



# Thermal and mechanical properties of concentrated rice starch gels of varying composition

Costas G. Biliaderis

Department of Food Science, University of Manitoba, Winnipeg, MB, R3T 2N2, Canada

&

Bienvenido O. Juliano

International Rice Research Institute, PO Box 933, 1099 Manila, Philippines

(Received 22 September 1992; accepted 23 February 1993)

The thermal and mechanical properties of concentrated (25–35% w/w) starch gels of 43 non-waxy rices, varying in their amylose content and gelatinization characteristics, were investigated by small amplitude oscillatory shear measurements and differential scanning calorimetry. The storage modulus ( $G'$ ) of freshly prepared gels increased exponentially with increasing amylose content. An inverse linear relationship was also found between  $\tan \delta$  and amylose content ( $r = -0.75$ ,  $p < 0.01$ ). All high-amylose (>29%) rice starch gels exhibited  $\tan \delta$  values less than 0.1, indicating well cross-linked networks. At starch concentrations within 10–25% (w/w), the dependence of storage modulus on concentration followed power law relationships;  $G'$  varies as  $C^{2.2-2.9}$  ( $r > 0.96$ ,  $p < 0.01$ ). Changes in gel rigidity ( $G'$ ) upon storage were not solely dependent on amylose content. Kinetic experiments on staling of gels (35% w/w, 8°C) by calorimetry indicated that granular lipids retarded this process; the effect was more pronounced for low-amylose rice starches.

## INTRODUCTION

Considerable variation in cooking and eating qualities of milled rice has been reported (Juliano, 1985a). It is generally accepted that cooking quality of rice is strongly influenced by the composition and physical properties of the starch component. More specifically, amylose content, gelatinization characteristics and granular lipids have been identified as the main quality determinants of cooked rice (Juliano, 1979a,b; Morrison & Azudin, 1987). Breeding programs aiming at improvement of grain quality have resulted in many genetically distinct lines of different amylose content and starch gelatinization properties. Thus, various rices with specific amylose-amylopectin ratios are grown and used for different rice products around the world. However, it must be noted that amylose content and structural organisation of starch granules are also affected by environmental conditions (Asaoka *et al.*, 1985a,b). Moreover, rice grains undergo textural changes during storage which have an imprint on end-product quality. Such effects are important in the implementation of effective quality assessment tests.

Most of the developed methods for texture evaluation of cooked rice involve measurements of hardness, stickiness or consistency by either instrumental (e.g. Instron tester) or sensory testing (IRRI, 1979). Although sensory and Instron texture analyses of the same rice samples do not always correspond (Del Mundo *et al.*, 1989), the use of objective methods is preferred for convenience and because it allows a description of texture in fundamental units. Furthermore, mechanical tests provide a means of monitoring textural changes in rice products upon cooking, cooling and storage.

Undeniably, the pasting and gelation behaviour of starch in rice has a major impact on texture of the cooked product. Rice starch exists as polyhedral granules with a mean size diameter of 4–7  $\mu\text{m}$  (Juliano, 1985b). Unlike other cereal starches, rice starch consists of a single-size granule distribution. Therefore, rice starch makes an excellent model system to study the effect of composition and physical characteristics of the granules on the thermomechanical properties of starch gels. The objective of this study was to examine the rheological and thermal properties of 43 purified non-waxy starches, isolated from various cultivars, for the purpose of identifying relationships between physical properties and composition of these materials. The varieties chosen represent a wide array of cooking

qualities, while the isolated starches gave a broad range of amylose contents and gelatinization temperatures. Gels of high starch concentration (25% w/w) were prepared to resemble the actual solids level of cooked rice (60–75% moisture). The viscoelastic properties and structural changes in thermoset gels were monitored by small strain dynamic rheometry and differential scanning calorimetry. It is believed that such studies could lead to a better understanding of how starch composition (amylose content, lipids, etc.) and molecular interactions between constituents affect the mechanical properties of rice-based products.

## MATERIALS AND METHODS

### Rice starches

Starches were prepared from milled rice or milled rice flour by homogenizing (after soaking in water) in a Waring blender at medium speed for 1 min and passing through a 170-mesh screen. Some of the starches were extracted (4–5 ×) with several volumes of 1.2% sodium dodecylbenzene sulfonate (DoBS) containing 0.12% Na<sub>2</sub>SO<sub>3</sub> until the extract was negative to the biuret test (Maniñgat & Juliano, 1980). The starch was then washed repeatedly with water and centrifuged (2500 g) until the wash water was neutral and then air-dried at 25–27°C. Some starches were purified by enzymic hydrolysis of proteins using Pronase (alkaline protease from *Streptomyces griseus*) according to Maniñgat and Juliano (1980). Wet-milled flour was treated twice with five volumes of 0.2% Pronase (45,000 U g<sup>-1</sup>, Calbiochem, San Diego, CA) in 0.03 M phosphate buffer, pH 7.4, for 24 h at 37°C using toluene as preservative, washed with water and air-dried. Pronase treatments produce starch with intact granular lipids, while the DoBS replaces some of the internal starch lipids (Maniñgat & Juliano, 1980).

Amylose was determined according to the method of Juliano *et al.* (1981) using potato amylose-waxy rice starch standard mixtures. For quantitation of non-starch lipids, extraction of granules (2 ×) at ambient temperature (30 min) was carried out with five volumes of water-saturated 1-butanol (WSB); the granules were recovered by centrifugation (2000 g; 10 min). The internal granular lipids were subsequently extracted by refluxing (92°C, 2 h) with five volumes of WSB (three extractions). The extracts in each case were pooled, evaporated to dryness under vacuum, taken up with CHCl<sub>3</sub>, dried with anhydrous Na<sub>2</sub>SO<sub>4</sub>, filtered, evaporated to dryness and weighed. Some starches were defatted (hot extraction with 85% v/v methanol, 48 h) to determine the effect of internal lipids on starch retrogradation.

