Evaluation of Biogas through Chemically Treated Cottonseed Hull in Anaerobic Digestion with/without Cow Dung: An Experimental Study

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Research Article

Keywords: Biogas, chemical pre-treatment, anaerobicdigestion, cow dung, cottonseed hull

Posted Date: December 29th, 2021

DOI: https://doi.org/10.21203/rs.3.rs-1022625/v1

Abstract

Cow dung is generally used as the feedstock material for the anaerobic digestion to produce biogas. A selection of alternate biomass material is needed to reduce the consumption or to eliminate the use of cow dung. Recently, cottonseed hull has been considered as the primary substrate to produce biogas. In this paper, the effect of biogas production on anaerobic co-digestion of cow dung with pre-treated cottonseed hull using different concentrations of sulfuric acid, hydrochloric acid, hydrogen peroxide, and acetic acid is investigated. Sodium hydroxide and calcium hydroxide are used at different concentrations for pre-treatment of cottonseed hull. The enhancement of biogas production from the batch reactors at mesophilic temperature (35 ± 2 ℃) is observed for mono- and co-digestion of cow dung with treated cottonseed hull. Maximum biogas yield is achieved for the treated cottonseed hull at 6% sodium hydroxide during mono digestion and at 6% calcium hydroxide during co-digestion.

Highlights

- Anaerobic co-digestion of cow dung with chemically treated cotton seed hull can be enhanced biogas production.
- 3% acetic acid was best for biodegradability of cottonseed hull and enhancement of biogas yield in acid treated rectors.
- In alkaline treated reactors, 6% calcium hydroxide was best for biodegradability of cottonseed hull and enhancement of biogas yield.
- Optimized biogas production was observed on the anaerobic co-digestion of cow dung and 6% calcium hydroxide treated cotton seed hull at the ratio 75:25.

1 Introduction

In the present world, the increase in the human population is proportional to the increase in energy demand. Energy extracted from fossil fuel is mostly used which creates issues such as scarcity, expensive in price, and causes air pollution mainly affects the environment [1]. Previous studies also suggested using biogas as a clean energy source for power generation, cooking, and heating. Furthermore, anaerobic digestion for biogas production is a potential option that may both augment and reduce the use of non-renewable energy sources like fossil fuels [2]. Cotton production is predicted to climb 4.7 percent globally by 2021/2022, according to a forecast by the United States Department of Agriculture. However, there is a 6.5 percent reduction in comparison to 2019/2020. China is predicted to be the largest producer, with 29.0 million bales of cotton, the highest level in the last six years, while India is estimated to produce roughly 475 kg/hectare, a 1% decrease from the previous year. India was first in cotton-producing countries in 2015, and second in 2019 and 2020, followed by the United States, Brazil, and Pakistan [3]. In India, surplus crop residue is composted, which takes up a lot of space, and then burned. Crop residues produce greenhouse gases when burned, contributing to increased air pollution [4]. Agricultural crop residues are used to produce biogas via anaerobic digestion to address this problem[5].

Agricultural crop residue such as cotton waste was generated around 11.4 Mt in India [6]. Appropriate technologies are employed for the production of clean energy to properly utilize waste and meet current energy requirements by reducing pollution. For the maximum degradation of organic waste, anaerobic digestion is the best technique opted for the generation of energy (Methane) in a more economical and environment-friendly manner [7, 8].

The alternative new renewable energy extraction can be done from biomass which could be a better solution. Biomass is the best alternative source that produces renewable energy and replaces fossil fuel products. Lignocellulose biomass comprises a large fraction of cellulose (40-50%), a smaller fraction of hemicellulose (20-40%), a very low fraction of lignin, and a small quantity of other alternatives. Cellulose is a glucose polymer with an average molecular weight of around 1,00,000, whereas hemicellulose is a heterogeneously branched polysaccharide with an average molecular weight of less than 30,000. Lignin has complex and cross-linked structures with high molecular weight. Lignocellulose biomass is difficult to degrade, naturally. For better degradation, a proper treatment method is followed [1]. The anaerobic biodegradability has been enhanced with the aid of treatment methodologies such as mechanical, thermal, chemical, ultrasonic, biological and microwave approaches through the anaerobic digestion process [9, 10]. Biogas is produced through an anaerobic digestion process on various biomass materials such as wheat straw [11], pearl millet straw [12], corn stover [13, 14], rice straw[15], yard waste, and leaves [16], and also with other wastes such as food waste with cow dung [17], food waste with fresh septic tank sludge [18], etc.

Anaerobic co-digestion is a promising technique to enhance the digestion performance and biodegradability of lignocellulose biomass [19]. Among all the pre-treatment processes of biomass, the chemical pre-treatment process has been reported to offer maximum methane yield through the anaerobic digestion process with the effective breaking of chemical bonds among cellulose, hemicellulose, and lignin through microorganisms [20]. In the chemical treatment methods, the following chemicals are commonly used such as H_2SO_4, HCl, H_2O_2, CH_3COOH, NaOH and $Ca(OH)_2$ for the biodegradability of lignocellulosic biomass materials through an anaerobic digestion process [21]. NaOH and $Ca(OH)_2$ are used as pre-treatment chemicals on giant reed materials for the enhancement of biogas production through anaerobic digestion [22]. Three plant species such as hay, straw, and bracken were pre-treated using $Ca(OH)_2$, maleic acid, and ammonium carbonate, and subsequent anaerobic digestion was carried out under mesophilic temperature for 40 days in a batch digester [23]. The highest methane production was achieved by pre-treating corn stalk with $Ca(OH)_2$ through an anaerobic co-digestion process with levulinic acid wastewater [24]. Pre-treated grass silage with NaOH increases biodegradability as well as biogas production through anaerobic digestion [25]. Sunflower stalks are pre-treated using various chemicals like NaOH, H_2O_2, $Ca(OH)_2$, HCl, and $FeCl_3$ to enhance methane production through anaerobic digestion [26]. Anaerobic co-digestion of cattle manure and corn stover was compared with that of ammonia solution and NaOH pre-treated samples which also results in higher methane yield under mesophilic conditions [27]. Pre-treatment of corn stalk using H_2SO_4 and H_2O_2 yields higher biogas production on the anaerobic co-digestion with swine manure [28]. Anaerobic co-digestion of cocoa pod husk with swine manure yields maximum biogas production where cocoa pod husk was pre-treated by using H_2SO_4 and H_2O_2 [29]. It was reported that methane production is directly proportional to the enzymatic degradation of the lignocellulose biomass, whereas methane potential is inversely related to the lignin content present in the lignocellulose biomass [16]. Remarkable biogas production was achieved on pre-treating NaOH with corn stover [13, 14, 30] and Asparagus stover [31] in the anaerobic digestion process. Pre-treatment of rice straw with H_2O_2 used in biodegradation enhances biogas production [32]. Co-digestion of wheat straw with chemicals like KOH, $Ca(OH)_2$ results in higher biodegradability with the increase in biogas yield [33]. The main advantage of chemical pre-treatment on lignocellulosic biomass is to improve the effective biodegradability and upsurge the bioenergy production [21].

