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Characterization of Snakehead Fish (*Channa striata*) Meat Simplicia as Raw Material for Wound Healing Drugs

(*KARAKTERISASI SIMPLISIA DAGING IKAN GABUS (CHANNA STRIATA) SEBAGAI BAHAN BAKU SEDIAAN OBAT PENYEMBUHAN LUKA*)

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ABSTRACT

Characterization is the initial stage of standardization to determine the quality of simplicia. The quality of simplicia is influenced by raw materials, manufacturing process, the storage. Snakehead fish from natural ingredients has been shown to contain nutrients that can accelerate wound healing. This study aims to determine the characterization of snakehead fish (*Channa striata*) and snakehead fish meat simplicia originating from Sungai Itik Village, Kubu Raya Regency, West Kalimantan as raw material for wound healing drug preparations. Characterization was carried out biologically including habitat and morphology, physically including organoleptic, water content, ash content, and acid insoluble ash content, and chemically including protein and albumin qualitative tests. The results of the characterization of snakehead fish come from the waters around the Itik River with environmental conditions that are still not polluted. The head shape of the snakehead fish is like a snakehead, the body is blackish brown with dark band-shaped rows (*striata*), the belly is white to yellowish-white, and there is a labyrinth. The results of simplicia characterization of snakehead fish meat had a water content of 81.6% and 8.97%, ash content of 0.813% and 5.17%, and acid insoluble ash content of 0.02%. The results of the qualitative test of protein and albumin were declared positive for containing protein and albumin. Based on the results of the characterization that has been carried out, it can be obtained snakehead fish meat simplicia with good quality.

Keywords: characterization, protein, simplicia, snakehead fish (*Channa striata*)

ABSTRAK

Karakterisasi merupakan tahapan awal dari standardisasi untuk mengetahui kualitas dan mutu simplisia. Kualitas dan mutu simplisia dipengaruhi oleh faktor bahan baku, proses pembuatan hingga penyimpanan bahan baku simplisia. Ikan gabus dari bahan alam telah terbukti mengandung nutrisi yang dapat mempercepat penyembuhan luka. Penelitian ini bertujuan untuk mengetahui karakterisasi ikan gabus (*Channa striata*) dan simplisia daging ikan gabus yang berasal dari Desa Sungai Itik, Kabupaten Kubu Raya, Kalimantan Barat sebagai bahan baku sediaan obat penyembuhan luka. Karakterisasi dilakukan secara biologi yang meliputi habitat dan morfologi, secara fisika meliputi organoleptis, kadar air, kadar abu, dan kadar abu tidak larut asam, serta secara kimia meliputi uji kualitatif protein dan albumin. Hasil karakterisasi ikan gabus berasal dari perairan disekitar aliran Sungai Itik dengan kondisi lingkungan yang masih tidak tercemar. Bentuk kepala ikan gabus seperti ular (*snakehead*), tubuh bewarna cokelat kehitaman dengan barisan gelap berbentuk pita (*striata*), perut bewarna putih hingga putih kekuningan, dan terdapat labirin. Hasil karakterisasi simplisia daging ikan gabus memiliki kadar air sebesar 81,6% dan 8,97%, kadar abu sebesar 0,813% dan 5,17% serta kadar abu tidak larut asam sebesar 0,02%. Hasil uji kualitatif protein dan albumin dinyatakan positif mengandung protein dan albumin. Berdasarkan hasil karakterisasi yang telah dilakukan, dapat diperoleh simplisia daging ikan gabus (*Channa striata*) dengan mutu dan kualitas yang baik.

Kata-kata kunci: ikan gabus (*Channa striata*), karakterisasi, protein, simplisia

INTRODUCTION

Wound healing is a natural phenomenon of the body's repair process in overcoming tissue damage to the skin (Prayugo *et al*., 2021). The natural rate of wound healing is very slow and the chance of being infected with microbes is very high. Therefore, high enough nutrients are needed to accelerate the wound healing process. Snakehead fish **(*Channa striata*)** from natural ingredients has been shown to contain nutrients that can accelerate wound healing because it is safer and more effective (Andrie and Sihombing, 2017). Based on previous research, it was shown that snakehead fish ointment has effectiveness on wound healing (Daisa *et al*., 2017).

