MINISTRY OF HEALTH OF UKRAINE ODESA NATIONAL MEDICAL UNIVERSITY Department of Medical Biology and Chemistry

> CONFIRMED by Vice-rector for scientific and pedagogical work Eduard BURIACHKIVSKYI

# METHODOLOGICAL DEVELOPMENT TO <u>THE INDEPENDENT WORK</u> ON THE EDUCATIONAL DISCIPLINE

Faculty, courseInternational faculty, 3d yearSpecialty226 "Pharmacy, industrial pharmacy"Academic disciplineBiological chemistry

The program was approved: at a meeting of **the Department of Medical biology and chemistry** Odesa National Medical University Minute  $N_{\underline{0}} \underline{1}$  dated  $\underline{26}$  august 2024 Head of Department \_\_\_\_\_\_\_\_\_ Hennadii STEPANOV

# **Developers:**

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#### Practical class №1

Topic: General characteristics, properties of enzymes. The protein nature of enzymes. Active, allosteric enzyme centers. Regulation of enzyme activity. Enzyme activators and inhibitors. Coenzymes, their role in catalysis. Coenzyme functions of vitamins. International classification and nomenclature of enzymes. Proenzymes. Activation of proenzymes, role in metabolism.

Goal: Learning basic laboratory methodsdetermination of the structure of enzymes

**Basic concepts:**<u>enzymes, amino acids, peptide, ester, glycosidic bonds, hydrolysis, food</u> <u>digestion</u>

Equipment: Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

-the chemical nature of enzymes, their structure, biological role and basic properties of proteins as biocatalysts;

-types of chemical reactions that form the basis of the International Classification of Enzymes and the principles of their nomenclature;

-characteristics of individual classes of enzymes and give examples of each class of enzymes; -meaning of four-digit codes of enzymes;

-specificity and types of enzyme specificity;

-the role of active and allosteric centers in the action of enzymes;

-principles of enzyme activity regulation;

-the mechanism of action of activators and inhibitors on the activity of enzymes (for example, amylase of saliva);

-diagnostic value of determination of amylase activity in urine (for pancreatitis).

The higher education applicant should be able to:

-determine the chemical nature of enzymes using reactions to proteins and amino acids

-determine the activity of amylase (diastase) in biological fluids (saliva and urine) in the presence of activators and inhibitors;

-trace and evaluate the action of amylase activators and inhibitors.

Questions to check basic knowledge on the topic of the class:

-Stages of development of the teaching about enzymes

-The role of domestic scientists in enzymology

-Chemical nature of enzymes

-Evidence of the chemical structure of enzymes

-Types of connection in the structure of proteins

-Levels of structure in protein molecules

-Principles of protein detection in solutions

-What are the main types of chemical reactions you know from the bioorganic chemistry course

-Which compounds are characterized by peptide, ester, and glycosidic bonds

-Chemistry of the biuret reaction

-Foley reaction principle

–What determines the speed of chemical reactions

-Factors affecting the rate of an enzymatic reaction

-Activation energy of chemical reactions

-Mechanism of acceleration of chemical reactions by enzymes

3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Determining the structure of the enzyme using the biuret reaction".

Recommendations for performing tasks.

Principle of the method: discovery of peptide bonds in proteins and peptides. These substances form a red-violet complex with copper sulfate in an alkaline environment.

Procedure: Pour 0.5 ml of the test solution into a test tube, add 0.5 ml of 10% NaOH solution and 1-2 drops of 1% CuSO4 solution and mix. In the presence of protein, a red-purple color appears.

Conclusion: the appearance of a red-violet color indicates the presence of peptide bonds in the enzyme molecule, that is, its protein nature.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.2 Demonstration and practical work "Foley's reaction to sulfur-containing amino acids". *Recommendations for performing tasks.* 

The principle of the method: when boiling with alkalis, sulfur, which is separated from sulfurcontaining amino acids, forms sodium sulfide, which with lead acetate gives a dark precipitate of lead sulfide.

Procedure: 5-6 drops of 30% NaOH solution and 1-2 drops of lead acetate are added to 0.5 ml of the test solution. The contents of the test tube are boiled. With a positive reaction to sulfur, the liquid in the test tube darkens.

Conclusion: the structure of the enzyme contains sulfur-containing amino acids.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.3 Demonstration and practical work "Determination of salivary amylase activity in the presence of activator and inhibitor".

Recommendations for performing tasks.

The principle of the method: the amylase activity of saliva and urine is determined by the amount of substrate (starch) broken down by 1 ml of saliva or urine over a set period of time (30 minutes), and is based on finding the maximum dissolution at which the investigated liquid breaks down starch to the stage of red coloration with iodine (formation of erythrodextrins). Sodium chloride accelerates the cleavage of starch under the action of amylase. Copper sulfate slows down the action of amylase. Normally, human urine contains little amylase. The amount of amylase in urine increases with pancreatitis, parotitis, orchitis.

Progress:

a) Preparation of a number of dilutions of saliva. Prepare three rows of tubes of 10 each. Pour 1 ml of distilled water from a burette into all test tubes. Add 1 ml of saliva, diluted 10 times, to test tubes under  $N_{2}1$ . Stir the liquid. Transfer 1 ml of the resulting saliva solution to test tube #2. Stir. Transfer 1 ml of liquid from the test tubes under  $N_{2}2$  to the test tube under  $N_{2}3$ , etc. Pour 1 ml of liquid from test tubes under  $N_{2}10$ . 3 rows of tubes with diluted saliva were obtained, in each subsequent tube the concentration of the enzyme is 2 times lower than in the previous one.

b) Addition of activator and inhibitor. Add 1 ml of water to 10 test tubes of the 1st row. This series will serve as a control against which amylase activity in the presence of activator and inhibitor is compared. Add 1 ml to the second row of test tubes. 0.85% NaCl solution, which is an amylase

activator. In the third row of test tubes, add 1 ml of CuSO4 solution, which inhibits the action of amylase.

c) Preparation of enzyme-substrate mixture. Pour 2 ml of starch solution into all test tubes and mix quickly. Start with test tube 10, that is, with the highest dilution of saliva, where the enzyme concentration is the lowest.

d) Incubation. Place all 30 tubes for 30 minutes in a water bath at 37°C. Add 2 drops of iodine (Lugol's solution) to each test tube, mix and observe the color range from yellow to blue.

Conclusion: In the test tube, where the liquid is blue, the cleavage of starch did not occur. Sufficient cleavage occurs, for example, in test tube N $_{2}$ 5, where saliva is diluted 320 times, that is, 1/320 ml of saliva splits 2 ml of 0.1% starch solution, and 1 ml of undiluted saliva splits X ml of starch.

$$X = 320 \cdot \frac{2}{1} = 640$$
 од.

This is depicted as:

A (амілазна активність) 
$$=\frac{37^0}{30^{"}}=640$$
 од.

Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

- Common and divergent structures of simple and complex enzymes
- Chemical nature of cofactor, apoenzyme, holoenzyme
- Enzyme activators
- Enzyme inhibitors. Reversible and irreversible inhibition
- Allosteric centers of enzymes
- The nature of allosteric effectors
- Methods of determination of enzyme activity
- Enzyme activity units
- The structure of the active center of enzymes
- Chemistry of determination of amylase activity in biological fluids

Test tasks.

1 What are enzymes?

- a) inorganic cell catalysts
- c) organic compounds that can trigger new reactions
- c) inhibitors of chemical reactions
- +d) biological catalysts of protein nature
- e) reaction substrates
- 2 What is catalysis?
- a) inhibition of the rate of chemical reactions

c) acceleration of chemical reactions contrary to the laws of thermodynamics

+c) change in the speed of chemical reactions under the influence of catalysts

d) the ability of enzymes to initiate reactions contrary to the laws of thermodynamics

f) the course of a chemical reaction without an enzyme

3 What causes the high specificity of enzymes?

- +a) the unique structure of the apoenzyme
- c) the unique structure of the coenzyme

c) conformational and electrostatic complementarity between the substrate and the enzyme

d) conformational and electrostatic complementarity between coenzyme and apoenzyme

f) the unique structure of the cofactor

4 What is the absolute specificity of an enzyme?

+a) the ability to catalyze the transformation of only one substrate

c) the ability to catalyze the breaking of one specific bond

c) the ability to catalyze the transformation of one specific group of substrates

d) the ability to catalyze the transformation of all substrates

e) the ability to catalyze the transformation of the general group of stereoisomers

5 What is the relative specificity of enzymes?

a) the ability to catalyze the transformation of a certain group of substrates

c) the ability to catalyze the transformation of only one substrate

+c) the ability to catalyze the transformation of a group of substrates with a certain type of bonds,

d) the ability to catalyze the transformation of the general group of stereoisomers

f) decrease in enzyme activity

6 What is an apoenzyme?

a) vitamins

+c) protein part of complex enzymes

c) hormones

d) enzyme cofactors

f) nucleotides

7 What is the prosthetic group of a two-component enzyme?

a) non-protein part of the enzyme that easily dissociates from the complex with the apoenzyme

c) vitamins

c) the protein part of the enzyme

+d) non-protein part of the enzyme, which is strongly (covalently) connected to the apoenzyme

f) hormones

8 What are the main functions of apoenzyme?

a) enhances the catalytic activity of the non-protein part of the enzyme

+c) determines the specificity of enzyme action

c) participates in the act of catalysis and stabilizes the enzyme

d) is responsible for the connection between the coenzyme and the substrate

f) is in direct contact with the substrate

9 What is the active site of an enzyme?

a) active region of the non-protein part of the enzyme

+c) part of the enzyme molecule on which the substrate is converted

c) a unique combination of active sites of apo- and coenzyme

d) the region of the apoenzyme responsible for contact with the substrate

f) coenzyme site

10 What temperature is optimal for the action of most enzymes?

a) 28-32 °C

+c) 37-43 °C

c) 45-50 °C

d) 50-60 °C

f) 90-100 °C

11 What is the optimal pH value for the action of pepsin?

a) pH 4.5-5.0

c) pH 6.8-7.0

c) pH 7.5-8.5

+d) pH 1.5-2.5

e) pH 9.3 10.5

12 What criterion is the basis for the classification of enzymes?

a) chemical nature of enzymes

c) chemical nature of substrates

+c) type of catalyzed reaction

d) the type of chemical bond for which the enzymatic reaction is carried out

f) chemical nature of inhibitors

13 How many classes are enzymes divided into?

a) by four

+c) by six

c) by twelve

d) by two

f) by nine

14 What reactions are catalyzed by oxidoreductases?

a) transfer of groups, radicals, etc.

c) dehydration

+c) redox

d) isomerization

f) synthesis

15 Which enzymes belong to the second class?

a) aldolase, aminotransferase, lipase

+c) aminotransferase, phosphotransferase, CoA-transferase

c) pepsin, trypsin, carbonic anhydrase

d) aspartate decarboxylase, ATPase, fumarate hydratase

e) hexokinase, pepsin, pyruvate decarboxylase

16 How are enzymes classified according to modern classification and nomenclature?

+a) for classes, subclasses, subsubclasses, where each enzyme has a serial number

c) on classes and subclasses, where each enzyme has a serial number

c) into groups and subgroups, where each enzyme has a serial number

d) into types and subtypes, where each enzyme has a serial number

f) only for subclasses

4. Summing up.

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

### Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

# Practical class №2

Topic: <u>Basic theories of biocatalysis. Kinetics of catalysis. Methods of qualitative</u> and quantitative determination of enzymes. Intracellular localization of enzymes. Use of enzymes in the clinic (fundamentals of medical enzymology). Enzymodiagnosis, enzyme therapy. Enzymopathies.

**Goal:** <u>Study of the basics of the theory of biocatalysis, kinetics of enzymatic reactions and the use of these data in medical enzymology</u>

**Basic concepts:** biocatalysis, dissociation constant, Michaelis-Menten constant, dependence of enzymatic reaction rate on substrate concentration, isoenzymes, enzyme diagnostics, enzyme therapy, hereditary enzymopathies

Equipment: Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

-cellular organization and interrelationship of enzymes;

-peculiarities of action of individual forms of enzymes and their importance in tissue metabolism;

-the role of isozyme spectra in metabolism and their significance in enzymodiagnostics;

-basics of enzyme diagnostics and enzyme therapy;

-enzyme systems that are damaged in the most common enzyme diseases.

The higher education applicant should be able to:

-to isolate cell organelles and blood components by centrifugation;

-make an assumption about a possible pathology based on the analysis of enzyme activity.

Questions to check basic knowledge on the topic of the class:

- Cellular organization of enzymes;
- The role of membranes in biocatalysis;
- Enzymodiagnostics and its role in medical practice;
- Enzyme therapy and its significance in medicine;
- Isoenzymes, their importance in biocatalysis;
- Organ specificity of LDH isozymes;
- The principle of detection of LDH isozymes.
- 3. Formation of professional skills and abilities.

3.1Evaluation of blood enzymograms of healthy and patients with diseases of the cardiovascular system and liver.

#### Recommendations for performing tasks.

Principle of the method: As a result of differences in the primary structure of the apoenzyme, isoenzymes have different affinity for the substrate and different physicochemical properties, in particular, mobility in an electric field. Therefore, they are separated by electrophoresis in an agar or polyacrylamide gel, incubated in a substrate-coenzyme mixture, and then stained using special dyes.

Procedure: 0.01 ml of biological fluid (blood serum, homogenate) is applied to the strip of polyacrylamide gel, the gel is placed in an electric field for 2 hours, and then incubated in a mixture containing buffer, substrate, coenzyme, phenazine metasulfate and nitroblue tetrazolium. After incubation, the electrophoregram is subjected to densitometry.



Conclusion: The LDH isoenzyme spectrum of skeletal muscles and myocardium is significantly different: LDH1 and LDH2, which function in aerobic conditions, prevail in the myocardium, and LDH5 and LDH4, which function in anaerobic conditions, prevail in skeletal muscles. When various organs are damaged, LDH isoenzymes are released into the blood, which prevail in the affected tissue due to impaired permeability of plasma membranes.

*Requirements for work results.* Enter the obtained data into the workbook. Make medical and biological conclusions.

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3.2 Demonstration and practical work "Observation of the kinetics of lipase action on milk fat. The influence of bile on lipase activity".

#### Recommendations for performing tasks.

Principle of the method: The speed of the enzymatic reaction can be determined by the amount of substrate split per unit of time. To study the kinetics of the reaction in separate portions of the enzyme-substrate mixture, which contains fat and lipase, the amount of acids formed is determined by determining the time interval. The results of the determination are expressed graphically. The graph shows that the hydrolysis of fat proceeds quickly in the first 15 minutes of incubation, then stops completely at the end. This course of the process is caused by a constant decrease in the amount of the substrate and an increase in the cleavage products. When bile is added to the sample, lipase is activated, and fat hydrolysis proceeds at a faster rate.

#### Progress:

a) Preparation of enzyme-substrate mixture. Measure 10 ml of boiled diluted milk 1:10 and 1 ml of extract from the pancreas, which contains lipase, into 2 chemical beakers of 50 ml each. Add 1 ml of water to one of the glasses, bile to the second. Mix the liquid in the glasses. Take 2 ml of the mixture from each glass, transfer it to a 50 ml flask, add 1-2 drops of phenolphthalein solution, titrate with a 0.01 N solution to a faint pink color. Mark the volume (ml). Write in the table.

b) Incubation with sampling for 15-30-45 minutes. Place the mixture left in the glass in a water bath at a temperature of 370 C. Every 15 minutes. take 2 ml of the mixture and titrate with 0.01 N NaOH solution. Record the titration time and amount of alkali in the table.

c) Graphic representation of fat hydrolysis. From the obtained data for determining the amount of alkali neutralized by acids formed from milk at different stages of incubation, plot a graph, put the time of incubation on the abscissa axis. in min. (15-45) on the ordinate axis - volume of alkali (ml) of each sample.

time	Incubation	Volume of alkali used for titration (ml)				
	(min.)	Sample without bile	Bile test			
	0					
	15					
	30					
	45					

Medical and biological evaluation of the obtained results.

In a sample with bile, fat hydrolysis proceeds faster than without it. Based on the evaluation of the curves on the graph, the activating role of bile was noted in the conclusions.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.3 Demonstration and practical work "The influence of bile on lipase activity".

#### Recommendations for performing tasks.

Principle of the method: The speed of the enzymatic reaction can be determined by the amount of substrate split per unit of time. To study the kinetics of the reaction in separate portions of the enzyme-substrate mixture, which contains fat and lipase, the amount of acids formed is determined by determining the time interval. The results of the determination are expressed graphically. The graph shows that the hydrolysis of fat proceeds quickly in the first 15 minutes of incubation, then stops completely at the end. This course of the process is caused by a constant decrease in the amount of the substrate and an increase in the cleavage products. When bile is added to the sample, lipase is activated, and fat hydrolysis proceeds at a faster rate.

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	30				
	45				

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Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

- Compartmentalization of enzymes
- Enzymopathies of carbohydrate metabolism
- Enzymopathies of amino acid metabolism
- Enzymatic parameters that can be used to detect myocardial damage
- Enzyme indicators that can be used to detect liver damage
- Enzyme diagnosis of muscular dystrophies
- Enzyme therapy for hereditary pathology

Test tasks.

1 What is catalysis?

a) inhibition of the rate of chemical reactions

c) acceleration of chemical reactions contrary to the laws of thermodynamics

+c) change in the speed of chemical reactions under the influence of catalysts

d) the ability of enzymes to initiate reactions contrary to the laws of thermodynamics

f) the course of a chemical reaction without an enzyme

2 What temperature is optimal for the action of most enzymes?

a) 28-32 °C

+c) 37 – 43 °C

c) 45-50 °C

d) 50-60 °C

f) 90-100 °C

3 What is the optimal pH value for the action of pepsin?

a) pH 4.5-5.0

c) pH 6.8-7.0

- c) pH 7.5-8.5
- +d) pH 1.5-2.5
- f) pH 9.3-10.5

4 What is the Michaelis constant?

a) a value equal to the concentration of the enzyme at which the reaction rate is maximal

c) ratio of enzyme and substrate concentrations

c) a value equal to the concentration of the reaction product at which the reaction rate is half of the maximum

+d) a value equal to the concentration of the substrate at which the reaction rate is half the maximum

e) a value equal to the concentration of the enzyme at which the reaction rate is half the maximum

5 What mechanisms of lowering the energy barrier occur during enzymatic reactions?

a) formation of additional covalent bonds between apo- and coenzyme

+c) formation of an intermediate enzyme-substrate complex

c) participation of macroergic compounds in enzymatic catalysis

d) reducing the area of the contact area between the enzyme and the substrate

f) participation of additional compounds in enzymatic catalysis

6 What are the stages of the catalytic action of enzymes?

a) initial and terminal

c) preparatory, initial, working and terminal

+c) attachment of the substrate to the enzyme, conversion of the substrate, cleavage of the final products of the reaction from the enzyme

d) activation of the substrate-enzyme complex of substrate conversion, cleavage of the modified enzyme from the substrate or reaction product

e) preparatory and terminal

7 The child was diagnosed with galactosemia. The concentration of glucose in the blood does not change significantly. Deficiency of which enzyme causes this disease?

+a) galactose-1-phosphate-uridyltransferases

c) amyl-1,6-glucosidases

c) phosphoglucomutase

d) galactokinase

e) glucokinase

8 Cataract (clouding of the lens), mental retardation, enlarged liver, galactosemia, galactosuria were found in the sick child. Which enzyme is deficient in the child's body?

+a) galactose-1-phosphate-uridyltransferases

c) galactokinase

c) sorbitol dehydrogenase

d) phosphoglucosmutase

f) amyl-1,6-glucosidases

9 In a patient with acute pancreatitis, the activity of one of the specified enzymes is sharply increased in the blood and urine, which confirms the diagnosis of the disease:

+a)  $\alpha$ -amylase

c) pepsin

c) dipeptidase

d) sucrase

e) hexokinase

10 Name the enzyme, the determination of which in the blood is the most informative in the first hours after the occurrence of a myocardial infarction:

+a) creatine phosphokinase

c) alanine aminotransferase

c) pyruvate decarboxylase

d) diastasis

f) dihydrolipoyltransacetylase

11 Name the enzyme whose determination in the blood is the most informative in Botkin's disease:

a) creatine phosphokinase

+c) ornithine carbomoyltransferase

c) lactate dehydrogenase

d) diastasis

f) pepsin

12 Trasilol and Kontrical are used to treat pancreatitis. What biochemical processes do these drugs affect?

+a) reduce the activity of proteinases

c) increase the activity of proteinases

c) increase amylase activity

d) reduce amylase activity

f) increase the activity of amylo-1,6-glucosidase

13 Congenital oligophrenia is accompanied by inhibition of the conversion of phenylalanine into tyrosine. A sign of the disease is the accumulation of organic acids in the body:

+a) Phenylpyruvine

c) Lemon

c) Pyruvinogradnoi

d) Dairy

f) Glutamine

14 An increase in LDH activity is characteristic of heart and liver diseases. What additional research should be conducted to clarify the localization of the pathological process?

+a) Determination of LDH isozymes

c) Determination of amylase activity

c) Determination of alkaline phosphatase activity

d) Determination of creatine kinase isoenzymes

f) Determination of aminotransferase activity

15 An increase in the activity of which enzyme can be observed in a patient with an early stage of muscular dystrophy:

+a) Creatine kinases

c) Alanine aminotransferases

c) Collagenases

d) Hyaluronidase

f) Glutaminase

16. The patient was diagnosed with toxic hepatitis, which arose against the background of the use of drugs. The activity of which serum enzymes should be determined to confirm this diagnosis?

+a) Ornithine carbomoyltransferases

c) Creatine phosphokinase

c) Pyruvate dehydrogenase

d) Maltases

f) Malate dehydrogenase

17 When prostate cancer metastasizes to other tissues, acid phosphatase activity increases in the serum. What class does this enzyme belong to?

+a) Hydrolases

c) Oxidoreductases

c) Transferases

d) Lyases

f) Synthetases

18 The patient complains of pain in the chest area that is not relieved by nitroglycerin, weakness, increased sweating. He has cyanosis of the lips, pallor of the skin, and bradycardia. 4 hours have passed since the beginning of the anginal attack. Determining the activity of which enzyme will make it possible to make a diagnosis of myocardial infarction?

+a) Creatine kinases (MV)

c) LDH4

c) LDH5

d) ASAT

f) AlAT

19 The patient, 55 years old, was hospitalized in the infectious department with a diagnosis of viral hepatitis. An increase in the activity of which organ-specific liver enzyme will be observed in the patient's blood serum?

+a) Sorbitol dehydrogenase

c) Acid phosphatases

c) Lactate dehydrogenase

d) Cholinesterases

f) Creatine kinases

20 The temperature at which the enzyme denatures:

a) 0 °C

+c) 80-100 °C

c) 20-30 °C

d) 30-40 °C

e) 92 °C

4. Summary:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

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7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

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1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

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Electronic information resources:

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- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

### Practical class №3

Topic: <u>Specific and general pathways of catabolism. Tricarboxylic acid cycle</u> (TCA). Sequence of reactions and characteristics of enzymes. Biological value of CTC. Bioenergetics of CTC. Anaplerotic and amphibolic reactions of the cycle of tricarboxylic acids.

**Goal:** to study the general ways of catabolism of biomolecules in living cells, as well as the sequence of reactions and the biological significance of the cycle of tricarboxylic acids as a universal way of oxidative catabolism of biomolecules

Equipment: Laboratory of the department

# Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- interpret patterns of biochemical metabolic features of catabolic, anabolic and amphibolic metabolic pathways;

- to analyze the regularities of the functioning of the tricarboxylic acid cycle and the mechanism of its regulation;

- explain the structure and significance of the pyruvate and  $\alpha$ -ketoglutarate dehydrogenase complex;

- to reveal the essence and meaning of anaplerotic reactions of CTC.

The higher education applicant should be able to:

-write the reactions of the cycle of tricarboxylic acids;

- -calculate the energy balance of the CTC;
- -give examples of anaplerotic and amphibolic reactions of CTC.

Questions to check basic knowledge on the topic of the class:

- Write the formulas of pyruvic, α-ketoglutaric and oxalic-acetic acids.
- Write the formulas of citric, isolimic, cis-aconic and oxalic-succinic acids.
- Write the formulas of succinic, fumaric and malic acids.
- 3. Formation of professional skills and abilities.
- 3.1 Demonstration and practical work "Detection of milk dehydrogenase".

#### Recommendations for performing tasks.

The principle of the method is that if you take formaldehyde as the oxidation substrate (hydrogen donor) and methylene blue as the hydrogen acceptor, then under the action of milk dehydrogenase, formic aldehyde will be oxidized by removing hydrogen, which is attached to methylene blue, being restored to a colorless compound. In the form of a scheme of reactions that take place in this case, it can be depicted as follows:



Reagents:

1. 1% formaldehyde solution.

- 2. Methylene blue solution.
- 3. Milk.

Equipment:

- 1. Test tubes.
- 2. Pipettes.
- 3. Electric tiles.

The main stages of the work:

a) pour 4-5 ml of milk into 2 test tubes;

b) boil the contents of the second test tube;

c) add 8-10 drops of formaldehyde solution and 1-2 drops of methylene blue solution to both test tubes;

d) mix the contents of both test tubes, place them in a water bath (at 37°C).

After a few minutes, observe the coloration of methylene blue in the first tube and its absence in the second tube.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.2 Demonstration and practical work "Detection of succinate dehydrogenase in muscles". *Recommendations for performing tasks.* 

The principle of the method is that succinate dehydrogenase catalyzes the oxidation of succinic acid to fumaric acid. In the presence of succinate dehydrogenase, succinic acid and methylene blue, the oxidation of succinic acid and the reduction of methylene blue occur. At the same time, the latter is discolored. The reaction proceeds according to the following scheme:



- 1. Muscle porridge.
- 2. Neutralized solution of succinic acid.
- 3. Methylene blue solution.
- Equipment:
- 1. Test tubes.
- 2. Pipettes.
- 3. Electric tiles.
- The main stages of the work:

a) pour 4-5 ml of muscle slurry (muscle homogenate) into 2 test tubes. Add 0.5 ml of neutralized succinic acid solution to the first test tube. Add 2 drops of methylene blue solution to both test tubes;
b) mix the contents of both test tubes and place them in a water bath (at 37°C).

After a few minutes, observe the discoloration of methylene blue in the 1st test tube and its absence in the 2nd test tube. After decolorization, shake the 1st test tube strongly, the blue color will appear again due to the oxidation of methylene blue.

Requirements for work results.

Enter the obtained data into the workbook.

Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

1. Exchange of substances (metabolism) - general patterns of catabolic and anabolic processes.

2. Common stages of intracellular catabolism of biomolecules: proteins, carbohydrates, lipids.

3. Cycle of tricarboxylic acids. Localization, sequence of enzymatic reactions, significance in metabolism.

4. Energy balance of the cycle of tricarboxylic acids. Physiological significance of CTC reactions.

5. Amphibolic pathways and anaplerotic reactions. Examples.

Test tasks.

- 1 The concentration of which metabolite affects the work of CTC?
- a) ADP
- b) Glucose
- c) Oxaloacetate
- d) Alanine

- e) Phospholipases
- 2 In the course of CTC reactions, CO2 is formed, the amount of which is:
- a) 2
- b) 1
- c) 5
- d) 3
- e) 4

3 In the reactions of CTC, reduced flavin dehydrogenases are formed, which can lead to the synthesis of:

- a) 2 ATP
- b) 1 ATP
- c) 3 ATP
- d) 5 ATP
- e) 4 GTF

4 In the reactions of CTC, reduced pyrimidine dehydrogenases are formed, which can lead to the synthesis of:

- a) 9 ATP
- b) 7 ATP
- c) 3 ATP
- d) 5 ATP
- e) 2 GTF

5 CTC reactions ensure the synthesis of macroergic compounds (ATP and GTP). At the same time, the number of these molecules, the synthesis of which is not related to the work of the electron transport chain, is:

- a) 1
- b) 7
- c) 3
- d) 2
- e) 5

6 A 10-year-old child has Weber's optic neuropathy, the cause of which is a decrease in the activity of NADH-dehydrogenase of the respiratory chain of mitochondria in the cells of the optic nerve. The development of neuropathy is associated with a violation of which process?

- a) Oxidative phosphorylation
- b) Glycolysis
- c) Cycle of three carboxylic acids
- d) ketogenesis
- e) oxidation of fatty acids

7 The main energy process in the body is the cycle of three carboxylic acids, discovered in the 1930s. Outstanding biochemist, Nobel Prize laureate:

- a) H. Krebsom
- b) D. Sumner
- c) O. Warburg
- d) P. Mitchell
- e) O. Meyerhof

8 During the oxidation of carbohydrates, lipids, and proteins, a large amount of energy is generated, the main part of which is synthesized in the cycle of three carboxylic acids from acetyl-CoA. How many molecules of ATP are formed when one molecule of acetyl-CoA is oxidized?

- a) 12
- b) 24
- c) 38
- d) 1
- e) 3

9 During the oxidation of acetyl-CoA in the cycle of three carboxylic acids, many enzymes, which include non-protein substances, take part. Name the non-protein substance that is necessary for the conversion of succinic acid into fumaric acid:

- a) FAD
- b) OVER
- c) FMN
- d) TPF
- e) Coenzyme Q

10 A decrease in the activity of enzymes of the Krebs cycle, which occurs as a result of the influence of alcohol on the body, can cause the development of hypoxia:

- a) Fabric
- b) Respiratory
- c) Hypoxic
- d) Circulatory
- e) Chemical

11 A low level of which of the metabolites listed below causes inhibition of the Krebs cycle and increased ketogenesis in hepatocytes under the condition of limited utilization of carbohydrates:

- a) Oxaloacetate
- b) Acetyl-CoA
- c) ATP
- d) Fatty acids
- e) ADP
- 12 Anabolism processes are characterized by:
- a) Breakdown of metabolites into simpler compounds
- b) Convergence of metabolic pathways
- c) Formation of ATP molecules
- d) Formation of various organ- and tissue-specific biomolecules
- e) Formation of joint metabolites
- thirteen Uncharacteristic for the second stage of catabolism:
- a) Entry of monomers from the gastrointestinal tract into the blood and tissue cells
- b) Formation of key metabolites
- c) Formation of endogenous water
- d) The release of the energy of chemical bonds in the form of high-energy hydrogen
- e) Entry into cells of monomers that are formed as a result of intracellular catabolism of own carbohydrates, lipids and proteins
  - 14 In the third phase of catabolism:
  - a) Completion of cleavage of key metabolites
  - b) Formation of final products CO2 and H2O
  - c) Energy release
  - d) Inclusion of the Krebs cycle a common cyclic metabolic pathway
  - e) All answers are correct
  - 15 Substrates of biological oxidation can be:
  - a) Starch, glycogen, triacylglycerols, lactose food
  - b) Own proteins of blood and tissues
  - c) Nucleic acids and other high-molecular compounds
  - d) Amino acids, glucose, fatty acids, alcohols, oxyacids, etc.
  - e) Cellulose
  - 16 At all stages of biological oxidation:
  - a) Catabolic transformation of substrates
  - b) The release of the same amount of free energy
  - c) Air oxygen consumption
  - d) Consumption of carbon dioxide

- e) Release of various amounts of free energy
- 17 In the Krebs cycle, oxidation undergoes:
- a) A central key metabolite
- b) Glucose
- c) Pyruvic acid
- d) Glycerol
- e) Amino acid
- 18 The biological functions of CTC are:
- a) Maintenance of the physiological concentration of PVC in the cell
- b) Formation of substrates for gluconeogenesis
- c) Formation of high-energy hydrogen in the form of four portions of reduced cof actors
- d) Formation of endogenous water
- e) Formation of biologically active substances

19 To increase the performance of athlete K.'s training, the doctor recommended taking citric acid or products containing it, because it:

- a) CTC substrate
- b) The source of the starting material for the synthesis of fatty acids
- c) Fatty acid synthesis activator
- d) Activator of gluconeogenesis
- e) Inhibitor of glycolysis

20 A patient with atrophic gastritis has a sharply reduced content of erythrocytes in the blood. The biochemical mechanism of the development of this pathology is a violation of the central nervous system. The concentration of which metabolite affects the work of CTC?

- a) ADP
- b) Glucose
- c) Oxaloacetate
- d) Alanine
- e) Phospholipases

21 A woman with signs of metabolic acidosis was brought to the hospital. In the course of CTC reactions, CO2 is formed, the amount of which is:

- a) 2
- b) 1
- c) 5
- d) 3
- e) 6

22 The biochemical basis of the increase in the content of ketone bodies in pathological conditions is a decrease in the degree of utilization of acetyl-CoA in the central nervous system due to a violation of carbohydrate metabolism. This is due to the outflow from the CTC:

- a) Oxaloacetate
- b) Ketoglutarate
- c) Fumarate
- d) Malatu
- e) Succinate

23 Which carboxylic acid - an intermediate product of CTC - takes part in the regulation of blood calcium transport level?

- a) Citrate
- b) Isocitrate
- c) Oxaloacetate
- d) Succinate
- e)  $\alpha$ -ketoglutarate

24 Which of the components of CTC forms tightly bound complexes with calcium in dentin and takes part in the processes of mineralization and demineralization?

- a) Citrate
- b) Isocitrate
- c) Oxaloacetate
- d) Succinate
- e) Malat

25 The central intermediate product of all exchanges (proteins, lipids, carbohydrates) is:

- a) Acetyl-CoA
- b) Succinyl-CoA
- c) Oxaloacetate
- d) Pyruvate
- e) Citrate

26 A patient suffering from diabetes has ketonuria and ketonemia. Indicate which of the substances listed below is a precursor of ketone bodies?

- a) Acetyl CoA
- b) Oxaloacetate
- c) Unsaturated fatty acids
- d) Alpha-ketoglutarate
- e) Cholesterol

27 In the brain, ammonia, which is formed during the deamination of amino acids and amines, binds to alpha-ketoglutaric and glutamic acids. Therefore, the toxic effect of ammonia on the central nervous system is due to the suppression of:

- a) Cycle of three carboxylic acids
- b) Ornithine cycle of urea formation
- c) Pentose phosphate cycle
- d) Glycolysis
- e) Gluconeogenesis

28 In the human body, CTC reactions provide the synthesis of macroergic compounds (ATP and GTP). At the same time, the number of these molecules, the synthesis of which is connected with the work of the electrolyte transport chain, is:

- a) 1
- b) 3
- c) 5
- d) 2
- e) 4

29 During the oxidation of acetyl-CoA in the CTC, many enzymes, which include non-protein substances, take part. Name the non-protein substance that is necessary for the conversion of succinic acid into fumaric acid.

- a) FAD
- b) OVER
- c) FMN
- d) TPF
- e) Coenzyme Q

30 In the tissues of a healthy person, in the reactions of CTC, reduced pyrimidine dehydrogenases are formed, which can lead to the synthesis of:

- a) 9 ATP
- b) 7 ATP
- c) 3 ATP
- d) 6 ATP
- e) 2 ATP

31 Name the common end product of the second stage of catabolism of carbohydrates, lipids, and proteins:

a) Acetyl CoA

- b) Pyruvate
- c) Citrate
- d) Acyl-CoA
- e) ATP

32 The biochemical basis of the increase in the number of ketone bodies in pathological conditions is a decrease in the degree of utilization of acetyl-CoA in the central nervous system due to a violation of carbohydrate metabolism. This is due to the leakage from the CTC:

- a) Oxaloacetate
- b) Ketoglutarate
- c) Fumarate
- d) Malatu
- e) Succinate

33 During the oxidation of carbohydrates, lipids, and proteins, a large amount of energy is generated, the main part of which is synthesized in the central nervous system from acetyl-CoA. How many molecules of ATP are formed when one molecule of acetyl-CoA is oxidized:

- a) 12
- b) 24
- c) 36
- d) 4
- e) 2
- 34 Anaplerotic reactions are:

a) Reactions that increase the concentration of CTC substrates, forming them from intermediates of other metabolic pathways

b) Reactions that use CTC substrates for the formation of intermediates necessary for biosynthetic processes

- c) Nucleotide decay reactions
- d) Reactions of heme biosynthesis
- e) Reactions of protein biosynthesis.
- 4. Summary:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

#### Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

#### Practical class №4

Topic: <u>Molecular mechanisms of tissue respiration. The structure of</u> <u>mitochondria. Respiratory enzymes. Electron transport chain. Oxidative phosphorylation in</u> <u>the respiratory chain. Macroergic compounds. Peroxide and microsomal oxidation.</u> <u>Antioxidant systems.</u>

**Goal:** learn the basic principles of the mitochondrial respiratory chain, the role of redox enzymes in tissue respiration and the effect on the electron transport chain of biologically active and toxic substances

**Basic concepts:** *biological oxidation, tissue respiration, oxidative phosphorylation, macroergic compounds* 

Equipment: Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

Requirements for theoretical readiness of higher education applicants to perform practical classes:

The higher education applicant should know:

- explain the structure of the respiratory chain and the purpose of its main links (enzymes, coenzymes);

- analyze the structure and biological role of the respiratory chain; to explain the formation mechanism and biological role of redox potential in the respiratory chain;

- to analyze the mechanisms of action of medicines, biologically active and toxic substances in the processes of tissue respiration;

- the structure of the atom;
- active forms of oxygen;
- lipid peroxidation processes;
- "lines of defense" against free radicals;
- characteristics of monooxygenase reactions;
- characteristics of the microsomal oxidation system.

The higher education applicant should be able to:

-reproduce the scheme of transfer of protons and electrons in the respiratory chain;

-indicate the conjugation points of oxidation and phosphorylation;

-write reactions of formation of free radicals;

-sequence of microsomal oxidation enzymes.

Questions to check basic knowledge on the topic of the class:

- 1. Write the formula for NADP+
- 2. Write the formula for ATP.
- 3. Define free radicals.
- 4. Localization and components of the microsomal oxidation chain.
- 5. Write the formula of vitamin PP. Which coenzymes does it include?
- 6. Write the formula for vitamin B2. Which coenzymes does it include?

- 7. Write the formula for OVER.
- 8. Write the formulas of FAD and FMN.
- 9. Which complex proteins include iron?
- 3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Discovery of oxidase (tyrosinase) in potatoes". *Recommendations for performing tasks.* 

The principle of the method consists in catalyzing oxidation of substrates by free oxygen by oxidases. Tyrosinase is the most typical oxidase, found in potatoes and in animal organisms. It catalyzes the oxidation of tyrosine with its transformation into dark substances similar to pigments (melanins). Oxidation occurs in several stages:



In the human body, tyrosinase catalyzes the transformation of adrenaline into the pigment adrenochrome.

Reagents:

1. Tyrosine solution.

2. Potatoes.

Equipment:

1. Pipettes.

The main stages of the work:

Apply a few drops of tyrosine solution to the potato slice and leave it until the end of the class. A red color is observed on the cut of the potato.

Requirements for work results.

Enter the obtained data into the workbook.

Make medical and biological conclusions.

3.2 Demonstration and practical work "Discovery of peroxidase in horseradish extract". *Recommendations for performing tasks.* 

The principle of the method consists in peroxidase catalyzing the oxidation of phenols, polyphenols, aromatic amines due to peroxide oxygen. Peroxidases are found in plants (especially abundant in horseradish) and animal organisms. Hemoglobin, myoglobin, cytochromes have weak peroxidase activity.

Reagents:

- 1. 10% alcohol solution of guava resin.
- 2. 2% hydrogen peroxide.
- 3. Extract from horseradish.
- 4. Distilled water.
- Equipment:
- 1. Test tubes.
- 2. Pipettes.

The main stages of the work:

a) put 5 drops of 10% alcohol solution of guava resin and 5 drops of 2% hydrogen peroxide into two test tubes;

b) add 5 drops of fresh horseradish extract to the first test tube;

c) add 5 drops of water to the second test tube;

d) mix the contents of the test tubes.

A change in color is observed. Pine resin, being oxidized, acquires a blue color.

Requirements for work results.

Enter the obtained data into the workbook.

Make medical and biological conclusions.

3.3 Demonstration and practical work "Discovery of catalase in blood".

Recommendations for performing tasks.

The principle of the method consists in catalase of blood catalyzing the decomposition of H2O2 into O2 and H2O:

# $H_2O_2 \longrightarrow O_2 + H_2O_2$

The biological role of catalase is to neutralize hydrogen peroxide.

Reagents:

1. Citrate blood.

2. 2% hydrogen peroxide solution.

Equipment:

1. Test tubes.

2. Pipettes.

The main stages of the work:

Pour 10-15 drops of 2% hydrogen peroxide and one drop of blood into the test tube.

Foaming of the liquid due to the violent release of oxygen bubbles is observed.

Requirements for work results.

Enter the obtained data into the workbook.

Make medical and biological conclusions.

3.4 Demonstration and practical work "Quantification of blood catalase (catalase number) according to Bach and Zubkova.

Recommendations for performing tasks.

The catalase number is the number of milligrams of H2O2 that is decomposed by one microliter of the examined blood ( $1\mu$ l=1mm3).

The principle of determining the catalase number is based on the following reaction:

 $2KMnO4 + 5 H2O2 + 4 H2SO4 \rightarrow 2KH SO4 + 8 H2O + 5 O2 + 2MnSO4$ ,

that is, the amount of decomposed peroxide is judged by the difference in the amount of potassium permanganate spent on titration before and after the action of catalase.

