# Physical Properties of Polyvinyl Alcohol/Chitosan Films with the Addition of Anthocyanin Extract from Butterfly Pea for Food Packaging Applications

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*Received: January 4, 2023*
*Accepted: June 8, 2023*

**DOI:** 10.22146/ijc.80946

**Abstract:** *Composites of polyvinyl alcohol (PVA) and chitosan (CH) polymers, with the addition of anthocyanin (AN) obtained from the butterfly pea flower, were prepared using drop-casting. The composites were made by adding different concentrations of 5–40% anthocyanin with 5 wt.% PVA and 2 wt.% CH solutions (weight ratio of PVA/CH is 80:20). These polymers solution was mixed at 80 °C and dried using the drop-casting method at 25 °C for 48 h. The composites were characterized using a scanning electron microscope (SEM), Fourier-transform infrared (FTIR), ultraviolet-visible (UV–vis spectroscopy), contact angle, antibacterial properties, and food packaging applications. The morphology obtained using an SEM showed that the PVA/CH surface with AN and glycerol was smoother than that of PVA/CH. The increased absorption at a wavelength of 650–700 nm from UV-vis spectroscopy confirmed the success addition of AN. The contact angles of PVA/CH/AN and PVA/CH/GS/AN films were 15°–66°, which showed that the films were hydrophilic. The simple antibacterial test with* Escherichia coli *and* Staphylococcus aureus *showed 16 mm inhibition zone by adding AN. The test results of these characteristics show the potential for using PVA/CH/AN and PVA/CH/GS/AN composite film to be used as an excellent development food packaging material.*

**Keywords:** *anthocyanin; butterfly pea; chitosan; food packaging; polyvinyl alcohol*

## ■ INTRODUCTION

Plastic consumption is increasing in all foods and beverage applications. Plastic has been used in various ways in everyday life, such as food preparation and household equipment. Based on the data from the Ministry of Industry of the Republic of Indonesia in 2018, the national demand for plastic products was 4.6 million tons. It increased by 5% in the last five years. Indeed, this problem could raise pollution because plastic is a substance that is difficult to degrade and is not environmentally friendly [1].

Reducing the amount of plastic usage can prevent environmental pollution [2]. In 2015, the United Nations implemented the 2030 program (Sustainable Development Goals) that applies ecological food packaging materials, which are easy to be recycled, nontoxic, and can be used long-term [3]. Some materials that are often used in the manufacture of food packaging are polyvinyl alcohol (PVA) because they have good mechanical properties and flexibility, water resistance, and high melting temperatures [4]. PVA is often used as a matrix in manufacturing food packaging with a chitosan (CH) filler. CH is a polysaccharide macromolecule mainly obtained from shells of marine animals, such as shrimp, which has biodegradability, compatibility, nontoxic, and easily renewable properties [5]. However, in a previous study, pure CH processed into food packaging materials had hydrophobic properties that reduced the flexibility of a film [6]. In recent years, the PVA/CH film has often been mixed with anthocyanin (AN) [7] to cause food spoilage, as observed in the changes in the food color based on the pH indicator [8].

The mixture of PVA and CH showed much better stability, mechanical properties, and material compatibility than the combination of pure PVA and pure CH [9]. As a result, it became a good development as food packaging material. Several studies have been conducted by mixing PVA/CH with glycerol (GS) to increase the flexibility of films so that they cannot be easily destroyed [10]. AN was previously extracted from red cabbage [11] and purple sweet potato [12]. This work extracted AN from butterfly pea to fabricate PVA/CH film for food packaging. From the Ultraviolet–visible spectroscopy (UV-vis) characterization, it was found that as the AN concentration was increased, the absorbance increased.

## ■ EXPERIMENTAL SECTION

### Materials

The plant used in this study is the butterfly pea flower from Kulon Progo Regency, Special Region of Yogyakarta. PVA (Sigma-Aldrich MW 85,000–150,000) with +99% hydrolyzed as a matrix, CH (Sigma-Aldrich MW medium) as a filler, and 85% GS ($C_3H_8O_3$) as a plasticizer were used in this study. Acetic acid ($CH_3COOH$, 100%) from PT. Brataco was used for diluting CH.

### Instrumentation

The SEM used in the test was the Thermo Fisher (Nicolet iS10). The thickness was measured using a micrometer 150 mm QST-600. The FTIR spectrometer used was the Thermo Fisher (Nicolet iS10). The tests were conducted using UV–vis Ocean Optics (USB4000).

### Procedure

#### *Extraction of AN from the butterfly pea*

The AN extraction process from Butterfly pea used the maceration method [11]. Maceration is a widely used method, and it was quickly applied by adding dried butterfly pea flowers to a 70% ethanol solution [12]. As much as 10 g of dried butterfly pea flowers were soaked in 50 mL of ethanol solution for 3 d at room temperature (25 °C). After 3 d, the results of the first immersion were filtered, and then a second immersion was performed on the dregs of the first immersion with 25 mL of ethanol. The soaked and dried process was conducted in the same way as the first process. The results of the first and second filtrations were mixed and then heated at 50 °C to remove ethanol from the extract. AN levels were measured using the pH differential method [13]. The AN content obtained from butterfly pea is 36 ± 1%wb.

