

Optimization of Anaerobic Co-Digestion of Canna Indica Rhizome and Cow Manure

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Abstract - This study investigates the potential of *Canna indica* rhizome as a promising substrate for biogas production through anaerobic co-digestion with cow manure. Experimental results demonstrate that a 50:50 mixture of *Canna indica* rhizome and cow manure optimizes biogas yield, achieving a methane content of 64.48%. This synergistic effect is attributed to the complementary chemical composition of the substrates.

To provide an initial estimate of biogas production, an existing mathematical model was adapted without significant modifications. Despite its original design for different substrates, the model exhibited satisfactory performance in predicting biogas production trends, highlighting the general applicability of its underlying principles to this specific co-digestion system.

Keywords- Anaerobic co-digestion, *Canna indica* rhizome, Cow manure, Mathematical model, Synergy.

I. Introduction

Anaerobic co-digestion, a biological process of converting organic matter into biogas, represents a promising alternative for waste valorization and renewable energy production ([11], [21], [31], [2], [7], [17], [26], [29]). However, this process is far from simple. The diversity of substrates, complex interactions within the microbial consortium, and the sensitivity of the process to variations in operating conditions make it a highly dynamic and challenging system to model ([10], [13], [14], [24], [41]). This complexity necessitates a multidisciplinary approach combining microbiology, chemistry, process engineering, and thermodynamics ([7], [23], [26], [30], [36], [43]). By combining two or more substrates, whether rich in soluble compounds or lignocellulosic materials, a favorable environment can be created for the development of diverse microbial communities capable of degrading a wide range of organic compounds ([9], [12], [15], [16], [26]).

The concepts of synergy and antagonism are crucial in anaerobic co-digestion. Synergy refers to the beneficial effect resulting from the interaction between different substrates, where their combination enhances biogas production compared to the individual digestion of each substrate ([3], [9], [12], [16], [19], [25], [32], [34], [40]). Conversely, antagonism denotes negative interactions where the presence of certain substrates inhibits the degradation of others, thereby reducing the efficiency of the process ([32], [40], [7], [9], [12], [16], [25], [34], [42]). Understanding these interactions is essential for optimizing biogas production.

In this context, our study focuses on the co-digestion of two specific substrates: cow manure and canna rhizome. Cow manure is a substrate rich in organic matter and nutrients, while canna rhizome provides lignocellulosic compounds and fibers. The combination of these two substrates could potentially create a beneficial synergy due to the complementarity of their chemical compositions and physical properties.

The objectives of this study are manifold. Firstly, we aim to identify the optimal mixing ratio between cow manure and canna rhizome to maximize biogas production. Secondly, we will apply an existing model, developed by another researcher, to predict the volume of biogas produced based on key parameters such as pH, temperature, and digestion time [37]. Although this model was developed for different co-substrates, it could provide valuable insights into the synergistic and antagonistic interactions in our co-digestion system. Finally, we will evaluate the relevance and robustness of this model for our specific combination of substrates, with a view to its future adaptation for practical applications.

In summary, this study aims to deepen our understanding of the mechanisms of anaerobic co-digestion and to develop optimal strategies for the valorization of organic waste, thereby contributing to the sustainable and efficient production of renewable energy ([1], [7], [8], [39]).

II. MATERIALS AND METHODS

A. Instrumented digester

The experimental setup for this study consisted of a main digester (D) made of a 20-liter plastic tank. Two other identical tanks of the same volume completed the system: one of the tanks served as a gasometer, filled with water and connected to the digester by a pipe. It allowed to measure the volume of biogas produced, and the third tank, empty, was used to collect the water expelled from the gasometer under the pressure of the biogas produced.

Temperature sensors, a pH probe and gas analyzers were integrated into the digester to ensure real-time and automated monitoring of the key parameters of the anaerobic digestion process. (Fig.1.).

