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Methods of Determining the Total Amount of Plasma Proteins in Liver Diseases, Refractometer, Keld Reaction with Biuret, Nephelometer Method

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ABSTRACT

This article describes in detail the methods of determining the total amount of proteins in plasma in liver diseases, and provides theoretical information about the Refractometer, Keld's biuret reaction, and the Nephelometer method.

The liver performs more than ten vital functions, it can be compared to a huge factory. Its employees - hepatocytes - form unique communities. There are many such functional units in the liver, and they are able to take over the work of their dead counterparts. Therefore, if this gland becomes infected, symptoms of pathology may not appear immediately. Changes in the concentration of individual plasma proteins are observed in many physiological and pathological conditions. Changes in the ratio of different protein fractions (dysproteinemia), the appearance of unusual proteins, or the absence of any serum protein (paraproteinemia) are of great diagnostic value. Numerous proteins contained in blood plasma, differing in structure, physicochemical properties and functions, are designated as total protein and represent the sum of all plasma proteins. The concentration of total protein in plasma is determined by three factors:

a) the rate of synthesis;

b) removal rate;

c) volume of distribution.

Blood plasma proteins are classified according to their function, and the same protein can perform several different functions. Thus, serum albumin maintains its oncotic pressure, providing the volume of circulating blood and microcirculation of tissues, participates in the transport of various substances by the blood, and is an acute phase protein. The concentration of total blood protein can change significantly within a short time; when moving from a horizontal to a vertical position of the body, after 20 minutes the concentration of total protein can increase by 10-20% of the initial value. A false increase in the level of total protein in plasma is observed during venipuncture with a tourniquet applied to the shoulder and, especially, massaging or the so-called. "hand work" when taking blood from a vein. In these cases, the concentration of total protein increases after 2-3 minutes. This is due to the fact that even a short venostasis leads to an increase in intravenous hydral pressure and to an increase in the permeability of water by the venous wall, causing blood clotting. Changes in the concentration of total blood protein are due to changes in the concentration of either albumin or immunoglobulins. Other fractions of blood proteins, when their concentrations change, do not have a noticeable effect on the concentration of total protein. A rapid increase in the concentration of total blood protein (hyperproteinemia) is always associated with a decrease in the volume of extracellular fluid (dehydration), and a rapid decrease (hypoproteinemia) is associated with hyperhydration (excess water retention). According to the mechanism of development, hypo- and hyperproteinemia can be absolute and relative. Many liver diseases are insidious at a treatable stage. Symptoms begin to be felt when the liver is already significantly damaged and serious disorders occur. In severe cases, liver disease can lead to the death of the patient. Among the common and dangerous pathologies of this organ, cirrhosis takes one of the leading places. But cirrhosis rarely develops "by itself". An exception is primary biliary cirrhosis, in which liver cells are damaged by the immune system. In most cases, it occurs as a result of untreated chronic problems.

Absolute hyperproteinemia develops with an increase in any protein, mainly immunoglobulins. It is never associated with increased albumin biosynthesis, therefore, hyperalbuminemia indicates dehydration or technical errors in the analysis (violation of the procedure for taking blood - venous stasis with prolonged squeezing of the veins with a tourniquet, squeezing and unclenching the hand at the time of taking blood. Hyperimmunoglobulinemia as a cause of absolute hyperproteinemia is observed in chronic liver diseases (chronic and subacute hepatitis), autoimmune diseases (rheumatoid arthritis, dermatomyositis). Relative hyperproteinemia is a consequence of dehydration. Increased water loss with repeated vomiting, diarrhea, profuse sweating, shortness of breath causes blood clots, increased hematocrit and an increase in total protein. This equally increases the concentration of hemoglobin, the number of red blood cells; with dehydration and increased blood viscosity, ESR usually decreases. Absolute hypoproteinemia is usually due to a decrease in albumin concentration, either as a result of increased loss of albumin in the urine, through the intestinal mucosa, damaged skin, or as a result of a decrease in albumin biosynthesis by the liver in its pathology (cirrhosis).

Physical exertion (fast walking, climbing uphill, heavy lifting), nervous breakdown, eating large amounts of food, cold, wet and windy weather, strong excitement (including TV shows under the influence), sex, stopping taking drugs (antianginal, hypotensive, antiarrhythmic) leads to the development of angina attacks. Angina attacks are sometimes manifested in atypical forms, pain can be transmitted to the jaw, fingers, epigastric region, under the right and left rib cage. In some cases, the pain is of secondary importance, the patient feels shortness of breath, tightness of the chest, severe weakness, fear of death. Violation of bilirubin absorption by the liver: as mentioned above, the process of bilirubin absorption by the liver cells is accompanied by the separation of this pigment from albumin. this condition can be observed in patients. When the integrity of the liver cells is broken, the excretory activity of the liver is impaired to a greater extent when bilirubin binds to glucuronide. Therefore, in most hepatocellular diseases, hyperbilirubinemia is

mainly associated with free bilirubin.

The nephelometry It consists in measuring the radiation caused by particles (in solution or suspension), thereby measuring the power of the scattered radiation at a different angle from the direction of the incident radiation. When a suspended particle hits a beam of light, some of the light is reflected, some is absorbed, some is deflected, and the rest is transmitted. Therefore, when light falls on a transparent medium containing a suspension of solid particles, the suspension appears cloudy. A nephelometer is an instrument used to measure suspended particles in a liquid or gas sample. Thus, a photocell placed at an angle of 90 ° to the light source detects the radiation from the particles present in the suspension. Also, the light reflected by the particles towards the photocell depends on the density of the particles. In nephelometry, it is very important to have a source of radiation that emits high light. There are many different types, from xenon lamps and mercury vapor lamps, to tungsten halogen lamps, laser beams and more. In cuvettes, any agent external to the studied solution, inside or outside the cuvette, reduces the light beam on the way to the detector (defective cuvettes, dust sticking to the walls of the cuvette). Noise: the presence of some microbial contaminant or turbidity scatters the radiant energy, increasing the intensity of the dispersion. Fluorescent compounds: These are compounds that, when excited by incident radiation, produce false and high scattering density readings.

Since the radiation intensity of the detected radiation is proportional to the mass concentration of the particles, nephelometric studies have theoretically higher metrological sensitivity than other similar methods (for example, turbidimetry). In addition, this technique requires dilute solutions. This allows to minimize absorption and reflection phenomena. In this method, the rate of complex formation is constantly monitored. The reaction rate depends on the antigen concentration in the sample. Here, measurements are taken as a function of time, so the first measurement is taken at time "zero" (t = 0). Kinetic nephelometry is the most widely used method, because the study can be performed in 1 hour compared to the endpoint method. The dispersion coefficient is measured only after the addition of the reactant. Nephelometry is commonly used to analyze the chemical quality of water, determine clarity, and control treatment processes. In addition, the concentration of harmful substances is used to measure air pollution, which is determined by the dispersion formed in the light they fall on.

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