

Biobased polymer films from avocado oil extraction residue: Production and characterization

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ABSTRACT: In view of the increasing interest in the use of residues from the food industry as source for packing applications, the by-product of the mechanical extraction of avocado oil is an attractive product as it consists basically a hydrated and defatted pulp, rich in proteins, fibers, and oil. This work aims to produce biobased polymer films from avocado oil extraction residue. Seven film-forming solutions were elaborated from puree and the additives tested were glycerol, cassava starch, and microcrystalline cellulose. The films obtained from the pure residue presented brittle behavior. All films presented low values of water vapor permeability (0.064 to 0.446 g mm⁻²·kPa⁻¹ h⁻¹) and medium water soluble fraction (43.79 to 56.92%). The films with cassava starch and glycerol presented the best results, with mechanical (tensile: 2.70 MPa; elongation: 13.7%) and thermal properties in the range typically found in the literature for biobased films. © 2016 Wiley Periodicals, Inc. *J. Appl. Polym. Sci.* **2016**, *133*, 43957.

KEYWORDS: biopolymers & renewable polymers; films; mechanical properties; thermogravimetric analysis

Received 15 January 2016; accepted 22 May 2016

DOI: 10.1002/app.43957

INTRODUCTION

The use of by-products or residues from the food industry as source for packaging and other new applications has been receiving increasing interest both for its environmental appeal and its potential as a way to increase profit in industrial processes.^{1–5} In this sense, edible and biodegradable films and coatings produced from biopolymers are of special interest as alternatives for plastic packages,^{6–8} considering their fast degradability and the fact that they come from renewable sources.^{8,9}

Biopolymer-based packaging can be produced either from non-processed agricultural products or by-products from food industry, including proteins, lipids, polysaccharides, resins and combinations thereof.¹⁰ Blends of different materials in the film-forming solutions have been studied as a form of improving the characteristics of biopolymer-based packaging.^{11–13} The addition of lipids results, generally, in films with better moisture barrier properties and increased rigidity, also affording opaque characteristics.¹⁴ Protein-based films have hydrophilic character, resulting in poor water barrier, but good oxygen, carbon diox-

ide, and lipid barrier properties in comparison to conventional synthetic polymers. Their main drawback is the fact that they usually exhibit lower mechanical strength.¹⁵ Addition of polysaccharides (such as starch, microcrystalline cellulose, nanoclays, fibers, among others) to the formulation generally leads to improved mechanical properties and moisture content of the film.¹⁴ Extensibility and elasticity of these films may be improved using proper plasticizers.¹⁶ Glycerol is a plasticizer commonly used in this kind of product because, due to its polar nature and low molecular weight, it reduces the interaction between the macromolecules in the film-forming solution.^{17,18}

The avocado pulp is rich in proteins, fibers, and oil, and poor in polysaccharides when compared with other fruits.¹⁹ In the extraction process of avocado oil—a product of high aggregate value—great amount of residual pulp is generated. Extraction by chemical solvents is the most used process for oil production due to high efficiency.^{20,21} Conversely, extraction by centrifugation is an option for small producers, as it does not require large initial investments.^{21,22} Besides, it is attractive from an

environmental point of view and leads to high purity both of the extracted oil and the by-product from the process. This by-product consists basically in a hydrated and defatted pulp, with great potential as raw material for food industry and other applications. Thereby, based on the results obtained by other researchers with other fruit pulps, such as mango,²³ acerola,²⁴ and apple puree,²⁵ this material may represent an interesting alternative for application in film-forming solutions.

The use of avocado pulp or the residue of avocado oil extraction as source for film-forming solution has not yet been explored in the literature. Being so, the aim of this research is, primarily, to use the residue of avocado oil extraction by centrifugation to obtain biobased polymer films. Different film-forming solutions are proposed and characterization of their mechanical, optical, barrier and thermal properties is performed to analyze the potential of application of the produced biobased polymer films.

EXPERIMENTAL

Materials

The avocado oil extraction residue (hydrated and defatted avocado pulp) was provided by a local producer (AvocadoBR Essential Oils, Venâncio Aires, RS, Brazil). The avocado fruit, after pulping, undergoes a thermal treatment (50 °C) before centrifugation for the oil extraction.

The samples were placed in PET bottles under nitrogen atmosphere (100%), frozen by rapid freezing with dry ice, and stored at -18 °C under these conditions until use. Glycerol p.a. (Dinâmica, Brazil) was used as plasticizer. Microcrystalline p.a. (MCC) (density: 0.26–0.32; average molecular weight: 162, degree of polymerization: 350; Synth, Brazil) and cassava starch (CS) (amylose: 18%, amylopectin: 82%; Yoki, Brazil) were used as reinforcing agents.

Nutritional Composition of the Residue

The residue was characterized by physicochemical methods, according to the official AOAC method,²⁶ for the determination of moisture, protein, fat and ash. Moisture was determined by drying the sample until constant weight in an oven (DeLeo, model 48 TLK, Brazil) at 105 °C. Total protein content was calculated by the Kjeldahl method (correction factor: 6.5). Total lipid content was obtained using a Soxhlet extractor (Foss Soxtec, model 2055, Denmark) with petroleum ether as solvent. The ash content was determined by incineration at 550 °C using a muffle for 12 h (Elektro Therm Linn, 312.6 SO LM 1729, Germany). The quantity of soluble dietary fiber and insoluble dietary fiber was determined by enzymatic-gravimetric method, using the total dietary fiber assay kit TDF 100A (Sigma Aldrich). Carbohydrates were obtained by difference, subtracting from 100% the sum of the other components. All the analyses were done in triplicate for each sample and the results expressed in grams per 100 g of dry matter.