Naturally, agricultural biomass waste like cotton stalks was generally burnt and lead to air pollution. Cotton stalks can be effectively converted into useful energy through anaerobic digestion by various treated chemicals such as KOH, NaOH, $Ca(OH)_2$, alkali Hydrogen per-oxide (AHP), H_2SO_4, H_3PO_4, and steam explosion method [34]. Wheat straw was pre-treated with H_2O_2 and also co-digested with cattle manure for the enhancement of biodegradability and also methane yield [35]. Cotton wastes such as cotton stalks, cottonseed hull (CSH), and cotton oil cake can be effectively converted for biogas production in the presence of basal medium through anaerobic digestion [36]. For sustainable environments, energy extraction has been done by performing effective pre-treatment studies to improve energy production [37].

Biochemical methane potential (BMP) test was conducted in an anaerobic co-digestion of cow dung (CD) with cottonseed hull (CSH) at different ratios (CD: CSH) i.e., 0:100, 50:50, 25:75, 75:25 and 100:0. Maximum biogas yield was obtained from the anaerobic co-digestion at the ratio of 75:25 among other proportions. Mono-digestion of CSH results in less biogas yield compared to the co-digestion of CD and CSH at the ratio of 75:25. From the previous studies, it is found that the optimum biogas yield is obtained from the anaerobic co-digestion of CD with CSH at the ratio of 75:25, and verified both experimentally [38] and using kinetic studies [39].

From the above literature review, it is found that the importance of chemical treatments on different biomass and the triggering of biogas generation through anaerobic digestion. Besides, the chemicals used for treatment break the bonding between the complex biomass structures in generating the anaerobes at a rapid rate. The CSH has the potential to generate biogas during mono- and co-digestion with CD. Very few literatures have reported on the biogas generation from anaerobic mono- and co-digestion of CD with chemically treated CSH.

The performance of the digester with CD and CSH in the ratios of 75:25 and 0:100 with pre-treatment with various chemicals such as sulfuric acid, hydrochloric acid, hydrogen peroxide, acetic acid, sodium hydroxide, and calcium hydroxide at different concentrations for the enhancement of biogas yield has been investigated in this paper. The main objective of this paper is to find out the maximum biogas yield among all chemically pre-treated CSH and the best concentration in each acid and alkaline. The best-treated chemical for CSH for the complete replacement of CD in the existing gobar gas plant is also suggested.

2 Materials And Methods

2.1 Feedstock material

Feedstock material such as CD and CSH was collected from a local village in Karur city, Tamilnadu, India (Latitude - 10.69 °N, Longitude - 78.42 ° E). Before the usage of CSH material, air drying and shredding was done to make it into small particles with a spice pulverizer (Vikrem commercial heavy-duty pulverizer, India. Vik-9A model). Then, the material was sieved for 10 minutes through a particle sieve analyzer (make: Versatile equipment Private Limited, India) which ranges from 0.5 mm to 0.75 mm. Fresh CD was collected from a small household located in the same village. The inoculum was taken from the batch anaerobic digester where CD was used as the main feedstock material at the same location. Prior to the BMP test, the collected feedstock material was kept at a temperature of 4 °C. The effluent was maintained at a temperature of 37 °C for seven days and dilute it with water[38]. The physical and chemical characteristics of the feedstock material are listed in Tables 1 and 2. CSH substrates consist of 32 ± 2.1% cellulose, 18 ± 0.9% hemicellulose and 20 ± 1.7% lignin were founded using sequential extraction and weighing method [12].

Table 1
Physical characteristics of the feedstock materials used

Samples used	Moisture present (%)	Total solid (TS) (%)	Volatile solid (VS) (%)	Fixed solid (FS) (%)	Reported
Cow dung	86 ± 1	14 ± 1	68 ± 4	32 ± 4	[38]
Cotton seed Hull	11 ± 0.1	90 ± 0.1	90 ± 0.1	10 ± 0.1	
Inoculum	92 ± 0.1	9 ± 0.1	62 ± 0.2	38 ± 0.2	
± denotes the standard deviation values for triplicate observation					

Table 2
Chemical characteristics of the feedstock materials used

Feedstock materials	Total C (%)	Total H (%)	Total N (%)	Total S (%)	C/N ratio	Reported
Cow dung	39.64 ± 1.25	1.85 ± 0.01	1.21 ± 0.02	2.24 ± 0.03	32.76	[38]
Cotton seed Hull	38.31 ± 0.3	2.24 ± 0.25	1.05 ± 0.01	1.47 ± 0.01	36.38	
Inoculum	30.36 ± 1.5	0.88 ± 0.02	1.53 ± 0.01	1.37 ± 0.02	19.91	

2.2 Pre-treatment

A 500 ml-conical flask is filled with 300 ml of distilled water and 100 g of CSH sample. Based on the weight/weight basis, acids and alkaline are added to the conical flask. From the previous study, the concentration of each acid and alkaline was chosen for the pre-treatment of CSH [21]. Acids are mixed with distilled water of proportions 1%, 2%, and 3%, whereas alkaline is mixed with distilled water at proportions 4%, 6%, and 8%, respectively. The conical flask was kept inside the incubator at a temperature of 25 ± 2 °C for seven days. After the incubation period, all the samples were kept in the electric oven for two days at a temperature of 80 °C [21]. Then, the dried samples were taken out without washing and again maintained at a temperature of 4 °C in the refrigerator until the start of the BMP test.

2.3 Anaerobic digestion experiments

Biogas production was determined through anaerobic digestion in a batch process with two sets of experiments. Each pre-treated CSH was used as the feedstock material, where untreated CSH was used as the control. In the first set of experiments, mono-digestion of acid and alkaline treated CSH was carried out at different concentrations. In the second set of experiments, co-digestion of CD and treated CSH at the ratio of 75:25 was carried out with the corresponding pre-treated CSH using early mentioned chemicals. All the experiments were conducted in a 0.5 L conical flask of which 0.3 L was taken as working volume. Each conical flask was filled with 0.3 L of inoculum, and feedstock material was added based on a final concentration of 1.5 gram volatile solid (VS) per liter. The main reason for using inoculum from the anaerobic digestion process, which stimulates the methanogenic activity inside the batch reactors. All the conical flasks were stirred well and incubated under the mesophilic condition at a temperature of 35 ± 2 °C for the digestion period of 45 days [40]. All the flasks were flushed with nitrogen, then closed by using a septa cap and kept in the incubator to create favorable conditions for generating anaerobes. Each flask was stirred manually for one minute before measuring the biogas yield. All the BMP tests with each sample were performed in duplicate.

