Carbohydrate Polymers 89 (2012) 777–782

Contents lists available at SciVerse ScienceDirect

Carbohydrate Polymers

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

The effect of rice aging on the freeze–thaw stability of rice flour gels

Wattinee Katekhong, Sanguansri Charoenrein*

Department of Food Science and Technology, Faculty of Agro-Industry, Kasetsart University, Bangkok 10900, Thailand

ARTICLE INFO

Article history:
Received 24 November 2011
Received in revised form 10 February 2012
Accepted 4 April 2012
Available online 11 April 2012

Keywords:
Rice
Aging
Freeze-thaw stability
Gels

ABSTRACT

This study investigated the effect of aging rice on the freeze–thaw stability of rice flour gels since repeated freeze–thaw cycles can lead to reduced food quality. A rice grain cultivar called 'Khoa Dawk Mali 105' was aged for three different time periods, ranging from 0 to 12 months. Rice gels made from the aged rice were then freeze–thawed for up to 5 cycles. Repeated freeze–thaw cycles lead to an increase in syneresis values and hardness with increasing rice aging. Differential scanning calorimetry showed an increase in the enthalpy of melting of the amylose–lipid complex after 5 freeze–thaw cycles and an increase in peak gelatinization temperature and gelatinization enthalpy with longer rice aging. Moreover, aging length of the rice correlated significantly with a decrease in peak viscosity and breakdown but also an increase in final viscosity and setback. These results demonstrate that aging the rice reduced the freeze–thaw stability of the rice flour gels.

1. Introduction

In recent years, the ready-meal market has grown in developed and developing countries. Many ready-to-eat meals including starch-based frozen food products have been developed and launched upon the world market. During the freezing process, when starch pastes or gels are frozen, phase separation can occur upon formation of ice crystals. Then when thawing, a phenomenon known as syneresis occurs in which starch pastes and gels harden because the water can be easily expressed from the dense network (Karim, Norziah, & Seow, 2000). Repeated freezing and thawing cycles encourage phase separation and ice growth. As ice crystals become larger, syneresis and sponge formation occurs more readily (Eliasson & Kim, 1992). Multiple freeze–thaw cycles wherein samples are repeatedly frozen and then intermittently thawed to room temperature over a period of 2–4 h are known to drastically accelerate retrogradation and syneresis (Radley, 1976).

Starch-based frozen food products undergo textural changes related to amylose and amylopectin retrogradation and show syneresis after thawing. These changes have been attributed to starch retrogradation (Ferrero, Martino, & Zaritzky, 1994; Jacobson & BeMiller, 1998; Varavinit, Anuntavuttikul, & Shobsngob, 2000) and may make such products unacceptable to consumers (Ferrero, Martino, & Zaritzky, 1993).

Aging of rice is a normal step between harvest and consumption. In addition, a rice stock is necessary as an insurance against possible crop failures or poor yields in the following year. During aging of stored rice, a number of physicochemical properties of the rice are subject to change (Chrastil, 1990; Perdon, Marks, Siebenmorgen, & Reid, 1997; Villareal, Resurreccion, Suzuki, & Juliano, 1976; Zhou, Robards, Helliwell, & Blanchard, 2003). The changes which occur during the aging process have been reported to be greater in non-waxy rice than in waxy rice (Villareal et al., 1976). Perez and Juliano (1981) reported that 3 months was the minimum aging period for major changes to occur in the hardness of cooked rice, gel consistency and amylograph viscosity values. The texture of the cooked aged rice was found to be harder and less sticky than when the same freshly harvested rice was cooked. Moreover, the aged rice was found to exhibit an increased volume expansion and water absorption during the cooking process (Noomhorm, Kongseree, & Apintanapong, 1997; Pushpamma & Reddy, 1979). Chrastil (1990) reported that the number of disulfide bonds and the average molecular weight of oryzenin, which is a major protein in rice, increased during aging of the rice grains. Such changes in protein properties contribute to the effect of aging on the pasting properties of rice (Zhou et al., 2003). Removal of starch surface protein and lipid from wheat and maize starches led to more swollen and fragile granule than untreated starch (Debet & Gidley, 2006, 2007). Hamaker and Griffin (1993) reported that proteins with disulfide bonds in the rice flour restrict starch granules from swelling during gelatinization and made the swollen granules less susceptible to disruption by shear. The gelatinization temperature and gelatinization enthalpy increased with increasing rice aging times (Fan & Marks, 1999). Teo, Karim, Cheah, Norziah, and Seow (2000) found that the thermal properties of purified rice starch remained unchanged upon aging and concluded that it was the non-starch constituents, particularly protein, which were responsible for the increased gelatinization temperature and enthalpy in the rice flour. Several researchers

* Corresponding author. Tel.: +662 562 5025; fax: +662 562 5021.
E-mail address: fagisscr@ku.ac.th (S. Charoenrein).

