

Development of a Potential Prebiotic Film Based on Sago Flour and Kepok Plantain Peel Starch with Prebiotic Properties to Support *Lactobacillus plantarum* Growth

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Abstract

Innovative prebiotic carriers that can be directly integrated into foods are increasingly sought for functional food development. This study developed prebiotic starch films using sago flour and starch from kepok plantain (*Musa paradisiaca*) peel, designed as intrinsic prebiotic components rather than packaging. Films were formulated with 1%, 3%, and 5% plantain peel starch and characterized for physicochemical properties, morphology, and their prebiotic effect on *Lactobacillus plantarum*. Higher starch concentrations increased film thickness (0.13–0.18 mm) and moisture content (11.42–16.24%), while significantly decreasing water solubility (55.31–41.14%) ($p < 0.05$). Tensile strength was low (0.17–0.27 MPa) and elongation high (89.28–178.10%). FTIR confirmed polysaccharide functional groups; SEM revealed heterogeneous fibrous structures. Resistant starch in films (1.89–3.76%) was lower than raw starch (38.91%) due to gelatinization. The 5% starch film supported the highest *L. plantarum* viability (26×10^8 CFU/mL), compared to 3% (23×10^8 CFU/mL) and 1% (21×10^8 CFU/mL), demonstrating that plantain peel starch acts as an effective intrinsic prebiotic without commercial prebiotics. The composite film shows potential as active packaging or a probiotic carrier in functional foods, although mechanical and water barrier optimization is still required.

Keywords

Prebiotic Film, Kepok Plantain Peel Starch, *Lactobacillus plantarum*, Probiotic Viability, Sago Starch

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1. INTRODUCTION

The increasing consumer awareness of the link between diet and health has prompted the food industry to develop functional foods that provide specific physiological benefits, particularly through prebiotics (Precup et al., 2022). Prebiotics are substances that are selectively utilized by host microorganisms and offer health benefits. Their incorporation into convenient, ready-to-eat formats is in high demand (Ji et al., 2023). Starch-based films present a unique advantage: they are fully digestible, can be produced from underutilized agricultural resources, and act as edible carriers for bioactive compounds, eliminating the need for synthetic packaging materials. Unlike traditional edible films that are primarily designed for food preservation (Valdés et al., 2017), starch films intended for direct consumption as functional food components must retain their structural integrity during storage while also promoting the release of prebiotic substrates in the gastrointestinal tract (Vilela et al.,

2026).

The raw material widely used for such edible prebiotic matrices is starch because it is easily absorbed by the body and has biodegradable properties (Karnwal et al., 2025b). The advantages of starch-based films include their ability to protect products from oxygen and carbon dioxide, as well as their good mechanical properties (Karnwal et al., 2025a). Banana peels, especially those from plantains (*Musa paradisiaca*), are agricultural waste with great potential due to their high carbohydrate content, including starch (Zaini et al., 2022; Anggraeni et al., 2025). Ripe plantain peels also contain bioactive compounds such as flavonoids, phenolics, saponins, alkaloids, and tannins that can inhibit pathogenic bacteria (Sa'diyah et al., 2024), as well as cellulose fiber in a sufficiently high percentage (Serratos et al., 2025). A comprehensive study analyzed by Elvinna and Sadek (2024) with the chemical composition and functional properties of banana peel flour from ambon, kepok, and cavendish varieties reported that kepok banana

peel exhibited the highest total dietary fiber (TDF) content at 47.96% compared to the other varieties. These comparative study and quantitative compositional (Table 1) advantages justify the selection of kepok banana peel starch as the functional component in the development of prebiotic films in this study.

Recent advances in starch-based films have demonstrated their potential as platforms for both prebiotic incorporation and probiotic delivery, yet a critical research gap persists. [Urango and Silva \(2025\)](#) systematically investigated the incorporation of prebiotic dietary fibers (β -glucan, citrus pectin, and inulin) into potato starch-based films, reporting enhanced mechanical properties and functional characteristics suitable for food applications. In parallel, [Yaghoubi et al. \(2025\)](#) highlighted the potential of probiotic-incorporated active packaging solutions for meat products in a comprehensive review, emphasizing that such systems can extend shelf life while delivering health benefits to consumers. Furthermore, [Shoukat et al. \(2024\)](#) explored starch-based encapsulation to enhance probiotic viability under simulated gastrointestinal conditions, achieving 85% encapsulation efficiency and significantly improved probiotic stability. These studies collectively underscore the promising role of starch-based films in prebiotic incorporation and probiotic protection.

