RESEARCH ARTICLE

Characterization of films and film-forming solutions of chitosan/thermoplastic rice starch associations: Role of starch oxidation and plasticizer type in the molecular interactions

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Abstract

Polysaccharide films are alternative sources to replace fossil-based compounds. This study aimed to use rice starch modified by gelatinization and oxidation processes. Different properties were observed after blending it with chitosan and polyols, as evaluated on the film-forming solutions by rheology. The interaction between chitosan and oxidized starch appeared higher, as the plasticizers exhibited a low effect on the rheological behavior. Polyols played an essential role in chitosan/gelatinized associations, driven by the number of OH groups. Films were evaluated by thermogravimetry (TGA), differential scanning calorimetry (DSC), water vapor permeability (WVP), water solubility, and tensile tests. The effect of the plasticizer type on chitosan/gelatinized starch was related to an increase in the temperature of the endothermic peak observed by DSC, confirming the role of hydroxyl groups of polyols in the polymer–polymer chain associations. The WVP and solubility decreased with starch oxidation due to increased chitosan–starch interactions. The decrease in tensile strength with adding polyols was correlated to reduced direct interactions between chitosan and starch chains. A drop of about 77% in elongation at break was observed in chitosan/oxidized starch film. On the other hand, elongation at break increased by around 14% in films with sorbitol (SO).

Highlights

- Starch oxidation reduces the viscosity of film-forming solutions.
- The number of OH groups influences the gelation point measured by rheology.
- The WVP is reduced in oxidized starch/chitosan associations.
- Plasticizers enhanced the elongation at break of polysaccharide films.

KEYWORDS

chitosan, rheology, rice starch, thermoplastic

1 | INTRODUCTION

Research focusing on developing biodegradable films from renewable sources has been growing in the last decades due to environmental concerns. Biodegradable films based on natural polymers can be produced at a low cost and on a large scale and are a favorable alternative to current materials derived from nonrenewable fossil sources.[1] Due to the variety of natural polymers available and the different processing methods, biodegradable films are classified as biodegradable, compostable, and even edible, which opens new application possibilities.[2] Therefore, a criterion evaluation of the components and manufacturing processes is essential in designing new biopolymeric materials. Among all biopolymers, polysaccharides play an essential role in developing films as alternatives to synthetic polymer materials due to their renewable and recyclable nature.[3]

Starch has been extensively used in the preparation of films due to its inexpensiveness, renewability, and biodegradability.[4] Starch is the reserve polysaccharide in plants and is widely used in the food, paper, pharmaceutical industries, and others.[5] When native starch does not exhibit a required property that limits its application, several methods can be applied to overcome that restriction. The most known method is blending biodegradable polymers to achieve specific properties. Martins et al.[6] described the addition of rice and potato starch nanocrystals (SNCs) to a low-density polyethylene (LDPE) matrix with an increase in elongation at break of around 250% when compared with the standard LDPE film. In another study, low-density polyethylene/high-density polyethylene/thermoplastic starch (LDPE/HDPE/TS) blends were prepared at different ratios showing an influence of the polysaccharide in the rheological properties.[7] In our study, we focused on blending starch with another biodegradable polymer. Vásconez et al.[8] proposed the blending with chitosan to change the physical and functional properties of starch, like reducing water affinity and improving its mechanical properties due to the formation of intermolecular hydrogen bonds between amino and hydroxyl groups of chitosan and hydroxyl groups of starch.

Chitosan is the main derivative of chitin, found in the exoskeletons of arthropods, cell walls of fungi, and shells of crabs, squid, and shrimp.[9] It is the only alkali polysaccharide found in nature, and the amino groups present in its backbone can be easily protonated at a pH below 4. This characteristic allows chitosan to interact with other acids or neutral polysaccharides, like starch.[10]

Even though the simple blending approach is commonly found in the literature, molecular structure modification of starch can also be considered using physical, chemical, or enzymatic methods. In general, chemical treatments are the most exploited to bring new functionalities to starch and access new properties.[11] The oxidation process provides unique functional properties such as low viscosity, high stability, and excellent film-forming characteristics.[2] Periodic acid is a selective oxidizing agent responsible for cleaving the C-2–C-3 linkages of glucose units and producing dialdehyde groups[12] in the starch backbone, as shown in Figure 1.

Thus, evaluating how the chemical modification of starch affects the final properties of the chitosan/starch association is one of the objectives of this study. Nevertheless, due to the interaction between the components, polymeric films are often brittle and stiff, which limits their mechanical properties and applications.[13] The insertion of plasticizes in the polymeric system reduces polymer–polymer interactions resulting in a soft and flexible film. Common plasticizers described in the literature include monosaccharides, lipids, and polyols. Different behavior in properties is obtained depending on the type of plasticizer used.[14]

Polyols are low molecular weight molecules containing multiple hydroxyl groups often used in polymeric systems. Incorporating polyols facilitates the processability and workability of polymers, such as enhancing flexibility and reducing the rigidity of the films.[15] Glycerol (GL) is the most used plasticizer due to its availability and plasticization efficiency. Liu and coworkers described that GL facilitates the interaction between chitosan and starch by hydrogen bonds as Fourier-transform infrared spectroscopy (FTIR) bands related to GL-free have shifted to higher wavenumbers after the polyol addition.[16] Nevertheless, other small alcohol molecules are less explored, even though can provide enhanced mechanical and physicochemical properties to biopolymer films.

Then, the objective of this study was to assess both the effect of starch oxidation and evaluate the influence of plasticizer agents (GL, sorbitol [SO], and ethylene glycol [EG]) on the rheological, thermal, mechanical, and water vapor permeability (WVP) properties of chitosan/rice starch blends.
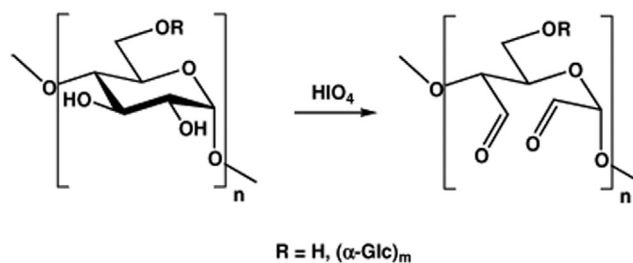

R = H, (α-Glc)ₘ

FIGURE 1 Dialdehyde starch oxidation.