2.4 Analysis and calculations

The volume of biogas produced from the digester was determined by using the water displacement method. The volume of biogas was finally calculated by subtracting it from the volume of biogas collected in the inoculum. Physical characteristics like total solid, volatile solid, fixed solid, and moisture content was found using standard methods (APHA 1995). Chemical characteristics like Carbon, Nitrogen was calculated using Vario EL III- Germany Elementar analyzer. C/N ratio was calculated as the ratio of total Carbon to total Nitrogen. pH values were determined using a pH meter (pH 827 modules, Metrohm India Ltd.). The composition of cellulose, hemicellulose, and lignin presented in CSH substrate was measured using sequential extraction and weighing method on a dry weight basis [12]. CSH samples were mixed in 75 ml of water and boiled for one hour. Residing hot water was removed after an hour by adding fresh water and boil for one more hour. Then, the CSH samples were cleaned with cold water, and the sample was dried in an oven for 15 hours at a temperature of 60 °C and weighed. The dried CSH sample was again mixed with 30 ml of water, 2 ml of acetic acid (i.e., 10%), and 0.6 g of sodium chlorite, and heated at 75 °C for 1 hour to calculate the lignin content. Similar steps were followed one more time and heated for 2 hours. Distilled water, acetone, and ether were used to wash the CSH samples 5 times, 2 times, and 1 time, respectively. After that, the same CSH sample was dried at a temperature of 105 °C for a one-and-half hours and weighed the sample. Hemicellulose content was calculated by appending 24% KOH, i.e., 20 ml and left in air at a temperature of 20 °C. Then the sample was soaked 5 times with water and one time into 5% acetic acid. Again, the same sample was bathed one more time with water, acetone, and ether. After bathing, the CSH sample was maintained at a temperature of 105 °C for a one-and-half hours and weighed the same. The left-over weight of the CSH sample was considered as the weight of the cellulose content.

3 Results And Discussion

3.1 Effect of pre-treatment on pH

Before anaerobic digestion, the initial pH values of all the reactors ranged from 7.21 to 7.44 which is mainly responsible for microbial growth and gives a positive impact on biogas production [42]. Since micro-organisms are more sensitive to pH, methanogenic bacteria perform maximum reproduction over the pH range of 6.6 to 7.8 for biogas generation. After anaerobic digestion, the final pH values were from 7.08 to 7.48 for all the digesters at the end of 45 days. The pH values at the end of the anaerobic digestion process were stable when it lies within 7.5 [43]. pH values for the pre-treated acid and alkaline at various concentrations before and after the anaerobic digestion process are shown in Fig. 1 and Fig. 2.

3.2 Effect of chemical pre-treatment on mono-digestion

3.2.1 Effect of H_2SO_4 treated CSH on biogas generation with mono-digestion

Maximum biogas yield of 120 ml/g VS was observed at 3% pre-treated H_2SO_4 with CSH. There was no significant difference in biogas yield between 1% and 2% H_2SO_4 treated CSH as the final yield was 106 ml/g VS. Biogas yield was 3 to 4-fold higher than untreated CSH. The difference in biogas yield between treated CSH and untreated CSH was observed as 73 ml/g VS,73 ml/g VS, and 87 ml/g VS at the concentration of 1%, 2%, and 3% H_2SO_4. Results produced are consistent in the previous study as the cumulative methane yield lies nearer range 176 ml/g VS for the 2% H_2SO_4 treated corn straw [21]. The maximum biogas difference was observed as 34 ml/g VS between 3% pre-treated H_2SO_4 and co-digestion of CD with untreated CSH at 75:25 (CD: CSH). A higher concentration of pre-treated CSH with H_2SO_4 leads to maximum bioconversion which could improve biogas production.

3.2.2 Effect of HCl treated CSH on biogas generation with mono-digestion

Different concentration of HCl pre-treated CSH offers different biogas yields as compared to untreated CSH. A higher biogas yield of 116 ml/g VS was observed at 1% HCl pre-treated CSH. 73 ml/g VS and 87 ml/g VS of biogas yield were observed for 2% and 3% HCl treated CSH. Results produced are persistent in the previous study as the cumulative methane yield lies nearer range 163 ml/g VS for the 2% HCl treated of corn straw [21]. The difference in biogas yield between treated CSH at the concentration of 1%, 2%, and 3% HCl and raw CSH was founded as 83,40 and 54 ml/g VS. 30 ml/g VS was observed as the difference in biogas yield at the 1% HCl treated CSH and untreated CSH at the ratio of 75:25(CD: CSH). Cumulative biogas production from the treated HCl on CSH was 2 to 4-fold greater than untreated CSH. A lower concentration of HCl on pre-treated CSH also improves biodegradability.

3.2.3 Effect of H_2O_2 treated CSH on biogas generation with mono-digestion

H_2O_2 pre-treated CSH substrate offered cumulative biogas production of 91 ml/g VS, 183 ml/g VS, and 171 ml/g VS respectively for 1%, 2%, and 3% under hydraulic retention time (HRT) of 45 days. The cumulative methane yield reported in the previous study for the 3% H_2O_2 pre-treated corn straw was observed as 217 ml/g VS [21]. It was observed that the difference of biogas yield between H_2O_2 treated at the concentration of 1%, 2%, and 3% and raw CSH as 58,150, and 138 ml/g VS. The maximum difference in biogas yield of 97 ml/g VS was observed between the 2% H_2O_2 treated CSH and CD with untreated CSH at the ratio of 75:25(CD: CSH). More than 3 to 6 times higher biogas yield was observed over untreated CSH. Increasing the loading of pre-treated H_2O_2 with CSH upsurge the bioconversion during anaerobic digestion.

3.2.4 Effect of CH_3COOH treated CSH on biogas generation with mono-digestion

A total of 295 ml/g VS of biogas yield was observed at 3% CH_3COOH, whereas 202 ml/g VS and 158 ml/g VS were observed at 1% and 2%, respectively. Previous results obtained cumulative methane yield as 145 ml/g VS for the 4% CH_3COOH treated on corn straw [21]. Biogas yield obtained at the difference between treated 1%,2%, and 3% CH_3COOH and raw CSH was 169,125 and 262 ml/g VS. Remarkable difference in biogas yield around 209 ml/g VS was observed between 3% CH_3COOH and untreated CSH at the ratio of 75:25 (CD: CSH). The maximum difference of 102 ml/g VS in yield was also observed between 3% CH_3COOH and CD alone. More than 5 to 9-fold higher biogas yield was observed with untreated CSH. The higher concentration of CH_3COOH was directly related to the bioconversion of CSH.

