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Mitrukhina O.B.
Silantyeva D.I.
Shaidullov I.F.,
Ananev A.S.
Svitko S.O.
Sitdikova G.F.

Lab manual of

visceral physiology

Kazan Federal
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Lab manual of
VISCERAL PHYSIOLOGY

Practical textbook

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Практикум по ВИСЦЕРАЛЬНОЙ ФИЗИОЛОГИИ

Учебно-методическое пособие
на английском языке

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Авторы

Митрухина О.Б., Силантьева Д.И.,
Шайдуллов И.Ф., Ананьев А.С., Свитко С.О.,
Ситдикова Г.Ф.

Рецензенты

кандидат биологических наук,
доцент кафедры физиологии человека и животных
Казанского федерального университета
Хаертдинов Наиль Назимович

кандидат медицинских наук,
доцент кафедры нормальной физиологии
Казанского государственного медицинского университета
Телина Эвелина Николаевна

L11 **Lab manual of Visceral Physiology**: учебно-методическое пособие на английском языке / Митрухина О.Б., Силантьева Д.И., Шайдуллов И.Ф., Ананьев А.С., Свитко С.О., Ситдикова Г.Ф. – Казань: КФУ, 2025. –82 с.: ил.

Настоящее учебно-методическое пособие представляет собой комплексное руководство по курсу “Нормальная физиология” для студентов медицинского профиля, обучающихся на английском языке. Пособие разработано для организации самостоятельной работы обучающихся и полностью соответствует требованиям образовательной программы по нормальной физиологии.

This practical book is a comprehensive guide to the course "Normal Physiology" for medical students studying in English. The manual is designed to organize students' independent work and fully complies with the requirements of the educational program for Normal Physiology.

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Lab 1. PHYSIOLOGY OF BLOOD

Blood is a functional system that delivers oxygen and nutrients to tissue cells and removes metabolic waste products from organs and interstitial spaces. Blood is a connective tissue composed of a liquid extracellular matrix called blood plasma that dissolves and suspends various cells and cell fragments. Blood performs numerous functions, including the transportation of respiratory gases, nutritive molecules, metabolic wastes, and hormones. Blood is circulated throughout the body in a system of vessels coming from and returning to the heart.

Blood has three **general functions**:

Transportation. The substances essential for cellular metabolism are transported by the blood. These substances can be categorized as follows: respiratory, nutritive and excretory. Blood transports oxygen from the lungs to the cells of the body and carbon dioxide from the body cells to the lungs for exhalation. It carries nutrients from the gastrointestinal tract to body cells and hormones from endocrine glands to other body cells. Blood also transports heat and waste products to various organs to remove them from the body.

Regulation. Circulating blood helps to maintain homeostasis of all body fluids. Blood helps regulate pH through the use of buffers. The circulation of the blood contributes to both hormonal and temperature regulation. The blood carries hormones from their site of origin to distant target tissues where they perform a variety of regulatory functions. Temperature regulation is aided by the diversion of blood from deeper vessels to more superficial cutaneous vessels or vice versa. When the body temperature is high, diversion of blood from deep to superficial vessels helps to cool the body, and when the body temperature is low, the diversion of blood from superficial to deeper vessels helps to keep the body warm.

The blood **protects** against blood loss from injury and against pathogens, including foreign microbes and toxins introduced into the body. The clotting mechanism protects against blood loss when vessels are damaged. The immune

function of the blood is performed by the leukocytes (white blood cells) that protect against many disease-causing agents (pathogens).

Blood consists of formed elements that are suspended and carried in fluid called plasma. The ratio between plasma and formed elements is called the **hematocrit** and this number is relatively constant. In humans, the percentage of plasma volume is 55–60% and volume of cells is 40–45% of the total blood volume. The hematocrit, or packed cell volume, essentially represents the percentage of erythrocytes in the total blood volume and characterizes the degree of water content in the blood.

Plasma, which is a fluid, consists of 90% water. Many inorganic and organic substances are dissolved in the plasma. **Inorganic constituents** account for about 1% of plasma weight. The most abundant electrolytes in the plasma are: Na^+ (142 mol/l) and Cl^- (103 mol/l). Smaller amounts of HCO_3^- (24 mmol/l), K^+ (4.4 mol/l), Ca^{2+} (2.5 mol/l) and other ions are present. The most notable functions of these ions are their roles in membrane excitability, osmotic distribution of fluid between the extracellular fluid and the cells, and buffering of pH changes. The most plentiful organic constituents by weight are the **plasma proteins**, which make up 6% to 8% of plasma weight. The total protein content in plasma is 6.6–8.2% of the plasma volume (in adults 200–300 g), the main of them are **albumin** (4–4.5%), **globulins** (2.8–3.1%) and **fibrinogen** (0.1–0.4%). Plasma also contains components which concentration varies, such as enzymes (lipase and amylase), vitamins, hormones, soluble products of hydrolysis of nutrients in the gastrointestinal tract and waste products for excretion.

The formed elements of the blood include three principal components: red blood cells (RBCs), white blood cells (WBCs), and platelets. All formed elements are formed in the bone marrow from a single pluripotent stem cell.

Red blood cells, or **erythrocytes** are the most abundant cells of the blood. A healthy adult male has about 4.5–5 million RBCs per 1 µl (microliter) of blood, and a healthy adult female has about 3.7–4.7 million RBCs per 1 µl. During pregnancy, the number of RBCs may decrease (many researchers consider that this is the norm). In a person with a body weight of 60 kg, the total number of RBCs is 25 trillion. There is a slight fluctuation of the number of RBCs. In the

case of various diseases, the number of RBCs may decrease. This condition is called **erythropenia** and is often associated with **anemia**. An increase in the number of RBCs is referred to as **erythrocytosis**.

Mature red blood cells have a simple structure. Their plasma membrane is both strong and flexible, which allows them to deform without rupturing as they squeeze through narrow capillaries. The main function of RBCs is a transportation of O_2 and CO_2, amino acids, polypeptides, proteins, carbohydrates, enzymes, hormones, fats, cholesterol, various biologically active compounds (prostaglandins, leukotrienes, etc.), microelements, and others. The protective function of RBCs is that they play a significant role in the specific and non-specific immunity and take part in hemostasis, blood coagulation and fibrinolysis.

Leukocytes or **white blood cells** (WBCs) have nuclei and do not contain hemoglobin. They are almost invisible under the microscope unless they are stained; therefore, they are classified according to their staining properties. WBCs are divided into two large groups: granular (granulocytes) and agranular (agranulocytes). The stain used to identify white blood cells is usually a mixture of a pink-red stain called eosin and a blue-purple stain (methylene blue), which is called a "basic stain." **Granular leukocytes** with pink-staining granules are therefore called **eosinophils**, and those leukocytes with blue-staining granules are called **basophils**. Those leukocytes with granules that have little affinity for either stain are **neutrophils**. **Agranulocytes** include **lymphocytes** and **monocytes** (Fig. 1.4). The number of the WBCs normally ranges from 4000 to 9000 in 1 mm^3 in adults. An increase in the number of leukocytes is called **leukocytosis**, a decrease in the number of leukocytes is called **leukopenia**.

The **thrombocytes** or **platelets** are formed from giant cells of the red bone marrow (**megakaryocytes**). Platelets have a round or slightly oval shape and their diameter do not exceed 2–3 microns. Platelets do not have nucleus, but have a large number of granules (up to 200) of various structures. Platelets play an important role in blood clotting. They constitute most of the mass of the clot, and phospholipids in their cell membranes activate the clotting factors in plasma that results in threads of **fibrin**, which reinforce the platelet plug. The number of platelets in a healthy person normally is 180–320 thousand in 1 µl. An increase

in number of platelets is called **thrombocytosis**, a decrease is called **thrombocytopenia**.

1.1. Red blood cell count (RBC count)

The RBC count is the number of red blood cells that are present in a sample of blood. The number of RBCs is very high and their counts are performed by determining the number of cells in the sample, which must be diluted 200 times with appropriate diluting fluid. Then obtained number is converted to the results: the number of cells per 1 liter of the whole blood.

Requirements

A counting chamber of Goryaev (improved Neubauer's chamber; Fig. 1.1), graduated pipettes (1 ml and 0.02 ml), a solution of 3% NaCl, a cotton wool, two tubes, conserved blood, a microscope.

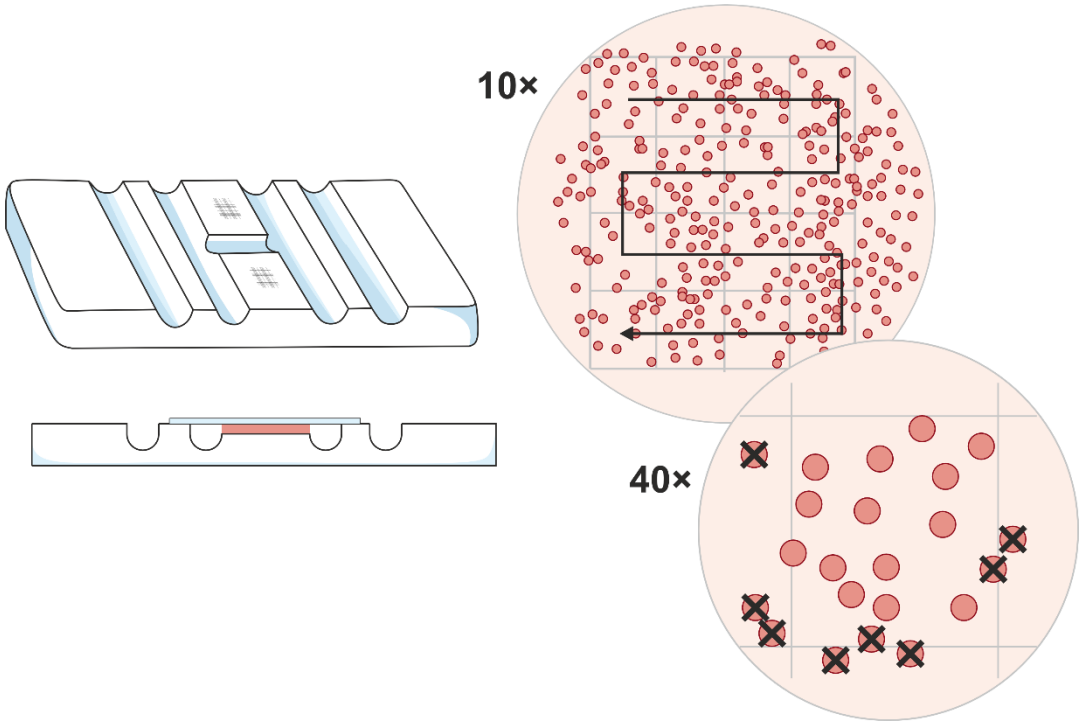

Figure 1.1. Counting chamber of Goryaev: 1 big square contains 16 small squares and example of counting the RBC in small square

A counting chamber consists of a thick glass slide divided into two central platforms (Fig. 1.1). The central platform is slightly lower than the sides thus the depth under the coverslip and the central platform is 0.1 mm (Fig. 1.1). When the

chamber is filled with diluted blood, a thin film of a fluid of a certain volume is spread on the central platform. This volume is used to count the number of cells.

Procedure

Pour 4 ml of 3% NaCl solution into a test tube. Add 0.02 ml of blood into a test tube with a pipette. Stir the solution thoroughly and leave it for 5–10 minutes. Cover the central part of counting chamber with a coverslip. Then place the tip of the pipette with RBC sample on the edge of coverslip and allow the solution to flow under the coverslip by capillary force. Allow the cells to settle for one minute before starting the cell count. Focus on the lines of the chamber and RBC under the low power of the microscope to get a general impression of the distribution of the RBC. The cells are to be counted in 5 large squares, which are located diagonally, each with 16 smallest squares (in total, there are 80 small squares). Count the RBC in each small square and cells lying on its left and upper lines; ignore those cells lying on its right and lower lines (Fig. 1.1). The chamber consists of 225 large squares, 25 of them are divided into 16 small squares. The side of the smallest square is known, it is 1/20 mm. The square of the smallest square is 1/400 mm^2 (1/20 × 1/20). The volume of the smallest square is 1/4000 mm^3 (side = 1/20 mm, depth = 1/10 mm, volume = 1/20 × 1/20 × 1/10 = 1/4000 mm^3). Use the formula bellow to calculate the number of RBC

$$X = \frac{a \times 4000 \times b}{c},$$

where **X** – the number of RBC in 1 mm^3 of blood, **a** – the total number of RBC in the 80 smallest squares; b – the coefficient of the blood dilution with 3% NaCl (which is 200); **c** – the number of the smallest squares (80).

Calculate the number of RBC per 1 liter of undiluted blood. Express your results in: 1 mm^3 (for example: 4.8×10^6 RBC/mm^3) and in 1 liter (for example: 4.8×10^{12} RBC/l). Compare your results to normal values. If they are below or above the average, assume the reasons for this deviation.

Normal range of RBC counts: males $4.5–5.5\times10^{12}$ RBC/l. females $4–5\times10^{12}$ RBC/l. In newborn children, the number of RBC may be higher than normal: $6–7.5\times10^{12}$ RBC /mm^3 or $6–7.5\times10^{12}$ RBC/l; 2–3 % of these RBC are the reticulocytes.

The value of the RBC count may vary during the 24 hours by 5 %. The count is lowest during the sleep, and then it gradually rises and becomes maximal in the evening. Muscular exercise raises the count temporarily. At higher altitude, the number of RBC raises, whereas at lower altitude it falls. High external temperature increases the number of RBC. Any condition, which lowers the oxygen tension of arterial blood, increases the RBC count. Emotional excitement also increases the number of RBC.

1.2. Sahli's method for the estimation of hemoglobin

Hemoglobin is a conjugated protein present in red blood cells. The method to estimate hemoglobin concentration is based on the fact that hemoglobin is converted to acid **hematin** by the action of HCl. The solution of acid hematin is further diluted until its color matches exactly with that of the permanent standard of the comparator block of Sahli's haemoglobinometer. The concentration of Hb can be read directly from the calibration tube with a sample of diluted solution.

Requirements

Sahli's hemoglobinometer, 0.1 mol/l of HCL solution, distilled water, 0.02 ml of blood. Sahli's haemoglobinometer contains two comparator blocks on the sides (Fig. 1.2) and hemoglobin tube (Fig. 1.2A) graduated in gram percent of Hb (2–24) in the middle, glass pipette and a glass stirrer (Fig. 1.2).

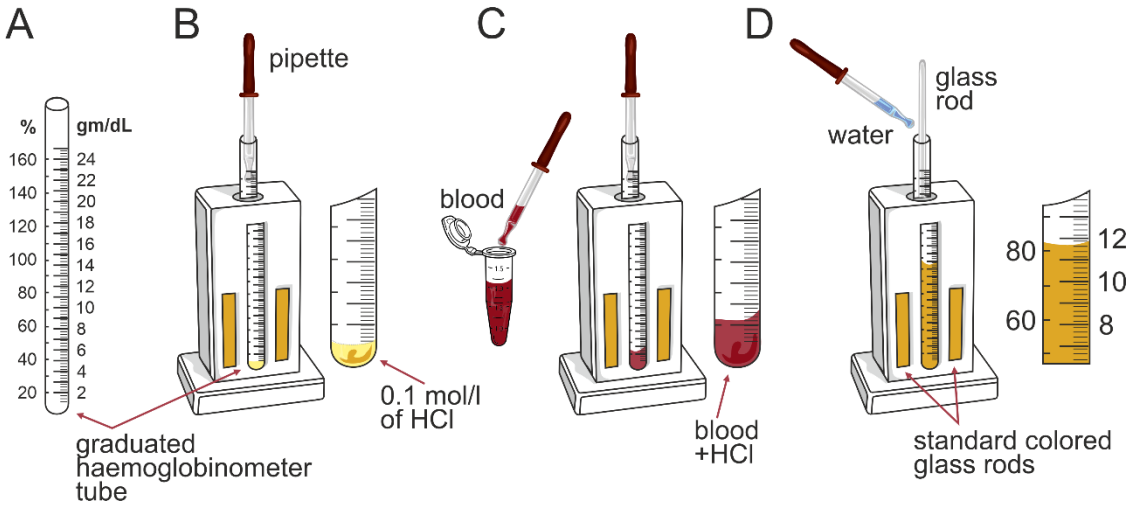

Figure 1.2. Using Sahli's Hemoglobinometer: a step-by-step guide

Procedure

Fill a clean and dry tube with 0.1 mol/l of HCl solution up to the lower ring mark and set it in the hemoglobinometer (Fig. 1.2B). Take 0.02 ml of blood sample with a pipette and immediately transfer it to the hemoglobinometer tube containing HCl (Fig. 1.2C). Gently mix the content of the tube by shaking. Put the tube back in the comparator and wait for about 10 minutes. During this time, all red cells will be destroyed and the hemoglobin will be released into the acid solution. The hemoglobin converts into hematin, which has a brown color. After 10 minutes, dilute the acid hematin by adding distilled water drop by drop (Fig. 1.2D). After addition of each drop of distilled water, the solution should be stirred with a glass stirrer and the color of the solution should be compared with the color in the comparator blocks on the side of the hemoglobinometer. If the color of the test solution is darker, then continue dilution till it matches with the standard. As soon as it happens, note the reading of the lower meniscus of the solution. Write down hemoglobin concentration in g per 100 ml of blood by multiplying the obtained reading by 10. The average concentrations of hemoglobin are presented in the table 1.1. Compare your results to normal values. If they are below or above the average, assume the reasons for this deviation.

Table 1.1. The average concentrations of hemoglobin

Sex	Average	Normal range
Males	140 g/l	135–150 g/l
Females	130 g/l	125–140g/l

1.3. Estimation of color index

The **color index** (CI) is a useful clinical test, which helps to determine the types of **anemia**. CI indicates the ratio of the amount of hemoglobin to the number of red blood cells in a given volume of blood.

Procedure

Use your results from lab works 1.1 and 1.2 to calculate the concentration of hemoglobin in 1 liter of blood (g/l). Calculate CI using formula:

$$\mathrm{CI} = \frac{\mathrm{Hb}}{\mathrm{RBC}} \times 3,$$

where **Hb** is the concentration of hemoglobin in gram per liter, **RBC** is the first 3 digits of the quantity of erythrocytes in 1 mm^3 of blood. For example, the concentration of Hb is 140 g/l and quantity of erythrocytes in 1 mm^3 is 4.8×10^6 mm^3, the calculation will look like:

$$\mathrm{CI} = \frac{140}{480} \times 3 = 0.87$$

CI indicates the proportion of hemoglobin present in each red cell relative to normal. The value of normal CI is 1, but usually CI is slightly lower, i.e. 0.85. The low CI indicates **hypochromic anemia**. The value of CI above 1 indicates **hyperchromic or macrocytic anemia**. Compare your results to normal values. If they are below or above the average, assume the reasons for this deviation.

1.4. White Blood Cells count

The number and types of WBC is represented in a percentage. For differentiation of WBC type the blood smears are stained according to Romanovsky staining procedure. After staining, the blood smear is examined under an oil-immersion objective of microscope, which allows to identify the different types of WBC. The experimenter should count a minimum of 100 different WBC.

Requirements

Prepared stained blood smears, a microscope, an immersion oil.

Procedure

Examine the smear under a low power objective (40×) for scanning the cells distribution. Put a drop of the immersion oil onto the smear. Bring the oil-immersion objective into position until it touches the drop of oil above the smear. Adjust the objective and focus on the cells. Start count from one edge of the smear and move the slide to another edge then return to the first edge. Continue counting until 100 cells will be found. Fixate your counts. For differentiation of the types of WBC, use Fig. 1.3.

