

**Experimental Approach to Produce Biogas from Fallen Leaves available in IUT
Campus**

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**A Thesis submitted in partial fulfillment of the requirement for the degree of Bachelor
of Science in Mechanical Engineering**



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Candidate's Declaration

This is to certify that the work presented in this thesis, titled, "**Experimental Approach to Produce Biogas from Fallen Leaves available in IUT Campus**", is the outcome of the investigation and research carried out by me under the supervision of **PROF.DR.MD.HAMIDUR RAHMAN, Professor, MPE, IUT**. It is also declared that neither this thesis nor any part of it has been submitted elsewhere for the award of any degree or diploma.

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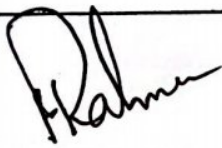
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Abstract

This paper describes an experimental approach to produce biogas from fallen leaves available on the campus of IUT. Four types of leaves were selected for this experiment, namely *Mangifera indica* (mango leaves), *Tectona grandis* (teak leaves), *Swietenia macrophylla* (mahogany leaves), and *Artocarpus heterophyllus* (jackfruit leaves). These leaves were collected from the IUT campus and used as the substrate for biogas production. The collected leaves were first chopped and mixed with water to form a slurry. This slurry was then added to an anaerobic digester, where it underwent the process of anaerobic digestion. The biogas produced was analyzed for its composition, and the results showed that it was primarily composed of methane and carbon dioxide. The production of biogas from fallen leaves has several benefits, including the reduction of greenhouse gas emissions and the generation of a renewable energy source. Furthermore, the use of fallen leaves as a substrate for biogas production can help to reduce the amount of waste on the IUT campus. Overall, this study demonstrates that fallen leaves can be used as a viable substrate for biogas production and highlights the potential of this approach to be used in other similar settings. The experimental approach used in this study has shown that fallen leaves can be used as a substrate for biogas production, and that this approach has the potential to be used in other similar settings. The study also highlights the importance of anaerobic digestion as a means of producing renewable energy and reducing waste on college campuses. Further research is needed to optimize the process and to explore the potential of this approach to be used on a larger scale.

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Nomenclature

Food Waste	FW
Cow Dung	CD
Food and Agriculture Organization	FAO
Islamic University of Technology	IUT
Municipal Solid Waste	MSW
Anerobic Digestion	AD
Volatile Fatty Acid	VFAs
Total Solid	TS
Volatile Solid	VS

Chapter One: Introduction

1.1 Background of the study

The production of biogas from organic waste materials has been a topic of increasing interest in recent years, as it offers a sustainable solution for the disposal of waste and the generation of clean energy. Among the various organic waste materials, fallen leaves have been identified as a potential source for biogas production. However, the utilization of fallen leaves as a feedstock for biogas production has received limited attention in the literature. This study aims to explore an experimental approach to producing biogas from fallen leaves and to investigate the effect of different process parameters on the biogas yield. The world is transitioning away from petroleum-based national economies to ones based on biofuels due to the growing expense and environmental impact of fossil fuels. Different experiments have been done with various sources to gain more usable energy from natural resources. Producing biogas from organic objects is one of them. According to IRENA's latest study, nations want to add 5.4 terawatts (TW) of renewable energy capacity by 2030. Bangladesh also has made ambitious goals for the use of renewable energy at COP26. A target of 4,100 MW of renewable energy is included in the Nationally Determined Contribution (NDC) for 2030.

Fallen leaves are leaves that have fallen from trees and other plants. In locations with vegetation and trees, they can find them on the ground. Seasonal variations, environmental conditions, and the tree's general health are only a few reasons why leaves fall from trees. Seasonal variations are one of the primary causes of leaf fall from trees. To save energy and make it through the winter, deciduous trees, or trees that lose their leaves every year, shed their leaves in the fall. The tree's metabolism slows down and stops generating chlorophyll, the pigment that gives leaves their green hue, as the days become shorter and the temperature drops. The other pigments in leaves, such as carotenoids and anthocyanins, become more apparent when chlorophyll is absent, giving the leaves their yellow, orange, and red hues. Leaves can fall off trees due to environmental causes such as dryness, high or low temperatures, and insect infestations. The tree may get stressed during a drought and lose leaves to retain moisture.

To produce methane from biomass such as Leaves, Anaerobic digestion is the most efficient process, Anaerobic digestion is a biological process that transforms organic materials, such as food waste, agricultural waste, and sewage, into biogas and a nutrient-rich fertilizer. Without oxygen present, the process takes happens in an anaerobic environment. It occurs in several phases. Enzymes convert complex organic molecules into simpler ones, such as sugars and amino acids, during the first stage of hydrolysis. Bacteria transform these simpler substances into volatile fatty acids, alcohols, and other tiny organic molecules during the second step of acidogenesis. Different bacteria transform these small molecules into acetic acid, hydrogen, and carbon dioxide during the third step of acetogenesis. Methane-producing bacteria convert acetic acid, hydrogen, and carbon dioxide into methane and carbon dioxide during the process known as methanogenesis. The biogas produced through anaerobic digestion is composed primarily of methane and carbon dioxide and can be used as a source of energy for heating or electricity generation.

Co-digestion combines the breakdown of several kinds of organic waste in an anaerobic digestion system. There may be a variety of food wastes, agricultural wastes, and other organic items included in this. Co-digestion aims to improve anaerobic digestion by combining various organic wastes to provide the best possible environment for microorganisms to break down organic material. Co-digestion is a process that can be carried out in several ways, but it is most frequently carried out by combining several forms of organic waste in a single digester. This could include a mix of organic, agricultural, and food waste.

Compared to conventional anaerobic digestion techniques, co-digestion has several advantages. One key benefit is that it makes it possible to handle a wider variety of organic wastes, which can increase methane output and enhance process stability. Co-digestion can also aid in lowering the price of disposing of organic waste and expanding the supply of renewable energy.

1.2 Statement of the problem

Bangladesh's dependence on imported fossil fuels exposes it to global price volatility, which has a detrimental effect on its balance of payments. The pace of leaf fall varies amongst trees. In settlements, these discarded leaves often decayed or were burnt as fuel. However, methane gas may also be made from these leaves. Several tree species typically cover a sizable space at

Bangladesh's institutions. At IUT, one of Bangladesh's universities, many Jackfruit and Shogun leaves have fallen and are still on the ground or in the fields. By making biogas from these dried fallen leaves, they may be used.