2 | EXPERIMENTAL

2.1 | Materials

Chitosan was extracted from squid pens (*Loligo* sp.). Rice starch containing approximately 75% amylopectin and 25% amylose was supplied by Sigma-Aldrich. Analytical grade GL with a molar mass of 92 g mol^{-1} was supplied by Aldrich. Sorbitol (SO) with a molar mass of 182 g mol^{-1} and EG with a molar mass of 62 g mol^{-1} were purchased by Merck. All other chemicals were analytical grade.

2.2 | Chitosan extraction and characterization

Chitosan was obtained and characterized (molecular weight 403 kDa; degree of acetylation 9%) as previously described.[17,18] In short, the raw material was immersed in a 0.55 mol L^{-1} HCl solution at room temperature for 2 h to remove inorganic materials. Next, the precipitate solid was immersed in a 0.3 mol L^{-1} NaOH solution at 80°C for 1 h to eliminate proteins. The last step consisted of alkaline deacetylation of β-chitin in a 40% (w/w) NaOH solution for 3 h at 80°C, followed by water washings and drying at 40°C for 5 days. One gram of the extracted chitosan was dissolved in 1% (w/w) acetic acid using a constant overnight stirring.

2.3 | Starch gelatinization

Starch powder and 50 mL of deionized water were added to a three-neck flask with a stirrer, thermometer, and nitrogen flow. The mixture was mechanically stirred, gelatinized at 90°C for 30 min, and cooled at room temperature, leading to a 2% (w/w) homogeneous solution.[19] During the heating process, the excess water breaks the crystalline structure of the starch molecules, exposing their hydroxyl groups and increasing the swell ability and solubility of the starch.

2.4 | Starch oxidation

The starch powder was oxidized with 5% (w/w) periodic acid (H$_5$IO$_6$) for 24 h at 25°C under stirring following an adapted method described by Hoffmann et al.[20] The obtained material was dialyzed against deionized water for 72 h and then lyophilized. A 2% (w/w) solution was prepared by dissolving the oxidized rice starch in deionized water under constant magnetic stirring.

2.5 | Fourier-transform infrared spectroscopy (FTIR)

FTIR analysis was employed to analyze the characteristic bands of the chitosan extracted from the squid pens waste source. Additionally, the spectroscopic technique was applied to investigate the effectiveness of the oxidation process on the starch backbone. FTIR spectra were obtained using a Bomem Michelson Series at a 400 to 4000 cm^{-1} interval with a resolution of 4 cm^{-1}.

2.6 | Film-forming solution

Each plasticizer was first dispersed in the 2% (w/w) starch solution (gelatinized or oxidized) under vigorous stirring. In sequence, the mixture was added to the 1% (w/w) chitosan solution resulting in a 2:1:1 (w/w/w) ratio blend. Samples were denominated as CSge or CSox, representing the starch form used (gelatinized or oxidized), followed by the capital letters (EG, GL, or SO) identifying the plasticizer agent used, EG, GL, and SO, respectively. The blends were immediately subjected to rheological measurements. For the blends without plasticizers, a proper amount of solvent was added to avoid dilution effects.

2.6.1 | Rheology measurements

The rheological properties were determined in a tension-controlled rheometer AR1000N from TA Instruments. A cone geometry (60 mm in diameter and angle of 0.5°) was employed as a measuring system using a gap of 15 μm. Initially, the viscoelastic region was determined using a constant frequency of 1 Hz and temperature of 25°C, which allows the measurement of the G' and G'' as a function of strain. The frequency sweep test was carried out within the viscoelastic region (strain of 10%), using a frequency range from 0.1 to 100 rad s^{-1} at 25°C. The temperature dependence behavior was accessed by varying the temperature from 25 to 75°C, applying a heating rate of 5°C min^{-1} and strain of 10%. Flow properties were evaluated as a function of shear rate from 0.01 to 100 s^{-1} at a temperature of 25°C.

2.7 | Films preparation

Films of starch/chitosan/plasticizers were prepared by casting in Teflon® molds the film-forming solutions at room temperature for 24 h and relative humidity of 50%. After the solvent evaporation, films were peeled and

stored in a humidity-controlled chamber (65%). A constant amount of 6 g of total solution was cast onto 16.5 cm^2 of the tray area, which allows the comparison of the thickness depending on the type of starch and the presence of the plasticizer agent.

2.8 | Characterization of the films

2.8.1 | Thermal analysis

DSC experiments were recorded on a DSC-2010 (TA Instruments). Samples (6 mg) were heated at the rate of $10°C$ min^{-1} from 25 to $450°C$ under a dynamic synthetic air atmosphere. A thermogravimetric analyzer (TGA-2050 TA Instruments) was employed to measure the weight loss of films under heating. Samples (10 mg) were heated between 25 and $800°C$ at $10°C$ min^{-1} in a synthetic air atmosphere.

2.8.2 | Film thickness

Film thickness was measured using a micrometer (Model M110-25, Mitutoyo MFG. Co. Ltd., Japan). Thickness measurements were taken at 10 different points along the film, and the mean values were used to calculate film tensile strength and WVP.

