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Research Article

Extraction and Characterization of Natural Protein (Keratin) From Waste Chicken Feather

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Abstract

Currently, the abundant quantity of chicken feathers produced by the poultry industry as waste is one of the major problems. The burning and burying disposal method of chicken feathers results in the release of more carbon dioxide, and methane respectively. Thus, it is imperative to convert these wastes into valuable products like keratin. The use of keratin in biomedical applications, and nanoparticles for food, medical, cosmetics, and pharmaceutical industry become a current global issue. Therefore, it is important to extract keratin from waste chicken feathers. In this research, keratin was extracted from waste chicken feathers at lab scale using different concentrations of sodium sulfide as a reducing agent. The main processes include; pretreating, solution preparation, filtration, centrifugation, precipitation, and purification. From different experimental work conducted, about 63.25% of pure keratin was extracted using 0.5 M of sodium sulfide at optimum temperature and PH value of 30°C and 10-13 respectively. The effect of temperature, the concentration of sodium sulfide, retention time and PH value were investigated. It was observed that as the concentration of sodium sulfide increase to about 0.5 M with an optimum temperature of 30°C, PH value restriction of 10-13 and an extraction time of 6 h the yield also increased. The result of characterization using a UV-visible spectrophotometer and a biuret test showed that keratin was already extracted. By this research, it can be concluded that keratin can be extracted from waste chicken feathers and can be used for several purposes.

Keywords: Extraction; Keratin; Characterization; Waste Chicken Feather; Reducing agent.

Introduction

Keratin proteins of waste chicken feathers have generated much interest recently. It is the most abundant structural fibrous protein of bird feathers, hair, skins, bristles, horns, and hooves. The feather is composed of about 91% of keratin, which has a structural characteristic of materials of high mechanical strength [1, 2, 3]. Many billions of tons of keratinous wastes are generated annually globally, especially in the wool textile industry and in poultry slaughterhouses [4]. Also in Ethiopia, billions of tons of chicken feather wastes produced from the poultry industry, and slaughterhouses [5]. Five percent of the body weight of poultry is feathers; the slaughterhouse with a capacity of 50,000 chickens can easily produce 2-3 tons of dry feathers per day [6]. Chicken feathers discarded during the production of poultry for human consumption are a big problem since chicken feathers can pose hazards to human and environmental health as they often contain viruses and bacteria.

There is little demand for waste chicken feathers and most poultry producers disposes of more than five billion tons of feathers produced annually worldwide by burying or burning the feathers or grinding them up for addition to livestock feed. Burning is the most common disposal technique and can result in the release of 50 times more carbon dioxide than the coal industry which is a serious problem to the environment [7]. Further, burning feathers in special installations are economically ineffective. Thus, extraction of keratin from feather waste and using it for useful bio-products, in biomedical engineering applications, micro and nanoparticles for the purpose of food, medical,

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fertilizers, textile and clothing industries, cosmetics, and bio-plastics would be very valuable for decreasing the environmental problems and to overcome the current and future demand of keratin [8, 9]. The keratin protein solution can be used for several purposes such as anti-aging cream, shampoo, and conditioner and medical purposes such as bone replacement and bone grafting.

Currently, there is an increasing interest in the development of materials that are environment-friendly, obtained from renewable resources [10]. The main renewable materials are obtained from polysaccharides, lipids, and proteins. Proteins are polymers formed by various amino acids capable of promoting intra- and inter-molecular bonds, allowing the resultant materials to have a large variation in their functional properties. Protein deficiency is a serious cause of ill health and death in developing countries. The protein shortage for food and other extra applications obliges to look for new protein sources, including waste products. Poultry feathers are bio-resource with high protein content.

The present research was undertaken to extract natural protein from chicken feathers by using reducing agents that will decrease the stability of keratin fibers in the solid form found in feathers. The reducing agents help in decreasing the stability of keratin fibers in the solid form found in feathers. These reagents break down disulfide bonds, hydrogen bonds and salt linkages of the keratin fibers to dissolve it into protein solution. By this study, keratin was extracted at lab scale using sodium sulfide as a reducing agent. The extraction was conducted using different procedures. Waste chicken feathers were collected, pretreated, ground, dissolved using a different concentration of sodium sulfide, separated and finally purified.

Materials and methods

During this research, the chemical extraction method which comprises different concentrations of sodium sulfide as a reducing agent was used. The extraction method was carried out by different stages of experimental procedures as listed below.

