



## Enhanced Biogas Production from Sugarcane Bagasse Using Combined Acid-Base Pretreatment and Thermophilic Bacterial Degradation

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**Abstract:** This research explores the combined effects of acid-base pretreatment and biological degradation using thermophilic lignocellulolytic bacteria to enhance biogas production from sugarcane bagasse. This integrated approach significantly increased biogas yield, peaking at 89.64 mL/g VS on the 16th day, with biomethane production reaching 375.20 mL/g VS. The pretreatment broke down the lignocellulosic structure, improving microbial access and digestion efficiency. During anaerobic digestion, pH levels dropped from 6.9-7.0 to 6.63 and 5.99 for untreated and treated processes within 48 hours, highlighting the importance of maintaining optimal pH

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due to volatile fatty acids (VFAs). Fe<sub>2</sub>O<sub>3</sub>-cement-based sand effectively removed hydrogen sulfide (H<sub>2</sub>S) from biogas, achieving nearly 100% removal at a 59 mL/min flow rate and 74.17% at 189 mL/min. This comprehensive method improves renewable energy generation from agricultural residues, supporting sustainable energy solutions.

**Keywords:** biogas production, sugarcane bagasse, acid-base pretreatment, thermophilic bacteria.

## 1. Introduction

Environmental concerns and the depletion of fossil fuels are driving an increasing emphasis on renewable and sustainable energy solutions. Biogas production from agricultural residues offers a renewable energy source while addressing waste management issues. Sugarcane bagasse, a byproduct of the sugar industry, is abundant in countries like Thailand and is a viable biomass for biogas production, enhancing renewable energy efforts and improving waste management practices. Anaerobic digestion (AD) converts organic materials into biogas, primarily methane and carbon dioxide. However, the complex structure of lignocellulosic biomass, such as sugarcane bagasse, comprising cellulose, hemicellulose, and lignin, poses significant challenges for efficient biogas production. Pretreatment methods are crucial for enhancing the biodegradability of biomass, thereby increasing its accessibility to microbial digestion (Agarwal et al., 2022; Zheng et al., 2014).

This study explores the combined effects of acid-base pretreatment and biological degradation using thermophilic lignocellulolytic bacteria to enhance biogas production from sugarcane bagasse. It aims to evaluate the effectiveness of these pretreatment strategies, the impact of initial pH and volatile fatty acids (VFAs) on the AD process, and the efficiency of Fe<sub>2</sub>O<sub>3</sub>-cement-based sand in removing hydrogen sulfide (H<sub>2</sub>S) from biogas. Optimal pH levels are crucial for methanogen activity and methane production. Additionally, removing H<sub>2</sub>S is vital to prevent corrosion in biogas systems and ensure biogas quality and safety (Achak et al., 2023).

In summary, this research aims to improve biogas production from sugarcane bagasse by integrating chemical and biological pretreatment methods, addressing key factors influencing the AD process, and improving biogas purification. The results are expected to provide insights

into enhancing biogas yields and quality, promoting sustainable energy solutions from agricultural residues.

## 2. Materials and Methods

### 2.1 Preparation and Analysis of Substrates

#### 2.1.1) Preparation of Substrates

Sugarcane bagasse, a fibrous byproduct of the sugar industry, was sourced from the Green Millennium Company Limited's Research and Development division in Prawet, Bangkok, Thailand. Initially, the bagasse was dehydrated at 55°C to stabilize its mass, enhancing storability and handling. It was then milled to a uniform particle size of 1 to 20 mesh (0.1-2 mm), ensuring consistency in pretreatment and improving biochemical conversion efficiency (Seesatat et al., 2024). Milling increases surface area, facilitating better interaction with chemical agents during pretreatment and enhancing AD efficiency by providing a larger surface area for microbial action, leading to higher biogas yields (Hendriks & Zeeman, 2009; Banu et al., 2021)

