



# Influence of acidogenic headspace pressure on methane production under schematic of diversion of acidogenic off-gas to methanogenic reactor

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## ABSTRACT

This study investigated the effects of 12.6 psi (T1), 6.3 psi (T2), 3.3 psi (T3) and ambient (T4) headspace pressure on the metabolic pathways in the acidogenic leach bed reactor (LBR) and overall methane recovery during two-phase anaerobic digestion of food waste. Diversion of biogas from LBR enhanced COD and soluble product generation in T2, T3 and T4 whereas, high pressure (T1) resulted in comparatively higher lactate production and low protein degradation. A pressure of 3–6 psi (T2 and T3) improved the production of COD by ~22–36%, soluble products by ~9–43%, volatile solid reduction by ~14–19%, and CH<sub>4</sub> production by ~10–31% compared with control. Besides, ~3–6 psi headspace pressure positively influenced the composition of soluble products resulting in enhanced methane recovery adding advantage to the two-phase system. A headspace pressure of ~3–6 psi is recommended to enhance the hydrolysis-acidogenesis; however, the actual hydrogen concentration should be considered.

## 1. Introduction

Hydrogen (H<sub>2</sub>) and carbon dioxide (CO<sub>2</sub>) are the important co-products during the primary fermentation of soluble organic substrates and these two components contribute to the headspace pressure of the acidogenic reactor. The reactions of converting hydrolysis products to short-chain intermediates can only be processed under low pressure of H<sub>2</sub> due to their unfavourable thermodynamic conditions under standard conditions (Huang et al., 2016). In addition, it was confirmed that high H<sub>2</sub> partial pressure (pH<sub>2</sub>) would inhibit the process of hydrolysis as well as further H<sub>2</sub> generation (Kongjan et al., 2009; Wang et al., 2008). However, it is the fact that H<sub>2</sub> and CO<sub>2</sub> produced in acidogenic reactor account for as high as 30% of the consumed substrates (Schievano et al., 2012); which means under the natural process, H<sub>2</sub> & CO<sub>2</sub> would accumulate in the acidogenic headspace and inhibit the process of hydrolysis/acidogenesis.

Headspace flushing is a common way to mitigate headspace inhibition. It was demonstrated that flushing the headspace with a gas mixture of 20% CO<sub>2</sub> significantly increased the methane production by over 20% (Koch et al., 2015). However, the drawbacks of headspace flushing are lowering substrate conversion efficiency and threatening the sustainability of the environment. Hence, a more favourable

strategy is needed to achieve a sustainable way for waste disposal. In a previous study, it was demonstrated that diverting the acidogenic off-gas from acidogenic leach bed reactor (LBR) into the methanogenic upflow anaerobic sludge blanket (UASB) reactor influenced the composition of the acidogenic leachate in LBR and increased the methane recovery in UASB (Yan et al., 2016). Reutilization of acidogenic off-gas from LBR definitely alleviated the inhibition of H<sub>2</sub>. However, the degree of acidogenic off-gas diversion would certainly be a critical factor that affects the acidogenic performance and overall methane production from two-phase anaerobic digestion (AD). Hence, it is necessary to study the effect of varying headspace pressures under the schematic of diversion of acidogenic off-gas to methanogenic reactor.

As H<sub>2</sub> and CO<sub>2</sub> are also the by-products of acidogenesis, consequently the headspace pressure could influence the acidogenic reactions and the microenvironment. H<sub>2</sub> is an important factor that influences the metabolic pathway occurs in acidogenic reactor. The composition of acidogenic products in the leachate is affected by the balance between H<sub>2</sub> and reducing equivalent. Higher H<sub>2</sub> pressure in the headspace could inhibit the acetogenic biomass growth rate, since high concentration of H<sub>2</sub> inhibits the generation of propionic and butyric acids (Lyberatos and Skiadas, 1999). Further, higher headspace pressures could lead to saturation of CO<sub>2</sub> in the reactors medium which

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could affect the pH and alter the microbial activity, thus shifting the anaerobic metabolic pathway (Valero et al., 2016). Therefore, maintaining appropriate headspace pressure in the acidogenic reactor is the key to control the metabolic pathways towards the target product.

Besides the effect of headspace pressure, the composition of the headspace gases has also been reported to regulate product spectrum. Researchers have considered altering the composition of headspace gases and decreasing the pressure of H<sub>2</sub> by sparging gases into the liquid phase of the reactor (Cazier et al., 2015). Different compositions of headspace with anaerobic microorganisms resulted in considerable differences in the generation and distribution of volatile fatty acids (VFAs) (Hillman et al., 1991). Similar observations were also reported by Valero et al. (2016) and Karlsson et al. (2008). However, studies with N<sub>2</sub>-sparging performed by Mizuno et al. (2000) and Kim et al. (2006) did not result in significant change in the composition of liquid end products, even when H<sub>2</sub> yield was increased. Headspace gas composition not only affects H<sub>2</sub> production, but also influences the overall energy efficiency of AD system. Recently, Patra and Yu (2013) observed that the headspace gas composition and bicarbonate concentrations in the media could affect methane production and other characteristics of rumen fermentation in *in vitro* gas production systems.

