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Three under-utilised sources of starch from the Andean region in Ecuador. Part II. Rheological characterisation

S. Santacruz[a,b,1], J. Ruales[a], A.-C. Eliasson[b,*]

[a]*Instituto de Investigación Tecnológica, Escuela Politécnica Nacional, P.O. Box 1701 2759, Quito, Ecuador*
[b]*Department of Food Technology, Lund University, P.O. Box 124, S-221 00 Lund, Sweden*

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Abstract

The dynamic rheological behaviour of *Arracacha xanthorriza* (white carrot), *Canna edulis* (achira) and *Oxalis tuberosa* (oca) starch gels were studied in strain-sweep mode. The starch was diluted with water to cover particle volume fractions of 1–3. The starch suspensions were heated at 89 °C for 30 min, and their swelling power and amylose leaching were measured. *C. edulis* showed the highest swelling power and particle rigidity (the elastic modulus, G', of a starch gel at a particle volume fraction of 1 is defined as rigidity), forming stronger gels than *O. tuberosa* and *A. xanthorriza*. Storage of the three starch gels at 4 °C showed that *A. xanthorriza* formed gels which were stable in G' and phase angle for three days of storage. *C. edulis* and *O. tuberosa* gels kept at the same refrigeration conditions showed a high increase in G' during the first day of storage. A decrease in pH from 6.5 to 4.0 produced a loss of structure in the three starch gels, as was showed by the reduction of G'. Storage at freezing temperature (−20 °C) produced higher changes in G' than refrigeration conditions. There was a rapid formation of an elastic structure in *C. edulis* and *O. tuberosa* starch gels on the first day of storage in both refrigeration and freezing conditions.

Keywords: *Arracacha xanthorriza*; *Canna edulis*; *Oxalis tuberosa*; Retrogradation; Freezing storage; Refrigeration storage

1. Introduction

In addition to being a main component in raw materials such as wheat flour, rice, maize, etc. starch is a common additive in the food industry. The most frequently used native starches do not always perform in an optimal way. Thus to obtain starch of suitable properties it may be necessary to modify the starch, chemically, physico-chemically or genetically. However, another approach could be to search for new sources of starch; the present investigation was performed as a part of the characterisation of the physico-chemical properties of three Andean starches, namely starches from *Arracacha xanthorriza*, *Canna tuberosa* and *Oxalis tuberosa* (Santacruz, Koch, Svensson, Ruales, & Eliasson, 2002).

Foods that contain starch are in the majority of cases heated in the presence of water. This results in the gelatinisation of the starch, producing favourable changes in the appearance and texture of the food. The changes in rheological behaviour during heating of a starch suspension are thus considerable, and rheological data may therefore be very helpful both in evaluating the behaviour of starch, and for the development of new products with controlled rheological behaviour (Steeneken, 1989). Starch concentration, rigidity of the starch particles and the rheological characteristics of the matrix phase are the main parameters that influence the rheology of starch gels (Doublier, 1990; Eliasson & Bohlin, 1982; Evans & Haisman, 1979). Storage of food containing gelatinised starch, either in refrigeration or freezing conditions, often produces undesirable changes in the texture (Kulp & Ponte 1981). Some of these changes are related to the retrogradation process, i.e. changes from an amorphous to a more ordered or crystalline state (Eliasson & Gudmundsson, 1996). The rheological properties will change, and increase of firmness or rigidity is measured. Retrogradation depends on time and temperature and, moreover, the retrogradation of native starch gels is affected by a serie of factors, such as the amylopectin/amylose ratio, percentage of water, lipids, pH and botanical source (Collison, 1968).

The rheological properties are also sensitive to pH, and under acid treatment, most native starches lose their

* Corresponding author. Fax: +46-46-222-9517.
E-mail address: ann-charlotte.eliasson@livsteki.lth.se (A.C. Eliasson).
[1] Present address: Department of Food Science, Swedish University of Agricultural Sciences, P.O. Box 7051, S-750 07 Uppsala, Sweden.