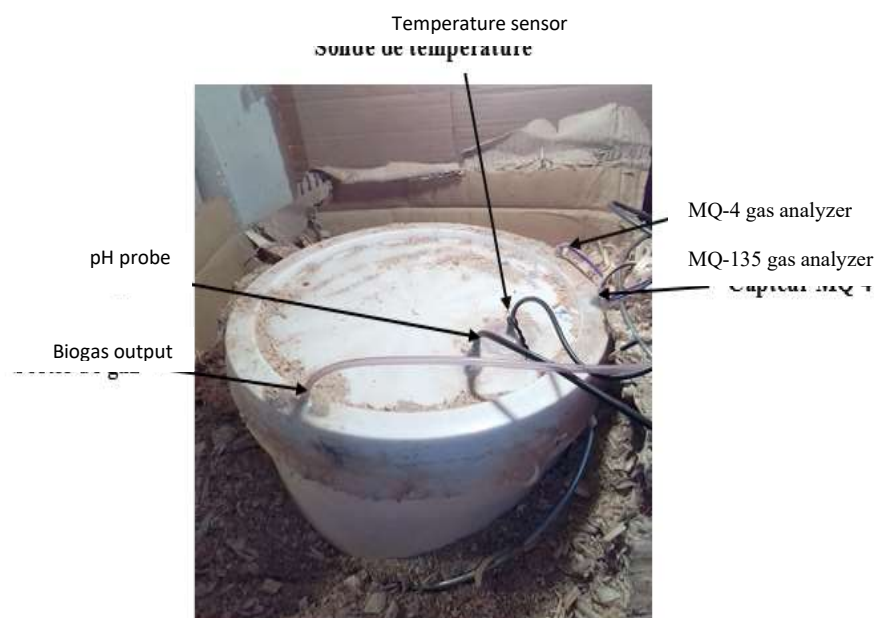


Fig.1. Schematic diagram of the experimental setup

B. Preparation of raw materials, collection, and experimental location

Samples of *Canna indica* were collected from the open field in the village of Ambohitra, located in the district of Andramasina in Madagascar.

After collection, the mature rhizomes underwent a cleaning process to remove soil and were then ground. Cow manure was collected from the vicinity of Ankatso University.

C. Experimental setup and procedure for biogas production

Batch digester experiments were conducted to evaluate the co-digestion of canna rhizome (CR) with varying proportions of cow manure (CM). Biogas production was quantified using the displacement method. Anaerobic digestion was maintained under mesophilic conditions (25-32°C) for a retention time that ranged from 22 to 29 days. Throughout the study, biogas production, biogas composition, temperature, and pH of the reaction medium were monitored in real-time.

D. Data Analysis and Multi-Start Optimization

To enhance our understanding of anaerobic co-digestion, we conducted a comprehensive analysis of biogas production data, employing rigorous statistical methods. We refined our model using a multi-start optimization approach, which involves starting from multiple random points and selecting the best solution. The model's accuracy was validated using MATLAB R2017a and R². Multi-start is a powerful technique for global optimization, ensuring thorough exploration of the parameter space and avoiding local minima [26].

III. RESULTS AND DISCUSSION

A. Daily biogas production

The daily biogas production profile exhibits a characteristic curve with distinct phases. Fig.2. illustrates this trend for both canna rhizome (CR) mono-digestion and CR/CM co-digestion.

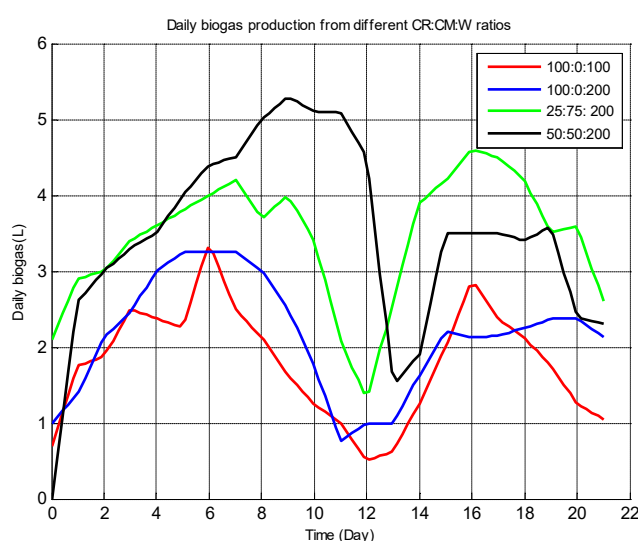


Fig.2. Daily Biogas Production Curves for Various Canna Rhizome (CR): Cow Manure (CM): Water (W) Ratios

