

Serum total protein

Proteins are biologically important compounds composed of α -amino acids. Proteins are major constituents of cells and hence of living bodies.

Plasma proteins is a collective term for the proteins present in the blood. Plasma contains > 300 different proteins. Plasma proteins are synthesized mainly by the liver with the exception of γ – **globulins which** are synthesized by plasma cells present in lymph nodes, spleen and bone marrow. Many diseases can affect the total concentration of proteins as well as affecting the ratios of protein fractions that comprise the total concentration.

Plasma proteins are heterogenous in nature and so are involved in many functions in the body.

Functions of plasma proteins

- Transport (Albumin, prealbumin, globulins)
- Maintain plasma oncotic pressure (Albumin)
- Defense (Immunoglobulins and complement)
- Clotting and fibrinolysis (Thrombin and plasmin)

Normal range of serum proteins:

Total protein:	64 - 83 g/l	or	6.4 - 8.3 g/dl
Albumin:	35 - 52 g/l	or	3.5 - 5.2 g/dl
Globulin:	20 - 35 g / l	or	2.0 - 3.5 g/dl

Serum total protein level gives an approximate measure of all serum proteins (with the exception of fibrinogen as the analysis is on a clotted blood sample).

Many diseases can affect the total concentration of proteins as well as affecting the ratios of protein fractions that comprise the total concentration. Abnormal serum protein levels may be primary (the cause is inherited) or secondary (the result of a wide range of diseases).

Pre-analytic causes of changed serum total protein level:

- The total serum protein, will be approximately 0.25 g/dl lower than total plasma proteins due to the fact that fibrinogen protein is found in plasma only and not in serum.
- In case of serum protein test, a prolonged application of a tourniquet may increase protein level due to hemoconcentration.
- The taking of blood from the arm into which an intra-venous infusion is flowing can cause dilutional hypoalbuminaemia.
- Plasma albumin concentrations may be 5-10 g/l lower in the recumbent than in the upright position because of the redistribution of body fluids.

Hyperproteinemia:

It is a state of is **an increase in the concentration of protein in the bloodstream.**

Causes of hyperproteinemia:

- 1- Increased synthesis such as in infections like viral hepatitis B or C, or HIV.
- 2- Increased synthesis in some cancers such as multiple myeloma or certain types of lymphoma.
- 3- Dehydration that results from conditions like vomiting, diarrhea, excessive sweating, and diabetic acidosis.

Hypoproteinemia:

It is a state of is **a decrease in the concentration of protein in the bloodstream.**

Causes of hypoproteinemia:

- 1- Prolonged starvation.
- 2- Malnutrition and malabsorption such as in Celiac disease.
- 3- Decreased synthesis by the liver such as in liver cirrhosis or failure.
- 4- Increased loss of proteins as in kidney disease (e.g., nephrotic syndrome) or intestinal disease (protein-losing enteropathy).
5. excessive bleeding.
6. Extensive burns.
7. Catabolic states after injury or tissue damage.
8. Inflammatory conditions that are associated with hypoalbuminemia

Serum protein electrophoresis

Serum protein electrophoresis (SPEP) is a simple method of separating proteins based on their net charge, size, and shape. The 2 major types of protein present in the serum are albumin and globulin proteins. Albumin is the major protein component of serum and represents a high peak that lies closest to the positive electrode. Globulins make up a much smaller fraction of the total serum protein but represent the primary focus of interpretation of serum protein electrophoresis. Five globulin categories are represented: alpha-1, alpha-2, beta-1, beta-2, and gamma, with the gamma fraction being closest to the negative electrode.

