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Surface modification of sodium caseinate films by zein coatings

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ABSTRACT

The objective of this study was to improve the barrier ability of sodium caseinate (SC) films using corn zein via direct coatings and structural inversion approach. The effects of zein coatings on barrier properties (oxygen and water vapor) and surface properties (surface hydrophobicity, surface free energy and surface topography) of the films were extensively investigated. Zein coating via the structural inversion approach heightened surface hydrophobicity of SC films, and polar parameter (γ^{AB}) of surface free energy (γ) decreased from 11.5 to 3.4 mJ/m^2, whilst apolar parameter (γ^{LW}) of γ increased from 25.3 to 32.1 mJ/m^2. In contrast, direct coating of zein presented an opposite pattern. Zein coatings increased surface roughness of SC films, structural inversion approach led to apparent surface irregularities with high irregular projections, whereas the well-defined zein nanospheres were evenly distributed in film surface via direct depositing. Zein coatings produced via structural inversion approach enhanced the barrier of SC films against water vapor and oxygen, whereas the direct coating approach only decreased oxygen permeability (OP) of the films. The present data pointed to a possible arrangement and orientation of hydrophilic and hydrophobic moieties of zein on surface of SC films under different conditions. A schematic illustration of the formation pathway of zein coatings on SC films was proposed to relate it with barrier and surface properties of the films.

1. Introduction

Biopolymer-based films have shown promising potential as a substitute of petroleum-based plastic package films to reduce environmental impact (Mariniello et al., 2003). The characteristics of biopolymer-based films are determined mainly by the chemical nature of the biopolymer. Among the biopolymer-based films, protein-based films are the most attractive due to their impressive gas barrier properties (Ou, Kwok, & Kang, 2004). However, protein-based films display weak gas barrier properties in high relative humidity conditions. Therefore, their application is limited by the high water-vapor permeability (WVP) due to their hydrophilic nature (McHugh & Krochta, 1994; Phan The, Debeaufort, Voilley, & Luu, 2009) and by adding plasticizing agents to obtain stretchable films. In many applications, a better barrier against water vapor is desirable for food packaging since low levels of water activity must be maintained in low-moisture foods to prevent texture degradation and to minimize deteriorative reactions, e.g., microbial growth (Yang & Paulson, 2000). In addition, dry and crispy cereal foods may become soggy and lose their crispness upon water absorption, and soft-textured foods may become hard upon loss of water (Yang & Paulson, 2000).

Sodium caseinate (SC) is a water-soluble polymer obtained by the acid precipitation of casein (Audic & Chaufer, 2005). SC has a satisfactory thermal stability and can easily form films from aqueous solutions, due to its random coil nature and ability to form extensive intermolecular hydrogen, electrostatic and hydrophobic bonds (Arvanitoyannis & Biliaderis, 1998; Janjarasskul & Krochta, 2010). However, as with other protein-based films, the highly hydrophilic nature of SC films limits their moisture-barrier ability as compared with commonly used synthetic-plastic films. One way of improving the moisture-barrier properties of SC films is through including lipidic materials in their formulations, such as fatty acids or essential oils (Fabra, Talens, & Chiralt, 2008; Morillon, Debeaufort, Blond, Capelle, & Voilley, 2002). However, the addition of lipid may weaken gas barrier ability of the films, probably

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due to the formation of channels and pores in the network (Bertan, Fakhouri, Siani, & Grosso, 2005; Morillon et al., 2002; Valenzuela, Abugoch, & Tapia, 2013). On the other hand, oxygen solubility in lipids is higher than correspondent within protein matrix, the addition of lipids promotes the formation of lipid droplets within the film matrix, thus facilitates the oxygen transfer. A lipid can also be laminated on protein matrix to form bi-layer films, but these bi-layer films tend to delaminate over time, develop pinholes or cracks (Quezada Gallo, Debeaufort, Callegarin, & Voilley, 2000). These phenomena may be attributed to the weak adhesion between protein matrix and lipid layer.

Zein, a prolamin, is not soluble in water but soluble in 40–90% ethanol. The water-insoluble characteristic of zein makes it a good candidate for the development of natural biopolymeric colloidal particles or films (Wu, Wen, Yang, Xu, & Yin, 2011; Zhong & Jin, 2009). The formation of zein coating on hydrophilic protein-based films via the specific protein–protein interactions has a promising potential to improve their barrier capability. Zein contains sharply defined hydrophobic and hydrophilic domains at its surface, and is capable of self-assembly (Wang & Padua, 2012; Wang, Wang, Geil, & Padua, 2004). Many applications of zein are related to its ability for self-assembly. Evaporation-induced self-assembly (EISA) results in microspheres, packed spheres, and films, depending on zein concentration and the ethanol–water ratio of the solvent (Wang & Padua, 2012). Zein self-assembly also appears in aqueous ethanol solutions to form different micelle-like structures, depending on the composition of the solvent mixture (Kim & Xu, 2008). At lower than 90% ethanol, zein aggregates form micelle-like structures in which the hydrophilic moiety is oriented toward the solvent medium and hydrophobic moieties clump together (Kim & Xu, 2008). The orientation of each molecule will be reversed when the solvent medium turns hydrophobic, which occurred at around 90% aqueous ethanol (Kim & Xu, 2008; Yamada, Noguchi, & Takahashi, 1996). The orientation of hydrophilic and/or hydrophobic moiety of zein aggregates lies on the condition of solvent medium. The absorption and orientation of zein on surface of SC films may provide SC film with appropriate surface and barrier properties.

