Carbohydrate Polymers 87 (2012) 1507–1514

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

journal homepage: www.elsevier.com/locate/carbpol

Nature of thermal transitions of native and acid-hydrolysed pea starch: Does gelatinization really happen?

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ARTICLE INFO

Article history:
Received 22 March 2011
Received in revised form 17 August 2011
Accepted 13 September 2011
Available online 29 September 2011

Keywords:
Starch
Pea
Pisum sativum
Acid hydrolysis
Thermal transition
Swelling and dissolution

ABSTRACT

Thermal transitions of native and acid-hydrolysed starch were investigated for the first time by employing differential scanning calorimetry (DSC) in combination with field emission scanning electron microscopy (FE-SEM) to examine morphological changes in the starch after hydrothermal treatment in the DSC pans. The characteristic, well-defined endothermic transition was observed in the DSC traces for native starch and starch after one day of acid hydrolysis, but after two days of hydrolysis the endothermic transition became broad and undefined. After heating in DSC pans, native starch was observed visually and with SEM to have formed a gel, whereas starch granules hydrolysed for one day appeared to have undergone only limited swelling and coalescence. Starch that had been hydrolysed for two or more days appeared powdery and SEM images revealed that the granules had undergone very little swelling. The thermal transitions of native and acid-hydrolysed starch are interpreted in terms of the theory of polymer swelling and dissolution. Native granules and granules with only minimal acid damage are able to swell within the constraints of the water-limited conditions of DSC, but as acid hydrolysis progresses the capacity of the granules to swell is decreased and endothermic transitions are increasingly due to dissolution of polymer chains. An exothermic transition that followed the main endothermic transition is proposed to be due to the condensation of water vapour on the lid of the DSC pan.

1. Introduction

Native starch granules contain two types of α-D-glucose polymers: amylose, an essentially linear molecule with degree of polymerization of 1000–10,000, and the highly branched amylopectin, which may have several million glucose units (Delcour & Hoseney, 2010). Amylose and amylopectin molecules are organized into semi-crystalline granules, which vary in form and properties between and within species. When heated in excess water, native starch granules undergo an irreversible process termed gelatinization, which involves a sequence of events that result in the loss of molecular organization and granular morphology (Atwell, Hood, Lineback, Varriano-Marston, & Zobel, 1988). During gelatinization, the glucan double helices of amylopectin unwind, the crystalline structures collapse, and the granules swell with dissolution and leaching of the majority of amylose molecules and some of the amylopectin molecules (the usage in this paper of the term “dissolution” is intended to include the hydration and formation of a molecular dispersion of the polymer). The conditions for these events, and the extent to which they occur, depends on the starch source and its structural properties (Shamekh, Forssell, & Poutanen, 1994; Tester & Morrison, 1990a). Gelatinization behaviour is generally monitored by differential scanning calorimetry (DSC) in combination with other complementary techniques such as wide angle X-ray diffraction (XRD), small angle X-ray scattering (SAXS), nuclear magnetic resonance (NMR), optical microscopy and scanning electron microscopy (SEM) (Ratnayake & Jackson, 2009). In DSC traces of starch:water mixtures (commonly 2:1, w/w), the main endothermic transition (sometimes termed the gelatinization endotherm) is considered to be associated mainly with the disruption of the double helices in amylopectin, and to a lesser extent, the melting of crystallites. However, combined DSC, XRD and SAXS studies have shown that residual crystallinity and lamellar organization remain at the end of the so-called gelatinization endotherm (Jenkins & Donald, 1997; Vermeylen et al., 2006). These studies indicate that the endothermic transition measured by DSC does not represent complete gelatinization. The gelatinization behaviour of starch is a major determinant of its functionality in foods, but as a recent review article indicates, there is as yet no universally accepted explanation of the mechanism of the process (Ratnayake & Jackson, 2009).

Many studies have shown the gelatinization characteristics of starches to be greatly influenced by acid hydrolysis. The main endothermic transition of acid hydrolysed starch is generally observed to be broader and has its peak shifted to higher

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doi:10.1016/j.carbpol.2011.09.047

temperatures, compared to native starch (Atichokudomchai, Varavinit, & Chinachoti, 2002; Donovan, 1979; Donovan & Mapes, 1980; Garcia et al., 1996; Jacobs, Eerlingen, Rouseu, Colonna, & Delcour, 1998; Komiya & Nara, 1986; Morrison, Tester, Gidley, & Karkalas, 1993; Muhr, Blanshard, & Bate, 1984) These effects on the gelatinization endotherm are usually explained as being due to an increase in crystallinity or molecular order with acid hydrolysis, and greater heterogeneity of the hydrolysis product. Other interpretations include that the preferential hydrolysis by acid of amorphous regions attenuates the destabilizing effect of swelling in amorphous regions on the melting of the crystallites (Donovan, 1979), or that it permits the formation of longer amylopectin double helices as a result of the removal of branch points (Morrison et al., 1993). In contrast, a decrease in the temperature of the endothermic transition has been reported for waxy starches (Genkina, Wikman, Bertoft, & Yuryev, 2007; Perera, Lu, Sell, & Jane, 2001; Shi & Seib, 1992; Tester & Morrison, 1990b). Genkina et al. (2007) proposed that this decrease is due to the acid partially disrupting the crystalline lamellae of waxy starches and increasing the defects on the edges of the crystallites. Furthermore, they argued that gelatinization events in these acid-hydrolysed waxy starches occur predominantly within the crystalline regions.

