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Microwave-Assisted Alkaline (NaOH) Pretreatment of Empty Palm Fruit Bunches (EPFB) in the Production of Second-Generation Bioethanol

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Abstract. The scarcity of fossil fuels drove the development of renewable energy, including second-generation bioethanol derived from lignocellulosic biomass. Empty Fruit Bunches of Oil Palm (EFBOP) represented an abundant agro-industrial waste rich in cellulose, thus showing potential as a bioethanol feedstock. This study aimed to analyze the effectiveness of the Microwave Assisted-Alkaline Pretreatment method in optimizing the conversion of EFBOP into bioethanol. The pretreatment process employed NaOH and microwave radiation to reduce lignin content, increase cellulose concentration, and decrease crystallinity levels. The study also evaluated the influence of Microwave Power (380 W, 500 W, 700 W) and alkali concentration (0.5 M, 0.6 M, 0.7 M) on lignin, cellulose, hemicellulose content, and ethanol production. The research sought to enhance the efficiency of converting EFBOP waste into renewable energy, supporting environmental sustainability and energy security. Results revealed the optimal pretreatment condition with 0.6 M NaOH concentration and 500 W microwave power, achieving a lignin reduction from 40.73% to 29.48%. The final bioethanol yield reached 14%.

Keywords : Second-Generation Bioethanol, Empty Fruit Bunches of Oil Palm (EFBOP), Microwave Assisted-Alkaline Pretreatment, Lignocellulose, Renewable Energy.

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Introduction

Scarcity of fuel, especially subsidized fuel, has begun to be felt in various regions of Indonesia. This is due to the declining natural resources that serve as raw materials for fuel. One common type of fuel can be replaced by bioethanol, a fuel produced through the fermentation of biomass [1]. In the production of first-generation bioethanol, the main raw materials come from starch-containing plants. However, since starch is a food material, the production cost of bioethanol is somewhat high. Therefore, second-generation bioethanol has been developed, which uses raw materials from lignocellulosic sources[2]. In their research, Stated that Empty Fruit Bunches of Oil Palm (EFB) are one of the sugar sources rich in cellulose, with content reaching 75-80%. Hence, EFB has great potential to be utilized as the main raw material for bioethanol production [2].

Lignocellulosic biomass is a renewable and sustainable raw material used in bioethanol production. Its abundant sources include residues from energy crops, agro-industrial products, and food processing waste. This biomass is a promising alternative to reduce environmental impacts caused by energy demand and uncertainties in oil supply. However, due to its complex structure, its conversion process requires a physical or thermochemical pretreatment stage, followed by various methods to enhance carbohydrate accessibility and reactivity. Subsequently, enzymatic hydrolysis is carried out to produce fermentable sugars, which then undergo microbial fermentation to produce bioethanol as fuel [3].

In second-generation bioethanol production based on lignocellulosic biomass waste, the initial step required is pretreatment to generate cellulose, which is then subjected to hydrolysis and fermentation to produce bioethanol. Bioethanol production from lignocellulosic materials involves pretreatment, the process begins with pretreatment as the first stage. The purpose of this stage is to remove lignin content, reduce cellulose crystallinity, increase porosity, and convert EFB into pulp. Lignin removal is an important factor in the development of commercial technology for lignocellulosic-based bioethanol production [4].

Lignin removal is a crucial factor in the development of commercial technologies for lignocellulosic-based bioethanol production[2], he addition of NaOH causes cellulose to swell. The swelling of cellulose increases the surface area of lignocellulose, leading to the breakdown and separation of lignin from cellulose, or decreasing the lignin content in lignocellulose. The addition of acid is used to hydrolyze hemicellulose and lignin, dissolving most of these components from the plant cell wall structure and enhancing enzyme accessibility to cellulose. Acid hydrolysis is done by hydrolyze ing the hemicellulose fraction while leaving cellulose and lignin intact in the residual solids. Concentrated acids such as H2SO4 or HCl can be used in this process. The drawbacks of this method include toxicity, corrosiveness, and the hazardous nature of concentrated acids [5].