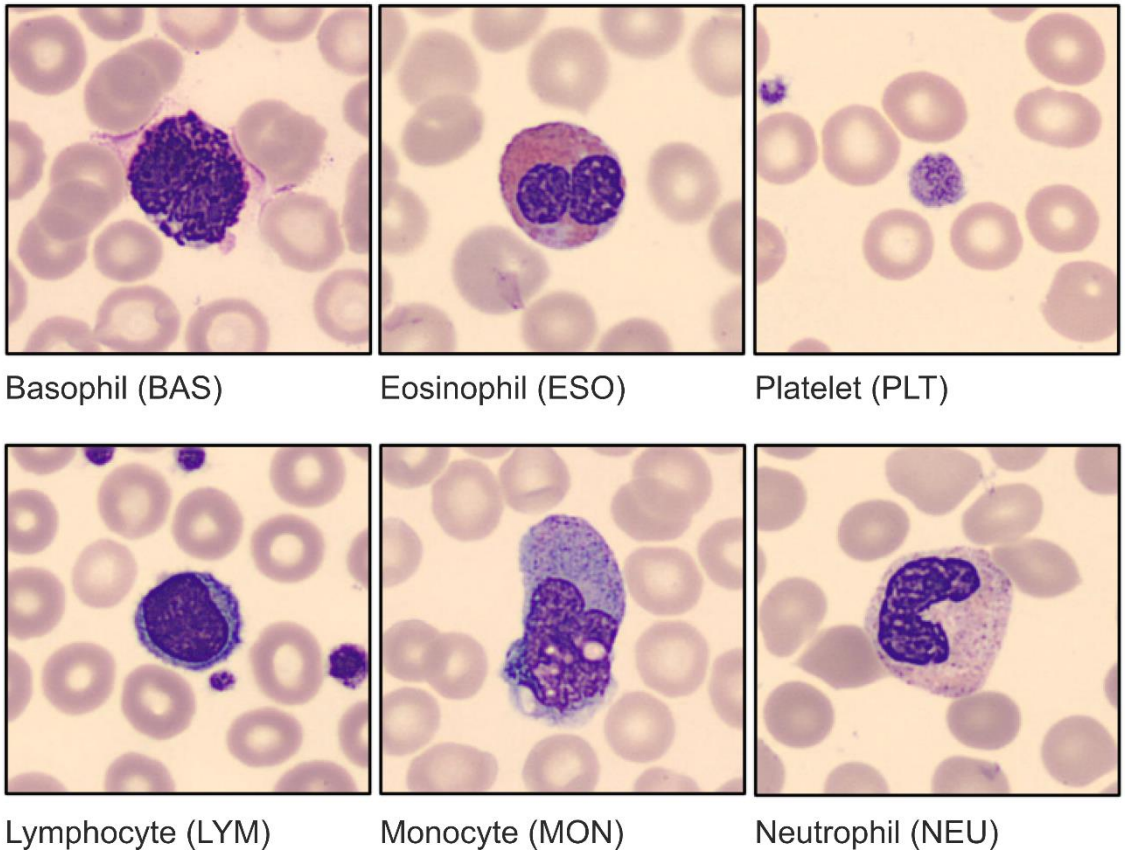

Figure 1.3. Types of white blood cells

Results and Discussion

Write down your results into table 1.2 and compare them with the normal values. If there is any deviation from normal, assume which process in the body caused these changes. Describe functions of each type of WBC.

Table 1.3. White blood cells count results

WBC type	Normal values	Counted results
Neutrophils	62%	
Eosinophils	2.3%	
Basophils	0.4%	
Lymphocytes	30 %	
Monocytes	5.3%	
Total WBC	100%	

1.5. Determination of blood groups. ABO system

The surface of erythrocytes contains genetically determined antigens. Based on the presence or absence of various antigens, blood is categorized into different blood groups. There are at least 24 blood groups and more than 100 antigens that can be detected on the surface of red blood cells. The ABO system is based on two glycolipid antigens called **A and B antigens (agglutinogens)**. People whose erythrocytes only have antigen A belong to type A blood group. Those who have only antigen B are type B. Individuals who have both A and B antigens are type AB; those who have neither antigen A nor B are type O. Blood plasma usually contains **antibodies called agglutinins**. These are the **anti-A (α) antibody** (which reacts with antigen A), and the **anti-B (β) antibody** (which reacts with antigen B).

Table 1.4. Antigens and antibodies of the ABO blood types.

Blood type	**A**	**B**	**AB**	**O**
RBC contain	A antigen	B antigen	A and B antigen	No agglutinogens
Plasma contain	Anti-B antibody	Anti-A antibody	No agglutinins	Anti-A and anti-B antibodies

Requirements

White glass slides with three round cavities, pipettes, physiological solution, standard anti-A serum, anti-B serum and anti-AB serum.

Procedure

Take a white glass slide with three round cavities. Subsequently add in cavities 0.1 ml of anti-A serum (pink), anti-B serum (blue) and anti-AB serum (yellow). Add 0.01 ml of blood to each cavity with serum using different pipettes then mix the contents of each hole using the individual glass sticks. Wait for 2 minutes and find in which cavity the agglutination occurs. Examples of different blood types sample's reaction to different serums are shown on Fig. 1.4.

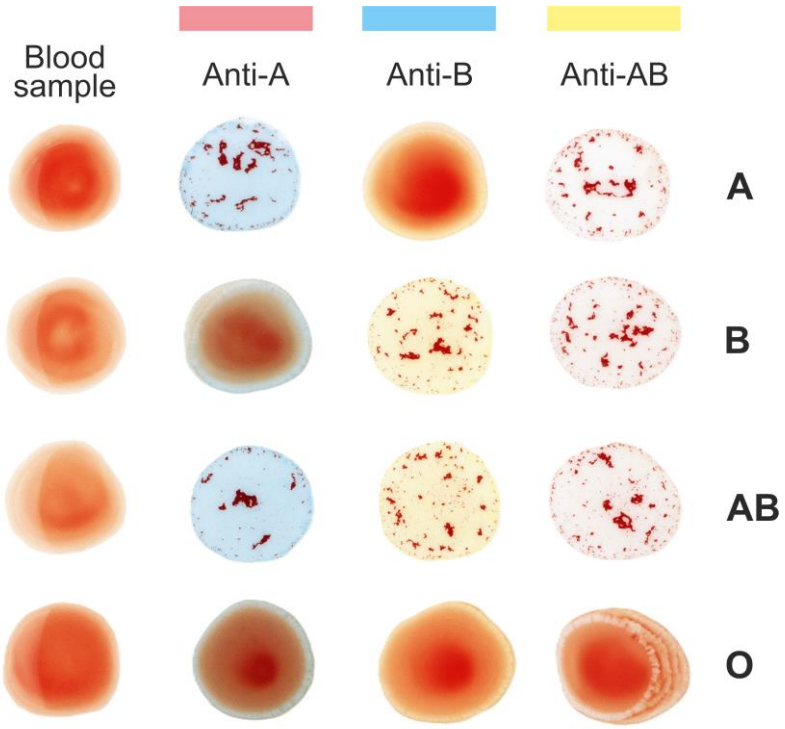

Figure 1.4. Blood typing. Agglutination (clumping) of red blood cells occurs when cells with A-type antigens are mixed with anti-A antibodies and when cells with B-type antigens are mixed with anti-B antibodies. No agglutination would occur with type O blood

Results and Discussion

Make conclusion about blood type of the sample. Explain the process of agglutination. Describe the main rules and principles of blood transfusion.

1.6. Osmotic resistance of erythrocytes

The osmotic resistance of erythrocytes can be evaluated as resistance of erythrocytes to hemolysis in hypotonic solutions. **Haemolysis** is the disruption of erythrocytes and the release of their contents (hemoglobin) into surrounding fluid. Normally, in humans, the osmotic minimum resistance of erythrocytes is 0.36%, the maximum is 0.42%.

Requirements

1% solution of NaCl, distilled water, 6 tubes, pipettes for 500 and 1000 µl (1ml) and measuring tube.

Procedure

Prepare solutions of various concentrations of NaCl in 6 test tubes:

1. 1 ml of distilled water + 9 ml of 1% NaCl solution (0.9% NaCl)
2. 2 ml of distilled water + 8 ml of 1% NaCl solution (0.8% NaCl)
3. 4 ml of distilled water + 6 ml of 1% NaCl solution (0.6% NaCl)
4. 6 ml of distilled water + 4 ml of 1% NaCl solution (0.4% NaCl)
5. 8 ml of distilled water + 2 ml of 1% NaCl solution (0.2% NaCl)
6. 10 ml of 1% NaCl solution.

Dilute 500 µl of blood in 2.5 ml of 1% NaCl solution. Then add 500 µl of acquired solution into each test tube, gently mix and leave in a rack for 1 hour. After 1 hour, analyze in which tubes the hemolysis occurred (the solution in the tube transfers to transparent fluid with light red color), i.e. determine the osmotic stability of erythrocytes in the blood.

Lab 2. PHYSIOLOGY OF CARDIOVASCULAR SYSTEM

2.1. Blood pressure by Korotkoff's auscultatory method

One of the most important indicators of the state of the cardiovascular system (CVS) is **blood pressure (BP)**. The blood pressure is determined by two factors: BP in aorta and the resistance of the vascular walls. The **systolic pressure** is maximal BP in the artery following ventricular systole and usually it is equal to 110–140 mm Hg in a healthy person. The **diastolic blood pressure (DP)** is the level to which the arterial BP falls during ventricular diastole, normal range is 65–80 mm Hg.

The **auscultatory or Korotkoff method** of measuring BP is the traditional noninvasive approach for measuring systolic and diastolic BP. The tonometer is the device consisting of the rubber cuff and manometer. It is used in clinics to conduct auscultatory measurements. The inflation of the cuff wrapped around the patient's arm causes the occlusion of brachial artery and its temporal collapse. As the cuff is gradually deflated, blood flow is restored and this process is accompanied by sounds that can be heard with a stethoscope held over the brachial artery. This method was proposed by the Russian scientist Nikolai Korotkov in 1905.

Requirements

A tonometer and stethoscope.

Procedure

Wrap the cuff around the subject's shoulder. Install the stethoscope strictly in the elbow bend above the brachial artery (Fig. 2.1). By injecting air into the cuff, create a pressure in it until maximum, then, slightly release the air by opening the tap until rhythmic sounds (Korotkoff's tones) appear. The manometer readings at the moment of appearance of first sound corresponds to the **systolic blood pressure (SP)**. Continue to decrease the pressure in the cuff until the sound disappears. The pressure in the manometer at the moment of last sound is equal to the **DP**.

Blood pressure should be measured while sitting in a calm environment, avoiding physical activity or strong emotions prior to measurement. When

measuring blood pressure, sit with your feet flat on the floor and do not cross or touch your legs. To get the most accurate reading, position the arm with the cuff at heart level, comfortably on a flat surface with no tension. Apply the cuff 2–3 cm from the elbow bend so that two fingers can easily fit under it. If your hand is on your knee or hanging down, the tonometer may inflate to an incorrect value. When the arm is below heart level, blood has difficulty returning to the heart. To improve blood return, compress the blood vessels in the arm by positioning it above heart level. **Do not talk during measurement.**

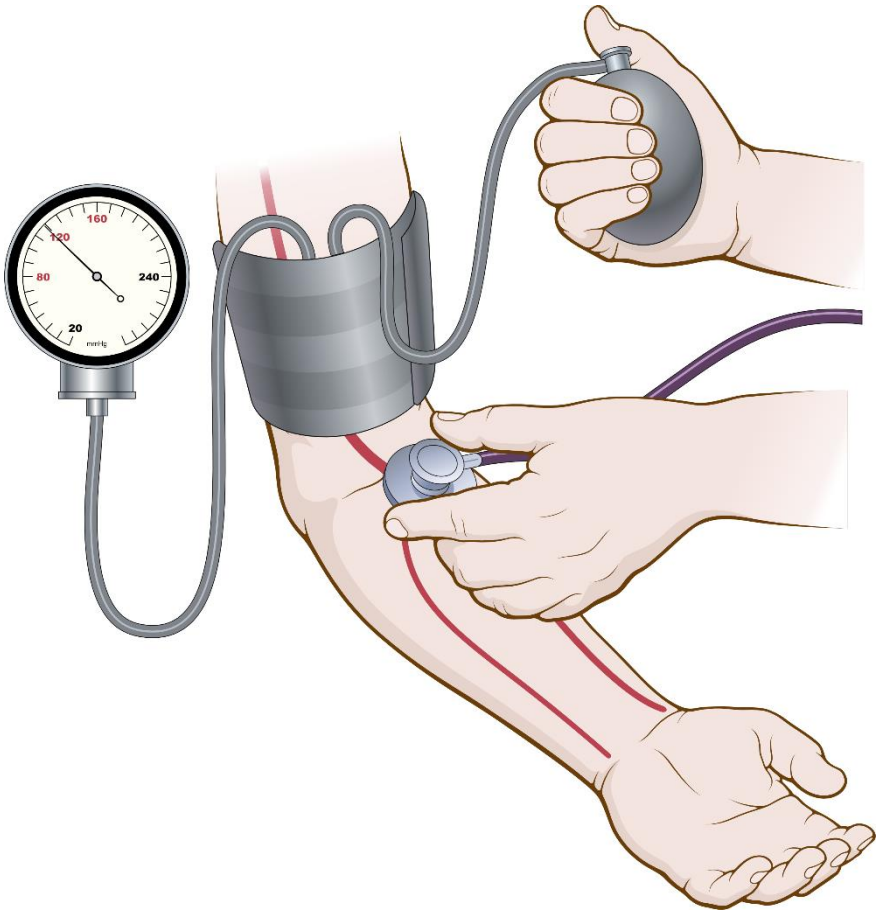

Figure 2.1. Auscultatory method for measuring systolic and diastolic arterial pressures

2.2. Determination of the functional state of cardiovascular system

During physical activity, the demand of the muscles and the human brain for nutrients and oxygen increases. A necessary condition for this is an increase in **blood flow velocity** and **minute blood volume (MBV)**. Minute blood volume depends on indicators such as **heart rate (HR)** and **stroke volume (SV)**, which in turn depends on BP. An increase in systolic BP during physical training is

an indicator of increased cardiac activity. If during physical training diastolic BP decreases, this indicates the expansion of blood vessels and a decrease in the resistance of the vascular walls to the blood movement. If the diastolic BP increases, which is associated with a neuro-emotional influence, it means that the vascular tone has greater tension. **Pulse pressure (PP)** is the difference between the maximum and minimum BP.

$$\mathrm{PP} = \mathrm{SP} - \mathrm{DP}$$

Physical exercises increase PP. An increase in pulse pressure usually occurs with an increase in systolic pressure and depends on the severity of load. There are several functional tests for evaluation of endurance of CVS.

2.2.1. Ruffier-Dickson test

The Ruffier-Dickson test is a simple cardiovascular endurance test, which involves measuring heart rate before and after physical load. The test was originally developed in the 1950s by J. E. Ruffier and modified by J. Dickson, who developed the **Ruffier-Dickson index (RDI).**

Requirements

Stopwatch

Procedure

1. Measure the pulse for 15 seconds ($\mathbf{P_1}$) at rest in a standing position.
2. Make 30 squats per 45 seconds.
3. Measure pulse of the first 15 seconds ($\mathbf{P_2}$) and the last 15 seconds ($\mathbf{P_3}$) of the first minute after load in a standing position.
4. Calculate the index of cardiac activity or RDI by the formula:

$$\mathrm{RDI} = \frac{4 \times (\mathrm{P_1} + \mathrm{P_2} + \mathrm{P_3}) - 200}{10}$$

5. Evaluate your results using table 2.1.

Table 2.1. RDI evaluation

Less than 0	Excellent
From 0 to 3	Very good
From 3 to 6	Reasonably good
From 6 to 9	Average
From 9 to 12	Moderate
From 12 to 15	Poor
From 15 and more	Very poor

2.2.2. Martinet-Kushelevsky test

Requirements

Sphygmomanometer, stopwatch.

Procedure

1. Measure heart rate (HR), systolic (SP) and diastolic (DP) pressure at rest in a sitting position.
2. Offer first load to the subject: 20 squats in 30 seconds. Measure HR and BP in 3 min after the load. After this load, the recovery period lasts on average 3 min.
3. Offer the second load to the subject: 80 squats for 2 minutes. Measure heart rate and BP in 5 min after the load. After this load, the recovery period lasts on average 5 min.

Evaluation is carried out over a three-point system of the largest difference between the measured parameters before and after the load. Compare columns 2 to 3, and 2 to 4 (Table 2.2). If the difference is not more than 5 then the result is considered **good**; from 5 to 10 is **satisfactory**; more than 10 is **unsatisfactory**. All calculations must be shown in your report.

Table 2.2. Martinet-Kushelevsky test results

Parameters	**At rest**	**Recovery after the first load (3min)**	**Recovery after the second load (5min)**
HR			
SP			
DP			

2.2.3. The influence of the physical load on the blood pressure

There are following types of response of the cardiovascular system to the physical exercises:

Normotensive type of reaction is characterized by the fact that along with increased heart rate, pulse pressure increases due to the pronounced increase of the maximum (systolic) pressure and a moderate reduction of the minimum (diastolic) pressure. The recovery period is short.

Hypertensive reaction is characterized by dramatic increase of the maximum pressure (up to 200 mmHg and above) and significant increase of the heart rate

Hypotonic type of reaction is described as a low rise of the maximum pressure and significant increase in pulse rate, recovery period is prolonged. Minimum pressure usually rises, whereby the pulse pressure can also be reduced.

Dystonic type of reaction in which the phenomenon of "Infinite tone" (non-vanishing sound pulsation) in determining minimum pressure can be observed. Maximum pressure often increases, causing a strong increase in pulse amplitude. Recovery is slowed down.

“Stair Step” type reaction is characterized by the fact that directly after the load maximum pressure is less than on the 2nd and even 3rd minute of the recovery period. Often there was a significant minimal pressure drop and heart rate increasing.

Requirements

Manometer, stethoscope.

Procedure

1. Measure the BP, HR (heart rate) and RR (respiratory rate) at res in a sitting position.
2. The subject has to do 20 deep squats. Measure the BP, HR and RR immediately after the load in a sitting position.
3. Measure the same parameters every minute during 3 minutes of recovery period after the load.
4. Write down the data to table 2.3.

Table 2.3. CVS parameters at rest and after physical load

The registration period		Systolic Pressure	Diastolic Pressure	Pulse Pressure	Heart Rate per 1 min	Respiratory rate per 1 min
Before the load (1)						
During the load (2) (immediately after the load)						
Recovery period	1 min (3)					
	2 min (4)					
	3 min (5)					

Results and Discussion

Make a conclusion about the type of cardiovascular response to the load by calculating following parameters and indicators.

1. Parameters of CVS at rest (Use control measurements (1) before the load):

Vegetative index (Kerdo's vegetative index in %)

$$\text{VIK} = 100 \times \left(1 - \frac{\text{DP}}{\text{HR}}\right),$$

where **DP** is diastolic pressure; **HR** is heart rate, measured for 1 min. If the result is 10% the subject's sympathetic and parasympathetic tones are balanced. If VIK is more than 10% than the sympathetic tone is dominant, if less 10% the parasympathetic tone prevails.

