Modelling the Kinetic of Biogas Production from Co-digestion of Pig Waste and Grass Clippings

Anthony Njuguna Matheri[1], *Member, IAENG*, Mohamed Belaid[2], *Member, IAENG*, Tumisang Seodigeng[3], Catherine Jane Ngila[4]

Abstract— **This work investigated the use of laboratory batch anaerobic digester to derive kinetics parameters for anaerobic co-digestion of pig waste and grass clippings. Laboratory experiment data from 10 litres batch anaerobic digester operating at ambient mesophilic temperature of 37 ^{0}C and pH of 6.9 was used to derive parameters for modified Gompertz model. The carbon/nitrogen (C/N) ratio of Pig waste was found to be 16.16 and grass clippings to be 20.54. Through co-digestion in ratio of 1:1, the C/N ratio settled at 17.28. The actual biogas yield was found to be 7725 ml/g COD. In the model of biogas production prediction, the kinetics constants of A (ml/g COD), μ (ml/g COD. day), λ (day) was 7920.70, 701.35, 1.61 respectively with coefficient of determination (R^2) of 0.9994. Modified Gompertz plot showed better correlation of cumulative biogas production and these results show biogas production can be enhanced from co-digestion of substrates.**

Keywords— **Anaerobic, Co-digestion, Kinetics, Mesophilic Temperature, Modified Gompertz**

I. INTRODUCTION

THE energy consumption worldwide is spontaneously increasing due to industrialization, population growth and state of development in both developing and developed countries. The need for alternative sources of energy for centralized and decentralized power generation has led to researchers looking for alternative source of renewable energy.

With the fast depletion of non-renewable energy sources such as fossil fuel, coal and petroleum which has led to global climate change, human health problems and environmental degradation. The commercial production of biogas and other alternative energy source such as solar energy, wind energy, hydropower, geothermal will definitely give a drive for the development of the economy. Energy derived from biogas is used in the form of fuel, heat and electricity. It is desirable to create sustainable and with zero carbon emissions world-wide energy system [1, 2].

Biogas is a renewable source of energy derived from biodegradable substrates such as agricultural wastes, animal wastes, domestic wastes, crops and industrial waste. It is produced by anaerobic digestion, which is a biochemical process in absence of oxygen. The main product of biogas is methane and carbon dioxide [3, 4].

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II. BIOCHEMICAL PROCESS OF ANAEROBIC DIGESTION

Biogas production follows four fundamentals processes. These processes include; hydrolysis, acidogenesis, acetogenesis and methanogenesis [5]. Fig. 1 shows a simplified generic anaerobic digestion process [6].

The anaerobic system is as the result of complex interactions among different of bacteria. The major functional groups of bacteria according to their metabolic reactions are [7]: Fermentative bacteria, hydrogen-producing acetogenic bacteria, hydrogen-consuming acetogenic bacteria, carbon dioxide reducing methanogens and aceticlastic methanogens.

A.Hydrolysis

Organic waste used in anaerobic digestion are originally made up of large carbon molecules called biomass. In the first stage of the AD process, they are hydrolyzed into smaller soluble molecules [8]. The products from this stage are normally monosaccharaides from carbohydrates, amino acids from proteins, and long-fatty acids and glycerine from lipids. Hydrolysis is mediated by extracellular enzymes produced by fermentative bacteria including cellulases,

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amylases, proteases, lipases and protease [9]. This process is reported to be a rate-limiting stage in anaerobic digestion while its inhibition is dependent on the type of substrates used during the process and temperature of the digester [9].

