O. N. RYNEISKAYA, E. M. ERMOLENKO

BIOORGANIC CHEMISTRY

Manual for laboratory classes

Minsk BSMU 2018

МИНИСТЕРСТВО ЗДРАВООХРАНЕНИЯ РЕСПУБЛИКИ БЕЛАРУСЬ
БЕЛОРУССКИЙ ГОСУДАРСТВЕННЫЙ МЕДИЦИНСКИЙ УНИВЕРСИТЕТ
КАФЕДРА БИООРГАНИЧЕСКОЙ ХИМИИ

О. Н. Ринейская, Е. М. Ермоленко

БИООРГАНИЧЕСКАЯ ХИМИЯ

BIOORGANIC CHEMISTRY

Практикум

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Ринейская, О. Н.

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Содержит методические рекомендации для подготовки к лабораторным занятиям по биоорганической химии на английском языке. К каждой теме даны цель занятия, вопросы для обсуждения, письменные задания, а также указана литература для подготовки. Приведены описания и протоколы лабораторных опытов. Первое издание вышло в 2015 году.

Предназначен для студентов 1-го курса медицинского факультета иностранных учащихся, обучающихся на английском языке по специальности «Лечебное дело».

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REGISTRATION FORM

Student name _____

№	Theme	Date	Mark	Signature of teacher
	Classification and nomenclature of organic compounds			
	Chemical bond structure and atom effects in the organic molecules			
	Stereoisomerism, its role for biological activity demonstration			
	Hydrocarbons			
	Monofunctional hydrocarbon derivatives			
	Biologically important reactions of aldehydes and ketones			
	Carboxylic acids and their derivatives			
	Concluding test «Theoretical fundamentals of basic classes of organic compound structure and reactivity»			
	Poly- and heterofunctional compounds			
	Biologically important heterocyclic compounds			
	Carbohydrates. Monosaccharides			
	Oligo- and polysaccharides			
	Structure and reactivity of amino acids			
	Peptides. The levels of protein organization			
	Nucleosides. Nucleotides. Nucleic acids			
	Lipids. Structure, properties. Lipid peroxidation			
	Steroids			
	Concluding test «Biopolymers and their structural components»			

LABORATORY SAFETY RULES

1. Dress appropriately for the lab. Wear white lab coat. Tie back long hair.
2. Know what safety equipment is available and how to use it. This includes eye-wash place, fire blanket, fire extinguisher and sand.
3. Know the dangers of the chemicals in use, and read labels carefully. Do not taste or sniff chemicals.
4. Dispose of chemicals according to instruction. Use designated disposal sites, and follow the rules. Never return unneeded chemicals to the original containers.
5. Always add acids and bases to water slowly to avoid splattering. This is especially important when using strong acids and bases that can generate significant heat, form steam, and splash out of the container.
6. Never point test tubes at yourself or others. Be aware of reactions that are occurring so that you can remove them from the heat if necessary.
7. Do not eat or drink in the lab! It is too easy to take in some dangerous substance accidentally.
8. Follow all directions. Never occasionally mix chemicals. Pay attention to the order in which chemicals are to be added to each other, and do not deviate!
9. After the end of the experiment each student should submit an account of the work that have been done, then to wash up chemical crockery, clean a workplace and ask the student on duty to check it.

Responsibilities of the student on duty:

– to get all the necessary equipment from the laboratory assistant;

– to keep an order the laboratory room;

– student on duty should leave the laboratory the last, after receiving permition from the lab assistant.

I agree _____20___year _____
(date) (signature)

PRECAUTIONS

Work with alcohol lamps

Careless work with an alcohol lamp can result in a fire, that is why it is necessary to follow the below requirements:

– the wick of an alcohol lamp should tightly enter the aperture of a metal bush; the topping should be put forward for 1 cm and fluffed up;

– the bush should close the aperture of a alcohol lamp tightly; the alcohol lamp should be filled with alcohol no more than 2/3 of the volume;

– the lighting of an alcohol lamp should be carried only by matches, it is strictly forbidden to light an alcohol lamp from another alcohol lamp, because the bush can stoop and coming out steams of alcohol can be fired;

– to blow out an alcohol lamp only by covering it with bell-glass;

– when heating up substances in chemical glassware it is necessary to heat them at the top or mid-range flame, not touching a wick, because a wick is always cool, and when hot glass contacts with it, glass may burst.

Work with chemical glassware

Heating substances in glassware should be performed gradually, slightly rotating it and cautiously shaking from time to time. When heating a test tube with a liquid on the open fire, splasing of a liquid is possible. Because of this fact, the aperture of a test tube should be directed aside from you and from your neighbours. Especially it is necessary to avoid injuring the eyes with hot splashes, that it is why it is forbidden to bend forward to the test tube and look inside. When heating the test tube, it should be kept at the angle of inclined position (45º), so that splashes will hit walls of a glassware and will not be thrown outside. If the liquid starts to rise in an exhaust tube, it is necessary to let down a test tube immediately, so that the fluid level in it will become lower than the end of an exhaust tube.

Work with inflammable liquids (IL)

IL (diethyl ether, alcohol, toluene, acetone, acetoacetic ether) are kept always in a fume hood. Experiments with these substances are carried out under draught, far from open fire and the turned on small stoves. If an ignition of the IL happened in a vessel, it is necessary to cover it quickly with a fire-prevention blanket. If the burning liquid has been spilt, it must be extinguished by sand. If the clothes begin to fire, it is necessary to wrap up quickly and densely in a fire-prevention blanket.

Work with acids and alkalis

Concentrated solutions of nitric, sulfuric, hydrochloric acids, nitrosulfuric acid are kept in a fume hood. All experiments with concentrated acids and alkalis are carried out only in the fume hood. It is necessary to cover carelessly spilt on the floor acids and alkalis by sand and after that to clean up.

Work with toxicants

Toxic organic substances — aniline, methyl amine, toluene, picric acid are kept in a fume hood. It is necessary to be cautious with these substances, not to inhale their steams, to avoid injuring the hands as they can penetrate through the skin. In case of emergency when these substances got on hands, it is necessary to wash up quickly the hands with warm water and soap. If inhaled the steams — immediately to go out in the fresh air.

First-aid treatment in case of accidents:

– in case of hands are cut with glass first of all it is necessary to remove all the splinters out of the wound, then to treat the wound with an alcohol solution of iodine and to put a bandage;

– in case of thermal burns happen it is necessary to treat the burnt place with the 70 % solution of ethanol;

– in case of burns are caused by solutions of acids or alkalis it is necessary to wash up the burnt site with water quickly and to put an aseptic bandage;

– in case of acids or alkalis hit the eyes it is necessary wash them with water carefully and to refer the victim to the outpatient clinic;

– in case of skin burns caused by bromine it is necessary quickly to wash the injured place off with ethanol and to put anti-burn emulsion;

– in case of burns caused by hot organic liquids it is necessary to wash out the injured place with ethanol;

– in case of burns caused by liquid phenol it is necessary to massage the emerged sites of white skin with a glycerin until normal skin color is restored then to wash with water and to put the gauze bandage moistened with a glycerin solution;

– after providing the first-aid treatment it necessary to address to the health center of the university or to the outpatient clinic.

LABWORK № 1
CLASSIFICATION AND NOMENCLATURE OF ORGANIC COMPOUNDS

Objective: to study composition unity, configuration and conformation concept for organic molecules.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 19–32.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 27–39.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 3–7.

Problems for discussion:

1. Introduction into bioorganic chemistry: the definition of subject, objects learned by bioorganic chemistry.
2. Classification of organic compounds: a) according to the carbon chain structure; b) according to the functional groups.
3. Nomenclature of organic compounds: a) trivial (or common) nomenclature; b) systematic nomenclature IUPAC.

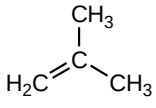
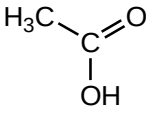
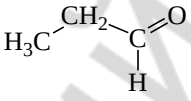
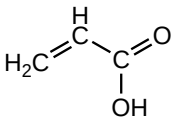
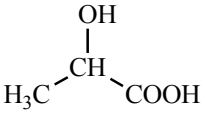
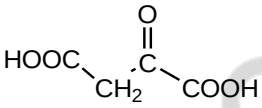
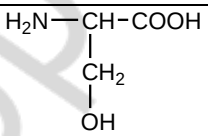
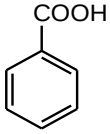
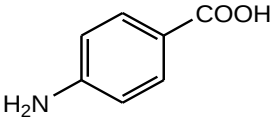
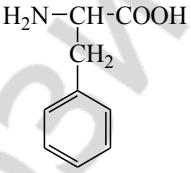
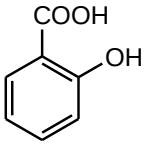
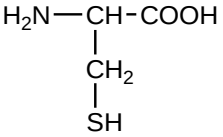
Exercises

1. Write the formulas of the following compounds:

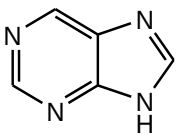
methane	ethane	propane
butane	ethene	propene
but-1-ene	but-2-ene	2-methylpropene
ethanol	pentan-1-ol	propan-2-ol
butan-2-ol	propanone	ethanethiol
methanoic acid	propanoic acid	benzene

phenol	benzoic acid	toluene
ethanedioic acid	butanedioic acid	butenedioic acid
2-aminopropanoic acid	2-oxopentanedioic acid	

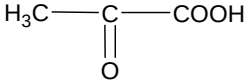
2. Give the IUPAC names for the following compounds:

TEST CONTROL

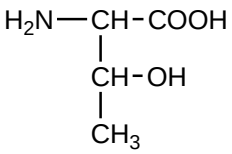
1. Give the name for the heterocycle: 

1) pyrrole; 2) purine; 3) pyridine; 4) pyrimidine.

2. Give the IUPAC name for the following compound: 

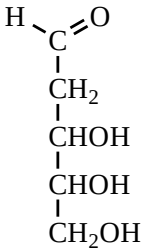
1) α-ketopropionoic acid; 3) pyruvic acid;
2) 2-oxopropanoic acid; 4) oxaloacetic acid.

3. Choose the IUPAC name of the amino acid (threonine):



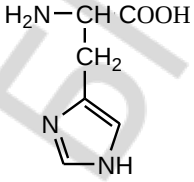
1) 2-hydroxypentanoic acid;
2) 2-aminobutanoic acid;
3) 2-amino-3-aminopropanoic acid;
4) 2-amino-3-hydroxybutanoic acid.

4. Choose the IUPAC name of the deoxyribose:



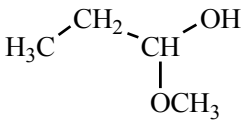
1) 1,3,4,5,6-pentahydroxyhexanone-2;
2) 2,3,4,5,6-pentahydroxyhexanal;
3) 2,3,4,5-tetrahydroxypentanal;
4) 3,4,5-trihydroxypentanal.

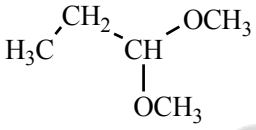
5. Choose the IUPAC name of the following compound

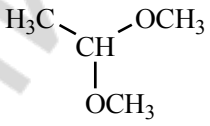


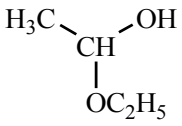
1) 2-amino-3-imidazolylpropanoic acid;
2) 2-amino-3-indolylpropanoic acid;
3) 2-amino-4- imidazolylpropanoic acid;
4) 2-hydroxy-3- imidazolylpropanoic acid.

6. Select the structural formula of the 1-methoxypropanol:

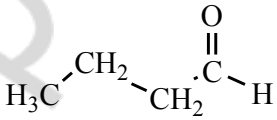
1) 

2) 

3) 

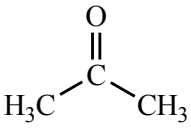
4) 

7. Choose the name of the following compound:



1) propanoic acid;
2) propanal;
3) butanal;
4) butanoic acid.

8. Select the IUPAC name of the following compound:

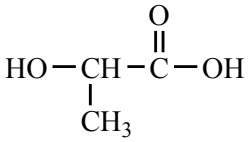


1) acetone;
2) propanone;
3) propanal;
4) propanoic acid.

9. Select unsaturated compound(s):

1) but-2-ene; 2) ethane; 3) cyclohexene; 4) benzene.

10. Select the trivial name of the compound:



1) 2-hydroxypropanoic acid;
2) alanine;
3) lactic acid;
4) malic acid.

