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Improving methane production during the anaerobic digestion of waste activated sludge: Cao-ultrasonic pretreatment and using different seed sludges

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Abstract

Three individual seed sludges, which domesticated by filter paper (SS1), food waste (SS2) and grease (SS3)，respectively, were used for enhancing the methane production of waste activated sludge (WAS). Also CaO-ultrasonic pretreatment was performed on WAS to evaluate the effectiveness on improving efficient anaerobic digestion (AD). The results showed that WAS being acidated for 24 h after CaO-ultrasonic pretreatment was an effective method for increasing initial methane production rate. The daily concentration of volatile fatty acids (VFAs) during the AD course showed that the propionic was easier to be reduced after adding seed sludge. The optimum seed sludge for improving methane production and biodegradability of WAS was SS3, which led to an increase in the methane production of 68.92% and VS reduction of 69.20% higher than the control. This pretreatment combined with adding optimum seed sludge can greatly improve clean energy generation from WAS.

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Keywords: Waste activated sludge; anaerobic digestion; Cao-ultrasound pretreatment; seed sludge; methane production;

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1. Introduction

Activated sludge process is an effective method for sewage treatment. During AD process, large amounts of excess WAS will be produced in the process. Early in 2007, 4.54×10^6 tons of dry WAS was produced in China[1]. And according to prediction, the production of WAS in China will be more than 6×10^8 tons[2]. WAS not only contains various pathogens and persistent organic pollutants, but also contains large amount of inorganic matters. Traditional handing methods such as landfill and incineration will lead to secondary environment pollution[3].

AD treatment has been widely studied for stabilizing WAS[4]. The advantages of this method were obvious: (1) effective reduction in sludge volume, (2) produce clean energy in the form of biogas, (3) environment friendly reaction products, (4) the lower treatment cost requirement and spaces than the traditional process[5,6,7]. Though AD process brought us environmental and economic benefits, there were still some defects: (1) the limited conversion of organic matter, (2) long retention times (3) non-biodegradable organic structures[8,9,10]. These defects are mainly caused by the retarded hydrolysis of WAS. The microbial cells and membrances in the WAS are not easily to be penetrated by the hydrolytic enzymes[11]. Therefor, various cell disruption methods have been explored to enhance the hydrolysis rate and methane production such as physical, chemical, biological and combined pretreatments[12].

Among these technologies, ultrasonic pretreatment has the advantage of destroying microbial cells to extract intracellular material[13]. Additionally, the short chain acids production can be enhanced and AD process can be shortened due to the release of soluble organic substrates from WAS[14]. In Teihm's study, the VS removal of WAS increased from 21.5% to 33.7% (+36%) after ultrasonic pretreatment at 41 kHz for 150min[15]. Another research showed that 24% increasement of biogas production was reached after ultrasonic pretreatment at 20 kHz for 60s[16].

Similarly, alkaline pretreatment was examined to be an effective method to destroy the cell surface structure and make the the cellular substances more susceptible to enzymatic action. Moreover, alkaline pretreatment needed simple device which was easy to be operated[17]. Combined pretreatment of alkaline and ultrasound pretreatment was demonstrated to be able to reduce the dewaterability of WAS more effectively[18]. NaOH and KOH was used as the alkaline part because of the strong alkalinity. However, a high concentration of Na^+ or K^+ may cause AD process slow down and even inhibit the activity of methanogens[19]. $Ca(OH)_2$, as a kind of alkalis, also can improve the AD efficiency of WAS and have less inhibiting effect than NaOH or KaOH[20]. Lime, as a cheap industrial raw material which mainly contains CaO, can improve carbohydrate digestion and biogas production with smooth cordgrass as fermentation substrate[21]. However, few reports have been published about pretreatment of WAS using CaO as an alternative to NaOH or KaOH.

