz equation as shown in Eq 1 and other models in Eq 2-14. The kinetic constants were determined by non-linear regression. The results model simulation along with experimental are presented in Figure 3 and Figure 4. These results suggest that the pH, Alkalinity (ALK) and VFA has the strong effect on methane yield, P, and R_m but not for lag phase period or minimum time to produce biogas (λ). The digester 2 had the highest value of cumulative methane production (P). This means the ratio of ash to added 2 g to POME 170 ml was an optimum ratio which brought to a good environmental condition in terms of pH and buffer capacity and suitable for bacterial growth, thus the biogas generated maximally. The suitable condition such as buffer capacity, pH, alkalinity, and VFA are the necessary in anaerobic digestion. In the case of pH, the results were in agreement with Chen *et al.* [2] who reported that since fatty acid forms ammonia which has been suggested to be the actual toxic agent [23].

It is interesting to note that all digester it took the shortest time to accumulate the methane gas and reached the final values within approximately 15-20 days. However, lowest of methane yield in digester 1, as considered the very high initial COD of POME, suggested the presence of toxic products or excessive acid accumulation (as shown by low final pH and high VFA/ALK). This brought about the cessation of methanogenesis. This was verified by the experiments using ash to add into palm oil wastewater for adjusting pH, maintaining pH to within methanogen's active limited (6.8 – 7.2 pH). Our results for palm oil mill wastewater were in agreement with the study of Paepatung *et al.* (2009) [14]. And compared the experimental data with kinetic models (Modified Gompertz, Schnute, and Monod) showed that the best-fit curve of the Monod model was still high in the digester 2. In addition the solutions for Gompertz-Monod parameter matching in the study of Siripatana *et al.* (2016) [18]. This a model that better to describes microbial activity than those other models, which showed that Monod-type model is more interpretive, giving better insight on the mechanistic explanation of the biogas data.

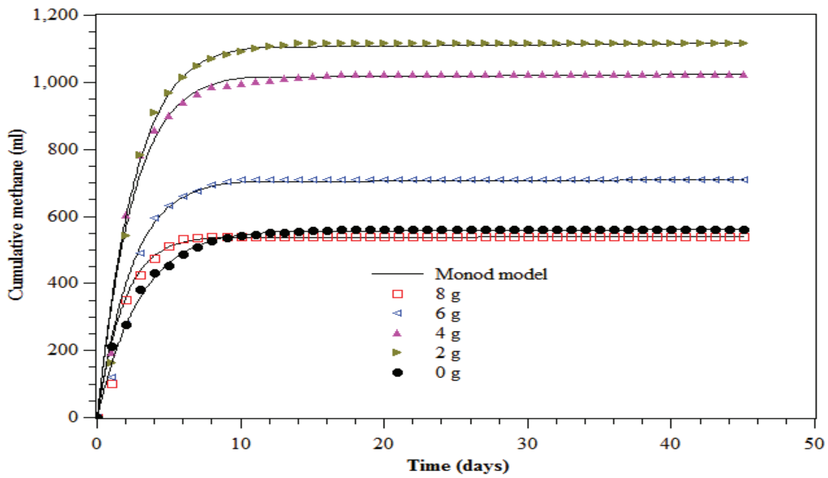

FIGURE 4. Comparison of experimental data and Monod model

TABLE 5. Kinetic constant of methane production in experiment 2

Models	**Parameter**	**0g**	**2g**	**4g**	**6g**	**8g**
General parameters	Initial COD (g l^{-1})	72	72	72	72	72
	P_∞(ml)	561	1116	1025	711	541
	S_0 (g/l)	9.28	9.28	9.28	9.28	9.28
Modified Gompertz equation	R_m(ml d^{-1})	106.40	298.36	307.52	194.85	201.20
	λ(d)	-0.5482	0.3401	0.2479	0.4828	0.4127
	R^2	0.9986	0.9932	0.9871	0.9976	0.9930
Schnute model	r_0(d^{-1})	2,546.4	5,5716	7,0080	63.306	9.4021
	α(d^{-1})	0.3018	0.5851	0.6296	0.5338	0.8707
	β	1.2847	0.4840	0.5386	0.5000	0.3622
	R^2	0.9975	0.9970	0.9919	0.9954	0.9950
Monod	$K = \frac{K_1}{\mu_m}$	2.75	2.3	2.2	2.2	1.7
	P'_0 (ml)	10,000	10,000	10,000	10,000	10,000
	μ_m (d^{-1})	0.4	0.5	0.5	0.4	0.4
	Y_{ps} (ml/mg COD)	0.103	0.219	0.215	0.149	0.159
	K_s (ml/L)	112,840	58,335	56.407	63,266	45,093
	R^2	0.9990	0.9990	0.9990	0.9990	0.9990

CONCLUSION

The results showed that the all kinetic models fitted the experimental data well. The parameters in modified Gompertz model P and R_m are very useful in performance comparison for all substrate sources in anaerobic batch digesters. Thus, the initial COD, pH and buffer capacity has a strong effect on methane yield, P, and Rm but not for lag phase period or minimum time to produce biogas (λ) in this study. However, too high initial COD concentration created conversion imbalance, resulting in excessive accumulation of organic acid, thus brought about lowering pH which caused the methanogen to stop functioning. However, lag phase period (λ) was not affected by the initial COD concentration and initial pH. In addition, in some cases, lag phase period (λ) became negative because the initial biomass concentration was high and active at the same time when exposed to the readily degradable substrate at the initial period of digestion, created the appearance of negative time lag. Finally, our results the best-fit curve of the Monod-type model showed that gave better to describe mechanistic in the batch anaerobic process.

ACKNOWLEDGMENT

The authors would like to thank Yala Rajabhat University grant for the financial support.

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