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RESEARCH ARTICLE

Antibacterial and Structural Properties of Chitosan-Gelatin Edible Films Enhanced with Liquid Smoke from Rice Husks and Cajuput Twigs

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ABSTRACT

This study developed edible films incorporating liquid smoke derived from rice husks and cajuput twigs as both a chitosan solvent and an active antimicrobial–antioxidant agent. The fabricated films exhibited excellent mechanical integrity in accordance with Japanese Industrial Standards and possessed homogeneous microstructures, as evidenced by Fourier Transform Infrared Spectroscopy (FTIR) and Scanning Electron Microscopy (SEM) analyses. Antibacterial tests demonstrated that higher concentrations of chitosan and liquid smoke enhanced inhibition against *Escherichia coli* and *Bacillus subtilis*, with inhibition zone diameters ranging from 6.08 mm to 11.68 mm for *E. coli* and 6.05 mm to 9.18 mm for *B. subtilis*. Antibacterial activity against *Bacillus subtilis* was enhanced in edible films containing rice husk and cajuput twig liquid smoke, with increases observed alongside rising concentrations of the liquid smoke. These results suggest that liquid smoke derived from biomass enhances the mechanical and antimicrobial properties of edible films, rendering them an eco-friendly and sustainable packaging solution.

Keywords: Cajuput (*Melaleuca leucadendron*) twigs, Chitosan, Edible films, Liquid smoke, Rice husks (*Oryza sativa*)

Introduction

Edible film is a thin sheet of biopolymers such as polysaccharides, proteins, fats, or a combination of these materials. Edible film is biodegradable and is used for food packaging.^{1,2} Their significance lies in their potential to replace petroleum-based plastics, which are known for their slow decomposition and contribution to environmental pollution. As biodegradable materials, edible films provide an eco-friendly alternative that helps prevent moisture loss, reduces fat oxidation, preserves color, aroma, and taste, and inhibits microbial growth. This makes

them an ideal substitute for harmful preservatives used in food and agricultural products. In addition, edible film can reduce browning and water loss³ in post-harvest fruit to maintain its quality. The addition of antimicrobial and antioxidant compounds to the edible film manufacturing formula can extend the shelf life of the product.⁴ This dual functionality aligns with sustainability goals, offering an effective solution for food preservation and quality maintenance in an environmentally conscious manner.

Biopolymers, such as chitosan (a polysaccharide obtained by partial deacetylation of chitin⁵) and gelatin (a protein-based natural polymer, which is produced

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by partially hydrolyzing collagen from animal skin or bones, and is used for gel formation⁶), are widely used as foundational materials for film production due to their inherent antibacterial and antifungal properties, making them suitable for both individual and combined applications. Edible films consisting of chitosan and gelatin demonstrate improved antimicrobial and antioxidant activities primarily due to chitosan, whereas gelatin serves predominantly to enhance the film’s structural integrity and flexibility. Gelatin is soluble in water and functions as a stabilizer, emulsifier, thickener, and binder.^{7–9} In contrast, chitosan is a weak base and insoluble in water but easily soluble in acidic conditions.¹⁰ The solubility of chitosan directly reduces its antibacterial properties. Therefore, chemical modification is needed to enhance its effectiveness. The addition of liquid smoke into the formulation of edible films containing chitosan and gelatin improves their quality due to its acidity and contains antimicrobial and antioxidant substances. Previous studies have shown that liquid smoke is an effective preservative that prevents decay by inhibiting the growth of pathogenic bacteria and fungi.^{11,12} Furthermore, because liquid smoke contains acetic acid,^{13–15} its use in producing edible films provides additional benefits by acting as a solvent for chitosan.

