Enhancement of Biogas Production in Two-Phase Anaerobic Fermentation System for Lower-Temperature Applications

Hongqiong Zhang, Wenzhe Li, Yong Sun and Xiangyou Wang*

College of Engineering, Northeast Agricultural University, Harbin, 150030, China

Abstract: For the purpose of enhancing the biogas production and operation stability of system at low temperature, a two-phase anaerobic fermentation facility for rural household energy generation was proposed. In this facility, the quantity of the fermented materials could be balanced by controlling the hydraulic retention time, which based on pH of the acidification tank and the fermentation tank. In addition, a portion of the biogas generated could be used to heat the acidification tank and the fermentation tank. Results shows that the optimal daily production rate of biogas for 1 m^3 fermentation liquid was about 1.47 L/L d^{-1} at a mixing ratio of cow: swine: chicken manure was 3: 1: 0.5. The production rate obtained in this study was more than four times higher than that from traditional single-phase processes (0.35 L/L d^{-1}). About 5.43 m^3 biogas can be produced daily per household with an average CH_4 content of 76.8%. The two-phase process developed in this work will also reduce environmental pollution and increase energy production efficiencies.

Keywords: Two-phase fermentation, Manure, Swine, Cow, Chicken, Anaerobic digestion, Biogas.

1. INTRODUCTION

Being the bio-energy of anaerobic digestion (AD) from ubiquitous animal manure, biogas is of great potential clean energy which can not only achieve sustainable development but also develop ecological agriculture and reduce environmental pollution in rural areas. AD is a multi-functional process that integrates environmental protection, renewable energy production, nutrients and water recycling [1, 2] It is also a common and yet very complex biological process in nature which decomposes agricultural wastes using anaerobic fermentation technology [3, 4]. The important factors that affect this process include temperature of tanks, pH, total solids, ratio of carbon to nitrogen of fermentation liquid, gaseous conditions, nutrients, fermentation liquid loading, stirring, degree of crushing, etc. [5-7]. Carbon is the main source of energy for anaerobic digestion and it is also the building blocks of microbial cells. Nitrogen is an essential nutrient for protein synthesis by the microbes [8, 9]. Chicken manure has rich nutrient but it is not suitable for anaerobic fermentation under normal temperature. Swine manure can be digested completely and more easily biodegradable and hence a good waste material for anaerobic fermentation [10, 11]. The ratio of carbon to nitrogen is an important factor that affects the microbial activities [12]. Cow manure was used as a ubiquitous feedstock in previous studies [13], and with a mixture of swine, cow and chicken manures can greatly improve the yield of biogas through anaerobic fermentation [14]. Recent investigation found that the methane production efficiency can be greatly increased by using mixed manures during anaerobic fermentation [15-17]. For example, pure chicken manure could not be anaerobically digested at a pH value lower than 6.0. However, a mixture of cow and chicken manure (1:1 or 2:1) was digested under the same condition. Results from recent experiments showed that the biogas production rate increased significantly at 25℃ or 30℃ [18].

Considering present issues of low temperature operation in conventional single-phase tanks, such as longer fermentation cycle, lower biogas production rate, slower degradation of raw material, more structure damages [19, 20], and it is difficult to maintain a balance between production and utilization of fatty acids [21, 22] and between acidogenesis and methanogens microorganisms with single-phase reactor, stable and economical operation of two-phase anaerobic fermentation system was proposed, which has potential to meet energy needs of rural households and ensure year-round production in the low temperature area of China [23-25], and it would play an important role in biomass utilization and rural industrialization. The existing researches of two-phase anaerobic fermentation were mostly focus on the influences of reactive conditions, co-acidification of cassava dregs and pig manure at a 4:6 ratio can improve the methane yield [26], pressure effects affected the pH value, biogas production and process stability *via* anaerobic filter [27]. However, there is no biogas digesters which can run stably throughout winter in low temperature area, even though several advantages of two-phase fermentation system, its performance has been restricted the utilization in cold regions [28]. This study showed a two reactors with

*Address correspondence to this author at the College of Engineering, Northeast Agricultural University, Harbin, 150030, China; Tel/Fax: +86-533-2786558; E-mail: wxy@sdut.edu.cn

E-ISSN: 1929-6002/14

one reactor was used to culture digestion bacteria and the other to store methanogens during the two-phase anaerobic digestion process. The optimal growth conditions for two different kinds of microorganisms could be achieved by controlling the operation parameters. The two-phase process would improve activity of organic load and methanogens [29] resulting in greater production of biogas, which is expected to stabilize the operation and improve the tolerance of the reaction system to load impact, and thus improve hydrolysis rate and anaerobic digestion. The presence of a large quantity of fibers in cow manure may result in a high conversion efficiency for methane because of the high carbon-nitrogen ratio [30-32].