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Table 1
Swelling power, amylose leaching, amylose content and granule size, together with close-packing concentration, rigidity of fully swollen granules and rigidity index of the native starches *A. xanthorriza*, *C. edulis* and *O. tuberosa*

Starch	Amylose content[a,b] (%)	Swelling power[b] (g/g)	Solubles[b,c] (%)	Amylose leaching[b] (%)	Granule size[a] (μm)	Close-packing concentration[b] (%)	Rigidity[b,d,e] (Pa)	Rigidity index[b] (G/c) (Pa ml g^{-1})
A. xanthorriza	4.0	57.9	2.6	2.3	7–23	1.8	12.5	610
C. edulis	23.8	65.5	21.4	16.9	35–101	1.7	25.8	2720
O. tuberosa	18.4	63.4	15.8	14.4	22–55	1.7	14.2	2880

[a] Santacruz et al. (2002).
[b] $n = 2$.
[c] Drying at 120 °C of a supernatant of a starch suspension heated previously at 89 °C.
[d] Rigidity of fully swollen granules.
[e] Elastic modulus G' of the starch gel at a particle volume fraction of 1.

thickening ability (Fennema, 1985). These changes vary with the botanical source of the starch, and in the manufacturing of products such as jelly gum candies, a more acid tolerant starch would be useful.

The present work is the second part of the physico-chemical characterisation of *A. xanthorriza*, *C. edulis* and *O. tuberosa* tuber starches (Santacruz et al., 2002). In the first part it was found that all these starches exhibited a B type of X-ray diffraction pattern, and that the starch granule sizes were between 35 and 101 μm for *C. edulis*, 20 and 55 μm for *O. tuberosa* and 7 and 23 μm for *A. xanthorriza*. Amylose content was highest for *C. edulis* with, 23.8%, followed by *O. tuberosa* (18.4%) and *A. xanthorriza* (4%). The chain length distribution of amylopectin showed different structures among the three starches, with a β-amylolysis limit of 67.6% for *C. edulis*, 64.5% for *O. tuberosa* and 56.6% for *A. xanthorriza*. Comparison of gelatinisation results for the three starches showed that *C. edulis* and *A. xanthorriza* had a higher temperature of gelatinisation than *O. tuberosa*. In the present study, the rheological properties of starch gels from these starches, as influenced by storage conditions and pH were investigated.

2. Materials and methods

2.1. Materials

Tubers of *A. xanthorriza*, cultivar FB-001, and *C. edulis*, cultivar MH-1173, were both identified and obtained from the International Potato Centre (CIP), Quito, Ecuador, whereas *O. tuberosa* was bought directly at the local market in Quito, Ecuador. The starches were extracted as described elsewhere (Santacruz et al., 2002). Double-distilled water was used in all the experiments. Citric acid was used to regulate the pH of the starch suspensions.

2.2. Determination of swelling power and amylose leaching

The swelling power and amylose leaching were determined according to the modified method of Schoch (1964). Starch suspensions (0.5% w/w, dry matter) were heated in test tubes at 89 °C (91 °C boiling point in Quito, Ecuador) and carefully agitated by hand to avoid sedimentation at the beginning of the heating process. The tubes were kept in a water bath at 89 °C for 30 min, and centrifuged at 1000 × g for 15 min in a Sorvall RC 5 (Connecticut, USA). The sediment fraction was weighed and its mass related to the mass of dry starch was expressed as swelling power (w/w). The amount of released amylose was measured as the absorbance of the supernatant, according to the blue value method of Gilbert and Spragg (1964), by means of a Beckman DU-50 Spectrophotometer (Beckman, USA). Amylose from potato (Sigma A-0512, USA) was used for the calibration curve. The quantity of solubles was obtained by drying the supernatant at 120 °C overnight in a heating cabinet. The results given are the mean values of two measurements. In no case did the difference between the extremes exceed 6% of the mean value.

2.3. Determination of the close-packing concentration

The close-packing concentration was determined according to Steeneken (1989). The swelling power (q) was defined as the volume occupied by 1 g of dry starch after swelling to equilibrium in excess solvent. At a low particle volume fraction (cq, where c is the concentration of the starch in the suspension), the granules are completely swollen. These systems are not homogeneous, in the sense that the internal starch concentration is higher than the external one. An increase in the starch particle volume fraction leads to a situation where the fully swollen granules just fill the available space. At this space-filling concentration, the system becomes homogeneous, and the particle volume fraction has the value 1 ($cq = 1$). This specific concentration is the close-packing concentration (c_{cp}), which is equal to $1/q$, ($c_{cp} = 1/q$) (Steeneken, 1989). The close-packing concentration results given are the mean values of two measurements.