3.2.5 Effect of NaOH treated CSH on biogas generation with mono-digestion

Cumulative biogas yield of 212, 430, and 155 ml/g VS was observed respectively at 4%, 6%, and 8% pre-treated NaOH with CSH. It was obvious from data that 5 to 13-fold greater biogas yield was obtained raw CSH during the anaerobic digestion process. 8% NaOH treated corn straw produced a cumulative methane yield of 164 ml/g VS [21]. The difference in biogas yield between the NaOH treated CSH at the concentration of 4%, 6%, and 8% and untreated CSH was observed as 179,397, and 122 ml/g VS. The maximum difference in biogas yield was observed as 344, 237 ml/g VS with the 6% treated NaOH between untreated CSH at the ratio of 75:25 (CD: CSH) and CD. Medium concentration (6%) of NaOH offered maximum biogas generation as compared to too low and too high concentrations.

3.2.6 Effect of $Ca(OH)_2$ treated CSH on biogas generation with mono-digestion

Maximum biogas of 279 ml/g VS was produced for the 6% $Ca(OH)_2$ pre-treated CSH, whereas 100 ml/g VS and 150 ml/g VS were observed for 4% and 8% $Ca(OH)_2$ pre-treated CSH, respectively. This was 3 to 8-fold greater biogas yield compared to the mono-digestion of raw CSH. 8% $Ca(OH)_2$ pre-treated corn straw produced a cumulative methane yield of 207 ml/g VS [21]. 4%,6%, and 8% $Ca(OH)_2$ treated CSH produces a difference in biogas yield of 67, 246, and 117 ml/g VS with untreated CSH. The maximum difference in biogas yield was observed as 193, 86 ml/g VS with the 6% treated $Ca(OH)_2$ between untreated CSH at the ratio of 75:25 (CD: CSH) and CD. Medium concentration (6%) of $Ca(OH)_2$ also offered maximum biogas yield compared to both too low and too high concentration cases.

3.2.7 Acidic Vs alkaline treatment effect on mono-digestion

With mono-digestion of pre-treated CSH, the obtained biogas yield was found to follow the order, 6% NaOH > 3% CH_3COOH > 6% $Ca(OH)_2$,> 2% H_2O_2> 3% H_2SO_4> 1% HCl where the obtained cumulative biogas yields respectively were 430 ± 11 ml/g VS, 295 ± 9 ml/g VS, 279 ± 14 ml/g VS, 183 ± 11 ml/g VS, 120 ± 9 ml/g VS, and 116 ± 9 ml/g VS. A previous study also reports that the maximum concentration of alkaline added to lignocellulose material does not produce any effect on chemical links between lignin and hemicellulose, which only reduces the lignin solubilization [44]. The difference in maximum biogas yield between acid and alkaline treated CSH during mono-digestion is around 135 ml/g VS. Pre-treatment on lignocellulose biomass was reported to improve biogas production by accelerating the growth of the micro-organisms at a rapid reaction rate through an anaerobic digestion process [45]. Effective alkaline treatment had a high potential to increase the biogas yield since it degrades the biomass completely by reducing the lignin content during anaerobic digestion [46, 47].

3.3 Effect of chemical pre-treatment on co-digestion

3.3.1 Effect of H_2SO_4 treated CSH on biogas generation with co-digestion of CD

75% of Cow dung was co-digested with 25% pre-treated H_2SO_4 through anaerobic digestion process yields cumulative biogas of 197, 106, and 126 ml/g VS for the concentration of 1%, 2%, and 3% H_2SO_4, respectively. This was 3 to 6-fold greater biogas yield over raw CSH, whereas 1 to 2-fold greater yield was obtained over co-digestion of CD with raw CSH at the same ratio. 164,73, and 93 ml/g VS was observed as the difference in biogas yield between 1%, 2%, 3% H_2SO_4 treated and untreated CSH. The maximum biogas difference of 111, 4 ml/g VS was observed with the 1% H_2SO_4 treated CSH between untreated CSH at the ratio of 75:25 (CD: CSH) and CD. A lower concentration of pre-treated H_2SO_4 itself enhances the maximal biodegradability of CSH.

3.3.2 Effect of HCl treated CSH on biogas generation with co-digestion of CD

200, 170, and 173 ml/g VS of biogas were generated during the co-digestion (75:25) of CD with HCl pre-treated for the concentration of 1%,2%, and 3%, respectively. This was a 5 to 6-fold increment achieved over mono-digestion of untreated CSH and also 2-fold greater than co-digestion of CD with untreated CSH at the same ratio. 167, 137, and 140 ml/g VS was obtained as the biogas yield difference between 1%, 2%, 3% HCl treated and raw CSH. 1% HCl treated CSH resulted in maximum biogas difference as 114, 7 ml/g VS between untreated CSH at the ratio of 75:25 (CD: CSH) and CD. Results revealed that a lower concentration of HCl produced maximum biogas generation.

3.3.3 Effect of H_2O_2 treated CSH on biogas generation with co-digestion of CD

The co-digestion (75:25) of CD with pre-treated H_2O_2 on CSH yielded cumulative biogas of 139,195 and 145 ml/g VS for 1%,2%, and 3% concentrations, respectively, which was 4 to 6-fold higher than mono-digestion of untreated CSH. Similarly, this yield was 2-fold greater than the one obtained with co-digestion of CD with untreated CSH at the same ratio. The biogas yield difference was observed as 106,162, 112 ml/g VS between the 1%, 2%, 3% H_2O_2 treated and untreated CSH. A remarkable biogas yield difference was noticed as 109, 2 ml/g VS between untreated CSH at the ratio of 75:25 (CD: CSH) and CD at the 2% H_2O_2 treated CSH. The medium concentration of treated H_2O_2 on CSH leads to maximum biogas generation.

3.3.4 Effect of CH_3COOH treated CSH on biogas generation with co-digestion of CD

277, 171, and 345 ml/g VS of biogas were produced during the co-digestion (75:25) of CD with CH_3COOH pre-treated CSH for the concentration of 1%,2%, and 3% which was 5 to 10-fold greater than the yield obtained with mono-digestion of untreated CSH, and 2 to 4-fold greater than the yield obtained with co-digestion of CD with untreated CSH for the same ratio. The biogas yield difference between 1%,2%, 3% CH_3COOH treated and untreated CSH was observed as 244,138, and 312 ml/g VS. At 3% CH_3COOH treated CSH, the maximum biogas yield difference was observed as 259, 152 ml/g VS between the untreated CSH at the ratio of 75:25 (CD: CSH) and CD. A higher concentration of CH_3COOH treated CSH leads to maximum biogas generation.