1.3 Goal and Objectives

- Producing Methane from tree leaves that can be burned as fuel
- Studying the effect of Cow Dung on Biogas Production from Green Leaves
- Studying the impact of Urea on Bio Gas Production
- Comparing the production of Biogas between different plant leaf types.
- Observing the impact of pro-biotics in the formation of Bio-Gas

CHAPTER TWO: LITERATURE REVIEW

2.1 Anaerobic Digestion

Biogas, mostly made up of methane and carbon dioxide, is produced by anaerobic digestion, a biological process that involves the breakdown of organic waste in the absence of oxygen. We used a 2-liter digester and a gas collection apparatus to perform anaerobic digestion in the lab. To keep everything at a comfortable $30^{\circ}\text{C} + 3^{\circ}\text{C}$, electric lighting was strategically positioned near the digesters. Dried, crumbled leaves were combined with an appropriate amount of cow dung and water to meet the specified total solids concentrations. Fresh cow dung or slurry from a functioning digester served as the inoculum. For a total of seven days, gas output was tracked every five days. At its core, the gas is made up of methane and carbon dioxide, but their relative abundances change as the reaction develops. At first, carbon dioxide is the most abundant, but later on, methane production increases. Few gases were created by the green jackfruit, green mango, and dry mango leaves. After being treated with urea, a sample of dried teak leaves and jack fruit leaves generated 60-65% methane. Since urea aided in anaerobic digestion, we obtained a methane gas concentration in excess of 60%. Large quantities of leaves are often burned as fuel in rural areas, which is bad for the ecology. For this reason, we decided to concentrate on developing a method

to extract biogas from tree leaves. In this study, biogas was produced from cow manure, tree leaves, urea, and lake water in a digester.

2.2 Biogas from anaerobic process

Biogas, along with minor quantities of other gases, is produced during the fermentation process caused by AD. The whole transformation procedure consists of many steps. Biogas and methane are often produced by a number of different bacterial species engaging in a complex sequence of chemical reactions. An overview of the processes involved in creating biogas or methane is provided.

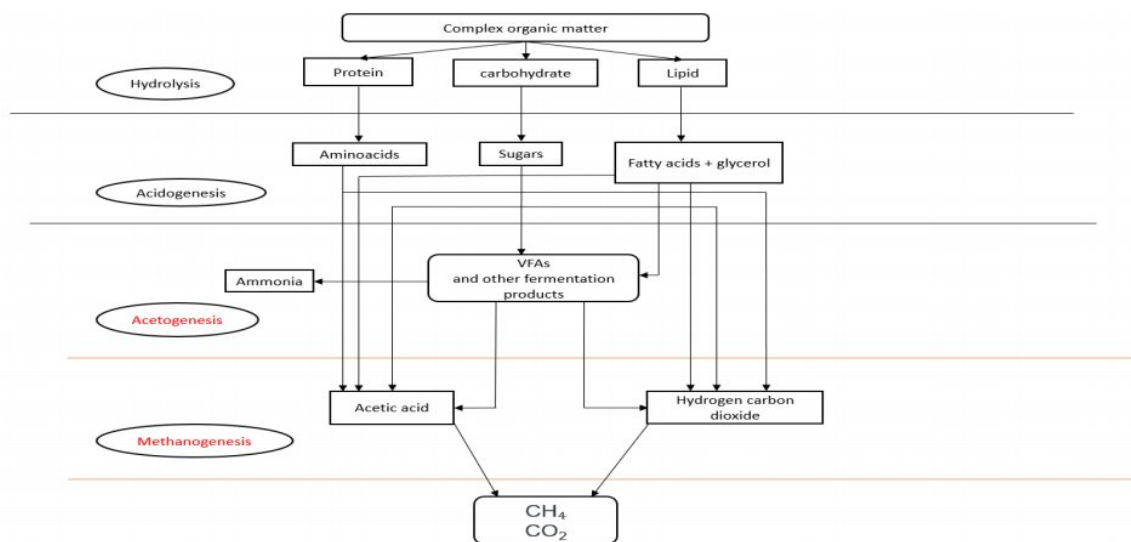
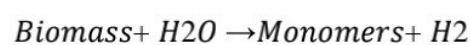


Figure 1 Anaerobic Digestion Process

2.2.1 Hydrolysis

The chemical process of hydrolysis reduces large organic molecules to their constituent monomers. Consider the following chemical process as an example of MSW hydrolysis. Fermentation and acetogenesis are the two processes that make acid. During this time period, glucose is often converted into ethanol and propionate. All of the acetogenesis reactions—converting glucose to acetate, ethanol to acetate, propionate to acetate, and bicarbonate to acetate—are required for acetogenesis to occur.



The various types of tree leaves were anaerobically digested for 55 days to produce biogas (for complete digestion). There is a close relationship between pH and anaerobic digestion; a balanced system will keep the pH somewhere in the range of 6.8 to 7.4. The anaerobic digestion process is also influenced by the digester and ambient temperatures. The first two days included making a slurry out of a 1:5 weight ratio of fresh cow dung to regular tap water. Using a conventional method for assessing water and waste water, the total solids, volatile solids, moisture content, and ash content of the water were calculated. It is everyone's obligation to "reuse, reduce, and recycle" as the world's population and demand for resources continue to rise. Utilizing eco-friendly technologies like anaerobic digestion allows us to reduce emissions of dangerous greenhouse gases and have a beneficial impact on our planet.

2.2.2 Acidogenesis

Ethanol, acids (including propionic and butyric acid), acetate, H₂O, and CO₂ are all byproducts of microorganisms that ferment carbohydrates and amino acids into their soluble organic monomers. Besides hydrogen gas, ammonia is another byproduct of amino acid degradation. It is the ability of acidogenic bacteria to take in the byproducts of hydrolysis via their cell membranes that allows them to produce intermediate volatile fatty acids (VFAs) and other chemicals. The class of organic acids known as volatile fatty acids includes both smaller organic acids such as acetates and larger organic acids such as propionate and butyrate (VFAs). In day-to-day life, it is not uncommon to come across ratios ranging between 75 and 15 to 10. Although trace quantities of ethanol and lactate may still be detectable. Contradictory data from various research suggest that the particular concentrations of intermediates generated during the acidogenesis stage may shift depending on digester settings, and it has been observed that VFA concentrations may vary greatly across digesters operating at different pH. Because acidogenic bacteria may reproduce in a shorter amount of time than 36 hours, it is common practice to presume that acidogenesis occurs quicker than the other stages of anaerobic digestion. Because of the speed with which it takes place, VFA acidification is often the root cause of digester failure. Methanogenesis, the last stage

in the process, occurs after naturally occurring volatile fatty acids (FFAs) are produced during the fermentation portion of the process. These bacteria are employed to breakdown the rubbish in a manner that is similar to the Bokashi composting method. When amino acids are deaminated to ammonia, the process of anaerobic digestion is slowed down.

