

# The effect of andiroba oil and chitosan concentration on the physical properties of chitosan emulsion film

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## Abstract

Chitosan film is used as a dressing to heal burns. The physical and biological properties of the film can be modified by the addition of phytotherapeutic compounds. This work used the casting -solvent evaporation technique to prepare chitosan film containing andiroba oil (*Carapa guianensis*) which has anti-inflammatory, antibiotic, and healing properties. The objective of this study was to determine the effect of the concentrations of chitosan and andiroba oil on the physical properties of chitosan films. The emulsion films were evaluated concerning the mechanical properties and fluid handling capacity. Additionally, scanning electron microscopy and thermal analysis were performed. The results showed that the barrier and mechanical properties were affected by the addition of andiroba oil, and these may be modulated as a function of the concentration of oil added to the film. The thermal analysis showed no evidence of chemical interactions between the oil and chitosan.

**Keywords:** biopolymers, dressings, *Carapa guianensis*.

## 1. Introduction

Andiroba (*Carapa guianensis*) is a tree of the Meliaceae family found in Central and South America, especially in the Amazon basin region. Many studies have reported the pharmacological properties of products obtained from the andiroba flower<sup>[1,2]</sup>, from the ethanolic extract of the andiroba leaf<sup>[3,4]</sup> and especially products derived from andiroba seed oil<sup>[5-7]</sup>. Andiroba oil is widely used in popular medicine in the Amazon basin region.

According to Cabral et al.<sup>[8]</sup>, andiroba oil is composed mostly of triacylglycerols, with high levels of unsaturated and saturated fatty acids such as oleic (51.81%), palmitic (25.76%), stearic (9.08%), and linoleic (8.3%). The medicinal properties of andiroba oil have been attributed to the presence of limonoids, which are tetranortriterpenoids<sup>[6]</sup>. Andiroba oil also contains triterpenes, tetraterpenes, alkaloids, and glycerides<sup>[9]</sup>.

Andiroba seed oil is currently considered to be acaricides<sup>[10]</sup>, larvicidal against *Aedes aegypti*<sup>[11]</sup>, as well as being an antiplasmodial<sup>[5]</sup>, anti-inflammatory<sup>[12]</sup>, anti-allergic<sup>[7,13,14]</sup>, and also suitable for wound healing<sup>[3,4]</sup>.

The ethanolic extract of *Carapa guianensis* leaves was evaluated for antibacterial and wound healing activity, using excision, incision, and dead space wound models in rats. The results showed an increased rate of wound contraction and hydroxyproline content (a biochemical marker for tissue collagen), which indicates the potential application of *Carapa guianensis* in wound healing<sup>[4]</sup>.

Many wound dressings have been developed for the treatment of severe burn wounds or ulcers. Damaged tissue requires biocompatible materials like chitosan that has a high film-forming capacity. The most cited advantages of chitosan are its physico-chemical and biological

properties. Chitosan promotes activation and proliferation of inflammatory cells in granular tissues<sup>[15]</sup>, stimulates cell proliferation and histoarchitectural reorganization of the tissue<sup>[16]</sup>, and affects the functioning of macrophages, thus accelerating the healing process<sup>[17]</sup>. The use of chitosan resulted in a substantial decrease in healing time and minimal scarring in several animals<sup>[18]</sup>. These and other properties can be potentiated with the incorporation of andiroba oil. As previously described, andiroba oil has many interesting properties for use in dressings.

The preparation of emulsified films presents some challenges, for example, maintaining the stability of the emulsion during the process of drying the film. The stability of chitosan films can be achieved by adding surfactants but also by emulsification with rotor-stator homogenizer (> 20,000 rpm)<sup>[19-21]</sup>, or even less vigorously (13,500 rpm)<sup>[22]</sup>. The small concentration of chitosan in oil emulsions also helps to maintain the stability of the emulsion. Usually, this concentration ranges from 0.1 to 1%<sup>[19-21]</sup>, but it can reach values up to 3%<sup>[22]</sup>. Therefore, in this work, we decided to work with concentrations between 0.1 and 1% and agitation (24,000 rpm).

This work aims to study the effect that the concentrations of andiroba oil and chitosan have on the physical and chemical properties of chitosan film used for wound dressings.

## 2. Materials and Methods

### 2.1 Materials

Commercial chitosan (deacetylation of approximately 82% and molar mass of approximately  $1.47 \times 10^5$  g/mol) was supplied by Polymar (Fortaleza, Brazil) without prior

purification. Acetic acid (Synth, Brazil) was used as an acidic medium. Andiroba oil was provided by MPR Indústria e Comércio de Óleos Vegetais Ltda (Brazil).

