# LABORATORY MANUAL FOR NORMAL PHYSIOLOGY

for specialty “Dentistry”

In two parts

Part 1

Student _____ group _____

Lecturer _____

Minsk BSMU 2019

МИНИСТЕРСТВО ЗДРАВООХРАНЕНИЯ РЕСПУБЛИКИ БЕЛАРУСЬ
БЕЛОРУССКИЙ ГОСУДАРСТВЕННЫЙ МЕДИЦИНСКИЙ УНИВЕРСИТЕТ
КАФЕДРА НОРМАЛЬНОЙ ФИЗИОЛОГИИ

# РУКОВОДСТВО К ЛАБОРАТОРНЫМ ЗАНЯТИЯМ ПО НОРМАЛЬНОЙ ФИЗИОЛОГИИ

# LABORATORY MANUAL FOR NORMAL PHYSIOLOGY

Практикум для специальности «Стоматология»

В двух частях

Часть 1

Под редакцией О. С. Никитиной, В. А. Переверзева

Минск БГМУ 2019

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ББК 28.707я73
Р84

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Р е ц е н з е н т ы: канд. мед. наук, доц. Я. Н. Борисевич; канд. мед. наук, доц. А. Г. Кадушкин

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Р84

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Представлены вопросы к лабораторным занятиям и к итоговым семинарам по всем разделам курса нормальной физиологии; описания лабораторных работ и протоколы их выполнения; необходимая дополнительная информация по темам занятий. Приведены задания для организации самостоятельной работы студентов.

Предназначен для студентов 1–2-го курсов медицинского факультета иностранных учащихся, обучающихся на английском языке по специальности «Стоматология».

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**ISBN 978-985-21-0228-5**

**Navigation**: http://etest.bsmu.by/ → For Students with Training in English → Dentistry → Normal Physiology;
www.bsmu.by (switch to "eng") → Educational materials → Normal Physiology.

| | Topics | Passed |
|---|---|---|
| 1. | Introduction. Physiology as a scientific basis of medicine. The significance of human physiology for dentists | |
| 2. | The concept of chemical and electrical signaling. Receptors, their types. Excitable tissues and their general properties. Biological potentials. Electroodontodiagnostics | |
| 3. | Conduction of excitation by nerve fibers and synapses. Physiological basis of conductive anesthesia in dental practice | |
| 4. | Physiology of skeletal muscles | |
| 5. | Physiology of muscles of maxillofacial region. Physiology of smooth muscles. Notion of the myoepithelial and glandular cells | |
| 6. | Physiology of the nervous system. Excitation and inhibition in CNS. Reflexes. General principles of CNS activity coordination | |
| 7. | **Colloquium "Excitable tissues"** | |
| 8. | Nervous regulation of somatic functions | |
| 9. | Nervous regulation of autonomic functions (physiology of autonomic nervous system) | |
| 10. | Humoral regulation of functions. Physiology of the endocrine system. Lesson № 1 | |
| 11. | Humoral regulation of functions. Physiology of the endocrine system. Lesson № 2 | |
| 12. | Regulation of calcium and phosphorus in the body, in the bone tissue and in the teeth | |
| 13. | **Colloquium "Mechanisms of regulation of functions"** | |
| 14. | Body fluids (blood, lymph, cerebrospinal fluid, saliva, etc.) | |
| 15. | Blood cells. Erythrocyte sedimentation rate. The common clinical blood analysis. Haematopoiesis | |
| 16. | Blood group systems. Components of the blood. Blood substituting solutions. Hemostasis | |
| 17. | **Colloquium "Body fluids"** | |
| 18. | **Credit Test** | |

| Short list of lectures | Passed |
|---|---|
| 1. Excitable tissues | |
| 2. Skeletal muscles | |
| 3. Central nervous system | |
| 4. Autonomic nervous system | |
| 5. Endocrine system | |
| 6. Blood. Blood cells. Hematopoiesis. | |
| 7. Blood groups. Hemostasis. | |
| 8. Heart physiology. | |

**INTRODUCTION**:
2nd semester of the 1st year.
Lectures — 16 hours (8 lectures),
Lab classes — 54 hours (18 lessons),
Colloquiums (the concluding lessons) — 3 lessons (№ 7, 13 and 17).

**Admission to Credit Test**:
– without missed lectures and practical classes;
– all lectures are written;
– completed and signed lab manual.

*"The right and wrong answers should come from your heart"*

## Lesson 1. INTRODUCTION. PHYSIOLOGY AS A SCIENTIFIC BASIS OF MEDICINE. THE SIGNIFICANCE OF HUMAN PHYSIOLOGY FOR DENTISTS

DATE OF CLASSES
«____» _____ 20____
day month year

| **BASIC QUESTIONS:** | LITERATURE |
|---|---|
| 1. Physiology as a scientific basis of medicine. The significance of human physiology for dentists.<br>2. Stages of development of Physiology (a brief history). The contribution of Belarusian / Russian scientists in the development of Physiology.<br>3. The concept of physiological research methods. Safety rules in performing physiological studies.<br>4. The cell as a structural and functional basis of a living organism, its main features and functions.<br>5. Modern concept of the structure and functions of the cell membranes. Mechanisms of transport of a substances across the cell membrane.<br>6. Ion channels in cell membranes: sodium, potassium, calcium, chloride and water channels. Principles of their classification. | Main<br>1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 7–8.<br>Additional<br>3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 33–36, 41–42, 45–52.<br>4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 3–14, 47–58. |

### WORK 1.1. STAGES OF DEVELOPMENT OF PHYSIOLOGY (A BRIEF HISTORY). THE CONTRIBUTION OF BELARUSIAN/RUSSIAN SCIENTISTS IN THE DEVELOPMENT OF PHYSIOLOGY

The Nobel Prize in Physiology or Medicine 1904
Ivan Pavlov

Official date of the physiology originating is _____ year.
William Harvey (1578–1657) _____
_____
Stage 1 _____
_____
Stage 2 _____
_____
I. P. Pavlov (1849–1936) _____
_____
Stage 3 _____
_____
The significance of Human Physiology for dentists: _____

## WORK 1.2. SAFETY RULES WHEN PERFORMING PHYSIOLOGICAL STUDIES.

The teaching program at the Department of Normal Physiology envisages practical works performed by the students, mastering their practical skills of operating some electric devices, computer techniques, research equipment, laboratory dishes, chemical reagents and biological fluids.

In addition, students may be allowed to do research work in the laboratories of the Department during their out-of-classes hours.

**General requirements.**

1. The student should put on a lab coat (medical gown) before entering an academic room.
2. To assign the student on duty.

**A student on duty should:**

– observe the order, rules and requirements of safety provisions while working in practical rooms;

– receive the practical rooms key and various materials necessary for carrying out practical works — in the laborant's room **№ 131**;

– at the end of practical classes — switch off the water and lights and return the received materials into room **№ 131**.

**Safety rules in operating electrical equipment.**

Cases of electric trauma and fires may occur while working with electric equipment. They may be caused by:

– working with defective electric equipment (knife-switches, sockets, etc.);
– absence of electric appliances grounding;
– breaking rule of operating electric devices;
– touching current-carrying elements with hands and metal objects.

In case of revealing a defect of the electric device or electric equipment it is necessary to inform the teacher about it.

While operating the electric equipment and electric devices it is strictly forbidden to:

– check the presence of electric voltage with fingers and touch current-carrying parts;
– operate ungrounded electric equipment and devices if not allowed by the device instruction;
– use defected electric equipment and electric wiring;
– leave an electric circuit under tension without supervision.

**General rules of giving the first aid.**

The first aid to victims should be given immediately and properly. It may affect the life, consequences of injuries, burns and poisonings. You'll get acquainted with specific rules of rendering it at clinical departments.

In case of serious injuries, burns due to electric trauma an ambulance should be called in (telephone number 103). If the injuries are mild, the victims should be given the first aid and directed to a medical care institution. It should be kept in mind that rendering aid to a person under electric current you shouldn't touch him with bare hands. First of all, the setting (device), which the victim touches, should be switched off or you should separate the victim from current-carrying parts using sticks, boards and other dry objects not conducting electric current or cut off wires by an axe with a dry axe handle.

*In all cases, you must call the duty laboratory assistant, who is in the room **№ 131**, or a lecturer of the Department.*

**Actions taken in case of fire.**

In case of fire one should immediately switch off the power, call in the assistance (**room 131**) or lecturer and start extinguishing the fire. There are fire extinguishers in rooms **104**, **131**, **135** and **138**. For extinguishing the fire one can also use available fire hoses: unreel the hose and open the hydrant. The fire hydrants with hoses are at the end of the corridor next to room **136**, in the niche between rooms **139** and **140**, **133** and **132**, and opposite room **104**.

**Directions for recording the Protocol:**

After the completion of safety rules studying it is necessary to put your name and signature in the "Safety Register for students" in the computer class, room 104.

**PROTOCOL**

I have read and have been instructed by safety rules:

| _____ | _____ | _____ |
|---|---|---|
| Date | Student's signature | Student's name (completely and legibly) |

**WORK 1.3. LEARNING METHODS OF WORKING IN THE COMPUTER CLASS (№ 104)**

| | |
|---|---|
| ***A.* Working in computer library** | Desktop → Link "Дистанционное обучение" → Log in (use data from student's card) → For Students with Training in English → Dentistry → Normal Physiology → Enroll → "Lectures and Books" and "Lesson 1" (or Link "Кафедра нормальной физиологии" → For English Students → Lesson 1).<br>Use the materials of "Lecture 1. Introduction" (or "Lesson 1) to fill in a table at work 1.1 |
| ***B.* Performing a computer tests** | Steps are on figures: |

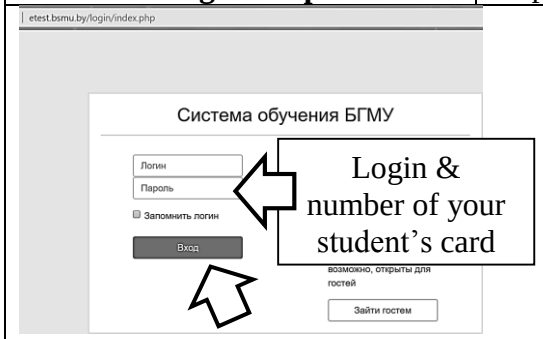



Fig. 1.1

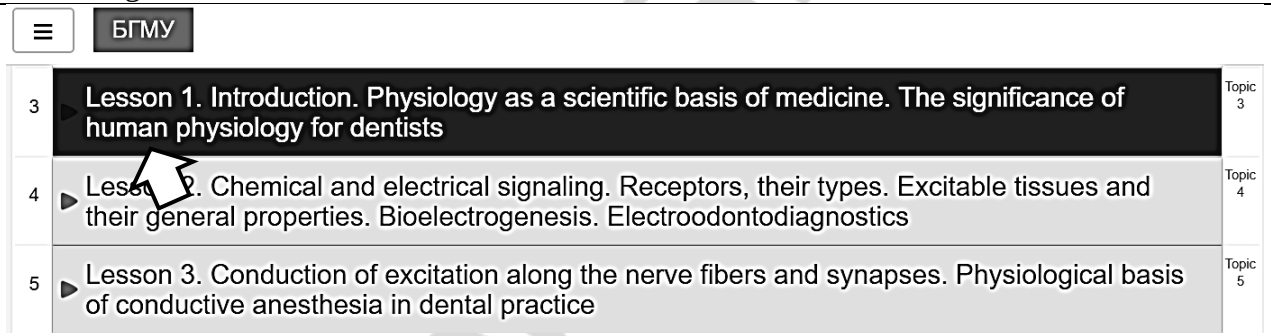



Fig. 1.2

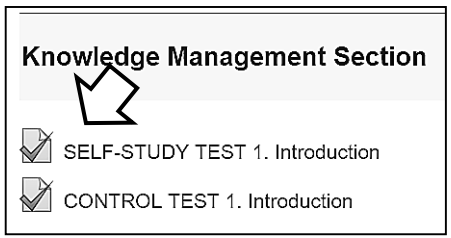



Fig. 1.3

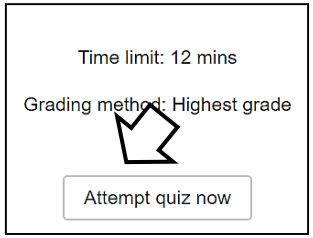



Fig. 1.4

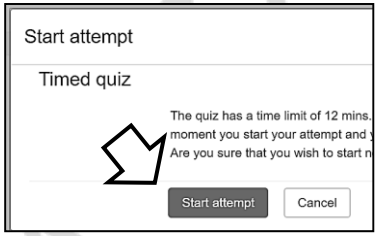



Fig. 1.5


Fig. 1.6

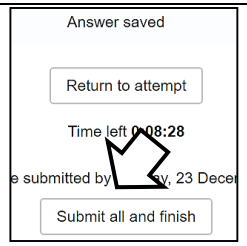



Fig 1.7

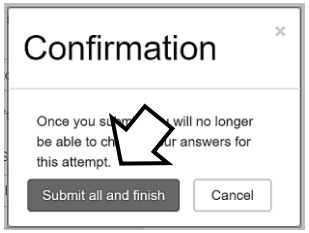



Fig 1.8

| | |
|---|---|
| **Started on** | Sunday, 23 December 2018, 8:08 PM |
| **State** | Finished |
| **Completed on** | Sunday, 23 December 2018, 8:15 PM |
| **Time taken** | 7 mins 9 secs |
| **Grade** | **7** out of 10 (**73%**) |

Grade / 10.0
7.3

Fig. 1.9

| % | Grade |
|---|---|
| 91-100 | 7 points |
| 81-90 | 6 points |
| 76-80 | 5 points |
| 70-75 | 4 points |
| 50-69 | 3 points |
| 30-49 | 2 points |
| 0-29 | 1 point |

**Grades 7 and up student has after the teacher's control**

**PROTOCOL**

The testing result is:

_____ %, mark is _____

## WORK 1.4. STUDYING THE EFFECT OF ADRENALINE ON THE HEART RATE


Fig. 1.10

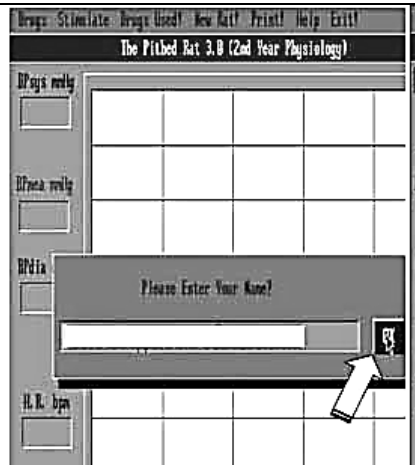



Fig. 1.11

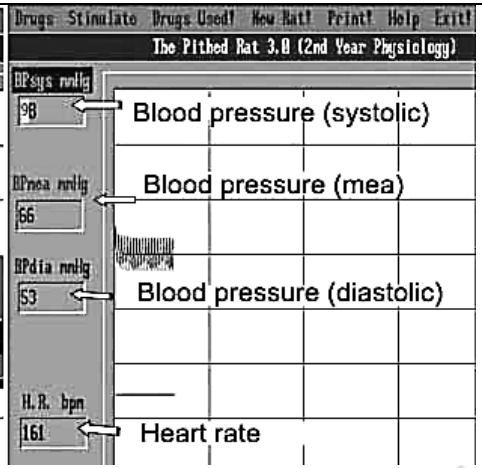



Fig. 1.12

Use the program "**PHYSIOL 2**". Next steps are on figures:

**PROTOCOL**

| Effects | | Dosage | HR, bpm |
|---|---|---|---|
| Rat 1 | Initial value | – | |
| | Inject. of Adrenaline | 5 μg/kg | |
| Click on "New rat" button | | | |
| Rat 2 | Initial value | – | |
| | Inject. of Propanalol | 100 mg/kg | |
| | Inject. of Adrenaline | 5 μg/kg | |

Conclusion: _____

_____

_____

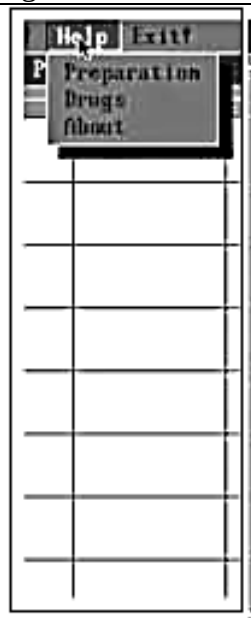



Fig. 1.13

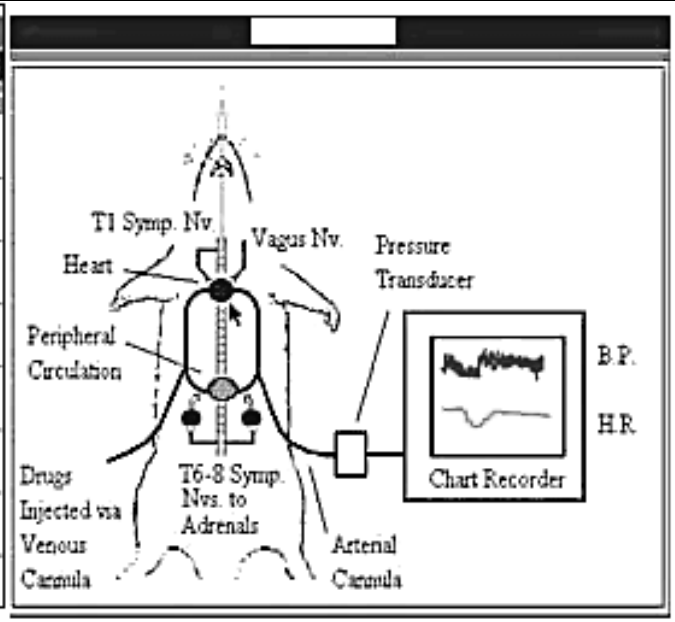



Fig. 1.14

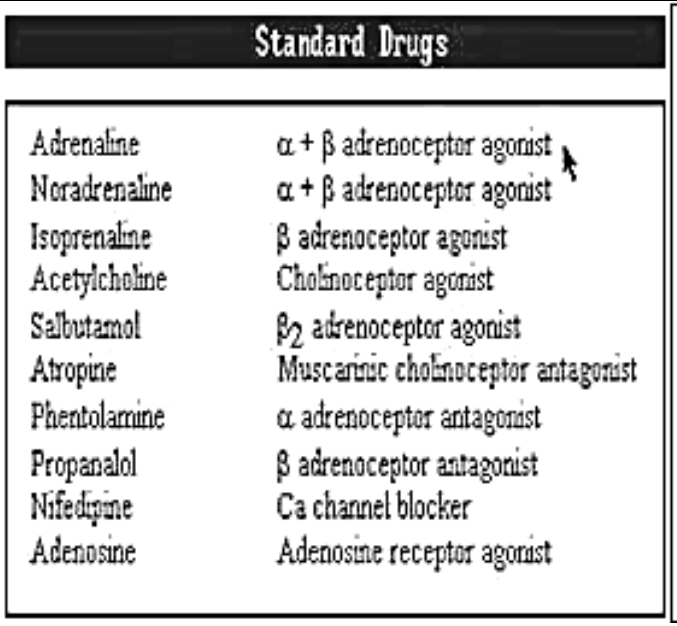



Fig. 1.15

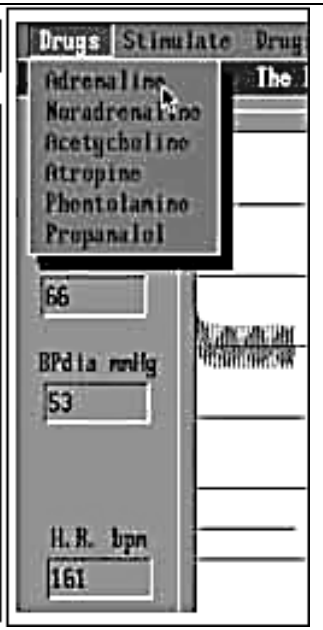



Fig. 1.16

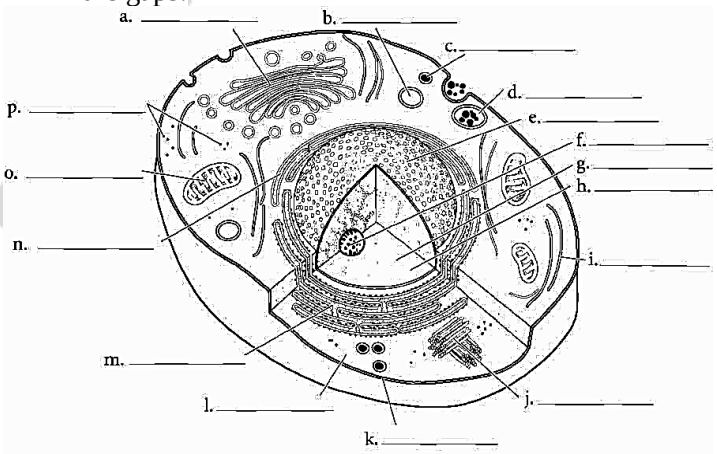
## WORK 1.5. THE CELL AS A STRUCTURAL AND FUNCTIONAL BASIS OF A LIVING ORGANISM, ITS MAIN FEATURES AND FUNCTIONS

Cells consist of an enclosing **plasma membrane**, an inner **cytoplasm** with numerous **organelles,** and other cellular structures. The fluid portion of the cell is called the **cytosol.** *Color the cytosol in last after you color the rest of the cellular structures*. One of the major structures in the cell is the **nucleus.** It is the genetic center of the cell and consists of fluid **karyoplasm, chromatin** (containing **DNA**), and the **nucleolus**. Color these features and label them on the illustration.

**The cytoskeleton** consists of microtubules, intermediate filaments and microfilaments. It is involved in maintaining cell shape, fixing organelles, and directing some cellular activity. Label the organelles of the cell and use a different color for each one. The **mitochondria** are the energy-producing structures of the cell while the **Golgi apparatus** assembles complex biomolecules and transports them out of the cell.

Proteins are made in the cell by ribosomes. If the ribosomes are found by themselves in the cytoplasm, they are called **free ribosomes**. If they are attached to the **rough endoplasmic reticulum,** they are called **bound ribosomes**. The **smooth endoplasmic reticulum** manufactures lipids and helps in breaking down toxic materials in the cell. Other structures in the cell are **vesicles** (sacs that hold liquids). **Phagocytic vesicles** ingest material into the cell. **Lysosomes** contain digestive enzymes while **peroxisomes** degrade hydrogen peroxide in the cell. *After you label and color the organelles make sure to go back and shade in the cytosol*. **Centrioles** are microtubules grouped together and arc involved in cell division.

Fill in the gaps:




The stucture of the eucariotic cell

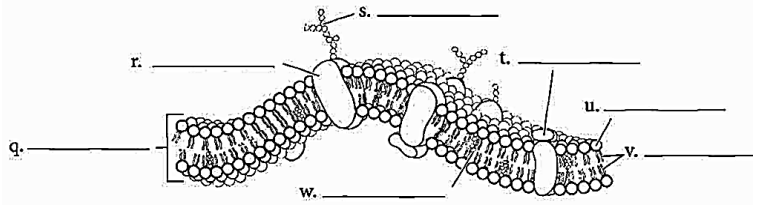
## WORK 1.6. MODERN CONCEPT OF THE STRUCTURE AND FUNCTIONS OF THE CELL MEMBRANES. MECHANISMS OF TRANSPORT OF A SUBSTANCES

The cell membrane is a lipid bilayer, consisting mainly of phospholipids. Dynamic properties are due to the protein component, which includes pumps, channels, receptors, and carriers. **Simple diffusion** and **facilitated transport** are both passive processes (independent of energy) due to concentration gradients.

The rate of protein-mediated transport will increase with increased substrate delivery until the carriers are saturated. The maximum rate (carrier saturation) is called TM and this rate is directly proportional to the number of functioning carriers presented in the system. **Secondary active transport** is driven by the sodium gradient across the cell membrane, which is maintained by the Na/K-ATPase pump (**active transport**).

**Endocytosis** and **exocytosis** are the active uptake and extrusion of macromolecules via vesicular transport.

Fill in the gaps:




The structure of the cytoplasmic membrane

The **plasma membrane** is composed of a **phospholipid bilayer. Cholesterol molecules** occur in the membrane and, depending on their concentration, can make the membrane stiff or more fluid.

*Color the **phosphate molecules** on the outside and inside of the membrane one color and the **lipid layer** another color.*

Proteins that are located on the outside of the membrane are called **peripheral proteins** while proteins that pass through the membrane are called **integral proteins**. Often they make up gates or channels that allow substances to pass through the membrane. **Carbohydrate chains** are attached to proteins on the cell membrane. These provide cellular identity.

***Label and color the cell membrane structures***.

Answer Key: a. Golgi apparatus, b. Lysosome, c. Peroxisome, d. Phagocytic vesicle, e. Nucleus, f. Nucleolus, g. Chromatin, h. Karyoplasm, i. Cytoskeleton, j. Centrioles, k. Plasma membrane, I. Cytoplasm, m. Rough endoplasmic reticulum, n. Smooth endoplasmic reticulum, o. Mitochondrion, p. Free ribosomes, q. Phospholipid bilayer, r. Integral protein, s. Carbohydrate chain, t. Peripheral protein, u. Phosphate molecule, v. Lipid layer, w. Cholesterol molecule.

## WORK 1.7. ION CHANNELS IN CELL MEMBRANES. PRINCIPLES OF THEIR CLASSIFICATION

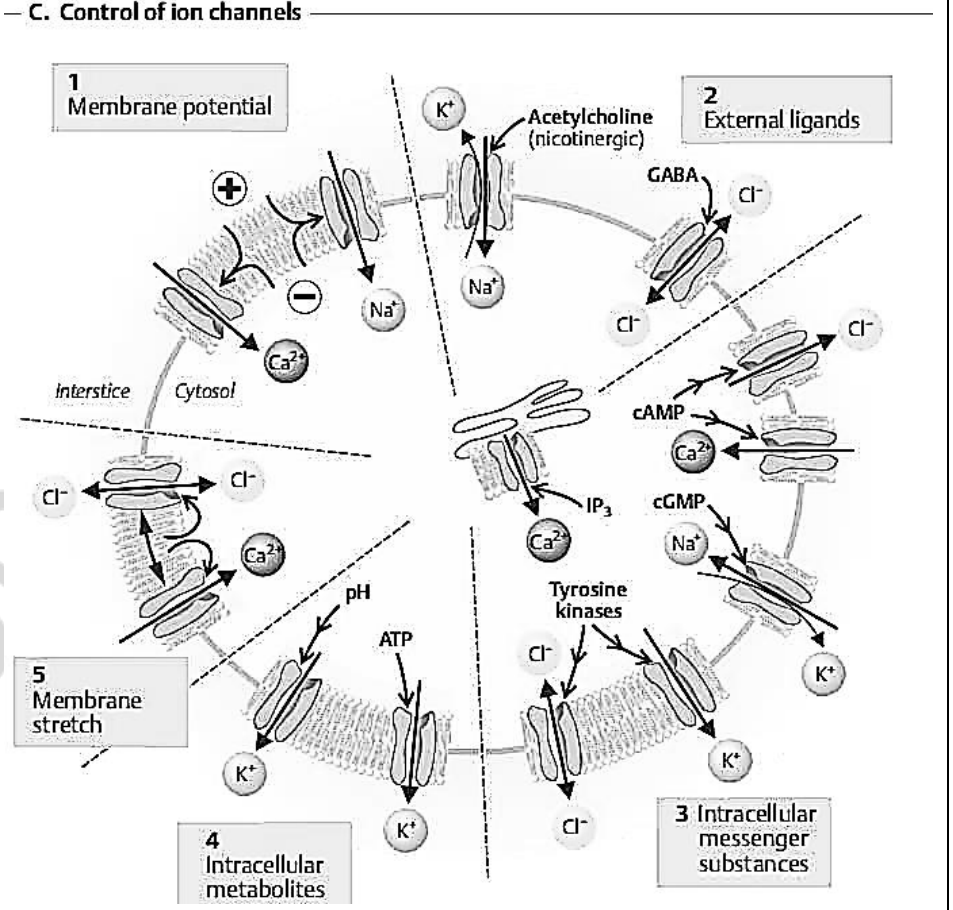
Channel open-probability is controlled by five main factors:

- Membrane potential (1), especially in Na+, $Ca^{2+}$ and $K^+$ channels in nerve and muscle fibers (**voltage-gated channels**).
- External ligands that bind with the channel (2 – **ligand-gated channels**). This includes **acetylcholine** (ACh) on the postsynaptic membrane of nicotinic synapses (cation channels), glutamate (cation channels), and glycine or (gamma-aminobutyric acid) GABA ($Cl^-$ channels).
- Intracellular messenger substances (3) such as:

– cAMP (e. g., in $Ca^{2+}$ channels in myocardial cells and $Cl^-$ channels in epithelial cells);
– cGMP (plays a role in muscarinergic effects of acetylcholine and in excitation of the retinal rods);
– IP3 (e. g. opening of $Ca^{2+}$ channels of intracellular $Ca^{2+}$ stores);
– G-proteins (e. g. $Ca^{2+}$ channels of the cell membrane);
– Tyrosine kinases (e. g. $Cl^-$ and $K^+$ channels during apoptosis);
– $Ca^{2+}$ (affects, for instance, $K^+$ channels and degree of activation of rapid $Na^+$ channels) ets;

- Intracellular metabolites (4) such as ATP (e. g., in $K^+$ channels in the heart and B cells in pancreatic islets) or $H^+$ ions (e. g., in $K^+$ channels in renal epithelial cells);
- Membrane stretch (5 – **mechanically gated** or **leakage channels**), the direct or indirect effects of which play a role in $Ca^{2+}$ channels of smooth muscle fibers and generally in normal $K^+$ and $Cl^-$ channels in swelling cells.

| Buzzword | Full name | Main function |
|---|---|---|
| cAMP | | |
| cGMP | | |
| IP3 | | |
| GABA | | |
| ACh | | |
| ATP | | |




THE LABORATORY WORKS ARE PASSED WITH MARK

Teacher's signature

## SECTION "PHYSIOLOGY OF EXCITABLE TISSUES"

## Lesson 2. THE CONCEPT OF CHEMICAL AND ELECTRICAL SIGNALING. RECEPTORS, THEIR TYPES. EXCITABLE TISSUES AND THEIR GENERAL PROPERTIES. BIOLOGICAL POTENTIALS. ELECTRODONTODIAGNOSTICS

DATE OF CLASSES
«____» _____ 20____
day month year

**BASIC QUESTIONS:**

1. The concept of chemical and electrical signaling. Information exchange between the cell and the environment.
2. Concepts: information, signal. Types of signals. The concept of cellular (molecular) receptors and its functions. The receptor mechanisms of signals perception. Basic ways of signal transmission.
3. General properties of excitable tissues. Excitation and forms of its manifestation. Indicators (parameters) excitability. Electrodontodiagnostics (electric dental pulp test), its use in dentistry.
4. Biopotentials, their types. Resting membrane potential, its origin. Galvanism.
5. The action potential (AP). Phases and ion mechanisms of AP generation. Excitability alteration during AP.
6. Basic laws of excitable tissues response to the stimulus action. Chronaximetry, its use to study the excitability of muscles and nerves.
7. Sensory receptors: definition, classification, functions, basic properties. Receptor and generator potentials. Basic principles of information coding in sensory receptors.

LITERATURE

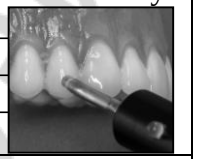
Main


1. Lecture & E-learning system.
2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et all] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 9–27, 144–148, 635–645.
3. *Severina, T. G.* Physiology of blood. Lecture notes / T. G. Severina. 2nd ed. Minsk : BSMU, 2017. P. 14–18.


Additional


4. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 53–64, 89–93, 159–160.
5. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 61–74, 577–578, 931–935.


Electroodontodiagnostics (electric dental pulp test), its use in dentistry:
_____
_____
_____



Galvanism is _____
_____
_____
_____

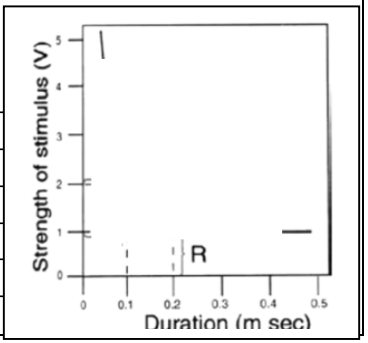
Chronaximetry, its use to study the excitability of muscles and nerves.

