



Experimental and Numerical Optimization of Biogas Energy Production Using the RSM Method

Othmane Maakoul^{*✉}, Abdellah Boulal²

Laboratory IMII, Faculty of Sciences and Technology, University Hassan First, 26000 Settati, Morocco

* Correspondence: Othmane Maakoul (othmane.1992.mkl@gmail.com)

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Abstract: This study aims to optimize biogas production at the laboratory scale using a batch-mode bioreactor and Response Surface Methodology (RSM). The main objective is to assess the effects of key parameters substrate composition (household waste and cow manure), pH, fermentation temperature, and agitation speed on biogas yield. A series of experiments were designed using a central composite RSM to evaluate the influence of substrate composition and temperature. The experimental data were analyzed through ANOVA to assess model significance and accuracy. The results show that the developed quadratic models are statistically significant, with a determination coefficient (R^2) of 0.90 for cumulative biogas production. These findings confirm the adequacy of the models and the effectiveness of RSM in identifying optimal operating conditions for enhanced biogas yield.

Keywords: Biogas production; Batch bioreactor; Response Surface Methodology; Substrate composition; pH; Agitation speed; ANOVA; Model optimization

1 Introduction

The growing energy demand across various sectors is accelerating the depletion of fossil fuels and contributing significantly to global warming. This alarming trend has intensified global efforts toward the development of renewable energy sources [1]. Simultaneously, both household and industrial activities generate substantial volumes of biodegradable solid waste, highlighting the urgent need for effective management and sustainable valorization methods [2]. Among the most promising waste-to-energy strategies is anaerobic digestion, a biological process wherein microorganisms break down organic matter in the absence of oxygen, generating a biogas [3].

Biogas, composed primarily of methane and carbon dioxide, is recognized as a clean, renewable energy source that provides both environmental and economic benefits [4]. This process typically operates under mesophilic (30–45°C) or thermophilic (50–60°C) conditions [5].

Research indicates that thermophilic digestion (around 55°C) offers higher biodegradation efficiency [6], although it requires additional thermal input to maintain operating temperatures. Several factors including substrate composition, pH, dry matter content, and mixing regime significantly affect the biogas production process [7, 8].

While many studies have analyzed these parameters individually, more recent work emphasizes the importance of their interactions [9, 10]. In particular, co-digestion using mixed substrates has shown improved nutrient balance, enhanced microbial activity, and better biogas yields with reduced inhibition risks [11]. Therefore, optimizing these interrelated variables is critical for stable and efficient biogas production. However, conventional trial-and-error methods are often time-consuming, resource-intensive, and unsuitable for capturing complex interactions [12].

To address this, researchers increasingly employ computational and statistical tools to optimize processes and reduce experimental costs [13, 14]. Among them, Response Surface Methodology (RSM) has proven particularly useful. RSM enables the systematic evaluation of multiple variables and their interactions, minimizes the number of experiments required, and facilitates the identification of optimal operating conditions through mathematical modeling [15, 16].

Despite extensive global research on anaerobic digestion, there remains a noticeable gap in region-specific studies, particularly in Southeast Asia and Indonesia. Local conditions such as tropical climate, unique waste composition,

and decentralized rural energy needs warrant targeted investigations. Few studies to date have explored the combined application of experimental and modeling approaches, such as RSM, in optimizing small-scale biogas systems under these specific regional constraints.

This study aims to bridge this gap by evaluating the effects of critical operating parameters (substrate composition, temperature, agitation speed) on biogas production under conditions relevant to the Indonesian context. Using RSM, we seek to develop a predictive model for process optimization, which will support the design of efficient, low-cost biogas systems suited to local needs.

2 Method and Materials

2.1 Experimental Optimization of Biogas Production at the Laboratory Scale

The literature review conducted highlighted several gaps in understanding the functioning of biogas production units [17–19]. This paper aims to study, model, experiment, and optimize the biogas production process from organic waste. The influences of several parameters are examined, such as the physicochemical characteristics of the organic waste to be fermented, operating temperature, pH value, agitation speed, and useful volume. Mastering these operating parameters is crucial for optimizing a large-scale pilot system [20, 21].

This work focuses on the optimization of biogas production at the laboratory scale, aiming to improve the efficiency and effectiveness of the process. The objective is to explore different methods, variables, and experimental setups that can influence the yield of biogas when using organic waste. By conducting controlled laboratory experiments, we aim to determine the key factors that maximize the production of biogas and evaluate how different parameters such as substrate composition, temperature, pH, agitation speed, and reactor volume affect the biogas yield.

