



Effective recovery of casein from its highly diluted solution by using a technology of foam fractionation coupled with isoelectric precipitation



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ABSTRACT

A technology of foam fractionation coupled with isoelectric precipitation was developed for effectively recovering casein from its highly diluted solution without using any surfactants as stabilizers due to its poor surface activity. The results showed that foam stability could be improved by using the pore diameter of gas distributor 125 μm and increasing the liquid phase height to 1750 mm. In the two-stage continuous foam fractionation, the enrichment ratio and the recovery percentage of casein were obtained as high as 12.1 and 92.3%, respectively. Then, the foamate was treated by precipitation and the supernatant could be used as the feed solution of the first stage foam fractionation. By using the coupled technology, the enrichment ratio was further increased to 52.6 and the recovery percentage was 91.7%. The concentration of casein 0.01 g/L and its mass percent 5% were got in the residual solution and the precipitated solution, respectively.

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1. Introduction

Dairy industry is one of the important economic sectors and it has to do its part in meeting the nutritional needs of a growing population (Augustin et al., 2013; Menrad, 2003). However, the dairy industry has also been generally regarded as the largest source of food processing wastewater in many countries because water is used throughout all steps of dairy production, including cleaning, sanitization, heating and cooling (Sarkar et al., 2006; Fernández et al., 2010). The dairy industrial wastewater can easily induce serious environmental pollution without expensive sewage treatment (Dragone et al., 2009; Golalikhani and Razavi, 2016). Currently, the wastewater is usually discharged to a sewage plant and treated by an active sludge process (Fang, 1991). However, this method for treating the wastewater may result in the massive loss of a phosphorous protein, namely, casein which is the main protein component in mammalian milk. Thus, it is significant to develop a cost-effective technology to recover casein from the wastewater.

The reported methods for isolating a protein from its highly diluted solution included membrane separation, isoelectric precipitation, aqueous two-phase extraction, affinity chromatography and foam fractionation and so on (Zhao et al., 2013; Tomasula et al., 1997; Atasever et al., 2013; Li et al., 2014b). Among these methods, foam fractionation has become the most desirable one to enrich casein from the wastewater, in which the concentration of casein was in a range of 0.05–0.45 g/L (Perle et al., 1995; Rivas et al., 2011; Wei et al., 2010). In recent years, foam fractionation has been proposed to recover trace biomaterials in the downstream processing of biotechnology (Burghoff, 2012). Foam fractionation has been applied for separating nisin from its fermentation broth in an industrial scale by our team for many years (Liu et al., 2010). Thus, the recovery of casein from the dairy industrial wastewater by using foam fractionation has a great potential to be industrialized.

Casein is an ampholytic biosurfactant and it structurally contains high levels of hydrophobic and hydrophilic amino acid residues, but the strong tendency to self-assemble into micelles results in inhibiting its surface activity (Lin et al., 2009). It is well known that the basic requirement of separating a material from its highly diluted solution by using foam fractionation is to form a stable foam layer above the liquid phase. If the foam ability of a desired material cannot meet the above requirement, a surfactant with a strong

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surface activity will be added into its feed solution and used as foam stabilizer based on co-adsorption effect (Green et al., 2000). Burapatana et al. (2005) have evaluated the effects of three surfactants (e.g. SDS, CTAB and Pluronic F-68) on the foam ability of cellulase and achieved the effective recovery of cellulase from an aerated water solution by using foam fractionation. In fact, however, the used surfactants would seriously affect the processing characteristics of a desired material if they were not effectively removed from the foamate, which is the concentrated solution from the deforming foam discharging from the top of a foam column. So in this work, a new technology was developed for the effective recovery of materials which could not generate stable foams without using any surfactants as stabilizers.

Foam fractionation mainly contains two essential steps: interfacial adsorption in the liquid phase and foam drainage in the foam phase (Li et al., 2014b). In order to separate a material with a weak surface activity, it is necessary to increase foam stability and facilitate the foam discharge after foam drainage. The foam stability is affected by bubble size and the adsorption density of a desired material on the bubble surface. The smaller bubble size is, the more stable foam is. In foam fractionation, adsorption density is directly determined by the liquid phase height (Li et al., 2014a). When bubbles rise in the foam phase, foam drainage can induce the occurrence of their coalescence and disproportionation inside the foam, resulting in the reduction of foam stability. Then foam drainage is also closely related to the structure parameters of the foam fractionation column. In this work, a technology of foam fractionation coupled with isoelectric precipitation was developed to effectively recover casein from its highly diluted solution. A new two-stage foam fractionation was designed for enriching casein into the foamate as much as possible. Then, the foamate was treated by adjusting its pH to the isoelectric point of casein (pI 4.8) and the precipitated liquid could be used as the raw material of spray drying for the production of casein. Then, the supernatant could be drained back to the foam fractionation column as the feed solution. This work is expected to develop a new technology to effectively recover a material with a weak surface activity from its highly diluted solution.

