# Assessment of Defatted Protein Concentrate from Flying Fish Roe Filament and Its Amino Acid Profile

*Vritta* Amroini Wahyudi[1,*], *Rohmin* Hidayanti[1], *Hanif* Alamadin Manshur[1], *Noor* Harini[1], and *Mohammed* Ali Wedyan[2]

[1]Department of Food Technology, University of Muhammadiyah Malang, Jl. Raya Tlogomas No. 246, Malang 65144, East Java, Indonesia
[2]Department of Biological Sciences and Biotechnology, Faculty of Science, The Hashemite University, 13115 Zarqa, Jordan

**Abstract.** Flying fish roe filament has a high protein content that can be explored as an alternative nutrition food. Unfortunately, those roe filaments still have a fishy odor caused by volatile compounds and fatty acid derivatives. This study aims to eliminate the fishy odor by removing the fat content using the defatting method. The research design used in this study was a simple Randomized Block Design (RDB) consisting of four treatments (control, 1 h, 3 h, and 5 h). The method used in this study for determining amino acids is a descriptive method using HPLC (High-Performance Liquid Chromatography). The best treatment for extraction time was 5 h with the results of moisture content 10.5 %, ash content 7.5 %, protein content 47.925 %, fat content 8.65 %, total volatile base (TVB) 4.939 %, and yield 86.63 %. TVB value proved that the treatment could reduce the fatty acid content, so the protein concentrate becomes not fishy (from 9.64 to 4.939 %). The best treatment showed the highest content of lysine (149.75 mg 100 $g^{-1}$) than another essential amino acid.

**Keywords:** Health food, *Hirundichthys oxycephalus*, protein extraction, stunting prevention, tobiko.

## 1 Introduction

Exploration of marine materials continues to be carried out, including finding sources of protein that can be utilized in food technology [1]. Previous studies have examined flying fish [*Hirundichthys oxycephalus* (Breder, 1928)] roe filament as protein sources. Flying fish roe filament comes from white fibers surrounding flying fish roe called “tobiko” [2]. In eastern Indonesia, flying fish can be found in the Natuna Sea, Makassar Strait, Flores Sea, Aru Sea, Arafura Sea, and Papua. On the other hand, flying fish can be found in North Sulawesi, Sabang Waters, West Sumatra Coast, Bali Strait, and East Java [3].

* Corresponding author: vritta@umm.ac.id

Exploring protein from flying fish roe filaments is interesting because previous studies have stated that the amount of filament protein is higher than that of flying fish roe. For example, the protein content of flying fish roe filament is 73 % ± 0.07 % [2], resulting in 72 % to 74 % [4]. Another study reported that the protein content from flying fish roe filament is 33.70 % to 40.10 %. Meanwhile, protein content from flying fish roe ("tobiko") is 30.27 % to 37.53 % [5].

Protein from flying fish roe contains several essential amino acids such as leucine (5.86 %), lysine (3.69 %), valine (3.41 %), isoleucine (3.33 %), threonine (2.86 %), phenylalanine (2.30 %), histidine (1.38 %), methionine (1.21 %), and also non-essential amino acid such as glutamate (7.08 %), arginine (6.11 %), alanine (3.82 %), aspartic acid (3.75 %), serine (3.05 %), glycine (1.84 %), tyrosine (1.46 %) [2]. However, the protein concentrate from flying fish roe filament still has a fishy odor. This is because the raw materials that cause this fishy odor come from marine sources, and trimethylamine oxide's interaction with the double bonds of unsaturated fatty acids produces trimethylamine [6, 7]. In addition, volatile compounds derived from fatty acids include aldehydes and ketones [8]. Therefore, defatting is one way to get rid of the fishy odor [9].

The defatting method is an extraction process using non-polar solvents to remove fat content and reduce Total Volatile Base (TVB) with a specific extraction time [10, 11]. The separation process between fat and material is carried out with stirring so that the fat binding phase by the solvent can occur adequately [12]. The type of solvent that is commonly used in defatting extraction in the manufacture of protein concentrates, the solvent used is the dissolving of Isopropyl Alcohol (IA) [11].