An overview of previous research reveals a significant focus on developing and characterizing carrier probiotic films from various starch sources including banana peels and other materials with emphasis on their mechanical, barrier, viability prebiotic and basic antimicrobial properties. Research by [Rahma et al. \(2025\)](#) successfully developed pregelatinized cassava starch-based edible films as carriers for *Bacillus coagulans* demonstrating that probiotic viability could be maintained around 8 log CFU/g after 90 days of storage at room temperature. Similarly on the other side, [Coimbra et al. \(2023\)](#) investigated starch-based edible films enriched with agrifood residues (quince, potato, and orange peels) as potential carriers for *Lactobacillus rhamnosus*, reporting that films containing agrifood residues exhibited a slower loss of probiotic viability during storage compared to plain starch films, attributed to the presence of antioxidant compounds. Furthermore, studies have explored the prebiotic potential of resistant starch from different origins, noting its fermentability by gut microbiota to promote health-beneficial metabolites like SCFAs ([Munir et al., 2024](#); [Shin et al., 2023](#)). However, some researchers only focus on integrating prebiotic functions directly into an prebiotic film matrix to create active probiotic deliveries. Moreover, no one has quantitatively investigated its effectiveness as a prebiotic substrate to stimulate the viability of specific probiotics such as *Lactobacillus plantarum*.

The current study addresses two research gaps: first, to develop a composite prebiotic starch film using sago flour and banana peel starch, and the second to quantitatively demonstrate the effectiveness of the film as a natural prebiotic substrate without the addition of commercial prebiotics by evaluating its ability to enhance the viability of *Lactobacillus plantarum*. This approach shifts the focus to active and consumable functional

food ingredients, offering innovative and sustainable methods to deliver prebiotics derived from agricultural waste. Therefore, this study pioneers the development of a composite prebiotic starch film made from sago flour and kepok plantain peel starch. It quantitatively demonstrates the film's intrinsic prebiotic functionality by enhancing the viability of *Lactobacillus plantarum*, all without the addition of any commercial prebiotic compounds.

2. EXPERIMENTAL SECTION

2.1 Extraction of Plantain Peel Starch

This procedure was based on [Kaur et al. \(2022\)](#) with modifications. Plantain peel samples were obtained from fried food vendors on Sultan Mansyur Street, Ilir Barat I District, Palembang City. The plantains were chosen when they were yellow, based on ripeness considerations. Prepare about 10 kg of plantain peels. The plantain peels are separated from the fruit, washed thoroughly, and then cut into small pieces. Next, the plantain peel is mashed using a blender, and water with ratio 1:2 respectively is added to facilitate the crushing process. The crushed plantain peel was strained through a cheesecloth and transferred to a clean container. The filtered pulp is mixed with water and filtered again to remove any remaining starch until the filtered water appears clear. The filtrate obtained was allowed to settle for 24 hours at room temperature until the starch settled completely below the surface. The supernatant liquid was discarded, and the starch sediment was dried in an oven at $\pm 50^{\circ}\text{C}$ until dry. The dried starch sediment was ground and sieved to produce starch of uniform size.

2.2 Procedure and Formula for Prebiotic Film Production

The prebiotic film formula from sago flour and plantain peel starch is based on that used by [Putri et al. \(2023\)](#), with slight modifications. The prebiotic film solution was made by mixing 3% sago flour with varying concentrations of banana peel starch (1, 3, and 5%) into a beaker containing 50 mL distilled water. The solution mixture was homogenized and filtered using cheesecloth with. Next, the starch solution was heated to $\pm 70^{\circ}\text{C}$ for 25 minutes and stirred with a stirrer. Subsequently, 1% (w/v) carboxymethyl cellulose (CMC), 3% (v/v) glycerol were added to the solution while stirring continuously at ± 300 rpm for 10 minutes until fully homogeneous. The total volume of each film-forming solution was adjusted approximately 100 mL with distilled water. After homogenization, the solution was poured into Petri dishes and dried at 55°C in an oven for 14 hours. Before analysis, the prebiotic film was stored in a desiccator. The table composition of formula variation prebiotic film was shown in Table 2 based on [Rahmawati et al. \(2025\)](#) with slightly modification.

2.3 Testing the Physical, Mechanical, and Morphological Properties of Prebiotic Films

Evaluation of the physical and mechanical properties of prebiotic films includes thickness, tensile strength, elongation, moisture content, and water solubility testings with each procedures performed triplicate. The film thickness was measured using

Table 1. Proximate Analysis, Dietary Fiber Fractions, Phenolic Compounds, and Mineral Composition of Banana Peel

Component	Value	Reference
<i>Proximate</i>		
Carbohydrate	61.34 ± 0.37%	Khamsaw et al. (2024)
Protein	3.76 ± 0.12%	Khamsaw et al. (2024)
Lipid	5.22 ± 0.11%	Khamsaw et al. (2024)
<i>Dietary Fiber</i>		
Crude Fiber	16.56 ± 0.04%	Khamsaw et al. (2024)
Total Dietary Fiber	~ 43–50%	Emaga et al. (2008)
Pectin	Up to 20%	Emaga et al. (2008)
<i>Phenolic Compound</i>		
(+)-catechin	Detected	Khamsaw et al. (2024)
Gallic acid	Detected	Khamsaw et al. (2024)
Ellagic acid	16.19 mg/100g DE	Behiry et al. (2019)
Rutin	973.08 mg/100g DE	Behiry et al. (2019)
Myricetin	11.52 mg/100g DE	Behiry et al. (2019)
Naringenin	8.47 mg/100g DE	Behiry et al. (2019)
<i>Mineral/Ash</i>		
Ash	8.20 ± 0.15%	Khamsaw et al. (2024)
Potassium (K)	4.39 ± 0.15 mg/100g	Hassan et al. (2018)
Calcium (Ca)	59.10 ± 0.85 mg/100g	Hassan et al. (2018)
Phosphor (P)	211.30 ± 1.24 mg/100g	Hassan et al. (2018)
Moisture Content	4.93 ± 0.04%	Khamsaw et al. (2024)