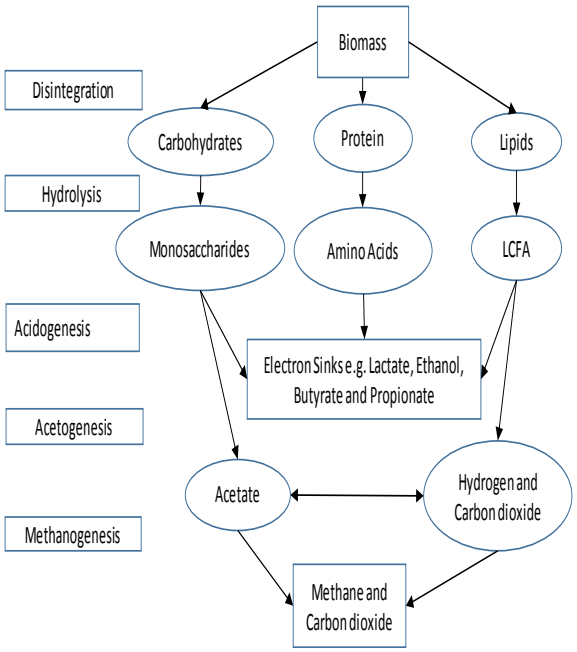

Fig.1. Degradation steps of anaerobic digestion process.

B. Acidogenesis

Acidogenesis, also known as acid formation stage, is the second step of anaerobic digestion. It is usually the fastest reaction in the overall anaerobic digestion process [10]. This process involves further breaking down of the simple molecules created through hydrolysis to a mixture of organic acid (lactate, butyrate, ethanol and propionate), hydrogen and carbon dioxide. The bacteria responsible for this stage are called acidogenesis (fermentative) bacteria or hydrogen-producing acetogenic bacteria. Apparently, the main product of this process is depended on the anaerobic microbial species present, and culture conditions. At low partial pressure of hydrogen, acetate and /or hydrogen dominate the product, while at high partial pressure of hydrogen; ethanol or organic acid is produced [11, 12]. Since acidogenesis bacteria are strictly anaerobic, thus obligate and facultative such as *Peptococcus anaerobes*, *Clostridium* ssp, and *Lactobacillus* and *Escherichia coli* are involved for the removal of oxygen, whenever available [13]. During acidogenesis, an acidic environment in the digester is created due to the generation of ammonia, H_2, CO_2, H_2S, shorter volatile fatty acids, carbonic acids, alcohols, as well as trace amounts of other by-products [14]. The volatile fatty acid concentration accumulated in the digester have a significant impact in the overall performance of the process, since acetic and butyric acids are the preferred precursor for methane formation [15].

C. Acetogenesis

In the third stage, the products from acidogenesis are completely converted to acetate, hydrogen and carbon dioxide by group of bacteria know as hydrogen-consuming acetogenic bacteria or acetogenic bacteria [16]. The entire products from this stage are used up for methane production. During this process, 17% of the energy is converted to acetic acid and 13% to hydrogen [10]. Table 1 shows the reactions and free energy changes of lactate, ethanol, butyrate, propionate, methanol, hydrogen-CO_2 and Palmitate during acetogenesis.

Acetogenic organisms are the connection between hydrolysis/acidogenesis and methanogenesis [16]. Acetogenesis is regarded as the most important stage, as it produces the main substrate for the last stage which are hydrogen, carbon dioxide and acetate. Tesfaye [15] reported that methanogenic bacteria are unable to process any substrate other than acetate, carbon dioxide and hydrogen. Thus, the performance of this stage is depended on the hydrogen partial pressure. If the hydrogen partial pressure is kept below 10-3 atm, hydrogen, acetate and carbon dioxide dominate the product, but if the hydrogen partial pressure is above the standard fatty acids will be produced which makes methanogenesis unfavorable. The pressure is controlled through efficient removal of hydrogen by hydrogen-consuming organisms such as hydrogenotrophic methanogens. Acetogenic bacteria are also sensitive to physical changes such as fluctuation in organic loading rate [17].

TABLE I
STOICHIOMETRY AND CHANGE OF FREE ENERGY (ΔG°′) FOR ACETOGENIC REACTIONS [18].