The main stages of the work:

Preparation of the basic blood solution (1:1000). Pour about 10 ml of distilled water into a 100 ml measuring flask. Add 0.1 ml of the tested blood to the flask with a micro pipette, having previously wiped the tip of the capillary from the blood that has stuck to the outside. Rinse the pipette with liquid from the volumetric flask. Add distilled water to the volumetric flask up to the mark. Note the time when the blood was diluted.

Preparation of enzyme-substrate mixture. Pour 7-8 ml of distilled water into two conical flasks. Add 1 ml of the basic blood solution to both flasks. Boil for 2 minutes. the contents of the first flask for the destruction of catalase. Let both flasks stand at room temperature for 30 minutes, counting from the moment of blood dilution.

Incubation at room temperature. Add exactly 2 ml of H2O2 solution to each flask and leave again for 30 minutes.

The interaction of the remaining peroxide with sulfuric acid. Add 4-5 ml of H2SO4 solution to each flask.

Determination of the amount of H2SO4 that remained in the flask. Titrate the contents of each flask with 0.1N KMnO4 solution until a pink color appears.

Catalase decomposes part of H2O2. Therefore, more KMnO4 solution will be used for the titration of the contents of the second flask than for the titration of the contents of the first flask, where the catalase was destroyed by boiling. This difference is multiplied by 1.7 and the catalase number of the examined blood is obtained.

Medical and biological evaluation of the obtained results. Normally, the catalase number is 12-20. The content of catalase in the blood decreases in some diseases (cancer, anemia, tuberculosis).

Requirements for work results.

Enter the obtained data into the workbook.

Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

1. Definition of the concept of tissue respiration.

2. Action of nicotinamide enzymes. Write the chemistry of the process of converting the oxidized form of NAD into the reduced form.

3. Action of flavin enzymes. Write the chemistry of the process of converting the oxidized form of FAD into the reduced form.

4. The action of coenzyme A. Write the chemistry of the process of converting the oxidized form of ubiquinone into the reduced form.

5. Action of cytochromes. Schematically depict the process of electron transfer through the cytochrome system to oxygen.

6. Electron transfer from cytochrome oxidase to oxygen.

7. The interaction of highly reactive oxygen  $O_2$ -with 2H+ protons.

8. Values of redox potentials. Their value in determining the sequence of enzymes of the respiratory chain.

9. Draw a diagram of the main, longer and shorter respiratory chains.

Test tasks.

1 A 10-year-old child has Weber's optic neuropathy, the cause of which is a decrease in the activity of NADH-dehydrogenase of the respiratory chain of mitochondria in the cells of the optic nerve. The development of neuropathy is associated with a violation of which process?

- a) Oxidative phosphorylation
- b) Glycolysis
- c) Cycle of three carboxylic acids
- d) ketogenesis
- e) oxidation of fatty acids
- 2 In all living nature, including the human body, the main macroergic compound is:
- a) ATP
- b) Creatine phosphate
- c) Phosphoenolpyruvate
- d) Acetyl-CoA
- e) Diphosphoglycerate

3 The patient underwent a long course of treatment with the antibiotic oligomycin. What kind of metabolic disorder can this cause?

- a) Inhibition of ATP synthesis
- b) Increase in ATP synthesis
- c) Inhibition of tissue respiration

- d) Inhibition of NAD-dependent dehydrogenase
- e) Inhibition of cytochromes

4 A patient in a state of acute hypoxia after hydrogen sulfide poisoning was taken to the hospital. What is one of the possible mechanisms of action of this gas on the body?

- a) Inhibits tissue respiration in mitochondria
- b) Inhibits dehydrogenation reactions
- c) Inhibits CTC
- d) It separates tissue respiration and phosphorylation
- e) Inhibits glycolysis
- 5 The organelles that belong to the "energy stations" of the cell are:
- a) Mitochondria
- b) Lysosomes
- c) Plasma membrane
- d) Golgi apparatus
- e) Ribosomes
- 6 In the bilipid layer of membranes, the protein molecule is fixed with the help of bonds:
- a) Electrostatic and hydrophobic
- b) Peptide and their disulfide
- c) Hydrophobic and ionic
- d) Ionic and hydrogen
- e) Hydrogen and disulfide
- 7 In the human body, compounds that contain macroergic connections are represented:
- a) Succinate
- b) 3-phosphoglycerate
- c) 2-phosphoglycerol
- d) pyruvate
- e) ATP, GTF, UTF
- 8 Substrates of biological oxidation can be:
- a) Starch, glycogen, triacylglycerols, lactose food
- b) Own proteins of blood and tissues
- c) Nucleic acids and other high-molecular compounds
- d) Amino acids, glucose, fatty acids, alcohols, oxyacids, etc.
- e) Cellulose
- 9 At all stages of biological oxidation:
- a) Catabolic transformation of substrates
- b) The release of the same amount of free energy
- c) Air oxygen consumption
- d) Consumption of carbon dioxide
- e) Release of various amounts of free energy
- 10 In the process of tissue respiration, the following occurs:
- a) Oxidation of reduced cofactors
- b) Transport of hydrogen (H+ and e–) from oxidizing substrates to air O2
- c) Complete oxygen recovery
- d) Formation of the final product of biological oxidation endogenous water
- e) All answers are correct
- 11 Tissue respiration is associated with processes:
- a) Oxidative decarboxylation
- b) Substrate phosphorylation
- c) Transdeamination
- d) Oxidative phosphorylation
- e) glycolysis
- 12 The number of areas of conjugation in tissue respiration depends on:

a) From the oxidizing substrate

b) From the amount of energy released at each transfer of an electron along the electron transport chain

- c) From the supply of tissue with oxygen
- d) From the supply of tissue with carbon dioxide
- e) From the total amount of energy released

thirteen Separating substances:

- a) Do not affect the transfer of electrons in mitochondria
- b) Contribute to the formation of heat
- c) Suppress the synthesis of ATP from ADP and inorganic phosphate

d) Prevent the emergence of a gradient of hydrogen ion concentration between the two sides of the mitochondrial membrane

- e) All answers are correct
- 14 The intensity of tissue respiration in mitochondria depends on:
- a) Exclusively from the amount of oxidation substrate
- b) From the concentration of phosphoric acid
- c) From the concentration of H+
- d) From the concentration of OH-
- e) From the relationship
- 15 The most intense redox processes are:
- a) In the cytoplasm
- b) In mitochondria
- c) In lysosomes
- d) In ribosomes
- e) In EPR

16 Manifestations of polyneuritis with hypovitaminosis B1 are mainly the result of impaired energy supply to the brain due to:

- a) Reduction of oxidative decarboxylation of keto acids
- b) Decreased substrate phosphorylation
- c) Deficiency of ATP
- d) Low creatine kinase activity
- e) High activity of adenylate cyclase

17 Acrychin is prescribed for enterobiosis - a structural analog of vitamin B2. Violation of the synthesis of which enzymes takes place under the action of this drug?

- a) FAD-dependent dehydrogenases
- b) Cytochrome oxidases
- c) Peptidase
- d) NAD-dependent dehydrogenases
- e) Aminotransferase
- 4. Summary:

5. List of recommended literature (main, additional, electronic information resources):

#### Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

#### Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

#### Practical class №5

Topic: Intracellular catabolism of glucose. Glycolysis: reactions. Comparison of glycolysis and alcoholic fermentation.Glycogenolysis: regulation. Differences from glycolysis. Stages of aerobic oxidation of glucose: oxidative decarboxylation of pyruvate. Bioenergetics of the process.

**Goal:**ATintroducing higher education applicants to the biological role and main pathways of carbohydrate conversion in the human body, glucose oxidation by anaerobic glycolysis, the differences between glycolysis and alcoholic fermentation, glucose oxidation reactions in aerobic conditions, the structure of the pyruvate dehydrogenase complex, as well as the structure and biological role of glycogen, the ways of its cleavage in various organs and tissues.

**Basic concepts:**carbohydrates, glycolysis, glycolytic oxidation reduction, alcoholic fermentation, glycogenolysis, oxidative decarboxylation of pyruvate

**Equipment:**laboratory of the department.

#### Plan:

**1. Organizational measures:**greetings, verification of those present, announcement of the topic, purpose of the class, motivation

### 2. Control of the reference level of knowledge

# **2.1. Requirements for theoretical readiness of higher education applicants:**

1. What compounds are carbohydrates?

- 2. Which formulas can be used to represent the structure of glucose and fructose
- 3. Classification of carbohydrates.
- 4. Types of isomerism characteristic of representatives of the class of carbohydrates.
- 5. What is an asymmetric carbon atom, how is it designated? Optical isomerism.

### 2.2. Questions to check basic knowledge on the topic of the class:

1. Write the sequence of glycolysis reactions.

- 2. How does the process of alcoholic fermentation differ from glycolysis.
- 3. Write the sequence of fermentation reactions.
- 4. Name the end products of anaerobic glycolysis.
- 5. What is glycolytic redox.
- 6. What enzymes and coenzymes are part of the pyruvate dehydrogenase complex
- 7. Sequence of reactions of complex oxidative decarboxylation of pyruvate
- 8. Regulation of the activity of the pyruvate dehydrogenase complex
- 9. Distinguish between glycolysis and glycogenolysis
- 10. Peculiarities of glycogenolysis in the liver and skeletal muscles
- 11. Regulation of glycogenolysis.

12. Calculate the energy balance of aerobic glycolysis and glycogenolysis

**3. Formation of professional skills and abilities:**demonstration and practical work.

### **3.1.** Content of demonstration and practical work

**3.1.1.**Reproduction of "in vitro" process of glycolysis, study of final products.

### **3.2. Implementation recommendations:**

**3.2.1.**1. Preparation of the reaction mixture:

Animal muscles (1 g) are ground in a mortar with glass sand, adding 2-3 ml of phosphate buffer with glucose content. Pour 3 ml of the same buffer into two test tubes. 1 ml of 20% THO solution is added to the first test tube (control) to precipitate proteins and end the enzymatic processes. Add 0.5 ml of muscle slurry to both test tubes and close them with stoppers.

2. Thermostat:

Both test tubes are placed in a thermostat for 45 to 30 minutes<sup>at</sup>C. Before the end of incubation, prepare two test tubes.

3. Carrying out the Uffelman reaction:

10 drops of 1% solution of Ufelman's reagent are poured into the prepared test tubes. The contents of the incubation tubes are filtered through a cotton filter, after which 0.5 ml of the filtrate is added dropwise to Ufelman's reagent. A change in coloration from purple to yellow-green in the test sample is observed.

**3.1.2.**Sample for alcoholic fermentation.

**3.2.2.**1. Preparation of the reaction mixture:

1 gbaker's yeast is ground in a mortar while constantly adding a 5% glucose solution (about 30 ml). The resulting liquid is introduced into the fermentation tube in such a way that the upper knee is completely filled, and a little liquid remains in the area of expansion.

2. Thermostating the sample:

The fermentation tube is placed in the thermostat for 40 minutes.

3. Observation of fermentation:

Observe CO release<sub>2</sub>it is necessary to add 2-3 ml of 10% solution of NaOH to the tube and tightly close the opening of the tube with the pad of the thumb, after which the tube is turned over several times, stirring its content CO2 is absorbed by alkali and the pad of the finger is sucked into the opening of the tube.

4. Detection of ethanol:

To detect ethanol, a part of the contents of the tube is taken (3-5 ml), filtered through a cotton filter, the pH is checked (if the filtrate is acidic, it is alkalized with NaOH to a weak alkaline reaction), a few drops of iodine solution are added and heated slightly. A characteristic smell of iodoform is formed.

**3.1.3.** Determination of pyruvate content in blood serum.

**3.2.3.**<u>Principle of the method:</u>

After precipitation of serum proteins, pyruvic acid without proteinof the filtrate reacts in an acidic environment with 2,4-DFH, forming the corresponding hydrazone. After the addition of alkali to the incubation medium, a characteristic color develops, the intensity of which is proportional to the concentration of pyruvate, which is estimated from a standard solution containing 5.0  $\mu$ g of pyruvate in 1 ml. The method is not specific enough, since hydrozones are also formed by other keto compounds, but since the increase in the inhibition level of hydrazones can occur mainly due to pyruvate, it can be used for diagnostic purposes.

Progress of work. 1. Obtaining the researched material:

0.7 ml of H is added to 0.3 ml of blood serum<sub>2</sub>Oh, mix, add 1 ml of 10% THO. Stir and after 2-3 minutes centrifuge at 1500 rpm for 15 minutes. Take two test tubes, mark the experiment on one, and the control on the second.

<u>Experiment</u>	<u>CONTROL</u>			
1 ml of centrifuge	1 ml of pyruvate No			
0.5 ml of 2,4-DFH solution	0.5 ml of 2,4-DFH solution			
leave for 20 minutes at room temperature in a dark place				

After 10 minutes, colorimeter against water. Measurements are carried out with experimental and control samples. The obtained measurements are entered in the experiment protocol and the calculation is carried out according to the formula:

#### $Xh = \underline{Sst \ x \ Est}$

E samples

#### CONCLUSION AND MEDICO-BIOLOGICAL ASSESSMENT OF RESEARCH RESULTS Summary table of data on lactate and pyruvate

Metaboli	Lactate	mmol/l	Pyruvate mmol/l		
c					Arterial-venous
State	artery	vei	artery	vein	difference
		n			
NORM	0.55	0.	50.0	70.0	Lactate-0.3 mmol/l;
		80			Pyruvate-20.0
					mmol/l
HYPOXI	0.70	1.	150.0	120.	Lactate-0.50
A		20		0	mmol/l;
Lung					Pyruvate-30 mmol/l
average	2.15	1.	200.0	160.	Lactate-0.65
		70		0	mmol/l
					Pyruvate-0.40
					mmol/l
Heavy	3.60	2.	240.0 or	200.	Lactate-1.10
	or more	50 and	more	0 or more	mmol/l
	(death-	more			Pyruvate-0.40
	case)				mmol/l

**3.3. Requirements for work results:** in a notebook for demonstration and practical work, write down the method of work performance, the results of observation. Provide a medico-biological evaluation of the obtained results. Draw conclusions.

**3.4.** Control materials for the final stage of the class:test tasks for the topic:

1. The erythrocyte needs energy in the form of ATP for its vital activity. What process provides this cell with the required amount of ATP?

- A Anaerobic glycolysis
- B Aerobic oxidation of glucose
- C The pentose cycle
- D Beta-oxidation of fatty acids
- E Cycle of tricarboxylic acids

2. Anaerobic breakdown of glucose to lactic acid is regulated by appropriate enzymes. Specify which enzyme is the main regulator of this process?

- A Enolase
- B Glucosel-6-phosphate isomerase
- C Aldolase
- D Phosphofructokinase
- E Lactate dehydrogenase

3. The concentration of glucose in the blood plasma of a healthy person is within the following limits:

- A 3.5-5.5 mmol/l
- B 2-4 mmol/l
- C 10-25 mmol/l
- D 6-9.5 mmol/l

#### E 1-2 mmol/l

- 4. What substance is the main source of energy for brain tissue?
- A Glycerin
- B Fatty acids
- C Glucose
- D Amino acids
- E Lactic acid

5. During short-distance running, untrained people experience muscle wasting due to the accumulation of lactate. Indicate the strengthening of which biochemical process this may be associated with.

- A Glycogenesis.
- B Gluconeogenesis.
- C pentose phosphate pathway.
- D Lipogenesis.
- E Glycolysis.

6. A 7-year-old girl has obvious signs of anemia. Laboratory established deficiency of pyruvate kinase in erythrocytes. Disruption of which process plays the main role in the development of anemia in a girl?

- A Anaerobic glycolysis
- B Oxidative phosphorylation
- C Tissue respiration
- D Decomposition of peroxides
- E Deamination of amino acids

7. A large number of metabolites of glucose oxidation are dissolved in the cytoplasm of myocytes. Name one of them, directly converted to lactate.

- A Oxaloacetate
- B Pyruvate
- C Glycerophosphate.
- D Glucose-6-phosphate.
- E Fructose-6-phosphate.

8. During long-distance running, the skeletal muscles of a trained person use glucose to obtain ATP energy for muscle contraction. Indicate the main process of utilization of glucose under these conditions.

- A Anaerobic chandnumberandwith.
- B Aerobic chandnumberandwith.
- C Seeandcogenolandwith.
- D Gluconeogenesis.
- E Glycogenesis.

9. Carbohydrates, especially sucrose, play a significant role in the spread of dental caries, especially in childhood. Which of the carbohydrate conversion pathways is accompanied by the formation of acids that leads to dentin demineralization?

- A. Gluconeogenesis
- B. The pentose cycle
- C. Glycolysis
- D. Synthesis of fatty acids
- E. Cholesterol synthesis

10. After long-term physical exertion during the physical education class, the students developed muscle weakness. The cause of its occurrence was the accumulation of lactic acid in the skeletal muscles. It was formed after activation in the body of students:

- A Lipolysis.
- B Gluconeogenesis.
- C Pentose phosphate cycle.
- D Glycolysis.
- E Glycogenesis.

11. The patient was taken to a medical institution in a comatose state. From the words of the attendants, it was found that the patient fainted during training at the final stage of the marathon distance. Who is most likely to be suspected of

- A. Hyperglycemic.
- B. Hypoglycemic.
- C. Acidotic.
- D. Hypothyroidism.
- E. Pechinkova.

12. In the process of glycolysis, hexose D-fructose-1,6-diphosphate is synthesized, from which two trioses are subsequently formed: dioxyacetone phosphate and glyceraldehyde-3-phosphate. The enzyme that catalyzes this reaction is:

- A. Fructose-1,6-diphosphate aldolase
- B. Phosphohexose isomerase
- S. Triosephosphatisomerase
- D. Phosphofructose isomerase
- E. Enolase

13. Patient A. was in an unconscious state after a brain injury. During the examination, an increase in the content of lactic acid and pyruvic acid in the blood serum was established, the pH of the blood was 7.2. These indicators indicate a violation of which metabolic process?

A Strengthening of aerobic glycolysis

In Inhibition of gluconeogenesis

C Enhancement of gluconeogenesis

D Enhancement of anaerobic oxidation of glucose

E Enhancement of glycogenolysis

14. A 42-year-old man ate a large portion of spaghetti, a piece of cake, and drank a glass of sweet tea. The activity of which enzyme of hepatocytes is activated to the greatest extent after consumption of high-carbohydrate food?

A. Hexokinase

- B. Glucose-6-phosphatase
- S. Glucose-6-phosphate dehydrogenase
- D. Glycogen phosphorylase
- E. Beta-galactosidase

15. The child is frail, apathetic. The liver is enlarged and a liver biopsy revealed a significant excess of glycogen. The concentration of glucose in the blood is below normal. What is the reason for the low concentration of glucose in the blood of this patient?

A Decreased (absent) activity of glycogen phosphorylase in the liver.

- B Reduced (absent) hexokinase activity.
- C Increased activity of glycogen synthetase.

D Decreased (absent) activity of glucose-6-phosphatase.

E Deficiency of the gene responsible for the synthesis of glucose-1-phosphaturidine transferase.

16. After blood circulation is restored in the damaged tissue, the accumulation of lactate stops and the rate of glucose consumption decreases. The activation of which process is caused by these metabolic shifts?

- A Anaerobic glycolysis
- B Aerobic glycolysis
- C Lipolysis
- D Gluconeogenesis
- E Glycogen biosynthesis

17. A 34-year-old patient has reduced endurance for physical exertion, while the glycogen content in skeletal muscles is increased. A decrease in the activity of which enzyme is this can be explained

A. Glycogen synthases

- B. Glucose-6-phosphate dehydrogenases
- C. Phosphofructokinase
- D. Glycogen phosphorylases
- E. Glucose-6-phosphatases

18. A 10-year-old child constantly complains of pain and muscle cramps after physical exertion. When examining the blood, the content of glucose, lactate and creatine corresponds to the physiological norm. Myoglobin is determined in the urine. A deficiency of glycogen phosphorylase, an enzyme that catalyzes the conversion of:

A. Glycogen into glucose-1-phosphate

- B. Glucose-6-phosphate to glucose
- C. Glucose-6-phosphate in glycogenase
- D. Glucose into glucose-6-phosphate
- E. "Seeds" of glycogen into glycogen

19. Patients with alcoholism often have signs of hypovitaminosis B1: polyneuritis, memory impairment. The metabolism of which substances is changed in this case?

A Violation of anaerobic oxidation of glucose

In Violations of aerobic oxidation of glucose

C Violation of amino acid oxidation

D Violation of beta - oxidation of fatty acids

E Violation of lipolysis in adipose tissue

20. A 2-year-old child is diagnosed with Gierke's disease, which is manifested by severe hypoglycemia. The reason for this condition is the absence of the glucose-6-phosphatase enzyme. What process is this pathology associated with?

A Glycogen synthesis In Mobilization of glycogen With Ketogenesis D Glycolysis E Gluconeogenesis

21. In the process of long-distance running, the skeletal muscles of a trained person use glucose to obtain ATP energy for muscle contraction. Indicate the main process of utilization of glucose under these conditions.

A Aerobic chandnumberandwith.

B Anaerobic chandnumberandwith.

C Seeandcogenolandwith.

D Gluconeogenesis.

E Glycogenesis.

### 4. Summary of the class. Assessment.

# 5. List of recommended literature

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

1. https://info.odmu.edu.ua/chair/biology/-

- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

### Practical class №6

Topic: Alternative ways of monosaccharide metabolism. The pentose phosphate pathway of glucose oxidation: scheme, biological significance, features of functioning in various tissues. Metabolic pathways of fructose and galactose conversion: hereditary enzymopathies of their metabolism.

**Goal:**Formation of systemic knowledge on the pathways of monosaccharide metabolism. Study of molecular mechanisms of the pentose phosphate pathway, transformations of fructose and galactose.

**Basic concepts:**pentose phosphate pathway of oxidation of glucose, fructose, galactose, hereditary metabolic enzymopathies.

**Equipment:**laboratory of the department.

Plan:

**1. Organizational measures:** greetings, verification of those present, announcement of the topic, purpose of the class, motivation

### 2. Control of the reference level of knowledge

### 2.1. Requirements for higher education applicants' theoretical readiness:

1. Formulas of the main representatives of monosaccharides

- 2. General ideas about the mechanism of the entry of carbohydrates into cells
- 3. The concept of hyper- and hypoglycemia

4. The amount of carbohydrates contained in the blood of a healthy person

5. Write the reactions of glycolysis.

6. Calculate the energy balance of aerobic and anaerobic glycolysis

2.2. Questions to check basic knowledge on the topic of the class:

1. What enzymes and coenzymes are involved in the pentose phosphate pathway of carbohydrate metabolism?

2. What is the biological significance of PFSH?

3. What mechanisms of PFSh regulation do you know?

4. Clinical aspects of the pentose phosphate pathway

5. Ways of converting fructose in the liver and adipose tissue.

6. Ways of conversion of galactose in the liver

7. Hereditary enzymopathies of fructose and galactose metabolism: fructosemia, hereditary fructose intolerance, galactosemia

3. Formation of professional skills and abilities: demonstration and practical work.

#### 3.1. Content of demonstration and practical work

Detection of fructose by the Selivanov reaction

#### **3.2. Implementation recommendations:**

Principle of the method:

Fructose and other ketohexoses, both in the free state and when separated from more complex substances (for example, sucrose), have a cherry-red color when heated with hydrochloric acid and resorcinol. The color of the sample depends on the reaction of resorcinol with oxymethylfurfural, which appears when ketosis is heated with acid.

Aldoses are also able to form oxymethylfurfural when heated with acids, but this reaction with aldoses occurs much more slowly, which leads to sufficient specificity of the Selivanov reaction

The main stages of the work:

Add 1-2 ml to the test tube. Selivanov's reagent, 1-2 drops of fructose solution and heat to boiling. A red color is observed.

**3.3. Requirements for work results:** in a notebook for demonstration and practical work, write down the method of work performance, the results of observation. Provide a medico-biological evaluation of the obtained results. Draw conclusions.

**3.4.** Control materials for the final stage of the class:test tasks for the topic:

1. The biosynthesis of the purine ring takes place on ribose-5-phosphate by the gradual increase of nitrogen and carbon atoms and the closing of the rings. The source of ribose phosphate is the process:

- A glycolysis
- B pentose phosphate cycle
- C glyconeogenesis
- D gluconeogenesis
- E glycogenolysis

2. During the transformation of glucose in the pentose cycle, various phosphates are formed monosaccharides. Which of these substances can be used for the synthesis of nucleic acids?

- A Ribose-5-phosphate
- B Ribulose-5-phosphate
- C Erythrose-4-phosphate
- D Sedoheptulose-7-phosphate
- E Xylulose-5-phosphate

3. In a 2-year-old boy, there is an increase in the size of the liver and spleen, cataracts. The concentration of sugar in the blood is increased, but the glucose tolerance test is normal. Indicate the hereditary disorder of the metabolism of which substance is the cause of this condition?

A Glucose

- B Fructose
- C Galactose
- D Maltose
- E Sucrose

4. A high content of galactose was found in the child's blood, and the concentration of glucose was reduced. Cataracts, mental retardation, and fatty degeneration of the liver develop. What disease is there?

- A Galactosemia
- B Diabetes
- C Lactosemia
- D Steroid diabetes
- E Fructosemia

5. In the sick child, a delay in mental development, an enlarged liver, and deterioration of vision were detected. The doctor associates these symptoms with a deficiency of galactose-1-phosphaturidyltransferase in the child's body. What pathological process is observed?

A Galactosemia;

- B Fructosemia;
- C Hyperglycemia;
- D Hypoglycemia;
- E Hyperlactatacidemia.

6. An infant was brought to the intensive care unit with the following symptoms: vomiting, diarrhea with impaired growth and development, cataracts, mental retardation. A diagnosis of galactosemia was established. Deficiency of which enzyme occurs?

A. Galactose-1-phosphaturidyltransferases

- B. Glucokinases
- C. UDF of glucose-4-epimerase
- D. UDF of glucose-pyrophosphorylase
- E. Glucose-6-phosphate dehydrogenase

7. The child was diagnosed with galactosemia. The concentration of glucose in the blood does not change significantly. This disease is caused by a deficiency of which enzyme?

- A. Hexokinase
- B. Amylo-1,6-glucosidases
- C. Phosphoglucomutases
- D. Galactokinase
- E. Galactose-1-phosphate-uridyltransferases

8. A sick child with galactose-1-phosphaturidyltransferase deficiency was found to have cataracts, mental retardation, and an enlarged liver. What is the cause of this pathological condition?

- A. Galactosemia
- B. Fructosemia
- C. Hyperglycemia D. Hypoglycemia
- E. Hyperlactatacidemia
- 2. Hypothecial of a contraction of the contraction

9. The pentose phosphate pathway of glucose oxidation has important physiological significance for the functioning of anabolic mechanisms. This is because it is a provider of:

A Ribose-5-phosphate

In Phosphoenolpyruvate

C Glycerol-3-phosphate

D Fructose-1-phosphate

E Galactose-1-phosphate

#### 4. Summary of the class. Assessment.

# 5. List of recommended literature

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

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- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

### Practical class №7

Topic: Glucose biosynthesis: physiological significance, reactions, regulatory enzymes. Substrates of gluconeogenesis. Glucose-lactate and glucose-alanine cycles. Regulation of glucose metabolism. Glucosemia: normal state and its disorders. Glycogen biosynthesis. Regulation of carbohydrate metabolism. Glycogenoses.

**Goal:**Formation of systemic knowledge on ways of glucose metabolism in the human body. Study of molecular mechanisms of glycogenesis and gluconeogenesis and their regulation

**Basic concepts:**Glycogenesis, gluconeogenesis, glucose-lactate, glucose-alanine cycles, glycogenoses.

**Equipment:**laboratory of the department.

Plan:

**1. Organizational measures:**greetings, verification of those present, announcement of the topic, purpose of the class, motivation

### 2. Control of the reference level of knowledge

2.1. Requirements for higher education applicants' theoretical readiness:

1. General ideas about the mechanism of glucose delivery to cells

2. Glycolysis: reactions, enzymes. Irreversible reactions of glycolysis

3. How dangerous is the accumulation of lactate? The mechanism of reutilization of lactate, which is formed in the process of anaerobic glycolysis. Corey cycle.

4. The concept of normo-, hyper- and hypoglycemia

5. Glycogen, structure and functions
6. Glycogenolysis. Regulation of the process of glycogen splitting in the human body.

#### 2.2. Questions to check basic knowledge on the topic of the class:

1. What enzymes and coenzymes are involved in gluconeogenesis?

2. What is the biological significance of gluconeogenesis?

3. What mechanisms of gluconeogenesis regulation do you know?

4. Why is blood glucose determined?

5. Glycogenesis. Reactions, enzymes, process regulation.

6. Glycogenoses. Types. Causes of occurrence.

7. Diabetes.

**3. Formation of professional skills and abilities:**demonstration and practical work.

#### 3.1. Content of demonstration and practical work

**3.1.1.** Detection of glucose content in the blood by the Hagedorn-Jensen method.

#### **3.2. Implementation recommendations:**

#### **3.2.1.** Principle of the method:

With the help of the Hagedorn-Jensen method, not only glucose is determined in the blood, but also some other reducing substances (for example, uric acid, glutathione, creatine). At the same time, the total regenerative capacity in the blood is taken for "sugar". The method is based on the ability of glucose in a protein-free blood filtrate in an alkaline medium when heated to reduce red blood salt ( $K_4$ Fe(CN)6);

Due to the reversibility of this reaction,  $K_4Fe(CN)6$  under the action of zinc sulfate (ZnSO4) is converted into an insoluble salt K2Zn3(Fe(CN)6)2, which is taken in excess and its residue unused in the reaction is determined iodometrically in an acidic medium (for example, in the presence of acetic acid), titrating the amount of iodine formed with sodium thiosulfate.

Starch is used as an indicator for molecular iodine. Glucose content is calculated according to a special table. The table is compiled in such a way that it shows the amount of sodium thiosulfate used for the titration of iodine, and therefore the excess ( $K_4Fe(CN)6$ ), corresponds to the number of milligrams of glucose that reacted in the reaction.

The main stages of the work:

1. 1 ml of 0.1N NaOH is placed in two test tubes. 0.1 ml of blood is added to one of them (sample), and 0.1 ml of distilled water is added to the other (control).

Then add 5 ml of 0.45% ZnSO4, place in a boiling bath for 2-3 minutes, then filter through a cotton swab inserted into the funnel.

2. Add 2 ml of 0.005 N alkaline solution of K3Fe(CH)6 to the filtrate, boil in a water bath for 15 minutes.

3. The mixture is cooled and 3 ml of solution (ZnSO4 + KJ + NaCI) is added, and then 2 ml of CH3COOH and 2 drops of starch (1% p-p) are added

4. The released iodine is titrated with a 0.005N solution of sodium thiosulfate until the blue color disappears.

Н										
ypo-	0	0	0	0	0	0	0	0	0	0
S	.00	.01	.02	.03	.04	.05	.06	.07	.08	.09
ul-										
f										
it										
i										
n										
junior										
0	0	0	0	0	0	0	0	0	0	0
.0	.358	.382	.379	.376	.373	.370	.367	.364	.361	.358
0	0	0	0	0	0	0	0	0	0	0
.1	.355	.352	.350	.348	.345	.343	.341	.338	.336	.333

5. Calculation of results is carried out using a table

0	0	0	0	0	0	0	0	0	0	0
.2	.331	.329	.327	.325	.323	.321	.318	.316	.314	.312
0	0	0	0	0	0	0	0	0	0	0
.3	.310	.308	.306	.304	.302	.300	.298	.296	.294	.292
0	0	0	0	0	0	0	0	0	0	0
.4	.290	.288	.286	.284	.282	.280	.278	.276	.274	.272
0	0	0	0	0	0	0	0	0	0	0
.5	.270	.268	.266	.264	.262	.260	.259	.257	.255	.253
0	0	0	0	0	0	0	0	0	0	0
.6	.251	.249	.247	.245	.243	.241	.240	.238	.236	.234
0	0	0	0	0	0	0	0	0	0	0
.7	.232	.230	.228	.226	.224	.222	.221	.219	.217	.215
0	0	0	0	0	0	0	0	0	0	0
.8	.213	.211	.209	.208	.206	.204	.202	.200	.199	.197
0	0	0	0	0	0	0	0	0	0	0
.9	.195	.193	.191	.190	.188	.186	.184	.182	.181	.179
1	0	0	0	0	0	0	0	0	0	0
.0	.177	.175	.173	.172	.170	.168	.166	.164	.163	.161
1	0	0	0	0	0	0	0	0	0	Δ
	v	0	U	0	U	0	0	0	0	0
.1	.159	.157	.155	.154	.152	.150	.148	.146	.145	.143
.1	.159 <sup>°</sup> 0	.157	.155	.154	.152	.150	.148	.146	.145	.143 0
.1 1 ,2	.159 0 .141	.157 0 .139	.155 0 .138	.154 0 .136	.152 0 .134	.150 0 .132	.148 0 .131	.146 0 .129	.145 0 .127	.143 0 .125
.1 1 ,2 1	.159 0 .141 0	.157 0 .139 0	.155 0 .138 0	.154 0 .136 0	.152 0 .134 0	.150 .132 0	.148 0 .131 0	.146 0 .129 0	.145 0 .127 0	.143 0 .125 0
.1 1 ,2 1 .3	.159 0 .141 0 .124	.157 0 .139 0 .122	.155 0 .138 0 .120	.154 0 .136 0 .119	.152 0 .134 0 .117	.150 0 .132 0 .115	.148 0 .131 0 .113	.146 0 .129 0 .111	.145 0 .127 0 .110	.143 0 .125 0 .108
.1 ,2 1 .3 1	.159 0 .141 0 .124 0	.157 0 .139 0 .122 0	.155 0 .138 0 .120 0	.154 0 .136 0 .119 0	.152 0 .134 0 .117 0	.150 0 .132 0 .115 0	.148 0 .131 0 .113 0	.146 0 .129 0 .111 0	.145 0 .127 0 .110 0	.143 0 .125 0 .108 0
.1 ,2 1 .3 1 .4	.159 0 .141 0 .124 0 .106	.157 0 .139 0 .122 0 .104	.155 0 .138 0 .120 0 .102	.154 0 .136 0 .119 0 .101	.152 0 .134 0 .117 0 .099	.150 0 .132 0 .115 0 .097	.148 0 .131 0 .113 0 .095	.146 0 .129 0 .111 0 .093	.145 0 .127 0 .110 0 .092	.143 0 .125 0 .108 0 .090
$ \begin{array}{r}     .1 \\     .2 \\     .3 \\     .4 \\     .4   \end{array} $	.159 0 .141 0 .124 0 .106 0	.157 0 .139 0 .122 0 .104 0	.155 0 .138 0 .120 0 .102 0	.154 0 .136 0 .119 0 .101 0	.152 0 .134 0 .117 0 .099 0	.150 0 .132 0 .115 0 .097 0	.148 0 .131 0 .113 0 .095 0	.146 0 .129 0 .111 0 .093 0	.145 0 .127 0 .110 0 .092 0	.143 0 .125 0 .108 0 .090 0
$ \begin{array}{r}     .1 \\     .2 \\     .3 \\     .4 \\     .5 \\   \end{array} $	.159 0 .141 0 .124 0 .106 0 .088	.157 0 .139 0 .122 0 .104 0 .086	.155 0 .138 0 .120 0 .102 0 .084	.154 0 .136 0 .119 0 .101 0 .083	.152 0 .134 0 .117 0 .099 0 .081	.150 0 .132 0 .115 0 .097 0 .079	.148 0 .131 0 .113 0 .095 0 .077	.146 0 .129 0 .111 0 .093 0 .075	.145 0 .127 0 .110 0 .092 0 .074	.143 0 .125 0 .108 0 .090 0 .072
$ \begin{array}{r} .1 \\ 1 \\ .2 \\ 1 \\ .3 \\ 1 \\ .4 \\ 1 \\ .5 \\ 1 \\ \end{array} $	.159 0 .141 0 .124 0 .106 0 .088 0	.157 0 .139 0 .122 0 .104 0 .086 0	.155 0 .138 0 .120 0 .102 0 .084 0	.154 0 .136 0 .119 0 .101 0 .083 0	.152 0 .134 0 .117 0 .099 0 .081 0	.150 0 .132 0 .115 0 .097 0 .079 0	.148 0 .131 0 .113 0 .095 0 .077 0	.146 0 .129 0 .111 0 .093 0 .075 0	.145 0 .127 0 .110 0 .092 0 .074 0	.143 0 .125 0 .108 0 .090 0 .072 0
$ \begin{array}{r}     .1 \\     .2 \\     .3 \\     .4 \\     .5 \\     .6 \\   \end{array} $	.159 0 .141 0 .124 0 .106 0 .088 0 .070	.157 0 .139 0 .122 0 .104 0 .086 0 .068	.155 0 .138 0 .120 0 .102 0 .084 0 .066	.154 0 .136 0 .119 0 .101 0 .083 0 .065	.152 0 .134 0 .117 0 .099 0 .081 0 .063	.150 0 .132 0 .115 0 .097 0 .079 0 .079 0 .061	.148 0 .131 0 .113 0 .095 0 .077 0 .059	.146 0 .129 0 .111 0 .093 0 .075 0 .057	.145 0 .127 0 .110 0 .092 0 .074 0 .056	.143 0 .125 0 .108 0 .090 0 .072 0 .054
$ \begin{array}{r}     .1 \\     .2 \\     .3 \\     .4 \\     .5 \\     .6 \\     .1 \\     .6 \\   \end{array} $	.159 0 .141 0 .124 0 .106 0 .088 0 .070 0	.157 0 .139 0 .122 0 .104 0 .086 0 .068 0	.155 0 .138 0 .120 0 .102 0 .084 0 .066 0	.154 0 .136 0 .119 0 .101 0 .083 0 .065 0	.152 0 .134 0 .117 0 .099 0 .081 0 .063 0	.150 0 .132 0 .115 0 .097 0 .079 0 .079 0 .061 0	.148 0 .131 0 .113 0 .095 0 .077 0 .059 0	.146 0 .129 0 .111 0 .093 0 .075 0 .057 0	.145 0 .127 0 .110 0 .092 0 .074 0 .074 0 .056	.143 0 .125 0 .108 0 .090 0 .072 0 .054 0
$ \begin{array}{r}     .1 \\     .2 \\     .3 \\     .4 \\     .5 \\     .6 \\     .7 \\   \end{array} $	.159 0 .141 0 .124 0 .106 0 .088 0 .070 0 .052	.157 0 .139 0 .122 0 .104 0 .086 0 .068 0 .050	.155 0 .138 0 .120 0 .102 0 .084 0 .066 0 .048	.154 0 .136 0 .119 0 .101 0 .083 0 .065 0 .046	.152 0 .134 0 .117 0 .099 0 .099 0 .081 0 .063 0 .045	.150 0 .132 0 .115 0 .097 0 .079 0 .061 0 .043	.148 0 .131 0 .113 0 .095 0 .077 0 .059 0 .041	.146 0 .129 0 .111 0 .093 0 .075 0 .057 0 .039	.145 0 .127 0 .110 0 .092 0 .074 0 .056 0 .038	.143 0 .125 0 .108 0 .090 0 .072 0 .054 0 .036
$ \begin{array}{r}     .1 \\     1 \\     .2 \\     1 \\     .3 \\     1 \\     .4 \\     1 \\     .5 \\     1 \\     .6 \\     1 \\     .7 \\     1 \\     .7 \\     1 \end{array} $	.159 0 .141 0 .124 0 .106 0 .088 0 .070 0 .070 0 .052 0	.157 0 .139 0 .122 0 .104 0 .086 0 .086 0 .068 0 .068 0 .050 0	.155 0 .138 0 .120 0 .102 0 .084 0 .084 0 .066 0 .048 0	.154 0 .136 0 .119 0 .101 0 .083 0 .083 0 .065 0 .046 0	.152 0 .134 0 .117 0 .099 0 .081 0 .063 0 .045 0	.150 0 .132 0 .115 0 .097 0 .079 0 .079 0 .079 0 .061 0 .043 0	.148 0 .131 0 .113 0 .095 0 .077 0 .077 0 .059 0 .059 0 .041 0	.146 0 .129 0 .111 0 .093 0 .075 0 .075 0 .057 0 .057 0 .039 0	.145 0 .127 0 .110 0 .092 0 .074 0 .056 0 .038 0	$\begin{array}{c} .143 \\ 0 \\ .125 \\ 0 \\ .108 \\ 0 \\ .090 \\ 0 \\ .072 \\ 0 \\ .072 \\ 0 \\ .054 \\ 0 \\ .036 \\ 0 \\ \end{array}$
$\begin{array}{c} .1 \\ 1 \\ .2 \\ 1 \\ .3 \\ 1 \\ .4 \\ 1 \\ .5 \\ 1 \\ .6 \\ 1 \\ .7 \\ 1 \\ .8 \\ \end{array}$	.159 0 .141 0 .124 0 .106 0 .088 0 .070 0 .052 0 .034	$\begin{array}{c} .157 \\ 0 \\ .139 \\ 0 \\ .122 \\ 0 \\ .104 \\ 0 \\ .086 \\ 0 \\ .068 \\ 0 \\ .068 \\ 0 \\ .050 \\ 0 \\ .032 \\ \end{array}$	.155 0 .138 0 .120 0 .102 0 .084 0 .084 0 .066 0 .048 0 .048 0 .031	.154 0 .136 0 .119 0 .101 0 .083 0 .065 0 .046 0 .029	.152 0 .134 0 .117 0 .099 0 .099 0 .081 0 .063 0 .045 0 .027	.150 0 .132 0 .115 0 .097 0 .097 0 .097 0 .079 0 .079 0 .061 0 .043 0 .025	.148 0 .131 0 .113 0 .095 0 .077 0 .059 0 .059 0 .041 0 .024	.146 0 .129 0 .111 0 .093 0 .075 0 .075 0 .057 0 .039 0 .022	$\begin{array}{c} .145 \\ 0 \\ .127 \\ 0 \\ .110 \\ 0 \\ .092 \\ 0 \\ .074 \\ 0 \\ .074 \\ 0 \\ .056 \\ 0 \\ .038 \\ 0 \\ .020 \\ \end{array}$	.143 0 .125 0 .108 0 .090 0 .090 0 .072 0 .054 0 .036 0 .019
$ \begin{array}{c} .1\\ .2\\ .3\\ .3\\ .4\\ .5\\ .6\\ .7\\ .8\\ .8\\ .8\\ .1 \end{array} $	.159 0 .141 0 .124 0 .106 0 .088 0 .070 0 .052 0 .052 0 .034 0	$\begin{array}{c} .157 \\ 0 \\ .139 \\ 0 \\ .122 \\ 0 \\ .104 \\ 0 \\ .086 \\ 0 \\ .068 \\ 0 \\ .068 \\ 0 \\ .068 \\ 0 \\ .050 \\ 0 \\ .050 \\ 0 \\ .032 \\ 0 \\ 0 \\ .032 \\ 0 \\ 0 \\ 0 \\ 0 \\ .032 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ $	.155 0 .138 0 .120 0 .102 0 .102 0 .084 0 .066 0 .048 0 .048 0 .048 0 .031	.154 0 .136 0 .119 0 .101 0 .083 0 .065 0 .046 0 .046 0 .029 0	.152 0 .134 0 .117 0 .099 0 .099 0 .081 0 .063 0 .045 0 .045 0 .027 0	.150 .132 0 .115 0 .097 0 .079 0 .061 0 .043 0 .043 0 .025 0	.148 0 .131 0 .113 0 .095 0 .077 0 .077 0 .077 0 .059 0 .041 0 .024 0	.146 0 .129 0 .111 0 .093 0 .075 0 .075 0 .057 0 .039 0 .039 0 .022 0	$\begin{array}{c} .145 \\ 0 \\ .127 \\ 0 \\ .110 \\ 0 \\ .092 \\ 0 \\ .074 \\ 0 \\ .074 \\ 0 \\ .056 \\ 0 \\ .038 \\ 0 \\ .038 \\ 0 \\ .020 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 $	$\begin{array}{c} .143 \\ 0 \\ .125 \\ 0 \\ .108 \\ 0 \\ .090 \\ 0 \\ .072 \\ 0 \\ .072 \\ 0 \\ .072 \\ 0 \\ .072 \\ 0 \\ .072 \\ 0 \\ .019 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ 0 \\ 0 \\ .019 \\ .010 \\ 0 \\ .019 \\ 0 \\ .010 \\ 0 \\ 0 \\ .010 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 \\ 0 $

**3.1.2.** Detection of glycogen in the liver.

3.2.2. Principle of the method:

Determination of glycogen is carried out with the help of Lugol's solution, and is also confirmed by its hydrolysis to glucose under the action of saliva amylase.

