Understanding Starch Organisation in Gluten-Free Pasta from Rice Flour

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Abstract

Starches extracted from parboiled rice flour and pasta samples produced by two extrusion processes – a conventional one carried out at 50 °C and an extrusion-cooking process at 115 °C - were evaluated by DSC and SEC analysis. Molecular changes induced by both pasta-making process and following cooking in boiling water were also investigated using iodine absorption properties of samples expressed as the ratio of absorption to scattering spectra (K/S) and X-ray diffraction. A decrease in polymer chain mobility and iodine binding capacity were observed after pasta-making process. While the characteristic A-type crystalline pattern remained, the exposure to iodine vapor changed the peak intensity of starch samples, especially at 0.97 a_w. The higher melting temperature of pasta samples in comparison with parboiled rice flour reflected the decrease in mobility of the amorphous regions detected by K/S spectral analysis. The pasta making-process also affected the molecular size distribution of starch samples. In particular, the elution peak shifted toward lower fraction numbers with increasing extrusion temperature, showing a higher molecular size for starch after the extrusion-cooking. All the differences detected between starch samples according to extrusion conditions were deleted during cooking. Compared to the uncooked samples, starch from cooked pasta showed higher K/S value at all wavelengths, highlighting the increase in mobility of the amorphous region. Moreover, beside the increase in melting temperature, a decrease in endothermic enthalpy was detected, confirming the loss of order observed by X-ray diffraction.

Keywods: milled rice, starch, iodine, pasta-making

List of Abbreviations: Cooked C, starch from cooked pasta prepared by conventional extrusion; Cooked E, starch from cooked pasta prepared by extrusion-cooking; ΔH_{TS}, gelatinization enthalpy expressed on total starch content; DSC, Differential Scanning Calorimetry; GF, gluten-free; PRS, starch from parboiled rice flour; SEC, Size-Exclusion Chromatography; Starch C, starch from raw pasta prepared by conventional extrusion; Starch E, starch from raw pasta prepared by extrusion-cooking; Tc, gelatinization conclusion temperature; To, gelatinization onset temperature; Tp, gelatinization peak temperature.

1. INTRODUCTION

Since milled rice consists of about 90% starch, the structure of this macromolecular fraction and its physicochemical properties are the primary characteristics used to select rice cultivar and rice starch for specific industrial applications (Bao, 2001). Several studies have been carried out on physicochemical, morphological, thermal, rheological properties of rice starch isolated from flour (Singh et al., 2006; Vandeputte et al., 2003a; Vandeputte et al., 2003b; Vandeputte et al., 2003c). However, no studies are present on rice starch properties in a complex matrix, such as pasta, where its properties and the extent of its modifications could be influenced by the presence and the interactions with other components.

Due to the absence of gluten, rice is recommended as safe for people affected by celiac disease and it is commonly used to produce gluten-free (GF) pasta, alone or in combination with other no-gluten cereals and/or additives (Marti et al., 2010). In a preliminary study, the effect of pasta-making process on pasting, thermal, textural, and cooking properties have been evaluated in pasta from brown and milled rice flour, considering the whole product and, thus, the interactions among all the biopolymers (starch-starch and starch-protein interactions in sample from milled rice; starch-starch, starch-protein, starch-lipid, and starch- non-starch polysaccharides interactions in pasta from brown rice) (Marti et al., 2010). In order to complete and better understand the starch architecture in pasta-product it could be advisable to investigate the starch macromolecular characteristics after