*Draw "force-duration" curve, label it and explain*:

Rheobase (R) — is _____
_____

Utilization time (UT) — is _____
_____

Chronaxia (Chr) — is _____
_____

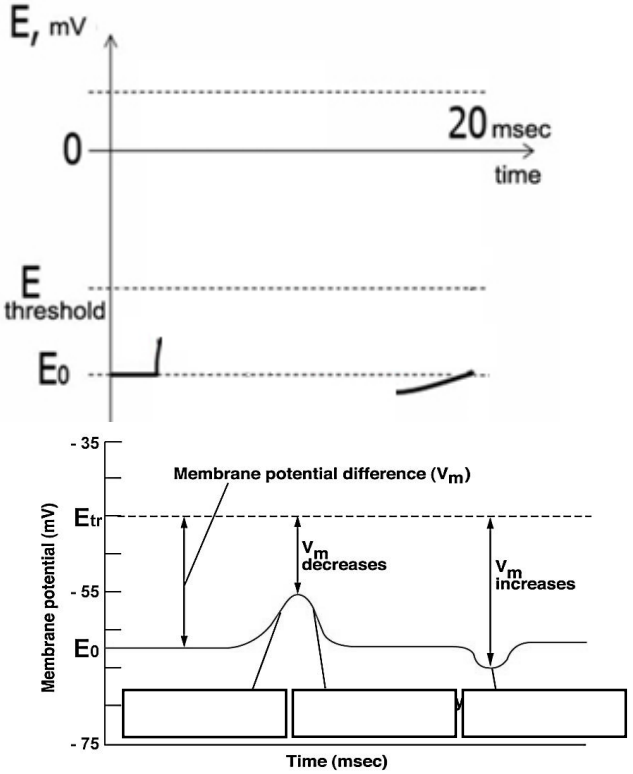


| **WORK 2.1. BUZZWORD** | |
|---|---|
| Irritation — | Generator potential — |
| Irritability — | Action potential — |
| Excitation — | Refractory period — |
| Excitability — | Law “all-or-none” — |
| Excitable tissues — | Law of force — |
| Resting potential — | Law of time (duration of stimulation) — |
| Depolarization — | “Force-time” law (“strength-duration” curve) — |
| Repolarization — | law of polar excitation — |
| Hyperpolarization — | Lability — |
| Local potential — | Adaptation — |
| Receptor potential — | Accommodation — |
| The ions permeability ratio at rest ($P_{K^+} : P_{Na^+} : P_{Cl^-}$) — **1** : : . | The ions permeability ratio during the excitation ($P_{K^+} : P_{Na^+} : P_{Cl^-}$) — **1** : : . |
| | |
| The main factors determining resting membrane potential (RMP) value are: | Types of biopotentials: |
| 1)_____<br>2)_____<br>3) | 1)_____<br>2)_____<br>3) |

**WORK 2.2. THE ACTION POTENTIAL AND CHANGES OF MPS**

*1. Draw **action potential**.*

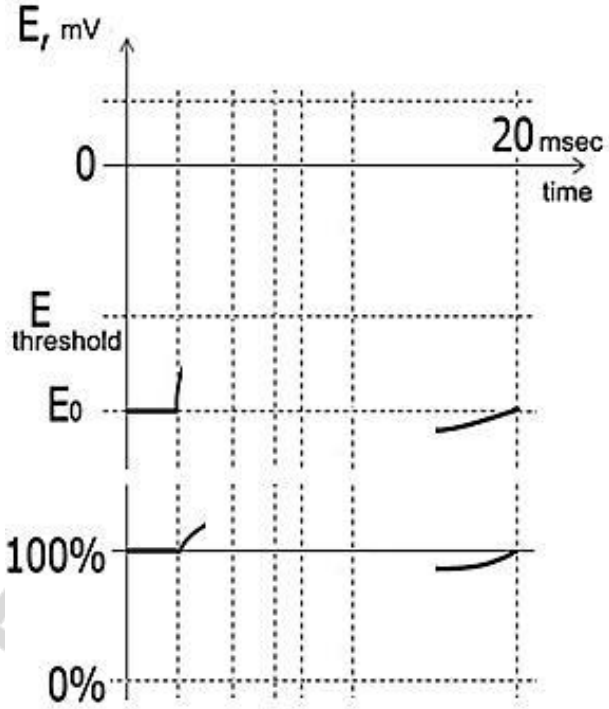
*2. Indicate the **phases** and describe the **ion mechanisms**.*




**WORK 2.3. EXCITATION AND EXCITABILITY OF CELL MEMBRANE**

*1. Draw **action potential** and synchronous changes of cell membrane **excitability**.*

*2. Indicate the **phases**.*




*0* – MRP

AP:
*1* – depolarization
*2* – overshoot
*3* – repolarization
*4* – after depolarization
*5* – hyperpolarization

Excitability alteration:
*6* – 100% excitability
*7* – supernormal period
*8* – absolute refractory period
*9* – absolute refractory period
*10* – relative refractory period
*11* – supernormal period
*12* – subnormal period

**WORK 2.4. BASIC LAWS OF EXCITABLE TISSUES RESPONSE TO THE STIMULUS ACTION**

*List and describe basic laws of excitable tissues response to the stimulus action:*

1. _____ 2. _____

_____ _____

_____ _____

3. _____ 4. _____

**WORK 2.5. THE EFFECT OF $Na^+$ AND $K^+$ IONS ON THE MEMBRANE RESTING POTENTIAL AND ACTION POTENTIAL. VIRTUAL PROGRAM "NMJ" ("NEUROMUSCULAR JUNCTION")**

1. This work is done in a computer class (No. 104). The student uses the **NMJ** program (link on the desktop). The NMJ program is a virtual simulator of operations on an isolated neuromuscular preparation placed in Ringer's solution (Fig. 2.1). It is possible to stimulate both the muscle fiber and the nerve, and to change the concentration of ions in the solution.

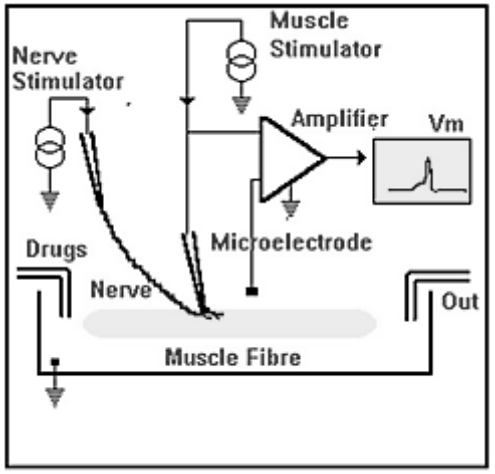



Fig. 2.1

2. Click in the upper line (menu):
1) Ions → potassium (K+) → 5 mM, sodium (Na+) → 120 mM;
2) Stimulated → Nerve;
3) Clipboard → Copy to clipboard (fig. 2.2–2.4).

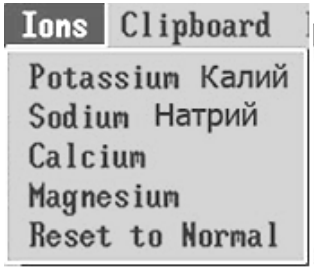



Fig. 2.2

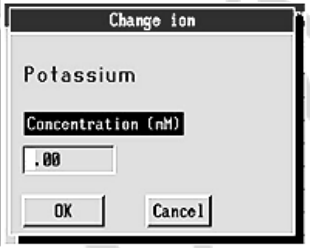



Fig. 2.3

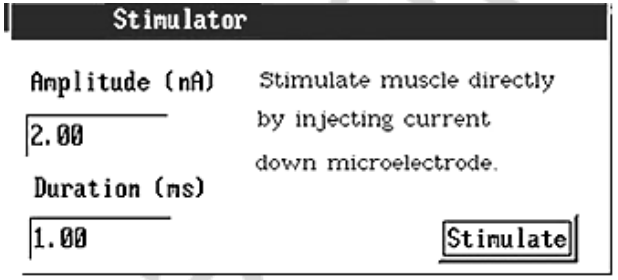



Fig. 2.4

3. You will receive an image, two graphics, as in Fig. 2.5, where the value of RMP (resting membrane potential) (arrow 1) is shown under conditions of optimal content of K + (potassium) and Na + (sodium) ions in Ringer's solution (arrow 2) (Fig. 2.5) and AP (action potential) graphs during electrical stimulation of muscles with electrical current with a force of 2 mA for 1 ms (Fig. 2.4).

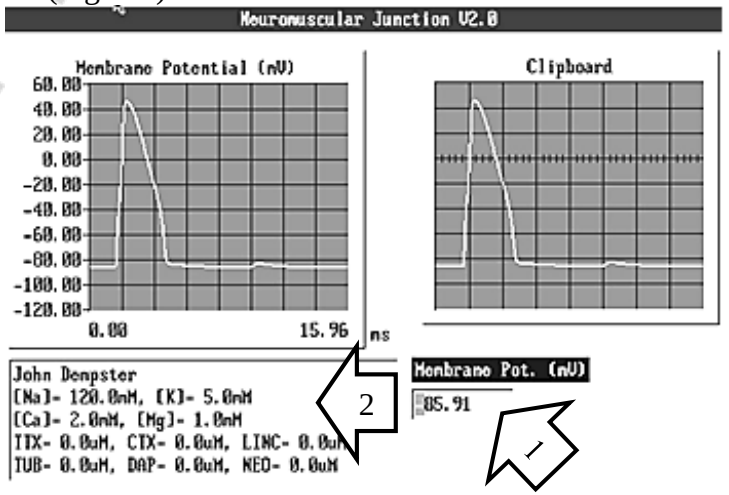



Fig. 2.5

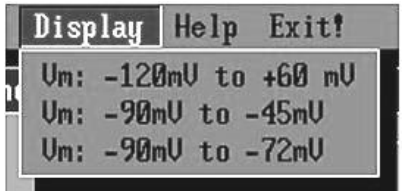



Fig. 2.6

4. After that the program allows you to change the concentrations of electrolytes (potassium and sodium) in solution using commands "Ions" (fig. 2.2.) and "Change ion concentration" (fig. 2.3). You mast record the values of RMP and AP (fig. 2.6) after the indirect electrical stimulation of muscle with the same single electric current amplitude of 2 mA for 1 ms: "stimulate → nerve".

**WORK 2.5.** (sequential)

**Directions for recording the Protocol**:

1. Simulate the change in membrane potentials (RMP and AP) by indirect electrical stimulation of muscles under conditions of optimal concentration of K+ and Na+ ions, as well as increasing and decreasing their concentration (according to the instructions in Table 2.1) in the solution surrounding the neuromuscular preparation.

2. Record the results of RMP and AP changes in the table 2.1.

3. Draw on fig. 2.7 the resulting graphs of RMP and AP using colored pencils. There should be 4 graphics of different colors.

4. Explain the effect of the changes in the concentration of the $K^+$ and $Na^+$ ions on the values of RMP and AP.

**PROTOCOL**

*Table 2.1*

| The extracellular concentration of ions | | | The magnitude of the potentials | |
|---|---|---|---|---|
| potassium | sodium | | resting (MRP) | action (AP) |
| **5 mM** | 120 mM | Copy to clipboard | –85,9 mV | +45 mV |
| **8 mM** | 120 mM | Copy to clipboard | | |
| **2 mM** | 120 mM | Copy to clipboard | | |
| Clipboard → clear | | | | |
| 5 mM | **120 mM** | Copy to clipboard | –85,9 mV | +45 mV |
| 5 mM | **160 mM** | Copy to clipboard | | |
| 5 mM | **100 mM** | Copy to clipboard | | |

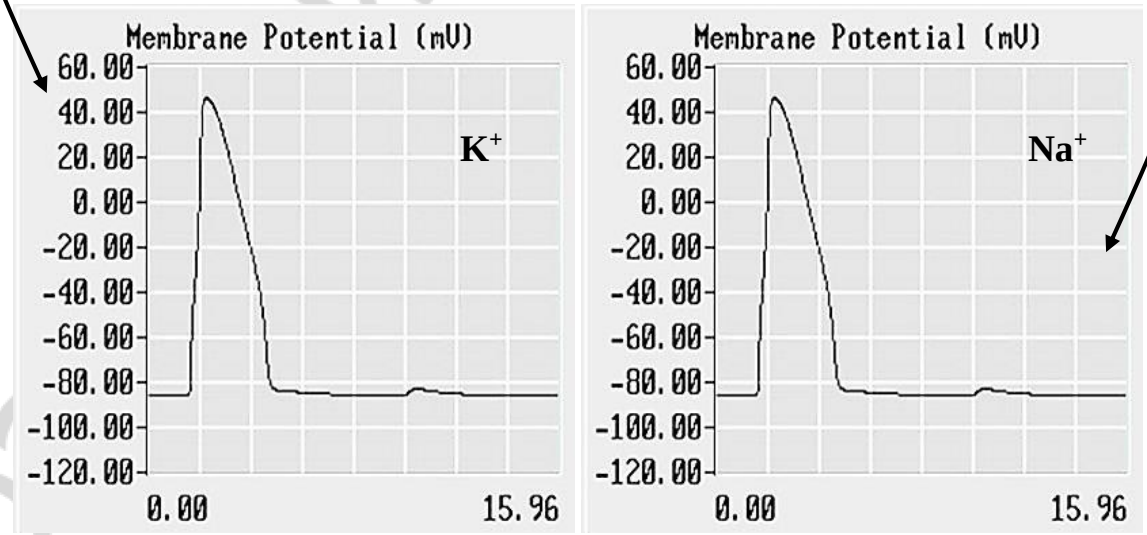



Fig. 2.7

**Conclusion**: the concentration of potassium ions in the extracellular fluid determines magnitude of the *resting/action* potential, while the content of sodium ions determines magnitude of the *resting/action* potential.

**WORK 2.6. THE RECEPTOR MECHANISMS OF SIGNALS PERCEPTION. RECEPTORS AND THEIR TYPES**

Molecular (cellular) receptors —

Sensory receptors —

| **Classification of molecular (cellular) receptors:** | **Corresponding ligands (examples):** | **Classification of sensory receptors:** | **The main categories of information signals:** |
|---|---|---|---|
| Membrane receptors:<br>1._____<br>2._____<br>3._____<br><br>Intracellular receptors:<br>1. _____<br>2. _____ | _____<br>_____<br>_____<br><br>_____<br>_____ | | of the chemical nature:<br>_____<br>_____<br>of the physical nature:<br>_____<br>_____<br>_____<br>of the physico-chemical nature:<br>_____<br>_____<br>Signals, indicating complex events:<br>_____ |

*Draw a schematic structure of membrane receptors & describe a mechanism of its working:*

*Draw a schematic structure of sensory neurons & mark with arrows the direction of propagation of excitation:*

| 7-TMSRs | 1-TMSRs | LGICs | Pseudo unipolar (sensory somatic or autonomic) neuron | Bipolar (smell or vision) neuron |
|---|---|---|---|---|
| | | | | |

THE LABORATORY WORKS ARE PASSED WITH MARK:

_____

Teacher's signature

## Lesson 3. CONDUCTION OF EXCITATION BY NERVE FIBERS AND SYNAPSES. PHYSIOLOGICAL BASIS OF CONDUCTIVE ANESTHESIA IN DENTAL PRACTICE

DATE OF CLASSES
«____» _____ 20____
day month year

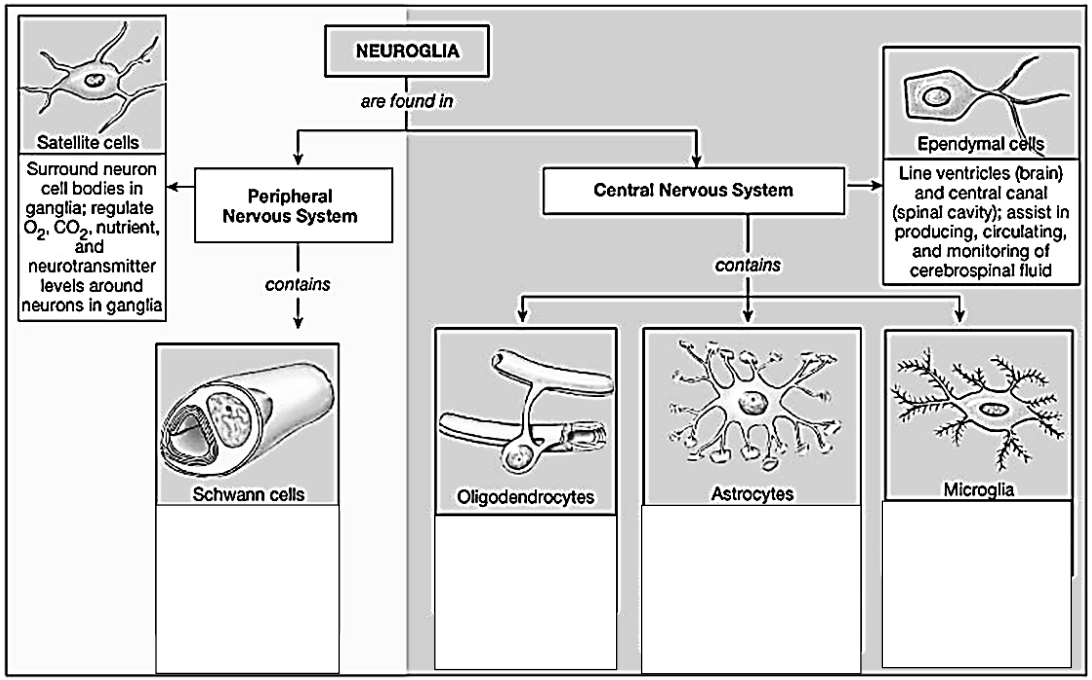
| **Basic questions:** | LITERATURE<br>Main |
|---|---|
| 1. Nerve fibers: structure and functions. Classification of nerve fibers.<br>2. Mechanisms and laws of excitation conduction by myelinated and unmyelinated nerve fibers.<br>3. Physiological basis of conductive anesthesia in dental practice.<br>4. Transport of substances in nerve fibers: types, functions.<br>5. Synapses: structure, classification, functions. Functional properties of synapses.<br>6. The mechanisms of excitation conduction in synapses. Excitatory neurotransmitters. EPSP. An End Plate Potential (EPP), its transformation into an action potential. Role of acetylcholinesterase.<br>7. Inhibitory synapses, its neurotransmitters. Ion mechanisms of IPSP. Summation.<br>8. The possibilities of directed pharmacological influence on synaptic transmission. | 1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 17–18, 47–54, 66–75.<br>Additional<br>3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 85–90, 93–95, 121–135.<br>4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 69, 71–72, 89–92, 580–592. |

**Buzzword**

| | |
|---|---|
| Nerve fibers — is _____<br>_____ | Types of nerve fibers:<br>1. _____,<br>2. |
| Continuous conduction — _____ | Saltatory conduction — _____ |
| Nervous tissue consist of cell types: 1. _____<br>and 2. _____ | EPSP — _____<br>EPP — _____<br>IPSP — |

**WORK 3.1. STUDYING THE NERVE FIBERS, ITS STRUCTURE, TYPES, AND FUNCTIONS.**

| *Draw a neuron, indicate its departments and functions:* | *Draw a scheme of continuous conduction:* | *Draw a scheme of saltatory conduction:* |
|---|---|---|
| | | |




## WORK 3.2. NEUROGLIA

*Using the material of the lectures and E-learning system, write the functions of the cells of the neuroglia*

**Two types in the PNS**
Schwann cells
satellite cells
**Four types in the CNS**
Astrocytes
Oligodendrocytes
Microglia
Ependymal cells

## WORK 3.3. STUDYING THE LAWS OF EXCITATION CONDUCTION BY NERVE FIBERS

1. _____
_____
_____
2. _____
_____
_____
3. _____
_____
_____

## WORK 3.4. STUDYING THE CLASSIFICATION OF NERVE FIBERS AND ITS SENSITIVITY TO ANESTHESIA

*Table 3.1*

**Classification of nerve fibers**

| Fiber type | Mielinization | Diameter (μm) | Conduction rate (m/s) | Sensitivity to anesthesia | Function according to fiber type |
|---|---|---|---|---|---|
| $A_\alpha$ | + | 12–22 | 70–120 | + | Skeletal muscle efferent, afferents in muscle spindles (Ib) and tendon organs (Ib) |
| $A_\beta$ | + | 8–12 | 40–70 | ++ | Mechanoafferents of skin (II) |
| $A_\gamma$ | + | 4–8 | 15–40 | ++ | Muscle spindle efferents |
| $A_\delta$ | + | 1–4 | 5–15 | ++++ | Skin afferents (temperature and “fast” pain) (III) |
| B | + – | 1–3 | 3–18 | ++++ | Sympathetic preganglionic, visceral afferents |
| C | – | 0,5–1,5 | 0,5–3 | ++++ | Skin afferents (“slow” pain), sympathetic postganglionic afferents (IV) |

**WORK 3.5. STUDYING THE MECHANISMS OF TRANSPORT OF SUBSTANCES IN NERVE FIBERS**

*Indicate*: a — anterograde transport is mediated by kinesin, b — retrograde transport is mediated by dynein, c — synaptic cleft, d — neurotransmitter, e — receptor

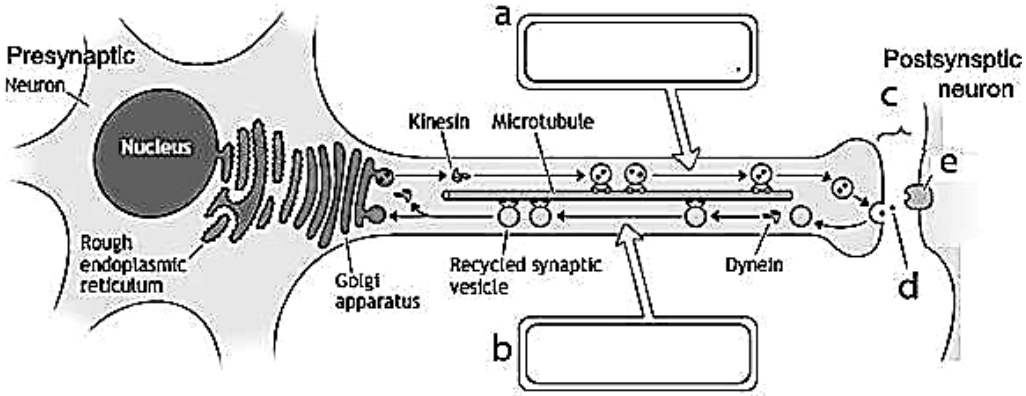



Fig. 3.1. Transport of substances in nerve fibers

**WORK 3.6. CHEMICAL SYNAPSES: STRUCTURE, CLASSIFICATION, FUNCTIONS**

| Excitatory synapse | Inhibitory synapse |
|---|---|
| EPSP — | IPSP — |
| Principle neurotransmitters (write examples): | |
| | |
| Choose the changes of membrane potential: | |
| -5, -30, -90, -120 mV | -5, -30, -90, -120 mV |
| Is it depolarization or hyperpolarization: | |
| | |

Characteristics of Postsynaptic Potentials (EPSP and IPSP).
All postsynaptic potentials have certain characteristics in common. Importantly, the local potential is a graded potential; that is, its amplitude is proportional to the size of the stimulus. Measurement of a local potential uses the membrain resting potential as its baseline. If the membrane's resting potential is depolarized from – 80 to – 70 mV during the local potential, the local potential has an amplitude of 10 mV. This potential change is one of decreasing negativity (or of *depolarization* = excitatory postsynaptic potential EPSP), but it could also be one of increasing negativity (or of *hyperpolarization* = inhibitory postsynaptic potential IPSP).

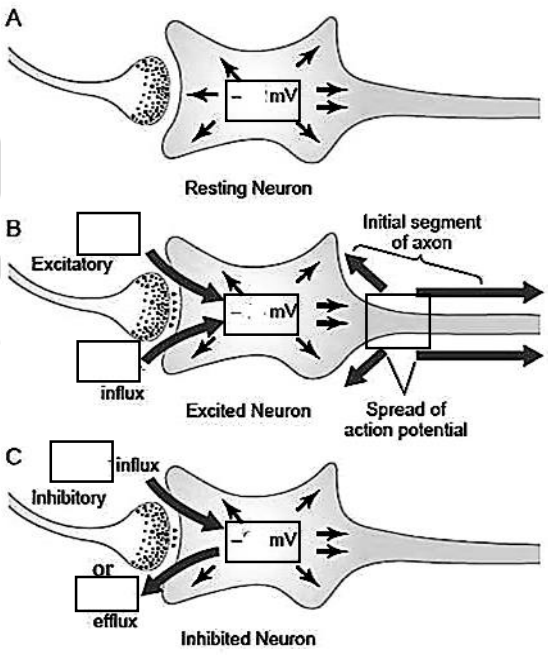



Fig. 3.2

*Draw the diagram of summation of EPSPs (red) and IPSP (blue) that will result firing of AP:*

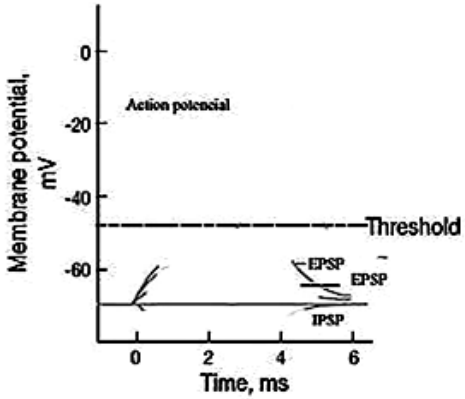
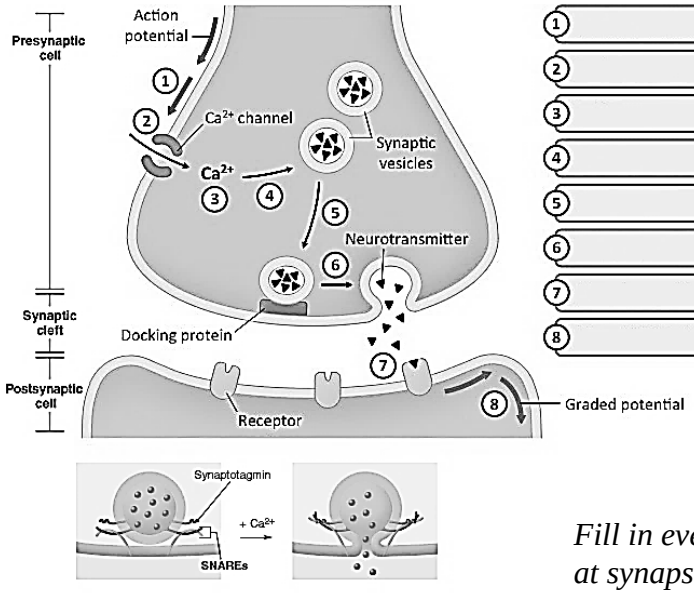



Fig. 3.3

## WORK 3.7. STUDYING THE MECHANISMS OF CONDUCTION IN CNS SYNAPSES




1 _____

2 _____

3 _____

4 _____

5 _____

6 _____

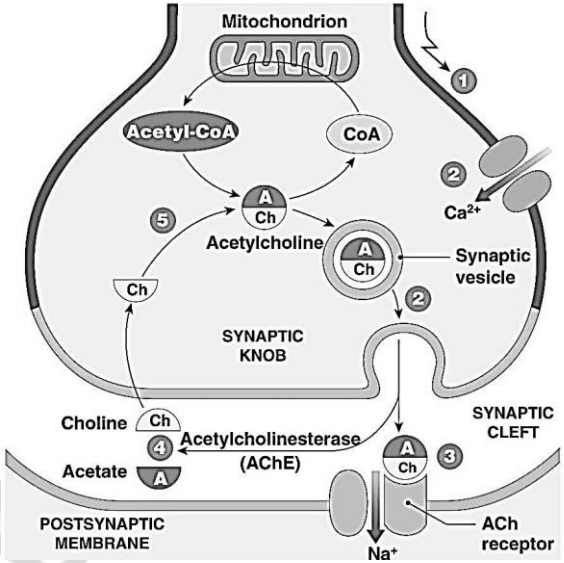
7 _____

8 _____

*Fill in events, occurring at synapse*

## WORK 3.8. STUDYING THE ROLE OF ACETYLCHOLINESTERASE

**The events that occur at a cholinergic synapse**




**Events Occurring at Synapse**

1 _____

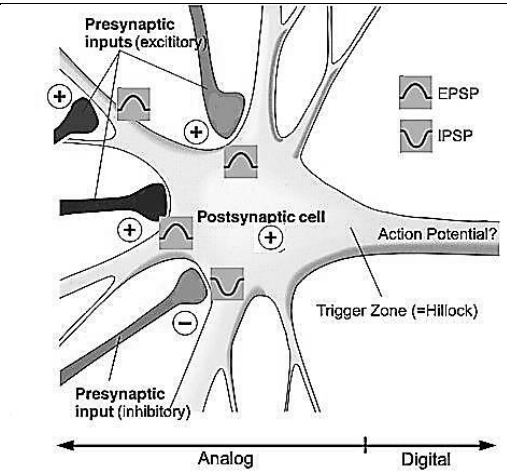
2 _____

3 _____

4 _____

5 _____

## WORK 3.9. STUDYING THE MECHANISMS OF SUMMATION AT CENTRAL SYNAPSES




_____

*yes or no?*

*Draw the scheme of temporal summation of graded potentials*

*Draw the scheme of spatial summation of graded potentials*

## WORK 3.10. STRUCTURE OF NEURO-EFFECTOR JUNCTION (WITH SMOOTH MYOCYTES, GLANDULAR CELLS, MYOEPITHELIAL CELLS)

*Indicate*:

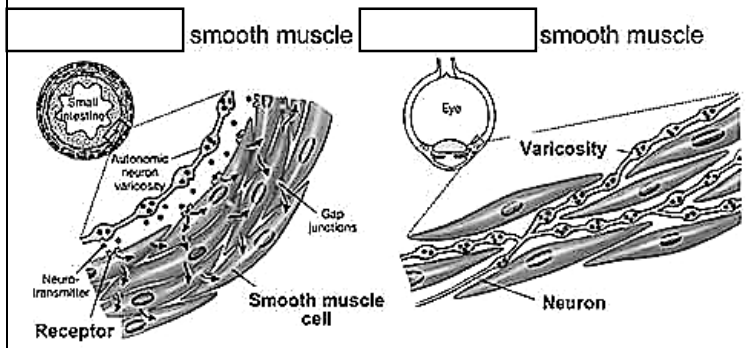



Fig. 3.4. Diagram of structure of neuro-effector junction

## WORK 3.11. STUDYING THE PHYSIOLOGICAL BASES OF CONDUCTION ANESTHESIA

Anesthesia (anaesthesia) in modern dentistry is a set of procedures aimed at the reduction or complete relief of pain during the treatment.

Two broad categories of anesthesia exist: local and general.

Local or regional anesthesia block transmission of nerve impulses from a specific part of the body. It has includes the injection and application techniques.

There are two main kinds of regional anesthesia — peripheral nerve block and infiltrative anesthesia, which is carried out by introducing of local anesthetics into the tissue surrounding the conductive nerve trunks (nerve block or regional nerve blockade) or its sensory endins (infiltrative anesthesia).

Local anesthetics (procaine, lidocaine, etc.) reversibly block impulse conduction along the axon membrane and other excitable membranes that use sodium channels as the main generator of action potentials.

The mechanism of action of local anesthetics are associated with their effects on receptors located near a intracellular inactivation gates (h-gate) of the sodium channel, which leads to a noticeable increase in the time of their inactivation and to the block of voltage gated sodium channels.

Thus, local anesthetics interfere inflow of sodium ions through the membrane and its depolarization. As a result, the generation of action potentials in nerve (pain) terminals at the injection and/or application anesthesia area, as well as conduction of excitation (action potentials) along the nerve fibers with nerve blockade, is disturbed. Recovery of sodium channels from blockade by local anesthetic is 10–1000 times slower than from normal physiological inactivation of channels. Regional (conductive anesthesia or nerve block) anesthesia is achieved by introducing anesthetic into the region of the conductive nerve trunks or plexuses. In this case, the pain sensitivity of the entire anatomical region, located far from the injection site of the anesthetic solution, is turned off.

Local anesthetics can block the transmission of a signal along any nerve fibers, but the sensitivity of the latter to anesthetic effects depends on its myelination, size, frequency of impulses on them, position of the fibers in the bundle.

*At first, sensory conductivity is blocked in type B and C fibers, then in Aδ fibers. Thus, the pain disappears first, then other kinds of sensitivity are suppressed, and motor functions the last one.*

Myelinated fibers are blocked earlier than non-myelinic fibers of the same diameter. To stop the initiation of myelinated fibers, it is necessary for the blockade to extend to three consecutive node of Ranvier. The effect of anesthesia is more expressed in actively acting axons, which are more accessible to local anesthetics. Aδ and C-fibers have a small diameter and participate in the transmission of high-frequency pain impulses. Therefore, they are blocked earlier and by lower concentrations of local anesthetics than Aα-fibers.

## Lesson 4. PHYSIOLOGY OF SKELETAL MUSCLES

DATE OF CLASSES
«____» _____ 20____
day month year

**Basic questions:**

1. Types of muscle tissue. Motor units, their types, structural and functional properties.
2. Physiological properties of skeletal muscles and their functions.
3. Neuromuscular synapse: mechanisms of signal transduction.
4. Structural and functional characteristics of muscle fiber. Sarcomere. Main proteins of myofilaments, their functions.
5. Mechanisms of contraction and relaxation of a single muscle fiber and a whole muscle. Excitation-contraction coupling.
6. A single contraction of muscle fiber and its phases. Types and regimes of skeletal muscle contraction. Tetanic muscle contraction and its types.
7. Force and work of muscle contraction. Nature of muscle tone. Muscle fatigue.
8. Dynamometry of a hand and back muscles.

LITERATURE

Main

1. Lecture & E-learning system.
2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 28–44, 81–83.

Additional

3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 99–111.
4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 75–95.

**BUZZWORD**

There are three types of muscle tissue: 1. _____
2. 3.

Sarcomere — _____

Functions of musculature system:
1 _____, 2 _____, 3 _____
4 _____, 5 _____

Physiological properties of muscle tissues:
1. _____ 2._____
3. 4.

