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ORIGINAL ARTICLE

Effect of Cross-Linking with *Aloe vera* Gel on Surface and Physicochemical Properties of Edible Films Made from Plantain Flour

Tomy J. Gutiérrez[1,2] · **Gema González**[3]

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Abstract Films made from plantain flour with incorporation of different concentrations (0, 2, 4 and 6 %) of a natural filler (*Aloe vera* gel - Av gel) were obtained by the casting method. The aim of this paper was to characterize the surface and physicochemical properties on plantain flour edible films. The average molecular weight, moisture, infrared spectroscopy, contact angle and mechanical properties were determined. Also, microstructural characterization was performed by atomic force microscopy and scanning electron microscopy. The Av gel produced cross-linking on the starch that is found in plantain flour, resulting in films with the following characteristics: smoother, more transparent, more rigid and plastics, less moist and with more hydrophobic surfaces. Finally, the surface properties of these materials were defined by the surface energy of material, which depends on the intermolecular interactions such as van der Waals-type interactions (hydrogen bond) and new bonds (cross-linking) formed between biopolymeric chains (plantain flour).

Keywords Edible films · Cross-linking · Natural fillers · Surface

✉ Tomy J. Gutiérrez
tomy.gutierrez@ciens.ucv.ve; tomy_gutierrez@yahoo.es

[1] Departamento Químico Analítico, Facultad de Farmacia, Universidad Central de Venezuela, Apartado 40109, Caracas 1040-A, Venezuela

[2] Instituto de Ciencia y Tecnología de Alimentos, Facultad de Ciencias, Universidad Central de Venezuela, Apartado 47097, Caracas 1041-A, Venezuela

[3] Centro de Ingeniería de Materiales y Nanotecnología, Instituto Venezolano de Investigaciones Científicas, Apartado 20632, Caracas 1020-A, Venezuela

Introduction

In the last decade, the elaboration of edible films based on bio-renewable polymers has been intensively studied in order to reduce the consumption of petroleum-derived polymers [1]. Among plant-derived materials, starch offers several advantages as a raw material for the plastic industry, including its low cost, non-toxicity, biodegradability, composability, and worldwide availability [2]. Nonetheless, properties such as mechanical resistance, water sensitivity and barrier properties have to be improved to compete with films of synthetic polymers [3–6]. Different alternatives have been proposed to improve the mechanical and physicochemical properties of edible films based on starch, among which are chemical and/or physical modifications, which seem to have been very effective [7]. Also, the properties of polysaccharide-based films can be modified and enhanced by blending starch with other biopolymers [8].

Aloe vera (Av) gel is a polysaccharide rich in over 75 potentially active compounds that has been used for centuries due to its medicinal and therapeutic properties [9–14]. For example, tannic acid that is found in the Av gel allows wound healing. Likewise, Robson et al. [15] found lactates in the Av gel, which can have analgesic properties provoking an aspirin-like effect. Av gel is obtained from the leaf pulp of Av and consists of approximate 99.5 % water and 0.5 % solid material including polysaccharides, vitamins, minerals, enzymes, phenolic compounds and organic acids [12]. The major polysaccharides are cellulose, hemicellulose and storage polysaccharides such as glucomannans, mannose derivatives and acetylated compounds.

Av gel has drawn attention in scientific community due to its unique biological properties such as anti-inflammatory, anti-cancer, anti-oxidant, anti-diabetes, anti-septic, anti-biotic and anti-microbial [16–21]. Besides, Av gel has important

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implications for wound healing [21], the reduction of cholesterol and triglyceride levels in blood as well as prevention of the gastrointestinal, renal and cardiovascular problems [10, 20, 22, 23]. The healing properties of Av gel are mainly attributed to polysaccharides present in plant, while the antiseptic and antimicrobial activity are related with the presence of antiseptic agents such as lupeol, salicyclic acid, urea nitrogen, cinnamonic acid, phenols and sulfur, which have inhibitory activities against fungi, bacteria and viruses [12].

More recently, the Av gel has found its place in the food industry as an ingredient of functional foods in ice-cream and beverages [24, 25], and due to its antifungal activity, it has been applied as an edible coating (plain or in combination with other components) to extend the post-harvest storage of arctic snow [26], table grapes [23, 27], sweet cherry [28], apple slices [29] and papaya fruits [30]. Moreover, coatings based on Av reduce weight loss and ethylene production in raw peaches and plums [31]. In a recent study with fresh-cut kiwifruit, Av was effective in reducing pectin depolymerization and microbial proliferation. Furthermore, sensorial quality was enhanced [32]. Additionally, an important advantage of Av gel at industrial level is its easy preparation.

On the other hand, organic acids such as citric acid, among others, that are found in Av gel can be potential cross-linking agents and could improve the physicochemical properties of edible films [33]. According to Majzoobi and Beparva [34], the organic acids have large applications in the food industry. For this reason, understanding its action in food matrices is important. In this regard, organic acids may alter the physicochemical properties of starches, since these may originate crosslinking reactions between the starch polymer chains [34].

In this sense, recently Gutiérrez and Álvarez [35], obtained an effect of cross-linking with the addition of *Aloe vera* (Av) gel in the plantain flour-based films, which caused a lower in vitro digestibility rate. According to the authors, the effect of cross-linking obtained was caused by organic acids contained in the Av gel, which can act as cross-linking agents between the starch chains. It is worth noting that the cross-linking reactions increase the average molecular weight of the starch and introduce chemical bridges between different molecules, thus increasing thermal resistance of the films [36, 37]. In addition, these reactions reduce the high sensitivity to moisture of these materials, and enhance both the mechanical and barrier properties [37].

Moreover, Av contains several polyphenolic compounds that can improve the physico-chemical and mechanical properties of the edible films, producing a plasticizing effect on the films made from starch [38].

On the other hand, recent studies have demonstrated the potential of plantain flour as a renewable resource for food packaging production. In this sense, the plantain flour has higher performance than the plantain starch. Therefore, the plantain flour has lower production costs, which brings as a result that this matrix has greater competitiveness with respect to synthetic materials made from petroleum [39–41].

Therefore, the objectives of the present work were to study the potential benefits of combining the two complex polymeric matrices (plantain flour and Av gel) and the effect of incorporating different concentrations of Av gel on the surface and physicochemical properties of edible films made from plantain flour.

