

Recovery And Valorization Of Snakehead Fish (Channa Striata) Surimi Wash Water As Stock Albumin Tablet

Ikbal Syukroni, Wini Trilaksani, Uju

Abstract: Surimi washing process is aimed to concentrate the myofibril protein by removing, cathepsin enzyme, fat, pigment, blood, and sarcoplasmic protein which is soluble in wash water. The soluble substances cause trouble environment if it was untreated. In addition recovery protein will give benefit both in reducing trouble environment and utilizing soluble protein as sources of albumin protein. The objectives of research were to recover albumin from snakehead fish surimi wash water and to valorize as stock albumin tablet. Recovery of albumin use 0.05 μm ultrafiltration membrane and the valorization of albumin tablets was by direct compression. The protein band with molecular weight of 67.741 kDa on the retentate was detected as albumin. Concentration of protein recover by ultrafiltration membrane increased 89.98% and the albumin content 3.5 \pm 0.4 g/dl. Based on the result of chemical composition and microbiology analysis, albumin of snakehead surimi wash water appropriate with Indonesia National Standard (SNI) quality requirement about snakehead fish albumin extract. The best formulation in the preparation of surimi wash water albumin tablet was by using corn starch excipients with uniformity weight value 410.39 \pm 0.09 g, hardness value 7.65 \pm 0.8 Kp, uniformity size of tablet with diameter 1 cm and thickness 0.59 cm, friability value 2.3% and disintegration time of the tablet is 2 minutes 16 second.

Key words: ,albumin, recovery, sarcoplasm, tablet, valorization

1. Introduction

Fishery processing industry will generate waste liquid that comes from the process of cutting, washing, and processing products. These fluids contain blood and pieces of small fish and fish entrails, skin, head of the fish that have no economic value (Bawono, 2015). Fishery waste, particularly liquid waste, has not been widely utilized. Waste liquid fishery that is without treatment will cause environmental pollution, because the content of organic compounds is very high. Waste water washing minced fish has a content over the limit threshold set by Reg-51/MoEF/10/1995, for some parameters including Chemical Oxygen Demand (COD), Biological Oxygen Demand (BOD), Total Suspended Solid (TSS) and Total Dissolved Solid (TDS). COD and TSS levels of surimi waste is higher compared to the wastewater processing of fillets, and the water used in rinsing and thawing of shrimp (Vandanjon et al., 2002). One of the fishery processing industry that produces the most liquid waste is surimi industry. Surimi is a minced fish which is washed to remove fat, blood, enzymes and proteins sarcoplasm and stabilized in frozen conditions with adding cryoprotectant (Balange & Benjakul, 2009). According Jaouen & Quemeneur (1992) large volumes of water for surimi 9-15 liters / kg. The raw material requirements to produce good quality in the manufacture of surimi including white flesh fish, low fat and have good gel properties (Park & Morrissey, 2000).

One types of fish that can be processed to surimi is Snakehead fish (*Channa striata*). According to stastical data of fisheries MoMAF RI (2015) it was seen from the composition by type of fish that the production volume of capture fisheries in openwaters in 2014 was dominated by snakehead fish, with total volume of production reached 39,030 tons (8.74%), followed by tilapia 28,637 tons (6.41%) and species of baung fish 27,157 tons (6.08%). The high volume of catch was not proportional to the utilization of snakehead fish. Some provinces such as south sumatera and south borneo utilizing snakehead fish as raw material of fishery products. Snakehead fish processing needs to be developed to increase the commercial value and shelf life, one of them as raw material in surimi industry. Snakehead fish is known to have a very high albumin content, one of the protein sarcoplasm or water soluble protein. Suzuki (1981) states that characteristic of the sarcoplasm protein are soluble in water and insoluble in salt or acid. Surimi washing process causes concentration of myofibril protein in surimi and increased sarcoplasmic protein, fat, pigment, blood, fats, and enzymes cathepsin dissolves with surimi washing water (Novanti, 2015). According to Putri & Agustina (2016) albumin has the advantage that is to accelerate healing of incision wounds on white rat. Valorization of water soluble protein fraction (sarcoplasm) from surimi washing water not only eliminating negative effects to the environment but also can increase the value added of surimi industrial waste. Previous research about protein recovery were conducted by Trilaksani et al. (2007) utilizing surimi waste water as edible film ; Bourtoom et al. (2009) get component of soluble water in surimi wash water with precipitation technique. Method of recovery has been done using absorption (Shukoor et al. 2007), precipitation (Kurinomaru et al., 2014), evaporation (Schuck et al., 2015), and filtration (Uju et al., 2009). Among these methods, ultrafiltration has an advantage because it does not require chemical material, sustainable, and remove impurity components (Kumar & Lawler, 2014). Membrane process keeps the quality of the compound concentrated eg protein, compared to heat or chemical process because it can be done on

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room temperature (Dumay et al., 2008). Based on the background, reinforcing the reason for obtaining extracted albumin by ultrafiltration technique from snakehead fish surimi wash water to make an albumin tablet stock.

