



## Changes in properties of starch isolated from whole rice grains with brown, black, and red pericarp after storage at different temperatures



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### ABSTRACT

The aim of this study was to evaluate the physicochemical, morphological, crystallinity, thermal, and pasting properties of starches isolated from rice grains with brown, black, and red pericarp. Starch was isolated from the rice grains at initial storage time, and after 6 months of storage at different storage temperatures (16, 24, 32 and 40 °C). Starch isolated from the grains stored for 6 months at 40 °C showed darker coloration, surface deformation of granules, and a significant reduction in the extraction yield, final viscosity, enthalpy, and crystallinity, independent of the grain pericarp coloration. The time and storage temperature not influence the swelling power and solubility of starch isolated from grains with brown pericarp, while for the grains with black and red pericarp there was reduction in swelling power and solubility of starches isolated of grains stored at 40 °C. Grains stored at 16 °C showed minimum changes in starch properties.

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### 1. Introduction

Rice (*Oryza sativa* L.) is a major source of energy owing to the high concentration of starch, proteins, minerals, and B vitamins. It is the staple food of more than half of the world population, and increasingly popular for its beneficial health properties (Falade & Christopher, 2015; Gunaratne et al., 2013). Brown rice grains have approximately 75% carbohydrates, predominantly starch (Sompong, Siebenhandl-Ehn, Linsberger-Martin, & Berghofer, 2011). In recent years, the interest in rice starch has increased because of its specific characteristics: white color, odorless, easily digestible, hypoallergenic, and small granular form. These characteristics allow multiple applications of starch, both in food and in non-food industries (Ashogbon & Akintayo, 2012).

Rice is a warm climate crop, produced seasonally, and needs to be stored throughout the year to maintain an abundant supply to the processing industries and consumers. The time and temperature of storage are important factors that affect the grain quality. Park, Kim, Park, and Kim (2012) reported an increase in free fatty

acids in polished rice grains that were stored for 4 months at 30 °C and 40 °C, with 16.5% humidity. Likewise, Paraginski, Vanier, Berrios, Oliveira, and Elias (2014) reported a reduction in protein solubility and pH of corn grain stored with 14% humidity, for 12 months at 35 °C. Amino acids and free fatty acids may form complexes with amylose short chain and amylopectin, changing the physicochemical properties and nutritional value of the final products (Hasjim et al., 2010; Salman & Copeland, 2007).

The consumption of foods with nutraceuticals and bioactive properties has gained importance in recent years, specifically that of full pigmented rice grains (red and black pericarp) (Finocchiaro, Ferrari, & Gianinetti, 2010; Sompong et al., 2011). Some studies have shown that phenolic compounds can interact with starch during gelatinization, and change its pasting, thermal, and digestibility properties (Zhu, 2015). However, there are no studies on the effect of storage temperature of whole grains of rice with brown, black and red pericarp and their changes in starch during the storage of grains. In a study of brown rice with husk stored at 38 °C for 9 months with 12.5% moisture content, Patindol, Wang, and Jane (2005) found a reduction in amylose content, length of amylopectin chains, and starch extraction yield. However, starch properties of rice grains with black or red pericarp and their behavior during storage remain to be evaluated. The aim of this study was to investigate the physicochemical, morphological, crystallinity, thermal, and pasting properties of starches isolated from whole

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rice grains with brown, black, and red pericarp stored for 6 months at different storage temperatures (16, 24, 32 and 40 °C).

## 2. Materials and methods

### 2.1. Material and sample preparation

The rice cultivars with brown pericarp (IRGA 417), black pericarp (IAC-600), and red pericarp (MPB-10) were cultivated under an irrigation system in Jaguarão (32° 33' 37" S, 53° 22' 52" W, 23 m), Rio Grande do Sul, Brazil. The rice grains were harvested with an approximate moisture content of 20%, and transported to the Postharvest Laboratory, Industrialization, and Grain Quality Federal University of Pelotas. The grains were cleaned, and dried to a moisture content of 13%. Afterwards, the grains were treated with aluminum phosphide to prevent interference of insects during the experiment. Then, the grains were dehusked using a Zaccaria machine (model PAZ-1-DTA, Zaccaria S/A, São Paulo, Brazil), and conditioned in low-density polyethylene bags with 0.2-mm thickness and 0.9-kg capacity. The processed grains were stored at temperatures of 16 °C, 24 °C, 32 °C, and 40 °C, for 6 months, in triplicate. The grains were covered with aluminum foil to block the light. Samples were collected at 1st day, and after 6 months of storage, for starch isolation and analysis.