Analysis of pretreatment of EFB as raw material for ethanol production showed that microwave usage was effective in increasing cellulose content from 33.84% to 36.87%, while lignin content decreased from 16.81% to 13.74%. Additionally, the analysis revealed a decrease in crystallinity at the cellulose peak after EFB was pretreated with microwave assistance, which facilitates the hydrolysis process. This decrease in crystallinity can be attributed to the effect of microwave irradiation, which generates rapid heating and molecular vibration within the biomass structure. Such effects disrupt the ordered arrangement of cellulose chains, reduce crystallinity, and consequently enhance the accessibility of enzymes during hydrolysis. In the initial treatment stage, 2% NaOH was used because alkaline pretreatment is effective in breaking down the lignocellulosic structure. The use of NaOH promotes the swelling of cellulose, reduces lignin content, and increases the porosity of the biomass, thereby improving enzyme accessibility in the subsequent hydrolysis stage. This fundamental reason explains why the present research also employed NaOH as the pretreatment reagent [2].

Higher concentrations of alkaline catalyst significantly reduced lignin content in the remaining pulp. Initially, Szarvasi biomass contained 78.4 mg lignin, equivalent to 19.6% of the total 400 mg. After microwave pretreatment with 0.6 M NaOH, lignin remaining in the pulp was only 3.3 mg (or 2.3% of total 146 mg), indicating that 96% of lignin had been hydrolyzed under these conditions. In contrast, the addition of acid catalysts did not show the same effectiveness. At the lowest concentration, 0.1 M sulfuric acid, the initial reduction of 78.4 mg lignin before pretreatment was not very significant.[6]

From the explanation above, research utilizing empty fruit bunch waste as an alternative raw material for second-generation bioethanol production has not been maximally utilized, and the use of microwave-assisted alkaline pretreatment remains relatively limited compared to conventional methods. The microwave-assisted alkaline pretreatment method offers a new approach by using microwaves as a tool to accelerate alkaline pretreatment, which can reduce time and energy required, as well as enhance effectiveness in breaking lignocellulosic bonds. Based on this background, a study entitled "Microwave Assisted-Alkaline Pretreatment of Empty Fruit Bunch (EFB) in Increasing Second-Generation Bioethanol Production" was conducted.

Experimental

Production of Bioethanol. The materials used in this study were empty palm oil bunches (EPFB), sodium hydroxide (NaOH), sulfuric acid (H_2SO_4), distilled water, and Saccharomyces cerevisiae. The main equipment included a microwave, oven, autoclave, fermenter, and distillation apparatus. Analytical instruments used were a Brix refractometer, alcohol refractometer, analytical balance, and thermometer. Standard laboratory glassware and tools were also utilized. This research was conducted at the Energy Engineering Laboratory, Sriwijaya State Polytechnic, Palembang, from March to July 2025.

EPFB Preparation. EPFB was dried, cut, and ground to obtain uniform particle size before use.

Delignification (Alkaline Pretreatment). 40 g of EPFB was mixed with NaOH solution (0.5 M, 0.6 M, and 0.7 M) in a 1:10 biomass-to-solution ratio. The mixture was heated using a microwave (370 W, 500 W, and 700 W) for 20 minutes. The sample was filtered, squeezed, and oven-dried at 60–70°C for 24 hours to obtain delignified biomass.