3.3.5 Effect of NaOH treated CSH on biogas generation with co-digestion of CD

During the anaerobic co-digestion (75:25) of CD with pre-treated NaOH on CSH yielded cumulative biogas of 166, 395, and 104 ml/g VS for the concentrations of 4%, 6%, and 8%, respectively which was 3 to 12-fold higher than mono-digestion of untreated CSH, and 1 to 5-fold greater than the yield achieved during the co-digestion of CD with untreated CSH at the same ratio. The biogas yield difference was observed as 133,362, 71 ml/g VS between the 4%, 6%, 8% NaOH treated and untreated CSH. A remarkable biogas yield difference was noticed as 309, 202 ml/g VS between untreated CSH at the ratio of 75:25 (CD: CSH) and CD at the 6% NaOH treated CSH. The medium concentration of treated CSH with NaOH yielded maximum biogas generation.

3.3.6 Effect of $Ca(OH)_2$ treated CSH on biogas generation with co-digestion of CD

151, 489, and 431 ml/g VS of biogas were generated during anaerobic co-digestion (75:25) of CD with treated $Ca(OH)_2$ for the concentrations of 4%, 6%, and 8%, respectively. It was observed that 5 to 15-fold greater yield was achieved over the mono-digestion of untreated CSH, and a 2 to 6-fold higher yield was achieved over the co-digestion of CD with untreated CSH at the same ratio. The biogas yield difference between 4%,6%, 8% $Ca(OH)_2$ treated and untreated CSH was observed as 118,456, and 398 ml/g VS. At 3% $Ca(OH)_2$ treated CSH, the maximum biogas yield difference was observed as 403, 296 ml/g VS between the untreated CSH at the ratio of 75:25 (CD: CSH) and CD. The medium concentration of treated CSH with $Ca(OH)_2$ yielded maximum biogas generation.

3.3.7 Acidic Vs alkaline treatment effect on co-digestion

The co-digestion of CD with pre-treated CSH at the ratio of 75:25, resulted in the cumulative biogas production of 489 ± 12 ml/g VS, 395 ± 16 ml/g VS, 345 ± 9 ml/g VS, 200 ± 9 ml/g VS, 197 ± 8 ml/g VS, 195 ± 9 ml/g VS (ranked in the order of 6% $Ca(OH)_2$ > 6% NaOH > 3% CH_3COOH > 1% HCl > 1% H_2SO_4 > 2% H_2O_2). The maximum difference in biogas yield of 144 ml/g VS is observed during co-digestion of CD with acid and alkaline treated CSH (75:25). The cumulative biogas production on both untreated CD and CSH at different ratios were observed to be 193 ± 6 ml/g VS (100:0), 33 ± 2 ml/g VS (0:100), and 86 ± 7 ml/g VS (75:25) after 45 days[38].

The cumulative biogas yield from the treated CSH with the chemical concentration of 1% CH_3COOH, 3% CH_3COOH, 4% NaOH, 6 % NaOH, 6 % $Ca(OH)_2$ produces the maximum results as compared with the CD alone. All other untreated CD with chemically pre-treated CSH produces extraordinary cumulative biogas production when compared to the untreated CD with CSH for the ratio of 75:25. Untreated CSH offered only very low cumulative biogas production as compared with both chemically treated CSH and untreated CD. In the 100% pre-treated CSH, the maximum biogas generation was seen through the mono-digestion process, especially in 6% pre-treated NaOH, whereas in the co-digestion of CD with pre-treated CSH, it was observed for the 6% of $Ca(OH)_2$. Table 3 describes the cumulative biogas production of untreated CD and CSH at the ratio of 100:0, 0:100, 75:25 along with the treated CSH, 0:100, and 75:25.

Table 3
Cumulative biogas production for all the reactors

Blend ratio	Chemicals treated	Cumulative Biogas yield (ml/g VS)	Reported
CD (100:0)	0%	193 ± 16	[38]
Untreated CSH (0:100)	0%	33 ± 2	
Untreated CD with CSH (75:25)	0%	86 ± 7	
Treated CSH (0:100)	1 % H_2SO_4	106 ± 11	Present study
Treated CSH (0:100)	2 % H_2SO_4	106 ± 9	
Treated CSH (0:100)	3 % H_2SO_4	120 ± 9	
Treated CSH (0:100)	1 % HCl	116 ± 9	
Treated CSH (0:100)	2 % HCl	73 ± 13	
Treated CSH (0:100)	3 % HCl	87 ± 11	
Treated CSH (0:100)	1 % H_2O_2	91 ± 12	
Treated CSH (0:100)	2 % H_2O_2	183± 11	
Treated CSH (0:100)	3 % H_2O_2	171 ± 10	
Treated CSH (0:100)	1 % CH_3COOH	202± 9	
Treated CSH (0:100)	2 % CH_3COOH	158 ± 9	
Treated CSH (0:100)	3 % CH_3COOH	295 ± 9	
Treated CSH (0:100)	4 % NaOH	212± 11	
Treated CSH (0:100)	6 % NaOH	430 ± 11	
Treated CSH (0:100)	8 % NaOH	155 ± 13	
Treated CSH (0:100)	4 % $Ca(OH)_2$	100 ± 9	
Treated CSH (0:100)	6 % $Ca(OH)_2$	279 ± 14	
Treated CSH (0:100)	8 % $Ca(OH)_2$	150 ± 8	
CD with treated CSH (75:25)	1 % H_2SO_4	197 ± 8	
CD with treated CSH (75:25)	2 % H_2SO_4	106 ± 9	

Blend ratio	Chemicals treated	Cumulative Biogas yield (ml/g VS)	Reported
CD with treated CSH (75:25)	3 % H_2SO_4	126 ± 9	
CD with treated CSH (75:25)	1 % HCl	200 ± 9	
CD with treated CSH (75:25)	2 % HCl	170± 8	
CD with treated CSH (75:25)	3 % HCl	173 ± 9	
CD with treated CSH (75:25)	1 % H_2O_2	139± 8	
CD with treated CSH (75:25)	2 % H_2O_2	195 ± 9	
CD with treated CSH (75:25)	3 % H_2O_2	145 ± 7	
CD with treated CSH (75:25)	1 % CH_3COOH	277 ± 14	
CD with treated CSH (75:25)	2 % CH_3COOH	171± 9	
CD with treated CSH (75:25)	3 % CH_3COOH	345 ± 9	
CD with treated CSH (75:25)	4 % NaOH	166 ± 8	
CD with treated CSH (75:25)	6 % NaOH	395 ± 16	
CD with treated CSH (75:25)	8 % NaOH	104 ± 9	
CD with treated CSH (75:25)	4 % $Ca(OH)_2$	151 ± 11	
CD with treated CSH (75:25)	6 % $Ca(OH)_2$	489± 12	
CD with treated CSH (75:25)	8 % $Ca(OH)_2$	431 ± 12	

Remarkably, biogas production was enhanced through co-digestion of CD with pre-treated CSH when compared with mono-digestion of CD and CSH. Previous studies were also reported that maximum biogas production achieved through the co-digestion process which indicates the greater solubility and the biodegradability of hemicellulose, cellulose, and lignin. This in turn shows the importance of anaerobic bacteria [35].