PRACTICAL PART

1. Antioxidant activity of ascorbic acid.

Take the two test tubes. In both tubes, place 2 drops of ethanol∗. To one of them add a few crystals of ascorbic acid * with a glass spatula. Then add 1 drop of KMnO4 solution (14) and 2 drops of H_2SO_4 solution (23) to each tube, and shake. Heat each tube to the boil and discoloration of the solution. Note the appearance of apples smell in one of the test tubes.

Observed changes: _____

_____

Signature of teacher:

LABWORK № 2
CHEMICAL BOND STRUCTURE AND ELECTRONIC EFFECTS IN THE ORGANIC MOLECULES

Objective: to develop knowledge about chemical bond structure, dimensional and electronic effects of substituents.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 5–17, 33–44.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 13–25, 40–49.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 7–16.

Problems for discussion:

1. An electronic and dimensional structure of sp^2-hybridized carbon atom.
2. Conjugated systems. Conjugation energy.
3. Cyclic conjugated systems. Aromaticity. Huckel's rule. Aromaticity of benzoic and non-benzoic systems.
4. Aromaticity of heterocyclic systems (pyrrole, pyridine).
5. Inductive effect.
6. Mesomeric effect.
7. Electron donating and electron withdrawing substituents.

Exercises:

1. Write down electronic configuration of carbon atom at the base and exited condition.

Draw the spatial structure of …

a) sp^3-hybridized carbon atom

b) sp^2-hybridized carbon atom

2. Write the formulas of the following compounds. Indicate compounds with conjugated system.

but-1,3-diene	hex-2,4-diene

pent-1,4-diene	but-2-ene

3. Determine the type of conjugated system:

2-methylbut-1,3-diene	propanoic acid
propenal	pyrrole
propenoic acid	pyridine

4. Write the compound formulas. Select aromatic ones from them (using the Huckel's rule):

benzene	pyridine
cyclohex-1,3-diene	pyrrole
imidazole	cyclohexane
pyrimidine	purine

phenanthrene	cyclopentane

5. ***Electronic effects*** — …

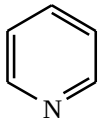
Show the electron density distribution in the molecules with inductive and mesomeric effects:

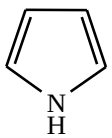
1-chlorobutane	propanal
benzaldehyde	propenal
ethanol	phenol

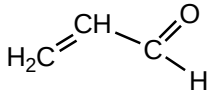
TEST CONTROL

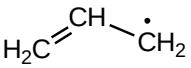
1. Indicate formulas of compounds with conjugated double bonds:
1) ethane;
2) pentadiene-1,3;
3) cycloheptatrienyl cation;
4) propenoic acid.

2. Indicate formulas of compounds with conjugated p-π double bonds:

1) 

2) 

3) 

4) 

3. Compounds with conjugated p-π double bonds are following:
1) benzene;
2) naphthalene;
3) cyclopentadienyl anion;
4) vinylamine.

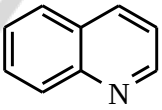
4. Indicate correct statements about pyridine: 1) everyone atom are in the sp^2-hybridization; 2) nitrogen gives in the conjudated system 2 electrons; 3) is π-deficient aromatic system; 4) nitrogen gives in the conjudated system 1 electron; 5) is π-excessive aromatic system.
1) 1, 4, 5; 2) 1, 2, 3; 3) 1, 3, 4; 4) 1, 2, 5.

5. What electronic effect(s) does hydroxyl group possess in propanol:
1) +I, –M; 2) –I; 3) –I, +M; 4) –I, –M.

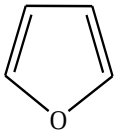
6. Which substitutions possess electron donor properties towards benzene:
1) – COOH; 2) – CH_3; 3) – OH; 4) – $NHCH_3$.

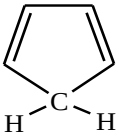
7. What electronic effect(s) does hydroxyl group possess in phenol:
1) +I, –M; 2) –I; 3) –I, +M; 4) –I, –M.

8. How many electrons are in cyclic conjugated system of quinoline:
1) 14; 2) 8; 3) 12; 4) 10.



9. Which of the following compounds are aromatic:

1) 
2) 

3) 

4) 

10. Indicate electronic effects of functional groups in the following compound:

A) benzyl alcohol;	1) –I, –M;
B) phenol;	2) –I < +M;
C) ethanol;	3) –I;
D) chlorobenzene.	4) –I > +M.

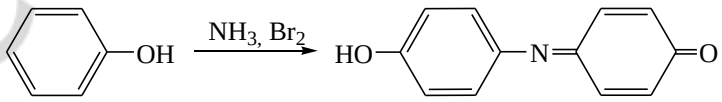
PRACTICAL PART

1. Indophenol test.

In a test tube, place 1 drop water emulsion of phenol *, 3 drops of ammonia solution *, and 3 drops of a saturated solution of bromine water *. Note the appearance of the characteristic staining.

Observed changes: _____

_____



indophenol

Conclusion: _____

_____

Signature of teacher:

LABWORK № 3
STEREOISOMERISM, ITS ROLE FOR BIOLOGICAL ACTIVITY DEMONSTRATION

Objective: to study the dimensional organization and discuss a stereoisomerism role for interaction specificity on a molecular scale understanding.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 61–81.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 76–81, 149–161.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 16–27.

Problems for discussion:

1. Stereoisomerism. Classification of stereoisomers.
2. A spatial structure of a sp^3-hybridized carbon atom. Configuration. Stereochemical formulas. Molecular models.
3. Ethane configuration and conformations, torsion strain. Newman projections.
4. Buthane conformations. Van der Waals strain. Long-chain compound conformations.
5. Carbocyclic compound conformations, angle strain. Cyclohexane conformations. A cyclohexane ring in the biologically important compounds.
6. Chiral and achiral molecules. Chiral centers. Optical activity is the property inherent chiral molecules.
7. Fischer's projective formulas. Enantiomers.
8. Relative D-,L-nomenclature of stereoisomers. Glyceraldehyde as the configuration standard. R, S-system of a configuration designation.
9. Racemic mixtures. Methods of racemic substance division.
10. Diastereoisomerism. Stereoisomers of tartaric acid.
11. Cys-, trans-isomerism. Stereoisomers of butenedioic and oleic acids.

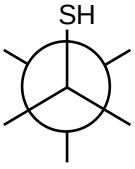
Exercises

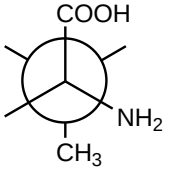
1. Write all possible conformations by means of Newman projections for the following compounds.

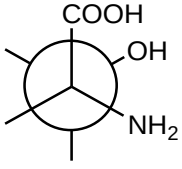
a) ethane

b) butane

2. Write the structural formulas for the following Newman projections:

a) 

b) 

c) 

3. Draw the possible chair conformations of the cyclohexanol.

4. Draw the preferred conformation of the 2-methylcyclohexanol.

5. ***Chiral center*** — is …

Write structural formulas: butanol-2, 2,3-dihydroxypropanal, 2-aminopropanoic acid, 3-hydroxybutanoic acid. Mark (with *) chiral centers.

6. Write Fisher projections of 2-aminopropanoic acid. What is the enantiomers? What is the racemic mixture?

7. Write Fisher projections of 2-amino-3-hydroxybutanoic acid (2 chiral centers). Show the pairs of enantiomers and diastereomers.

8. Write R- and S-isomers for the 2-hydroxypropanoic acid.

9. Draw *cis*- and *trans*-isomers of butenedioic acid.

TEST CONTROL

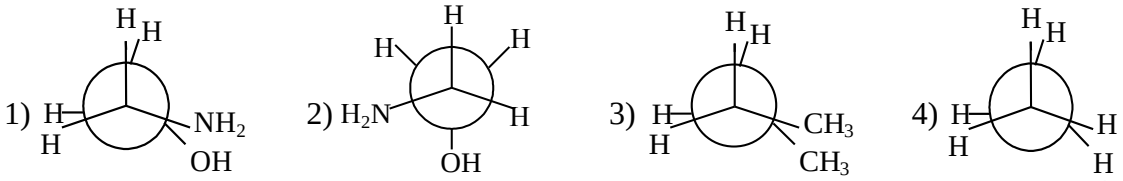
1. Repulsive interaction between electron clouds in the C-H bond is called:
1) Van der Waals strain;
2) angle strain;
3) Baeyer strain;
4) torsion strain.

2. Indicate compounds with chiral centers:
1) 2,3-dihydroxybutanedioic acid;
2) methanol;
3) 2-aminobutanoic acid;
4) butanol-2.

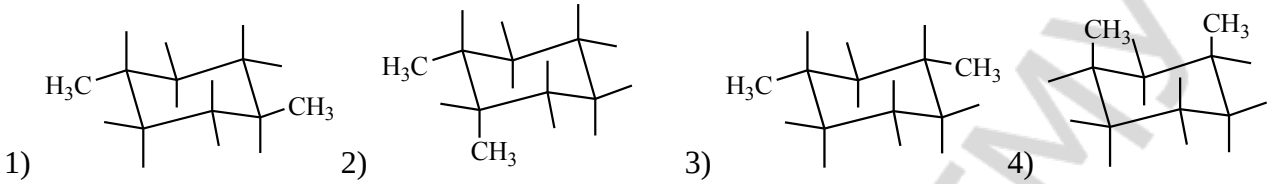
3. Various spatial arrangement of the atoms in molecular that differ only after rotation about C-C single bonds are:
1) enantiomers;
2) configuration;
3) diastereomers;
4) conformation.

4. Less stable butane conformation is:
1) stagged; 2) eclipsed; 3) skew; 4) zigzag.

5. Select conformations with the maximal Van der Waals strain:



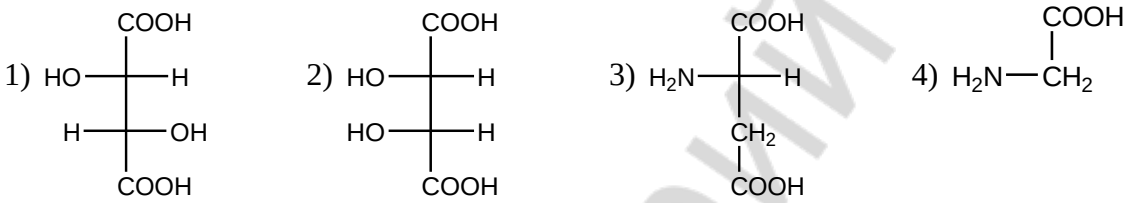
6. Less stable 1,3-dimethylcyclohexane conformation is:



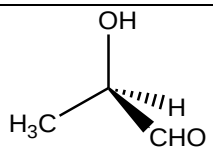
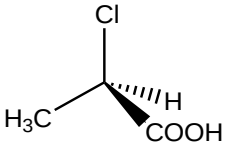
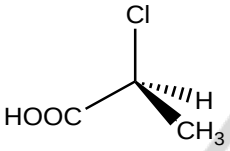
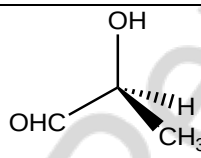
7. Select compounds with 2 chiral centrals:

1) 2-amino-3-methylpentanoic acid;
2) 2,3-dihydroxybutandioc acid;
3) 2-amino-3-methylbutanoic acid;
4) 2-hydroxyethanoic acid.

8. Select L-stereoisomers:



9. Select names for the corresponded structures:

A) 	1) R-2-chloropropanoic acid
B) 	2) R-2-hydroxypropanal
C) 	3) S-2- hydroxypropanal
D) 	4) S-2- chloropropanoic acid

10. Diastereoisomers — are:

1) pairs of stereoisomers which concern to each other as a subject and its display in an ideal plane mirror, possess in achiral surrounding identical chemical and physical properties, except for a sign on optical rotation;
2) pairs of stereoisomers of the same substance not being a mirror image of one another and possessing various chemical and physical properties;

3) pairs of stereoisomers of the same substance not being a mirror image of one another and possessing the same chemical and physical properties;
4) pairs of stereoisomers which consist in migration of some groups within a molecule and is a accompanied by redistribution of electron density.

Signature of teacher:

LABWORK № 4
HYDROCARBONS

Objective: to develop knowledge about classification and mechanisms of organic reactions; to develop skills of carrying out of qualitative tests for double bond detection in organic compounds.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 94–110.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 52–57, 75, 82–96.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 27–28, 30–35.

