Development of a Low Glycemic Maize Starch: Preparation and Characterization

Xian-Zhong Han,[†,‡] Zihua Ao,[†] Srinivas Janaswamy,[†] Jay-Lin Jane,[§] Rengaswami Chandrasekaran,[†] and Bruce R. Hamaker*[,†]

Whistler Center for Carbohydrate Research and Department of Food Science, 745 Agriculture Mall Drive, Purdue University, West Lafayette, Indiana 47907-2009, and Department of Food Science and Human Nutrition, Iowa State University, Ames, Iowa 50011

Received December 23, 2005; Revised Manuscript Received February 13, 2006

A low glycemic index starch was developed by partial α-amylase treatment, and its fine structure responsible for slowly digestible and resistant properties was investigated. Different digestion rates were obtained for gelatinized, retrograded starch by varying the enzyme dosage and reaction time. Analysis by high performance size-exclusion chromatography (HPSEC) coupled with multiangle laser-light scattering indicated that the molecular weighs of amylopectin and amylose were reduced during the digestion, to less than 100 kDa. A debranched chain length study using high performance anion-exchange chromatography equipped with an amyloglucosidase reactor and a pulsed amperometric detector and HPSEC revealed that short chains of amylopectin and noncrystalline amylose were rapidly digested, while DP_n 121 chains showed resistance, followed by DP_n 46 chains. X-ray diffraction analysis revealed that the crystalline structure in the treated starches survived cooking. These starches not only have slowly digestible and resistant character, but also retain some branched structure for adequate functionality.

Introduction

Starch can be classified as containing rapidly digestible starch (RDS), slowly digestible starch (SDS) and resistant starch (RS) related to its physiological effect after consumption.[1] Substantial RDS correlated to high glycemic index (GI) and has been shown in rats to induce nonreversible insulin resistance,[2−4] stimulate fatty acid synthase and lipogenesis,[5] and promote body fat deposit.[6,7] In obese teenagers, the rapid absorption of glucose after consumption of the high GI meals was shown to induce a sequence of hormonal and metabolic changes that promoted excessive food intake.[8] In contrast, in the low GI meals, less starch is digested rapidly and often is absorbed slowly. Low GI meals lower nonfasting plasma glucose, plasma triacylglycerol, and adipocyte volume in rats[9] and prolonged satiety in obese adolescents.[10] Slowly digestible starch has low GI and provides sustained postprandial levels of glucose in blood that may be related to satiety, since transient declines in blood glucose signal meal initiation in rats[11] and humans.[12] Resistant starch, on the other hand, reduces starch availability for digestion and produces short-chain fatty acids in the large bowel through fermentation, which is beneficial for colon health[13] and may protect against colorectal cancer.[14,15]

Among the different resistant starches, the retrograded resistant starch (RS3) has great commercial importance, since its crystalline polymorphs exhibit an endothermic transition from 120 to 165 °C[16,17] that typically survive most, but not all, food processing conditions. RS3 was initially hypothesized as a crystalline state of amylose double helices.[18] Later, the noncrystalline part of retrograded starch was also found to be enzyme resistant.[19,20] Gidley et al.[21] characterized resistant starch as aggregated/gelled amylose with high double helix content, low crystallinity, and a degree of polymerization (DP) of junction zones ranging from 10 to 100. Eerlingen et al.[22] reported that DP of the RS crystallites varied only between 19 and 26 and is independent of the chain length of the amylose. Resistant starch isolated from ileostomates showed[23] two main proportions of DP 38 and over 100. Compared to current understanding of resistant starch, little is known about the nature of slowly digestible starch, although its existence in raw starch[24] and creation by retrogradation of debranched starch have been well-documented.[25]

As normal maize starch contains both linear amylose and branched amylopectin with short and long chains of different sizes, our approach was to manipulate it at the molecular level through physical and enzyme treatments to attenuate starch digestion properties. The objectives of this work were (1) to make a cooking-stable starch having slowly digestible and resistant properties and (2) to study the structure of the created starch with modified digestion. It is demonstrated that, by controlling the digestions of retrograded normal maize starch to varying extents with further processing, starches of different digestion rates can be achieved.

Experimental Section

Materials. Normal maize starch was from Tate & Lyle North America, Decatur IL. Porcine pancreatic α-amylase was from Sigma-Aldrich, St. Louis, MO (Sigma A-3176).

Preparation of Low GI Starch. Normal maize starch (5 wt % in diluted pH 6.9 buffer solution containing 0.0625 mM sodium glycerolphosphate-HCl, 1.563 mM NaCl, and 0.313 mM $CaCl_2$) was heated to over 80 °C with stirring to gelatinize starch, and thereafter autoclaved (121 °C) for 15 min. The gelatinized starch solution was cooled and stored at 4 °C for 12 h to allow retrogradation. It was then warmed to 37 °C and digested by porcine pancreas α-amylase at 15 U/g of starch

* Corresponding author. Fax: (765) 494-7953. E-mail: hamakerb@purdue.edu.

† Purdue University.

‡ Current address: Research and Development, Tate & Lyle North America, Decatur, IL 62525.

§ Iowa State University.

10.1021/bm050991e CCC: $33.50
Published on Web 03/22/2006

for 1 h, 30 U/g of starch for 2 h, and 45 U/g of starch for 4 h (U is defined as the amount of enzyme that liberates 1.0 mg of maltose from starch in 3 min at pH 6.9 at 20 °C). Immediately after the reaction, the solutions were autoclaved for 15 min and cooled in a water bath and 10% of ethanol (v/v) was added to facilitate precipitation of retrograded starch. The mixtures were transferred to 4 °C and stored overnight (12 h). The precipitated starch was collected by centrifugation at 6000*g* for 15 min, washed with purified water (half of the original volume of buffer), and collected by centrifugation twice. This starch was mixed with water (half of the original volume of buffer) and spray-dried at a flow rate of 30 mL/min with inlet and outlet temperatures of 120 and 75 °C, respectively (NIRO Automizer, Copenhagen, Denmark).