0144-8617/$ – see front matter
http://dx.doi.org/10.1016/j.carbpol.2012.04.007

also reported that the peak viscosity and breakdown of fresh rice flour was higher than in aged rice flour (Noomhorm et al., 1997; Tulyathan & Leeharatanaluk, 2007; Zhou, Robards, Helliwell, & Blanchard, 2002; Zhou et al., 2003).

However, no reports are as yet available on the effects of rice aging time on the freeze–thaw stability of rice flour gels. Freeze–thaw stability is an important property that is used to evaluate the ability of starch to withstand the undesirable physical changes which occur during freezing and thawing. Therefore, the objective of this study was to determine the freeze–thaw stability, i.e. syneresis values and textural properties of rice flour gels made from rice aged for 0 to 12 months. The microstructures of the freeze–thawed rice flour gels as well as the thermal and pasting properties of these rice flour samples were also examined. The insights gained in this research have the potential to help improve the quality of frozen rice-based products.

2. Materials and methods

2.1. Materials

Milled rice of the Khao Dawk Mali 105 (KDML 105) cultivar with three different aging time periods, i.e. 0 (newly harvested), 3 and 12 months were examined in this study. Samples were packed in nylon pouches under vacuum and aged at room temperature (25 ± 2 °C). The samples were then ground and sieved for analysis. Using the method of Juliano (1971), the apparent amylose content of rice aged for 0, 3 and 12 months was determined to be 22.0%, 21.9% and 20.6%, respectively. The moisture content of rice aged for 0, 3 and 12 months were 10.20, 9.09 and 9.63%, respectively (AACC, 2000).

2.2. Flour gel preparation

The flour gels were prepared following the method of Charoenrein, Tatirat, and Muadklay (2008). Rice flour suspensions (11% total solid w/w wet basis) were prepared by mixing the flour in distilled water and stirring continuously at 250 rpm for 1 h followed by 200 rpm at 85 °C for 25 min. The suspensions were then loaded into 10 ml syringes (20 mm in diameter) for syneresis measurements and loaded into a 60 ml aluminum cans (size 5 oz.) for texture measurements. After that, the samples were steamed for 9 min and placed in an incubator at 25 °C for 2 h.

2.3. Freezing and thawing

Flour gel samples were frozen in a cryogenic cabinet freezer (Minibatch 1000 L; Bangkok Industrial Gas Co., Bangkok, Thailand) which allowed the flow rate of liquid nitrogen to be adjusted creating a cold atmosphere of −20 °C. Then the samples were stored in a chest freezer (Sanyo refrigerator, model SF-C1497) at −18 °C for 22 h and thawed at ambient temperature (25 ± 2 °C) for 2 h. This freeze–thaw cycle was repeated for up to 5 cycles. The freezing experiments were carried out in two separate trials.

2.4. Syneresis measurement

The syneresis measurements followed the method of Charoenrein et al. (2008). The thawed flour gel samples were removed from their syringes and put in a cylindrical plastic tube with a perforated bottom which was covered with filter paper (Whatman No. 41). These tubes were then placed in centrifuge tubes and centrifuged at 100 × g (centrifuge CN-1050, MRC Ltd., Holon, Israel) for 15 min. The cylindrical plastic tube with cover was then removed from the centrifuge tube and the liquid which had separated from the flour gel was weighed. The percentage of syneresis was calculated as the ratio of the weight of liquid separated from the gel to the total weight of the gel before centrifugation and multiplied by 100. The data were reported as the average of five measurements.

2.5. Texture measurement

Texture was measured using a Stable Micro System (TA-XT plus) Texture Analyzer (five replicates per treatment). Thawed flour gel samples which had been stored in an aluminum can (25 mm) were compressed with a P/25 cylinder probe at the test speed of 1 mm/s. The deformation level was 10 mm. The maximum force from the texture profile was recorded as the hardness of the flour gel. The slope of the linear portion of the curve which is related to the elastic properties (Kidmose & Martens, 1999) was also recorded.

2.6. Frozen structure by scanning electron microscope (SEM)

The freeze–thawed samples were cut into sections of 1–3 mm thickness using a razor blade and gradually dehydrated in 50%, 70%, 90% and absolute ethanol at room temperature for 24 h at each concentration and finally dehydrated using a critical point dryer. The cut surface samples were mounted on a stub, coated with gold and observed with a JSM-5600LV microscope (JEOL, England). The accelerating voltage and magnification are shown on the micrographs.

2.7. Retrogradation properties

The retrogradation properties of samples freeze–thawed for 1 and 5 cycles were determined using a differential scanning calorimeter (DSC, Pyris-1, Perkin Elmer, Norwalk, CT, USA). After the gelatinization properties (Section 2.9) were analyzed, the samples were prepared in a stainless steel pan and then stored in a chest freezer (Sanyo refrigerator, model SF-C1497) at −18 °C for 22 h and thawed at room temperature (25 ± 2 °C) for 2 h. The samples were heated in the DSC at a rate of 10 °C/min from 25 to 140 °C. The peak temperature (T_p) and enthalpy of melting the amylose–lipid complex were calculated. All measurements were performed in two replicates.

