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Investigating the Potential of Indica Canna Rhizome for Bioethanol Production

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Abstract— Madagascar's energy landscape is heavily reliant on finite fossil fuels and charcoal, raising concerns about sustainability and environmental impact. The burgeoning industrial sector and escalating energy demands necessitate a shift towards eco-friendly and renewable alternatives. This research delves into the potential of Canna indica, a rhizomatous plant, for bioethanol production.

Bioethanol extraction was achieved by harnessing the starch content of Canna indica rhizomes through a multi-step process involving enzymatic hydrolysis, fermentation, and distillation. The starch content of the peeled dry extract was found to be 74.5% basis on dry matter, yielding 72.30 ml of bioethanol ranging from 6° to 45°. The unpeeled dry extract produced 64.67 ml of bioethanol ranging from 5° to 30°, while the fresh rhizome yielded 64.79 ml of bioethanol ranging from 6° to 44°. The remaining waste biomass, including leaves, stems, and distillation residues, was utilized for the production of biochar and briquettes, as detailed in another article.

Keywords—Bioethanol; Canna indica; Rhizome; Renewable energy; Sustainability; Biomass.

I. INTRODUCTION

The global reliance on fossil fuels, particularly petroleum, has significantly influenced economies and lifestyles over the decades. However, the depletion of these non-renewable resources and their environmental impacts necessitate a transition to sustainable energy sources. Climate change, characterized by rising global temperatures, increased frequency and intensity of extreme weather events, and ocean acidification, is a major consequence of this dependency ([20], [21]).

Biofuels, such as bioethanol, offer a promising alternative to reduce greenhouse gas emissions and diversify the energy mix [20]. Traditionally, bioethanol is produced from crops like sugarcane, corn, or sugar beet ([7], [11], [12], [16]). However, new biomass sources are being explored, including Canna indica, a plant native to South America but widely cultivated in Madagascar.

Canna indica has recently gained attention as a potential biomass source for bioethanol production. The plant's rhizome is rich in starch, making it an attractive candidate for bioethanol production. Unlike other energy crops, Canna indica is well-adapted to Madagascar's unique soil and climate conditions, including poor soils. This adaptability makes it a resilient and sustainable option for biofuel production in the region. Additionally, Canna indica does not compete directly with food crops, which is crucial for maintaining food security in Madagascar.

Despite the promising potential of Canna indica for bioethanol production, there is limited research on the subject. The feasibility and efficiency of using Canna indica rhizomes for bioethanol production need to be carefully studied. Understanding the agronomic characteristics of the plant, its yield potential, and the conversion efficiency of its rhizomes into bioethanol is essential for determining its viability as an energy crop ([3], [4], [5], [14], [18], [19], [22], [23]).

The production of bioethanol involves several key steps. First, the biomass is pretreated to break down the complex carbohydrates into simpler sugars. This is typically achieved through mechanical, chemical, or enzymatic methods [12]. Mechanical pretreatment involves grinding or milling the biomass to increase its surface area, making it more accessible to enzymes or chemicals. Chemical pretreatment uses acids, bases, or solvents to break down the biomass structure. Enzymatic pretreatment employs enzymes to hydrolyze the complex carbohydrates into fermentable sugars ([13], [15], [17]).

The resulting sugars are then fermented using yeast or bacteria to produce ethanol. Fermentation is a critical step in the bioethanol production process, as it determines the yield and quality of the final product. Various fermentation techniques can be employed, including batch, fed-batch, and continuous fermentation. Batch fermentation involves adding all the substrate at the beginning of the process, while fed-batch fermentation involves gradually adding the substrate to maintain optimal conditions for the microorganisms. Continuous fermentation, on the other hand, involves a steady flow of substrate and product, allowing for higher productivity and efficiency [1].

Finally, the ethanol is distilled and purified to obtain the final product. Distillation separates the ethanol from the fermentation broth, while purification removes any remaining impurities. The efficiency of these processes can vary significantly depending on the type and quality of the biomass used ([2], [8]).

This study aims to evaluate the potential of Canna indica as an energy crop in Madagascar. The objectives include assessing the agronomic advantages of Canna indica, including its adaptability to local soil and climate conditions, studying the starch content and yield potential of Canna indica rhizomes, evaluating the conversion efficiency of Canna indica rhizomes into bioethanol, and highlighting the development prospects of Canna indica as a biomass source for bioethanol production in Madagascar.

