serum protein and A/G ratio

Blood plasma contains at least 125 individual proteins. Serum (as contrasted with plasma) is deficient in those coagulation protein which are consumed during the process of blood coagulation. The value of total serum protein will be approximately 0.25 gm/100 ml lower than for plasma protein because of the absence of fibrinogen (which was converted to insoluble fibrin which make up the blood clot). Only a few of the plasma proteins have been isolated, characterized and their function determined.


Serum protein include albumin and Globulin but most of the Fibrinogen is removed in the clothing processes.

Albumin, the most prevalent protein (about 60% by weight), has a low molecular weight (66.000) and has an important function in the regulation of the osmotic pressure of the plasma, and in distribution of water between blood plasma and the tissues. Albumin is also important as nonspecific transport mechanism for many physiologic substances as well as drugs, antibiotics, various ions, amino acids and hormones. Albumin also serves as a precursor for tissue proteins and in nutrition, proteins play a small part in maintaining the plasma pH. They are negatively charged at body pH and so act as bases, accepting hydrogen ions. The wide range of enzymes present in plasma are proteins. The immunoglobulins are antibodies which provided a defense against infection.

Many plasma proteins, including albumin, fibrinogen and most
globulins are formed in the liver; immunoglobulins are produced by the reticuloendothelial system, the lymph nodules and plasma cells; the enzymes are produced or released from various organs.

The oldest procedure used for protein estimation is the kjeldahl procedure which is a method used for determining the total nitrogen content in biological materials. The nitrogen containing compound in serum are converted to NH$_4^+$ by oxidation in a digestion mixture of cone. H$_2$SO$_4$, catalyst and a salt to increase the boiling point of the mixture. The NH$_4^+$ may also be quantitated photometrically by Nessler's reagent. The protein containing 16% nitrogen by weight. The total nitrogen content in (gm/l) of a sample minus the nonprotein nitrogen is multiplied by 6.25 to obtain the protein content in gm/l.

The most frequently used colorimetric method for determining total protein in serum is the biuret method. In this reaction cupric ion reacts with the peptide linkages of protein in a basic solution to form blue violet colored complex with an absorption maximum at 540 nm.

One of the simple and important techniques for quantitaion of serum proteins is their separation in an electric field, this procedure is referred to as serum protein electrophoresis (SPE). An electric field is applied to a medium containing charged particles, at pH 8.6 all the proteins are negatively charge and tend to move toward the anode, while the positively charge particles migrate towards the cathode. At a higher pH there is increasing danger of denaturation of the protein. Barbitone buffer of ionic strength between 50-140 mmol/l have been mostly used. At lower ionic strength the buffering capacity is too low, at higher ones too much heat is developed so that the possibility of denaturation increases. Demarcation of the bands sharpens with increase ionic strength.
Separation can be followed by permanent fixation of the fraction at the position in the medium to which they migrate and by using certain pH value and certain voltage at the fixed time.

The rate of movement is manly dependent on the charge on the protein molecule (proteins at a certain pH value differ in their net charge) the greater the charge the more rapidly it moves, but it is also influenced by the mass and shape of the molecule, increase in mass exerting a retarding effect.

A variety of support media has been used in recent years for protein separations, among which are cellulose acetate, agar gel, agarose, starch gel and acrylamide gel. The major differences is that because in the last two the pore size is smaller and is comparable with the size of the protein molecules there is superimposed a molecular sieve effect. This enables molecules of the same charge but different size to be separated and has led the use of double runs.

Cellulose- acetate electrophoresis is most widely used for human serum proteins separation by using barbital buffers at pH 8.6 for 30 min and the applied voltage is 90-100 volts. Six zones are recognized for normal sera at pH 8.6 which are represent: albumin, $\alpha_1$, $\alpha_2$, $\beta_1$, $\beta_2$, and $\gamma$ (gamma)- globulin- for plasma protein electrophoresis a seventh zone was obtained for fibrinogen which is present in plasma.
Clinical significance:

The total plasma protein concentration ranged between 63-79 gm/liter, albumin may be taken as 37-53 gm/liter and globulins 18-36 gm/liter with fibrinogen 2-4 gm/liter, giving an albumin /globulin ratio which can vary from 1.2-2.5.

An increase of total proteins may be caused by dehydration and a decrease from overloading with water. The increase in disease arise from an increase in total globulin most often the gamma- globulin, the albumin remaining normal or being reduced to a lesser extent. A decrease in total protein concentration is almost always the result of a fall in that of albumin while the globulin do not change or increase by a smaller amount. Although the changes in albumin and globulin concentration may cancel each other, the albumin- globulin ratio may range from below 0.5 to well over 2.5.

Albumin was increased in dehydration due to reduced of plasma water, chronic vomiting and diarrhea may impair intestinal absorption of amino acids so reducing plasma albumin formation. A decrease in plasma albumin concentration occurs with excessive losses (a) in the urine as in the nephritic syndrome (b) into the intestine as in protein- losing naturopathy (c) from widespread burns (d ) in severe hemorrhage . Also decreased in defective anabolism may be due to( a ) reduce synthesis in liver diseases( b ) defective digestion or malabsorption( c) impaired intake of protein in malnutrition.
Increased globulin concentration usually occurs in the y- globulin fraction (or immunglobulins) and there is a corresponding increase in the total protein concentration, the most common causes of hypergammaglobulinemia are chronic infections, liver diseases, and lymphoma.

Decreased level of γ - globulin was observed in association with a tumor of thymus gland which is a very rare tumor, and in chronic lymphocytic leukemia.