Film Preparation

Seven film-forming solutions were elaborated using the residue as main component. The first one, called control (AC), was produced using exclusively avocado oil extraction residue. The other six formulations were prepared with the addition of

Table I. Composition of Film-Forming Solutions Prepared with Avocado Oil Extraction Residue

Film ^a	Residue (g)	Glycerol (g)	Cassava starch (g)	Cellulose (g)
AC	100	—	—	—
AG05	99.5	0.5	—	—
AG10	99	1	—	—
ASG05	98.5	0.5	1	—
ASG10	98	1	1	—
AMG05	99	0.5	—	0.5
AMG10	98.5	1	—	0.5

^a Avocado films (A) without any additive (C → control sample) and with addition of 0.5 and 1 g of glycerol (G05 and G10), cassava starch (S), or microcrystalline cellulose (M).

different concentrations of glycerol (0.5 or 1 g), CS (1 g), and microcrystalline cellulose (0.5 g) to the residue, as described in Table I. For clarity and concision, the abbreviations AG, ASG, and AMG will be used along the text to refer to the corresponding pairs of samples (i.e., AG05 and AG10, ASG05 and ASG10, and AMG05 and AMG10, respectively).

The CS solution was previously prepared from a suspension of 10 g of starch in 200 g of distilled water. The CS solution was heated until 75 °C with constant stirring for approximately 20 min, until the complete gelatinization of the starch. This was made to avoid the degradation of the components of the residue. Then, the CS solution was maintained in a water bath at 40 °C.

All film-forming solutions were stirred at 300 rpm using an overhead stirrer (Edutec, EEQ9034 model, Brazil), for 10 min at 25 °C, to obtain a good mixture of the different components and a homogeneous solution. The films were produced by casting method. The solutions were transferred to acrylic petri dishes in an amount of 0.20 g cm⁻². The samples were dried in a laboratory oven (DeLeo, model B5AFD, Brazil) with forced air circulation at 40 °C for 16 h.

Characterization

All samples were conditioned at 50 ± 5% RH and 25 ± 2 °C for at least 48 h before the analyses, in a desiccator with saturated sodium bromide solution (Dinâmica, Brazil). All the analyses, when applicable, were performed on the side of film exposed to air during the drying process.

Thickness and Moisture Content. Thickness was measured using a digital micrometer (Digimes, IP40 model, Brazil) with a precision of ± 0.001 mm. Thirty six different positions of four samples of each film formulation were measured, and the average value and standard error were calculated.

The moisture content was determined in an analytical balance (Shimadzu, AY 220 model, Japan) with ± 0.001 g precision by measuring the weight loss of the films after dried at 105 °C for 24 h. Seven samples with 2 cm of diameter were used for each formulation.

Grammage. Grammage (g cm⁻²) or mass per unit area was measured according to TAPPI T 410 om-13.²⁷ The samples were

cut in squares (10 cm × 5 cm) and weighted in an analytical balance (Shimadzu, AY 220 model, Japan).

Water Soluble Fraction (WS). The WS was determined gravimetrically using the dried 2 cm diameter samples obtained at the end of the moisture content determination procedure. These samples were put in 30 mL of distilled water and placed in shaking water bath (Novatecnica, NT145, Brazil) slowly (80 rpm) and periodically agitated at 25 °C for 24 h. After that, the samples were dried again in an oven at 105 °C for 24 h and then weighted. WS was calculated as the percentage of dry matter solubilized after the 24 h of immersion.

Morphology. The morphology of the produced films was evaluated by scanning electron microscopy (SEM). The film samples were put on aluminum stubs using a double-sided adhesive tape and coated with a thin layer of gold. The images of the films surface were taken with a SEM (JEOL, JSM 6060 model) with an acceleration potential of 5 kV.

Film Color. Color analysis was performed in a colorimeter (Minolta, CR400, Japan) using D65 at day light. The parameters L^* (luminosity), a^* (green – red), and b^* (blue – yellow) were measured.²⁸ The film samples were analyzed on a white disk ($L_0 = 97.67$; $a_0 = 0.13$, and $b_0 = 1.57$) and the total color difference (ΔE) was calculated using eq. (1). High ΔE values indicate high color intensity.²⁹ Twelve measurements were made on different areas of each sample.

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (1)$$

where $\Delta L^* = L^* - L_0$, $\Delta a^* = a^* - a_0$, $\Delta b^* = b^* - b_0$.

Opacity Index. The light transmittance was measured at 600 nm using a UV spectrophotometer (Shimadzu, CPS 240 A model, Japan) according to Shiku *et al.*³⁰ modified using a quartz test cell. Rectangular film samples (1 mm × 5 mm) were cut and placed on a quartz test cell and then in the spectrophotometer. An empty quartz test cell was used as the reference.

The opacity index (OI) of the films was calculated as proposed by Han and Floros³¹:

$$OI = (-\log T_{600})/x \quad (2)$$

where T_{600} is the transmittance at 600 nm, and x is the film thickness (mm). According to eq. (2), higher values of OI indicate higher opacity and, consequently, lower transparency. No less than twelve measurements were made for each sample.

