

Potential of Liquid Smoke from Corn Cobs as an Antioxidant Source in Mackerel Preservation

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Abstract:

This study aims to determine the antioxidant potential contained in liquid smoke using the DPPH method, as well as the effects of the concentration and soaking duration of liquid smoke from corn cobs on phenol content, moisture content, and organoleptic testing. The study first tested the antioxidant activity of liquid smoke from corn cobs by measuring the 50% Inhibition Concentration (IC₅₀) value. Subsequently, liquid smoke was used at various concentrations (0%, 5%, 7%, and 10%) with a 60-minute soaking time, and a 7-day storage period was followed by testing for phenol content, moisture content, and organoleptic properties. Data analysis was performed using SPSS.26 with ANOVA testing. The results showed an IC₅₀ value of 11.642 ppm in the antioxidant test of liquid smoke, indicating its effectiveness in neutralizing free radicals. The addition of liquid smoke to mackerel with different concentration variations affected phenol content, moisture content, and organoleptic properties. The optimal concentration of liquid smoke for use as a fish preservative is 10%.

Keywords: Liquid Smoke, Antioxidants, Mackerel, Corn Cobs.

1. Introduction

Currently, in Gorontalo, the use of corn cob waste is limited to its application as animal feed, while the remainder is often just burned, which can pose environmental pollution risks. As an alternative, one technological approach to handling corn cob waste is to apply pyrolysis techniques aimed at producing smoke condensate [1].

According to Xin et al. [2] Liquid smoke is a liquid formed through the condensation of pyrolysis products and can serve as a natural antioxidant in food processing, including in fish and its processed products. According to Sri harti et al. [3] Substances present in the liquid resulting from the pyrolysis of corn cobs include phenol compounds, aldehydes, hydrocarbons, acids, and esters.

Mackerel (*Rastrelliger sp.*) is one of the most well-known pelagic fish species with high economic value in Indonesia [4]. Mackerel faces challenges in maintaining its freshness over extended periods because it tends to spoil quickly. Daily production variability of mackerel is also an additional factor affecting this issue [5].

Fish contain compounds with significant potential for human health benefits. Due to their high protein content, fish are prone to spoilage. Fish can only last 5-8 hours in open air before beginning to emit unpleasant odors, indicating the onset of spoilage. Therefore, efforts are needed to preserve mackerel to prevent spoilage and oxidation, one of which is using smoking methods with liquid smoke from corn cobs as a natural preservative, since liquid smoke contains phenolic compounds that function as antioxidants.

According to Gulcin [6] antioxidant compounds are substances that can inhibit the oxidation process caused by free radicals. Free radicals are molecules capable of damaging cells and other biological materials through oxidation processes, which can ultimately lead to tissue damage [7].

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According to Ali et al. [9] Liquid smoke is a liquid formed through the condensation of pyrolysis products and can serve as a natural antioxidant in food processing, including in fish and its processed products. according to Zeng et al. [10] Substances present in the liquid resulting from the pyrolysis of corn cobs include phenol compounds, aldehydes, hydrocarbons, acids, and esters.

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2. Materials and Methods

This research was conducted at the Department of Chemistry Laboratory, Faculty of Mathematics and Natural Sciences, Gorontalo State University, and the Integrated Laboratory, Islamic University of Indonesia.

The equipment used in this research includes test tubes, clamps and stands, burettes, dropper pipettes, plastic containers, Erlenmeyer flasks, desiccators, ovens, porcelain dishes, Petri dishes, analytical glasses, spatulas, stirring rods, test tube racks, UV-Vis spectrophotometers, distillation apparatus, reaction tubes, smoke delivery pipes, condensers, tar collectors, pyrolysis reactor covers, liquid smoke collectors, and waste smoke discharge pipes.

Corn cobs, mackerel, DPPH solution, 0.1N KBrO₃, KBr, 3N HCl, KI, starch indicator, methanol, and Na₂S₂O₃.

