

## Evaluation of Crude Protein and Amino Acid Analysis in the Scales of A Rohu Species, *Labeo rohita* Collected from Korangi Fish Harbor, Pakistan

<sup>1</sup>Zubia Masood, <sup>2</sup>Farhat Iqbal, <sup>3</sup>Muhammad Samee Haider, <sup>3</sup>Omer Mukhtar Tarar,  
<sup>3</sup>Lakht-e-Zehra, <sup>2</sup>Shagufta Saddozai, <sup>4</sup>Wali Muhammad Achakzai,  
<sup>2</sup>Wajeeha Razzaq, <sup>2</sup>Nighat Din, <sup>2</sup>Nosheen Rafique, <sup>5</sup>Nelofer Jamil and <sup>2</sup>Hina Gul Gharsheen

<sup>1</sup>Department of Zoology, University of Karachi-75270, Pakistan

<sup>2</sup>Department of Zoology, Sardar Bahadur Khan Women's University, Quetta, Balochistan, Pakistan

<sup>3</sup>Food and Marine Resources Research Centre,

PCSIR Laboratories Complex, Karachi, Off University Road, Karachi-75280, Pakistan

<sup>4</sup>Department of Zoology, University of Balochistan, Quetta, Pakistan

<sup>5</sup>Department of Chemistry, Sardar Bahadur Khan Women's University, Quetta, Balochistan, Pakistan

**Abstract:** The present study was aimed to evaluate the amount of crude protein and amino acids found in the scales of *Labeo rohita* belongs to the family Cyprinidae and are commonly known as 'Rohu'. About eighteen amino acids were analyzed in the present study. The obtained results revealed that the scales of *Labeo rohita* contain 82.29 % of crude protein (CP) on dry weight basis. Among all the evaluated amino acids, both glycine and proline were present in highest concentration as compare to the other amino acids, while Cysteine, methionine and tryptophan were totally absent in the crude protein extracted by scales of *Labeo rohita*. Thus, it had been concluded that scales of *Labeo rohita* are rich source of protein and certain essential and non-essential amino acids that later could be extracted and used in the production of certain products for pharmaceutical and cosmetic industries.

**Key words:** Scales • Crude Protein • Amino Acids • *Labeo rohita*

### INTRODUCTION

The body of the fish is covered with tough stiff protein made structures called scales, which protect the fish from being injured by sharp objects in their natural environment and also from the attack of certain diseases and predator [1-3]. They also help in locomotion [4-5]. Scales display a wonderful combination of flexibility, strength and resistance. Fish scales are variable in their size, shapes, composition and arrangement among the different fish species or even a single fish also contain great variations in the scale microstructures that are found in the different body regions [4, 6-7].

According to the several previous investigations, fish scale is consists of 40-80% protein and 16 to 59% minerals on dry weight basis [8-10]. About 70% of organic portion of scale is consists of only collagen protein [11]. In addition, one more protein named as ichthylepidin had

also been reported in the scales of cartilaginous fishes for about 24% on dry weight basis [12].

*Labeo rohita* is belongs to the family Cyprinidae, which is included in the major carps of India and commonly used as food fish throughout the world. *Labeo rohita* is commonly cultured fish species that contributes about 35% of the total carp fish production in India [13]. Among other cyprinids, it is very appetizing and prominent fish species among other Indian major carps [14]. It is widely abundant fresh water fish species that occurs throughout the world [15]. *Labeo rohita* is selective in its feeding habits as it gives partiality to plants and animal protein diet [16]. Only cycloid type scales are observed on *Labeo rohita*, which contain irregular dorsal and smooth ventral surfaces [17].

As large amount of waste materials in fish market is generated by fish scales, because their processing is a serious problem due to their hard structures [18].

But nowadays, like other animal wastes, fish scales have been used commercially for the production of important materials in various fertilizers, pharmaceuticals and cosmetic industries, because they are cheap and rich source of protein and certain essential amino acids [1-3, 7]. In this regards, our present study was conducted to estimate the amount of crude protein and amino acids in the scales of *Labeo rohita* in order to evaluate that whether its scales can be cheap and rich source for essential and non-essential amino acids. This information could be valuable in the production of certain materials in industries such as, fishmeal, organic fertilizers, pharmaceuticals and cosmetics. Furthermore, this investigation could be an initiative for the usage of crude protein and amino acids obtained from fish scales for valuable purposes.

