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Fabrication and Characterization of Pectin-Chitosan Edible Coatings with a *Cosmos caudatus* Leaf Extract for Tomato Preservation

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Cite This: *ACS Omega* 2025, 10, 7204−7210

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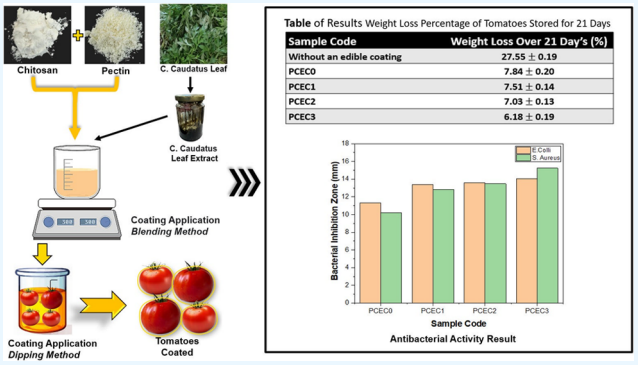
ABSTRACT: An edible coating based on pectin-chitosan and *Cosmos caudatus* leaf extract has been created. *Cosmos caudatus* leaf extract, which contains several bioactive compounds, aims to produce an edible coating with antibacterial properties. *C. caudatus* extract was incorporated at concentrations of 1, 2, and 3 g into a mixture of 1.5 g of pectin and 1 g of chitosan. The edible coating was applied to the tomatoes using the dipping method. The coated tomatoes were analyzed for 21 days at room temperature to determine the weight loss value. The edible coating was characterized, including FTIR analysis, X-ray diffraction, surface morphology, thermal stability, viscosity, and antibacterial activity. The research results reveal that *C. caudatus* extract contains anthocyanins with antibacterial properties, has an amorphous crystalline structure, and has a textured surface with partial aggregation. Thermal stability analysis using differential scanning calorimetry (DSC) shows a decrease in thermogravimetric (TG) values with increasing extract concentration. The optimal weight loss (6.18%) was found in the pectin-chitosan composition containing 3 g of *C. caudatus* extract. At this concentration, the inhibition zones against *Escherichia coli* and *Staphylococcus aureus* were 16.4 and 15.6 mm, respectively. These findings indicate that the *C. caudatus* leaf extract, particularly at 3 g, enhances the antibacterial properties of the edible pectin-chitosan coating, demonstrating its potential to extend the shelf life of tomatoes safely.



1. INTRODUCTION

In recent years, there has been a growing emphasis within the food industry on developing sustainable and environmentally friendly preservation methods.[1] Edible coatings have emerged as a promising solution for extending the shelf life of perishable foods while offering an alternative to synthetic packaging.[2] These coatings, composed of natural polymers and bioactive compounds, present a biodegradable option that provides essential functional benefits such as moisture control, oxidation prevention, and antimicrobial activity.[3] As concerns over environmental impact and food safety increase, the exploration of such coatings has become increasingly relevant.[4]

Edible coatings serve multiple critical functions. They create a thin, protective layer on the surface of fruits and vegetables, which helps in maintaining freshness, reducing moisture loss, and minimizing oxidation.[5] Additionally, these coatings can act as barriers to microbial contamination, thereby enhancing food safety and quality. The ability to incorporate antimicrobial agents into these coatings further amplifies their protective capabilities.[6] By addressing microbial spoilage, edible coatings can significantly contribute to reducing food waste and improving shelf life, which are essential for advancing food preservation technologies.

Among various materials used in edible coatings, pectin and chitosan have gained considerable attention due to their unique properties.[7,8] Pectin, a natural polysaccharide derived from fruits, is known for its gelling and film-forming abilities.[9] It provides structural integrity and can form a gel-like network when combined with other substances.[10] Previous research also indicated that pectin could maintain the firmness of fruit, thereby reducing the ripening rate for up to 12 days at 20 °C.[11] Chitosan, a biopolymer obtained from chitin, enhances the coating's ability to interact with negatively charged surfaces due to its positive charge.[12] Previous studies have also shown that chitosan used as a coating material can effectively preserve the quality of tomatoes during storage at 8 °C for 21 days.[13] The combination of pectin and chitosan in edible coatings takes advantage of their complementary properties, creating a robust and versatile coating system that offers both mechanical

Received: November 25, 2024
Revised: January 7, 2025
Accepted: January 31, 2025
Published: February 10, 2025