2.8. Pasting properties

The pasting properties of the rice flour suspensions (8%, w/w) were determined using a Rapid Visco Analyser (RVA3D, Newport Scientific Instrument & Engineering, Australia) (AACC, 2000). All measurements were performed in triplicate.

2.9. Gelatinization properties

The gelatinization properties of flour were modified from methods of Thirathumthavorn and Charoenrein (2005) and analyzed using DSC (Pyris-1, Perkin Elmer, Norwalk, CT, USA). Rice flour (6 mg, dry basis) at a 70% moisture content was prepared in a stainless steel pan. Samples were hermetically sealed and allowed to stand for 1 h at room temperature (25 ± 2 °C) before being heated in the DSC. The empty stainless steel pan was used as a reference. The samples were heated at a rate of 10 °C/min from 25 °C to 140 °C and cooled at a rate of 2.5 °C/min to 25 °C.

2.10. Statistical analysis

The experiments were ordered in a completely randomized design. The data were analyzed using an analysis of variance (ANOVA) and the differences between means were determined

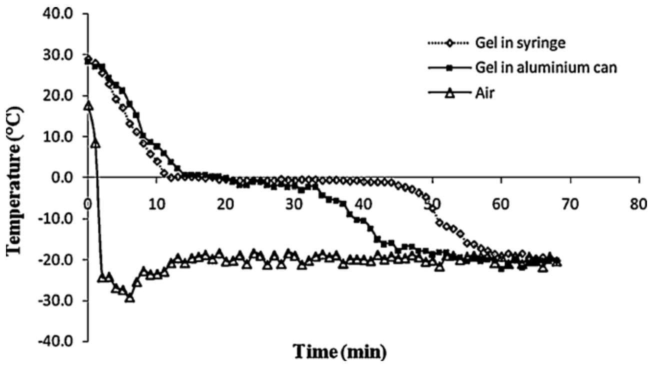

Fig. 1. Freezing profile of rice flour gels (11%, w/w) at −20 °C in a cryogenic freezer.

using the Duncan's new multiple range test. All statistical analyses were performed using SPSS 12.0 for windows.

3. Results and discussion

3.1. Freezing of rice flour gel

Fig. 1 shows the freezing curve for freezing rice flour gel at −20 °C in a cryogenic freezer. It should be noted that the temperature inside the freezer was reduced to −20 °C in only 2 min due to the high efficiency of the freezer. In this study, the freezing time of the flour gels for the syneresis measurement was 70 min and the freezing time of the flour gels for the texture measurement was 57 min. A previously reported undercooling phenomenon that can cause a low % syneresis and high hardness (Charoenrein & Preechathammawong, 2010) was not detected.

3.2. % Syneresis

The determination of % syneresis from the freeze–thawed flour gels is used to evaluate the ability of starch to withstand the undesirable physical changes which occur during freezing and thawing (Charoenrein & Preechathammawong, 2010). Syneresis in freeze–thawed gels is due to an increase in molecular associations between starch chains, in particular the retrogradation of amylose (Morris, 1990) which results in the expulsion of water from the gel structure (Saartratra, Puttanlekb, Rungsardthong, & Uttapap, 2005). Thus, the amount of water released due to syneresis is a useful indicator of the tendency of starch to retrograde (Karim et al., 2000).

For the first to fifth cycles, the analysis of variance results showed that storage duration of the milled rice affected the percentage of syneresis of the rice flour gels, in particular during the fourth and fifth cycles. The results showed a significant increase in % syneresis with increasing time periods of rice aging (Table 1). Rice flour gels from rice aged for 0 months were found to have a 0.44% syneresis after the first cycle, which only showed a slight increase to 0.83 after 2 freeze–thaw cycles. After that the syneresis value changed dramatically through cycles 3–5. Rice flour gels made from rice aged for 3 months had % syneresis values which were not significantly different ($P > 0.05$) to rice flour gels made from rice aged for 0 months. On the other hand, rice flour gels made form rice aged for 12 months had a 0.56% syneresis after the first cycle, and showed a progressive increase in syneresis value of 2.02–10.76% through the next 3 cycles. Furthermore, after the fourth and fifth freeze–thaw cycles, the % syneresis greatly increased to 27.76 and 36.38, respectively. These results were significantly different ($P \leq 0.05$) from rice aged for 0 and 3 months. Furthermore, the analysis of variance results showed an interaction between aging time and a number of freeze–thaw cycle to % syneresis of rice flour gels ($P \leq 0.05$). The increase in the % syneresis was attributed to an increase in retrogradation of the rice flour gels. This result was correlated to setback (Section 3.6) which increased with increasing aging duration of the rice, implying an increase in retrogradation of the amylose. To study freeze–thaw stability, we used the textural properties, microstructure (SEM) and retrogradation properties of the freeze-thawed flour gels as described in Sections 3.3–3.5, respectively, to better understand the effects of rice aging on the freeze–thaw stability of the rice flour gels. These results clearly show that the aging of rice affected the freeze–thaw stability of the rice flour gels.