The enthalpy change associated with the melting of the crystallites and disruption of the molecular order of the granules is referred to as the enthalpy of gelatinization (ΔH_{gel}). In most studies, ΔH_{gel} was observed to decrease in acid-hydrolysed starches, which has been explained on the basis that enthalpy change is mainly contributed by amorphous regions, which are preferentially hydrolysed by acid (Muhr et al., 1984). However, an increase in ΔH_{gel} in acid-hydrolysed starch has been noted in some studies and was attributed to the increase in crystallinity (Jayakody & Hoover, 2002; Morrison et al., 1993). Further, some researchers reported that there was little effect on ΔH_{gel} even after prolonged acid hydrolysis, which was explained as due to there being little effect of acid hydrolysis on the crystalline structure (Donovan & Mapes, 1980; Jenkins & Donald, 1997).

Although the effects of acid hydrolysis on the thermal behaviour of starch have been interpreted in different ways, most explanations are based on the assumption that gelatinization of starch still occurs even after prolonged hydrolysis. The present paper challenges this assumption, particularly as there is a lack of information on the morphological changes of starches that have undergone thermal transitions in DSC pans.

The preparation and analysis of acid-hydrolysed starch by a combination of physical, chemical and imaging methods are described in an another paper (Wang, Blazek, Gilbert, & Copeland, 2011). Crystallinity was shown to increase in the initial stages of hydrolysis, indicating that the amorphous regions were being degraded. Evidence is also presented to show that amylose is more concentrated in the core of the granule, and that both amylose and amylopectin are located on the surface of the granules and are attacked simultaneously during the initial stages of hydrolysis. More extensive hydrolysis resulted in the simultaneous disruption of amorphous and crystalline regions. The present paper describes the application of a novel concept of semi-preparative DSC, which involves thermal transition processing of starch samples in DSC pans and subsequent morphological characterization. The nature of thermal transitions is explored from the new perspective of the swelling and dissolution of polymer chains in combination with the morphological changes of starches after thermal transition in DSC pans using field-emission scanning electron microscopy (FE-SEM). In this context, the term “endothermic transition” is used rather than gelatinization for the DSC studies.

2. Experimental

2.1. Materials

Field pea varieties Maki, Kaspa and PRL131 were obtained from the Plant Breeding Institute (Narrabri, The University of Sydney, NSW). The origin of these varieties and the isolation of starch are described in Wang, Sharp, and Copeland (2011). The moisture content of the isolated starches was 11.1% (Maki), 11.8% (Kaspa), and 11.4% (PRL131), and the amylose content was between 35 and 38%. The acid-hydrolysed starch was prepared according to the method of Wang, Yu, and Yu (2008), which is similar to the method used generally to prepare Lintnerized starch.

2.2. Differential scanning calorimetry

Measurements were made using a Modulated Differential Scanning Calorimeter MDSC 2920 instrument (TA Instruments Inc., Delaware, USA) equipped with a thermal analysis data station and data recording software. Approximately 3 mg of starch were weighed accurately into an aluminium sample pan. Distilled water was added with a microsyringe to obtain a starch:water ratio of 1:2 (w/w) in the DSC pans. Care was taken to ensure that the starch samples were completely immersed in the water by gentle shaking before the pans were sealed, reweighed and left overnight at room temperature before DSC analysis. An empty pan was used as a reference. The pans were heated from 30 to 95 °C and 30 to 125 °C at a scanning rate of 10 °C/min. The instrument was calibrated using indium as a standard. The Universal Analysis 2000 software was used to analyse the transition temperatures and enthalpy change (ΔH) of the endotherm. The transition temperatures calculated were: (T_s), defined as the inflexion point at which endothermic heat flow increased on the DSC trace; the onset temperature (T_o), the intersection point of tangents to the trace at T_s and the down slope of heat flow; T_p, the temperature of maximum heat flow; and conclusion T_c, the intersection point of tangents to the trace at the up slope after T_p and an estimate of the baseline.

After heating, the pans were cooled to room temperature and reweighed. Those pans without any weight loss were collected. The pans were opened carefully and left to dry under atmospheric conditions overnight before the starch samples were removed carefully with a spatula.

2.3. Field-emission scanning electron microscopy

The dried starch specimens were fixed on to the surface of double-sided, carbon-coated adhesive tape attached to an aluminium stub. The mounted samples were sputter coated with Palladium/Gold prior to imaging by FE-SEM (Carl Zeiss ULTRA plus, Germany). The accelerating voltage was 1.01 kV and the magnification is shown on the micrographs.