Pulse pressure (PP)

Pulse pressure reflects the difference between systolic and diastolic arterial pressure.

$$\text{PP} = \text{SP} - \text{DP},$$

where **SP** is systolic pressure, **DP** is diastolic pressure. The value of this parameter depends on arterial wall elasticity and stroke volume and it can be an indicator of atherosclerosis and heart disease. Normal pulse pressure is 40 mmHg.

Systolic volume or stroke volume (SV) is the volume of blood ejected from the ventricles during systole or during one contraction cycle. Systolic volume can be calculated using **Starr's formula**:

$$\text{SV} = (90.97 + 0.54 \times \text{PP}) - (0.57 \times \text{DP} + 0.61 \times \text{A}),$$

where **A** is the age (the total quantity of years), **PP** is the pulse pressure; **DP** is the diastolic pressure. Normally, the systolic volume is 60–80 ml.

Minute volume (MV) of blood is the volume of blood pumped by the heart during 1 minute. It can be calculated by the following formula

$$\mathrm{MV} = \mathrm{SV} \times \mathrm{HR}$$

where **HR** is the heart rate per minute, **SV** is systolic volume. Normally, this value is 3500–5000 ml/min for men and 3000–4000 ml/min for women.

Average dynamic pressure (ADP) is the resulting variable of all values of the pressures measured during one cardiac cycle. It can be estimated according to the **Hickem formula**:

$$\mathrm{ADP} = \frac{\mathrm{PP}}{\mathrm{A}} + \mathrm{DP}$$

where **PP** is the pulse pressure; the **DP** is the diastolic pressure and **A** is the age (the total quantity of years). Normally, the ADP is 80 mm of Hg.

Vascular peripheral resistance (PR) or **total peripheral resistance** is the resistance of the blood vessel to the blood flow through the circulatory system. It can be calculated according to the **Poiseuille's formula**:

$$\mathrm{PR} = \frac{\mathrm{ADP} \times 60 \times 1333}{\mathrm{MV}}$$

where **ADP** is the average dynamic pressure, **MV** is minute volume of blood. For healthy person the value of peripheral resistance at rest varies between 1095 and 2500 dynes/s/cm^{-5}. **Functional hyperemia** occurs during muscle activity and leads to a reduction of total peripheral resistance, which is proportional to the power of the produced work and an increase in the minute volume of blood circulation.

2. Parameters of CVS after the load (Measurement (2) immediately after the load):

To assess the state of the cardiovascular system, depending on the physical load, compare values of the following coefficients in the resting state and after the load.

The coefficient of increase in systolic pressure (K_1)

$$K_1 = \frac{(SP_a - SP_b)}{SPb},$$

where **SP_a** is systolic pressure **after** exercise, **SP_b** is systolic pressure **before** exercise (at rest).

The coefficient of increase in the pulse rate (K_2)

$$K_2 = \frac{(HR_a - HR_b)}{HR_b},$$

where **HR_a** is the pulse rate **after** exercise, **HR_b** is the pulse rate **before exercise**. If **$K_1 > K_2$**, the regulation of cardiovascular activity is **normal**. If **$K_1 < K_2$**, there is probably a **risk of heart failure**.

Endurance factor at rest (REF)

$$REF = \frac{HR_b \times 10}{PP_b},$$

where **HR_b** is the pulse rate before exercise and **PP_b** is the pulse pressure before exercise. This factor characterizes the training level of fitness of the cardiovascular system at rest. Normal value of REF is 16.

Endurance factor after the load (LEF)

$$LEF = \frac{HR_a \times 10}{PP_a},$$

where **HR_a** is the pulse rate after physical exercises and **PP_a** is the pulse pressure after physical exercises. This factor characterizes the degree of training level of the cardiovascular system to load. The increase in LEF compared with REF serves as an indicator of an untrained cardiovascular system.

The quality of the reaction (RQ)

$$RQ = \frac{PP_a - PP_b}{HR_a - HR_b},$$

where **PP_a** is the pulse pressure after physical exercises, **PP_b** is the pulse pressure before exercise, **HR_a** is the pulse rate after physical exercises and **HR_b** is the pulse rate before physical exercises. The **quality of the reaction** is a parameter that allows to evaluate the degree of training level of the cardiovascular system;

it characterizes the recovery of blood pressure and pulse rate after the end of the exercise.

Usually the **normal value** of **RQ<1**. The increase in RQ indicates an unfavorable reaction of the cardiovascular system to the load.

There is a relationship between the parameters of cardiovascular and respiratory systems of a person measured during exercises. This relationship can be expressed in terms of the **pulse-to-breath ratio before exercise (PBR_b)** and **pulse-to-breath ratio after load (PBR_a)**

$$\mathrm{PBR_b} = \frac{\mathrm{HR_b}}{\mathrm{RR_b}},$$

$$\mathrm{PBR_a} = \frac{\mathrm{HR_a}}{\mathrm{RR_a}},$$

where **RR_b** is the respiratory rate before physical exercises and **RR_a** is the respiratory rate after exercises, **HR_a** is the pulse rate after physical exercises and **HR_b** is the pulse rate before physical exercises. The value of the PBR_b is 4–5, and it increases during physical activity. The less is the difference between PBR_a and PBR_b, the better is **cooperation between CVS and respiratory system**. A sharp increase of the PBR_a indicates the **overload** of the cardiovascular system, and its decrease indicates the **decompensation** in the respiratory system.

Results and Discussion

Write down the obtained results to the copybook, compare the results obtained at rest and under physical load, assess the functional state of the cardiovascular and respiratory system on the basis of all calculated indicators of CVS state. Define the type of response of the cardiovascular system to a physical load. Define which one from the fifth types mentioned in the beginning of the work is yours. Explain the nature of the change of indicators during exercises.

2.3. Registration and analysis of ECG at rest and after physical load

When the cardiac impulse passes through the heart, electrical current also spreads from the heart into the adjacent tissues surrounding the heart. A small portion of the current spreads all the way to the surface of the body. If electrodes

are placed on the skin on the opposite sides of the heart, electrical potentials generated by the current can be recorded; the recording is known as an electrocardiogram.

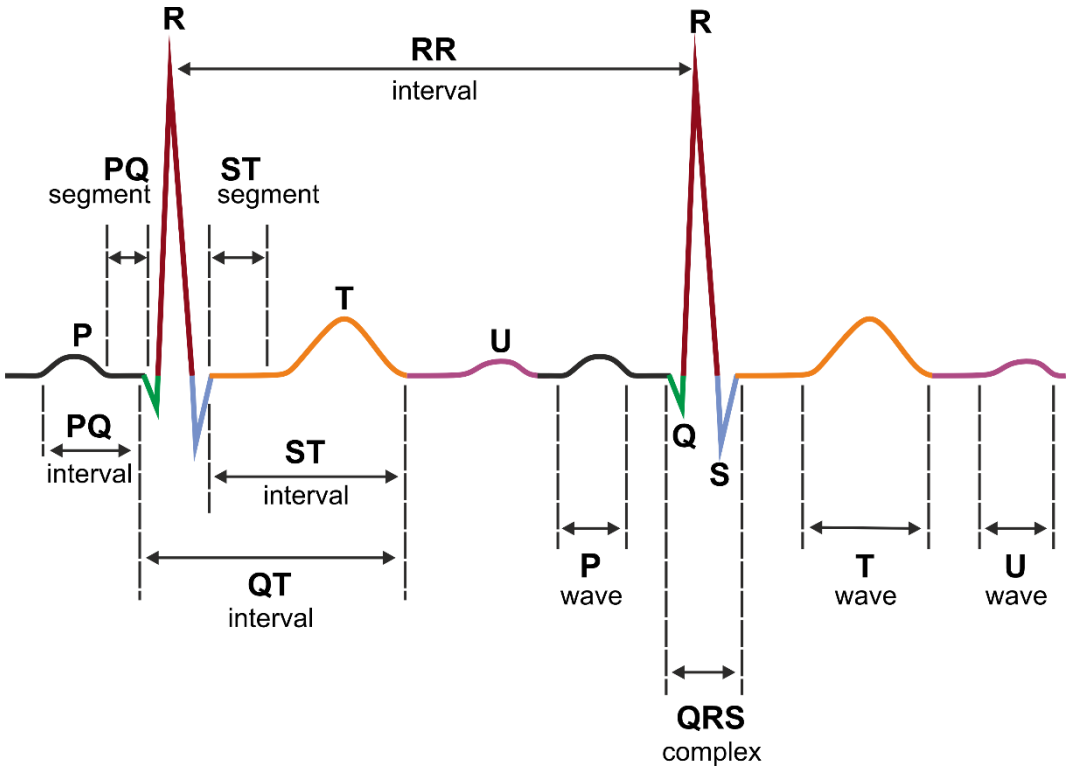

Figure 2.2. ECG of a heart in normal sinus rhythm

The normal electrocardiogram (Fig. 2.2) is composed of P wave, QRS complex, and T wave. The P wave is caused by electrical potentials generated when the atria depolarize before beginning of contraction. The QRS complex is caused by potentials generated when the ventricles depolarize before contraction, that is, as the depolarization wave spreads through the ventricles. T wave is known as a repolarization wave.

Requirements

Electrocardiograph, electrolyte gel, cotton pads, alcohol, physiological solution.

Procedure

The subject should lie down on the medical couch. Wipe the skin about 5 cm above the wrists and ankles on the internal side, using cotton pads and alcohol. Put some electrolyte gel (or napkin moisturized with physiological solution) to the place where the electrodes will be attached (See Fig. 2.3). Connect electrodes

following the color code (Table 2.4 or Fig. 2.3). Set the paper-speed selector switch to 25 mm/sec and vertical gain in 10 mm/mV.

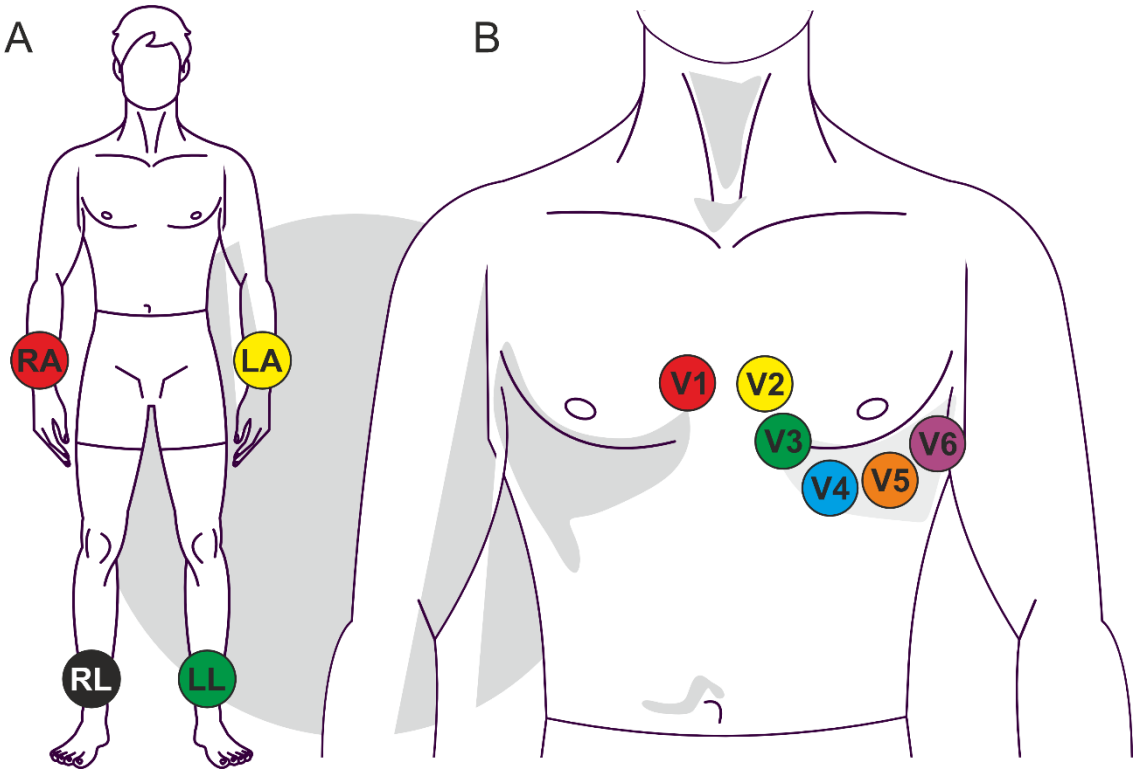

Figure 2.3. The electrocardiograph lead and their placement

Record the electrocardiogram of I, II and III standard leads at rest and after physical load (20 squats in 30 seconds).

Table 2.4. Color code for the electrodes placement.

Color	Position of electrodes
Red	Right arm
Yellow	Left arm
Black	Right leg
Green	Left leg

2.3.1. Analysis of the ECG waves

Give the **characteristics of waves** (Fig. 2.2), such as **direction** (positive, negative), **duration** (sec), and **amplitude** (mm). For example: "Waves P and T have positive direction, because the projection of vector is directed towards the side

of the positive electrode lead; waves Q, S are negative, because the projection of vector is directed towards the side of the negative electrode lead".

Measure the amplitude of the waves and put the values in a table 2.5

Table 2.5. Amplitude values of ECG waves on II standard lead.

Waves	**Normal range (mV)**	**Subject's parameters at rest**		**Subject's parameters after exercise**	
		mm	mV	mm	mV
P	**0.1–0.2**				
Q	**0.02–0.03**				
R	**0.8–1.5**				
S	**0.02–0.03**				
T	**0.05–0.3**				

Measure the durations of ECG intervals and put values in a table 2.6.

Table 2.6. Durations of ECG intervals.

Intervals	**Normal range (sec)**	**Subject's parameters at rest**		**Subject's parameters after exercise**	
		mm	sec	mm	sec
PQ	**0.12–0.2**				
QS	**0.06–0.10**				
QT	**0.35–0.46**				

Calculate **frequency of cardiac contractions (FCC)** by using following formula:

$$\mathrm{FCC} = \frac{60}{\mathrm{RR}},$$

where 60 is a number of seconds in 1 minute; RR is the duration of interval between two R waves (sec). Normal range of FCC is 60–80 per minute.

Calculate corrected **QT interval.** The **QT interval** is the time from the start of ventricular depolarization to the end of ventricular repolarization, it corresponds to the time of mechanical systole. Corrected duration of the QT interval is determined by **Bazett formula**:

$$\mathrm{QT_c} = \frac{\mathrm{QT}}{\sqrt{\mathrm{RR}}},$$

where **QT_c** is the corrected interval between **QT** and **RR** is the interval between two neighboring R waves (sec). Normal range of corrected **QT_c** interval is 0.45 second (450 msec) in men and 0.46 second (460 msec) in women.

Systolic index (SI) is characterized by the ratio of **QT** interval toward duration of cardiac cycle (**RR**). Normal range of SI is 40 ± 5%.

$$SI = \frac{QT}{RR} \times 100\%$$

2.3.2. Determination of the electric axis of the heart using the EEG record

The projection of the resulting vector of QRS on a frontal plane is called the **electrical axis of the heart**. Localization of the electrical axis of the heart is defined by **angle α**, which is formed by the electrical axis of the heart and positive axis of the first standard lead (Fig. 2.3). The **α** angle can be determined by a graphical method. To build the electrical axis of the heart 1st and 3rd (I and III) standard leads are used.

For determination of the electrical axis of the heart, calculate the algebraic sum of the Q, R, S waves of the 1st and 3rd lead (as shown on Fig. 2.4A). Construct the projection of this sum on the corresponding axis (as shown on Fig.2.4A). Draw the lines, which are perpendicular to the axes, I and III respectively and find a crossing point. Then, connect this crossing point to the zero point of the graph (See Fig. 2.4). Measure the angle α. Normally, the α angle depends on the type of body constitution. Normally, the α angle depends on the type of body constitution. In normostenic person α = 30–69°, in hyperstenic person – 0–29° and in astenic person – 70–90° (See Fig. 2.4B).

Results and Discussion

Make conclusion about the identified axis of the heart. If there are any deviations from the normal, explain them.

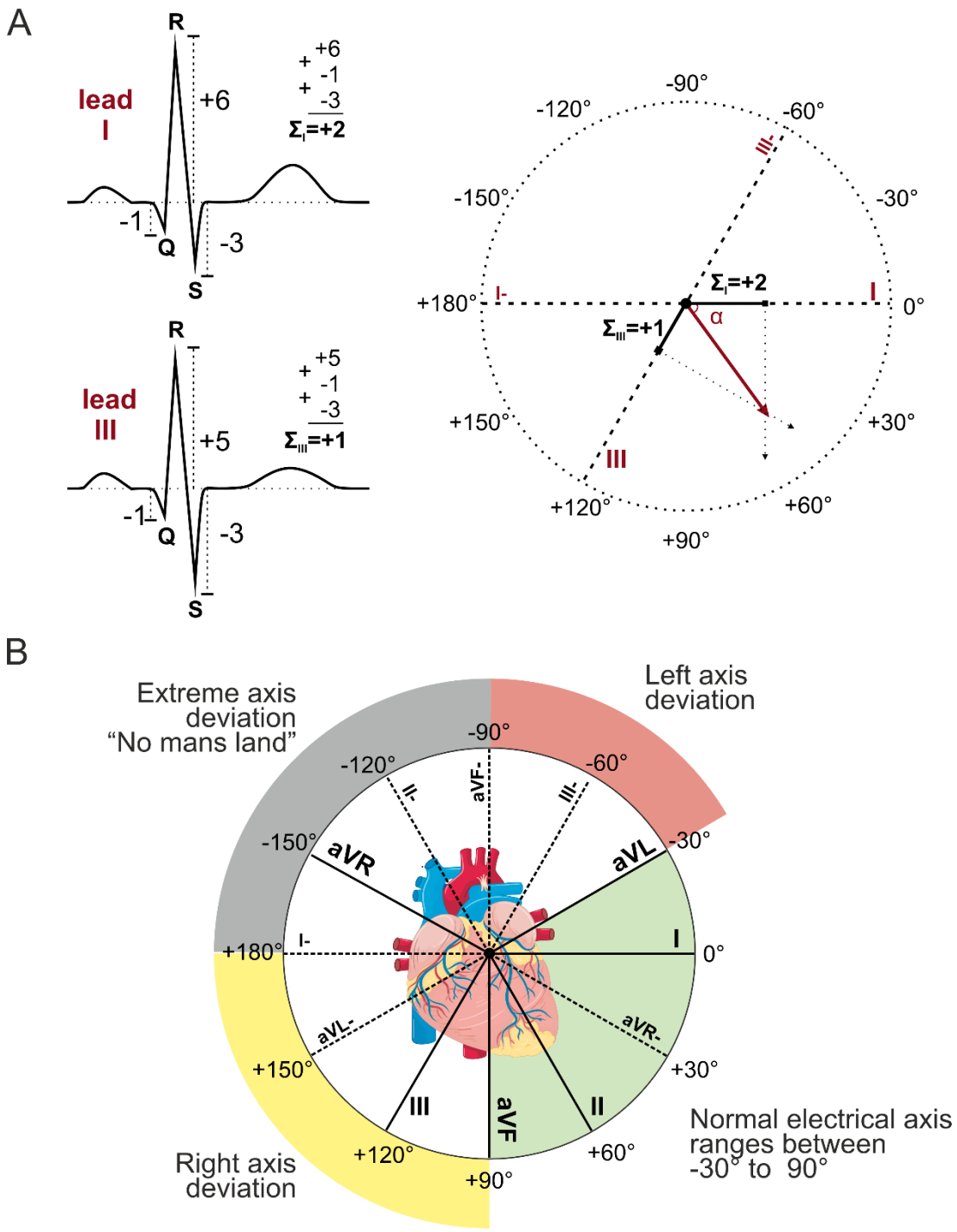

Figure 2.4. A Determination of direction of electrical axis of the heart. On the right graph: I is the axis of 1st lead, III is the axis of the 3rd lead. On the left graph, there are the algebraic sums of the QRS complexes of the 1st and 3rd leads. On the right graph, there is a resultant vector from the 1st and 3rd leads sums, and α angle. **B** Axis deviations

2.4. The effect of electrical stimuli on cardiac activity (virtual lab)

Open file `LUPRAFISIM.exe > START > English version > CONTENTS > HEART`. Do virtual works, following instructions in them.

