

Studies on the in vitro starch digestibility and the glycemic index of six different indigenous rice cultivars from the Philippines

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Abstract

Starch degradability was studied in six indigenous Philippine rice cultivars differing in amylose contents. An in vitro enzymatic starch digestion method was applied in order to estimate the expected glycemic index in vivo based on the kinetics of starch hydrolysis in vitro. Two different treatments were investigated: first, samples were cooked and analysed immediately; second, samples were cooked and stored in a refrigerator for 24 h at a temperature of 4 °C in order to induce a retrogradation effect. The results indicate substantial differences in the estimated glycemic index between rice cultivars. Values ranged between 68 and 109 for cooked rice and between 64 and 87 for stored rice containing retrograded starch. Starch hydrolysis tended to be more rapid and more complete for waxy cultivars than for high amylose cultivars. Storing rice in the refrigerator led to a reduction of the estimated glycemic index for all cultivars. The highest decrease in starch hydrolysis after cool storing was seen for the waxy cultivars.

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1. Introduction

Starch digestibility is known to vary among different starchy foods and has increasingly attracted interest in the context of non-insulin-dependent diabetes treatment (Jenkins et al., 1988). Numerous studies have been carried out to assess blood glucose response after the intake of different plant sources of carbohydrates (e.g. Behall, Scholfield, Yuhaniak, & Cabary, 1989; Jenkins et al., 1982; Jenkins, Wolever, Jenkins, Josse, & Wong, 1984). Blood glucose response is commonly estimated using the glycemic index (GI), which relates the response of a test food to that of a reference food, usually fresh white bread. The GI is usually obtained by dividing the incremental postprandial blood glucose production by the corresponding production after ingestion of an equicarbohydrate portion of the reference food (Jenkins et al., 1983). A low glycemic response is considered beneficial from a nutritional point of view, especially for individuals suffering from

impaired glucose tolerance. This has led to the dietary recommendation of legumes (Jenkins et al., 1982, 1983) due to their low carbohydrate level and slow rate of enzymatic starch digestion. Wolever and Mehling (2002) recently demonstrated the beneficial effect of high-carbohydrate-low-glycemic index diets on insulin secretion in supporting β -cell function. Differences in the amounts of carbohydrate ingested from the same source, however, do not seem to influence the glycemic index, as was shown by Rasmussen and Gregersen (1992) in the case of parboiled white rice.

Aside from the glycemic index, resistant starch (RS) content has been established as an important measure to characterize starch digestibility. Resistant starch acts as a fermentation substrate in the colon, similar to non-starch carbohydrates, with positive implications for the prevention of food-borne diseases, such as colon cancer and hypolipidemia. Moreover, it is likely to be negatively correlated with the glycemic index (Annison & Topping, 1994; Asp, van Amelsvoort, & Hautvast, 1996).

Differences in starch digestibility have been ascribed to various factors, including the botanical source (Goñi, Garcia-Alonso, & Saura-Calixto, 1997; Jenkins et al.,

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1984), food processing (Bravo, Siddhuraju, & Saura-Calixto, 1998; Jenkins et al., 1982; Sagum & Arcot, 2000), physiochemical properties (particularly gelatinization characteristics) (Panlasigui, Thomson, Juliano, Perez, Yiu, & Greenberg, 1991), particle size (Snow & O'Dea, 1981), amylose/amylopectin ratio (Goddard, Young, & Marcus, 1984; Juliano & Goddard, 1986) and the presence of lipid-amylose complexes (Goddard et al., 1984; Guraya, Kadan, & Champagne, 1997).

Rice, being one of the primary dietary sources of carbohydrates worldwide, is of particular interest when assessing variability in starch digestibility. The glycemic response of rice is known to be relatively high compared to other starchy foods. Jenkins et al. (1984) report a glycemic index of 96 for brown rice and 83 for white rice. Miller, Pang, and Bramall (1992) likewise classified rice as a high glycemic index food with values ranging from 64 to 93 in the freshly cooked form. The cooling and storing of cooked rice is known to entail starch retrogradation, thus increasing the level of enzyme-resistant starch through recrystallization (Englyst, Kingman, & Cummings, 1992; Sievert & Pomeranz, 1989). The term retrogradation refers to changes that occur in gelatinized starch upon cooling, which imply fully reversible recrystallization in the case of amylopectin and partially irreversible recrystallization in the case of amylose (Björck, 1996). Baik, Kim, Cheon, Ha, and Kim (1997) found a higher degree of recrystallization in waxy rice starch (consisting exclusively of amylopectin) than in non-waxy rice starch (consisting of amylose and amylopectin). Experimental data on the starch digestibility of retrograded rice starch are not available.

