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# Influence of lactic acid on the two-phase anaerobic digestion of kitchen wastes

ZHANG Bo<sup>1,2</sup>, CAI Wei-min<sup>2</sup>, HE Pin-jing<sup>1,\*</sup>

 State Key Laboratory of Pollution Control and Resources Reuse, Tongji University, Shanghai 200092, China. E-mail: solidwaste@mail.tongji.edu.cn
School of Environmental Science and Environmental Scienc

2. School of Environmental Science and Engineering, Shanghai Jiaotong University, Shanghai 200240, China

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#### Abstract

To evaluate the influence of lactic acid on the methanogenesis, anaerobic digestion of kitchen wastes was firstly conducted in a two-phase anaerobic digestion process, and performance of two digesters fed with lactic acid and glucose was subsequently compared. The results showed that the lactic acid was the main fermentation products of hydrolysis-acidification stage in the two-phase anaerobic digestion process for kitchen wastes. The lactic acid concentration constituted approximately 50% of the chemical oxygen demand (COD) concentration in the hydrolysis-acidification liquid. The maximum organic loading rate was lower in the digester fed with lactic acid than that fed with glucose. Volatile fatty acids (VFAs) and COD removal were deteriorated in the methanogenic reactor fed with lactic acid compared to that fed with glucose. The specific methanogenic activity (SMA) declined to 0.343 g COD/(gVSS-d) when the COD loading were designated as 18.8 g/(L-d) in the digester fed with lactic acid. The propionic acid accumulation occurred due to the high concentration of lactic acid fed. It could be concluded that avoiding the presence of the lactic acid is necessary in the hydrolysis-acidification process for the improvement of the two-phase anaerobic digestion process of kitchen wastes.

Key words: lactic acid; kitchen wastes; anaerobic digestion; methanogenesis

# Introduction

Kitchen wastes contain high concentration of biodegradable organic compounds and therefore are predominant renewable resource in municipal solid wastes (Fang, 1999; Wang and Nie, 2001). Owning to efficient resource recovery and lessened environmental impact, anaerobic digestion compares favorably with alternative treatments, such as incineration, landfill and composting (Foresti, 2001; McCarty, 2001; Gilzen, 2002; Lema and Omil, 2001; van Lier *et al.*, 2001; De Baere, 2000; Suh and Rousseaux, 2002).

Anaerobic digestion of organic wastes is a complex process involving the hydrolysis, fermentative acidogenesis, acetogenesis and methanogenesis. Due to the high organic wastes content, the single-phase anaerobic digestion of kitchen wastes easily leads to a subsequent accumulation of intermediary products with a resultant fall in pH, thus giving rise to unbalanced fermentation and diminishing the stability of the process (Ince, 1998; Jeyaseelan and Matsuo, 1995). The two-phase anaerobic digestion could optimize the conditions for the hydrolytic acidogenic group of bacteria as well as for the acetogenic-methanogenic group and enhance the stabilization of organics and gasification rates (Yilmazer and Yenign, 2002; Banks and Wang, 1999; Bhattacharya *et al.*, 1996; Qi *et al.*, 2003). Most studies involved a solid waste

\*Correspondence author. E-mail: solidwaste@mail.tongji.edu.cn.

reactor and a high-rate anaerobic wastewater reactor for kitchen wastes (Cho and Park, 1995; Wang et al., 2002a, 2003a; Xu et al., 2002; Han et al., 2002). In these studies, high-rate methanogenic reactors such as UASB (upflow anaerobic sludge bed), UBF (upflow blanket filter) were used to recover methane from the acidified leachate. The methanogenic phase was focused on, as it is the energyyielding phase, while not much attention has been paid to the acidogenic phase. However, the methanogenesis is based on the hydrolysis and acidification step and the overall metabolic rate and operational stability of the methanogenic phase depend intensely on the fermentation products from the hydrolysis-acidification reactor. Therefore, it is essential to investigate the distribution of hydrolysis-acidification products and the influence of predominant products on the subsequent methanogenesis.

The lactic acid was found to be the main fermentation products for kitchen wastes (Wang *et al.*, 2001, 2002b). Due to the wide application of lactic acid in many regions including food industry, pharmaceutical, leather and textile industries, a new reducing and recycling system that produce lactic acid from kitchen wastes was developed. Despite of that, the recovery of biogas from kitchen wastes is still a predominant recycling route. Nevertheless, little is known about the influence of lactic acid on the two-phase anaerobic digestion process from the point of view of the methanogenesis.

The objective of this research was to evaluate the influence of lactic acid on the subsequent methanogenesis in the two-phase anaerobic digestion of kitchen wastes. The performance of two digesters feeding with lactic acid and glucose was compared, focusing on the effluent propionic acid concentration and the properties of methanogenic microorganism in the process of increasing COD loading.