3.3. Texture

KDML 105 rice has a low amylose content ranging from 12 to 20% (Juliano, 1992; Prathepha, Daipolmak, Samappito, & Baimai, 2005). Therefore, flour gels generated from KDML 105 are soft gels. In our study, the amylose content of the rice aged for 0–12 months did not change significantly (21.96–20.60%). Similar results were reported by some other investigators (Indudhara Swamy, Sowbhagya, & Bhattacharya, 1978; Noomhorm et al., 1997; Villareal et al., 1976).

The texture properties of the rice flour gels before and after the freeze–thaw cycle were determined and the results are shown in Table 2. The analysis of variance results showed an

Table 1
Percent of water separated (syneresis) of rice flour gels made from KDML 105 rice aged for different durations.

Storage duration (months)	% Syneresis				
	1 cycle	2 cycle	3 cycle	4 cycle	5 cycle
0	$0.44^{aA} \pm 0.04$	$0.83^{aA} \pm 0.20$	$7.71^{aB} \pm 1.42$	$17.28^{aC} \pm 1.10$	$29.08^{aD} \pm 0.67$
3	$0.54^{aA} \pm 0.18$	$1.45^{bA} \pm 0.08$	$7.12^{aB} \pm 0.66$	$18.03^{aC} \pm 0.13$	$29.97^{aD} \pm 1.39$
12	$0.56^{aA} \pm 0.17$	$2.02^{cA} \pm 0.20$	$10.76^{aB} \pm 1.72$	$27.76^{bC} \pm 1.01$	$36.38^{bD} \pm 0.43$

[a–c]Means with different letters in the same column are significantly different ($P \leq 0.05$).
[A–D]Means with different letters in the same row are significantly different ($P \leq 0.05$).

Table 2
Hardness of the rice flour gels made from milled KDML 105 rice aged for different durations.

Storage duration (months)	Hardness (N)			
	Fresh	1 cycle	3 cycle	5 cycle
0	$0.99^{aA} \pm 0.01$	$0.96^{aA} \pm 0.05$	$1.87^{aB} \pm 0.35$	$2.84^{aC} \pm 0.06$
3	$1.04^{aA} \pm 0.02$	$0.92^{aA} \pm 0.02$	$1.70^{aB} \pm 0.24$	$2.90^{aC} \pm 0.16$
12	$1.05^{aA} \pm 0.05$	$1.02^{aA} \pm 0.09$	$1.89^{aB} \pm 0.25$	$3.75^{bC} \pm 0.07$

[a,b]Means with different letters in the same column are significantly different ($P \leq 0.05$).
[A–C]Means with different letters in the same row are significantly different ($P \leq 0.05$).

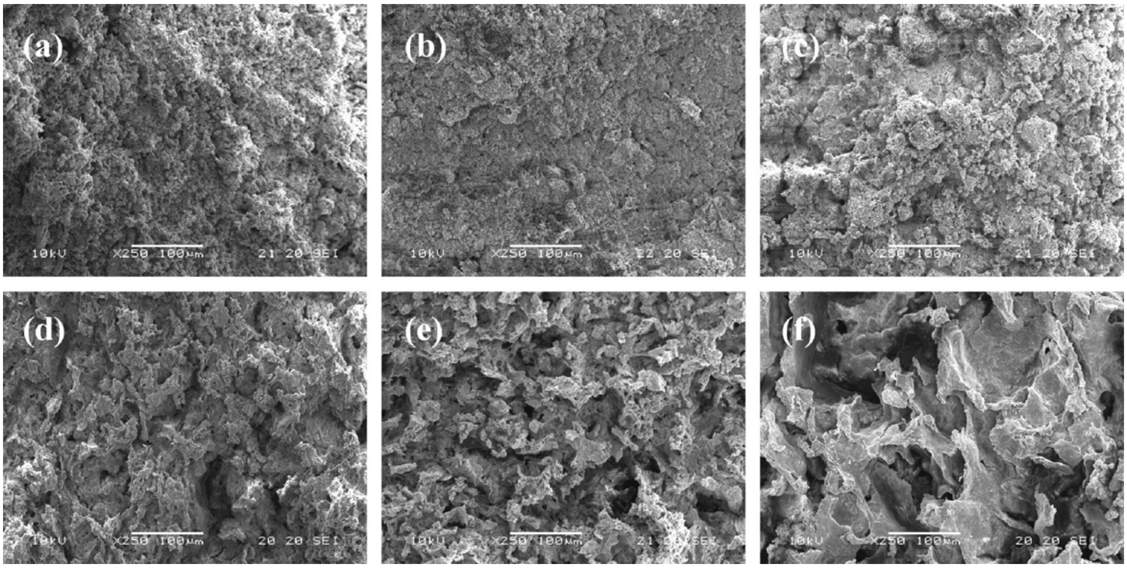

Fig. 2. SEM images of unfrozen and the fifth freeze–thaw cycle of the rice flour gels from rice aged for 0 month (a, d), 3 months (b, e) and 12 months (c, f), respectively.