2.1 The Process of Making Liquid Smoke

The first step before the burning process is to dry the corn cobs under the sun for about one day. After drying, the corn cobs are chopped into pieces measuring 5-10 cm. A total of 25 kg of corn cobs is placed into the pyrolysis reactor and sealed tightly to prevent any air from entering or escaping. The burning process is carried out for 5 hours at a temperature of 300°C. the liquid obtained from the condensation is classified as grade 3 liquid smoke.

2.2 Purification of Liquid Smoke

A sample of grade 3 liquid smoke is placed into a distillation flask that has been assembled. The flask is then sealed and a thermometer is attached. The mixture is heated to a temperature between 95-100°C. The resulting grade 2 liquid smoke has a pale yellow color with a less intense aroma.

2.3 Identification of Liquid Smoke Components

The determination of chemical components in the liquid smoke from corn cob waste is conducted using GC-MS (Gas Chromatography-Mass Spectrometry) spectrophotometer.

2.4 Antioxidant Activity Test on Liquid Smoke

The antioxidant activity test is performed using the DPPH method. A 1 ml sample of liquid smoke is mixed with 2 ml of methanol. The test mixture is then incubated at 37°C for 30 minutes in a dark place. The absorbance of the solution is measured using a UV-VIS spectrophotometer [14].

$$AA (\%) = \frac{A_0}{A_1 - A_0} \times 100 \quad (1)$$

Description:

% AA = Antioxidant Activity

A₀ = Blank Absorbance

A₁ = Sample Absorbance

2.5 Preparation of Mackerel Fish Samples

The mackerel used as a sample is washed and cleaned of any impurities, with its head and internal organs removed. After that, the fish is drained for 5 minutes and then soaked in a solution of liquid smoke from corn cobs.

2.6 Application of Liquid Smoke on Mackerel Fish

2.6.1 Determination of Optimal Concentration for Grade 2 Liquid Smoke

The cleaned mackerel is soaked in 250 ml of liquid smoke solution at concentrations of 0%, 5%, 7%, and 10%.

2.6.2 Determination of Storage Duration

The mackerel, soaked in liquid smoke at concentrations of 0%, 5%, 7%, and 10% for 60 minutes, is then stored for 1 day, 3 days, and 7 days. The analysis includes measuring phenol content, moisture content, and conducting organoleptic tests.

2.7 Phenol Content Test on Smoked Mackerel Fish

First, a 2 gram sample of mackerel is weighed and dissolved in 100 ml of distilled water in a volumetric flask. Next, 5 ml of this solution is transferred to an Erlenmeyer flask, to which 5 mL of 0.1 N KBrO₃ Solution, 0.2 grams of KBr, and 3 ml of 3 N HCl solution are added. The mixture is stirred and allowed to stand for 30 minutes, then 5 mL of KI solution is added. Following this, 8 drops of starch indicator are added, and the solution is titrated with 0.1 N thiosulfate solution until the blue color disappears [15].

$$\% \text{ Phenol} = \frac{(b-a) \times (BM_f / 6) \times 1000}{0.1 \times \text{sample weight}} \times 100 \quad (2)$$

Description :

b = Blank Volume

a = Sample Volume

BM_f = Molecular Weight of Phenol

6 = Number of Bromine Atoms Used

2.8 Moisture Content Test on Smoked Mackerel Fish

The dish is heated and then placed in an oven at 150°C for 1 hour. After heating, the dish is transferred to a desiccator and cooled to room temperature for 30 minutes, then weighed to obtain the weight of the empty dish. A 2-gram sample of untreated mackerel and a 2-gram sample of mackerel that has been soaked in liquid smoke from corn cobs are weighed. These samples are placed into the dish, and the total weight of the dish and samples is recorded. The dish containing the samples is then heated in the oven at 105°C. After heating, the dish is cooled in the desiccator. The dish and sample, which have lost moisture, are weighed again to measure the weight after heating. The heating process in the oven is repeated until the sample weight reaches a stable (constant) condition [15].

