Available online at www.sciencedirect.com

ScienceDirect

Carbohydrate Polymers 70 (2007) 265–273

Carbohydrate Polymers

www.elsevier.com/locate/carbpol

Influence of storage time at room temperature on the physicochemical properties of cassava starch films

Lucía Famá [a,1], Silvia Goyanes [a,2], Lía Gerschenson [b,*,2]

[a] *Departamento de Física, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Ciudad Universitaria, (1428) Buenos Aires, Argentina*

[b] *Departamento de Industrias, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Ciudad Universitaria, (1428) Buenos Aires, Argentina*

Received 23 September 2006; received in revised form 3 March 2007; accepted 4 April 2007
Available online 12 April 2007

Abstract

The influence of storage time on colour, moisture content, crystallinity and mechanical performance of edible cassava starch films containing glycerol and sorbate, were studied. The effect of the pH of the film forming systems was also evaluated.

It was observed that, for storage times of 4 weeks or longer, tan δ curves shifted to higher temperatures independently of the pH. Films obtained from systems of pH 5 showed a decrease in the intensity of the loss tangent-peak observed between -30 and 10 °C, fact that is in accordance with a slight increase in the crystalline fraction. At room temperature, E' increased and tan δ decreased between 2 and 4 weeks trends that are in accordance with the increase in crystallinity and the decrease in moisture content observed with storage time.

Colour parameters and sorbate content did not change significantly along 8 week-storage, showing that sorbate was not destroyed along the period studied.

It can be concluded that changes of starch along storage were mostly responsible for the changes observed in mechanical properties.

Keywords: Edible starch films; Storage; Physicochemical properties

1. Introduction

Research on edible films and coatings in foods is partly driven by industry due to the high demand of consumers for longer shelf-life and better quality of foods as well as of environmentally friendly materials. These biodegradable polymeric films offer alternative packaging options with no contribution to the environmental pollution while being obtained from renewable sources with low cost (Lu, Tighzert, Berzin, & Rondot, 2005; Tharanathan, 2003).

Cellulose, gums, starches and proteins are the basic materials for edible film and plasticizers such as glycerol or sorbitol are commonly included in their formulation, to improve film flexibility and strength (Jongjareonrak, Benjakul, Visessanguan, Prodpran, & Tanaka, 2006; Sobral, do, dos Santos, & García, 2005).

Starch is a commonly used agricultural raw material. It is a renewable resource, inexpensive and widely available (Lourdin, Valle, & Colonna, 1995) and it is capable of edible film formation. Recently, Veiga-Santos, Suzuky, Cereda, and Scamparini (2005) remarked that starch-based films have low mechanical resistance and high moisture sensitivity when compared to traditional petroleum plastic films. Recent research has demonstrated that several polysaccharide based films and coatings have good mechanical properties (Famá, Flores, Gerschenson, & Goyanes, 2006; Hsu, 2002; Le Tien et al., 2000).

---

[*] Corresponding author. Tel.: +54 11 45763300x336; fax: +54 11 45763366.
E-mail address: lia@di.fcen.uba.ar (L. Gerschenson).

[1] Fellow of Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET).

[2] Member of Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET).

0144-8617/$ - see front matter © 2007 Elsevier Ltd. All rights reserved.
doi:10.1016/j.carbpol.2007.04.003

Cassava is produced in Latin America and its importance as a source of starch is growing rapidly, especially because of its low price in the world market when compared to starches from other sources (FAO, 2004). The potential use of cassava starch as a matrix for the development of edible films is a topic that has been previously considered (Famá, Rojas, Goyanes, & Gerschenson, 2005).

In the last years, research has been performed concerning the use of edible films for surface application or slow release of different antimicrobials (Buonocore et al., 2003; Chung, Chikindas, & Yan, 2001; Franssen, Rumsey, & Krochta, 2004; Park, Daeschel, & Zhao, 2004). Sorbic acid and its potassium salt (sorbates) are considered GRAS additives and are active against yeast, molds and many bacteria the effects being greater at low pH (Sofos, 1989). Addition of sorbates to edible films has been proposed as a way of minimizing surface microbial contamination (Cagri, Ustunoi, & Ryser, 2001; Chen, Yeh, & Chiang, 1996; Flores, Haedo, Campos, & Gerschenson, 2006; Han & Floros, 1997).

In the case of starch-based films, physical and chemical changes as a result of ageing (Delville, Joly, Dole, & Bliard, 2003) are of great importance because they can affect their functionality as a consequence of starch molecules re-association into crystalline segments (retrogradation). The rate and extent to which this occurs depends on factors such as chain length, concentration of starch, system pH and composition as well as time and temperature.

Different researchers studied the effect of various polyols on physical properties of starch-based films (Talja, Helén, Roos, & Jouppila, 2007), the combined effect of plasticizers and surfactants on physical properties of starch-based films (Rodríguez, Osés, Ziani, & Maté, 2006); the effect of starch-based films containing glycerol on food shelf life (García, Martino, & Zaritzky, 1998; Viña et al., 2006). Some information is also available concerning the change of physical properties of plasticized films based on starch with storage (Famá et al., 2005; Mali, Grossman, Garcia, Martino, & Zaritzky, 2006). More systematic information is needed concerning changes in the properties of these films on storage due to starch changes and/or to its interaction with other components of the film with the object of understanding the influence of those changes on film functionality.

In this research, the influence of storage time on physicochemical properties of edible cassava starch films containing glycerol and sorbate was studied along 8 weeks of storage at 25 °C, under controlled relative humidity. Changes observed in mechanical and thermochemical properties were studied through DMTA and DSC at low temperatures. It was also explored the effect of pH of film forming system on those properties.

2. Materials and methods

2.1. Materials, film obtention and storage

Cassava starch was provided by Industrias del Maíz S.A. (Argentina). Glycerol, citric acid (Mallickrodt, Argentina) and potassium sorbate (Sigma, St. Louis, Missouri) used were of analytical grade.

Edible films were constituted from aqueous suspensions containing 5.0 g/100 g cassava-starch, 2.5 g/100 g glycerol, 0.2 g/100 g potassium sorbate and 92.3 g/100 g of water. The suspension pH was 6.7. To adjust the pH of 100 g of suspension to a value of 5.0, 0.25 ml of citric acid aqueous solution (50 g/100 g solution) was used.

The preparation method involved a gelatinization process under constant agitation followed by drying step as described by Famá et al. (2006).

