

Analysis of Protein Concentration in Different Tissues of Indian Major Carp *Catala catala*

Milind Kale

Department of Zoology, Govt. Vidarbha Institute of Science and Humanities (Autonomous) Amravati, Maharashtra, INDIA 444604

Email: [milindrck19\[at\]gmail.com](mailto:milindrck19[at]gmail.com)

Abstract: *Fish is highly nutritive, and easily digestible. A significant portion of the population of the world, particularly in developing nations, is very interested in it. Commercial fisheries, both in freshwater (about 10% of all catches) and in the oceans (about 90%), include wild fisheries and fish farms. Worldwide, about 500 million people are economically dependent on fishing. Fishes are regarded as an important high grade protein containing food staple of Indian people. In view of this, the investigations on different tissue contain protein concentration in fresh water carp Catala catala have been carried out. The changes in protein content of selected tissues like muscle, liver and gill were examined. The study revealed a highest percentage of protein content in muscle, liver and gill tissues was observed.*

Keywords: biochemical; body tissue. Introduction:

1. Introduction

Fish is highly nutritive, and easily digestible. A significant portion of the population of the world, particularly in developing nations. Every year the Indian seafood industry exports around 13.77.244 tonnes of seafood worldwide. As per studies the Indian seafood industry is expected to generate a revenue of \$10 billion by 2022.

The primary export destinations for Indian seafood are Asia, Middle East, USA, U.K., China and European countries. The major seafood producing states of India are Maharashtra, Goa, Karnataka, Tamil Nadu, Andhra Pradesh, Kerala and Gujarat. The seafood export industry contributes around 40% to the economy of the country (Seafood Exporters In India 2023 by moneeca moitra January 13, 2023). India is the third largest fish producing country and the world's second largest producer of fish, aims to increase export of fish and fishery product by more than 50% in value terms to reach Rs 1 lakh crore every year in the next five years from the current Rs 45,000 crore, animal husbandry and fisheries (minister Girijaj Singh 2019). India contributes about 7% to the global fish production. The 10% of the global fish biodiversity and is one of the 17-mega biodiversity rich countries. Around 14 million people are engaged in fisheries and its allied activities more than 30% of their animal protein needs (Uttam K Sarkar, JK Jena, Shri Prakash Singh, AK Singh and SC Rebello 2012). Moreover, fish is a considerable source of all amino acids. Hence, fish protein can be employed to enhance the overall protein quality of a varied diet and complement the amino acid pattern. Animal proteins are excellent forms of protein for people because they contain all the essential amino acids.

Protein is one of the most important constituents of meat products (Mahmudul et al., 2021). Animal foods, particularly lean meat, chicken and fishery products, are an excellent source of "high quality" protein for humans. (Animal Frontiers, Volume 2, Issue 4, October 2012). Improvements in diet composition are essential for improving animal welfare and health, as well as livestock output and performance. Meat and fish represent a valuable constituent

of most balanced diet. Consumption of different types of meat and fish has been associated with both beneficial and adverse health effects. While white meats and fish are generally associated with positive health outcomes, red and especially processed meats have been associated with colorectal cancer and other diseases, but it also has healthy outcomes.

The protein content estimation in the most repeated spectrometric methods for the quantification of soluble proteins are Biuret, Lowry's and Bradford's method. The optimum conditions for proteins quantification, using BSA (Bovine serum albumin) as standard proteins were studied. (Lowry OH, Rose Brough NJ, Bradford, (1976). The obtained results from spectrophotometric and literature methods were statically compared is an important and essential parameter to determine in order to ensure the quality and safety of food.

Practical terms such as the requirement of work standardization between laboratories have a comparative technique that essentially retests a measurement result (Yamamoto et al. 2009). Many total protein assays have a variable analytical response per unit of protein, depending on the type of protein. Out of 100 grams of body weight of fishes contain 38 grams of protein. According to dietary reference, 0.8 protein per kilograms of body weight or 0.38 gm per pound should consume daily means 46-56gm per day are required for human being (Kris Gunnars 2004).

Proteins in animal food products have been thoroughly separated, described, and identified using proteomics techniques (Boccaletto et al., 2018; Chandhini and Rejish Kumar, 2019) The most important processes in an electrophoretic analysis are sample preparation and extraction in order to get accurate results. The food industry has been driven to investigate various techniques for producing fish protein ingredient from various raw materials as a result of the growing understanding that dried fish protein can be used for food fortification and the creation of value added/functional meals. Scientifically proven health benefits of eating aquatic food, lipids, and oil have

significantly boosted the nutritional significance of aquatic food in recent years. Fish makes a significant contribution to our diet's provision of both macro and micronutrients (The State of World Fisheries and Aquaculture 2020).