The results of this study could contribute to diversifying biomass sources for bioethanol production in Madagascar and strengthening the country's energy security. By exploring the potential of Canna indica, this research aims to provide valuable insights into the feasibility of using this plant as a sustainable and renewable energy source ([3], [5], [14], [18], [19], [22], [23]).

II. MATERIALS AND METHODS

A. Raw material, Plant Identification and Analysis

The rhizomes of Canna indica were used as the raw material for this study. These rhizomes were collected from Ambohitra village, located in the Andramasina district of Madagascar.

To ensure the accurate identification of the plant, botanical experts from the Herbarium of the Tsimbazaza Botanical and Zoological Park (PBZT) were consulted. Identification was confirmed using a herbarium specimen, which involves carefully arranging the entire plant on a large sheet of paper with adhesive strips to showcase all its distinctive features.

Analyses to determine the starch content were conducted at the CNRE Laboratory of Food and Water Analysis and Control.

B. Preparation of rhizome extracts

Three types of extracts were used in the experiments: fresh material, dried unpeeled material, and peeled dried material. The rhizomes were washed after being removed from the soil, regardless of the type of extract required.

For the first type of extract, only the cortex was removed, and then the rhizomes were ground using a mixer. For the other two types of extracts, they were dried directly in the sun, after peeling for the peeled dried extract, and then they were weighed every six hours until their weights remained constant. Finally, they were ground.

C. Ethanol Production from Canna Rhizome: A Three-Step Process

a. Hydrolysis, Enzyme Addition, and Gel Cooking

The enzymatic hydrolysis of complex starch molecules into simple sugars, such as glucose and maltose, was catalyzed using a solution containing amylases. Cereal milk, specifically paddy (rice) milk, served as the enzyme source. This milk contains two types of amylases: α-amylase and β-amylase. α-amylase randomly cleaves the 1-4 bonds of amyloses, leading to liquefaction, while β-amylase primarily releases maltose by breaking the 1-4 bonds.

Starch breakdown into simple sugars (hydrolysis) occurred in three stages: 1) gelatinization (heating and color change) for uniform cooking, 2) cooling to 50°C to protect the added enzyme, and 3) enzyme addition to break down branched starch chains (visible liquefaction). Vigorous mixing ensured even cooking and enzyme activity.

To optimize bioethanol production from Canna indica rhizomes, we conducted a series of experiments by varying several key parameters during the enzyme addition and gel cooking steps. These parameters included the quantity of added enzymes, cooking temperature, cooking duration, and the amount of hydrolyzed wort obtained. These experimental conditions were carefully adjusted for each type of extract to maximize biomass conversion into fermentable sugars. Table 1 summarizes the experimental conditions used for each extract and the corresponding quantity of hydrolyzed sweet juice obtained.

b. Fermentation

Following hydrolysis, the cooled hydrolyzed sweet juice (30°C) was inoculated with Saccharomyces cerevisiae yeast and fermented under dark conditions at 29°C. A 5% (w/w) yeast inoculum was employed. Successful fermentation was evident by the presence of bubbling in each container.

c. Distillation and alcohol volume

The fermented mixture was distilled at 60-90°C to separate the ethanol. The alcohol volume was measured using an alcoholmeter.

III. RESULTS AND DISCUSSION

A. Plant Identification and Starch Content in Canna Indica Rhizome

The botanical experts at PBZT confirmed the identification of the plant as Canna indica, a member of the Cannaceae family. A detailed taxonomic classification is provided below:

Kingdom: Plantae

Subkingdom: Tracheobionta

Division: Magnoliophyta

Class: Liliopsida

Subclass: Zingiberidae

Order: Zingiberales

Family: Cannaceae

Genre: Canna

Species: indica (Linné, 1907).

Analyses conducted at the CNRE Laboratory revealed a substantial starch content in the Canna indica rhizomes. Starch constitutes 74.5% of the rhizome's dry matter: this high starch concentration suggests significant potential for ethanol production.

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B. Enzymatic hydrolysis of starch

The results of the enzymatic hydrolysis of the three types of extracts are shown in the following table1.