Table 2. Formulation of Prebiotic Film Variation

Material	Formulation		
	F1 (1%)	F2 (3%)	F3 (5%)
Sago Flour (g)	3	3	3
CMC (g)	1	1	1
Glycerol (mL)	3	3	3
Plantain Peel Starch (g)	1	3	5
Aquadest (mL)	Ad 92	Ad 90	Ad 88

a digital micrometer 0–25 mm (Mitutoyo 547-526s) with an accuracy of 0.01 mm at five different points, and the average value was reported (Ratna et al., 2024). Tensile strength and elongation measurements are specifically conducted in the integrated BRIN laboratory, in accordance with ASTM D882 standards. This process uses the Shimadzu AGS-X Universal Testing Machine (UTM), equipped with a 100N load cell and pneumatic film grip, as well as a 0.001 mm micrometer and a 10 mm die cutting strip (Sinaga et al., 2020). Water content was determined using the gravimetric method, where a 1 g sample was dried in an oven at 105°C until constant weight was achieved, and the weight loss was calculated as the percentage of water content (Pak et al., 2020). Meanwhile, water solubility was tested by soaking 0.1 g of the sample in 50 mL of distilled water for 24 hours; the undissolved film portion was then filtered, dried at 105°C for one hour, and the weight difference was used to calculate the percentage of solubility (Indrianti et al., 2018). Statistical analysis was performed using

one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT) as a post hoc test to determine significant differences among mean values at a 95% confidence level ($p < 0.05$). Statistical computations were carried out using SPSS Statistics version 25 (IBM Corp., Armonk, NY, USA). The prebiotic film was characterized using Scanning Electron Microscopy analysis (SEM-EDX; FEI Quanta 250) to examine its surface morphology and by Fourier Transform Infrared spectroscopy (FTIR; Shimadzu IR Prestige-21) to identify functional groups and molecular interactions between film components.

2.4 Resistant Starch Test with AOAC Multienzyme Method

The enzymatic digestibility of the prebiotic film and control was analyzed using a sequential enzymatic hydrolysis method adapted from Do et al. (2022) with major modification. Briefly, samples (0.5 g) were suspended in 25 mL of 0.1 M phosphate buffer (pH 7) and hydrolyzed with 0.1 mL of α -amylase (Sigma Aldrich) at 100°C for 15 minutes. After cooling, the mixture was acidified with 5 mL of 1N HCl and digested with 1 mL of 1% pepsin at 40°C for 1 hour. Subsequently, the pH was neutralized with 5 mL of 1 N NaOH, and hydrolysis was continued with 0.1 mL of β -amylase (Sigma Aldrich) at 40°C for another hour. The resulting solution was filtered, and the hydrolysate was analyzed for its starch content by measuring absorbance at 630 nm using a spectrophotometer. The resistant starch test was performed in triplicate ($n = 3$) which statistical analysis was performed using one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT)

with mean values at a 95% confidence level ($p < 0.05$) using SPSS Statistics version 25 (IBM Corp., Armonk, NY, USA).

2.5 Viability Test of *Lactobacillus plantarum* Bacteria

The viability of *Lactobacillus plantarum* FNCC 0020 (AgaviLab) was tested by preparing MRS Agar and MRS Broth media (Himedia) that were sterilized in an autoclave (121°C, 15 minutes). The bacteria that had been rejuvenated on MRS Agar medium were inoculated into MRS Broth and incubated (37°C, 48 hours) to create a stock suspension. The test samples, consisting of 1% probiotics (control) and their combinations with 1%, 3%, and 5% prebiotic film, were incubated in MRS Broth (37°C, 24 hours). Viability was calculated using the total plate count method; samples were serially diluted to 10^{-8} , plated in duplicate on MRS Agar, and incubated (37°C, 24–48 hours). The bacterial viability assay was performed in triplicate ($n = 3$) and the number of colonies that grew was then counted according to SNI 2897:2008 standards which statistical analysis was performed using one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT) with mean values at a 95% confidence level ($p < 0.05$) using SPSS Statistics version 25 (IBM Corp., Armonk, NY, USA) (Legowo et al., 2025).

3. RESULTS AND DISCUSSION

3.1 Physical and Mechanical Properties in Prebiotic Film

The prebiotic film products in this study were made in 3 samples with the addition of plantain peel starch and a control made of sago flour. The control prebiotic film product, which was made only from sago flour, CMC, and glycerol, produced a white color, while the prebiotic film product with the addition of plantain peel starch showed a brown color, as seen in Figure 1. The characterization of prebiotic films incorporated with kepok plantain peel starch at varying concentrations (1%, 3%, 5%) revealed significant trends in their properties. The tensile strength values (Table 3) are very low (0.27 MPa, 0.17 MPa, and 0.20 MPa, respectively), showing a fluctuating trend with the 1% concentration effect of banana peel starch being the best effect on film strength. The low tensile strength value can be influenced by the inherent properties of banana peel starch and the glycerol plasticizer, which weaken the hydrogen bonds between the polymers and reduce the mechanical integrity of the film (Chandrasekar et al., 2023). Although the tensile strength value is relatively low compared to typical packaging films, the main function of the developed film is intended as a prebiotic carrier film that can be directly consumed. Therefore, the mechanical requirements are not too strict and can be tolerated as long as the film maintains its integrity during storage and handling as a functional food (Warkoyo et al., 2022).