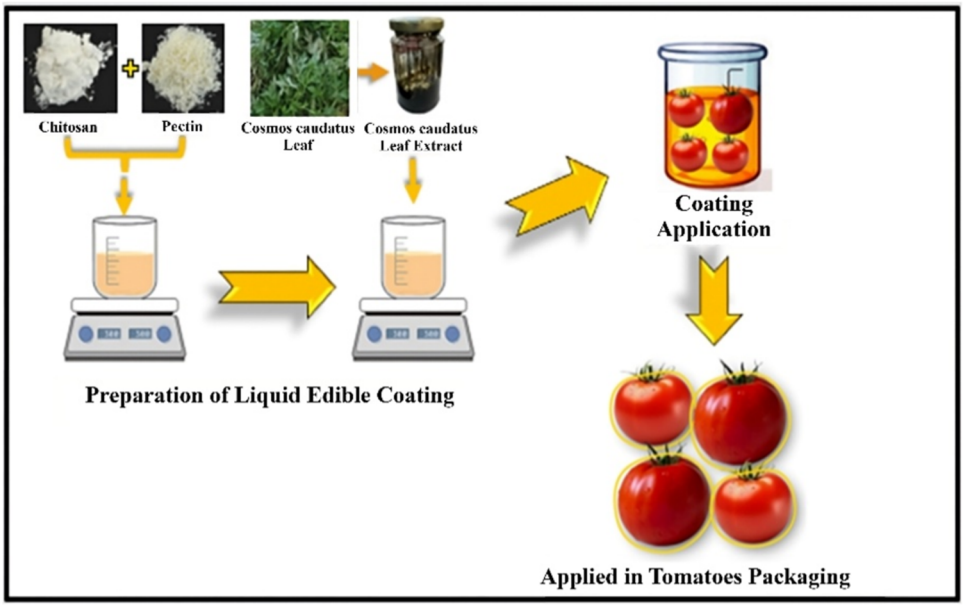


Figure 1. Schematic diagram of the experimental procedures in this study.

strength and functional benefits.[14] Tomatoes coated with chitosan exhibit lower ethylene production, reduced soluble solid concentration, and better retention of total acidity and vitamin C content.[15] Notably, chitosan coating also helps preserve the nutraceutical quality of tomatoes by slowing the degradation of bioactive compounds, such as lycopene and phenolics, during storage.[15]

The integration of antimicrobial agents into edible coatings addresses the challenge of microbial spoilage in perishable foods.[16] Microbial contamination is a major cause of food degradation, leading to spoilage and potential health risks.[17] Antimicrobial agents can inhibit or kill bacteria, fungi, and other microorganisms that contribute to food spoilage.[18] Natural extracts, such as those derived from plants, provide a sustainable source of antimicrobial compounds.[1,19] By incorporating such extracts into edible coatings, it is possible to enhance their protective properties while minimizing the reliance on synthetic preservatives, aligning with the growing trend toward natural and ecofriendly food preservation solutions.

The *Cosmos caudatus* (*C. caudatus*) leaf extract, derived from *C. caudatus*, has shown significant potential as an antibacterial agent in recent studies.[20] This plant extract is rich in bioactive compounds with known antimicrobial properties, which can effectively inhibit the growth of bacteria and fungi. The incorporation of the *C. caudatus* leaf extract into edible coatings could enhance their ability to prevent microbial contamination and extend the shelf life of coated foods.[17] Given the promising antibacterial properties of the *C. caudatus* leaf extract, it represents a valuable addition to the formulation of pectin-chitosan coatings. Due to its quercetin content, which exhibits anticancer properties, *C. caudatus* leaves (*C. caudatus* Kunth.) have been applied as a complementary therapy for breast cancer. The primary goal of this application is to formulate a nanosuspension containing the *C. caudatus* leaf extract with cytotoxic activity against MCF-7 breast cancer cells.[21] Recent studies have explored the potential use of the *C. caudatus* leaf extract (*C. caudatus*) in edible films as a mouth freshener, highlighting its antioxidant and antibacterial properties.[22]

The aim of this study is to investigate the structural and antimicrobial properties of pectin-chitosan edible coatings that are enhanced with the *C. caudatus* leaf extract.[23] Specifically, the study focuses on characterizing the surface morphology and crystallinity of the coatings, evaluating their antimicrobial efficacy, and assessing how varying concentrations of pectin and kenikir leaf extracts influence these properties. By addressing these objectives, the research seeks to identify the most effective coating formulations that balance structural stability with antimicrobial effectiveness. Understanding the structural characteristics of edible coatings is crucial to evaluating their performance. Techniques such as Scanning Electron Microscopy (SEM) and X-ray Diffraction (XRD) provide insights into the surface morphology and crystalline properties of the coatings, respectively. These analyses help in assessing how different formulations impact the coating's ability to form a uniform barrier and interact with microbial agents. Additionally, evaluation of antimicrobial activity through tests against common foodborne pathogens provides information on the effectiveness of the coatings in preventing microbial growth. The novelty of this research is the incorporation of the *C. caudatus* leaf extract in a pectin-chitosan matrix; it's unique due to the bioactive profile, including anthocyanins, flavonoids, and phenolics, which are not commonly used in similar studies.