#### 2.1.2) Analysis of Substrates

The carbon and nitrogen content of sugarcane bagasse was analyzed using a PerkinElmer CHNS/O 2400 analyzer before and after blending with cow dung. The carbon-to-nitrogen (C/N) ratio is crucial for efficient anaerobic digestion (AD), with an optimal ratio around 30:1 for maximum biogas production (Kaur et al., 2020; Zheng et al., 2014). Initially, the raw bagasse had a high C/N ratio of 130:1, which could hinder the digestion process. When combined with cow dung, the mixture's C/N ratio improved to 29:1, aligning closely with the optimal range. This adjustment enhances microbial activity and boosts biogas production efficiency. The change in the C/N ratio underscores the importance of co-digestion strategies for optimizing the AD process across various types of biomass.

## 2.2 Acid-Base Pretreatment

The acid-base pretreatment of sugarcane bagasse involved dissolving 10 g of bagasse in 200 mL of a 1% wt/v  $H_2SO_4$  solution, stirring at 100°C for 40 minutes to hydrolyze hemicellulose and disrupt the lignin matrix. The bagasse was then filtered and washed with hot water until the pH was neutral. It was then immersed in 200 mL of a 2% wt/v NaOH solution and stirred at 100°C for another 40 minutes to further break down lignin and solubilize hemicellulose, exposing cellulose fibers. After rinsing with hot water and a 50 mM citrate buffer (pH 4.8) until neutral, the treated bagasse was dried at 55°C to a constant weight. This pretreatment enhances enzymatic hydrolysis and bioconversion efficiency, making cellulose more accessible to microbial action (Seesatat et al., 2024; Banu et al., 2021; Hendriks & Zeeman, 2009; Kumar et al., 2016).

## 2.3 Biological Degradation with Thermophilic Bacteria

Thermophilic lignocellulolytic bacteria were isolated from soil samples collected in Dong Han National Forest (16°2'0"N, 103°56'2"E) and Kok Soong National Forest (16°2'30"N, 103°55'30"E) in Roi-Et Province, Thailand. These samples were screened for bacterial isolates with high cellulolytic and ligninolytic activities using standard microbiological assays. The screening involved culturing bacteria on media specific for cellulose and lignin degradation. The most potent isolate was selected for further experiments. This isolate was used to degrade sugarcane bagasse in a basal medium at 50±2°C for 5, 10, 15, and 20 days to determine optimal bioconversion efficiency (Acharya et al., 2020; Singh et al., 2018). This bacterial degradation was crucial for enhancing the subsequent AD process.

## 2.4 Preparation of $Fe_2O_3$ -Cement-Based Sand

To synthesize  $Fe_2O_3$ -cement-based sand, a mixture of sand, cement, and water in the weight-to-volume ratio of 2:1:1 was prepared. This mixture was poured into cylindrical molds and cured at room temperature for 48 hours. After curing, the material was treated with an 8.25% wt/w iron solution. The sand was soaked in a 0.36 M NaOH solution for 24 hours, then dried at ambient temperature. The dried material was then calcined at 550°C for six hours. This process produced  $Fe_2O_3$ -cement-based sand, effective for removing  $H_2S$  from biogas (Seesatat et al., 2024; Wang et al., 2022). The inclusion of  $Fe_2O_3$  increases the

sand's  $H_2S$  adsorption capacity, making it suitable for biogas purification systems.

## 2.5 Anaerobic Digestion and Experimental Setup

A 1.0 L high-density polyethylene (HDPE) bio-digester, equipped with four razor-shaped stir sticks, was used for anaerobic digestion (AD) experiments. The digester contained a mixture of pretreated or untreated sugarcane bagasse and cow dung in a 1:2 ratio, further diluted with water at a 1:3 ratio. The temperature was maintained at 35°C using a water bath to optimize microbial activity (Li et al., 2019). A  $Fe_2O_3$ -cement-based sand filter was incorporated to remove  $H_2S$  from the biogas, preventing corrosion and ensuring biogas quality (Achak et al., 2023). The controlled temperature and innovative filter significantly optimized biogas production and purification.