2.2.4 Acetogenesis

Acetogenesis is a process that occurs during anaerobic digestion, in which microorganisms convert carbon compounds such as sugars and organic acids into acetate, hydrogen, and carbon dioxide. This process is typically carried out by a group of microorganisms known as acetogens.

The main reactions that occur during acetogenesis include:

1. Conversion of sugars (such as glucose) into organic acids (such as acetic acid) through the process of acidogenesis.
2. Conversion of organic acids (such as acetic acid) into acetate through the process of acetogenesis.
3. Conversion of hydrogen into methane through the process of methanogenesis.

The overall reaction for acetogenesis can be represented by the following equation:



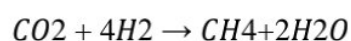
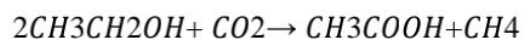
In this equation, glucose ($\text{C}_6\text{H}_{12}\text{O}_6$) is converted into acetic acid (CH_3COOH) and carbon dioxide (CO_2) through the process of acidogenesis. Then, acetic acid is converted into acetate (CH_3COO^-), carbon dioxide and hydrogen through the process of acetogenesis.

It's worth noting that acetogens are the only organisms that are capable of converting acetic acid into acetate, thus playing a crucial role in the overall anaerobic digestion process. Additionally, the hydrogen produced during acetogenesis can be used by methanogens in the process of methanogenesis to produce methane, which is a valuable energy source.

2.2.3 Methanogenesis

Methanogenic bacteria are responsible for the last step of the process, which consists of the conversion of hydrogen and acetic acid into methane gas and carbon dioxide. Methanogenesis is affected by a number of characteristics in the reactor, including temperature, feed content, and organic loading rate, among others.. Biogas mostly consists of methane (CH₄) and carbon dioxide (CO₂), but it also contains nitrogen, oxygen, and hydrogen. Hydrogen sulfide (H₂S) is responsible for the rotten egg smell. The methane content in combustible biogas must be at least 45 percent since the more CH₄ there is, the more energy there is in the gas. The production of methane by methanogenic microbes is the ultimate stage of anaerobic digestion. A need for anaerobic archaea such as methanogenic bacteria, 99 percent of whose cells die off after only 10 hours in the presence of oxygen. Methanogenic bacteria are very oxygen-sensitive and selective in their substrate preferences. Methanogenesis has also been seen using methanol, methylamines, and formates. The redox potential of methanogenic bacteria is expected to be lower than in the first stages of anaerobic digestion, which has led to significant laboratory culture difficulties. Methanogens seem to regenerate at a considerably slower pace than other species. Anaerobic bacteria may live for anywhere between five and sixteen days without oxygen. A good example of a fast-growing hydrogenotrophic bacterium is *Methanococcus maripaludis*, which may double in population in under two hours. Extreme resistance to environmental conditions such high ammonia, salt, and acetate concentrations and rapid pH shifts has been shown for *methanosarcina* spp. Reducing biogas output in batch reactors stops methanogenesis, which may take up to 40 days. The concentration of volatile solids and the ability to be dewatered are two measures that may be used to assess the quality of digested sludge.

The process of methanogenesis occurs at the end of anaerobic digestion. The results of these activities are fed into a series of reactions, the most important of which produces methane. The most often seen reactions during methanogenesis are:



It's important to note that many other types of bacteria contribute to methanogenesis, not only *Methanobacterium*, *Methanobacillus*, *Methanococcus*, and *Methanosarcina*. Anaerobic digesting bacteria are possible inhabitants of our own digestive tracts and are unique from other biofuel-producing enzymes.

Anaerobic digesters may be fed a wide variety of organic materials including manure and litter, food wastes, green wastes, plant biomass, and sewage sludge. To break down cellulose and hemicellulose, two chemical components that disintegrate extremely slowly, hydrolysis is the rate-limiting step. Examples of chemical compounds that the body cannot degrade include lignin, peptidoglycan, and membrane-associated proteins. As we have shown, pretreatment of biomass prior to ethanol synthesis has the potential to increase the efficiency of anaerobic digestion. Pretreatment reduces the recalcitrance of the biomass, making it easier to hydrolyze cellulose and hemicellulose.

2.3 Effect of urea addition on anaerobic digestion's ability to produce methane

Urea is an important additive in the production of biogas as it increases the efficiency of the anaerobic digestion process. Urea is a source of nitrogen, which is an essential nutrient for the growth of methanogens, the microorganisms responsible for the production of methane in biogas. The addition of urea to the substrate can increase the rate of methanogenesis and thus increase the yield of biogas. According to scientific literature, the addition of urea to the substrate can also increase the overall efficiency of the anaerobic digestion process. Studies have shown that the addition of urea to the substrate can increase the methane content of the biogas and decrease the amount of volatile fatty acids (VFAs) produced during the process. This is because the addition of urea can shift the balance of the microbial community in the anaerobic digester towards methanogens, which results in a higher methane yield and a lower accumulation of VFAs. Additionally, urea can also improve the stability of the anaerobic digestion process. The addition of urea can increase the pH of the substrate, which can inhibit the growth of acidogenic bacteria and thus prevent pH drops that can inhibit the growth of methanogens. This can lead to a more stable and consistent biogas production over time.