3. Results

3.1. Thermal transitions of native and acid-hydrolysed starches

On heating to 95 °C in the DSC instrument, only native starch and starch hydrolysed with acid for one day showed the typical endothermic transition. As the DSC profiles of the three starches were similar, only the trace for variety Kaspa is presented in Fig. 1. For the three native starches, the T_s was 53–54 °C and T_o 60–62 °C, the temperature for the endothermic minimum (T_p) was close to 68 °C, and the temperature for the conclusion of the endothermic transition (T_c) was 73–74 °C (Table 1). The temperature range T_c–T_s was about 20 °C, and the enthalpy change (ΔH) was about 9 J/g. The respective temperatures except T_s were about 10 °C higher for

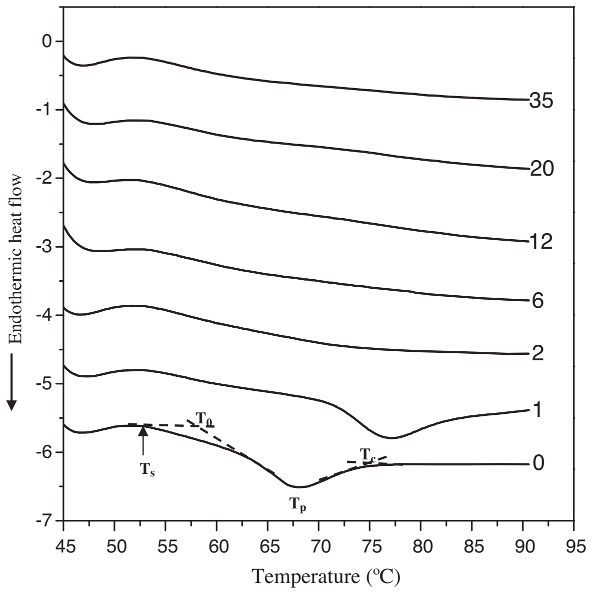

Fig. 1. DSC thermograms of native and acid-hydrolyzed starches heated to 95 °C. The numbers represent the hydrolysis time in days.

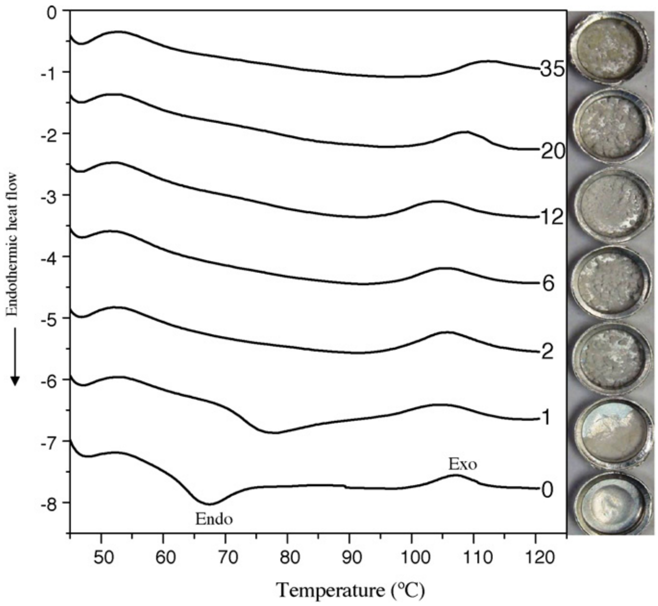

Fig. 2. DSC thermograms of native and acid-hydrolysed starches heated to 125 °C. The numbers next to each curve represent the hydrolysis time in days. The images in the panel on the right are of the corresponding starches after they were heated in the DSC.

starch that had been incubated with acid for one day (Table 1). The temperature range (T_c–T_s) and enthalpy change (ΔH) of the endothermic transition also increased (Table 1). The T_c of starch hydrolysed for one day was less distinct than for native starch. No distinct endothermic transitions were observed with starch that had been incubated with acid for two days or longer, rather the DSC traces showed a gradual endothermic decline.

To determine whether an endothermic transition might occur in acid hydrolysed starch at higher temperatures, a separate experiment was performed in which the starches were heated in the DSC instrument to 125 °C. Again, only native starch and starch hydrolysed with acid for one day displayed a characteristic distinct endothermic transition (Fig. 2). Starches hydrolysed for two or more days did not display the endothermic transition typical of native starch but a broad and undefined endotherm when heated to 125 °C, from which thermal transition parameters could not be calculated (Fig. 2). However, an exothermic transition following the endothermic transition was observed for all samples (Fig. 2). The maximum value for this transition occurred at temperatures between about 105 and 115 °C (Fig. 2).