The optimization process will involve both experimental and numerical approaches to improve the process parameters. The use of advanced methods such as RSM will help in determining the best combination of variables to optimize biogas production. This section provides insights into the techniques used to study, model, and experiment with biogas production, as well as the tools and equipment required for such experiments.

2.1.1 Influence of the usable volume and operating temperature

Numerous studies have demonstrated that increasing temperature positively influences specific stages of anaerobic digestion. Within an appropriate temperature range, it can notably accelerate the digestion process. The fermented substrate needs to be heated to reach the optimal fermentation temperature, and in general, higher temperatures lead to faster reaction rates [22, 23].

However, it is important to note that excessively high temperatures may disrupt the stability of organic matter and negatively affect its functionality. Indeed, too high a temperature can lead to the destruction of the bacteria necessary for the process; while too low a temperature will slow down the degradation of organic matter.

Influence of the usable volume of bioreactors:

Our goal through this experimental trial is to study the influence of the usable volume of bioreactors on the amount of biogas produced as shown in Figure 1. Experimental trials were conducted to analyze the impact of the usable volume of the bioreactor on biogas production. Five bioreactors operating in batch mode were used for these experiments, in order to analyze the biogas production performance based on different working volumes of the bioreactors.



Figure 1. Experimental setup for biogas production

The tested usable volumes, expressed as a percentage of the total volume of the bioreactors, are: 50%, 60%, 70%, 80%, and 90%. This approach allows the evaluation of the impact of varying the usable volume on the efficiency of biogas production and identifies the optimal conditions to maximize the performance of the anaerobic digestion process. At the exit of the five Erlenmeyer, storage balloons are connected to collect the produced biogas, which is then, weighed using precision balances to determine the amount of biogas in grams.

Influence of temperature:

To study the influence of temperature on biogas production, a thermal regulation system was implemented to ensure precise temperature control inside the bioreactors. This system allows the bioreactors to be maintained under optimal conditions for both mesophilic and thermophilic regimes, with temperatures varying between 25°C and 45°C for the mesophilic regime, and between 45°C and 60°C for the thermophilic regime. For this study, eight Erlenmeyers were used as bioreactors operating in discontinuous mode. Each Erlenmeyer was subjected to a specific temperature, adjusted and regulated through a thermostatic bath.

The temperatures of each bioreactor were measured using thermocouples, carefully immersed in the fermentation medium to ensure precise and continuous readings. These thermocouples were connected to a control and data acquisition system via a DSP card, allowing for real-time collection and recording of temperature variations.

2.1.2 Influence of pH value on biogas production

The pH is a key factor in regulating the growth and activity of bacteria, but excessive variations in pH can inhibit their activity, affecting the yield of anaerobic digestion [24]. The main objective of this study is to determine the optimal pH value that maximizes biogas production, while ensuring a favorable environment for bacterial growth, and to better understand the interdependence between acid-base conditions and the biological processes of anaerobic digestion.

An experimental trial as shown in Figure 2, was designed using six 1000 mL Erlenmeyers, serving as bioreactors operating in batch mode. These bioreactors were fed with a homogeneous substrate, and the experimental conditions were controlled to observe the impact of pH on anaerobic fermentation and biogas yield. The pH of the reaction medium was adjusted to values ranging from 2 to 10, covering a wide range of acidic, neutral, and alkaline conditions. Regular measurements are taken to maintain and adjust the pH to specific levels, in order to identify the optimal range for biogas production. The amount of gas produced was measured for each pH value to study the effect of this parameter on fermentation yield.



Figure 2. Experimental setup for pH monitoring

2.1.3 Influence of stirring on methanization

Proper stirring helps prevent the formation of crusts and the settling of dense particles, which aids in breaking the floating layer and facilitates the escape of biogas [25]. It thus promotes thermal and metabolic transfers during chemical reactions. Insufficient stirring may limit the contact between the substrate and methanogenic bacteria, while excessive stirring could lead to gas loss or stress for the microorganisms. In order to study the impact of stirring on biogas production, experimental systems using 1000 mL Erlenmeyer were set up, exposed to ambient temperature, and operated in batch mode.

The homogenization of the reaction medium is ensured by a magnetic stirrer, with variable rotation speeds. In this experiment, the stirring speed of the bioreactors was controlled and regulated, ranging from 20 to 150 RPM, in order to evaluate the effect of stirring on the fermentation process, particularly on the homogeneity of the mixture and the accessibility of the substrate to microorganisms. The volume of biogas produced is measured using the liquid displacement method, which provides an accurate measurement of the gas generated based on the displacement of a liquid in a specific device.