Various authors have suggested in vitro starch hydrolysis methods for predicting in vivo glycemic response (e.g. Englyst, Veenstra, & Hudson, 1996; O'Dea, Snow, & Nestel, 1981). The objective of this study was to assess the variability of in vitro digestibility of morphologically and chemically different indigenous rice cultivars from the Philippines and to estimate their glycemic index based on a model established previously by Goñi et al. (1997). Rice cultivars varying widely in amylose content (between 0 and 26.9%) were chosen in order to estimate the glycemic index. In rural areas of the Philippines, rice is commonly pre-cooked 1 day in advance to save labour and firewood, with subsequent cool storing for up to one day. The impact of this practice on starch digestibility was estimated by analysing rice samples in the retrograded form.

2. Materials and methods

2.1. Samples

Rice samples were collected during a research excursion to the Philippines in the Province of Aklan in the

year 2000. All six analysed cultivars are indigenous upland rice cultivars grown in subsistence oriented farming systems. They were selected from a large set of samples consisting of 51 cultivars. The selection criteria were amylose content and popularity among the rice-growing population. The local names of the cultivars are *Milagrosa*, *Manumbaeay*, *Kutsiyam*, *Kinaures*, *Bagoean*, and *Karaya*. The samples were collected directly from the fields or taken from farmer's storages and dried at a temperature of 40 °C to attain a moisture content of approximately 10%. Rough rice was de-hulled, using a mechanical rubber-roll de-huller (Satake, Japan). All analyses were carried out on brown rice, since this is the form in which rice is consumed in the sample collection area.

2.2. Processing methods

Approximately 50 mg of sample material, which corresponds to 2–4 entire rice grains, depending on the variety, were weighed into a 30-ml Erlenmeyer flask. Subsequently, the samples were cooked at 180 °C for 30 min in 4 ml of distilled water, using a thermostat-controlled heater. For the first experiment, the in vitro starch digestion procedure was started immediately after cooking, in order to avoid any effect of retrogradation. For the second set of experiments, the samples were stored in the refrigerator, at a temperature of 4 °C for 24 h after cooking, in order to allow for a retrogradation effect. 4 °C is the temperature at which the degree of recrystallization is expected to be the highest in rice starch (Baik et al., 1997). The enzymatic hydrolysis was performed on the following day in the same flasks as the cooking, in order to avoid any losses of sample material.

2.3. Analytical procedures

2.3.1. Total starch (TS)

This was determined enzymatically according to the modified Goñi et al. (1997) method. Raw rice samples were ground to pass through a 0.5-mm sieve. Subsequently, 25–35 mg of ground sample material were dispersed in 6 ml of 2 M KOH and shaken vigorously for 30 min at room temperature. Solubilized starch was then hydrolyzed by adding 60 µl of amyloglucosidase from *Aspergillus niger* (ref. 102 857, Roche; Basel, Switzerland), and by incubating the samples at 60 °C for 45 min in a shaking waterbath. After centrifugation (10 min, 3000 g), glucose concentration in the supernatant was determined by using a glucose oxidase-peroxidase kit (ref. 510-A, Sigma; Taufkirchen, Germany). Colour absorption was measured at a wavelength of 450 nm and glucose concentration was converted into starch content by applying the factor 0.9. Each cultivar was analysed in duplicate.

2.3.2. Resistant starch (RS)

This was likewise determined according to Goñi et al. (1997). Around 100 mg of ground sample material were first incubated with a pepsin solution containing 20 mg of pepsin (ref. 7180, Merck; Darmstadt, Germany) for 60 min at 40 °C for protein removal. Then starch was hydrolysed at 37 °C for 16 h after adding an enzyme solution containing 40 mg of α -amylase (ref. A-3176, Sigma). After α -amylase hydrolysis, samples were centrifuged and the supernatants were discarded. The residue was analysed for remaining starch as described earlier, using amylogucosidase as hydrolysing enzyme.

