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Minerals and Heavy Metals of Banana Puffer Fish from Sea of Region Gebang, Cirebon, West Java

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Abstract

This banana puffer fish empirically very liked by fishermen in the area Gebang, Cirebon. The purpose of this study was to determine the chemical composition and to identified heavy metals and minerals of its meat, viscera and skin. The meat had greater contain of protein and moisture than that of the skin and viscera. The chemical composition of the banana puffer fish was analyzed using AOAC method for proximate content, minerals and heavy metals were analyzed by atomic absorption spectrophotometer (AAS) of Shimadzu AA-7000. The fat and carbohydrate content were hightest in viscera, amounting to 36.83% (db) and 20.27% (db). The viscera had ash content of 11.66% (db) and the value was greater than that of meat and skin. The highest macro mineral content in the meat and the skin was K (1,469.34 ppm and 495.32 ppm), while the viscera were Mg (13,607.48 ppm). The viscera macro mineral content higher than meat and skin. Micro minerals dominant in the meat and the skin was Zn (73.63 ppm and 41.80 ppm), while the viscera were Fe (167.03 ppm). The meat and viscera contained Pb which exceeds the allowable limit, as well Cd in the viscera.

Keywords

Banana Puffer Fish, Heavy Metals, Minerals, Proximate

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1. Introduction

Banana Puffer fish (*Lagochepalus Lunaris*), which is carnivorous, is a slow swimmer fish species. This fish can be found almost all over the Indonesia waters. In fact, it can live in different ecosystems--seas, estuaries and freshwater areas (Wahyuni *et al*. 2004). In Japan, this fish is known as *dokusabafugu*, and it is one of the puffer species often consumed in Japan besides puffer fish called *Takifugu rubripes*. According to Nurjanah *et al.* (2014), empirically banana Puffer fish becomes the favorite of the fishermen in Gebang, Cirebon Regency.

Noguchi and Arakawa (2008) stated that the fish had a high level of toxicity in the muscle tissue that was more than 1000 MU / g, while the fish skin had a toxicity level between 100-1000 MU / g. A low toxicity level was found in the fish liver, which was less than 10 MU / g, but the blood of banana puffer did not contain tetrodotoxin. One MU (mouse unit) was defined as the amount of toxin required to kill the male rats (20 g) within 30 minutes after intraperitoneal administration, and the minimum lethal dose (MLD) for humans was estimated to be around 10000 MU (≈ 2 mg).

The economic value of banana puffer fish in Indonesia is not very high due to the people's lack of knowledge concerning the management and utilization, especially since the fish is considered as deadly toxic fish. Banana Puffer fish is so delicious and nutritious that in Japan the fish is a highly priced commodity (Noviyanti 2004). The fishermen in Gebang, Cirebon, eat banana puffer fish with fried.

Minerals play an important role in maintaining body functions because they set enzyme work, maintain acid-base balance, and help a bond formation (hemoglobin formation)

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(Almatsier 2004). Essential mineral elements in the body consist of two categories: macro minerals and micro minerals. The macro minerals are necessary for the formation of body organ components, while the micro minerals are needed in very small amounts which are generally found in tissues with very small concentrations (Arifin 2008).

Heavy metals are natural components contained in the earth's crust that cannot be degraded or destroyed. They are dangerous substances because their bioaccumulation and toxicity can threaten living organisms (Agustina 2010). Permatasari (2006) stated that the metal could not be separated from daily life because it is used in agriculture, medicine, and industry. However, metals can be harmful to health if present in food or water with excessive concentration. Since the information on the content of heavy metals and minerals found in banana puffer fish in Gebang, Cirebon, was not available, this study was conducted to determine the chemical composition, the residues of mineral and heavy metal in the meat, viscera and skin of banana puffer fish.

2. Materials and Methods

2.1. Materials and Tools

The main material used in this study was banana puffer fish (*Lagochepalus Lunaris*) obtained from the sea of Gebang, Cirebon, West Java. The materials needed for proximate analysis were fat solvents such as hexane, selenium, concentrated H2SO4 merck KGaA, distilled water, NaOH 40% merck KGaA, boric acid (H3BO3) 2% merck KGaA, indicator of Bromcresol Green-Methyl Red merck KGaA in pink color, N HCl solution 0.1 merck KGaA. The analysis of minerals and heavy metals used an apparatus called atomic absorption spectrophotometer (AAS) of Shimadzu AA-7000.

