BIOGAS PRODUCTION KINETIC FROM VINASSE WASTE IN BATCH MODE ANAEROBIC DIGESTION

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ABSTRACT Bottom product of distillation unit from bioethanol industry is often called as vinasse waste. Anaerobic treatment is one of good choice to convert vinasse into biogas. The purpose of this research was to study the biogas production kinetc from vinasse waste in batch mode anaerobic digestion. The kinetic model of biogas production was developed through modified Gompertz equation. Meanwhile, the kinetic of biodegradability of organic material was developed based on first order kinetic reaction. The researh resulted the kineticconstant of biogasproduction were biogas production potential (A),maximum biogas production rate (U), and minimum time to produce biogas (λ) of83,982 mL/(kg COD), 19,71 mL/(kg COD.day), and1.004 days, respectively. Kinetic constant of organic biodegradability material (k) was-0,059day^{-1}. Kinetic model could be used to design volume of batch digesteranaerobic with the formula $V_{digester}$ = 3 * ym (1-exp(-k*t)) * m.

(**Keywords** : batch mode, biogas production, COD/N, kinetic, vinasse, bioethanol industry)

INTRODUCTION

The anaerobic digestion of waste organic materials has two advantages, i.e. treating waste and generating biogas which can be used as alternative energy source. In the anaerobic digestion, organic materials will be converted by bacteria into the biogas through four major phases i.e. hydrolysis, acidonegesis, acetonegesis and methanogenesis[1-2]. In the hydrolysis phase: complex organics (carbohydrate, protein, fat) are converted into simple organics (sugar, amino acid, LVFA); the acidonegesis phase: simple organics are converted into organic acids; the acidogenesis phase: organic acids are converted into acetic acid; the methanogenesis phase: acetic acid is converted into CH_4 and CO_2. Biogas contains 50-75% CH_4, 25-48% CO_2 and other gases in small amount [3-4].

Many authors reported that solid waste such as cattle manure, food waste, municipal solid waste [5-7] had potential to generate biogas using anaerobic digestion treatment. In addition, liquid waste such as vinasse was treated more effectively by using anaerobic treatment than that by using aerobic treatment [8-10]. Vinasse is bottom product of distillation from production ethanol by fermentation. Vinasse contains high COD, more than 100,000 mg/L [11-12].

In ethanol industry, production 1 liter ethanol will generate 8-15 liters [11,13-15]. Because of its COD content, vinasse cannot be discharged directly into the river, so the treatment of vinasse must be done and the best choice is anaerobic treatment. Espinoza-Escalantea[8] studied the effect of initial pH with variation of 4.5; 5.5; 6.5 and the effect of digestion temperature with variation of 35 and 55^{o}C to biogas production. The result showed that initial pH neutral (6.5) and mesophilic temperature (35^{o}C) produced the most biogas yield. Soeprijanto *et al.* [16] studied the effect of COD substrate with variation of 5,000; 10,000; 15,000 to biogas production.

Buitron and Carjaval [9] studied the effect of temperature and HRT with variation 25;35^{o}C and 12;24 hours respectively. From the some other authors that conducted research about production biogas from vinasse above, can be conclude that study of COD/N ratios did not report yet. Wastewater containing COD will be destroyed and converted into biogas optimally if COD/N of substrate in range 350/7 – 1000/7 [17].

Many authors have developed kinetic model of production biogas rate and biodegradability of organic material in anaerobic digestion. Several researchers [5-6,18-21] used modified Gompertz equation that was modified by Zwietering *et al.* [22] to make the kinetic model of biogas production. Meanwhile, Yusuf *et al.*[23] and Yusuf and Ify [24] developed simple kinetic model of biodegradability of organic material based on the first order kinetics. This research studied the kinetic model of biogas production and kinetic model of biodegradability organic material in batch anaerobic digestion.

MATERIALS AND METHODS

Wastewater and Inoculum

The wastewater used was vinasse obtained from an ethanol industry. The ethanol industry located in Solo, Central Java, Indonesia, that produced ethanol from molasses. Properties of vinasse that was used as biogas feedstock are shown in Table 1.

The rumen fluid was used as inoculum. In this study, rumen fluid that was in fresh condition was obtained from slaughterhouse in Semarang, Central Java, Indonesia.

Table 1: Vinasse properties

Parameters	Values
COD	299,250±1.060
TS	27.865±0.000
VS	284,659±0.000
pH	3.25±0.212
N	1,458±0.000

*Remarks: COD, chemical oxygen demand (mg/L); TS, total solid (%); VS, volatile solid (mg/L), pH, power of hydrogen; N, nitrogen content (mg/L).