### **1** Materials and methods

### 1.1 Two-phase anaerobic digestion of kitchen wastes

Kitchen wastes were taken from a student restaurant located in Shanghai Jiao Tong University. The obtained kitchen wastes mainly contained cooked rice, vegetables, meat, eggs, potatoes and salts. Table 1 shows the physical and chemical characteristics of the sampled kitchen wastes.

Table 1 Sampling components of kitchen wastes

Parameter	Value	Parameter	Value
Particle size (mm)	<2	C/N	49.9
TS (%)	12.9	$NH_4^+$ -N (mg/L)	<10
VS (%)	12.5	TP (mg/L)	182.5
SS (%)	9.6	S-TP (mg/L)	172.5
VSS (%)	9.4	Orthophosphate (mg/L)	104.8
pH	3.9	C/P	910.5
VFA (g/L)	3.6	Protein (%TS)	16.1
TOC (%TS)	46.8	S-protein (g/L)	2.6
S-TOC (g/L)	11.8	Carbohydrate (%TS)	69.3
COD(g/L)	166.2	S-carbohydrate (g/L)	21.6
S-COD (g/L)	53.2	Lipids (%TS)	10.6
Total nitrogen (g/L)	3.3	S-lipids (g/L)	2.2

The index with the first term S represented the characteristics of centrifuged liquid.

Anaerobic digestion of kitchen wastes was conducted in a two-phase anaerobic digestion process. The experimental set-up is shown as Fig.1. The hydrolysis-acidification reactor and the methanogenic reactor were fed intermittently and were operated with a cycle of 12 h including 30 min of feeding and discharging, 30 min of sedimentation and 11 h of reaction. The kitchen wastes were pumped into the hydrolysis-acidification reactor when the valve 1 and valve 2 were on. The feeding of kitchen wastes

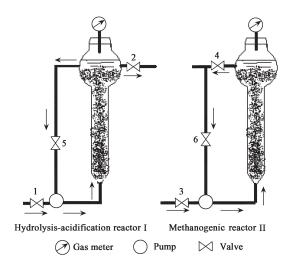


Fig. 1 Experimental set-up of two-phase anaerobic digestion process.

and the discharging of fermentation liquid were carried out simultaneously. The methanogenesis reactor had the same operation as the hydrolysis-acidification reactor. The hydrolysis-acidification products were centrifuged firstly. The acidified liquid was pumped into the subsequent methanogenic reactor, while the residual solid was recycled into the hydrolysis-acidification reactor. Both the hydrolysis-acidification and the methanogenic reactors were mixed with the inner-phase leachate recirculation. The valve 5 and valve 6 were shut down when the feeding was conducted. The kitchen wastes were diluted with tap water and about 100 g/L of TS was obtained. The feeding volume was increased from 10 ml to 20, 40, 60, 80 and 100 ml. The methanogenic effluent was recycled into the hydrolysis-acidification feeding to control hydrolytic retention time (HRT) at 5 d. HRT of the methanogenic reactor was controlled at 5 d.

# 1.2 Comparisons of two digesters feeding with lactic acid and glucose

Performance of two digesters feeding with lactic acid and glucose was compared to determine the influence of lactic acid on the methanogenic process. Constituents of the feeding synthetic medium are shown in Table 2. The constituents of the synthetic medium in Reactor I (abbreviated as RI) were according to the distribution of the hydrolysis-acidification products obtained in this study. The feeding concentration of two anaerobic reactors was both designated as 25 g/L and the feeding constituents are shown in Table 1. In RI, the lactic acid concentration was half of the total feeding COD concentration. In Reactor II (abbreviated as RII), the glucose concentration was main feeding substrate.

Table 2 Constituents of the feeding synthetic medium

Feeding constituent	Reactor I	Reactor II
Glucose (COD%)	25	75
Sodium acetate + Sodium propionate + Butyric acid	25	25
( <i>m</i> Sodium acetate: <i>m</i> Sodium propionate: <i>m</i> Butyric acid =1:1:1 COD%)		
Lactic acid (COD%)	50	0

The two digesters had the same dimensions with working volume of 1.6 L and operated under the same conditions. The operation of two reactors was the same as the above-mentioned methanogenic reactor. Granular sludge originating from the mesophilic anaerobic reactor treating beer wastewater was used as inoculum of the anaerobic sequence batch reactor (ASBR). The loading rate was increased with the feeding COD concentration. The two sets of systems were maintained at 35–37°C and operated for 120 d.