2.8.3 | Water vapor permeability

The WVP of the films was calculated using ASTM E96, as described by Garcia et al.,[21] with some modifications. Each film sample was fixed in a 0.007 m^2 circular hole containing anhydrous calcium chloride (0%). The cups were weighed and placed in a humidity chamber at 75% relative humidity (NaCl saturated solution) and $25°C$. The cups were weighed six times throughout 9 h. The water vapor transferred through the films at different time intervals was determined from the weight gain of the cups. A plotted curve of changes in weight (g) over time (min) was obtained to determine the slope, used to calculate the water vapor transmission rate (WVPR), as shown in Equation (1):

$$WVPR = \frac{m}{t} \cdot \frac{1}{A}$$ (1)

where m/t is the slope of the curve (g over min), and A is the permeation area of the sample (m^2). Thus, WVP in the films was determined using Equation 2:

$$WVP = \frac{WVPR.t}{sp(RH_1 - RH_2)}$$ (2)

where t is the film thickness (mm), sp is the water vapor pressure at $25°C$ (3119 Pa), RH_1 is the relative humidity inside the chamber (75%), and RH_2 is the relative humidity inside the cup (0%).

2.8.4 | Solubility

Films were previously dried overnight in an oven set up at $50°C$. Pieces of known weight were swollen in 50 mL of deionized water at $25°C$ for 1 h. The swollen films were removed from the solution and dried at $60°C$ until constant weight. The percentage of total soluble material of the chitosan/starch films was calculated as shown in Equation (3). Films were analyzed at minimum in triplicate.

$$Solubility = \left[\frac{(Initial\,dry\,weight) - (Final\,dry\,weight)}{Initial\,dry\,weight} \right] \times 100$$

(3)

2.8.5 | Mechanical properties

Mechanical properties were measured at $25°C$ in a DMA 2980 (TA Instruments) operating with the tension clamp, using a force rate of 1 N min^{-1} and a distance between clamps of 10 mm. The films (15 mm long$° \times °$6.3 mm wide) were stored in controlled humidity (65%), and tests were replicated five times for each type of film.

3 | RESULTS AND DISCUSSION

The film-forming solutions were, in all cases, homogeneous transparent solutions without the presence of precipitates or undissolved materials. The films were prepared by casting through solvent evaporation and easily peeled from the Teflon trays. In general, brittle films were obtained in the absence of plasticizers. Nevertheless, all maintained the shape of the mold used for preparation. Additionally, films prepared with oxidized starch appeared yellowish compared to those with gelatinized starch.

3.1 | FTIR

The FTIR spectra of chitosan, gelatinized rice starch, and oxidized rice starch are shown in Figure 2.
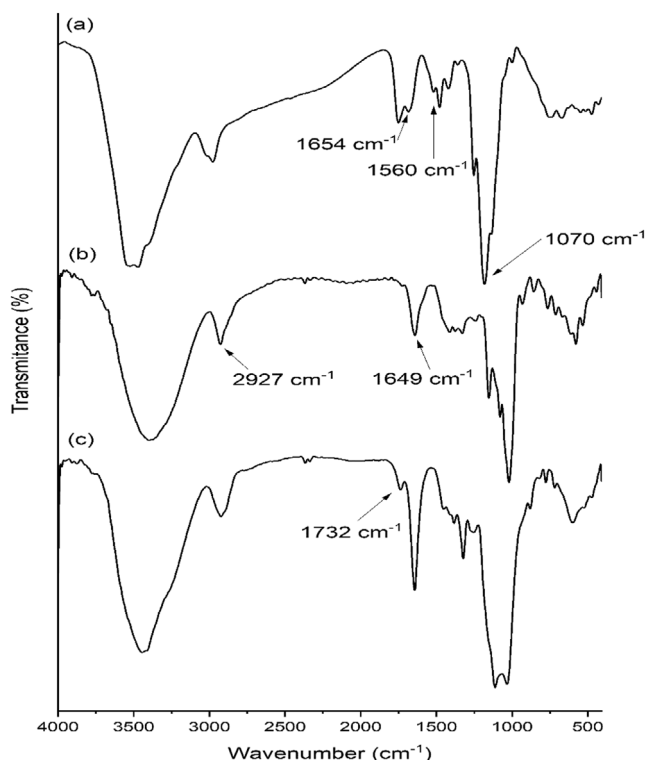
FIGURE 2 FTIR spectra of (A) chitosan; (B) gelatinized rice starch, and (C) oxidized rice starch.

Characteristic bands for chitosan (Figure 2A) were observed: 1654 cm^{-1}—carbonyl stretching of amide I; 1560 cm^{-1}—due to N—H in-plane bending vibrations and C—N stretching of amide II; 1323 cm^{-1}—CO—N group; 1070—1030 cm^{-1} C—O stretching; 3450 cm^{-1} O—H stretching. The main bands detected in the chitosan spectrum agree with other studies,[22,23] which indicate a successful extraction from a waste raw material, such as squid pens.

The spectrum of starch (Figure 2B) depicted characteristic absorption bands at 3450 cm^{-1} corresponding to OH stretching; 2927 cm^{-1} referring to C—H stretching; and 1649 cm^{-1} assigned to the deformation of OH bending of water. The C—O bond stretching is related to the 765 and 1120 cm^{-1} bands. Changes in the chemical structure of oxidized starch were also examined by FTIR. The presence of an additional band at 1732 cm^{-1} (Figure 2C) confirms the oxidation process in the starch molecule.[24] This band is attributed to the C=O valence vibration of the aldehyde group generated by the ring-opening of the glucose units. The aldehyde groups of oxidized starch are essential to the formation of a network between the amino groups of chitosan.

3.2 | Rheological characterization

The viscous and elastic properties of the film-forming solutions were evaluated by rheological studies. The linear

viscoelastic region (LVE) was initially determined by an amplitude sweep measurement. The viscoelastic properties, defined by the elastic modulus (G') and viscous modulus (G''), were measured as a function of strain and provided the range in which further tests could be carried out without destroying the structure of the samples. The diagram of moduli against strain indicates the LVE region, characterized by a constant moduli value. Then, a strain value (10%) within this plateau range was chosen for the subsequent oscillatory measurements.

