

## Albumin Identification in Catfish ( *Clarias sp* ) Using Spectrophotometry UV-Vis

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### ABSTRACT

Albumin is a simple protein with globular protein molecules. Albumin is water soluble and can be coagulated with heat. Albumin solution in water can be precipitated by adding ammonium sulfate until saturated. Albumin functions to regulate the osmotic pressure in the blood, maintain the presence of water in the blood plasma so that it can maintain the volume of blood in the body and as a means of transportation and transport. Catfish contains 15.6 grams of high-quality protein in each fish so that it can meet the needs of the amino acids needed by the body. The high-quality protein in catfish helps the body build lean brain mass and the protein content in catfish also helps increase the effectiveness of immune function. The purpose of this study was to determine albumin levels in catfish by extracting catfish meat using the oven method and determining the maximum albumin wavelength. In the identification test with the heating method and UV-Vis spectrophotometry test. This research method uses the experimental method. The extraction results were then heated in an oven to observe coagulation and determine the wavelength using UV-Vis spectrophotometry using Bovine Serum Albumin (BSA) as a reference solution and Bromocresol green as a reagent. The conclusion of this study is that catfish meat extract was coagulated using the oven method and maximum absorption was obtained at a wavelength of 625 nm using UV-Vis spectrophotometry.

**Keywords :** Albumin , Catfish, UV-Vis Spectrophotometry.

## INTRODUCTION

Protein is a food nutrient that is very important for the body, functions as a building substance in the body. Protein is a source of amino acids that contain the elements Carbon (C), Hydrogen (H), Oxygen (O), and Nitrogen (N) which are not owned by fat or carbohydrates. (Apriyana, 2014) .

Classification of proteins according to their structure is divided into two major groups, namely simple proteins and combined proteins. Simple proteins are proteins that consist of amino acid molecules, while compound proteins are proteins that consist of protein and non-protein groups. Simple proteins are divided into two parts according to their molecular shape, namely fiber proteins and globular proteins (Sasongko et al., 2010)

Albumin is the main protein in human plasma and makes up about 60% of the total plasma protein. The liver produces 12 grams of albumin per day which is 25% of the total liver protein synthesis and half of all protein secreted by the organ (Galuh Sayang Kusumaningrum, 2012).

Albumin is a simple protein with a globular protein molecule shape. Albumin is water soluble and can be coagulated with heat. Albumin solution in water can be precipitated by adding ammonium sulfate until it is saturated (Sasongko et al., 2010) .

The spectrophotometric method is one method that is widely used to analyze the characteristics of a material by analyzing the spectrum produced by the material. The spectral image of a material, which is the result of the interaction between radiation energy and the atoms or molecules that make up the material, is expressed as a variation of radiation intensity or absorbance as a function of wavelength (Maknumah, 2015)

Fish is one of the aquatic products that is widely used by humans because of several advantages. Fish is a very potential source of animal protein and usually the protein content is around 15-24 percent depending on the type of fish. Fish protein has a very high digestibility, which is around 95% (Chasanah et al., 2015) .

Catfish contains 15.6 grams of high-quality protein in each fish so that it can meet the needs of the amino acids needed by the body. The high-quality protein in catfish helps the body build lean brain mass and the protein content in catfish also helps increase the effectiveness of the body's immune function (Puspitaningsih, 2018).

Seeing the background above, it can be seen that fish contain albumin, therefore researchers are interested in conducting research on the presence or absence of albumin in catfish and determining the maximum wavelength using a spectrophotometer.

## **TOOLS AND MATERIALS**

The tools used include: analytical balance, knife, 100 ml beaker glass, test tube, steamer pan, measuring cup, volumetric flask, syringe, spectrophotometer, water bath, petri dish, cuvette and stir bar.

The materials used include: catfish, distilled water, Bovine Serum Albumin (BSA), Bromocressol green (BCG) and pH 4 buffer.