In a separate experiment, a dilute suspension of native starch granules (1 mg in 25 μl of water) was heated to 125 °C in the DSC instrument. The resulting thermogram had a broad, undefined endothermic transition, which was followed at high temperature by an exothermic transition (Fig. 3). When water alone was heated in the DSC as a comparison, a similar endothermic transition was noted (Fig. 3), but the intensity of the heat input was less than with samples containing native or acid-hydrolysed starch. The exothermic transition at the conclusion of the endothermic heat flow was also present.

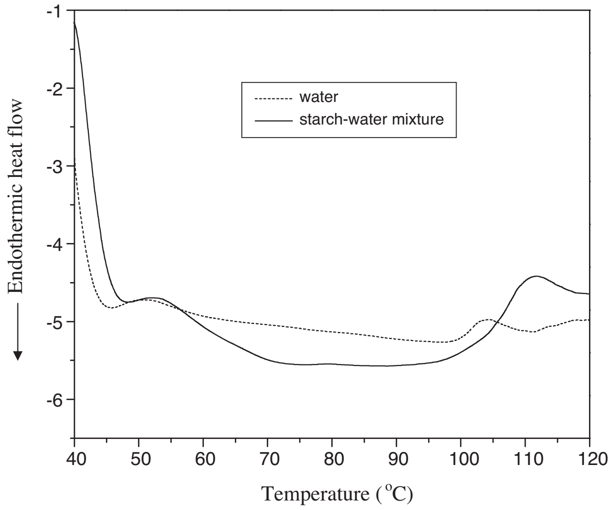

Fig. 3. DSC thermograms of water and a dilute starch-water mixture (1:25 w/v).

3.2. *Morphology of native and acid-hydrolysed starches after DSC measurement*

When the cooled DSC pans were opened after recording the thermograms, the native starch was observed visually to have formed a clear gel, whereas starch hydrolysed for one day formed

Table 1
Thermal transition parameters of native and acid-hydrolysed starches. The transition temperatures indicated are: T_s, defined as the inflexion point at which endothermic heat flow increased on the DSC trace; T_o, the onset temperature, T_p, the temperature of maximum heat flow; and T_c, conclusion temperature. The relationship of these temperatures to the DSC thermogram is shown in Fig. 1.

Starches	Hydrolysis (d)	T_s (°C)	T_o (°C)	T_p (°C)	T_c (°C)	T_c–T_s (°C)	ΔH (J/g)
Maki	0	53.5 ± 0.2	59.8 ± 0.7	67.6 ± 0.0	73.2 ± 0.5	19.7 ± 0.2	8.6 ± 0.6
	1	53.2 ± 0.2	69.6 ± 0.8	77.5 ± 0.1	84.4 ± 1.2	31.2 ± 0.7	13.0 ± 1.6
Kaspa	0	53.7 ± 0.1	61.9 ± 0.1	68.9 ± 0.5	74.2 ± 0.4	20.5 ± 0.2	9.6 ± 0.6
	1	53.6 ± 0.1	72.0 ± 0.9	78.1 ± 0.8	84.5 ± 1.0	30.9 ± 1.0	15.4 ± 1.4
PRL131	0	53.2 ± 0.2	61.0 ± 0.2	68.2 ± 0.2	73.8 ± 0.1	20.6 ± 0.3	8.8 ± 0.2
	1	53.0 ± 0.2	70.9 ± 0.8	76.9 ± 0.7	83.2 ± 1.6	30.2 ± 1.0	13.0 ± 1.3

a translucent material with some gel-like features. Surprisingly, all of starches hydrolysed for two days or more appeared at first as a white glassy material after DSC heating, which subsequently took on a powdery appearance after exposure to the atmosphere overnight (Fig. 2).

SEM images of the heated and cooled native starch granules showed that a more-or-less continuous gel phase had formed with the loss of granular morphology (Figs. 4A1 and 5A1). Residual traces of swollen granules were visible after heating to 95 °C (Fig. 4A2), but these had almost disappeared after heating to 125 °C (Fig. 5A2). Starch granules that were hydrolysed for one day were still able to absorb water and swell and form a gel-like material after heating. Images of starch granules that were hydrolysed for one day and heated to 95 or 125 °C, showed more limited swelling and coalescence than native granules (Figs. 4B2 and 5B2). The granular shapes could still be clearly made out and the granular morphology was not completely lost. Cracks appeared on the surface of starch granules hydrolysed for two and six days but there was little evidence of swelling and coalescence (Figs. 4C, D and 5C, D). The starch granule fragments after 12, 20 and 35 days of hydrolysis did not display swelling behaviour on heating to 95 or 125 °C, although the granule fragments appeared to have undergone some form of fusion during DSC (Figs. 4E–G and 5E–G). The absence of swelling behaviour and subsequent gel formation in starch after two days of hydrolysis was consistent with the DSC traces for these samples, which did not have the characteristic endothermic transitions.

