IJBPAS, April, 2022, 11(4): 1847-1852

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

QUANTITATIVE DETERMINATION AND COMPARATIVE ANALYSIS OF AMINO ACID COMPOSITION OF BARALI (*Wallago attu*) IN FRESH AND ICE-PRESERVED CONDITION

BASUMATARI D[1*], SHARMA R[2] AND DOLOI D[3]

1: Assistant Professor, Department of Zoology, Cotton University, Guwahati, Assam-781001, India

2: Research Scholar, Department of Zoology, Cotton University, Guwahati, Assam-781001, India

3: Research Scholar, Department of Zoology, Cotton University, Guwahati, Assam-781001, India

[*]**Corresponding Author: Dr. Devjit Basumatari: E Mail: devajitbasu@yahoo.in**

Received 21st May 2021; Revised 22nd June 2021; Accepted 19th July 2021; Available online 1st April 2022

https://doi.org/10.31032/IJBPAS/2022/11.4.6018

ABSTRACT

The amount of protein in fish is influenced by fat and water content and as a whole all these are influenced by the various environmental conditions. It has been found that post-preservation, there are various changes observed in the fish irrespective of the types or methods of preservation. In the present study, *Wallago attu* was chosen for amino acid analysis in two conditions – fresh and ice-preserved (for 15 days in 0°C, -4°C, -10°C, and -20°C). The identification and quantification of different amino acids were made from the HPLC characteristics of authentic samples (Sigma). It was observed that preservation in ice leads to loss of amino acid content, with highest losses at -4°C. The reason for this loss is not entirely known.

Keywords: Amino acids, Ice-preserved, Post-preservation, *Wallago attu*

INTRODUCTION

Systematic investigations of the nutritive value of fish proteins were initiated by Drummond **[1]** in 1918, who isolated the protein of herring, cod, and salmon by means of heat coagulation in an acid medium, followed by consecutive extractions with water, hot alcohol, and ether. This protein extract was determined to be as effective as beef protein and superior to casein in promoting the growth of rates when fed at a 6% level. Drummond **[2]** came to the conclusion that because of its nutritive value fish should figure prominently in the diet of the British nation as a "substitute" for meat.

From this initial work, a series of studies proved that fish proteins are of the superior nutrients and it contains all the essential amino acids that are necessary for human nutrition. The interesting experiments of Wenderoth **[2]** (1960) have shown that premature babies show good response after being fed frozen and dried fish powder as a protein supplement to breast milk. Owing to supplement of fish meal powder in the poultry industry, importance has been laid on the quality and biological importance of fish protein. In order to understand the nutritive value of fish protein, it is desirable to establish its amino acid composition.

In order to fully evaluate the nutritive value of fish protein, it is desirable to establish its amino acid composition. Data published in the past 30 years employing chemical, enzymic, microbiological, and chromato-graphic methods for the amino acid determination, have been compiled and reviewed by Geiger **[3]** (1948) and others. Many of these data were obtained without the benefit of the more recent technical improvements, and besides that, the method of amino acid determination, as well as the sampling procedures and the method employed for the hydrolysis of the protein, frequently were unsatisfactory. In addition, the values presented by different investigators are not strictly comparable, since the variable no. protein nitrogen content of fish has not been considered in many cases in the calculation of the amount or proportion of amino acid present.

Each species appears to have a characteristics basic composition of its pool (Duchateau and Florkin 1954 & 1957) **[4, 5]**, (Block and Bolling, 1951) **[6]**. Histidine is a unique constituent which is entirely absent in most other animals, but quite high in carp (Yadav, 1950 **[7]**; Duchateau and Florkin, 1954) **[8]**, lamprey (Duchateau and Florkin. 1957) **[4]**, and the herring-mackerel-tuna group

(Shewan, 1955) **[8]**. In this latter case, histamine appears during the initial spoilage. This was confirmed for the herring by Hughes (1959) **[9]**.