2.2 Optimization of Biogas Production Using the RSM

The effect of operating parameters of a bioreactor on the kinetics of biogas production is studied individually in repeated experiments, and results observed in the literature have shown the interaction of these process parameters on biogas production [10]. However, processes fed with mixed substrates have shown a balanced nutritional content, promoting a high reaction rate, reduced inhibition, and better biogas production [11].

Traditional methods are not suited for optimization studies because they require extensive testing [12]. The RSM stands out as an effective tool for simulating, optimizing, and predicting biogas production. In the literature, optimization tools are widely used as approaches to optimize complex systems [26]. The RSM tool allows for reducing the number of experimental trials, determining interactions, and evaluating the influence of operating parameters in an effective and optimal manner.

This part of the paper presents the modeling and optimization of biogas production from mixed substrates, with varied proportions, under mesophilic and thermophilic conditions. The retention time was 30 days, and the working volume was 1000 mL. Data from twenty biogas fermentations conducted at laboratory scale were used to form bioreactors operating in batch mode. The effect of biogas production parameters, such as substrate mixture composition and temperature, are considered and selected as input features for RSM. A central composite design RSM is used to define the experimental design for anaerobic co-digestion. Based on the results from analysis of variance (ANOVA), the obtained models were highly significant, with a determination coefficient (R^2) of 0.90 for the cumulative biogas production. The results also show that the quadratic models obtained are acceptable for the relationship between the studied parameters (mixture composition, temperature) and the response (volume of biogas produced).

2.2.1 Physico-chemical characteristics of substrates

In this study, the main substrates used are bovine slurry (S1) and household waste (S2), which are collected, ground, and mixed. To measure the moisture content of the substrates, 10 g samples were placed in an oven at a temperature of 105°C for 24 hours. After drying, their weight was measured again. The weight difference (before and after drying) allows for determining the initial moisture content of the substrates. Total solids (TS) are defined as the solid content remaining after the evaporation of the water content of the sample. Table 1 presents the results of physicochemical characterisation.

Table 1. Physicochemical characteristics of the organic waste

Organic Waste	pH	Density	Moisture Content	Dry Matter	Mineral Matter	Organic Matter
Household waste	4.30	0.97 g/ml	47%	53%	34.4%	65%
Cattle manure	8	1.3 g/ml	70%	30%	43%	57%

A 1000 mL Erlenmeyer flask with a total volume were used as Batch-type digesters and placed in a water bath with adjustable temperature (between 25°C and 55°C). Necessary precautions were taken to make the setup airtight. The temperature of the bath was measured and controlled by a thermocouple immersed inside the thermostatic tank, and this thermocouple was connected to a data acquisition card for collecting the various measured temperature values.

The substrates to be fermented were mixed with water at a ratio of 1/3 to control the TS in the substrate and generate methanogenic bacteria within the substrate. The temperature inside the tank was regulated to ensure operation under thermophilic and mesophilic conditions, with a temperature ranging from 35°C to 55°C. The factors influencing biogas production were selected and are as follows: The composition of the substrates and the fermentation temperature.

2.2.2 Design of the RSM

The factors influencing biogas production were investigated experimentally using RSM. The relationship between the independent variables and the biogas yield was analyzed through this methodology. For the numerical experiments, the central composite design (CCD) was determined to be the most suitable model [14]. Table 2 presents the independent variables along with their coded and actual values.

Table 2. Encoded and real values

Factor	Name	Low Level	High Level
X_1	S1 (%)	0%	100%
X_2	S2 (%)	0%	100%
X_3	T (°C)	35°C	55°C

The trial version of Design Expert 7.0 was employed to carry out the optimization and prediction of the experimental outcomes. The system's behavior was modeled using a second-order polynomial function.

$$R = \alpha_0 + \sum_{i=1}^3 \alpha_i \cdot X_i + \sum_{i=1}^3 \alpha_{ii} \cdot X_i^2 + \sum_{i=1}^3 \sum_{j=i+1}^3 \alpha_{ij} \cdot X_i \cdot X_j \quad (1)$$

where,

X_i and X_j represent the independent parameters being studied;

R is the predicted response;

α_0 and α_i are the intercept and linear effects;

α_{ii} and α_{ij} are the quadratic and interaction coefficients.