Elongation at break (Table 3) demonstrates a clear and significant trend of decreasing values with the increasing concentration of banana peel starch. Specifically, the elongation values decline from 178.10% at 1% concentration to 129.17% at 3% and further to 89.29% at 5%. This inverse relationship aligns with the observed increase in film stiffness as the solid

content rises. Higher concentrations of starch enhance the density of intermolecular hydrogen bonds and chain entanglement, which restrict the mobility of the polymeric chains and diminish the film's capacity for plastic deformation (Pan et al., 2024). Nevertheless, all formulations surpass the 70% minimum elongation required by JIS standards, signifying adequate flexibility for handling as a consumable food product.

Film thickness increased proportionally from 0.13 mm to 0.18 mm, well within acceptable limits. Moisture content rose with starch concentration (11.42% to 16.24%), whereas water solubility exhibited an inverse relationship, decreasing from 55.31% to 41.14%. The resistant starch content in the films, though substantially lower than that of the native starch (38.91%), also increased with the addition level, ranging from 1.89% to 3.76%. The data of the test can be seen in Table 3.

The thickness of the prebiotic film increased with the addition of kepok plantain peel starch concentration from 1% to 5%, from 0.13 mm to 0.18 mm. All these values are still below the Japanese Industrial Standard (JIS) maximum limit of 0.25 mm (Warkoyo et al., 2022), indicating that all formulation variations have met the basic physical criteria for prebiotic film. This phenomenon of increasing thickness is linearly driven by the increase in total solids in the film-forming solution, where a higher starch concentration will form a denser and thicker matrix (Pan et al., 2024), as also observed in previous studies Rahmawati et al. (2025).

The hydrophilic nature of starch also significantly affects the characteristics of barrier films. The film's moisture content increased significantly from 11.4% to 16.2% as the starch concentration increased from 1% to 5%. This is due to the abundance of hydroxyl groups ($-OH$) in starch, which are naturally hydrophilic and capable of strongly binding water molecules within the film matrix (Indrianti et al., 2018; Daza et al., 2021). The same property is also responsible for the high-water film solubility value, which ranges from 41.1% to 55.3%. The relatively high-water solubility of the film is beneficial for prebiotic films consumed directly, as rapid dissolution in the digestive tract will facilitate the release of prebiotic oligosaccharides and their availability for fermentation by gut microbiota (Allahverdi and Dadmehr, 2026).

Furthermore, proximate analysis showed that the film-making process caused a drastic reduction in resistant starch content, from 38.9% in native starch to below 4% in all film samples. This decrease is caused by the gelatinization and heating processes during film production, which destroy the crystalline structure of starch granules (RS2 type), the main form of resistant starch in raw starch (Huang et al., 2022). Additionally, the addition of glycerol inhibits the retrogradation process necessary for the formation of new type RS3 resistant starch (Gutiérrez and Álvarez, 2016). Although its value decreased, the presence of detectable resistant starch still indicates the functional potential of kepok plantain peel starch. Overall, this study successfully characterized prebiotic films from kepok plantain peel starch and identified critical areas for further development, such as optimizing the type and concentration

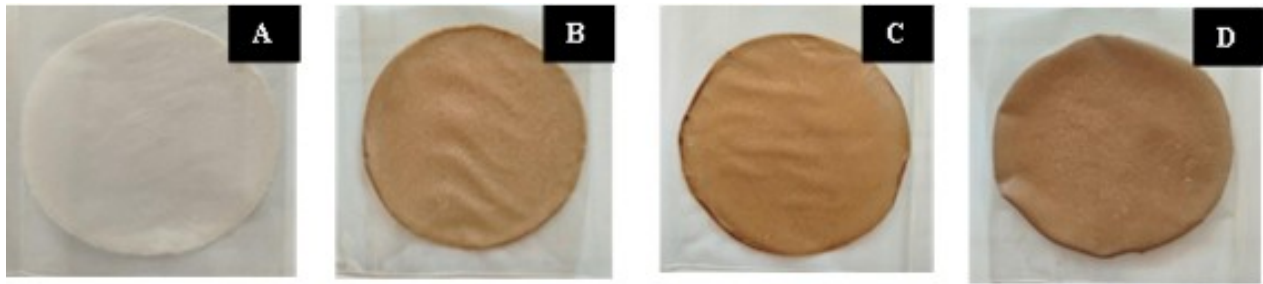


Figure 1. (A) Control Prebiotic Film, (B) Prebiotic Film with 1% Plantain Peel Starch, (C) Prebiotic Film with 3% Plantain Peel Starch, (D) Prebiotic Film with 5% Plantain Peel Starch