Preparation substrate

In this study, substrate used was obtained from our previous study. In previous study, authors conducted investigation to know the effect of concentration of solid in substrate to biogas production. Vinasse diluted using water with ratio of vinasse:water of 1:0; 1:1; 1:2; 1:3; 1;4; 1:5. The result showed that substrate with ratio vinasse:water of 1:3 (TS 7.015±0.007%) produced the most cumulative biogas production. Therefore, in this study, authors used substrate with ratio of vinasse:water of 1:3.

Experimental set up

Anaerobic digesters were made from polyethylene bottles which have a volume of 5 L. The bottles were plugged with rubber plug and were equipped with valve for biogas measurement. Anaerobic digesters were operated in batch system and at room temperature. Biogas formed was measured by liquid displacement method as also has been used by the other authors [5, 23-24]. The anaerobic digestion of experimental laboratory set up is shown in Fig. 1.

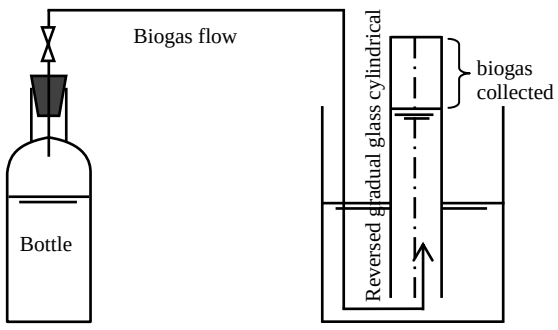


Figure 1: The batch anaerobic digestion of experimental laboratory set up

Experimental design

Anaerobic digestions of experimental laboratory using 5-liter volumes were operated in batch system. 1-liter substrate was put in the digester. Rumen fluid as methanogenic bacteria provider that was added into the digester as much as 10% v/v substrate. From Table 1, can be known that ratio COD/N of vinasse was 1436/7. Meanwhile, ratio COD/N is optimum to produce biogas at range 350/7 – 1000/7 [17]. Furthermore, urea as nitrogen source was added into the digester to make COD/N ratios of 400/7, 500/7, 600/7, 700/7. Initial pH for all variables was adjusted 7.0 by using NaOH solution 10 N. The variables in this study can be seen in Table 2.

Table 2: Variation of COD/N ratios

Digester	Substrate (mL)	Rumen (mL)	COD/N
A	1000	100	1436/7 (control)
B	1000	100	400/7
C	1000	100	500/7
D	1000	100	600/7
E	1000	100	700/7

*Remarks: COD, Chemical Oxygen Demand; N, total nitrogen

Experimental procedures

Biogas formed was measured every once in two days to know biogas production with water displacement method (Fig 1). pH substrates in the digester were measured by pH meter every once in two days to know pH profile daily.

Kinetic model of biogas production

Biogas production kinetic was modeled through modified Gompertz equation[23]. Kinetic of biogas production in batch condition was assumed that had correspondence to specific growth rate of methanogenic bacteria in digester [5-6, 18-21, 23]. The modified Gompertz equation as follows:

$$P = A.\exp\left\{-\exp\left[\frac{U.e}{A}(\lambda - t) + 1\right]\right\} \qquad (1)$$

Where P is cumulative of specific biogas production (mL/g COD), A is biogas production potential (mL/g COD), U is maximum biogas production rate (mL/g COD.day), λ is lag phase period or minimum time to produce biogas (days), t is cumulative time for biogas production (days) and e is mathematical constant (2.718282). Kinetic constant of A, λ and U was determined using non-linear regression with help of polymath software [5, 20-21].

Kinetic model of biodegradability of organic material

Authors developed kinetic model of biodegradability of organic material based on first order reaction. This concept also was developed by Yusuf *et al.* [23] and Yusuf and Ify [24]. Assumsion:

$$\underset{(C)}{\text{COD}} \longrightarrow \underset{(B)}{\text{Biogas}} \qquad (2)$$

In the first order reaction, organic material (COD, symbolized C) was converted into biogas (symbolized B) with reaction rate formula, V = -k*C = k*B, with k = reaction rate constant.

