# PROJECT REPORT

TITLE OF THE REPORT: BIO-METHANATION

POTENTIAL ANALYSIS

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# **Bio-methanation Potential of Various Substrates**

#### Introduction

The process of anaerobic digestion, otherwise known as biomethanation, has been the most promoted environmentally friendly and renewable source of technology for recovering energy from organic wastes. It is a biologically operated process that decomposes organic matter in the absence of oxygen by bacteria involved in this process in order to produce biogas that contains primarily methane (CH<sub>4</sub>) and carbon dioxide (CO<sub>2</sub>). Methane content in the biogas is an important renewable energy which may be utilized for electricity generation, heating purposes, and even for transport. It provides a route towards reduction in dependence on fossil fuels. The present study is aimed at assessing the potential for biomethanation of various organic substrates with emphasis on the comparison of yields over a 30-day period. The substrates chosen include Micro Cell, Animal Feed, Wheat Straw, Sunflower Seed, and Inoculum. These cut across the categories of organic materials readily available in either an agricultural setting or an industrial setting. Their methane generation potential is an area that requires much understanding, for it would influence the optimization of anaerobic digestion processes and feedstock selection for biogas plants.

In the pressure gauge manometric method, this study will use gas from each substrate to estimate cumulative methane produced. The volume of methane produced can be measured and converted to the pressure that gas produces in a closed system. It is precise with the advantage of measuring production of gas continuously over time, and thus, it brings about insight into kinetics during the biomethanation process for each substrate. Methane yield of a substrate would directly indicate the efficiency with which the substrate produces biogas, which means that the higher the methane yield, the greater the fraction of organic matter existing in the substrate that is being converted to biogas, hence, a potential for greater energy recovery. The cumulative methane yield of the five substrates under study will be compared and checked for best performance in methane generation. Micro Cell It contains high TS and VS; thus, it may well be regarded as a potentially high-energy feedstock. However, the actual methane yield is affected by the extent of biodegradability of organic matter present in it.

Such feed being mainly rich in nutrients and organic content, significant methane production is expected from animal feed; again, such feed could well be regarded as the feedstock that contains unbiodegradable material, which may affect the overall methane yield.

Wheat straw, as an agro-product, is readily available in huge quantities at low cost. Hence, this appears to be an interesting substrate for biomethanation. Methane from wheat straw is dependent upon the breakdown of the lignocellulosic material, which is bothersome unless subjected to some pre-treatment. This is yet another agricultural byproduct sunflower seed waste, which is much richer in organic content and might hold great potential for significant yields of methane due to easily degradable components in it. This is the control or baseline in the experiment. Inoculum carries the microbial population which will be required to operate the anaerobic digestion but will contribute only minimally to the methane production as it has low organic contents. The result of this study is expected to provide a critical comparison of the cumulative methane yields of these substrates, thus providing insights into their relative efficiencies for biogas production. The study would identify substrates that yield the highest possible amount of methane, thereby contributing to optimizing feedstock selection for anaerobic digestion processes. Optimizing such feedstock selection may be expected to decrease operational costs for biogas plants while raising energy yields and help scale up biomethanation as a renewable source of energy. Possibly, pre-treatment of substrates like Wheat Straw and Micro Cell may be necessary if the latter contains such complex organic compounds like lignin resistant to microbial degradation. Pre-treatment in the form of mechanical, thermal, or chemical treatment can break down such complex organic compounds with increased bioavailability of the organic matter eventually leading towards an increase in the yield of methane.

Bio-methanation not only provides a means of recovering energy but also presents solutions to the problem of waste. Untreated organic wastes result in decomposition, and at its largest concentration, it is methane, which has a higher global warming potential than carbon dioxide. Methane capture and utilization in controlled anaerobic digestion systems aid in abating the emission of this potent gas resulting from waste. In addition, the digestate that is produced as a byproduct of the biomethanation process can be utilized as a biofertilizer, which closes the loops in agricultural waste management. In a circular economic sense, utilizing waste converts it into valuable resources without contributing much to environmental degradation and ensures resource efficiency.