2. MATERIALS AND METHODS

The experimental stages of this study include materials, preparation, application, physical-chemical characterization, and antimicrobial activity, as illustrated in Figure 1.

2.1. Materials. The materials used for the experiment included glacial acetic acid, 96% ethanol, glycerol, and chitosan, which were purchased from Sentra Chemical Laboratory (Indonesia), and pectin from citrus peel, which was purchased from IR & Co. (Indonesia). *C. caudatus* leaves and fully mature tomatoes were collected from the local markets in North Sumatra, Indonesia.

https://doi.org/10.1021/acsomega.4c10344
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2.2. Preparation. *2.2.1. C. caudatus Leaf Extraction.* Fresh *C. caudatus* leaves were washed and cut. They were then dried at 40 °C in an oven for 3 days, followed by pulverizing and sieving through a 40 mesh sieve before extraction. The *C. caudatus* leaves powder was extracted using a direct maceration method. In detail, as much as 100 g of the *C. caudatus* leaves powder was mixed and soaked in 1 L of ethanol for 24 h at room temperature with occasional shaking. All of the extract solutions were then filtered using Whatman No.1 filter paper, and the filtrate was evaporated using a rotary evaporator (Rotavapor R-100, BUCHII) at 40 °C to obtain the dry extract.[24] The concentrations of the *C. caudatus* extract used in this study (1, 2, and 3 g) are well within the limits of natural plant extract usage in food applications. The bioactive compounds, including phenolics and flavonoids, are derived from plants traditionally consumed in various cuisines, which supports their biocompatibility and safety.

2.2.2. Preparation of the Liquid Edible Coating of Pectin, Chitosan, and C. caudatus Extract. The liquid edible coating of pectin, chitosan, and *C. caudatus* extract was prepared using the mixing method. Briefly, as much as 1.5 g of pectin was dissolved in 100 mL of distilled water and stirred at 500 rpm for 30 min. Then, a chitosan solution (1 g of chitosan dissolved in 10 mL of acetic acid) was added. The mixture was stirred at 500 rpm for 1 h at 80 °C. Next, the *C. caudatus* leaf extract, in varying amounts of 1, 2, and 3 g, was added to the solution. The samples were then named PCEC0 (the control sample, with no addition of the *C. caudatus* leaf extract), PCEC1 (1 g of the *C. caudatus* leaf extract), PCEC2 (2 g of the *C. caudatus* leaf extract), and PCEC3 (3 g of the *C. caudatus* leaf extract). After cooling for 30 min, 1.5 mL of glycerol was added to each variation of the mixture, and it was stirred for an additional 30 min.

2.3. Application in Tomato Coating. The tomatoes were selected based on uniformity in size, maturity (mature red), and absence of external damage. They were washed with running water and dried naturally at room temperature. Afterward, they were dipped into edible coating solutions for one min, and tomato without edible coating was used as the control in the experiment. The coated tomato samples were stored at room temperature for 21 days, and weight loss was observed.

2.4. Physical-Chemical Characterization of the Liquid Edible Coating of Pectin, Chitosan, and *C. caudatus* Extract. The sample was pressed into a solid form on a glass slide before being measured with FT-IR spectroscopy, powder X-ray diffraction (XRD), morphology measurement, and differential scanning calorimetry (DSC) analyses. In addition to these analyses, viscosity and weight loss measurements were also conducted.

2.4.1. FT-IR Spectroscopy. Fourier transform infrared (FT-IR) spectra were recorded using an IR spectrometer (Jasco FT/IR 200, Japan) with an infrared microscope with an ATR-1000-VS objective. The spectra were obtained as the average of 50 scans recorded at a resolution of 4 cm^{-1} in the range from 4000 to 500 cm^{-1} with a DLATGS detector.