2.4. Preparation of starch gels

Starch suspensions of particle volume fractions (cq) between 1 and 3 were prepared by manually shaking

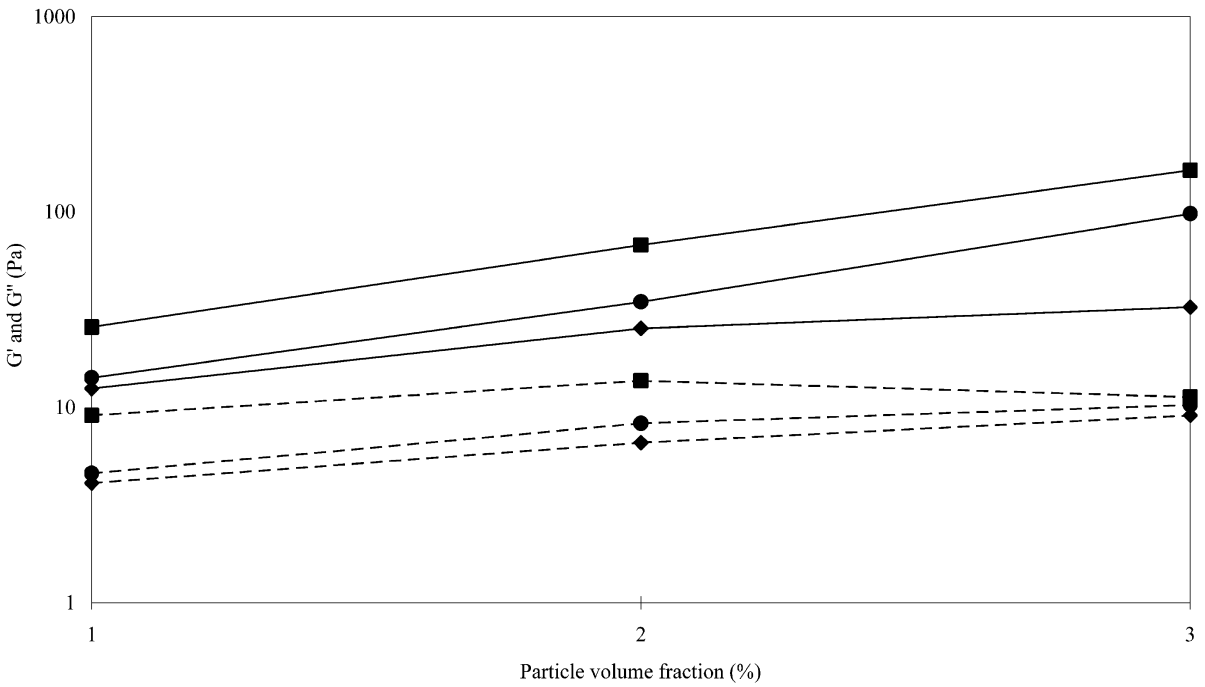

Fig. 1. Elastic (G') and viscous (G'') moduli at different particle volume fractions. (◆) *A. xanthorriza*, (■) *C. edulis*, (•) *O. tuberosa*. Solid lines G' and dotted G''. Measurements at 2 Hz.

appropriate amounts of starch and distilled water in tubes in a water bath at 89 °C for 30 min. The amount of starch was based on the close-packing concentration calculated previously. The prepared samples were stored either at 4 °C in a cool room or at −20 °C in a freezer room for up to 3 days. The stored samples were thawed at room temperature (25 °C) for 2 h before the measurements were made. Measurements were made at 0, 1, 2 and 3 days. When the effect of pH was studied, starch suspensions at close-packing concentration were prepared as previously described. Three pH values, 4, 5 and 6.5, were used for the analyses. The pH of the suspension was measured by using a pH meter (PHM 85, Radiometer, Copenhagen) and adjusted to the chosen pH with citric acid. The samples were stored at 4 °C in a cool room for up to 3 days, and measurements were made at 0, 1, 2 and 3 days.

2.5. Rheological measurements

Strain-sweep measurements were made using a plate and plate system, gap 1 mm, either with torsion bars of 0.245 or 1.60 g cm in a VOR Bohlin rheometer (Metric Analysis, Stockholm, Sweden). The elastic modulus (G'), viscous modulus (G'') and phase angle (δ) were evaluated from the rheological measurements. G', G'' and δ were taken from the linear region of the strain-sweep measurements. The results given are the average and standard deviation of at least three measurements. The strain range was from 0.1 to 100%. The frequency was constant and equal to 2 Hz. The sample was loaded into the rheometer and allowed to stabilise for 15 min before the measurement started. A thin layer of silicon oil was added on top of the sample to prevent loss of moisture.