Table 3. Thickness, Tensile Strength, Elongation, Moisture Content, Water Solubility, Resistant Starch, and Viability Probiotic Test for Prebiotic Films

Group	Thickness (%)	Tensile Strength (MPa)	Elongation (%)	Water Content (%)	Solubility in Water (%)	Resistant Starch (%)	Viability <i>L. plantarum</i> (CFU/mL)
Control	-	-	-	-	-	-	1.9×10^{8c} ± 0.047
variation 1%	0.13 ^c ± 0.003	0.27 ^a ± 0.064	178.10 ^a ± 0.065	11.42 ^b ± 0.034	55.31 ^a ± 0.011	1.89 ^d ± 0.087	21×10^{8b} ± 0.044
variation 3%	0.16 ^b ± 0.004	0.17 ^b ± 0.087	129.17 ^b ± 0.073	16.65 ^a ± 0.047	49.20 ^b ± 0.025	2.95 ^c ± 0.068	23×10^{8ab} ± 0.013
variation 5%	0.18 ^a ± 0.002	0.20 ^c ± 0.038	89.29 ^c ± 0.059	16.24 ^a ± 0.064	41.12 ^c ± 0.043	3.76 ^b ± 0.085	26×10^{8a} ± 0.011
plantain peel paste	-	-	-	-	-	38.91 ^a ± 0.067	-

Values are presented as mean \pm standard deviation ($n = 3$ for all parameters). Different superscript letters within the same column indicate statistically significant differences among film formulations at $p < 0.05$, based on one-way ANOVA followed by Duncan's Multiple Range Test (DMRT).

of plasticizers and cross-linking modification to improve mechanical properties and water barrier.

3.2 Morphological Properties of Prebiotic Films

FTIR characterization (Figure 2) indicated that the functional groups of the sago starch-based prebiotic film were not significantly altered by the addition of plantain peel starch, with key absorptions observed for glycosidic bonds (C–O–C) at 1041 cm^{-1} , C–H bending of polysaccharides at $1328\text{--}1432 \text{ cm}^{-1}$, bound water (O–H bending) at 1634 cm^{-1} , and a broad hydroxyl (O–H) stretch at $3200\text{--}3550 \text{ cm}^{-1}$ influenced by glycerol and starch. The presented FTIR spectral analysis shows that there is no fundamentally significant difference between the control prebiotic film spectra (based on sago flour) and the prebiotic film enriched with kepok plantain peel starch, indicating good compatibility between the two materials. The second spectrum of both samples shows the characteristic absorption bands of polysaccharides at a wavenumber of 1041 cm^{-1} , representing the glycosidic bond (C–O–C), C–H bending vibrations at $1432\text{--}1328 \text{ cm}^{-1}$, and the O–H bending peak of bound water at 1634 cm^{-1} , which confirms the hygroscopic

nature of the film. Strong absorption in the region of $3550\text{--}3200 \text{ cm}^{-1}$ represents the stretching of the hydroxyl (–OH) group, which is enhanced by the presence of glycerol and hydroxyl groups from plantain peel starch, while alkane (C–H) absorption is observed at $2990\text{--}2850 \text{ cm}^{-1}$.

The consistency of this spectral pattern with the findings of Ferreira-Villadiego et al. (2018) on plantain peel starch, Ningrum et al. (2020) on sago films, and Irmayanti and Anwar (2024) on plantain peel starch films confirms that the addition of kepok plantain peel starch does not change the fundamental functional groups of the polymer matrix, but strengthens the characteristic spectral fingerprint of starch-based materials by increasing the intensity of the hydroxyl group peaks, indicating stronger hydrogen interactions within the film system. The increased O–H band intensity prebiotic films suggests that the additional hydroxyl groups from banana peel starch and glycerol participate in forming a more extensive hydrogen bond network, contributing to the cohesive film structure observed in SEM micrographs.

SEM micrographs (Figure 3) suggested a relatively smooth and homogeneous matrix with some fine cracks and ridges,