2. Materials and methods

2.1. Materials

Casein was obtained from Tianjin Damao Chemical Reagent Factory, China. Hydrochloric acid and sodium hydroxide (Tianjin Yingdaxigui Co. Ltd., China) were used to adjust the initial pH of a feed solution. Coomassie Brilliant Blue (Beijing Dingguo Biotechnology Co. Ltd., China), phosphoric acid 85% (Tianjin Fengchuan Chemical Technology Co. Ltd., China) and ethanol 95% (Tianjin Dengfeng Chemical Reagent Factory, China) were used to determine the concentration of casein. All the reagents above were analytical grade.

2.2. Equipment

Fig. 1 shows the schematic diagram of the two-stage continuous foam fractionation process. All the columns were constructed by using polymethyl methacrylate tubes with an inner diameter of 45 mm and their heights were 700, 900, 1100, 1300, 1500, 1800 and 2100 mm, respectively. There were three gas distributors of lacunaris sintered glass and their pore diameters were 125, 180 and 425 μm , respectively. The air was introduced into the columns by an air compressor (AC0-318, Guangdong Hailea Group Co. Ltd., China). A rotameter (LZB-3WB, 0.03–0.3 L/min, Tianjin Meter Factory, China) was used to measure and control volumetric air flow rate. A

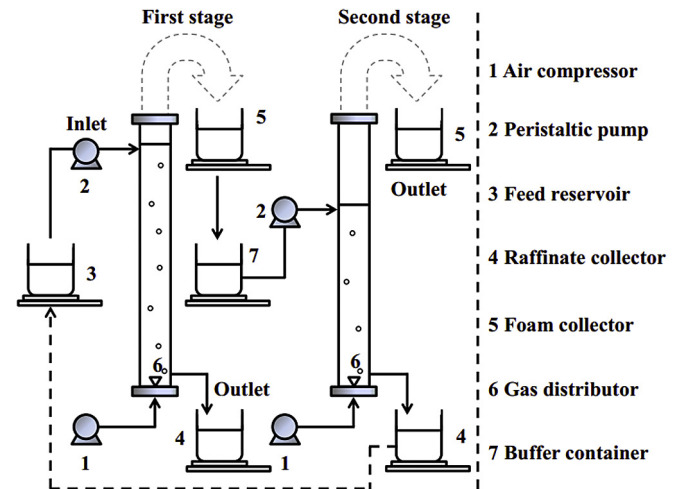


Fig. 1. Schematic diagram of the process of separating casein by two-stage continuous foam fractionation.

PHS-25 pH meter and a 721 spectrophotometer (Shanghai Precision & Scientific Instrument Co. Ltd., China) were used for measuring pH and casein concentration, respectively.

During the process of batch foam fractionation, a feed solution was injected into the column and the air was introduced into the column through the gas distributor mounted at its bottom. The foam flowed out from the top of the column and was collected in a foam collector. Volumetric air flow rate was controlled by a rotameter.

During the process of continuous foam fractionation, the inlet was 10 mm below the surface of the liquid phase and the outlet was at the bottom of the column. The feed solution was continuously pumped into the column from the inlet and the residual solution was continuously discharged from the outlet. The liquid phase height was controlled by the flow rate of the residual solution.

During the process of two-stage foam fractionation, the feed solution was continuously pumped to the column of the first stage from its inlet, the residual solution was discharged from its outlet. The foam flowed out from the top of the column and then formed a foamate in a foam collector. The foamate of the first stage was used as the feed solution of the second stage; the residual solution of the second stage was discharged from its outlet and then returned to the first stage as its feed solution. The foam flowed out from the top of the column of the second stage and its foamate had a high enrichment ratio of casein.

2.3. Measurement of casein concentration

Casein concentration was measured by the Coomassie Brilliant Blue assay at the maximum absorption wavelength of 595 nm. A linear relationship between the absorbance, A , and casein concentration, C (g/L), was $A = 5.205C + 0.0523$ with the linear correlation coefficient $R^2 = 0.9994$. The range of C was from 0.01 g/L to 0.10 g/L.

2.4. Measurements of foaming height and foam half-life

Foaming height characterizes the foamability of an aqueous solution and decides whether the solution can be used in foam fractionation. In this work, a foam was generated by flowing the air through a gas distributor into a liquid phase. When the casein concentration of a feed solution was high enough, the foam went up and flowed out from the top of the column and then foaming height

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