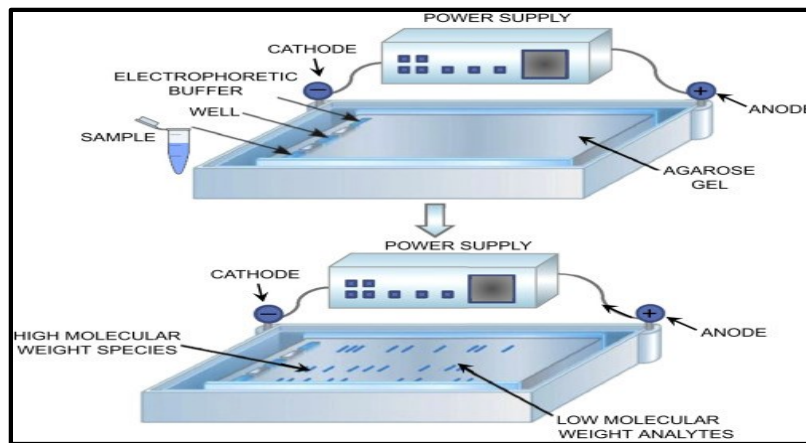


Figure 1: Electrophoresis instrument

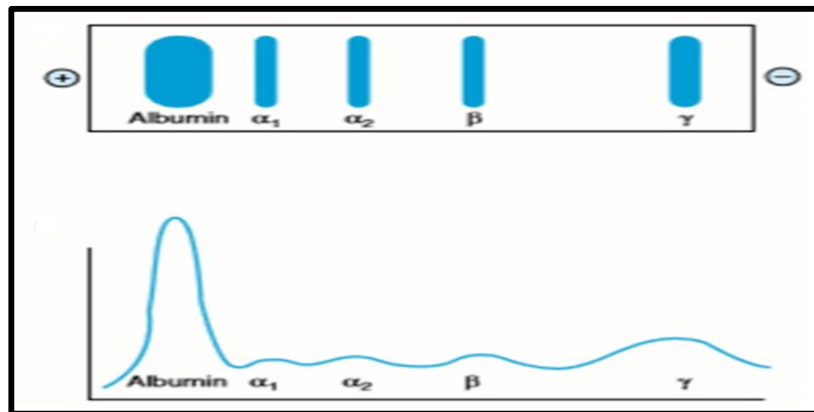


Figure 2: Electrophoresis pattern or diagram

Examples of abnormalities in electrophoresis of serum protein fractions

Protein in electrophoretic pattern	Detection of disease
Albumin	<ul style="list-style-type: none"> - Malnutrition - Malignancy - Liver disease - Kidney disease
α1-globulin	<ul style="list-style-type: none"> - Neural tube defects (α-FP ↑) - Acute & Chronic inflamm dis ↑ - α1-antitrypsin deficiency ↓
α 2- globulin	<ul style="list-style-type: none"> - Wilson's disease (↓ ceruloplasmin) - Hemolytic disorders ↓ - Nephrotic syndrome ↑
β 1- globulin (↑)	<ul style="list-style-type: none"> - Infection ↑ - Inflammation ↑ - Some hyperlipidemias (↑ apo B)
β 2- globulin	<ul style="list-style-type: none"> - Renal failure ↑ in glomerular and ↓ in tubular disease - iron deficiency/excess (↑ transferrin)
γ – globulin ↑	<ul style="list-style-type: none"> - Chronic Liver disease - Acute Infections - Autoimmune disease - Chronic inflammatory conditions like RA - Multiple myeloma

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Principle of measurement serum total protein (biuret method):

A protein (in serum) containing two or more peptide bonds that react with Cu^{+2} ions in alkaline solution (Biuret's reagent) to form blue - violet colored complex , the intensity of its color increase with the increase protein concentration in serum .

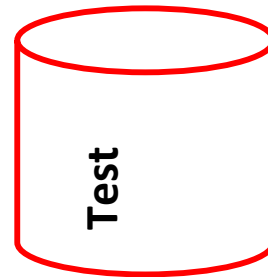
Peptide bond (proteins) + Cu^{+2}  blue violet colored complex

Procedure :

2 ml Biuret reagent
(working reagent)

+

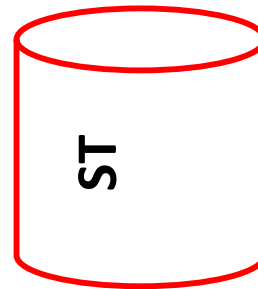
20 μl Serum



2 ml Biuret reagent
(working reagent)

+

20 μl Standard



Mix and stand for 10 min in 37 °C, then read at 550 nm

Concentration of standard 8 g/ dl

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