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RESEARCH ARTICLE

Effects of Steaming on the Functional Properties and Mineral Content of *Lumi-Lumi* Fish (*Harpodon Nehereus*) Meal

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Abstract

The purpose of this study is to evaluate the mineral content, phenolic content, antioxidant activity, and soluble protein of *Lumi-lumi* fish meal (*Harpodon nehereus*) treated with and non-steamed for 30 min. The results revealed that iron (Fe) mineral content was significantly higher in the steamed than non-steamed treatment but not in calcium (Ca). However, there was a decrease in the minerals magnesium (Mg), phosphorus (P), and zinc (Zn) during the steamed process, which was not significant. Then, during steamed, there was a significant decrease in antioxidant activity but not in total soluble protein, whereas phenolic content increased. Based on the findings of this study, *Lumi-lumi* fish (*Harpodon nehereus*) meal steamed treatment could be the best treatment because it contained the best phenolic and Fe and Ca minerals.

Keywords: Lumi-lumi; *Harpodon nehereus*; Steamed; Functional properties; Minerals

Introduction

Lumi-lumi fish (*Harpodon nehereus*), known as Bombay duck, is a demersal fish prospective to be developed in Meulaboh. The abundant availability of *Lumi-lumi*, especially in the West-South (Barsela) waters, makes it a characteristic of Meulaboh. However, data on the production of catching it has yet to be available in Aceh. *Lumi-lum*i fish is also known by different names in various regions of Indonesia, such as Nomei fish in Kalimantan and Lomek fish in Riau. The naming of *Lumi-lumi* itself is a local name in the waters of Barsela Aceh. *Lumi-Lumi* is reported to be able to breed well in river estuaries, beaches, and mud and can live all year round (Setiawan et al., 2020) with extensive distribution in Indonesia, including the Arafuru Sea, Bay of Bengal, Sumatra, Sea Java, South Sulawesi, Kalimantan, the Bay of Bengal, and the South China Sea (Nugroho et al., 2014). Unfortunately, it has yet to report the research results on its functional and micronutrient properties of it, especially in Indonesia. It is very potential for health, considering it contains minerals and protein, which are relatively high. *Lumi-lumi* is reported to contain mineral calcium (Ca) of 370 mg/100 g, iron (Fe) of 3.11 mg/100 g, and phosphorus (P) of 832 mg/100 g (Nazir & Magar, 1965). It also contains various crude proteins, namely 50.64%, 58.33%, and 59.00% (Ratrinia et al., 2019; Siddique & Aktar, 2011; Mithun et al., 2021). Geographical factors influence the diversity of nutritional value of *Lumi-lumi.* Therefore, identifying functional properties and micronutrients in different water areas, especially Barsela, is essential.

Until now, the development of processed *Lumi-lumi* is still straightforward. Coastal communities generally market in dry conditions through a salting process. It is because it contains up to 89.1% water, so it has a high risk of damage (Nazir & Magar, 1965) and has a soft texture and a fishy smell. This condition causes the selling value of *Lumi-lumi* to be relatively low and often not sold in traditional markets.

In this study, we evaluated the functional properties and the mineral content of *Lumi-lumi* fish (*Harpodon nehereus*) to help stakeholders promote their superiority and encourage producers to produce diversified food products based on local resources to improve small fishermen's welfare directly. In this method, *Lumi-Lumi* was treated with steamed and non-steamed. The steamed process was chosen

because it is a healthy cooking method. However, this method can affect the number of minerals and their functionality. Therefore, identification is necessary. The findings of this study can encourage efforts to use *Lumi-lumi* fish to produce functional food products based on local *Lumi-lumi* fish flour, which can improve the welfare of small fishermen and promote the superiority of local resources.

Methods

Material

The *Lumi-lumi* fish (*Harpodon nehereus*) used in this study was obtained from the central market, Meulaboh, Indonesia.

Preparation of *Lumi-lumi* Fish Meal Samples

Samples were prepared in two treatments: *Lumi-Lumi* fish, which was steamed and non-steamed. In the steamed treatment, the *Lumi-lumi* removed the head and abdominal part. Then steamed for 30 min and dried at 50^{o}C for 48 h. Then grind it using a blender. In the non-steamed treatment, the fish removed from the insides and heads were immediately dried at 50^{o}C for 48 h.

