Aerogenic Characters of Cellulose Digestion under Simulated Landfill Condition

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Abstract: Anaerobic digestion of cellulose is the main contributor to landfill gas generation since 91% of the methane potential of the MSW was contained in the cellulose and hemicelluloses. It is therefore important to reveal the characteristics of anaerobic digestion and landfill gas (LFG) generation of cellulose under landfill condition. In this paper, effects of temperature, initial pH, moisture content as well as inocula on cellulose digestion under simulated landfill condition were investigated. The results showed that inocula and moisture content are two important factors for LFG generation from cellulose. And the highest cumulative gas and methane yield, 620.4 ml g⁻¹ and 8.59×10⁻² g g⁻¹ were obtained respectively at the end of the experiment (the 109th day) at 30℃, with 60% moisture content when anaerobic sewage sludge were severed as inocula.

Keywords: cellulose; anaerobic digestion; landfill; methane

1 Introduction

The most common disposal method for municipal solid waste (MSW) is buried in landfills [1]. Aerobic digestion of the organic matter is limited by the small amount of air caught during landf illing. When oxygen is exhausted in the initial aerobic MSW degradation, anaerobic conditions are established. Observations made at various landfill sites have shown that the end of aerobic respiration (due to the lack of oxygen) occurs in a few weeks after MSW were buried, while gas production goes on for more than 30 years after landfill completion. Finally, most of biodegradable organic compounds are converted into CH₄ and CO₂ [2]. The yield of landfill gas reaches the maximum value when biodegradable organic compounds are totally consumed.

However, these organic matters can hardly convert totally and part of them remains in the landfill [3]. The contribution of each organic matter to landfill gas is different. The essential component of biodegradable organic compounds is carbohydrate, protein and fat. And the carbohydrate is composed of cellulose, hemicellulose and lignin. The cellulose and hemicellulose is decomposable under anaerobic conditions whereas lignin is not [4]. Around 71% of the cellulose and 77% of the hemicellulose were degraded finally in landfills. And 91% of the methane potential of the MSW was contained in the cellulose and hemicellulose [4].

Although a general scheme of organic carbon mineralization in anaerobic environments can be outlined as described above, detailed information on special matter, especially the break-down of cellulose in landfill are still lacking. Most of the test about methane production in landfill chose mixed material, but not pure substrates to be subjected. Study in the scope of cellulose digestion characterization under landfill condition is lacking. On the other hand, most of the studies of pure cellulose anaerobic digestion concentrated on the functional behavior of microbial communities in animal rumen and defined mixed cultures [5-8].

The purpose of our study was to characterize the anaerobic digestion and gas generation of cellulose and the effects of inocula, moisture content, pH and temperature on cellulose digestion under simulated landfill conditions were investigated. The results can provide important evidence for aerogenic character of cellulose digestion in sanitary landfill.

2 Materials and Methods

2.1 Effect of Inocula on Anaerobic Cellulose Digestion

The tests were carried out in 500-ml sealed saline bottles. 50g microcrystalline cellulose were mixed with different kind of inocula at a ratio of 100:5 (wt:wt). Four kinds of inocula were used in the test, including sewage sludge from anaerobic digestion reactor of wastewater treatment plant, municipal solid waste from landfill, rice paddy soil, and cow dung. Municipal solid waste was sampled from a 30-meter-deep layer in Xiaping landfill in Shenzhen, China. The moisture content of the mixture in each trail was adjusted to 60% with distilled water. After flushing the headspace with N₂ gas for 2 min, the bottles were sealed with butyl rubber with gas-guide tubes connected to 1000-ml gas gathering cylinder, and incubated at 35℃ in a water-bath. The gas yields were recorded daily, and CH₄ and CO₂ concentrations were
measured by Gas Chromatograph (SQ-206, Beijing Bei- fen Analytical Instruments, China).

2.2 Effect of Moisture Content on Anaerobic Cellulose Digestion

Aerogenic character of cellulose digestion was investigated at four moisture levels, i.e. 40%, 50%, 60%, 70% with the same experimental setup as described above. Anaerobic sludge which proved to be the most efficient inoculum in previous test was chosen and mixed thoroughly with substrate (cellulose) before incubation at 35 °C.

