MINISTRY OF HEALTH OF THE REPUBLIC OF MOLDOVA
STATE MEDICAL AND PHARMACEUTICAL UNIVERSITY
NICOLAE TESTEMITANU

Dumitru CROITORU, Victor VOVC
Ion COJOCARU

Practical Papers of Medical Biophysics

Second edition

CHISINAU
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PREFACE

Biophysics is a science of interdisciplinary character being at the border, which parts and links at the same range physics, mathematics and biology. It has special relations with some sciences of interdisciplinary character such as biochemistry (that studies chemical aspects of structures and biological phenomena), biomathematics (mathematical aspects of structures and biological phenomena), bioengineering (application of principles and engineering processes in biomedical issues), bionics (application in techniques of nature inventions), chemical physics (physical aspects of chemical phenomena). It has the close relations with physiology, molecular and cell biology, cybernetics, electronics, etc.

Biophysics taught at medical department studies three general subjects:

1) physical analysis of primary mechanisms referring to composition, structure, existence and development of living matter (physical aspects of liquids and biological tissues, biological membranes, the transmembrane transportation, biopotentials and transportation of nervous influx, cell energy, physics of complex elements, etc.)

2) familiarization of future specialists with physical techniques applied in the biomedical exploitation (luminescent analysis, spectral analysis, diffraction methods by X - rays, electrophoresis methods, polarimetry, electronic microscopy, spectrometers of magnetic resonance), and also the constructive principles and functioning of modern equipment with diagnostic and therapeutic purpose (of electrography, ultrasound, quantum generators, detectors and dosimeters of ionizing radiation, etc.);

3) analysis of biological effects of physical factors from the environment, issues assumed by biophysics within the fundamental level of interaction of these factors with biological systems.

Medical biophysics offers the student the possibility to understand the mode, in which ions, molecules and molecular con-

glomeration participate in different biological processes, in which the energy is transformed from one form into another.

The study process in laboratories is organized so that to form in students scientific skills for research which includes three main components: measurement of studied phenomenon parameters, mathematical analysis of experimentally obtained results, establishment of functional dependence between the tested factor and response reaction. The content of all biophysics practical paper corresponds to this criterion. The studied subject in laboratory is of biological nature (liquids, cells, tissues, and organs) within optimal possible limits.

Each paper includes:

1) theoretical notions which are at the base of the method;
2) explanations regarding the construction and functioning of applied devices and equipment;
3) information about the importance of the applied method for research and medical practice;
4) working technique;
5) representation form of experimentally obtained results.

An advantageous didactical peculiarity with respect to the handbooks of the same kind existing presently is the alternation of practical papers with qualitative – illustrative experiments well integrated into the content of the paper. The positive emotional effects produced by these expiments stimulate students'desire to familiarize more profoundly with the essence of physical phenomena implied in harmonious modality and economically into vital processes.

The devices and applied installations with this purpose were performed at the Biophysics and Informatics Department of the State Medical University "N. Testemitanu" of the Republic of Moldova within many years. Having an original construction they are protected by invention patents.

To perform practical papers in biophysics always means to make a people – device interface, it also means contact with the tool, to be maneuvered, regulated, used in optimal way.

Nowadays collaboration of medicine with physics and engineering is more necessary, the language barrier between the physiologist and physicist, between the physician and engineer endangers the capacity to understand each other. With this aspect the biophysical practical paper offers future doctors elements both of language and thinking which allow discussing constructively with physicists and engineers.

Offering the students this handbook we desire not only to give necessary knowledge but also to contribute within possibilities to obtain fundamental science gifts, to make the interest in the application work of physical performances and modern technique in medicine.

We acknowledge to all who will give any suggestions and critical objections for improving a future edition.

Authors

1. DETERMINATION OF VISCOSITY OF BIOLOGICAL LIQUIDS

Purposes

- presentation of theoretical notions of liquid viscosity;
- study of the relative method of viscosity determination by the Ostwald viscometer;
- determination of plasma viscosity;
- confirmation of the Stokes law by demonstrative experiment

Theoretical notions

The cohesion forces between molecules of fluid are relatively small, therefore different layers of liquid can slip easily one on another. An ideal liquid will be that which is not compressed and the removing of the molecules of which takes place without any friction. The apparition of friction force for the real liquids follows the removing of the molecules.

The forces appearing during the moving of liquid layers are called the internal friction forces or viscosity.

We shall consider a surface S_1 into a mass of liquid in moving, parallel to the direction of flowing that removes with respect to another surface of the same value S_2 being removed at the distance Δx (Fig. 1.1)

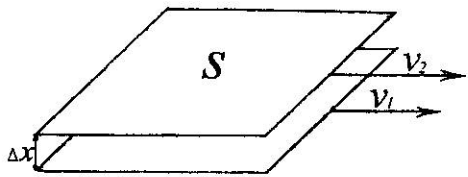


Fig. 1.1

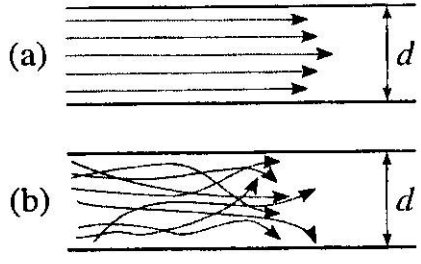
The surface S_1 exercises the friction force **F** on the surface S_2 with the value calculated by the law of Newton:

$$F = \eta \cdot S \cdot \frac{\Delta V}{\Delta d} \qquad (1)$$

Where

$$\frac{\Delta V}{\Delta d} = \frac{V_1 - V_2}{\Delta d}$$

is the gradient of speed, η – the coefficient of dynamical viscosity of the liquid.



Schema reprezentation of the flows:
a – Laminar; *b* – turbulent;

The definition equation of dynamical viscosity coefficient is established from the equation (1):

$$\eta = F \frac{1}{S \cdot \frac{\Delta V}{\Delta d}} \qquad (2)$$

As $S = 1$ and $\frac{\Delta V}{\Delta d} = 1$ we obtain $\eta = F$, and it follows from this

that the dynamical viscosity coefficient is equal numerically to the friction force from the interior of the liquid, which is exercised by a monomolecular layer on another monomolecular layer (both are equal as the surface to the unit and parallel to the flowing direction), when the gradient of the relative speed is a unit.

The dimensional equation of the dynamical viscosity coefficient is:

$$\dim \eta = ML^{-1}T^{1}$$

The measure unit is obtained from the definition equation (2) in I.S:

$$[\eta]_{I.S} = N\frac{1}{m^2\frac{m/s}{m}} = \frac{N\cdot s}{m^2} = N\cdot s\cdot m^{-2}$$

$$\frac{N}{m^2} = P_a \Leftrightarrow [\eta]_{I.S} = P_a\cdot s$$

The CGS unit of the dynamical viscosity coefficient has the name of poise (P) after the name of Poiseuille and is 10 times smaller than the I. S unit: $1P = 0.1N\cdot s\cdot m^{-2}$.

A submultiple of poisel named centipoise (cP) often used in practice is 100 times smaller than the poisel: $1cP = 0.01P$. In the laboratory practice are also used:

– the coefficient of cinematical viscosity numerically equals to the ratio between the dynamical viscosity coefficient of the liquid and its density;

– the relative viscosity coefficient determined by the ratio between the viscosity of studied substance and the standard substance.

The importance of the determination of viscosity coefficient for medical practice

The knowledge of viscosity coefficient values is useful for revealing various diseases affecting the state of some biological liquids (the blood, serum, gastric juice, etc.).

The relative viscosity of the whole human blood ranges between 3.5 and 5.4 at the normal temperature. The viscosity of the blood depends both on the viscosity of the serum (1.5 – 2.2), plasma (1.9 – 2.3) and the number of erythrocytes and leukocytes which are in the unit volume, and also on its own volume.

The viscosity of the normal urine is a little greater than that of distilled water. The presence of proteins leads to the increase of viscosity.

The measurement of viscosity coefficient of liquids by the Ostwald viscometer

The method of determination of the relative viscosity coefficient of liquids by the Ostwald viscometer is based on the law of Poiseuille.

Studying the laminar flowing through capillary tubes, Poiseuille established that the volume debit of one liquid (the flowed volume in the time unit) is proportional to the fourth power of tube radius, with the loss of the pressure on length unit and inversely proportional to the dynamical viscosity coefficient:

$$Q = \frac{V}{\Delta t} = \frac{\pi r^4 \Delta P}{8\eta l} \quad (3) \qquad V = \frac{\pi \cdot r^4 \cdot \Delta p}{8 \cdot \eta \cdot l} \cdot t \quad (3a)$$

Here $d_p = p_1 - p_2$ and p_1 and p_2 are the input and output pressures of tube respectively.

Writing this law with the respective indices of the researched liquids and the distilled water (as the standard liquid), we obtain: for the distilled water (a):

$$Q_a = \frac{V}{\Delta t_a} = \frac{\pi r^4 \Delta P_a}{8\eta_a l} \qquad (4)$$

and for the researched liquid (x)

$$Q_x = \frac{V}{\Delta t_x} = \frac{\pi r^4 \Delta P_x}{8\eta_x l} \qquad (5)$$

When the hydrostatical pressure assures the flowing:

$$\Delta P_a = \rho_a g h \qquad \text{Bat} \qquad \Delta P_x = \rho_x g h$$

Dividing the equation (4) to the equation (5), the working formula is obtained:

$$\eta_x = \eta_a \cdot \frac{\rho_x \cdot \Delta t_x}{\rho_a \Delta t_a} \qquad (6)$$

The description of equipment

The determination of relative viscosity of liquid is performed by the Ostwald viscometer. The device represents a glass tube of the U form, which has only a bulb at different levels in the both branches, the tube has a capillary part between them (fig. 1.2).

The upper margin of the inferior bulb from the branch a is reached with the researched liquid. This is brought by absorption to the superior bulb from the branch **b** above the grade **m**, using a rubber pear.Then, the flowing time of the liquid between the marks **m** and **n** situated over and under the inferior bulb is determined by a chronometer.

The same operation is repeated with the distilled water. After that the viscometer is cleaned and dried. Let **t** be the flowing time of researched liquid and t_0 that of distilled water.

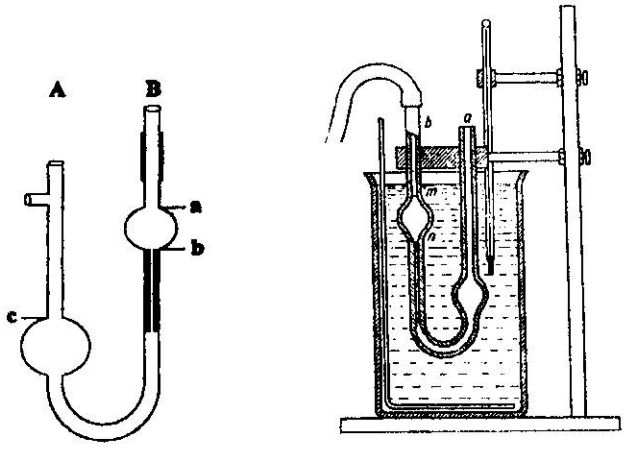


Fig. 1.2

Work procedure

The researched liquid is poured into the viscometer, so that the level of it reaches the grade **c** (the upper limit of the reservoir R_2).

The mark of liquid is aspired by the syringe till the level exceeds the grade **a**, situated above the reservoir R_1. Removing the syringe from the tube linked to the viscometer, the necessary time interval for the liquid to flow from the level of grade a up to the level **b** is measured. The necessary interval of time that the volume **V** of the liquid will flow through the capillary DE, is measured by the chronometer. Ten measurements are performed, after that the viscometer is emptied and washed with distilled water.

The procedures 1 and 2 are repeated 10 times with distilled Water. The work temperature is measured and the found value is marked. The density of researched liquid is determined.

The presentation of experimental results

Work temperature: t = ...°C

The dynamical viscosity coefficient of distilled water according to the table corresponding to this temperature is:

$$\eta_a = ...P$$

but its absolute density is (at the same temperature):

$$\rho_a = ...\text{g/cm}^3$$

Table 1.1

Nr of experiment	η_a, cP	ρ_a, g/cm^3	ρ_x, g/cm^3	t_a, s	t_x, s	η_x, cP
1						
2						
3						
4						
5						

The absolute density of the researched liquid determined by the densimeter is:

$$\rho_x = ...\text{g/cm}^3$$

The demonstrative experiment
The verifying of Stokes law

The resistance force in the case of flowing of the sphere of radius **r** into a liquid depends on its viscosity and is given by the Stokes law:

$$F = 6\pi\eta rV \qquad (7)$$

Where V is the moving speed of the sphere.

The moving of sphere will be uniform when the influence of those three forces (the weight force, the Archimede force and the resistance force) will be balanced. The following expression results from the equilibrium condition:

$$6\pi\eta rV = \frac{4}{3}\pi r^3 g(\rho - \rho') \qquad (8)$$

Where ρ is the density of the ball, but ρ' is the density of liquid.

The following expression results from the equation (8):

$$\eta = \frac{2}{9}\frac{r^2 g(\rho - \rho')}{V} \qquad (9)$$

As the concomitant falling of two balls with the different radius $\mathbf{r_1}$ and $\mathbf{r_2}$ are examined, the following expression results:

$$\eta = \frac{2}{9}\frac{r_1^2 g(\rho - \rho')t}{S_1} \qquad (10)$$

And respectively:

$$\eta = \frac{2}{9}\frac{r_2^2 g(\rho - \rho')}{S_2} \qquad (11)$$

The expressions are obtained respectively by equaling the right parts of equations (10) and (11):

$$\frac{r_1^2}{S_1} = \frac{r_2^2}{S_2} \quad or \quad \frac{r_1^2}{r_2^2} = \frac{S_1}{S_2} \qquad (12)$$

According to the equation (12) the passed distances of balls in the same interval of time always is divided as respective square radii balls.

The experimental confirmation of equation (12) is also an indirect verification of the Stokes law.

The devices used with this purpose are represented in fig. 1.3.

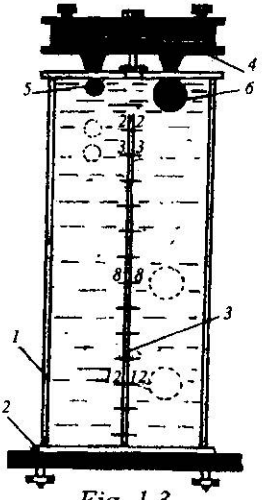


Fig. 1.3

The vase **1** is filled with the viscous liquid (for example, glycerin). In order to perform the experiment the coil of electromagnet is supplied from the continuos current supply. The balls are brought In the initial position by rotating of the device around the horizontal axis.

As the current is disconnected the falling of both the balls begins concomitantly. So the ball speeds are relatively small. It is established visually that at the moment when the small ball passes the division **2** of the scale drawn on the frontal wall of the vase, the big ball passes the division **8** (further the small ball - the division 3,

but the big one - the division 12). The passed distances of the balls in each equal time interval are divided as $1/4$.

The result of the experiment confirms the equation (12) and the law of Stokes.

Questions and tasks for self- control

1. Using the formula of Newton explain the physical meaning of viscosity coefficient; prove the dimensions of measure units into IS and in medical practice
2. Using the mathematical expression of the Poiseuille law prove the formula for the determination of viscosity coefficient value by the method of Ostwald
3. What does the blood viscosity depend on?
4. What are the normal values of viscosity coefficient for the basic biological liquids?
5. What is the practical application of viscosity method into medical practice?
6. What does the demonstrative device for performing the experiment that proves experimentally the Stoxes law consists of?

2. DETERMINATION OF SURFACE TENSION OF BIOLOGICAL LIQUIDS

Purposes:

- Presentation of theoretical notion of surface tension coefficient.
- The measurement of surface tension coefficient of liquids by stalagmometer.
- The study of surface tension dependence on solution by concentration.

Theoretical notions

The surface tension forces are born as a result of molecule interactions at the border between two phases and lead to the decrease of contact surface. These forces lead to the decrease of the surface of liquid. These forces act perpendicularly on the contour of each portion of the contour of free surface and tangentially to the liquid surface.

In the present case the forces of surface tension are of great interest, they appear at the border between the liquid phase and air.

The value of surface tension force corresponding to the unit of length of the free surface contour is called the surface tension coefficient σ :

$$\sigma = \frac{F}{l} \qquad (1)$$

The equation can also define the coefficient of surface tension:

$$\sigma = \frac{\Delta W}{\Delta S} \qquad (2)$$

Where ΔW is the variation of free energy, but ΔS - the variation of interphase layer.

It results that the value of surface tension coefficient is equal to the value of mechanical work for the decrease of liquid free surface by a unit.

The dimensional formula deduced from the equation (1) has the form:

$$\dim \sigma = \frac{ML/T^2}{L} = M \cdot T^{-2}$$

$$[\sigma]_{Practic} = \frac{dyn}{cm}$$

The unit measure is:

$$[\sigma]_{I.S} = \frac{N}{m} = \frac{J}{m^2}$$

Using the CGS unit system the following equation results:

$$[\sigma] = dyn/cm = erg/cm^2$$

We note that the surface tension coefficient is a constant, its value is influenced by the nature of two phases being in contact and their concentration, temperature, and also the possible action of different chemical or physical factors.

It results from the formula (1) that in order to determine the surface tension coefficient we must measure the force necessary for breaking liquid surface on the contour of free surface of liquid of the certain length. The most known methods of determination of surface tension coefficient are:

the method of plucking of drops;

the method of plucking of ring;

the method of liquid lifting into a capillary.

The importance of determination of surface tension coefficient for medical practice

Biological liquids usually represent the water saline solutions and have the surface tension coefficient smaller than that of the distilled water. Proteins are the substances that decrease the surface tension coefficient according to the Gibbs law, and they are concentrated at the surface of these liquids in the monomolecular layers playing a great role in the exchange of substances at the cell level.

The surface tension coefficient of the blood serum at the body temperature is about 67 dyn/cm. The normal value is modified by the presence of some substances or by the existence of some pathological processes (jaundice, neoplasia and others).

The existence of biliary acids, and the salts of these, decreases the value of urine surface tension coefficient that allows the easy detection of their presence.

The phenomenon of surface tension is the base of different living and pathological process taking place in the human organism such as hemotaxis, gaseous emboli, etc.

The dropper is built on the basis of surface tension phenomenon used for dosing some medicinal substances.

a) Measurement of surface tension coefficient of urine by the Traube stalagmometer

The stalagmometric method is based on the Tate law confirming that a drop of liquid comes off from the inferior extremity of a capillary oriented vertically when its weight becomes equal to the surface tension force exercised on the contour of drop. So:

$$G = F \quad mg = F \Leftrightarrow F = \sigma \cdot l$$

Where $mg = V \cdot \rho \cdot g$ is the weight of the drop, but $F = 2 \cdot \pi \cdot r \cdot \sigma$ - the surface tension force (V - the volume of one drop, ρ - the density of liquid), σ - surface tension coefficient,

g - free fall acceleration, r - the radius of capillary tube (Fig.2).

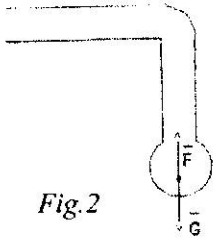


Fig.2

$$M = m \cdot n \Leftrightarrow mn = \rho V$$

$$m = \frac{\rho V}{n}$$

Description of the device

The Traube stalagmometer represents a capillary fixed vertically on a support. The middle portion of the capillary is a reservoir with the volume V bordered with the superior mark **a** and llic inferior *one* **b**, and with only 40 equal divisions between them (fig. 2.1).

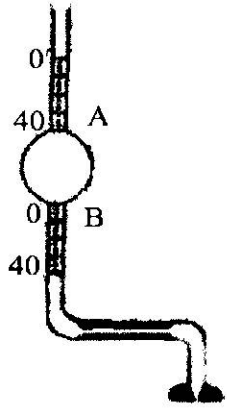


Fig. 2.1

A rubber tube is attached to the superior extremity, in which the liquid is aspired by the stalagmometer.

As **n** drops are flowed from the volume **V** of reservoir of stalagmometer, then the equation (3) becomes:

$$\frac{V}{n} \cdot \rho \cdot g = 2 \cdot \pi \cdot r \cdot \sigma$$

For the researched liquid (x) this equation becomes:

$$\frac{V}{n_x} \cdot \rho_x \cdot g = 2 \cdot \pi \cdot r \cdot \sigma_x \qquad (4)$$

And for the standard liquid (a):

$$\frac{V}{n_a} \cdot \rho_a \cdot g = 2 \cdot \pi \cdot r \cdot \sigma_a \qquad (5)$$

$$\frac{\rho_x n_a}{\rho_a n_x} = \frac{\sigma_a}{\sigma_x}$$

Dividing each other, (4) and (5), we find the equation

$$\sigma_x = \sigma_a \frac{n_a}{n_x} \cdot \frac{\rho_x}{\rho_a} \qquad (6)$$

Equipment and materials

The Traube stalagmometer, a Berzelius glass for the distilled water (the standard liquid), a Berzelius glass for the researched liquid (urine), thermometer.

Work procedure

The measurements are reduced to the determination of the number of drops flowing from the volume V of the reservoir of

stalagmometer for the researched liquid and the standard one (under the same conditions).

The liquid volume of 10 – 15 cm^3 (distilled water or the researched liquid) is poured from the glass on the table into a Berzelius glass. The liquid is aspired in the stalagmometer by the syringe attached at the superior extremity over the mark "O" of the graded scale situated above of the reservoir V.

The initial level of liquid is chosen so that taking off of one drop is assured at the moment when the liquid level is in the upper part of the superior graded scale (for example, at the division "3"). The level of the liquid is retained at the moment when the following drop is taking off by the free fall (for example, at the division "25").

All the drops which are born are counted till taking off of one drop takes place, when the level of liquid reaches the inferior scale (for example, at the division "9", 45 drops are formed).

Ten measurements are performed for the distilled water and ten measurements for the researched liquid respectively, and ten pairs of values written in the table 2.1 are obtained respectively.

The presenting of experimental results

The work temperature: t =...0C and the surface tension coefficient of the distilled water for this temperature is respectively:

$$\sigma_a = ...dyn/cm$$

The absolute density of water at the same temperature is:

$$\rho_a = ...g/cm^3$$

The absolute density of the liquid is determined by the densimeter:

$$\rho_x = ...g/cm^3$$

. The number of drops and fractions of drop for each measurement is obtained analogously with the numerical example mentionned in the work procedure.

We have a drop from division 3 to division 25 (the superior scale), so that 25 – 3 = 22 corresponds to one drop. The number of drops formed only from the volume V of stalagmometer reservoir results in subtracting the number of drops of supplementary volumes from 45 (15 divisions up of reservoir – 9 divisions under the reservoir is equal to 24 divisions). One drop corresponds to 22 divisions, but ч drops correspond to 24 divisions. So, that

$$x = \frac{24}{22} = 1.1 \ drops$$

As a result, the number of drops **n** from the volume V is:

$$n = 45 - 1.1 = 43.9 \ drops$$

The obtained experimental results are given in table 2.1

Table 2.1

Nr of experiments	σ_a, dyn/cm	ρ_a, g/cm^3	ρ_x, g/cm^3	The number of drops		σ_x, dyn/cm
				n_a	n_x	
1						
2						
3						
4						
5						
6						

b) Study of solution surface tension dependence on its concentration by the method of ring taking off

Take a metal ring for which the researched liquid is adherent.

The ring is brought in contact with the free surface of liquid, a thin layer of liquid is adhered to the ring, the surface forces act on it that are composed into the result F_t (fig. 2. 2).

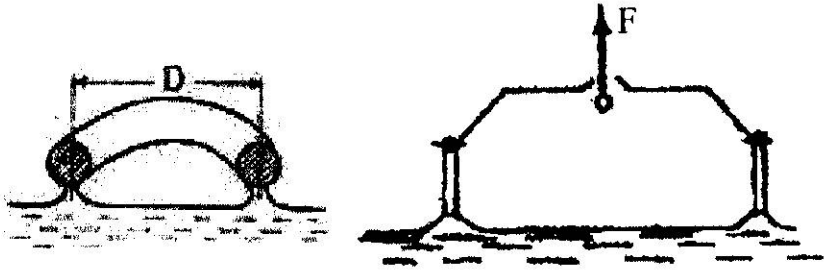


Fig.2.2

Taking into consideration the fact that the mentioned layer has two surfaces (interior and exterior), the result force of surface tension is determined by the equation:

$$F_t = 2 \cdot \pi \cdot r_1 \cdot \sigma + 2 \cdot \pi \cdot r_2 \cdot \sigma = 2 \cdot \pi \cdot \sigma (r_1 + r_2) \quad (7)$$

Where σ is the surface tension coefficient.

r_1 – the internal radius of the ring,

r_2 – the external radius of the ring.

To take off the ring from the surface of liquid we must act with the force F:

$$F = F_t$$

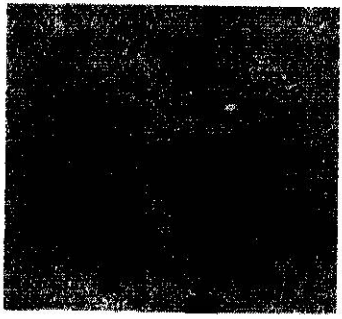
Taking into consideration the equation (7) and determining this force by the torsion balance the work formula is obtained.

$$\sigma = \frac{F}{\pi (d_1 + d_2)} \quad (8)$$

Where d_1 and d_2 are the diameters of the ring respectively.

Description of the equipment

The mechanism of torsion balance used by us is mounted on the metallic support 1 and protected by the screen 2 (fig. 2.3).



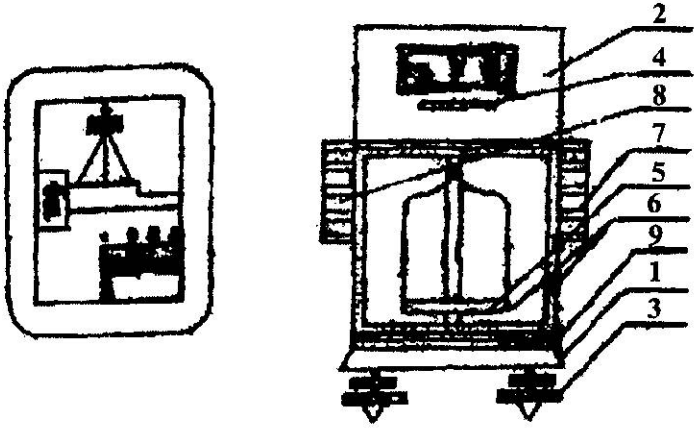


Fig.2.3

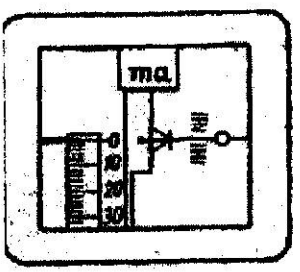
The support is fixed horizontally by two adjusting screws 3 and is retained after the level 4.

The scale 5 is suspended by the extremity of one mobile lever. The glass door 6 excludes the influence of the air moving in the vicinity of apparatus. The adjusting knob 7 is situated at the right

part of the balance used for the setting the mobile needle at the zero division of the graded scale.

The knob 8 is situated in the left part of the balance used for the rotation of the drum with the graded scale, the result of weighing is maintained on it. The knob 9 serves for blocking the lever mobility when the balance does not work or during transportation.

Weighing is performed in the following way: the protective door is opened and the weighed body is placed on the scale by tweezers. Then the door is closed and the left hand rotates the knob 8 (from us), thus assuring the coincidence of mobile indicator needle with the red mark of equilibrium. Concomitantly with the rotation of the knob 8 the mobile graded scale is rotated. The mass of the researched body is retained on this scale after the indication of immobile needle, the value of one division is 1 mg.



After reading the result the graded scale is returned in the initial position, the knob 8 is rotated to us.

Work procedure

We determine the value of surface tension force formed at the moment when the ring is taken of from the surface of liquid by using the examined torsion balance.

In order to perform it, the metal ring is hung instead of the scale. The diameters of the ring (interior and exterior) are indicated on the worktable.

The solutions of ethyl alcohol (96%, 75 %, 50%, 255 and 0%) are used as the searched liquids.

Three measurements are performed for each concentration and the average value of the surface tension force is determined.

The obtained results are written in table 2.2

Table 2.2

Nr of experiments	The concentration	F, dyn	Dyn/cm
1 2 3			
Average value			

The diagram of the dependence of surface tension coefficient on the solution concentration is built on the scale paper, placing the values of surface tension coefficient on the ordinate axis, and the respective concentrations on the abscise axis.

Their concentrations are identified using the drawn diagram and the surface tension coefficient of solutions is determined experimentally.

Questions and tasks for self – control

1. What are the two definitions of surface tension coefficient and units of measure in IS?

2. Describe the construction of the devices and explain the proper methods of surface tension coefficient measurement (the stalagmometric method and the method of ring plucking).

3. TENSIOACTIVITY OF PULMONARY SURFACTANT

Purposes:

- presentation of theoretical notions of the molecular structure of tensioactive substances;
- study of molecular composition, specific properties and physiological role of pulmonary surfactant;
- familiarization with medical practice aspect of pulmonary surfactant;
- study of preparation method of surfactant and modes of molecular layer production;
- drawing of hysteresis according to the experimental data obtained in the laboratory;
- visualizion of elasticity properties of surfactant molecular layer.

Theoretical notions

The force of surface tension leads to the decrease of liquid surface energy. For pure liquids the possible way is the decrease of free surface.

Another way is reali3ed when the liquid contains impurities of tensioactive substances that are stored at the surface of liquid.

The molecules of tensioactive substances are made of polar grouping (COOH, OH, NH_2) represented by a circle and a nonpolar paraffin part under the form of chain (fig. 3.1).

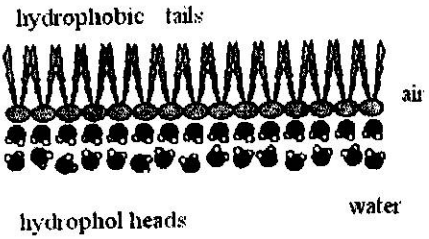


Fig. 3.1

The polar grouping is hydrated by the water medium and is named hydrofoil, but the nonpolar one, not dissolved in the water, is named hydrophobia.

As a result, the molecules of tensioactive substances "are floating" on the liquid surface in the vertical position and have the paraffin chain at the upper part. The decrease of surface tension lasts till the moment when the surface layer is saturated with the molecules of tensioactive substance.

At small concentrations a monomolecular layer is formed on the surface of the liquid in which the so -called surface pressure is manifested. The equation determines the value of this pressure:

$$P = \sigma_l - \sigma_m \qquad (1)$$

σ_l is the surface tension coefficient of the pure liquid, but

σ_m - the surface tension coefficient of monomolecular layer. For the concrete conditions σ_l is a constant but value, σ_m varies dependending on the monomolecular layer surface (of surface concentration of tensioactive molecules).

At the contraction of the surface σ_m is decreased, but the surface pressure c is increased; at the dilatation of the surfaces the inverse phenomenon takes place.

Such a process takes place in the pulmonary alveoli due to the monomolecular surfactant layers situated on their walls. On inspiration the surface pressure decreases, but on expiration it increases.

The dependence of pressure surfactant surface on the surface value is expressed by a closed curve named hysteresis (fig. 3.2).

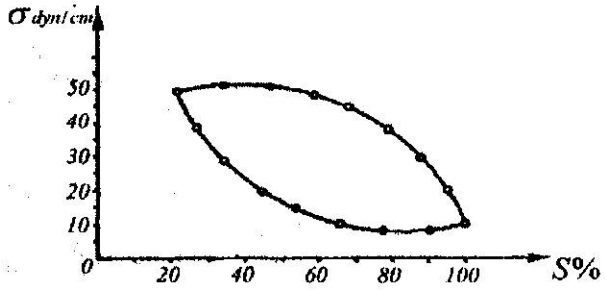


Fig. 3.2

The area of hysteresis is equal to the used energy for the contraction - dilatation act.

The property of a tensioactive substance to form a stable monomolecular layer is characterized by the stability index:

$$IS = \frac{2(\sigma_{max} - \sigma_{min})}{\sigma_{max} + \sigma_{min}} \qquad (2)$$

Where σ_{max} is the maximal value of surface tension coefficient lor the surface of 100% of monomolecular layer (the beginning of contraction), but σ_{min} – the minimal value corresponding to the surface of 20 % of monomolecular layer (the end of contraction)

For the alveolar stratum of mammals the value of stability index is not smaller than 0.8. The smaller values mean the insufficiency of surfactant or the loss of tensioactive properties.

Importance for medical practice and pharmaceutics

A series of biological compounds possesses tensioactive properties, which are of a great importance for vitality processes. As it was already mentioned, the normal functioning of pulmonary alveolus takes place in the presence of tensioactive substances named surfactant and synthesized during the whole life by special

cells situated in the alveolus walls. The loss of quality and an insufficient production of surfactant are the causes of different diseases of the respiratory system.

The tensioactive substances also occur in the digestive process. The biliary acids are tensioactive in comparison with burliness. Decreasing the surface tension they lead to the division of burliness drops, thus increasing the contact surface of ferments with the food substances and the digestive speed respectively. These substances present an interest from the pharmaceutical point of view because they allow performing mixture of two liquids by the decrease of interfacial tensions, which normally are not mixed.

Description of the installation

The working installation is presented in fig. 3.3 The principal components are the torsion balance 1 and the tub 2 with the paraffin internal part.

The value of one balance division is 0.98 dyn.

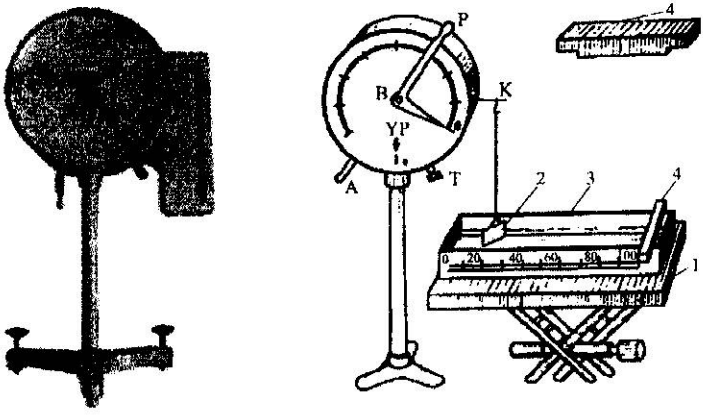


Fig. 3.3

A metallic mount 3 is hung on the plate of the balance (it is recommended to be of Pt) with the length **l** and the thickness **d**. In this case the balance can be graded in the units of surface tension, the value of one division is:

$$K = \frac{0.98dyn}{2 \cdot (l+d)cm} = \frac{0.98}{2 \cdot (l+d)cm} dyn/cm$$

The tub in which the physiological solution is poured is supplied with the graded scale and immobile barrier 4.