In the present study, a two-phase anaerobic fermentation facility was designed and investigated for the energy production from animal manures. The facility can be operated at moderate temperature (35℃± 1℃). The main objective of the present work was to investigate the optimal operational parameters. The results will be valuable to large-scale energy production through continuous methane production, and reducing the impact of animal wastes on environments in low temperature area. Other benefits from proposed technology may include reduction in the cost for organic waste treatment and increase in the applicability of efficient anaerobic fermentation in biomass energy utilization in rural area.

2. MATERIALS AND METHODS

2.1. Experimental Set-Up

The experiments were performed in the greenhouse of Beidahuang Modern Agricultural Park of Harbin, China. The greenhouse was oriented north and south, and 6° of west. The dimension of the green house was about 100 m long and 7.5 m wide. The digester was built underground on the west side of the greenhouse. Durable no-dip PVC film with a thickness of 0.12 mm was used to cover the greenhouse. In the winter, a cotton quilt with a thickness of 40 mm was used to insulate the device from air.

Maintaining appropriate temperatures is a major challenge to operating an AD facility in north China. The heating system for the digester is shown in Figure **1**.

A hot-water boiler (20 in Figure **1**) was used to provide hot water to heat the acidified liquid in the acidification tank (1) and the fermentation liquid in the fermentation tank (2). The heating system was designed to balance the cost and energy consumption.

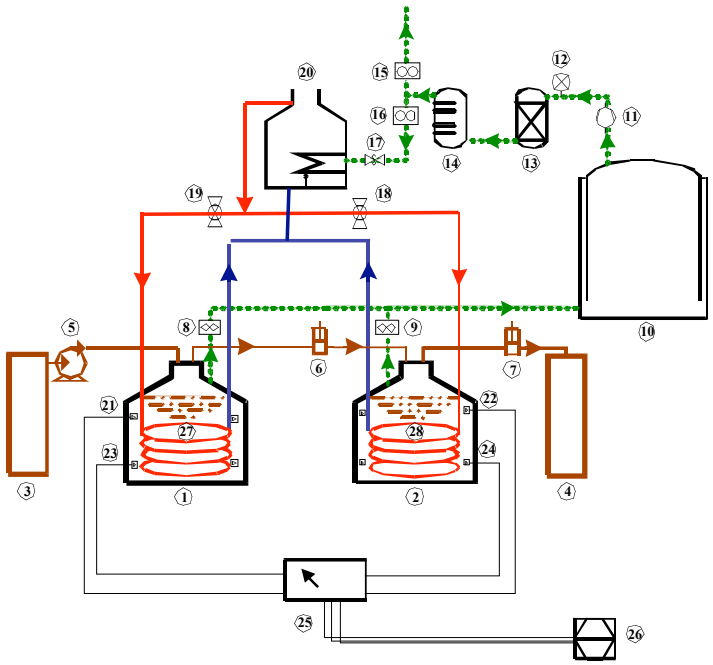

Figure 1: The heating system for the digester. Numbers are defined as follows: 1: Acidification tank; 2: Fementor tank; 3: Storing tank; 4: Biogas slurry tank; 5: Vacuum pump; 6 and 7: Portable pumps; 8,9,15, and 16: Flow meters of biogas 10: Biogas bag; 11: Booster pump; 12: Pressure gauge; 13: Desulfurizer; 14: Dehydrating tower; 17: Air valve; 18 and 19: Ball valves; 20: Marsh gas boiler; 21 and 23: Temperature sensors 22 and 24: PH sensors; 25: Data acquisition unit; 26: Computer memory; 27 and 28: Tube exchanger. Red lines represent hot water lines and blue for cooled water lines. Dashed green lines denote biogas lines. Dark yellow lines represent the liquid lines. Black thin lines denote the controlled lines.