#### *Preparation PVA, CH, and GS*

Fabrication films used several polymer materials. The first process is PVA solution, 50 mL of distilled water was put into a beaker glass, and the distilled water solution was heated at 120 °C. When the temperature of the distilled water solution reaches 80 °C, 5 g of PVA powder was added into the distilled water. PVA powder was dissolved in distilled water for 4 h at 80 °C to obtain a homogeneous solution. To make 2 w/v% CH solution, 10 mL of acetic acid was heated at 80 °C, then added by 2 g of CH powder into the solution. Stirring was carried out for 3 h to obtain a CH solution. The weight ratio of PVA/CH was 80/20. The mixed solution was carried out for 1 h at 60 °C, and then 0.1432 mL of GS was added. The mixed solution was poured into the petri dish and left at 25 °C for 24 h to obtain PVA/CH composite without AN.

Characterization SEM testing was performed by observing the film surface and observing the level homogeneity from the PVA, CH, AN or PVA/CH/AN and PVA, CH, GS, AN or PVA/CH/GS/AN films. If the surface film was smooth, then the homogeneity of the solution was good.

Film thickness measurements were carried out with the aim of knowing the resulting film thickness. The thickness of the film was measured by cutting the size film 1 × 1 cm.

The FTIR test was conducted to determine the functional groups of compounds formed during the mixture of PVA/CH/GS and AN. The test was performed by cutting a 1 mg film to form pellets for the testing film.

Changes in the color of the food packaging film greatly affected the AN levels in the film. The film color would be darker if it was given a large anthocyanin variation, such as 40% composition. UV–vis spectroscopy on the film color was performed by attaching a film to a 1 × 1 cm quartz blank and then observing the transmittance and absorbance of the film.

The film properties can be determined by measuring the contact angle of the film. The contact angle was measured by dripping a drop of water on the surface of the 2 × 2 cm film. Then, the droplets on the surface were observed using a loop, and a photo was taken. The image result was processed using the application to determine the angle produced by the films.

Antibacterial testing of the film used two types of bacteria: *Escherichia coli* (ATCC 27922) and *Staphylococcus aureus* (ATCC 25923). Antibacterial observations were performed by inserting a 1 × 1 cm film into a 12 cm petri dish containing bacteria and then incubating it at 125 °C for 7 d.

### *Application of the PVA/CH/GS/AN films as active packaging for broccoli*

The food packaging film in this study was applied to the broccoli vegetable wrap. The PVA/CH/AN and PVA/CH/GS/AN films were used to wrap broccoli by observing changes in broccoli's color and physical properties for 7 d. Based on the observations, the color change of the film would be known when the broccoli was put in a rotten state. Observations were made at a room temperature (25 °C) and a refrigerator temperature of 4 °C.

## ■ RESULTS AND DISCUSSION

### Characterization of the Films

The mixture of several materials, such as PVA, CH, GS, and AN, in the PVA/CH/AN and PVA/CH/GS/AN films produced a different film surface under the level homogeneity of the solution materials. The surface film test results based on the SEM test are shown in Fig. 1.

The SEM results in Fig. 1(a) show that the film's surface morphology looks smoother than the surface film with the addition of GS and AN. Fig. 1(b) shows the presence of a few small grains, which are suspected of being CH powder. Fig. 1(c) shows black spots on the surface, which are supposed as dregs from the CH extract. Adding more AN concentration made the film darker and produced black holes of dregs on the sample surface [13]. Moreover, a smoother surface can be observed by adding GS, which could cover the spots on the film surface. In Fig. 1(d), adding GS produced the smoothest surface.

The addition of GS could cover the cracks on the film surface. When GS was mixed with a solution of CH and PVA, hydrogen bonds formed, which could inhibit the flexibility of macromolecule chains. Films showed the stiffness of the material mixture, which could improve the mechanical properties, such as tensile strength and flexibility, of thin films for food packaging [14]. Adding AN to the mixture of materials could also