It is placed on the table 5 with the mechanism 6 for lifting and lowering.

Creation of surfactant monomolecuiar layer

A gram of pulmonary tissue prepared from the rat lung is broken being mixed with 10 ml of physiological solution. After the filtration 150 ml of physiological solution is added and poured in the tub 2. To form the monomolecuiar layer we have to wait for 15 – 20 minutes. The mentioned quantitative proportions correspond to the concrete dimensions of tub 21 x 5, 5 x 2 cm^3.

The work process

The tub with the solution is placed on the table 5. The mobile obstacle is fixed at the division 100 %. After the equilibrium of the balance the table is lifted till the top of the blade comes in the solution. Then by taking off the mount the value of surface tension coefficient σ_{100} is determined. The measurements are made at the consequent contraction with 10 % of monomolecuiar layer by the respective removal of the obstacle 4.

The last measurement being at the division of 20%, the monomolecuiar layer is dilated, repeating the measurements in the inverse order.

For each measurement the surface pressure c is calculated according to the equation (1).

The experimentally obtained results are written in table 3.1

The diagram P=F(S) is drawn according to the data from the table. The index of monomolecuiar layer stability is calculated according to the equation (2).

Table 3.1

Compressed				Dilated		
S%	balance indication s, n	Surface tension coefficient, σ, dyn/cm	P, dyn/cm	N	σ, dyn/cm	P, dyn/cm
100						
90						
80						
70						
60						
50						
40						
30						
20						

Note:

Before performing the measurements, the tub must be washed with distilled water, but the mount is washed with ether, alcohol and distilled water.

The functioning of the balance is verified by measuring the surface tension coefficient for the distilled water. The result must be of the order 72 dyn/cm.

Questions and tasks for self- control

1. What is the molecular structure of tensioactive substances?
2. What mathematical expression is the value of surface pressure determined by?
3. What is the chemical composition and preparation method of pulmonary surfactant?
4. Describe the constructive elements of the used device for the determination of surface pressure of surfactant layer.

4. CELL OSMOTIC PHENOMENA

Purposes

- Theoretical notions of osmotic phenomena.
- Study of the devices applied for pointing out and measurement of osmotic pressure.
- Turgescent and plasmolysis phenomena.
- Evaluation of the cell dimension by the method of two micrometers.
- Familiarization with biological and medical aspects of osmotic pressure.

Theoretical notions

Take a vase divided in two parts by the dividing semipermeable wall. In one part there is water, but in the other part the substance solution is, their molecules do not pass through this separable wall (fig. 4.1).

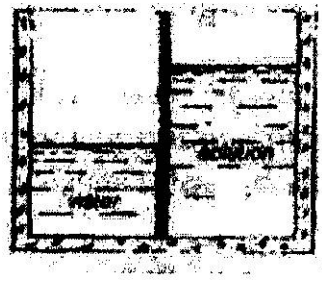


Fig.4.1

Practice shows that the water passes through the separator wall and after a while the difference of level is established in the divisions of the vase and, the difference of pressures, named the osmotic pressure of solution. Thus the osmotic pressure is the pressure, which must be exercised on the solution to bring it in the equili-

brium with the pure solvent separated by it through the semipermeable membrane.

Living organisms contain water in the proportion 50–90 % of their total weight. Different substances are dissolved in water making a solution. The membranes of living cells being semipermeable and selective let pass water and different substances as a function of metabolic necessities. Just in the case of small concentrations, the osmotic pressure can obtain the essential values. In different cells of vegetable nature its values can be of 5–20 atmospheres, assuring the penetration of water from ground to the high heights (for example, up to the tops of trees). The action of osmotic pressure is felt by swimmers when they open the eyes in water (especially in the lake water); the interocular pressure is increased due to the penetration of water through the cornea.

Experiments confirm that the osmotic pressure is similar to the pressure of ideal gases having the same laws.

The first law of osmoses is the law of concentrations according to which the osmotic pressure of dilute solutions at the constant temperature is in proportion to the molar concentration of the solvent.

$$P_{osm} = K_T \cdot C^M \qquad K_T = const \Leftrightarrow \frac{N \cdot m}{mol}$$

The law of temperature: for the examined solution, the osmotic pressure increases in proportion to the coefficient 1 + αt, the value α being equal to 1/273. (At higher temperatures this law is not respected).

$$P_{osm} = K_C \cdot T \qquad K_C = const \Leftrightarrow \frac{N \cdot m^{-2}}{^oK}$$

The law of Vant Hoff: the osmotic pressure does not depend on either the solvent nature or the nature of the dissolved substance, it depending only on the number of particles from the volume of solution.

It follows from the laws of osmoses that the osmotic pressure can be calculated after the formula of Mendeleev-Clapeiron:

$$P_{osm} \cdot V = n \cdot R \cdot T \qquad (1)$$

Where V is the volume occupied by the solution, R – gas universal constant, T – absolute temperature, n – number of moles. The following equation results from the equation (1):

$$P_{osm} = \frac{nRT}{V} = C^M RT \qquad (2)$$

Where $C^M = \frac{n}{V}$ is the molar concentration of solution.

The law of Dalton is confirmed for a mixture of solutions.

Acording to this the total osmotic pressure is equal to the sum of osmotic pressures of each solution, taking in consideration that each dissolved substance has its own osmotic pressure, as though it were llngle in the whole quantity of solvent.

The osmotic pressure of two different solutions, which have the Oommon solvent, is the same at the equal temperatures and molar I onccntrations. These solutions are named isotonic.

The medium with the less or greater osmotic pressure or greater with respect to another medium is named **hypotonic** or **hypertensive** respectively. The solutions being in contact with the semipermeable membrane bear a flux of solvent always oriented to the hypertensive medium.

Noilet observed the phenomenon of osmoses in 1748 for the lirst time. The detailed study belongs to Dutrochet.

The devices used for revealing and measurement of osmotic pressure are named osmometers. The osmometer of Dutrochet is represented in Fig 4.2; it is made of glass vase, in which the bottom is replaced by a semipermeable membrane (a pig's urinary bladder or cellophane).

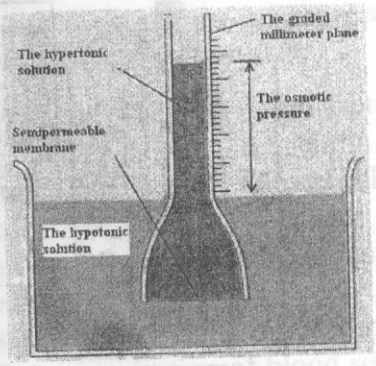


Fig. 4.2

The vase is prolonged at the superior part with a capillary tube placed on the graded millimeter plane. The solution of sugar is introduced in the osmometer up to the inferior level of the vertical tube and is sunk into the vase with distilled water that must be at the same level with the solution from the interior. The liquid from the internal vase will rise slowly in the capillary tube. The maximal height of the column determines the osmotic pressure of solution.

There is a series of indirect methods of osmotic pressure measurements based upon the procedures of determination of solution molar concentration after which the osmotic pressure is calculated. In biology this method is called the **cryoscopy** (the molar concentration is determined after the solidification point of solution).

The fall of solidification point of a solution is in proportion to the number of dissolved molecules in the unit of its volume that is with its molar concentration C_m:

$$\Delta t = t_0 - t_1 = K \cdot C^M$$

Where t_0 is the freezing temperature of pure solvent; t_1 - the freezing temperature of solution.

The determination of cryoscopic point of solution is made by the Beckman cryoscop (fig. 4.3)

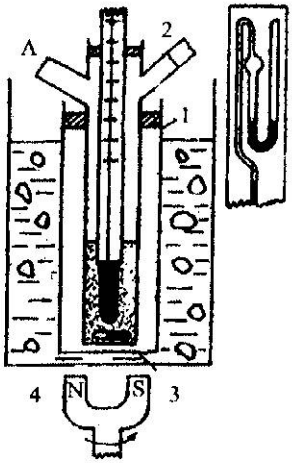


Fig. 4.3

It is supplied with a vase made of glass in which a cooling mixture is introduced (ice and salt). In this mixture two coaxial test tubes of glass are introduced. The interior test tube 2 is supplied with two lateral branches and on the axes with a punched cork through which the thermometer Beckman is introduced inside. The air stratum between two test tubes allows a slow cooling of solution.

The state of extra fusion is performed by stirring the magnetic bar 3, being in rotation moving by the magnet 4 in the form of a horseshoe. The magnet is fixed on the axes of electrical motor lUpplied with the electrical set.

The used thermometer named the Beckman thermometer has the reduced scale graduated in hundredths of degree. In the upper part it is supplied with a supplementary reservoir with mercury, which communicates with the inferior reservoir. The mercury from ihis reservoir is used for adjusting mercury quantity from the inferior reservoir, that is necessary to make possible the measurement ol temperature variation in the needed interval.

The importance of osmosis phenomenon study for medical practice

The osmotic pressure is the important factor that assures the maintenance of the volume and consequently: the cell structure, the morphological and functional integrity of cells.

The osmoses take place in the exchanges of substances between organisms and an environmental medium between cells and an extracellular medium.

The determination of cryoscopic point can give the information of the molecular concentration of blood serum or other biological liquids. The calculation of osmotic phenomenon allows the study of functioning of various physiological systems. Comparing the osmotic pressures of different biological liquids in the normal and pathological state, some aspects of hydroelectric metabolism of renal function can be followed.

The passage of some substances between the vascular and intercellular compartments takes place under the influence of pressure difference on the both parts of the capillary wall.

The cell volume varies as a dependence of extra cellular medium concentration. This variation offers the model for the study of permeability phenomena of cell membranes.

We note that in order to introduce various solutions in the blood circulation, these must be isotonic, and not make modifycations of cell volume.

Turgescent and plasmolysis phenomena in the vegetable cells

The principle of method. In order to take into account the volume modifications of the cells produced by the concentrations of intercellular media the optical microscope is used.

In hypotonic media water penetrates by the endosmosis to the interior of the cell leading to turgescent in vegetable cells.

This increase of volume is limited by the cell wall, rigid in respect to the plasmatic membrane of the vegetable cell. (Fig. 4.4, a)

For animal cells the absence of rigid wall limiting the increase of volume in the hypotonic medium leads to a break of cell membrane. In the particular case of red cells the phenomenon is called haemolysis.

In the hypertensive media the water is going out by the exosmose from cells resulting in the separation of those two membranes of the vegetable cells; this phenomenon is named plasmolysis. The cell is decreased (wrinkled) and the plasmatic membrane is taken off from the cell coat. (fig. 4.4, b). In the hypertensive medium the red cell decreases the volume by itself.

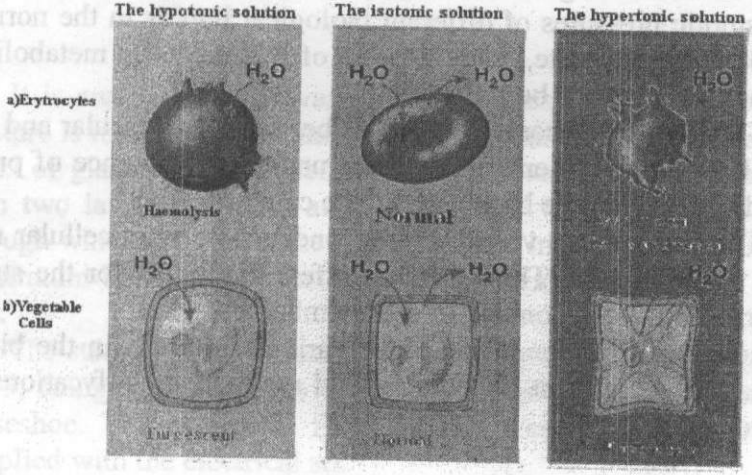


Fig.4.4

Equipment and materials

The binocular microscope with the ocular micrometer and object lens micrometer; 4 glass blades; 4 mounts; 3 Petri vessels; 20 ml of NaCl isotonic solution; 20 ml of 20% NaCl solution (the hypertensive solution); 20 ml of distilled water (the hypotonic

medium); 3 Pasteur pipettes; lint; xilol, razor blade; 6-8 leaves of *Elodeea canadiensis* or of *Valisneria spiralis*.

Work procedure

Only 20 ml of distilled water, NaCl isotonic solution and hypertensive solution are poured in those three Petri vessels, respectively. Some leaves of alga are put in each Petri vase. In 30 minutes each leaf from each medium is examined under the microscope.

In order to examine the leaf of *Valisneria spiralis* under the microscope the leaf is sectioned thickly by the razor blade, and then a more transparent fragment is maintained. The leaf of *Elodeea canadiensies* does not need any sectioning because it is transparent.

For this item of the paper we shall follow only the morphologic aspect of vegetable cell from those three media, then their diameters are measured according to the subitem b.

Evaluation of cell dimensions by the method of two micrometers

The principle of the method is the comparison of the measured object by the overlapping its microscopic images of a preliminary standard graded scale

The objective micrometer is a glass mount in its center where there is a segment of 1 mm divided in 100 equal parts (the value of 1 division is 0.01 mm or 10 μm). The ocular microscope is a glass disk on which the equidistant divisions as a grading are drawn.

The objective micrometer is set on the microscope plate. The ocular micrometer is introduced in one of the ocular microscopes. The ocular system with the ocular micrometer is introduced in the optical tube. The adjustment screws are maneuvered so that the overlapping and cleared images of those two micrometers are obtained in the field of vision.

The number of divisions of the objective micrometer corresponding to a square of ocular micrometer are noted and, knowing the value of one division of objective micrometer (10 μm), the number of microns representing the side of one square of ocular micrometer is determined.

A mount with a fragment of the leaf from the isotonic medium replaces the objective micrometer. The number of squares from the ocular grading overlapping on each longitudinal and transversal diameter is noted for 10 cells.

The same procedure is performed for the same number of cells with the leaves fragments from hypotonic and hypertensive media.

Knowing the dimensions of a square of ocular micrometer, the values of transversal and longitudinal diameters of the measured cells are transformed in μm. The experimental results are written in table 4.1.

Table 4.1

The number, of cells	The hypotonic medium		The isotonic medium		The hypertensive medium	
	The longitudinal diameter (μm)	The Transversal diameter (μm)	The longitudinal diameter (μm)	The Transversal diameter (μm)	The longitudinal diameter (μm)	The Transversal diameter (μm)
1 2 3 4 5 6 7 8 9 10						
The average values						

Questions and tasks for self-control

1. Describe the devices used for pointing out and measurement of osmotic phenomena.
2. Formulate and write the mathematical formulae of osmoses laws (the law of concentrations, the law of temperature, the law of Vant Hoff, the law of Dalton).
3. What is the technique for the plasmolysis and turgescent phenomena in vegetable cells?
4. What are medical biological peculiarities of the osmotical pressure and the method of osmometry?

5. ULTRASOUND EFFECTS. OBTAINIG EMULSION BY ULTRASOUND METHOD

Purposes

- obtaining emulsion of castor oil in water by the ultrasound method;
- obtaining emulsion of castor oil in water by the mechanical method;
- evaluation of the obtained emulsions by ultrasound and mechanical methods.

Theoretical notions

Ultrasounds are mechanical longitudinal waves, their frequency ranges between $2 \cdot 10^4$ Hz and 10^{10} Hz. The upper limit of ultrasound oscillations can be considered theoretically 10^{13} Hz, thus it can correspond to the frequencies at which the wavelength becomes commensurable with the intermolecular distances. The biggest obtained frequencies for solid bodies are called 10^{11} Hz.

Ultrasound emitters called ultrasound transducers obtain ultrasound. Ultrasound emitters have two main components: a generator that produces an alternative current of the required frequency, and a vibrator that transforms electrical energy of this current into mechanical vibrations.

In order to create ultrasound waves two phenomena are used at present: the piezoelectric effect (electrostriction) and magnetostrictive effect.

The ultrasound piezoelectrical emitter is based on the inverse piezoelectrical effect. It consists of the fact that some crystalline materials (tourmaline, the cut quartz after certain axis, the lithium sulfate, the monosulfite of potassium and of ammonium, etc.) are deformed as a result of electrical field influence by the application of potential difference at two chosen points of the material.

The material vibrates by the application of alternative voltage and emits ultrasound waves.

The frequency of the field coincides with that of the crystal and fa amplitude of vibration becomes maximal in the case of resonance. The resonance frequency of quartz mount depends on its dimension:

$$\frac{285}{d,\ kHz} \qquad (1)$$

Where **d** is the thickness of the plate in cm.

The intensity of the ultrasound as a function of used alternative voltage frequency (v) is calculated after the equation:

$$I = 3.6 \cdot 10^8 v^2 u^2 / \rho C$$

Where I is the intensity in W/cm^2, U – the voltage in V, pC – the characteristical impedance of the medium (p- the density of the medium, C – the speed of ultrasound waves in the given medium).

The magnetostrictive emitter is based on the magnetostritive effect that consists of dimension modifications of one ferromagnetical body under the influence of magnetical field. A rod of length I suffers shorting at each alternation of the field, thus oscillating with the double frequency in comparison of the applied voltage. The equation gives the own frequency of vibration of the rod:

$$v = \frac{1}{2l}\sqrt{\frac{E}{\rho}} \qquad (2)$$

in which **l** is the length of the rod, **E** - the elastical module, but ρ - the density of the material of which the rod is made.

The ultrasound waves like sonorous waves can propagate in solid, liquid and gaseous bodies. The speed of propagation of ultrasound waves depends on the elasticity and density of the medium and ranges between $3 \cdot 10^2$ m/s and $6 \cdot 10^3$ m/s. As the wavelength (λ) of ultrasound is small (0.2 and 0.3 m), it propagates in the form of strictly oriented narrow beams. The laws of reflection

at the interface between two media are corrected for the ultrasound and, therefore, it can be oriented from the emitter in the strictly determined directions.

The intensive absorption by the medium is characteristic of ultrasound which is expressed qualitatively by the equation:

$$I = I_0 e^{2\mu \cdot l} \qquad (3)$$

Where **I** is the intensity of ultrasound passing through the substance stratum with the thickness **l**; $\mathbf{I_0}$ - the intensity of incident ultrasound on the substance layer; $\boldsymbol{\mu}$ - the absorption coefficient.

Ultrasound is absorbed intensively by gases. So, for example, $\boldsymbol{\mu}$ - for the gases is 1000 times greater than for water. Therefore, its action on the object by some intermediary medium that assures a good contact, takes place. These media are oil, vaseline, water, etc.

As the volume density of wave energy ($W = \rho \cdot A^2\omega^2 / 2$) is proportional to the square pulsation ω, the ultrasound sources can emit oscillations of considerable energies (up to 20 W/cm^2) with the pressures often atmospheres. So, the pressure varies million times per second with ten atmospheres in microvolumes of substance. Such mechanical action leads to a series of phenomena, we note cavitation among them. The enormous concentration of energy, considerable acceleration transmitted to a substance, compressions and dilatations in one point become so strong, that the liquids "are broken". The small gaseous bubbles are created. They are filled with vapors that dilate rapidly and return to the extremely small volume (implosions). These implosions create shock waves of hundred and thousand atmospheres causing a break in chemical bounds and ionization and, as a result, the destruction of substance microstructure.

For technical production of ultrasounds the electro – and magneto – mechanical transducers are used. They transform electrical or magnetic energy into mechanical energy.

The transducer (emitter) of ultrasounds consists of two principal parts: a generator producing an alternative electrical voltage of the certain frequency and a vibrator transforming the electrical energy into mechanical vibrations.

The ultrasound piezo – electrical transducer is based on inverse piezoelectric effect that consists in the elastic deformation of some crystals on the application of potential difference between their faces. Such crystals are: quartz, Segnette salt, some ferrum – electrical pottery materials with induced piezoelectricity by cooling into an electrical field, etc.

The application of alternative electrical voltage on the plate 1 (fig. 5.1) on the surface of which are applied the electrodes 2 connected to the generator 3 determines the successive compressions nul dilatations of it, emitting acoustical waves (ultrasounds).

The resonance intensity (maximal) of these waves is obtained when the frequency of alternative voltage coincides with the own frequency of vibration of the used crystal. It depends on the crystal plate dimension, its orientation in respect to the crystalline planes fend is given by the empirical equation (1).

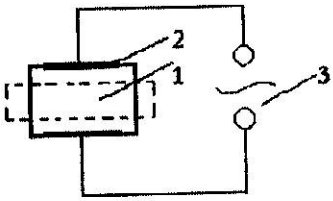


Fig. 5.1

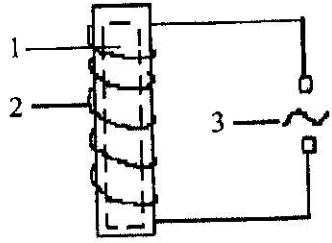


Fig. 5.2

The magnetostrictive transducer (emitter) is based upon the magnetostrictive effect that consists of the modification of the dimensions of a ferromagnetic body situated in the longitudinal magnetic field.

The magnetostrictive emitter consists of a bar **1** (ferromagnetic body) of the length *l* (fig.5.2) placed into a solenoid **2** supplied with an alternative electric current from generator **3**. The bar is compressed when the instantaneous electrical current from solenoid reaches the maximal values and returns to the initial dimensions when the values of electrical intensities pass through null values. So the bar experiences shortenings at each alternation of magnetic field that oscillates with a double frequency in respect to the applied voltage frequency.

The own frequency is given by the equation (2).

A device (the receiver of ultrasounds) working on the base of direct piezoelectric effect performs the reception of ultrasounds. The ultrasound waves lead to the polarization of piezoelectric crystal and then to the generation of alternative electrical field, the electrical signal being amplified.

The propagation of ultrasound waves through elastic media takes place with the speeds depending on the elasticity and density of medium, ranging within $3 \cdot 10^2$ and $6 \cdot 10^3$ m/s. Having a small wavelength (λ) (0.2 m and 0.3 m), the ultrasounds propagate in the form of narrow strict oriented fluxes. They can be localized by mirrors or acoustical lenses, obtaining the concentration of energy up to 5 kW/cm^2.

The propagation of ultrasounds is followed by reflection, refraction, diffraction, interference and absorption phenomena of the transported energy.

Reflection laws of ultrasounds take place at the interface of two media with different acoustical impedance. The waves reflect well at the interface muscle – periosteum, on the surfaces of cavity organs, etc. Thus the position and the dimensions of internal organs of cavities, etc. (ultrasound location) can be determined.

The specific acoustical impedance of air is about $\approx 3 \cdot 10^3$ times greater than the specific acoustical impedance of biological media. Therefore, if the ultrasound emitter is stuck on the human body,

the ultrasounds will not penetrate into it, but they will reflect from the thin air layer between the emitter and the human body. To exclude any air layer, an oil, vaseline, water layer, etc. must cover the surface of the ultrasound emitter.

The diffraction of ultrasounds depends on the correlation between their wavelength (λ) and the dimensions of bodies which the waves suffer diffraction from. A body with the greater dimensions than the wavelength of ultrasounds becomes an obstacle for them, the "ultrasound shade" appears that allows the neglecting of waves diffraction in these cases.

The intense absorption by the medium is characteristic of the ultrasounds. The intensity of the waves (I) after they have passed through the layer of thickness (d) with the attenuation coefficient by absorption or penetration power for the ultrasounds (μ), decreases after the exponential law and it is expressed by the above equation(3).

The blood absorption coefficient is 1.55, for muscles – 13.00, hone – 155.00.

Physical effects of ultrasounds

Physical effects of ultrasounds are mechanical, thermal, rical, optical, chemical. Cavitation, dispersion, precipitation, coagulation, degasing of liquids, etc. result from mechanical effects.

An especially complex phenomenon followed by important

mdary effects called cavitation appears on the propagation of Ultrasounds with high energies in liquids.

The volume density of ultrasound wave energy is proportional to the square pulsation ω:

$$E = \frac{1}{2}\rho \cdot A^2\omega^2 \qquad (4)$$

Therefore, the ultrasound sources can emit oscillations of a considerable energy (up to 20 W/cm2) and the pressures of 10 Atmospheres. In this case the local breaking of liquid occurs with the apparition of some cavities initially vacuumed, and then filled rapidly with the rarefied gases (born from dissolved gases) or with liquid vapors. The pressure varies by ten atmospheres in created bulbs with the rhythm of ultrasound frequency. Each zone of liquid necessary for some successive compressions and dilatations. If the own resonance frequency of the cavity (it depends on its radius) greater than the ultrasound frequency, the compressions and dilatations in a point become so strong that the liquids "are broken". The small gaseous bulbs are formed, that are filled with vapors, which are dilated rapidly, then they return to the extremely small volume named implosion.

The implosion lasts for (0.01–0.1) μs, and is developed adiabatically and leads to the local increase of temperature up to $\approx 10^4$ °C.

They produce shock waves with pressures up to ten thousands atmospheres causing the break in chemical bounds and the ionization and, finally, the break in substance microstructure.

Another mechanical effect of ultrasounds is dispersion taking place at intensities and great frequencies. The irradiation by ultrasounds allows the formation of some disperse systems, emulsions, aerosols, hydrosols. The effect takes place in the substance particles that oscillate with different amplitudes, especially in liquids due to cavitation. The shock wave from the moment of implosion leads to the break in substance particles.

The inverse phenomenon of dispersion is the precipitation that appears at the small intensities of ultrasounds. Under the influence of acoustical pressure the movement of particles is amplified and leads to the increase of collision probability and to the formation of molecular aggregates.

Relatively stable cavities are linked and lifted to the surface of liquid, emitting dissolved gases.

This phenomenon is named degasing of liquid. The thermal effects are due to the absorbed energy of the substance, depending on the intensity and ultrasounds frequency and on the increase of absorption coefficient μ. It is manifested at small intensities by the increase of temperature of the tissues due to ultrasound energy transforming into heat.

The electrical effects consist of apparition of alternative voltage into the liquid as a result of oscillations of electrical charge carriers. On the separation surface between two media a double ionic layer is formed. The electrical potential differences appear in the cavities between their walls and they can produce electrical discharges into rarefied gases.

The optical effects consist in the modification of refractive index of the substance as a result of successive compressions and dilatations of the medium that the ultrasound waves are propagated in. The periodical variations of the density take place. A diffraction grading is obtained for the light beam, allowing the visualizion of ultrasound wave. The break of bulbs during the cavitation is followed sometimes by the emission of ultraviolet radiation.

Chemical effects depend on medium temperature and substance concentration. They are related by cavitation consisting in the development and acceleration of some chemical reactions.

Biological effects of ultrasounds

Biological effects depend on the characteristics of ultrasound weve: intensity, frequency, doses.

Ultrasounds were divided conventionally into three groups from the biological point of view: those of small intensity ($0.5–1.5$ W/cm^2), of middle intensity ($1.5–3$ W/cm^2) and great intensity ($3 – 10$ W/cm^2).

In small intensities the tissues do not suffer morphological

changes producing only functional modifications. A cytoplasm in rent appears and stimulates physiological processes.

In middle intensities cytoplasm currents become strong and stop the normal development of cell mechanisms. The permeability of membrane is modified but the effects remain reversible.

In great intensities the irreversible structural modifications take place. The embryo tissues and, generally, internal cells are more lensible to the irradiation by ultrasounds than mature cells.

For a living biosystem cavitation is very dangerous due to the high temperature and pressures following a shock wave of implosion capable of breaking the structures or the macromolecules from the zone. At the molecular level the oxidation and polymerization as well as the break of biological molecules take place, free radicals appear, the decrease of γ globulin is observed, etc. At the cell level haemolysis (in dilute solutions), breaking and sprains of some cilium are produced, the permeability of cell membrane is modified, the volume of mithochondria is increased; in the hepatic cell the number of lysozymes increases or decreases in dependence on radiation parameters, etc.

At the organism level ultrasounds have a lethal effect on larvae of mosquitoes, fish, frogs. Mice die at (1-2 W/cm^2) and frequency of 20 kHz. In the superior animals muscle contraction increases due to the formation of exciting products as a result of microbreak. Waves P, Q, T of electrocardiogram are modified as a function of ultrasound intensity and extrasystoles and bradycardia are produced. At the law frequencies (under than 1000 kHz) the ultrasounds are little absorbed by air, therefore they propagate at the long distances and can have effects on the staff working in vicinity of ultrasound installations. At the frequencies of (16-25) kHz and intensities over 100 Db a series of neurovegetative manifestations were observed such as perturbation of suprarenal gland function, thermoregulation processes, psychical disorders (hallucinations), equilibrium disorders, bulimia.

Importance of ultrasound effects for medical practice

In medical – biological sphere ultrasound is applied mainly in two directions – in diagnostics and action.

Methods of location belong to the first direction. Among them are: echoencephalography – revealing of tumors and cerebral edema; ultrasonic cardiography - determination of heart size by dynamics.

The Doppler effect in the case of ultrasound is used in the study of the moving character of the heart valves, in the measurement of blood flow, in the visualizion of different internal organs.

Ultrasonic therapy belongs to the second direction. Ultrasound is used in therapeutical purposes with the frequency of 800 kHz and the intensity of about 1 W/m^2.

The possibilities to use ultrasound in the laboratory and pharmaceutical industry are manifold.

It can reach the condition of true colloidal solutions by the ultrasounding of emulsions and medical suspense. The inverse effect of destroying the disperse system can occur. Thus, the ultrasounding of aerosol leads to the increase of particles followed by sedimentation.

Ultrasound is also used for rapid, fine and uniform crystallization of extrasaturated solutions, in drying of thermosensitive materials, extraction of active components from vegetable products.

Ultrasound breaking of different medical substances is the basis of functioning of modern inhalators.

Ultrasound breaking action on the microorganisms is used in **sterilization.**

In medicine the Doppler effect is used. It is caused by the fact that the frequency of ultrasound waves registered by a detector differs from the frequency of waves emitted by the source when the source and detector (or at least one of them) are removed in respect to the propagation medium. This effect is characteristic of

waves of any nature, including ultrasound waves and it is named the Doppler affect

The frequency variation of the Doppler effect V_D is called also the Doppler variation. The speed of ultrasounds (V_U) is greater than I he speed of objects (V_0) in medical applications and for these cases V_D is given by the equation:

$$V_D = V_U$$

The Doppler effect is used for determination of blood circulation speed: the variation of reflected ultrasound frequency of red cells being in moving is registered. The speed of valve moving and that of heart walls are determined (the Doppler echography); the variation of ultrasound frequency reflected by the heart walls during the pulsation is determined.

The ultrasound breaking of different medicament substances is the basis of functioning of modern inhalators. For example, let us get acquainted with the construction and function principles of ultrasound inhalator of IU – 001 type used for the prophylaxis and treatment of the respiratory tract by aerosols with preparations dissolved in water at home and at medical institutions.

External view of inhalator

Inhalator is made of two connected parts: the electronic block 1 and the pulverization room in which the converter 2 is inserted (fig. 5.3)

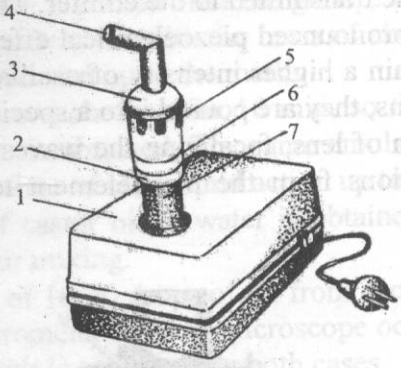


Fig. 5.3

The pulverization room has a form of glass closed over by the lid 3 and with the respiration tube 4. On the bottom of the glass the electroacoustic converter is inserted. Three cuts of different breadth are made on the lateral surface of the lid. The diaphragm 5 is situated at the lateral part of the glass for the regulation of aerosol pulverization productivity by the linking of a respective cut with the diaphragm. The control marks 6 are on the lower part of the lateral surface of the glass inside the liquid level (preparation for treatment) must be in order to assure the normal pulverization.

In the lower part of the pulverization room there is a plug for electrical connection with the electronic block. The electronic block contains a generator of high frequency producing the voltage of frequency 3.64 MHz indispensable for the excitation of pulverization room converter.

The inhalator is supplied with alternative current of frequency 50 Hz and voltage 220 V from the electrical net.

Description of installation

The ultrasonic installation presents a ultrasound generator with the piezoelectrical emitter. The ultrasonic oscillations from

the generator are transmitted to the emitter. The piezoceramics element with the pronounced piezoelectrical effect is used as emitter. In order to obtain a higher intensity of oscillations in the preparation of emulsions, they are poured into a special glass with the bottom in the form of lens, focalizing the waves. The medium transmitting oscillations from the piezoelement to the glass is water (fig. 5.4).

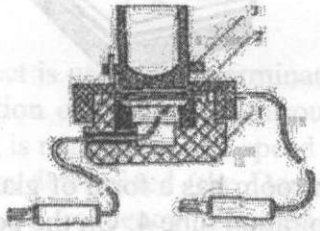


Fig.5.4

Equipment and materials

The ultrasound generator, emitter, optical microscope, the micrometer scale of the ocular, castor oil, distilled water, glass, mechanical mixing.

Work procedure

The emitter 1 is connected up into the sockets of ultrasound generator.

The water is poured upon the piezoelement 2.

The ultrasound generator is connected up into the electrical net and the knob "frequency regulator" is rotated till the "filling" of the water on the piezoelement is obtained.

The glass 3 in the form of lens is placed on the emitter, so that between the emitter and glass bubbles of air are not created.

The distilled water is poured into the glass 2 mm lower than the mark. The active substance with the mass of about 0.1 g is added. The castor oil is poured up to the mark.

The generator is switched on for 10 min, so the emulsion of castor oil in the water is obtained. The same emulsion is obtained in the increase of the time of ultrasound action up to 30 min.

The emulsion of castor oil in water is obtained by means of mechanical shaking or mixing.

The dimensions of 10-20 drops of oil from each emulsion are measured by the micrometer scale of microscope ocular

The obtained result is compared in both cases.

The results of measurements are written in table 5.1

Table 5.1

The method of emulsion preparation	Duration of preparation	Diameters of 10-20 drops, mm	D_{aver}, mm
Ultrasound	10		
Ultrasound	20		
Mechanical	30		

Questions and tasks for self-control

1. Explain the physical phenomena that are on the basis of production and receiving of ultrasounds.
2. What effects can occur in the interaction of ultrasound with the substance?
3. What are the principal medical – biological applications of ultrasound? Explain the technique of each application on the basis of ultrasound properties and ultrasound effects.
4. What physical phenomenon (characteristic of all wave processes) explains the "paradox" of ultrasound transparency through the metallic plates of the thickness $\lambda/2$ and $\lambda/4$.

