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Chemical composition and functional properties of native chestnut starch (*Castanea sativa* Mill)

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ABSTRACT

Starch isolation methods can change their physico-chemical and functional characteristics hindering the establishment of a starch-food functionality relation. A simple high yield and soft isolation method was applied for chestnut (*Castanea sativa* Mill) starch consisting in steeping and fruit disintegration in a 25 mM sodium bisulfite solution and purification by sedimentation. Starch integrity, physico-chemical composition, morphology and functional properties were determined, being observed significant differences from previous described methods for chestnut starch isolation. The X-ray pattern was of B-type, with a degree of crystallinity ranging from 51% to 9%, dependent on the starch moisture content. The onset, peak, and conclusion gelatinization temperatures were 57.1 °C, 61.9 °C and 67.9 °C, respectively. Total amylose content was 26.6%, and there was not found any evidence for lipid complexed amylose. Swelling power at 90 °C was 19 g/g starch, and the amount of leached amylose was 78% of the total amylose content. Native chestnut starch presents a type B pasting profile similar to corn starch but with a lower gelatinization (56.1 °C) and peak viscosity (79.5 °C) temperatures, making native chestnut starch a potential technological alternative to corn starch, especially in application where lower processing temperatures are needed.

1. Introduction

Chestnut kernels (*Castanea sativa* Mill.) are a highly appreciated seasonal nut in the Mediterranean countries. They are mainly consumed fresh, after cooking, with roasting, boiling or frying being the most common cooking methods. Although being a highly perishable product, nowadays chestnuts can be found on the market all around the year due to the availability of frozen and boiled frozen chestnuts. Other important chestnut products are available on the market, among them the high added value and highly appreciated "Marrons Glacés" (Comba, Gay, Piccarolo, & Aimonino, 2009), and chestnut flour obtained by grinding dried chestnuts, used for valorization of small chestnuts or chestnuts with double embryos (Sacchetti, Pinnavaia, Guidolin, & Rosa, 2004). Chestnut flour is then used as a confectionery paste for producing desserts and jams. It is evident that a step of heat treatment of whole chestnuts or chestnut flour is always used before consumption. The heat treatments, like cooking, change considerably the sensorial and nutritional properties of chestnuts, many of these changes being directly or indirectly related with starch gelatinization. For example, chestnut cooking results in large changes in the macromolecular structure of starchy material and these are correlated with changes of digestibility (Pizzoferrato, Rotilio, & Paci, 1999). For cooked chestnuts, besides its sweetness and color, their texture like firmness and elasticity are important quality attributes (Mellano, Beccaro, Bounous, Trasino, & Barrel, 2009). Starch is one of the main components of chestnut kernels (*C. sativa*), accounting for approximately 50% of the chestnut kernel dry matter (Borges, Gonçalves, de Carvalho, Correia, & Silva, 2008; Vasconcelos, Bennett, Rosa, & Ferreira-Cardoso, 2009; Pereira-Lorenzo, Ramos-Cabrer, Díaz-Hernández, Ciordia-Ara, & Rios-Mesa, 2005), so it is expected that the quality attributes and behavior during industrial processing and transformation will be related to the physico-chemical and functional properties of starch from the different chestnut cultivars available, as observed for other starchy foods. For example, the quality of cooked rice is related with its starch chemical composition and properties, with cooked rice with low amylose content being soft and sticky, while rice with high amylose content being firmer and fluffy (Juliano, 1985). Also starch gelatinization during hydrothermal treatment of cassava may play an important role in defining the final characteristics of the cooked product (Beleia, Butarelo, & Silva, 2006).

Chestnut starch has been previously isolated from oven-dried chestnut flours either from *C. sativa* (Correia & Beirão-da-Costa, 2010; Demiate, Oetterer, & Wosiacki, 2001), *Castanea crenata* S. (Yoo, Lee, Kim, & Shin, 2012) and *Castanea mollissima* Bl. (Zhang,

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Chen, & Zhang, 2011), and in this last work, also from freeze-dried chestnut flour. The drying method clearly affected the properties of the Chinese chestnut starches, both in composition, physico-chemical, thermal, pasting, and functional characteristics (Zhang et al., 2011). Furthermore the starch isolated from *C. sativa* flours and characterized, was obtained from an oven dried chestnut flour at 60 °C during approximately 24 h (Correia & Beirão-da-Costa, 2010, 2012; Correia, Leitão, & Beirão-da-Costa, 2009) using different isolation methods (Correia & Beirão-da-Costa, 2010). The drying process at 60 °C changed significantly the chemical composition and functional characteristics of the chestnut starch (Correia & Beirão-da-Costa, 2010, 2012; Correia et al., 2009), as also observed previously by other authors (Attanasio, Cinquanta, Albanese, & Matteo, 2004). The starch chemical composition and functional properties of the chestnut starch previously described are therefore not representative of the native chestnut starch. Moreover the main method used in all purification procedures of chestnut starch has been the alkaline steeping method (Correia & Beirão-da-Costa, 2010; Demiate et al., 2001; Yoo et al., 2012). There are several reported methods for starch isolation and purification in the literature (Liu, 2005, chap. 7). The preferred method for a particular source is dependent on the easiness of releasing the starch granules from the plant cell matrix without damaging their structure, the amount and nature of contaminants present, and the simplicity and economy of the procedure. Although the alkaline stepping method is superior for rice starch isolation due to the presence of insoluble protein (Adoracion, Li, Okita, & Juliano, 1993), it is also known that the alkaline extraction of starch can significantly change the physico-chemical properties of the isolated starch (Cardoso, Putaux, Samios, & da Silveira, 2007; Thys et al., 2008). The objective of this work was to apply a simple and soft isolation and purification method for chestnut (*C. sativa*) starch in order to maintain its native physico-chemical composition, a requirement needed to study native chestnut starch functional properties aiming in the near future to correlate the physico-chemical and functional properties of starch with the quality attributes of processed chestnuts from different cultivars. Functional properties of native chestnut starch were compared with commercial potato and corn starches in order to evaluate potential technological advantages of the native chestnut starch.