Collection and Surfactant pretreatment of waste chicken feathers

Waste chicken feathers were collected from chicken processing small poultry house of Jimma town Kochi kebele (Ato.Addis Zeleke poultry house). The collected feathers were soaked in ether for 24 h to clean the feathers from stains, oil, and grease. The feathers were then washed with soapy water and dried to remove its moisture content and to make an easily crushable sample, at 60^{o}C for 6 hr in the oven. The dried chicken feathers were ground by using both mortar and miller. The particle sizes of the ground sample were 0.25 mm, 0.5 mm, and 2 mm. Each sample was kept in a sealed plastic bag at room temperature until the next stage of the experiment.

Solution preparation, heating, centrifugation and filtration

1 L of 0.3 M, 0.4 M, and 0.5 M sodium sulfide solution were prepared in a 1 L flask separately. 25 g of the ground chicken feathers were weighed and added to each sodium sulfide solution. The solution was heated to the temperature of 30^{o}C, pH was maintained in basic media between 10 -13 and the solution was continuously stirred for 6 h. The solution was then filtered and centrifuged at 10,000 rpm for 6 min. The supernatant liquid was carefully collected then filtered using filter paper to make it particle free.

Precipitate preparation

175 g of ammonium sulfate was dissolved in 0.25 L deionized water. The solution was stirred until all the ammonium sulfate particles dissolved and were filtered to make it particle free.

Natural protein (keratin) precipitation

A filtrate solution collected earlier was placed in a beaker and stirred continuously. A precipitate was added slowly dropwise to the filtrate. The two components were with a 1:1 ratio. The solution was centrifuged at 10,000 rpm for 6 min and the solids particles were collected. The supernatant liquids were collected separately and step 2 and 3 was repeated with it carefully.

Keratin purification

The solid particles were collected and added into 20 ml deionized water and stirred. The solution was centrifuged at 10,000 rpm for 6 min and the solid particles were gathered

carefully. The collected solid particles were dissolved in 100 ml of 2 M sodium hydroxide solution. The solution was centrifuged again at 10,000 rpm for 6 min and all the liquids were collected carefully and stored. The precipitating, washing and dissolving step was repeated at least for 3 to 4 times.

Biuret test method characterization procedures

1 % copper sulfate solution and 1 % potassium hydroxide solution was prepared, and 6 ml of the pure keratin solution was collected and mixed with potassium hydroxide solution with a 1:1 ratio. After the whole, three drops of copper sulfate solution were added to the mixture solution and changes in the solution were observed and recorded.

Spectrophotometer (UV-Vis-Sp method) characterization procedure

A small sample of keratin protein extracted after purification was prepared and diluted using different solvents as basic media (sodium hydroxide solution, biuret test solution, and deionized water), and the spectrophotometer was adjusted using these basic media solutions. The liquid form of keratin protein was feed to the spectrophotometer and the absorption versus wavelength of keratin was observed and analyzed.

Result and discussion

Figure 1 shows that as the concentration of Sodium Sulfide increases, the yield of keratin protein also increased.