3.4 Comparison of acid and alkaline pre-treated CSH on biogas production

The biodegradability of lignocellulosic material mainly depends on the degradation of cellulose, hemicellulose, and lignin. Lignin plays a major role in biodegradation in anaerobic conditions [48]. For higher biodegradability, lignin content present in the lignocellulosic material must be lower. If lignin content is more in the lignocellulosic material, the solubilization rate will be low, and special chemical treatment will create a positive effect on biodegradability [16, 23, 42]. Alkali treatments are best in maximum solubilization of hemicellulose and lignin since they broke the chemical bonded linkages on ester bonds between them presented on the cell wall [10, 49]. Maximum methane yield was observed when there was greater enzymatic degradation, and contents of cellulose and hemicellulose with less lignin in the biomass [16, 45, 50]. Saturated lignocellulose presented in biomass samples sinks the biogas production, whereas alkaline pre-treatment upsurge the biogas production through the increase of microbial fermentative organism available in the soluble organic substance [35, 45, 50, 51].

In the acid pre-treatment of CSH, the maximum biogas production was observed in the 3% CH_3COOH whereas, in the alkaline pre-treatment of CSH, maximum biogas production was observed in the 6% NaOH from the mono-digestion of CSH. For the co-digestion of 75% CD with 25% of pre-treated CSH, the maximum biogas was produced in the 3% CH_3COOH in the acid pre-treatment and 6% $Ca(OH)_2$ in the alkaline pre-treatment. During mono digestion of treated CSH, 13-fold improvement was observed at 6% NaOH over untreated CSH, whereas in co-digestion of CD with treated CSH, it is achieved 15-fold improvement over untreated CSH at 6% $Ca(OH)_2$. This result was consistent with a previous study where biogas production was up to 162.2% [44] and also CD co-digested with 6% of $Ca(OH)_2$ pre-treated CSH at the ratio 75:25 increased by 153.4%. This implies that co-digestion improves the biodegradability of CSH when compared with mono-digestion of pre-treated CSH. The major advantage of alkaline pre-treatment on lignocellulose biomass was to broke the ester bonds and disintegrate cellulose, hemicellulose, and lignin content to reduce the digestion time and also to improve the biogas production. Previous researchers also reported that 6% of alkaline pre-treatment on lignocellulose biomass showed the best results of biogas production among other concentrations [31, 52, 53]. Too low or too high concentration of alkaline pre-treatment may affect the anaerobic degradation of biomass substrate which will result in lower biogas yield. Acid pre-treatment on lignocellulose biomass substrate resulted in maximum biogas production at a lower concentration. Since it degrades the structure of the substrate and creates the composition favorable to generate micro anaerobes. At higher concentrations, lower biogas yield was obtained as it loses the substrates of dry matter rapidly which inhibits the growth of micro-organisms [52]. Results shown in the current study were consistent with previous studies and it can be concluded that alkaline pre-treatment will produce maximum biogas yield as compared to acid pre-treatment on various biomass substrates [53–55]. For the HRT of 45 days, with different chemically pre-treated CSH, the corresponding biogas production is shown in Fig. 3 and Fig. 4.

4 Conclusions

The variation of biogas production on anaerobic digestion of chemically treated mono-digestion of cottonseed hull and co-digestion of cow dung with treated cottonseed hull was studied under batch digester conducted for 45 days. Remarkable biogas production was achieved in the co-digestion of cow dung with treated cottonseed hull at the ratio of 75:25 when compared with the treated mono-digestion of cottonseed hull. Among all digesters, it was observed that 153.4% of biogas production in the co-digestion of 75% cottonseed hull treated with 6% calcium hydroxide as compared to untreated 25% cow dung. It is suggested that pre-treated 75% cottonseed hull at 6% calcium hydroxide will replace 75% of cow dung consumption in the existing biogas plant with increased biogas production. Among various acid pre-treatments, 3% acetic acid was best for biodegradability of cottonseed hull and enhancement of biogas yield and in alkaline pre-treatments, 6% calcium hydroxide was best. Chemical pre-treatment on cottonseed hull enhances biogas production with reduced duration of digestion time compared with untreated cottonseed hull.

Declarations

competing Interests The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgment

We would like to record our sincere thanks to Dr.R. Gandhimathi, Associate Professor, and Professor G. Swaminathan, Head, Department of Civil Engineering, National Institute of Technology, Tiruchirappalli for providing opportunities to utilize the facilities in Environmental Laboratory.