Snakehead fish (*C. striata*) is a predatory fish that lives in freshwater native to Indonesia (Sumatra, Java, and Kalimantan). Snakehead fish live in rivers, lakes, rice fields, and swamps (Haryanto, 2019). Snakehead fish has biomedical benefits such as antimicrobial, anti-inflammatory, anti-nociceptive pain, and anti-cancer properties (Alviodinasyari *et al*., 2019). Snakehead fish contains protein compounds that are higher than other types of fish. The protein content of snakehead fish can reach 25.5% higher than sardines (21.1%), milkfish (20%), snapper (20%), and goldfish (16%) (Alfarisy, 2014). Snakehead fish contains protein with albumin as the main content of about 6.22%, amino acids, fatty acids, and minerals such as Zn, Cu, Ca, and Fe (Sulfitri *et al*., 2020).

Snakehead fish albumin is effective in helping the healing process of postoperative wounds, burns, internal and external wounds, and helps increase albumin levels and body resistance (Asikin and Kusumaningrum, 2017). Amino acids in snakehead fish have a good ability in the synthesis of collagen fibers, proliferation fibroblasts, and source of energy for cell proliferation thereby accelerating re-epithelialization of the wound healing process (Karliman *et al*., 2021). Fatty acids play a role in inhibiting inflammation by reducing leukocytes. Minerals in snakehead fish play a role in cell proliferation, blood vessel growth factors, and oxygen affinity so that the remodeling process occurs and accelerates wound closure (Saputra *et al*., 2019). The magnitude benefits of the bioactive compounds in snakehead fish on the wound healing process, and the simplicia of snakehead fish need to be characterized.

Characterization has the aim of knowing the specifications of the simplicia to be studied. Specifications are carried out to determine the clarity of the material under study because the origin of the environment in which they live affects the content of the active compound (Handayani *et al*., 2017). The quality of snakehead fish meat simplicia is influenced by raw material, manufacturing process, and storage (Pujiasmanto *et al.*, 2021). Length, weight, food, and habitat are also factors that can affect the quality of snakehead fish meat simplicia (Alfarisy, 2014).

Characterization is the initial stage to determine the quality of simplicia so that the results of the characterization can be used as a reference in future development (Supriningrum *et al*., 2020). Based on the results of the existing literature search, so far research related to the characterization of snakehead fish (*C. striata*) and snakehead fish meat simplicia, especially in Sungai Itik Village, Kubu Raya Regency of West Kalimantan, Indonesia, has never been carried out. Therefore, researchers are interested in conducting this study to obtain the characteristics of snakehead fish and snakehead fish meat simplicia so that it is expected to produce good quality snakehead fish meat simplicia. The characterization in this study was based on biological aspects (habitat and morphology), physical (organoleptic test, moisture content test, ash content, and acid insoluble ash content), and chemistry (protein qualitative test and albumin qualitative test). Snakehead fish samples were obtained from Parit Tuampe RT 013 / RW 005, Cempaka Hamlet, Sungai Itik Village, Sungai Kakap District, Kubu Raya Regency, West Kalimantan, Indonesia.

RESEARCH METHODS

The materials used in this study consisted of several groups; the manufacture of snakehead fish meat simplicia using snakehead fish (*C. striata*) obtained from Sungai Itik Village, Sungai Kakap District, Kubu Raya Regency, West Kalimantan, Indonesia. In testing the water content, ash content, and acid insoluble ash content using snakehead fish meat simplicia, 25 mL of dilute HCl (HCl 0.2 N), and distilled water. Qualitative testing of protein and albumin includes snakehead fish meat simplicia, aquades, and biuret reagent (*Indo Reagen*®). The tools used in this study consisted of several groups, including making simplicia using knives, cutting boards, scissors, basins, scales (*Kris Chef*®) , packaging containers, and freezer boxes (*AQUA Chest Freezer-200 Liter*®). The

water content test includes a closed weighing bottle, desiccator, oven, and analytical balance. The ash content test includes porcelain dishes, electric furnaces, and analytical balances. Tools for acid-insoluble ash content tests include water bath, electric furnace, filter paper, and porcelain dishes. Qualitative testing of protein and albumin includes test tubes (*PYREX*®), tube clamps, 100 mL, and 500 mL beakers (*PYREX*®), 10 mL measuring cups (*PYREX*®), dropper, analytical balance (*OHAUS SCOUT*®), filter, thermometer, mortar and stamper and *magnetic stirrer* (*MAGNETIC STIRRER REXIM RSH-1DR*®).