Motor units — is

Tone —

Tetanic contraction —

Fatigue —

**WORK 4.1. TYPES OF MUSCLE FIBERS**

*Complete the table using lecture and e-learning materials.*

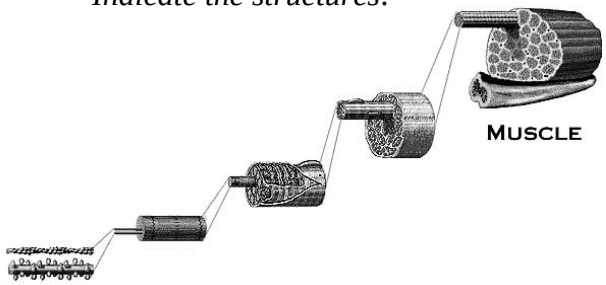
| Type | I (Slow) | IIa (FOG) | IIb (FG) |
|---|---|---|---|
| description | | | |
| myoglobin | | | |
| mitochondria | | | |
| fatigues | | | |
| color | | | |
| diameter | | | |

**WORK 4.2. MOTOR UNITS**

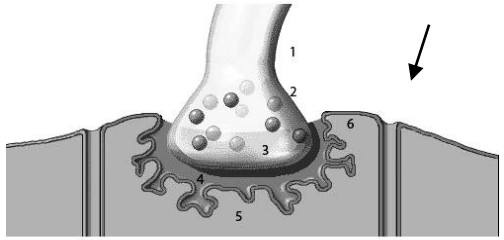
*Draw a motor unit.*

**WORK 4.3. LEVELS OF ORGANIZATION OF SKELETAL MUSCLE**

*Indicate the structures*:



## WORK 4.4. NEUROMUSCULAR SYNAPSE: MECHANISMS OF SIGNAL TRANSDUCTION




*Draw a sarcomere structure*

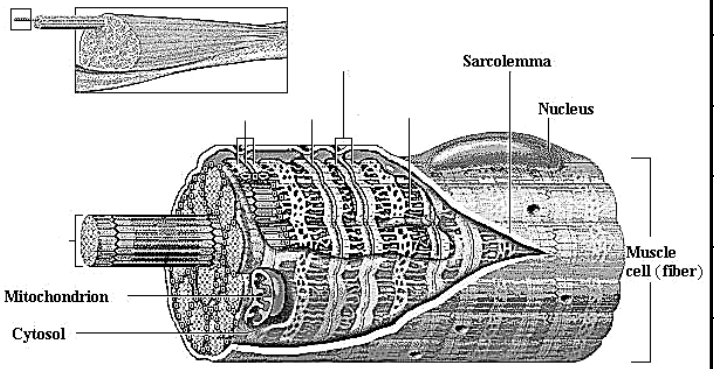
*List the mechanisms of excitation-contraction coupling*:

1. AP
2. _____
3. _____
4. _____
5. EPSP → AP
6. _____
7. _____
8. _____
9. _____

## WORK 4.5. STRUCTURAL AND FUNCTIONAL CHARACTERISTICS OF MUSCLE FIBER

*Indicate structures using a lecture or program Interactive Physiology materials.*

INTERNAL STRUCTURE OF A SKELETAL MUSCLE CELL

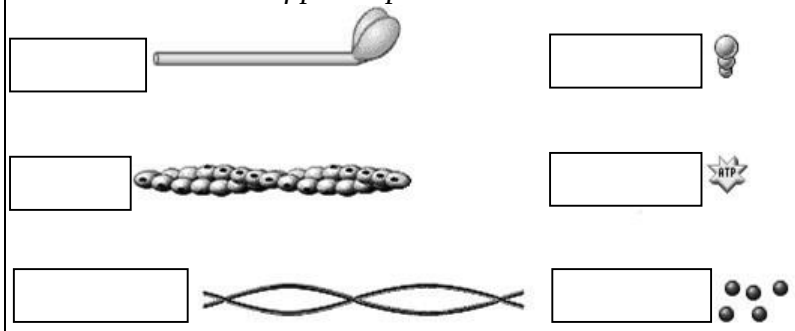



Write the functions of the following structures:

| Structure | Function |
|---|---|
| Myofibril | |
| Triad | |
| T-tubule | |
| Sarcoplasmic reticulum (CR) | |
| Terminal cisternae | |

## WORK 4.6. MOLECULAR PARTICIPANTS OF CONTRACTION

*Indicate the names of participants*:




Phases of muscle twitch:

| Phase | Explanation |
|---|---|
| 1. | |
| 2. | |
| 3. | |

## WORK 4.7. MECHANISM OF CONTRACTION AND RELAXATION OF WHOLE MUSCLE

*Open "IP" → Muscular → Contraction of whole muscle → read and go to p. 4 (muscle twitch) → draw a graph of muscle twitch and indicate contraction phases.*

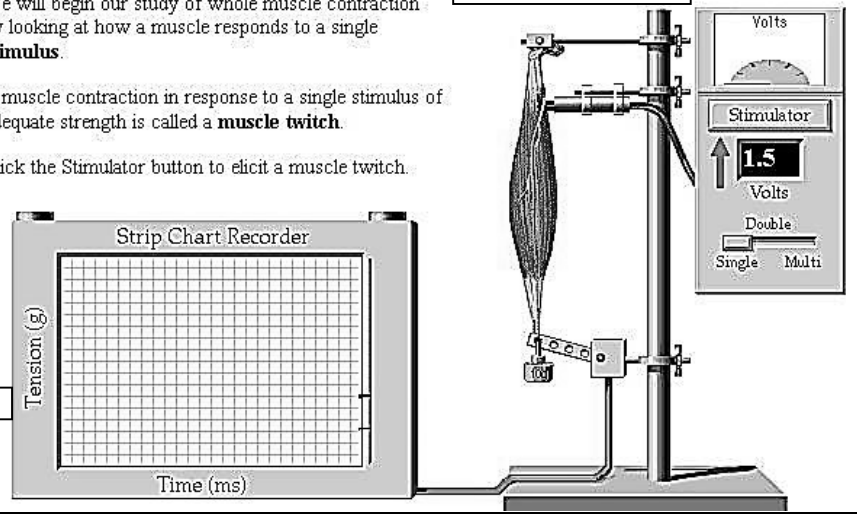
MUSCLE TWITCH

**PROTOCOL**

We will begin our study of whole muscle contraction by looking at how a muscle responds to a single **stimulus**.

A muscle contraction in response to a single stimulus of adequate strength is called a **muscle twitch**.

Click the Stimulator button to elicit a muscle twitch.



## WORKS 4.8. ELECTROMYOGRAPHY (EMG)

Electromyography (EMG) — a method of the detection and recording of electrical activity generated by muscle fibers.

Muscle biopotential abduction is carried out using surface (cutaneous, overhead) or needle (injected) electrodes.

The advantage of total EMG is non-invasive research and, as a rule, the absence of electrical stimulation of muscles and nerves. This method allows us to investigate the nature of the muscle biocurrents at rest and with voluntary contractions, which ensured its wide use in physiological and clinical practice.

An electromyogram is the result of the interference of a multitude of action potentials asynchronously arising in different motor units. At present, the quantitative analysis of EMG is carried out using special instruments that allow measuring the frequency of oscillations, carrying out spectral analysis and estimating the total and average amplitude of pulses. One of the most common methods for analyzing muscle currents is their integration, that is, the summation of all amplitudes per unit time. When dividing the total amplitude by the number of pulses, their average amplitude is calculated. This indicator is proportional to the size of the developed muscular effort.

At rest, a low amplitude EMG (5–10 μV) is recorded, associated with the redistribution of muscle tone while maintaining posture. With a weak contraction and muscle tension, an increase in electrical activity is observed, which reaches a maximum with an voluntary effort (the amplitude of biocurrents can increase to 3000 μV at a frequency of up to 100 Hz)..

**Materials and equipment:** silver surface electrodes (6 pcs.), electrically conductive paste, 70% ethanol solution, cotton gauze swabs, rubber clips (2 pcs.), a set of weights from 0.5 to 3 kg, biopotential amplifier, recorder, oscillographic indicator and myography analyzer.

**Accomplishment**. A bipolar electrodes are applied to the subject in a standing position on the skin above the biceps muscle of the shoulder of the right hand. Common electrode impose on the skin of the shoulder not far from the points of registration of the EMG. In the places where the electrodes are applied, the skin is degreased with alcohol beforehand, and the electrodes are smeared with a conductive paste, and then EMG is recorded and analyzed under various functional conditions:

a) rest: arms loosely lowered, muscles relaxed;

b) flexion of the arm at the elbow joint from position “a”;

c) extension of the arm from position “b”;

d) the arm is fixed at the elbow so that the forearm is in a horizontal position, and loads of increasing weight are put on the palm.

**Directions for recording the Protocol:**

1. Draw the EMG recorded under different conditions.
2. Make a conclusion about the state of the motor center activity that innervates the shoulder biceps under the experiment

**PROTOCOL**

1. Figures of EMG under different conditions:

| EMG drawing of the biceps under various conditions | rest | arm flexion | arm extension | fixation and holding the load |
|---|---|---|---|---|
| | | | | |

2. **Conclusion**: The electrical activity of the shoulder biceps and that of nerve centers innervating it, under experiment (when bending arm at the elbow and especially with the additional muscle tension for holding the weights) versus the state of rest is considerably _____ (increased or reduced), it being testified by _____ (increase or decrease) of amplitude and frequency of the waves of EMG.

## WORK 4.9. DYNAMOMETRY OF HANDS AND BACK MUSCLES

Dynamometry is a method of measuring of the strength of muscle contraction.

Muscle strength is an important indicator of their contractility, as well as the physical development of the human body. It is estimated by the weight of the load that can be held by the muscle at its maximum excitation, without changing the length of the muscle. Muscle strength depends on its physiological cross-section, initial length, contraction rate and other factors. Muscle contraction is measured by dynamometers and expressed in absolute units (kg or N, as well as in kg/cm$^2$ of muscle cross section (ranging from 2 to 10 kg/cm$^2$)) or in relative units (relative to body mass, expressed in %). Dynamometry (especially manual) is widely used in medicine and in the physiology of labor and sports activities.

**Materials and equipment**: manual dynamometer, back muscles dynamometer, medical scale (fig. 4.1, 4.2, 4.3).

Fig. 4.1. Manual dynamometer (at laboratory room № **131**)

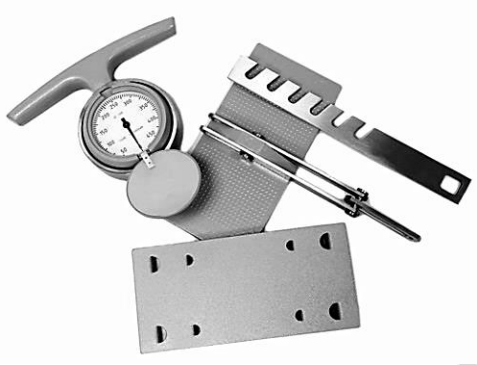


Fig. 4.2. Back muscles dynamometer (at laboratory room № **131**)

Fig. 4.3. Medical scale (at laboratory room № **135**)

**Accomplishment**. The strength of the hands is determined using a manual dynamometer. The dynamometer is held parallel to the position of the floor (fig. 4.4).

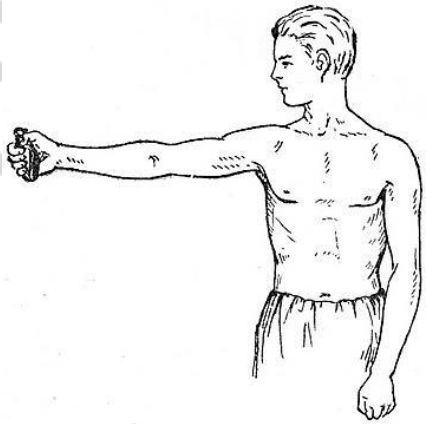


Fig. 4.4

Perform maximum compression of the dynamometer with your hand. The measurement is carried out three times with each hand. Of the three dimensions (for each hand) choose the largest.

Measure your body mass (without shoes) on medical scales and subtract 1 kg from it (conditionally, the weight of the clothes is taken), obtaining an indicator of body weight.

*Calculate* the Hand Strength Index (HSI) for right and left hands by the formula:

$$\textbf{HSI} = \frac{\text{muscle strength in kg} \times 100\ \%}{\text{body mass in kg}}$$

$\textbf{HSI}_r = \frac{\quad \times \quad}{\quad} = \_\_\_\_\_$; $\textbf{HSI}_l = \frac{\quad \times \quad}{\quad} = \_\_\_\_\_$.

**Satisfactory HSI** for men is **55 units**, for women — **50 units**.

Evaluation of the relative strength of the muscles of the hands is given in table 4.1. The strength of the muscles of the hands of students over the past ten years has decreased significantly. In this regard, in table 4.1 are given the standards of hand strength, taking into account the trend of its change in students of BSMU (boys and girls) in the last years.

The strength of the extensor muscles of the back is measured by a back muscles dynamometer (fig. 4.5) three times and the highest value is selected.

Fig. 4.5

*Calculate* the Back Strength Index (BSI) by the formula:

$$\textbf{BSI} = \frac{\text{Muscle strength in kg}}{\text{Body mass in kg}} = \frac{\qquad}{\qquad} = \_\_\_\_\_$$

**Satisfactory** BSI for men is **2 units**, for woman — **1.5 units**.

*Table 4.1*

Hand strength index of young humans

| Sex | Level of hand strength index (%) | | | | |
|---|---|---|---|---|---|
| | low | below the average | average | above the average | high |
| Male | < 41 | 41–50 | 51–60 | 61–70 | > 70 |
| Female | < 21 | 21–25 | 26–30 | 31–40 | > 40 |

**Directions for recording the Protocol**:

1. Put down the obtained data into the Protocol
2. Calculate HIS and BSI.
3. Evaluate muscle strength of the tested person and make a conclusion.

**PROTOCOL**

1. Body mass _____ (kg), sex ____ (м. or f.)

| Muscles | Muscle strength | Muscle strength index (in units) |
|---|---|---|
| Right hand | | HSI = |
| Left hand | | HSI = |
| Back extensors | | BSI = |

2. **Conclusion**: Level of right hand strength index is _____ _____, level of left hand strength index is _____ (*low, below the average, average, above the average, high*).
Back strength index is _____ (*satisfactory, unsatisfactory*).

THE LABORATORY WORKS ARE PASSED WITH MARK

_____
Teacher's signature

## Lesson 5. PHYSIOLOGY OF MUSCLES OF MAXILLOFACIAL REGION. PHYSIOLOGY OF SMOOTH MUSCLES. NOTION OF THE MYOEPITHELIAL AND GLANDULAR CELLS

DATE OF CLASSES
«____» _____ 20____
day month year

Main questions:

1. The concept of the components of the masticatory system and their functional interaction. Movement of the mandible. Physiological occlusion.
2. Muscles of the maxillofacial region and their functions. Functional purpose of individual muscles of mastication.
3. Work and strength of the muscles of mastication. Regulation of its contraction.
4. Parodont, its endurance to the pressure developed by the masticatory muscles. Gnathodynamometry.
5. Physiological properties and characteristics of smooth muscle. Smooth muscle tone.
6. Transmission of information from nerve fibers to smooth muscle. Neuroeffector connections of smooth muscle.
7. The concept of myoepithelial cells (salivary and other exocrine glands) and its functions.
8. Glandular epithelium, glands: functions, properties, especially bioelectrogenesis.

LITERATURE

Main


1. Lecture & E-learning system.
2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 28–46, 496–501.


Additional


3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 115–118.
4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 97–105.


**BUZZWORD**

Masticatory system —

Physiological occlusion —

Centric occlusion —

Centric relation —

Intercuspal position (2–4 mm) —

Electromyography —

Gnathodynamometry —

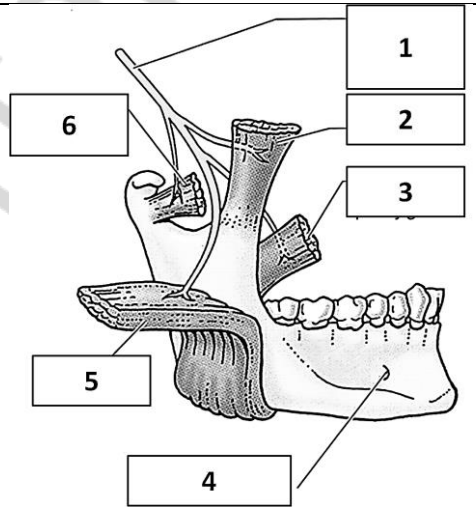



Fig. 5.1

**WORK 5.1. MUSCLES OF MASTICATION**

| Structure | Function |
|---|---|
| 2. | |
| 3. | |
| 5. | |
| 6. | |
| 1. | |
| 4. | |

## WORK 5.2. ELECTROMYOGRAPHY OF THE MUSCLES OF MASTICATION

This is a method of bipolar recording of the total electrical activity of the chewing muscles. When chewing food, the lower jaw with respect to the upper one makes movements in six directions due to the complex structure of the temporomandibular joint and the location of the chewing muscles.

Chewing muscles, especially the masseter and medial pterygoid muscles, refers as a muscles of power. According to Weber, these muscles with a cross section of 1 $cm^2$ can develop a strength of 10 kg, i.e. more than the gastrocnemius (5.9 $kg/cm^2$).

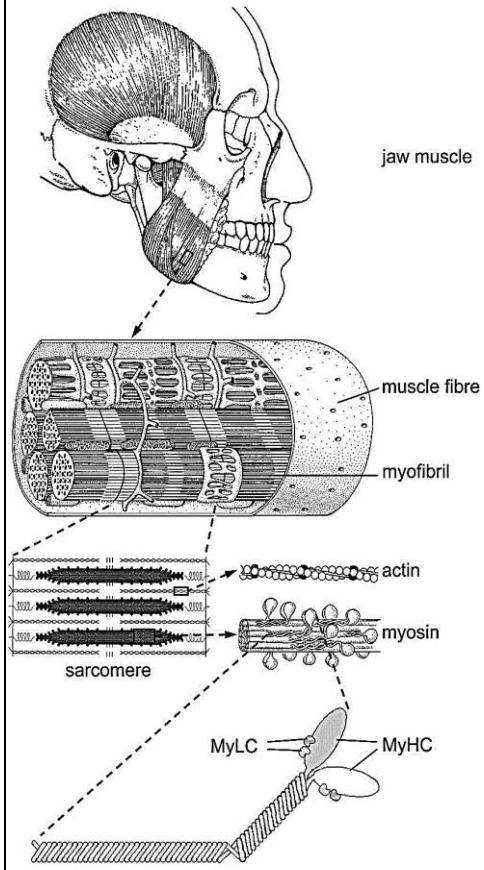


Fig. 5.2

The mandibular branch of the trigeminal nerve performs innervation of the muscles of mastication. Development of pathological processes in the lower jaw leads to impaired digestion in the oral cavity. Pain reflex reduces contractility and electrical activity of the masticatory muscles.

Currently, the quantitative analysis of EMG is carried out using special instruments that allow one to measure the frequency of oscillations, to carry out spectral analysis and to estimate the average amplitude of the pulses. One of the most common methods of EMG analysis is its integration (determining the average amplitude value per unit time, i.e., dividing the total amplitude by the number of measurements, for example, per second). This indicator is proportional to the size of the developed muscular effort. In a state of functional rest, low amplitude electrical activity of the muscles is recorded, which reaches a maximum with arbitrary compression of teeth.

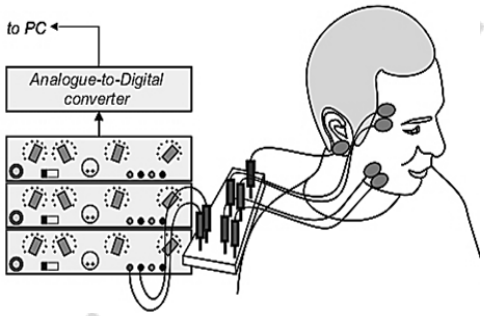


Fig. 5.3

**Accomplishment**. The subject is seat in a chair; the skin is degreased at the points where the electrodes are applied. Above the masseter and digastric muscles of the face, glue on two electrodes, after having previously smeared them with a paste. The common electrode is fixed on the earlobe with a clip, and then the electrical activity of the muscles is recorded in various functional states of the oral cavity:

a) rest: the facial muscles are relaxed, the jaws and teeth are open, the lower jaw is slightly lowered;
b) opened mouth: jaws is wide opened;
c) closed mouth: jaws is tightly closed;
d) chewing a standard chewing gum;
e) maximum compression of the jaws through the standard chewing gum.

### PROTOCOL

1. Draw an EMG in different conditions. 2. In the conclusion, evaluate the results of the study of electrical activity of the masticatory muscles in various conditions.

| Recorded EMG from muscles | rest | open mouth | closed mouth | chewing |
|---|---|---|---|---|
| masseter | | | | |
| digastric | | | | |

2. **Conclusion**. At rest, the electrical activity of the masseter and digastric muscles is ___ (↓ or ↑). With an open mouth increases electrical activity in the _____ muscle. When the mouth is closed, electrical activity in the _____ muscle increases. When chewing, the frequency and amplitude of the impulses in the studied muscles _____ (↓ or ↑).

## WORK 5.3. STUDYING MANDIBULAR MOVEMENTS IN DIFFERENT PLANES. GOTHIC ARCH MANDIBULAR MOVEMENTS

In lateral movements, the condyle appears to rotate with a slight lateral shift in the direction of the movement. This movement is called the **Bennett movement** and may have both immediate and progressive components. By the use of recording equipment such as a pantograph or kinesiograph, it is possible to record mandibular movements in relation to a particular plane of reference (e.g., sagittal, horizontal, or frontal planes). If a point (the incisive point) located between the incisal edges of the two mandibular central incisors is tracked during maximal lateral, protrusive, retrusive, and wide opening movements, such movements are seen to take place within a border or envelope of movements. Functional and parafunctional movements occur within these borders. However, most functional movements such as those associated with mastication occur chiefly around centric. Border movements in the horizontal plane are shown in fig. 5.4.

At fig. 5.4. right mandibular movement with schematic representation of movement at the incisal point in the horizontal plane *(CR, LL, P, RL)* and at the condyle *(W, C, B, P)* made by a pantograph are presented. Teeth are not in occlusion. *CR*, Centric relation; *LL*, left lateral; *P*, protrusive; *RL*, right lateral; *CO*, centric occlusion; *IEC*, incisal edge contact. On the right side, the condyle moves from *C* (centric) to right working *(W).* On the balancing side, the left condyle moves from *C* along line *B* and makes an angle *BG*, called the *Bennett angle. C* to *P*, Straight protrusive movement.

The ***maximum opening movement*** is 50 to 60 mm, depending on the age and size of the individual. An arbitrary lower limit for normal of 40 mm may be in error, inasmuch as some individuals may have no difficulty incising a large apple and have no history of TMJ muscle dysfunction. The ***maximum lateral movement*** in the absence of TMJ muscle dysfunction, including pain, is about 10 to 12 mm. The ***maximum protrusive movement*** is approximately 8 to 11 mm, again depending on the size of the subject and skull morphology.

**Materials and equipment:** millimeter ruler.

**Accomplishment.** Suggest to the tested person to open his mouth as wide as possible. Measure the distance between the upper and lower incisors with an accuracy of 1 mm. Normally, it is 40–60 mm. Ask the subject to insert between the incisors 3 middle fingers of his non-working hand. Normally, with the maximal lowering of the mandible, the distal phalanges of the 3 middle fingers should fit between the incisors of the jaws. Ask the tested person to describe the "**gothic arch**" with the lower jaw, first without contact, and then in contact with the teeth of the upper jaw. In the case of the normal function of the chewing system, the mandible evenly (right to left or left to right) describes the "gothic arch" within the scope of its movements both without contact and in contact with the teeth of the upper jaw.

**Directions for recording the Protocol**:

1. Measure the distance between the incisors of the upper and lower jaw at the maximum opening of the mouth.
2. Make a visual assessment of whether the mandible of the tested person describes the "gothic arc" while moving.
3. Make a conclusion about the amount of movement of the lower jaw.

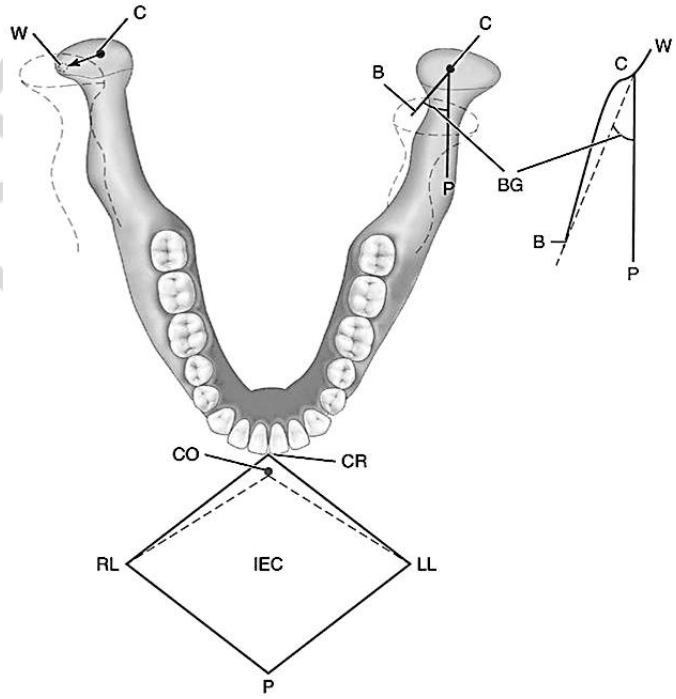



Fig. 5.4

### PROTOCOL

1. The distance between the incisors of the upper and lower jaw at the maximum opening of the mouth is _____ mm (normal range is from _____ to_____ mm).

2. During the movement of the mandible, the "gothic arch" is described _____ (completely or interrupted).

3. **Conclusion**. The amount of movements of the mandible of the tested person is _____ (full or limited).

## WORK 5.4. STUDYING THE OCCLUSION (IN DENTISTRY)

**Occlusion**, in a dental context, means simply the contact between teeth. More technically, it is the relationship between the maxillary (upper) and mandibular (lower) teeth when they approach each other, as occurs during chewing or at rest.

**Malocclusion** is the misalignment of teeth and jaws, or more simply, a "bad bite". Malocclusion can cause number of health and dental problems. **Static occlusion** refers to contact between teeth when the jaw is closed and stationary, while **dynamic occlusion** refers to occlusal contacts made when the jaw is moving, as with chewing. **Centric occlusion** is the occlusion of opposing teeth when the mandible is in centric relation. Centric occlusion is the first tooth contact and may or may not coincide with maximum intercuspation. It is also referred to as a person's habitual bite, bite of convenience, or intercuspation position (ICP). *Centric relation*, not to be confused with *centric occlusion*, is a relationship between the upper and lower jaw.

### MANDIBULAR POSITIONS

Basic jaw positions are usually described as **centric occlusion, intercuspal position, centric relation**, retruded contact position, and rest position of the mandible. *Centric occlusion* or *intercuspal position* is defined as maximum intercuspation of the teeth. Centric relation is a position of the mandible (or path of opening and closing without translation of the condyles) in which the condyles are in their uppermost position in the mandibular fossae and related anteriorly to the distal slope of the articular eminence. Because the mandible appears to rotate around a transverse axis through the condyle in centric relation movement, guidance of the jaw by the clinician in opening and closing movements that do not have translation is referred to as **hinge axis movement**. In this position, the condyles are considered to be in the terminal hinge position. Under physiological conditions of the masticatory system, centric relation is used to transfer the position of the mandible (in relation to the maxilla) to an articulator. Figure 5.5 schematic representation of mandibular movement envelope in the sagittal plane. *CR*, Centric relation; *CO*, centric occlusion; *F*, maximum protrusion; *R*, rest position; *E*, maximum opening; *B* to *CR*, opening and closing on hinge axis with no change in radius *(r)*. In the natural dentition, centric occlusion is, in the majority of people, anterior to centric relation contact on the average by approximately 1 mm. Centric occlusion (or **acquired** or **habitual centric** as it is sometimes called) is a tooth-determined position, whereas centric relation is a jaw-to-jaw relation determined by the condyles in the fossae. Closure into occlusion occurs usually anterior to centric relation; however, a coincidence of centric relation contact and the intercuspal position is evident in about 10 % of the population.

Rest position is a postural position of the mandible determined largely by neuromuscular activity and to a lesser degree by the viscoelastic properties of the muscles. Thus, because tonicity of muscles may be influenced by the central nervous system as a result of factors such as emotional stress and by local peripheral factors such as a sore tooth, the rest position of the mandible is not consistent.

The **interocclusal space** with the mandible in rest position and head in upright position is about 2 to 4 mm at the incisors but has considerable normal variance even from 1 up to 8 to 10 mm without evidence of dysfunction.

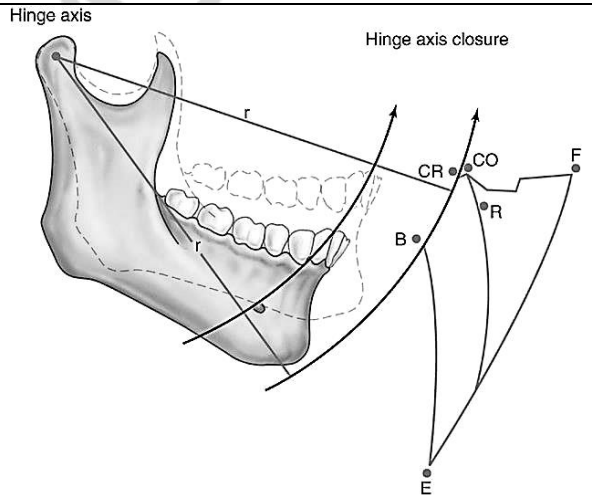



Fig. 5.5

**Materials and equipment:** pencil (or handle), ruler, caliper.

**Accomplishment.** Mark two points on the skin, one at the tip of the nose, the other – on the chin along the midline of the face. Ask the tested person to sit up straight, close his lips and completely relax the muscles of the face. With complete relaxation of muscles, the lower jaw occupies the physiological **rest position**. Using a caliper measure the distance between the marked points. It is the **centric occlusion**. Then ask the subject to clench the teeth. Measure the distance between the same points on the skin. It is the **centric occlusion**. The difference between the **rest position** and a **centric occlusion** is **interocclusal space**.

1. Indicate values of the **rest position** and **centric occlusion**.
2. Calculate the magnitude of the **interocclusal space**.
3. Make a conclusion about the size of the **interocclusal space**.

### PROTOCOL

1. **Rest position** _____ mm; **centric occlusion** _____ mm.
2. **Interocclusal space =** _____ - _____ = _____ mm.
3. **Conclusion**. The size of the **interocclusal position** is _____ (normal, increased, reduced).