Water Vapor Permeability. The water vapor permeability (WVP) was determined according to ASTM E96/E96M – 15³² with the modification proposed by Pagno *et al.*²⁹ The samples were positioned in permeation cells (inner diameter = 63 mm, height = 25 mm) filled with granular anhydrous calcium chloride. The cells were subsequently hermetically sealed (0% RH) and stored at 25 °C and in a glass chamber with saturated sodium chloride solution (75% RH). The cells were weighed before and after 24 h of storage in an analytical balance (Shimadzu, AY 220 model, Japan), precision ± 0.001 g. The samples were analyzed in triplicate and WVP was calculated as follows:

$$WVP = (W \times L) / (A \times t \times \Delta p) \quad (3)$$

where W is the weight of water that permeated through the film (g), L is the thickness of the film (mm), A is the permeation area (m²), t is the time of permeation (h), and Δp is the water vapor pressure difference between the two sides of the film (2.376 kPa, 25 °C).

Mechanical Properties. Tensile strength (σ), elongation at break (ϵ), and Young's modulus (E) were determined in a texture analyzer (TA.XT2i, Stable Micro Systems, UK), according to the modified ASTM D882-12.³³ The samples were cut into strips (80 mm × 25 mm) and their thickness was measured at three random positions for each strip. The tests were performed at a speed of 10 mm min⁻¹ with an initial grip separation of 50 mm. Twelve samples of each formulation were tested. σ was obtained by dividing the maximum load by the cross-sectional area of the sample, while E was calculated as the slope of the initial linear portion of the stress versus relative elongation (i.e., the ratio between the absolute elongation at considered point and the initial gage length) curve. Conversely, all values of elongation at break are reported in terms of percent elongation (i.e., relative elongation multiplied by 100).

Thermogravimetric Behavior. Thermogravimetric analysis (TGA) was performed in a Shimadzu Instrument (TGA-50 model, Japan), under argon flow of 50 mL min⁻¹ with a heating rate of 10 °C min⁻¹, from room temperature up to 650 °C, using 8 ± 0.5 mg for each sample. The derivative thermogravimetric (DTG) curve was obtained directly from the TGA curve. All samples were analyzed in duplicate.

Statistical Analysis

The statistical analysis of the obtained data was performed with the Statistica 12.0 software (Statsoft Inc., Tulsa), using analysis of variance and Tukey's test at a significance level of 0.05. To express the results of the Tukey's test, each group of statistically equal means was represented by a different letter.

RESULTS AND DISCUSSION

Composition of the Residue

The analysis of the avocado oil residue composition showed that it has 95.63% moisture content, and other elements on a dry basis were 10.45% protein, 36.53% fat, 10.70% soluble dietary fiber and 15.27% insoluble dietary fiber, 7.60% ash, and 19.44% carbohydrates. The quantities of oil and protein still present in the residue, after de extraction, were high if compared to the composition of the pulp without any treatment (oil: 55.8%; protein: 2.7%; total fibers: 24.5%; carbohydrate: 6.9% for Hass avocado on a dry basis).³⁴ This result is intrinsically related to the species of avocado used and the specific characteristics of the extraction by centrifugation. The avocado fruits used in the oil extraction process that generated the residue under study are, generally, species of small size, like Hass and Fuerte, which have high content of oil. Additionally, extraction by centrifugation has low efficiency, mainly because it does not use solvents, leaving a considerable quantity of oil in the extraction residue.