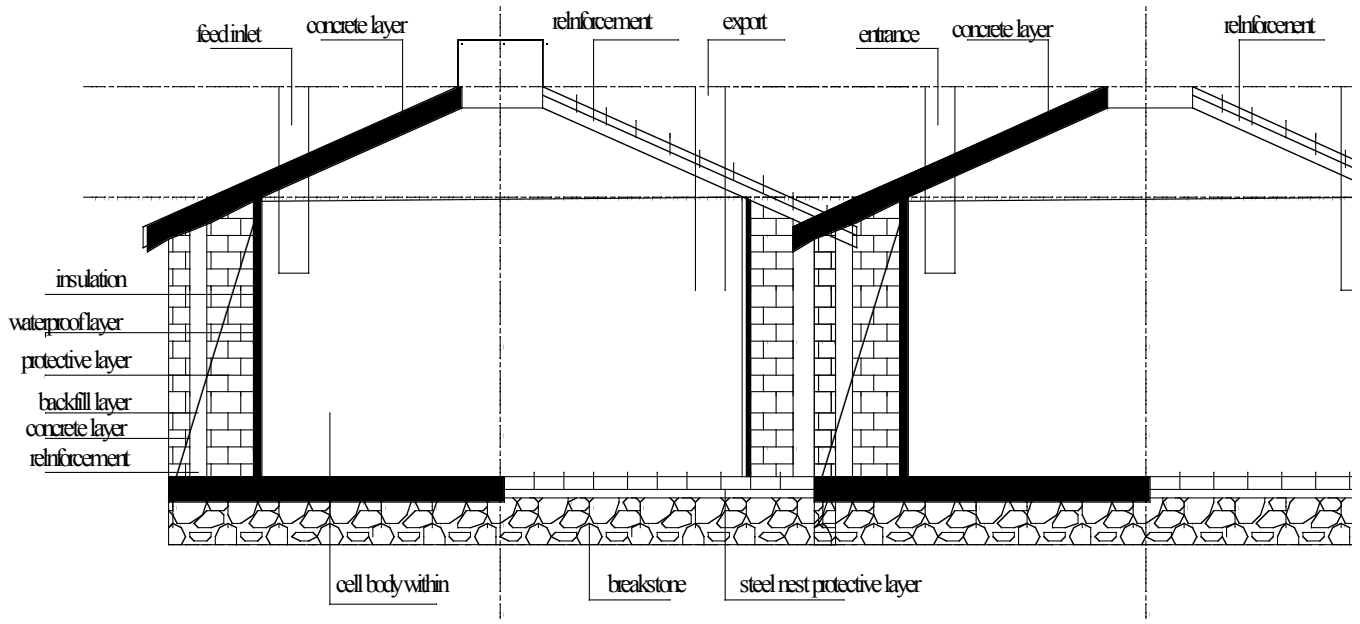

Figure 2: The structure of the rigid PVC tanks used in building the digester.

Biogas was recirculated from a biogas tank (10) through an air valve (17) after the gas was desulfurized (13) and dehydrated (14) into the boiler as the fuel to boil the water. The acidified liquid and fermented liquid were heated through a heat exchangers inside the acidification tank and the fermentation tank respectively. The temperature of the acidification tank and the fermentation tank was kept at about 35±1℃, in order to ensure desirable microbial activities for biogas production and to save energy by recirculating the biogas as the heating fuel.

2.2. Design of Biogas Digester

The two-phase anaerobic fermentation system (the digester) consisting of an acidification tank and a fermentation tank are shown in Figure **2**.

The tanks (Heilongjiang Jian Xin Technology Co., Ltd) were made of rigid PVC which is able to sustain freezing and bursting which may occur due to large temperature fluctuation. A cement liner was used to prevent corrosion and aging. The tanks were easy to install, maintain and replace. The tanks were covered by concrete tops. The pressures inside the tanks can be adjusted by an automatic device for stabilizing pressure, so that bursting of the tanks due to excessive pressures can be prevented. In addition, potential poisoning accidents can be avoided because operators are not necessary to enter the tanks for maintenance. The construction process for the digester is illustrated in Figure **3**.

2.3. Feedstock for Biogas Systems

Swine and cow manures were taken from Xiangfang Experimental Farm, chicken litter from Northeast Agricultural University Experimental Station. The inocula were collected from a composite microbial system in our lab. Table **1** shows the main properties of those waste materials. Manure had the highest TS% and VS% among the used waste materials. Cow-dung had the highest C/N ratio and chicken litter had the highest nitrogen content among those materials.

The experiments began from January 25 and ended at February 25. Experimental group I and experimental group II were performed at TS ratios of 3:1:1 and 3:1:0.5 (cow manure: swine manure: chicken litter), prescribed based on our previous studies, respectively [14]. The TS of the initial liquid (2 m^3) entering the digester was adjusted to 10%, and then inoculated into the acidification tank all at once. Experimental group I and experimental group II were performed four times, and each experiment period lasted 4 days. Two thermometers were installed inside and outside the greenhouse respectively. Two temperature sensors and two pH sensors were installed in the acidification tank and fermentation tank at a liquid level of about 20 cm and 45 cm, respectively. Data were acquired during the entire experiment period. The biogas flow rate was recorded manually every 12 h. Samples were taken from the acidification tank and fermentation tank for tests described in 1.6.

a

b

Figure 3: Construction process.