Liquid smoke is produced from various biomass sources, including cocoa pods,^{13,14} rice husks,^{15,16} and cajuput twigs.¹⁵ The potential of rice husks and cajuput twigs as raw materials for liquid smoke production is significant due to the large quantities of waste they generate. Rice husks and cajuput twigs are locally abundant in many Southeast Asian regions, especially Indonesia, but are often regarded as waste materials. Their utilization aligns with circular economy principles and contributes to environmental value. This study presents a novel approach by utilizing rice husks and cajuput twigs as raw materials for liquid smoke production, which is then incorporated into starch-based edible films. However, there has been limited research on the use of liquid smoke from rice husks and cajuput twigs in the development of edible films. Liquid smoke from these sources contains compounds such as acetic acid and phenols, which exhibit antioxidant and antibacterial properties against both Gram-positive bacteria (such as *Listeria monocytogenes*, *Bacillus subtilis*, and *Staphylococcus aureus*) and Gram-negative bacteria (including *Salmonella typhimurium*, *Pseudomonas aeruginosa*, and *Escherichia coli*).¹⁵ The objective of this study was to investigate the characteristics of edible films made from a combination of two biopolymers—chitosan and gelatin—supplemented with liquid smoke derived from rice husks and cajuput twigs.

Materials and methods

Materials

Liquid smoke used in this study was derived from rice husks and cajuput twigs through pyrolysis at 500 °C for 8 hours, following established procedures.¹⁵ The liquid smoke condensation products from the 5-kg pyrolysis reactor are gathered in a container and allowed to stand for 1 day so that tar can precipitate. Aquadest was produced from a Millipore Milli-Q device. Edible bovine gelatine was purchased from PT. Brataco, Indonesia (pH=3.8-7.8). Chitosan of low molecular weight from shrimp shells was purchased from Sigma-Aldrich (DD = 97.03 %). Glycerol pro analysis 85% was purchased from Merck (Darmstadt, Germany).

Preparation of edible films

Aquadest was heated to 100 °C using a hot plate, and edible films were formulated as described in [Table 1](#). Gelatin was then added to the heated water, stirred at 250 rpm with a magnetic stirrer for 15 minutes, and allowed to cool to 50 °C. After cooling, liquid smoke and chitosan were added, followed by stirring for an additional 30 minutes. Glycerol was incorporated next, and the mixture was stirred for 10 minutes. The homogeneous solution was poured into Petri dishes and dried in an oven at 50 °C for 20 hours with a typical laboratory relative humidity of approximately 50–60% RH.

Table 1. Composition of the edible film formulation.

Composition (w/v.%)	Sp ₁	Sp ₂	Sp ₃	Rk ₁	Rk ₂	Rk ₃
Chitosan	0.20	0.40	0.60	0.20	0.40	0.60
Gelatin	8.20	8.20	8.20	8.20	8.20	8.20
Glycerol	4.92	4.92	4.92	4.92	4.92	4.92
Liquid smoke	4.92	4.92	4.92	4.92	4.92	4.92
Aquadest	81.76	81.57	81.36	81.76	81.57	81.36

Note: Sp = Rice husks, Rk = Cajuput twigs.

Characterization

Thickness, tensile strength, and elongation of each edible film were measured using a 10 KN Universal Testing Machine (Shimadzu AGS - 10kNG), following ASTM standard D882 - 18. Fourier Transform Infrared (FTIR) Spectroscopy (Thermo Scientific Nicolet iS-10) was used to identify functional groups, while X-ray fluorescence (XRF) (S2 PUMA – Bruker) was employed to determine the elemental composition of the edible films. The morphology of the edible films was analyzed using Scanning Electron Microscopy (SEM)

and Energy Dispersive X-ray (EDS) (Hitachi SU-3500 and Jeol JSM-IT200).

Antibacterial activity test

The antibacterial activity of edible films was evaluated using the Kirby-Bauer method, as outlined in previous studies.¹⁶ *E. coli* ATCC 25992 and *B. subtilis* ATCC 6633 were adjusted to a 0.5 McFarland standard and evenly swabbed onto Mueller-Hinton Agar (MHA) plates. Samples of the edible films, cut into discs with a diameter of 6 mm, were placed on the MHA media that contained the bacterial culture, along with a positive control (Gentamicin 10 µg/disc) and a negative control (distilled water). After 24 hours of incubation, the diameter of the inhibition zones formed by the bacteria around the discs was measured. Statistical evaluation was performed using one-way ANOVA and the Least Significant Difference (LSD) test, utilizing SPSS version 22 software.

