



Valorization of Rice Panicle Straw and Watermelon Rind as an Innovative Medium for Eco-Friendly Production of SCOBY Biofilm

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ABSTRACT

The valorization of agro-food waste represents a promising pathway toward sustainable bio-based materials. This study aimed to formulate a fermentation medium from rice panicle straw (merang) and watermelon rind for producing eco-friendly SCOBY (Symbiotic Culture of Bacteria and Yeast) biofilm. The waste materials were thermally extracted without filtration to retain active solids and subsequently fermented with lactic acid bacteria for 14 days. The fermentation broth was enriched with soybean flour as a nitrogen source and Ziziphus mauritiana leaf extract as an antioxidant additive. Phytochemical screening was conducted using colorimetric reactions analyzed through chromaticity coordinates for flavonoids and grayscale intensity for alkaloids. SCOBY growth was evaluated by measuring biofilm thickness, wet and dry weight, tensile strength, and elongation at break. The results revealed that the combination of these residues provided sufficient nutrients and bioactive compounds to support SCOBY development. The best treatment produced a biofilm with 4.51 mm thickness, 17.49 g wet weight, 3.86 g dry weight, 43.11 MPa tensile strength, and 6.66% elongation. Colorimetric evaluation showed a red chromaticity spectrum for flavonoid presence (dominant wavelength 620 – 625 nm, purity 81.04%), and a clear white grayscale range (220 – 240) for alkaloids. In conclusion, this formulation offers a simple, low-cost approach to converting agricultural waste into functional biofilms. Further investigations are recommended to evaluate the shelf life and potential of liquid SCOBY as a plant immunomodulator and biofilm as organic packaging.

Keywords: agro-food waste, chromaticity, fermentation medium, SCOBY, valorization

1. INTRODUCTION

Agro-industrial waste management is one of the most pressing issues in developing countries, where large volumes of agricultural residues remain underutilized or improperly disposed. Among various strategies for waste mitigation, valorization of agro-food residues into value-added products offers a sustainable solution to reduce environmental burden while supporting green innovation. In this context, microbial fermentation using waste-derived substrates emerges as a promising approach for producing biodegradable materials such as SCOBY



(symbiotic culture of bacteria and yeast) biofilm, which has received increasing attention for its applications in food packaging, agriculture, and environmental bioremediation.

Rice panicle straw (*merang*) and watermelon rind are abundant by-products of rice farming and fruit processing, respectively. Although frequently discarded or burnt (Masullo, 2017 and Harindintwali et al., 2020), both materials contain residual carbon and nutrients that may support microbial metabolism (Walling et al., 2019). However, their use as a combined medium for fermentative SCOBY biofilm production has rarely been investigated. Most prior studies focus on conventional substrates such as sweetened tea (de Miranda et al., 2022) or fruit extracts (Chong et al., 2023), leaving a research gap in the utilization of low-cost lignocellulosic and fruit-waste-based media (non-tea substrates) for symbiotic microbial cultures (Zulaikha Sudin & Azila, 2024).

Understanding the biochemical environment of the fermentation medium is essential to optimizing SCOBY development. Phytochemical components such as flavonoids and alkaloids may influence microbial growth, and their presence can be semi-quantitatively assessed through color-based screening techniques, including chromaticity and grayscale analysis. These visual indicators serve as a non-destructive tool for estimating the relative concentration of active compounds in fortified media.

This study aims to formulate an innovative fermentation medium using rice panicle straw and watermelon rind, evaluate its potential to support SCOBY biofilm growth, and analyze the colorimetric profile of phytochemical content through visual and digital color analysis. The integration of waste valorization and phytochemical screening provides a novel, low-cost, and eco-conscious strategy for advancing microbial biopolymer research.

