

Comparative Study of Nutritional Content and Amino Acid Profile on Rono Poso Fish (*Adrianichthys oophorus*) and Rono Lindu Fish (*Oryzias sarasinorum*) Endemic of Central Sulawesi

Jamaluddin^{1*}, Meita¹, Syariful Anam¹, Agustinus Widodo¹, Pitriani²

¹ Department of Pharmacy, Mathematics and Natural Science of Faculty, Tadulako University, Palu, Central Sulawesi, Indonesia

² Department of Environmental Health, Public Health Faculty, Tadulako University, Palu, Central Sulawesi, Indonesia

* Corresponding Author. E-mail: jamal_farmasi02@yahoo.co.id

Received: 12 September 2021 / Revised: 01 November 2021 / Accepted: 12 January 2022

ABSTRACT: Rono Poso fish (*Adrianichthys oophorus*) originating from Lake Poso and Rono Lindu fish (*Oryzias sarasinorum*) from Lake Lindu, is central Sulawesi endemic fish that consumed the communities around the lake, but it is not known nutritional content. This study aims to determine the nutrient content and amino acid profile in Rono Poso and Rono fish Lindu. Ash content analysis testing using the dry ashing method, testing the moisture content using the oven method, using soxhlet fat, and protein content using the method and the testing Kjeldhal amino acid profile using Chromatography High Performance Liquid (HPLC). The results showed that several nutrients and amino acid profiles differ between the two samples. In Rono Poso fish containing ash 11.00%, water 6.11%, 58.88% protein, fat, and carbohydrates 22.68% 1:33%. In Rono Lindu containing ash 18.84%, water 9.77%, 58.97% protein, fat, and carbohydrate 10:35% 2:07%.

KEYWORDS: Nutrient Content; Amino Acids; *Oryzias sarasinorum*; *Adrianichthys oophorus*.

1. INTRODUCTION

Indonesia is well-known as the largest archipelago country in the world and as the second country *Mega-biodiversity* after Brazil which has 1300 species of freshwater fish with 0.72 types of density / 1000 km² (FAO, 2010). In Sulawesi, freshwater fish recorded as many as 62 species and 52 species of them are part of endemic species (Kottelat et al. in Mamangkey, 2010)

Sulawesi is included in the region of Wallace which has been known as a center of biodiversity. This is due to the high degree of endemic fauna between the original (Whitten et al. in Mamangkey, 2010). The famous lake in Central Sulawesi is Poso Lake and Lindu Lake.

Soeroto, et al (1996) declared that new species have been discovered at Poso Lake, one of them was Rono Fish (*Adrianichthys oophorus*). This fish is an endemic fish of Poso Lake that has been identified its existence in 1988 by Kottelat. Rono Fish (*Adrianichthys oophorus*) is a small fish that has a beautiful color, greenish-black. Beside at Poso Lake, it is also located at Lindu Lake as endemic fish with other species, namely Rono Fish (*Oryzias sarasinorum*) (Lukman, 2002). However the nutritional content such as protein, carbohydrates, fats, vitamins, and minerals of the two types of fish is not known, so it is necessary to test the nutritional intake that will be consumed by the body.

Fish contains protein, fat, vitamins, excellent and prospective minerals for the human body (Panagan et al, 2012) and a certain type of fish has higher protein content than meat. Also, protein is an ingredient that forming new networks that always happens in the body. During the process of forming tissue growth occurs on a large scale. In every cell of his life, protein is a very important part of most tissues of the body. In addition to the protein content of the fish also contain fat.

The main role of fat in the diet is as an energy source. According to (Muchtadi, et al, 2000) Fat is a form of excess energy stored by the animal, so the amount of fat in animals used as food ingredients determined by the energy balance of the animal. In addition to fat, fish also contain carbohydrates. Carbohydrate is the body serves to prevent the onset of ketosis, preventing excessive body protein breakdown, prevent the mineral loss, and to help metabolize fat as an energy source. Carbohydrates in the human body can be formed of several amino acids and glycerol fatty portions of Gandjar, et al (2007).

Besides protein, fat, and carbohydrates, fish also contain minerals as same as the number of minerals in milk such as calcium. Small fish and dried fish provide a high percentage of lime (calcium) derived from bones because these fish are eaten all over the body without being discarded. Calcium is a mineral that is most abundant in the body and calcium among other functions for the formation of bones, teeth formation, growth, muscle contraction, muscle flexing, maintaining body fluid balance, prevent osteoporosis (Anonymous, 2007).