$$\text{Moisture Content (\%)} = \frac{b - c}{b - a} \times 100$$

Introduction:

- a = Weight of The Empty Cup (grams)
- b = Weight of Sample + Cup (Before Oven Drying) (grams)
- c = Weight of Sample + Cup (After Oven Drying) (grams)

2.9 Organoleptic Test on Mackerel Fish

2.9.1 Taste Test

- (a) The mackerel samples are first cooked by steaming.
- (b) Next, the mackerel samples that have been soaked in grade 2 liquid smoke are tasted.
- (c) The tasting is conducted by 15 panelists

2.9.2 Texture Test

- (a) The mackerel samples that have been soaked in grade 2 liquid smoke are touched y hand and assessed by 15 panelists.

2.9.3 Aroma Test

- (a) The mackerel samples are sniffed from a distance of ½ cm from the nose to identify the odor of samples that have been soaked in grade 2 liquid smoke
- (b) The test is conducted by 15 panelists

2.10 Data Analysis

The data from the phenol content test, moisture content test, and organoleptic tests are analyzed using statistical hypothesis testing, specifically ANOVA.

3. Result And Discussion

3.1 Analysis of Liquid Smoke

The GC-MS analysis results of the liquid smoke from corn cobs pyrolysis show 8 peaks. The GC Chromatogram of the liquid smoke is displayed in Figure 1, which shows 8 peaks, each representing the spectrum of compounds present in grade 2 liquid smoke from pyrolysis. The GC chromatogram of the corn cob liquid smoke can be seen in Figure 1, and the characteristics of grade 2 liquid smoke are presented in Table 1.

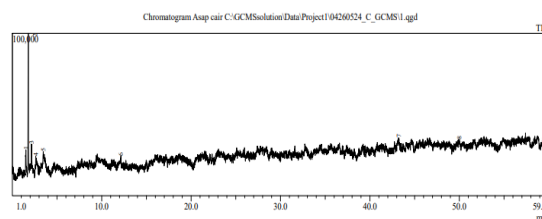
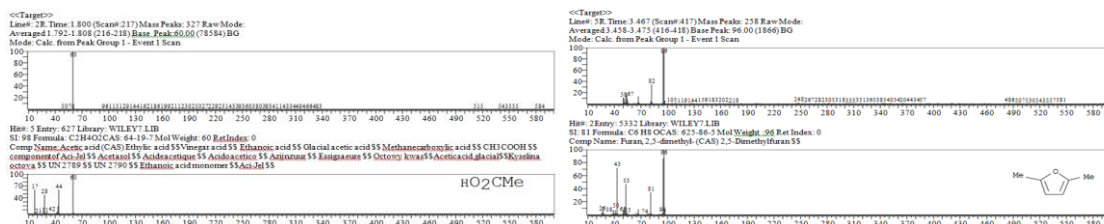