Experimental

Materials

The plantain flour and Av gel were obtained by the method recently described by Gutiérrez and Álvarez [35]. Food grade glycerol (Aldrich, product code: G7893) was used as a plasticizer for the formation of the films. Dimethyl sulfoxide (DMSO) (Aldrich, product code: 472,301) was used as solvent for determining the average molecular weight of the developed films.

Film Formation

A group of films were prepared from film-forming solutions (*FFS*) made by mixing 2 % *w/v* of native plantain flour and **1.5 % *w/v* of glycerol** in 500 mL distilled water. Different concentrations of Av gel (0, 2, 4 and 6 %) were added to the films. Another second group of films were prepared containing 2 % w/v of native plantain flour, **0 % w/v glycerol** and 500 mL distilled water with different concentrations of Av gel (0, 2, 4 and 6 %). The latter group was prepare to evaluate the potential plasticizing effect of Av gel. However, as will be discussed later, these films did not meet the minimum required mechanical properties, since they were very fragile, which made unviable its handling to carry out the tests. These films were difficult to handle to perform the different characterization tests. Both groups of films were prepared following the methodology described by Gutiérrez et al. [40, 41]. The solutions were then heated in a water bath with constant shaking at 98 °C for 30 min. to ensure starch gelatinization and inactivation of enzymes contained in the Av gel [42]. The gel obtained was poured into stainless steel trays 40 × 30 cm, and dried in a Mitchell dehydrator (Model 645,159) for 24 h at 45 °C. The resulting thermoplastic flour films containing glycerol: plantain flour with 0 % of Av gel (*TPF-PF*), plantain flour with 2 % of Av (*TPF-PF + 2 % Av*), plantain flour with 4 % of Av (*TPF-PF + 4 % Av*) and plantain flour with 6 % of Av (*TPF-PF + 6 % Av*) were then carefully removed from the casting molds. Before characterization, the films were conditioned at ~57 % relative humidity (*RH*) for one week at 25 °C. During this period, containers were

protected of light in a dark room in order to avoid the photo-oxidation of polyphenolic compounds.

Characterization of the Films

Determination of Average Molecular Weight (M_w) of the Films Developed by the Use of Ostwald Viscometer

DMSO was used as solvent for performing solutions at 1 % *w/v* of each film assessed. Next, successive dilutions were prepared different solutions at 0.125, 0.25 and 0.50 % w/v. Afterwards, the resistance time to the flow of the solutions prepared was determined, taking as reference the passage between two areas marked in Ostwald viscometer (Cannon-Fenske 200 series, model 120,205, IVA, Argentina) at 25.0 ± 0.1 °C. After obtaining the residence time of different solutions prepared, we proceeded to determine the relationship η_{exp}/C through Eq. 1, which was empirically demonstrated by Kramer [43] and Huggins [44]:

$$\eta_{exp}/C = {}^{1}/_{C} + {}^{t-t_o}/_{t_o} \quad (1)$$

where $\eta_{exp} = (\eta - \eta_o/\eta_o)$, intrinsic viscosity; η_o = viscosity of the pure solvent; C = concentration of the solutions % *w/v*; t = residence time of the solutions and t_o = residence time of the pure solvent.

By using Eq. 2:

$$\eta_{exp}/C = \eta + KC\eta^2 \quad (2)$$

And graphing η_{exp}/C vs C, a straight of cut point equal to η was obtained. At the same time, using the Mark-Houwink-Sakurada-Staudinger equation (Eq. 3):

$$\eta = KM_w{}^a \quad (3)$$

where: η = intrinsic viscosity; M_w = polymer molecular weight; K and ^{a} = constants reported for the solvent used (DMSO); K = 0.0112 mL/g at 25 °C and ^{a} = 0.72 at 25 °C.

Finally, the average molecular weight (M_w) of the films tested was calculated from Eq. 4:

$$M_w = \left({}^{\eta}/_{K}\right)^{1/a} \quad (4)$$

Moisture Content

The moisture content of the films was calculated using Eq. 5. It was analyzed gravimetrically by drying the samples for 24 h at 105 °C [45].

$$\%\text{moisture} = \frac{\text{initial mass}_{(g)} - \text{final mass}_{(g)}}{\text{initial mass}_{(g)}} \text{ x } 100 \quad (5)$$

The experiments were carried out by triplicate.

Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy (ATR/FTIR)

The IR spectra of the films were determined using an infrared spectrometer (FTIR) (Nicolet 8700). The spectra were obtained in an Attenuated Total Reflectance mode (ATR) between 700 and 4000 cm^{-1}, using 40 scans at a resolution of 4 cm^{-1}. Each sample was scanned three times observing good reproducibility.

Atomic Force Microscopy (AFM)

The films surface were analyzed by AFM using an Agilent 5500 in Tapping Mode with silicon nitride (Si_3N_4) tips. Sections of the films were stuck to glass slides using double-sided adhesive tape and the top surface was imaged by AFM. The surface analyzed was the side of the surface exposed to drying air during film preparation. The AFM images were taken at center and periphery of surface of the films. The surface analyzed was 20 × 20 μm. The cantilever had a spring constant of 0.2 N m^{-1} and a Si_3N_4 V-shaped tip, which was positioned over sample at ambient conditions. The cantilevers were driven into oscillation at 10 % below their fundamental resonant frequency (typically around 320 kHz) and the feedback loop operated using amplitude control. The set-point was kept at minimal value (~90 % of the free amplitude) that enabled stable tracking of the sample surface in order to avoid any damage to sample. Each scan was carried out at a relatively slow rate of 0.5 Hz to allow proper imaging of the corrugated surfaces of the films. The AFM images were processed with PicoView image software.

