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1. Introduction

Biogas produced in anaerobic digesters consists of methane (50%-80%), carbon dioxide (20%-50%), and trace levels of other gases such as hydrogen, carbon monoxide, nitrogen, oxygen, and hydrogen sulfide. Anaerobic digestion is a biological process in which organic material is decomposed by bacteria in the absence of air. The general technology of anaerobic digestion of complex organic matter is well known and has been applied for over 60 years as part of domestic sewage treatment to stabilize organic wastes. Bal & Dhagat (2001) points out that the anaerobic process is more advantageous than the aerobic process in organic waste treatment because of the high degree of waste stabilization, low production of excess biological sludge, low nutrient requirement and production of methane gas as a useful byproduct. Several studies have been carried out for evaluating kinetic parameters and model equations for anaerobic digestion by Siles et al. (2010), Borja et al. (2005), Jimenez et al. (2004), Raposo et al. (2009), Rincon et al. (2009) and Hu et al. (2002); these are all based on the Monod kinetic model (Monod 1950) and on the revised kinetic model developed by Chen et al. (1980) and Hashimoto et al. (1981).

In the microbiology of methanogenic process four different bacterial groups are identified as being responsible for carrying out the anaerobic digestion of complex organic matter. The first group of bacteria is hydrolytic bacteria which catabolizes carbohydrate, protein, lipid and other minor components of organic matter to fatty acids, $\text{H}_2$ and $\text{CO}_2$. The second group of bacteria is hydrogen producing acetogenic bacteria which catabolizes certain fatty acids and neutral end products to acetate, $\text{CO}_2$ and $\text{H}_2$. The third group of bacteria is homoacetogenic which synthesizes acetate using $\text{H}_2$, $\text{CO}_2$ and formate, and hydrolyzes multcarbone compound to acetic acid. Finally, the fourth group of bacteria i.e. methanogenic bacteria utilizes acetate, carbon dioxide and hydrogen to produce methane. The concerted action of these four bacterial groups ensures process stability during anaerobic digestion of the complex organic matter.

The reactions involved in these steps are given below:

- **Phase-I. Solubilization of carbohydrate via hydrolysis**
  \[ [\text{C}_n\text{H}_{2n}\text{O}_x]_n + n\text{H}_2\text{O} = n\text{C}_n\text{H}_{2n}\text{O}_6 \]  

- **Phase-II. Acidogenesis fermentation of glucose to acetate**
\[ \text{C}_6\text{H}_{12}\text{O}_6 + 2\text{H}_2\text{O} = 2\text{CH}_3\text{COOH} + 4\text{H}_2 + 2\text{CO}_2 \]  
(2)

- Phase-III. Methanogenic reaction

\[ \text{CH}_3\text{COOH} = \text{CH}_4 + \text{CO}_2 \]  
(3)

\[ 4\text{H}_2 + \text{CO}_2 = \text{CH}_4 + 2\text{H}_2\text{O} \]  
(4)

### 1.1 Chen–Hashimoto kinetic model of anaerobic digestion

Chen–Hashimoto model was used for kinetic analysis of the experimental data. In a completely mixed continuous digester the rates of change of cell mass and substrate concentration are expressed by the following equations:

\[
\frac{dX}{dt} = \mu X - \frac{X}{\theta} \quad (5)
\]

\[
\frac{dS}{dt} = -r + \frac{S_0 - S}{\theta} \quad (6)
\]

Where

- \( X \) is the concentration of cell mass
- \( \mu \) the specific microbial growth rate
- \( \theta \) the hydraulic retention time
- \( S_0 \) the concentration of substrate in the influent,
- \( S \) the concentration of substrate in the effluent
- \( r \) is the volumetric substrate utilisation rate

The relationship between \( r \) and \( \mu \) is defined by the following equation:

\[
\mu = \frac{Yr}{X} \quad (7)
\]

Where

- \( Y \) is the yield coefficient (cell mass/substrate mass) and is considered constant (Chen & Hashimoto, 1978). In the steady-state, \( \frac{dX}{dt} = 0 \) and \( \frac{dS}{dt} = 0 \), hence

\[
\mu = \frac{1}{\theta} = D \quad (8)
\]

Where

- \( D \) is the dilution rate

\[
r = \frac{S_0 - S}{\theta} \quad (9)
\]

and

\[
X = Y(S_0 - S) \quad (10)
\]

Substituting these expressions in Contois’ equation:

\[
\mu = \frac{\mu_{\text{max}}S}{\mu_{\text{max}} + S} \quad (11)
\]
Kinetics of Biogas Production from Banana Stem Waste

Where
\[ \mu_{\text{max}} \] is the maximum specific microbial growth rate
\[ \beta \] is a dimensionless kinetic parameter

\[ \frac{S}{S_0} = \frac{K}{\mu_{\text{max}} \theta - 1 + K} \]  \hspace{1cm} (12)

Where
\[ K \] is an dimensionless kinetic parameter.