#### **Materials and Methodology**

The Water Displacement Method simply represents the easy, accurate method for the evaluation of Biomethane Potential of organic substrates. This method is the capture by displacement of volume through water during the production of gas within an anaerobic digestion process. The amount of water displaced is equal in volume to that quantity of gas produced; consequently, it is directly proportional with biogas production within time. The experimental procedure for the estimation of BMP through the water displacement method is illustrated below in the steps:

## **Materials and Equipment**

## **Anaerobic Digestion System Components:**

Batch reactors: Glass bottles or serum bottles fitted with tight caps Substrates like organic waste materials such as food waste, agricultural residues, etc. Inoculum for the microbial degradation process such as anaerobic sludge or digestate. Nutrient medium if required to support microbial activity.

## **Gas Collection Apparatus:**

Water displacement setup consisting of an inverted graduated cylinder or gas collection tube that is filled up with water. silicone tubing to connect the gas outlet coming out from the reactor to the gas collection cylinder. Water bath or tray to support the water displacement system.

**Measurement Tools:** Pressure gauges if available to record the pressure. Graduated cylinders for recording the volume of the gases Thermometer to monitor the temperature in reactors and water bath. pH meter or pH strips for pH monitoring in reactors.

# **Incubator or Temperature Control Unit:**

To maintain the digestion temperature at either mesophilic conditions  $35^{\circ}\text{C} \pm 2^{\circ}\text{C}$ , or thermophilic conditions,  $55^{\circ}\text{C} \pm 2^{\circ}\text{C}$ , depending on experimental set up.

# **Experimental Procedure**

#### 1. Preparation of Reactors

Substrate loading: Organic substrates like food waste and agricultural residues are weighed and prepared in an appropriate manner to obtain their total solids content and volatile solids content. The load applied from one reactor to another aims at standardization. The amount of substrate added is calculated based on desired organic load, usually between 1 to 3 g VS per liter of reactor volume.

Inoculum Addition: Anaerobic inoculum typically coming from a functional anaerobic digester was added to each reactor. The inoculum-to-substrate ratio was kept at about 2:1 (VS basis) to allow for adequate numbers of microbes to carry on anaerobic digestion.

Seal the Reactors: Each reactor was sealed with an airtight rubber stopper or cap to prevent oxygen from entering the system. Reactors were next flushed with nitrogen gas to establish anaerobic conditions.

# 2. Water Displacement System Preparation

Gas Collection System: The reactors are connected to the water displacement system by gas-tight tubing. One end of the tubing goes into the reactor's gas outlet and the other end into an inverted graduated cylinder that contains water.

Displacement Apparatus: The inverted graduated cylinder is submerged in a water bath so that all the biogas produced is captured. As soon as the biogas is produced in the reactor, it comes through the tubing and pushes out the column of water in the cylinder. That is, the volume of the water displaced will be equal to the volume of biogas produced.

## 3. Incubation and Monitoring

Incubation: The reactors are incubated or water bathed in a water bath set and maintained at a constant temperature around 35°C for mesophilic digestion. The incubation of the reactors may take about 30 to 60 days depending on the rate of degradation of the substrates.

Biogas Volumes: The total produced biogas is recorded daily or at intervals by measuring water displaced in a graduated cylinder. Errors are avoided, and adequate time is given for the equilibration of all dissolved gases in the water.

## 4. Compositional Analysis of Biogas (Optional)

Methane Content: If this is crucial, one can measure methane content from the biogas composition using GC. This would be important where the BMP has a particular emphasis on methane yield. This may be carried out by taking a gas sample from the headspace of the graduated cylinder and then analyzing for methane, carbon dioxide, among other gases.

Correction to Standard Temperature and Pressure (STP): The water volume displaced by biogas as measured corrected to standard temperature and pressure so that the reported volumes of gas are consistent.

#### 5. Calculation of Biomethane Potential

Cumulative Biogas and Methane Yield. The total volume of biogas produced during an experimental period is measured. When gas composition data are available, the volume of methane may be calculated from the proportion of methane in biogas. The cumulative yield of methane is then expressed as liters of CH<sub>4</sub> produced per kilogram of volatile solids added L CH<sub>4</sub>/kg VS added.