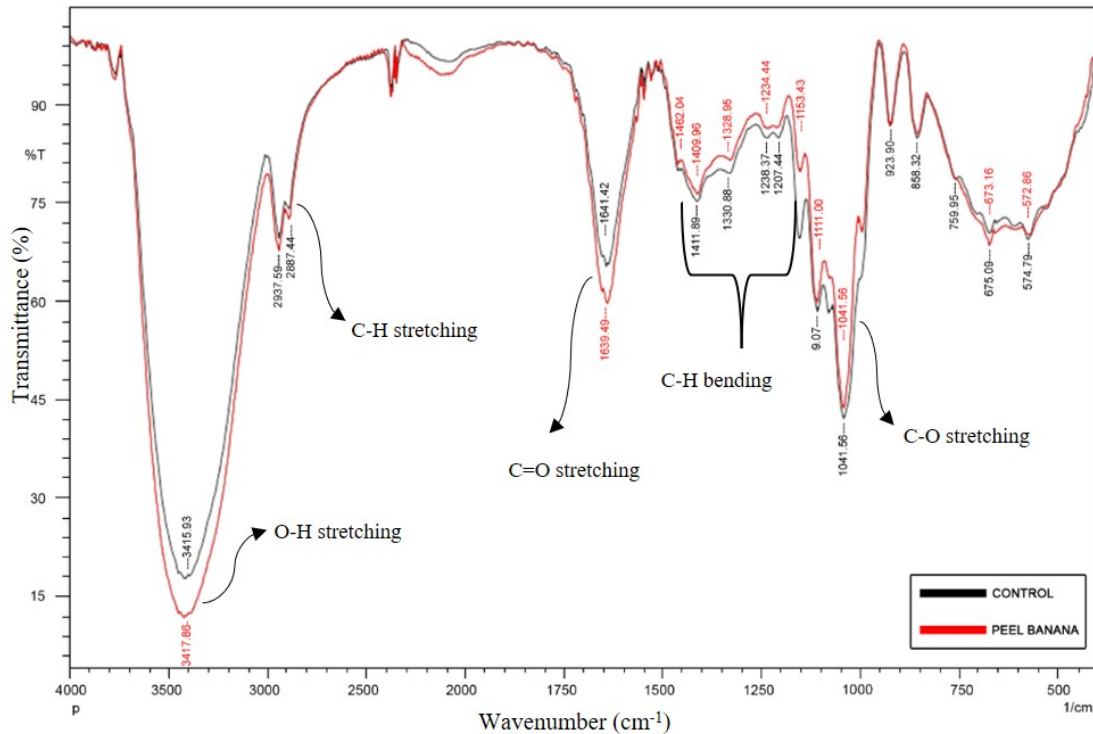


Figure 2. Comparative Fourier Transform Infrared (FTIR) Spectra of Prebiotic Films on The Control Film (without addition) and The Film Incorporated with Plantain Peel Starch

characteristic of gelatinized sago starch, while the incorporation of plantain peel starch introduced a more heterogeneous structure with fibrous or granular features. Morphological analysis of SEM results showed a significant difference between prebiotic films made from pure sago flour and a combination of sago flour and kepok plantain peel starch. Prebiotic films from pure sago flour exhibited a relatively smooth and homogeneous surface with even matrix distribution, although some fine cracks (microcracks) and ridges were present, which are characteristic of starch-based films undergoing gelatinization and retrogradation. This fine morphology indicates good plasticizer dispersion within the polymer matrix and high compatibility between its constituent components (Chen et al., 2024). Conversely, prebiotic films with the addition of kepok plantain peel starch showed a more heterogeneous morphology with the presence of fibrous and granular structures as well as a tendency for particle agglomeration. The presence of these fibrous structures stems from the natural characteristics of kepok plantain peel starch, which has larger granules and a higher fiber content, resulting in a more complex network within the film matrix.

These morphological differences have direct implications for the functional properties of prebiotic films. The smooth surface of pure sago film is generally correlated with better barrier properties against water vapor and gas migration, due to the minimal presence of defects that can act as diffusion pathways (Esfandiari et al., 2025). Conversely, the structural

heterogeneity in composite films can create more complex diffusion pathways, which can either enhance or diminish barrier properties depending on phase distribution (Pires et al., 2021). On the other hand, it can provide mechanical reinforcement through the interlocking mechanism of its fibrous structure. The inhomogeneity in the distribution of these particles indicates challenges in achieving perfect compatibility between the two types of starch, which need to be addressed through optimization of the mixing process and the addition of compatibilizers in further research.

SEM micrographs revealed that the incorporation of banana peel water extract induced a structural transformation in the prebiotic film, shifting from the compact, continuous surface of the control film to a more amorphous and less dense morphology. This amorphous structure is functionally significant for the film's performance as a prebiotic delivery system. Amorphous matrices are characterized by higher molecular mobility and free volume, which facilitate water penetration and matrix disintegration, thereby directly supporting our observed increase in film solubility (Saršūns et al., 2026; Wernisch et al., 2024). Enhanced solubility is desirable for ensuring rapid release of prebiotic substrates in the gastrointestinal tract. Furthermore, the less compact, amorphous morphology likely increases surface area and matrix porosity, which can improve bacterial accessibility to encapsulated prebiotics by providing greater colonization sites and facilitating enzymatic degrada-