The daily biogas production curves for various Canna rhizome (CR): cow manure (CM): water (W) ratios reveal intricate dynamics within anaerobic digestion. Each curve exhibits distinct phases of growth, stabilization, and decline, offering valuable insights into the biodegradation process for each mixture.

Mixtures containing only Canna rhizome (100:0:100 and 100:0:200) displayed similar patterns, characterized by an initial peak around day 7-8, followed by a rapid decline and a smaller secondary peak near day 18-20. While the 100:0:200 ratio consistently yielded more biogas, both ratios demonstrated lower overall production compared to manure-containing mixtures, suggesting nutrient limitations or microbial diversity issues with Canna rhizome alone. Notably, the higher biogas production observed with the 100:0:200 ratio suggests that a certain degree of dilution may be beneficial for optimizing Canna rhizome digestion.

Mixtures incorporating both Canna rhizome and cow manure (25:75:200 and 50:50:200) exhibited significantly higher biogas production and more complex patterns. Initially, both ratios demonstrated a rapid increase in production until day 7-8, indicative of rapid microbial growth and easy substrate utilization. A subsequent decline around day 8-9 suggested the depletion of easily degradable substrates. However, a notable secondary rise began around day 14-15, peaking near day 18-20, possibly due to microbial adaptation to less readily degradable compounds or the onset of methanogenesis from intermediate products. Production eventually decreased after day 20, signaling the depletion of biodegradable substrates.

The 50:50:200 ratio displayed the highest peaks and overall production, while the 25:75:200 ratio maintained a more consistent production level, especially in later stages. These patterns reflect the complex interplay of various factors in anaerobic digestion, including substrate composition, microbial population dynamics, and metabolic pathways.

B. Cumulative biogas production

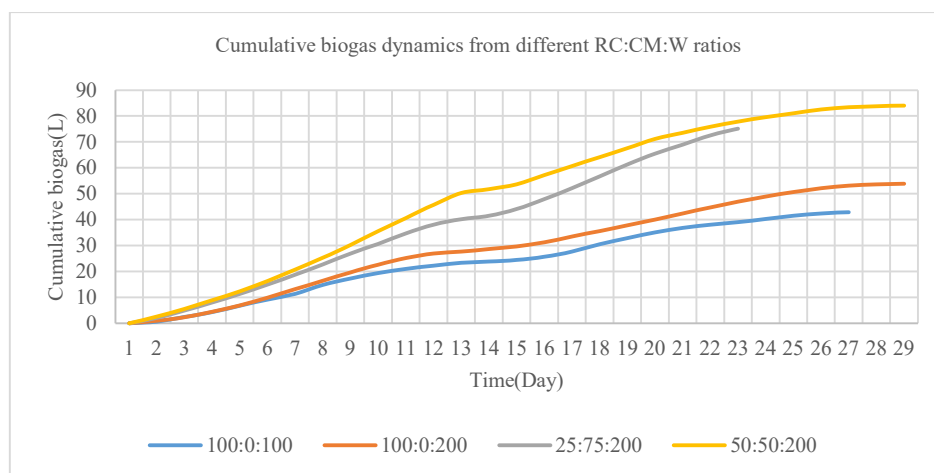


Fig.3. Temporal evolution of biogas production as a function of mixture ratio

The ratio 100:0:100, with a lower water content, produces less biogas compared to the ratio 100:0:200. Higher water content creates a more favorable environment for microbial growth, improves nutrient availability, and reduces mixture viscosity. These factors ultimately enhance mass and heat transfer, which are essential for efficient biogas production. This is evident in the graph, where the 100:0:200 ratio (orange curve) consistently outperforms the 100:0:100 ratio (blue curve) throughout the 29-day period.