Based on the description above, this research will test the content of the ash content, moisture content, carbohydrate, protein, and fat and amino acids of Rono fish taken from Lindu Lake (*Xenopoecilus sarasinorum*) and Poso Lake (*Xenopoecilus oophorus*).

2. EXPERIMENTAL SECTION

2.1. Materials

The materials that have been used in this study include: distilled (H₂O), phenol (C₆H₅OH), sulfuric acid (H₂SO₄), hydrochloric acid (HCl), alcohol (OH), sodium hydroxide (NaOH), hexane, methanol, boron trifluoride (BF₃), Florida saturated sodium (NaCl), n-hexane and the standard solution of fatty acids.

2.2. Methods

2.2.1. Sample preparation

This research used Rono Fish taken from Lindu Lake and Poso Lake as a sample. The Fish put in Styrofoam containers that had ice cubes before processing for extraction. Furthermore, the samples were washed and removed its entrails and then drained. Samples that have been drained and then dried using an oven at a temperature 60°C. Rono Fish taken from Lindu Lake dried for 24 hours and Rono Fish are taken from Poso Lake dried for 36 hours. After drying, the samples blended to a powder and stored at room temperature (20-25°C) in a steel container stainless (Endriyani in Mustard, 2013).

2.2.2. Ash content

As much as 2-3 grams of dried sample was heated with a Bunsen burning, then put in a furnace at a temperature of 50-60°C, and performed for 6 hours, thus incineration is perfect.

$$\text{Ash Content} = \frac{C - A}{B} \times 100\%$$

2.2.3. Water content

As much as 10 grams of wet samples included in the rate cup is heated at a temperature of 105°C for eight hours, included in the desiccators, and weighed to get a constant weight.

$$\text{Water Content} = \frac{(A + B)}{B} \times 100\%$$

2.2.4. Carbohydrate Content

$$\text{Carbohydrate} = 100\% - (\text{Ash}\% + \text{Water}\% + \text{Protein}\% + \text{Fat}\%)$$

2.2.5. Levels of protein

Weigh carefully 1 gram of trailer (for the high protein 0.3-0.5 grams) into the tube kjeltec. Added 2 grams of a mixture of selenium. Added 12 ml of concentrated H₂SO₄, In Turn, digestion block, destruction at a temperature of 400°C for 1 hour, In Turn off the digestion block, chill. Create sample sequences on kjeltec with the AN300 program. Install kjeltec funds to run a sequence. Make a blank determination (Jamaluddin., Nur Atina, and Yuyun Y, 2019).

$$\text{Protein} = \frac{(Vp - Vb) \times NP \times 1,4007 \times FK}{\text{sample weight (gram)}}$$

Description:

V_p : Volume pillar

N_p : Normality pillar

F_p : Dilution factor

F_k : Factor correction

2.2.6. Levels of fat

As much as 5 grams of sample is inserted into the lead and subsequently entered into the Soxhlet flask, N-hexane solvent is added and the heating process is performed for 8 hours.

$$\text{Levels of Fat} = \frac{C - A}{B} \times 100\%$$

Weighed 2-3 grams of blow-dry powder included in a paper sleeve (Hulls) and dried in an oven for ± 1 hour at a temperature <80°C. A sample contained in a paper sleeve is extracted by methods soxhletation using hexane solvent for ± 6 hours. Hexane solvent extracted and the extracted fat dried at 105°C. Subsequently, the oil extract is cooled in an exicator then weighed to extract heavy oils have fixed weights (Jamaluddin, Muhsinah, NA., and Widodo, A. 2019).