2.3.3. Digestible starch (DS)

This was calculated as the difference between TS and RS.

2.3.4. Free glucose (FG) content

This was determined according to Bravo et al. (1998) in order to correct the TS values obtained as described earlier. 100 mg of sample material were dispersed in 2 M KOH and treated with invertase (ref. 104914, Boehringer Mannheim; Mannheim, Germany) for 30 min at 37 °C. After centrifugation, a 1 ml aliquot was precipitated with 2 ml of 96% ethanol. The whole sample was centrifuged again and glucose was analysed from the supernatant using the Sigma kit mentioned previously.

2.3.5. Amylose content

This was determined in accordance with the method established by Mestres, Matencio, Pons, Yajid, and Fliedel (1996), by carrying out differential scanning calorimetry (DSC). This method is based on the enthalpy change which occurs during the exothermic formation of complexes between amylose and phospholipids during cooling. According to Mestres et al. (1996) the amylose contents obtained from differential scanning calorimetry are comparable with those obtained from the classical colorimetric method, based on complex formation between amylose and iodine ('Blue Value'). Differences between the methodologies were continuously found to be less than 1% in that previous study, with the DSC method being time-saving and more easily repeatable.

Between 10 and 11 mg of ground sample material were weighed into a medium pressure inox pan (70 μ l) and 50 ml of a 2% L- α -lysophosphatidylcholine (LPC) solution from egg yolk (ref. L-4129, Sigma) were added. The pan was then hermetically sealed and differential scanning calorimetry measurements were performed using a Perkin-Elmer DSC 7 instrument (Perkin-Elmer, Norwalk, USA). A sample pan was placed in the sample cell at 35 °C, whereas an inox pan containing 50 ml of water was placed in the reference cell. Temperature was first raised to 160 °C at 10 °C/min and then held constant for 2 min. Subsequently, the samples were cooled

at 10 °C/min down to a temperature of 60 °C. Energy data were automatically collected during the cycle. The exotherm of the LPC-amylose complex formation, occurring during cooling at a temperature of 80–85 °C was used to determine the amylose content. A base line correction was made by subtracting the energy data obtained from a cycle with two reference pans. For calculation of the amylose content, the area under the exothermic peak of each sample was divided by the corresponding area of a reference sample containing pure potato amylose. Each cultivar was analysed in duplicate.

2.3.6. In vitro kinetics of starch digestion

These were analysed following the procedure suggested by Goñi et al. (1997). Fifty-milligram rice portions were prepared in 30-ml Erlenmeyer flasks as explained earlier. Subsequently, 10 ml of HCl-KCl buffer pH 1.5 were added and samples were homogenized for 2 min using an Ultra Turrax homogenizer (T25, Ika Labortechnik). Then, 0.2 ml of a solution containing 1 mg of pepsin from porcine gastrin mucosa (ref. 107195, Merck) in 10 ml HCl-KCl buffer, pH 1.5, were added to each sample, followed by 60 min of incubation in a shaking water bath at 40 °C. The volume was raised to 25 ml by adding 15 ml of Tris-Maleate buffer (pH 6.9) and the pH was adjusted accurately. To start starch hydrolysis, another 5 ml of Tris-Maleate buffer containing 2.6 IU of α -amylase from porcine pancreas (ref. A-3176, Sigma) were added to each sample. The flasks were placed in a shaking waterbath at 37 °C with moderate agitation. Aliquots (0.1 ml) were taken from each flask every 30 min from 0 to 3 h. α -Amylase was inactivated immediately by placing the tubes containing the aliquots in a boiling waterbath for 5 min. Then, 1 ml of 0.4 M sodium-acetate buffer, pH=4.75, and 30 μ l of amylogucosidase from *Aspergillus niger* (ref. 102 857, Roche) were added. In order to hydrolyse digested starch into glucose, samples were incubated at 60 °C for 45 min. Finally, glucose concentration was measured using the glucose oxidase-peroxidase kit (ref. 510-A, Sigma). The rate of starch digestion was expressed as a percentage of the total starch hydrolysed at different times (30, 60, 90, 120, 150, and 180 min). Each cultivar and each treatment was analysed in triplicate.