2.4.2. X-ray Powder Diffraction (XRD) Measurements. A Shimadzu XRD 7000 diffractometer with Ni filtered Cu Kα radiation (λ = 1.5406 Å), 2θ ranging between 10 and 80°, was used for the compositional and structural characterization of the samples through X-ray diffraction (XRD).

2.4.3. Morphological Measurements. Microscopic morphology of obtained dried coatings was explored by using a FEI Quanta Inspect F scanning electron microscope, operated at 25 kV. A thin and continuous layer of gold was deposited on the sample surface before the SEM investigation.

2.4.4. Differential Scanning Calorimetry (DSC) Measurements. Thermal analysis was carried out utilizing a differential scanning calorimeter (DSC) (1 Mettler Toledo) using indium in an ultrahigh purity nitrogen environment (flow rate 50 mL/min) by TA Instruments. In two heating−cooling cycles, 8−10 mg samples were tested at a 20 °C/min heating/cooling ramp. In the first cycle, film samples were heated to 300 °C at 20 °C/min and then isothermed for 1 min after being equilibrated at 100 °C for 1 min. Film samples were equilibrated at 300 °C and isothermed for 1 min in the second cycle and then cooled to 100 °C at 10 °C/min and isothermed for 1 min. Each sample had three separate thermal scans, and the average results were presented. Using information from the results, the glass transition temperature (T_g) was calculated.[25]

2.4.5. Viscosity Measurements. The viscosity of the coating solution is measured using viscometers with the AMETEK Brookfield brand, using a No. 4 spindle at 100 rpm. As much as 200 mL sample of the solution was measured for 5 min at 20 °C. The result is expressed in centipoise (cP).[26,27]

2.4.6. Weight Loss Determination. The total weight loss fruit samples were determined by difference in weight between the initial and the final storage time[28] by eq 1.

$$\text{total weight loss (\%)} = \frac{w_1 - w_2}{w_1} \times 100 \quad (1)$$

where w_1 is the initial weight at day 0 and w_2 is the final weight at the end of storage studies. The experiment was performed independently in triplicates.

2.5. Antimicrobial Activity. In vitro antimicrobial activity was measured by using the disk diffusion method. Bacterial suspensions (108 CFU/mL) of *Staphylococcus aureus* and *Pseudomonas aeruginosa* were swabbed into solidified Mueller−Hinton agar. After 5 min, the 6 mm cut PGTC films were placed on top of solidified agar in 9 cm Petri dishes. The Petri dishes were incubated at 37 °C for 24 h in an incubator. Using a caliper, we determined the inhibition zones of the nanocomposite films. The index of antimicrobial activity can be calculated using eq 2.

$$\text{index of antimicrobial activity (mm)} = \frac{\text{diameter of inhibition zone} - \text{diameter of film (6 mm)}}{\text{diameter of film (6 mm)}} \quad (2)$$

3. RESULTS AND DISCUSSION

3.1. FT-IR Analysis. FT-IR analysis was conducted to determine the components present in the liquid edible coating layer, both without and with the addition of the *C. caudatus* leaf extract. In Figure 2, the control sample (PCEC0) shows a sharp peak at a wavelength of 3307.06 cm^{-1}, indicating the absorption of the −OH group, specifically phenol. Similarly, a medium peak at 1594.66 cm^{-1} corresponds to the absorption of the aromatic C=C group. Peaks at wavelengths of 1029.69 and 1108.05 cm^{-1} indicate the absorption of the C−O group, which aligns with the functional groups found in pectin and chitosan.

The graphs for the PCEC1, PCEC2, and PCEC3 compositions, shown in Figure 2, indicate that anthocyanin compounds from the *C. caudatus* leaf extract are bound to the

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ACS Omega 2025, 10, 7204−7210