Sample thickness was measured to the nearest 0.01 mm, using an optical microscope (Nikon AFX II, Japan) at three different locations in each specimen. Average values were calculated and are reported.

Films obtained were stored for 8 weeks, at 25 °C, over a saturated solution of NaBr (water activity, $a_W \cong 0.575$).

Samples were obtained at 2, 4 and 8 weeks of storage for their characterization.

2.2. Methods for film characterization

2.2.1. Potassium sorbate dosage

Potassium sorbate content was determined through the oxidation technique proposed by the AOAC (1990) which involves distillation and a colourimetric reaction using thiobarbituric acid. Determinations were performed in duplicate and the average is reported.

2.2.2. Moisture determination

Samples ($\sim$0.5 g) were dried over calcium chloride, in a vacuum convection oven at 70 °C, until constant weight. The reported results represent the average of, at least, three samples.

2.2.3. Colour

Measurements were carried out with a Minolta CM-508d colourimeter (Minolta, Kyoto, Japan) employing the Hunter and CIE scale.

The films to be tested were cut into circles of 2 cm diameter (sample) for these tests. Colour differences (ΔC) and yellow index (YI) were evaluated in three films for each time and pH studied and each sample was evaluated in, at least, 10 positions randomly selected. The average is reported.

Colour difference is the magnitude of the resultant vector of three component differences: lightness difference, ΔL; red-green chromaticity difference, Δa; and yellow-blue chromaticity difference, Δb (Valencia Rodríguez, 2001). Colour differences were calculated as:

$$\Delta C = \sqrt{\Delta a^2 + \Delta b^2 + \Delta L^2}$$

where L, represents lightness; a, redness; b, yellowness; and $\Delta a = a_i - a_0$, $\Delta b = b_i - b_0$ and $\Delta L = L_i - L_0$. The index i, indicates the values observed for each storage time (2, 4 and 8 weeks) and index 0, indicates the references used.

In our case, we used the colour parameters of the white standard ($L = 94.8$, $a = -0.78$, $b = 1.43$) as the reference (Sobral et al., 2005).

Yellow index (YI) parameter evaluates the degree of yellowness for certain nearly colourless transparent plastics. It was calculated according to the ASTM D1925 (1988) method:

$$\mathrm{YI} = \frac{100}{Y}\ (1,28X - 1,06Z)$$

using the CIE (Commission International de l'Eclairage) values. X is the tristimulus value for red, Y is the tristimulus value for green and Z is the tristimulus value for blue.

2.2.4. X-ray diffraction analyses

Diffraction of samples was recorded using a Philips X-ray diffractometer with vertical goniometer operating at 40 kV and 30 mA (Cu Kα radiation $\lambda = 1.542$ Å). Samples mounted on a glass plate and conditioned at a water activity (a_W) of 0.575 and 25 °C were attached to the equipment holder. X-ray intensity was determined with a scintillation counter in a scattering angle range from 3° to 33° (2θ) with a scanning speed of 1°/min (Biliaredis, Lazaridou, & Arvanitoyannis, 1999; Famá et al., 2005; Valle, Buleon, Carreau, Lavoie, & Vergnes, 1998). Distances between the planes of the crystals d(Å) were calculated from the diffraction angles (°) obtained in the X-ray pattern, according to Bragg's law (Kittel, 1986).

From the scattering spectrum, the effective percent crystallinity of films was determined, according to Hermans and Weidinger (1961), as the ratio of the integrated crystalline intensity to the total intensity. Crystalline fraction was estimated by the area above the smooth curve drawn on the basis of the main peaks (main d-spacing) (García, Martino, & Zaritzky, 2000; Koksel, Ahbaz, & Ozboy, 1983; Snyder & Bish, 1989).

2.2.5. Mechanical properties

Mechanical measurements were performed using a Dynamic Mechanical Thermal Analyzer (DMTA IV) Rheometric Scientific equipment (Rheometric Scientific Inc., New Jersey, USA) in the rectangular tension mode. Two types of test were carried out for characterizing the viscoelastic properties of the material: dynamic and quasi-static tests.

Dynamic tests were performed at 1 Hz, in the temperature range from −85 to 20 °C, at a heating rate of 2 °C/min. The samples were subjected to a cyclic strain of 0.04%. The strain values used were sufficiently small to assure that the mechanical response of the specimen was within the linear viscoelastic range (Brostow & Corneliussen, 1986; Famá, Rojas, Goyanes, & Gerschenson, 2003). The dependence with the temperature of E' and $\tan\delta$ were recorded for each storage time and pH condition. The loss tangent curves were used to evaluate the relaxation processes and the glass transition temperature was taken, as the maximum of the $\tan\delta$ peak (Chartoff, 1981).

Quasi-static tests were performed at room temperature employing a strain rate fixed at a value of $5 \times 10^{-3}\ \mathrm{s}^{-1}$. From the quasi-static test information was obtained about the effect of storage time and pH on the $\sigma - \varepsilon$ curves.

In both cases the samples were cut according to ASTM D4092 (2001), considering the highest elongation allowed by the Dynamic Mechanical Thermal Analyzer used. Sample dimensions were 15.0 mm long, 5.1 mm wide and 0.35 mm thick. To avoid an excessive deformation in compression of the head of the sample, while maintaining a firm holding, a constrainer of that deformation was placed in the contact area of the grips (Famá et al., 2005).

Microscopic examination of specimens to be tested was performed and pieces, where flaws were detected, were discarded.

Determination of dynamic parameters was performed in triplicate. Results reported from quasi-static tests are the average of, at least, seven samples.

2.2.6. Differential scanning calorimetry (DSC)

A differential scanning calorimeter (Mettler Toledo Schwerzenbach) was used to determined changes in enthalpy (ΔH) and glass transitions (T_g) of the films. Indium and zinc were used to calibrate temperature and heat flux. Ten milligrams of samples contained in hermetically sealed aluminium pans was heated twice, in the temperature range −100 to 20 °C, at a scanning rate of 10 °C/min, under a nitrogen atmosphere. Changes of phase or state and the corresponding, enthalpy were determined from the second heating thermogram. Glass transition was assigned to the middle temperature of the relaxation range (Biliaredis et al., 1999; Chartoff, 1981).

Results obtained through DSC were used to understand trends observed in the DMTA studies.