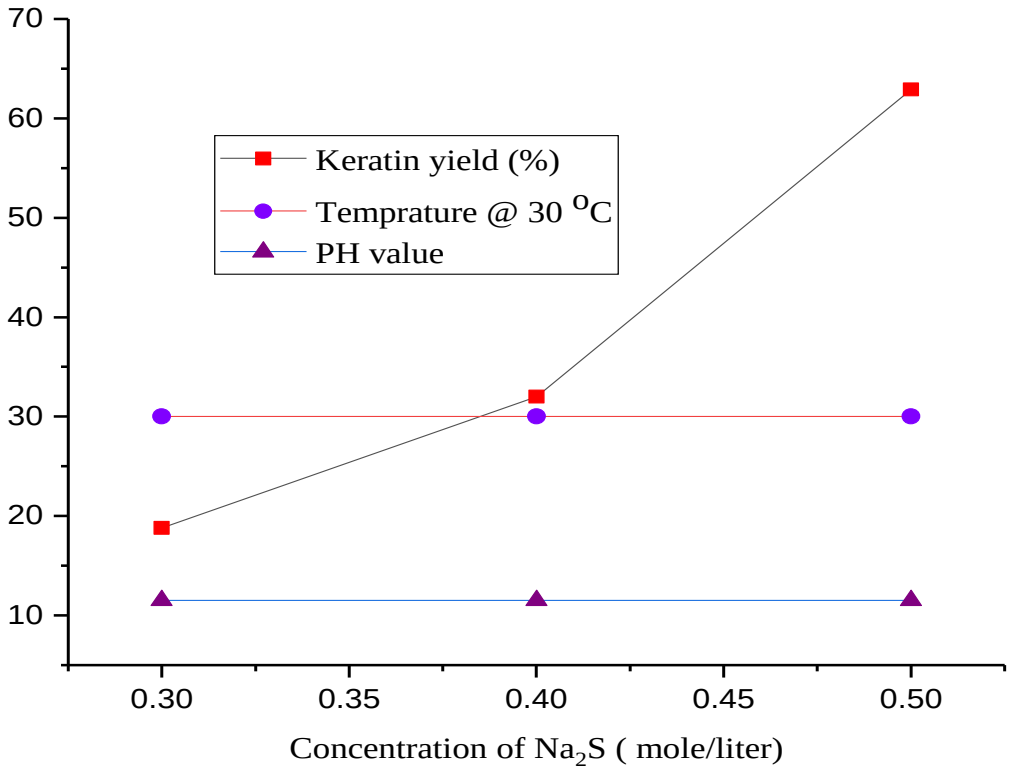

Figure 1. Effect of concentration of sodium sulfide at constant temperature and pH

As observed from Figure 2, the minimum and maximum yield of keratin protein was 18.8 and 63.25% at a constant temperature of 30°C and pH value of 10 – 13 with 0.3 M and 0.5 M respectively. It is shown that a further increase in the concentration of Na_2S results in a decrease of the total yield. Thus, the concentration of the extraction medium is an important parameter, as the concentration increased, the disulfide bonds decreased [11]. Therefore, an effective concentration of sodium sulfide, which is most probably 0.5M, can extract keratin at an optimum temperature of 30°C and pH 10–13 with a retention time of 6 h.

One major factor in the yield of keratin protein is particle size. Figure 3 shows that less keratin was recovered from a particle of mesh size > 0.5 mm and ≤ 0.25 mm. The main reason is that a larger particle with smaller contact surface area, has more resistance to the solvent entrance and it is difficult to break down the disulfide bonds. Therefore, less amount of keratin will be extracted. In another way, very fine particles specifically less than 0.25 mm can agglomerate and the extraction process becomes ineffective, which then reduces the yield of keratin.

