

FEDERAL AGENCY FOR EDUCATION

Lobachevsky State University of Nizhny Novgorod

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PHYSIOLOGY OF BLOOD AND CARDIOVASCULAR SYSTEM

Educational and methodical manual

Recommended by the methodological commission of the Institute of biology and biomedicine for UNN students studying in the field of training: 31.05.01 "Medicine", 31.05.03 "Dentistry", 30.05.01 "Medical biochemistry", 30.05.02" Medical Biophysics", 30.05.03" Medical Cybernetics", 06.03.01" Biology", 05.03.06"Ecology".

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Reviewer

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The training manual contains materials for practical work on the course of human and animal physiology and test questions on the topics of the lesson. This training manual is intended for students studying in the following areas: 31.05.01 "Medicine", 31.05.03 "Dentistry", 30.05.01 "Medical biochemistry", 30.05.02" Medical Biophysics", 30.05.03" Medical Cybernetics", 06.03.01" Biology", 05.03.06"Ecology".

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INTRODUCTION

Almost all of our knowledge in the field of human physiology is based on the results of laboratory experiments, thanks to which the information presented in textbooks and lectures was obtained. The experimental approach is used to solve many of the remaining mysteries in the body's work, and only experiment makes it possible to understand physiology as a science. In addition, physiology is the theoretical basis of medicine, its foundation, and, consequently, the physiological experiment are considered as an important stage of scientific clinical research. It is obvious that the laboratory workshop should be an integral part of teaching students the basics of human and animal physiology.

General objectives of the workshop:

- * demonstrate how the physiological processes studied in the theoretical course actually occur in a living organism;
- * give a general idea of some of the physiological methods and devices used to observe physiological phenomena and measure their parameters;
- * study the mechanisms that control the physiological functions of the body.

ETHICS OF PHYSIOLOGICAL EXPERIMENT

Compliance with the rules of humane treatment of animals is a prerequisite for conducting physiological experiments. In 1984, International recommendations for conducting biomedical research using animals were approved. This document of the European research Council and the Advisory Committee on medical research sets out the most important principles for setting up a biomedical experiment.

In Russia, the procedure for using animals in an experiment is determined by a number of documents approved by the heads of all departments where this type of research can be conducted. The main document is the "Rules for working with experimental animals". This document stipulates that the researcher has the right to use animals in the experiment, but a number of provisions must be strictly observed:

- * You can only conduct an experiment using animals in public institutions that have an appropriate experimental base.
- * Setting up such experiments is allowed only in institutions where there is an equipped vivarium, served by the staff who serve animals.
 - Only people with higher education in biological, medical, veterinary and zootechnical field can conduct experiments with animals.
- * The model of the experiment must meet the requirements of humane treatment of animals and in the case of any painful manipulations, it is necessary to use anesthesia.
- * Compliance with the requirements of humane treatment of animals should be noted when presenting the methodology and results of the experiment in scientific publications or compiling reports.

* Healthy animals of the appropriate species should be selected for experiments, limited to the minimum number required to obtain scientifically reliable results.

* The next in order and degree of importance of the recommendations is the requirement of ethical treatment of animals. Researchers and other personnel should always treat animals as sensitive to various types of impacts and consider it their ethical duty to treat and use animals in such a way as to minimize the inconvenience and pain caused to them.

MEASURES FOR INJURIES, CONTACT WITH BLOOD AND OTHER BIOLOGICAL MATERIALS

If contact with blood or other fluids occurred with a violation of the integrity of the skin (prick, cut), the person must do the following:

- * remove gloves with the work surface inside;
- * squeeze the blood out of the wound;
- * wash your hands under running water with soap, and then treat the damaged area with one of the disinfectants (70% alcohol, 5% tincture of iodine);
- * apply a patch to the wound, put on a fingertip;
- if it is necessary to continue work - wear new gloves.

If the skin is contaminated with blood or other biological fluid without damage, you must:

- * treat the skin with one of the disinfectants (70% alcohol, 3% chloramine solution);
- * wash the contaminated area with soap and running water and re-treat with 70% alcohol.

If biological material gets on the mucous membranes:

- * oral cavity - rinse with 70% alcohol;
- * nasal cavities-drip a 30% solution of albucide;
- eyes - rinse with water (clean hands), drip a 30% solution of sulfacetamide. A 0.05% solution of potassium permanganate can be used to treat the nose and eyes.

If the biomaterial gets on the robe, clothing:

- * disinfect gloves;
- * remove clothing and soak it in a disinfectant solution (except for 6 % hydrogen peroxide, neutral calcium hypochloride, which destroy tissues) or place it in an autoclave bag;
- * wipe the skin of the hands and other parts of the body with 70% alcohol under contaminated clothing, then rinse with soap and water and re-wipe with alcohol;
- * clean contaminated shoes twice with a rag soaked in a solution of one of the disinfectants.

FIRST AID KIT FOR EMERGENCY MEDICAL CARE:

- * finger pads (or gloves) at the rate of 1-2 per student per shift;
- * adhesive tape - 1 coil;
- * potassium permanganate in attachments of 0.05 g;
- * potassium permanganate dilution tank;
- * ethyl alcohol 70%;
- * a tube-dropper of a 30% solution of sulfacetamide ;
- * 5% iodine tincture and 3% hydrogen peroxide solution;
- * rubber gloves - 3 pairs, glasses., plastic aprons, 4-layer masks;
- * large plastic bag for collecting contaminated clothing;
- * attachments of disinfectants: chloramine 30 g; 3 attachments (each stored separately);
- * container for diluting disinfectants.

After reading the rules and receiving instructions on safety, the student signs in the "Journal of control sheets for instructing students on safety".

PREPARATION OF REPORTS ON PRACTICAL WORK

The accumulation of knowledge in any field occurs through active communication of scientists, which consists in publishing the results of experiments in scientific journals and presentations at conferences, congresses and symposiums. Therefore, the task of the workshop is not only to get acquainted with the basics of experimental work, but also to teach students the rules for presenting the results of scientific work in the form of written reports (protocols) and oral messages. The written report is proposed to be based on the same rules that are usually imposed on the publication of experimental materials by scientific journals. The report on practical classes includes the main sections that are present in the scientific article - "Introduction", "Methodology", "Results", "Discussion", "Conclusions" and "Literature".

Introduction. It contains a small amount of basic information about the problem under study, and sets out the goals of the experiment. The definition of the main physiological phenomena and concepts under study is given, and the expected results of experiments can be described. You must remember to correctly quote all the information sources used in this part of the report. Include in this part of the report only the information that is relevant to this work!

Methods (methodology). This includes a brief description of the object of research, materials, devices, equipment, substances and reagents, as well as methodological approaches used in the experiment. The description of the methods should be detailed enough for other researchers to repeat the experiment. At the same time, you should avoid excessive detail, and it is better to refer to the original literary

source, where the methodological techniques are considered in detail. If you have made any modifications to the experiment, this must be reflected in the description. Do not forget to specify the doses and concentrations of the drugs used.

Result of work. This section can be designed separately or together with the next section "discussion of results".

The results obtained in the experiment can be presented in the form of original recordings on the tape recorder, cardiograph or electroencephalograph. You must specify the speed of the tape, the parameters of the applied stimuli with accurate recording of the moment of application and termination of the stimulus (in the captions to the illustrations, appropriate explanations are given). If the registration was performed from the oscilloscope screen, on the scale of the pressure gauge, etc., then it is more convenient to present the results of the experiment in the form of a table. The table contains the obtained values of the studied parameters and their units of measurement.

If it is possible to identify the main regularities of the studied phenomena, graphs are built based on the obtained data. They should be neat and clear. You do not need to build each graph on an A4 sheet, but you should not reduce it to the size of a postage stamp. Graphs must have a title, parameter designations along the axes with units of measurement, number and explanations of the symbols used in it (legend); all experimental points and calculated parameters are entered in them.

Discussion of results. This is the most important section of the report that reveals the depth of understanding of the problem being studied and the ability to apply theoretical knowledge in explaining the results obtained in a real experiment. Discuss your results from the perspective of modern science concepts. Try to imagine the mechanisms underlying the observed phenomena. Explain the significance of the discovered method of regulation in the work of the whole organism. If the results obtained differ from the theoretically expected results, try to identify possible reasons for these discrepancies. When making assumptions, don't forget about the limitations that any measurement technique has.

Conclusions. They briefly list the main results and patterns found in the experiment. For example: "when the amplitude of the stimulus increases from ... mV to ... there is an increase in the amplitude of the muscle response. Further amplification of the stimulus does not change the muscle response." (It is not necessary to explain the mechanisms of the observed phenomena again – they are already set out in the "Discussion" section.)

Literature. At the end of the work, all the literature sources that you used in the report design and referenced in the theoretical introduction should be listed in alphabetical order.

Thus, it is clear that the laboratory report (Protocol) should be brief and objective. The key point is the completeness and consistency of the above mentioned. A thoughtful approach to explaining discrepancies between the results of the experiment and the theory is much more correct than trying to ignore them! This will teach you, as future researchers, to be accurate and critical in evaluating the results obtained.

Tools for dissection

The following set of tools can be used to perform the work described in the workshop (Fig. 1):

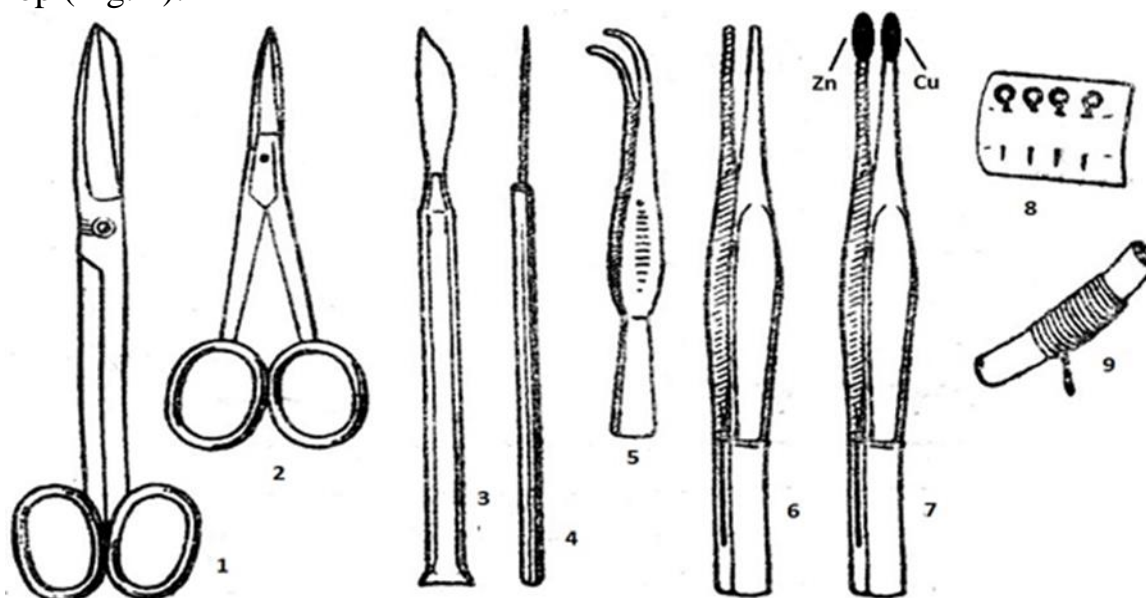


Fig. 1 Tools for dissection (explanations in the text)

Scissors are large with straight ends, one of which is sharp (1).

Small (eye) scissors for fine preparation, necessary for most work on the physiology of the nervous system and the physiology of blood circulation (2).

Tweezers: anatomical (6), ocular (5).

Preparation needle (4). Pins (mainly for attaching a frog to a plate). They should be at least four, and it is better not to scatter them in a box, but to give them fixed in a paper substrate (8).

The scalpel (3) is issued only for certain works, for example, for opening the eye, for operations to the experiment of Sechenov braking. Knife for Stripping contacts, electrodes, wires, etc.

Ligature or thread (9) (for convenience, wound on a rubber tube).

Galvanic tweezers (7) the Tweezers are made of surgical tweezers with cu and Zn plates on the branches.

