Separation and Purification Technology 132 (2014) 577–583

Contents lists available at ScienceDirect

Separation and Purification Technology

journal homepage: www.elsevier.com/locate/seppur

Extraction of keratin with ionic liquids from poultry feather

Yimei Ji, Jinyang Chen *, Jingxiao Lv, Zhilian Li, Luyao Xing, Siyuan Ding

School of Environmental and Chemical Engineering, Shanghai University, Shanghai 200444, China

ARTICLE INFO

Article history:
Received 11 November 2013
Received in revised form 8 May 2014
Accepted 17 May 2014
Available online 16 June 2014

Keywords:
Imidazole ionic liquid
Extraction
Poultry feather
Keratin
1-Buty-3-methylimidazolium chloride

ABSTRACT

Ionic liquid (IL) is a new eco-friendly solvent to accelerate dissolution of natural polymer and several common water-soluble imidazole ionic liquids (ILs) have been selected to dissolve poultry feather and extract keratin. [Bmim]Cl has shown the best solubility, thus it has been studied to extract the keratin. Because Na_2SO_3 can unfold disulfide bond of polypeptide chains in different keratin molecules in feather by forming R-SSO_3Na and the addition of water can increase solubility of Na_2SO_3 and decrease viscosity of solution, Na_2SO_3 and water have been used to improve the extraction process. During the solubilization process, SEM pictures show that the smooth structure of the feather was destroyed and followed by the dissolution of the keratin into the liquid phase. The keratin was easily separated from the liquid phase as a solid precipitate after adding some more water owing to the miscibility of the IL with water and the immiscibility of the keratin with water. The keratin precipitate was filtrated and the liquid was distilled to remove water, then the IL and Na_2SO_3 can be recycled. The optimum extraction conditions for keratin are: 20 wt.% of water in IL–water, 10 wt.% Na_2SO_3 in liquid phase, the weight ratio of liquid/feather = 20, extraction temperature at 90 °C, and extraction time of 60 min; under these conditions, the dissolution rate of feather is 96.7% and the yield of keratin is 75.1%.

1. Introduction

The poultry industry produces a great amount of feather each year especially with popular fast food such KFC and only a little part of them is used as filler, adornment and forage. Although the feather can be biodegraded, it not only needs large dumping ground but also discharges the landfill leachate and pollution gas. Thus if not treated in time it causes an environmentally disposal problem [1]. Recently it is very common to hydrolyze keratin in poultry feather with strong acid to produce amino acid in China, but the process results in greatly acid pollution. Enzymatic catalysis hydrolysis is a potentially alternative method, but up to date owing to the outer protective film and compact structure, it is far from industrial application. Therefore, it is very urgent to recycle feather with eco-friendly way.

It is known that keratin is rich in feathers, usually more than 70 wt.%, owing to its unique molecular structure and crystal arrangement [2], keratin has important applications in biomaterials [3], biomedical [4], flocculants [5] and adhesive [6]. In addition, the hydrolysis to produce amino acid is indeed the hydrolysis of keratin in feather. If the pure keratin is extracted from feather, the hydrolysis is no doubt easier because there is no protective film and compact structure and the eco-friendly enzymatic hydrolysis can be industrial application. Therefore, from both environmental and economic point of views, it is quite desirable to develop effective processes to extract keratin from poultry feather.

Feather keratin is a structural protein, characterized by high cystine content and a significant amount of hydroxyl amino acids, especially serine. Feather keratin involves a range of non-covalent interactions (electrostatic forces, hydrogen bonds, hydrophobic forces) and covalent interactions (disulfide bonds) [7]. In addition, its molecular chains also have complex structures of α-helix and β-sheet [2], and these structures must be destroyed during the extraction of keratin. Therefore, it is difficult to dissolve feather to extract keratin. The traditional methods of keratin extraction from feather usually used strong acid, alkali or high concentration of salt solutions [7,8]. These processes were multi-steps and could result in degradation of protein. Furthermore, owing to the consumption of a large quantity of reagents that could not be recovered, they polluted our environment. Although the use of superheated water has been shown as an eco-friendly processing method [9], it could break down the peptide bonds in keratin molecules and result in degradation of protein. Thus, it is urgent to develop a new simple and eco-friendly method to extract feather keratin.

Recently, ionic liquids (ILs), a group of salts existing as liquids at relatively low temperatures, have drawn intense attention as a type of eco-friendly and safe solvents with their advantages of non-volatility (i.e., no discharge comparing with traditional organic solvent),

* Corresponding author. Tel.: +86 21 66137729; fax: +86 21 66137725.
E-mail address: chenjy@shu.edu.cn (J. Chen).

http://dx.doi.org/10.1016/j.seppur.2014.05.049
1383-5866/

non-flammability, chemical and thermal stability and easy recycling. They have remarkable solubility ascribed to their own ionic structure comparing with traditional solvents. In 2002, Swatloski et al. [10] reported that cellulose could be dissolved in ILs and the regenerated cellulose was tested with no significant changes in the degree of polymerization and dispersibility. This discovery has received great attention for the study of ILs as solvents for the natural polymer materials. Owing to the strong polar ILs can destroy the usually strong inter-molecule effect, such as hydrogen bonding in natural polymers, and enhance the dissolution of polymers, so that ILs have been developed to a new class of eco-friendly solvents for natural polymers and the structures of the anions and cations had great influence on the dissolution [11–14].

Up to date, the type of ILs with imidazole cation has been studied to show excellent dissolution for pretreatment of lignocellulose [15–19], wool keratin [20–23], and extraction of proteins [24]. In addition, the imidazole ILs are very common and relatively inexpensive, which is great advantage to decrease the obstacle of the high price for industrial applications. Thus in this study, several water-soluble imidazole ILs were tested to dissolve feather and extract keratin.

