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Rice Amylopectin Fine Structure Variability Affects Starch Digestion Properties

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The possibility to identify or develop new rice cultivars with low glycemic response was investigated. Twelve rice cultivars with a narrow range of amylose contents were selected based on their wide variation in rapid viscoanalyzer (RVA) pasting breakdown to study the relationship between starch digestibility and amylopectin fine structure and pasting properties. Rice flour samples were cooked for in vitro digestibility analysis using the standard Englyst assay. RVA was performed for pasting properties of starches. Results showed that rapidly digestible starch (RDS) was highly and negatively correlated ($r = -0.86$, $p < 0.01$; $r = -0.81$, $p < 0.01$) with FrI long and FrII intermediate/short debranched amylopectin linear chains, respectively, and positively correlated ($r = 0.79$; $p < 0.01$) with FrIII very short linear chains. Slowly digestible (SDS) starch was positively correlated ($r = 0.80$, $p < 0.01$; 0.76, $p < 0.01$) with FrI and FrII, respectively, and negatively correlated ($r = -0.76$, $p < 0.01$) with FrIII. RVA breakdown viscosity was positively correlated ($r = 0.88$, $p < 0.01$) with RDS and negatively correlated ($r = -0.89$, $p < 0.01$) with SDS. Thus, the RVA method potentially could be used as a screening tool for starch digestion properties. This study reveals a molecular basis in amylopectin fine structure variability for starch digestion properties in rice cultivars and could have value in identifying slowly digesting cultivars as well as developing a breeding strategy to produce low glycemic rice cultivars.

KEYWORDS: Rice; starch; RVA; amylopectin; digestibility

INTRODUCTION

The glycemic response of foods, especially starchy foods, has become a topic of interest and research in North America, Asia, and Europe. High glycemic foods typically result in a high initial blood glucose level about 30 min after consumption followed by a rapid drop, usually resulting in a mild hypoglycemic episode (below baseline for blood glucose). The glycemic index (GI) was initially conceived as a tool for dietary management of carbohydrates, particularly for diabetic patients (*1*). The continuous consumption of high GI foods has been implicated in diabetes and prediabetes, cardiovascular disease, and obesity (*2*). Food with a low glycemic response may be beneficial to health. It would be desirable to identify ways to reduce the starch digestion rate so that the glycemic response or index (GI) can be moderated and perhaps additionally to provide extended glucose (energy) release. The GI of rice is known to be relatively high as compared to other starchy foods (*3*), specifically in the normal and waxy rices. The mechanism leading to starch that is low GI and slowly digested is not well-understood. Understanding the factors responsible for this variation would allow breeders to both identify and develop new low GI rice cultivars.

Rice starch, polymers of glucose, is composed of about 15−20% amylose by weight and about 80−85% amylopectin by weight. Amylose is a smaller (although still quite large) linear molecule, and amylopectin is a huge branched molecule often over 1 million glucose units. Generally, starchy foods differ in their digestion rates. This has been attributed to many factors. Generally, it is thought that starches with a higher amylose content have a lower digestion rate. However, to emphasize the complexity of factors influencing digestion properties, Rao (*4*) found that rice varieties with a high amylose content (22%) showed higher starch digestion rates and glycemic responses as compared with rice varieties low in amylose content (15%). Moreover, Panlasigui et al. (*5*) reported a study with three high-amylose rice varieties, IR42, IR36, and IR62 (26.7−27.0%). Samples were cooked under the same conditions and tested for in vitro digestibility as well as blood glucose and insulin responses in healthy human volunteers. Digestion properties differed among the varieties, and these authors concluded that the amylose content alone is not a good predictor of the starch digestion rate. Differences in physicochemical (gelatinization)

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properties among rice varieties with similar high-amylose contents could influence starch digestibility and blood glucose response (*5*). Several studies have demonstrated that differences in physicochemical properties of rice starches are related to starch structure (*6*−*18*). Pertinent to our study, Han and Hamaker (*9*) reported a high negative correlation between amylopectin long linear chains (>DP 100) and viscosity breakdown among rice starches with a fairly narrow range of amylose content.