The interactions between chitosan and starch were studied in relation to the frequency sweep properties of a wide range of systems (Figures 3 and 4). The rheological behavior of chitosan/gelatinized starch association (CSge) denotes a typical gel-like nature, in which G' was always higher than G'' over the whole frequency range. When plasticizers were added to the CSge sample, differences were observed between the distinct agents, suggesting different physical associations between the components (Figure 3).

EG was the plasticizer that showed less effect in. the chitosan/gelatinized starch interaction, as at low-frequency G' was below G'' and after around 1 rad s^{-1}, the contribution of the elastic and viscous moduli was barely the same. On the other hand, the addition of GL led to the CSgeGL sample exhibiting a rheological behavior typical a viscoelastic solution, whereby $G'' > G'$ at low frequency, but G' is higher than G'' at high frequency values (above 10 rad s^{-1}). This is commonly related to flow associated with chain disentanglement. Additionally, a crossover in the moduli was observed at 0.8 rad s^{-1}, implying a high relaxation time of the polymeric chains.[25] The CSgeSO sample displayed the G'' higher than G' all over the frequency studied range. Even though a crossover of moduli was not obtained in the measurement interval, it is observed that it will probably occur at a frequency value up to 100 rad s^{-1}. The frequency tests indicate that GL and SO are the plasticizers that affect the polymeric association. Furthermore, GL induced an entanglement density between the components,[25] reflecting a high plasticization effect due to the crossover observance at a low frequency compared to the CSgeSO sample.

The samples prepared by mixing chitosan and oxidized starch solutions exhibited a different behavior than when gelatinized starch was used (Figure 4). In general, G' values were at least 10 times lower in the sample prepared with oxidized starch. The elastic modulus is a prompt indication of the strong characteristic of polymeric associations. Indeed, a high G' modulus implies that a high association network was formed.

For all the samples, G'' was higher than G' in all the frequency interval exhibiting a predominant viscous behavior. In the studied range, no crossover of the moduli
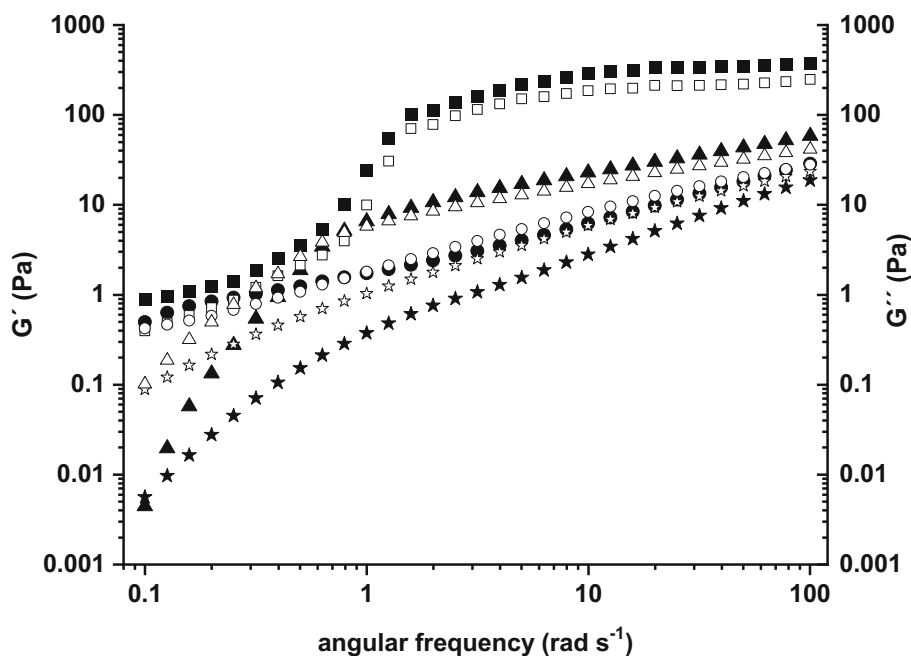
FIGURE 3 G' and G'' moduli as a function of angular frequency for CSge —■— G', —□— G''; CSgeGL —▲— G', —△— G''; CSgeEG —●— G', —○— G'' and CSgeSO —★— G', —☆— G''.
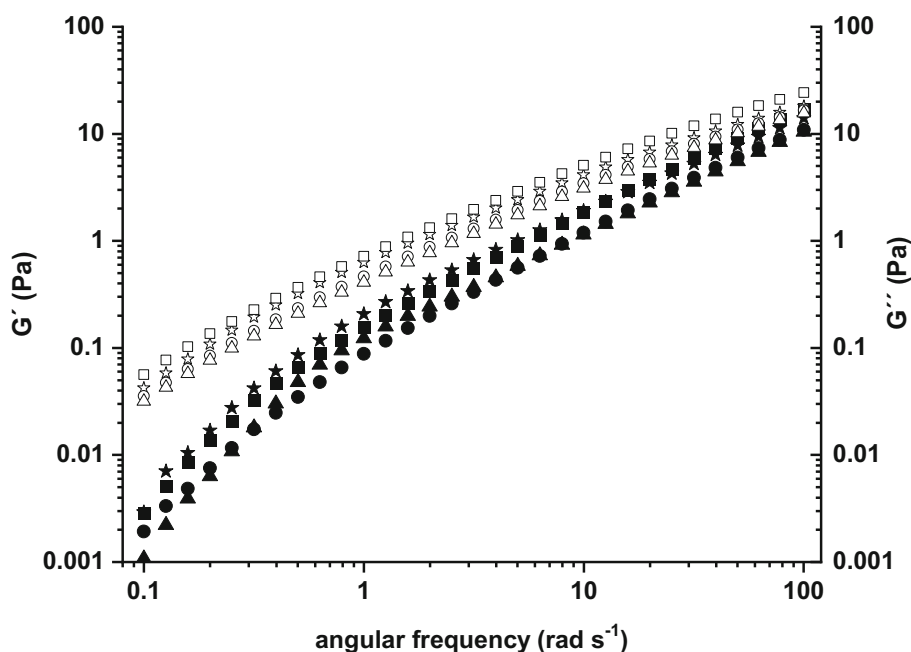