Table 1: Characteristic of Fresh Manures

Character	TS%[a]	VS%[b]	C/N[c]	Nitrogen (g/kg)
Cow	19.3	76.6	26.5	16.33
Swine	40.4	79.9	14.5	23.7
Chicken	27.1	63.3	9.31	24.5
Inocula	4.40	28.1	--	--

[a]TS% is percentages of the total solid content; [b]VS% is volatile solid content; [c]C/N is the carbon to nitrogen ratio.

2.5. Data Processing

Temperature and pH were measured with the thermometers (21 and 23 in Figure **1**) and pH sensors (22 and 24 in Figure **1**), respectively. The volumetric rate of biogas was measured using a biogas flow meters (8, 9, 15 and 16 in Figure **1**). Total solid content (TS) and volatile solids content (VS) were measured using a drying method. The CH_4 content was measured using gas chromatography (Agilent 6890, Agilent) [14]. Pure CH_4 and pure CO_2 were compared with unknown sample to obtain chromatogram map, and then the area of chromatographic peak is calculated for the content of CH_4.

$m_i\% = \frac{A_i}{\sum_{i=1}^{n} A_i} \times 100\%$, A_i is area of chromatographic peak. (1)

The volatile fatty acid (VFA) was measured using gas chromatography (Agilent 6890, Agilent). Sample was diluted by distilled water with a ratio of 1:1 and a sulfuric acid solution of 6 mol·L^{-1} was added. The solution was centrifuged for about 10 min with a speed of 12 000 r·min^{-1} and then a microporous filtering film of 0.45 mm was used for filter press. The pH value was adjusted to 2 by adding formic acid. An average value was taken from three samples. The daily volume of the produced biogas for 1 m^3 fermentation liquid is calculated according to (2),

$$V = Q / D \cdot G \quad (2)$$

Where V represents average rate of biogas daily (m^3 $m^{-3}d^{-1}$), Q represents production (m^3), D represents fermentation days (d) and G represents volume of biogas tank (m^3).

Weight percent is M_0,

$$M_0 = \frac{\sum X_i m_i}{\sum X_i + W} \times 100\% \quad (3)$$

Where M_0 represents Weight percent (%), W represents water (Kg), m_i represents Weight percent of TS (%) and Xi represents quality of material (Kg).

3. RESULTS AND DISCUSSION

3.1. Indoor and Outdoor Temperature Variability

The history of the indoor and outdoor temperatures during the 32 days experiment period is shown in Figure **4**.

The average temperature outdoor was -17.8℃, and the average temperature indoor was 12.0℃, and the average temperature difference between indoor and outdoor was about 30℃. Note that under such a cold condition there is a need for developing digesters that can be operated continuously throughout the winter to produce enough biogases from household waste materials to meet household energy needs in low temperature area.

3.2. The pH Values in the Acidification Tank

The pH values in the acidification tank decreased steadily with time during hydrolysis acidification process was showed in Figure **5**.

The two experiments following the same trend for the pH values. During hydrolysis acidification, high molecular weight compounds were degraded acidogenesis microorganisms into low molecular weight compounds, volatile fatty acid (VFA), which lowered the pH values. The initial organic matters provided nutrients for growth and reproduction of

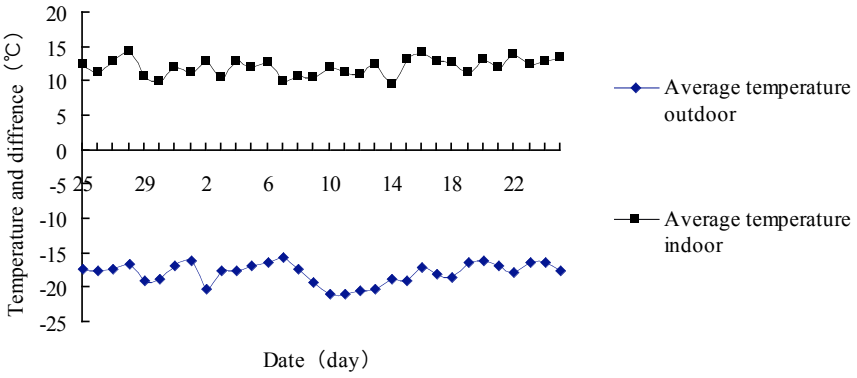

Figure 4: Temperature condition during test.