### Gelatinization temperature, gel viscosity and consistency

Gelatinization temperature (*GT*) of starch was determined as the temperature at which 90–98% of the granules lose their birefringence when heated on a microscope hot stage (Schoch & Maywald, 1956). Gel

consistency of rice starch was determined according to Cagampang *et al.* (1973) using 160 mg starch and 2.0 ml 0.2 M KOH. The viscosity of gels was taken with a Wells–Brookfield cone and plate microviscometer Model RVT with 1.565° and 4.8 cm cone diameter at 30°C and 25 rpm.

### Dynamic rheological testing

Starch gels were prepared by heating starch slurries at a specified concentration in a hermetically sealed stainless steel container with dimensions 80 mm (internal diameter) × 1.0 mm thickness (Biliaderis & Tonogai, 1991). Using this device, 1-mm thick gels were cast (by immersion in boiling water for 15 min, followed by rapid cooling in a water bath at 25°C, for 15 min) without loss of water or mechanical damage of the gel network. Gel discs (30 mm diameter) were cut to fit the parallel plate geometry of the rheometer. For gels of low-amylose starches, because of their viscous character, gelation was carried out in sealed stainless steel cylinders (125 mm i.d. × 33 mm length) following the same heating–cooling regimes. A certain amount of gel material was forced to attain the geometry of the gap by lowering the upper plate of the rheometer onto the sample to a preset gap width. The samples were allowed to relax for 5 min before measurements were taken. To minimize contributions from retrogradation events, all rheological tests were carried out immediately after gelation.

A Bohlin VOR rheometer (Bohin Reologi Inc., Edison, NJ) equipped with a parallel plate geometry (30 mm diameter) and a torque element of 93.2 g·cm was employed for the small amplitude oscillatory rheological tests. Measurements were made at 25°C over a frequency range of 0.1–20.0 Hz and strain less than 4.0% in all cases; however, data comparisons were made using values obtained at a specified frequency. The dynamic rheological parameters used to evaluate the gel networks were: storage modulus,  $G'$ ; loss modulus  $G''$  dynamic viscosity  $\eta' = G''/2\pi f$  ( $f$  is frequency); and  $\tan \delta$  or loss tangent ( $\tan \delta = G''/G'$ ).

The kinetic aspects of structure development in aging gels (25% w/w starch concentration, 8°C) were monitored over a period of 17.5 h at 0.2 Hz and strain less than 2.0%; data were collected at 30-min intervals. In these experiments, after loading the gel on the geometry, a thin layer of paraffin oil was used to cover the sample and prevent moisture loss due to evaporation during storage (Biliaderis & Zawistowski, 1990). Preliminary work indicated negligible contributions to the rheological parameters from the oil itself and that there is no slip between plate and sample surfaces. All dynamic tests were performed at low enough strains to ensure linear viscoelasticity.

### Calorimetry

Differential scanning calorimetry (DSC) of starches was performed on a Dupont 9900 Thermal Analyzer equipped with a 910 DSC pressure cell (pressure 1400

kPa with N<sub>2</sub>). Aqueous starch suspensions (35% w/w, 3.65–3.85 mg starch) were sealed in hermetic DSC pans. For the staling studies, the starch samples were first heated to 140°C (10°C/min) to melt the granules, then cooled and stored at 6°C for a designated time (4, 16, 48, 98 and 168 h) before DSC analysis of the staling process was carried out. All other conditions for calibration and operation of the calorimeter were essentially the same as described by Biliaderis *et al.* (1985).

## RESULTS AND DISCUSSION

### Composition and properties of rice starches

The amylose content and physical properties of the 43 granular starches used in this study are summarized in Tables 1, 2 and 3; a broad range of amylose content and gelatinization characteristics was found among the samples. Surprisingly, hard gel consistency (low values, 26–40 mm) was observed even among low-amylose

**Table 1. Properties of purified low-amylose rice starches prepared by either DoBS or alkaline protease treatment**

| Sample                                      | Method   | Amylose (%) | Final <i>GT</i> (°C) | <i>Tm</i> <sup>a</sup> (°C) | $\Delta H$ <sup>a</sup> (J g <sup>-1</sup> ) | Gel properties   |                   | Storage modulus <i>G'</i> (Pa) <sup>b</sup> | tan $\delta$ <sup>b</sup> |
|---|----------|-------------|----------------------|-----------------------------|--|------------------|-------------------|---|---------------------------|
|   |          |             |                      |                             |  | Consistency (mm) | Viscosity (cP)    |   |                           |
| IR37307-8-ch                                | Protease | 9.8         | 70                   | 68.9                        | 12.7   | 39 <sup>c</sup>  | 4350 <sup>c</sup> | 502 ± 29                                    | 0.21                      |
| IR841-67-1                                  | DoBS     | 18.3        | 69                   | 69.4                        | 12.1   | 30               | 4970              | 1393 ± 96                                   | 0.21                      |
| Khao Dawk Mali 105                          | DoBS     | 18.4        | 64                   | 67.1                        | 11.6   | 84               | 1170              | 193 ± 13                                    | 0.35                      |
| IR24  | DoBS     | 19.5        | 66                   | 66.5                        | 12.8   | 84               | 1720              | 412 ± 92                                    | 0.24                      |
| IR43  | DoBS     | 21.3        | 66                   | 66.6                        | 15.9   | 75               | 1780              | 828 ± 38                                    | 0.23                      |
| Akibare                                     | DoBS     | 23.6        | 68                   | 65.0                        | 14.5   | 66               | 1450              | 487 ± 86                                    | 0.23                      |
| Milyang 23                                  | DoBS     | 23.4        | 66                   | 64.4                        | 12.5   | 60               | 1450              | 377 ± 25                                    | 0.24                      |
| Tongil                                      | DoBS     | 24.0        | 65                   | 65.4                        | 9.9  | 43               | 3220              | 861 ± 50                                    | 0.16                      |
| IR2071-137-5                                | DoBS     | 16.0        | 75                   | 77.7                        | 14.0   | 58               | 1760              | 182 ± 8                                     | 0.36                      |
| Century Patna 231                           | DoBS     | 17.4        | 76                   | 76.9                        | 14.9   | 44               | 2445              | 204 ± 12                                    | 0.25                      |
| Coefficient of variation<br>( <i>n</i> = 3) |          | <1.2%       | <1.4%                | <0.6%                       | <3.8%  |                  |                   |   |                           |

<sup>a</sup> From DSC measurements at 35% (w/w) starch concentration; C.V. of *Tm* (peak temperature) and  $\Delta H$  did not exceed 0.30% and 4.0%, respectively.