In Vitro Digestibility Assay. In vitro digestibility was analyzed using the method of Zhang and Hamaker[26] with minor modification. Starch (500 mg) with 5 mL of purified water in a sealed 30-mL bottle was cooked in a boiling water bath for 10 min. Buffer (20 mL) containing 1 mM sodium glycerophosphate-HCl, pH 6.9, 25 mM NaCl, 5 mM $CaCl_2$ was added to the cooked starch. The solution was equilibrated at 37 °C for 0.5 h and 150 U of α-amylase added. Enzyme digestion was carried out at 37 °C, and 0.5 mL aliquots of hydrolyzed solution were withdrawn at selected times. The equivalent reducing sugar value of maltose was determined using the Somogyi−Nelson method.[27]

In Vivo Assay of Acute Glucose Response in Rats after Gavage of Starch. Starch samples (10 wt % in water) were cooked in boiling water for 10 min and cooled at room temperature for 1 h. Sprague−Dawley rats (age 44−48 days and weight 175−199 g) (10 rats for each treatment) were fed 2.3 mL (10% starch) of cooked starch samples, and blood samples were drawn at 0, 10, 20, 30, 45, 60, 90, 120, and 180 min after oral gavage. Seven rats in the control and eight rats in the testing sample successfully completed the blood sampling. Blood glucose was determined by the colorimetric method using a Cobas Mira Plus autoanalyzer (Roche Diagnostics, Mannhein, Germany).[28]

High-Performance Liquid Chromatography. Molecular weight distribution of low GI starch was analyzed using high performance size-exclusion chromatography (HPSEC) coupled with a multiangle laser-light scattering (MALLS) and a refractive index (RI) detector.[29] Starch samples (100 mg) were dissolved in 95% dimethyl sulfoxide (10 mL) and boiled with continuous stirring for 1 h. The samples were cooled to room temperature and stirred for an additional 24 h. The completely dissolved samples were precipitated with ethanol (40 mL) and centrifuged at 3800*g* for 15 min. The precipitates were washed with 40 mL of ethanol and collected with centrifugation at 3800*g* for 15 min two times. The precipitates were dried for 24 h in a vacuum at room temperature. Dried samples (15 mg) were added with 5 mL of purified water and boiled with stirring for 10 min. The samples were cooked in a pressure cooker (121 °C) for 25 min and immediately injected into an HR 16/50 Pharmacia column (Sephacryl S 500 HR gel, exclusion range M_r 4×10^5 to 2×10^7, Pharmacia, Sweden) with a Varian 9012 HPLC solvent delivery system, a Varian 9040 reflective index detector (Varian Associates, Inc., Walnut Creek, CA), and a multiangle laser light scattering detector (Wyatt/Optilab 903 interferometric refractometer and DAWN DSP laser photometer) (Wyatt Technology Corp., Santa Barbara, CA). The mobile phase was 0.02% sodium azide aqueous solution filtered through a 0.22 μm GV Durapore membrane (GA Durapore Membrane Filters, Millipore, Ireland), and the flow rate was 1.5 mL/min.

Chain Length Distribution of Debranched Starch Analysis. The chain length distribution of debranched starch was analyzed using high performance anion-exchange chromatography equipped with an amyloglucosidase reactor and a pulsed amperometric detector (HPAEC-ENZ-PAD) and high performance size exclusion chromatography (HPSEC). Starch samples were debranched by isoamylase using the procedure by Jane and Chen.[30] Chain-length distributions of debranched starch samples were analyzed using HPAEC-ENZ-PAD (Dionex, Sunnyvale, CA) following the method of Wong and Jane.[31] A PA-100 anion-exchange analytical column and a guard column (Dionex, Sunnyvale, CA) were used. The separation gradient, composed of eluent A (100 mM sodium hydroxide) and eluent B (100 mM sodium hydroxide, 300 mM sodium nitrate), was as follows: 0−5 min, 99% A and 1% B; 5−30 min, linear gradient to 8% B; 30−150 min, linear gradient to 30% B; 150−200 min, linear gradient to 45% B. Samples were analyzed in duplicate. The debranched starch chain distribution was also analyzed to determine long and extra-long chains using an HPSEC equipped with an RI detector.[32] Standards (pullulan and glucose) were used to estimate molecular weights (MW).[30]

X-ray Diffraction. X-ray diffraction patterns were obtained using a Philips PW3710 diffractometer with a step width of 0.01° in the 2θ range 10°−35°. Cu Kα ($\lambda = 1.5418$ Å) radiation was used and the tube was operated at 40 kV and 25 mA. The time spent at each step was 3 s. Moisture levels of all starch samples were equilibrated in desiccators containing a saturated solution of K_2CO_3 at room temperature for a week. In the assessment of cooking on the crystallinity of starch, starch (10% in water) was cooked in a boiling water bath in a sealed 30-mL tube for 10 min and immediately transferred to a 37 °C water bath. After 30 min, the starch solution was frozen in liquid nitrogen and kept in a freezer (−20 °C) overnight. The frozen starch samples were freeze-dried (VirTis Genesis 25 ES, VirTis, Gardiner, NY) and moisture levels equilibrated using the same method as above. Powdered starch (~1 g) was packed in an aluminum holder, and X-ray data were collected at room temperature. Starch crystallinity was measured on the basis the ratio of crystalline to amorphous material.[33]

Differential Scanning Calorimetry (DSC). The thermal properties of starch samples were determined using a differential scanning calorimeter (DSC-7, Perkin-Elmer, Norwalk, CT). Approximately 6 mg of starch was weighed in a stainless steel pan, mixed with 18 mg of deionized water, and sealed. Sample was allowed to equilibrate for 2 h and scanned at a rate of 10 °C/min over a temperature range of 25−170°C. An empty pan was used as the reference.

Viscosity of Cooked Starch Solution at Different Shear Rates. Starch solution (10% in purified water) was cooked in boiling water for 10 min and then placed in a 27°C water bath for 2 h. Starch solution (1.2 mL) was transferred onto the center of the plate of a controlled stress rheometer (ReoLogica Instruments AB, Sweden). Measurements were conducted using a cone and plate system with a cone of 4 cm diameter and 4° angle. Steady shear measurements were conducted at 27 °C using a range of shear rates of 5.6−238 1/s and the resulting flow curves analyzed. All the measurements were conducted in duplicate.