Using the topographical data from AFM micrographs and the software previously mentioned, roughness was evaluated. The average roughness (Ra) is the arithmetic mean of the deviations of the heights:

$$Ra = \frac{\sum_{i=1}^{N} Z_i - Z}{N} \quad (6)$$

with

$$Z = \sum_{i=1}^{N} \frac{Z_i}{N} \quad (7)$$

where Zi is the height value (nm), Z is the arithmetic mean of the height (nm) and N is the number of observed points. The mean square roughness (Rq) has the following expression [46]:

$$Rq = \sqrt{\sum_{i=1}^{N} \frac{(Z_i - Z)^2}{N}} \quad (8)$$

Table 1 Surface energy components of probe liquids for contact angle measurements

(mN/m)	Ethylene glycol	Di-iodomethane
Total surface energy (γ)	47.7	50.8
Dispersive component (γ^d)	30.1	48.5
Polar component (γ^p)	17.6	2.3

Contact Angle

The contact angles (θ) were calculated by analyzing the images to determine the angles formed by the intersection of the liquid-solid interface (drop of water-surface of the film) and the liquid-vapor interface (tangent to the boundary of the drop) [47]. An USB Digital Microscope (model DIGMIC200X, China) equipped with Image Analysis Software 220X 2.0MP video, with 0.0001° precision was used for determining the contact angle. Contact angle of the surface exposed to drying air during film preparation were determined by depositing a drop of distilled water (2 μL) onto the film surface. The drop of distilled water was carefully dropped on films through a syringe (KDL Corp., Shanghai, China) and contact angles were quickly measured before swelling started. The measurements were carried out in open air, at a room temperature of 25 °C. The contact angles were expressed as the average of 12 measurements per film.

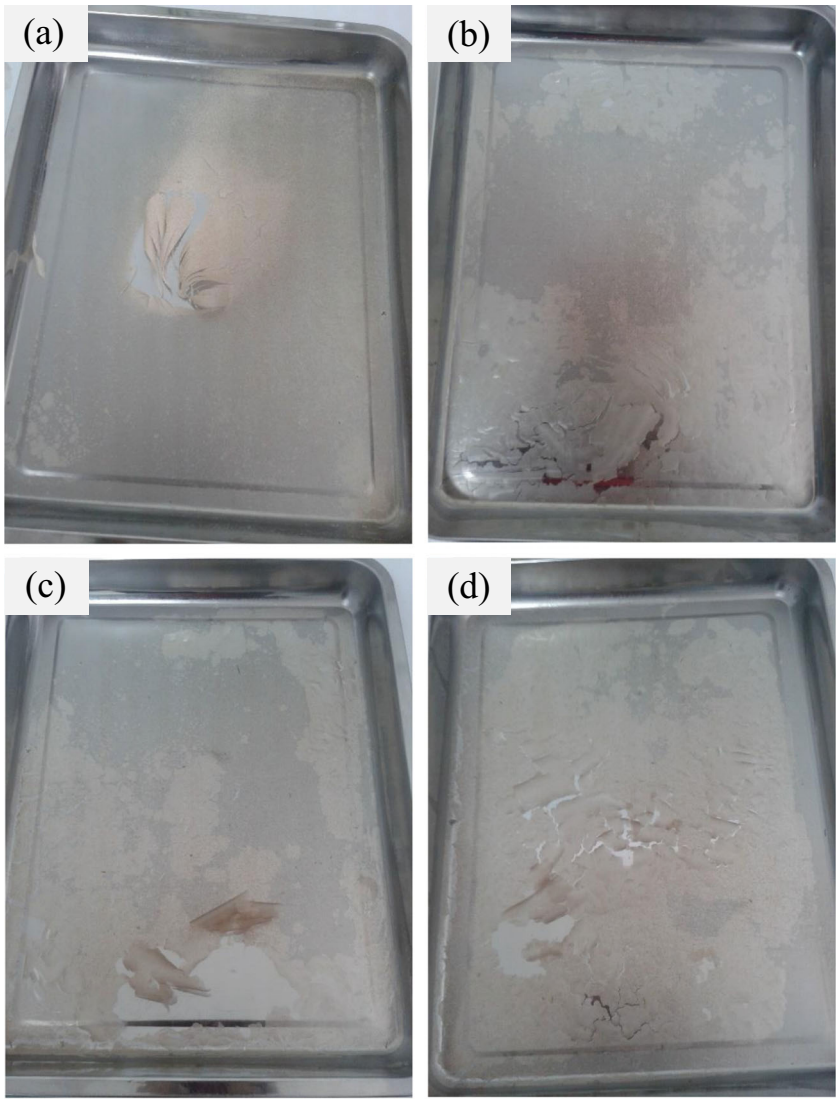

Fig. 1 Appearance of the different films studied without glycerol: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

The surface energy of the materials and their polar and dispersive components were determined by Owens and Wents equation [48].

$$0.5\gamma_2\,(1+cos\theta) = \left(\gamma_1^d\gamma_2^d\right)^{0.5} + \left(\gamma_1^p\gamma_2^p\right)^{0.5} \quad (9)$$

where 1 and 2 refers to the solid and liquid, respectively; γ^d is the dispersive component and γ^p the polar component of the surface energy; θ is the contact angle. If the contact angles made by two liquids of known (ethylene glycol and di-iodomethane) γ^d and γ^p are measured, it is possible to solve the Eq. (9) and infer the γ^d_1 and γ^p_1 for the material. Then, the total surface energy was estimated from Eq. 10:

$$\gamma_1 = \gamma_1^d + \gamma_1^p \quad (10)$$

The surface energy components of probe liquids used for the contact angle measurements are listed in Table 1 [49].

Scanning Electron Microscopy (SEM)

The surface of the films exposed to drying and the fracture surface of each material were analyzed using a JEOL JSM-6460 LV instrument. Films were cryo-fractured by immersion in liquid nitrogen. For the analysis of both surfaces, the films were mounted on bronze stubs and sputter coated with a thin layer of gold for 35 s.

Uniaxial Tensile Tests

The mechanical properties were carried out for films cut in rectangular sections (16 mm long and 8 mm width). Samples were mounted and clamped with grips in a texturometer (TA.XT2i Texture Analyzer, Stable Micro Systems, Surrey, UK) (5 kg) to determine force-distance curves. The samples were stretched at a constant speed of 0.5 mm/s at 25 °C until break.

The overall stress acting on sample during tension was expressed as the so-called true tensile strength (σ), which is the force normal to the film cross-section F (N) divided by initial area A_0 (m^2) of sample [50]:

$$\sigma = {}^{F}/_{A_o} \quad (11)$$

The overall strain (ε) of films was expressed as percent of the initial height as follows:

$$\varepsilon = {}^{L}/_{L_o}\ \text{x}\ 100\% \quad (12)$$

where Lo and L (mm) are initial and final longitude of film after deformation, respectively.