A generic word for a deficiency of some or all of the dietary components essential for human health is malnutrition. Because fish protein has a high nutritional value and contains all the essential amino acids in the right amounts, it also has a high biological value (National Research Council (US)). The majority of people eat fish, which is a food contain high in protein concentration. Due to its economic value, easily accessibility, flavour, and palatability, fish is consumed by a higher percentage of people than due to its nutritional value (R.D Miles and F. A. Chapman 2).

Indian rivers are the major source of irrigation system, drinking water and fish as food. There are list of freshwater fishes in Indian river, few most popular names of freshwater fishes of India are Rohu, Catla, Mahseer, Magur, and Vaam.

The fresh water fish resource of Maharashtra constitutes 6 orders 25 families and 160 species. There are many species like Oriochromis, Grass carp, silver carp, etc. that have been introduced in the inland water of Maharashtra. The Maharashtra state often an excellent opportunity particularly in inland fisheries measures on Maharashtra include 16,000 km river.

Indian rivers are the major source of irrigation system, drinking water and fish as food. There are list of freshwater fishes in Indian river, few most popular names of freshwater fishes of India are Rohu, Catla, Mahseer, Magur, and Vaam.

The fresh water fish resource of Maharashtra constitutes 6 orders 25 families and 160 species. There are many species like Oriochromis, Grass carp, silver carp, etc. that have been introduced in the inland water of Maharashtra. The Maharashtra state often an excellent opportunity particularly in inland fisheries measures on Maharashtra include 16,000 km river.

2. Material and Method

Sample of Liver, Gills, Muscle of fish were collected from local market.

Chemicals and Reagents:

Biuret Reagent: Dissolve 1.5 g of copper sulphate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) and 4.5 g of sodium ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$) and 4.5 g of sodium potassium tartrate in 250 ml of 0.2 mol/litre sodium hydroxide; add 2.5 g of potassium iodide and makeup to 250 litre with 0.2 mol/litre sodium hydroxide. That is 500ml of Biuret reagent.

Protein Standard: 5 mg BSA/ml (for 10ml, 0.25g of BSA)

UV visible Spectrophotometer:

A number of methods have been used to determine protein concentration, which is based on UV-visible spectroscopy. These methods use either the natural characteristic of proteins to scatter light in the UV-visible region of the

electromagnetic spectrum, or they physically or chemically modify the proteins to make them absorb light in the UV-visible region. First, a curve of absorbance (Abs) versus protein concentration is prepared by using a series of protein solutions of known bovine serum albumin (BSA) concentrations, i.e., 0, 200, 400, 600, 800 and 1000 μg per mL (Figure 1). Then the absorbance of the solution being analysed, compared, and measured at the same wavelength of 750 nm, and the unknown protein concentration was determined from the calibration curve.

Preparation of Protein Sample:

Fresh samples of liver, muscles, and gills were collected from the market. Each sample was cut into small pieces. Then 3gm of each sample tissue was taken by the help of digital weighing scale. After weighing homogenised tissue with SSC buffer was transferred to centrifuge tube for centrifugation at 3000 rpm for 10 minutes. After the 10 minutes of centrifugation, supernatant was discarded and 2ml NaCl was added. Again the suspension was centrifuge at highest possible speed for 15 to 20 minutes. Then supernatant was collected.

Estimation of total protein content by Biuret method:

- Pipette out 0.0, 0.2, 0.4, 0.6, 0.8, 1, 1.2, 1.4, 1.6, 1.8 and 2ml of working standard in to the series of labelled test tubes.
- Pipette out 2 ml of the each given sample in another test tubes.
- Make up the volume to 2ml in all the test tubes. A tube with 2ml of distilled water serves as the blank.
- Now add 3 ml of Biuret reagent to all the test tubes including the test tubes labelled 'blank' and 'unknown'.
- Mix the contents of the tubes by vortexing / shaking the tubes and warm at 37°C for 10min.
- Now cool the contents to room temperature and record the absorbance at 540 nm against blank.
- Then plot the standard curve by taking concentration of protein along X-axis and absorbance at 540 nm along Y-axis.
- Then from this standard curve calculate the concentration of protein in the given samples.