Results and discussion

Properties of edible films: Thickness, tensile strength, and elongation

Table 2 indicates that the tensile strength of edible films ranges from 0.903 to 4.607 MPa. The Rk₃ sample exhibits the highest tensile strength at 4.607 MPa, while the Rk₁ sample has the lowest at 0.903 MPa. Notably, the Rk₁ sample also has the greatest thickness at 0.764 mm, which correlates with its lower tensile strength and elongation. Thickness serves as a divisor in the calculation of tensile strength, and higher concentrations of chitosan generally lead to increased tensile strength across all edible film samples. Although the tensile strength and elongation at break of the films meet the performance criteria specified in the JIS standard, the thickness of several samples—such as Rk₁, which measured 0.764 mm exceeds the JIS maximum allowable thickness of 0.25 mm. This discrepancy may be attributed to the relatively high solid content in the film-forming solution and the swelling properties of the biopolymer components. In particular, the addition of chitosan and gelatin,

both of which have high water absorption and film-forming capacities, likely contributed to the increased film thickness. While the films demonstrate satisfactory mechanical performance, it is important to note that not all samples fully comply with the thickness specifications of the JIS standard, which correlates with its lower tensile strength and elongation. This finding aligns with the results reported by Rahmasari and Yemis,¹⁷ who measured the thickness of edible films made from coconut shell-based liquid smoke combined with ginger starch to be between 0.1716 mm and 0.1730 mm. In contrast, Soazo et al.¹⁸ found that liquid smoke did not affect the thickness of the films. However, other research has indicated that the addition of liquid smoke can enhance tensile strength¹⁹ by promoting interactions and hydrogen bond formation between phenolic compounds, starch, and gelatin.²⁰

According to Sancakli et al.,²¹ gelatin was found to enhance both the tensile strength and elongation of plasticized gelatin films. However, glycerol, a commonly used plasticizer, enhances flexibility by reducing molecular tension between the components of the film matrix, which in turn decreases tensile strength.^{22,23} The samples analyzed in this study exhibited a thickness ranging from 0.410 to 0.498 mm. The addition of chitosan affects the elongation value, which ranges from 70.45% to 230.5%. In samples Sp₃ and Rk₃, there was a decrease in the elongation value because of the higher amount of chitosan added. The addition of a higher amount of chitosan causes the formation of empty spaces, which reduces the elasticity of the film. Glycerol, on the other hand, can improve elasticity by reducing hydrogen bonding and increasing the space in the matrix, thereby enhancing the flexibility of the films.²³ According to the Japanese Industrial Standard (JIS),²⁴ as shown in Table 3, which specifies a minimum tensile strength of 0.3 MPa for edible films, all samples in this study meet JIS standards for tensile strength.

Characteristics of edible films

The chemical element of each edible film was analyzed using XRF characterization, and the results

Table 2. Thickness, tensile strength, and percent elongation of edible films.

Liquid smoke	Sample	Thickness (mm)	Tensile Strength (MPa)	Elongation (%)
Rice husks	Sp ₁	0.451±0.02	2.875±0.18	135.8±16.23
	Sp ₂	0.448±0.01	3.369±0.32	165.9±34.92
	Sp ₃	0.498±0.04	3.430±0.53	151.5±39.58
Cajuput twigs	Rk ₁	0.764±0.20	0.903±0.14	70.45±34.88
	Rk ₂	0.410±0.08	3.011±0.74	230.5±53.98
	Rk ₃	0.436±0.04	4.607±0.81	142.6±9.789

Table 3. Edible films' standards according to JIS (1975).

Edible films' standards	Japanese Industrial Standard
Thickness	Max. 0.25 mm
Tensile strength	Min. 0.3 MPa
Elongation	Min. 10%

Table 4. XRF test results.

Element	Value (%)					
	Sp ₁	Sp ₂	Sp ₃	Rk ₁	Rk ₂	Rk ₃
Ca	5.7	2.8	19.5	24.5	22.3	12.5
Cl	20.1	24.4	18.9	16.8	17.1	20.3
K	0.7	0.7	0.4	0.3	0.4	0.5
Mg	2.2	2.2	2.4	2.1	2.1	2.4
P	1.3	1.2	0.8	0.8	0.8	2.2
S	22.8	21.8	16.9	15.6	15.4	18.1
Si	3.3	3.5	2.5	2.0	3.0	3.4
Fe	-	0.9	-	-	0.7	-

Table 5. Composition of components in edible films based on EDS results.