2.4 Operational parameters

2.4.1 Temperature

It is between 30 and 40 degrees Celsius that mesophilic microorganisms thrive best, whereas for anaerobic bacteria, the ideal temperature is 37 degrees Celsius. Bacteria and archaea grow between 30 and 40 degrees Celsius in mesophilic environments, and above 60 degrees Celsius in thermophilic environments (optimum temperature 55 degrees Celsius). Digesters function more efficiently at mesophilic temperatures because mesophilic microbial populations are more resilient to fluctuations in their surroundings and consume less energy. Since there is less ammonia present when temperatures are lower, ammonium's influence is muted. More biogas will be produced if mesophilic bacteria have more time to develop in the digester. Thermophilic activity speeds up breakdown by more than 50%, which is particularly helpful for fatty substances. This results in more biogas being produced. The CO₂ content in biogas is increased by 2-4 percent in thermophilic digesters because CO₂ becomes less soluble at higher temperatures. While running the digester at thermophilic temperatures has some potential benefits, the extra energy requirements and instability make it impractical for use in impoverished countries.

2.4.2 pH

AD plants with a notable biogas production are typically stable at a pH between 6 and 7. Compared to methanogenesis, which occurs at a higher pH (>6.5), acidogenesis happens after digestion at a lower pH (5.5-6.5). (6.5–8). A constant buffering capacity of 3,000 mg/L is required at all times. Lime is often used to raise the pH in AD systems if it is excessively low. Sodium bicarbonate, on the other hand, may be used to adjust the pH level. Some area firms may even give out lime solutions if they have too much. Lime is usually more affordable than other building supplies. Lime's most typical unwanted consequences include precipitation and pipe blockage. Costs are greater because neither sodium bicarbonate nor sodium hydroxide ever precipitates. When compared to lime, sodium bicarbonate and sodium hydroxide might be harder to come by. Na salts

are suggested for quick relief. For substrates with a pH lower than 7, lime may be employed as a backup pH adjuster.

Chapter 3: Methodology

3.1 Leaves Collection

Fallen leaves were collected from different places of the IUT campus. Jackfruit tree leaves were collected from infront of the North Workshop. Fallen leaves from Teak and Mahgony was



Figure 2 IUT Campus Outline

collected from OIC tree park. Mango leaves were collected from behind the North Hall of Residence. An outline of IUT is attached for reference.

3.2 Leaves Processing

The leaves were collected and kept to dry for 3 days. After the moisture or any exterior water particle dried up, the leaves were mashed using a blender. The leaves were grounded. The fine size of the particles will allow the bacteria to have greater contact area thus better digestion.



Figure 4 Leaves Processing with Blender



Figure 3 Measuring Cow-dung

3.3 Temperature Control

An incubator-like setup was used to control the temperature. The setup included light bulbs and a temperature controller called a W1209. The controller was set to keep the temperature at 37 degrees Celsius. Once the temperature reached 37 degrees, the light bulbs would turn off. If the temperature dropped to 36.5 degrees, the light bulbs would turn back on and stay on until the temperature reached 37.5 degrees. The inside of the setup was insulated to prevent heat from escaping. Once the desired temperature of 37 degrees was reached, the setups were placed inside a temperature-controlled box and left there for a certain amount of time.

3.4 Initial Setup



Figure 5 Initial Setup of Digesters

Single bottle setup was done to examine the presence of methane in the digester. Leaf samples of 50g with 500 mL solution and 2% urea was mixed. This ratio of mixture yields the best results. The 500 mL solution was then transferred into 2L plastic bottles. The plastic bottles had a head with two outlets, one with ball valve and another with screw valve. The bottle was tightened and the air was removed using a vacuum pump. The plastic bottles were kept inside a black box at 35°. Gas chromatograph was used to analyze the gas and determine the percentage of different gases present in the digester. Gas samples were collected directly to the analyzer with the gas outlets of the bottle. pH meter was used to monitor the pH of the solution during every reading.

3.5 Inoculum

Cow-dung was collected and kept in a plastic tank for 30 days for bacteria to grow. It was kept in a sealed tank under atmospheric condition. It allowed the bacteria to adapt to mesophilic condition. After 30 days 5g cow-dung was mixed with the digester which was already under regular monitoring. Also, in one of the digesters, pro-biotics were added to understand the potential of probiotics in creating methanogenesis bacteria.

Figure 7 Vacuuming of the digester



Figure 6 Measurement of Urea



3.6 Slurry Preparation

Different reactors were used to digest different leaves. Urea was added to maintain the C/N ratio and to maintain the pH levels.

Digester	Sample	Amount of Urea	Inoculum after 30 days
Reactor 1	Green Jackfruit Leaves	1g Urea	2g Pro-biotic
Reactor 2	Dry Mango Leaves	1g Urea	5g Cow-dung
Reactor 3	Dry Teak Leaves	1g Urea	5g Cow-dung
Reactor 4	Dry Jackfruit Leaves	1g Urea	5g Cow-dung
Reactor 5	Green Mahgony Leaves	1g Urea	5g Cow-dung

3.7 Gas Analyzing

Gasboard Analyzer-3200 Plus was used to analyze the gas at certain intervals

3.7.1 Gas Analyzer Specification

For monitoring anaerobic digestions projects, Gasboard analyzer-3200 plus is widely used. There are 4 different sensors, which are CH₄, CO₂, H₂S, O₂.

Figure 8 Gas Board 3200 Plus Specification

Measuring gases	CH ₄ , CO ₂	NDIR
	O ₂ , H ₂ S	ECD
Measuring rang	CH ₄	0~100 %
	CO ₂	0~50 %
	O ₂	0~25 %
	H ₂ S	0~10000 ppm
Accuracy	CH ₄	± 2%FS
	CO ₂	± 2%FS
	O ₂	± 3.0%FS
	H ₂ S	± 3.0%FS
Repeatability	CH ₄ , CO ₂ , H ₂ S, O ₂	≤1.5%
Lithium battery pack	2200mAh	
Power supply	DC5V 2A	
Flow	(0.7-1.2) l/min	
Warm up time	90seconds once power on	
GPS sensor	Positioning and location	
Working temperature	(-10~40) ℃	
Ambient pressure	(700 ~ 1200) mbar	
Relative humidity	0~95% non-condensing water	
Dimension	276 × 195 × 66 mm (Length×width×height)	
Casing material	ABS/ Polypropylene and rubber molding	
Keyboard	Film panel keyboard	
Display	High-resolution colored 3.2-inch	
Communication	Micro USB port, bluetooth 4.0	

3.7.2 Data Collection

The analyzer comes with a companion software which allowed us to collect and store gas data. The data was recorded and stored using a laptop. It was analyzed and interpreted using Microsoft Excel. The analyzer's inlet port was linked to the setup's valve output. In between, air filters and scrubbers were utilized to separate the gases from contaminants. The toggle valve was gently opened, and the analyzer's pump was turned on, allowing the gas generated in the bottle to travel through the analyzer.