The inclusion of cow manure significantly enhanced biogas production, as seen in the superior performance of the 25:75:200 (grey curve) and 50:50:200 (yellow curve) ratios. This suggests that the added organic matter and microbial inoculum from manure contributed to a more active and efficient anaerobic digestion process.

Ratios containing manure (25:75:200 and 50:50:200) displayed more stable production profiles and higher cumulative production levels. The graph shows cumulative biogas production over 29 days. The 50:50:200 ratio produced the most biogas, reaching about 85 L after 29 days, closely followed by the 25:75:200 ratio at approximately 75 L.

Interestingly, the 25:75:200 ratio showed a steeper slope towards the end of the 29-day period compared to the 50:50:200 ratio. This indicates that the 25:75:200 mixture still had significant biogas production potential at the end of the experiment, suggesting a longer retention time might be beneficial for this ratio.

C. Biogas composition: impact of mixing ratios

Our experiments yielded biogas with methane concentrations ranging from 60% to 64.48%, aligning with typical reported values of 40% to 75% [18]. A consistent trend showed initial low methane content, increasing steadily as digestion progressed, indicative of methanogenic microorganism growth. This pattern mirrors typical anaerobic digestion dynamics, validating our experimental setup and allowing reliable comparisons with other studies.

Gas composition variations highlight the complex interplay between substrate, microbes, and biogas production. Optimizing feedstock mixing ratios is crucial for efficient anaerobic digestion and high-quality biogas, offering valuable insights for designing better systems and improving yields.

We studied four ratios: two mono-digestion and two co-digestion mixtures of *Canna indica* (*C. indica*) and cow manure, focusing on co-digestion due to its potential to enhance biogas yield and quality through synergistic effects and improved process stability. Over 22-29 days, biogas samples were collected:

- 50% *C. indica* - 50% Cow Manure yielded 64.48% methane ;
- 25% *C. indica* - 75% Cow Manure yielded 62% methane.

Significant differences were observed, with the 50-50 mixture yielding slightly higher methane content, hinting at benefits from *C. indica*. Both mixtures generated high-quality biogas with methane concentrations above 60%, confirming the viability of co-

digestion. The modest difference (2.48%) indicates process stability. Exploring ratios near the 50-50 mark and factors like substrate pretreatment, temperature, and pH could optimize methane production and support larger-scale applications.

Table I presents a comparative analysis of methane concentrations produced from anaerobic co-digestion using various co-substrates, as reported by multiple studies.

Table I. Methane Concentration from Anaerobic Co-Digestion: A Comparative Analysis

Substrate	Co-substrate	Methane (%)	Reference	
Corn	Cow manure	51-62	Abdoli et al.(2014)	[1]
Canteen wastewater	Food waste	61	Pongthornpruek et al. (2017)	[35]
Water hyacinth	Cow manure	67.11	Ali et al. (2022)	[5]
Water hyacinth	Pig and poultry droppings	64.92	Akindele et al. (2019)	[4]
Waste fruits	Waste vegetables	57	Salim et al. (2023)	[38]
C. indica (1% CaO)	Buffalo dung	60.92	Muronda et al. (2023)	[33]
C. indica (2% CaO)	Buffalo dung	64.93	Muronda et al. (2023)	[33]
C. indica (3% CaO)	Buffalo dung	55	Muronda et al. (2023)	[33]
C. indica 50%	Cow manure 50%	64.48	This study	
C. indica 25%	Cow manure 75%	62	This study	

Based on the provided table comparing methane content from various substrate and co-substrate combinations, we can draw several conclusions:

- High methane yields: the methane yields achieved in our study (64.48% for 50% C. indica - 50% cow manure and 62% for 25% C. indica - 75% cow manure) exceed the average methane content reported in the literature for similar substrate combinations, demonstrating the effectiveness of C. indica as a substrate.
- Consistency with previous studies: our results are consistent with other studies using similar substrates, such as corn and cow manure.
- Comparison with other plant-based substrates: C. indica performs similarly to or better than other plant-based substrates like water hyacinth and waste fruits/vegetables.
- Effect of mixing ratios: our study's results demonstrate the importance of optimizing substrate ratios for maximum methane production. The 50:50 mixture slightly outperformed the 25:75 mixture.
- Versatility of animal manure: animal manure, including cow, buffalo, pig, and poultry, consistently performs well as a co-substrate.
- Comparison with pretreated substrates: our study's results without pretreatment compete favorably with those obtained by Muronda et al. (2023) using CaO pretreatment. This suggests that proper substrate selection and mixing can be as effective as or more effective than certain chemical pretreatments.
- Future research: future studies could explore combining optimized mixing ratios with mild pretreatment methods to potentially achieve even higher methane yields.

Our study's results for C. indica and cow manure co-digestion are promising for biogas production. The high methane yields achieved without additional treatments or additives suggest the potential for further optimization through careful substrate selection and mixing.

D. Biogas Volume Prediction Model

The production of biogas through anaerobic co-digestion is a complex process influenced by numerous factors, including temperature, pH, and digestion time. To better understand and optimize this process, it is essential to have a model capable of predicting the volume of biogas produced based on these parameters. In this study, we utilize an existing mathematical model developed by researchers Rajaonahy et al., to predict the volume of biogas produced from the anaerobic co-digestion batch experiments of cow manure and canna rhizome.

The model used is described by the following equation [37]:

$$V = aT^b pH^c \sin(\omega t + \phi)$$

with

V : Volume of biogas

T : Temperature

pH : Hydrogen potential

t : Time

a, b, c, ω, ϕ : Model parameters

This model takes into account the combined effects of temperature, pH, and time on biogas production. The parameters a, b, c, ω, ϕ are adjusted to reflect the specific conditions of our co-digestion system. By using this model, we can not only predict the volume of biogas produced but also analyze the synergistic and antagonistic interactions between the substrates, as well as the influence of environmental conditions on the digestion process.

The model developed by Rajaonahy et al. has been previously applied to different co-substrates, such as food waste and agricultural residues, other than cow manure and canna rhizome. However, its application to our specific combination of substrates can provide valuable insights into the synergistic and antagonistic interactions in our co-digestion system.

The objective of this part of the study is to apply this existing model to our experimental data, evaluate its accuracy and robustness, and discuss its potential for practical applications in optimizing biogas production. By better understanding the underlying mechanisms of anaerobic co-digestion, we can develop more effective strategies for the valorization of organic waste and the production of renewable energy.

a. Parameter Estimation

To apply the model to our experimental data, the parameters a, b, c, ω, ϕ were estimated using the MultiStart method. This method involves multiple starting points to find the global minimum of the parameter estimation problem, ensuring a more accurate fit to the experimental data.

Figure 4 illustrates the execution of the local solver for determining the model constants for all mixture.