The main stages of the work:

1. Filtrate production:

2.5 grams of liver are mixed in a porcelain mortar, to which 4 ml of dist. H2O and grind with glass sand. The homogenate is quantitatively transferred to a flask and 20 ml of dist. H2O, after which the sample is boiled in a water bath for 20-30 minutes. For more complete precipitation, add 5-10 drops of 1% acetic acid solution to the boiling liquid. The protein precipitate is separated by filtering through a paper filter moistened with water.

2. Carrying out qualitative reactions:

For this, take 4 test tubes and number them with a glass pencil:

№1 control,

№2 glycogen deposition,

№3 digestion of glycogen, №4 pure glycogen.

	1	2	3	4
no				
	H2O dist	Filtrate-5	Filtrate-5	Filtrate-5 drops.
1.	5 drops.	drops.	drops.	
	Lugol's	2. sulfuric	2. saliva	2. Fehling's solution - 2
2	solution - 2	acid	(1:10)-2 drops.	drops. heat up
	drops.			
		3. Lugol's	3. Lugol's	
	-	solution - 2	solution - 2	_
3		drops.	drops.	

A change in color is observed

**3.3. Requirements for work results:** in a notebook for demonstration and practical work, write down the method of work performance, the results of observation. Provide a medico-biological evaluation of the obtained results. Draw conclusions.

**3.4.** Control materials for the final stage of the class:test tasks for the topic:

1. In a patient exhausted by starvation, the process in the liver and kidneys intensifies:

- A Bilirubin synthesis
- B Urea synthesis
- C Gluconeogenesis
- D Formation of hippuric acid
- E Uric acid synthesis

2. For the synthesis of polysaccharide chains of glycogen, a precursor - the active form of glucose - is used. The direct donor of glucose residues in the process of glycogen synthesis is:

- A ADP-glucose
- B glucose-1-phostat
- C UDF-glucose
- D glucose 6-phosphate
- E glucose-3 phosphate

3. When there is a lack of blood circulation during intense muscle work, lactic acid accumulates in the muscle as a result of anaerobic glycolysis. What is her future fate?

- A Involved in gluconeogenesis in the liver
- B It is removed through the kidneys with urine
- C It is used in the muscle for the synthesis of amino acids
- D It is used by tissues for the synthesis of ketone bodies
- E It is used in tissues for the synthesis of fatty acids

4. A characteristic sign of glycogenosis type V (McArdle's disease) is muscle pain during physical work. Congenital deficiency of which enzyme causes this pathology?

A Glycogen synthases

- In Glucose-6-phosphatase
- C Glycogen phosphorylase
- D Amylo-1,6-glycosidases
- E Lysosomal glycosidase

5. In Itsenko-Cushing's disease (hyperfunction of the adrenal cortex with increased production of corticosteroids), hyperglycemia occurs. What process is stimulated in this case?

A Gluconeogenesis

- B Glycogen phosphorolysis
- C Krebs cycle
- D Pentose phosphate pathway of glucose oxidation
- E Glycolysis

6. As a result of prolonged starvation in the human body, reserves of carbohydrates quickly disappear. Which of the metabolic processes restores the glucose content in the blood?

- A Glycogenolysis
- B Anaerobic glycolysis
- C Aerobic glycolysis
- D Hyukoneogenesis
- E Pentophosphate pathway

7. A 40-year-old woman with complaints of thirst and increased appetite is being treated in the endocrinology department with a diagnosis of diabetes. What pathological components were found during the laboratory examination of the patient's urine?

- A Glucose, ketone bodies
- B Protein, amino acids
- C Protein, creatine
- D Bilirubin, urobilin
- E Blood

8. With a chronic overdose of glucocorticoids, the patient develops hyperglycemia. Name the process of carbohydrate metabolism due to which the concentration of glucose increases:

- A Aerobic glycolysis
- B Seeandcogenolandwith
- C Gluconeogenesis
- D Pentose phosphate cycle
- E Seeandcogenesis

9. A 40-year-old woman has Itsenko-Cushing's disease - steroid diabetes. During biochemical examination: hyperglycemia, hypochloremia. Which of the following processes is activated first?

- A Gluconeogenesis
- B Glycogenolysis
- C Reabsorption of glucose
- D Transport of glucose into the cell
- E Glycolysis

10. In the patient's blood, the glucose content on an empty stomach is 5.6 mmol/l, after 1 hour after the sugar load - 13.8 mmol/l, and after 3 hours - 9.2 mmol/l. Such indicators are likely for:

- A Hidden form of diabetes
- B A healthy person
- C Thyrotoxicosis
- D Itsenko-Cushing diseases
- E Acromegaly

11. In a patient with a diagnosis of Itsenko-Cushing's disease (hyperproduction of the adrenal cortex), an increased concentration of glucose, ketone bodies, and sodium was determined in the blood. What biochemical mechanism is the leading cause of hyperglycemia?

- A Glycogenesis
- **B** Gluconeogenesis
- C Glycogenolysis

- D Glycolysis
- E Aerobic glycolysis

12. A one-year-old child lags behind his peers in mental development. In the morning: vomiting, convulsions, loss of consciousness. Fasting hypoglycemia in the blood. Which enzyme defect is this associated with?

A. Glycogen synthases

- B. Phosphorylases
- C. Arginases
- D. Sucrase
- E. Lactases

13. A patient with signs of acute alcohol poisoning was brought to the clinic. What changes in carbohydrate metabolism are characteristic of this condition

- A. The rate of gluconeogenesis decreases in the liver
- B. Glycogen breakdown increases in the liver

C. Anaerobic breakdown of glucose prevails in muscles

D. Gluconeogenesis increases in the liver

E. Aerobic breakdown of glucose increases in muscles

14. As a result of prolonged fasting, hypoglycemia occurs, which is aggravated by alcohol, because it slows down:

And Lipolysis

In Glycolysis

C Glycogenolysis

D Gluconeogenesis

E Proteolysis

# 4. Summary of the class. Assessment.

# 5. List of recommended literature

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

# Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

# **Electronic information resources:**

1. https://info.odmu.edu.ua/chair/biology/-

2. http://libblog.odmu.edu.ua/

3. https://moodle.odmu.edu.ua/login/index.php

#### Practical class No8

Topic: The role of lipids in the structure and functions of biological membranes. Establishment of molecular mechanisms of lipolysis regulation. Tissue, intracellular exchange of lipids. Oxidation of higher fatty acids and glycerol. Bioenergetics of the oxidation of VHF.

**Goal:**Formation of systemic knowledge on the structure of biomembranes, their role in cell protection, transmission of nerve impulses, creation of intercellular contacts. Study of molecular mechanisms of lipolysis and its regulation. Compilation of the energy balance of the oxidation of VHL and glycerol.

**Basic concepts:**lipids, biomembranes, lipolysis, hormonal mechanisms of lipolysis regulation, higher fatty carboxylic acids, beta-oxidation, glycerol, energy balance of oxidation.

Equipment: laboratory of the department.

Plan:

**1. Organizational measures:** greetings, verification of those present, announcement of the topic, purpose of the class, motivation

## 2. Control of the reference level of knowledge

## 2.1. Requirements for higher education applicants' theoretical readiness:

1. Define lipids as esters.

2. Alcohols included in the structure of lipids (glycerol, myricyl alcohol, cholesterol)

3. Higher fatty acids are structural components of lipids. Saturated and unsaturated higher fatty acids. Give their formulas.

4. What is the biological role of polyunsaturated fatty acids in the body.

5. Classification of lipids (simple, complex, derivatives of lipids). Give the formulas of simple and complex lipids.

6. Fats (neutral - triglycerides, charged - phospholipids). Features of the structure of solid fats and liquid oils.

#### 2.2. Questions to check basic knowledge on the topic of the class:

1. Biological role of lipids and higher fatty acids in the body.

- 2. Types, functions and chemical composition of biomembranes.
- 3. Lipolysis. Hormonal regulation of lipolysis

4. Enzymatic digestion of lipids.

5. Modern ideas about the mechanisms of absorption of lipids from the intestines into the blood and lymph. The role of bile acids in the absorption of lipid digestion products.

6. Write the activation reaction of glycerol and the scheme of complete oxidation of glycerol to CO2 and H2O.

7. Calculate the number of ATP molecules formed during the oxidation of glycerol to  $CO_{2}$  and H2O.

8. Write the activation reaction of a higher fatty acid (acyl), as well as the reaction of acyl-CoA transport through the inner membrane of mitochondria.

9. Write the sequence of acyl-CoA oxidation reactions inside mitochondria to form acetyl-CoA.

10. What is the meaning of oxidation of higher fatty acids in tissues? Calculate the energy balance of the oxidation of palmitic and stearic acids (by the number of ATP molecules formed).

11. Give examples of the relationship between carbohydrate and lipid metabolism. Name the common metabolites of carbohydrate and lipid oxidation processes.

**3. Formation of professional skills and abilities:**demonstration and practical work.

**3.1.** Content of demonstration and practical work

**3.1.1.**Observation of the effect of bile on fat emulsification.

#### **3.2. Implementation recommendations:**

**3.2.1.**Measure 1 ml of water into 4 test tubes. Add 2 drops of oil to the first test tube, 2 drops of 0.5% baking soda and 2 drops of oil to the second, 1 ml of protein and 4 drops of oil to the third.

In the fourth - 1 ml of bile and 2 drops of oil. Mix the contents of the test tubes by vigorous shaking. Note: bile lowers the surface tension of fat droplets, forming stable emulsions.

**3.1.2.** Observation of the effect of pancreatic lipase on milk fat with and without bile.

**3.2.2**.Pour 1 ml of milk into 3 test tubes. Add 0.5 ml of water to the first and second. Add 0.5 ml of bile to the first test tube. Shake all the test tubes, add 2 drops of an alcoholic solution of phenolphthalein and an aqueous solution of KHSO3 to each of them until it turns pale pink. Place all tubes in a thermostat at 37°C for 30 min. draw a conclusion

Note: pidina will become discolored when the pH of the medium changes due to the formation of fatty acids during the splitting of milk fat by lipase.

**3.3. Requirements for work results:** in a notebook for demonstration and practical work, write down the method of work performance, the results of observation. Provide a medico-biological evaluation of the obtained results. Draw conclusions.

**3.4.** Control materials for the final stage of the class:test tasks for the topic:

1. What lipids make up the basis of fat depots in the body?

And steroids

In phospholipids

With triglycerols

D sphingolipids

E glycolipids

2. Which enzyme catalyzes the hydrolysis of triglycerides in the intestinal cavity?

And monoglyceride lipases

In acetylcholinesterase C transacetylase

D lipase

E phospholipase

3. High lipase activity was detected in the gastric juice of a 6-month-old child. What is the optimal pH of this enzyme?

A- 7.8

B - 5.5

C - 3.2

D - 1.5

E - 9.5

4. Which pancreatic enzyme is activated by bile acids? And proelastase In lipase C oligo-1-6-glucosidase

D trypsinogen

E chymotrypsinogen

5. Which protein of the pancreas takes part in the emulsification of fats?

And elastase

In trypsin

C collagenase

D chymotrypsin

E colipase

6. Which of the following substances are surface active and take part in the emulsification of fats?

And glycosidases

In bicarbonates With bile acids D proteases E glycosaminoglycans

7. Deficiency of which enzyme is most often the cause of incomplete digestion of fats in the gastrointestinal tract and an increase in the amount of neutral fat in feces?

And enterokinase In intestinal lipase With pancreatic lipase D gastric lipase E liver lipase

8. In the bilipid layer of membranes, a protein molecule is fixed with the help of bonds: And hydrophobic and ionic
In peptide and disulfide
With electrostatic, hydrophobic
D ionic and hydrogen
E of hydrogen and disulfide

9. The gradual process of lipolysis in adipocytes is enzymatic. Which of these enzymes controls the slowest stage of lipolysis?

And diglyceridlipase In triglyceride lipase C monoglyceride lipase D glycerol acyltransferase E phosphotidat phosphatase

10. The activity of tissue triglyceride lipase is regulated by hormones And vasopressin, oxytocinIn adrenaline, insulin, glucagon, somatotropinC cortisol, corticosteroneD thyroxine, triiodothyronineE prostaglandins

11. A patient with pulmonary emphysema was prescribed oxygen treatment, which will probably lead to an increase in the activity of oxygen radicals in the body. What can this lead to?

And destruction of membrane phospholipids In increasing the number of membranes With a violation of the structure of protein components of membranes D increase of transport proteins on membranes E oxidation of cholesterol.

12. The doctor recommended a bile preparation to the patient to improve the digestion of fatty food. What components of this drug are involved in the emulsification of fats?

And diglycerides; Cholesterol and its esters; C higher fatty acids; D bilirubin, glucuronides; E bile acids.

13. What is the transformation of glycerol formed during the breakdown of triacylglycerols in

the first stage? And phosphorylation In oxidation With methylation D recovery E acylation

14. The central intermediate product of metabolism (proteins, lipids, carbohydrates) is:And citrateIn succinyl-CoAC oxalic-acetic acidD pyruvateE acetyl-CoA

15. During the oxidation of organic substances in the body, endogenous water is formed, which in conditions of "dry starvation" often compensates for the lack of water. Which of these substances gives the largest amount of water (per unit mass of this substance)?

And glycerol In glucose With proteins D glycogen Fats

16. Which of these substances transports acyl-CoA from the cytoplasm to the mitochondria? And carnitineIn citrateC oxaloacetateD creatinineE carnosine

17. Fatty acids, as high-calorie compounds, undergo transformations in the mitochondria, as a result of which a large amount of energy is produced. Name this process.

And demining In decarboxylation With transamination D beta oxidation E recovery

18. Cardiac muscle is characterized by the aerobic nature of substrate oxidation. The main ones

are:

- A fatty acids
- B triacylglycerols
- C glycerol
- D glucose
- E amino acids

19. To improve results, the athlete was recommended to use a drug that contains carnitine. What process is activated to the greatest extent by carnitine?

- A transport of fatty acids in mitochondria
- B synthesis of ketone bodies
- C synthesis of lipids
- D tissue respiration

E synthesis of steroid hormones

20. The administration of mildronate, an inhibitor of carnitine synthesis, to patients with coronary artery disease leads to a significant inhibition in cardiomyocytes:

And  $\beta$ -oxidation of fatty acids In gluconeogenesis With the synthesis of fatty acids D synthesis of 2,3-DFH E absorption of glucose

21. A 1-year-old child with signs of muscle damage was admitted to the clinic. After the examination, a deficiency of carnitine in the muscles was revealed. The biochemical basis of this pathology is a violation of the process:

- A transport of fatty acids in mitochondria
- B regulation of Ca level<sup>2+</sup>in mitochondria
- C substrate phosphorylation
- D utilization of lactic acid
- E synthesis of actin and myosin

22. With systematic intense physical exertion, the content of fat in adipose tissue decreases. It leaves the cells in the blood in the form of:

- A glucose
- B chylomicrons
- C lipoproteins
- D ketone bodies
- E free fatty acids and glycerol

23. A vitamin-like compound is necessary for the activation and transfer of UVK through the mitochondrial membrane. Specify it.

- A biotin
- B carnitine
- C riboflavin
- D uiquinone
- E thiamine

#### 4. Summary of the class. Assessment.

## 5. List of recommended literature

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

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Additional:

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2. http://libblog.odmu.edu.ua/

3. https://moodle.odmu.edu.ua/login/index.php

#### *Practical class №9* Topic: Biosynthesis of glycerol, VHL and glycerides. Formation of phospholipids

**Goal:**Formation of systemic knowledge on intracellular lipid exchange. Study of the molecular mechanisms of the biosynthesis of VLKA, glycerol, triglycerides and phospholipids.

**Basic concepts:**higher fatty carboxylic acids, biosynthesis, glycerol, triglycerides, phospholipids.

**Equipment:**laboratory of the department.

## Plan:

**1. Organizational measures:** greetings, verification of those present, announcement of the topic, purpose of the class, motivation

2. Control of the reference level of knowledge

**2.1. Requirements for theoretical readiness of higher education applicants:** 

1. What components are necessary for the biosynthesis of simple fat - triglyceride?

2. What is the name of the active form of glycerin? Write the formula.

3. What is the active form of higher fatty acids called?

4. Name the sources of glycerol formation in the body?

5. What components are included in the composition of phospholipids - lecithin and kephalin?

2.2. Questions to check basic knowledge on the topic of the class:

1. Explain the necessity and mechanism of acetyl-CoA transport from mitochondria to cytoplasm.

2. Write the equation for the formation of malonyl-CoA, explain the role of active CO2 in this process.

3. Structure of the multienzyme complex "synthase of higher fatty acids".

4. Write the stages of biosynthesis of a saturated higher fatty acid:

A) binding of acetyl-CoA to APB;

B) binding of malonyl-CoA to APB;

C) condensation of acyl from malonyl derivatives on APB;

D) reduction of  $\beta$ -keto-butyryl-APB;

D) dehydration of  $\beta$ -hydroxybutyryl-APB;

E) reduction of enoyl-APB followed by elongation of the butyryl-APB carbon chain.

5. Write the equation of the enzymatic reactions of the formation of the active form of glycerol in adipose tissue and in the liver.

6. Write the reaction of the synthesis of phosphatidic acid followed by the formation of triglyceride.

7. Phospholipid synthesis reactions.

**3. Formation of professional skills and abilities:**demonstration and practical work.

3.1. Content of demonstration and practical work

Determination of total lipids in blood serum by the method of Bang.

## **3.2. Implementation recommendations:**

The principle of the method

Determination of total lipids in blood serum is carried out after their extraction with alcohol and ether. At further stages of determination, lipids are subjected to oxidation with potassium dichromate followed by iodometric titration of dichromate, sodium hyposulfite solution in test and control samples. Based on the difference between the number of milliliters of sodium hyposulfite used for the titration of control and experimental samples (K - D), the formula calculates the amount of total lipids in the blood, which normally ranges from 350 to 800 mg/100 ml, or 3.5 to 8. 0 g/l serum.

The main stages of work performance.

1. Extraction of lipids according to Bloor: add 0.2 ml of blood serum drop by drop to 4 ml of alcohol with ether /3:1/. Heat in a sand bath for 15 minutes. filter through a paper filter, which is washed with 1 ml of a mixture of alcohol and ether, and bring the volume to 15 ml.

Notes: the normal concentration of total lipids is 3.5 -8.0 gper liter of blood serum or 350-800 mg/per 100 ml of serum.

Pour 2.5 ml of filtrate into a test tube, corresponding to 0.1 ml of blood serum, add 1.0 ml of 1% caustic sodium solution and heat in a sand bath until the ether smell disappears. The control sample instead of blood serum contains 1.5 ml of water and 1 ml of 1% sodium hydroxide solution.

Note: All of the above is performed by the educational laboratory.

2. Oxidation of lipids with dichromate. After removing the ether, add 2 ml of 0.1N potassium dichromate solution and 3 drops of concentrated sulfuric acid to the filtrate. Place the test tube in a boiling water bath for 15 minutes, after heating carefully add water to the test tube, transfer all the liquid to a flask and add up to 100 ml of distilled water.

3. Iodometric titration of dichromate. Add 2 ml of potassium iodide solution, 2 drops of 1% starch solution, and titrate the contents of the flasks with 0.01N sodium hyposulfite solution to a pale green color. In parallel with the test sample, put a control, where instead of blood serum, take 1.5 ml of water and 1 ml of 1.0% caustic sodium solution. The difference between the control and the experiment divided by 2.45 corresponds to the amount in mg of lipids in the blood.

Counter - experiment Calculation: <u>• 10,000 g/l</u> 2.45 Make a calculation and give a medical biological accessment of the resu

Make a calculation and give a medical - biological assessment of the result.

Medical and biological evaluation of the obtained results.

An increase in the content of total lipids in the blood (hyperlipemia) is a physiological phenomenon that occurs 1-3 hours after eating. This is the so-called alimentary hyperlipemia, which is temporary in healthy people.

In diabetes, hyperglycemia occurs simultaneously with hyperlipemia (i.e. 500 - 1000 mg or more / 100 ml)

In acute hepatitis, hyperlipemia is a constant phenomenon, especially in the presence of jaundice.

In acute and chronic nephritis accompanied by edema, the amount of lipids in the blood is increased.

With lipoid nephrosis, the increase in the amount of lipids in the blood reaches 1000 mg or more /100 ml.

With chronic malnutrition and starvation, the amount of total lipids in the blood is increased due to the mobilization of fat from fat depots.

**3.3. Requirements for work results:** in a notebook for demonstration and practical work, write down the method of work performance, the results of observation. Provide a medico-biological evaluation of the obtained results. Draw conclusions.

**3.4.** Control materials for the final stage of the class:test tasks for the topic:

1. What substances transport acetyl-CoA from the mitochondria to the cytoplasm? And citrate, carnitine

In malonyl-CoA, butyryl-CoA C malate, lactate D acyl-CoA, acetoacetyl - CoA E carnosine, anserine

2. A 65-year-old patient with signs of general obesity and the risk of fatty liver dystrophy is recommended a diet enriched with lipotropic substances, among which the content in products is important:

A methionine B cholesterol C glucose D vitamin C E glycine

3. What process is the source of NADPH2 for the synthesis of VHL?And mitochondrial oxidationIn the pentose phosphate pathway of glucose oxidationC oxidation of pyruvate to acetyl-CoAD glucose oxidation to lactateE oxidation of ketoglutarate

4. In which tissue does the biosynthesis of triglycerides occur?And musclesIn the intestinesC adipose tissueD liverE mammary gland during lactation

5. Name the enzyme that catalyzes the formation of the active form of glycerol only in the liver.

And dioxyacetone phosphate dehydrogenase

B glycerol phosphate dehydrogenase;

C glycerol phosphate transferase;

D glycerol acyltransferase;

E glycerol kinase.

6. Fatty degeneration of the liver develops in the absence or insufficient formation of lipotropic factors in the human body. Which of the following substances can be classified as lipotropic?

- A Cholesterol
- B Holin
- C Triacylglycerides
- D Fatty acids
- E Riboflavin

7. Which enzyme of biosynthesis of VLDL is biotin-containing:

And acetyl-transacetylase

In acetyl-CoA-carboxylase

C  $\beta$ -ketoacyl-APB reductase

D β-hydroxybutyryl-APB dehydratase

E enoyl-APB reductase

8. When the substance A labeled by carbon is administered to experimental rats, the label is

incorporated into glycerophospholipids and triglycerides, thus A is a common precursor in the biosynthesis of these lipids. Name the substance A.

And methionine; In ethanolamine; C choline; D cytosine triphosphate; E phosphatidic acid.

9. The drug "Heptral", which is used for liver diseases, contains S-adenosine methionine. In what processes is this active amino acid involved?

And heme synthesis In the synthesis of bile acids Synthesis of triacylglycerols D synthesis of cholesterol E synthesis of phospholipids

10. Excessive consumption of carbohydrates (600 g per day), which exceeds the energy needs of a 28-year-old person, will be accompanied by the activation of:

A lipolysis B lipogenesis C of glycolysis D gluconeogenesis E β-oxidation of fatty acids

11. Fatty degeneration of the liver during starvation and diabetes develops because hepatocytes: And the intake of fatty acids from adipose tissue increases Fatty acid oxidation decreases

C reduces the formation of ketone bodies from fatty acids

And the synthesis of triacylglycerols decreases

And the formation of high-density lipoproteins decreases

12. Biosynthesis of fatty acids is constantly taking place in the body. Which of the listed substances is the main source of their biosynthesis?

A succinyl-CoA B glucose-6-phosphate C acetyl-CoA D acyladenylate E aminoacyladenylate

13. The patient consumes several raw eggs daily, which contain the antivitamin biotin-avidin. What disorders in lipid metabolism can occur in this case?

A biosynthesis of cholesterol B biosynthesis of fatty acids C oxidation of glycerol D absorption of lipids E transport of lipids in the blood

14. The doctor recommended a patient with coronary heart disease to use fats containing polyunsaturated higher fatty acids. What components of biomembranes are synthesized with the participation of these substances?

A phospholipids B cholesterol C proteins

# D glycolipids

## E lipoproteins

15. In the human body, both triacylglycerols [neutral fats] and glycerophospholipids are synthesized from the same precursor, namely from:

A acetic acid.

B orotic acid.

C phosphatidic acid.

- D lipoic acid.
- E malonic acid.

4. Summary of the class. Assessment.

## **5.** List of recommended literature

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

# Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

# **Electronic information resources:**

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

# Practical class №10

Topic: Structure, biological role and metabolism of cholesterol. Cholesterol biosynthesis. Disorders of lipid metabolism. Lipoproteins, structure and functions. Exchange of acetoacetic acid. Ketone bodies.

**Goal:**Formation of systemic knowledge on intracellular lipid exchange, as well as lipid transport by blood and lymph. Study of molecular mechanisms of synthesis of cholesterol and ketone bodies. Determination of their biological role and causes of lipid metabolism disorders.

**Basic concepts:**cholesterol, acetoacetic acid, ketone bodies, blood plasma lipoproteins, atherosclerosis, gallstone disease.

**Equipment:**laboratory of the department.

Plan:

**1. Organizational measures:**greetings, verification of those present, announcement of the topic, purpose of the class, motivation

2. Control of the reference level of knowledge

#### 2.1. Requirements for higher education applicants' theoretical readiness:

1. How do you understand the process of fat mobilization from fat depots? Under the action of which enzymes does it occur? Name them.

2. What products are formed from the fat of fat depots under the action of tissue lipases, what type of chemical reactions do they catalyze?

3. Name the intermediate and final products of beta-oxidation of UVC.

4. Define steroids.

5. To which group of organic compounds (by chemical structure) does cholesterol belong? Write its formula.

6. Due to what is the cholesterol fund formed in the body?

7. How much cholesterol (on average) is in tissues?

## 2.2. Questions to check basic knowledge on the topic of the class:

1. Write the synthesis reactions of acetoacetic acid followed by the formation of betahydroxybutyric acid and acetone.

2. Pathways of acetoacetate metabolism in tissues.

3. Biological role of ketone bodies in the human body.

4. Explain the reasons (from a biochemical point of view) for the occurrence of acetonemia and acetonuria.

5. Give examples of the relationship between carbohydrate and lipid metabolism. Name the common metabolites of carbohydrate and lipid oxidation processes.

6. Which tissues of the human body are the richest in cholesterol?

7. What is cholesterol synthesized from? Write the initial reactions of cholesterol biosynthesis (up to and including mevalonic acid).

8. Name the intermediate products of cholesterol biosynthesis. Explain the structural features of squalene and lanosterol as important intermediates in cholesterol biosynthesis. What is the difference between them?

9. Regulation of cholesterol biosynthesis.

10. Cholesterol as a precursor of other steroids in the body. Biological role of cholesterol.

11. Name the transport forms of lipids in the blood. Their structure.

12. General concepts about the mechanisms of development of atherosclerosis and gallstone disease

3. Formation of professional skills and abilities: demonstration and practical work.

## **3.1.** Content of demonstration and practical work

**3.1.1.**Qualitative reactions to acetone (iodoform, nitroprusside)

#### **3.2. Implementation recommendations:**

**3.2.1.**Lieben's iodoform test. Pour 2 ml of the tested urine into the test tube, add 1 ml of 10% caustic potassium solution and 6 drops of Lugol's solution. Mix the contents of the tube by shaking. Make a conclusion.

Notes:

In the presence of acetone, the solution becomes cloudy and a light yellow precipitate falls out. Rother's nitroprusside test.Pour 2 ml of the tested urine into the test tube, add 0.5 ml of sodium nitroprusside and 0.5 ml of saturated ammonium sulfate solution. Mix the contents of the test tube by

shaking and carefully pour 2 ml of ammonia solution along the wall. Watch. Make a conclusion.

Notes:

In the presence of acetone, a purple ring /coloring/ appears at the liquid boundary.

**3.1.2.**Quantitative determination of acetone in urine by the Rudogi method

**3.2.2.** Pour 1 ml of saturated sodium chloride solution into 6 test tubes (except the first one), and 2 ml of the tested urine into the first one. From the first tube, transfer 1 ml to the second, and from the second to the third, etc., take 1 ml from the last. Thus, in each subsequent test tube, the amount of urine will be 2 times less than in the previous one. That is, we will obtain a number of dilutions of 2, 4, 8, 16 and 32 times. Then add 0.5 ml of sodium nitroprusside solution and 1 ml of

saturated ammonium sulfate solution to all test tubes and carefully layer 2 ml of ammonia solution; determine the layering time. Note in which test tube a purple ring appeared before the end of 4 min. Do the calculation, taking into account the dilution of urine in this test tube. Rate the result.

#### Notes

The time of appearance of color depends on the concentration of acetone. At a concentration of 0.85 mg%, a colored ring appears between 3.5-4 min. after ammonia layering.

Medical and biological evaluation of the obtained results

Acetone and other acetone bodies, if they are present in urine in detectable quantities, are pathological components of urine. If acetone is present in the urine, it means that it also contains other acetone bodies (acetoacetic and beta-hydroxybutyric acids). This condition - acetonuria, ketonuria - indicates a violation of lipid metabolism. During the intensive splitting of fats in the body (in the case of diabetes), a significant number of acetyl coenzyme-A molecules accumulate, which do not have time to be oxidized in the cycle of tricarboxylic acids to end products. The conditions for the transformation of acetoacetyl-CoA through an intermediate product to acetoacetic, beta-hydroxybutyric acids and acetone are created. Acetonuria is observed in severe forms of diabetes with impaired carbohydrate and lipid metabolism.

**3.1.3.**Qualitative response to the presence of cholesterol in the brain

**3.2.3.**Brain tissue is rich in cholesterol, which is a cyclic, monounsaturated alcohol. During the interaction of sulfuric acid with cholesterol, its dehydration occurs, i.e. splitting of H2O, as a result of which cholesterol turns into an unsaturated cyclic hydrocarbon of red-brown color.

Sequence of actions. Cholesterol is extracted from the brain by grinding a piece of tissue in a mortar, with the addition of 2 ml of chloroform.

Filter the resulting homogenate into a test tube through cotton wool.

To the obtained chloroform extract containing cholesterol, add (Caution!) 2 ml of conc. sulfuric acid (H2SO4).

Carefully mix the liquids. Let stand.

After settling and placement of liquids, observe the appearance of a red-brown color in the upper layer of the liquid, while the lower layer remains yellowish-green, the lower layer is occupied by sulfuric acid.

Medical and biological evaluation of the obtained results

Qualitative determination of cholesterol in the brain tissue confirms the known data that in the human body the most cholesterol is in the nervous tissue (myelin sheath) and in the cortex of the adrenal glands.

In human tissues - near140 gcholesterol Part of the cholesterol in the tissue is esterified by VHL, mainly oleic. Cholesterol esters are, as a rule, its stored or transport form. 2/3 of plasma lipoprotein cholesterol is esterified in blood, and the same amount of cholesterol is esterified in adrenal cells. In most other organs and tissues, cholesterol esters make up a smaller part of it (in the liver, for example, only 20-25%).

The cholesterol fund in the body is created due to food cholesterol (about 0.3 g per day). When eating plant-based food, low in cholesterol, the biosynthesis of cholesterol in the body is of leading importance.

**3.3. Requirements for work results:** in a notebook for demonstration and practical work, write down the method of work performance, the results of observation. Provide a medico-biological evaluation of the obtained results. Draw conclusions.

3.4. Control materials for the final stage of the class:test tasks for the topic:

1. During the examination of the patient, an increased content of low-density lipoproteins in the blood serum was found. What disease can be predicted in this patient?

- A Gastritis
- B Kidney damage
- C Acute pancreatitis
- D Atherosclerosis
- E Pneumonia

2. In the human body, the main place of deposition of triacylglycerols (TAG) is adipose tissue. However, their synthesis occurs in hepatocytes. In the form of what is the transport of TAG from the liver to adipose tissue?

- A LDL
- B Chylomicrons
- C LPDNSH
- D HDL
- E Complex with albumin

3. An increase in the level of HDL leads to a decrease in the incidence of atherosclerosis. What is the mechanism of antiatherogenic effect of HDL?

- A Cholesterol is removed from tissues
- B Cholesterol is supplied to tissues
- C Take part in the breakdown of cholesterol
- D Activate the transformation of cholesterol into bile acids
- E Contribute to the absorption of cholesterol in the intestine

4. During the patient's examination, bile stasis in the liver and gallstones in the gallbladder were found. Name the main component of gallstones that form in this condition.

- A White.
- B Triglycerides.
- C Calcium bilirubinate.
- D Cholesterol.
- E Mineral salts.

5. With diabetes and starvation, the content of acetone bodies, which are used as an energy material, increases in the blood. Name the substance from which they are synthesized:

- A Malat
- B Succinyl-CoA
- C Citrate
- D Acetyl CoA
- E Ketoglutarate

6. Ketone bodies were found in the patient's urine. In which disease do ketone bodies appear in the urine?

A Diabetes mellitus.

B Acute glomerulonephritis.

- C Urinary stone disease.
- D Kidney tuberculosis.
- E Renal infarction.

7. During a blood test, a high cholesterol content in the  $\beta$ -lipoprotein fraction was revealed. What are the possible consequences of this phenomenon for the body?

- A Hypertension
- **B** Diabetes
- C Obesity
- D Atherosclerosis
- E Jaundice

8. During the treatment of patients with coronary artery disease as a result of atherosclerotic degeneration of vessels, statins are prescribed, HMC-CoA reductase inhibitors, which inhibit the

synthesis of: A Fatty acids In ketone bodies With cholesterol D Corticosteroids E Bile acids

9. Analysis of the patient's blood plasma lipid metabolism revealed the risk of developing atherosclerosis due to an increase in the ratio of lipoproteins:

And LDL/LDL In VLDL/LDL With LDL/LDL D Chylomicrons/LDL E VLDL/Chylomicrons

10. When examining the patient's blood plasma 4 hours after he took fatty food, it was found that it is cloudy. The most likely cause of this condition is an increase in plasma concentration:

A Cholesterol B HDL C LPG D Chylomicrons E Phospholipids

11. Complaints and objective data suggest that the patient has an inflammatory process in the gallbladder, a violation of the colloidal properties of bile, and the possibility of the formation of gallstones. What can mainly cause their formation?

A Cholesterol

- B Urates
- C Oxalates
- D Chlorides
- E Phosphates

12. When examining a teenager with xanthomatosis, familial hypercholesterolemia was established. The concentration of which lipoproteins is significantly increased in the blood in this pathology?

- A LPDNSH
- B Chylomicrons
- C LDL
- D HDL
- E NEZHK

13. In a sick 12-year-old boy, the cholesterol content in blood serum is up to 25 mmol/l. There is a history of hereditary familial hypercholesterolemia, the cause of which is a violation of the synthesis of receptor proteins for:

A High-density lipoproteins

- B Low-density lipoproteins
- C Chylomicrons
- D Very low density lipoproteins
- E Intermediate density lipoproteins

14. A 58-year-old man has signs of atherosclerotic damage to the cardiovascular system. An increase in which of the following indicators of biochemical blood analysis is most characteristic of

this condition?

- A LDL ( $\beta$ -lipoprotein) level
- B Ericoproteins
- C HDL (α-lipoprotein) level
- D Activities of alanine aminotransferase
- E Activities of succinate dehydrogenase

15. When examining a patient with signs of atherosclerosis in the blood, a significant decrease in HDL, an increase in LDL, and a cholesterol concentration of 11 mM/l were found. A decrease in the activity of which enzyme is the most likely cause of such changes?

A Lecithin cholesterol acyltransferase

In blood Lipoprotein lipase

With Tissue lipases

D Pancreatic phospholipases

E Cholesterolesterase

#### 4. Summary of the class. Assessment.

## 5. List of recommended literature

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

## Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

#### **Electronic information resources:**

1. https://info.odmu.edu.ua/chair/biology/-

- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

#### Practical class № 11

**Topic:** Ways of formation and maintenance of the pool of amino acids in the body. Transport of amino acids into cells. Deamination of amino acids. Mechanism of indirect deamination of L-amino acids. Decarboxylation of amino acids: enzymes, physiological significance. Oxidation of biogenic amines. Transamination. Biochemical significance, mechanisms of action of aminotransferases. Diagnostic value of determination of aminotransferases in blood serum.

**Goal:**Learn about deamination reactions, which are the central link in the intracellular metabolism of amino acids. As a result of deamination processes, ammonia is formed - a toxic substance that is subject to temporary and final detoxification processes and is excreted in the urine

in the form of end products of nitrogenous metabolism, one of which are ammonium salts. During the decarboxylation of amino acids, biogenic amines are formed, which are mediators of the central nervous system and have a hormonal effect. Know the effect of alanine aminotransferase (ALT) and aspartate aminotransferase (AST), which is widely used in the clinic for early diagnosis of hepatitis of various etiologies and early diagnosis of myocardial infarction. Transamination reactions are a central link in the metabolism of amino acids, controlling the metabolism of proteins and carbohydrates, as well as lipids.

**Basic concepts:**pool of amino acids, transport of amino acids through the biological membrane, deamination, decarboxylation, aminotransferases, transamination of amino acids, shuttle mechanisms of obtaining glutamate and alanine.