Figure 1. Snakehead fish habitat

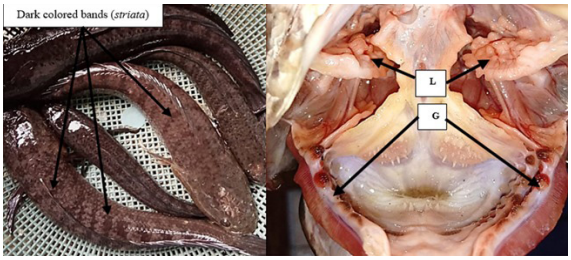

Figure 2. Snakehead fish morphology, L= labyrinth; G= gill

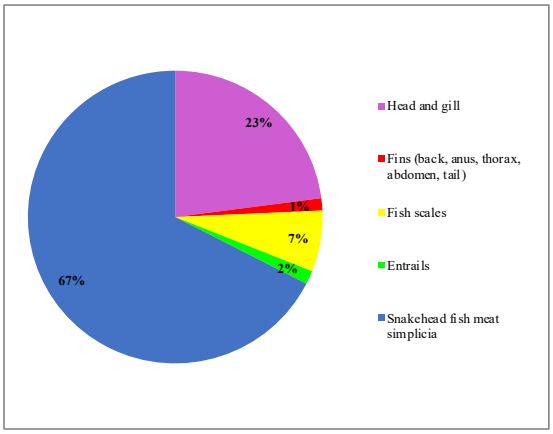

Figure 3. Snakehead fish yield percentage diagram

Characterization begins with observing the habitat of snakehead fish that lives in the wild including the origin of the waters (troughs, rivers, river estuaries, or lakes), water conditions, and environmental conditions. around the waters. Morphological observations of snakehead fish include a snakehead fish head shape (*snakehead fish*), the front body shape is almost round and flat straight towards the back, brownish-black in color with a combination of dark-colored rows in the form of ribbons (*striata*) (Sinaga *et al*., 2019). Snakehead fish have five fins consisting of dorsal fins, pectoral fins, pelvic fins, anal fins, and tail fins (Shasia *et al*., 2021). Snakehead fish have a respiratory system consisting of gills and a labyrinth (Pertiwi *et al*., 2017). Furthermore, the determination of the snakehead fish is carried out to ensure the truth regarding the clarity of the snakehead fish's identity. Snakehead fish was determined at the Biology Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Tanjungpura University, Pontianak, West Kalimantan.

The collection of snakehead fish samples were obtained from the results of fishing. The snakehead fish sample was selected with the criteria of a length of 30-50 cm and a weight of 500-1000 g/head. Next, sorting is done by separating the live snakehead fish from the dead snakehead fish. The live snakehead fish were selected with the criteria of body condition such as were the fish scales intact and were there any defects, were the fish's body looked bright and shiny. Dead snakehead fish were selected with criteria such as live snakehead fish, only that there were additional criteria organoleptically with the characteristics of appearance, flesh texture, and smell (SNI, 2013). The live snakehead fish are sacrficed and a weeding process is carried out which includes removing the entrails, scales, fins, and tail. After that, the snakehead fish was washed using running water until it was clean. The snakehead fish was cut into three parts, then each was weighed as much as 500 g. Fish are packed and labeled. Store fish in the *freezer* at a temperature of -20 °C to -10 °C (Kemenkes RI, 2017).