$$V_d\frac{dC}{dt} = Q_i{*}C_i - Q_o{*}C_o + V_d(-k{*}C) \qquad (3)$$

In batch system, flow of input (Q_i) = flow of output (Q_o) = 0. Whereas C_i and C_o were influent and effluent COD and V_d was volume of digester, so that the equation (3) can be written as:

$$V_d\frac{dC}{dt} = V_d\,(-k{*}C) \qquad (4)$$

Both sides of equation (4) were divided by Vd, so equation (4) can be written as:

$$\frac{dC}{dt} = (-k{*}C)$$

$$\frac{dC}{C} = -k{*}dt$$

$$\int_{Co}^{Ct}\frac{dC}{C} = -k\int_0^t dt$$

$$\ln\left(\frac{Ct}{Co}\right) = -k{*}t \qquad (5)$$

Correlation between substrate biodegradability and biogas yield at any time (y_t) can be developed assuming all substrate (COD) are converted into biogas as shown in Fig 2 [25]. From Fig 2, can be deduced that:

$$\frac{Co-Ct}{Co} = \frac{yt}{ym} \qquad (6)$$

$$\frac{Co}{Ct} = \frac{ym}{ym-yt} \qquad (7)$$

Substituting equation (5) into (7) to get (8)

$$\ln\left(\frac{ym-yt}{ym}\right) = -k{*}t \qquad (8)$$

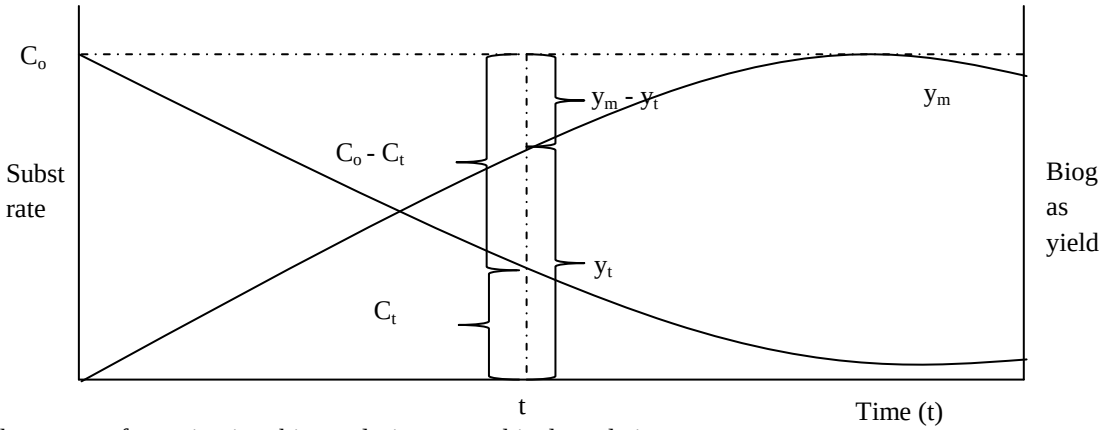


Figure 2: Subtrate transformation into biogas during anaerobic degradation

Rearrange (8) to get (9)

$$\frac{ym-yt}{ym} = \exp(-k*t)$$

$$ym\,(1-\exp(-k*t)) = yt \qquad (9)$$

From equation (9); ym, volume of biogas formed at maximum time (mL/kg COD); yt, volume of biogas formed at any time (t); -k, rate constant associated with degradation of the material organic (/day). Linearization of equation (9) by differentiation,

$$yt = ym\,(1-\exp(-k*t))$$

$$\frac{dyt}{dt} = 0 - (-k)*ym*\exp(-k*t)$$

$$\frac{dyt}{dt} = k*ym*\exp(-k*t) \qquad (10)$$

Taking natural logarithm on both sides of the equation (10)

$$\ln\left(\frac{dyt}{dt}\right) = \ln(k*ym*\exp(-k*t)$$

$$\ln\left(\frac{dyt}{dt}\right) = (\ln ym + \ln k) - k*t$$

$$\frac{1}{t}\ln\left(\frac{dyt}{dt}\right) = \frac{1}{t}(\ln ym + \ln k) - k \qquad (11)$$

Equation (11) represented straight line equation y = mx + c. Slope of straight line equation (m) represented the value of (lnym + ln k) and intercept of that (c) represented the value of (-k).

RESULTS AND DISCUSSIONS

Effect of COD:N ratio to kinetic model of biogas production

Biogas production for all variables was modeled based on modified Gompertz equation. Kinetic constant of A, U and λ was determined by using non-linear regression. Kinetic constants obtained were presented completely in Table 3. By plotting experimental data and simulation of modified Gompertz equation was obtained the graph as shown in Fig 3.