TABLE I. EXPERIMENTAL CONDITIONS FOR HYDROLYSIS OF CANNA INDICA EXTRACTS

Feature	Skinned dry matter	Unskinned dry matter	Fresh Matter
Nature	Flour	Flour	Thin slices
Quantity(g)	250	250	1000
Water quantity during gelation (ml)	3000	3000	3500
Gelation temperature (°C)	73-84	76-89	90
Gelation duration (min)	60	60	120
Color after gelation	Chocolate	Chocolate	Beige

Enzymatic hydrolysis of Canna indica extracts revealed significant differences among the three extract types. Fresh extracts, being thin slices, required more rigorous gelatinization conditions due to their higher moisture content and different structural properties. These extracts also necessitated larger quantities of water and initial raw material.

The final gel colors varied among extract types, indicating distinct chemical compositions. To optimize hydrolysis for each extract type, it is essential to tailor conditions to their specific characteristics. Fresh extracts may require additional pretreatment or specialized enzymes.

Chemical and microscopic analyses are necessary to gain a deeper understanding of the underlying mechanisms and further optimize hydrolysis. Factors such as starch, protein, fat, and phenolic compound content, as well as starch granule structure, likely play significant roles in the hydrolysis process.

C. Enzyme addition and gel cooking

To optimize the production of bioethanol from Canna indica rhizomes, we conducted a series of experiments by varying several key parameters during the steps of enzyme addition and gel cooking. These parameters include the quantity of enzymes added, the cooking temperature, the cooking duration, and the quantity of hydrolyzed wort obtained. These experimental conditions were carefully adjusted for each type of extract to maximize the efficiency of converting biomass into fermentable sugars. The following table summarizes the experimental conditions used for each type of extract.

TABLE II. EXPERIMENTAL CONDITIONS AND HYDROLYZED SWEET JUICE YIELD FOR CANNA INDICA RHIZOME EXTRACTS

Feature	Skinned dry matter gel	Unskinned dry matter gel	Fresh matter gel
Enzyme quantity (ml)	200	200	235
Cooking temperature (°C)	56-64	59-70	56-64
Cooking duration (min)	80	80	80
Hydrolyzed sweet juice quantity (ml)	2850	3010	3250

Hydrolysis yields differed among Canna indica extracts. Skinned dry extracts, despite identical flour and enzyme amounts, produced slightly less hydrolyzed sweet juice due to lower cooking temperatures. This suggests that higher temperatures favor starch hydrolysis.

Fresh extracts outperformed dry extracts, likely due to their increased rhizome and enzyme quantities. While cooking temperatures were similar, these additional factors may explain the higher hydrolysis yield.

C. Fermentation

After five days at a temperature varying between 27 and 31°C, the bubbling stops. This indicates that the fermentation is complete. The hydrolyzed juices are then fermented and ready for the final extraction step, distillation.

D. Distillation and alcohol yield

The volume and alcohol content obtained for each type of rhizome extract after distillation are shown in Tables III, IV, and V.

TABLE III. ETHANOL CHARACTERISTICS FROM DISTILLED JUICE OF PEELED DRY CANNA

Time (min)	**Volume (ml)**	**Alcohol Content (°)**	**Ethanol Volume (ml)**
43	85	45	38.25
11	50	17	8.5
35	80	17	13.6
16	65	13	8.45
15	70	05	3.5
		Total	**72.3**

Waste: 1908 g (1350 ml)

TABLE IV. ETHANOL CHARACTERISTICS FROM DISTILLED JUICE OF UNSKINNED DRY CANNA

Time (min)	**Volume (ml)**	**Alcohol content (°)**	**Ethanol volume (ml)**
15	72	30	21.6
19	78	15	9
25	75	12	8
15	80	10	8
17	71	08	5.68
14	67	07	4.69
21	80	05	4
		Total	**64.67**

Waste : 2150 g (1500 ml)

The following table summarizes the key characteristics of ethanol produced from the distillation of fermented juice extracted from fresh Canna.