The mechanical activity of the heart, known as **cardiac cycle**, is composed by the rhythmical succession of the distinct phases: **systole** (contraction phase of the cardiac muscle) and **diastole** (relaxation phase of cardiac muscle). The excitability of the cardiac muscle is changing during the cardiac: in systole the myocardium becomes unexcitable; in diastole the cardiac excitability reaches the highest level. Demonstration of the phases of cardiac cycle of the frog heart and the evolution of its excitability by using the graphical method.

Principle

The phases of the cardiac cycle of the frog heart are recorded on a graphical surface and the effect of electrical stimuli on the heart is determined. Graphical recording consists of two parts: recording of normal mechanical activity of the heart; recording of the mechanical activity under effect of electrical stimuli applied first during systole and then during diastole (Fig. 2.5).

The normal graphic of heart mechanical activity is sinusoid, where the ascending side is systole and the descending part is diastole.

Results and Discussion

1. Draw a graph of the cardiogram during normal conditions, after electrical stimulus applied during systole and after electrical stimulus applied during diastole.
2. Draw the conduction system of the human heart. What is a normal source of electrical activity in the heart?
3. Explain why in systole the myocardium becomes unexcitable and in diastole the cardiac excitability reaches the highest level. Provide your explanation with the drawing of the graphs of action potentials in typical and atypical cardiomyocytes, mention its ionic mechanisms and refractory periods, also the contraction-relaxation phases, aligned in time with excitation processes in typical cardiomyocytes.
4. What is extrasystole and compensatory pause? Explain their nature.

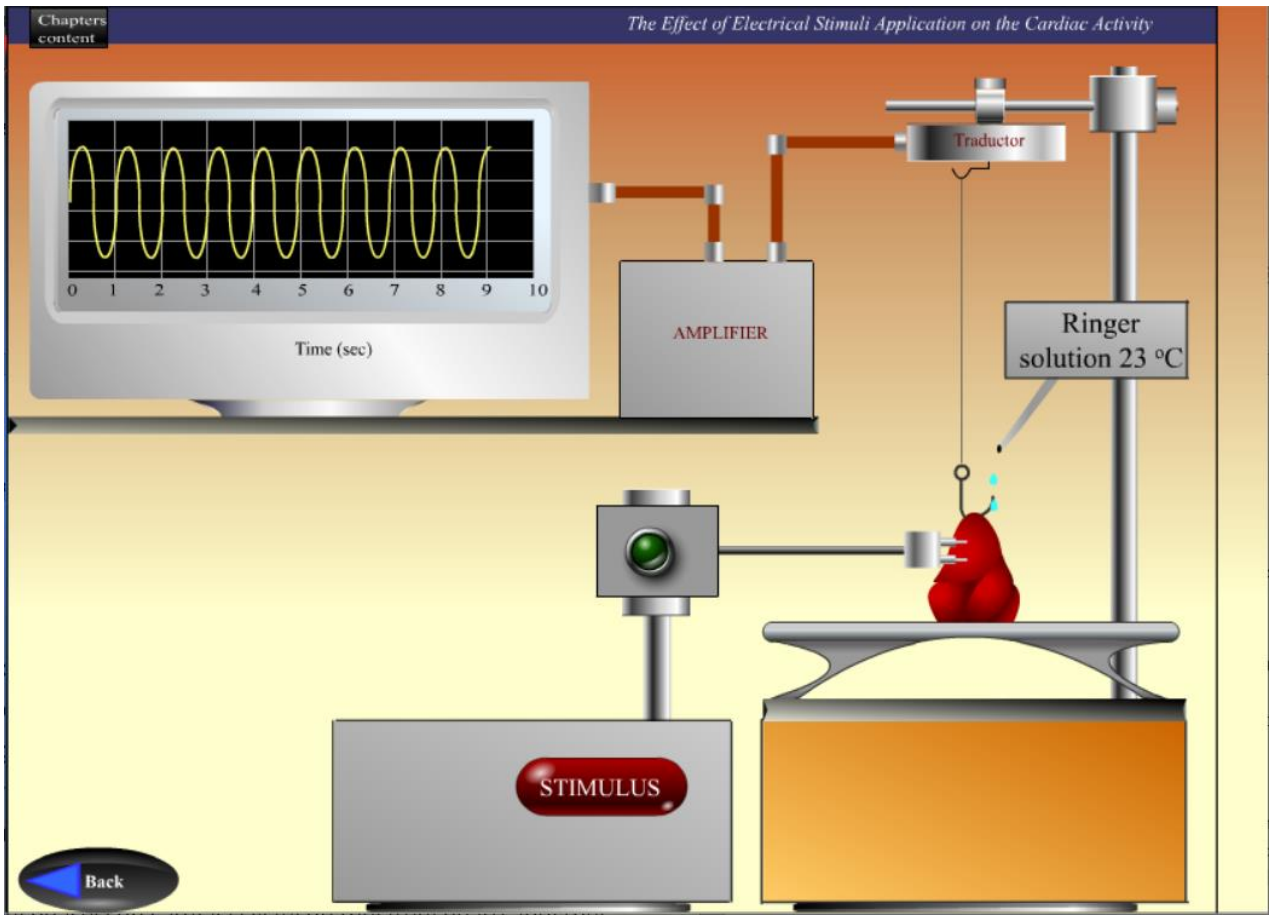

Figure 2.5. Rhythmic cardiac activity

2.5. The effect of several drugs and some chemical mediators on the cardiac activity (virtual lab)

The **cardiac automatism** is the ability of the cardiac muscle to contract rhythmically and independently without the intervention of other extracardiac regulating factors. This property provides the heart the ability to contract rhythmically even when all the nervous, vascular and physical connections of the organ with the rest of the body is ceased.

The completely isolated heart can continue its activity if the following conditions are assured: (1) perfusion (circulation of a liquid through the cardiac compartments) with solution under certain pressure; (2) the solution used for perfusion must provide the energetic substrate necessary to the cardiac activity; (3) optimal temperature. Under these circumstances, the heart can continue its activity for a long time.

Principle

Record the mechanical activity of the isolated frog heart, while the heart is being perfused with isotonic fluid containing various concentrations of ions and chemical mediators (epinephrine and acetylcholine).

Results and Discussion

1. Observe effects of Ca^{2+} and K^{+} concentration change and effects of epinephrine (adrenaline) and acetylcholine on mechanical activity (amplitude and frequency of contractions). Fill the table 2.7.

Table 2.7. Changing parameters of heart contraction

	Frequency of contraction	**Amplitude of contraction**
Decreased concentration of Ca^{2+}		
Increased concentration of Ca^{2+}		
Increased concentration of K^{+}		
Adrenalin		
Acetylcholine		

2. Draw the changes of the cardiogram in each case.
3. Explain the results and their physiological mechanisms (including effects on the excitability and contractile ability of typical and atypical cardiomyocytes; types of receptors for adrenaline and acetylcholine in the heart, intracellular changes after their activation).
4. What intracardiac mechanisms regulate frequency and amplitude of contraction?

2.6. The effect of Vagal Excitation on cardiac activity (virtual lab)

Demonstrate the effect of the vagus nerve on cardiac activity

Principle

Apply electrical stimuli on the vagus nerve and record mechanical activity of the heart without stimulation, after short vagus nerve stimulation and after prolonged vagus stimulation.

Results and Discussion

1. Draw a graph of the heart mechanical activity and its changes during short-term and long-term vagal stimulation.
2. Explain physiological mechanisms of extracardiac regulation of the heart.
3. What is ventricular escape? Why does it happen?
4. Draw the cardiac conduction system

2.7. The influence of pressure and viscosity of a fluid, radius and length of the vessel (virtual lab)

Open file `LUPRAFISIM.exe > START > English version > CONTENTS > BLOOD VESSELS`. Do virtual works, following instructions in them. Demonstrate effect of different parameters on the flow

Principle

Set values of blood pressure, blood viscosity, length and radius of the vessel and measure flow of the blood. Change these values separately.

Results and Discussion

Write what values change flow positively and negatively. What value has the most impact? What values can organism change rapidly? Which of these values are local? Explain the mechanisms of the regulation of the blood flow. Write your results into the table 2.8.

Table 2.8. Effect of different parameters on the blood flow

BP	BV	LV	RV	Blood flow

2.8. The influence of the cardiac output, the peripheral resistance and the vascular elasticity on arterial pressure (virtual lab)

The **arterial pressure** is the force exerted by the blood flow on arterial walls. The arterial pressure is generated by:

Ventricular contractions – in each systole the heart pushes in the arteries a new quantity of blood over the previous one, which leads to creating and maintaining a certain blood pressure in these vessels;

Arterial wall elasticity does not allow the systolic pressure to exceed a certain level and the diastolic pressure to fall under a certain level (maintains the diastolic pressure);

Peripheral resistance – created in arterioles by the frictional force of the blood with a large surface represented by the summation of walls of the arterioles.

Aim: Demonstrate the influence of the cardiac output, the peripheral resistance and the vascular elasticity on arterial pressure

Principle

By changing mentioned parameters, measure systolic and diastolic pressure.

Results and Discussion

Write your results into the table 2.9. Write what values change blood pressure positively and negatively. How these parameters are normally regulated in our body?

Table 2.9. Effect of cardiac output, the peripheral resistance and the vascular elasticity on arterial pressure

	Cardiac output, ml/sec	Peripheral resistance	Vascular elasticity	Arterial pressure, mmHg
Value				
Special situations				

2.9. The effect of adrenaline, acetylcholine and atropine on arterial pressure (virtual lab)

Aim: Examine effects of mentioned substances on the arterial pressure

Principle

Record blood pressure changes before and after intravenous administration of substances (Fig. 2.6). Firstly, apply acetylcholine inducing the same effects as the parasympathetic activation. Secondly, apply adrenaline to induce the same effects as sympathetic activation. Thirdly, apply atropine, blocking effects of the parasympathetic system. Lastly, during the effect of atropine, apply adrenaline again.

Figure 2.6. Effect of adrenaline on arterial pressure

Results and Discussion

Draw the changes of arterial pressure in normal conditions and after injections of substances. Explain how blood pressure is regulated and how these substances change it. Which receptors are activated in the blood vessels?

What is the mechanism of their activation and effect?

Lab 3. PHYSIOLOGY OF RESPIRATION

The process of gas exchange in the body is called **respiration**. It has three basic steps: **pulmonary ventilation**, **external respiration** and **internal respiration**. **Pulmonary ventilation** is the inhalation (inflow) and exhalation (outflow) of air and involves the exchange of air between the atmosphere and the alveoli of the lungs. **External respiration** is the exchange of gases O_2 and CO_2 between the alveoli of the lungs and the blood in pulmonary capillaries. **Internal respiration** is the exchange of gasses O_2 and CO_2 between blood in systemic capillaries and tissue cells.

3.1. Determination of pulmonary volumes and capacities

Spirometry is a simple method for studying pulmonary ventilation. It includes the measurements of the volume of air moved into and out of the lungs. For describing the events of pulmonary ventilation, the air in the lungs has been subdivided into four volumes and four capacities. The **pulmonary volumes** are following:

The tidal volume (TV) is the amount of air that a person inspires or expires with each normal breath. In a healthy young human adult tidal volume is approximately 500 ml per one respiratory cycle.

The inspiratory reserve volume (IRV) is the amount of air that can be inhaled over and above the normal tidal volume when the person inhales with full force. It is about 3000 ml.

The expiratory reserve volume (ERV) the amount of air that can be expired by forceful expiration after the end of a normal tidal expiration (about 1100 ml).

The residual volume (RV) is the volume of air remaining in the lungs after the most forceful expiration. It is about 1200 ml maximum amount of air which can be expelled by a person after maximal inspiration.

Pulmonary capacities (PC) can help to describe the events of respiratory cycles. The respiratory capacities consist of two or more pulmonary volumes.

The **inspiratory capacity (IC)** equals the tidal volume plus the inspiratory reserve volume (about 3500 ml). It represents the amount of air which can be breathed in during normal and maximal inspiration.

The **vital capacity (VC)** consists of the inspiratory reserve volume plus the tidal volume plus the expiratory reserve volume. It is about 4600 ml.

The **functional residual capacity (FRC)** equals the expiratory reserve volume plus the residual volume. This is the amount of air that remains in the lungs at the end of normal expiration (about 2300 ml).

The **total lung capacity (TLC)** is a sum of vital capacity and residual volume. It is the maximum volume of air to which the lungs can be expanded with the greatest possible effort (about 5800 ml);

The aim of present work is to determine the **tidal volume**, **reserve volumes** and **vital capacity** of the lungs.

Requirements

Spirometer, disinfectant solution.

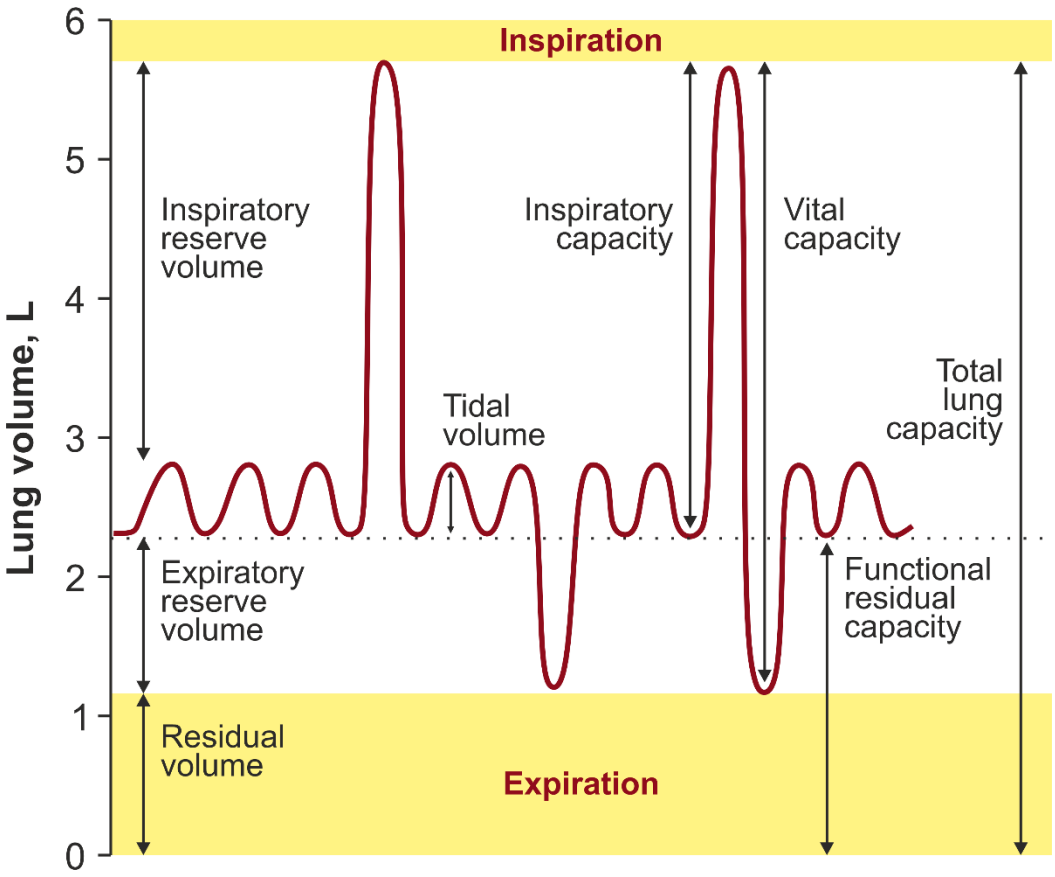

Figure 3.1. Pulmonary volumes and capacities

Procedure

Reset spirometer to zero. Wipe mouthpiece of spirometer with disinfectant solution. To define the vital capacity (VC) make 2 or 3 deep inhalations and

exhalations, then inhale as deeply as possible and exhale the air into the spirometer as much as you can. To determine the tidal volume (TV) make a normal exhale into the spirometer after a normal inhale. Repeat measurements 3 times. Calculate the average volume. To define expiratory reserve volume (ERV) you should make the deepest additional exhalation into the spirometer after the normal exhalation. To determine the inspiratory reserve volume (IRV) you can use the formula:

$$\mathrm{IRV} = \mathrm{VC} - (\mathrm{TV} + \mathrm{ERV})$$

Write down your results in table 3.1.

Table 3.1. Respiratory volumes before and after exercise

Respiratory volumes	Before exercise	5 min after exercise
VC		
TV		
RV_{exp}		
RV_{ins}		

The sum of TV, ERV and IRV is the **VC**. Compare measured and calculated VC. Explain the difference. Measure **all respiratory volumes 5 minutes after exercise** (30 squats). Compare volumes before and after exercise. VC can also be calculated by the formula:

For men: $\mathrm{VC} = 0.052 \times \mathrm{P} - 0.028 \times (\mathrm{A} - 3.2)$,

For women: $\mathrm{VC} = 0.049 \times \mathrm{P} - 0.019 \times (\mathrm{A} - 3.76)$,

where **P** – weight in kg, **A** – age in years. Compare the value of **VC** calculated by this formula with the measured by spirometer. Make conclusions.

3.2. Study of the functional state of the respiratory system

The aim of this work is to study the methods of defining the functional state of the respiratory system; rate the functional possibilities of the respiratory system and study the resistance to excess of carbon dioxide.

3.2.1. Apnea tests.

There are two different tests which determine the oxygen supply and overload of carbon dioxide in the body. It is used to determine the functional state of the respiratory system. **Shtange's test** evaluates the time of holding breath after inhale and **Genchi's test** determines the time of holding breath after exhale.

Requirements

A stopwatch.

Procedure

In a sitting position, make 2–3 deep breaths. For Shtange's test, make a deep **inhale** and measure the longest time you can hold your breath after inhale. For Genchi's test, make 2–3 deep breaths, after that make a normal **exhale** and measure the longest time, you can hold your breath after exhale. While holding the breath you should close your mouth and nose with your hands. Measure the maximum time at which you can hold your breath. Repeat 2–3 times, calculate the **average time**. Using table 3.1, rate the results.

Table 3.1. The results of Stange's and Genchi's tests.

Respiratory functional state	Time of holding breath after inhaling,sec	Time of holding breath after exhaling, sec
Excellent	>60	>50
Good	40–60	30–50
Satisfying	30–40	20–30
Unsatisfying	<30	<20

3.2.2. Resistance of the respiratory center to excess of carbon dioxide

Requirements

A stopwatch

Procedure

Measure the heart rate by counting the pulse during one minute in standing position. Using the result of **Genchi's test**, calculate the indicator of **resistance to excess of carbon dioxide (RI)**.

$$RI = \frac{HR}{T_a},$$

where **HR** is a heart rate and $\mathbf{T_a}$ is a time of apnea in Genchi's test. Normal value is RI⩽1. If RI is lower than 1, the resistance of the respiratory center to excess carbon dioxide is better. If RI from 1 to 1.19 there is a light pathology, RI in interval from 1.19 till 1.99 shows the pathology of medium degree, and from 2-5.99 is severe degree of pathology. If RI is from 6 to 11, this condition is dangerous for life.

