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Characterization of edible starch–chitosan film and its application in the storage of Mongolian cheese

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ABSTRACT

The physicochemical, mechanical, optical and structural properties based on different amylose content starch–chitosan films with the addition of hydrophilic glycerol and hydrophobic perilla oil were investigated, and the effects of the starch–chitosan coating on the physicochemical and microbial properties of Mongolian cheese were evaluated. The films were formed by casting method. Results showed that the incorporation of perilla oil resulted in a decrease in moisture content, solubility and mechanical properties and an increase in total color difference (ΔE^*). High water vapor permeability (WVP), good transparency and low solubility were observed with the addition of glycerol. Meanwhile, the film based on mung bean starch–chitosan (MSC) exhibited higher moisture content, WVP values, ΔE^* and less transparency than that based on water chestnut starch–chitosan (WSC). The morphology of films was also different based on MSC/WSC. The shelf life extension of Mongolian cheese was evaluated at 8 °C. The results showed that the cheese coated by WSC film containing perilla oil presented better treatment performance in terms of microbial growth delay, weight loss and shelf life length.

1. Introduction

Edible films and coatings are prepared from edible materials, which act as a barrier to the external elements (such as moisture, oil and microorganism) [1,2] and improve the quality of food products. Starch is one of the most commonly used materials to prepare edible films for its low cost, renewability and biodegradability [3]. Mung bean is the seed of Vigna radiate and is traditionally consumed in soup, pancake and cold noodle in China. By heating, the dispersion of mung bean starch becomes a transparent and strong gel with resilience, which is smooth and pliable in texture; and of good cooking quality [4–6]. Water chestnut (*Eleocharis dulcis*) is a fruit crop grown in China, India and Southeast Asia [7] and the cake made from its flour is a traditional dessert in the south of China [8,9]. The mung bean starch differs from water chestnut starch in the amount of amylase and the degree of accessibility of water [10].

Our study focused on the properties of film formation from mung bean/water chestnut starch and chitosan containing glycerol/perilla oil. The shelf life of Mongolian cheese coated by the films was monitored by the microbiological and physicochemical changes of Mongolian cheese after 30 days of storage period.

2. Materials and methods

2.1. Materials and chemicals

Mung bean starch and water chestnut starch were purchased from Jincheng Food (Yantai, China) and Hongyuan Agriculture (Guilin, China), respectively. Mongolian cheese was purchased from a local market in Keshiketeng Banner in Inner Mongolia. Perilla oil was obtained from Yetongren Food (Wenzhou, China). Plate Count Agar (PCA) and Potato Dextrose Agar (PDA) were from Huankai Microbial (Guangdong, China). Hydrochloric acid (HCl), Chitosan (5000 Da), Glycerol ($C_3H_8O_3$), acetic acid ($C_2H_4O_2$), phosphorus pentoxide (P_2O_5) and anhydrous calcium chloride ($CaCl_2$) were all obtained from Sinopharm Chemical (Shanghai, China).

2.2. Film preparation

Chitosan solution (1.3%, w/v) was prepared by dispersing chitosan in 1% (w/w) acetic acid and stirring with a magnetic stirrer (Huxi Analysis, Shanghai, China). After chitosan was dissolved completely, the solution was filtered with cheesecloth. Mung bean starch and water chestnut starch were dispersed in deionized water to obtain 1.5% (w/v) starch solution, heated at 85 °C and 75 °C, respectively, and each kept for 60 min under stirring to accomplish a complete starch gelatinization. Based on preliminary experiments, starch–chitosan solution was prepared by mixing 200 mL

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of starch solution and 200 mL of chitosan solution together. Then 1.5 g glycerol or 1.5 mL perilla oil was added and the solution was stirred for 1 h. Then the mixture was homogenized using a rotor–stator homogenizer (IKA T25-Digital Ultra-Turrax, Staufen, Germany) at 30 k rpm for 60 s. All the film-forming solutions (MSC: mung bean starch–chitosan solution; MSCG: mung bean starch–chitosan solution containing 0.5% (v/v) glycerol; MSCP: mung bean starch–chitosan solution containing 0.5% (v/v) perilla oil; WSC: water chestnut starch–chitosan film; WSCG: water chestnut starch–chitosan solution containing 0.5% (v/v) glycerol; WSCP: water chestnut starch–chitosan solution containing 0.5% (v/v) perilla oil) were prepared and adjusted to pH 4.0. Finally, a vacuum pump was applied for 1 h to remove air bubbles from the systems.

The films were prepared by casting 200 mL gelatinized suspensions on rectangular acrylic plates (25 cm × 25 cm). The starch suspensions were dried (25 °C, RH 50%) in a climatic chamber to constant weight (about 24 h); translucent films which can be easily removed from the plate were obtained. Films were peeled off and conditioned for 10 days at RH 50% before measurements.

