Anaerobic Digestion of Vegetable Wastes for Biogas Production in Single Chamber and Double Chamber Reactors
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Abstract
The compositions of solid wastes of a rural market of Bangladesh and the results of two sets of laboratory experiments on biogas generation from the easily biodegradable wastes under daily feed condition are presented in this paper. Cow dung, cauliflower stick, papaya and potato were the major biodegradable wastes. Daily average composition of the biodegradable wastes was used in the experiments. The average Total Solids (TS) and Volatile Solids (VS) concentrations of the raw substrate were determined as 18.90% and 15.10% respectively. The experimental setups were placed in a large closed chamber containing two room heaters. The room heaters were operated alternatively at 35°C to maintain a favorable condition for anaerobic digestion of the substrate. In the first setup, a single chamber reactor and a double chamber reactor were used. In the single chamber reactor, 750 g wastes and required amount of inoculum were added initially to make the effective volume of 2 L. For the double chamber reactor, each chamber was initially fed with 350 g wastes and inoculum was added to make the effective volume of 1 L. The single chamber reactor was operated for 58 days and the double chamber reactor was run for 23 days. From the 2nd day of operation, each reactor was fed daily with a mixture of 18.75 g wastes and the required volume of tap water to make the total volume of 50 mL after taking out equal volume of slurry from the reactor. The second set of experiment was similar to the double chamber reactor of the first setup, but it was operated for 54 days including the last 16 days operation at room temperature as the heaters became out of order. In case of the first setup, the temperature varied from 31°C to 36°C and the rate of biogas generation was not affected due to this variation. The results of the experiments revealed that for the Organic Loading Rate (OLR) of 1.42 g VS/L/d, the daily stable biogas generation rate was 0.22 m³/kg of VS added for the single chamber reactor, and apparently the daily stable average rate of biogas production was 0.37 m³/kg of VS added for the double chamber reactor. During the second set of experiment, the temperature varied in between 32°C and 36°C when the chamber-heater was on and the rate of biogas generation was not affected, and the stable rate of biogas generation was 0.26 m³/kg of VS added for the OLR of 1.42 g VS/L/d. The temperature of the chamber varied from 22°C to 25°C when the heater became out of order and the sudden drop of the temperature by about 10°C affected the rate of biogas production greatly. At the ambient temperature, the stable rate of biogas generation was only 0.08 m³/kg of VS added.

Keywords: Anaerobic digestion; Biodegradable waste; Biogas; Hydraulic retention time; Single chamber reactor; Double chamber reactor; Organic loading rate

Introduction
Cities and rural growth centers are places of rapid economic growth, trade, education and employment resulting in increasing the consumption of resources and generation of wastes. The world is facing the burning problems of management of large quantities of solid wastes produced in these places and meeting the energy requirements due to rapid growth of urban population. The vegetable markets of these areas produce large quantities of biodegradable wastes which are very poorly managed in developing countries producing malodorous gases, greenhouse gases, and leachate during on-site degradation and pollute the water when the wastes are thrown into local water bodies. Hence, the wastes should be properly managed to have clean environment and reduction of greenhouse gas emission to slow down the climate change. The high moisture and Volatile Solids (VS) contents in vegetable wastes make these more suitable for anaerobic digestion than incineration and composting. Anaerobic digestion of solid wastes is becoming popular day by day as a method of solid waste management as it produces biogas which can be used for steam heating, cooking and generation of electricity [1-4]. The residual slurry can be used as a bio-fertilizer and soil conditioner [5].

Huge amount of biodegradable solid wastes are generated in the markets of Bangladesh and these are the potential sources of biogas generation. An investigation on the type and quantity of solid wastes produced in a rural vegetable market of Bangladesh was carried out, and the biogas generation from the wastes was quantified through laboratory scale single chamber and double chamber reactors under daily feed condition at controlled temperature and sharp decrease in ambient temperature. The investigation was done to compare the results with those found in the available literature and to see the effect of the change in the ambient temperature on the gas generation. In addition, finding the difference in biogas production between single chamber and double chamber reactors were the objective as short circuiting of the added wastes is likely to happen in case of single chamber reactor. This paper presents the results of the investigation.

Anaerobic digestion process
Anaerobic digestion is a complex fermentation process brought about by the symbiotic association of different types of bacteria with ultimate products being mainly methane and carbon dioxide [6-9]. The products generated by one group of bacteria serve as substrates for the

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next group. The general process of anaerobic digestion is a series of four metabolic processes namely hydrolysis, acidogenesis, acetogenesis, and methanogenesis shown in Figure 1 [10].