Compound	Reaction	ΔG°'(kJ/mole)	Eqn
Lactate	$CH_3CHOHCOO^- + 2H_2O \rightarrow CH_3COO^- + HCO_3^- + H^+ + 2H_2$	-4.20	(1)
Ethanol	$CH_3CH_2OH + H_2O \rightarrow CH_3COO^- + H^+ + 2H_2$	9.60	(2)
Butyrate	$CH_3CH_2CH_2COO^- + 2H_2O \rightarrow 2CH_3COO^- + H^+ + 2H_2$	48.10	(3)
Propionate	$CH_3CH_2COO^- + 3H_2O \rightarrow CH_3COO^- + HCO_3 + H^+ + 3H_2$	79.10	(4)
Methanol	$4CH_3OH + 2CO_2 \rightarrow 3CH_3COOH + 2H_2O$	-2.90	(5)
Hydrogen-CO_2	$2HCO_3^- + 4H_2 + H^+ \rightarrow CH_3COO^- + 4H_2O$	-70.30	(6)
Palmitate	$CH_3(CH_2)_{14}COO^- + 14H_2O \rightarrow 8CH_3COO^- + 7H^+ + 14H_2$	345.6	(7)

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D. Methanogenesis

Methanogenes is the final stage of anaerobic digestion where hydrogen, carbon dioxide and acetate are converted to methane [19]. The formation of methane involves two biological reactions. The primary reaction is where acetate is degraded to methane and carbon dioxide as referred in "(8)" [20].

$$CH_3COOH \rightarrow CH_4 + CO_2 \quad . \quad (8)$$

The second reaction occurs when methanogenic archea reduce carbon dioxide using hydrogen as electro donor to form methane and water as indicated in "(9)" [16].

$$4H_2 + CO_2 \rightarrow CH_4 + 2H_2O \quad (9)$$

This reaction is known as a rate limiting stage in anaerobic digestion. During this process, 65-70% of methane is produced from acetate, then 27-30% from hydrogen and carbon dioxide [8, 16, 17]. Products from this process are the ones that make up the majority of the biogas. The metabolism and activity of microorganism governing this stage are highly affected by the change or fluctuation of pH and temperature than any other microbial balance in the digester [9]. In addition, methane producing bacteria are mostly likely to cease growth due to inhibition of ammonia to anaerobic process [21].

III. PARAMETERS AFFECTING ANAEROBIC DIGESTION

The activity of biogas production depends on various parameters like temperature, partial pressure, pH, hydraulic retention time, C/N ratio, pre-treatment of feedstock, trace of metals (trace elements) and concentration of substrate [2, 22-24].

A. Temperature

The anaerobic process is so sensible to temperature; change of acetic acid (acetate) to methane depends mostly on temperature but conversion to acetic acid will not affect much by slight temperature variations. Grimberg et al., [25] reported that the environmental temperature has a major influence on the anaerobic microbial systems, which affects the metabolic rate, ionization equilibria, substrate solubility and fats. Higher temperature affects the activity of hydrogenotropic methanogens in the anaerobic process and enriches hydrogen producing bacteria and spore forming bacteria [25]. Mesophilic digestion temperature is considered to be most suitable for anaerobic digestion on the ranges of 35-37℃. In thermophilic digestion, 55℃ is considered to be ideal [26]. Table II shows different thermal stages, process temperatures and typical hydraulic retention times for the AD process.

TABLE II
THERMAL STAGES, PROCESS TEMPERATURE AND TYPICAL HYDRAULIC RETENTION TIMES [27].

Thermal stages	Process temperature (^{0}C)	HRT(days)
Psychrophilic	<20	From 70-80
Mesophilic	From 30-42	From 14-40
Thermophilic	From 43-55	From 14-20

B. pH

The pH-value is the measure of alkalinity/acidity of a solution [28]. It affects the production of biogas because each group of the microorganisms have different optimum pH range. Methanogenic bacteria have an optimum pH between 6.5 and 7.5. They are extremely sensitive to pH. The fermentative micro-organisms are less sensitive to pH since they have wide optimum pH range between 4.0 and 8.5. Low pH level favours the production of acids such as butyric acid, propionic acids and acetic mainly at pH of 4.0. At pH higher than 8.0, ammonia is mainly produced. The presence of volatile fatty acids (VFAs) tends to decrease the pH and can lower the methanogenic bacteria activity and hence the biogas production [29].

