Lipid Addition to Improve Barrier Properties of Edible Starch-based Films and Coatings

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ABSTRACT: Effects of formulation (lipid presence, type of starch, and plasticizer) on microstructure, water vapor (WVP) and gas (GP) permeabilities of films and coatings were analyzed. Plasticizer was necessary to maintain film and coating integrity and to avoid pores and cracks. Films made from high amylose starch showed lower WVP and GP than regular corn starch films; permeabilities of films with sorbitol (20 g/L) were lower than those with glycerol. The addition of 2g/L sunflower oil to the formulations decreased WVP of starch-based films; X-ray diffraction and Differential Scanning Calorimetry experiments demonstrated that films with plasticizer and lipid showed lower crystalline-amorphous ratio compared to films without additives. Microstructural observations helped explain the decrease of the film permeabilities during storage.

Key words: starch films, edible coatings, lipid addition, water vapor, gas permeability, microstructure

Introduction

EDIBLE FILMS AND COATINGS CAN HELP MEET THE MANY CHALlenges involved with the marketing of foods that are nutritious, safe, of high quality, stable, and economical. They can control mass transfer between components of a product as well as between the product and the environment: the addition of antioxidants, antimicrobial agents; in their formulations improves their performance. The main components are generally recognized as safe substances (GRAS) such as lipids, proteins, cellulose derivatives, starch, and other polysaccharides. Film materials and their properties have been reviewed extensively (Guilbert 1986, Kester and Fennema 1986, Krochta and De Mulder Johnston 1997).

Hydrophilic coatings with differential gas permeability applied to fruits have shown their influence on fruit physiology by delaying senescence and extending storage life (Olorunda and Aworth 1984, Drake and others 1987, Dhalla and Hanson 1988, Avena Bustillos and others 1993 and 1994, García and others 1998a and b). Their use is frequently limited due to their poor water vapor barrier properties. Water loss from fresh fruits and vegetables leads to a decrease in turgor and weight of the product with the consequent quality and economical losses during commercialization (Avena Bustillos and others 1994). The most well known and oldest coating technique was the application of natural waxes and lipid coatings on specific fruits and vegetables to reduce dehydration, fungi attack, and abrasion during processing and to improve appearance by adding glossiness (Hardenburg 1967, Paull and Chen 1989, Drake and Nelson 1990 and 1991, Baldwin 1994, Hagenmaier and Baker 1994, Baldwin and others 1997). Limitations to their use are poor mechanical properties and oily appearance in some products (Baker and others 1994, Baldwin and others 1997).

Composite films and coatings can be formulated to combine the advantages of both lipid and hydrocolloid components (Baldwin and others 1997, Krochta and De Mulder Johnston 1997). The lipid component in the coating formulation can serve as a good barrier to water vapor while the hydrocolloid component can provide a selective barrier to oxygen and carbon dioxide and the necessary supporting matrix (Guilbert 1986, Kester and Fennema 1986, Drake and others 1987and 1991, Baldwin 1994, Wong and others 1994, Baldwin and others 1997). Starch is an appropriate matrix-forming material and is a commonly used agricultural commodity with a low cost relative to other alternatives. Amylose is responsible for the film-forming capacity of starch.

The functional, organoleptic, nutritional, and mechanical properties of an edible film can be modified by the addition of various chemicals in minor amounts. Plasticizers, such as glycerol, sorbitol, or polyethylene glycol are often used to modify the mechanical properties of the film. These compounds decrease intermolecular attractions between adjacent polymeric chains increasing film flexibility (Donhowe and Fennema 1994). Incorporation of these additives may, however, cause significant changes in the barrier properties of the film. Lipophilic compounds act as emulsifiers and plasticizers increasing film flexibility. Some lipids like vegetable oils, lecithin, fatty acids, and waxes are incorporated to films as plasticizers (Kester and Fennema 1986, Cuppet 1994, Donhowe and Fennema 1994, Hernández 1994). Lipids in a composite film can form an emulsion or a second layer, depending on film application (Baldwin and others 1997, Shellhammer and Krochta 1997). Water vapor transfer generally occurs through the hydrophilic portion of the film, thus, water vapor permeability (WVP) depends on the hydrophilic-hydrophobic ratio of the film components (Hernández 1994). WVP increases with polarity, unsaturation, and branching degree of the lipid, depending also on the water absorption properties of the polar part of the film (Gontard and others 1994). Barrier and mechanical properties depend on film microstructure, which in turn depend on film composition, its formation, and the method of application to the product (Cuq and others 1995). An insight into film microstructure through microscopy observation, studies of permeability, crystallinity, and so on would help explain differences in the macroscopic performance of edible films and coatings. In previous works (García and others 1998a and b) starch-based coatings were used to extend storage life of refrigerated strawberries. The controlling parameter was weight loss associated with a relative high water vapor permeability of coatings. Thus, the addition of lipids to our formulation appears to be an alternative to minimize this problem.

The objectives of the present work were 1) to reduce water vapor permeability and to measure gas permeability of starch-based films and coatings using sunflower oil as the hydrophobic phase and glycerol or sorbitol as plasticizers. 2) to study crystalline-amorphous ratio of the films or coatings by using differential scanning calorimetry (DSC) and X-ray diffraction to understand the effect of film formulation on barrier properties.

Materials and Methods

TWO STARCHES WERE USED FOR FILM AND COATING FORMULAtions: (1) commercial corn starch (Molinos Río de la Plata, Argentina) with 250g/kg amylose and (2) high amylose corn starch, (Amylomaize VII, Cerestar, Hammond, Ind., U.S.A.) with 650g/kg amylose.

In all cases, 20g/L starch aqueous solutions were gelatinized with 10g/L NaOH at room temperature, to obtain the films or coatings (Young 1984, García and others 1995). These suspensions were then neutralized with 7M H_3P0_4. Glycerol or sorbitol (Merck,Whitehouse Station, N.J., U.S.A.) were added as plasticizers after neutralization, at a concentration of 20g/L in the formulation (34.4% dry basis) which was the most effective concentration for this type of films and coatings (García and others 1998 b).