The micelle-like structures of zein aggregates and their structural inversion behavior lead us to the development of a new coating technology for surface modification of hydrophilic films. Thus, the objective of this study was to improve barrier ability of sodium caseinate (SC) films using corn zein via direct coatings and structural inversion approach. The effects of zein coatings on barrier properties (oxygen and water vapor permeability) and surface properties (surface hydrophobicity, surface free energy and surface topography) of the films were extensively investigated. In addition, a schematic illustration of the formation pathway of zein coatings on SC films was proposed to illuminate the possible relationship between the arrangement of hydrophilic and/or hydrophobic groups on film surface and some selected physical properties of the films.

2. Materials and methods

2.1. Materials

Zein was purchased from Sigma Chemical Co. (St. Louis, MO, USA). Glycerol was purchased Sinopharm Chemical Reagent Co., Ltd (Beijing, China). Sodium caseinate (food grade) was purchased from Arla Foods, UK. Liquids used for contact angle measurements (e.g., ethylene glycol, α-bromonaphthalene) were purchased from Sigma–Aldrich Co. (St. Louis, MO, USA). All other chemicals used were of analytical grade.

2.2. Preparation of films

Sodium caseinate aqueous solutions with protein concentrations of 5.0% (w/v) were prepared by dispersing sodium caseinate powder in de-ionized water and continuous stirring for 1 h at room temperature. Appropriate amounts of glycerol were added to achieve a glycerol-to-protein ratio of 0.3 g g^{-1}. Following degassing under vacuum, the film-forming dispersion (FFD) was cast onto rimmed, leveled glass plates coated with polyethylene films (Clorox China Co. Ltd., Guangzhou, China). The film thickness was controlled by casting the same solid content (3.25 g) on each plate (18 × 20 cm). The castings were air-dried in a thermostatic and humidistatic chamber [25 ± 1 °C, 50 ± 5% relative humidity (RH)] for 48 h, and then the films were used for surface modification.

2.3. Modification of sodium caseinate films

Precisely, zein (1 g) was dissolved in 20 mL of ethanol/water binary solvent (80:20 v/v) to form a stock solution. Twenty milliliters of zein solutions were poured into the glass plates with the formed SC film with a dimension of 18 × 20 cm, and then the glass plate was shaken slowly to spread zein solutions over the SC films. The structural inversion of micelle-like structure of zein aggregates was induced by increasing the ethanol concentration over 90% by adding anhydrous ethanol under ultrasonic circumstance for about 60 s, and then the zein solutions on the SC films were got rid of. The modified SC films were incubated at room temperature for about 30 s. Finally, the film surface was washed with 10 mL de-ionized water to wipe off the residues, and then air-dried in a thermostatic and humidistatic chamber [25 ± 1 °C, 50 ± 5% relative humidity (RH)] for about 4 h to yield zein coating on SC films using structural inversion approach. Direct formation of zein coating on surface of SC films was performed using above-mentioned procedure, without adding absolute ethanol.

2.4. Film thickness determination

Film thickness was measured with a digital micrometer (TAIHAI apparatus Co. Ltd., Shanghai, China) to the nearest 0.001 cm. Measurements were taken along the length of the specimen five times, and the mean values were used to calculate film tensile strength.

2.5. Contact angle measurements

Contact angles of three liquids, including MilliQ water(W), ethylene glycol(EG), and α-bromonaphthalene(B) on films were measured at 25 °C and 50% RH using an OCA 20 AMP (Dataphiscis Instruments GmbH, Germany). Lifshitz–van Der Waals (γ^{LW}) and Acid–Base (γ^{AB}) of the surface free energy (γ) of four liquids was listed in Table 1. A droplet of liquid (4 μL) was deposited on the film surface with a precision syringe. The drop image was recorded by a video camera, and the profile of the droplet was numerically solved and fitted to LaPlace–Young equation (Stacy, 2009). Ten parallel

Table 1
Values of the Lifshitz–van Der Waals (γ^{LW}) and Acid–Base (γ^{AB}) of the surface free energy (γ) of liquids, in mJ/m^2.

Liquid	γ(mN/m)	γ^{LW}	γ^{AB}
Water	72.8	21.8	51
Ethylene glycol	48	29.76	18.24
α-Bromonaphthalene	44.8	44.0	0.8

Taken from Jańczuk, Białopiotrowicz, Zdziennicka (1999).

measurements were performed for each film. The surface free energy was calculated using Owens, Wendt, Rabel and Kaelble (OWRK) procedure in SCA 20 software.