Agro-food waste valorization has become a critical component of circular bioeconomy strategies, especially in the context of climate change and resource scarcity. Valorization refers to the transformation of waste into higher-value products, reducing the burden on landfills and promoting sustainable material flows (Roy et al., 2023). Agricultural by-products such as straw, husks, peels, and rinds are rich in lignocellulosic materials, carbohydrates, and phenolic compounds, making them suitable for microbial bioconversion processes (Xu et al., 2019) and (Li & Wilkins, 2021). However, their effective use depends on the use of appropriate pre-treatment and formulation to enhance bioavailability and microbial compatibility.

Rice panicle straw, or *merang*, is a lesser-utilized portion of rice residues that supports the grain structure but is often overlooked compared to general rice straw. It contains fibrous cellulose, hemicellulose, and trace minerals that can serve as carbon sources in fermentation media (Hussain et al., 2019); and (Dörsam et al., 2017). Similarly, watermelon rind, though commonly discarded,

contains pectin, reducing sugars, and bioactive compounds such as citrulline and polyphenols (Access, 2023); (Vinhas et al., 2015); and (Balogun & Kang, 2024). These biochemical constituents make both materials promising candidates for low-cost microbial substrates, especially when properly fortified or combined.

Symbiotic culture of bacteria and yeast (SCOBY) forms a cellulose-rich biofilm during acetic acid fermentation, commonly used in kombucha tea production (Zulaikha Sudin & Azila, 2024). The biofilm is mainly composed of bacterial cellulose, synthesized by acetic acid bacteria in collaboration with yeast that ferments sugars to ethanol and carbon dioxide (Carvalho et al., 2022); and (Xu et al., 2021). The quality of SCOBY biofilm, including its thickness and wet mass, is heavily influenced by the fermentation medium's nutrient content and pH balance. Innovative media derived from agro-waste offer economic and environmental advantages, but require careful evaluation of microbial growth support and material consistency (Castagna et al., 2025) and (Sivakumar et al., 2022).

Phytochemicals such as flavonoids and alkaloids play a role not only in the bioactivity of fermented products but also in shaping the microbial ecology of the fermentation process. Rapid screening of these compounds can be performed using qualitative colorimetric reactions, with color changes serving as indicators of specific metabolite groups (Maheshwaran et al., 2024). The integration of chromaticity analysis Commission Internationale d'Eclairage (CIE 1931 diagram) and grayscale intensity based on RGB (Red-Green-Blue) values has recently been explored as a semi-quantitative approach to estimate compound concentration from visual data (Jorge & Silva, 2017); (Fernandes et al., 2020); and (Phuangsaibai et al., 2021). These methods offer non-invasive alternatives to early-stage screening, particularly useful when instrumental analysis is limited.

2. MATERIALS AND METHODS

Thermal extraction of phytochemicals and minerals from agro-food waste

The main materials used in this study consisted of agro-industrial lignocellulosic waste in the form of *Oryza sativa* panicle straw (locally referred to as *merang*) and *Citrullus lanatus* rind (watermelon rind). Both were cleaned, dried, and ground into a fine paste to enhance surface area for extraction. Distilled water was employed as the universal solvent in thermal extraction. Fermentation was initiated using a 5% (v/v) inoculum of lactic acid bacteria (LAB), with sucrose (5% w/v) added to enhance microbial activity during the 14-day incubation period at ambient temperature ($\pm 25^{\circ}\text{C}$). To improve the nutritional profile of the medium, soybean flour (80 mesh)

was incorporated as a nitrogen source, while a thermally extracted filtrate of *Ziziphus mauritiana* (jujube) leaves was used as an antioxidant and antibacterial enhancer.

The raw materials used in this study were rice panicle straw (*merang*) and watermelon rind, mixed at a 1:1 ratio (w/w). The samples were ground into a paste, then subjected to thermal extraction with distilled water at a solid-to-liquid ratio of 1:10 (w/v). Extraction was conducted at 100°C for 20 minutes under intermittent stirring (5-minute intervals). The extraction pH was maintained at neutral (pH 6 – 7) to prevent degradation of heat- and pH-sensitive bioactive compounds such as flavonoids and alkaloids. This process aimed to solubilize phytochemicals and mineral nutrients (especially K and Mg), yielding a crude extract with suspended solids for further lactic acid fermentation.