The elastic modulus G' of the starch gel at a particle volume fraction of 1 is defined as rigidity or deformability (Steeneken, 1989). The rigidity is constant, and characteristic of dilute regime, i.e. particle volume fraction <1. However, at particle volume fractions above the close-packing concentration, the rigidity becomes concentration-dependent (Steeneken, 1989), and is represented by the rigidity index (G/c) as follows:

$$(G/c) = (G' - G'_{cp})/(c - c_{cp})$$

where G'_{cp} is G' at close-packing concentration.

The results given are the average and standard deviation of two measurements. The standard deviation of 36% of the measurements was above 10% of the average.

Table 2
Light transmittance ($\lambda = 650$ nm) of starch gels (volume fraction $cq = 2$) stored at 4 °C

Time (days)	*A. xanthorriza* (%)	*C. edulis* (%)	*O. tuberosa* (%)
0	67	76	74
1	48	2	2
2	47	2	2

$n = 2$.

2.6. Light transmittance of starch gels

Light transmittance measurements were performed

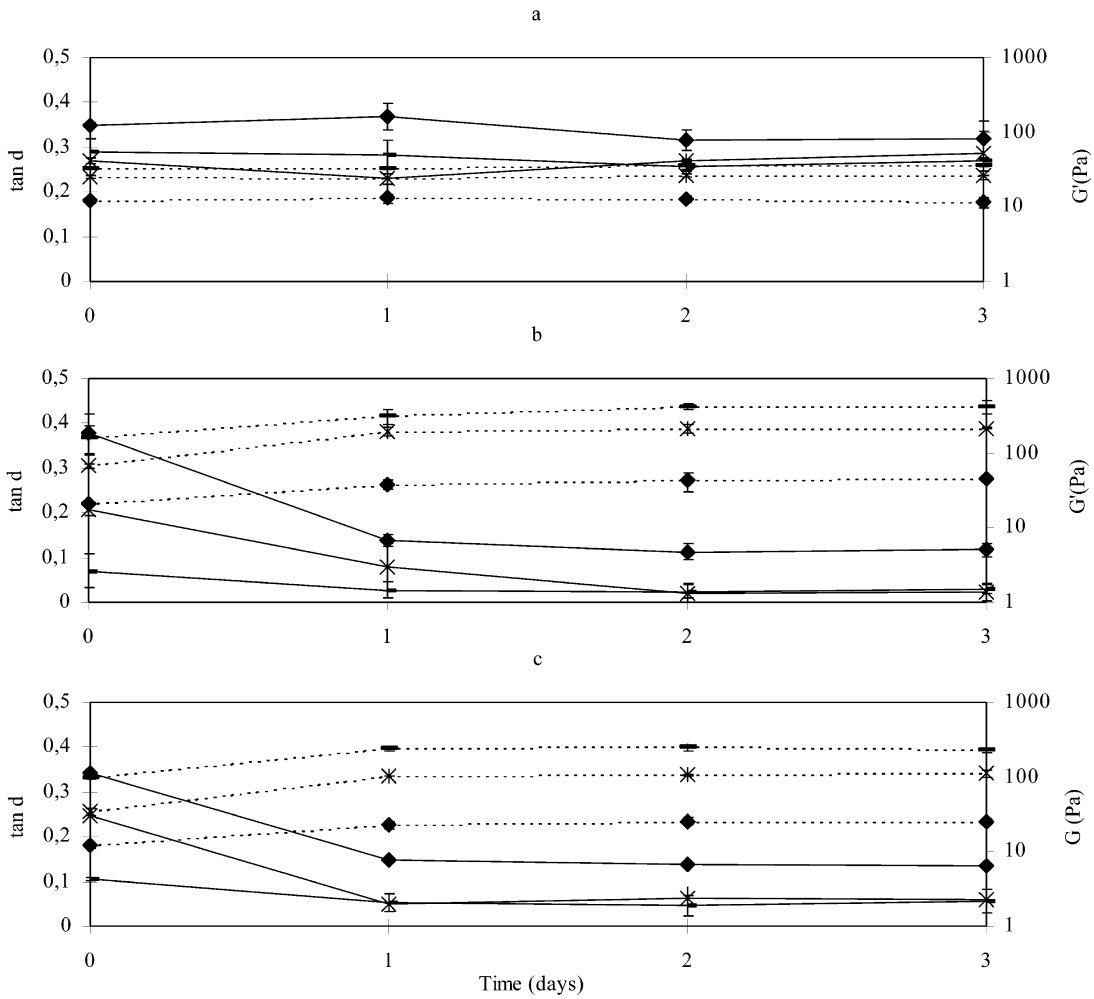

Fig. 2. Changes in the elastic modulus, G', and tangent of phase angle, tan δ, of (a) *A. xanthorriza*, (b) *C. edulis* and (c) *O. tuberosa* starch gels during storage at 4 °C, (◆) *cq* 1, (∗) *cq* 2 and (—) *cq* 3. Dotted lines G' and solid lines tan δ. Measurements at 2 Hz.