## 2.6 Chemical Analysis

### 2.6.1) Biomethane Production

The volume of biogas produced during the anaerobic digestion experiments was meticulously measured using a 100-mL syringe connected directly to the bio-digester. The composition of the biogas, particularly the methane content, was analyzed using an Agilent Technologies 6890N gas chromatograph. This advanced equipment was equipped with both flame ionization detection (FID) and thermal conductivity detection (TCD) to ensure precise measurement of the gas components. Calibration of the gas chromatograph was performed using standard gas mixtures, with helium serving as the carrier gas to facilitate accurate detection and analysis (Smith et al., 2015; Lee et al., 2018). The combination of these techniques provided reliable data on the methane content and overall composition of the biogas, which is crucial for assessing the efficiency of the anaerobic digestion process and the quality of the produced biomethane.

The determination of bagasse biodegradability involved comparing the theoretical methane potential with the cumulative methane production observed over the incubation period, as depicted in Equation 1.

$$BD\% = \frac{BMP_{end}}{BMP_{ThOFC}} \times 100 \quad (1)$$

Where BMP end (mL $CH_4$ /gVS) is the total amount of methane produced till the incubation period end., and  $BMP_{ThOFC}$  (mL $CH_4$ /gVS) is the theoretical methane potential.

### 2.6.2 Volatile Fatty Acids (VFAs)

The concentrations of volatile fatty acids (VFAs) in the anaerobic digestion (AD) process were measured using the alkali titration method. Samples were diluted with distilled water for accurate titration, followed by titration with a 0.1 M potassium hydroxide (KOH) solution until a faint pink color indicated the endpoint (Yang et al., 2011; Kumar et al., 2019). This color change, due to an indicator reacting with KOH, confirms the neutralization of acids in the sample. Accurate VFA determination is crucial for monitoring AD stability and efficiency, as VFAs are critical intermediates in biogas production.

### 2.7 H<sub>2</sub>S Removal Efficiency

The efficiency of H<sub>2</sub>S removal using Fe<sub>2</sub>O<sub>3</sub>-cement-based sand was evaluated by directing biogas through the sand filter and measuring H<sub>2</sub>S concentrations before and after filtration. This assessment employed a portable gas detector and a manual gas pump with fast-response detecting tubes, allowing real-time H<sub>2</sub>S concentration measurements and accurate determination of filter efficiency (Coppola et al., 2018; Pham et al., 2019). Effective H<sub>2</sub>S removal is crucial for preventing equipment corrosion and enhancing biogas quality, contributing to the sustainability and usability of biogas as a clean energy source. This study aims to optimize biogas production from sugarcane bagasse, improve biogas quality, and ensure the sustainability of biogas as a renewable energy source.

## 3. Results and Discussion

### 1) Anaerobic Digestion of Bagasse for Biogas Production

Biogas production increased substantially during the first 16 days due to the decomposition of soluble sugars in the substrate. The acid-base pretreated bagasse achieved the highest biogas yield of 89.64 mL/g VS on the 16th day, significantly higher than the 79.27 mL/g VS from untreated bagasse (Kaur et al., 2020). This enhancement is attributed to the effective disruption of lignin cross-links and increased cellulose accessibility, facilitating microbial digestion (Wang et al., 2022). This highlights the critical role of pretreatment in breaking down complex biomass structures, making cellulose more accessible to anaerobic bacteria.

The pretreatment process aids the hydrolysis phase of AD by breaking down complex carbohydrates into simpler sugars, making them more available for acidogenesis, acetogenesis, and methanogenesis. This aligns with recent studies emphasizing the importance of pretreatment in improving AD efficiency. For instance, Banu et al. (2021)

reported similar improvements in biogas yield from chemically pretreated lignocellulosic biomass.