Characterization of snakehead fish (*C. striata*) meat simplicia includes organoleptic, water content, ash content, acid insoluble ash content, and the qualitative test of protein and albumin. Tests for water content, ash content, and acid insoluble ash content were carried out by sending samples to the Laboratory of Research and Industrial Standardization of Pontianak

City, West Kalimantan, and at the Integrated Research and Testing Laboratory Gadjah Mada University, Yogyakarta. Qualitative tests of protein and albumin were carried out at the Laboratory of Pharmaceutical Chemistry, Department of Pharmacy, Faculty of Medicine, Tanjungpura University, Pontianak City, West Kalimantan.

Organoleptic test includes characteristics of appearance, smell, and texture (SNI, 2013). Testing the water content of snakehead fish meat simplicia using the gravimetric method by weighing the sample in a closed bottle (weight is known), then drying it in an oven at 105 °C for three hours. Let it cool in the desiccator, weighs, and repeat this work until a constant weight is obtained (SNI, 1992). The ash content test uses the gravimetric method by weighing the sample into a porcelain dish (weight is known). Charcoal over the flame of the burner, then ash in an electric furnace at a temperature of 600 °C for eight hours until a constant weight is obtained. Let cool in the desiccator, then weigh until constant weight (SNI, 1992). Measurement of acid-insoluble ash content using the gravimetric method in which the ash obtained from the determination of the total ash content was boiled in 25 mL of dilute HCl (HCl 0.2 N) for five minutes. The acid-insoluble portion was filtered through ash-free filter paper. After that, wash with hot water and incandescent until the weight remains. Acid insoluble ash content is calculated in %w/w of the dried material (SNI, 1992).

A Protein qualitative test was carried out with the biuret method. Qualitative test of albumin using the heating method. The snakehead fish meat simplicia as much as 25 g was mashed using a mortar and stamper. Add 5 mL of distilled water and grind the snakehead fish meat until it is mixed with aquadest. After that, separate the fish meat from the filtrate using a filter. Put 2 mL of the filtrate into the test tube. For the qualitative test of protein, 3-5 drops of "*Indo Reagent*®" into the test tube, and homogenized. A positive reaction is if a red violet or blue-violet color is formed (Purnama *et al*., 2019). For the qualitative test, albumin was filled in a 500 mL beaker with 250 mL of distilled water and placed in the water bath. Heat the sample at 70-90 °C for 30 minutes. The Positive reaction of albumin is indicated by the formation of the white lump (Taurina and Andrie, 2017).

RESULTS AND DISCUSSION

Habitat

Based on the results of observations and interviews conducted in Sungai Itik Village, Kubu Raya Regency, West Kalimantan, snakehead fish originated from dike ditches and creeks around the water flow of the Itik River which is overgrown by aquatic plants such as water *Eichhornia crassipes* and *Mimosa pudica*. The environmental conditions around the Itik River are still preserved in there are no industrial factories whose waste can pollute the river water. According to SNI 8074:2014, snakehead fish must come from unpolluted water (SNI, 2014).

Morphology

Snakehead fish (*C. striata*) was observed, including the shape of a snake-like head (*snakehead*), blackish-brown in color with a combination of dark-colored bands (*striata*), and a yellowish-white belly. The front of the body is almost round and the back at flatter (compressed). There are five fins, namely the dorsal fin, pectoral fin, pelvic fin, anal fin, and tail fin. The dorsal and anal fins are long and broad. The snakehead fish labyrinth is located at the top of the gills which serves to breathe air from the atmosphere (Pertiwi *et al*., 2017).

Determination of Sample

Determination was carried out to ensure the truth regarding the identity of the snakehead fish. In addition, determination also aims to avoid errors when sampling snakehead fish which is studied and used as raw material in research (Handayani *et al*., 2017). Results of determination carried out at the Biology Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Tanjungpura University, Pontianak City, West Kalimantan, stated that the snakehead fish species was *C. striata*.