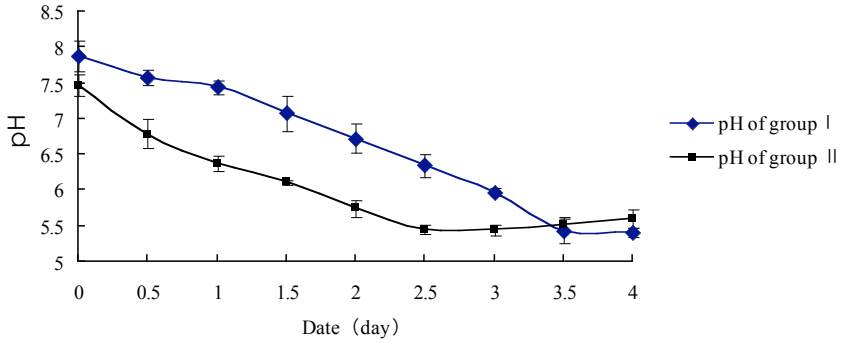

Figure 5: pH of acidification tank.

acidogenesis microorganisms, which adapted to the environment and metabolized rapidly. Since methanogens microorganisms adapted environment more slowly, their activities were inhibited by the rapidly decreasing pH, resulting in rapid accumulation of VFA and further increase in acidity.

In later time, the methanogens microorganisms gradually adapted to the environment, and began to consume fatty acid more rapidly. The nitrogen-containing organic matter began to be decomposed to produce amide salts (ammonification), pH value in the acidification tank declined and slowly rose. The pH value in experimental group II reached a minimum of 5.45 and then rose slowly after 2.5th day, compared with slight increase in pH value for experiment group I after 4 days. Because the nitrogen content in chicken manure was much more than cow and swine manures. The ammonium nitrogen in the manures could increase the activity of methanogens microorganisms, hence the basicity of the reaction system in the acidification tank [12, 28]. After acidification, the biogas slurry was sent to the fermentation tank and raw materials were added into the acidification tank.

3.3. VFA of Fermentation Tank

Initially from the first half day of the experiments, the VFA content for experimental group I had a sluggish stage at first and then decreased rapidly from the half past day as shown in Figure **6**.

The VFA content for experimental group II kept decreasing until it reached a minimum of 3700 mg L^{-1} on the 4th day. During the experiments, the acidogenesis microorganisms in the liquid were sent from acidification tank to fermentation tank, and those microorganisms continued to degrade organic compounds into low molecular organic acids. The organic acids could not be utilized by the methanogens immediately, and therefore the VFA content (7335 mg L^{-1}) was higher in the beginning of the experiments. Compared with that for experimental group I, the VFA content for experimental group II was higher in the beginning of the experiments with a TS ratio of 3: 1: 0.5. The VFA were reduced rapidly from 7335 mg L^{-1} to 3933 mg L^{-1}, after the methanogens microorganisms gradually adapted to the environment during fermentation process. However, the VFA content decreased more slowly at the end because content of organic matter was reduced, and the activity of microbes decreased during the fermentation process.

3.4. The Daily Biogas Production Rates

Figure **7** shows the daily biogas production rates for experimental group I and experimental group II.

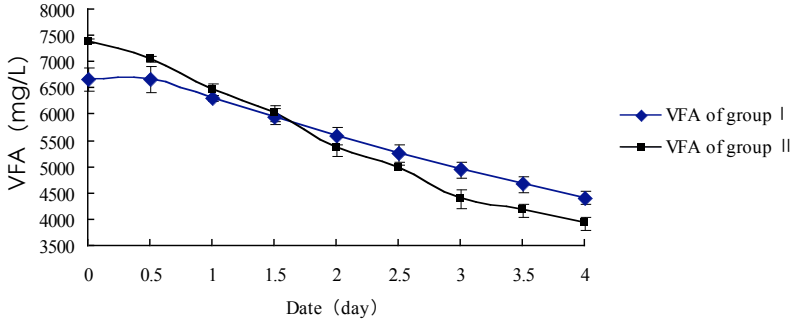

Figure 6: VFA of fermentation tank.

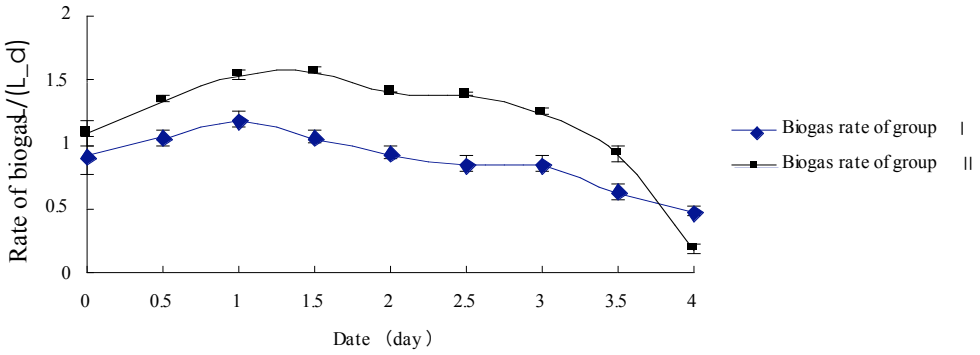

Figure 7: The rate of biogas.