2.2.7. Mathematical data treatment and statistical analyses

A two-factor (storage time and pH) experiment was performed. Three levels of time (2, 4 and 8 weeks) and two levels of pH (5.0 and 6.7) were studied. Data were analyzed through a two-way ANOVA with a probability level, α, of 0.05. A Tukey test was applied to test which treatments rendered differences between samples. Results are reported on the basis of their average and confidence interval (α: 0.05; Sokal & Rohlf, 1969).

Statgraphics Plus for Windows, version 3.0, 1997 (Manugistics Inc., Rockville, Maryland, USA) was used for data treatment and statistical analysis.

3. Results and discussion

The potassium sorbate concentration, moisture content, crystalline fraction, colour parameters and stress at 70% of strain for films obtained from systems prepared at pH 5.0 or 6.7 and after 2, 4 and 8 weeks of film storage, are shown in Table 1. Ageing had no significant effect (α: 0.05) on the sorbate content of the films. These results indicate that the

Table 1
Physicochemical characteristics of cassava starch biofilms

Storage time		2 weeks		4 weeks		8 weeks	
pH		6.7	5.0	6.7	5.0	6.7	5.0
Sorbate content (g/100 g film, dry basis)		2.19 ± 0.27	1.95 ± 0.15	2.0 ± 0.2	1.8 ± 0.1	2.2 ± 0.2	2.0 ± 0.2
Moisture (g/100 g film, dry basis)		36 ± 2	33 ± 4	22.4 ± 1.3	21.5 ± 1.5	24.8 ± 6.2	22.5 ± 1.5
Crystalline fraction (%)		22.2 ± 1.1	25.6 ± 1.3	26.2 ± 1.3	28.5 ± 1.4	30.3 ± 1.5	31.6 ± 1.6
Colour parameters	ΔC	11.6 ± 0.2	10.3 ± 0.5	12.0 ± 0.1	9.4 ± 0.1	12.9 ± 0.1	9.8 ± 0.1
	YI	11.8 ± 0.8	9.8 ± 1.6	12.2 ± 0.5	10.5 ± 0.8	10.5 ± 1.2	10.5 ± 0.5
σ (MPa) at $\varepsilon = 71\%$		1.19 ± 0.06	1.20 ± 0.04	2.0 ± 0.1	1.9 ± 0.1	2.8 ± 0.2	2.6 ± 0.2

Data are informed as $x \pm \varepsilon$, being x the average and $\varepsilon = t_{n-1}\, s/n^{1/2}$, where t is the Student parameter (α: 0.05); s is the standard deviation and n is the number of samples assayed (three for moisture content, two for sorbate content and crystalline fraction, six for colour parameters and, at least, seven for stress).

sorbate was not oxidized or degraded, at least, after 8 weeks of storage, independent of the pH of the system.

Colour parameters, ΔC and YI did not change with storage time (Table 1). This trend was observed for both pHs studied, but slightly lower values were obtained for pH 5.0. This behaviour is consistent with the null change of sorbate on storage as it is well known that this antimicrobial has an important tendency to participate in browning reactions (Gerschenson & Campos, 1995) affecting the colour of food and also with the fact that the rate of browning reactions decreases with pH (Cheftel, Cheftel, & Besancon, 1983; Valencia Rodríguez, 2001).

Fig. 1 shows X-ray diffraction patterns for films at (i) 2, (ii) 4 and (iii) 8 weeks of storage time and at pH 6.7 (panel a) and 5.0 (panel b), respectively. The crystalline peaks, were analyzed in the interval from 12° to 25° (2θ), identifying the most intense peaks and calculating the distances between the planes of the crystals d(Å) from the diffraction angles (°C). It was observed that X-ray pattern showed a B–V structure (García et al., 2000; Manzocco, Nicoli, & Labuza, 2003; Zobel & Illinois, 1986) which diffracted in the following planes $d \cong 3.6$, 4.0, 4.5, 5.2 and 5.9 Å. These peaks are identified in the Figs. 1(a) and (b). In general, tuber starches show a B-type crystal structure. The introduction of complexing agents into starch preparations disrupts double helix conformations by forming stable single chain V-conformation helices. The V-conformation is a result of amylose being complexed with substances such as aliphatic fatty acids, surfactants, emulsifiers, n-alcohols, glycerol and dimethyl sulfoxide. When amylose and polar lipids are present, V-structures can result from gelatinization, both during heating and upon cooling (Zobel, 1994).

According to the data base ICPDS (1997), the peaks at 5.9, 5.2 and 3.6 Å coincide with amylose peaks of high intensity. The peak at 4.0 and 4.5 Å are associated with amylose peaks of middle intensity and with high intensity potassium sorbate peaks.

The intensity of the crystalline peaks increased for 4 weeks of storage for both pHs analyzed; for longer times of storage, increase was not significant (α: 0.05) as can be observed in Table 1. Hale and Bair (1981), reported the same behaviour in acrylonitrile–butadiene–styrene (ASB) polymer materials.

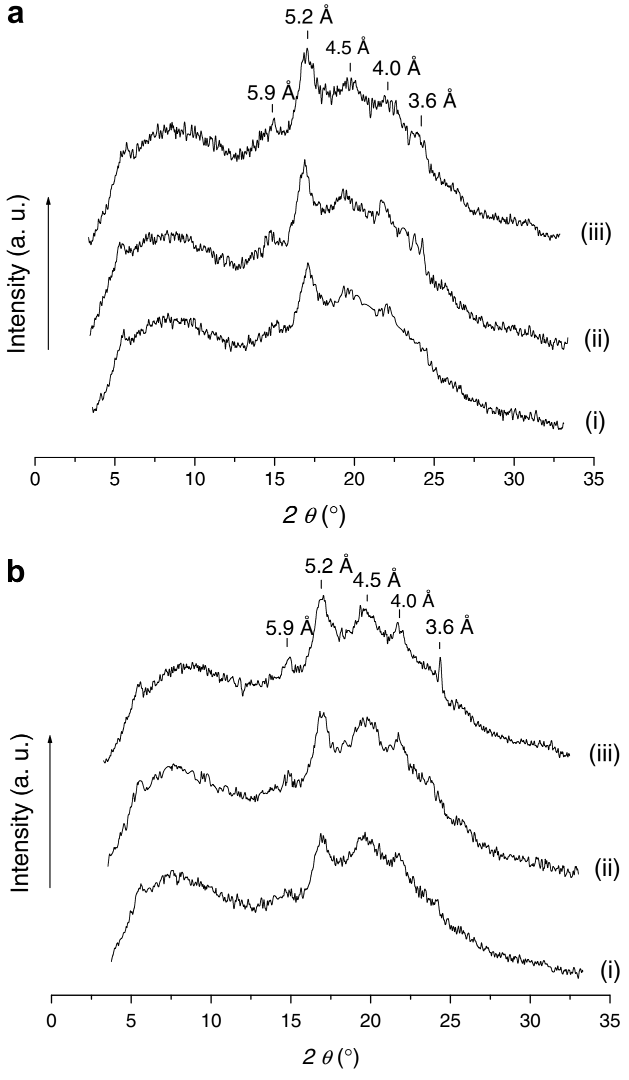

Fig. 1. X-ray diffraction pattern for films after storage times of 2 weeks (i), 4 weeks (ii) and 8 weeks (iii). (a) pH 6.7, (b) pH 5.0.