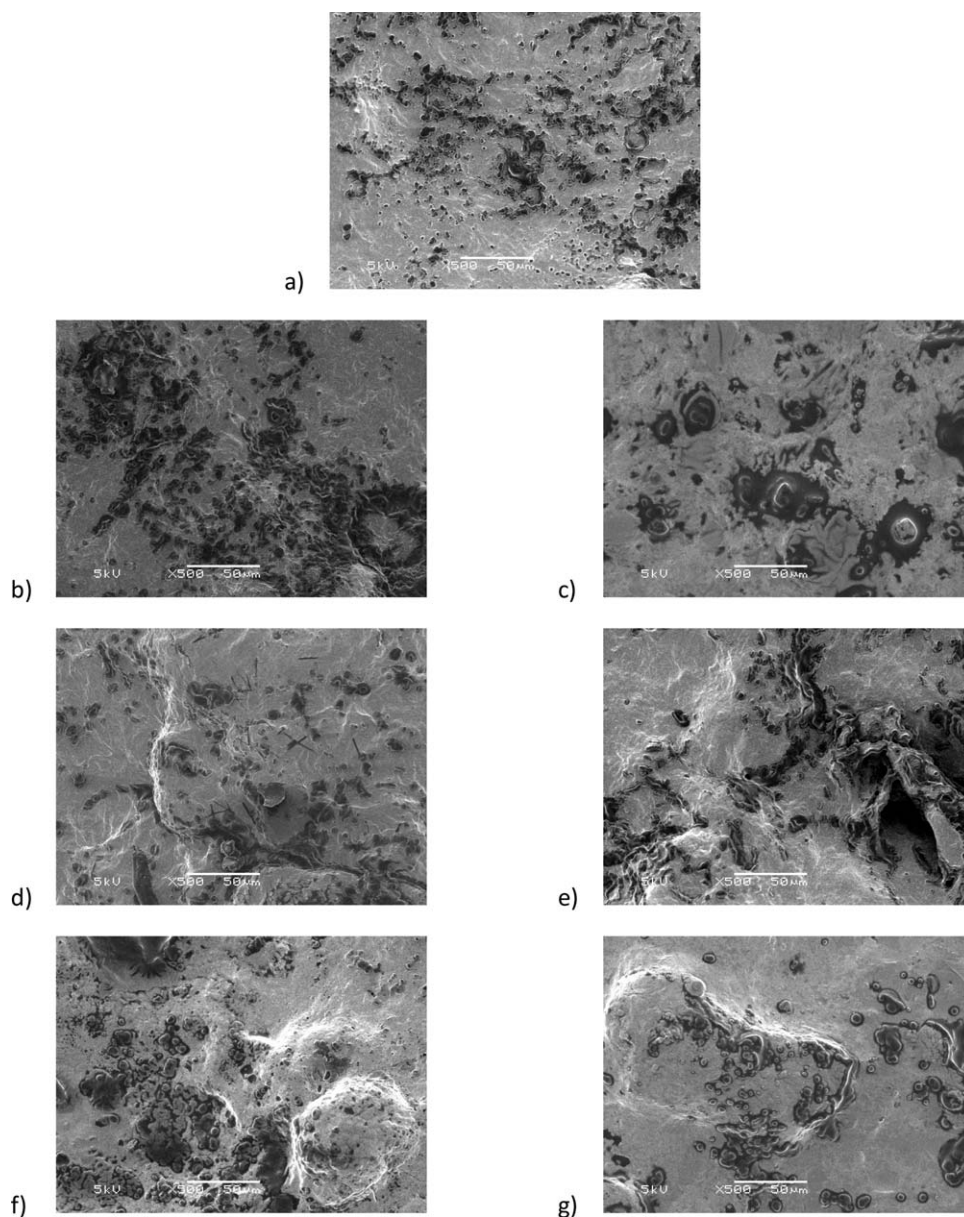


Figure 1. Micrographs of avocado biobased films: (a) AC, (b) AG05, (c) AG10, (d) AMG05, (e) AMG10, (f) ASG05, (g) ASG10.

Film-Forming Features

It was found that the avocado oil extraction residue has adequate properties as film-forming solution material, as it was possible to produce biobased films from the pure residue (formulation AC). This is probably due to its high content of proteins and residual oil. Furthermore, in the centrifugation process the partially hydrated pulp undergoes heat treatment and homogenization at approximately 50 °C. Hence, the proteins are expected to denature in some extent during this process, which facilitates the formation of the film-forming solution.^{35,36} Regarding the oil present in the film-forming solution, it seems to maintain, at least partially, the level of interaction with the other components of the mixture observed originally in the avocado fruit. This statement is based on the fact that regular and stable films were obtained, which would not be possible otherwise. It is important to remark that in most of the works related to the use of fruit

pulp in the production of biobased films, the pulp is used as an additive in gelatin, starch, alginate or pectin-based films.^{12,23–25} Similar results, that is, biobased films using the pulp as main ingredient, were found by Azeredo *et al.*³⁷ for mango films and Martelli *et al.*³⁸ for banana puree films.

Another aspect to be mentioned is that all films produced from pure avocado oil extraction residue exhibited brittle behavior. Therefore, six other formulations were prepared, aiming to obtain more flexible films. Glycerol, a known plasticizer, was tested in two different proportions to increase the flexibility of the formed films. In addition, fixed amounts of CS and MCC were also tested as additives.

Film Characterization

Scanning Electron Microscopy. Figure 1 shows the surface micrographs of the obtained films. All samples presented

Table II. Thickness, Moisture Content, Water Soluble Fraction, and Grammage of the Avocado Films

Film	Thickness (mm)	Moisture content (%)	Water soluble fraction (%)	Grammage (g cm ⁻²)
AC	0.0893 ± 0.0009 ^b	13.7 ± 0.6 ^a	45.8 ± 0.5 ^a	0.0081 ± 0.0002 ^a
AG05	0.0831 ± 0.0012 ^a	24.5 ± 1.2 ^b	46.1 ± 0.3 ^a	0.0089 ± 0.0004 ^{a,b}
AG10	0.0828 ± 0.0021 ^a	35.4 ± 1.4 ^c	46.8 ± 0.8 ^a	0.0093 ± 0.0006 ^{a,b}
ASG05	0.0838 ± 0.0021 ^{a,b}	16.3 ± 0.8 ^a	54.6 ± 0.8 ^b	0.0096 ± 0.0007 ^{a,b,c}
ASG10	0.0814 ± 0.0018 ^a	24.2 ± 1.6 ^b	56.9 ± 0.8 ^b	0.0108 ± 0.0004 ^{b,c}
AMG05	0.1259 ± 0.0025 ^d	22.9 ± 0.8 ^b	43.8 ± 0.8 ^a	0.0100 ± 0.0004 ^{b,c}
AMG10	0.1168 ± 0.0019 ^c	32.6 ± 0.7 ^c	44.9 ± 1.2 ^a	0.0115 ± 0.0006 ^c

The results are represented as the means ± standard error. Different letters in the same columns differ significantly ($P < 0.05$) by Tukey test. Avocado films (A) without any additive (C → control sample) and with addition of 0.5 and 1 g of glycerol (G05 and G10), cassava starch (S), or microcrystalline cellulose (M).