FIGURE 4 G' and G'' moduli as a function of angular frequency for CSox —■— G', —□— G''; CSoxGL —▲— G', —△— G''; CSoxEG —●— G', —○— G'' and CSoxSO —★— G', —☆— G''.

was detected, which probably occurs at frequencies higher than 100 rad s^{-1}, and samples can be classified as diluted solutions.[26] The moduli dependence is intrinsically related to the chitosan concentration. In a study by Serrero et al.,[25] chitosan concentrations of 1.9% and 2.3% associated with oxidized starch led to a crossover in moduli (at 4 rad s^{-1}) and the observance of G' higher than G'', respectively. In fact, in our study, a distinct behavior was displayed, probably related to a chitosan concentration of 1%. Furthermore, in a previous study, we observed a weaker interaction between the blend components wherein oxidized corn

starch was associated with chitosan.[27] Regardless of the slight changes after plasticizer addition, their effect is less evident. At low frequencies, the weak action of EG as a plasticizer agent is distinguishable.

The temperature sweep tests were carried out to determine the sol–gel transition of the samples (Figures S1 and S2). Depending on the composition, it was possible to observe the gelation behavior when samples were subjected to temperature ramping.

The moduli displayed a temperature-dependent profile in which a progressive increase in their values was

observed in the analyzed temperature range. It was noted that no gelation occurred between chitosan and gelatinized starch in the studied range. In fact, for CSge, G'' is higher than G', which implies the gelation point appeared below 25°C. Including plasticizers altered the gelation temperature of the samples, confirming that these additives are involved in the polymer chains association (Table 1).

The active hydroxyl groups of the alcohol molecules make them an effective plasticizer for polymers rich in —OH and —NH groups. As both polysaccharides possess these functional groups in their structures, the plasticizers used in this study can increase the free volume between the polymeric chains, reflecting high chain mobility. Even though the same molar amount of EG, GL, and SO were used in the composition of the samples, their effect is particularly related to the number of OH groups. EG, GL, and SO exhibit two, three, or six OH in their structure, respectively. As the plasticizer effect is associated with those interactions between OH groups, the polysaccharide chains are relatively more affected by SO molecules when components are in solution. Then, the increase in gelation temperature observed for CSgeEG, CSgeGL, and CSgeSO is closely related to the OH associations in the blends.

The sample prepared with oxidized starch (CSox) displayed the gelation temperature in the analyzed range, at 52.2°C. Additionally, the temperature where $G' = G''$ are higher for all the samples prepared with oxidized starch than those containing the gelatinized polysaccharide. This indicates that polyols play an essential role in increasing gel strength. The addition of plasticizers increased the gelation temperature, although their action is less pronounced. This meant that higher interaction occurred between chitosan and oxidized starch, and the plasticizers had less effect on occupying and increasing the space between the polymeric chains. These results agreed with the frequency sweep measurements, wherein plasticizers were effectively active in chitosan/gelatinized

starch associations, and less interference was observed for chitosan/oxidized starch samples.

Steady shear behavior reflected the structure of chitosan/starch film forming associations. It was performed to study their structure and properties after plasticizer addition and when oxidized starch was used in the composition. Also, the relationship between viscosity properties and chitosan/starch interactions can provide information about their processability, as flow characteristics are directly related to the thickness spreadability and uniformity of the blend film.

All the samples displayed a shear-thinning behavior, wherein the viscosity reduces with the increase of the shear rate. This behavior is typically found in polymers and their associations due to molecule entanglements observed at low shear rates. With the increase in shear rate, these entanglements are destroyed, and a reduction in viscosity is observed.

Many models have represented the flow behavior of non-Newtonian fluids, and the Power Law model satisfactorily fitted experimental data obtained by flow measurements ($R^2 > 0.9935$). For this model, the mathematical interpretation of the relationship between the shear stress and the shear rate is described by Equation (4):

$$\sigma = K\dot{\gamma}^n, \tag{4}$$

where K is the flow consistency index, σ is the shear stress, $\dot{\gamma}$ is the shear rate, and n is the flow behavior index (dimensionless). The flow curves for the blends are presented in the Figures S3 and S4. The parameters obtained by power law fitted curves are detailed in Table 2.

The K parameter is directly related to the viscosity characteristic of the samples. Values decrease with plasticizer addition for all blends. It is evident that the reduction in viscosity values is related to changes in entanglement networks due to the plasticizers action. This occurs as plasticizers can form strong bonding

TABLE 1 Gelation temperature of the samples.

Sample	Temperature (°C)
CSge	–
CSgeEG	43.7
CSgeGL	48.0
CSgeSO	52.8
CSox	52.2
CSoxEG	52.8
CSoxGL	56.7
CSoxSO	58.1

TABLE 2 Fitted parameters to power law model.

Blends	K (s)	n	r^2
CSge	4.25	0.45	0.9962
CSgeEG	2.91	0.49	0.9935
CSgeGL	1.77	0.53	0.9951
CSgeSO	3.32	0.48	0.9960
CSox	1.66	0.59	0.9946
CSoxEG	1.39	0.55	0.9943
CSoxGL	1.41	0.55	0.9939
CSoxSO	1.50	0.54	0.9935

interactions with starch and chitosan and weaken the interaction between the biopolymers, facilitating chain movement.

Depending on the plasticizer type, changes in the polymer–polymer associations are noticed when gelatinized starch is used in the sample preparation. For chitosan/oxidized rice starch film-forming solutions, the difference in K values after plasticizer addition was less evident, suggesting the weak influence of polyols in the polymer–polymer system. Additionally, the oxidation of starch molecules influences their chain interaction with chitosan. Indeed, CSge showed a higher value of K (4.25 s) than CSox (1.66 s). This low value can be attributed to an increase in the alignment of oxidized rice starch and chitosan molecules due to a higher interaction, favoring the flow of this blend.