6. DETERMINATION OF ION MOBILITY BY THE ELECTROPHORESIS METHOD

Purposes

- study of phenomena which are on the base of electrophoresis method;
- familiarization with application of electrophoresis method in biology and medicine;
- study of electrophoresis equipment construction; electrophoresis separation of protein fractions of blood serum;
- electrophoresis separation of inorganic ions (Cu^{2+} and Fe^{3+}); visualization of colored ions moving into the electrical field.

Theoretical notions

Electrophoresis is based on the electrokinetical phenomena, by which the movement processes of electrical charged particles, mespectively of their nature (ions, colloidal particles, bubbles of gas suspended in liquid, etc.) are understood.

The migration of any colloidal particles carrying charge in the external electrical field is called electrophoresis. Force F_e is exercised on a particle of mass **m** and charge $q = Z \cdot e$ in an electrical homogenous field by intensity E, given by the equation:

$$F_e = Z \cdot e \cdot E \qquad (1)$$

The direction of this force corresponds to the direction of the electrical field, when the particle has the positive charge and, in the opposite case, the direction of the field and the force are of the Contrary sense. As a result, the particle is accelerated according to the fundamental law of dynamics.

$$F_e = \frac{d(mV)}{dt} \qquad (2)$$

The friction force from the environment influencing the particle increases concomitantly. The value of friction force is determined by the Stokes law, that supposes the spherical form of the particle:

$$F_s = 6\pi\eta r V \qquad (3)$$

Here **r** is the radius of the particle, v the speed of the particle, η viscosity medium coefficient in which the particle moves, in a while, the friction coefficient is compensated by electrical force. From this moment the accelerated movement is conversed into the uniform movement. In this case the following equation is true:

$$F_e = F_s \quad or \quad Z \cdot e \cdot E = 6\pi\eta r V \qquad (4)$$

From this:

$$V = \frac{Z \cdot e \cdot E}{6\pi\eta r} = \frac{Z \cdot e}{6\pi\eta r} \cdot E \qquad (5)$$

It should be noted tha $\frac{Z \cdot e}{6\pi\eta r} = M$. Then from the equation (5) we obtain:

$$V = M \cdot E \qquad (6)$$

The value **M** is called the mobility of the charged electrical particle.

Concluding from the equation (6), we can say that the mobility of particle is a value numerically equal to the speed of its uniform motion under the influence of the electrical field by the intensity E=1 v/m .

It results from (5) that the mobility of the charged particle depends on the nature of the particle (Z e and r), the nature of medium and its temperature (because η depends on temperature). The unit measurement is:

$$[M]_{I.S} = m^2 \cdot s^{-1} \cdot v^{-1} \qquad [M]_{\text{Pract}} = cm^2 \cdot s^{-1} \cdot v^{-1}$$

The importance of electrophoresis method for medical practice

Electrophoresis is frequently used in medical practice for examination of biological liquids as to proteins – an important factor for the diagnostics of different diseases. Electrophoresis is a major method for separation of different fractions of serum proteins based upon physical-chemical properties of blood proteins, it being a qualitative and quantitative determination.

The principle of method

The electrophoresis methods use the equipment in which the migration is made in the tube of liquid, and equipment in which the migration is made in a porous body (filter paper, synthetic resin, gelose etc.) We shall use the migration on the filter paper.

The electrophoresis method is based upon the principle of charged particles migrating into the electrical field that due to theirs sizes and their charges, have different mobility and are separated among them in space, thus, obtaining the electrophoregram.

Description of electrophoreses equipment

The electrophoresis device with which we work in our laboratory is made of electrophoresis cavity and a continuum current supply source. The electrophoresis cavity (fig 6.1) is made of plastic material and contains two organic glass tubs 1 for the tampon solu-

tion. Each tub has two compartments: one with the penetrated electrode of Pt **5**, and the other with the penetrated end of paper band 2. These two compartments are connected with each other by diaphragms. The lid of organic glass 4 covers the electrophoresis cavity.

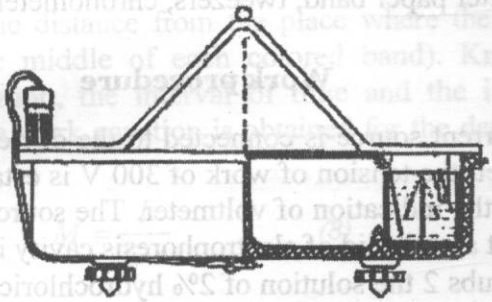


Fig.6.1

The supply source (fig.6.2) represents a changeable redresser (0-500 V, 0-40 mA) endowed with the tools of sensible measurements (voltmeter, milliammeter).

At the back part of the source there are sockets with the respective inscriptions "+" and "-" for the connection of electrophoresis cavity.

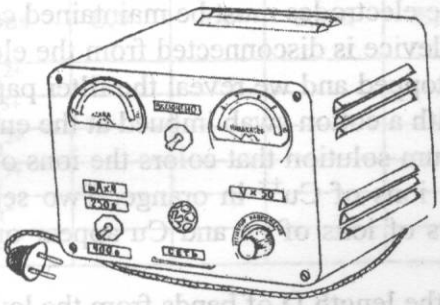


Fig. 6.2

Devices and materials

The electrophoresis cavity, the continuum current supply source, filter paper bands 2/26 cm and 2/20 mm, the researched product: a mixture of $FeCl_2$ and $CuCl_2$, micropipette of 0,01 ml, glass stick, cotton, tampon solution, colored solution (potassium ferrocyanide), niter paper band, tweezers, chronometer, scaled rule.

Work procedure

The current source is connected to the device and then to the electrical net, the tension of work of 300 V is established in accordance with the indication of voltmeter. The source is disconnected from the net and the lid of electrophoresis cavity is removed.

In the tubs 2 the solution of 2% hydrochloric acid is poured in the distilled water. (The tampon solution). Two bands of filter paper ire soaked in the tampon solution (2/26 cm); the combination of lhem is uniformed by placing between two pieces of usual paper. These bands arc placed horizontally in the electrophoresis cavity with the free ends in the tubs with the tampon solution. The electrophoreses is covered by the lid 4 in order to maintain the instant humidity inside.

The current source is connected to the electrical net, fixing concomitantly the time on the chronometer. During the experience the tension at the electrodes must be maintained constantly. Within 20–25 min the device is disconnected from the electrical net, the й hronometer is stopped and we reveal the filter paper bands using a stick of glass with a cotton swab imbued at the end by 5% ferrocyanide of potassium solution that colors the ions of ferrum Fe^{+++} in to bluc, but the ions of Cu^{++} in orange. Two separated bands of respective colors of ions of Fe and Cu appear on the filter paper hands.

Measuring the length D of bands from the level of tampon solutions in the tub of electrophoresis cavity till the level of the other

tub and knowing the value of the applied tension at the electrodes V, the intensity of electric field is determined by the equation:

$$E = \frac{V}{d} \qquad (7)$$

Subsequently, the average distance l is measured, moved by the respective ions (the distance from the place where the product is applied up to the middle of each colored band). Knowing the distance of migration, the interval of time and the intensity of electrical field, the work equation is obtained for the determination of ion mobility:

$$M = \frac{l}{E \cdot t} \qquad (8)$$

The measure unit is:

$$[M] = \frac{cm^2}{V \cdot s}$$

The experiment is performed three times for different values of tension and intervals of time.

Table 6.1 is filled in with the experimentally obtained data.

Table 6.1

Nr of experiment	Ions	U,V	t,s	d, cm	E, V/cm	l, cm
1	Cu^{2+}					
	Fe^{3+}					
2	Cu^{2+}					
	Fe^{3+}					
3	Cu^{2+}					
	Fe^{3+}					

After the mathematical processing of the experimental data, the value of ion mobility at the given conditions is expressed as:

$$U = \bar{U} \pm \Delta U \qquad (10)$$

Where: U is the average value; ΔU is the value of absolute error.

Questions and tasks for self-control

1. What factors determine the migration speed of a particle with the certain electrical charge into an electrical field of known intensity?
2. Write the mathematical equation that includes all these factors.
3. How is the mobility of charged particle into the electrical field defined and what is the measure unit in the IS?
4. Indicate the medical-biological fields where the electrophoresis method is used successfully.

7. ELECTRICAL METHODS OF TEMPERATURE MEASURING

Purposes:

1. presentation of theoretical notions about thermoelectric phenomena;
2. familiarization with the application of thermoelectricity in medical practice;
3. standardization of thermocouple and thermoresistor;
4. performing of the experiment with the demonstration of the Peltier effect.

Theoretical notions

In the direct contact between two heterogeneous metals free electrons pass through one metal to another due to the thermal movement. The diffusion fluxes from each metal at first are different due to the different concentrations of free electrons. Fig. **7.1, a** shows the contact between metals 1 and 2 with the concentrations of free electrons r_1 and n_2 respectively ($n_1 > n_2$). In this case the contact potential difference appears with the direction indicated in Fig. **7.1, a**. The potential difference leads to the equalization of the diffusion fluxes and the establishment of dynamical equilibrium. The potential difference value can be calculated resulting from the general condition of equilibrium - the equality of electrochemical potentials of metals coming into contact.

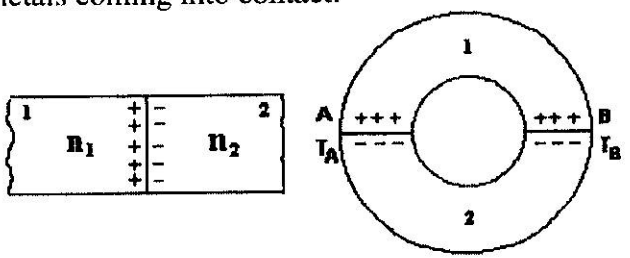


Fig. 7.1

$$RT \ln n_2 + z \cdot F \cdot \varphi_2 = RT \ln n_1 + z \cdot F \cdot \varphi_1$$

or

$$RT \ln \frac{n_2}{n_1} + z \cdot F \cdot (\varphi_2 - \varphi_1) = 0 \qquad (1)$$

As $z = -1$, we have got:

$$U = \varphi_2 - \varphi_1 = \frac{RT}{F} \ln \frac{n_1}{n_2} \qquad (2)$$

It results from (1) that the potential difference depends on the concentration difference of electrons and also on the temperature of contact. Let us examine a closed circuit made of metals 1 and 2 (fig. 7.1, b).

For the same temperature of contacts A and B the contact potential differences U_A and U_B are opposite – the current in the circuit is equal to zero.

At different temperatures (to be more exact, we assume that $T_A > T_B$) we obtain from the relation (2):

$$U_A = \frac{RT_A}{F} \ln \frac{n_1}{n_2} \qquad (3)$$

$$U_B = \frac{RT_B}{F} \ln \frac{n_1}{n_2} \qquad (4)$$

It is $U_A \neq U_B$ that evidently leads to the apparition of thermal dectromotion voltage (T. E. V):

$$\varepsilon_1 = U_A + U_B = \frac{R}{F} \ln \frac{n_1}{n_2} \cdot (T_A - T_B) \qquad (5)$$

Marking $\frac{R}{F}\ln\frac{n_1}{n_2}$ by β, we obtain:

$$\varepsilon_1 = \beta(T_A - T_B) \qquad (6)$$

For concrete metals β is the constant numerically equal to (T. E. V) that appears in the circuit when the temperature difference of contacts is equal to 1 K. For the majority of metals it is relatively small and is expressed in mV/K:

Copper – Ferrum	0.01 mV/K
Constantan – Copper	0.041 mV/K
Constantan – Ferrum	0.053 mV/K

(T. E. V) can be measured by galvanometer (microvoltmeter) connected in the circuit of thermocouple.

A direct proportional dependence exists between the number of divisions at which the needle of galvanometer II and (T. E. V)

$$N = \gamma \cdot \varepsilon_1 \qquad (7)$$

Substituting et from (6) in the expression (7), we obtain:

$$N = \beta \cdot \gamma \cdot (T_A - T_B) = \alpha \cdot (T_A - T_B) \qquad (8)$$

where $\alpha = \frac{N}{(T_A - T_B)}$ presents the deviation of the needle at the temperatures difference equal to 1K and is named the **sensitivity of installation.**

As the temperature difference is included in (8), then we can use °C. Maintaining constantly the temperature cool contact (T_B=const), the temperature of warm contact will become the function of galvanometer needle deviation:

$$T_A = \frac{N}{\alpha} + T_B \qquad (9)$$

Thus, the standardization of thermocouple is possible to be used as a thermometer.

Another electrical method of thermometry is based on the property of some metals and semiconductors to change essentially the specific resistance depending on temperature. The standardization of thermoresistor as a thermometer is performed using the electrical scheme represented in fig. 7.2. Although the resistance R and R_e are linked consecutively, then:

$$\frac{R}{R_e} = \frac{U}{U_e} \quad \text{or} \quad R = \frac{R_e \cdot U}{U_e} \qquad (10)$$

Where U is the voltage decrease on the thermoresistor, but U_e – the voltage decrease on the standard resistance.

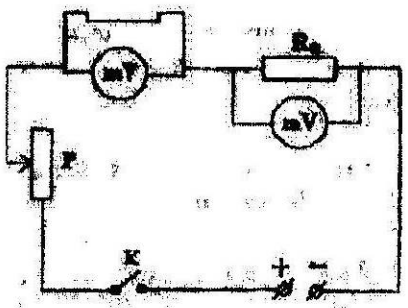


Fig. 7.2

R – the value of thermoresistance, R_e – the value of standard resistance

The importance for medical practice

Thermoelectricity has three main practical applications: 1) for production current generators, in which the thermal energy is transformed into electrical energy; 2) The standard thermocouple is used as a thermometer.

Such a thermometer is used in medicine and biology both for

the determination of animals' and vegetal tissues temperature. Having small dimensions and thermal capacity, it allows the determination of temperature in small spaces with the sufficient precision and very rapidly. It can measure the skin temperature as well as the body temperature at a distance.

3) For the measuring of both visible and ultraviolet and infrared radiation power.

The Peltier phenomenon presents a great importance for medical practice. It is used for the construction of different devices for cooling with different dimensions.

The minicooler can be mentioned for example. It is used in ophtalmological surgery for extraction of the defected crystalline from the eye.

A cooler in the form of a needle is introduced in the crystalline. The crystalline sticks to it when the needle is cooled on the basis of the Peltier phenomenon.

Standardization of thermocouple and thermoresistor as a thermometer

a) Standardization of thermocouple

1) In order to perform measurements the installation is mounted according to the scheme shown in fig. 7.3:

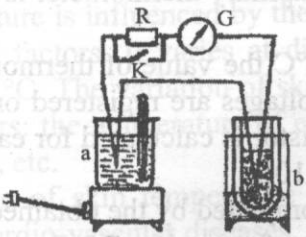


Fig. 7.3

1) Warm welding of thermocouple is inserted in the vase a, containing water with ice and situated on the electrical range.

A cool welding is inserted in the Dewar vase filled with the same mixture of ice and water. The indications of galvanometer correspond to the null position of the needle. Introducing the range in the plug, the temperature of welding is from 5° to 5° by the thermometer with Hg, as well as the respective indications of galvanometer. During the measurements the switcher K is closed. The measurements last till the temperature of boiling water is achieved.

2) The experimentally obtained values are written in table 7. $T_B = 0$ *(const).*

According to the data from table 7.1, the scheme of dependence between T_A and N is drawn.

Using the middle portion of the scheme, the sensitivity value of the working installation is calculated.

It is recommended to determine the skin temperature using the installation and standardization scheme.

Standardization of thermoresistor

The standardization of thermoresistor is made by the method used in the standardization of thermocouple.

1) The standard – resistance is maintained at the constant temperature (0°C), but the thermoresistor is inserted into the vase posed to heating. A capillary thermometer is situated in the vicinitz of the thermometer.

2) After each 10°C the value of thermoresistor temperature is registered, and the voltages are registered on each resistance. The value R of thermoresistor is calculated for each measurement acording to the equation (10).

3) Table 7.2 is completed by the obtained experimental data.

4) Using the data from table 7.2 the scheme of dependence R=F(t) and the graphic U=F(t) are drawn on the same millimeter paper. In this case it is recommended to determine the skin temperature using the installation and the second scheme.

Table 7. 1

Nrof experiments	1	2	...	19	20
T_A, ^0C					
N					

Table 7.2

Nr of experiments	t, (°C)	R_e,(Ω)	U_e,(V)	U,(V)	R,(Ω)
1					
2					
3					
4					
5					
6					

Determination of skin temperature by the thermocouple

The skin temperature is influenced by the internal temperature and different external factors. It varies at different points of the body from 23.2 to 35 °C. The variation of skin temperature is due to the following factors: the temperature of environment, the kind of food and work, age, etc.

The determination of skin temperature is a procedure used currently in various cardio-vascular diseases, giving the diagnostic indications, prognoses, and the efficiency of different used therapeutic maneuvers.

As the skin temperature is dependent on the blood peripheral circulation, the action of vasodilator substances, for example,

tolazoline, histamine, methyl salicylate, etc., all these factors will influence the skin temperature.

The present paper will focus on the influence of one of these substances used as an ointment in the temperature.

The installation consists of:

- the measuring instrument, galvanometer "Multiflex", the scale with 100 divisions;
- the thermocouple with cool welding is introduced into a recipient with water at the room temperature; the warm welding has the form of rod made of plastic material (fig. 7.5).

Work process

- The switch button of the measuring board of the tool is verified whether it is at the position "0".
- The measuring tool is connected up to the plug (~ 220 V); a light spot will appear on the instrument scale at the division point *"0"*.
- These two linking wires of thermocouple are connected up to the sockets of the tool observing the polarity.
- The tool switcher is put in the position 1:1, the light spot will move to the right (depending on laboratory temperature).
- On the arm tegument of the left hand a thin layer of the solution
- of vasodilator substance is applied, the initial temperature of which was determined.
- The mentioned region is frictioned well for about 12 minutes.
- The variation of temperature at the mentioned place is followed every 30 seconds;
- The read values are written in table 7.3 and according to them the respective chart is drawn.

• **Warning: In case when the scale 1:10 was used, it is not necessary to omit the preliminary multiplying of n to 10.**

The variation of skin temperature under the influence of one vasodilator (arm, left hand).

Table 7.3

	Time (s)	0	30	60	90...
	Initial temperature, t_i				
n					
t					

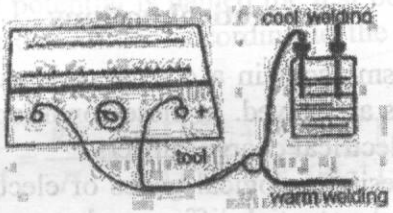


Fig.7.5

Questions and tasks for self – control

1. What is the cause of apparition of contact potential difference between two heterogeneous metals and under what formula its value is determined?
2. What is the thermocouple? Write the formula for the determination of thermal electromotion voltage and explain the symbols.
3. What are the main applications of thermoelectricity and Peltier effect in medical practice?

8. REGISTERRATION OF ACTION AND BREAKING POTENTIALS IN FROG CORD

Purposes:

- presentation of theoretical aspects regarding bioelectrogenesis;
- biphase wave;
- monophase wave and the measurement of breaking potential in the frog cord;
- familiarization with the use of registration of biopotentials in medical practice

Theoretical notions

Living organisms contain a considerable quantity of water in which mineral salts are solved. In these conditions biological media are rich in ions - electrical charge carriers.

Bioelectrogenesis (biological genes of electrical potentials) is due to the electrical potential difference between the internal face and the external one of the cell membrane. These potential differences are of 50-100 mV, the potential is negative in the internal part in respect to the external part of the membrane cell.

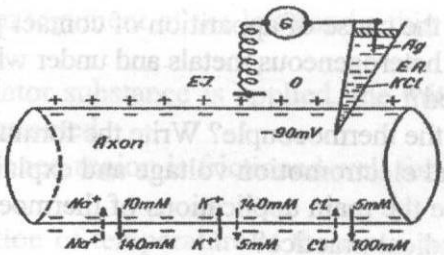


Fig.8.1

The scheme of membrane potential measurement in the gigant axon $E \cdot A$ – active electrode, EI – indifferent electrode – passive transportation, – active transportation.

The transmembrane potential is caused by inter- and extracellular concentration differences of ions Na^+, K^+, and Cl^-. These concentration differences are due to the activity of potassium - sodium pump activity and in a less degree of the Donnan equilibrium.

The voltage between two faces of the cell membrane (membrane potential) was measured by microelectrode technique. An electrode is placed at the external part of the membrane (as a rule the axon of the nerve) and a thin edge of glass microelectrode with the diameter less than 1 μm is introduced in the internal part. The microelectrode is filled with a saturated solution of KC1 in contact with the Ag electrode on which AgCl is formed (fig. 8.1).

The measured potential is named the rest potential. The value of this potential is determined according to the given equation of Goldman:

$$\Delta\varphi = V_i - V_e = \frac{RT}{F} \ln \frac{P_K[K^+]_i + P_{Na}[Na^+]_i + P_{Cl}[Cl^-]_e}{P_K[K^+]_e + P_{Na}[Na^+]_e + P_{Cl}[Cl^-]_i}$$

Where C is the permeability of membrane for the respective ions; [ ] represents the concentration of the respective ions; the indices i and e refer to the internal and external parts of the cell membrane.

It should be noted that the numerical values of the rest potential vary with the cell type, the species, etc.

A common property for all types of living cells is excitability followed by a series of phenomena, among them there is also electrical behavior. The latter is, in fact, the expression of disequilibrium of the rest of electrical phenomena and the apparition of action potential. According to the hypotheses by Hodgkin and Huxley the permeability of K^+ and Na^+ is modified essentially on the excitation.

At rest we obtain:

$$\frac{P_K}{P_{Na}} = \frac{1.0}{0.04}$$

On the excitation:

$$\frac{P_K}{P_{Na}} = \frac{1.0}{30}$$

As on the excitation moment $P_{Na} >> P_K$ and $P_{Na} >> P_A$ (A - the anion of NaCl salt) the following condition can be considered $P_K \rightarrow 0$ and $P_A \rightarrow 0$. Making this supposition the following pression is obtained from the Goldman equation:

$$\Delta\varphi_i = \frac{RT}{F} \ln \frac{[Na^+]_e}{[Na^+]_i}$$

This potential was named the Na potential and later the Inversion potential. $\Delta\varphi_i$ possesses different values for different lis, but it always has some tens of mV. The action potential (fig. 8.2) is:

$$\Delta\varphi_a + |\varphi_r| = |\varphi_i|$$

Where φ_r is the rest potential.

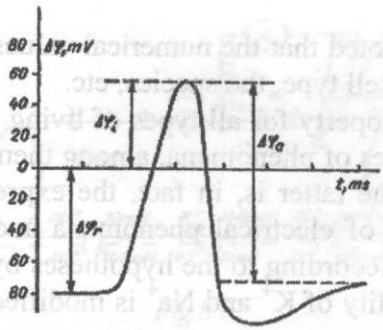


Fig. 8.2. **Action potential recorded by microelectrode technique on the giant axon.**

The membrane is polarized inversely at the moment of action potential existence: the excited portion becomes negative in the external part and positive in the internal part. This state is called depolarization.

After a while (milliseconds) the state of membrane is completely restored. This phenomenon is named repolarization.

The potential differences can be picked from an unbroken organ during its activity. In this case the electrical phenomenon represents a sum of electrical potential modifications from each cell. Studying experimentally the muscle the following facts are established:

– At rest the organ is polarized and no difference exists between points A and B, on which the picking electrodes are applied.

The galvanometer needle indicates zero and the horizontal line named the isopotential line is presented graphically (fig.8.3 - I).

The applied excitation in the point A produces a disequilibrium between the depolarized point A (electronegative) and point B still polarized (electropositive). An oriented vector from «-» to can represent this disequilibrium "+". An electrical current passes through the external circuit that causes the deviation of galvanometer needle in one sense and an abrupt ascendant line is recorded graphically (fig. 8.3 – II).

There is no potential difference between the points A and B when the excitation involves the whole muscle which is entirely depolarized and the galvanometer needle comes to zero, but the wave returns to the isopotential line where it is maintained for a short time 9 (fig. 8.3 – III).

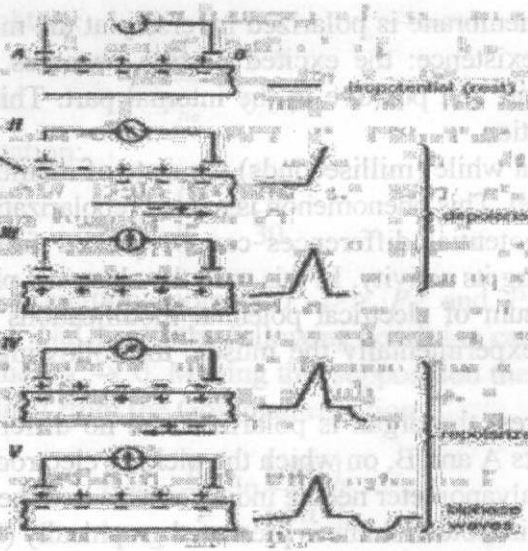


Fig. 8.3

Then the repolarization phase follows. It begins at the point A and propagates in the same sense with excitation. During repolarization another electrical disequilibrium is produced between still depolarized zone (electronegative) and the repolarized zone (electropositive). The galvanometer needle will be deviated to the opposite sense from depolarization. The opposite direction will also have the electrical vector. A wave is recorded graphically. It comes under the isoelectrical line but has a less ample development (fig. 8.3-IV).

When the entire fiber is repolarized the potential difference between two electrodes is absent, the galvanometer needle comes to zero, but the graphic to the isoelectrical line (fig. 8.3 V).

From the succession of these phenomena a biphase potential is registered (biphase wave).

The measurement of the created potential difference of the cell and tissue activity can be performed by means of:

– the direct methods when electrodes of picking are placed directly on the studied organ;

– the indirect methods by remote placing of electrodes, for example, on the body surface;

– the single polar picking, one of electrodes that is active and with the small contact surface is placed on the studied organ but another electrode named indifferent and with a great contact surface is placed in the same medium.

– the bipolar picking when both electrodes are situated on the studied organ, so that both electrodes are active;

– cell potentials are picked by the microelectrode method but those tissual – with the special nonpolarized electrodes. We shall use the electrode of the d'Arsonval type in this paper.

The paper has two processes

a) biphase wave and b) the measurement of the frog cord action potential by electrocardiograph using the bipolar method.

The principle of the method

The picked potential differences between the placed electrodes on the base and the edge of the cord in activity constitute electrocardiogram (ECG).

Its production mechanism is that of biphase wave. The curve of potential variations has a complex aspect due to anatomical, physiological and histological peculiarities of the miocardium (fig.8.4).

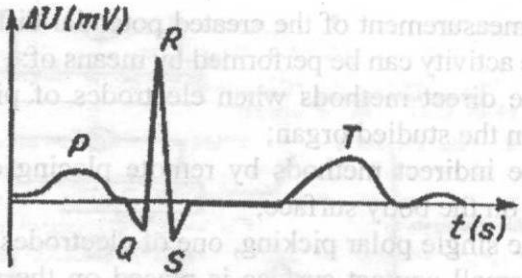


Fig. 8.4

The signification of electrocardiogram deviations is the following: the wave C corresponds to the depolarization of auricles; The complexity QRS - to the depolarization of ventricles but the wave T - to the repolarization of the entire myocardium.

We have to note that the aspect of ECG performed for different experimental animals is similar to that taken in people.

Description of working device

The device used to take an electrocardiogram on frog cord is named "salute" and it is produced in Russia. This portative device is used in medical practice. In fact, the apparatus amplifies the potential differences picked from the surface of the cord with two nonpolarizing electrodes of the d'Arsonval type and records them. The amplification of biocurrents is made by means of an electronic system. The amplified currents pass through solenoid, yield a variable magnetic field leading to the registering nib. This nib leaves imprints on the thermosensitive paper by heaters. The paper is involved in movement by an electrical engine with the known speed.

The block scheme is represented in fig. 8.5

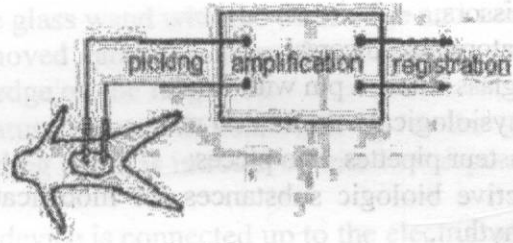


Fig. 8.5

The d'Arsonval electrodes are nonpolarizing electrodes consisting of a glass tube mounted on one of the extremities (fig. 8.6). As the zinc wand is sunk into the saturated solution of zinc sulfide on passing the current through the electrodes the ions of zinc

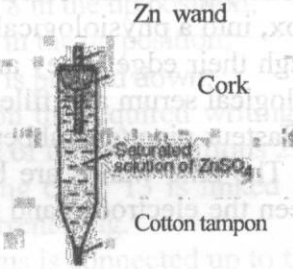


Fig.8.6

are discharged on the wand. So, the apparition of electromotive force of polarization is avoided.

The necessary materials

– the apparatus "salute" with wires for connection.
– thermosensitive paper for recording;
– special pipettes for preparation of electrodes;
– a plate for the dissection of a frog;

– scissors;
- anatomical pincers;
– a glass wand, a pin with head;
– physiological serum of 20 ml;
– pasteur pipettes, two pieces;
– active biologic substances for modification of the cardiac rhythm.

Work process

The registration of action potential on the frog cord is performed in the following work periods.

1) The preparation of nonpolarized electrodes and their inting on the supports. The pipettes for electrodes are set on the worktable into a Petry box, into a physiological serum with the coton tampon passed through their edge. They are taken from there, emptied from the physiological serum and filled with the saturated solution of $ZnSO_4$ by a Pasteur pipette. Rubber corks with zincids obturate each electrode. The zinc wands are bound to two wires that make binding between the electrodes and the device "Salute" by a coaxial cable.

The cotton filter of electrodes is washed in the physiological serum. Those two electrodes are adapted to the respective supports.

2) A needle breaks the bulb and the spine of the frog. Then the frog is placed on the dissection plate and the pins fix the limbs of the frog.

Anatomical pincers catch the teguments from the stern, and the isors make a buttonhole of 1 cm under the xiphoid appendices.

The incision of the skin soft parts and ribs to each humeral articulation is continued, thus performing a section in V – shape.

The rib plastron is lifted sectioning carefully the pericardia lterna reins. The cord appears as a pulsate formation in the center of the wound.

Using only the glass wand with the bond edge and scissors the pericardium is removed carefully. Then the electrodes are placed carefully with the edge on the frog cord.

3) The apparatus "salute" is connected up to the grounded plug and the recording paper is introduced into the respective support.

4) Before the device is connected up to the electric net the leading devices must be fixed in the initial position (fig. 8.6):

- the switcher 3 of derivations - in the 0 position;
- the switcher 1 of the net - in the position opposite to the one marked with a red point;
- the potentiometer 9 for the establishment of amplification level is rotated till a full stop.
- the switcher 8 in the up position;
- the button 7 in the up position;
- the button 5 is pressed down;
- depending on the required writing speed the switcher 10 is fixed in one of the two positions 25 or 50.

5) The recording cable 13 is linked and the apparatus is connected up to the current plug.

6) The apparatus is connected up to the electrical net, the switcher 1 is put in the "lit" position.

The lighting of the bulb indicates the setting on of the apparatus.

7) After 5-6 seconds the registering nib is released from the fixed state by pressing the button 5 (passage to the upper position).

8) The button 7 is pressed setting on in function the mechanism for the record paper moving. The level of warming of the nib is adjusted by the rotation of the knob 2 and then the thickness of recording line is (0.3 – 1 mm).

9) The standardization of electrocardiograph is performed establishing the sensitivity of 10 mm/ mV. In order to perform this the following conditions have to be taken into consideration:

– The mechanism for the paper band moving is coupled by pressing the button 7;

– The regulator 9 is rotated gently for the exchange of sensitivity, the button 4 is pressed and released successively several times till a nib deviation equal to 10 mm is obtained (fig.8.7) because the applied voltage for the standardization is about 1 mV;

– The mechanism for the registering paper moving is disconnected.

10) The switcher 3 of derivations is fixed in the position 1, which the electrodes of biopotentials picking are connected up to.

– The mechanism of paper development is set on and the ne]cessary number of electrocardiogram cycles is registered stopping then the development.

11) The experience is repeated by dropping previously a medicament substance on the cord that leads to the variation of the cordiac rhythm.

Note:

a) The paper with the recorded curves is not taken out in order lo perform properly point **b** of the paper.

b) The monophase wave and the measuring of breaking potential on the frog cord by the electrocardiograph.

Principle of the method

The created potential differences are picked between the broken and unbroken portions of the myocardium, both at rest and in activity.

The obtained graphical aspect and the explanation of product mechanism are of the breaking potential and of monophase wave.

The device, necessary materials, the conditions and the working technique are the ones described in the point **a**.

The electrode from the edge of the cord is lifted. The latter is broken by the pincers, thus, forming a lesion.

The lesion electrode is replaced and several monophase waves tfe written as a continuation of the obtained biphase waves on the unaffected cord.The obtained graphics for the standard pulse, the action potential on the uninjured cord (in the absence and presence of a medicament substance), the rest potential and the monophase wave will be placed as in fig. 8.7.

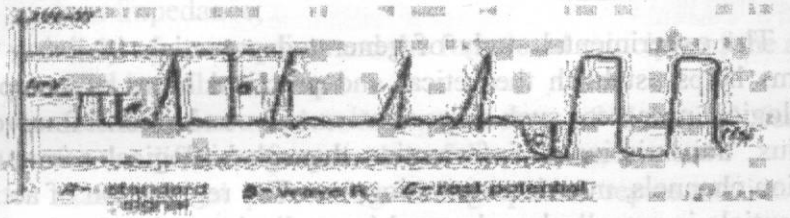


Fig. 8.7

Processing of experimental data

The amplitude of maximal positive deflection is measured (the wave R) by a graded ruler that represents the action potential of the uninjured cord.

The falling under the isoelectric line is also measured in millimeters after performing the lesion in the case when the cord is at rest (diastole).

Both the amplitude and the duration of those two phases of depolarization and repolarization are determined at the monophase wave. Using the result of standardization and the value of amplitude the respective potential value is determined. The obtained experimental values are written in the table below:

The wave or the signal	Amplitude (mm)	The potential value (mV)	Duration(s)
Standard signal	10	1	----------
The wave R			
Rest			

Continu

Depolarization wave			
Monophase repolarization			

The importance for medical practice

The experimental study of generated potentials in living systems helps establish theoretical and practical bases of complex biological processes such as generation and conduction of nervous influx, transmission of information through biological communication channels, muscle contraction, etc. The registration of action potentials is actually largely used in medical practice for the function examination of some vital organs. It is performed as electrocardiogram (ECG), electroencephalogram (EEG), electroretinagram (ERG), electromiogram (EMG) and others.