## WORK 5.5. SMOOTH MUSCLES. CONTRACTION OF SMOOTH MUSCLE

Mechanism and characteristics of contraction:

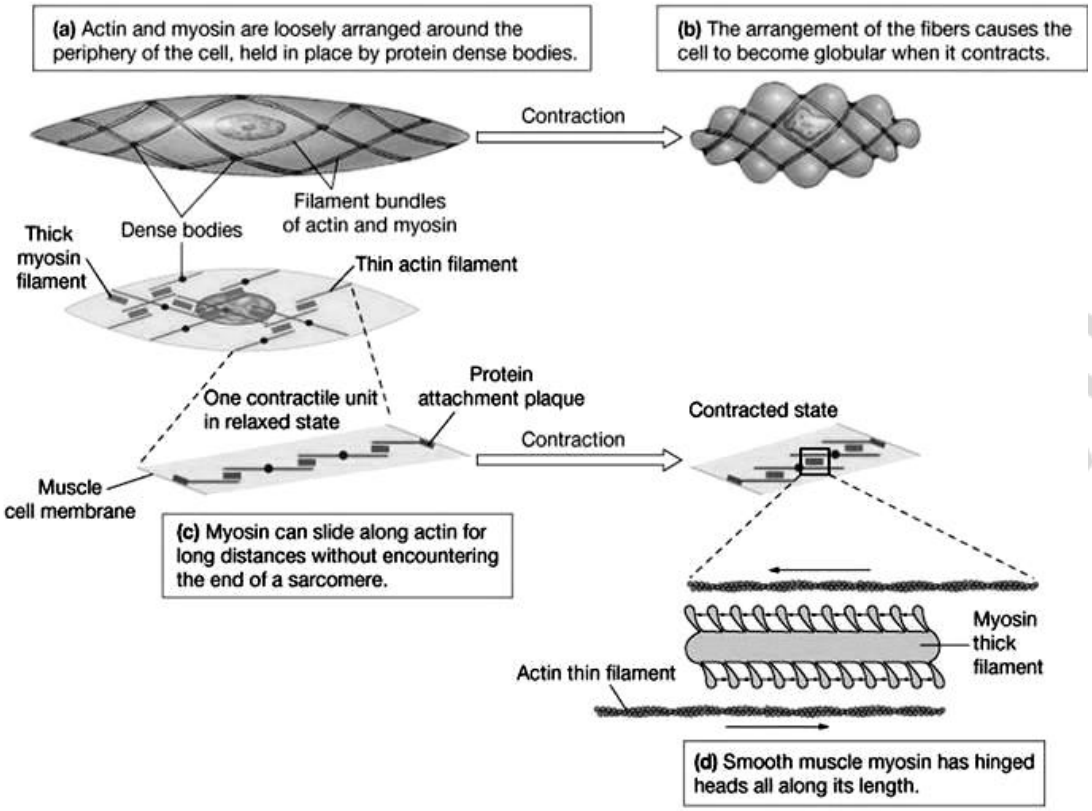



Fig. 5.6

*Main pecularetіes of SM contraction*: 1. _____ 2. _____
3. _____ 4. _____ 5. _____

## WORK 5.6. TYPES OF SMOOTH MUSCLE

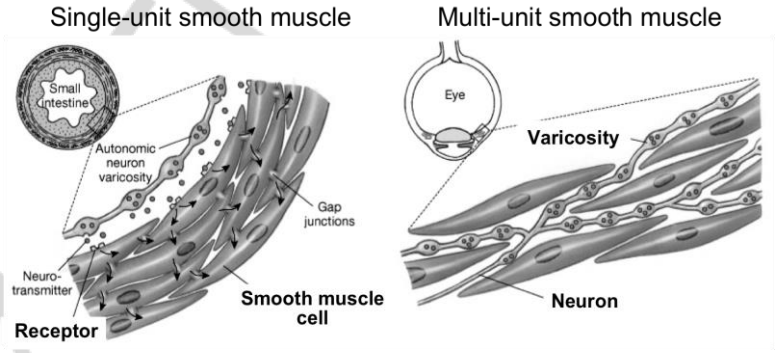



Fig. 5.7

*Write an examples of tissues formed from*:
1. Single-unit SM: _____
_____
2. Multi-unit SM: _____

## WORK 5.5. REGULATION OF SMOOTH MUSCLE CONTRACTION

*List the factors that may cause the contraction of a smooth muscle*:
1. _____
_____
2. _____
_____
3. _____

THE LABORATORY WORKS ARE PASSED WITH MARK

_____
Teacher's signature

## Lesson 6. PHYSIOLOGY OF THE NERVOUS SYSTEM. EXCITATION AND INHIBITION IN CNS. REFLEXES. GENERAL PRINCIPLES OF CNS ACTIVITY COORDINATION

DATE OF CLASSES
«____» _____ 20____
day month year

| **Basic questions:** | LITERATURE |
|---|---|
| 1. Nervous system and its role in ensuring the vital activity of the organism. Modern methods of investigation the functions of central nervous system.<br>2. Neuron: classification, structure, functions, properties, interactions with glial cells. Neuroglia functions. Cerebrospinal fluid: formation, composition, properties and functions.<br>3. The structure, functions and properties of nerve centers and nuclei. Their tone.<br>4. Reflex principle of the nervous system functioning. Types of reflexes. The structure of the reflex arch (somatic reflex). Feedback and its role.<br>5. Excitatory and inhibitory neurotransmitters, receptor mechanisms of their functioning.<br>6. Basic principles of propagation of excitation in the central nervous system. Interaction of excitation and inhibition processes. The concept of neuron integrative function.<br>7. Inhibition processes in the central nervous system, its manifestation forms and role. Classification: primary, secondary, their types. Concept of central inhibition mechanisms.<br>8. The basic principles of CNS activity coordination: convergence and divergence, reciprocal inhibition, feedback, final common pathway, dominant, subordination, plasticity. | Main<br>1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 54–79.<br>Additional<br>3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 123–130, 137–155.<br>4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 577–580, 595–606, 790–793. |

| **BUZZWORD** | Inhibition — |
|---|---|
| Nerve nuclei — | Coordination in CNS — |
| Nerve center — | Liquor — |
| Reflex — | Liquor composition — |
| Feedback — | Liquor functions — |

**WORK 6.1. CLASSIFICATION OF NEURONS**

| *Draw and indicate main types of neurons* | | |
|---|---|---|
| 1. | 2. | 3. |

## WORK 6.2. EXCITATORY AND INHIBITORY NEUROTRANSMITTERS, RECEPTOR MECHANISMS OF THEIR FUNCTIONING

*Fill in the table*:

| Classic excitatory neurotransmitters | Receptor types | Principle mechanisms of their functioning |
|---|---|---|
| | | |
| | | |

| Classic inhibitory neurotransmitters | Receptor types | Principle mechanisms of their functioning |
|---|---|---|
| | | |
| | | |

## WORK 6.3. THE STRUCTURE OF THE REFLEX ARCH

**Accomplishment**. The work is performed by the student independently while preparing for the lesson and is checked during the lesson.

**Directions for recording the Protocol:**

1. Draw somatic monosynaptic and polysynaptic reflex arch schemes (table 6.2).

2. Describe the five links of the reflex arch (table 6.1).

3. Indicate numbers of corresponding elements of reflex arch at picture in the table 6.1

*Table 6.1*

| Part | The full name of element |
|---|---|
| 1 | |
| 2 | |
| 3 | |
| 4 | |
| 5 | |

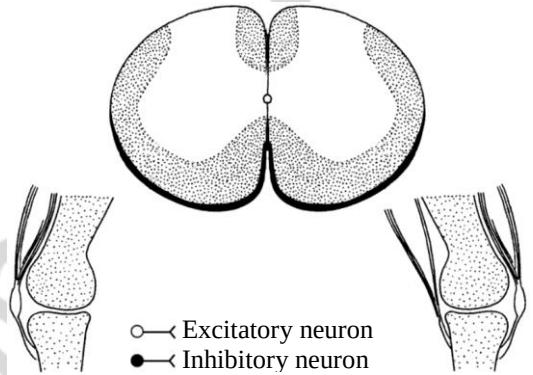
4. Fill in the table 6.2.

*Table 6.2*

**PROTOCOL**

Somatic monosynaptic reflex arch

Somatic polysynaptic reflex arch




| **Reflex arch elements of a somatic monosynaptic reflex:** | **Reflex arch elements of a somatic polysynaptic reflex:** |
|---|---|
| 1. Receptor part is presented by the following receptors of skeletal muscles: 1.1 | 1. Receptor part is presented by the following receptors of: 1.1 ; 1.2 |
| 2. Afferent part is presented by _____, its bodies are located in | 2. Afferent part is presented by _____, its bodies are located in |
| 3. Intercalated neuron | 3. Intercalated neuron |
| 4. Efferent part is presented by ____ or ___motor neurons, which are located in | 4. Efferent part is presented by ____ or ___motor neurons, which are located in |
| 5. Target organs are _____ and muscular fibers of skeletal muscles. | 5. Target organs are _____ muscular fibers of skeletal muscles. |
| Signal transmission rate (velocity of action potential [AP] propagation) is from _____ m/sec to _____ m/sec in efferent fibers, as they have _____ sheath and are referred to the type _____ | |
| Neurotransmitter in neuromuscular synapse is _____, that acts upon _____ type of _____-receptors. | |

**WORK 6.4. STUDYING OF A KNEE (TENDON) REFLEX**

Tendon reflexes participate in regulation of muscle tone and support of the body posture. In clinical practice tendon reflexes are studied to determine the functional state of different parts of the reflex arch and for the topic diagnosis of some CNS diseases.

**Materials and equipment**. A percussion hammer.

**Accomplishment**. A knee jerk reflex examination.

The examined person should sit down on the chair and put one his leg on the other. Hit the tendon of a quadriceps muscle of the hip below the patella with the percussion hammer. Observe the extension movement of the leg in the knee joint. Compare the reflex reaction on both extremities.

**Directions for recording the Protocol**:

1. Evaluate the expression degree of the reflexes, their symmetry.
2. Make a conclusion about the state of reflex reaction.

**PROTOCOL**

1. Knee reflex is _____ (*marked, absent*) on _____ (*one or both extremities*).
2. **Conclusion**: the reflex reaction is _____ _____ (*in norm, asymmetric, absent*)

*A knee jerk reflex*. This is an example of a monosynaptic stretch reflex.

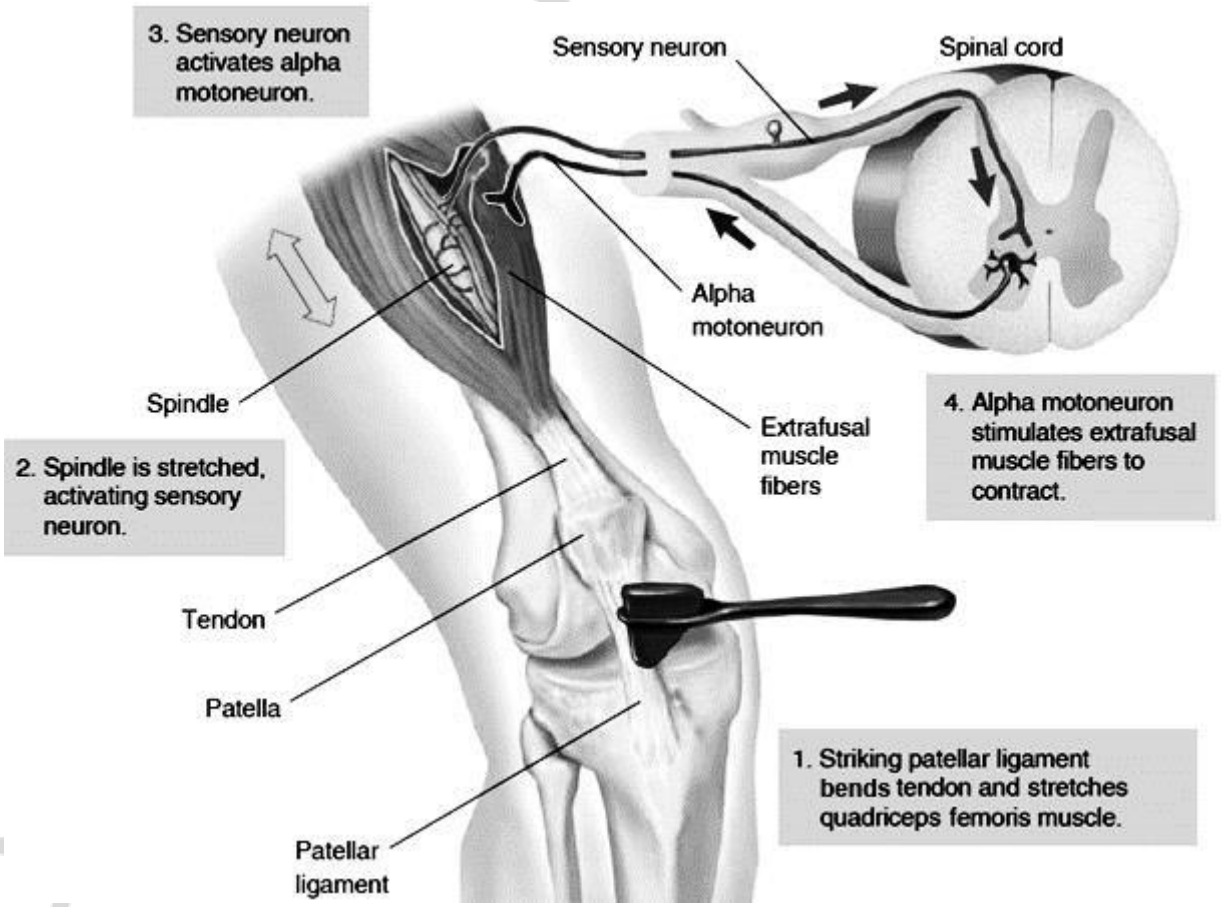



Fig. 6.1

## WORK 6.5. THE STUDY OF RECIPROCAL INHIBITION OF MOTOR REACTIONS BY ELECTROMYOGRAPHY

Electromyography is a recording method of total bioelectric activity of the muscle. Electromyogram (EMG) is a graphic record of the electrical activity of a muscle as recorded by an electromyography. Its reflects the tone state of the muscle at rest and its functional activity during contraction.

An electromyogram is made, when a person is awake and at rest, it having the character of continuous frequent oscillations with a very low amplitude (from 5 to 10 μV). When the contraction and tension are weak, an increase of electric activity is observed reaching its maximum in voluntary contraction (oscillation amplitude may reach 1000-2000 μV, oscillation frequency – 100 Hz).

Electromyographic studies are used in clinical practice, physiology of labor and sport.

**Materials and equipment**: superficial (cutaneous) electrodes, an electromyograph or an electroencephalograph for EMG recording; a set of weighs from 0.5 to 2 kg.

**Accomplishment**. Electrodes (bipolar) are applied to the arm skin of the examined in the region of biceps and triceps, they are attached to the electromyograph.

EMG is recorded under various conditions:

a) at rest;

b) the arm is flexed at the elbow;

c) the arm is extended;

d) at synergic tension of the arm biceps and triceps produced by increasing the load.

In the last case the examined person is standing with his hands down free. Then the examined person bends his elbow so that the forearm is in a horizontal position. Put weighs on the palm of the examined increasing their weight, e.g. 0.5, 1 and 2 kg and asking the tested person to keep the forearm horizontally.

**Directions for recording the Protocol**:

1. Draw an EMG recorded during the experiment.
2. Compare the character of EMG under various conditions (amplitude and frequency of impulses) visually.
3. Make a conclusion about the state of the motor center activity that innervates the shoulder biceps under the experiment.

### PROTOCOL

**1.** EMG drawing of the biceps under various conditions

| EMG recording from the muscle | Rest | Arm bending | Arm extension | Under tension (holding the load) |
|---|---|---|---|---|
| biceps | | | | |
| triceps | | | | |

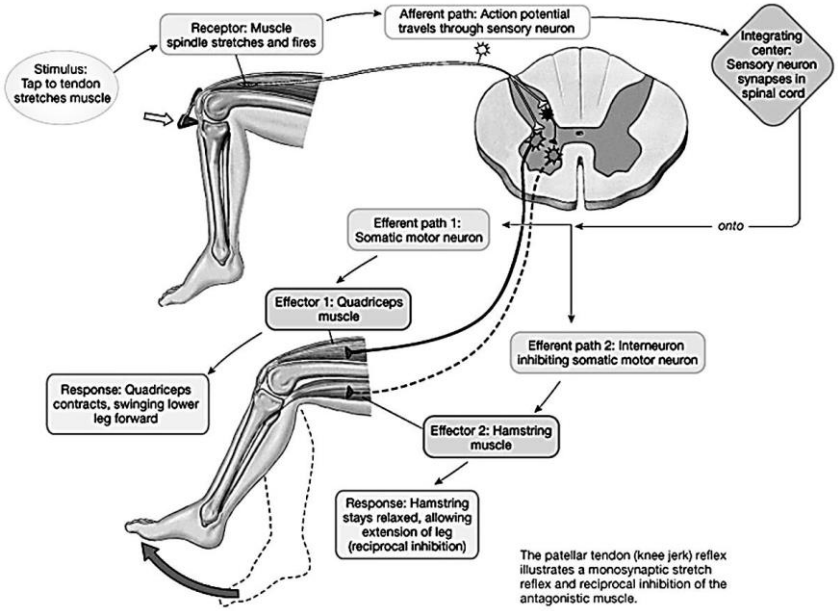
**2. Conclusion**: electric activity of the shoulder biceps and triceps muscles and that of nerve centers innervating it, under experiment (while bending the arm at the elbow and particularly in additional tension of the muscle for holding the weighs) versus the state of rest is considerably

_____

(*increased or reduced*), it being testified by

_____

(*increase or decrease*) of amplitude and frequency of EMG waves.



**WORK 6.6. STUDYING THE BASIC PRINCIPLES OF CNS ACTIVITY COORDINATION**

| List the basic principles of coordination | Description |
|---|---|
| 1. | |
| 2. | |
| 3. | |
| 4. | |
| 5. | |

**WORK 6.7. STUDYING THE CENTRAL INHIBITION MECHANISMS (use the lecture or E-learning materials)**

| Write a name of the main mechanism of central inhibition | Explain a basic mechanisms of inhibition |
|---|---|
| 1. | |
| 2. **Secondary** | |

**Central inhibition**

*Fill in the table. Write a classification and draw diagrams. Use the lecture or E-learning materials*

| **Primary** | | | |
|---|---|---|---|
| [illegible] | postsynaptic | | |
| *picture*: | *picture*: | *picture*: | *picture*: |
| | | | |
| *picture*: | *picture*: | *picture*: | *picture*: |

THE LABORATORY WORKS ARE PASSED WITH MARK

Teacher's signature

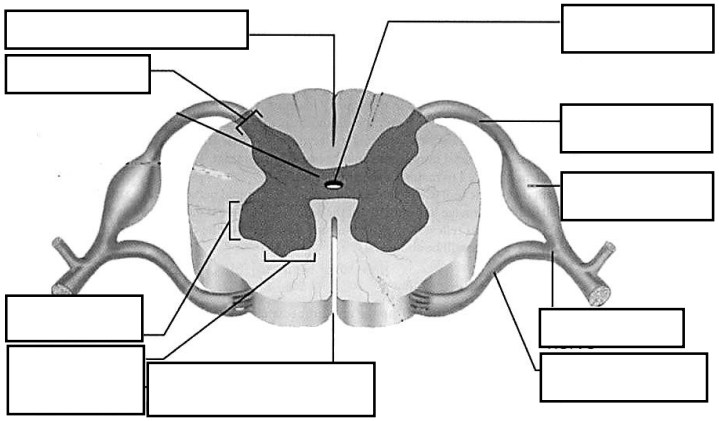
## Lesson 7. COLLOQUIUM "EXCITABLE TISSUES"

DATE OF CLASSES
«____» _____ 20____
day month year

Example of the ticket for writing part:

| BSMU | DEPARTMENT OF NORMAL PHYSIOLOGY | NAME: | | | |
|---|---|---|---|---|---|
| Normal physiology | Version 58 | DATE: | | | |
| **Colloquium "EXCITABLE TISSUES"** | | | | | |

The structure and function of the spinal cord. Indicate structures.



Dinamometry.

Muscle strength:
Right hand – 39
Left hand – 35
Back muscles – 112
Body mass – 68 kg
Sex – male

$HSI_r$ = ---------- = _____

$HSI_l$ = ---------- = _____

BSI = ---------- = _____

Conclusion:

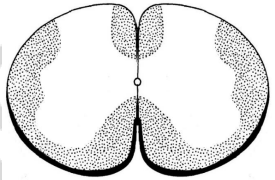
Describe the mechanisms of conduction in CNS synapses:

1.
2.
3.
4.
5.
6.
7.
8.

Write the function of the following structures:

| Structure | Function |
|---|---|
| Myofibril | |
| Triad | |
| T-tubule | |
| Sarcoplasmic reticulum (CR) | |
| Terminal cisternae | |

Somatic polysynaptic reflex arch diagram.
Draw it and fill in the table



Reflex arch parts of somatic polysynaptic reflex:
1. Receptor part is presented by the following receptors of:
1.1 _____; 1.2 _____
2. Afferent part is presented by _____, its bodies are located in _____
3. Intercalated neuron _____
4. Efferent part is presented by _____ or _____ motor neurons, which are located in _____
5. Target organs are _____ muscular fibers of skeletal muscles.
Signal transmission rate (velocity of action potential [AP] propagation) is from _____ m/sec to _____ m/sec in efferent fibers, as they have _____ sheath and are referred to the type _____
Neurotransmitter in neuromuscular synapse is _____, that acts upon _____ type of _____-receptors.

**Basic questions**:

1. Physiology as a scientific basis of medicine. The significance of human physiology for dentists.
2. Modern concept of the structure and functions of the cell membranes. Mechanisms of transport of a substances across the cell membrane.
3. Concepts: information, signal. Types of signals.
4. The concept of cellular (molecular) receptors and its functions. The receptor mechanisms of signals perception. Basic ways of signal transmission.
5. General properties of excitable tissues. Excitation and forms of its manifestation.
6. Indicators (parameters) excitability. Electrodontodiagnostics (electric dental pulp test), its use in dentistry.
7. Biopotentials, their types. Resting membrane potential, its origin. Galvanism.
8. The action potential (AP). Phases and ion mechanisms of AP generation. Excitability alteration during AP.
9. Basic laws of excitable tissues response to the stimulus action. Chronaximetry, its use to study the excitability of muscles and nerves.
10. Sensory receptors: definition, classification, functions, basic properties. Receptor and generator potentials. Basic principles of information coding in sensory receptors.
11. Neuron: structure, function, properties, interaction with glial cells. The role of glial cell.
12. Nerve fibers: structure, classification, function.
13. Mechanisms and laws of excitation conduction by myelinated and unmyelinated nerve fibers. Physiological basis of conductive anesthesia in dental practice.
14. Synapses: structure, classification, functions. Functional properties of synapses.
15. The mechanisms of excitation conduction in synapses. Excitatory neurotransmitters. EPSP. Inhibitory synapses, its neurotransmitters. Ion mechanisms of IPSP. Summation.
16. Neuromuscular synapse: mechanisms of signal transmission. An End Plate Potential (EPP), its transformation into an action potential. Role of acetylcholinesterase.
17. Types of muscle fibers. Motor units, their types, structural and functional properties.
18. Physiological properties of skeletal muscles and their functions.
19. Structural and functional characteristics of muscle fiber. Sarcomere. Main proteins of myofilaments, their functions.
20. Mechanism of contraction and relaxation of a single muscle fiber and a whole muscle. Excitation-contraction coupling.

LITERATURE

Main


1. Lecture & E-learning system.
2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 7–82, 144–148, 635–646.
3. *Severina, T. G.* Physiology of blood. Lecture notes / T. G. Severina. 2nd ed. Minsk : BSMU, 2017. P. 14–18.


Additional


4. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016.
5. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016.


**Form of colloquium:**
***computer control test "Lesson 07" at E-learning system with oral or writing part.***

| | |
|---|---|
| 21. A single contraction of muscle fiber and its phases. Types and regimes of skeletal muscle contraction. Tetanic muscle contraction and its types.<br>22. Force and work of muscle contraction. Nature of muscle tone. Muscle fatigue.<br>23. Dynamometry of a hand and back muscles.<br>24. The concept of components of masticatory system and their functional interaction. Movement of mandible. Physiological occlusion. Gnathodynamometry.<br>25. Physiological properties and characteristics of smooth muscle. Smooth muscle tone.<br>26. Transmission of information from nerve fibers to smooth muscle. Neuroeffector connections of smooth muscle.<br>27. Central nervous system (CNS) and its role in ensuring the vital activity of the organism. The structure, functions and properties of nerve centers and nuclei. Their tone.<br>28. Reflex principle of the nervous system functioning. Types of reflexes. The structure of the reflex arch (somatic reflex). Feedback and its role.<br>29. Basic principles of propagation of excitation in the central nervous system. Interaction of excitation and inhibition processes. The concept of neuron integrative function.<br>30. Inhibition processes in the central nervous system, its manifestation forms and role. Classification: primary, secondary, their types. Concept of central inhibition mechanisms.<br>31. The basic principles of CNS activity coordination: convergence and divergence, reciprocal inhibition, feedback afferentation (P. K. Anokhin), final common pathway (C. S. Sherrington), dominance principle (A. A. Uhtomsky), subordination (corticalisation), plasticity. | |

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## SECTION "MECHANISMS OF FUNCTIONS REGULATION"

## Lesson 8. NERVOUS REGULATION OF SOMATIC FUNCTIONS

DATE OF CLASSES
«____» _____ 20____
day month year

**Basic questions:**

1. The concept of physiological function and its regulation. Levels of regulation. Types of regulation.
2. Nervous and humoral mechanisms of regulation of functions, their comparative characteristics.
3. The structure and functions of the spinal cord. Spinal reflexes.
4. The concept of the spinal level of muscle tone regulation. The consequences of spinal cord injury.
5. Functions of the medulla oblongata, pons and midbrain. Vital centers of the brain stem and their functions. Reticular formation, its functions.
6. The functions of the cerebellum. The consequences of the cerebellum injury.
7. Diencephalon. The functions of the thalamus and hypothalamus.
8. Modern concept of localization of function in the cerebral cortex. Functional asymmetry of the cortex.
9. Forebrain structures. The concept of the basal nuclei, limbic system, their functions. The role of dophamine and acetylcholine neurotransmitter systems.

LITERATURE

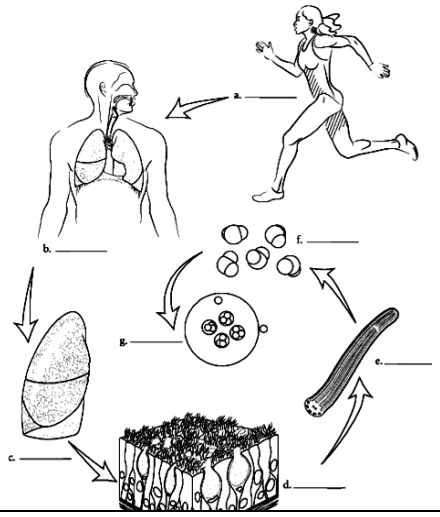
Main


1. Lecture & E-learning system.
2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 80–118.


Additional


3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 227–253, 263–273.
4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 6–10, 695–714, 721–745, 751–761.


### WORK 8.1. LEVELS OF ORGANIZATION




*Answer Key: a. Organism (human), b. Organ system (respiratory system) c. Organ (lung), d. Tissue (epithelium), e. Organelle (cilia), f Molecule, g. Atom, h. Cells*

### WORK 8.2. MECHANISMS OF REGULATION OF FUNCTIONS

| | | |
|---|---|---|
| – metabolites<br>– electrolytes<br>– neurohormones<br>– tissue hormones<br>– hormones | – somatic reflexes<br>– autonomic reflexes | – automaticity<br>– contraction after stretching<br>– plasticity |

*Answer Key: humoral, nervous, myogenic*

### BUZZWORD

Function — _____

Regulation — _____

Levels of regulation — _____

Types of regulation — _____

Reflex — _____

Vital centers — _____

Functional asymmetry — _____
_____

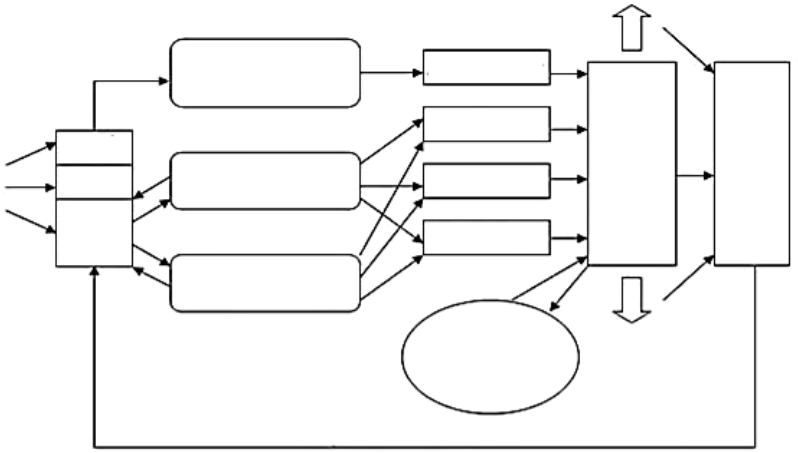
**WORK 8.3. MECHANISMS OF REGULATION OF FUNCTIONS, THEIR COMPARATIVE CHARACTERISTICS**

*Fill in the table*

| Indicator | Neural mechanism | Humoral mechanism |
|---|---|---|
| Regulation accuracy | | |
| Methods of communication | | |
| Speed of regulation | | |
| Duration of the regulation | | |

**WORK 8.4. THE GENERAL SCHEME OF THE FUNCTIONAL SYSTEM OF REGULATION OF FUNCTIONS "ON A DEVIATION"**

*Fill in the scheme*



**WORK 8.5. THE STRUCTURE AND FUNCTION OF THE SPINAL CORD. SPINAL REFLEXES**

The spinal cord is the simplest part of the central nervous system (CNS).

A. The spinal cord extends from the foramen magnum approximately to the level of the body of the $L_2$ vertebra in adults.

B. The spinal cord contains an inner core of gray matter that is completely surrounded by white matter (fig. 8.1).

C. The gray matter is divided into a dorsal horn and a ventral horn, which are separated by an intermediate zone; the gray matter of the spinal cord has a "butterfly" shape in transverse sections.

D. Dorsal horn contain the bodies of intercalated neuron, ventral horn — bodies of moto neuron, and intermediate zone (lateral horn) — neurons of autonomic nervous system.

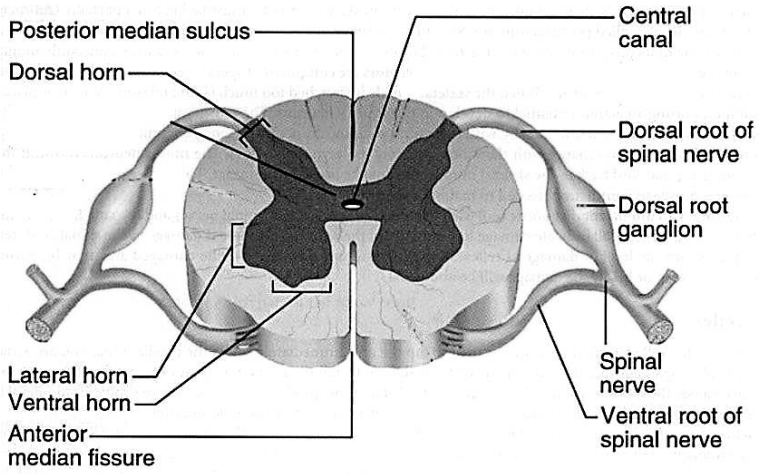


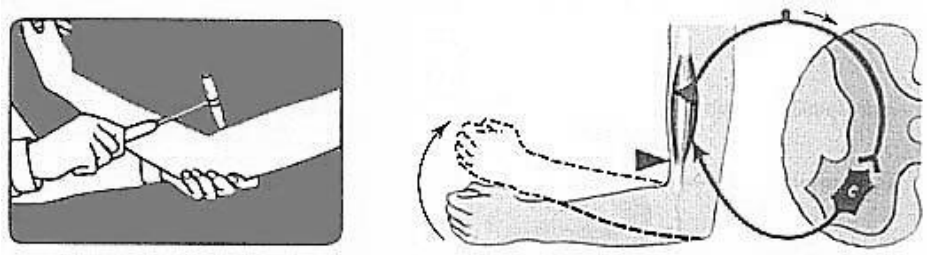

Fig. 8.1

**WORK 8.6. STUDYING THE STRETCH REFLEXES (OR MYOTATIC REFLEXES)**

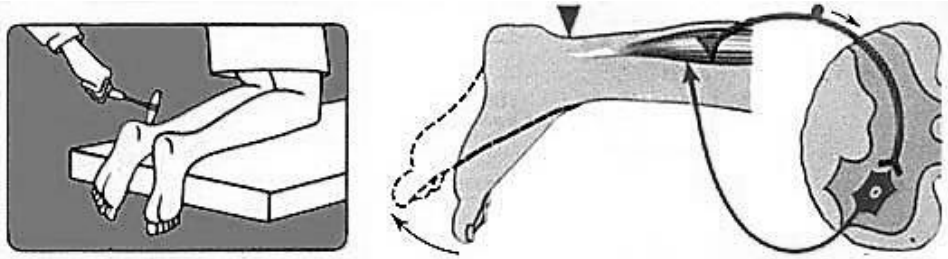
**Accomplishment**.

Write the names of reflexes shown at pictures and indicate spinal cord segments levels of a tendon reflexes moto neurons localization.

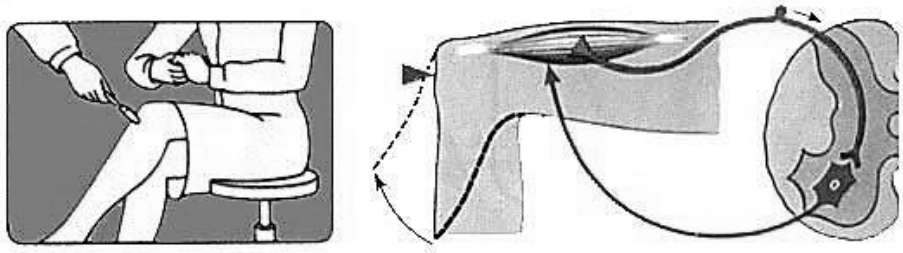
Study the reflexes like it shown at pictures. Evaluate the expression degree of the reflexes, their symmetry



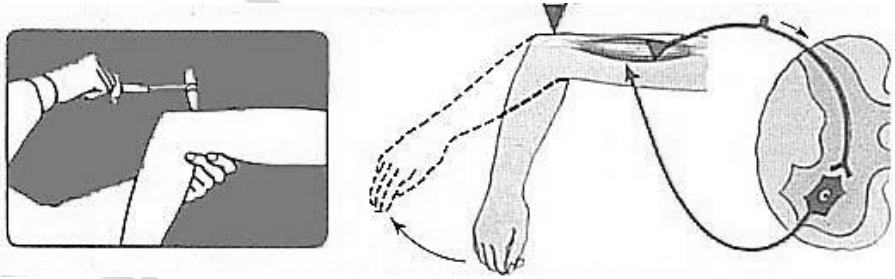
1. *Tendon flexion reflex of the upper extremity (elbow reflex), $C_4$-$C_5$*



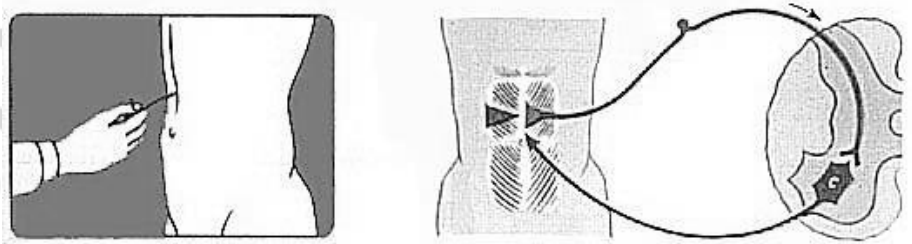
2. _____



3. _____



4. _____



5. _____

**PROTOCOL**

1. Reflexes are _____

*(marked or absent)*

and _____

*(symmetric or asymmetric)*

2. **Conclusion**: the reflex reactions is _____

*(in norm or impaired)*

## WORK 8.7. STUDY OF CEREBELLUM CONTROL OF MOTOR ACTIVITY

Efferent signals from the cerebellum regulate neuronal activity of vestibular (Deiters') and red nuclei, the thalamus nuclei, and through them the activity of peripheral (α- and γ-motor neurons of the spinal cord and nuclei of cranial nerves) and central (cortical) motor neurons. Through these pathways efferent signals from the cerebellum *regulate strength of muscle contractions* ensuring the ability for prolonged *tonic muscle contraction*, relate the *volume of a voluntary movement* with the distance to the aim of this movement, and *quickly change flexing to extending and vice versa*. The cerebellum provides the synergy of contractions in complex movements.