54 removing all the non-starch constituents from the samples. In fact, in a GF matrix, starch cannot be
55 considered an inert filler, but it substantially contributes to the structure and the quality of the
56 product. The textural and nutritional properties of GF pasta are related both to the organization of
57 raw-material native starch and to starch modifications promoted by thermal and mechanical events
58 occurring during the pasta-making process. In order to produce a GF pasta with similar appearance
59 and texture as conventional products of *durum wheat* semolina, a new starch organization, effective
60 in substituting the gluten network in the final product, can be formed applying one or more heating
61 and cooling treatments to GF raw-material. The sequence of thermal events during gelatinization
62 promotes a loss of native granular structure of starch and an extensive reticular and fibrillar network
63 after cooling (Resmini & Pagani, 1983).
64 Combining high temperature with a large amount of mechanical energy input, extrusion causes
65 an irreversible swelling or even disruption of the starch granules, depending upon the severity of the
66 treatment applied (Karim et al., 2000). After its gelatinization, during the cooling phase starch
67 undergoes retrogradation that involves the starch chains tend to reassociate in an ordered structure.
68 Since retrogradation profoundly affects quality, acceptability, shelf-life, and nutritional properties
69 of starch-based foods, the phenomenon is of great interest to food scientists and technologists
70 (Biliaderis, 1991). Although starch retrogradation is considered a negative phenomenon in baking,
71 it could positively affect the quality of extruded products as pasta (Karim et al., 2000). In fact, in
72 order to decrease stickiness, prevent the dissolution of solid matter into the boiling water, and
73 obtain a characteristic chewiness, the production of rice noodles involves cooling cycles (Tan et al.,
74 2009). However, in pasta products starch retrogradation is often associated with an increase in
75 firmness and springiness after cooking. The objective of this paper was to better understand starch
76 modification that occurred during pasta-making process and the relationship between starch
77 structure and processing conditions. Iodine was used as tool to highlight structural differences in the
78 arrangement of pasta samples prepared by using different extrusion conditions. At the same time,
79 the molecular changes promoted by cooking were investigated.

2. MATERIALS AND METHODS

2.1 Materials

Milled parboiled rice (Thai cultivar of commercial origin; total starch: 82.6% db; protein: 6.8% db; lipid: 1.0% db; amylose: 25% db) used in this study was provided by Riso Viazzo s.r.l. (Crova, Italy).

2.2 Pasta preparation

Two pasta samples were prepared in the pilot-plant of DiSTAM (University of Milan, Milan, Italy) by using a conventional extrusion (Process C) and an extrusion-cooking pasta-making process (Process E), as reported by Marti et al. (2010). Process C was carried out in a continuous press commonly used for the production of pasta from semolina (Braibanti, Milano, Italy). Parboiled rice flour (particle size less than 250 μm) and water were blended to produce a mixture with a final water content of 40%. After mixing, the dough was formed in Pasta C, maintaining the continuous press under vacuum and keeping the dough temperature below 50-55 °C, at an extrusion pressure of 10-11 MPa. A patented process was used to produce pasta by an extrusion-cooking process (Grugni et al., 2009). The mixture of flour and water (with a final water content of 40%) was heat-treated for 2 min in a Progel® extruder (single screw; Braibanti, Milano, Italy) fed with steam at 115 °C. After the first extrusion step, the heat-treated dough was formed in the continuous extruder used for the conventional process (Braibanti, Milano, Italy), obtaining Pasta E. Both pasta samples (Pasta C and Pasta E) were dried in an experimental drying cell (Braibanti, Milano, Italy) using a low-temperature drying cycle (50 °C for 14 hours) and stored at room temperature until analyzed. Before the analysis, rice pasta samples were ground in order to produce a product with less than 500 μm particle size.

2.3 Starch isolation

Starch samples were isolated from rice flour and rice pasta samples, according to Park et al. (2006) and Goesaert et al. (2009) with some modifications. The samples were mixed with protein extraction buffer (12.5 mM sodium borate buffer, pH 10, containing 0.5% SDS and 0.5% $Na_2S_2O_5$)

106 (solvent-to-flour ratios = 1:20). After stirring for 5 min and centrifugation (3000 rpm for 5 min at
107 10°C), the supernatant was discarded and the mixing step with protein extraction buffer was
108 repeated. The pellets obtained were dispersed and washed in deionized water for three times in
109 order to remove the residual bran. After the buffer treatment, an aliquot of 100 mL of a protease
110 solution containing 0.50 g of trypsin from porcine pancreas (13,000-20,000 BAEE units/mg protein;
111 Sigma-Aldrich Chemie) and 0.25 g of papain from Carica papaya (≥3 units/mg; Sigma) was added
112 to 10 g of sample. After overnight incubation at room temperature under magnetic stirring, the
113 suspension was centrifuged (10 min, 3000g). The residues were washed three times with deionized
114 water and twice with 10.0 mL of ethanol (95%). The enzymatic degradation, incubation (3 hours,
115 magnetic stirring, room temperature) and the washing step with water and ethanol (95%) were
116 repeated. The isolated starch was washed with acetone and dried overnight. The dry starch was
117 ground lightly with a mortar and pestle (with less than 500 μm particle size) for further analysis.
118 The same procedure was carried out to isolate starch from pasta samples after cooking in boiling
119 water. Pasta C and Pasta E were cooked as reported by Marti et al. (2010); after draining, samples
120 were dipped into liquid nitrogen and then freeze-dried before starch isolation. The rapid cooling
121 contains retrogradation phenomenon which extent is at 4-6°C.