<sup>b</sup> Values of 25% (w/w) starch gels (25°C) at 1.0 Hz (*n* = 3).

<sup>c</sup> Means of duplicate measurements; the individual values differ by less than 10.2% (consistency) and 9.6% (viscosity) of the mean.

**Table 2. Properties of purified intermediate-amylose rice starches prepared by either DoBS or alkaline protease treatment**

| Sample                                      | Method   | Amylose (%) | Final <i>GT</i> (°C) | <i>Tm</i> <sup>a</sup> (°C) | $\Delta H$ <sup>a</sup> (J g <sup>-1</sup> ) | Gel properties   |                   | Storage modulus <i>G'</i> (Pa) <sup>b</sup> | tan $\delta$ <sup>b</sup> |
|---|----------|-------------|----------------------|-----------------------------|--|------------------|-------------------|---|---------------------------|
|   |          |             |                      |                             |  | Consistency (mm) | Viscosity (cP)    |   |                           |
| Arlesienne                                  | DoBS     | 29.2        | 62                   | 57.6                        | 11.3   | 34 <sup>c</sup>  | 2300 <sup>c</sup> | 2348 ± 170                                  | 0.10                      |
| IR480-5-9                                   | DoBS     | 29.2        | 61                   | 60.9                        | 9.7  | 36               | 1650              | 1526 ± 80                                   | 0.10                      |
| IR48a                                       | DoBS     | 27.3        | 64                   | 62.6                        | 11.1   | 40               | 1400              | 1070 ± 62                                   | 0.17                      |
| IR48b                                       | DoBS     | 30.8        | 62                   | 63.9                        | 14.7   | 31               | 2350              | 1210 ± 108                                  | 0.15                      |
| BPI-121-407                                 | DoBS     | 30.7        | 62                   | 63.9                        | 9.9  | 34               | 2020              | 1463 ± 58                                   | 0.14                      |
| D25-4                                       | DoBS     | 23.5        | 70                   | 71.6                        | 12.3   | 60               | 2020              | 1900 ± 85                                   | 0.13                      |
| Basmati 370                                 | DoBS     | 26.0        | 71                   | 72.6                        | 11.8   | 34               | 2350              | 1070 ± 57                                   | 0.12                      |
| Dumsiah                                     | DoBS     | 26.9        | 71                   | 71.7                        | 13.2   | 42               | 2020              | 1165 ± 30                                   | 0.12                      |
| C4-63Ga                                     | DoBS     | 27.2        | 73                   | 75.2                        | 13.4   | 100              | 803               | 995 ± 63                                    | 0.16                      |
| C4-63Gb                                     | Protease | 26.6        | 76                   | 75.7                        | 13.3   | 29               | 3660              | 1120 ± 32                                   | 0.16                      |
| C4-63Gc                                     | Protease | 29.8        | 76                   | 77.0                        | 13.0   | 28               | 3270              | 872 ± 21                                    | 0.19                      |
| IR64a                                       | DoBS     | 26.6        | 70                   | 74.0                        | 13.1   | 50               | 957               | 840 ± 32                                    | 0.17                      |
| IR64b                                       | Protease | 27.4        | 74                   | 74.4                        | 13.7   | 28               | 3310              | 1425 ± 105                                  | 0.14                      |
| Intan                                       | DoBS     | 28.7        | 71                   | 71.7                        | 13.8   | 60               | 928               | 707 ± 60                                    | 0.16                      |
| Rojolele                                    | DoBS     | 28.4        | 71                   | 71.8                        | 13.7   | 92               | 659               | 974 ± 51                                    | 0.14                      |
| Labelle                                     | DoBS     | 28.4        | 74                   | 74.2                        | 14.9   | 80               | 778               | 817 ± 14                                    | 0.16                      |
| Ganagalla                                   | DoBS     | 28.6        | 76                   | 76.4                        | 14.0   | 28               | 2190              | 1537 ± 60                                   | 0.10                      |
| Yilan-chu-tze                               | DoBS     | 28.4        | 77                   | 76.1                        | 14.9   | 20               | 2710              | 1340 ± 80                                   | 0.09                      |
| Coefficient of variation<br>( <i>n</i> = 3) |          | <1.9%       | <1.8%                | <0.8%                       | <5.1%  |                  |                   |   |                           |

<sup>a</sup> From DSC measurements at 35% (w/w) starch concentration; C.V. of *Tm* (peak temperature) and  $\Delta H$  did not exceed 0.23% and 5.1%, respectively.

<sup>b</sup> Values of 25% (w/w) starch gels (25°C) at 1.0 Hz (*n* = 3).

<sup>c</sup> Means of duplicate measurements; the individual values differ by less than 11.4% (consistency) and 12.9% (viscosity of the mean).