The study of biopotentials is of a great importance for elaboration and application of an ample variety of electrical stimulators both for restoring and maintaining functional activity of different lystems of muscles and organs for a long time.

Questions and tasks for self- control

1. Write the Goldman equation for the determination of the rest membrane potential and explain the symbols.
2. What is the microelectrode technique? Show graphically the result of recording of action potentials by the microelectrode technique.
3. Draw the simplified scheme of recording and the form of biphase wave.
4. Kxplain the preparation procedure of the frog technique and registering of action and lesion potentials
5. What are the fields of application of the biopotential electro-graphic method in medicine?

9. DISPERSION OF ELECTRICAL IMPEDANCE OF BIOLOGICAL TISSUES

Purposes:

- presentation of theoretical notions regarding alternative current parameters and the peculiarities of biological tissues impedance;
- study of construction and the functional principle of the applied installation;
- determination of electrical parameters of biological tissue for the different frequencies of the current;
- mathematical processing and graphical representation of experimental results;
- familiarization with some applications of the researched method in medical practice.

Theoretical notions

As we know, the alternative current is produced by the electrical field that varies in time after the sinusoidal law:

$$E = E_m \sin \omega t \qquad (1)$$

Here E is the momentum value of electrical field, E_m –maximal value (of amplitude) ω – pulsation. ($\omega = 2\pi\nu$, ν -frequency). From this, the intensity I will follow the same law of variation in time:

$$I = I_m \sin(\omega t + \varphi_0) \qquad (2)$$

Here the value φ_0 is named the phase difference between the intensity and voltage.

If the conductor has the form of solenoid (coil), the phenolmenon of selfinduction takes place, it makes the intensity remain

behind the voltage, the phase difference has the value $\varphi_0 = \frac{\pi}{2}$

$\varphi_0 = -\pi/2$.

The pure resistance does not introduce any phase difference.

For a circuit made of pure resistance R, a coil with the inductance L and capacity C, linked consecutively and supplied by alternative current (fig.9.1), the total resistance Z(or the impedance) Is determined by the equation

$$Z = \sqrt{(R + R_L)^2 + (\omega L - \frac{1}{\omega C})^2} \qquad (3)$$

here $\omega L = X_L$ is inductive reactance, $\frac{1}{\omega C} = X_C$ capacitance reactance, but R_L- the resistance of continuum current of the coil. It is established that in living tissues the inductive reactance is absent.

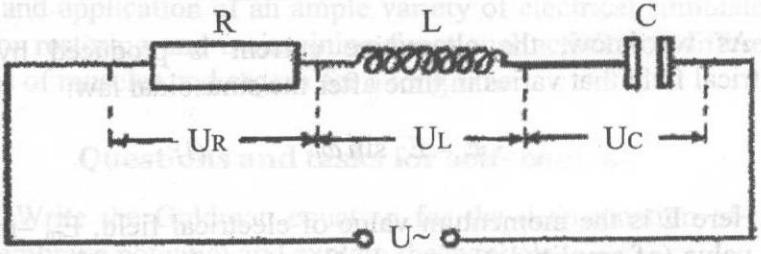


Fig.9.1

Taking in consideration this fact the impedance for living biological tissues is determined by the equation:

$$Z = \sqrt{R^2 + (1/\omega C)^2} \qquad (4)$$

The presence of capacitance reactance is caused especially by cell membranes, their capacities depend on their geometrical parameters and the state of the medium. Physical characteristics of this medium vary under the influence of external electromagnetic fild and also as a result of variation of permeability of cell membranes. Taking this into account, living tissues have a dependence on the the frequency existing not only for the capacitance reactance after the equation $X_C = \frac{1}{\omega C}$, but also for the capacity of structural components. From this living tissues have different electrical conductivity for alternative current of different frequency in comparison with the electrolytes their conductivity does not depend on the frequency of current (in the range from tens up to millions of Hz). The phenomenon of variation of impedance of living tissues as dependence on the frequency of electrical current is named the dispersion of impedance.

Graphically this dependence is represented by the so-called curve of dispersion (fig. 9.2):

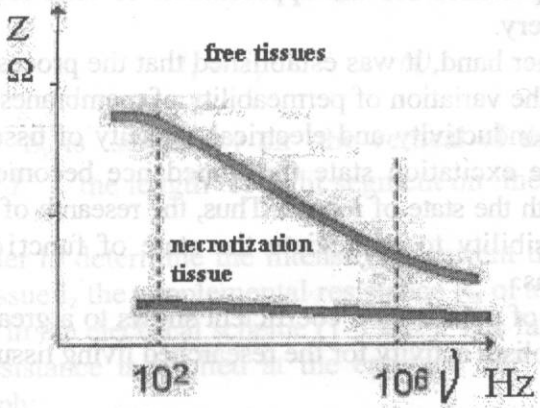


Fig. 9.2

The interval of frequencies in which the dispersion of impedance is observed is called the field of dispersion. Often instead of building the curve of dispersion, the so-called polarization coefficient is only determined:

$$K = \frac{Z_{y\min}}{Z_{y\max}} \qquad (5)$$

In the necrotization of tissue, the polarization coefficient tends to the unity. (The dependence on impedance of frequency disappears). The information mentioned above is characteristic of both the tissues of animal nature and vegetal nature.

The importance for medical practice

Though the necrotizated tissue and the electrolyte do not have the dispersion of impedance, the value of polarization coefficient characterizes the vital state of different tissues and organs. This has a practical importance for the appreciation of vital level of transplants in surgery.

On the other hand, it was established that the process of excitation provokes the variation of permeability of membranes that has an effect on the conductivity and electrical capacity of tissues. Due to this fact in the excitation state the impedance becomes miller in comparison with the state of repose. Thus, the research of impedance gives the possibility to appreciate the state of functionality of different organs.

The value of polarization coefficient shows to a great extent the level of metabolism activity for the researched living tissue.

Equipment and materials

Cathode oscillograph, the generator of sounds (ultrasounds), biological tissue - the object of research supplemental resistance, two electrodes of non-oxidative iron and flexes of connection.

Description of installation and calculation

The principal electrical scheme of the used installation is represented in fig. 9.3.

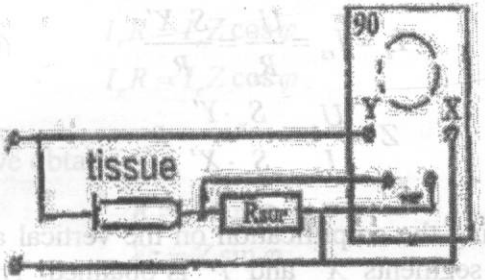


Fig. 9.3

The installation allows determining U_z and I_z. Then their values Z are calculated. The decrease of tension on the sector of the object between electrodes U_z is applied at the entrance "Y" of the cathode oscillograph. The value of this voltage is determined by the formula:

$$I_z = S_y \cdot Y' \qquad (4)$$

Here S_y is sensitivity on the vertical of the oscillograph, $V \cdot \min^{-1}$ Y' - the length of light segment on the screen in millimeters.

In order to determine the intensity of current through the researched tissue I_z the supplemental resistance R_s of a known value is connected in the electrical scheme (1-3 kΩ). The falling of voltage on this resistance is applied at the entrance "X" of the cathode oscillograph:

$$U_{rs} = S_x \cdot X' \qquad (5)$$

Here S_x is sensitivity on the horizontal of the oscillograph, X' - the length on the horizontal of the light segment on the screen. Knowing R_s and U_{rs}, we can determine I_z and Z using the law of Ohm:

$$I_z = I_{rs} = \frac{U_r}{R_s} = \frac{S_x X'}{R_s} \qquad (6)$$

$$Z = \frac{U_z}{I_z} = \frac{S_y \cdot Y'}{S_x \cdot X'} \cdot R_s \qquad (7)$$

Adjusting the amplification on the vertical and horizontal, the equality of segments X' and Y' is obtained. The equation for the calculation of electrical impedance is simplified:

$$Z = \frac{S_y}{S_x} \cdot R \qquad (8)$$

Knowing the value of impedance Z its components R and X_c can be determined using the vector diagram of tensions (fig. 9.4).

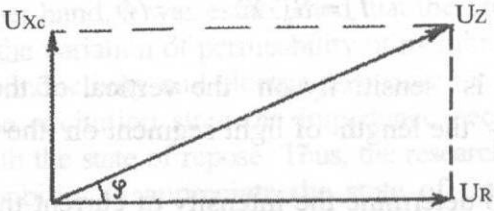


Fig.9.4

$$U_r = U_z \cdot \cos\varphi$$

$$U_r = U_z \cdot \cos\varphi$$

Here φ is the phase difference between the tension and current in the researched tissue. As R and C are considered to be connected in consecutively, then $I_r = I_c$ and:

$$I_r R = I_r Z \cos\varphi$$
$$I_c R = I_c Z \cos\varphi$$

Simplifying we obtain:

$$R = Z \cos\varphi$$
$$X_c = Z \sin\varphi$$

The phase difference φ is calculated after the parameters of the ellipse described on the screen of oscillograph as a result of composition of reciprocal perpendicular oscillations of the same frequency (fig. 9.5).

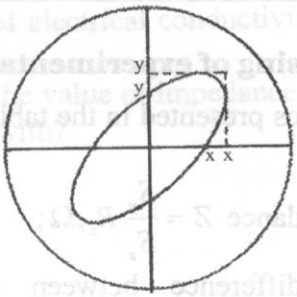


Fig. 9.5

Work procedure

1. The scheme of installation is made as in fig 9.3.
2. The apparatuses are connected up in the electrical net, establishing consequently their regime of work. The outlet tension of the generator of sound is 1 - 2 V, but the frequency is 500 Hz.

3. Shutting the basis of time, the amplification of oscillograph is adjusted on the vertical and horizontal in such a way, that the ellipse described on the screen will be written within a square with the lattice of 20 mm. Consequently, the condition is obtained as: $Y' = X'$

Y' and Y are measured on the screen. The obtained results as well as the values S_y and S_x determined from the graphics are introduced in the table below.

Table 9.1

Nrof exp.	Freq. kHz	Y, mm	Y', mm	ψ, grad	S_y, V/mm	S_x, V/mm	Z, Ω	R, Ω	Xc, Ω	C, ηF	K
1											
2											
3											
4											
5											

Processing of experimental results

For all frequencies presented in the table above, we can determine:

– electrical impedance $Z = \frac{S_y}{S_x} R_s, \Omega$;

– the phase difference between current and tension $\varphi = \arcsin(\frac{Y}{Y'}), grad$

– the active resistance of the researched tissue $R = Z\cos\varphi, \Omega$

– the capacitance resistance: $X_C = Z\sin\varphi, \Omega$;

– the capacity of tissue $C = \frac{10^6}{2\pi\gamma X_C}, \mu F$;

– the coefficient of polarization $K = \frac{Z_{0,5}}{Z_{50}}$

– the dependence of electrical impedance on current frequency, u five resistance and capacitance resistance of researched tissue are represented graphically.

Questions and tasks for self- control

1. Write the formula for the value of impedance into the circuit of alternative current (containing R; L and C connected consecutively) and the value of electrical impedance of living biological tissue.

2. What does the impedance dispersion phenomenon of living tissues mean? Write the formula for determination of the polarizing coefficient.

3. What are constructive elements of the didactic device that allows the modeling of electrical conductivity of biological membrane?

What purpose is the value of impedance of biological tissue in medicine determined with?

10. METHODS OF OPTICAL MICROSCOPY

Purposes

- presentation of theoretical notions of optical microscopy;
- study of the construction and the principle of functioning of the microscope;
- determination of linear enlargement;
- determination of red cells diameters;
- study of ultramicroscopic method;
- modeling of polarization microscope.

Theoretical notions

Optical microscopy consists of the unity of examination methods and the study of objects of very small dimensions that can not be seen clearly with the naked eye and it uses optical equipment called microscopes.

In the development of microscopic techniques the following device were invented: a simple microscope or a magnifying glass, a composed microscope, the devices of ultramicroscopy, of polarization, of phase contrast, etc. were adapted.

a) Simple microscope consists of a convergent lens with the big focal distance of 10 – 100 mm.

The image from the magnifying glass is virtual, right and bigger than the object itself. The researched object is placed between the focal of the lens and its optic center (Fig. 10.1). The most important characteristic values of the magnifying glass are: power P (or the convergence) is expressed by the angle under which the unit of the length of the object is seen and that is equal to the inverse focal distance of the lens:

$$P = \frac{\alpha'}{AB} = \frac{1}{F} \qquad (1)$$

The angular enlargement is the ratio between the angle α' under which an observer can see the object through the magnifying glass (fig. 10.1) and the angle α under which the same object is seen with ihr naked eye at the minimal distance of distinctive vision, (d):

$$G = \frac{\alpha'}{\alpha} \qquad (2)$$

$$for\ \alpha < 5^0, tg\alpha = \alpha\ and\ \alpha = \frac{AB}{d} \qquad (3)$$

Substituting α' from (1) and α from (3) equation 2) becomes:

$$G = \frac{\alpha'}{\alpha} = \frac{P \cdot AB}{\frac{AB}{d}} = P \cdot d$$

Thus, the enlargement of one magnifying glass is equal to the product of magnifying power and minimal distance of distinctive vision.

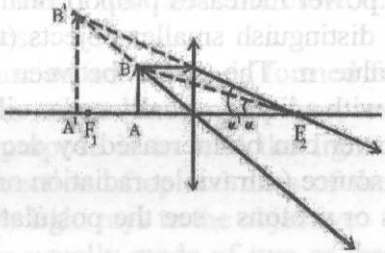


Fig. 10.1

The linear enlargement (the power of linear increase) is the ratio between the image value I and the object value O:

$$G = \frac{I}{O} = \frac{A'B'}{AB} \qquad (4)$$

The resolution power is the capacity of optical system to separate two near points, so that they can be received distinctly. It is inverse proportional to the minimal distance (d) between two light points of the object received separately. It is given by the Abbe formula:

$$d = \frac{0.526\lambda}{n \cdot \sin\alpha}$$

Where **n** is the refractive index of medium between the object and objective lens; α – the aperture angle (the angle between the optical axes of the system and the external ray of the flux); λ - the wavelength of the used light.

The value $n \cdot \sin\alpha$ is called the numerical aperture.

Usual microscopes allow to resolve some minute dimensions equal to about $\frac{\lambda}{2}$.

As the resolution power increases proportionally to the numerical aperture, we can distinguish smaller objects (the objects with immersion) by the value n. The space between the object and objective lens is filled with a liquid, usually cedar oil.

The resolution power can be increased by decreasing the wavelength emitted by a source (ultraviolet radiation or associated wavelengths of electrons or protons - see the postulate of L. de Broglie).

b) The composed microscope (fig. 10.2) is made of three principal parts: 1) mechanical part, 2) optical part, 3) illumination device of the object.

1) Mechanical part is composed of:

– basement of microscope (a) made of metal that assures the necessary stability;

– the table (b) of square or round form serves to sustain the object. An object is fixed on the platform by the gallants (c);

– the microscope tube (d) sustains the objective lens (e) and the ocular (f) which can have a rapid movement by the pot –hangers (g) fixed with a screw (h) and a fine movement by graded micrometer screw (i);

– the piece named the revolver (j) is placed at the inferior end of the tube, on which 2-4 objective lenses can be mounted and which allows to chose them rapidly by rotation of the support.

2) Ocular and objective lens compose the optical part. The ocular represents a composed magnifying glass formed usually by 2 lenses, an inferior convergent lens, and a divergent one placed from each other at the distance equal to half of the sum of the focal distance. Both lenses are fixed into a cylindrical body introduced into the superior orifice of the tube. The oculars are numerated as the objectives in the order of enlargement. The objective represents a system of lenses fixed into the support. The front lens, the socalled frontal lens, is the principal and single lens producing magnification. Others serve for correction of defects of the images reated by the frontal lens and, therefore, these lenses are named correction lenses.

3) The illumination device is formed from the mirror (k) and the lens (1). The mirror has two surfaces, one – plane and the other –concave; its purpose is to lead the light source rays (a frosted bulb) to the direction of optical axes of the microscope. The lens concentrates the light rays to the objective.

The lens is usually made of two or three lenses by which the ivllected light by the mirror is concentrated on the objective in the form of one strong convergent flux. In order to obtain a more clear image it is necessary to place object at the focal of this flux; and to

achieve this the lens is moved up or down by the screw. Some lenses have a variable diaphragm. The light source is placed at the basement of the microscope and the mirror is absent in modern microscopes. The illumination of microscope field must not be very strong because in this case the contrast is decreased ("drowning in the light"). *Fig.10.2*

Fig. 10.2

Formation of image in the microscope

The object O is beyond the external focal F_1 of the objective and very close to it. The image I' will be real, upset and bigger: it is formed near the ocular, between the focal of the ocular F_2 and the ocular (fig. 10.3) This intermediary image has the role of an object with respect to the ocular that works as the magnifying glass and gives the final image I, right, virtual and bigger than the image I'

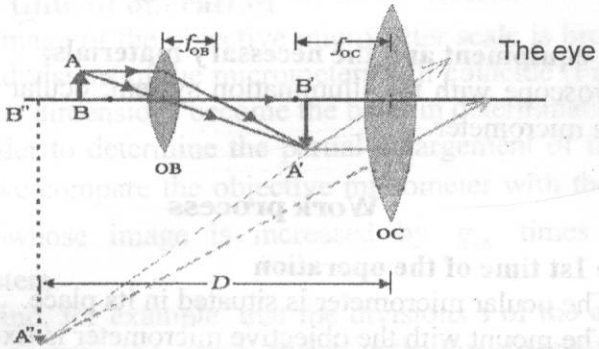


Fig. 10.3

The final image will be formed at the minimal distance of distinctive vision into a light plane named the optical field. As the image I' given by the objective is upset with respect to the object, the image given by the ocular is inverted with respect to the object. The enlargement of the microscope is given by the following formula:

$$G = \frac{I}{O}$$

Note: g_{ob} - the objective enlargement $g_{ob} = \frac{I'}{O}$ g_{oc} - the ocular enlargement $g_{oc} = \frac{I}{I'}$

The enlargement of the microscope can be written like this:

$$G = \frac{I}{O} = \frac{I'}{O} \cdot \frac{I}{I'} = g_{ob} \cdot g_{oc} \qquad (5)$$

Thus the total enlargement of the microscope is equal to the Iroduct of partial enlargements.

Determination of enlargement

The equipment and the necessary materials:

microscope with the illumination system; ocular micrometer; objective micrometer.

Work process

The 1st time of the operation

– The ocular micrometer is situated in its place.

– The mount with the objective micrometer is fixed on the microscope table, with the mounts up. The objective micrometer is situated into the black box of bakelite on the working table. **The micrometer is seized only by the edge; the divided portion is not touched by fingers.**

– The bending of the concave mirror is adjusted so that the maximal illumination of the image is obtained.

– The circle from the middle of the micrometer is brought in front of the objective by the screw that moves the table of the microscope.

– The smaller objective is placed (10 x).

– The objective is approached carefully to the micrometer till the micrometer divisions appear in the microscope field.

– **Warning: The objective must not touch the objective micrometer to avoid its breaking.**

– The microscope is adjusted so that the divisions of the objective micrometer and the image of ocular micrometer are seen at the same time.

– The ocular is rotated in its place till those two scales are parallel, enclosed, just partially superposed. In order to increase the contrast the lens is lifted or put down.

The 2nd time of operation

– The image of the objective micrometer scale is brought so that two big divisions of the micrometers will coincide (Fig. 10.3) The respective dimensions become the basis in determinations.

– In order to determine the partial enlargement of the objective (g_{ob}) we compare the objective micrometer with the ocular micrometer whose image is increased by g_{ob} times by the objective system.

– We find, for example, that the divisions n of the objective micrometer occupy the same space with the divisions m of ocular micrometer. We find the coincidence of divisions farther than zero in order to be exact.

The objective micrometer has the gradations of 0.01 mm; the ocular micrometer has the gradations of 0.1 mm.

Thus, we can write: $n \cdot \frac{1}{100} \cdot g_{ob} = m \cdot \frac{1}{10}$ from which the value of g_{ob} is found:

$$g_{ob} = \frac{100}{10} \cdot \frac{m}{n} \qquad (6)$$

For each objective (10x, 40x, and 90x) only three determinations are performed.

The 3rd time of operation

The total enlargement of the microscope is: $G = g_{ob} \cdot g_{oc}$. We determine the total enlargement of the microscope replacing g_{ob} with the average obtained value for each particular objective. The results are written in table 10.1.

Table 10.1

Objective	Nr. of determ	n	m	g_{ob}	G
10x	1				
	2				
	3				
40x	1				
	2				
	3				
90x	1				
	2				
	3				

The calculations are performed in this way.

$$g_{ob} = 10\frac{m}{n}$$

$$\overline{g}_{ob} = \frac{\sum g_{ob}}{3}$$

$$G = \overline{g}_{ob} \cdot g_{oc}$$

Determination of red cells diameters

The determination of red cells dimensions is of clinical importance helping us to establish some diagnosis along with other investtigations. For example, in anemia of the Bierman type the diameter is increased (8-10 μm), but in hypochromial anemia it is decreased (6.2-6.7 μm), etc.

The graphical transcription of the red cells percentage with the diameter in normal limits constitutes the curve Price – Jones, correct for the pathological deviations (for example, the curve is flat in haemolysis jaundice).

Equipment and necessary materials

- The microscope with the illumination system.
- Microscope mounts.
- Pipette.
- Blood.

Work process

The first time of operation

Smearing. A drop of blood is put on the cleaned mount. With the edge of another mount the drop is touched and stretched along the edge. The upper mount is moved rapidly in the direction of obtuse angle, the drop is stretched uniformly and in a thin layer on the mount. (Fig. 10.3) We let it dry.

The second time of operation

– The smear is put on the microscope table; it is worked with the bigger objective (90x). The image of particles is adjusted and the divisions c of the ocular micrometer are counted as the ones corresponding to one diameter (Fig. 10.4). Ten determinations are performed for different red cells. The value is calculated:

$$d = p \cdot \frac{1}{g_{ob}} \cdot \frac{1}{10} \quad (d \text{ in mm})$$

The normal value is of 7.2 – 7.9 μm, varying with the age and sex. The results are written in table 10.2:

Fig. 10.4

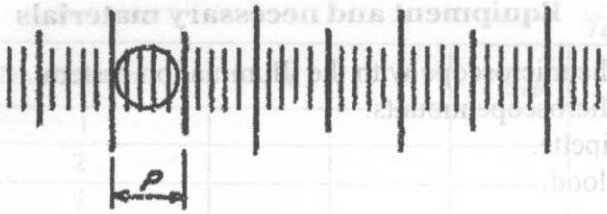


Fig. 10.5

Note: *d* represents the average value

Table 10.2

Nr. det.	*p(div)*	*d(μ)*
1		
2		
3		
4		
5		
6		
7		
8		
9		
10		
	$\overline{d}$ =	

Ultramicroscopy

Theoretical notions

As a powerful ray of light passes through the disperse molecular solution and we have a look at the solution in the perpendicular direction to this ray we can not see its way inside the solution, therefore this solution is called empty optical medium. If we replace the molecular solution with sol, all the trajectory of the ray is observed

very well due to a light diffusion by the colloidal particles. This phenomenon is called the Tyndall effect.

Ultramicroscopy is based on the Tyndall phenomenon. The principle of the method of ultramicroscopy can be schematically represented as follows: a light flux is concentrated by the electrical bulb and the lens (fig. 10.6) on the vase of parallelepiped form D that contains a colloidal system.

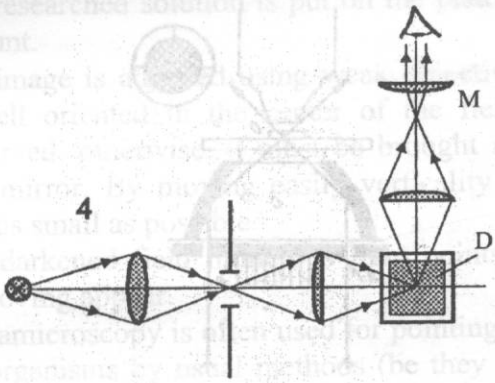


Fig. 10.6

The horizontal light is diffused by it in all directions. A part of this diffused light comes into the objective of the microscope M placed vertically and gives a diffraction figure that allows to observe the position and remove particles (for example, the Brownian moving). Sometimes the form of particles (spherical, lamellar, stretched, and ovoid) can be distinguished very vaguely. The particles up to three millimicrons can be observed by the ultramicroscope. In fact, each microscope can be transformed into ultramicroscope by introduction of usual lenses of light with devices Balled ultramicroscopic lenses.

The optical system of this device is built so that the light ray passing through it crosses the object in the lateral direction and es the Tyndall effect without penetrating into the microscope objecttive.

Of ultramicroscopic lenses we mention the cardioid and the parabolic lenses of Siedentrop. The passing of rays and schematic construction of the above mentioned lens is seen in (Fig. 10.7) where: A - light ray; B - the diaphragm obturating the base of the device C, leaving transparent only a circular zone near to the periphery of the lens; O - preparation.

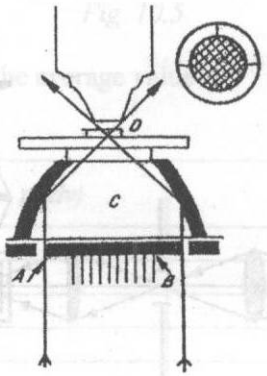


Fig. 10.7

The Brownian moving of colloidal particles can be observed by ihe ultramicroscope. The Brownian moving is due to the collisions at which the particle is submitted by the liquid molecules surrounding it. The particle has the approximate diameter of about 10^{-5} cm, but the molecule has the diameter about 100 times smaller.

Work process

– A parabolic or cardioid lens is fixed instead of the lens of the usual microscope.

– The lamp is fixed at the distance of about 10cm from the mirror of the microscope.

– The illumination system is verified.

– The lens is moved a little down and a drop of cedar oil is put on the superior lens of this.

– A plate of glass is put on the table, supplied with a groove for placing colloidal sol, after that a drop of oil cedar is put preliminary on its inferior part.

– The lens is lifted and those two drops of cedar oil are linked.

– The formation of air bubbles is avoided.

– The researched solution is put on the plate and is covered with the mount.

– The image is adjusted using weak objectives (10x). If the mirror is well oriented in the center of the field, a light spot will be observed: otherwise, it must be brought at the center by moving the mirror. By moving easily verticality of the lens we make a spot as small as possible.

On the darkened field numerous light points presenting the Brownian moving appear.

The ultramicroscopy is often used for pointing out difficult visible microorganisms by usual methods (be they scarcely colorable, or very small) as: treponema, leptospirals, rickettsia, etc.

Note: As the ultramicroscopic lenses have a very short focus (maximum 1.1 mm) the plates of glass with the thickness of about 0.8 – 1 mm. will be used.

Questions and tasks for self – control

1. Draw a simplified optical scheme of microscope.
2. Define and write the formula for the determination of parameters characterizing the quality of one optical microscope.
3. What physical phenomenon is the basis of the method called ultramicroscopy? Present the schematic construction and passing of the rays through the ultramicroscopic lens.
4. What are constructive elements and the work mode with the didactical device modeling the polarization microscope?

11. PHOTOVOLTAICAL CELL. LUXMETER.

Purposes

- Determination of theoretical notions regarding photoelectric effect.
- Determination of photocurrent variation as illumination and sensitivity of photoelectric cell for the white light.
- Standardization of the laboratory device as of luxmeter.

Theoretical notions

The photoelectric effect belongs to the phenomena that show the corpuscular character of light radiation. Hertz discovered this phenomenon in 1887. It was established experimentally that under the influence of light the energetic state of electrons from different substances undergoes modifications.

The emission phenomenon of free electrons from the metal under the influence of the light radiation is called external photoelectric effect conditioned by the following laws:

1. The number of emitted electrons by a substance in the time unit increases proportionally with the intensity of incident light radiation (I~E).

2. The frequency of incident radiation must be greater than a certain limit ν_0 (photoelectric threshold) to extract the electrons.

3. The kinetic energy of emitted electrons depends on light frequency ν (directly proportional). The basic equation of external photoelectric effect established by Einstein has the form:

$$h\nu = W_e + \frac{mV^2}{2}$$

Where h is the Planck constant ($6.624\ 10^{-34}$ J·s), ν – the frequency of incident light radiation; W_e – the minimal energy

necessary for pulling out electrons from metal, $\frac{mV^2}{2}$ – kinetic energy of electrons.

In case of dielectric media and semiconductors the internal photoelectric effect takes place and it consists in decreasing their electrical resistance under the influence of light.

In darkness an examined semiconductor has an electrical conductivity named the darkness conductivity. The illumination conductivity appears under the influence of light radiation named photoconductivity that allows their utilization as sensible resistances in illumination.

The photoelectric effect can take place and, producing electron holes, at the same time it assures their separation conditioned by the presence of a dam layer. This phenomenon is called photoelectric effect of valve obtained at the illumination of p – n junction. Such a device is called photovoltage cell and acts as a generator of electric current.

Practical realization of photovoltage cell is shown in fig. 11.1.

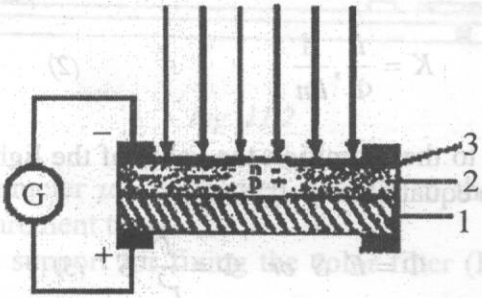


Fig. 11.1

It is made of the metal support plate **1** that serves as one of the electrode. The layer **2** of selenium with the hole conductivity on which semi transparent metal layer **3** is laid (the third electrode).

From the metallic layer **3** the atoms penetrate in the upper part of the selenium layer, thus imposing the electronic conductivity, but the lower parts of semiconductor preserve hole conductivity. Between the mentioned layers the junction p – n is formed.

The light radiation passing through semitransparent metallic layer makes hole – electrons pairs in the region of junction that disequilibrates the potentials from both faces of the dam layer and makes an electromotive voltage between the electrodes of the cell able to maintain a current in the external circuit as long as the cell is illuminated.

The following materials can be used as semiconductor materials: copper oxide, sulfur of silver, germanium, etc. The applications of this type of cell (sometimes called the cell with the dam layer) are numerous, such as: photometry, photocolorimeters, spectrophotometers, etc.

The main parameter of photovoltage cell is the integral sensitivity K, numerically equal to the current intensity i from the cell circuit when the incident flux on its active surface is equal to a lumen:

$$K = \frac{i}{\Phi}, \frac{A}{lm} \qquad (2)$$

According to the definition the value of the light flux is determined from the equation:

$$\Phi = E \cdot S \quad or \quad \Phi = \frac{I}{r^2} \cdot S \qquad (3)$$

$$E = \frac{\Phi_{inc}}{S}$$

Where I is the light source intensity, S – illuminated surface of photoelectric cell, r – the distance between the light source and active surface of the cell.

From the equations (2) and (3) we obtain:

$$K = \frac{i \cdot r^2}{I \cdot S}$$

The description of equipment

The photoelectric cell C is placed on the optical plate (fig. 11.2) in a fixed position. At the regulated distance the light source S is placed (the electric bulb of known voltage)

The voltage applied in the bulb is maintained constantly by the potentiometer **P** being indicated by the voltmeter **V**.

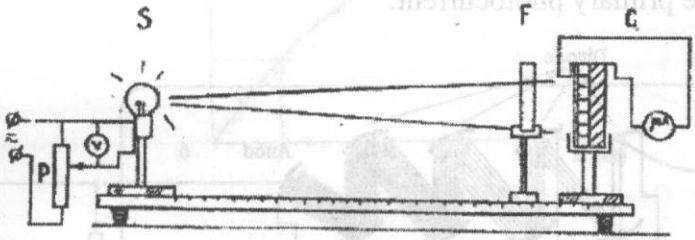


Fig. 11.2

A microammeter μA is coupled directly to the photoelectric Cell (the measurement tool).

There is a support for fixing the color filter (F) on the same optical plate.

Application in biology and medicine

The vacuum equipment or semiconductors the working principle of which is based on photoeffect are called photoelectric equipment. We will examine the construction of one of them.

To increase the intensity of photoelectric current the photo-elements filled with gases are used. In these gases nonautonomous obscure discharge in an inert gas and the secondary electronic emission occurs, that is electron emitting takes place as a result of metal surface bombing by the prime electronic flux. The bombing is used in the photoelectric multiplier (PEM).

The scheme of PEM is presented in fig. 11.3. Under the influence of incident photons the photocathode C eliminates electrons focused on the first cathode of the second emission (diode) E_1. As a result of electronic secondary emission from this diode a lot of electrons fly from it rather than fall on it, so, the multiplication of electrons takes place. As a result multiplying on other diodes, the electrons form a current of hundred and thousand times greater thcin the primary photocurrent.

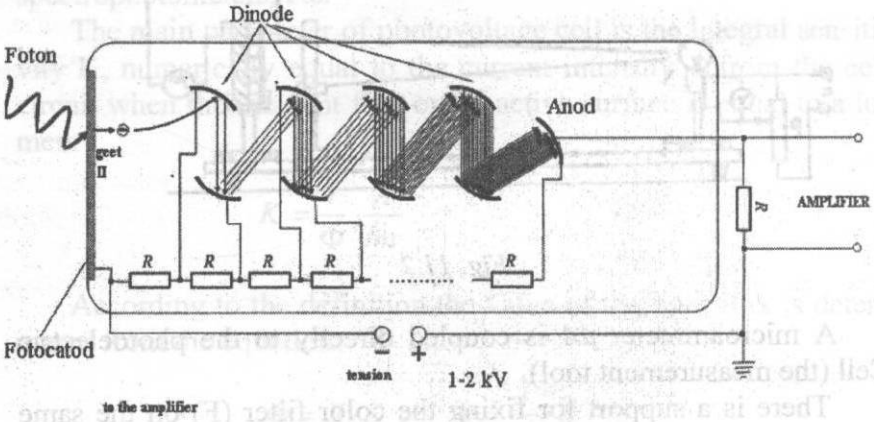


Fig. 11.3

The light "receivers" of visible range serve as photoelectric Mils in different automatic systems the spectral sensitivity of which reflects the eye spectral sensitivity.