In order to enhance the effective of WAS degradation, the pretreatment methods have been widely explored, but the effect of seed sludge addition was often ignored. Anaerobic seed sludges may come from the bottom from the pond, deep in the sewer, cattle cesspool or the pig cesspool[22]. Also inoculums can be domesticated by pig manure, cow dung, cornstalk, vinasse or food waste[23,24]. Different inoculum may result in different biogas production rate, total biogas production or methane percentage because of the special species and amounts distribution. Using the appropriate seed sludge can shorten start-up period and improve the methane production[24].

Consequently, in our research, three kinds of seed sludges domesticated with grease, food waste and filter paper, respectively were added into the WAS which pretreated in CaO-ultrasonic way, aim to: (1) evaluate the effect of CaO-ultrasonic pretreatment, (2) enhance the methane production rate, VS reduction and total methane production of WAS, (3) explore a better combined method to deal with WAS, (4) lay a theoretical foundation for further industrial application.

2.Materials and methods

2.1 WAS and seed sludges

The WAS was collected from department of dehydration of Xianyanglu wastewater treatment plant (Tianjin, China). Total solids (TS) of WAS was diluted from 20% to 8% before used in the experiments. The basic characteristics of WAS is shown in Table. 1.

The resource of seed sludges was from the sewage treatment plant in Chengdu, Sichuan, China. The sludge was separated into three parts and domesticated in the same conditions except only one factor, inoculation of carbon source. The first inoculum was grease with daily dosage of 1g·L-1d-1. The second and the last inoculums were food waste and filter paper with the same daily dosage. After a month of domesticating, the basic characteristics of the three kind of seed sludges are shown in Table 1.

Table 1. Characteristics of WAS, PWAS and seed sludges.

Sludges	Domesticated by	pH	TS (%)	VS (%)	VS/TS (%)
WAS	-	6.8	8.11	4.46	54.99
PWAS	-	9.5	8.53	4.44	52.07
APWAS	-	7.5	8.49	4.38	51.59
Seed sludge 1 (SS1)	Grease	6.7	3.81	1.65	43.41
Seed sludge 2 (SS2)	Food waste	6.8	0.89	0.55	61.24
Seed sludge 3 (SS3)	Filter paper	6.8	2.37	1.22	51.48

WAS was first mixed evenly with CaO with a dose of 40mg g-1 TS in a 5.0 L beaker. Then the mixed sample was disintegrated in an ultrasonic reactor (TZ-U1, Beijing, China) with a working volume of 2 L. The reactor equiped with two 75 W ultrasonic probes at its bottom which can produce a 20 kHz ultrasound. After a handling time of 30 min, CaO-ultrasonic pretreatment for WAS was finished.

2.3 Batch AD experiments

Batch AD was conducted in anaerobic batch reactors of 250 mL BPN culture bottles with hermetically sealed stoppers at capacity mesophilic conditions to examine biogas production, variation of VFAs and WAS degradability of raw and pretreatment sludge samples. Each bottle of test group was filled with 150 mL of substrate and 50 mL of seed sludge. And the control groups were filled with 150 mL of substrate and 50 mL of distilled water. The initial pH was natural. Because of a high pH at 9.5 of pretreated WAS may inhibit the AD process, another batch experiment was initiated with PWAS which was acidated under natural conditions for one day. The initial pH of acidulated PWAS (APWAS) was tested around 7.5 in the previous experiments. All the batch AD experiments were shown in Table 2. Each group set up three parallel experiments. The reactors were laid in a constant temperature incubator at 37±2℃ for 25 days. Samples were taken every 24 h to examine daily biogas production, methane content and VFAs conditions. In the end of batch AD experiments, samples were collected to test TS and VS conditions.

Table 2. Experimental design for batch AD experiments.