122 *2.4 Sepharose CL-2B chromatography*

123 Granular starches were prepared according to Klucinec & Thompson (1998). Starch (0.5 g)
124 was dispersed in 10 ml of 90% (v/v) dimethyl sulfoxide in water, by heating the mixture in a boiling
125 water bath with constant stirring for three hours. Following dispersion, 40 ml of ethanol was added
126 and the mixture was centrifuged at 10000 rpm for 15 minutes at 4 °C. The supernatant was
127 discarded and the pellets were washed by suspending them in 50 ml of ethanol, followed by
128 centrifugation (10000 rpm). The washing procedure was repeated once with ethanol and once with
129 acetone. The acetone-washed precipitate was dried in a forced air oven at 50 °C for 24 hours. The
130 nongranular starches were stored under desiccation ($CaSO_4$) before the analysis. Nongranular

131 starches (4 mg) were dispersed in 2 ml of NaOH 0.1 N for one night. The dispersed starches were
132 loaded into a Sepharose CL-2B (Sigma Aldrich Inc.) SEC column (50 cm x 1 cm, Pharmacia Fine
133 chemicals, Sweden) using gravity flow. The mobile phase in the system was NaOH, 0.1 N,
134 containing 0.02% (v/v) sodium azide. For each sample, 1 ml of fraction was collected by using a
135 fraction collector, at a flow rate of 0.3 ml/min and the analysis was repeated twice. Every two SEC
136 fraction was examined for total carbohydrate and iodine binding λ_{max}. Total carbohydrate of the
137 fractions was determined using the phenol-sulfuric acid assay of Dubois et al. (1956). The iodine
138 binding λ_{max} was determined using the procedure of Klucinec & Thompson (1998). An aliquot of
139 0.4 ml of a SEC fraction was neutralized with 25 μl of HCl, 1.0 N and mixed with 2.5 ml of iodine
140 solution. The iodine solution was prepared immediately before the analysis, diluting 0.5 ml of the
141 stock iodine solution (0.026 g of I_2 and 0.26 g of KI/ml water) in 130 ml of deionized water. The
142 absorbance of the solution (composed by SEC fraction, HCl, and iodine solution) was examined
143 between 450 and 800 nm (VARIAN Cary 1 Bio). The measurement was carried out in duplicate.
144 *2.5 Thermal properties*
145 The thermal properties of starch samples were determined using a Perkin Elmer Differential
146 Scanning Calorimeter (DSC-7/DX; Perkin Elmer, Cambridge, United Kingdom). Each sample was
147 weighed in a steel pan and an empty steel pan was used as reference. Distilled water was added to
148 the sample to make a 1:3 (w/w) sample:water ratio. The pans were hermetically sealed and allowed
149 to equilibrate 24 h at room temperature. Samples were heated from 25 to 180 °C at a rate of 10
150 °C/min. The onset temperature (To, °C), peak temperature (Tp, °C), conclusion temperature (Tc,
151 °C), and gelatinization enthalpy (ΔH, J/g) were determined using the software provided with the
152 equipment. All measurements were replicated at least twice.
153 *2.6 Colorimetric analyses of granular samples exposed to iodine vapor*
154 Samples of 2 g were equilibrated to the respective water activity (a_w), with final values of
155 0.33, 0.75, and 0.97 a_w using saturated solutions of $MgCl_2$ (EMD Chemicals Inc., Gibbstown, NY),
156 NaCl (Fisher Chemicals Inc., Fair Lawn, NJ), and K_2SO_4 (EDM Chemicals Inc., Gibbstown, NY)