**Table 3. Properties of purified high-amylose rice starches prepared by either DoBS or alkaline protease treatment**

| Sample                                      | Method   | Amylose (%) | Final <i>GT</i> (°C) | <i>Tm</i> <sup>a</sup> (°C) | $\Delta H^a$ (J g <sup>-1</sup> ) | Gel properties   |                    | Storage modulus <i>G'</i> (Pa) <sup>b</sup> | tan $\delta^b$ |
|---|----------|-------------|----------------------|-----------------------------|-----------------------------------|------------------|--------------------|---|----------------|
|   |          |             |                      |                             |                                   | Consistency (mm) | Viscosity (cP)     |   |                |
| IR8a  | DoBS     | 31.6        | 63                   | 64.3                        | 9.3                               | 28 <sup>c</sup>  | 2 080 <sup>c</sup> | 5 425 ± 119                                 | 0.05           |
| IR8b  | Protease | 31.0        | 68                   | 67.4                        | 10.4                              | 25               | 6 040              | 4 655 ± 260                                 | 0.05           |
| IR42a                                       | DoBS     | 31.2        | 63                   | 63.7                        | 11.5                              | 26               | 2 310              | 4 257 ± 230                                 | 0.04           |
| IR42b                                       | Protease | 30.8        | 65                   | 65.7                        | 12.8                              | 27               | 7 350              | 3 857 ± 154                                 | 0.05           |
| IR13754-5-2                                 | Protease | 30.8        | 67                   | 68.1                        | 10.3                              | 24               | 4 950              | 4 440 ± 96                                  | 0.04           |
| IR29723-88-2                                | Protease | 31.6        | 66                   | 66.3                        | 9.7                               | 24               | 4 810              | 4 146 ± 169                                 | 0.05           |
| BG11-11                                     | DoBS     | 26.4        | 74                   | 74.1                        | 12.7                              | 34               | 1 640              | 2 282 ± 45                                  | 0.08           |
| Podiwee A-8                                 | DoBS     | 25.8        | 73                   | 73.1                        | 13.3                              | 30               | 1 670              | 2 340 ± 76                                  | 0.09           |
| H4  | DoBS     | 27.6        | 75                   | 73.7                        | 13.5                              | 28               | 3 740              | 2 670 ± 119                                 | 0.07           |
| IR5   | DoBS     | 33.0        | 72                   | 73.2                        | 13.9                              | 76               | 936                | 3 660 ± 150                                 | 0.06           |
| IR32  | DoBS     | 32.0        | 74                   | 74.7                        | 9.9                               | 76               | 1 270              | 3 086 ± 233                                 | 0.08           |
| IR62  | Protease | 34.6        | 74                   | 75.0                        | 13.6                              | 26               | 1 380              | 1 960 ± 127                                 | 0.08           |
| IR70  | Protease | 30.2        | 74                   | 75.0                        | 14.9                              | 26               | 5 640              | 1 655 ± 35                                  | 0.06           |
| BKN-6987-62                                 | DoBS     | 29.2        | 75                   | 74.8                        | 13.4                              | 32               | 1 510              | 2 045 ± 115                                 | 0.08           |
| IR36 ae mutant                              | Protease | 48.4        | 73                   | 75.7                        | 10.9                              | 25               | 5 790              | 12 700 ± 720                                | 0.05           |
| Coefficient of variation<br>( <i>n</i> = 3) |          | <2.2%       | <1.6%                | <0.6%                       | <6.2%                             |                  |                    |   |                |

<sup>a</sup> From DSC measurements at 35% (w/w) starch concentration; C.V. of *Tm* (peak temperature) and  $\Delta H$  did not exceed 0.29% and 4.8%, respectively.

<sup>b</sup> Values of 25% (w/w) starch gels (25°C) at 1.0 Hz (*n* = 3).

<sup>c</sup> Means of duplicate measurements; the individual values differ by less than 12.4% (consistency) and 13.3% (viscosity) of the mean.

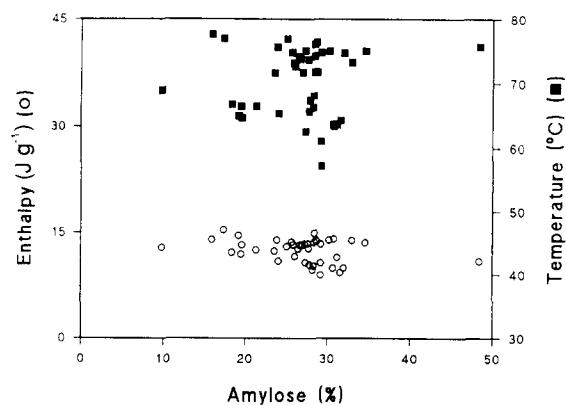
starches; these samples also had high viscosity. For the same variety, protease-treated starches tended to have harder gel consistency and higher gel viscosity than their DoBS-extracted counterparts (e.g. IR42a,b, IR64a,b, IR8a,b). A major factor for such property responses may be the changes in lipid composition of the DoBS-treated starches. Lipids have been identified as a major factor affecting gel consistency and viscosity (Perez, 1979). It is well known that DoBS partially replaces internal granular lipids (Maniñgat & Juliano, 1980), and this may alter the capacity of the starch granules to hydrate and swell. The slightly lower final *GT* temperatures observed for the DoBS-treated starches of the same variety (Tables 2 and 3) also suggests that granule destabilization is caused by this detergent.

The apparent gelatinization enthalpy ( $\Delta H$ ) values for the rice starches ranged between 9.3 and 15.9 J g<sup>-1</sup> (Tables 1, 2 & 3; 35% w/w starch dispersions). The peak temperature of the gelatinization endotherm (*Tm*) and final *GT* were significantly correlated ( $r = 0.96$ ,  $p < 0.001$ ) as has been previously reported for other granular starches (Biliaderis *et al.*, 1980; Russell & Juliano, 1983). Differences in amylose content among the samples were not reflected on the resistance of the granules to gelatinization (*Tm*) or the energy required to disrupt their structure upon heating ( $\Delta H$ ), as illustrated in Fig 1.

### Viscoelastic properties of rice starch gels

Many investigations have dealt with the flow behaviour of starch dispersions and pastes, as recently reviewed by Launay *et al.* (1986) and Doublier (1990). However,

interpretation of the rheological behaviour in terms of macromolecular organization and interactions among constituents has not been fully elucidated and it would require complementary analytical data to unravel the molecular processes involved in structure formation. When starch granules are heated in water above their melting temperature, irreversible swelling and amylose solubilization occur. On cooling sufficiently concentrated starch dispersions (>6.0% w/w), a gel network is established due to intermolecular associations in the polymer-rich regions. In contrast to other polysaccharide network systems (e.g. pectins, seaweed polysaccharides), starch gels are composite networks in which starch granules are embedded in a continuous matrix of physically cross-linked amylose chains (Ring & Stainsby, 1982; Ring, 1985). It is generally accepted that both the continuous phase (amylose) and the swollen granules contribute to the mechanical properties



**Fig. 1.** Relationships between amylose content of rice starch and gelatinization enthalpy, and peak melting temperature.

of the gel composite. Thus, granular size, amylose-amylopectin ratio, minor constituents (lipids, phosphorus, ect.), as well as starch concentration-temperature-time protocols are all important determinants of the viscoelastic behaviour of starch dispersions (Launay *et al.*, 1986; Biliaderis, 1992).