Experimental group II produced more gases than experimental group I until almost the end of the 4th day. However at the end of the 3th day, the production rates of experimental group II started to decrease more rapidly than experimental group I, because lower concentration of VFA for experiment II than for experiment I. Initially, the biogas production rate increased in the first day of the experiments and then reached a relative stable level for about another 2.5 days. Because the microbial inoculation had a lag phase when it was introduced into a new substrate, the microbes liquefied organic matter so that the biogas production rates were lower on the first day of the experiments. Meanwhile, the concentration of organic matter would affect the methane production rates. Low concentration of organic matter facilitated the biogas production. In the beginning of the experiments, the concentration of organic matter was higher, the microbe activity increased so that the biogas production rate was higher. However, the organic matter continued to be digested and the microbe activity decreased, so the biogas production rate reduced gradually. In addition, sulfate bacteria and profitless bacteria for digestion would compete for hydrogen with methanogens microorganisms, thus the rate of biogas production decreased gradually. The average biogas production rate for experimental group I was 0.80 L/L d^{-1} and experimental group II was 1.19 L/L d^{-1}. We can control the acidification time by regularly samples and analysis the pH value of acidification tank, so as to produce more organic acids that can increase the activity of the methanogens microorganisms.

3.5. Use of Biogas to Maintain Temperature

The biogas produced was burned in a marsh gas boiler (20 in Figure **1**) to provide heat to maintain the temperatures of acidification tank and fermentation tank. The amount of biogas needed to maintain temperature of the fermentation liquid is shown as Figure **8**. In the beginning of the experiments, more biogas was needed to heat both tanks. After the liquids were heated, the biogas consumption rates decreased and were kept at ~ 2 m^3/day. The largest amount of biogas that used to heat new liquid was 15.97 m^3 on the first day. Since the average biogas production was 8.01 m^3, household could get 5.43 m^3 biogas for cooking and heating every day.

3.6. Variation of CH_4 Content

The CH_4 content increased in the beginning and became stable after 2 days of experiments as was shown in Figure **9**.

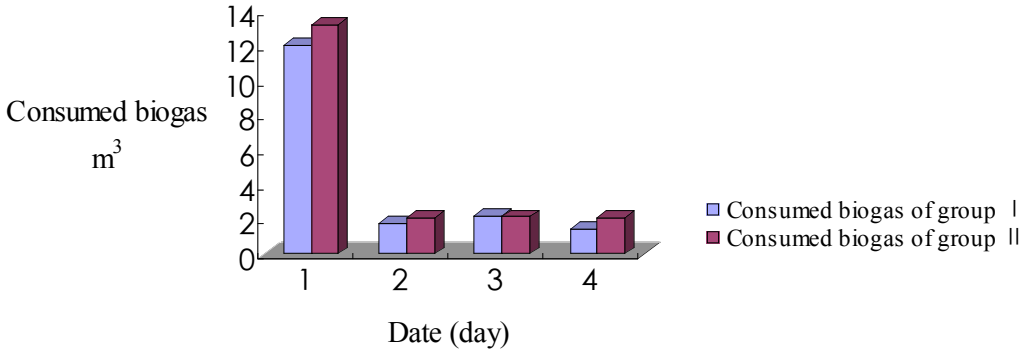

Figure 8: Consumed biogas.

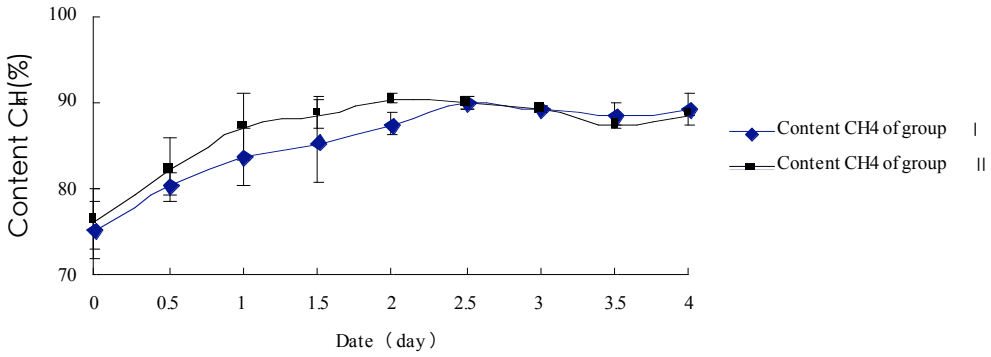

Figure 9: Variation of CH_4 content.

The maximum of CH_4 content for experimental group I and II were 85.5% and 86.8%, respectively, and the average are 65.1% and 76.8%, respectively.