The mechanical properties at break, maximum stress (σ_m) and strain at break (ε_b) were obtained from stress-distance curves. For this, these curves were transformed into stress-strain curves, as outlined in ISO 527–2 [51]. Young's modulus (E) was determined from the slope of linear regression of the stress-strain and toughness (T) was determined as area under stress-strain curves. At least 10 tests for each film were performed.

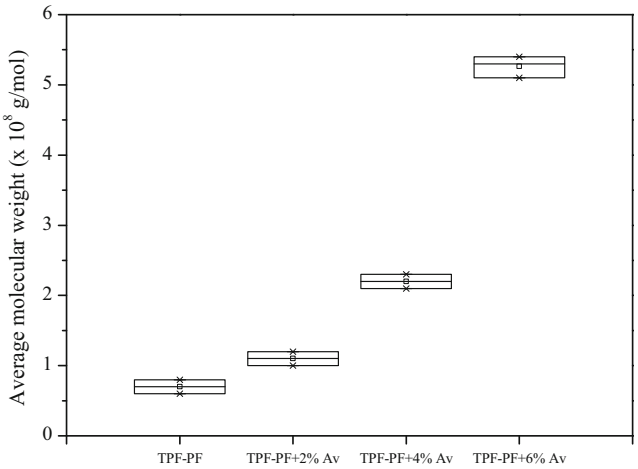

Fig. 2 Average molecular weight of the films based on: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

Statistical Analysis

Experimental data was analyzed using Statgraphics Plus 5.1. software (ManugisticsCorp., Rockville, MD). The one-way ANOVA procedure followed by Duncan's multiple range tests was adopted to determine the significant difference ($p < 0.05$) between treatment means and results were expressed as means ± SD.

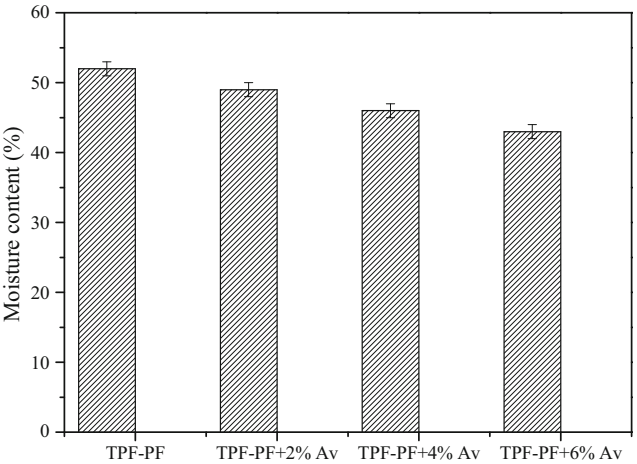

Fig. 3 Moisture content of the films based on: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

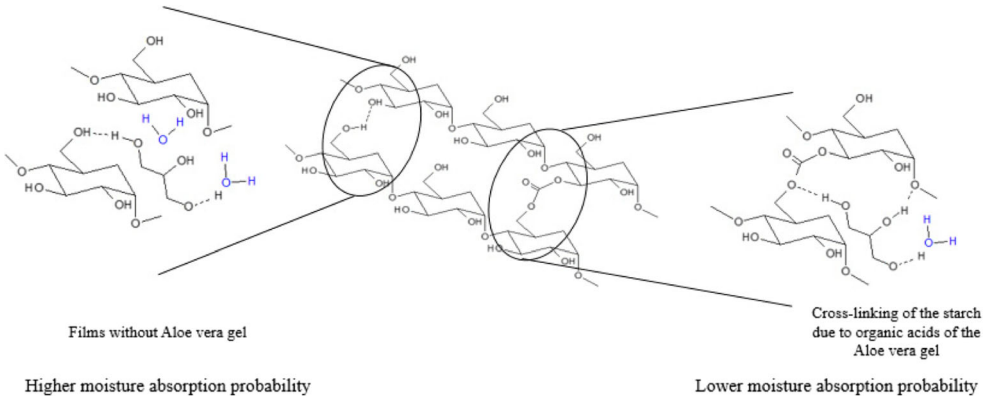

Fig. 4 Mechanism proposed to explain the effect caused by organic acids contained in Av gel on moisture content of the developed films