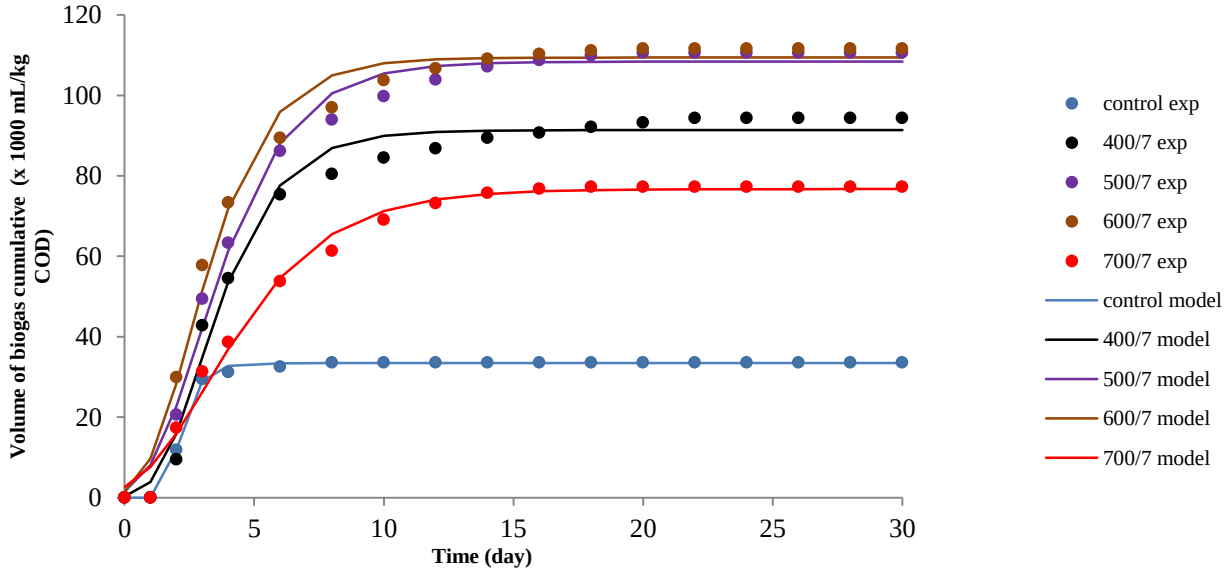
From Table 3., difference in the COD:N ratio affected value of kinetic constant. Control variable had the lowest value of A which was 33,429 mL/kg COD. That means control variable in prediction generated biogas in little amount. Meanwhile, variable with COD/N of 600/7 had the highest value of A which was 109,368 mL/kg COD.

Variable that had optimum COD/N ratio caused good condition for bacterial growth in the digester so that biogas will be generated maximally. COD/N ratio of substrate is necessary parameter in anaerobic treatment, optimum COD/N ratio is in range of 350/7 – 1000/7. If COD/N is out from that range, bacterial growth will be disturbed [17]. Nitrogen was needed by bacteria to build cell structures [26-27].

Table 3: Kinetic constant of biogas production

Variable	Total of Biogas Volume (mL/kg COD)	Modified Gompertz Equation (Model)			R^2
		A (mL/kg COD)	U (mL/kg COD. day)	λ (day)	
Control	33,591.78	33,429	24,165	1.505	0.998
400/7	94,376.29	91,354	19,954	1.255	0.986
500/7	110,550.60	108,444	20,156	0.908	0.991
600/7	111,649.80	109,368	23,466	0.803	0.988
700/7	77,233.62	76,712	10,807	0.572	0.991
Constant average		83,982	19,71	1.004	0.991

Remarks: A, biogas production potential; U, maximum biogas production rate; λ, minimum time to produce biogas; R^2, correlation coefficient.



Remarks: exp, data from experiment data; model, data from modified Gompertz model

Figure 3: Comparison of experimental data and modified Gompertz model

Variable with COD/N of 400/7 and 700/7 had more value of A constant and produced more biogas than control variable. In control variable, COD/N ratio was not appropriate, the amount of nitrogen total in substrate was too little so that bacteria could not build cell structures and finally death. On the other hand, variable with COD/N of 400/7 and 700/7 had less value of A constant than that of 500/7 and 600/7.

Protein and urea in the substrate was decomposed to be ammonia/ammonium. Ammonia/ammonium was used by bacteria as nitrogen source [28] but ammonia/ammonium in large amount became toxic to

bacterial activity. De-Baere *et al.* [29] reported that concentration of ammonia of 100-140 mg/L hampered bacterial growth in mesophilic temperature. Omil *et al.* [30] stated that ammonia with concentration 25 mg/L was minimum concentration that disturbed bacterial activity. Meanwhile according to Deublein and Steinhauser [31], ammonia concentration of 80 mg/L was minimum concentration to disturb bacterial growth and 150 mg/L was toxic to bacteria.Methanogenic bacteria was the least tolerant and the most easily killed to ammonia inhibition among the four anaerobic bacteria in four step biogas production there were hydrolysis, acidogenesis, acetogenesis, methanogenesis.

Ammonia (NH_3) formedammonium (NH_4^+) depend on pH condition. Ammonium had less toxic than ammonia. Ammonium will be toxic just in high concentration. Ammonium concentration of 1,500-10,000 mg/L was inhibition start and that of 30,000 mg/L was toxicity concentration [31].