### 2.2. Starch isolation

The whole rice grain with brown, black, and red pericarp were milled to reduce the particle 35-mesh (Perten 3100, Perten Instruments, Huddinge, Sweden) and defatted with petroleum ether. The milled and defatted grains were subjected to extraction of starch according to the method described by Wang and Wang (2004), with some modifications. The milled and defatted sample was soaked in 0.18% NaOH in a 1:2 (w/v) ratio for 18 h (4 °C ± 1). After the dispersion was submitted to constant stirring in a blender for 2 min. Then, the resulting material was passed through a 250-mesh screen and centrifuged at 1200g for 5 min at room temperature (25 °C ± 1). The soft top layer was carefully removed, and the underlying starch layer was re-slurried. The starch layer was then washed twice with 0.18% NaOH and centrifuged. The starch layer was washed with distilled water and neutralization with HCl 1 mol L<sup>-1</sup> to pH 6.5 and centrifuged. The neutralized starch was washed with distilled water three times and dried at 40 °C until 10.0 ± 0.5% moisture and milled in laboratory mill (Perten 3100, Perten Instruments, Huddinge, Sweden) to reduce the particle 35-mesh. The starch extraction yield was calculated by the dry starch percentage obtained after the extraction in relation to the amount of grains used for starch extraction.

### 2.3. Amylose content

The amylose content was determined by the colorimetric reaction with iodine, in accordance to the method of Mcgrane, Cornell, and Rix (1998). Starch (20 mg, dry basis) was dissolved in 90% dimethylsulfoxide (8 mL) in 10 mL screw-cap reaction vials. The contents of the vials were vigorously mixed for 20 min and then heated in a water bath (with intermittent shaking) at 85 °C for 15 min. The vials were then cooled to room temperature (25 ± 2 °C), and the content diluted with water to 25 mL in a volumetric flask. The diluted solution (1.0 mL) was mixed with water (40 mL) and 5 mL of I<sub>2</sub>/KI solution (0.0025 mol L<sup>-1</sup> I<sub>2</sub> and 0.0065 mol L<sup>-1</sup> KI) and then adjusted to a final volume of 50 mL. The content was allowed to stand for 15 min at room temperature (25 ± 2 °C), before absorbance measurements at 600 nm. For performing calibration curve used was 20 mg of pure amylose potato

subjected to the same method described for starch, with 0.2–1.0 mL with 0.2 mL interval to determine the absorbance.

### 2.4. Colorimetric profile

The colorimetric profile of the isolated starch was determined using a colorimeter (Minolta, CR-310, Osaka, Japan). The parameters used are the  $L^*$  (100 = white and 0 = black),  $a^*$  (positive = red and negative = green) and  $b^*$  (positive = yellow and negative blue).

### 2.5. Residual protein content

The nitrogen content was determined according AOAC (AOAC, 2006), and the protein content was obtained using a conversion factor of nitrogen to protein of 5.95.

### 2.6. Morphology of the starch granules

A small quantity of each starch sample was spread directly on the surface of the stub and coated with gold (20 nm) using a sputter coater (Desk V, New Jersey, USA). Starch morphology was examined using a scanning electron microscope (Jeol JSM6610LV, New Jersey, USA) at an accelerating voltage of 10 kV. The images were captured at magnifications of 4000 times.

### 2.7. Relative crystallinity

The crystallinity of starches was determined with an X-ray diffractometer (XRD-6000, Shimadzu, Brazil). The scanning region of the diffraction ranged from 5° to 30° with a target voltage of 30 kV, current of 30 mA and scan speed of 1°/min. The relative crystallinity (RC) of the starch granules was calculated as described by Rabek (1980) using the following equation:  $RC (\%) = (Ac/(Ac + Aa)) * 100$ ; where  $Ac$  is the crystalline area; and  $Aa$  is the amorphous area on the X-ray diffractograms.

### 2.8. Thermal properties

The gelatinization characteristics of the starches were determined using differential scanning calorimetry (TA-60WS, Shimadzu, Kyoto, Japan). Starch samples (approximately 2.5 mg on a dry basis) were weighed directly in an aluminum pan (Mettler, ME-27331), and distilled water was added to obtain a starch-water ratio 1:3 (g/g). The pan was hermetically sealed and allowed to equilibrate for 24 h before analysis. An empty pan was used as a reference. The sample pans were then heated from 30 to 130 °C at the rate of 10 °C/min. The onset temperature of gelatinization ( $T_o$ ), peak temperature ( $T_p$ ), conclusion temperature ( $T_c$ ), and gelatinization enthalpy ( $\Delta H$ ) were determined. The range of gelatinization was calculated by subtracting  $T_o$  from  $T_c$ .

### 2.9. Swelling power and solubility

The swelling power and solubility of the starches were determined as described by Adebooye and Singh (2008), with some modifications. Samples (1.0 g) were mixed with 50 mL of distilled water in centrifuge tubes. The suspensions were heated at 90 °C for 30 min. The gelatinized samples were then cooled to room temperature and centrifuged at 1000g for 20 min. The supernatants were dried at 110 °C until a constant weight was achieved so that the soluble fraction could be quantified. Swelling power was represented as the ratio of wet sediment weight to initial dry sample weight (deducting the amount of soluble starch). Solubility was expressed as the percentage of the dried solid weight based on the dry sample weight.

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