complex morphologies throughout the film surface at the micrometer level, in which two main phases are observed: a dispersed phase composed mainly of oil (dark areas) and a continuous phase composed of the remaining components of the avocado pulp. In the samples in which glycerol was the only additive used (AG05 and AG10), increase of glycerol content led to coalescence and increase of the dispersed oil phase dimensions, probably due to the increase of hydrophilicity promoted by glycerol. In the samples containing cellulose (AMG05 and AMG10) and starch (ASG05 and ASG10) this effect was not noticeable, perhaps due to the interaction between glycerol and those components. In the specific case of sample AMG05, the surface micrograph shows the presence of cellulose fibers on the film surface, which is in agreement with the rough texture empirically observed for these films. The fact that the cellulose fibers are not observed in the micrograph of the sample AMG10 can be attributed to the higher content of glycerol of this sample and the plasticizing effect exerted by glycerol on cellulose fibers.³⁹ The surface of the samples containing starch (ASG05 and ASG10) presents some high relief structures, probably constituted of gelatinized starch.

Thickness, Moisture Content, WS Fraction and Grammage. Table II shows the values of thickness, moisture content and WS fraction for each of the seven biobased film formulations. Thickness varied significantly among the films. By comparing the samples AC to AG (AG05 and AG10) and AMG05 to, AMG10 it is observed that an increase in the glycerol content decreased the film thickness. This result is opposite to those reported in the literature for films produced from binary protein/glycerol formulations⁴⁰ and can be related to the changes promoted by glycerol on the state of dispersion of the oil phase (Figure 1). Additionally, the films containing MCC (AMG05 and AMG10) presented the highest values of thickness, probably due to the more heterogeneous nature of the system as a consequence of the presence of suspended fibrous material.

Regarding moisture content, it can be seen that in all cases the addition of glycerol caused an increase of moisture. This correlation between addition of glycerol and moisture has been observed for many other biobased film formulations.^{40–43} It is attributed to the hygroscopic nature of glycerol, which is a consequence of the hydroxyl groups present in its structure. It is

also remarkable that in the two formulations containing CS (ASG05 and ASG10) the increase of moisture content with the addition of glycerol was lower than that observed when adding either only glycerol (AG05 and AG10) or MCC and glycerol (AMG05 and AMG10). This suggests that in the case of these formulations there must be some degree of interaction between glycerol and starch particles in such a way that the amount of glycerol available for moisture absorption is lower. Conversely, the moisture content of the AMG and AG formulations are statically equal when compared at the same content of glycerol, indicating that MCC has no significant effect on the moisture of the films.

Concerning the results for WS fraction it was observed that the films can be statistically divided in two groups, with the ASG films presenting higher solubility than all other formulations. This is a direct consequence of the fact that in the film-forming solution containing starch there is a decrease in the oil content in relation to the other formulations (Table I). So, in the ASG films the ratio between hydrophilic and hydrophobic species increases, promoting higher WS fraction. Similar behavior has been reported by Pérez-Mateos *et al.*⁴⁴ for cod gelatin films with sunflower oil. It is also interesting to observe that this effect is not observed in terms of moisture content of the samples, as there is not a correlation between moisture content and WS fraction values reported in Table II. This is possibly a consequence of the interaction between glycerol and starch particles discussed in the previous paragraph and of the conditions used for the determination of WS fraction. While the moisture content of a sample is defined under limited availability of water, allowing direct interaction between glycerol and starch, the WS fraction determination involves the contact of small piece of sample with a great volume of water, in such a way that each soluble species is directly in contact with water.

The grammage of the films did not vary significantly as a function of the amount of glycerol added. However, differences were observed with the addition of CS and microcrystalline cellulose. These compounds led to increase on grammage, probably due to their high molecular weight and hydrophilic characteristics.

Color and OI. The Table III exhibits the optical properties found for the films produced. There were significant differences between samples especially regarding ASG films. Films with

Table III. Optical Properties of Avocado Extraction Residue Films

Film	L^*	a^*	b^*	ΔE	OI
AC	53.3 ± 0.6^a	5.2 ± 0.1^b	36.8 ± 0.5^a	57.0 ± 0.2^b	15.0 ± 0.2^e
AG05	52.2 ± 0.5^a	6.3 ± 0.2^c	$37.6 \pm 0.5^{a,b}$	$58.4 \pm 0.2^{b,c}$	15.2 ± 0.4^e
AG10	54.2 ± 0.6^a	6.0 ± 0.2^c	$39.2 \pm 0.4^{b,c}$	$57.9 \pm 0.3^{b,c}$	14.5 ± 0.5^d
ASG05	59.3 ± 0.6^b	3.5 ± 0.2^a	$39.0 \pm 0.2^{b,c}$	53.8 ± 0.5^a	12.0 ± 0.4^c
ASG10	60.5 ± 0.9^b	3.4 ± 0.1^a	$39.4 \pm 0.3^{b,c}$	53.1 ± 0.6^a	11.9 ± 0.2^c
AMG05	53.2 ± 1.6^a	$5.7 \pm 0.4^{b,c}$	$37.9 \pm 0.9^{a,b,c}$	$57.9 \pm 0.8^{b,c}$	10.5 ± 0.2^a
AMG10	54.3 ± 0.7^a	6.1 ± 0.2^c	40.5 ± 0.4^c	58.7 ± 0.3^c	10.9 ± 0.3^b