Therefore, the objective of this study was to investigate the effect of varied acidogenic headspace pressures and the composition of headspace on the performance and energy recovery of two-phase AD under schematic of diversion of acidogenic off-gas to methanogenic reactor. It is expected that controlling the headspace of acidogenic reactor through accumulation of gases produced in the LBR could alter the pressure as well as the composition of gases. This can be achieved through simple control of the pressure with a cut-off pressure value beyond which the gases can be released from the reactor and transferred to methanogenic reactor for further utilization. The continuous transfer of the gases is expected to cause the shift of metabolic pathway in acidogenic reactor as well as the performance and methane recovery from two-phase AD.

## 2. Materials and methods

### 2.1. Substrate and inoculum

Artificial food waste with a total solid (TS, %) of 40.0 ± 2.5, volatile solid (VS/TS, %) of 98.0 ± 0.1, total organic carbon (TOC, %) of 45.9 ± 4.4 and total Kjeldahl nitrogen (TKN, g/kg) of 28.8 ± 0.5 was used as the substrate in this experiment. The artificial food waste used in the study was prepared as detailed in Yan et al. (2014). Anaerobically digested sludge with 2.3% TS and 76% VS/TS obtained from Shek Wu Hui wastewater treatment plant was used as the seed source.

### 2.2. Experimental set up and treatments

LBRs with a 5.4-L working volume and 2.9-L leach bed volume was used as acidogenic reactor while UASB with 10-L reactor volume was used as the second phase methanogenic reactor. Both LBRs and UASBs were prepared as detailed in a previous study (Yan et al., 2016).

LBRs were filled with 2.0 kg of food waste mixed with 20% (I/S, wet basis) of inoculum and 100 g of wood chips as bulking agent according to a previous study (Xu et al., 2012). The liquid to solid ratio of 1:1 was applied for the start-up of the experiment, which means 2.0 L of water was added to each LBR. The sampling frequency was 1 day. During each sampling, the leachate was taken out and exactly 50% (v/v) of the leachate with pH adjusted to 6.0 with NaOH (0.5 mol/L) was returned back to the LBR from the top; while the remaining 50% of the leachate was fed to UASB with 50 mL reserved for chemical analysis.

In order to investigate the effects of different headspace pressure levels on acidogenic performance of LBR and overall CH<sub>4</sub> recovery under the regime of reutilization of acidogenic off-gas in methanogenic reactor (Yan et al., 2016), the following four headspace pressure levels

were set according to gas production levels obtained from a preliminary experiment: 12.6 psi (T1), 6.3 psi (T2), 3.3 psi (T3) and ambient pressure (T4). The headspace pressure was controlled by online pressure sensor; and the acidogenic gases beyond the set pressure point was diverted to methanogenic UASBs.

### 2.3. Solubilization rate

The degree of solubilization (%) of organic particulates from the substrates in the LBR was calculated by the production of total soluble products (TSP) and recovery of chemical oxygen demand (COD) (TSP/COD).

$$\text{Solubilization rate (\%)} = \frac{\text{TSP}}{\text{COD}} \times 100 \quad (1)$$

### 2.4. Analyses

Methods for preparation and physicochemical analysis of the food waste and seed sludge were followed as detailed in a previous study (Yan et al., 2014). Acidogenic leachate samples collected from LBRs were analyzed for pH, COD, soluble products, NH<sub>4</sub><sup>+</sup>, and TKN, according to the methods described in Yan et al. (2016). Total soluble products are the sum of volatile fatty acids (acetate, propionate, *iso*- and *n*-butyrate, *iso*- and *n*-valerate and caproate), alcohols (ethanol, propanol and butanol), solvents (acetone) and lactate. Volume of acidogenic biogas in LBR was calculated by integration of real-time data of gas flow rate measured by a mass flow controller (MFC, Seven Star, China). The outlet acidogenic off-gas was collected using Tedler gas bags and 1-mL gas was used for the analysis of composition during each sampling point. Biogas from the UASB was continuously measured using a gas meter and analyzed for the methane contents using a gas chromatograph (HP7890) equipped with TCD and PLOT-Q column (30 m × 0.53 mm × 15 μm). Concentration of gases was calculated by referring to the corresponding standard curves, methods for which were described in Yan et al. (2016).

### 2.5. Statistical analysis

Analysis of variance (ANOVA) and general linear model (GLM) procedures of SPSS Statistics v19 were used to evaluate the effect of regulation of acidogenic headspace pressure on the performance of the two-phase AD digesters. It was considered significantly different at P < 0.05 level.

## 3. Results and discussion

### 3.1. Pressure profile

Fig. 1 shows the dynamics of headspace pressures in the experiment. Although the set values in T1 was 12.6 psi, the real pressure never reached this level while values ranging between 8 and 12 psi were observed during the first half of the digestion. Thereafter the headspace pressure gradually decreased till the end of this experiment, matching with a typical batch study. Most of the points in T2 reached the set value of 6.3 psi, while all the points in T3 could maintain the set value of 3.3 psi. Since ambient pressure was applied in T4, i.e., the gases produced are continuously transferred to the UASB, increase of pressure was not observed. The four levels of headspace pressures were set to investigate their effects on acidogenic performance and overall energy recovery from two-phase AD under the schematic of diversion of acidogenic off-gas.

### 3.2. Performance of LBR

Volumes of leachate production from all four treatments throughout

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