Various types of fish protein have been analyzed for the content of essential amino acids in desirable concentration for human beings. Several researches studied the amino acid make up of fish and found that fish and fish products provide proteins of the finest nutritive value or quality, when evaluated on the basis of its quantities of essential amino acids (Geiger, 1962. Gopakumar, 1997) **[10, 11]**.

Iced-preservation of fish is a very common technique in India. It has been reported that after preserving fish in ice, there is a decrease in total nitrogen (TN) and non-protein nitrogen (NPN) (Reddy & Shrikar, 1991) **[12]** and denaturation of myofibrillar proteins (Fredrick & Thomas, 1985) **[13]**.

Barali (*Wallago attu*), the freshwater catfish, finds itself in high demand in the Indian market. However, due to unavailability of this species and depletion of fish stock in some areas, it has to be transported with the help of ice-preservation. It is therefore necessary to document the changes in the amino acid composition upon preservation of this fish.

MATERIALS AND METHODS

The amino acid composition of fresh samples, different ice preserved samples collected from the markets, as well refrigerated samples were determined by hydrolyzing the samples in 6N HCL for 24 h at 110^0 C. The acids were removed by vacuum evaporation, made up to a known value with 0.05N HCL and then analyzed by HPLC (Shimadzu) on an ion exchanged column and a fluorescence detector after converting to 0-phthalaldehyde derivatives. Tryptophan content of the samples were determined after alkali hydrolysis. The identification and quantification of different amino acids were made from the HPLC characteristics of authentic samples (Sigma).

RESULTS AND DISCUSSION

The present studies on the content of essential and non essential amino acids have provided interesting information on the quality of fishes available in the markets. The ice-preserved samples collected from different markets showed a loss of both varieties of amino acids. It is difficult to interpret how the loss of the content of amino acids is regulated during the course of its preservation. The ice preserved samples sold in different markets have no temperature control, and being strewn on the floor of the market place deteriorates their quality till furthers several microorganisms are available

in the keeping place which have direct effects on the protein quality. Further, the freezing and thawing process goes on continuously, as there is no attempt to keep the quality as well as the amount of ice and the fish. Further, transportation enhances the thawing time. Several external and internal factors lead to the deterioration of the quality as well as the amino acids content of the fish.

The control refrigeration under -4^0 C, -10^0 C and -20^0 C shows an effective technique of preservation where loss of both essential and non-essential amino acids are under controlled conditions. There are no statistically significant differences on the samples preserved in -10^0C and -20^0 C. However, a few amino acids showed a significant loss in - 4^0 C preservation for 15 days (Arginine, Threonine, Glutamic acid). Results are shown in **Table 1** and **Table 2**.

Table: 1 Amino acids composition of *Wallago attu* in fresh and ice-preserved condition (on 15th day in 0°C, -4°C, -10°C, and -20°C) (shown in mg/g). The values are mean value of 3 samples of same size and sex collected