Cerebellum functions disorder is manifested by: decrease of muscle contraction force (asthenia); loss of the ability to prolonged muscle contraction that makes standing, sitting difficult (astasia); involuntary change of muscle tone (dystony); finger trembling at rest (tremor); movement impairment revealed as excessive or insufficient movement (dysmetry); coordination impairment (ataxia) that is manifested as "drunk" (swaying) gait and etc.; speech motor disorders (dysartria); swinging rhythmic twitching of eyeballs (nystagmus); impairment of alternating opposite movements (adiadochokinesis), etc.

**Directions for recording the protocol:**

1. Point out, if the examined succeeded correctly (without faults) to perform the offered tests.
2. Make a conclusion about the quality of the cerebellum control of motor activity.

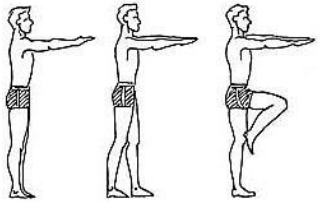
### PROTOCOL

1. The tests for ataxia in the examined were _____ (+ *or* –), as in Romberg's pose he _____ (*kept or didn't*) balance, his gait was _____ (*normal or impaired*); tests for dysmetry and tremor were _____ (+ *or* –); dysarthry _____ (*was or wasn't*) revealed.
2. **Conclusion**. The cerebellum control of motor activity in the examined was _____ (*in norm or impaired*)

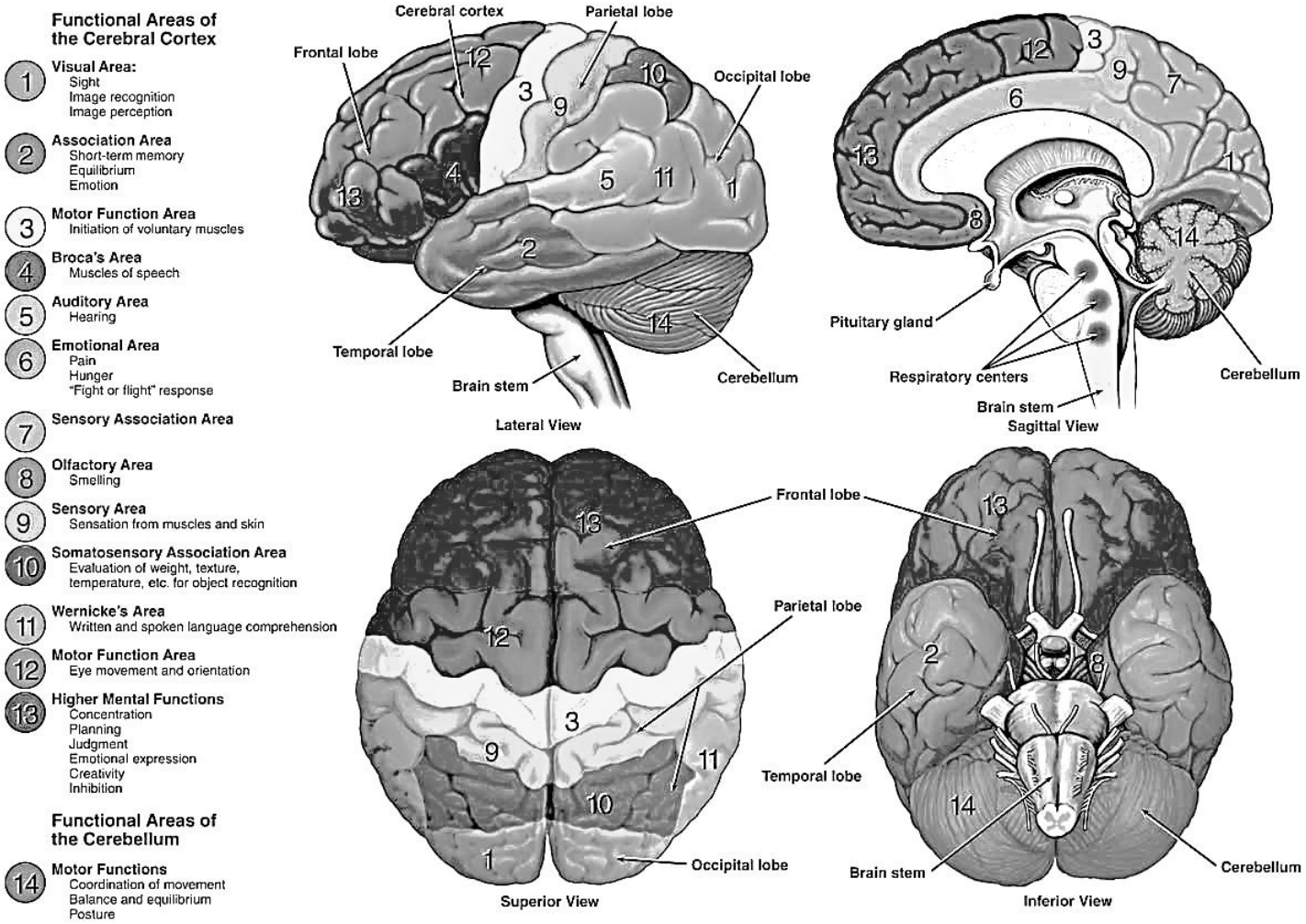
**Materials and equipment:** a glass, a book.

**Accomplishment.** The examined performs actions and exercises indicated in table 8.1. *Table 8.1*

**Cerebellum control of skeletal muscles motor activity**

| Type of experiment | Technique |
|---|---|
| Romberg's pose (coordination assessment of movements or ***abasia*** test) | The examined should stand with feet close and hands stretched forward, at first with open and then with closed eyes. In norm the person keeps the balance in Romberg's pose (i. e. the abasia test is negative)  |
| Gait (assessment of movements coordination or ***ataxia*** test) | Examined should walk about the room forward and backward with open and closed eyes. In norm the gate of a healthy person is usual, without swaying to the sides and broad placing his feet (i. e. the ataxia test is negative)   |
| ***Dysmetria*** test | The examined should take from the table and put back some object (a book, a glass). In norm the person puts the subject to the same place with an error ±2 cm (i. e. the dysmetry test is negative) |
| Speech (***dysarthria*** test) | The examined should repeat some words difficult for pronunciation (*adiadochokinesis*, *atrioventricular*, *deoxyhemoglobin* etc.). Note, if there is slowed down, irregular or discontinuous speech |
| Finger-nose test (for ***dysmetry*** and ***tremor***) | The examined should point with his index finger (at first of the left and then of the right hand) to the tip of his nose with open and closed eyes. In norm the person touches his nose tip with accuracy of ±1 cm without tremor of fingers (i. e. the test for dysmetria and tremor is negative). Persons having cerebellum disorder miss the nose tip and their fingers tremble while reaching the nose   |

## WORK 8.8. STUDYING THE FUNCTIONS OF THE CEREBRAL CORTEX

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## Lesson 9. NERVOUS REGULATION OF AUTONOMIC FUNCTIONS (PHYSIOLOGY OF AUTONOMIC NERVOUS SYSTEM)

DATE OF CLASSES
«____» _____ 20____
day month year

| **Basic questions**: | LITERATURE |
|---|---|
| 1. The role and functions of the autonomous nervous system (ANS).<br>2. Comparative characteristics of somatic and autonomic nervous system (sensory receptors, afferent, efferent, and intercalary divisions, effector organs).<br>3. Differences of neuroeffector connections of smooth muscle and neuromuscular synapses of skeletal muscle.<br>4. Comparative characteristics of the structure and neurochemical mechanisms of the sympathetic and parasympathetic parts of ANS, as well as their influence on the effector organs. Relative antagonism and synergism of their effect.<br>5. The concept of metasympathetic part of ANS.<br>6. Basic objective and subjective indices for evaluation the functional state of ANS parts.<br>7. The concept of the principles of the autonomic functions correction (for example, salivation) by affecting the neurotransmitter-receptor mechanisms in ANS ganglia and of the effector cells. | Main<br>1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 119–133.<br>Additional<br>3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 255–267.<br>4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 773–785. |

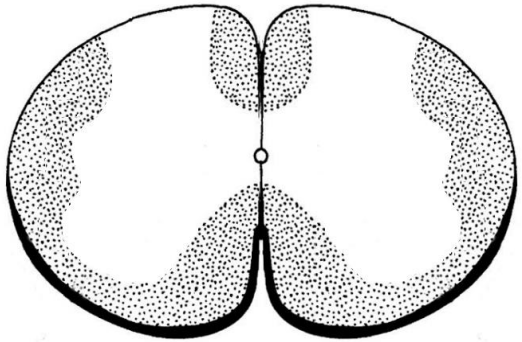
**BAZZWORD**

| | |
|---|---|
| Autonomous nervous system — | Major neurotransmitter of *sympathetic* postganglionic neurons and their receptors — _____, _____-_____receptors |
| Sympathetic nervous system — | Major neurotransmitter of *parasympathetic* postganglionic neurons and their receptors — _____, _____-_____receptors |
| Parasympathetic nervous system — | The peculiarities of ANS innervation of the adrenal glands medulla — |
| Metasympathetic nervous system — | The peculiarities of sweat glands innervation by ANS — |
| Autonomic ganglion — | Influence of *parasympathetic* part of ANS on salivation — |
| Neurotransmitter and receptor of ANS ganglia — | Influence of *sympathetic* part of ANS on salivation — |
| Antagonism — | Synergism — |

**WORK 9.1. DESCRIPTION OF SPINAL REFLEXES OF THE SYMPATHETIC AND SOMATIC NERVOUS SYSTEM**

**PROTOCOL**

Draw the scheme of **somatic** polysynaptic reflex arch **Autonomous (sympathetic)** reflex arch



| Indicate the reflex arch structural parts of a somatic reflex: | Indicate the reflex arch structural parts of a sympathetic reflex: |
|---|---|
| 1. Receptor part is presented by the following receptors of skeletal muscles: 1.1 _____; 1.2 _____ | 1. Receptor part is presented mainly by _____ receptors. |
| 2. Afferent link is presented by_____, which are located in _____ | 2. Afferent part is presented by _____, which are located in _____ |
| 3. Interneurons are located in _____ | 3. Interneurons are located in _____ |
| 4. Efferent part is presented by _____ or _____ motor neurons, which are located in _____ | 4. Efferent part consist of 2 neurons, which are located: $4^1$ in _____ _____ and $4^2$ in _____ respectively. |
| 5. Working organs. They are _____ and _____ muscle fibers of skeletal muscles. | 5. Working organs. They are _____ muscle cells; cardiomyocytes; gland cells, myoepitheliocytes. |
| 6. Signal (AP) transmission rate is from _____ m/sec to _____ m/sec in efferent fibers, as they have _____ sheath and are referred to the type _____ | 6. Signal (AP) transmission rate is from _____ m/sec to _____ m/sec in efferent postganglionic fibers, as they do not have _____ sheath and are referred to the type _____ |
| 7. Neurotransmitter of the neuromuscular synapse is _____, which binds to the _____ type of _____ receptors. | 7. Main neurotransmitter in neuroeffector junction is _____, which binds to the _____ and _____ types of _____ receptors. |

## WORK 9.2. CLINOSTATIC REFLEX

Reflex study allows determining the functional state of parasympathetic and sympathetic centers regulating the heart function. When a man passes from standing to lying position, the heart beat rate **decreases by 4–6 beats/min**. Pulse retardation over 6 beats/min evidences the increased tone of the parasympathetic part of ANS that regulates the heart functioning. The absence of reaction or its paradox character — pulse acceleration — evidences the prevalenced tone of the sympathetic part of ANS regulating heart functioning.

**Materials and equipment**: a couch, a stop-watch.

**Accomplishment**. At first the pulse of the examined is counted (per 15 sec and multiplied by 4), when he is standing. Then, in 10–25 seconds after the examined lay down, the pulse is again calculated in the same way.

**Directions for recording the Protocol**:

1. Put down the pulse rate (PR) in standing position and then in lying position, count the pulse difference.
2. Make a conclusion of the tone of the sympathetic and parasympathetic departments of ANS regulating the heart functioning of the examined.

**PROTOCOL**

| Pulse Rate, beats/min | | |
|---|---|---|
| In standing position | In lying | Pulse difference [PR lying – PR standing] |
| | | |

**Conclusion**: _____

## WORK 9.3. ORTHOSTATIC REFLEX

Reflex study allows determining the functional state of sympathetic and parasympathetic centers regulating the heart functioning. When a man passes from lying to standing position, the heart beat rate **increases normally 6–24 beats/min**. Pulse acceleration over 24 beats/min evidences the tone dominance of the sympathetic department of ANS, under 6 beats/min — that of the parasympathetic department of ANS.

**Materials and equipment**: a coach, a stop-watch.

**Accomplishment**. The pulse of the examined is counted when he is lying (the man is lying quietly for 4–6 min before the count starts). Then he is asked to stand up and his pulse is counted in 15–25 sec again.

**Directions for recording the Protocol**:

1. Put down the pulse rate (PR) in lying and standing position, calculate the pulse difference.
2. Make a conclusion of the tone of the sympathetic and parasympathetic departments of ANS regulating the heart functioning in the examined.

**PROTOCOL**

| Pulse Rate, beats/min | | |
|---|---|---|
| In lying position | In standing | Pulse difference [PR standing – PR lying] |
| | | |

**Conclusion**: _____

## WORK 9.4. HERING'S RESPIRATORY-CARDIAC REFLEX

Reflex study allows determining the functional state (tone) of the parasympathetic center regulating the heart functioning. When respiration is held on after a deep inhalation, the tone of *n. vagi* nuclei and heart beat rate **decreases normally by 4–6 beats/min**. Pulse retardation by 8–10 beats/min and over evidences the parasympathetic ANS part tone increase, under 4 beats/min — tone decrease.

**Materials and equipment**: a stop-watch.

**Accomplishment**. The pulse is counted when the examined is sitting, then he is asked to make a deep inhalation and hold on the breath and the pulse is counted again.

**Directions for recording the Protocol**:

1. Put down the pulse rate (PR) before the breath is held on and when breath is held on during inspiration. Calculate the pulse difference.
2. Make a conclusion about the tone of the ANS parasympathetic department regulating the heart function in the examined.

**PROTOCOL**

| Pulse Rate, beats/min | | |
|---|---|---|
| Before breath holding | During breath holding after inspiration | Pulse difference [PR breath holding – PR before BH] |
| | | |

**Conclusion**: the tone of the ANS parasympathetic department _____

**WORK 9.5. ASSESSMENT OF NEUROTRANSMITTER MECHANISMS OF THE EFFECT OF SYMPATHETIC AND PARASYMPATHETIC DEPARTMENTS OF ANS ON THE HEART FUNCTIONING (demonstrative computer work)**

**Accomplishment**. The program "**Physiol 2**" is used; it has allows to perform various virtual experiments on rats.

The description of work with the program is given in work 1.4.

For making the experiment in menu:

1. Choose Help → Preparation.
2. Help → Drugs
3. Drugs → Injected, or Stimulate
4. New Rat

**Directions for recording the Protocol:**

1. Make an experiment and analyze the data from the table of the protocol.

Abbreviations: HR — Heart Rate, $BP_{syst}$ — Systolic Blood Pressure, $BP_{diast}$ — Diastolic Blood Pressure, $BP_{mean}$ — Mean Hemodynamic Blood Pressure.

2. Make a conclusion about the effect of the ANS sympathetic and parasympathetic parts on the force of heart contraction and heart rate as well as about neurotransmitter mechanisms of these effects.

**PROTOCOL**

| Effects of the heart | | HR | $BP_{sys}$ | $BP_{diast}$ | $BP_{mean}$ |
|---|---|---|---|---|---|
| 1. | Initial values (baseline) | 161 | 98 | 53 | 66 |
| 2. | Stimulation Symp. Nerves to heart $T_1$ | 210 | 130 | 95 | 106 |
| 3. | New Rat + Injection of noradrenaline, 5 μg/kg | 212 | 130 | 95 | 106 |
| 4. | New Rat + Phentolamine(α-adrenoblocker), 100 mg/kg | 161 | 98 | 53 | 66 |
| 5. | New Rat + Phentolamine (α-adrenoblocker), 100 mg/kg + Stimulation Symp. Nerves to heart $T_1$ | 210 | 114 | 98 | 106 |
| 6. | New Rat + Propranolol (β-adrenoblocker), 100 mg/kg | 161 | 98 | 53 | 66 |
| 7. | New Rat + Propranolol (β-adrenoblocker), 100 mg/kg + Stimulation Symp. Nerves to heart $T_1$ | 170 | 99 | 65 | 75 |
| 8. | New Rat + Stimulation Vagus Nerve to heart | 112 | 42 | 30 | 40 |
| 9. | New Rat + Acetylcholine, 5 μg/kg | 115 | 31 | 19 | 28 |
| 10. | New Rat + Atropine (M-cholineblocker), 10.0 mg/kg | 161 | 98 | 53 | 66 |
| 11. | New Rat + Atropine (M-cholineblocker), 10.0 mg/kg + StimulationVagus Nerve to heart | 152 | 82 | 44 | 57 |

**Conclusion**: _____

_____

_____

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THE LABORATORY WORKS ARE PASSED WITH MARK

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Teacher's signature

## Lesson 10. HUMORAL REGULATION OF FUNCTIONS. PHYSIOLOGY OF THE ENDOCRINE SYSTEM. LESSON № 1

DATE OF CLASSES
«____» _____ 20____
day month year

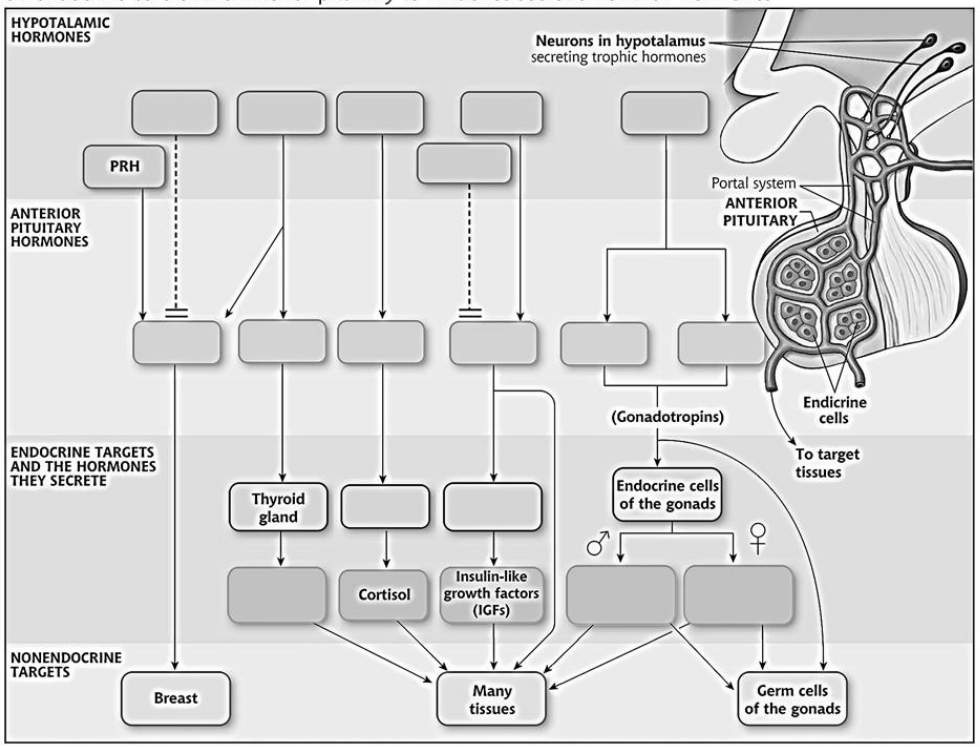
| **Basic questions**: | LITERATURE |
|---|---|
| 1. Endocrine system. Its role in the regulation of physiological functions.<br>2. The structures of the endocrine system (glands of internal secretion, diffuse elements) and its functions.<br>3. The concept of an autocrine, paracrine, endocrine and neuroendocrine.<br>4. Hormones, their chemical and functional classification, mechanisms of action. Basic ways of signal transmission. Second messengers.<br>5. Methods of investigation of the endocrine system functions in humans.<br>6. The pituitary gland, its connection with the hypothalamus.<br>7. Hormones of the pituitary gland (hypophysis) and hypothalamus, their role in the regulation of endocrine and not endocrine organs.<br>8. The concept of the endocrine function of pineal gland (melatonin).<br>9. Gonads. Male and female sex hormones and their physiological role. | Main<br>1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 134–154, 215–249.<br>3. *Severina, T. G.* Physiology of blood. Lecture notes / T. G. Severina. 2nd ed. Minsk : BSMU, 2017. P. 13–21.<br>Additional<br>4. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 299–335, 389–427.<br>5. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 925–950, 1021–1054. |

| **BUZZWORD** | |
|---|---|
| Endocrine system — | Statins — |
| Autocrine — | Tropic hormones — |
| Paracrine — | Effector hormones — |
| Endocrine — | LGIC — |
| Neuroendocrine — | 1 TMS receptor — |
| Circadian rhythm — | 7 TMS receptor — |
| Hormone — | First messenger — |
| Lipophilic hormones — | Second messengers — |
| Hydrophilic hormones — | Diabetes insipidus — |
| Liberins — | Acromegaly — |

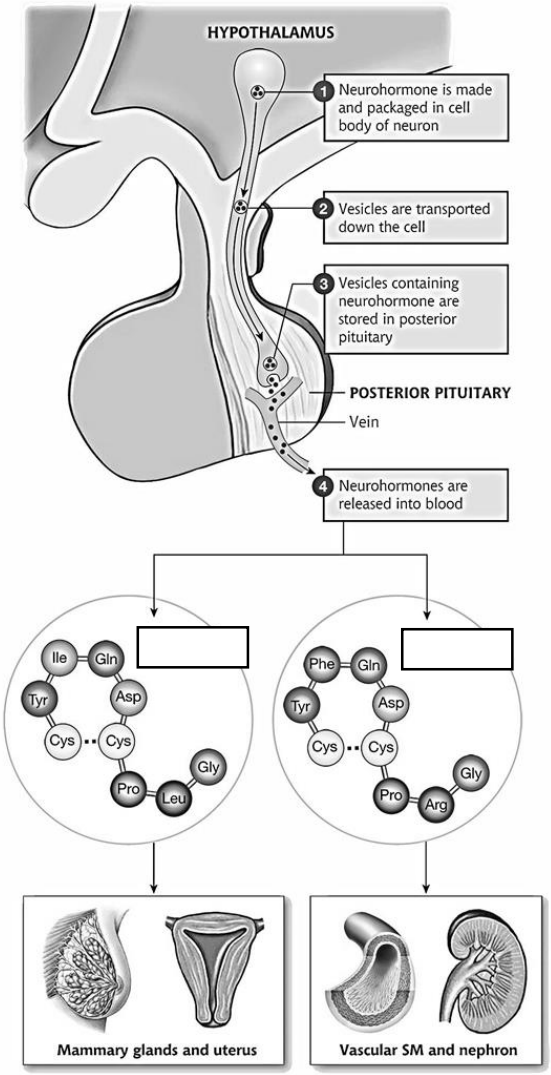
## WORK 10.1. STUDYING THE HORMONES OF THE HYPOTHALAMIC-PITUITARY SYSTEM

*Fill in the scheme (empty spaces). Use the materials of lectures, E-learning system, and text-book.*

The hypothalamus secretes releasing hormones (-RH) and inhibiting hormones (-IH) that act on endocrine cells of the anterior pituitary to influence secretion of their hormones




| ANTERIOR PITUITARY HORMONE | HYPOTALAMIC RELEASING HORMONE | HYPOTALAMIC INHIBITING HORMONE |
|---|---|---|
| | Prolactoliberin, Prolactin-releasing hormone (PRH) | Dopamine, prolactin-inhibiting factor (PIF) |
| Thyrotropin, Thyroid-stimulating hormone (TSH) | | |
| | Corticoliberin, Corticotropin-releasing hormone (CRH) | |
| | | Somatostatin, Growth hormone–inhibiting hormone (GHIH) |
| Gonadotropins (GTH): Follicle-stimulating hormone (FSH); Luteinizing hormone (LH) | | |



**WORK 10.2. STUDYING HORMONES OF ENDOCRINE SYSTEM AND THEIR ACTION ON TARGET CELLS (TISSUES, ORGANS)**

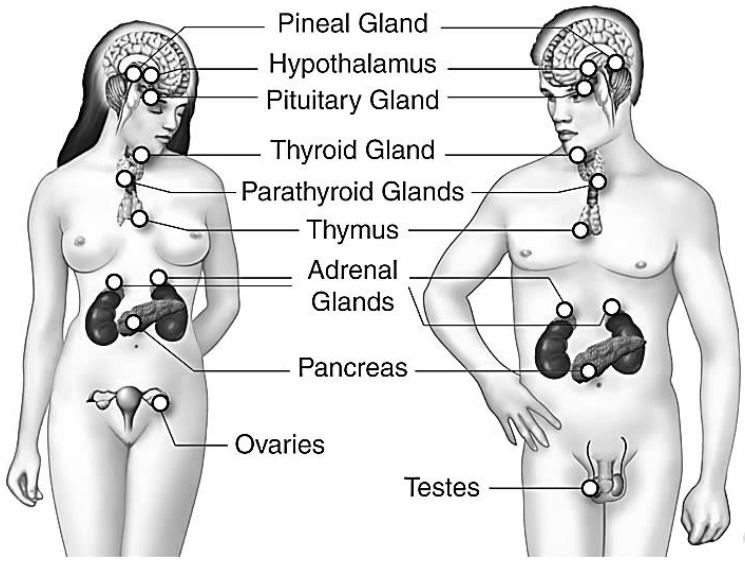



Fig. 10.1

*Write the names of the endocrine glands, shown in fig. 10.1, of the hormones secreted by them and the main functions of the hormones*

| Source (gland) | Hormone name | Chemical structure | Main function |
|---|---|---|---|
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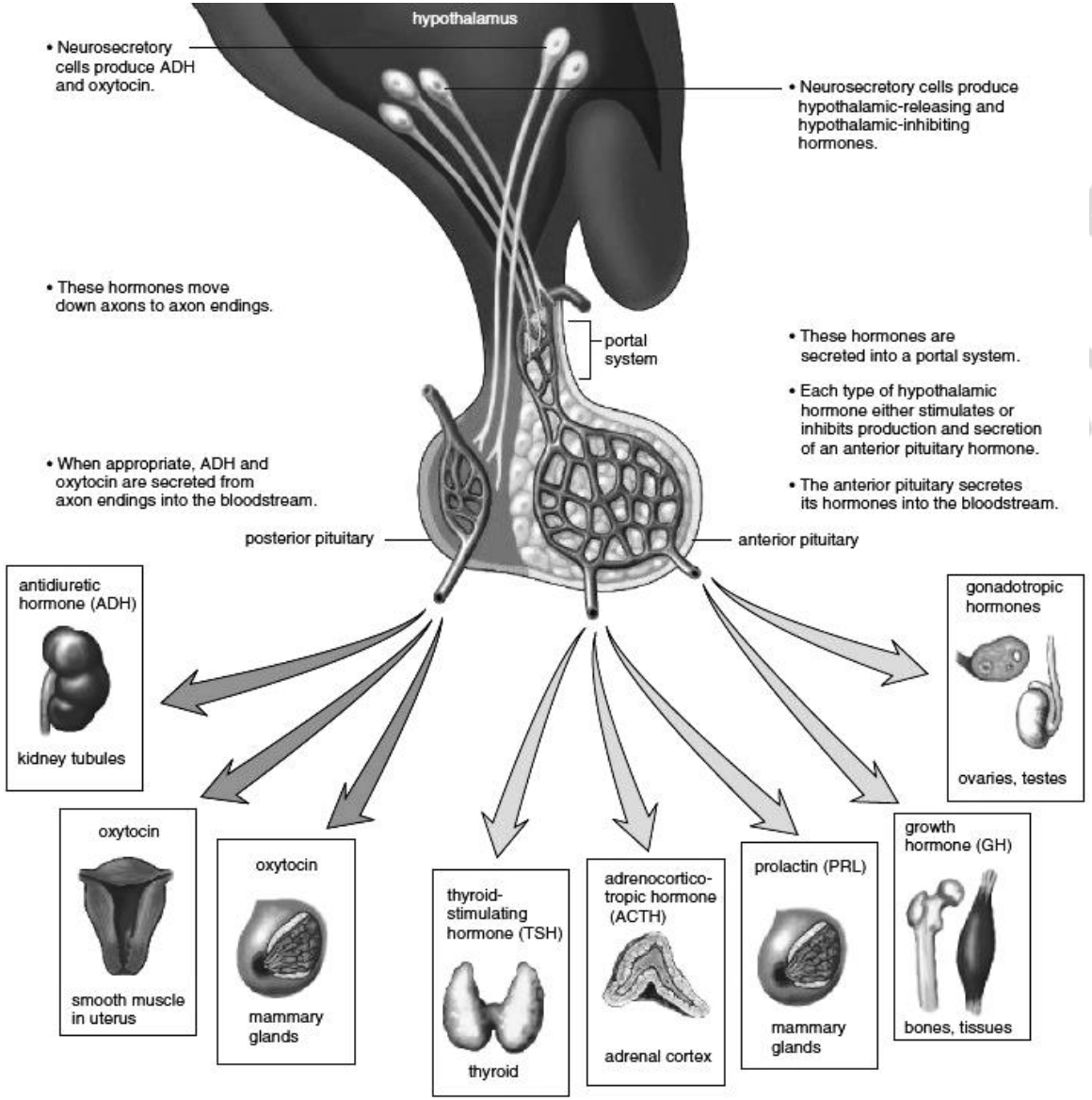


Fill in the table. Describe the manifestation of excess and insufficiency of main hormones *of hypotalamic-pituitary system*.

| Hormone | Function | Excess (hyper-function) | Insufficiency (hypo-function) |
|---|---|---|---|
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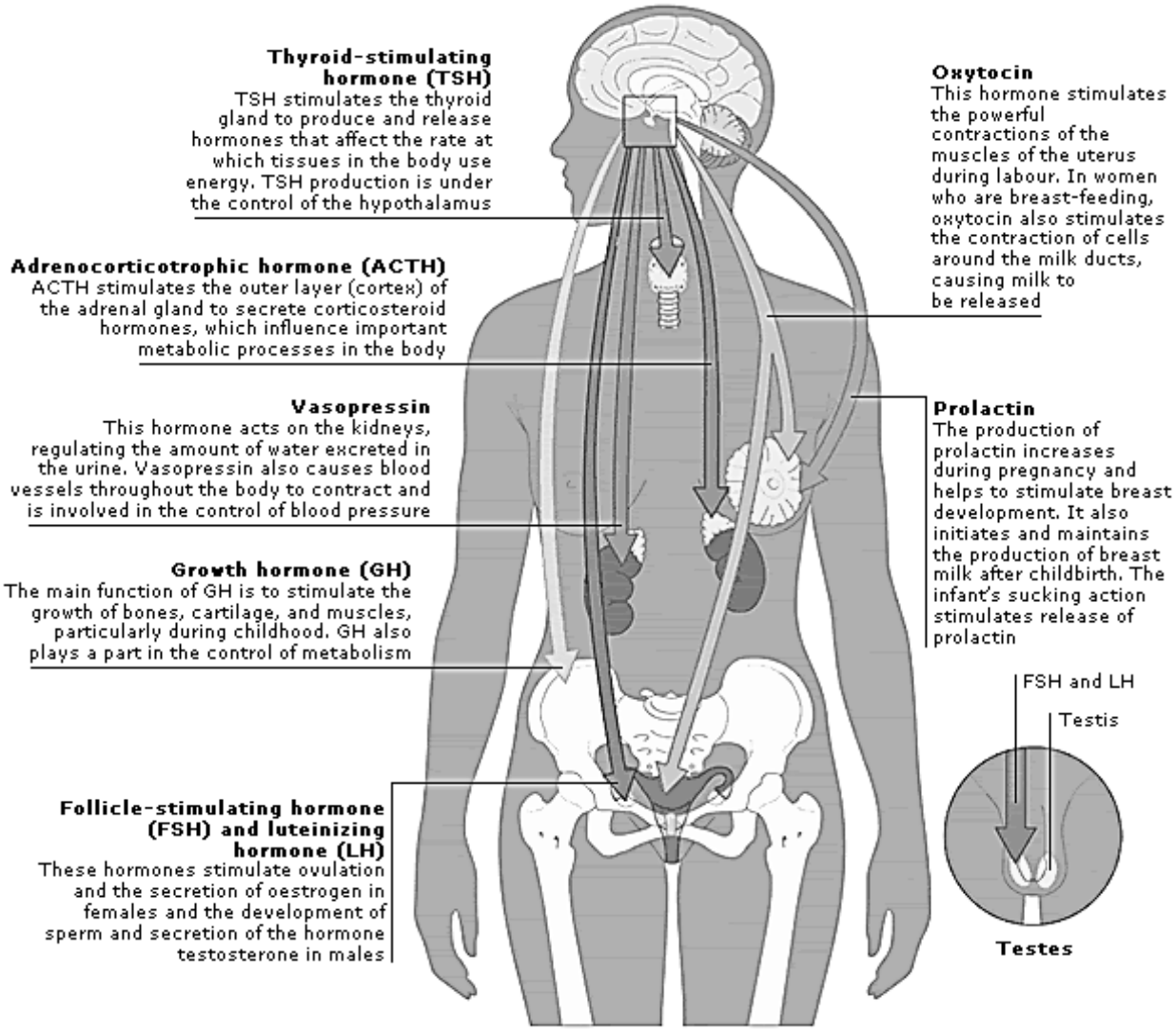
*Describe the endocrine functions of gonads. List the male and female sex hormones and their physiological role.*

Male sex hormones: _____

Female sex hormones: _____

Additional information



## WORK 10.3. HUMAN HEIGHT EVALUATION

The body growth is an irregular process. Maximum growth rate is noted in newborns and infants and then it considerably decreases. Some increase of growth rate is noted in girls from 9 to 14 years and in boys from 11 to 16 years, then it decreases again. By 16 years in girls and by 18 years in boys the body growth is practically completed and in norm it does not exceed 1 cm/year. Complete ossification occurs by 20–23 years in a female organism and by 21–25 years in a male. The height of an adult of 130–200 cm in males and 120–190 cm in females is considered normal. Men less than 130 cm in height and women <120 cm in height are dwarfs. People-giants are women higher than 190 cm and men higher than 200 cm.

