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Phase equilibria in water–protein–polysaccharide systems

III. Water-soy bean globulins-polysaccharide systems

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With 18 figures and 3 tables

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1. Introduction

It has been shown earlier (1, 2) that incompatibility of proteins and polysaccharides in aqueous media is common phenomenon. Proteins and polysaccharides are incompatible only under conditions preventing from formation of protein-polysaccharide complexes. Thus pH and ionic strength affect strongly thermodynamic stability of water-protein-polysaccharide systems (1, 3). Conditions of incompatibility of protein and polysaccharides (pH and ionic strength of the system) differ depending on physico-chemical and molecular properties of the chosen pairs of the biopolymers (1–5). Examination of main factors affecting incompatibility of casein with acidic and neutral polysaccharides (1, 3) pointed to identify of conditions for destabilization and separation of water-casein-acidic polysaccharide and water-casein-neutral polysaccharide systems under conditions favorable for self-association of the protein. It permitted conclusion that the connection between self-association of biopolymers and their incompatibility is quite common. Phenomenological studies on incompatibility of protein and polysaccharides are not only of theoretical interest but also quite important for processing proteins in food production (6–9).

In this work we present the results of studies on thermodynamic incompatibility of soy bean globulins (SBG) with acidic and neutral polysaccharides (APS and NPS respectively) in aqueous media. Polysaccharides we used were pectin (P), sodium alginate (A), carboxymethylcellulose (CMC), arabic gum (AG), dextran (D), and dextran sulphate (DS). Choice of SBG was stipulated by their high biological value, relatively low production cost, and actuality of problems of processing proteins.

2. Experimental

2.1. Materials

In this work we used SBG isolated as follows. Soy beans were treated with tissue desintegrator Rt-1 ("EMA", USSR). The beans were separated from the broken coats on the sieves with hole diameters 3.16 and 1.6 mm, and after separation the beans were crushed by a hammer crusher. The meal was defatted by petroleum ether (in ratio 1:10) at room temperature and intensive stirring for three days. The solvent was renewed each 24 hours. To separate albumins we performed water extraction (pH 4.5) with a meal:water ratio = 1:20. Extraction procedure took 30 minutes. The meal was separated from the extract by centrifuging at 2,000 g during 30 minutes, then SBG were extracted by water (pH 8–9) within an hour. The ratio defatted meal: protein solution was 1:20. The extract was separated by centrifuging (2,000 g, 10 minutes). To obtain more complete isolation of globulins the extraction was repeated. The extracts were clarified by centrifuging at 50,000 g (30 minutes), after which SBG were precipitated at pH 4.5. The precipitate was centrifuged within 2–3 minutes (2,000 g), water washed (pH 4.5), dissolved in water (pH 8–9), filtered, and lyophilized. The yield was 20% of the defatted meal. The protein content was 90%, moisture content 2.5%. Sedimentation analysis showed that fractions 2,7,11 S, and >11 S amounted to 32.9; 32.1; 28.5 and 6.5 percent of total content, respectively. Lipid content was 0.7%, ash –4.5%, calcium –0.4%, phosphorus –0.8%, carbohydrates –0.5%. According to (13), weight average molecular weight of SBG is equal to ~220 • 10^5.
3. Results and discussion

Thermodynamic compatibility of SBG with polysaccharides has been studied for the following systems: water-SBG-pectin (W-SBG-P), water-SBG-sodium alginate (W-SBG-A), water-SBG-carboxymethylcellulose (W-SBG-CMC), water-SBG-arabic gum (W-SBG-AG), water-SBG-dextran sulphate (W-SBG-DS), and water-SBG-dextran (W-SBG-D).

3.1. Phase diagrams of W-SBG-PS systems

By phase analysis isothermal phase diagrams have been obtained for W-SBG-A, W-SBG-P, W-SBG-CMC, W-SBG-AG, W-SBG-DS, and W-SBG-D systems (figs. 1–6). For phase diagrams we have estimated coordinates of critical points expressed in weight (\( w_{2c} \) and \( w_{3c} \)) and molal (\( m_{2c} \) and \( m_{3c} \)) concentrations of the protein and polysaccharide (table 1), limits of compatibility: \( w_a \) characterizing the limit solubility of a polysaccharide in the concentrated \(^1\) solution of the protein, \( w_p(2.4\%) \) characterizing the limit solubility of the protein in the 2.4% solution of the polysaccharide \(^2\) (table 2), and the ratio between the molal concentrations of protein and polysaccharide in the critical point, characterizing symmetry of the phase diagrams (table 3).

\[ K_m = \frac{m_{3c}}{m_{2c}}. \]  

[1]

The character of the obtained phase diagrams points to incompatibility of SBG with the polysaccharides in aqueous media in a wide range of concentrations of macromonomers. The comparison of the phase diagrams of the W-SBG-APS system, obtained under identical conditions, shows that the concentration range of the two-phase state expands in a series W-SBG-AG, W-SBG-A, W-SBG-CMC, W-SBG-P.

The phase diagrams of the W-SBG-PS systems are highly asymmetrical. All studied two-phase systems can be arranged in order of increasing asymmetry of the diagrams: W-SBG-P < W-SBG-A < W-SBG-CMC < W-SBG-DS < W-SBG-AG. \( K_m \) varies disproportionately when passing from one polysaccharide to another. The greatest distinctions in the asymmetry of the phase diagrams

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\(^1\) If an increase in the concentration of the macromonomer \( (A_1) \) in the solution did not cause a decrease of the limit solubility of the second macromonomer in the solution of \( A_1 \), we considered the solution of \( A_1 \) concentrated.

\(^2\) Because of a high viscosity of the phase enriched with an acid polysaccharide, it was difficult to determine \( w_p \) from phase diagrams. Therefore, the protein limit solubility was determined with respect to 2.4% solution of polysaccharide. In this case it was possible to use the phase diagrams to compare the limit solubilities of the protein in the solutions of all studied polysaccharides.