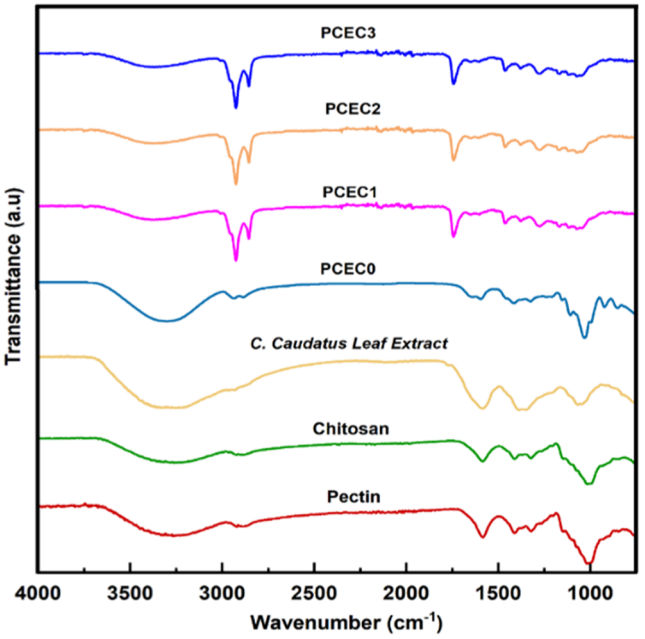

Figure 2. FTIR spectra of pectin, chitosan, *C. caudatus* leaf extract, and liquid edible coatings (solid form) under different *C. caudatus* leaf extract weights.

edible coating matrix. This is evidenced by a peak at 3368.01 cm^{-1}, where the O−H group, specifically phenol, exhibits broadening. Additionally, at wavelengths of 667.76 and 731.13 cm^{-1}, the C−H group also shows broadening. Peaks at 1073.32 and 1378.32 cm^{-1} correspond to the absorption of the C−O group, indicating the presence of alcohol.

The increased addition of the *C. caudatus* leaf extract to the edible coating solution caused the absorption peak of the O−H group to broaden. This broadening results from the increased number of OH groups, reflecting the antimicrobial properties of the *C. caudatus* leaf extract.[29]

3.2. X-ray Diffraction Analysis. The crystalline properties of the edible coatings were analyzed by using X-ray Diffraction (XRD) to evaluate the effect of varying concentrations of the *C. caudatus* leaf extract on the structural organization of the pectin-chitosan edible coatings. Figure 3 shows the XRD

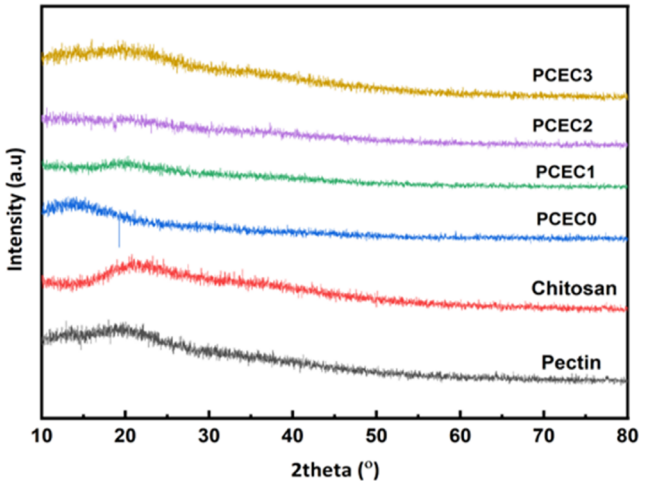

Figure 3. XRD pattern of pectin, chitosan, and pectin-chitosan edible coating with the *C. caudatus* leaf extract.

diffraction patterns of all pectin-chitosan edible coating samples containing the *C. caudatus* leaf extract. A broad crystalline peak was observed for both pectin and chitosan in the 2θ range of 15−25°, indicating their amorphous nature. This result is consistent with the findings of Benhabiles et al.[26] The PCEC0 exhibited a broad peak in the 2θ range of 10−18°. The incorporation of the *C. caudatus* leaf extract into pectin-chitosan edible coatings did not significantly change the peak position. PCEC1 exhibited a broad and diminished peak in the 2θ range of 16−26°. The peak of the edible coating was almost completely diminished for PCEC2. However, PCEC3 exhibited stronger and broader peaks in the 2θ range of 10−29°. It appeared that the crystalline structure of the edible coatings was independent of the increased concentration of the *C. caudatus* leaf extract. We suggest that the *C. caudatus* leaf extract was molecularly dispersed into the pectin and chitosan matrix, creating interactions between the phenolic compounds of the *C. caudatus* leaf extract and the functional groups of pectin and chitosan. Therefore, it could be concluded that the incorporation of the *C. caudatus* leaf extract into the pectin and chitosan matrix did not affect the amorphous structure of the edible coating. Therefore, it could be concluded that the incorporation of the *C. caudatus* leaf extract into the pectin and chitosan matrix did not affect the amorphous structure of the edible coating.