4. CONCLUSIONS

The work proposed an anaerobic digestion facility that separate acidogenic and methanogenic phases into two independent processing units. The facility created optimal environment for acidogenic bacteria and methanogenic bacteria by controlling hydraulic retention time making the activities of both bacteria maximized, which can improve the treatment efficiency and ensure stable operation, a small proportion of the biogas can used to heat the acidification tank and the fermentation tank. In addition, the equipment built underground can reduce energy consumption. The biogas production rates and the CH_4 content increased significantly by using a mixture at the ratio of cow: swine: chicken manure was 3: 1: 0.5 and the optimal biogas production rate was 1.47 L d^{-1}. This can produce a volume of about 5.43 m^3 biogas daily with a CH_4 content of about 76.8%, containing energy sufficient for daily cooking and heating for a three people family household. The conversion of animal waste materials into a useful energy source will alleviate environmental pollution and provide an alternative promising energy source in winter in the low temperature area.

ACKNOWLEDGEMENTS

The study was supported by grants from Doctoral Fund of Ministry of Education of China (20122325120024), Natural Science of Fund Heilongjiang Province (E201320), The Key Technology Projects of Heilongjiang Province (GZ11B503), Natural Science Project of Education Department of Heilongjiang Province (12521029), and the 12th Five Years Key Programs for Science and Technology Development of China (2011BAD15B04). Heilongjiang Longrui Renewable-energy Technology Co., Ltd is acknowledged for helping with materials and equipment. We thank Dr. Nalladurai Kaliyan and Dr. Min Min (Department of Bioproducts and Biosystems Engineering, University of Minnesota, Twin Cities, USA) for their guidance and suggestions. We thank Richard Griffith (3M Corporate Research Process Lab) for better suggestions. Heilongjiang Jian Xin Technology Co., Ltd is acknowledged for helping with materials of the tanks, and thank Beidahuang Modern Agricultural Park of Harbin for helping with the test area.

REFERENCES

[1] Chang JS, Lee KS, Lin PJ. Biohydrogen production with fixed-bed bioreactors. Int J Hydrogen Energy 2002; 27: 1167-74. http://dx.doi.org/10.1016/S0360-3199(02)00130-1

[2] Sun ZJ. Biomass industry and its developmental trends in China. Trans CSAE 2004; 20: 1-5.

[3] Ward AJ, Hobbs PJ, Holliman PJ, Jones DL. Optimization of the anaerobic digestion of agricultural resources. Bioresour Technol 2008; 99: 7928-40. http://dx.doi.org/10.1016/j.biortech.2008.02.044

[4] Angelidaki I, Ahring BK. Methods for increasing the biogas potential from the recalcitrant organic matter contained in manure. Water Sci Technol 2000; 41: 189-94.

[5] Vavilin VA, Rytov SV, Lokshina LY. A description of hydrolysis kinetics in anaerobic degradation of particulate organic matter. Bioresour Technol 1996; 56: 229-37. http://dx.doi.org/10.1016/0960-8524(96)00034-X

[6] Sánchez, E, Borja, R, Weiland P, Travieso, L, Martín, A. Effect of temperature and pH on the kinetics of methane production, organic nitrogen and phosphorus removal in the batch anaerobic digestion process of cattle manure. Bioprocess Eng 2000; 22: 247-52. http://dx.doi.org/10.1007/s004490050727

[7] Balat M, Balat H. Biogas as a Renewable Energy Source-A Review. Energy Sources, Part A: Recovery, Utilization, and Environmental Effects 2009; 31: 1280-93. http://dx.doi.org/10.1080/15567030802089565

[8] Ismail Z K, Abderrezaq SK. Employment of Anaerobic Digestion Process of Municipal Solid Waste for Energy. Energy Sources. Energy Sources, Part A 2007; 29: 657-68.

[9] Lissens G, Vandevivere P, De Baere L, *et al*. Solid waste digester: process performance and practice for municipal solid waste digestion. Water Sci Tech 2001; 44: 91-102.

[10] Ren NQ, Liu M, Wang AJ, *et al*. Organic Acids Conversion in Methanogenic-phase Reactor of the Two-phase Anaerobic Process. China J Environ Sci 2003; 24: 89-93.

[11] Li WZ, Wang ZJ, Wang LL, *et al*. Factors affecting acetic acid content in the process of high-concentration hydrolysis and acid genesis of cow manure. Trans CSAE 2008; 24: 204-8.

[12] MØller HB, Sommer SG, Ahring BK. Biological degradation and greenhouse gas emissions during pre-storage of liquid animal manure. J Environ Qual 2004; 33: 27-36. http://dx.doi.org/10.2134/jeq2004.2700

[13] Dan L. The Experimental Research on Anaerobic Fermentation Characteristics of Mixed Livestock Manure. J Northeast AgricUniv 2008; 22-27: 33-5.