In order to improve extracting efficiency, some additives could be used. For example, the imidazole ILs with a little NaOH can mutual enhance pretreatment of corn stover [25]. As for the extraction of keratin from feather, the disulfide bonds of cystine in keratin are the main obstacle. Na_2SO_3 can unfold the disulfide bonds according to the following reaction to accelerate the dissolution of keratin.

$$RSSR' + SO_3^{2-} \rightarrow RSSO_3^- + R'S^-$$

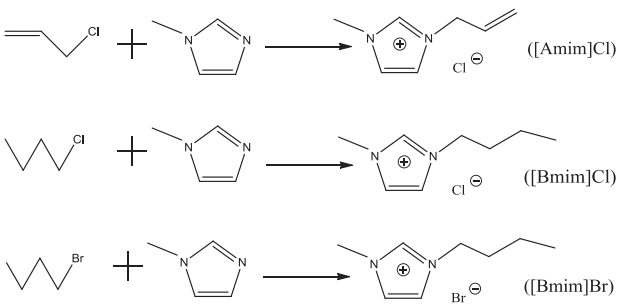
The Na_2SO_3 is a weak alkaline salt, therefore, it cannot destroy the keratin and can be used as an accelerant to extract keratin from feather.

2. Experimental

2.1. Material

Duck feather is a typical poultry feather and the sample of duck feather was provided by Xinyi Hanling Biological Engineering Co., Ltd. (Xuzhou, China). N-methylimidazolium, 3-chloropropene, 1-chlorobutane, 1-bromobutane, Na_2SO_3, iodine, $NaHCO_3$, $Na_2S_2O_3$, and starch were purchased from Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China).

ILs [Amim]Cl, [Bmim]Cl and [Bmim]Br were prepared following the three one-step reactions:



[Bmim]NO_3, [Hmim]CF_3SO_3 and [Bsmim]HSO_4 (1-sulfobutyl-3-methylimidazolium hydrogen sulfate) were purchased from Shanghai Cheng Jie Chemical Co. Ltd. (China).

2.2. Keratin dissolution from feather and precipitation

Duck feathers were washed with water, dried and cut into small pieces. The different weight ratios of IL, duck feathers, Na_2SO_3, and water were put into beakers and heated to fixed temperature under magnetic stirring condition. After desired time, the residual solids were removed by suction filtration immediately under the high temperature because the increase of viscosity with decreasing temperature will cause difficulty in filtration. Then, the keratin solution was added with some water at room temperature to precipitate keratin. The solid keratin and liquid phase were separated by suction filtration. IL was recovered from the liquid IL–water phase by evaporating water, and it can be used repeatedly.

The dissolution rate of feather and yield of keratin were used to evaluate the optimal extraction condition. The dissolution rate of feather is expressed as $[(M_0 - m_t)/M_0] \times 100\%$, where M_0 is the initial weight of feather and m_t is the weight of residual feather. The yield of keratin is expressed as $[M_t/M_0] \times 100\%$, where M_t is the weight of keratin.

2.3. Analysis of the feather and extracted keratin

The content of protein in the feather and the obtained keratin were determined by hydrolysis with 6 mol/L HCl solution to produce amino acids at the reflux temperature of about 105 °C during 24 h. Then the total amino acids were analyzed by amino acid analyzer (AAA-Direct, DIONEX) and the measuring condition is the same as that described in the previous paper [26]. The FTIR spectrum of keratin was measured on an Avater370 FTIR spectrograph (Nicolet Co., USA) by KBr method. The molecular weight of extracted keratin was determined by gel permeation chromatography (GPC) (HP-1100) system, which consists of a Phenogel mixed column and a RID (G1362A) detector. THF was used as a mobile phase at a flow rate of 1.0 mL/min and the injection volume was 20 μL. Calibration of GPC was carried out with a standard polystyrene sample.

2.4. Content of Na_2SO_3 in recovered IL

Although Na_2SO_3 benefits the dissolution of keratin by opening the disulfide bonds, the process of keratin precipitation should cause the recovery of these opened disulfide bonds. If the disulfide bonds are recovered with the precipitation of keratin, the Na_2SO_3 should dissolve in IL–water solution and the content of Na_2SO_3 gives the information about the precipitation and extraction of keratin. This is a chemical titration measurement based on the following two equations:

$$SO_3^{2-} + I_2 + 2HCO_3^- = SO_4^{2-} + 2I^- + 2CO_2 \uparrow + H_2O \quad (1)$$

$$2S_2O_3^{2-} + I_2 = 2I^- + S_4O_6^{2-} \quad (2)$$

First, a certain amount of solution with excess iodine and sodium bicarbonate was added to the liquid after filtration of keratin, such that SO_3^{2-} was reduced by I_2 according to Eq. (1). Then, according to Eq. (2), sodium thiosulfate solution was added to oxidize the excess I_2 in the above solution with starch solution as indicator. By subtracting the excess I_2 derived from Eq. (2) from the original total I_2, the amount Na_2SO_3 in the recovered IL was determined.

3. Results and discussions

3.1. In situ change of feather and precipitation of keratin

Some photographs showing dissolving process of the feathers in the IL system are given in Fig. 1 under the temperature of 90 °C, with 20 wt.% of water in IL–water, weight ratio of liquid/feather = 20, and 10 wt.% Na_2SO_3. The branches of feathers dissolved very quickly and at about 20 min these parts were mainly dissolved. At 40 min, the main residual feathers were the stalks and usually they were totally dissolved at 60 min.