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Preparation and characterization of octenylsuccinylated plantain starch

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ABSTRACT

Plantain starch was esterified with octenylsuccinic anhydride (OSA) at two concentrations (3 and 15% w/w) of OSA. The morphology, granule size distribution, pasting, gelatinization, swelling, and solubility of granules and structural features of the starch polymers were evaluated. Granules of the OSA-modified starches increased in size during cooking more than did the granules of the native starch, and the effect was greater at the higher OSA concentration. Pasting viscosities also increased, but gelatinization and pasting temperatures and enthalpy of gelatinization decreased in the OSA-modified starches. It was concluded that insertion of OS groups effected disorder in the granular structure. Solubility, weight average molar mass, $\overline{M_w}$, and z-average radius of gyration, R_{Gz}, of the amylopectin decreased as the OSA concentration increased, indicating a decrease in molecular size.

1. Introduction

Plantain starch has been reported to be less reactive than normal corn starch during various chemical modifications [1–3,10,11].

Starches with different amylose/amylopectin ratios and granule sizes have been modified by reaction with OSA and the products tested for emulsifying capacity [12]. Results showed that granule size had the greatest impact on the stabilizing capacity of modified intact granules, and that the starch with the smallest granule size (quinoa) had the best emulsifying properties. It was also reported that intact OS quinoa starch granules efficiently stabilized oil droplets by creating Pickering-type emulsions [13–15]. The aim of this study was to prepare octenylsuccinylated (OS) plantain starch with different degrees of substitution (DS) and to determine the physicochemical and molecular characteristics of the products.

2. Materials and methods

2.1. Materials

Unripe plantain (*Musa paradisiaca* L.) were purchased at a local market in Cuautla, Morelos State, Mexico. Starch was isolated according to the procedure of Flores-Gorosquera et al. [16]. 2-Octen-1-ylsuccinic anhydride (OSA) was purchased from Sigma-Aldrich Co. (St. Louis, MO, USA).

2.2. Preparation of OSA-modified starches

Starch was esterified (triplicate) according the procedure of Han and BeMiller [17]. Plantain starch (100 g dry wt) was dispersed in distilled water (225 mL) with agitation. The pH of the slurry was adjusted to 8.5–9.0 with 1 M NaOH. To this mixture, different amounts of OSA (3 and 15.0% w/w) were added, and agitation was continued at room temperature (≈ 25 °C) while the pH was maintained at 8.5. After 6 h, the starch slurry was neutralized to pH 7 with 1 M HCl. The resulting starch-OSA derivative was collected by centrifugation (4200 × *g*) and washed 3 times with distilled water and once with acetone, and air-dried.

2.3. Determination of degree of substitution (DS) and OSA substitution

The DS of OS starches was determined by titration following the methodology proposed by Song et al. [18]. An accurately weighed OS starch sample (5 g, dry weight) was dispersed by stirring 30 min in 25 mL of 2.5 M HCl-isopropanol. Isopropanol–water (9:1 v/v) (100 mL) was added, and the slurry was stirred an additional 10 min. The suspension was filtered through a glass filter,

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and the residue was washed with 90% isopropanol until the washing was free of chloride ion as determined by adding a solution of silver nitrate (0.1 M). The washed starch was re-dispersed in 300 mL of distilled water, and the dispersion was heated in a boiling water bath for 20 min. The starch solution was titrated with 0.1 M standardized NaOH solution, using phenolphthalein as the indicator. An unmodified starch sample was simultaneously titrated as a blank. The DS was calculated utilizing the reported equation.

Additionally, the percentage of carboxyl groups from OS on the starch granules was determined using a titration method reported by Timgren et al. [12].

2.4. Light microscopy

A polarized light microscope (Eclipse 80i, Nikon, Japan) equipped with a 20x objective lens and a digital camera (Digital imaging Head, DC330 camera MTI, Japan) was employed. Dry native starch was sprinkled on a slide, and a cover slip was added to the slide. A drop of deionized water was added to the edge of the cover slip and the images were captured under polarized light. Samples from the starch pasting cell (Section 2.7) were removed when they reached at 60 °C, then they were dispersed in hot distilled water to a final concentration of 0.4 w/w. One drop of diluted pastes was observed by light microscopy.

2.5. Scanning electron microscopy

Starch granules were fixed to conductive tape mounted on a brass disk. The granules were then coated with gold using a Polaron E5100 (Polaron equipment Ltd., Watford, UK). Images of starch granules were captured at 2000× and 2500× using a scanning electronic microscope model JSM-5800LV (JEOL, Tokyo, Japan).