Results and Discussion

Digestibility of Normal Maize Starch after α-Amylase Treatment. Raw starch was treated at three specific dosages and digestion times, 15 U/g of starch for 1 h, 30 U/g of starch for 2 h, and 45 U/g of starch for 4 h, denoted hereafter as Tr1, Tr2, and Tr3, respectively. Subsequently, after cooking, α-amylase digestion rates were found to be significantly lower than for the untreated starch (Figure 1). For example, at 30 min, the digestions were reduced by 21, 35, and 46% from the control, respectively. Even after 3 h, this trend was maintained (13, 21, and 28%). Thus, it was hypothesized that there are rapidly digestible, slowly digestible, and resistant starch fractions in the treated samples. To test this, rats were fed (by gavage) Tr2 and normal maize starch after boiling 10% of each in sealed bottles for 10 min. Their blood glucose levels were then analyzed up to 3 h (Figure 2). Postprandial glycemic response was substantially lower in the treated than in the original starch, implying reduced digestion or absorption of the treated starch (Figure 2). The area under the glucose curve, namely glycemic index (GI), showed 35% reduction. The GI in rats has been related to GI in humans.[34] In addition, the peak glucose level is related to the amount of insulin released to blood.[35] The

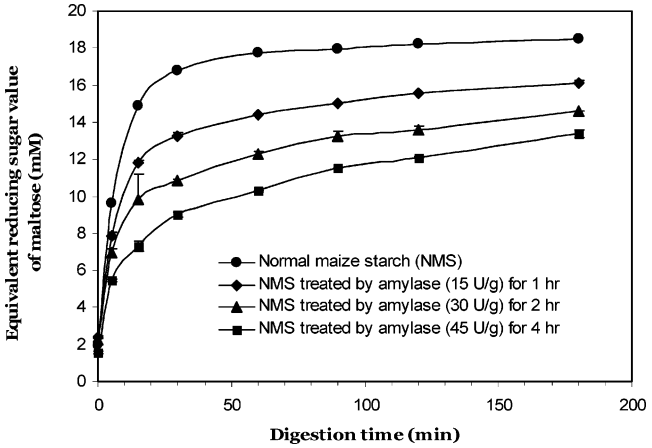

Figure 1. In vitro digestibility of normal maize and α-amylase-treated normal maize starches after cooking.

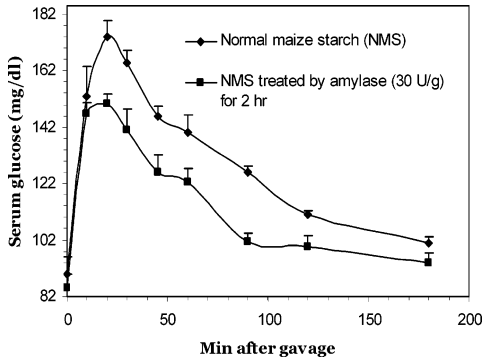

Figure 2. Acute postprandial glucose response of rats after gavages of cooked normal maize and α-amylase-treated normal maize starches.

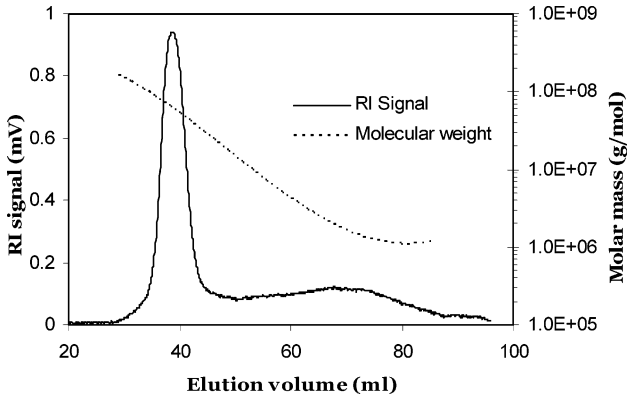

Figure 3. Chromatogram and molar mass distribution of normal maize starch by high performance size-exclusion chromatography coupled with MALLS.

observed difference of 24 mg/dL in the peak glucose levels at 20 min digestion was much more than that after 2 h (11.4 mg/dL) or 3 h (7.3 mg/dL) digestion. This indicates that glucose absorption from cooked normal maize starch by rats is more rapid and more starch is digested slowly in the α-amylase-treated starch.

Molecular Weight and Debranched Chain Length Distribution. The changes in molecular size after amylase treatment were studied to understand how the treated starch structure was prone to be more slowly digested and resistant to digestion. Figure 3 shows the HPSEC/MALLS-generated molecular weight (MW) profile of normal maize starch. After treatment with gelatinization, retrogradation, and amylase digestion, the most significant change was that the MW was considerably reduced

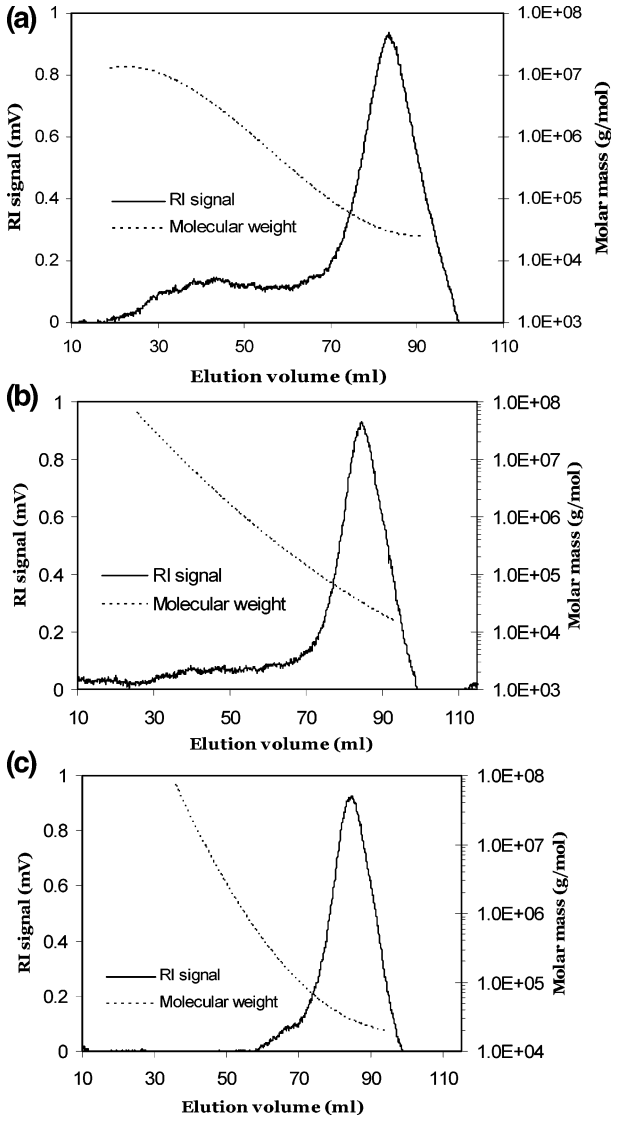

Figure 4. Chromatogram and molar mass distribution of α-amylase-treated normal maize starch determined by high performance size-exclusion chromatography coupled with MALLS: (a) 15 U/g of starch for 1 h, (b) 30 U/g of starch for 2 h, and (c) 45 U/g of starch for 4 h.