157 (Greenspan, 1977), as described by Saibene & Seetharaman (2006). Following equilibration, the
158 moisture content of samples was measured according to the AACC method 44-15A (2000). To
159 determine the iodine binding, 0.2 g of the equilibrated sample were placed in the corresponding a_w
160 desiccator, and exposed to iodine vapor generated from 2 g of iodine crystals (J.T. Baker,
161 Phillipsburg, NJ) for 24 h at room temperature. Each sample was equilibrated and exposed to iodine
162 vapour twice. A CM 3500-d Spectrophotometer (Konika Minolta, Mahwah, NJ, USA) was used to
163 evaluate the color (expressed as luminosity) and the K/S value of stained samples. The K/S value of
164 the samples after iodine exposure was measured at wavelength range from 400 to 700 nm, at 10 nm
165 intervals, using unstained starch as the target color.

166 *2.7 Wide Angle X-ray Powder Diffraction*

167 Wide angle X-ray diffraction measurements were carried out on starch samples (0.1 g) after
168 equilibration, before and after iodine exposure (Rigaku Powder Diffractometer equipment - Rigaku
169 Co., Tokyo, Japan). CuKα1 radiation (λ = 1.54 Å) was selected using a quartz monochromator and
170 the operational settings for the diffractometer were 40mA and 40kV. For this instrument, the
171 diffractometer had a 0.5° divergence slit, a 0.33 mm receiving slit and a 0.5° scattering slit. The
172 samples were scanned in the range 3-35° 2θ at a rate of 1° 2 θ per minute. Data were smoothed
173 using Jade 6.5 software and were normalized to equal total scattering in 3-35° 2θ range. The
174 analysis was done in duplicate.

175 **3. RESULTS AND DISCUSSION**

176 *3.1 Sepharose CL-2B chromatography*

177 The Sepharose CL-2B chromatograms of starch samples are shown in Figure 1. Two main fractions
178 can be distinguished for each sample: the amylopectin peak showing iodine absorption at about 550
179 nm, and the amylose fraction with high iodine binding absorption at more than 600 nm. In parboiled
180 rice starch (PRS), the molecular weight of the peak corresponding to amylopectin was smaller than
181 the corresponding peaks observed for the starch from pasta samples (Starch C and Starch E). Pasta
182 samples exhibited a sharp transition from the low wavelength fraction to the high wavelength

183 fraction (Figure 1). Compared to PRS, the increase of the molecular weight of amylopectin peak
184 after pasta-making process was accompanied, at the same time, by the formation of amylose
185 fraction characterized by lower λ_{max}. The extrusion conditions affected the molecular size
186 distribution in pasta samples; the elution peak shifted toward lower fraction numbers with
187 increasing extrusion temperature, indicating an increase of the molecular size after extrusion-
188 cooking (Figure 1). The results suggest that the amylopectin matrix is likely a combination of
189 amylose and amylopectin chains. The average molecular weight of amylose and amylopectin, as
190 well as their molecular organization within the granule, affected starch functionality. During
191 extrusion-cooking, the higher extrusion temperature and mechanical stress associated with the
192 screw turning promoted starch gelatinization, involving the disruption of molecular order within the
193 starch granule. After heating, retrogradation occurred and starch polymers showed a tendency to
194 reassociate in an ordered structure, resulting in a new rearrangement (Atwell et al., 1988).
195 Moreover, the amylopectin structure seems to explain the differences in texture among rice
196 varieties. According to Reddy et al. (1993), the presence of a large number of long unbranched
197 chains at the exterior region of the amylopectin molecule leads, by their interaction, to the
198 formation of strong and elastic starch aggregates as a consequence of the numerous interactions
199 arising among chains: the new organization could account for firm and non-sticky cooked rice. On
200 the other hand, the lack of such chains makes starch material weak and fragile, and the cooked rice
201 results to be soft and sticky (Reddy et al., 1993). In the present study, the structure of the
202 amylopectin could account for the texture of the cooked product: Starch E, characterized by high
203 molecular weight distribution (Figure 1) resulted in a product characterized by extreme firmness
204 after cooking, as recently reported by Marti et al. (2010).
205 Further changes occurred in starch samples after cooking, according to the extrusion
206 conditions (Figure 1). Compared to the uncooked sample, the SEC chromatograms of cooked starch
207 from conventional extrusion shifted towards lower fraction numbers, indicating that the cooking
208 process resulted in an increase in molecular size, while the absorbance values were lower at values

209 similar to rice flour starch. On the other hand, the profile related to the amylose fraction did not
210 change after cooking, keeping the iodine binding λ_{max} values between 600 and 640 nm. In Starch E,
211 cooking did not change the SEC profile compared to uncooked pasta, suggesting that the starchy
212 structure created during the extrusion-cooking was still present in the product, but with even higher
213 wavelength of absorption. It is likely that this structure provides the stability and lower cooking loss
214 (4.2%) observed for this pasta compared to the conventionally processed pasta (15.9%), as reported
215 in a previous publication (Marti et al., 2010).