Various clips and cannulas (issued when performing the relevant work and are not included in the permanent set of tools).

It is recommended that each student be given a set of tools that is necessary for this laboratory task.

Methods of immobilizing a frog

In conducting a physiology workshop, it is necessary to immobilize the frog either by destroying the brain and spinal cord, or by anesthesia. Destruction of the frog's brain and spinal cord can be performed in the following ways:

Destruction of the brain and spinal cord (Fig.2). Take the frog in your left hand with your hand back up, so that your thumb is on its back. Place your index finger on the frog's upper jaw and tilt its head down. In this position, the location of the occipital

fossa is clearly visible. Through the fossa between the occipital bone, insert the needle into the skull cavity and destroy the brain. Then turn the needle in the opposite direction with the spine and insert it into the spinal canal, destroying the spinal cord with several turns of the needle. The General relaxation of the frog's muscles and the lack of reflex reactions indicate complete destruction of the brain and spinal cord. With this method of immobilizing the frog, very little blood is lost.

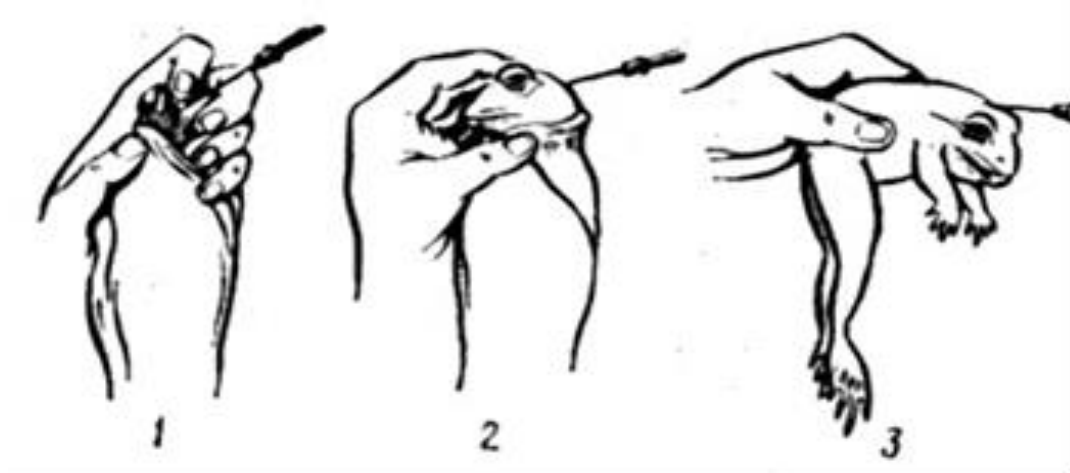


Fig. 2 Scheme of destruction of the brain and spinal cord

Decapitation followed by destruction of the spinal cord. Take the frog in your left hand, and with your right one, insert the lower blade of the scissors as deep as possible into the mouth under the back of the upper jaw behind the eyes. With a quick movement, cut off the upper jaw at the level of the rear end of the eardrums (save the lower jaw). Insert a dissecting needle into the opening of the spinal canal and destroy the spinal cord.

The use of anesthesia (ether, alcohol, urethane). Anesthesia is rarely used in the training workshop. To anesthetize the frog, a 10% solution of alcohol or a 2% solution of ether is used. The frog is released into the solution for 10-15 minutes. Muscle relaxation and lack of motor activity are good indicators of sufficient anesthesia action.

Fixing a frog

In a number of works, the spinal preparation of a frog is used - a frog whose brain is destroyed and the spinal cord is preserved. When preparing the corresponding nerves and muscles and conducting research, the spinal frog must be fixed to the plate motionless. It is best to fix it on a cork (or paraffin) plate with a size of 20x10 cm.

When fixing a frog on a plate, it is important to stretch its limbs well so that they are stationary and do not interfere with the recording of responses. Pins must be inserted in the direction opposite to the movement of the limb: otherwise, the legs slide on the pin, and fixation is not provided.

TOPIC 1. PHYSIOLOGY OF BLOOD

Practical work №1. Determination of the number of red blood cells by test tube method

Red blood cells is the most numerous form of blood elements. The main component of red blood cells is hemoglobin, which is 95% of the dry matter of red blood cells. Normally, mature red blood cells have the form of biconcave disks and do not contain a nucleus.

The main function of red blood cells is the transfer of oxygen from the lungs to the tissues and carbon dioxide from the tissues to the lungs. In addition, red blood cells are involved in many other physiological processes: the adsorption of amino acids, lipids, and toxins, as well as in enzymatic processes. Due to the content of hemoglobin in them, red blood cells play an important role in regulating the acid-base balance of the body. Red blood cells (hemoglobin) account for about 30% of the buffer properties of blood that protect the blood pH from shifting towards acidosis. Red blood cells also have antigenic properties and participate in hemostasis.

Purpose of the work: to determine the number of red blood cells in the blood by the test tube method.

For work, you need: Test blood; centrifuge tubes; Goryaev's camera, microscope; 20 ml dispenser, or capillary from the Sali hemometer; 0.9% NaCl solution; 3% NaCl solution; glass sticks; cotton wool; alcohol; cover glasses; distilled water.

Progress of work. The cover glass is lapped to the counting chamber before the appearance of Newtonian rings and the grid is examined under a microscope. 4 ml of 3% NaCl solution is added to the centrifuge tube. With a pipette from the Sali hemometer or a dispenser, take 20 microl. of blood and, wiping the tip of it with cotton, quickly blow the blood into a test tube. Without removing the pipette from the solution, rinse it with the solution. After that, the contents of the test tube are mixed. Then the prepared counting chamber is filled with diluted blood (Fig.1).

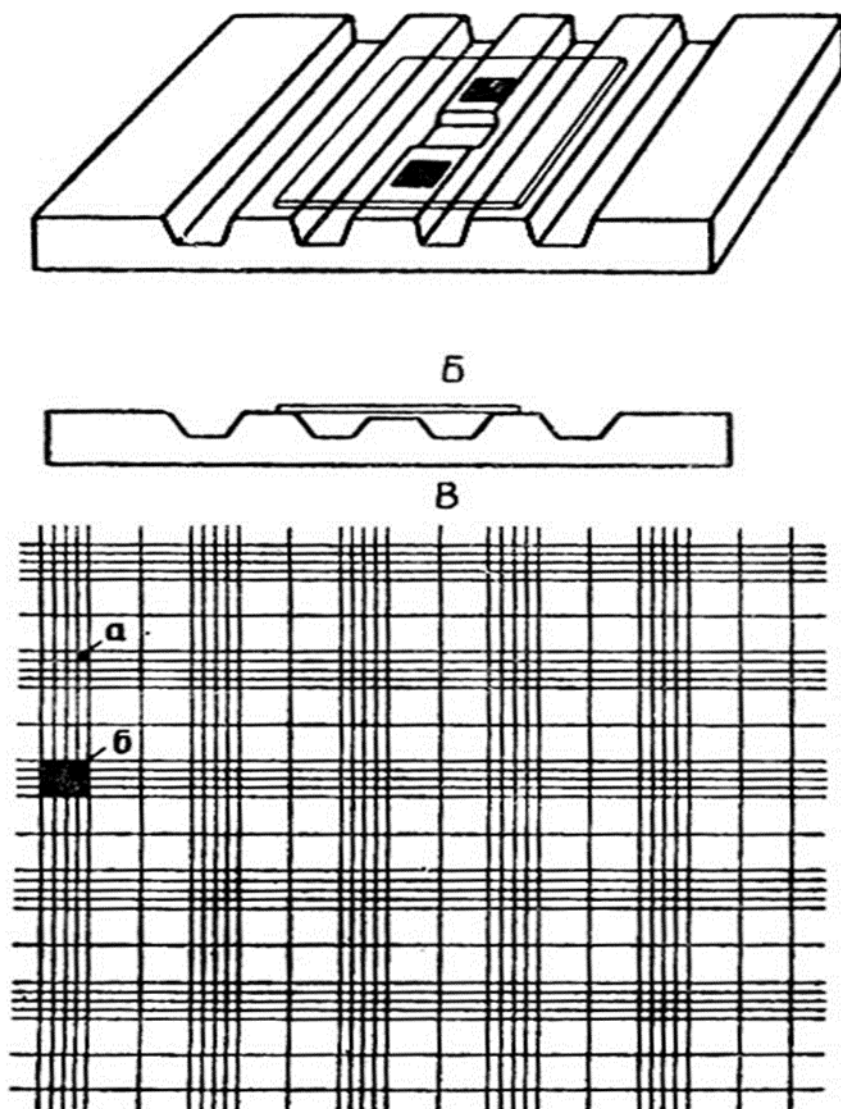


Fig. 1 Goryaev's camera

(a-small square, b-large square);

B – side view; C-grid

After filling the camera, put it under a microscope and count the red blood cells in 5 large squares, divided into 16 small ones, diagonally (top left, right down) (Fig. 2).

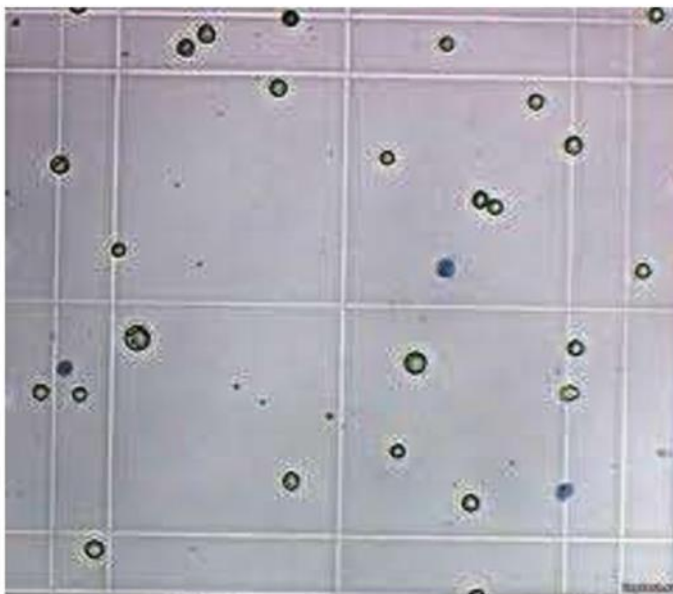
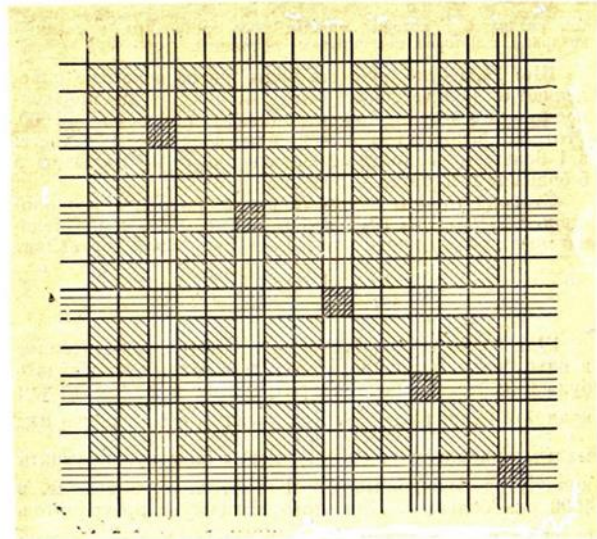
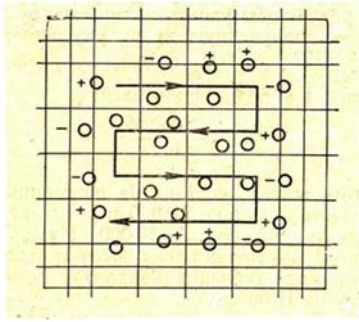


Fig. 2 Counting scheme and type of red blood cells in the Goryaevs camera

Counting is done in a darkened field of view. Count all red blood cells inside the square and on its upper and left sides (Egorov's rule) (Fig. 3).

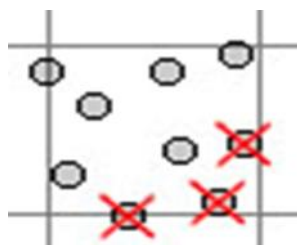


Fig. 3 Yegorov's rule

In a square, the cells that lie inside it, as well as those that touch the left and upper borders are considered. Cells that touch the right and lower borders are not counted.