Work process

1. Varying successively the distance **r** between the photoelectric cell and the bulb the current **i** indicated by microammeter is read every time. The active surface of photoelectric cell S and the Intensity of the light source are given. The values E and K are calculated according to the equations (3) and (4).
2. Numerical results obtained are written in table 11.1
3. According to the data from table 11.1 the scheme i=f(E) is drawn, being the same as in fig. 11.4

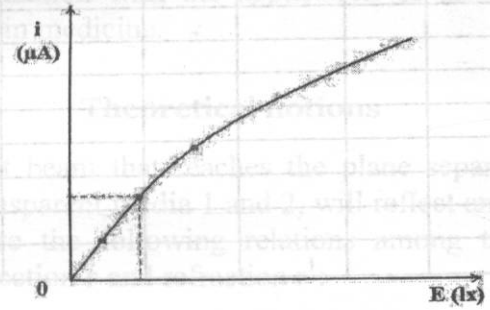


Fig. 11.4

Standardization of laboratory device in quality of luxmeter

Simple transformations of equation (4) allow finding the univocal correspondence between the values of photoelectric current and respective values of illumination:

$$E = \frac{I}{r^2} = \frac{i}{KS} \qquad (5)$$

The equation (5) allows standardizing of microammeter as the illumination measurement tool.

It is recommended to determine illumination at different places of the room.

In this case when the needle deviation of the tool exceeds the limits of the scale, the diaphragm limits the active surface of photoelectric cell. The value of illumination is determined according to the equation (5), where S will represent the slit of diaphragm.

Table 11.1

No of experiment	I, cd	S, m^2	r, m	I, μA	E, lx	K, $\frac{\mu A}{lm}$
1						
2						
3						
4						
5						
6						
7						
8						
9						
10						

Questions and tasks for self- control

1) What are the laws of external photoelectric effect? Write the equation of Einstein for the external photoelectric effect and explain the symbols.

2) Having the example of selenium photovoltage cell, explain the mechanism of valve photoelectric effect performed at the illumination of p – n junction.

3) What is the device and the principle of action of photomulti-plifier?

12. DETERMINATION OF SERUM PROTEIN SUBSTANCE CONTENT BY REFRACTIVE INDEX

Purposes

- presentation of theoretical notions of the refractometer method;
- principles of method and construction of the apparatus;
- determination of the protein present in the researched blood serum;
- familiarization with the application of the refractometer method in medicine.

Theoretical notions

An incident beam that reaches the plane separation surface between the transparent media 1 and 2, will reflect and refract (fig. 12.1). There are the following relations among the angles of incidence i, reflection **r** and refraction r^1:

$$i = r\,;\ \frac{\sin i}{\sin r^1} = \frac{V_1}{V_2} = n_{21} \qquad (1)$$

Where V_1 is the speed of the light into the media 1.

V_2 – the speed of the light into the media 2. The coefficient n_2 is called the refractive relative index of the medium 2 with respect to the medium 1.

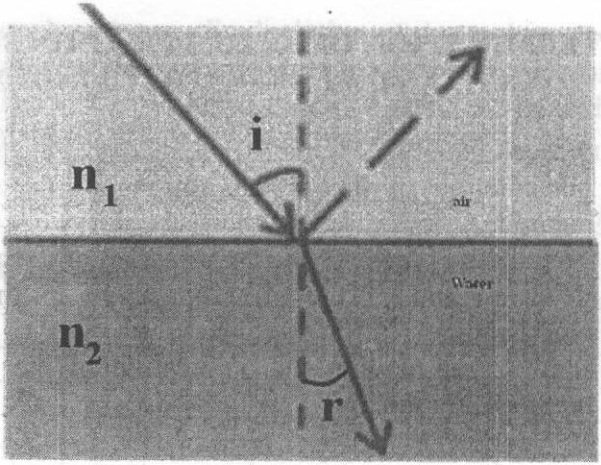


Fig. 12.1

The absolute refractive index of a substance is defined with respect to vacuum. Considering that the first medium is vacuum then:

$$\frac{\sin i}{\sin r^1} = \frac{C}{v_2} = n_2 \qquad (2)$$

The relative refractive index can be defined as the ratio between the absolute refractive indexes of those two media:

$$n_{21} = \frac{V_1}{V_2} = \frac{n_2}{n_1} \qquad (3)$$

The refractive index of a substance depends on a series of factors such as the nature of respective substance, $n_1 < n_2$ wavelength of the applied light, temperature, pressure, etc. From these facts the refractive index is marked, for example, in the determinations and tables as n_D^{20}, meaning that the determination was performed at 20°C using a radiation from a visible range with the wavelength of 589.3 nm (the line D of sodium).

The refractive index of the given substance varies with density and its aggregation state. The increase of density leads to the increase of the refractive index. A certain function of refractive index $F(n)$ varies directly proportional to the density ρ:

$$F(n) = r_s \cdot \rho$$

Where r_s is a characteristic coefficient for the given substance and it is called specific refraction which depends neither on external conditions nor on aggregation state of the substance. In order to determine the value of specific refraction physicists H. Lorenz and I. Lorenz established the equation:

$$r_s = \frac{n^2 - 1}{n^2 - 2} \cdot \frac{1}{\rho}$$

The product between the specific refraction of substance and molecular weight is called molecular refringence power (molecular refraction).

The molecular refringence power of a chemical compound is equal to the sum of atomic refringence powers of constitutive elements.

In organic compounds double or treble binds bring modifications of molecular refringence power. It means that the molecular refringence power depends on the chemical structure.

Principle of the method and construction of apparatus

It is known that when one light beam passes from a dense medium into a less dense medium, refraction deviates from the normal one. The refraction angle increases more rapidly than the incident one.

A value of incident angle is reached which the refractive angle of 90° corresponds to (fig. 12.2). It is a situation in which the

refracted ray is tangent to the separation surface. The incident angle into the dense medium, which the refractive angle of 90° corresponds to, is named limit angle (i_L). The rays, coming on the separation surface under the incident angles greater than the limit angle are totally reflected into a denser medium. For the limit angle the second law of refraction will be:

$$n_2 \sin i_1 = n_1 \sin 90^0 \text{ But } \sin 90° = 1 \text{ so:}$$

$$n_1 = n_2 \sin i_L \qquad (5)$$

As the refraction index of the denser medium is known the index of another medium can be determined by measuring the limit angle, whose value is introduced in the equation (5).

The advantage of this process is that a single angle must be measured but in order to observe the total reflection only the presence of the plan separation surface is necessary.

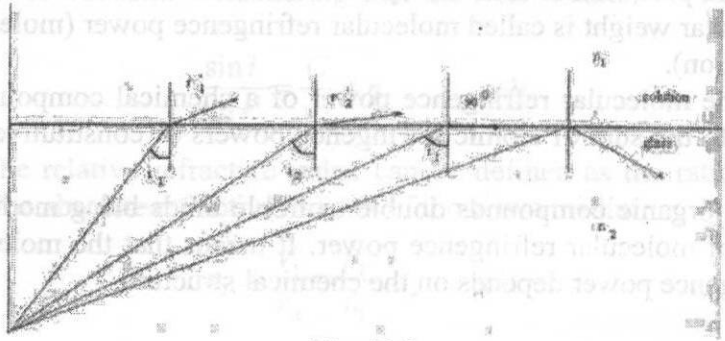


Fig. 12.2

Special apparatuses called refractometers determine the limit angle.

This work will be done using the Pulfrich refractometer with prisms warmed by water or the refractometer of the Abbe type. The constitutive principal parts of a refractometer are the prism of measurement, optical tube or compensator.

The Pulfrich refractometer is made of prism of measurement and one of illuminations, both fixed in empty metallic supports in order to penetrate the water for temperature regulation, coming from the ultrathermostat through the rubber tube.

Although the surface of the prism of measurement is oriented up and down from the illumination prism, the position of the separation limit is turned, therefore, the obtained values must be subtracted from 100.

The researched liquid is put on the prism surface in a very small quantity (0.1 – 0.2 ml) and the illumination prism is put on. The researched liquid forms a less dense medium whose refractive index must be determined. The optical tube of refractometer is supplied with an objective and an ocular with the scale of divisions from – 5 to 105 for reading the limit between the light zone and the darkened one of the field of vision.

The compensator is formed of a system of prims playing the role of recombining the dispersed light. In order to illuminate the prism we will use a sodium lamp that sends a yellow monochromatic light beam (λ= 589 – *550nm)to* the device.

The principle scheme of the Pulfrich refractometer is presented in fig. 12.3 and fig. 12.4.

The micrometer scale is in the right part of the ocular. It serves for appreciation of decimals. There is a correction system of standard scale next to it.

The placed substance between the two prisms whose index must be determined makes the optical contact with the measurement prism, on which separation surface the refraction and total reflection take place. The separation limit of both illuminated and darkened zones appearing in the lunette of the ocular corresponds to the limit ray. (The ray determining the limit angle with the normal one in the denser medium).

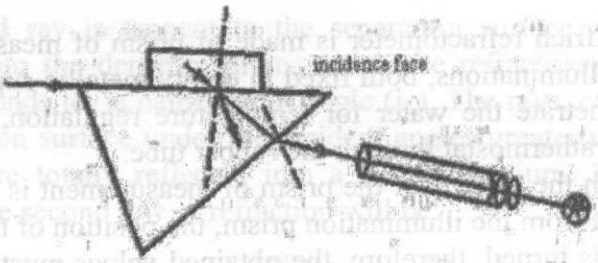
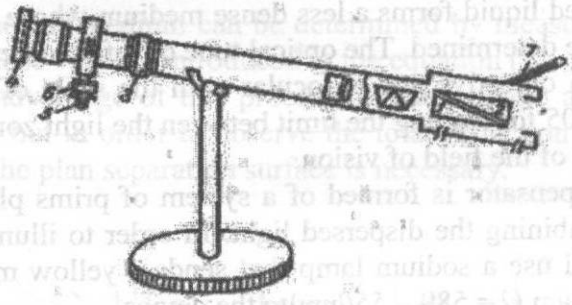


Fig. 12.3



Fulfrich refractometer

1. prism of measurement
2. prism of illumination
3. compensator
4. ocular
5. micrometiical screw
6. system of standardization
7. thermometer

Fig. 12.4

Ultrathermostat is a device with the help of which we can obtain water at a perfectly regulated temperature. The water is moved by means of an engine to the prisms of the refractometer through

rubber tubes. A sensible thermometer is installed on the prisms measuring the temperature around them. The determinations are made at the temperature of 17.5 °C or 20°C.

Special tables with values of refractive index convert the values of limit angle read from the refractometer scale. As in biology we are interested, practically, in the researched medium concentration the values of the limit angle are transformed directly into percentage concentrations by means of special tables called the Wagner tables. The Wagner table for proteins is on the work desk.

The Abbe refractometer presents the same constitutive parts as the Pulfrich refractometer (fig. 12.5) and the principal part of this device is also a double flint prism formed from two halves: one of measurement and another of illumination. The prism of measurement is placed in the support (4) and it is mobile on the horizontal axes. A small space exists between the prism planes that do not touch each other and the probe is introduced in it.

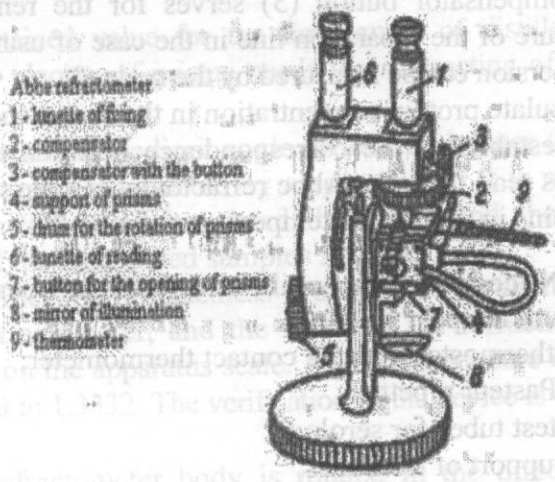


Fig. 12.5

The researched liquid must have a refractive index smaller than that of the prism.

In order to maintain the constant temperature the both prisms are surrounded by a metallic cloak and water stream flows from the ultrathermostat through it. The temperature can be read from the scale of the thermometer (9) attached to the prisms.

A sodium lamp is used as the light source. The light rays are reflected in the direction of the illumination prism by the mirror (8).

Two reticular wires are fixed in the lunette ocular 1) perpendicular between them. The limit between the darkened space and light one are brought exactly at the crossing point (fig. 12.3) by rotating the prism by the drum (5). The position, which the prisms have when the direction of incident rays on the liquid layer reaches the limit angle, is read from the refractometer scale in the form of arch. On this scale the values of refraction indexes n_D from 1.3–1.7 are represented directly. The circular scale is graded up to the units of the third decimal; the forth decimal is estimated by a vernier. The exact reading on the vernier is made by the magnifying glass.

The compensator button (3) serves for the removal of the colored picture of the separation line in the case of using white light and the dispersion can be measured by the scale (2).

To calculate protein concentration in the serum the annex table is used; it establishes the correspondence between the refractive index values read from the Abbe refractometer scale and the quantity of proteins in g/l for the temperature of 17.5 °C.

Necessary materials and work process

- sodium lamp;
- ultrathermostat with the contact thermometer;
- two Pasteur pipettes;
- two test tubes for serology;
- the support of test tubes;
- 5 ml. of blopd serum;
- 5 ml. of distilled water;

– gauze;
– paper filter.

The exactness of the Pulfrich refractometer is verified before the proceeding:

– the sodium lamp is cpnnected up to the electrical net by the falling transformer;

– the ultrathermostat is also connected up to the electrical net;

– the work temperature is established at 17,5 °C by putting the contact thermometer in front of the respective division;

– several drops of distilled water are introduced by means of Pasteur pipette between the two prisms through the diaphragm of the prism armatures.

– the micrometer screw is fixed at zero;

– the ocular is rotated for establishing scale image clearance and demarcation limit;

– the number from the scale is noted, the limit between the light zone and darkened zone of sight field is situated in front of it; for distilled water it is 85,5. The micrometer screw determines decimals.

The obtained value for the limit angle of distilled water is 14.5, bicause placing of prisms needs the subtracting of value 85.5 from 100.

If the value of the limit angle for distilled water differs from 14.5 the apparatus is adjusted by a specialist or it is necessary to like into consideration the stated difference and the respective (Direction of the researched samples is made.

The verification of exactness is also made by distilled water at lie Abbe refractometer, and the value of the refractive index of Nter is read on the apparatus scale. At the temperature of 17.5 °C it imii be equal to 1.3332. The verification of the device is performed II follows:

– the refractometer body is rotated in the direction of the marked arrow on the device up to the extreme position;

– the prism body is opened by the screw (7). In this position he bright face of the prism of measurement is approximately horizontal. In the contrary case, the drum (5) is maneuvered. The surkces of both prisms are cleaned by a light wet rag soaked in ether / alcohol and they are left to dry;

– 1-2 drops of distilled water are put on the bright surface of the prism by a glass pipette with the rounded edge;

– the prism body is closed by the screw (7) and the refractlometer is brought to the working position. It takes several minutes to equal the temperature of the researched liquid with the ultrathermostat water. The temperature is read from the lateral thermometer (9);

– the mirror (8) is placed so that the illumination diaphragm must be equally illuminated and the field of the lunette 1 is bright;

– the separation line is brought to the intersection of lunette wires by the drum (5).

The water refractive indexes are read in this position if one looks through the lunette 6. If the apparatus is exact this value is the one indicated previously.

– the prisms are cleaned and the refractive index of blood serum is measured.

To determine proteins, fresh and nonhaemolysed serum is used. It is obtained by sampling 2-3 ml of venous blood into a cleaned test tube. The serum is eliminated by coagulation at the room temperature.

2-3 drops of serum are put between the Pulfrich or Abbe refractometer prisms, and the described method for distilled water is samely performed.

The values read by means of the Pulfrich refractometer are processed with the help of Table A19 from the annex to find the proteins.

The values of the refractive indexes read from the scale of the Abbe refractometer are transformed into concentration of proteins by means the respective table from the annex.

Bach student in the subgroup will perform three determinations written in the table below.

Table 12.1

The number of determinations	Scale division of the Pulfrich refractometer	The n_d value from the Abbe refractometer	Proteins (g%)
1			
2			
3			
4			
5			
6			
7			

The average value is calculated by making simple arithmetical average of the values of the proteins from of the same serum obtained from many readings.

The importance of refractometer method for medical practice

The refractometry is a relatively simple method and it allows determining the value of the refractive index with the precision of 10^{-4} order. The refractive index measurement can give information of the degree of a substance purity.

Taking into consideration the total volume of the emitted urine during 24 hours, the variations of the refractive index of the urine can give data about the purging degree of the body.

The modification of refractive indexes of refringence media of the eye produces different degrees of sight perturbation.

The refractometry remains the most frequently used method in clinical laboratories for measuring protein concentrations of blood serum (proteins).

The modifications of protein values give data about the evolution of an affection and the possibilities of recovery of the diseased body because reflects the protein metabolism at the level of blood serum proteins, whose normal value is 6–9 % and they have the values of refractive indexes values ranging between 1.3487 and 1.3517 respectively.

The description of the device

The examined installation for the demonstration of total reflection into a jet of water (fig. 12.6) consists of the following components: light source is He–Ne laser; the vase of Mariotte with the section of square form; protection screen;

The orifice of the from lower part of the vase is covered with a transparent organic glass stopper with parallel faces and is well polished.

Work process

The vase of Mariotte is filled with distilled water and placed in the way of red light flux irradiated by laser radiation, thus the light will penetrate into the vase perpendicularly to the opposite wall with respect to the diaphragm and go out through the cork ol organic glass. A light circle of red color is observed on the screen. In case of removing of the cork the direction of light flux is led by the water current into the trickling vase as a result of total reflection.

When the water level in Mariotte vase becomes lower than the end of the adjusting tube the water current is bent more abruptly, the conditions of total reflection disappear; the light leaves the water current making again the colored circle on the screen.

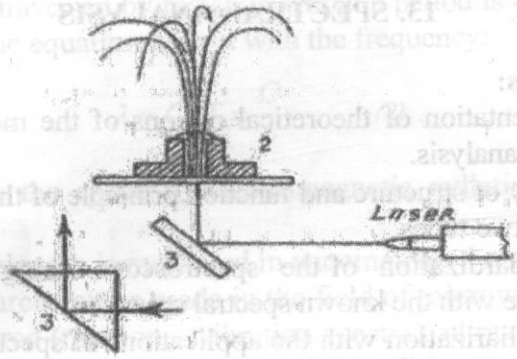


Fig. 12.6

A necessary optical guide for the performing the demonstration can be prepared with available means. From a photographic piece cleaned preventively of emulsion the narrow strips about 0.5 cm are cut wide. Each strip is warmed on the gas lamp till softening, then it is stretched rapidly, obtaining a thin glass fiber. Thus, 15–20 fibers of 1 are prepared and are linked into the frame 2 and polished (fig. 12.6)

The light flux irradiated by He–Ne laser passes by a prism of total reflection 3 through the section of the frame making a fountain with the bright points formed by diffused light from fibers ends.

Questions and tasks for self- control

1) Write the mathematical expressions and explain the notions: relative refractive index, absolute refractive index.

2) Explain the construction and principle of functioning of the refractometer based on the total reflection phenomenon.

3) What are the constructive elements and how does the device reflecting transmission of images and optical fibers operate?

4) What are the fields of practical applications of optical fibers?

13. SPECTRAL ANALYSIS

Purposes:

- Presentation of theoretical notions of the method of spectraln analysis.
- Study of structure and function principle of the spectroscope with two tubes.
- Standardization of the spectroscope using the radiation source with the known spectral structure
- Familiarization with the applications of spectral analysis in medical - biological research.
- Sodium found in biological products by means of emission spectral analysis.
- Determination of oxyhemoglobin spectrum.
- Visualization of rotation dispersion of the polarized light.

Theoretical notions

The spectral analysis is a method of physical analysis used for determination of chemical composition of different substances by studying their electromagnetic radiation spectrum accomplished by electronic transitions between different energetic levels or energetic transfer provoking the variation of rotation or rotation – vibration movement of molecules.

Radiation will be emitted on the transition of an atom or molecule from the upper energetic state E_2 into a low energetic state with the energy equal to the difference of energies of the two states:

$$E = E_2 - E_1 = h\nu \qquad (1)$$

Here н is the frequency of radiation, but **h** is the Planck constant (h=6.62 ·$10^{-34}$$T \cdot s$).

The road traversed by radiation within a period is called the wavelength λ. The equation links it with the frequency:

$$\lambda = C \cdot T = \frac{C}{\nu} \qquad (2)$$

Here c is the speed of electromagnetic radiation in the vacuum.

The wavelength is measured in nanometers (1 nm= 10^{-9}m)

The research can be made in the field of microwaves (rotation ipectra), infrared (rotation – vibration spectra), ultraviolet and visible (electronic spectra) according to the condition or substance.

Thus, we refer to the spectroscopy in the field of vision. The vision spectrum is represented by the radiation with the wavelengths ranging within 750 nm and 400 nm.

The phenomena of refraction and its decomposition into consecutive colored bands from the red to the violet when white light passes through the separation surface between two transparent media occur simultaneously. This phenomenon of decomposition of white light in the colors it is formed from is called dispersion. The dispersion takes place because the speed of light propagation in the same medium depends on the wavelength. As the wavelength decreases (from the red to violet) the speed decreases too and the respective index of refraction increases. The dispersion is better pronounced in the passage of white light through a prism of heavy glass giving an essential difference between the indexes of refraction for different wavelengths. The passage of beams through the prism is given in fig. 13.1

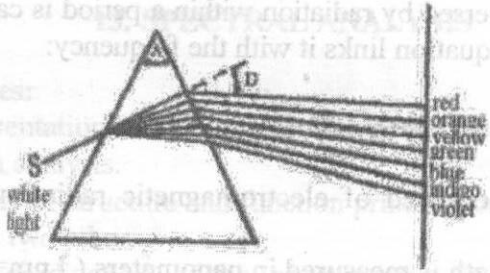


Fig. 13.1

While passing through the prism each of the monochromatic rays is deviated from the initial direction by the angle D, whose value for small incidences is calculated by the equation:

$$D = (n_1 - n_2)A$$
$$D = (n-1)A$$

Here n and A are the refractive index and angle of the prism respectively.

If the screen is placed against the rays dispersed by the prism, a colored image is obtained on it. It is called spectrum.

From the point of view of production, the spectra are classified into emissive spectra and absorption spectra.

The emissive spectra are obtained when the radiation emitted by the substance comes directly in the spectral apparatus.

The absorption spectrum is obtained when the white radiation is analyzed by the spectral apparatus, after traversing a transparent substance.

The emissive absorption spectra can be of a linear form, bands or continuous, they having a.particular structure that depends on the chemical composition and physical state of the researched substance. An emissive spectrum is characterized by the presence of bright lines (of different colors) placed on the dark background in comparison with the absorption spectrum which is determined

by the existence of some lines or dark bands placed on the background of the continuum spectrum. There is a direct correspondence conditioned by the Kirchoff law or the law of "inversion of spectral lines" between the emissive and absorption spectra obtained in identical conditions.

The spectral analysis can be qualitative or quantitative.

Each chemical element has a proper electronic structure and emits monochromatic radiation with the wavelengths strictly determined. From here results the possibility to identify a chemical element after the linear spectrum emitted - the qualitative spectral analysis.

The quantitative spectral analysis is based on the fact that the intensity of spectral lines is proportional to the concentration of the respective element.

To perform the spectral analysis the apparatuses called spectroscopes are used if the spectrum is studied by the naked eye or a spectrograph.

The mentioned apparatuses use the optical prism or the diffraction gradation as disperse elements. Both deviation and dispersion are inverse with respect to the wavelength on the optical prism. The optical gradation realizes the deviation proportional to the wavelength in comparison with the prism, as it is shown in fig. 13.2

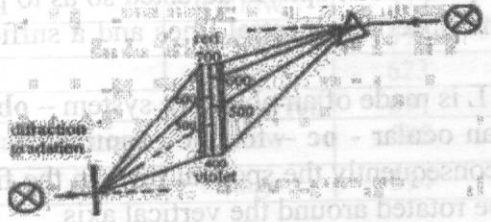


Fig.13.2

Description of spectroscope with two tubes

The spectroscope with two tubes is made of the optical prism P and two tubes - the collimator C and lunette L (fig. 13.3).

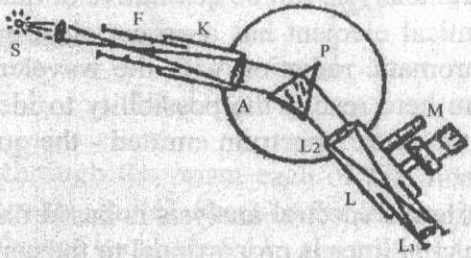


Fig. 13.3

The prism serves for obtaining dispersion of the light. Its composition must correspond to the field of work (usual glasses for the visible, quartz for the ultraviolet, salt gem for the infrared fields, in some cases the prisms with spectral compositions are used). The used spectroscope contains a prism of usual glass.

The collimator **k** is made of convergent lens and adjustable split placed in the focus of lens. The collimator is destined to send narrow beams of rays on the incidence face of the prism parallel to the lattice of the prism. The split is adjusted so as to provide a proper finesse of the observed spectral lines and a sufficient luminosity.

The lunette L is made of an objective system – **ob** – oriented to the prism, and an ocular - **oc** – for the examiner to look through them. To bring consequently the spectral lines in the field of vision, the lunette can be rotated around the vertical axis.

To obtain a clear image of spectrum the ocular is removed in the longitudinal axis of the lunette matching the examiner's eye.

a) Standardization of spectroscope

The standardization of spectroscope consists in establishing a dependence between the divisions of the scale and wavelengths of radiation. For this purpose sources emitting radiation with known wavelengths and spread all over the visible spectral field are used.

Each spectral line is fixed consequently against the indicator (dark triangle needle) registering the respective angle from the scale of spectroscope every time. The value of a division on the horizontal scale is 1°, and that of the scale of the drum is 0,02°. Then the curve of standardization is drawn on the millimeter paper by placing the divisions of the scale on the abscise of one rectangular axis, and on the other one – the respective wavelengths. The curve of standardization of the spectroscope is obtained by linking all the points in a continuous line. In our case we will use some of the emitted radiation by a luminescent lamp with the Hg vapors and a neon lamp, the wavelengths of which are represented in the table below.

Table 13.1

No of experience	Standard light source	The color of radiation	Wavelength (ran)	The division of the scale
1	Standard Hg	Violet	408	
2		Indigo	436	
3		Green	546	
4		Yellow	577	
5		Yellow	579	
6		Red	623	
7		Darkened red	691	
8	standard neon	Yellow	585	
9		orange	594	

The importance of spectral analysis for medical practice

The apparatuses of spectral analysis are frequently used in medical – biological research to study the chemical structure of various organic molecules. Aminoacids and nucleic acids present characteristic bands of absorption upon which the identification and their dosing in the solutions can be realized.

The spectral qualitative analysis is largely used in the field of medicine for identification of blood traces and establishment causes of different intoxications. These analyses are based on the fact that hemoglobin and its derivatives have characteristic absorption spectra.

The method of flamphotometry dosing of some alkali ions from biological products is a variety of spectral emissive quantitative analysis. Actually, in clinical laboratories spectrophotometers are needed strictly for identification of some substances and also for stating results of some laboratory reactions.

Equipment and materials

The spectroscope with two braces, sources of known spectrum, a luminescent tube with mercury vapors and a neon lamp, asbestos board mounted on a support for fringing to the incandescence of the researched product, research products in the liquid state.

b) Sodium in biological products

The used method is qualitative and it allows revealing the presence of sodium or other elements with the characteristic spectra in some liquid product.

In order to point out the presence of sodium in the researched products, its yellow line is identified (the double line D containing two very close lines): D_1 =589.6 nm and D_2=*589,0 nm doing* the following:

- the spirit lamp is lit;
- the researched product taken by the pipette is placed on the

asbestos board (for each researched product a respectively marked board of asbestos is used);

– the eventual spectra are studied by removing the lunette in the horizontal plane, and the value of each division of the scale of spectroscope is retained, corresponding to each distinctive spectral line;

– the presence of sodium is determined by matching the obtained spectra with those from spectrographic atlases,

c) Determination of oxyhemoglobin spectrum

As the darkened lines appearing in the absorption spectroscopy are received distinctly it is necessary to find a certain concentration called optimal concentration. A series of dilutions are made within which the position of the bands and the absorption intensity are studied. The absorption is represented graphically in dependence on concentration.

The dilution is made like this: 10 cm^3 of distilled water is poured into 10 tests tubes and the following quantities of blood are added: into the test tube 1–0.05 cm^3, the second one–0.08 cm^3, I he third one – 0.11 cm^3 and so on.

The test tubes with the diluted blood are interposed gradually between the natural light source (day light) and the slit of collimator. Two bands of absorption appear in the spectroscope (the darkened bands), one – in the green region and another in the yellow region, this being characteristic of oxyhemoglobin.

For each test tube the wavelength of these absorption bands is read and noted using the micrometer scale and the standardization curve. Totally, there are four values for each test tube. The obtained results are written in table 13.2

Table 13.2

The limits of oxyhemoglobin absorption bands

The number of test tube	Dilution	Yellow band		Green band	
		Left	Right	Left	Right
1					
2					
. . .					
10					

The chart (fig. 13.4) shows the values from table 13.2

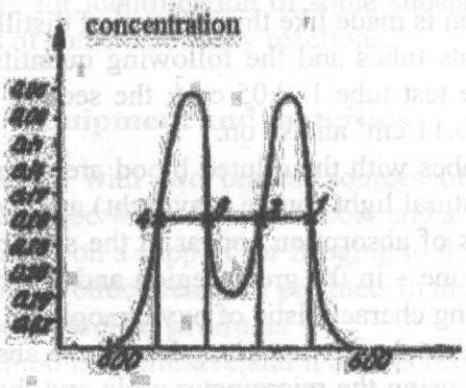


Fig.13.4

The optimal dilution in case of oxyhemoglobin is that which allows the determination of two distinctive darkened bands. This dilution will be used for practical determinations in the clinical laboratory (the optimal dilution for the oxyhemoglobin is approximately of 1%).

Questions and tasks for self-control

1) What is the meaning of dispersion phenomenon and what are the principal devices performing the dispersion of the light?

2) Draw correctly the passage of rays through the prism and write the mathematical expression for the determination of the deviation angle.

3) What do emissive and absorption spectra have in common and how do they differ?

4) What are the fields of practical application of spectral analyses?

14. INTERFERROMETRIC ANALYSES OF SOME GASES OF BIOLOGICAL INTEREST

Purposes:

- study of physical phenomena which are the basis of of interferrometers construction, for example, the Jamen refractometer;
- familiarization with the installation Chipp for obtaining CO_2;
- measuring for determination carbon dioxide percentage in a mixture of different gases.

Theoretical notions

The interferrometer is a measuring device of measurement based on the wave interference. There are two interferrometers for sound waves and electromagnetic waves (radio waves and optical waves).

The principle of functioning of all interferrometers is the same and it differs only by the method of obtaining coherent waves.

The light beam is divided spatially into two or more beams by certain means. Consequently they pass different optical distances, and then as the result of their overlapping an interference picture is observed.

The Jamen interferrometer applied for the determination of refractive indexes of gases and liquids can serve as an interferrometer with two rays (fig. 14.1)

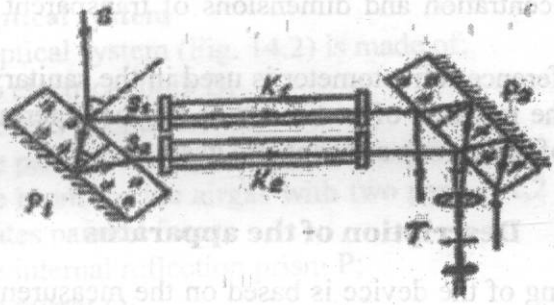


Fig. 14.1

After the reflection from both surfaces of the glass plate P_1 the light beam S is divided into two beams S_1 and S_2. Passing through the tubs K_1 and K_2 and reflecting from the surfaces of the glass plate P_2, the rays penetrate into the visual tube T, where they interfere making right fringes of equal bending. As one of the tubs contains the substance with the refractive index n_1, and another -the substance with the refractive index n_2, then after the removal of the interference picture (be it with **m** fringes) from the null position (when both tubs contain the same substance) the following equation can be determined:

$$\Delta n = |n_1 - n_2| = m\lambda / l$$

Where λ is the wavelength of the light

l – the length of the tub

The importance of medical biological research

The optical interferrometer is frequently used for the measurement of wavelengths, refractive indexes of transparent media, for the check of qualities of optical devices and others. The supply of the interferrometer with a double beam and a microscope called interference microscope is used in biology for measurement of dried

substance concentration and dimensions of transparent micro-objects.

The interference refractometer is used in the sanitary – hygienic field, in the research of environment for the determination of the presence of dangerous atmospheric gases.

Description of the apparatus

Functioning of the device is based on the measurement of the interference picture removal as a result of the composition variation of the researched gas present in the way of one of the two coherent rays. The value of removal is proportional to the difference between the refractive indexes of the researched gas and atmospheric air. The interference picture is symmetrical with respect to the light fringe of the zero order. The zero fringes are limited from the left and the right by darkened fringes with colored edges. The initial position of measurement is a position of the darkened picture from the right. The device scale is graded in volume portions in per cent. The value of one division is 2.5 % CH_4. In measuring the volume portion of carbon dioxide the scale for CH_4 is used, but the obtained result is multiplied to the factor of correction equal to 0.95. The internal part of the device is divided into three sections. In one of them the optical details are placed. There is a wrapped coil with a tube of polychlorvinyl in the second part. There is also a fixed electrical element here.

The absorption chuck is in at the third section. The connecting piece is also present here and a rubber pear - shaped tube is linked to it. This tube is used for pumping pure air into the respective tub. After pumping, the connecting piece is closed by a rubber cork. The chuck with the electrical illumination bulb is fixed on the lower part of the section.

a) Optical system

The optical system (Fig. 14.2) is made of:

- the electrical bulb B;
- the lens - condenser K;
- the parallel faces plate (the mirror) O;
- the room for the airgas with two parts – 1,2 limited by the plates parallel faces;
- the internal reflection prism P;
- the mirror O_1
- the observing tube with the objective OB, ocular OC and slit – diaphragm with the measurement scale S.

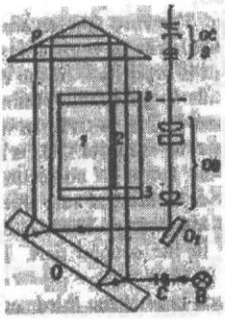


Fig. 14.2

The passage of rays during measurements is represented in fig. 14.2; the beams creating the interference picture pass through different tubs (the tub 1 contains atmospheric air, but the tub 2 - the researched gas). Although the length of the optical way of interference rays is the same for the given case, the interference picture remains stable, independent of gas nature in the tub.