according to the modified method of Craig, Maningat, Seib, and Hoseney (1989). Starch suspensions at volume fractions of 2 were prepared by manually shaking appropriate amounts of starch and distilled water in a tube on a water bath at 89 °C for 30 min. The starch gels were studied after 0, 1 and 2 days of storage at 4 °C by means of a Shimadzu spectrophotometer (Japan) at $\lambda = 650$ nm. The results given are the mean of two measurements. The difference between the duplicates did not exceeded 10% of the mean value.

2.7. Differential scanning calorimetry

The influence of the change of pH on the gelatinisation of starch was analysed according to Santacruz et al. (2001) on a Seiko SII 6200 DSC (Seiko, Japan) equipped with standard software. The samples were investigated at a water/starch ratio of 2:1 in coated pans from TA Instruments (TA Instruments, USA). Enough citric acid was added to obtain a starch solution of pH 3 (approximately 30% starch basis). A pan containing Al_2O_3 was used as a reference. The

Table 3
Swelling power and rigidity of starch gels at close packing concentration to different pH

	A. xanthorriza		*C. edulis*		*O. tuberosa*	
pH	Swelling power	Rigidity (Pa)	Swelling power	Rigidity (Pa)	Swelling power	Rigidity (Pa)
4.0	46.2	6.3	45.8	8.6	41.3	6.3
5.0	n.d.	9.9	n.d.	11.3	n.d.	6.9
6.5	52.5	12.1	54.9	16.6	58.4	10.5
8.4	54.7	n.d.	55.3	n.d.	60.0	n.d.

n.d.: not determined; $n = 2$.