2.2. Research Methods

The raw material used in this study (banana puffer fish) were obtained from the waters of Gebang, Cirebon Regency. The samples were delivered in a styrofoam box containing ice, shipped to Bogor (IPB) for about 5 hours. The samples arrived at the laboratory in a fresh condition, after which the morphometric measurement and preparation and were directly conducted. Collectig of banana puffer fish sample from course by product is by using the fishermen payang at 3 a.m until 2 p.m. Measurement of banana puffer fish is used about 9 sampel of fish and is done 3 times repetitions with the lenght of 13 to 15.5 cm which had a juvenile size.

The preparation aimed to ease the process of separating the fish meat, viscera and skin, followed by weighing and measuring the rendemen**t**. The proximate analysis was performed on the samples of meat, viscera and skin with AOAC method 2005. The analysis of mineral and heavy metals was carried out using atomic absorption spectrophotometer (AAS) of Shimadzu AA-7000. The samples to be tested in an effort to find out the mineral content first had to go through wet ash process. The wet ash process was carried out with a sample of 1 g which was put in 125 mL Erlenmeyer, added by 5 mL HNO3, and allowed without any treatment for 1 hour. The flask was placed on a hotplate at a temperature of 80-100°C for 4-6 hours, added by 0.4 mL of concentrated H2SO4 and mixed with 3 drops of $HClO_4$ and HNO_3 until the color became light yellow. The samples were cooled and added 2 mL of distilled water and 0.6 mL of concentrated HCl. The sample solution was then diluted to 100 mL using distilled water in the flask. The standard solution, blank, and sample were flowed into the Atomic Absorption Spectrophotometer (AAS) of Shimadzu AA-7000 with a detection limit of 0.002 ppm.

3. Results and Discussion

3.1. Characteristics of Banana Puffer (*L. Lunaris*)

Banana puffer fish used in this study had the following characteristics: oval-shaped body, gray-brown in color from head to tail fin, and white abdomen. The fish has a dorsal fin, tail fin, anal fin, and a pair of pectoral fins. The banana puffer fish used in this study consisted of 9 samples and 3 repetitions which had an average total length of 14.07 ± 1.08 cm, average standard length 11.83 ± 1.04 cm, average height 4.63 ± 0.12 cm, average width 2.50 ± 0.20 cm and average weight 67.67 ± 1.53 g. Data on the average weight and morphometric of banana puffer fish is presented in Table 1.

Table 1. Data on the average weight and morphometric of banana puffer fish

Parameter	Size	Unit
Standard lenght	11.83±1.04	cm
Total length	14.07±1.08	cm
Height	4.63±0.12	cm
Width	2.50±0.20	cm
Weight	67.67±1.53	gram

The body weight and total length of banana puffer used in the research indicated that the fish were of juvenile size. The total length of adult banana puffer ranged from 14.5 to 35 cm (Auawithoothij and Noomhorm 2012). Nurjanah *et al.* (2014) stated that the difference in size and weight of puffer was influenced by growth. The growth was influenced by several factors which could be grouped into outer and inner factors. The external factors were the factors that could be controlled while internal factors were those which were hard to control such as heredity.

The puffer whose morphometric had been measured was then prepared by separating the meat, viscera, skin, bones and head in order to calculate the rendement. The parts of banana puffer fish used in this study were meat, viscera and skin, which were chopped to ease the process of chemical analysis.

3.2. Rendement

The rendement of the body parts banana puffer fish in this research was in the form of meat, viscera and skin. The rendement calculations were performed three times for each sample. The results of the rendement calculations are presented in Figure 1.

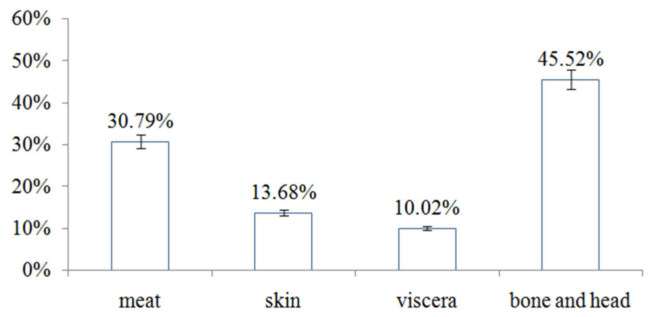

Figure 1. Rendement body part buffer fish

Figure 1 showed that the rendement of the bone and heads of banana puffer fish had the highest value 45.52% compared to the other parts. Puffer bones and heads could be used and processed to make fish bone powder and gelatin of high economic value. According to Kaya *et al.* (2007), the utilization of the fish bones to make fish bone meal had many advantages in that the bone meal contains minerals, especially calcium and phosphorus that can be used as a source of the fulfillment of minerals. Astawan *et al.* (2002) stated that the fish bone can be used as an alternative source of producing gelatin which is generally derived from pig skin.