### 1.3 Analytical methods

pH, total solid (TS), volatile solid (VS), suspended solid (SS), volatile suspended solid (VSS), COD, orthophosphate, NH<sub>4</sub><sup>+</sup>-N and total phosphate (TP) were analyzed according to the standard method (APHA, 1995). Total

organic carbon (TOC) was measured by catalytic oxidation on a multi N/C 3000 analyzer (Analytik Jena, German). The protein content was determined based on Kjeldahl nitrogen, which was measured by acid hydrolysis of the insoluble organic nitrogen and the Kjeldahl nitrogen titration procedure (APHA, 1995). The process of the digestion and distillation was finished with the nitrogen analyzer (BÜCHI, Digestion Unit K-424; BÜCHI, distillation Unit B-324). The measured Kjeldahl nitrogen was multiplied by 6.25 to give the protein content (APHA, 1995). Lipids concentration was determined gravimetrically after extraction of lipids by petroleum ether according to the Soxlett extraction method (Nielsen, 2002). Carbohydrates in the wastes were determined according to phenol-sulphuric acid method (Nielsen, 2002). The VFA was determined by a high performance liquid chromatography (2010A, Shimadzu, Japan) equipped with a C18 column. A gas chromatograph (GC-14B) was used to measure the biogas composition. The gas chromatogram (GC) was equipped with a 3 mm×2 m TDX-102 filled column and a thermal conductivity detector (TCD). For the dissolved COD, TOC, TP, protein, carbohydrate, lipids, the sample was centrifuged at 10000 r/min for 30 min, and then the same method as the above was applied.

The measurement of specific methanogenic activity (SMA) was performed in duplicates in serum vials (120 ml) based on the reported method (Hwang and Cheng, 1991). Sludge samples were taken for the SMA analysis when the performance of the reactor attained stable. The methanogenic acitivity of anaerobic granule sludge (1 g VSS) was measured for the sodium acetate (5 g COD/L).

The microbial examination was carried out with scanning electron microscopy (SEM). The sample preparation procedures were as reported by Fang and Chui (1995).

### 2 Results and discussions

A similar result as the study of Wang *et al.* (2001, 2002b) was obtained that lactic acid was the main fermentation products of kitchen wastes. As shown from the distribution of fermentation products at different COD loading rate in Fig.2, lactic acid concentration constituted approximately 50% of the COD concentration in

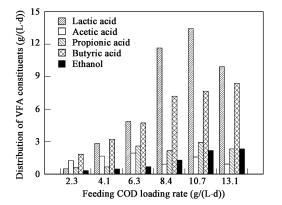


Fig. 2 Variations of the fermentation products at different feeding COD loading rates.

the hydrolysis-acidification liquid. There were a variety of factors affecting the generation of lactic acid such as substrate characteristics, seed inoculum, pH and temperature (Wang et al., 2001, 2002b, 2003b). Kitchen wastes contain rich nutrition, including carbohydrate, lipid and proteins. Carbohydrates are broken down to sugars of low molecule weight such as maltose, glucose, fructose and galactose with the saccharification process. Soluble sugars are substrates available for the growth of lactic acid bacteria. In this study, there are two factors favorable to the production of lactic acid. Firstly, the carbohydrates are the main constituents of kitchen wastes. Secondly, pH in the hydrolysis-acidification reactor kept in approximately 5 following the COD loading increase to 8.4 g/(L·d), while the optimum pH of lactic acid production was thought to between 5 and 6 (Fu and Mathews, 1999).

It can be obviously seen from Fig.3 that the lactic acid had an intensely negative influence on the performance of methanogenesis. Compared to RII, RI had a higher effluent COD concentration, a lower biogas productivity rate, and a higher effluent VFA concentration with an increase in the COD loading rate. At the low COD loading rate, a lag period in the COD removal and biogas production was found in the RII. The pH and alkalinity in RI indicated a slightly higher value than that in RII. RII could obtain a higher COD loading rate than RI. Therefore, it can be concluded that high concentration of lactic acid feeding limited the increase in the COD loading rate.

The influence of lactic acid on the effluent propionic acid concentration in Fig.4 partly explained the different operational performance for RI and RII. The effluent propionic acid concentration increased almost linearly with the feeding lactic acid loading rate in RI. About 19% of the lactic acid was converted into the propionic acid per d. Some researchers have reported that the lactic acid was easily degraded into propionic acid under conditions that the hydrogen partial pressure was over 100 Pa (Ren et al., 1997). Furthermore, lactic acid was usually considered to be the precursor of propionic acid during the anaerobic digestion from the point of view of microorganism (Min, 1993; Costello et al., 1991). Skiadas et al. (2000) also considered that lactate was easily converted into propionic acid and acetic acid. It was widely studied on the influence of the accumulation of propionic acid on the methanogenesis process (Ren et al., 2002). Since the methanogenesis of propionate was slower compared with acetate and butyrate, propionate was an undesirable intermediate product in the methanogenic process (Fang and Yu, 2002). The presence of a large amount of propionic acid in RI feeding with lactic acid indicated that the production of lactic acid should be avoided considering the subsequent methanogenesis.