The biodiversity of rice genotypes is larger than that of other cereal grains (*19*). From a practical point of view, this large biodiversity offers the opportunity to select rice starches with different thermal and pasting properties, amylopectin fine structures, and starch digestibility rates. The purpose of the present study was to develop a knowledge of the factors that determine digestion rate and lower rice GI and to moderate starch digestion rate and lower rice GI. The study focused on the relationship between amylopectin fine structures, its branching characteristics, and starch digestion rate and pasting properties. Our hypothesis, supported by preliminary work, was that amylopectin with a high proportion of long chains may moderate starch digestion properties. Therefore, a range of rice cultivars that may differ in proportion of long chains was chosen based on differences in rapid viscoanalyzer (RVA) paste breakdown [as noted above (*9*)]. Studied were the following: (i) in vitro starch digestion including determination of rapidly digestible (RDS), slowly digestible (SDS), and resistant starch (RS), (ii) amylopectin fine structure analysis, and (iii) starch pasting profiles and thermal properties.

MATERIALS AND METHODS

Rice Samples. Twelve U.S. long and medium grain rice cultivars with a narrow range of amylose contents were grown at the Rice Research and Extension Center, University of Arkansas (Stuttgart, Arkansas). Cultivars M202, BNGL, and RU0301127 were medium grain, and cultivars MBLE, CCDR, CHNR, LGRU, DREW, RU0301041, RU0201139, RU0301081, and RU0201142 were long grain. Rice samples from the 2003 crop year were dried and stored in a conditioning chamber to achieve approximately 13% kernel moisture. Rice grains were milled and debraned according to standard procedures at the Rice Research and Extension Center. All milled rice flours were kept in sealed bags at 4 °C until the time of analysis.

Starch Isolation. The wet milling procedure described by Banks and Greenwood (1975) was used to isolate starches. Flour was deproteinized with a mixture of 0.05 M NaCl and toluene in water (ratio 1:10). Isolated starch was defatted at room temperature for 24 h with 85% methanol and dried overnight at 40 °C.

Total Starch and Amylose Quantification. The amylose quantification kit of Megazyme (Ireland) was used to determine the amylose content of isolated starch. In this procedure, the true amylose content was measured following a separation step to remove amylopectin with concanavalin A. Amylose was then measured following digestion with α-amylase + glucoamylase, and the resulting glucose was measured by the glucose oxidase−peroxidase reaction. The total starch quantification kit of Megazyme (Ireland) was used to determine the starch content of flours from the 12 rice varieties.

Determination of RDS, SDS, and RS. Quantification of RDS, SDS, and RS was performed as described in the method of Englyst et al. (*20*). The rice flour samples were cooked (boiled) for 20 min in water followed by an overnight cooling step at 4 °C. The different starch fractions were measured after incubation of cooked flour pastes (550 mg of flour and 5 mL of water) with an amylase cocktail at 37 °C in capped conical flasks in a shaking water bath. Fifty milligrams of guar gum was added to samples to standardize the viscosity, and glass beads were added to disturb the food particles. A value for RDS was obtained by measuring the amount of glucose released after 20 min of incubation (G20). A second measurement was obtained as the glucose released after a further 100 min of incubation (G120). The starch that remained unhydrolyzed after 120 min of incubation was measured as RS. The SDS fraction was that starch digested between G20 and G120. The glucose released at each time was determined using the glucose oxidase−peroxidase assay kit (Megazyme, Wicklow, Ireland).