On ageing, starch molecules can reassociate into crystalline segments or "retrograde" (Aguilera & Stanley, 1999). Generally, during starch storage, amylose crystallisation occurs within hours. Then, a slower development of the crystallinity occurs that is attributed to amylopectin (Delville et al., 2003). Biliaredis et al. (1999) gave evidence of partial crystallisation of the starch component from films stored at an a_W above 0.64.

In the analysis of the results of this research, it was supposed that glycerol was stable for the storage time studied. This supposition is based in reports of Orliac, Rouilly, Silvestre, and Rigal (2003) for starch–glycerol films and of Gueguen, Viroben, Noireaux, and Subirade (1998) who studied pea protein–glycerol films.

It is interesting to remark that the decrease of the pH of the film forming system used from 6.7 to 5.0, produced a slight increase in the crystalline fraction of the film, at each time studied and non-significant decrease in its moisture content and this behaviour is in accordance to that reported by Dumoulin, Alex, Szabo, Cartilier, and Mateescu (1998) for dextrins. The influence of the pH in the crystalline fraction of films decreased with the storage time.

In Table 1, a significant decrease in the moisture content, between the second and the fourth week, is observed independent of the pH of the system. This can be ascribed to the increase in the crystalline fraction with the storage time as it is known that the crystalline phase of a semicrystalline material is highly linked to the decrease in its moisture content (Chang, Chea, & Seow, 2000). Between 4 and 8 weeks no further decrease in moisture content is observed.

3.1. Dynamic tests and differential scanning calorimetry

Figs. 2 and 3 show the loss tangent, tan δ (a), and storage modulus, E' (b), as a function of the temperature for the films obtained from aqueous systems of pH 6.7 and 5, respectively. In the tan δ curves (Figs. 2(a) and 3(a)), two relaxation peaks can be observed independent of storage time and pH: a very important one around −62 °C and other, wide and with low intensity, between −30 and 10 °C. The molecular relaxation around −62 °C is associated to a glycerol-rich phase (Famá et al., 2005; Wilhelm, Sierakowski, Souza, & Wypych, 2003). Wilhelm et al. (2003) reported that glycerol-native cara-root starch films presented two relaxation peaks, one at −74 °C and the other, at 188 °C. They attributed these peaks to two phases originated due to the partial miscibility of starch and glycerol: the relaxation at −74 °C was attributed to a glycerol-rich phase and the other, to an amylose-rich phase.

As can be observed in Figs. 2(a) and 3(a), the molecular relaxation associated with the glycerol-rich phase, shifted slightly to higher temperatures for 4 weeks of storage time, independently of pH. No differences were observed between 4 and 8 weeks. This fact could be attributed to the moisture decrease observed after 4 weeks of storage (see Table 1). Water is an important plasticizer and the effect of plasticizer incorporation to a system is the movement of their relaxations to lower temperatures (Chartoff, 1981; Goyanes, 2000). Therefore, a decrease in moisture content should produce a shift in the transition temperature to higher values. A similar effect is observed in the dependence of E' with temperature (Figs. 2(b) and 3(b)). The abrupt fall (~1 GPa) in the storage modulus in that region of temperatures is associated with the molecular relaxation related with glycerol-rich phase transition. Independently of the pH, the curves of samples with 4 and 8 weeks of storage suffered a shift to higher temperatures with respect to that observed after 2 weeks of storage.

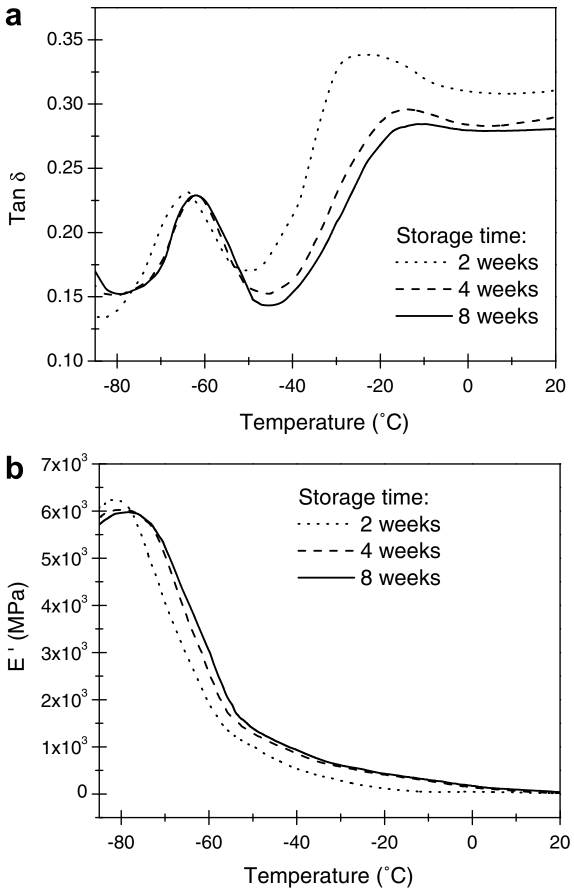

Fig. 2. Variation of the loss tangent, tan δ (a) and storage modulus, E' (b) with the temperature, for films of pH 6.7.

From Figs. 2(b) and 3(b) it can be obtained information about the intensity of the main relaxation, $\Delta E'$, defined as the difference between the E' values in the glassy zone and the E' value en the rubbery zone. This relaxation is associated with the amorphous part of the films. Then, a decrease in this relaxation should be related to an increase in the crystalline fraction. The pH seems not to affect the $\Delta E'$ value while the storage time exerted a slight effect on $\Delta E'$, being obtained the maximum value for 2 weeks of storage (6.2 ± 0.1 GPa at 2 weeks, and 5.9 ± 0.1 GPa at 8 week storage).