After this stage, sunflower oil (Aceitera General Dehesa S.A., Córdoba, Argentina) was added; oil composition was 99.93% lipids, from which 25.71% were monounsaturated and 64.29% polyunsaturated. Sunflower oil was preferred over a wax because the latter has a high melting point and high temperatures must be avoided when coatings are applied on perishable fruits. Lipid concentrations were 1, 2, 3, 4, 5, 6, and 8 g/L (starch/plasticizer/oil ratio varied between 33.8/33.8/1.7 to 30.2/30.2/12.1, dry basis). Emulsions with lipid as the disperse phase and starch based suspension as the continuous phase were homogenized in an emulsifying equipment (Ultra Turrax, Staufen, Germany) using a dispersing device S 25 N-10 G. Suspensions were dispersed at 12000 and 7800 rpm for 5 min and then were degassed under vacuum. Emulsions were observed by phase-contrast and dark field under a light Ortholux 2, Vario Orthomat (Leitz, Wetzlar, Germany) microscope with a built in camera. Oil droplet sizes were analyzed immediately after preparation and after 30 and 60 minutes to follow emulsion stability.

Starch systems with and without plasticizer were also analyzed. All suspensions were used immediately to obtain bothcoatings and films, depending on the method of preparation. Coatings were used for water vapor permeability determinations and films for gaseous permeabilities, Differential Scanning Calorimetry and X-ray Diffraction determinations.

To obtain films, 15g suspensions were cast on polyethylene films in Petri dishes of 6 cm in diameter and placed in a controlled temperature oven with leveled stages. Samples were dried at 60 °C until constant weight (approximately 8 h). Films were peeled from polyethylene and were stored in a room under controlled conditions (20 °C and 63.8% relative humidity, RH) until they were used for DSC (Differential Scanning Calorimetry), X ray diffraction and gas permeability determinations. The surfaces of films containing lipids were observed under stereomicroscope Zeiss (Zeiss, Germany) after 1 and 7 d of storage at 20 °C and 63.8% RH to analyze lipid migration.

Edible emulsified coatings were applied on slices of carrots (*Daucus sativa*) that were selected as a biological model system with a regular surface area to facilitate the determination of water vapor permeability. Peeled carrots were cut with a core-borer into cylinders 3.7 cm in diameter and 0.5 cm high; then, samples were dipped once in the starch based formulations and dried with an air stream (1.2 m/sec, 20 °C and 84.8% RH) over approximately 2 h (García and others 1998 a and b).

Thickness determination

Thickness values of films and coatings are important for permeability determinations. Thickness of the films was determined using a digital coating thickness gauge Elcometer A 300 FNP 23 (England) for nonconductive materials on nonferrous substrates. Reported values are an average of at least 15 measurements. A stereomicroscope (Zeiss, Germany) was used to determine coating thickness (δ) and to check coating uniformity on carrot slices. To improve coating visualization coated carrots were stained with iodine solution (García and others 1998 a). 5 measurements per carrot slice and 3 slices per formulation were determined under the stereomicroscope using micrographs.

Gas permeability

CO_2 and O_2 permeabilities of the films were assessed by the accumulation method in an especially designed stainless steel cell as described in a previous work (García and others 1999b). The quasi-isostatic method used was based on the measurement of the amount of gas diffusing through a film. The experiment was performed under conditions in which total pressure difference across the test material was zero and the partial pressure difference for the test gas was approximately 1 atm, (Mc Hugh and Krochta, 1994 c). Gas permeability assays were performed in a room under controlled conditions (20 °C and 63.8% RH).

Oxygen and carbon dioxide permeability determinations were done by placing the test-film between two compartments and closing the cell tightly. The volume of each compartment was 29.45 cm^3 and the transfer area was 4.91 cm^2. A stream of test gas passed through one compartment and the pressure was adjusted to 1 atm. To assess test-gas concentration 500 µL gas samples were withdrawn with a gas syringe (Hamilton, St Louis, Mo., U.S.A.) from the other compartment, initially filled with air. Gas concentration was measured in a gas chromatograph Varian Star 3400 cx (Walnut Creek, Calif., U.S.A.) with a Porapack Q column with an inner diameter of 3mm and a length of 1.5m to determine CO_2 concentration and molecular sieve column (0.5 nm pore size) to determine O_2 concentration, using He as carrier. From the moment that differences in CO_2 and O_2 were detected, samples were withdrawn at suitable intervals, 30 minutes for CO_2 and 60 minutes for O_2. At least eight measurements were performed within the range in which gas concentration vs. time was linear. For each tested film formulation, the whole experiment was performed at least in 5 replicates.

Calibration curves to determine gas concentration were obtained with pure gases; concentrations were calculated from the peak areas.

Based on Fick's law the equation governing the permeation of a gas A through a plane sheet (film) of thickness δ (m) in steady state is:

$$JA = D\,S\,(p_{A1} - p_{A2}(t)) \,/\, \delta \qquad (1)$$

where JA is the molar flux of the permeant A, (moles of A/ cm^2 sec), D is the diffusion coefficient (cm^2/ sec), S is the solubility in the film (moles of A/ cm^3 Pa), p_{A1} is the partial pressure of A in compartment 1 and p_{A2} is the partial pressure of A in compartment 2 which varies with time t (sec). Gas solubility(S) relates permeant A concentration in the surface of the film with the external partial pressure. The permeability P' (moles of A m/cm^2 sec Pa) is defined as P' = D S.

Macroscopic mass balance in compartment 2, where the concentration of gas (CO_2 or O_2) is measured by gas chromatography is given by:

$$V\,(dCA/dt) = P'\,(A/\delta)\,(p_{A1} - p_{A2}(t)) \qquad (2)$$

where A is the film permeating area (cm^2) and V is the compartment volume (cm^3), C_A is the molar concentration of gas A (moles/cm^3) and t is the time (sec). Considering ideal gas behavior, the total molar concentration (C) and permeant A concentration are expressed as:

$$C = (n/V) = p/(R\,T) \quad \text{and} \quad C_A = p_A/(RT) \qquad (3)$$

Thus,

$$dp_A/dt = (R\,T/V)\,P'\,(A/\delta)\,(p_{A1} - p_{A2}(t)) \qquad (4)$$

By integrating eq. (4) the obtained solution is:

$$\ln\,[(y_{A1} - y_{A2,0})\,/(y_{A1} - y_{A2})] = (P'\,/\delta)\,RT\,(A/V)\,t \qquad (5)$$

where Dalton's law was applied; y_{A1} is the gas molar fraction in compartment 1, $y_{A2,0}$ is the initial gas molar fraction in compartment 2 at initial time and y_{A2} is the gas molar fraction in compartment 2 at each time (t). Plotting $\ln\,[(y_{A1} - y_{A2,0})/(y_{A1} - y_{A2})]$ vs time a straight line was obtained. The value of the permeability was calculated from the slope, knowing compartment volume (V), film area (A) and film thickness (δ) . Permeability was calculated as $P = P'/(RT)$ and was expressed in [cm^3 /m sec Pa].