Lignocellulose Composition Analysis (Chesson Method). 2 g sample was refluxed in hot water (100°C, 2 h). The residue was treated with 72% H_2SO_4 (4 h, room temperature) and refluxed (100°C, 2 h). The residue was treated with 72% H_2SO_4 (4 h, room temperature), then diluted to 0.5 M and refluxed again (100°C, 2 h). The dried residue was powdered, and lignin, cellulose, and hemicellulose contents were calculated using standard equations.

$$\text{Lignin content} = \frac{d-e}{a} x\ 100 \quad (1)$$

$$\text{Cellulose content} = \frac{c-d}{a} x\ 100 \quad (2)$$

$$\text{Hemicellulose Content} = \frac{b-c}{a} x\ 100 \quad (3)$$

Notes:

There are several parameters used to measure the weight of a sample. Parameter a is the dry weight of the sample, expressed in grams. Next, b is the weight of the first sample after being refluxed with water, c is the weight of the second sample after being refluxed with H_2SO_4, d is the weight of the third sample after being soaked and refluxed with H_2SO_4 again, and e is the weight of the fourth sample after the sample has been reduced to ash, all of which are also expressed in grams.

Acid Hydrolysis. 20 g of delignified EPFB was mixed with 2.5% H_2SO_4 at a 1:10 biomass-to-solution ratio. Hydrolysis was carried out in an autoclave (121°C, 60 min). The hydrolysate was neutralized with NaOH solution.

Glucose Analysis. Glucose concentration was measured using a Brix refractometer after calibration with distilled water.

Fermentation. Neutralized hydrolysate was placed in a fermentation vessel. S. cerevisiae (5% of substrate weight) was added, and fermentation was carried out for 7 days at room temperature under anaerobic conditions.

Distillation. The fermented broth was distilled at 78–80°C to collect ethanol. Distillation was stopped near 100°C to avoid water co-distillation.

Ethanol Analysis. Ethanol concentration was measured using an alcohol refractometer after calibration.

Data Analysis. Based on the results (in Table 1) of the research conducted and Chesson Data analysis, the results of the lignocellulose content analysis in the form of lignin, cellulose, and hemicellulose content from empty palm fruit bunches (TKKS) were obtained using the microwave pretreatment method. The lignocellulose was obtained from Chesson data analysis. while the Ethanol Content Observation data is shown in Table 2.

Table 1. Results of Lignocellulose Content Observation with Variations in Microwave Power and Alkali Concentration

Sample Mass (Gram)	Time (Minutes)	*Microwave* Power (W)	NaOH Alkali Concentration (M)	Lignocellulose Content		
				Lignin	Selulosa	Hemiselulosa
40	0	0	0	40.73	34.02	14.36
40	20	380	0,5	31.2	14.6	32.20
			0,6	31.47	32	33.00
			0,7	29.82	27.6	29.20
40	20	500	0,5	33.95	34.2	28.80
			0,6	29.48	28.2	28.20
			0,7	32.8	24.7	28.40
40	20	700	0,5	30.0	21.8	27.10
			0,6	34.28	9.8	20.10
			0,7	30.74	19.12	25.80

Table 2. Results of Ethanol Content Observation

Sample No.	Sample Mass (Gram)	*Microwave Power* (W)	NaOH Alkali Concentra-tion (M)	Volume of Ethanol (mL)	Ethanol Content (%)
1	200	380	0,7	160	8
2	200	500	0,6	145	14
3	200	700	0,5	148	7

Result and Discussion

Figure 1 shows the results of the analysis of the content of empty oil palm fruit bunches, using nine samples with variations in microwave power of 380W, 500W, and 700W and alkali concentration (NaOH) of 0.5M, 0.6M, and 0.7M for each power level. As a result, the lignin content ranged from 29.48% to 34.28%. The lowest lignin content was found in the NaOH concentration of 0.6 with a microwave power of 600W, at 29.48%. The highest lignin content was found in the NaOH concentration of 0.6 with a power of 700 W, at 34.28%. Based on the analysis results, it has been proven that the pretreatment process using microwave waves successfully increases the cellulose content while reducing the lignin content. This condition facilitates the subsequent hydrolysis process [1]. Increasing the concentration of NaOH enhances the breaking of lignin bonds, thereby facilitating the removal of lignin from the lignocellulosic structure. As a result, the measured lignin content in the solid biomass decreases because part of the lignin is solubilized into the alkaline solution. However, when the NaOH concentration is excessively high, it can lead to over-degradation of lignin and partial damage to cellulose and hemicellulose, causing loss of structural carbohydrates along with dissolved lignin in the solvent [7]. The use of 0.6 M NaOH with a microwave power of 600 W can increase cellulose