2.2.7. Amino Acid Testing

The sample solution was weighed 0.1 gram sample was added 5 ml of 6N HCl, vortex, is hydrolyzed to 22 hours at a temperature of 110°C, cooled, transfer to a 50 ml measuring flask, add aquadets to mark boundaries, filtered with a 0.45 µm filter, pipette 500 mL of filtrate AABA ± 40 µm add 460 mL of distilled water, pipette 10 mL of the solution, add 70 mL AccQ-fluorine borate, vortex, add 20 mL reagent A fluorine, vortex, let stand 1', incubation 10' at a temperature of 55°C, an injection at HPCL. A standard solution pipette 40µl standard mix of amino acids added 40 mL

of internal standard AABA, added 920 mL of distilled water, homogenize, taken 10 mL of standard, 70 mL AccQ-Fluor borate, vortex, let stand 1', incubation 10' at a temperature of 55°C, an injection HPLC (Jamaluddin, Mappiratu, Septiawan, and Yuyun Y., 2016).

$$\text{Amino Acid Testing (\%)} = \frac{\text{Rasioanalit sample (pmol)} / 1000000000 \times \text{BM} \times \text{fp} \times 100}{\text{Rasioanalit standard} \times \text{Sample weight (g)}}$$

3. RESULTS

Proximate Analysis Results of Rono Fish taken from Poso Lake

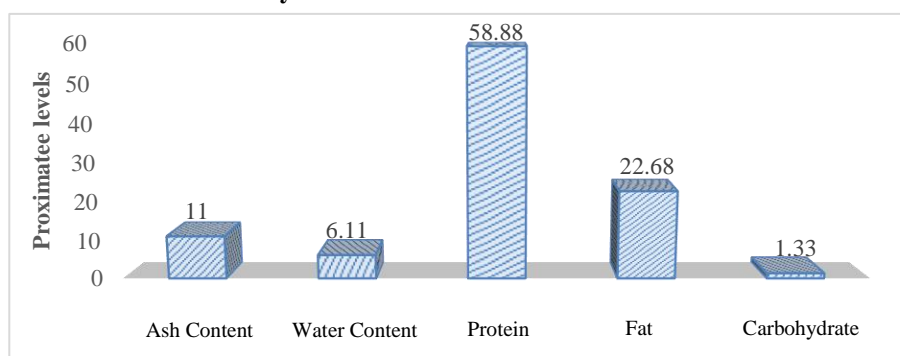


Figure 1. Proximate levels Fish Rono Poso

Proximate Analysis Results of Rono Fish taken from Lindu Lake

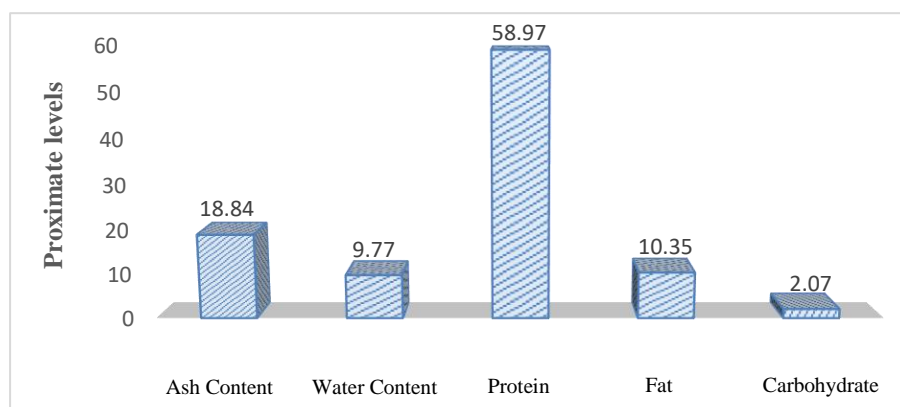


Figure 2. Proximate levels Fish Rono Lindu

Amino Acid Result Analysis of Rono Fish taken from Poso Lake

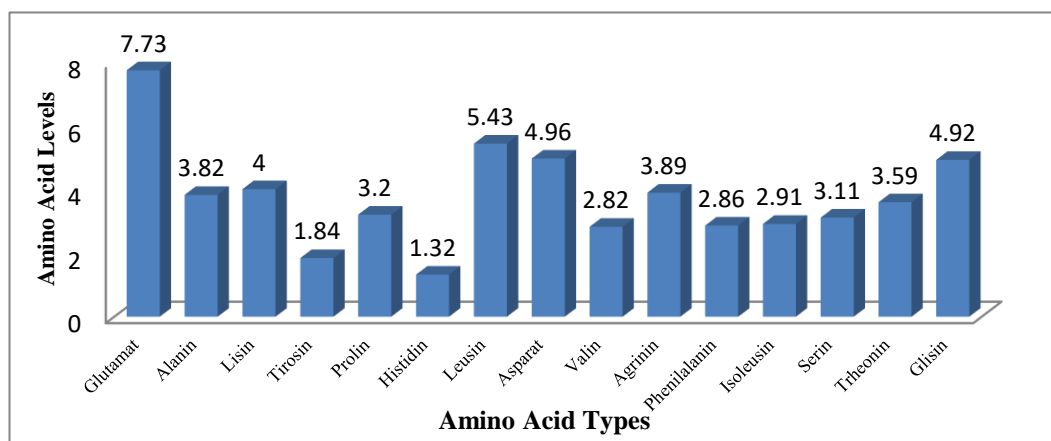


Figure 3. Amino Acid Levels Rano Poso

Amino Acid Result Analysis of Rono Fish taken from Lindu Lake

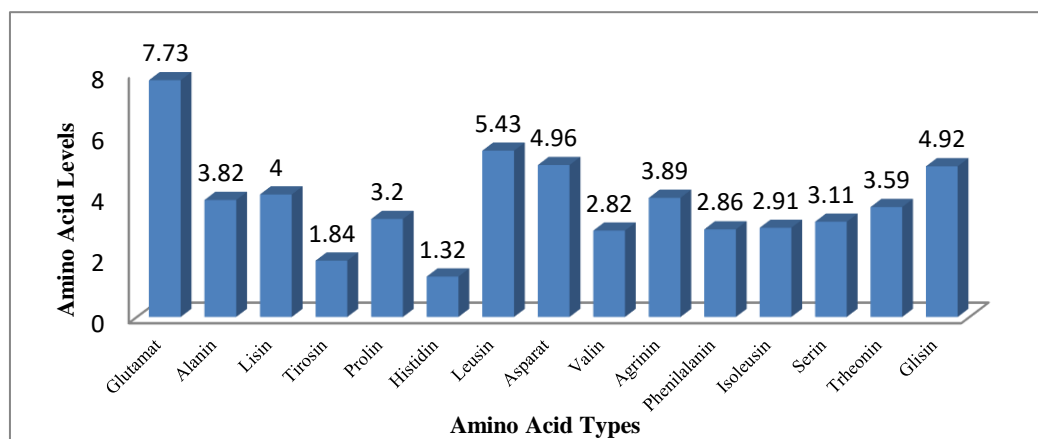


Figure 4. Amino Acid Levels Rano Lindu

4. DISCUSSION