A total of twenty experimental runs were performed in this design. The lack of fit and the experimental variability were assessed through a central run. The suitability of the models, along with the statistical relevance of the process parameters, was examined using ANOVA. The adequacy of the quadratic model was confirmed by calculating the coefficient of determination (R^2). Moreover, the p-value and the F-test (F-value) were employed to evaluate the statistical significance of the quadratic model.

2.2.3 Analysis of statistical data from the RSM

A total of 20 experimental runs were planned using the CCD within the framework of RSM. The aim was to determine the maximum cumulative biogas production and to optimize the independent variables: the substrate compositions (S1 and S2) and the temperature. These parameters were varied within their specified lower and upper limits to study their effects on the selected response. The optimization parameters used in the design are summarized in Table 3.

Table 3. Optimization parameters

Parameters	Value
Study Type	Response Surface
Design Type	Central Composite
Design Model	Quadratic
Groups	7
Subtype	Split-plot
Runs	20

ANOVA was performed to assess the statistical significance of the quadratic model equations. The main and interaction effects, reflecting the relationship between the response variables and the quadratic models, are summarized in the ANOVA tables. The coefficient of determination (R^2) for the quadratic model of cumulative biogas production was 0.90. In addition, the p-values of the models were below the corresponding F-values and less than 0.05, confirming that the quadratic equations are statistically significant and suitable for describing the system.

3 Results and Discussion

The temperatures of the bioreactor and the water heater were monitored and recorded using LabView software, interfaced with a data acquisition and control card. Measurements were taken at 30-minute intervals over a period of 14 hours daily for 30 days. The experimental results from these series will be presented in the form of graphs.

Figure 3 illustrates the evolution of the temperature of the heating system of the 50-liter bioreactor. Figure 4 shows the temperature inside the bioreactor, where it is observed that after an initial adjustment phase, the temperature tends to stabilize around 40°C. This stabilization indicates that the heating system has reached thermal equilibrium, allowing the bioreactor to maintain a constant and optimal temperature for the ongoing biological process. The importance of this thermal regulation lies in the fact that a stable temperature is crucial for the proper functioning of biological reactions, as slight temperature fluctuations can affect the activity of the microorganisms or enzymes involved in the process.

Figure 5 presents the results obtained from the study of the influence of the usable volume of the Erlenmeyer flask on biogas production. These results show variations in the amount of biogas produced depending on the percentage of the usable volume of the bioreactor used and reveal that a usable volume of 70% of the total volume generates the maximum biogas production. The results indicate that managing the usable volume of the bioreactor is essential to optimize biogas production. A usable volume of 70% seems to be the best configuration, producing the highest amount of biogas (103 g) compared to other volumes (50%, 60%, 80%, 90%). This highlights the importance of selecting the optimal working volume for each methanization system to ensure maximum efficiency in biogas production.