Then determine the number of red blood cells in 1 microl. by the formula:

$$X = \frac{\vartheta \cdot 4000 \cdot 200}{80},$$

where X- is the desired number of red blood cells;

ϑ – number of red blood cells in 5 large squares;

200-blood dilution;

4000-given that the volume of the chamber over one small square is equal to $1/4000 \text{ mm}^3$, to determine the number of red blood cells in $1 \mu\text{l}$, multiply the found number by 4000;

80 – the number of small squares in 5 large ones.

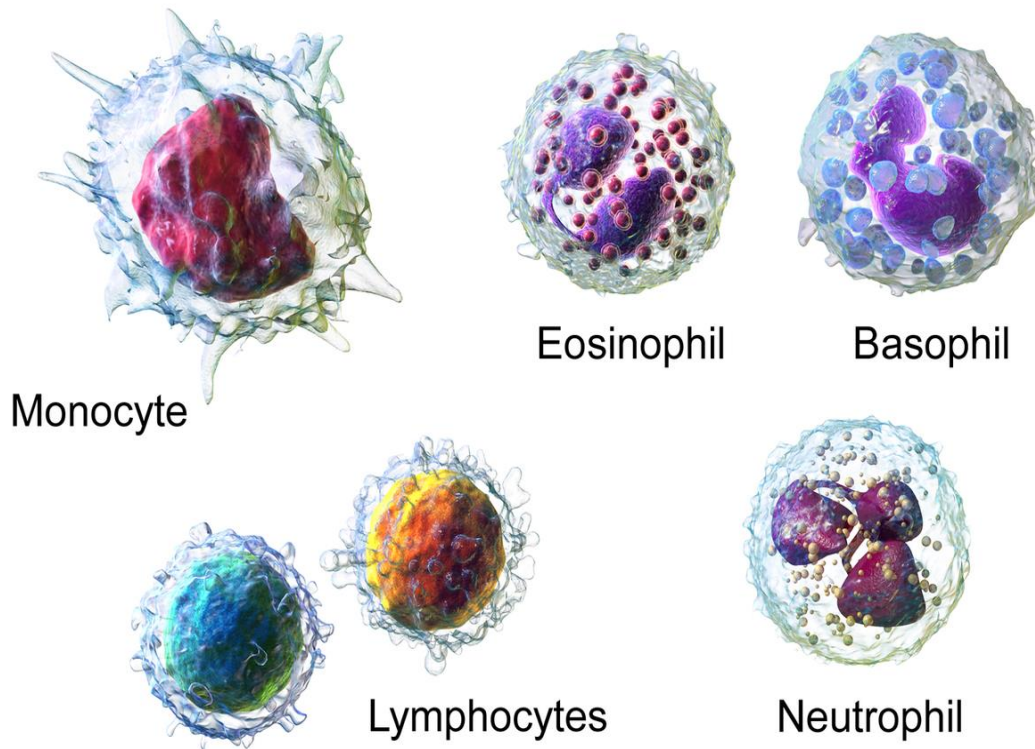
Normally, the number of red blood cells in women is $3.5\text{-}4.5 \times 10^{12}/\text{l}$; in men- $4\text{-}5.5 \times 10^{12}/\text{l}$. an Increase in these indicators is called erythrocytosis, a decrease is called erythropenia.

Recommendations for the design of the work. As a result, specify the obtained number of red blood cells, as well as the limits of fluctuations and the average figure for students of the subgroup. In the analysis, compare the results with the norm. Draw conclusions.

Practical work №2. Determination of the number of white blood cells by test tube method

The study of white blood cells is one of the most common in laboratory practice. Counting the number of white blood cells is included in the General blood test, carried out by all inpatient and outpatient patients and during medical examination.

White blood cells are highly organized cells that perform protective functions due to phagocytic activity, participation in cellular and humoral immunity, and exchange of histamine and heparin. White blood cells are divided:



White Blood Cells

Neutrophils are active microphages. Having the ability to move independently, they form false legs, like an amoeba, capture foreign particles, mainly bacteria and viruses, and destroy them - neutrophil granules contain a rich set of enzymes that can digest almost any biological material. In addition, neutrophils take part in all stages of inflammation, being the first to appear at the site of the inflammatory reaction.

Eosinophils have an antihistamine effect: using the enzyme histaminase, they destroy excess histamine, thus participating in immediate allergic reactions. Like neutrophils, eosinophils are able to move independently, although they move slower than neutrophils. Eosinophils are weak phages and phagocytize mainly coccal forms of bacteria, and also secrete substances that neutralize the poisons of microorganisms, that is, they perform an antitoxic function.

Basophils synthesize histamine, which takes part in allergic reactions and affects the permeability of blood vessels, and contain heparin in the grains, which has an anti-clotting effect. They have weak phagocytic activity.

Lymphocytes by origin and function are divided into T-lymphocytes, which in their development pass through the thymus gland (thymus) and provide cellular immunity, and B-lymphocytes, formed in lymphoid tissue and responsible for humoral immunity, that is, the production of antibodies. T-lymphocytes are the only cells in the body that can distinguish their own protein from a foreign one. T-lymphocytes are of three types: killers, helpers, and suppressors. Killers find alien cells and destroy them.

Helpers (assistants) transmit information about a foreign protein to b-lymphocytes and activate them. T-suppressors inhibit the production of excessive amounts of antibodies.

Monocytes carry out phagocytosis of large microorganisms, old and tumor cells, foreign bodies. After maturation in the bone marrow, monocytes briefly circulate in the blood, and then pass into tissues where they are called macrophages - macrophages of the spleen, bone marrow, alveolar macrophages of the lungs, etc.

Purpose of the work: to Determine the number of white blood cells in the blood by the test tube method.

For work, you need: Test blood; centrifuge tubes; Goryaev's camera, microscope; 3-5% acetic acid solution, tinted with an aqueous solution of methylene blue to color the nuclei of white blood cells and facilitate their counting; 20 microl. dispenser or capillary from the Sali hemometer; 0.9% NaCl solution; glass sticks; cotton wool; alcohol; cover glasses; distilled water.

Progress of work. Prepare the Goryaev's camera for operation by lapping the cover glass so that rainbow rings appear. A centrifuge tube is filled with 0.4 ml of 5% solution's acetic acid, tinted with methylene blue. The acid destroys the shells of the shaped elements, and the dye stains the nuclei of white cells. In this case, red blood cells become invisible and do not interfere with the counting of white blood cells. With a pipette from the Sali hemometer or a dispenser, 20 μ l of whole blood is collected and blown into a test tube. Then dilute blood (dilution 1: 20) fill the chamber and put it under the microscope. Leave the filled counting chamber for 1 minute in a horizontal position for settling of white blood cells. The score starts from the upper-left corner of the Goryaev camera grid. White blood cells are counted at low magnification in 100 large squares, undivided into small ones (Fig. 4). Count white blood cells, as well as red blood cells, according to the Egorov's rule.

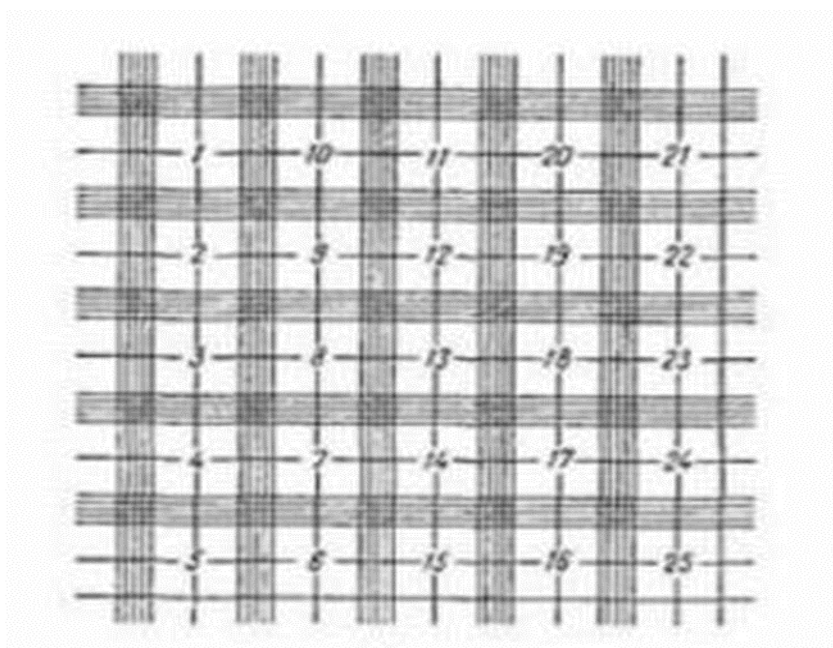


Fig. 4 Squares for counting white blood cells

The number of white blood cells is determined by the formula:

$$X = \frac{a \cdot 4000 \cdot 20}{1600}$$

Where X - is the number of white blood cells in 1mcl of blood;

a - the number of white blood cells counted in 100 large squares;

4000 – Volume conversion factor of 1 μ l, based on the volume of a small square, which is 1 μ l.

1600 – Number of small squares counted;

20 – Dilution of blood.

To convert the number of white blood cells into SI units (in 1 l. of blood), the resulting figure is multiplied by 10^6 .

To determine the content of white blood cells in 1l of blood, the number of white blood cells counted in 100 large squares of the counting chamber is multiplied by 50, divided by 1000 (that is, move the comma 3 characters to the left) and multiplied by 10^9 . The normal number of white blood cells in the blood is $4-9 \times 10^9/l$. an increase in the number of white blood cells is called leukocytosis, a decrease is called leukopenia.

Recommendations for the design of the work. As a result, specify the received number of white blood cells, as well as the limits of fluctuations and the average figure for students of the subgroup. In the analysis, compare the results with the norm. Draw conclusions.

Practical work №3. Determination of the leukocyte formula

The leukocyte formula (the percentage of different types of white blood cells) is calculated in colored blood smears.

The morphology of blood cells is described according to a certain scheme:

1. The size and shape of cells.
2. Nuclear-cytoplasmic ratio.
3. Characteristics of the nucleus: its size, shape, location in the cell (Central, eccentric), color, structure, presence of nucleoli and vacuoles.
4. Characteristics of the cytoplasm: width, color, presence of specific granularity (color of grains, their number and size), presence of non-specific azurophilic granularity, vacuoles, phagocytic elements, perinuclear zone.

Rod-core neutrophils (NP/I) have a size of 10-15 microns. The nuclear-cytoplasmic relationship is shifted towards the cytoplasm. The core is in the form of a bundle or stick, curved in the form of a Latin " S "or Russian letter "C", without significant constrictions, dark purple, with an uneven large-toothed structure. The cytoplasm is pink, contains a specific neutrophilic granularity (pink-purple grains, very small, pulverized, the number of grains is plentiful).

Segmental nuclear neutrophils (NS/I) differ from rod-core neutrophils only in the shape of the nucleus – segmental nuclear neutrophils have a narrow nucleus consisting of 2-5 segments. The connections between the individual segments are a single contour, a thin thread in which lumps of chromatin are not visible, in contrast to the rod-core neutrophil, in which the connections between the individual parts of the nucleus are wider (double contour, bridges, chromatin is visible in them).

Eosinophils (E) have a diameter of 12-15mcm. The nuclear-cytoplasmic relationship is shifted towards the cytoplasm. The core is purple in color and usually consists of two (rarely three) segments that resemble drops in shape. The core structure is uneven and coarse-grained. The cytoplasm is pale pink, contains characteristic eosinophilic granules – large, round, of the same size and shape, pink-red or yellow-red (the color of ket caviar). They fill the entire cytoplasm, so that it is almost invisible.

Basophils (B). The size of the cells is 8-12mcm. The core has a purple color, uneven large-scale structure, undefined shape, sometimes resembling a leaf, it is not clearly visible because of the grain. The cytoplasm is pale pink in color and contains a specific basophilic granularity. Basophilic granules are colored in a dark purple, almost black color, unequal in size (large ones predominate, but there are also small ones), are located throughout the cell, including superimposed on the nucleus. When coloring drugs, some of the granules dissolve, so the basophils look blurry, diffusely colored in purple, and only a few individual granules are visible.