Height is an integral factor of the effect of genetic, hormonal, tissue and external factors on the bony and other tissues of the organism. The height genetic program is realized through the endocrine system including all known hormones (thyroid, insulin, calcium-regulating, adrenal, sex), but the most important is hypothalamic-pituitary regulation of growth, the central link of which is somatotropin. Somatotropin (somatotropic hormone or growth hormone) is a basic hormone stimulating linear growth. Somatotropin stimulates growth of bones in length, growth and differentiation of internal organs, development of muscle tis-sue. A basic effect of somatotropin at a bony tissue level is its stimulation of cartilage growth, protein synthesis and cell mitosis induction. Somatotropin effects are mediated by insulin-like growth factors (IGF-I, IGF-II) or comatomedins that are synthesized under the action of this hormone mainly in the liver and kidneys. The linear human growth is completed, when growth zones have become closed under the effect of sex hormones.

The most simple and accessible method of studying the somatotropin function is antropometric, i. e. the human height is evaluated versus its predicted height calculated on the basis of an average height of his parents. To determine the final height range the following formula is used:

**Predicted final height of a male = (father's height + mother's height + 13 cm) : 2**

**Predicted final height of a female= (father's height + mother's height – 13 cm) : 2**

The measured height of an adult must coincide with a predicted height or deviate from a calculated value no more than 2 standard deviations (SD), i.e. ±10 cm from a calculated height value. Deviations of the measured height exceeding 2 SD from a calculated height value evidence a pathologically low or high hu-man height. In this case it is necessary to perform detailed studies of the pituitary somatotropic function to clear up the cause of growth impairment, as well as to study the state of other glands (first of all sex and thyroid glands).

**Materials and equipment**: a height meter.

***TO PERFORM THE WORK ONE SHOULD KNOW THE HEIGHTS OF THE PARENTS!***

**Accomplishment**. Height measurement is performed in standing position with the height meter. The examined should stand without shoes (in thin socks) in the right position: arms down; heels together; heels, buttocks and scapulae are pressed to the board of the height meter. The head is in position of "Frankfurt's plane", i.e. the lower edge of the eye and the external auditory canal should lie on one horizontal line. Measurements are performed on exhalation. The plank of the height meter is lowered to the level of the head of the examined. Measurements are performed with precision of 0.5 cm.

**PROTOCOL**

1. Height of the examined is _____ cm.
Sex of the examined _____.

2. Parents' height of the examined: father's ____ cm; mother's ___ cm.

3. Calculation of predicted height of the examined (PHE)
**PHE** = (father's height + mother's height ± 13 cm) : 2 = _____ + _____ + or – _____ = _____ **cm**.

4. **Conclusion**. Height of the examined is _____ (*in norm, pathologically high, pathologically low*).

5. **Excess** of growth hormone in childhood or adolescence or insufficiencies of sex hormones may result in pathologically _____ _____ height. **Insufficiency** of growth hormone in childhood and adolescence or excess of sex hormones may result in pathologically _____ height.

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Teacher's signature

## Lesson 11. HUMORAL REGULATION OF FUNCTIONS. PHYSIOLOGY OF THE ENDOCRINE SYSTEM. LESSON № 2

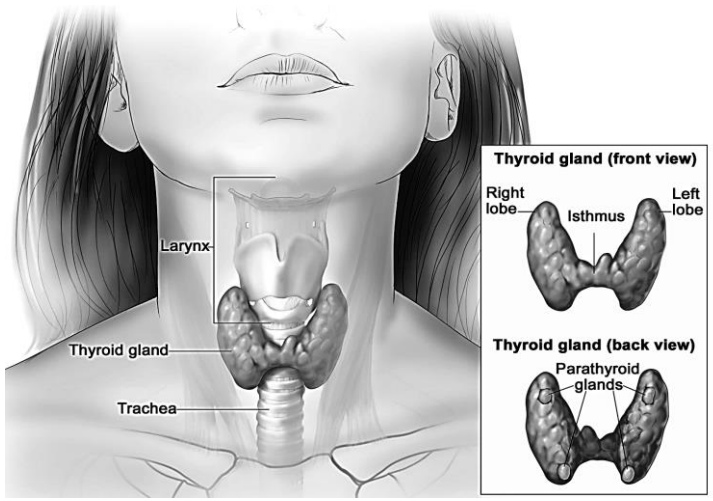
DATE OF CLASSES
«____» _____ 20____
day month year

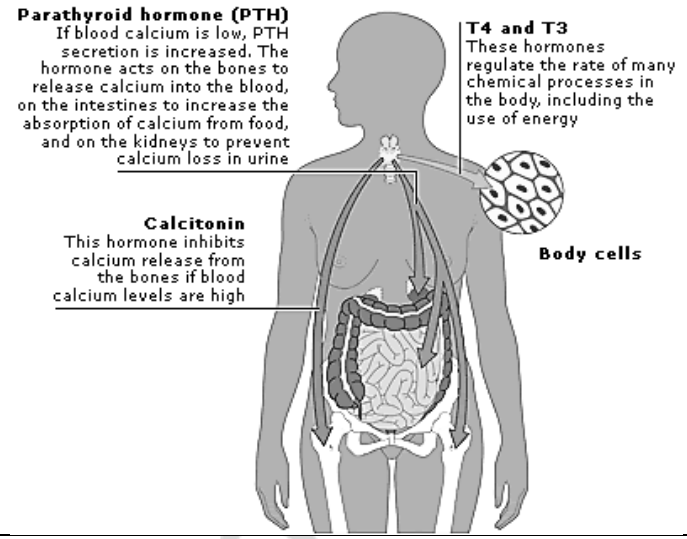
| **Basic questions**: | LITERATURE<br>Main |
|---|---|
| 1. Endocrine function of the thyroid and parathyroid glands.<br>2. Physiology of the adrenal glands. Hormones of the adrenal cortex and medulla in the regulation of body functions.<br>3. The concept of stress, its mechanisms and methods of prevention.<br>4. Endocrine function of the pancreas and its role in the regulation of carbohydrate, fat and protein metabolism.<br>5. The concept of the endocrine function of the heart (atrial natriuretic peptide), kidney (calcitriol, erythropoietin), fat tissue (leptin), salivary glands (parotin), liver (somatomedin C, thrombopoietin, 1 (OH)-VitD3).<br>6. Characteristic manifestations of excessive and insufficient secretion of hormones.<br>7. General concept of physiological approaches to the use of hormones for functional correction of the body functions. | 1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 155–181, 192–214.<br>Additional<br>3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 337–374, 429–449.<br>4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 951–1000. |

**BAZZWORD**

| | |
|---|---|
| Downregulation — | Atriopeptid — |
| Upregulation — | Erythropoietin — |
| Negative-feedback mechanism — | 1 (OH)-VitD3 — |
| Positive-feedback mechanism — | Vitamin D3 — |
| Sympathoadrenal system — | Calcitriol (cholecalciferol)— |
| IGF I — | Calcitonin — |
| Somatomedin C — | T3 — |
| Thrombopoietin — | T4 — |
| Parotin — | Stress — |

## WORK 11.1. THE ENDOCRINE FUNCTION OF THE THYROID AND PARATHYROID GLANDS

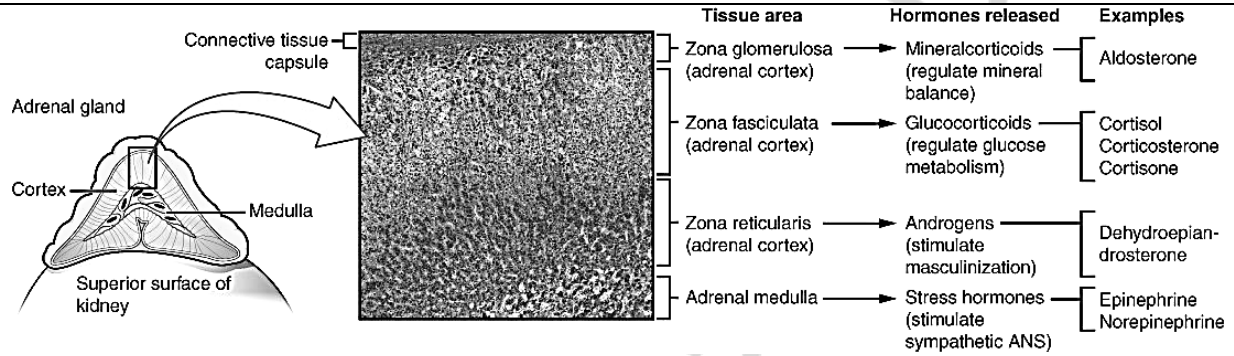


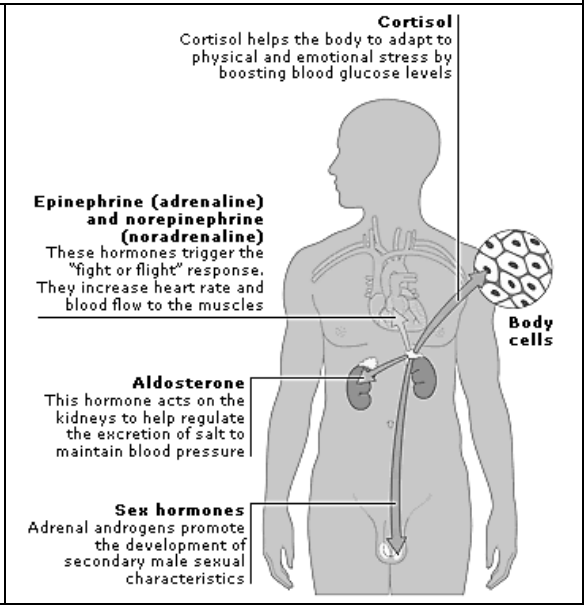




*Fill in the table*

| **In children** |
|---|
| Excess of $T_3$ and $T_4$ |
| |
| Insufficiency |
| |
| **In adults** |
| Excess of $T_3$ and $T_4$ |
| |
| Insufficiency |
| |

## WORK 11.2. PHYSIOLOGY OF THE ADRENAL GLANDS




*Fill in the table. Describe the main hormons of adrenal gland*

| Hormone | Functions | Excess | Insufficiency |
|---|---|---|---|
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**WORK 11.3. STUDYING EFFECTOR HORMONES OF ENDOCRINE SYSTEM AND THEIR ACTION ON TARGET CELLS (TISSUES, ORGANS)**

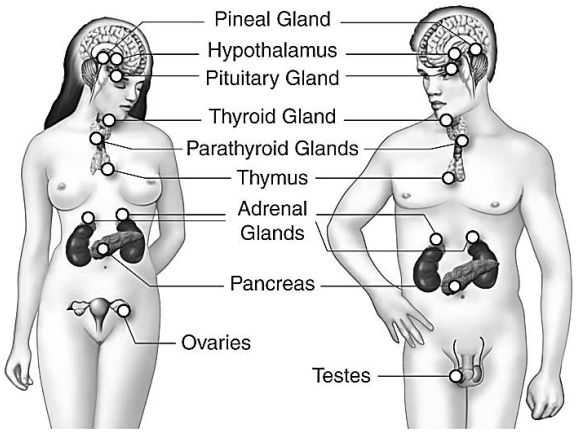


Fig. 11.1

Fill in the table. Describe the manifestation of excess and insufficiency of main *effector hormones*.

| Source (gland) | Hormone name | Chemical structure | Basic functions | Excess (hyper-function) | Insufficiency (hypo-function) |
|---|---|---|---|---|---|
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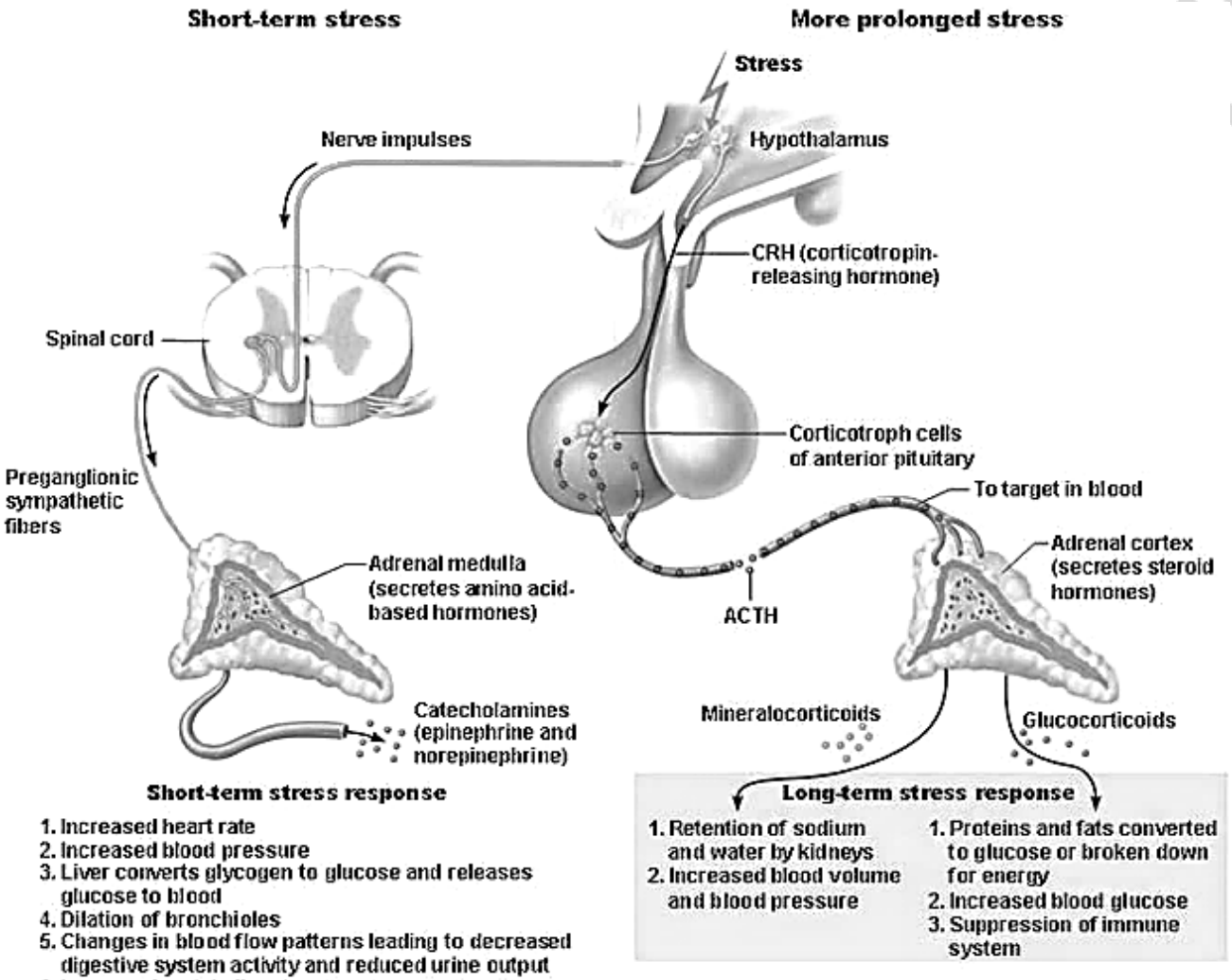
## WORK 11.3. GENERAL ADAPTATION SYNDROME (STRESS): NERVOUS AND HORMONAL MECHANISMS OF THEIR DEVELOPMENT




*Describe the mechanisms of stress development*

1. Short-term stress response: _____

2. Long-term stress response: _____

**WORK 11.4. ANALYSIS OF THE EFFECT OF CATECHOLAMINES AS HORMONES (OF ADRENAL MEDULLA) AND AS NEUROTRANSMITTERS (OF THE SYMPATHETIC DEPARTMENT OF ANS) ON CARDIOVASCULAR SYSTEM**
**(demonstrative computer work)**

**Accomplishment**. The work is performed as virtual experiment on rats in the program "Physiol 2".

**Directions for recording the Protocol**:

1. Fill in the table. Abbreviations: HR – Heart Rate, BPsyst – Systolic Blood Pressure, BPdiast – Diastolic Blood Pressure, BPmean – Mean Hemodynamic Blood Pressure.

2. Make a conclusion, what is the difference between the action of catecholamines as neuro-transmitters of sympathetic nerves and as hormones of the adrenal medulla. Indicate, by what types of adrenoreceptors the effect of noradrenalin and adrenalin on the cardio-vascular system is predominantly realized.

**PROTOCOL**

| Effect on the heart | HR | $BP_{syst}$ | $BP_{diast}$ | $BP_{mean}$ |
|---|---|---|---|---|
| **Initial values** | | | | |
| **Stimulation** Symp. Nerves to heart $T_1$ | | | | |
| **Stimulation** Symp. Nerves to adrenals $T_{6–8}$ | | | | |
| Phentolamine (α-adrenoblocker), 100 mg/kg + **stimulation** Symp. Nerves to heart $T_1$ | | | | |
| Propranolol (β-adrenoblocker), 100 mg/kg + **stimulation** Symp. Nerves to heart $T_1$ | | | | |
| Propranolol (β-adrenoblocker), 100 mg/kg + **stimulation** Symp. Nerves to adrenals $T_{6–8}$ | | | | |
| **Injection** noradrenaline, 5μg/kg | | | | |
| **Injection** adrenaline, 5μg/kg | | | | |

**Conclusions**: as the blockage of α adrenergic receptors by phentolamine _____ (does or does not) prevent the sympathetic nerves effect on the heart, and the blockage of β adrenergic receptors _____ (does or does not) prevent this effect, the sympathetic nerves effect on the heart is achieved through ____ adrenergic receptors.

Effect of *noradrenaline* is achieved mainly through _____adrenergic receptors, while effect of *adrenaline* is achieved through both _____- and _____adrenergic receptors.

THE LABORATORY WORKS ARE PASSED WITH MARK

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Teacher's signature

## Lesson 12. REGULATION OF CALCIUM AND PHOSPHORUS IN THE BODY, IN THE BONE TISSUE AND IN THE TEETH

DATE OF CLASSES
«____» _____ 20____
day month year

| **Basic questions**: | LITERATURE |
|---|---|
| 1. The role of calcium and phosphate in the body, their compounds and content in bone tissue and teeth.<br>2. Bone tissue: functions, features of the structure and composition, age-related changes. The concept of remodeling of bone tissue.<br>3. Hard tissues of teeth: types, functions. Enamel: structure, properties, functions, nutritional features.<br>4. Dental formula for milk and permanent teeth.<br>5. The balance of calcium and phosphate in the body and in bone tissue: age-specific features, mechanisms of regulation. The daily requirement in calcium, phosphate and fluoride.<br>6. The concept of homeostasis. Mechanisms to maintain a constancy of internal environment and functions of the organism as well as the mechanisms regulating them (for example, the regulation of calcium levels in the blood: calcitonin, calcitriol and PTH).<br>7. Factors of preserving the health of bone tissue and teeth. | Main<br>1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 181–192.<br>Additional<br>3. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 62, 375–388.<br>4. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 4. 1001–1020. |

**BAZZWORD**

| | |
|---|---|
| Bone composition — | Bite — |
| Osteoclasts — | Daily requirements in calcium for children _____, for adult _____, for pregnant _____ |
| Osteoblasts — | Daily requirements in phosphate — |
| Enamel — | Daily requirements in fluoride — |
| Dentine — | Homeostasis — |
| Pulp — | PTH — |
| Hydroxyapatite — | Absorption — |
| Dental formula — | Reabsorption — |

## WORK 12.1. EVALUATION OF A DENTAL FORMULA. BITE ANALYSIS

The teeth are arranged so that their crowns form an arc or a row on the upper and lower jaws. The dentition consists of 10 deciduous (milk) teeth (4 incisors, 2 canines, and 4 molars) in children and from 16 permanent teeth (4 incisors, 2 canines, 4 premolars, and 6 molars) in adults. In total, a person has a total of 20 temporary teeth and 32 permanent teeth. The eruption of milk teeth begins at 6–8 months and ends at 2.5–3 years, and its loss begins at 6–7 years and ends at 11–13 years. The eruption of permanent teeth begins at 6–7 years and is completed by the age of 17–22.

The eruption of milk and permanent teeth is an important indicator of physical development ("dental" age) and reflects the interaction of local (humoral) and endocrine (thyroid hormones, growth hormone, etc.) factors in the regulation of this process.

For example, the development of thyroid function in humans coincides with the period of differentiation of the rudiments of milk teeth. Therefore, both the premature onset of the functioning of the thyroid gland and congenital hypothyroidism cause hypoplasia (insufficient formation of tissue elements) of the teeth and violation of the terms of their eruption.

The dentition of the upper and lower jaws is closed in a certain position. The ratio of the dentition of the upper and lower jaws with the most complete closure of antagonist teeth is called bite. There is another definition of bite. Bite – the nature of tooth closure in the position of the central occlusion.

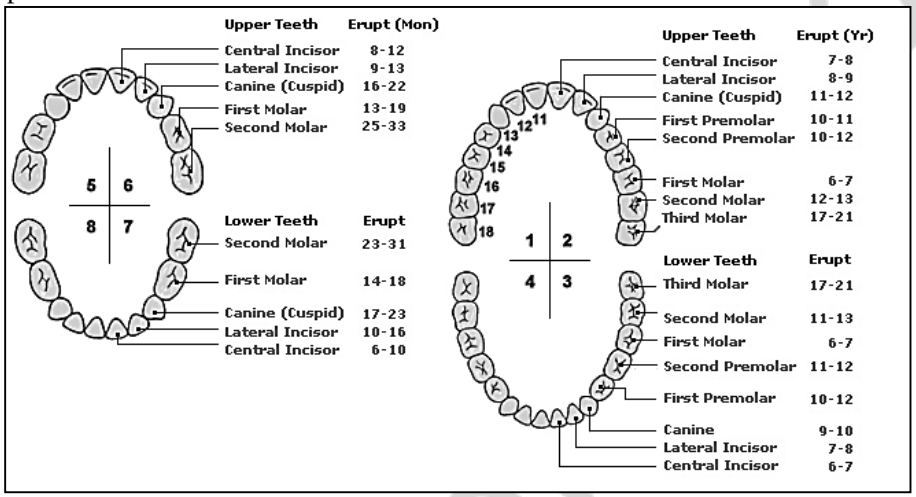



Fig. 12.1. Dental formula for milk (left) and permanent (right) teeth

**Materials and equipment**: dental mirror (preferably an individual (personal) for each student), a glass with disinfecting solution (chloramine, septocide etc.).

**Accomplishment**. Ask the tested person to open his mouth as much as possible and inspect the presence and location of the teeth with the help (or without help) of a dental mirror. Then ask the tested to close his jaws and grin his teeth. Consider the nature of the ratio of the teeth in the position of the central occlusion (overlap of the incisors, as well as the ratio of the first antagonistically located premolars) and evaluate the bite variant of the subject.

**Directions for recording the Protocol**:

1. Write the normal clinical dental formula proposed by World Health Organization for milk and permanent teeth of a healthy child and adult.
2. Write the dental formula of the examined and the permanent bite in the tested person. Indicate the groups of teeth.
3. Evaluate the "dental" age (passport compliance) of tested person.

**PROTOCOL**

1. Dental formula for milk teeth:

*teeth groups*

*r.* *l.*

2. Dental formula for permanent teeth:

3. Age _____ years. Dental formula for permanent teeth *of examined*:

*Only existing teeth indicate. Pay attention to the presence of third molars!*

4. **Conclusion**: "Dental" age corresponds _____ (yes or no) to passport age.

**WORK 12.2. STUDYING THE MECHANISMS OF REGULATION OF HOMEOSTASIS OF CALCIUM AND PHOSPHORUS IN THE ORGANISM**

*Fill in the schemes (empty spaces) of calcium homeostasis maintenance*

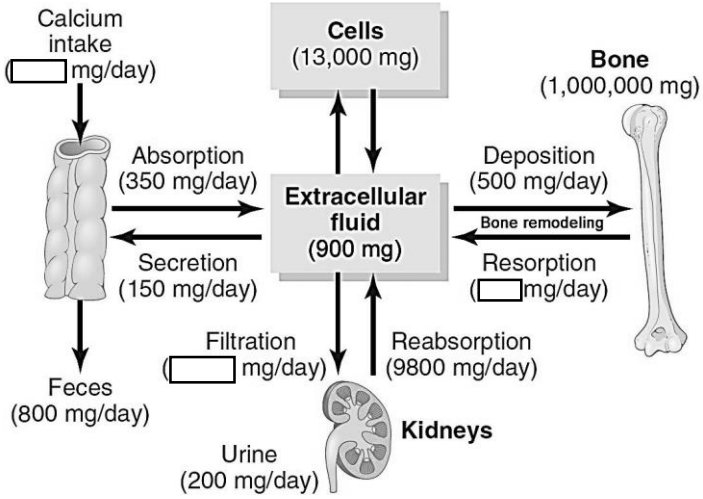



Fig. 12.2. $Ca^{2+}$ homeostasis in an adult eating 1000 mg/day of elemental $Ca^{2+}$

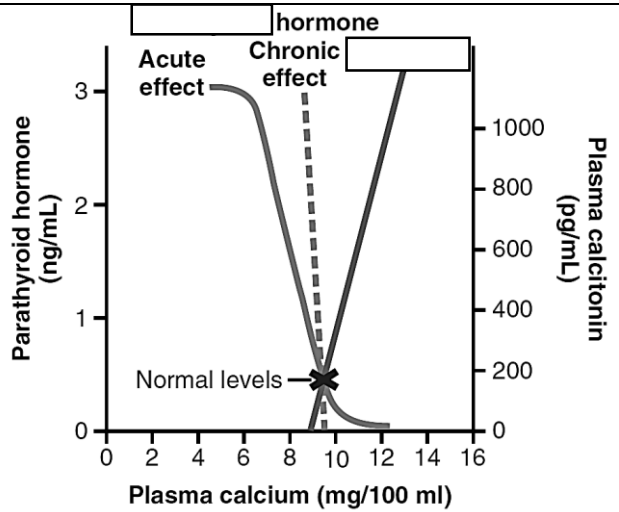



Fig. 12.3. The approximate effect of plasma calcium concentration on the plasma concentrations of parathyroid hormone and calcitonin

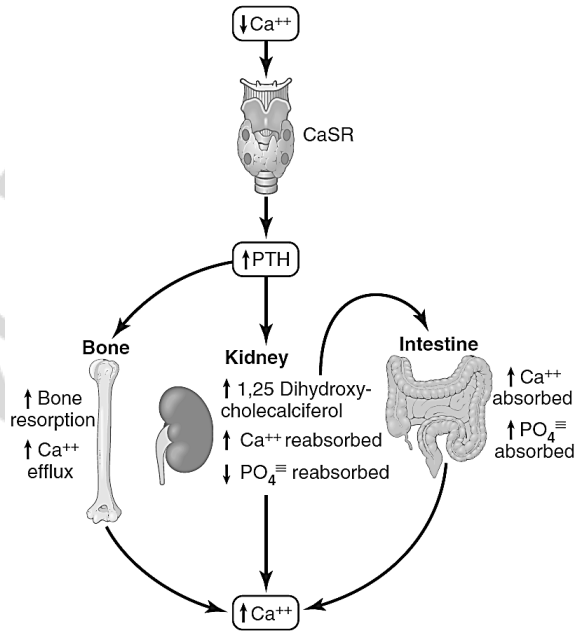



Fig. 12.4. Summary of effects of PTH in response to decreased $Ca^{2+}$ concentration. CaSR – calcium-sensing receptor

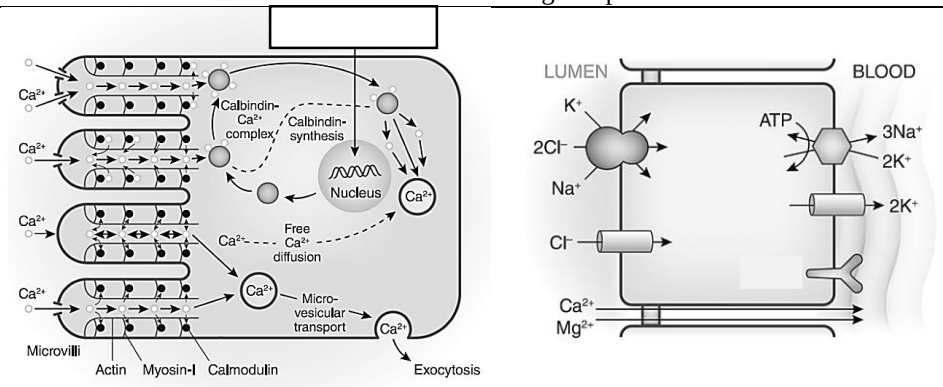



Fig. 12.5. Intestinal (left) and ascending limb of Henle (right) pathways for calcium absorption

*Fill in the schemes (empty spaces) of phosphate & $Ca^{2+}$ homeostasis maintenance*

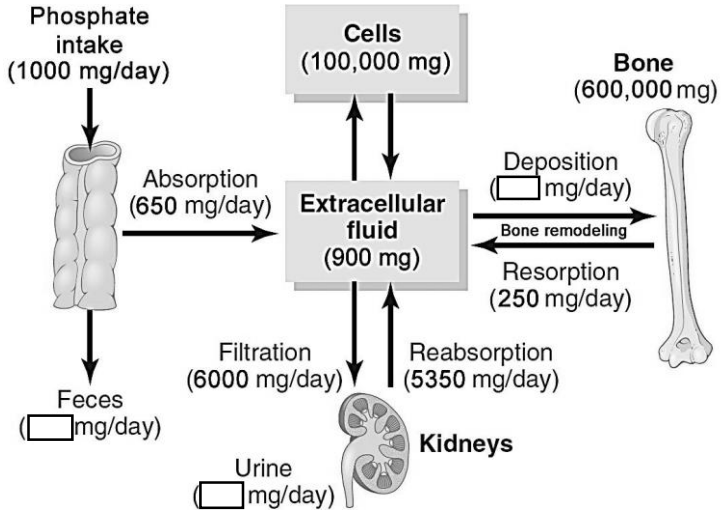
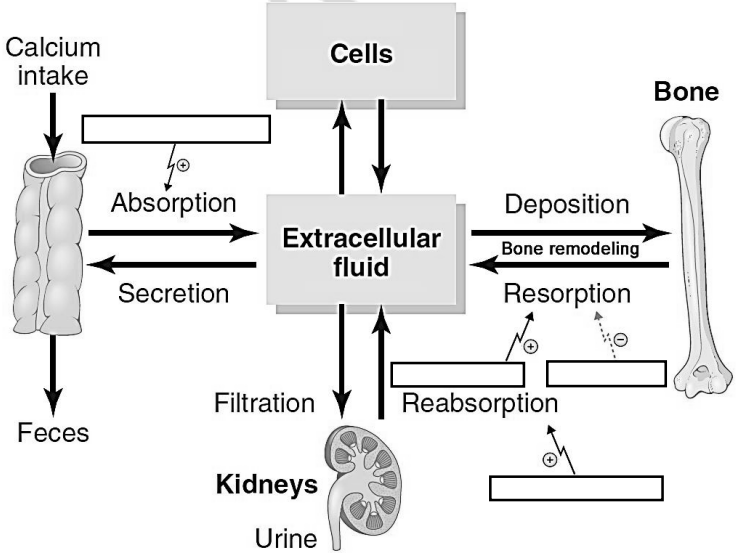



Fig. 12.6. $P_i$ homeostasis in an adult eating 1000 mg/day of elemental $P_i$




*Describe the main effects of Ca2+ and $P_i$-regulating hormones:*

| Effects | Regulation of secretion | Plasma $Ca^{2+}$ concentration | Plasma $P_i$ concentration | Intestinal $Ca^{2+}$ & $P_i$ reabsorption | Bone $Ca^{2+}$ & $P_i$ reabsorption | Kidney reabsorption of $Ca^{2+}$ | Kidney reabsorption of $P_i$ |
|---|---|---|---|---|---|---|---|
| PTH | | | | | | | |
| $VitD_3$ | | | | | | | |
| Calcitonin | | | | | | | |

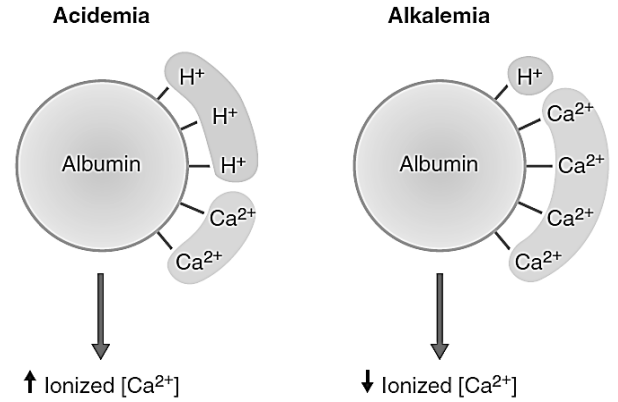



Fig. 12.7. Effects of acid-base disturbances on plasma protein-binding of $Ca^{2+}$ and the ionized $Ca^{2+}$ concentration in blood

THE LABORATORY WORKS ARE PASSED WITH MARK

Teacher's signature

## Lesson 13. COLLOQUIUM "MECHANISMS OF REGULATION OF FUNCTIONS"

DATE OF CLASSES
«____» _____ 20____
day month year

**Main questions:**

1. The concept of physiological function and it regulation. Levels of regulation. Types of regulation.
2. Nervous and humoral mechanisms of functions regulation, their comparative characteristics.
3. The structure and functions of the spinal cord. Spinal reflexes.
4. The structure and functions of the brain. Multilevel system of muscle tone regulation
5. Modern concept of localization of function in the cerebral cortex. Functional asymmetry of the cortex. Forebrain structures and its functions.
6. Comparative characteristics of somatic and autonomic nervous system (sensory receptors, afferent, association and efferent divisions, effector organs).
7. Differences of neuroeffector connections of smooth muscle and neuromuscular synapses of skeletal muscle.
8. Comparative characteristics of the structure and neurochemical mechanisms of the sympathetic and parasympathetic parts of ANS, as well as their influence on the effector organs.
9. The concept of the principles of the autonomic functions correction (for example, salivation) by affecting the neurotransmitter-receptor mechanisms in ANS ganglia and of the effector cells.
10. Endocrine system. The pituitary gland, its connections with the hypothalamus.
11. Hormones of the pituitary gland (hypophysis) and hypothalamus, their role in the regulation of endocrine and not endocrine organs.
12. Endocrine function of the thyroid and parathyroid glands.
13. Physiology of the adrenal glands. Hormones of the adrenal medulla, its role in the regulation of body functions.
14. Hormones of the adrenal cortex, its role in the regulation of body functions.
15. Endocrine function of the pancreas and its role in the regulation of carbohydrate, fat and protein metabolism.
16. Gonads. Male and female sex hormones and their physiological role.
17. The concept of the endocrine function of pineal gland (melatonin), heart (atrial natriuretic peptide), kidney (calcitriol, erythropoietin, and other), fat tissue (leptin), salivary glands (parotid and others), liver (somatomedin, thrombopoetin, cholecalciferol).
18. Physiological properties of skeletal muscles and their functions.