3.2.3. The maximum time of apnea after a dosed physical load

Requirements

A stopwatch.

Procedure

Hold the breath for the maximal possible time in a sitting position after the normal exhale. 5 minutes later make 20 squats within 30 seconds. Immediately after squats hold your breath after exhaling in a sitting position and measure the time. Rate your result using table 3.2.

Table 3.2. Results of the functional test of holding the breath before and after a dosed physical load.

Category	Normal time of holding the breath at rest, sec	Time of holding the breath at rest, sec	Time of holding the breath after 20 squats
Healthy trained	46–60	More than 100% from normal	More than 50% from normal
Healthy untrained	36–45	70–100% from normal	30–50% from normal
With health dysfunctions	20–35	Less than 70% from normal	30% and less from normal

3.3. The mechanism of respiration, pulmonary volumes and capacities, the role of diameter of the airways (virtual lab)

Open file `LUPRAFISIM.exe > START > English version > CONTENTS > RESPIRATORY SYSTEM`. Do virtual works, following instructions in them.

The aim of this work to observe a graphical demonstration of different volumes and capacities of the lungs, and evaluate the influence of trachea radius on the pulmonary measurements.

Principle

Graphically record a series of tidal inspirations and expirations followed by forcible inspirations and expirations and measure the pulmonary volumes and capacities. Repeat the experiment after reducing the radius of the trachea.

1. Click the `“START”` button of the experimental device closely observing the recording of the pneumatogram during the tidal respiration and then during the forcible respiration.
2. Record the data regarding the pulmonary volumes and capacities shown by the device. This data is acquired while the radius of trachea was set as 5 cm.
3. Reduce the radius of the trachea using the right buttons of the device and then repeat steps 1 and 2. In the Fig. 3.2 the radius of the trachea was set as 4 cm. Note the difference in measurements between the first and second experiment.
4. Perform more measurements, setting the radius of trachea as 4.5 cm and 3 cm and observing the pneumatogram. Fill the following table 3.3 with your measurements.

Results and Discussion

1. Draw the graph of all respiratory volumes and capacities. What parameters of the respiratory system pulmonary volumes and capacities can describe?
2. How can they be useful in the clinical practice of the doctor to evaluate the functional state of the respiratory system?
3. How does the radius of trachea affect the act of respiration and measurements of pulmonary volumes and capacities? How it’s regulated?
4. Make your conclusions about the influence of tracheal radius on parameters of respiration.

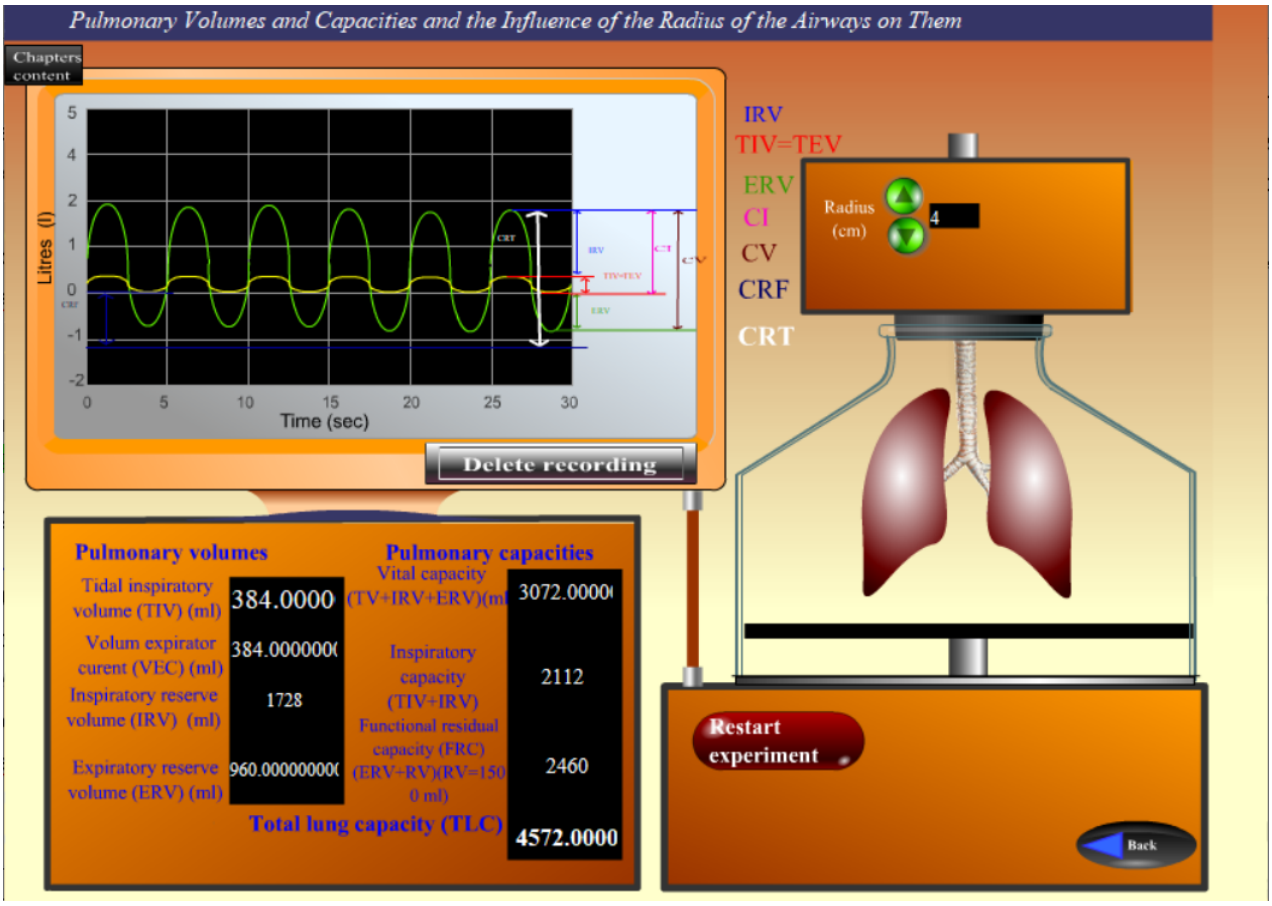

Figure 3.2. Pulmonary volumes and capacities. Yellow line in the graph represents tidal respiration, while the green line represents forceful respiration

Table 3.3. Pulmonary volumes and capacities change with airway diameter

Radius of trachea, cm	**3**	**4**	**4.5**	**5**
Pulmonary volumes, ml				
Tidal inspiratory volume (TIV)				
Volume expiratory current (VEC)				
Inspiratory reserve volume (IRV)				
Expiratory reserve volume (ERV)				
Pulmonary capacities, ml				
Vital capacity (TV+IRV+ERV)				
Inspiratory capacity (TIV+IRV)				
Functional residual capacity (FRC) (ERV+RV)				
Total lung capacity (TLC)				

3.4. The influence of pulmonary space pressure on pulmonary ventilation (virtual lab)

The pleural space is the space between the visceral pleura (covering the lungs) and the parietal pleura (lining the thoracic cavity). This space is normally filled with a small amount of pleural fluid, which acts as a lubricant to allow the lungs to smoothly expand and contract during breathing.

Pleural space pressure has lower values than atmospheric pressure. As a consequence, there is a permanent contact between the external surface of the lungs and the thoracic wall, allowing the lungs to closely follow the movements of the thoracic wall.

Pneumothorax is the condition where air enters the pleural space, either through a communication between the pleural cavity and the exterior, or from a rupture of the lung itself. When this happens, the volume of the lungs diminishes, and they are no longer able to follow the movement of the thoracic walls during respiratory movements.

The aim of the work to observe normal respiratory movements of the lungs and to witness the influence of artificially induced pneumothorax on the presence of respiratory movements.

Principle and technique

Record the respiratory movements before and after opening the pleural cavity (producing pneumothorax).

1. Click the "START" button of the experimental device
2. Observe closely the respiratory movements and the graphical recording. Under the normal conditions, you will witness rhythmic movements of the lungs accompanied by graphical recordings of inhalation and exhalation acts:
3. Click the "OPEN VALVE" button to produce pneumothorax condition.

Once the valve is opened, you will witness quick and abrupt deflation (decrease of volume) of the lungs. Note that inhale-exhale movements of the lungs immediately stopped after deflation.

Results and Discussion

Write down your observations about the presence of respiratory movements in normal conditions and after pneumothorax was evoked. In the discussion part, answer the following questions.

1. Why is pleural space pressure important to maintain the normal act of breathing?
2. Why are lungs able to maintain respiratory movements in normal conditions?
3. Why does disruption of normal pleural space pressure lead to inability to maintain respiratory movements?
4. What can lead to disruption of pleural space pressure?
5. Make conclusions about the importance of pleural space in the act of breathing.

3.5. The influence of surfactant on pulmonary ventilation (virtual lab)

The surfactant is a complex of phospholipids secreted by the type II alveolar cells. These cells are located on the inner side of the alveoli, and their surface tension lowering action prevents the collapse of the alveoli. This way the alveoli remain open even at the end of extremely forcible expirations.

The aim of the work to observe the effect of adding surfactant on the intensity of the respiratory movements.

Principle and technique

Record the pneumogram before and after adding different amounts of surfactant into the lungs.

1. Click the "START" button of the experimental device.
2. Observe the movements of the lungs and the recording of the respiratory movements.
3. Record the tidal volume (the volume of air that moves in or out of the lungs with each respiratory cycle). With the absence of surfactant in the lungs, you will see the following measurements in Fig. 3.3.

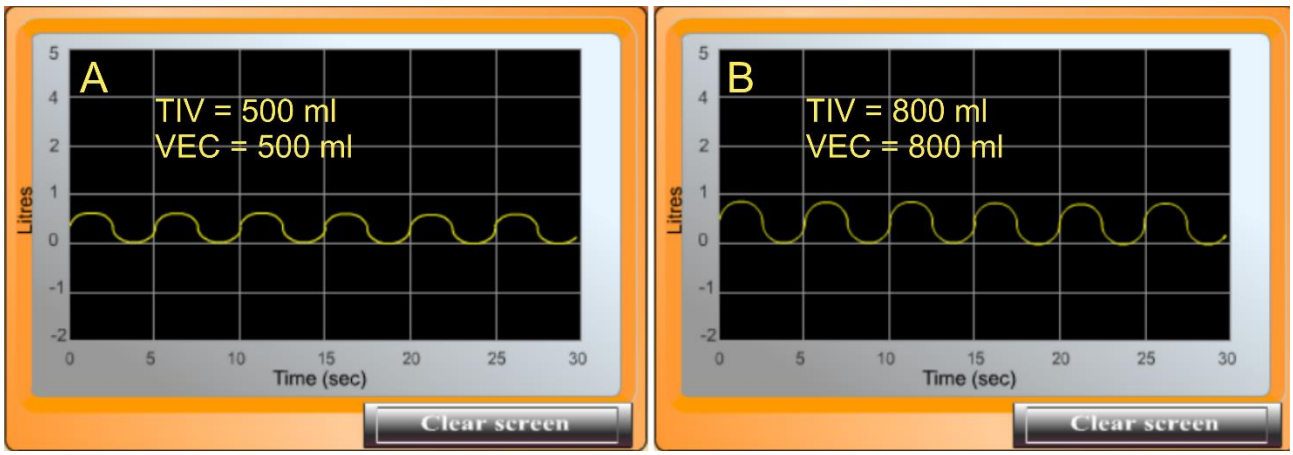

Figure 3.3. Tidal volume change without and with surfactant

4. Click the "SURFACTANT" button (this introduces surfactant into the lungs) 3 times.
5. Click the "START" button again.
6. Observe closely the pulmonary movements and their recording.
7. Record the value of the tidal volume and compare it to the value obtained at the previous step. Under these conditions, you will witness more pronounced pulmonary movements and increased tidal volume, which will be 800.
8. Repeat the same experiment, adding more surfactant (for example, click on the "SURFACTANT" button 6 times and next, 9 times). Observe the pulmonary movements and record the values of tidal volume.

Results and Discussion

Write down obtained values of tidal volume and draw graphs of respiratory movements in following conditions: (1) in the absence of surfactant; (2) in the presence of a relatively small amount of surfactant; (3) in the presence of a relatively large amount of surfactant. In the discussion part, answer the following questions.

1. What is surfactant and how is it important to maintain the normal respiratory act? How can the amount of surfactant affect the intensity of the respiratory movements?
2. Make assumptions based on scientific sources about reasons, which can lead to the absence or deficiency of surfactant in the lungs.
3. Make conclusions about the importance of the surfactant in the act of breathing.

Lab 4. PHYSIOLOGY OF DIGESTION

4.1. Enzymatic properties of human saliva

Ingested food is mixed with saliva in the oral cavity. The ducts of the salivary glands open to the mouth. Saliva is a watery substance (98.5–99.5% of water), which contains enzymes, organic substances, salts, anions and other micronutrients. The salivary **enzyme amylase** begins the digestion of starches, by catalyzing the hydrolysis of carbohydrates.

Requirements

A thermostat or a water bath, spirit lamp, rack of 10 tubes, pipettes, human saliva, 1% solution of boiled starch, 1% solution of iodine, a solution of 10% NaOH, 0.5% HCl solution, ice (snow), 2% copper sulfate solution ($CuSO_4$), sheets of filter paper.

Procedure

Collect 5 ml of saliva into the test tube. Dilute it with distilled water (1:1) and filter it with paper. Take 8 tubes, mark them from 1 to 8, add following components and mix it carefully:

1. 3 ml of boiled starch + 1 ml of diluted human saliva
2. 3 ml of boiled starch + 1 ml of diluted boiled human saliva
3. 3 ml of boiled starch + 1 ml of distilled water (H_2O)
4. 3 ml of boiled starch + 1 ml of diluted human saliva + 1 ml of HCl (0.5%)

Put all tubes at the thermostat with 37°C for 10 min. Divide solutions of each tube into two parts. Take the first parts and make the **Trommer's test**, which allows determining the presence of monosaccharides. For this purpose, add 0.5 ml of NaOH (10%), 2–3 drops of 2 % solution of sulfuric acid copper ($CuSO_4$) into each tube. The mixed solutions turn to blue, due to the formation of copper hydroxide $Cu(OH)_2$. Then, gently heat each tube on a spirit lamp. In the presence of monosaccharides, the blue staining of the solution changes to yellow, and then the solution becomes orange-red.

Take the second part of tubes. Determine the rate of the starch hydrolysis by adding 2–3 drops of iodine solution. If unhydrolyzed starch is presents, then the

color of the solution becomes blue (positive reaction). The more intensive is the color, the more starch is present in the solution.

Results and Discussion

Write down the experimental results in the table 4.1. Make conclusion based on your results. Describe optimal conditions for salivary amylase activity.

Table 4.1. Experimental results

Tube	**Solution content**	**Trommer's reaction** (positive or negative, color)	**Starch hydrolysis test** (positive or negative, color)
1	3 ml of boiled starch + 1 ml of diluted human saliva		
2	3 ml of boiled starch + 1 ml of diluted boiled human saliva		
3	3 ml of boiled starch + 1 ml of distilled water (H_2O)		
4	3 ml of boiled starch + 1 ml of diluted human saliva + 1 ml of HCl (0.5%)		

4.2. Effect of bile on fats

Bile is produced by the liver and released into the duodenum. Bile contains specific organic substances: bile acids, bile pigments (bilirubin and biliverdin), lecithin, cholesterol, mucin and inorganic salts. It activates the enzymes secreted by the pancreas and intestinal glands by creating alkaline medium, emulsifies fats thus promoting their splitting and absorption, enhances bowel movement and excites secretion of pancreatic juice. During the day, the person produces from 0.5 to 1 liter of bile.

Requirements

A magnifying glass, slides, tubes, funnels, a pipette, fresh bile, oil, paper filters, water.

Procedure

The influence of bile on fats can be observed in the following way: apply a drop of water and a drop of bile by the pipette on a glass slide. Add a small amount of oil in each drop, mix it and see the contents of the two drops under

a magnifying glass. Put 2 tubes in a rack. In each tube, set the funnel and put a piece of filter paper in the funnel. In tube #1: soak the filter paper in funnel with water, tube #2 with bile. Fill the both funnels with the same amount of oil (2 ml). Leave for 30 minutes. Determine the amount of filtered oil in tubes #1 and #2.

Results and Discussion

How is fat distributed in a drop of water and in a drop of bile? Sketch it in the notebook. Which of the bile functions does it explain? Explain the results.

4.3. Substrate specificity of salivary amylase (virtual lab)

Open file `LUPRAFISIM.exe > START > English version > CONTENTS > DIGESTIVE SYSTEM`. Do virtual works, following instructions in them.

Enzymes are characterized by **substrate specificity**, which is the capacity of the enzyme to identify and act on a certain substrate **(absolute substrate specificity)** or identify and act on multiple substrates **(relative substrate specificity)**.

Salivary amylase is a glycolytic enzyme whose main substrates are **starch and glycogen**. It breaks down polysaccharides into monosaccharides, like maltose and glucose. Its activity is enhanced by the Cl^- ion and it is most efficient at a temperature of 37–38°C and a slightly alkaline pH (7.5–8).

Salivary lipase is an enzyme that breaks down fats (lipids) in the oral cavity. Its main substrates are triglycerides, which it hydrolyzes into fatty acids and glycerol. Salivary lipase is active in the slightly acidic environment of the mouth (pH 6–7).

The aim of this work to demonstrate the substrate specificity of salivary amylase.

Principle and technique

Salivary amylase is mixed with three different carbohydrates. The **Trommer's reaction** is performed to identify monosaccharides. The red color that appears at the end of the reaction proves that only starch is decomposed by the enzyme, demonstrating the substrate specificity of salivary amylase.

Equipment

Substrates for reaction (starch, sucrose, cellulose), salivary amylase, $CuSO_4$ 10%, NaOH 10%; thermostat, glass tube, Bunsen burner.

Procedure

1. Introduce sucrose and salivary amylase in a test tube.
2. Click the "START" button of the warming device.
3. Add a few drops of NaOH after the incubation period.
4. Add $CuSO_4$ 10% in the test tube.
5. Click "WARM UP THE SAMPLE" and the content of the test tube is boiled.
6. Analyze the resulting color.
7. Click the "RESTART EXPERIMENT" button.
8. Repeat the same steps, but instead of sucrose, add cellulose to the test tube.
9. Repeat the same steps, but instead of cellulose, add starch to the test tube and warm it up.

Results and Discussion

Write down your observations about performed experiments in the table 4.2. In the discussion part, answer the following questions.

Table 4.2. Trommer's reaction of determination of aldoses with $Cu(OH)_2$

	Substrate	**Trommer's reaction** (positive or negative)
1	Sucrose	
2	Cellulose	
3	Strach	

1. Based on the Trommer's reaction, which of the substances were not affected by amylase? Explain how does Trommer's reaction work.
2. What kinds of digestive enzymes are present in the human saliva?
3. What is substrate specificity of the enzyme?
4. Describe the chemical properties of salivary amylase, such as substrate, chemical media.
5. What is the specific action of salivary amylase to its substrate?
6. Make your conclusions about functions of salivary amylase as digestive enzyme of the oral cavity.

4.4. The influence of pH on the action of pepsin (virtual lab)

Pepsin is a proteolytic enzyme synthesized as inactive pepsinogen by the main cells of the **gastric glands**. Pepsinogen is turned into active pepsin when the pH is lower than 5. This is usually achieved by the presence of hydrochloric acid (HCl) in the gastric liquid. Pepsin belongs to the group of endopeptidases. It **splits peptides into polypeptidic chains** and is most efficient at a pH of about 2.