2.3 Effect of PH on Anaerobic Cellulose Digestion

Aerogenic character of cellulose digestion was investigated at three pH levels, i.e. 6.45, 7.00, 7.65, and no adjustment (control, pH=6.8). The moisture content was adjusted to 60% which proved optimal for anaerobic digestion of cellulose in previous test. Other conditions were the same with the above section.

2.4 Effect of Temperature on Anaerobic Cellulose Digestion

Six temperature levels, i.e. 30°C, 35°C, 40°C, 45°C, 50°C, 55°C were set to simulate temperature vary in different layers of sanitary landfills. Other conditions were the same with the above section.

2.5 Mass Balance Analysis

Mass balance of cellulose anaerobic digestion was analyzed by determining of LFG production and solid mass weight lost, which was calculated by subtracting residue from initial mass. LFG is composed of methane, carbon dioxide, water vapor and other trace gases. From a volumetric point of view, the water vapor and trace gases were neglected and considered only the dry biogas (i.e. CO2 and CH4) for determining mass removals [9]. Conversion of as-measured biogas volumes to standard conditions at 0°C and 101.3 kPa was done in a way similar to that used by Richards [10]. The biogas mass was calculated using the molecular weights of methane and carbon dioxide (16 and 44 g/mol, respectively), the molar volume of an ideal gas at STP (22.4 l/mol) and normalized individual gas contents (vol%) as follows:

\[ G_{CO_2} = V_d \times (44 \times V_{CO_2} / 100) / 22.4 \]  
\[ G_{CH_4} = V_d \times (16 \times V_{CH_4} / 100) / 22.4 \]

Where Gx, the individual biogas mass (g); Vd, the dry biogas volume at STP (l); VCH4, the biogas normalized methane content (vol%); VCO2, the biogas normalized carbon dioxide content (vol%).

2.6 Statistical Analysis

All the results reported are the means of three replicates. Anova and LSD were used to test for differences in biocconversion of cellulose in different temperatures. All tests were two-tailed and the level of significance was set at P < 0.01. All statistical calculations were performed using Statistical Package for Social Science (SPSS for Windows package release 10.0; SPSS Inc, Chicago, IL).

3 Results and Discussion

3.1 Effect of Inocula on Anaerobic Cellulose Digestion

Figure 1a illustrates the effect of inocula on LFG production during cellulose digestion. LFG productions were detected in all the trails with different inoculants. And the LFG production presented a trend of monophasic curves during experimental duration. Maximum LFG production rate of cellulose occurred before 20 to 30 days of incubation. However, there was significant difference among four treatments. It showed that the LFG yield in the trail inoculated with anaerobic sludge increased at the highest rate of 3.85ml g⁻¹ day⁻¹ from the beginning to the 22nd day. Then, the rate of LFG generation began to decrease, and the cumulative LFG yield reached 117.4 ml g⁻¹ at the end of incubation (46 days).

Moreover, the cumulative CH4 yield of cellulose was also the highest when inoculated with anaerobic sludge (Fig 1b). Aerogenic rate was maintained at a high value of 8.68×10⁻⁴ g g⁻¹ day⁻¹ from the beginning to the 22nd day. Then the slope of LFG yield curve turned smaller. The cumulative methane yield reached 2.6×10⁻² g g⁻¹ at the 46th day. CH4 from cellulose with anaerobic MSW and anoxic rice field soil were also detected. The cumulative methane yields were 2.8×10⁻³ g g⁻¹ and 3.0×10⁻⁴ g g⁻¹ respectively. No methane production was detected in the trail inoculated with cow dung. Sewage sludge from anaerobic digestion reactor of wastewater treatment plant was therefore chosen as the optimal inoculum for further study.
The presence of methanogenic bacteria, particularly for the sludge which had already been anaerobically digested, was the basis for the explanation regarding the capacity of accelerating degradation of cellulose. This positive influence has been widely investigated [11-12]. Many researchers have suggested that the addition of sludge from wastewater treatment plants is an efficient method for improvement of landfiling [13-14]. Our results are in accordance with the conclusions in literatures. In fact, most of the landfill in China receives sludge from waste water treatment plant, but due to lack of proper treating technologies and facilities, the drawbacks linked to operational difficulties may hinder the movement of vehicles above the waste. It is therefore a practical theme to mix certain amount of sludge during landfiling. However, pretreatment has to be performed to reduce the moisture content of sludge. For instance, a possible solution could be that of mixing the sludge with ashes from power-plants with low degree of humidity, whose environment compatibility has been confirmed in recent studies [15].