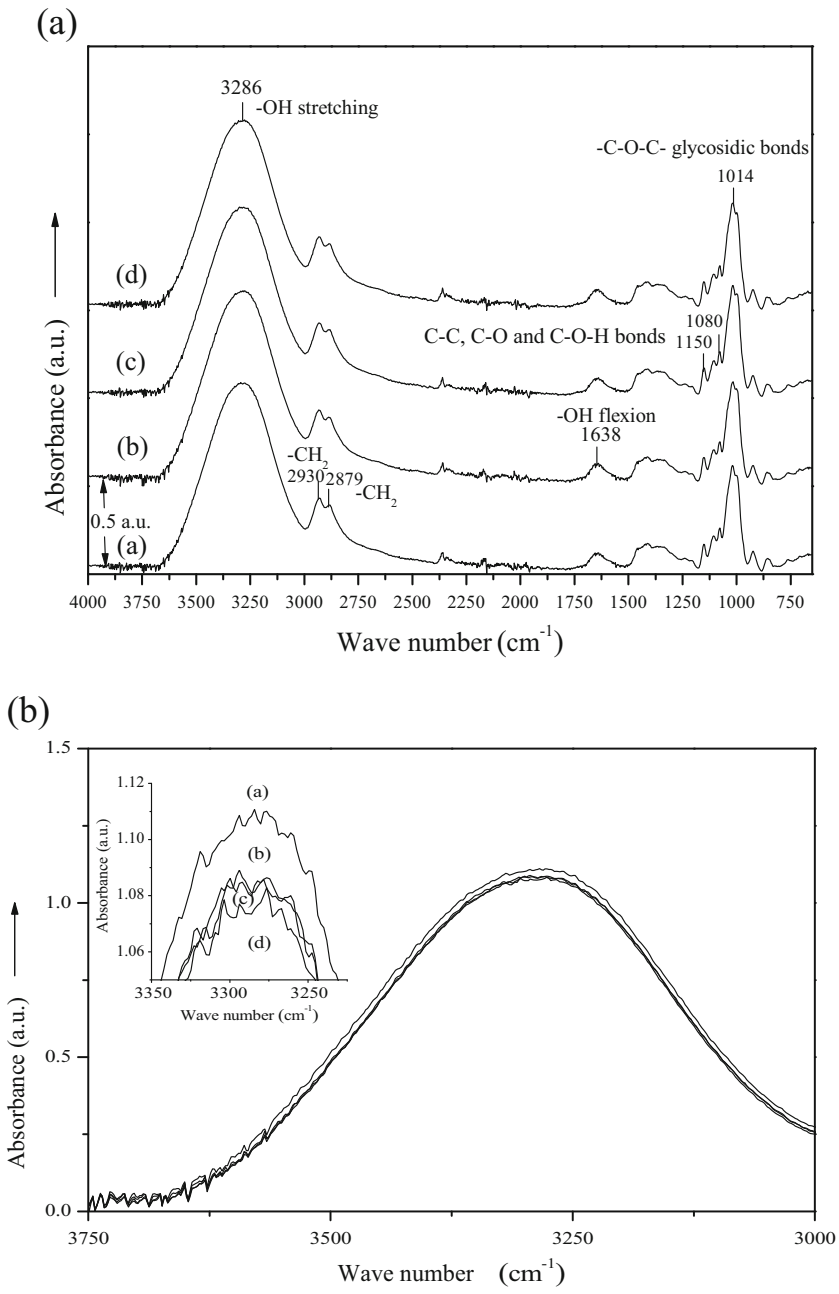

Fig. 5 Panel A- FTIR spectra of the different films studied in all the range absorption: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av). Panel B- FTIR spectra in the range of absorption corresponding to C-O group (OH stretching) of the different films studied: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

Results and Discussion

Although, Av gel contains polyphenolic compounds, which are widely known by their plasticization effect in starch films [35, 38]. However, group of films made from plantain flour with addition of different concentrations of Av gel (0, 2, 4 and 6 %), but without the addition of glycerol did not have the minimum required properties for characterization, as can be observed in Fig. 1.

Average Molecular Weight (M_w) of the Developed Films

Figure 2 shows the average molecular weight of the different films studied. A significant tendency ($p \leq 0.05$) in increasing the molecular weight of the developed films was observed when increasing the Av gel content. According to Mizoguchi and Ueda [52] an increase in average molecular weight of this type of materials is related to the cross-linking of polymers. Therefore, organic acids that are found in Av gel, in particular citric acid found in the Av gel could provoke crosslinking of the starch chains [35], leading to the increase of the average molecular weight of the films. Similar values of average molecular weight have been reported by Yoo and Jane [53] for this type of materials, e.g. the normal and waxy maize starches had average molecular weights of 4.9×10^8 and 8.3×10^8 g/mol, respectively.

Likewise, Yoo and Jane [53] reported that normal and waxy wheat starches had average molecular weights of

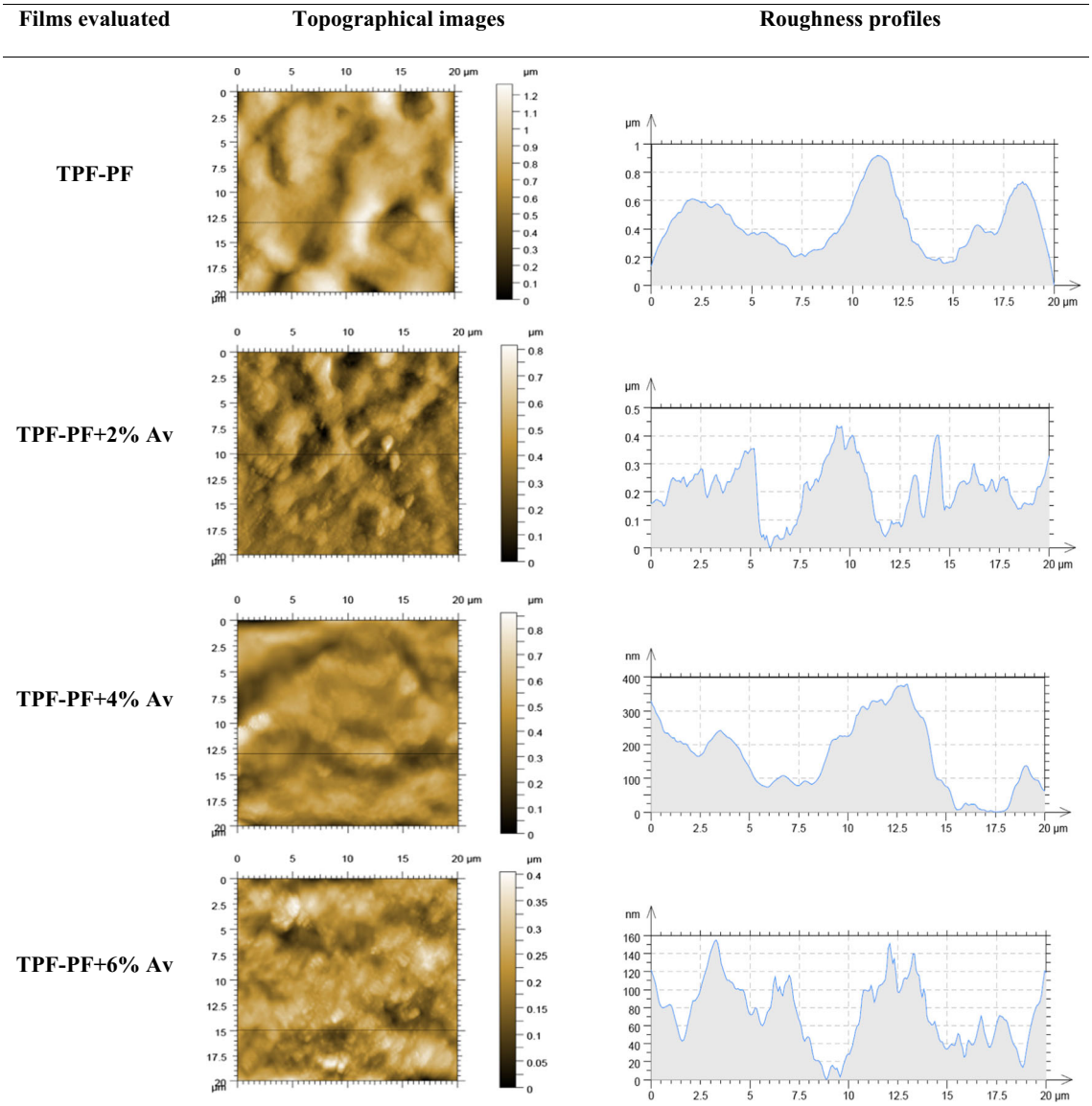

Fig. 6 AFM images of the films: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

3.1×10^8 and 5.2×10^8 g/mol, respectively, i.e. materials rich in amylopectin starch have higher average molecular weights than those materials rich in amylose.