Eq. 12 shows that effluent substrate concentration depends on the influent substrate concentration.

The minimum retention time indicating when the washout of micro-organisms occurs is numerically equal to the reciprocal of the maximum growth rate:

\[ \theta_{\text{min}} = \frac{1}{\mu_{\text{max}}} \]  \hspace{1cm} (13)

There are two different approaches generally used to study the kinetics of biogas production of lignocellulosic waste: one approach is to find the rate-limiting substrate for the kinetic evaluation; another approach is using chemical oxygen demand or volatile solids concentration as an indicator of the substrate concentration (Chen & Hashimoto, 1978).

There are difficulties in using COD or VS as the gross substrate since a portion of the COD or VS is not available to the microbes as substrate. The laboratory test for COD of high strength residues requires at least 100 times dilution which generally yields unreliable data. Also, some of the volatile acids in the effluent are volatilised during the VS determination. Because the volatile acids are precursors of biogas production, their volatilisation during the VS determinations causes errors in the calculated amount of substrate utilised.

Biogas production is directly correlated with COD reduction. Since no oxidising agent is added, the only way COD reduction can occur is through the removal of organic material from the waste, such as through the evolution of methane and carbon dioxide. The other avenues of COD reduction through hydrogen sulpsulphide and hydrogen gas evolution are insignificant (Chen & Hashimoto, 1978). A reduction of 1 g COD is equivalent to the production of 0.35 l of methane at STP. Knowing the COD loading to the reactor and the volume of methane produced, the remaining COD in the digester can be calculated.

The biodegradable COD in the reactor will be directly proportional to \( (B_0 - B) \) where \( B \) denotes the volume (in litres) of methane produced under normal conditions of pressure and temperature per gram of substrate (COD) added to the digester and \( B_0 \) is the volume of methane produced under normal conditions of pressure and temperature per gram of substrate added at infinite retention time or for complete utilization of substrate and \( B_0 \) will be directly proportional to the biodegradable COD loading (Chen & Hashimoto, 1978). Therefore, from Eq. (12) one obtains:

\[ \frac{B_0 - B}{B_0} = \frac{K}{\mu_{\text{max}} \theta - 1 + K} \]  \hspace{1cm} (14)

From Eq. (14) one obtains:

\[ \theta = \frac{1}{\mu_{\text{max}}} + \frac{K}{\mu_{\text{max}} (B_0 - B)} \]  \hspace{1cm} (15)
Thus, by first calculating the value of $B_0$, the graph of $\theta$ versus $B/(B_0-B)$ produces a straight line with an intercept of $1/\mu_{max}$ and with a slope of $K/\mu_{max}$. To obtain the parameter $B_0$ one uses the following equation, which is easily derived from Eq. (14):

$$B = B_0 \left| 1 - \frac{K}{\mu_{max} \theta - 1 + K} \right|$$  (16)

Since $B$ is the methane production per gram of added COD, the volumetric methane production rate ($\delta$) equals $B$ multiplied by the loading rate:

$$\delta = \frac{BS}{\theta} = \frac{B_0 S_a}{\theta} \left| 1 - \frac{K}{\mu_{max} \theta - 1 + K} \right|$$  (17)

Where $\delta$ has the dimensions of volume methane per volume digester per unit time.

The objective of the present study is to develop kinetic parameters for two-stage biogas production using banana stem waste as substrate.

2. Materials and methods

2.1 Acclimatization

In the acclimatization step, soil sludge containing mixed culture from soil (MCS) will be collected from banana plantation soil using polyvinyl chloride pipe in 10 cm depth from surface to make sure the anaerobic mixed culture available in the sample. The end of the pipe is then closed with rubber stopper and need to be process in 4 hours after collection was made.

Subsequently, 20 ml of the soil sludge is put into each of 20 serum bottles that contained 20 mg of the banana stem waste (BSW). The bottle is then closed with bottle cap or rubber stopper and flushed with nitrogen for 5 minutes. The flushed bottle is then incubated in anaerobic container in ambient temperature (28°C to 30°C) and dark condition for a month. Then, the content in all serum bottles are put into 1 liter anaerobic container which contained of 10 g of BSW. The 1 litre anaerobic container is then closed and flushed with nitrogen for 5 minutes and left for incubation in ambient temperature (28°C to 30°C) for two months.