Table 1. Total Starch and Amylose Contents of the 12 Rice Cultivars

cultivar	total starch	amylose
RU0201142	79.71 ± 1.1	16.53 ± 0.5
M202	77.02 ± 0.4	15.91 ± 1.4
BNGL	79.21 ± 0.7[a]	16.02 ± 0.7
RU0301127	81.74 ± 1.3	15.98 ± 1.6
MBLE	76.17 ± 0.7	15.79 ± 0.9
DREW	79.04 ± 0.9	16.82 ± 0.6
LGRU	78.37 ± 1.2	16.23 ± 0.7
RU0301041	77.19 ± 0.8	17.01 ± 1.0
RU0301081	75.16 ± 0.8	16.72 ± 0.3
RU0201139	79.04 ± 0.8	17.18 ± 0.8
CHNR	75.84 ± 1.5	16.07 ± 0.7
CCDR	75.33 ± 0.7	15.84 ± 0.9

[a] Values are means ± standard deviations.

Pasting Profile. Starch samples (3 g, on 14% moisture basis) were mixed with water (25 mL) in a canister, heated in a RVA (Newport Scientific Pvt. Ltd., NSW, Australia) at a rate of 5 °C/min to 95 °C, maintained at 95 °C for 7 min, and cooled at a rate of 6 °C/min to 50 °C. Changes in viscosity during heating, cooking, and cooling were recorded, and the gelatinization temperature and peak, breakdown, and setback viscosities were noted. RVA measurements were performed in duplicate.

Amylopectin Structural Analysis. Amylopectin was first isolated and collected chromatographically using a SEC system with Sephacryl S-500 HR media (Amersham Biosciences, Piscataway, NJ), then pooled, and freeze-dried. Amylopectin α-1,6 linkages were hydrolyzed using isoamylase for 24 h, and the α-1,4 linear chains were analyzed using a HPSEC system with Superdex 200 and 30 SEC columns (Amersham Biosciences) with a flow rate of 0.4 mL/min. Aqueous ammonium sulfate (6.5 mM, pH 3) was used as the mobile phase. Typical bimodal profiles were obtained, and proportions of fractions I (long), II (intermediate and short), and III (very short) linear chains were determined by peak integration software (Star Chromatography Workstation, version 4.51, Varian Associates, Sugarland, TX).

Statistics. Variations among the 12 rice starch cultivars were examined by SPSS 12.0 for Windows software using Pearson's correlation.

RESULTS AND DISCUSSION

Amylopectin Fine Structure. Table 1 shows total starch and amylose contents of the 12 U.S. long grain rice cultivars. Amylose contents were verified to be similar among the rice cultivars (this was a criteria for their selection). Isolated and debranched amylopectins of the 12 rice cultivars were fractionated into three molecular weight groups denoted by FrI (DP > 33), FrII (13 < DP < 33), and FrIII (DP < 13) representing proportions of long, intermediate/short, and very short linear glucan chains, respectively (**Figure 1**). Chromatograms of cultivars CCDR and RU0201142 showed a bimodal linear chain molecular weight distribution. The percentage areas of fractions among the 12 cultivars are shown in **Table 2**. **Figure 1** shows differences between the two distribution profiles for cultivars CCDR and RU0201142, particularly in the proportions of intermediate and short linear chains. Overall, CCDR and CHNR had the highest proportion of both long (FrI) and intermediate/short chains (FrII) and lowest proportion of very short chains (FrIII) (**Table 2**). In contrast, BGNL, RU0301127, M202, and RU0201142 linear chain distributions showed a higher proportion of very short chains and a lower proportion of long and intermediate/short chains. This variability among rice cultivars