Aim: To demonstrate the influence of pH on the efficiency of pepsin by incubating ovalbumin and pepsin with and without hydrochloric acid, and then analyzing the degree of protein digestion.

Principle and technique

Incubation of ovalbumin and pepsin at 38°C for 3 hours, with and without hydrochloric acid, estimating the degree of protein digestion (diminishing of the dimensions of ovalbumin fragments).

Equipment

Experimental tube with clots of ovalbumin (protein of egg whites), thermostat, distilled water, HCl, pepsin.

Procedure

1. Add pepsin and HCl in the test tube that contains ovalbumin.
2. Click the "START" button of the warming device.
3. Analyze the degree of protein digestion.
4. Add combination pepsin and distilled water in the test tube that contains ovalbumin and repeat actions from points 1, 2, and 3.
5. Add hydrochloric acid and distilled water in the test tube that contains ovalbumin and repeat actions from points 1, 2, and 3.

Results and Discussion

Write down the experimental steps and your observations on performed experiments in the table 4.3. In the discussion part, answer the following questions.

1. What essential enzymes are responsible for digestion of proteins? Where they are synthesized? Which of them are endopeptidases and exopeptidases? How do they work?
2. What environment is necessary for gastric pepsin to function properly?

3. What can prevent proper digestion of food proteins in the stomach?
4. Make conclusions about the general role of gastric enzyme pepsin in digestion of food.

Table 4.3. The influence of pH on the efficiency of pepsin

	Combination of substances	**Reduction of ovalbumin** (positive or negative)
1	Ovalbumin + pepsin + HCl	
2	Ovalbumin + pepsin + H_2O	
3	Ovalbumin + HCl + H_2O	

4.5. Demonstration of the action of pancreatic lipase in the presence and absence of bile (virtual lab)

Pancreatic lipase is a lipolytic enzyme, which decomposes lipids into glycerol and fatty acids. The best temperature for the activity of pancreatic lipase is 37–38°C and the best pH is slightly alkaline (pH more than 7). The activity of pancreatic lipase is enhanced by **bile produced in pancreas**, which has tensioactive properties (enlarging the area of action of this enzyme).

The pancreatic **lipolytic enzymes** are lipase, phospholipase, and esterase, which digest fats. **The glycolytic** (carbohydrate digesting) enzymes are lactase and amylase, which breaks down starch into maltose, maltotriose, and dextrins.

The aim of this study to demonstrate the influence of bile on the activity of pancreatic lipase. The experiment involves comparing the results with and without the presence of bile to observe its effect on the activity of pancreatic lipase.

Principle and technique

Pancreatic lipase and vegetable oil are introduced in two test tubes, with and without bile. They are kept at a temperature of 37°C. Phenolphthalein (a pH indicator that becomes red/pink when the medium is alkaline (pH 8–10) and is colorless in more acidic pH 4–7) is added to both test tubes to check for the presence of fatty acids, which are produced as a result of the decomposition of lipids.

Procedure

1. Introduce vegetable oil, bile, and pancreatic lipase in the test tube.
2. Click the "START" button of the warming device.
3. Add phenolphthalein in the test tube after the incubation period.
4. Analyze the resulting color.
5. Introduce vegetable oil and pancreatic lipase in the test tube and repeat the same steps in the experiment.
6. Introduce bile and pancreatic lipase in the test tube and repeat actions from points 1, 2, 3, and 4.

Results and Discussion

For the results part, fill the following table 4.4 with your observations during experiments. For the discussion part, answer the following questions.

1. What general role pancreas play in digestion?
2. What enzymes are produced by pancreas and what is their importance?
3. What role does bile play in digestion?
4. How can the lack of pancreatic enzymes affect digestion?
5. Make conclusions about observed results on activity of pancreatic lipase.

Table 4.4. Reactions for phenolphthalein

	Combination of substances	**Color of solution after the addition of phenolphthalein**	**pH** (alkaline or acidic)	**Explain the pH value**
1	Oil +bile+ pancreatic lipase			
2	Oil + pancreatic lipase			
3	Oil + bile			
4	Pancreatic lipase + bile			

Lab 5. PHYSIOLOGY OF METABOLISM

Metabolism comprises the processes that the body needs to function. **Basal metabolic rate (BMR)** is the amount of energy that person needs to keep the body functioning at rest. Some of these processes are breathing, blood circulation, controlling body temperature, cell growth, brain and nerve function, and contraction of muscles.

5.1. Determination of the deviation of the BMR

The intensity of the metabolic processes in the organism is accompanied with some changes in activity of the internal organs, as, for example, in the cardiovascular system. If you know heart rate and the arterial pressure value of the subject, you can identify a deviation rate of the BMR from the normal BMR by the **Reed's formula** or **nomogram**.

Requirements

Sphygmomanometer, Reed's nomogram.

Procedure

Measure the pulse (P) and arterial pressure (systolic and diastolic pressure) of the subject at rest in a sitting position.

5.1.1. Calculation of BMR deviation using Reed's formula

Calculate the pulse pressure (PP) using the following formula

$$\mathrm{PP} = \mathrm{SP} - \mathrm{DP},$$

where **SP** is systolic pressure and **DP** is diastolic pressure.

Calculate the BMR deviation using the **Read's formula**:

$$\mathrm{BMR}_{\mathrm{dev}} = 0.75 \times (\mathrm{P} + \mathrm{PP} \times 0.74) - 72$$

The BMR deviation shows the percentage of deviation of the personal BMR from normal range.

5.1.2. Determination of BMR deviation using Reed's nomogram

Draw the Reed's nomogram in your lab notebook (Fig. 5.1). Then point value of pulse (**P**) on the left scale and the value of a pulse pressure (PP) on the right scale of the Reed's nomogram. Connect these points using the ruler. The crossing point with a middle line shows the BMR deviation value from normal BMR in percent. Compare results obtained by both methods: Reed's formula and Reed's nomogram.

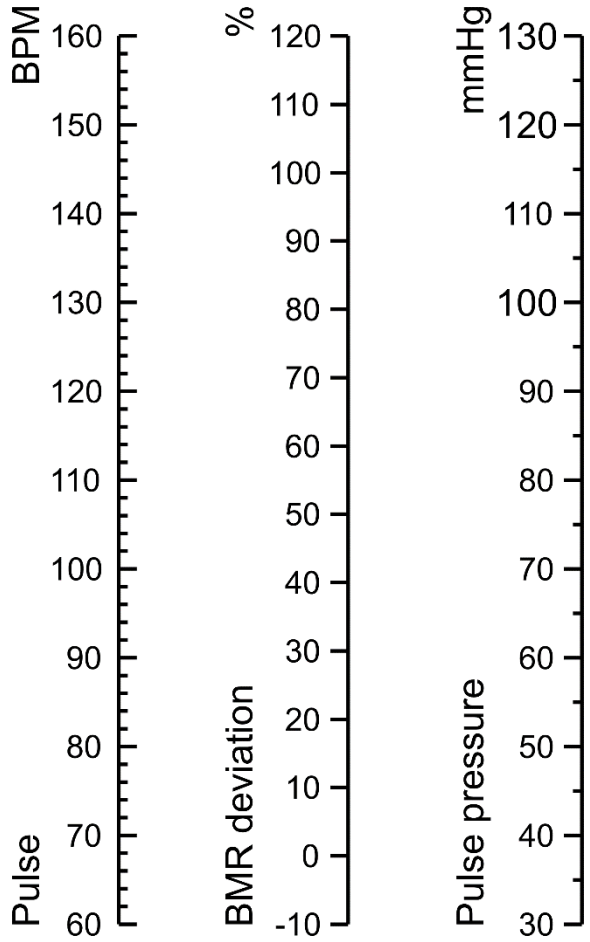

Figure 5.1. The Reed's nomogram

5.2. Determination of the BMR using the tables

One of the most widely used methods of evaluating the basal metabolic rate is the method for evaluating basal metabolism by tables of Harris-Benedict. There are two types of tables — for men (See Supplement 1) and for women (See Supplement 2). Each of them contains 2 parts, A and B. From the first part number A is found, it depends on body mass; and from the second part number

B, depending on height and age. The sum of these numbers (A+B) gives the value of BMR.

Requirements

Floor scales, height measurement device, standard tables for determining the BMR values (Supplement 1 and 2).

Procedure

Measure body weight (in kg) and height (in cm). Determine the BMR value using the appropriate tables (Supplement 1 for male, Supplement 2 for female). For example, the man is 25 years old, his height is 180 cm and body weight is 75 kg. Standard table consists of two parts: A and B. Find the subject's body weight (75 kg) in part A and find the value of the first part of BMR (1098 kcal). Then find age across part B (25 years), on vertical line find height (180 cm) and find the second part of BMR (732 kcal) at the intersection of lines. Sum the number from part A (1098 kcal) and part B (732 kcal). It will be the average statistical level of the subject's BMR. It is equal to 1830 kcal/day.

Calculate your own BMR using tables. Then use your results from Reed's nomogram to calculate your real BMR (considering percentage of your BMR deviation). For example, if BMR deviation is +10%, then your real BMR is equal 1830×1.1=2013 kcal/day.

5.3. Calculation of the energy expenditure during daily activity

Daily energy expenditure (EE) includes energy expenditure for all of the types of daily activities.

Procedure

Fill in the table 5.1, which is presented below due to your own types and duration of the activities during the day.

Table 5.1. Types and duration of the activities during the day.

Type of activity	Duration of activity, hours	Energy expenditure due to subject's body weight, kcal/hour × kg of body weight	Energy expenditure during the activity time, kcal

Use Supplement 3 to calculate the values of the third column of table 5.1. Energy expenditure during the activity time is calculated by multiplying the second and third columns. Total number of hours must be 24. Count your own EE for each type of activity (Table from Supplement 3 value multiply to your body weight and duration of this activity). Amount of all of the EE during the day of the subject, (summated values of column 4 in the table 5.1), will compose the daily energy expenditure.

Results and Discussion

Calculate your daily energy expenditure.

5.4. Analysis of the energy value of students' daily diet

Caloric value of the daily diet must cover all EE and BMR of the person.

Procedure

Compose an approximate menu for one day of your life. Separate breakfast, lunch, dinner, supper and snacks. Using any diet application or information on the package, calculate the energy value and containing of your own daily diet. Compare it with your daily EE value summated with your real BMR. Put it in the table 5.2.

Results and Discussion

Compare total energy value of your diet with EE and BMR found in previous work. Make conclusions about the ratio of proteins, carbohydrates and fats in your diet, mention which correction should be done to make it healthier.

Table 5.2. Energy value of daily diet

	Dishes	**Amount per serving**		**Carbohydrate, g**	**Protein, g**	**Fat, g**
		Size, g	Calories, kcal			
Breakfast						
Lunch						
Dinner						
Total						

5.5. Body mass index and body composition scaling

Body mass index (BMI) is a value derived from the mass (weight) and height of a person. The BMI is defined as the body mass divided by the square of the body height, and is expressed in units of kg/m^2, resulting from mass in kilograms (kg) and height in meters (m).

$$\mathrm{BMI} = \frac{\text{body mass}}{(\text{body height})^2}$$

Requirements

Smart scale

Procedure

Search `"Fitdays"` in App Store or Google Play. You also can scan below QR code to download the APP (Fig. 5.2). App will indicate you to turn on Bluetooth, and turn on GPS & location permission if your phone is Android system when you start the App.

Click to `“Test”` as showed on Fig. 5.2 (step 1), add personal data (step 2–4) and confirm (step 5). Entering the wrong gender, age or height may results in measurements being inaccurate.

Tap your foot on the scale to activate the display. Wait until the display shows `“0.0”` (step 6). Step on the scale with **bare feet** to turn the scale on. If stepping on the scale with socks, only weight and BMI will be measured and displayed on the main page. The number on the display flashes when the scale is being weighed and analyzed. Continue to stand on the scale until the circle is stable and shows your phone has received all the data from the scale.

You should save or share your report by scrolling down (steps 7–8). Write down your results in table 5.3. standard part contains evaluation from the app (low/standard/ high/ too high etc.). Make conclusion on basis of your results.

Table 5.3. Recorded parameters

Indicator	BMI	Body fat rate	Visceral fat	Body water	Skeletal muscle rate	Muscle mass	Bone mass	Protein	BMR
Value									
Standard									

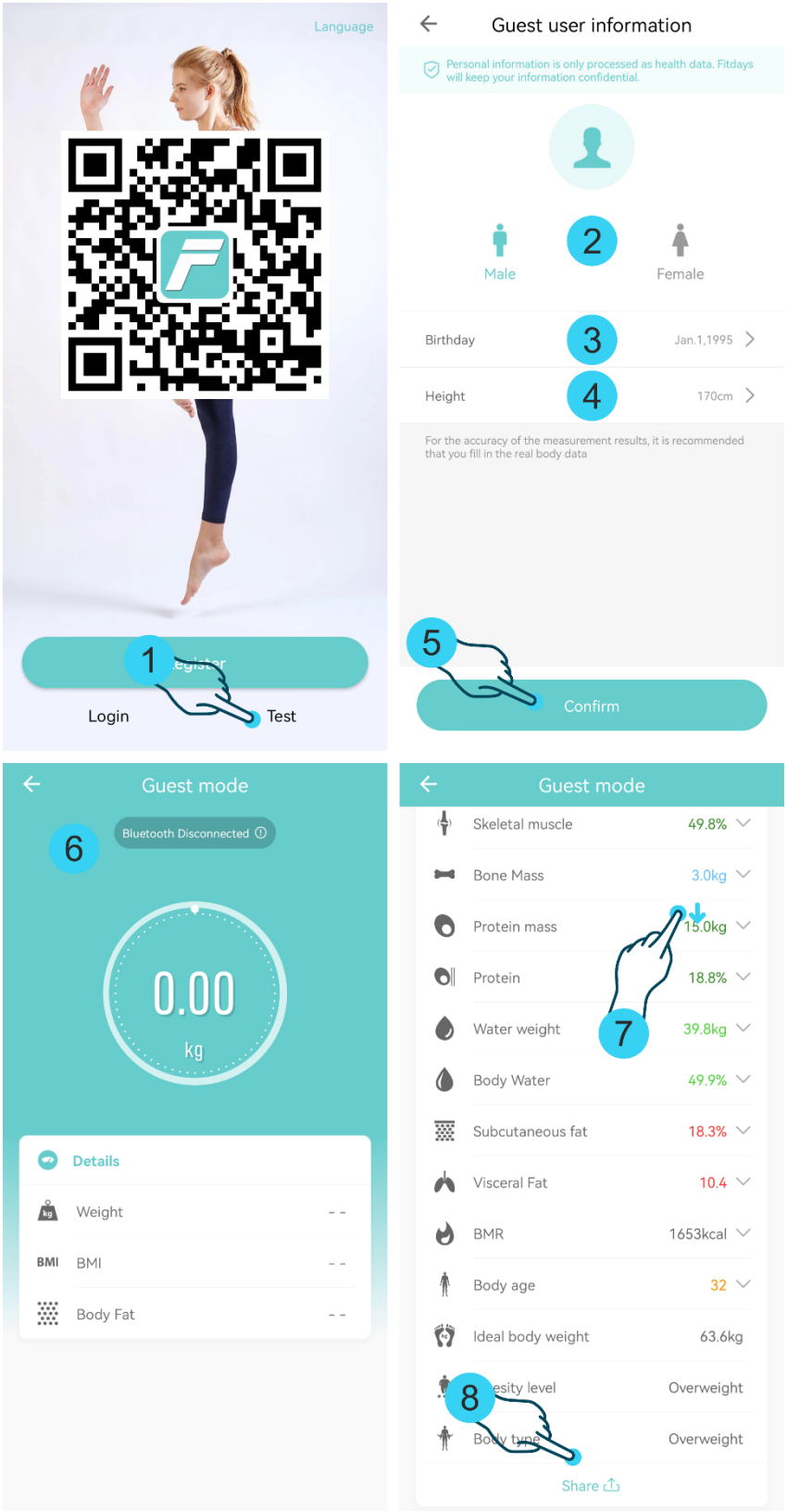

Figure 5.2. Instruction for Fitdays App

Lab 6. PHYSIOLOGY OF EXCRETION

6.1. Simulating glomerular filtration (virtual lab)

This computerized simulation allows you to explore one function of a single simulated nephron, glomerular filtration. The concepts you will learn by studying a single nephron can then be applied to understand the function of the kidney as a whole. Please, open `PhysioEx > Exercise 9: Renal System Physiology >` from the top of menu click `Experiment > Simulating Glomerular Filtration` (Fig. 6.1). The main features on the screen when the program starts are a **simulated blood supp**ly at the left side of the screen, a **simulated nephron** within a supporting tank on the right side, and a **data control unit** at the bottom of the display. The left beaker is the **"blood" source** representing the general circulation supplying the nephron. The **"blood pressure"** in the beaker is adjustable by clicking the (+) and (–) buttons on top of the beaker. A tube with an adjustable radius called the **afferent flow tube** connects the left beaker to the simulated glomerulus. Another adjustable tube called the **efferent flow tube** drains the glomerulus. The afferent flow tube represents the **afferent arteriole** feeding the glomerulus of each nephron, and the efferent flow tube represents the **efferent arteriole** draining the glomerulus. The outflow of the nephron empties into a **collecting duct**, which in turn drains into another small beaker at the bottom right part of the screen. Clicking the valve at the end of the collecting duct (which currently reads **"valve open"**) stops the flow of fluid through the nephron and collecting duct.

The **Glomerular pressure** window on top of the nephron tank displays the pressure within the glomerulus. The **Glomerular Filt**. **Rate** window indicates the flow rate of the fluid moving from the lumen of the glomerulus into the lumen of the renal tubule.

Procedure

The concentration gradient bathing the nephron is fixed at 1200 milliosmoles (mOsm). Clicking `"Start"` begins the experiment. Clicking `"Refill"` resets the equipment to begin another run.

If you need help identifying any piece of equipment, choose "Balloons On/Off" from the "Help" menu and move the mouse pointer onto any piece of equipment visible on the computer's screen. As the pointer touches the object, a pop-up window appears identifying the equipment.