Afterward, the materials in the 1 litre anaerobic container are put into two anaerobic container (5 litres) which each of container contained 50 g of BSW. It was found by the inventors of the present invention that acclimatization increased the microbe amount in reactor.

Subsequently, the acclimatized MCS will be used as inoculum for biogas production. Biogas production will be done in 10 L anaerobic fed-batch bioreactor with gas outlet (Fig. 1). Bioreactor equipped with temperature controller and agitator to ensure the inoculum and BSW mixed evently. To maintain anaerobic condition in the reactor nitrogen will be purged everytime the inlet and outlet were done. 5000 mg/l inoculum mixed with BSW at HRT of 12 d and OLR 1.5 gTS/l.d and ambient temperature (28°C to 30°C) for biogas production.
2.2 Experimental set-up

All experiments were done in 20 L anaerobic sequencing batch reactor followed by 10 L fixed bed reactor with gas outlet (Fig. 1). All the reactors were seeded with anaerobic acclimatized banana stem sludge. The anaerobic digestion system was varied at different reaction temperatures using water bath. The HRT and OLR for this system were 9 d and 4 g TS/Ld respectively. The process was conducted at ambient temperature for the first stage and thermophilic temperature for the second stage. Daily withdrawal of an appropriate volume from the reactor corresponding to the determined HRT or OLR was done by a draw-and-fill method. Biogas evolved from the fixed bed reactor was measured and collected in a gas holder by water displacement. Samples were collected and analyzed for performance evaluation.
2.3 Two-stages biogas production system description

2.3.1 Bioreactor description

This system consists of 4 components which are hydrolysis reactor, liquid-solid separator, storage tank and methanogenesis reactor. The dimensions of those four components are listed in Table 1. Detailed of each component are as follows:

<table>
<thead>
<tr>
<th></th>
<th>CRR</th>
<th>Solid-liquid separator</th>
<th>Storage tank</th>
<th>BPR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume (l)</td>
<td>20</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Diameter (in)</td>
<td>12</td>
<td>9</td>
<td>10</td>
<td>Upper-10 Lower-6</td>
</tr>
<tr>
<td>Length (in)</td>
<td>15</td>
<td>16</td>
<td>10</td>
<td>35</td>
</tr>
<tr>
<td>Maximum pressure(bar)</td>
<td>2</td>
<td>2</td>
<td>-</td>
<td>2</td>
</tr>
<tr>
<td>Relieve</td>
<td>0.2</td>
<td>0.2</td>
<td>-</td>
<td>0.2</td>
</tr>
</tbody>
</table>

Table 1. Component dimension in two-stages biogas production

2.3.2 Hydrolysis reactor

Influent for the hydrolysis reactor is banana stem waste slurry. The type of reactor is anaerobic batch reactor. Reactor volume is 20 litre. Initial concentration of the biomass in teh reactor is 5000 mg/l which is mixed culture from acclimatized banana plantation soil. Inlet and outlet from this reactor is drawn manually (Scharer et al. 1981; Gavala et al. 2003).

2.3.3 Solid-liquid separator

The second component is solid-liquid separator. The tank volume is 10 litre. The function of this tank is to separate the solid and liquid from CRR effluent. The effluent from CRR will be sediment at the bottom of conical shaped separator tank. Sludge from separator will be recycled back to CRR and the liquid from separator will be transferred to storage tank (Batstone et al. 2002).

2.3.4 Storage tank

The storage tank with 10 litre function as storage for BPR influent.

2.3.5 Biogas Production Reactor (BPR)

BPR with 10 litre volume function as biogas production reactor. This reactor is anaerobic fixed bed reactor and contained plastic media for biomass support. The initial biomass concentration in the reactor is 5000 mg/l which is acclimatized mixed culture from banana plantation soil.

2.4 Analytical methods

COD concentration was spectrophotometrically analyzed using a HACH spectrophotometer and methods as in Spectrophotometric Instrument Manual. Gas
collection was done using daily water displacement. Methane content was analyzed using gas chromatography with thermal conductivity detector (GCTCD) with helium as the carrier gas. Acetic acid concentration (TVA) was determined using HPLC. Substrate concentration was measured as suspended solids according to the Standard Methods for The Examination of Water and Wastewater. 20 ml well-mixed sample was filtered through a weighed standard glass-fiber filter and the residue retained on the filter is dried to a constant weight at 103°C to 105°C. The increase in weight of the filter represents the total suspended solids (APHA 1989).