Problems for discussion:

1. Organic reaction mechanism definition. Homolytic and heterolytic mechanisms of bond cleavage. Classification of reagents in organic reactions
2. Organic reactions classification according to the direction and result of reaction.
3. Reactions of radical substitution (S_R). Alkanes and cycloalkanes.
4. Electrophilic addition (A_E) to alkenes: hydrogenation, halogenation, hydrohalogenation and hydration reactions. The Markovnikov's rule.
5. Mechanism of electrophilic substitution reactions (S_E) in aromatic compounds. I and II sort directing substitutients.

Exercises:

1. Indicate the type of reagent:

$^{+}CH_3$	HOH	$^{\cdot}CH_3$	$CH_3 – Cl$	$CH_3 – OH$

2. Write the scheme of chlorination reaction of propane. Indicate mechanism.

3. Write the schemes of addition reaction:
a) Br_2 to propene

b) HBr to propene

c) HOH to butenedioic acid

d) HBr to propenoic acid

4. Describe the reaction mechanism of:
a) chlorination of benzene ($AlCl_3$ as catalyst)

b) alkylation of toluene with $CH_3 – CH_2 – Cl$ ($AlCl_3$ as catalyst)

TEST CONTROL

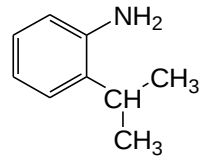
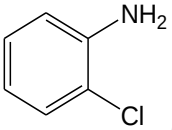
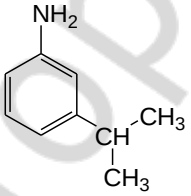
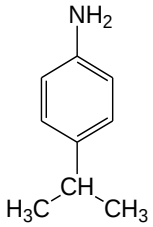
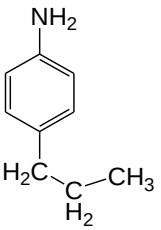
1. Nucleophile reagents are:
1) H^-; 2) HOH; 3) C_2H_5OH; 4) H^+; 5) CH_3NH_2.

2. Select properties of free radicals reactions:
1) molecular contain polar covalent bond;
2) covalent bonds breaks as a result of hemolysis;
3) acids and bases catalyze these reactions;
4) require violent conditions (high t°, pressure, irradiation).

3. Electrophilic addition reaction usually takes place in:
1) cyclohexene; 3) ethane;
2) but-2-enoic acid; 4) 3-methyl-1-chlorobutane.

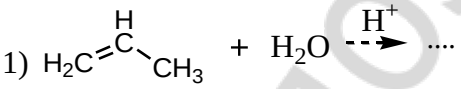
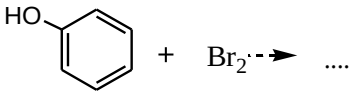
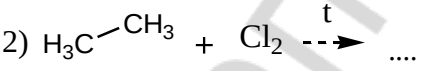
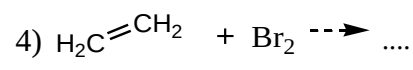
4. The following product is mainly formed as a result of interaction of 2-methylpenten-1 and HCl:
1) 4-methyl-3-chloropentane; 3) 2-methyl-2-chloropentane;
2) 4-methyl-2-chloropentane; 4) 2-methyl-1-chloropentane

5. The following product is mainly formed as a result of interaction of 2-chloropropane and aniline with catalyst:

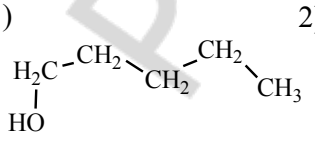
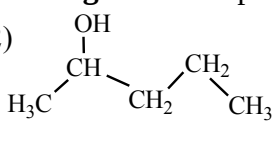
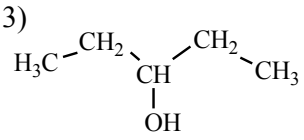
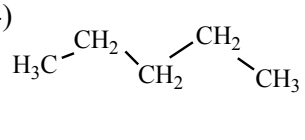
1) 
2) 
3) 
4) 
5) 

6. Hydration reaction is:
1) hydrogen addition; 3) hydrogen elimination;
2) water addition; 4) water elimination.

7. Select scheme(s) of electrophilic addition reaction(s):

1) 
3) 
2) 
4) 

8. Indicate product of following reaction: pent-1-ene + HOH $\xrightarrow{H^+}$ ……

1) 
2) 
3) 
4) 

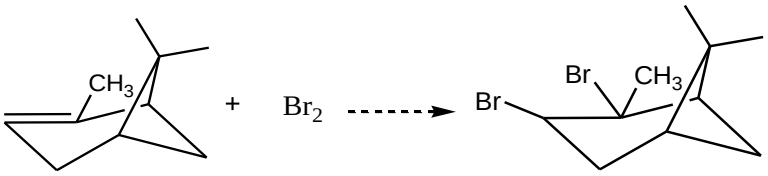
9. Select reactions which goes according Marcovnicov rules:
1) ethane hydration;
2) propenoic acid hydration;

3) propene hydration;
4) butene-2 hydrohalogenation;
5) butene-1 hydrohalogenation.

10. Indicate compound possessing strongest reaction ability in the S_E mechanism:
1) benzene; 2) toluene; 3) benzoic acid; 4) pyridine.

PRACTICAL PART

1. Qualitative test on the alkenes with bromine water.



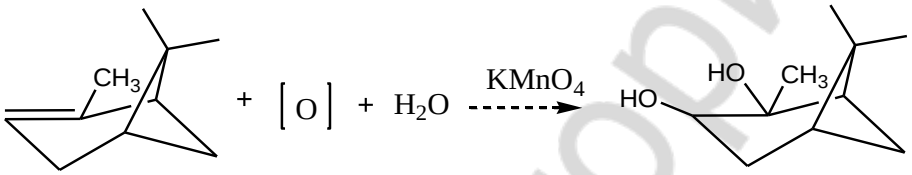
Accomplishment: to 4 drops of bromine water*[1] add 2 drops of α-pinene* and shake.

Observed changes: _____

Conclusion: _____

_____

2. Qualitative test on the alkenes with potassium permanganate.



α-pinene α-pinenglycol

Accomplishment: to 3 drops of $KMnO_4$ (14) solution add 1 drop of α-pinene* and shake.

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

[1] *Notice: reagents marked with asterisk (*) are in the fume.

LABWORK № 5
MONOFUNCTIONAL HYDROCARBON DERIVATIVES

Objective: to study structure and properties of monofunctional hydrocarbon derivatives; acidity and basicity of organic compounds; to generate skills for qualitative determination of organic compound acidity and basicity.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 47–59, 112–131.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 61–73, 101–119.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 28–30, 35–41.

Problems for discussion:

1. The Brensted theory of organic compound acidity and basicity. The Lewis electronic theory of organic compound acidity and basicity. Classification of organic acids.
2. The quantitative and qualitative characteristics of acidity. The factors influencing on the acidic properties of organic compounds.
3. Oxidation reactions of alcohols, thiols and phenols. Antioxidants and their role in processes of vital activity.
4. Basicity. The factors influencing on the basic properties of organic compounds.
5. Amphoteric properties of organic compounds. Hydrogen bonds.
6. Nucleophilic substitution reaction at sp^3-hybrid carbon atom. Elimination reaction.

Exercises

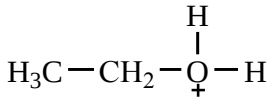
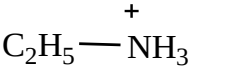
1. ***Brensted acid*** — …

Brensted base — …

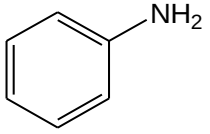
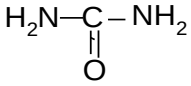
Lewis acid — …

Lewis base — …

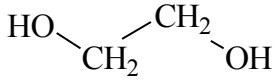
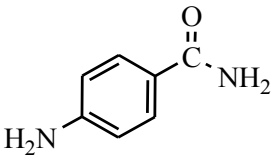
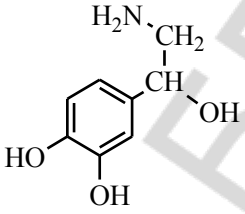
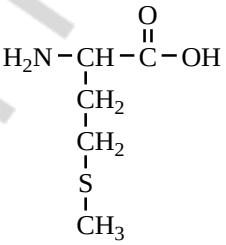
Write down the formulas of conjugate bases for the following acids:

CH_3COOH		H_2O		C_6H_5OH

Write down the formulas of conjugate acids for the following bases:

CH_3-NH_2	H_2O	OH^-	CH_3COO^-		

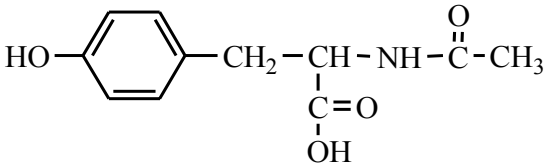
2. Indicate acidic and basic centers at the following compounds:

3. Compare acidity of compounds in the following groups:

ethanol and ethanthiol	ethanoic and ethanedioic acids
phenol and *p*-aminophenol	benzoic acid and *o*-hydroxybenzoic acid

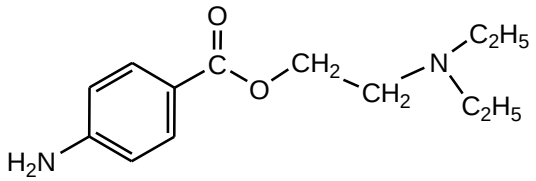
4. Indicate acidic centers at the N-acetyltyrosine:



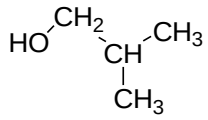
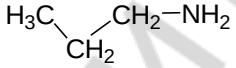
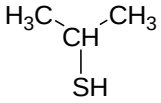
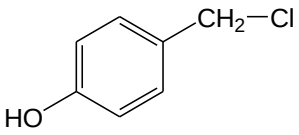
5. Compare basicity of compounds in the following groups:

ethylamine and ethanol	methylamine and dimethylamine
ethylamine and aniline	2-aminoethanol and ethylamine

6. Show the strongest basic center at the procaine molecule. Write the reaction of procaine with hydrochloric acid.



7. Indicate reactive sites in the following molecules:

8. Write the schemes of interaction reactions of:

a) 1-chloropropane and NaOH solution

b) propan-1-ol and HBr

c) 2-bromo-2-methylpropane and alcohol solution of NaOH

9. Write the scheme of dehydration reactions of 2-hydroxybutanedioic acid *in vivo.*

10. Write the ethanol oxidation reaction:
in vitro:

in vivo:

11. Write the scheme of oxidation reaction:
a) methanethiol

b) 2-amino-3-mercaptopropanoic acid

TEST CONTROL

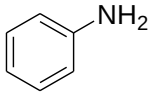
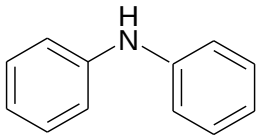
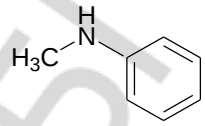
1. Acidity increases in the following row of acids:
1) acetic acid, oxalic acid, malonic acid;
2) acetic acid, malonic acid, oxalic acid;
3) oxalic acid, malonic acid, acetic acid;
4) malonic acid, acetic acid, oxalic acid.

2. Basicity according to the Bransted theory is ability of molecular or ion:
1) accept electrons;
2) donate electrons;
3) donate proton;
4) accept proton.

3. Indicate the correct statement about acidity comparison:
1) conjugation stabilizes anion and increase acidity;
2) electron donors increase acidity;
3) electron withdrawers increase acidity;
4) solvation effect influence on anion stability and acidity.

4. Select substances which are capable to link heavy metals:
1) 2-amino-3-mercaptopropanoic acid;
2) propanol-2;
3) 2,3-dimercaptopropanol-1;
4) diethyl disulfide.

5. Basicity decreases in the following row of amines:

1)  2)  3) CH_3NH_2 4) 

6. Acidity according to the Lewis theory is the ability of molecule or ion:
1) to accept proton; 3) to donate electrons;
2) to accept electrons; 4) to donate proton.

7. Indicate the factors which influence on the basicity:
1) polarizability of the basic site elements is in the same period of the periodic table;
2) electronegativity of the basic site elements is in the same period of the periodic table;
3) electronegativity of the basic site elements is in the same group of the periodic table;
4) polarizability of the basic site elements is in the same group of the periodic table.

8. Give characteristics for interaction reaction between butene-2 and H_2O (in acidic medium):
1) S_N mechanism; 2) water is electrophile; 3) S_E mechanism; 4) A_E mechanism.

9. Give characteristics for interaction reaction between benzene and isopropyl chloride (with $AlCl_3$ presence):
1) Cl^+ is electrophile; 3) S_E mechanism;
2) alkylation of benzene is result of this reaction; 4) S_N mechanism.