## 2.2 Chitosan suspension

Chitosan (1.0% or 2.0%, w/w) was dissolved in an aqueous acetic acid solution. The stoichiometric amount of acetic acid was calculated to achieve the protonation of all the  $\text{NH}_2$  sites and taking into account the sample weight and the degree of acetylation. Using this base amount plus an extra 50% gave the total stoichiometric amount to be used. The suspension was homogenized for 2 h prior to the preparation of the chitosan film, in order to complete chitosan solubilization.

## 2.3 Preparation of chitosan film

The films were prepared by the casting technique. The chitosan suspension and the andiroba oil was emulsified beforehand (24000 rpm for 10 min). We tested three concentrations of andiroba oil (g of andiroba oil/100 g of solution, i.e. % w/w): 0.1%, 0.5%, and 1.0% w/w. The chitosan emulsion was poured into polyethylene Petri dishes. The films were dried in a forced air oven at 40 °C for 24 h. The mass of the suspension applied to the Petri dishes was kept constant (0.21 g/cm<sup>2</sup>). The films then underwent various analyses.

## 2.4 Scanning Electron Microscopy (SEM)

SEM analysis was performed on fractured cross-sections and the surfaces of gold-sputtered films using an LEO 440i scanning electron microscope (LEO Electron Microscopy Ltda.) with 10 kV and 100 pCa.

## 2.5 Fluid Handling Capacity (FHC)

The fluid handling capacity (FHC) of the film is defined as the sum of the Absorbency (ABS) and Moisture Vapor Transmission Rate (MVTR). The FHC was examined according to the BS EN 13726-1 method<sup>[23]</sup> for hydrocolloids and dressings. In this test, samples of each film (or dressing) were applied to the modified Paddington cups (Figure 1), to which were added 20 mL of simulated exudate fluid (SEF).

The cups were weighed using a calibrated analytical balance, inverted so that the dressing came into contact with

the SEF – see Figure 1c – and the solution was placed in a temperature and humidity controlled incubator to maintain an environment of 37 °C ± 2 and a relative humidity below 20% for a period of 24 h. At the end of the test the cups were removed from the incubator and were allowed to equilibrate at room temperature for a period of 30 min prior to reweighing on the analytical balance. The FHC, ABS, and MVTR were calculated by the following equations:

$$MVTR = \frac{x - y}{\text{time} \times \text{surface}} \quad (1)$$

$$ABS = \frac{b - a}{\text{time} \times \text{surface}} \quad (2)$$

$$FHC = MVTR + ABS \quad (3)$$

where  $x$  is the complete system weight (film + SEF solution + cup) at the beginning of the test;  $y$  is the complete system weight (film + SEF solution + cup) after 24 h;  $b$  is the film weight at the beginning of the test; and  $a$  is the film weight after 24 h. Five repetitions were done per experiment.

## 2.6 Mechanical properties

Tensile testing was done in accordance with the ASTM D882 method<sup>[24]</sup>. Films were cut into 10.00 cm × 2.54 cm strips. The tensile strength, elongation at breaking point, and Young's modulus were measured using TexturePro CT V1.2 (Brookfield, CT3 50K Texturometer). The crosshead speed was set at 1 mm.s<sup>-1</sup>. Samples were pre-conditioned in a desiccator at 75% relative humidity for 48 hours. There were at least 10 repetitions per experiment.

## 2.7 Thermal analysis

Differential scanning calorimetry (DSC) and thermogravimetric analysis (TGA) studies were performed on chitosan film, pure chitosan, and andiroba oil. TGA was done with a TGA-60 (Shimadzu) analyzer. All analyses were performed with 5-10 mg samples in platinum pans in a dynamic nitrogen atmosphere (100 mL.min<sup>-1</sup>), between 30 °C and 700 °C. The experiments were done at a scanning rate of 10 °C.min<sup>-1</sup>. DSC analysis was performed with a DSC-60 (Shimadzu) analyzer. Samples (approx. 5-10 mg) were scanned in a sealed aluminum pan and heated to a