Figure 1: GC Chromatogram of Grade 2 Corn Cob Liquid Smoke



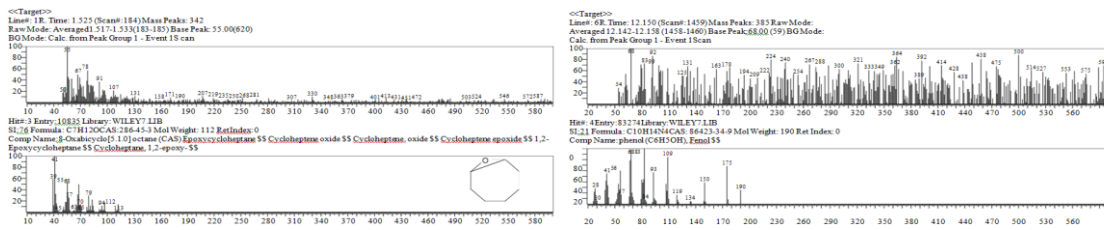


Figure 2: MS Chromatogram of Grade 2 Corn Cob Liquid Smoke

Table 1: Characteristics of Grade 2 Liquid Smoke

Peak	tR (menit)	Area %	Nama IUPAC	Molecular Formula
1.	1.522	7.46	Cycloheptene oxide	C ₇ H ₁₂ O
2.	1.797	39.08	Acetic acid	CH ₃ COOH
3.	2.150	6.76	Propanoic acid	C ₃ H ₆ O ₂
4.	2.626	11.19	1 butanol	C ₄ H ₁₀ O
5.	3.463	13.65	2,5-Dimethylfuran	C ₆ H ₈ O
6.	12.152	6.56	Phenol	C ₆ H ₆ O
7.	43.224	8.05	Pentanoic acid ethyl ester	C ₇ H ₁₄ O ₂
8.	50.001	7.23	Methyl ester	RCOOCH ₃

Based on Table 1, it can be concluded that grade 2 liquid smoke contains 8 compounds, analyzed based on retention time and different area percentages. The compound with the largest area percentage reaches 39,08% with a retention time of 1.797 minutes. This compound is acetic acid, which plays a role in extending the shelf life of products preserved with liquid smoke.

3.2 Antioxidant Activity Test of Liquid Smoke

The antioxidant activity of liquid smoke is tested to evaluate its ability to scavenge free radicals. By measuring the IC50 value, it is possible to determine how effectively the liquid smoke prevents oxidation, extended shelf life, reduces rancidity, and maintains the color stability of food products. The compounds with potential antioxidant properties in liquid smoke are phenolic compounds.