10. Find the accordance between scheme of the reaction and typical reaction mechanism:
A) toluene + CH_3Br ($FeBr_3$); 1) S_R;
B) propene + HCl; 2) A_E;
C) ethane + Cl_2 (light); 3) S_E;
D) *tert*-butyl alcohol+ $HBr_{(conc.)}$. 4) S_N.

PRACTICAL PART

1. Oxidation of primary alcohols.

Alcohol oxidation reaction is carried out in narrow term. Primary alcohols are oxidized to aldehydes.

$$3\,\underset{\text{ethanol}}{CH_3CH_2OH} + K_2Cr_2O_7 + 4H_2SO_4 \xrightarrow{t} \underset{\text{ethanal}}{H_3C-C(=O)-H} + K_2SO_4 + Cr_2(SO_4)_3 + 7H_2O$$

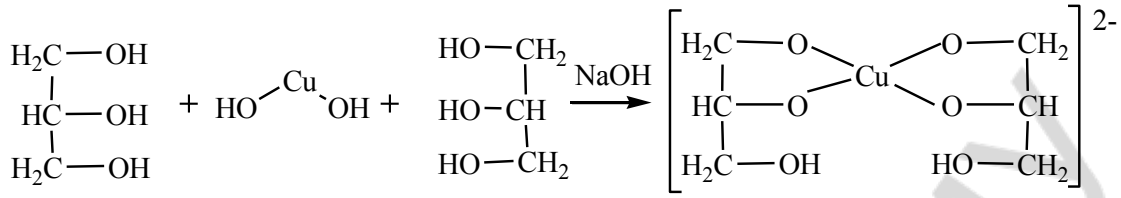
Accomplishment: add 2 drops of H_2SO_4 (23) dilute solution and 3 drops of C_2H_5OH* to 3 drops of $K_2Cr_2O_7$ (24). Carefully mix and heat.

Observed changes: _____

Conclusion: _____

2. Qualitative test on polyols.

Unlike primary alcohols polyols react not only with alkali metals but with some metal hydroxides. In reaction of glycerine with copper (II) hydroxide complex compound is formed:



Accomplishment: to 2 drops of NaOH (21) solution add 2 drops of solution $CuSO_4$ (26), shake, add 2 drops of glycerine (4), shake.

Observed changes: _____

Conclusion: _____

3. Sodium phenoxide production and its decomposition.

Phenols possess more strong acidic properties than alcohols because stability of phenoxide anion raises according to negative charge delocalization along bond conjugate system. Phenols unlike alcohols are capable to react with alkalis. Water-soluble sodium phenoxide is formed. Mineral acids replace phenol from phenoxides.

$$C_6H_5OH + NaOH \rightarrow C_6H_5ONa + H_2O$$

$$C_6H_5ONa + H_2SO_4 \rightarrow C_6H_5OH + NaHSO_4$$

Accomplishment: to 10 drops of phenol water emulsia* add on drops solution of NaOH (21) until transparent solution has been obtained. Add on drops dilute solution of H_2SO_4 (23), and again emulsia is formed.

Observed changes: _____

Conclusion: _____

4. Qualitative test on phenol.

This is a qualitative test on the hydroxyl group bound with unsaturated carbon atom. Phenol as an acid reacts with ion Fe^{3+} forming the complex compound.

Accomplishment: to 10 drops of phenol water emulsia * add 1–2 drops of solution of $FeCl_3$ (8), shake.

Observed changes: _____

Conclusion: _____

5. Comparison of the methyl amine and aniline basic properties.

Aliphatic radicals possessing positive inductive effect +I increase electronic density on the nitrogen atom therefore aliphatic amines are stronger bases than aromatic amines.

Accomplishment: one litmus band is moistened with water solution of methylamine* and another is with water solution of aniline*.

Observed changes: _____

Conclusion: _____

Signature of teacher:

LABWORK № 6
BIOLOGICALLY IMPORTANT REACTIONS OF ALDEHYDES AND KETONES

Objective: to study features of aldehydes and ketones reactivity and develop skills to carring out of qualitative reactions on aldehydes, ketones.

Recommended literature:

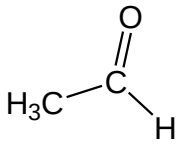
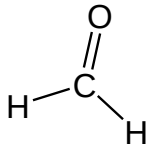
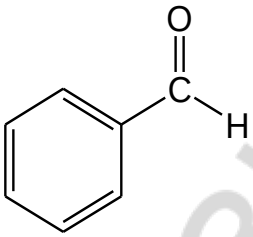
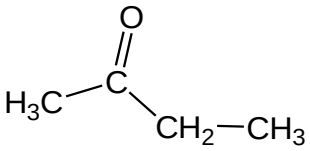
1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 133–147.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 121–133.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 28–30, 35–41.

Problems for discussion:

1. An electronic structure of a carbonyl group. The reactive centers in aldehydes and ketones.
2. Mechanism of nucleophilic addition reaction (A_N). Addition of water and alcohols. Reactions of aldehydes and ketones with amines. Reduction reactions.
3. Reaction of CH-acidic center. Aldol condensation reactions. Haloform reactions.
4. Oxidation reactions are qualitative tests on aldehyde group. Oxidation reactions of ketones. Disproportionation reactions.
5. Formaldehyde. Application in medicine. Toxicity.

Exercises

1. Indicate reactive centers in the carbonyl compound molecules:

2. Write down the equation reaction acetalization: interaction ethanal with 2 mol methanol

3. Describe the mechanism of intramolecular acetalization reaction to form cyclic hemiacetal of 5-hydroxyhexanal.

4. Write the interaction reaction of ethanal and methylamine.

5. Write reaction schemes of ethanal reduction:
in vitro:

in vivo:

6. Write reaction schemes of propanone reduction *in vitro.*

7. Write the scheme of aldol condensation reaction of 2-methylpropanal.

8. Write the scheme of aldol desintegration of 2-amino-3-hydroxypropanoic acid (proteinogenic amino acid serine).

9. Write the scheme of oxidation reaction of ethanal.

10. Write the scheme of dismutation reaction for the formaldehyde.

TEST CONTROL

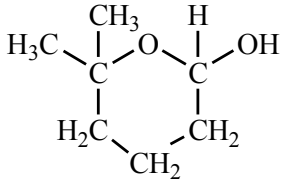
1. Indicate reaction sites in the 2,2-dimethylpropanal molecule:
1) CH-acidic site on α-carbon atom;
2) basic site on the oxygen atom;
3) electrophilic site on the carbonyl carbon atom;
4) nucleophile site on the carbonyl carbon atom.

2. Find the accordance between the carbonyl compounds and its reduction product:

A) 2-methylpropanal;	1) 2-hydroxybutandioic acid;
B) 2-oxopropanoic acid;	2) 2-methylpropan-1-ol;
C) 2-oxobutandioic acid;	3) propan-1-ol;
D) propanal.	4) 2-hydroxypropanoic acid.

3. Select the product of methanal and ethanol (1:2) interaction in acidic medium:
1) 2-methoxyethanol; 3) ethoxymethanol;
2) diethoxymethane; 4) 1,1-dimethoxyethane.

4. Select the hydrolysis product of the represented hemiacetal:



1) 4-hydroxy-5-methylhexanal;
2) 5-hydroxyhexanal;
3) 5-hydroxy-5-methylhexanal;
4) 5-hydroxy-5-methylheptanal.

5. Schiff's bases forms as a result of interaction between:
1) methylamine and ethanal;
2) methylamine and benzoic acid;
3) propanal and ethylamine;

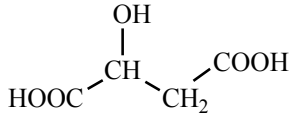
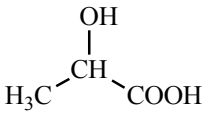
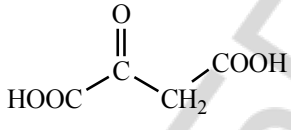
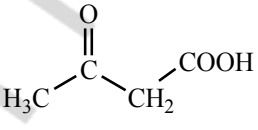
4) methylamine and ethylamine.

6. In aldol condensation reaction could undergo:
1) 2-methylpropanal; 3) benzaldehyde;
2) propanal; 4) 2,2-dimethylpropanal.

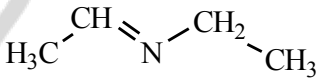
7. For qualitative detection of the aldehyde group are used:
1) Shiffs reagent; 3) $Cu(OH)_2$, heating;
2) $FeCl_3$; 4) Ag_2O in ammonia solution.

8. Choose carbonyl compound with the highest reactive ability in A_N reactions:
1) propanone; 2) butan-2-one; 3) ethanal; 4) methanal.

9. Select the product of 2-oxopropanoic acid reduction:

1) 
2) 
3) 
4) 

10. Represented substance forms as a result of interaction between:



1) methylamine and ethanal;
2) ethylamine and ethanal;
3) ethylamine and methanal;
4) ethylamine and methylamine.

PRACTICAL PART

1. Formaldehyde oxidation with $Cu(OH)_2$ in alkaline medium.

Qualitative tests on aldehydes are connected with easy oxidizability of aldehyde group with oxides or metal hydroxides in medium at heating, thus aldehydes turn into carboxylic acids with the same number of carbon atoms and the ion of metal is reduced. The Trommer's reagent (fresh obtained copper (II) hydroxide) is used as an oxidizer.

$$CuSO_4 + 2\ NaOH \rightarrow Cu(OH)_2 + Na_2\ SO_4$$

$$R - CHO\ + 2Cu(OH)_2 \xrightarrow{OH^-,t} R - COOH + 2CuOH\ + H_2O$$

$$2\ CuOH \longrightarrow Cu_2O + H_2O$$

Accomplishment: to 3 drops of formaline (32) add 5 drops of NaOH solution (21) and 1–2 drops of $CuSO_4$ (26). Mixture is heated to boiling point.

Observed changes: _____

Conclusion: _____

_____

2. Reaction of formaldehyde with Shiff's reagent.

Reaction goes according to the A_N mechanism with the Shiff's reagent without heating.

Accomplishment: to 2 drops of the Shiff's reagent* add 3 drop of formaldehyde solution (32).

Observed changes: _____

Conclusion: _____

3. Disproportiation reaction of formaldehyde.

Disproportionation reaction is interaction of two aldehyde molecules when one aldehyde molecule is reduced to alcohol due to another aldehyde molecule is oxidized to a carboxylic acid. Water formaldehyde solution has acidic medium of reaction.

$$2\ HCHO \rightarrow HCOOH + CH_3OH$$

Accomplishment: to 3–4 drops of formaline (32) add 1 drop of methyl red indicator*.

Observed changes: _____

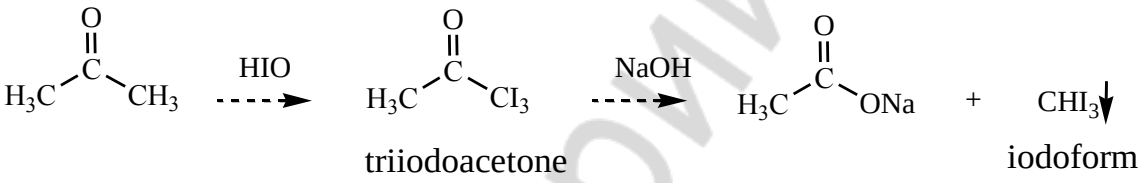
Conclusion: _____

_____

4. Acetone detection by transformation to iodoform (iodoform reaction).

Iodoform reaction is connected with ability of carbonyl containing compounds to substitute hydrogen atom at α-carbon atom on halogen and the following cleavage of carbon-carbon bond with iodoform (CHI_3) formation.

$$I_2 + NaOH \longleftrightarrow HIO + NaI$$



triiodoacetone iodoform

Accomplishment: to 3 drops of Lugol (47) solution (I_2 in KI solution) add NaOH solution (21) to disappearing of color, then pour 1-2 acetone drops*.

Observed changes: _____

Conclusion: _____

_____

5. Colored reaction on the acetone with sodium nitroprusside.

Reaction with sodium nitroprussiate $Na_2[Fe(CN)_5NO]$ is used in a clinical practice to discovery of acetone in urine at a diabetes. Aromatic carbonyl compounds do not yield this reaction.

Accomplishment: to 3 drops acetone* add 2 drops of sodium nitroprussiate $Na_2[Fe(CN)_5NO]$ (35) and 2 drops of NaOH (21) solution. In 2–3 minutes add 2 drops of acetic acid (36).