In the present investigations, rice starch gels were quickly set from aqueous suspensions of granules in hermetically sealed containers under static conditions, i.e. no shear was applied upon heating and cooling. Since there are no major differences in shape, size and size distributions of granules among low-, intermediate- and high-amylose starches, these materials offer a unique model system to explore the relationship between amylose content and mechanical properties of starch gels. For all gels,  $G'$  exceeded  $G''$  and both moduli showed little dependence on frequency (Fig. 2) within the frequency range 0.1–20.0 Hz. This rheological response is more typical of a 'true' gel (physical cross-links have a permanent character) than an entanglement network system (e.g. polysaccharide solutions/dispersions where transient cross-links arise from chain entanglements). Figure 3 shows the dependence of  $G'$  and  $\tan \delta$  on amylose content (9.8–48.4%) for the 25% (w/w) gels of all rice samples (at 1.0 Hz). An inverse linear relationship between  $\tan \delta$  and amylose content was found;  $r = 0.75$ ,  $p < 0.01$ . The  $\tan \delta$  is a more sensitive parameter than  $G'$  and  $G''$  in probing changes in the viscoelastic nature of polymer gels. The gels of all high-amylose-content rice starches exhibited  $\tan \delta$  values less than 0.1 (Table 3), pointing to well-cross-linked network structures. There was a general trend for increased  $G'$  values with increasing amylose content; an exponential relationship  $G' \propto (\text{amylose content})^{2.72}$ ,  $r = 0.76$ ,  $p < 0.01$ , was found. Between the two starch polymers, amylose gelation is effected at much lower polymer concentrations than amylopectin (Biliaderis & Zawistowski, 1990; Doublier & Choplin, 1989; Ring *et al.*, 1987). Thus, for the higher-amylose rice starches, one would expect greater amounts of exuded linear starch molecules at the intergranular spaces. This would increase the extent of interchain associations necessary for the development

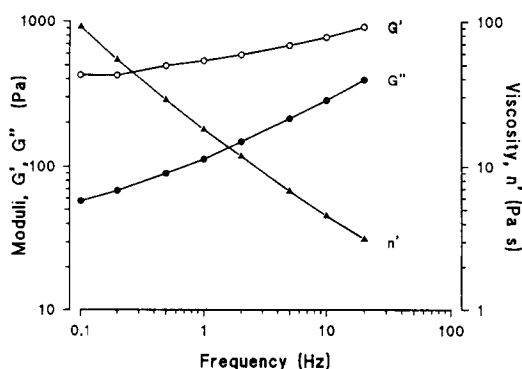


Fig. 2. Frequency dependence of dynamic moduli ( $G'$ ,  $G''$ ) and viscosity ( $\eta$ ) of IR37307-8-ch starch gel (25% w/w) at 25°C, strain  $< 3.0\%$ .

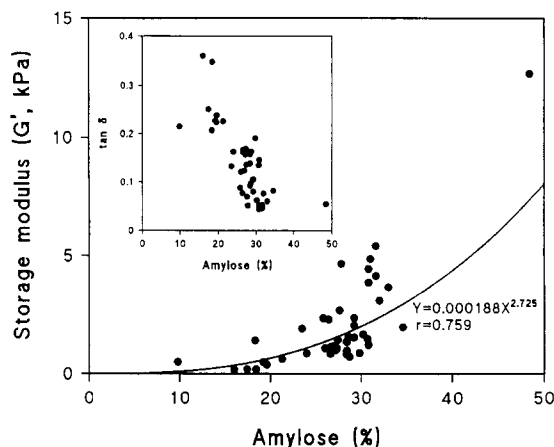


Fig. 3. Relationships between amylose content and  $G'$ , and  $\tan \delta$  of 25% (w/w) rice starch gels (at 1.0 Hz, 25°C).

of rigid three-dimensional network structure. However, the substantial variation in modulus values observed for the intermediate- to high-amylose content samples (Fig. 3) clearly indicates that other factors also contribute to the mechanical properties of starch gels. In a related study, Wong and Lelievre (1981) concluded that differences in the dynamic mechanical properties of wheat starch pastes ( $< 7.0\%$  w/w) from several cultivars may be due to variations in the swelling capacity and number fraction of large granules. Differences in the amounts of starch lipids among the rice samples could also affect their gel properties. Biliaderis and Tonogai (1991) have recently shown that removal of internal granular lipids from wheat and rice starches results in increased gel firmness ( $G'$ ) and viscosity ( $\eta$ ). A likely explanation is that granular lipids inhibit leaching of amylose from the granules during gelatinization and thereby yield a weaker continuous phase. An attempt was also made to examine whether the gelatinization temperature of starch has any effect on the mechanical properties of the thermo-set gel. Rice starches with low ( $T_m$ : 63–67°C,  $n = 15$ ) and high ( $T_m$ : 73–78°C,  $n = 16$ ) gelatinization temperatures were pooled separately and the amylose content, [% A], dependence of  $G'$  was determined. A slightly stronger dependence of modulus on [% A] was found for the low  $GT$  group;  $G' \propto [\% A]^{4.22}$  for low  $GT$  vs  $G' \propto [\% A]^{3.76}$  for high  $GT$ . This comparison may suggest that a better gel network structure is established with the low  $GT$  starches. However, a more extensive study would be required to further test this hypothesis. No significant relationships were found between  $G'$  and gel consistency ( $r = -0.44$ , ns) and  $G'$  and gel viscosity ( $r = +0.47$ , ns). This indicates that although empirical tests are useful as indirect indices of rice quality (Juliano, 1985a), such measurements are not related to the fundamental mechanical properties of the sample. Obviously, a more detailed description of the viscoelastic character of starch gels and the time-dependent consistency changes can be obtained by the use of non-destructive small strain rheological testing, as employed in the present studies.