A non-linear model established by Goñi et al. (1997) was applied to describe the kinetics of starch hydrolysis. The first order equation has the form:

$$C = C_{\infty}(1 - e^{-kt})$$

where C corresponds to the percentage of starch hydrolyzed at time t , C_{∞} is the equilibrium percentage of starch hydrolyzed after 180 min, k is the kinetic constant and t is the time (min). The parameters C_{∞} and k were estimated for each cultivar and each treatment

based on the data obtained from the *in vitro* hydrolysis procedure. Parameter estimation was carried out using the software SYSTAT version 9 for MS Office.

The area under the hydrolysis curve (AUC) was calculated using the equation

$$\text{AUC} = C_{\infty}(t_f - t_0) - (C_{\infty}/k)[1 - \exp[-k(t_f - t_0)]]$$

where C_{∞} corresponds to the equilibrium percentage of starch hydrolysed after 180 min, t_f is the final time (180 min), t_0 is the initial time (0 min) and k is the kinetic constant.

A hydrolysis index (HI) was obtained by dividing the area under the hydrolysis curve of each sample by the corresponding area of a reference sample (fresh white bread). Goñi et al. (1997), showed this hydrolysis index to be a good predictor of glycemic response. Expected GI was thus estimated using the model

$$\text{GI} = 39.71 + (0.549 \times \text{HI}).$$

2.4. Statistics

A one way analysis of variance (ANOVA) was performed to test the statistical significance of differences in the hydrolysis index and estimated glycemic index between cultivars and the retrogradation effect. ANOVA was followed by Duncan's multiple range test. The significance level was $P < 0.05$ and the software used was Statistica for Windows 97, version 5.1 (Statsoft inc., Tulsa, USA).

3. Results

Total starch and amylose contents are given in Table 1. TS values ranged between 72.0% and 82.0% on a dry matter basis, which is in agreement with literature data (Juliano, 1985). The resistant starch (RS) level was generally low, except for the cultivar Kutsiyam, which stands out with a relatively higher RS content (1.3%).

Table 1
Total starch, resistant starch, digestible starch, free glucose, and amylose contents

Cultivar	TS (%DM)	RS (%DM)	DS (%DM)	FG (%DM)	Amylose	
					%DM	%TS
Milagrosa	72.0	0.2	71.8	0.16	26.9	37.3
Manumbaeay	75.1	0.3	74.8	0.24	26.9	35.8
Kutsiyam	82.0	1.3	80.7	Traces	18.7	22.8
Kinaures	76.0	0.3	75.7	0.13	9.8	12.9
Bagoean	80.2	0.2	80.0	0.34	0	0
Karaya	81.0	0.1	80.9	0.30	0	0

TS, total starch; RS, resistant starch; DS, digestible starch; FG, free glucose, including that from sucrose; DM, dry matter.

As a result of the low RS values, digestible starch (DS) content was high in all cultivars. Free glucose (FG) occurred only in very small quantities: the values ranged from traces to 0.34%. The highest free glucose contents were found for the two waxy cultivars.

Two of the selected cultivars—Milagrosa and Manumbaeay—had high amylose contents, both 26.9%. The cultivar Kutsiyam had an intermediate amylose content (18.7%) and the cultivar Kinaures a low amylose content (9.8%). The remaining two cultivars—Bagoean and Karaya—were waxy, with starch consisting exclusively of amylopectin.

The results of the *in vitro* starch digestion are summarized in Table 2, including the estimated parameters C_{∞} and k in the starch hydrolysis model, the hydrolysis index HI and the estimated glycemic index GI. The starch hydrolysis curves for cooked and retrograded samples are shown in Fig. 1.