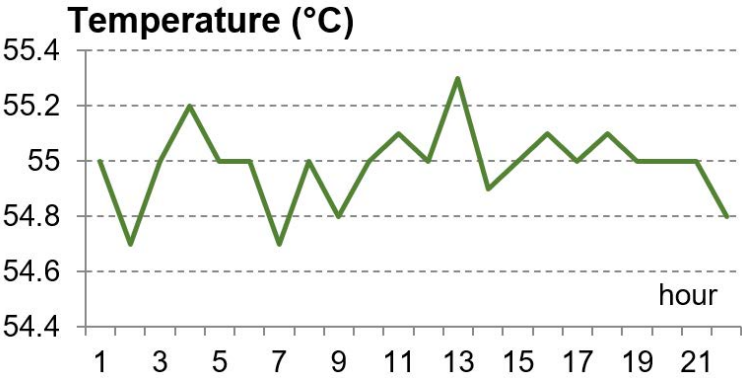


Figure 3. Temperature regulation at 55°C inside the water heater

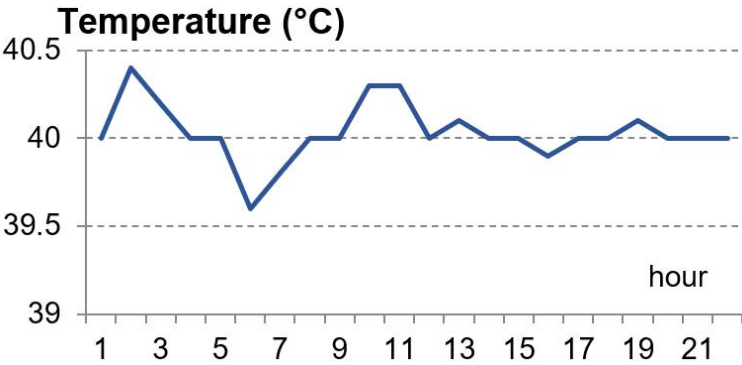


Figure 4. Temperature regulation at 40°C inside the bioreactor

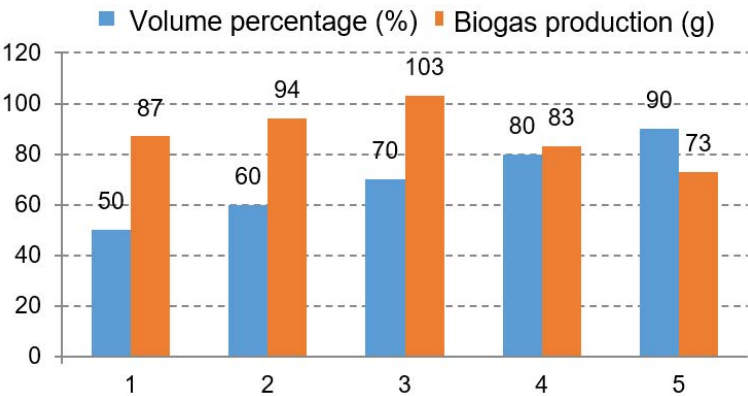


Figure 5. Biogas volume as a function of the usable volume

Figure 6 shows the marked influence of temperature on biogas production over time. The observation of a large volume of biogas produced at 40°C indicates that this temperature is optimal for anaerobic digestion in this particular case. At this temperature, the activity of methanogenic microorganisms is likely at its maximum, which accelerates

the degradation of the biodegradable substrate. This temperature is generally favorable for the fermentation of household waste, especially in the range of upper mesophilic conditions (around 40–45°C), where microbial activity is faster. This high volume of biogas produced at 40°C confirms the hypothesis that temperature favors the activation of enzymes and bacteria necessary for the degradation of organic matter.

Figure 7 shows the influence of pH value on biogas production, with the optimal pH being 7, which is neutral, at which biogas production reaches its maximum level. Neutral pH is generally optimal for most microorganisms involved in anaerobic digestion, particularly methanogens. These microorganisms grow and metabolize the substrate effectively, thus producing the maximum amount of biogas. Neutral pH promotes better enzymatic activity and greater stability in the process.