### Effect of starch concentration on gel rigidity

The dependence of rigidity modulus ( $G'$ ) on polymer concentration is often examined for physically cross-linked gelling biopolymer systems. For starch gels, the modulus is highly dependent on time due to retrogradation (Biliaderis, 1992). This means rheological tests must be carried out immediately after gelation under constant temperature and at a fixed frequency. For potato, wheat and maize starch gels, in the concentration range of 6–30% (w/w), linear relationships between the rigidity modulus (as determined with a Ward and Saunder's U-tube) and concentration have been reported (Ring, 1985). A linear relationship between  $G'$  and concentration was also shown for corn starch pastes (4–8% w/w) by small deformation oscillatory tests (Evans & Haisman, 1979). However, a stronger concentration dependence ( $\alpha c^{2.1-2.9}$ ) has been recently reported for rice and garbanzo bean starch gels (8–40% w/w) also using small amplitude oscillatory shear testing. Figure 4 shows the concentration dependence of  $G'$  for four different rice starches at concentrations between 10 and 25% (w/w). Linear relationships between  $G'$  and concentration were obtained when the data were plotted on a log-log scale ( $r > 0.96$ ,  $p < 0.01$  in all cases). From the slopes of these lines, the  $G'$  was found to vary with concentration as  $C^{2.2-2.9}$ . These exponent values are similar to those previously reported for rice

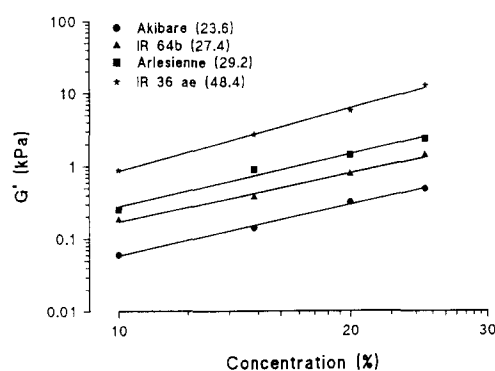


Fig. 4. Experimental  $G'$  values (at 1.0 Hz, 25°C) vs concentration of rice starch gels; numbers in parentheses are the corresponding amylose contents.

and garbanzo bean starches (Biliaderis & Tonogai, 1991). The above values are also closer to the theoretical limiting relationship  $G' \propto C^{2.0}$  predicted for biopolymer gels at high polymer concentrations (Clark & Ross-Murphy, 1987) than earlier published data on starch gels (Ring, 1985; Evans & Haisman, 1979; Wong & Lelievre, 1981).

### Kinetics of gelation-retrogradation of rice starches

It is well accepted that starch gels are non-equilibrium, kinetically constrained systems; changes in  $G'$  and crystallinity on storage reflect contributions from diffusion-controlled chain aggregation processes. Dynamic rheological tests are particularly useful in monitoring the kinetics of network development, provided that such measurements are within the linear viscoelastic regime (Biliaderis, 1992). Aging kinetics of 25% (w/w) of starch gels (8°C) were undertaken on selected samples varying in their amylose content (9.8–31.6%), as shown in Figure 5. The lipid composition of these starches is shown in Table 4. A large variation in gel rigidity development was observed among the samples. Previous studies on more concentrated (>30%, w/w) cereal and legume starch gels (Biliaderis & Zawistowski, 1990; Biliaderis & Tonogai, 1991) showed that the

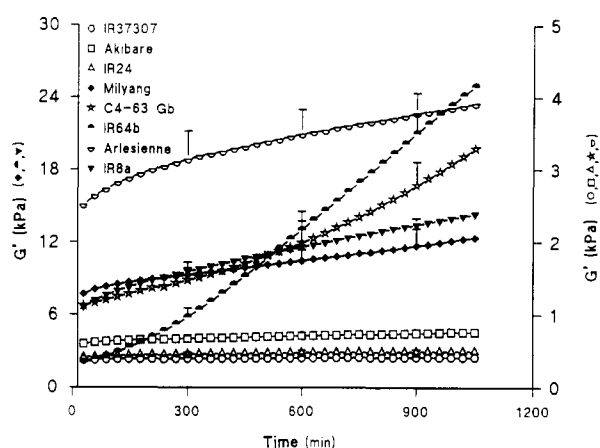


Fig. 5. Storage modulus ( $G'$ )-time profiles for rice starch gels (25% w/w) stored at 8°C (at 0.2 Hz).

Table 4. DSC staling endotherm enthalpies of 35% (w/w) rice starch gels stored at 6°C for four and seven days

| Sample                   | Lipids     | Lipids     |        | Amylose (%) | $\Delta H$ (J g <sup>-1</sup> ), 4 days |           | $\Delta H$ (J g <sup>-1</sup> ), 7 days |           |
|--------------------------|------------|------------|--------|-------------|---|-----------|---|-----------|
|                          |            | Non-starch | Starch |             | Native                                  | Defatted  | Native                                  | Defatted  |
| IR37307-8-ch             | (Protease) | 0.28       | 0.52   | 9.8         | 1.4 ± 0.1 <sup>a</sup>                  | 8.9 ± 0.5 | 1.9 ± 0.1                               | 9.8 ± 0.3 |
| IR24                     | (DoBS)     | 0.15       | 0.43   | 19.5        | 4.4 ± 0.1                               | 7.9 ± 0.2 | 6.2 ± 0.3                               | 8.2 ± 0.1 |
| Akibare                  | (DoBS)     | 0.21       | 0.51   | 23.6        | 5.5 ± 0.2                               | 5.7 ± 0.1 | 5.9 ± 0.1                               | 7.7 ± 0.1 |
| Milyang 23               | (DoBS)     | 0.24       | 0.57   | 23.4        | 4.7 ± 0.1                               | 6.9 ± 0.1 | 6.0 ± 0.2                               | 8.0 ± 0.1 |
| IR64b                    | (Protease) | 0.20       | 0.96   | 27.4        | 7.6 ± 0.2                               | 8.4 ± 0.1 | 8.5 ± 0.1                               | 8.8 ± 0.2 |
| C4-63Gb                  | (Protease) | 0.22       | 0.83   | 29.8        | 9.1 ± 0.3                               | 8.3 ± 0.1 | 9.3 ± 0.1                               | 9.8 ± 0.1 |
| Arlesienne               | (DoBS)     | 0.17       | 0.53   | 29.2        | 3.9 ± 0.2                               | 5.5 ± 0.2 | 4.7 ± 0.2                               | 6.9 ± 0.3 |
| IR8a                     | (DoBS)     | 0.20       | 0.90   | 31.6        | 4.9 ± 0.1                               | 6.4 ± 0.2 | 5.5 ± 0.1                               | 7.1 ± 0.2 |
| IR36 ae mutant           | (Protease) | 0.30       | 0.75   | 48.4        | 9.3 ± 0.2                               | 7.1 ± 0.2 | 10.2 ± 0.1                              | 8.5 ± 0.2 |
| Coefficient of variation |            | <2.6%      | <3.8%  |             |   |           |   |           |

<sup>a</sup> Means ± S.D. ( $n = 3$ ).