The results are represented as the means \pm standard error. Different letters in the same columns differ significantly ($P < 0.05$) by Tukey test. Avocado films (A) without any additive (C \rightarrow control sample) and with addition of 0.5 and 1 g of glycerol (G05 and G10), cassava starch (S), or microcrystalline cellulose (M).

starch often have high luminosity. In the process of gelatinization, water is retained by the starch granules causing their swelling.⁴⁵ When gelatinized starch is added, a dilution of the original solution occurs, thereby leading to a lighter color spectrum, with decrease in the color intensity of the samples. This can be observed through the increase of L^* and the decrease of a^* values of ASG films or their lower values of ΔE , when we compared to the other formulations.

The analysis of the OI data in Table III also enlightens the influence of the composition and related microstructural features on the final characteristics of the produced films. The AG films presented values of OI more similar to that of the control sample than the ASG and AMG films. Both ASG and AMG films presented a significant increase of transparency with relation to the control sample. In the case of the ASG films, this increase can be explained by considering the same aspects discussed in the previous paragraph concerning the effect of the addition of the gelatinized starch and also by the lower effective content of avocado residue of the ASG films. In turn, the interpretation of the low values of OI presented by the AMG films is not so straightforward. However, some insight can be gained by looking at these results in the light of those of thickness and microscopy. The fact that the AMG films presented the highest values of thickness and that irregularities were observed in some of their micrographs, as in Figure 1(e), suggest the presence of microvoids in these films, which would explain their higher

transparency. Additionally, it is observed that the samples with the highest presence of dark areas in the micrographies (AC and AG05) [Figure 1(a,b)] also present the highest OI values. This fact indicates that the opacity is also affected by the dispersed oil phase, which is in agreement with the results reported by Ma *et al.*¹³ for gelatin-based films.

Still regarding the OI data, it is worthy to mention the differences observed among formulations in terms of glycerol content influence. The AG films presented significant decrease of opacity with the increase of glycerol content, which is in agreement with results presented by Dick *et al.*⁴¹ for chia mucilage films and is due to the transparent nature of glycerol. For the ASG films, no significant change of the OI with the content of glycerol was observed, probably due to the interaction between glycerol and starch gelatinized particles, which reduces the effective amount of glycerol available for direct interaction with the avocado residue. Finally, the AMG films presented an inverted behavior, with increase of opacity with the increase of the glycerol content. This can be attributed to the plasticizing effect of glycerol on cellulose fibers and a consequent densification of the film.

Water Vapor Permeability. The values of WVP of the produced films were in the range from 0.064 to 0.446 g mm m⁻² kPa⁻¹ h⁻¹ (Table IV). These values are far below those reported in the literature for other biobased films of

Table IV. Mechanical and Barrier Properties of Avocado Oil Extraction Residue Film

Film	WVP (g mm m ⁻² kPa ⁻¹ h ⁻¹)	σ (MPa)	ϵ (%)	E (MPa)
AC	0.064 ± 0.003^a	4.20 ± 0.24^f	1.4 ± 0.2^a	418.2 ± 13.3^f
AG05	0.262 ± 0.008^c	0.90 ± 0.04^d	$9.4 \pm 0.5^{c,d}$	37.8 ± 1.8^d
AG10	0.383 ± 0.006^e	0.56 ± 0.02^b	13.9 ± 0.4^e	13.2 ± 0.5^a
ASG05	0.155 ± 0.004^b	2.70 ± 0.04^e	13.8 ± 0.4^e	98.7 ± 2.2^e
ASG10	0.243 ± 0.007^c	0.34 ± 0.01^a	10.4 ± 0.3^d	24.1 ± 1.2^b
AMG05	0.322 ± 0.013^d	0.76 ± 0.03^c	7.2 ± 0.5^b	35.9 ± 1.4^d
AMG10	0.446 ± 0.016^f	0.32 ± 0.01^a	$8.7 \pm 0.3^{b,c}$	13.8 ± 0.5^a

The results are represented as the means \pm standard error. Different letters in the same columns differ significantly ($P < 0.05$) by Tukey test. Avocado films (A) without any additive (C \rightarrow control sample) and with addition of 0.5 and 1 g of glycerol (G05 and G10), cassava starch (S), or microcrystalline cellulose (M).

similar composition, such as values between 2.7 and 1.38 $\text{g mm m}^{-2} \text{kPa}^{-1} \text{h}^{-1}$ for gelatin-glycerol-olive oil films (62.5:25:12.5),¹³ and between 4.37 and 5.25 ($\text{g mm m}^{-2} \text{kPa}^{-1} \text{h}^{-1}$) for alginate-apple puree-essential oils films.²⁵ Conversely, the WVP values of the avocado films were much higher than those of synthetic films as LDPE (0.0032 $\text{g mm m}^{-2} \text{kPa}^{-1} \text{h}^{-1}$, 0/90% RH),^{46,47} HDPE (0.0008 $\text{g mm m}^{-2} \text{kPa}^{-1} \text{h}^{-1}$, 0/90% RH),^{46,47} and PVCD (0.0008 $\text{g mm m}^{-2} \text{kPa}^{-1} \text{h}^{-1}$, 0/90% RH),^{46,47} being more similar to that of cellophane (0.293 $\text{g mm m}^{-2} \text{kPa}^{-1} \text{h}^{-1}$ 0/90% RH).^{46,47}

AC film presented the lowest WVP, which is in agreement with its higher oil content and finer dispersion of the oil phase [Figure 1(a)] taking into consideration that WVP depends on the quantity of water in the film⁸ and on the particle size distribution of the lipid phase.⁴⁸ This later effect is related to the fact that, under a fixed volume fraction, smaller lipid particles lead to higher contact surface between the lipid phase and the diffusing water vapor, increasing the resistance to this transport.