interaction between aging time and a number of freeze–thaw cycle to the hardness of rice flour gels ($P \leq 0.05$). The results showed that the hardness values of all samples increased after the repeated freeze–thaw cycles. However, all samples of the rice flour gel were not significantly different ($P > 0.05$) from fresh gels, and gels freeze–thawed for 1–3 cycles. On the other hand, after the fifth cycle, the rice flour gels made from rice aged for 12 months had significantly ($P \leq 0.05$) higher hardness values than rice only aged for 0 and 3 months. As can be seen from the SEM images of the freeze–thaw gels (Fig. 2), the spongy structure of the rice flour gels for rice aged for 12 months (Fig. 2f) was thicker than that of rice flour gels made from rice aged for 0 months (Fig. 2d). This could explain the harder texture noted in freeze–thawed flour gels with longer aging durations. In addition, a slope in a linear portion of the curve of all samples was significantly ($P \leq 0.05$) increased after freeze–thaw cycles (Table 3). The high values of this parameter indicate a firm sample (Kidmose & Martens, 1999). These results showed that the flour gels became harder with repeated freeze–thaw cycle.

3.4. Structure of freeze–thaw gels

To elucidate the relationship between the syneresis and structure of rice flour gels, the microstructure of the freeze–thawed gels were examined using SEM. Specimen images are shown in Fig. 2. Clear differences were observed in the microstructure of rice flour gels with different aging times after 5 freeze–thaw cycles. The repeated freeze–thawed of flour gels made a spongy structure of all samples clearly observation. The pores resulting from ice crystal formation were more clearly seen and the matrix surrounding the pores was thicker due to increasing retrogradation of the starch matrix from repeated freeze–thaw cycles (Charoenrein et al., 2008). In the fifth freeze–thaw cycle, rice flour gels aged for 0 months appeared to have smaller pores embedded in a weak matrix and a texture that was visually similar in appearance to mashed wet tissue paper (Fig. 2d). Flour gels made from rice aged for 3 months had slightly larger pores and a thicker matrix surrounding the pores than flour gels made from rice aged for 0 months. Furthermore, the microstructure of the flour gels from rice aged for 12 months was characterized by large pores in the gel and the matrix surrounding the pores which were thicker than flour gels made from rice aged for 0 and 3 months (Fig. 2f). The changes in microstructure correlated closely with the increase in % syneresis and hardness of the flour gels. These specimen images showed that aging of rice reduced the freeze–thaw stability of the rice flour gels.

3.5. Retrogradation properties

For the retrogradation of rice flour gels after the freeze–thaw cycle (Table 4), the result showed that the DSC could not detect a melting endotherm of a crystallized amylopectin occurring at temperature less than 100 °C. This implies that the freeze–thaw cycle does not produce a long term amylopectin retrogradation. However, we found a melting peak at a T_p of 101–102 °C which corresponded to a melting of crystallized amylose–lipid complex in the temperature range 94–106 °C (Vandeputte, Vermeylen, Geeroms, & Delcour, 2003).

In the first and fifth freeze–thaw cycles, the rice flour gels of all samples were not significantly different ($P > 0.05$) in melting peak temperature of the amylose–lipid complex. However, all samples showed an increase in enthalpy of the melting amylose–lipid complex, through 1–5 freeze–thaw cycles. In the first freeze–thaw cycle, the enthalpy of amylose–lipid complex of rice flour made from rice aged for 12 months was 0.7 J/g which was higher than for rice flour

Table 3
Texture of the rice flour gels made from milled KDML 105 rice aged for different durations expressed by slope.

Storage duration (months)	Slope (N/mm)			
	Fresh	1 cycle	3 cycle	5 cycle
0	$0.19^{aA} \pm 0.00$	$0.18^{aA} \pm 0.01$	$0.30^{aB} \pm 0.04$	$0.48^{aC} \pm 0.01$
3	$0.21^{bA} \pm 0.00$	$0.19^{aA} \pm 0.02$	$0.31^{aB} \pm 0.02$	$0.49^{aC} \pm 0.01$
12	$0.21^{bA} \pm 0.00$	$0.20^{aA} \pm 0.01$	$0.32^{aB} \pm 0.04$	$0.52^{aC} \pm 0.02$

^{a,b}Means with different letters in the same column are significantly different ($P \leq 0.05$).
$^{A–C}$Means with different letters in the same row are significantly different ($P \leq 0.05$).