$G'$ -time profiles often reveal a biphasic gelation process: an initial rapid rise in modulus (attributed to amylose gelation) followed by a phase of much slower  $G'$  development (reflecting retrogradation of amylopectin). For the starch gel concentrations adopted in this study (25% w/w), it appeared that the first phase has been rapidly completed during the gel preparation stage. The  $G'$ -time profiles in Fig. 5 most likely reflect aggregation processes of the amylopectin, as found by Biliaderis and Zawistowski (1990). Clearly, the gels of IR37307-8-ch and IR24 (having the lowest amylose contents) exhibited low initial  $G'$  values and a relatively slow  $G'$  development. In contrast, the IR64b and C4-63Gb gels showed sharp increases in  $G'$  at longer storage times. These samples also had high lipid contents (Table 4) which could explain the initial delay in modulus development; complexation of amylopectin with lipids may have a retarding effect on retrogradation. However, the IR8a gels with similar amylose and lipid contents to those of IR64b and C4-63Gb showed much slower kinetic responses. Obviously, the underlying mechanism(s) by which all pertinent molecular parameters affect gel network development of such composite hydrogels is complex and warrants further investigation.

The retrogradation process in rice starch gels (35% w/w) was also followed calorimetrically over longer storage periods (at 6°C up to seven days). Amylopectin retrogradation is accompanied by the development of the 'staling endotherm' ( $T_m \sim 45\text{--}60^\circ\text{C}$ ) as a result of chain reorganisation (mainly involving the outer short DP  $\sim 15\text{--}20$  chains of the molecule). The rate of staling endotherm development ( $\Delta H$ ) in an aging gel is generally much slower than that of  $G'$  (Biliaderis & Zawistowski, 1990). Kinetics of  $\Delta H$  development for nine different native and defatted rice starch gels are presented in Figs 6 and 7, respectively. Table 4 also includes the absolute mean  $\Delta H$  values of the gels following storage of four and seven days. It was evident from the DSC results that extraction of granular lipids brings about greater staling rates in all rice starches with the exception of IR36 ae mutant (Table 4, Figs 6 & 7). The greatest effect was observed for the low-amylose samples (e.g. IR 37307-8-ch and IR24).

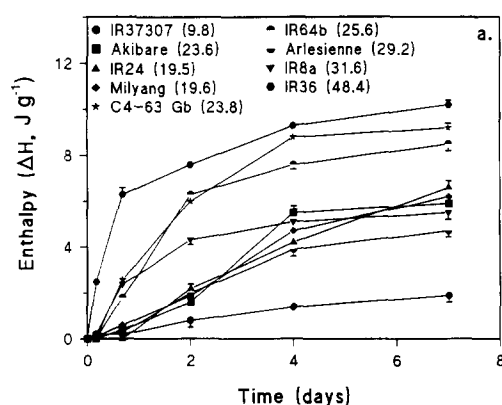


Fig. 6. Retrogradation of native rice starch gels (35% w/w, 6°C); numbers in parentheses are the corresponding amylose contents.

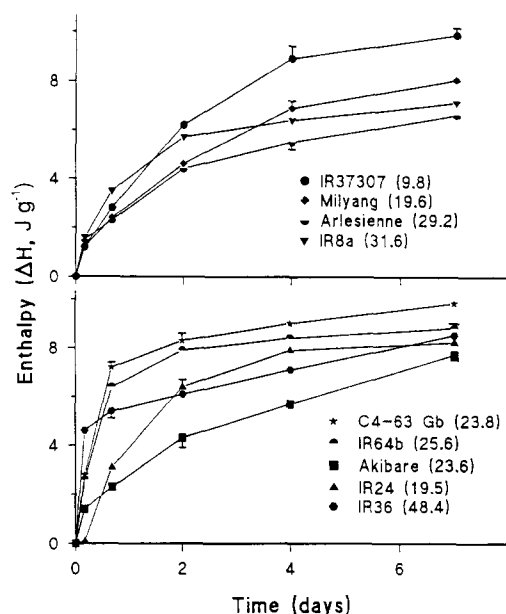


Fig. 7. Retrogradation of defatted rice starch gels (35% w/w, 6°C).

These results support the argument that granular lipids do associate with amylopectin during gelatinization, thereby lowering the tendency of this polymer to retrograde. Indeed, there has been indirect calorimetric evidence for complexation of amylopectin and lipids (Evans, 1986; Gudmundsson & Eliasson, 1990). Interestingly, there was a decrease in retrogradation of gels of the defatted IR36 ae starch as compared to its native counterpart. For such high-amylose-content starches, it is likely that removal of granular lipids enhances the interactions between amylose and amylopectin in their gels. This would retard the formation of ordered structures involving only the outer chains of amylopectin which gave rise to the staling endotherm. In highly concentrated gels, as those of the present study, it is unlikely that sufficient phase separation between the two starch constituents takes place. Instead, intermolecular associations between amylose and amylopectin would probably occur. Depending on the amylose-amylopectin ratio of each particular gel system, lipids (because of their ability to complex with both polymers) would modulate the extent of such interchain associations and thereby alter the thermal and mechanical properties of the composite gel.

## ACKNOWLEDGEMENTS

The financial support from the Natural Sciences and Engineering Research Council of Canada is gratefully acknowledged. The authors also acknowledge J. Tonogai for technical assistance.