3. Results and discussion

Chen et al. (1978) developed a kinetic model on substrate utilization based on the Contois model as follows:

\[
\frac{\mu_{\text{max}}}{\mu} = K \frac{S_{a}-S}{S} + 1
\]  

(18)

The kinetic model has been aptly used in many studies, notably in investigations on anaerobic digestion of high strength wastes (Mata-Alvarez et al. 1992; Sales et al. 2000).

Equation (18) can be written as:

\[
\frac{1}{\mu} = \frac{1}{\mu_{\text{max}}} + \frac{K}{\mu_{\text{max}}} \frac{S_{a}-S}{S}
\]

(19)

For a completely mixed system \(1/\mu = \theta\) and \(1/\mu_m = \theta_m\). Therefore

\[
\theta = \frac{1}{\mu_{\text{max}}} + \frac{K}{\mu_{\text{max}}} \frac{S_{a}-S}{S}
\]

(20)

here

\(S\) is substrate total effluent

The kinetic parameters, \(\mu_{\text{max}}\) and \(K\), were calculated with the aim of studying possible inhibition phenomena. Using the least squares method, the values for the kinetic parameters \(\mu_{\text{max}}\) and \(K\) can be obtained from the intercept and the slope of the adjusted lines. Thus, according to Eq. 20, \(\mu_{\text{max}} = 1/\text{intercept}\), and \(K = \text{slope/intercept}\). Through linear regression \(T\) vs \(S\) value of \(\mu_{\text{max}}\) and \(K\) could be determined (Fig. 2). Here

\(T=\theta\)

and

\(S = S_0 - S/S\).

In this study the value of \(\mu_{\text{max}}\) and \(K\) calculated were 0.111 d\(^{-1}\) and 0.330 g/g respectively.

The kinetics of methane fermentation as proposed by Chen and Hashimoto (1978) is described by

\[
\delta = \frac{R_{0}S_{a}}{\theta} \left[ 1 - \frac{K}{\mu_{\text{max}}(0-1+K)} \right]
\]

(21)

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Equation (21) states that for a given substrate loading rate ($S_0/\theta$), the daily volumetric methane production depends on the biodegradability of the wastewater ($B_0$) and the kinetic parameters $\theta$ and $K$ (Yeoh, 1997). These values are calculated from the values of methane volume (Table 2), taking into account that the influents used have a COD of 2000 mg/l. Least squares method were used to determine the intercept $B_0$. Through linear regression $Y$ vs $St$ the value of $B_0$ could be determined. Here $Y=\delta$ and

$$St=\frac{S_0}{\theta} \left[ 1 - \frac{K}{\mu_{\text{max}} \theta - 1 + K} \right].$$

Fig. 3 shows the regression to determine $B_0$. The value of $B_0$ from this study is 0.326 l methane/g COD.
### Table 2. Values of Y and St for linear regression to determine $B_0$

<table>
<thead>
<tr>
<th>St</th>
<th>Y</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.033</td>
<td>0.085</td>
</tr>
<tr>
<td>0.028</td>
<td>0.081</td>
</tr>
<tr>
<td>0.047</td>
<td>0.088</td>
</tr>
<tr>
<td>0.090</td>
<td>0.101</td>
</tr>
<tr>
<td>0.068</td>
<td>0.090</td>
</tr>
<tr>
<td>0.081</td>
<td>0.100</td>
</tr>
<tr>
<td>0.068</td>
<td>0.100</td>
</tr>
<tr>
<td>0.068</td>
<td>0.101</td>
</tr>
<tr>
<td>0.071</td>
<td>0.098</td>
</tr>
<tr>
<td>0.046</td>
<td>0.090</td>
</tr>
<tr>
<td>0.051</td>
<td>0.092</td>
</tr>
<tr>
<td>0.105</td>
<td>0.107</td>
</tr>
<tr>
<td>0.105</td>
<td>0.109</td>
</tr>
</tbody>
</table>