Water vapor permeability

Water vapor permeability (WVP) of the different coating formulations was determined using cylindrical carrot samples. Coated samples were placed in individual test cups and equilibrated for 24 h in a desiccator chamber maintained at 98.2% RH with a $K_2Cr_2O_7$ (Anedra, Argentina) saturated solution at 20 °C. Water activity (a_w) of carrots was measured in a thermoconstanter Humidat TH2/RTD-33/BS, model AG CH-8050 (Novasina, Zurich, Switzerland). Then, samples were weighed in an analytical balance and placed in another cabinet equilibrated at 33.3% RH with $MgCl_2.6H_2O$ (Anedra, Argentina) at 20 °C. At regular intervals samples were weighed to calculate weight loss with time. Data were obtained during a period of 3 h. It was assumed that during this period the loss of water was produced in conditions where water surface concentration remained constant and coincided with the equilibrium value ($a_w = 0.988$). A control test with uncoated carrots was also performed to determine the mass transfer coefficient of water vapor in air ($k_{air} = 1.3\ 10^{-8}$ g m^{-2} sec^{-1} Pa^{-1}), (García and others 1998 b).

Water vapor flux, Fl (g m^{-2} sec^{-1}), was calculated from the slope of the weight loss vs time curves (Δw/Δt) and the mass transfer area (A) was considered as the upper surface and the lateral area of the sliced carrots ($1.656\ 10^{-3}$ m^2).

$$FI = (\Delta w)/(\Delta t \times A) \qquad (6)$$

Coating permeance, P (g m^{-2} sec^{-1} Pa^{-1}), was determined, considering that water vapor is transferred in series through the coating and the surrounding air, using the following equation:

$$FI = (p_i - p_a)\,/\,[(1/P) + (1/\,k_{air})] \qquad (7)$$

where p_i is the partial water vapor pressure corresponding to the carrot-coating interface (3129.18 Pa) which was calculated considering the aw of the carrots and the total water vapor pressure at 20 °C (3167.19 Pa), p_a is the partial water vapor pressure in the environment with 33.3% relative humidity at 20 °C expressed in Pa and k_{air} is the mass transfer coefficient of water vapor in air.

Water vapor permeability was calculated as: $WVP = P\delta$, with δ being the coating thickness (m). The whole experiment was performed at least six times for each formulation.

Differential scanning calorimetry (DSC)

Samples of the formulated films were tested in a Polymer Laboratories DSC (Rheometric Scientific, Surrey, U.K.) working under a PL-V5.41 software. Indium was used to calibrate temperature and heat flux. Samples of 1.5 to 2mg were weighed in aluminum pans and were hermetically sealed; an empty pan was used as reference. Samples were run between 0 and 130 °C with 10 °C/min heating rate. Once analyzed, pans were punctured and dried until constant weight at 100 °C to get sample dried weight. Enthalpies ΔH (mJ/mg), onset and peak transition temperatures were obtained from the thermograms. Reported values are the mean of three measurements.

X ray diffraction

Samples were analyzed between $2\theta = 3°$ to $2\theta = 40°$ in a diffractometer Phillips PW 1390 (The Netherlands) using K_α Cu radiation ($\lambda = 1.5418$Å), 40 kV and 20mA. Modifications of thecrystalline structure of films stored in a room at 20 °C and 63.8% RH were evaluated.

Statistical analysis

Systat-software (SYSTAT, Inc., Evanston , Ill., U.S.A., 1990) version 5.0 was used for all statistical analysis. Analysis of variance (ANOVA), Fisher LSD means comparison test and regression analysis were applied. The significance levels used were 0.05 and 0.01.

Results

Film characterization

Stirring velocity of the emulsion system was an important factor to obtain homogenous films and coatings and had great influence on their performance. The highest velocity used (12000 rpm) produced foam that could not be eliminated with vacuum. Once dried these films showed irregular surfaces compared to the smooth and homogeneous surfaces obtained with 7800 rpm, this one was the selected stirring velocity for emulsion dispersion.

Light microscopy showed that emulsion stability was maintained at least for an hour that was the longest observation time analyzed. Coatings were homogeneous as seen by iodine staining. Emulsified films showed smooth surfaces and a compact structure by scanning electron microscopy (SEM) as reported in a previous work (García and others 1999a). These results indicated that lipids were homogeneously dispersed in the film matrix.

Lipid migration depends on the matrix microstructure; in the present work starch-based films containing less than 5 g/L sunflower oil did not show oil migration to the surface. Wong and coworkers (1992) reported that similar lipid concentrations did not produce oil migration in chitosan-fatty acid films; however, a marked lipid migration was reported in methylcellulose films (Greener and Fennema 1989; Vojdani and Torres 1989, 1990).

Thickness of films measured by an electronic gauge are shown on Table 1. Coating thicknesses determined with a stereomicroscope varied between 40-50 μm for all tested formulations.

Barrier properties of films

Gas permeability. ANOVA showed that CO_2 and O_2 permeabilities of films without plasticizer were significantly higher ($p < 0.05$) than plasticized films (Table 1 and Fig. 1). These results can be attributed to the presence of pores and cracks in the surface of unplasticized films observed by SEM in previous works (García and others 1999 a and b). Pores also may explain the higher dispersion of the permeability values compared to those of plasticized films (Table 1 and Fig. 1). For all the tested formulations, CO_2 permeability decreased significantly ($p < 0.05$) with storage time (Fig. 1).

ANOVA showed that lipid presence did not modify ($p > 0.05$) CO_2 permeability of plasticized starch based films, however the type of both plasticizer and starch used in film formulations were significant factors ($p < 0.05$) (Table 1). The formulation containing amylomaize, sorbitol and sunflower oil showed the lowest CO_2 permeability.

Films with sorbitol exhibited statistically lower ($p < 0.05$) oxygen permeabilities than films with glycerol. Mc Hugh and Krochta (1994 a, b) found similar results with protein films.

The lowest values of CO_2 permeability were obtained for amylomaize films either with sorbitol or glycerol. With regard to O_2 permeabilities, starch type was not a significant factor ($p > 0.05$) for sorbitol-plasticized films. However, amylomaize films with glycerol had lower O_2 permeability than corn starch films.

Table 1 shows that O_2 permeabilities were much lower than those for CO_2, indicating a selective action of these films on gas permeabilities. These effects can be attributed to a higher solubility of CO_2 in the starch films.