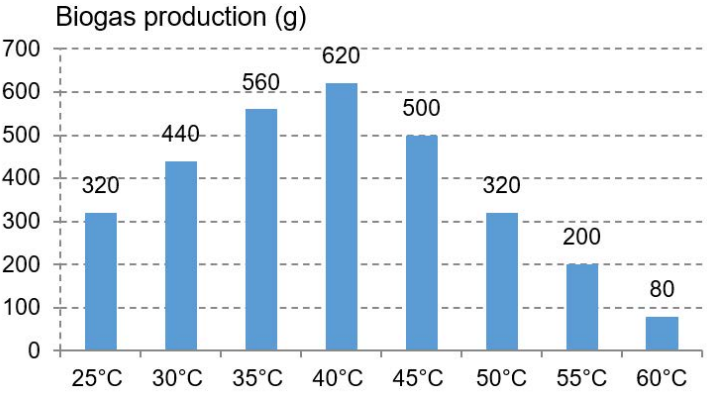


Figure 6. Biogas volume as a function of fermentation temperature

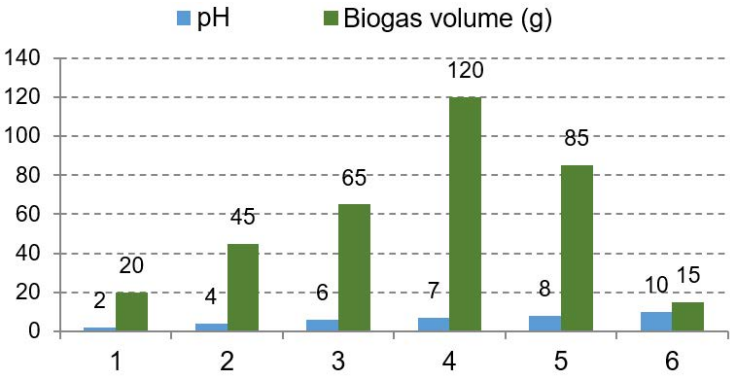


Figure 7. Influence of pH on biogas production

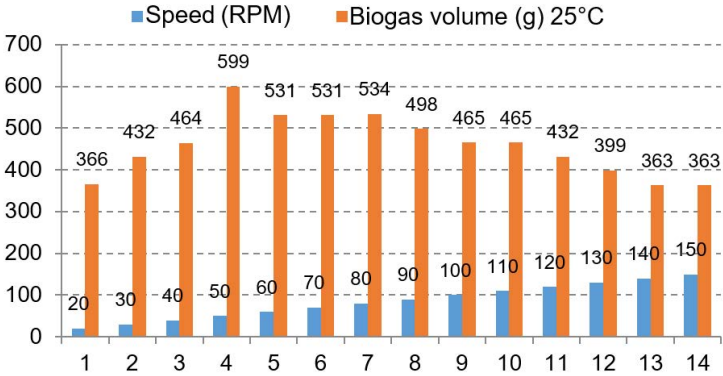


Figure 8. Biogas volume as a function of agitation speed

Figure 8 shows the influence of stirring speed on biogas production. At 50 RPM, biogas production reaches its maximum. This result suggests that this stirring speed optimizes the conditions for anaerobic digestion. At this

speed, the stirring is sufficiently high to ensure good substrate dispersion and maximum interaction between the microorganisms and the substrate. This allows for more efficient degradation of organic matter and optimal biogas production. Stirring at this speed also promotes the homogeneity of the mixture, ensuring an even distribution of nutrients and better efficiency in the metabolic process of the microorganisms involved in biogas production.

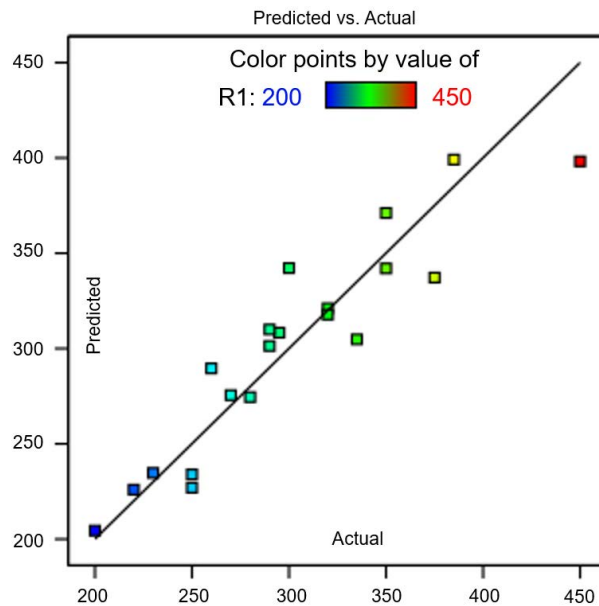


Figure 9. Correlations between predicted and observed data

Table 4. Optimization parameters

Run	Actual Value	Predicted Value	Residual	Internally Studentized Residuals	Externally Studentized Residuals	Standard Order
1	220	225.94	-5.94	-0.454	-0.593	20
2	250	233.95	16.05	0.775	0.707	19
3	270	275.55	-5.55	-0.259	0.026	3
4	300	342.23	-42.23	-2.281	-2.276	1
5	295	308.34	-13.34	-0.612	-0.355	2
6	260	289.59	-29.59	-1.248	-1.175	4
7	320	317.81	2.19	0.091	0.121	14
8	350	371.10	-21.10	-0.941	-0.928	13
9	335	304.83	30.17	1.363	1.151	7
10	290	301.37	-11.37	-0.486	-0.746	5
11	375	337.24	37.76	1.583	1.515	6
12	450	398.18	51.82	2.310	2.677	8
13	320	321.11	-1.11	-0.050	0.032	10
14	290	310.00	-20.00	-0.858	-0.818	9
15	350	342.05	7.95	0.333	0.357	18
16	385	399.17	-14.17	-0.625	-0.643	15
17	230	234.71	-4.71	-0.298	-0.215	17
18	200	204.49	-4.49	-0.210	-0.169	16
19	250	226.98	23.02	1.117	0.931	11
20	280	274.54	5.46	0.376	0.048	12

The "Predicted vs. Actual" plot presente in Figure 9 demonstrates a strong correlation between the experimental and model-predicted values of cumulative biogas production, with data points closely aligned along the diagonal line representing perfect prediction. This alignment indicates that the developed model exhibits high accuracy and reliability across the entire range of values, from 200 to 450. The color gradient, ranging from blue to red, reflects the variation in response values (R1) and confirms the model's consistency in predicting both low and high production outputs. The absence of significant outliers further supports the robustness of the model and its suitability for

optimizing biogas production under varying experimental conditions.