The null position is established by the upper mobile lens of the objective giving the possibility to remove the interference picture and fix it in the null position of the measurement scale.

b) The scheme of airgas

The scheme of airgas of interferrometer consists of two separated ducts - for the researched gas and for air. The duct for the gas contains:

– the tap for distribution intended for the variation of the moving direction of gas mixture depending on the nature of the researched substance (methane or carbon dioxide);

– the absorption chuck is separated in two parts. One of them contains an absorbent for carbon dioxide and the other – an absorbent for water vapors. Both parts of the absorption chuck have antidust filters and they are separated by a valve;

– linking rubber tubes;

– the gas section of the airgas room;

– the gas section of the airgas room of air duct.

Devices and materials

1. Interferrometer.
2. Chipp device.
3. A tub.
4. Pieces of marble.
5. Hydrochloric acid.
6. Barometer.
7. Thermometer.

Experimental process

Before performing the measurements with interferrometer it is necessary to check the null maximum position of the interference picture. In order to perform this the buttons I and K are pressed simultaneously (fig.14.3), looking at the position of the interference picture through the visor. If the black fringe from the right is not removed from zero, then the device is ready for work.

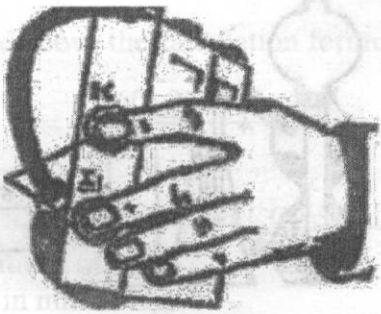


Fig. 14.3

If the black fringe is removed from zero, it must be removed to zero and then the device is ready for work. When a measurement is performed, at first the button I must be pressed.

In order to measure the volume portion of carbon dioxide from the given volume it is necessary the tap to be in the position "CO_2", and then the gas mixture must be pumped into the interferrometer. As the air contains CO_2, the interference picture is removed to the right of the scale. Looking through the ocular at the removal of the black fringe from the right of the interference picture, we read the indications of the device knowing that each division is 2.5%.

In order to perform measurements the installation is mounted according to the scheme in fig.14.4 where all the necessary parts are fixed.

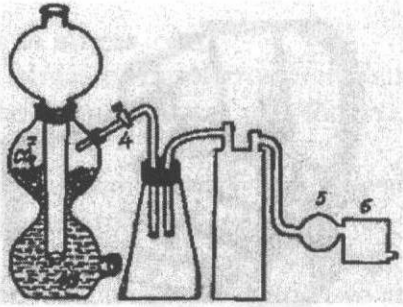


Fig. 14.4

1 – chipp device for obtaining $C0_2$;
2 – tub;
3 – interferrometer;
4 – tap;
5 – rubber switch;
6 – pump.

When opening the tap 4 (fig. 14.4) the hydrochloric acid from the Chipp device comes into contact with the marble pieces. As a result of the chemical reaction CO_2 is obtained. When closing the tap 4 the pressure in the volume 7 increases leading to disconnection of the acid from the marble pieces and the reaction is discontinued. There is **a** mixture of CO_2 and air in the tub 2. The mixture of the gases passes into the interferrometer 3 by means of the pump 6 with the rubber pear 5 and then the apparatus indications are read.

Representation and processing of experimental results

To determine the volume portion of CO_2 in per cent, the calculation formula must correspond to the conditions: 20° C and 760 mm of Hg. In case the temperature and pressure are different

from those mentioned above the calculation formula is as follows:

$$I_a = I \cdot \frac{760}{p} \cdot \frac{273+t}{293} \cdot 0.95$$

Where I_a is the volume portion of CO_2 calculated in %;

I – device indications in %;

P – the pressure in mm of Hg;

T – the temperature in °C

The experimental results are written in the table:

The number of experiment	t,°C	p, mm of Hg	I, %	I_a, %
1				
2				
3				

Questions and tasks for self-control

1. What are the main physical phenomena that are the basis of construction of interferrometers?
2. What are component elements of the installation for producing $C0_2$ in laboratory conditions?
3. Draw an optical scheme of the Jamen interferrometer.
4. What are medical-biological fields of application of the interferrometer method?

15. DETERMINATION OF CONCENTRATION OF OPTICAL ACTIVE SUBSTANCE

Purposes:

- study of phenomena and some methods forming the basis of obtaining polarized light;
- familiarization with the possibilities of using polarized light in biology and medicine;
- study of construction and work principles of polarimeter;
- use of polarimeter in the study of optically active substances, particularly in determination of sugar concentration (or glucose) in different biological solutions.

Theoretical notions

The vector of the variable electrical field intensity E and the vector of the variable magnetic field B characterizing the electromagnetic wave are inseparable. These vectors oscillate in reciprocal perpendicular planes containing simultaneously the direction of wave propagation. The electromagnetic waves the wavelengths of which are enclosed in the interval from 10^6 nm up to 10 nm make the optical spectrum. A small portion from this area (750 nm – 400 nm) perceived by human eye represents the visible spectrum or simply the light.

The light having the electromagnetic nature possesses both wave properties and corpuscular properties. The wave properties are manifested especially in the propagation phenomena (reflection, refraction, interference, diffraction, polarization, etc.), but the corpuscular ones – in the emissive phenomena or absorption (photoelectric effect, the Compton effect, etc.).

Chemical and biological action of the light, particularly the light action on the eye, is connected by the electrical component of the electromagnetic wave. Therefore, the vector E of the variable electrical field intensity from the electromagnetic wave is called the light vector and is considered the basic feature of the ligh.

The plane in which the oscillations of light vector takes place is called the polarization plane (variations under the value and direction).

In every source of light waves are emitted by milliards of atoms oriented chaotically and, therefore, the oscillations of the light vector are made in different planes (fig. 15.1).

Such a light is called nonpolarized. It follows the natural light and the one emitted by artificial sources is nonpolarized. The light in which the oscillations of light vector takes place only in a plane is called the plane polarized or linear polarized light. The plane in which the oscillations of the light vector are made in the plane-polarized light is perpendicular to the direction of propagation of light and it contains this direction in itself. If the oscillations of light vector take place strictly in a single plane, the light is totally or completely polarized; and otherwise, the light is partially polarized.

The polarized light can be obtained by many methods: a) by the reflection of light from the surface of the medium; b) by refraction of light in the transparent medium; c) by birefringence - double natural refraction – the phenomenon which takes place both in the anisotropic substances from the optical point of view and by artificial birefringence obtained in some isotropic bodies subject to some mechanical deformations, or in some solids, liquids or gases under the action of the electrical field (Kerr effect) or in some liquids under the influence of an intensive magnetic field (Cotton -Mouton phenomenon); d) by diffusion in some solutions (Raleigh); c) by absorption followed by reemission at the particle levels with a small ray in relation to the wavelength (Tyndall phenomenon), etc.

Both reflected and refracted lights are partially polarized; oscillations in the reflected ray take place in the incident plane, but in the refracted ray – in the plane perpendicular to the first one. (fig. 15.1) The polarization degree of the reflected light depends on

the incident angle. If the incident angle i satisfies the condition:

$$tgi = n \qquad (1)$$

where **n** is the refraction index of medium, from the surface of which the light is reflected, the total polarization of the reflected light takes place (the law of Brewster). According to the Brewster law the refracted ray is partially polarized and the polarization degree is maximal. Applying the formula (1) and the law of refraction, it can be demonstrated that at the total polarization of the reflected light the angle between the reflected ray 2 and the refracted ray 3 is equal to 90° (fig. 15.1).

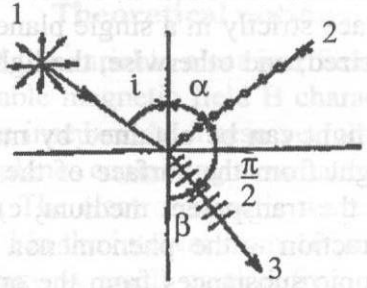


Fig. 15.1

The total polarization takes place also as a result of birefringence when the light passes through some transparent crystals *from the optical anisotropic active substance. The spat of Iceland* (carbonate of calcium) and quartz are well known among numerous birefringence crystals.

The birefringence is caused by optical anisotropy of some substances expressed by the property of these transparent substances to transmit the light in different ways and directions. If a ray of light falls on such a crystal it is doubled after the refraction. For one of them the laws of refraction works and therefore, this ray is

called ordinary, for the other one these laws do not work and the ray is called extraordinary. Both rays are totally polarized and have reciprocally perpendicular polarization planes. The oscillations of light vector in the ordinary ray take place perpendicularly to the principal plane of the crystal, but in the extraordinary ray - in the principal plane. The plane containing the optical axis and incident ray is called the principal plane of the crystal. The direction along which the birefringence does not occur and the ordinary and extraordinary rays are propagated with the same speed is called the optical axis of the crystal.

The devices (optical apparatuses) in which the light is polarized are called the polarizers. The devices used to study (to analyze) the polarized light are called analyzers.

The polaroid prisms nicol are used in practice for obtaining polarized light and analyzing it.

The nicol is a prism (fig. 15.2) made of anisotropic crystal, for example the spat of Iceland divided into two parts after the short diagonal and stuck together with the transparent glue, for example, balm of Canada, whose refraction index has the value ranging between the values of the refraction indexes of the Iceland spat for the ordinary and extraordinary rays. Choosing the angles of prism by the respective means allows to make the ordinary ray undergo the total reflection on the separation surface but the extraordinary one pass through the second part of the prism and go out from the nicol in parallel with its bottom surface.

The polaroid is a transparent film of celluloid containing a great number of small crystals of anisotropic dichroism substance samely oriented, for example, herapatitis (quinine iodsulfate) that polarizes the light and absorbs completely one of those two rays (usually the ordinary one).

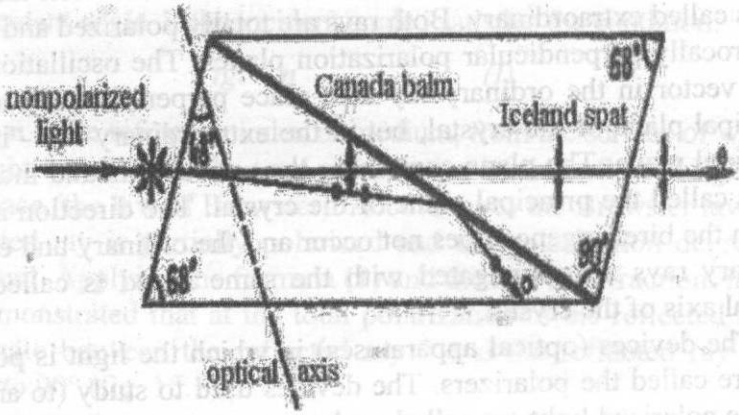


Fig. 15.2

If the second nicol N_2 is placed in front of the polarized ray going out from the first nicol N_1 (fig. 15.3, a), so that their principal planes are parallel, the light ray passes through this nicol. By rotating the second nicol we change the intensity of the light passing through it. It becomes null (fig. 15.3, b) when the angle formed between the principal planes of nicols is 90° (the nicols are crossed). In this case N_1 is a polarizer but N_2 - analyzer.

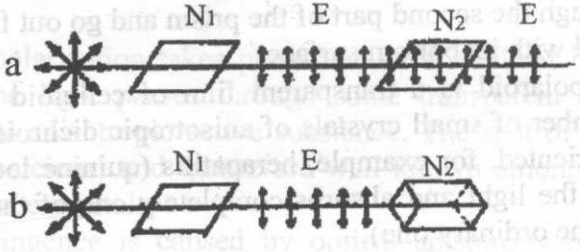


Fig. 15.3

The importance for medical practice

Living organisms are made to a great extent of optically active substances (carbon hydrates, albumin and their aminoacids, hormones, etc.). In order to study these substances the polarized light is usually used, therefore, the polarized light is applied on a large scale in biology and medicine, particularly in determination of optical activity of serum albumin with the purpose to make the diagnosis of cancer or in clinical practice to determine glucose or albumin concentration in the urine of diabetis patients.

The polarimetry is applied not only for determination of solution concentration, but as a method of studying structural transformations, particularly in molecular biophysics.

The polarizing microscopy serves to determine isotropy and optical anisotropy of different histological elements as well as in verification of bone plates, collagen fibers, chromatin, myelin, nervous fibers, cartilage, and darkened discs of muscular fibers.

The polarized light can be applied in studying molecules for applying mechanical tensions appearing in bone tissues.

The apparition of life needs natural syntheses of active optical compounds. Actually, in the laboratory optically active substances can be obtained out of action of living organisms only by photosynthesis in the circular polarized light.

Determination of sugar concentration in solutions by saccharimeter

Some optical transparent substances, through which the polarized light passes, have the property to rotate its polarization plane. Such substances are named optically active, and their property to rotate the polarization plane of the light – optical activity. Quartz crystals possess a property, in the direction of which the birefringence, for example, chlorate of potassium do not take place.

The rotation angle φ of polarization plane of the polarized light while passing through the crystals, is directly proportional to the thickness l of the crystal crossed by the light:

$$\varphi = [\alpha] \cdot l \qquad (2)$$

The coefficient [α] is characteristic of the examined crystal and is called specific rotation power (or the specific constant of rotation) of it and is expressed in grad/mm. The specific rotation power of crystalline substances is numerically equal to the rotation angle of the polarization plane referring to a millimeter of the distance crossed by the light ray.

Some substances are optically active in the solutions, for example, sugar solution in water. In this case the angle φ, with which the optical active substance rotates the polarization plane of polarized light, is proportional to the thickness l of solution column and the concentration C of the substance in the solution:

$$\varphi = \frac{[\alpha] C \cdot l}{100} \qquad (3)$$

In this formula l is expressed in dm, but C in % that is in grams per 100 cm^3 of solution.

The property of rotation of the polarized plane of the light is explained by the asymmetrical structure of molecules (in liquids) or of optical active crystals (in solids). Due to the molecular property the angle of rotation increases proportional to the number of molecules met by the light polarized beam, from which the dependence on contraction and crossed thickness layer respectively results.

The specific rotation power of optically active substance is determined conventionally at the temperature of 21°C and for the light wavelength $\lambda = 589.4$ *nm* (the yellow line D of sodium flame).

The specific rotation power [α] of optically active substance is equal numerically to the rotation angle increased by 100 times of the polarization plane of polarized light by the solution column with the thickness of 1 dm, the concentration of the substance being of 1 g in 100 cm^3 of solution at t=21°C and for λ= 589.4 *nm* – physical sense of *[α]*.

The specific rotation power *[α]* of optically active substance depends on the nature of substance, its temperature and the light wavelength, passing through it (inverse proportional to the square wavelength). One and the same optically active substance rotates the polarization plane of different wavelengths with different angles because the specific rotation power depends on the light wavelength λ. For example, a stratum of sugar solution with the thickness of 1 dm and the concentration of 1 g of sugar in 1 cm^3 of the solution rotates the polarization plane of the red light *(λ = 656nm)* by 66.5° and that of the green one *(λ = 535nm) – by* 821. Therefore, the light is decomposed in its components while white light passes through the stratum of optically active substance. This phenomenon is called disperse rotation.

Some optically active substances possess the right rotation, the others the left one. The substances rotating the polarization plane of the polarized light to the right (in the clockwise direction, if we look in the opposite direction of the light propagation) are named clockwise, but the substances rotating the polarization plane to the left are called counterclockwise. A lot of optical active substances exist in two varieties – clockwise and counterclockwise. Quartz can be an example.

The principle of method

The law expressed by the formula (3) for the optically active substances gives a rather precise method of determination of sugar or glucose concentration in the solution and allows distinguishing molecules of the same substance with different configuration of atoms (the stereoisomery).

Having a solution with known concentration C (standard solution), we can determine the specific rotation power of the given substance, resulting from the formula (15.3)):

$$[\alpha] = \frac{100 \cdot \varphi}{C \cdot l} \qquad (4)$$

The specific rotation power [α], according to the formula (4) is expressed in $\frac{grad \cdot cm^3}{g \cdot dm}$ if the concentration C is expressed in $\frac{g}{cm^3}$, or in $\frac{grad}{\% \cdot dm}$ if the concentration C is expressed in percent (%).

For sugar $[\alpha] = 66.5 \frac{grad \cdot cm^3}{g \cdot dm}$, or $[\alpha] = 66.5 \frac{grad \cdot cm^3}{\% \cdot dm}$ but for glucose $[\alpha] = 52.8 \frac{grad \cdot cm^3}{g \cdot dm}$.

Knowing the specific rotation power of the given optically active substance and the length of the tube, in which the solution of this substance is poured, we can determine the unknown concentration C_x of the substance in the solution by means of the formula:

$$C_x = \frac{100 \cdot \varphi_x}{[\alpha] \cdot l} \qquad (5)$$

In order to simplify the calculations while determining glucose concentration in the urine by the polarimeters (called urometers), the length of the tube 1 is chosen so that the angle φ with which the glucose rotates the polarization plane of the polarized light (in degrees) should be numerically equal to the concentration

C of glucose in percent (in grams at 100 cm^3 of solution), thus:

$$C_x = \varphi_x \qquad (6)$$

Substituting in (21.5) the equality (21.6) and the value *[α]* for the glucose in the yellow light, equal to 52.8 we obtain:

$$l = \frac{100}{[\alpha]}\,dm = \frac{100}{52.8}\,dm = 1.89dm \quad (7)$$

Writing the formula (21.3) for the solution of the researched optically active substance with the unknown concentration and for the standard solution with the same optically active substance, we obtain:

$$\varphi_x = [\alpha]\frac{C_x \cdot l}{100} \qquad (8)$$

$$\varphi_s = [\alpha]\frac{C_s \cdot l}{100} \qquad (9)$$

Dividing the equality 15.9 to that of 15.8 the following relation is obtained

$$\frac{\varphi_x}{\varphi_s} = \frac{C_x}{C_s} \qquad (10)$$

From it we obtain the formula that gives the possibility to determine the concentration of optically active substance by the relative or indirect method, knowing the concentration of the standard solution of the same optically active substance:

$$C_x = C_s \frac{\varphi_x}{\varphi_e}$$

This method needs certain conditions:

a) The temperature must be constant during the work.

b) The same tube must be used to assure the same thickness of the researched solution stratum and that of standard solution respectively.

c) The standard solution must contain the same optically active substance as the researched solution.

d) The tube of polarimeter must be washed perfectly with distilled water between the performed determinations with the researched solution and with the standard solution respectively.

e) The monochromatic light must be used to avoid the disperse rotation appearing in the case of white light due to a lot of wavelengths.

Description of apparatus

The method of polarized light used in the qualitative and quantitative analysis of different optically active substances is called polarimetry, but the devices used for the measurement of the angle rotation of the polarization plane of the polarized light are called polarimeters.

The optical scheme of the polarimeter is given in fig. 15.4.

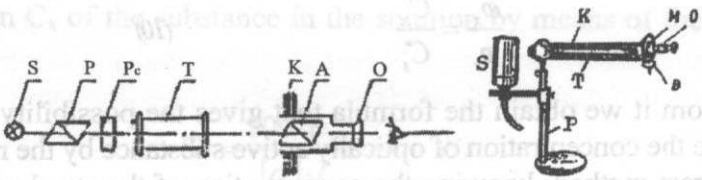


Fig. 15.4

The simplest polarimeter consists of two identical nicols: the polarizer P and the analyzer A. The optically active substance is introduced in the tube T. The rotation of the analyzer necessary for restoring the same field (usually uniformly darkened field) establi-

shed in the lack of the substance will give us the angle with which the optically active substance rotated the polarization plane of the polarized light.

In some polarimeters polaroids are used as polarizers and analyzers because of high cost of nicols. Polarimeters used for determining sugar concentration in a solution are called sacchari-meters.

The medical saccharimeter of SM type (fig. 15.4) consists of the support P, on which the source of light S and the metal tub K arc fixed. The optical system of the device (fig. 15.5) consists of the yellow filter of light F, the polaryzer P, the tube T with the standard solution or the researched solution, the analyzer A with the circle dial M and a vernier (circular vernier calipers) for reading angles, the mobile muff B, the ocular O and lunettes L for reading indications on the dial and vernier. In this saccharimeter polaroids as a polarizer and analyzer are used. The optical properties of the light filter and polarizer are combined so that in the obtained polarized light the maximum of intensity will correspond to the yellow line D from Na spectrum.

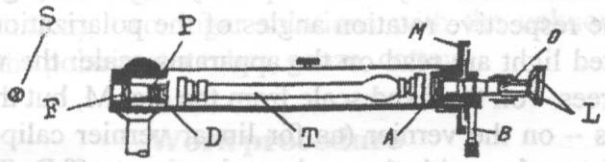


Fig. 15.5

The optical system of the device also includes the diaphragm D with a lunette of quartz allowing to perform photometric reading of angles with a higher precision. The quartz plate is placed in such a position that only the rays from the central part of the light beam will pass through it. Besides this, the polarization plane of the polarized light is rotated. It goes out from the polarizer with

$5 - 7^{\cdot 0}$. Thus, looking through the ocular O, we can note that at the crossing of the analyzer A and polarizer P (their principal planes being reciprocally perpendicular), the vision field is darkened in the lateral parts, but in the middle it is illuminated. So, the visual field is divided into three parts (fig. 15.6).

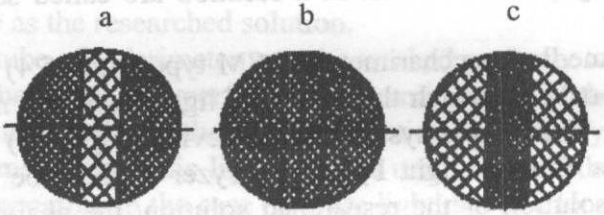


Fig. 15.6

By rotating the analyzer in some direction the illumination of the middle part of the vision field can be weakened, concomitantly the illumination of the lateral parts increases. In this case a triple field is also obtained, but the middle part is more darkened, and the lateral parts more illuminated (fig. 15.6, c). It is evident that we can find such a position of the analyzer at which the visual field will be more uniformly and completely darkened (fig.15.6,b).

The respective rotation angles of the polarization plane of the polarized light are read on the apparatus scale: the whole number of degrees – on the fixed scale from the dial M, but the decimals of degrees – on the vernier (as for linear vernier calipers), which is rotated together with the analyzer by the muff B. The reading is made by looking through one of the lunettes situated in the vicinity of the ocular.

The integral number of degrees is read on the fixed scale from its zero to zero on the vernier, but the number of decimals or hundreds (0.05) is indicated by the ordinal number of hachures (dash) from the vernier, coinciding with some hachure of the fixed scale. For example, in fig. 15.7 the position 2.65° is indicated.

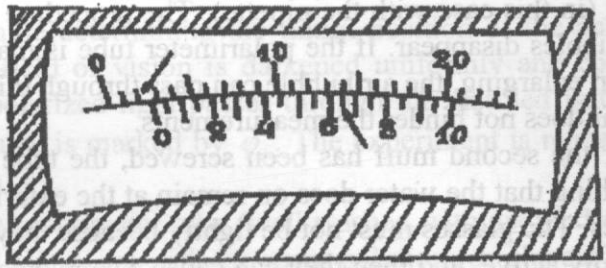


Fig. 15.7

The vernier allows determining the rotation angle of the analyzer with the accuracy of 0.05°, because it is graded so that 20 of its divisions are equal to *19* divisions of the fixed scale, so that each division of the vernier is smaller by $\frac{1}{20} = 0.05$ as the division of the fixed scale.

Equipment

Saccharimeter, sugar solution with the known concentration (standard solution), two sugar solutions with the unknown concentrations, the polarimeter tubes, gauze, cotton.

Work procedure

1) The polarimeter tube is prepared for work. The muffs with filet are discrewed, the glasses are taken out and are well cleaned on two sides with the cotton soaked in distilled water, then with the dry cotton (or gauze). One end of the tube is closed, but through the other one the distilled water is poured till a convex meniscus is made. The glass is placed carefully over the meniscus, so that the water from the meniscus will be removed and no air bubbles remain under the glass. If the air bubbles remain, a little water

is poured (in this case with the pipette). The procedure is repeated till the bubbles disappear. If the polarimeter tube is made of glass and has an enlarging, the air bubble can pass through this enlarging and then it does not hinder the measurements.

After the second muff has been screwed, the tube is cleaned out, watching that the water does on remain at the exterior parts of the glasses. The glasses must not be tightly because they can become optically active, and then they can cause a supplementary rotation of the light polarization plane, so, it can result in measurement errors.

2) The tube filled with distilled water is placed in the tub of the saccharimeter between the polarizer and the analyzer and is covered by the lid of the tub.

3) The plug is set on and the switcher in the circuit connects the electrical light bulb.

4) The clearance of the treble vision field is established (Fig. 15.6, a or c) by fixing the saccharimeter for each observer's eye, that is, it can be obtained by moving the mobile ocular O to the observer or from him.

5) The muff B (fig. 15.4) rotates the analyzer in one direction or another till such a position of the analyzer is established, that the field of vision is darkened uniformly (minimum illumination) (fig. 15.6, b). In this case the rotation angle of the polarization plane of the polarized light must be equal to zero, so that the zero of the vernier must coincide with the zero of the basic scale, because the distilled water is optically inactive and does not rotate the polarization plane of the polarized light, passing through it. If this coincidence does not take place, then it means that the device is fault, it can be used in the work, but the initial indications of the vernier calipers must be taken into consideration. This is the initial position of the analyzer. The initial position of the analyzer is often determined when the polarized light passes not through the distilled water but through air.

The angle determining the initial position of the analyzer, at which the field of vision is darkened uniformly and completely, when the polarized light passes through the distilled water (or through the air), is marked by φ_0. The experiment is repeated several times (3–5).

6) The polarimeter tube (well washed with distilled water and dried) is filled at first with the sugar solution (or glucose) with the known concentration C (standard solution), then with sugar solution for research. The concentrations C_x of those solutions must be determined avoiding the formation of air bubbles.

7) Before beginning the measurements for each solution, the clarity of treble field of vision is established (fig. 15.6, a or c).

8) The angles φ_s' and φ_x' are analogically determined 3-5 times.

This determines the position of the analyzer, for which the field is darkened uniformly and completely in case when the polarized light passes through the standard solution and the solutions with the unknown concentrations respectively.

9) The values φ_s and φ_x are determined 3-5 times by the formulae below, that are the angles for which the standard solution and solutions with the unknown concentrations rotate the polarization plane of the polarized light passing through these solutions:

$$\varphi_s = \varphi_s' - \varphi_0 \qquad (12)$$

$$\varphi_x = \varphi_x' - \varphi_0 \qquad (13)$$

10) Knowing the values of the angles φ_s and φ_x, the unknown concentrations C_x of sugar solutions (or the glucose) are determined by the formula (11).

11) The obtained results are given in table 15.1 and on their basis the proper conclusion is done.

12) Point 10 is repeated for one and the same researched sugar solutions, taking a tube with other lengths and on the basis of the obtained results the second conclusion is done.

The method described in this paper can be used in clinical laboratories, where a glucose solution with known concentration is taken as a standard solution, but a pathological urine is taken as a researched solution, i. e. the urine containing glucose.

Table 15.1

The number of researched solutions	The number of experiments	C_x, %	φ_0, degree	φ'_s, degree	$\varphi_s = \varphi'_s - \varphi_0$ degree	φ'_x, degree	$\varphi_x = \varphi'_x - \varphi_0$ degree	Cx, %
	1 2 3							
	1 2 3							

Questions and tasks for the self-control

1) As a result of what phenomena and by what devices the polarized light can be obtained? Write the mathematical expression of the Brewster law.

2) Draw an optical scheme of the polarimeter. Indicate the role of each constructive element.

3) What substances are named optically active? Explain the notion of the specific rotation power?

4) What are the medical biological fields of polarized light application?

5) What are the constructive elements and working technique of the didactical device for proving the existence of two modifycations of optically active substances?

16. DETERMINATION OF WAVELENGTH AND ENERGY OF LASER RADIATION QUANTUM

Purposes:

- study of phenomena that are the basis of laser functioning;
- study of the principle of laser He – Ne functioning;
- determination of wavelength and energy of quantum of laser radiation by means of diffraction grating;
- familiarization with the laser effects in the interaction with the substance and application of this radiation in medicine.

Theoretical notions
Spontaneous emission and inductive emission

According to the laws of quantum mechanics, the energy of electron, and that of atom, to which it is connected, can have only the discrete values E_1, E_2, E_3, ... called the energetic levels.

The transition of atom (or molecule) from one superior energetic state (for example, E_3) to a smaller energetic state (E_2) takes place on the emission of one energetic quantum. The equation determines the frequency of the emitted quantum:

$$\nu_{32} = \frac{E_3 - E_2}{h} \qquad (1)$$

Here **h** is the Planck constant.

The inverse process is produced at the absorption of one energetic quantum: the transition of the particle from a smaller energetic state (for example, E_2) to a higher energetic state (E_3) takes place. The frequency of the absorbed radiation is ν_{23} equal to ν_{32}.

The quantum of energy can be emitted in the environment as a photon or thermal energy. In the first case, the transition takes place with emission, in the second one – without emission.

The emission of energy quanta can take place in two processes. The first consists of the spontaneous transition of excited micro-particles from high energetic levels to levels of inferior energies with emission of radiation. Self-emission is determined by the internal interaction and does not depend on external fields applied on the quantum system. The emission produced as a result of self-emissive transitions is called spontaneous emission (fig. 16.1).

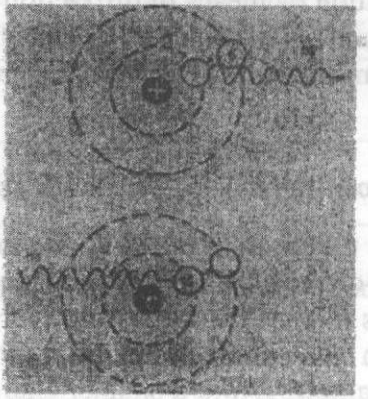


Fig. 16.1.

The spontaneous radiation has an aleatory character and presents a mixture of quanta with different frequencies and directions of propagation.

The second process of emission takes place under the action of external electromagnetic fields. The radiation born in this case is called inductive emission or stimulated emission. The hypothesis about the existence of induce radiation was formulated by Einstein in 1917.

In case of stimulated emission, the newly born quanta do not differ from the quanta provoking the respective transitions. They have the same frequency, phase and direction of propagation (fig. 16.2).

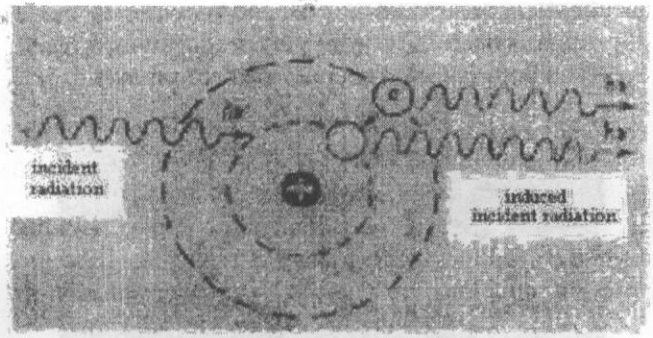


Fig.16.2.

The phenomenon of stimulated emission The mechanism of population inversion of energetic levels.

To obtain an increase of the number of particles on a higher level a series of methods are used.

We will study the method used in the gas lasers to which the He – Ne lasers belong. In the Equilibrium State the population of energetic levels is determined by the Boltzmann distribution, drawn dotty in the graphic from Fig.16.3, delimited by the function $\left(-\frac{E}{KT}\right)$. The system of the examined microparticles is chosen only with three levels. The radiation from the exterior ν_{31} allows the transition of atoms from level 1 to level 3.

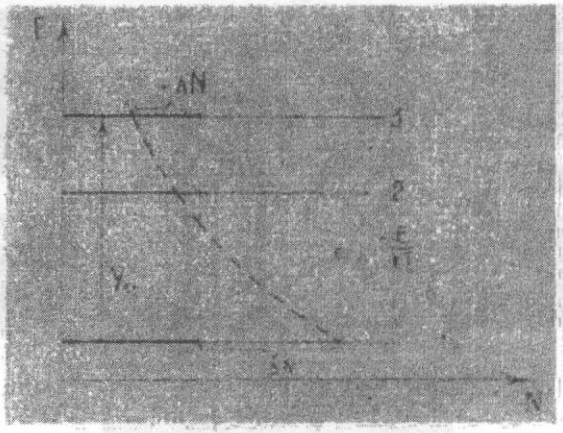


Fig. 16.3.

If the power radiation from the exterior is sufficiently great, then by the transition $1 \rightarrow 3$ the saturation will take place and then the populations of levels 1 and 3 can be considered equal (the population of level 1 falls to the value $-\Delta N$, but for the level 3 it increases by $+\ \Delta N$). In this way the inversion of population of the level 3 and 2 is obtained.

To obtain inversion of population between two energetic levels is easier if the electrical discharge in the mixture of two gases is used (be it A and B), respecting a number of requirements, so the base gas A and the added B must posses a level, whose energy coincides or is close, so:

$$\left|E_3^B - E_3^A\right| \leq KT$$

The energetic diagram of the gaseous mixture is given in fig.16.4

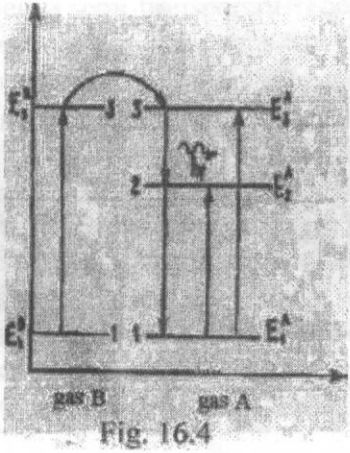


Fig. 16.4

Neon atoms play the role of the basis gas A but the role of added gas B belongs to the atoms of helium in the neon-helium laser. The first source of excitation of quantum systems consists of the electrons appearing at the electrical discharge through gases.

By means of collisions with the excited molecules the basic gas and the added one, the electrons produce the population of the levels 2 and 3 of gas A and level 3 of gas B.

The density of excited molecules from gas B is increased by increasing the discharge current. As a result of collision the transfer of energy from gas B to gas A takes place, obtaining the population of the superior level 3^A. The condition of inverse population is created between the levels 3^A and 2^A.

For each mixture of gases an optimal current of discharge exists in which the high inversion of population is assured. The typical dependence on inversion of population in a mixture of gases as a function of current value is given in fig. 16.5.