216 *3.2 Thermal properties*

217 The thermal properties of starch samples are shown in Table 1. Starch from PRS exhibited a
218 peak at 53.7 °C suggesting the presence of recrystallized amylopectin, and a peak at 95 °C
219 indicating the presence of amylose-lipid complexes, as can be expected in parboiled rice (Lamberts
220 et al., 2009). The endothermal peaks shifted to higher temperatures in pasta. The shift of T_p from
221 53.7 °C to 57.7 °C was accompanied by a slight decrease of gelatinization enthalpy from 3.8 to 3.1
222 J/g in pasta made using conventional process. A similar trend has been reported for pasta from
223 semolina, implying starch molecules underwent conformational reorganization as a result of the
224 extrusion step and the drying cycle (Yue et al., 1999). However, no differences were observed for
225 the amylose-lipid complex endotherm in pasta made using conventional process when compared to
226 PRS.

227 Starch in pasta made by using extrusion process (Starch E) exhibited a different endothermal
228 profile with a broad peak starting at 57.2 °C and ending at 96.4 °C. There were two unresolvable
229 melting peaks at 68.7 °C and 83.7 °C; and thus, it was not possible to calculate the endothermal
230 energy (Table 1). Nevertheless, this appears consistent with the data shown in the SEC profile,
231 wherein the amylopectin matrix has a larger molecular size (Figure 1). The higher melting
232 temperature detected in Starch E could highlight a higher thermal stability in the product,
233 confirming previous results (Marti et al., 2010). A similar trend was detected in pasta from semolina
234 and related to the stabilization of amylopectin crystallites (Yue et al., 1999).

Following cooking, the endothermic profile only exhibited a peak at 89 °C and no peak was observed at the lower temperature, for pasta made by using both processes. However Cooked E sample still had a higher end temperature that is consistent with the SEC profile that shows the amylopectin fraction with higher wavelength of absorption.

3.3 Moisture content of equilibrated samples

The moisture content of starch samples equilibrated above saturated solutions of $MgCl_2$, NaCl, and K_2SO_4 is shown in Table 2. The moisture content of the samples were similar when equilibrated above $MgCl_2$ (0.33 a_w) and, as expected, it increased with increasing a_w. Starch C had a lower moisture content compared to PRS and Starch E both at 0.75 and 0.97 a_w. Moisture of the starch from cooked pasta increased; however, no differences were observed between the two cooked pasta samples (Cooked C and Cooked E).

3.4 Color development in iodine exposed starches

The color development (in terms of luminosity, L*) of starch samples after iodine exposure following equilibration above $MgCl_2$ (0.33 a_w), NaCl (0.75 a_w), and K_2SO_4 (0.97 a_w) solutions is shown in Table 2. L* values were not significantly different between the starch samples following equilibration above $MgCl_2$. With increasing moisture content, the L* value decreased for each sample, indicating a darker color. These results are supported by previous studies (Saibene & Seetharaman, 2006; Saibene et al., 2008; Saibene & Seetharaman, 2010; Waduge et al., 2010). Moreover, at higher water activity, the effect of pasta-making process appeared more evident on the L* values of samples. In particular, following equilibration above NaCl solution, L* value increased slightly, reaching higher value in the extrusion-cooking process. An overall increase in L* value was also observed in the cooked samples, regardless of process conditions. A different trend was observed when starch samples were equilibrated above K_2SO_4 (0.97 a_w). Only starch from process C exhibited a higher L* value compared to PRS. A further decrease in L* value was detected after cooking, regardless the extrusion conditions ($p<0.05$).