This study examines the process of defatting protein concentrate from flying fish roe filaments and then analyzes its chemical character and amino acid profile using High Performance Liquid Chromatography (HPLC). The results of this study could potentially be used for enrichment food to solve several health problems such as stunting [13, 14], antiaging [15, 16], anticancer [16], anti-carcinogenic [17], antidiabetic [18, 19], antihypertensive [20, 21], antimicrobial [22], antioxidative stress [22], anti-inflammatory [23], cholesterol-lowering [23], growth-enhancing [24], immunomodulatory [25], mineral binding [26], radical scavenging [27], regulation of glucose and insulin homeostasis [28], and satiety regulating [29].

## 2 Methods

### 2.1 Description of the study sites

The flying fish roe filament was obtained from PT. Kelola Mina Laut, Sumenep, East Java, Indonesia. The manufacture of concentrate protein was carried out in the Food Technology Laboratory, University of Muhammadiyah Malang. The amino acid was analyzed at IPB University. The study consisted of two stages, (i) defatting and protein extraction from flying fish roe filament, (ii) analysis of the chemical properties and amino acid profile. This study used the simple Randomized Block Design (RBD) with a factor which is a defatting treatment that includes on it (protein concentrate without defatting, and protein concentrate with defatting from 1 h, 3 h, and 5 h of extraction). Amino acid profile was analyzed using HPLC (Thermo Scientific ODS-2 Hyersil, US).

## 2.2 Defatting and protein extraction from flying fish roe filament

Extracting fly fish roe filament protein concentrate for fat removal refers to the research defatting method [30]. First, flying fish roe filament was crushed using a cooper. After that, it was extracted with Isopropyl Alcohol (IA) in a ratio of 1:3 (w $v^{-1}$) at 50 °C for 1 h, 3 h, and 5 h. After that, filtering was carried out using filter paper to separate the solvent and the precipitate. After that, the precipitate was dried using a cabinet dryer at a temperature of 40 °C to 50 °C for 7 h. The next step refers to the previous study method [31] modified at the pH point. The dried precipitate was then added to distilled water (10 % w $v^{-1}$) and stirred with a water bath shaker at room temperature for 5 h. The sample was stored in the showcase for 24 h and filtered to obtain the filtrate. The filtrate obtained was then extracted with the addition of 1 M NaOH until a pH of 8.5 was obtained and stirred for 1 h with a magnetic stirrer at 25 °C. Finally, the protein filtrate was added with citric acid (1 M, 0.2 mL) until the pH become 7 (neutralization) and centrifuged at room temperature for 20 min at $1 \times 10^4$ rad $s^{-1}$ to obtain a precipitate. Then the precipitate obtained is put into the freezer to freeze and then put in a vacuum oven. Flying fish roe filament protein concentrate is received as a powder that is still slightly wet [2].

## 2.3 Protein concentrate analysis

The protein concentrate was analyzed: protein concentrate (Kjedahl method) [32], lipid content (Soxhlet method) [33], Total Volatile Based (TVB) [34], moisture content (Gravimetry method) [35], ash content (Gravimetry content) [35], and Yield (%).

## 2.4 Protein content analysis

The protein concentrate (0.2 g) was put into a Kjeldahl flask, a catalyst was added, and $H_2SO_4$ (0.1 M; 2 mL) was added. The sample was digested (4 h) until it became clear and cooled to room temperature. Next, the sample was added to distilled water (15 mL), and NaOH (0.1 M; 10 mL). The sample is distilled. The distillate was accommodated in a Erlenmeyer, then $HBO_3$ (0.1 M; 15 mL) was added until it turned turquoise [32]. The sample is then titrated with HCl (0.02 N) until it turns purple. Volumes are recorded, according Equation (1) and Equation (2) [36].

$$\text{Nitrogen (\%)} = \frac{(\text{mL HCl} \times \text{N HCl}) \times 14.008}{\text{sample (mg)}} \text{x } 100\ \% \quad (1)$$

$$\text{Protein content} = \%\ \text{Nitrogen} \times \text{conversion factor } (6.25) \quad (2)$$

## 2.5 Lipid content analysis

Protein concentrate (2 g) was weighed and put into thimble paper. The thimble paper is inserted into the Soxhlet. The lipid analysis process was carried out by Soxhlet using petroleum benzene solvent (85 °C, 4 h). The filtrate in the Soxhlet flask was then concentrated using a rotary evaporator (ZZKD Machine, US). Lipid content is calculated according to Equation (3) [33].