The effect of the S1+S2 mixture concentration was examined at various substrate ratios. Cumulative biogas production increased with a higher proportion of substrate S2 and elevated temperatures. The maximum experimental biogas production (approximately 450 mL) was observed when S2 constituted 70% and S1 30%. This initial increase in biogas yield with S2 is attributed to the abundance of easily biodegradable compounds in the substrate

Table 4 presents a comparison between the predicted and observed values of cumulative biogas production. Figure 9 illustrates the normal probability plots of the standardized residuals in the given sequence. The residuals were distributed uniformly around the regression line, indicating that the quadratic models are suitable for representing the selected response. Additionally, the interaction effects on the response variable (cumulative biogas production) were examined.

The three-dimensional surface plots illustrating the relationships between the response variable (cumulative biogas production) and the independent parameters (substrate S1, substrate S2, and temperature) are presented in Figure 10 and Figure 11. Figure 10 shows the combined effects of the independent variables on cumulative biogas production. Figure 11 presents the influence of the input parameters in more detail. It shows that cumulative biogas production increases with temperature, indicating that operation at 55°C enhances microbial activity and significantly improves biogas yield. Higher temperatures promote substrate biodegradability, stimulating the growth of methanogenic microorganisms and consequently increasing biogas production. Conversely, when the proportion of substrate S2 decreases, biogas generation tends to decline due to reduced microbial and enzymatic interactions within the mixture.

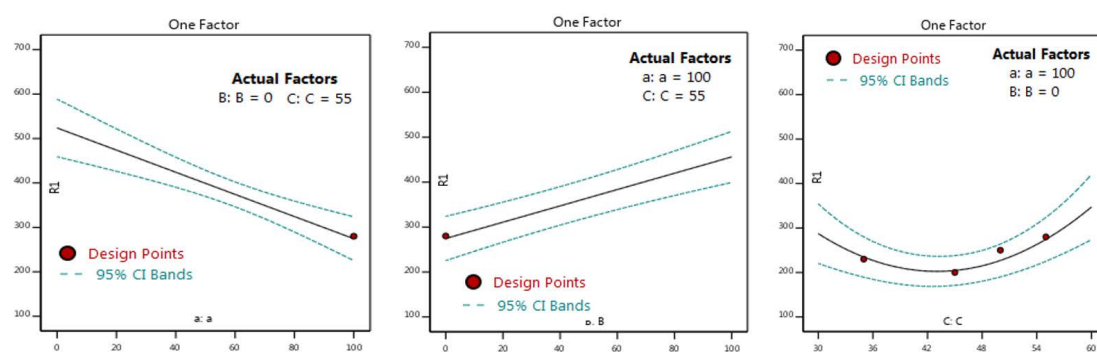


Figure 10. Effect of the combination of independent variable levels

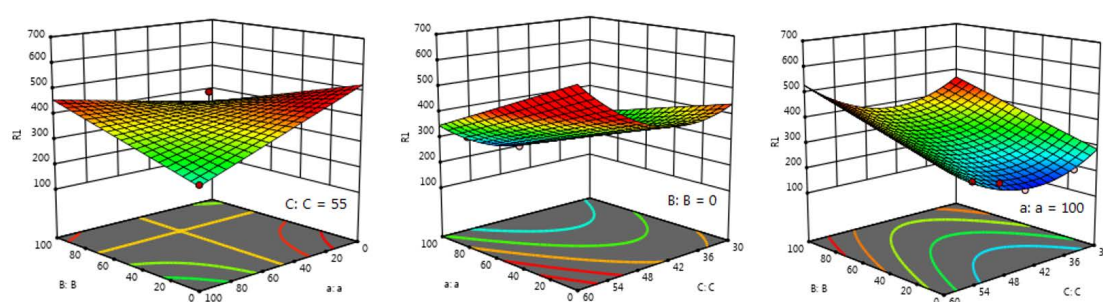


Figure 11. Influence of input parameters on biogas production

The second-order polynomial model applied in this study was used to determine the optimal condition aimed at maximizing biogas production (R1), and it was adjusted using quadratic minimization methods. The effect on the dependent variable R1 at different levels of the mixture of independent variables was studied to identify the concentrations of the substrates and temperature that result in the maximum value of R1.