## REFERENCES

- Asaoka, M., Okuno, K. & Fuwa, H. (1985a). Genetic and environmental control of starch properties in rice seeds. In *New Approaches to Research on Cereal Carbohydrates*,

- ed. R. R. Hill & L. Munck. Elsevier Science Publishers, Amsterdam, pp. 29–38.
- Asaoka, M., Okuno, K., Sugimoto, Y. & Fuwa, H. (1985b). Developmental changes in the structure of endosperm starch of rice (*Oryza sativa* L.). *Agric. Biol. Chem.*, **49**, 1973–9.
- Biliaderis, C. G. (1992). Characterization of starch networks by small strain dynamic rheometry. In *Developments in Carbohydrate Chemistry*, ed. R. J. Alexander & H. F. Zobel. American Association of Cereal Chemists, pp. 87–135.
- Biliaderis, C. G. & Tonogai, J. R. (1991). Influence of lipids on the thermal and mechanical properties of concentrated starch gels. *J. Agr. Food Chem.*, **39**, 833–40.
- Biliaderis, C. G. & Zawistowski, J. (1990). Viscoelastic behaviour of aging starch gels: effects on concentration, temperature and starch hydrolysates on network properties. *Cereal Chem.*, **67**, 240–6.
- Biliaderis, C. G., Maurice, T. J. & Vose, J. R. (1980). Starch gelatinization phenomena studied by differential scanning calorimetry. *J. Food Sci.*, **45**, 1669–74 & 1680.
- Biliaderis, C. G., Page, C. M., Slade, L. & Sirett, R. R. (1985). Thermal behaviour of amylose–lipid complexes. *Carbohydr. Polym.*, **5**, 367–89.
- Cagampang, G. B., Perez, C. M. & Juliano, B. O. (1973). A gel consistency test for eating quality of rice. *J. Sci. Food Agric.*, **24**, 1589–94.
- Clark, A. H. & Ross-Murphy, S. B. (1987). Structural and mechanical properties of biopolymer gels. *Adv. Polym. Sci.*, **83**, 57–192.
- Del Mundo, A. M., Kosco, D. A., Juliano, B. O., Siscar, J. J. H. & Perez, C. M. (1989). Sensory and instrumental evaluation of texture of cooked and raw milled rices with similar starch properties. *J. Texture Stud.*, **20**, 97–110.
- Doublier, J.-L. (1990). Rheological properties of cereal carbohydrates. In *Dough Rheology and Baked Product Texture*, ed. H. Faridi & J. M. Faubion. Van Nostrand Reinhold, pp. 111–55.
- Doublier, J.-L. & Choplin, L. (1989). A rheological description of amylose gelation. *Carbohydr. Res.*, **193**, 215–26.
- Evans, I. D. (1986). An investigation of starch/surfactant interactions using viscometry and differential scanning calorimetry. *Starch*, **38**, 227–35.
- Evans, I. D. & Haisman, D. R. (1979). Rheology of gelatinized starch suspensions. *J. Texture Stud.*, **10**, 347–70.
- Gudmundsson, M. & Eliasson, A.-C. (1990). Retrogradation of amylopectin and the effects of amylose and added surfactants/emulsifiers. *Carbohydr. Polym.*, **13**, 295–315.
- IRRI (1979). *Proc. Workshop on Chemical Aspects of Rice Grain Quality*. Int. Rice Res. Inst., Los Baños, Laguna, Philippines, 390 pp.
- Juliano, B. O. (1979a). The chemical basis of rice grain quality. In *Proc. Workshop on Chemical Aspects of Rice Grain Quality*. Int. Rice Res. Inst., Los Baños, Laguna, Philippines, pp. 69–90.
- Juliano, B. O. (1979b). Amylose analysis in rice. A review. In *Proc. Workshop on Chemical Aspects of Rice Grain Quality*. Int. Rice Res. Inst. Los Baños, Laguna, Philippines, pp. 251–60.
- Juliano, B. O. (1985a). Criteria and tests for rice grain qualities. In *Rice: Chemistry and Technology*, ed. B. O. Juliano. American Association of Cereal Chemists, pp. 443–524.
- Juliano, B. O. (1985b). Polysaccharides, proteins and lipids of rice. In *Rice: Chemistry and Technology*, ed. B. O. Juliano. American Association of Cereal Chemists, pp. 59–174.
- Juliano, B. O., Perez, C. M., Blakeney, A. B., Castillo, D. T., Kongseree, N., Laignelet, B., Lapis, E. T., Murty, V. V. S., Paule, C. M. & Webb, B. D. (1981). International cooperative testing on the amylose content of milled rice. *Starch*, **33**, 157–62.
- Launay, B., Doublier, J.-L. & Cuvelier, G. (1986). Flow properties of aqueous solutions and dispersions of polysaccharides. In *Functional Properties of Food Macromolecules*, J. R. Mitchell & D. A. Ledward. Elsevier, pp. 1–78.
- Maniñgat, C. C. & Juliano, B. O. (1980). Starch lipids and their effect on rice starch properties. *Starch*, **32**, 76–82.
- Morrison, W. R. & Azudin, M. N. (1987). Variation in the amylose and lipid contents and some physical properties of rice starches. *J. Cereal Sci.*, **5**, 35–44.
- Perez, C. M. (1979). Gel consistency and viscosity of rice. In *Proc. Workshop on Chemical Aspects of Rice Grain Quality*. Int. Rice Res. Inst., Los Baños, Laguna, Philippines, pp. 293–302.
- Ring, S. G. (1985). Some studies on starch gelation. *Starch*, **37**, 80–3.
- Ring, S. G. & Stainsby, G. (1982). Filler reinforcement of gels. *Prog. Food Nutr. Sci.*, **6**, 323–9.
- Ring, S. G., Colonna, P., l'Anson, K., Kalichevsky, M. T., Miles, M. J., Morris, V. R. & Orford, P. D. (1987). The gelation and crystallization of amylopectin. *Carbohydr. Res.*, **162**, 277–93.
- Russell, P. L. & Juliano, B. O. (1983). Differential scanning calorimetry of rice starches. *Starch*, **35**, 382–6.
- Schoch, T. J. & Maywald, E. C. (1956). Microscopic examination of modified starches. *Anal. Chem.*, **28**, 387–90.
- Wong, R. B. K. & Lelievre, J. (1981). Viscoelastic behaviour of wheat starch pastes. *Rheol. Acta*, **20**, 299–307.