Trials	Substrate (mL)			Seed sludge (mL)			Distilled water (mL)
	WAS	Preatment WAS (PWAS)	Acidated preatment WAS (APWAS)	SS1	SS2	SS3	
1	150	-	-	50	-	-	-
2	150	-	-	-	50	-	-
3	150	-	-	-	-	50	-
4	150	-	-	-	-	-	50
5	-	150	-	50	-	-	-
6	-	150	-	-	50	-	-
7	-	150	-	-	-	50	-
8	-	150	-	-	-	-	50
9	-	-	150	50	-	-	-
10	-	-	150	-	50	-	-
11	-	-	150	-	-	50	-
12	-	-	150	-	-	-	50

2.4 Analysis methods

Total solids (TS) , volatile solids (VS) and pH were measured according to the standard methods (APHA, 1995)[25].

The analyze of VFAs contained acetic acid, propionic acid and butanoic acid. 1ml of samples was extracted from the samples, and acidified by adding 5% vitriol before centrifuging at 10000 r/min for 10 min. The resulting supernatants of 2-μl were injected for gas chromatography analysis. VFAs were determined using a Shangfen GC112A gas chromatograph equiped with a packed column (GDX103, length 10 m; i.d. 2 mm; o.d. 3 mm). The flow rate of H_2 carrier was 20 ml/min. The temperature of oven and the flame ionization detection system were 210 ℃ and 270 ℃, respectively. After run time of 7 min, the VFAs were calculated by comparing the peak area with standard substances.

The biogas was collect and quantify by a gas device. The methane content of biogas was measured using a gas chromatograph (Micro GC490, Agilent) equipped with a packed column(MSA/PPU, Agilent). The injector and column temperatures were 100 and 50 ℃, respectively. H_2 was used as the carrier gas with a pressure condition at 100 k Pa.

3. Results and discussion

Biogas is produced from AD process and manly contains methane and carbon dioxide (CO_2). Methane, as the main energy material of biogas, is an important index to assess performance of AD[26]. The AD process can be divided into four steps: hydrolysis, acidification, acetogenesis and methanogenesis. In the first two steps, organic matters are converted into small molecules and turned into volatile fatty acids(VFAs), such as acetic acid, propionic acid and butanoic acid, which can be used to produce methane and CO_2 in the acetogenesis and methanogenesis process[27]. So the concentration of VFAs during AD is also an important factor to reflect the AD efficiency. To investigate the effects of CaO-ultrasonic pretreatment and different seed sludges, the cumulation of methane production and VFAs concentration during the AD of 25 d are shown in Fig. 1-3.

3.1. Comparison among different seed sludges for AD of WAS

The cumulative methane production of WAS with different seed sludges were calculated based on the daily methane production as shown in Fig.1(a). All the groups started to produce methane no more than 3 days. On the 15th day, the cumulative methane production of WAS with SS3 reached to 983.3 mL. The cumulative methane production of WAS were only 674.1(SS1), 736.7 mL(SS2) and 225.2 mL(control) at the same time. In the end of the 25 days' batch experiments, the cumulative methane production obtained by seed sludges with SS1, SS2 and SS3 were 1178.1 mL, 1236.8 mL and 1211.9 ml, respectively. The effect of different seed sludges for cumulative methane production of WAS was very similar. The enhancements of methane production were 43.3%(SS1), 50.5%(SS2) and 47.5%(SS3) higher than the control check of 821.9 mL, respectively. Thought the highest enhancement of methane production of WAS was achieved by SS2, it is easy to be found that there was a higher methane production rate of WAS by SS3 than other seed sludges and control check.