		Iced preserved samples at different temperatures			
Essential amino acids	**Fresh fish**	**0˚C**	**-4˚C**	**-10˚C**	**-20˚C**
Arginine	***8.5 ± 1.5**	**5.0 ± 1.0**	**5.5 ± 0.5**	**6.5 ± 1.0**	**7.5 ± 0.5**
Histidine	**4.8 ± 2.0**	**3.0 ± .05**	**3.5 ± .05**	**4.0 ± 1.0**	**4.0 ± .05**
Isoleucine	**4.5 ± 0.5**	**2.0 ± 0.5**	**2.5 ± 0.5**	**4.0 ± 0.5**	**4.0 ± 0.5**
Leucine	***12.5 ± 1.5**	***7.0 ± 1.0**	**7.5 ± 0.5**	***10.0 ± 0.5**	**11.0 ± 0.5**
Lysine	***13.5 ± 1.0**	**8.0 ± 0.5**	***9.5 ±0.5**	**11.0 ± 0.5**	***12.0 ± 1.0**
Methionine	**3.2 ±0.5**	**2.0 ±0.5**	**2.0 ±0.5**	**2.5 ±0.5**	**2.5 ±0.5**
Phenylalanine	**3.5 ±0.5**	**2.0 ±0.5**	**2.0 ±0.5**	**2.0 ±0.5**	**2.5 ±0.5**
Threonine	**3.5 ±0.5**	**2.0 ±0.5**	**2.0 ±0.5**	**2.5 ±0.5**	**3.0 ±0.5**
Valine	***6.5 ±1.5**	**5.0 ±1.0**	**5.0 ±0.5**	**5.0 ±0.5**	**5.5 ±0.5**
Tryptophan	**2.0 ±0.5**	**1.0 ±0.5**	**1.0 ±0.5**	**1.5 ±0.5**	**1.5 ±0.5**
Non-Essential amino acids					
Aspartic acid	***12.0 ±2.0**	**7.0 ± 1.0**	**8.0 ± 0.5**	***9.0 ± 1.5**	**10.5 ± 0.5**
Asparagine	**1.5 ± 0.5**	**0.5 ±0.5**	**0.5 ±0.5**	**0.6 ±0.5**	**0.5 ±0.5**
B-Alanine	***7.0 ±1.0**	**3.5 ±0.5**	***4.0 ±0.5**	**4.5 ±0.5**	**5.0 ±0.5**
Glutamine	**1.5 ±0.5**	**1.0 ±0.5**	**1.0 ±0.5**	**1.0 ±0.5**	**1.0 ±0.5**
Glutamic acid	***13.5 ±2.0**	**7.0 ±1.5**	***9.0 ±1.5**	***11.0 ±1.5**	**12.5 ±2.5**
Glycine	***5.5 ±1.0**	***3.0 ±0.5**	**3.5 ±0.5**	**3.5 ±0.5**	**3.5 ±0.5**
Proline	***6.0 ±1.5**	***3.0 ±0.5**	**3.5 ±0.5**	**4.5 ±0.5**	**5.5 ±0.5**
Serine	***4.5 ±0.5**	***3.0 ±0.5**	**3.0 ±0.5**	**4.0 ±0.5**	**4.0 ±0.5**
Tyrosine	***5.0 ±0.5**	***3.5 ±0.5**	**4.0 ±0.5**	**4.5 ±0.5**	**4.5 ±0.5**

***Significantly different (P≤0.5) at 5% level**

Table 2: Showing % of amino acids composition of *Wallago attu* during frozen conditioned at different temperature. The % have been calculated taking the value of fresh fish and other values of different temperatures as shown in Table 1

		Iced preserved sample at different temperatures			
Essential amino acids	**Fresh fish (mg/g)**	**0˚C**	**-4˚C**	**-10˚C**	**-20˚C**
Arginine	**8.5 ± 1.5**	**58.8**	**64.7**	**76.4**	**88.2**
Histidine	**4.8 ± 2.0**	**62.5**	**72.9**	**47.0**	**47.0**
Isoleucine	**4.5 ± 0.5**	**44.4**	**55.5**	**88.8**	**88.0**
Leucine	**12.5 ± 1.5**	**56.0**	**60.0**	**80.0**	**88.0**
Lysine	**13.5 ± 1.0**	**59.0**	**70.3**	**81.4**	**88.8**
Methionine	**3.2 ±0.5**	**62.5**	**62.5**	**78.1**	**78.1**
Phenylalanine	**3.5 ±0.5**	**57.1**	**57.1**	**57.1**	**71.1**
Threonine	**3.5 ±0.5**	**57.1**	**57.1**	**71.1**	**85.7**
Valine	**6.5 ±1.5**	**76.9**	**76.9**	**76.9**	**84.6**
Tryptophan	**2.0 ±0.5**	**50.0**	**50.0**	**75.0**	**75.0**
Non-Essential amino acids					
Aspartic acid	**12.0 ±2.0**	**58.3**	**66.6**	**75.0**	**87.5**
Asparagine	**1.5 ± 0.5**	**33.3**	**33.3**	**40.0**	**33.3**
B-Alanine	**7.0 ±1.0**	**50.0**	**57.1**	**64.2**	**71.5**
Glutamine	**1.5 ±0.5**	**66.6**	**66.6**	**66.6**	**66.6**
Glutamic acid	**13.5 ±2.0**	**51.8**	**66.6**	**81.4**	**92.5**
Glycine	**5.5 ±1.0**	**54.5**	**63.6**	**63.6**	**63.6**
Proline	**6.0 ±1.5**	**50.0**	**58.3**	**75.0**	**91.6**
Serine	**4.5 ±0.5**	**66.6**	**66.6**	**88.8**	**88.8**
Tyrosine	**5.0 ±0.5**	**70.0**	**80.0**	**90.0**	**90.0**