Elements	Composition (wt.%)					
	Sp ₁	Sp ₂	Sp ₃	Rk ₁	Rk ₂	Rk ₃
C	55.12	57.02	57.48	57.00	58.15	57.08
O	43.54	38.25	42.23	42.55	41.33	42.25
Mg	0.04	0.26	0.01	0.02	0.01	0.05
Si	0.01	0.47	0.02	0.06	0.07	0.11
Cl	0.12	2.42	0.17	0.19	0.23	0.26
K	0.01	-	-	-	-	-
Ca	1.14	0.50	0.03	0.11	0.09	0.11
Fe	0.02	-	0.01	0.02	-	0.05
S	-	-	0.06	0.05	0.11	0.10

are summarized in Table 4. The edible films exhibit high concentrations of elements such as calcium (Ca), chlorine (Cl), and sulfur (S), which exceed the levels of potassium (K), magnesium (Mg), phosphorus (P), and silicon (Si). Iron (Fe) is detected in low concentrations, particularly in the edible films Sp₂ (0.90%) and Rk₂ (0.70%). To further validate these findings and to examine the microstructure of the edible film, SEM and EDS analyses were conducted. The results of the EDS analysis are presented in Table 5. Notably, compared to the XRF data in Table 4, phosphorus (P) was not detected in the observed area. The EDS analysis also identified carbon (C) and oxygen (O) as the dominant elements, both of which are key components of the natural materials used to produce these edible films. Additionally, magnesium (Mg), silicon (Si), chlorine (Cl), potassium (K), and sulfur (S) were found in trace amounts. These elements are the ingredients used to make edible films, and they do not contain any contaminants.

The SEM analysis presented in Fig. 1 illustrates the surface microstructure of edible films. At 200 × magnification, the image reveals a clear microstruc-

tural view, showcasing a variety of structural forms for edible films made from rice husks and cajuput twig liquid smoke. The structures of edible films have different densities, with some appearing dense, while others are loose and wavy. The surface of each edible film is free of cracks, maintaining a flat, tight, and non-porous composition. This characteristic is crucial for achieving optimal barrier properties, as it minimizes air transfer and reduces moisture loss, ultimately enhancing the shelf life of food products.^{25,26}

FTIR spectroscopy, as shown in Fig. 2, indicates that during the formation process of the edible films, no new functional groups were generated. This observation suggests that the films made from liquid smoke, chitosan, gelatin, glycerol, and water (aquadest) are products of physical mixing rather than chemical transformation. Furthermore, the FTIR characteristics of liquid smoke derived from rice husks and cajuput twigs display wave numbers ranging from 4000 to 400 cm⁻¹. The FTIR results show identical absorption bands in liquid smoke samples from both sources. In the functional group analysis presented in Fig. 2, consistent absorption peak wavelengths are observed for each variant of the edible films (Sp₁–Sp₃ and Rk₁–Rk₃). For example, the absorption peak at 3295 cm⁻¹ in the Sp₁ sample and 3278 cm⁻¹ in the Rk₁ sample indicate the presence of the O–H group, which is indicative of phenolic compounds in the liquid smoke.²⁷ This finding supports the results of Mansur et al.,¹⁵ who reported a phenol content of 12.40% in rice husk liquid smoke and 11.52% in cajuput twig liquid smoke. All films exhibit a localized absorption peak around 3273 cm⁻¹, attributed to the presence of O–H groups.²⁸ Additionally, wave numbers of 2935 cm⁻¹, 1633 cm⁻¹, 1239 cm⁻¹, and 1035 cm⁻¹ signify the presence of C–H, C=C, amine, and carbonyl groups, respectively. The FTIR spectroscopy findings align with the presence of acetic acid (CH₃COOH) and other organic acid derivatives found in the smoke. Specifically, the liquid smoke from rice husks contains 41.13% acetic acid, while the smoke from cajuput twigs comprises 65.52% acetic acid.¹⁵ The band near 1633 cm⁻¹ is usually linked to the stretching vibration of C=O (carbonyl) groups, especially in conjugated or aromatic systems, or to C=C stretching in aromatic rings.^{29,30} The presence of this band is likely associated with the aromatic C=C stretching vibrations, which are typical in materials containing phenol or other aromatic structures from liquid smoke. The characteristics of chitosan and gelatin in the edible films are indicated by an absorption band at 1552 cm⁻¹, which signifies the presence of N–H and C–N groups. The frequency range of 900–1225 cm⁻¹, particularly at 921 cm⁻¹ and 1111 cm⁻¹, points to the presence of C–O groups. Overall, the FTIR analysis