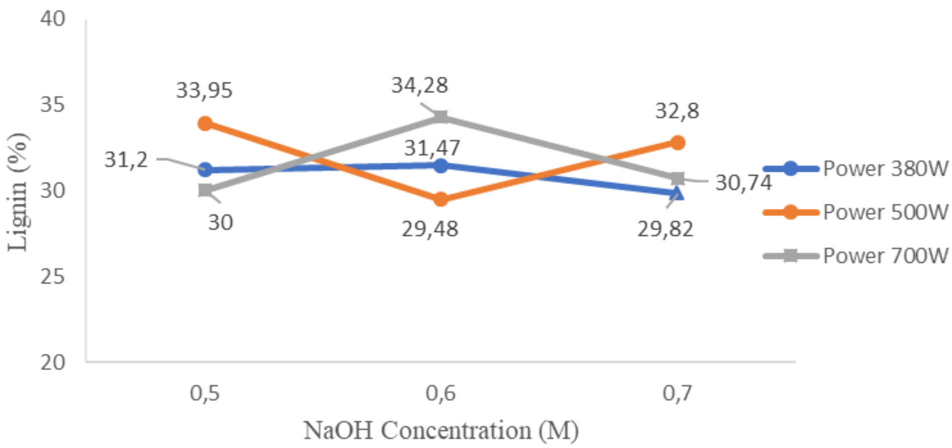

Figure 1. The Effect of NaOH Concentration and Microwave Power on Lignin Content

content and reduce lignin content more than other variations. This is because cellulose remains in the solid phase, with only a small amount in the liquid phase [8]. The reduction in lignin content in EFB using the microwave pretreatment method with 0.6 M NaOH at 500 W power achieved the highest lignin reduction compared to other variations. This is evidenced by the decrease in lignin percentage from 40.73% to 29.48%. This is because the use of a base in pretreatment can break down lignin into its monomers more effectively than other materials [9]. The use of a base, particularly sodium hydroxide (NaOH), in the pretreatment of lignocellulosic biomass is highly effective in breaking down lignin into its monomeric units. NaOH plays a crucial role by disrupting the ester and ether bonds that hold lignin polymers together, especially the aryl-ether linkages such as β-O-4 bonds, which are the most abundant in lignin's complex structure [9] This chemical action causes lignin to depolymerize into smaller, more soluble fragments, facilitating its removal from the biomass matrix.

The efficiency of this degradation process is significantly enhanced when NaOH treatment is combined with microwave radiation. Microwaves provide rapid, volumetric heating by converting electromagnetic energy into thermal energy within the material. Unlike conventional heating, microwave radiation directly interacts with polar molecules, such as water and NaOH, causing them to rotate and generate heat internally through dielectric heating [10]. This rapid heating accelerates the chemical reactions initiated by NaOH, leading to faster and more uniform lignin breakdown.

Moreover, the synergistic interaction between NaOH and microwave irradiation enhances the disruption of the lignocellulosic structure. The intense localized heating increases the solubility and mobility of lignin fragments, promoting their further breakdown into monomers. Simultaneously, the hemicellulose, which acts as a glue binding cellulose fibers, is also solubilized. As a result, the structural barriers within the biomass—primarily lignin and hemicellulose—are significantly reduced, improving the accessibility of cellulose for subsequent enzymatic hydrolysis or fermentation processes [11].

