Research Article

Enhancing Precision in Protein Estimation: A Comprehensive Analysis of Edible Meats and Cereals

Anuradha[1], Vinod Kumar Gupta[2]

[1,2]Rapture Biotech International Pvt Ltd, Noida, Uttar Pradesh, India.
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I N F O

Corresponding Author:
Anuradha, Rapture Biotech International Pvt Ltd,
D-201, D Block, Sector 10, Noida, Uttar Pradesh,
India.
E-mail Id:
nautiyalanuradha304@gmail.com
Orcid Id:
https://orcid.org/0009-0006-0601-9935

A B S T R A C T

Introduction: Protein is one of the main nutritional components of our life. It helps reduce many illnesses in the body. Protein contains genetic materials and also provides nourishment to cells. Here, we are studying cereals and meat varieties that contain the most protein. Based on our previous knowledge, we selected food sources that contain high amounts of protein such as green grams, black peas, goat liver, goat bone marrow, and fish muscle.

Method: In this research, we used Bovine serum albumin (BSA) as a standard protein. The Lowrey method was used to estimate the protein value in samples.

Results: The concentration of protein in goat bone marrow, goat liver, fish muscle, black peas, and green gram is 0.862, 0.497, 0.474, 0.105, and 0.018 mg/mL respectively. The highest protein concentration was found in goat bone marrow (0.862 mg/mL).

Conclusion: The study showed that goat bone marrow has the highest protein content as compared to goat liver, fish muscle, black peas, and green gram.

Keywords: Protein Quantification, Lowrey Method, BSA, Kwashiorkor, Standard, Deficiency, Metabolism

Introduction

The demand for a diet relies on numerous factors such as age, place, profession, and the fitness of an individual. Coronary artery diseases, obesity, elevated blood pressure, diabetes, and cancer are increasing due to fluctuating habits and increasing food adulteration.[1] The nourishment that can prevent disorders, protein, is one of them. Protein is a Greek word derived from 'proteios', which means 'primary' or of first importance. Proteins found in all animal and plant cells are nitrogen-containing biomolecules. In the body's waterless matter, protein constitutes nearly one-half.[2] Proteins offer nourishment for living organisms, act as genetic code, and play a vital role in cell metabolism, and that is the reason they are found in biological substances that are extremely important.[3] Around a billion people suffer when they are unable to meet their protein demand. In central Africa and south Asia, nearly 30% of young ones obtain very little amount of proteins in their food. Protein deficiency causes muscle wasting, but in critical conditions, it leads to kwashiorkor disease.[4] Protein deficiency affects most of the body functions, which are associated with many symptoms; some of these are listed here. The symptoms are associated with kwashiorkor even in slight deficiency of protein. Oedema is one of the important symptoms of protein deficiency, caused by the lack of Albumin which is found in blood and helps in the maintenance of blood circulation indirectly. Fatty liver is also a symptom linked with kwashiorkor. It causes accumulation in liver cells and

13

Anuradha & Gupta K V
Epidem. Int. 2023; 8(2)

finally leads to liver failure. Fatty liver mostly occurs in alcoholic people. Other symptoms are skin, hair and nail problems. As they are made up of proteins, a deficiency of protein affects them too. Children have peeling skin due to kwashiorkor which causes redness and patches of colored skin. Thinning and decolouring of hairs with excessive hair loss take place in that condition. Loss of muscle mass, greater risk of bone fractures and increased severity of infections are other symptoms of kwashiorkor which are caused by protein deficiency.[5]

The complex organic components, made up of a long chain of amino acids (which are bound together by peptide bonds) are proteins. Protein is a main ingredient of bones, cartilage, skin, blood, muscle etc., and synthesizes enzymes, hormones, and body chemicals. The body's roles, including modification of molecular reactions, DNA replication, responding to stimuli, providing structure to cells and organs, and transcribing molecules from one location to another, are performed by protein.[6]