Table 4
Thermal properties of KDML 105 rice aged for different durations after 1 and 5 freeze–thaw cycles.

Storage duration (months)	Amylopectin retrogradation	Amylose–lipid complex			
		T_p (°C)		ΔH (J/g)	
		Cycle 1	Cycle 5	Cycle 1	Cycle 5
0	nd	$102.3^{aA} \pm 0.4$	$101.4^{aA} \pm 0.0$	$0.4^{aA} \pm 0.0$	$0.6^{aB} \pm 0.0$
3	nd	$101.0^{aA} \pm 0.5$	$101.8^{aA} \pm 0.6$	$0.4^{aA} \pm 0.0$	$0.7^{bB} \pm 0.0$
12	nd	$101.0^{aA} \pm 1.6$	$101.7^{aA} \pm 0.5$	$0.7^{aA} \pm 0.2$	$0.9^{cA} \pm 0.0$

$^{a-c}$Means with different letters in the same column are significantly different ($P \le 0.05$).
$^{A-B}$Means with different letters in the same row are significantly different ($P \le 0.05$).
T_p = Peak temperature and ΔH = Enthalpy of melting of the amylose–lipid complex.
nd: the peak temperature and retrogradation enthalpy was not detected.

Table 5
Pasting properties of KDML 105 rice aged for different durations.

Storage duration (months)	Peak viscosity (RVU)	Trough (RVU)	Breakdown (RVU)	Final viscosity (RVU)	Setback (RVU)	Pasting temperature (°C)
0	$133.36^{c} \pm 1.00$	$96.69^{a} \pm 2.54$	$36.67^{c} \pm 1.59$	$171.64^{a} \pm 1.40$	$74.94^{a} \pm 1.18$	$87.43^{a} \pm 1.92$
3	$130.06^{b} \pm 0.80$	$97.64^{a} \pm 3.08$	$33.42^{b} \pm 1.92$	$179.25^{b} \pm 2.16$	$81.61^{b} \pm 0.92$	$89.43^{ab} \pm 1.30$
12	$120.22^{a} \pm 0.77$	$104.97^{b} \pm 0.34$	$15.25^{a} \pm 0.43$	$201.25^{c} \pm 1.30$	$96.28^{c} \pm 0.96$	$91.92^{b} \pm 0.98$

$^{a-c}$Means with different letters in the same column are significantly different ($P \le 0.05$).

aged for 0 months (0.4 J/g). Some researchers reported an increase in amylose–lipid complex formation due to aging of rice (Zhou et al., 2002, 2003). During the storage of rice, lipid hydrolysis was initiated by the action of lipases. This resulted in an increase of free fatty acids which could complex with amylose to amylose–lipid complex (Dhaliwal, Sckhon, & Nagi, 1991; Nishiba, Sato, & Suda, 2000). Thus, the enthalpy of the amylose–lipid complex of rice aged for 12 months was higher than for rice aged for 0 and 3 months. Moreover, the enthalpy of the amylose–lipid complex of all of the samples after 5 freeze–thaw cycles was higher than after 1 freeze–thaw cycle. When a starch gel is frozen, starch-rich regions are created in the matrix. High solid concentration in the regions facilitates the starch chains to associate forming thick filaments, whereas water molecules aggregate into ice crystals forming a separate phase. Upon thawing, ice transforms to bulk phase water, which can be readily released from the polymeric network (syneresis) (Ferrero et al., 1993). The expulsion of water from the gel structure causes the amylose chains to come close to a free fatty acid which allows an increase in the amount of amylose–lipid complex. The repeated freezing and thawing increases this formation and thus increases the melting enthalpy of the amylose–lipid complex.

3.6. Pasting properties

The pasting properties of the milled rice flour after aging are shown in Table 5. From the RVA analysis, the samples showed significant decreases in peak viscosity and breakdown after being aged. The change in some of the pasting properties during aging can be attributed to starch granule characteristics. As reported by Whistler and Paschall (1965), the height of peak viscosity during the heating cycle is a measure of the ability of the rice granules to swell markedly before rupture. The changes in peak viscosity show that the starch granules of aged rice were more resistant to swelling than those of fresh rice. The decrease in breakdown indicates that the capacity of the starch granules to rupture after cooking was reduced significantly by aging of the starch granules (Noomhorm et al., 1997; Tulyathan & Leeharatanaluk, 2007; Zhou et al., 2003). Our findings agree with those of Tulyathan and Leeharatanaluk (2007) who found that the peak viscosity and breakdown of KDML 105 decreased with longer aging times, reaching a plateau after 6 months of aging time. In addition, the final viscosity and setback of the milled rice flour showed a significant increase with increasing rice aging duration. The direct relationship between increased setback and increased retrogradation of amylose for longer aging times correlates well with the previously discussed increases in % syneresis (Section 3.2) and rice flour gel hardness (Section 3.3).