and to a fairly narrow distribution (Figures 4a−c). Also, increasing enzyme dosage and digestion time resulted in progressively smaller amounts of higher molecular weight material.

To further characterize the structure of the α-amylase degraded starches, chain length distributions of debranched starch were analyzed by both HPAEC-ENZ-PAD and HPSEC-RI. Detailed short and medium size chains are shown by HPAEC-ENZ-PAD, whereas HPSEC-RI was used to detect the long and extra-long chains of amylopectin.[32] In normal debranched maize starch, significant amounts of short chains of DP_n 12−13 were detected (Figure 5). In contrast, the proportion of short chains was significantly reduced in treated starches (Figures 6a−c), and this number decreased considerably with increasing amylase dosage and digestion time. The inference, therefore, is that short chains of amylopectin molecules in the retrograded material were preferentially digested by amylase.

The chain length distribution of debranched starches obtained by HPSEC-RI confirmed that the short chains around DP_n 12 were markedly less due to amylase treatment (Figure 7). Surprisingly, amylose (DP_n 665) was also found to be preferentially digested. In this regard, it can be postulated that such

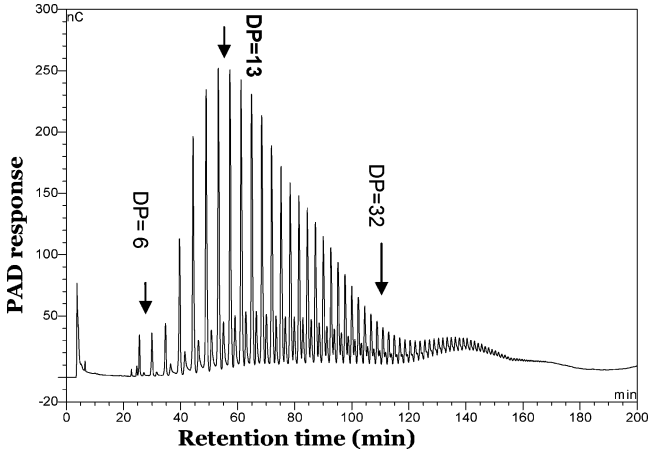

Figure 5. Chromatogram of isoamylase debranched normal maize starch determined by high performance anion-exchange chromatography equipped with an amyloglucosidase reactor and a pulsed amperometric detector.

long chains may be involved in retrogradation or formation of junction zones to nucleate an ordered domain, while a significant part of its structure would still be in an amorphous state. It is nearly impossible for these chains to form a perfect crystalline structure. On this basis, it is proposed that the amorphous regions of retrograded amylose are easy targets for α-amylase during treatment. Thus, these short chains and amorphous regions of amylose appear to account for the rapidly hydrolyzed starch during α-amylase treatment.

In the case of Tr3, the predominant peak (DP_n 121) of branched chains of the digested material extended to about DP 46 (Figure 7). Since linear chains of DP_n 121 existed in smaller amounts in normal maize starch and progressively increased (in proportion) upon enzyme treatment, they are likely to be retrograded glucan stretches and either resistant or slowly digestible in nature. Among the three treated samples, DP_n 46 chains showed a similar trend, but the effect was the most pronounced for Tr2. Thus, we infer that these chains are likewise slowly digestible and/or partially resistant to amylase digestion.

It has been reported that the isolated RS after dietary fiber analysis has DP between 19 and 26, regardless of the original chain length.[22] Similarly, the RS of DP 43 is formed after porcine pancreatic α-amylase digestion and that of DP 32 after acid hydrolysis.[17] Both studies showed more extensive hydrolysis due to higher enzyme or acid concentration than in our experiments. Therefore, the linear chains of the larger DP_n 121 in Figure 7 are due to relatively milder hydrolysis. Starch linear chains of DP_n 121 may still be digestible under more extensive digestion conditions, but they would be digested at a much slower rate than short chains or part of the extra-long chains. These chains may also contribute, to some extent, to form crystalline structure along with nearby short noncrystalline regions. Small-angle X-ray scattering of retrograded starch by others showed much lower crystallinity than native starch granules.[36,37] Thus it can be speculated that the short noncrystalline regions between crystalline regions in our treated samples could be slowly digested or resistant to digestion, depending on the length of these noncrystalline regions and their locations (surface or interior) in crystallites.

X-ray Analysis. X-ray powder diffraction patterns of normal maize starch and the three treated starches (Tr1, Tr2, Tr3) are shown in Figure 8. Their crystallinities are 24.7, 27.2, 24.6, and 21.5%, respectively. Normal maize starch had a typical A-type pattern with the most intense bands corresponding to Bragg angles (2θ) 15, 17, 18.1, and 23.3° as reported by