3.2 Effect of Moisture Content on Anaerobic Cellulose Digestion

Figure 2a illustrates the effect of moisture content on the production of LFG from cellulose digestion. It showed that the LFG generation rate of cellulose digestion was higher at moisture content of 60% than other levels. The LFG yield in the trail with 60% moisture content maintained much higher than other treatments, as evidenced by its production rate of 380.6 ml g⁻¹, compared with 93.6 ml g⁻¹ (40%), 80.2 ml g⁻¹ (70%) and 72.0 ml g⁻¹ (50%) in other trails.

The trends of methane yield were similar with that of LFG yield as Figure 2b showed. The cumulative methane yield of cellulose anaerobic digestion at different moisture content could be ranked as: moisture content 60% > moisture content 70% > moisture content 40% > moisture content 50%. Methane yield at moisture content of 60% reached the highest value of $4.7 \times 10^{-2}$ g g⁻¹.

There were studies showed that the methane production rate increases with increasing moisture content of the waste from 25% to 60% [16-17]. Recommended moisture content in literatures ranged from a minimum of 25% to optimum levels of 40% to 60% [14]. Besides diluting the inhibitors, the main effect of the increased water content is probably the facilitated exchange of substrate, nutrients, buffer, and spreading of microorganisms in the micro environments of waste. However, too much water in landfill can reduce the degradation rate of organic matters, especially for big molecule organics such as cellulose and lignocelluloses due to slow metabolic activities of anaerobes under strict anaerobic condition. Therefore, moisture content should be controlled and distributed as homogeneous as possible during the landfiling, and seepage should be avoided.

On the other hand, too low moisture content inhibits the microbial activity and will hinder the fermentation of organics. Leachate recirculation appears to be an effective method to increase moisture content in a controlled landfill unit [18-19]. The successful operation of several bioreactor landfills has proved that moisture content regulation through leachate recirculation showed signifi-
cant advantages in accelerating waste degradation, enhancing gas production rates, and settlement promoting. The rapid treatment of the waste also facilitates the operation of a bioreactor landfill as a reusable landfill.

### 3.3 Effect of PH on Anaerobic Cellulose Digestion

The effect of initial pH on LFG generation during cellulose digestion is shown in Figure 3a. The LFG yield with no pH adjustment (pH = 6.8) increased at the highest rate (7.08 ml g⁻¹ day⁻¹). However, bio-conversion of cellulose with pH adjustment seemed to be inhibited as evidenced by much less cumulative LFG yields compared with that not controlled. Totally 474 ml g⁻¹ LFG was produced in trail without pH adjustment on the 66th day, but only 282.5 ml g⁻¹, 231.7 ml g⁻¹, and 2 ml g⁻¹ were obtained in the trails with initial pH of 7.00, 7.65, and 6.45, respectively.

Similar trend was observed with methane generation as shown in Figure 3b. The inhibition of pH adjustment to methane generation was obvious: only the methane yield in trail with no pH adjustment increased at a high rate, and the cumulative methane yield at the end of the test reached 6.2×10⁻² g g⁻¹. But that in other trails were only 2.8×10⁻¹ g g⁻¹ (pH = 7.00), 2.6×10⁻³ g g⁻¹ (pH = 7.65) and 8.8×10⁻⁶ g g⁻¹ (pH = 6.45) respectively. Initial pH adjustment was therefore not recommended in further study.