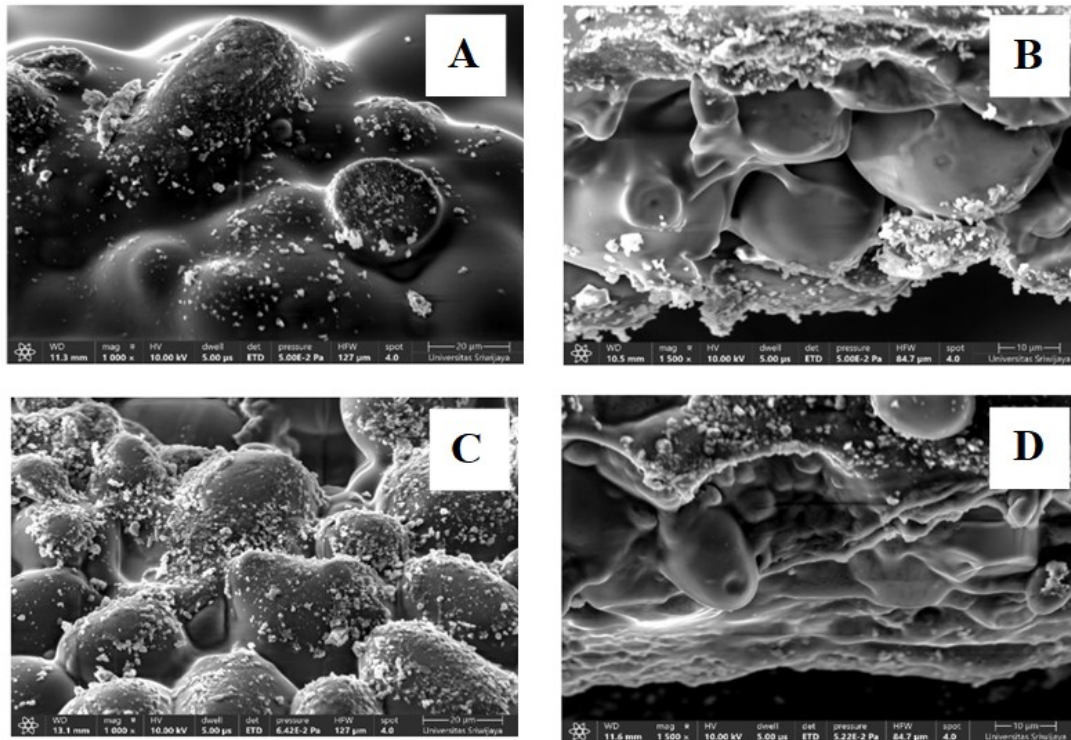


Figure 3. Morphology from SEM Micrographs of Prebiotic Film Surfaces at Different Magnifications with (A) Control Film (Sago Flour) at 5.000×; (B) Control Film at 10.000×; (C) Composite Film (Sago Flour with Kepok Plantain Peel Starch) at 5.000×; (D) Composite Film at 10.000×

tion (Shanuke et al., 2025; Luca and Oroian, 2021). These structure-function relationships underscore that the physical architecture of the film is intrinsically linked to its prebiotic potential, where the amorphous state promotes both dissolution and substrate bioavailability for gut microbiota. This morphology is favorable for the film's intended use as a digestible prebiotic matrix, as it enables faster saliva and gastric fluid penetration, accelerating the exposure of prebiotic substrates to intestinal bacteria.

3.3 Viability Test of *Lactobacillus plantarum* Bacteria for Prebiotic Film

The viability test of probiotic bacteria (Table 3) revealed that the prebiotic film with 5% kepok plantain peel starch supported the highest growth of *L. plantarum* at 26×10^8 CFU/mL, followed by the 3% and 1% formulations with 23×10^8 and 21×10^8 CFU/mL, respectively. Viability test for probiotic on prebiotic film showed that the addition of kepok plantain peel starch significantly increases the viability of the probiotic bacterium *L. plantarum* in prebiotic films. These results confirm that kepok plantain peel starch acts as an effective source of carbon and nutrients for bacterial growth, where an increase in starch concentration is linearly correlated with an increase in cell viability. This finding aligns with the research by Kusuma and Zubaidah (2016), who reported high viability of *L. plantarum* in kepok plantain peel flour medium (1.92×10^{11} CFU/mL), supported

by the simple sugar (glucose, sucrose, fructose) and essential micronutrient content in plantain peels.

This mechanism for increasing viability can be explained by the dietary fiber and complex carbohydrate content in plantain peel starch, which acts as a prebiotic substrate, facilitating the metabolism and proliferation of probiotic bacteria. The study by Tan et al. (2024) reinforces these findings by demonstrating that banana peel supplementation can increase the *L. plantarum* population to over 10 log CFU within 24 hours. Thus, prebiotic films based on kepok plantain peel starch not only have potential as active packaging but also as an effective probiotic carrier for functional food applications, where a concentration of 5% shows optimal efficacy without significant differences compared to lower concentrations. A schematic illustration of the proposed prebiotic film mechanism can be seen in Figure 4.

The prebiotic mechanism enhancing the viability of *L. plantarum* in the composite film can be explained by the synergistic effects of the various components found in the starch of kepok banana peel. The skin of kepok plantain peel starch contains a considerable amount of total dietary fibre (~ 48%, Table 1), which includes pectin and hemicellulose fractions. These components are resistant to digestion in the upper gastrointestinal tract and reach the large intestine intact, where they serve as fermentation substrates for beneficial bacteria (Han et al., 2023;