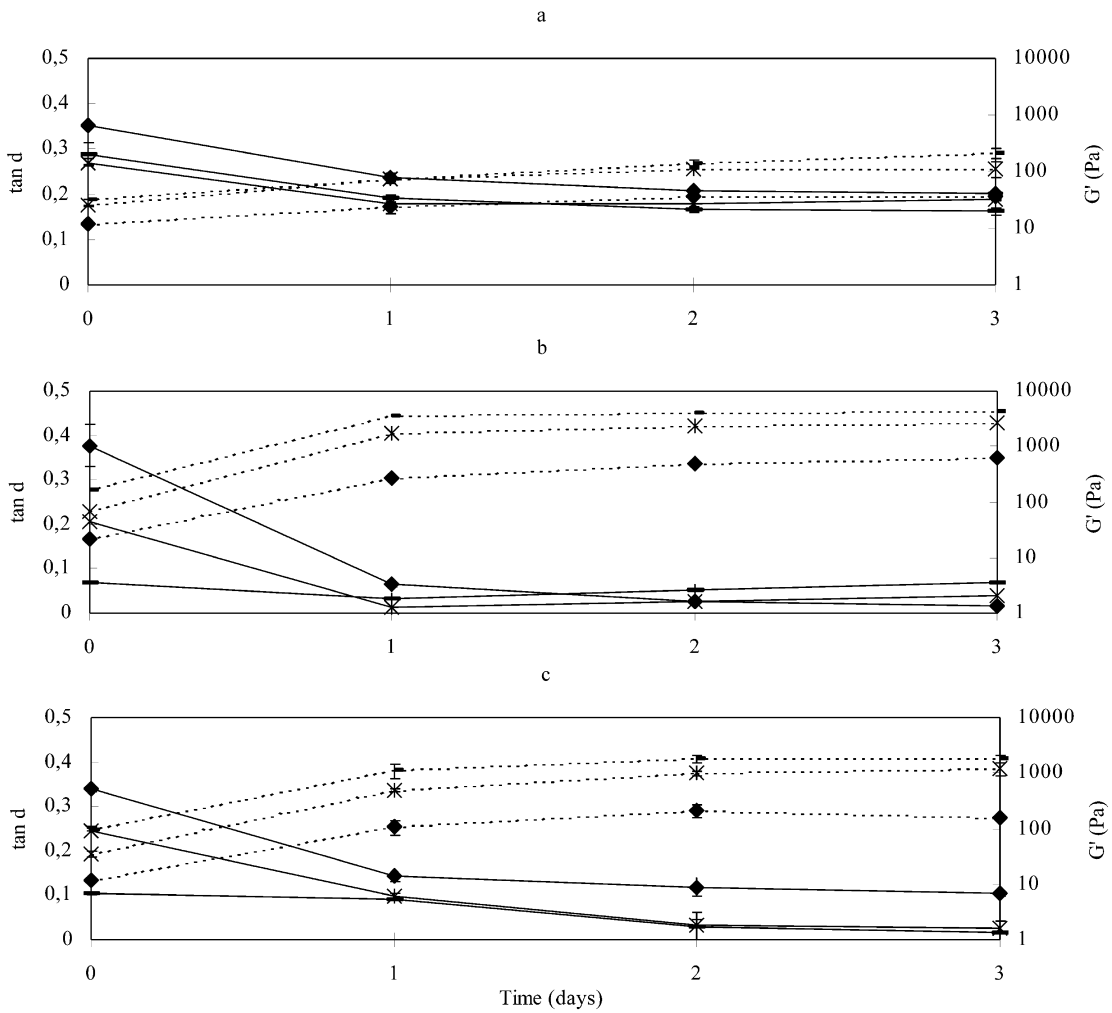

Fig. 3. Changes in the elastic modulus, G', and tangent of phase angle, tan δ, of (a) *A. xanthorriza*, (b) *C. edulis* and (c) *O. tuberosa* starch gels during storage at 20 °C, (◆) *cq* 1, (∗) *cq* 2 and (—) *cq* 3. Dotted lines G' and solid lines tan δ. Measurements at 2 Hz.

enthalpies of gelatinisation were calculated on dry-matter basis, which was determined by puncturing and drying the pans in an oven at 105 °C for 2 h.

2.8. Statistical analysis

The analysis of the rheological properties of the three starch gels were performed by using STATGRAPHS computer software. ANOVA test or regression analysis were used to determine the significance of the affecting factors on G'.

3. Results

3.1. Swelling and amylose leaching

The changes of *A. xanthorriza*, *C. edulis* and *O. tuberosa* starches during the gelatinisation process resulted in the highest swelling power and amylose leaching at 89 °C for *C. edulis*, followed by *O. tuberosa* and *A. xanthorriza* (Table 1). The swelling power, amylose leaching and percentage of solubles showed high positive correlation with the amylose content (correlation coefficients > 0.98). On the basis of the swelling power results, the close-packing concentrations were calculated according to Steeneken (1989). These close-packing concentrations were similar among the three starches, *A. xanthorriza* having a somewhat higher value, 1.8%, followed by *O. tuberosa*, and *C. edulis*, both of which had 1.7%. (Table 1).

3.2. Concentration regimens

All dynamic measurements were performed in the linear strain region, where the viscoelastic parameters are independent of the deformation. G' and G'' increased with the increase of particle volume fraction (Fig. 1). G' of *C. edulis* was the highest among the three starches, followed by *O. tuberosa* and *A. xanthorriza*. The difference between G' and G'' increased with the increase in particle volume fraction, showing a more rapid increment of G' for the three starches.