The methanogenic bacteria are activated only within a narrow pH range of 6 to 8 [20]. The pH adjustment, however, is unnecessary even harmful, because microbial action can change the pH of substrate. The neutralization of acetic acid with sodium hydroxide may decrease yield of CO₂, which will lead to excessively high pH of anaerobic digestion and inhibit further methanation process [21]. It can explain that the LFG (methane) yield of cellulose digestion without pH adjustment was higher than those adjusted. Therefore, it is not necessary to control pH during landfilling operation. On the other hand, the presence of buffering material in the landfill (e.g. demolition waste, soil) will significantly improve the ability of the landfill environment to maintain a reasonable pH range during anaerobic digestion.

### 3.4 Effect of Temperature on Anaerobic Cellulose Digestion

In the experimental setups, the LFG production from cellulose lasted for 109 days at different temperatures (Figure 4a). The course of bioconversion of cellulose can be divided into three phases: 1) Adaptation phase (about 0–10 days). The rate of LFG generation was low at this stage due to limited population and low activity of the microorganisms. 2) Swift growth phase (about 11–55 days). The cumulative LFG yield increased rapidly. Also, the highest value of LFG generation rate was detected in this stage. 3) Decay phase (56 days later). The cumulative LFG yield increased very slowly due to the negative growth of microbial population. The rate of LFG generation thus began to fall down. Moreover, the rates of LFG production differed significantly at different temperatures. The cumulative LFG yields of cellulose digestion at low temperatures (30°C and 35°C) were higher than the higher temperatures (40°C ~ 55°C), i.e. 30°C ~ 35°C is the optimum temperature for mesophilic bacteria.

Figure 4b shows the methane production at different temperatures over the experimental duration. The highest cumulative methane yield was obtained at 30°C, which was 8.59×10⁻² g g⁻¹. The trends of methane production for cellulose digestion in these trails were also characterized by three phases as described above. For instance, CH₄ production rate was low for the first 10 days when incubated at 30°C, and it increased steadily till the 60th day and decreased thereafter. While the methane yields in other trails were 7.13 × 10⁻² g g⁻¹ (40°C), 5.15 × 10⁻² g g⁻¹ (45°C), 3.33 × 10⁻² g g⁻¹ (50°C), 2.62 × 10⁻² g g⁻¹ (35°C) and 1.89 × 10⁻² g g⁻¹ (55°C) respectively. That means the anaerobic digestion was
inhibited significantly due to the slow microbial growth at unfavorable temperature. Figure 4c shows the CO$_2$ accumulation during the process of cellulose digestion. The highest cumulative CO$_2$ yield was also obtained at 30°C, which was 5.30 × 10$^{-1}$ g g$^{-1}$.

The results of mass balance showed that the initial material mass was equal to the sum of LFG mass and residual mass (Figure 5). And solid mass reduction at 30°C was the maximum, which accounted for 67.8% of initial material mass. Statistical analysis showed that average of remained material, CH$_4$ and CO$_2$ production in six temperature levels were significantly different (P < 0.01) (Table 1). This is in accordance with the above results that the effect of temperature on anaerobic bioconversion of cellulose was significant.

The bioconversions of cellulose with mesophilic (anaerobic sludge) and thermophilic (refuse sample from 30 meters deep in MSW landfill) inocula were compared to correct the bias possibly caused by predominant microbial germplasm in different inocula. The results corroborated the above test as evidenced by the higher LFG yields in trails with mesophilic inocula than that of

![Figure 4a. Effect of temperature on cumulative gas yield of anaerobic cellulose digestion](image)

![Figure 4b. Effect of temperature on CH4 yield of anaerobic cellulose digestion](image)

![Figure 4c. Effect of temperature on CO2 yield of anaerobic cellulose digestion](image)

![Figure 4d. The comparisons of cumulative gas yield with mesophilic and thermophil inocula](image)

![Figure 5. Mass balance of anaerobic cellulose digestion](image)