Substrate with COD/N ratio of 400/7 might contain nitrogen total that was too much, so that ammonia/ammonium formed caused toxicity for bacterial activity. Whereas, substrate with COD/N ratio of 700/7 contained nitrogen total that was not in appropriate amount yet.Although, COD/N of 400/7 and 700/7 was included in optimum range that was stated by Speece [17]. The good COD/N ratio in this study was 500/7 - 600/7.

From Table 3, the U constant value of control variable was the highest of all variables, which was 24,165 mL/kg COD.day. That was caused by cumulative time that needed to produce biogas. Control variable produced biogas just until 8th day fermentation (Fig 3.), whereas the other variables generated biogas until up to 15th day of fermentation. Although control variable had value of U constant was highest, it had value of A was lowest because biogas production process took in short time.

Control variable had high value of λ. Budiyono *et al.* [5] stated that variable that had little value of kinetic constant of λ, needed just little time to produce biogas. Zwietering *et al.* [22] reported that value of λ indicated the time that was required for bacteria to adapt. Based on that, bacteria in control variable needed much time to adapt and produce biogas which was 1.505 days.Whereas, variable with COD/N ratio of 400/7, 500/7, 600/7, 700/7 needed less time than control variable. Bacteria needed nitrogen to build cell structures, so availability of nitrogen in appropriate amount caused good growth of bacteria in digester. If bacteria is not lack of nutrient, degradation activity is done well and biogas will be generated immediately.

Effect of COD:N ratio to kinetic model of biodegradability of organic material

From equation (11), we had straight line equation $\frac{1}{t}\ln(\frac{dyt}{dt}) = \frac{1}{t}(\ln ym + \ln k) - k$. The value of k obtained by plotting $\frac{1}{t}\ln(\frac{dyt}{dt})$ against $\frac{1}{t}$. The results of plotting that can be seen in Fig. 4. Yusuf *et al.* [23] stated that the term (-k) was a measure of the rate of removal of the biodegradable fractions as the biogas yield increased with time. This rate constant was an aspect of the first order rate constant. The more negative the value of (-k), the faster the rates of removal of the biodegradable fractions. The value of (-k) for control, 400/7, 500/7, 600/7, 700/7 was (-0.6466), (-0.1852), (-0.2365), (-0.2876), (-0.2355) respectively with good value of R^2 in range 0.9867 – 0.9996.

From Figure 4, the most negative of (-k) value was in control variable but it generated the least biogas total. Bacteria in control variable generated biogas in large amount at beginning fermentation. This was caused by characteristic of vinasse. Vinasse contained simple organic materials such as acetic acid, lactic acid and glycerol [32], so that bacteria could easily degrade them into biogas. After 8th day, biogas was not generated. Meanwhile in variable with COD/N ratio of 400/7 – 700/7, biogas was generated biogas in large amount at beginning (at 2nd – 3rd day), then decreased until 18th – 22nd day. In control variable, the process of decreasing of biogas production took the shortest time of all variable so that biodegradability rate was high although biogas formed was little. Biogas production daily can be seen in Fig 5.