The VFAs concentration during the AD process of WAS by different seed sludges is shown in Fig. 1(b). On the first day of AD process, the total VFAs concentration reached up to 1814.7 mg/L, 1677.3 mg/L, 1005.8 mg/L and 1603.7 mg/L by the seed sludges of SS1, SS2, SS3 and control, respectively. Acetic acid was the main component of VFAs (more than 70%) except control. The control group contained butanoic acid of 52.8%, propanoic acid of 7.4% and acetic acid of 39.8%. The highest concentration of VFAs were achieved on day 3 (2187.6 mg/L), day3 (1869.5 mg/L), day 1 (1005.8 mg/L) and day 12 (1321.9 mg/L) by SS1, SS2, SS3 and control, respectively. Then the VFAs concentration of each group gradually decreawsed after reaching the highest VFAs level. This result showed that the efficiency of hydrolytic and acidification had been improved by the seed sludges addition. As we know, acetic acid can be more easily used by methanogens to produce methane and CO_2 than propionic acid[28]. In our research, as the experiments went on, acetic acid and butanoic acid decreased gradually. And the butanoic acid disappeared after 13 d in all groups. The propionic acid had been accumulated from the beginning of the batch experiments to the highest of 966.3 mg/L on day 13(SS1), 1150.0 mg/L on day 13(SS2), 530.9 mg/L on day 12(SS3) and 1121.9 mg/L on day 16(CK). And the propionic became the main component of VFAs instead of acetic acid. On the 15th day, the concentration of propionic acid decreased to 21.4 mg/L by SS3. At the same time, there still were a large amount of propionic acid in other groups, which were 788.5 mg/L(SS1), 971.6 mg/L(SS2) and 622.3 mg/L(SS3), respectively. The concentration of propionic acid decreased to a low level of 25.4 mg/L by SS1 on day 19. And 2 days later, the concentration of propionic acid decreased to 28.7 mg/L by SS2. Even in the end of batch experiments, there was still a lot of propionic acid of 879.6 mg/L left in control check. It is demonstrated that SS3 has stronger ability turning VFAs into methane than other seed sludges or control check.

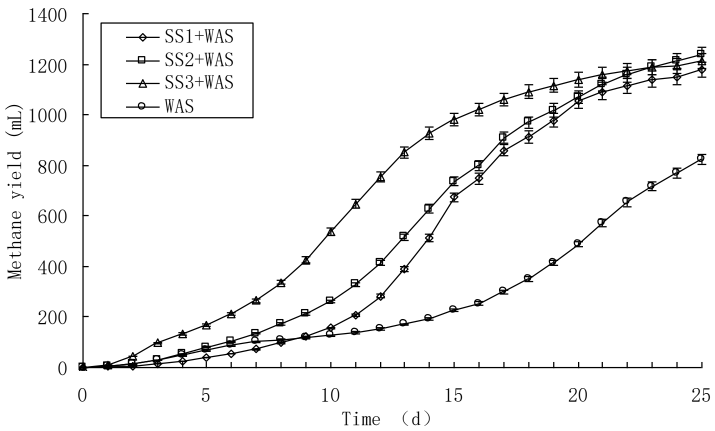

Fig.1 (a) Effect of seed sludges on methane yield of WAS.

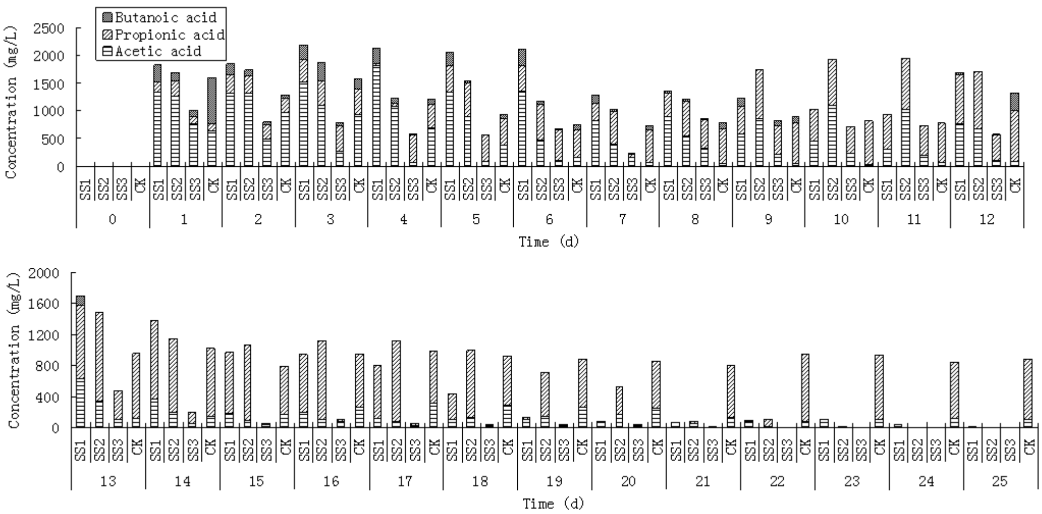

Fig. 1(b) The VFAs concentration during the AD process of WAS.