Permeability strongly depend on the interaction between the polymer matrix and the permeating gas and the environmental conditions such as temperature and relative humidity. Synthetic materials show lower gas permeabilities like LDPE with 2.16 10^{-11} for O_2 and 9.45 10^{-11} ($cm^3\ m^{-1}\ sec^{-1}\ Pa^{-1}$) for CO_2 but have low selectivity between CO_2 and O_2 (around 4). Cuq and coworkers (1995) compared the CO_2 to O_2 permeability ratio for several synthetic and edible films and reported that edible films show higher selectivity than synthetic ones with ratios between 8 to 30. In the present work we found an average ratio of 13. The development of edible films and coatings with selective gas permeability could be very promising for controlling respiratory exchange and improving the conservation of fresh or minimally processed vegetables (Cuq and others 1998; García and others 1998a and b).

Water vapor permeability. The optimum lipid concentration was selected by WVP measurements. The lowest WVP of coatings was obtained with 2 g/L sunflower oil; higher oil concentrations increased WVP (Fig. 2).

Another factor analyzed in permeability measurements was the stirring velocity of the emulsifying equipment. WVP of films prepared at 12000 rpm was 2.01 ± 0.57 10^{-10} ($g\ m^{-1}\ sec^{-1}\ Pa^{-1}$) and for those prepared at 7800 rpm was below 1.94 ± 0.42 10^{-10} ($g\ m^{-1}\ sec^{-1}\ Pa^{-1}$). Thus, a speed of 7800 rpm was selected because foam formation was minimized and WVP of coatings was lower.

Table 1—Effect of lipid phase addition, plasticizer and starch type on thickness and gas permeabilities of starch-based films.

Starch base	Additives	Film thickness (μm)	CO_2 permeability 10^9 ($cm^3\ m^{-1}\ sec^{-1}\ Pa^{-1}$)	O_2 permeability 10^{10} ($cm^3\ m^{-1}\ sec^{-1}\ Pa^{-1}$)
Corn	—	119.92 ± 32.52[a]	29.21 ± 13.89	15.92 ± 2.99
	Glycerol	104.34 ± 24.24	5.69 ± 0.97	4.61 ± 0.51
	Glycerol + SO	109.72 ± 13.99	5.87 ± 0.58	Nd[b]
	Sorbitol	110.71 ± 23.11	4.19 ± 0.81	2.48 ± 0.32
	Sorbitol + SO	115.77 ± 22.96	4.72 ± 0.65	Nd
Amylomaize	—	157.45 ± 35.48	28.05 ± .37	26.45 ± 2.48
	Glycerol	128.70 ± 20.82	3.85 ± 1.28	3.21 ± 0.19
	Glycerol + SO	135.04 ± 26.43	4.39 ± 0.90	Nd
	Sorbitol	134.34 ± 25.13	2.96 ± 0.46	2.28 ± 0.26
	Sorbitol +SO	137.60 ± 24.84	3.43 ± 0.21	Nd

Glycerol and sorbitol concentration = 20g/L
SO = sunflower oil (2g/L)
[a] Value ± standard deviation
[b] Not determined

Table 2—Lipid phase addition, starch-based and plasticizer type effect on water vapor permeability of coatings.

Starch base	Additives	Water vapor permeability 10^{10} ($g\ m^{-1}\ sec^{-1}\ Pa^{-1}$)
Corn	—	3.68 ± 2.24[a]
	Glycerol	2.57 ± 1.04
	Glycerol + SO	1.92 ± 0.47
	Sorbitol	1.75 ± 0.14
	Sorbitol +SO	1.22 ± 0.11
Amylomaize	—	2.62 ± 1.39
	Glycerol	2.14 ± 0.75
	Glycerol + SO	1.76 ± 0.37
	Sorbitol	1.21 ± 0.15
	Sorbitol +SO	0.97 ± 0.08

Glycerol and sorbitol concentration = 20g/L
SO = sunflower oil (2g/L)
[a] Value ± standard deviation

Lipid presence decreased significantly ($p < 0.05$) water vapor permeability (WVP) of starch based films due to its hydrophobicity (Table 2). ANOVA showed that the addition of a plasticizer improved barrier properties of starch-based films and coatings for

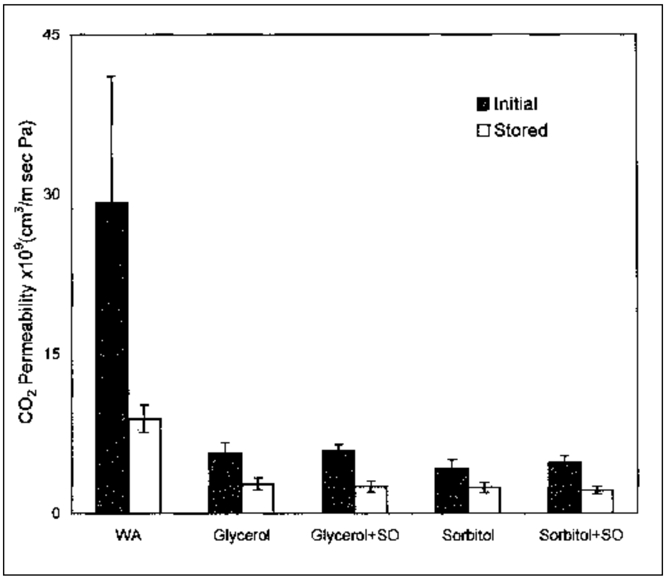

Fig. 1—Effect of storage time (20 d at 20 °C and 63.8% relative humidity) on CO_2 permeability of corn starch-based films. WA: without additives, SO: sunflower oil (2g/L), plasticizer concentration (glycerol or sorbitol): 20g/L

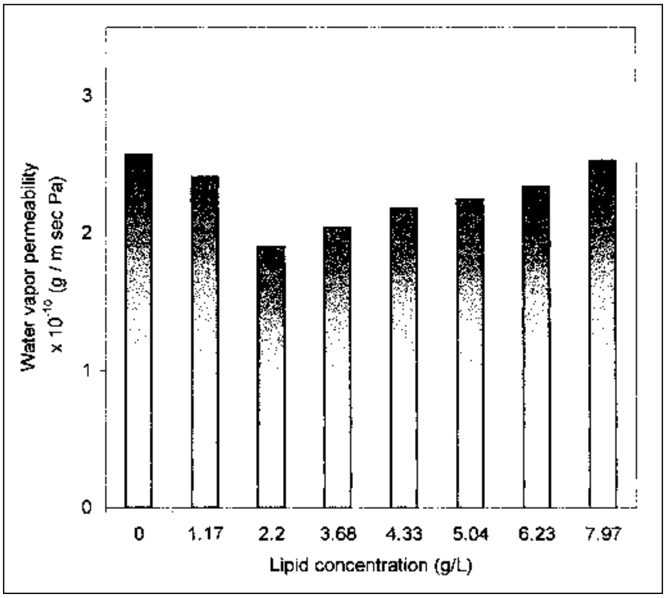

Fig. 2—Effect of lipid concentration (sunflower oil) on water vapor permeability of corn starch films plasticized with 20g/L glycerol. Maximum standard deviation = 0.53

both types of films with and without lipids (Table 2). Sorbitol gave lower WVP values compared to glycerol plasticized coatings. Mc Hugh and Krochta (1994a) found similar results for alginate and pectin films using the same plasticizer concentrations.