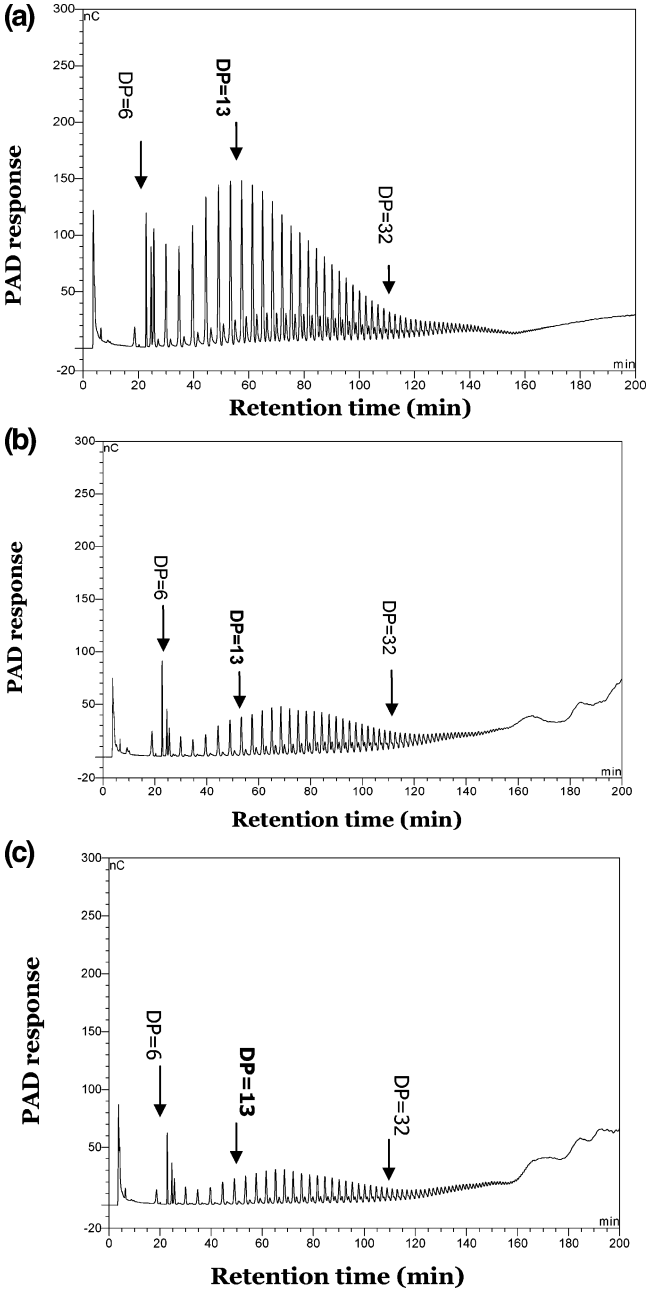

Figure 6. Chromatogram of isoamylase debranched α-amylase-treated normal maize starch determined by high performance anion-exchange chromatography equipped with an amyloglucosidase reactor and a pulsed amperometric detector: (a) 15 U/g of starch for 1 h, (b) 30 U/g of starch for 2 h, and (c) 45 U/g of starch for 4 h.

Planchot et al.[38] On the other hand, the diffraction profiles for the treated samples were of a different nature. All the three treated samples diffracted similarly (Figure 8) and correspond to a composite of the B-type crystalline structure and V-form of amylose−ethanol or amylose−lipid complex. It is known that lipid-complexed amylose in the amorphous region of starch can be partially hydrolyzed to form B-type crystalline structures that are resistant to hydrolysis during lintnerization.[39] It is also known that when ethanol is used in the precipitation of amylase-treated starches, it complexes with amylose and hence appears in the amylose−ethanol diffraction pattern, which is similar to that of the V-form of the amylose−lipid complex.[40,41]

After cooking and freeze-drying, no crystalline pattern was discerned for normal maize starch (Figure 9). However, all the three treated starches recovered their original diffraction patterns

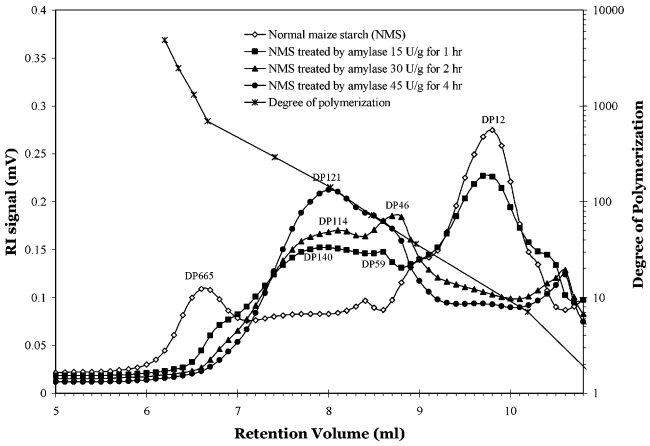

Figure 7. Chromatograms of debranched normal and α-amylase-treated normal maize starches determined by high performance size-exclusion chromatography.

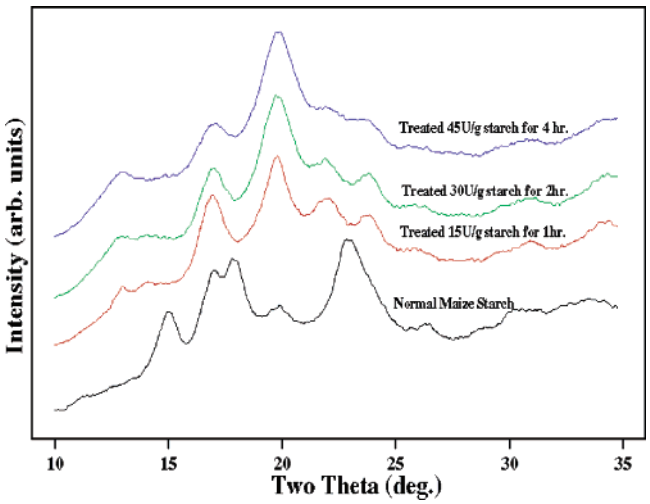

Figure 8. X-ray diffraction patterns of native and α-amylase-treated normal maize starches.