The rendement of banana puffer fish meat was 30.79%. Puffer meat tastes delicious and savory, luring puffer meat eaters. The rendement of this fish skin reached 13.68% and the rendement of banana puffer viscera was 10.02%. Nurjanah *et al.* (2014) said that the skin is usually utilized to make fish skin cracker by the villagers of Gebang, Cirebon. Fish viscera has a great potential as a source of fish oil for feed ingredients. Fish viscera could be used to substitute fish meal in the feed formulation through the production of silage (Putra 2001).

3.3. Chemical Composition

The proximate analysis was conducted to determine the nutrient content of the crude parts of banana puffer fish, which include moisture, ash, protein, fat and carbohydrates. The carbohydrate content was calculated by different. The chemical composition tested was the samples of meat, skin and viscera of banana puffer fish, which are presented in Table 2.

Table 2. Chemical composition banana puffer fish

Chemical composition	Meat		Skin		Viscera	
	Wb (%)	Db (%)	Wb (%)	Db (%)	Wb (%)	Db (%)
Moisture	79.39±0.70	-	74.00±0.50	-	74.10±0.33	-
Ash	0.62±0.01	3.01±0.00	1.81±0.56	6.96±0.02	3.02±0.36	11.66±0.01
Fat	0.15±0.03	0.73±0.00	0.49±0.05	1.88±0.00	9.56±0.31	36.83±0.01
Protein	18.92±0.41	91.8±0.02	23.08±0.41	88.78±0.02	8.07±0.32	31.15±0.01
Carbohydrates*	0.92±0.52	4.46±0.03	0.61±0.85	2.35±0.03	5.25±1.10	20.27±0.04

*bydifference

Table 2 showed that the water level had the highest value on a wet basis in all the three samples of banana puffer fish. The protein had the second highest value after the water sample levels of meat and skin, and viscera was the second highest of the fat content.

The highest water content was in the banana puffer fish meat (79.39 ± 0.70%), followed by the viscera (74.10 ± 0.33%), and the lowest was skin (74.00 ± 0.50%). The water content in food greatly affects the quality and shelf life of the food material. The higher the water content in a material, the more quickly the material quality will degenerate, which may become a source of living microorganisms to grow. Immediate actions must be taken to maintain the product quality quickly and carefully by still keeping the cold chain. According to Ayas and Ozugul (2011), the difference in water levels could be caused by the types, biota ages, differences in environmental conditions, and the levels of the organism freshness.

The ash content is a mixture of inorganic or mineral components in food. Incineration is the sample preparation stage to conduct mineral analysis. The analysis result of the ash content in the banana puffer fish showed the highest value in the viscera samples, that is, 3.02 ± 0.36% (wb) and 11.66 ± 0.01% (db). The high ash content in the banana puffer fish viscera indicates the high mineral content in it. When biological material is burned, all organic compounds will be damaged, and most of the carbon turns into carbon dioxide (CO2), hydrogen into water vapor and nitrogen vapor nitogen be (N2). Minerals will be left behind in the form of ash and simple inorganic compounds, and the merger will occur between individuals or with oxygen to form inorganic salts (Arifin 2008).

Fat is an energy source that is more effective when compared with carbohydrate and protein, since 1 gram of fat can produce 9 kcal. The value is higher compared to the energy produced by 1 gram of protein and carbohydrates, which is 4 kcal. Fat can also be used as a source of essential fatty acids and vitamins (A, D, E, K) (Winarno 2008). Based on the analysis of fat content, the viscera samples were found to have the highest values of 9.56 ± 0.31% (wb) and 36.83 ± 0.01% (db). Parts of fish viscera such as liver are fat storage. In addition, in the viscera there are also stomach and intestines, which both serve to store food before being ingested, causing the accumulation of fat that contributes to the high levels of fat in the viscera. The fat content in the fresh meat of banana puffer fish was 0.15 ± 0.03% (wb), higher than that of *Takifugu rubripes* based on the research result by Saito and Kunisaki (1998), which was only 0.7%. *Takifugu rubripes* is a different kind of puffer fish, so the fat content in the body is not necessarily the same. According to Jacoeb *et al.* (2008), the fat content in fish is influenced not only by the type of fish but by feeding habit, type of food, age, environment, seasons, and nutrition sufficiency level (TKG).