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TABLE V. ETHANOL CHARACTERISTICS FROM FRESH CANNA JUICE

Time (min)	Volume (ml)	Alcohol content (°)	Ethanol volume (ml)
18	60	44	26.4
18	60	30	18
25	53	19	10.07
15	57	12	6.84
09	58	06	3.48
		Total	**64.79**

Waste: N/A

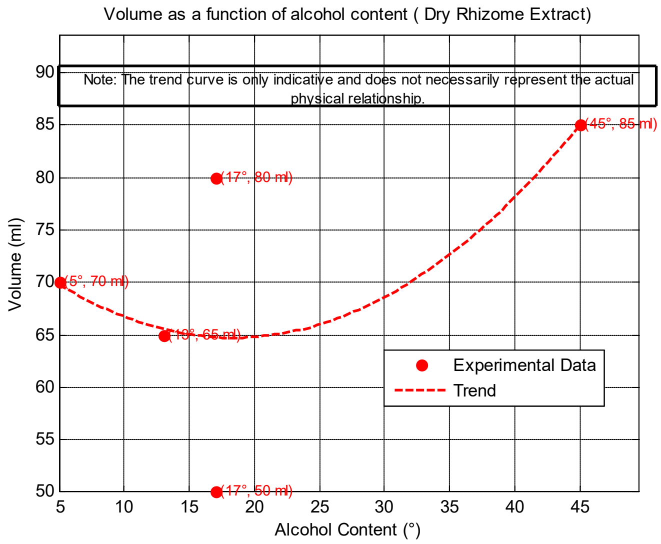

Fig.1: Alcohol Volume vs. Alcohol Content from 250 g Peeled Dry Rhizome

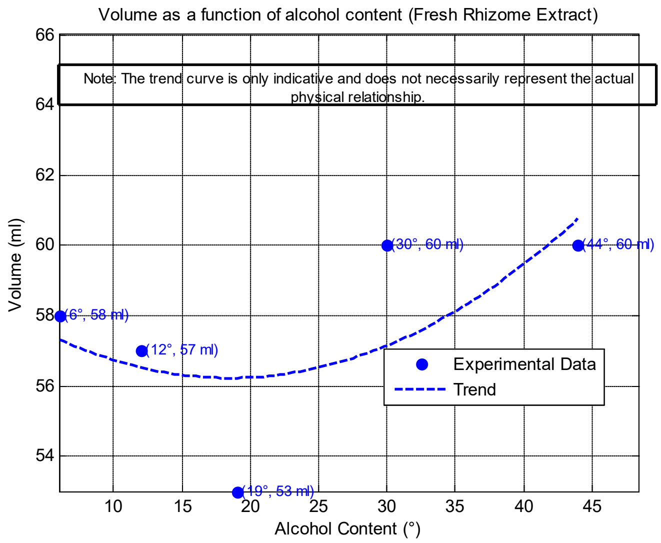

Fig. 2. Ethanol Yield vs. Alcohol Content from 1 kg Fresh Rhizome.

The table VI compares bioethanol production from fresh and dried Canna indica rhizomes, highlighting differences in yield, stability, and process complexity.

TABLE VI. COMPARISON OF BIOETHANOL YIELD FROM FRESH AND DRIED CANNA INDICA RHIZOMES AT VARYING ALCOHOLIC STRENGTHS

Feature	Dried Rhizome	Fresh Rhizome
Yield	Generally higher, especially at high alcohol degrees, 200 -340ml/kg	More stable, 55-60 ml/kg
Stability	More variable	More stable
Process complexity	More complex, requires more optimization	Less complex
Improvement potential	Higher potential, especially for high alcohol degrees	Lower potential

Fresh rhizomes yield a more consistent and predictable bioethanol product, but with limited room for optimization. In contrast, dried rhizomes offer higher yields, especially at higher alcohol concentrations, but the process is more variable and complex.

Ultimately, the choice between fresh and dried rhizomes depends on the specific production goals: those seeking a stable and reliable process may prefer fresh rhizomes, while those prioritizing high yields and exploration of new methods might opt for dried rhizomes.

B. Discussion

a. Comparison of Ethanol Yields from Different Canna Rhizome Extracts

Based on the ethanol yield results presented, the following interpretations can be made:

- Skinned Dry Rhizome Extract: This produced the highest ethanol yield of 289.2 ml per kg of flour, making it the most efficient extract for ethanol production. It requires the least amount of material (3.46 kg) to produce 1 liter of ethanol (83.53 L/ton).
- Unskinned Dry Rhizome Extract: This had a slightly lower ethanol yield of 258.68 ml per kg of flour compared to the skinned dry rhizome. It requires a bit more material (3.87 kg) to produce 1 liter of ethanol (66.84 L/ton).
- Fresh Rhizome Extract: This produced the lowest ethanol yield of 64.79 ml per kg of flour, making it the least efficient extract for ethanol production. It requires a significantly higher amount of material (15.43 kg) to produce 1 liter of ethanol (4.198 L/ton).