Lymphocytes (L). Usually have a size of 7-10mkm, rarely (in large lymphocytes) – up to 15mkm. The nuclear-cytoplasmic ratio is shifted towards the nucleus. The core is rounded, less often bean-shaped, dark purple in color, located more often eccentrically. The chromatin structure is compact and large-toothed. The cytoplasm is transparent and has the appearance of a narrow blue rim. In broad-cytoplasmic (activated) lymphocytes, a wide zone of gray-blue cytoplasm is visible, which may contain non-specific azurophilic (pink-red, reddish-purple) granularity. All lymphocytes have a pronounced perinuclear zone – the zone of enlightenment around the nucleus.

Monocytes (Mon). The largest peripheral blood cells have a diameter of 12-20mkm. The nucleus occupies an equal part of the cell with the cytoplasm, often located centrally. The core is colored in light purple and has a polymorphic shape: rounded, lobed, lobed, bean-shaped, in the form of a mushroom, butterfly, etc. the Contour of the nuclei is serrated, scalloped. The structure of the core is characteristic: chromatin strands form a wide grid-a loose, evenly non-grid structure. The cytoplasm

is wide, opaque, has a smoky, bluish-gray color, may contain vacuoles, phagocytic elements, dust-like azurophilic granules (Fig. 5).






Leukocyte	Category	Cytoplasm	Nucleus	Function
neutrophil segmented neutrophil, segmenter, seg, polymorphonuclear leukocyte (PMN), poly	granulocyte 	large, pale granules that do not stain either red or blue	three or more lobes	engulf and destroy bacteria
eosinophil eo	granulocyte 	large granules that stain bright pink to red	two lobes	engulf and destroy foreign cells (pollen, animal dander, etc.) and release chemicals that kill parasites
basophil baso	granulocyte 	large granules that stain dark blue to purple	more than one lobe	release histamine at the site of tissue injury, release heparin to limit the size of a forming blood clot
lymphocyte lymph	agranulocyte 	few or no granules	round	engulf and destroy viruses and produce antibodies (immunoglobulins)
monocyte mono	agranulocyte 	few or no granules	kidney bean-shaped	engulf and destroy micro- organisms, cancerous cells, dead leukocytes, and cellular debris

Fig 5. Types of white blood cells in dry blood smears

Purpose of the work: to determine the leukocyte formula in dry blood smears.

For work, you need: Dry blood smears, colored according to Romanovsky-Gimza; immersion oil; microscopes; alcohol; cotton wool; a counter for counting the leukocyte formula.

Progress of work. A blood smear is placed under a microscope and white blood cells are counted in the immersion system. You must view at least 100 cells. The smear is moved either from the top edge to the bottom, then moved to 2-3 fields of view along the edge, then go in the opposite direction; or from the edge, move 5-6 fields to the middle of the smear, then as much sideways, then back to the edge. Move a few fields to the side and repeat the progress again until 100 cells are counted (Fig. 6).

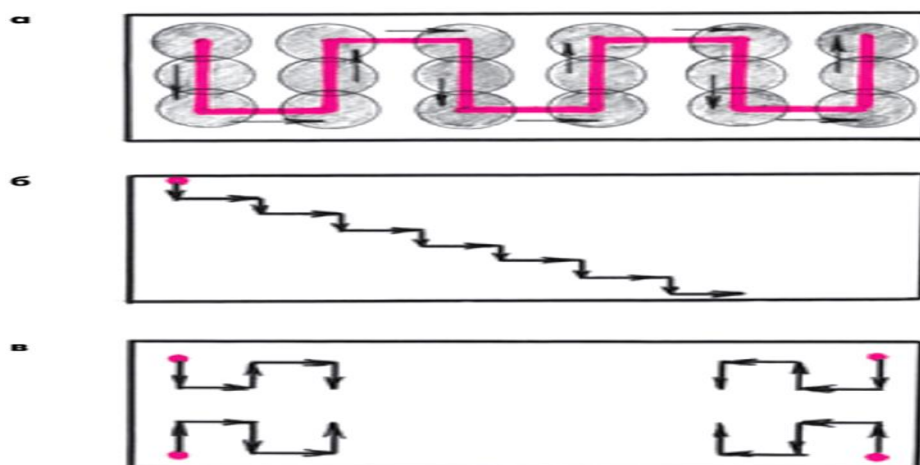


Fig. 6 Options for counting white blood cells in dry blood smears

Four such sections are viewed under a microscope at 4 corners of the smear. Each cell is marked on an 11-key counter, which has keys for all types of white blood cells. After counting the 100 cells, the leucoformula is recorded.

Recommendations for the design of the work. In the results, write the calculated leukocyte formula in the table.

Total number of white blood cells in 1 mm ³ of blood	Granulocytes (%)					Agranulocytes (%)	
	Basophils	Eosinophils	Neutrophils			Lymphocytes	Monocytes
			Juvenile	Band-nuclear	Segment-nuclear		
Normal value							
4000-9000	0-1	0-5	0-1	1-6	47-72	19-37	3-11
Received result							

In addition, taking the number of white blood cells in work 1, calculate the absolute number of individual types of white blood cells. In the analysis, compare these indicators with the norm. Draw conclusions.

Practical work №4. Determination of hemoglobin level by Sali method

Hemoglobin [from the Greek. haima blood + globus ball] – blood pigment contained in red blood cells and giving the blood a red color. The main functions of

hemoglobin are the transfer of oxygen from the lungs to the tissues and carbon dioxide from the tissues to the lungs, as well as maintaining a constant blood pH.

By chemical structure, hemoglobin (Hb) belongs to complex proteins-chromoproteins. Its prosthetic group, which includes bivalent iron, is called heme, and the protein component is called globin. The hemoglobin molecule contains 4 hemes and one globin. Globin consists of two pairs of polypeptide chains, which, depending on the amino acid composition, are designated as α , β , γ and δ chains.

Hemoglobin types differ in the structure of globin polypeptide chains. There are physiological and pathological types of hemoglobin.

The physiological types of hemoglobin include hemoglobin A, F, and P.

Hb A-adult hemoglobin [from English. Adult adult]. Hemoglobin A consists of two α -and two β -chains ($\alpha_2\beta_2$). There are several fractions of hemoglobin A: A1, A2, A3. In normal adults the fraction A1 is a basic and amounts to 96-98%, fraction A2 is less than 3%, A3 – in the form of traces.

Hb F-fetal hemoglobin [from lat. fetus the fetus]. This type of hemoglobin consists of $\alpha_2\gamma_2$ chains and is contained in the fetus from 3 months. In newborns, the content of HbF is about 20%, the rest of the hemoglobin is represented by HbA. In the future, HbF continues to decrease and by 4-5 months reaches the values of an adult-1-2%.

Hb P-primitive hemoglobin, is contained in the fetus at the early stages of embryonic development (up to 3 months). Hemoglobin compounds are physiological and pathological. The physiological compounds of hemoglobin include oxyhemoglobin (a compound of hemoglobin with oxygen), carbohemoglobin (a compound of hemoglobin with carbon dioxide) and reduced hemoglobin-a compound of hemoglobin with a water molecule.

In the lungs, hemoglobin combines with oxygen to form oxyhemoglobin, which is carried with the current of arterial blood to all organs and tissues. Here, oxyhemoglobin dissociates (breaks down) into oxygen, which is used by cells for oxidative processes, and hemoglobin, which attaches a water molecule and becomes reduced hemoglobin. The free valences of the reduced hemoglobin bind to carbon dioxide. The resulting carbo-hemoglobin with venous blood flow is delivered to the lungs, where it dissociates into its constituent parts. Carbon dioxide is released with the exhaled air, and hemoglobin attaches a new portion of oxygen and the whole process is repeated again.

Pathological hemoglobin compounds include carboxyhemoglobin, methemoglobin, and sulfhemoglobin. Pathological hemoglobin compounds are very resistant, not capable of dissociation, so they can not carry oxygen and when they are formed in the body, oxygen deficiency develops.

Methemoglobin is a compound of hemoglobin with oxygen, in which bivalent iron is replaced by trivalent iron. The formation of methemoglobin occurs when

poisoning with amido and nitro compounds of the benzene series. A specific laboratory sign of methemoglobin formation is the presence of Heinz cells in red blood cells.

Carboxyhemoglobin is a compound of hemoglobin with carbon monoxide (CO). Carbon monoxide is a toxic product of incomplete combustion of carbon-containing substances. The formation of carboxyhemoglobin in the blood occurs when sanitary, hygienic requirements are not met, and technological processes are violated in blast furnace and open-hearth production, auto garages, furnace heating, accidents and explosive works in mines, and fires. The affinity of hemoglobin for carbon monoxide is several hundred times greater than for oxygen, so even a small concentration (0.07%) of carbon monoxide in the air binds more than 50% of the hemoglobin available in the body and is fatal.

Sulfhemoglobin is detected when using sulfonamides and when poisoning with benzene compounds simultaneously with the formation of methemoglobin.

Purpose of the work: to determine the level of hemoglobin in the blood by the Sali method.

For work, you need: test blood; Sali hemometer; microscope; 20 ml dispenser, or capillary from Sali hemometer; 0.9% NaCl solution; 0.1 n HCl solution; glass sticks; cotton wool; alcohol; distilled water; pipettes.

Progress of work. In the middle tube of the hemometer pour 0.1 n p-p HCl to the lower mark (Fig. 7).



Fig. 7. Sali Hemometer

By a pipette or a dispenser take 20 microl. of blood and blow it to the bottom of the tube so that the top layer remains unpainted. Without removing the pipette from the acid rinse it. Then the contents of the test tube are stirred by tapping the bottom of the test tube with your finger and left it for 5-10 minutes. During this time, hydrochloric acid hemin is formed. In the future, adding a drop of distilled water, bring the color of the contents of the test tube to the color of the liquid in the side tubes of the hemometer. According to the lower meniscus of the fluid in the middle tube, the hemoglobin content in g % or g/l is noted. If the test tube is graded in g%, convert the value to g/l, the resulting figure must be multiplied by 10.

Normal hemoglobin content in the blood: men 130-160 g / l; women 120-140 g/l.

A decrease in the concentration of hemoglobin in the blood is the main laboratory sign of anemia. A moderate decrease in hemoglobin content is more common in iron-deficiency anemia, and a significant decrease is typical for acute blood loss, hypoplastic and B12-deficient anemia. However, for the diagnosis of anemia, it is not enough to detect a decrease in the concentration of hemoglobin – this only establishes the presence of anemia. To clarify the nature of anemia, additional studies are required (determining the number of red blood cells, their morphology, calculated red blood cell indices, the number of reticulocytes, etc.). **Recommendations for the design of the work.** In the results, record the resulting hemoglobin concentration. Compare the result with the norm.

Practical work №5. Determination of blood group and Rh-supplies using standard isohemagglutinins serum

The ABO system is the main blood compatibility system. It is represented by agglutinogens A and B, which are glycoproteins located on the surface of red blood cells, and agglutinins alpha and beta, belonging to the class of IgM immunoglobulins and circulating in the blood plasma. Depending on the combination of these agglutinogens and agglutinins, 4 blood groups are isolated according to the ABO system.

Blood group	Red blood cell agglutinogens	Plasma agglutinins
I	0	$\alpha\beta$
II	A	β
III	B	α
IV	AB	0

The first (I) blood group (the most common in the European population, 42 % of the population) is also called the O-group, with no agglutinogens A or B on the surface of red blood cells, and agglutinins alpha and beta are detected in the plasma.

The second (II) blood group (37%) is also called A-group, agglutinin A is present on the surface of red blood cells, and agglutinin beta is detected in the plasma.

The third (III) blood group (13 % of the population) is also called B-blood group, agglutinin B is present on the surface of red blood cells, and agglutinin alpha is detected in the plasma.

The fourth (IV) blood group (the rarest, only 8 % of the population) is also called AB-blood group, there are agglutinogens of both types A and B on the surface of red blood cells, and there are no agglutinins alpha and beta in the plasma.