## **METHOD**

### **Research procedure**

#### **A. Preparation of albumin extract from catfish meat (Clarias sp.)**

The catfish is cleaned from the head and entrails of the catfish, scales, fins, tail and the part of the meat eaten (filleted) is taken. Wash the catfish meat with distilled water until it is clean and no part of the stomach contents remains. Weigh the filleted catfish meat, then put it in a petri dish, place it sideways, put it in the oven with a temperature of 100°C, a yellowish white extract comes out of the meat after 42 minutes (Abdulgani, Ulfin, & Biologi, 2013). The result of pressing is the juice of catfish meat. The resulting catfish extract was reheated in a saucepan over low heat for 30 minutes. The results show positive albumin (coagulation occurs) (Fitriyani, 2018)

#### **B. Preparation of BCG Reagent (Bromocresol green)**

The required BCG reagent is a BCG solution with a concentration of 0.01%, so it is necessary to make a stock BCG reagent with a concentration of 1%. Preparation of 1% stock BCG reagent was carried out by weighing 100 mg of BCG powder and then dissolving it into 10 ml using a buffer solution of pH 4 and stirring until homogeneous. BCG reagent 0.01% was prepared by taking 1 ml of BCG reagent stock and then diluting it using a buffer solution of pH 4 to 10 ml. All dilutions were made using a volumetric flask (Alfarisy, 2014) .

#### **C. Determination of Standard Wavelength**

Albumin stock solution was prepared by weighing 100 mg of BSA (Bovine Serum Albumin) then dissolved in 100 ml of distilled water and homogenized. The standard solution was prepared

with a concentration of 10 ppm from the dilution of the mother liquor. A standard 10 ppm albumin solution is prepared by taking 1 ml and then dissolving it to the mark. Then take 0.5 and add 2.5 ml of 0.01% BCG and leave for 10-15 minutes with a wavelength of 500-650 nm (Safir et al., 2022) .

#### D. Identification of Albumin by UV-Vis Spectrophotometry

Albumin stock solution was prepared by weighing 100 mg of the sample and then dissolved in 100 ml of distilled water and homogenized. The sample solution was prepared with a concentration of 10 ppm from the mother liquor dilution. A 10 ppm sample solution was prepared by taking 1 ml and then dissolving it to the mark. Then take 0.5 and add 2.5 ml of 0.01% BCG and leave for 10-15 minutes with a wavelength of 500-650 nm (Safir et al., 2022) .

### RESULTS

#### 1. Albumin extraction .

$$\% \text{ Extract} = \frac{\text{weight of extract}(g)}{\text{weight of sample}(g)} \times 100\%$$

$$\% \text{ Extract} = \frac{58 \text{ gram}}{875 \text{ gram}} \times 100\%$$

$$\% \text{ Extract} = 6,62 \%$$

#### 2. Maximum wavelength determination

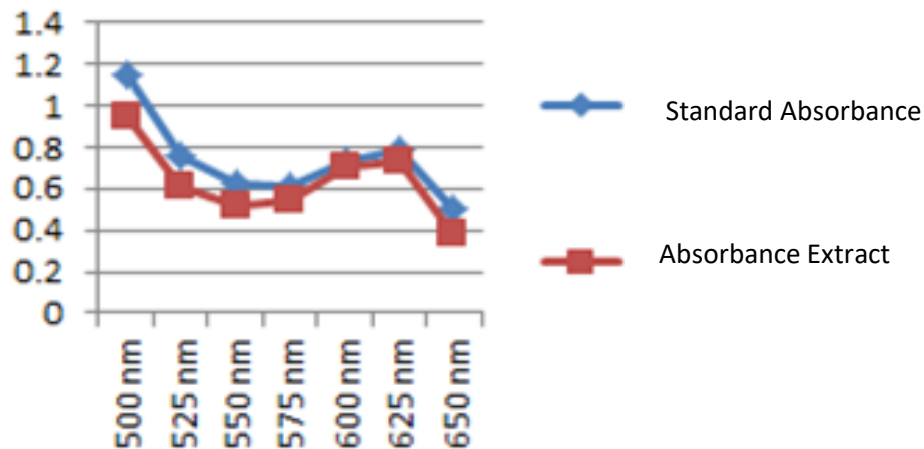


Figure 1. The maximum wavelength of albumin.

## DISCUSSION

Albumin extraction.