Table 6
Gelatinization properties of KDML 105 rice aged for different durations.

Storage duration (months)	T_p (°C)	ΔH (J/g)
0	$67.0^{a} \pm 0.5$	$1.7^{a} \pm 0.0$
3	$67.1^{a} \pm 0.4$	$1.7^{a} \pm 0.0$
12	$70.3^{b} \pm 1.1$	$2.0^{b} \pm 0.1$

$^{a-b}$: Means with different letters in the same column are significantly different ($P \le 0.05$), T_p = Peak temperature and ΔH = Gelatinization enthalpy.

3.7. Gelatinization properties

The double helical and crystalline structures of starches are disrupted during gelatinization. The gelatinization properties of milled rice flour made from rice aged for varying durations are shown in Table 6. Rice aged for 12 months had the highest peak temperature and gelatinization enthalpy; and these parameters were significantly different from rice aged for 0 and 3 months. For the shorter 0 and 3 months aging times, the peak temperature and gelatinization enthalpy were not significantly different. These results indicate that the extended aging of rice restricted the gelatinization of starch granules. This property was most likely due to the molecular weight of oryzenin which increased significantly after extended rice aging, and which correlated with an increase in disulphide bonding (Chrastil, 1990; Chrastill & Zarins, 1992; Martin & Fitzgerald, 2002). This resulted in starch granules of rice aged for 12 months which were more resistant to gelatinization than rice aged for 0 or 3 months.

4. Conclusions

Increased rice aging times caused an increase in % syneresis and hardness of rice flour gels. Repeated freeze–thaw cycles also led to increases in % syneresis and hardness of these gels. These results were also shown to be related to changes in the microstructure of the rice flour gels which showed an increase in retrogradation after 5 freeze–thaw cycles. In addition, an increase in amylose–lipid formation was found after repeated freeze–thaw cycles and after longer aging times. These results suggest that the aging of milled rice influences the pasting and gelatinization properties of rice flour which correlated to a reduction in the freeze–thaw stability of rice flour gels.

Acknowledgements

Financial supports from the Graduate School, Kasetsart University (Graduate Study Research for International Publication) and the Kasetsart University Research and Development Institute (KURDI) are gratefully acknowledged.

References

AACC. (2000). *Approved methods of American Association of Cereal Chemists* (10th ed.). St. Paul, MN: American Association of Cereal Chemists.

Charoenrein, S., & Preechathammawong, N. (2010). Undercooling associated with slow freezing and its influence on the microstructure and properties of rice starch gels. *Journal of Food Engineering*, *100*, 310–314.

Charoenrein, S., Tatirat, O., & Muadklay, J. (2008). Use of centrifugation–filtration for determination of syneresis in freeze–thaw starch gels. *Carbohydrate Polymers*, *73*, 143–147.

Chrastil, J. (1990). Protein-starch interaction in rice grains. Influence of storage on oryzenin and starch. *Journal of Agricultural and Food Chemistry*, *38*, 1804–1809.

Chrastill, J., & Zarins, Z. M. (1992). Influent of storage on peptide subunit composition of rice oryzenin. *Journal of Agricultural and Food Chemistry*, *40*, 927–930.

Debet, M., & Gidley, M. J. (2006). Three classes of starch granule swelling: Influence of surface proteins and lipid. *Carbohydrate Polymers*, *64*, 452–465.

Debet, M., & Gidley, M. J. (2007). Why do gelatinized starch granules not dissolve completely? Role for amylose, protein, and lipid in granule ghost integrity. *Journal of Agricultural and Food Chemistry*, *55*, 4752–4760.

Dhaliwal, Y. S., Sckhon, K. S., & Nagi, H. P. S. (1991). Enzymatic activities and rheological properties of stored rice. *Cereal Chemistry*, *68*, 18–21.

Eliasson, A. C., & Kim, H. R. (1992). Changes in rheological properties of hydroxypropyl potato starch pastes during freeze–thaw treatments. 1. A rheological approach for evaluation of freeze–thaw stability. *Journal of Texture Studies*, *23*, 279–295.

Fan, J., & Marks, B. P. (1999). Effects of rough rice storage conditions on gelatinization and retrogradation properties of rice flours. *Cereal Chemistry*, *76*, 894–897.

Ferrero, C., Martino, M. N., & Zaritzky, N. E. (1993). Stability of frozen starch pastes: Effect of freezing, storage and xanthan gum addition. *Journal of Food Processing and Preservation*, *17*, 191–211.

Ferrero, C., Martino, M. N., & Zaritzky, N. E. (1994). Corn starch-xanthan gum interaction and its effect on the stability during storage of frozen gelatinized suspensions. *Starch/Stärke*, *46*, 481–498.