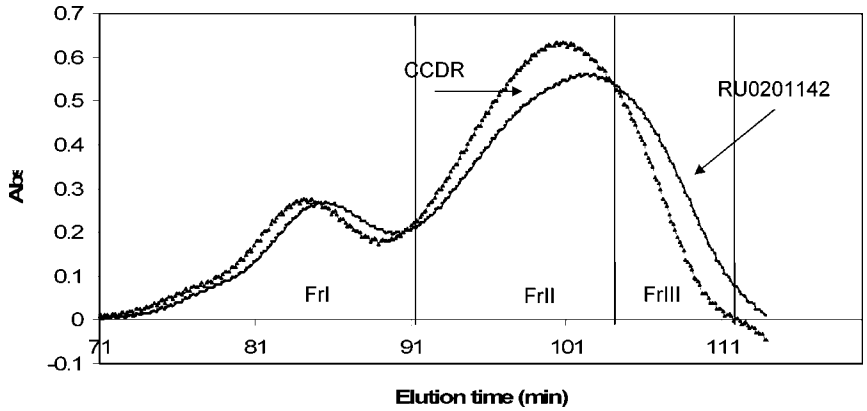

Figure 1. Chromatograms of isolated and debranched amylopectin from CCDR and RU0201142 rice cultivars.

Table 2. Percentage Area of FrI, FrII, and FrIII in Chromatograms of Debranched Amylopectin from Starch of 12 Rice Cultivars

cultivar	FrI	FrII	FrIII
RU0201142	24.3 ± 1.6	40.7 ± 1.1	35 ± 1.3
M202	24.7 ± 1.9	40.9 ± 1.7	34.3 ± 2.1
BNGL	25.9 ± 0.7[a]	41.6 ± 0.6	33.5 ± 1.0
RU0301127	26.5 ± 1.7	42 ± 1.5	31.6 ± 1.2
MBLE	26.7 ± 1.9	45.3 ± 2.1	28.5 ± 1.4
LGRU	26.0 ± 1.7	41.3 ± 1.3	25.9 ± 1.3
DREW	27.8 ± 1.3	45.2 ± 1.6	27 ± 1.5
RU0301041	28 ± 1.2	48 ± 1.7	24.1 ± 1.0
RU0301081	29.5 ± 2.8	47.2 ± 1.8	23.4 ± 2.1
RU0201139	28.2 ± 1.7	47.8 ± 1.9	24.2 ± 1.5
CHNR	30.4 ± 0.6	48 ± 0.6	21.6 ± 0.4
CCDR	30.7 ± 0.8	48.8 ± 1.2	21.5 ± 0.9

[a] Values are means ± standard deviations.

Table 3. In Vitro Starch Digestibility Values (% of Total Starch) of 12 Rice Cultivars[a]

cultivar	RDS	SDS	RS
RU0201142	77.39 ± 0.3	13.7 ± 2.51	8.88 ± 1.31
M202	77.84 ± 1.35	13 ± 1.53	9.17 ± 1.38
BNGL	75.97 ± 0.6[b]	16.1 ± 1.21	7.91 ± 1.1
RU0301127	74.75 ± 2	18.6 ± 0.88	6.67 ± 1.3
MBLE	70.45 ± 2.31	23.7 ± 2.09	5.86 ± 2.21
LGRU	71.39 ± 2.41	20.1 ± 1.61	8.48 ± 1.79
DREW	76.25 ± 1.5	17 ± 0.8	6.77 ± 1.1
RU0301041	71.89 ± 2.74	20 ± 1.45	8.14 ± 1.98
RU0301081	73.31 ± 2.11	17.3 ± 1.45	9.42 ± 1.78
RU0201139	72.87 ± 2.56	19.7 ± 2.35	7.46 ± 2.42
CHNR	65.35 ± 2.11	25.5 ± 1.69	10.14 ± 1.75
CCDR	64.34 ± 2.56	25.4 ± 1.53	11.28 ± 1.88

[a] RDS is rapidly digestible starch, SDS is slowly digestible starch, and RS is resistant starch. [b] Values are means ± standard deviations.

Table 4. Correlation between Debranched Amylopectin Chain Fractions and RDS, SDS, and RS

fraction	RDS	SDS	RS
FrI	−0.87[a]	0.79[a]	NS
FrII	−0.81[a]	0.77[a]	NS
FrIII	0.79[a]	−0.76[a]	NS

[a] Correlation is significant at the 0.01 level. NS, not significant.