2. Materials and methods

2.1. Materials

Chestnut (*C. sativa* Mill var. Longal) fruits were collected from Castanha da Terra Fria, a Protected Designation of Origin region of Portugal. Chestnut fruits harvested at a mature stage, were dehulled, chopped, freeze-dried and then milled. Commercial grade corn (Sigma, S-4126) and potato (Ramazzotti, S. A., Portugal) starches were used as standards when necessary.

2.2. Starch extraction

Chestnut starch was isolated from chestnut flours obtained from freeze-dried chestnuts by steeping in a 25 mM sodium bisulfite aqueous solution followed by sedimentation in water. The chestnut flours (25 g) were steeped in a 25 mM sodium bisulfite solution (250 mL) for 1 h at room temperature and further disintegrated using a Waring blender for 3 min. The starch was separated from cell debris by sedimentation, and the top brown mucilaginous layer was scrapped off the surface. The sedimentation process in water was repeated eight times after re-suspension of the starch slurry in 250 mL of water. The starch precipitate obtained was filtered, washed with water (250 mL) followed by washing with ethanol (250 mL) and dried in the air at room temperature.

2.3. General composition of chestnut starch

Total and damaged starch were determined according to the AACC methods 76.13 and 76-31, respectively, using K-TSTA and K-SDAM kits of Megazyme (Megazyme International Ireland Ltd., Co.). Total amylose and free amylose contents were determined by iodine binding according to Chrasil (1987), with and without lipid extraction with 85% aqueous methanol, respectively. Lipid-complexed amylose was calculated as the difference between total and free amylose content. Nitrogen and phosphorous content of the starch was determined after acid digestion according to the method of Novozamsky, Houba, van Eck, and van Vark (1983), by the methods described by Houba, Novozamsky, and Temminghoff (1994) and Coutinho (1996). Protein content was obtained by multiplying the nitrogen content by 6.25. Water content was determined by oven drying of the starch at 104 °C until constant weight (AOAC, 2000).

2.4. Determination of granule structure and size of chestnut starch by scanning electron microscopy

Chestnut starch morphology was analyzed using the FEI Quanta 400 Scanning Electron Microscope (FEI Company, USA) in environmental mode at 6 mbar using a Large Field Detector (LFD). Starch samples were suspended in water during 24 h with occasional stirring. One drop of the starch suspension was applied on carbon glue and let dry at room temperature. An accelerating voltage of 30 kV was used. The granule size (diameter) was obtained using the Image Tool software Version 3.0 for windows (UTHSCSA, 2002).

2.5. Color evaluation

Chestnut starch color was evaluated with a Chroma Meter CR-300 Minolta (Osaka, Japan). CIE Lab coordinates were obtained using D65 illuminant a 10 observer as reference system. L^*, a^* and b^* parameters were calculated from the average of five color measurements. The equipment was calibrated with a white standard ($L^* = 97.71$; $a^* = -0.59$ and $b^* = 2.31$). For reference, commercial corn (Sigma, S-4126) and potato (Ramazzotti, S. A., Portugal) starches were used. From these values, chroma was calculated using Eq. (1):

$$C^* = \sqrt{a^{*2} + b^{*2}} \quad (1)$$

saturation difference (ΔC^*) between chestnut and reference standards was calculated as $C^*_{\text{chestnut starch}} - C^*_{\text{standard}}$. Hue difference ($\Delta H^*$) was calculated according to Eq. (2):

$$\Delta H^* = \sqrt{(\Delta E^*)^2 - (\Delta L^*)^2 - (\Delta C^*)^2} \quad (2)$$

where ΔE^* is the total color difference calculated according to Eq. (3):

$$\Delta E^* = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (3)$$

2.6. X-ray pattern and relative crystallinity

Powder X-ray diffraction (XRD) data were collected at room temperature with the PANalytical X'Pert Pro diffractometer, equipped with X'Celerator detector and secondary monochromator in $\theta/2\theta$ Bragg–Bentano geometry. The measurements were carried out using a CuKα radiation ($\lambda_{\alpha 1} = 1.54060$ Å and $\lambda_{\alpha 2} = 1.54443$ Å) in a 4–60° 2θ angular range, a step width of 0.017° and a counting time of 100 s/step.