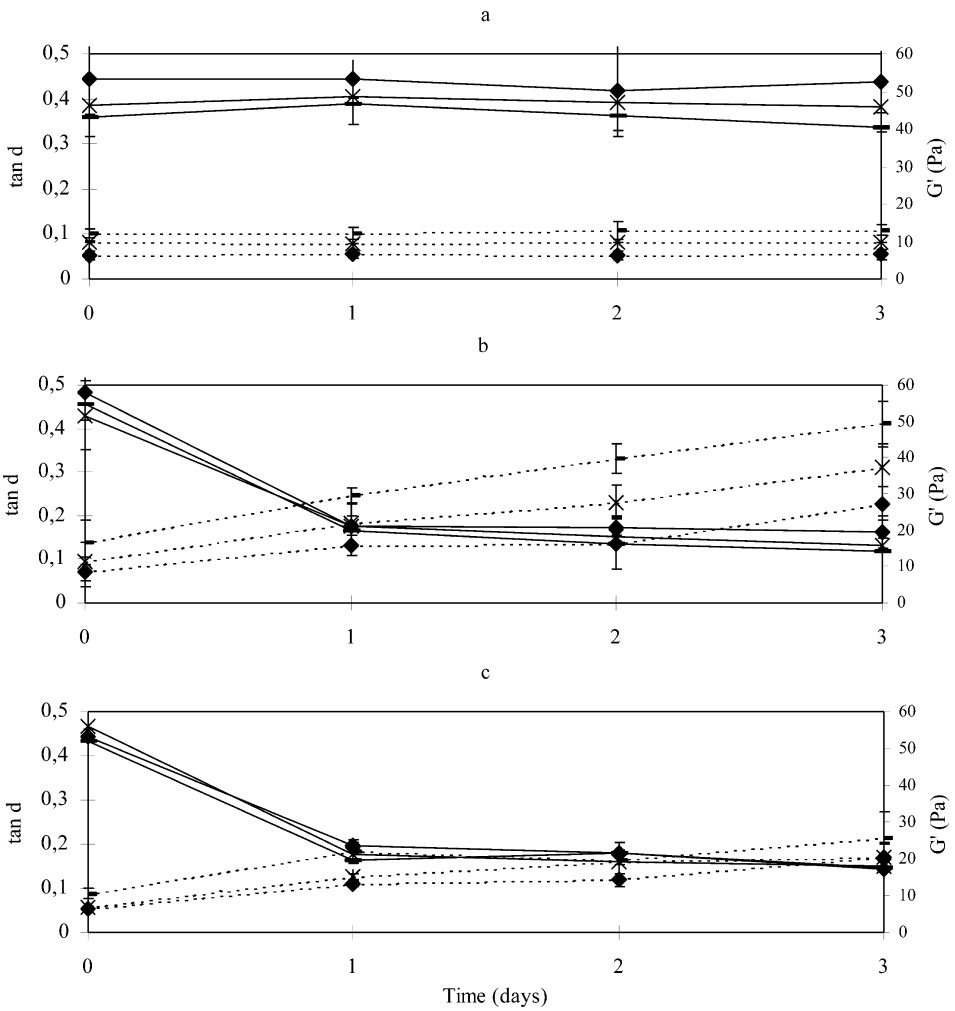

Fig. 4. Changes in the elastic modulus, G', and tangent of phase angle, tan δ, of (a) *A. xanthorriza*, (b) *C. edulis* and (c) *O. tuberosa* starch gels during storage at 4 °C at different pH, (◆) pH 4, (∗) pH 5 and (—) pH 6.5. Dotted lines G' and solid lines tan δ. Measurements at 2 Hz.

3.3. Concentration dependence of rigidity

The rigidity of *C. edulis* was the highest in the dilute regime, 25.8 Pa, followed by *O. tuberosa* and *A. xanthorriza*, with 14.2 and 12.5 Pa, respectively (Table 1). At particle volume fractions > 1, the rigidity, expressed by the rigidity index, increased more rapidly for *O. tuberosa* and *C. edulis* starch gels than for *A. xanthorriza* (Table 1). Regression analysis of data of starch gels prepared at particle volume fractions from 1 to 3 showed that G' had a high dependence on rigidity and particle volume fraction (data not shown). Similar conclusions were obtained for corn starch (Tsai, Li, & Lii, 1997).

3.4. Storage of starch gels under refrigeration

Fig. 2 shows the changes in G' and tan δ for starch gels at pH 6.5 stored at 4 °C. *A. xanthorriza* starch gels of particle volume fractions between 1 and 3 showed only minor changes in the G' and tan δ during 3 days of storage. *C. edulis* and *O. tuberosa* starch gels of particle volume fractions between 1 and 3 showed an increase in G' during storage at 4 °C. The tan δ of *C. edulis* and *O. tuberosa* decreased rapidly on the first day of storage at 4 °C, after which there were only minor changes in tan δ. The decrease in tan δ was slower for the higher volume fractions.

Storage of *A. xanthorriza*, *C. edulis* and *O. tuberosa* starch gels at 4 °C reduced the light transmittance of the gels (Table 2). Changes in the light transmittance of starch gels from *C. edulis* and *O. tuberosa* were higher than those for *A. xanthorriza*. The highest reduction in the light transmittance of *C. edulis* and *O. tuberosa* was shown after 1 day of storage, from 76 to 2% and from 74 to 2%, respectively. Light transmittance of *A. xanthorriza* starch gel was reduced from 67 to 48% after 1 day of storage. The storage of starch gels during 2 days at 4 °C reduced the light transmittance to 47% for *A. xanthorriza* and to 2% for *C. edulis* and *O. tuberosa.*

3.5. Storage of starch gels under freezing conditions

The changes in G' and tan δ during frozen storage (-20 °C) and thawing is shown in Fig. 3. Separation of

water (syneresis) after thawing the starch gels was not observed. The G' of all three starches increased during 3 days of storage. The tan δ of *A. xanthorriza*, *C. edulis* and *O. tuberosa* decreased rapidly during the first day of storage at -20 °C, after which there were only minor changes in tan δ.