4. Discussion

When native starch granules were heated in DSC pans, the characteristic endothermic transition was observed. Subsequent SEM images of the starch showed that the granules had swelled and coalesced into a gel structure. The presence of residual traces of the swollen granules, which were much more evident after heating to 95 °C than to 125 °C, indicated that the swelling of the starch granules under the conditions in the DSC pans was incomplete. In comparison, SEM of native starch granules after heating and swelling in a large excess of water (for example in swelling power tests) produced a homogenous material with no residual traces of swollen granules (Wang & Copeland, unpublished results). On the basis of the results from the DSC and SEM observations, we conclude that only partial swelling of starch granules took place during DSC, and that the endothermic transition in DSC traces did not represent complete gelatinization of the starch granules. The water/starch ratio employed in DSC measurements is generally between 2:1 and 5:1 (it was 2:1 in the present study), which has been considered sufficient for complete gelatinization of starch (Donovan, 1979). However, some studies have shown that in this amount of water, swelling of starch granules is incomplete (Tester & Morrison, 1990a). Our conclusion is consistent with the demonstration by XRD and SAXS that there is residual crystallinity and lamellar organization at the completion of the DSC endotherm (Jenkins & Donald, 1998; Vermeylen et al., 2006).

After one day of acid hydrolysis, the endothermic transition of the starch became broader, with T_p and T_c shifted to higher temperatures and an increase in ΔH, compared to native starch. On heating for DSC, the starch after one day of acid hydrolysis still had some capacity to swell and form a gel-like structure, although much less than for the native starch. The outlines of the granules were clearly visible and more evident after heating to 95 °C than to 125 °C. The swelling capacity of native starch granules is mainly dependent on the structural integrity of amylopectin and can be greatly reduced as the structure is disrupted (Tester & Morrison, 1990b; Wang & Copeland, unpublished result). The first paper of this series showed that amylose and amylopectin chains were both attacked during the initial stages of acid hydrolysis, and that after one day of acid hydrolysis there is mainly corrosion on the outer surface of the granules, with both amylose and amylopectin molecules remaining relatively intact and only minor effects to the inner part of the granules (Wang, Blazek et al., 2011; Wang et al., 2008).

The behaviour of starch granules that are heated in excess water may be considered to have similarities to the dissolution of a mixture of linear and cross-linked polymers in a compatible solvent, which involves the slow diffusion of the solvent into the polymer matrix followed by rapid chain disentanglement (Miller-Chou & Koenig, 2003). In contrast to low molecular weight solutes which dissolve instantaneously, polymers tend to swell before subsequent dissolution, whereas large, highly branched polymers tend to swell to an equilibrium point without subsequent dissolution (Flory & Krigbaum, 1951). In comparison, two separate events occur when starch granules are heated in excess water: swelling of the macromolecular constituents, followed by their partial dissolution. By analogy, one can propose that on heating in water the linear amylose chains in starch swell and eventually dissolve, whereas most of the large, predominantly intact branched amylopectin molecules only swell. Some smaller amylopectin molecules may be leached with amylose (Doublier, Paton, & Llamas, 1987; Hoover & Vasanthan, 1992; Shamekh et al., 1994; Tester & Karkalas, 1996). The starch residue after acid hydrolysis would have freed of highly soluble small glucan fragments by cold water washing. However, some less soluble linear or branched fragments (referred to subsequently as slightly soluble chains) are likely to have remained and these would have absorbed heat to dissolve. This assumption is supported by parallel studies which show that the majority of acid hydrolysed starch dissolves when heated in a 25:1 (w/w) excess of water (Wang et al., unpublished result). Hence, the endothermic transition of starch after one day of hydrolysis reflects mainly energy uptake due to the swelling of relatively intact starch chains, but there may also be a contribution from dissolution of slightly soluble chains.

The shift to higher temperature of the endothermic transition has long been interpreted as an increase in gelatinization temperature due to acid hydrolysed starch having increased crystallinity. However, the shift to higher temperatures of the endothermic transition is more likely due to broadening of endothermic transition as a result of the occurrence of multiple thermal events. This possibility is consistent with the assumption that the endothermic transition of starch after one day of acid hydrolysis reflects heat input required for both swelling and dissolution of polydisperse molecular chains. In contrast, the occurrence of multiple thermal events could possibly lead to the decrease in endothermic temperature, which is in agreement with some previous experimental observations (Genkina et al., 2007; Perera et al., 2001; Shi & Seib, 1992). The extent of broadening of the endothermic transition on acid hydrolysis, and whether the endothermic temperature shifts to higher or lower temperature, will depend on the structure of the starch chains and the ability of the limited amount of water in DSC pans to penetrate into the starch matrix in the absence of mixing. These factors will influence the sequence and extent to which thermal events that represent swelling and/or dissolution of individual polymer chains occur and contribute to the endotherm, which is a composite for the whole material.