## MATERIALS AND METHODS

**Fish Sampling:** About 20 fish specimens of *Labeo rohita* were collected from the local market of Korangi fish harbor, Karachi on 15<sup>th</sup> March 2014. These specimens were instantly transferred to the laboratory of the department of Zoology, University of Karachi. Scales samples were obtained through direct scrap from the fresh and healthy specimens of *Labeo rohita*. Scales were air dried for two days to obtain constant weight. Then these dried samples of scales were converted into powder form in mortar, which was then later utilized to calculate the concentrations of crude protein and amino acids.

**Crude Protein Analysis:** The amount of crude protein in the scales of *Labeo rohita* was determined by using micro-Kjeldhal method followed by Zubia *et al.* [3, 7].

**Amino Acid Analysis:** Liquid chromatography was done for the evaluation of amino acids in the scales of *Labeo rohita* on the Amino Acid Analyzer by using the method followed by Zubia *et al.*[7].

## RESULTS AND DISCUSSION

In the present study, the amount of crude protein and amino acids in the scales of the *Labeo rohita* and were presented and recorded in Tables 1-2 and Figure 1, respectively.

**Crude Protein Content (CP):** The obtained result revealed that the concentration of crude protein in the cycloid scales of *Labeo rohita* was found to be 82.29% on dry weight basis, which was found to be high as compare to those of Silver carp, *Hypophthalmichthys molitrix* (43.43%) and low as compare to blue tilapia, *Oreochromis aureus* (88.65%) as previously reported by Zhang *et al.* [19] and Zubia *et al.* [3]. Furthermore, our present study was also indicating that though *Labeo rohita* scales were also rich source of protein content but variations in the protein content in the scales obtained from the different fish species might be due to their different feeding habits, seasons, age, size, stages of growth as well as the habitat of fish as previously reported by Saraswat [20].

**Amino Acid Contents:** Methods used for the analysis of amino acid contents of the scales of *Labeo rohita* allows to determine the concentration of eighteen amino acids. During the evaluation process, amount of amino acid in hundred gram of crude protein was calculated and represented in the Table 1. Among these eighteen extracted amino acids, threonine, valine, methionine,

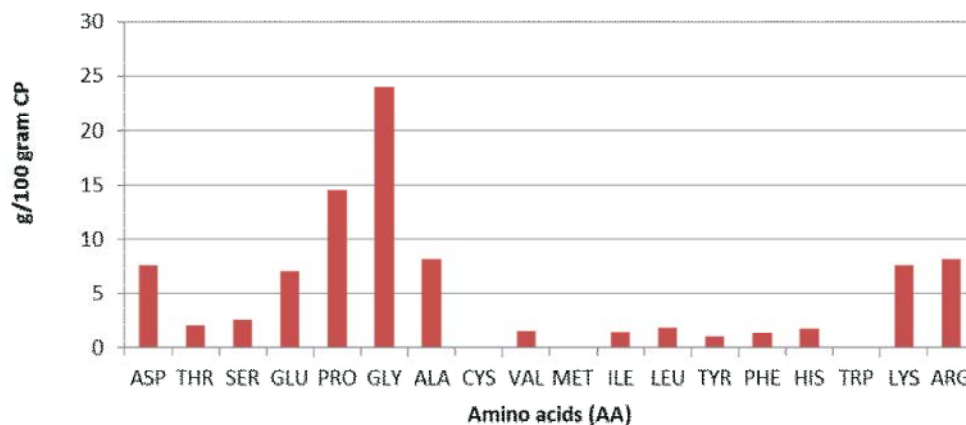


Fig. 1: Amount of cruid protein and amino acids in the scales of labeo rahita

Table 1: The concentration of amino acids in 100 grams of crude protein in the scales of *Labeo rohita*.