The film-forming solution with small viscosity is commonly related to a high spreadability property,[28] facilitating the processability of the blend. That makes the addition of a plasticizer advantageous in the film-forming solution.

The n values were lower than 1, indicating the shear-thinning (pseudoplastic) behavior, as Newtonian fluids have n equal to 1. The CSge showed the lower n value and polyol addition promotes an increase in pseudoplasticity, probably to a decrease in inter and intramolecular interactions of chitosan and starch in the blends.

On the other hand, CSox displayed the highest flow index, a behavior attributed to an increase in interactions between polymers (amino groups of chitosan and aldehyde groups of oxidized starch). The n values decreased when polyol was incorporated. However, no remarkable differences were observed between CSoxEG, CSoxGL, and CSoxSO film-forming solutions. It suggested the weak influence of polyols in the polymer–polymer system when oxidized rice starch was used in sample preparation.

3.3 | Film characterization

All films were homogeneous, transparent, thin, flexible, and easily removed from the casting plate. Chitosan/oxidized rice starch films were slightly yellow in appearance (Figure S5).

3.3.1 | Thermal analysis

Thermogravimetric analysis is a simple and accurate method for studying the decomposition pattern and the thermal stability of polymers. Thermogravimetric curves (Figures S6 and S7) characterize the thermal decomposition in synthetic air of chitosan/rice starch films. The weight loss occurs in three stages: the first refers to the loss of structural water (25–200°C); the second (200–400°C) is a complex process involving the dehydration of the saccharide rings, depolymerization, and decomposition of the acetylated and deacetylated polymer units;[29] and the third stage (400–750°C) corresponds to the carbonization of polymeric materials. The close similarity of TG curves for films prepared with gelatinized starch indicated that the thermal decomposition processes have a similar pattern.

For chitosan/oxidized starch films, the same weight loss events were observed: one stage referring to the dehydration and two stages related to the decomposition. Starch oxidation increases the amount of water observed in the first stage (Table 3). This is related to the fact that the dialdehyde groups are more hydrophilic than native starch, as already observed by Yu et al.[30]

Table 3 reports the T_{onset} point, which corresponds to the initial decomposition temperature of the films. Starch oxidation decreases the decomposition temperature from 266.3°C (CSge) to 204.8°C (CSox). This behavior occurs due to the breaking of the starch rings during the oxidation reaction, thus reducing their thermal stability.[31] In addition, the presence of polyols decreases T_{onset} values for both forms of starch. Polyol addition reduces the attractive intermolecular forces of polymer chains, reducing the energy required for degradation and consequently decreasing the decomposition temperature.[21] For films prepared with gelatinized starch, there seems to be a relation between the number of OH groups in the plasticizer molecules and degradation stability, as CSgeSO exhibited the lowest value (250.7°C).

A decrease in degradation temperature was observed for the chitosan/oxidized starch films. Nevertheless, the plasticizer type showed no effect on the T_{onset} values (CSoxEG and CSoxSO) or a slight decrease for CSoxGL.

DSC curves of chitosan/gelatinized films and chitosan/oxidized films (Figures S8 and S9) exhibit an endothermic peak (around 100°C) attributed to the water evaporation that was not removed completely when the films were dried. Polysaccharides exhibit a strong affinity to water, and due to the arrangements that macromolecule chains assume in the solid state, their structures can be easily hydrated. Depending on how ionic or hydrogen interactions exist and how strong they are among the components, distinct peak areas and position of peak temperature of endotherm thermal events are observed. Indeed, these differences can be associated with the water-holding capacity of polysaccharides associations[32] or in the presence of an additive, like plasticizers. This behavior is frequently observed in many polysaccharides, such as cellulose and chitin derivatives.[33–35]

TABLE 3 % Weight loss and T_{onset} values of chitosan/rice starch (gelatinized or oxidized) films.

Film	1 stage (25–200°C)	2 stage (200–400°C)	3 stage (400–750°C)	T_{onset} (°C)
CSge	11.1	49.3	37.5	266.3
CSgeEG	9.4	49.6	38.4	264.1
CSgeGL	9.6	54.5	34.2	261.5
CSgeSO	11.6	56.1	31.7	250.7
CSox	16.0	42.3	40.3	204.8
CSoxEG	13.2	44.5	40.6	192.6
CSoxGL	16.8	44.9	36.9	177.0
CSoxSO	14.8	45.7	38.9	192.4

TABLE 4 Temperature of endothermic peak and enthalpy values of chitosan/rice starch films.

Films	Temperature (°C)	ΔH (kJ mol^{-1})
CSge	105.7	314.4
CSgeEG	109.2	270.6
CSgeGL	110.1	224.1
CSgeSO	120.4	194.2
CSox	110.7	407.1
CSoxEG	110.3	311.6
CSoxGL	116.0	396.4
CSoxSO	109.7	265.9

The temperature values of the endothermic peaks obtained by DSC are listed in Table 4.

The starch oxidation process shifted the temperature of the endothermic peak from 105.7°C (CSge) to 110.7°C (CSox) and increased the ΔH value, which is related to the heat required to remove the water from the films. This result indicates an increase in water when oxidized starch was used, which is coherent with the thermogravimetric results. Supare et al.[36] described that an increase in chitosan-starch interactions shifts the endothermic peak to a high temperature.

The effect of the plasticizer type on chitosan/gelatinized starch is related to an increase in the temperature of the endothermic peak, confirming the role of hydroxyl groups of polyols in the polymer-polymer chain associations. Nevertheless, the enthalpy involved in this thermal event is lower than CSge, suggesting that the water molecules are connected less strongly, and less energy is required to vaporize the water in films.

Starch oxidation also influenced both the endothermic peak and ΔH values of plasticized films. Nevertheless, the observed temperature is compared to each other, confirming the behavior noticed in the rheological measurements and the weak effect of the plasticizers due to a high interaction between both polysaccharides.