2.3. Physicochemical properties

Thickness of the films was determined using a spiral micrometer (Shanghai Measuring & Cutting Tool Works, Shanghai, China) at 10 random spots of the film. The mean standard deviation within the film was about 5% of the average thickness. The moisture content of the films was determined by the gravimetric method, after being dried at 105 °C for 24 h in triplicate. For this study, film solubility (FS) in water was defined as the ratio of the water-soluble dry matter of film that is dissolved after immersion in distilled water. A square film sample was cut from each film, dried at 105 °C for 24 h in a ventilation drying oven, and weighed to determine the initial dry weight ($M_{initial}$). FS was measured from immersion assays in 50 mL of distilled water with constant stirring for 12 h at 25 °C. Then, the remaining pieces of films were taken out and dried at 105 °C until constant weight (final dry weight, M_{final}). The FS was calculated using Eq. (1):

$$FS\% = \frac{M_{initial} - M_{final}}{M_{initial}} \times 100 \quad (1)$$

Labscan XE (Hunterlab, Reston, U.S.A.) was used to determine the change of film color. The CIE Lab scale was used. Total color difference (ΔE^*) and color intensity(c^*) were calculated using Eqs. (2) and (3) [11,12].

$$\Delta E* = [(\Delta L*)^2 + (\Delta a*)^2 + (\Delta b*)^2]^{1/2} \quad (2)$$

$$c* = [(a*)^2 + (b*)^2]^{1/2} \quad (3)$$

The film opacity was determined by measuring the absorbance at 600 nm with a spectrophotometer (UV 1800, Shimadzu Corporation, Japan) and calculated by the following equation.

$$O = \frac{A_{bs600}}{\delta} \quad (4)$$

where O is the opacity, A_{bs600} is the value of absorbance at 600 nm and δ is the film thickness (mm).

2.4. Mechanical properties

Mechanical properties of the films were tested according to the method described by Park and Zhao [13] with some improvements and the tests were carried out using a TA-XT2i texture analyzer (Stable Micro Systems Ltd., UK) with load cell of 50 kg. Films were cut into strips (20 mm × 80 mm) and mounted between the tensile grips. Initial grips separation was 50 mm and cross-head speed was 0.8 mm/s. The machine was capable of maintaining a uniform cross-head speed. Tensile properties for different types of films and treatments were determined with 2 individually prepared films as the replicated experimental units and 5 or 6 sub-samples tested from each film.

The elongation (%) was calculated by dividing the extension-at-break of the specimen by the initial gauge length. Tensile strength was calculated on the basis of the original cross-sectional area of the test specimen using Eq. (5).

$$TS = \frac{F}{A} \quad (5)$$

where TS is the tensile strength (Pa), F is the force (N) at maximum load, and A is the initial cross-sectional area (m^2).

2.5. WVP

WVP was determined gravimetrically at 25 °C, according to the method described by Sothornvit and Talja [14,15] with some modifications. The samples (discs of paper, 70 mm diameter) were placed in permeation cells (inner diameter: 42 mm, height: 25 mm) filled with granular (Φ <2 mm) anhydrous calcium chloride and hermetically stamped on the edges with a mixture of refined paraffin and microcrystalline wax. The stagnant air gap under the films is less than 5.0 mm. The permeation cells were placed in a glass chamber, with saturated sodium chloride solution, providing RH gradients of 0/75% at 25 °C. The permeation cells were weighed at regular time intervals until changes in the weight were recorded to be the nearest 0.001 g. WVP was calculated as follows:

$$WVP = \frac{m\delta}{At\Delta p} \quad (6)$$

where m is the weight of water permeated through the film (g), δ is the thickness of the film (m), A is the permeation area (m^2), t is the time of permeation (s), and Δp is water vapor pressure difference across the film (Pa).

2.6. Scanning electron microscope (SEM)

The films were previously cleaned and dried at 45 °C for 3 days and kept in a desicator. Pieces of 1.0 mm × 5.0 mm were cut from films and then fractured in liquid nitrogen. Prior to observation, the films were coated with a thin conducting layer of gold. Microstructure observations of surface were carried out using a NOVA Nano SEM 230 (FEI, USA) low vacuum ultra-high resolution field emission scanning electron microscope with vCD. The accelerating voltage was 5 kV and the working distance was 5 mm.

2.7. Cheese coating and analysis

The Mongolian cheeses (6.0 cm × 4.0 cm × 5.0 cm) were coated with the selected solution by brushing the surface until all of it was covered. Then the cheeses were left at 8 °C (92% RH) until the coating was dry. Packaged and uncontrolled samples were stored at 8 °C for 30 days for evaluation in terms of several quality factors.

Moisture, weight loss and microbiological analyses were carried out on the cheeses by the 0th, 15th and 30th day after the application of the coating. The moisture content in Mongolian cheese was determined by the gravimetric method, after being dried at 105 °C for 24 h in triplicate. The weight loss was evaluated by weighing at the beginning of the experiment and during the storage period. Microbiological analysis was performed by the determination of the total bacterial (TB) and total fungi (TF) count. In order to do this, a 20 g sample was removed aseptically from a standardized area of the cheese surface, diluted 1:10 (v/v) in sterile 0.9% (w/v) sodium citrate and blended. Appropriate dilutions of the sample