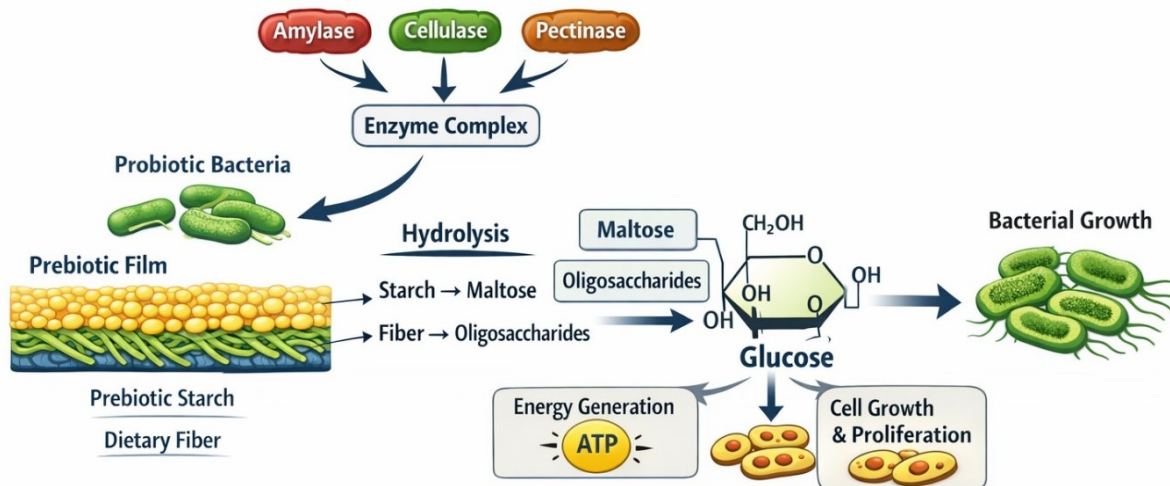


Figure 4. Proposed Mechanism of Probiotic-Derived Enzyme Complex in Hydrolyzing Prebiotic Film Components

Guarino et al., 2020). Although there is a significant reduction in resistant starch content following the gelatinisation process (from 38.91% to 1.89–3.76%, Table 3), the remaining resistant starch within the film matrix remains enzymatically accessible to bacterial amylase. *L. plantarum* is known for its production of extracellular α -amylase and other glycoside hydrolases (Behera et al., 2018; Hao et al., 2024), which can hydrolyse the α -1,4 and α -1,6 glycosidic bonds in starch, thereby releasing glucose, maltose, and malto-oligosaccharides that support bacterial proliferation. The natural simple sugars found in banana peels provide an immediately accessible carbon source for rapid initial growth. The morphology of the amorphous and less dense film, as observed through SEM (Figure 3), supports this mechanism by facilitating water absorption, matrix swelling, and enzyme infiltration, which in turn increases the surface area available for enzymatic hydrolysis and substrate release. The structural, compositional, and morphological characteristics create an environment that promotes the sustained release of prebiotics, gradually nourishing *L. plantarum*.

The enhancement in viability achieved in this study is comparable to or exceeds that research for other starch-based prebiotic or probiotic delivery systems. Shoukat et al. (2024) reported 85% encapsulation efficiency and improved stability under simulated gastrointestinal conditions for probiotic on starch-based film, yet they did not evaluate prebiotic-driven proliferation. Rahma et al. (2025) developed pre-gelatinised cassava starch films as carriers for *Bacillus coagulans*, demonstrating probiotic viability of approximately 10^8 CFU/g after 90 days of storage, but the film functioned solely as a protective carrier, not as a prebiotic growth substrate. Coimbra et al. (2023) enhanced starch films using agrifood waste for the delivery of *Lactobacillus rhamnosus*, noting a reduced rate of viability loss during storage, attributed to antioxidant protection rather than prebiotic stimulation. In contrast, our research shows a net

increase in probiotic viability reaching up to 26×10^8 CFU/mL on *L. plantarum* during incubation with the film without the need for external prebiotic supplementation. This positions the current prebiotic starch film as an active nutritional medium rather than a mere passive carrier. Moreover, the achieved viability level on this research aligns with the therapeutic range recommended for probiotic products (10^6 – 10^8 CFU/g), highlighting the practical applicability of the formulated film as a functional food component. However, the stability of *L. plantarum* within the prebiotic starch film during prolonged storage at various temperatures and relative humidities was not evaluated. Evaluating storage stability is a critical subsequent step in determining the shelf life of the film as a functional food product.

4. CONCLUSION

This study successfully developed the first composite prebiotic starch film from sago flour and kepok plantain peel starch, designed as a directly consumable functional food ingredient rather than a packaging material. The key scientific contribution is the demonstration that, despite partial loss of resistant starch during gelatinization, the whole plantain peel starch within the film matrix retains significant prebiotic capacity, effectively enhancing *Lactobacillus plantarum* viability up to 26×10^8 CFU/mL without any commercial prebiotic additives. This positions the film as a novel, waste-derived dietary prebiotic carrier that can be consumed as a snack, supplement, or food component to support gut health. The film exhibited acceptable handling properties, and its relatively high-water solubility is an advantage for prebiotic release in the digestive tract. From an industrial perspective, this prebiotic starch film offers a sustainable strategy to valorize agricultural waste into a functional food product. Future work should focus on optimizing mechanical properties through plasticizer adjustment or

cross-linking to improve handling and storage stability, as well as evaluating the film's prebiotic efficacy *in vivo* and in real food matrices to establish its commercial viability as a functional food.

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