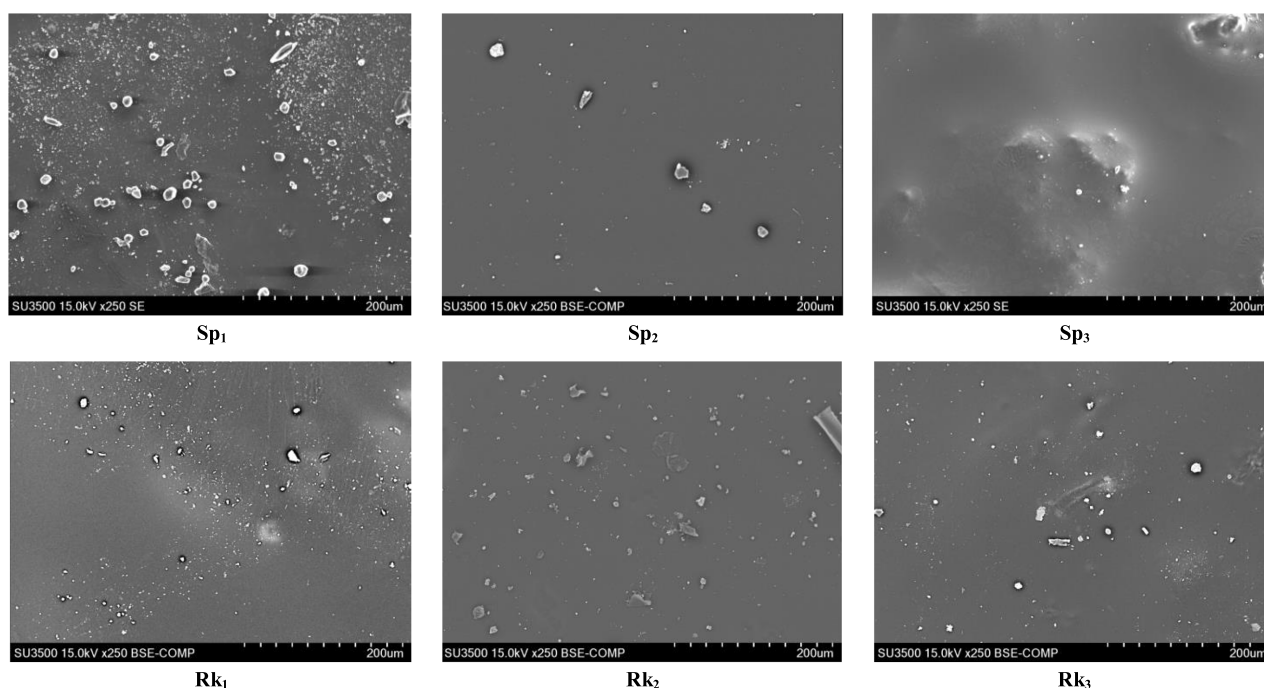


Fig. 1. Microstructure of edible films.