Fig. 4 shows the heating thermograms obtained for all films studied (pH 6.7, Fig. 4(a) and pH 5, Fig. 4(b)). In all cases, only one relaxation, around −76 °C, was observed. This is associated with the T_g of a glycerol-rich phase (Famá et al., 2005; Wilhelm et al., 2003). Besides, in the same figure, it can be seen that an increase in the storage time decreased the change in the enthalpy (ΔH) during the relaxation processes. This effect is not affected

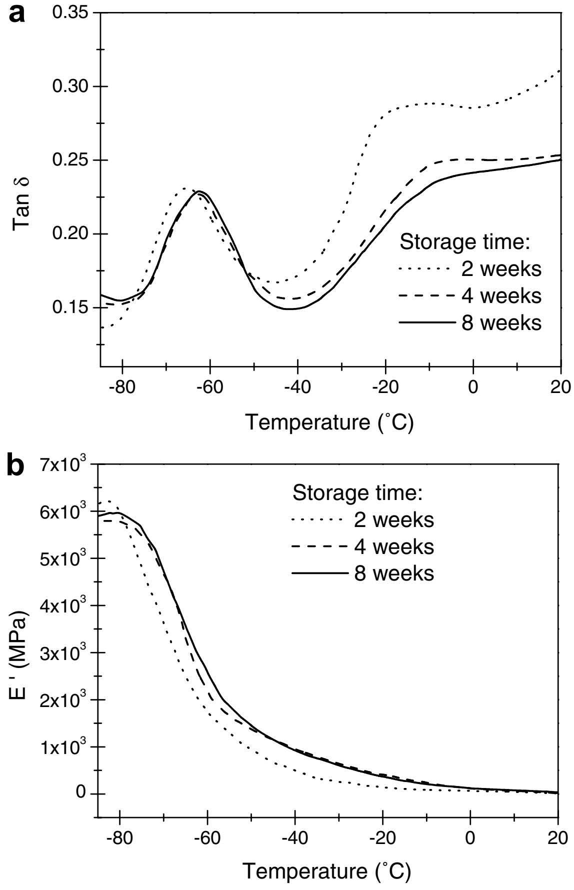

Fig. 3. Variation of the loss tangent, tan δ (a) and storage modulus, E' (b) with the temperature, for films of pH 5.

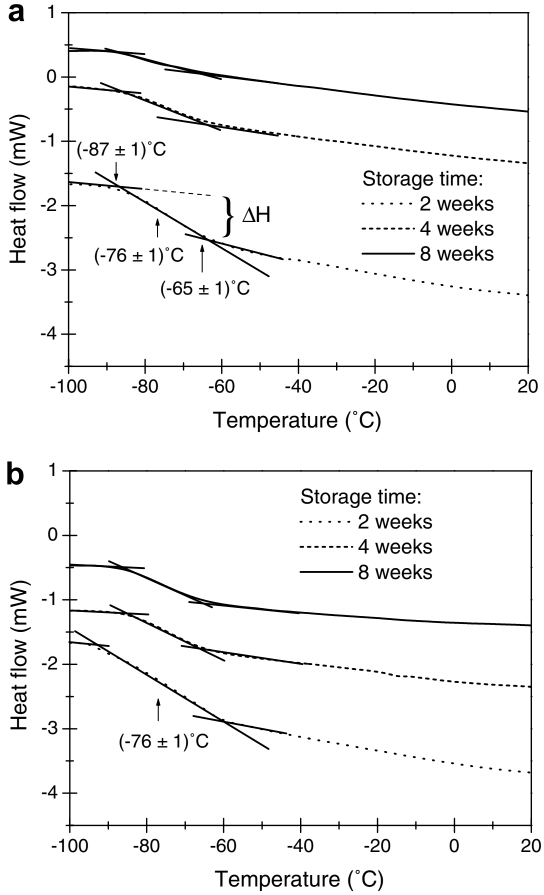

Fig. 4. Heating thermograms for different storage times. (a) pH 6.7, (b) pH 5.0.

by the pH value. It is well known that the polymer glass physical ageing process is generally accompanied by the decrease of enthalpy during the relaxation processes (Brostow & Corneliussen, 1986). In particular, Chung and Lim (2003, 2004) found in normal and waxy rice starch stored a similar behaviour for ΔH to that reported in this paper.

According to Bair (1981), it is possible to estimate from ΔH, the fraction of the material participating in a particular phase of a multicomponent system. Taking into account that the relaxation observed corresponds to an amorphous relaxation associated with the T_g of a glycerol-rich phase, the decrease in ΔH with the storage time is in accordance to the decrease observed in $\Delta E'$ (Figs. 2(b) and 3(b)).

In order to better analyze the relaxation processes observed between −30 and 10 °C in Figs. 2(a) and 3(a), a detail of the tan δ curve is shown in Figs. 5(a) and (b) for films of pH 6.7 and 5.0, respectively. There is a strong effect of the storage time on the loss tangent curves in this temperature range. The peak observed between −30 and 10 °C showed a shift to higher temperatures when the storage time increased from 2 to 4 weeks, but little increase between 4 and 8 weeks of storage. These results can be related to the crystalline fraction. To clarify this idea, the same Figure includes a bar plot of dependence of crystalline fraction with storage time for both pHs. It can be observed that the increase in the crystalline fraction with time led to a decrease in the peak intensity and to an increase in the temperature peak. Chartoff (1981) for polycarbonate showed that the tan δ peak was partially suppressed and shifted to higher temperatures with ageing time. Similar observations were reported by Hale and Bair (1981) for an ASB polymer. Comparing Figs. 5(a) and (b) it can be concluded that the peak observed in that range of temperature was less pronounced when the pH decreased. This trend might be ascribed to changes in the crystalline fraction; as previously reported, X-ray studies showed that the crystalline fraction was slightly greater for films at pH 5.0 than for the ones at pH 6.7.