Coatings without plasticizer gave significantly ($p < 0.05$) higher WVP values than those with plasticizer (Table 2). This difference was attributed to the presence of pores and cracks in unplasticized coatings, as was previously discussed in gas permeability section. According to Donhowe and Fennema (1993) amylose molecules without plasticizer produce crystalline and brittle films. Amylomaize coatings with the highest amylose content and plasticized either with sorbitol or glycerol showed lower WVP values ($p < 0.05$) than those of corn starch coatings (Table 2). A similar trend was observed by other workers analyzing the effect of amylose content in starch gel structures (Miles and others 1985 a, b; Noel and others 1992). However, the effect of amylose content of the starch matrix was no longer significant under the presence of lipids in the coating formulation (Table 2).

Formulations containing both sorbitol and sunflower oil were the most effective in decreasing WVP of starch based coatings. Even though lipid addition decrease WVP of starch-based films, values are still higher than those of synthetic films, for example: $8.4\ 10^{-11}$ (g m^{-1} sec^{-1} Pa^{-1}) for cellophane, $3.6\ 10^{-13}$ for LDPE and $2.2\ 10^{-13}$ for PVDC (Shellhammer and Krochta, 1997). However, our results are within the range of other edible films (Table 2). Gennadios and coworkers (1994b) improved the commonly standard method used to measure WVP and reviewed several hydrophilic edible film data. Reported WVP values were $5.3\ 10^{-10}$ (g m^{-1} sec^{-1} Pa^{-1}) for corn zein-glycerin, $7.0\ 10^{-10}$ for wheat gluten-glycerin, $3.8\ 10^{-10}$ for amylose and $1.9\ 10^{-10}$ for hydroxypropylmethylcellulose-polyethyleneglycol-hydrogenated cottonseed and soybean oils.

Crystalline structure of the films

Differential scanning calorimetry (DSC). At initial storage time, corn starch and amylomaize films did not show any peak on DSC thermograms, indicating that starch gelatinization with NaOH was complete. However, stored films showed an endothermic transition with a peak temperature between 50 and 54 °C. With increasing storage times the peak became narrower and peak temperature and the corresponding enthalpy (ΔH) increased (Table 3). This thermal transition could be associated with crystal growth and recrystallization processes that take place during storage. Amylomaize formulated films due to its higher amylose content, showed higher ΔH values (Fig. 3a) compared to those of corn starch films along storage time (Fig. 3b).

Lower enthalpy (ΔH) and peak temperatures were obtained with stored plasticized films compared to unplasticized ones (Table 3 and Fig. 3a and b). Both plasticizers glycerol and sorbitol limited crystal growth and recrystallization because they interact with the polymeric chains hindering their alignment and, thus, crystal formation. Corn starch films with sorbitol showed lower crystallinity, characterized by lower ΔH values as compared to films plasticized with glycerol (Fig. 3b). Since sorbitol resembles better than glycerol the molecular structure of glucose units that build the starch polymeric chains, sorbitol chances to interact with polymeric chains are higher.

Sunflower oil concentration of 2g/L, that led to the lowest WVP values, did not modify significantly ΔH compared to time curves compared to those of plasticized films without lipid. However, higher oilconcentrations (> 2g/L) in the film formulation decreased enthalpy values and increased gas permeability (Fig. 2). Thus, film crystallinity decreased probably by interfering polymeric chain association due to steric hindrances.

X ray diffraction

X-ray diffraction patterns of corn starch films without addi-

Table 3—Effect of lipid phase and plasticizer (film additives) on DSC measurements of corn starch-based films.

Film additives	Storage[a] time (days)	Peak temp. (°C)	Peak range (°C)	Enthalpy ΔH (mJ/mg)
Without additives	0	—[d]	—	—
	15	51.63	7.53	113.74
	30	53.32	6.95	138.01
Glycerol[b]	0	—	—	—
	15	45.21	16.07	68.07
	30	47.75	15.66	93.70
Glycerol + SO[c]	0	—	—	—
	15	46.27	17.41	62.80
	30	46.51	14.11	89.50
Sorbitol[b]	0	—	—	—
	15	43.42	18.29	41.67
	30	45.79	18.07	55.20
Sorbitol + SO	0	—	—	—
	15	44.94	18.04	47.12
	30	45.17	15.65	65.43

[a] Storage at 20 °C and 63.8% relative humidity [b] Glycerol and sorbitol concentration = 20g/L
[c] SO = sunflower oil (2g/L)
[d] Not detected