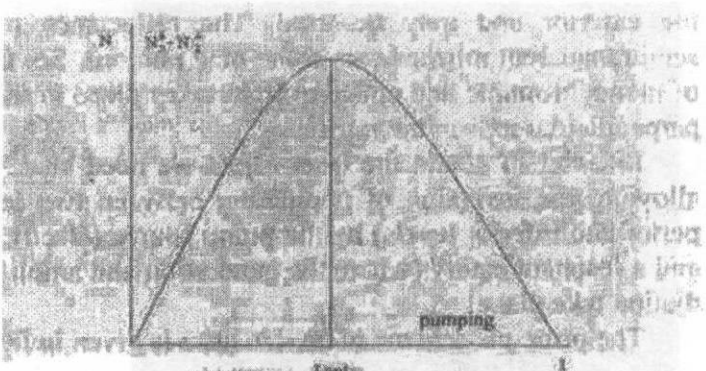


Fig. 16.5.

Different types of mixtures of gases can be used, their mechanism of excitation is near to that described. These mixtures are helium and neon, helium and xenon, neon and oxygen.

The amplifying process

The active medium with the inversion of population between two given energetic levels is able to amplify the electromagnetic radiation propagated through it when its frequency coincides with the frequency of quantum transition between energetic levels, in which the inversion of population exists. So, an active medium in this condition plays the role of quantum amplifier.

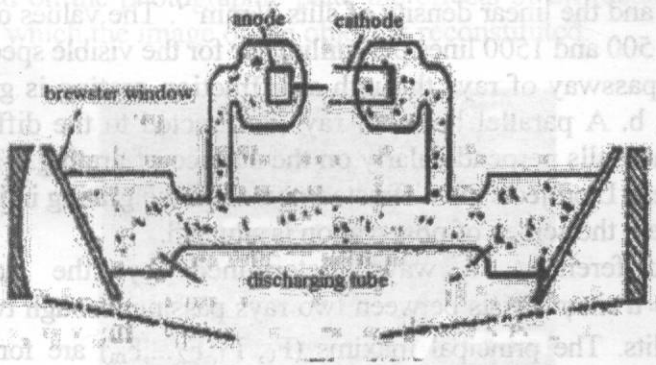
The condition of self-oscillation. The realization of population inversion is a necessary condition, but not sufficient for the laser effect to take place. It is also necessary to perform the so-called condition of self-oscillation.

For the light to lead the emission of atoms, it is necessary that a part of emitted radiation remained in the interior of the cavity leading to the inductive emission of new atoms. The mirrors perform it. The working substance is placed between plane parallel

mirrors, one of them being semi transparent. The light wave emitted in one place as a result of spontaneous transition is amplified on the account of induced radiation at its propagation through the working substance. Reaching the semitransparent mirror it partially passes in the exterior and may be used. The reflectance part from the semitransparent mirror bears some new photons. So, the generation of monochromatic and coherent light takes place in the direction of perpendicular axis to the mirrors.

In order to obtain the laser effect we need an active medium, allowing the inversion of population between two levels (the superior and inferior levels) by the pump source (electrical discharge) and a resonant cavity (where the generation and amplification of radiation take place).

The principle scheme of He-Ne laser is given in fig. 16.6.



Diffraction grating

A diffraction grating (fig. 16.6, a) is a system of parallel slits of the equal size **a** separated by opaque spaces of size **b**.

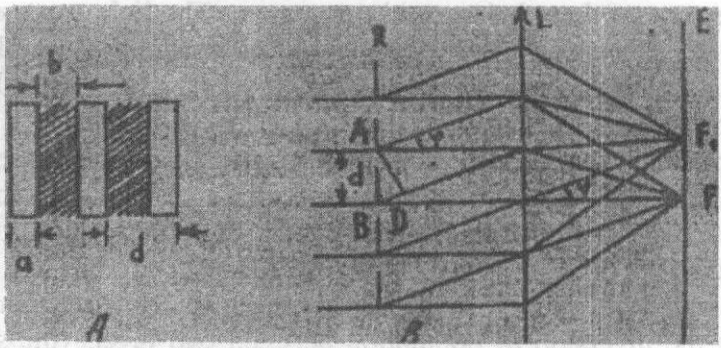


Fig. 16.7

In order to perform a grating a series of parallel rectilinear scratches is drawn by a mechanical device of high fines on the surface of glass. A diffraction grating is characterized by the constant $d = a + b$ and the linear density of slits $N \cdot mm^{-1}$. The values of N can be within 500 and 1500 lines per millimeter for the visible spectrum.

The passway of rays through a diffraction grating is given in fig. 16.7, b. A parallel beam of rays is directed to the diffraction grating that falls perpendicularly on the surface of grating. The convergent lens L projects the diffracted rays from the grating in its focal plane where the screen of observation is situated.

A difference of ways determined by the equation $\Delta = BD = d \sin\varphi$ exists between two rays passing through two successive slits. The principal maxims (F_0, F_1,F_2,...,F_m) are formed in those directions for which the difference of ways is the whole multiple of wavelength, thus:

$$\frac{d}{m}\sin\varphi = m\lambda$$

Here m = 0,±1,±2,...

Knowing the constant of grating and the position of the respective maximum on the screen, the wavelength of researched radiation can be determined from this equation.

Principle of holography

The holography is a method of registration and restoration of wave fronts or, otherwise defined, is a method through which the image of an object can be obtained from the general figure of diffraction, made by that object.

Holography is a procedure consisting of two steps. In the first step the registration of wave fronts is performed on the photo, a background of coherent light, the so-called reference light, through which the diffraction figure is overlapped, produced by the respective object lens illuminated with the coherent light waves (fig. 16.8,a) the hologram will contain all the information regarding the amplitude and the phase of the wave diffracted by the object.

In the second step (fig. 16.8 b) the hologram constitutes the registered wave fronts and due to the variations of optical density presented on the photographic plate the effects of diffraction appear through which the image of the object is reconstituted.

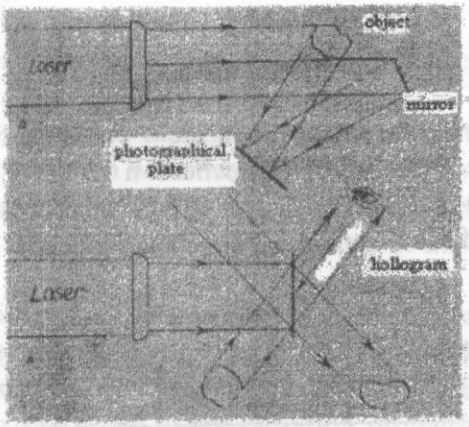


Fig. 16.8.

Equipment

The He - Ne laser; two diffraction gratings with different constants; optical bank; graded ruler; hologram; the screen of millimeter paper.

Measurement of wavelength of laser radiation by diffraction grating

The laser is switched on and the diffraction image is obtained on the screen by adjusting the optical system.

The distance L from the grating to the screen is measured and the distance s of n maximum from the central maximum (n=0). The relation s/L restores the numerical value of tgφ. Using the trigonometric table the respective value of sinφ is found out. Note that for small angles $\sin\varphi \approx \mathrm{tg}\varphi$

The obtained experimental results are introduced in table 16.1.

The equation of the diffraction grating determines the wavelength:

$$\lambda = \frac{d}{m}\sin\varphi$$

The energy of one quantum of He – Ne laser radiation is determined by the equation E=hν, where h is the Planck constant ($h=6.62\cdot10^{-34}$ j·s) The above mentioned measurements are repeated for the maxims of the second order (n=2) and the third order (n=3).

Table 16.1

Nr of experiments	m	L, cm	S, cm	$\sin\varphi$	λ, nm	ν, Hz	E, j
1	1						
2	2						
3	3						
					$\lambda_m =$	$\gamma_m =$	$E_m =$

Restoring of hologram

The diameter of laser radiation beam is increased by the divergent lens or lens system. The anterior performed hologram of the object is placed in front of the radiation flux, between the system of lenses and the eye of the observer. The optimal quality of the image is obtained in the plane of the hologram by varying the incidence angle of laser radiation.

Interaction with substance and applications of laser radiation in medicine and biology

The interaction of laser radiation with the substance can determine a series of effects depending on energetic intensity of laser radiation. These effects can be of two categories: **destructive and nonlinear.**

Destructive effects. They are manifested as a result of a very great energy concentration of laser radiation in the impact zone of the substance. On these places a "thermal shock" is produced because of the instantaneous increase of temperature of millions degrees.

The destructive and thermal effects are due to the electrical field from laser radiation which can reach the values of 10^{12} $V \cdot m^{-1}$ in case of localized beam. Under the influence of such a very intense electrical field the irreversible modifications at the molecular level are produced.

The great intensity of the electrical field is also associated with a great intensity radiation pressure (the exercised pressure by associated photons of the electromagnetic wave upon the irradiated body in the direction and sense of the radiation propagation). In case of some laser beams with small energetic intensity the orientations of non-spherical molecules are produced in the direction of radiation propagation (the energy falling on the unit surface in a time period).

The intensity of electrical field is very small in comparison with the internal microscopic electrical field in the light wave obtained from the usual source acting on the substance electrons.

Nonlinear effects of the light interaction with the substance are manifested in case of laser radiation, whose electrical field is comparable with the intensity with the interatomic electrical fields. So, it was observed that a laser beam passing a transparent medium is self-focalized and self – channeled. It was also observed, if the laser light passes through some crystal, a mixture of light from the incident beam and radiation with double frequency (for example, of ultraviolet radiation) is observed at the exit of the crystal. The other phenomena from the category of nonlinear effects (with the frequency of 30 GHz order) are the two laser beams representing the biphoton excitation and possibility of hypersound generation. The quantum interpretation of these phenomena offer the possibility of studying the interaction among photons by laser, the phenol-menon that can not be observed in nonlinear optics, where light radiation is supposed to be independent but optical properties of media (for example, the refractive index) do not depend on light intensity.

It has become possible to establish advantages of laser application in some methods of investigation in therapy and surgery due to accumulation of new data and progress in laser technology.

As the specialization research presents interaction with the living matter, the laser radiation can produce the following effects: thermal, photobiostimulant and photochemical. Two types of thermal effects can be produced as the function of the reached temperature by the contact with tissues: coagulation (for the temperatures between 60 and 100°C) and volatilization (for temperatures higher than 100°C).

As laser appeared, the following applications, covering new activity areas, (meteorology, processing and transmitting of information, holography, cybernetics, etc.) were found. A very special increase of laser application in the medical – biological field is also observed.

Due to distinct properties of laser radiation it is used as a working tool in studying living cells and different cell organs by the spectroscopic and microscopic techniques from the experimental cytology. The application of laser radiation in this research at the submicroscopic level is based on the so-called effect "needle – effect", due to possibilities of obtaining some laser beams with very small diameters (of microns order). The nucleus of the cell was eliminated of the cell by laser radiation, observing the behavior of the cell, the steps of agony and necroses, the chromosomes from the living cell were destroyed leaving other elements untouched. It can also act on some groups of genes at the ultramicroscopic level.

In medicine the application of laser allows the development of some medical techniques that will replace more efficiently the conventional techniques or create new modes of investigations and treatment. So, a laser that emits in IR can determine different blood substances, apply the usual method of blood sampling. The devices allow the precise and rapid measurement of glucose content, cholesterol, uric acid and alcohol (ethanol) from blood, this offering the possibility of detection of some diseases. (Tuberculoses, diabetes, etc.)

Lasers can be used in **surgery and microsurgery** in the treatment of glioma, in removing some tumors from the main blood vessels, in vaporization of some ventricles tumors, in extirpation of some intense cerebral tumors, in welding of some blood vessels, in detachment of bone muscles, in clinical endoscopy for checking gastrointestinal hemorrhage, for cicatrizing stomach ulcer, etc.

By this new surgery technique in which the laser beam plays the role of a classical lancet, some negative influences on the surrounding tissues are avoided. The rapid interventions are not bleeding (due to tissue vaporization), post-operative complications do not appear, but there are also other advantages.

In **ophtalmogy** laser is used in diabetic retinopathy, in occlusions of retina vessels, in prevention and extension of hemorrhages, in plucking or breaking retina, in surgery of eyelid tumors, in some forms of cataract and glaucoma.

In otorhinolaryngology laser allows the treatment of some pathologic conditions by photocoagulation or vaporization, such as in case of larynges papilomatose, vocal nodules, vocal cords poly-pile, in removing some benign tumor formations (papillomas), palate tonsillitis (in chronic tonsillitis).

Another field of laser applications is **dermatology** where both thermal effect and photobiostimulant and photochemical are used, some of indications are still at the research stage. The photocoagulation and vaporization of cuperoses, in removing benign and malignant skin tumors and in the treatment of dermatological affections are used.

A favorable photobiostimulant effect is used in epithelization of chronic ulcers, in acupuncture, in regeneration of hair by the action on pilose bulb, in producing of caloric vasodilatation and others.

The research on the application of laser at orthopedy and traumatology proved a stimulation effect of trophicity of tissues, cicatrized and anti-inflammatory. As a result, the laser radiation is indicated in the treatment of a number of pathologic conditions:

postrheumatical arthrosis and rheumatism, calcium periarthritis, osteoporosis, retardation of consolidation of bones, dyskeratosis and spondylosis and others.

The increase of specialists' interest and scientific research for the laser technique is also manifested by the study of physiology (for the local heating of some nerve portions or muscle tissue), of stomatology (for the treatment of dental carics), of bio – ecology, etc.

Therapy by laser radiation supposes a wide range of laser devices with functional optimal characteristic of a type of pathology. The laser of CO_2, the He – Ne laser, the Ar laser and others

are present among laser devices applied in medical field. In some cases the application of laser cannot replace the classical therapeutic techniques, but amplify the possibilities by finesse, precision, rapidity and efficacy.

Questions and tasks for self-control

1) Explain by diagrams what are the peculiarities of stimulated emission with respect to spontaneous emission.

2) Explain those three necessary conditions in functioning of quantum generator: the inversion of population, photon pumping and resonant cavity.

3) What are the effects of laser radiation interaction with the matter?

4) What are the fields of medical and biological application of laser radiation?

5) What are constructive elements and working technique of the examined installation to demonstrate the elimination of light energy loss at the reflection?

17. DETECTION OF NUCLEAR RADIATION

Purposes:

- study of effects produced by nuclear radiation at the interaction with atoms of the substance;
- analysis of peculiarities of ionized radiation interaction with a living matter;
- analysis of phenomena which are on the base of nuclear radiation detectors;
- detection of radioactive fond using the installation of "B- 4" type;
- research of r radiation attenuation in different substances;
- familiarization with the application of radioactive isotopes in medicine and biology.

Theoretical notions
Interaction of nuclear radiation with substance atoms

The nuclear radiation is formed by nuclear particles in movement. The nuclear particles are either elementary particles $\left({}_1^1p, {}_0^1n, e^-, \gamma\right)$ or nuclei $\left({}_1^2H, {}_2^4He\right)$ emitted by unstable nuclei.

The nuclear radiation interacts with the atoms of the substance that they penetrate and produce a series of effects. The effects of radiation can be used in applications. On the other hand, they are dangerous and must be limited. It is necessary to know interaction phenomena for both situations.

The nuclear radiation is divided in two categories after the charge of particles that they form:

A. The radiation formed by the electrically charged particles

These particles can have a great rest mass, can be nuclei or nuclear fragments (protons, particles α, fission fragments) or can have a small rest mass (electrons and positrons).

Ionizing. The most important interaction of charged particles with the substance in both cases is the ionization and excitation of medium atoms. Ions and free electrons are created by means of ionization phenomenon. In the process of atom ionization and excitation of the medium the particle gives a part of its kinetic energy that it transforms in the energy of excitation and ionization. As a result, the kinetic energy of an incident particle is decreased. The excitation energy can be transferred to the photon or to the particles of medium, thus increasing the energy of their thermal agitation.

At the formation of one pair of ions, the particle transfers energy ε to the other systems. If the particle penetrates a certain distance Δx into the medium, it will produce a number of ionizations in its way, its energy decreasing by ΔE. The value $\Delta E/\Delta x$ is called the loss of energy by ionization on the traversed unit. It is as great as the electrical charge of the particle is, and its speed is smaller. After that has kinetic energy decreased under the ionization energy, the particle captures the electrons and is stopped into the medium making a neuter atom (if it is a proton or α particle).

The route. The average way passed by charged particles into the substance is called the linear route. Its value depends on medium nature and the charge and kinetic energy of particle.

B. Radiation formed by neuter particles

Here we will have to study separately the behavior of the photons having the null rest mass or that of neutrons with the rest mass different from zero.

Passing of photons X or γ through the substance is followed by two phenomena: the photoelectric effect and the Compton effect.

The photoelectric effect (fig. 17.1) consists of absorption of the photon with the emission of electron with the kinetic energy E_{fe} given by the equation:

$$E_{fe} = E_{\gamma} - W_{el}$$

Where E_{γ} is the energy of the absorbed photon and W_{el} is the

bound energy of electron in the atom or crystalline structure. The absorption of photons by the photoelectric effect occurs especially at small energy and in the materials with the large atomic number Z.

The Compton effect (fig. 17.2) is a phenomenon of elastic scattering of photons on free electrons (very weakly bound) from the substance. After the interaction the energy of the photon is so small as it is scattered by a greater angle. The repulse electron takes the difference of energy. This electron can have kinetic energy included between zero and the maximal value corresponding to the scattering of the photon by 180^0.

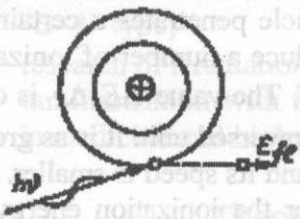


Fig. 17.1

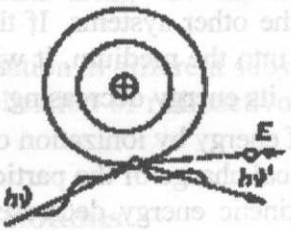


Fig. 17.2

By means of the photoelectric effect and the Compton effect the photons pull the rapid electrons into the medium and can produce in their turn other ionizing processes.

This phenomenon prevails at high energies of γ photons of about 5-10 Mev and at the interactions in the medium containing heavy nuclei. The energy of the photon is materialized as the form of electron – positron (fig. 17.3).

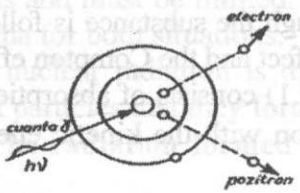


Fig. 17.3. **The creation of pairs**

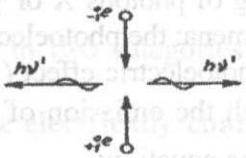


Fig. 17.4. **The scheme of annihilation reaction**

The positron disappears by annihilation (by combining with an electron) producing two γ quanta of energy 0.51 MeV (fig. 17.4).

Interaction of neutrons with substance

The neutron flux is also an ionized radiation, because, as a result of interaction with atomic nuclei, the charged particles and the radiation γ are formed.

The nuclear reactions produced by the capture of neutrons can be an example:

$$^{27}_{13}Al + ^{1}_{0}n = ^{24}_{12}Na + ^{4}_{2}\alpha \qquad ^{14}_{7}Al + ^{1}_{0}n = ^{14}_{6}C + ^{1}_{1}p$$

The nuclear reactors are the bases for producing neutrons.

The interaction of neutrons with the substance differs from the interaction of other particles at first due to the fact that the neutron interacts with the nuclei producing the nuclear reactions. The probability of these reactions is very great because the neutron has no charge and the electrostatic repelling does not stop its penetrating into the nucleus. The neutrons are absorbed in the nuclear processes. The knowledge of capture processes of the neutron into the nuclei is used for the protection of neutron particles. The materials which have the great probability of capture are $^{10}_{5}B$ and $^{113}_{48}Cd$:

$$^{10}_{5}B + ^{1}_{0}n = ^{4}_{2}He + ^{7}_{3}Li$$
$$^{113}_{48}Cd + ^{1}_{0}n = ^{114}_{48}Cd + \gamma$$

We should mention the fact that the neutrons interact at a small speed by capture processes.

Rapid neutrons interact elastically with the nuclei, assuring the maximal transfer of energy when the mass of nucleus is equal to the mass of neutron, that is, that of hydrogen. In biological tissues for the neutrons with the energy of 10 MeV more than 90 % of transfer energy is made by the elastic collisions with the hydrogen

nuclei. The rest of energy is used as non - elastic collisions (capture processes).

Those exposed confirm the application of materials enriched in hydrogen (paraffin, polyethylene) for screening neutrons sources.

Interaction of ionized radiation with living matter

The interaction of ionized radiation with a living matter is based on the fundamental processes of interaction occurred in nonliving matter. Those that differentiate the two actions are the consequences to which the ionized radiation leads in the living matter.

The physical – chemical mechanisms of ionized radiation action with the living matter are: a) **direct** by the impact of quantum or radiant particle with the biological macromolecules (excitation, ionization, photoelectric effect, Compton effect, the form of pairs); b) **indirect,** by the interaction with the atoms and liquid molecules from the organism and especially of water producing free radicals which then are combined with biomolecules.

These two mechanisms are known as the "theory of target" and "the theory of free radicals".

According to the first theory, there are certain sensible zones in the living cell, especially at the level of nucleic acids, bombardment of which with the particles or radiant quanta will lead to destroying of the cell.

The second theory states that the modifications produced by the action of ionized radiation are the consequence of some chemical dissociation especially in the cell water solution. The ionized molecule dissociates into ions and free radicals. Two radicals can be linked making, for example, peroxides, which, coming into reactions with macromolecule, produce essential changes for the cell.

Ions, radicals and peroxides produce a series of chemical reactions including oxidation chain reactions. These gradually impair the biochemical processes and normal functions developing ra-

diation disease. The chain oxidation reactions can easily take place in the substrates rich in double bounds (fig. 17.5) The devolving of oxidation reactions in lipids produces the defect of membrane structure and increase of its permeability.

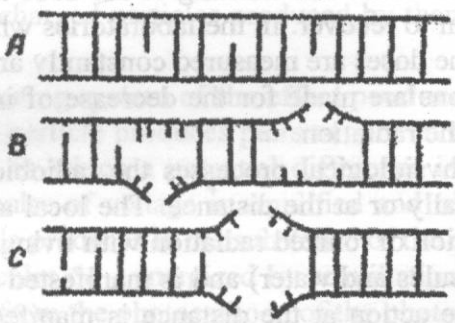


Fig. 17.5

These consequences of interactions of ionized radiation were confirmed by both the conductimeter method and electronic microscopy method.

Under the influence of ionized radiation the biological macromolecules modify the molecular weight, aminoacids are decarboxylized, disamminars are produced in peptide chains which are unbounded and reconstitute into another order, the colloidal substances can coagulate, but enzymes are essentially inactivated.

The effect of radiation is as great as the intensity of source is great and the irradiation time is longer and the distance with respect to the source is smaller. The effect of radiation depends on the type of radiation from which the fascicle is formed. According to this fact it is better to memorize that α particles and rapid neutrons are by 10-20 times more dangerous than the radiation X, γ or β.

The value indicating the global effect of radiation upon the organism is called biological dose. Its calculation takes into consideration all the factors indicated above. The dose is measured with

detectors of radiation called dosimeters. From the observations made during the years from the moment when the nuclear processes began to be applied widely, some limits for the biological doses were established biological effects of which do not affect the organism adversely during the entire life. This value is called the maximal permitted dose. The limiting of the dose at this value allows the organism to recover. In the laboratories where nuclear radiation is used, the doses are measured constantly and special measures of protections are made for the decrease of irradiation level in working with the radiation.

Regarding physiological processes the radiobiological effect is manifested locally or at the distance. The local action refers to the direct interaction of ionized radiation with living matter (biological macromolecules and water) and is manifested at the place of irradiation but the action at the distance is manifested in the organism at the neuro – humor level.

As a rule, the harmful effects do not appear immediately, but after a latent period.

The latent period is considered to be the time during which the biochemical complex reactions are produced and finally can lead to death of a cell or the organism. This latent time can last from several seconds to several years.

Detectors of nuclear radiation

The detector of nuclear radiation is a system that registers the nuclear radiation and allows the measurement of its quantity and other characteristics such as the energy or mass of particles.

Two parts form a detector:

a) **The detection body constituted by a medium where the nuclear radiation produces a specific effect.** The effect is given by the interaction of the nuclear particle with the substance of the detector; the charged particles produce ionization or scintillation, the γ photons produce the electrons by photoelectric or the Comp-

ton effect, the electrons which, in their turn, produce ionization, the neutrons produce nuclear reactions during which the charged particles appear and produce ionization. The response of the detector is linked with the electrical interactions of the particles. The particles which have not got the charges are not detected directly $(\gamma, {}^{1}_{0}n)$ but by charged particles produced by them in the material of the detector.

The registering system of the effect produced by a particle. In case when a particle produces pairs of ions in the detector, certain electrodes, in which a potential difference is applied, collect these and the pulse of voltage is amplified and registered. In case when the particle produces in the substance of the detector the excitations of molecules de – energized by scintillation, the photons release electrons from the photocathode of the photomultiplier which are multiplied; and the obtained voltage pulse on the photo-multiplier anode is amplified and registered.

The detectors can be classified after a series of criteria. The classification by the functioning principle parts divides detectors into:

1) **The detectors based on the gaseous ionization phenomenon.** While one charged particle passes through the detector gas, the electron – ion pair is produced and the pair is collected by two electrodes at which a potential difference is applied.

Such a detector is called the ionization room. The potential difference between the electrodes must be sufficiently greater so that the ions do not recombine in removing to them. The principle scheme of the ionizing room is given in fig. 17.6.

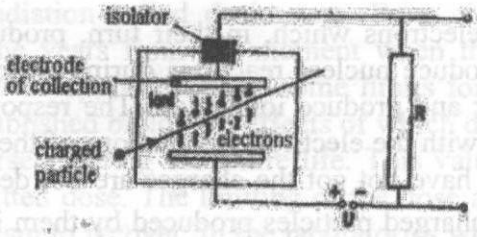


Fig. 17.6. **Principal scheme of the ionizing room.**

The positive ions and electrons are produced along the trajectory of the charged nuclear particle penetrating the gas of the room, directed to the electrodes between which the potential difference is applied. The current obtained on the collector electrode is amplified and measured. The ionizing current is proportional to the total number of the electron – ion pairs created by particles in the ionization room in the time unit. The current is small in the ionization rooms, the fact, leading to the complicated registering scheme. Therefore, another gas detector called the contour Geiger Muller is used more frequently. The scheme of this contour is given in fig. 17.7. The contour is made of a cylindrical capacitor. The detectors of radiation are the tools by which the nuclear radiation can be revealed and measured. These radiation produces directly or indi-recttly the ionization of substance with which it interacted. The above mentioned characteristic is on the base of the functioning principle of radiation detectors. One of the widely used detectors is the detector Geiger - Muller.

Such a contour presents a glass tube or metallic tube T (fig. 17.7) filled with some gases at the pressure of approximately 0.1 atm. (usually argon mixed with a small portion of alcohol or other organic vapors), with two electrodes.

The electrode 1 having the form of a very thin thread of tungsten placed axially across the tube, serves as anode. The cathode is a metallic layer, applied to the interior part of the tube (or the tube in case when it is made of metal).

It is seen from the installation scheme of the contour that the high continuum tension is applied between electrodes. On passing of one charged particle or a γ quantum the ions and electrons are produced and will be led by the electrical field of contour. In the vicinity of anode, where the intensity of electrical field is high, the shock of ionization by collisions takes place. As a result a small current Δi passes through the circuit of contour.

At the terminals of the resistance R (with the value of 1–2 $M\Omega$) a surplus of tension $\Delta U = R\Delta I$ transmitted through the capacity C of a amplifier A passes to the electronic circuit of the counting N. The capacity C plays also the part in discontinuation of the passage of high continuum tension to the contour.

The tension on the resistance R decreases to the same degree the tension between the electrodes of contour; on the other hand, the organic vapors take over more from the shock produced by collisions.

The 'extinction' of discharge through the contour occurs due to these phenomena, so that the reestablishment to the initial state ready to evidence the following particle with charge or photon.

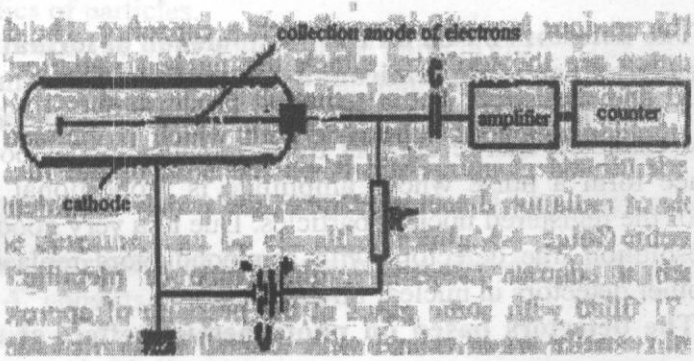


Fig. 17.7. **Principal scheme of Geiger-Muller contour.**

2) **The detectors based on the apparition of scintillation produced in the substance of the detector by the charged particle.**

The first detector of α particles was a zinc sulfide screen, on which the rapid α particles produce sparks and which were observed by means of a microscope. Now the scintillation detectors are widely used. The applied phenomenon is the apparition of scintillation in the crystals on passing of a charged particle. The photons are registered by a photomultiplier producing a pulse of voltage fig. 17.8 The amplitude of pulse is proportional to the number of scintillations produced by the charged particle while passing through the crystal and, thus, with its energy. Due to this fact the detector is used both in counting of nuclear radiation and measurement of their energy.

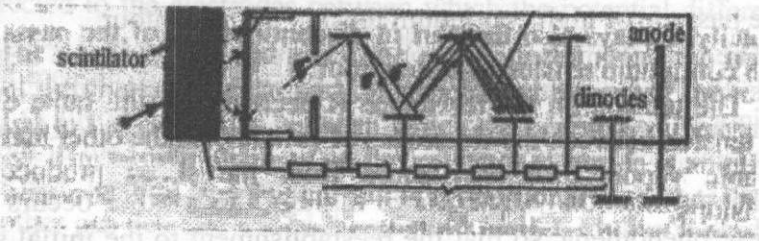


Fig. 17.8. **Principal scheme of scintillation detector.**

3) **The detectors based on the phenomenon of formation of hole – electron pairs into semiconductor crystals.** The number of electron – hole pairs is proportional to the energy of the particle.

The collected charge carriers by the application of potential difference form a pulse whose amplitude is proportional to the energy of the registered nuclear particles. Due to a very good energetic resolution, the detectors with semiconductors replace gradually other detectors in nuclear physics research.

4) **The detectors based on photochemical effect of radiation.**

Such a detector is a nuclear emulsion being a thicker photographic emulsion. On passing of the charged particle, the silver halide is decomposed and the developing centers of metallic silver are created. After developing, they are observed easily by a microscope.

The nuclear emulsion allows to visualize the trajectory of the particle and then the determination of energy and its types.

5) **The detectors based on the returning of medium at the normal state in the vicinity of ions formed along the trajectory of charged rapid particle, into a gas or liquid being in the metastable state.** Such a detector is the fog **room** built by Wilson in 1912. In the cavity filled with a gas a sudden falling of pressure creates the supersaturated vapors. The charged particles produce ions along their trajectory and these ions become condensation centers. The liquid drops are eliminated laterally and photographed, giving an image of the particle trajectory.

To register particles of high energy the **bubble rooms** are used. Their functioning is the same as that of the fog room. It is based on the fact that under some conditions a liquid can be warmed over the boiling temperature (the state of superheating). The boiling is not produced as the centers of boiling do not appear. These are the ions created on passing of the electrical charged particle through the liquid of the room. The gas bubbles formed along the trajectory are photographed and the photos give the information of the characteristics of particles.

To obtain the information of the particle charge, the fog rooms and bubble room are placed in the magnetic field perpendicularly to the direction of the movement of the particles.

Determination of radioactive fond by the installation B-4

The description of the installation of B-4 type. The installation "B-4" consists of two blocks:

1) the block of contours;

2) the main block.

The main block contains the electronic installation for supplying the contour with high voltage, the request electronic circuits of counting and registering pulses.

On the heading panel (fig 17.9) we have a series of command buttons with the respective inscriptions, the switch of polarity, and also six dekatrons serving for registering pulses, their maximal number can be 1000000.

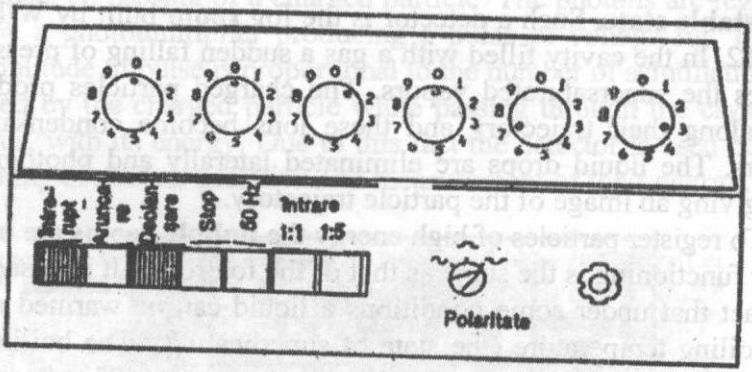


Fig.17.9

Each dekatron represents a tube of discharging in the gas that recalculates at ten, so for each 10 registered pulses a single pulse transmitted to the next dekatron appears.

The dekatron is made of a central anode as a disk and 30 cathodes as wire footing. The leading cathodes in the number of 10 are marked with the respective digits from 0 to 9 (in fig. 17. 10 white color).

The groups with different functions present the primary subcathodes (emphasized) and secondary subcathodes (of black color). A common foot in the socket of dekatron is destined to the cathodes of each group. Only the leading -zero cathodes with the independent foot present the exception. While coupling the installation in the circuit, some of the cathodes are lit aleatorily. By pressing the button "cancel" all the cathodes are turned off with the exception of "zero" cathodes.

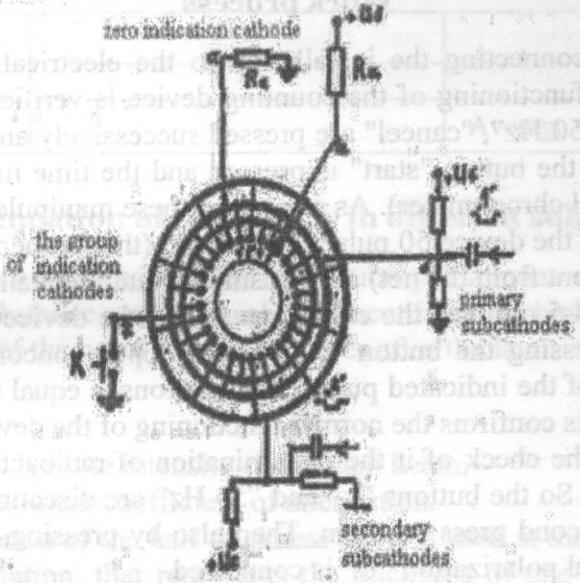


Fig. 17.10.