(medium and long grain) emphasizes the role of variants in starch-synthesizing enzymes on amylopectin molecular structure.

Differences in amylopectin fine structure reflect up/down regulation of starch enzyme expression and activity (*21*). This regulation is under genetic and environmental control. Jiang et al. (*22*) reported that high temperature decreases activity of starch branching enzyme and reduces its branching activity, thus altering amylopectin fine structure. For instance, increased temperature resulted in a reduction of amylopectin short chains (*16*). In the present study, the 12 rice cultivars were grown in the same location and under the same environmental conditions. Thus, the variability among cultivars shown in **Table 2** indicates genetic differences in rice cultivars that affect amylopectin fine structure, specifically in the length of the branched linear chains and their relative proportions.

Amylopectin Fine Structure and Starch Digestion Properties. On the basis of the study of Han and Hamaker (*9*) that showed high correlation between RVA breakdown viscosity and proportion of amylopectin long chains, RVA breakdown viscosity was used to select rice cultivars that differed in amylopectin fine structure. This correlative relationship was verified in the present study as discussed in the next section below.

Starch digestibility data are presented in **Table 3**, and correlations between digestibility values (RDS and SDS) and amylopectin fine structure parameters are found in **Table 4**. Results show that RDS is lowest in cultivars with the highest proportion of long (FrI) and intermediate/short (FrII) chains and with the lowest proportion of very short chains (FrIII) (**Tables 2** and **3**). Accordingly, cultivars with the highest SDS similarly showed the highest proportion of FrI and FrII and the lowest proportion of FrIII. Correlation analysis showed that RDS was negatively correlated ($r = -0.87$, $p < 0.01$; $r = -0.81$, $p < 0.01$) with FrI and FrII, respectively, and positively correlated ($r = 0.79$; $p < 0.01$) with FrIII (**Table 4**). SDS was positively correlated ($r = 0.79$, $p < 0.01$; $r = 0.77$, $p < 0.01$) with FrI and FrII, respectively, and negatively correlated ($r = -0.76$, $p < 0.01$) with FrIII. These correlations are for the mixture of long and medium grain rice cultivars. Correlation analysis run only on long grain rice samples showed equally high coefficients. RDS was negatively correlated ($r = -0.72$, $p < 0.05$; $r = -0.71$, $p < 0.05$) with FrI and FrII and positively correlated ($r = 0.73$, $p < 0.05$) with FrIII (**Table 5**). In contrast, SDS was negatively correlated ($r = -0.68$, $p < 0.05$) with FrIII and positively correlated ($r = 0.69$, $p < 0.05$; $r = 0.7$, $p < 0.05$) with FrI and FrII (**Table 5**). RDS has been shown in humans to correlate positively to GI (*20*), and high SDS may be beneficial in terms of extended glucose, or energy, release. Therefore, rice cultivars with high FrI and FrII and low FrIII should have lower GIs and delayed glucose release.

In the present study, RS contents ranged from 5.86 to 11.28%, reflecting the storage of the cooked pastes overnight at 4 °C. When immediately cooked and analyzed, Englyst et al. (*20*) found that cooked long grain rice did not have RS. Because of

Table 5. Correlation between Debranched Amylopectin Chain Fractions and RDS, SDS, and RS among Only Long Grain Cultivars

fraction	RDS	SDS	RS
FrI	–0.72[a]	0.69[a]	NS
FrII	–0.71[a]	0.70[a]	NS
FrIII	0.73[a]	–0.68[a]	NS

[a] Correlation is significant at the 0.05 level. NS, not significant.

the overnight cooling of cooked rice samples, amylopectin retrogradation was likely the parameter affecting RDS and SDS values, accounting for the high correlations observed.