2.6. Oxygen permeability (OP)

The oxygen permeability (OP) of the films was determined using a VAC-V1 Oxygen Permeation Analyzer (Jinan Languang M & E Technology Co., Ltd, Jinan, ShanDong, China) at 23 °C and 45 ± 3% RH, according to the ASTM D1434 procedure (ASTM, 2009). Film samples with the diameter of 9.7 cm were preconditioned at 25 °C and 58 ± 3% RH in a desiccator containing magnesium nitrate saturated solution [$Mg(NO_3)_2 \cdot 6H_2O$] for at least 2 days prior to analysis, then placed between 2 aluminum masks with a circular area of 38.46 cm^2 to determine oxygen permeability.

2.7. Water vapor permeability

A TSY-H1H Water Vapor Permeability Analyzer (Jinan Languang M & E Technology Co., Ltd, Jinan, ShanDong, China) was employed to measure water vapor permeability (WVP) of films according to the ASTM E96 procedure (ASTM, 1995). Film samples were chosen for the WVP testing based on a lack of physical defects such as cracks, bubbles or pinholes. Film samples with the diameter of 7.5 cm were preconditioned at 25 °C and 58 ± 3% RH in a desiccator containing magnesium nitrate saturated solution [$Mg(NO_3)_2 \cdot 6H_2O$] for at least 2 days prior to analysis.

The WVP (g m^{-1} s^{-1} Pa^{-1}) of films was calculated with the equation

$$\mathrm{WVP} = \Delta m \times L/(A \times \Delta t \times \Delta P) \qquad (3)$$

Where Δm is the weight gain (g) of the glass cups during time Δt (s). L is the thickness of films (m), A is the exposed area of films (33 cm^2). Δp is the partial water vapor pressure (Pa).

2.8. Atomic force microscopy images

Surface topography of films was measured by using atomic force microscopy (AFM) equipped with a Multimode SPM and Nanoscope IIIa controller (Veeco Instruments, NY, USA). The films were cut to 2.5 × 2.5 mm and pasted on the sample stage with a double-sided tape. The microscope was operated in the contact mode, wherein changes in the contact amplitude of the cantilever tip on the surface of the film provide feedback signals for measuring variations in surface topography. All AFM images with a scan size of 10 × 10 µm were acquired from the air side of the films. The surface roughness of the films was calculated on the basis of the root mean square (RMS) deviation from the average height of peaks after subtracting the background using Images were processed by the Digital Nanoscope Software (Version 5.3 or 3, Digital Instruments). The following parameters related with surface roughness were calculated: average roughness (R_a: average of the absolute value of the height deviations from a mean surface), root-mean-square roughness (R_q: root-mean-square average of height deviations taken from the mean data plane) according to the method ASME B46.1 (ASME, 1995).

2.9. Statistical analysis

An analysis of variance (ANOVA) of the data was performed using the SPSS 13.0 statistical analyses system, and a least significant difference (LSD) with a confidence interval of 95% was used to compare the means.

3. Results and discussion

3.1. Surface properties

3.1.1. Surface hydrophobicity

The contact angle (θ) is a variable that determines the wettability of a surface. The surface hydrophobicity of film was evaluated using contact angle upon film surface by sessile drop method. The information given by contact angle measurements can be exploited in a static manner at time 0 s when the drop is just deposited onto the test surface. The contact angles (θ) of three liquids, including MilliQ water, ethylene glycol, and α-bromonaphthalene on film surface (air side) are shown in Table 2. The contact angle is determined by the force balance between adhesive (the forces between liquid and solid) and cohesive (the forces within the liquid). Therefore, a water-wettable surface may indicate its hydrophilic property. The contact angle of water decreased from 62.9° (SC film) to about 24.8° (the direct formation of zein coating) on SC films, on the contrary increased to 76.8° after the formation of zein coatings via structural inversion of micelle-like structure of zein aggregates (Table 2). In fact, air is hydrophobic, leading to the amphipolar protein conformation changes at the air/films interface. As a result, most of the hydrophobic groups of SC are expected to migrate toward the film–air interface. This is the reason why SC film surfaces are slightly hydrophobic ($\theta > 60$). As expected, the contact angle of α-bromonaphthalene (an apolar liquid) presented an opposite trend after the surface modifications. The contact angle change of bipolar liquids indicates a density alteration of polar groups on the film surface, also pointing to the gradual transformation of the films to hydrophilic or hydrophobic surface by using different coating approaches.