$$\text{Lipid content} = \frac{\text{lipid extract (g)}}{\text{sample (g)}} 100\ \% \quad (3)$$

### 2.6 Amino acid HPLC analysis

Amino acid profile analysis was carried out at the Laboratory of IPB University, HPLC (Thermo Scientific ODS-2 Hyersil, US). The injection rate was set at 1 mL $min^{-1}$, using a fluorescence detector and Buffer A and Buffer B as the mobile phase. Buffer A is dissolved in 1 L of water. Buffer B was dissolved in methanol (95 %) and water. Amino acid analysis was carried out by dissolving the sample (protein concentrate) with HCl (0.01 N, 10 mL) then, filtered with millipore paper, then adding buffer (potassium borate, pH 10.4; 1:1). The analysis was carried out by entering the sample (5 L) and OPA (o-Phthaldialdehyde reagent solution) (25 µL) and left for 1 min. After that, the sample was injected into the HPLC column (5 µL) and then waited until all the amino acids were separated and detected [37]. All chemicals in this research using pro-analysis.

### 2.7 Statistical analysis

The research design used is a simple random group design consisting of four treatments, namely the control of raw materials before the defatting process (raw material) and the treatment of the length of extraction time, which is 1 h, 3 h, and 5 h. Each treatment is repeated twice. The second study design used descriptive methods for determining amino acids using HPLC.

## 3 Result and discussion

### 3.1 Result

Based on the various analysis results, it was found that the extended treatment of the extraction time of the defatting method had no significant effect on the total yield produced (sig > 0.05). The results of the filament yield data of flying fish roes defatting can be seen in Table 1.

**Table 1.** Results of analysis of the yield of flying fish roe filaments various treatments.

| Treatment | Yield (%) |
|---|---|
| Extraction 1 h | 89.20 ± 0.05 |
| Extraction 3 h | 89.35 ± 0.06 |
| Extraction 5 h | 86.63 ± 0.05 |

Based on Table 1, the average yield of flying fish roe filaments ranges from 86.63 % to 89.20 %. The average yield of flying fish roe filaments has increased and decreased insignificantly. According to Mardina *et al.* [38], the longer the extraction time, the higher the yield. However, the yield obtained with the long extraction time treatment is 5 h (86.63 %) less than the long treatment of extraction time 1 h (89.20 %) and 3 h (89.35 %).

#### *3.1.1 Total Volatile Base (TVB)*

Based on the results of various analyses conducted, the long treatment of defatting extraction time has a significant effect on the Total Volatile Base (TVB) filaments of flying fish roes (sig > 0.05). From the results obtained, there is a decrease in the value of TVB. Total Volatile Base (TVB) data results can be seen in Table 2.

**Table 2.** Results of Total Volatile Base (TVB) analysis of flying fish roe filaments after defatting.

| Treatment | Total volatile base (mg N 100 $g^{-1}$) |
|---|---|
| Control | 9.64 ± 0.33d |
| Extraction 1 h | 6.59 ± 0.00c |
| Extraction 3 h | 5.88 ± 0.00b |
| Extraction 5 h | 4.94 ± 0.33a |

Note: The average value followed by the same letter is no different in significant terms according to Duncan α = 5 % test.

The results of the study data total volatile base filaments of flying fish roes with control treatment and length of extraction time of 1 h, 3 h, and 5 h, namely 9.64 mg N 100 $g^{-1}$, 6.59 mg N 100 $g^{-1}$, 5.88 mg N 100 $g^{-1}$, and 4.94 mg N 100 $g^{-1}$.

### *3.1.2 Fat content*

Based on the results of various analyses conducted, the long treatment of defatting extraction time has a significant effect on the fat content of flying fish roe filaments (sig > 0.05). The results of data on the fat content of flying fish roes after defatting can be seen in Table 3.

**Table 3.** Results of fat content analysis of flying fish roes after defatting

| Treatment | Fat content (%) |
|---|---|
| Control | 11.05 ± 0.02c |
| Extraction 1 h | 9.20 ± 0.05b |
| Extraction 3 h | 8.75 ± 0.03a |
| Extraction 5 h | 8.65 ± 0.04a |

Note: The average value followed by the same letter is no different in significant terms according to Duncan α = 5 % test.