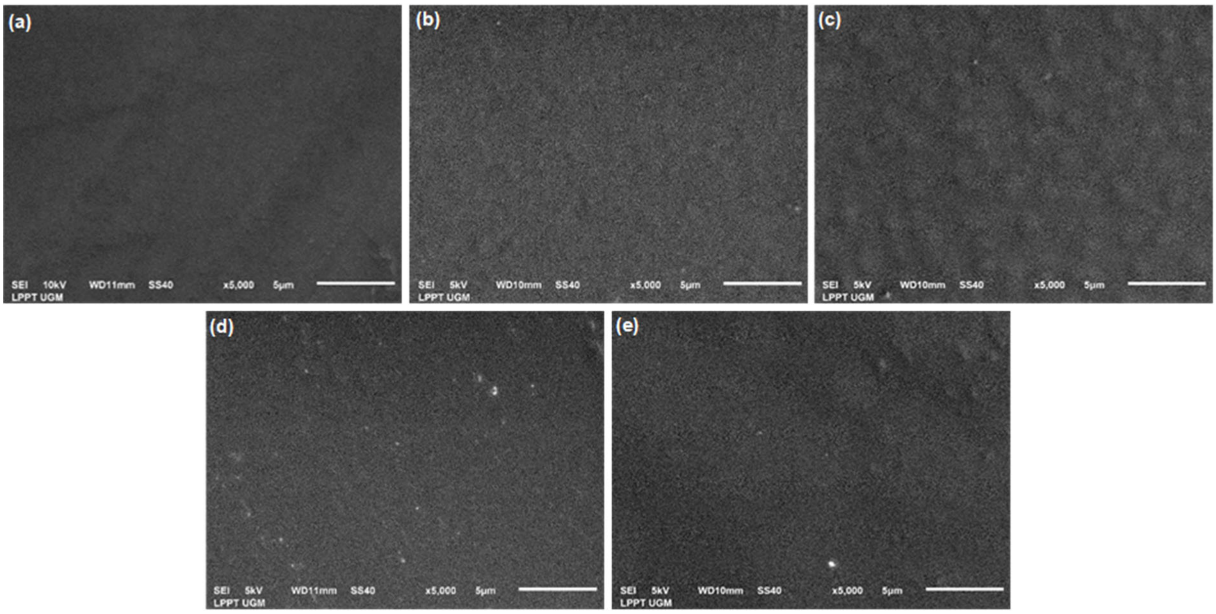



**Fig 1.** SEM morphology: (a) PVA/CH, (b) PVA/CH with 10% AN, (c) PVA/CH with 40% AN, (d) PVA/CH/GS with 10% AN, and (e) PVA/CH/GS with 40% AN

improve the biopolymer microstructure. This result is attributed to O–H groups in the AN structure, which interacted with H bonds in biopolymers, such as CH, resulting in the distribution and dispersion according to the biopolymer matrix [15].

With the addition of AN, PVA/CH films, with or without GS, were involved in several functional groups of PVA, CH, AN, and GS. The functional groups could be observed via FTIR spectroscopy. Fig. 2 shows the FTIR of PVA/CH, adding 5, 10, 20, and 40% AN without GS (Fig. 2(a)) and with GS (Fig. 2(b)). The presence of CH with N–H groups showed at the wavenumber of 3896 $cm^{-1}$, and O–H groups showed the existence of PVA and AN. With the addition of AN, the wavenumber increased drastically.

At the absorption peak of 2931 $cm^{-1}$, symmetrical and asymmetrical strains of C–H were produced in various layers. The absorption peak in the PVA/CH layer shows the C–O group at a wavenumber of 2331 $cm^{-1}$, which illustrates a deformation $CH_3$ absorption event [16]. The increase in peak intensity in the 1635 $cm^{-1}$ wave indicated a rise in the amide absorption area. The aromatic ring from the 1635 $cm^{-1}$ anthocyanin alkene bond is a C=C group that produces 1118 $cm^{-1}$ C–N group as a mixed group of CH and AN [17].

## Transparency

The PVA/CH and PVA/CH/GS films with 5, 10, 15, 20, 30, and 40% AN produced different color. Films with or without GS and AN had a blue-purple color. The addition of AN had a darker film color due to neutral quinoids and anionic quinoidal bases in equilibrium [18]. AN formed quinoidal blue due to the bathochromic displacement caused by the AN molar absorption coefficient–the transparency and UV-vis results tested on films with different AN variations.

Based on the transparency test results, different AN created darker color film. The lightest color was found in the PVA/CH and PVA/CH/GS films with an additional 5% AN variation, and the darkest color was found in the PVA/CH and PVA/CH/GS films with 40% AN with or without the addition of GS. Fig. 3 shows the transparency of PVA/CH/AN and PVA/CH/GS/AN films, where the dark color of AN can protect food from spoilage because it absorbs UV rays. Thus, avoiding UV radiation by slowing food spoilage [19].

Fig. 4(a) and 4(b) show the UV-vis films with the addition of 10–40% AN. The PVA/CH/AN and PVA/CH/GS/AN films were gradually increased at 580 and 630 nm peaks. The addition of AN produced a blue-purple color at the 550–650 nm peak. The ratio changes in the intensity of absorbance of the butterfly pea by increasing the AN at 580 and 630 nm can be attributed to the stability of the AN in the film. Mainly, the color change in the food packaging film of the butterfly pea resulted in a shift in the increase in the maximum AN absorption peak [8].