Correction for Gas Solubility: The dissolved gas in the liquid phase of the reactor has to be accounted for using Henry's law, especially in long-term experiments.

Plotting Cumulative Methane Yield vs. Time: The kinetics of methane production can be understood based on the types of the substrate used. A comparison can be made of the results for different substrates to compare their biomethane potential.

## 6. Control and Replicates

Control Reactors: An empty control reactor is filled with inoculum but no substrate for methanogenesis. This allows the results obtained for methanogenesis in the experimental reactors to be corrected.

Replicates: All experiments are done in triplicate. The data obtained from three replicates are averaged, and standard deviations are calculated to quantify the variability.

# **Advantages of Water Displacement Method**

Ease and Economy: The method of water displacement is very easy to construct, and does not demand any complicated or very expensive equipment.

**Direct Measurement:** In the method of water displacement, direct measurement of gas volumes is possible. Thus, there are no pesky calculations or conversion needed to get reliable data.

**Real-Time Monitoring:** Gas production can be monitored real-time over the course of the experiment allowing for time-based kinetic analysis.

#### Limitations

**Gas Saturation:** Measured values may be less accurate, as dissolved gases evaporate over time, if the displacement method is used without properly accounting for saturating gases.

**Gas Content:** While the displacement method can provide an overall measure of biogas production, direct determination of methane content requires additional instrumentation, such as gas chromatography.

**Corrections in Temperature and Pressure:** The volume of biogas should be corrected at standard conditions for better reporting of results. This requires careful observation of the ambient conditions in the experiment setup.

#### **Results and Discussion**

The results of the cumulative methane yield for the substrates—Micro Cell, Animal Feed, Wheat Straw, Sunflower Seed, and Inoculum—over a period of 30 days provide significant insights into the potential of these materials for biomethanation. The data from the experiments, summarized in the graph and table, offer a comparative analysis of methane

production efficiency, with total solids (TS) and volatile solids (VS) percentages playing an essential role in determining the effectiveness of each substrate.

#### 1. Cumulative Methane Yield

The cumulative methane yield is measured in litres of methane produced per kg of volatile solids added (L CH4/kg VS added) over a period of 30 days. The graph illustrates the distinct methane production profiles of each substrate:

- **Sunflower Seed** emerged as the most promising substrate, producing the highest cumulative methane yield of approximately 370 L CH4/kg VS added by the 30th day.
- Wheat Straw followed with a cumulative yield of around 325 L CH4/kg VS added.
- **Animal Feed** displayed moderate methane production, reaching approximately 275 L CH4/kg VS added by day 30.
- **Micro Cell** exhibited a lower methane yield of around 210 L CH4/kg VS added by the end of the observation period.
- **Inoculum**, used as a control, showed negligible methane production (close to zero) throughout the experiment, as expected due to its low VS content.

# 2. Analysis Based on Total Solids (TS) and Volatile Solids (VS)

The total solids (TS) and volatile solids (VS) content of each substrate are key factors in determining their biomethanation potential. The table below shows the TS and VS values for each substrate:

Substrate	TS (%)	VS (%)
Micro Cell	97.00	93.17
Animal Feed	89.70	81.11
Wheat Straw	91.17	86.49
Sunflower Seed	92.41	89.56
Inoculum	2.81	1.89