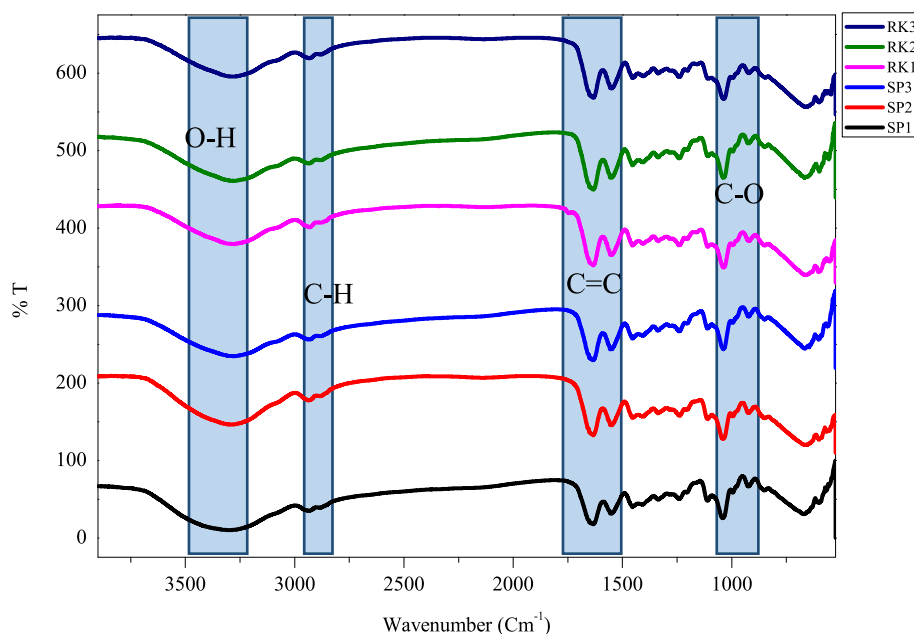


Fig. 2. FTIR spectroscopy of edible films.

of edible films, composed of liquid smoke, chitosan, glycerol, and gelatin, reveals wavelength values similar to those of the raw materials, confirming that the original chemical structures have been maintained.

Antibacterial properties of edible films

The antibacterial efficacy of edible films derived from liquid smoke produced from rice husks and ca-

juput twigs against *E. coli* is presented in Table 6. The results indicate that the diameter of the clear zone, which reflects antibacterial activity, increases with higher concentrations of chitosan. This positive correlation is consistently observed across all samples, from Sp₁ to Sp₃ and Rk₁ to Rk₃. The Rk₃ sample demonstrates the highest antibacterial activity, with a clear zone diameter of 11.68 mm, while Sp₁ exhibits the lowest at 6.08 mm. The Analysis

Table 6. The diameters of the inhibition zone of edible films against *E. coli*.

Liquid smoke	Sample	Diameter inhibitory zone (mm)
Rice husks	Sp ₁	6.08 ^A
	Sp ₂	11.25 ^B
	Sp ₃	11.31 ^B
	Control -	0
	Control +	20.31
Cajuput twigs	Rk ₁	6.12 ^A
	Rk ₂	10.92 ^B
	Rk ₃	11.68 ^B
	Control -	0
	Control +	20.49

Control +: Gentamicin, Control -: Aquadest.

of Variance (ANOVA) test reveals significant variation ($\alpha = 0.05$) in antibacterial activity against *E. coli* among the different treatments. In comparison, the negative control (distilled water) shows no bacterial inhibitory response (0 mm), while the positive control (Gentamicin) exhibits a strong inhibition zone of 20.31 mm for liquid smoke from rice husks (Sp) and 20.49 mm for liquid smoke from cajuput twigs (Rk). Gentamicin, as a broad-spectrum antibiotic, achieves a strong antibacterial efficacy category (> 20 mm).²⁹ Samples Sp₁ and Rk₁ display weak antibacterial activity (< 10 mm), measuring 6.08 mm and 6.12 mm, respectively. The interaction involving 0.20% chitosan does not yield significant bacterial inhibitory power, while 0.40% chitosan demonstrates strong inhibitory capabilities. Additionally, the inclusion of 0.60% chitosan produces an inhibitory effect comparable to that of 0.40% chitosan. This suggests that the optimal condition for inhibiting *E. coli* growth is achieved at a chitosan concentration of 0.40%. Post-hoc analysis using LSD's indicated that the inhibition zone at 0.40% chitosan was not significantly different from 0.60% chitosan ($p = 0.78$), while both were significantly higher than 0.20% and 0.30% chitosan ($p < 0.05$). The sample size for each treatment was $n = 3$. Furthermore, treatments with Sp₂, Sp₃, Rk₂, and Rk₃ exhibit moderate antibacterial activity, with respective inhibitory zone areas of 11.25 mm, 11.31 mm, 10.92 mm, and 11.68 mm, all classified in the medium category (> 10 mm).³¹

The antibacterial efficacy of edible films containing liquid smoke from rice husks and cajuput twigs against *Bacillus subtilis* was assessed by measuring the inhibition zone diameter Table 7. The findings showed that every edible film sample with liquid smoke had an inhibitory effect on *B. subtilis*, whereas the negative control displayed no inhibition zone. This confirms that the antimicrobial effect came from the components of the liquid smoke. The rice husk-based liquid smoke films exhibited inhibition zone diameters between 6.02 and 7.11 mm. While

Table 7. The diameters of the inhibition zone of edible films against *B. subtilis*.