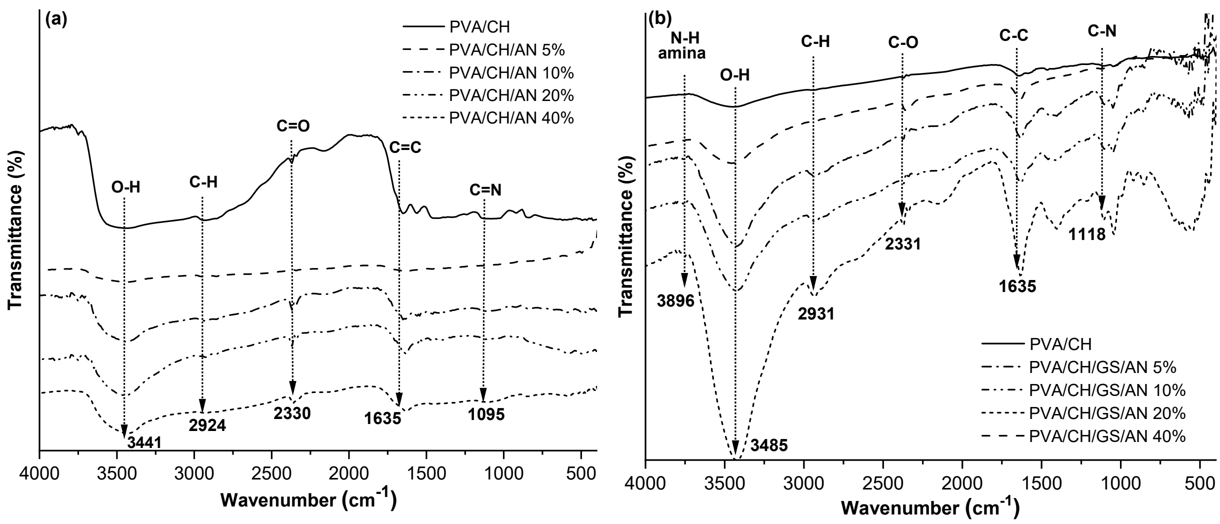



**Fig 2.** FTIR of PVA/CH/AN films: (a) without and (b) with glycerol

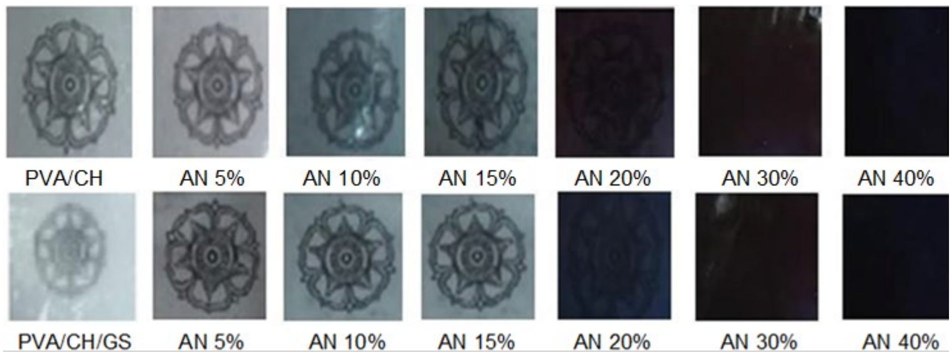



**Fig 3.** Transparency of PVA/CH and PVA/CH/GS with the addition of AN

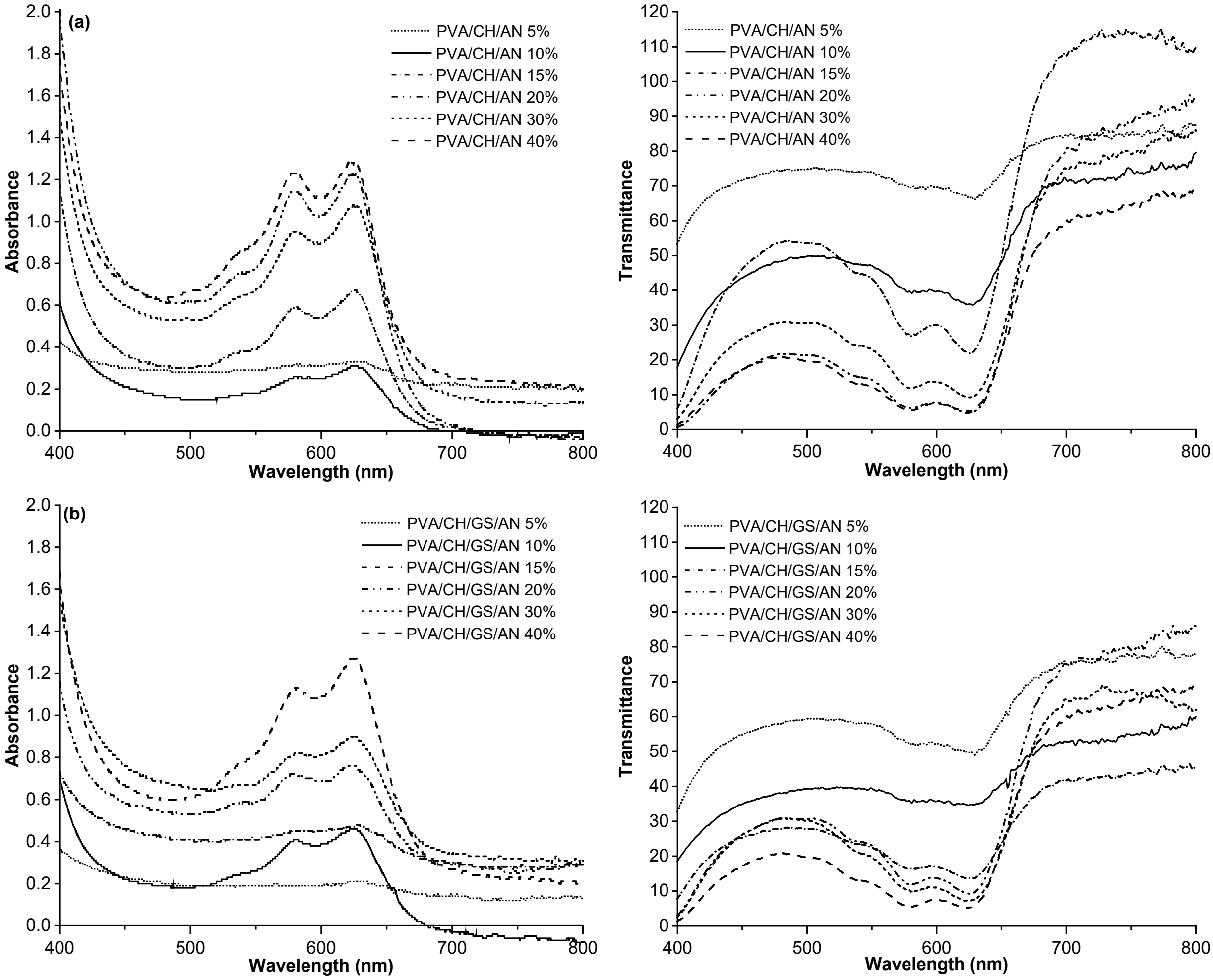



**Fig 4.** UV-vis film: (a) PVA/CH/AN and (b) PVA/CH/GS/AN

Meanwhile, Fig. 4(a) and 4(b) found that the transmittance gradually decreases by increasing AN composition at 200–300 nm. It is shown that there were more reflections than wave absorption. With a great reflection, the films could not block UV rays, considering the acceleration of the oxidation of food due to UV radiation. The PVA/CH/AN and PVA/CH/GS/AN films have good potential when used as food packaging. The UV-vis test data showed that the more AN added to the film, the more food oxidation would be prevented.

AN affects film contact angle with the increasing number of AN resulting in smaller contact angle values

[16]. Table 1 shows the composition of PVA/CHA/N 40% (40° ± 4°) and PVA/CH/GS/AN 40% (15° ± 1°). All the contact angles of the films with the addition of AN had a hydrophilic contact angle of < 60°. In brief, the addition of AN caused the film to absorb water.

Water absorption in the film was caused by pores on the film surface, which formed when the hydroxyl group bonded with PVA and CH. The highest contact angles were found in the PVA/CH (65° ± 1°) and PVA/CH/GS/AN 5% (66° ± 