Figure 10 Measuring pH level with pH meter



Figure 9 Measuring Gas Percentage with Gas Analyzer

3.7.3 Verification of presence of methane

The methane production was tested by setting a flame at the gas outlet. Continuous flame was seen which confirms the presence of methane in the gas present in the digesters.

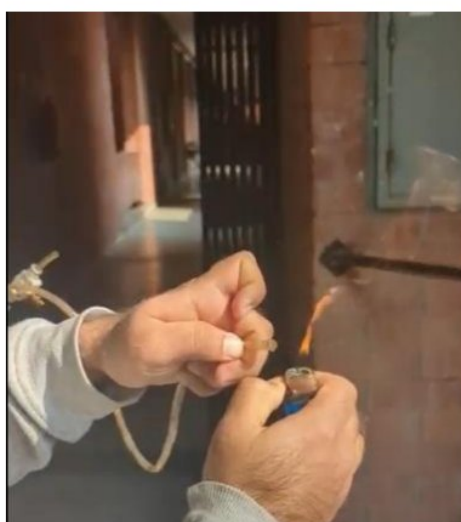


Figure 11 Flame test of produced methane

3.8 New Setups

The objective of the second phase of our experiment was to improve the efficiency of methane production by reducing the required time to achieve a methane concentration of 60%. Based on the preliminary findings, jackfruit leaves were selected as the primary substrate due to their favorable methane production characteristics and availability, as well as their high leaf litter rate.

To investigate the effects of different parameters on methane production, four new setups were designed. The first setup consisted of jackfruit leaves combined with cow dung, representing a co-digestion system. This choice was inspired by a previous study that demonstrated the enhanced biogas production achieved through anaerobic co-digestion of pre-treated press mud and molasses-based distillery wastewater.

Reactor	Sample	Preparation
Reactor 1	Dry Jackfruit Leaves	500 ml solution with 50 g leaves 5ml cow dung 2g yeast 2g urea
Reactor 2	Dry Jackfruit Leaves	500 ml solution with 50 g leaves 5ml cow dung 5g yeast 2g urea
Reactor 3	Dry Jackfruit Leaves	500 ml solution with 50 g leaves 5ml cow dung 5g molasses 2g urea
Reactor 4	Dry Jackfruit Leaves	500ml solution with 50 g leaves 5g Cow dung 2g Urea

Figure 12 New Setup Details

In the second setup, 2g of yeast was added to the substrate. This decision was influenced by research indicating the feasibility of co-digestion of surplus yeast and brewery wastewater, where no negative effects on digestion were observed. Moreover, the same study reported improved stability in the anaerobic digestion system with the addition of yeast, as evidenced by analysis of indicators such as volatile organic acids, alkalinity, and propionic acid. This setup aimed to explore the potential benefits of yeast addition on methane production efficiency.

In the third setup, 5g of yeast was added to the substrate, further examining the effects of yeast dosage on methane production. By comparing the methane yields of the setups with 2g and 5g of yeast, insights could be gained into the optimal dosage for maximizing biogas production.

Finally, molasses was included as an additive in the fourth setup. This decision was inspired by the study that demonstrated the positive impact of molasses-based distillery wastewater on biogas production during anaerobic co-digestion [1]. The addition of molasses aimed to assess its potential role in improving methane production efficiency.

Each setup was replicated multiple times to ensure the reliability and validity of the results. The experiment was conducted in anaerobic digesters maintained under controlled temperature and pH conditions. Methane production was monitored regularly using gas chromatography to assess the progress and efficiency of the anaerobic digestion process.

To quantify the methane concentration, samples were collected from the digesters at regular intervals and analyzed using established scientific methods. The methane production rate, time required to reach 60% methane concentration, and any observed variations or abnormalities in the biogas composition were recorded and analyzed.

In summary, the methodology involved the selection of jackfruit leaves as the primary substrate, followed by the creation of four distinct setups, each exploring the effects of different parameters on methane production efficiency. The decision to include yeast and molasses was based on previous studies highlighting their potential benefits in anaerobic digestion processes. The experimental setups were replicated to ensure robustness, and methane production was monitored and analyzed using established scientific techniques.

Chapter 4 Result and Discussion

4.1 Setup 1: Gas Analyzing Results

From graph 1 Gas percentage and pH vs Days were plotted where gas percentage was in the primary axis and pH was in the secondary axis. No methane was produced from this setup. The maximum amount of CO₂ in this setup 22.59% and maximum O₂ is 2.95%

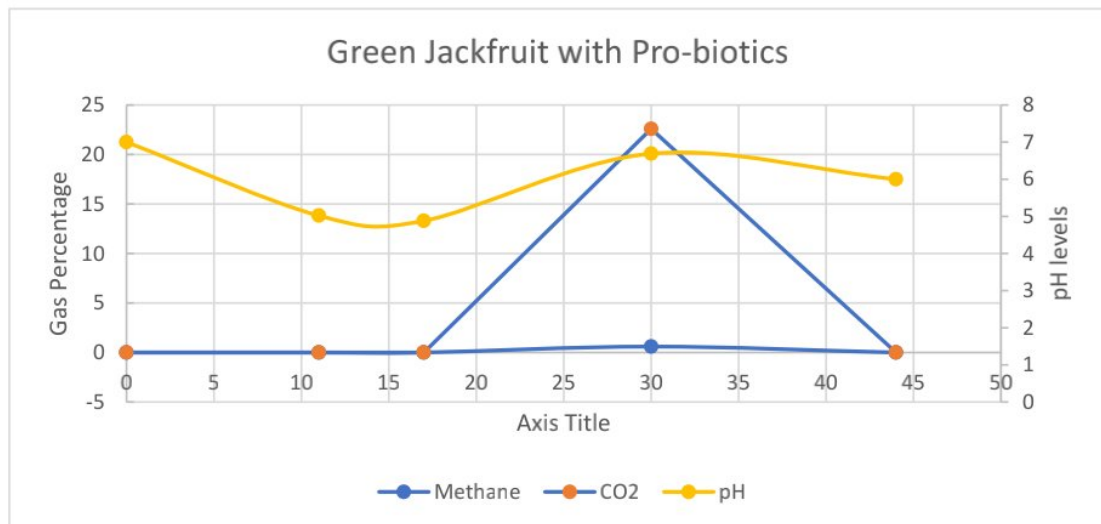


Figure 13 Green Jackfruit Gas Percentage and pH

4.2 Setup 2: Gas Analyzing Results

This setup contained Dry Mango leaves and cow dung, and co-digestion was observed. The production of methane was observed but the percentage was really low, at a maximum of 7.48%. The pH was observed to be in a optimum range.