**Equipment:**Laboratory of the department

#### Plan:

1. Organizational measures (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- the composition of the biological membrane
- replaceable and essential amino acids
- α-ketoglutarate shuttle mechanism
- mechanism of decarboxylation
- The role of pyridoxal phosphate in metabolic processes.
- concept of transamination
- action of transferases
- where transamination occurs
- function of ALT and AST in transamination
- why alanine is an important substrate in the liver
- Schiff foundations
- structure of vitamin B6 and its coenzymes
- mechanism of transamination
- trasnreamination mechanism

The higher education applicant should be able to:

- write structural formulas of amino acids
- explain the role of glutathione in the transport of amino acids
- which are central nervous system mediators
- indicate reactions of amino acid metabolism
- explain the role of biogenic amines
- explain the role of PALF in the metabolism of amino acids
- know the shuttle mechanisms of pyruvate and  $\alpha$ -ketoglutarate
- distinguish the process of tracing from deamination
- explain the role of aminotransferases
- which enzyme from transamination decreases, and which increases during a heart attack
- indicators of transamination enzymes in hepatitis
- transferase research methods
- explain the creation of Schiff bases

- distinguish the action of ALT from the action of AST (with which substrates do they work)

Questions to check basic knowledge on the topic of the class:

- structural formula of pyruvate
- structural formula of  $\alpha$ -ketoglutarate
- functions of catecholamines
- structural formulas of 20 amino acids

- biological role of aminoxylotes in the human body

- the difference between replaceable and essential amino acids

Write transamination reactions between alanine and L-ketoglutarate, asparagine and L-ketoglutarate

- What is transdeamination

- What is transreamination

- Reactions of catabolism of nitrogen-free amino acid residues

- Which amino acids take part in gluconeogenesis reactions

- As a result of the exchange of which amino acids, ketone bodies are formed

- Interrelationship of amino acid and carbon exchange

- Scheme of inclusion of amino acids in CTC

3. Formation of professional skills:

3.1 Demonstration and practical work: Quantitative determination of ammonia nitrogen (ammonium salts) in urine by the Model method.

Recommendations for performing tasks

Principle of the method:

Nessler's reagent forms an orange-colored complex salt with ammonium salts, the color intensity is proportional to the amount of ammonium salts, therefore a colorimetric method of determination is used.

Progress:

Pour 0.5 ml of 10-fold diluted urine into one test tube, and 0.5 ml of standard (NH4)2SO4 solution containing 0.025 mg of nitrogen into the second. Add 0.5 ml of distilled water and 0.5 ml of ferrous salt to both test tubes. Mix, add 0.1 ml of Nessler's reagent to both test tubes.

Colorimetry on the FEK (photoelectrocolorimeter) with a green light filter (wavelength 500-600 nm) in a cuvette with a layer thickness of 1 cm against water.

The calculation is made according to the formula:

$$C_x = \frac{C_{CT} \times E_x}{E_{CT}}$$
, where

 $C_{CT}$  - 0.025 mg of nitrogen

 $E_{CT}$  - optical density of the standard sample

 $E_x$  - optical density of the test sample

 $C_x$  - the nitrogen content of 0.5 ml of urine diluted 10 times.

When completed, dilution and daily diuresis (D) are counted. According to the norm, the composition of nitrogen salts in urine is 0.5-1.2 g per day.

Clinical and diagnostic value of the method:

The amount of ammonium salts in the urine increases in chronic and severe forms of diabetes accompanied by acidosis, diffuse liver diseases with impaired urea synthesis. The amount of ammonium salts decreases with a plant-based diet and kidney disease

Make medical and biological conclusions.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.2. Demonstration and practical work "Detection of alanine aminotransferase (AlAT) in normal and pathological blood serum."

Recommendations for completing tasks:

The method is based on the study of the activity of alanine aminotransferase expressed in micromoles of pyruvic acid, which was formed during the incubation of 1 ml of serum for 1 hour at 370C according to the formula:  $x=C*2*\times10$ , where C is micromoles of pyruvate, which is found using a calibration curve, 10 is the conversion factor for 1 ml of serum, 2 is the conversion factor for

1 hour of incubation. Normally, the activity of alanine aminotransferase is equal to 0.1-0.68  $\mu$ M/h, the activity of alanine aminotransferase is 0.1-0.45  $\mu$ M/h.

#### Progress.

Measure 0.5 ml of 1% solution into two test tubes $\alpha$ - acid glutarate, 0.5 ml of 1% analin solution and 1 ml of 0.1% KHSO3 solution. Add 0.5 ml of the patient's serum to one test tube. Mix, place both test tubes for 30 minutes in a thermostat at a temperature of 37C0.

After incubation, add 0.5 ml of 2,4-dinitrophenylhydrazine solution and 0.5 ml of 0.4 N NaOH solution to each test tube, mix. Give color. If the activity of alanine aminotransferase is low, the color will be pale, if it is high, it will be dark.

We measure color on a photoelectrocolorimeter (PEKi) with a green light filter (wavelength 500-560 nm) and find the optical density. Then we calculate the activity of alanine aminotransferase with the help of a calibration curve according to the formula given earlier

Medical and biological evaluation of the obtained results.

The activity of analin and aspartate aminotransferase in the blood increases in diseases that penetrate with necrosis and tissue damage - mainly the heart muscle and liver. During a myocardial infarction, AST activity reaches its maximum after 6-12 hours, and ALT activity rises less noticeably. With infectious hepatitis, we observe the second picture - the activity of alanine aminotransferase increases much more noticeably than the activity of aspartame aminotransferase. Therefore, the aminotransferase test is a valuable diagnostic test.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Test tasks:

A 7-year-old child was brought to the emergency hospital in a state of allergic shock, which developed after a wasp bite. The blood concentration of histamine is increased. As a result of which reaction is this amine formed?

A - restoration;

B - dehydrogenation;

C - deamination;

D - hydrolysis;

E\* - decarboxylation.

A 24-year-old patient was injected with glutamic acid to treat epilepsy. The therapeutic effect in this disease is due not to glutamate itself, but to the product of its decarboxylation.

A - adrenaline:

B\* - GABA;

C - histamine;

D - serotonin;

E - dopamine.

3. A 32-year-old man was diagnosed with acute radiation sickness. A sharp decrease in the level of serotonin in platelets was established in the laboratory. The most possible reason for a decrease in platelet serotonin is a violation of the decarboxylation process:

A - serine;

B - histidine;

C - pyruvic acid;

D - tyrosine;

E\* - 5 - oxytryptophan.

The patient, who is in the gastroenterology department, is prescribed a histamine test. For what purpose is histamine administered to the patient?

A - to study the secretory function of the stomach;

B - to stimulate digestion of lipids in the intestine;

C - to assess the activity of proteolytic enzymes of the pancreas;

D - for activation of limited proteolysis in the intestine;

E\* - for studying the nitrogen balance.

5. There are several ways of neutralizing ammonia in the body, but there are specific ones for individual organs. What is the path of neutralization of ammonia typical for brain cells?

A\* - formation of glutamine;

B - formation of urea;

C - formation of asparagine;

D- formation of ammonium ion;

E - the formation of a fold.

What is the difference between reamination and deamination?

A\* – transfer of amino groups from an amino acid to a keto acid;

B – there is no difference – amino acid by deamination to keto acid;

C - in the organism of higher animals and humans, peramination does not occur; islot

D- by transferring the amino group to asparagine and glutamine with the formation of amides;

E - the formation of hydrochloric acids.

Reamination processes provide all processes, with the exception of:

A\* – binding of ammonia;

B – deamination of a number of amino acids;

C – transamination;

D-synthesis of individual (replaceable) amino acids;

E - formation of ammonia.

The vitamin takes part in transamination processes:

A – oscorbic acid;

B-thiamine;

C – biotin;

D\* – pyridoxamine;

E is routine.

ALT activity is slightly elevated. What additional sign will help establish a patient with gallstone disease, and not hepatitis?

A – transketolosis;

B – cholinesterosis;

C – glycogen synthetase;

D\* – alkaline phosphatase;

E - arginase.

The patient complained of nausea, increased fatigue. When examining blood serum, the activity of ALT was 2.3 mmol/hour, LDH was 14 mmol/hour, and the content of LDH-5 was increased. Previous diagnosis:

A – gastritis;

B-myocardial infarction;

C – gallstone disease;

D\* – hepatitis;

E - glomerulonephritis.

For a long time, the patient experienced chest pain radiating under the left scapula, and was admitted to the clinic due to the deterioration of his health. In the study of blood serum, the activity of AST-1.2 mmol/h.L, LDH-16 mmol/h.L, increased content of LDH-1. What disease can you think of?

A – gastritis;

 $B^*$  – myocardial infarction;

C – gallstone disease;

D – hepatitis;

E - glomerulonephritis.

## 4. Summary.

## 5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

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6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

#### Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

#### **Electronic information resources:**

1. https://info.odmu.edu.ua/chair/biology/-

2. http://libblog.odmu.edu.ua/

3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №12

**Topic:**Ammonia metabolism in the human body. Urea. Ways of ammonia formation. Ammonia toxicity and mechanisms of its neutralization. Transport forms of ammonia (glutamine and asparagine). Urea biosynthesis: enzyme reactions, genetic anomalies.

**Goal:**To study the main mechanism of ammonia detoxification in the human body, urea is the main end product of nitrogen metabolism.

Basic concepts:ornithine cycle, ammonia toxicity, ways of its neutralization, glutamine, asparagine.

**Equipment:**Laboratory of the department

## Plan:

1. Organizational measures (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- the amount of ammonia formed during the deamination of amino acids
- blood urea level
- transport forms of ammonia
- reasons for increasing or decreasing the level of urea in liquids
- urinary function of the liver
- excretory function of the kidneys
- state of protein metabolism

- the role of urinary function

The higher education applicant should be able to:

- write the reactions of the ornithine cycle
- write the formula of carbomoyl phosphate
- explain the difference between urea and urine
- write the formula of the vitamin present in the reaction with carbomoyl phosphate
- write reactions with consumption of ATP
- Questions to check basic knowledge on the topic of the class:

- The role of dicarboxylic acids in the processes of binding and transport of ammonia in the blood

- A compound formed from ammonia and carbon dioxide in the presence of ATP

- What vitamin is necessary for the functioning of carbamoyl phosphate synthetase
- Reaction catalyzed by ornithine carbamoyltransferase.
- The reaction of formation of citrulline, arginine in the process of urea biosynthesis.
- The energy of how many ATP molecules is spent on the synthesis of one molecule of urea
- Under which pathological conditions can the synthesis of urea increase

- What lesions of the liver lead to a decrease in its urea-forming function

3. Formation of professional skills:

3.1 Demonstration and practical work: "Quantitative determination of urea in urine" *Recommendations for performing tasks.* 

Content of laboratory work

Principle of the method: urea forms a red complex with diacetyl monooxin in the presence of Fe3+ ions and thiosemicarbazide, and its concentration is determined by the intensity of the color.

**Progress:**solutions of diacetyl monoxide, biological fluid or physiological solution and thiosemicarbazid are measured sequentially in test tubes, in accordance with the table. Urine must be diluted 30 or 100 times before starting the analysis, multiply the obtained results by the dilution factor.

Measured liquid	Experimental	Sample calibration	empty
Diacetyl monooxime	1.0 ml	1.0 ml	1.0 ml
Biological fluid	-	0.01 ml	-
Calibration solution of urea	-	0.01 ml	-
Physiological solution	-	-	0.01 ml
Thiosemicurea solution	1.0 ml	1.0 ml	1.0 ml

The test tubes are covered with foil, the contents are mixed and simultaneously placed in a boiling water bath for exactly 10 minutes. Then the test tubes are cooled under a stream of cold water. Then colorimetry is performed on a photoelectrocolorimeter.

The concentration of urea is calculated according to the formula:

Ex.

C = ----- 8.32 mmol/l, where

Ekal

C - concentration of urea; Ex. - optical density of the test sample; Ekal - optical density of the calibration sample.

Norm: blood - 2.5-8.3 mmol/l, urine - 330-580 mmol/l.

Clinical and diagnostic value of the method

The content of urea in the blood serum of healthy people is 3.3-8.3 mmol/l (20-50 mg%). A decrease in this indicator is observed in parenchymal hepatitis, cirrhosis and liver dystrophy, which are accompanied by a sharp decrease in urea biosynthesis, as well as during pregnancy and eclampsia.

An increase in the content of urea in the blood serum is one of the main signs of nephritis and tuberculosis of the kidneys, but it is also observed with increased breakdown of proteins in the body, with loss of fluid (dehydration, vomiting, diarrhea), with sepsis, fever, and excess protein nutrition.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Test tasks.

How is ammonia neutralized in the body?

A - is partially used to neutralize acids;

B - by synthesis of urea;

C - in the form of amides;

D - goes to the synthesis of ammonium salts;

E\* - in all the specified ways.

How much urea is formed per day in an adult?

A\* - 25-35 g (depending on the amount of proteins in the diet);

B - 10-35 g;

C - 35-50 g;

D - 50-75 g;

E - 2-5 g.

Ammonia is a very poisonous substance, especially for the nervous system. Which compound is particularly active in neutralizing ammonia in brain tissues?

A. Lysine.

B. Glutamic acid

C. Proline.

D. Histidine

E.\* Alanine

Citrulline and a high level of ammonia are determined in the urine of newborns. The formation of which substance is most likely disturbed?

A. Ammonia.

B\*. Uric acid.

C. Urea.

D. Creatinine.

E. Creatine

4. Summary.

5. List of recommended literature (main, additional, electronic information resources):

Main:

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#### Practical class № 13

**Topic:** Ways of metabolism of the nitrogen-free skeleton of amino acids in the human body. Glycogenic and ketogenic amino acids. Specialized ways of exchange of acyclic amino acids. Glutathione, its role in the exchange of organic peroxides. Arginine exchange, biological role of nitric oxide, NO-synthase. Features of branched-chain amino acid metabolism: participation of coenzyme forms of vitamin B12 in amino acid metabolism. Metabolic pathways of cyclic amino acids. Hereditary enzymopathies of cyclic and acyclic amino acid metabolism.

**Goal:** to investigate the ways of metabolism of the nitrogen-free skeleton of amino acids, to know their biological significance, the function of glitaton, the metabolism of cyclic amino acids.

**Basic concepts:** ways of exchange of acyclic and cyclic amino acids, synthesis of catecholamines, creatine, creatinine.

Equipment: Laboratory of the department

#### Plan:

1. Organizational measures (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- inclusion of amino acids in CTC

- structural formula of glutamine
- synthesis of glycine
- functions in serine and glycine metabolism
- threonine metabolism
- functions of individual nitrogenous and nonnitrogenous amino acids
- pathologies of amino acid metabolism
- The higher education applicant should be able to:
- describe maple syrup disease
- know the concept of cystinuria
- describe phenylketonuria
- violation of amino acid metabolism in albinism
- sources of tryptophan
- Explain the metabolic schemes of transformations of individual amino acids
- To characterize the chemistry of creatine synthesis and cleavage
- Determine creatinine in blood serum
- Interpret results regarding serum creatinine concentration

Questions to check basic knowledge on the topic of the class:

- Specialized ways of exchange of acyclic amino acids. Exchange of glycine and serine.
- The role of folate in the transfer of one-carbon radicals. Dehydrofolate reductase inhibitors.
- Exchange of sulfur-containing amino acids: methylation reactions; creatine synthesis.
- Glutathione, its role in the exchange of organic peroxides.
- Features of the exchange of amino acids with branched chains.

- Participation of coenzyme forms of vitamin B12 and biotin in amino acid metabolism.

- Arginine exchange; biological role of nitric oxide, NO-synthase.

- Specialized pathways of cyclic amino acid metabolism.

3. Formation of professional skills:

3.1. Demonstration and practical work: "Reaction to phenylpyruvic acid (Fehling's test)" *Recommendations for performing tasks.* 

Principle of the method. Phenylpyruvic acid forms a blue-green complex compound with trivalent ferrum ions.

Procedure. Add 8-10 drops of 5% FeCl3 solution to 2 ml of freshly filtered urine. In the presence of phenylpyruvic acid in the urine, a blue-green color appears after 30-60 seconds, which gradually disappears within 5-30 minutes, depending on the concentration of phenylpyruvic acid in the urine.

Clinical and diagnostic significance. Congenital absence of the enzyme phenylalanine-4monooxygenase in the liver of children leads to the blocking of the oxidation of phenylalanine to tyrosine and, accordingly, all subsequent metabolic transformations of tyrosine. Accumulation of phenylalanine and its breakdown products, including phenylpyruvic acid, in the blood and tissues causes intoxication of the body. The consequence of this is a violation of the normal development of the brain and severe nervous disorders. The diagnostic criterion of this hereditary disease is the increased content of phenylalanine in the blood, the presence of phenylpyruvic acid in the urine. Normally, the average concentration of phenylalanine in the blood of children is: up to 1 month -0.133 mmol/l, from 1 month to 1 year - 0.095 mmol/l, from 1 year to 14 years - 0.115 mmol/l.

The test for phenylpyruvic acid can be performed on filter paper. A strip of filter paper is moistened with urine, dried in air and a drop of 10% FeCl3 solution is applied. A positive test gives a blue-green color. A similar test can be performed on a dry or wet baby diaper.

#### Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Test tasks:

The patient has disturbed sleep, there is a weakening of the activity of inhibitory processes in the central nervous system, which is associated with a violation of the formation of gamma-aminobutyric acid. What substance is the precursor of GABA?

- A Histidine
- B Tryptophan
- C Methionine
- D Valin
- E Glutamate

When forming the tertiary structure of most proteins, nonpolar amino acid residues form the inner hydrophobic part of the globule. Name one of these hydrophobic amino acids.

- A valine
- B lysine
- C arginine
- D glutamic acid
- E aspartic acid

The hormone of local action, histamine, is produced in the lungs, digestive system, and skin. He is vasodilator. Indicate which compound it is as a result of decarboxylation

- is formed:
- A Histidine
- B Valina
- C Alanine
- D Serena

## E Threonine

In the process of decarboxylation of 5-hydroxytryptophan, a biogenic amine is formed, which has vasoconstrictor effect. Name this biogenic amine.

- A serotonin
- B histamine
- C gamma-aminobutyric acid
- D putrescine
- E cadaverine

In a man who suffers from chronic intestinal obstruction, the decay of proteins increases in the large intestine. What toxic substance is formed in this case with

tryptophan:

- A Indole
- B Bilirubin
- C Lactate
- D Creatine
- E Glucose

The patient warned that the use of painkillers can cause

allergic shock. An increase in the amount of which biogenic amine in the blood can be the cause such a state?

- A Histamine;
- B GABA;
- C Cadaverine;
- D Dopamine;
- E Putrescin

L-DOPA and its derivatives are used to treat Parkinson's disease. From which

Amino acids form this substance?

- A Tyrosine
- B Asparagine
- C Glutamate
- D Tryptophan
- E Arginine

In the course of histidine catabolism, a biogenic amine is formed, which has a significant vasodilating effect. Specify this substance.

- A Histamine
- B Serotonin
- C DOFA
- D Thyroxine
- E Dopamine

The patient has pronounced allergic symptoms: rashes on the body, swelling of the face, itching. From the increase in the formation of which biogenic amine is it related to?

- A Histamine
- B Serotonin
- C adrenaline
- D Norepinephrine
- E to Holin

The structure of the lateral radical is the basis of the structural classification of amino acids. Which of the listed amino acids belongs to diaminomonocarbonic ones?

- A Lysine
- B Proline
- C Valin
- D Leucine
- E Methionine

## 4. Summary.

## 5. List of recommended literature (main, additional, electronic information resources):

Main:

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7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

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Electronic information resources:

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## Practical class №14,15

Topic: Nucleotides' metabolism in tissues: purine and pyrimidine nucleotides catabolism. Disorders of purine metabolism (gout). Purine and pyrimidine nucleotides biosynthesis. Regulation of nucleotide biosynthesis. Deoxyribonucleotide biosynthesis. Formation of thymidine nucleotides: dTMF biosynthesis inhibitors as antitumor agents.

**Goal:** providing knowledge about the biological rolenucleic acids and nucleoproteins as carriers of genetic information, Studying the processes of nucleotide synthesis and the mechanisms of their regulation, using the acquired knowledge to understand the principles of inhibition of tumor processes

**Basic concepts:**nucleoproteins, nucleic acids and their levels of organization, mononucleotides, end products of purine and pyrimidine metabolism, disorders of purine metabolism, *synthesis of nucleotides, regulation of synthesis, deoxyribonucleotides, synthesis inhibitors as antitumor agents.* 

Equipment:Laboratory of the departmentPlan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- the structure of nucleoproteins and nucleic acids, their localization in the cell,
- structure of constituent parts of nucleic acids mononucleotides (DNA and RNA)
- formation of end products of purine and pyrimidine metabolism (urea and uric acid)
- disorder of purine metabolism (in gout)

- synthesis processes of purine nucleotides to inosinic acid, and then the formation of adenyl and guanyl nucleotides.

- synthesis processes of pyrimidine nucleotides with features of deoxyribonucleotide formation

- mechanisms of regulation of synthesis of purine and pyrimidine nucleotides.
- inhibitors of deoxyribonucleotide biosynthesis (dTMF) as antitumor agents.

The higher education applicant should be able to:

- whoLkandsleep to determine artandArt urinaryeatacid in urineand
- understand the synthesis schemes of purine and pyrimidine nucleotides.
- to determine inhibitors of deoxyribonucleotide synthesis.
- distinguish the causes of Lesch-Nyhan disease.

Questions to check basic knowledge on the topic of the class:

- -What are nucleoproteins and what do they consist of?
- -How are nucleosides and mononucleotides different?
- –What simple proteins are part of nucleoproteins?
- -How do you form the name of a nucleotide?
- –What are the functions of DNA and RNA in the body?
- -What is the role of nucleic acids (DNA, RNA) in protein biosynthesis (general provisions)?
- -What nitrogenous bases are in the composition of nucleic acids?
- -Define complementarity
- -Chargaff rules
- -What carbohydrates are part of DNA, RNA?
- -Types of RNA and their functions.
- What amino acids and other substances are used for the synthesis of nitrogenous bases:
- −a) purine; b) pyrimidine?
- What is the difference between purine mononucleotides and pyrimidine mononucleotides?
- Where does the synthesis of nucleic acids take place?
- Name the sources of synthesis of purine bases of nucleotides.
- Name the sources of synthesis of pyrimidine bases of nucleotides
- Biosynthesis of mononucleotides (scheme).

#### 3. Formation of professional skills and abilities.

# 3.1 Demonstration and practical work "Quantitative determination of uric acid in urine". *Recommendations for performing tasks.*

Principle of the method: discovery of uric acid salts in urine by titration with potassium permanganate.

Procedure: Measure 50 ml of urine into a flask, add 12 ml of uranium-ammonium reagent for precipitation of mucin and phosphates. Leave for 10-15 minutes, filter. Measure 60 ml of filtrate, add 5 ml of 25% ammonia to the flask and leave for 24 hours. for precipitation of uric acid salt. WARNING! This stage of work is performed by the laboratory! After 24 hours (on the second day) higher education applicants will receive samples with precipitates of uric acid salt and continue

working with them.

After receiving a sample with sediment, decant the solution, and collect the sediment on a filter, wash it 2 times with 10% ammonium sulfate, and transfer the filter together with the sediment to a flask for titration. Add 15-20 ml of distilled water and 5 ml of concentrated sulfuric acid H2S04. Titrate the contents of the flask with potassium permanganate KMpO4 (0.02N) to a pink color that does not disappear within 10 seconds.

Calculation example:

For example, 2 ml of KMpO4 solution was used for sample titration, 1 ml of KMpO4 corresponds to 1.5 mg of uric acid, and 2 ml - x. Then  $x = (2 \times 1.5):1=3$  mg of uric acid in the sample (in 50 ml). Calculate daily diuresis: 3 mg of uric acid in 50 ml, and x mg - in 1200 ml, x = 360 mg/day.

The norm is 0.3-0.5-1.2 g of uric acid per day.

Conclusion:

The presence of uric acid in terms of daily diuresis normally should not exceed 1.2 g. If this value is exceeded, it is a pathology of the breakdown of purine nucleotides.

Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

- Give the full name of the substance, which is abbreviated as UTF.
- Write the name of the substance denoted as GDF.
- Write the nucleotide consisting of AMP and uridine acid.
- What is the name of a nucleotide whose molecule contains 2.6-dihydroxypyrimidine?

- Name the nucleotide:



- Which nucleotide contains the nitrogenous base of 2-amino-6-hydroxypurine?

- What are minor nitrogenous bases?
- Name Chargaff's rules.
- What are the options for adding phosphoric acid to nucleosides?
- The phenomenon of transformation.
- Hypochromic effect; denaturation and renaturation of DNA.
- Free nucleotides and their participation in metabolic processes.
- Methods of nucleic acid research.
- Compare the reactions of sequential cleavage of AMP and HMF.
- Reactions of microsomal oxidation in the process of catabolism of purine nucleotides.
- Gout: causes and symptoms of the disease.
- Lesch-Nyhan syndrome: describe the symptoms and causes of the disease.
- Scheme of conversion of cytidyl nucleotide by cleavage to ammonia.
- Compare the processes of splitting uracil and thymine.
- Name the sources of synthesis of purine bases of nucleotides.
- Give the scheme of the synthesis of purine nucleotides.

- Give the reactions of the biosynthesis of pyrimidine nucleotides.

- How are UTF and CTF formed?
- How is the synthesis of purine nucleotides regulated?
- What enzymes regulate the synthesis of pyrimidine nucleotides?
- How are thymidyl nucleotides formed?
- Explain the mechanism of conversion of ribonucleotides to deoxyribonucleotides.
- Explain the causes of Lesch-Nyhan disease.

#### Test tasks.

1. A 37-year-old man came to the therapist with complaints of periodic intense pain attacks in the joints of the big toe and their swelling. When analyzing urine, its sharply acidic character and pink color were established. Such changes in urine can be associated with the presence of what substances?

- A \* Uric acid salts
- B Chlorides
- C Ammonium salts
- D Calcium phosphate
- E Magnesium sulfate

2. For the treatment of gout, the patient was prescribed allopurinol, a structural analogue of hypoxanthine, which led to an increase in the excretion of hypoxanthine in the urine. What process is blocked at the same time?

- A \* Formation of uric acid
- B Spare path of synthesis of purine nucleotides
- C The main path of synthesis of purine nucleotides
- D Urea synthesis
- E Decay of pyrimidine nucleotides

3. A 48-year-old patient turned to the doctor with complaints of severe pain, swelling, redness in the joints, temperature rise to 38°C. A high content of urates was found in the blood. A possible cause of this condition may be a metabolic disorder:

- A \* Purinov
- B Collagen
- C Cholesterol
- D Pyrimidines
- E Carbohydrates

4. A 46-year-old patient turned to the doctor with a complaint of pain in the joints, which worsens on the eve of a change in weather. An increase in the concentration of uric acid was detected in the blood. Increased decay of which substance is the most likely cause of the disease?

- A \* AMP
- B TsMF
- C UTF
- D UMF
- E TMF

5. The patient has increased uric acid content in the blood, which is clinically confirmed by a pain syndrome due to the deposition of urates in the joints. As a result of which process is this acid formed?

- A \* Decay of purine nucleotides
- B Decay of pyrimidine nucleotides
- C Heme catabolism
- D Cleavage of proteins
- E Recycling of purine bases

6. Nitrous acid is formed in the body from nitrates, nitrites and nitrosamines, which causes the oxidative deamination of the nitrogenous bases of nucleotides. This can lead to a point mutation-substitution of cytosine to:

A.\* Uracil B.Guanine

C.Timin

D.Adenine

E.Inosine

7. A 37-year-old man came to the therapist with complaints of periodic intense pain attacks in the joints of the big toe and their swelling. When analyzing urine, its sharply acidic character and pink color were established. With the presence of which substances can be

A.\* Uric acid salts

**B**.Chlorides

**C.**Ammonium salts

**D.**Calcium phosphate

E.Magnesium sulfate

**8.** The patient's joints are enlarged and painful. The patient's blood has an elevated level of urates. What is this pathology called?

A.\*Gout

**B.**Rickets

C.Scurvy

**D.**Pellagra

**E.**Caries

**9.** The patient has pain in small joints, the joints are enlarged. There is an increased content of urates in the blood serum. The exchange of what substances

A.\* Purinov

**B.**Amino acids

**C.**Disaccharides

**D.**Pyrimidines

**E.**Glycerin

**10.** A patient with suspected gout was admitted to the clinic. What biochemical analysis should be prescribed to clarify the diagnosis?

A.\* Determination of uric acid in blood and urine

**B.**Determination of urea in blood and urine

C.Determination of creatine in the blood

**D**.Determination of uricase activity in blood

E.Determination of amino acids in blood

**11.** On the basis of laboratory analysis, the diagnosis of gout was confirmed in the patient. What analysis was carried out for the production

A.\* Determination of uric acid in blood and urine

**B.**Determination of creatinine in urine

C.Determination of residual nitrogen in the blood

**D**.Determination of urea in blood and urine

E.Determination of ammonia in urine

**12.** A 46-year-old patient turned to the doctor with a complaint of pain in the joints, which worsens on the eve of a change in weather. An increase in the concentration of uric acid was detected in the blood. Enhanced decay of which substance is the most likely cause

A.\*AMP B.TsMF C.UTF D.UMF

#### E.TMF

thirteen.An 8-year-old boy has Lesch-Nyhan disease. The concentration of uric acid in the blood is increased. Indicate the violation of which process is the cause of this hereditary disease.

A.\*Decomposition of purine nucleotides

**B.**Synthesis of purine nucleotides

C.Synthesis of pyrimidine nucleotides

**D**.Decay of pyrimidine nucleotides

E.Formation of deoxyribonucleotides

**14.**Methotrexate, a structural analogue of folic acid, is prescribed for the treatment of malignant tumors, which is a competitive inhibitor of dihydrofolate reductase and therefore suppresses

A.\* Nucleotides
B.Monosaccharides
C.Fatty acids
D.Glycerophosphatides
E.Glycogen
Pterin derivatives

15. Pterin derivatives - aminopterin and metatrexate - are competitive inhibitors of dihydrofolate reductase, as a result of which they inhibit the regeneration of tetrahydrofolic acid from dihydrofolate. These drugs lead to inhibition of the intermolecular transport of one-carbon groups. The biosynthesis of which nucleotide is inhibited in this case?

A.\* dTMF B.IMF C.UMF D.OMF E.AMP 16. The stri

16. The structural analogue of glutamine, the antibiotic azaserine, a powerful inhibitor of purine nucleotide synthesis, was prescribed to the patient for tumor chemotherapy. What type of inhibition is characteristic of this drug?

A.\* Competitive

**B.**Irreversible

C.Non-competitive

**D.**Uncompetitive

**E.**Allosteric

17. The child has delayed growth and mental development, a large amount of orotic acid is excreted in the urine. This hereditary disease develops as a result

A.\*Synthesis of pyrimidine nucleotides

**B.**Decay of pyrimidine nucleotides

C.Synthesis of purine nucleotides

D.Breakdown of purine nucleotides

E.Conversion of ribonucleotides into deoxyribonucleotides

18. A 58-year-old man underwent surgery for prostate cancer. After 3 months, he underwent a course of radiation and chemotherapy. The complex of medicines included 5-fluorodeoxyuridine - a thymidylate synthase inhibitor. The synthesis of which substance is primarily blocked under the action of this drug?

A.+ DNA B.i-RNA C.p-RNA D.t-RNA E.Squirrel

19. For the treatment of malignant tumors, methotrexate is prescribed, a structural analog of folic acid, which is a competitive inhibitor of dihydrofolate reductase and therefore suppresses

A.\* Nucleotides
B.MonosaccharidesC.Fatty acidsD.Glycerophosphatides

E.Glycogen

20. Methotrexate, a structural analogue of folic acid, is prescribed for the treatment of malignant tumors, which is a competitive inhibitor of dihydrofolate reductase and therefore suppresses the synthesis of nucleic acids at the level of:

A.\*Synthesis of mononucleotides
B.Replications
C.Transcriptions
D.Reparations
E.Processing

21. With hereditary orataciduria, the excretion of orotic acid is many times higher than the norm. The synthesis of which substances will be disturbed in this pathology?

**A.**\*Pyrimidine nucleotides **B.**Purine nucleotides

C.Biogenic amines D.Uric acid E.Urea

4. Summing up.

5. List of recommended literature (main, additional, electronic information resources):

## Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

## Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №16, 17, 18

Topic: <u>Biosynthesis of nucleic acids. Molecular mechanisms of DNA replication. Stages of synthesis of daughter chains of DNA molecules. Molecular mechanisms of transcription.</u> <u>Stages and enzymes of RNA synthesis. Processing - post-transcriptional modification of RNA.</u> Antibiotics-inhibitors of transcription. Biosynthesis of proteins in ribosomes. Genetic code: triplet code structure, its properties. Post-translational modification of peptide chains. Broadcast regulation. Regulation of gene expression. Mechanisms of DNA mutations and repairs.

**Goal:** <u>Studying the mechanisms of DNA and RNA synthesis, studying the processes of protein synthesis from amino acids, translation of information from the language of the genetic code, regulation of translation and post-translational transformations of peptide chains. Get information about the possibility of gene expression, the mechanisms of mutations and the formation of recombinant DNA.</u>

Basic concepts:replication, repair, transcription, splicing, keeping,processing,genetic code, translation, regulation of gene expression, mutations, recombinant DNA.Equipment:Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- structural differences between DNA and RNA;
- the role of DNA and RNA in the storage and transfer of genetic information;
- stages of synthesis of daughter chains of the DNA molecule;
- mechanisms of matrix (programmed) RNA biosynthesis;
- antibiotics capable of inhibiting the transcription process;
- processes of post-transcriptional modification of RNA;

- mechanisms of matrix (programmed) biosynthesis of nucleic acids, as the basis for

understanding matrix biosynthesis of proteins;

- mechanisms of matrix synthesis of proteins and its regulation;

- effects of physiologically active compounds on protein synthesis mechanisms.

The higher education applicant should be able to:

- -explain the meaning of using the genetic code to encode amino acids;
- -explain the stages of broadcasting;

-Give examples of the influence of physiologically active compounds on the stages of protein synthesis.

Questions to check basic knowledge on the topic of the class:

- What nitrogenous bases (as part of nucleic acids) do you know?
- What carbohydrates are part of DNA, RNA?
- Localization of RNA and DNA in cells.
- What is the difference between DNA and RNA?
- Define complementarity.
- Name the types of RNA.
- What are the functions of DNA and RNA in the body?
- -Ribosomal protein-synthesizing system
- Aminoacyl-t-RNA synthetases.
- Genetic code, its properties.

-Explain the meaning of the terms: transcription, translation, codon, anticodon.

-Explain the stages of biosynthesis: recognition, initiation, elongation, termination, informosomes.

- Formation of higher levels of the protein structure.
- Molecular mechanisms of protein biosynthesis Stages of translation.
- How is protein biosynthesis regulated?

- Functional organization of a gene: what is a gene-regulator, gene-operator, structural gene, cistron, operon?

-Mechanisms of protein biosynthesis regulation.

-Violation of protein biosynthesis. Molecular mutations as the primary source of genetic changes.

- Influence of physiologically active compounds on translation processes.

- Antibiotics - translation inhibitors, their biomedical application.

3. Formation of professional skills and abilities.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

-What is the role of nucleic acids (DNA, RNA) in protein biosynthesis (general provisions)?

-What is meant by matrix biosynthesis of nucleic acids and proteins?

-Where does the synthesis of nucleic acids take place?

-From what and how does DNA biosynthesis occur? Name the main stages, draw a general scheme of DNA biosynthesis; possible options.

-Molecular mechanisms of DNA reduplication.

-What regularity underlies the structure of copies of polynucleotide chains?

-What do DNA replication errors lead to?

-From what and how does RNA biosynthesis occur? Name the main stages, possible variants of biosynthesis: state the general scheme of RNA biosynthesis.

-The process of "maturation" of RNA (splicing, processing). The role of enzymes in this process (nucleases, RNA ligases).

-Describe the functions of the most important groups of nucleases (DNAases,

exodeoxyribonucleases, polynucleotide phosphorylases, restriction enzymes, etc.).

-What regularity underlies the structure of copies of polynucleotide chains?

-How is protein biosynthesis induced?

-How is repression of protein biosynthesis carried out?

-Violation of protein biosynthesis: what are enzymopathies, hemoglobinopathies, hemoglobinoses?

-Hereditary diseases, as one of the manifestations of biochemical polymorphism, ideas about the biochemical mechanisms of hereditary molecular diseases (examples).

- Name the causes of: phenylpyruvic oligophrenia (phenylketonuria), alkaptonuria, galactosemia, albinism.

- Name antibiotics - inhibitors of protein synthesis in prokaryotes, determine their role in the treatment of infections.

- What is the role of diphtheria toxin in inhibition of protein synthesis?

- Individual features of the antigenic composition of organisms as the basis of tissue (transplantation) incompatibility.

- Regulation of protein biosynthesis.

-Influence of physiologically active compounds on translation processes.

- Antibiotics - translation inhibitors, their biomedical application.

- Differential activity of genes as a mechanism of cell differentiation in ontogenesis.

- Molecular mutations as the primary source of genetic variability.

- Biochemical polymorphism of proteins (on examples of hemoglobins and isozymes).

- What are mutations?

- What types of mutations can you name?

- Gene, genomic, chromosomal mutations.

- Mutagens.

- Mechanisms of DNA repair.

- Recombinant DNA.

- Genetic engineering.

Test tasks.

1.In experimental studies, it was established that steroid hormones affect proteosynthesis. Indicate which stage of this process they influence.

A.\*Synthesis of specific m-RNAs.

**B.**Synthesis of ATP.

**C.**Synthesis of specific t-RNAs.

**D.**GTP synthesis.

**E.**Synthesis of specific r-RNAs.

2. The blood of the child and the alleged father was submitted for forensic examination to establish paternity. Specify which chemical components must be identified in the test blood.

A.\* DNA. B.t-RNA. C.p-RNA.

**D.**m-RNA.

**E.**my-RNA.

3. In patients with xeroderma pigmentosum, the skin is extremely sensitive to sunlight, skin cancer may develop. The reason is a hereditary deficiency of the enzyme UV-endonuclease. As a result of this defect, the process is disrupted:

A.\*DNA repairs
B.DNA replication
C.Transcriptions
D.Reverse transcription
E.Broadcasts

4. In the experiment, it was shown that ultraviolet-irradiated skin cells of patients with xeroderma pigmentosum restore the native structure of DNA more slowly than cells of normal people due to a defect in the repair enzyme. Choose the enzyme of this process.

A.\* EndonucleaseB.RNA ligaseC.PrimazaD.DNA polymerase Sh

**E.**DNA gyrase

5.The antibiotic rifomycin, which is used in the treatment of tuberculosis, affects certain biochemical processes. Name them.

A.\*Inhibits RNA polymerase at the initiation stage

**B.**Inhibits DNA polymerase at the initiation stage

C.Inhibits DNA ligase

D.Inhibits aminoacyl RNA synthetase

6. Quinolones are used to treat urogenital infections - DNA gyrase enzyme inhibitors. Indicate which process is disrupted by quinolones in the first place.

A.\*replication

**B.**reparation

C.gene amplification

**D**.recombination of genes

E.reverse transcription

7. A 58-year-old man underwent surgery for prostate cancer. After 3 months, he underwent a course of radiation and chemotherapy. The complex of medicines included 5-fluorodeoxyuridine - a thymidylate synthase inhibitor. The synthesis of which substance is primarily blocked under the action of this drug?

A.DNA

B.i-RNA C.p-RNA D.t-RNA E.Squirrel

8. When poisoned with amanitin - the poison of the pale toadstool, RNA polymerase B(II) is blocked. At the same time, it stops:

A.\*Synthesis of mRNA

**B.**Synthesis of tRNA

C.Reverse transcription

**D.**Synthesis of primers

**E.**Maturation of mRNA.

9. Patients with xeroderma pigmentosum are characterized by an abnormally high sensitivity to ultraviolet light, the result of which is skin cancer, due to the inability of enzyme systems to restore damage to the hereditary apparatus of cells. What process is this pathology associated with?

A.\* DNA repair

**B.**Gene conversion

C.DNA recombination

**D**.Gene complementation

**E.**DNA reduplication

10. A 40-year-old woman was hospitalized in serious condition with symptoms of Amanita phalloides (pale toadstool) poisoning. It is known that one of the toxins of these mushrooms blocks the synthesis of mRNA precursors. This toxin:

A.\* amanitinB.actinomycinC.taurineD.ribophorinE.bicuculline

11. A 57-year-old patient diagnosed with viral hepatitis was prescribed interferon. The antiviral effect of this drug is based on:

A - \*Suppression of the translation of virus envelope proteins

- B Inhibition of viral DNA replication
- C Activation of the complement system
- D Formation of antiviral antibodies
- E Inhibition of the synthesis of transforming growth factors.

12. For the formation of a transport form of amino acids for protein synthesis on ribosomes, it is necessary:

A - \*Aminoacyl-tRNA synthetase

In - GTF

C - m-RNA

- D ribosome
- E revertase

13. The transport form of AK for protein synthesis on ribosomes acts as:

A - \*aa tRNA

B - Aminoaceladenylate

 $C-S\mbox{-}adenosylmethionine$ 

D - phosphoadenosine phosphosulfate (FAPS)

E-Aminoacyladenylate

14. The intensity of gene expression is controlled by a developed system of transcription regulation signals. Effective activating elements of such a system are specific DNA sequences, which are called:

A - \*Enhancers

- **B** Silencers
- C Repressors
- D Operators
- E-Inductors

15. The human genetic apparatus contains about 30,000 genes, and the number of antibody variants reaches millions. What mechanism is used for the formation of new genes responsible for the synthesis of such a large number of antibodies?