Observations of catfish albumin extract obtained in line with Wirahadikusuma's 2016 stated that extraction with temperatures above 70°C produced fish extracts that were cloudy white in color due to the large amount of precipitate. Turbidity in processed fish extract at temperatures above 70°C can be caused by some of the plasma proteins being coagulated by heat (Fitriyani, 2018) .

The extract obtained was then subjected to an albumin identification test which aimed to ensure that the extract obtained contained albumin. Identification of albumin was carried out by the heating method, namely by heating catfish extract (*Clarias sp.*) for 30 minutes over low heat in a pan. Albumin will coagulate and form clots. Based on the results of the identification test of catfish extract (*Clarias sp.*) It produces lumps after being heated so that it can be seen that the catfish extract (*Clarias sp.*) positively contains albumin (Fitriyani, 2018) .

The lumps that occur are characterized by being shaped like a gel, containing water and also quite sticky (Fitriyani, 2018) . According to Poedjiadi, albumin belongs to a group of globular proteins which are generally round and elliptical in shape and consist of folded polypeptide chains (Alauddin, 2016) .

Albumin also has water soluble properties and can be coagulated with heat. Protein denaturation is a change in protein properties due to the breaking of hydrogen bonds and non-polar bonds in protein molecules. Protein denaturation can occur by heating, heating albumin will cause denaturation followed by coagulation (Maknumah, 2015). If the globular protein is denatured, no covalent bonds in the polypeptide chain are damaged, but in biological activity almost all proteins are damaged causing their solubility to decrease. (Alauddin, 2016) .

Maximum wavelength determination

In determining the maximum wavelength of albumin (standard solution and sample) a 100 ppm mother liquor is made, where 100 mg (standard solution and sample) is dissolved with 100 aquadest up to the mark in a volumetric flask, then a series of 10 ppm solution is made by dissolving 1 ml of solution parent stock using distilled water up to the mark on the volumetric flask. Then 0.5 ml was taken and 2.5 bromocresol green (BCG) was added (Safir et al., 2022) .

Perform a maximum wavelength determination test with a UV-Vis Spectrophotometer. The results obtained are maximum absorption values at wavelengths in the range of 500 nm – 650 nm (Maknumah, 2015) . There was agreement between the sample results and the standard indicating that the catfish (*Clarias sp.*) extract sample contained albumin according to the Bovine Serum Albumin standard with a wavelength of 625 nm. For catfish (*Clarias sp.*) extract solution and Bovine Serum Albumin (BSA) standard solution, see Appendix 6.

In measuring albumin, standard solutions and extracts, bromocresol green (BCG) was added using a spectrophotometer. Examination of albumin with BCG in citrate solution forms a color complex. The absorbance of this color complex is proportional to the concentration of albumin in the sample. The intensity of the green color indicates albumin levels in serum (Dwi Harjanto, 2016) and the addition of bromocresol green (BCG) because it is not affected by interfering compounds such as bilirubin and salicylate, bromocresol green (BCG) is commonly used to measure albumin (Safir et al., 2022) .

The absorbance value obtained at a wavelength of 500 nm (standard solution and sample) above 0.8 if the absorbance value is very large ( $> 0.8$ ) indicates the concentration absorbed is very large, likewise the dilution should not be too concentrated, because if the concentration is too concentrated can also produce an absorbance value above 0.8 because the concentration absorbed is very high (Khasani & Astuti, 2019) .

Factors that affect absorbance include the type of solvent, pH, temperature, high electrolyte concentration and the presence of interfering substances. Cleanliness will also affect absorption including finger prints on the walls of the tube should be cleaned with a tissue and only hold the top end of the tube before measurement. In addition to these factors, the absorbance value of the existing sample solution is measured at each wavelength and must be replaced, not using the same sample solution. This will result in the accuracy of the absorbance value being reduced, because the solution passed by the monochromatic light beam will affect the actual absorbance value. The next factor is that the sample solution to be tested is colorless, if it is colorless then the sample solution must be colored by giving a certain color-forming reagent (Khasani & Astuti, 2019) .

## CONCLUSION

Albumin of catfish (*Clarias sp.*) can be detected by the presence of lumps through the heating process. then identified by the UV-Vis spectrophotometry method. The maximum wavelength in the standard solution and extract of catfish (*Clarias sp.*) Maximum absorption is obtained at a wavelength of 625 nm.

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