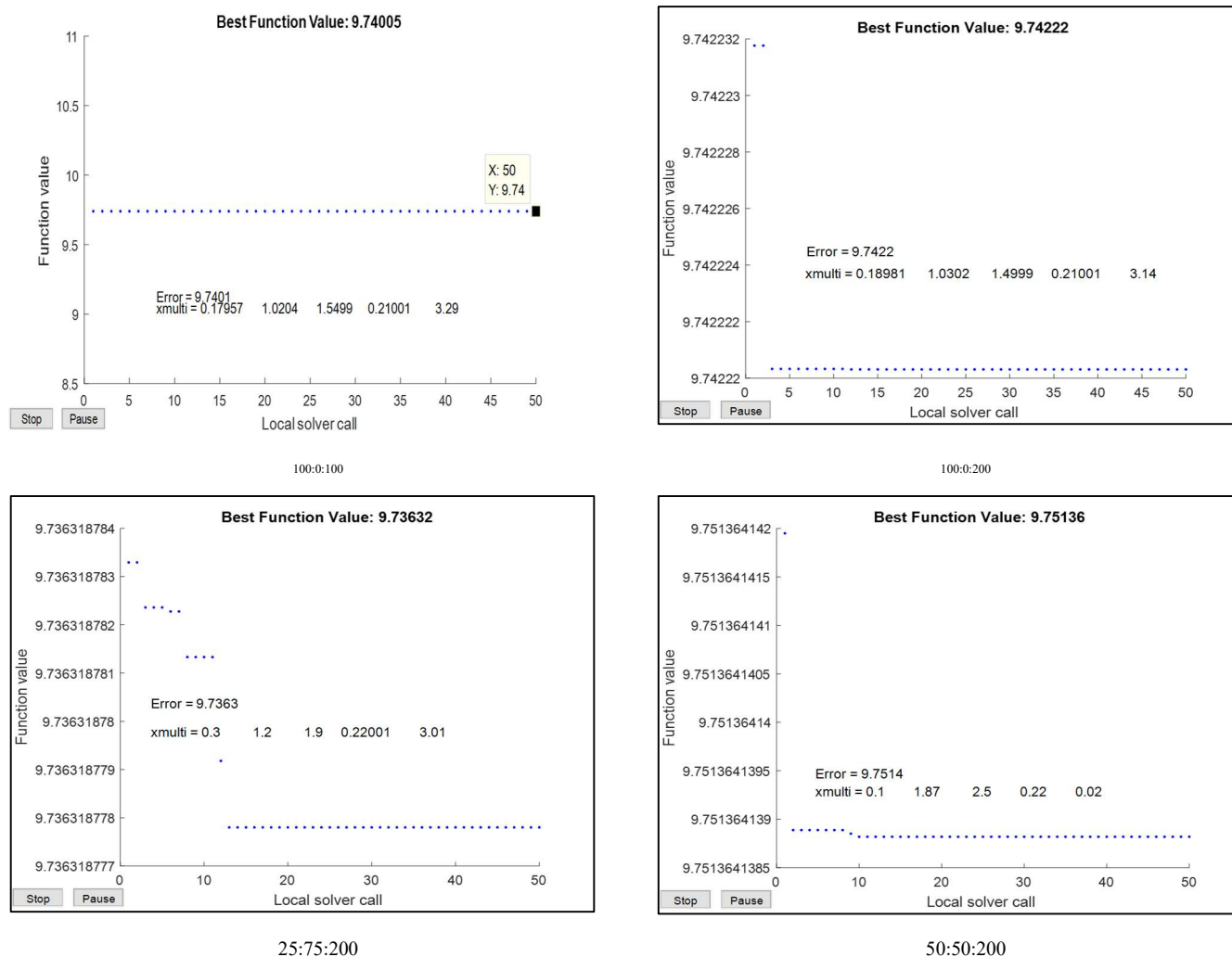


Fig.4. Local solver execution for model constant determination for RC: CM: W blends

b. Comparison of Predicted Results with Experimental Data: R^2 Values and Model Performance

To assess the model's accuracy, we compared its predictions to experimental data. As shown in Figures 5, 6, 7 and 8, the predicted values closely align with the experimental measurements.

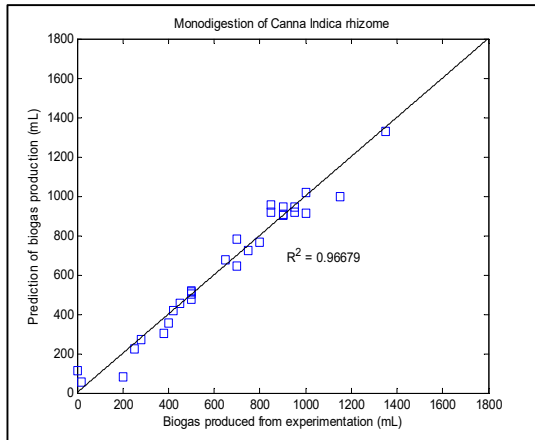


Fig.5. Scatter plot of theoretical and experimental biogas production for cow manure and Canna indica rhizome mixture 100:0:100