Hamaker, B. R., & Griffin, V. K. (1993). Effect of disulfide bond containing protein on rice starch gelatinization and pasting. *Cereal Chemistry*, *70*, 377–380.

Indudhara Swamy, Y. M., Sowbhagya, C. M., & Bhattacharya, K. R. (1978). Changes in the physicochemical properties of rice with aging. *Journal of the Science of Food and Agriculture*, *29*, 627–639.

Jacobson, M. R., & BeMiller, J. N. (1998). Method for determining the rate and extent of accelerated starch retrogradation. *Cereal Chemistry*, *75*, 22–29.

Juliano, B. O. (1971). A simplified essay for milled-rice amylase. *Cereal Science Today*, *16*, 334–360.

Juliano, B. O. (1992). Structure, chemistry and function of the rice grain and its fraction. *Cereal Foods World*, *37*, 772–779.

Karim, A. A., Norziah, M. H., & Seow, C. C. (2000). Methods for the study of starch retrogradation. *Food Chemistry*, *71*, 9–36.

Kidmose, U., & Martens, H. J. (1999). Changes in texture, microstructure and nutritional quality of carrot slices during blanching and freezing. *Journal of the Science of Food and Agriculture*, *79*, 1747–1753.

Martin, M., & Fitzgerald, M. A. (2002). Protein in rice grains influence cooking properties. *Journal of Cereal Science.*, *36*, 285–294.

Morris, V. J. (1990). Starch gelation and retrogradation. *Trends in Food Science and Technology*, *1*, 2–6.

Nishiba, Y., Sato, T., & Suda, I. (2000). Convenient method to determine free fatty acid of rice using thin-layer chromatography and flame-ionization detection system. *Cereal Chemistry*, *77*, 223–229.

Noomhorm, A., Kongseree, N., & Apintanapong, M. (1997). Effect of aging on the quality of glutinous rice crackers. *Cereal Chemistry*, *74*, 12–15.

Perdon, A. A., Marks, B. P., Siebenmorgen, T. J., & Reid, N. B. (1997). Effect of rough rice storage condition on the amylograph and cooking properties of medium grain cv. Bengal. *Cereal Chemistry*, *74*, 864–867.

Perez, C. M., & Juliano, B. O. (1981). Texture changes and storage of rice. *Journal of Texture Studies*, *12*, 321–333.

Pushpamma, P., & Reddy, M. U. (1979). Physico-chemical changes in rice and jowar stored in different agro climatic regions of Andhra Pradesh. *Bulletin of Grain Technology*, *17*, 97–108.

Prathepha, P., Daipolmak, V., Samappito, S., & Baimai, V. (2005). An assessment of alkali degradation, waxy protein and their relation to amylose content in Thai rice cultivars. *ScienceAsia*, *31*, 69–75.

Teo, C. H., Karim, A. A., Cheah, P. B., Norziah, M. H., & Seow, C. C. (2000). On the roles of protein and starch in the aging of non-waxy rice flour. *Food Chemistry*, *69*, 229–236.

Thirathumthavorn, D., & Charoenrein, S. (2005). Thermal and pasting properties of acid-treated rice starches. *Starch/Stärke*, *57*, 217–222.

Tulyathan, V., & Leeharatanaluk, B. (2007). Change in quality of rice (*Oryza sativa* L.) cv. Khao Dawk Mali 105 during storage. *Journal of Food Biochemistry*, *31*, 415–425.

Radley, J. A. (1976). *Industrial uses of starch and its derivative*. London: Applied Sciences Publishers.

Saartratra, S., Puttanlekb, C., Rungsardthong, V., & Uttapap, D. (2005). Paste and gel properties of low-substituted acetylated canna starches. *Carbohydrate Polymers*, *61*, 211–221.

Vandeputte, E. G., Vermeylen, R., Geeroms, J., & Delcour, J. A. (2003). Rice starch. I. Structural aspects provide insight into crystallinity characteristics and gelatinization behavior. *Journal of Cereal Science*, *38*, 61–68.

Varavinit, S., Anuntavuttikul, S., & Shobsngob, S. (2000). Influence of freezing and thawing techniques on stability sago and tapioca starch pastes. *Starch/Stärke*, *52*, 214–217.

Villareal, R. M., Resurreccion, A. P., Suzuki, L. P., & Juliano, B. O. (1976). Changes in physicochemical properties of rice during storage. *Starch/Stärke*, *28*, 88–94.

Whistler, R. L., & Paschall, E. F. (1965). *Starch: Chemistry and technology*. New York, NY: Academic Press.

Zhou, Z., Robards, K., Helliwell, S., & Blanchard, C. (2002). Aging of stored rice: Change in chemical and physical attributes. *Journal of Cereal Science*, *35*, 65–78.

Zhou, Z., Robards, K., Helliwell, S., & Blanchard, C. (2003). Effect of rice storage on pasting properties of rice flour. *Food Research International*, *36*, 625–634.