Zein aggregates form a micelle-like structure in which the hydrophilic moiety is oriented toward the solvent medium at lower than 90% ethanol (Kim & Xu, 2008). As it has been previously mentioned, some of hydrophobic moieties of zein molecules and SC films may clump together. Concomitantly, hydrophilic moieties of zein aggregates may protrude toward air, with subsequent decrease in surface hydrophobicity (water contact angle) (Table 2). In structural inversion approach, when the absolute ethanol was added the solvent medium turned hydrophobic, the orientation of zein aggregates will be reversed to form micelle-like aggregates with the hydrophobic moiety exposed to the surface and hydrophilic moieties clumped together (Kim & Xu, 2008). Thus, some of hydrophilic part of zein molecules and film surface may clump together via hydrogen bond or electrostatic interaction. As a consequence, the hydrophobic moieties may protrude toward air, with subsequent increase in water-contact angles (Table 2). Similarly, Kim and Xu (2008) reported the absorption properties of zein molecule on hydrophilic (glass spheres) or

Table 2
Contact angle of untreated and zein coated SC films.[a]

Films type	MilliQ water	Ethylene glycol	α-Bromonaphthalene
SC film	62.9 ± 2.7b	59.78 ± 2.7a	40.1 ± 1.7b
Direct coating[b]	24.8 ± 2.5c	66.4 ± 5.9a	58.7 ± 1.1a
Structural inversion approach[c]	76.8 ± 5.0a	61.58 ± 0.7a	34.5 ± 2.2c

Letters (a–c) indicate significant ($P < 0.05$) difference within the same column.
[a] Values are expressed as the means and standard deviation of eight measurements.
[b] Direct coating: direct formation of zein coating on SC film using 5% zein in 80% ethanol (v/v).
[c] Structural inversion approach: the formation of zein coating via structural inversion approach.

hydrophobic surface (toner) could be tuned by adjusting ethanol concentration.

3.1.2. Surface free energy

The surface free energy (γ) of the films was used to further clarify the alteration in surface hydrophobicity of the SC films due to surface modifications. Furthermore, the calculations of Lifshitz–van Der Waals (γ^{LW}) and Acid–Base (γ^{AB}) components of surface free energy (γ) provided more detailed information on the surface properties of the films. Table 3 summarizes the surface free energy components and parameters of the films determined by using three test liquids. Generally, the contact angles for three-liquid systems including two polar and one apolar liquid gave reasonable values of the components and parameters of the surface free energy of a solid (Jánczuk, Chibowski, Bruque, Kerkeb, & Gonzalez-Caballero, 1993). The contact angle values for water, ethylene glycol and α-bromonaphthalene were used to calculate the surface energy characteristics. The γ, γ^{LW} and γ^{AB} of the SC films were 42.5, 18.2 and 24.1 mJ/m^2, respectively. This result was consistent with the fact that SC is an amphiphilic polymer, possessing high polar amino acid content. The alteration of γ, γ^{LW} and γ^{AB} of the films depended on the forming process of zein coating.

In the direct formation of zein coating case, the surface free energy (γ) and its polar component (γ^{AB}) of the films increased from 36.8 and 11.5 to 54.8 and 44.2 mJ/m^2, respectively. Concomitantly, the apolar γ^{LW} decreased from 25.3 to 10.6 mJ/m^2 (Table 3). This phenomenon may be associated with the arrangement or orientation phenomenon of hydrophilic and hydrophobic moieties of zein on the surface of SC films. Some of hydrophobic moieties of zein molecules and SC film surface clumped together via hydrophobic interactions. Concomitantly, the hydrophilic part was oriented toward the solvent medium, and was deposited on surface of SC films after air-drying, resulting in a density increase of polar groups on SC surface, and the increased γ^{AB} parameter.

In structural inversion approach, the surface free energy (γ) and components (γ^{AB}, γ^{LW}) of the films presented a completely different pattern as compared with the direct formation of zein coating. The apolar γ^{AB} decreased from 11.5 (SC films) to 3.4 mJ/m^2, whereas the polar γ^{LW} increased from 25.3 (SC films) to 32.1 mJ/m^2 (Table 3). This result supported the aforementioned observation in surface hydrophobicity section, further pointing out that structural inversion of zein aggregates resulted in the different absorption pattern of zein molecules on the surface of hydrophilic SC films, where hydrophobic moiety may orient toward air after air-drying of coated SC films.

3.2. Film microstructure

The effects of surface modification on the surface characteristics of SC films were evaluated by AFM. Fig. 1A–C shows typical 3D surface topographies of the films, and the corresponding results of roughness parameters, the root-mean-square roughness (R_q) and average roughness (R_a) are shown in Table 4. The 3D plots were obtained for the height of the films with respect to a reference plane. SC films showed a relatively smooth and continuous matrix without pores and cracks and with good structural integrity, similar result was reported by Fabra, Talens, and Chiralt (2009). Surface modification of SC films via zein coatings significantly altered the surface morphology of the films, showing an increased trend in the surface roughness of the films (Fig. 1BC). The change in the surface roughness was quantified by the R_q value, which refers to the average size of peaks and valleys within the interest area. Lower R_q numbers indicate a smoother surface. The alteration of surface roughness of the films depended on the formation approach of zein coatings. Clearly, the direct deposition of zein coatings on SC films resulted in an increase of the R_q from 1.42 ± 0.03 to 12.64 ± 1.06. The R_q of the films increased up to 34.54 ± 3.10 when zein coatings were formed by structural inversion of zein aggregates (Table 4), and topographical images showed that these films had the highest projections. The average roughness (R_a) presented a similar trend (Table 4). Conventionally, surface roughness and low surface energy material are the two key factors in determining the surface hydrophobicity (related to water contact angle values) of the film (Feng et al., 2002). The data of surface roughness of the films were consistent with the results of surface hydrophobicity of the films, determined by initial contact angles.