Here, we selected some meat samples (goat bone marrow, goat liver, and fish muscle), alongside some cereals (such as green gram and black peas). Goats, notably the Black Bengal breed found in India possess desirable traits such as higher prolificacy, hostility against common diseases, easy adaptability to adverse environmental conditions, early maturity, high fecundity rate, and tender meat with low intramuscular fat.[7] Fish contains high-quality protein with different varieties of vitamins and minerals such as vitamin A, vitamin D, phosphorus, magnesium, selenium, and iodine, thereby providing significant nutritional value. Fish protein is easily digestible. Studies show that a small amount of fish protein can make up for the essential amino acids necessary for human health.[8] Green gram poses 20%–50% of total dry weight, in which globulin (60%) and albumin (25%) are the main storage protein. It is examined as an eccentric source of dietary protein. During sprouting, the breaking of the peptide bonds between amino acids is even higher.[9] Black peas, another protein-rich option, are particularly suitable for weight reduction diets, contributing 24.5%–28.4% of total nourishment in protein content.[10]

There are many ways of protein quantification, such as the Biuret method, Lowrey method, μ-Kjeldahl method, spectrophotometric method, BCA method, etc. For this study, protein analysis was conducted using the Lowrey method. The main purpose of the study is to determine and compare the protein content in different edible foods.

Materials and Method

Samples were collected from a nearby meat shop in Sector 10, Noida and cereals from a home kitchen in Sector 12, Noida in 2023. The study was conducted in Rapture Biotech International Pvt Ltd, Sector 10, Noida, India.

Requirements

UV visible spectrophotometer, centrifuge machine, Eppendorf, micropipette, test tubes, Sodium dodecyl sulfate (SDS), Folin–Ciocalteu, sodium carbonate, distilled water, and BSA

Protein Quantification by the Lowrey Method

Reagents

Reagent 1: Reagent A (2% Na_2CO_3 in 0.1 N NaOH) + Reagent B (a specified percentage of sodium potassium tartrate dissolved in 5 mL distilled water) + Reagent C (1% $CuSO_4.5H_2O$ dissolved in 10 mL distilled water)

Reagent 2: Diluted Folin–Ciocalteu reagent in a 1:1 ratio

Standard BSA concentration: 1 mg/ml[11]

Method

1. Pipette out 30, 60, 120, and 240 μL of BSA into different labelled test tubes in series.
2. Pipette out 100 μL of each sample into other test tubes.
3. Fill one test tube with 1 mL of distilled water, which serves as a blank. Fill another test tube with distilled water to make up the volume 1 mL.
4. Add 4.5 mL of reagent 1 to all the test tubes and incubate for 10 min.
5. Add 500 μL of reagent 2 to all the test tubes and incubate for 30 min in the dark.
6. Record the absorbance at 660 nm against a black background[11] (Table 1).

Table 1.Methodology of Lowrey Method

Tube	BSA Standard/ Sample (μL)	DW (μL)	Reagent 1 (mL)	Incubate for 10 min	Reagent 2 (μL)	Incubate for 30 min	Absorbance at 660 nm
Blank	0	1000	4.5		500		
T1	30	970	4.5		500		
T2	60	940	4.5		500		
T3	120	880	4.5		500		
T4	240	760	4.5		500		
GBM	100	900	4.5		500		

Anuradha & Gupta K V
Epidem. Int. 2023; 8(2)

14

| GL | 100 | 900 | 4.5 | | 500 | | |
FM	100	900	4.5		500		
BP	100	900	4.5		500		
GG	100	900	4.5		500		

GBM: Goat bone marrow, GL: Goat liver, FM: Fish muscle, BP: Black peas, GG: Green grams

Results and Discussion

This study aimed to compare the protein amount of different edible meat and cereals. The results showed the following protein amounts, ranked from highest to lowest: goat bone marrow (0.862 mg/mL), goat liver (0.497 mg/mL), fish muscle (0.474 mg/mL), black peas (0.105 mg/mL), and green gram (0.018 mg/mL).

BSA served as the standard protein, with a concentration of 1 mg/mL. This means that 1 mg of protein is present in 1 mL of the BSA sample.