The addition of plasticizer increased the WVP of the films for all formulations. This probably occurs because glycerol, that has a hydrophilic and hygroscopic nature, attracts water molecules forming a hydrodynamic plasticizer-water complex. According to Wang *et al.*,¹² such effect may increase the distance between polymer molecules, promoting the creation of free volume on the film matrix and increasing the WVP. This interpretation also agrees with the aforementioned decrease of OI from sample AG05 to AG10.

The increase of WVP in relation to the control sample was smaller for the ASG films. Similar behavior has been reported by Zhong and Xia⁴⁹ and Mali *et al.*⁵⁰ This can be understood as a result of the more compact structure and stronger intermolecular interaction in the gelatinized starch particles compared to the other two systems. In AG and AMG films, as discussed before, the plasticizing effect of glycerol was more intense, which would lead to higher free volume and facilitate the passage of water vapor. It is worth mentioning that these results are in qualitative agreement with those found for moisture content (Table II). Nevertheless, the fact that the AMG samples presented the highest values of WVP can be taken as support for the discussion presented in the previous section about the possibility of occurrence of microvoids in the AMG samples.

Mechanical Properties. Tensile strength (σ), elongation at break (ϵ), and Young's Modulus (E) of the films are shown in Table IV. AC film presented a brittle response, with low values of σ and ϵ but a high E , probably on account of oil presence.⁴⁸ The addition of glycerol decreased tensile strength and elastic modulus, confirming the plasticizing effect of this component. AMG and AG films presented similar values of tensile strength and elastic modulus, indicating that the addition of MCC was not effective as reinforcement. This latter result is in agreement with the morphological aspects discussed in the previous section.

The ASG films presented significant differences in relation to the AG and AMG formulations. The addition of starch together with glycerol led to reduction of brittleness in relation to the control sample, but with a much lower reduction of tensile strength and modulus when compared to the AG and AMG

films. Besides, for the ASG samples the elongation at break decreased with the increase in glycerol content, contrarily to what occurred for the AG and AMG formulations and some reports in the literature.^{50,51} Additionally the reduction of tensile strength with the increase of the glycerol content was much more intense (nearly 9 times) for the ASG films. This could be explained in terms of a non-homogeneous plasticizing effect for the highest content of glycerol, with the excess of glycerol causing the matrix to deform much more easily than the gelatinized starch particles. Ramos *et al.*⁴² found similar response for glycerol-whey protein films and attributed this behavior to the saturation of the film matrix by glycerol.

Finally, it is worthy to mention that the values of mechanical properties reported in Table IV are in the same order of magnitude as those reported in the literature for other types of biobased films, such as those obtained from açai puree-pectin-thyme oil (σ : 0.59 to 2.74 MPa, ϵ : 27.58 to 114%, E : 3.17 to 13.36 MPa),⁵² carrot puree (σ : 2.46 to 18.08 MPa, ϵ : 4.36 to 21.94%),¹² alginate-apple puree-essential (σ :2.47 to 2.90 MPa, ϵ : 51.06 to 58.33%, E : 5.75 to 7.07 MPa),²⁵ alginate-acerola-cellulose whiskers (σ : 3.16 to 6.10 MPa, ϵ : 16.60 to 28.26%, E : 15.35 to 50.16 MPa),²⁴ and CS-yerba mate extract-mango pulp (σ : 1.36 to 4.03 MPa, ϵ : 55.15 to 69.36%).²³ In comparison to synthetic plastics such as HDPE (σ : 17.3 to 34.6 MPa, ϵ : 150 to 300%),^{46,47,53} LDPE (σ : 8.27 to 31.71 MPa, ϵ : 68.7 to 500%),^{46,47,53} and PVC (σ : 48.4 to 138 MPa, ϵ : 20 to 40%)⁴⁷ the avocado films presented lower values of tensile strength and elongation at break.

Thermogravimetric Behavior. Figure 2 shows the TGA and DTG curves of the produced films. It can be seen in all samples the presence of three peaks in the temperature range of 50–120 °C. These weight losses are related in the literature as associated with the loss of free and bound water present in the film.^{52,54,55} This hypothesis is confirmed by the area increase of these peaks with increasing glycerol content, which results in increased moisture content of the sample (Table II).

The mass loss stage between 160 and 230 °C shows the contribution of the glycerol degradation,⁵⁶ which occurs in this temperature range. The increase of this peak with the increase of glycerol content supports this statement. However, this peak must also contain the contribution of simple polysaccharides degradation and lower molar weight peptides decomposition that occur in this region,⁵⁵ as it can be observed in the AC TGA curve.