Figure 2 shows the results of the analysis of the content of empty oil palm fruit bunches, where this study used nine samples with variations in microwave power of 380W, 500W, and 700W and alkali concentration (NaOH) of 0.5M, 0.6M, and 0.7M for each power level. As a result, the cellulose content ranged from 9.8% to 34.2%. The lowest cellulose content was found at an NaOH concentration of 0.6M with a microwave power of 700W, amounting to 9.8%. The highest cellulose content was found at an NaOH concentration of 0.5M with a microwave power of 500W, amounting to 34.2%. It is estimated that pretreatment combined with electromagnetic radiation (microwave) can enhance the overall effectiveness of the pretreatment process, thereby increasing the cellulose content in the raw material [1]. The average cellulose content obtained showed a decrease, which is likely due to cellulose dissolving in the NaOH solution. The observed decrease in cellulose content during pretreatment can be attributed to the disruption of the ordered crystalline structure of cellulose, which occurs when exposed to an alkaline solvent such as NaOH. Sodium hydroxide penetrates the cellulose fibers and breaks hydrogen bonds within and between cellulose chains, leading to swelling and partial solubilization of amorphous regions. As a result, some cellulose molecules become dispersed in the NaOH

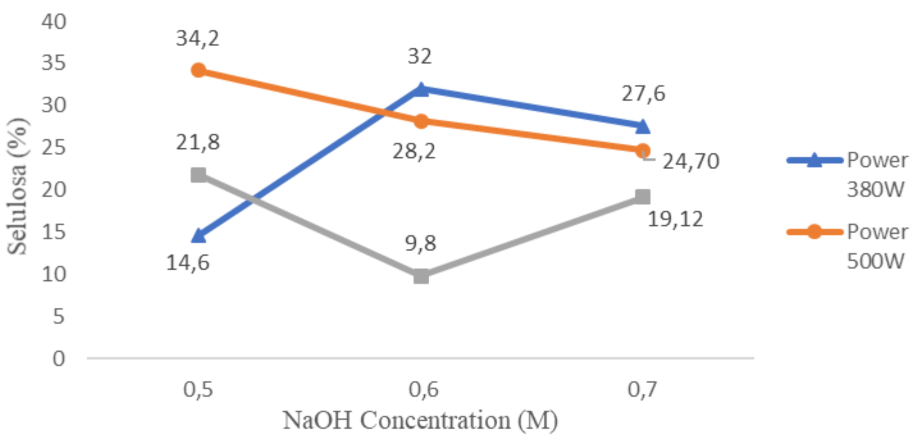

Figure 2. The Effect of NaOH Concentration and Microwave Power on Cellulose Content

solution. During the subsequent filtration step, these freely dispersed cellulose fragments may be lost along with the filtrate, leading to a measurable reduction in the solid cellulose content [12].

Conversely, an apparent increase in cellulose content may occur due to the degradation and removal of lignin and hemicellulose during pretreatment. As lignin breaks down, it releases the embedded cellulose and hemicellulose fractions that were previously bound within the lignocellulosic matrix, making more cellulose accessible and quantifiable in the residue [12]. This effect is strongly influenced by the duration of microwave irradiation. Microwave radiation induces rapid internal heating through dielectric interactions, enhancing the efficiency of the NaOH pretreatment. As the exposure time increases, more lignin and hemicellulose are degraded, leaving behind a higher proportion of cellulose in the solid residue. This selective breakdown not only improves cellulose purity but also contributes to better digestibility in subsequent processing steps [1].

Figure 3 Show The results of the analysis of empty palm fruit bunch (EPFB) samples show the effects of combined microwave-assisted alkali pretreatment. This study used nine samples, each treated with varying microwave power levels (380W, 500W, and 700W) and sodium hydroxide (NaOH) concentrations (0.5 M, 0.6 M, and 0.7 M). The hemicellulose content across all treatments ranged from 20.10% to 33%. The lowest hemicellulose content (20.10%) was observed at a NaOH concentration of 0.6 M with 700 W microwave power, while the highest (33%) was found at 0.6 M with 380 W.