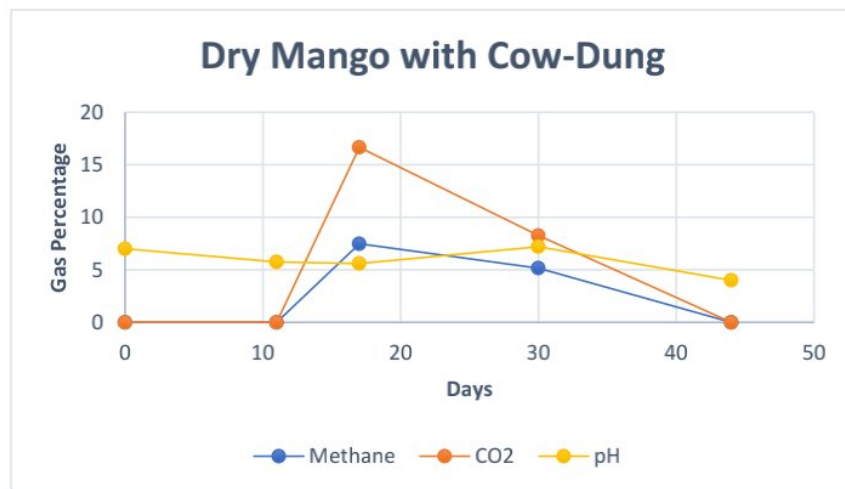


Figure 14 Dry Mango Gas Percentage and pH

4.3 Setup 3 Gas Analyzing Result

This setup contained Dry Teak leaves and cow dung, and co-digestion was observed. The desired amount of methane production was observed from this setup. The methane percentage was 64.3%

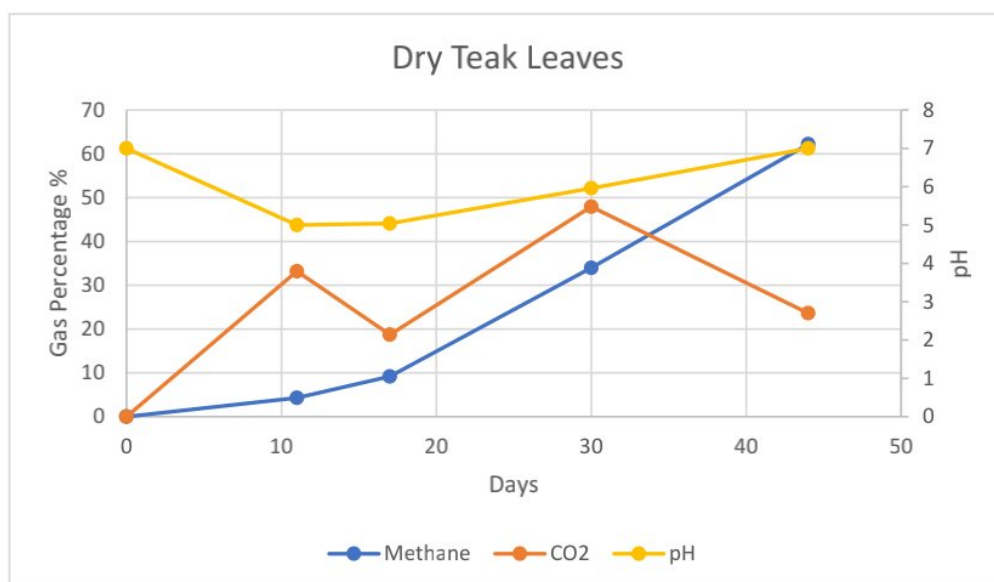


Figure 15 Dry Teak Leaves Gas percentage and pH

(>60%) which is enough for burning as fuel. Also, the pH levels were between 5-7, which is optimum for methane production

4.4 Setup 4 Gas Analyzing Result

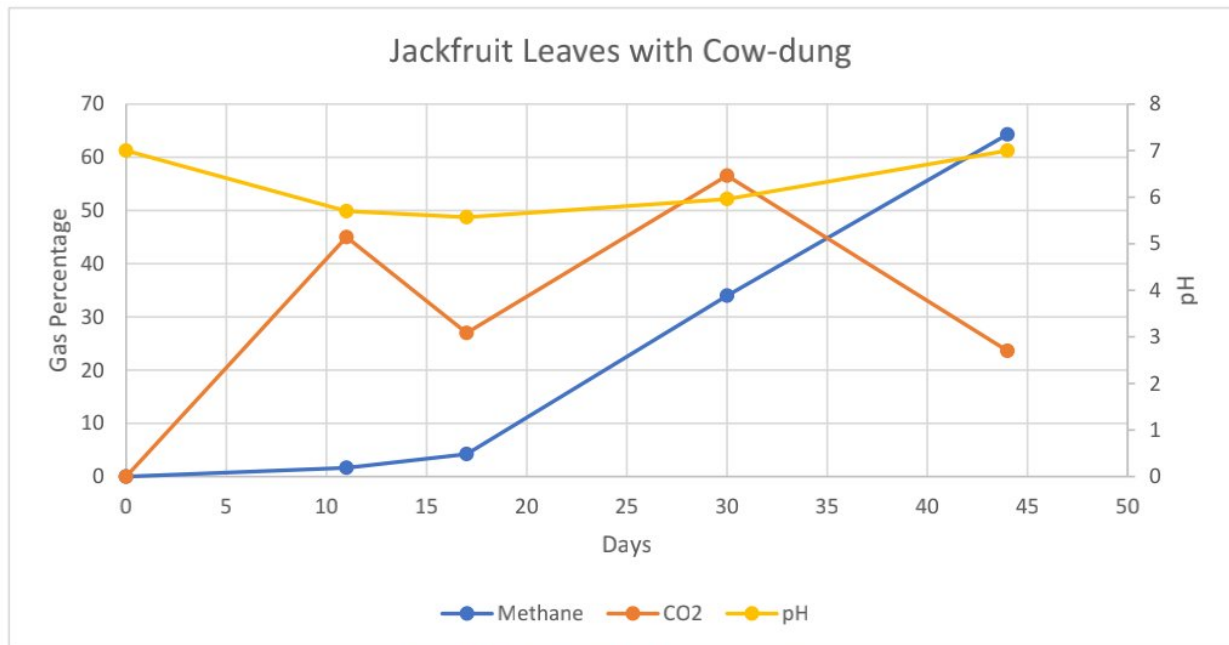


Figure 16 Dry Jackfruit Gas Percentage and pH

Co-digestion was seen in this experiment, which included Dry Jackfruit leaves and cow manure. This configuration produced the necessary amount of methane. The methane content was 62.2% (>60%), which is sufficient for use as fuel. Furthermore, the pH levels were between 5-7, which is ideal for methane synthesis.