The ΔH of acid-hydrolysed starch can be considered to be the sum of the swelling enthalpy of relatively intact starch chains and dissolution enthalpy of slightly soluble chains. For starch after one day of acid hydrolysis, swelling of relatively intact starch chains predominated over the dissolution of slightly soluble chains, as seen from the gel-like structure observed in SEM images. According to theory of polymer dissolution (Miller-Chou & Koenig, 2003), the endothermic enthalpy would be expected to decrease as a result

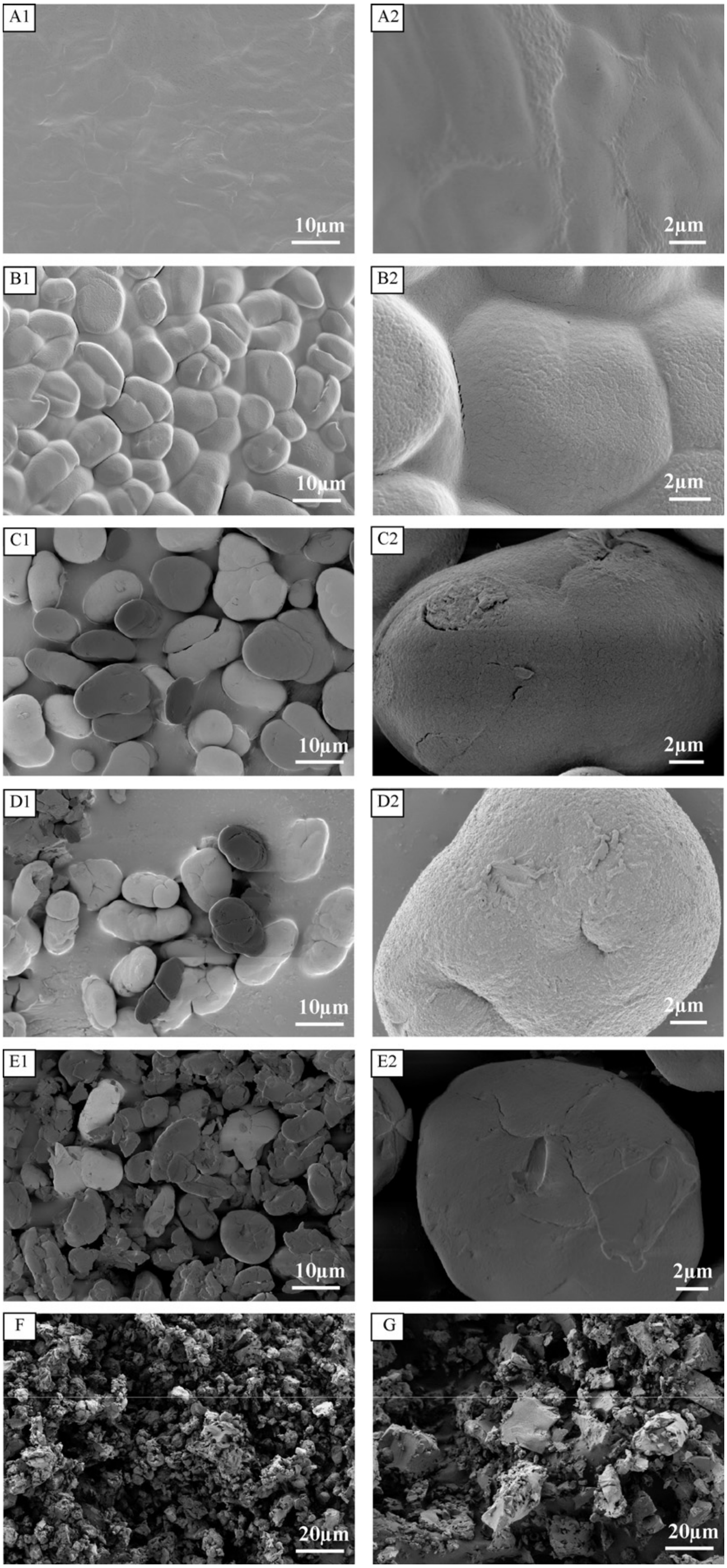

Fig. 4. Scanning electron micrographs of native and acid-hydrolysed starches heated in DSC to 95 °C. The panels show native starch (A1, A2), and starch hydrolysed for 1 (B1, B2), 2 (C1, C2) and 6 (D1, D2), days. The scale and magnification are shown on the panels.