The highest fat content of flying fish roe filaments is found in the control treatment of 11.05 %. As for the lowest fat content, there is in the treatment of the long extraction time of 5 h which is 8.65 %.

### *3.1.3 Protein content*

Based on the results of various analyses conducted, the long treatment of extraction time has a significant effect on the protein content in the filaments of flying fish roes due to defatting (Sig > 0.05). The results of the filament protein levels of fish roes flying after defatting can be seen in Table 4.

**Table 4.** Results of protein content analysis of flying fish roes after defatting

| Treatment | Protein content (%) |
|---|---|
| Control | 47.49 ± 0.04c |
| Extraction 1 h | 31.94 ± 0.03a |
| Extraction 3 h | 39.24 ± 0.05b |
| Extraction 5 h | 47.93 ± 0.04c |

Note: The average value followed by the same letter is no different in significant terms according to Duncan α = 5 % test.

Based on Table 4, the average protein content of flying fish roe filaments with various treatments (47.49 %; 31.94 %; 39.24 %; 47.93 %).

### *3.1.4 Moisture content*

Based on the analysis results, the variety of treatments for long extraction times in the defatting method significantly affects the moisture content of flying fish roes (sig > 0.05). Here is a percentage of the moisture content of flying fish roe filaments after extraction by defatting method Table 5.

**Table 5.** Results of moisture content analysis of flying fish roes after defatting.

| Treatment | Moisture content (%) |
|---|---|
| Control | 16.37 ± 0.07d |
| Extraction 1 h | 11.65 ± 0.07c |
| Extraction 3 h | 9.15 ± 1.20a |
| Extraction 5 h | 10.05 ± 1.34b |

Note: The average value followed by the same letter is no different in significant terms according to Duncan α = 5 % test.

The study data results showed that the percent moisture content contained in the filaments of flying fish roes after going through the defatting extraction process decreased compared to the filaments of flying fish roes control treatment. The moisture content of the filament of the flying fish roe treatment of control by 16.37 %, after going through the defatting extraction process with long treatment extraction times of 1 h, 3 h, and 5 h are 11.65 %; 9.15 % and 10.05 %.

### *3.1.5 Ash content*

Based on the results of variety analysis, the long treatment of extraction time has a significant effect on the content of ash levels in the filaments of flying fish roes (sig > 0.05). Data on the ash levels of flying fish roes after defatting can be seen in Table 6.

**Table 6.** Results of analysis of ash levels of flying fish roes after defatting.

| Treatment | Ash content (%) |
|---|---|
| Control | 8.85 ± 0.00c |
| Extraction 1 h | 9.95 ± 0.07ab |
| Extraction 3 h | 7.75 ± 0.21b |
| Extraction 5 h | 7.5 ± 0.28ab |

Note: The average value followed by the same letter is no different in significant terms according to Duncan α = 5 % test.

The results of the analysis of the ash levels of flying fish roes with control treatment, the length of extraction time of 1 h, 3 h, and 5 h in a row are 8.85 %; 9.95 %; 7.75 %; and 7.5 %.

### *3.1.6 Amino acid profile*

Based on the amino acid HPLC test results on flying fish roe filaments, and the filament concentrate of flying fish roes obtained, 15 types of amino acids. The results of the amino acid test of flying fish roe filaments after defatting and protein concentrates of flying fish roe filaments can be seen in Table 7. The amino acids found in the filaments of flying fish roes consist of nine types of essential amino acids (Histidine, threonine, valine, methionine, isoleucine, leucine, phenylalanine, lysine, and arginine) and six types of non-essential amino acids (aspartate acid, glutamic acid, serine, glycine, alanine, tyrosine).