The results showed that among the three kinds of seed sludges, the most effective for AD of WAS was SS3 addition, which can greatly improve utilization rate of VFAs and methane production.

3.2. Comparison among different seed sludges for AD of PWAS

The effect of seed sludges on methane yield from PWAS AD is shown in Fig 2(a). Unlike WAS, there was a long lag time of methane production for PWAS. In the first week, there was almost no methane produced in all groups. Then the PWAS with SS1 and SS3 first began to produce methane. The lag time of PWAS with SS2 lasted for 18 days. And the longest lag time(23 days) for methane production was achieved by control check (PWAS with no seed sludges). Zhai et al. studied the effect of 5 different initial pH (6.0, 6.5, 7.0, 7.5 and 8.0) on AD. He found that the lag-phase times of treatments with initial pH of 6.5 and 7.5 were 10 days shorter than the group with pH of 8.0[29]. In Zhang's research, to achieve the maximum total biogas production, the optimal initial pH was controled at 6.81. And a high pH inhibited the AD process and reduce the biogas production in a certain time[30]. In our research, as shown in table 1, the pH of PWAS was 9.5, which was the main factor to cause such a long lag time during AD process of PWAS.

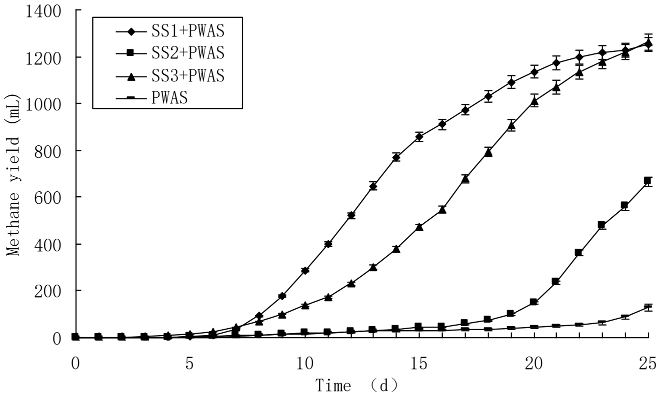

Fig. 2(a) Effect of seed sludges on methane yield of PWAS.

After 25 days' batch experiment, the cumulative methane production of PWAS without seed sludges was only 128.6 mL(control check). And the cumulative methane production obtained by seed sludges with SS2 was 666.7 mL, which was 46.1% less than that of WAS (1238.8 mL). The cumulative methane production of PWAS with SS1 and SS3 was 1251.1 mL and 1262.2 mL, which were 6.2% and 4.2% higher than those of WAS(1178.1 mL and 1211.9 mL) with same seed sludges. Though the negative effects of pH were obvious, SS1 and SS3 had a strong ability to adapt the initial alkaline environment. Moreover, the cumulative methane production of WAS after CaO-ultrasonic pretreatment had been enhanced in SS1 and SS3 groups.

The concentration of VFAs during the AD process of PWAS is shown in Fig.2(b). During batch experiments, the total VFAs of each group gradually accumulated and reach to the highest at 4194.2 mg/L(SS1), 4883.9 mg/L(SS2), 3879.0 mg/L(SS3), and 4980 mg/L(CK) on the 5th day, 9th day, 7th day and 13ed day, respectively. Because of the consumption of VFAs to produce methane by methanogens and the reduction of available organic matter in the sludge, the concentration of total VFAs began to reduce after reaching the highest level. Among the three kinds of VFAs, acetic acid was the main component in early period of all groups. And the proportion of propionic acid gradually increased with the reduction of acetic acid. The total VFAs first reduce to a low level at 98.8 mg/L on day 19 by SS1. Then on day 22, it reduce to a low level at 90.1 mg/L by SS3. In the end of the batch experiments, there were still 1659.1 mg/L total VFAs by SS1. And the CK group still contained 3476.0 mg/L of total VFAs. The results showed that though a high pH can inhibit methanogenesis, there was no harmful effect on VFAs production process.