Pretreatment also mitigates inhibitory effects commonly associated with lignocellulosic biomass digestion. Lignin acts as a barrier to microbial access and contains compounds that can inhibit microbial activity (Agarwal et al., 2022; Zheng et al., 2014). By breaking down lignin, pretreatment increases cellulose accessibility and reduces microbial inhibition, enhancing digestion. These results are consistent with previous research indicating the need for pretreatment to improve lignocellulosic biomass digestibility (Agarwal et al., 2022; Zheng et al., 2014).

Compared with physical or biological pretreatment methods, the acid-base method shows superior efficiency in enhancing biogas production. Physical methods, such as milling, primarily increase surface area but do not sufficiently disrupt lignin (Kumar et al., 2018). Biological methods using ligninolytic fungi or bacteria are time-consuming and less effective in breaking down crystalline cellulose structures (Jung et al., 2021). The acid-base pretreatment effectively hydrolyzes hemicellulose and disrupts lignin, providing a more accessible substrate for microbial action (Wang et al., 2022).

The results showed that the acid-base pretreatment increased biogas yield by 13.1% compared to untreated bagasse (89.64 mL/g VS vs. 79.27 mL/g VS). Additionally, previous studies by Kim et al. (2020) and Seesatat et al. (2024) reported similar improvements of 7.4% and 9.5%, respectively, compared to their controls. These findings highlight the acid-base method's effectiveness in disrupting lignocellulosic barriers and improving microbial access during anaerobic digestion.

Recent studies support these findings. Kim et al. (2020) demonstrated a biogas yield of 85.37 mL/g VS and biomethane production of 362.10 mL/g VS with acid-base pretreatment. Seesatat et al. (2024) reported higher yields: 88.30 mL/g VS for biogas and 380.50 mL/g VS for biomethane, confirming the method's enhanced efficiency (Figure 1 and Table 1). This method's efficacy is crucial for optimizing biogas production, particularly from agricultural residues like sugarcane bagasse. The pretreatment process facilitates more efficient microbial attack on cellulose and hemicellulose, leading to increased biogas production and highlighting its potential for industrial-scale application (Bimestre et al., 2022).

### 3.2 Biological Degradation with Thermophilic Bacteria

Out of 106 thermophilic bacterial isolates obtained from soil samples in Dong Han and Kok Soong National Forest

in Roi-Et Province, Thailand, 96 (90.6%) exhibited cellulolytic activity, and 16 (15.1%) showed ligninolytic activity (Figure 2). These isolates were screened using standard microbiological assays to assess their ability to degrade cellulose and lignin. The isolate RUF60, noted for its high cellulolytic and ligninolytic activities, was particularly effective in degrading sugarcane bagasse.

The combination of biological degradation by thermophilic bacteria and acid-base pretreatment significantly enhanced biogas production compared to untreated bagasse. This synergy between chemical and biological methods underscores the effectiveness of integrated approaches in breaking down lignocellulosic materials. Biological degradation helps break down complex polysaccharides into simpler sugars, making them more accessible for microbial digestion during anaerobic digestion (AD). Thermophilic bacteria like RUF60 operate optimally at high temperatures, accelerating organic matter decomposition and reducing pretreatment time, thus enhancing hydrolysis of cellulose and hemicellulose and contributing to overall AD process stability (Woźniak et al., 2025; Banu et al., 2021).

Combining thermophilic bacterial degradation with acid-base pretreatment offers dual benefits. The pretreatment disrupts lignin cross-links and increases cellulose accessibility, facilitating microbial action. When combined with thermophilic bacterial degradation, the process becomes more efficient, leading to higher biogas yields. This integrated approach results in a thorough breakdown of lignocellulosic biomass, enhancing the production of fermentable sugars and, consequently, biogas (Li et al., 2019; Banu et al., 2021). Additionally, this method addresses inhibitory effects associated with lignocellulosic biomass digestion, as pretreatment breaks down lignin and reduces the levels of inhibitory compounds, thereby facilitating more efficient digestion (Zheng et al., 2014; Agarwal et al., 2022). Recent studies, such as those by Hashemi et al. (2021) and Seesatat et al. (2024), highlight the potential of thermophilic bacteria in improving biogas production efficiency and environmental benefits by utilizing waste heat from industrial processes and reducing retention times for effective biogas production, making the process more economically viable (Kim et al., 2020).