**Table 7.** Amino acid compotition.

| Amino acid | Result | | | |
|---|---|---|---|---|
| | **Filament (% w $w^{-1}$)** | **Filament* (% w $w^{-1}$)** | **Protein concentrate (mg 100 $g^{-1}$)** | **Protein concentrate** (% w $w^{-1}$)** |
| Essential | | | | |
| Histidine | 2.24 ± 0.03 | 3.51 ± 0.09 | 17.71 ± 0.02 | 1.38 ± 0.02 |
| Threonine | 2.79 ± 0.02 | 1.90 ± 0.01 | 56.25 ± 0.02 | 2.86 ± 0.02 |
| Arginine | 1.49 ± 0.03 | 2.56 ± 0.08 | 21.79 ± 0.02 | 6.11 ± 0.02 |
| Methionine | 0.66 ± 0.04 | 1.13 ± 0.03 | 2.16 ± 0.02 | 1.21 ± 0.02 |
| Valine | 5.95 ± 0.02 | 2.83 ± 0.08 | 65.50 ± 0.02 | 3.41 ± 0.02 |
| Phenylalanine | 2.90 ± 0.04 | 1.30 ± 0.09 | 37.94 ± 0.02 | 2.30 ± 0.02 |
| I-leucine | 1.39 ± 0.02 | 1.63 ± 0.02 | 34.03 ± 0.02 | 3.33 ± 0.02 |
| Leucine | 3.99 ± 0.01 | 2.52 ± 0.16 | 67.49 ± 0.02 | 5.86 ± 0.02 |
| Lysine | 3.37 ± 0.02 | 2.32 ± 0.11 | 149.75 ± 0.02 | 3.69 ± 0.02 |
| Total | 24.78 ± 0.02 | 19.70 ± 0.02 | 452.62 ± 0.02 | 30.15 ± 0.02 |
| Non essential | | | | |
| Aspartate acid | 4.00 ± 0.03 | 3.36 ± 0.02 | 61.37 ± 0.02 | 3.75 ± 0.02 |
| Glutamate acid | 10.36 ± 0.03 | 7.43 ± 0.17 | 64.61 ± 0.02 | 7.08 ± 0.02 |
| Serine | 6.70 ± 0.04 | 2.50 ± 0.00 | 47.52 ± 0.02 | 3.05 ± 0.02 |
| Glycine | 3.85 ± 0.02 | 2.25 ± 0.15 | 54.88 ± 0.02 | 1.84 ± 0.02 |
| Alanine | 3.97 ± 0.03 | 3.21 ± 0.17 | 43.34 ± 0.02 | 3.82 ± 0.02 |
| Tyrosine | 5.49 ± 0.02 | 1.71 ± 0.04 | 60.80 ± 0.02 | 1.46 ± 0.02 |
| Total | 34.37 ± 0.02 | 20.46 | 332.52 ± 0.02 | 21.00 ± 0.02 |

Note: *Azka *et al.* [5], **Wahyudi *et al.* [2].

The highest essential amino acid content in flying fish roe filaments is valine by 5.95 %, while the highest non-essential amino acid is glutamic acid at 10.36 %. Valine is an essential amino acid that can increase energy and endurance, lower blood sugar levels, and grow. On the other hand, glutamic acid is a non-essential amino acid that supports brain function, strengthens memory, speeds the healing of wounds in the intestines, and dampens emotions [39]. The highest level of essential amino acids in the protein concentrate of flying fish roe filaments is lysine at 149.75 mg 100 $g^{-1}$. On the other hand, the highest non-essential amino acid is glutamic acid at 64.61 mg 100 $g^{-1}$ [2].

## 3.2 Discussion

The average yield of flying fish roe filaments ranges from 86.63 % to 89.20 %, the average yield of flying fish roe filaments has increased and decreased insignificantly. The longer the extraction time, the higher the yield [31]. However, the yield obtained with the long extraction time treatment is 5 h (86.63 %) less than the long treatment of extraction time 1 h (89.20 %) and 3 h (89.35 %). Decreased yield in flying fish roe filaments after defatting can be caused by the extraction method used, sample particle size, storage conditions and time, length of extraction time, and comparison of the number of solvents and materials used [4, 5]. During the extraction process, flying fish roe filaments are separated from flying fish roes that are still attached. So, the yield obtained from the extraction results of a long time of 5 h is lower than the treatment of 1 h and 3 h. This study has a higher yield than previous research [2], that uses 20 min of extraction (73.52 % ± 0.07 %). The greater the volume of the solvent and the longer the extraction time, the greater the yield value.