The study of Kohyama et al. (*23*) on wheat starch showed that retrogradation was more advanced when the amylopectin polymer has more long chains than short chains. This supports our digestibility and amylopectin fine structure results. On the other hand, a shortening of amylopectin chains, performed using β-amylase, caused no retrogradation to occur when the external chains were less than 11 glucose units in length (*21*). This shortening inhibited the amylopectin retrogradation process. In the current study, rice cultivars with higher proportion very short chains (DP < 13) had lower SDS. We speculate that this is likely due to its lesser tendency to form double helices and crystallites during retrogradation. On the other hand, rice cultivars with a higher proportion of longer linear chains may have the ability to form more readily double helices due to more free movement in the amylopectin polymer for A chains to interact, a theory proposed by Klucinec and Thompson (*24*). To reiterate, starches from rice cultivars with high FrI and FrII and low FrIII had reduced RDS and higher SDS contents.

Prediction of Rice Starch Digestibility Properties. There is currently a perceived need to develop starchy foods with low GI and perhaps extended release of glucose in the human small intestine (SDS) with physicochemical characteristics desired in the food industry. The present study demonstrates that such development of low GI starches with corresponding elevated levels of SDS could be based on studies on the modification of amylopectin fine structure.

For purposes of breeding, or other genetic manipulation of starch structure, a rapid method to screen for the low GI/high SDS trait is needed. In this regard, we thought that the RVA breakdown viscosity measurement might be an appropriate tool for a fairly rapid assessment of digestibility, through its correlative relationship with amylopectin fine structure parameters (*9*). The RVA viscosity parameters of the 12 rice cultivars starches are presented in **Table 6**. Breakdown was highest in BGNL and RU0301127 and lowest in CCDR and CHNR. In addition, BGNL and RU0301127 had lower pasting temperatures than CCDR and CHNR (**Table 3**). RVA breakdown describes the viscosity behavior and the fragility of swollen starch granules. With increasing heat treatment, granular structure absorbs water and starts to swell. During the swelling, amylose and some amylopectin leaches out. Swollen granules, as well as the leached soluble components, cause a rise in viscosity (*25*). With increasing heat, swollen granules become more fragile and may break resulting in a decrease in viscosity (*26*). The difference between the highest level of viscosity of swollen and gelatinized starch granules and their disintegration is recognized as paste breakdown.

Rice cultivars BNGL, RU0301127, M202, and RU0201142 showed the highest breakdown and lowest FrI and FrII fractions (**Tables 3** and **7**). The lowest RVA paste breakdown was shown for CHNR and CCDR cultivars with the highest proportion of

Table 6. Rapid Visco Analyzer Pasting Characteristics of Starch from 12 Rice Cultivars[a]

	CP				
cultivar	breakdown	setback	final visc	peak time	pasting temp (°C)
RU0201142	1371	1484	3461	5.933	75.1
M202	1349	1090	2717	5.7996	75.9
BNGL	1220	1002	2752	5.9996	72.65
RU0301127	1114	1124	2809	6.0663	72.55
MBLE	942	1490	2819	5.7996	78.15
LGRU	949	1418	3109	5.8663	76.7
DREW	945	1602	3081	5.7996	78.1
RU0301041	883	1882	3344	5.999	81.45
RU0301081	895	1624	3389	5.999	77.4
RU0201139	910	1757	3600	5.933	83.85
CHNR	652	1134	1979	5.533	78.25
CCDR	671	1274	2360	5.4663	80.8

[a] Values are means ± standard deviations.

Table 7. Correlations between RVA Breakdown and Pasting Temperature and Debranched Amylopectin Linear Chains Fractions

	FrI	FrII	FrIII
RVA breakdown	–0.92[a]	–0.87[a]	+0.96[a]
pasting temp (° C)	+0.67[b]	+0.83[a]	–0.80[a]

[a] Correlation is significant at the 0.01 level. [b] Correlation is significant at the 0.05 level.

Table 8. Correlation RVA Breakdown and Starch Digestibility Rate

	RDS	SDS	RS
RVA breakdown	+0.88[a]	–0.89[a]	NS
pasting temp (° C)	–0.64[b]	+0.61[b]	NS

[a] Correlation is significant at the 0.01 level. [b] Correlation is significant at the 0.05 level. NS, not significant.