Fig.5 shows the variations of the SMA for the anaerobic granule sludge in two reactors. The SMA is an indicator for evaluating the methanogenic acitivity of the biomass under a condition in which the supply of substrate is not a limiting factor. There is no obvious difference in SMA with the COD loading below 2.5 g /(L·d) in two reactors. However, SMA in RI showed an obvious decrease when the COD loading continued to increase. The VFA-

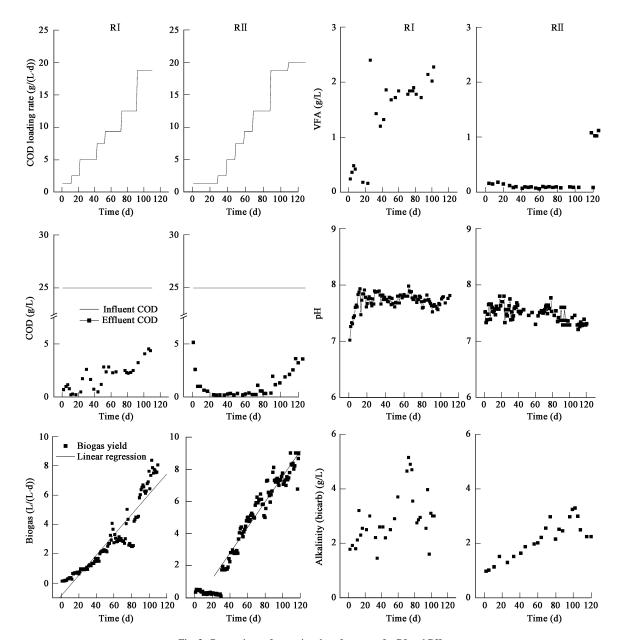
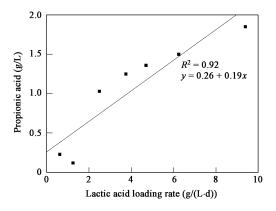


Fig. 3 Comparison of operational performance for RI and RII.



1.25 1.00 0.75 0.75 0.25 0.25 ---- Reactor II ---- Reactor II ---- Reactor I 0 ---- Reactor I ---- Reactor I

Fig. 4 Influence of lactic acid on the effluent propionic acid concentration.

degrading activity of granule was the highest for butyrate, and the lowest for propionate (Shin *et al.*, 2001). Since the high concentration of lactic acid led to the accumulation

Fig. 5 Comparison of SMA for RI and RII.

of propionic acid in the methanogenic effluent, the SMA in RI declined to 0.343 g COD /(  $gVSS \cdot d$ ) when the COD loading was designated as 18.8 g /(L·d).

248

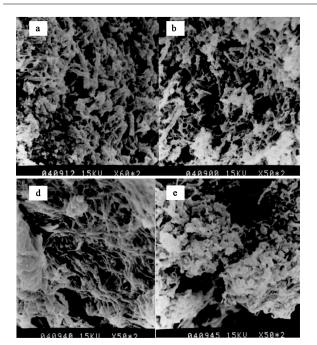


Fig. 6 SEM pictures of the anaerobic granule sludge for RI and RII; (a) seeding sludge granule; (b) sludge granule in RII; (d, e) sludge granule in RI.

The seeding sludge granule and most of the sludge granule in RII were black, while the sludge granule in RI was yellow. Fig.6 shows the anaerobic microorganism in the sludge granules from the seed and two reactors. The methanococcus and the methaobacillus predominate over the sludge granule in the inoculum and RII. However, there appeared the methanofilamentous colony and large cavities in the sludge granule in RI. The methanofilamentous colony could lead to the loose structure of the sludge granule, and further cause the decrease in SMA (Liu, 2001).

The discovery of propionic acid accumulation in the effluent in the methanogenic reactor feeding with the lactic acid provided a possible explanation for the general failure of anaerobic digestion system. Generally, the propionic acid accumulation was considered to be the bottle-neck of anaerobic digestion process. It was found in this study that the lactic acid easily led to the production of the effluent propionic acid. Therefore, to some extent, the research in this paper gives an answer why the propionic acid accumulation occurs in some anaerobic systems. The production of lactic acid in the hydrolysis-acidification stage could be an important reason. Thereby it is necessary to avoid the presence of the lactic acid in the hydrolysisacidification process for the improvement of the two-phase anaerobic digestion process.

# **3** Conclusions

It could be concluded that the lactic acid predominated over fermentation products of the hydrolysis-acidification stage in the two-phase anaerobic digestion process for kitchen wastes. It was found that high concentration of lactic acid feeding could decrease the COD load and deteriorate the effluent of the subsequent methanogenic process due to the propionic acid accumulation in the effluent. Therefore, it is necessary to avoid the presence of the lactic acid in the hydrolysis-acidification process for the improvement of the two-phase anaerobic digestion process of kitchen wastes.

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