3.3.2 | Water vapor permeability

Both biodegradability and good gas barrier properties are the main features of polysaccharide-based films. Plasticizers, such as polyols, are often added to modify the mechanical properties of films, and these may cause significant changes to the barrier properties (e.g., WVP).[16]

Figure 5 shows the influence of the different studied formulations on the WVP values for all films. The values are also listed in Table 5. WVP values of chitosan films prepared with gelatinized starch showed higher WVP values (7.82×10^{-8} g m^{-1} s^{-1} Pa^{-1}), while those with oxidized starch showed lower WVP values (1.23×10^{-8} g m^{-1} s^{-1} Pa^{-1}). This change in values is related to increased interactions between chitosan and oxidized rice starch, hindering the diffusion of water molecules through the film. A similar result was described by Pranoto et al.,[37] wherein an increase in ionic interactions between gellan and gelation reduced the WVP of the films.

The increased molecular interaction resulted in films with compact structures, and low WVP values was detected by adding plasticizers in chitosan/gelatinized starch. Notably, the WVP decreased with the addition of polyols with high hygroscopic characteristics, and there was a sharp reduction of around 77% in the film containing sorbitol (CSgeSO). This behavior is closely related to improving the interaction between OH groups of polyols and NH_2 and OH from the polysaccharides. An increase in interactions restricts the mobility of polysaccharide chains, which leads to a long and tortuous path for water molecules through the film, reducing the diffusion and, consequently, the WVP.[38]

CSox showed the lowest WVP value due to an increase in chitosan-starch interactions that reduce the diffusion of water molecules through the film. The addition of polyols as plasticizers agents promoted an increase in the values due to the higher interaction between the polyol hydroxyl groups and water molecules. In a study
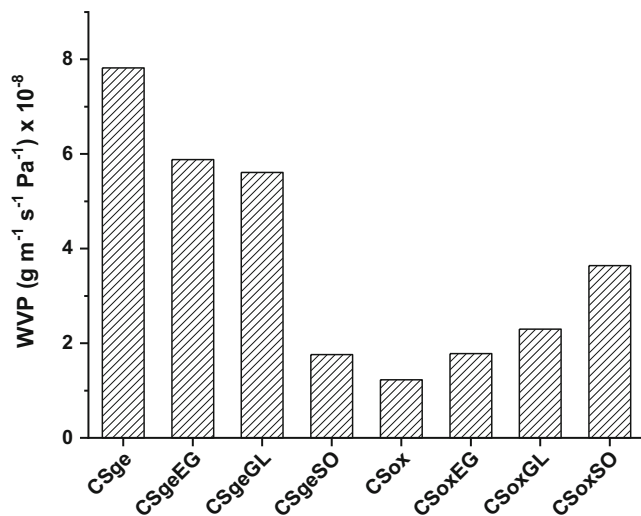
FIGURE 5 Effect of different polyols on the water vapor permeability (WVP) values of chitosan/rice starch films.

TABLE 5 Thickness, solubility, and WVP values of chitosan/rice starch films.

Films	Solubility (%)	Thickness (mm)
CSge	16.15 ± 0.34	0.059 ± 0.003
CSgeEG	41.15 ± 0.82	0.047 ± 0.002
CSgeGL	51.98 ± 0.16	0.051 ± 0.003
CSgeSO	32.88 ± 1.91	0.051 ± 0.001
CSox	10.13 ± 1.10	0.217 ± 0.004
CSoxEG	21.96 ± 0.60	0.108 ± 0.002
CSoxGL	40.43 ± 0.43	0.117 ± 0.002
CSoxSO	15.75 ± 0.71	0.135 ± 0.002

comparing SO and GL as plasticizers for starch/gelatin films, Al-Hassan and Norziah[39] found high WVP values when SO was used in the composition of the film.

Low WVP values suggest these films can be used particularly in highly humid conditions. Thus, the properties of films prepared with chitosan and starch may vary, depending on the starch form (gelatinized or oxidized) and the presence and structure of the plasticizer.

3.3.3 | Thickness and solubility

Table 5 shows the thickness and solubility values for all films at 25°C. Thickness was influenced by polyol addition since lower values were observed in the presence of the plasticizer. Moreover, the starch oxidation process increased the thickness values from 0.059 mm (CSge) to 0.217 mm (CSox), indicating that the reduction of direct associations between chitosan and starch restrains the gel retraction and consequently increases film thickness.

The thickness for chitosan/gelatinized starch was comparable, ranging from 0.059 mm (CSge) to 0.047 mm (CSgeEG). On the other hand, chitosan/oxidized starch films exhibited a broad range of values.

In general, due to polymer chain hindrance, an extension in the route for water molecules occurs, reflecting an improvement in the water resistance in films.[40] The solubility values of the films can indicate integrity in an aqueous system since higher solubility is due to lower resistance to dissolution in water.[41] The solubility values of the films can indicate integrity in an aqueous system.

Solubility was reduced by 40% in CSox compared to the film prepared with the unmodified starch form

(CSge). It is apparent that the presence of plasticizers controls solubility. Films prepared with both types of starch (gelatinized or oxidized) showed a rise in the values. Nevertheless, solubility values were found for chitosan/gelatinized rice starch films. In general, the polyols exhibited a lower influence in the chitosan/oxidized starch, probably due to the weak interaction of the plasticizers in the polymer–polymer association.

Film solubility can be modified by changes to the starch form and plasticizer type, allowing a wide range of possible applications. In some applications, films with low solubility that maintain structural integrity and with higher water resistance are required. However, in other cases, higher solubility is necessary, and solubility in water might be beneficial.[41]

3.3.4 | Dynamic mechanical analysis

Tensile strength is defined as the maximum tensile stress that a film can sustain. Elongation at break is the maximum change in length of the test specimen before breaking.[42] Figure 6 shows the effect of starch oxidation on elongation at the break and tension at the break for all films tested. The values are listed in the Table S1.