FrI and FrII (**Tables 3** and **7**). Paste breakdown in the present study was highly and negatively correlated ($r = -0.92$, $p < 0.01$; $r = -0.86$, $p < 0.01$) with FrI and FrII, respectively (**Table 7**). FrIII was positively correlated ($r = 0.96$, $p < 0.01$) with breakdown viscosity. Cultivars with low breakdown had a high pasting temperature and proportion of long and intermediate/short chains. Han and Hamaker (*9*) speculated that a higher proportion of long chains (FrI) may help to maintain the gelatinized starch granule structure. **Table 8** shows high correlation between RVA breakdown and starch digestibility. RVA breakdown was highly and positively correlated (+0.88, $p < 0.01$) with RDS and negatively (−0.89, $p < 0.01$) with SDS. These correlations demonstrate the potential of using RVA breakdown as a screening tool for selection of rice cultivars with low RDS and high SDS.

In conclusion, the present study shows that variability in rice starch amylopectin fine structure can significantly affect its in vitro cooked starch digestion properties. This finding could have significant importance for the U.S. rice industry.

It not only shows that U.S. rice cultivars can differ in starch digestion properties to likely result in low glycemic response, but also demonstrates that an easy physical test (RVA paste breakdown) can be used as a rice breeding selection tool for a lower GI/slowly digestible property. Therefore, we show the possibility that lines can be selected with high proportion of amylopectin long and intermediate/short, and low proportion of very short, linear chains for comparably low RDS and high SDS contents.

LITERATURE CITED

(1) Jenkins, D. J.; Wolever, T. M.; Taylor, R. H.; Barker, H.; Fielden, H.; Baldwin, J. M.; Bowling, A. C.; Newman, H. C.; Jenkins, A. L.; Goff, D. V. Glycemic index of foods: A physiological basis for carbohydrate exchange. *Am. J. Clin. Nutr.* **1981**, *34*, 362−366.

(2) Ludwig, D. S. The glycemic index: Physiological mechanism relating to obesity. Diabetes and cardiovascular disease. *J. Am. Med. Assoc.* **2002**, *287*, 2414−2423.

(3) Miller, J. B.; Pang, E.; Bramall, L. Rice: A high or low glycemic index food?. *Am. J. Clin. Nutr.* **1992**, *56*, 1034−1036.

(4) Rao, P. S. Studies on the nature of carbohydrate moiety in high yielding varieties of rice. *J. Nutr.* **1971**, *101*, 879−884.

(5) Panlasigui, L. N.; Thompson, L. U.; Juliano, B. O.; Perez, C. M.; Yiu, S. H.; Greenberg, G. R. Rice varieties with similar amylose content differ in starch digestibility and glycemic response in humans. *Am. J. Clin. Nutr.* **1991**, *54*, 871−877.

(6) Lu, S.; Chen, L. N.; Lii, C. Y. Correlations between the fine structure, physicochemical properties, and retrogradation of amylopectins from Taiwan rice varieties. *Cereal Chem.* **1997**, *74*, 34−39.

(7) Lai, V. M. F.; Lu, S.; Lii, C. Y. Molecular characteristics influencing retrogradation kinetics of rice amylopectins. *Cereal Chem.* **2000**, *77*, 272−278.

(8) Lai, V. M. F.; Shen, M. C.; Yeh, A. I.; Juliano, B. O.; Lii, C. Y. Molecular and gelatinization properties of rice starches from IR24 and Sinandomeng cultivars. *Cereal Chem.* **2001**, *78*, 596−602.

(9) Han, X. Z.; Hamaker, B. R. Amylopectin fine structure and rice starch paste breakdown. *J. Cereal Sci.* **2001**, *34*, 279−284.

(10) Lin, Y. S.; Yeh, A. I.; Lii, C. Y. Correlation between starch retrogradation and water mobility by differential scanning calorimetry (DSC) and nuclear magnetic resonance (NMR). *Cereal Chem*, **2001**, *78*, 647−653.