In the hydrolysis step, large protein macromolecules, fats and carbohydrate polymers (such as starch, cellulose, hemicellulose, pectin, and lignin) are broken down or depolymerized through hydrolysis by extracellular enzymes excreted by fermentative microorganisms to amino acids, glycerol, long-chain fatty acids, and sugars before being taken up by acidogenic bacteria. Hydrolysis is generally a relatively slow step and it can limit the rate of the overall anaerobic digestion process, especially when using solid wastes as the substrate. The rate of hydrolysis is a function of pH, temperature, composition, particle size of the substrate and concentrations of intermediate products [11].

In acidogenesis step, the products of hydrolysis are converted by acid-forming bacteria to higher organic acids such as propionic, lactic, butyric, and acetic and valeric acids, hydrogen and carbon dioxide [12]. In acetogenesis step, these fermentation products are transformed to acetic acid, carbon dioxide, and hydrogen by acetogenic bacteria. The hydrogen gas produced in acetogenesis step can inhibit the metabolism of aceticogenic bacteria if it is not consumed by methane-producing bacteria functioning as hydrogen-scavengers to generate methane [13].

Methane is formed by methanogenic bacteria in the last step. Formic acid, acetic acid, methanol, carbon monoxide, carbon dioxide and hydrogen are metabolized by these bacteria to methane. As the methanogenic bacteria are slow growing and extremely sensitive to changes in the environment and can assimilate only a narrow array of relatively simple substrates, they are crucial to anaerobic digestion process. The anaerobic degradation of cellulose-poor wastes like fruit and vegetable wastes is limited by methanogenesis rather than by hydrolysis [14,15] as they are very rapidly acidified to Volatile Fatty Acids (VFA) and tend to inhibit methanogenesis when the feedstock is not adequately buffered [16].

Typical chemical reactions involved in the four steps of anaerobic digestion of complex wastes can be given as follows [17]:

### Hydrolysis

\[ C_{6}H_{10}O_{5} + 2H_{2}O \rightarrow C_{6}H_{12}O_{6} + H_{2} \]

### Acidogenesis

- **Acetotrophic methanogenesis:** \( CH_{3}COOH \rightarrow CO_{2}+CH_{4} \)
- **Hydrogenotrophic methanogenesis:** \( CO_{2}+4H_{2} \rightarrow CH_{4}+2H_{2}O \)
- **Methylotrophic methanogenesis:**
  - \( CH_{3}OH+CO_{2} \rightarrow CH_{4}+CH_{3}COOH \)
  - \( 2CH_{3}CH_{2}OH+CO_{2} \leftrightarrow 2CH_{3}CH_{2}COOH+2H_{2}O \)

### Acetogenesis

- \( CH_{3}CH_{2}COOH + 2H_{2}O \leftrightarrow CH_{3}COOH + CO_{2} + 3H_{2} \)
- \( CH_{3}CH_{2}COOH + 3H_{2}O \leftrightarrow CH_{3}OH + CO_{2} + H_{2} + HCO_{3}^{-} + 3H_{2} \)
- \( CH_{3}CH_{2}CH_{2}COOH + 2H_{2}O \leftrightarrow 2CH_{3}COOH + 2H_{2} \)

### Methanogenesis

The pathways along with the stoichiometries of the overall chemical reactions are:

- **Acetotrophic methanogenesis:** \( CH_{3}COOH \rightarrow CO_{2}+CH_{4} \)
- **Hydrogenotrophic methanogenesis:** \( CO_{2}+4H_{2} \rightarrow CH_{4}+2H_{2}O \)
- **Methylotrophic methanogenesis:**
  - \( CH_{3}OH+CO_{2} \rightarrow CH_{4}+CH_{3}COOH \)
  - \( 2CH_{3}CH_{2}OH+CO_{2} \rightarrow CH_{3}CH_{2}COOH \)

As the reactions in the anaerobic digestion process take place sequentially all products of a previous metabolic stage are converted into the next one without significant buildup of intermediate products in a well-balanced digestion process, and the anaerobically biodegradable organic matter is converted nearly completely into end products like methane, carbon dioxide, hydrogen sulfide and ammonia [18].