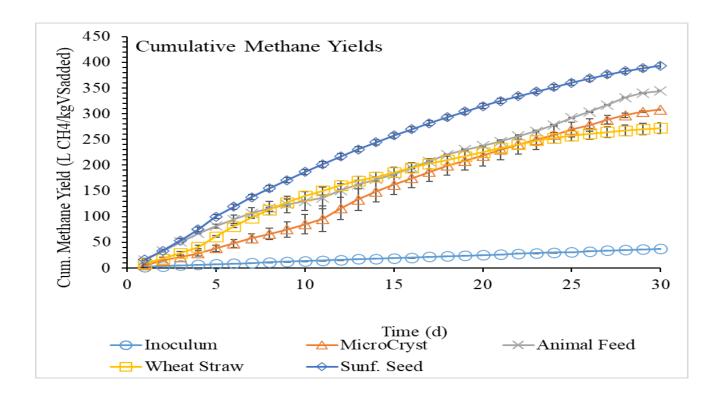
- Sunflower Seed (TS = 92.41%, VS = 89.56%) produced the highest methane yield, likely due to its high volatile solids content, which contributed to enhanced microbial activity and biogas production. The nearly 90% VS content means that most of the substrate's solid content was degradable organic matter, leading to the higher cumulative methane yield.
- Wheat Straw (TS = 91.17%, VS = 86.49%) also showed strong methane production. The slight reduction in VS content compared to Sunflower Seed explains its slightly lower methane yield. However, Wheat Straw remains a viable candidate for methane production due to its high organic matter content.
- **Animal Feed** (TS = 89.70%, VS = 81.11%) yielded lower methane compared to Sunflower Seed and Wheat Straw. The lower VS content in Animal Feed contributed to the reduced biogas production, as the substrate contained more non-degradable material, which limited its potential for conversion to methane.
- Micro Cell (TS = 97.00%, VS = 93.17%) was expected to perform better due to its high volatile solids content, but its cumulative methane yield was lower compared to other substrates. This discrepancy might be attributed to the nature of the organic matter in Micro Cell, which could be less bioavailable for microbial degradation, thus slowing down methane production.
- **Inoculum** (TS = 2.81%, VS = 1.89%) acted as a control in the experiment. As it contained minimal organic matter, its methane production was negligible. This result aligns with expectations, as the inoculum was primarily used to initiate the anaerobic digestion process and did not contribute significantly to methane production.

# 3. Temporal Methane Production Patterns

The temporal patterns of methane production for each substrate reveal insights into the kinetics of biomethanation:

- Sunflower Seed and Wheat Straw both exhibited rapid methane production during the first 10 to 15 days, followed by a gradual leveling off towards day 30. This behaviour suggests that the readily biodegradable fractions of these substrates were exhausted early in the process, and the remaining organic matter took longer to break down.
- Animal Feed showed a slower initial rate of methane production compared to Sunflower Seed and Wheat Straw. However, its methane yield continued to increase steadily throughout the 30-day period, indicating a more prolonged degradation process.

- Micro Cell displayed the slowest methane production rate among the organic substrates. This could indicate that the complex structure of Micro Cell limited microbial access to the organic matter, resulting in slower breakdown and methane generation.
- **Inoculum** remained relatively flat over the entire 30-day period, with no significant methane production, as expected.



**Significance of graph:** the cumulative methane yield versus time graph illustrates the total amount of methane produced over a given period. Initially, methane production begins slowly as microbial communities acclimate to the substrate. Then, it enters a rapid increase phase, indicating the peak of microbial activity and substrate digestion. Over time, the slope of the graph plateaus as the readily degradable organic matter becomes limited, leading to reduced methane production. The final plateau represents the maximum methane yield, showing that most available substrate has been converted to biogas. This graph helps in assessing the biodegradability and efficiency of substrates for biogas production.

#### 4. Discussion on Substrate Selection

The data clearly indicate that **Sunflower Seed** and **Wheat Straw** are the most effective substrates for biogas production in this experiment. The high VS content in both substrates provides ample degradable organic matter, leading to superior methane yields. The rapid production of methane in the early stages of the experiment suggests that these substrates contain easily degradable organic compounds, making them ideal for quick biogas generation.

On the other hand, **Animal Feed** and **Micro Cell**, while still viable, exhibited lower yields, likely due to either a lower VS content (in the case of Animal Feed) or the complex composition of the substrate (in the case of Micro Cell), which hindered microbial degradation. Future studies could explore pre-treatment options to enhance the bioavailability of these substrates, potentially increasing their methane yield.

#### Conclusion

The study indicates that Sunflower Seed and Wheat Straw are promising substrates for biomethane production. The higher volatile solids content in these substrates contributed to their superior methane yields. Micro Cell and Animal Feed also showed significant potential, although their yields were slightly lower. Inoculum, as expected, did not contribute substantially to methane production. These results highlight the importance of selecting the right substrates based on their volatile solids content for optimizing biomethane production.