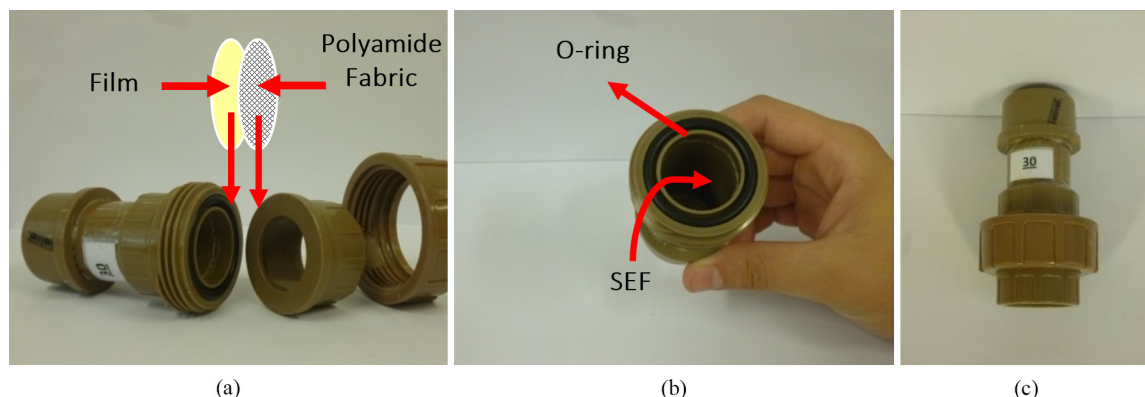


Figure 1. Modified Paddington cups used for the determination of FHC.

temperature of 450 °C at a rate of 10 °C.min<sup>-1</sup>, in a nitrogen atmosphere, and with a flow rate of 50 mL.min<sup>-1</sup>.

## 2.8 Statistical analysis

All the characterizations were done in replicate. The Tukey's test was done for comparison of means, using BioEstat 5.3<sup>[25]</sup>.