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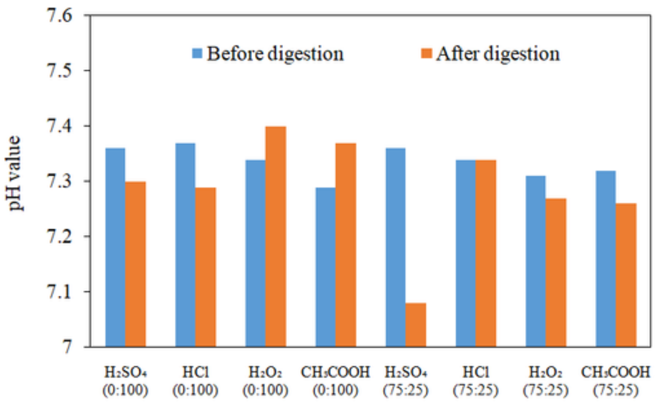
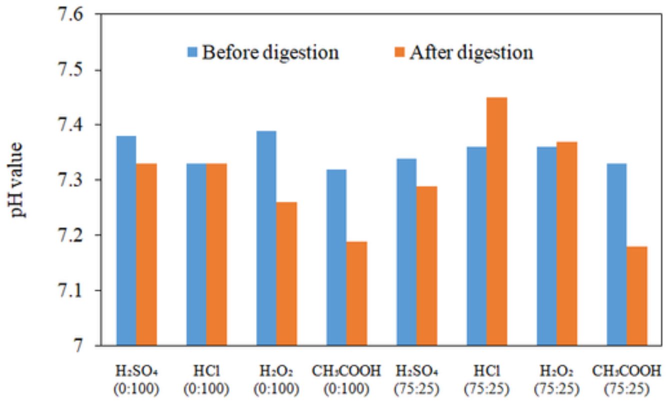
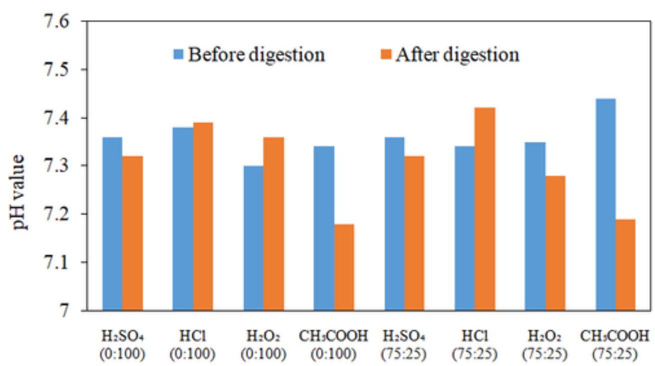
Figures