2. Material and Methods

2.1. Material and Equipment

Materials used in this study were Snakehead fish (*Channa striata*) obtained from Anyar Market, Bogor. Materials used for manufacture surimi, albumin extract and tablet consists of aquadest, ice, Avicel 102, PVP, Corn Starch, Mg stearate, Aerosil, Talc, chemical reagents such as reagents Bromocresol green (BCG) and bovine serum albumin (BSA). Other ingredients including materials for chemical analysis of albumin's snakehead fish surimi wash water. The equipment used for the preparation and preparation of surimi manufacture, is among the polypropylene ultrafiltration membranes with an average pore size of 0.05 µm with membrane surface area of 1.8 m² and Deng Yuan KJ-2600 pump, tablet molding (Rimek mini pres- II), hardness tester (YD-3), moisture balance (AMB 50, UK), electric stove, waterbath, volume pipette, cuvet, oven.

2.2. Research Prosedure

2.2.1. Preparation of Snakehead surimi wash water

Preparation process of snakehead surimi wash water in this research based on Bourtoom method (2009). Fresh snakehead fish was prepared and filleted. Snakehead fish was chopped using a knife and grinded into using meat grinder, then snakehead's minced meat was washed with cold water 10°C with water and meat ratio of 3:1 (v/b). Washed meat squeezed the water using gauze, water washing then used as research material

2.2.2. Concentration and characterization of Albumin Snakehead Fish Surimi Wash Water

Pretreatment was done to recover albumin with other proteins and disable the pathogen that contaminated in snakehead fish surimi wash water by modifying method of Burnouf (1991). Surimi wash water was fed into the feed tank, then heated to a temperature of 60°C, following concentration of the protein refers to Benhabiles et al. (2013) with modifications to the transmembrane pressure used. Protein was concentrate using a polypropylene ultrafiltration membrane with an average pore size of 0.05µm. The result of the process at the concentration consists of two fractions, the fraction that can through the pores of the membrane is called permeate and fractions that can not be passed pores called retentate. 2L of surimi wash water inserted into a feed tank. The surimi wash water flowed towards membrane using a pump. Concentrating was carried out at the transmembrane pressure 0.5 Bar and 30°C. Concentrating was stopped when the retentate volume reaches 320ml or at a concentration factor of 6.25.

2.2.3. Formulation of Albumin Snakehead Surimi Wash Water Tablet

Powder extract of albumin was made using Avicel 102 as absorbent on each albumin extract with ratio (extract of albumin : Avicel 102) was 0.5:1 (v/w). The mixture was dried in an oven of 50°C for 1 hour until the moisture content is less than 5%. Formulations of extract albumin surimi wash water tablets made in 3 formulas with different types of excipients ie Formula 1 (Avicel 102), Formula 2 (PVP) and Formulation 3 (Corn starch) quantify effect on evaluation value of the tablet, while other additives remain. Tablets made were round with a weight of 410Mg per tablet. Tablet molding machine was prepared for a while, then granule mass put into tablet molding machine. After tablet has been printed, the evaluation of tablet was done by testing uniformity weight, hardness test, uniformity of size, friability test and disintegration time (Ministry of Health, 1995).

Table 1. Formulation of Snakehead fish surimi wash water stock albumin tablet

Component	Formulation		
	Formulation 1 (Avicel 102)	Formulation 2 (PVP)	Formulation 3 (Corn Strach)
Snakehead fish albumin extract	224mg	224mg	224mg
Avicel 102	44.6mg	-	-
PVP	-	44.6mg	-
Corn strach	-	-	44.6mg
Aerosil	1.5%	1.5%	1.5%
Talc	1%	1%	1%
Magnesium Stearat	0.5%	0.5%	0.5%