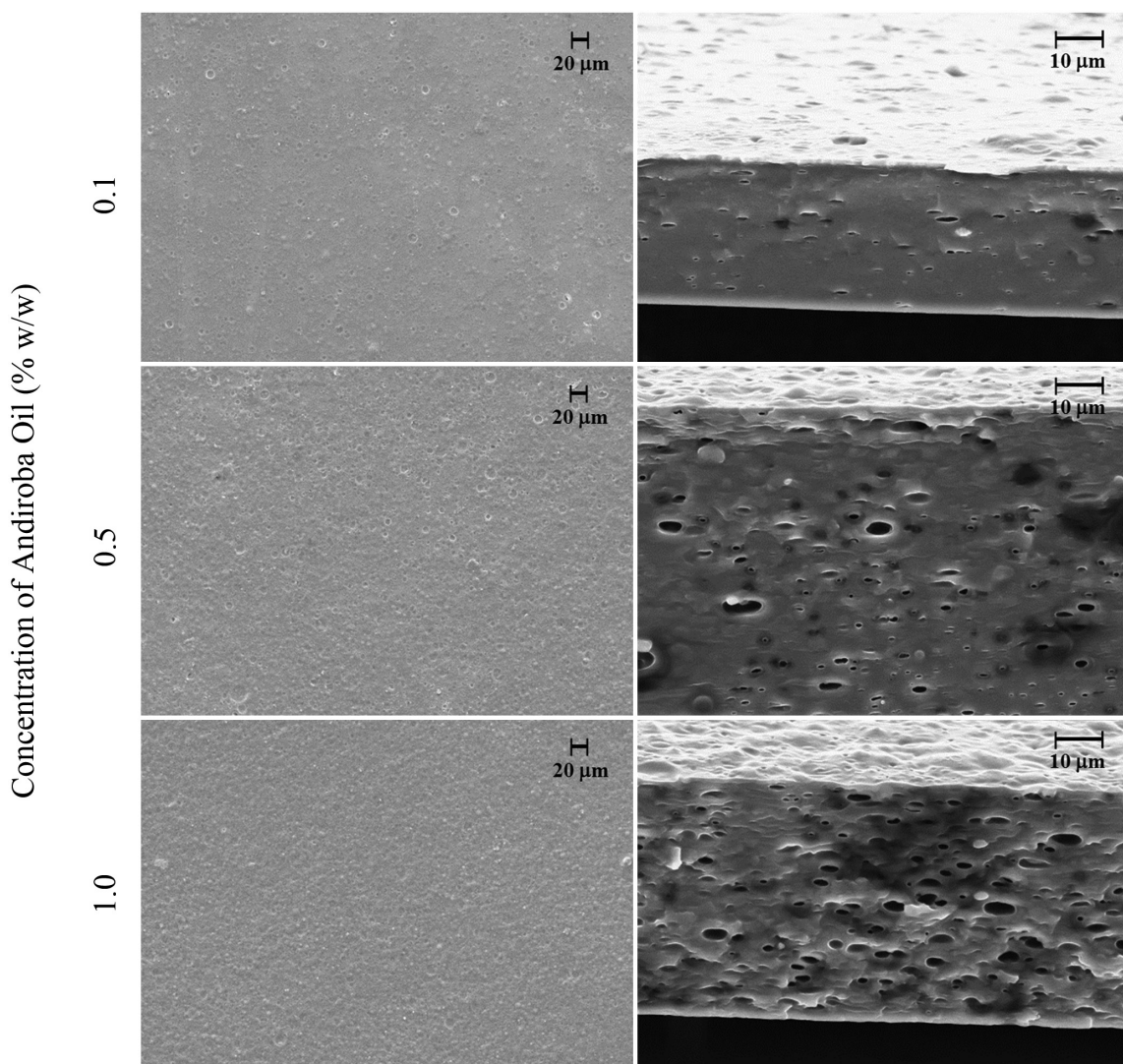
## 3. Results and Discussion

### 3.1 Scanning electron microscopy

No macroscopic phase separation was observed after drying for any of the samples prepared from chitosan/andiroba oil, which indicates that oil droplets were stabilized in the chitosan suspension. SEM was used to evaluate the morphology and distribution of oil droplets in the films. This analysis allows us a better understanding of mechanical and barrier properties<sup>[26]</sup>. Figure 2 shows SEM micrographs of the chitosan

films containing andiroba oil for 2% chitosan. Micrographs of films for 1% (w/w) chitosan were quite similar, and they are not presented. The emulsified films showed structural discontinuities associated with the formation of two phases (lipid and polymer) in the matrix. The oil-free films had a smooth and homogeneous microstructure with no irregularities like air bubbles or oil droplets detected (micrograph not shown), as our group had previously published<sup>[27]</sup>. The number of oil droplets increases as the concentration of oil increases. The cross-sections of the emulsified films show that droplets have an ellipsoidal shape, which was also verified by other authors<sup>[26,28,29]</sup>. This ellipsoidal shape can be attributed to the weight of the chitosan over the droplets during the drying. It is important to mention that no surfactant was used in the preparation of our films.

Kokoszka et al.<sup>[30]</sup> studied whey protein/rapeseed oil emulsion film. Unlike what was observed in this study, they verified that the oil is not well distributed throughout



**Figure 2.** SEM of the surface (left) of chitosan film viewed at a magnification of 500×, and cross-sections (right) viewed at a magnification of 3000× (Chitosan 2.0% w/w).



the film on both sides. According to Kokoszka et al.<sup>[30]</sup>, the oil droplets are more concentrated on the side exposed to the air since the film retraction during drying produces changes in its structure, which becomes denser, and the oil droplets migrate towards the side exposed to the air, thus favoring coalescence.

### 3.2 Fluid handling capacity

The moisture content of the wounds must be carefully controlled to achieve optimal rates of wound healing. The healing process can be influenced by changes in the moisture content of a wound and the skin around it. A wound that is too dry may delay or impair the healing while excess fluid can cause maceration or infection. Thus, the optimal healing environment is achieved by applying an appropriate dressing that should be removed in time to avoid maceration or adherence<sup>[31]</sup>. Donor sites, unspecified granulating wounds, and third-degree burns generate between 3.4 and 5.1 g of exudate per 10 cm<sup>2</sup> over a 24 hour period<sup>[32]</sup>.

The MVTR, ABS, and FHC are presented in Table 1. Thomas and Young<sup>[31]</sup> evaluated these properties for two commercial dressings: ActivHeal (Advanced Medical Solutions) and Allevyn Adhesive (Smith & Nephew) – both are film-backed foam dressings. They verified the absorbencies to be 3.44 and 4.32 g/10 cm<sup>2</sup>/24 h, respectively, for ActivHeal and Allevyn Adhesive. Our results for absorbency are very close to these values, especially when the concentration of chitosan is 2.0% w/w. Furthermore, the MVTR were 1.67 and 12.35 g/10 cm<sup>2</sup>/24 h, respectively, for ActivHeal

and Allevyn Adhesive<sup>[31]</sup>. Regarding the MVTR, our results were close to those for ActivHeal. Also, it is important to mention that both these commercial dressings are foam dressings, so these properties are usually greater than those for film dressings.