(a) 1% concentration



(b) 2% concentration



(c) 3% concentration

Figure 1

pH values of acid treated reactors

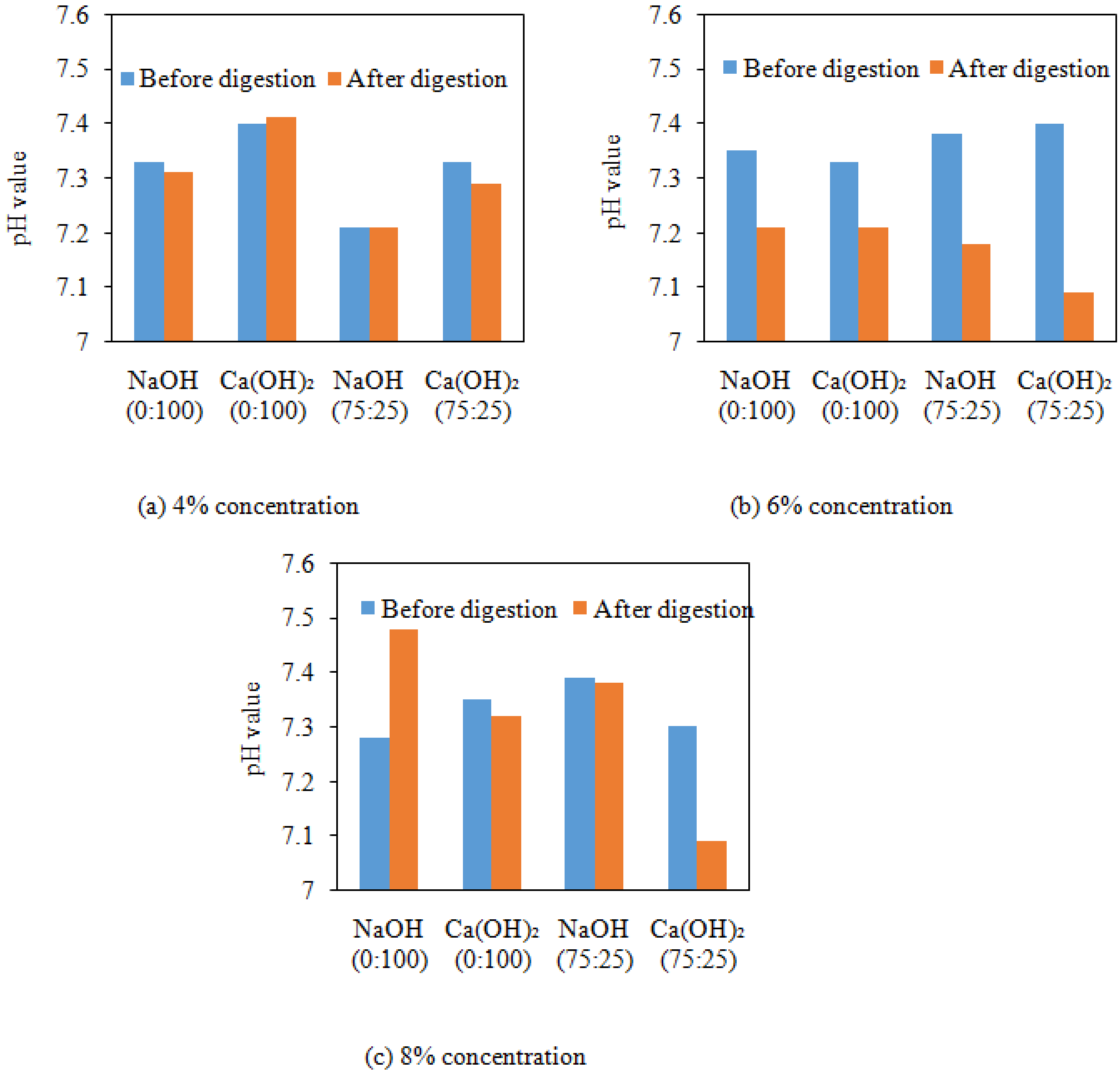


Figure 2

pH values of alkaline treated reactors

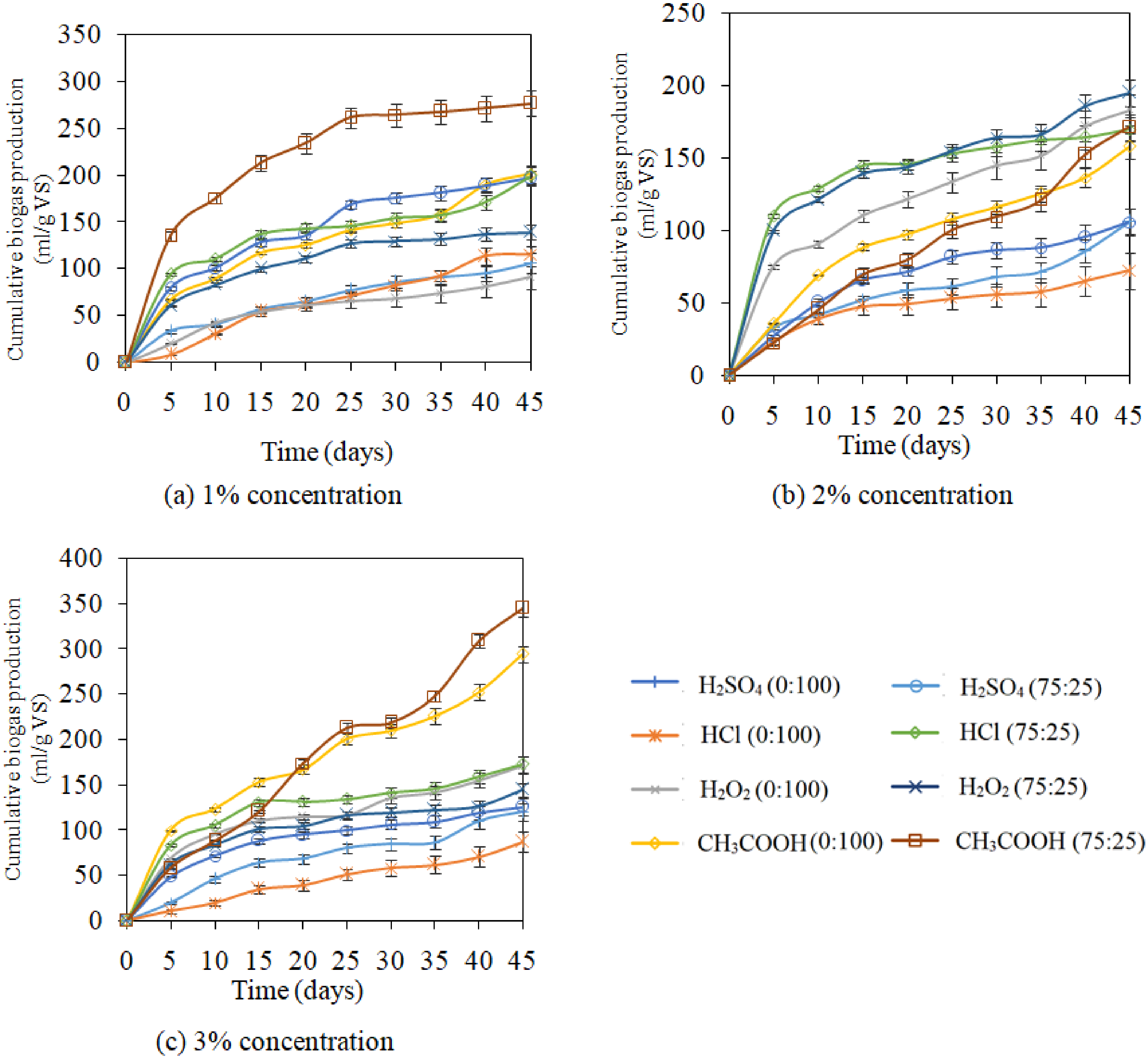


Figure 3

Biogas production from acid treated reactors

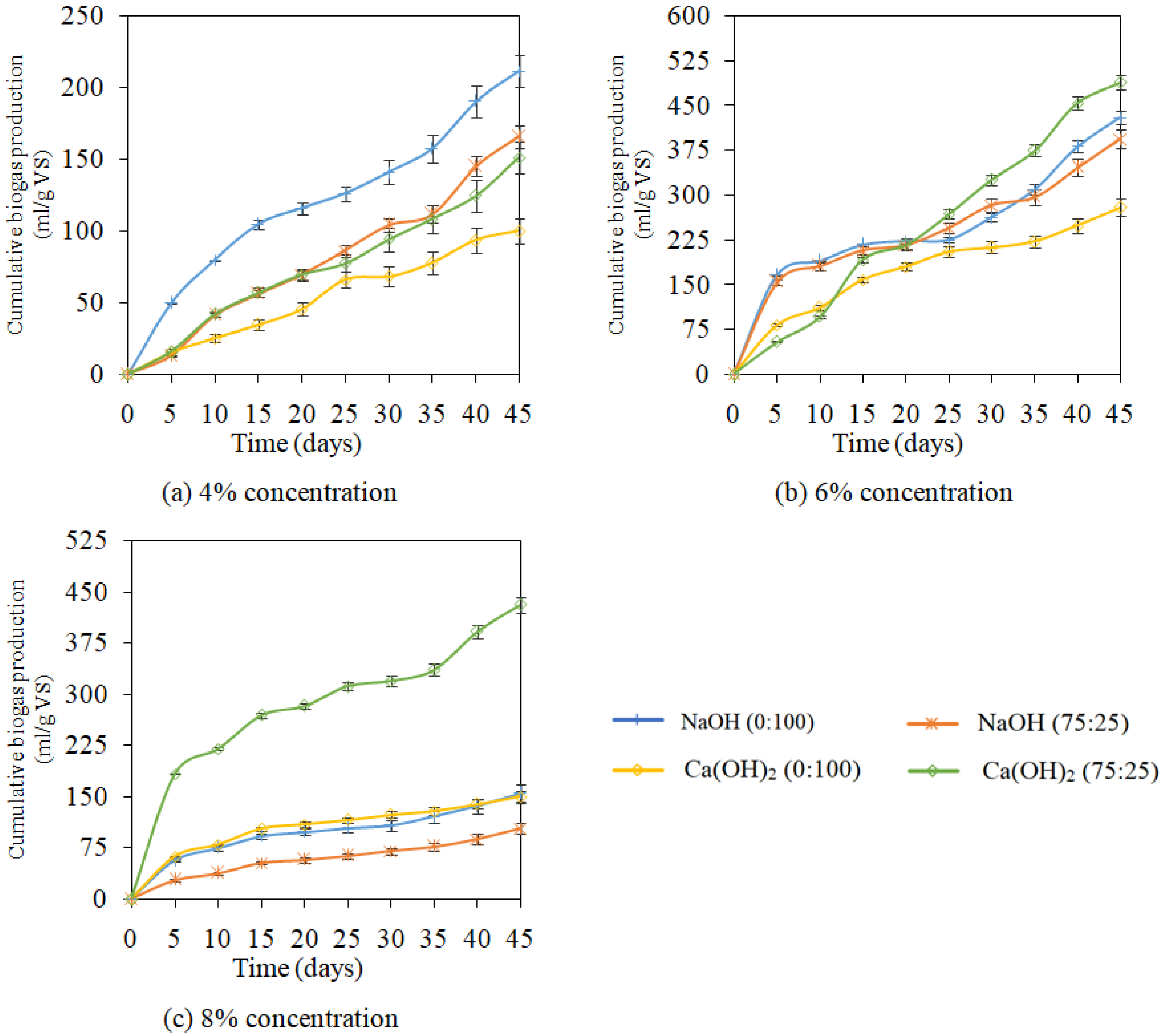


Figure 4

Biogas production from alkaline treated reactors

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