Biofilm properties were assessed by measuring wet and dry weights, thickness using a Vernier caliper, and mechanical strength (tensile strength and elongation at break) using a universal testing machine (UTM – ASTM D 882). For colorimetric analysis, a digital camera and calibrated color analysis software were used to extract chromaticity coordinates (x, y) mapped onto the CIE 1931 diagram, and interpreted using purity and wavelength metrics (Kutz, 2016). Grayscale analysis was applied in the case of non-chromatic or whitish sediment for alkaloid identification.

Lactic acid fermentation (LAF) to produce LAB filtrate

Unfiltered crude extract was subjected to lactic acid fermentation (LAF) by inoculating 5% (v/v) lactic acid bacteria (LAB) starter culture. Sucrose was added to a final concentration of 5% (w/v) to support bacterial growth. The fermentation was carried out at room temperature (25°C) for 14 days. The filtrate, enriched with organic acids (primarily lactic acid), served as the fermentation medium for SCOBY growth.

Preparation of fermentation media for SCOBY growth

The BAL filtrate was supplemented with soybean flour (80 mesh) at various concentrations: 1:0.125, 1:0.250, and 1:0.375 (v/b), and sugar at 1:10 (w/v). Each treatment also received 100 ml/500 ml of thermal extract from *Ziziphus mauritiana* (bidara leaves), which contributed antibacterial, antioxidant, and nutritional enhancement properties. The formulation consists of 4 levels as follows:

- F₀ (control): BAL extract only (no soybean flour, no bidara extract).
- F₁: F₀ + soybean flour (1:0.125) + 50 g sugar/500 ml.
- F₂: F₀ + soybean flour (1:0.250) + 75 g sugar/500 ml + bidara extract.
- F₃: F₀ + soybean flour (1:0.375) + 100 g sugar/500 ml + bidara extract.

SCOBY fermentation and biofilm production

Each formulation was inoculated with 5% (v/v) SCOBY starter culture and incubated at room temperature ($\pm 25^{\circ}\text{C}$) for 14 days. The resulting biofilms were harvested and analyzed for thickness, wet and dry weight, tensile strength, and elongation at break. These physical-mechanical parameters served to assess the performance of the valorized fermentation medium in supporting sustainable SCOBY biofilm production.

Phytochemical color-based screening

Qualitative phytochemical analysis was conducted using standard colorimetric assays: 1) Flavonoids were detected using the Wilstatter reaction, indicated by reddish coloration. 2) Alkaloids were identified via the Mayer test, characterized by the formation of clear white precipitates. The visual results were further interpreted using the CIE 1931 chromaticity diagram (for chromophoric compounds) and grayscale RGB analysis (for non-chromophoric precipitates) to infer concentration levels through color intensity and purity semi-quantitatively.

The SCOBY (Symbiotic Culture of Bacteria and Yeast) was sourced from a previously fermented kombucha culture and served as the starter for biofilm production. Colorimetric phytochemical screening was conducted using Mayer's reagent to detect alkaloids (white precipitate) and the Wilstatter reaction to detect flavonoids (colorimetric red shift). All extractions were performed using a hot-plate magnetic stirrer equipped with temperature control, with contact times and stirring cycles optimized based on preliminary trials. The pH of the medium was maintained in the neutral range (6 – 7) to prevent degradation of flavonoids and alkaloids.

Phytochemical screening was conducted to identify key bioactive compounds, namely flavonoids and alkaloids through visual colorimetric reactions. The Wilstatter test method was used to identify flavonoids, indicated by the formation of a reddish solution upon reaction, while Mayer's reagent was used to detect alkaloids, indicated by the formation of a white precipitate. Following the development of color, the visible spectral properties were analyzed using a chromaticity-based method. The colors observed in the screening reactions were captured under standardized lighting and plotted on the CIE 1931 xy chromaticity diagram (Figures 1 & 2).