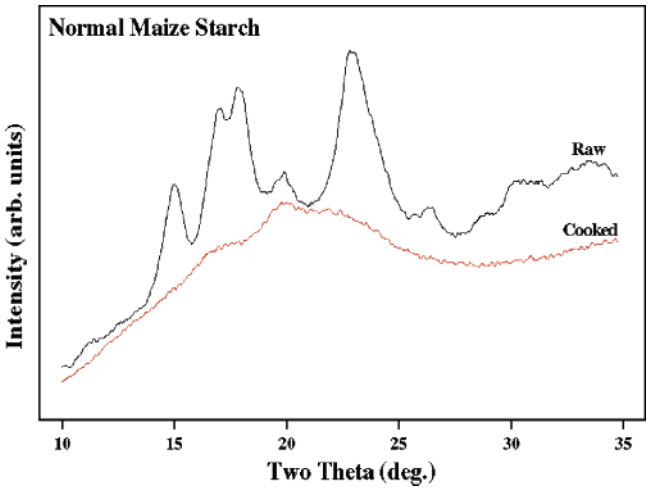

Figure 9. X-ray diffraction patterns of normal maize starch before and after cooking.

to varying degrees (Figures 10a−c). The resulting crystallinity (19.0% for Tr1, 17.9% for Tr2, and 18.6% for Tr3) is likely due to the resistance of crystallites to melt at the boiling temperature (around 100 °C), but conceivably could be due to the ability to re-form a crystalline structure quickly during the cooling process. The applied enzyme dosage and treatment time was positively correlated with the retaining of crystallinity, with

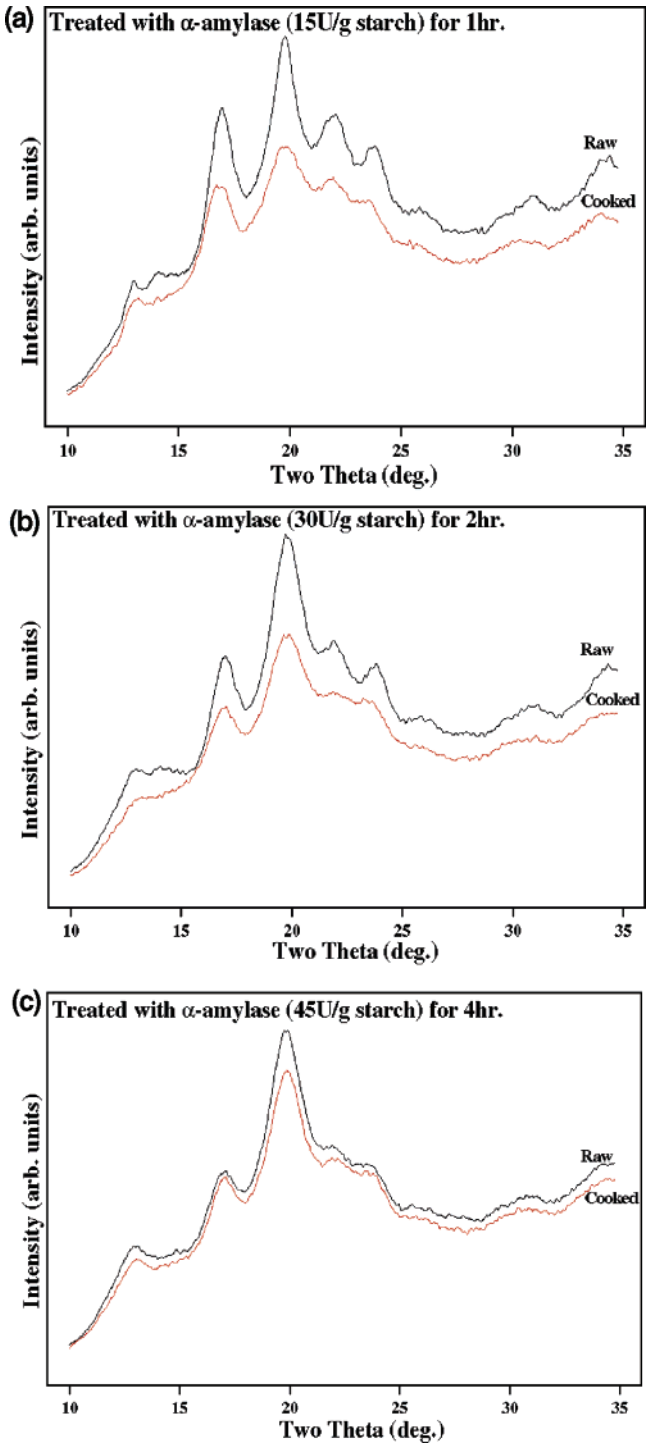

Figure 10. X-ray diffraction patterns of α-amylase-treated normal maize starches before and after cooking: (a) 15 U/g of starch for 1 h, (b) 30 U/g of starch for 2 h, and (c) 45 U/g of starch for 4 h.

Tr3 almost abutting its raw sample. Because of this ability to maintain initial crystallinity, it is not surprising that the treated samples are less digestible.

DSC Analysis. DSC thermal profiles are shown in Figure 11. In the case of normal maize starch, the first endothermic peak was at 71 °C (Tp) and the second at 93 °C, corresponding to the melting of crystallites in the native starch granules and amylose−lipid complex, respectively.[40,42] In all the three treated samples, the 71 °C gelatinization peak was absent, indicating the complete melting of the original crystalline structure. Instead, the first new endothermic peak appeared at roughly 24 °C higher, 96, 95, and 94 °C, for the three samples. The observed

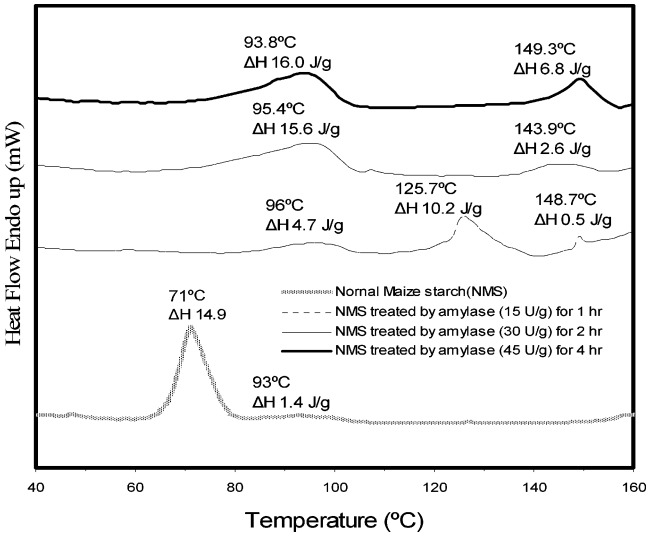

Figure 11. DSC analysis of normal and α-amylase-treated maize starches. Peak melting temperatures and melting enthalpies (Δ*H*) are shown above the thermograms.