The higher is the particle volume fraction, the lower is the decrease in tan δ during the first day of storage (Fig. 3). Starch gels stored at -20 °C had a higher G' during the storage period than similar gels stored at 4 °C.

3.6. Effect of pH on storage under refrigeration

The changes in G' and tan δ of the three starch gels at $cq = 1$ and stored at 4 °C at pH 4, 5 and 6.5, are shown in Fig. 4. Changes of pH from 6.5 to lower values produced a reduction in the G' and an increase in tan δ in the three starch gels. The G' and tan δ of *A. xanthorriza* starch gels showed only minor changes during 3 days of storage at 4 °C. G' of *C. edulis* and *O. tuberosa* increased during the 3 days of storage, whereas tan δ of *C. edulis* and *O. tuberosa* decreased rapidly on the first day of storage at 4 °C.

Lowering of pH from 8.4 to 4.0 (Table 3) reduced swelling power and rigidity. DSC data indicated that enthalpy and temperatures of gelatinisation were not affected by the addition of citric acid (data not shown).

4. Discussion

A more rapid increase in G' compared to G'' with the increase in volume fraction revealed that the starch concentration had a higher effect on the elastic behaviour of the three starch gels than on the viscous behaviour. The rigidity of the starch granules was positively correlated with swelling power and amylose content (Table 1). The amorphous regions of the starch, which are mainly amylose and the branching points in amylopectin, could reinforce the structure of the granule, thus increasing the rigidity. Similar conclusions were drawn by Wong and Lelievre (1981) and by Sandhya Rani and Bhattacharya (1989). However, Lii, Tsai, and Tseng (1996) suggested that rigidity might be inversely proportional to the swelling power. The reason for this opposite conclusions may be that the use of different species of starch involves differences in the amylopectin fractions (Tester & Morrison, 1990).

A high thickening power of a starch gel requires a high particle rigidity index (G/c) for concentrations above the close-packing concentration. However, those starches that exhibit a high viscosity at a low concentration are of great interest, because the utilisation of minor quantities of those starches leads to economic benefits. To meet both requirements, rigidity index (G/c) and swelling power must be as high as possible (Steeneken, 1989). *C. edulis* showed both high swelling power and high rigidity index.

Besides the increase of G' during storage at 4 and -20 °C, a change of texture of the starch gels was observed. The altered texture was described as lumpy, grainy or sticky (Albrecht, Nelson, & Steinberg, 1960; Schoch, 1968). The major changes in G' and tan δ were produced in *C. edulis* and *O. tuberosa* starch gels during the first day of storage at both 4 and -20 °C. The changes during the first day of storage must be produced by retrogradation of the amylose fraction, reinforcing the structure of the system during cooling and ageing (Hamann, Purkayastha, & Lanier, 1990; Lii et al., 1996; Tsai et al., 1997). After the first day of storage, only minor changes in G' and tan δ were observed. The initial rapid retrogradation in *C. edulis* and *O. tuberosa* starch gels stored at 4 °C was also revealed by light-transmittance analysis (Table 2). Doublier (1990) and Jacobson, Obanni, and BeMiller (1997) reported that changes in the light transmittance of starch gels from amylose-containing starches, e.g. *C. edulis* and *O. tuberosa* (Table 1), could be the result of a transformation from an amylose network structure to a dense aggregated state.

C. edulis and *O. tuberosa*, with higher rigidity indexes than *A. xanthorriza*, showed a higher increase in G' during storage. Higher rigidity and volume fractions lead to higher G'. However, G' must depend not only on rigidity; leached amylose could strengthen the gel during cooling and ageing (Evans & Haisman, 1979; Lii et al., 1996).

A reduction of tan δ to values close to zero indicates the formation of a purely elastic network, and could be used as a marker for gel formation. The gel formation could be due to the presence of amylose, as in *C. edulis* and *O. tuberosa* starches. On the other hand, *A. xanthorriza* did not show a reduction in tan δ during storage.

A decrease in pH produced a loss of structure in the three starch gels, as shown by the increase in tan δ and reduction of G'. Muhrbeck and Eliasson (1987) made similar observations on potato starch gels. DSC results revealed that pH did not affect temperature and enthalpy of gelatinisation. However, the reduction of pH affected the gelatinisation process by diminishing the swelling power.

The present study shows that *A. xanthorriza* starch gels exhibited only minor changes in G' and tan δ, compared with *C. edulis* and *O. tuberosa*, during storage under refrigeration. However, all three starches were affected during storage under freezing conditions.

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