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 7
CARBOXYLIC ACIDS AND THEIR DERIVATIVES

Objective: to study features of carboxylic acids reactivity and develop skills to carring out of qualititative reactions on carboxylic acids.

Recommended literature:

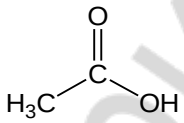
1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 149–159.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 135–135.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 47–52.

Problems for discussion:

1. Reactions sites of carboxylic acids and derivatives.
2. Acidic properties of carboxylic acids.
3. Decarboxylation reaction. Biogenic amines. Cyclic anhydrides.
4. Nucleophilic substitution reactions. Esterification reaction.
5. Amides, acyl chlorides, anhydrides. Their hydrolysis.

Exercises

1. Indicate reactive sites at the carboxylic acid molecule:



2. Compare the acidity of ethanoic and ethanedioic acids. Write the reaction of salt formation of the stronger acid with $Ca(OH)_2$.

3. Write down the decarboxylation reaction of the following compounds:

a) propanedioic acid (malonic)

b) 2-aminopentanedioic acid

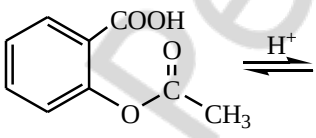
4. Write the dehydration reaction of pentanedioic acid.

5. Write the formulas of the functional derivatives of carboxylic acids:

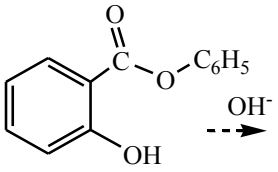
anhydride of acetic acid	acetyl chloride
ethylethanoate	ammonia acetate
full amide of oxalic acid	full amide of carbonic acid

6. Write the esterification reactions of the methanoic acid with ethanol.

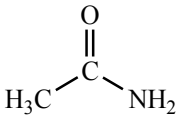
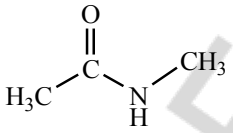
7. Write down the acidic hydrolysis reaction of the following compound:



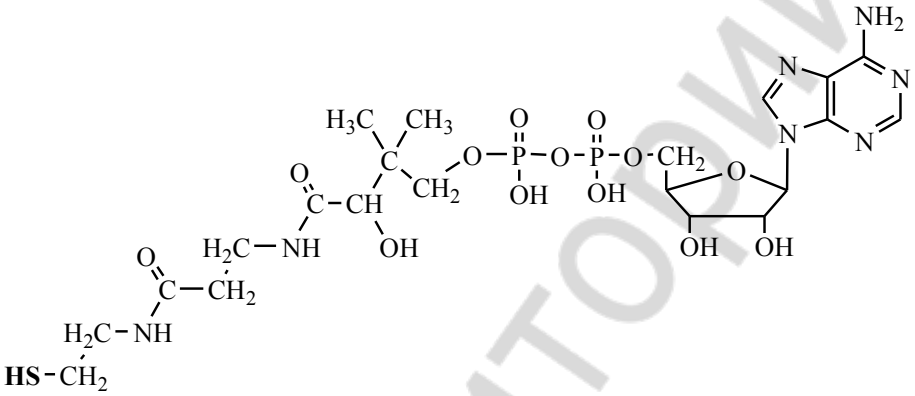
8. Write down the alkaline hydrolysis reaction of the following compound:



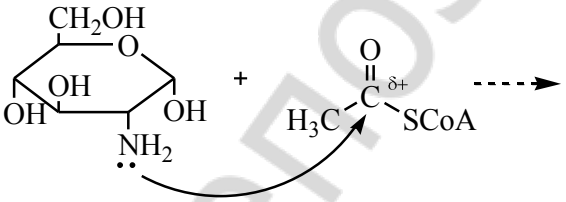
9. What products would be obtained from the hydrolysis of each of the following amides:

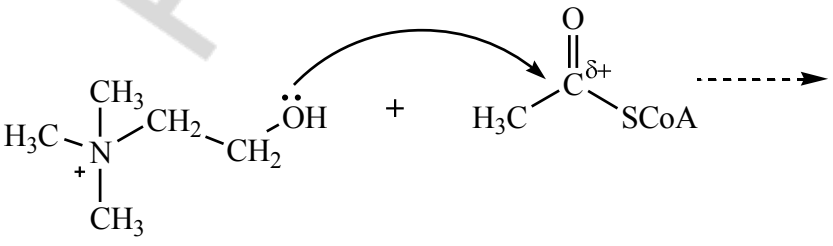
	

10. Mark the ester, amide, anhydride bonds at the coenzyme A molecule. Write the scheme of acetylcoenzyme A formation (using short formula for coenzyme A — CoA-SH.



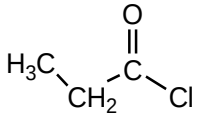
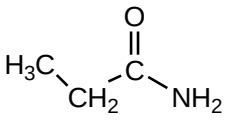
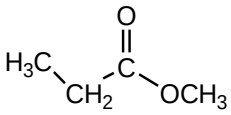
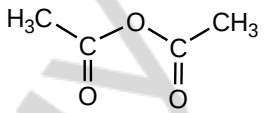
11. Write the schemes of acylation reactions:





TEST CONTROL

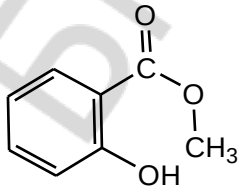
1. Arrange in order of decreasing of reactivity in S_N reactions of the following compounds:

1) 
2) 
3) 
4) 

2. Find accordance between compound and its decarboxylation products:

A) ethandioic acid;	1) propanone;
B) 2-amino-3-hydroxypropanoic acid;	2) 2-aminoethanol;
C) propandioic acid;	3) ethanoic acid;
D) 3-oxobutanoic acid.	4) methanoic acid.

3. Methyl salicylate forms as a result of acidic hydrolysis of:



1) methanol and *o*-hydroxybenzoic acid;
2) *o*-hydroxybenzoic acid and methanoic acid;
3) *p*-hydroxybenzoic acid and methanol;
4) *o*-hydroxybenzoic acid and phenol.

4. Choose correct statement(s):

1) RS-group possess less +M effect than RO-group;
2) RS-ions more stable than RO-ions and are more easily leaving group;
3) RO-ions more stable than RS-ions and are more easily leaving group;
4) partial positive charge on carbonyl carbon atom in thioesters is higher than its in esters.

5. Electron density distribution in propanoic acid molecule is characterized by presence:

1) O – H acidic site in the carboxyl group;
2) nucleophile site on the carbon atom of carboxylic group;
3) C – H acidic site in the alkyl group;
4) basic site on the oxygen atom in the carboxyl group;
5) electrophilic site on the carbon atom of carboxylic group.

6. Indicate type of the following reaction $CH_3COCl + CH_3OH \rightarrow CH_3COOCH_3 + HCl$:

1) elimination;
2) nucleophilic substitution;
3) electrophilic substitution;
4) nucleophilic addition.

7. Indicate acids which are stronger than acetic acid?

1) 2-chloroacetic acid;	3) propanoic acid;
2) hydrochloric acid;	4) formic acid.

8. To increase reactive ability of carboxylic acids we should:

1) conduct reaction in alkaline medium;
2) conduct reaction in acidic medium;
3) enter more strong electron withdrawer in a side chain;
4) enter more strong electron withdrawer in a carboxyl group.

9. Select functional derivatives of carboxylic acids:

1) ethanoic acid;	3) acetic anhydride;
2) ethyl chloride;	4) methyl benzoate.

10. Choose products of the butandioic acid heating in acidic medium:

1) H_2O;	3) propanoic acid;

2) CO_2; 4) succinic anhydride.

PRACTICAL PART

1. Ethyl acetate formation.

To detect the carboxylic acids the esters production reaction can be used if esters have specific smell. The reaction is carried out according to the nucleofilic substitution mechanism (S_N).

$$CH_3\,COONa + H_2SO_4 \rightarrow CH_3\,COOH + NaHSO_4$$

$$CH_3\,COOH + C_2H_5OH \rightarrow CH_3COOC_2H_5 + H_2O$$

Accomplishment: to 3 drops of ethanol* add 5 drops of H_2SO_4 concentrated solution* and waterless CH_3COONa (42), heat. Pour solution to another test-tube with water.

Observed changes: _____

Conclusion: _____

_____

2. Oxalic acid decarboxylation (demonstration).

Result of the oxalic acid decarboxylation is carbon dioxide which forms $CaCO_3$ when mixed with the lime water (solution of $Ca(OH)_2$)

$$HOOC - COOH \xrightarrow{t} CO_2 + HCOOH$$

$$CO_2 + Ca(OH)_2 \rightarrow CaCO_3\downarrow + H_2O$$

Accomplishment: in dry test-tube add crystal oxalic acid* (mass ≈ 0,5 g). Test-tube is closed by flatus tube and heat. The end of flatus tube put into test-tube with 15 drops of lime water ($Ca(OH)_2$) (2).

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 8
CONCLUDING TEST "THEORETICAL FUNDAMENTALS OF BASIC CLASSES OF ORGANIC COMPOUND STRUCTURE AND REACTIVITY"

Remind the program material from the theme № 1 to № 7.

Recommended literature: study the literature from the theme № 1 to № 7.

Questions to the concluding test:

1. Conformations. Newman projections. Types of strains. Energetic characteristic of eclipsed, gauche and staggered conformations (butane). Conformational structure of hydrocarbon radicals of fatty acids. Cyclohexane conformations. Types of strains (angle, torsion, Vander-Waals). Inversion of cycle. 1,3-diaxial interaction.
2. Configuration of organic compounds. Stereoisomerism. Fischer projections. Relative configuration and D, L-convention. Glyceraldehyde as the configurational standart.
3. Stereoisomerism of molecules with one chiral centre (lactic acid as an example). Enantiomers. Optic activity. Racemic mixtures.
4. Absolute configuration of stereoisomers. R, S-convention. Relationship of spatial structure with biological activity.
5. Electronic effects in organic molecules (inductive and mesomeric), their role in the reactivity centers in the molecule. Electron donors and withdrawers.
6. Conjugation (π,π- and p,π-conjugations). Conjugated systems with open chain (butadiene-1,3).
7. Conjugated systems with close chain. Aromaticity, criteries of aromaticity, Huchel's rule (benzene, naphtaline, phenantrene).
8. Acidity and basicity of organic compounds; Brensted and Lewis theories. Acidic properties of organic compounds (alcohols, phenols, thiols, carboxylic acids, amides). Factors of anion stability.
9. Basic properties of organic compounds (alcohols, ethers, thioesters, amines). Comparing of basic properties of aliphatic and aromatic amines; salt formation.
10. Classification of organic reactions (substitution, addition, elimination, isomerisation, red-ox, acid-basic interaction). Classification of organic reactions on the mechanism of covalent bond cleavage (radical and ionic). Electronic and spatial structure of free radicals, carbocations and carboanions.
11. Oxidation reactions of organic compounds (alcohols, thiols, phenols). Antioxidants (2,3-dimercaptopropanol, ascorbic acid, phenols and others).
12. Radical substitution reactions. Propane chlorination as an example of free radical substitution. Initiators of radical reactions. Antioxidants.
13. Electrophilic addition reactions of alkenes. Hydration reactions of alkenes. Acidic catalys. Markovnikov's rule.
14. Electrophilic substitution reactions of aromatic hydrocarbons. Substituent effects in the aromatic ring on the reactivity of aromatic hydrocarbons.
15. Alkylation reactions of aromatic compounds.
16. Electronic and spatial structure of the carbonyl group. Comparative reactivity of aldehydes and ketones.
17. Oxidation and reduction reactions of carbonyl compounds. Visual tests on the aldehyde group (silver mirror test, Trommer test). Reduction reactions *in vivo*, NADH as a hydride ion donor.

18. Nucleophilic addition reactions of aldehydes and ketones; addition of water and alcohols.

19. Addition of amines to carbonyl compounds, mechanism. Schiff's bases.

20. Electronic and spatial structure of the carboxylic group. Acidic properties of the carboxylic acids: mono-, dicarboxylic, aliphatic saturated, aliphatic unsaturated, aromatic carboxylic acids.

21. Nucleophilic substitution at sp^2-hybridized carbon atom in the carboxylic group: esterification reaction. Properties of esters, hydrolysis.

LABWORK № 9
HETEROFUNCTIONAL COMPOUNDS OF ALIPHATIC, BENZENE AND HETEROCYCLIC SERIES, METABOLITES AND BIOREGULATORS

Objective: to develop skills to predict chemical properties biologically important heterofunctional compounds taking into account a structure and interference of various functional groups.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 161–171.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 163–172.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 52–61.