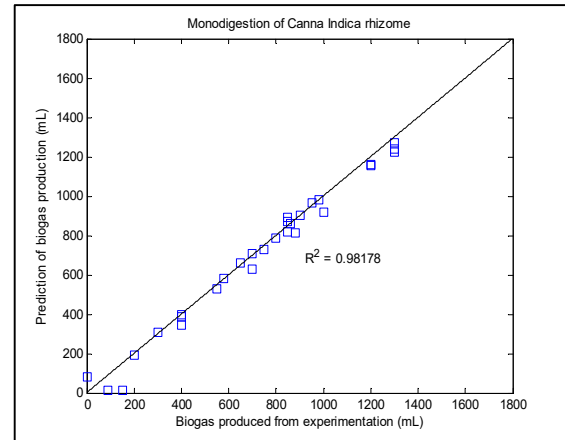


Fig.6 Scatter plot of theoretical and experimental biogas production for a mixture of cow manure and Canna indica rhizome 100:0:200

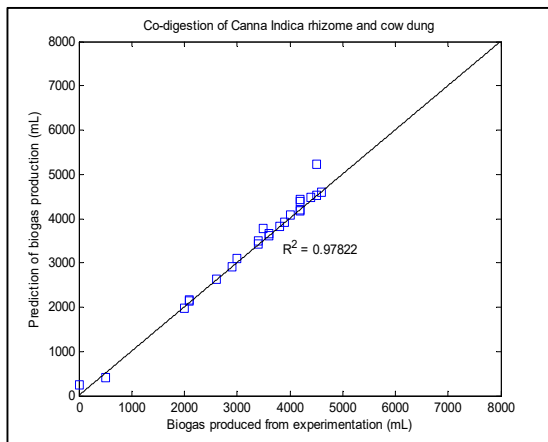


Fig. 7. Experimental vs. theoretical results for a mixture of cow manure and Canna indica rhizome (25:75:200)

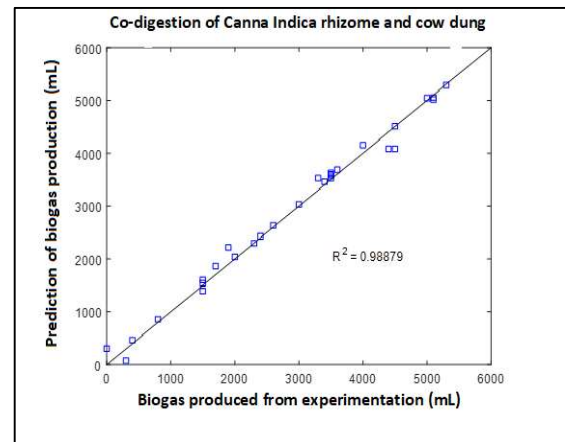


Fig. 8 Comparison of experimental and theoretical results for cow manure and Canna indica mixture 50:50:200

For all four ratios studied, the comparison graph between predicted and experimental biogas production values demonstrates an excellent correlation. The coefficient of determination, approaching 1, indicates that the model is highly effective in estimating biogas production within the context of the investigated co-digestion system. The data points are tightly clustered around the regression line, further confirming the reliability of the model's predictions.

c. Results summary: kinetic parameter values and model fit quality for each ratio

Table II provides a comprehensive summary of the model's optimal parameters, obtained through the MultiStart method. It also includes the coefficient of determination (R^2) for each scenario, a key measure of the model's ability to explain the variability in the experimental data.

Table II. Optimal Model Parameters and Corresponding Goodness of Fit (R^2)

Ratio	Constante					Coefficient of determination R^2
	a	b	c	ω	Φ	
100 :0 :100	0,17957	1,0204	1,5599	0,21001	3,29	0,96679
100 :0 :200	0,18981	1,0302	1,4999	0,21001	3,14	0,98178
25 :75 :200	0,30	1,2	1,9	0,22001	3,01	0,97802
50 :50 :200	0,10	1,87	2,50	0,22	0,02	0,98879

The analysis of the model $V = aT^b pH^c \text{abs}[\sin(\omega t + \varphi)]$ and its constants for different substrate ratios reveals valuable information, especially when compared to the results of Rajaonahy model applied to other co-substrates.