4.5 Setup 5 Gas Analyzing Result

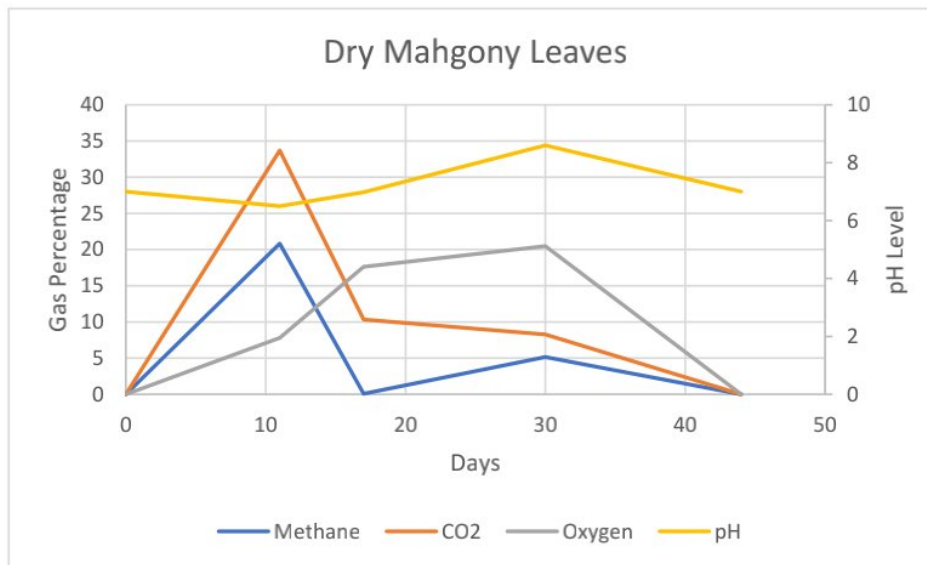


Figure 17 Dry mahgony leaves gas analysis and pH

This experiment, which contained Dry Mahgony leaves and cow manure, revealed co-digestion. This arrangement generated methane but not desired amount. The methane level was 20.83% (<60%), which is not adequate for burning as fuel. Although, the pH levels were optimal for methane production, ranging from 5-7.

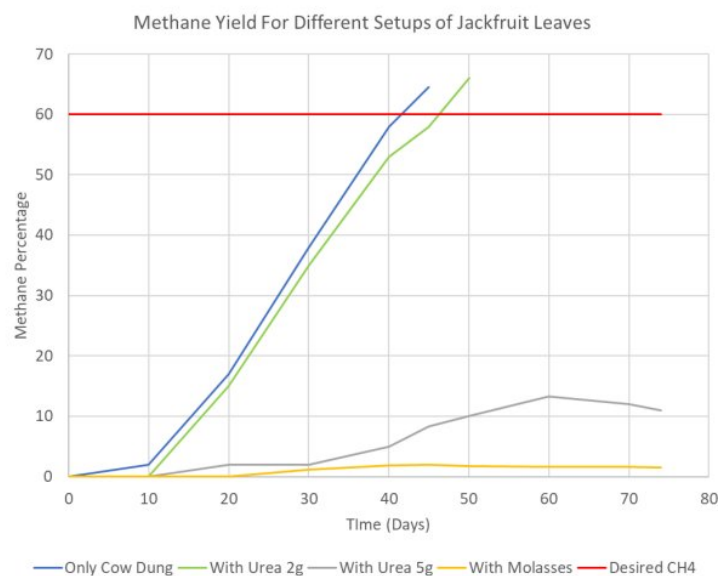
4.6 Phase 2 of experiment

The objective of the second phase of our experiment was to enhance the efficiency of methane production by reducing the time required to achieve a methane concentration of 60%. However, our findings indicate that none of the tested setups yielded favorable results in terms of both methane production and time efficiency.

Among the setups tested, the one employing 2g of yeast demonstrated the highest methane concentration, surpassing the targeted 60%. While this outcome is promising in terms of methane production, it came at the expense of an increased time requirement compared to the initial setup. This observation suggests a potential trade-off between methane yield and production time, indicating the need for further investigation into optimizing these variables simultaneously.

Interestingly, the setups utilizing 5g of yeast and molasses exhibited significantly low levels of methane production. This unexpected outcome raises questions about the influence of yeast and molasses quantities on the overall efficiency of methane production. Further exploration is warranted to understand the underlying mechanisms that led to this reduced methane output and to identify potential factors inhibiting the desired microbial activity.

These findings underscore the complexity of optimizing methane production and highlight the challenges associated with achieving both high methane concentrations and reduced production time. It is evident that a careful balance between yeast quantity, molasses addition, and fermentation time needs to be struck in order to achieve optimal results.



To overcome the limitations encountered in this phase of the experiment, future studies could explore alternative strategies, such as modifying the yeast strain or adjusting the nutrient composition of the medium. Additionally, analyzing the microbial dynamics and metabolic pathways involved in methane production could provide valuable insights for process optimization.

In conclusion, our second phase of experiments aimed at improving the time efficiency of methane production did not yield favorable results. While the setup using 2g of yeast demonstrated increased methane production, it also incurred a longer time requirement. Conversely, the setups with 5g of yeast and molasses exhibited unexpectedly low methane production. These findings emphasize the need for further research to understand the underlying factors influencing methane production and to develop optimized strategies that can balance both methane yield and production time.

4.7 Effect of yeast and molasses

The second phase of our experiment aimed to improve the time efficiency of methane production, focusing on the effects of yeast and molasses addition. However, the observed outcomes did not align with our expectations, prompting further analysis and discussion.