The WVTR decreases with the oil concentration in both chitosan concentrations; this could be attributed to the hydrophobicity of the andiroba oil. Concerning the Absorbency, the incorporation of andiroba oil decreased this parameter especially for the 2% chitosan films.

### 3.3 Mechanical properties

To adequately protect a wound, the film must maintain its integrity against external stress during the manipulation and application, even when it is on the wound. Tensile strength indicates the maximum tensile stress that the film can sustain, elongation at breaking point is the maximum change in length of a test specimen before breaking, and the Young's modulus is a measure of the stiffness of the film<sup>[21]</sup>. The mechanical properties of chitosan film are shown in Table 2. Both the concentrations significantly affected the mechanical properties of the emulsified films.

Young's modulus and the tensile strength of the chitosan film increased when andiroba oil was incorporated into the chitosan matrix. These two properties are usually higher for samples with 2.0% of chitosan. The addition of low concentrations (0.1%) of andiroba oil causes an increase in these properties if compared to oil-free film. Whenever the concentration of oil increases, these two properties decrease

**Table 1.** Fluid handling properties of the film dressing for different concentrations of andiroba oil.

Concentration (% w/w)		MVTR (g/10cm <sup>2</sup> /24h)	Absorbency (g/10cm <sup>2</sup> /24h)	FHC (g/10cm <sup>2</sup> /24h)
Chitosan	Andiroba oil			
1.0	0.0	1.73 ± 0.07 <sup>a</sup>	1.67 ± 0.12 <sup>a</sup>	3.39 ± 0.14
	0.1	2.83 ± 0.28 <sup>b</sup>	0.62 ± 0.06 <sup>b</sup>	3.45 ± 0.29
	0.5	1.53 ± 0.12 <sup>c</sup>	1.59 ± 0.16 <sup>a</sup>	3.12 ± 0.20
	1.0	1.31 ± 0.13 <sup>d</sup>	1.49 ± 0.08 <sup>a</sup>	2.81 ± 0.15
2.0	0.0	3.24 ± 0.22 <sup>c</sup>	3.43 ± 0.21 <sup>c</sup>	6.67 ± 0.30
	0.1	1.88 ± 0.12 <sup>a</sup>	2.32 ± 0.18 <sup>d</sup>	4.20 ± 0.21
	0.5	1.28 ± 0.13 <sup>d</sup>	2.74 ± 0.26 <sup>c</sup>	4.02 ± 0.29
	1.0	1.36 ± 0.07 <sup>c,d</sup>	2.63 ± 0.21 <sup>d,e</sup>	3.98 ± 0.22

Different superscripts within the same column indicate significant differences among formulations (p<0.05).

**Table 2.** Effect of the concentrations of chitosan and andiroba oil on Young's modulus, tensile strength, and elongation at breaking point, for the emulsified films.

Concentration (% w/w)		Young's modulus (MPa)	Tensile strength (N/m <sup>2</sup> )	Elongation at breaking point (%)
Chitosan	Andiroba oil			
1.0	0.0	36.52 ± 1.49 <sup>a</sup>	169.99 ± 14.06 <sup>a</sup>	13.59 ± 1.20 <sup>a</sup>
	0.1	67.67 ± 5.32 <sup>b</sup>	263.47 ± 22.62 <sup>b</sup>	13.48 ± 1.29 <sup>a</sup>
	0.5	55.39 ± 4.96 <sup>c</sup>	204.16 ± 18.71 <sup>c</sup>	13.07 ± 0.40 <sup>a</sup>
	1.0	34.63 ± 3.99 <sup>a</sup>	172.60 ± 17.16 <sup>a</sup>	13.69 ± 1.03 <sup>a</sup>
2.0	0.0	62.02 ± 6.75 <sup>b,c</sup>	171.21 ± 14.01 <sup>a</sup>	22.56 ± 1.86 <sup>b</sup>
	0.1	98.50 ± 9.58 <sup>d</sup>	272.55 ± 17.00 <sup>b</sup>	9.26 ± 0.89 <sup>c</sup>
	0.5	81.98 ± 3.55 <sup>c</sup>	250.20 ± 22.77 <sup>b</sup>	9.23 ± 0.92 <sup>c</sup>
	1.0	57.32 ± 4.37 <sup>c</sup>	182.67 ± 17.11 <sup>a,c</sup>	12.43 ± 1.14 <sup>a</sup>

Different superscripts within the same column indicate significant differences among formulations (p<0.05).

and are closer to those of the control film (oil-free chitosan film). A decrease in these two properties with the increase of oil was also observed in emulsified films with chitosan/basil essential oil<sup>[19]</sup>, chitosan/thyme essential oil<sup>[20]</sup>, chitosan/tea tree oil<sup>[22]</sup>, cassava starch-chitosan/oregano essential oil<sup>[33]</sup>, and whey protein/olive oil<sup>[34]</sup>.