Soluble Protein Assay

Soluble protein assay using the Lowry method. Determining soluble protein involves preparing a standard solution, Lowry A reagent, and Lowry B reagent.

Sample Preparation

The sample preparation stage is to carry out a multilevel retail process up to 10,000 times. 1 mL of the *Lumi-lumi* fish meal sample solution was transferred to a test tube containing 9 mL of distilled water to obtain a dilution factor of 10 times (FP=10). Next, 1 mL of the sample solution was dissolved in 9 mL distilled water (FP = 100). Furthermore, it was carried out with the same steps until a dilution factor of 10,000 times (FP = 10,0000) was obtained.

Standard Solution

0.1 g of Bovine Serum Albumin (BSA) was put in a beaker, then added 10 mL of distilled water and homogenized (10 mg/mL). Next, 1 mL of BSA solution was transferred to a test tube containing 9 mL of distilled water (1 mg/mL). Standard solutions are prepared from BSA solution (1 mg/mL) with a 200-1000 ppm concentration.

Lowry A and B reagents

Lowry A reagent consists of folin-ciocalteau and distilled water in a ratio of 1: 1. Lowry B reagent is prepared by combining 30 mL of 2% Na_2CO_3 solution in 0.1 N NaOH with 1% Na-K-Tartrate solution, and 1% $CuSO_4$ solution, respectively - each as much as 0.3 mL.

Determination of Total Soluble Protein

2 mL of sample was added with 2.75 mL of Lowry B reagent, then vortexed and incubated for 15 min. Then 0.25 mL of Lowry reagent A was added, vortexed, and incubated for 30 min. After incubation, the absorbance of the solution was measured at a wavelength of 660 nm using a spectrophotometer. Total soluble protein was expressed in milligrams of BSA equivalent per mL sample (mg BSA/mL sample).

Sample Extract Preparation

The sample extract refers to the procedure of Marjoni et al. (2012). 50 g of *Lumi-lumi* fish meal sample was weighed, and 500 mL distilled water was added. Subsequently, it was stirred until homogeneous. After that, boil the solution in an infusion pan at 90^{o}C for ± 15 min with occasional stirring. Then it is filtered, and the result is referred to as the filtrate. Materials that do not pass are rinsed again with distilled water. The filtrate is then dried using a rotary evaporator. Furthermore, the dry samples were used to analyze phenolic content and DPPH antioxidant activity.

Analysis of Phenolic Content

Analysis of phenolic content refers to Orak's procedure (2006). 100 mg of the extract was dissolved in distilled water up to 10 mL (10 mg/mL). Then 1 mL of the 10 mg/mL solution was taken to be transferred to a test tube, then diluted to 10 mL (1 mg/mL). Next, 0.2 mL of the extract was taken, 15.8 mL of distilled water, and 1 mL of folin-ciocalteu reagent was added, then vortexed. The solution mixture was allowed to stand for 8 min, and then 3 mL of 10% Na_2CO_3 solution was added and vortexed. The solution was incubated for 120 min at room temperature. After incubation, the sample solution was measured with a UV-VIS spectrophotometer at a wavelength of 765 nm. Total phenolic compounds were measured in milligrams of gallic acid equivalents per gram of extract (mg GAE/g extract).

Analysis of Antioxidant Activity

Analysis of antioxidant activity used the Blois procedure (1958) using the 2,2-diphenyl-1-picrylhydrazyl. (DPPH) method. 1 g of extract dissolved in 10 ml of ethanol. Next, 0.2 mL was mixed with 3.8 mL of 0.1 mM DPPH solution (DPPH dissolved in ethanol), then dissolved with ethanol until a purple color appeared. Then vortexed and incubated for 30 min. After incubation, it was vortexed again, and the absorbance was measured with a UV-VIS spectrophotometer at a wavelength of 515 nm. The antioxidant activity of *Lumi-lumi* fish meal extract in capturing DPPH free radicals is expressed as a percentage using the following equation:

$$\text{Antioxidant Activity (\%)} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

Figure 1. Equation of antioxidant activity

The absorbance of control consisted of a mixture of 0.2 mL ethanol and 3.8 mL DPPH, then measured at a wavelength of 515 nm.