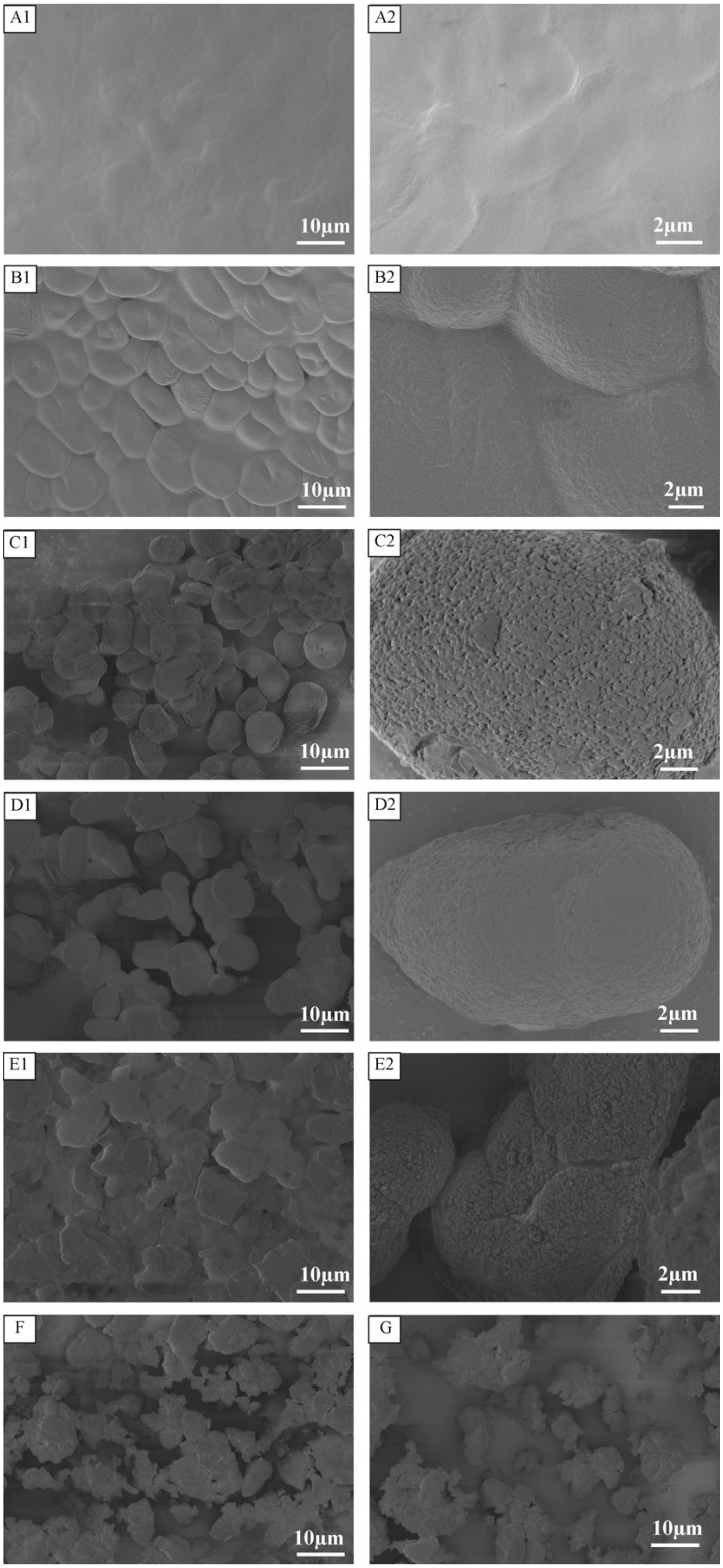

Fig. 5. Scanning electron micrographs of native and acid-hydrolysed starches heated in DSC to 125 °C. The panels show native starch (A1, A2), and starch hydrolysed for 1 (B1, B2), 2 (C1, C2), 6 (D1, D2), 12 (E1, E2) 20 (F) and 35 days (G). The scale and magnification are shown on the panels.

of acid hydrolysis causing a reduction in the swelling power of the starch chains. As there was an increase in endothermic enthalpy, it can be concluded that the increase in endothermic enthalpy must involve the substantial contribution of dissolution enthalpy of slightly soluble chains. This conclusion is consistent with the broadening and shift to higher temperatures of the endothermic transition due to the dissolution of polydisperse molecular chains. Similarly, the enthalpy changes with acid hydrolysis are dependent on the extent of involvement of these endothermic events. Our conclusion is in agreement with the early hypothesis that dissolution of short chains occurs during thermal transitions of acid-hydrolysed starches (Morrison et al., 1993; Muhr et al., 1984). In contrast to the earlier studies, the dissolution enthalpy of the short chains was evident in the DSC traces in the present study.