2.2.4. Analysis of Albumin Content (Bartholomew & Delaney, 1966)

Analysis of albumin content requires reagent of albumin that is Bromocresol Green (BCG), albumin standard solution prepared from Bovine Serum Albumin (BSA). Tests of albumin content were performed by measuring absorbance in the sample. Albumin extraction was taken as much as 0.5 ml then added 2.5 ml of BCG reagent 0.01% and left for 10 - 15 minutes. The mixture then is put into the

cuvette and measured absorbatation value at 636 nm wavelength

3. Result and Discussion

3.1. Chemical Composition and Microbiology Albumin Snakehead Surimi Wash Water

Chemical analysis results (proximate and heavy metals) and microbiology compared with terms of quality and safety of albumin extract (SNI, 2014) are presented in Table 2. Characteristics of albumin snakehead fish surimi wash water were examined from chemical composition and microbiology have fulfilled the requirement of albumin quality according to Indonesia National Standard (SNI) 8074:2014 about snakehead albumin extract (*Channa striata*). Albumin snakehead surimi wash water have high protein and albumin content. High content of protein on albumin snakehead fish surimi wash water caused by the fraction of water soluble protein from snakehead fish. Research of DeWitt & Morrissey (2001) describes some of the results of wash water, about 40-50g/100g of fish minced solids dissolve in wash water, resulting in wash water contains a water soluble protein fraction (sarcoplasm). High content of albumin on this study is caused by the concentrating by ultrafiltration membrane. According to Cheryan (1998) in the ultrafiltration process of pressure difference across the membrane ultrafiltration will force the smaller solvents and molecules through the membrane pores, while larger molecules will be retained and fed as retentate. Fish Serum Albumin (FSA) has a characteristic similar to *Human Serum Albumin* (HSA). Fish *Channa gachua* and *Channa gariepinus* (catfish from Africa) have albumin in the form of a monomer with a molecular weight of 70 kDa, in tune with *Human Serum Albumin* (HSA) on the secondary structure (Hasnain et al., 2004). In this research, the fat content of albumin is very low around 1.14% (dry basis). Snakehead Fish is a high-protein and low-fat fish (Osibona et al., 2009). In addition, membrane filtration process with ultrafiltration membrane as well reject the fat as described in Gringer et al. (2015) ultrafiltration membrane with an average pore size of 0.04 μm can reduce the content protein and fat in marinated herring (*Clupea harengus*) 76% and 100% respectively.

Table 2. Chemical and microbiology composition albumin of snakehead fish surimi wash water

Parameter	Unit	Albumin of Snakehead fish Surimi Wash Water	Indonesia National Standard (SNI)
Chemical Composition			
• Protein	%	77.15±0.13	Min. 70
• Albumin	%	35±0.4	Min. 15
• Fat	%	1.14±0.06	Maks. 8
• Zinc (Zn)	mg/kg	0.2498±0.04	Min. 1
• Iron (Fe)	mg/kg	0.293±0.01	Min. 0.3
• Calcium (Ca)	mg/kg	0.681±0.02	Min. 120
• Arsenic (As)	mg/kg	Not detected	Maks. 1
• Cadmium (Cd)	mg/kg	0.009±0.01	Maks. 0.1
• Lead (Pb)	mg/kg	0.191±0.024	Maks. 0.4
• Mercury (Hg)	mg/kg	No detected	Maks. 0.5
Microbiology			
• <i>Escherichia coli</i>	APM/g	Negative	Negative
• <i>Salmonella</i>	Per 25 g	Negative	Negative

Albumin snakehead fish surimi wash water contains essential minerals that have physiological functions for the body, namely zinc (Zn), Iron (Fe) and Calcium (Ca). According to Mustafa et al. (2012) essential minerals found in snakehead fish correlated with albumin that have function of maintaining the body's metabolism, integrity with some vitamins and with the immune system. Based on the results of heavy metal analysis and microbiology (Table 2) which showed that the heavy metal and microbiology in albumin snakehead surimi wash water is safely below the standard threshold set by Indonesia National Standard (SNI) for the product albumin snakehead fish extract therefore safe to be consumed.

3.2. Concentration of Sarcoplasm Protein Albumin Snakehead fish Surimi Wash Water

Concentrations of protein increased 89.98% after filtrated by Ultrafiltration membrane from 0.9997 % (surimi wash water) be 1.111% (retentate). Sotoft et al. (2015) explained that herring marinade concentration using 50 kDa ultrafiltration membrane can increase the protein concentration up 57.8%. The increasing in measured protein concentration because there has been a separation to the water content of the feed into the permeate whereas most the protein was retained as the retentate redirected to the feed tank, so higher protein concentration in the feed. The result of measurement of protein concentration albumin snakehead fish surimi wash water can be seen in table 3.