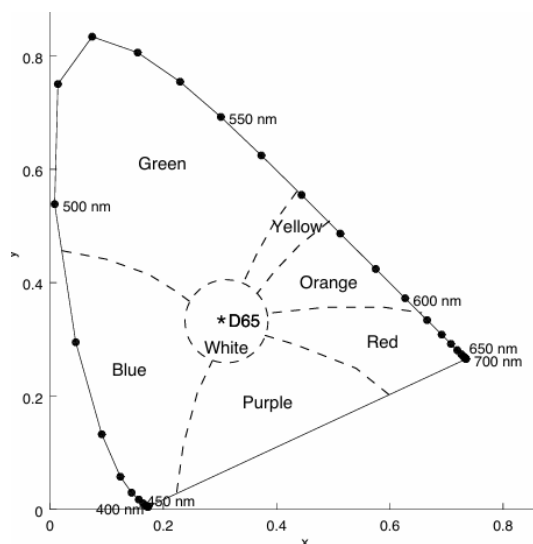


Figure 1. CIE 1931 chromaticity diagram (xy) with the D65 point marks the standard white illuminant (Centore, 2020)

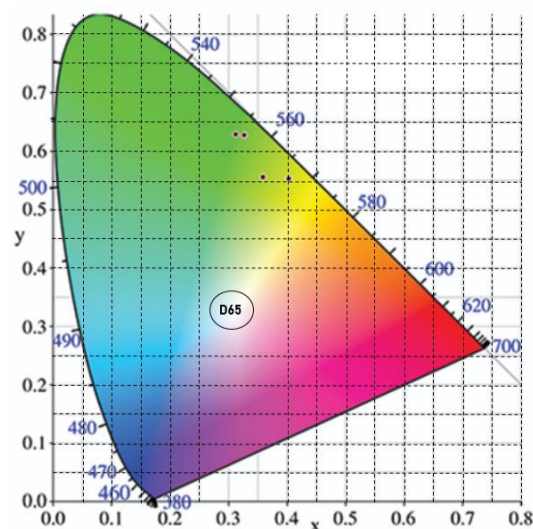


Figure 2. CIE 1931 color chart (xy) chromaticity diagram based on the visible spectrum with a wavelength scale (Zuo, 2019), and (Rosetta et al., 2020)

Figure 1. The CIE 1931 chromaticity diagram illustrates color representation based on x and y chromaticity coordinates. The outer boundary defines the spectral locus of pure spectral colors, with corresponding dominant wavelengths labeled in nanometers. The D65 point marks the standard white illuminant. This diagram is commonly used to determine both dominant wavelength and color purity in phytochemical screening based on visible color responses.

Figure 2. The CIE 1931 chromaticity diagram presents the distribution of human color perception mapped through x and y chromaticity coordinates. The horseshoe-shaped boundary delineates pure spectral hues, each annotated with its dominant wavelength in nanometers. The central white region corresponds to the standard illuminant D65, while radial lines represent isohue trajectories (hue angles). This visual model supports analytical interpretation of colorimetric results, particularly in color-based phytochemical identification.

In the context of chromaticity, the CIE 1931 xy diagram provides a visual representation of color perception, with the peripheral spectral locus representing highly saturated, pure hues, and saturation decreasing gradually toward the center. Colors that appear achromatic, such as white or neutral grays, are clustered near the central region of the diagram. This central zone is typically associated with the standard illuminant D65, which has chromaticity coordinates at approximately ($x = 0.31$, $y = 0.33$) (Centore, 2020). These principles are essential in interpreting visual outputs

from phytochemical screening, particularly when assessing intensity or purity of colors derived from bioactive compounds.

In digital image analysis, grayscale intensity represents the brightness level of each pixel and typically ranges from 0 to 255 in 8-bit images, where 0 indicates absolute black and 255 indicates pure white (Padmavathi & Thangadurai, 2016). Grayscale values are derived from the RGB color model, which is based on the additive combination of red, green, and blue light (Prabowo, 2020). White is represented as (255,255,255) – (Figure 3), and black is defined as (0,0,0) (Wang & Mogos, 2021).



Figure 3. RGB color model diagram showing additive color mixing (Prabowo, 2020).