In terms of profitability for ethanol extraction, skinned dry Canna rhizome is the most promising option due to its higher ethanol yield and lower material requirement. However, additional considerations such as processing costs, ease of handling the different extract types, and the nature and potential use of the waste associated with each extract type could also influence overall profitability.

Overall, the results suggest that skinned dry Canna rhizome is the most efficient and cost-effective option for ethanol production from Canna rhizome. The distillation process varies based on the alcohol content obtained (Fig. 1 & Fig. 2). The volume of ethanol is variable for each step. For the dry rhizome, the maximum alcohol content is 45° with a volume of 85 ml, and for the fresh rhizome, the maximum alcohol content is 44° with a volume of 60 ml. The alcohol content obtained decreases with each subsequent distillation.

After analysis, the starch content in Canna indica is 74.5% of the dry matter. By summing up, the quantity of pure ethanol (98 to 99°) after extraction from 250 g of rhizome flour is 72.3 ml, and 64.8 ml from 1 kg of fresh rhizome. Therefore, it requires 3.5 kg of rhizome flour to obtain 1 liter of pure ethanol and 15.5 kg of fresh rhizome to obtain the same volume of pure ethanol. According to these results, the use of flour is the most efficient for extraction.

The result of the second extract is slightly lower than that obtained from the same extract of a similar plant (Canna edulis). Indeed, 11-12 kg of fresh rhizome of Canna edulis yielded 1 liter of pure ethanol [5]. This difference may be due to the use of other additives such as NPK and urea.

b. Comparison with Other Raw Materials

To provide a broader context, it is useful to compare the ethanol production from Canna indica with that from other common raw materials such as cassava and sugarcane. Preliminary findings indicate that these crops show higher ethanol yields compared to Canna indica.

For instance, cassava has a starch content that can vary between 20% and 35% of its dry weight, making it a highly efficient raw material for ethanol production. Sugarcane, with its high sugar content ranging from 10% to 15% of its fresh weight, is also very efficient for ethanol production. Preliminary data suggest that the ethanol yield from cassava can be around 180 liters per ton of fresh cassava, while sugarcane can yield up to 70 liters per ton of fresh sugarcane. In contrast, Canna rhizome yields 83.53 liters per ton.

These results highlight that cassava is the most efficient raw material for ethanol production, while sugarcane has a lower yield. The Canna rhizome falls between the two, offering an interesting but less performant alternative to cassava.

In terms of profitability and efficiency, cassava is clearly the most advantageous choice for ethanol production. However, cassava is also an important food source in Madagascar, which could pose competition issues with food production. The Canna rhizome, on the other hand, does not directly compete with food crops and presents notable potential for ethanol production.

These findings highlight the need for further optimization of the extraction process from Canna indica to make it more competitive.

c. Comparison of Ethanol Yields in Different Countries

Studies on bioethanol production from canna rhizomes have yielded varying results across different regions and methodologies.

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- India: Srivastava et al. achieved a bioethanol yield of 320 liters per ton of fresh rhizomes by combining enzymatic saccharification using cellulases and β-glucosidases with alcoholic fermentation conducted by Saccharomyces cerevisiae under mesophilic conditions [23].
- Thailand: A yield of 277 liters per ton of dry rhizomes was attained using acid saccharification followed by fermentation with Zymomonas mobilis, a thermophilic bacterium [3].
- Our study, conducted in Madagascar using the Canna indica extract, resulted in bioethanol yields of 83.53 L/ton, 66.84 L/ton, and 4.198 L/ton for skinned dry rhizome, unskinned dry rhizome, and fresh rhizomes, respectively. We employed an optimized enzymatic saccharification process followed by fermentation with Saccharomyces cerevisiae. These results surpass those reported in the literature for other canna varieties, indicating the significant potential of this species for bioethanol production in our local context.