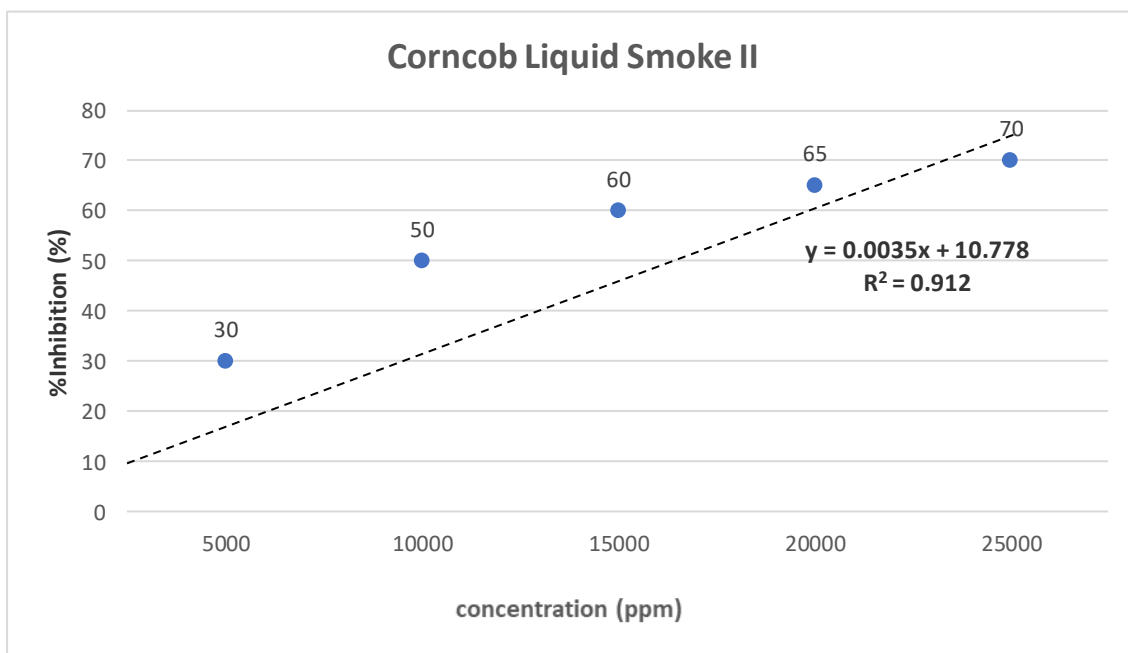


Figure 3: Antioxidant Activity Diagram (DPPH) of Corn Cob Liquid Smoke

Antioxidant activity can be measured using the 50% Inhibition Concentration (IC50) value. In the context of antioxidant activity testing of liquid smoke, IC50 (Inhibitory Concentration 50) represents the concentration of liquid smoke required to inhibit 50% of oxidative activity of free radicals in the test system El-lateef et al. [16] Each known inhibition percentage and concentration value can be plotted on a graph to derive a linear equation $y = mx + c$. By substituting y with 50 in this linear equation, we can find the x value, which represents the IC50 value of the sample.

Based on the results obtained from the linear equation in Figure 3, the IC50 value of the liquid smoke is calculated to be 11.64 ppm. A smaller IC50 value indicates a higher antioxidant capacity of the sample, demonstrating its greater ability to inhibit free radicals.

3.3 Phenol Content Test in Smoked Mackerel

Phenol is a natural antioxidant with potential benefits in food. In addition to its antioxidant function, phenol also acts as a flavor enhancer in smoked food products [17]. The analysis of phenol content aims to determine the amount of phenol absorbed by the mackerel after being soaked in liquid smoke. This information is crucial for evaluating the effectiveness of phenol as an antioxidant in the fish preservation process. The percentage of phenol content in mackerel (*Rastrelliger sp.*) is assessed.

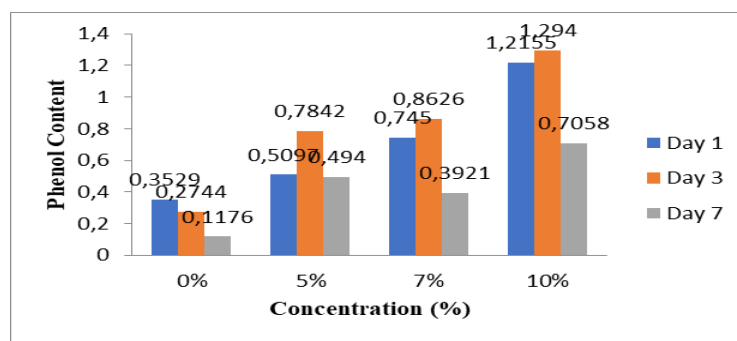


Figure 4: Bar Chart of Phenol Content Percentage in Mackerel at Different Liquid Smoke Concentrations During Storage Periods

From the observations, it is evident that all samples soaked in liquid smoke at concentrations of 5%, 7%, and 10% show significant differences in phenol content. The increase and decrease in phenol content are attributed to the storage process at room temperature, which causes phenol to evaporate from the mackerel. A similar trend was observed in the study by Chavoshizadeh et al. [15] where the phenol content in fish increased and decreased from day 1 to day 7. The control treatment also contains phenol, as the phenolic compounds present in the brine come from the sweater.

3.4 Moisture Content Test in Smoked Mackerel

Moisture content is a critical property in food because it affects appearance, texture, and flavor. High moisture levels in food can impact the shelf life of fish, as it creates an ideal environment for the growth of bacteria, mold, and yeast, potentially leading to spoilage [18]. Fish meat soaked in liquid smoke solution will experience a reduction in moisture content due to the osmosis process, where the amount of free water in the fish decreases as smoke components are absorbed.

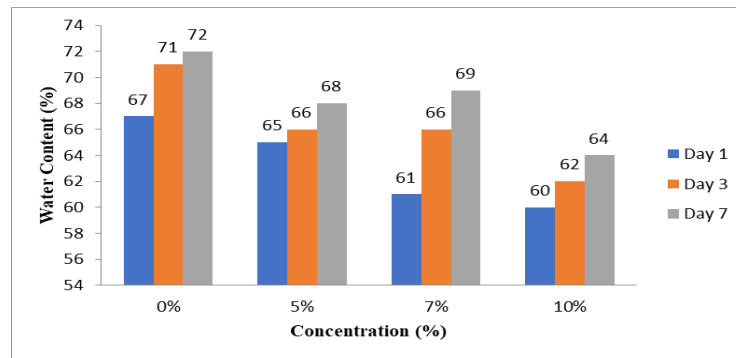


Figure 5: Bar Chart of Moisture Content Percentage In Mackerel at Different Liquid Smoke Concentrations During Storage Periods