The equation used to calculate the relationship between the protein content (y) and the concentration (x) is represented as follows: $y = 4.4083x + 0.021$ (Figure 1).
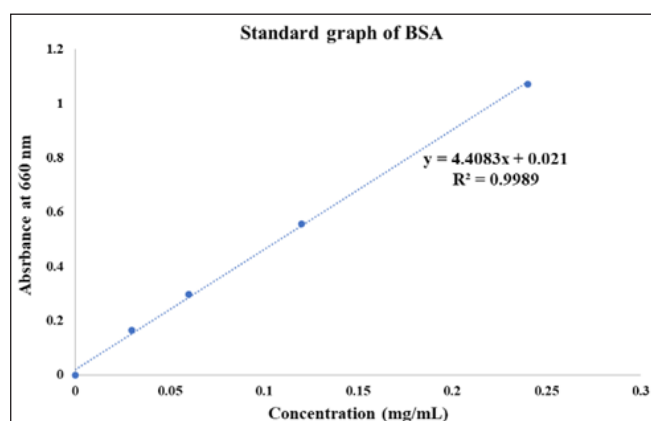
Figure 1.Standard Curve of Bovine Serum Albumin (BSA) (30–240 μL), Optical Density (OD) at 660 nm

The amount of protein present in samples is shown in Table 2.

Table 2.Protein Content in Samples

S. No.	Sample	Absorbance at 660 nm	Concentration (mg/mL)
1	GBM	0.965	0.862
2	GL	0.6	0.497
3	FM	0.577	0.474
4	BP	0.208	0.105
5	GG	0.085	0.018

GBM: Goat bone marrow, GL: Goat liver, FM: Fish muscle, BP: Black peas, GG: Green grams

Protein is very important for the growth of human muscles and is essential for a healthy body. High-protein foods can help to reach fitness goals. Globally, in 2019, there were 147,672,757 cases of protein-energy malnutrition, with a confidence interval between 130,405,923 and 167,471,359. The associated deaths were 212,242, within a confidence interval of 185,403 to 246,217. Protein-energy malnutrition accounted for a total of 15,256,524 disability-adjusted life years (DALYs) in 2019, ranging from 12,565,114 to 18,327,803 in the confidence interval. Among these DALYs, 71.5% were attributed to years of life lost.[12] Global challenges like the exhaustion of natural resources, the imperative to sustain a burgeoning human population, and the surge in non-communicable diseases have prompted scientists, businesses, and policymakers to promote a transition from animal-centric to plant-centric diets. Worldwide, approximately 57% of protein intake is sourced from plants, leaving the remaining 43% to animal-derived products. Nevertheless, with the projected global population reaching 9.8 billion by 2050, there is an anticipated 70% surge in the demand for animal-based food.[13]

Serum proteins are divided into two groups: albumin, and globulin. Proteins act as transporters of hormones, vitamins, minerals, lipids, and other materials. In addition, proteins help balance the osmotic pressure of blood tissue.[6] Presuming that standard food for every raised human population is crucial for growth, survival, development, and maintaining good health throughout life. Also, underdeveloped nations are more important as there is global protein undernutrition. In the category of highly nutritious foods, fishes are included. Proteins represent a wide range of roles and supply energy. The necessity of protein varies with many factors such as age, body ability, lactating women, individuals during infections or illness, and tension.[14] To enhance the efficiency of nutrient utilization and overall well-being in livestock, avian, and aquatic species, as well as companion animals, it is imperative to broaden our perspective on individual amino acids beyond their conventional role as protein building blocks. A reconsideration of the ideal protein concept is necessary, taking into account the diverse functions of amino acids. Embracing the latest advancements in amino acids, biochemistry, and nutrition is crucial for optimising animal production globally and requires thinking innovatively outside traditional boundaries.[15]

Goats are primarily raised in a natural environment; they

15

Anuradha & Gupta K V
Epidem. Int. 2023; 8(2)

demonstrate resilience in harsh conditions, requiring minimal medication and typically avoiding the use of feed additives and chemical promoters. Consequently, the meat obtained from goats could be deemed organic when compared to other meat products. Goats can thrive on a low concentrate supply, reducing competition for food grains with others.[16] Fish is a primary and valuable source of animal protein in the diet, contributing a substantial quantity, typically 70% or more, of protein with high biological value. This is especially noteworthy due to its rich content of sulphur-containing amino acids.[17]

Conclusion

Protein is a key source of antibodies and enzymes for all living beings and makes up an essential part of muscles, hair, bones, cartilage, skin, blood, and other bodily components. The protein estimation analysis was conducted to understand the significance of consuming certain food items for healthcare, as protein is vital for a healthy life. A deficiency of protein can cause kwashiorkor disease, with symptoms such as hair loss, nail and skin problems, muscle wasting, oedema, fatty liver, etc. For the biochemical analysis of protein content, various food items were selected and subjected to protein estimation. The results were obtained through centrifugation of collected items. The biochemical parameter, protein, was then estimated using the required chemicals and a spectrophotometer. Then the statistical analysis of concluded data was done using an Excel sheet. The results of this experiment prove that goat bone marrow has the highest protein content than goat liver, fish muscle, black peas and green gram. These samples were chosen due to their availability in the market and increasing consumption. This could be very helpful in addressing protein deficiency and malnutrition in the world, where the population is growing rapidly.