C. Volatile fatty acid

The VFA's uptake play a crucial role in the whole degradation kinetics of organic waste digestion, as the accumulation of the intermediate products, VFAs, is the rate-limiting step [25]. High concentrations VFAs in the digester lower the pH, inhibit methanogenic activity and cause possible failure of the anaerobic digestion process [25].

D. Carbon/Nitrogen ratio

The carbon/nitrogen (C/N) ratio represent the relationship between the amount of carbon and nitrogen present in organic substrate. The optimal C/N ratio for anaerobic digestion is considered to be in the range of 15-30 [30]. If the C/N ratio is too high, the nitrogen is consumed rapidly by the methanogens bacteria to meet their protein requirement and is no longer available to react on the left-over carbon content in the material. As a result the biogas production is reduced [30]. If the C/N ratio is too low, nitrogen is liberated and accumulates in the form of ammonia [30]. This increases the pH of the digestates. When pH value rises higher than 8.5, it begins to exert a toxic effect on the methanogenic bacteria [30]. To maintain

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the C/N level of the digester substrate at optimum levels, substrate of high C/N ratio can be co-digested with substrate of low C/N ratio [30].

E. Retention time

Retention time is the time required to degrade the organic matter (substrate) completely and for bacterial to grow. The retention time depends on process temperature and batch composition, meaning retention time for waste treated in a mesophilic condition than thermophilic conditions [31], the residence time is generally positively correlated with methane content. There are two important types of retention time that include; solid retention time (SRT) and hydraulic retention time (HRT). SRT is the average time the bacteria (solids) are in the anaerobic digester, and HRT is the volume of the biological reactor per influent flow rate in time, which is defined by following equation: Digestion time inside the reactor is one of the main factors influencing the CH_4 yield [31]. Effective hydraulic retention time depends on the type of substrate, loading rate, and reaches up to a couple of weeks. Shorter HRT usually results in accumulation of VFAs, whereas at HRT longer than optimal, the digester components are not effectively utilized [31].

F. The organic loading rate (OLR)

The organic loading rate (OLR) is the amount of volatile solids (VS) to be fed into the digester each day in a continuous process. As the OLR increases, the biogas yield increases to some extent but above the optimal organic loading rate, the volatile solids degradation and biogas yield decreases due to overloading [32]. The maximum possible OLR depends on the process temperature and its retention time.

G. Toxicity

Mineral ions, especially of trace elements are among the materials that inhibit the growth of bacteria in a digester. Small amount of mineral (calcium, sodium, potassium, sulphur, magnesium and ammonium) stimulate the microorganisms growth, but higher concentrations have a toxic and inhibition effect [26]. Heavy metals such as zinc, nickel, cobalt, copper, lead and chromium are essential for bacterial growth in very small quantities, but higher quantities have a toxic bacteria effect. Organic solvents and antibiotic also inhibit the bacteria. Recovery of digesters can only be achieved by flushing the content, cessation of feeding, or diluting the contents to lower the concentration of inhibitory substances to below the toxic level [26].

H. Ammonia

Studies in the past have showed that ammonia is an important source of nitrogen for bacteria, low concentrations of ammonia is valuable to the process [33], although some findings showed that the specific activity of methanogenic bacteria decreases with increasing in concentrations of ammonia [33]. The mechanisms ammonia inhibition are change in the intracellular pH, increase of maintenance energy requirement as well as inhibition of a specific enzyme reaction [33]. And high concentration of ammonia in the digester decreases the deamination activity of proteolytic bacteria [34].