Sample Collection

Snakehead fish is obtained from fishing in Itik River Village, Kubu Raya Regency, West Kalimantan. Snakehead fish fishing is done in the afternoon and evening. The snakehead fish selected from the fishing results are fresh in living conditions with a length of 30-50 cm and a weight of 500-1000 g/head. According to SNI (2014), the minimum weight of snakehead fish is 500 g/head. The results of research by Suwandi *et al*. (2014) showed that the highest protein content of 20.4% was found in snakehead fish

weighing 1 kg. According to Chasanah (2015), natural snakehead fish with a length of 32-41 cm contains a protein content of 19.85%. The results of Asikin *et al*. (2018) research stated that medium-sized snakehead fish (600-900 g) produced protein extracts with the highest albumin content compared to small snakehead fish (300-600 g) and large snakehead fish (901-1200 g) (Asikin and Kusumaningrum, 2018).

Simplicia Preparation

Sorting is done to separate the live snakehead fish from the dead snakehead fish. Weeding is done by removing the head, entrails, scales, fins, and tail. After that, it was washed using running water until it was clean. The goal is to clean the impurities that might stick. The simplicia used is snakehead fish meat accompanied by bones and skin. It is known that the weight of one snakehead fish is 742 g which are divided into 170 g of head and gills, 10 g for the fins (top, bottom, chest, anus) and tail, 50 g of scales, 11 g of stomach contents, and 500 g of snakehead fish meat simplicia. From these results, the percentage yield of one snakehead fish resulted in a simplicia of snakehead fish meat of 67.38%. The next step is to cut the snakehead fish meat simplicia into three parts, then weigh as much as 500 g. The goal is to facilitate the process of taking snakehead fish meat simplicia which will be made into raw material for wound healing drug preparations. The snakehead fish meat simplicia is put in a package, labeled, and stored in the *freezer* at temperatures of -20 ^{o}C to -10 ^{o}C (Kemenkes RI, 2017). Storage of snakehead fish meat simplicia in the *freezer* aims to preserve the fish so that microorganisms and enzymes that cause spoilage can be deactivated to prevent damage to simplica.

Table 1. Test results for water content, ash content, and acid insoluble ash

Test parameters	LPPT	BARISTAND
Water content	8.97%	81.6%
Ash content	5.17%	0.813%
Acid insoluble ash	0.02%	-

Description: LPPT= sample in the form of snakehead fish meat simplicia that has been freeze dried (in powder form); BARISTAND= sample in the form of fresh snakehead fish meat simplicia

Table 2. Protein and albumin qualitative test results

Test parameters	Methods	Results
Protein	Biuret	+
Albumin	Heating	+

Description: positive (+); negative (-)

Organoleptic

The organoleptic examination aims to provide an initial introduction to simplicia using the five senses produced simply and subjective (Depkes RI, 2000). Organoleptic examination of fish using the five senses is the most common and easy way to measure the freshness and quality of fish (Nurjanah and Abdullah, 2019). The organoleptic examination of snakehead fish showed that the eyes of snakehead fish were convex and bright, the eye membranes were clear and not cloudy, and the pupils were black. The gills are bright red with a little mucus. The smell is very strong specific to the type of fish. The scales are firmly attached to the skin. Mucus on the body surface is clear, thin, shiny, and clear in color. The flesh is brilliant white, shiny, and elastic.

Moisture Content

Water is one of the important components because water can affect the appearance, freshness, and texture of simplicia. A water content test is carried out to determine the amount of water in simplicia (Umage *et al*., 2019). Based on the test results at the Laboratory of Research and Industrial Standardization of Pontianak, West Kalimantan, the water content of fresh snakehead fish meat simplicia is 81.6%. Research by Suwandi *et al*. (2014) and Ahmed *et al*. (2012) also showed that the water content of snakehead fish was 80.41% and 82.66%, respectively (Ahmed *et al.*, 2012; Suwandi *et al*., 2014). Snakehead fish that comes from nature has high water content (Suwandi *et al*., 2014; Chasanah *et al.*, 2015). The composition of fish contains 65-80% water. The water content is also influenced by the type of food and the habitat of snakehead fish (Asikin and Kusumaningrum, 2017).