Fig. 6 shows the dependence of E' and tan δ on the storage time for films at pH 6.7 and 5. These values were obtained from Figs. 2 and 3 at room temperature (20 °C). As can be seen, there is an increase in E' on storage, and a large decrease in tan δ between 2 and 4 weeks, with a smaller decrease up to 8 weeks. Those results are in accordance with the increase in crystallinity and decrease in moisture content with storage time, previously reported (Table 1). It is well known that an increment in water content leads to a decrease in Young modulus (Chartoff, 1981). Similar behaviour has been observed by Lens et al. (2003) for wheat gluten films. Forssell, Hulleman, Myllarinen,

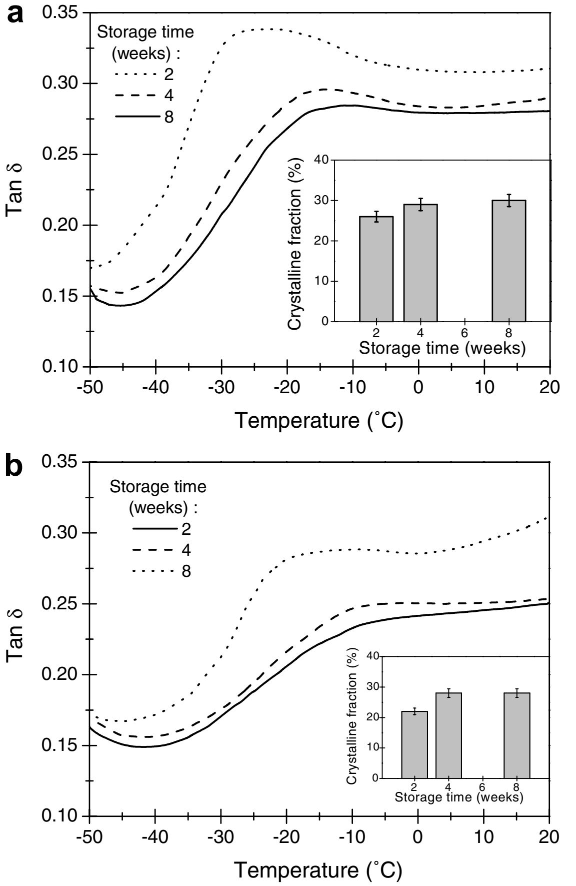

Fig. 5. Loss tangent change with temperature. (a) Films with pH 6.7. (b) Films with pH 5.0. It is also included a bar plot showing the effect of storage time on crystalline fraction.

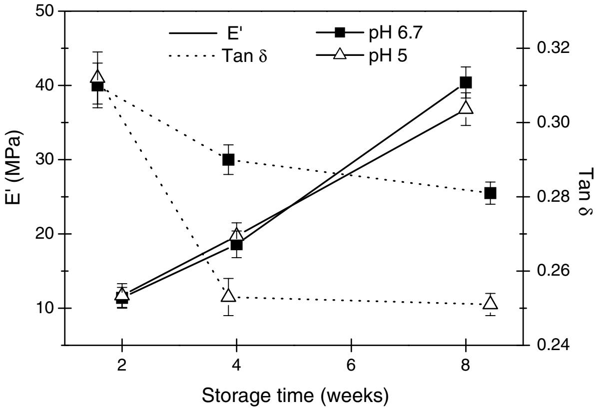

Fig. 6. Dependence with the storage time of storage modulus (E') and loss tangent ($\tan\delta$) at room temperature.

Moates, and Parker (1999) stated that the explanation for the increase in tensile modulus with storage time is the crystallization of starch components. The different pH for film forming systems did not change the values of E' for each time; however, $\tan\delta$ for films at pH 5 was significantly (α: 0.05) lower for 4 and 8 weeks when compared with the ones for films obtained from systems at pH 6.7.

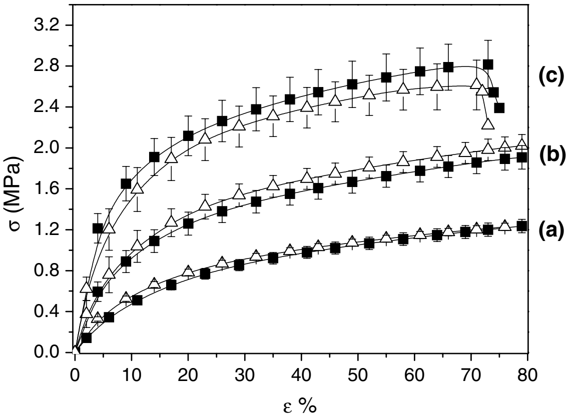

Fig. 7. Stress (σ) versus strain (ε) curves for films after storage times of 2 weeks (a), 4 weeks (b) and 8 weeks (c). (■) pH 6.7; (Δ) pH 5.0.

3.2. Quasi-static tests

Fig. 7 shows the influences of storage time on the stress (σ)–strain (ε) relationship for samples with pH 6.7 and 5. As can be observed, these films showed a small strain range where the stress increased linearly with the strain, then a non-linear behaviour started, without arriving to plastic deformation region in the strain range studied. Independently of the pH of film forming system, the curves reported in Fig. 7 show an increase of the hardening (defined as a bigger tension to achieve the same deformation) with the increment of the storage time. This effect is correlated with a smaller rupture deformation for the sample that had the longest period of storage, as can be seen in Fig. 7 (curve c). The changes in the hardening, shown in the curves $\sigma - \varepsilon$, are linked to an increase in the crystallinity and/or to the decrease in the moisture content with storage time, as was observed through X-ray studies and moisture content determination.

4. Conclusions

During the eighth week of storage at room temperature, it was observed that sorbate content and yellow index did not change. The null change in colour precludes the occurrence of undesired changes in food organoleptical characteristics and suggests that antimicrobial activity does not change.

Between the second and eighth weeks of storage, at room temperature, of edible cassava starch films containing glycerol as plasticizer and potassium sorbate as antimicrobial agent, the moisture content of the films decreased and crystallinity increased regardless of the pH of the film forming system. These changes were reflected in the mechanical properties, showing a slight shift in the $\tan\delta$ peak to higher temperatures when storage time increased from 2 to 4 weeks in the temperature range −30 to 10 °C. E' evaluated at 20 °C increased, approximately, 300% after eight weeks and $\tan\delta$ decreased more than 10% between 2 and 4 weeks. Only slight changes from there on were observed. Starch ageing was mostly responsible for the changes observed in the mechanical properties.

The depression of the pH of the aqueous systems from which the films were formed, resulted in a slight increment in film crystallinity but did not change the transition observed at around −62 °C; however, the peak between −30 and 10 °C was less intense as the pH decreased.