This study was conducted to analyze the nutritional content such as ash, water, carbohydrates, protein, fat, and amino acid profile of the Rono fish taken from Lindu Lake (*Oryzias sarasinorum*) and Poso Lake (*Adrianichthys oophorus*), so it has a function as the nutritional value of Central Sulawesi endemic fish.

At the stage of the analysis of the ash content using dry ashing method or directly ashing was used for the determination of total ash content of foodstuffs and agricultural products, but it is also to get the maximum burning so only behind mineral elements left. The ash content is a mixture of inorganic or mineral components contained in food. Foodstuffs consist of 96% inorganic material and water, while the rest are mineral elements, the elements are also known as organic matter or ash. The ash content can show total minerals in foodstuffs. Organic ingredients in the combustion process will be burned but a not inorganic component, due to that is referred to the ash content. Determination of total ash can be used for many purposes, among others, to indicate whether or not a treatment, knowing the type of material used, and as a parameter determining the nutritional value of a foodstuff (Astuti, 2011).

The ash content that was obtained is different for each species. High and low levels of ash were caused by different types of organisms and the natural environment of the organism. Each organism has a different capability to regulated and absorbing metal, it essentially will affect the ash content in the materials (Rusyadi, 2006). The result of this research showed the different result of the ash content of the two types of Endemic fish of Central Sulawesi, Ikan Rono was taken from Lindu Lake (*Xenopoeilus sarasinorum*) and Poso Lake (*Xenopoeilus oophorus*). Ikan Rono taken from Poso Lake contained high levels of ash 11.00 (%) and Ikan Rono was taken from Lindu Lake contained 18.84 (%).