Table 2 indicates substantial differences between rice cultivars in terms of starch digestibility and glycemic response. Estimated glycemic indices of freshly cooked rice samples ranged between 109 for the waxy cultivar Karaya and 68.0 for the high amylose cultivar, Milagrosa. Amylose content had an obvious impact on starch degradation and thus on the predicted glycemic response. GI was the highest for the waxy cultivar, Karaya (GI 109.2), followed by the low amylose (9.8%) cultivar, Kinaures (GI 96.9), and another waxy cultivar, namely Bagoean (GI 92.3). However, amylose content was not the only factor determining starch digestibility as reflected in the statistical differences occurring between the waxy cultivars. The lowest GI was estimated for the high amylose (26.9%) cultivar, Milagrosa (GI 68.0), followed by the intermediate (18.7%) amylose cultivar, Kutsiyam and the high amylose (26.9%) rice, Manumbaeay. The low GI of the cultivar Kutsiyam is in line with its relatively high resistant starch content (1.3%). As with the waxy varieties, a statistically significant difference occurred between the two cultivars, Milagrosa and Manumbaeay, which are equal in amylose content on a dry matter basis.

Storing the cooked samples at 4 °C for 24 h led to a reduction of HI and estimated GI for all varieties. The range of retrograded rice, GI, was between 63.5 and 82.6. The reduction of HI and GI after storage was statistically significant for all samples except for the high amylose cultivar Milagrosa, and the intermediate (18.7%) amylose cultivar, Kutsiyam. The highest retrogradation effect was observed for the waxy cultivars: GI decreased from 109.2 to 82.6 for Karaya and dropped from 92.3 to 65.7 for Bagoean, which corresponds to a reduction by 26.6 in both cases. The two high amylose cultivars behaved differently; while retrogradation effect was rather weak and not statistically significant for Milagrosa, the predicted GI dropped substantially from 87.3 to 74.2 for Manumbaeay. The intermediate amylose

Table 2
Model parameters, hydrolysis index and estimated glycemic index of cooked and retrograded rice samples

Cultivar	Amylose content (%DM)	Treatment	C_{∞}	k	Calculated HI	Estimated GI
Milagroa	26.9	Cooked	44.3±6.0	0.022	51.4±6.7ef	68.0±3.7ef
		Retrograded	39.8±3.4	0.021	45.3±4.1f	65.0±2.2f
Manumbaey	29.9	Cooked	66.6±4.1	0.028	86.8±4.9cd	87.3±2.7cd
		Retrograded	50.9±5.0	0.035	62.9±6.5e	74.2±3.6e
Kustiyam	18.7	Cooked	41.5±2.7	0.031	52.4±5.9ef	68.5±3.2ef
		Retrograded	38.0±4.7	0.023	43.5±2.7f	63.6±1.5f
Kinaures	9.8	Cooked	73.4±6.4	0.080	104.2±4.5b	96.9±2.5b
		Retrograded	68.3±2.4	0.030	85.6±6.4cd	86.7±3.5cd
Bagoean	0	Cooked	68.0±6.4	0.062	95.8±8.8bc	92.3±4.8bc
		Retrograded	35.0±1.5	0.043	47.3±2.3f	65.7±1.2f
Karaya	0	Cooked	85.4±0.7	0.129	126.6±1.6a	109.2±0.9a
		Retrograded	53.5±7.1	0.112	78.2±8.7d	82.6±4.7d

HI, hydrolysis index; GI, estimated glycemic index; Different letters (a–f) within one column denote statistically significant differences ($P < 0.05$).

