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Optimization of starch isolation from red sorghum using response surface methodology

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A R T I C L E I N F O

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A B S T R A C T

A Wet-milling process for isolation of starch from red sorghum grains was optimized in this paper. A method combining a Plackett-Burman design (PBD), the steepest ascent method, and a Box-Behnken design (BBD) was used to evaluate the effects of major treatment process variables, including the soaking solid-liquid ratio, NaOH concentration, soaking temperature, soaking time, stirring rate, ultrasonic power, milling solid-liquid ratio, milling time and milling rate, on the starch isolation. The response value was a composite value (that is, the sum) of starch yield and sorghum peeling rate. Soaking temperature, soaking time and milling time were significant factors. The conditions of adjusting the temperature of soaking to 50 °C, soaking time to 36 min and milling time to 8 min and 10 s would favor maximum response value obtainment. A mathematical model with high determination coefficient ($R^2 = 0.9825$) was developed and showed good consistency between the experimental values (149.26%) and predicted values (148.87%), which indicated the suitability of the developed model. Starch with high yield (54.58%) and satisfactory color was obtained from red sorghum after optimization.

1. Introduction

Sorghum [*Sorghum bicolor* (L.) Moench] is an annual herb belonging to the sorghum genus of grass family, and is mainly distributed in tropical arid and semi-arid areas of Africa, Asia and the Americas (Beta, Obilana, & Corke, 2001). Sorghum is the world's fifth cereal crop that produced behind only corn, rice, wheat, and barley (Food and Agriculture Organization of the United Nations Statistics Division, 2014), and it is an abundant food staple in underdeveloped countries. Sorghum has been reported to have 570–750 g/kg starch content (Dicko, Gruppen, Traoré, Voragen, & Berkel, 2006; Souilah et al., 2014), and it is an invaluable source of starch for both domestic and industrial uses (Olayinka, Adebowale, & Olu-Owolabi, 2013). Research has shown that sorghum starch contains a higher level of resistant starch than maize starch and other cereals starch (Niba & Hoffman, 2003), which enables its use as a functional ingredient for health benefits, such as preventing and controlling diabetes mellitus (Rooney & Awika, 2005). Therefore, there is tremendous potential for the commercial use of sorghum starch, which is beneficial for promoting economic of countries where sorghum is the principal cereal crops (Easterly & Levine, 2003).

Starch can be isolated using different processes, depending on the source and end use of the starch. Wet-milling procedures have been employed for sorghum starch production (Higiro, Flores, & Seib, 2003), which minimizes the degree of destruction of starch granules and keeps the physicochemical properties of sorghum starch. In general, corn and sorghum starches have similar properties (Watson, 1970). However, sorghum grain available for wet milling tends to be less, resulting in the fact that the application value of sorghum starch has not been taken seriously. The reason is mainly due to the special structure of sorghum and the influence of the components. Sorghum starch is pretty difficult to separate resulting from its combination with highly cross-linked kafirin proteins (Munck, 1995). Additionally, Sorghum cultivars show a wide range of seed colors, these pigments could contaminate starch during wet-milling operations and hindrance their value in food applications (Boudries et al., 2009; Perez Sira & Lares, 2004). For this reason, peeling has been proposed as an effective treatment before wet-milling (Lazaro & Favier, 2001; Yang, Seib, & Yang, 1995). Research

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has been done in order to optimize the isolation methods for sorghum starch (Belhadi, Djabali, Souilah, Yousfi, & Nadjemi, 2013; Beta, Corke, Taylor, & Rooney, 2007; Park, Bean, Wilson, & Schober, 2006; Perez Sira & Lares, 2004). However, most studies have been conducted to just increase starch yield, and less has been done to simultaneously improve starch yield and appearance of isolated sorghum starch; and these methods required a long isolation procedure time.

At present, reports on sorghum starch isolation and characterization have been constrained to small, laboratory-scale conditions. To extended the production of sorghum starch to a pilot plant scale, a starch isolation process must feature a high yield, fine color, low input, and low cost of operation. Hence, the purpose of the present study was to establish optimal technological parameters for isolating starch from sorghum grains, in order to simultaneously enhance a higher yield and improve the appearance of isolated sorghum starch. In a previous work, thirty-two sorghum cultivars grains from National Sorghum Modern Technology Industry System were analyzed for attribution, and the variety suitable for wet-milling has been found. In this study, a method combining a Plackett-Burman design (PBD), the steepest ascent method, and Box-Behnken design (BBD) was used to optimize the production of starch and peeling of sorghum. Our work proposed a simple and quick isolation method of potential use in the sorghum starch manufacturing industry.

2. Materials and methods

2.1. Materials

The samples of red sorghum (Jiliang 1) were obtained from National Sorghum Modern Technology Industry System, Dongying Experimental Field, Jinan. It was harvested in July 2016 and dried under natural condition. The sorghum seeds were cleaned to remove impurities and broken kernels and then stored in a cold chamber at 10 ± 2 °C for experiments. All reagents/chemicals used were of analytical grade.