The tensile strength (TS) of CSge was 45.42 MPa, showing that the chitosan/starch mixture produces a brittle material that must be handled carefully. As films were produced by casting, the higher tensile strength of CSge was attributed to a high formation of intermolecular hydrogen bonds between NH_3^+ of chitosan backbone and OH^- of gelatinized rice starch. The amino groups (NH_2) of chitosan were protonated to NH_3^+ due to dissolution in acetic acid solution. In contrast, the gelatinization process destroyed the ordered crystalline structure of starch, exposing OH^- groups that readily form hydrogen bonds with the protonated amino group. For the CSox film, electrostatic interaction occurs within aldehyde

FIGURE 6 Elongation at the break (dense filling) and tensile strength (sparse filling) for chitosan/starch films.
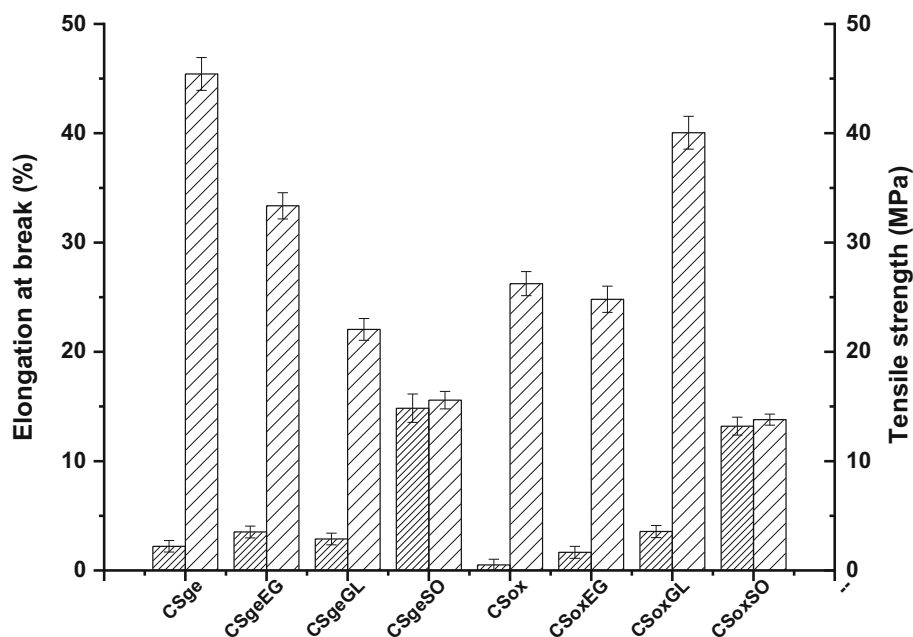
groups of oxidized starch and chitosan-protonated amino groups. This higher interaction between NH_3^+ and OH^- or the dialdehyde group of starch is responsible for a reduction in the brightness of the material.

Plasticizer insertion was generally associated with decreased tensile strength behavior and increased elongation at break. Based on these results, there was a reduction in the intermolecular forces between the chains of adjacent macromolecules, increasing the free volume and reducing mechanical resistance.[43] Thus, the decrease in tensile strength with the addition of polyols is due to a decrease in intermolecular interaction between chitosan and starch.

A decrease in tensile strength in chitosan/gelatinized starch films was noticed after polyol was incorporated into the network, thanks to reduced direct interactions between chitosan and starch chains. Therefore, the ease of chain movement decreases the tensile forces. In addition, the oxidation process decreases the tensile strength values in CSox compared to CSge, indicating a loss of resistance in the films when oxidized starch was used for film preparation.

The values of elongation at break were affected by the addition of polyol as plasticizers enhance film flexibility due to the interference in polymer–polymer associations and reduce the rigidity of the network, increasing the capacity for polymer chain movements.[44]

Changes in elongation at break were observed when oxidized starch was used to prepare the films, with values of 2.21% and 0.51% for CSge and CSox, respectively. It is possible to conclude that film elasticity decreases when oxidized starch is used.

Nevertheless, the SO seems to increase elongation at break values, independently of the starch form used to produce the films. The values found were 14.84% for CSgeSO and 13.20% for CSoxSO, showing that this polyol highly affects the elasticity property, probably due to its structure with six OH groups. Similar behavior was previously observed in starch/gelatin films.[39]

4 | CONCLUSION

Starch was oxidized to produce aldehyde groups in the polysaccharide chain. The chemical modification was compared with the traditional gelatinization process conducted in starch. Additionally, the insertion of different plasticizer types was evaluated in this study. It seems distinct interactions are involved between the components in the range of formulations studied. An increase in polysaccharide interaction between chitosan and oxidized starch was observed. As detected by rheological measurements, polyols could not interfere with these polymer–polymer associations. On the other hand, in chitosan/gelatinized blends, adding polyols induced an entanglement density between the components, mainly driven by the OH groups. The oxidation of starch changes the properties of chitosan/starch films, decreasing the WVP values due to the higher interactions between the amino groups of chitosan and dialdehyde groups of oxidized starch. In addition, the chemical modification to the starch structure influences the amount of water in the films. Stress–strain results showed a decrease in percentage elongation at break when oxidized starch was

used for preparing the films, reducing their flexibility and handling. On the other hand, adding plasticizers improves mechanical properties, especially elongation at break, resulting in increased film flexibility.

AUTHOR CONTRIBUTIONS

Conceptualization, methodology: Marilia M. Horn and Virginia C. Amaro Martins. *Formal analysis, investigation, writing—original draft*: Marilia M. Horn. *Resources, supervision*: Ana Maria de Guzzi Plepis. *Original draft and review and editing*: Virginia C. Amaro Martins and Ana Maria de Guzzi Plepis. All authors read and approved the final manuscript.

CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest concerning this study.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available in the Supporting Information of this article.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.