The peak of degradation that occurs around 300 °C appears to be related only to components of the oil extraction residue, because its area does not present relevant variation when comparing all formulations. Considering literature data, this peak may be associated with larger-size or higher interacted protein fractions degradation.^{57,58}

The last mass loss stage occurs between 320 and 400 °C. It is probably related to degradation of more complex and high weight components, like fibers and complex polysaccharides,⁵⁵ and evaporation of oleic (360 °C) and palmitic (352 °C) acids, which are the main fatty acids present in avocado oil. In addition, in this temperature range it also occurs the degradation of CS and cellulose.^{59,60} For the AG films [Figure 2(a)], the area of

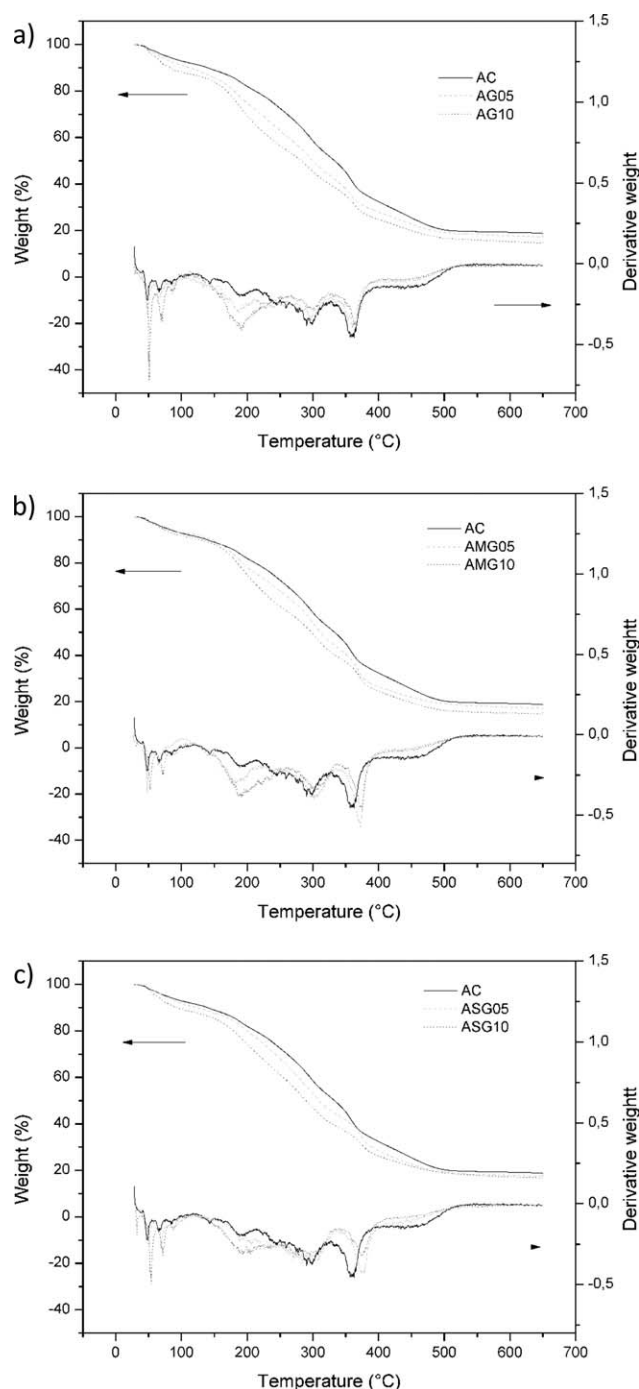


Figure 2. TGA and DTG curves of samples: (a) AG, (b) AMG, (c) ASG. Results for sample AC are also presented, in all cases, as basis of comparison.

this peak decreased with the increase in glycerol content, as a result of the dilution of the components present in the pulp. For AMG and ASG films, this peak was shifted by about 15 °C, indicating that the interaction between the components of oil extraction residue and MCC or starch are strong enough to promote a light increase of the thermal stability of the formulation. This interaction is probably due to the residual oil present in the raw material, as the literature reports increased stability in CS or gellan films additivated with oil.^{57,58,61}

CONCLUSIONS

The residue of the avocado oil extraction has properties of a film-forming solution and it can be used to produce biobased plastic films. Although it was possible to obtain films from the pure residue, these films were brittle in nature. Glycerol was an efficient plasticizer for the films produced from the residue of avocado oil extraction, increasing significantly the elongation at break of the formulations. Despite that some of the results indicated some level of interaction between MCC and the other components of the pure residue; this additive was not effective in improving the mechanical properties of the formulations, presenting the poorest mechanical behavior among the additivated films. CS, conversely, showed higher interaction with the other components of the formulation and improved the mechanical response of the produced films, resulting in films with mechanical properties in the range typically found for biobased films presented in the literature. It is also relevant the fact that for ASG films elongation at break decreased with the increase of the glycerol content, indicating that saturation of the film matrix by glycerol can occur in the presence of CS. The avocado films show good WVP properties when compared to other biobased films of similar composition and cellophane, presenting WVP values much higher than those of some widely used synthetic plastics. Therefore, the residue of the avocado oil extraction can be pointed as an alternative raw material to produce biobased packaging films for food industry.

ACKNOWLEDGMENTS

The authors are grateful to Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES, Brazil) for the financial support and to the Electronic Microscope Center (CME) of Federal University of Rio Grande do Sul (UFRGS) for technical assistance. We also acknowledge AvocadoBR Essential Oils for the donation of the avocado pulp used in this research.

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