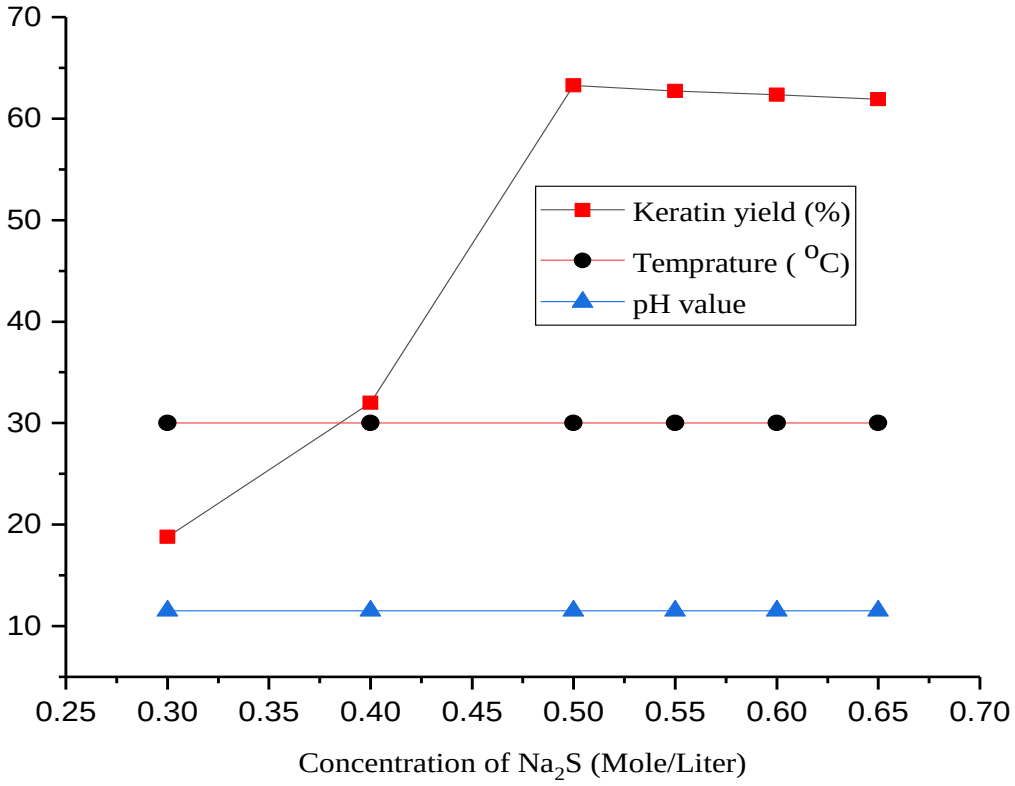

Figure 2. Investigating effective concentration of Sodium sulfide to maximize keratin yield

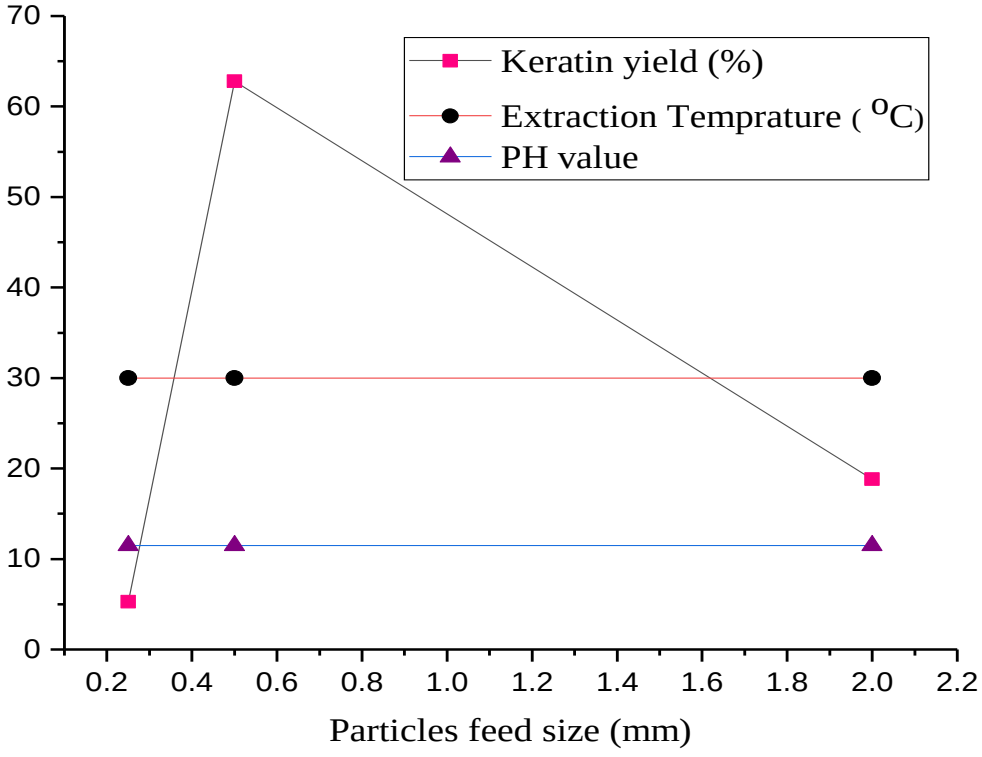

Figure 3. Effect of particle size during extraction

Temperature is an important factor that affects the yield of keratin and the rate of dissolution of feather keratin at various temperatures was determined while keeping other conditions constant. With the increase of reaction temperature a gradual decrease in the yield of keratin was recorded (Figure 4). Therefore, 30°C was selected as an optimal extraction temperature. The main reason is that it denatures the natural protein and decreases the total yield of keratin. Extraction temperature showed a significant effect on the yield of keratin extracted [12].

As observed from above Table 1, the maximum and minimum wavelength is 301 nm and 276 with the absorbance of 0.47 and 0.107 respectively. The color change obtained from the biuret test was similar to the purple plate with a wavelength of 315 nm and 253 nm (Table 1). The result from UV has to be from 250-280 nm and 320-410 nm [13].Therefore; keratin was extracted from chicken feathers at the lab scale.