3.5 Iodine absorption spectra

261 Absorption spectra of starch samples after iodine exposure, presented as a ratio of
262 absorption/scattering (K/S), are shown in Figure 2. At the lowest a_w value (0.33 a_w), all the samples
263 exhibited a low K/S value at all wavelengths (data not shown). Absorption intensities increased for
264 all samples with increasing water activity, as expected (Figure 2a, 2b). This is due to the greater
265 mobility of longer chains with increasing moisture content (Saibene & Seetheraman, 2006). A
266 similar effect was reported previously with potato, corn, and wheat starches (Saibene et al., 2008;
267 Saibene & Seetharaman, 2010; Waduge et al., 2010).
268 After equilibration of samples above NaCl solution, PRS had the highest absorption values,
269 although no distinct peaks are evident, followed by starches extracted from pasta; starches from
270 cooked pasta had the lowest absorption values. This trend is similar to the L* values reported in
271 Table 2; higher the iodine absorption, darker the color and higher the K/S values. Therefore, with
272 increasing the intensity of physical (both thermal and shear) stresses associated with processing,
273 there was a decreasing amount of mobile polymers capable of binding iodine. Furthermore,
274 differences were also observed between starches processed using the two extrusion conditions.
275 Starch from raw pasta (Starch C) and cooked pasta (Cooked C) processed using the conventional
276 process had higher iodine absorption values compared to the respective starches processed using
277 extrusion cooking (Starch E and Cooked E). In particular, the wavelength maxima was 420 nm for
278 starch from pasta made using conventional extrusion, while 480 nm for Starch E, suggesting the
279 chain length of the mobile polymers in starch from conventionally processed pasta was smaller.
280 This is also reflected in the SEC data (Figure 1) wherein Cooked E sample had a higher wavelength
281 of absorption. The different chain mobility of samples is likely related to molecular changes that
282 occurred during pasta-making process.
283 As expected, all starch samples exhibited higher absorption values at 0.97 a_w compared to
284 when equilibrated to 0.75 a_w (Figure 2). This is due to increased plasticization of polymers at the
285 higher moisture contents allowing more polymers to complex with iodine. However, two interesting
286 differences were observed in iodine absorption following equilibration at 0.97 a_w (Fig. 2b) when

compared to samples equilibrated over NaCl. First of all, starch from PRS had iodine absorption values similar to starches from both cooked pasta; while starches from the raw pasta had lower absorption values. Secondly, starch from conventionally processed pasta had lower absorption values compared to starch from extrusion cooked pasta, a switch from what was observed at 0.75 a_w. Cooking the pasta appeared to increase the mobility and amount of polymers available to complex with iodine, likely due to starch gelatinization and the consequent rearrangement of the starch polymers. However, the differences in raw pasta starch are interesting, because it seems that Starch E has higher amount of mobile polymers compared to Starch C. the cooking in boiling water promoted further change in chain mobility, which overcome the differences among uncooked samples.

3.6 X-ray powder diffraction

The X-ray diffractograms of starch samples before and after iodine exposure at 0.75 and 0.97 a_w are shown in Figure 3. Sample PRS showed a typical A-type diffraction pattern with peaks at 15°, 17°, 18°, and 23° 2θ, as reported by previous study (Marti et al., 2010). Pasta samples also exhibited similar peaks, suggesting that the pasta-making process and the extrusion condition did not change the A-type organization of starch polymers. However, in starches from cooked pasta, the peaks at 17° and 18° 2θ disappeared; highlighting a loss in crystallinity as a consequence of starch gelatinization. At a_w value of 0.75, the effect of iodine exposure on crystalline order was evident only in cooked samples. In particular, Cooked E starch exhibited higher V-type peak at 20° θ compared to Cooked C sample. This is consistent with the high presence of longer chains complexing with iodine to form single helical complexes in Cooked E sample compared to Cooked C; as is evident in the SEC profile (Figure 1).

Greater differences among the iodine exposed samples were observed at high a_w values (0.97) (Figure 3b). Iodine did not change the polymorphic pattern of PRS and pasta samples. The same behavior has been previously observed in corn (Cheethaman & Tao, 1998) and wheat starches (Waduge et al., 2010). However, an overall change in X-ray diffraction intensity after iodine

exposure was observed for all starches, suggesting an alteration and disruption of crystallinity of the granules as a consequence of the formation of the iodine-starch complex. After iodination, starch samples exhibited a slight increase in intensity at 7° and 20° 2θ peaks – both related to single helical complexes; and a decrease in peak intensity at 13°, 15°, and 17-18° 2θ (Figure 3b).