Moisture Content

Figure 3 shows the moisture content of the different films studied. As can be observed, the films derived from plantain flour with higher Av gel content showed a tendency to decrease moisture content. This may be due to strong starch-glycerol interactions, thus reducing water absorption from atmosphere [3, 54]. These results agree with those reported by Cyras et al. [55] on films based acetylated potato starch. A similar behavior has also been recently reported by Gutiérrez et al. [41, 56] for edible films based on cassava and cush-cush yam starch, and plantain flour chemically modified by cross-linking with sodium trimetaphosphate (STMP). Several authors have reported a decrease in moisture content in starch-based films, due to cross-linking with different organic acids such as citric acid, among others [57–59]. This suggests that possibly organic acids of the Av gel caused cross-linking of the starch. A graphical representation is given in Fig. 4.

Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy (ATR/FTIR)

Figure 5a shows FTIR spectra of the different films studied in all the range absorption and the functional groups associated with each band. Specifically in the region between 3350 and 3250 cm^{-1}, it can be observed differences in terms of band intensity (Fig. 5b). A lower absorbance at this peak has been associated to a reduction of free water content [14]. This agrees with the results obtained here. Although, it could also be explained by the loss of the hydroxyl groups of starch during the cross-linking reaction. It is worth remembering that hydroxyl groups of the starch, can act as a nucleophile in the cross-linking reactions, this provokes loss of the hydroxyl functional group, since on this group occurs the addition of the cross-linking agent. Therefore, increasing the Av gel content reduces the hydroxyl group's content of the starch, which is contained in plantain flour-based films.

Atomic Force Microscopy (AFM)

The topography and roughness profiles of the edible films analyzed by AFM are shown in Fig. 6. Films made from plantain flour with higher Av gel content had a trend to decrease surface roughness. This behavior is consistent with luminosity results reported by Gutiérrez and Álvarez [35] for these same systems, i.e. smoother surfaces allows obtaining more transparent materials, therefore, light would not be adsorbed by material surface. Similar results have been reported by Gutiérrez et al. [41] and Reyes [60], for films with a high roughness profile, which were also more opaque, i.e. less transparent. In this context, starch films with higher roughness may indicate the starch retrogradation [41, 61]. The last statement is based on the results obtained of luminosity and roughness of the developed films [41, 61].

Rq and Ra parameters were determined in order to quantify the surface roughness of the films. TPF-PF film had the roughest surface, since Rq was 1.0 μm and Ra was 0.9 μm, while roughness parameters significantly ($p \leq 0.05$) diminished for TPF-PF + 2 % films (Rq 0.5 μm and Ra 0.4 μm) and for TPF-PF + 6 % Av films (Rq 0.16 μm and Ra 0.12 μm). These values confirm qualitative results obtained from SEM images of film surfaces. This reduction in roughness was positively

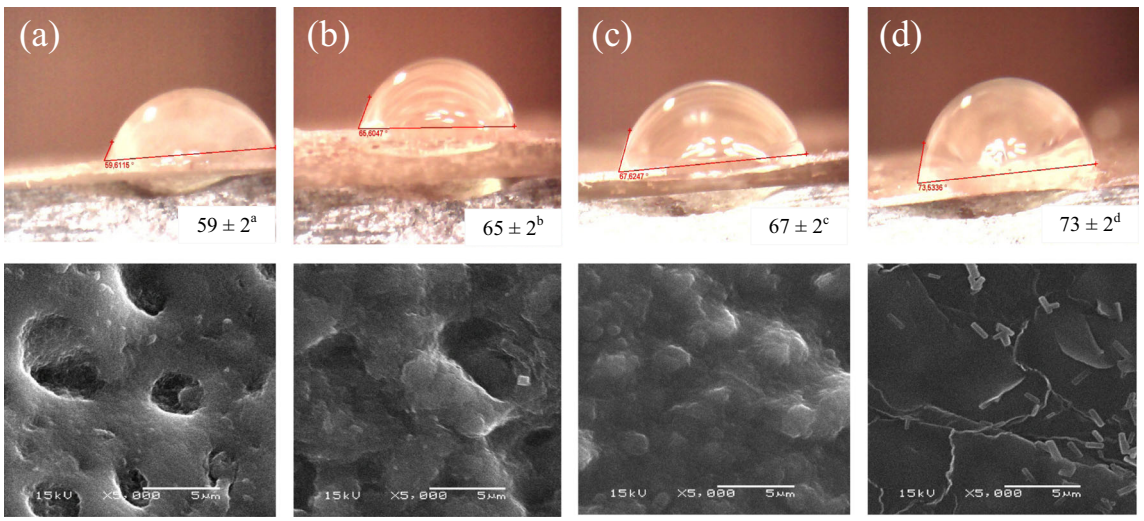

Fig. 7 Contact angles and SEM micrographs of surface of the films based on: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

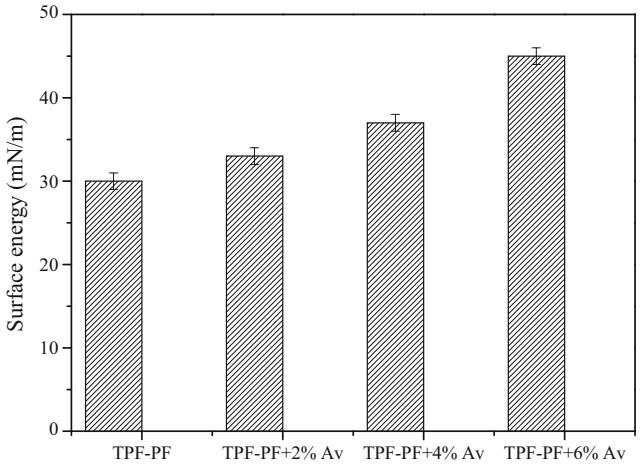

Fig. 8 Surface energy of the films based on: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

correlated with increasing the Av gel content. Therefore, the crosslinking effect caused by the Av gel, prevents the torsion of starch molecules (starch retrogradation), thereby, the surface was smoother, when the concentration of Av gel in plantain flour film systems was increased.