Phase analysis of moisture content using the oven method, the sample was heated at 105 ° C of temperature aimed to evaporate the water, desiccator was used as a coolant and as a residual remover to obtain a fixed or constant weight at the stage of this analysis. The water content is an important parameter in food as it can affect the texture, appearance, and flavor of food. The water content plays an important role in determining the lasting power of food as it can affect the physical properties, physical changes, changes in microbiological and enzymatic changes (Bucket *et al.* 1987). From this research, it can be stated that the content of water from the two types of Endemic fish of Central Sulawesi, Rono Fish taken from Poso Lake and Rono Fish taken from Lindu lake was Different. , Rono Fish are taken from Poso Lake contained water content amounted to 6.11 (%) and Rono Fish are taken from Lindu lake of 9.11 (%).

The Kjeldahl method was used to analyze the proteins. Kjeldahl is a simple method for the determination of total nitrogen in protein and nitrogen-containing compounds in foodstuffs, in the analysis process has three stages: destruction, distillation, and titration. From the results of the study, the content of protein from the two types of Endemic fish of Central Sulawesi, Rono Fish taken from Poso Lake, and Rono Fish was taken from Lindu lake was Different. Rono Fish taken from Poso Lake contained 29.62 (%) of protein Rono Fish taken from Lindu lake contained 11.26 (%). This difference was caused by several factors, including habitat, age, and food is digested (Winarno, 2008). The function of the protein in the body is as a builder of substance and guardian. The high levels of proteins show odds utilization as a source of animal protein.

Soxhlet method was used to analyze the lipid content. Soxhlet is the best way to extraction because the solvents that have been used can be recovered. The extraction phase is deemed complete when the solvent down to a fat pumpkin colorless (clear). Fat is an organic compound consisting of carbon atoms (C) hydrogen (H) and oxygen (O). Fat is soluble in fat solvents, such as benzene, ether, petroleum, and so on. Animal fats contain many sterols called cholesterol. Fat as a backup meal in the body, because the excess carbohydrates are converted into fat and stored in adipose tissue (Winarno, 2008).

Analysis of lipid aims to determine the fat content of food, there are many different methods of analysis of lipids, this study used soxhlet extraction methods. The results of the study stated that the total fat content of the fish of Endemic fish of Central Sulawesi, Rono Fish are taken from Poso Lake and Rono Fish taken from Lindu lake were Different. Rono Fish are taken from Poso Lake 29.62 (%) and Rono Fish are taken from Lindu lake (%). The differences between the fat content of the fish above can occur due to the influence of how factors, such as age and size of the habitat.

The results of carbohydrate content were obtained by the *difference* calculating, known as well as a *carbohydrate by the difference* that is by calculating the difference of the number 100 with the number of components of other materials (moisture content, ash content, protein content, and fat content). Carbohydrate is the main component of food which has the main function in processing food. Carbohydrates give sweetness to food, especially mono and disaccharides (Farida, 2006). Carbohydrates as energy-saving for plants. For humans, carbohydrates serve as a source of energy, materials forming the various compounds of the body, the material forming the essential amino acid, normal metabolism of fat, save the protein, enhance the growth of intestinal bacteria, maintain bowel movements (especially fiber), increase the consumption of protein, minerals, and vitamins B (Winarno, 1997). From the results of the study stated that the content carbohydrate of the fish of Endemic fish of Central Sulawesi Rono Fish taken from Poso Lake and Rono Fish are taken from Lindu lake were Different. Rono Fish taken from Poso Lake contained 29.62 (%) of carbohydrates and Rono Fish is taken from Lindu lake at 11.26 (%).

High-Performance Liquid Chromatography (HPLC) method was used to analyze the amino acids. The amino acid is an organic component containing amino and carboxyl. The composition of the amino acid content can determine the quality of the protein. Proteins which contain all the essential amino acids in the necessary quantities body (Winarno, 2008). Amino acids are usually soluble in water and insoluble in non-polar organic solvents are ether, acetone, and chloroform (Sitompul, 2004). because based on the chemical properties of amino acids is a weak base, so that first performed the hydrolysis aims to generate free amino acids (Winarno, 2008).

After the results of the hydrolyzate was filtered, then a solution of internal standard or AABA (α -amino-N-butyric acid) was added for correction factor error volumetric during the sample preparation and correct the loss of amino acid residues during the hydrolysis process that will be detected by reduced internal standard, so the use of an internal standard solution can improve precision. Once that is done by adding a derivatization process AccQ 70 mL and 20 mL borate Fluor fluorine reagent An into the filtrate. derivatized amino acids first to form a derivative that can fluoresce, so when the ongoing analysis process can improve the detection, change the molecular structure or polarity of the analyte so that it will produce a peak chromatograms better.