Liquid smoke	Sample	Diameter inhibitory zone (mm)
Rice husks	Sp ₁	6.02 ^A
	Sp ₂	7.05 ^B
	Sp ₃	7.11 ^B
	Control -	0
	Control +	18.11
Cajuput twigs	Rk ₁	6.05 ^A
	Rk ₂	8.12 ^B
	Rk ₃	9.18 ^B
	Control -	0
	Control +	17.39

Sp₁ demonstrated the smallest inhibition zone (6.02 mm), Sp₂ and Sp₃ exhibited significantly enhanced antibacterial activity of 7.05 mm and 7.11 mm, respectively. The letters in superscript (A and B) represent that these differences reached statistical significance ($p < 0.05$), meaning that increasing the concentration of liquid smoke improves the antibacterial properties of the edible films. Films with cajuput twig liquid smoke also exhibited a comparable trend, showing inhibition zones between 6.05 mm and 9.18 mm. The increase in antibacterial activity from Rk₁ to Rk₃ indicates that higher concentrations of phenolic and acidic compounds in the cajuput-derived liquid smoke contributed to greater bacterial inhibition. Rk₃ demonstrated the highest antibacterial activity (9.18 mm), approaching that of the positive control (17.39 mm), thereby indicating a strong antibacterial potential.

Liquid smoke is known to possess antibacterial properties, primarily due to its composition of phenolic and acid compounds, as described by Desvita et al.³² Among these, phenol and its derivatives—such as 2-methoxyphenol, 3-methylphenol, 2-methoxy-4-methylphenol, 2-ethylphenol, and 4-ethyl-2-methoxyphenol—are the second-largest component found in liquid smoke derived from rice husks and cajuput twigs.¹⁵ These phenolic compounds result from the degradation of lignin during the pyrolysis process, while acid compounds are produced from the pyrolysis of cellulose and hemicellulose.³³ Phenol targets bacterial cell walls by disrupting peptidoglycan bonds,³⁴ which leads to incomplete formation of the cell layer. At low concentrations, phenol forms weak protein-phenol complexes that cause the denaturation of proteins, while at high concentrations, it induces cell lysis.³⁵ Phenolic compounds and their derivatives readily create protein complexes through hydrogen bonding and other non-covalent interactions with the polymer chains. In addition to phenol, acetic acid found in liquid smoke is an important antibacterial agent.^{16,36} Acetic acid destabilizes various cellular functions and

structures, enhancing its antibacterial properties.^{35,37} These interactions may affect the matrix's diffusional routes, crosslink density, and mechanical strength. Beyond liquid smoke, chitosan is effective against Gram-positive bacteria, Gram-negative bacteria, and fungi.^{38,39} Chitosan exerts its antibacterial effects by damaging intracellular components and bacterial cell membranes. This occurs through interactions between the positively charged amino groups in chitosan and the negatively charged molecules on bacterial cell membranes.^{37,38} This study's findings can be interpreted in light of known mechanisms by which acetic acid and phenolic compounds interact with polymer matrices and contribute to microbial inhibition. Acetic acid can impact the structure of the polymer network through plasticization and pH-dependent protonation effects.⁴⁰ Chitosan dissolves in acidic environments,¹⁰ because its amino groups become protonated at low pH, leading to increased solubility.⁴¹ Liquid smoke contains organic acids such as acetic acid and phenolic chemicals, which reduce the pH sufficiently to protonate the $-NH_2$ groups of chitosan, allowing it to dissolve.⁴²