4°) films. This result can be attributed to the ionic interaction between CH and pectin, which reduced the hydrophilic groups in PVA and AN [20]. The higher the number of hydrophobic acetyl groups in CH, the higher the contact angle. The contact angle is also related to the surface roughness of the film, with a rough surface containing grains in the PVA/CH composition, as shown in the SEM results in Fig. 1.

## Antibacterial Activity

Based on several characterization tests, such as the contact angle test to determine the hydrophilic nature of the film and the addition of CH polymer to films, bacterial testing was required to observe the antibacterial properties of CH and the effect of the addition of AN on the hydrophilic properties of films to inhibit bacteria when films were applied as food packaging. Mixing PVA and CH in manufacturing film food packaging can increase antibacterial activities. CH is a hydrophobic polysaccharide macromolecule, so bacteria cannot easily invade [21]. A mixture of CH and PVA was used to reduce the hydrophilic nature of PVA so that bacteria cannot easily infect the film [22]. The composition of PVA/CH/AN 5% and PVA/CH/AN 10% had an excellent absorption property because there are $NH_2$ and OH groups in the mixture. Accordingly, the PVA and CH materials showed film quality in inhibited antimicrobials but had poor stretching [23]. Hence, GS was added to PVA/CH/GS/AN 5% and PVA/CH/GS/AN 40%.