### Materials and Methods

#### Selected market and composition of wastes

Tekerhat Shangkardirpar bazaar (a very large size market) of Rajoir upazilla of Madaripur district of Bangladesh was selected for the study. It consists of about 700 shops (400 are permanent) and a slaughter house. During haat days (Saturday, Wednesday) the number of temporary vegetable shops is about 200. There are about 200 carpentry shops, 5 restaurants and 6 tea-stalls in the bazaar. Haat days are typically one or two days of every week when large number of people come to rural or semi-urban bazaars (markets) with their commodities for sale. People also make greater quantity of purchase on the haat days.

A field survey was conducted to acquire the composition of wastes produced on a haat day and a normal day. Two labors were engaged to collect and gather all the wastes of each particular day. Then each type of the wastes was separated manually and measured by an electronic weighing scale using a 30 L bucket. The weight of every item was recorded individually to know the composition of the wastes and ensure that representative sample is used to conduct laboratory experiments.

The amount of total wastes generated on a haat day and a normal day obtained from the survey at Shangkardirpar bazaar are shown in Table 1.

It can be seen from Table 1 that the quantity of wastes generated on a haat day was more than 3 times of the amount produced on a normal day. A total of 480 kg waste was produced on a haat day of which 254 kg...
(53%) was biodegradable. On a normal day 152 kg waste was produced of which 89 kg (58%) was biodegradable. Cow dung, cauliflower stick, dry paddy straw, banana leaves, papaya, and potato were the major biodegradable wastes. Garlic and onion peel, paperboard and polythene were the major non-biodegradable wastes. Cow dung possessed about an average of 40% of the total biodegradable waste.

The experiments were conducted using the same composition of the biodegradable portion of the market wastes except the dry paddy straw. When it was included as substrate along with other biodegradable market wastes during the preliminary experiment in the laboratory for biogas generation, it was found that the dry paddy straw was very poorly degraded in the biogas digester even after a very long digestion period [19]. Daily average composition of the biodegradable wastes was determined on the basis of the average waste generation on the haat day and normal day of a week and was used as the substrate of the experiments (Table 2). Calculation shows that 48% of the total daily average wastes were easily biodegradable.

The wastes were cut into small pieces (maximum dimension of 4 mm) and were mixed thoroughly to obtain a uniform mixture. Fresh wastes were collected once per week and were preserved in a refrigerator after the sizing. Daily feeding was done with this sample only for the particular week. During the experiments, the TS (Total Solids) and VS (Volatile Solids) of the substrate were determined for only for the particular week. During the experiments, the TS (Total Solids) and VS (Volatile Solids) of the substrate were determined for the experiments (Table 2). Calculation shows that 48% of the total daily average wastes were easily biodegradable.

Experimental setup

Two sets of laboratory experiments were run in semi-continuous feeding mode (one-time daily feeding) of the anaerobic digesters/reactors. Suitable arrangements for feeding, biogas collection and draining of residues was made for each reactor. A closed chamber made of Thai Aluminum was placed on a wooden table to run the experiments. To maintain a constant temperature within the chamber, two electrical room heaters were placed inside it. One single chamber reactor and one double chamber reactor were operated in the first setup. The heaters were operated alternatively setting the temperature at 35°C. In case of the single chamber reactor, a 5 L capacity plastic container was initially loaded with 750 g wastes and 1300 mL inoculum (collected from a field biogas plant) was added to make the effective volume of 2 L. To measure the generated biogas, water displacement mechanism from an inverted cylinder was used (Figure 2).

Two plastic containers connected in series, each having a capacity of 1.5 L were used as double chamber reactor. The interconnection was made near the bottom so that solids accumulated at the bottom of the inlet container could move easily to the outlet container. Initially, each container was fed with 375 g wastes and 650 mL inoculum to make the effective volume of the reactor of 2 L. The biogas generated in each container was measured using the same method of the single chamber reactor. The setup is shown in Figure 3.

The second set of experiment was a duplication of the semi-continuous feeding double chamber reactor as the data on gas production during the initial days of the reactor were apparently somewhat inconsistent. So, the same setup was used but it was operated for a much longer period (54 days).

Operation of anaerobic digesters

The single chamber reactor was operated from 18th August to 14th October, 2016 (total 58 days of operation). Since from the batch studies conducted earlier [19], optimum HRT was found to be 40 days, daily feeding was calculated based on this. Accordingly, from the 2nd day of operation, 50 mL of slurry was taken out from the reactor and then 50 mL mixture of 18.75 g wastes and tap water was added to the reactor daily at a particular time. The biogas production and the temperature within the enclosure were recorded daily.