Problems for discussion:

1. Polyfunctional compounds: classification, chemical properties.
2. Heterofunctional compounds: classification, a role in biological processes.
3. Aminoalcohols: their biological role.
4. Hydroxyacids. A structure, typical and specific properties of α-, β-, γ-hydroxy and amino acids.
5. A citric acid: a structure, properties. Citrates.
6. Oxoacids. Acid properties and reactivity. Ketone bodies.
7. Keto-enol tautomerism.
8. Amides of carbonic acid. Urea.
9. Salicylic acid, its derivatives.
10. Para-aminobenzoic acid, its derivatives.
11. Sulfanylamides.

Exercises

1. Write the structural formulas of the following polyfunctional compounds:

glycerol	ethylene glycol
inositol	catechol

hydroquinone	resorcinol
oxalic acid	malonic acid
succinic acid	glutaric acid
fumaric acid	maleic acid

2. Write the structural formulas of the amino alkohols:

2-aminoethanol	choline

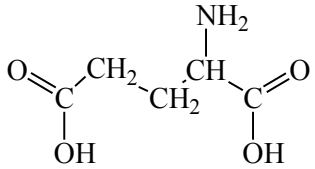
3. Write the structural formulas of the hydroxy acids:

lactic acid salts — …	malic acid salts – …
citric acid salts — …	

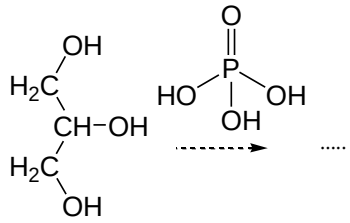
4. Write the structural formulas of the oxo acids:

pyruvic acid	oxaloacetic acid	α-oxoglutaric acid

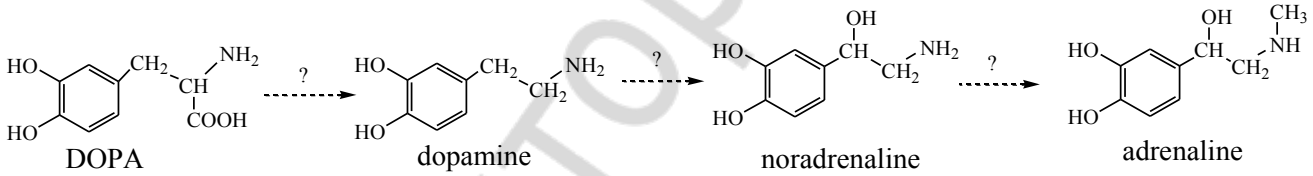
5. Indicate the acidic and basic centers in the following molecule and write its ionic forms:



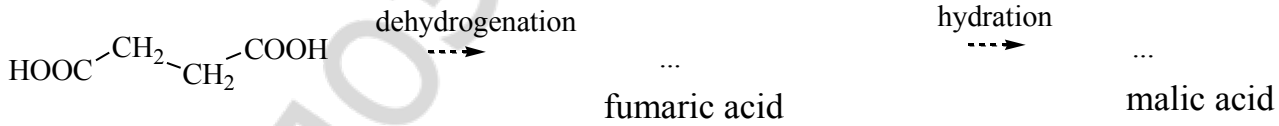
6. Fill in the scheme of the α-glycerophosphate formation:



7. Show the catechol at the catecholamine molecules and its precursor DOPA (3,4-dihydroxy-phenylalanine). Write the name of the reactions occurring during the synthesis of catecholamines *in vivo* (alkylation, decarboxylation, hydroxylation). Mark the chiral centers in molecules.



8.Complete the scheme of the reactions *in vivo*:

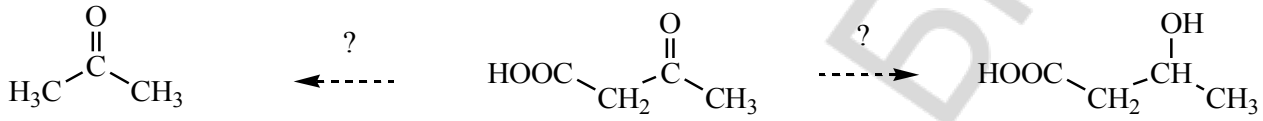


9. Write scheme of the oxidation reaction *in vivo* of malic acid.

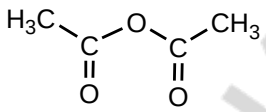
10. Write scheme of the reduction reaction *in vivo* of pyruvic acid.

11. Write down the tautomeric forms of oxaloacetic acid:

12. Write the names of the ketone bodies formation according to the following scheme:



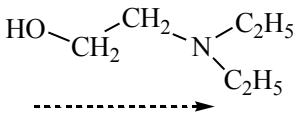
13. Fill in the scheme of the acetylsalicylic acid formation reaction.



salicylic acid

acetylsalicylic acid

14. Fill in the scheme of the *p*-aminobenzoic acid derivatives formation reaction:



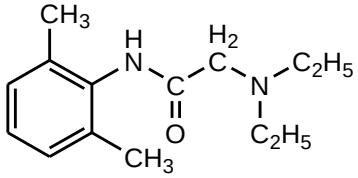
ethanol

benzocaine

p-aminobenzoic acid

procaine

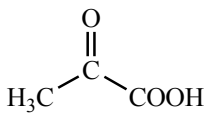
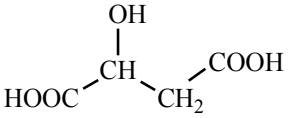
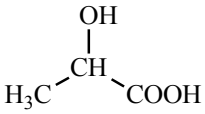
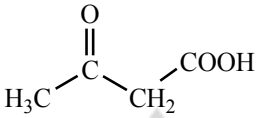
15. Explain the structure peculiarities of the modern anesthetic remedies such as lidocaine.



lidocaine

TEST CONTROL

1. Indicate the product of malic acid oxidation *in vivo*:

1) 
2) 
3) 
4) 

2. Salicylic acid is stronger than benzoic acid because of:
1) both functional groups are acidic;
2) mesomeric effect of phenol OH-group decrease anion stability;
3) formation of intermolecular hydrogen bond between ionized carboxyl group and phenol hydroxyl group;
4) mesomeric effect of phenol OH-group increase anion stability.

3. Novocain possess less long-term anesthetic action in comparison with ultracaine because of:
1) it has ethers bonds;
2) it is Shiff"s base which hydrolyzes easy;
3) it has esters bond which hydrolyze easier then amide bond;
4) it has glycoside bond.

4. As a result of decarboxylation of 2-amino-3-hydroxypropanoic acid decarboxylation forms CO_2 and:
1) propanon; 2) 2-aminoethanol; 3) ethanoic acid; 4) methanoic acid.

5. Indicate correct statements about oxaloacetic acid:
1) refer to ketoacids;
2) posses optical activity;
3) exist in toutomeric forms in solution;
4) undergo in nucleophilic substitution reaction.

6. Choose the carbonic acid derivatives:
1) carbamic acid; 2) carbamide; 3) uric acid; 4) urea.

7. As result of interaction of salicylic acid and acetic anhydride forms:
1) acetylsalicylic acid; 3) methyl salicylate;
2) phenyl salicylate; 4) ethyl salicylate.

8. Indicate correct statements about urea:
1) gives acidic properties of medium;
2) possess basic properties;
3) is the final product of nitrogen metabolism in human body;
4) oxygen is protonated after interaction with acid;
5) nitrogen is protonated after interaction with acid.

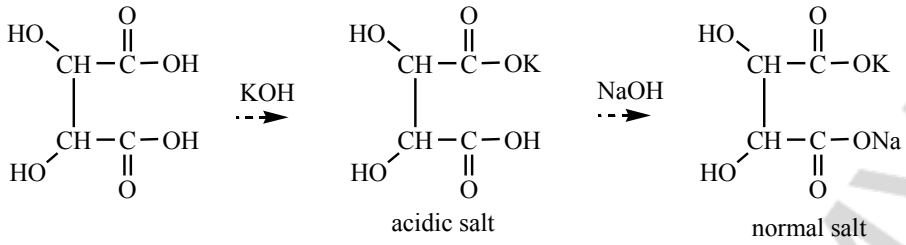
9. Which acids undergo elimination reaction:
1) 4-hydroxypentanoic acid; 3) 3-hydroxybutanoic acid;
2) 2-hydroxy-3-methylbutanoic acid; 4) 3-aminopentanoic acid.

10. Which one of the following compounds forms gamma-lactone under heating:
1) 4-hydroxy-2-methylbutanoic acid; 3) 3-hydroxybutanoic acid;
2) 2-hydroxybutanoic acid; 4) 5-hydroxypentanoic acid.

PRACTICAL PART

1. Evidense of two carboxyl groups in tartaric acid structure.

Tartaric acid as dioic forms two salts — acid salt and neutral [normal] salt which differ with water solubility.



Accomplishment: to 3 drops of tartaric acid (50) add 2–3 drops of KOH solution (51), intensively intermix rubbing with glass rod against walls of a test tube. There is a crystal deposit. Add 2–3 drops of NaOH solution (21) into a test tube to form the solution of **segnetic salt** (sodium and potassium tartrate). Save this solution for next experiment.

Observed changes: _____

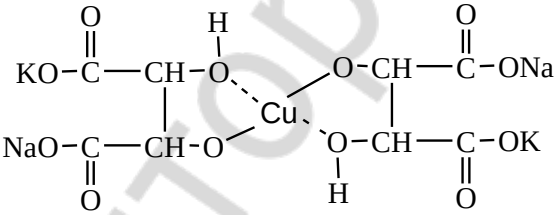
Conclusion: _____

_____

2. Evidense of two hydroxyl groups in tartaric acid structure.

Qualitative test on polyols is used. Tartaric acid reacts with $Cu(OH)_2$ and forms copper (II) alcoholate (chelate).

$$CuSO_4 + 2NaOH \rightarrow Cu(OH)_2 + Na_2SO_4$$



Copper alcoholate of sodium and potassium tartrate is called the Fehling's reagent and is used for qualitative and quantitative definition of carbohydrates.

Accomplishment: Pour 2 drops of 5 % solution of $CuSO_4$ (26) and 2 drops of 10 % solution of NaOH (21) in the test tube. Then to the formed mixture add the solution of segnetic salt received at the last experiment.

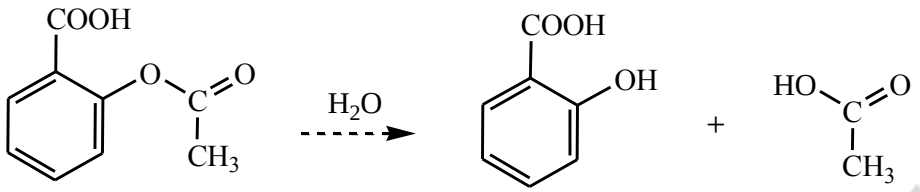
Observed changes: _____

Conclusion: _____

_____

3. Test on the high quality of aspirin.

At hydrolysis of aspirin *o*-hydroxybenzoic acid is formed which with Fe (III) chloride forms complex compound.



Accomplishment: place some grains of aspirin* and 5–6 drops of water in a test tube, shake it. Divide the test tube contents into 2 parts. To one part add 1 drop of $FeCl_3$ (8), another part boil for half a minute and then add 1 drop of $FeCl_3$.

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 10
BIOLOGICAL IMPORTANT HETEROCYCLIC COMPOUNDS. ALKALOIDS

Objective: to develop knowledge about structures and properties of biological important compounds, derived heterocycles.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 172–187.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 230–244.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 61–69.

Problems for discussion:

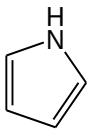
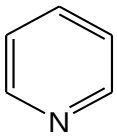
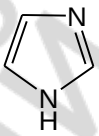
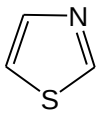
1. Heterocycles with 1 heteroarom: pyrrol, furan, thiophene, indol, pyridine, quinoline. Heterocycles with more than 1 heteroarom: pyrazole, imidazole, pyrimidine, purine.
2. Acidity and basicity of heterocycles.
3. Porphyrins: aromaticity, biological importance.
4. Furan. Furfural, 5-nitrofuranic derivatives. Thiophene. Biothin.
5. Indol is structural component of biogenic amines serotonine, tryptamine and so on.
6. Imidazole. Prototropic tautomerism. Imidazole is structural component of biogenic amine histamine.
7. Biolodically important derivatives of pyridine: nicotinamide, pyridoxal. Participation of nicotinamide at the biological oxidation.
8. Quinoline and drugs derived quinoline.
9. Pyrimidine. Vitamin B_1. Barbituric acid and its derivatives.
10. Hydroxy pyrines: hypoxantine, xantine, uric acid, their taunomerism. Uric acid salts.
11. Alkaloids.