Fig. 1a–c shows the 2D AFM images (top view) of the films. SC films did not show any characteristic structure. We found that the surface structure of SC films can be altered by the use of different surface modification techniques. The well-defined structures on the surface of SC films occurred after the direct formation zein coating, where the individual nanospheres with particle size of 0.5–1.0 μm were distributed in films matrix. The zein nanospheres across the film surface spread sparsely and evenly, and their heights found to be 0–34 nm by the use of "sectional analysis" function of the AFM software (Fig. 2B).

When zein coating was formed via structural inversion of micelle-like zein aggregates, the modified SC films showed apparent surface irregularities. Microsphere-like structures in the range of 1–5 μm were observed in top view of AFM images, and the height of peaks and deepness of valleys were up to 67.6 nm and 45 nm, respectively, according to the "sectional analysis" of the AFM software (Fig. 2C). Generally, the surface of zein freestanding films bears many cracks, voids, and wrinkles reported by Shi, Yu, Rao, and Lee (2012). In contrast, zein coating produced by directing coating or structural inversion approach exhibited a different pattern. This phenomenon may be associated with the arrangement or orientation of hydrophilic and hydrophobic moieties of zein molecules on the surface of SC films, further pointed out that distinct zein coating on SC films could be formed by zein self-assembly strategy through modulating solvent composition.

Table 3
Surface free energy (γ) and its Lifshitz–van Der Waals (γ^{LW}) and Acid–Base (γ^{AB}) of untreated and zein coated SC films (mJ/m^2).

Films type	Surface free energy		
	γ	γ^{LW}	γ^{AB}
SC film	36.8	25.3	11.5
Direct coating[a]	54.8	10.6	44.2
Structural inversion approach[b]	39.8	32.1	3.4

[a] Direct coating: direct formation of zein coating on SC film using 5% zein in 80% ethanol (v/v).
[b] Structural inversion approach: the formation of zein coating via structural inversion approach.

3.3. Barrier properties

This section deals with the evaluation of oxygen and water vapor properties of SC films modified by two coating approaches.

3.3.1. Water vapor permeability

The WVP of the films is shown in Table 5. The WVP of SC films was $5.12 \pm 0.25 \times 10^{-10}$ g m^{-2} s^{-1} Pa^{-1}. It was comparable with the result reported by Li, Yin, Yang, Tang, and Wei (2012), where the WVP was approximately 2.4 g mm m^{-2} h^{-1} kpa^{-1} (6.67×10^{-10} g m^{-2} s^{-1} Pa^{-1}). The direct formation of zein coatings did not significantly enhance water vapor barrier the films, whereas