4. CONCLUSIONS

The molecular size distribution, the DSC profile, the iodine binding, and the X-ray diffraction analysis elucidated differences in starch organization/architecture in rice flour and pasta samples according to the extrusion conditions adopted for pasta-making. In particular, the extrusion-cooking promoted the formation of a starchy structure characterized by amylopectin fraction with higher wavelength of absorption, broad melting temperature, and high mobility and V-type peak at 20° θ of longer chains complexing with iodine.

During cooking two main phenomena occurred:

- starch gelatinization with loss of crystalline order and increase of both mobility and amount of polymers available to complex with iodine;
- starch polymers rearrangement resulting in high molecular size fractions, with high melting temperature.

Further studies are underway to investigate if the differences in starch structure among the pasta samples could influence starch digestibility and blood glucose response.

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Table 1. Thermal properties of starch samples.

Sample	First peak				Second peak			
	T_o (° C)	T_p (° C)	T_c (° C)	ΔH (J/g)	T_o (° C)	T_p (° C)	T_c (° C)	ΔH (J/g)
PRS	46.3 ± 0.3	53.7 ± 0.0	62.9 ± 1.5	3.8 ± 0.1	76.8 ± 0.0	86.2 ± 0.7	95.0 ± 0.4	2.0 ± 0.2
Starch C	50.3 ± 0.2	57.7 ± 0.5	64.6 ± 0.2	3.1 ± 0.0	77.0 ± 0.5	86.0 ± 0.5	94.3 ± 1.1	2.2 ± 0.2
Starch E	57.2 ± 1.6	68.7 ± 1.4	-	-	-	83.7 ± 1.4	96.4 ± 0.9	2.1 ± 0.2
Cooked C	-	-	-	-	81.0 ± 0.8	89.2 ± 0.2	95.5 ± 1.1	2.1 ± 0.2
Cooked E	-	-	-	-	80.1 ± 0.4	89.0 ± 0.0	108.4 ± 4.8	2.2 ± 0.2

T_o = gelatinization onset temperature
T_p = gelatinization peak temperature
T_c = gelatinization conclusion temperature
ΔH = gelatinization enthalpy

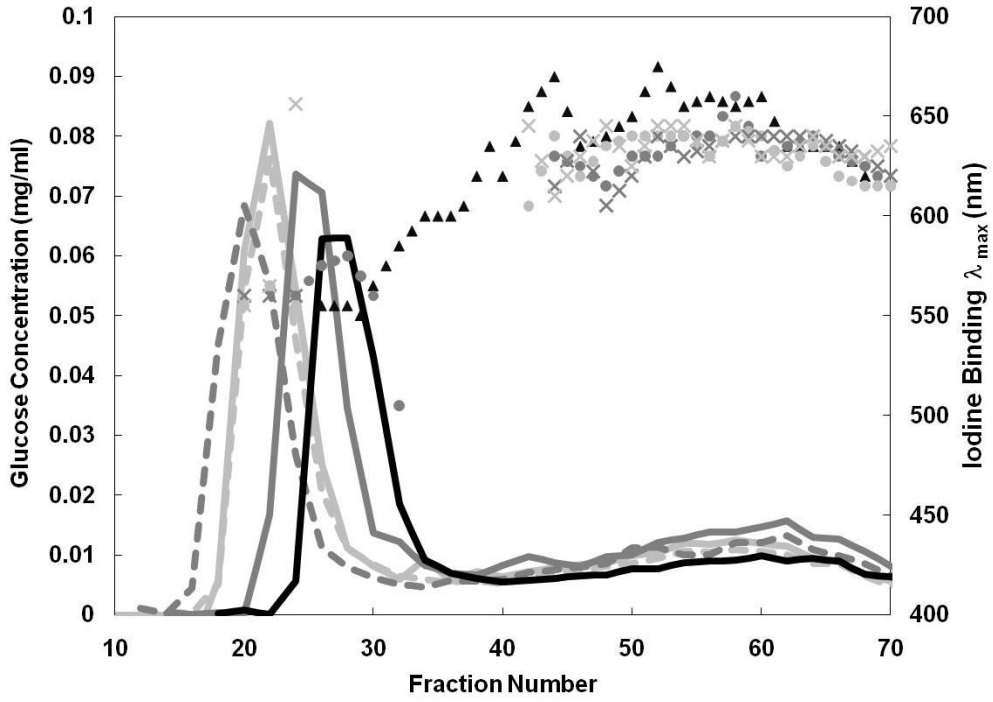
PRS = starch from parboiled rice flour
Starch C= starch from raw pasta prepared by conventional extrusion
Starch E = starch from raw pasta prepared by extrusion-cooking
Cooked C = starch from cooked pasta prepared by conventional extrusion
Cooked E = starch from cooked pasta prepared by extrusion-cooking