Exercises

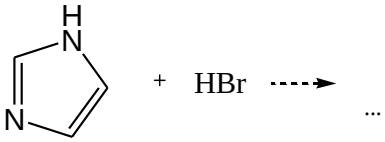
1. Write formulas of the following heterocycles:

pyrrole	thiophene	furan	indol
pyrazole	imidazole	pyridine	pyrimidine
quinoline	isoquinoline	purine	

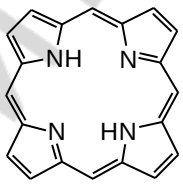
2. Indicate acidic and basic centers:

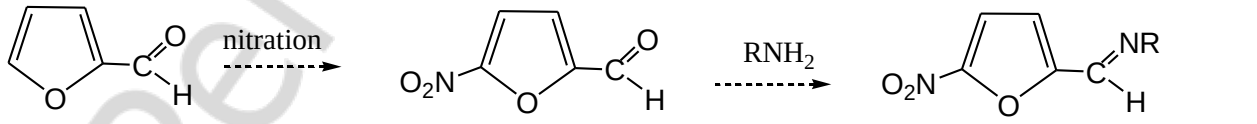
Complete the reaction scheme:



3. Indicate the pyrrol and pyridine nitrogen atoms at the porphine molecule. Estimate the aromaticity of porphine.

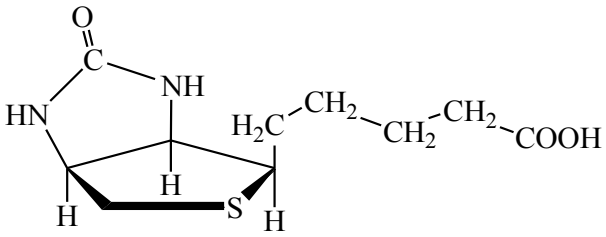


4. Indicate the reaction mechanism of formation of 5-nitrofurfural derivatives, which possess antiseptic action.



furfural — 5-nitrofurfural — the general formula of 5-nitofuranic derivatives

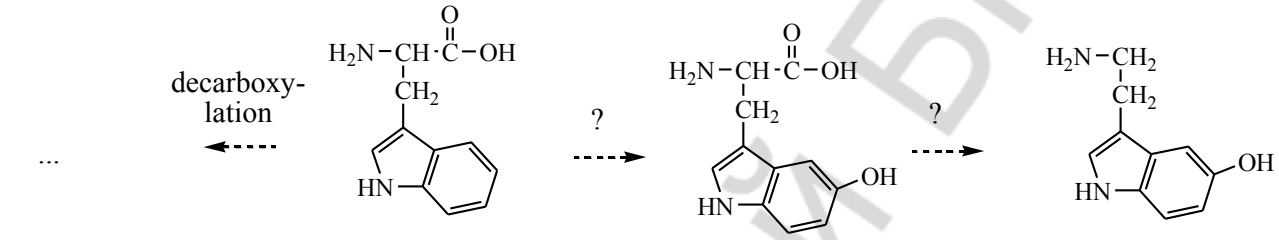
5. Select the fragments of tetrahydrothiophene, urea and pentanoic acid at the biothin molecule (vitamin H). Designate the chiral centers (3).



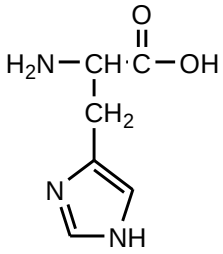
biothin

Biothin participates at the carboxylation reaction *in vivo.*

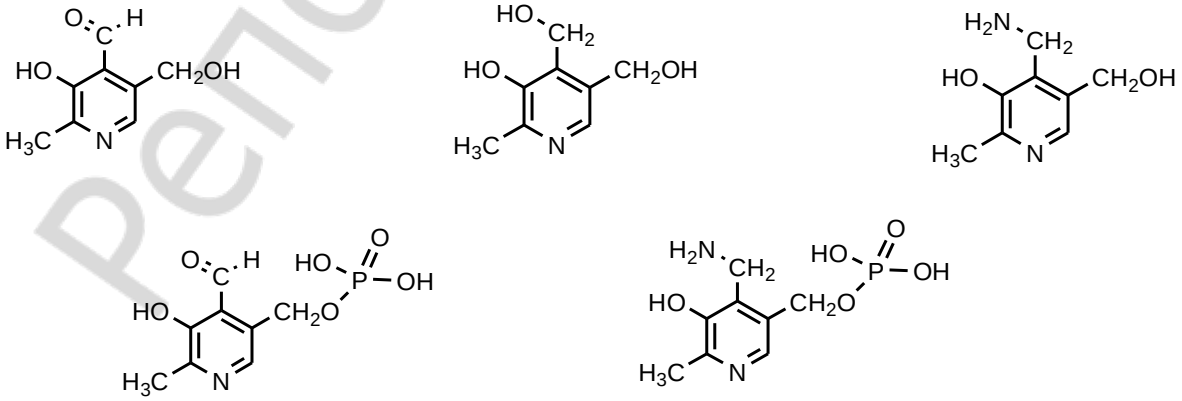
6. Fill in the scheme reactions with tryptophane participation. Write the names of the reactions *(decarboxylation, hydroxylation).*



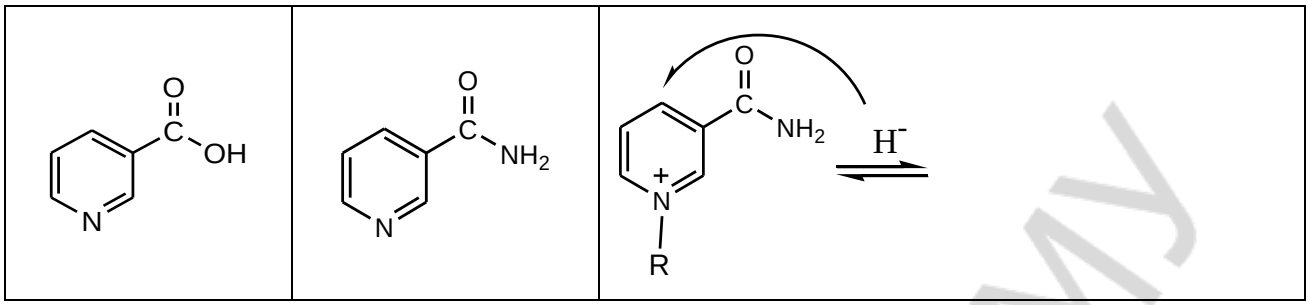
7. Indicate acidic and basic centers at the histigine molecule. Write it at the ionic form. Write the decarboxylation reaction of histidine.



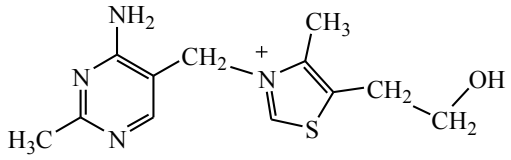
8. Vitamin B_6 containes pyridine. Call different forms of vitamin B_6 (*phosphate of pyridoxamine, pyrodoxol, pyridoxal, pyridoxamine, phosphate of pyridoxal*).



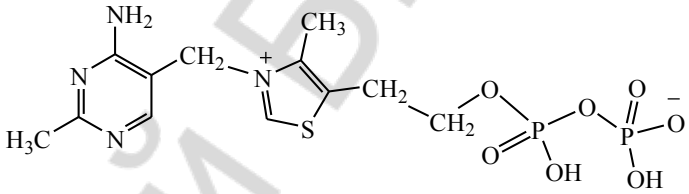
9. Give the names of vitamin PP forms; write the reaction interaction of vitamin PP and hydride ione.



10. Thiamine (vitamine B_1) participares in the decarboxylation reaction of α-oxo acids. Select and call heterocycles, which are structural components of thiamine.



thiamine

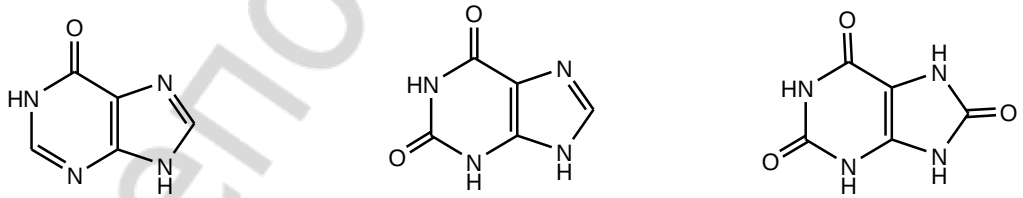


diphosphate of thiamine

11. Write the formula of barbituric acid and its derivatives.

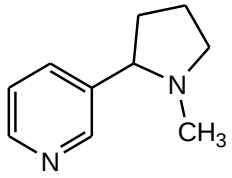
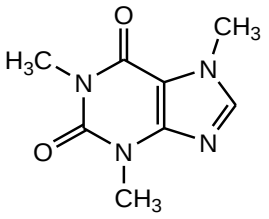
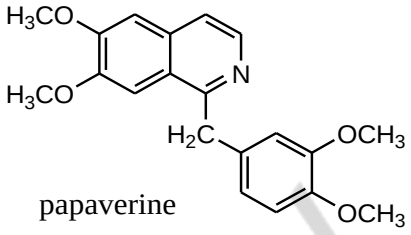
barbituric acid	5,5-diethylbarbituric acid	5-phenyl-5-ethylbarbituric acid

12. Write down the names of the following compounds:



Salts of uric acid is called … — …

13. Select well-known heterocycles at the following formulas of alkaloids:

 nicotine	 caffeine	 papaverine

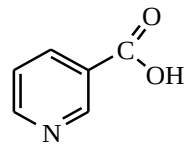
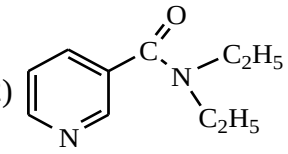
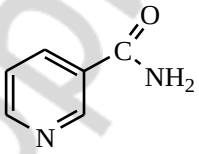
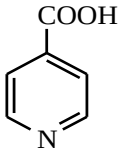
TEST CONTROL

1. Biogenic amine serotonin contains:
1) purine; 2) naphthaline; 3) quinoline; 4) indole.

2. Select the correct statements for pyridine: 1) all atoms are sp^2-hybridezed; 2) N gives 2 electrons to conjugation system; 3) it is π-deficient aromatic system; 4) N supplies 1 electron to conjugation system; 5) it is π-rich aromatic system.
1) 1, 4, 5; 2) 1, 2, 3; 3) 1, 3, 4; 4) 1, 2, 5.

3. Indicate correct statements about structure and properties of uric acid:
1) it reacts with 3 mol of NaOH;
2) water insoluble;
3) alkaline insoluble;
4) it has lactam and laclim forms.

4. Barbituric acid has the following forms of tautomerism:
1) keto-enol; 2) amino-imino; 3) lactam-lactim; 4) cyclo-chaine.

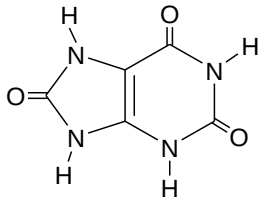
5. Select 2 forms of vitamin PP:
1) 
2) 
3) 
4) 

6. Indicate the derivatives of pyridine:
1) thiamine; 2) pyridoxal; 3) nicotinic acid; 4) furfural.

7. Select the compounds with the most basic properties:
1) imidazole; 2) dimethylamine; 3) pyrrol; 4) methylamine.

8. Indicate compounds which are formed from purine bases in vivo:
1) urea;
2) xantine (2,6-dihydroxypyrine);
3) hypoxantine (6-hydroxypyrine);
4) uric acid (2,6,8-trihydroxypurine).

9. Indicate the trivial name of compound:
1) uric acid;
2) urea;
3) hypoxantine;
4) xanthine;
5) caffeine



10. Alkaloids are the derivatives of:
1) oligosaccharides;
2) monosaccharides;
3) oxygen containing heterocycles;
4) nitrogen containing heterocycles;

5) fatty acids.

PRACTICAL PART

1. Copper salt of nicotinic acid formation.

Accomplishment: place 1 spatula of nicotinic acid* in a test tube, add 10–15 drops of water, heat up to boiling. To a hot solution add 1–2 drops of acetic acid (36) and 3–4 drops of $CuSO_4$ (26).