### *3.2.1 Total volatile base (TVB)*

The results of the study data total volatile base filaments of flying fish roes with control treatment and length of extraction time of 1 h, 3 h, and 5 h, namely 9.64 mg N 100 $g^{-1}$, 6.59 mg N 100 $g^{-1}$, 5.88 mg N 100 $g^{-1}$, and 4.94 mg N 100 $g^{-1}$. Based on the results of the data obtained the longer the extraction time of TVB content contained in the material is decreasing, so it is proven to reduce the fishy odor in the filaments of flying fish roes. Fishy odor in fish and its derivatives can be caused by the formation of volatile compounds derived from the oxidation of fatty acids [40]. During fish respiration, volatile compounds can be absorbed by water and stored under lipid tissues. Volatile compounds derived from lipids are aldehydes and ketones. TVB is a protein degradation compound that produces several volatile bases, such as ammonia, histamine, hydrogen sulfide, and trimethylamine, that cause a foul odor [41, 6].

One of the compounds that because fishy odor is trimethylamine oxide which is located naturally in the muscle muscles of fish. At the end of the biochemical process, fish meat will cause a fishy odor because it produces high enough trimethylamine and ammonia compounds caused by the rapid breakdown of proteins [42]. The limit of TVB levels declared rotten for fishery products is 30 mg N 100 $g^{-1}$ [43]. The results of TVB research data on fish roes fly lower than the threshold of TVB value of decay of fish-derived products. It proves that the filament of the flying fish roe control treatment and the length of extraction time of 1 h, 3 h and 5 h does not experience decay or damage physically and chemically. This result is according to previous research about the length of interaction between fat and nonpolar solvent [2].

### *3.2.2 Fat content*

The highest fat content of flying fish roe filaments is in the control treatment of 11.05 %. As for the lowest fat content, there is in the treatment of the long extraction time of 5 h which is 8.65 %. The acquisition of fat levels with long extraction times between 3 h and 5 h does not differ much. The results of the fat content of flying fish roes in this study is relatively high because it has a fat content of more than 7 %, so it is necessary to remove fat by using the defatting method. Isopropyl alcohol and ethanol are solvents often used to extract fish protein concentrates. Alcohol solvents can increase protein levels and lower fat levels. Oils and fats have common properties that dissolve in non-polar solvents such as ether, benzene, and chloroform. However, several types of fats can dissolve in alcohol solvents. Polar fat groups include phospholipids, glycolipids, and proteolipids [44, 12]. The extraction process is carried out only once. It can be the cause of the fat content of the roes of flying fish extracted is still relatively high. The extraction process that is done repeatedly will be able to help reduce fat levels in the material. Other factors that affect fat levels are the extraction method, the way of drying, and the length of extraction time [45]. Compared with previous research, this study showed that defatting proved effective in reducing the fish odor. The fat content without defatting was reported as 37.57 % [5].

### *3.2.3 Protein content*

The average protein content of flying fish roe filaments with various treatments (47.49 %; 31.94 %; 39.24 %; 47.93 %) is lower than the results of the study by Azka *et al.* [5] which is 59.69 % and previous research which is 73.80 % [4]. It can be due to several factors in which the fish species are used, and the solvent used during the extraction process. Isopropyl alcohol is a solvent used when defatting some organic solvents that are polar so that some proteins

are dissolved during the extraction process. Previous study Kumoro *et al.* [46] stated that the extraction process using alcohol solvents would increase protein levels and lower fat levels in the material. However, low protein levels can be caused by the extraction process done only once without any repetition, so it is less maximal in producing flying fish roe filaments with high protein and low-fat content.

### *3.2.4 Moisture content*

The study data results showed that the percent of moisture content contained in the filaments of flying fish roes after going through the defatting extraction process decreased compared to the filaments of flying fish roes control treatment. For example, the moisture content of the filament of the flying fish roe treatment of control by 16.37 % after going through the defatting extraction process with long treatment extraction times of 1 h, 3 h and 5 h is 11.65 %; 9.15 %, and 10.05 %.

The long treatment extraction time affects the moisture content in the filaments of flying fish roes. In the extraction process carried out using isopropyl alcohol solvent which is a polar organic solvent so that the water contained in the filament of flying fish roes is dissolved [30]. In addition, the decreased moisture content is affected by drying factors carried out after the extraction process. Drying is conducted using an instrument cabinet dryer. Drying is done to evaporate the solvent isopropyl alcohol on the filament of the flying fish roe after going through the extraction process so as not to odor strong isopropyl alcohol. In addition, it is also used to evaporate the volatile content that resides in the filaments of flying fish roes.