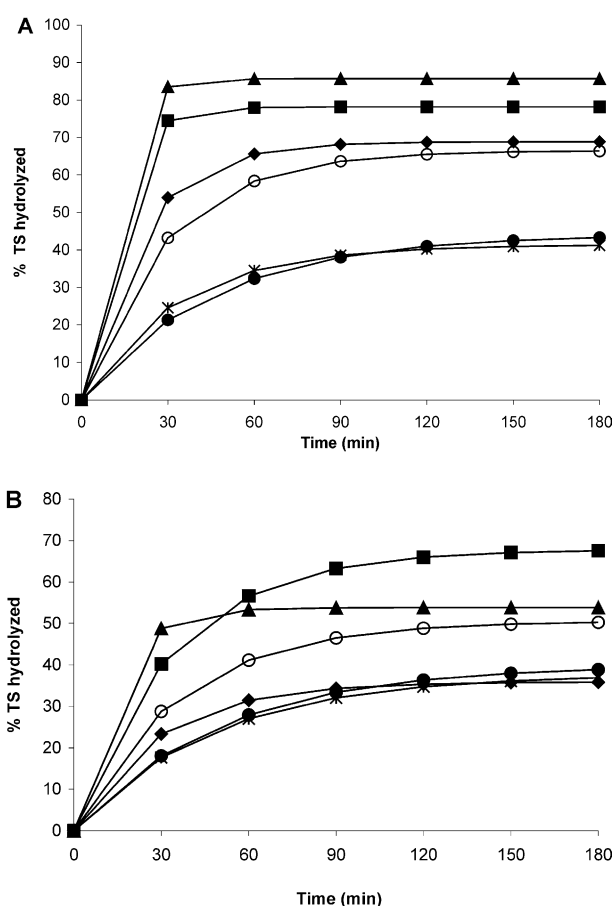


Fig. 1. In vitro starch hydrolysis rate of (A) cooked and (B) retrograded rice samples: Milagroa (●), Manumbaey (○), Kustiyam (*), Kinaures (■), Bagoean (◆), and Karaya (▲).

cultivar, Kustiyam also showed only a weak and non-significant retrogradation effect, while Kinaures fell significantly by 10.2 from 96.9 to 86.7.

Fig. 1 shows the starch hydrolysis curves for all freshly cooked (A) and retrograded (B) samples. The curves exhibit significantly different kinetics of starch

hydrolysis between cultivars but also between treatment groups. In the case of freshly cooked samples, the cultivars Karaya (waxy) and Kinaures (9.8% amylose) had already reached an equilibrium after 60 min, and the cultivar Bagoean (waxy) after 90 min. The cultivar Manumbaey (high amylose) reached its equilibrium after 150 min and, for the remaining two cultivars, hydrolysis continued slowly until the end point at 180 min. After 24 h of storage in the refrigerator, only the waxy cultivator, Karaya, reached an early equilibrium after 90 min, whereas hydrolysis continued and did not seem to be terminated after 180 min in the case of all other cultivars.

4. Discussion

Previous studies on rice starch digestibility led to the conclusion that rice should generally be classified as a high glycemic index food (Björck, 1996; Jenkins et al., 1984; Miller et al., 1992). In contrast, this study reveals that the selection of a certain cultivar as well as the adaptation of processing might offer the possibility of substantially reducing the glycemic response to ingested rice. The investigated samples can be classified into high GI and low GI rice cultivars. The waxy cultivar, Karaya, for example, shows a GI value higher than that of white bread (100). The extremely rapid starch hydrolysis might be ascribed to the relatively long cooking time (30 min). GI values higher than 100 have previously been reported for millet, cornflakes, baked and instant potato and sugars (Jenkins et al., 1984). In contrast, the low GI values exhibited by some cultivars (Milagroa, Kustiyam), especially after storing, fall within range of the given literature data for legumes (Bravo et al., 1998; Goñi et al., 1997; Jenkins et al., 1984). Substantial differences in starch digestibility based on the cultivar and the treatment are thus obvious.

The influence of amylose content on starch digestibility stated previously (Goddard et al., 1984; Juliano et al., 1986; Miller et al., 1992) could be confirmed. However, the statistically significant differences in digestibility between cultivars equal in amylose content (Milagrosa–Manumbaeay and Bagoean–Karaya) indicate that other factors, such as physicochemical properties, granule size and degree of crystallinity might also substantially affect starch digestibility. Panlasigui et al. (1991) found a high gelatinization temperature, a high minimum cooking time, a low amylograph consistency, and a low volume expansion upon cooking to be good predictors of a low glycemic response, aside from a high amylose content.

The GI values found for the two waxy cultivars are relatively high, with that of the cultivar Karaya being even higher (109) than that of fresh white bread. The lower glycemic response of high amylose cultivars, as compared with waxy varieties, has been attributed to the formation of complexes between amylose and lipids upon heating, thus entailing reduced enzyme susceptibility (Goddard et al., 1983). Guraya et al. (1997) demonstrated substantially reduced digestibility of non-waxy starch after complex formation, particularly with long chain saturated emulsifiers, whereas waxy starch did not form complexes with most emulsifiers. Amylopectin side-branches are likely to be too short for complex formation. Other suggested explanations for the lower glycemic response of high amylose starches include incomplete gelatinization of amylose under normal cooking conditions (Björck, 1996). Gelatinization temperature, which is directly related to cooking time (Juliano, 1985), is known to be positively correlated with amylose content (Frederiksson, Silverio, Andersson, Eliasson, & Aman, 1998). This, however, is not likely to be a crucial factor in this study, since samples were cooked for 30 min and gelatinization was presumably completed for all samples.