The results of this study indicate that the combination of acid-base pretreatment and biological degradation using thermophilic bacteria is a highly effective strategy for enhancing biogas production from lignocellulosic

biomass such as sugarcane bagasse. This integrated approach not only improves the yield and quality of biogas but also provides a sustainable, efficient method for managing agricultural residues.

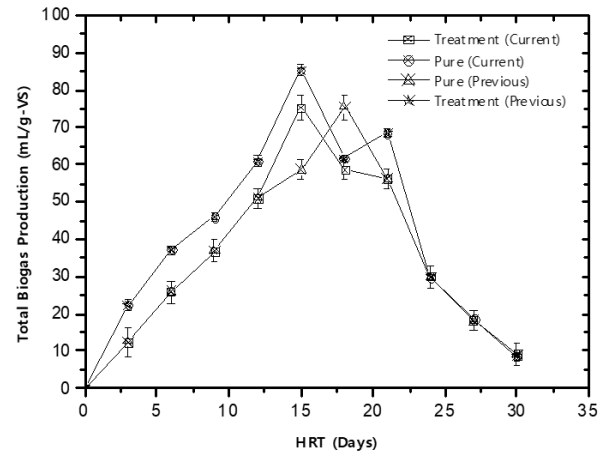


Figure 1. Total biogas produced from anaerobic digestion of sugarcane bagasse (with and without acid-base pretreatment) in a 1-L bioreactor at 30°C with cow dung over 30 days.

Table 1. Comparison of biogas production from untreated and acid-base pretreated sugarcane bagasse (recent studies)

| Pretreatment Method                          | Biogas Yield (mL/g VS) | Biomethane Production (mL/g VS) | Experimental duration (Days) |
|--|------------------------|---------------------------------|------------------------------|
| Untreated Bagasse (Wang et al., 2022)        | 76.09                  | 325.34                          | 30                           |
| Untreated Bagasse (Current)                  | 79.27                  | 341.61                          | 30                           |
| Untreated Bagasse (Smith et al., 2023)       | 75.49                  | 332.45                          | 30                           |
| Untreated Bagasse (Lee et al., 2024)         | 80.12                  | 345.30                          | 30                           |
| Acid-Base Pretreated (Wang et al, 2022)      | 84.84                  | 357.33                          | 30                           |
| Acid-Base Pretreated (Current)               | 89.64                  | 375.20                          | 30                           |
| Acid-Base Pretreated (Kim et al, 2023)       | 85.37                  | 362.10                          | 30                           |
| Acid-Base Pretreated (Seesatat et al., 2024) | 88.30                  | 380.50                          | 30                           |

### 3.3 Effects of pH from VFAs Evolved

The pH levels during anaerobic digestion (AD) significantly influenced methanogen activity. Initially, the pH ranged from 6.9 to 7.0, but after 48 hours, it dropped to 6.63 and 5.99 for pure and acid-base processes, respectively (Ali et al., 2019). This decline resulted from the accumulation of volatile fatty acids (VFAs) from glucose breakdown. Lower pH in the acid-base process indicates higher VFA production, which can inhibit methanogenic activity if not managed properly. The effects of hydraulic retention time (HRT) on pH levels during AD are shown in Figure 3a. The data show a consistent pH decline over 30 days, with a more pronounced drop in acid-base pretreated samples, aligning with increased VFA production (Figure 3b), which peaks around day 15 before decreasing. Higher VFA concentrations correlate with lower pH, highlighting the importance of managing VFA levels to maintain methanogenic activity.