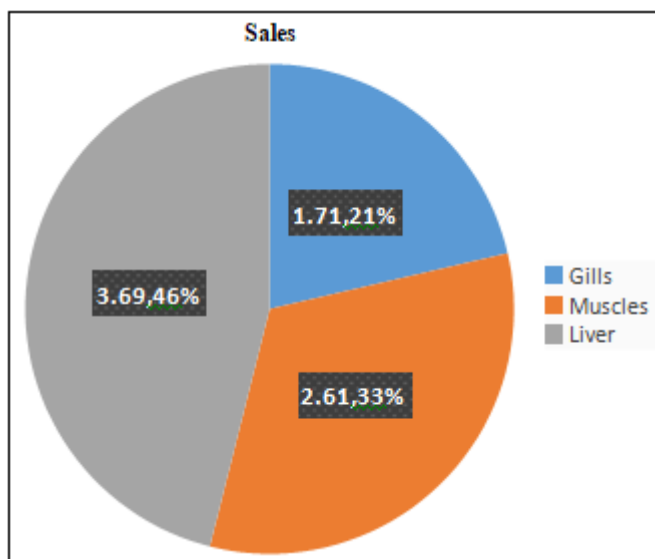
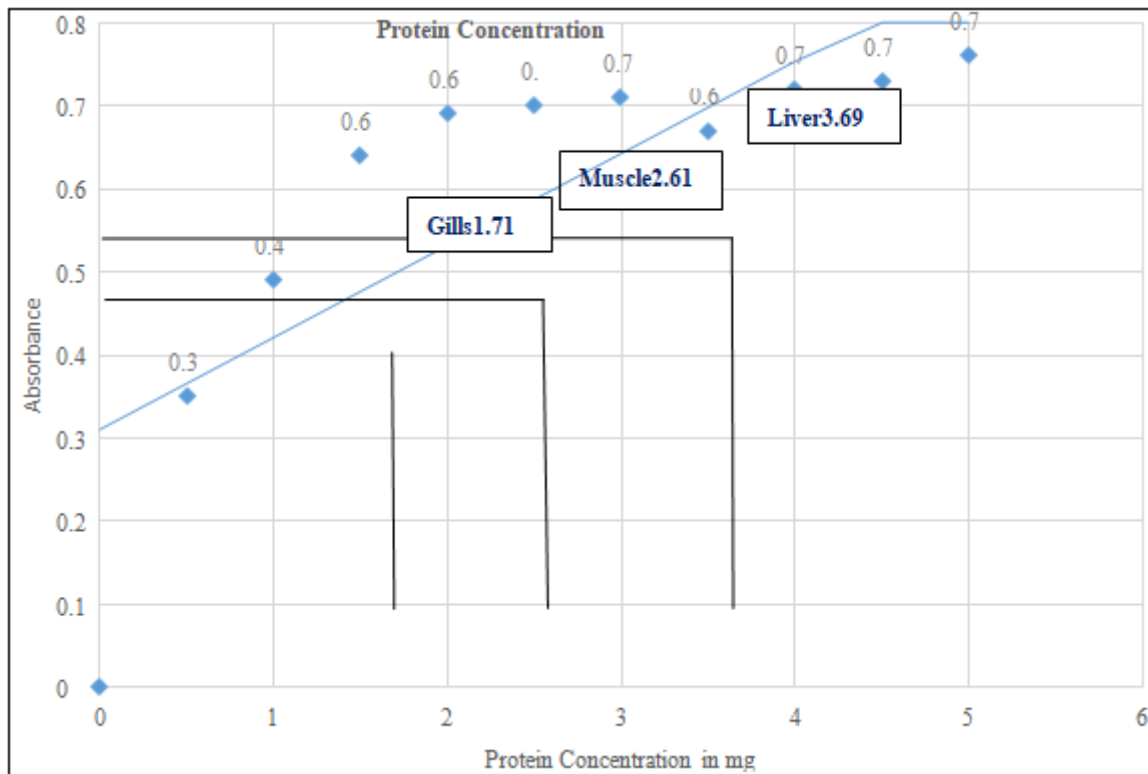
3. Result and Discussion

Table 1: Total protein contains in fish (*Catla catla*) in Muscle, Liver and Gills

Test Tube Number	Volume of Standard BSA (ml)	Distilled Water (ml)	Biuret Reagent (ml)	Absorbance at 750 nm
1	0.0	2	3	0.00
2	0.2	1.8	3	0.35
3	0.4	1.6	3	0.49
4	0.6	1.4	3	0.64
5	0.8	1.2	3	0.69
6	1.0	1.0	3	0.70
7	1.2	0.8	3	0.71
8	1.4	0.6	3	0.67
9	1.6	0.4	3	0.72
10	1.8	0.2	3	0.73
11	2	0.0	3	0.76

Table 1: Absorbance of different Bovine Serum albumin (BSA) concentration

Sr. no.	Sample	Absorbance (540nm)	Amount of Protein (mg)
1.	Muscle	0.60	2.61
2.	Liver	0.72	3.69
3.	Gills	0.5	0.60



Onyia, 2010). The protein performs a wide range of functions and provides energy. The protein requirement varies with many factors like age, body ability lactating women, the individuals like during infections and illness or stress. Fishes serves as a good source of nutritionally important biochemical constituents like carbohydrate, proteins and lipids. The protein content varies according to sex, size, food availability and seasonal maturity etc. Fish protein has since long been considered having high nutritional value (Sargent, 1997).