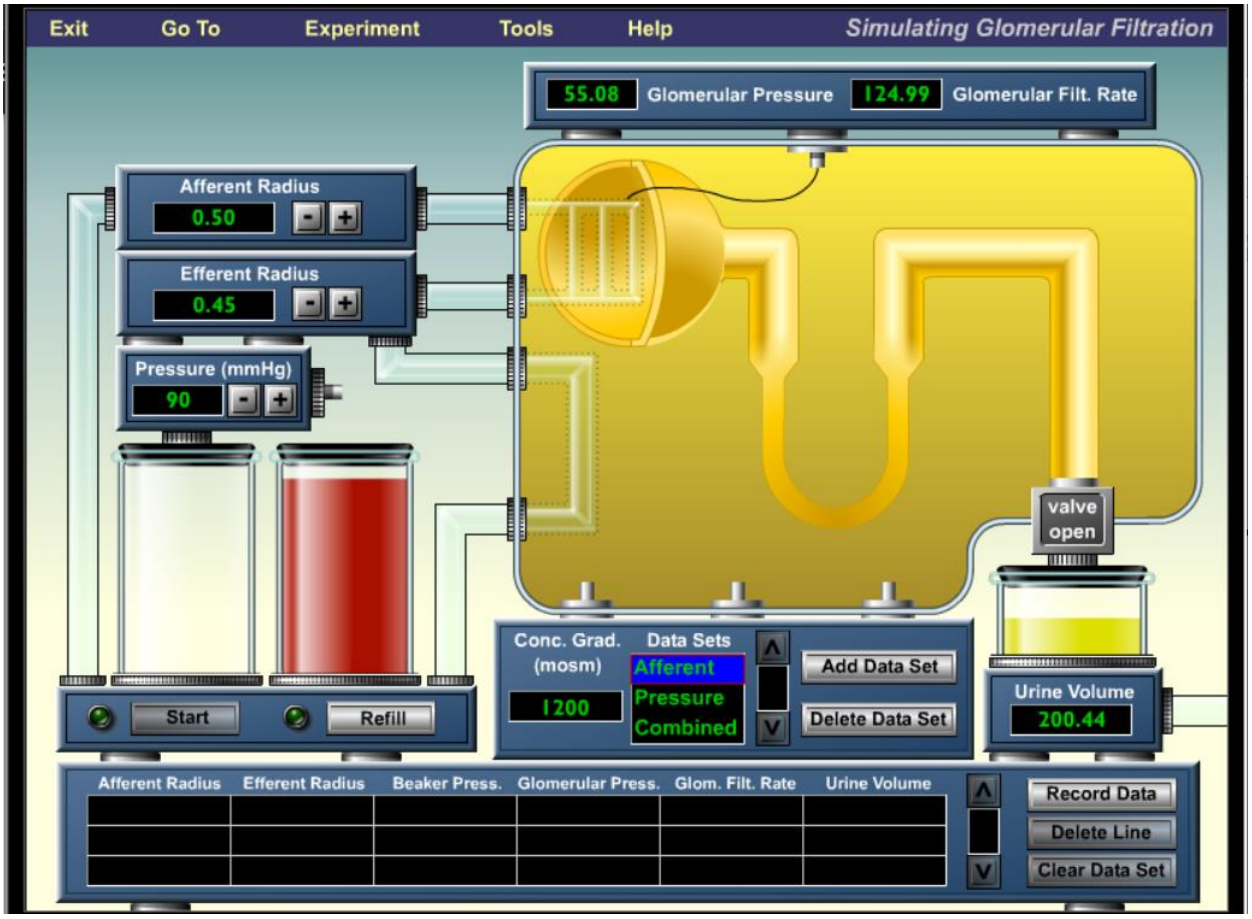

Figure 6.1. Opening screen of the simulating glomerular filtration experiment

6.1.1. Investigating the effect of flow tube radius on glomerular filtration (virtual lab)

Procedure

Click "Start" to see the on-screen action. Continue when you understand how the simulation operates. Click "Refill" to reset the experiment.

6. The "Afferent line" in the "Data Sets" window of the data control unit should be highlighted in bright blue. If it is not, choose it by clicking the "Afferent" line. The data control unit will now record **filtration rate variations due to changing afferent flow tube radius**.
7. If the data grid is not empty, click "Clear Data Set" to discard all previous data.

8. Adjust the afferent radius to 0.35 mm and the efferent radius to 0.40 mm by clicking the appropriate (+) or (–) buttons.
9. If the left beaker is not full, click "Refill".
10. Keep the beaker pressure at 90 mm Hg during this part of the experiment.
11. Click "Start", and watch the blood flow. Simultaneously, filtered fluid will be moving through the nephron and into the collecting duct. The Glomerular Filtration Rate window will display the fluid flow rate into the renal tubule when the left beaker has finished draining.
12. Now click "Record Data" to record the current experiment data in the data grid. Click "Refill" to replenish the left beaker and prepare the nephron for the next run.
13. Increase the afferent radius in 0.05 mm increments and repeat steps 6 and 7 until the maximum radius (0.6 mm) is achieved. Be sure to click "Record Data" after each trial. If you make an error and want to delete a single value, click the data line in the data grid and then click "Delete Line".

Results and Discussion

What happens to the glomerular filtration rate as the afferent radius is increased? What happens to the glomerular pressure as the afferent radius is increased? Predict what effect increasing or decreasing the efferent radius will have on glomerular filtration rate. Use the simulation to check your prediction and record your results. Make a general conclusion about the effects of flow tube radii and pressures on the rate of glomerular filtration. Put your results into the table in your lab notebooks according to Fig. 6.1 in data control unit.

6.1.2. Studying the effect of pressure on glomerular filtration (virtual lab)

Both the blood pressure supplying the glomerulus and the pressure in the renal tubule have a significant impact on the glomerular filtration rate. In this activity, the data control unit will record filtration rate variations due to changing pressure.

Procedure

1. Click the “Pressure” line in the “Data Sets” window of the data control unit.
2. If the data grid is not empty, click “Clear Data Set” to discard all previous data.
3. If the left beaker is not full, click “Refill”.
4. Adjust the pressure in the left beaker to 70 mm Hg by clicking the appropriate (+) or (–) button.
5. During this part of the experiment, maintain the afferent flow tube radius at 0.55 mm and the efferent flow tube radius at 0.45 mm.
6. Click “Start”, and watch the blood flow. Filtrate will move through the nephron into the collecting duct. At the end of the run, the **glomerular filtration rate** window will display the filtrate flow rate into the renal tubule.
7. Now click “Record Data” to record the current experiment data in the data grid. Click “Refill” to replenish the left beaker.
8. Increase the pressure in the left beaker in increments of 10 mm Hg and repeat steps 6 and 7 until the maximum pressure (100 mm Hg) is achieved. Be sure to click “Record Data” after each trial. If you make an error and want to delete a single value, click the data line in the data grid and then click “Delete Line”.

Results and Discussion

What happened to the glomerular filtration rate as the beaker pressure was increased? What was the effect on glomerular pressure as beaker pressure increased? Write formula for the glomerular filtration rate (GFR). Based on your results draw the graph that shows dependence between pressure and GFR. Make a general conclusion about impact of blood pressure on GFR.

6.2. Simulating urine formation (virtual lab)

This part of the simulation allows you to explore some aspects of urine formation by manipulating the **interstitial solute concentration**. Other activities

include investigating the effects of **aldosterone** and **ADH (antidiuretic hormone)** and the role that **glucose carrier proteins** play in renal function.

Procedure

Choose "Simulating urine formation" from the "Experiment" menu (Fig. 6.2).

Click "Dispense" to fill the tank through the jets at the bottom of the tank with the chosen solute gradient. Click "Start" to begin a run. While the experiment is running, the concentration probe can be clicked and dragged over the nephron to display the solute concentration within.

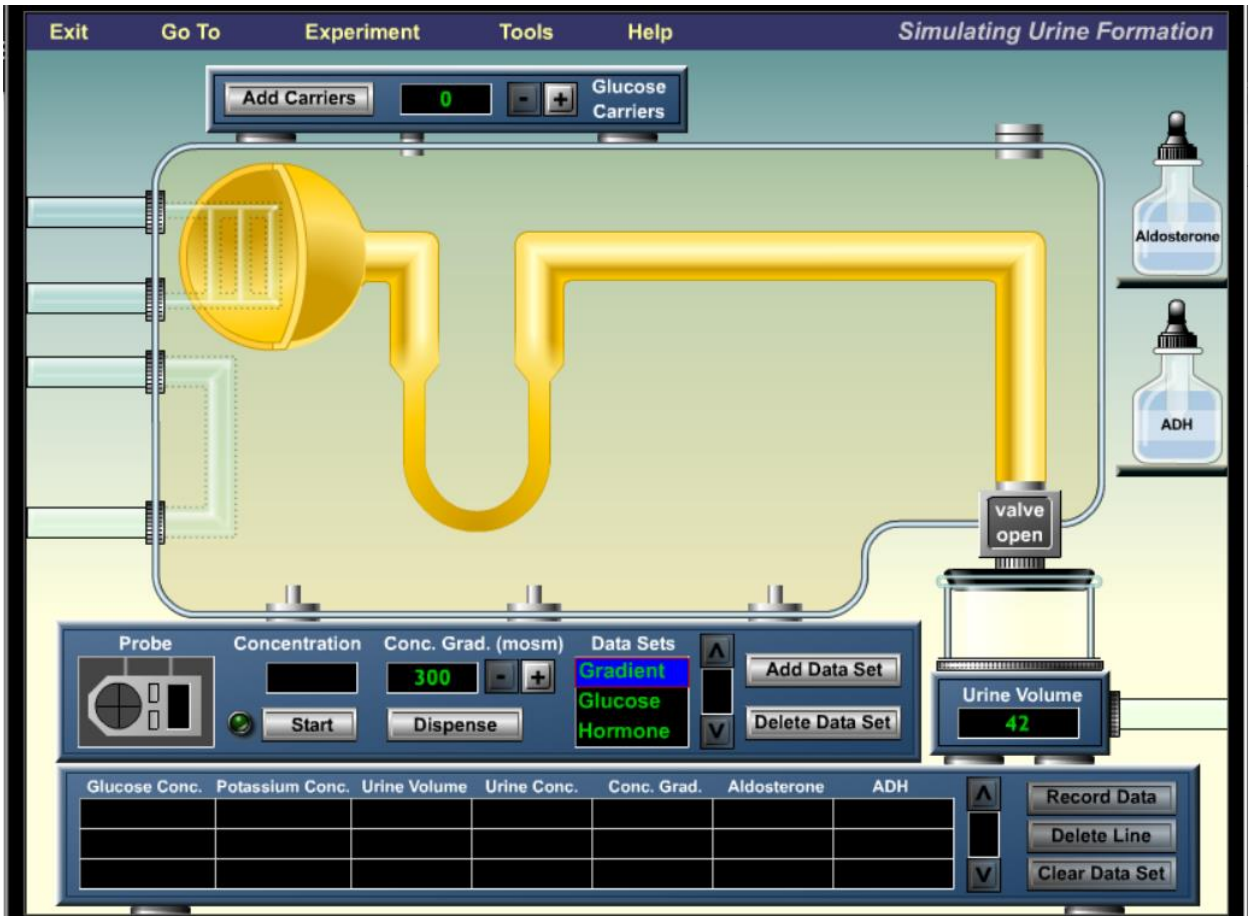

Figure 6.2. Opening screen of the simulating urine formation experiment

Hormone is dispensed by dragging a hormone dropper cap to the gray cap button in the nephron tank at the top of the collecting duct and then letting go of the mouse button.

The (+) and (–) buttons in the glucose carrier control are used to adjust the number of glucose carriers that will be inserted into the simulated proximal convoluted tubule when the "Add Carriers" button is clicked.

6.2.1. Exploring the role of the solute gradient on maximum urine concentration achievable (virtual lab)

In the process of urine formation, solutes and water move from the lumen of the nephron into the interstitial spaces. The passive movement of solutes and water from the lumen of the renal tubule into the interstitial spaces relies in part on the total solute gradient surrounding the nephron. When the nephron is permeable to solutes or water, an equilibrium will be reached between the interstitial fluid and the contents of the nephron. **Antidiuretic hormone (ADH)** increases the water permeability of the distal convoluted tubule and the collecting duct, allowing water to flow to areas of higher solute concentration, usually from the lumen of the nephron into the surrounding interstitial area. You will explore the process of passive reabsorption in this experiment.

Procedure

1. "Gradient" in the "Data Sets" window of the data control unit should be highlighted in bright blue. If it is not, then click "Gradient"
2. If the data grid is not empty, click "Clear Data Set" to discard all previous data.
3. Click and hold the mouse button on the "ADH bottle" cap and drag it to the gray cap at the top right side of the nephron tank. Release the mouse button to dispense ADH onto the collecting duct.
4. Adjust the maximum total solute concentration of the gradient (**Conc. Grad.**) to 300 mOsm by clicking the appropriate (+) or (–) button. Because the blood solute concentration is also 300 mOsm, there is no osmotic difference between the lumen of the nephron and the surrounding interstitial fluid.
5. Click "Dispense".
6. Click "Start" to begin the experiment. Filtrate will move through the nephron and then drain into the beaker below the collecting duct.
7. While the experiment is running, watch the **Probe**. When it turns red, click and hold the mouse on it, and drag it to the urine beaker. Observe the total solute concentration in the **Concentration** window.

8. Now click “Record Data” to record the current experiment data in the data grid.
9. Increase the maximum concentration of the gradient in 300 mOsm increments, and repeat steps 3 through 8 until 1200 mOsm is achieved. Be sure to click “Record Data” after each trial. If you make an error and want to delete a single value, click the data line in the data grid and then click “Delete Line”.

Results and Discussion

What happened to the urine concentration as the gradient concentration was increased? What happened to the volume of urine? Draw a graph showing how the volume of urine changes depending on concentration gradient. What factor limits the maximum possible urine concentration? Was equilibrium achieved? Explain the mechanism of the effect of ADH on water permeability in the membrane (receptor, intracellular process) and make a general conclusion based on the results.

6.2.2. Studying the effect of glucose carrier proteins on glucose reabsorption (virtual lab)

Because carrier proteins are needed to move glucose from the lumen of the nephron into the interstitial spaces, there is a limit to the amount of glucose that can be reabsorbed. When all glucose carriers are bound with the glucose they are transporting, excess glucose is eliminated with urine. In this experiment, you will examine the effect of varying the number of glucose transport proteins in the proximal convoluted tubule.

1. Click “Glucose” in the “Data Sets” window of the data control unit.
2. If the data grid is not empty, click “Clear Data Set” to discard all previous data.
3. Set the concentration gradient (**Conc. Grad.**) to 1200 mOsm.
4. Click “Dispense”.
5. Adjust the number of glucose carriers to 100 (an arbitrary figure) by clicking the appropriate (+) or (–) button.

6. Click “Add Carriers”. This action inserts the specified number of glucose carrier proteins per unit area into the membrane of the proximal convoluted tubule.
7. Click “Start” to begin the run after the carriers have been added.
8. Click “Record Data” to record the current experimental data in the data grid. Glucose presence in the urine will be displayed in the data grid.
9. Now increase the number of glucose carrier proteins in the proximal convoluted tubule in increments of 100 glucose carriers, and repeat steps 6 through 8 until the maximum number of glucose carrier proteins (500) is achieved. Remember to click “Add Carriers” each time. Be sure to click “Record Data” after each trial. If you make an error and want to delete a single value, click the data line in the data grid and then click “Delete Line”.

What happened to the amount of glucose present in the urine as the number of glucose carriers was increased? Draw the graph showing your results. Explain how glucose is transported back from the nephron to the blood. How does the glucose transporter work?

6.2.3. Testing the effect of hormones on urine formation (virtual lab)

The concentration of the urine excreted by our kidneys changes depending on our immediate needs. For example, if Renal System Physiology 127 a person consumes a large quantity of water, the excess water will be eliminated, producing dilute urine. On the other hand, under conditions of dehydration, there is a clear benefit in being able to produce urine as concentrated as possible, thereby retaining precious water. Although the medullary gradient makes it possible to excrete concentrated urine, urine dilution or concentration is ultimately under hormonal control. In this experiment, you will investigate the effects of two different hormones on renal function, aldosterone produced by the adrenal gland and ADH manufactured by the hypothalamus and stored in the posterior pituitary gland.

Procedure

1. Click “Hormone” in the “Data Sets” window of the data control unit.

2. If the data grid is not empty, click "Clear Data Set" to discard all previous data.
3. During this part of the experiment, keep the concentration gradient at 1200 mOsm.
4. Click "Dispense" to add the gradient, and then click "Start" to begin the experiment.
5. Now click "Record Data" to record the current experiment data in the data grid.

You will use this baseline data to compare with the conditions of the filtrate under the control of the two hormones.

6. Keeping all experiment conditions, the same as before, do the following:
   a. Drag the "Aldosterone" dropper cap to the gray cap on the top right side of the nephron tank, and release the mouse to automatically dispense aldosterone into the tank surrounding the distal convoluted tubule and collecting duct.
   b. Click "Start", and allow the run to complete.
   c. Click "Record Data".
7. Drag the "ADH" bottle cap to the gray cap on the top right side of the nephron tank, and release it to dispense ADH.
   a. Click "Start", and allow the run to complete.
   b. Click "Record Data".

Results and Discussion

How does the volume of urine differ from the previously measured baseline volume? Explain the difference in the total amount of potassium in the urine between this run and the baseline run. How does the volume of urine differ from the baseline measurement? Is there a difference in the total amount of potassium in this run and the total amount of potassium in the baseline run? Explain your answer. (Hint: The urine volume with ADH present is about one-tenth the urine volume when it is not present.) Are the effects of aldosterone and ADH similar or antagonistic? Explain difference in mechanism of ADH and aldosterone's effects on urine formation process.

6.3. The effect of hydrostatic pressure, osmotic pressure and diameters of the glomerular afferent and efferent arterioles on urine flow (virtual lab)

Open file `LUPRAFISIM.exe > START > English version > CONTENTS > KIDNEY`. Do virtual works, following instructions in them.

Urine is formed in the kidney as a result of three processes: glomerular filtration, tubular reabsorption and tubular secretion.

Glomerular filtration consists of a transfer of water and low molecular weight substances from plasma that flows through the glomerular capillaries into glomerular capsules. This process is influenced by the total surface of the glomerular filtrating membrane, the **net filtration pressure** and the **glomerular filtration rate**.

The net filtration pressure (P_f) results from the **hydrostatic glomerular blood pressure** (P_b=70mmHg)**, oncotic blood pressure** (P_o=25mmHg) and **intracapsular pressure** (P_i=5mmHg), according to the following formula:

$$P_f = P_b - (P_o + P_i)$$

The glomerular filtration rate varies with the vasodilatation or the vasoconstriction of the afferent and efferent glomerular arterioles.

Aim: Demonstration of the influence of the glomerular filtration rate, the hydrostatic blood pressure and oncotic blood pressure on urine flow.

Technique

1. Click the `“START”` button and wait until the determination is made;
2. Record the value of the urine flow;
3. The value of the diameter of the glomerular afferent arteriole is increased decreased and actions from points 1 and 2 are repeated;
4. The value of the diameter of the glomerular efferent arteriole is decreased and actions from points 1 and 2 are repeated;
5. The value of the diameter of both afferent and efferent arterioles is decreased and actions from points 1 and 2 are repeated;
6. The value of the blood pressure is increased and then decreased and actions from points 1 and 2 are repeated;
7. The value of the oncotic blood pressure is increased and then decreased and actions from points 1 and 2 are repeated.

Results and Discussion

1. Write down the results into Table 6.1.
2. Explain your results, make conclusions based on them.
3. Draw a nephron and explain processes of filtration, reabsorption and secretion
4. Write down the formula of the net filtration pressure, explain it.

Table 6.1. Changing of urine flow

Conditions	**The diameter** (dilated/constricted)		**Oncotic pressure, mmHg**	**Blood pressure, m Hg**	**Intensity of diuresis, ml/min**
	Afferent arterioles	Efferent arterioles			
Control					
Decreased diameter of the glomerular afferent arteriole					
Decreased diameter of the glomerular efferent arteriole					
Both arterioles' diameters are decreased					
Increased blood pressure					
Decreased blood pressure					
Increased oncotic pressure					
Decreased oncotic pressure					

6.4. Influence of the aldosterone and the antidiuretic hormone on the urine flow (virtual lab)

Aldosterone (a mineralocorticoid hormone) is synthesized in the zona glomerulosa of the adrenal cortex.