The influence of organic load (OL) on biogas production and composition is well-documented [51]. Higher OL has been associated with increased production of various bioproducts, although their respective yields tend to be higher at lower OL levels. In our study, the addition of yeast and molasses likely contributed to an increased OL, which may have influenced the methane yield. The higher organic load could have led to alterations in the microbial community dynamics and metabolic pathways, potentially impacting methane production efficiency.

Another factor that might have affected methane production is the inhibition of aceticlastic microorganisms. Previous research on anaerobic digestion of baker's yeast molasses wastewater revealed that high pH and increased acetic acid concentration inhibited aceticlastic microorganisms crucial for methane production [52]. It is plausible that a similar inhibition occurred in our experimental setups due to the addition of yeast and molasses, leading to suboptimal methane production levels.

Furthermore, the imbalance in microbial reaction rates within the anaerobic digestion process is critical for optimal biogas production. The mechanism of syntrophy, which relies on the cooperation between different microbial species, plays a vital role in maintaining this balance [53]. Factors such as organic acid concentration, pH, temperature, and organic loading rates influence this microbial synergy. The introduction of yeast and molasses in our setups may have disrupted the delicate equilibrium of microbial reaction rates, resulting in unfavorable outcomes.

To address the limitations encountered in our experiment, future studies could explore potential strategies to mitigate the inhibitory effects observed. For instance, adjusting the pH levels, optimizing the organic acid concentrations, or modifying the organic loading rates could help restore the balance of microbial reactions and enhance methane production efficiency.

Moreover, a comprehensive analysis of the microbial community dynamics and metabolic pathways involved in the anaerobic digestion process would provide valuable insights for process optimization. By understanding the complex interactions and dependencies between different microorganisms, it may be possible to develop targeted interventions to improve methane production and mitigate inhibitory effects caused by the addition of yeast and molasses.

In conclusion, our findings highlight the potential influence of the organic load, inhibition of aceticlastic microorganisms, and imbalances in microbial reaction rates on the efficiency of methane production when yeast and molasses are introduced. The observed unfavorable results underscore the complexity of optimizing the anaerobic digestion process. Future studies should focus on strategies to mitigate inhibition and restore microbial balance, as well as deepen our understanding of the microbial dynamics involved. Such efforts are crucial for advancing methane production techniques and promoting the development of sustainable energy systems.

Chapter 5 Conclusion and Future Recommendation

5.1 Conclusion

5.1.1 Findings

In conclusion, our study has provided valuable insights into the factors influencing methane production in anaerobic digestion processes. The key findings of our investigation shed light on the role of pH, substrate selection, and the effects of yeast and molasses addition on biogas yields.

Firstly, our research confirms the critical role of pH in anaerobic digestion. pH levels have a significant impact on microbial activity and metabolic pathways involved in methane production. It is crucial to maintain an optimal pH range to promote favorable conditions for methanogenic microorganisms and maximize biogas yields.

Secondly, our study highlights that not all substrates yield the same amount of methane. This finding emphasizes the importance of carefully selecting suitable substrates for anaerobic digestion to enhance biogas production. Factors such as substrate composition, nutrient content, and degradation rates should be considered to optimize the overall efficiency of the process.

Furthermore, our investigation revealed unexpected outcomes regarding the addition of yeast and molasses. Contrary to our initial expectations, their inclusion did not result in the desired enhancement of biogas production. This unexpected finding indicates the complexity of the interactions between added components and the microbial community in anaerobic digestion systems. Further research is necessary to elucidate the underlying mechanisms and identify potential strategies to optimize the utilization of yeast and molasses for methane production.

Overall, our study contributes to the understanding of factors influencing methane production in anaerobic digestion and highlights the need for careful consideration of pH, substrate selection, and the effects of additives. These findings have practical implications for the design and operation of biogas production systems, aiming to maximize methane yields and promote the development of sustainable energy generation.

5.2 Recommendations

Based on the findings of this study, further research and exploration are warranted to optimize the methane production process, increase methane yields, and reduce the methane generation time. The following recommendations outline potential avenues for future work:

Study the effects of yeast on methane production: Investigating the influence of yeast on methane generation in anaerobic digestion systems is essential. This research could involve studying different yeast strains, dosage levels, and fermentation conditions to identify optimal parameters that maximize methane production. Additionally, exploring the interactions between yeast and other microorganisms within the microbial community could provide valuable insights into improving biogas yields.

Investigate the effects of sample size on methane generation: Understanding the impact of sample size on methane production is crucial for process optimization. Conducting experiments with varying sample sizes can help determine the relationship between sample size and methane yields. This research could provide insights into the scalability of the anaerobic digestion process and guide the design and operation of larger-scale biogas production systems.

Experiment with co-digestion using cafeteria waste: Exploring co-digestion strategies by incorporating cafeteria waste as an additional substrate can enhance methane production and improve process efficiency. Investigating the synergistic effects of different feedstock combinations, including dry jackfruit, teak leaves, and cafeteria waste, could lead to improved methane yields and a more sustainable utilization of organic waste resources.

Optimize the utilization of additives: To unlock the full potential of additives such as yeast and molasses, further exploration is necessary. Research efforts should focus on identifying optimal dosage levels, application methods, and potential alternative additives that can enhance biogas production. Understanding the mechanisms behind the interactions between additives, microorganisms, and the anaerobic digestion process is crucial for their effective utilization.

Conduct comprehensive life cycle assessments and economic analyses: In addition to technical optimization, conducting comprehensive life cycle assessments and economic analyses is vital to evaluate the environmental impacts and economic feasibility of biogas production systems. Assessing the entire life cycle, from feedstock cultivation to waste management, will provide a holistic understanding of the environmental benefits and potential economic viability of biogas production. These assessments can guide decision-making processes and help prioritize sustainable and economically viable biogas projects.

In conclusion, future research endeavors should focus on optimizing the methane production process by investigating the effects of yeast, sample size, and co-digestion with cafeteria waste. Exploring different dosage levels, application methods, or alternative additives can also enhance biogas production. Additionally, conducting comprehensive life cycle assessments and economic analyses will provide valuable insights into the environmental and economic feasibility of biogas production systems. By addressing these research gaps, we can advance the field of anaerobic digestion and contribute to the development of sustainable energy solutions.

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