After two days of hydrolysis, only a broad and undefined endothermic transition was observed. This has long been assumed to be the gelatinization transition characteristic of acid-hydrolysed starches, which has been noticed less reproducible to determine the thermal transition parameters (Atichokudomchai et al., 2002; Jacobs et al., 1998; John, Raja, Rani, Moorthy, & Eliasson, 2002; Sandhu, Singh, & Lim, 2007; Shi & Seib, 1992; Singh, Sodhi, & Singh, 2009). After two and six days of hydrolysis, the interior of the starch granules was also damaged, which resulted in the severe degradation of amylose and amylopectin chains and increased amounts of slightly soluble chains in the acid-hydrolysed starch. On heating in water, these severely degraded starch chains were not able to hold water and swell, and hence dissolution of these chains predominated over the swelling of any remaining intact starch chains. Morphological changes after the DSC measurement provided further evidence for this, in that the acid-hydrolysed starches were visually powdery and had no obvious gel-like form. After prolonged hydrolysis (over 12 days), most of starch granules were cracked and smaller fragments that had broken away were noted. When these cracked starch granules were heated for DSC, they no longer absorbed water to swell but rather they dissolved. Apart from the fusion of dissolved slightly soluble chains, no swelling and subsequent formation of the gel or gel-like structure was observed, but more cracks occurred in the granules. The appearance of more cracks was assumed to be caused by steam pressure produced during heating in the sealed aluminium pan. Damaged starch granules are less resistant to pressure treatment due to cracks in their structure (Bauer, Hartmann, Sommer, & Knorr, 2004). Taken together, these results lead us to conclude that the broad endothermic transition observed in DSC traces of starch subjected to acid hydrolysis for two or more days was predominantly attributable to the dissolution of slightly soluble chains. Because of the polydispersity of the chain fragments their dissolution occurs over a wide temperature range, resulting in a complex endothermic trace.

4.1. Swelling and dissolution of starch chains during heating

In order to test the hypothesis that swelling and/or dissolution of different starch chain segments takes place during heating in DSC pans, the thermal transition of dilute starch–water system (the ratio of water/starch 25 μl:1 mg) was measured under the same DSC conditions. To our knowledge, it is the first time that the thermal transition recording of such dilute starch–water system has been made. This dilute starch–water system showed similar thermal transition traces with those of acid-hydrolysed starches over 2 days, presenting a wide endothermic transition followed by an exothermic transition. The transformation from narrow endotherm observed at a water/starch of 2:1 to the broad one indicated that swelling of the starch chains was partially translated into dissolution, during which continuous heat input was required. This result further supports the above conclusion that the endothermic transition measured by DSC only reflected the limited swelling of starch granules at given water level, which could not result in the complete loss of crystalline/molecular structures (Jenkins & Donald, 1998; Vermeylen et al., 2006). By analogy, we can conclude that upon heating in water, swelling and/or dissolution of multiple chain segments in acid-hydrolysed starches could concomitantly occur depending on the depolymerization degree of starch chains and water availability.

4.2. Nature of the exothermic transition

An important observation made in the present study was the exothermic transition in all of the samples at temperatures between about 105 and 115 °C. The exothermic transition for native starch and acid-hydrolysed starch was previously taken as the conclusion point of the gelatinization endothermic transition (Atichokudomchai et al., 2002; Donovan & Mapes, 1980; Garcia et al., 1996; Jacobs et al., 1998; Komiya & Nara, 1986; Morrison et al., 1993; Muhr et al., 1984; Shi & Seib, 1992). However, what does this exothermic transition really represent if gelatinization does not occur? It is well-known that the phase transition of water from liquid to vapour requires heat input. The broad endothermic transition of water alone was similar to those of acid-hydrolysed starches over two days, except for a reduced intensity of heat input. Hence, it is proposed that the exothermic peak was due to the phase transformation of water from vapour to liquid. The observation of condensed water droplets on the inner crucible lids after DSC measurement after quickly opening the crucible lids further confirmed the above proposal. The difference in the intensity of heat input between water alone and starch that had been treated with acid for two days further indicated that dissolution of starch chains in the residual granules actually occurred and required heat input.

5. Conclusions

The thermal transitions of native and acid-hydrolysed starches have been investigated using the novel concept of semi-preparative DSC and FE-SEM. To our knowledge, this is the first report on morphological changes that native and acid-hydrolysed starches undergo after heating in DSC pans. We conclude that the endothermic transition of native starch (water/starch ratio 2:1) measured by DSC is complex and reflects limited swelling behaviour of starch granules and does not represent complete gelatinization. The broadening of the endothermic transition of starch after one day of acid hydrolysis was concluded to be due to the occurrence of multiple thermal events including swelling of relatively intact starch chains and dissolution of slightly soluble chains. The balance of these events changed as the period of hydrolysis increased. Traditionally assumed gelatinization behaviour did not occur for starches hydrolysed over two days. The broad and undefined endothermic transition in DSC traces was mainly due to the dissolution of slightly soluble chains. The transformation of the shape of the endothermic transition from narrow to broad reflected the changes of thermal transition behaviour from limited swelling of amylose and amylopectin chains to the predominant dissolution of slightly soluble chains and minor swelling of starch chains relatively intact, and to the final dissolution of slightly soluble chains. The exothermic nature of the second transition next to the endothermic transition was elucidated, which represented the phase transition of water from vapour to liquid.

Acknowledgements

SW is the recipient of a University of Sydney Postdoctoral Research Fellowship Scheme (U2527-2009/2012). We thank Dr. Hank De Bruyn for assistance with the DSC, Dr Peter Hines and Dr.

Yanyan Sun in the Australian Centre for Microscopy and Microanalysis for assistance with the FE-SEM, and Dr. Jinglin Yu for helpful discussions and technical support.

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