Table 3. Concentration of sarcoplasm protein snakehead fish surimi wash water

Sampel	Concentration of Protein (%)
Snakehead fish surimi wash water	0.990 ± 0.013
Retentate	1.111 ± 0.041

3.3. Protein Profile of Albumin Snakehead Fish Surimi Wash Water

Protein profile of albumin snakehead fish surimi wash water using SDS-PAGE method. Protein band with a molecular weight of 67,741 kDa (1) in the retentate detected as albumin with type quite similar Human Serum Albumin (HSA) (Denizli, 2011). Results of protein profile analysis presented in Fig. 2. There are different protein profiles in the feed, retentate, and permeate. Protein profile on retentate less and thin. This can be caused low molecular weight proteins in retentate have passed through the ultrafiltration membrane. Based on Cheryan (1998) in the process ultrafiltration, pressure difference across the ultrafiltration membrane will force the solvent and smaller molecules pass through membrane pores, while large molecules will be retained and flowed as retentate. It was proven on permeate, which only found very thin bands of protein (2) with a molecular weight of 37.178 kDa. Reduced number of bands protein on permeate shows a protein with a molecular weight above 50 kDa does not pass through the ultrafiltration membranes used. Ultrafiltration membranes have been rejected protein with a minimum molecular weight of 50 kDa, as evidenced by the thickness protein bands on the permeate which is thicker than the retentate.

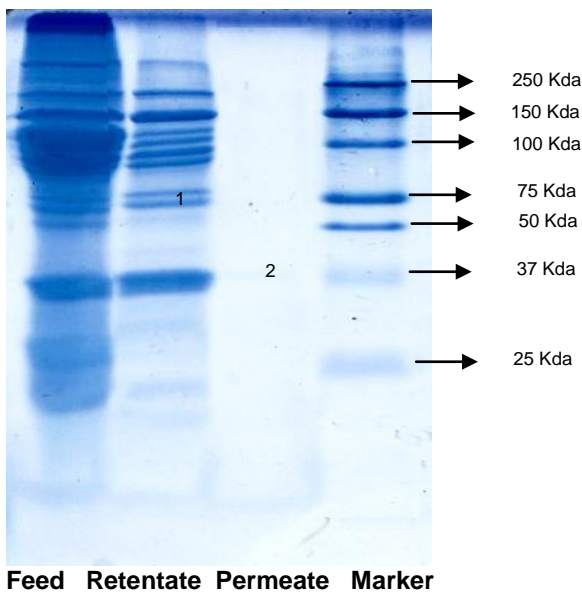


Figure 2. Molecular weight electrophoresis of albumin snakehead fish surimi wash water

3.4. Amino Acid of Albumin Snakehead Fish Surimi Wash Water

The composition of the amino acid albumin of snakehead fish surimi wash water was presented in Table 1. Based on the results of the study, it was found that the cucumber fish wash water albumin contained amino acid L-Glutamic acid, L-Aspartic acid, L-Proline and L-Leucine most. Nugroho (2014) explains that in the body of snakehead fish contains amino acid L-Glutamate Acid and L-Aspartate is very high followed by several other essential amino acids.

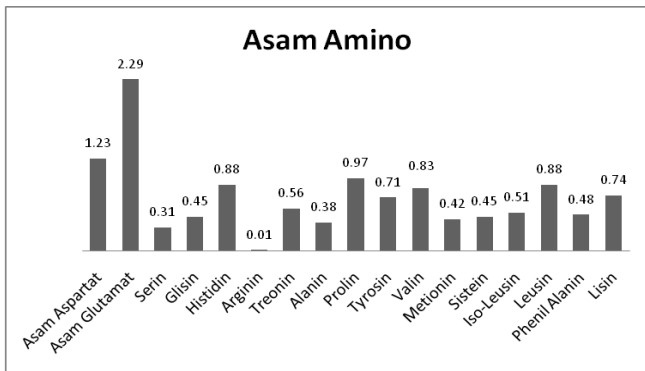


Figure 1. Composition of the amino acid albumin of snakehead fish surimi wash water

3.5. Evaluation of Albumin Snakehead Fish Surimi Wash Water Tablet

Evaluation of Tablet	Formulation				Standard of Tablet (Ministry of Health 1995)
	F0	F1	F2	F3	
Weight Uniformity (g)	369.86 ± 0.18	410.14 ± 0.16	410.45 ± 0.11	410.39 ± 0.09	5% from tablet's weight
Size Uniformity a. Diameter (cm)	1	1	1	1	3x Thickness of tablet
b. Thickness(cm)	0.59	0.59	0.59	0.59	1 ^{1/3} thickness of tablet
Hardness (Kp)	6.36 ± 0.38	8.1 ± 1.4	18.49 ± 1.51	7.65 ± 0.8	4-8 Kp
Friability (%)	2.6%	0%	0%	2.3%	<1%
Disintegration(minute)	>15 minute	4 minute 25 second	4 minute 12 second	4 minute 16 second	< 15 minute