increase in enthalpy (Δ*H*) of this peak with the extent of amylase treatment suggests that this starch fraction is partially resistant to digestion. Two possible explanations exist. First, it is possible that the endotherms at an elevated temperature of ~95 °C could represent the melting of a crystalline structure composed of longer chains of amylopectin. This is in accord with the rapid retrogradation of long chains of amylopectin in a DSC study of rescanning high amylose starch in aqueous solution.[43,44] Second, it is possible that this peak represents the amylose-lipid complex and is partially resistant to porcine α-amylase digestion. This is consistent with a report that the amylose−lipid complex slows down the digestion by pancreatic α-amylase compared to noncrystalline amylose.[45]

The second endothermic transition peak in treated starches occurred at 126, 144, and 149 °C, respectively (Figure 11). It may be recalled that B-type crystals with longer chains have higher melting temperatures than those with shorter chains.[46] The 126 °C portion of Tr1 was either digestible or partially digested during its processing, since this peak did not appear in Tr2 or Tr3 starches. A third peak for Tr1 was at about 150 °C, comparable to the second for Tr2 and Tr3. This usually is attributed to the melting of retrograded amylose.[16] However, the 144 °C peak for Tr2 was larger but broader than the corresponding one for Tr1. Hence, we hypothesize that the 126 °C peak in Tr1 represents the melting of an amylopectin crystalline structure composed of medium chains or partially crystallized long chains. With further amylase treatment for 2 h (Tr2), both short and medium chains of amylopectin were significantly reduced, thereby likely leading to its disappearance and resulting in a crystalline mixture of long chains of amylopectin and amylose of varying sizes so that the melting profile at 144 °C was broader and lower. As the amylase treatment was increased to 4 h (Tr3), more and more of the short and medium chains were digested, and the long chains of amylopectin and of partially degraded amylose of relatively uniform sizes formed crystalline structures that melted at the higher temperature of 149 °C. The increased melting enthalpy (Δ*H*) of 6.8 J/g at the high-temperature endotherm is an indication of formation of high-temperature stable crystalline structure.

Viscosity of Treated Starches after Cooking. To assess the rheological properties of the treated starches, viscosities were measured and compared in Figure 12. The appreciable viscosity

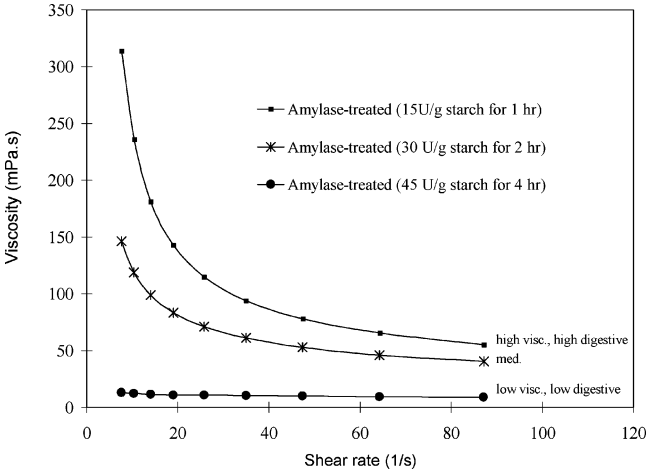

Figure 12. Viscosity of pastes of cooked α-amylase-treated normal maize starches under different shear rates.

noticed at low shear is a positive attribute of the lesser treated starches. The viscosity, however, decreased with an increase in the severity of α-amylase treatment during starch processing. Thus, the viscosity is inversely related to the digestibility of the treated starches (Figures 1 and 12). This is fully in accordance with the X-ray observations (Figure 10). In addition, the removal of short chains of amylopectin may significantly increase the thermal stability of the crystalline structure made up of longer chains that would otherwise decrease the solubility and hence viscosity of cooked starch.

Conclusions

Starches with resistant and slowly digestible properties retained after cooking were produced upon amylase treatment of normal maize starch. Postprandial glycemic response in rats was reduced. The native amylopectin and amylose molecular size was significantly and rapidly reduced during enzyme treatment to a narrow, but defined, molecular weight range. The chain length distribution of debranched starch showed that the short chains of amylopectin and noncrystalline amylose were preferentially digested (rapidly) during starch processing, while the chains of DP_n 121 were resistant to amylase treatment, followed by those of DP_n 46. Also, substantial crystallinity persisted in the treated starches after cooking. With an increase in the time and amount of α-amylase treatment, DSC enthalpy values increased for the primary peak around 95 °C and the higher melting peak around 140−150 °C. The mildly treated starches (Tr1 and Tr2) maintained appreciable viscosity following recooking. In short, our study indicates that, with α-amylase treatment, maize starch with resistant as well as slowly digesting nature retained after cooking that corresponds to a low glycemic index, can be produced. Similar behavior could be expected for treatment of other starches as well, perhaps even improving resistant and slowly digestible properties. However, its validation must await further research.

Acknowledgment. We thank Dr. Berdine Martin for her gracious help in the rat study.

References and Notes

(1) Englyst, H. N.; Kingman, S. M.; Cummings, J. H. *Eur. J. Clin. Nutr.* **1992**, *46* (Suppl. 2), S30−S50.
(2) Byrnes, S. E.; Miller, J. C.; Denyer, G. S. *J. Nutr.* **1995**, *125*, 1430−1437.