Amino acids	Code	Amino acid per 100 gram of crude protein (CP)
Aspartic acid	ASP	7.62
Threonine	THR <sup>a</sup>	2.09
Serine	SER	2.63
Glutamic acid	GLU	7.07
Proline	PRO	14.57
Glycine	GLY	24.04
Alanine	ALA	8.25
Cysteine	CYS	ND
Valine	VAL <sup>a</sup>	1.54
Methionine	MET <sup>a</sup>	ND
Isoleucine	ILE <sup>a</sup>	1.46
Leucine	LEU <sup>a</sup>	1.88
Tyrosine	TYR	1.05
Phenylalanine	PHE <sup>a</sup>	1.39
Histidine	HIS <sup>a</sup>	1.75
Tryptophan	TRP <sup>a</sup>	ND
Lysine	LYS <sup>a</sup>	7.54
Arginine	ARG	8.25
Total		91.14

Note: <sup>a</sup>shows essential amino acids.

Table 2: Total concentrations of Essential (EAA) and non-essential amino acid (NEAA) contents in the scale protein of *Labeo rohita* on dry weight basis

Amino acid (AA)	Amount of amino acids per 100 gram of crude protein (CP)
Total amino acid (TAA)	91.14
Total non-essential amino acids (TNEAA)	73.48
Total essential amino acid (TEAA)	
-With Histidine	17.65
-Without Histidine	15.9

isoleucine, leucine, phenylalanine, histidine, tryptophan and lysine were essential amino acids. The results of the present study revealed that Rohu scales contain about 91.14 grams amino acids in hundred grams of crude protein (CP) on dry weight basis. Among all the eighteen analyzed amino acids, glycine shows the highest value that is 24.01g/100g crude protein, while cysteine, methionine and tryptophan were totally absent in the crude protein extracted from the scales of *Labeo rohita*.

In the present study, the concentration of glycine and proline was high, which was in accordance with Zubia *et al.* [3, 7] who observed the similar results for the mullets and blue tilapia species. The investigations conducted by Bin Wang *et al.* [19] also showed the highest amount of glycine in the scales of Croceine Croaker (*Pseudosciaenacrocea*). Such highest amount of glycine and proline revealed that these two amino acids are responsible for collagen formation [20], therefore, scales of *Labeo rohita* was found to be contain highest collagen content.

While in contrast, very less amount of serine, threonine, leucine, histidine, valine, isoleucine, phenylalanine and tyrosine were also extracted from its scales. The presence of amino acids like tyrosine, histidine and tryptophan in least amount was actually indicating the occurrence of type I collagen in scales of this species [5, 21-22].

## CONCLUSION

From the obtained results, it had been concluded that revealed that though fish scales that were considered as a waste materials that are the major constituents of environmental pollution, but now after the analysis organic and inorganic components of fish scales revealed that fish scales can also be utilized as rich source of protein and certain amino acids as well as important minerals. Moreover, such information's will be valuable for the promoting the use of fish scales as animal derivatives in the preparation of various materials for fertilizers, cosmetics and pharmaceutical industries in the future.

## REFERENCES

1. Vernerey, F.J. and F. Barthelat, 2010. On the mechanics of fish scale structures. *International Journal of Solids and Structure*, 47: 2268-2275.
2. Zubia, M., Y.F. Rehana, S.M. Haider, O.M. Tarar, L. Zehra, M.U. Ain, H.U. Rehman, U. Asim, U.H. Ihsan, M. Bilal and Md. H. Yeamin, 2015. Comparative studies of the scale characters in four Mugilid species (Family Mugilidae: Order Mugiliformes) from Karachi Coast, Pakistan. *Biological Forum-An International Journal*, 7(1): 410-418.
3. Zubia, M., A. Malik, M.S. Haider, O.M. Tarar, L. Zehra, H.U. Rehman, N. Jamil, N. Din, W. Razzaq, N. Nazeer and Q. Jahangir, 2015. Evaluation of Crude Protein and Amino Acid Contents in the Scales of Blue Tilapia, *Oreochromis aureus* from Pakistan. *Global Veterinaria*, 14(4): 603-607.
4. Torres F.G, O.P. Troncoso and J. Nakamatsu, 2008. Characterization of nanocomposite laminate structure occurring in fish scales from *Arapaima giga*. *Materials science & Engineering C-Biomimetic and supramolecular Systems*, 28: 1276-1283.
5. Ikoma, T., H. Kobayasi, J. Tanaka, D. Walsh and S. Mann, 2003. Physical properties of type I collagen extracted from the fish scales of *Pagrus major* and *Oreochromis niloticas*. *International Journal of Biological Macromolecules*, 32(3-5): 199-204.