The interpretation of the model constants will be addressed first:

- The 'a' parameter, ranging from 0.10 to 0.30, indicates that the overall magnitude of the observed phenomenon is susceptible to changes in the mixture composition. The increase in 'a' with the addition of the second component indicates that it might amplify the overall effect.
- The exponents 'b' and 'c,' respectively representing temperature and pH, exhibit substantial variability. 'b' ranges from 1.0204 to 1.87, while 'c' varies from 1.4999 to 2.50. These values, always greater than 1, indicate a strong non-linear relationship between $V(t)$ and these two factors. The increase of these exponents in more balanced ratios suggests that temperature and pH have a more pronounced influence under these conditions.
- The parameter ω remains relatively stable around 0.21-0.22 for all ratios. This consistency indicates that the cycle frequency in the system is little affected by changes in mixture composition.
- In contrast, Φ shows great variability, ranging from 0.02 to 3.29. This significant variation in phase shift suggests that the cycle timing is strongly influenced by the mixture composition.

These observations on the model constants are even more interesting when viewed in perspective with the obtained coefficients of determination (R^2). Our model consistently displays R^2 values above 0.96, even reaching 0.98879 for the 50:50:200 ratio. These values are remarkable, especially when compared to the results of the model applied to other co-substrates, where R^2 varied from 0.86 to 0.96[37].

This comparison highlights several points:

- The robustness of our model: It maintains very high R^2 values across different substrate ratios, even surpassing the performance of the model in some cases.
- The suitability of the chosen substrates: The tested ratios seem particularly well-adapted to our model, allowing superior adjustments.
- The flexibility of the model: The model's ability to effectively adjust to different ratios while maintaining high R^2 values suggests that the parameters a, b, c, ω , and Φ efficiently capture the underlying dynamics of the system.

In conclusion, the model not only demonstrates excellent predictive capability for the studied substrates and ratios, but it also offers a finer understanding of the influence of different factors (temperature, pH, mixture composition) on the studied phenomenon. This superior performance, combined with the interpretability of the model constants, opens interesting perspectives for optimizing processes involving these substrates and could justify further exploration of the model's applicability to other similar systems.

IV. CONCLUSION

This study demonstrates the significant potential of canna rhizome (CR) as a promising substrate for biogas production. By optimizing its co-digestion with cow manure, we achieved a substantial increase in biogas yield, with a 35.53% increase for the 50:50 mixture compared to the monodigestion of individual substrates, highlighting the synergistic benefits of substrate combinations in enhancing biogas generation.

The developed model exhibits exceptional performance in predicting biogas production from the studied co-substrates, with a coefficient of determination (R^2) exceeding 0.98. This remarkable accuracy underscores the model's robustness and its ability to capture the complexities of the co-digestion process.

While these results are encouraging, further research is warranted to comprehensively characterize the biochemical composition of RC and to assess the influence of various operating conditions on biogas yield and quality. A deeper understanding of these factors will facilitate the development of optimized co-digestion strategies.

Rigorous experimental testing is necessary to validate the model's robustness and ensure its generalizability. A sensitivity analysis will help identify the most influential parameters and assess the uncertainty associated with the model's predictions.

A thorough comparison with existing models will provide insights into the strengths, limitations, and potential improvements of the developed model.

The obtained results can be used to optimize co-digestion conditions, such as temperature, retention time, and C/N ratio, to maximize biogas production and reduce costs.

In the long term, developing a simulation tool can assist industries in sizing and optimizing their co-digestion facilities according to their specific needs.

This study paves the way for sustainable valorization of canna rhizome, offering new opportunities for diversifying biogas production substrates. The results obtained lay the groundwork for future research aimed at refining co-digestion processes and developing accurate predictive models. By combining RC with other co-substrates, it is possible to enhance digester performance and accelerate the energy transition.

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