The mechanical properties obtained by the addition of oil may be related to the structural arrangement of the lipid phase in the chitosan matrix. A number of discontinuities increases as the concentration of andiroba oil increases, which could explain the decrease in Young's modulus and tensile strength.

No significant effect on elongation at breaking point was observed for 1% of chitosan and when the andiroba oil concentration increased. This effect was also reported by other authors when adding oil to a chitosan matrix<sup>[29,35,36]</sup> and it could also be attributed to the structural discontinuities provoked by the incorporation of the oil. Moreover, the incorporation of oil promoted a substantial reduction in the elongation of the films containing 2% chitosan.

The effects of chitosan concentration on the mechanical and physical properties of the films seem to be greater than the effect of the oil. Most likely, the oil concentrations are not sufficiently high so that they could be more important than chitosan concentration. But it would be difficult to produce films with higher oil concentration because we would have exudation of the oil from the film, even if we had used one tensoative.

### 3.4 Thermogravimetric analysis

TGA was performed to evaluate the thermal stability of the chitosan powder, andiroba oil, and chitosan-andiroba oil films. Thermal degradation is displayed in Table 3 and Figures 3, 4.

Table 3 and Figure 3 show that chitosan powder mainly loses mass due to decomposition between 200 °C and 400 °C, and especially in the range of 200 °C to 300 °C. On the other hand, the weight loss of the pure andiroba oil is mainly between 300 °C and 400 °C, while above 500 °C andiroba oil is entirely decomposed. The TGA curves have different behavior above and below the temperature of the oil degradation for andiroba oil/chitosan films. Below the temperature of oil degradation, the oil seems to

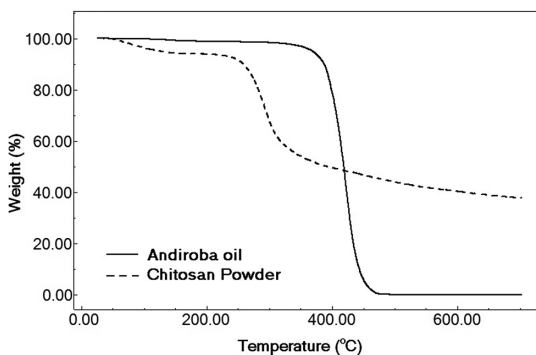


Figure 3. TGA thermograms of andiroba oil and chitosan powder.

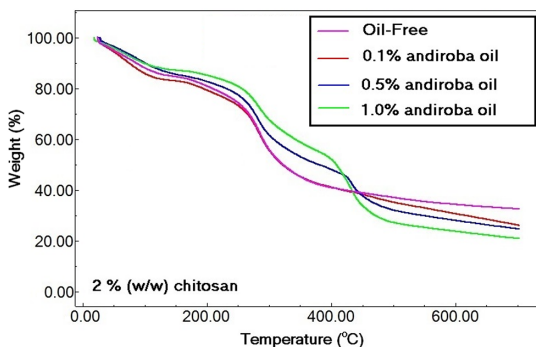
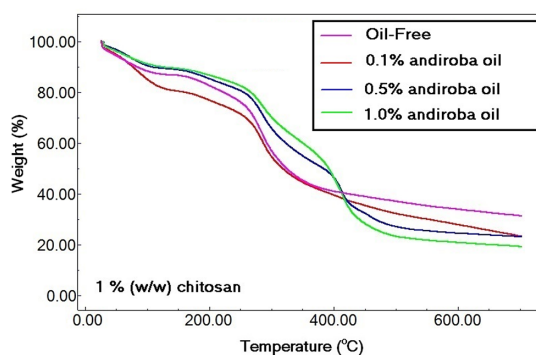


Figure 4. TGA thermograms of 1% and 2% (w/w) chitosan films containing andiroba oil.

Table 3. Thermal analysis of chitosan, andiroba oil, and andiroba oil/chitosan film in an N<sub>2</sub> atmosphere.