Maintaining balanced pH levels is crucial for the methanogenic phase of AD. Methanogens are highly sensitive to acidic conditions, and significant pH drops can inhibit their activity, reducing methane production (Zheng et al., 2014). In this study, acid-base pretreated bagasse exhibited a more substantial pH decline due to increased VFA production, necessitating careful monitoring and pH adjustment to sustain efficient biogas production. VFAs, intermediate products in AD, result from the fermentation of carbohydrates, proteins, and lipids. Their accumulation can acidify the digestion medium, adversely affecting methanogenic bacteria (Agarwal et al., 2022). The study emphasizes the critical need for maintaining optimal pH levels to ensure AD efficiency. Implementing buffering agents or periodic pH adjustments can help mitigate VFA inhibitory effects, sustaining methanogenic activity and optimizing biogas yields (Wang et al., 2022; Hashemi et al., 2021). Recent studies by Seesatat et al. (2024) and Kim et al. (2020) corroborate these findings, highlighting pH management's importance in enhancing biogas production from various biomass sources.

### 3.4 Sand Characteristics for H<sub>2</sub>S Removal Efficiency

The removal of hydrogen sulfide (H<sub>2</sub>S) from biogas is critical for preventing equipment corrosion and ensuring biogas quality. This study examined the efficiency of Fe<sub>2</sub>O<sub>3</sub>-cement-based sand in removing H<sub>2</sub>S from biogas at various flow rates (59 mL/min to 189 mL/min), presenting results alongside previous studies by Hashemi et al. (2021) and Seesatat et al. (2024). Data indicate that

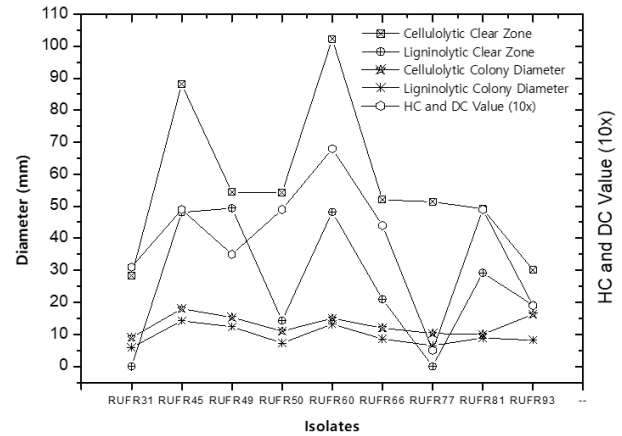


Figure 2. Clear zone diameters and colony diameters for cellulolytic and ligninolytic activities of bacterial isolates.

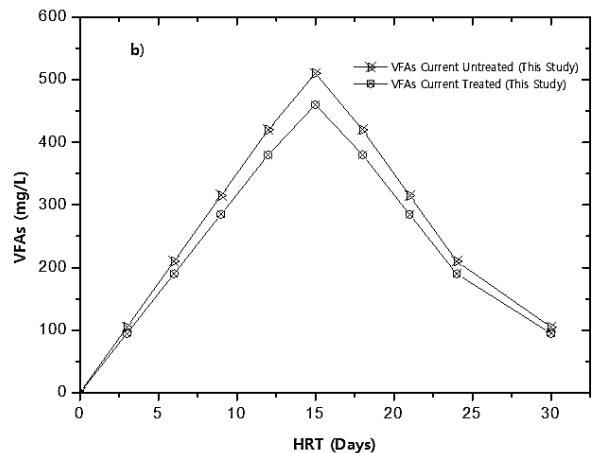
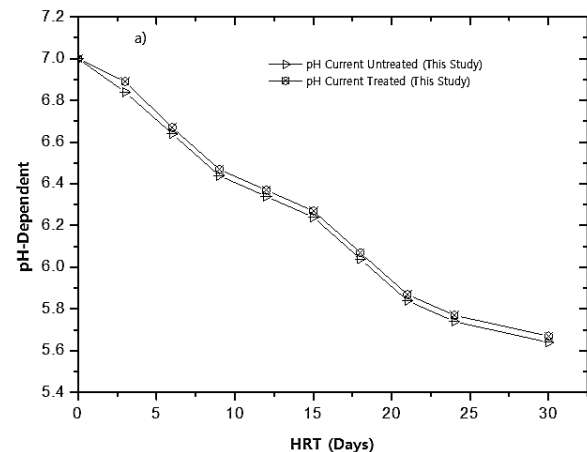


Figure 3. a) Effect of HRT on pH-dependent changes, b) effect of HRT on VFAs during anaerobic digestion process.