Operation of the double chamber reactor was started on 21st...
September and continued up to 14th October, 2016 (total 23 days of operation). The daily feeding was started on the 2nd day of the operation and the procedure was similar as that of the single chamber reactor i.e. taking 50 mL slurry out from the outlet chamber and adding 50 mL mixture of 18.75 g wastes and tap water. On each day of the operation period, the biogas generated in each chamber of the reactor was measured separately and the temperature within the enclosure was recorded.

The second set of experiment was started on 26th October and continued up to 18th December, 2016 (total 54 days of operation). As described above, the daily feeding was started on the 2nd day of operation taking 50 mL slurry out from the outlet chamber and adding 50 mL mixture of 18.75 g wastes and tap water. It is to be noted that at the end of the 2nd December, 2016, the heater was out of order and the experiment was continued at the room temperature to see the effect of sudden fall of temperature. On each day of the operation period, the biogas generated in each chamber of the reactor was measured separately and the temperature within the enclosure was recorded.

Methods of measurement

The biogas generated was measured daily by inserting the gas outlet tube into an inverted measuring cylinder filled with water and placed in a water jar. The gas produced displaced the water from the measuring cylinder and the displaced volume was recorded daily. The inverted cylinder was refilled as and when needed. The temperature displayed by the heater was noted several times daily and the average value was determined. The TS and VS of the wastes were determined according to the APHA standard methods [20].

Results and Discussion

The TS and VS of the biodegradable portion of the market wastes were determined three times and the results are presented in Table 3 below.

The average of the three measurements is found to be 18.90% for TS and 15.10% for VS. Calculations for all the experiments were based on these average values. The variations in TS and VS measurements were due primarily to sampling cow dung from the stomach of slaughtered cow and actual cow dung.

In the first setup, the single chamber reactor was operated for 58 days with 750 g waste in the reactor and the double chamber reactor was run for 23 days with a total of 750 g wastes in the two chambers with one-time daily feeding.

The change in the daily gas generation in the single chamber reactor and the daily average temperature with time are shown in Figure 4.

It is observed that during the first two days the gas generation per day was very high owing to availability of high amount of easily biodegradable organic matter (cow dung) in the reactor. Then a sharp drop was noticed in the daily gas production and the drop became less prominent for the next few days. Then the gas generation fluctuated significantly at times for the next 22 days. It is seen that a more or less stable condition was obtained after 40 days of operation, and it continued up to the end of the experiment. The initial feed, the biodegradable matter present in the added inoculum and the daily feed had pronounced effect on the rate of biogas generation initially and then the effect was slowly disappeared and a stable condition was attained. The average daily temperature was within 32°C to 36°C and this small variation had no effect on the rate of gas generation. The stable rate of gas generation is found to be about 625 mL per day (0.31 m³/m²/d). The stable gas yield was 0.22 m³/kg of VS. It is considerably lower than the rate of gas generation (0.35 m³/kg of VS) under batch study for the same OLR (1.42 g VS/L/d) [21]. The reason is the effect of presence of high concentration of biodegradable organic matter in the inoculum used and it was nullified at the steady state condition (Figure 5).
It is seen that after a fluctuation in gas production during the initial days, the gas production was more or less stable for the rest of the days of operation and the difference in gas production between the two chambers were negligible. The inlet chamber produced a slightly higher amount of gas due to addition of fresh feed to this chamber only. The gas production during the later periods was 550 mL/day for the inlet chamber and it was 500 mL/day for the outlet chamber. The fluctuation in the temperature of the enclosure was from 33°C to 36°C did not affect the gas generation. The apparent stable rate of gas generation was found to be about 0.28 m³/m³/d and 0.25 m³/m³/d for it the inlet chamber and outlet chamber respectively. The corresponding gas yield was 0.19 m³/kg of VS and 0.18 m³/kg of VS respectively. As the experiment was run only for 23 days, much higher rate of stable gas generation was achieved compared to the of the single chamber reactor.

The comparison between gas production between the single chamber reactor and double chamber reactor is shown in Figure 6. It is evident that the gas production in the two systems differed significantly during the initial 15 days– the single reactor producing much higher quantity of gas in general, but then the difference became insignificant. It appears that the inoculum added to the double chamber reactor had less concentration of biodegradable organic matter compared to that in the inoculum added to the single chamber reactor.

However, it can be predicted that if the operation of double chamber reactor would continue for the same period of time (58 days), comparable results would be obtained.

In the second setup, the double chamber reactor was run for 54 days with a total of 750 g wastes in the two chambers with one-time daily feeding. The variations in gas generation in each chamber and the daily average temperature with date of operation are depicted in Figure 7.