A - \* Gene recombination

- B Amplification of genes
- C DNA replication
- D DNA repair
- E Formation of Okazaki fragments

16. Development of the methodin viewandlenience geneandinandwith'isdnaeath in new ones comboandnationalandyah became new bandOhandmandwe do achievement genetic exandgin.For with'isdna two chain DNA,asandviewandflaxand andwith pandof them bodyandzmandin, eath processed:

- 2. A \* restriction endonuclease
- 3. B lyase
- 4. C helicase
- 5. D transferase
- 6. E synthetase.

4. Pipounding pandbagandin:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

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7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

## Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

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- 2. http://libblog.odmu.edu.ua/

3. https://moodle.odmu.edu.ua/login/index.php

 Practical class № 19,20

 Topic:
 Intermediate control for the semester

 Goal:
 Determine the level of assimilation of knowledge for the semester

 Equipment:
 Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. List of questions for preparation for control:

1. Enzymes: definition; properties of enzymes as biological catalysts.

2. Classification and nomenclature of enzymes, characteristics of individual classes of enzymes.

3. Structure and mechanisms of action of enzymes. Active and allosteric (regulatory) centers.

4. Cofactors and coenzymes. The structure and properties of coenzymes, vitamins as precursors in the biosynthesis of coenzymes. Coenzymes: The types of reactions catalyzed by particular classes of coenzymes.

5. Isoenzymes, features of structure and functioning, importance in diagnosis of diseases.

6. Mechanisms of action and kinetics of enzymatic reactions: dependence of the reaction rate on substrate concentration, pH, and temperature. Principles and methods of detecting enzymes in biological objects. Units of activity and amount of enzymes.

7. Enzyme activators and inhibitors: examples and mechanisms of action.

8. Types of enzyme inhibition: reverse (competitive, non-competitive) and irreversible inhibition.

9. Regulation of enzymatic processes. Ways and mechanisms of regulation: allosteric enzymes; covalent modification of enzymes. Cyclic nucleotides (cAMP, cGMP) as regulators of enzymatic reactions and biological functions of the cell.

10. Enzymopathies - congenital (hereditary) defects in the metabolism of carbohydrates, amino acids, porphyrins, and purines.

11. Enzyme diagnosis of pathological processes and diseases.

12. Enzymotherapy - use of enzymes, their activators and inhibitors in medicine.

13. General characteristics of energy metabolism. Metabolism: catabolism, anabolism and amphibolic pathways.General and specific pathways of catabolism. Stages of intercellular catabolism of biomolecules: proteins, carbohydrates, lipids.

14. The Citric Acid Cycle (CAC). Location in the cell. Enzymes of CAC. Importance of CAC. Bioenergetics of Citric Acid Cycle. Physiologic importance of CAC.

15. Reactions of biologic oxidation. Types of reactions: dehydrogenase, oxidase, oxygenase reactions and their biologic importance. Chemical nature of dehydrogenases. Role of coenzymes in the functions of enzymes.

16. Tissue respiration.Flavoenzymes. Chemical nature and biochemical role.Cytochromes. Cytochrome oxidase, its chemical nature, the biochemical role and mechanism of participation in the respiratory chain.

17. Oxidative phosphorylation: coupling points of electron transport and phosphorylation, coefficient of oxidative phosphorylation.

18. The chemiosmotic theory of oxidative phosphorylation. ATP synthase of mitochondria. Inhibitors of electron transport and uncouplers of oxidative phosphorylation.

19. Microsomal oxidation. Cytochrome P450 and molecular organization of the electron transport chain. Detoxification of xenobiotics and formation of biological active compounds.

20. Aerobic and anaerobic oxidation of glucose, general characteristics of the processes.

21. Anaerobic oxidation of carbohydrates. Glycolysis.

22. Oxidative-reduction steps of anaerobic glycolysis, its importance. Shuttle mechanism for the transfer of reducing equivalents of NADH.

23. Aerobic oxidation of glucose. The sequence of reactions and enzymes of glycolysis.Oxidative decarboxylation of pyruvate. Enzymes, coenzymes and the sequence of reactions in a multienzyme complex.

24. The comparison of bioenergetics of aerobic and anaerobic oxidation of glucose and glycogen. Pasteur's effect.

25. Pentose phosphate pathway of carbohydrates metabolism.Regulation of glycogen phosphorylase activity.

26. Biosynthesis and catabolism of glycogen in the liver. Regulation of glycogen synthase activity.

27. Mechanism of reciprocal regulation of glycogenolysis and glucogenesis by cAMP dependent cascade phosphorylation of enzymes. The role of adrenaline, glucagon and insulin in hormonal regulation of glycogen metabolism in muscles and liver.

28. Genetic disorders of glycogen metabolism (glycogenosis, aglycogenosis).

29. Gluconeogenesis. Mechanism in the different tissues and organs. Corey's cycle and the glucose alanine cycle.

30. Blood glucose (glucosemia): normoglycemia, hypo- and hyperglycemia, glucosuria. Diabetes mellitus is a pathology of glucose metabolism. Hormonal regulation of blood glucose concentration and metabolism.

31. Pentose phosphate pathway of glucose oxidation: scheme of the process and biological significance. Metabolic ways of conversion of fructose and galactose; hereditary enzymopathies of their metabolism.

32. Catabolism of triacylglycerols in adipocytes of adipose tissue: sequence of reactions, mechanisms of regulation of triglyceride lipase activity. Neurohumoral regulation of lipolysis with the participation of adrenaline, norepinephrine, glucagon and insulin).

33. Fatty acid oxidation reactions ( $\beta$ -oxidation); the role of carnitine in the transport of fatty acids in mitochondria. Energy value of in-oxidation of fatty acids in cells.

34. Glycerol oxidation: enzymatic reactions, bioenergetics.

35. Ketone bodies. Reactions of biosynthesis and disposal of ketone bodies, physiological significance. Violation of the metabolism of ketone bodies under pathological conditions (diabetes mellitus, starvation).

36. Biosynthesis of higher fatty acids: reactions of biosynthesis of saturated fatty acids (palmitate) and regulation of the process. Biosynthesis of mono- and polyunsaturated fatty acids in the human body.

37. Biosynthesis of triacylglycerols and phosphoglycerides. Metabolism of sphingolipids. Genetic abnormalities of sphingolipid metabolism - sphingolipidosis.

38. Cholesterol biosynthesis: scheme of reactions, regulation of cholesterol synthesis. Ways of biotransformation of cholesterol: esterification; formation of bile acids, steroid hormones, vitamin D3.

39. Pathologies of lipid metabolism: atherosclerosis, obesity, diabetes.

40. Circulatory transport and deposition of lipids in adipose tissue. Endothelial lipoprotein lipase. Blood plasma lipoproteins: lipid and protein (apoprotein) composition. Hyperlipoproteinemia.

41. The pool of the amino acids in the body. Routes for transport and utilization of amino acids in tissues. Glucogenic and ketogenic amino acids.

42. Direct and indirect deamination of free L-amino acids in tissues.

43. Transamination of amino acids: reactions and their biochemical significance, mechanisms of action of aminotransferases.

44. Decarboxylation of L-amino acids in the human body. Physiological value of the products formed. Oxidation of biogenic amines.

45. Ways of formation and neutralization of ammonia in the body. Urea biosynthesis: the sequence of enzymatic reactions of biosynthesis, genetic anomalies of urea cycle enzymes.

46. Biosynthesis and biological role of creatine and creatine phosphate. Glutathione: structure, biosynthesis and biological functions of glutathione.

47. Specialized ways of metabolism of cyclic amino acids - phenylalanine and tyrosine. Hereditary enzymopathies of the exchange of cyclic amino acids - phenylalanine and tyrosine. Metabolism of the cyclic amino acid tryptophan and its hereditary enzymopathies.

48. Nitrogen bases, nucleosides and nucleotides as the composite components of the nucleic acids. Minor nitrogen bases and nucleotides.Free nucleotides: ATP, NAD+, NADP+, FAD, FMN, CTP, UTP, 3',5'-cAMP, 3',5'-cGMP, their biochemical functions.

49. Nucleic acids. General characteristics of DNA and RNA, their biological importance in the storage and the transfer of genetic information.Features of DNA and RNA primary structure. Chemical bonds, which are responsible for the formation of nucleic acids primary structure.

50. Secondary structure of DNA, role of hydrogen bonds in its formation (Chargaff's rules, Watson-Crick model), anti-parallelity of strands.Tertiary structure of DNA. Physical and chemical properties of DNA, interaction with cation ligands, formation of nucleosomes.

51. Molecular organization of nuclear chromatin of eukaryotes; nucleosome organization, histone and non-histone proteins. Nucleoproteins: structure, biological functions.

52. Structure, properties and biological functions of RNA. Types of RNA: m-RNA, t-RNA, r-RNA. Features of the different type of RNA structural organization.

53. Biosynthesis of purine nucleotides; scheme of IMP synthesis reactions.Formation of AMP and GMP from IMP, mechanisms of regulation.

54. Biosynthesis of pyrimidine nucleotides; scheme of reactions, regulation of synthesis. Biosynthesis of deoxyribonucleotides. Formation of the thymidine nucleotides. Inhibitors of TMP synthesis as anti-cancer medicines.Catabolism of purine nucleotides, hereditary disturbances of the uric acid metabolism.

55. Replication of DNA, its biological importance, and semiconservative mechanism of replication.

56. RNA transcription: prokaryotes and eukaryotes RNA-polymerases, signals of transcription: promoter, initiator and terminator fragments of genome. Processing and post-translational modification of synthesized RNA.

57. Genetic (biologic) code, triplet structure and properties. Transport RNA and transportation of amino acids. Amino acyl-tRNA-synthetases. Steps and mechanism of translation (protein synthesis) in ribosomes: initiation, elongation and termination.

58. Post-translational modification of peptide chains. Regulation of translation.Inhibitors of transcription and translation in prokaryotes and eukaryotes. Antibiotics and interferons, they use in medicine. Diphtheria toxin.

59. Regulation of prokaryote gene expression: regulatory and structural fragments of lactose, Lac-operon, gene regulator, promoter, operator.

60. Gene engineering: construction of recombinant DNA, gene cloning. Genetic engineering of enzymes, hormones, interferons, etc.

61. Mutations: genome, chromosome, gene. Mechanisms of mutagen activity; role of the induced mutations in the origin of the enzymopathology and hereditary human diseases.Biological importance and mechanisms of DNA reparations. Reparation of UV-induced gene mutations: xeroderma pigmentosum.

3. Summing up.

4. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

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6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

#### Practical class №21

# Topic: Hormones general concept. Classification, mechanisms of action of hormones on target cells. Hormones of the hypothalamus and pituitary gland hormones.

**Goal:**To study and be able to characterize the hormones of the hypothalamic-pituitary system according to the following plan: 1) the name of the hormone; 2) place of synthesis; 3) features of the structure; 4) mechanism of action, biological role; 5) violation of synthesis.

**Basic concepts:**hormones, classification of hormones, protein nature of hormones, the mechanism of action of hormones depends on their nature.

Equipment: Laboratory of the department

#### Plan:

1. Organizational measures (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know :

- General ideas about hormones and other signaling molecules;

- Properties of hormones and features of endocrine system functioning;

- Classification of hormones: 1) by the city of synthesis, 2) by chemical nature, 3) by ensuring and maintaining homeostasis, 4) by primary contact with the cell.

- To study the concept of receptors, their structure, localization and interaction with hormones The higher education applicant should be able to:

- hormones of the hypothalamic-pituitary system
- the name of the hormone
- place of synthesis
- features of the structure
- mechanism of action, biological role
- violation of synthesis

Questions to check basic knowledge on the topic of the class:

- General characteristics of hormones, the role of hormones in the system of intercellular integration of human body functions.

- Hormone research methods.

- Properties of hormones and features of endocrine system functioning.

- Classification of hormones by the city of synthesis, chemical nature, provision and maintenance of homeostasis.

- Name groups of hormones and representatives of each of them.

- Mechanisms of action of hormones of protein-peptide nature and derivatives of amino acids.

- Biochemical systems of intracellular transmission of hormonal signals: G-proteins, secondary messengers (cAMP, cGMP, Ca2+/calmodulin, IF3, DAH).

- Adenylate cyclase messenger system. Structure of ATP and cyclic 3',5'-AMP.

- Hormones of the hypothalamus - liberins and statins. Their structure and role in neurohumoral regulation.

- Hormones of the anterior lobe of the pituitary gland. Pathological processes associated with a violation of their synthesis.

- Group "growth hormone (somatotropin) - prolactin - chorionic somatomammotropin"; pathological processes associated with a violation of their functions.

- A group of glycoproteins - tropic hormones of the pituitary gland (thyrotropin, gonadotropins).

- Vasopressin and oxytocin: structure, biological functions. Pathology associated with a violation of vasopressin production.

3. Formation of professional skills:

3.1 Demonstration and practical work:

Study of the nature of hormones using the biuret reaction.

Recommendations for performing tasks.

Principle of the method: discovery of peptide bonds in proteins and peptides. These substances form a red-violet complex with copper sulfate in an alkaline environment.

Procedure: Pour 0.5 ml of the test solution into a test tube, add 0.5 ml of 10% NaOH solution and 1-2 drops of 1% CuSO4 solution and mix. In the presence of protein, a red-purple color appears.

Conclusion: the appearance of a red-violet color indicates the presence of peptide bonds in the enzyme molecule, that is, its protein nature.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Test tasks.

What hormone stimulates the activity of the enzyme adenylate cyclase?

- A. Adrenaline
- B. Aldosterone
- S. Testosterone
- D. Progesterone
- E. Calcitriol

In which glands are steroid hormones synthesized?

A. Shield-like

V. Pidzhudzkova

S. Adrenal cortex

D. The cerebral part of the adrenal glands

What hormone regulates the function of the thyroid gland?

- A. Tyroliberin
- V. Transkortin
- S. Cortisol

#### D. Somatoliberin

E. Somatotropin

What hormone regulates water balance and osmotic pressure of blood plasma, stimulates contraction of smooth muscles?

A. Prolactin

B. Somatostatin

S. Corticoliberin

- D. Vasopressin
- E. Glucagon

For the purpose of analgesia, a substance that imitates the effects of morphine, but is synthesized in the central nervous system, can be used. Name this substance.

A. Somatoliberin

B. Oxytocin

S. Vasopressin

- D. Calcitonin
- E. Endorphin

The patient has a headache, changes in appearance (increasing the size of the limbs, brow ridges, nose, tongue), hoarse voice, memory impairment. The disease started about three years ago. The reason for this condition can be:

A. Aldosterone deficiency

B. Glucagon deficiency

S. Thyroxine deficiency

D. Hyperproduction of somatotropin

E. Hyperproduction of corticosteroids

Products of hydrolysis and modification of some proteins are biologically active substances - hormones. Lipotropin, corticotropin, melanotropin and endorphins are formed from which protein in the pituitary gland?

A. Proopiomelanocortin

B. Neuroalbumin

- S. Neurostromin
- D. Neuroglobulin
- E. Thyroglobulin

Products of hydrolysis and modification of some proteins are biologically active substances - hormones. Lipotropin, corticotropin, melanotropin and endorphins are formed from which protein in the pituitary gland?

A. Proopiomelanocortin

- B. Neuroalbumin
- S. Neurostromin

D. Neuroglobulin

E. Thyroglobulin

Ca2+ ions play the role of secondary messengers in cells. They are activators of a number of processes if they interact with:

A. Calcitonin

V. Calmodulin

S. Calciferol

D. Myosin kinase

E. Phosphorylase S

After a cerebral hemorrhage with damage to the hypothalamic nuclei, the patient developed diabetes insipidus, which is accompanied by polyuria as a result of:

A. Hypoglycemia

B. Reduction of reabsorption of potassium ions

S. Acceleration of glomerular filtration

D. Hyperglycemia E. Reduction of water reabsorption

Which of the listed hormones is hydrophilic and does not require a special transport protein: A. Dihydrotestosterone

B. Progesterone

S. Paratyrin

D. Aldosterone

E. Estradiol

The husband was diagnosed with angina pectoris. A phosphodiesterase inhibitor is included in the complex of drugs prescribed to the patient. The concentration of which substance in the heart muscle will increase?

A. AMF

V. GMF

S. cAMP

D. ADP

E. ATP

The patient turned to the doctor with complaints of frequent and excessive urination, thirst. Urinalysis revealed: daily urine output -19 L, urine density -1.001. For which disease are these indicators characteristic?

A. Steroid diabetes

B. Diabetes mellitus

S. Thyrotoxicosis

D. Diabetes insipidus

E. Addison's disease

The boy is being examined in the hospital for short stature. Over the past two years, it has grown by only 3 cm. This condition is caused by the lack of which hormone?

A. Somatotropin

B. Corticotropin

S. Gonadotropin

D. Thyrotropin

E. Parathyroid hormone

4. Summary.

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

## Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

#### Practical class №22

Topic: Thyroid and parathyroid glands hormones. Structure and synthesis of thyroid hormones. Thyroid gland pathology. Regulation of phosphorus-calcium metabolism. Metabolic disorders of calcium homeostasis.

Goal: Thyroid hormones, iodothyronine and calcitonin, dysfunction of the thyroid gland, calcitonin, parathormone.

**Basic concepts:**thyroid gland, regulation of phosphorus-calcium metabolism, iodothyronine and calcitonin.

**Equipment:**Laboratory of the department

Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know :

- Hormones of the anterior lobe of the pituitary gland. Pathological processes associated with a violation of their synthesis.

- Thyroid hormones: iodothyronine and calcitonin. The main types of dysfunction of the thyroid gland.

- Hormones of the parathyroid glands. The relationship between parathyrin and calcitonin in the regulation of phosphorus-calcium metabolism. Signs of parathyroid gland dysfunction.

The higher education applicant should be able to:

- Determine the type of diabetes

- The role of thyroid glands

- Hypothyroidism and thyroiditis

Questions to check basic knowledge on the topic of the class:

- thyroid hormones

- names of hormones
- mechanism of synthesis
- features of the structure

- mechanism of action, biological role

- violation of the synthesis of thyroid hormones
- dysfunction of the thyroid gland
- parathyroid hormones

3. Formation of professional skills and abilities.

Demonstration and practical work "Determination of iodine in the thyroid gland".

Recommendations for completing tasks:

The method is based on the separation during acid hydrolysis of thyroid hormones (iodothyronines) of iodide acid, which reacts with potassium iodate to release free iodine:

 $5NH + KJO3 + HNO3 \rightarrow 3J2 + KNO3 + 3H2O$ 

In chloroform, iodine has a purple color.

**Progress.** Place several crystals of thyroidin in a test tube, add 10 drops of concentrated nitric acid and heat for 3-5 minutes in a water bath. Then add 20 drops of 10% potassium iodate solution. Mix the contents and cool. Add 15 drops of chloroform to the test tube, mix by shaking. Color development is observed.

Des	ign	ing	the	work	:fill	in	the	tabl	e
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Hormones	Place of synthesis	Chemical structure	Qualitative response	Reaction mechanism	Color
Insulin					
Iodothyronine					

In clinical and biological laboratories, methods of qualitative and quantitative analysis are widely used to determine hormones in biological material for the purpose of diagnosis and prognosis of various endocrine diseases.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Test tasks.

What hormones regulate the content of calcium and phosphorus in the blood?

- A. Parathyroid hormone
- B. Calcitonin

S. Adrenocorticotropin

D. Progesterone

E. Testosterone

What hormone stimulates glycogen biosynthesis and enhances anabolic processes?

A. Adrenaline

B. Norepinephrine

S. Cholecystokinin

Violation of hormone synthesis in the body leads to various pathological conditions. What hormone is synthesized in an insufficient amount in the body when growth stops in childhood, but at the same time mental activity is not disturbed?

A. Thyroxine

B. Prolactin

S. Somatotropin

D. Adrenaline

E. Gonadotropin

A 9-year-old boy had multiple fractures of his limbs due to the fragility of his bones. The function of which endocrine organ is impaired?

A. Parathyroid glands

B. Thyroid gland

S. Epiphysis of the adrenal glands

E. Pancreas

Some substances contribute to the uncoupling of respiration and phosphorylation processes, thereby causing the activation of Na+, K+-ATP-ase. Name this substance.

- A. Vasopressin
- B. Insulin
- S. Oxytocin
- D. Adrenaline
- E. Thyroxine
- D. Insulin
- E. Thyroxine

3. The patient has tachycardia, increased body temperature, weight loss, irritability. An increase in the content of which hormone in the blood can lead to such a condition?

A. Thyroxine

B. Vasopressin

S. Somatotropin

D. Insulin

E. Adrenocorticotropin

It is known that endemic goiter is widespread in some biogeochemical zones. Deficiency of which biogenic element is the cause of this disease.

A. Zaliza

V. Yodu

S. Tsink

D. Midi

E. Cobalt

The patient's blood shows hypercalcemia, hypophosphatemia, and hyperphosphaturia in the urine. What is the possible cause of this condition?

A. Enhanced secretion of parathyroid hormone

B. Inhibition of parathyroid hormone secretion

S. Enhanced secretion of calcitonin

D. Inhibition of calcitonin secretion

E. Enhanced secretion of thyroxine

A patient living in a specific geochemical area has been diagnosed with endemic goiter. What type of post-translational modification of thyroglobulin is disturbed in the patient's body?

A. Methylation

B. Acetylation

S. Phosphorylation

D. Glycosylation

E. Iodination

With hyperfunction of the thyroid gland, there is a decrease in weight and an increase in body temperature. What biochemical processes are activated at the same time?

A. Anabolism

B. Catabolism

S. Gluconeogenesis

D. Lipogenesis

E. Steroidogenesis

Parathyroid glands were mistakenly removed during thyroid surgery in a patient with diffuse toxic goiter. Convulsions, tetany occurred. The exchange of which bioelement is disturbed?

A. Calcium

B. Magnesium

S. Kaliya

D. Iron

E. Sodium

4.Summary:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

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## Practical class №23

# Topic:Steroid hormones. Hormones of the adrenal cortex and gonads. Their structure and biochemical mechanisms of action.

Goal:characterize hormones of steroid nature, mechanism of action, structures of hormones, biological role

**Basic concepts:** steroid hormones, nomenclature, classification, synthesis scheme, hormones of the adrenal cortex (C21-steroids) - cortisol, corticosterone, aldosterone, biochemical effects of corticosteroids.

**Equipment:** Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know :

- adrenal cortex hormones

- glucocorticoids

- Itsenko-Cushing syndrome

- the role of aldosterone in the regulation of water-salt metabolism; aldosteronism

- Female sex hormones: estrogens - estradiol, progesterone physiological and biochemical effects, regulation of synthesis and secretion

- Male sex hormones (androgens) – testosterone, dihydrotestosterone, regulation of synthesis and secretion

Questions to check basic knowledge on the topic of the class:

- Mechanism of action of steroid hormones

- Structural formulas of steroid hormones

- Steroid hormones of the adrenal cortex (C21-steroids) – glucocorticoids and mineralocorticoids; structure, properties.

- Female sex hormones: estrogens, progesterone. Physiological and biochemical effects; connection with the phases of the ovulatory cycle

- Male sex hormones (C19-steroids). Physiological and biochemical effects of androgens; regulation of synthesis and secretion.

- Catecholamines (adrenaline, norepinephrine, dopamine): structure, biosynthesis, physiological effects, biochemical mechanisms of action.

3. Formation of professional skills and abilities.

3.1. Refractometric determination of protein in blood serum

Work progress: At the beginning, the refractive index of distilled water is determined, with the correct setting of the device, the refractive index is 1.3330. To do this, open the chamber and apply 1-2 drops of water to the lower half of the prism so that it completely wets the chamber. The camera is closed and the sharpness of the eyepiece is set so that the field of vision and sight lines are clearly visible. If the chiaroscuro boundary is blurred, clarity is achieved by rotating the dispersion limb screw. The chiaroscuro boundary is set at the point of intersection of the scale with the three sighting lines of the eyepiece. Then lift the upper half of the chamber, wipe both prisms and apply 2 drops of blood serum. At the same time, the boundary of chiaroscuro shifts, the sight lines of the eyepiece are set on it, and the refractive index is taken on the reference scale.

Refractive	Protein	Refractive	Protein
index	in blood serum	index	in blood serum
	1n %		1n %
1.33705	0.63	1.34575	3.68
1.33743	0.86	1.34612	5.90
1.33781	1.08	1.34650	6,12
1.33820	1.30	1.34687	6.34
1.33858	1.52	1.34724	6.55
1.33896	1.74	1.34761	6.77
1.33934	1.96	1.34798	6.98
1.33972	2.18	1.34836	7.20
1.34000	2.40	1.34873	7.42
1.34048	2.62	1.34910	7.63
1.34086	2.84	1.34947	7.85
1.34124	3.06	1.34984	8.06
1.34162	3.28	1.35021	8.28
1.34199	3.50	1.35058	8.49
1.34237	3.72	1.35095	8.71
1.34275	3.94	1.35132	8.92
1.34313	4.16	1.35169	9,14
1.34350	4.38	1.35205	9.35

Calculation of protein percentage by refractive index:

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions

Control materials for the final stage of the class:

Questions to check the final level of knowledge

- Mechanism of action of estrogens
- Biological significance of steroid hormones
- Mechanism of action of androgens
- Synthesis of catecholamines
- Biological value of aldosterone, cortisol
- Glucocorticoids
- Structural formulas of steroid hormones

Test tasks

Arachidonic acid as an irreplaceable component of food is a precursor of biologically active substances. What compounds are synthesized from it?

A. Ethanolamine

V. Kholin

S. Norepinephrine

D. Prostaglandin E1

E. Triiodothyronine

A patient complained of constant thirst to the doctor. Hyperglycemia, polyuria, and increased content of 17-ketosteroids in urine were established. What disease is possible?

A. Steroid diabetes

B. Non-insulin-dependent diabetes

S. Myksedema

D. Glycogenosis of type I

E. Addison's disease

Hyperglycemia occurs in Itsenko-Cushing's disease. What process is stimulated in this case?

A. Glycolysis

B. Phosphorolysis of glycogen

S. Krebs cycle

D. Pentose phosphate pathway of glucose oxidation

E. Gluconeogenesis

What hormone stimulates the synthesis of corticosteroids?

A. Parathyroid hormone

B. Thyrotropin

S. Corticoliberin

D. Calcitonin

E. Corticosterone

The patient was found to have hypernatremia, hypervolemia, and hypokalemia. What is the possible cause of this condition?

A. Hyperaldosteronism

B. Hypoaldosteronism

S. Addison's disease

D. Based's disease

E. Diabetes

Taking oral contraceptives containing sex hormones inhibits the secretion of pituitary hormones. The secretion of which of the following hormones is inhibited at the same time?

A. Vasopressin

B. Somatotropin

S. Oxytocin

D. Follitropin

#### E. Corticotropin

Testosterone and its analogues increase the mass of skeletal muscles, which allows them to be used for the treatment of dystrophies. This action of the hormone is determined by the interaction with which cell substrate?

A. Membrane receptors

B. Ribosomes

S. Nuclear receptors

D. Transcription activator proteins

E. Chromatin

The patient, who has been taking glucocorticoids for a long time, had an exacerbation of the disease, a decrease in blood pressure, and weakness as a result of the withdrawal of the drug. What explains this?

A. Occurrence of adrenal insufficiency

B. Cumulation

C. Addiction to the drug

D. Hyperproduction of ACTH

E. Sensitization

The patient has been taking glucocorticoids for a long time. After abrupt withdrawal of the drug, he complains of myalgia, increased fatigue, emotional instability, headache. Glucocorticoid withdrawal syndrome developed. What drugs are prescribed to correct this condition?

A. AKTG

B. Glucocorticoids

S. Mineralocorticoids

D. Adrenaline

E. Corticosteroids

A patient with Itsenko-Cushing syndrome has an increased cortisol content in the blood. Which endocrine gland pathology is it connected with?

A. The cortical part of the adrenal glands

B. The cerebral part of the adrenal glands

S. Pancreas

D. Hypophysis

E. Thyroid gland

A man who has been in a state of stress for a long time has a significantly increased content of 17-ketosteroids in his urine, which primarily indicates increased secretion:

A. Estradiol

B. Aldosterone

S. Adrenaline

D. Cortisol

E. Progesterone

The woman showed signs of virilism (hair growth on the body, irregular menstrual cycle). Hyperproduction of which hormone can cause such a condition?

A. Estriola

B. Testosterone

S. Relaxin

D. Oxytocin

E. Prolactin

The woman has a "moon-shaped" face, obesity of the upper part of the body, stretch marks on the front abdominal wall, hirsutism. The urine has an elevated level of 17-oxyketosteroids. Such manifestations are characteristic of:

A. Pheochromocytomas

B. Kon's syndrome

S. Itsenko-Cushing syndrome

D. Primary hypoaldosteronism

E. Secondary hyperaldosteronism

A patient suffering from Itsenko-Cushing's disease was consulted about excess body weight. The survey revealed that the energy value of the consumed food is 1700-1900 kcal/day. What is the leading cause of obesity in this case?

A. Hypodynamia

B. Insufficiency of insulin

C. Excess insulin

D. Insufficiency of glucocorticoids

E. An excess of glucocorticoids

Glucocorticoids and nonsteroidal anti-inflammatory drugs are widely used in practical medicine. One of the negative consequences of long-term glucocorticoid therapy is:

A. Polyuria

B. Hyponatremia

C. Hyperkalemia

D. Osteoporosis

E. Hypotension

4. Summary:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

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7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

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Electronic information resources:

1. https://info.odmu.edu.ua/chair/biology/-

2. http://libblog.odmu.edu.ua/

3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №24.

Topic: Pancreas and adrenal medulla hormones. Chemical structure and mechanism of action. Hormonal regulation of blood sugar. Local hormones, their structure, biological role. Hormones of the digestive tract. **Goal**: to study the mechanism of action of insulin, glucagon, the mechanism of their action, diabetes mellitus, the stimulating effect of insulin.effect of digestive tract hormones, types of hormones, synthesis of gastrin, secretin, main digestive tract hormones. Eicosanoids.

**Basic concepts:**metabolism, digestion of carbohydrates, lipids and proteins, hypoglycemia, hyperglycemia.

**Equipment:**Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- Mechanisms of influence of insulin and glucagon on metabolism
- Insulin: structure
- Synthesis of insulin
- Influence of insulin on the metabolism of carbohydrates, lipids and proteins
- The difference between glucagon and insulin
- The nature of gastrointestinal hormones, their mechanism of action on target cells.
- Origin and mechanism of action of leukotrienes.
- Enzyme cyclooxygenase, what reactions does it catalyze.

- Features of clinical manifestations of functional and organic diseases of the esophagus, stomach and duodenum.

- The concept of eneterohormone.
- Groups of gastrointestinal hormones.
- The concept of gastroinhibitory polypeptide
- The higher education applicant should be able to:
- Mechanism of insulin synthesis
- The structure of the tyrosine kinase receptor
- The influence of pancreatic hormones on the metabolism of biopolymers
- Hypoglycemia, index
- Questions to check basic knowledge on the topic of the class:
- insulin: structure,
- biosynthesis and secretion of pancreatic hormones
- influence on the metabolism of carbohydrates, lipids, proteins.
- growth-stimulating effects of insulin.
- be able to explain the work of the insulin receptor
- types of diabetes
- representatives of gastrin
- representatives of secretin
- what are gastrointestinal hormones by nature
- action of gastrin
- action of secretin
- action of motilin
- renin-angiotensin system
- action of angiotensinogen I
- action of angiotensinogen II

3. Formation of professional skills.

3.1 Demonstration and practical work: "Determination of sugar in urine by Fehling's reagent"

Principle of the method: In Fehling's reagent, copper (P) ions are in the form of a complex compound with tartrates. This reaction for glucose in its general form can be represented by equations:



The advantage of Fehling's reagent is that copper does not fall out in the form of copper (II) oxide when the reagent is in excess.

Procedure: 1 ml of urine and 1 ml of Fehling's reagent are introduced into the test tube. The mixture is stirred and heated to boiling. In the presence of sugar in the urine, a brick-red precipitate of copper (I) oxide appears.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Test tasks:

What hormone stimulates glycogen biosynthesis and enhances anabolic processes?

A. Adrenaline

B. Norepinephrine

S. Cholecystokinin

D. Insulin

E. Thyroxine

Glucose is used by its transport from the extracellular space through the plasma membrane into cells. This process is stimulated by the hormone:

A. Insulin

B. Glucagon

S. Thyroxine

D. Aldosterone

E. Adrenaline

The patient is in a state of hypoglycemic coma. An overdose of which hormone can lead to such a situation?

A. Insulin

**B.** Cortisol

S. Somatotropin

D. Progesterone

E. Corticotropin

The parents of a five-year-old child turned to the hospital. During the examination, it was found: retardation in mental development and growth, the child is sedentary. General exchange is reduced. What disease does the child have?

A. Cretinism

B. Lesch-Nyhan syndrome

S. Phenylketonuria

D. Hyperparathyroidism

E. Endemic goiter

Insulin, like other hormones of a protein-peptide nature, has receptors on the surface of the cytoplasmic membrane. Name the mechanism of realization of the effect of insulin in target cells?

A. Adenylate cyclase messenger system

B. Guanylate cyclase messenger system

C. Protein kinase cascade

D. Phosphoinositide messenger system

F. All answers are correct

The patient mistakenly took an excess dose of thyroxine. What changes in the secretion of thyroliberin and thyrotropin will this lead to?

A. The secretion of hormones will increase

A. There will be no change in secretion

C. Secretion of thyroliberin will increase, thyrotropin will decrease

D. Secretion of thyrotropin will increase, and thyroliberin will decrease

F. Hormone secretion will decrease

Acetylsalicylic acid is used in the treatment of rheumatism. For what process does acetylsalicylic acid affect?

A Synthesis of prostaglandins

B Breakdown of glucose

C Glycogen synthesis

D Synthesis of amino acids

E Fat breakdown

Various biologically active compounds are involved in blood pressure regulation. Which can peptides entering the blood affect the tone of blood vessels?

A Quinines

B Leukotrienes

C Enkephalins

D Iodothyronine

E Endorphins

The anti-inflammatory effect of a number of drugs is due to inhibition of the release of arachidonic acid. This acid is a precursor of which biologically active substances?

A Prostaglandins

B Uric acid

C Urea

D Hema

E Cholesterol

The exchange of arachidonic acid is accompanied by the formation of biologically active compounds. Name one of them that is formed with the participation of the lipoxygenase pathway.

A Leukotrienes.

B Quinines.

C Catecholamines.

D Bile acids.

E Steroids.

Nonsteroidal anti-inflammatory drugs are used in medical practice for treatment rheumatoid arthritis, osteoarthritis, inflammatory connective tissue diseases.

The activity of which enzyme is inhibited by these drugs?

A cyclooxygenase

B hexokinase

C succinate dehydrogenase

D aminotransferases

E xanthine oxidase

The patient was prescribed aspirin as an anti-inflammatory agent that inhibits production prostaglandins What enzyme is blocked at the same time?

- A Cyclooxygenase
- B Monooxygenase
- C Dioxygenase
- D Lipoxygenase
- E Peroxidase

Indomethacin is an active nonsteroidal anti-inflammatory drug that

used in medical practice for the treatment of rheumatoid arthritis,

osteoarthritis, inflammatory diseases of connective tissue. Which process slows down indomethacin?

- A synthesis of prostaglandins
- B formation of kinins
- C formation of angiotensin II
- D synthesis of amino acids
- E synthesis of purines

Steroid hormones activate the synthesis of the inhibitor of phospholipase A2, in connection with which their anti-inflammatory action consists in inhibiting the synthesis:

- A Prostaglandins
- B Kallikrein
- C bradykinin
- D Kininogens
- E Histamine

Biosynthesis of prostaglandins begins with the release of arachidonic acid from phosphoglycerides. What enzyme catalyzes this process?

- A Phospholipase A2
- B Cholesterolesterase
- C Sphingomyelinase
- D Triacylglyceride lipase
- E Lipoprotein lipase

A patient with rheumatism was prescribed prednisolone. The anti-inflammatory effect is due to the release of arachidonic acid. This acid is a precursor of which biologically active substances?

- A Prostaglandins
- B Urea
- C Hemu
- D Uric acid
- E Cholesterol

During the utilization of arachidonic acid on the cyclooxygenase pathway, biologically active substances are formed. Will indicate them.

- A Prostaglandins
- B Interferons
- C Biogenic amines
- D Somatomedins
- E Insulin-like growth factors

In the therapy of chronic inflammatory processes, a number of drugs are used. Indicate which of them reversibly inhibits cyclooxygenase of arachidonic acid

- A Indomethacin
- B Antimycin
- C Vikasol
- D Carnitine
- E Cholecalciferol

The activity of cyclooxygenase can be inhibited by the use of certain drugs. Which of them irreversibly inhibits the action of this enzyme?

A Aspirin

- B Insulin
- C Oligomycin
- D Allopurinol
- E Aminalon

## 4. Summary:

**5. List of recommended literature**(main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

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7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

# Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

# Practical class №25

Topic: Digestion of carbohydrates, lipids, proteins, nucleoproteins in the gastrointestinal tract. Enzymes, biochemical mechanisms. Chemical composition of gastric and intestinal juice. Hereditary disturbances of digestion.

**Goal:** To inform higher education applicants that nutrition is a necessary prerequisite for human life, which ensures normal metabolism, the dynamic state of all biomolecules, cellular and extracellular structures.

**Basic concepts:**—*digestion of foodnutrients, nutrients, components of normal nutrition; biological value of individual nutrients.* 

Equipment: Laboratory of the department

# Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know :

-organization of the digestive tract,

-enzyme spectrum and optimal pH values in separate departments of the digestive tract, -end products of digestion of carbohydrates, lipids, nucleoproteins and proteins in the small intestine.

The higher education applicant should be able to:

-determine all forms of acidity of gastric juice, make a medical-biological conclusion and predict the type of pathology under which the acidity of gastric juice will increase or decrease.

Questions to check basic knowledge on the topic of the class:

1. Components of normal human nutrition. Macrocomponents, microcomponents.

2. The needs of the human body in nutritional compounds.

3. Digestion of nutrients.

3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "*Determination of all forms of acidity of gastric juice*". *Recommendations for performing tasks.* 

Principle of the method: Quantitative determination of the acidity of gastric juice is performed by titrating a portion of the filtered juice with 0.1 n NaOH with indicators, taking into account the amount of alkali used for titration (alkali neutralization). Distinguish between total acidity, total HCl, free and bound HCl.

Procedure: Measure 5 ml of filtered gastric juice into a flask, add 2 drops of dimethylaminoazobenzene and 2 drops of phenolphthalein. Titrate with 0.1 N NaOH until an orange color appears. Note the amount of alkali used for titration (V1). Continue the titration to a lemon-yellow color, note the amount of alkali (V2) that went into the titration, count from zero. Continue the titration until a pink color, note the amount of alkali (V3), count from zero.

Calculation:

V1 – corresponds to free HCl;

V2 is auxiliary, used for calculation. Corresponds to total HCl:

$$\frac{V_2 + V_3}{2}$$

V3 corresponds to total acidity.

Acidity values are determined by the formula:  $X = \frac{V (M\pi) \cdot 1000 \cdot 0,1}{5}, MMOЛЬ/Л$ 

Bound HCl is found by the difference between total and free HCl. Normally in adults: free HCl – 20-40 mmol/l; total HCl – 30-50 mmol/l;

bound HCl - 10-20 mmol/l; total acidity - 40-60 mmol/l.

Conclusion: The obtained result should be evaluated from the point of view of belonging to normal parameters of acidity of gastric juice or differences from them. Based on the received data, propose a diagnosis.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

1. Biochemistry of human nutrition: components and nutrients of normal nutrition.

2. Digestion and biological value of carbohydrates. Enzymes of the stomach and intestines.

3. Digestion and biological value of lipids. Enzymes of the stomach and intestines.

4. Digestion and biological value of proteins. Enzymes of the stomach and intestines.

5. Digestion of nucleoproteins.

6. Disruption of digestion of certain nutrients in the stomach and intestines. Hereditary enzymopathies of digestive processes.

7. Microelements in human nutrition. Biological functions of individual trace elements; manifestations of trace element deficiency.

Test tasks.

1. Substances in the digestive system undergo certain changes. Enzymes of which class mainly carry out enteral transformations?

A \*Hydrolases

B Oxidoreductases

C Transferases

- D Lyases
- E Ligases

2. The most important phase of the breakdown of starch and glycogen occurs in duodenum under the action of pancreatic enzymes. What enzyme splits (-1,4-glycosidic bonds

in these molecules?

A \* alpha-amylase

- B Amylo-1,6-glycosidase
- C aldolase
- D hexokinase
- E maltase

3. In a patient with complaints of poor appetite, weight loss, pain in the epigastric area, the analysis of gastric juice showed the presence of achillea. What does this term mean?

A \* Absence of free NSI and pepsin.

In the absence of free NSI

C Lack of acidity

D Absence of free and bound NSI

E Absence of gastromucoprotein

4. The patient complains of loss of appetite, weight loss, unpleasant belching, pain and heaviness in the epigastric region. Analysis of gastric juice showed: total acidity 20 mM/l, free HCI absent, bound HCI 18 mM/l, lactic acid present. What disease can be assumed?