Table 4. Evaluation value of snakehead fish surimi albumin tablet

3.5.1. Uniformity Weight of Tablet

Based on evaluation of albumin tablets presented in Table 4, the uniformity weight of each tablet in all tested formulas was eligible uniformity weight of tablet because there are no two tablets of each formula, that has weight deviation exceed 5% and there was no single tablet that weight deviate from an average weight greater than 10% (Ministry of Health, 1995). Uniformity weight affects the uniformity of the dosage and the dose of the drug to achieve therapeutic goal (Lieberman et al., 1989). Uniform weights can be influenced by the flow properties of the excipient, if have a good flow properties, material will fill print space well during tablet forming process. Analysis of uniformity weight used for solid preparations that contain one or more active substances (Ministry of Health, 1995). Based on statistic analysis with One Way ANOVA test on α=0.05, results showed that the treatment of excipient differences on each formulation has a significant effect on weight uniformity on tablets of albumin snakehead surimi wash water.

3.5.2. Hardness Test of Tablet

Hardness test on tablets has been conducted to determine about compactness and resistance of the tablet when mechanic pressurized. Requirements of hardness value according to Ministry of Health (1995) is 4-8 Kp per tablet. Based on the results of research, tablet formulas with excipients Avicel 102 and Corn Strach have ideal hardness requirements of tablets with hardness value each other 8.1 ± 1.4 Kp and 7.65 ± 0.8 Kp. The tablet formula with the addition of PVP excipient with a value of 18.49 ± 1.51, due to the excipient PVP as a binder having properties as adhesives which is good in water or alcohol solvent, PVP also has the ability as dry binder that has good consistency (Herawati et al., 2014). Based on statistic analysis with One Way ANOVA test α=0.05, the results showed that the treatment of excipient differences in each formulation gives effect significant to hardness value on each tablet of albumin snakehead fish surimi wash water.

3.5.3. Uniformity Size of Tablet

The uniformity size test on tablet of albumin snakehead fish surimi wash water showed that formulation 1, formulation 2 and formulation 3 has a diameter of 1 cm tablet and 0.59 cm tablet thickness. The results revealed that three tablets of albumin snakehead fish surimi wash water were eligible uniformity of tablet size that was tablet diameter not more than 3 times thick tablet and not less than 1 1/3 times thicker tablets. The thickness of the tablet was influenced by several things including pressure at the time of printing the tablet, the amount of mass that was loaded in the tablet's print space and mass density of printed tablets (Lachman et al., 1994) where as tablet diameter was affected by the size of the tablet's print space (Voight, 1994).

3.5.4. Friability Test of Tablet

Based on the results of friability test, formulation 1 and formulation 2 had a better percentage of friability than formulation 3 which is have not requirements of friability tablet that is minimum value of friability does not exceed 1% (Ministry of Health, 1995). Difference percentage of tablet friability was affected by hardness value, higher hardness value of the tablets causes bonding between the particles on tablet more powerful, so that have smaller percentage friability of tablet (Rori et al., 2016). Tablet friability may also be affected by the nature of the excipient and the amount of excipient in the formulation tablet, according to Agoes (2008) Avicel 102 and PVP are excipient types has good compatibility value against the tablet so it has an effect on tablet friability

3.5.5. Disintegration Time of Tablet

Disintegration time test is important to know soluble time of tablet. To provide a therapeutic effect, the tablet must be destroyed into smaller particles and can be absorbed in the gastrointestinal tract. Result of the disintegration time test showed that all formulation was eligible for the crushed timing test of the Ministry of Health (1995) that is disintegration time under 15 minutes. This phenomenal affected by excipients, in this research formulation tablet of albumin surimi wash water by using corn starch has fast enough. According Rahayuningsih (2010) excipients with materials starch can accelerate water absorption so it will be possible to draw water rapidly through capillary work.

4. Conclusion

Based on the result of chemical composition and microbiology analysis, albumin of snakehead surimi wash water appropriate with Indonesia National Standard (SNI) quality requirement about snakehead fish albumin extract. Concentration protein after concentration using ultrafiltration membrane increases and electrophoresis results detecting protein in albumin with band protein indication that is similar with a type of Human Serum Albumin (HSA). The best formulation is the types of excipient corn starch with the value of uniformity weights, hardness value, value of uniformity size, friability value and disintegration time inside quality requirement of tablet Ministry of Health Indonesia.

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