![Table 1. Result of mass balance of anaerobic cellulose digestion with different temperature](table)

<table>
<thead>
<tr>
<th>Temperature</th>
<th>Remained material</th>
<th>CH$_4$</th>
<th>CO$_2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>30°C</td>
<td>16.10 ± 0.11</td>
<td>4.29 ± 0.04</td>
<td>26.51 ± 0.07</td>
</tr>
<tr>
<td>35°C</td>
<td>29.80 ± 0.10</td>
<td>1.31 ± 0.01</td>
<td>17.12 ± 0.11</td>
</tr>
<tr>
<td>40°C</td>
<td>34.50 ± 0.14</td>
<td>3.56 ± 0.07</td>
<td>9.97 ± 0.10</td>
</tr>
<tr>
<td>45°C</td>
<td>32.30 ± 0.17</td>
<td>2.57 ± 0.01</td>
<td>12.66 ± 0.08</td>
</tr>
<tr>
<td>50°C</td>
<td>37.60 ± 0.21</td>
<td>1.65 ± 0.03</td>
<td>9.04 ± 0.06</td>
</tr>
<tr>
<td>55°C</td>
<td>43.20 ± 0.20</td>
<td>0.94 ± 0.07</td>
<td>5.28 ± 0.13</td>
</tr>
</tbody>
</table>

(Mean ± SD) Data within a column with the different letter are significantly different (ANOVA, LSD, P<0.01)
with thermophilic inocula (Figure 4d). The LFG yields at the 88th day ranked as: anaerobic sludge (30°C) > anaerobic sludge (50°C) > anaerobic MSW (50°C) > anaerobic MSW (30°C), which demonstrated that cellulose digestion was improved by inoculating anaerobic sludge either at high temperature or at mesophilic temperature. These results also corroborated the effect of inocula on cellulose digestion in previous tests.

The anaerobic degradation rate of waste was significantly affected by temperature and microbial population. The methanogenic bacteria contain a mesophilic group with a rate maximum around 40°C and a thermophilic group with a maximum around 70°C. Only the former group is relevant in landfills. The methane production rate has been proven to increase significantly (up to 100 times), when the temperature is raised from 20 to 30 and 40°C [17][22]. And Ranade (1988) reported that the optimum temperature of anaerobic digestion ranges from 30 to 40°C. The methane production rate with significant inhibition observed at temperatures above 50 to 55°C [23]. The results suggested that rapid degradation of cellulose occurs at the layer with lower temperature in landfill site, such as bottom and up layer.

Temperature control at full-scale landfills may be difficult to achieve. Introduction of air and the consequent onset of aerobic activity has been suggested to stimulate methane production. Another potential method of temperature control under investigation is recirculated leachate such as used in Sweden’s experiment “Energy Loaf”

However, at this point, temperature control in the landfill bioreactor is not practical and its benefit has not thoroughly demonstrated.

4 Conclusions and Perspectives

Effect of initial pH, moisture content, temperature and inocula on cellulose digestion under simulated landfill condition have been investigated. It showed that inocula, temperature as well as moisture content are key parameters affecting the degradation rate of cellulose.

Anaerobic sludge was deemed to the optimal inoculants through the different inocula tests. The yields of both cumulative LFG and methane at the 46th day reached the maximum when cellulose was inoculated with anaerobic sludge, with the values of 117.4 ml g⁻¹ and 0.026 g g⁻¹ respectively.

Moisture content is a crucial parameter for cellulose fermentation, and 60% showed to be the optimum moisture content among the four tested moisture content levels. Maximum yields of both the cumulative LFG and methane, which were 380.6 ml g⁻¹ and 0.047 g g⁻¹ respectively, were obtained at the end of test with the moisture content of 60%.

The cellulose anaerobic digestion tests at different temperatures indicated that mesophilic temperature (30°C) favored high yield of landfill gas generation.

The result also indicated that no pH adjustment is needed for cellulose degradation under anaerobic condition, which is also practical for landfill operation.

These results provide practical guides for operation of bioreactor landfills to get stabilization at relative short time.

References