I. Agitation/Mixing

Mixing is required to maintain fluid homogeneity, hence process stability, temperature distribution, within a digester. The objectives of mixing are to combine the incoming substrate with the bacteria, to reduce the formation of scum, and to avoid pronounced temperature gradients within the digester. Very rapid mixing can disrupt the microbial balance while too slow stirring can cause short-circuiting and inadequate mixing. [26].

J. Dilution

Water should be added, if necessary, to the substrate to generate a slurry which is neither too thick nor too thin. If a slurry is diluted too much, the solid particles may settle down in the digester and may not get degraded properly. If the slurry is too thick, it may be difficult to stir and may impede the flow of gas to the upper part of the digester. Different systems can handle different levels of slurry density, generally in the range of 10-25% of solids [26].

K. Solid Residue/Slurry

When the anaerobic degradation is nearly complete, the solid residue or digestate is removed and is normally cured aerobically and screened for items such as plastic pieces, glass, shards etc., before being disposed on land as fertilizer [26].

L. Grinding

Grinding or breaking down of substrate into small pieces before feeding them into the digester will decrease the retention time in digestion and enhance biogas production. Since materials grinded increases their surface area of contact with anaerobic bacteria and thus simplifying the digestion process [35].

M. Co-digestion

Studies show that co-digestion is a way of minimising HRT and improving methane production [36]. The other substrate should be manure which is dominated by high levels of organisms that have the ability to hydrolyse lingo-cellulose material. Co-digestion of biomass waste can produce more methane than manure itself, but the challenge in this process is to achieve completely break down of organic material in stage of hydrolysis [36]. The importance of co-digestion is to stabilize conditions or other parameters in digestion process such as C:N ratio as well as pH,

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macronutrients and micronutrients, inhibitors and dry material [36].

N. Substrate pre-treatment

Pre-treatment is done to increase the efficiency of anaerobic digestion technology and increase the production of biogas [37]. Pre-treatment can be classified as thermal, mechanical, biochemical pre-treatment. Pre-treatment is necessary since the nature of a substrate has an effect on the rate of biogas production [37].

IV. Modelling of Anaerobic Digestion

The option to convert biogas to natural gas is purely relevant in large scale production. Development of appropriate models are the best steps for complete process. For nearly 40 years, scientists have developed and improved on the anaerobic digestion models of organic substances [38]. Primary modelling allows to determine optimal working conditions or parameters which are theoretically possible, to analyse and estimate variety of different process possibilities. The most prominent advantages of the use of the models in anaerobic digestion is [7]; This reduce additional costs for continuous and repeated experiments, the possibility of saving time and money in the process of technology/process selection, rapid comparison of options and comparison of the system performance in a quantitative instead of a qualitative way allows in many cases for easier decision-making [7], monitoring parameters, possibility of minimizing risks and enhance plant efficiency. By using model, 'what if' scenarios can be examined in a quantitative way in respect of what the effects of potential risks are [7].

Biogas can be produced from co-digestion of various substrates. In the present study, anaerobic digestion of pig waste and grass clippings were studied in laboratory experiments in a 10 liters digester under constant temperature of 37 ^{0}C. The data obtained from this experiment was used to check fitness of modified Gompertz equation that well described kinetics of biogas production. Several researchers [39-44] have used modified Gompertz equation that was developed by Zwietering et al. [6] for kinetics of biogas production. Kinetics parameters A (ml/g COD)-biogas production potential, μ (ml/g COD. day)-maximum biogas production rate and λ (day)-lag phase period were estimated. The modelling of the biogas production help to analyse kinetic models and other parameters that can be used to design and scale-up of laboratory experiments into industrial size applications.