[14] Sun Y, Li WZ, Zhang HQ. Biogas System in Low temperature area by Two-Phase Anaerobic Fermentation Process. Trans CSAE 2012; 9: 196-201.

[15] Mata-Alvarez J, Cecchi F, Pavan P, Llabres P. The performance of digesters treating the organic fraction of municipal solid wastes differently sorted. Biological Wastes 2000; 33: 181-99. http://dx.doi.org/10.1016/0269-7483(90)90004-C

[16] Dan M. Making electricity from manure. Biocycle 2003; 44: 52-5.

[17] Liu RH, Hao YY, Wu LJ. Effect of temperature on biogas yield and property during anaerobic fermentation process of pig dung. Renew Energ 2006; 5: 32-5.

[18] Xu HW. The study of anaerobic fermentation efficiency of cow and chicken manure in different temperature. J Northeast Agric Univ 2007; 23-34.

[19] Cantrell KB, Ducey T, Ro KS, Hunt PG. Livestock waste-to-bioenergy generation opportunities. Bioresour Technol 2008; 99: 7941-53. http://dx.doi.org/10.1016/j.biortech.2008.02.061

[20] Chynoweth, DP, Owens JM, Legrand R. Renewable methane from anaerobic digestion of biomass. Renew Ener 2000; 22: 1-8. http://dx.doi.org/10.1016/S0960-1481(00)00019-7

[21] Labatut RA, Angenent LT, Scott NR. Biochemical methane potential and biodegradability of complex organic substrates. Bioresour Technol 2011; 102: 2255-64. http://dx.doi.org/10.1016/j.biortech.2010.10.035

[22] Yu FB, Luo XP, Guang LB, *et al*. Research advances in biogas fermentation microorganism. J Anhui Agric Sci 2008; 36: 15658-660.

[23] Sinbuathong N, Sirirote P, Sillapacharoenkul B, *et al*. Biogas Production from Two-stage Anaerobic Digestion of Jatropha curcas Seed Cake. Energy Sources, Part A 2012; 34: 2048-56. http://dx.doi.org/10.1080/15567036.2012.664947

[24] Neves L, Oliveira R, Alves MM. Co-digestion of cow manure, food waste and intermittent input of fat. Bioresour Technol 2009; 100: 1957-62. http://dx.doi.org/10.1016/j.biortech.2008.10.030

[25] Bouallagui H, Torrijos M, Godonc JJ, *et al*. Two phases anaerobic digestion of fruit and vegetable. Wastes: bioreactors performance. Biochem Eng J 2000; 21: 193-7. http://dx.doi.org/10.1016/j.bej.2004.05.001

[26] Ren JW, Yuan XF, Li J, Ma Xg, *et al*. Performance and microbial community dynamics in a two-phase anaerobic co-digestion system using cassava dregs and pig manure. Bioresour Technol 2014; 155: 342-51. http://dx.doi.org/10.1016/j.biortech.2013.12.120

[27] Chen YL, Rößler B, Zielonka S, Lemmer A, Anna-Maria W, Jungbluth T. The pressure effects on two-phase anaerobic digestion. Appl Energ 2014; 116: 409-15. http://dx.doi.org/10.1016/j.apenergy.2013.11.012

[28] Xia JQ, Bi JY, Zheng GX, *et al*. Temperature test surface of earth and effects on biogas generating pit in winter. J Northeast Agric Univ 2009; 40: 121-4.

[29] Bougrier C, Delgenes JP, Carrere H. Impacts of thermal pre-treatments on the semi-continuous anaerobic digestion of waste activated sludge. Biochem Eng J 2007; 34: 20-7. http://dx.doi.org/10.1016/j.bej.2006.11.013

[30] Castillo M, EF, Cristancho DE, Arellano AV. Study of the operational conditions for anaerobic digestion of urban solid wastes. Waste Management 2006; 26: 546-56. http://dx.doi.org/10.1016/j.wasman.2005.06.003

[31] Yaldiz O, Sozer S., Caglayan N, *et al*. Methane Production from Plant Wastes and Chicken Manure at Different Working Conditions of a One-stage Anaerobic Digester. Energy Sources, Part A 2011; 33: 1802-13. http://dx.doi.org/10.1080/15567030903419463

[32] Ghash S, Henry MP, Sajjad A, *et al*. Pilot-Scale gasification of municipal solid wastes by high-rate and two-phase anaerobic digestion (TPAD). Water Sci Tech 2000; 41: 101-10.

Received on 15-05-2014 Accepted on 17-06-2014 Published on 29-08-2014

DOI: http://dx.doi.org/10.6000/1929-6002.2014.03.03.5