6. Kardong, K., 2008. Vertebrates: Comparative anatomy, function, evolution. 2<sup>nd</sup> edition. McGraw-Hill.
7. Zubia, M., Y.F. Rehana, S.M. Haider, O.M. Tarar, L. Zehra and Md. H. Yeamin, 2015. Evaluations of crude protein and amino acid contents from the scales of four mullet species (Mugilidae) collected from Karachi fish harbour, Pakistan. Indian Journal of Geo-marine Sciences, 44(5): (in press).
8. Nagai, T., M. Izumi and M. Ishii, 2004. Fish scale collagen. Preparation and partial characterization.. International Journal of Food Sciences and Technology, 39: 239-244.
9. Courtemanche, D.A., F.G. Whoriskey, V. Jr, Bujold and R.A. Curry, 2006. Assessing anadromy of brook char (*Salvelinus fontinalis*) using scales microchemistry. Canadian Journal of Fisheries and Aquatic Sciences, 63: 995-1006.
10. Mitsuhiro, O., T. Masaki, T. Motohiro, T. Tonegawa A. Hashimoto, H. Nobutaka and T. Ikoma, 2009. Elemental distribution analysis of type I collagen fibrils in tilapia fish scale with energy-filtered transmission electron microscope. Micron, 40(5-6): 665-668.
11. Pati, F., S. Dhara and B. Adhikari, 2010. Fish collagen: A potential material for biomedical application. Students' Technology Symposium (Tech. Sym.), IEEE. IEEE, pp: 34-38
12. Seshaiya R.V., P. Ambujabai and M. Kalyani, 1963. Amino acid composition of ichthylepidin from fish scales. In: Aspects of protein structure (Eds. G.N. Ramachandran), Academic Press, pp: 343-356.
13. FAO, 2001. Fishery Statistics. Food and Agriculture Organization of the United Nations, Rome: Vol., 92(2).
14. Abid, M. and M.S. Ahmed, 2009. Growth response of *Labeo rohita* fingerlings fed with different feeding regimes under intensive rearing. J. Animal & Plant Sci., 19(1): 45-49.
15. Khan, M.A, A.K. Jafri, N.K. Chadha, 2004. Growth and body composition of rohu, *Labeo rohita* (Hamilton), fed compound diet: winter feeding and rearing to marketable size. Journal of Applied Ichthyology, 20: 265-270.
16. Haniffa, M.A, A.G. Murugesan and A.T. Flemming, 1987. Influence of plant and animal food on food utilization of fresh water carp (*Labeo rohita*) Hamilton. Curr., Sci., 56(16): 846-848.
17. Tandon, K.K. and S.K. Sharma, 1977. The structure of the lateral line scales of some marine fishes of India. Vestnik. Cs. Spol. Zool., 41(3): 218-222.
18. Mejia-S. J.E., K.N. Waliszewski, M.A. Garcia, R. Cruz-Camarillo. 2006. The use of crude shrimp shell powder for chitinase production by *Serratia marcescens* WF. Food Technol. Biotechnol., 44(7): 646-651.
19. Bin Wang, W. Yu-Mei, C. Chang-Feng, L. Hong-Yu, D. Shang-Gui and M. Jian-Yin, 2013. Isolation and Characterization of Collagen and Antioxidant Collagen Peptides from Scales of Croceine Croaker (*Pseudosciaenacrocea*). Marine Drugs, 11(11): 4641-4661.
20. Zhang, J., R. Duan, Chao-Ye and K. Konno, 2010. Isolation and characterization of collagens from scales of Silver carp (*Hypophthalmichthys molitrix*). Journal of food chemistry, 34(6): 1343-1354.
21. Saraswat, R.C., 1976. Some organic constituents of scales of the fish *Channa punctatus* at different stages of m growth. Indian Journal of Fisheries, 23(1 and 2): 265-268.
22. Muyonga, J.H., C.G.B. Cole and K.G. Duodu, 2004. Extraction and physicochemical characterization of Nile perch (*Lates niloticus*) skin and bone gelatin. Food Hydrocolloids, 18: 582-591.