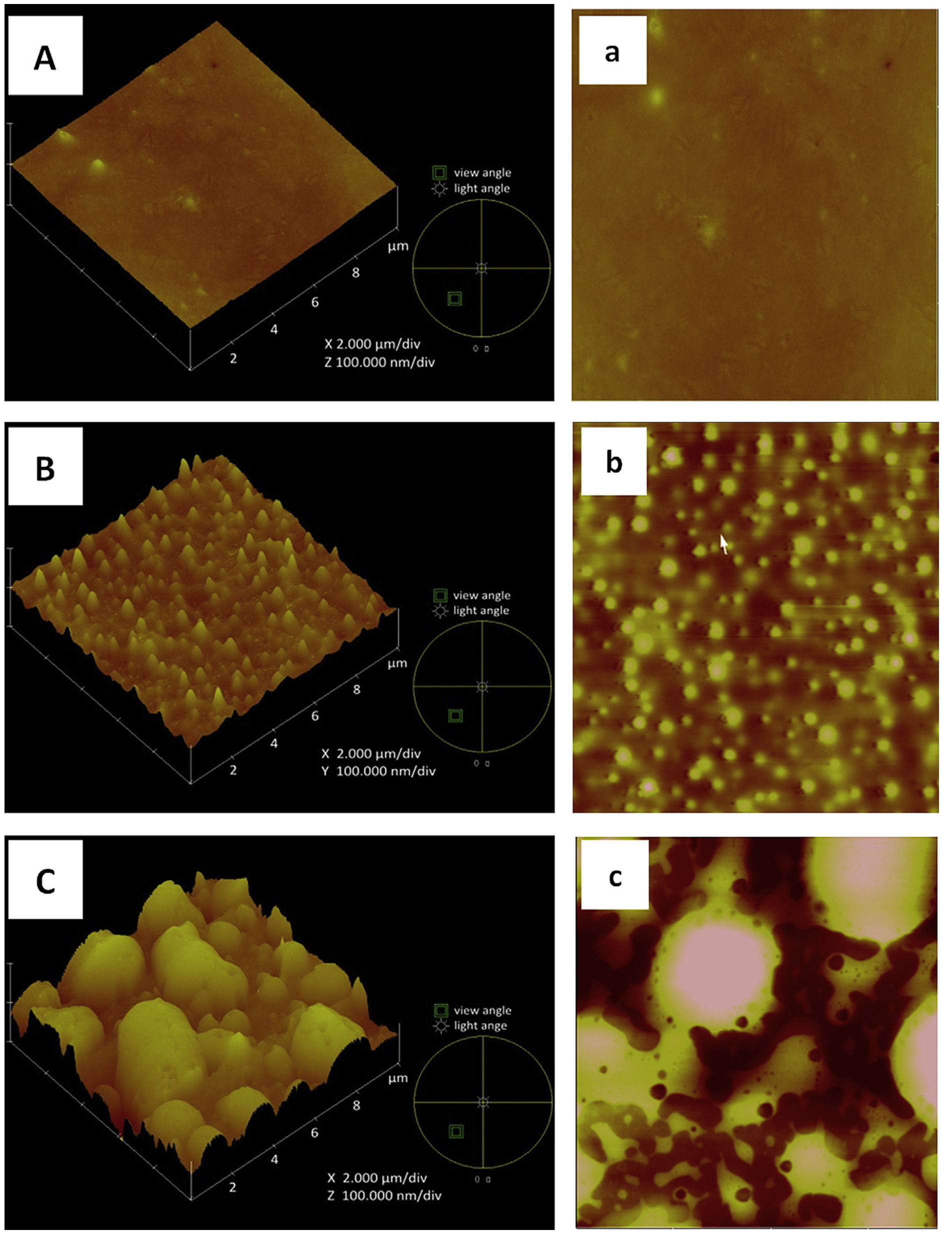

Fig. 1. 3D AFM view (lefthand of) 2D AFM view (right) SC films modified by corn zein via direct coating and structural inversion approach. Panel A & a: SC films. Panel B & b: the direct coating of zein. Panel C & c: zein coating via structural inversion approach. Scan size = 10 μm, data scale = 100 nm.

the WVP of the films was decreased by 22% from $5.12 \pm 0.25 \times 10^{-10}$ to $3.95 \pm 0.50 \times 10^{-10}$ g m^{-2} s^{-1} Pa^{-1} when zein coatings were formed via structural inversion approach (Table 5).

This response of the WVP toward to surface coating approaches was consistent with the surface hydrophobicity and surface free energy analysis of the films. In fact, Zein coating via the structural inversion approach heightened surface hydrophobicity of SC films, and the γ^{AB} of the films decreased from 11.5 to 3.4 mJ/m^2, whilst the apolar γ^{LW} increased from 25.3 (SC films) to 32.1 mJ/m^2, reflecting a density decrease of polar groups on SC surface after. On the other hand, the direct coating resulted in a slight density increase in polar groups on SC surface, which may contribute to the unaffected WVP of SC films by this approach. This result further confirmed that the arrangement of hydrophobic moiety of zein molecule on the surface of SC films play a crucial role in restricting the migration of water vapor across the films.

3.3.2. Oxygen permeability (OP)

Reducing the oxygen transmission rate in food packaging is important because it is related to the decrease in the development of off-flavors, off-odors and nutritional loss associated with oxidation in foodstuffs (Ozdemir & Floros, 2004). Table 5 shows the oxygen permeability (OP) values of the films. Both surface modified approaches presented a slight improvement in barrier property of SC films against oxygen, the OP value of the films decreased by 46% from 1.73 to about 0.94 cm^3 m^{-2} d^{-1} Pa^{-1}. Such result is expected, corn zein films are excellent oxygen barriers due to the presence of polar interactions in their structure (Padua & Wang, 2002). Oxygen permeability of corn zein is far lower than that of polyolefins such as PP and PE, and is comparable to the well-known oxygen barrier materials, e.g., EVOH and PVDC (Miller & Krochta, 1997; Padua & Wang, 2002; Tihminlioglu, Atik, & Öen, 2010). Several studies showed significant decreases in oxygen permeability with corn zein

Table 4
Roughness parameters obtained from AFM analysis of untreated and zein coated SC films.[a]

Film types	R_q (nm)[d]	R_a(nm)[e]
SC films	1.42 ± 0.03c	1.08 ± 0.21c
Direct coating[b]	12.64 ± 1.06b	9.39 ± 0.55b
Structural inversion approach[c]	34.54 ± 3.10a	27.78 ± 2.12a

Letters (a–c) indicate significant ($P < 0.05$) difference within the same column.
[a] Values are expressed as the means and standard deviation of three measurements.
[b] Direct coating: direct formation of zein coating on SC film using 5% zein in 80% ethanol (v/v).
[c] Structural inversion approach: the formation of zein coating via structural inversion approach.
[d] R_q, root-mean-square roughness.
[e] R_a, average roughness.