Based on results at the Integrated Research and Testing Laboratory (LPPT) Gadjah Mada University, Yogyakarta showed the water content of dried powdered snakehead fish meat simplicia was 8.97%. Wirawan *et al*. (2018) in their research stated that the chemical characteristics in testing the water content of

snakehead fish meal had met the standards set by the *Food and Agriculture Organization* (FAO) of 10% (Wirawan *et al*., 2018). This means that the simplicia of snakehead fish powder is resistant to being stored for a long time and tends to avoid fungal and bacterial contamination (Utami *et al.*, 2017).

Ash Content

Ash content test aims to see the internal and external mineral content and the influence of the manufacturing process from the beginning to the formation of simplicia (Sarnia *et al*., 2018). Based on the test results at the Laboratory of Research and Industrial Standardization Institute Pontianak, West Kalimantan, and the Integrated Research and Testing Laboratory Gadjah Mada University, Yogyakarta, the ash content of snakehead fish meat simplicia was 0.813% and 5.17%, respectively. That is, the remaining inorganic mineral content contained in the simplicia of snakehead fish is 0.813% and 5.17% (Rosidah *et al.*, 2020). Prasetyo *et al.* (2012) stated that the ash content of snakehead fish of 4.5%. The results of research by Suwandi *et al*. (2014) also obtained the ash content of snakehead fish of 7.50%. The ash content contained in snakehead fish is influenced by the mineral content found in the type of food and living habitat of the fish (Suwandi et al., 2014).

Acid Insoluble

Ash content test for acid-insoluble ash aims to determine the amount of ash originating from external factors sourced such as impurities originating sand or silicate soil (Salim *et al*., 2016). The insoluble ash content of snakehead fish meat simplicia acid was 0.02%. Acid insoluble ash content reflects the presence of metal contamination that is not acid soluble in a simplicia. High levels of insoluble ash in acid indicate the presence of silicate content from soil or sand, soil, and metallic elements silver, lead, and mercury due to environmental contaminants (Evifania *et al*., 2020).

Protein Qualitative Test

Snakehead fish protein contains main nutrients consisting of protein and fat of 78% and 2%, respectively (Prayugo *et al*., 2021). Snakehead fish protein content can reach 25.5% (Alfarisy, 2014). Qualitative protein test should be done to determine whether there is the protein content in snakehead fish meat simplicia (Purnama *et al*., 2019). Test of qualitative protein indicated that in snakehead fish meat simplicia showed positive results indicated by a change in the color of the sample to purple. The color change occurs because copper/Cu^{2+} from the biuret reagent in alkaline conditions reacts with polypeptides on proteins to form purple complex compounds (Sulfitri *et al*., 2020).

Albumin Qualitative Test

Snakehead fish meat contains 70% and 21% protein and albumin, respectively (Kordi, 2010). Snakehead fish contains protein with albumin as the main content of about 6.22% (Sulfitri *et al*., 2020). A qualitative test of albumin was carried out to identify the presence of albumin chemically through precipitation by heating (Jamaluddin *et al.*, 2020). The results showed positive results indicated by the formation of precipitate after a sample was heated. The precipitate was formed due to excess heat treatment of albumin resulting in an irreversible reaction, it can be seen by the increase in water-insoluble protein by forming a precipitate.

CONCLUSION

The characterization of snakehead fish meat simplicia consisted of organoleptic, water content, ash content, acid insoluble ash content, the qualitative test of protein, and albumin. The organoleptic of snakehead fish meat simplicia is elastic flesh texture, brilliant white flesh color, and has a distinctive and strong odor specific to the type of fish. The water content is 81.6% and 8.97%, the ash content is 0.813% and 5.17% and the acid insoluble ash content is 0.02%. The results of the protein qualitative test and albumin were positive for containing protein and albumin.

SUGGESTION

Further research is necessary to characterize snakehead fish (*C. striata*) extract. Characterization of snakehead fish extract can include several parameters, namely assays for protein, albumin, amino acids, fatty acid, and minerals (Zn, Cu, Ca, Fe) levels. The heavy metal content test includes arsenic (Ar), cadmium (Cd), and lead (Pb). Microbial contamination test includes *Escherichia coli* and *Salmonella*.

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