The RH system also consists of several antigens, the main of which is called antigen-D, or RH factor. Approximately 85% of people can detect RH factor (RH-positive blood) on the surface of red blood cells. Human blood belonging to a certain group according to the ABO system and the rhesus system is genetically determined and does not change throughout life. Blood groups are determined by the antigenic properties of red blood cells, which are set using standard serums containing known agglutinins.

Purpose of the work: to Determine blood group and RH affiliation with standard isohemagglutinins sera.

For work, you need: Test blood, standard serums of 4 blood groups and the standard universal reagent antiresus anti-D, marked plates, slides, alcohol, cotton swabs, 0.9 % sodium chloride solution, pipettes, dispenser.

Progress of work: On a marked plate (tablet) (Fig. 8) apply 1-2 drops of standard isohemagglutinating serum of groups I, II, III, and IV to the appropriate sector.



Fig. 8 Porcelain plate (tablet) for determining blood groups and Rhesus factor

Serums from each vial should be taken with a separate pipette. The dispenser should take a little blood (the amount of test blood should be approximately 10 times less than the amount of serum with which it is mixed), add a drop of group 1 serum and mix well. Replacing the nozzle, add blood to a drop of group 2 serum, etc. Then mix the blood with the standard serum with a glass stick. Slightly swaying the plate, monitor the agglutination of red blood cells (Fig. 9).



Fig. 9. Scheme for determining the blood group using a tablet

Within 3 minutes after the onset of agglutination, add 1 drop of 0.9 % sodium chloride solution to exclude non-specific agglutination. The final result is defined in 5 minutes. Then the RH factor is determined similarly. On a marked plate (tablet) (Fig. 8), apply to the sector 1 drop of the standard universal reagent antiresus anti-D. the dispenser should take a little blood (the amount of blood being tested should also be approximately 10 times less than the amount of serum with which it is mixed), add a drop of serum and mix it well. Then, with a glass stick, the blood is mixed with the standard serum. Slightly swaying the plate, monitor the agglutination of red blood cells (Fig. 9)

Recommendations for the design of the work. Determine the groups of blood in the investigated samples. Write the composition of its agglutinogens and agglutinins. Draw a picture of the interaction of serum and blood for each group and rhesus group. Explain the reason for the appearance or absence of agglutination. Draw conclusions.

Topic 2. PHYSIOLOGY OF THE CARDIOVASCULAR SYSTEM

Practical work № 1. Recording of frog heart contractions

The purpose of the work: To master the technique of cardiography and to study the phases of heart activity.

The activity of the heart consists of three interrelated phases: atrial systole, ventricular systole, and general pause.

With 75 heart contractions per minute, the duration of the human atrial systole is about 0.1 seconds, the ventricular systole is 0.3 seconds, the total pause is 0.4 seconds, and the entire cardiac cycle is 0.8 seconds.

For work, you need: a board for fixing frogs, dissecting set, the lever of Engelman, universal tripod, kymograph, electrometrical with electromagnetic timer, the ringers solution for cold-blooded, wool, frog.

Progress of work: The frog is fixed with its belly up, pinning its legs to the preparation plate (made of foam, cork or wood with a cork inserted at the corners of the plank) with pins. After that, proceed to the opening of the chest cavity.

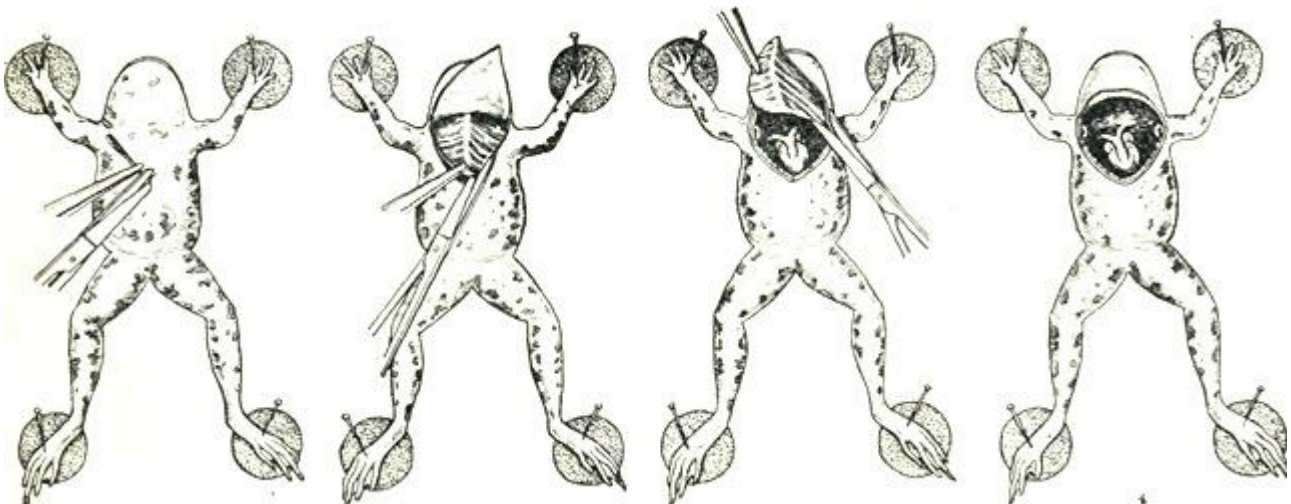


Fig. 10. Diagram of dissection of the frog's thoracic cavity

Make an incision of the skin 0.5 cm caudal to the end of the sternum, after which the skin is cut in the direction of the shoulder joints (Fig. 10). Grab the sternum with tweezers, pull it up and make an incision of the muscles at its caudal end. Dissect the muscles in the direction of the shoulder joints (Fig. 10). The resulting musculoskeletal flap, carefully lifting, is separated from the underlying tissues and cut off at the base (Fig.10). A pulsing heart can be seen in the resulting wound. With the help of eye anatomical tweezers and small scissors, the pericardium is opened and the frenulum of the heart (a thin cord that fixes the posterior surface of the heart to the underlying

tissues) is taken for ligature. To do this, tweezers are placed under the ventricle and lift the heart with them.

With tweezers, grab the ligature and stretch it under the bridle. To prevent the latter from breaking, it should be bandaged as close to the heart as possible. They cross the bridle and, lifting the heart by it, grab its top with a clothespin. Attach the clothespin with a thread to the Engelmann lever (Fig. 11) to get the maximum swing of the lever, and proceed to record the work of the heart.



Fig. 11. Diagram of installing a clothespin on the heart of a frog

Assemble the installation according to the scheme shown in Fig. 12, install the Engelmann lever in a horizontal position, lowering or raising the plank with the frog.

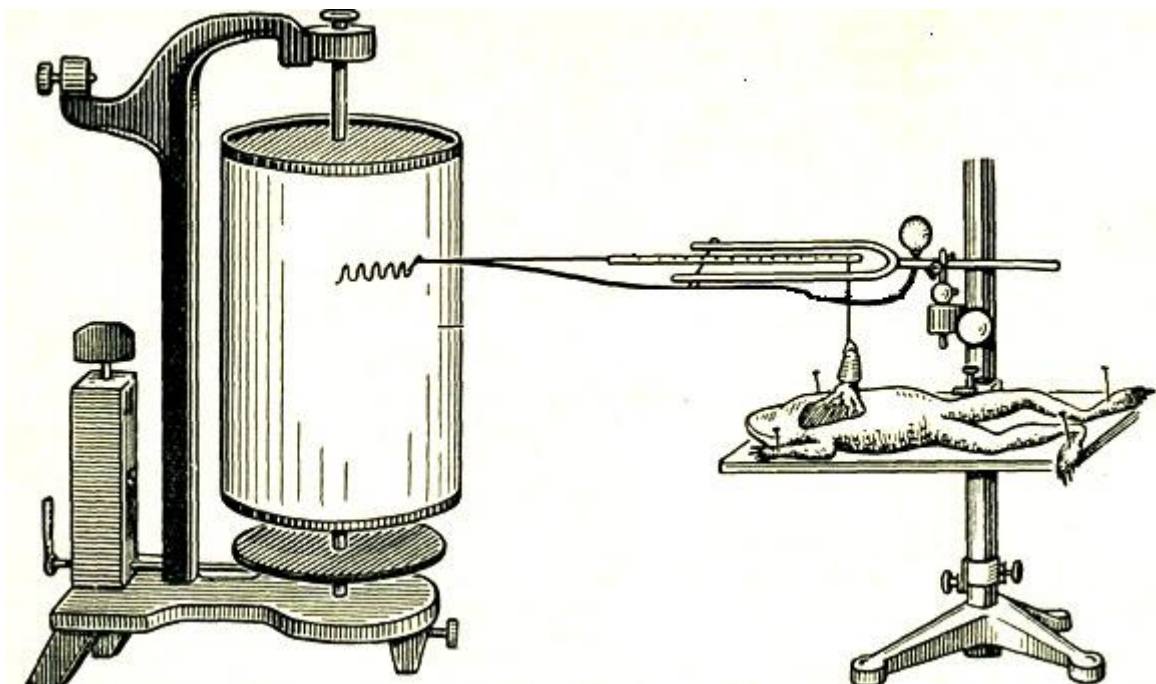


Fig. 12. Installation diagram for recording the frog heart mechanogram

Press the lever Peschici of Engelman to the paper, include kymograph, conduct a recording of the heart's contractions (Fig.13)



Fig. 13. Recording of the frog's heart rate

1 - atrial systole; 2-ventricular systole; 3 - period of relaxation of the ventricular muscles; 4-general diastole of the heart

In the process, it is necessary to irrigate the heart with Ringer's solution to prevent it from drying out.

Recommendations for the design of the work: 1. Draw a diagram of the experience. 2. Cut out and paste the cardiogram into the notebook. 3. On the cardiogram, mark the phases of heart contractions. 4. Calculate the duration of the cardiac cycle and each of its phases separately.

Practical work №2. Study of the degree of automatism of various parts of the frog heart. Ligatures of Stannius.

The purpose of the work: To study the degree of automatism of various parts of the heart.

The heart muscle has the ability to contract without external influences under the influence of impulses arising in it. This property is called automatic. Thanks to this property, the heart, separated from the body, retains the ability to contract. The automatism of the heart is caused by rhythmic excitations that occur in the atypical muscle tissue of the heart, called the conducting system, through which these excitations spread from one part of the heart to another. In the conducting system of the frog heart, there are several departments that have different degrees of automatism:

- 1) the Remak node located between the venous sinus and the atria, which has the highest degree of automatism and is the driver of the heart rhythm (Fig. 14);
- 2) Bidder's node, located in the atrial septum at the border with the ventricles, from which Purkinje fibers go into the ventricular wall;
- 3) Dogel nodes, located below the previous node on the nerve stems extending from it; their role in the heart's automatism is not fully understood.

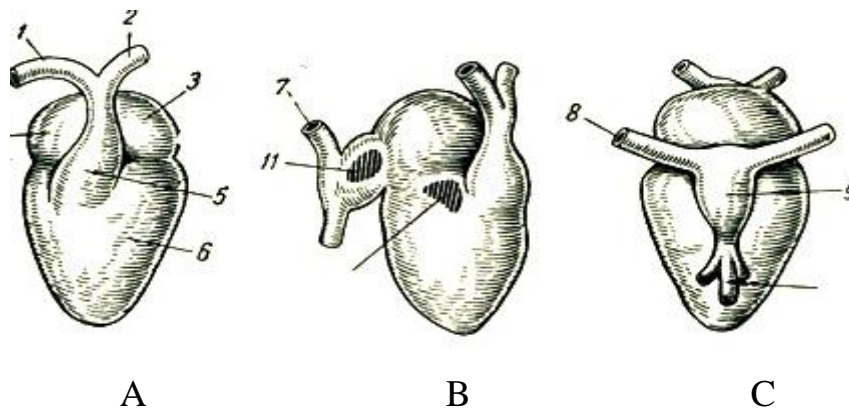


Fig. 14. Anatomical diagram of the frog heart

A - view from the abdominal side; B - side view; C - view from the back

1-left aortic arch; 2 - right aortic arch; 3-left atrium; 4 - right atrium; 5 - aortic bulb; 6-ventricle; 7-right anterior vena cava; 8-left anterior vena cava; 9-venous sinus; 10-posterior vena cava; 11-Remac node; 12-Bidder node

To determine the role of each node of the conducting system and their functional connections, they resort to applying ligatures that separate the heart departments from each other, and based on the results of the experiment, they judge the role of the nodes located in these departments.