This diagram shows the principle of additive mixing of primary colors of light: red, green, and blue. The mixture of the three primary colors in the middle produces white, while the mix of two primary colors produces secondary colors, such as yellow (R+G),

magenta (R+B), and cyan (G+B). This image is relevant to explain the basic theory of RGB to grayscale color conversion in digital image analysis.

The chromatic coordinates (x, y) of the resulting color were extracted using digital imaging and color analysis software. To quantify the color purity (saturation), the following vector-based calculation was applied (equations 1, 2, and 3) (Rosetta et al., 2020). Purity calculation for point (x, y); to calculate the color saturation (purity), the following equation is used:

Formula to calculate the distance from W to P (sample color)

$$d_{W \rightarrow P} = \sqrt{(x_P - x_W)^2 + (y_P - y_W)^2} \dots \dots \dots (1)$$

Formula to calculate the distance from W to S (point on the spectral locus in the same direction)

$$d_{W \rightarrow S} = \sqrt{(x_S - x_W)^2 + (y_S - y_W)^2} \dots \dots \dots (2)$$

$$\text{Purity} = \frac{\text{Distance (W} \rightarrow \text{P)}}{\text{Distance (W} \rightarrow \text{S)}} \times 100\% \dots \dots \dots (3)$$

Where, W = refers to the coordinates of the standard white point D65 ($x = 0.3127$, $y = 0.3290$); P = represents the chromaticity coordinates of the observed sample color ($x = 0.625$, $y = 0.290$); and S = denotes the point on the spectral locus that lies along the line extending from the white point through point P, i.e., the intersection with the chromaticity boundary (x_S , y_S). The spectral locus (point S) is determined using CIE chromaticity data. This calculation is applied to evaluate the color purity (saturation) of the colored solution derived from phytochemical extraction.

3. RESULTS AND DISCUSSION

Visual colorimetry in phytochemical analysis

In phytochemical screening, color development serves not only as a qualitative indicator but also offers quantitative insight into the concentration of bioactive compounds. Colorimetric observations commonly used in classical tests, such as the Wilstatter reaction for flavonoids and the Mayer test for alkaloids, can be objectively interpreted through digital color models. Among these, the CIE 1931 (x, y) chromaticity diagram has emerged as a robust tool for mapping color stimuli perceived by the human eye based on standardized spectral distributions.

The chromaticity coordinates (x, y), extracted from the RGB values of the reaction product, enable researchers to locate the color point within the CIE diagram, thus allowing the determination of the dominant wavelength and color purity. These metrics are beneficial for comparing phytochemical extract intensities across treatments and formulations.

Figures 4 and 5 illustrate how chromaticity data can be visualized and analyzed using this diagram. They not only capture the spectral position of sample colors but also provide a geometrical framework to compute saturation or purity, an indicator of compound concentration. This methodology bridges the gap between subjective visual scoring and quantitative color science, enhancing the reproducibility and analytical depth of color-based phytochemical assays.

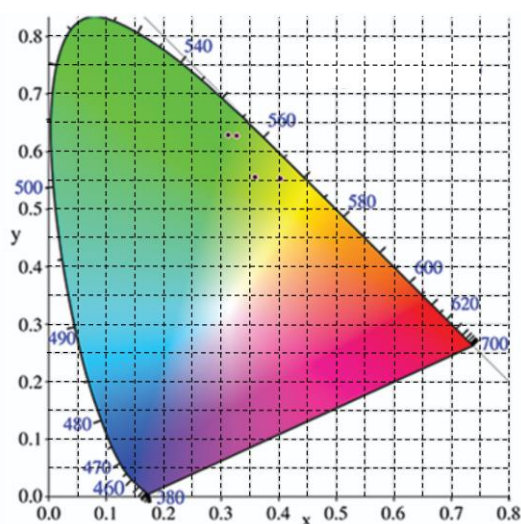


Figure 4. CIE 1931 (xy) colored diagram with coordinate grid and spectral wavelength labels