Table 2. Moisture content and luminosity (L*) of starch samples, following equilibration at 0.33 a_w ($MgCl_2$), 0.75 a_w (NaCl), and 0.97 a_w (K_2SO_4).

Starch samples	Moisture content (%)			Luminosity (L*)		
	0.33 a_w ($MgCl_2$)	0.75 a_w (NaCl)	0.97 a_w (K_2SO_4)	0.33 a_w ($MgCl_2$)	0.75 a_w (NaCl)	0.97 a_w (K_2SO_4)
PRS	9.7 ± 0.4	11.6 ±0.2	18.2 ± 0.7	46.0 ± 1.3	11.5 ± 0.4	9.2 ± 0.3
Starch C	9.4 ± 0.3	10.6 ± 0.1	17.5 ± 1.2	43.5 ± 1.5	13.4 ± 0.2	12.0 ± 0.1
Starch E	9.3 ± 0.3	12.8 ± 0.1	20.0 ± 0.5	45.6 ± 3.2	14.6 ± 0.2	10.4 ± 0.1
Cooked Starch C	10.3 ± 0.1	13.4 ± 0.2	19.2 ± 1.2	44.9 ± 1.0	16.3 ± 0.3	9.2 ± 0.1
Cooked Starch E	9.8 ± 0.2	13.2 ± 0.3	18.3 ± 1.3	41.1 ± 1.4	19.9 ± 0.3	9.5 ± 0.3

PRS = starch from parboiled rice flour
Starch C= starch from raw pasta prepared by conventional extrusion
Starch E = starch from raw pasta prepared by extrusion-cooking
Cooked C = starch from cooked pasta prepared by conventional extrusion
Cooked E = starch from cooked pasta prepared by extrusion-cooking

Figure 1. Size-exclusion chromatograms of nongranular starch samples.



Total carbohydrate: PRS (black solid line), Starch C (dark grey solid line), Starch E (light grey solid line), Cooked C (dark grey broken line), Cooked E (light grey broken line).

Iodine binding λ_{max}: PRS (▲), Starch C (●), Starch E (●), Cooked C (X), Cooked E (X).

PRS = starch from parboiled rice flour
Starch C= starch from raw pasta prepared by conventional extrusion
Starch E = starch from raw pasta prepared by extrusion-cooking
Cooked C = starch from cooked pasta prepared by conventional extrusion
Cooked E = starch from cooked pasta prepared by extrusion-cooking

462 Figure 2. K/S values of starch samples exposed to iodine vapor following equilibration at 0.75 a_w
463 (a) and 0.97a_w (b).

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466 PRS = starch from parboiled rice flour
467 Starch C= starch from raw pasta prepared by conventional extrusion
468 Starch E = starch from raw pasta prepared by extrusion-cooking
469 Cooked C = starch from cooked pasta prepared by conventional extrusion
470 Cooked E = starch from cooked pasta prepared by extrusion-cooking
471

Figure 3. X-ray powder diffraction spectra of starch samples following equilibration at 0.75 a_w (a) and 0.97 a_w (b). Light color represents the control starch, while dark color represents the iodine exposed starch.

PRS = starch from parboiled rice flour
Starch C= starch from raw pasta prepared by conventional extrusion
Starch E = starch from raw pasta prepared by extrusion-cooking
Cooked C = starch from cooked pasta prepared by conventional extrusion
Cooked E = starch from cooked pasta prepared by extrusion-cooking