The addition of AN in-manufacturing food packaging films also affected their antibacterial properties. PVA/CH/AN *E. coli* (Fig. 5(a)) and PVA/CH/AN 10% *S. aureus* (Fig. 5(b)) films had clear zones of no microbial growth around 16 mm. In contrast, with the addition of GS, PVA/CH/GS/AN 5% *E. coli* (Fig. 5(e)) and PVA/CH/GS/AN 5% *S. aureus* (Fig. 5(f)) had clear zones of no microbial growth around 5 mm. Meanwhile, with the addition of more anthocyanin, PVA/CH/AN 5% *E. coli* (Fig. 5(c)), PVA/CH/AN *S. aureus* (Fig. 5(d)), PVA/CH/GS/AN 40% *E. coli* (Fig. 5(g)), and PVA/CH/GS/AN 40% *S. aureus* (Fig. 5(h)) had no clear zones of no microbial growth around. The area is decreased because the increasing levels of AN cover the function of CH as an antibacterial. Fig. 6 shows the antibacterial zone of all variants. The addition of 40% AN could inhibit the antibacterial properties of CH. Additional AN is significantly suitable for the body because they contain antioxidant properties, which can be applied to food and beverages to reduce oxidative stress [24].

## Potential Test Films

After antibacterial observations, the potential of films was tested to examine the ability of the PVA/CH/AN and PVA/CH/GS/AN films to increase the shelf life of food. The test was performed on broccoli, a vegetable that quickly rots. The test was conducted by wrapping broccoli using several types of films, such as

**Table 1.** Contact angles of the (a) PVA/CH/AN and (b) PVA/CH/GS/AN films

| **(a)** Materials | Contact angle (°) | **(b)** Materials | Contact angle (°) |
|---|---|---|---|
| PVA/CH | 65 ± 1 | PVA/CH/GS | 62 ± 4 |
| PVA/CH/AN 5% | 65 ± 1 | PVA/CH/GS/AN 5% | 66 ± 4 |
| PVA/CH/AN 10% | 56 ± 2 | PVA/CH/GS/AN 10% | 51 ± 1 |
| PVA/CH/AN 15% | 62 ± 4 | PVA/CH/GS/AN 15% | 42 ± 6 |
| PVA/CH/AN 20% | 51 ± 2 | PVA/CH/GS/AN 20% | 60 ± 1 |
| PVA/CH/AN 30% | 51 ± 2 | PVA/CH/GS/AN 30% | 43 ± 4 |
| PVA/CH/AN 40% | 39 ± 4 | PVA/CH/AN 40% | 15 ± 1 |

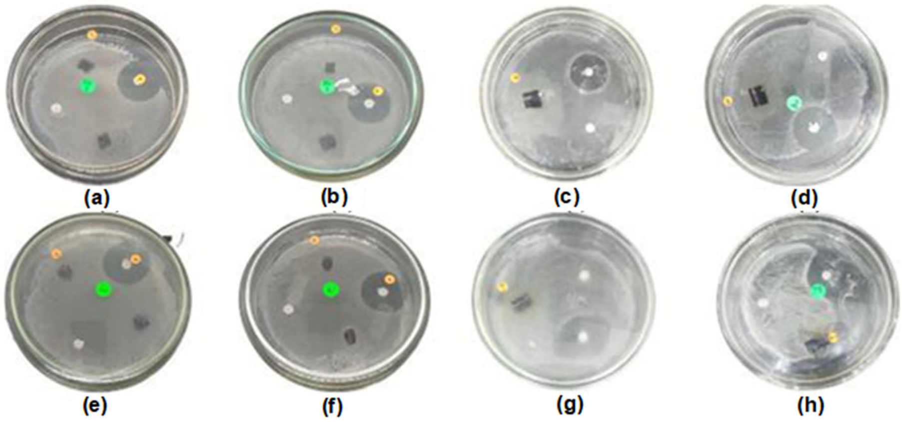



**Fig 5.** Bacterial test films: (a) PVA/CH/AN 10% *E. coli*, (b) PVA/CH/AN 10% *S. aureus*, (c) PVA/CH/AN 40% *E. coli*, (d) PVA/CH/AN 40% *S. aureus*, (e) PVA/CH/GS/AN 5% *E. coli*, (f)PVA/CH/GS/AN 5% *S. aureus*, (g) PVA/CH/GS/AN 40% *E. coli*, and (h) PVA/CH/GS/AN 40% *S. aureus*