2.2. Methods

2.2.1. Isolation of sorghum starch

According to the general starch processing technology, the sorghum starch was isolated by wet-milling using the procedure described by Belhadi et al. (2013) with certain modifications. Procedures of starch isolation from sorghum consist of steeping, wet milling, centrifugation, isolation and drying. The weighed sorghum grains (100 ± 0.1 g) were steeped in a certain volume (The solid-liquid ratio (g/mL) was 1:2–1:3. Here, g/mL refers to the ratio of sorghum grains to the solvent.) of sodium hydroxide solution (50–100 g/L), then heated (30–60 °C) and sonicated (80–120 W for Watt) in an ultrasonic container (model AS20500A, Auto Science, Hangzhou, China) for 8–15 min with agitation (50–150 r/min; Electric Mixer JJ1, Jintan, China). Then soaking solution was recovered, and the sorghum grains were washed with a large amount of distilled water until the seed coat was removed. Then, a certain percentage of distilled water (The solid-liquid ratio (g/mL) was 1:5–1:8. Here, g/mL refers to the ratio of sorghum grains to the distilled water.) was added, and the sorghum grains without pericarp were milled by a crusher (High-speed Tissue Crusher DS1, Shanghai, China) for 3–5 min at a speed of 5000–12000 rpm. The significant factors and their optimal values were identified through screening and optimization using a PBD, the steepest ascent method and RSM (BBD). The suspension was allowed to stand for 1 h, and the supernatant was removed; the sediment was centrifuged at 760 × g for 10 min (Yang & Seib, 1996). The gray-colored top protein layer was removed using a spatula. Distilled water was added again to suspend the sediment and the slurry was centrifuged for 10 min. Washing and centrifugation were repeated 5 times until the top starch layer became white. The starch isolate was dried for 24 h at 40 °C in hot air drying oven (Electro-Thermostatic Blast Oven, CIMO, Shanghai, China) (Beta, Rooney, Marovatsanga, & Taylor, 2000). The dried starch was ground and passed through 0.15 mm sieve and stored in a tightly closed container.

2.2.2. Starch chemical characterization

The method of Goñi, Garcia-Alonso, and Saura-Calixto (1997) was used to determine starch content in starch isolate, and the purity of starch isolate was evaluated by total starch analysis. Moisture of starch isolate were determined by a moisture analyser (Kern & Sohn GmbH, Balingen-Frommern, Germany), following the method described by AACC (2000). The yield (X%) of starch was calculated using Eq. (1).

$$X\% = A_g/B_g{*}100 \tag{1}$$

Where, A_g represents dry weight of isolated starch; B_g represents dry weight of sorghum grain.

2.2.3. Starch physical analyses

The color of the isolated starch was measured using a HunterLab ColorFlex (USA). This measurement was quantified by Hunterlab (1958) system given by L, a, and b parameters (Boudries et al., 2009).

2.2.4. Peeling rate detection

Peeling rate (R%) of sorghum seeds was calculated by Eq. (2).

$$R\% = M/N{*}100 \tag{2}$$

Where, M represents the number of seeds whose coat has been removed; N represents the total number of sorghum grains. The number of seeds was determined by counting.

2.2.5. Experiment design

2.2.5.1. Plackett-Burman design (PBD) for screening of significant factors. PBD is a two-level experimental design method that can study up to $K= N-1$ variables (N is generally a multiple of 4) by N experiments. The design is based on the first order model: $Y= \beta_0 + \beta_i x_i$ (Y is the predicted response, β_0 and β_i are constant coefficients, and x_i is a coded independent factor.) (Li et al., 2008; Shen et al., 2014). It can estimate the main effect of the factors with the minimum experiments, and rapidly and effectively screen out the most important factors for further study.

Nine factors (Table 1) influencing the isolation of sorghum starch were investigated in this study. Each factor was tested at two levels with a high level and a low level, coded as −1 and +1, respectively. The factors of selection and their levels were derived from single factor pre-experiments. The Plackett-Burman experimental design and corresponding results were shown in Table 1. Design-Expert 8.0.6 software was used for the experimental design and data analysis to determine the significant factors.

2.2.5.2. Steepest ascent method for determining the optimal range of significant factors. The method of steepest ascent can quickly improve the level of each factor screened by the Plackett–Burman design to the optimal response value, and finally establish an effective response surface fitting equation (Zhu et al., 2012). In this study, a suitable direction of steepest ascent was determined by increasing or decreasing the values of variables according to the response value, and the step size in the direction was determined by the estimated coefficient from PBD results and practical experience. The experimental design of the steepest ascent method and results were shown in Table 3.

2.2.5.3. Response surface methodology (RSM) to optimize the optimal conditions. RSM is a mathematical statistical method to find the optimal conditions in multi-factor systems. It is based on multiple linear regressions that take into account the main, quadratic, and interaction effects in accordance with Eq. (3) (Shen et al., 2014).

$$Y= \beta_0 + \sum \beta_i x_i + \sum \beta_{ii} x_i^2 + \sum_{i=1}^{n} \beta_{ij} x_i x_j + \varepsilon \tag{3}$$