A \*Stomach tumor

In Gastric ulcer disease

C Hyperacid gastritis

D. Hypoacidic gastritis

E Ulcerative disease of the duodenum

5. The patient notes periodic pains in the epigastric area, heartburn. When examining the gastric juice, hyperacidity was established. Which of the given data corresponds to this condition?

A \* 80 mM/l

In 40 mM/l With 60 mM/l D 25 mM/l E 55 mM/l

6. A large amount of fluoride was found in the water of the river, which is the source of the city's water supply. What pathology can develop among the inhabitants of this city?

A \*Fluorosis In Thyrotoxic goiter C Juvenile current D Caries It's beri-beri

7. A patient with hypofunction of the pancreas is prescribed pancreatin. When is it most rational to take this drug?

A \*Before food In After eating C During meals D Before going to bed E Any time

8. At the final stage of protein breakdown, trace elements Zn, Mn, Mg, Co play an important role, which:

A \* They increase the activity of peptidases

Contribute to the absorption of proteins

C They reduce the activity of peptidases

D They inhibit the absorption of proteins

E Inactivate peptidases

9. The patient has impaired protein digestion in the stomach and small intestine. This process is caused by a deficiency of which enzymes?

A Peptidase

B Synthetase

C Amylase

D Lipase

E Transferase

10. Pancreatic enzymes are secreted in the duodenum in an inactive state. Specify which enzyme activates trypsinogen.

- A Enterokinase
- B Gastrixin
- C Lipase
- D Pepsin
- E Elastase

11. The child complains of a toothache. The dentist diagnosed caries

enamel damage. The amount of which mineral substances is decreasing in the region carious damage:

A \* Phosphorus, fluorine, calcium;

- B Sodium, calcium, potassium;
- C Potassium, phosphorus, fluorine;
- D Magnesium, fluorine, calcium;
- E Phosphorus, magnesium, potassium.

12. A woman, an employee of a confectionery shop, turned to a dentist. The patient drew attention to increased sensitivity to caries. For the purpose of remineralizing therapy, the doctor prescribed fluoride preparations. What is the role of fluoride in this therapy?

- A \*Increase in the formation of fluorine apatite;
- B Increased permeability of enamel;
- C Suppression of alcoholic fermentation;
- D Decreased synthesis of proteoglycans;
- E Activation of salivary proteases.

13. The patient is hyperacidic. Name the hormone that stimulates the secretion of HCl and pepsinogen in the stomach.

- A Gastrin
- B Insulin
- C Somatotropin
- D secretin
- E Glucagon

14. Parenteral nutrition is recommended for a patient with a damaged esophagus. Indicate which of the indicated pharmaceutical preparations is a hydrolyzate of amino acids?

- A Hydrolysin
- B Asparkam
- C Rheopolyglukin
- D Polyglukin
- E Panangin

15. To obtain amylase enzyme in its pure form from the pancreas of animals, the method of affinity chromatography with a ligand attached to the carrier is used. Which of the following substances is used as a ligand?

- A Starch
- B Cellulose
- C Lactose
- D Sucrose
- E Glucose

16. In the patient, a stone in the common bile duct blocked the flow of bile to the intestine. Indigestion of what substances is observed?

- A Zhiriv
- B Proteins
- C Water-soluble vitamins
- D Microelements
- E Carbohydrates

17. The patient was diagnosed with achlohydria. This leads to a decrease in the activity of which enzyme?

- A Pepsin
- B Trypsin
- C Chymotrypsin
- D Elastase
- E Aminopeptidases

18. The drug tannin is used in practical medicine as an astringent for acute and chronic intestinal diseases. The astringent effect of tannin is related to its ability to:

- A Denature proteins
- B Hydrolyze proteins
- C Renature proteins
- D Salt the proteins
- E Oxidize proteins

19. In the patient, as a result of the study, a violation of protein digestion in the stomach and small intestine was revealed. Lack of which enzymes leads to to such a violation?

- A Peptidases
- B Transferases
- C Amylase
- D Lipases
- E Oxidoreductases

20. Nucleosidases and nucleotidases of the gastrointestinal tract catalyze the hydrolysis of nucleic acids and mononucleotides, as well as those medicinal substances that have the following chemical bond in the molecule:

- A Phosphodiester
- B Hydrogen
- C Peptide
- D Glycoside
- E Amid

21. In children, the pH of gastric juice ranges from 4.0 to 5.0. Name the enzyme of gastric juice that is active under these conditions.

- A Renin
- B Pepsin
- C Chymotrypsin
- D Trypsin
- E Elastase

22. According to the result of the analysis of the patient's saliva, it was established that the pH is 8.0, i.e. shifted to the alkaline side. This state of saliva contributes to:

- A \* Formation of tartar;
- B Development of caries;
- C Development of fluorosis;
- D Development of tooth tissue hyperplasia;
- E Development of tooth tissue hypoplasia.

23. The patient was prescribed a bile preparation to improve the digestion of fatty food. What are the components?

Does this drug take part in the emulsification of fats?

- A Bile acids;
- B Cholesterol;
- C Diglycerides;
- D Amino acids;
- E Higher fatty acids.

24. The pancreas secretes an enzyme capable of hydrolyzing  $\alpha$ -1,4-glycosidic bonds in the glycogen molecule. Point to this enzyme.

- A α-Amylase
- B Phosphatase;

- C Enterokinase
- D Chymotrypsin
- E Lysozyme.
- 25. With exocrine insufficiency of the pancreas sometimes with

the drug "festal", which contains pancreatic enzymes, for improvement Bile acid preparations are recommended for digestion. For what purpose is such an additive used?

- A \*For emulsifying fats
- B To activate proteolytic enzymes
- C For activation of ?-amylase
- D To stimulate the secretion of pancreatic juice
- E To stimulate intestinal peristalsis

26. Inhibition of the synthesis of bile acids from cholesterol in the experimental liver animals led to impaired digestion of lipids. What is the role of these acids in lipid digestion?

- A \*Emulsify food lipids
- B Participate in resynthesis of lipids
- C They are part of LDL
- D Maintain an alkaline environment in the intestines
- E Activate the formation of chylomicrons
- 4. Summing up.
- 5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

- 2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.
- 3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

## Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

# Practical class №26

# Topic: Biochemical characteristics and classification of vitamins. Water-soluble vitamins B1, B2, B6, PP. Their coenzyme role and symptoms of hypovitaminosis.

**Goal:** <u>Study of the mechanism of action and biological role of this group of vitamins.</u> Show the possibility of using them in practical medicine.

To acquaint higher education applicants with the coenzyme functions of vitamins B1, B2, B6, RR.

To teach higher education applicants to carry out qualitative determination of vitamins B1, B2, B6, RR.

 Study of coenzyme forms of vitamins and their role in the catalytic activity of enzymes.

 Basic concepts:
 vitamins, coenzymes, prosthetic group

 Equipment:
 Laboratory of the department

## Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know :

- -what vitamins are, their biological role for the body
- -chemical composition and structure of vitamins;
- -Symptoms and consequences of hypovitaminosis B1, B2, B6, PP.

Questions to check basic knowledge on the topic of the class:

- Coenzyme function of vitamins
- What are the groups of coenzymes?
- Differences of coenzymes from prosthetic groups
- The role of coenzymes in catalysis
- The structure of the most common coenzymes

-Causes of hypovitaminosis B1, B2, B6, PP.

- Symptoms of hypovitaminosis B1, B2, B6, PP and their consequences for the body.

3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "*Qualitative reactions to vitamins B1, B2, B6, PP*". *Recommendations for performing tasks.* 

## A. Qualitative determination of thiamine

The principle of the method: during oxidation, thiamine turns into thiochrome, which has the ability to fluoresce blue in ultraviolet light.

Work progress: 1. Oxidation of thiamine to thiochrome:

Pour 0.5 ml of 5% thiamine solution into the test tube, then 1.0 ml of 5% ferric cyanide and 2.0 ml of 10% NaOH solution. Mix thoroughly and leave for 10 minutes.

2. Extraction of thiochrome:

After 10 minutes, 1.0 ml of isobutyl alcohol is added to the test tube, shaken and allowed to settle for 5 minutes.

3. Registration of indicators and conclusion:

The test tube is brought to the source of ultraviolet light. A solution of thiochrome in isobutyl alcohol fluoresces blue.

## **B.** Qualitative determination of riboflavin

Principle of the method:

In ultraviolet light, riboflavin is able to fluoresce in a yellow-green color. When it is reduced with sodium hyposulfite, it loses this property.

The main stages of work performance.

1. Preparation of material for research:

Take 2 test tubes and label them "experiment" and "control". Pour 1.0 ml of 0.02% riboflavin solution into both test tubes.

2. Restoration of riboflavin:

A few crystals of sodium hyposulfite are added to the test tube marked "experiment". The solution turns from bright yellow to pale yellow.

3. Comparative fluorometry:

Both test tubes are raised to the source of ultraviolet light. Reconstituted riboflavin in a test tube does not fluoresce under ultraviolet light.

## **B.** Qualitative determination of pyridoxine

Principle of the method:

If iron chloride is added to the pyridoxine solution, a red color of the complex salt appears, similar to red iron phenolate.

The main stages of work performance.

1. Preparation of material for research:

Pour 0.5 ml of 5% pyridoxine solution into the test tube.

2. Carrying out the reaction:

Pour 0.5 ml of 5% ferric chloride solution into the test tube and shake it. The mixture turns red.

## G. Qualitative determination of vitamin RR

Principle of the method:

Nicotinic acid, when heated with a solution of copper acetic acid, forms a blue soluble precipitate of the copper salt of nicotinic acid.

The main stages of work performance.

1. Preparation of material for research:

Pour 1.0 ml of 1% nicotinic acid solution into the test tube.

2. Formation of the copper salt of nicotinic acid:

Add 1.0 ml of 5% copper acetic acid solution to the nicotinic acid solution. They stir. They heat up. A poorly soluble blue precipitate of the copper salt of nicotinic acid is formed.

#### Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

- Participation of nicotinamide coenzymes in catalysis
- Participation of flavin coenzymes in catalysis
- Participation of pyridoxal coenzymes in catalysis

Questions to check the final level of knowledge:

1. Representatives of water-soluble vitamins.

2. Structure, physical and chemical properties of vitamin B1 (thiamine). Coenzyme form of thiamine. Mechanism of action and biological role of thiamine. Avitaminosis and hypovitaminosis. Use in medical practice.

3. Structure, physical and chemical properties of vitamin B2 (riboflavin). Coenzyme form of vitamin B2. Mechanism of action and biological role. Avitaminosis and hypovitaminosis. Use in medical practice.

4. Structure, physical and chemical properties of vitamin B6 (pyridoxine). Coenzyme forms of pyridoxine. Mechanism of action and biological role of pyridoxine. Avitaminosis and hypovitaminosis.

5. Structure, physical and chemical properties of vitamin PP (niacin, nicotinic acid, nicotinamide). Coenzyme forms of vitamin PP. Mechanism of action and biological role of niacin. Avitaminosis and hypovitaminosis.

Test tasks.

1.Inactivation of vitamin PP and its coenzyme forms is carried out by methylation to Nmethylnicotinamide, which is excreted in the urine. Therefore, with the long-term appointment of vitamin PP in high doses, the following dietary recommendation is appropriate:

And \*enrichment of the diet with proteins rich in methionine

In Enriching the diet with proteins rich in tryptophan

C Enrichment of the diet with unsaturated fatty acids

D Enrichment of the diet with carbohydrates

E Sharp restriction of carbohydrates

2. Vitamin derivatives are part of the coenzymes of the respiratory chain. NAD is the coenzyme form of which vitamin?

A \*RR

- B B6
- C B1

D B2

E B3

3. Tuberculosis patients take a drug that is an anti-vitamin of nicotinic acid. Specify this substance.

- A \*Isoniazid
- B Sulfanilamide
- C Akrichin
- D Isoriboflavin
- E Oxythiamine

4. The patient complains of lack of appetite, nausea, abdominal pain, diarrhea, headache, memory impairment. Dermatitis is observed in the neck and face. Which vitamin is deficient?

- A \*Vitamin B5
- B Vitamin B1
- C Vitamin B3
- D Vitamin B2
- E Vitamin B6

5. A patient with a severe form of diarrhea, dermatitis and dementia is prescribed vitamin PP. State the role of vitamin PP in metabolism.

A \*Participation in redox processes

- B Participation in the hydrolysis of peptide bonds
- C Participation in isomerization reactions
- D Participation in oxygen transport
- E Participation in the formation of peptide bonds

6. Determination of the activity of some transaminases is widely used in medical practice for the purpose of diagnosing damage to internal organs. The cofactor of these enzymes is the active form of the vitamin

A \*B6 B B1 C B2 D B12 E RR

7. Determination of the activity of alanine aminotransferase (AlT) and aspartate aminotransferase (AsT) is widely used in medical practice for the purpose of diagnosing damage to internal organs. The prosthetic group of these enzymes is the coenzyme form of which vitamin?

A \*B6 B B1 C WITH D B5 E R

8. Insufficiency of which vitamin causes a decrease in the activity of aminotransferases and decarboxylases?

- A \*B6 B B12 C B2 D B3
- E B15

9. The patient has dry lips, cracks and "crusts" in the corners of the mouth, a bright red tongue, seborrheic dermatitis of the nasolabial folds, photophobia and conjunctivitis. What vitamin deficiency is this associated with?

- A \*Riboflavin
- B Cholecalciferol
- C Cobalamin
- D Pyridoxine
- E Ascorbic acid

10. The patient has neurasthenic syndrome, diarrhea, dermatitis. What vitamin deficiency is this associated with?

- A \*Nicotinic acid
- B Vitamin D
- C Vitamin K
- D Vitamin B12
- E Folic acid

11. A compound containing isoalloxazine was found in the patient's urine cycle. What is this compound?

- A \*Vitamin B2;
- B Vitamin B5;
- C Vitamin B6
- D Vitamin B1

E Vitamin B3

12. In clinical practice, the drug isoniazid is used, which is competitively included in the coenzyme structure, which cannot participate in redox processes, and this leads to the stoppage of the growth of Koch's bacillus. Specify which enzyme systems are inhibited:

- A \*NAD-dependent enzymes
- B FAD-dependent enzymes
- C CoQ
- D Cytochrome c
- E Cytochrome a1

13. A 38-year-old patient suffering from chronic alcoholism has edema, muscle atrophy, cardiovascular insufficiency, and peripheral nerve pain. Such symptoms are caused by the lack of which vitamin in the body?

A \*vitamin B1

- B vitamin A
- C vitamin E
- D vitamin B6
- E vitamin K

14. The patient has pain along the course of peripheral nerves. This can be caused by a lack of which vitamin?

- A \*vitamin B1
- B vitamin A
- C vitamin E
- D vitamin B12
- E vitamin K

15. Redness of the mucous membrane of the mouth, cracks in the corners and lips of the patient, peeling of the skin, dryness and inflammation of the conjunctiva on the face, sprouting of the vascular mesh

cornea The probable cause of this pathology is a lack of vitamin:

A	*B2
В	WITH
С	IS
D	Κ
E	D

16. Nicotinic acid amide plays an important role in metabolism. What a disease occurs with his hypovitaminosis?

- A \*Pellagra
- B Rickets
- C Anemia
- D Xerophthalmia
- E Beri-Beri

17. Pyridine-dependent dehydrogenases act as the primary hydrogen acceptor during tissue respiration. Which of the vitamins is necessary for the formation of the corresponding coenzyme (NAD +)?

A \* Vitamin PP

- B Vitamin C
- C Vitamin B1
- D Vitamin B2
- E Vitamin B6

18. Vitamins must be included in a person's diet. Which one is used to treat pellagra?

- A \*Vitamin B5
- B Vitamin B1
- C Vitamin C
- D Vitamin A
- E Vitamin D

19. For the treatment of heart diseases, the drug cocarboxylase is used. This drug is the coenzyme form of which vitamin?

A \* B1 B B6 C B12 D WITH E R

20. Water-soluble vitamins in the body are converted into coenzyme forms. Coenzyme form of which vitamin is thiamine diphosphate (TDP)?

- A \* vitamin B1 B vitamin B2 C vitamin C
- D vitamin B6
- E vitamin B12

21. The biochemical function of water-soluble vitamins depends on their ability to transform into coenzyme forms. What coenzyme form can vitamin B2 (riboflavin) convert into:

- A \* FMN (flavin mononucleotide)
- B NAD+ (nicotinamide adenine dinucleotide)
- C TMF (thiamine monophosphate)
- D TDF (thiamine diphosphate)
- E PALF (pyridoxal phosphate)

22. A 30-year-old man with pulmonary tuberculosis is prescribed isoniazid. Insufficiency of which vitamin can develop as a result of long-term use of this one drug?

- A \* Pyridoxine
- B Tocopherol
- C Cobalamin
- D Ergocalciferol
- E Retinol
- 4. Summary:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

### Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

1. https://info.odmu.edu.ua/chair/biology/-

2. http://libblog.odmu.edu.ua/

3. https://moodle.odmu.edu.ua/login/index.php

### Practical class №27

Topic: Water-soluble vitamins C, biotin, folic acid, B12, pantothenic acid. Structure, biological role and symptoms of hypovitaminosis.

**Goal:**<u>To make higher education applicants aware of the importance of studying the mechanism</u> of action and biological role of this group of vitamins. Show the possibility of using them in practical <u>medicine</u>.

<u>To acquaint higher education applicants with the peculiarities of the coenzyme functions of the vitamins biotin, folic acid, vitamin B12, with the peculiarities of the influence of vitamin C on metabolic processes.</u>

To teach higher education applicants the quantitative determination of vitamin C in plant objects.

Basic concepts: vitamins, coenzyme functions, symptoms of hypo-vitaminosis, antivitamins Exprime article and the department

Equipment: Laboratory of the department

### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know :

-structural formulas of vitamins Bc, C, B12, P, H.

-the role of these vitamins in metabolic processes as coenzymes or prosthetic groups.

-causes, symptoms and consequences of hypovitaminosis.

The higher education applicant should be able to:

-to determine the amount of vitamin C in a plant object.

Questions to check basic knowledge on the topic of the class:

1. Which water-soluble vitamins are coenzymes?

- 2. Sources of vitamin C, daily requirement. What does vitamin C cause?
- 3. Coenzymes-nucleotides.
- 4. Coenzymes-phosphorus esters of vitamins.
- 3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Quantitative determination of vitamin C in products by the Tillmans method.

Recommendations for performing tasks.

Principle of the method: Determination of the content of vitamin C (ascorbic acid) is based on its reaction with 2,6-dichlorophenolindophenol. Using the change in color, based on the amount of the reagent spent on the oxidation of vitamin C, it is possible to determine its amount in the studied object.

Progress:

1. Preparation of material for research.

Take 1.0 g of rose hips or needles, grind it in a porcelain mortar, then add 10.0 ml of 2% hydrochloric acid. Quickly filter the hood to a dry flask.

2. Determination of content by titration.

Measure 3.0 ml of the filtrate into a conical flask and titrate with a 0.001 N solution of the sodium salt of 2,6-dichlorophenolindophenol to a pale pink color that does not disappear within 30 seconds.

The calculation is carried out according to the following formula:

$$C = , where \frac{M \cdot 0,088 \cdot 10}{A \cdot B}$$

C is the concentration of vitamin C in the solution;

M is the amount of 2,6-dichlorophenolindophenol used for titration;

0.088 – the amount of vitamin C that binds 1 ml of 2,6-dichlorophenolindophenol;

10 - the number of hoods;

A is the amount of extract taken for titration;

B is the amount of researched material in

Conclusion: with the help of the described method, it is possible to determine the content of vitamin C in plant objects.

Requirements for work results.

Enter the obtained data into the workbook.

Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

-1. Physico-chemical characteristics of folic acid, biological role and mechanism of action.

-2. Physico-chemical characteristics of biotin, biological role and mechanism of action.

-3. Physico-chemical characteristics of vitamin C, biological role and mechanism of action. The main signs of hypovitaminosis.

-4. Structure, physical and chemical properties, biological role, mechanism of action of vitamin

P.

-5. Structure, physical and chemical properties, biological role, mechanism of action of vitamin

B12.

-6. Structure, physical and chemical properties, biological role, mechanism of action of pantothenic acid.

-7. Hypovitaminosis of the above-mentioned vitamins.

Test tasks.

1. Insufficiency of which vitamin leads to the development of hypercoagulation syndrome? A \*S

- In B12
- S A
- D B6
- E D2

2. The doctor diagnosed the patient with scurvy. This disease was a manifestation of which pathological condition associated with improper nutrition?

A \* Nutritional imbalance In Malnutrition C Overeating D Stomach ulcer

E Helminthosis

3. The mechanism of antimicrobial action of sulfonamide drugs is based on their structural similarity with:

A \* Para-aminobenzoic acid In Glutamic acid With folic acid D Nucleic acid E Antibiotics

4. In the process of biotransformation of the anesthetic drug novocaine, paraaminobenzoic acid is formed. Therefore, with the combined use of novocaine and sulfonamides, the following is most likely to occur:

A \*Decreasing the antimicrobial effect of sulfonamides

In Strengthening the antimicrobial action of sulfonamides

C Reduction of the anesthetic effect of novocaine

D Strengthening of the anesthetic effect of novocaine

E Decrease in the pharmacological activity of both drugs

5. Lack and absence of ascorbic acid in food cause various diseases. Name the main one of them.

- A Scurvy
- B Rickets
- C Beri-Beri
- D Pellagra
- E Gout

6. Complex proteins in the human body perform various functions. Which glycoprotein contained in raw egg white disrupts the absorption of vitamin H and can lead to acute biotin deficiency?

- A Avidin
- B Fibrinogen

- C Hemoglobin
- D Interferon
- E Caseinogen

7. The formation of collagen in the human body requires the hydroxylation of proline, which occurs with the participation of prolyl hydroxylase. What substance activates this process:

- A Ascorbic acid
- B NAD
- C FAD
- D Biotin
- E Pyridoxine phosphate

8. The patient was diagnosed with pernicious anemia. Indicate which vitamin deficiency this may be due to.

- A
   B12

   B
   B3

   C
   B2
- D WITH
- E IS

9. Vitamins, when used simultaneously, can enhance each other's effects. Which of the vitamins potentiates the antihyaluronidase activity of vitamin P?

- A \*Vitamin C
  B Vitamin B1
  C Vitamin D
  D Vitamin A
- E Vitamin B2

10. Certain conditions are necessary for the assimilation of vitamins by organisms. For the absorption of which vitamin, the presence of Castle's factor (a glycoprotein produced by the lining cells of the gastric mucosa) is necessary?

- A \*Vitamin B12
- B Vitamin C
- C Vitamin B5
- D Vitamin E
- E Vitamin B6

11. A patient with angina was prescribed a sulfonamide drug, the antimicrobial action of which is due to a violation of the synthesis of folic acid. With which substance do sulfonamides compete for the active center of the enzyme?

- A \* Para-aminobenzoic acid
- B Ubiquinone
- C Succinate
- D Glutamic acid
- E Citric acid

12. Bacteriostatic sulfonamides suppress the synthesis of nucleotides, nucleic acids and proteins in microbial cells, but in pharmacological doses they do not affect the synthesis of these substances in the human body. This difference is due to the fact that eukaryotic cells:

- A \*Folic acid is not synthesized
- B Para-aminobenzoic acid is not synthesized
- C Nucleotides are not synthesized

- D Impermeable to sulfonamides
- E Very quickly inactivate sulfonamides

13. Antimicrobial sulfonamide drugs are metabolized in the body by acetylation. At the same time, their bacteriostatic action:

- A \*Lost
- B is growing
- C Does not change
- D It changes to toxic in the human body
- E Intensifies

14. The patient has megaloblastic anemia (Addison-Birmer disease). It is advisable to treat with vitamin B12 in combination with the following drug:

- A \*Folic acid
- B Lipoic acid
- C Ascorbic acid
- D Nicotinic acid
- E Pangamic acid

15. The patient consumes several raw eggs daily, which contain the anti-vitamin of biotin - avidin. What disorders in lipid metabolism can occur in this case?

- A \*Biosynthesis of fatty acids
- B Cholesterol biosynthesis
- C Oxidation of glycerol
- D Absorption of lipids
- E Transport of lipids in the blood

16. Bacterial cells use folic acid to synthesize a certain vitamin, the derivatives of which are coenzymes of a number of important bacterial enzymes. Sulfanilamide drugs block the formation of these coenzymes, as they are antivitamins:

- A \* Para-aminobenzoic acid
- B Pyridoxine
- C Nicotinic acid
- D Riboflavin
- E to Holin

17. In a patient with complaints of pain in the stomach, a decrease in the secretory function of the stomach, which was accompanied by anemia, was established during the biochemical examination. Indicate which of the vitamins has an anti-anemic effect:

- A \* Cobalamin
- B Tocopherol
- C Retinol
- D Thiamine
- E Nicotinic acid

18. Some vitamins inhibit the formation of lipid peroxides in cells membranes and ensure the stability of biological membranes. Specify one of vitamins that have this effect.

- A \* ascorbic acid
- B naphthoquinone
- C cholecalciferol
- D pantothenic acid

E folacin

19. In a patient with frequent bleeding in the internal organs and mucous membranes in the composition

proline and lysine were found in collagen fibers. Lack of which vitamin leads to violation of their hydroxylation.

A \*Vitamin C

- B Vitamin E
- C Vitamin K
- D Vitamin A
- E Vitamin B1

20. The parents of a 10-year-old boy went to the doctor with a complaint about stunted growth. During the examination, the doctor found changes in the mucous membranes and suspicion of malignant anemia. He suggested that this disease is associated with vitamin deficiency. Indicate which vitamin deficiency can lead to

the development of such a condition?

- A \* Folic acid
- B Nicotinic acid
- C Orotic acid
- D to Holin
- E Arachidonic acid

21. The result of a violation of which biochemical reaction is the appearance of hemorrhages at scurvy disease.

- A \*hydroxylation of proline
- B glucose phosphorylation
- C dehydrogenation of isocitric acid
- D isomerization of phosphodioxyacetone
- E deamination of glutamic acid

22. A patient with tuberculosis was treated with isoniazid (tuberculostatic drug). Later, he developed signs of dermatitis, diarrhea, and damage to the central nervous system. Which vitamin should be prescribed for this patient?

- A \*vitamin RR
- B vitamin C
- C lipoic acid
- D vitamin A
- E vitamin B1.

23. In medical practice, sulfonamide drugs are used, which are

antimetabolites of paraaminobenzoic acid, which is synthesized by microorganisms. Indicate the synthesis of which vitamin is blocked.

- A \* Folic acid
- B Pangamic acid
- C Orotic acid
- D Nicotinic acid
- E Ascorbic acid

24. The patient complains of bleeding gums, spot hemorrhages. which vitamin preparation can be used in this case?

- A \* Askorutin
- B Thiamine hydrochloride

- C Cyanocobalamin
- D Nicotinic acid
- E Pyridoxine hydrochloride

25. Most vitamins undergo transformation in the human body. What vitamin takes part in the formation of coenzyme of acetylation (CoASH)?

- A \*Vitamin B3
- B Vitamin C
- C Vitamin D
- D Vitamin A
- E Vitamin K

26. A 50-year-old patient developed hypovitaminosis C as a result of long-term irrational nutrition. A decrease in the activity of which enzyme is the basis of connective tissue damage in this pathology?

- A \*Proline hydroxylases
- B Alanine aminotransferases
- C Pyruvate carboxylase
- D Tryptophan hydroxylase
- E Glutaminase

27. The enzyme hyaluronidase splits hyaluronic acid, as a result of which intercellular permeability increases. Which vitamin helps to strengthen the walls of blood vessels and inhibits the activity of hyaluronidase?

- A \* Vitamin R
- B Vitamin A
- C Vitamin B1
- D Vitamin B2
- E Vitamin D
- 4. Summary:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

### Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

Topic: <u>Digestion of carbohydrates, lipids, proteins, nucleoproteins in the gastrointestinal</u> <u>tract. Enzymes, biochemical mechanisms. Chemical composition of gastric and intestinal juice,</u> <u>hereditary enzymopathies. Biochemical characteristics and classification of vitamins. Water-</u> <u>soluble vitamins B1, B2, B6, PP. Coenzyme role, hypovitaminosis.</u>

**Goal:** <u>To inform higher education applicants that nutrition is a necessary prerequisite for human life, which ensures normal metabolism, the dynamic state of all biomolecules, cellular and extracellular structures. To study the mechanism of action and biological role of this group of vitamins.</u>

Show the possibility of using them in practical medicine.

<u>To acquaint higher education applicants with the coenzyme functions of vitamins B1, B2, B6,</u> <u>RR.</u>

To teach higher education applicants to carry out qualitative determination of vitamins B1, B2, B6, RR.

Study of coenzyme forms of vitamins and their role in the catalytic activity of enzymes.

**Basic concepts:**—*digestion of foodnutrients, nutrients, components of normal nutrition; biological value of certain nutrients, vitamins, coenzymes, prosthetic group* 

Equipment: Laboratory of the department

## Plan:

6. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

7. Control of the reference level of knowledge.

The higher education applicant should know:

-organization of the digestive tract,

-the enzyme spectrum and optimal pH values in certain departments of the digestive tract,

-end products of digestion of carbohydrates, lipids, nucleoproteins and proteins in the small intestine.

-what vitamins are, their biological role for the body

-chemical composition and structure of vitamins;

-symptoms and consequences of hypovitaminosis B1, B2, B6, PP.

The higher education applicant should be able to:

-determine all forms of acidity of gastric juice, make a medical-biological conclusion and predict the type of pathology under which the acidity of gastric juice will increase or decrease.

Questions to check basic knowledge on the topic of the class:

- 1. Components of normal human nutrition. Macrocomponents, microcomponents.
- 2. The needs of the human body in nutritional compounds.
- 3. Digestion of nutrients.
- 4. Coenzyme function of vitamins
- 5. Causes of hypovitaminosis B1, B2, B6, RR.
- 1. Symptoms of hypovitaminosis B1, B2, B6, PP and their consequences for the body.

8. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Determination of all forms of acidity of gastric juice". *Recommendations for performing tasks.* 

Principle of the method: Quantitative determination of the acidity of gastric juice is performed by titrating a portion of the filtered juice with 0.1 n NaOH with indicators, taking into account the amount of alkali used for titration (alkali neutralization). Distinguish between total acidity, total HCl, free and bound HCl.

Procedure: Measure 5 ml of filtered gastric juice into a flask, add 2 drops of dimethylaminoazobenzene and 2 drops of phenolphthalein. Titrate with 0.1 N NaOH until an orange color appears. Note the amount of alkali used for titration (V1). Continue the titration to a lemon-yellow color, note the amount of alkali (V2) that went into the titration, count from zero. Continue the titration until a pink color, note the amount of alkali (V3), count from zero.

Calculation:

V1 – corresponds to free HCl;

V2 is auxiliary, used for calculation. Corresponds to total HCl:

$$\frac{V_2 + V_3}{2}$$

V3 corresponds to total acidity.

Acidity values are determined by the formula:

$$X = \frac{V (мл) \cdot 1000 \cdot 0,1}{5}, \text{ммоль/л}$$

Bound HCl is found by the difference between total and free HCl.

Normally in adults: free HCl - 20-40 mmol/l;

total HCl – 30-50 mmol/l; bound HCl – 10-20 mmol/l; total acidity - 40-60 mmol/l.

Conclusion: The obtained result should be evaluated from the point of view of belonging to normal parameters of acidity of gastric juice or differences from them. Based on the received data, propose a diagnosis.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

*a.* Demonstrationsjust now-practical work"Qualitative reactions to vitamins B1, B2, B6, PP".

Recommendations for performing tasks.

#### A. Qualitative determination of thiamine

The principle of the method: during oxidation, thiamine turns into thiochrome, which has the ability to fluoresce blue in ultraviolet light.

Work progress: 1. Oxidation of thiamine to thiochrome:

Pour 0.5 ml of 5% thiamine solution into the test tube, then 1.0 ml of 5% ferric cyanide and 2.0 ml of 10% NaOH solution. Mix thoroughly and leave for 10 minutes.

2. Extraction of thiochrome:

After 10 minutes, 1.0 ml of isobutyl alcohol is added to the test tube, shaken and allowed to settle for 5 minutes.

3. Registration of indicators and conclusion:

The test tube is brought to the source of ultraviolet light. A solution of thiochrome in isobutyl alcohol fluoresces blue.

## **B.** Qualitative determination of riboflavin

Principle of the method:

In ultraviolet light, riboflavin is able to fluoresce in a yellow-green color. When it is reduced with sodium hyposulfite, it loses this property.

The main stages of work performance.

1. Preparation of material for research:

Take 2 test tubes and label them "experiment" and "control". Pour 1.0 ml of 0.02% riboflavin solution into both test tubes.

2. Restoration of riboflavin:

A few crystals of sodium hyposulfite are added to the test tube marked "experiment". The solution turns from bright yellow to pale yellow.

3. Comparative fluorometry:

Both test tubes are raised to the source of ultraviolet light. Reconstituted riboflavin in a test tube does not fluoresce under ultraviolet light.

### **B.** Qualitative determination of pyridoxine

Principle of the method:

If iron chloride is added to the pyridoxine solution, a red color of the complex salt appears, similar to red iron phenolate.

The main stages of work performance.

1. Preparation of material for research:

Pour 0.5 ml of 5% pyridoxine solution into the test tube.

2. Carrying out the reaction:

Pour 0.5 ml of 5% ferric chloride solution into the test tube and shake it. The mixture turns red.

### G. Qualitative determination of vitamin RR

Principle of the method:

Nicotinic acid, when heated with a solution of copper acetic acid, forms a blue soluble precipitate of the copper salt of nicotinic acid.

The main stages of work performance.

1. Preparation of material for research:

Pour 1.0 ml of 1% nicotinic acid solution into the test tube.

2. Formation of the copper salt of nicotinic acid:

Add 1.0 ml of 5% copper acetic acid solution to the nicotinic acid solution. They stir. They heat up. A poorly soluble blue precipitate of the copper salt of nicotinic acid is formed.

Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

- Participation of nicotinamide coenzymes in catalysis
- Participation of flavin coenzymes in catalysis
- Participation of pyridoxal coenzymes in catalysis

—

Questions to check the final level of knowledge:

1. Representatives of water-soluble vitamins.

2. Structure, physical and chemical properties of vitamin B1 (thiamine). Coenzyme form of thiamine. Mechanism of action and biological role of thiamine. Avitaminosis and hypovitaminosis. Use in medical practice.

3. Structure, physical and chemical properties of vitamin B2 (riboflavin). Coenzyme form of vitamin B2. Mechanism of action and biological role. Avitaminosis and hypovitaminosis. Use in medical practice.

4. Structure, physical and chemical properties of vitamin B6 (pyridoxine). Coenzyme forms of pyridoxine. Mechanism of action and biological role of pyridoxine. Avitaminosis and hypovitaminosis.

5. Structure, physical and chemical properties of vitamin PP (niacin, nicotinic acid, nicotinamide). Coenzyme forms of vitamin PP. Mechanism of action and biological role of niacin. Avitaminosis and hypovitaminosis.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

1. Biochemistry of human nutrition: components and nutrients of normal nutrition.

2. Digestion and biological value of carbohydrates. Enzymes of the stomach and intestines.

3. Digestion and biological value of lipids. Enzymes of the stomach and intestines.

4. Digestion and biological value of proteins. Enzymes of the stomach and intestines.

5. Digestion of nucleoproteins.

6. Disruption of digestion of certain nutrients in the stomach and intestines. Hereditary enzymopathies of digestive processes.

7. Microelements in human nutrition. Biological functions of individual trace elements; manifestations of trace element deficiency.

8. Coenzyme function of vitamins

9. Structure of the most common coenzymes

10. Causes of hypovitaminosis B1, B2, B6, RR.

11. Symptoms of hypovitaminosis B1, B2, B6, PP and their consequences for the body.

Test tasks.

9. Substances in the digestive system undergo certain changes. Enzymes of which class mainly carry out enteral transformations?

A \*Hydrolases

B Oxidoreductases

- C Transferases
- D Lyases
- E Ligases

10. The most important phase of the breakdown of starch and glycogen occurs in duodenum under the action of pancreatic enzymes. What enzyme splits (-1,4-glycosidic bonds in these molecules?

A \* alpha-amylase

- B Amylo-1,6-glycosidase
- C aldolase
- D hexokinase
- E maltase

11. In a patient with complaints of poor appetite, weight loss, pain in the epigastric area, the analysis of gastric juice showed the presence of achillea. What does this term mean?

A \* Absence of free NSI and pepsin.

In the absence of free NSI

C Lack of acidity

D Absence of free and bound NSI

E Absence of gastromucoprotein

12. The patient complains of loss of appetite, weight loss, unpleasant belching, pain and heaviness in the epigastric area. Analysis of gastric juice showed: total acidity 20 mM/l, free HCI absent, bound HCI 18 mM/l, lactic acid present. What disease can be assumed?

A \*Stomach tumor

In Gastric ulcer disease

C Hyperacid gastritis

D. Hypoacidic gastritis

E Ulcerative disease of the duodenum

13. The patient notes periodic pains in the epigastric area, heartburn. When examining the gastric juice, hyperacidity was established. Which of the given data corresponds to this condition?

A \* 80 mM/l In 40 mM/l With 60 mM/l D 25 mM/l E 55 mM/l

14. A large amount of fluoride was found in the water of the river, which is the source of the city's water supply. What pathology can develop among the inhabitants of this city?

A \*Fluorosis In Thyrotoxic goiter C Juvenile current D Caries It's beri-beri

15. A patient with hypofunction of the pancreas is prescribed pancreatin. When is it most rational to take this drug?

A \*Before food In After eating C During meals D Before going to bed E Any time

16. At the final stage of protein breakdown, trace elements Zn, Mn, Mg, Co play an important role, which:

A \* They increase the activity of peptidases

Contribute to the absorption of proteins

C They reduce the activity of peptidases

D They inhibit the absorption of proteins

E Inactivate peptidases

9. The patient has impaired protein digestion in the stomach and small intestine. This process is caused by a deficiency of which enzymes?

- A Peptidase
- B Synthetase
- C Amylase
- D Lipase
- E Transferase

10. Pancreatic enzymes are secreted in the duodenum in an inactive state. Specify which enzyme activates trypsinogen.

- A Enterokinase
- B Gastrixin
- C Lipase
- D Pepsin
- E Elastase

11. The child complains of a toothache. The dentist diagnosed caries

enamel damage. The amount of which mineral substances is decreasing in the region carious damage:

- A \* Phosphorus, fluorine, calcium;
- B Sodium, calcium, potassium;
- C Potassium, phosphorus, fluorine;
- D Magnesium, fluorine, calcium;
- E Phosphorus, magnesium, potassium.

12. A woman, an employee of a confectionery shop, turned to a dentist. The patient drew attention to increased sensitivity to caries. For the purpose of remineralizing therapy, the doctor prescribed fluoride preparations. What is the role of fluoride in this therapy?

- A \*Increase in the formation of fluorine apatite;
- B Increased permeability of enamel;
- C Suppression of alcoholic fermentation;
- D Decreased synthesis of proteoglycans;
- E Activation of salivary proteases.

13. The patient is hyperacidic. Name the hormone that stimulates the secretion of HCl and pepsinogen in the stomach.

- A Gastrin
- B Insulin
- C Somatotropin
- D secretin
- E Glucagon

14. Parenteral nutrition is recommended for a patient with a damaged esophagus. Indicate which of the indicated pharmaceutical preparations is a hydrolyzate of amino acids?

- A Hydrolysin
- B Asparkam
- C Rheopolyglukin
- D Polyglukin
- E Panangin

15. To obtain amylase enzyme in its pure form from the pancreas of animals, the method of affinity chromatography with a ligand attached to the carrier is used. Which of the following substances is used as a ligand?

- A Starch
- B Cellulose

- C Lactose
- D Sucrose
- E Glucose

16. In the patient, a stone in the common bile duct blocked the flow of bile to the intestine. Indigestion of what substances is observed?

- A Zhiriv
- B Proteins
- C Water-soluble vitamins
- D Microelements
- E Carbohydrates

17. The patient was diagnosed with achlohydria. This leads to a decrease in the activity of which enzyme?

- A Pepsin
- B Trypsin
- C Chymotrypsin
- D Elastase
- E Aminopeptidases

18. The drug tannin is used in practical medicine as an astringent for acute and chronic intestinal diseases. The astringent effect of tannin is related to its ability to:

- A Denature proteins
- B Hydrolyze proteins
- C Renature proteins
- D Salt the proteins
- E Oxidize proteins

19. In the patient, as a result of the study, a violation of protein digestion in the stomach and small intestine was revealed. Lack of which enzymes leads to to such a violation?

- A Peptidases
- B Transferases
- C Amylase
- D Lipases
- E Oxidoreductases

20. Nucleosidases and nucleotidases of the gastrointestinal tract catalyze the hydrolysis of nucleic acids and mononucleotides, as well as those medicinal substances that have the following chemical bond in the molecule:

- A Phosphodiester
- B Hydrogen
- C Peptide
- D Glycoside
- E Amid

21. In children, the pH of gastric juice ranges from 4.0 to 5.0. Name the enzyme of gastric juice that is active under these conditions.