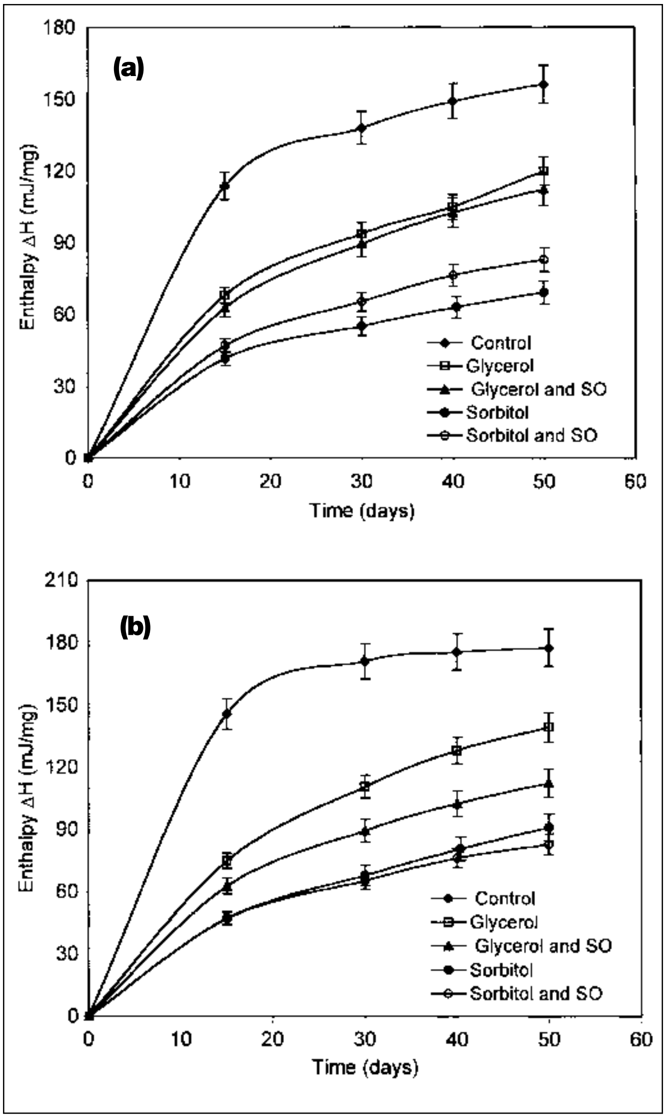

Fig. 3—Effect of plasticizer and lipid presence on transition enthalpy of (a) corn starch films and (b) amylomaize films stored at 20 °C and 63.8 % relative humidity. (◆)Control films without plasticizer, (□) films with 20g/L glycerol, (▲) films with 20g/L glycerol and 2g/L sunflower oil, (●) films with 20g/L sorbitol, (○) films with 20g/L sorbitol and 2g/L of sunflower oil

tives at initial storage time is shown in Fig. 4. They are characterized by sharp peaks associated with the crystalline diffraction and an amorphous zone. The amorphous fraction of the sample can be estimated by the area between the smooth curve drawn following the scattering hump and the baseline joining the background within the low- and high-angle points. The crystalline fraction can be estimated by the upper region above the smooth curve (Snyder and Bish, 1989; Köksel and others 1993).

Corn starch films (Fig. 4) showed larger amorphous zones than amylomaize formulations (Fig. 5a). Crystalline fraction increased at the expense of the amorphous zone with storage time for all formulations and was more evident in films without additives (Fig. 5). All films tended to a common crystalline structure of higher stability with storage time. The peak pattern observed at 60 d of storage was similar to that reported by Bader and Göritz (1994) for films obtained with autoclaved amylomaize, being 2θ = 17° the peak with the highest intensity.

These results agree with the DSC studies that showed endothermic transitions with lower peak temperatures and lower ΔH for films with plasticizer and lipid than for control films. The X-ray pattern of films with plasticizer and lipid was not strongly modified with storage time, (Fig. 5 c, d). This observation was attributed to the additive presence, which facilitated polymeric chain mobility allowing a rapid development of the most stable structure, and reduced crystal growth by interfering with polymeric chain arrangement.

X-ray analysis of films plasticized with sorbitol showed a similar trend compared to glycerol ones; peak intensities of films with sorbitol slightly increased with storage time.

Discussion

As FOR SYNTHETIC MATERIALS, GAS AND VAPOR PERMEABILITIES of edible films and coatings depend on several factors such as: the integrity of the film, the ratio between crystalline and amorphous zones, the hydrophilic-hydrophobic ratio and the polymeric chain mobility. The interaction between film-forming polymer and the plasticizer or other additives are also important factors in film permeability.

According to Donhowe and Fennema (1993) permeability increases with decreasing crystalline-amorphous ratio. Crystalline structures are extremely tightly packed and tend to be impermeable but fragile. The decrease of CO_2 permeability of the films during storage (Fig. 1) could be explained by considering that the crystalline-amorphous ratio increased with time. The longer the storage time, the higher the crystalline-amorphous ratio and the lower the permeability.

Permeability of starch films without additives was high due to the presence of pores and cracks; the increase of crystallinity with time markedly reduced the effect of pores. For example for corn starch films CO_2 permeability decreased from 29.2 10^{-10} at initial time to 8.9 10^{-10} ($cm^3\ m^{-1}\ sec^{-1}\ Pa^{-1}$) after 20-d storage, (Fig. 1).

Amylomaize films with higher amylose content showed higher crystallinity and lower permeabilities than corn starch films (Table 1).

In the present work the addition of plasticizers improved barrier properties of starch-based films by avoiding pores and cracks (Table 1 and 2). Glycerol and sorbitol are compatible with amylose and improved mechanical properties of films, by decreasing intermolecular attraction and interfering with the amylose packing (Donhowe and Fennema 1993). Generally in protein films, plasticizer addition increases polymer mobility and permeability to gases (Gennadios and others 1994a, Mc Hugh and Krochta 1994a, b; Park and others 1994, Parris and others 1995). However, Herald and others (1996) reported that in polysaccharide films like those of methylcellulose, permeability is only slightly modified by the addition of glycerol. Plasticized starch-based films and coatings showed selective gas permeabilities (higher to CO_2

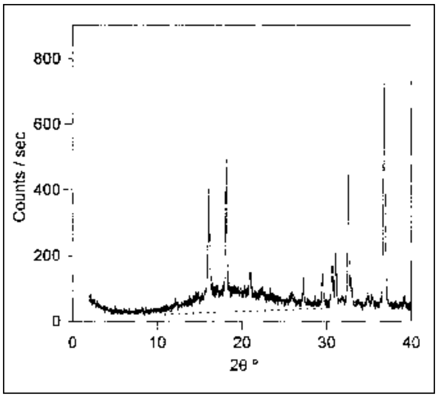

Fig. 4 (above)—X-ray diffraction patterns of corn starch films without plasticizer nor lipid at initial storage time