3.3. Morphological Analysis. The morphology of edible coatings made from PCEC0 is shown in Figure 4a at 1000× magnification and Figure 4c at 5000× magnification, where the surfaces of the pectin and chitosan layers appear smoother. However, some substances do not interact well with each other, resulting in clumps identified as chitosan.[30]

The morphological structure of edible coatings with the addition of the *C. caudatus* leaf extract (PCEC3) indicates that higher extract concentrations can lead to partial aggregation or uneven distribution in the coating, creating a more textured or rough surface due to the *C. caudatus* leaf extract's properties.[31] As shown in Figure 4b at 1000× magnification and Figure 4d at 5000× magnification, air voids are clearly visible, caused by the interaction between the *C. caudatus* leaf extract and the biopolymer components (pectin-chitosan).[32]

3.4. Differential Scanning Calorimetry (DSC) Analysis. Thermal analysis was conducted to determine the thermal stability of the edible coating layer sample. Figure 5 shows the thermal analysis graph of several sample variations, where the control sample (PCEC0) shows a value of 137.16 °C. The addition of the *C. caudatus* leaf extract to the samples shows a decrease in thermal value, as seen in sample PCEC1, which has a value of 126.81 °C, and sample PCEC2, which has a value of 99.94 °C. This is due to the *C. caudatus* leaf extract containing bioactive components such as flavonoids, tannins, and saponins, leading to molecular interactions in the pectin-chitosan polymer chain in the edible coating.[33,34] However, in sample PCEC3, the thermal value increases to 122.06 °C, which is still below the control sample. With the decrease in thermal value, it can affect the characteristics of the edible coating toward O_2 and CO_2 gases, which can slow down the respiration and ripening process or shelf life of tomatoes.[35]

3.5. Viscosity. The viscosity of the edible coating is crucial, as it affects both the application process and the final texture of the coating. Viscosity test results for various compositions PCEC0, PCEC1, PCEC2, and PCEC3 are presented in Table 1. The research results indicate that the PCEC0 composition has the highest viscosity (476.3 cP) compared with other

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ACS Omega 2025, 10, 7204−7210