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References

1. Duhan N, Barak S, Mudgi D. Bioactive lipids: chemistry & health benefits. Biointerface Res Appl Chem. 2020;10:6676-87. [Google Scholar]

2. Rosenfeld L. Origins of clinical chemistry. Elsevier Science; 2012. p. 1-17. [Google Scholar]

3. Satpathy L, Dash D, Sahoo P, Anwar TN, Parida SP. Quantification of total protein content in some common edible food sources by lowry protein assay. Platin Open Access J. 2020;9:1275-83. [Google Scholar]

4. Goldenberg JZ, Day A, Brinkworth GD, Sato J, Yamada S, Jönsson T, Beardsley J, Johnson JA, Thabane L, Johnston BC. Efficacy and safety of low and very low carbohydrate diets for type 2 diabetes remission: systematic review and meta-analysis of published and unpublished randomized trial data. BMJ. 2021;372:4743. [PubMed] [Google Scholar]

5. Verma P. Protein deficiency and poisoning. Res Rev J Med. 2023;13(1):9-14. [Google Scholar]

6. Pradhan S, Parida SP, Mohanty S. Protein estimation in seven different sun dried fish species in CUTM campus, Bhubaneswar. Int J Creat Res Thought. 2020;8:402-6.

7. Siddiki AZ, Baten A, Billah M, Alam MA, Shawrob KS, Saha S, Chowdhury M, Rahman AH, Stear M, Miah G, Kumkum M, Islam MS, Hossain MA, Mollah AK, Khan MK. The genome of the Black Bengal goat (Capra hircus). BMC Res Notes. 2019 Jun 27;12(1):362. [PubMed] [Google Scholar]

8. Balami S, Sharma A, Karn R. Significance of nutritional value of fish for human health. Malays J Halal Res. 2019;2(2):32-4. [Google Scholar]

9. Ganesan K, Xu B. A critical review on phytochemical profile and health promoting effects of mung bean (Vigna radiata). Food Sci Hum Wellness. 2018 Mar 1;7(1):11-33. [Google Scholar]

10. Kamani MH, Meera MS. Assessment of black gram milling by-product as a potential source of nutrients. J Food Sci Technol. 2021 Oct;58(10):3844-52. [PubMed] [Google Scholar]

11. Mahesha HB. Estimation of proteins by Lowry's method. Mysore: Yuvaraja's College; 2012.

12. Gastaldello A, Giampieri F, De Giuseppe R, Grosso G, Baroni L, Battino M. The rise of processed meat alternatives: a narrative review of the manufacturing, composition, nutritional profile and health effects of newer sources of protein, and their place in healthier diets. Trends Food Sci Technol. 2022;127:263-71. [Google Scholar]

13. Jiang W, Zhao Y, Wu X, Du Y, Zhou W. Health inequalities of global protein-energy malnutrition from 1990 to 2019 and forecast prevalence for 2044: data from the Global Burden of Disease Study 2019. Public Health. 2023;225:102-9. [PubMed] [Google Scholar]

14. Rath S, Parida SP. Comparative studies of protein estimation in different fresh water fishes. Int J Sci Res. 2020;2:2319-7064.

15. Wu G, Bazer FW, Dai Z, Li D, Wang J, Wu Z. Amino acid nutrition in animals: protein synthesis and beyond. Annu Rev Anim Biosci. 2014 Feb:2:387-417. [PubMed] [Google Scholar]

16. Lalhriatpuii M, Singh AK. Goat meat: no less source of protein in comparison to other meat for human consumption. In: Kukovics S, editor. Goat science-environment, health and economy. IntechOpen; 2021 May 13. [Google Scholar]

17. Pawar SM, Sonawane SR. Fish muscle protein highest source of energy. Int J Biodivers Conserv. 2013 Jul;5(7):433-5. [Google Scholar]