Contact Angle

Figure 7 shows the measurements of the contact angles of the films studied. A statistically significant increase ($p \leq 0.05$) in contact angle values were observed with increasing content of Av gel in the films made from plantain flour. It is well known that water contact angle increases with an increase in surface hydrophobicity [62]. Karbowiak et al. [47] suggested that an increase in water contact angle of biopolymers could be due to strong inter-molecular hydrogen bonding under the film surface. This means that the more polar sites (Lewis sites) would be affected, thus generating a reduction in surface polarity of the biopolymer films. Thus, surface of starch films is dryer because it does not contain enough energy to break up the cohesive force of water [63]. This fits well with the results obtained here, since previously has been discussed that the Av gel produces stronger hydrogen-bond interactions between starch and glycerol, probably due to cross-linking of the starch, caused by organic acids of the Av gel.

Figure 7 shows the contact angles and SEM micrographs of surface of the films studied. The surface of the films with higher Av gel contents were more hydrophobic than those films with lower Av gel contents. This is correlates with the SEM micrographs of surface of the films (Fig. 7). According to Vogler [63] a hydrophobic surface requires Lewis sites in order to increase the humidity. The flour based films derived

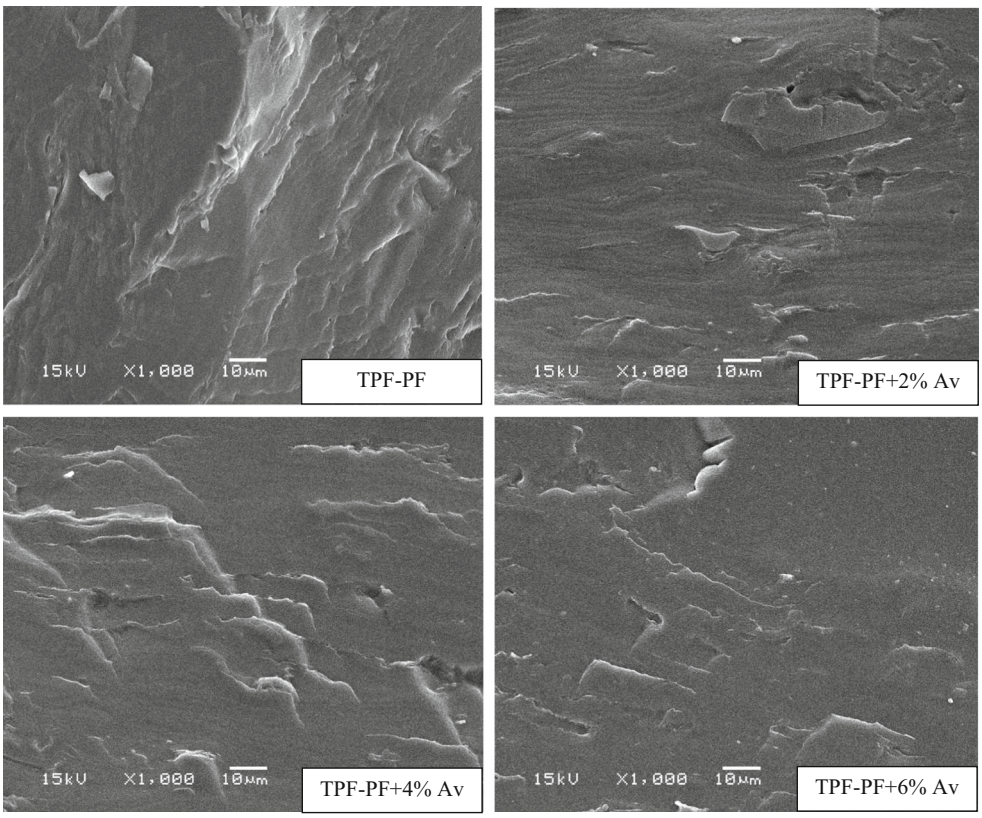

Fig. 9 SEM micrographs of the cryogenic fractured surface of the films: plantain flour (TPF-PF), plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av). At 1kX of magnification

from plantain with higher Av gel content had a closed morphological, which act as a physical barrier to water.

Interestingly, this kind of water structure requires that the hydrogen-bond network of water directly adjacent to a non-polar surface is interrupted, yielding "dangling hydrogen bonds". These dangling hydrogen bonds have been theoretically predicted [64] and spectroscopically resolved from hydrogen bonds in bulk water [65–67]. An increase in contact angle between surface and water would generate dangling hydrogen bonds in the films with higher Av gel contents. The lower contact angle values of the TPF-PF film were due to the weak interactions between starch and glycerol leading to an increase in density of Lewis sites on surface. The Lewis sites on the TPF-PF film would come close to hydrogen bond network in water, thus competing with cohesive forces and leading to collapse of water structure on a hydrophilic surface with a corresponding decrease in contact angle.

Surface roughness can be explained by an increase in contact area and is strongly related to molecular re-organization of the polymeric matrix [47]. In the plantain flour films without Av gel, hydrogen-bonding (starch-starch) would act in support to starch retrogradation. This creates twisting forces in starch macromolecules generating crater-like holes at surface. The holes increase surface roughness of the films (Fig. 6) by creating physical obstacles.

Moreover, recently Gutiérrez and Álvarez [35], determined that higher Av gel content in these same systems decreases in vitro digestibility. Therefore, a closed and compact morphology could act as a physical barrier to enzymatic action.

Additionally, the contact angle method was used to compare the surface energy of the biopolymer composites. The surface energy calculated by means of the Owens and Went equation [48] for flour films are shown in Fig. 8. An increase in the surface energy was observed by increasing the Av gel content. This behavior could be related to strong intermolecular interactions, mainly hydrogen bonding between glycerol and starch below the surface of the material.