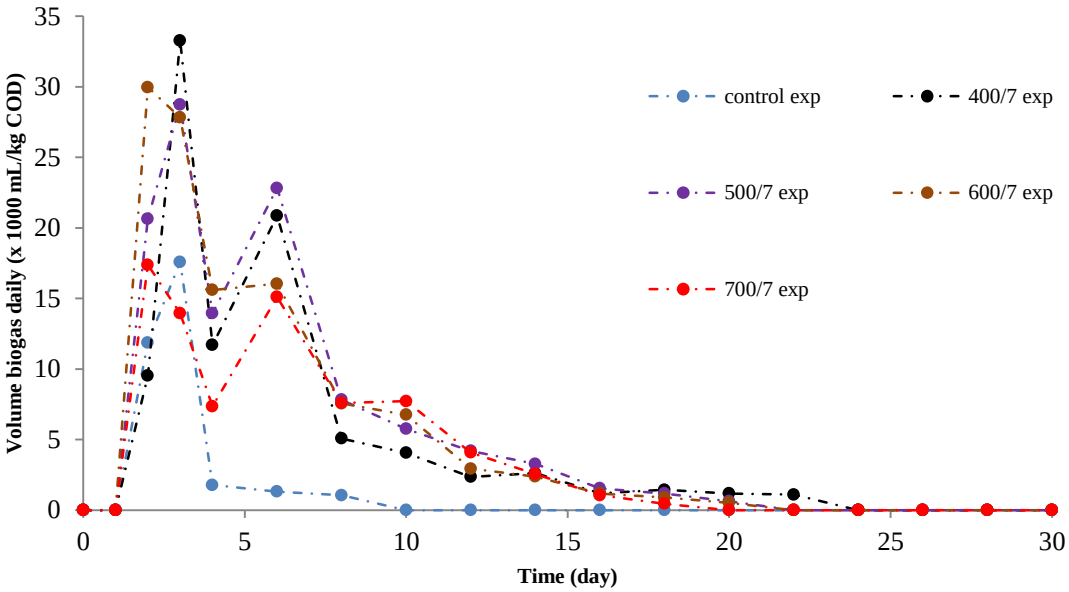
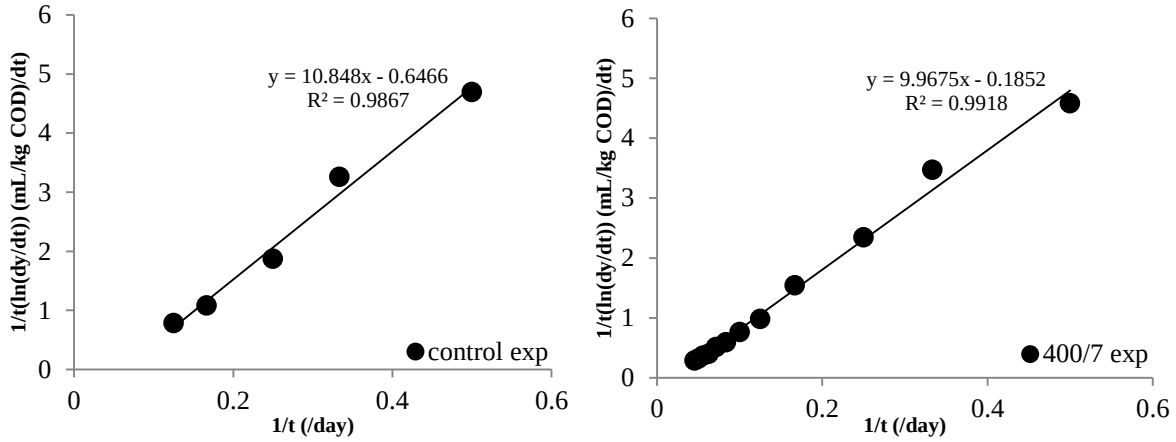
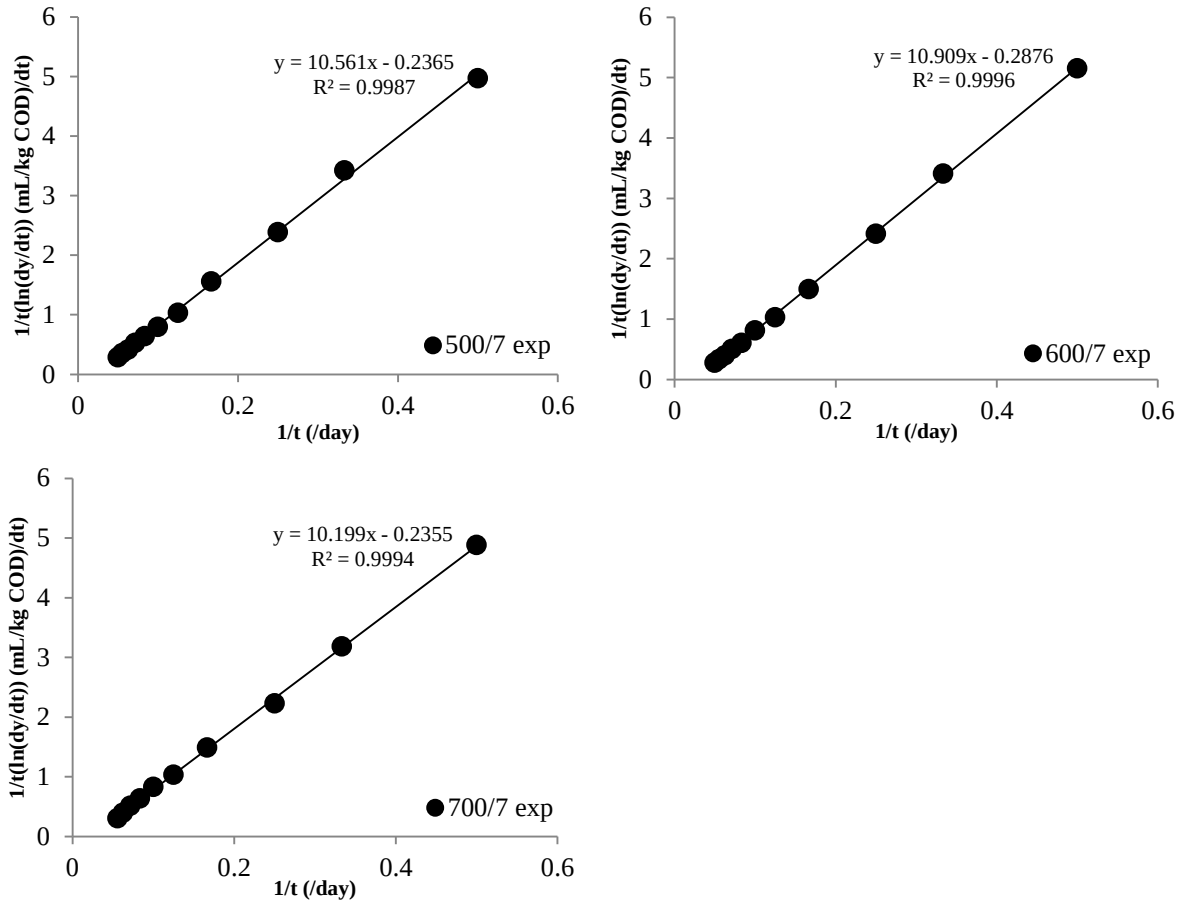