The resistance Ra from the circuit of anode excludes the concomitant turning on of two cathodes. When the leading device under the influence of the arrived pulse from contour transfers a negative pulse to the primary subcathodes, the voltage between the latter and the anode becomes higher than between the anode and "zero" cathode. Due to this the electrical discharge is transferred from "0" cathode to "1" cathode.

The next pulse from the contour analogously leads to the transfer of the electrical discharge from cathode "1" to the cathode "2" and so on. The tenth pulse from the contour makes the return of electrical discharge to the "0" cathode, concomitantly the out pulse of counting dekatron of units is transferred to the next counting dekatron of tens of units. The latter adds 100 impulse forms the out impulse addressed to the next counting dekatron of hundreds and so on.

Work process

After connecting the installation to the electrical circuit the quality of functioning of the counting device is verified. The buttons "≈", "50 Hz", "cancel" are pressed successively and then concomitantly the button "start" is pressed and the time marker is set on (or usual chronometer). As a result of these manipulations at the entrance of the device 50 pulses per second (the frequency of alternative current from the net) are transmitted automatically.

After 4-5 minutes the chronometer and the device of recounting by pressing the button "stop" are stopped concomitantly. If the speed of the indicated pulses by dekatrons is equal to 300 pulses/min, this confirms the normal functioning of the device.

After the check of it the determination of radioactive fond is performed. So the buttons "≈" and "50 Hz" are disconnected, making the second press on them. Then also by pressing, the button of the signal polarization "U" is connected.

Performing the above mentioned manipulations, the number of pulses H arrived from the Geiger Muller contour are registered during 5 minutes. The measurements are repeated several times, determining every time the number of pulses which refers to a minute $n = \frac{N}{t}$ (the characteristics of radioactive fond).

The obtained results are written in table 17.1

Table 17.1

Nr. of experiment	The duration registration T (min)	The number of registered impulses N	Radioactivelbnd n=N/t (pulses/min)
1			
2			
3			

Continu

4			
5			
			Average value n_{aver}=

The attenuation of radiation γ in different substances

As a result of those three interaction phenomena of radiation with the substance at passing of absorbent layer of thickness d, the intensity **I** of the beam **X** or **γ** decreases after the law:

$$I = I_0 e^{-\mu x}$$

Where: I_0 is the intensity of incident beam;

M – the linear coefficient of attenuation.

The notion of the half thickness ($d_{1/2}$) is used at the protection against radiation, that represents the thickness of one attenuating material reducing to the half the initial intensity of the radiation beam:

$$\frac{d}{2} = \frac{0.7}{\mu}$$

for the attenuation lead is used. The half thickness is: 0.92 cm for the radiation γ of ^{60}Co; 0.35 cm for the radiation γ of ^{131}J, etc.

In the table below the values of linear attenuation coefficients for the photons of energy 1.3 MeV in different substances are presented.

Table 17.2

Absorbent	$_6C$	$_{13}Al$	$_{26}Fe$	$_{29}Cu$	$_{82}Pb$
μ (cm^{-1})	0.115	0.156	0.455	0.509	0.8

The activity of radioactive source is a value of great practical importance. It is defined as the number of the produced disintegration per a time unit.

The activity in I. S. is measured as the disintegration per second (becquerel). The used symbol is Bq. The unit named curie (C_i) is more frequently used:

$$1C_i = 3.7 \cdot 10^{10} Bq$$

In this part of the practical paper on the use of the installation $B - 4$ we will determine the dependence of counting speed on the function of contour – source distance, and also the decrease of counting speed by the screening of the contour Geiger - Muller with the plates of different metals (Al, Fe, Cu, Pb) of the same thickness.

Although the used radiation source in the laboratory has the activity smaller than $1\mu C_i$ and does not present any danger, during the work the protection rules must be strictly respected.

Work process

With the rubber gloves and by special tongs the source is transferred from the box into the device for fixing (fig. 17.11) the Geiger – Muller contour situated above.

Varying the contour – source distance from 10 to 10 cm every time the total number of pulses of counting speed is registered. The experimentally obtained results are written in table 17.3.

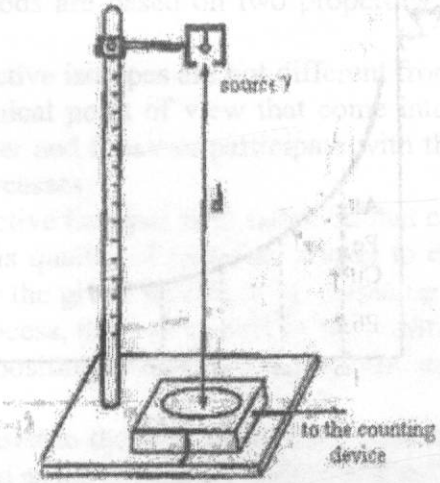


Fig. 17.11.

Table 17.3

Nr. of experiments	d (cm)	H (pulses)	$V = N/t$ (pulses/min)
0	∞	H_{fond}	V_{fond}
1	10		
2	20		
3	30		
4	40		
5	50		

Then the graphic of the function V=F(d) is drawn, analogously to the one represented in fig. 17.12.

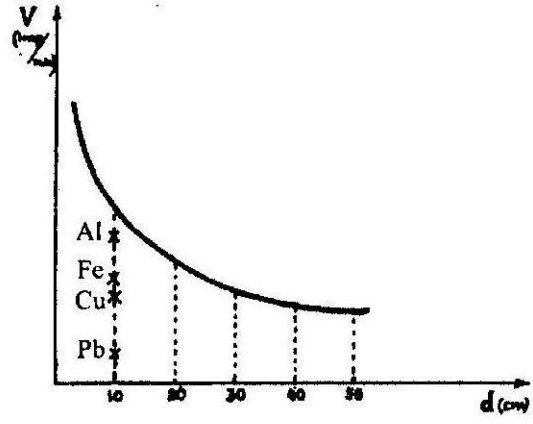


Fig.17.12

The application of radioactive isotopes in medicine and biology

Isotopes are atoms (exactly nuclides) with the same atomic number Z, but a different mass number A, that is a different number of neutrons. As the nucleus of these atoms has the same number of protons, that is the same electrical charge, they occupy the same place into periodical system of elements. The isotopes have the same electronic covering, that is the same the identical chemical properties, differing only in physical properties.

Due to their properties, the radioactive isotopes obtained in the reactors as a result of different nuclear reactions are used in various fields of science and engineering. They are used in industry, geology, archeology, chemistry, etc.

Radioactive isotopes are used in medicine and biology in three important directions: research, diagnostics and treatment. Their use with these purposes is explained by the possibilities and advantages offered by a series of studies and biochemical and biophysical research works of high sensitivity and complexity performed "in vivo".

Isotopic methods are based on two properties of radioactive isotopes, namely:

1) The radioactive isotopes are not different from stable isotopes from the chemical point of view that come into the constitution of living matter and thus can participate with them in all studied biological processes.

2) The radioactive isotopes emit radiation that can be detected and measured. This quality of radiation allows to establish metabolic processes for the given substance participating in the intensity of metabolic process, the way passed by the substance, the place and the time of depositing, the ways and the form under which it is eliminated, etc.

While being used in the mentioned three directions radioactive isotopes are utilized as tracer (marked atoms) and radiation sources.

Tracers. They represent radioactive isotopes of a chemical element which if being introduced in small quantity into a certain system, allows watching the route of that element into the system of the studied process. These radioactive isotopes participate in all processes and phenomena as the stable isotopes of the respective element, but the radiation emitted by them allows performing some research in the zones of observation inaccessible by other methods.

Radiation sources. They represent isotopes used to study some properties of tissues and organs or to produce a series of modifications.

It is observed that the radioactive isotopes can be used in two categories of studies: those of dynamic (variable in time) and the static characters.

Radioactive isotopes can be used as tracers in a variety of research aspects, such as:

a) study of metabolic processes "in vivo" at different structural and functional levels;

b) revealing of some metabolic processes and identification of some intermediary products;

c) study of metabolism of simple bioelements or compounds at different biological processes such as:

– the way passed from one organism to another during a lot of successive generations by the gametes (sexual cells) and zygote (fecundated ovule) that is the hereditary "biological way";

– the passed trajectory into the organism from introduction to elimination, that is the "physiological way".

– knowledge of successive intermediary compounds, mechanisms and processes as the chains of biochemical reactions in which the marked bioelements are implicated, that is the "biological way";

d) study of biosynthesis (the synthesis of one biological product by the living cell) of some organic compounds with the complex structure (as, for example, lipids, carbohydrates, proteins).

As the radiation sources, the isotopes are used in the study of properties of some organs and tissues; knowledge of bioelements repartition into the organs and tissues; the study of diagnostics in a number of diseases of the thyroid, liver, brain, in case of anemia and polycythemia (the increase of red cells in the blood); the revealing of some tumors (renal, pulmonary, gastric, etc.); study of pathogenic agents (for example, brucellosis, etc.). The radiation emitted by some isotopes is used also for the producing somatic and genetic effects, this way the abnormal cell proliferation is stopped (mitoses) as in case of cell formations with congenital defects. In radiotherapy the best results are obtained when isotopes are concentrated mainly in the organ submitted to the treatment (called"critical organ"). The property of nuclear radiation to stop the cell proliferation is used also to destroy microorganisms. This way the sterilization of some materials as, for example, suture fibers, plastic materials, etc. (in this case the physical properties are saved), surgery instruments and food products (assuring good pre-

serving and conservation) are made. The genetic effects examine also the production of genetic mutations finally followed by a selection action and crossing to improve or obtain new sorts of plants and animal breedings.

The application of isotopic methods in research presents the advantage of performing of some studies of high sensitivity, by noninvasive techniques, which give the possibility to observe metabolic processes "in vivo". These biophysical methods open large perspectives for a series of functional exploration, in diagnostics and treatment of some diseases.

Table 17.4

Some characteristics of radioactive nuclides

The nuclide	The half time	The energy of radiation in MeV		Critical organ
		β	γ	
^{3}H	12.4 years	0.018	-	The whole organism
^{14}C	5720 years	0.15	-	The adipose tissue
^{24}Na	14.97 hours	1.39	2.76	The whole organism
^{32}P	14.7 days	1.71	-	Bones
^{35}S	87.1 days	0.17	-	Skin
^{42}K	12.4 hours	3.6	-	Muscle
^{45}Ca	163 days	0.26	-	Bones
^{51}Cr	27.7 days	-	0.28	Blood
^{59}Fe	47.1 days	0.46	1.1	Blood
^{60}Co	5.26 years	0.30	1.33	Liver
^{92}Sr	28 years	0.5	2.26	Bones
^{131}I	8 days	0.69	0.36	Thyroid
^{137}Cs	30 years	0.51	0.66	Muscle

Questions and tasks for self-control

1) Name the types of nuclear radiation and their characteristics.

2) What are the mechanisms of interaction of nuclear radiation with the matter?

3) What phenomena are on the base of nuclear radiation detector construction?

4) Draw the simplified scheme of turning ombudsman of gas contour and explain its principle of functioning.

5) According to what law does the attenuation of /rays in the substance take place?

6) What are the fields of application of radioactive isotopes in medicine?

18. DETERMINATION OF RADIATION DOSE BY THE DEVICE "Chid-2"

Purposes

- presentation of theoretical notions of values and measure units in roentgenology and radiobiology;
- determination of radiation dose.

Theoretical notions

Biological effects of irradiation by ionized radiations depend on a lot of factors, such as the nature of radiations, the irradiation geometry, etc. It is difficult to propose a value easy to measure and universally valuable for biological effects of ionized radiations. Although the biological effects are to a great degree cumulative, the value called radiation dose was chosen. Two systems of dose measuring exist – roentgenological and radiobiological.

The roentgenological system can be applied only for the radiations r and roentgen *(x)* for which the photons energy is included between 0.03 and 3 MeV. (The electron volt *eV* is the energy of one electron obtained in passing through a potential difference of 1V into a vacuum. 1 eV=1.6 $\cdot 10^{-12}$ ergs). The radiobiological system includes all the radiations including γ and *x* radiations.

The roentgenological system

The fundamental value of the roentgenological system is the r or X radiation dose (Δ). The unit measure of this dose was called roentgen (the symbol R but sometimes r).

Roentgen is the radiation dose γ or x, with the emission of a number of ions into 0.001293 g of air, the total charge of them being 3.3356310 ^{10}coulombs (C).

(0.001293 of air is the mass of one cm^3 of dried atmospheric air at 0°C and 760 torrs).

The debit of dose (δ) the ratio between the radiation dose and the irradiation time t:

$$\delta = \frac{\Delta}{t}$$

The unit of debit is the roentgen per second R/s (hour R/h).

Resulting from the roentgen definition we can define this unit: a roentgen per second yields a current of about $3.33 \cdot 10^{-10}$ A into 1 cm^3 of dried air under the above mentioned conditions.

The integral dose (Δ_i)is the product between the radiation dose (Δ) and the mass m of the irradiated tissue.

$$\Delta_i = \Delta \cdot m$$

The unit measure of the integral dose is kilogram-roentgen (Kg ·m)

The radiobiological system

This system is referred to all types of penetrating radiations as well as to secondary effects caused by radiations.

The fundamental value from the radiobiological system is the **absorbed dose** (D). The equation gives it:

$$D = \frac{W}{m}$$

Where W is the energy absorbed from the radiation, but m - the substance mass or irradiated tissue.

The unit measure of IS is **gray** (Gr) that represents the dose of radiation at which the mass of 1 kg absorbed 1 j of energy. The derivative unit is called rad (the symbol rad) the short name of "roentgen absorbed doses". 1 rad = 0.01 Gr = 100 $\frac{ergs}{g}$.

The equation gives the debit of the absorbed dose (d):

$$d = \frac{D}{t}$$

where **t** is the irradiation time. The unit is rad/s (rad/h) that represents the debit of dose of 1 rad absorbed by a tissue per second (hour).

The integral absorbed dose (D_i) $D_i = D \cdot m$ where **m** is the mass of tissue yielded by the absorbed dose **D**. The unit measure is kilogram – rad (Kg · rad) or gram · rad (g-rad)

The biological dose (B)

The biological effect of ionized radiation depends not only on the absorbed dose but also on radiation type. This fact resulted in the introduction of the notion of biological dose. It is given in the equation:

$$B = \eta \cdot D$$

where the coefficient η is called relative biological efficacy. It represents the ratio between the absorbed dose of reference radiation and the dose of examined radiation yielding the same biological effect into the tissue under the same conditions of the initial state.

The reference radiation for defining η is the radiation X given by the installations of 200 kV and easily filtered:

$\eta = 1$ for the radiations β and γ;

$\eta = 2-5$ for the neutrons;

$\eta = 10-20$ for the radiations α;

As the incident radiation consists of different radiations, the total biological dose will be the sum of partial biological doses for each type of radiations. As a result, the advantage of biological dose (B) with respect to the absorbed dose (D) is that it allows evaluation of the total biological effect resulting from simultaneous irradiations or successive irradiations with different nuclear radiations.

The unit of measure of biological dose is rem (symbol rem) - the short name of "roentgen equivalent man".

Rem is the biological dose received by a gram of irradiated tissue when it absorbs the energy of 100/η ergs by ionized particles;η is the relative biological efficacy of incident radiations.

$$1rem = \frac{100}{\eta} ergs / g$$

Debit of biological dose (b) is **the** ratio between biological dose (B) and irradiation time **(t):**

$$b = \frac{B}{t}$$

Rem per second (hour, etc.) is the debit of biological dose of J rem per second (hour, etc.)

Generally, the effect increases with the increase of biological dose debit (for example, if B received by a person is 600 rem, the administrated dose in 40 years is b=0.041 rem/day that is not a dangerous dose. As this dose is received for a day, then b=600 rem/day, it leads probably to a rapid death; generally, the lethal dose is 400 rem received at a time.

The admitted maximum biological debit for the professional irradiation in working zones with the radioactive substances is considered to be 5 rem/year or 100 mrem/week.

The integral biological dose (β):

$$\beta = B \cdot m$$

wheres the mass of tissue exposed to the biological dose B.

The unit measure is kilogram (gram) – the rem

This value is important because it does not mattr whether the organism or only a part of it (for example, hand, arm, etc) received the biological dose.

Determination of radiation dose by the device "Chid-2". Description of device

The device "Chid – 2" is intended for determination of dose radiation Δ of χ and γ radiations within limits 0.05 - 1 R having energies of photons of about 150 keV up to 2 MeV by individual detectors. It consists of the load – check device and a set of individual detectors (fig. 18.1).

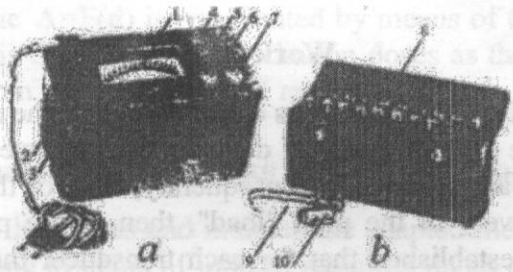


Fig. 18.1

The device of load – check (fig. 18.1,a) is supplied with continuum current supply with the voltage of 26 V, or from an alternative current net (220 V; 50 Hz).

The window (1) for reading values is on the front panel of the load – check device.

The measuring instrument has two scales with the respective gradations of two fields: 0.005 - 0.05 and 0.05 - 1R. The gradation 0.017 R from the first scale marked by the red color corresponds to the admitted maximal dose during 24 hours for professional irradiation in working zones with the radioactive substances or generators of X rays. On the same panel the following elements used during work are also mounted:

- the switcher (2);
- the knob for fixing the initial position of the indicator needle (3);

– the plug "measurement" (4);
– the plug "load" (5);
– the knob for adjusting sensitivity for the range 0.05 R (6);
– the knob for adjusting sensitivity for the 1R domain.

The garnish for 20 individual detectors (8) is given in fig. 24,1b. Each detector is made of two parts: the transducer of radiations with the limit of 0.05R (9) and the transducer of radiations with the limit of 1R.

Work process

The load – check device is connected up to the alternative current net. One of the individual detectors is discrewed and the parts (9) and (10) are separated. Consequently, each of them is introduced successively in the plug "load", then in the plug "measurement". It is established that for each transducer the indicator needle of the respective scale is removed to the right of the zero division. It also confirms the fact that the load – check device works normally.

Those two components of the detector are screwed in their places, setting it in the respective place of the garnish. The other 19 individual detectors are worked with samely.

As a source of ionized radiation the X ray device is optimal to use in the laboratory conditions, as it radiates in a single direction and does not present any danger for surroundings.

The direction of irradiation of the device is oriented vertically down. Two individual detectors are placed from ten to ten cm on the working table at different distances from this direction.

Then the device of X-rays is turned on for 30 min.

After this duration the device is disconnected and the fixed radiation doses are measured by each individual detector.

The obtained results are given in table 18.1

Table 18.1

Distance d(cm)	Radiation dose Δ (R)
0	
10	
.....	
90	

The graphic Δ=F(d) is represented by means of the data from the table, that is the variation of radiation doses as the function of the distance from the direction of X rays flux.

Questions and tasks for self- control

1) Define the values and indicate the adapted measuring units in two measuring systems of ionized radiation doses and radiobiology.

2) What are the constructive elements of the individual detector of ionized radiations from the garnish "Chid – 2"?

ANNEX
INTERNATIONAL SYSTEM OF UNITS(IS.)

In order to measure physical values, a proper measure unit can be chosen in the arbitrary way. The totality of measuring units defined arbitrarily and independently from others is called the incoherent system of measure units.

The use of incoherent systems of measure units presents two great disadvantages: 1) a great number of measure units must be defined and the standards must be performed for each separate case; 2) the mathematical formulae by which the physical laws are expressed will contain some coefficients depending on used measure units.

The development of sciences and techniques imposes that the measuring of certain values has the same signification for all. This result can be obtained by choosing a standard for each measured value, defining an admitted universal unit with precision.

The universality of these could not be performed only by an international convention. This was the purpose of the meter convention, the treaty signed in Paris in 1875. There 7 measure units were defined arbitrarily and called fundamental units. Other measure units whose value is not arbitrary but defined by fundamental units are called derivative units. The totality of fundamental and derivative units is called the coherent system of measure units.

In October 1960, at the XIth general conference of measures and weights held in Paris the measure units system was proposed with the name International System of Units (I.S), the adopted system as single and compulsory for the majority of the world countries. In May 1977 the World Health Organization decided to introduce I. S in all medical professions expressing, thus, the importance of I.S. for medicine.

The I. S units for measuring physical values are of three types: a) fundamental; b) derivative; c) supplementary.

a) The fundamental units are presented in Table A. 1 together

with the material element and its property used for each definition from the 7 fundamental units.

The symbols of units are written with small Latin letters; if the symbols are built from proper names, then they are marked with capital letters. The point is not used after a symbol.

b) **The class of I. S. derivative units.** It includes the units that can be formed by combining the fundamental units on the basis of certain chosen algebraic equations. A lot of these algebraic expressions can be replaced by the names and special symbols, which can be used for expressing some derivative units more simply than on the basis of fundamental methods.

Some derivative units result from multiplying of fundamental units by themselves, or by combining two or more basic units by simple multiplication or division.

The derivative units can be classified into three groups.

Table A. 1

	Value	**Material element**	**Used property**	**Name and definition of the unit**
1.	Mass	The conserved international prototype into the pavilion Breteuil (Sevres)	Mass	Kilogram 1 kg = the mass of standard prototype
2.	Time	The atom of Cs^{133} in the fundamental state	The correspond-ding radiation between two superfine levels	Second 1 s = the duration of 9192631770 periods of radiation
3.	Length	The Krypton atom 86	The wavelength into a vacuum of corresponding radiation for the transition $2\,pl_0 - 5d_5$	Meter 1 m = 1650763,73 lengths of waves of radiation

Continu

4.	Thermody-namic temperature	Pure water	The temperature of treble point (the equilibrium solid - liquid -steam)	Kelvin The temperature of treble point 273. 16 K
5.	Luminous intensity	Black body at the freezing temperature of Pt	The radiation of 1/60 cm^2 in the direction perpendicular to the surface	1 cd =the luminous intensity of radiation
6.	The quantity of matter or substance	Carbon atoms 12	The quantity of matter contained in 12 grams ofC	Mole 1 mole = quantity of atoms of C^{12}
7.	The intensity of electrical current	Two parallel rectilinear infinite conductors situated at the distance of 1 m into a vacuum passed by the same electrical current	The force on the meter length appearing between two conductors	Ampere 1 A - the current making the force to appear

Table A. 2

Value	The I.S	
	Name	Symbol
Area	Square meter	m^2
Volume	Cubic meter	m^3
Speed	Meter per second	m/s
Acceleration	Meter per square second	m/s^2
Volume mass	Kilogram per cubic meter	kg/m^3
Magnetic field	Ampere per meter	A/m
Concentration (of substance quantity)	Mole per cubic meter	mol/m^3

Table A. 3

Value	The I. S			
	Name	Symbol	Expression in other units	Expression in fundamental I. S
Frequency	Hertz	**Hz**		s^{-1}
Force	Newton	**H**		$m \cdot kg \cdot s^{-2}$
Pressure, mechanical tension	Pa,	**Pa**	**N/m²**	$m^{-1} \cdot kg \cdot s^{-2}$
Energy, mechanical work, heat quantity	Joule	**J**	**Nm**	$m^2 \cdot kg \cdot s^{-2}$
Power	Watt	**W**		$m^2 \cdot kg \cdot s^{-3}$
Quantity of electricity, electrical charge	Coulomb	**C**	***J/s***	A·s
Electrical potential, electrical voltage, Electromotive voltage	Volt	**V**	W/A	$m^2 \cdot kg \cdot s^{-3} \cdot A^{-1}$
Electrical capacity Electrical resistance	Farad Ohm	**F Ω**	C/V	$m^2 \cdot kg \cdot s^4 \cdot A^2$ $m^2 \cdot kg \cdot s^{-3} \cdot A^2$
Temperature Celsius	Grade	**°C**	V/A	K
Light flux Illumination	Lumen Lux	lm **lx**		Cd · sr
Radionuclide activity		**Bd**	Lm/m^2 s^{-1}	$m^{-2} \cdot cd \cdot sr$
Absorbed dose, The index of absorbed dose	Gray	**Gy**	s ***J/kg***	$m^2 \cdot s^{-2}$

1) The derivative units expressed as the function of fundamental units

The examples of such derivative units are:

Table A. 4

Value	I. S. unit		
	Name	Symbol	Expression in 1 the fundamental I. S units
Dynamical viscosity coefficient	Pascal – second	*Pa·s*	$m^{-1} \cdot kg \cdot s^{-1}$
Surface tension coefficient	Newton per meter	*N/m*	$kg \cdot s^{-2}$
Thermal capacity, entropy	Joule per Kelvin	J/K	$m^2 \cdot kg \cdot s^{-2} \cdot K^{-1}$
Mass thermal capacity, mass entropy	Joule per kilogram Kelvin	J/(kg·K)	$m^2/s^{-2} \cdot K^{-1}$
Mass energy	Joule per kilogram	J/kg	$m^2 \cdot s^2$
Thermal conductivity	Watt per meter Kelvin	W/(m·K)	$m \cdot kg \cdot s^{-3} \cdot K^{-1}$
Molar energy	Joule per mole	J/mole	$m^2 \cdot kg \cdot s^{-2} \cdot mol^{-1}$
Exposition (of X - ray and γ)	Coulomb/kilogram	C/kg	$kg^{-1} \cdot s \cdot A$
Debit of the absorbed dose	Gray per second	Gy/s	$m^2 \cdot s^{-2}$

II. A series of I. S. derivative units received special names, the name of a scientist is used frequently with the essential contribution in the field.

Examples:

III. – The third group of derivative I. S. units contains the units expressed by using special names.

Examples:

c) **the class of supplementary I. S. units** contains some units for which the general Conference have not decided yet whether they belong to the fundamental units or to derivatives units. These units can be treated as fundamental or derivative.

Now this class does not contain more than two pure geometrical units.

Table A.5

Value	I. S unit	
	Name	Symbol
Plane angle Spatial angle	Radian steradian	Rad Sr

A series of names and symbols for the prefixes were adopted due to the formation of multiples and decimal submultiples of I. S units between the values 10^{18} and 10^{-18}. The range $10^{3} - 10^{-12}$ is used in biology and medicine.

Table A. 6

10^{3}	Kilo	k
10^{2}	Hecto	h
10	Deca	da
10^{-1}	Deci	d
10^{-2}	Centi	c
10^{-3}	Mili	m
10^{-6}	Micro	μ
10^{-9}	Nano	n
10^{-12}	Pico	P

The World Health Organization makes the following statements for the field of biology and medicine:

1. As the interaction of chemical substances takes place in certain proportions as a function of relative molar mass, the quantity of substance is measured using the mole as a unit.

2. The measurement of substance concentration in moles instead of mass units is adopted in clinical biochemistry (for example, milligrams per liter).

3. The adoption of mole unit implies an essential change in dimensions, from the "mass" to the "active particles".

4. The concentration of a substance is measured in mol/m^3 or moles/I: the molar in moles/kg; the molar fraction or molar ratio in mole/mole.

5. The use of mole unit is exemplified in the instructions of World Health Protection (WHP) for measuring immune globine concentration, hemoglobin hydrogen ions, enzymes, the flux of substances eliminated by the urine; the use of unit Pa is also exemplified (instead of mm Hg) for measuring partial pressure of gases, blood and interocular pressure, etc; unit joule instead of calorie, unit newton instead of kilogram force, unit kilopascal – second per liter for vascular resistance.

6. The unit of joule per kilogram replaces rad and rem, coulomb per kilogram replaces roentgen; gray (gy) is used for measuring the absorbed dose.

Table A.7

Density of some solid bodies (at 20 ^{0} C)

Substance	ρ, kg/m^3	substance	ρ, kg/m^3
Brass	8300-8700	Bone	1800 - 2000
Aluminum	2690	Iron	7700 - 8000
Silver	10500	Paraffin	870 – 930
Aurum	19300	Lead	11220 – 11440
Bronze	8700	Cork	240
Constantine	8800	Platinum	21200 – 21700
Quartz	2650	Kitchen salt	2080 – 2200
Copper	8600-8900	Island spat	2710
Diamond	3400-3600	Glass	2400 – 2600
Ebonite	1800	Rhombic	2700
Iron	7860	sulfur	1960
Ice	880-920	Monoclinic	19100

Continu

Cast	2170-2310	sulfur	1590
Graphite	1900-2300	Wolfram	6860 – 7240
Marble	2520-2840	Sugar	
I Nickel	8400-9200	Zinc	

Table A. 8

Density of water at different temperatures

t,°c	ρ , kg/m^3	t,°C	ρ , kg/m^3	t,°C	ρ , kg/m^3
0	0,99987	13	0,99940	26	0,99681
1	0,99993	14	0,99927	27	0,99654
2	0,99997	15	0,99913	28	0,99626
3	0,99999	16	0,99897	29	0,99597
4	1,00000	17	0,99880	30	0,99567
5	0,99999	18	0,99862	31	0,99537
6	0,99997	19	0,99843	32	0,99505
7	0,99993	20	0,99823	33	0,99472
8	0,99988	21	0,99802	34	0,99440
9	0,99981	22	0,99780	35	0,99406
10	0,99973	23	0,99757		
11	0,99963	24	0,99732		
12	0,99952	25	0,99707		

Table A. 9

Density of ethyl alcohol at different temperatures

t,°C	ρ , kg/m^3	ρ ,°C	ρ , kg/m^3
0	806,25	35	776,71
5	802,07	40	772,20
10	797,88	45	767,20
15	793,67	50	762,94
20	789,45	55	785,62
25	785,22	60	754,10
30	780,97		

Table A. 10

Density of water solutions of ethyl alcohol (at 20°C)

C, %	ρ, kg m^3	C,%	ρ, kg m^3
5	989,38	55	902,61
10	981,85	60	891,15
15	975,22	65	879,50
20	968,70	70	867,70
25	961,69	75	855,70
30	953,85	80	843,49
35	944,98	85	830,96
40	935,24	90	817,95
45	924,84	95	804,15
50	913,86	100	789,33

Table A. 11

Psychometric table of relative humidity of air

Dried therm	Difference between the dried thermometer and humid one										
	0	1	2	3	4	5	6	7	8	9	10
0	100	81	63	45	26	11					
2	100	84	68	51	35	20					
4	100	85	70	56	42	28	14				
6	100	86	73	60	47	35	23	10			
8	100	87	75	63	51	40	28	18	7		
10	100	88	76	65	54	44	34	24	14	4	
12	100	89	78	68	57	48	38	29	20	11	
14	100	90	79	70	60	51	42	33	25	17	9
16	100	90	81	71	62	34	45	37	30	22	15
18	100	91	82	73	64	56	48	41	34	26	20
20	100	91	83	74	66	59	51	44	37	30	24
22	100	92	83	76	68	61	54	47	40	34	28
24	100	92	84	77	69	62	56	49	43	37	31
26	100	92	85	78	71	64	58	50	45	40	34
28	100	93	85	78	72	65	59	53	48	42	37
30	100	93	86	79	73	67	61	55	50	44	39

Table A. 12

Water surface tension coefficients at temperatures absolute from 0° *to* 80° C

Tempe-rature in the scale Celsius	Surface tension coefficient in dyn/cm	Tempe-rature in the scale Celsius	Surface tension coefficient in dyn/cm	Tempe-rature in the scale Celsius	Surface tension coefficient in dyn/cm
0	1,7921	21	0,9810	70	0,4061
5	1,5188	22	0,9579	80	0,3565
10	1,3077	23	0,9358	90	0,31365
15	1,1404	24	0,9142	100	0,2838
16	1,1111	25	0,8937		
17	1,0828	30	0,8007		
18	1,0559	40	0,6560		
19	1,0299	50	0,5494		
20	1,0000	60	0,4688		

Table A. 13

Water surface tension coefficients at temperatures from 0° to 80° C

Temperature in the scale Celsius	Surface tension coefficient in dyn/cm	Temperature in the scale Celsius	Surface tension coefficient in dyn/cm
0	75,625	45	68,592
5	74,860	50	67,699
10	74,113	55	66,894
15	73,350	60	66,040
20	72,585	65	65,167
25	71,810	70	64,274
30	71,035	75	63,393
35	70,230	80	62,500
40	69,416		

Table A. 14

Water viscosity coefficient between 15 °C and 100 °C expressed in centipoise (cP)

Liquid	Viscosity coefficient
Water	1.00
Acetone	0.33
Alcohol	1.2
Petrol	0.53
Glycerine	850
Arterial blood	4.5
Blood plasma	1.6 – 2.4
Urine	1.02 – 1.14

Table A. 15

Specific resistance of some metals and alloys in ($\Omega \cdot m$)

Pure metals and alloys	ρ x 10^4 $\Omega \cdot m$	Pure metals and alloys	ρ x 10^4 $\Omega \cdot m$	Pure metals and alloys	ρ x 10^4 $\Omega \cdot m$
Brass	0.0008	Copper	0.017	Platinum	0.107
Aluminum	0.029	Iron	0.086	Lead	0.21
Silver	0.016	Iridium	0.053	Potassium	0.070
Aurum	0.023	Manganin	0.43	Radium	0.060
Bismuth	1.20	Mercury	0.958	Sodium	0.049
Cadmium	0.076	Nickel	0.070	Antimony	0.45
Cobalt	0.090	Niccolite	0.42	Tantalum	0.12
Constantan	0.50	Nichrome	1.1	Wolfram	0.056
Tin	0.113	Palladium	0.107	Zinc	0.060

Table A.16

Wavelength of certain spectral lines, in nm (10 m)

Element	λ, nm	Element	λ, nm	Element	λ,nm
Ba	455,4	H	397,0	Li	
	493,4		410,2		610.4
	553,5		434,0		670
	577,8		486,1	Na	589,0
	597,2		656,3		589.6
	614,2	He	388,9	Sr	460,7
	649,6		402,6		638,6
Ca	445,5		447,1		640, S
	487,8		471,3		
	527,0		492,2		
	534,9		501,6		
	559,0		587,6		
	585,7		657,8		
	612,2		706,3		
	616,2	K	404,5		
	643,9		691,1		
Cu	402,3		693,9		
	406,3		766,5		
	427,5		769,9		
	637,8				
	458,7				
	515,3				
	521,8				
	570,0				
	578,2				

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