2.6. Granule size distribution

Size distribution of starch granules was determined by laser diffraction (Cilas 1090L, Cilas, France). Uncooked starch granule (250 mg) powders were dispersed in distilled water (100 mL) in the liquid dispersion module with mechanical stirrer at 250 rpm by 2 min and ultrasonic at 25 W. Starch pastes from the starch pasting cell (Section 2.7) were diluted with distilled water at 60 °C to a concentration of 15% (w/w). Then 10 mL samples were taken and dispersed in distilled water (100 mL) with mechanical agitation and ultrasound (as done with uncooked granules) for 2 min. Particle size is expressed as the median diameter D [v, 0.5], which is defined as the diameter for which 50% of particles by volume are larger, and cumulative volume distribution.

2.7. Pasting properties

Native and OS starches were cooked in a stress rheometer (AR-1500ex, TA Instruments, USA) using a starch pasting cell (SPC) with an impeller blade at 500 s^{-1} under the following sequential steps: (a) heating (2.5 °C/min) from 30 to 92 °C; (b) holding at 92 °C for 10 min; (c) cooling (2.5 °C/min) to 30 °C. Starch concentration was 4% on a dry weight basis.

2.8. Gelatinization

Thermal properties were determined using a differential scanning calorimeter (DSC) (Q10, TA Instruments, USA). A mass of 2 mg of starch was placed in an aluminum pan at room temperature (25 °C) and 7 μL (7 mg) of deionized water was added to obtain a moisture content of about 70%. The pans were hermetically sealed, and allowed to stand for 12 h at room temperature for even distribution of water. Sample pans were heated at a rate of 10 °C/min from 20 to 120 °C. An empty pan was used as reference. Onset temperature, peak temperature, conclusion temperature, and enthalpy of gelatinization (ΔH) were recorded.

2.9. Swelling and solubility

Aliquots of starch paste necessary to prepare a dilution containing 0.5% starch at 60 °C were taken from the SPC. Samples were rapidly cooled to 25 °C and 4 mL of the 0.5% dilution were centrifuged at 700 × g for 15 min. A separate 4-mL volume of the same 0.5% dilution was dried in an oven at 100 °C for 24 h to calculate the mass of dry starch. The supernatant of the centrifuged portion was carefully separated from the residue. The total sugar content of the supernatant was determined by reference to a calibration curve (0–50 mg/mL of glucose) using the phenol-sulphuric method [19]. Solubility (%) was calculated using the following equation:

$$\text{solubility}\,(\%) = \left(\frac{m_{SS}}{m_{DS}}\right) \times 100$$

where m_{SS} is the mass of starch in the supernatant (calculated as the product of the concentration of total sugars in the supernatant times the volume of supernatant) and m_{DS} is the mass of dry starch in the aliquot (calculated as the product of the ratio of dry to wet mass of the residue times the volume of dilution assuming a density of 1000 kg/m^3). Swelling was calculated according to equation:

$$\text{swelling}\,(\text{g water/g starch}) = \frac{m_{RH}}{m_R}$$

where m_{RH} is the mass of wet residue and m_R is the mass of dry residue.

2.10. Weight average molar mass and z-average radius of gyration

Starch granule solubilization was performed following the procedure described by Bello-Pérez et al. [20]. Briefly, 20 mg of starch were added into a Teflon cup containing 10 mL of deionized water. The cup was introduced into a polycarbonate vessel (Parr Instrument Co., Moline, IL, USA) and centred inside a microwave oven at maximum power during 1 min (1100 W). Cooling step was carried out by immersion in an ice bath for 30 min. The solution was filtered through a 5 mm nylon membrane (Millipore, Bedford, MA, USA) and immediately injected onto the high-performance size exclusion chromatography (HPSEC) column.

HPSEC analysis combined with multi-angle laser-light scattering (MALLS) (Dawn 8+ Heleos, Wyatt Technology Corporation, CA, USA) and refractive index (Agilent 1100, model G1362A, Agilent Technologies, UK) detectors were used to estimate weight average molar mass ($\bar{M}_w$) and z-average radius of gyration ($\bar{R}_{Gz}$) of the starches. A Shodex OH PAK SB-807 HQ analytical column (Showa Denko K.K., Tokyo, Japan) was used to separate starch molecules. The mobile phase was distilled-deionized water with 0.02% sodium azide; flow rate was 0.3 mL/min. Data obtained from MALLS and RI detectors was analyzed using Astra software (version 5.3.4.14, Wyatt Technology Corporation, CA, USA) to obtain the molecular weight distribution.

3. Results and discussion

3.1. Degree of substitution (DS) and percent OS groups by weight

Reaction with 3% OSA by weight produced a low DS (0.011) whereas reaction with 15% OSA by weight resulted in a higher DS (0.038). The OSA starch treated with 3% of reagent resulted in the maximum allowable level by the USFDA. The weight percent