Mineral Analysis

Mineral analysis followed the procedure of Association of Offical Analytical Chemistry (AOAC) (2005). The minerals tested were Calcium (Ca), Magnesium (Mg), Copper (Cu), Iron (Fe), Iron (Zn), and Phosphorus (P). Determining minerals began with wet ashing, in which 1 g of sample was weighed, then 10 mL of HNO_3 was added. Next, 3 mL of 60% $HClO_4$ was added and heated slowly over the digestion tool until the foam that formed stopped. If a black solution is formed, 10 mL of HNO_3 has added again until the solution becomes clear. Then the solution was cooled, and 10 mL HCl was added and continued for 5 min of heating which was counted after boiling, then filtered using Whatman paper no. 1 and measured in a measuring flask up to 50 mL. The filtrate from wet ashing was analyzed for minerals using an Atomic Absorption Spectrophotometer (AAS).

Data analysis

The presentation of the data was processed using Microsoft Excel and SPPS version 22 software. The Shapiro-Wilks test was performed to determine whether the data were normally distributed. Furthermore, statistical data were analyzed using an independent t-test with an alpha of 5%.

Results

Soluble Proteins, Antioxidants, and Phenolic Content

The values for phenolic content, antioxidant activity, and soluble protein were obtained, and presented in Table 1. Both contents of phenolic and soluble protein did not significantly affect treated with and non-steamed. However, the antioxidant activity decreased significantly while steaming for 30 min

Table 1. Content of phenolic, antioxidant activity, and soluble protein of *Lumi-lumi* Fish Meal (*Harpodon nehereus*)

Functional Characterization	Steamed	Non-steamed
Phenolic content (mg GAE/g Extract)	8,30±3,09 [a]	6,76±0.72 [a]
Antioxidant activity (%)	76,96±3,50[a]	83,58±3.64 [b]
Soluble protein (mg BSA/mL sample)	7276,8±1.65[a]	7678,3±0.99[a]

Data presented were mean ± standard deviation; n = 3 (triple). Numbers with different letters in the same column and row showed a significant differences at the 5% test level.

Macro and Micro Minerals

The study of the macro and micro mineral content analysis in *Lumi-lumi* fish meal with steamed and non-steamed treatment is presented in Table 2. The Mg, P, and Zn minerals decreased non-significant during steaming, but the Fe mineral was significantly higher in the steamed than non-steamed treatment, but not in calcium (Ca).

Table 2. Macro and Micro Mineral Content of *Lumi-lumi* Fish Meal (*Harpodon nehereus*)

Mineral Compound	Steamed	Non-steamed
Macro Minerals (mg/100 g)		
Magnesium (Mg)	6,10± 0,00 [a]	6,17± 0,06 [a]
Phosphorus (P)	1350± 56,57 [a]	1423,33± 37,86 [a]
Calcium (Ca)	531,15± 27,22 [a]	517,20± 5,35 [a]
Micro Minerals (mg/100 g)		
Iron (Fe)	4,57± 0,42 [a]	3,07± 0,15 [b]
Copper (Cu)	<LOD*	<LOD*
Zinc(Zn)	5,30± 0,36 [a]	5,33± 0,93 [a]

Data presented were mean ± standard deviation; n = 3 (triple). Numbers with different letters in the same column and row showed a significant differences at the 5% test level. LOD = Limit of Detection. Results under test detection (<0.2*

mg/100g)

Discussion

In previous research, the yields of *Lumi-lumi* fish meal treated with steam and without steam were obtained respectively at 10.23% and 10.57% (aSafrida et al., 2022). The yield value amount aims to calculate the economic value, namely the number of parts that can be utilized to produce *Lumi-lumi* fish meal. Based on the study's results, Table 1 showed that the steamed treatment for 30 min had a significant effect on the antioxidant activity ($p<0.05$) of *Lumi-lumi* fish meal, which was 76.96% and 83.58% non-steamed. The decrease in antioxidant activity in the steamed treatment was thought to be due to several proteins in the *Lumi-lumi* experiencing denaturation during the heating process, resulting in total soluble protein in the steamed treatment of 7276.8 µg/L and 7678.3 µg/L in the non-steamed treatment. It follows the statement of Jian et al. (2018) that protein during heating will experience denaturation resulting in low molecular weight and a decrease in dissolved protein. Proteins are composed of amino acid strands where the peptide's size and solubility determine the antioxidant activity's capacity (Bordbar et al., 2018). It strengthens the suspicion of why there is a decrease in antioxidant activity in the steamed treatment.