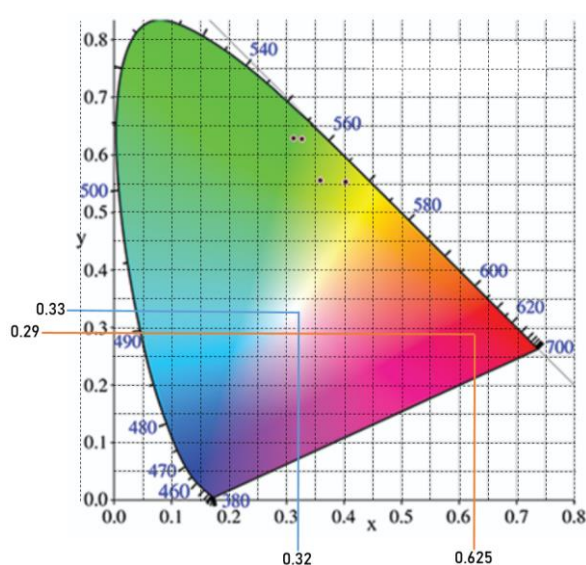


Figure 5. CIE 1931 diagram with color coordinate marking and reference lines for purity calculation (added by the author)

(modified by the author)

(Zuo, 2019), and (Rosetta et al., 2020)

(Zuo, 2019), and (Rosetta et al., 2020)

Figure 4 illustrates the full-range color representation within the CIE 1931 chromaticity space, where the horseshoe boundary defines pure spectral hues annotated with their respective dominant wavelengths (in nm). The overlaid x–y grid enables precise visual mapping and interpolation of colors derived from phytochemical reactions, thereby supporting accurate identification of dominant wavelength and calculation of color purity. Figure 5 presents an overlay chart featuring sample coordinates from phytochemical screening, including: a) Horizontal and vertical guide lines at the sample point ($x = 0.625$, $y = 0.29$); b) The standard white point (D65, $x = 0.3127$, $y = 0.3290$) (G. Wang et al., 2021) and (Zhu et al., 2022); and c) Intersection with the spectral locus (dominant wavelength point). These elements form a geometric construct enabling purity calculation defined as the ratio of the distance from white point to sample ($W \rightarrow P$) over the distance from white point to spectral locus ($W \rightarrow S$), which quantifies saturation level. A higher purity percentage indicates a more chromatic (less diluted) solution, correlating to greater chromophore concentration.

Integration with phytochemical screening

In practical application, the red shift observed during the test for flavonoids plotted on this diagram ($x = 0.625$, $y = 0.29$) corresponds to high purity (80%), suggesting a strong presence of flavonoid chromophores in the extract. For colorless or white precipitates resulting from Mayer's alkaloid test, a similar chromaticity approach is less meaningful; instead, grayscale analysis ($G = R = B$) is employed to quantify brightness, a proxy for non-chromatic compound concentration. This dual methodology aligns with modern colorimetric sensor approaches, where chromaticity and grayscale provide complementary insights (Fernandes et al., 2020), color purity for chromophoric compounds, and luminance-based analysis for achromatic substances.

Following the chromatic representation discussed in the CIE 1931 chromaticity diagram (Figures 3 and 4), further interpretation is required to link specific color responses to the presence of bioactive compounds. In this study, two color-based phytochemical tests were employed, the Wilstatter test, which detects flavonoids through the formation of a red chromophore, and the Mayer test, which identifies alkaloids via the appearance of white precipitates. These color reactions were analyzed by plotting their chromaticity coordinates (x , y) and grayscale values to quantify their optical characteristics. A summary of the chromatic and achromatic data obtained from this study, providing insight into the concentration and type of phytochemicals present in the tested extracts, is presented (Table 1).



Table 1. Chromaticity and grayscale-based interpretation of phytochemical screening color reactions using Wilstatter^{*)} and Mayer^{**)} test methods.