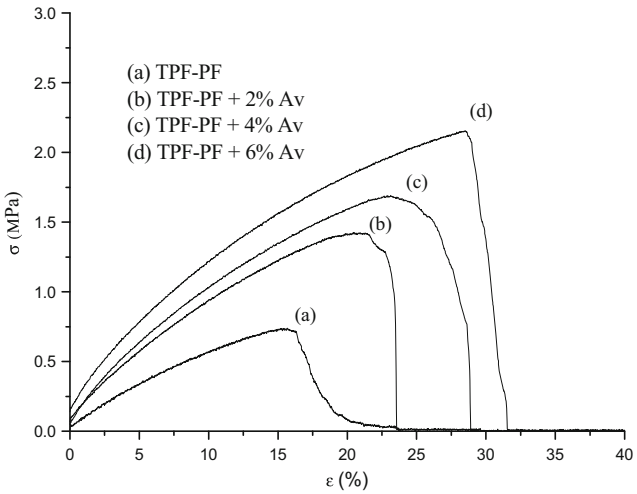

Fig. 10 Stress (σ) - strain (ε) curves of the films: (**a**) plantain flour (TPF-PF), (**b**) plantain flour with incorporation of 2 % *Aloe vera* (TPF-PF + 2 % Av), (**c**) plantain flour with incorporation of 4 % *Aloe vera* (TPF-PF + 4 % Av) and (**d**) plantain flour with incorporation of 6 % *Aloe vera* (TPF-PF + 6 % Av)

Table 2 Parameters of the uniaxial tensile tests: Young's modulus (E), maximum stress (σ_m), strain at break (ε_b) and tenacity (T)

Material	E (KPa)	σ_m (MPa)	ε_b (%)	T ($\times 10^3$) (J/m^3)
TPF-PF	56 ± 6^a	0.81 ± 0.07^a	24 ± 1^a	13.7 ± 0.9^a
TPF-PF + 2 % Av	82 ± 2^b	1.43 ± 0.04^b	25 ± 1^a	24 ± 2^b
TPF-PF + 4 % Av	108 ± 2^c	1.52 ± 0.10^b	30 ± 2^b	30 ± 2^c
TPF-PF + 6 % Av	190 ± 27^d	2.1 ± 0.2^c	31 ± 1^b	45 ± 7^d

Equal letters in the same column indicate no statistically significant difference ($p \leq 0.05$)

Scanning Electron Microscopy (SEM)

Figure 9 shows the SEM images of the cryo-fractured surfaces of the different films. It can be seen that the films obtained are non-porous. All systems presented a compact morphology, but this was far more evident in the films made from plantain flour with higher Av gel content [6, 68]. Similarly this happened on surface exposed to drying the films (Fig. 7). Gutiérrez et al. [6] and Pelissari et al. [39] indicated that a more compact morphology leads to lower water adsorption, since the starch-glycerol and starch-water interactions are less likely to decrease the polar nature of the films [7]. The moisture content results obtained for the plantain flour-based systems (Fig. 3) is agree with the morphological observed by SEM. Similar results and morphologies have been reported for cassava starch-protein films by Saavedra and Algecira [69]. Therefore, surfaces with more compact morphologies could be a physical barrier (steric hindrance) for moisture absorption from the environment.

Uniaxial Tensile Tests

Packaging films are required to have good mechanical properties in order to withstand the stress that occurs during shipping, handling and storage of foods [70]. The stress-strain curves of each film system studied are shown in Fig. 10. A small linear zone followed by a nonlinear zone up to breaking point was observed regardless of the Av gel content incorporated. Table 2 clearly shows a positive effect on all mechanical parameters upon increasing the Av gel content. A higher rigid behavior was found for the films with higher Av gel content (Table 2). Similar results were reported by Pereira et al. [14] for films made from alginate and Av gel. According to Pereira et al. [14] higher Av gel content in alginate-based films, possibly causes cross-linking between the alginate and the Av gel in presence of calcium chloride, thus an increase in maximum

stress was obtained, resulting in more rigid materials. This is in good agreement with the results obtained in this study. Other similar results of maximum stress have recently been published by Gutiérrez et al. [40], in intelligent films made from native and phosphated plantain flour.

As mentioned above, the glycerol interacts more strongly with matrix (plantain flour) upon increasing the Av gel content, probably due to formation of hydrogen bonds between the glycerol and the starch. This glycerol-starch interaction would weaken the intra and intermolecular interactions between starch macromolecules leading to an increase in the movement and rearrangement of their chains and thus in the flexibility of starch films [5, 6, 40]. This results in an increase in the strain at break values of the films with higher Av gel content. Similar results were reported by Gutiérrez et al. [5, 6, 40] for edible films based on corn, cassava and cush-cush yam starch, and plantain flour chemically modified by cross-linking with STMP.

Saavedra and Algecira [69] proposed that cassava starch-protein films have high elasticity values due to its compact morphology. This agrees with results of present work, since the SEM images obtained for films with higher Av gel content (Figs. 7 and 9) correspond to the greater elasticity observed.

A statistically significant increase ($p \leq 0.05$) in Young's modulus and toughness was observed upon increasing the Av gel content. According to Gutiérrez et al. [40], this means that these films could absorb more energy without being transmitted to food, minimizing damage caused by blows in foods during transport and storage. Therefore, films made from plantain flour with higher Av gel content could be biodegradable films to be applied as food containers.

It should be noted that based on the results so far analyzed, the Av gel allows cross-linking of the starch chains, but simultaneously allows a better glycerol-crosslinked starch interaction, thus avoiding intermolecular interactions between the macromolecules of starch without reacting, thereby, the glycerol works as a lubricant for displacement, mobility and rearrangement between starch chains. This fits well with the established in Fig. 4.

Conclusions

The surface of plantain flour-based films with added *Aloe vera* (Av) gel were characterized in order to analyze the effect of incorporating different concentrations of Av gel on the surface and physicochemical properties. In this regard, the effect of crosslinking caused by the organic acids from Av gel on the plantain flour provoked an increase in the hydrophobic nature of the surface and increase the surface energy due to the higher hydrogen bonding interactions. As a result a more compact and smooth morphology was associated with lower values of moisture content in the developed materials. Additionally, the surfaces can be properly associated with the mechanical properties of the films.

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Compliance with Ethical Standards

Conflicts of Interest The authors declare no conflict of interest.

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