The stress at a certain deformation increased along storage for all films studied. Samples stored for 8 weeks showed the smallest rupture deformation (≅75%) but for shorter storage periods, films showed rupture deformations higher than 80%. It can be concluded that storage, in general, did not impair mechanical characteristics of the films.

Results permit to expect integrity and effectiveness of films when applied to foods.

References

Aguilera, J. M., & Stanley, D. W. (1999). *Microstructural principles of food processing and engineering* (3rd ed.). Gaithenburg, Maryland: A Chapman & Hall Food Science Book, Aspen Publishers, Inc.

AOAC (1990). Official methods of analysis (13th ed.). Washington, DC: Association of Official Analytical Chemists.

ASTM D1925 (1988). Standard Test Method for yellowness index of plastics. Philadelphia: American Society for Testing and Materials.

ASTM D4092 (2001). Plastics: Dynamic Mechanical Proper ties. Philadelphia: American Society for Testing and Materials.

Bair, H. E. (1981). In E. A. Turi (Ed.). *Thermal characterization of polymeric materials* (Vol. 2, p. C 10). USA: Academic Press.

Biliaredis, C. G., Lazaridou, A., & Arvanitoyannis, I. (1999). Glass transition and physical properties of polyol-plasticised pullulan-starch blends at low moisture. *Carbohydrate Polymers, 40*, 29–47.

Brostow, W., & Corneliussen, R. D. (1986). *Failure of plastics*. Munich, Vienna, New York: Hanser publishers, pp. C 3–4.

Buonocore, G. G., Nobile, M. A., Panizza, A., Bove, S., Battaglia, G., & Nicolais, L. (2003). Modeling the lysozyme release kinetics from antimicrobial films intended for food packageing applications. *Journal of Food Science, 68*(4), 1365–1370.

Cagri, A., Ustunoi, Z., & Ryser, E. T. (2001). Antimicrobial, mechanical, and moisture barrier properties of low pH whey protein-based edible films containing aminobenzoic or sorbic acids. *Journal of Food Science, 66*(6), 865–870.

Chang, P., Chea, P. B., & Seow, C. C. (2000). Plasticizing-antiplasticizing effects of water on physical properties of cassava starch films in the glassy state. *Journal of Food Science*(3), 445–451.

Chartoff, R. P. (1981). In E. A. Turi (Ed.). *Thermal characterization of polymeric materials* (Vol. 1, pp. C 3). USA: Academic Press.

Cheftel, J. C., Cheftel, H., & Besancon, P. (1983). *Introducción a la bioquímica y tecnología de los alimentos* (Vol. 2). Zaragoza, España: Editorial Acribia.

Chen, M. H., Yeh, G. H., & Chiang, B. H. (1996). and physicochemical properties of methylcellulose and chitosan films containing a preservative. *Journal of Food Processing and Preservation, 20*, 379–390.

Chung, D., Chikindas, M., & Yan, K. (2001). Inhibition of *Saccharomyces cerevisiae* by slow release of propyl paraben from a polymer coating. *Journal of Food Protection, 64*(9), 1420–1424.

Chung, H. J., & Lim, S. T. (2003). Physical ageing of glassy normal and waxy rice starches: Thermal and mechanical characterization. *Carbohydrate Polymers, 57*, 15–21.

Chung, H. J., & Lim, S. T. (2004). Physical ageing of glassy normal and waxy rice starches: Effect of ageing time on glass transition and enthalpy relation. *Carbohydrate Polymers, 17*, 855–861.

Delville, J., Joly, C., Dole, P., & Bliard, C. (2003). Influence of photocrosslinking on the retrogradation of wheat starch based films. *Carbohydrate Polymers, 53*, 373–381.

Dumoulin, Y., Alex, S., Szabo, P., Cartilier, L., & Mateescu, M. (1998). Cross-linked amylose as matrix for drug controlled release. X-ray and FT-IR structural analysis. *Carbohydrate Polymers, 37*, 361–370.

Famá, L. M., Rojas, A. M., Goyanes, S., & Gerschenson, L. (2003). Películas comestibles de aplicación industrial. *Anales de las Jornadas SAM – CONAMET – Simposio Materia, 2003*, 898–901. Argentina.

Famá, L., Rojas, A. M., Goyanes, S., & Gerschenson, L. (2005). Mechanical properties of cassava-starch edible films containing sorbates. *Lebensmittel Wissenschaft und Technologie, 38*, 631–639.

Famá, L., Flores, S., Gerschenson, L., & Goyanes, S. (2006). Physical characterization of cassava starch biofilms with special reference to dynamic mechanical properties at low temperatures. *Carbohydrate Polymers, 66*(1), 8–15.

FAO (2004). In Proceedings of the validation forum on the global cassava development strategy (Vol. 6). Global cassava market study business opportunities for the use of cassava. International fund for agricultural development, Rome.

Flores, S., Haedo, A., Campos, C., Gerschenson, L. (2006). Antimicrobial performance of potassium sorbate supported in cassava starch edible films. European Food Research and Technology. Online from July 26th, 2006.

Forssell, P. M., Hulleman, S. H. D., Myllarinen, P. J., Moates, G. K., & Parker, R. (1999). Ageing of rubbery thermoplastic barley and oat starches. *Carbohydrate Polymers, 39*, 43–51.

Franssen, L. R., Rumsey, T. R., & Krochta, J. M. (2004). Whey protein film composition effects on potassium sorbate and natamycin diffusion. *Journal of Food Science, 69*(5), 347–350.

García, M., Martino, M., & Zaritzky, N. (1998). Starch-based coatings: Effect on refrigerated strawberry quality. *Journal of the Science of Food and Agriculture, 76*, 411–420.

García, M., Martino, M., & Zaritzky, N. (2000). Lipid addition to improve barrier properties of edible starch-based films and coatings. *Journal of Food Science, 65*(6), 941–947.

Gerschenson, L., & Campos, C. (1995). Sorbic acid stability during processing and storage of high moisture foods. In G. Barbosa Canovas & J. Welti Chanes (Eds.), *Food preservation by moisture control* (pp. 761–791). Lancaster, USA: Technomic Publishing Co.

Goyanes, S. N. (2000). Dynamic mechanical behaviour of atactic and high impact polystyrene. *Journal Applied Polymer Science, 75*, 865–873.

Gueguen, J., Viroben, G., Noireaux, P., & Subirade, M. (1998). Influence of plasticizers and tratments on the properties of films from peo proteins. *Industrial Crops and Products, 18*, 91–100.