Protein is a substance that is very important for the body. In addition to functioning as an energy source, it also serves as regulating and building substances. Protein is a source of amino acids containing the elements of C, H, O and N which are not owned by fats and carbohydrates (Winarno 2008). The analysis results of protein showed that the highest values on the meat samples of dry basis (db) were 91.8 ± 0.02%, skin 88.78 ± 0.02% and vicera 31.15 ± 0.01%, all of which decreased in protein content. The determination of dry basis was necessary to find out the real decrease in the protein samples of meat, skin and viscera of banana puffer fish by ignoring the water content. Most body tissues are water, followed by protein. Half or 50% of the cell dry weight in the tissue was in heart and flesh, consisting of proteins (Winarno 2008).

Carbohydrates calculated by different showed that banana puffer fish viscera had the highest values of 20.27 ± 0.04% (db) and 5.25 ± 1.10% (wb). Meanwhile, the low carbohydrate was found in the skin samples of 2.35 ± 0.03% (db) and 0.61 ± 0.85% (wb). The banana puffer fish meat had carbohydrates of 4.46 ± 0.03% (db) and 0.92 ± 0.52% (wb). The results obtained were not much different from the research conducted by Nurjanah *et al.* (2014), where the carbohydrate content of the fresh banana puffer fish was 4.42%. The carbohydrate in the fish meat was polysaccharide, that is, glycogen. Glycogen contained in the fishery products was approximately 1%. Carbohydrate content in fish meat ranged from 0.05 to 0.85% glycogen, 0.038% glucose, and 0.006 to 0.43% lactic acid (Adawyah 2007).

3.4. Composition of Mineral and Heavy Metal of Banana Puffer Fish

The mineral analysis were performed on seven minerals consisting of 4 macro minerals (Ca, Mg, Na and K), 3 micro minerals (Cu, Fe, and Zn) and heavy metals (Hg, Pb, Cd and As). The mineral and heavy metal contents of banana puffer fish can be seen in Table 3.

Table 3. Mineral and heavy metal contents of banana puffer fish

Type	Meat	Skin	Viscera	Max. limit SNI 2009
Mineral macro (ppm)				
Ca	146.96±1.72	25.21±0.14	7.186,71±88.92	
Mg	216.62±1.45	126.69±.,82	13.607,48±140.26	
Na	20.61±2.49	145.86±0.91	211.34±5.68	
K	1.469,34±6.92	495.32±3.42	1.289.95±7.29	
Mineral micro (ppm)				
Fe	6.85±0.01	22.07±0.19	167.03±1.48	
Zn	73.63±0.60	41.80±0.71	10.33±0.15	
Cu	0.43±0.01	0.25±0.01	5.03±0.02	
Heavy metals (ppm)				
Pb	0.51±0.01	0.05±0.00	0.57±0.02	0.3
Cd	0.02±0.00	0.02±0.00	0.19±0.01	0.1
As	*tt	*tt	*tt	0.5
Hg	*tt	*tt	*tt	1.0

Minerals (metals) are divided into two groups, namely the essential and non-essential minerals that are used in life activities. Essential minerals are needed in the physiological processes of animals, so this group of metals is the elements of essential nutrients that, if deficiency occurs, can cause an abnormal physiological process called mineral deficiency disease. These minerals are usually bound to proteins, including enzymes for metabolic processes, was Ca, P, K, Na, Cl, S, Mg, Fe, Cu, Zn, Mn, Co, I and Se. Non-essential metals are the metals that are not useful. At least, the benefits have not been found in animals, so that the presence of these elements more than normal can cause poisoning. The metals such as Pb, Hg, As, Cd and Al are even very harmful to living things (Darmono 1995; Permatasari 2006; Arifin 2008).