The average moisture content of mackerel increased from the control group to the concentrations of 5%, 7%, and 10%. The study shows that the average moisture content of mackerel ranges from 60% to 72%. On days 3 and 7, the mackerel products are no longer suitable for consumption as the moisture content has exceeded the maximum limit set by the Indonesian National Standard (SNI 2725:2013), which is 60-65%.

Based on the collected data, there is a significant increase in the moisture content of smoked mackerel from the first day to the seventh day of storage. A similar trend was observed in the study by Angelovič et al. [19], where moisture content increased over the storage period. According to Song et al. [20], the increase in moisture content in smoked fish is attributed to bacterial activity that produces water during metabolic processes during storage.

However, at a concentration of 10%, there is a decrease in moisture content from day 1 to day 7 compared to concentrations of 5% and 7%. This occurs because the 10% concentration contains higher levels of active compounds such as phenols and organic acids, which accelerate the dehydration process through stronger osmotic pressure and deeper penetration into the fish tissue. This results in a more rapid loss of water from the fish cells. In contrast, at lower concentrations, the active compounds are not strong enough to cause significant dehydration, leading to an increase in moisture content due to ongoing microbial activity that produces water.

3.5 Organoleptic Testing of Smoked Mackerel

One method to assess consumer responses to the evaluated product is through sensory testing, which includes evaluating taste, odor, and texture.

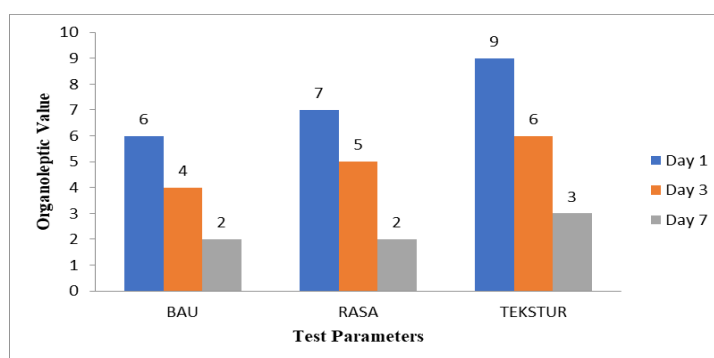


Figure 6: Bar Chart of Organoleptic Percentage of Smoked Mackerel with Liquid Smoke During Storage Periods.

Value 1-9 represent that preference parameters as follows:

1 = Strongly Dislike

6 = Somewhat Like

- 2 = Dislike Very Much 7 = Like
3 = Dislike 8 = Like Very Much
4 = Somewhat Dislike 9 = Strongly Like
5 = Neutral

3.5.1 Aroma

Aroma refers to the ability to perceive scents through our sense of smell. This sense plays a crucial role in determining whether a product is liked or disliked based on its aroma. The storage duration of smoked mackerel significantly affects the organoleptic evaluation of its smell. Based on the responses from 15 panelists, the average aroma rating of smoked mackerel is as follows: on the first day, with an interval value of 6 (Somewhat Like); on the third day, with an interval value of 4 (Somewhat Dislike); and on the seventh day, with an interval value of 2 (Dislike Very Much). According to Liu et al. [21], the presence of microorganisms on the fish also causes changes in its smell. The organoleptic rating for smell on the third and seventh days was disliked by the panelists, while on the first day, the product's aroma was considered good and favorable due to the residual smoke odor. In smoked mackerel, the decline in aroma is attributed to fungal growth, which can lead to a rancid smell.