Figure 5: Volume biogas daily (experiment)

Among of COD/N ratio with variation of 400/7, 500/7, 600/7 and 700/7, COD/N of 600/7 had the most negative of (-k) value which was (-0.2876). That means organic material (COD) that was contained in substrate, was faster to be degraded than that in the other variables. COD/N of 600/7 also had the highest value of biogas production potential constant (A). Yusuf *et al.* [23] reported that the more negative of (-k) value was obtained from first order model, the more biogas production potential (A) was obtained from modified Gompertz model. In this study, variable with COD/N ratio of 500/7 and 600/7 that had the most value of A (108,444 and 109,368 mL/kg COD respectively) had the most negative of k value (-0.2365 and -0.2876 /day respectively), so this result was similar with the result of Yusuf *et al.* [23].





*Remark: exp, based on experiment data

Figure 4: Plot of 1/t (ln(dy/dt)) (mL/kg COD/dt) against 1/t (/day)

pH profile

pH profile for all variables is shown in Fig 6. Elbeshbishy and Nakhla [33] stated that decreasing of pH was caused by VFAs (organic acid) production at beginning of fermentation. Vinasse used in this study was obtained from ethanol industry that produced ethanol from molasses. Ethanol was produced by fermentation yeast. Ethanol formed was separated from fermentation broth by distillation. The bottom product of distillation process was vinasse. Therefore, vinasse contained short chain molecular that was easy to be degraded by bacteria into VFAs (organic acid) so pH medium was easy to drop.

Deublein and Steinhauser [31] reported that there were two kinds of organic acid which were dissociated and not-dissociated acid. Composition of them in substrate was depended by pH condition. The more acidic of pH substrate, the more amount of not-dissociated acid was in substrate. Presence of not-dissociated acid hampered bacterial activity, because it was penetrated into cell through cell membrane and then spoiled the protein of bacteria.