Generally, increasing the NaOH concentration from 0.5 M to 0.6 M led to a decrease in hemicellulose content, particularly at the highest microwave power of 700 W, where the hemicellulose content dropped from 27.10% to 20.10%. This suggests that stronger alkali conditions, when paired with sufficient energy input, enhance the delignification and hydrolysis of hemicellulose. NaOH acts as a solvent by breaking down ester and glycosidic linkages within the hemicellulose structure, solubilizing it into the solution. This dissolution process is crucial, as it separates hemicellulose from the lignocellulosic matrix, allowing it to be removed during filtration.

However, a further increase in NaOH concentration from 0.6 M to 0.7 M resulted in an unexpected rise in hemicellulose content at all power levels. This could be due to re-precipitation of degraded fragments, incomplete solubilization at higher concentrations, or reduced efficiency of microwave absorption due to excessive ion content in the solution, which may interfere with uniform heating. Notably, the highest hemicellulose content was still observed at the lowest microwave power (380 W), indicating that insufficient energy input may not effectively disrupt hemicellulose, even under high alkali conditions.

Microwave heating plays a critical role in this process by converting electromagnetic energy into thermal energy. The rapid internal heating caused by microwave interaction with polar molecules (such as water and NaOH) leads to cell wall rupture and the breakdown of hemicellulose, which binds cellulose fibers. The combined chemical and thermal effects help loosen the lignocellulosic structure, promoting the solubilization of hemicellulose. This is essential, as hemicellulose, if retained, can

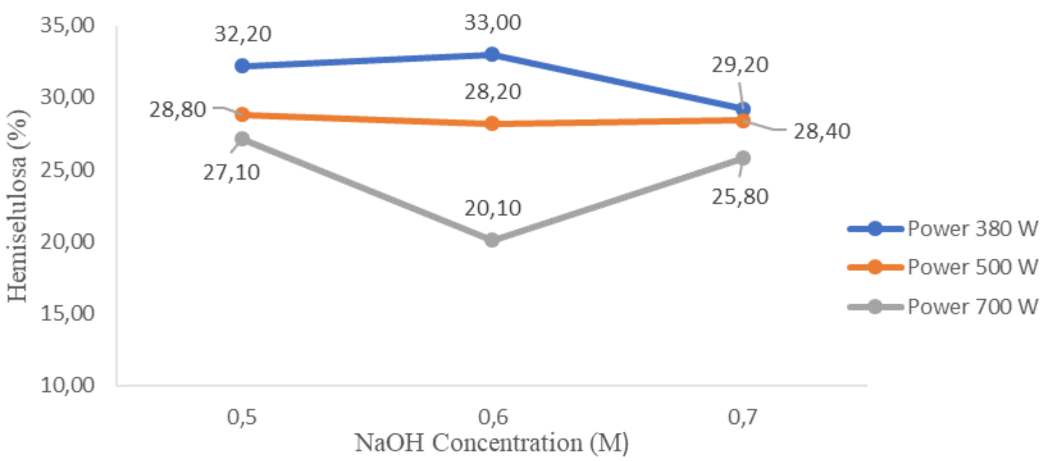

Figure 3. The Effect of NaOH Concentration and Microwave Power on Hemicellulose Content