- A Renin
- B Pepsin
- C Chymotrypsin
- D Trypsin
- E Elastase

22. According to the result of the analysis of the patient's saliva, it was established that the pH is 8.0, i.e. shifted to the alkaline side. This state of saliva contributes to:

- A \* Formation of tartar;
- B Development of caries;
- C Development of fluorosis;
- D Development of tooth tissue hyperplasia;
- E Development of tooth tissue hypoplasia.

23. The patient was prescribed a bile preparation to improve the digestion of fatty food. What are the components?

Does this drug take part in the emulsification of fats?

- A Bile acids;
- B Cholesterol;
- C Diglycerides;
- D Amino acids;
- E Higher fatty acids.

24. The pancreas secretes an enzyme capable of hydrolyzing  $\alpha$ -1,4-glycosidic bonds in the glycogen molecule. Point to this enzyme.

- A α-Amylase
- B Phosphatase;
- C Enterokinase
- D Chymotrypsin
- E Lysozyme.

25. With exocrine insufficiency of the pancreas sometimes with

the drug "festal", which contains pancreatic enzymes, for improvement Bile acid preparations are recommended for digestion. For what purpose is such an additive used?

A \*For emulsifying fats

- B To activate proteolytic enzymes
- C For activation of ?-amylase
- D To stimulate the secretion of pancreatic juice
- E To stimulate intestinal peristalsis

26. Inhibition of the synthesis of bile acids from cholesterol in the experimental liver animals led to impaired digestion of lipids. What is the role of these acids in lipid digestion?

- A \*Emulsify food lipids
- B Participate in resynthesis of lipids
- C They are part of LDL
- D Maintain an alkaline environment in the intestines
- E Activate the formation of chylomicrons
- Test tasks.

27.Inactivation of vitamin PP and its coenzyme forms is carried out by methylation to Nmethylnicotinamide, which is excreted in the urine. Therefore, with the long-term appointment of vitamin PP in high doses, the following dietary recommendation is appropriate:

And \*enrichment of the diet with proteins rich in methionine

In Enriching the diet with proteins rich in tryptophan

C Enrichment of the diet with unsaturated fatty acids

D Enrichment of the diet with carbohydrates

E Sharp restriction of carbohydrates

28. Vitamin derivatives are part of the coenzymes of the respiratory chain. NAD is the coenzyme form of which vitamin?

A \*RR B B6

- C B1
- D B2
- E B3

29. Tuberculosis patients take a drug that is an anti-vitamin of nicotinic acid. Specify this substance.

- A \*Isoniazid
- B Sulfanilamide
- C Akrichin
- D Isoriboflavin
- E Oxythiamine

30. The patient complains of lack of appetite, nausea, abdominal pain, diarrhea, headache, memory impairment. Dermatitis is observed in the neck and face. Which vitamin is deficient?

- A \*Vitamin B5
- B Vitamin B1
- C Vitamin B3
- D Vitamin B2
- E Vitamin B6

31. A patient with a severe form of diarrhea, dermatitis and dementia was prescribed vitamin PP. State the role of vitamin PP in metabolism.

- A \*Participation in redox processes
- B Participation in the hydrolysis of peptide bonds
- C Participation in isomerization reactions
- D Participation in oxygen transport
- E Participation in the formation of peptide bonds

32. Determination of the activity of some transaminases is widely used in medical practice for the purpose of diagnosing damage to internal organs. The cofactor of these enzymes is the active form of the vitamin

A	*B6
В	B1
С	B2
D	B12
E	RR

33. Determination of the activity of alanine aminotransferase (AIT) and aspartate aminotransferase (AsT) is widely used in medical practice for the purpose of diagnosing damage to internal organs. The prosthetic group of these enzymes is the coenzyme form of which vitamin?

A \*B6 B B1 C WITH D B5 E R 34. Insufficiency of which vitamin causes a decrease in the activity of aminotransferases and decarboxylases?

A	*B6
В	B12
С	B2
D	B3
Е	B15

35. The patient has dry lips, cracks and "crusts" in the corners of the mouth, a bright red tongue, seborrheic dermatitis of the nasolabial folds, photophobia and conjunctivitis. What vitamin deficiency is this associated with?

- A \*Riboflavin
- B Cholecalciferol
- C Cobalamin
- D Pyridoxine
- E Ascorbic acid

36. The patient has neurasthenic syndrome, diarrhea, dermatitis. What vitamin deficiency is this associated with?

- A \*Nicotinic acid
- B Vitamin D
- C Vitamin K
- D Vitamin B12
- E Folic acid

37. A compound containing isoalloxazine was found in the patient's urine cycle. What is this compound?

- A \*Vitamin B2;
- B Vitamin B5;
- C Vitamin B6
- D Vitamin B1
- E Vitamin B3

38. In clinical practice, the drug isoniazid is used, which is competitively included in the coenzyme structure, which cannot participate in redox processes, and this leads to the cessation of growth of Koch's bacillus. Specify which enzyme systems are inhibited:

- A \*NAD-dependent enzymes
- B FAD-dependent enzymes
- C CoQ
- D Cytochrome c
- E Cytochrome a1

39. A 38-year-old patient suffering from chronic alcoholism has edema, muscle atrophy, cardiovascular insufficiency, and peripheral nerve pain. Such symptoms are caused by the lack of which vitamin in the body?

- A \*vitamin B1
- B vitamin A
- C vitamin E
- D vitamin B6
- E vitamin K

40. The patient has pain along the course of peripheral nerves. This can be caused by a lack of which vitamin?

- A \*vitamin B1
- B vitamin A
- C vitamin E
- D vitamin B12
- E vitamin K

41. Redness of the mucous membrane of the mouth, cracks in the corners and lips of the patient, peeling of the skin, dryness and inflammation of the conjunctiva on the face, sprouting of the vascular mesh into the cornea. The probable cause of this pathology is a lack of vitamin:

А	*B2
В	WITH
С	IS
D	Κ
E	D

42. Nicotinic acid amide plays an important role in metabolism. What a disease occurs with his hypovitaminosis?

- A \*Pellagra
- B Rickets
- C Anemia
- D Xerophthalmia
- E Beri-Beri

43. Pyridine-dependent dehydrogenases act as the primary hydrogen acceptor during tissue respiration. Which of the vitamins is necessary for the formation of the corresponding coenzyme (NAD +)?

- A \* Vitamin PP
- B Vitamin C
- C Vitamin B1
- D Vitamin B2
- E Vitamin B6

44. Vitamins must be included in a person's diet. Which one is used to treat pellagra?

- A \*Vitamin B5
- B Vitamin B1
- C Vitamin C
- D Vitamin A
- E Vitamin D

45. For the treatment of heart diseases, the drug cocarboxylase is used. This drug is the coenzyme form of which vitamin?

A \* B1 B B6 C B12 D WITH E R

46. Water-soluble vitamins in the body are converted into coenzyme forms. Coenzyme form of which vitamin is thiamine diphosphate (TDP)?

A \* vitamin B1

- B vitamin B2
- C vitamin C
- D vitamin B6
- E vitamin B12

47. The biochemical function of water-soluble vitamins depends on their ability to transform into coenzyme forms. What coenzyme form can vitamin B2 (riboflavin) convert into:

- A \* FMN (flavin mononucleotide)
- B NAD+ (nicotinamide adenine dinucleotide)
- C TMF (thiamine monophosphate)
- D TDF (thiamine diphosphate)
- E PALF (pyridoxal phosphate)

48. A 30-year-old man with pulmonary tuberculosis is prescribed isoniazid. Insufficiency of which vitamin can develop as a result of long-term use of this one drug?

- A \* Pyridoxine
- B Tocopherol
- C Cobalamin
- D Ergocalciferol
- E Retinol
- 9. Summing up.

10. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

### Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/

3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №28

## Topic: <u>Fat-soluble vitamins. Vitamins of groups A, D, E, K. Structure, biological</u> <u>role. Hypo- and hypervitaminosis.</u>

**Goal:**\_\_\_To study the mechanism of action and biological role of this group of vitamins. Show the possibility of using them in practical medicine.

<u>Higher education applicants should study and know the structure, biochemical characteristics</u> of fat-soluble vitamins dissolved in fats A, D, E, K, know the mechanisms of action of these vitamins and signs of hypovitaminosis and hypervitaminosis of vitamins A, D, K.

 Teach higher education applicants the qualitative determination of vitamins A, D, E, K.

 Basic concepts:
 fat-soluble vitamins, hypo- and hypervitaminosis, causes, consequences, symptoms

Equipment: Laboratory of the department

### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- know the role of each of the vitamins listed above in metabolism

The higher education applicant should be able to:

i.write the formulas of vitamins A, D, E, K;

- to qualitatively determine the vitamins in the studied sample.

-

Questions to check basic knowledge on the topic of the class:

-1. Characteristics of fat-soluble vitamins.

- -2. What are carotenoids.
- -3. The role of bile acids in the absorption of fat-soluble vitamins.
- -4. Participation of fat-soluble vitamins in metabolism.

3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "*Qualitative reactions to fat-soluble vitamins A, D, E, K* (*Vikasol*)".

### Recommendations for performing tasks.

## A. Qualitative determination of vitamin A

<u>Principle of the method:</u> If chloroform and concentrated sulfuric acid are added to fish oil containing vitamin A, a purple ring forms at the interface between the two liquids.

### The main stages of work performance.

1. Preparation of the hood:

2 drops of fish oil are dripped into the test tube, and then 5 drops of chloroform. They shake

2. Formation of a colored complex:

Add 0.5 ml of concentrated sulfuric acid to the obtained extract. Mix carefully. A purple ring appears at the interface between the two liquids, which then turns brown. The appearance of such a ring indicates that the solution contains vitamin A.

### B. Qualitative determination of vitamin D

<u>Principle of the method:</u> When vitamin D interacts with hydrochloric acid aniline, a red color is observed.

The main stages of work performance.

1. Preparation of the reaction mixture:

Pour 0.5 ml of fish oil into a dry test tube, then add 1.0 ml of aniline hydrochloric acid solution. *2. Boiling:* 

The contents of the test tube are heated to boiling with constant stirring and boiled for 30 seconds. The yellow emulsion first acquires a dirty-green, and then a brown-red color. This indicates that the solution contains vitamin D.

### **B.** Qualitative determination of vitamin **E**

<u>Principle of the method:</u> When an alcoholic solution of tocopherol reacts with concentrated nitric acid, the reaction mixture turns red.

The main stages of work performance.

Pour 0.5 ml of an alcoholic tocopherol solution into a dry test tube and add 1.0 ml of concentrated nitric acid. As a result of the reaction, a product of quinoid nature is formed, which gives a red color. This color indicates the presence of vitamin E in the solution.

### **D.** Qualitative determination of vitamin K

<u>Principle of the method:</u> If cysteine and NaOH are added to a solution containing vitamin K, the solution will acquire a lemon-yellow color.

The main stages of work performance.

Pour 0.5 ml of Vikasol into the test tube, then add 0.5 ml of cysteine and one drop of 10% NaOH solution.

In the presence of cysteine, the Vikasol solution in an alkaline environment acquires a lemonyellow color.

### Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

Control materials for the final stage of the class.

Questions to check the final level of knowledge:

1. Vitamin A, its structure, forms in the body, biological role and mechanism of action. Avitaminosis, hypervitaminosis A.

2. Vitamin D, its structure, biological role and mechanism of action on mineral metabolism. Avitaminosis, hypervitaminosis D.

3. Vitamin E, its chemical nature, biological role and mechanism of action. Hypovitaminosis E.

4. Vitamin K, its structure, biological role and mechanism of action on blood coagulation processes. Hypovitaminosis K, signs of hemophilia.

5. Hypervitaminoses. Toxic effect of large doses of vitamins.

### Test tasks.

1 The doctor recommends the patient to take Vikasol under conditions of increased risk of bleeding. What vitamin is this drug an analogue of?

A \*Vitamin K

B Vitamin A

C Vitamin B5

D Vitamin B12

E Vitamin B6

2. Administration of dicoumarol into the body causes a sharp decrease in blood clotting factors. Dicoumarol is an antivitamin of which vitamin?

A \*Vitamin K

- B Vitamin C
- C Vitamin B2
- D Vitamin E
- E Vitamin R

3. The patient suffers from thrombophlebitis. Which of the vitamins that enhances the synthesis of blood coagulation factors can provoke an exacerbation of this disease?

- A \* vitamin K
- B vitamin E
- C vitamin B2
- D vitamin D
- E vitamin B1

4. In the large intestine, microorganisms synthesize vitamins that participate in the biochemical processes of the body. What vitamins are mainly synthesized by microflora?

- A \*K, B12
- B A, S
- C E, RR
- D B1, B2
- E B6, E

5. A patient with chronic hepatitis has bleeding gums, hemorrhages in the skin even with a minor injury. These manifestations are most likely associated with a violation of the metabolism of which vitamin?

A \*K B D C IS D RR E IN

6. Vitamin D is necessary for the formation of the bone system of the fetus during intrauterine development. This vitamin is a derivative of which chemical compound?

- A Cholesterol;
- B Glycerol;
- C Sphingosine;
- D Inositol;
- E Ethanol.

7. Some vitamins ensure the stability of biological membranes. Name one of the vitamins that has this effect.

- A \* tocopherol
- B naphthoquinone
- C cholecalciferol
- D pantothenic acid

E riboflavin

8. Vitamin A quickly oxidizes in the air, which leads to the loss of biological activity. Which component of food products mainly prevents oxidation of the vitamin?

- A \*tocopherol
- B nicotinic acid
- C Table salt
- D White
- E Fat

9. Deficiency of which vitamin will most likely cause activation of lipid peroxidation processes?

- A \*Vitamin E
- B Vitamin D
- C Vitamin K
- D Vitamin B12
- E Vitamin B6

10. An elderly woman complains of impaired vision at dusk. Which of the listed vitamins should be prescribed in this case?

- A \*AND
- B WITH
- C IS
- D D
- E RR

11. The patient was hospitalized with intestinal bleeding. What drug should be included in the treatment regimen?

- A Vikasol
- B Sulfanilamide
- C Cocarboxylase
- D Aspirin
- E Riboflavin

12. The state of the patient's antioxidant system was assessed based on the determination of the content of one of the endogenous antioxidants. Which one exactly?

- A \*Alpha-tocopherol
- B Trivalent Ferum
- C Ornithine
- D Hydrogen peroxide
- E Cholecalciferol

13. The ophthalmologist found that the patient had an increase in the time it took for the eye to get used to the dark. The lack of which vitamin can be the cause of such a symptom?

- A \* vitamin A
- B vitamin C
- C vitamin K
- D vitamin B1
- E vitamin B6

14. 20 minutes after the skin cut, the woman noticed that the wound did not stop bleeding. Deficiency of which vitamin causes this condition?

- A \* vitamin K;
- B vitamin A;
- C vitamin D;
- D vitamin E;
- E vitamin B12.

15. A woman contacted a pediatrician about the ill health of her 8-month-old child: sweating, increased size of the relative, delay in teething. Which drug should be prescribed first?

- A \*Cholecalciferol;
- B Cyanocobalamin;
- C Calcium gluconate;
- D Thiamine bromide;
- E Calcium pangamate

16. The child did not receive vitamin D in a timely manner. After a while, everyone appeared

symptoms of rickets. A decrease in the activity of which blood enzyme is observed in this case?

- A \*Alkaline phosphatase;
- B Acid phosphatase;
- C Alpha amylases;
- D Choline esterase;
- E Creatine kinases.

17. During the examination of a one-year-old child, the doctor paid attention to the late eruption of teeth, their incorrect location. The lack of which vitamin can cause such a condition?

- A \*Vitamin A;
- B Vitamin C;
- C Vitamin E;
- D Vitamin D;
- E vitamin B2.

18. During the examination of the patient's oral cavity, the dentist determined dryness mucous membrane, numerous erosions. The lack of which vitamin caused these phenomena?

- A \*Vitamin A;
- B Vitamin K;
- C Vitamin P;
- D Vitamin H;
- E Vitamin PP.

19. In the last month of pregnancy, the doctor recommends that women take Vikasol according to the scheme. Which vitamin is it an analogue of?

А	*Vitamin K	

- B Vitamin B12
- C Vitamin B5
- D Vitamin B6
- E Vitamin A

20. Vitamins are the amines of life. What vitamin in the body is formed from provitamin beta-carotene?

A \*A1 B B1 C B12 D WITH

E D

21. With long-term use of the sulfonamide drug, the patient's microbial flora in the intestine was disrupted. During the examination in the polyclinic, small point hemorrhages were found on the skin. Their reason is most likely:

- A \*Deficiency of vitamin K in the body
- B Lack of vitamin C in the body
- C Enterocolitis (inflammation of the mucous membrane of the small and large intestine)
- D Liver disease
- E Hereditary defect of blood coagulation factors

22. A patient with cirrhosis of the liver is noted to have impaired vision at dusk. Which of the following is the most likely cause?

- A \* Violation of absorption of vitamin A in the intestine
- B Insufficient supply of vitamin A with food products
- C Excessive intake of vitamin A with food products
- D Violation of trans-retinal conversion into cis-retinal
- E Violation of rhodopsin synthesis

23. To stimulate the activity of the blood coagulation system, vitamin K is used. Its effect is based on participation in the process:

- A \*Carboxylation of amino acid residues of blood coagulation factors II VII, IX and X
- B Glycosylation of amino acid residues of blood coagulation factors II VII, IX and X
- C Phosphorylation of amino acid residues of blood coagulation factors II VII, IX and X
- D Deamination of amino acid residues of blood coagulation factors II VII, IX and X
- E Limited proteolysis of amino acid residues of blood coagulation factors II VII, IX and

Х

24. As a result of intoxication, the patient has impaired synthesis of beta-lipoproteins. This is accompanied by malabsorption:

- A \*Vitamin E.
- B Vitamin C.
- C Vitamin B6.
- D Vitamin H.
- E Vitamin B12.

25. The patient complains of loss of appetite, headache, bad sleep. Inflammation of the eyes, hair loss, and general exhaustion of the body were noted. It is known from the anamnesis that the patient took fish oil. What disease can be suspected?

- A \*Hypervitaminosis of vitamin D
- B Hypovitaminosis of vitamin D
- C Hypervitaminosis of vitamin A
- D Vitamin A hypovitaminosis
- E Hypervitaminosis of vitamin F

26. Dicoumarol is prescribed for the prevention of thrombosis. Antivitamin of which vitamin is it?

- A \*K B D
- C C
- D A

E RR

27. A patient with symptoms of increased blood coagulation has been taking anticoagulant drugs - coumarol derivatives for a long time. As a result, the patient developed signs of bleeding. The appointment of which vitamin will quickly and effectively eliminate unwanted complications?

A \*K B AND C D D WITH E IS

28. Under the influence of ultraviolet rays and ionizing radiation, active forms of oxygen are formed in the body. Substances exhibiting antioxidant properties are used to stabilize redox reactions. Specify them.

- A \*Vitamin E
- B Vitamin B12
- C Vitamin B2
- D Vitamin B6
- E Vitamin B1

29. A 35-year-old patient was prescribed vikasol (a synthetic analogue of vitamin K) in the preoperative period. What is the mechanism of antihemorrhagic action of this drug?

- A \* Stimulation of prothrombin synthesis
- B Activation of plasminogen
- C Stimulation of tissue thromboplastin synthesis
- D Activation of the Hageman factor (XII)
- E Activation of the complement system

30. Antivitamins are substances of various structures that limit the use of vitamins in the body and have the opposite effect. Specify among the listed substances the anti-vitamin of vitamin K:

- A \*Dicumarol
- B Deoxypyridoxine
- C Aminopterin
- D Sulfapyridazine
- E Isoniazid

31. A certain vitamin is necessary for the normal development and functioning of the reproductive system in humans. Specify it.

A	*IS
В	WITH
С	Ν
D	D
E	AND

32. The process of blood coagulation cannot normally be carried out without the participation of certain vitamins. What vitamin is involved in this process?

- A \*vitamin K
- B vitamin B6
- C vitamin C
- D vitamin E
- E vitamin D

33. The child was given vitamin D in a dose of 50 mg/day to prevent rickets, which led to the appearance of signs of vitamin intoxication. Select the signs of hypervitaminosis D

- A \* Bone demineralization
- B "Chicken Blindness"
- C Anemia
- D Violation of blood coagulation
- E Gout

34. Vitamins regulate various biochemical processes. What vitamin ensures the conversion of prothrombin to thrombin?

- A \*Vitamin K
- B Vitamin A
- C Vitamin E
- D Vitamin B1
- E Vitamin D

35. A man suffering from enterocolitis was treated for a long time with the sulfonamide drug Phthalazol. At the last examination in the polyclinic, small point hemorrhages were found on the patient's skin. Their reason is most likely:

A \* lack of vitamin K in the body

Insufficiency of vitamin C in the body

C Enterocolitis (inflammation of the mucous membrane of the small intestine)

D Liver disease

E Hereditary defect of blood coagulation factors

36. The blood coagulation process normally cannot be carried out without the presence of certain vitamins. What vitamin is involved in this process?

And \*vitamin K B vitamin B6 C vitamin C D vitamin E And vitamin D

4. Summary:

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

#### Practical class №29

Topic: Biochemical characteristics and functions of blood. Biochemical composition of blood plasma. Characteristics of protein fractions of blood. Characteristics of non-protein substances of blood plasma. Residual blood nitrogen, its components. Diagnostic value of determination of residual blood nitrogen. Respiratory function of blood. Hemoglobin, structure, synthesis in the body. Role in the transport of oxygen and carbon dioxide.

**Goal:** To study theoretical material on blood biochemistry: fractions of plasma and serum proteins, classification of enzymes and their value for differential diagnosis of pathology, acute phase proteins and the value of their determination in clinical diagnosis. Be able to determine the total protein in blood serum by the biuret method and explain the diagnostic value of quantitative determination of protein in blood serum. Learn the structure and functions of hemoglobin. To study the mechanism of gas transportation by blood.

Basic concepts: <u>hemoglobin, cooperative effect, Bohr effect, hemoglobinosis, thalassemia,</u> <u>porphyria</u>

Equipment: Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

-functions and composition of blood;

- -components of residual blood nitrogen;
- -structure and function of lipoproteins;
- -the role of lipoproteins in the development of atherosclerosis;

- mechanisms of regulation of the acid-base state with the participation of individual organs and types of violations and mechanisms of compensation of the acid-base state;

- the structure of hemoglobin and its synthesis in the human body. Abnormal types of hemoglobin;

- mechanisms of hemoglobin participation in the transport of oxygen and carbon dioxide;

- the main breakdown products of hemoglobin.

The higher education applicant should be able to:

- evaluate the main biochemical indicators of blood composition in healthy people and in a number of diseases;

- to quantitatively determine the total protein of the blood plasma, individual fractions of proteins and to know their biochemical characteristics and biological significance for the body;

- - determine the hemoglobin content in the blood.

Questions to check basic knowledge on the topic of the class:

1. Comparative characteristics of the chemical composition of plasma and blood serum in the norm.

2. Biochemical functions of blood proteins. Characteristics of the main fractions of blood plasma proteins (albumin,  $\alpha$ -,  $\beta$ -,  $\gamma$ -globulins). The concept of hypo-, hyper-, para- and dysproteinemia. Dyslipoproteinemia.

3. Clinical and biochemical characteristics of proteins of the acute phase of inflammation.

4. Classification of blood plasma enzymes, their use in the diagnosis of diseases.

5. Blood buffer systems. Violation of acid-base balance: types of acidosis and alkalosis, mechanisms of their occurrence.

6. The main organic non-protein nitrogen-containing and nitrogen-free components of plasma, characteristics and significance of determination in pathology.

7. General ideas about the mineral composition of blood plasma in normal and pathological conditions.

8. Composition and properties of erythrocytes:

9. What is the number of erythrocytes in the blood, does it depend on gender and life requirements;

10. Name the structural and functional features of erythrocytes;

11. List the metabolic features of erythrocytes;

12. What is the partial pressure of carbon dioxide in venous blood and tissues?

3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Quantitative determination of blood proteins by the biuret method".

Recommendations for performing tasks.

The principle of the method is that alkaline solutions of proteins and peptides, thanks to peptide bonds, become colored like biuret when a solution of copper sulfate is added.

Progress. To 0.1 ml of serum, add 8 ml of a 4.8% solution of lye, 3 ml of a 20% solution of copper sulfate. Centrifuge at 3000 rpm. 5 minutes. Colorimeter on FEK with a green light filter against the mixture in which the serum is replaced by water.

A red-violet color appears, the intensity of which is directly proportional to the amount of protein. Multiply the optical density by 12 to find a quantitative representation of the protein content (%).

*Requirements for work results.* 

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.2 Demonstration and practical work "Refractometric determination of protein in serum". *Recommendations for performing tasks.* 

The principle of the method consists in the protein's ability to increase the refractive index of light passing through the solution.

Progress. Place 2-3 drops of water between the prisms and install a zero shunt on the 1.322 section of the light refraction scale. Place 2-3 drops of serum instead of the eyepiece, move the handle of the eyepiece along the scale until the viewing scale aligns with the border of the dark and light parts of the field of vision. According to the table The flight determines the appropriate amount of protein.

Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

3.3 Demonstration and practical work "Fractionation of blood serum proteins by the method of salting out".

#### Recommendations for performing tasks.

The principle of the method is the ability of proteins to coagulate at different concentrations of salts, depending on the molecular weight of the proteins.

Progress. Add an equal volume of saturated ammonium sulfate solution to 3 ml of serum. A precipitate of globulins falls out. They filter. Ammonium sulfate is added to the filtrate until saturation. A precipitate of albumin falls out. The precipitate of albumins and globulins is dissolved in 4 ml of water, a biuret reaction is carried out and the content of globulins and albumins is determined.

### Medical and biological evaluation of the obtained results

Hypoproteinemia can be caused by a lack of protein in the diet, a violation of the processes of digestion and absorption of proteins, a violation of protein synthesis (for example, when the liver is damaged), protein loss during acute and chronic bleeding, kidney damage.

Hyperproteinemia can be absolute - with an increase in the protein content in the blood plasma - for example, an increase in the number of  $\gamma$ -globulins in infectious diseases, the appearance of abnormal proteins in the blood in myeloma, in macroglobulinemia. Loss of water in the body (diarrhea, vomiting, extensive burns) can lead to an increase in plasma protein, that is, to relative hyperproteinemia.

### Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions.

3.4.Demonstration and practical work «Determination of the hemoglobin content in the blood". *Recommendations for performing tasks.* 

The principle of the method consists in transforming blood hemoglobin into hydrochloric acid hematin and comparing the obtained color with the standard available in the device. Sali's hemometer is used for determination. It consists of two sealed tubes with a standard colored liquid (1% solution of hematin hydrochloride in glycerol) containing 16.67 g% hemoglobin (16.67 g per 100 ml of blood). Between them is a graduated test tube with two scales. One - with distributions from 0 to 23 is used to determine hemoglobin in grams per 100 ml of blood, that is, in grams as a percentage; another scale with divisions from 0 to 140 shows hemoglobin units (hemoglobin percentage).

Progress. A 0.1N solution of hydrochloric acid is poured into a graduated test tube located in the middle slot up to the beginning of the scale. Then, from the injection site on the flesh of the finger, Sally collects blood up to the mark of 0.02 ml (20 mm3) with a pipette, sucking it through the mouth through a rubber tube with a glass mouthpiece attached to the upper end of the pipette. The tip of the pipette is wiped from the blood and lowered into a test tube with hydrochloric acid, carefully blowing out the contents so that air bubbles do not form. Hitting the bottom of the test tube with a finger, thoroughly mix the blood and leave it for 5 minutes. for the formation of hydrochloric acid hematin. During this time, blood is collected for another part of the analysis. After this time, distilled water is added to the test tube drop by drop, stirring with a glass rod until the color of the solution of the tested blood completely equals the color of the standard liquid.

### Medical and biological evaluation of the obtained results

Normally, the hemoglobin content in gram percentages in men varies from 13.3 to 18 g%, in women - from 11.7 to 15.8 g% (on average 13.7); in units (percentages) in men - from 80 to 108 units, in women - from 70 to 95 units.

#### Requirements for work results.

Enter the obtained data and calculations into the workbook. Make medical and biological conclusions. Control materials for the final stage of the class.

Questions to check the final level of knowledge:

- 1. List and explain the main functions of blood.
- 2. Chemical composition of blood plasma.

3. What is the difference between plasma and blood serum? Methods of obtaining them.

4. Specify the structure of the protein fractions of the blood and their role in the vital activity of the body.

5. Pathological conditions associated with quantitative and qualitative changes in blood plasma proteins.

- 6. Blood plasma enzymes. Diagnostic value of determination of blood plasma enzymes.
- 7. Non-protein organic blood compounds.
- 8. Nitrogen-containing compounds. Residual (rest-nitrogen) blood plasma. Its components.
- 9. Methods of determining the level of residual blood nitrogen.
- 10. Hyperazotemia. Types and causes of occurrence.
- 11. Nitrogen-free compounds.
- 12. Inorganic components of blood plasma.
- 13. What is the structure of hemoglobin?
- 14. What is the structure of the protein part of hemoglobin A?
- 15. What types of hemoglobin can be found in the body?
- 16. What is the peculiarity of abnormal hemoglobins?
- 17. What is the heme molecule synthesized from and what role do vitamins play in this?
- 18. How does hemoglobin carry oxygen?
- 19. How is the transfer of carbon dioxide by hemoglobin?
- 20. What is the Bohr effect and the cooperative effect of oxygen?
- 21. Oxyhemoglobin dissociation curve.
- 22. What role does 2,3-bisphosphoglycerate play in supplying tissues with oxygen?
- 23. Why is a mixture of carbon dioxide and not pure oxygen used in respiratory mixtures used in the clinic?
  - 24. What role does methemoglobin play in tissues?
  - 25. Why is hyperbolic oxygenation necessary for carbon monoxide poisoning?

Test tasks.

Plasma includes mineral salts. What physical and chemical properties of blood are due to their presence?

And osmotic pressure In oncotic pressure With an active blood reaction D blood viscosity E SHOE

The buffer systems of the blood support the constancy of its acid-alkaline balance. Which substances of one of the buffer systems have amphoteric properties?

And plasma proteins In the bicarbonate system C hemoglobin D phosphate system E electrolytes

Cyanides are strong poisons for the human body. Indicate which compound will be the best to connect them?

- A Methemoglobin
- B Carboxyhemoglobin
- C Carbhemoglobin
- D Oxyhemoglobin
- E Hem

A diabetic patient has a hyperglycemic coma as a result of long-term elevated blood glucose. What is the most likely mechanism for the development of such a condition?

And \* Ketonemia. A change in blood pH and, as a result, a decrease in the affinity of Hb to O2. B Increased BBB to glucose.

Coma due to high glucose content in brain neurons

D Decrease in neurocirculation

E Acidosis. Methemoglobinemia due to a change in the valence of Fe under the influence of a high concentration of glucose and the formation of a stable compound Hb with O2.

Liver diseases (hepatitis, cirrhosis, tumor) lead to all the following disorders of protein metabolism except:

A \* Hyper-alpha2-globulinemia In hypoalbuminemia With Hemorrhage D Hyperaminoacidemia E of Azotemia

A number of biochemical tests (samples) are used to diagnose liver diseases. Which of the following pathological conditions is most likely indicated by an increase in the concentration of alpha-fetoprotein in the blood plasma.

A \* Liver cancer In Cirrhosis of the liver C Viral hepatitis D Cholestasis E Fatty infiltration of the liver

Which of the following conditions develops when a large volume of  $5\$  glucose solution is introduced.

A \* Hypoosmolar hyperhydration

B. Hypoosmolar dehydration

C Isoosmolar dehydration

D. Hyperosmolar dehydration

E Hyperosmolar hyperhydration

All of the pathological conditions listed below can be accompanied by hyperosmolar dehydration except:

A \*Burns In Hyperaldosteronism C Heart failure D Jade E Diabetes

The formation of carbonic acid from CO2 occurs in the presence of the enzyme carbonic anhydrase. Where does this process take place?

And in erythrocytes In leukocytes C in plasma proteins D in platelets E in plasma For the prevention and therapy of radiation sickness, among various radioprotectors, sodium nitrite is used, which causes the development of hypoxia. What is the mechanism of its hypoxic action:

A \* Stimulates the formation of methemoglobin

Stimulates the breakdown of hemoglobin

C Inhibits the dissociation of oxyhemoglobin

D Inhibits the activity of tissue respiration enzymes

E Activates lipid peroxidation

A 40-year-old man was hospitalized due to carbon monoxide poisoning. Which of the listed fractions of hemoglobin will be elevated in this patient?

- A Carboxyhemoglobin
- B Methemoglobin
- C Carbhemoglobin
- D Oxyhemoglobin
- E Glycosylated hemoglobin

Hemoglobin shows the ability to form a very strong compound with carbon monoxide, which is dangerous to life. What is the name of this compound?

- A carboxyhemoglobin
- B methemoglobin
- C carbhemoglobin
- D oxyhemoglobin
- E myoglobin

Some proteins in the body have buffering properties. Due to the content of which amino acid does hemoglobin show its buffering properties?

- A Histidine
- B Valin
- C Isoleucine
- D Threonine
- E Alanine

Nitrogen oxides can oxidize Fe2+ in the hemoglobin molecule to Fe3+ with the formation of its derivative, which cannot attach oxygen. Name this substance:

- A Methemoglobin
- B Carboxyhemoglobin
- C Carbhemoglobin
- D Oxyhemoglobin
- E Deoxyhemoglobin

Hemoglobin of red blood cells binds and carries oxygen from the lungs to the tissues. What level of structural organization of hemoglobin ensures the respiratory function of blood:

- A quaternary
- B tertiary
- C secondary
- D primary
- E Does not matter

Sickle-cell anemia is caused by a mutation of the gene responsible for the synthesis of the protein part of hemoglobin. At the same time, a polar amino acid is replaced by a non-polar one,

which leads to a decrease in the solubility of hemoglobin and a change in the solubility of erythrocytes. Indicate what substitution takes place in the hemoglobin molecule?

- A Glutamic acid on valine
- B Alanine to phenylalanine
- C Glutamic acid to aspartic acid
- D Valine on serine
- E Glutamic acid on lysine

The concentration of carbon monoxide in the air increased due to non-compliance with safety technology rules. Which type of hemoglobin will this lead to?

- A Methemoglobin
- B Carboxyhemoglobin
- C Oxyhemoglobin
- D Cyanohemoglobin
- E Carbhemoglobin

Many proteins have a quaternary structure, that is, they consist of several polypeptides chains Name one of these proteins.

- A \* hemoglobin
- B myoglobin
- C albumin
- D elastin
- E prealbumin

Hemoglobin is a complex protein that transports oxygen to and from the body carbon dioxide from it. Indicate to which class of substances it belongs.

- A \* Chromoproteins
- B Nucleoproteins
- C Metalloproteins
- D Lipoproteins
- E Glycoproteins
- 4. Summing up.

5. List of recommended literature (main, additional, electronic information resources):

## Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

# Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.
2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

- 1. https://info.odmu.edu.ua/chair/biology/-
- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №30

Topic: Biochemistry of coagulation, anticoagulation and fibrinolytic blood systems. Functional and biochemical characteristics of the homeostasis system in the human body: coagulation and vascular-platelet hemostasis. Blood coagulation system, characteristics of individual components (coagulation factors). Mechanisms of coagulation. Blood clotting system, anticoagulants. The role of vitamin K in coagulation reactions. Hereditary disorders of the blood coagulation system.

**Goal:** to study the mechanisms of blood coagulation and anticoagulation, disorders in the functioning of these processes. evaluate the molecular structure of coagulants and anticoagulants, the mechanisms of hemophilia of various genesis

**Basic concepts:**<u>blood coagulation system, external coagulation pathway, internal coagulation</u> <u>pathway, anticoagulation system of blood, fibrinolytic system of blood, anticoagulants, hemophilia,</u> <u>CVD syndrome</u>

Equipment: Laboratory of the department

#### Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- molecular mechanisms of blood coagulation and anticoagulation systems, the role of the liver in this process.

- molecular structure of coagulants and anticoagulants, mechanisms of hemophilia of various genesis.

The higher education applicant should be able to:

- determine biochemical indicators of the state of the coagulation system and give them a medical and biological assessment.

Questions to check basic knowledge on the topic of the class:

1. Composition and properties of erythrocytes and platelets:

2. Chemical composition of blood plasma;

3. Physico-chemical properties of proteins;

4. Glycosaminoglycans. Their structure and functions;

5. Blood plasma enzymes.

3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Determination of the amount of fibrinogen in blood plasma".

Recommendations for performing tasks.

Progress. 0.1 ml of 5% calcium chloride solution is added to 1 ml of clear plasma. Fibrin is wound on a stick, dried with filter paper and weighed. The weight of fibrin is multiplied by a factor of 22.2 and expressed in mg%. Normally, the amount of fibrinogen in blood plasma is 200-400 mg%.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.2 Demonstration and practical work "Determination of prothrombin time".

Recommendations for performing tasks.

The principle of the method. With excess thromboplastin and optimal calcium content, the time of clot formation in plasma depends on the activity of II, VII, IX, X factors.

Progress. Add 0.1 ml of 1% thrombolastin solution to 0.1 ml of plasma, incubate for 1 minute and add 0.1 ml of 0.025 M calcium chloride solution, turn on the stopwatch until a dense clot forms. A dense clot is formed. Time is expressed in seconds.

$$PAK = \frac{A}{B} x 100\%$$

A - the time of a healthy person (20 seconds)
In - the time of a sick person *Requirements for work results.*Enter the obtained data and calculations into the workbook.
Make medical and biological conclusions.

3.3 Demonstration and practical work "Definition of plasma recalcification".

Recommendations for performing tasks.

Progress. Blood is collected in a centrifuge tube with 1.34% sodium oxalate in a ratio of 9:1 and centrifuged for 10 minutes. at 1500 rpm. 0.2 ml of 0.025 M calcium chloride and 0.1 ml of physiological solution are mixed in a test tube and placed in a water bath. Then add 0.1 ml of plasma and start the stopwatch. Clotting is complete if blood does not flow when the tube is inverted. Normally, the recalcification time is 60-120 seconds. when fibrin threads appear.

Medical and biological evaluation of the obtained results

Violations of coagulation hemostasis can be genetically determined and acquired. Plasma recalcification time increases in hemophilia A, Hagemann's defect, factor VII deficiency. Is prothrombin activity reduced in hemophilia B, factor VII deficiency? V, X. With hypofibrinogenemia, all coagulation tests are sharply increased. In case of enteropathy and damaged liver, hypovitaminosis of vitamin K, deficiency of factors I, II, IX, X, XI, XII is possible.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Questions to check the final level of knowledge:

- 1. What are the phases of blood coagulation?
- 2. Where are blood coagulation factors located and how are they affected?
- 3. How many blood clotting factors found in plasma and erythrocytes have been studied?
- 4. What is the process of autocatalysis on the example of activation of coagulation factors?
- 5. What factors of plasma and platelets take part in the 1st phase of blood coagulation?
- 6. What factors of plasma and platelets take part in the II phase of blood coagulation?
- 7. What factors of plasma and platelets take part in the III phase of blood coagulation?

8. The absence of which factors causes hemophilia A and which stages of coagulation are disrupted?

9. The absence of which factors causes hemophilia B and which stages of coagulation are disrupted?

10. Why is Vikasol administered to patients with bleeding?

11. What systems make up the anti-coagulation system?

12. What is the mechanism of action of heparin?

13. How is the liquid state of the blood in the vessels ensured?

14. What antivitamins are used to strengthen the anticoagulation system and their mechanism of action?

15. Medicines affecting fibrinolysis processes.

Test tasks.

A patient with thrombophlebitis is prescribed complex therapy, which affects various stages of thrombus formation. Which of the mentioned means helps

restoration of vascular patency?

A. Neodicumarin

B. Fibrinolysin

C. Acetylsalicylic acid

D. Dipyridamole

E. Heparin

A patient suffering from a streptococcal infection developed hemorrhagic diathesis. What is the cause of increased bleeding?

A Increased fibrinolysis

Lack of vitamin A

C Increase in the amount of kallikrein in the blood plasma

D Increase in the amount of heparin in the blood plasma

E Lack of vitamin C

Heparin is used to prevent blood clotting. What class of complex proteins does it belong to? A Glycoprotein

In Metalloprotein

C Hemoprotein

D Lipoprotein

E Phosphoprotein

In a patient with a streptococcal infection, diffuse bleeding was observed after tooth extraction, which is a consequence of:

A Activation of fibrinolysis In Violation of the coagulation system With hypovitaminosis of vitamin K D Insufficiency of anticoagulants

E Violation of calcium metabolism

4. Summing up.

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

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6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

## Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

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- 3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №31

Topic: Biochemical functions of the liver, its role in the metabolism of carbohydrates, lipids, and proteins. Hemoglobin breakdown. The role of the liver in the exchange of bile pigments. Pathobiochemistry of jaundice, hereditary (enzymatic) jaundice. Detoxification function of the liver: biotransformation of xenobiotics and endogenous toxins. Types of reactions of biotransformations of foreign chemical compounds. Reaction of microsomal oxidation, inducers and inhibitors of microsomal monooxidases. Conjugation reactions in hepatocytes: biochemical mechanisms, functional significance.