(11) Fujita, N.; Kubo, A.; Suh, D. S.; Wong, K. S.; Jane, J. L.; Ozawa, K.; Takaiwa, F.; Inaba, Y.; Nakamura, Y. Antisense inhibition of isoamylase alters the structure of amylopectin and the physicochemical properties of starch in rice endosperm. *Plant Cell Physiol.* **2003**, *44*, 607−618.

(12) Nakamura, Y.; Sakurai, A.; Inaba, Y.; Kimura, K.; Iwasawa, N.; Nagamine, T. The fine structure of amylopectin in endosperm from Asian cultivated rice can be largely classified into two classes. *Starch/Stärke* **2002**, *54*, 117−131.

(13) Patindol, J.; Wang, Y. J. Fine structures of starches from long-grain cultivars with different functionality. *Cereal Chem.* **2002**, *79*, 465−469.

(14) Wang, Y. J.; Wang, L. Structures of four waxy rice starches in relations to thermal, pasting, and textural properties. *Cereal Chem.* **2002**, *79*, 252−256.

(15) Yao, Y.; Zhang, J.; Ding, X. Partial β-amylolysis retards starch retrogradation in rice products. *J. Agric. Food Chem.* **2003**, *51*, 4066−4071.

(16) Noda, T.; Nishiba, Y.; Sato, T.; Suda, I. Properties of starches from several low-amylose rice cultivars. *Cereal Chem.* **2003**, *80*, 193−197.

(17) Qi, X.; Tester, R. F.; Snape, C. E.; Ansell, R. Molecular basis of the gelatinisation and swelling characteristics of waxy rice starches grown in the same location during the same season. *J. Cereal Sci.* **2003**, *37*, 363−376.

(18) Vandeputte, G. E.; Derycke, V.; Geeroms, J.; Delcour, J. A. Rice starches. II. Structural aspects provide insight into swelling and pasting properties. *J. Cereal Sci.* **2003**, *38*, 53−59.

(19) Ziu, J.; Gale, M. D.; Quarrie, S.; Jackson, M. T.; Bryan, G. J. AFLP markers for the study of rice biodiversity. *Theor. Appl. Genet.* **1998**, *96*, 602−611.

(20) Englyst, H. N.; Veenstra, J.; Hudson, G. J. Measurement of rapidly available glucose in plant foods, a potential in vitro predictor of the glycaemic response. *Br. J. Nutr.* **1996**, *75*, 327−337.

(21) Würsch, P.; Gumy, D. Inhibition of amylopectin retrogradation by partial beta-amylosis. *Carbohydr. Res.* **1994**, *256*, 129−137.

(22) Jiang, H.; Dian, W.; Wu, P. Effect of high temperature on fine structure of amylopectin in rice endosperm by reducing the activity of the starch branching enzyme. *Phytochemistry* **2003**, *63*, 53−59.

(23) Kohyama, K.; Matsuki, J.; Yasui, T.; Sasaki, T. A. Differential thermal analysis of gelatinization and retrogradation of wheat starches with different amylopectin chain lengths. *Carbohydr. Polym.* **2004**, *58*, 71−77.

(24) Klucinec, J.; Thompson, D. Amylose and amylopectin interact in retrogradation of dispersed high-amylose starches. *Cereal Chem.* **1999**, *76*, 282−291.

(25) Mohan, B. H.; Gopal, A.; Malleshi, N. G.; Tharanathan, R. N. Characteristics of native and enzymatically hydrolyzed ragi (*Eleusine coracana*) and rice (*Oryza sativa*) starches. *Carbohydr. Polym.* **2005**, *59*, 43−50.

(26) Whistler, L.; BeMiller, J. N. *Carbohydrate Chemistry for Food Scientists*; American Association of Cereal Chemists: St. Paul, MN, 1997; pp 91−115.

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