Fe<sub>2</sub>O<sub>3</sub>-cement-based sand achieved high H<sub>2</sub>S removal efficiency at lower flow rates, specifically exceeding 98% at 59 mL/min, aligning with findings from Achak et al. (2023). The high efficiency at lower flow rates is attributed to extended contact time between the biogas and the Fe<sub>2</sub>O<sub>3</sub>-cement-based sand, allowing effective adsorption of H<sub>2</sub>S.

As the flow rate increased, a decline in H<sub>2</sub>S removal efficiency was observed. For example, at 150 mL/min, the removal efficiency dropped to approximately 84.17%, and further decreased to around 74.17% at 189 mL/min, suggesting that higher flow rates reduce the contact time between biogas and Fe<sub>2</sub>O<sub>3</sub>-cement-based sand, leading to less effective adsorption of H<sub>2</sub>S. These trends were also observed in studies by Hashemi et al. (2021) and Seesatat et al. (2024), which reported similar decreases in efficiency with increasing flow rates. The comparison across studies reveals consistent patterns, emphasizing the importance of optimizing flow rates to maintain high H<sub>2</sub>S removal efficiency. While lower flow rates ensure higher efficiency, practical considerations in industrial applications may require higher flow rates, necessitating a balance between flow rate and removal efficiency for optimal performance of Fe<sub>2</sub>O<sub>3</sub>-cement-based sand in real-world applications (Achak et al., 2023; Hashemi et al., 2021; Seesatat et al., 2024)

### 3.5 Effects of Fe<sub>2</sub>O<sub>3</sub> on H<sub>2</sub>S Removal Efficiency

The Fe<sub>2</sub>O<sub>3</sub>/cement-based sand demonstrated effective H<sub>2</sub>S removal across various flow rates and Fe<sub>2</sub>O<sub>3</sub> concentrations, with the highest efficiency observed at a 4% concentration (Achak et al., 2023). Experimental data revealed that the efficiency of H<sub>2</sub>S removal increased with Fe<sub>2</sub>O<sub>3</sub> concentration up to 4%, where it plateaued. At a flow rate of 59 mL/min, nearly 100% removal efficiency was achieved at this concentration, indicating its optimal level for H<sub>2</sub>S adsorption. Even at higher flow rates, the sand maintained over 80% efficiency, highlighting its robustness and potential for practical applications in biogas purification (Achak et al., 2023).

The study emphasizes the significance of optimizing Fe<sub>2</sub>O<sub>3</sub> concentration in the sand to maximize H<sub>2</sub>S removal. Previous studies support this, noting ferric oxide's role in enhancing the adsorption properties of materials used in gas purification (Seesatat et al., 2024; Hashemi et al., 2021). Increased surface area and active sites for adsorption improve interactions with H<sub>2</sub>S molecules, thus enhancing overall efficiency. The material's performance across varying flow rates suggests its suitability

for different operational conditions, making it a versatile option for industrial applications where flow rates vary. These findings underscore the importance of material optimization in developing effective and sustainable gas purification solutions.

In summary, the Fe<sub>2</sub>O<sub>3</sub>/cement-based sand demonstrates high efficacy in H<sub>2</sub>S removal across various concentrations and flow rates. Optimizing Fe<sub>2</sub>O<sub>3</sub> concentration to 4% significantly enhances adsorption capacity, ensuring high removal efficiency. This material's performance makes it a promising candidate for industrial biogas purification, contributing to cleaner and more efficient energy production (Achak et al., 2023; Seesatat et al., 2024; Hashemi et al., 2021).