For work, you need: a preparation kit, a ligature hook, thread, a stopwatch, a Ringer's solution for cold-blooded people, cotton wool, a frog.

Progress of work: Immobilize the frog and pin the belly up to the dissecting board. They bare the heart (work № 1). The number of heart contractions is counted, then a thread is inserted under the venous sinus and a ligation is made at the border between the sinus and the atria (Fig. 15).

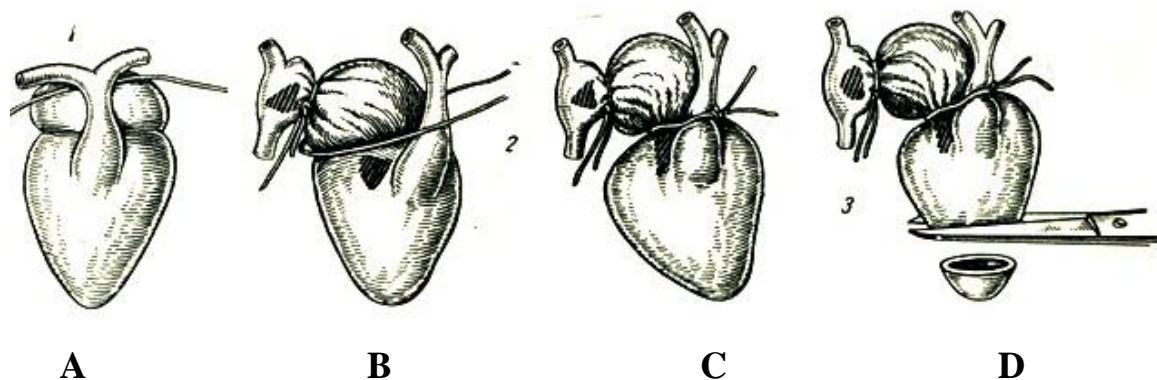


Fig. 15. Switching off individual nodes of the conducting system using Stannius ligatures

a-ligature for ligation of the venous sinus; b-the venous sinus is separated by a ligature from the atria; a second ligature is connected to separate the atria from the ventricles; c - the atria from the ventricles are separated; d-cutting off the apex of the heart.

The rhythm of the venous sinus contractions usually does not change, and the atria and ventricle stop or begin to contract in a more rare rhythm. Count the number of contractions of the venous sinus and note the state of the atrioventricular heart. If, after applying the first ligature, the contractions of the atria and ventricles do not recover on their own, then a second ligature is applied, which will irritate the Bidder node and cause its automatic activity. The second ligature is applied to the atrioventricular sulcus (Fig. 8). Now, only the ventricle or only the atrium will contract, depending on how the ligature lies in relation to the Bidder node (below the node or above it). If the atrial and ventricular automatics recovered independently after the first ligature was applied, then the second ligature is applied not along the atrioventricular sulcus, but slightly higher, which will more clearly show the leading role of the atrioventricular node. Also count the number of contractions of the working parts of the heart. Then a third ligature is applied to the lower third of the ventricle and the condition of the apex of the heart is noted. Usually, the apex of the heart does not contract. In order to make sure that the ability of the apex of the heart to contract is preserved, it is cut off (Fig. 8) and placed on a slide with a drop of Ringer's solution. Irritating the tip of the heart with needle pricks, note its reaction.

Recommendations for the design of the work: 1. Draw a diagram of the ligature overlay on the frog's heart. 2. Make a table of changes in the frequency of contractions of the venous sinus, atria and ventricles of the heart after applying each ligature. 3. Explain the change in the frequency of contractions of different parts of the heart after applying the first, second and third ligature.

Practical work № 3. Features of excitability of the heart and extrasystole

The purpose of the work: To get acquainted with one of the most important properties of the heart muscle - excitability.

During contractions in the heart muscle, phase changes in excitability are observed. Figure 16 shows a schematic representation of the action potential, excitability and contraction phases. Immediately after the start of arousal, the heart muscle completely loses excitability (absolute refractory phase). After this phase, the excitability gradually recovers, but all the time remains below normal-the relative refractory phase. After the relative refractory phase, a short-term phase of increased excitability of the heart muscle occurs, and then its excitability returns to its original level.

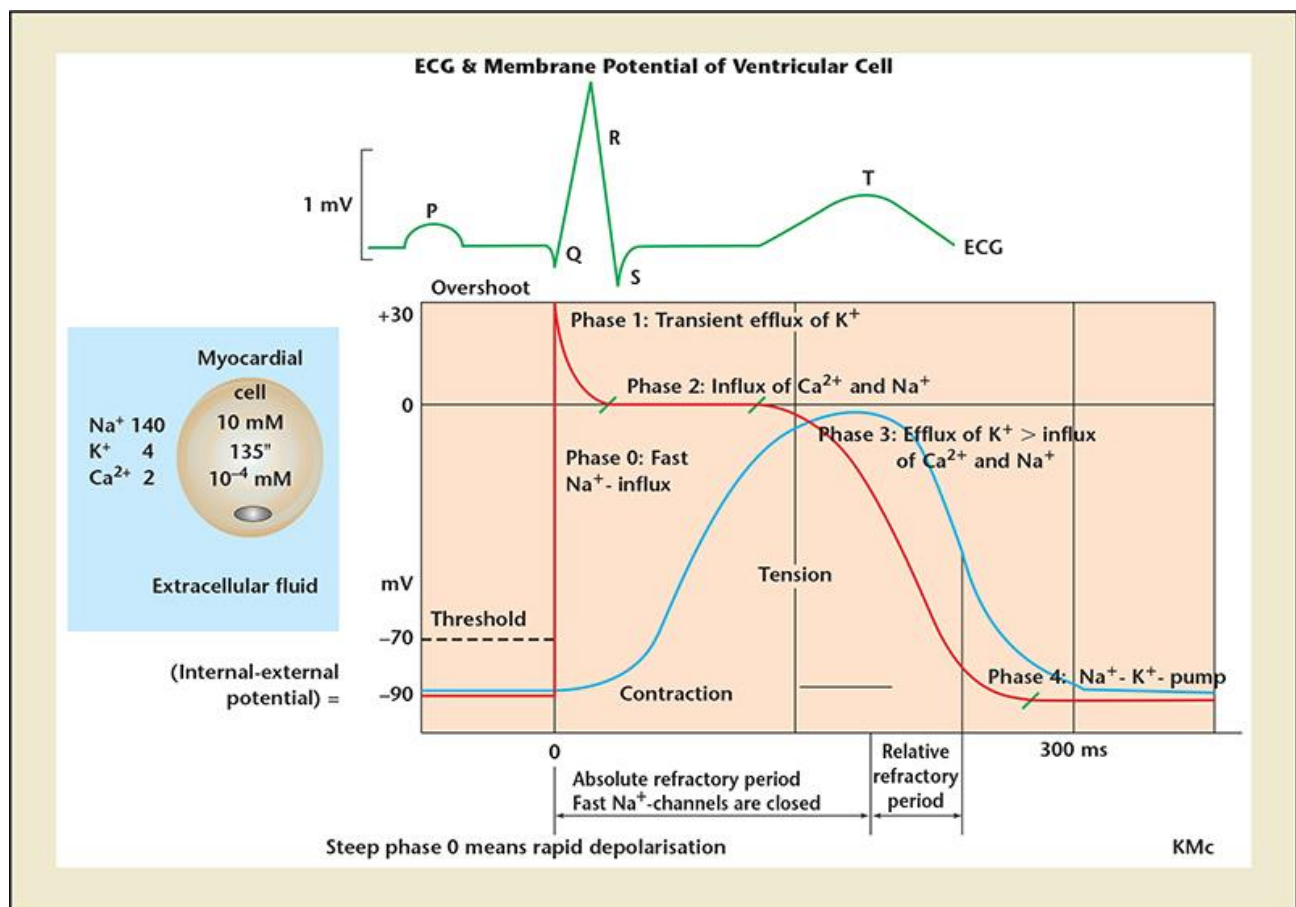


Fig. 16. Action potential, excitability and contraction phases of the heart muscle

Due to the relatively long duration of the absolute refractory phase, the heart responds to frequent intermittent irritations with rhythmic contractions and does not go into a state of tetanus - a long shortening. If you apply an extraordinary irritation during the relative refractory phase, the heart muscle can respond with extraordinary contractions, which are commonly called extrasystole. After the ventricular extrasystole, the time of its diastole (compensatory pause) is prolonged (Fig. 17). The mechanism of the compensatory pause is as follows: the next pulse, which originated in the sinus node, comes to the ventricle when it is in the absolute refractory phase as a result of

extrasystole and therefore does not respond to this pulse. Figure 10 shows a diagram explaining the occurrence of extrasystole and compensatory pause.

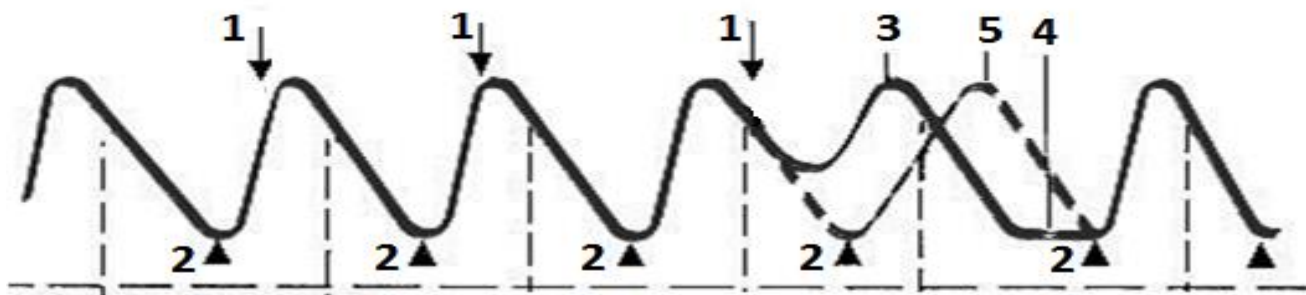


Fig. 17. Extrasystole and compensatory pause

1 – The moments of application of an electrical stimulus, 2-the moments of receipt of impulses from the sinus node, 3-extrasystole, 4-compensatory pause, 5-dropped contraction

For work, you need: dissecting kit, kymograph, universal tripod, dissecting Board, the lever of Engelman, pacemaker electrodes, ringer solution for cold-blooded, pipette, wool, frog.

Progress of work: Immobilize the frog and prepare it for cardiography (work № 1). Attach the stimulator electrodes to the base of the heart ventricle (Fig. 18). Write the initial contraction of the heart.

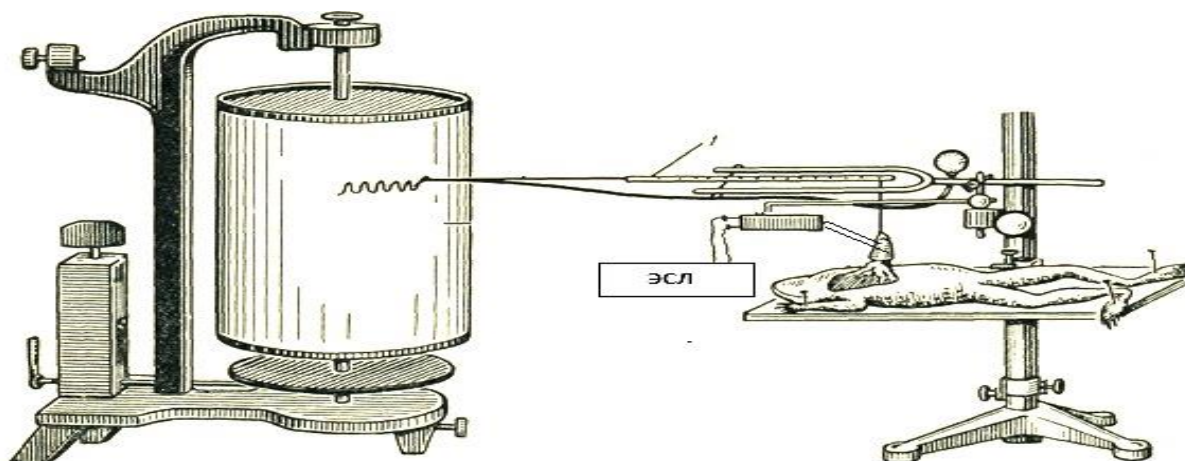


Fig. 18. Scheme of the installation for registering the extrasystole

After making sure that the recording is going well, pressing the "irritation" button on the electric stimulator, select the optimal voltage. Keeping a record of heart contractions, single stimuli are simultaneously applied (pulse duration 1 Ms, frequency 1-5 uti/s) in the following phases of the cardiac cycle: in the middle of the ventricular

contraction, at the height of the ventricular contraction, in the middle of its relaxation and during diastole.