Besides amino acid compounds and bioactive peptides, antioxidant activity is also determined by several phenolic compounds in fish. Phenolic components affect antioxidant activity by acting as free radical scavengers (Safrida et al., 2020; bSafrida et al., 2022). The steamed treatment did not significantly affect the phenolic content in the research results. It is suspected that bioactive peptides influence antioxidant activity. Soluble protein testing measures the amount of protein easily absorbed by the human digestive system.

However, in the analysis of the Mg and P minerals, there was a non-significant decrease in Mg which was 6.10 mg/100g from 6.17 mg/100g (non-steamed), and mineral P, which was 1350 mg/100g from 1423.33 mg/100g (non-steamed) which presented in Table 2.

. The decrease in these minerals can be caused during the heating process, some divalent minerals such as Mg and P become dissolved, and muscle proteins are thought to experience denaturation, especially myofibrillar and sarcoplasmic (Barbosa et al., 2021). As a result of this denaturation, the protein will experience disconnection and dehydration in the muscle fibrils so that the protein structure changes and complex bonds are formed between the protein-minerals, which result in the solubilization of Mg and P minerals (Bastías et al., 2017; Ochiai & Ozawa, 2020; Barbosa et al., 2021).

In the steamed treatment, Ca mineral content was higher but not significantly different from the non-steamed with successive values, namely 531.15 mg/100 g and 517.20 mg/100 g. These results are different from those reported by Barbosa et al. (2021), where there was a decrease in Ca content in both types of fish, namely gilthead seabream (*Sparus aurata*) and common carp (*Common carp*) with a long steamed process of 15 min.

In the Fe mineral, the results were obtained: the steamed process significantly affected the Fe content with a value of 4.57 mg/100g and 3.07 mg/100g in the non-steamed treatment. These results follow those reported by Barbosa et al. (2021), gilthead seabream (*Sparus aurata)* from a type of marine fish and common carp (*Common carp)*, namely freshwater fish, both of which were treated by steamed for 15 min containing higher Fe minerals than non-steamed. Iron is essential in preventing anemia (Briguglio et al., 2020), with a total requirement of 18 mg per day for adult women and 27 mg per day for pregnant women (Burke et al., 2014). The amount of Fe content in the results of this study was slightly different from that reported by Nazir & Magar (1965), Bombay ducks without being given a heating treatment which was 3.11 mg/100 g. Differences in the number of minerals in fish are affected by their habitat, size, and weight (Barbosa et al., 2021).

This study showed that the Cu mineral content under test detection was less than 0.2 mg/100 g. The results of this study were the same as those reported by Nazir & Magar (1965). Namely, the Cu content was obtained at 0.126 mg/100 g.

In Zn minerals, it was found that *Lumi-lumi*, which was given heating treatment, was slightly lower than non-steamed, with values of 5.30 mg/100 g and 5.33 mg/100 g. This result is the same as that Mnari et al. (2012) reported. It was found that the mineral content of Zn in Sea Bram (*Sparus aurata*) fish treated with steam significantly decreased the amount of Zn compared to non-steamed. This study's decrease in Zn minerals is thought to be due to complex bonds between proteins and Zn minerals during heating (Bastias et al., 2017; Barbosa et al., 2021). The mineral Zn is reported to play a critical role in regulating the function of the immune system

both specifically and non-specifically and is involved in synthesizing DNA, RNA, and protein (Weyh et al., 2022). The recommended daily requirement for Zn is 10 mg daily for children and 15 mg daily for adults (Mnari et al., 2012).

Conclusion

The steamed process of *Lumi-lumi* fish meal (*Harpodon nehereus*) affects functional properties and mineral content characterization. The results showed that the steamed process significantly decreased the antioxidant activity but not significantly the total soluble protein. On the contrary, the phenolic content were higher. The Fe mineral content was found to be significantly high, and Ca minerals were obtained to be insignificant. The content of other minerals, namely Mg, P, and Zn, decreased during the steamed process but showed no statistically significant difference. It shows that the steamed process does not significantly affect the mineral content.

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Author Contributions and Competing Interests

All authors contributed to this study and declared no conflict of interest.

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