Color	x	y	Dominant wavelength (nm)	Purity (%)	Interpretation
Red	0.625	0.29	620 – 625	81.04	Flavonoid (red color spectrum ^{*)})
Color	x	y	Grayscale range		Interpretation
White	0.32	0.33	220 – 240		Alkaloid (clear white sediment ^{**)})

Identification of secondary metabolites through color-based phytochemical screening provides a rapid and informative preliminary method for detecting bioactive compounds in plant extracts. This study employed two distinct yet complementary approaches: (i) chromatic evaluation using the CIE 1931 chromaticity diagram for Wilstatter's reaction targeting flavonoids, and (ii) achromatic grayscale evaluation for Mayer's test, commonly applied to detect alkaloids.

The red color coordinates resulting from the Wilstatter reaction were mapped at $x = 0.625$ and $y = 0.29$, located on the right spectral limb of the chromaticity horseshoe curve. This position corresponds to a dominant wavelength in the range of 620–625 nm, characteristic of the red spectrum typically associated with reduced flavonol compounds. A calculated color purity of 81.04% indicates a high chromatic saturation, suggesting the color is close to a pure spectral hue. This further implies a substantial concentration of flavonoids within the extract.

The result aligns with the reaction mechanism of the Wilstatter test, in which magnesium ions facilitate the reduction of flavonoid carbonyl groups under acidic conditions, forming red chromophores indicative of conjugated aromatic systems (Aribowo et al., 2021). Thus, the colorimetric data corroborate the qualitative identification of flavonoid presence and offer a semi-quantitative insight into its relative abundance.

In contrast, the detection of alkaloids using Mayer's reagent produced a distinct white precipitate, which cannot be represented through chromaticity analysis, as the color coordinates are located near the neutral white point D65 ($x = 0.32$; $y = 0.33$) with very low purity values. In this context, the white color is achromatic and is more appropriately analyzed using grayscale values derived from the RGB color model, which indicates visual brightness or luminance.

In this study, a grayscale range of 220 – 240 was applied to classify the intensity of clearly defined white precipitates. This range was established based on the standard RGB-to-grayscale (Table 2), luminance conversion using the formula (4) (Padmavathi & Thangadurai, 2016):



Table 2. RGB grayscale conversion for assessment of sediment intensity (alkaloids)

Visual	Grayscale	Score	Interpretation
Very strong	> 240	*****	Thick white sediment (high alkaloids)
Strong	220 – 240	****	Clear sediment
Moderate	200 – 220	***	Moderate sediment
Weak	180 – 200	**	Thin sediment
Almost none	< 180	*	Very weak sediment

$$Y_{\text{gray}} = 0.299R + 0.587G + 0.114B \dots \dots \dots (4)$$

as recommended by the ITU-R BT.601 standard for digital visual representation (Setiawan & Faisal, 2020). A high grayscale intensity reflects a significant level of alkaloid content, even though no dominant wavelength is associated with the response.

This combined analytical approach is crucial, considering that not all chemical reactions produce chromatic outputs interpretable via dominant wavelength. For reactions yielding achromatic visual changes, such as white, turbid, or clear precipitates, grayscale analysis provides a practical and quantitative alternative. This study contributes to the development of a semi-quantitative visual classification system for the preliminary evaluation of bioactive compounds based on digital color analysis, particularly in scenarios where reference standards are unavailable or spectrophotometric instrumentation is not employed.

The next stage of the study focused on evaluating the effectiveness of the formulated fermentation media in producing SCOBY-derived biofilm. The functionalization of the medium through the integration of rice panicle straw (merang), watermelon rind, soybean flour, and *Ziziphus mauritiana* (bidara) extract was expected to enhance microbial cellulose biosynthesis. This section presents the morphological and mechanical properties of the resulting biofilms, providing insights into how nutrient enrichment affects the structural quality and performance of the SCOBY matrix. Table 3 summarizes key parameters, including thickness, wet and dry weight, tensile strength, and elongation at break, which collectively determine the suitability of the biofilm for potential biodegradable applications.

Table 3. SCOBY media formulation from food waste rice panicle straw (*merang*), and watermelon rind, and biofilm results.