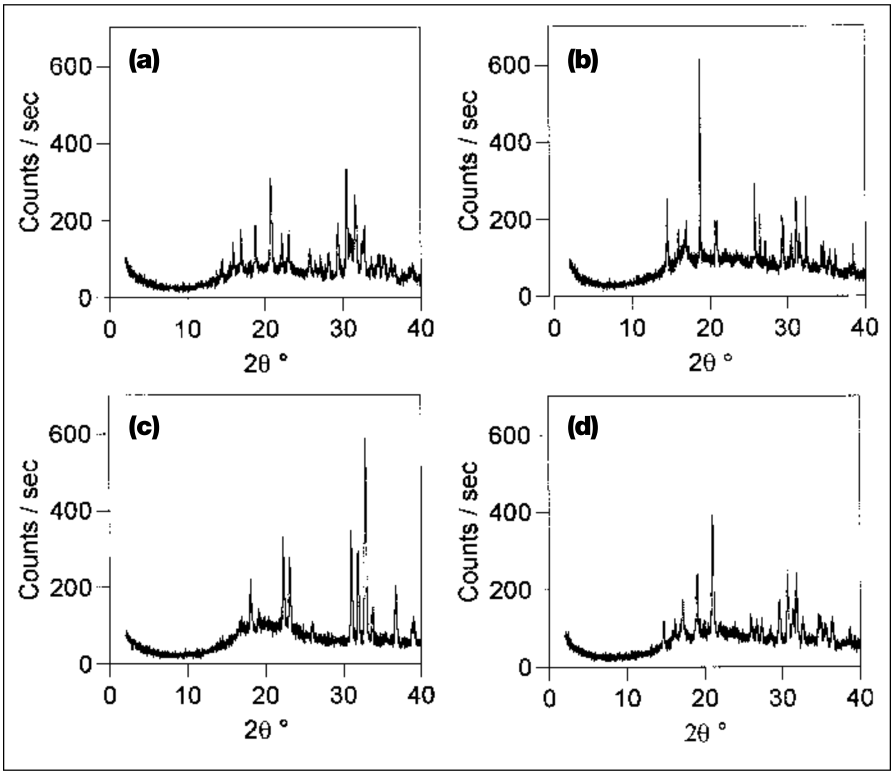

Fig. 5 (right)—X-ray diffraction patterns of amylomaize films: (a) control (without plasticizer nor lipid) at initial storage time; (b) control at 60-d storage; (c) films containing 20 g/L glycerol and 2g/L sunflower oil at initial storage time; (d) films containing 20 g/L glycerol and 2g/L sunflower oil at 60-d storage. Films were stored at 20 °C and 63.8% relative humidity.

than to O_2) and lipid addition did not modify significantly these properties. Gas selectivity helps extending storage life of fruits and vegetables by controlling respiratory exchange and reduces oxidation of foods with high lipid content (Cuq and others 1998).

The addition of lipids to polysaccharide-based films produced good barriers to water vapor due to its hydrophobicity. The decrease in WVP is important in the case of fresh fruits and vegetables because composite coatings retard moisture loss and subsequent shriveling of fresh products.The optimum lipid concentration is a balance between hydrophilic-hydrophobic ratio and crystalline-amorphous ratio. Increasing oil concentration above the optimum (2g/L) although increased the hydrophobic fraction, WVP increased (Fig. 2). This result was attributed to oil migration and to a decrease of the crystalline-amorphous ratio as indicated by DSC.

Additives like plasticizers and lipids reduced modifications of crystalline-amorphous ratio with time (as evidenced by DSC and X-ray diffraction) because they interfere with polymer chain alignment and with crystalline growth (Fig. 3 and 4).

Plasticizer and lipid addition improved starch-based coatings performance by increasing barrier properties to water vapor maintaining the selective gaseous permeability.

References

Avena-Bustillos RJ, Cisneros-Zeballos LA, Krochta JM, Saltveit ME. 1993. Optimization of edible coatings on minimally processed carrots using response surface methodology. Am Soc of Agric Engin 36 (3): 801-805.

Avena-Bustillos RJ, Krochta JM, Saltveit ME, Rojas-Villegas RJ, Sauceda-Pérez JA. 1994. Optimization of edible coating formulations on zucchini to reduce water loss. J Food Eng. 21: 197-214.

Bader HG and Göritz D. 1994. Investigations on high amylose corn starch films. Part 1: Wide angle X-ray scattering (WAXS). Starch/Starke 46 (6): 229-232.

Baker RA, Baldwin EA, Nisperos-Carriedo MO. 1994. Edible coatings for processed foods. In: Edible coatings and films to improve food quality. Krochta JM, Baldwin EA, Nisperos-Carriedo M, Editors. p. 89-104. Technomic Pub Co. Lancaster Pa.

Baldwin EA. 1994. Edible coatings for fruits and vegetables, past, present and future. In: Edible coatings and films to improve food quality. Krochta JM, Baldwin EA, Nisperos-Carriedo M, Editors. p. 25-64. Technomic Pub. Co. Lancaster, Pa.

Baldwin EA, Nisperos-Carriedo MO, Hagenmaier RD, Baker RA. 1997. Use of lipids in coatings for food products. Food Technol 51 (6): 56-64.

Cuq B, Gontard N, Guilbert S. 1995. Edible films and coatings as active layers. In: Active food packaging. Rooney ML, Editor. p. 111-142. Chapman & Hall, UK

Cuq B, Gontard N, Guilbert S. 1998. Proteins as agricultural polymers for packaging production. Cereal Chem 75 (1): 1-9.

Cuppet SL. 1994. Edible coatings as carriers of food additives, fungicides and naturals antagonists. In: Edible coatings and films to improve food quality. Krochta JM, Baldwin EA, Nisperos-Carriedo M, Editors. p. 121-137. Technomic Pub Co. Lancaster Pa.

Dhalla R and Hanson SW. 1988. Effect of permeability coatings on the storage life of fruits. II Pro-long treatments of mangoes (Mangifera indica L cv. Julie). Int J Food Sci Technol. 23: 107-112.

Donhowe IG and Fennema OR. 1993. The effects of plasticizers on crystalinity, permeability, and mechanical properties of methylcellulose films. J of Food Process Preserv. 17: 247-257.

Donhowe IG and Fennema OR. 1994. Edible films and coatings: characteristics, formation, definitions and testing methods. In: Edible coatings and films to improve food quality. Krochta JM, Baldwin EA, Nisperos-Carriedo M, Editors. p. 1-21. Technomic Pub. Co. Lancaster Pa.

Drake SR, Fellman JK, Nelson JW. 1987. Post-harvest use of sucrose polyesters for extending the shelf-life of stored Golden Delicious apples. J Food Sci. 52 (5): 1283-1285.

Drake SR and Nelson JW. 1990. Storage quality of waxed and nonwaxed "Delicious" and "Golden Delicious" apples. J Food Qual. 13: 331-334.

Drake SR, Cavalieri R, Kupferman EM. 1991. Quality attributes of "D'Anjou pears after different wax drying and refrigerated storage. J Food Qual. 14: 455-465.

García MA, Martino MN, Zaritzky NE. 1995. Comparison of amylose enrichment procedures for food applications. Cereal Chem. 72 (6): 552-558.

García MA, Martino MN, Zaritzky NE. 1998a. Starch-based coatings: effect on refrigerated strawberry (Fragaria ananassa) quality. J Sci Food Agric. 76: 411-420.