Hale, A., & Bair, E. (1981). In E. A. Turi (Ed.). *Thermal characterization of polymeric materials* (Vol. 1, pp. C4). USA: Academic Press.

Han, J. H., & Floros, J. D. (1997). Casting antimicrobial packageing films and their physical properties and antimicrobial activity. *Journal of Plastic Films and Sheeting, 13*, 287–298.

Hermans, P. H., & Weidinger, A. (1961). On the determination of the crystalline fraction of polyethylenes from X-ray diffraction. *Macromolecular Chemistry*, 24–36.

Hsu, C. K. (2002). Thermal decomposition properties of polymer fibers. *Thermochimica Acta*, 163–167.

ICPDS (1997). International Centre for Diffraction Studies. PCPDFWIN, V.130.

Jongjareonrak, A., Benjakul, S., Visessanguan, W., Prodpran, T., & Tanaka, M. (2006). Characterization of edible films from skin gelatin of brownstripe red snapper and bigeye snapper. *Food Hydrocolloids, 20*, 492–501.

Kittel, C. (1986). Introduction to solid State Physics (pp. 48–51). New York, London, Sydney, Toronto: J. Wiley & Sons, Inc., p. C 2.

Koksel, H., Ahbaz, F., & Ozboy, O. (1983). Influence of wheat-drying temperatures on the birefringence and X-ray diffraction patterns of wet-harvested wheat starch. *Cereal Chemistry, 70*(4), 481–483.

Lens, J. P., de Graaf, L. A., Stevels, W. M., Dietz, C. H. J. T., Verhelst, K. C. S., Vereijken, J. M., et al. (2003). Influence of processing and storage conditions on the mechanical and barrier properties of films cast from aqueous wheat gluten dispersions. *Industrial Crops and Products, 17*, 119–130.

Le Tien, C., Letendre, M., Ispas-Szabo, P., Mateescu, M. A., Delmas-Patterson, G., Yu, H-L., et al. (2000). Development of biodegradable films of whey proteins by cross-linking and entrapment in cellulose. *Journal of Agricultural and Food Chemistry, 48*, 5566–5575.

Lourdin, D., Valle, G. D., & Colonna, P. (1995). Influence of amylose content on starch films and foams. *Carbohydrate Polymers, 27*, 261–270.

Lu, Y., Tighzert, L., Berzin, F., & Rondot, S. (2005). Innovative plasticized starch films modified with waterborne polyurethane from renowable sources. *Carbohydrate Polymers, 61*, 174–182.

Mali, S., Grossman, M. V. E., Garcia, M. A., Martino, M. N., & Zaritzky, N. E. (2006). Effects of controlled storage on thermal, mechanical and barrier properties of plasticized films from different starch sources. *Journal of Food Engineering, 75*(4), 453–460.

Manzocco, L., Nicoli, M. C., & Labuza, T. (2003). Study of bread staling by X-ray diffraction analysis. *Italian Food Technology, XII*(31), 17–23.

Orliac, O., Rouilly, A., Silvestre, F., & Rigal, L. (2003). Effects of various plasticizers on the mechanical properties, water resistance and ageing of thermo-moulded films made from sunflower proteins. *Industrial Crops and Products, 18*, 91–100.

Park, S. L., Daeschel, M. A., & Zhao, Y. (2004). Functional properties of antimicrobial lysozyme-chitosan films. *Journal of Food Science, 69*(8), 215–221.

Rodríguez, M., Osés, J., Ziani, K., & Maté, J. I. (2006). Combined effect of plasticizers and surfactants on the physical properties of starch based edible films. *Food Research International, 39*(8), 840–846.

Snyder, R. L., & Bish, D. L. (1989). Quantitative analysis. In D. L. Bish & J. E. Post (Eds.), *Modern powder diffraction* (pp. 101–144). Washington, DC: Mineralogical Society of America.

Sobral, P. J., do, A., dos Santos, J. S., & García, F. T. (2005). Effect of protein and plasticizer concentrations in films forming solutions on physical properties of edible films based on muscle proteins of a Thai Tilapia. *Journal of Food Engineering, 70*, 93–100.

Sofos, J. N. (1989). *Sorbate food preservatives*. CRC Press Inc.

Sokal, R. R., & Rohlf, J. B. (1969). *Biometry. The principles and practice of statistics in biological research*. San Francisco, California: W.H. Freeman and Company.

Talja, R. A., Helén, A., Roos, Y. H., & Jouppila, K. (2007). Effect of various polyols and polyol contents on physical and mechanical properties of potato starch-based films. *Carbohydrate polymers, 67*(3), 288–295.

Tharanathan, R. N. (2003). Biodegradable films and composite coatings: past, present and future. *Trends in Food Science and Technology, 14*, 71–78.

Valencia Rodríguez, M. T. (2001). Efecto del tratamiento de preservación por depresión e la actividad acuosa en la calidad del alga. Tesis de Magíster en Bromatología y Tecnología de la Industrialización de Alimentos. FCEN-UBA. Argentina.

Valle, G. D., Buleon, A., Carreau, P. J., Lavoie, P. A., & Vergnes, B. (1998). Relationship between structure and viscoelastic behaviour of plasticized starch. by The Society of Rheology, Inc. *Journal of Rheology, 42*(3), 507–525.

Veiga-Santos, P., Suzuky, C. K., Cereda, M. P., & Scamparini, A. R. P. (2005). Microstructure and colour of starch-gum films: Effect of gum deacetylation and additives, Part 2. *Food Hydrocolloids, 19*, 1064–1073.

Viña, S. Z., Mugridge, A., García, M. S., Ferreyra, R. M., Martino, M. N., Chaves, A. R., Zaritzky, N. E. (2006). Effects of polyvinylchloride films and edible starch coatings on quality aspects of refrigerated Brussels sprouts. Food Chemistry. Available online 30 October 2006.

Wilhelm, H. M., Sierakowski, M. R., Souza, G. P., & Wypych, F. (2003). Starch films reinforced with mineral clay. *Carbohydrate Polymers, 52*, 101–110.

Zobel, H. F., & Illinois, A. (1986). Starch crystal transformations and their industrial importance. *Verlagsgesellschaft mbff. D-6940 Weinheim, 40*(1), 1–7.

Zobel, H. F. (1994). Starch granule structure. In R. J. Alexander & H. F. Zobel (Eds.), *Developments in carbohydrate chemistry* (pp. 1–36). St. Paul, Minnesota: The American Association of Cereal Chemists.