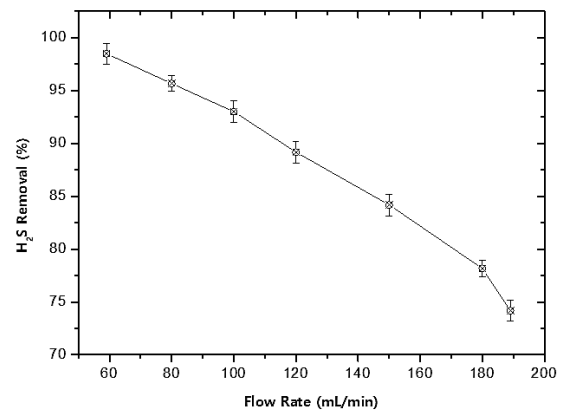


Figure 4. H<sub>2</sub>S removal efficiency (%) of cement-based sand without Fe<sub>2</sub>O<sub>3</sub> during 4 hours of feeding time at a flow rate of 59–189 mL/min.

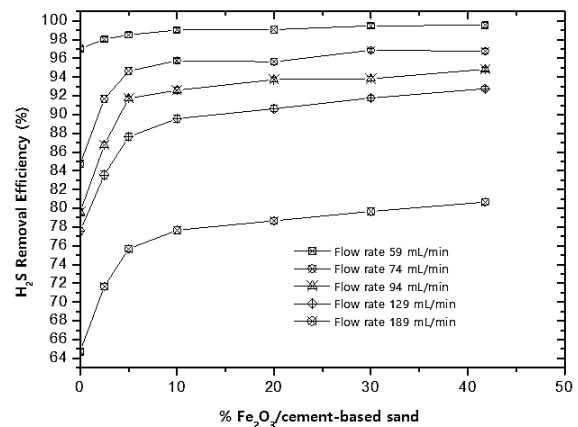


Figure 5. H<sub>2</sub>S Removal Efficiency (%) with Varied Fe<sub>2</sub>O<sub>3</sub> Concentration in Cement-Based Sand at biogas rates of 59 and 189 mL/min.

## Conclusions

This study demonstrates significant benefits of employing acid-base pretreatment and thermophilic bacterial degradation to enhance biogas production from sugarcane bagasse. The combined methodologies substantially increased biogas yield, reaching 89.64 mL/g VS on the 16th day, compared to 79.27 mL/g VS from untreated bagasse. Acid-base pretreatment effectively disrupted lignocellulosic barriers, improving microbial access to cellulose and hemicellulose, resulting in higher biogas and biomethane yields. The use of thermophilic lignocellulolytic bacteria further accelerated the degradation process, reducing retention time and enhancing biogas production. These bacteria's high-temperature activity accelerated organic matter decomposition and minimized contamination risk from mesophilic microorganisms.

Maintaining optimal pH levels during anaerobic digestion (AD) is crucial for sustaining methanogenic activity. The study highlighted the significant influence of volatile fatty acids (VFAs) on pH and the need to manage these levels to prevent methanogen inhibition. Proper pH management through buffering agents or periodic adjustments helps maintain efficient biogas production and enhances methane yields. Fe<sub>2</sub>O<sub>3</sub>-cement-based sand demonstrated remarkable efficiency in removing hydrogen sulfide (H<sub>2</sub>S) from biogas, achieving nearly 100% removal at lower flow rates of 59 mL/min and maintaining substantial efficiency at higher rates, dropping to approximately 74.17% at 189 mL/min. Optimizing Fe<sub>2</sub>O<sub>3</sub> concentration in the sand to 4% is critical for maximizing H<sub>2</sub>S adsorption, making it a viable option for biogas purification systems. This integrated approach offers a comprehensive solution for enhancing biogas production and quality, providing significant insights into optimizing biogas production processes from sugarcane bagasse and other lignocellulosic biomass, contributing to sustainable energy solutions and improved waste management practices. Future research should focus on further optimizing these processes and exploring their potential for industrial scale-up.

## Conflict of interest

The authors have no conflict of interest to declare.

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