The observed differences among the studies can be attributed to various factors, including canna variety, cultivation conditions, and pretreatment methods, choice of enzymes and microorganisms, and fermentation parameters. Our findings underscore the importance of optimizing each step of the production process to enhance the yield and cost-effectiveness of bioethanol production from canna.

d. Advantages despite Lower Yields in Madagascar

Despite the lower ethanol yields obtained in Madagascar compared to other regions, there are several advantages to using Canna indica for bioethanol production:

- Non-Competition with Food Crops: Unlike cassava and sugarcane, which are important food sources, Canna indica does not compete with food production. This makes it a more sustainable option for biofuel production.
- Adaptability: Canna indica is a robust and adaptable plant that can grow in a variety of soil conditions and climates. This makes it suitable for cultivation in different regions of Madagascar.
- Environmental Benefits: The cultivation of Canna indica can help in soil conservation and improvement, as it can prevent erosion and enhance soil fertility.
- Economic Potential: The development of a bioethanol industry based on Canna indica can create new economic opportunities for local farmers and communities. It can also reduce dependence on imported fossil fuels, contributing to energy security.
- Scalability: The production process can be scaled up to meet increasing demand, and further research can lead to improvements in yield and efficiency.

By carefully selecting the canna variety, refining cultivation techniques, and fine-tuning the pretreatment and fermentation processes, we can maximize the efficiency and sustainability of bioethanol production. This approach not only improves the economic viability of the process but also contributes to the development of a more robust and environmentally friendly biofuel industry in Madagascar. Future research should focus on further optimizing these processes and exploring additional factors that can enhance bioethanol production from canna rhizomes. Despite the lower yields, the advantages of using Canna indica make it a promising feedstock for bioethanol production in Madagascar.

IV. CONCLUSION

Our comprehensive research into ethanol production from Canna indica rhizomes has revealed promising potential for this feedstock as a viable source of bioethanol. One of the key findings is that skinned and dry canna exhibits significantly higher ethanol yields (289.2 ml/kg) compared to other processing methods (unskinned dry: 258.68 ml/kg, fresh: 64.79 ml/kg). This underscores the importance of feedstock preparation in enhancing the efficiency of bioethanol production.

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While Canna indica rhizomes may yield slightly lower ethanol yields (83.53 liters per ton) compared to cassava (180 liters per ton), but higher than sugarcane (70 liters per ton), it offers interesting potential as a complementary energy crop, as it does not compete with food crops.

To significantly improve ethanol yields from Canna indica rhizome, it is essential to optimize pretreatment methods. Techniques such as acid or mixed hydrolysis, along with the exploration of new enzymes and the use of other ferments, or the addition of fertilizers like NPK and urea, could significantly enhance the conversion of biomass into fermentable sugars. These advancements would not only increase ethanol yields but also make the process more efficient and cost-effective. For instance, previous research by Dewi et Al. has demonstrated that the addition of NPK and urea can boost ethanol yields by 33.04%.

A thorough cost-benefit analysis is necessary to determine the economic viability of ethanol production from Canna indica. This analysis should consider all aspects of the production process, from feedstock cultivation to ethanol distillation. Additionally, an assessment of the environmental impact of each process stage is crucial to ensure that the production pathway is ecologically sustainable. This includes evaluating factors such as water usage, energy consumption, and waste management.

The competitiveness of bioethanol production hinges on energy efficiency. By minimizing energy consumption through process optimization and the adoption of energy-saving technologies, producers can significantly reduce costs and improve the overall efficiency of their operations.

Canna indica presents Madagascar with a unique opportunity to develop a sustainable and competitive ethanol industry. As a non-food crop, it offers an abundant and affordable resource. To maximize its potential, it is essential to accurately quantify the available biomass, identify the most suitable local varieties, optimize production processes, and assess socioeconomic and environmental impacts. A precise assessment, particularly of the lower heating value (LHV) of each fraction, will enable the optimal utilization of this resource.

Looking ahead, an exciting perspective for future research is the exploration of bioethanol production from the flowers of Canna indica. The flowers of this plant contain significant amounts of sugars and other fermentable compounds, making them a potential valuable feedstock for ethanol production. By utilizing both the rhizomes and flowers, we could maximize the overall yield of bioethanol from this plant, thereby enhancing the economic and environmental benefits of this production pathway.

Another important avenue for improvement is the development of rustic combustion materials that can be used by households to produce their own bioethanol. By making bioethanol production accessible at the household level, we can empower local communities to become more self-sufficient and reduce their dependence on fossil fuels. This not only contributes to a more sustainable energy transition but also has the potential to improve the quality of life for many families.