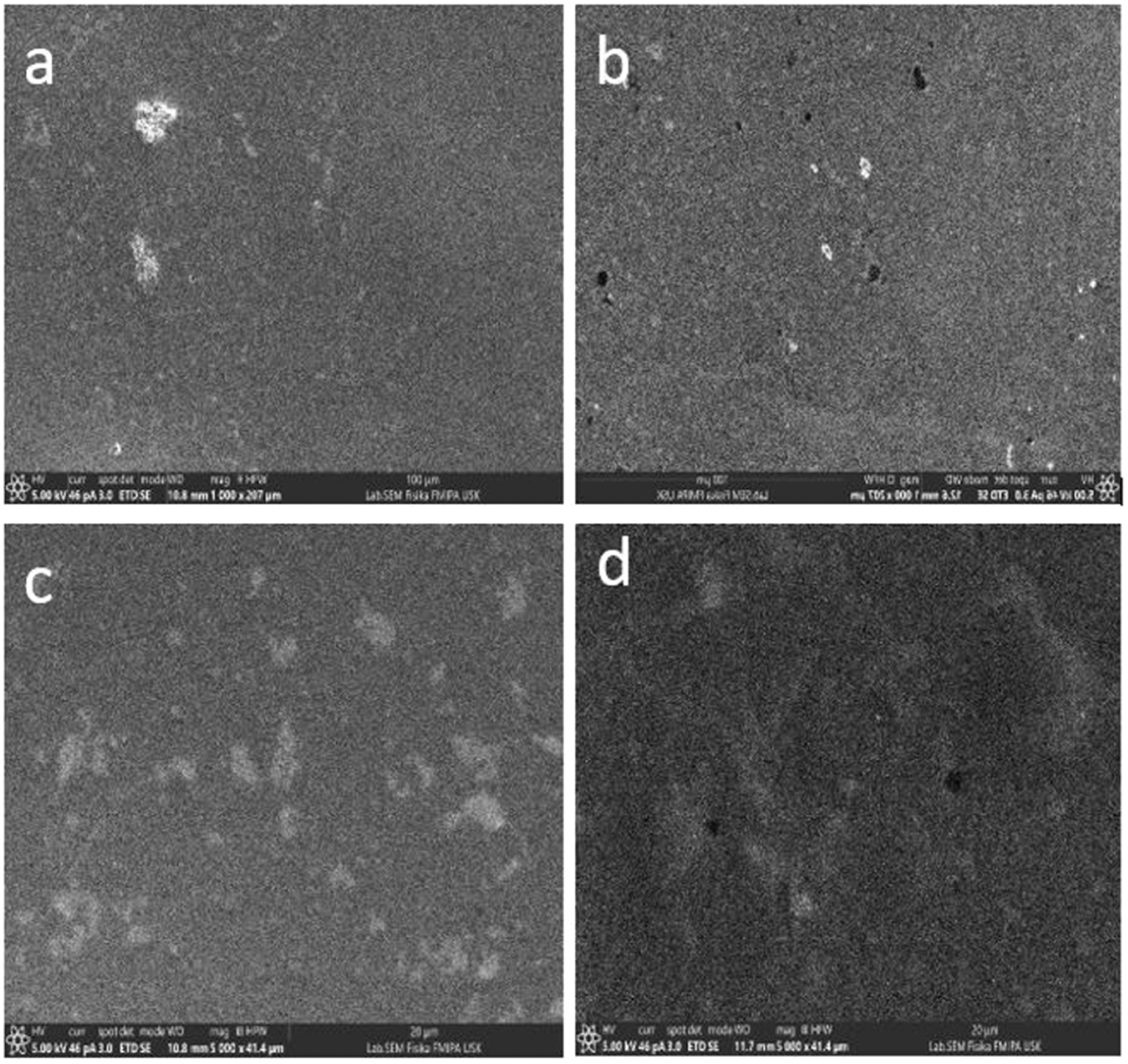

Figure 4. SEM images of (a) pectin-chitosan (1000 times magnification), (b) pectin-chitosan with the *C. caudatus* extract (1000 times magnification), (c) pectin-chitosan (5000 times magnification), and (d) pectin-chitosan with the *C. caudatus* extract (5000 times magnification).

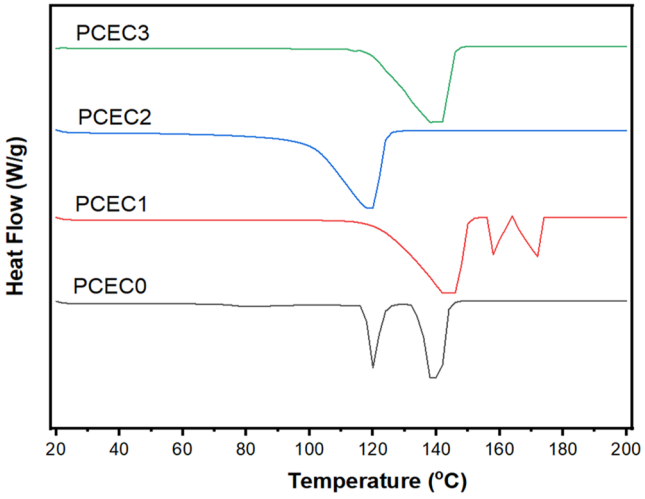

Figure 5. Thermograms of differential scanning calorimetry (DSC).

compositions. This is primarily due to the pectin content, which plays a significant role in the solution's viscosity. Pectin contains a high level of methoxyl groups, enhancing the sample's gel-forming capacity and increasing viscosity.[10] The

Table 1. Viscosity of the Pectin-Chitosan Edible Coating with the *C. caudatus* Leaf Extract

sample code	viscosity (cP)
PCEC0	476.3 ± 0.02
PCEC1	462.4 ± 0.10
PCEC2	455.6 ± 0.11
PCEC3	420.8 ± 0.10

increase in the percentage of the *C. caudatus* leaf extract leads to a reduction in viscosity: 462.4 cP for PCEC1, 455.6 cP for PCEC2, and 420.8 cP for PCEC3. This decrease is attributed to the essential oils in the *C. caudatus* leaf extract, which enhance fluidity and act as a solvent in the edible coating mixture.[36]

3.6. Weight Loss Analysis. Weight loss measurement is one of the important aspects of analyzing the effect of edible coatings. As seen in Table 2, tomatoes without a coating experienced the highest weight loss: 8.84% after 7 days, 16.64% after 14 days, and 27.55% after 21 days. This indicates that without a coating, tomatoes are more susceptible to respiration and transpiration processes. After coating PCEC0, there was a significant reduction in the weight loss percentage: 6.04% after

https://doi.org/10.1021/acsomega.4c10344
ACS Omega 2025, 10, 7204−7210

Table 2. Weight Loss Percentage of Tomatoes Stored for 7, 14, and 21 Days

sample code	weight loss over 7 days (%)	weight loss over 14 days (%)	weight loss over 21 days (%)
without an edible coating	8.84 ± 0.29	16.64 ± 0.38	27.55 ± 0.19
PCEC0	6.04 ± 0.09	7.01 ± 0.12	7.84 ± 0.20
PCEC1	5.42 ± 0.15	6.2 ± 0.10	7.51 ± 0.14
PCEC2	5.35 ± 0.22	6.53 ± 0.13	7.03 ± 0.13
PCEC3	4.06 ± 0.06	5.24 ± 0.29	6.18 ± 0.19