LITERATURE

Main


1. Lecture & E-learning system.
2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 80–249.
3. *Severina, T. G.* Physiology of blood. Lecture notes / T. G. Severina. 2nd ed. Minsk : BSMU, 2017. P. 13–21.


Additional


4. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016.
5. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016.


**Form of colloquium:**
***computer control test "Lesson 13" at E-learning system with oral or writing part.***

| | |
|---|---|
| 19. Force and work of muscle contraction (masticatory muscles). Gnatodinamometry. Dynamometry of hands and back muscles.<br>20. Motor units and their characteristics in different muscles. Types of muscle fibers. Muscle fatigue.<br>21. Contraction of whole muscle. Types and regimes of skeletal muscle contraction. Summation. Tetanic muscle contraction and its types.<br>22. Mechanisms of contraction and relaxation of a single muscle fiber (sliding filaments theory).<br>23. Functional purpose of individual muscles of mastication. Movement of the lower jaw (mandible). Physiological occlusion (bite, central occlusion, "gothic" arch).<br>24. Physiological properties and characteristics of smooth muscle compared to skeletal. Smooth muscle tone. The concept of myoepithelial cells.<br>25. Bone tissue: functions, features of the structure and composition, age-related changes. The concept of remodeling of bone tissue.<br>26. Hard tissues of teeth: types, functions. Enamel: structure, properties, functions, nutritional features.<br>27. Dental formula for milk and permanent teeth.<br>28. The balance of calcium and phosphate in the body and in bone tissue: age-specific features, mechanisms of regulation. The daily requirement in calcium, phosphate and fluoride.<br>29. The concept of homeostasis. Mechanisms to maintain the constancy of the internal environment of the body (for example, regulation of calcium levels in the blood: calcitonin, calcitriol and PTH). | |

THE COLLOQUIUM IS PASSED WITH MARK

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## SECTION “BODY FLUIDS”

## Lesson 14. BODY FLUIDS (BLOOD, LYMPH, CEREBROSPINAL FLUID, SALIVA, ETC.)

DATE OF CLASSES
«____» _____ 20____
day month year

| **Basic questions**: | LITERATURE<br>Main |
|---|---|
| 1. The role of water for vital functions The content and distribution of water in the organism. The main fluid compartments of the body. Water balance.<br>2. Blood. The concept of the blood system. The composition of the blood, its physiological properties and functions. Basic physiological constants of blood.<br>3. Acid-base balance and the mechanisms of its maintaining. Buffer systems of blood.<br>4. Blood plasma, its quantity, composition, and properties. Hemolysis and its types.<br>5. The electrolyte composition of blood plasma. The osmotic pressure of the blood and its regulation (ADH, RAAS, natriuretic peptides (ANP & BNP)).<br>6. Blood plasma proteins, their characteristics. Colloid osmotic (oncotic) blood pressure.<br>7. Lymph, its composition, physicochemical properties and functions. Lymph formation.<br>9. Cerebrospinal fluid (CSF), its quantity, composition and function.<br>10. Fluids of the oral cavity: oral fluid (“mixed saliva”), gingival fluid, and saliva of salivary glands. Acid-base condition of the oral cavity. | 1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 250–254, 605–607, 613–630.<br>3. *Severina, T. G.* Physiology of blood. Lecture notes / T. G. Severina. 2nd ed. Minsk : BSMU, 2017. P. 3–12, 22-25.<br>Additional<br>4. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 553–554, 562–564, 567, 582, 695–706, 603–604.<br>5. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 4–6, 305–320, 381–387, 389–390, 396, 397–398, 401–416, 790–793. |

| **BUZZWORD** | |
|---|---|
| Compartment — | Isotonic solution — |
| Blood system — | Saline — |
| Blood plasma — | Oral fluid (“mixed saliva”) — |
| Hemolysis — | Gingival fluid — |
| Strict physicochemical constants of blood — | pH of oral fluid — |
| Blood pH — | RAAS — |
| Osmotic pressure — | ANP — |
| Oncotic pressure — | BNP — |

## WORK 14.1. METHODS OF TAKING CAPILLARY BLOOD (DEMONSTRATION). SAFETY RULES FOR INFECTION PREVENTION

Common clinical blood analysis is one of the most widespread laboratory examinations. Capillary blood is often used for this purpose.

Working with blood one should remember that blood can be virulent (HIV, hepatitis, etc.) and physitians and laboratory assistants performing serological and clinical tests are at risk of getting infected. That is why while making blood tests one should follow regulations of the Ministry of Health of the Republic of Belarus on prophylaxis of viral hepatitis and HIV in medical workers engaged in taking and analyzing blood.

While performing laboratory tests of the blood and other biological fluids one should use individual protective means: a medical gown and rubber gloves, spectacles, a mask (or a shield).

Any injury of the skin, mucous membranes, getting blood or other biological fluid of the patient there should be qualified as a possible contact with the material containing HIV or other infected agent.

*1. In case of integument lesions (puncture, cut) when working with biological material the victim should*:

– quickly take off the gloves with the working surface inside and immerse them in a container with a disinfectant solution or place them in a waterproof bag for subsequent disinfection;

– wash the hands with soap under running water and wash the wound abundantly with water or isotonic NaCl solution (saline);

– rinse the injured site with 3 % peroxide solution.

*2. In case of contamination of the skin with biological material without cutaneous lesions*:

– wash the contaminated skin area thoroughly with soap and water and treat with an antiseptic.

*3. When biological material has got on mucous membranes:*

– immediatly take off the gloves with the working surface inside and immerse them in a container with a disinfectant solution or place them in a waterproof bag for subsequent disinfection;

– wash the contaminated site with running water and soap and wash the mucosa abundantly (do not rub) with water or saline.

*When biomaterial gets on the sanitary and hygienic clothing (hereinafter referred to as SHC) – gown (lab coat), or personal clothes, footwear*:

– wash the glove surface, without removing it from your hands, under running water with soap or an antiseptic solution, disinfectant;

– remove contaminated CHS, personal clothes, shoes;

– SHS, personal clothing and shoes should be folded into waterproof bags for subsequent disinfection;

– take off the gloves with the working surface inside and immerse them in a container with a disinfectant solution or place them in a waterproof bag for subsequent disinfection;

– wash your hands with soap and running water and treat the skin in the area of the projection of the pollution of CHS, personal clothes, shoes in accordance with paragraph 2 of this order.

*5. In case of contamination with biomaterial of environmental objects*, it is should be disinfected with a disinfectant solution and removed from the surface, followed by wet cleaning.

**Materials and equipment**: disposable scarifiers, sterile cotton wool, alcohol, iodine, rubber gloves, masks, disinfectant solution.

**Accomplishment**. Demonstration is shown as teaching video (in the computer room).

Taking capillary blood probe from the patient should be done as follows:

1. The patient should sit opposite the doctor, the patient's hand (better non-working) should be on the table.
2. Taking blood is done from the 4$^{th}$ (ring) finger, as its synovial sheath is isolated preventing the spread of an inflammatory process to the wrist in case of infecting the site of puncture.
3. The finger skin is disinfected with disinfectant solution.
4. The scarifier is taken by the middle with the hand by the end opposite to a puncturing one.
5. A skin puncture is done in the side of the ball of the finger across the lines of the fingerprint, not parallel to them, the scarifier being thrust to a full depth of a cutting surface.
6. The first blood drop is wiped away with dry sterile cotton wool or gauze (to avoid specimen dilution with interstitial fluid), the finger is carefully wiped out (the skin should be dry).
7. The next blood drop should have a convex meniscus and not spread about the finger, this drop and the next ones are taken for analysis.
8. Having taken the blood the puncture site is treated with an antiseptic.

**Answer to the questions**:

Why isn't the first blood drop recommended to be used for analysis:

_____

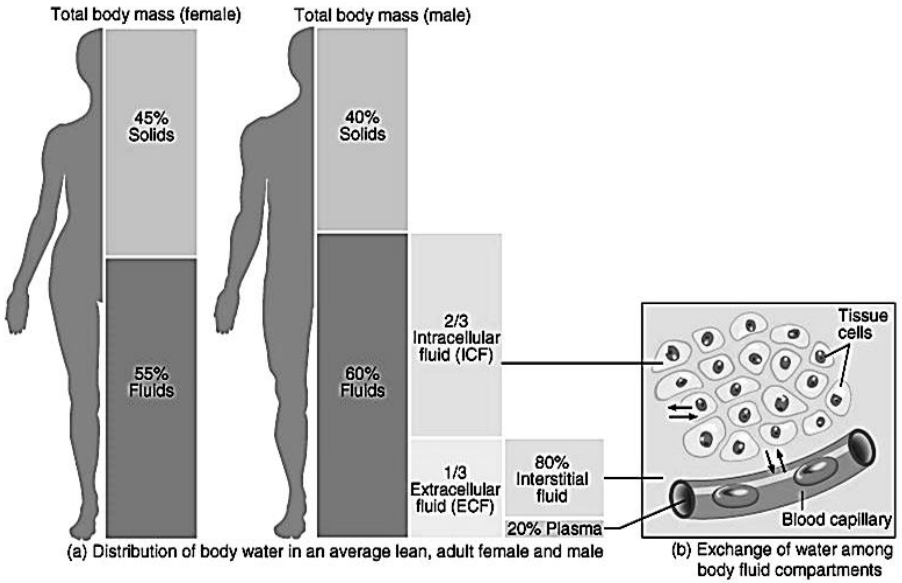
Why is the blood usually taken from the 4$^{th}$ finger of a non-working hand?

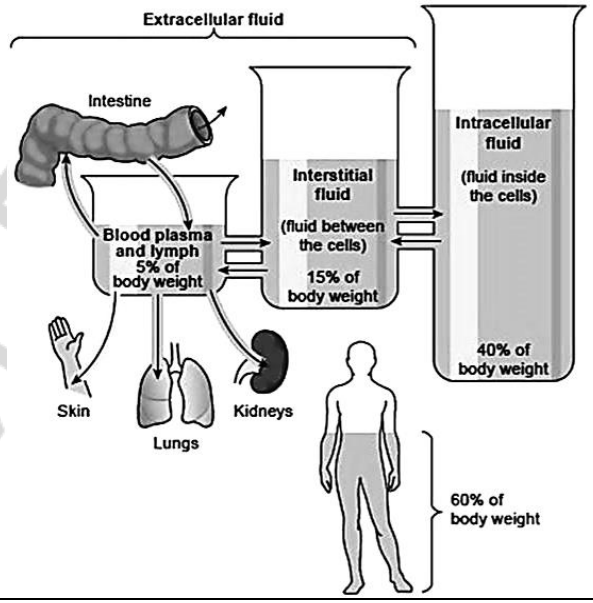
_____

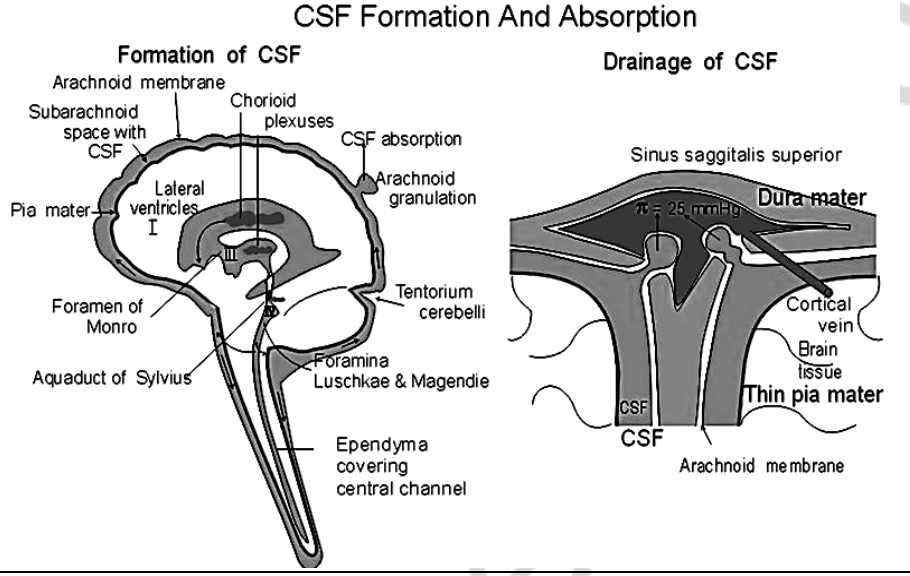
_____

With safety provisions while performing practical works with blood and other biological fluids as well as with tissues has been acquainted and instructed _____ (**student's signature**)

**WORK 14.2. STUDYING THE BODY FLUIDS DISTRIBUTION AND MAINTENANCE MECHANISMS**

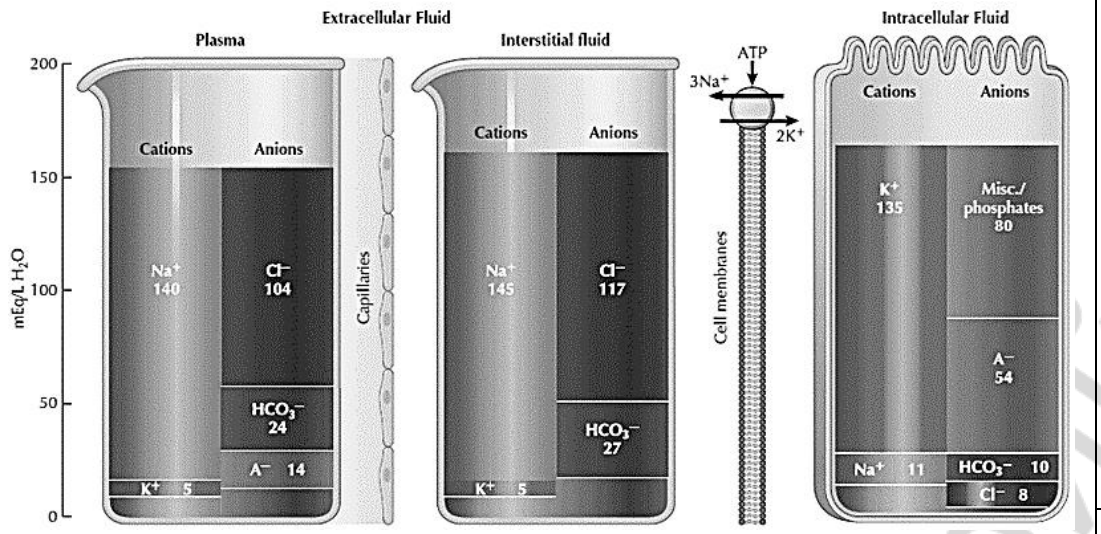


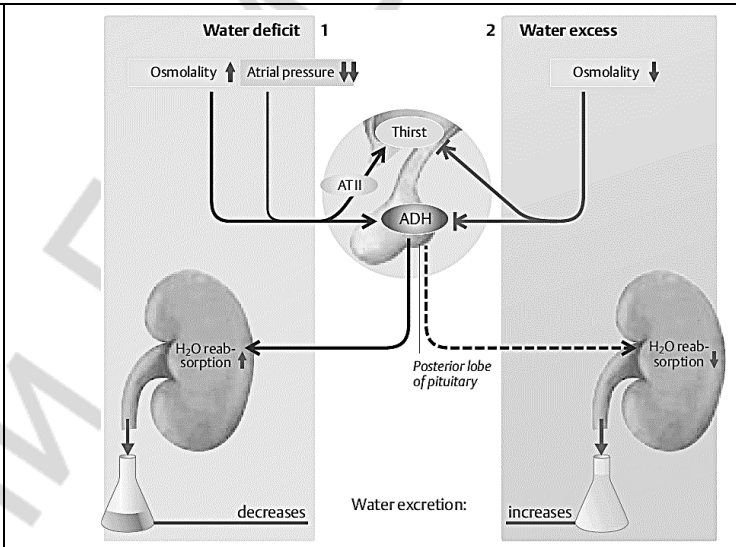



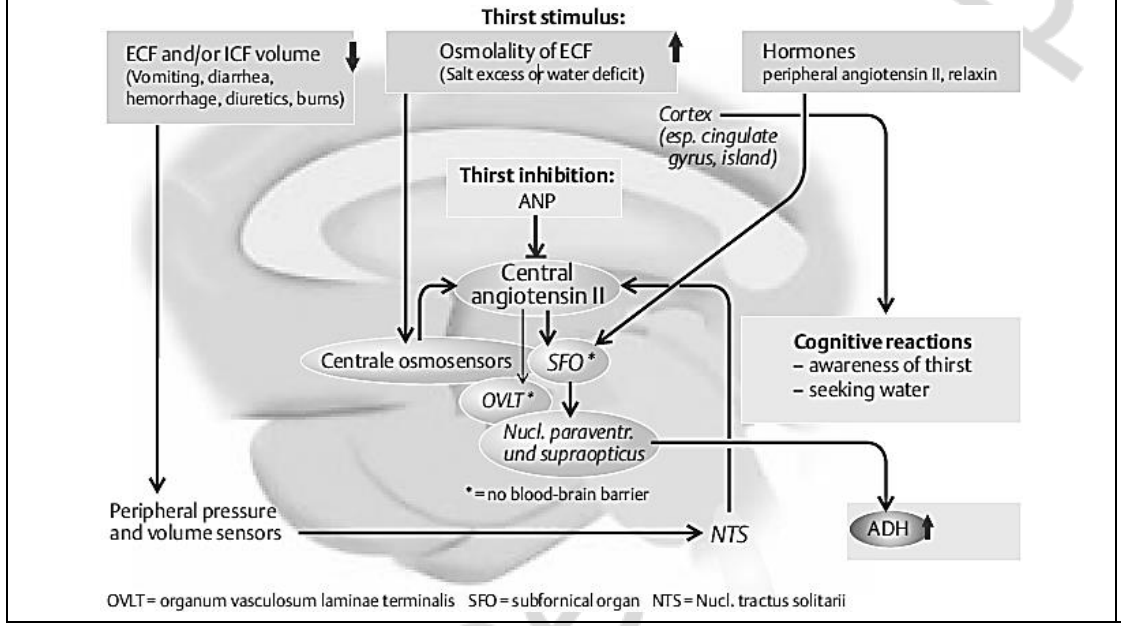



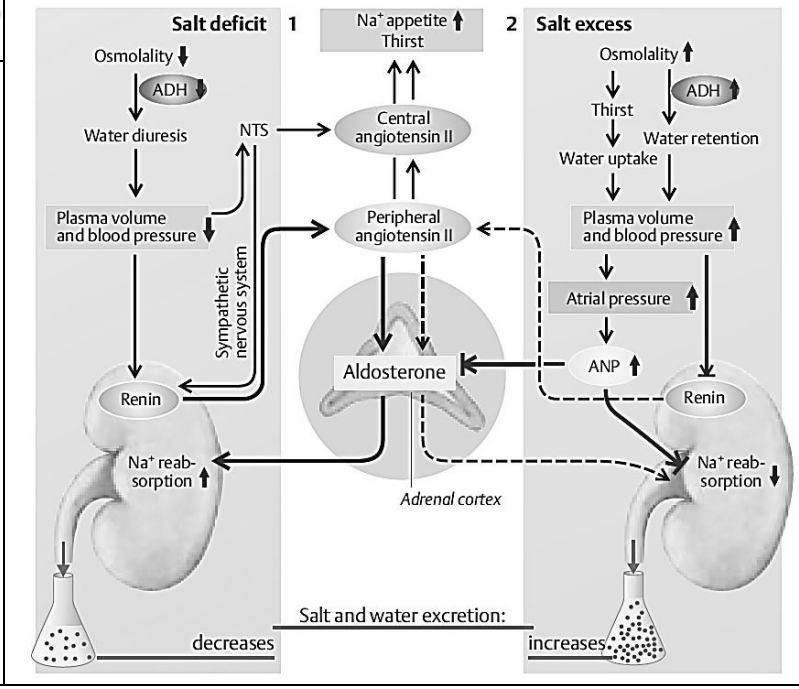

*Complete daily water intake and loss (L/day)*:

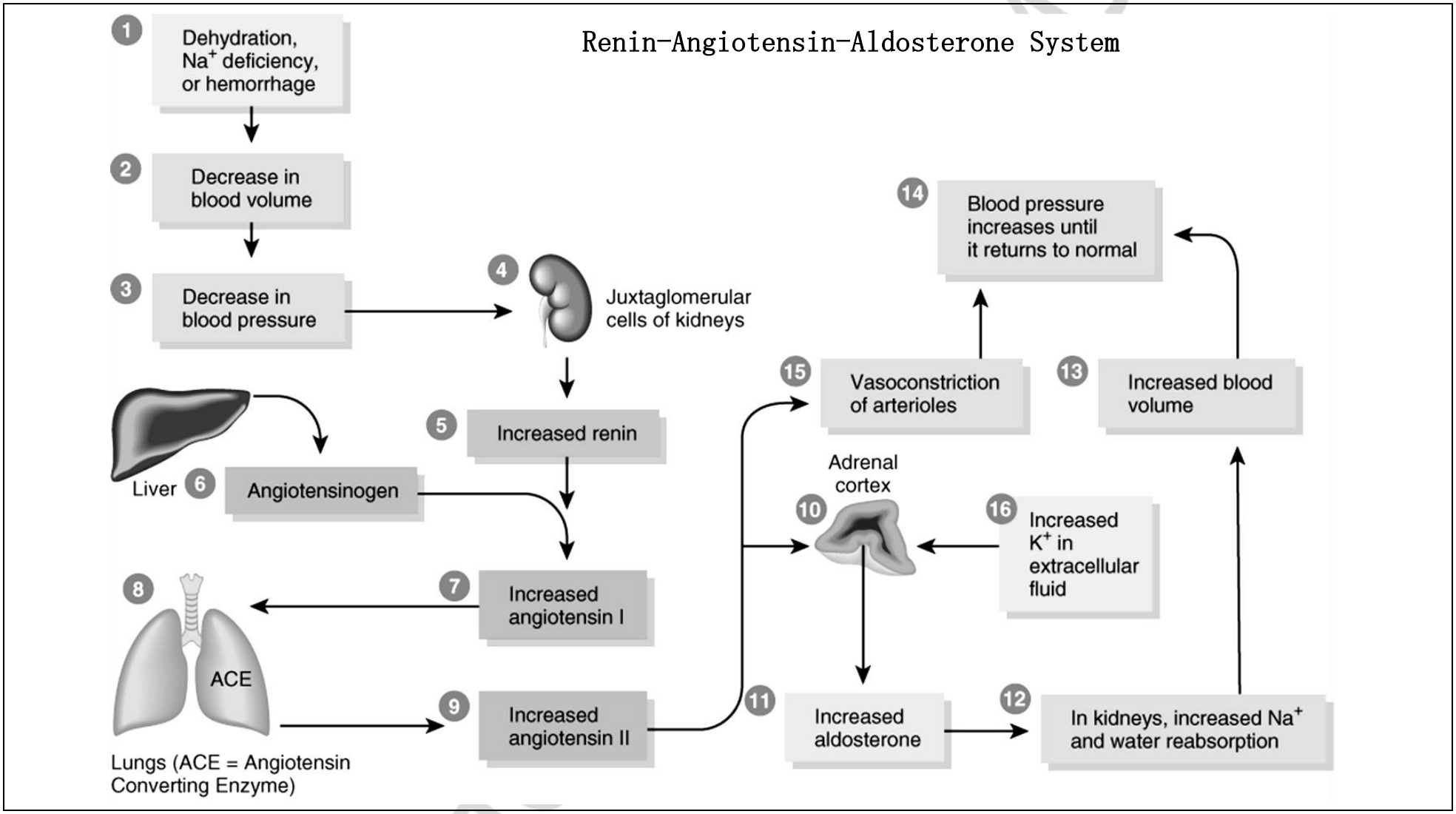
| Way | Intake | Output |
|---|---|---|
| | | |
| | | |
| | | |
| **Total** | | |


Extracellular Fluid
Plasma
Interstitial fluid
Intracellular Fluid
mEq/L $H_2O$
200
150
100
50
0
Cations
Anions
$Na^+$ 140
$Cl^-$ 104
$HCO_3^-$ 24
$A^-$ 14
$K^+$ 5
Capillaries
$Na^+$ 145
$Cl^-$ 117
$HCO_3^-$ 27
$K^+$ 5
ATP
$3Na^+$
$2K^+$
Cell membranes
$K^+$ 135
Misc./ phosphates 80
$A^-$ 54
$Na^+$ 11
$HCO_3^-$ 10
$Cl^-$ 8


Water deficit
1
2
Water excess
Osmolality
Atrial pressure
Osmolality
Thirst
ATII
ADH
$H_2O$ reab-sorption
Posterior lobe of pituitary
$H_2O$ reab-sorption
decreases
Water excretion:
increases


Thirst stimulus:
ECF and/or ICF volume
(Vomiting, diarrhea, hemorrhage, diuretics, burns)
Osmolality of ECF
(Salt excess or water deficit)
Hormones
peripheral angiotensin II, relaxin
Cortex
(esp. cingulate gyrus, island)
Thirst inhibition:
ANP
Central angiotensin II
Centrale osmosensors
SFO*
OVLT*
Nucl. paraventr. und supraopticus
*= no blood-brain barrier
Cognitive reactions
– awareness of thirst
– seeking water
Peripheral pressure and volume sensors
NTS
ADH
OVLT = organum vasculosum laminae terminalis SFO = subfornical organ NTS = Nucl. tractus solitarii


Salt deficit
1
$Na^+$ appetite
Thirst
2
Salt excess
Osmolality
ADH
Water diuresis
NTS
Central angiotensin II
Osmolality
ADH
Thirst
Water retention
Water uptake
Plasma volume and blood pressure
Peripheral angiotensin II
Plasma volume and blood pressure
Sympathetic nervous system
Atrial pressure
ANP
Aldosterone
Renin
Renin
$Na^+$ reab-sorption
Adrenal cortex
$Na^+$ reab-sorption
Salt and water excretion:
decreases
increases




THE LABORATORY WORKS ARE PASSED WITH MARK

Teacher's signature

## Lesson 15. BLOOD CELLS. ERYTHROCYTE SEDIMENTATION RATE. THE COMMON CLINICAL BLOOD TEST. HAEMATOPOIESIS

DATE OF CLASSES
«____» _____ 20____
day month year

**Basic questions**:

1. Erythrocytes (RBC): peculiarities of the structure and properties, their functions. RBS count. RBC evaluation methods. Hematocrit (HTC), its level and evaluation.
2. Hemoglobin (Hb): its concentration, structure, main types and compounds, their physiological significance. Hemoglobin evaluation methods. Color Index (CI).
3. Erythrocyte sedimentation rate (ESR): definition, factors affecting it. Diagnostic significance of ESR. Methods of its evaluation.
4. Leukocytes (WBC), their types, quantity, properties and functions. Leukocyte formula, its peculiarities with age, "left" and "right" shifts. Leukocytosis and leukopenia.
5. The concept of levels and mechanisms of nonspecific and specific protection (resistance) of the organism. The concept of innate and adaptive immunity.
6. Platelets (PLT), their count, structure, properties and functions.
7. Common clinical blood test (hematology tests) and physiological evaluation of its results.
8. Hematopoiesis. Stem cell, its microenvironment. Nervous and humoral mechanisms of regulation. The role of vitamins ($B_{12}$, $B_9$ and others) and trace elements ($Fe^{2+}$ and others).

LITERATURE

Main


1. Lecture & E-learning system.
2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 254–264, 283.
3. *Severina, T. G.* Physiology of blood. Lecture notes / T. G. Severina. 2nd ed. Minsk : BSMU, 2017. P. 23–41.


Additional


4. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 554–558.
5. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 445–476.


**BUZZWORD**

Blood cells —

Hematopoiesis —

Stem cell —

Vitamin $B_9$ — ; $B_{12}$ —

Hemoglobin A —

ESR —

Hematocrit —

Leukocyte formula —

"Left" shift —

Leukocytosis —

Draw a **scheme of hematopoiesis**.
*Use the materials of lectures, E-learning system, and textbook.*

## WORK 15.1. EVALUATION OF A COLOR INDEX AND MCH

To evaluate an *absolute content* of hemoglobin in every erythrocyte the **MCH** (Mean Corpuscular Hemoglobin) index is used. It's approximately equal to 30 pg (reference range 25.4–34.6 pg). Its value is obtained by division of the hemoglobin (HGB) content in 1 liter by red blood cells count in 1 liter: **MCH = HGB / RBC**.


The **Color Index** (CI) is a relative value of hemoglobin content in red blood cells. CI is calculated by division of the hemoglobin content in g/l by the number of the first three digits of red blood cells count in 1 liter of blood with multiplication of the received value by 3: **CI = 3 × HGB / RBC**. For example, the blood hemoglobin content is 152 g/l, the erythrocyte count is $4.56 \times 10^{12}$/l; then CI is equal to 3 × 152 : 456 = 1.00.

CI of a healthy person is **0.8–1.05** (*normochromia*). In decreased hemoglobin content in Red Blood Cells CI is **less than 0.8** (*hypochromia* that usually occurs in iron deficiency in the organism), in **increased — over 1.05** (*hyperchromia* which is noted in insufficiency of vitamin $B_{12}$ and/or folic acid in the organism).

**Directions for recording the Protocol**:

Calculate MCH andCI of the tested blood using the data of work 15.3. (fig. 15.1)

**PROTOCOL**

1. Hemoglobin content in tested blood is equal to _____ g/l.
   Red Blood Cells count in tested blood is equal to _____ × $10^{12}$/l.

| Index | Normal range |
|---|---|
| **MCH** = : = | |
| **CI** = 3 × : = | |

2. **Conclusion**: _____ (normo-, hypo- or hyperchromia)

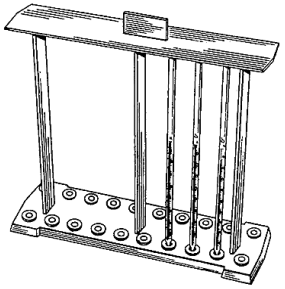
## WORK 15.2. ESR EVALUATION BY PANCHENKOV'S METHOD (demonstration)

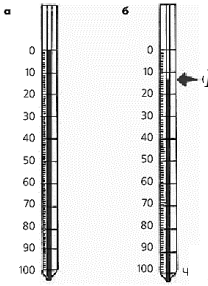
Unless the blood is not coagulated, red blood cells sediment to the test-tube bottom as their specific weight (1.096 g/ml) is higher than that of plasma (1.027 g/ml). **Normal values of erythrocyte sedimentation rate (ESR)** in healthy people are: **in male 1–10 mm/h**; **in female 2–15 mm/h**. The *most important factors affecting ESR* are the proportion of various kinds of blood plasma proteins as well as red blood cells content. An increase of globulins and fibrinogen, and/or decrease of albumins in plasma as well as decrease of red blood cells is associated with an increase of ESR. An increase of red blood cells in the blood as well as an increase of albumin and bile pigments results in a decrease of ESR. A higher value of the ESR norm in women is associated with a less red blood cells content.

Under physiological conditions an increased ESR is noted during pregnancy, in eating dry food and fasting, after vaccination (due to an increase of globulins and fibrinogens in plasma). Delayed ESR can be noted in blood thickening due to enhanced perspiration (for example, in high external temperature) or enhanced formation and content of erythrocytes in blood (for example, in Alpine residents and mountaineers).