Table 5
Water vapor permeability and oxygen permeability of untreated and zein coated SC films.[a]

Films type	WVP × 10^{-10} (g m^{-2} s^{-1} Pa^{-1})	OP (cm^3 m^{-2} d^{-1} Pa^{-1})
SC film	5.12 ± 0.25a	1.73 ± 0.19a
Zein coatings[b]	4.75 ± 0.66a	0.91 ± 0.11b
Inversion plus water wash[c]	3.95 ± 0.50b	0.94 ± 0.11b

Letters (a–b) indicate significant ($P < 0.05$) difference within the same column.
[a] Values are expressed as the means and standard deviation of three measurements.
[b] Direct coating: direct formation of zein coating on SC film using 5% zein in 80% ethanol (v/v).
[c] Structural inversion approach: the formation of zein coating via structural inversion approach.

coatings on PP and PE films (Tihminlioglu et al., 2010). Thus, zein coating approaches enhanced the oxygen barrier efficiency of SC films, make them as a potential oxygen-barrier food packaging.

3.4. Schematic illustration

Zein coating via the structural inversion approach heightened surface hydrophobicity of SC films, and the polar γ^{AB} of surface free energy (γ) decreased from 11.5 to 3.4 mJ/m^2, whereas the apolar γ^{AB} of γ increased from 25.3 (SC films) to 32.1 mJ/m^2. In contrast, the direct formation of zein coating presented an opposite trend (Tables 1 and 3). The well-defined zein nanospheres with particle size of 0.5–1.0 μm were evenly distributed in film surface via direct coating approach, whereas topographical images showed that zein coating produced via structural inversion approach presented apparent surface irregularities, with high projections about 1–5 μm in diameter (Figs. 1 and 2). These phenomena pointed to a possible arrangement and orientation of hydrophilic and hydrophobic moiety of zein aggregates on surface of SC films when the medium was changed.

Herein, a schematic illustration of the formation pathway of zein coatings on SC films was proposed to illuminate the possible relationship between the arrangement and/or orientation of hydrophilic and hydrophobic groups and some selected physical properties of the films (Fig. 3). SC contains globally a great amount of hydrophilic groups but it also possesses hydrophobic groups, especially β- and *k*-caseins are amphiphilic (Horne, 1998). Because air is hydrophobic most of the hydrophobic groups are expected to migrate toward the film/air interface. Thus, coexistence phenomena of hydrophilic and hydrophobic groups occurred on the surface of SC films (Fig. 3A). Zein has unusual amino acid sequence, containing over 50% hydrophobic residues (Pomes, 1971). Matsushima, Danno, Takezawa, and Izumi (1997) proposed a prism structural model, in which 9–10 helical segments are aligned in an

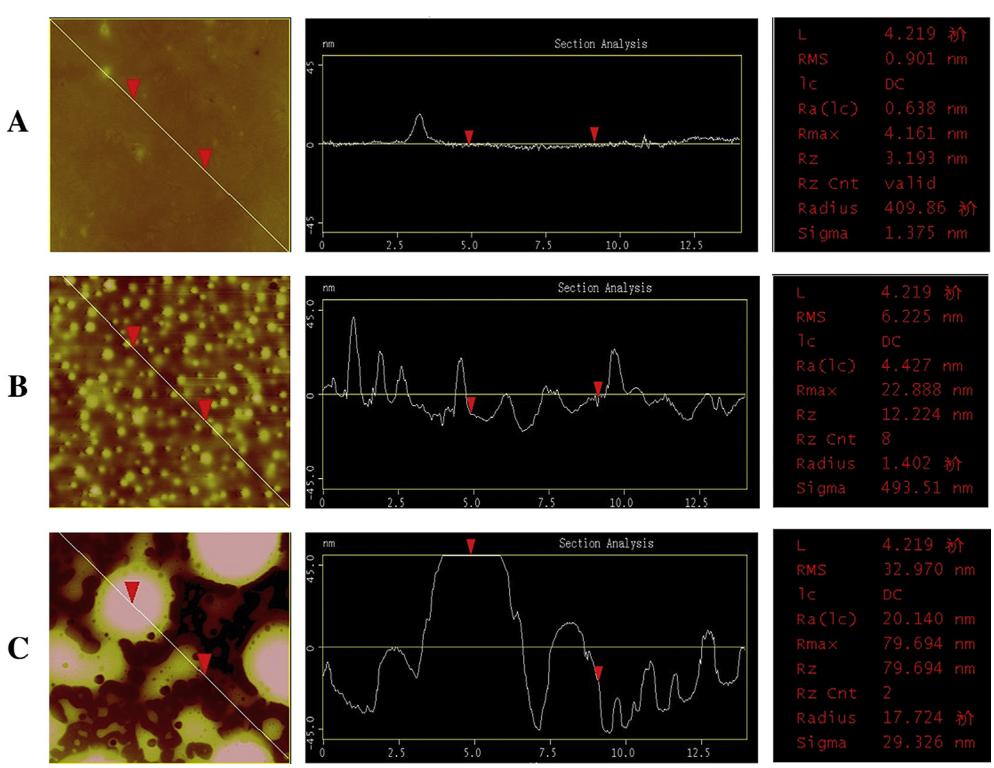

Fig. 2. 10 μm scan of surface modified and untreated SC films, top view (left image), section analysis (middle) and section analysis results (right image). Panel A: SC films. Panel B: the direct coating of zein. Panel C: zein coating via structural inversion approach.