Heavy metals are known as elements with a high level of toxicity which has the ability to accumulate in the human body. Some dangerous heavy metals are lead (Pb), mercury (Hg), arsenic (As) and Cadmium (Cd). According to Darmono (1995), metal toxicity level is influenced by several factors: the level of heavy metals consumed, duration of consumption, age, species, sex, physical condition and the ability of the body's tissues to accumulate metals.

The results of this study indicated that the highest macro calcium (Ca), magnesium (Mg) and sodium (Na) were found in the viscera samples of bananas puffer fish, where the meat samples were found to contain high potassium (K). The food types of banana puffer fish have a high mineral content. According to Noviyanti (2004), the main foods of banana puffer fish are fish and crustaceans. The types of food found in the stomach of banana puffer fish consisted of eight types of fish (Sciaenidae and Silaginidae families), mussels (Trigonidae family), gastropods (Phasianellidae Family), shrimp (Peneidae family, genus Penaeaus family), crabs (Ocypodidae family) and squid (Loliginidae family).

The micro mineral content in the samples of bananas puffer fish analyzed showed that the content of iron (Fe) was the highest in the viscera, 167.03 ± 1.48 ppm. This is due to the factors of the food consumed by banana puffer fish. Besides, the foods eaten by the fish are all processed in the stomach. This is consistent with the results of the research conducted by Brown et al. (2004) which explains that most Fe is stored in the liver, spleen, and bone marrow. Fe content in the animal body varies, depending on nutrition, age, sex and species. The content of zinc (Zn) was the highest in the meat, 73.63 ± 0.60 ppm. On the one hand, Zn mineral is essential to the organism; on the others, the amount of Zn-containing waste from households or industry that goes into the waters is hard to control. According to Darmono (1995), Zn metal has a higher maximum concentration limit of Cu because Zn is widely available in the enzymes used in the process of metabolism and helps growth. The highest content of copper (Cu) in the viscera was 5.03 ± 0.02 ppm and the lowest copper value in the skin was 0.25 ± 0.01 ppm. The high mineral content of Cu in the banana puffer fish viscera was supported by Winarno (2008) who said that there was a lot of copper in the liver, kidney, and brain. The low concentration of copper in the skin was because the Cu content in the aquatic environment of Gebang, Cirebon Regency was lower in concentration. Copper (Cu) is needed by humans as Cu complex protein that functions in the formation of hemoglobin, collagen, blood vessels, and brain meylin. However, Cu would be very dangerous when consumed in excess (Palar 1994). The content of Cu in banana puffer if constantly consumed by humans will have a bad effect on the human health. Nurjanah *et al.* (2005) stated that if the body lacks copper, there will be an increase in lipid peroxides.

The content of non-essential heavy metals in banana puffer fish such as lead (Pb) and (Cd) had the highest value of Pb in the viscera and meat, that is, 0.57 ± 0.02 ppm and 0.51 ± 0.01 ppm, while the highest Cd in viscera was 0.19 ± 0.01 ppm. The puffer fish skin samples had the lowest Pb and Cd values. Heavy metals Pb and Cd are toxic and hazardous waste materials, so that when the dose exceeds the normal limit it can lead to poisoning. According to ISO 7387: 2009, the maximum limit of Pb in food is 0.3 ppm, and the maximum limit of Cd contamination in food is 0.1 ppm.

The contents of arsenic (As) and Hg (mercury) in all the three samples of banana puffer were not detected. This was probably due to the low contents of As and Hg, making them below the limit of detection equipment (<0.002 ppm). Leung et al. (2001) stated that the low content of heavy metals in organisms was due to several factors, among others, differences in growth rate, metabolic rate, body sensitivity (to the intake levels of certain heavy metals) and physiological needs for the metal. Mercury can exist in all living creatures of the sea, especially those living in polluted water. When the seafood is consumed by humans, the heavy metals will automatically be transferred into the body (Agustina 2010).

4. Conclusion

The viscera had ash content of 11.66% (db) and the value was greater than that of meat and skin. The highest macro mineral content in the meat and the skin was K (1,469.34 ppm and 495.32 ppm), while the viscera were Mg (13,607.48 ppm). The viscera macro mineral content higher than meat and skin. Micro minerals dominant in the meat and the skin was Zn (73.63 ppm and 41.80 ppm), while the viscera were Fe (167.03 ppm). The meat and viscera contained Pb which exceeds the allowable limit, as well Cd in the viscera.

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