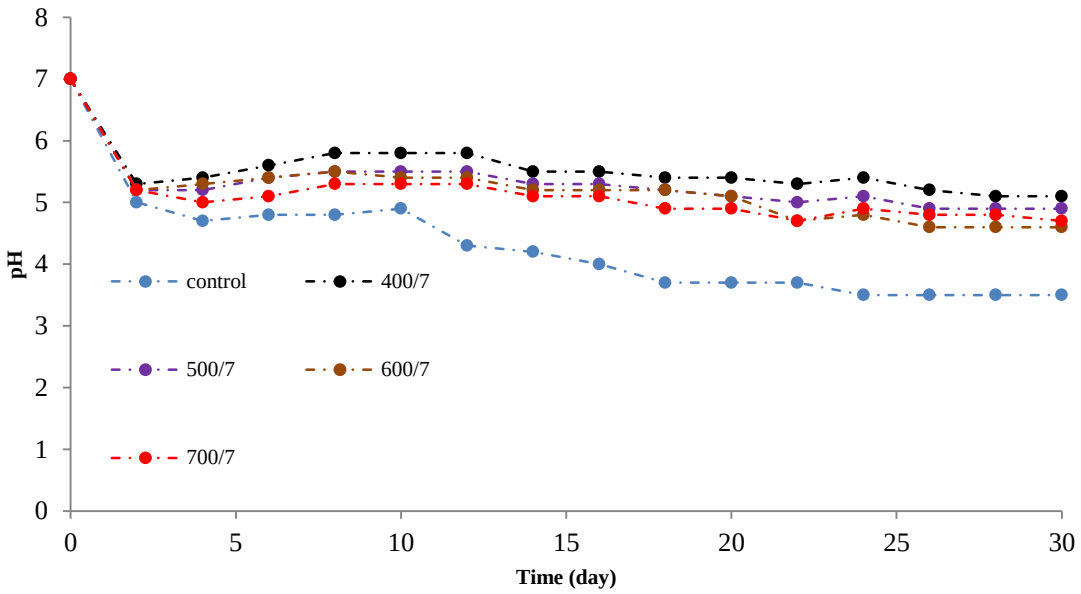


Figure 6: pH profile

Profile of pH for all variables showed that decreasing pH from beginning until ending of fermentation. Control variable had the sharpest trend of decreasing pH substrate of all variables. This phenomenon was caused by presence of total nitrogen. According to Mata-Alvarez *et al.* [34], substrate contained carbohydrate in large amount potentially produced VFAs that caused acidity in pH. Whereas substrate that contained nitrogen in large amount potentially produced NH_4^+ that caused alkalinity in pH.

Elbeshbishy and Nakhla [33] reported that decreasing in the pH could be due to the rapid VFAs production at the beginning, while the increasing in the pH from 3rdday to 10thday could be due to generation of NH_4^+ during protein degradation, as ammonia which was a base combines with carbon dioxide and water to form ammonium bicarbonate (a natural pH buffer). However, in this experiment, pH decreased until the end (Fig 6). This phenomena was caused accumulation of VFAs production in the digester was very large. Condition of pH in the substrate was very acid and methanogenic bacteria was death. Ammonium production from degradation urea and protein could not increase the pH because VFAs was produced rapidly.

Design of anaerobic batch digester using kinetic model

Yusuf and Ify[24] designed batch digester to treat cow dung, waste paper and water hyacinth. A ratio of volume gas chamber:volume of anaerobic digester of 1:3 was used by Yusuf and Ify [24] and Igoni *et al.*. [35]as establishing basic to design batch digester. Volume gas chamber (V_{gc})was proportional to the volume of biogas formed.

$$V_{gc} = 1/3\ V_{digester} \quad (11)$$

$$3\ V_{gc} = V_{digester} \quad (12)$$

V_{gc} (mL) = yt (mL/kg COD) * m (mass of COD fed into the digester), (13)

And from equation (9), we had

$$yt = ym\ (1-\exp(-k*t)) \quad (9)$$

Subtituting equation (13) into (12)

$$V_{digester} = 3 * yt * m \quad (14)$$

And, substituting equation (9) into (14)

$$V_{digester} = 3 * ym (1-exp(-k*t)) * m \quad (15)$$

ym was volume of biogas formed at maximum time (mL/kg COD), so we can replace value of ym with kinetic constant A which was 109,368 mL/kg COD (in variable with COD/N of 600/7). Assuming we want to treat 10 L vinasse that contained 299,250 mg/L COD, so substrate contained 2,992,500 mg COD = 2.992 kg. Retention time that was needed based on Fig 5. was 20 days. So $V_{digester}$= 3 * 109,368 mL/kg COD (1-exp(-0.2876/day*20 days)) * 2.992 kg. Thus, $V_{digester}$ = 978568 mL = 978.6 Liter. Detail calculation of $V_{digester}$ for all variables can be seen in Table 4.

Table 4: Calculation of digester volume for treat 10 L vinasse (contains 299,250 mg/L COD)

Variable	m (kg COD)	A (mL/kg COD)	-k (/day)	t (based on Fig 5) (day)	$V_{digester}$ (Liter)
Control	2.992	33,429	-0.6466	8	298.4
400/7	2.992	91,354	-0.1852	22	806.1
500/7	2.992	108,444	-0.2365	20	964.8
600/7	2.992	109,368	-0.2876	20	978.6
700/7	2.992	76,712	-0.2355	18	678.6

Remarks: m, mass of COD fed into digester; A, biogas production potential; -k, biodegradability rate of COD; t, retention time

CONCLUSION

Variation of COD/N ratio affected value of kinetic constant on kinetic model of biogas production that was modeled through modified Gompertz model. Variable with COD/N of 600/7 that was the best variable, had the values of A (biogas production potential), U (maximum biogas production rate) and λ (minimum time to produce biogas) which were 109,368 mL/kg COD; 23,466 mL/kgCOD.day; 0.803 day. On kinetic model of biodegradability of organic material, variable with COD/N of 600/7 had the most negative value of (-k) (biodegradability rate constant, -0.2876 /day), that means organic material of substrate that had COD/N of 600/7 was easy to be degraded by bacteria. Volume of digester could be designed with formula of $V_{digester}$ = 3 * ym (1-exp(-k*t)) * m.

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