Many diseases are accompanied by ESR changes. Thus, an increased ESR is noted in the majority of infectious, inflammatory and autoimmune diseases (due to hyper-globulinemia and/or hyperfibrinogenemia), kidney diseases with nephrotic syndrome (due to a loss of albumins with urine and development of hypoalbuminemia), malignant tumors and hemoblastoses (due to an increased content of large-molecular proteins in the blood and/or depression of erythropoiesis and development of anemia), endocrine diseases (thyrotoxicosis and diabetes mellitus) and anemia of different genesis. A decreased ESR, up to a complete stop of sedimentation, occurs in erythrocytosis.

**Materials and equipment**: Panchenkov's device, a watch glass or test tube, scarifiers in sterilizers or disposable, rubber gloves, masks, sterile cotton wool, antiseptic, iodine, 3 % solution of chloramine, 5 % solution of sodium citrate.






**Accomplishment**. Panchenkov's device is used to evaluate ESR. A pipette (capillary) of the device is washed with 5 % solution of sodium citrate. The taken blood is carefully stirred with sodium citrate on the watch glass. The mixture is drawn into the pipette to mark 0. The pipette is placed into the stand for 1 hour in a strictly vertical position. The result is assessed by a decrease of a red column of red blood cells in the capillary from point 0 (in millimeters).

While evaluating ESR this should be followed strictly: the proportion of sodium citrate and blood 1:4; verticality of the pipette in the stand; the temperature in the room — 18–22 °C (in lower temperature ESR decreases and in higher — increases).

**PROTOCOL**

1. ESR of tested blood = _____ mm/h. Tested person sex is _____.
2. ESR reference range (normal values): in males _____ mm/h; in fimales _____ mm/h;
3. While evaluating ESR the blood is mixed with 5% solution of Na citrate with the aim _____
4. **Conclusion**: ESR is _____ (in norm, increased or decreased)

## WORK 15.3. PHYSIOLOGICAL ASSESSMENT OF THE COMMON CLINICAL BLOOD TEST

Common clinical blood test (hematology tests) is one of the most common laboratory examinations. It includes evaluation of the following indices:

1) Red Blood Cells count per 1 liter of blood;
2) Hemoglobin content (g/l);
3) calculation of Color Index;
4) White Blood Cells count per 1 liter of blood;
5) Leukocyte formula (WBS differentiation);
6) Erythrocyte Sedimentation Rate (ESR). Result depends on technique (e. g. Panchenkov, Westergren, Wintrobe or Seditainer).

Additional examinations include: evaluation of platelets in 1 liter of blood, count of reticulocyte percentage and some other indices. Modern hematologic analyzers allow additional evaluation of: the hematocrit, mean volumes of Red Blood Cells, White Blood Cells and platelets; mean hemoglobin content in Red Blood Cell, etc.

Using common blood test indices the physician may assess the respiratory function of the blood (by the Hemoglobin content, Red Blood Cells count); erythropoesis intensity (by the reticulocyte count); suggest the presence of infectious, inflammatory and autoimmune processes in the organism (by the White Blood Cells count, "left shift" of the leukocyte formula and ESR changes) etc.

**Directions for recording the Protocol**:

Fill in the table of the common clinical blood test indices.

**PROTOCOL**

| Factor | Normal range | Main function |
|---|---|---|
| 1. Red Blood Cells (RBC) | $(3,9–5,1) \times 10^{12}$/l, male<br>$(3,7–4,9) \times 10^{12}$/l, female | |
| 2. Hemoglobin (HGB) | 130–170 g/l, male<br>120–150 g/l, female | |
| 3.Color index (CI) | 0,8–1,05 | |
| 4. White Blood Cells (WBC) | $(4–9) \times 10^{9}$/l | |
| 5. Leukocyte formula: | Per 100 cells (100 %) | |
| 6.1. Basophils | 0–1 % | |
| 5.2. Eosinophils | 1–5 % | |
| 5.3. Neutrophils:<br>myelocytes<br>young<br>rod nuclear<br>segmented | <br>0 %<br>0–1 %<br>1–5 %<br>46–68 % | |
| 5.4. Monocytes | 2–9 % | |
| 5.5. Lymphocytes | 18–40 % | |
| 6. Erythrocyte sedimentation rate (ESR) | 1–10 mm/h, male<br>2–15 mm/h, female | |

Abbreviations used for hematologic tests

1. WBC (White Blood Cells) — total leukocyte count;
2. RBC (Red Blood Cells) — erythrocyte count;
3. Hb or HGB (Hemoglobin) — hemoglobin content;
4. HCT (Hematocrit) — hematocrit factor;
5. MCV (Mean Corpuscular Volume) — mean Red Blood Cells volume;
6. MCH (Mean Corpuscular Hemoglobin) — mean hemoglobin content in an Red Blood Cell;
7. MCHC (Mean Corpuscular Hemoglobin Concentration) — hemoglobin content in 100 ml of Red Blood Cells (hemoglobin concentration in one Red Blood Cell);
8. PLT (Platelets) — thrombocyte count;
9. W-SCR — percentage of small leukocytes, i.e. lymphocytes;
10. W-LCR — percentage of large leukocytes, i.e. total percentage of neutrophils + monocytes + basophils + eosinophils;
11. W-SCC — or LYMPH — absolute small leukocyte count, i.e. lymphocytes;
12. W-LCC — or MO + GR — absolute count of large cells, i.e. total count of neutrophils + monocytes + basophils + eosinophils;
13. RDW (Red Cell Distribution Width) — distribution width of Red Blood Cells by the volume;
14. PDW (Platelet Distribution Width) — distribution width of platelets by the volume;
15. MPV (Mean Platelet Volume) — mean thrombocyte volume.

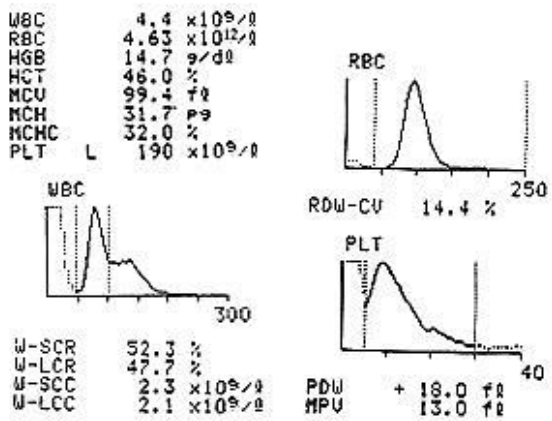



Fig.15.1

## Lesson 16. BLOOD GROUP SYSTEMS. BLOOD PREPARATIONS. BLOOD SUBSTITUTING SOLUTIONS. HEMOSTASIS

DATE OF CLASSES
«____» _____ 20____
day month year

| **Basic questions**: | LITERATURE<br>Main |
|---|---|
| 1. Human blood group systems (ABO, Rh, and other). The concept of human leukocyte antigen (HLA) system.<br>2. Antigens (agglutinogens) and antibodies (agglutinins) of ABO and Rh blood types, their characteristics. Determination of ABO and Rh system blood group. Blood typing using the standard and monoclonal sera.<br>3. The concept of blood preparations and blood substitution solutions. Principles of blood matching. Consequences of mismatched blood transfusion in ABO or Rh system.<br>4. The concept of the hemostasis system and its parts.<br>5. Primary (vascular-thrombocyte) and secondary (plasma-coagulation) hemostasis. Basic methods of evaluation. Bleeding time after tooth extraction.<br>6. Concept of an anticoagulant system. The main anticoagulants.<br>7. Concept of the fibrinolytic system, its mechanisms. | 1. Lecture & E-learning system.<br>2. *Moroz, V. M.* Physiology : textbook / V. M. Moroz [et al.] ; ed. by V. M. Moroz, O. A. Shandra. 2nd ed. Vinnitsia : Nova Knyha, 2016. P. 264–267, 281–292.<br>3. *Severina, T. G.* Physiology of blood. Lecture notes / T. G. Severina. 2nd ed. Минск : БГМУ, 2017. P. 41–52.<br>Additional<br>4. *Ganong, W. F.* Review of medical physiology / W. F. Ganong. 25th ed. McGraw-Hill Companies, Inc., 2016. P. 558–562, 564–567.<br>5. *Hall, J. E.* Guyton and Hall textbook of medical physiology / J. E. Hall. 13th ed. Elsevier, 2016. P. 477–494. |

| **BUZZWORD** | |
|---|---|
| Blood group system — | Hemostasis — |
| Agglutination — | Primary hemostasis — |
| Agglutinogen — | Secondary hemostasis — |
| Agglutinin — | Coagulation system — |
| Standart sera — | Petechiae — |
| Monoclonal sera — | Main anticoagulants — |
| Rh incompatibility — | Fibrinolysis — |

## WORK 16.1. BLOOD TYPING IN THE ABO SYSTEM USING STANDARD SERA (demonstration)

The ABO system blood type is determined by the presence of agglutinogens in red blood cells which is revealed by the hemagglutination reaction using standard sera. The interaction between red blood cells antigens of the tested blood and a corresponding antibodies (agglutinins) of the standard serum under-lies the bases of such reaction. As antibodies contained in standard sera are known, red blood cells antigens of the tested blood and consequently the blood type in the ABO system are determined by the presence or absence of agglutination.

**Materials and equipment:** standard sera of Oαβ(I), Aβ(II), Bα(III) and $AB_0$(IV) groups of two various series; pipettes for them; special plate; glass sticks; isotonic (0.9 %) solution of NaCl; scarifiers in sterilizers or disposable; cotton wool; antiseptic; iodine; rubber gloves; masks; disinfectant solution.

**Standard serums of Oαβ(I), Aβ(II), Bα(III), and $AB_0$(IV) groups of two various series**

**Accomplishment**. Blood typing should be done in the room with sufficient illumination and at the temperature of **15–25 °C**.

Determination is done on special plate. 0.1 ml (1 large drop) of every standard serum of two series is applied to appropriate depressions of the plate.

The blood for the test is taken from the finger in compliance with all necessary rules. The first blood drop is taken off with a gauze ball. Then the blood is added with glass sticks **(5–10-fold less than the serum)** to every drop of the serum and carefully stirred. The obtained mixture is mixed again by rocking the plate.


**Special plate**

scarifiers

**Four different combinations of the reaction are possible:**

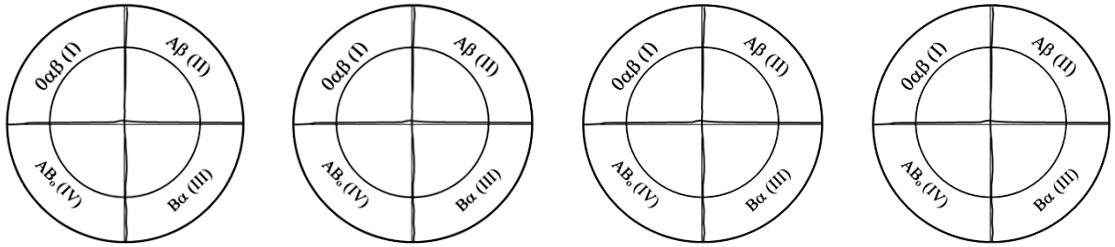
1. Agglutinins of standard serums of all 3 groups did not cause agglutination, and all drops stayed regularly stained in red. In this case the blood belonged to group Oαβ(I) (*type O*).

2. Agglutinins of standard serums of groups Oαβ (I) and Bα (III) caused a positive reaction of agglutination, and serums of group Aβ (II) — a negative one. The tested blood belongs to group Aβ (II) (*type A*).

3. Agglutinins of standard serums of groups Oαβ (I) и Aβ (II) caused a positive reaction of agglutination, while serums of group Bα (III) — a negative one. The tested blood belongs to group Bα (III) (*type B*).

4. Agglutinins of standard serums of all three groups caused a positive reaction of agglutination. The tested blood belongs to ABo(IV) group (*type AB*).

*Draw a scheme for each blood typing experiment*:
**(blood drop 5–10-fold less than the serum, temperature 15–25 °C )**




| 0αβ(I) /type O/ | Aβ (II) (type A) | Bα (III) (type B) | ABo(IV) (type AB) |
|---|---|---|---|

In case of typing $AB_0$(IV) group, before giving such a conclusion, to exclude non-specific agglutination, it is necessary to do an additional control test with the standard serum of $AB_0$(IV) group by the same technique. The absence of agglutination in this test allows to consider the former reactions specific and refer the tested blood to $AB_0$(IV) group. The presence of agglutination with the serum of $AB_0$(IV) group reveals non-specific agglutination. In this case the test should be repeated with washed Red Blood Cells.

**Revealing other combinations of agglutination reactions testifies to improper blood typing!**

*Errors* while determining blood groups are possible in situations, when agglutination is not revealed or a false agglutination occurs.

The absence of agglutination may be due to the following causes: 1) retardation of this reaction at high temperature of the environment >25 °C (blood typing should be done only at the room temperature of 15–25 °C); 2) addition of an excess of tested blood to standard serums resulting in a decrease of agglutinin titer in their content (remember that a drop of the applied blood should be 5–10 times less than that of the serum); 3) weak activity of the standard serum or low agglutinin ability of Red Blood Cells.

Revealing false agglutination in its real absence may be due to drying of a serum drop and formation of Red Blood Cells "monetary columns" (nummiform red cells aggregarion) or appearance of cold agglutination at the temperature less than 15 °C. The addition of a drop of isotonic NaCl solution to the tested mixture of serum and blood and performing the test at the temperature higher than 15 °C allow to avoid the mentioned errors.

The reaction of agglutination is observed during 5 minutes. Usually the agglutination reaction starts during the first 10-30 seconds, however agglutination may be late, e.g. with Red Blood Cells of $A_2\beta$(II) group. As agglutination occurs, but not earlier than in 3 minutes, 1 drop of NaCl isotonic solution is added into those drops, where agglutination has already occurred, and observation is continued followed by rocking the plate for 5 minutes, and only then the final result is assessed.

The reaction in every drop may be either positive or negative. In a positive reaction there appear small red granules (agglutinates) seen with naked eye in the mixture; they consist of glued Red Blood Cells. Step-by-step they cluster and form larger granules or flakes of irregular shape. Meanwhile the serum becomes completely or partially decolorized. In case of a negative reaction the content of drops stays regularly stained in red, and agglutinates are not revealed there. The results of the reaction in both serum series should be identical.

***N o t e***. In case of a doubtful or unclear result during the first determination of blood group a repeated test of the blood group of the same blood with standard serums of other series should be done. If the results remain still unclear, the blood group should be determined by a cross-method using standard serums and standard red blood cells or monoclonal antibodies (see the supplement) in laboratory.

**Directions for recording the Protocol:**

Fill in tables 16.1 and 16.2.

Indicate in table 16.2, when agglutination occurs (+) and when doesn't (–).

*Table 16.1*

| Blood groups | Serum agglutinins | Red Blood Cells agglutinogens |
|---|---|---|
| 0αβ (I) | | |
| Aβ (II) | | |
| Bα (III) | | |
| $AB_0$ (IV) | | |

*Table 16.2*

| Blood groups | Standard serums | | | |
|---|---|---|---|---|
| | 0αβ (I) | Aβ (II) | Bα (III) | AB (IV) |
| 0αβ (I) | | | | |
| Aβ (II) | | | | |
| Bα (III) | | | | |
| $AB_0$ (IV) | | | | |

## WORK 16.1. BLOOD TYPING IN THE ABO SYSTEM USING MONOCLONAL SERA (demonstration)

**Technique for determining human blood groups of the AB0 system and Rh systems using monoclonal sera.**

Apply one large drop of anti-A, anti-B and anti-D (anti-Rh) reagents to the special tablet or porcelain plate under the appropriate inscriptions (anti-A, anti-B and anti-D). Next to the drops of reagents are placed on a small drop of the test blood (10:1 ratio). The reagent is thoroughly mixed with blood with glass sticks. Observation of the course of the agglutination reaction is carried out with a slight rocking of the plate for 1–2.5 minutes.

Agglutination with monoclonal reagents usually occurs within the first 3-5 sec. But observation should be carried out for 2.5 minutes due to the possibility of a later onset of agglutination with red blood cells containing weaker antigens.

**Accomplishment**. Work is performed using the computer program "**PhysioEx**". To get started, select “Exercise 11: Blood Analysis” → “Activity 4: Blood Typing” → “Introduction (tab in the top menu)” and study the distribution of agglutinogens on the surface of red blood cells of different groups (Figure 11.3, page 1 of 3), and method of determining blood groups using monoclonal antibodies (video: Blood Typing wet-lab video, page 2 of 3).

Click on the "Experiment" tab. Take a tablet from the Blood Typing Slide Dispenser to determine blood types and drag it onto the worktable. After that, the wells on the tablet become denoted by the symbols A, B, and Rh. Drop the testing blood “Sample 1” into the wells, starting with “A”, then drop by drop anti-A, anti-B and anti-Rh reagents. Mix the blood with reagents with the help of Stirring Sticks of the appropriate color. Dispose of used sticks in the waste biomaterial package (Biohazard).

Place the tablet on the light table on the right side and click “Light”.

Click the “positive” on the image that appears under the drops, in which blood agglutination occurred, and “negative” under the drops in which agglutation did not occur. To record the results, click “Record Data”. Throw the tablet in a package for waste biomaterials (Biohazard).

Answer the question of the program and click "Submit".

By the same way, analyze blood samples number 2-6. To start the analysis, place the tablets on the desktop. After the finishing of the last sample analyzing, determine the test blood groups by selecting the appropriate line in the electronic protocol and clicking the “A”, “B”, “AB” or “O” and “+” or “–” buttons to specify the Rh factor. Enter the data in the laboratory protocol.

After the answering the program's question: “Why people with ABO (IV) $Rh^-$ blood type are known as universal recipients” and clicking “Check Answer” → “Submit” → “Submit”, you can, if necessary, re-see the results of blood group determination by clicking "View Experiment Results".

**Directions for recording the Protocol**:

1. Record the results. Determine the blood group in the test.
2. In the conclusion, indicate what the differences in the determination of blood groups are with the help of standard isohemagglutinating sera and monoclonal anti-bodies. Explain the reason for the differences.

**PROTOCOL**

| Blood sample | Presence of agglutination (list «+» or «–» ) | | | Blood group |
|---|---|---|---|---|
| | Anti-A | Anti-B | Anti-Rh | |
| 1 | | | | |
| 2 | | | | |
| 3 | | | | |
| 4 | | | | |
| 5 | | | | |
| 6 | | | | |

Conclusion: _____

_____

_____

_____

## WORK 16.3. EVALUATION AND PHYSIOLOGICAL ASSESSMENT OF PRIMARY HEMOSTASIS INDICES

The term **hemostasis** means a complex of reactions to stop bleeding in vascular injuries and maintenance of blood liquid state in vessels.

Since bleeding and thrombus formation in vessels of various sizes have different courses, there are two basic mechanisms of hemostasis:

**microcirculatory,** vascular-thrombocyte or primary mechanism of hemostasis. It starts reactions of hemostasis in capillaries, venous and arterial vessels **up to 200 μm in diameter**. This process involves platelets and endothelium of vessels. Almost 80 % of bleedings and 95 % of thrombus formations are associated with the impairment of this mechanism.

**Macrocirculatory,** hemocoagulatory or secondary mechanism starts as a rule on the basis of the primary one and follows it. It is accomplished by the blood coagulation system. Due to the secondary hemostasis a red thrombus is formed, it consists mainly of fibrin and blood cells. It provides a final stop to bleeding from injured macro vessels (**over 200 μm in diameter**).

Primary (vascular-thrombocyte, microcirculatory) hemostasis means fast (within several minutes) formation of platelet clots at the site of vessel injury what is very important for stopping bleeding from small vessels with low blood pressure.

The components of the primary hemostasis are vascular wall, platelets and their coagulation factors.

**The primary hemostasis stages are:**

*1) **spasm of vessels** (local vasoconstriction);*

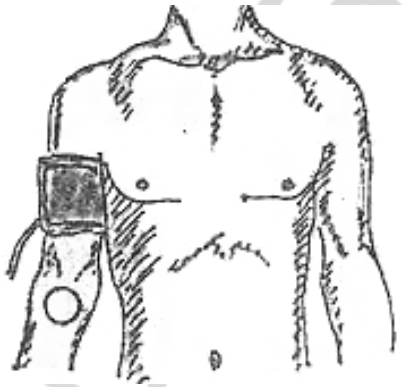
*2) **platelets adhesion** (involving Willebrand's factor), their **activation** and secretion of platelets granules (involving thromboxane $A_2$ through a phospholipase mechanism), as well as platelets **aggregation** (at first it is reversible and then irreversible due to the action of thrombin and fibrin traces) with the **formation of a platelets plug**;*

*3) **retraction** (constriction and consolidation) of the platelets plug.*

The most important screening indices characterizing the primary hemostasis are: **bandage test, platelets count, bleeding time by Aivy or Duke.**

**A. Bandage test** (evaluation of a vascular component of the primary hemostasis)

The method is based on the fact that dosed mechanic action (pressure) on skin capillaries of a healthy person does not cause any substantial changes. When the normal state of a capillary wall is impaired, increased vascular fragility occurs and after mechanic action at the site of the pressure multiple petechiae or hemorrhage appear manifesting the impairment of a vascular component of hemostasis.



**Materials and equipment**: a tonometer, a stop-watch, a circle of dense card-board 2.5 cm in diameter, a pen or a pencil.

**Accomplishment**. The test is done on the forearm. A circle 2.5 cm in diameter is outlined 1.5–2.0 cm from the ulnar pit. To do a test one should check if there are any hemorrhages in this circle (and their number if there are any). The blood pressure cuff is applied and the pressure of 80 mm Hg is created. The pressure is sustained at this level for 5 minutes pumping the air if necessary. The arm of the examined person should be relaxed and lie freely.

All **petechiae** that appeared in the outlined circle are counted in 10–15 minutes (taking into consideration those present before). In healthy persons petechiae are not formed or their number does not exceed 10 in the circle and their sizes are not more than 1 mm in diameter (negative bandage test). An increase of the petechiae number over 10 and petechiae sizes over 1 mm in diameter or the presence of a hemorrhage (positive bandage test) evidence the following: wall defects of micro vessels due to endocrine changes (menstrual period); infectious-toxic effect (sepsis etc.); insufficiency of vitamin C; the impairment of Willebrand's factor formation, etc.; the presence of thrombocytopenia or thrombocytopathia etc.

**PROTOCOL**

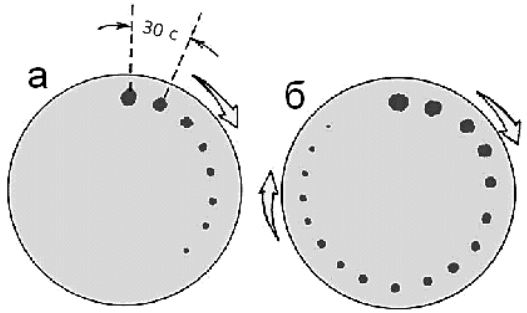
1. Petechiae number in the circle before the test _____ (no, 1, 2, 3…)
   Petechiae number in the circle in 10–15 minutes after the test ____ (no, 1, 2, 3,…).
   If petechiae are present, indicate their diameter ____ (below 1 mm or over 1 mm).
2. **Conclusion**: bandage test _____
   (negative= without petechiae or positive = with petechiae )

**B. Time of bleeding by Duke**

The time of bleeding evaluated by Duke's method gives a general idea, if the primary hemostasis function is normal (and first of all it allows evaluating the function of platelets, their ability for adhesion or aggregation). An increase of bleeding time evidences the impairment of the primary hemostasis due to thrombocytopenias, thrombocytopathias, vascular wall injuries or a combination of these factors. Reducing the bleeding time evidences only an enhanced spastic ability of peripheral vessels.

**Materials and methods**: a stop-watch, sterile filter paper, disposable scarifiers, cotton wool, antiseptic, iodine, rubber gloves, masks, disinfectant solution.

**Accomplishment**. Puncture the 4th finger-cushion to the depth of 3 mm. If this is done properly, the blood is discharged spontaneously without pressure. Having made a puncture, switch on the stop-watch. Touch the first appearing blood drop with a strip of sterile filter paper that absorbs the blood. Then take off further blood drops with sterile filter paper every 30 sec. Avoid touching the skin with filter paper, as it stimulates premature stop of bleeding. Continue till blood traces are absent on the filter paper. **In norm duration of the bleeding time by Duke is 2–4 min**.

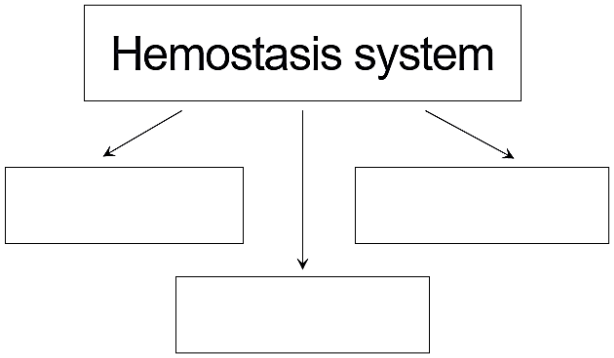



**PROTOCOL**

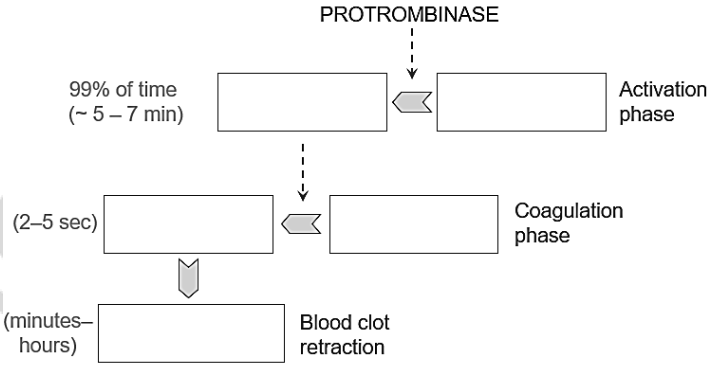
1. Bleeding time is _____ min _____ sec.
2. **Conclusion**: Bleeding time is _____
(normal, increased, reduced)

**WORK 16.4. HEMOSTASIS SYSTEM**

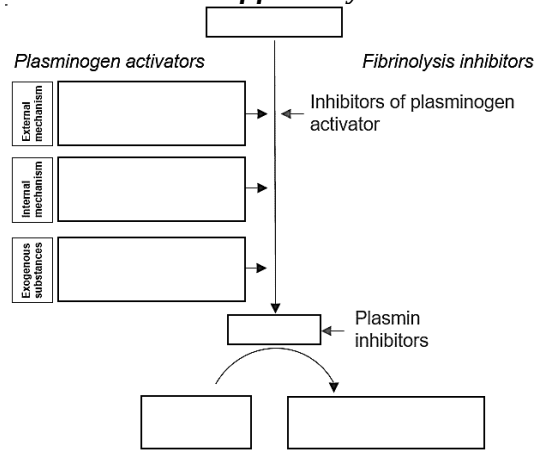
*Fill in the **general scheme of hemostasis system***




*Fill in the **scheme of coagulation by Moravitz***



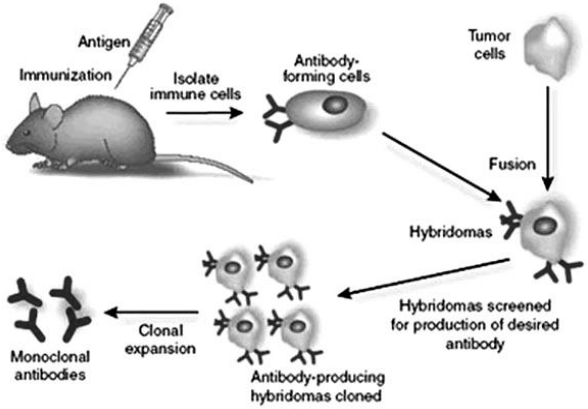

*Fill in the **scheme of fibrinolyzis***



**SUPPLEMENT. MONOCLONAL SERA: APPLICATION OF MONOCLONAL ANTIBODIES IN BLOOD TYPING**

At present ABO-typing reagents produced from the human or animal serum with antibodies to red blood cells agglutinogens are still often used. These antibodies are the result of a polyclonal immune response, i.e. they come from various clones of antibody-forming cells and are the mix of immunoglobulins of various classes. To get such serums a great amount of donor blood is needed. Besides, the titer of natural antibodies in the human blood is usually low, that is why produced serums have low activity and one has to use serums obtained from specially immunized people.

Antibody-producing technology based on the fusion of a malignant myeloma cell and an antibody-forming lymphocyte of mice, becomes more and more widespread. As a result of fusion a hybrid cell (hybridoma) is formed inheriting basic properties of its parents: immortality and the ability to constant growth — from a tumor cell, and the ability to produce antibodies — from a B-lymphocyte.




Antibodies secreted by cells-descendants of such hybrids are monoclonal, i.e. they come from one cellular clone, belong to one class of immunoglobulins, are aimed at one antigen, are standard and able to grow both in culture and in the mouse's organism as an ascite tumor producing antibodies in high concentrations, up to some tens of grams per liter.

To obtain ABO-typing monoclonal reagents it is enough to make a wash-out of tissue culture or take some ascite fluid and dilute these fluids as the titer of antibodies in them is very large (often for dilution 0.3 M solution of NaCl is used). At present ABO monoclonal reagents are commercially produced in England, Germany, Canada, Russia, Belarus, and other countries.

The benefits of monoclonal reagents are their high activity, standardization, reliability of revealing appropriate antigens, absence of false-positive reactions that is due, first of all, to the absence of antibodies of other specificity. Monoclonal reagents are not products of human cells that it excludes the possibility of transmitting viruses of hepatitis and HIV.

Two types of monoclonal reagents are necessary for blood typing — anti-A and anti-B that are produced by two different hybridomas and contain correspondingly α- and β-agglutinins.

Blood typing in the ABO system using monoclonal serums


Blood groups
Reaction of tested Red Blood Cells with monoclonal reagents

| | anti-A (α) | anti-B (β) |
|---|---|---|
| 0 (I) | – | – |
| A (II) | + | – |
| B (III) | – | + |
| AB (IV) | + | + |

Per one large drop of anti-A and anti-B reagents is applied on a special plate or a porcelain dish under corresponding signs "anti-A" and "anti-B". Next to reagent drops small drops of the tested blood are applied (proportion 10:1). The reagent is carefully mixed with the blood with glass sticks. Observation of the course of the reaction is done by rocking the plate for 1–2.5 minutes.

Agglutination with monoclonal reagents usually occurs within the first 3–5 sec. But the observation should be continued for 2.5 min due to a possibility of late agglutination with red blood cells containing weak types of antigens A and B.

THE LABORATORY WORKS ARE PASSED WITH MARK

Teacher's signature

## Lesson 17. COLLOQUIUM "BODY FLUIDS"

DATE OF CLASSES
«____» _____ 20____
day month year

**Basic questions:**

1. The role of water for vital functions. The content and distribution of water in the organism. The main fluid compartments of the body. Water balance.
2. Blood. The concept of the blood system. The composition of the blood, its physiological properties and functions. Basic physiological constants of blood.
3. Blood plasma, its quantity, composition, and properties. Hemolysis and its types.
4. Blood acid-base balance and the mechanisms of its maintaining. Buffer systems of blood.
5. Electrolyte composition of blood plasma. Osmotic blood pressure and its regulation (ADH, RAAS, and others).
6. Blood plasma proteins, their characteristics and quantity. Colloid osmotic (oncotic) blood pressure, its role in regulation of volume of the blood.
7. Erythrocytes (RBC): peculiarities of the structure and properties, their functions. RBS count. Hematocrit (HTC), its level and evaluation.
8. ESR (erythrocyte sedimentation rate): definition, factors affecting it. Diagnostic significance of ESR.
9. Hemoglobin (Hb): its concentration, structure, main types and compounds, their physiological significance. Hemoglobin evaluation methods. Color Index (CI).
10. Leukocytes (WBC), their types, quantity, properties and functions. Leukocyte formula, its peculiarities with age, "left" and "right" shifts. Leukocytosis and leukopenia.
11. Platelets (PLT), their count, structure, properties and functions.
12. The concept of the hemostasis system. Primary and secondary hemostasis and the basic methods of their assessment. The duration of bleeding after tooth extraction. The concept of anticoagulant and fibrinolytic systems.
13. Human blood group systems (ABO, Rh, and other). HLA system. Antigens (agglutinogens) and antibodies (agglutinins) of ABO and Rh blood types, their characteristics.
14. Determination of ABO and Rh system blood group. Blood typing using the standard and monoclonal sera. Principles of blood matching. Consequences of mismatched blood transfusion in ABO or Rh system. Risk factors when working with blood: for medical professionals, recipients, and donors.

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**Form of colloquium:**
***computer control test "Lesson 17"***
***at E-learning system with oral or writing part.***

| | |
|---|---|
| 15. Hematopoiesis. Stem cell, its microenvironment. Nervous and humoral mechanisms of regulation. The role of vitamins ($B_{12}$, $B_9$ and others) and trace elements ($Fe^{2+}$ and others).<br>16. Fluids of the oral cavity: oral fluid ("mixed saliva"), gingival fluid, and saliva of salivary glands. Acid-base condition of the oral cavity. | |

THE COLLOQUIUM IS PASSED WITH MARK

_____

Teacher's signature

## СПИСОК ИСПОЛЬЗОВАННОЙ ЛИТЕРАТУРЫ


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**CONTENTS**

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# РУКОВОДСТВО К ЛАБОРАТОРНЫМ ЗАНЯТИЯМ ПО НОРМАЛЬНОЙ ФИЗИОЛОГИИ
# LABORATORY MANUAL FOR NORMAL PHYSIOLOGY

Практикум для специальности «Стоматология»

На английском языке

В двух частях

Часть 1

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