Recommendations for the design of the work: 1. Draw a diagram of the experience. 2. Insert the resulting cardiogram into the notebook. 3. Mark the extrasystole and compensatory pause on the cardiogram.

Answer the questions: 1. What are the relative and absolute refractory phases of the heart? 2. What is the significance of the refractory period for the functions of the heart? 3. What is a beat and a compensatory pause?

Practical work № 4. Human electrocardiography

Electrocardiography is a method of recording the electrical potentials of a working heart. The electrocardiogram is a curve consisting of five prongs – PQRST. The P wave reflects the atrial excitation and is the algebraic sum of the potentials that arise when the right and left atria are excited. The QRST teeth are a ventricular complex that reflects the process of ventricular excitation. In the normal position of the heart, the ECG has the greatest amplitude of the teeth in the second lead, the smallest in the third. To explain the different voltage of the teeth, Einthoven proposed schematically depicting the human body in the form of a triangle. The electrical axis of the heart is located in the center of the triangle parallel to its left side. The projection of this axis on the side of the triangle corresponds to the potential difference recorded by the galvanometer (Fig. 19).

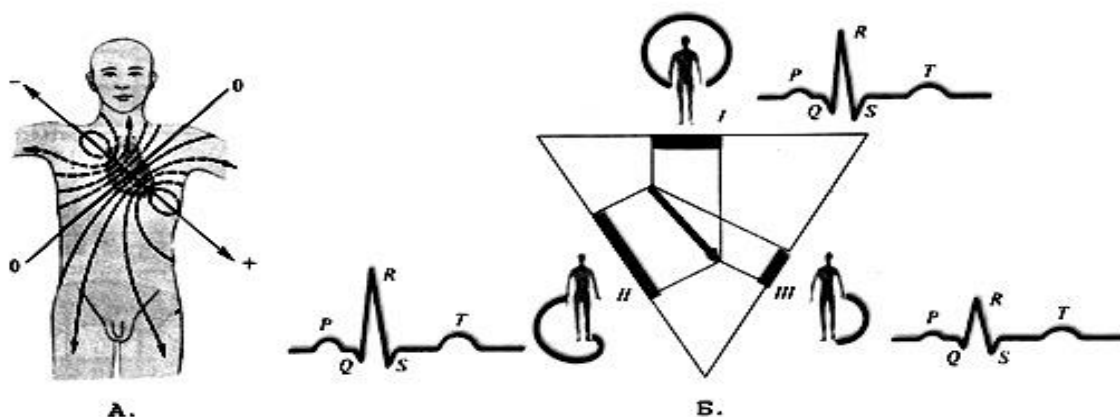


Fig. 19. A - the electric field of the heart during the formation of the electrocardiogram; B - the Einthoven triangle and the projection of the electric axis of the heart on the sides of the triangle: I, II, III leads

Three standard leads are used for ECG recording: I-right arm – left arm; II-right arm-left leg; III - left arm – left leg. Thoracic leads and unipolar leads from the extremities are also used.

The single-channel portable electrocardiograph "Malysh", designed for ECG registration, consists of the following main components: an amplifier, a tape drive mechanism, a stabilizer and a power supply unit (batteries). Push-button switches are located on the front panel of the amplifier. The "On" button is used to turn on the power supply voltage of the device. Buttons "50" and "25" - to select the speed of stretching the paper tape-25 and 50 mm/s. The "Record" button is designed to supply voltage to warm up the pen and turn on the tape-drawing mechanism. The "1:2" button provides a step-by-step adjustment of the sensitivity in the range of 5 mm/mv; the "2:1" button - 20mm/mv.

The purpose of the work: To get acquainted with the methodology of human electrocardiography and the analysis of the electrocardiogram.

For work, you need: An electrocardiograph, gauze, saline solution.

Progress of work. Before switching on, ground the electrocardiograph and turn on the device. Fix the electrodes on the test subject with rubber bands (between the skin and the electrodes, a gauze pad is placed, previously moistened with a solution of table salt). Connect the colored leads of the lead cable to the superimposed electrodes in the following order: right hand - red, left hand – yellow, left leg – green, right leg – black. To remember the location of the electrodes by color, remember (the rabbit chews green garlic, or red, yellow, green, black.)

After applying the electrodes to the patient and connecting the lead cable, do the following: set the lead switch to the "K" position, turn on the "Record" button and press the "1mB" button. The calibration signal will be recorded. Set the lead switch to the "1" position, turn on the "Record" button, record the required number of ECG cycles (Fig. 20), turn off the "Record" button. After the operation is finished, press all the buttons on the switch on the front panel and turn off the device.

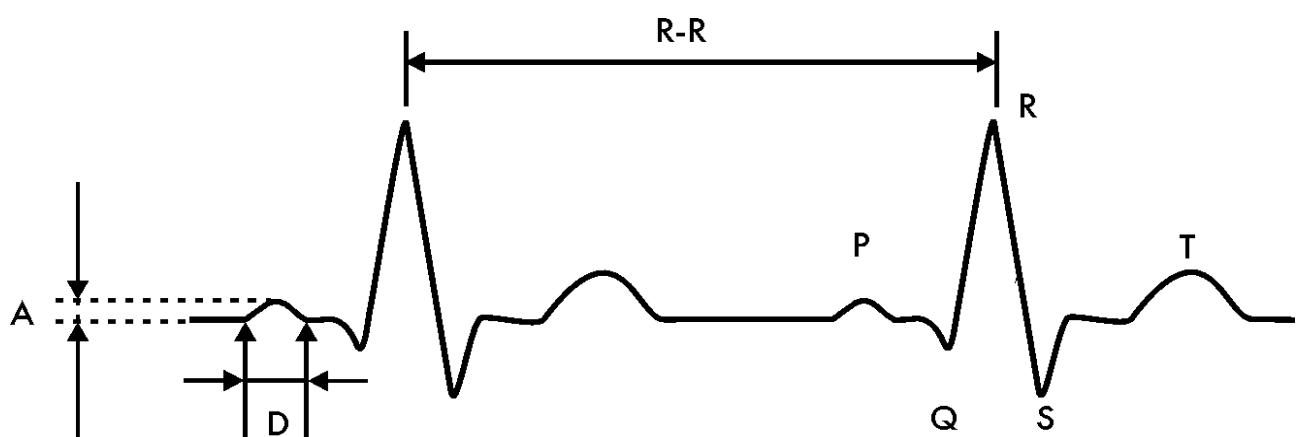


Fig. 20. Diagram of a normal human electrocardiogram

Perform an analysis of the ECG in three leads, and then write the data in Table 1.

Normal values of the electrocardiogram

The amplitude of the teeth (A) in mV and their duration (D) in s									
P		PQ	Q	R	S	QRS	T		QRST
A	Д	Д	A	A	A	Д	A	Д	Д
0-0,25	0,06-0,11	0,12-0,18	0-0,25	0,15-2,4	0-0,6	0,06-0,09	0,05-0,3	0,05-0,25	0,36

Recommendations for the design of the work: Conclusions are drawn by comparing the obtained ECG values with the norm.

Practical work № 5. Measurement of arterial blood pressure

Blood pressure is the blood pressure in a person's major arteries.

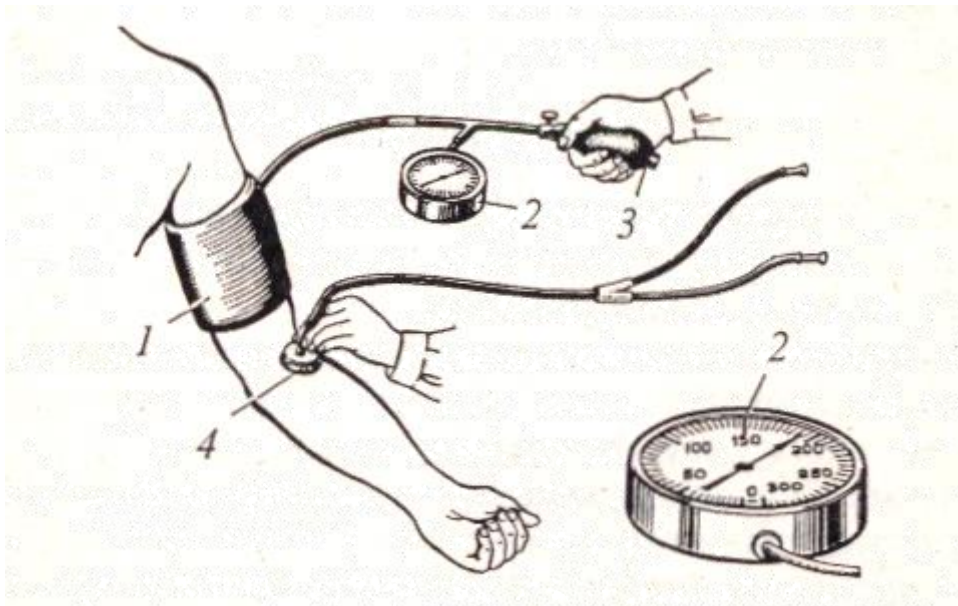


Fig. 21. Scheme of measuring blood pressure in humans by the Korotkov method

Blood pressure is measured in millimeters of mercury, abbreviated mm Hg. The value of the value of blood pressure 120/80 means that the value of systolic pressure is 120 mm. Mer. Col., and the value of diastolic blood pressure is 80 mm. Mer. Col.

There are two indicators of blood pressure:

- systolic (upper) blood pressure (SP) is the level of blood pressure at the time of maximum contraction of the heart, describes the condition of the myocardium of the left ventricle and is equal to 100-120 mm. Mer. Col.
- diastolic (bottom) blood pressure (DP) is the level of blood pressure at the moment of maximum relaxation of the heart, characterizes the tone of the arterial walls and is equal to 50-80 mm. Mer. Col.

The difference between the values of systolic and diastolic pressure is called pulse pressure (PP). It shows how much systolic pressure exceeds diastolic pressure, which is necessary for the opening of the semilunar valve of the aorta during systole. Normally, the pulse pressure is 35-55 mm. Mer. Col. Only under such conditions, during the systole of the left ventricle, the valve opens completely, and the blood enters the large circulatory circle. If the systolic pressure becomes equal to the diastolic pressure, the movement of blood will be impossible and death will occur. Increase in pressure for every 10 mm. Mer. Col. increases the risk of developing cardiovascular diseases by 30%.

The amount of blood pressure depends on three main factors:

- the frequency and strength of the heart rate;
- the values of peripheral resistance, i.e. the tone of the walls of blood vessels, mainly arterioles and venules;
- the volume of circulating blood.

The blood pressure of a healthy person is a fairly constant value, but it is always subject to small fluctuations depending on the phases of heart activity and respiration. Blood loss leads to a decrease in blood pressure, and the transfusion of a large amount of blood increases blood pressure. The amount of pressure depends on the age. In children, blood pressure is lower than in adults, because the walls of the vessels are more elastic.

Methods for measuring blood pressure

To measure blood pressure, direct and indirect methods are currently used:

The indirect Korotkov method was developed by the Russian surgeon N. S. Korotkov in 1905 and allows you to measure blood pressure with a very simple device. The Korotkov method is based on measuring the amount of pressure that is necessary for the complete compression of the artery and the cessation of blood flow in it.