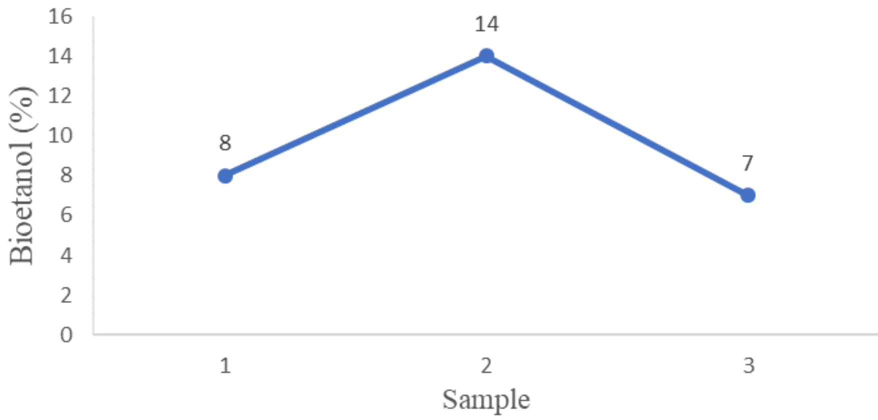

Figure 4. The effect of NaOH concentration and microwave power on bioethanol content

contribute to brittleness in the final material [13], reducing its suitability for further processing such as enzymatic hydrolysis or material reinforcement.

Figure 4 above shows the results of the analysis of the effect of NaOH concentration and microwave power on the bioethanol content produced. Thus, the ethanol content was found to be in the range of 7%–14%. The lowest ethanol content was found in sample 3, which had a NaOH concentration of 0.5 M and microwave power of 700 W, at 7%. The highest ethanol content was found in sample 2 with a NaOH concentration of 0.6 M and microwave power of 500 W, at 14%, indicating the most optimal conditions for breaking down lignocellulose structure, thereby producing high glucose content that can be fermented into bioethanol. The conversion of TKKS into bioethanol through acid hydrolysis can be hindered by lignin, which protects cellulose and hemicellulose, thereby reducing the effectiveness of the process. Therefore, pretreatment of TKKS is necessary to disrupt the lignin structure, making it easier for cellulose and hemicellulose to react more efficiently in producing bioethanol[14]. The delignification process using strong bases shows that the higher the concentration of a solution, the more molecules from that solution can help break down the lignin structure [14]. The highest reduction in lignin content and increase in cellulose content were observed in pretreatment using strong bases, specifically NaOH at a concentration of 0.6 M and microwave power of 500 W, which influenced glucose production, which is subsequently converted into ethanol.

Based on the research, the glucose produced will determine the ethanol produced. Supported by the results of the pretreatment study, the highest reducing sugar content from TKKS without pretreatment was 5.6 mg/100mL, which was obtained through hydrolysis with a concentration of 0 M H2SO4 for 15 minutes. This result is lower compared to the reducing sugar content obtained from TKKS hydrolysis with pretreatment [15]. In this study, a reducing sugar content of 6% was obtained in 2000 mL, indicating that pretreatment using 0.6 M NaOH base at 500 W power is the best treatment. This is because the prolonged contact time and high temperature during hydrolysis cause cellulose and hemicellulose to degrade more easily into glucose, resulting in a more complete hydrolysis reaction and higher ethanol production [15].

Conclusion

Based on the results of research on the Effect of NaOH Concentration and Microwave Power on Lignocellulose Content and Bioethanol Content from Empty Fruit Bunch (EFB) of Oil Palm, the following conclusions were obtained are the optimal conditions for facilitating lignin bond cleavage using microwave energy are a NaOH concentration of 0.6 M and microwave power of 500 W. This results in a reduction in lignin content from 40.73% to 29.48% and the optimal conditions for the highest bioethanol fermentation yield were obtained at 14% in sample 2, with a NaOH concentration of 0.6 M and microwave power of 500 W. The lowest bioethanol yield was in sample 3, with a NaOH concentration of 0.5 M and microwave power of 700 W, at 7%.

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Author Contributions

R.C. designed and conducted the microwave-assisted alkaline pretreatment experiments and contributed to data interpretation. P.R. performed the acid hydrolysis experiments using an autoclave and analyzed the resulting data. M.H.P. carried out the fermentation process and ethanol yield analysis. R.D., L.T., Z., and N.K. supervised the project, provided guidance throughout the research, and contributed to the refinement of the manuscript. All authors engaged in scientific discussions and approved the final version of the manuscript.

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