

# METHODS OF QUANTITATIVE ANALYSIS IN CLINICAL LABORATORY DIAGNOSTICS

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When conducting biochemical analyses in clinical diagnostic laboratories use methods of quantitative determination of components in biological fluids on the basis of modern advances in medical science and technology, providing high quality research, mechanisation and automation of laboratory work.

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These methods include the following. 1. Weight (gravimetric) analysis, based on the isolation of a substance as a result of certain reactions, drying and accurate weighing of it on analytical or torsion scales. An example of this analysis is the determination of fibrinogen content by the Rutberg method. 2. Volumetric (titrometric) analysis, based on accurate measurement of volumes of substances reacting with each other in equivalent (equal) quantities. Volumetric analysis includes neutralisation method, method of redox reactions, complexometry, precipitation method, etc. Examples are determination of acidity of gastric juice, chlorides in biological fluids by titrometric method, etc.

3. electro-volumetric (electroanalytical) methods based on electrochemical properties of solutions. This group includes conductometry, potentiometry,



potentiometry, voltammetry, polarography, etc. An example of these methods is the determination of the concentration of hydrogen, chlorine, sodium, potassium, calcium ions in biological fluids using ion-selective electrodes.

4. optical methods including refractometry, polarimetry and photometry. The most common methods in clinical diagnostic laboratories are photometric methods, which are divided into absorption and emission methods. Absorption photometry includes spectrophotometry, nephelometry and turbidimetry. Emission photometry includes fluorimetry and flame photometry.

Absorption photometric analysis is based on the physicochemical property of a substance to selectively absorb a monochromatic (of a certain wavelength) flux of light energy. Instruments designed for absorption photometry are called optical analysers or photometers (absorption photometers).

**Photometers** include colorimeters, photoelectrocolorimeters, spectrophotometers. Colorimeters (colour - colour, metric - measure) are devices designed to measure the wavelength of the visible region of the spectrum (400-800 nm). The principle of colorimetry is based on measuring the colour intensity of the solution of the analysed substance. Analysers that allow operation in both visible and invisible spectral regions are called spectrophotometers. These instruments allow measurement in the ultraviolet (190-400 nm), visible (400-800 nm) and infrared (800-2000 nm) regions of the spectrum. Nephelometry is based on the measurement of the intensity of light scattering by suspended particles of the substance under investigation. The more turbid a solution is, the more it scatters light and, consequently, the less it transmits. Turbidimetry is the measurement of the absorbed light flux by the particles of the substance under study. The more turbid the solution, the more it absorbs light and transmits less. Emission photometry is based on the ability of organic substances to give characteristic emission spectra (emission, luminescence) in an energetically excited state. Atoms and molecules of substances are able to absorb energy coming to them from the outside and move to higher energy levels, and then, returning to the normal energy state, give up excess energy in the form of a quantum of light. Fluorimetry is based on the effect of fluorescence (luminescence) resulting from



energetic excitation of the substance under study after irradiation with ultraviolet or other short-wave rays. There are several types of luminescence analysis: luminescence microscopy, luminescence chromatography, fluorimetric quantitative analysis. The instruments used for fluorimetric quantitative analysis are called fluorimeters. They are used to determine the concentration of vitamins, adrenaline, noradrenaline, serotonin and other biologically active substances. In flame photometry, a gas burner flame is used as an energy source causing the excitation state of the sample under study. Metal atoms, falling into a high-temperature flame, capture part of the thermal energy and then release it in the form of a quantum of light. Instruments designed for this type of research are called flame photometers. They are used to determine the concentrations of potassium, sodium, lithium ions, etc. Currently, flame photometry is being replaced by new instruments for the determination of electrolytes - ion-selective analysers. Atomic absorption spectrophotometry possesses high accuracy reproducibility of measurement results, with the help of which it is possible to determine a wide range of elements not only in biological fluids, but also in various environmental objects.

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