(3) Wiseman, C. E.; Higgins, J. A.; Denyer, G. S.; Miller, J. C. B. *J. Nutr*. **1996**, *126*, 410−415.
(4) Higgins, J. A.; Miller, J. C.; Denyer, G. S. *J. Nutr*. **1996**, *126*, 596−602.
(5) Kabir, M.; Rizkalla, S. W.; Quignard-Boulangé, A.; Guerre-Millo, M.; Boillot, J.; Ardouin, B.; Luo, J.; Slama, G. *J. Nutr*. **1998**, *128*, 1878−1883.
(6) Pawlak, D. B.; Bryson, J. M.; Denyer, G. S.; Brand-Miller, J. C. *J. Nutr*. **2001**, *131*, 99−104.
(7) Pawlak, D. B.; Kushner, J. A.; Ludwig, D. S. *Lancet* **2004**, *364*, 778−85.
(8) Ludwig, D. S.; Majzoub, J. A.; Al-Zahrani, A.; Dallal, G. E.; Blanco, I.; Roberts, S. B. *Pediatrics* **1999**, *103*, E261−E266.
(9) Lerer-metzger, M.; Rizkalla, S. W.; Luo, J.; Champ, M.; Kabir, M.; Bruzzo, F.; Bornet, F.; Slama, G. *Br. J. Nutr*. **1996**, *75*, 723−732.
(10) Ball, S. D.; Keller, K. R.; Moyer-Mileur, L. J.; Ding, Y.-W.; Donaldson, D.; Jackson, W. D. *Pediatrics* **2003**, *111*, 488−494.
(11) Campfield, L. A.; Smith, F. J. *Int. J. Obes.* **1990**, *14* (Suppl. 3), 15−33.
(12) Campfield, L. A.; Smith, F. J.; Rosenbaum, M.; Hirsch, J. *Neurosci. Biobehav. Rev.* **1996**, *20*, 133−137.
(13) Moreau, N. M.; Champ, M. M.; Goupry, S. M.; Bizec, B. J. L.; Kremp, M.; Nguyen, P. G.; Dumon, H. J.; Martin, L. J. *J. Nutr*. **2004**, *134*, 493−500.
(14) Emenaker, N. J.; Calaf, G. M.; Cox D.; Basson, M. D.; Qureshi, N. *J. Nutr*. **2001**, *131*, 3041S−3046S.
(15) Leu, R. K. L.; Brown, I. L.; Hu, Y.; Young, G. P. *Carcinogenesis* **2003**, *24*, 1347−1352.
(16) Sievert, D.; Pomeranz, Y. *Cereal Chem*. **1989**, *66*, 342−347.
(17) Pomeranz, Y. *Eur. J. Clin. Nutr*. **1992**, *46* (Suppl. 2), S63−S68.
(18) Jane, J.-L.; Robyt, J. *Carbohydr. Res*. **1984**, *132*, 105−118.
(19) Berry, C. S.; I'Anson, K.; Miles, M. J.; Morris, V. J.; Russell, P. L. *J. Cereal Sci*. **1988**, *8*, 203−206.
(20) Sievert, D.; Czuchajowska, Z.; Pomeranz, Y. *Cereal Chem*. **1991**, *68*, 86−91.
(21) Gidley, M. J.; Cooke, D.; Darke, A. H.; Hoffmann, R. A.; Russell, A. L.; Greenwell, P. *Carbohydr. Polym.* **1995**, *28*, 23−31.
(22) Eerlingen, R. C.; Deceuninck, M.; Delcour, J. A. *Cereal Chem*. **1993**, *70*, 345−350.
(23) Faisant, N.; Champ, M.; Colonna, P.; Buléon, A. *Carbohydr. Polym*. **1993**, *21*, 205−209.
(24) Gérard, C.; Colonna, P.; Buléon, A.; Planchot, V. *J. Sci. Food Agric*. **2001**, *81*, 1281−1287.
(25) Shi, Y.-C.; Cui, X.; Birkett, A. M.; Thatcher, M. G. U.S. Patent 2003/0215562 A1, 2003.
(26) Zhang, G.; Hamaker B. R. *Cereal Chem*. **1998**, *75*, 710−713.
(27) Chaplin, M. F.; Kennedy, J. F. *Carbohydrate Analysis*—A Practical Approach; Oxford University Press Inc.: Oxford, 1994; p 4.
(28) Gokcel, A.; Karakose, H.; Ertorer, E. M.; Tanaci, N.; Tutuncu, N. B.; Guvener, N. *Diabetes Care* **2001**, *24*, 1957−1960.
(29) You, S. G.; Lim, S.-T. *Cereal Chem*. **1999**, *76*, 116−121.
(30) Jane, J.-L.; Chen, J.-F. *Cereal Chem*. **1992**, *69*, 60−65.
(31) Wong, K. S.; Jane, J. *J. Liq. Chrom. Relat. Technol*. **1997**, *20*, 297−310.
(32) Yoo, S.-H.; Jane, J.-L. *Carbohydr. Polym*. **2002**, *49*, 297−305.
(33) Cairns, P.; Bogracheva, T. Y.; Ring, S. G.; Hedley, C. L.; Morris, V. J. *Carbohydr. Polym.* **1997**, *32*, 275−282.
(34) Wolever, T. M. S.; Jenkins, D. J. A.; Jenkins, A. L.; Josse, R. G. *Am. J. Clin. Nutr*. **1991**, *54*, 846−854.
(35) Jeppesen, J.; Hollenbeck, C. B.; Zhou, M. Y.; Coulston, A. M.; Jones, C.; Chen, Y. D. I.; Reaven, G. M. *Arterioscler. Thromb. Vasc. Biol*. **1995**, *15*, 320−324.
(36) Suzuki, T.; Chiba, A.; Yano, T. *Carbohydr. Polym*. **1997**, *34*, 357−363.
(37) Shamai, K.; Shimoni, E.; Bianco-Peled, H. *Biomacromolecules* **2004**, *5*, 219−223.
(38) Planchot, V.; Colonna, P.; Buléon, A. *Carbohydr. Res*. **1997**, *298*, 319−326.
(39) Morrison, W. R.; Tester, R. F.; Gidley, M. J.; Karkalas, J. *Carbohydr. Res*. **1993**, *245*, 289−302.
(40) Biliaderis, C. G.; Galloway, G. *Carbohydr. Res*. **1989**, *189*, 31−48.
(41) Rondeau-Mouro, C.; Le Bail, P.; Buleon, A. *Int. J. Biol. Macromol*. **2004**, *34*, 309−315.
(42) Villwock, V. K.; Eliasson, A.-C.; Silverio, J.; BeMiller, J. N. *Cereal Chem*. **1999**, *76*, 292−298.
(43) Brumovsky, J.; Thompson, D. B. *Cereal Chem*. **2001**, *78*, 680−689.
(44) Thompson, D. B.; Brumovsky J. U.S. Patent 6468355B1, 2002.
(45) Holm, J.; Björck, I.; Ostrowska, S.; Elisson, A.-C.; Asp, N.-G.; Larsson, K.; Lundquist, I. *Starch* **1983**, *35*, 294−297.
(46) Moates, G. K.; Noel, T. R.; Parker, R.; Ring, S. G. *Carbohydr. Res*. **1997**, *298*, 327−333.

BM050991E