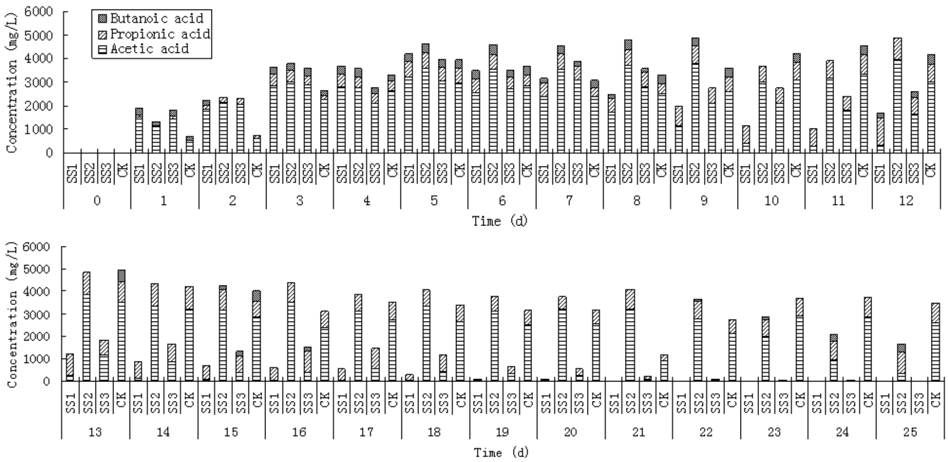

Fig.2 (b) The VFAs concentration during the AD process of PWAS.

Accoding to the changing of VFAs concentration and methane yield during the AD process, both SS1 and SS3 were the suitable seed sludges for PWAS AD. Cao-ultrasonic pretreatment united with suitable seed sludge addition can enhance the total methane production.

3.3. Comparison among different seed sludges for AD of APWAS

To make a better initial environment for AD, the PWAS was stayed for 24 h under nature conditions to acidate. The production of organic acid can counteract alkalinity caused by CaO. After acidification, the pH of APWAS reduced from 9.5 to 7.5 as shown in Table. 1. And as shown in Fig. 3(b), the initial total VFAs was increased to 1814.7 mg/L. Then the APWAS was used as substrate for batch experiments. The cumulative methane production of APWAS by seed sludges is shown in Fig. 3(b). There was almost no lag time for methane production by SS2 and SS3 addition. The SS1 group and control check contained a lag time of around 5 days. Though there was a similar methane production rate by SS2 and SS3 groups in the early period of APWAS AD process, there was a large distance of cumulative methane production in the end of batch experiments. The cumulative methane production by SS2 was only 927.3 mL, which was even 7.5% less than control check of 1002.4 mL. SS2 may consume more organic matter for microorganism growth than other seed sludges or produce other products during AD process. The reason remained to be further researched and proved. The cumulative methane production by SS1 and SS3 was 1249.3 mL and 1383.2 mL , which were 38.0% and 24.6% higher than the control. Comparing with the cumulative methane production of WAS (821.9 mL), the cumulative methane production of APWAS by SS3 was increased by 68.92%. That means after CaO-ultrasonic pretreatment and using optimum seed sludge of SS3 under a comfortable condition, the methane production performance got an obvious improvement.