García MA, Martino MN, Zaritzky NE. 1998b. Plasticizer effect on starch-based coatings applied to strawberries (Fragaria ananassa). J Agric Food Chem. 46: 3758-3767.

García MA, Martino MN, Zaritzky NE. 1999a. Effect of lipid and plasticizer addition on microstructure of starch-based films. J Scanning Microscopies. 21(2): 154-155.

García MA, Martino MN, Zaritzky NE. 1999b. Edible starch films and coatings characterization: SEM, water vapor and gas permeabilities. J Scanning Microscopies. 21(5): 348-353.

Gennadios A, Mc Hugh TH, Weller CL, Krochta JM. 1994a. Edible coatings and films based on proteins. In: Edible coatings and films to improve food quality. Krochta JM, Baldwin EA, Nisperos-Carriedo M, Editors. p. 201-277. Technomic Pub. Co. Lancaster Pa.

Gennadios A, Weller CL, Gooding CH, 1994b. Measurement errors in water vapor permeability of highly permeable, hydrophilic edible films. J Food Eng. 21, 395-409.

Gontard N, Duchez C, Cuq J-L, Guilbert S. 1994. Edible composite films of wheat gluten and lipids: water vapor permeability and other physical properties. Int J Food Sci Technol. 19: 39-50.

Greener IK and Fennema OR. 1989. Evaluation of edible, bilayer films for use as moisture barriers for foods. J Food Sci. 54: 1400-1406.

Guilbert S. 1986. Technology and application of edible protective films. In: Food packaging and preservation. Theory and Practice. Mathlouthi M (Ed.) p. 371-394. Elsevier Applied Science Publishing Co. London UK

Hagenmaier RD and Baker RA. 1994. Wax microemulsions and emulsions as citrus coatings. J Agric. Food Chem. 42: 899-902.

Hardenburg RE. 1967. Wax and related coatings forhorticultural products - A bibliography. Agric Res Serv Bull 965. Cornell Univ. Ithaca NY

Herald TJ, Hachmeister KA, Huang S, Bowers JR. 1996. Corn zein packaging materials for cooked turkey. J Food Sci. 61 (2): 415-417.

Hernandez E. 1994. Edible coatings for lipids and resins In: Edible coatings and films to improve food quality. Krochta JM, Baldwin EA, Nisperos-Carriedo M, Editors. p. 279-304. Technomic Pub Co. Lancaster Pa.

Kester JJ and Fennema OR. 1986. Edible films and coatings: a review. Food Technol. 12: 47-59.

Köksel H, Ahbaz F, Özboy Ö. 1983. Influence of wheat-drying temperatures on the birefringence and X-ray diffraction patterns of wet-harvested wheat starch. Cereal Chem 70 (4):481-483.

Krochta JM and De Mulder-Johnston C. 1997. Edible and biodegradable polymer films: challenges and opportunities. Food Technol. 51(2): 61-77.

Mc Hugh TH and Krochta JM. 1994a. Milk-protein-based edible films and coatings. Food Technol. 48 (1): 97-103.

Mc Hugh TH and Krochta JM. 1994b. Sorbitol- vs glycerol- plasticized whey protein edible films: integrated oxygen permeability and tensile property evaluation. J Agric Food Chem. 42 (4): 841-845.

Mc Hugh TH and Krochta JM. 1994c. Permeability properties of edible films. In: Edible coatings and films to improve food quality. Krochta JM, Baldwin EA, Nisperos-Carriedo M, Editors. p. 139-187. Technomic Pub Co. Lancaster Pa.

Miles MJ, Morris VJ, Orford PD, Ring SG. 1985a. The roles of amylose and amylopectin in the gelation and retrogradation of starch. Carbohydr Res. 135: 271-281.

Miles MJ, Morris VJ, Ring SG 1985b. Gelation of amylose. Carbohydr Res. 135: 257-269.

Noel TR, Ring SG, Whittman MA. 1992. The structure and gelatinization of starch [Review]. Food Sci Technol Today 6 (3): 159-162.

Olorunda AO and Aworh OC. 1984. Effects of Pro-long, a surface coating agent, on the shelf-life and quality attributes of plantain. J Sci Food Agric. 35: 573-578.

Park HJ, Bunn JM, Weller CL, Vergano PJ, Testin RF. 1994. Water vapor permeability and mechanical properties of grain protein-based films as affected by mixtures of polyethylene glycol and glycerin plasticizers. Trans. of the ASAE 37 (4): 1281-1285.

Parris N, Coffin DR, Joubran RF, Pessen H. 1995. Composition factors affecting the water vapor permeability and tensile properties of hydrophilic films. J Agric Food Chem. 43: 1432-1435.

Paull RE and Chen NJ. 1989. Waxing and plastic wraps influence water loss from papaya fruit during storage and ripening. J Am Soc Hort Sci. 114: 937-942.

Shellhammer TH and Krochta JM. 1997. Whey protein emulsion film performance as affected by lipid type amount. J Food Sci. 62 (2): 390-394.

Snyder RL and Bish DL. 1989. Quantitative analysis. In: Modern Powder diffraction. Bish DL and Post JE, Editors. p. 101-144. Mineralogical Society of America. Washington DC

Vojdani F and Torres JA. 1989. Potassium sorbate permeability of methylcellulose and hydroxypropylmethylcellulose multi-layer films. J Food Process Preserv. 13: 417-430.

Vojdani F and Torres JA. 1990. Potassium sorbate permeability of methylcellulose and hydroxypropylmethylcellulose coatings: effect of fatty acids. J Food Sci. 55(3): 841-846.

Wong DWS, Gastineau FA, Gregorski KS, Tillin SJ, Pavlath AE. 1992. Chitosan-lipids films: microstructure and surface energy. J Agric Food Chem. 40: 540-544.

Wong DWS, Tillin SJ, Hudson JS, Pavlath AE. 1994. Gas exchange in cut apples with bilayer coatings. J Agric Food Chem. 42: 2278-2285.

Young AH. 1984. Fractionation of starch. In: Starch, chemistry and technology. Whistler RL, Be Miller JN, Paschall EF, Editors p. 249-283. Academic Press. Orlando FL USA.

MS 19991027 received 10/11/99; revised 3/8/00; accepted 4/10/00.

The financial support provided by CONICET (Consejo Nacional de Investigaciones Científicas y Técnicas), Proyecto BID 802 OC-AR PID N° 358 and University of La Plata is gratefully acknowledged.

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