Fig. 3. Determination of $B_0$

\[
y = 0.3263x + 0.074
\]

\[
R^2 = 0.9026
\]
The kinetic values corresponding to the substrate used (banana stem waste) is of the same order of magnitude with those obtained in the mesophilic anaerobic digestion process of some solid wastes such as vegetable waste, banana peel and palm oil (Table 4). From the kinetic constants (K, $\mu_{max}$ & B₀), the theoretical daily volumetric methane production values ($\delta$) were calculated by using Eq. (21). It can be seen that the experimental results were reproduced with errors equal to or less than 5% in all cases. As can be seen in Table 3 and Fig. 4, the values of $\delta$ increased when influent substrate concentration (COD in) increased. Therefore, the kinetic parameters were found to be influenced by the influent substrate concentration. When the influent substrate concentration increased from 0.835 to 2.463 g COD/l, the $\delta$ values also increased from 0.085 to 0.109 l methane/g COD. The mixed culture in biomass also gives effect to methane production. As can be seen in Table 3, when the biomass in the bioreactor concentration (X) increased from 0.23 g/l to 0.93 the $\delta$ increased from 0.085 to 0.109 was, therefore, multiplied by a factor of 1.3. A similar behaviour was observed in the anaerobic digestion process of traditional olive mill wastewaters (Borja et al., 1995). In this work, the minimum hydraulic retention time, $\theta_{min}$ (days), at which the washout of the micro-organisms occurs was: 9.04 days. These values were calculated by using Eq. (13) and taking into account that this retention time is numerically equal to the reciprocal of the maximum micro-organisms growth rate ($\mu_{max}$).

Fig. 4. Graph for influent substrate concentration (COD in), biomass in bioreactor concentration (X) and biomass in effluent concentration (Xe)
### Table 3. Values of influent substrate concentration (COD in), biomass in bioreactor concentration (X), biomass in effluent concentration (Xe) and daily volumetric methane production (δ)

<table>
<thead>
<tr>
<th>Study</th>
<th>Substrate</th>
<th>B₀ (l methane/g COD)</th>
<th>K (g/g)</th>
<th>μ_max (day⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>This study</td>
<td>Banana stem waste</td>
<td>0.326</td>
<td>0.330</td>
<td>0.111</td>
</tr>
<tr>
<td>Gunaseelan (2004)</td>
<td>Banana peel</td>
<td>0.277*</td>
<td>-</td>
<td>0.089</td>
</tr>
<tr>
<td>Faisal &amp; Unno (2001)</td>
<td>Palm oil mill wastewater</td>
<td>0.381</td>
<td>-</td>
<td>0.304</td>
</tr>
<tr>
<td>Gunaseelan (2007)</td>
<td>Banana peel</td>
<td>0.322*</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Zhao &amp; Viraraghavan (2004)</td>
<td>Treatment plant wastewater</td>
<td>0.366</td>
<td>0.079</td>
<td>1.16</td>
</tr>
<tr>
<td>Maya-Altamira et al (2008)</td>
<td>Vegetable product-peas &amp; fried onion</td>
<td>0.36</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Vegetable product-leek</td>
<td>0.36</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

*1 methane/g VS added

### Table 4. Comparison of kinetics parameter to other research

The values of B₀, μ_max and K is fixed for different type of wastewater. So the Eq. (21) and values of B₀, μ_max and K could be used in scaling up biogas production process. The values of B₀, μ_max and K was compared to other research in Table 4. B₀ is a good parameter to determine biodegradability of any particular waste (Torres-Castillo et al. 1995). From Table 1 it was shown that banana stem waste from this research have a good methane production potential and comparable with other agricultural waste such as POME and sugar cane waste. μ_max is microorganism maximum growth rate and the value of μ_max in this study is...
considered low compared to other research. This is because the value of kinetic parameter $\mu_{\text{max}}$ and $K$ is dependent on each other. High value of $K$ could lower down the value of $\mu_{\text{max}}$ and vice versa. However the value of $\mu_{\text{max}}$ reported in this research is still within acceptable range.

4. Conclusion

A kinetic model for studying the anaerobic digestion process of banana stem waste was proposed on the basis of the two-stages process data obtained. The two-stages system comprising two bioreactors for acidogenic and methanogenic phases respectively. The experiments were conducted with hydraulic retention time (HRT) of 9 d corresponding to organic loading rate (OLR) of 4 gTS/l.d. The parameters obtained represent and predict the activity of the mixed culture in the biogas production of this waste. Kinetic evaluation of the experimental data provided $\mu_{\text{max}}$ (maximum microorganism growth rate) and $K$ (kinetic constant) values as 0.111 d$^{-1}$ and 0.330 g/g respectively based on COD. The waste biodegradability ($B_0$) was graphically evaluated to be 0.326 l methane/g COD.

5. References


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This book contains research on the chemistry of each step of biogas generation, along with engineering principles and practices, feasibility of biogas production in processing technologies, especially anaerobic digestion of waste and gas production system, its modeling, kinetics along with other associated aspects, utilization and purification of biogas, economy and energy issues, pipe design for biogas energy, microbiological aspects, phyto-fermentation, biogas plant constructions, assessment of ecological potential, biogas generation from sludge, rheological characterization, etc.

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