The VFAs concentration during AD process of APWAS is shown in Fig 3(b). The initial total concentration of VFAs was 1814.7 mg/L. Then the total VFAs reached to the highest in SS1, SS2, SS3 and CK groups at 2541.1 mg/L, 2203.5 mg/L, 2322.6 mg/L and 2405.9 mg/L on day 3, day 1, day 1 and day 5, respectively. After reaching the highest methane yield, the total VFAs began to reduce and propanoic acid became the main component instead of acetic acid gradually. On day 12, the concentration of total VFAs reduced to a low level at 157.2 mg/L by SS3. And there was only 20.7 mg/L total VFAs remained on the next day. SS1 and SS2 groups consumed VFAs to a low level at 103.4 mg/L and 78.4 mg/L on day 15. In the end of batch experiments, there was still 204.2 mg/L total VFAs remained in control check. It is easy to be found that SS3 can use VFAs faster than other seed sludges. And for control group, the conversion of VFAs was lower than that of seed sludges addition groups. This is one of the reasons that a higher methane production had been achieved by adding seed sludges than CK.

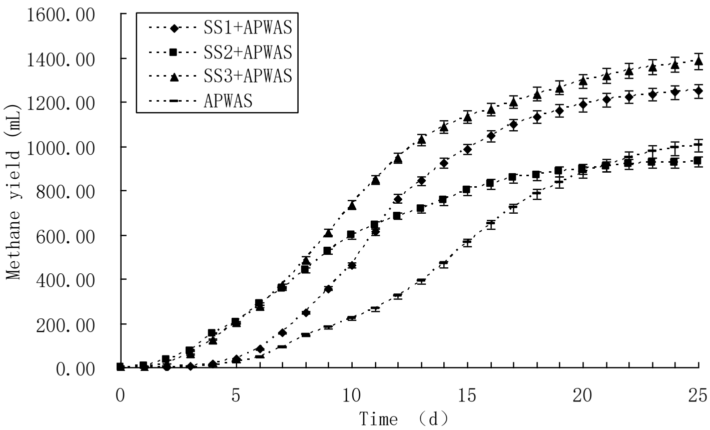

Fig.3 (a) Effect of seed sludges on methane yield of APWAS.

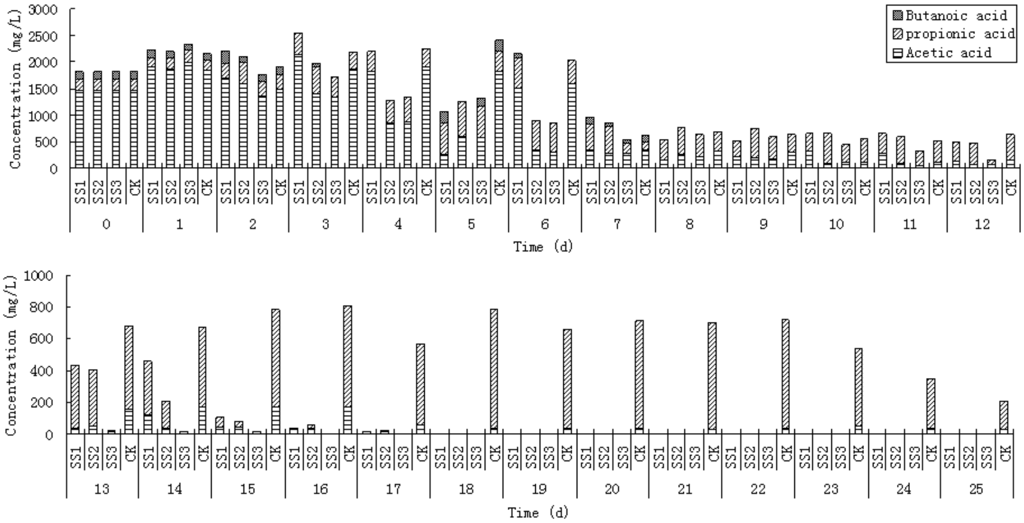

Fig. 3(b) The VFAs concentration during the AD process of APWAS.

In our research, the VFAs reduction match up to the accumulate methane production. The faster VFAs reduced, the higher methane production rate can be achieved. So the accumulation and reduction of VFAs can be an important factor to reflect AD system efficiency during batch experiments.