Concentration (% w/w)		% weight loss						
Chitosan	Andiroba oil	100 °C	200 °C	300 °C	400 °C	500 °C	600 °C	700 °C
1.0	0.0	12	17	43	59	63	66	69
	0.1	14	22	44	59	66	71	75
	0.5	10	15	34	54	74	76	78
	1.0	9	13	30	54	77	80	82
2.0	0.0	12	19	44	59	63	66	67
	0.1	14	21	44	59	65	69	74
	0.5	10	17	38	52	68	72	75
	1.0	10	15	32	48	73	76	79
Chitosan powder		4	6	32	50	56	60	62
Andiroba oil		0	1	2	21	100	100	100

stabilize the film, but when the temperature is higher than the temperature of the oil degradation, the oil appears to promote the opposite. If we compare the weight loss up to 400 °C (Figure 4 and Table 3), the increase of andiroba oil concentration decreases the weight loss values (the TGA curves are in a superior position for 0.5 and 1.0% of andiroba oil). However, when the temperature is above 400 °C, there is one inversion and the TGA curves become inferior for these higher concentrations of andiroba oil. We also observed in Figure 4 that the incorporation of andiroba oil in chitosan film tends to shift the thermal degradation zone to higher temperatures. Such change is attributed to an increase in thermal stability by the incorporation of andiroba oil, and they were also supported by the differential thermogravimetric analysis - DTG (data not shown). Regardless, Figure 4 shows that both chitosan concentrations presented the same behaviour when the andiroba oil concentration increases.

Pelissari et al.<sup>[33]</sup> verified that the addition of oregano essential oil to chitosan-starch films did not influence the thermal stability of these films; however, they observed an increase in residue percentage after the incorporation of the oregano essential oil.

### 3.5 Differential scanning calorimetry

Figures 5, 6 shows the DSC results for the chitosan powder, andiroba oil, and andiroba oil/chitosan films. The results obtained from the DSC include the temperatures, and their respective  $\Delta H$  values are presented in Table 4.

The DSC heating curve for pure andiroba oil showed one endothermic peak at 42.57 °C and some exothermic peaks between 180 °C and 360 °C. Oils are one complex mixture of triacylglycerols (TAGs) acting also as a solvent for minority components, such as vitamins, pigments, phenolic compounds, phospholipids, free fatty acids, and mono- and diacylglycerols<sup>[37]</sup>. The five top TAGs present in Andiroba oil are, in descending order: Palmitic-Oleic-Oleic, Palmitic-Palmitic-Oleic, Palmitic-Oleic-Stearic, Oleic-Oleic-Oleic/Stearic-Oleic-Linoleic, and Palmitic-Linoleic-Oleic<sup>[8,37]</sup>. Thus, the first endothermic peak is probably associated with the fusion of free fatty acids. The exothermic peaks are probably associated with the decomposition of TAGs.

Matos<sup>[38]</sup> conducted a thermal study of pure fatty acids by DSC. He observed endothermic peaks for melting temperatures at 64.55 °C ± 0.67 and 71.36 °C ± 0.30, respectively, for the palmitic acid and stearic acid. Exothermic peaks of decomposition were observed at 235.24 °C ± 7.57, 241.87 °C ± 5.61, 238.79 °C ± 12.11, and 268.26 °C ± 18.16, respectively, for the palmitic, stearic, oleic, and linoleic acids. These fatty acids also showed a sequence of many exothermic peaks above 350 °C<sup>[38]</sup>.

The exothermic peaks between 200 °C and 300 °C are probably due to the decomposition of the TAGs and fatty acids into smaller chains while the sequence of peaks above 350 °C can be attributed to the decomposition of these compounds into even smaller chains. The chitosan-andiroba oil film presented two peaks ( $T_1$  and  $T_2$ ) while the andiroba oil-free film showed also two peaks ( $T_1$  and  $T_2$ ) with a shoulder on the descending side. These shoulders could be attributed to the decomposition described above ( $T > 350$  °C). The increase in andiroba oil concentration promotes a decrease

in the temperature  $T_1$ , which is attributed to the evaporation of water associated with the chitosan and could also be attributed to the hydrophobic properties of the andiroba