CONCLUSIONS

- Refrigerated samples preserved in -10^0C and -20^0 C did not show significant changes on the overall concentration of both essential and non-essential amino acids. However, preservation in -4^0 C showed the loss of both essential and non-essential amino acids.
- Ice-preserved marketed samples showed poor quality in terms of loss of both essential and non-essential amino acids. There is formation of histamine, which is a metabolic product of histidine.

REFERENCES

[1] Drummond, J.C.1918: The nutritire value of certain fish. *J. physiol.*, **52**, 95-109.

[2] Wenderoth, H. 1960: Fische in der Ernahrung von gesunden and kranken. In “Fett and Eiweiss in der

Ernahrung des gesunden and Kranken Menschen" (H.D. cresner, ed.). Liebniz University Giessen Germany.

[3] Geiger, G. 1948: Biochemistry of Fish protein. Fortschr.Chem.org Naturstoffe. **5**, 267-275.

[4] Duchateau, G., and M. Florkin 1957: Type de composition du pool d"acides amines non-proteiaues des muscles de lamproic. Arch. Intern. physical. et biochim., **65** (**2**), 378-379.

[5] Duchateau, G. and M. Florkin 1954: Constitution qualitative de la composante proteidique et non proteique du muscle de carpe. *Compt*. *Rend*. *Soc*. *biol*., **148**, 1287-1289.

[6] Block, R. J. and D. Bolling 1951: The Amino Acid Composition of Proteins and Foods. 1st ed., 576 pp. C Thomas, Springfield, Illinois.

[7] Yadav, N.A. 1950: Content of histidine, carnosine, and anserine in muscle of some fish. Doklady Akad. Nauk S. S. S. R. 70, 279-282

[8] Shewan, J. M. 1955: Nitrogenous entractires from fresh fish muscle III. Comparison of several flatfishes and members of the herring mackerel group. *J. Sci., Food Agr.,* **8**, 471 – 478.

[9] Hughes, R.B.1959: Chemical studies on the herring (Clupea harengus). II. Free amino acids of herring flesh and their behaviour during post mortem spoilage. *J. Sci*. *Food Agr*., **10**, 558 – 564.

[10] Geiger, G. 1962: Fish protein-nutritive aspects. In: Fish as Food, Ed G Borgstrom. Academic Press, New York. Vol. 2, 29.

[11] Gopakumar, K. 1997: Biochemical Composition of Indian Food Fish. Special Publication. Central Institute of Fisheries Technology. Cochin.

[12] Reddy, G. V. S. & Shrikar, L. N. (1991). Effect of ice storage on protein and related changes in pink perch (*Nemipterus japonicus*). J. Food Sci. Technol., India, 28(2), 101~104.

[13] Fredrick, W. W. & Thomas, B. L. (1985). Spoilage of marine and freshwater food products. In Processing Aquatic Food Products. John Wiley & Sons, New York, pp. 233-239.