3.5.2 Taste

Taste is a crucial factor in determining whether consumers will accept or reject a food product. Phenolic and carbonyl compounds play a significant role in defining the taste characteristics of smoked fish [22]. Based on the responses from 15 panelists, the average organoleptic taste rating for smoked mackerel is as follows: on the first day, with an interval value of 7 (Like); on the third day, with an interval value of 5 (Neutral); and on the seventh day, with an interval value of 3 (Dislike). The storage duration can alter the taste due to various chemical and physical processes occurring during storage, which can affect the organoleptic characteristics of the fish.

3.5.3 Texture

Storage duration can alter texture due to various chemical and physical processes occurring during storage, which can affect the organoleptic characteristics of the fish [23]. The highest average organoleptic rating for texture is 9 (Strongly Like), recorded on the first day of storage, while the lowest is 3 (Dislike), observed on the seventh day of storage. According to the study, the smoked mackerel sample was favored by panelists on the first day; however, by the third and seventh days, it was no longer liked. The decline in texture rating is due to increasing concentrations, which make the texture of the smoked mackerel harder over time.

4. Conclusion

Based on the result obtained, the following conclusions can be drawn Liquid smoke from corn cobs grade 2 contains 8 compounds, analyzed based on retention time and different percentage of area. The compound with the largest area is acetic acid, reaching 39.08% with a retention time of 1.797 minutes. The antioxidant activity test results indicate that the liquid smoke has a very high potential as an antioxidant. This is evidenced by the IC50 value of 11.642 ppm, which signifies its significant effectiveness in neutralizing free radicals. With this value, liquid smoke shows promise as a potential antioxidant source. Overall, the addition of liquid smoke to mackerel with varying concentrations affects phenol content, moisture content, taste, aroma, and texture.

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Authors Profile



Ishak Isa

Ishak Isa is a lecturer and professor specializing in analytical chemistry, holding both a Doctorate and Master of Science degrees. He is currently affiliated with Gorontalo State University as a Professor at the Faculty of Mathematics and Natural Sciences. In 2013, he published a research article focusing on community waste recycling, titled 'Utilization of Banana Peel Waste to Produce Ethanol through Hydrolysis and Fermentation Using *Saccharomyces cerevisiae*.' In 2019, he released a scientific work titled 'The Effect of Environmental-Based Laboratory Learning to Reduce Students' Misconceptions Through Remedial Teaching on Acid-Base Solution Material at SMA Negeri 1 Limboto.' His scholarly works are inspirational to many, especially among students and educated youth.



Windi Bukani

Windi Bukani, a student of Gorontalo State University, Faculty of Mathematics and Natural Sciences, who is pursuing an S1 in Chemistry Department. She has authored a commendable scientific article titled "Potential Of Liquid Smoke From Corn Cobs As An Antioxidant Source In Mackerel Preservation." This article aims

to explore the potential of corn cobs as an antioxidant source in liquid smoke for preserving mackerel, helping to educate the public about this preservation method.



La Ode Aman

La Ode Aman is a doctor in the Faculty of Sports and Health, currently serving as a senior lecturer at the same faculty. He obtained his bachelor's degree from Gorontalo State University in 2002, continued his academic studies at the Bandung Institute of Technology (ITB), where he completed his master's degree in 2010, and earned his doctoral degree from ITB in 2021. This year, he has published an impressive article titled "Molecular Docking and Molecular Dynamics Study of 3-Hydroxybutyrate with Polymers for Diabetic Ketoacidosis-Targeted Molecularly Imprinted Polymers."



Hendri Iyabu

Hendri Iyabu is a lecturer in the Faculty of Mathematics and Natural Sciences at Gorontalo State University. He holds the position of Head of the Department of Mathematics and Natural Sciences and also serves as a senior lecturer within the faculty. He completed his master's degree at Brawijaya University in 2011. In 2020, he conducted research titled "Empowerment of Pangi Village Community in Organic and Inorganic Waste Processing," aimed at developing a product for processing organic and inorganic waste.



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