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Structural and rheological properties of xanthan gum/lysozyme system induced by in situ acidification

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ABSTRACT

Structural and rheological properties of xanthan gum/lysozyme (XG/Ly) system induced by electrostatic interaction through in situ acidification were investigated. The two biopolymers transited from co-solubility state to form soluble complexes, and finally produced tenuous network as the pH further decreased. The fluorescent images indicated that the network was cross-linked of XG chains by Ly, liking a polymerization process which resulted in the sol-gel transition by electrostatic interactions. High Ly content could accelerate the phase transition at the same pH condition, while XG played a contrary role. XG addition could enhance the thermal stability of Ly. The phase transition was also illustrated by ζ-potential at different pHs. The boundary parameters were determined to distinguish the phase transition regions. At the pH higher than pH_{φ}, the negatively liked XG and Ly were in co-soluble state. They formed soluble complexes at the pH between pH_{φ} and pH_g, and gel was obtained with net microstructure as the pH continuously decreased (lower than pH_g). The paper provides practical parameters that may be applicable in controlling the structure, texture, and stability of polysaccharide/protein system, as well as in food and medicinal application with various purposes.

1. Introduction

Proteins and polysaccharides, two kind of essential biopolymers, are widely used in food, cosmetic and other industries. Their simultaneous use commonly occurs in food products (Schmitt & Turgeon, 2011; Zeeb, Zhang, Gibis, Fischer, & Weiss, 2013). Controlling and regulating the phase transition of proteins/polysaccharides systems is always considered as a crucial issue in food field which dominantly relates the structural and textural properties of food products (Gupta, Bohidar, & Aswal, 2007; Scholten, Moschakis, & Biliaderis, 2014). Both proteins and polysaccharides are so sensitive to the changes of external stress that their phase transition is easy to happen and hard to control (Dong et al., 2015; Hanazawa & Murray, 2013; Jones & McClements, 2011). Either segregative or associative phase transition depends mainly on the electrical charges of the associated biopolymers, and therefore on the factors of the ionic strength and pH which are also closely concerned in food industry (Jara, Sánchez, Patino, & Pilosof, 2014; Li et al., 2012; Thongkaew, Hinrichs, Gibis, & Weiss, 2015). There is therefore a need to understand the phase transition behavior of protein/polysaccharide mixtures to serve food processing, new food design and the development of novel biomaterials that we desired.

Phase transition of many kinds of proteins/polysaccharides systems suffered from external changes has been widely studied. Heated whey protein/pectin soluble complex could be formed by heating protein and pectin together at pH 7.0 and 85 °C for 30 min (Zhang, Hsieh, & Vardhanabhuti, 2014). The complexation of chitosan and soy protein fractions (glycinin and β-conglycinin) has been widely explored under different pH, mixing ratio, heat treatment (Yuan, Wan, Yang, & Yin, 2014). The Influence of low-methoxyl pectin on mechanical behavior and microstructure of bovine serum albumin gels was evaluated (Donato, Garnier, Novales, Durand, & Doublier, 2005). However, the successive phase transition driven by electrostatic interaction without thermal, enzymatic or any other denaturing treatment is rarely reported.

Here, the phase transition of mixed solutions of the globular protein lysozyme (Ly) and xanthan gum (XG) was reported. Both biopolymers are extensively used in food and medicinal application because of their natural occurrence. XG, is an extracellular anionic bacterial polysaccharide, consisted of β-1, 4-linked D-glucose backbone, substituted alternately with a trisaccharide side chain linked to every second glucose residue (Mao, Klinthong, Zeng, & Chen, 2012; Xu et al., 2014). It is widely used in the food industry as a stabilizer and thickener of food products (Toniazzo et al., 2014). Ly, the main protein component

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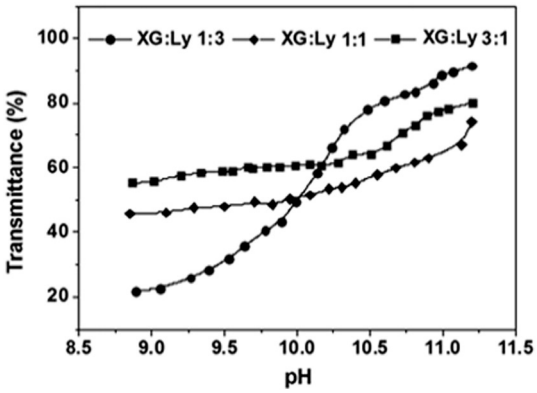

Fig. 1. Transmittance of XG/Ly system with different ratios during acidification process.

of the egg white fraction, has a molar mass of 14.3 kDa and has been received attentions for its use as a kind of food preservative (Vocadlo, Davies, Laine, & Withers, 2001; Zhang et al., 2015). Since now, no report about the phase transition of XG/Ly system has been found.