Reduction of the glycemic response through retrogradation has been demonstrated for both waxy and non-waxy starch. The slowdown of starch hydrolysis through storage is caused by recrystallization of previously gelatinized starch. Retrogradation of amylose is an irreversible process (Englyst & Cummings, 1986) and leads to the formation of an indigestible and physiologically important starch fraction which has been classified as resistant starch type 3 (Englyst et al., 1992, Englyst, Kingman, Hudson, & Cummings, 1995). Retrogradation of amylopectin is a more complex phenomenon and depends largely on the botanical source, the structure and the storage conditions (Frederiksson et al., 1998). It has been stated that retrogradation of amylose reaches a limit after 2 days, whereas retrogradation of amylopectin continues until 30–40 days after gelatinization. Long-term storing and retrogradation processes, however, usually lead to sensory

properties which are unacceptable for consumption (Eliasson & Gudmundsson, 1996). Reduced enzyme susceptibility of recrystallized amylopectin was observed previously, e.g. by Eerlingen, Jacobs, and Delcour (1994) in the case of maize. In that study, particle size and physical accessibility were also cited as factors reducing retrograded amylopectin enzyme susceptibility, aside from crystallinity. In the current study, the extent to which the digestibility of the waxy varieties was reduced after only 24 h of storing is surprising and may be due to the fact that digestion was not terminated after 180 min. This would be in agreement with the findings of Eerlingen et al. (1994) who found a high level of resistant starch (42%) in retrograded waxy maize starch after 120 min of enzymatic incubation but almost none after 6 h. Although the conditions of retrogradation were optimal at a temperature of 4 °C (Baik et al., 1997) and do not quite correspond to the actual storage conditions occurring in rural areas in developing countries, it can be assumed that the habit of pre-cooking rice—as was observed in the area of sample collection—affects rice starch digestibility through retrogradation. Likewise, the potential of this alternative processing in the formulation of low GI diets is demonstrated, which is particularly relevant for individuals suffering from an unnatural carbohydrate metabolism (Jenkins et al., 1984, 1988). These findings should be further consolidated through direct in vivo measurement of GI. Furthermore, it has been suggested that slow carbohydrate digestibility is usually associated with a reduced subjective sensation of hunger in the hours following ingestion, due to a slowdown of the carbohydrate oxidation rate (Sparti et al., 2000). This is not only relevant to persons trying to control their food intake but also has great significance in regions where food is temporarily scarce, as can be the case in developing countries.

5. Conclusion

While previous studies found notable differences in glycemic response to carbohydrate foods of different plant origins, it has been demonstrated that starch degradability can differ significantly within one botanical food source. The identification of favourable cultivars is particularly relevant in the case of rice, one of the most important carbohydrate foods worldwide.

The results clearly demonstrate that numerous factors, aside from the botanical source, must have an impact on starch digestibility. Specific chemical properties, such as amylose content, as well as the treatment, have been demonstrated to be two of these factors. Both high amylose and waxy cultivars respond to cool storing with a slowdown of starch digestion. The decrease in estimated glycemic index was shown to be most substantial for

those cultivars with a particularly high estimated glycemic index in the freshly cooked form, and more specifically the waxy ones. Our results show that it would be worthwhile to re-evaluate the indigenous knowledge of rice preparation and consumption. The mechanisms leading to a lower digestibility of high amylose starch are not fully understood and explanations found in literature are somewhat vague. Furthermore, starch retrogradation and its possible application in the adaptation of diets to diabetic metabolism need to be further studied, including *in vivo* trials. More knowledge is also needed of the influence of storage condition and storage time, as well as the amylose/amylopectin ratio, on retrogradation behaviour.

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