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QUALITATVE ANALYSIS OF PROTEIN: EGG ALBUMIN AND MILK

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*For Correspondence:	ABSTRACT			
Department of Pharmacy, Prasad	As the name suggests amino acids are organic compounds that contain			
Institute of Technology, Jaunpur	amino and carboxyl groups. The above formula stands for different			
U.P 222001	chemical groups and this determines the characteristics of the amino			
Received: 29.09.2018	acids. The color tests have frequently been used for qualitative			
Accepted: 22.03.2019	detection of amino acids. Not all amino acids contain the same reactive			
Access this article online	groups. For this reason, the various color tests yield reactions varying in intensity and type of color according to the nature of groups contained			
Website:	in the particular amino acid under examination. Ninhydrin test, Biuret			
www.drugresearch.in	test, Xanthoproteic acid test and Millon's test were present in egg			
Quick Response Code:	albumin and milk. This is essential substances in human body for the maintenances of protein. KEY WORDS: Egg Albumin and Milk, Ninhydrin test, Biuret test, Xanthoproteic acid test and Millon's test.			

INTRODUCTION

PROTEIN: The word "protein" is derived from the Greek word "proteios", which means "of primary importance". In fact, proteins play an important role in all biochemical and physiological body processes; they act as enzymes, hormones, receptors, antibodies and are required for the structural integrity of cells. Proteins are the most essential for the maintenance of human life. Tissue, muscles, organs, enzymes and hormones are protein in nature. The body can't store excess protein. Protein requirement is expressed in terms of grams/ kg body weight. The daily body requirement of protein for an adult is about 1g/kg body weight. It is desirable that one fifth of it should be animal protein. Protein deficiency amounts to amino acids deficient and is accompanied by deficiency of mineral and vitamins. Protein calorie malnutrition (PCM) means deficiency of calories due to deficiency of protein, it is also known as protein Energy Malnutrition (PEM).^[1] A protein is said to be all the essential amino acids are present in the required proportion by the human body that is egg albumin.

Essential and non-essential amino acids: There are 20 standard amino acids which differ in their side chain. Some of them are considered "**essential**" since they cannot be synthesized in our body and must be therefore provided in the diet (e.g. tryptophan & phenylalanine), while others are "**non-essential**" and can be synthesized in the body (e.g. alanine & cysteine). A good source of protein is often a combination of various foods, because different foods are rich in different amino acids. A

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good source of dietary protein meets two requirements:^[2] The requirement for the nutritionally indispensable amino acids (histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine) under all conditions and for conditionally indispensable amino acids (cystine, tyrosine, taurine, glycine, arginine, glutamine, proline) under specific physiological and pathological conditions

• The requirement for nonspecific nitrogen for the synthesis of the nutritionally dispensable amino acids (aspartic acid, asparagine, glutamic acid, alanine, serine) and other physiologically important nitrogen-containing compounds such as nucleic acids, creatine, and porphyrins.

Protein deficiency and malnutrition (PEM) can lead to variety of ailments including mental retardation and kwashiorkor. Symptoms of kwashiorkor include apathy, diarrhea, inactivity, failure to grow, flaky skin, fatty liver and edema of the belly and legs. This edema is explained by the action of lipoxygenase on arachidonic acid to form leukotrienes and the normal functioning of proteins in fluid balance and lipoprotein transport. ^[3,4] PEM is fairly common worldwide in both children and adults and accounts for 6 million deaths annually. In the industrialized world, PEM is predominantly seen in hospitals, is associated with disease, or is often found in the elderly.^[5] Albumin and globulin are two types of protein in your body. The total protein test measures the total amount albumin and globulin in your body. It's used as part of your routine health checkup. It may also be used if you have unexpected weight loss, fatigue or the symptoms of a kidney or liver disease.

Purpose of the total protein test

A total protein test is completed as part of your routine health checkup. It's one of the tests that make up your comprehensive medical panel (CMP). It may be ordered if you have:

- unexplained weight loss
- fatigue
- edema, which is swelling caused by extra fluid in your tissues
- symptoms of kidney or liver disease

The total protein test measures the total amount of protein in your blood and specifically looks for the amount of albumin and globulin. This test will also look at the ratio of albumin to globulin in your blood. This is known as the "A/G ratio."

MATHODS

Using the provided solutions of albumin (egg white), casein (milk protein) and gelatin (animal collagenous material), perform the following ^[6,7,8].

A. General tests

- B. Color reactions
- C. Precipitation reactions

A. GENERAL TESTS FOR PROTEINS

- 1. Ninhydrin test: Ninhydrin is most commonly used as a forensic chemical to detect "fingerprints", as amines left over from proteins sloughed off in fingerprints react with ninhydrin giving a characteristic purple color.
- Procedure & observation:

To 1 ml amino acid solution in a test tube, add 1 drop of ninhydrin. Put in a boiling water bath and observe the formation of a purple color.

- 2. Biuret test: The biuret reagent (copper sulfate in a strong base) reacts with peptide bonds in proteins to form a blue to violet complex known as the "biuret complex".
- Procedure & observation:

To 2 ml of protein solution in a test tube, add 3 drops of 10% sodium hydroxide solution and 3-6 drops of 0.5% copper sulfate solution. Mix well; a blue to violet color is obtained with albumin, casein & gelatin.

B. COLOR REACTIONS OF PROTEINS

1. Xanthoproteic acid test: Nitric acid gives a color when heated with proteins containing tyrosine (yellow color) or tryptophan (orange color); the color is due to nitration.

• Procedure & observation:

To 2 ml of protein solution in a test tube, add 2 drops of concentrated nitric acid. A white precipitate is formed and upon heating in a boiling water bath, it turns yellow with "tyrosine" and orange with the essential amino acid "tryptophan" indicating a high nutritive value.

2. Millon's test: Millon's reagent (Hg/HNO3) gives positive results with proteins containing the phenolic amino acid "tyrosine".

• Procedure & observation:

Took 2 ml of protein solution in a test tube, add 3 drops of Millon's reagent. Mix well and heat directly on a small flame. A white ppt is formed with albumin and casein (but not gelatin); the ppt gradually turns into brick red.

S.No.	Tests	Egg Albumin	Milk
1.	Ninhydrin test: Test solution and added 2drops Ninhydrin reagent and heat on water bath- Purple colour	Present	Present
2.	Biuret test: Test sample and added Biuret Reagent- Purple or pink colour	Present	Present
3.	Xanthoproteic acid test: Test solution and added 2 drops of nitric acid then heat on water bath- Yellow colour	Present	Present
4.	Millon's test: test sample +2ml millon's reagent and boil for 10min on water bath + 5drops NaNO ₂₋ Brick red colour	Present	Present

RESULTS

Ninhydrin test, Biuret test, Xanthoproteic acid test and Millon's test were present in egg albumin and milk. This is essential substances in human body for the maintenances of protein.

CONCLUSION

The normal range for total protein is between 6 and 8.3 grams per deciliter (g/dL). This range may vary slightly among laboratories. Low albumin is considered albumin below 3.4 g/dL. It's associated with decreased effectiveness of medications used for ulcerative colitis. Low albumin levels may result in complications during or after surgery.

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