Treatment	Thickness biofilm (mm)	Wet weight (g)	Dry weight (g)	Tensile strength (MPa)	Elongation at break (%)
F ₀	2.86±0.23	11.30±0.90	2.33±0.19	25.20±3.78	2.78±0.50
F ₁	3.33±0.27	13.51±1.06	2.83±0.16	31.92±4.68	3.72±0.68
F ₂	3.77±0.59	15.73±1.34	3.41±0.47	38.00±5.70	6.06±1.12
F ₃	4.51±0.09	17.49±1.22	3.86±0.31	43.11±6.08	6.66±1.18



Table 3 presents the physical and mechanical characteristics of SCOBY biofilm produced using fermentation media formulated from agro-food waste, specifically rice panicle straw (*merang*) and watermelon rind. The treatments (F_0 to F_3) represent increasing levels of nutritional fortification, particularly from soybean flour and *Ziziphus mauritiana* (bidara) leaf extract (La Torre et al., 2024), alongside a constant sugar source.

An increasing trend is observed in biofilm thickness, with F_0 (control) yielding a mean thickness of 2.86 mm, while F_3 (the highest fortification) resulted in 4.51 mm. This progressive enhancement is attributed to the presence of protein and mineral content from soybean flour and bioactive compounds from bidara leaves, which support the growth of acetic acid bacteria and yeast during SCOBY formation (Nguyen et al., 2015); (Lee et al., 2024); (Birhanu, 2021); (Ogodo et al., 2018); (Huang et al., 2018); (Sukhikh et al., 2022); and (Situmeang et al., 2022). The biofilm matrix becomes denser due to improved microbial proliferation and extracellular polysaccharide secretion (Karygianni et al., 2020).

The wet and dry weights of the films also significantly increased, indicating a higher biomass yield. F_3 exhibited the highest dry mass at 3.86 g, suggesting better substrate conversion and structural accumulation. Notably, mechanical properties improved in parallel, with tensile strength rising from 25.20 MPa (F_0) to 43.11 MPa (F_3), and elongation at break increasing from 2.78% to 6.66%. These findings reflect enhanced polymer cross-linking and matrix integrity, potentially due to protein integration from soybean and polyphenol-stabilized microbial cellulose networks.

Such biofilms possess mechanical robustness comparable to conventional biodegradable polymers and may serve as sustainable packaging materials. Furthermore, the combined valorization of lignocellulosic and fruit-processing waste offers a low-cost and environmentally conscious platform for microbial biopolymer production (Gebre et al., 2023); (Lucarini et al., 2021); and (Sarker et al., 2023).

4. CONCLUSION

This study demonstrated that the valorization of agro-food residues, specifically rice panicle straw (*merang*) and watermelon rind, can be effectively applied as fermentation media for the production of SCOBY biofilms. Incorporating thermal-based phytochemical extraction, lactic acid fermentation, and nutrient fortification with soybean flour and *Ziziphus mauritiana* extract, a nutrient-rich medium was formulated to support microbial symbiosis. Chromaticity-based

colorimetric screening and grayscale analysis successfully confirmed the presence of flavonoids and alkaloids, serving as early indicators of bioactive compound availability.

The addition of nutrient and antioxidant components significantly enhanced the mechanical properties of the resulting SCOBY biofilms. A progressive increase in thickness, wet and dry weight, tensile strength, and elongation at break was observed with each level of formulation, reaching a tensile strength of 43.11 MPa and elongation above 6.6% at the highest treatment level. These findings support the potential of using low-cost, lignocellulosic food waste in biotechnological processes to generate biodegradable, high-performance biofilms.

The approach presented herein offers a simple, integrative, and eco-friendly strategy to upcycle agricultural residues into functional biomaterials, aligning with circular economy principles and sustainable material innovation. Future research should explore the potential of LAB fermentation residues as chelating agents for heavy metal pollutants and microplastics in soil/planting media, the use of SCOBY solutions for immunomodulation in plants, livestock, fisheries, and the potential applications of biofilm derivatives as vegan leather or organic packaging.

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