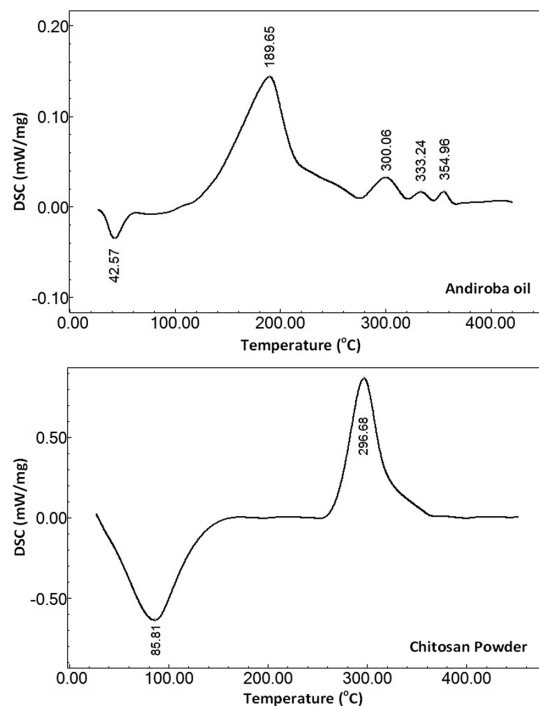


Figure 5. DSC run curves for andiroba oil and chitosan powder.

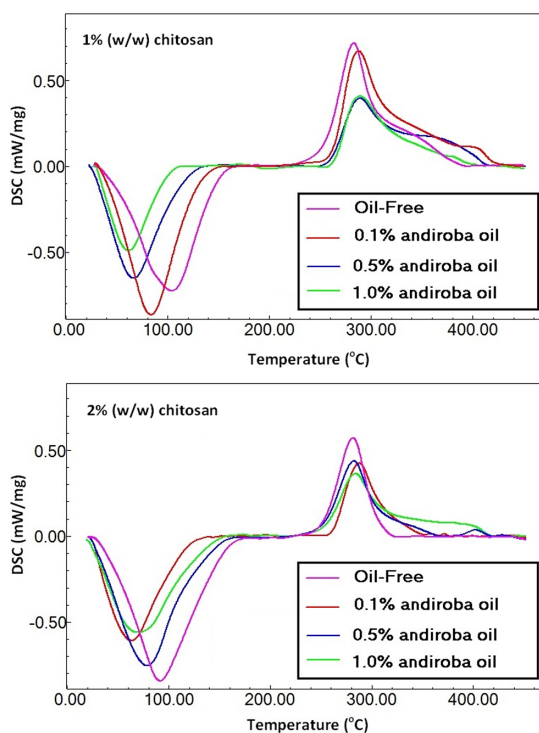


Figure 6. DSC run curves for 1% and 2% (w/w) chitosan films containing andiroba oil.

**Table 4.** Temperatures and enthalpy measured by DSC.

Concentration (% w/w)		Temperature (°C)		Enthalpy (J/g)	
Chitosan	Andiroba oil	T <sub>1</sub>	T <sub>2</sub>	ΔH <sub>1</sub>	ΔH <sub>2</sub>
1.0	0.0	103.98	283.02	-288.99	233.48
	0.1	83.60	288.00	-297.25	268.72
	0.5	66.51	288.88	-210.18	181.10
	1.0	61.29	289.43	-112.70	155.37
2.0	0.0	91.07	282.37	-340.13	266.23
	0.1	63.36	287.69	-203.67	102.58
	0.5	77.92	282.47	-259.22	116.44
	1.0	69.55	283.72	-228.50	138.21
Chitosan powder		85.81	296.68	-248.53	198.83

oil. Such decrease in this temperature was, even more, prominent for 1% chitosan films. This fact can be explained by the hydrophobic character of andiroba oil and also by the reduction in enthalpy related to this evaporation ( $\Delta H_1$ ).

The temperature T<sub>2</sub> for the decomposition of chitosan is little affected by the addition of andiroba oil; however, the enthalpy ( $\Delta H_2$ ) of the chitosan decomposition is reduced, which shows that andiroba oil reduces the heat generated by the decomposition of chitosan, thus confirming the increased thermal stability of the film that was observed in the TGA results.

#### 4. Conclusions

Chitosan films containing andiroba oil were obtained with satisfactory properties for use as a dressing to heal wounds. The addition of andiroba oil significantly modified the mechanical and barrier properties of the films and promoted greater thermal stability for the films. About the barrier properties, it was found that all of the films exhibited properties which were compatible with commercial dressings, and the films with the highest concentration of chitosan showed best results. Concerning the mechanical properties, the Young's modulus and tensile strength increased with the addition of andiroba oil; however, as the concentration of andiroba oil increases, the values for these properties approach the values observed for the film without andiroba oil. While elongation remained practically unchanged for the films with lower concentrations of chitosan (1.0%), for the films with higher concentrations of chitosan, the addition of andiroba oil promoted a decrease in elongation.

#### 5. Acknowledgements

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