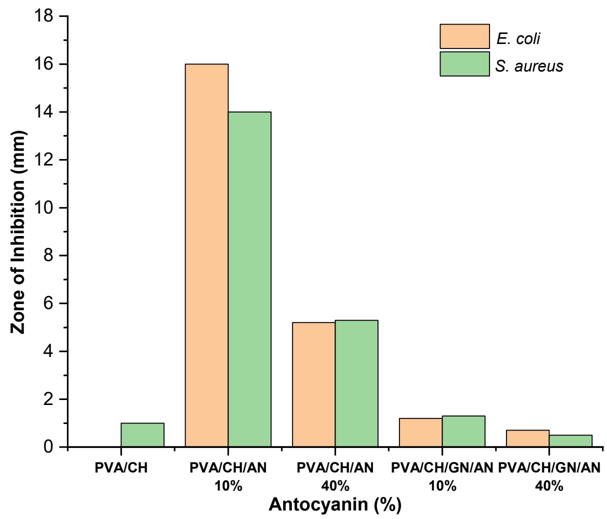


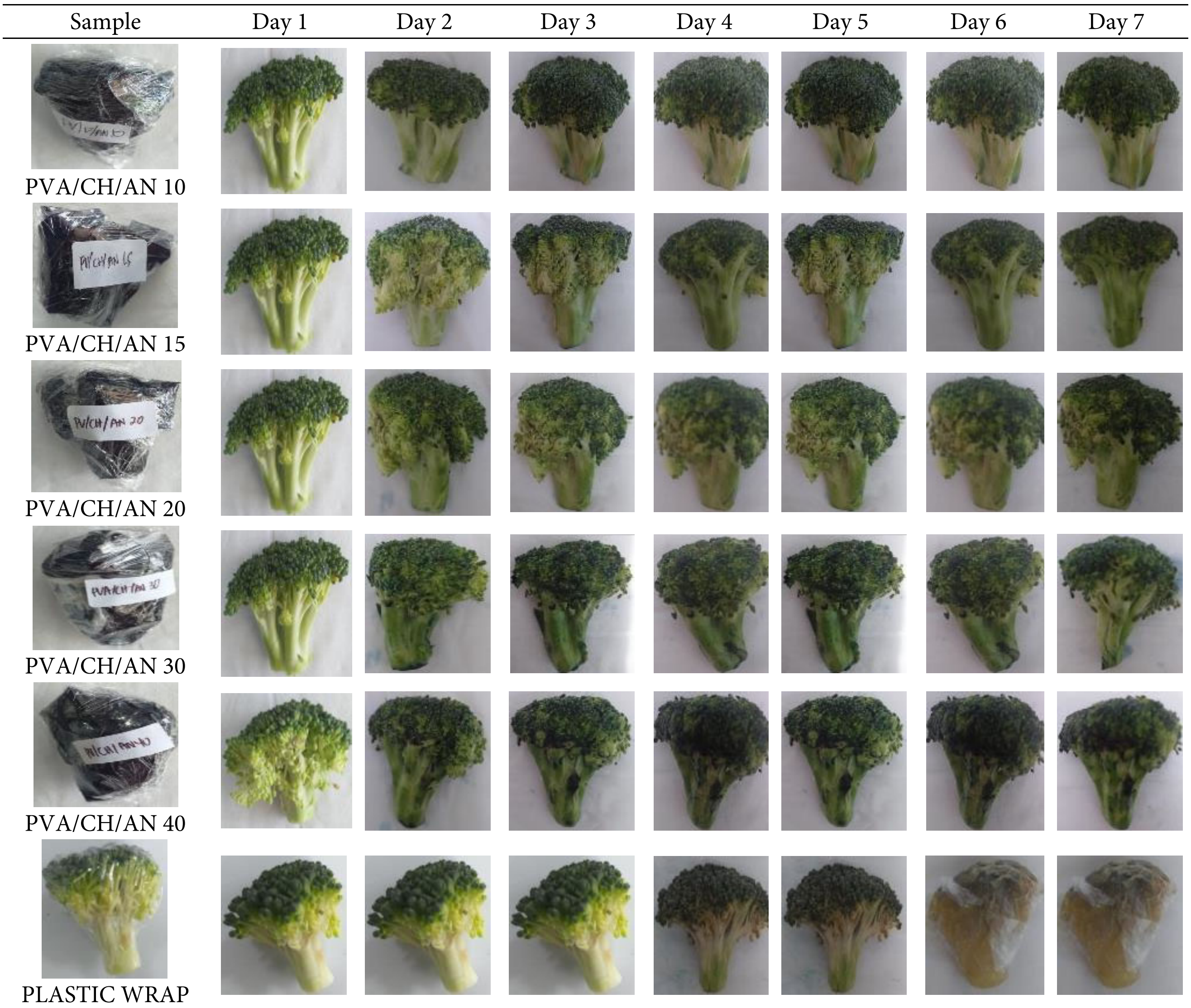

**Fig 6.** Zone of inhibition with different concentrations of anthocyanin

PVA/CH/AN and PVA/CH/GS/AN, with AN variation. Table 2 shows the application of the PVA/CH/AN film in the refrigerator. The test was performed by inhibiting the broccoli wrapped in films and then leaving it at refrigerator temperature. Observations were made for 7 × 24 h.