Not all amino acids can be made in our body when the terms of the formation of amino acids are divided into two groups: the exogenous and endogenous amino acids. Exogenous amino acids also are known as essential amino acids and endogenous amino acid is also called non-essential amino acids. Essential amino acids are amino acids that can not be made in the body and must be obtained from food sources of a protein called amino acids also exogenous whereas non-essential amino acid is an amino acid in the body can debut (Winarno, 2008).

5. CONCLUSION

From the research results, it was concluded that can be obtained were both amino acids from both samples of Rono fish taken from Lindu Lake and Poso Lake as many as 15 types had different content. 15 types of amino acids had the highest levels of amino acid glutamate, Rono Fish were taken from Poso Lake derived amino acid glutamate levels of 64869.84 ppm, and Rono Fish are taken from Lind Lake amounted to 77369.33 ppm. the amino acid histidine had the lowest level, in Rono Fish taken from Poso Lake 18777,96 ppm dan Rono Fish taken from Lindu Lake 13215,01 ppm.

Acknowledgments: The authors thank Chemistry Laboratory Hasanudin University, Makassar, Indonesia for the analysis of nutrient content and amino acid profile in Rono Poso and Rono fish Lindu.

Author contributions: Concept – J., S.A.; Design – J., A.W.; Supervision – P.; Materials – M; Data Collection and/or Processing – J., A.W.; Analysis and/or Interpretation – S.A., P.; Literature Search – M., S.A.; Writing – P.; Critical Reviews – J., A.W.

Conflict of interest statement: The authors declare no potential conflict of interest.

REFERENCES

- Apriyantono, An et al. Hint 1989. Food Analysis Laboratory. IPB, Bogor.
- Almatsier S 2005, the Basic Principles of Nutrition, PT Gramedia Pustaka Utama, Jakarta
- Anonymous, 2007, Nutrition and Public Health Faculty of Public Health, Jakarta
- Astuti. 2011. Abu Kadar. [http://astutipage.wordpress.com/tag/kadar ash /](http://astutipage.wordpress.com/tag/kadar%20ash/). Retrieved March 12, 2020, at Makassar
- Astawan, M., 2008. Noodle Making dan Bihun. Penebar Swadaya, Jakarta.
- Atkins, C, Robert, 2007, Atkins Diet, Alex Media Komputindo PT Gramedia Group, Jakarta
- Coal, .U.N 2009, Analysis of Protein, Calcium, And Fats In Fish Pora-Pora, thesis: Faculty of Public Health, University of North Sumatra, Medan
- Buckel., 1987. Food Science, Hayyuningsih, D. R. W., Sarbini, D and Kurnia, P., Translator, University of Indonesia. Jakarta
- FAO-fisheries and Aquaculture Department. 2010. The state of world fisheries and aquaculture 2010, the Food and Agriculture Organization of the United Nations. Rome. 197p.