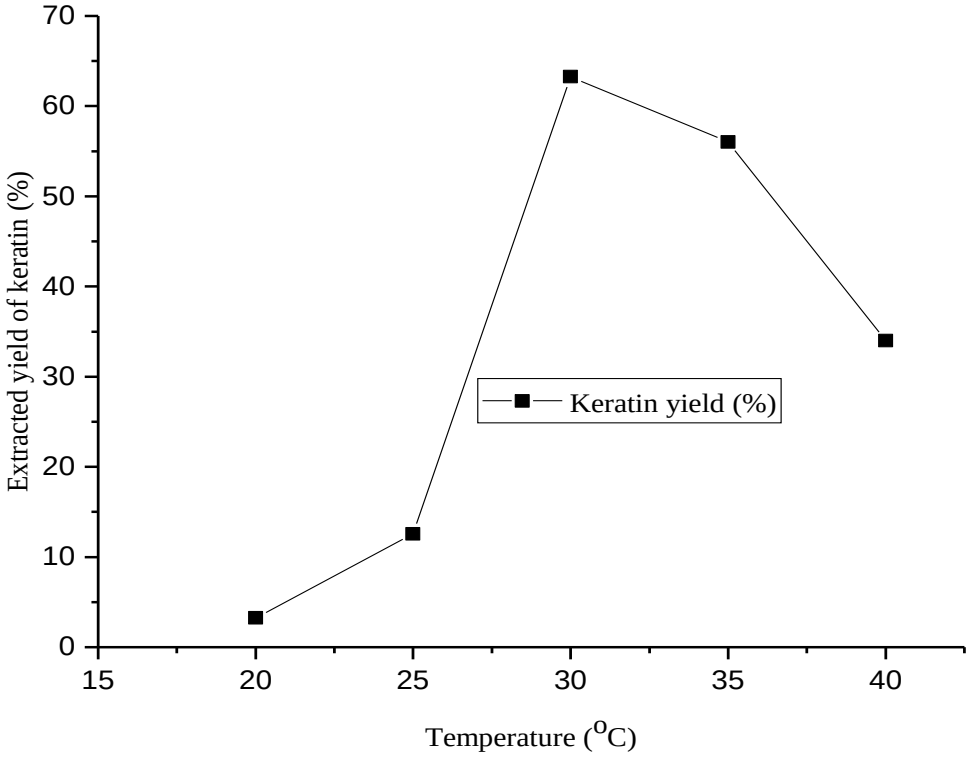

Figure 4. Effect of temperature on yield of keratin

Table 1. Characterization result of keratin protein by UV and biuret test

Analysis type	Basic media	Absorbance	Color change
UV analysis of biuret test	KOH	2.385 at 315nm and 0.287 at 253nm	Similar to purple plate
UV Spectro analysis of pure keratin	Deionized water	0.474 at 301nm and 0.107 at 276nm	

As a whole, from this experimental work, the sodium sulfide solution can extract keratin protein from chicken feathers. As more chicken feathers are dissolved at optimum temperature and PH value, the yield of keratin protein increased. About 63.25% of keratin protein was extracted using 0.5 M of sodium sulfide through an extraction time of 6 h. keeping pH value in alkaline media of 10-13 at 30^{o}C, facilitate the extraction of keratin. In another way, particle size and moisture were other factors that affect the extraction of keratin. By the biuret test, a purple plate color was observed.

Conclusions

Chicken feathers are one of the main sources of keratin protein. However, these feathers simply discarded to the environment from poultry processing houses and industry, which then pose environmental and health problems due to its time-consuming decomposition. Thus, valorizing these waste materials to meet its applications for industrial and personal purposes is an imperative way in countries like Ethiopia where such materials are considered as invaluable. By this study, keratin protein was extracted from waste chicken feathers using a sodium sulfide solution as a reducing agent. Through its whole process of extracting the study tried to relate different factors that affect the yield of a product like a temperature, retention time, the concentration of the solvent and pH. By controlling the pH of 10-13 and temperature value of 30^{o}C, keratin was extracted. Variation of temperature affects the pH value which then affects the product yield. As a whole, temperature, the concentration of the reducing agent and pH are the interrelated

parameters that affect the extraction of keratin. Accordingly, through the experimental work of the chemical extraction method used to extract keratin, about 63.25% of keratin was obtained.

Conflict of interest

Authors declare no conflicts of interest.

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