Through numerical analysis, a set of solutions was obtained to determine the maximum levels of the selected response and the optimal values for each of the three parameters. Using RSM, the optimal conditions were identified as S1 at 26.66%, S2 at 55.55%, and a temperature of 59.33°C. Under these conditions, the predicted maximum cumulative biogas production was 483.89 mL. These optimal conditions were then tested experimentally to validate the proposed model equations. The observed results in Figure 12 was closely matched the values predicted by the quadratic models, indicating that RSM is an effective approach for optimizing the biogas production process.

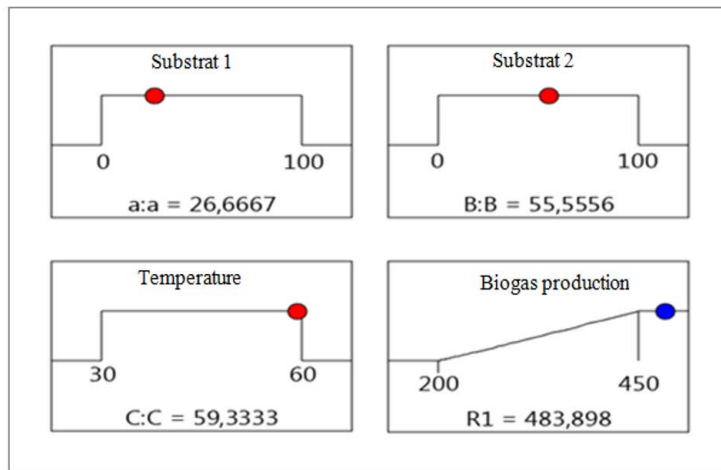


Figure 12. Optimal parameters for maximum response

The second-order polynomial equation is framed to signify the final regression model for biogas production:

$$R_1 = 1029.74 + 0.6133X_1 - 40.378X_3 + 0.03747X_1X_2 - 0.02118X_1X_3 + 0.5045X_3^2$$

4 Conclusions

The laboratory-scale optimization of biogas production has provided valuable insights into the key operational parameters influencing methanogenesis, namely temperature, agitation, substrate concentration, and pH. The experimental findings emphasize the necessity of precise control over these factors to achieve maximum biogas yield. Through the application of RSM, the study successfully identified optimal operating conditions, with the predictive models showing strong alignment with experimental data demonstrating the robustness of this approach for process optimization.

The validated models confirm the feasibility of using RSM to enhance anaerobic digestion processes, providing a scalable framework for the sustainable management of organic waste. These findings hold significant implications for the development of decentralized energy systems, particularly in rural and peri-urban areas of Southeast Asia and Indonesia, where organic waste is abundant and energy access is often limited.

To translate laboratory findings into real-world solutions, policies should promote investment in agricultural biogas units, provide financial incentives to replace coal with renewables, encourage co-digestion of agricultural and municipal waste, support research on integrating biogas with other green technologies, and enforce technical standards to ensure biogas quality for grid injection or bio-CNG production.

Future research should aim to validate these optimal parameters in industrial-scale bioreactors, considering real-world variability in substrates and environmental conditions. Moreover, integrating advanced monitoring technologies including artificial intelligence and machine learning can further improve process stability and energy output. Investigating co-digestion strategies and multi-resource integration would also enhance sustainability and system resilience. Ultimately, the advancement of optimized biogas production, backed by clear policy frameworks and investment strategies, offers a concrete path toward cleaner energy systems and circular waste management particularly in emerging economies like Indonesia.

This study has some limitations related to uncertainties in energy demand growth, technology costs, and policy implementation. Future demand may vary due to economic or demographic changes. Technology costs could fluctuate, affecting feasibility. Policy delays or shifts may alter outcomes. Future research should use scenario analysis or sensitivity testing to address these uncertainties.

Author Contributions

Conceptualization, O.M. and A.B.; methodology, O.M. and A.B.; software, O.M.; validation, O.M. and A.B.; formal analysis, O.M.; investigation, O.M.; resources, A.B.; data curation, O.M.; writing—original draft preparation, O.M.; writing—review and editing, O.M. and A.B.; visualization, O.M.; supervision, A.B.; project administration, A.B.; funding acquisition, A.B. All authors have read and approved the published version of the manuscript.

Data Availability

The data used to support the research findings are available from the corresponding author upon request.

Conflicts of Interest

The authors declare no conflict of interest.

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