7 days, 7.01% after 14 days, and 7.84% after 21 days. The addition of the *C. caudatus* leaf extract further reduced the weight loss percentage. A higher concentration of the *C. caudatus* leaf resulted in a lower weight loss percentage, with the lowest weight loss observed for PCEC3 samples: 4.06% after 7 days, 5.24% after 14 days, and 6.18% after 21 days. This indicates that the edible coating with the addition of the *C. caudatus* leaf extract can slow down respiration and transpiration rates, which is related to the antibacterial properties of the extract, thereby providing optimal protection for tomatoes during storage and maintaining their quality for longer.[37]

3.7. Antibacterial Activity Analysis. Measuring antibacterial activity is a crucial aspect of evaluating edible coatings. Antibacterial properties in a coating can help slow the decay process caused by bacterial growth. By comparing the antibacterial activity of pure pectin-chitosan samples with those containing the *C. caudatus* leaf extract, the impact of the extract can be effectively analyzed. Table 3 presents the results of

Table 3. Results of Antimicrobial Analysis

	zone of inhibition (mm)	
sample code	*E. coli*	*S. aureus*
PCEC0	11.32 ± 0.1	10.21 ± 0.1
PCEC1	13.4 ± 0.1	12.83 ± 0.1
PCEC2	13.96 ± 0.1	13.47 ± 0.1
PCEC3	14.06 ± 0.1	15.23 ± 0.2

measuring the diameter of the inhibition zones for *Escherichia coli* and *S. aureus* bacteria in the samples. The PCEC0 sample produced an inhibition zone of 11.32 mm for *E. coli* and 10.21 mm for *S. aureus*. The addition of the *C. caudatus* leaf extract enhanced the inhibition zones for *E. coli*, increasing to 13.40, 13.96, and 14.06 mm for PCEC1, PCEC2, and PCEC3, respectively. Similarly, the inhibition zones for *S. aureus* increased to 12.83 mm (PCEC1), 13.47 mm (PCEC2), and 15.23 mm (PCEC3). These results indicate strong antibacterial activity, as inhibition zones larger than 11 mm are classified as strong, while those smaller than 6 mm are considered weak.[38]

In general, this study shows that the increase in the concentration of the *C. caudatus* leaf extract in the edible coating is directly proportional to the increase in the diameter of the inhibition zone, which identifies a fairly antibacterial solid effect. This is due to the high content of bioactive compounds, specifically polyphenols and flavonoids, in *C. caudatus* leaves, which have antibacterial and antioxidant properties.[20] The *C. caudatus* extract is nontoxic and safe for consumption; however, to mitigate residue concerns, it is recommended that a lightly rinse-coated product be consumed before consumption. Additionally, given that this coating is edible and biodegradable, the residue does not pose significant health risks.

4. CONCLUSIONS

This study successfully developed pectin-chitosan edible films enhanced with the *C. caudatus* leaf extract, demonstrating improved antibacterial properties. The addition of the *C. caudatus* leaf extract to pectin-chitosan coatings significantly enhanced the preservation qualities of tomatoes by reducing weight loss during storage and increasing the antibacterial activity. The integration of bioactive compounds, such as anthocyanins, into the coating matrix contributed to these improvements, making the developed coatings a promising option for extending the shelf life of tomatoes. The enhanced antibacterial effects, particularly against *E. coli* and *S. aureus*, underscore the potential of these coatings for safe and effective food preservation applications.

■ ASSOCIATED CONTENT

Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author.

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Author Contributions

E.S.: Conceptualization, methodology, validation, formal analysis, investigation, writing—original draft. E.F.: Methodology, resources, formal analysis, investigation, writing—review, editing, supervision. Z.S.: Investigation, conceptualization, methodology, formal analysis, resource. T.S.: Formal analysis, term, validation.

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

The authors are grateful for Post Graduate Program Department of Physics, Faculty of Mathematics and Natural Sciences, Universitas Sumatera Utara.

https://doi.org/10.1021/acsomega.4c10344
ACS Omega 2025, 10, 7204−7210

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https://doi.org/10.1021/acsomega.4c10344
ACS Omega 2025, 10, 7204−7210