Table 3 shows the application of PVA/CH/GS/AN in the refrigerator. Based on the film observation, the first fungus in the broccoli was found in the food packaging film with the PV/CH/GS/AN composition on the fourth day at room temperature. At room temperature, bacteria multiplied, whereas, at refrigerator temperature, bacterial growth is inhibited because of the cold. The food packaging film with GS had an oilier surface than the film without GS. Hence, the addition of GS made the

**Table 2.** Application of the PVA/CH/AN film in a refrigerator

| Sample | Day 1 | Day 2 | Day 3 | Day 4 | Day 5 | Day 6 | Day 7 |
|---|---|---|---|---|---|---|---|
|  PVA/CH |  |  |  |  |  |  |  |
|  PVA/CH/AN 5 |  |  |  |  |  |  |  |

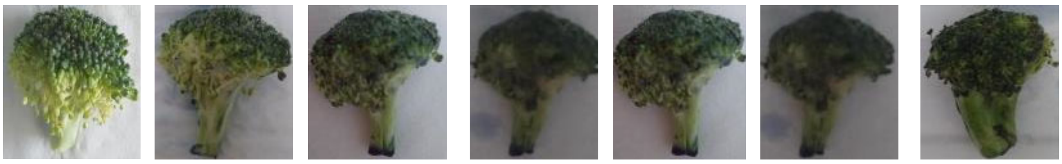
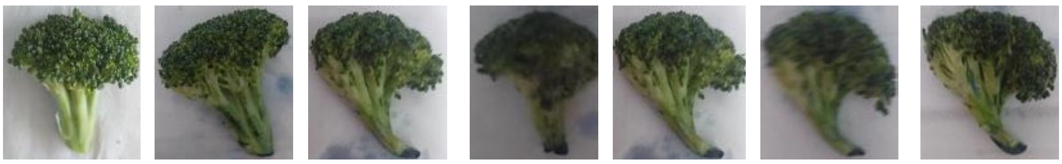



food oxidation occur quickly, such that bread wrapped using a PV/CH/GS/AN film had more moldings [13].

Furthermore, adding GS to the food packaging film resulted in the plasticization effect of GS and PVA, which can reduce the function of CH as an antibacterial. The antibacterial properties of CH bind positively charged amino groups to negatively charged sites on the bacterial wall, which binds to the cytoplasmic membrane, preventing the diffusion of nutrients into cells while destroying bacteria [25]. Adding GS also created a high level of interaction between the active groups of PVA, with the polymer leaving few free amino groups in the CH chain. This condition reduced the antibacterial activity of the mixed material when applied to food packaging.

The color of AN can changes in acid, neutral, and wet conditions. In addition, the film in the acidic condition is reddish-purple, the film in the neutral condition is blue-purple, and the film in the wet condition is blue green. The PVA/CH/AN and PVA/CH/GS/AN films with all variations provided the food information with discoloration of films when applied as food packaging. In short, AN added in PVA/CH/AN and PVA/CH/GS/AN films can be used as a pH indicator considering the color produced.

**Table 3.** Application of the PVA/CH/GS/AN film in a refrigerator

| Sample | Day 1 | Day 2 | Day 3 | Day 4 | Day 5 | Day 6 | Day 7 |
|---|---|---|---|---|---|---|---|
|  PVA/CH/GS |  | | | | | | |
|  PVA/CH/GS/AN 5 |  | | | | | | |
|  PVA/CH/GS/AN 10 |  | | | | | | |
|  PVA/CH/GS/AN 15 |  | | | | | | |
|  PVA/CH/GS/AN 20 |  | | | | | | |
|  PVA/CH/GS/AN 30 |  | | | | | | |
|  PVA/CH/GS/AN 40 |  | | | | | | |
|  PLASTIC WRAP |  | | | | | | |

## CONCLUSION

In this study, PVA/CH/AN and PVA/CH/GS/AN films with the addition of AN showed some change in physical properties. The highest contact angle film was observed in a 10% AN concentration of PVA/CH/AN composition. The film's surface looked smoother in the composition of PVA/CH/GS/AN compared to PVA/CH/AN. The FTIR test showed that the PVA/CH/AN and PVA/CH/GS/AN compositions with AN variations (5–40%) contained an AN peak. UV-vis spectroscopy shows the increase at 550–650 nm with increasing AN composition. The potential film test showed a change in the film color when the broccoli was rotting, with the fastest decay occurring at room temperature. The PVA/CH/AN composition had clear antibacterial zones of no microbial growth around 16 mm. Briefly, adding AN in small amounts, i.e., 5 and 10%, increased the antibacterial value of the films, and therefore it can be applied as a food packaging film.

## ACKNOWLEDGMENTS

We would like to thank Universitas Gadjah Mada for the research facilities at the Laboratory of Instrumentation and Material Physics.

## AUTHOR CONTRIBUTIONS

Siti Khanifah and Alda Dwi Karina Legowo conducted the experiment. Siti Khanifah, Ari Dwi Nugraheni, and Sholihun wrote and revised the manuscript. All authors agreed to the final version of this manuscript.

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