In our past work, we have studied the phase behavior and self-assembly behavior of XG/Ly system after heat treatment (Xu et al., 2015; Xu, Jin, Hu & Li, 2014). But in the present work, the phase transition of XG/Ly system induced by in situ acidification at room temperature was investigated. The spontaneous changes were monitored by transmittance, ζ-potential, rheological behavior and micro-structure. The transition from co-solubility to soluble complex, and finally to net structure was captured at different pH stage. Finally, the transition process was proposed to determine the boundary parameters to distinguish the phase transition regions. The attempt may be applicable to regulate the phase transition of these protein/polysaccharide systems, and to design desired structures, stability and texture of new food and cosmetic products with various purposes.

2. Materials and methods

2.1. Materials

Xanthan gum (XG) was purchased from Shanghai source biological technology co., LTD. Lysozyme (Ly, Mw = 14.3 kDa) from chicken egg white was obtained from Sinopharm Chemical Reagent co., LTD. Other chemicals were reagent grade and used without purification. All the solutions used in the experiments were prepared using ultrapure water through a Millipore (Millipore, Milford, MA, USA) Milli-Q water purification system.

2.2. Sample preparation

Both XG and Ly solutions (1.0 mg/mL) were stirred with purified water used magnetic stirrer (Ika-Labor-techink, Germany) at room temperature for 6 h and 2 h, respectively. Before XG/Ly mixtures preparation, the initial pH of both the biopolymers solutions was regulated to 11.2 using 1 mM HCl and 1 mM NaOH. The three XG/Ly ratios taken into consideration were: 3:1, 1:1 and 1:3. The amount of glucono-δ-lactone (GDL) used in each sample finally reached 0.008% (w/v). The pH of the XG/Ly mixture was continuously monitored using a FiveEasy Plus pH meter (Mettler Toledo, Germany).

2.3. Transmittance measurements

Transmittance of the XG/Ly system was measured using 722E visible-infrared spectrometer at 600 nm against a distilled water blank as the function of time after GDL was added as 25 °C (Hosseini, Rezaei,

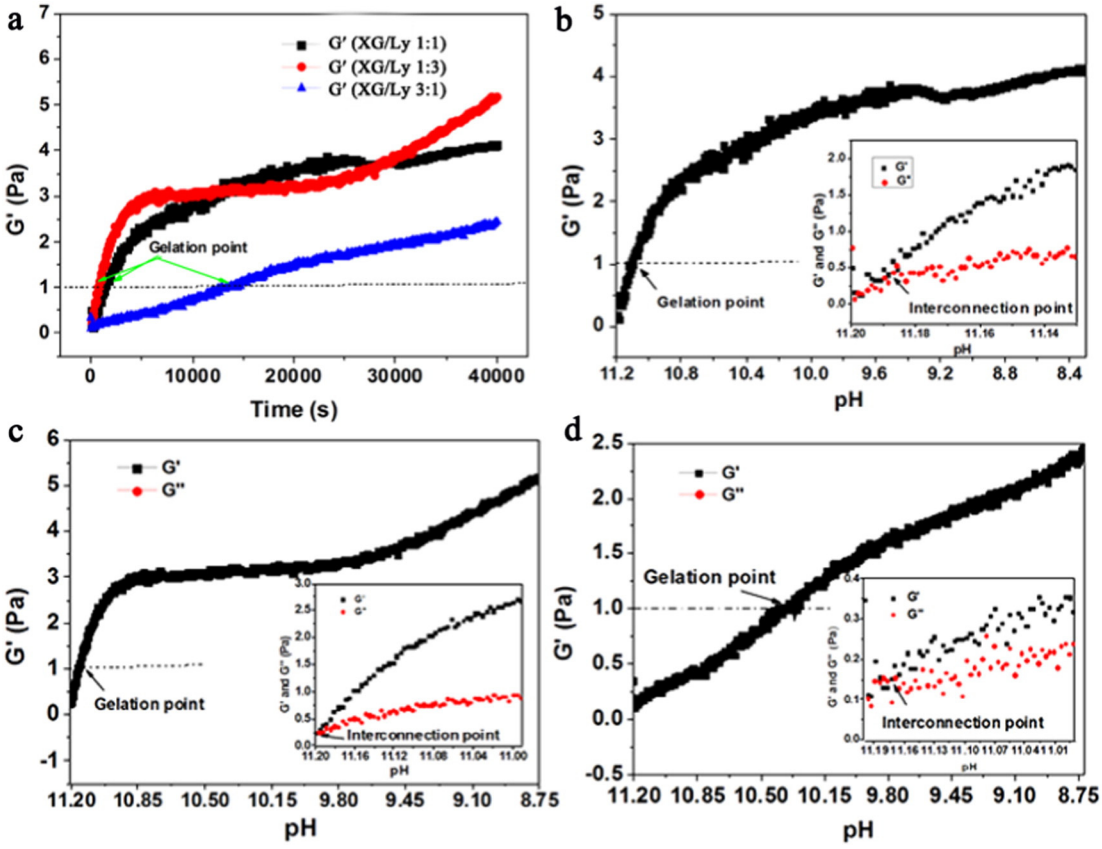

Fig. 2. Evolution of storage modulus as a function of time (a), and storage modulus and loss modulus as a function of pH of (b, c, d) for XG/Ly mixture with XG/Ly ratio of 3:1 (b), 1:1 (c), and 1:3 (d).