- German, p.m, 1998, Introduction to Food Science, Nutrition and Microbiology, Translator: Murdijati Gardjito, et al. Gadjah Mada University Press Yogyakarta.
- Gandjar, I.G., & Rohman, A., 2010, Pharmaceutical Analysis, Library Student, Jakarta.
- Hart, H, et al, 2003. Organic Chemistry Edisi Eleventh, PT.Gelora Literacy, Jakarta
- Havivariful 2010, Analytics, SSL HTTP: //www.scribd.com/doc/38043085/Analiti- SSA (Retrieved at 28th March 2020)
- Hutagalung, H 2004, carbohydrates, Faculty of Medicine, University of North Sumatra, North Sumatra.
- Jamaluddin, Mappiratu, Septiawan and Yuyun Y., 2016. Analysis of Fatty Acid and Amino Acid Profile of “Meti” Mussels (*Batissa violacea* L. Von. Lamarck, 1818) in La'a River of Petasia District North Morowali Regency. RASAYAN Journal of Chemistry. Vol. 9. No. 4; 673 – 679. https://rasayanjournal.co.in/admin/php/upload/78_pdf.pdf
- Jamaluddin., Nur Atina and Yuyun Y, 2019. The Analysis of Protein Level and Amino Acid Profile in Eels (*Anguilla marmorata* (Q.) Gaimard and *Anguilla bicolor*) of Lake Poso. Journal of Pharmacy and Nutrition Sciences, 9 (1): 32-37. <https://setpublisher.com/downloads/jpansv9n1a6/>.
- Jamaluddin, Muhsinah, NA., and Widodo, A. 2019. Comparative Study on Fatty Acid Profile of Eel Fish (*Anguilla bicolor*) in Elver Eel and Silver Eel Phase from Palu River and Lake Poso. *Research Journal of Pharmacy and Technology*, Vol. 12, No. 12. <http://rjptonline.org/AbstractView.aspx?PID=2019-12-12-26>
- Kordi. 2009. Maintenance jetted tilapia pond. Jakarta: PT Perca.
- Kottelat, M., A.J. Whiten., S. and S. N. Kartikasari Wirjoatmodjo. 1993. Freshwater Fishes of Western Indonesia and Sulawesi. Periplus Editions (HK) Ltd. (EMDI), Republic of Indonesia. 291 pp.
- Lukman, 2002, Characteristics of Air Quality area of Lake Lindu, Central Sulawesi. Proceedings of the National Seminar on Limnology. Research Center for Limnology-LIPI. It 109-117.
- Lukman, 2007, Lake Lindu shade that Merindu, LIPI Press, Jakarta, Page 21 -32.
- Mamangkey, J. J ., Biopopulasi 2010. Endemic Fish Butini (*Glossogobius matanensis*) In Towuti Lake, South Sulawesi. Dissertation (S3). Graduate School of Bogor Agricultural University. Bogor.
- Mustard. 2013. Study Making Shredded Fish Cork (*Ophiocephalus striatus*) As the Food Supplements (Food Supplement) Study Of Making Snakehead Shredded (*Ophiochpalus striatus*) As Food Supplement. Essay. Program of Food Science and Technology, Department of Agricultural Technology, Faculty Pertanian Universitas Hasanuddin, Makassar.
- Muchtadi MS, et al, 2000. Substance Nutrient Metabolism, Pernebit Pustaka Sinar Harapan in Jakarta
- Muliani, 2012, sports Increase Calcium Absorption Mechanism, Faculty of Medicine, University of Udayana, Bali.
- Panagan A. T., et al., 2012. Quantitative and Qualitative Analysis of Unsaturated Fatty Acids Omega-3, Omega-6 and Characterization of Oil Catfish (*Pangasius pangasius*). Science Research Journal Volume 15 Number 3
- Rasmina, 2014. Study Ekobiologi *Xenopocilus sarasinorum* Endemic Fish Lake Lindu As Basis For Raising, Thesis: Faculty of Animal Husbandry and Fisheries Tadulako University, Palu
- Rendi, 2013. Domestication *Xenopocilus oophorus* Endemic Fish Lake Poso In Controlled Containers With Different Feed dosage, Thesis: Thesis: Faculty of Animal Husbandry and Fisheries Tadulako University, Palu
- Saanin, H. 1968. Taxonomy and Key Identify Fish. Volume 1. Bina Cipta. Bandung. 508
- Sediaotama, A.D. 1997. Ilmu Diet Nutrition And Science In Tropical Regions, publisher Balai Pustaka Jakarta.
- Soeroto, B, F, Tunga., 1996. The Island Fishes and The Distribution of Adrianichthyoidea of Sulawesi Island, With Special Comment on The Endangered Species in Lake Poso In Kitchener. D. J and A. Suyanto. Petroleum Pty Ltd. Melbourne. Australia. 5 pp
- Sudarmadji, S.B., Haryono, and Suhardi. 1997. Analysis Procedure For Foodstuff and Agriculture. Liberty. Yogyakarta.
- Winarno, F.G. Food and Nutrition Chemistry, Gramedia PT. Pustaka Utama, Jakarta, 1997, Edition VIII, Page 80. <https://www.goodreads.com/book/show/6044215-kimia-pangan-dan-gizi>
- Winarno, F.G.. Food and Nutrition Chemistry. M. Brio Press, Jawa Barat, 2008, Edition XI, Page 250 – 253. <https://id.mbrifofood.com/product-page/kimia-pangan-dan-gizi>