Lecture 1/Second Year

Serum Albumin

Subject Lecturer

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Estimation of total protein and Albumin/Globulin Ratio

Total Protein

CLINICAL SIGNIFICANCE

Proteins form the major portion of dissolved substances in the plasma. They form the basic structural components of the body. They constitute the enzymes present in our body & also act as secondary source of energy. The other functions include distribution of water, buffering, transport of various components, defense & coagulation of blood in our body. Increased levels are found in dehydration & myeloma. Decreased levels are found in liver disorders, Nephrotic syndrome, malnutrition & protein due to haemorrhage.

PRINCIPLE

Protein in serum or plasma forms a blue/violet complex when mixed with copper ions in alkaline solution (Biuret reaction) each copper ion binding with 5 or 6 peptide bonds. Tartrate is added as a stabilizer and iodide is used to prevent auto reduction of the alkaline copper complex

The absorbance of this complex at 546 nm is proportional to the protein concentration

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REAGENT COMPOSITION

TOTAL PROTEIN (SL)

Sodium hydroxide	200 mmol/L
Potassium sodium tartrate	32 mmol/L
Copper Sulphate	18 mmol/L
Potassium Iodide	30 mmol/L

Protein Standard

Protein standard concentration	8 g/dL

ASSAY

Wavelength	546nm
Cuvette	1 cm light path
Temperature	20-25°C
Measurement	Against reagent blank

PROCEDURE

Pipette into cuvettes	Blank	Standard	Sample
T. Protein reagent	1000 µL	1000 µL	1000 µL
Standard	--	20 µL	--
Sample	--	--	20 µL
Mix and incubate for 5 minutes at 20-25°C (RT). Measure the absorbance of the sample (As) and the standard (Astd) against the reagent blank within 30 minutes.			

CALCULATION

$$\text{Total Protein Conc. (g/dL)} = \frac{\Delta A \text{ sample}}{\Delta A \text{ standard}} \times 8 \text{ (Std.conc.)}$$

Albumin/Globulin Ratio

The albumin globulin ratio expresses the balance between albumin and globulins in serum. Albumin acts as the main carrier protein and helps maintain oncotic pressure. Globulins include antibodies and transport proteins that support immunity and other functions.

Clinicians use the ratio as a quick screen to detect shifts in protein production or loss. The result does not diagnose a specific disease alone. Instead, clinicians treat it as a clue that guides further testing.

The albumin–globulin (A/G) ratio is a calculated parameter used to evaluate the relative proportions of albumin and globulins in serum and provides insight into liver function, nutritional status, and immune activity.

It is calculated as:

A/G ratio = Serum albumin ÷ Serum globulin,

where globulin is obtained by subtracting albumin from total protein.

The normal A/G ratio is approximately 1.0–2.0, as albumin normally exceeds globulins in concentration.

A decreased A/G ratio may result from reduced albumin synthesis (e.g., liver disease), increased globulin production (e.g., chronic infections, autoimmune disorders, multiple myeloma), or protein loss.

An increased A/G ratio is less common and may be seen in conditions associated with low globulin levels, such as immunodeficiency states or leukemia.