V. Methodology

Pig waste were collected from a farm in Gauteng province while grass clippings were collected from University of Johannesburg, South Africa. Waste characterization was done to ascertain the composition. These included physical and chemical composition with regards to C/N ratio, volatile solids, total solids and elemental analysis for carbon, nitrogen, sulphur and hydrogen in accordance with the standard method (APHA 1995) [45]. To determine biogas production rate, a batch digester was fed with the co-digested substrates and inoculum under pre-set conditions of 37 ^{0}C and pH of 7 as shown in Fig.2. pH was neutralized by a solution of 8g NaOH in 100 ml and H_2SO_4. The digester was flushed with nitrogen to expel the oxygen and make the process anaerobic. It was then immersed in the water bath and kept under constant temperature. The gas produced was measured using downwards displacement method on a daily basis until the end of retention time.

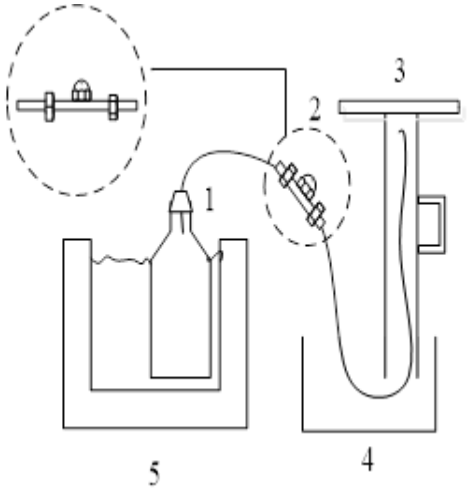

Fig. 2. Biogas Production set-up (1 Digester, 2 T-union, 3 Measuring Cylinder, 4 Water Bucket, 5 Thermostatic Water Bath)

The scope of this research was to evaluate kinetics of biogas production with regards to prediction of biogas production. Modified Gompertz equation was used in this study to model cumulative biogas production. Equation 10 shows modified Gompertz equation.

$$Y(t) = A\exp[-\exp(\frac{\mu e}{A}(\lambda - t) + 1)] \quad (10)$$

Where:

Y = Cumulative of specific biogas production (ml)

A = Biogas production potential (ml)

μ =Maximum biogas production rate (d^{-1})

λ = Lag phase period

t = Cumulative time for biogas production (days)

e = Mathematical constant (2.718282)

The kinetics constant A, μ and **λ** were determined using non-linear regression approach for the best fittings with the aid of solver command in Microsoft excel [1, 46, 47].

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VI. RESULTS AND DISCUSSION

In this study, co-digestion of pig waste and grass clippings were evaluated for the purpose of getting the bio-methane potentials and bio-chemical kinetics at optimum temperature (37 ^{0}C) and initial pH of 7. Table III shows the substrate characterization. Grass clippings were found to contain more volatile solids compared to pig waste which had more nutrients. The elemental analysis of pig waste indicated low C/N ratio compared to grass clippings. Through co-digestion, the C/N ratio increased to 17.28.

TABLE III
SUBSTRATE CHARACTERIZATION

Parameters	Grass clippings	Pig waste
C	19.1	42.26
H	1.04	0.7
N	0.93	2.62
S	0	0
TS (g)	0.88	0.77
VS (g)	0.64	0.56
TS (%)	64.08	55.7
VS (%)	87.88	76.8
C/N ratio	20.54	16.16

Where:
C – Carbon
H – Hydrogen
N – Nitrogen
S – Sulphur
TS – Total Solids
VS – Volatile Solids

TS is the sum of dissolved solids and suspended solids. TS and pH are important to assess anaerobic digestion process efficiency [14, 19]. VS is the organic portion of TS that biodegrade in anaerobic process. C/N ratio is an important factor in bacteria stability in anaerobic process. The C/N ratio required for production of biogas is from 15-30 [43, 48]. TS and VS are calculated using "(2)" and "(3)" respectively while C/N ratio is calculated using "(4)".

$$VS(\%) = \frac{M_{dried} - M_{burned}}{M_{wet}} \quad (11)$$

$$TS(\%) = \frac{M_{dried}}{M_{wet}} \quad (12)$$

Where:

M_{dried} = Amount dried sample (mg)