3.4. Methane yield and VS reduction

The methane yield and VS reduction of all groups after the digestion for 25 d are shown in Fig. 4. The methane yield of raw WAS was found to be 123.3 mL/g·VS with a VS reduction of 16.95%. The methane yield can be increased to 176.6, 185.5 and 181.8 mL/g·VS with VS reduction of 23.04%, 24.75% and 23.1% by adding SS1, SS2 and SS3, respectively. After Cao-ultrasonic pretreatment, the methane yield increased to 187.7 and 189.3 mL/g·VS with a VS reduction of 21.69% and 22.50% by SS1 and SS3. The methane yield reduced to 100 mL/g·VS with VS reduction of 14.55% by SS2. The AD process of control group was serious inhibited and the methane yield was only 19.3 mL/g·VS with VS reduction of 11.01%. After Cao-ultrasound pretreatment, the pH of WAS turn to alkalinity, which strongly inhibited the ability of microorganism to use organic matter to produce meth. SS1 and SS3 can adapt the alkaline condition more quickly than SS2 or microorganism of CK itself by the results. So the VS reduction and biogas production of SS2 and CK groups were lower than the other two groups. The best results were attained with CaO-ultrasonic pretreatment WAS acidated for one day. The methane yield increased to 150.4 mL/g·VS with VS reduction of 23.4% for control group. The optimum seed sludge was found to be SS3, which can increase the methane yield from 123.3 mL/g·VS to 208.3 mL/g·VS and VS reduction from 16.95% to 28.68%. The methane yield and VS reduction by SS3 were 68.92% and 69.20% higher than those of raw WAS group.

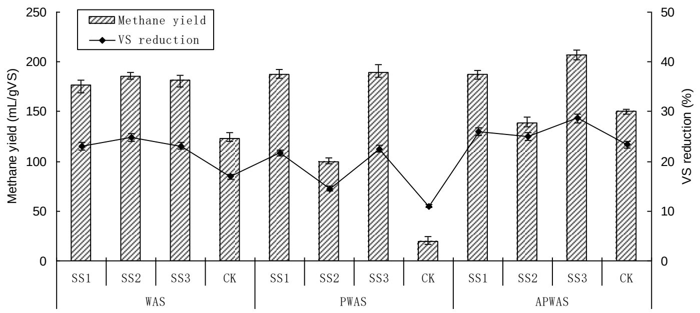

Fig. 4 The methane yield and VS reduction of all groups.

In previous studies of other researchers, ultrasound and alkalized pretreatment have been used to enhance AD performance for WAS, and a certain achievement was obtained. In Perez-Elvira's study, the biogas production from WAS increased by 42% after ultrasonic pretreatment[31]. Neis found that the biogas production was enhanced by 30% after ultrasonic pretreatment[32]. In another research of treated at ultrasonic parameters beyond 2.0 W/mL and 15 min for WAS, more organic substances were released[33]. Alkaline pretreatment by NaOH can increase the methane production from 251.2 mL/L·d to 362.2 mL/L·d[34]. In our research, we first combined CaO-ultrasound pretreatment with seed sludges addition to improve the methane production and VS reduction successfully. A methane increasement of 68.92% higher and VS reduction of 69.20% higher than control had been achieved. The effective of methane yield and VS reduction by combained method (Cao ultrasound pretreatment, acdification and seed sludges addition) was higher than that of using a single method, respectively.

4. Conclusion

In this study, a new method was explored for improving AD performance of WAS. For AD process of WAS, CaO-ultrasound pretreatment would cause a initial alkaline environment, which could inhibit the methanogens to produce methane. This problem can be solved after PWAS laying for a day under natural surroundings. After 25 days' batch experiments, the CaO-ultrasound pretreatment with acdification and optimum seed sludge addition of SS3, which had a stronger ability of degrading WAS and producing methane than SS1 and SS2, can increase 68.92% of methane yield and 69.2% of VS reduction. The effective of WAS AD by SS3 a stronger This study may pave a new way for industrial application of dealing with WAS.

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