Description of devices:

To measure blood pressure by the Korotkov method, mechanical and electronic meters with light and digital indication are used. Mechanical meters consist of a mechanical pressure gauge, a cuff with a pear and a phonendoscope. These devices are mainly used in professional medicine, since without special training, errors in the determination of indicators can be allowed.

For home use, electronic meters are most suitable. They are semi-automatic (fig. 22, a) and automatic (fig. 22, b). Their use does not require any prior training and, following simple guidelines, allows you to get accurate blood pressure data by pressing a single button. The principle of their operation is based on the registration of the device pulsations of air pressure that occur in the cuff, when blood passes through the compressed section of the artery.

a.



b.



Fig.22. Blood pressure measuring device: (a) semi-automatic, (b) automatic

The purpose of the work: To get acquainted with the method of measuring blood (arterial) pressure in humans according to the Korotkov method and learn how to determine it in humans.

For work, you need: a blood pressure cuff, a stethoscope, an examinee.

Progress of work:

1. Wash your hands.
2. Treat the phonendoscope membrane with 70% alcohol by double wiping.
3. Place the patient's hand correctly: in the extended position, palm up, the muscles are relaxed.
4. Place the cuff on the patient's bare shoulder 2-3 cm above the elbow bend; clothing should not squeeze the shoulder above the cuff; secure the cuff so tightly that only one finger passes between it and the shoulder.
5. Connect the pressure gauge to the cuff. Check the position of the pressure gauge arrow relative to the zero point of the scale.
6. Feel the pulse in the area of the ulnar fossa and put the phonendoscope in this place.
7. Close the valve on the pear and pump air into the cuff: pump air until the pressure in the cuff, according to the pressure gauge, exceeds by 25-30 mm. Mer. Col. the level at which the pulsation of the artery has ceased to be determined.
8. Open the valve and slowly release the air from the cuff. At the same time, use the phonendoscope to listen to the tones and monitor the readings of the pressure gauge scale.
9. Note the amount of systolic pressure when the first distinct sounds appear above the brachial artery.
10. Note the value of the diastolic pressure, which corresponds to the moment of complete disappearance of the tones.
11. Write down the blood pressure measurement data in the form of a fraction (in the numerator – systolic pressure, and in the denominator – diastolic pressure), for example, 120/75 mm. Mer. Col.

Remember! Blood pressure should be measured two to three times on both hands at intervals of 1-2 minutes, and the lowest result should be considered a reliable blood pressure. The air from the cuff must be released completely each time.

Practical work №6. Sphygmography

Purpose of the work: Get acquainted with the technique of registering a sphygmogram. Perform a visual and quantitative analysis of the sphygmogram at rest and after exercise.

For work, you need: Device for wireless registration of biological signals "Bio-rod ®, P2 sensor equipped with two piezo sensors, Software" Powergraph 3.3 X ® ", Computer with Windows XP and higher, equipped with a USB port, Band-aid, Electrode gel, a set of textile fasteners, Case for Bio-rod, Stationery clips.

Progress of work:

1. Turn on the computer.
2. Connect the wireless signal receiver to the USB port.
3. The sensor P2(purple PP) is connected to the body of the Bio-rod.
4. If the connection is made correctly, the blue light on the amplifier housing will start to pulse intermittently.
5. The subject is put on ammunition, the Bio-rod is placed in the cover with the connector up. The cover is securely fastened with a Velcro tape on the body of the subject (see Figures 1.3 and 9-11 of the introductory section).

INSERT A DRAWING

6. The subject is seated on a chair, hands along the torso, palms turned outwards. The subject is asked to relax.
7. On the base of the last phalanx, on the pad of the ring finger of the left hand, a strip of adhesive plaster is not tightly attached to the red sensor.
8. * Feel the pulsation of the brachial artery at the level of the elbow joint. The course of the artery can be marked on the skin with a marker.

At the point of pulsation, two strips of adhesive plaster crosswise fix the sensor of black color. The wires from the sensors are fixed to the clothes with stationery clips, making sure that there is no tension when moving.

9. * Feel the pulsation of the external carotid artery, moving from the midline to the edge of the sternocleidomastoid muscle at the level of the cricoid cartilage. The course of the artery can also be marked on the skin with a marker.
10. The experimenter starts the program "Powergraph Pro", in the menu "Selection 86 - 86 - Physiology of the cardiovascular system of the ADC" selects the item "Viorecorder".
11. After starting the program, in the "File" menu, select: "Load settings" - "Sphygmography", which will result in a field for two-channel recording.

12. The experiment consists of three parts, each beginning when you click on the "Start" button and ending manually.

13. Press the "Start" button, and perform the registration at rest. The appearance of the curve should correspond to Fig. 23.

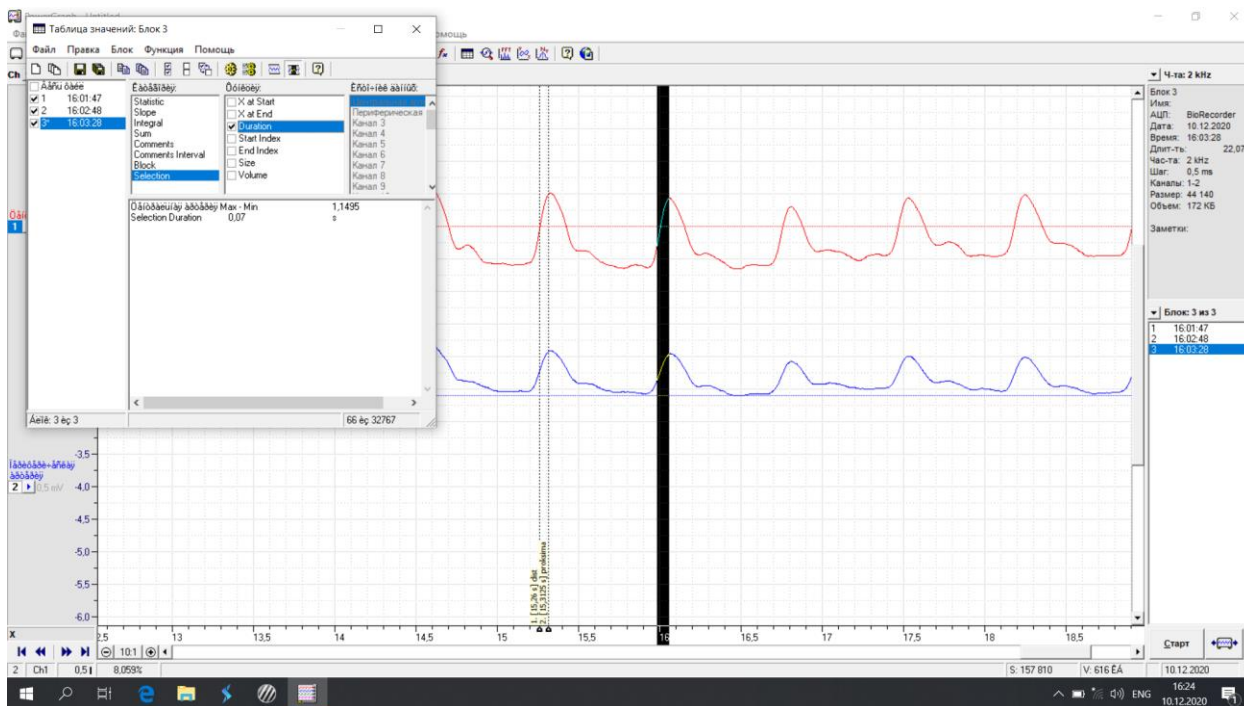


Fig. 23 Appearance of the pulse wave curve

14. The subject is asked to do 20 squats. Again, press the "Start" button, and register after physical activity.

15. Save the data (menu "File"- "Save") and start analyzing the received data. 16. Compare the appearance of sphygmograms of the artery of the finger, brachial and carotid arteries. Similarities and differences are recorded in the protocol.

17. Measure the amplitude of the teeth of the sphygmogram. To do this, select one prong, select the "Table of values" item in the "Analysis" menu. In the "Statistics" category, select the "Max-min" function, and then select "Calculate" in the "Function" submenu. In each fragment of the record, at least three teeth are measured, calculating the arithmetic mean, after which the results are entered in Table 2.

18. Measure the duration of one cycle of the sphygmogram: for this purpose, select the section of the curve between the maximum values of the teeth, and then select the "Table of values" in the "Analysis" menu. In the "Category" section, select "Selection", and in the "Function" section, select "Duration". After calculating the duration of one cycle (Menu-Function Calculate), repeat the measurements for the same interval 3 times, calculating the average value.

The value of the maximum teeth of the sphygmogram under different conditions.

Name of the record fragment	Tooth value (mV)	Average duration of one cycle	Name of the artery
At rest			
After physical activity			

19. Determine the heart rate for each fragment. Knowing the duration of one cycle, one cycle, calculate the heart rate according to the formula: heart rate= 60/duration of the average cycle (s). For example, the average cycle = 0.81 s, heart rate=60/0, 81=74 beats. min. 24. 20.

Formulate conclusions.

Questions for the Colloquium:

1. The main functions of the blood. The amount and composition of blood. Physical and chemical properties of blood. Colloid-osmotic (oncotic) pressure.
2. Buffer properties of blood.
3. Plasma and blood serum. Plasma proteins and lipoproteins.
4. The formed elements of blood. Structure and functions of red blood cells.
5. Platelets, structure and function.
6. White blood cells and their classification. White blood cell formula.
7. Hematopoiesis and its regulation.
8. Blood groups. The RH factor. Agglutinate of red blood cells.
9. Homeostasis and blood clotting.
10. Vascular-platelet link of hemostasis.
11. Coagulation hemostasis.
12. Fibrinolysis.
13. Neurohumoral regulation of the liquid state of the blood.
14. The structure of the heart of warm-blooded people. Cardiac cycle. Automation. The conducting system of the heart. Atrio-ventricular delay and its functional meaning.
15. Electrocardiogram, registration method and informative value.
16. Systolic and minute volume of the heart.
17. Mechanisms of regulation of cardiac activity: myogenic mechanisms (Frank-Starling and Anrep patterns); local intracardiac reflexes.
18. Central (extracardial) mechanisms of nervous regulation.
19. The effects of sympathetic and parasympathetic nerves on the heart.
20. The main reflexogenic zones involved in the reflex regulation of the heart.
21. Basic principles of hemodynamics and factors that determine the value of blood pressure.
22. General characteristics of changes in pressure and linear velocity of blood flow in different parts of the bloodstream.
23. Arterioles, their structure and role in the regulation of blood flow.
24. Venous blood flow and its features. Innervation of blood vessels.
25. Bulbar vasomotor center.

26. The main reflexogenic zones of the vascular bed.

27. Humoral mechanisms of blood flow regulation.

Literature:

1. Human physiology / Under the editorship of V. M. Pokrovsky, Federal state-funded healthcare. 1998. - pp. 326-344, 347-349.
2. Human physiology / Under the editorship of V. M. Smirnov. 2001. - pp. 271-288, 331-333 (for ped. faculty), 335-337.
3. Compendium on normal physiology for students of the Medical and Psychological Faculty: Textbook / V. V. Zinchuk, O. A. Balbatun, Yu. M. Emelianchik. Ed. Zinchuk V. V.-Grodno: GrSMU, 2004. (see the corresponding section).
4. Fundamentals of human physiology / Edited by B. I. Tkachenko, St. Petersburg, 1994, Vol. 1, pp. 246-262.
5. Korobkov A.V., Chesnokova S. A. Atlas of normal physiology / Edited by N. A. Aghajanyan. M., 1987. - pp. 38-45, 52-57.
6. Human Physiology / Under the editorship of G. I. Kositsky. 1985. - p. 239-257
7. Physiology of the fetus and children / Ed. by V. D. Glebovsky. 1988. - pp. 27-31, 33-40, 41-50. (for ped. faculty members)
8. Lectures on the topic of the lesson.

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Marina Vianorovna **Zolotova**

PHYSIOLOGY OF BLOOD AND CARDIOVASCULAR SYSTEM

Educational and methodical manual

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