**Goal:** to study the main biochemical functions of the liver, its role in the exchange of proteins, carbohydrates, and lipids; to learn the main stages of the breakdown of hemoglobin in the body with the formation of bile pigments, to be able to carry out quantitative and qualitative determination of bilirubin in serum and give it a medical and biological assessment. To interpret the mechanisms of biotransformation of xenobiotics and endogenous toxins. To study theoretical material on the biochemistry of the immune system.

**Basic concepts:**<u>total bilirubin, conjugated bilirubin, unconjugated bilirubin, UDF-</u><u>glucuronyltransferase, hemolytic jaundice, parenchymal jaundice, obstructive jaundice;microsomal</u><u>oxidation, cytochrome P-450, animal indican, hippuric acid, immunoglobulins, cytokines</u>

Equipment: Laboratory of the department

## Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

- the main biochemical functions of the liver, its role in the exchange of proteins, carbohydrates, and lipids;

- the main stages of the breakdown of hemoglobin in the body with the formation of bile pigments;

- the main types of jaundice;

- the main mechanisms of neutralization of xenobiotics and endogenous toxins;

- cellular and biochemical organization of the immune system; mediators and hormones of the immune system;

- biochemical components and mechanisms of complement system activation; biochemical

mechanisms of immunodeficiency states.

The higher education applicant should be able to:

- carry out quantitative and qualitative determination of bilirubin in serum and give it a medical and biological assessment;

- determine hippuric acid and indican in urine;
- assess the state of the body's immune system.

Questions to check basic knowledge on the topic of the class:

1. List and explain the main functions of the liver.

2. Microsomal oxidation: cytochrome P-450; molecular organization of the electron transport chain. Biological significance.

3. Formation of peroxides. Antioxidant systems.

- 4. Structure and functions of blood plasma  $\gamma$ -globulins.
- 5. Formed elements of blood. Structure and functions.
- 6. Anatomical and physiological features of the lymphoid system.
- 7. The main ways of metabolism of carbohydrates, lipids and proteins;
- 8. Structural and functional features of erythrocytes;
- 9. List the metabolic features of erythrocytes;

10. The structure of hemoglobin and its synthesis in the human body. Abnormal types of hemoglobin.

3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Determination of total, direct and indirect bilirubin in blood serum".

Recommendations for performing tasks.

The principle of the method is that the direct bilirubin of the blood serum gives a pink color when the diazo reagent is added; indirect bilirubin reacts with the diazo reagent only after adding caffeine. The color intensity is proportional to the amount of bilirubin.

Progress. a) determination of total bilirubin: add 3.5 ml of caffeine reagent and 0.5 ml of diazo reagent to 1 ml of serum - a pink color appears, after 5 minutes the resulting mixture is placed in a SF cuvette with a layer thickness of 1 cm with a green light filter against the compensation solution , containing 1 ml of serum, 3.5 ml of caffeine solution and 0.5 ml of water.

Calculation: C = 6.34xE - 0.05 (C - bilirubin concentration)

b) determination of direct bilirubin: to 1 ml of serum add 3.5 ml of physiological solution and 0.5 ml of diazo reagent and 0.5 ml of diazo reagent - a pink color appears, the following stages are similar to point a)

c) the concentration of indirect bilirubin is determined by the difference between total and direct bilirubin.

Medical and biological evaluation of the obtained results

An increase in indirect bilirubin in the blood is observed with hepatic jaundice - hemolytic anemias of various etiologies, as well as posthepatic hyperbilirubinemia, jaundice of newborns.

With mechanical jaundice, direct bilirubin is increased in the blood. In parenchymal jaundice, an increase in both direct and indirect bilirubin is observed.

## Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.2 Demonstration and practical work "Determination of hippuric acid and indican in urine". *Recommendations for performing tasks.* 

The principle of the method is that alkaline solutions of proteins and peptides, thanks to peptide bonds, become colored like biuret when a solution of copper sulfate is added.

Progress. To 0.1 ml of serum, add 8 ml of a 4.8% solution of lye, 3 ml of a 20% solution of copper sulfate. Centrifuge at 3000 rpm. 5 minutes. Colorimeter on FEK with a green light filter against the mixture in which the serum is replaced by water.

A red-violet color appears, the intensity of which is directly proportional to the amount of protein. Multiply the optical density by 12 to find a quantitative representation of the protein content (%).

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.3 Demonstration and practical work "Determination of indican in urine".

Recommendations for performing tasks.

The principle of the method consists in the transformation of indican into indoxysulphuric acid and its subsequent oxidation (with iron chloride or potassium permanganate) to blue or red indigo.

Progress. Mix 8-10 ml of urine with an equal volume of hydrochloric acid, add 1-2 ml of chloroform and 1-2 drops of potassium permanganate. Close the test tube and invert several times without shaking.

In the presence of indican, chloroform turns blue or pink.

Note: In the presence of iodide salts in the urine, chloroform also gives a pink color. In this case, a hyposulfite crystal is added. The disappearance of the pink color of chloroform indicates the presence of iodide salts. In the presence of indican, the pink color does not disappear.

Medical and biological evaluation of the obtained results

In normal urine, indican is contained in a small amount, which is not detected by ordinary quality tests. Turkey anuria occurs with intense decay of protein substances in the large intestine (colitis, colon abscess, peritonitis, constipation), as well as with increased breakdown of proteins in the body (tumor, emphysema, abscesses, pulmonary tuberculosis).

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Questions to check the final level of knowledge:

1. List and explain the main functions of the liver.

2. What role does the liver play in carbohydrate metabolism?

3. What role does the liver play in lipid metabolism?

4. What role does the liver play in protein metabolism?

5. What are the main stages of hemoglobin breakdown?

6. What forms of bilirubin are formed when hemoglobin breaks down?

7. What indicators of pigment metabolism change:

a) with hemolytic jaundice?

b) with parenchymal jaundice?

c) with obstructive jaundice?

8. What are Gelber, Dubin-Johnson and Crigler-Nayar syndromes related to?

9. Diagnostic value of determination of bound and free bilirubin in jaundice of various etiology.

10. List the compounds that have adverse, toxic effects on both individual cells and the higher organism as a whole.

11. Name the types of biotransformation reactions of xenobiotics and endogenous toxins.

12. What role do microsomal and peroxidation reactions play in detoxification of toxic substances?

thirteen. What role do conjugation reactions play in the detoxification of xenobiotics?

14. Name the most common conjugation reactions?

15. Name the main classes of lymphocytes. Their structure and functions.

16. Name the main factors of the non-specific immune system.

17. Name the main classes of immunoglobulins. Their structure and functions.

18. The main classes and biological role of cytokines.

19. Molecular mechanisms of antiviral action of interferons.

20. Tumor necrosis factors, colony-stimulating factors and transforming growth factors. Their biological role.

21. Biological organization and ways of activation of the complement system.

22. Mechanisms of impaired functioning of the human immune system. Primary and secondary immunodeficiencies.

Test tasks.

1. A patient with signs of acute poisoning was admitted to the hospital. A high content of methemoglobin was found in the blood. Which of the following compounds led to this:

A. - Lead salts

B. - Alkaloids

C. - Tetrachloromethane

+D. - Nitrates

E. - Radionuclides

2. A child born prematurely has hypoglycemia in the first days due to:

+A. - Deficiency of gluconeogenesis enzymes

B. - Violation of glycogen synthesis

C. - Violation of glycolysis

D. - Hyperinsulinemia

E. - Uncoupling of tissue respiration and oxidative phosphorylation

3. A 56-year-old patient complains of general weakness, nausea, and poor appetite. Yellowish skin, hyperbilirubinemia (direct bilirubin), light, foamy urine, acholic stools are noted. For which condition are these changes most characteristic?

A. - Dubin-Johnson syndrome

B. - Parenchymal jaundice

+C. - Obstructive jaundice

D. - Hemolytic jaundice

E. - Gilbert's disease

4. Patient F., 44 years old, notes pain in the right hypochondrium after minor physical exertion, sometimes at rest, periodic nausea, loss of appetite, swelling of the legs and trunk. During a biochemical blood test: total bilirubin 88.4  $\mu$ M/l, indirect 58  $\mu$ M/l, direct 30.4  $\mu$ M/l, AlAT - 22.4  $\mu$ mol/h-/ml, AsAT-14.7  $\mu$ mol/h/ml, total protein 35 g/l, albumins 15 g/l, globulins 20 g/l, K-2.2 mM/l, Na-1-8 mM/l. The patient probably has:

A. - Hepatic jaundice

+B. - Cirrhosis

C. - Obstructive jaundice

D. - Chronic pancreatitis

E. - Chronic cholecystitis

5. A patient was admitted to the infectious department with complaints of itching, jaundice of the skin and mucous membranes. Laboratory: increased concentration of bilirubin in blood serum due to direct, acholic stool. Determining the activity of which serum enzyme will allow to confirm the clinical signs of cholestasis in this patient?

A. - Aspartate aminotransferase

B. - Creatine phosphokinase

C. - Lactate dehydrogenase

D. - Alpha amylase

+E. - Gamma-glutamyltranspeptidase

6. The baby has an unstable light yellow color of the skin, icteric sclera. Laboratory: anemia is not determined, hyperbilirubinemia, mainly due to the fraction of unconjugated bilirubin. Diagnosis: Gilbert's syndrome. Which enzyme defect is one of the causes of this pathology?

A. - Glycogen synthases

B. - Alanylaminotransferases

C. - Glucose-6-phosphatases

+D. - UDP-glucuronyltransferase

E. - Biliverdin reductases

7. A 52-year-old female patient has been bothered by attacks of pain in the right hypochondrium for the past few days after eating fatty food. Yellowing of sclera and skin, acholic stool, "beer-colored" urine is visually determined. The presence of which substance in the patient's urine caused the dark color of the urine in obstructive jaundice?

A. - Ketone bodies

- +B. Bilirubin glucuronides
- C. Urobilin
- D. Glucose
- E. Stercobilin

8. In a patient with liver disease, there is no urobilinogen in the urine in the presence of bilirubin, this is due to a violation of:

+A. - The influx of bile into the intestines

- B. Formation of direct bioirubin
- C. Kidney function
- D. Conversion of bilirubin in the intestine
- E. Formation of stercobilin

9. Wilson-Konovalov disease (hepatocerebral degeneration) is accompanied by a decrease in the concentration of free copper in blood serum, as well as the level of:

A. - Transferrin

B. - Albumin

- +C. Ceruloplasmin
- D. C-reactive protein

E. – Fibrinogen

10. Indicate which of the following biochemical blood parameters is most important for confirming the diagnosis of liver cirrhosis?

- +A. Hypoalbuminemia
- B. Hypercholesterolemia
- C. Hyperglycemia

D. - Hypoglycemia

E. – Hyperglobulinemia

11. Yellowing of the skin is observed in a newborn. Specify the blood index, the increase of which led to such a condition:

A. - Creatine

B. - Urea

C. - Direct bilirubin

D. - Uric acid

+E. – Indirect bilirubin

12. Hyperbilirubinemia with an increase in the conjugated form was found in the patient. The thymol test is normal, a slight increase in the activity of alanine aminotransferase is noted. Choose a possible diagnosis.

A. - Viral hepatitis

- +B. Mechanical jaundice
- C. Hemolytic jaundice
- D. Acute cholecystitis
- E. Polyarthritis

13. In jaundice, the content of total bilirubin in the blood is increased due to indirect bilirubin, in the feces and urine there is a high content of stercobilin. Name the type of jaundice.

- A. Jaundice of newborns
- B. Biliary
- C. Mechanical
- D. Hemolytic
- +E. Parenchymatous

14. In which jaundice hyperbilirubinemia is not accompanied by bilirubinuria?

- A. Terminal
- B. Parenchymatous
- C. Obturational
- +D. Hemolytic
- E. Mixed

15. With hemolytic jaundice, the level of direct bilirubin:

- +A. It is growing
- B. Does not change
- C. Decreases
- D. Not defined
- E. It fluctuates

Neutralization of toxic substances and inactivation of biologically active substances in hepatocytes is carried out by various reactions. Sulfonamides are converted by which of the following reactions?

A \* Acetylation In Demining C Conjugation with glucuronic acid D Oxidation E Conjugation with glycine

Drug metabolism in hepatocytes is carried out mainly: A \*In the endoplasmic reticulum In the plasma membrane C In the core D In mitochondria E In lysosomes In the process of microsomal oxidation, binding and transformation of the substrate is carried out by:

A \* Cytochrome P450 In Flavoprotein C Iron-containing non-heme protein D NADF E OVER

Neutralization of toxic and inactivation of biologically active substances in hepatocytes is carried out in different ways. How is benzoic acid neutralized?

- A Conjugation with glycine
- B Restoration
- C Methylation
- D Oxidation
- E Acetylation

In a boy with intestinal obstruction, the urinary excretion of indican, which is formed in the liver as a result of the reaction of conjugation of indoxyl with:

A with phosphoadenosine phosphosulfate

- B Glycine
- C Glutathione
- D Acetyl-Co A
- E Taurine

Neutralization of toxic substances and inactivation of biologically active substances in hepatocytes is carried out by various reactions. Barbiturates are converted by which of the following reactions?

- A Oxidation
- B Deamination
- C Acetylation
- D Restoration
- E Conjugation with glycine
- 4. Summing up.

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

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5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

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Electronic information resources:

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- 3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №32

Topic: Biochemistry of muscles. Features of the chemical composition and metabolism in muscles. Molecular mechanisms of muscle contraction. Bioenergetics of muscle tissue: sources of ATP in muscles.Features of the biochemical composition and metabolism of the nervous system. Biochemical composition of the brain. Energy metabolism of the human brain, the value of aerobic oxidation of glucose. Neurotransmitters: acetylcholine, norepinephrine, dopamine, serotonin. Molecular basis of bioelectrical processes on neuron membranes.

Goal: Master the featureand metabolism in muscle and nerve tissues.

**Basic concepts:** <u>sarcomere, actin, myosin, tropomyosin, troponin, creatine phosphate,</u> <u>adenylate kinase, biogenic amines, dopamine, serotonin, GABA, gamma-aminobutyrate shunt</u>

Equipment: Laboratory of the department

## Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

-molecular organization of muscle tissue and the structure of the sarcomere;

-structure and functions of the main proteins of muscle tissue;

-mechanisms of muscle contraction and relaxation;

-energy sources for muscle contraction;

-biochemical diagnosis of muscle tissue pathology;

-peculiarities of the morphology and chemical composition of the nervous system;

-peculiarities of the metabolism of carbohydrates, lipids, proteins and free amino acids in nervous tissue;

-mechanisms of formation, functioning and disposal of mediators of the nervous system;

-the structure of the synapse and the main biochemical mechanisms of the functioning of neural networks (synaptic processes, formation of excitation and inhibition);

-features of Na+/K+- ATPase functioning.

-neurochemical mechanisms of action of psychotropic drugs

The higher education applicant should be able to:

- determine the content of creatine and creatinine in blood, urine and be able to correctly interpret the obtained results;

- to carry out methods of analysis of the chemical composition of nervous tissue in order to assess its specificity.

Questions to check basic knowledge on the topic of the class:

1. Anaerobic glycolysis;

2. Krebs cycle;

3. Tissue respiration and oxidative phosphorylation;

- 4. Mechanisms of formation and metabolism of creatine and creatinine.
- 5. The main ways of carbohydrate metabolism;
- 6. The main ways of lipid metabolism;
- 7. The main pathways of protein and amino acid metabolism. Formation of biogenic amines;
- 8. The main ways of ATP formation in the human body.
- 9. Morphology and cytoarchitectonics of the brain, functional load of its parts and departments.
- 3. Formation of professional skills and abilities.

3.1 Demonstration and practical work "Determination of the level of creatinine and creatine in urine".

#### Recommendations for performing tasks.

The principle of the method. The detection and quantification of creatinine is based on the color reaction of creatinine in an alkaline medium with picric acid, resulting in the formation of yellow-brown creatinine picrate



The mass concentration of creatinine in the test mixture can be calculated from the colorimetrically determined color intensity of the control (standard 1% potassium dichromate solution, which corresponds to the color of 2.5 mg of creatinine in 250 ml of solution) and the test sample. When determining the mass concentration of creatine in urine, the concentration of creatinine in it is first determined. Then creatine is converted into creatinine by boiling urine in an acidic medium and the creatinine content is also determined. The amount of creatine in the urine is calculated based on the difference between the second and first determinations, taking into account the ratio of creatinine to creatine.

Progress. Determination of the mass concentration of creatinine and creatine in urine is carried out in two portions. In one of them, the creatinine content is immediately determined. For this purpose, 2.0 ml of filtered urine, 5 ml of saturated picric acid solution, 2 ml of 10% sodium hydroxide solution are added to the volumetric flask and thoroughly mixed. After 10 minutes, the contents of the flask are brought to a volume of 250 ml with distilled water and colorimetered. The mass concentration of creatinine in urine (mg/ml) is calculated using the formula

C1 = D1Q / D2V

where D1 and D2 are optical density values of the standard potassium dichromate solution and the test solution, respectively; Q is the mass of creatinine in 250 ml of solution, equivalent in color to a 1% standard solution of potassium dichromate (2.5 mg); V is the volume of filtered urine taken for analysis (2 ml).

In another portion of urine, creatine is converted into creatinine and the amount of creatinine formed is determined. To do this, add 2 ml of the studied urine and 5 ml of a saturated picric acid solution to a 250 ml flask. Note the level of liquid in the flask, add another 100 ml of distilled water and boil for one hour. If during this time the liquid in the flask boils above the level of the mark, distilled water is added, if less, the liquid is evaporated to the mark.

After cooling, add 2 ml of 10% sodium hydroxide solution to the flask, after 10 minutes pour into a 250 ml volumetric flask, add distilled water to the 250 ml mark and colorimeter. The mass concentration of creatinine in the second portion of urine is calculated using the same formula as the first.

The difference between the values in the second and first portions, taking into account the conversion factor of creatinine into creatine, is equal to the content of the mass concentration of creatine in the examined urine

 $C = (C2 - C1) \times K$ 

where C1 and C2 are the mass concentration of creatinine in the first and second portions of urine, respectively; K is the conversion factor of creatinine into creatine (1.16). The coefficient of 1.16 is obtained as follows: the molecular weight of creatine is 131, and the molecular weight of creatinine is 113; 131 : 113 = 1.16.

Medical and biological evaluation of the obtained results

The amount of creatinine that is excreted in the urine of adults during the day ranges from 1.5 to 2.4 g. An increase in the amount of creatinine in the urine is observed during intense physical work, during fevers, etc.

There is no creatine in normal adult urine. Creatinuria occurs in cases associated with increased tissue breakdown, for example, with postpartum involution of the uterus. Children's urine always contains both creatine and creatinine.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Questions to check the final level of knowledge:

- 1. What are the types of muscle tissue?
- 2. What body functions does muscle tissue take part in?
- 3. What subcellular formations of muscles do you know?
- 4. What is ER sarcoma? Its structure.
- 5. What types of muscle tissue proteins do you know? Their structure and functions
- 6. Structural and biological role of anserine and carnosine in the functioning of muscle tissue.
- 7. Muscle tissue enzymes. Diagnostic value of determination of muscle tissue enzymes.
- 8. Molecular mechanisms of contraction and relaxation.
- 9. Sources of energy for muscle contraction depending on the intensity of physical activity.
- 10. Pathology of muscle tissue.
- 11. Structure of a neuron.
- 12. Content and features of carbohydrate metabolism in nervous tissue.
- 13. Peculiarities of lipid metabolism in nervous tissue.
- 14. Peculiarities of protein metabolism in nervous tissue.

15. Content and features of amino acid metabolism in nervous tissue. Metabolism and biological role of glutamic acid.

16. Energy exchange in the human brain. Value of aerobic oxidation of glucose; changes in the conditions of physiological sleep and anesthesia.

- 17. Mechanisms of formation, functioning and disposal of mediators of the nervous system.
- 18. Brain peptidergic system: opioid peptides, opioid peptide receptors.
- 19. Functional features and biological role of the metabolic  $\gamma$ -aminobutyrate shunt.
- 20. Molecular mechanisms of nerve impulse transmission.
- 21. Features of Na+/K+- ATPase functioning.

22. Disorders of the exchange of brain mediators and modulators in mental disorders. Neurochemical mechanisms of action of psychotropic drugs.

Test tasks.

A patient with progressive muscular dystrophy was treated

biochemical examination of urine. The appearance of which substance in large quantities in urine can confirm muscle disease in this patient?

A. Porphyrins

B. Hippuric acid

C. Urea

D. Creatinine

E. Creatine

A patient with a crush injury was brought to the traumatology department muscle tissue. What will be the biochemical indicator of urine in this case enlarged?

A. Mineral salts

- B. Glucose
- C. Total lipids
- D. Uric acid
- E. Creatinine

When examining the patient's blood, a significant increase was found activity of MB-forms of CPK (creatine phosphokinase) and LDH-1. Which one the most likely pathology?

- A. Myocardial infarction
- B. Pancreatitis
- S. Hepatitis
- D. Rheumatism
- E. Cholecystitis

The patient has muscle atony. Name the muscle enzyme

tissue, the activity of which may be reduced in such a condition.

- A. γ-Glutamyltransferase
- B. Catalase
- C. Amylase
- D. Creatine phosphokinase
- E. Transketolase

A 47-year-old man was admitted to the intensive care unit

diagnosis of myocardial infarction. Which of the fractions of lactate dehydrogenase (LDH) will prevail in the blood serum during the first two days?

- A. LDH 3
- B. LDH5
- C. LDH1
- D. LDH4
- E. LDH2

As a result of the exhausting muscular work, the employee has a lot decreased blood buffer capacity. The arrival of which acidic substance can this phenomenon be explained in the blood?

- A. Pyruvate
- B. Lactate
- C. 3-phosphoglycerate
- D. 1,3-bisphosphoglycerate
- E. alpha-ketoglutarate

An increase in the activity of LDH1, LDH2, AST was found in the patient's blood, creatine kinase. In which organ of the patient is the most likely development pathological process? A. Pancreas B. Skeletal muscles

C. Liver

D. Kidneys

E. Heart

A 46-year-old patient has been suffering from progressive muscular dystrophy for a long time Duchenne dystrophy. A change in the level of an enzyme in the blood diagnostic test in this case?

A. Glutamate dehydrogenase

- B. Lactate dehydrogenase
- C. Pyruvate dehydrogenase
- D. Creatine phosphokinase
- E. Adenylate cyclase

An 18-year-old boy was diagnosed with muscular dystrophy. Increase in blood serum content of which substance is most likely with this pathology?

A. Myoglobin

- B. Alanine
- C. Creatine
- D. Myosin
- E. Lactate

Which ions play the most important role in the formation of the action potential

- +A. K+, Na+
- B. Na+, Mg2+
- C. K+, Li+
- D. Na+, Li+
- E. K+, Mg2+

Choose from the following compounds which is not a neurotransmitter in the human nervous system:

- A. Serotonin
- B.-Nore pine phrine
- C.-Acetylcholine
- +D.  $\alpha$ -alanine
- E. GABA

Specify a pathology not associated with a violation of synaptic transmission:

- A. Myasthenia
- +B. Phenylketonuria
- C. Schizophrenia
- D. Parkinson's disease
- E. Manic-depressive psychosis

Central nervous system inhibitory mediators include:

- A. Aspartate
- B. Glutamate
- +C. GABA
- D. Dopamine
- E. Acetylcholine

At rest, the brain is supplied with energy almost entirely due to:

- A. Anaerobic glycolysis
- B. Pentose phosphate oxidation of glucose
- C. catabolism of ketone bodies
- D. Gluconeogenesis
- +E. Aerobic breakdown of glucose

Detoxification of ammonia in nervous tissue is carried out by:

- A. Urea synthesis
- B. Gluconeogenesis
- C. Reductive amination of glycine
- D. Regenerative amination of alanine
- +E. Regenerative amination of glutamate

Choose a neurotransmitter from the following compounds:

- A. Glutathione
- +B. GABA
- C. Histidine
- D. Cortisol
- E. Tryptophan

Choose a neurotransmitter from the following compounds:

- A. Oxytocin
- B. Histidine
- +C. Serotonin
- D. Cortisol
- E. Tryptophan

4. Summing up.

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

# Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

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- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

## Practical class №33

Topic: The role of the kidneys in regulating the electrolyte composition and pH of body fluids. Biochemical mechanisms of the urinary function of the kidneys. Pathobiochemistry of kidneys. Biochemical composition of human urine in normal conditions and under conditions of pathological processes, nephrolithiasis. Clinical and diagnostic value of urine composition analysis.

Goal:Learn the peculiarities of metabolism in the kidneysBasic concepts:primary urine, secondary urine, urea, clearance, ACE inhibitorsEquipment:Laboratory of the department

## Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

2. Control of the reference level of knowledge.

The higher education applicant should know:

-kidney function;

-peculiarities of metabolism in the kidneys;

-the role of the kidneys in maintaining acid-base balance;

-biochemical mechanisms of the urinary function of the kidneys;

-the biochemical composition of human urine in normal conditions and under the conditions of pathological processes;

-clinical and diagnostic value of urine composition analysis;

-hormones involved in the regulation of water-salt metabolism and kidney functions

-the principle of action of hypotensive drugs - angiotensin-converting enzyme inhibitors

The higher education applicant should be able to:

- perform qualitative reactions to protein in urine;
- determine the amount of protein in urine;
- determine the amount of glucose in the urine.

Questions to check basic knowledge on the topic of the class:

- 1. The structure of the nephron.
- 2. The mechanism of urine formation: filtration, reabsorption, secretion.
- 3. pH of biological fluids in the body.
- 4. What are the functions of the kidneys?
- 5. What are the organs of excretion other than the kidneys do you know?
- 6. What is urine?
- 7. How much urine is released per day from the body of a healthy person?
- 8. What is diuresis?

9. What is the ratio between daily water consumption and diuresis in the norm?

10. Hormones of the hypothalamic-pituitary system and the cortex of the adrenal glands, which take part in the regulation of water-salt metabolism.

11. Components of the renin-angiotensin system and its role in maintaining blood pressure.

12. Natriuretic factors of the atrium and other tissues

3.1 Demonstration and practical work "Qualitative determination of protein in urine (heating and acid precipitation".

Recommendations for performing tasks.

Progress

A) Boiling test: urine is tested in advance using litmus. If the urine has an acidic reaction, then it (2-3 ml) is immediately boiled in a test tube, and if the urine has an alkaline reaction, then it is first acidified according to litmus, adding 1% acetic acid solution drop by drop. In the presence of protein during boiling, turbidity or a precipitate of coagulated proteins is formed, which does not dissolve during repeated boiling after adding 3-5 drops of 10% acetic acid to the liquid.

B) Precipitation of protein with concentrated nitric acid (Heller's test): about 1 ml of concentrated nitric acid is poured into a test tube and urine is carefully poured from a pipette along the wall of the test tube. In the presence of protein, a white amorphous layer or turbidity, the so-called protein ring, forms at the boundary of both liquids. In the absence of protein in the urine, a colored transparent ring appears on the border between the two liquids, caused by a change in the pigments of the urine under the influence of nitric acid.

C) Precipitation of protein with sulfosalicylic acid: 2-3 drops of freshly prepared 20% solution of sulfosalicylic acid are added to 1-2 ml of urine. In the presence of protein in the urine, a white precipitate or turbidity is formed.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

3.2 Demonstration and practical work "Quantitative determination of protein by the Branderg-Stolnikov method".

#### Recommendations for performing tasks.

The principle of the method. The method is based on an experimentally established fact: the appearance of a barely noticeable protein ring in the Heller test occurs between the second and third minutes at a urine concentration of 0.0033% protein, i.e. 0.033 g/l. By successively diluting urine and layering it on nitric acid, such a maximum dilution of urine is achieved that a ring appears between the second and third minutes. Multiply the dilution by 0.033 g/l and get the protein content in the urine.

Progress. A Heller's test is performed with normal and pathological urine, for which 20 drops of concentrated nitric acid are placed in a test tube and the urine is carefully layered with a pipette. If the urine contains protein, then after 2-3 minutes. a white turbidity in the form of a ring is formed. Urine with a positive Heller test is used for protein quantification, for which a urine dilution is prepared. Pour 2 ml of distilled water into five test tubes. Add 2 ml of urine to the first, mix and take 2 ml of the mixture, transfer to the second, etc. Discard 2 ml of the collected liquid from the last test tube. Urine diluted 2, 4, 8, 16, 32 times is obtained. 2 ml of concentrated nitric acid is measured into the other five test tubes and a suitable sample of diluted urine is carefully layered on the acid using a pipette. They note the maximum dilution of urine, in which a cloudy white ring appears between the second and third minutes. The found urine dilution is multiplied by 0.033 g/l. For example, a ring of denatured protein was formed in the fourth test tube, where the dilution is equal to 16. Therefore, the protein content in the test urine is 0.033x16=0.548 g/l.

Medical and biological evaluation of the obtained results

Normal urine does not contain protein because it is unable to pass through the capillary walls. The appearance of protein in the urine indicates kidney disease. Protein can appear in the urine either as a result of a pathological change in the permeability of the capillary walls, when they begin to pass protein into the urine, or during inflammatory processes in the kidneys. For example, protein in the urine appears in glomerulonephritis (that is, inflammation of the glomeruli of the kidneys, when their permeability increases), in case of heart failure, sometimes during pregnancy.

3.3 Demonstration and practical work "Quantitative determination of glucose in urine using a polarimeter".

## Recommendations for performing tasks.

Principle of the method: glucose rotates the plane of the polarized beam to the right. The amount of glucose in the urine is determined by the angle of rotation.

Progress. Determination is carried out strictly according to the instructions for the polarimeter. The polarimeter tube is filled with filtered urine (without air bubbles), covered with ground glass, screwed tightly, wiped dry and inserted into the device. Determination is carried out 2-3 minutes after filling the tube, because fluctuations of liquid particles interfere with the study. When the color or intensity of illumination of a part of the field of view changes, the fields are equalized by the rotation of the disk and the angle of deviation of the polarized beam is determined, which is expressed in degrees of the scale of the device. With a tube length of 18.94 cm, the deflection angle of 10 corresponds to 1% glucose; if the length of the tube is 9.47 cm, then the obtained results are multiplied by 2, if the length is 4.74 cm, then by 4. After the work is finished, the urine is poured out, the tube and the glass of the polarimeter are washed with distilled water and dried.

Requirements for work results.

Enter the obtained data and calculations into the workbook.

Make medical and biological conclusions.

Questions to check the final level of knowledge:

- 1. Biochemical processes in the cortical and medullary layers of the kidneys.
- 2. Glomerular filtration.
- 3. Reabsorption and secretion.
- 4. Involvement of kidneys in regulation of volume, electrolyte composition and pH of body

## fluids.

5. Specify the acidity of urine. What buffer system plays an important role in maintaining the constancy of urine pH?

- 6. Amount, color, smell, transparency, reaction of normal urine.
- 7. Clinical and diagnostic value of quantitative and qualitative analysis of urine.
- 8. How much urea, uric acid, creatinine is excreted in the urine per day?
- 9. How much residual nitrogen is in urine?
- 10. What is Creatine Ratio?
- 11. What causes the active reaction of normal urine?

12. How many chlorides, sulfates, phosphates are excreted in the urine per day, their sources of origin.

4. Summing up.

5. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

3. Lehninger. Principles of Biochemistry. 7th edition. NY, United States. 2017.

4. Jeremy M. Berg, John L. Tymoczko, Gregory J. Gatto. Biochemistry. 8th Revised edition. 2015.

5. Lippincott Illustrated Reviews: Biochemistry. Philadelphia :Wolters Kluwer, 2017. 560 p.

6. Baynes J., Dominiczak M. Medical Biochemistry. 5th Edition. Elsevier, 2018. 712 p.

7. Donald Voet, Judith G. Voet, Charlott W. Pratt. Fundamentals of Biochemistry: Life at the Molecular Level. ISBN: 978-1-118-91840-1 February 2016, 1184 P.

Additional:

1. William Marshall, Marta Lapsley, Andrew Day, Kate Shipman. Clinical Chemistry. Elsevier, 2020. 432 p.

2. Harper's Illustrated Biochemistry / V.W. Rodwell, D.A. Bender, K.M. Botham et al. – Mc Graw Hill Education, 2015. – 817 p.

Electronic information resources:

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- 2. http://libblog.odmu.edu.ua/
- 3. https://moodle.odmu.edu.ua/login/index.php

# Practical class №34,35 Topic: Intermediate control for the semester Goal: Determine the level of assimilation of knowledge for the semester Equipment: Laboratory of the department

# Plan:

1. Organizational activities (greetings, verification of those present, announcement of the topic, purpose of the class, motivation of higher education seekers to study the topic).

- 2. List of questions for preparation for control:
- 1. Hormones and their general characteristics. Role of hormones and other bioregulators in the system of the intracellular integration of the human organism functions.
- 2. Classification of hormones and bioregulators in correspondence of structure and mechanisms of hormone activity.
- 3. Reaction of the target cells on the hormone action. Membrane (ionotropic, metabotropic) and cytosol receptors.Biochemical systems of the hormonal signals intracellular transfer: G-proteins, and secondary messengers cAMP, Ca<sup>2+</sup>-calmodulin, inositol-3-phosphate, and diacylglycerol.Molecular cell mechanisms of the steroid and thyroid hormone activity.
- 4. Neuropeptides of hypothalamus. Liberins and statins, their mechanisms of activity and biologic role.
- 5. Hormones of pituitary gland: melanotropin, thyrotropin, corticotropin, lutropin, somatotropin (growth hormone), prolactin. Their mechanisms of activity and biological role. Pathological processes related to the disturbances of hormone functions. Hormones of posterior lobe of pituitary gland. Vasopressin and oxytocin: structure, biological functions. Pathological processes related to the disturbances of hormone functions.
- 6. Insulin: structure, biosynthesis and secretion.Mechanism of insulin activity on the carbohydrate metabolism.Mechanism of insulin activity on the lipid metabolism.Mechanism of insulin activity on the protein and nucleotide metabolism.Glucagon and its mechanisms of activity on the carbohydrate and lipid metabolism.
- 7. Thyroid hormones, their structures, biological effects of T3 and T4. Disturbances of metabolic processes due to hypo- and hyperthyreosis.
- 8. Epinephrine, norepinephrine, dopamine, their structure, biosynthesis, physiological effects, biochemical mechanisms of activity. Pathological processes related to the disturbances of hormone functions.
- 9. Steroid hormones of the suprarenal glands (C<sub>21</sub>-steroids), glucocorticoids and mineralocorticoids, their structures and properties.Mechanisms of glucocorticoids activity on the carbohydrate and lipid metabolism. Mechanisms of glucocorticoids activity on the protein and nucleotide metabolism.

- 10. Female sex hormones estrogens, progesterone. Physiological and biochemical effects, related to the ovulation cycle phases. Male sex hormones ( $C_{19}$ -steroids). Physiological and biochemical effects of androgens, regulation of synthesis and secretion.
- 11. Eicosanoids: structure, biological and pharmacological properties. Aspirin and other nonsteroidal anti-inflammatory drugs as inhibitors of prostaglandin synthesis.
- 12. Biochemistry of human nutrition, the food components and nutrients, biological value of certain nutrients.
- 13. Mechanisms of conversion of nutrients, proteins, carbohydrates, and lipids, in the digestive tract. The saliva, stomach and intestine enzymes.Digestion disorders of certain nutrients in the stomach and intestines, and hereditary enzymopathies of digestive processes.
- 14. Microelements in human nutrition. Biological functions of certain microelements, and microelement deficiency manifestations.
- 15. Vitamins in human nutrition. Water-soluble and fat-soluble vitamins; exogenous and endogenous causes of vitamin deficiency.
- 16. Vitamin B<sub>1</sub> (thiamin): structure, biological properties, mechanism of action, the sources, daily need.Vitamin B<sub>2</sub> (riboflavin): structure, biological properties, mechanism of action, the sources, daily need.Vitamin PP (nicotinic acid, nicotinamide): structure, biological properties, mechanism of action, manifestations of deficiency, sources, the daily need.
- 17. Vitamin  $B_6$  (pyridoxine): structure, biological properties, mechanism of action, the sources, the daily need.Vitamin B12 (cobalamin): biological properties, mechanism of action, manifestations of deficiency, sources, the daily need.Vitamin  $B_c$  (Folic Acid): biological properties, mechanism of action, the sources, the daily need.
- 18. Vitamin H (biotin): biological properties, mechanism of action, the sources, the daily need.Vitamin B<sub>3</sub> (pantothenic acid): biological properties, mechanism of action, the sources, and the daily need.Vitamin C (ascorbic acid): structure, biological properties, mechanism of action, manifestations of deficiency, sources, the daily need.Vitamin P (flavonoids): structure, biological properties, mechanism of action, manifestations of deficiency, sources, the daily need.
- 19. Vitamin A (retinol, retinal, retinoic acid): biological properties, mechanism of action, manifestations of deficiency, sources, the daily need. Vitamin D<sub>3</sub> (cholecalciferol): biological properties, mechanism of action, manifestations of deficiency, sources, the daily need.
- 20. Vitamin K (phylloquinone, farnohinon): biological properties, mechanism of action, manifestations of deficiency, sources, the daily need.Vitamin E (α-tocopherol): biological properties, mechanism of action, manifestations of deficiency, sources, the daily need.
- 21. Biochemical and physiological functions of blood in the human body. Respiratory function of erythrocytes.Hemoglobin: mechanisms of it's' participation in the transport of oxygen and carbon dioxide. Types and pathological forms of human hemoglobin.The decomposition of hemoglobin. Differential diagnosis of jaundice.
- 22. Buffer systems of blood. Acid-base balance disturbances in the body, metabolic and respiratory acidosis, and alkalosis. Mechanisms of compensation.Biochemical composition of human blood. Plasma proteins and their clinical and biochemical characteristics.
- 23. Enzymes of plasma; its' importance in enzyme diagnostics of diseases.Kallikrein-kinin system of blood and tissues. Medicines, which are antagonists of kinin formation.
- 24. Non-protein organic compounds of plasma. Inorganic plasma components.
- 25. Biochemical and functional characteristics of the hemostasis system.
- 26. Coagulation and characterization of certain factors of coagulation, mechanism of function of the cascade blood clotting system. The role of vitamin K in the reactions of coagulation, therapeutic agents, which are agonists and antagonists of vitamin K.
- 27. Anticoagulation blood system and characteristics of anticoagulants. Hereditary disorders of blood coagulation. Fibrinolytic system of blood. Medicines that affect the processes of fibrinolysis.

- 28. Immunoglobulins and biochemical characteristics of individual classes of human immunoglobulins. Mediators and hormones of the immune system: interleukins, interferons, protein-peptide factors regulating growth and cell proliferation. Complement system, biochemical components of human complement, and the classical and alternative ways of activation. Biochemical mechanisms of immunodeficiency states: primary (hereditary) and secondary immunodeficiencies.
- 29. Biochemical functions of liver: carbohydrate and protein synthetic, urine formation, cholepoietic, regulation of lipid composition of blood.
- 30. Detoxicative function of liver. Types of biotransformation reactions of xenobiotics and endogenous toxins.
- 31. Microsomal oxidation reactions. Cytochrome P450 and related electron-transport chain in the membranes of the endoplasmic reticulum of hepatocytes.
- 32. Metholism of porphyrins: structure of heme. Scheme of protoporphyrin IX and heme biosynthesis. Hereditary disorders of the porphyrins biosynthesis, types of porphyrias.
- 33. Catabolism of hemoglobin and heme (scheme); formation and structure of bile pigments.Pathobiology and types of jaundice; biochemical diagnosis of jaundice.The role of the liver in the metabolism of bile pigments. Pathobiology of jaundice. Types of hereditary (enzyme) jaundice.
- 34. Water-salt metabolism in the body. Intracellular and non-cellular water; metabolism of water, sodium, potassium. The role of the kidneys in regulating the volume, electrolyte composition and pH of the body fluids. Biochemical mechanisms of urine-forming function of the kidneys.
- 35. Renin-angiotensin system of the kidneys. Hypotensive drugs angiotensin converting enzyme inhibitors.
- 36. Biochemical composition of muscles. Myocyte proteins: myosin, actin, tropomyosin, troponin.Molecular mechanisms of muscle contraction. The role of Ca<sup>2+</sup>ions in the regulation of muscle contraction and relaxation.Bioenergy of muscle tissue. ATP sources. The role of creatine phosphate in providing energy for muscle contraction.
- 37. Biochemistry of the nervous system: features of the biochemical composition and metabolism of the brain. Energy metabolism in the human brain. The value of aerobic oxidation of glucose; changes in conditions of physiological sleep and anesthesia.
- 38. Biochemistry of neurotransmitters; receptors of neurotransmitters and physiologically active compounds.Peptidergic system of the cerebrum: opioid peptides, receptors of opioid peptides.
- 39. Violations of the metabolism of mediators and modulators of the brain in mental disorders. Neurochemical mechanisms of psychotropic drugs action.
  - 3. Summing up.

4. List of recommended literature (main, additional, electronic information resources):

Main:

1. Gubsky Yu.I., I.V. Nizhenkovska, Korda M.M. Biological and Bioorganic Chemistry: in 2 books. Book 2. Biological Chemistry: textbook. 2021. 544 p.

2. Satyanarayana U. Biochemistry. 5th edition. India 2020. 777 p.

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