Nonetheless, only in the last decade, research has also

focused on the beneficial health effects of fish protein in human nutrition (Rudkowska et al., 2010; Pilon et al., 2011). Furthermore, fish protein hydrolysates are considered as superior from a nutritional point of view due to the excellent amino acid composition and easily digestible proteins.

Acknowledgement

I express my sincere thanks to the Head of the Department of Zoology Prof. Kishor G. Patil for the continue encouragement and support. I also express deep sense of gratitude to our Director Prof. Anjali Deshmukh.

References

- [1] International Journal of Science and Research (IJSR) ISSN: 2319-7064 ResearchGate Impact Factor (2018): 0.28 | SJIF (2019): 7.583 Volume 9 Issue 4, April 2020 Soumyashree Rath1 , Siba Prasad Parida2 Department of Zoology, School of Applied Sciences, Centurion University of Technology and Management, Bhubaneswar, Odisha, India.
- [2] The Pharma Innovation Journal 2022; SP-11(4): 2091-2095 Comparative analysis of different protein estimation methods.
- [3] Proteomics in fish health and aquaculture productivity management: Status and future perspectives. Sarika Jaiswal , Kiran D. Rasal, Tilak Chandra, Ratna Prabha, Mir Asif Iquebal, Anil Rai, Dinesh Kumar.
- [4] Protein Determination—Method Matters _ Hanne K. Mæhre,* Lars Dalheim, Guro K. Edvinsen, Edel O.

- Elvevoll, and Ida-Johanne Jensen VRSANSKA MARTINA¹, KUMBAR VOJTECH² ¹ Department of Chemistry and Biochemistry ² Department of Technology and Automobile Transport Mendel University in Brno Zemedelska 1, 613 00 Brno CZECH REPUBLIC. A
- [5] COMPARISON OF BIURET, LOWRY AND BRADFORD METHODS FOR MEASURING THE EGG'S PROTEINS.
- [6] Haifeng Wang, Zhi Yang, Huijuan Yang, Jing Xue, Yunyan Li, Shitong Wang, Lijun Ge, Qing Shen, Manman Zhang.
- [7] Comparative study of body composition of four fish species in relation to pond depth December 2006 International journal of Environmental Science and Technology.
- [8] Zekry, Abdelhalim & Abouelatta-Ebrahim, M.. (2015). LED Based Spectrophotometer can compete with conventional one. International Journal of Engineering & Technology. 4(2). 10.14419/ijet.v4i2.4504.
- [9] Aryal S. Spectrophotometer- Principle, Instrumentation, Applications | Instrumentation | Microbe Notes [Internet]. Microbe Notes. 2019 [cited 13 October 2019]. Available from: <https://microbenotes.com/spectrophotometer-principle-instrumentation-applications/>
- [10] Comparative study on nutritional composition of fish available in Portugal April 2019 Nutrition & Food Science Maria Ivone Santos Marques, Goreti Botelho
- [11] M Karthigarani, G Chelladurai, N Buvaneshwari Department of Zoology, Yadava College, India Department of Zoology, G Venkataswamy Naidu College, India. Comparative Study of Nutritional Evaluation between Three Types of Edible Marine Fishes of Landing Site of Tuticorin Coast and Marine Market Fishes of Madurai District.
- [12] Spencer H, Samachson, Fowler J, Kulka MJ (1971). Availability in man of protein and minerals from fish protein concentrate. *Am J Clin Nutr* 24(3): 311-317.
- [13] Jaclyn M, Stacey JB, Ashley SR (2010) Omega-3 fatty acids and pregnancy. *Rev Obstet Gynecol* 3(4): 163-171.
- [14] Spencer H, Samachson, Fowler J, Kulka MJ (1971). Availability in man of protein and minerals from fish protein concentrate. *Am J Clin Nutr* 24(3): 311-317.
- [15] Jaclyn M, Stacey JB, Ashley SR (2010) Omega-3 fatty acids and pregnancy. *Rev Obstet Gynecol* 3(4): 163-171.
- [16] Abdullahi, S. A., Abolude, D. S., & Ega, R. A. (2001). Nutrient quality of four oven dried freshwater catfish species in Northern Nigeria. *Journal of Tropical Bioscience*, 1(1), 70–76 Ababouch L. (2006).
- [17] Assuring fish safety and quality in international fish trade. *Marine Pollution Bulletin*, 53(10), 561–568
- [18] Ahmed M., Lorica M. H. (2002). Improving developing country food security through aquaculture development – Lessons from Asia. *Food Policy*, 27, 125–141.
- [19] A comparative study of the aquaculture practices adopted by fish farmers in Andhra Pradesh and West Bengal July 2010 TJ Abraham West Bengal University
- [20] Fishery training is provided by the Central Institute of Fisheries Education in Mumbai, which has ancillary institutions at Barrackpore in Uttar Pradesh and Hyderabad in Telangana.