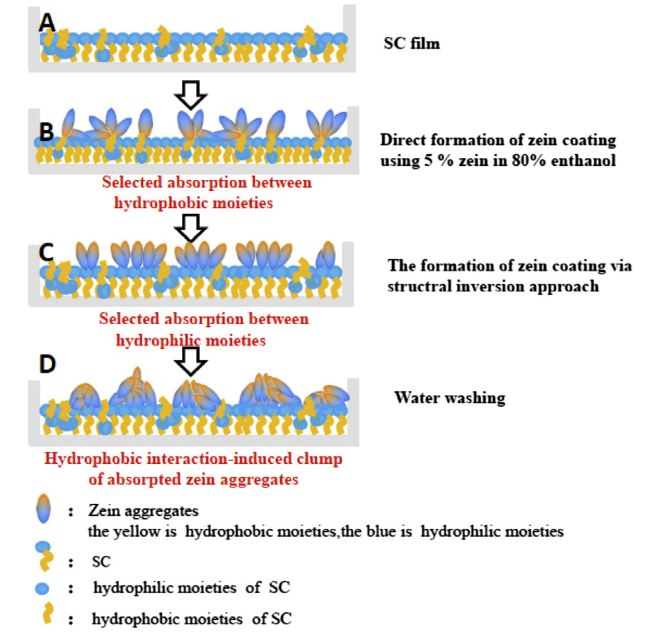

Fig. 3. A schematic illustration of formation pathway of zein coatings on SC films.

antiparallel fashion, and helices are connected at each end by glutamine-rich bridges. According to this model, the sides of the prism formed by the outer surface of the helices are hydrophobic, while the top and bottom surfaces of the prism containing the glutamine-rich bridges are hydrophilic (Lai, Geil, & Padua, 1999). Zein has an amphiphilic character due to sharply defined hydrophobic and hydrophilic domains at its surface (Wang et al., 2004).

Amphiphilicity is one of the main driving forces for zein self-assembly (Löwik & van Hest, 2004). Zein aggregates form a micelle-like structure in which the hydrophilic moiety is oriented toward the solvent medium at 80% ethanol. Some of hydrophobic moieties of zein molecules and SC film surface may clump together. As a result, the hydrophobic sites on the surface of the films may be masked, and the hydrophilic moieties of zein aggregates may protrude toward air, resulting in zein nanospheres on SC surface of SC films (Figs. 1B and 3B). Thus, the direct deposition of zein resulted in the decrease in surface hydrophobicity and apolar Lifshitz–van Der Waals (γ^{LW}) for the films, as well as the increase in polar Acid–Basic γ^{AB}. In structural inversion approach case, the orientation of zein aggregates will be reversed to form micelle-like aggregates with the hydrophobic moiety exposed to the surface and hydrophilic moieties clumped together when the absolute ethanol was added the solvent medium turns hydrophobic. Thus, some of hydrophilic moieties of zein molecules and film surface may clump together via hydrogen bond or electrostatic interaction. As a consequence, the hydrophobic moieties may protrude toward air (Fig. 3C). Water washing induced the hydrophobic interaction between hydrophobic moieties formed on the surface of films, making protruded hydrophobic moiety aggregate to form irregular peak and valley structures, the size and height of the peaks increased accordingly (Fig. 1Cc and Fig. 3D), resulting in increased surface roughness and surface hydrophobicity of the films. As a consequence, the polar γ^{AB} component of the films decreased from 11.5 to 3.4 mJ/m^2, whereas the apolar γ^{AB} increased from 25.3 (SC films) to 32.1 mJ/m^2 (Table 3). The present result indicated that the changes in barrier properties are moderate in spite of a significant change in the film surface properties (evaluated by static and initial contact angle measurement). This phenomenon may be associated with the fact that the transport phenomena, including both sorption and diffusion mechanisms are not only related to the surface of the films. The nature of polymer matrix may play a more important role in barrier properties of biopolymer films.

4. Conclusions

A novel surface modification strategy has been proposed to improve the barrier properties of sodium caseinate films by using amphiphilic zein. The arrangement and orientation of hydrophilic and/or hydrophobic moieties of zein aggregates on surface of SC films was achieved by tuning solvent medium of zein. Zein coating via the structural inversion approach produced more hydrophobic films, while direct deposition of zein produced more hydrophilic films. This phenomenon was supported by surface free energy data. Two coating approaches produced different surface morphology of the films, structural inversion approach lead to apparent surface irregularities of the films with high irregular projections, whereas the well-defined zein nanospheres were evenly distributed in film surface via direct deposition. Zein coatings produced via structural inversion approach enhanced the barrier of SC films against water vapor and oxygen whereas the direct coating approach only decreased oxygen permeability (OP) of the films. A schematic illustration of the formation pathway of zein coatings on SC films was proposed to relate it with barrier and surface properties of the films.

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