Conclusion

In conclusion, the use of liquid smoke derived from rice husks and cajuput twigs in edible film formulations containing chitosan and gelatin has the potential to be developed. The combination of these three materials increases the mechanical strength and antibacterial properties of the film. The interactions between chitosan and the bioactive components of liquid smoke, especially phenolic compounds and acetic acid, greatly affect the functional performance of the films. The mechanical properties (tensile strength 0.903–4.607 MPa; elongation 70.45–230.5%) were connected with JIS standards, demonstrating that the film structure remained stable despite variations in chitosan content. The antibacterial effects against *E. coli* revealed that inhibition zones (6.08–11.68 mm) and *B. subtilis* with inhibitory zones ranged from 6.05 to 9.18 mm. This expanded as chitosan concentrations increased, in line with the increased availability of protonated amino groups. While films with 0.40% chitosan demonstrated a practical balance of mechanical strength and antimicrobial efficiency, those containing 0.60% chitosan exhibited comparable or slightly enhanced inhibition. This suggests that the optimal formulation likely falls within this range. Further statistical validation, including analysis of variance (ANOVA) and p-value assessment (<0.05), was performed to confirm whether the observed differences among the

variables were statistically significant. These findings highlight the effectiveness of biomass-derived liquid smoke in manufacturing edible films. This study also encourages the exploration of liquid smoke from other biomass sources in edible film applications according to the availability of different raw materials. Thus, the use of liquid smoke in edible films is an innovative and sustainable food preservation solution. Further research is recommended to study a broader range of antimicrobial activity and evaluate the quality of the films under various storage and environmental conditions.

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Authors' declaration

- Conflicts of Interest: None.
- We hereby confirm that all the Figures and Tables in the manuscript are ours. Furthermore, any Figures and images that are not ours have been included with the necessary permission for republication, which is attached to the manuscript.
- No animal studies are present in the manuscript.
- No human studies are present in the manuscript.
- Ethical Clearance: The project was approved by the local ethical committee at the National Research and Innovation Agency, BJ. Habibie Science and Technopark, Serpong, South Tangerang, Banten 15314, Indonesia.

Authors' contributions statement

M. F. Supervision, Validation, Investigation, and Writing-review & editing. D. M. Conceptualization, Investigation, Supervision, Validation, Writing-original draft. H. D. Conceptualization, Methodology, Data curation, and Writing-original draft, and Writing-review & editing.

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الخصائص المضادة للبكتيريا والبنية لأغشية الكيتوزان-الجيلاتين الصالحة للأكل المعززة بالدخان السائل المستخلص من قشور الأرز وأغصان الكاجوبوت

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الخلاصة

طوّرت هذه الدراسة أغشية صالحة للأكل تتضمن دخانًا سائلًا مستخلصًا من قشور الأرز وأغصان الكاجوبوت، وذلك باعتباره مذيبيًا للكيتوزان وفي الوقت نفسه عاملاً فعالاً مضاداً للميكروبات ومضاداً للأكسدة. أظهرت الأغشية المُحضّرة سلامة ميكانيكية ممتازة وفقاً لمعايير المواصفات الصناعية اليابانية، كما امتلكت تراكيب مجهرية متجانسة، وهو ما أكدته تحاليل مطيافية الأشعة تحت الحمراء بتحويل فورييه (FTIR) والمجهر الإلكتروني الماسح (SEM). أظهرت الاختبارات المضادة للبكتيريا أن زيادة تراكيز الكيتوزان والدخان السائل أدت إلى تعزيز فعالية التثبيط ضد كل من *Bacillus subtilis* و *Escherichia coli*، حيث تراوحت أقطار مناطق التثبيط بين 6.08 مم و 11.68 مم بالنسبة لـ *E. coli*، وبين 6.05 مم و 9.18 مم بالنسبة لـ *B. subtilis*. كما زادت الفعالية المضادة للبكتيريا ضد *Bacillus subtilis* في الأغشية الصالحة للأكل المحتوية على دخان سائل مستخلص من قشور الأرز وأغصان الكاجوبوت، مع ملاحظة ارتفاع في التأثير بالتزامن مع زيادة تركيز الدخان السائل. تشير هذه النتائج إلى أن الدخان السائل المستخلص من الكتلة الحيوية يعزز الخصائص الميكانيكية والمضادة للميكروبات للأغشية الصالحة للأكل، مما يجعلها حلاً مستداماً وصديقاً للبيئة في مجال التغليف.

الكلمات المفتاحية: أغصان الكاجوبوت (*Melaleuca leucadendron*)، الكيتوزان، الأغشية الصالحة للأكل، الدخان السائل، قشور الأرز (*Oryza sativa*).