From Figure 7, it is revealed that the gas production was generally highly fluctuating in both the chambers as long as the heater was working. It appears that passing of substrate through the interconnected tube was not uniform resulting in the high variation of the gas production in both the chambers. However, during operation at the low temperature, the difference in gas generation between the two chambers was not generally significant.

The variations in temperature and total daily gas production with time in the double chamber reactor are shown in Figure 8. It is seen that the total gas production was more or less stable (750 mL/day as long as the heater was working) after 17 days of operation and the stable gas production for 40 days of Hydraulic Retention Time (HRT) was 0.38 m³/m³/d. The stable gas yield was 0.26 m³/kg of VS added. It is about 18% higher than the stable production rate (0.22 m³/kg of VS added) found in the single chamber reactor of the 1st set of experiments. As short circuiting of fresh feed could not occur in case of the double chamber reactor, it was more productive than the single chamber reactor. But when compared with the same range of operating days, the single chamber generated on average about 1160 mL gas per day (0.41 m³/kg of VS added), about 58% more gas than the double chamber reactor–the reason appears to be the higher organic content in the inoculum of the single chamber reactor.

From Figures 7 and 8, it is seen that due to sudden drop of the temperature by about 10°C, the gas generation reduced drastically (about 220 mL/d from 750 mL/d) because it came as a shock to the microbes and it continued until the end of the experiment. At this condition the total average rate of gas generation was about 0.13 m³/m³/d and the corresponding gas yield was 0.08 m³/kg of VS added which is about one-third of the values at higher controlled temperature. Hence, sudden drop of the ambient temperature of the anaerobic digesters must be prevented to obtain satisfactory production of biogas.

Anaerobic digestion of vegetable wastes in a fed-batch reactor with a HRT of 30 days and an OLR of 2.25 g/L/d was studied Velmurugan & Ramanujam [22]. They found the biogas yield as 0.59 L/g VS added.
They also reported methane yield in the range of 0.15-0.732 per g VS added for vegetable, fruit and mixture of fruit and vegetable wastes. Boualleugi et al. [11] conducted batch studies using fruit and vegetable wastes, and determined the methane generation rate of 0.16 m³/kg of VS added for OLR of 1.06 having HRT of 47 days and 0.26 m³/kg of VS added for OLR of 0.9 g VS/L/d with HRT of 32 days. Babaee and Shayan [23] obtained biogas generation rate in the range of 0.30-0.47 m³/kg of VS added for vegetable wastes with OLR in the range 1.4-2.75 g VS/L/d with HRT of 25 days in case of daily feed reactor. They also reported biogas generation rate of 0.26-0.47 m³/kg of VS added for OLR in the range of 0.30-1.6 g VS/L/d for fruit & vegetable waste and municipal solid wastes from literature review. Sridevi et al. [24] conducted daily feed two phase studies (acidogenic, HRT=2 days and methanogenic, HRT=15-25 days) with OLR varying from 1.50-4.50 g VS/L/d and found biogas production in the range of 0.24-0.72 m³/kg of VS added. Patil and Deshmukh [25] reviewed past literature on biogas yield from a mixture of vegetable wastes in the range of 0.36-0.90 m³/kg of VS added. In case of batch reactors, Jalil et al. [4] reported average biogas generation rate of 0.27 and 0.39 m³/kg of VS added for OLR of 0.83 and 1.24 g VS/L/d respectively for 40 days HRT. For the same HRT, they found the stable rate of biogas produciton as 0.24 and 0.30 m³/kg of VS added for single chamber reactor (OLR=1.18 g VS/L/d) and the double chamber reactor (OLR=0.96 g VS/L/d) respectively. The results of these studies are in good agreement with the data obtained from the present study.

Conclusion

Based on the results of the present study, the following conclusions can be made:

1. About 48% of the wastes generated in the market were easily biodegradable. Cow dung, cauliflower stick, dry paddy straw, banana leaves, papaya, and potato were the major easily biodegradable wastes.
2. The TS and SS contents of the easily biodegradable portion of the market wastes were 18.90% and 15.10% respectively.
3. Under daily feed condition at favorable temperature, the stable biogas generation rate was 0.22 m³/kg of VS added and 0.26 m³/kg of VS added for single chamber reactor and double chamber reactor respectively for OLR of 1.42 g VS/L/d and 40 days HRT.
4. Sudden and sustained change of ambient temperature changed the biogas production rate drastically.

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