M_{wet} = Amount of wet sample (mg)

M_{burned} = Amount of burned sample (mg)

$$\frac{C}{N} = \frac{(F * C_F) + (S * S_f)}{(F * N_f) + (F * N_f)} \quad (13)$$

Where:

F = First substrate

S = Second substrate

C_f = Carbon composition for the first substrate

C_s = Carbon composition for the second substrate

N_f = Nitrogen composition for the first substrate

N_s = Nitrogen composition for the second substrate

A good substrate characterisation is important on modelling and especially on prediction of biogas potential from different substrates. The moisture content (MC) of substrates ranged from 55-95%. These indicated that the substrates had enough moisture content for AD. The volatile solids (VS) of substrate ranged from 55-65%. These indicated that the substrates were rich in organic solid content that was to be converted to biogas as highlighted by Zhang et al., (2012) [49]. C/N ratio was important factor in bacteria stability in anaerobic process. Higher C/N ratio is adventurous to digestion stability, high carbon content provided carbon content required for bacteria growth and hence production of biogas. The increased in carbon content gave rise to more carbon dioxide formation and lowered the pH value. Low C/N ratio indicates higher nitrogen content to carbon and thus causes ammonia accumulation. Ammonia accumulation leads to increase in pH above 8.5 which again leads to low methane production according to Mojapelo et al., (2014) [48]. The C/N ratio was from 15-30 for pig waste and grass clippings required for production of biogas.

The study of biogas production from Pig waste and grass clippings were conducted in a laboratory batch anaerobic digester. Biogas production was monitored and measured until there was no more biogas produced. The modified Gompertz model was used to fit the cumulative biogas production using non-linear regression as shown in Fig. 3.

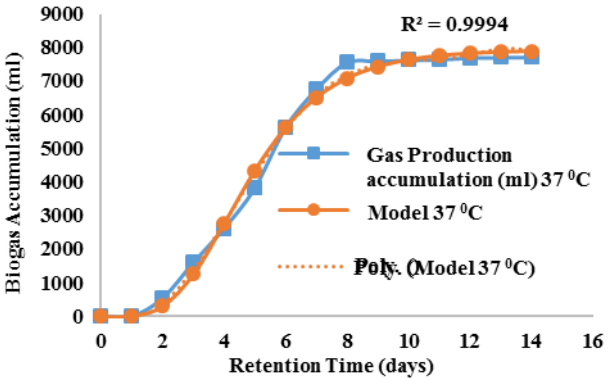

Fig. 3. Biogas Prediction using Modified Gompertz model for pig waste/grass.

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Fig. 3 shows effect of mesophilic temperature on AD 37 ^{0}C. It was observed that there was shorter lag phase which indicated the digester had essential microbes and enriched seeding (inoculum) to enhance anaerobic digestion. Between 1-10 days the rate of conversion increased with retention time. This was because with time, the conversion rate/percent of reactants to products increased. Temperature played an important role in dissociating old /reactant particle to form new species. Conversion rate increased with increased in temperature. And conversion of reactants increased with time, until an equilibrium state was reached.

The kinetics parameters evaluated are shown in Table IV. The kinetics constants A (ml/g COD)-Biogas production potential, μ (ml/g COD. day)-maximum biogas production rate, λ (day)-lag phase period were 7920.70, 701.35, 1.61 respectively with R^2- coefficient of determination of 0.9994 [1].

TABLE IV
MODIFIED GOMPERTZ PARAMETERS

Digester	Temp	Biogas Yield (ml)	Modified Gompertz parameters (model)			R^2
			A ml	**λ** (d)	μ d^{-1}	
Pig Waste Grass Clippings	37 ^{0}C	7725.0	7920.70	701.35	1.61	0.94

VII. CONCLUSION

Biogas production from co-digestion of pig waste and grass clippings was established to be feasible at a temperature of 37 ^{0}C. The application of modified Gompertz equation in studying the biogas production was able to predict biogas production with retention time.

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