The release of aldosterone into the blood flow is controlled by the **renin-angiotensin-aldosterone system (RAAS)**. Lowering of the blood pressure from the glomerular arterioles determines a release of the proteolytic enzyme **renin** from the kidneys. Renin converts the plasmatic **angiotensinogen** into **angiotensin I**, which is further converted into **angiotensin II** through the action of **angiotensin-converting enzyme**. Angiotensin II stimulates the synthesis and release of aldosterone by the adrenal cortex. The main actions of aldosterone are: (1) retention of Na^+, Cl^-, bicarbonic ions and water; (2) reduction of the urine flow; (3) increase of the blood pressure.

The **antidiuretic hormone (ADH or vasopressin)** is a neurohypophyseal hormone which is synthesised in the hypotalamus and is stored in the *pars nervosa* of the hypophysis. ADH is then released into the bloodstream when: (1) the hypothalamic osmoreceptors perceive a **lowering of the blood osmotic pressure**; (2) the baroreceptors from the aorta and the carotid artery perceive a **lowering of the blood pressure**.

The main actions of ADH are: (1) retention of water; (2) reduction of the urine flow; (3) increase in blood pressure.

Aim: Demonstration of the effect of aldosterone and ADH on urine flow. Determination of the urine flow before and after aldosterone administration and then before and after ADH administration.

Technique

1. Click the "START" button and wait until the determination is made;
2. Record the value of the urine flow;
3. Give aldosterone by clicking on the arrow and repeating actions from points 1 and 2.
4. Give ADH by clicking on the arrow and repeat actions from points 1 and 2.

Results and Discussion

1. Write down the results into Table 6.2.
2. Explain your results, make conclusions based on them.
3. Describe regulation of aldosterone and ADH synthesis, explain renin-aldosterone-angiotensin system (in the form of scheme).
4. Draw the scheme, showing cellular mechanisms of aldosterone and ADH effects on the cells of the distal and collecting tubules of nephron.
5. What is overall effect of ADH and aldosterone on the diuresis and blood pressure?

Table 6.2. Influence of hormones on the urine flow

Conditions	Intensity of diuresis, ml/min
Control	
Aldosterone	
ADH	

6.5. Influence of glucose on urine flow (virtual lab)

An increase in the level of blood glucose, which is characteristic of diabetes mellitus, influences the urine flow in the following way:

– the renal threshold for glucose is surpassed and glucose is found in the urine;
– the urine flow is enhanced.

Aim: Demonstration of the enhancement of urine flow and of glucose level in the urine as a result of an increase in blood glucose. Urine flow and glucose level in the urine are estimated before and after the intravenous administration of a concentrated glucose solution.

Technique

1. Click the "START" button and wait until the determination is made;
2. Record the value of the urine flow;
3. Click the "COLLECT A SAMPLE" button;
4. Add NaOH in the test tube;
5. Add $CuSO_4$ in the test tube;
6. Click the "WARM THE SAMPLE" button and wait until the determination is made;
7. Give glucose by clicking the arrow and repeat actions from points 1 – 6.

Results and Discussion

1. Write down the results into Table 6.3.
2. Explain your results, make conclusions based on them. How does the presence of glucose affect the amount of final urine?
3. Describe what is happening with glucose during the processes of filtration, reabsorption and secretion processes.
4. Draw the scheme, showing cellular transporters for glucose in the kidney cells. What kind of transport is that?
5. What can be the reason for the presence of glucose in the final urine?

Table 6.3. Influence of glucose on urine flow

Conditions	**Trommer reaction** (positive or negative)	**Intensity of Diuresis, ml/min**
Control		
Glucose		

Lab 7. ENDOCRINE SYSTEM

Open file `LUPRAFISIM.exe > START > English version > CONTENTS > ENDOCRINE SYSTEM`. Do virtual works, following instructions in them.

7.1. The effect of thyroxine, TSH and propylthiouracil on metabolism (virtual lab)

Metabolism consists of all the exchanges of matter and energy between the organism and the environment. Its value depends on the following:

- species (the larger the animal, the lower the value of metabolism);
- sex (the metabolism of a male is more intense than that of a female);
- age (the older the animal, the lower the value of metabolism).

The main hormones, which are responsible for the regulation of metabolism, are the **thyroid hormones** (thyroxine and triiodothyronine). They are synthesized by the follicular cells of the **thyroid gland** and **TSH** (thyroid stimulating hormone), which is synthesized by the anterior pituitary gland, enhances their secretion. **Propylthiouracil** is a substance, which inhibits the synthesis of thyroid hormones.

The intensity of exchanges between organism and environment may be estimated by measuring the heat, which comes out of the organism into the environment **(calorimetry method)**.

Direct calorimetry consists of measuring the heat, which comes out of the organism into the environment per time unit. For this purpose, a complex experimental device is necessary.

Indirect calorimetry offers the possibility to evaluate metabolism by means of simpler methods, which do not use complex experimental devices. Such methods are the **method of nutritive balance** and the **method of respiratory exchanges**.

The method of respiratory exchanges relies on the principle that the intensity of metabolism is proportional with the amount of oxygen, which is consumed by the organism in a time unit:

$$\text{Metabolic rate} = \text{ml of consumed } O_2/\text{kg/hour}$$

Objective

Demonstration of the influence of thyroxine, TSH and propylthiouracil on metabolism at three different rats. One rat is normal, the second had the thyroid gland extirpated and the third had the pituitary gland extirpated.

Principle

Metabolism of the three rats is measured before and after administering them thyroxine, TSH and propylthiouracil.

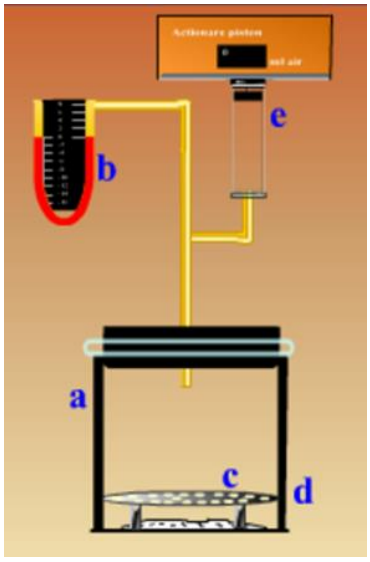

Figure 7.1.

The experimental device is made up of (Fig. 7.1):

- respiratory room (**a**) with a closed circuit, which has:
- a simple manometer (**b**) – an U-shaped tube with liquid inside;
- a grill (**c**);
- sodium limestone (**d**) (a substance which absorbs carbon dioxide from the respiratory room).
- device for introducing air in the respiratory room (**e**).

Technique

1. Introduce the normal rat in the respiratory room.
2. Click the "START" button.
3. Wait for 60 seconds and notice how the level of the liquid decreases in the left side of the manometer, as the oxygen from the respiratory room is consumed (in the same time, the carbon dioxide which is produced by the rat is absorbed by the sodium limestone).
4. After 60 seconds, click the button in order to introduce air in the respiratory room, until the level of the liquid is the same in the two sides of the manometer.
5. Determine the metabolic rate, using the following formula:

$$\text{Metabolic rate} = \frac{\text{ml}O_2 \times 60 \times 1000}{\text{rat body weight in g}}, \text{ml}O_2/\text{kg/hour}$$

6. Repeat actions from points 1, 2, 3, 4 and 5 after administering to the rat: thyroxine, TSH and propylthiouracil.

The mentioned techniques are applied on "normal", "thyroidectomized" and "hypophysectomized" rats. Put your calculations into the table 7.1.

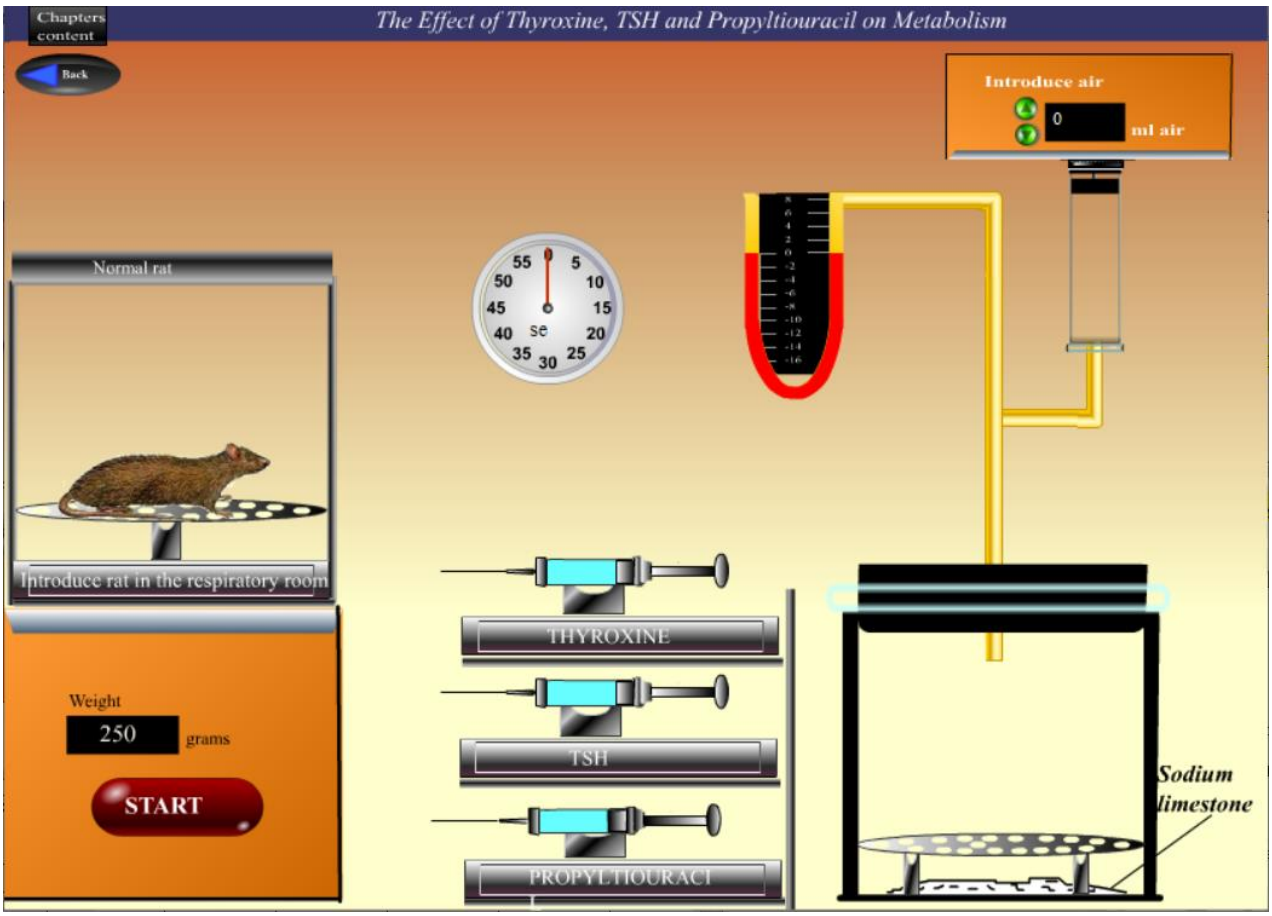

Figure 7.2. Screenshot of the lab with a "normal" rat

Table 7.1. Amount of consumed oxygen

Types of rats	Rat's weight, g	Amount of consumed oxygen, ml			
		Control	Thyroxine	TSH	PTU
Normal					
Thyroidectomized					
Hypophysectomized					

Determine the metabolic rate, using the following formula and put your calculations into the table 7.2.

$$\text{Metabolic rate} = \frac{\text{ml}O_2 \times 60 \times 1000}{\text{rat body weight in g}}, \text{ml}O_2/\text{kg/hour}$$

Table 7.2. Results of metabolic rate

Types of rats	Metabolic rate, mlO_2/kg/hour			
	Control	Thyroxine	TSH	PTU
Normal				
Thyroidectomized				
Hypophysectomized				

Results and Discussion

1. Make conclusions about the effect of thyroxine, TSH and propylthiouracil on metabolism of different kinds of rats.
2. Draw the scheme of the hypothalamic-pituitary-thyroid axis with feedback mechanisms of their synthesis.
3. Draw the scheme, showing cellular mechanisms of thyroid hormones' effect on the target cell (types of receptors, metabolic changes).
4. Describe general metabolic changes and effects of thyroid hormones in different tissues of the body.

7.2. The effects of insulin and alloxan on blood glucose (virtual lab)

Insulin is a polypeptidic hormone, which is synthesized by the cells from the Langerhans islets of the pancreas.

The main metabolic action of this hormone is to lower the level of the glucose in the whole blood, by increasing the transfer of glucose across the plasmatic membrane of target cells, where it enhances:

– glycolysis;
– inclusion of glucose in the glycogen molecule (in the hepatic and muscular tissues);
– transformation of glucose in lipids and proteins.

Diabetes mellitus is a metabolic disorder which may be classified into types I and II:

– insulin-dependent diabetes mellitus (type I), which is caused by the insufficient synthesis of insulin in the endocrine pancreas;
– non-insulin-dependent diabetes mellitus (type II), which is characterized by a sufficient synthesis of insulin, without a suitable response from the target cells.

Objective

Demonstration of the effect of insulin on the healthy rat and on the rat with insulin-dependent diabetes mellitus (produced by administering alloxan, a substance that destroys the cells from the Langerhans islets of the pancreas).

Principle

Following step-by-step instructions in the program, analyze glucose levels in the blood of a normal rat before and after insulin administration, and then repeat the same procedure with the rat with diabetes mellitus (after administration of alloxan). Put your results into the table 7.3.

Table 7.3. Insulin-dependent and independent glucose levels

Type of rat	**Glucose level, mg/dL**	
	Before insulin administration	After insulin administration
Normal		
Diabetic (injected with alloxan)		

Results and Discussion

Make conclusions about the effect of insulin and alloxan on blood glucose level of rats.

1. Describe regulation of insulin synthesis
2. Draw the scheme, showing cellular mechanisms of insulin effect on the target cell (types of receptors for glucose transport, receptors for insulin, its effects on glucose metabolism).
3. Describe general metabolic changes and effects of insulin in different tissues of the body.
4. Compare different types of Diabetes mellitus and its effect on the body.

Supplement 1. Harris-Benedict table of counting basal metabolism for male (1 kcal = 4.19 joules)

A						**B**										
Weight, kg	Calories, kcal	Weight, kg	Calories, kcal	Weight, kg	Calories, kcal	Height cm	15	17	19	21	23	25	27	30	35	40
40	617	61	905	82	1194	124	420	393	368							
41	630	62	919	83	1208	128	460	433	408							
42	644	63	933	84	1222	132	500	473	448							
43	658	64	947	85	1235	136	540	513	486							
44	672	65	960	86	1249	140	580	553	528							
45	685	66	974	87	1263	144	620	593	568							
46	699	67	988	88	1277	148	660	633	608							
47	713	68	1002	89	1290	152	700	673	648	619	605	592	578			
48	727	69	1015	90	1304	156	713	713	678	669	625	612	598			
49	740	70	1029	91	1318	160	780	743	708	659	645	631	618	598	564	530
50	754	71	1043	92	1332	164	810	773	738	679	665	652	638	623	589	555
51	768	72	1057	93	1345	168	840	803	768	699	685	672	658	648	614	580
52	782	73	1070	94	1345	172	860	823	788	719	705	692	678			
53	782	74	1084	95	1373	176	880	843	808	729	725	718	698	673	639	605
54	809	75	1098	96	1387	180	900	863	828	759	745	732	718	664	664	630
55	823	76	1112	97	1406	184	920	883	846	779	765	752	738			
56	837	77	1125	98	1414	188	940	903	868	799	785	772	752			
57	850	78	1139	99	1428	192	923	888	819	805	792	776				
58	864	79	1153	100	1442	196	908	839	825	812	798					
59	878	80	1167													
60	892	81	1180													

Supplement 2. Harris-Benedict table of counting basal metabolism for female (1 kcal = 4.19 joules)

A						B										
Weight, kg	Calories, kcal	Weight, kg	Calories, kcal	Weight, kg	Calories, kcal	Height cm	15	17	19	21	23	25	27	30	35	40
40	1038	61	1238	82	1439	124	101	101	82							
41	1047	62	1248	83	1449	128	117	107	98							
42	1057	63	1258	84	1458	132	133	123	114							
43	1066	64	1267	85	1468	136	140	139	130							
44	1076	65	1277	86	1478	140	165	165	146							
45	1085	66	1286	87	1487	144	181	171	162							
46	1095	67	1296	88	1497	148	197	187	178							
47	1 105	68	1305	89	1506	152	212	201	192	183	174	164	155	138	113	90
48	1 114	69	1315	90	1516	156	227	215	206	190	181	172				
49	1 124	70	1325	91	1525	160	242	229	220	198	188	179	170	155	132	109
50	1 133	71	1334	92	1535	164	257	243	234	205	196	186	177	164	142	119
51	1 143	72	1 344	93	1544	168	271	255	246	213	203	194	184	169	146	124
52	1 152	73	1353	94	1554	172	285	264	258	220	211	201	192	175	151	128
53	1 162	74	1363	95	1564	176	299	279	270	227	218	209	199	184	160	137
54	1 172	75	1372	96	1573	180	313	291	282	235	225	216	207	193	169	146
55	1 181	76	1382	97	1583	184	327	303	294	242	233	223	214			
56	1 191	77	1391	98	1592	188	322	304	250	240	231	221				
57	1200	78	1401	99	1602	192	333	314	257	248	238	229				
58	1210	79	1411	100	1661	196	334	324	264	255	246	236				
59	1219	80	1420													
60	1229	81	1430													

Supplement 3. Adult's energy expenditure of the different physical activities

Activities	Value*	Activities	Value	Activities	Value
Aerobic	7.3	Automobile repair	3.3	Basketball	7.5
Ballet	6.3	Butchering	3.0	Boxing	12.3
Ballroom	11.3	Chess game	1.5	Eating	1.5
Bicycling	7.0	Child care	3.5	Football	8.0
Calisthenics	7.5	Cooking	3.5	Guitar	2.5
Circuit training	6.0	Driving	2.8	Hairstyling	2.5
Cleaning	3.3	Food shopping	3.3	Hockey	8.0
Fishing	3.5	Hammering nails	3.0	Patient care	2.3
Flamenco	8.5	Handwashing	1.3	Piano	2.3
Folk	9.8	Home repair	6.0	Skateboarding	5.0
Home exercise	3.8	Knitting	1.3	Skating	14.0
Hunting	2.5	Laundry	2.3	Skiing	6.0
Jumping rope	9.0	Lying	1.0	Skipping	11.5
Kitchen activity	3.3	Moving furniture,	5.8	Swimming, butterfly	13.8
Mopping	2.5	Painting	5.0	Swimming, craw	8.0
Pilates	2.8	Planting	4.3	Swimming, freestyle	9.8
Pole dancing	4.5	Scrubbing floors	3.5	Tennis	6.8
Polishing floors	4.5	Sitting	2.3	Triathlon	13.0
Salsa	6.0	Sleeping	1.0	Typing	1.3
Stretching	2.3	Standing	2.0	Video game	2.3
Virtual reality	7.9	Vacuuming	3.0	Violin	2.5
Wash dishes	2.0	Walking	3.5	Volleyball	6.0
Yoga	2.3	Watering plants	2.0	Water polo	10.0
Zumba	6.5	Writing	1.8	Wrestling	6.0

*Value of energy expenditure due to 1 kg of the body weight, kcal/hour

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Авторы
Митрухина О.Б., Силантьева Д.И.,
Шайдуллов И.Ф., Ананьев А.С., Свитко С.О.,
Ситдикова Г.Ф.

Lab manual of VISCERAL PHYSIOLOGY

Учебно-методическое пособие
на английском языке

Department
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