### *3.2.5 Ash content*

The results of the analysis of the ash levels of flying fish roes with control treatment, the length of extraction time of 1 h, 3 h, and 5 h in a row are 8.85 %; 9.95 %; 7.75 % and 7.5 %. The ash levels of long extraction treatments of 1 h, 3 h, and 5 h are not significantly different, while the three treatments differ markedly from the control treatment. The results of the ash level of the control treatment were the same as the results of the study by Azka *et al.* [5] which is 8.28 % and higher than the ash levels of flying fish roes (6.62 %). Flying fish roes contain the most sodium (Na) and potassium (K) minerals [4]. In addition, the mineral chloride (Cl) can also be said to be high, this is because the fibers of flying fish roes come from seawater rich in NaCl. The average ash content of protein hydrolysis in fish is 1.76 % to 25.94 % [47].

### *3.2.6 Amino acid profil*

The highest essential amino acid content in flying fish roe filaments is valine at 5.95 %, while the highest non-essential amino acid is glutamic acid at 10.36 %. Valine is an essential amino acid that can increase energy and endurance, lower blood sugar levels, and grow [48]. Glutamic acid is a non-essential amino acid that supports brain function, strengthens memory, speeds the healing of wounds in the intestines, and dampens emotions. The highest level of essential amino acids in the protein concentrate of flying fish roe filaments is lysine at 149.75 mg 100 $g^{-1}$. The highest non-essential amino acid is glutamic acid at 64.61 mg 100 $g^{-1}$. Lysine is an essential amino acid that plays a role in the growth and prevention of osteoporosis [48, 36]. In addition, the amino acid lysine helps increase calcium in the body and maintain mineral excretion in the body. Lack of amino lysine acid will impact the body, such as impaired growth [49, 48, 13, 14].

## 4 Conclusion

There is a long-time influence of defatting extraction time on moisture content and ash levels resulting from the extraction of flying fish roe filaments with control treatment, 1 h, 3 h, and 5 h in a row, namely moisture content (16.37 %; 11.65 %; 9.15 %; 10.05 %) and ash levels (8.85 %; 9.95 %; 7.75 %; 7.5 %). The length of time of defatting extraction did not affect protein content (47.20 %; 31.94 %; 39.24 %; 47.96 %) and fat content (11.05 %; 9.2 %; 8.75 %; 8.65 %). The amino acids found in the filaments of flying fish roes and the protein concentrates of flying fish roe filaments are nine types of essential amino acids consisting of histidine (2.24 % and 17.71 mg 100 $g^{-1}$), threonine (2.79 % and 56.25 mg 100 $g^{-1}$), valine (5.95 % and 65.50 mg 100 $g^{-1}$), methionine (0.66 % and 2.16 mg 100 $g^{-1}$), I-leusine (1.39 % and 34.03 mg 100 $g^{-1}$), leusine (3.99 % and 67.49 mg 100 $g^{-1}$), phenylalanine (2.90 % and 37.94 mg 100 $g^{-1}$), lysine (3.37 % and 149.75 mg 100 $g^{-1}$), arginine (1.49 % and 21.79 mg 100 $g^{-1}$) and six types of non-essential amino acids consisting of aspartic acid (4.00 % and 61.37 mg 100 $g^{-1}$), glutamic acid (10.36 % and 64.61 mg 100 $g^{-1}$), serine (6.70 % and 47.52 mg 100 $g^{-1}$), glycine (3.85 % and 54.88 mg 100 $g^{-1}$), alanine (3.97 % and 43.34 mg 100 $g^{-1}$), tyrosine (5.49 % and 60.80 mg 100 $g^{-1}$). There is a long-time influence of defatting extraction on the Total Volatile Base (TVB) extract of flying fish roe filaments and the data generated with control treatment, extraction of 1 h, 3 h, and 5 h in a row is 9.64 mg N 100 $g^{-1}$; 6.59 mg N 100 $g^{-1}$; 5.88 mg N 100 $g^{-1}$ and 4.94 mg N 100 $g^{-1}$. The best length of extraction time is 5 h.

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