Observed changes: _____

Conclusion: _____

_____

2. Formation of uric acid salts.

Accomplishment: place some grains of uric acid (7) and add 10 drops of water in a test tube. You can see that uric acid water insoluble. But after addition 1 drop 1 % solution of NaOH (21) transparent solution of uric acid disodium salt is obtained. A half of solution place to another tube and add 1 drop solution of NH_4Cl (10). Precipitation of ammonium urate occurs.

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 11
CARBOHYDRATES. MONOSACCHARIDES

Objective: to develop knowledge of a stereochemical structure, consider important properties of monosaccharides and gain skills to carry out qualitative reactions on monosaccharides.

Recommended literature:

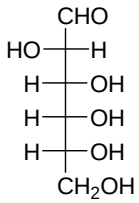
1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 189–199.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 195–207.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 76–85.

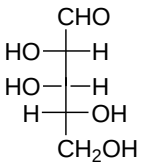
Problems for discussion:

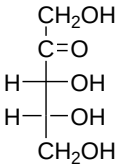
1. Carbohydrates: definition, biological role.
2. Monosaccharides, definition, classification, stereoisomerism. Epimers.
3. Monosaccharide tautomerism. Anomers. Tautomeric forms of D-glucose, D-galactose, D-fructose, D-ribose, 2-deoxy-D-ribose. Fisher and Haworth formulas. Conformations of cyclic forms.
4. Chemical properties of monosaccharides. Glycosides (O- and N-glycosides).
5. Monosaccharide esters. A biological role of monosaccharide phosphates.
6. Monosaccharide oxidation: aldonic, aldaric and uronic acids.
7. Monosaccharide reduction. Xylitol and sorbitol.
8. Aminosugars. Their structure, properties and a biological role.
9. Ascorbic acid (vitamin C) as water-soluble antioxidant.

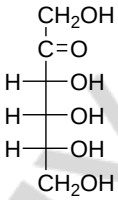
Exercises

1. Classify the following monoses according to the type of carbonyl group and the number of carbon atoms. Show the chiral centers.





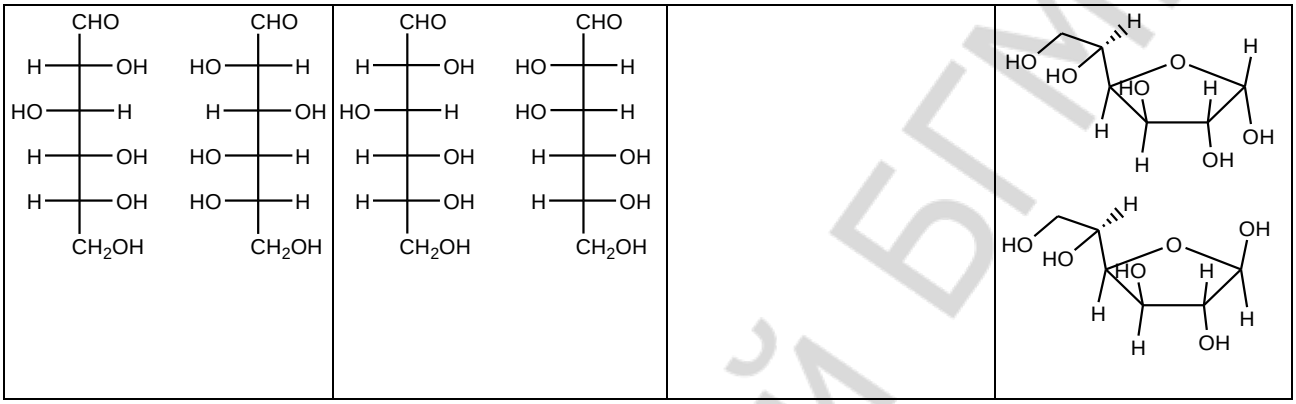




2. Write the all tautomeric forms of D-glucose (according to Fisher and Haworth). Indicate the pairs of anomers.

3. Write the all tautomeric forms of D-fructose.

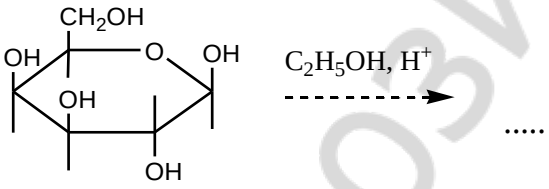
4. Call the pairs of isomers *(epymers, anomers, functional isomers, enantiomers).*



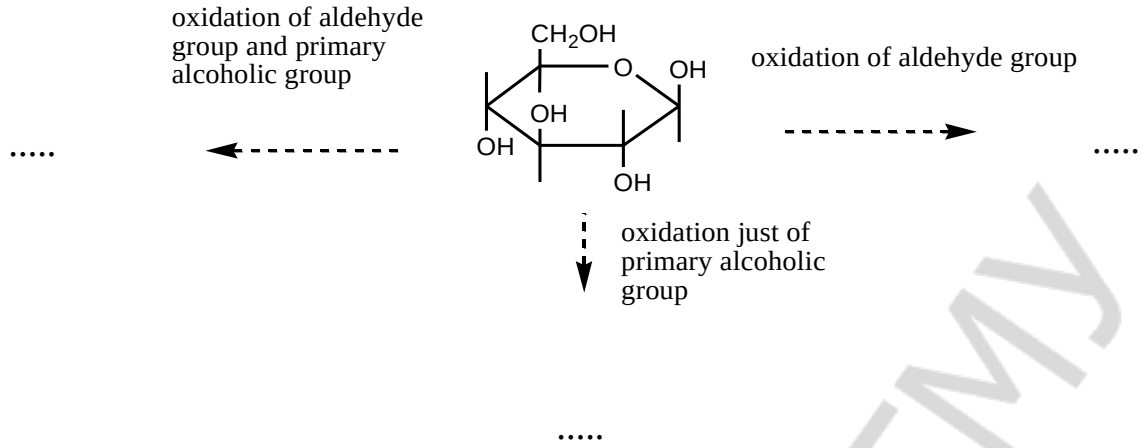
5. Write the formulas of β-D-ribofuranose и β-D-deoxyribofuranose.

6. ***Glycosides*** — are the …

Complete the scheme of the reaction:



7. Write down the formulas of product of the reactions.

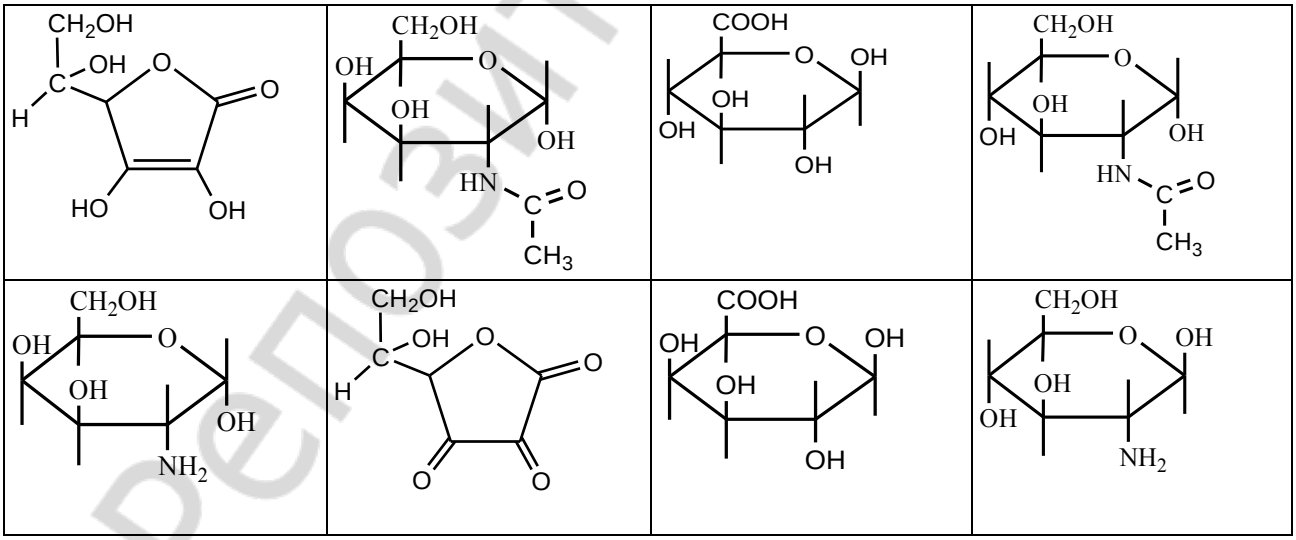


8. Write down the formulas of reduction products of monoses.

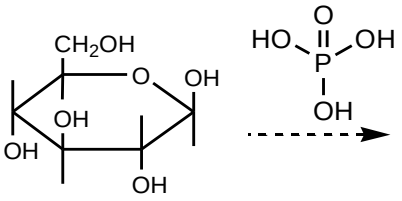
D-xylose $\xrightarrow{H_2,\ Pd}$ .....

D-glucose $\xrightarrow{H_2,\ Pd}$ .....

9. From the formulas raw select the following compounds: 2-deoxy-2-amino-β-D-glucopyranose, 2-deoxy-2-amino-α-D-galactopyranose, N-acetylgalactosamine, N-acetylglucosamine, glucuronic acid, galacturonic acid, reduce form of ascorbic acid, oxidized form of ascorbic acid.



10. Phosphorylation is the way of formation of metabolic active forms of compound. Write down the products of phosphorylation reactions.



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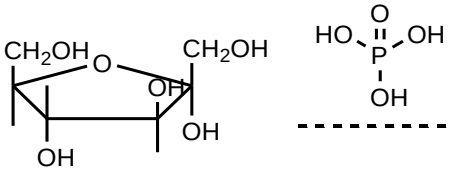
6 - phosphate-D - glucopyranose



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1,6 - diphosphate-D - fructofuranose

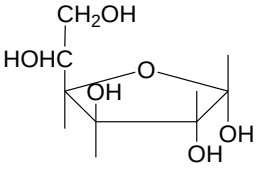
TEST CONTROL

1. Select monosaccharaides which refer to aldohexoses:
1) mannose; 2) galactose; 3) xylose; 4) glucose; 5) fructose.

2. Find characteristics for D-glucose:
1) refer to hexose; 2) is aldose; 3) refer to pentose; 4) is ketoses.

3. Choose a type of glucose fermentation where hydrogen liberate?
1) lactic-acid; 2) alcoholic; 3) butyric-acid; 4) citric-acid.

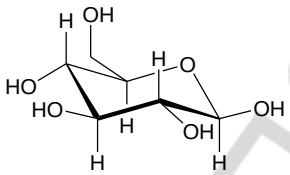
4. How many chiral carbon atoms in cyclic glucose form?
1) 4; 2) 5; 3) 3; 4) 6; 5) 2.

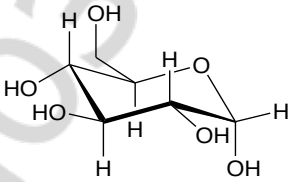


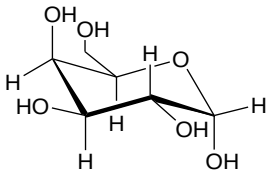
5. Give the name of the following compound:
1) α-D-galactopyranose;
2) α-D-glucofuranose;
3) α-D-fructofuranose;
4) β-D-glucopyranose.

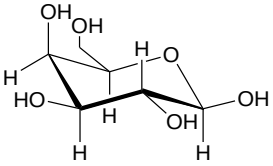
6. D-glucose and D-mannose are stereoisomers which are called:
1) enantiomers;
2) epimers;
3) functional isomers;
4) anomers.

7. Find β-D-galactopyranose:

1) 

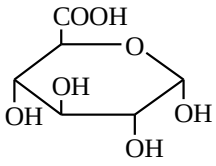
2) 

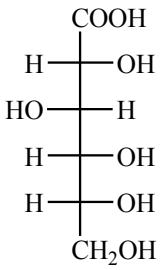
3) 

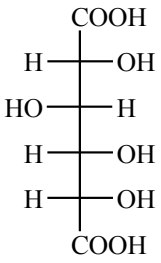
4) 

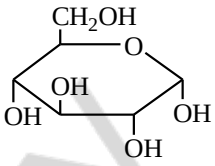
8. Point out the product of interaction between α-D-glucopyranose and methanol (with HCl presence):
1) 2,3,4,6-tetramethyl-D-pyranose;
2) 2,3,4,6- tetramethyl-O-methyl-D-glucopyranoside;
3) methyl-α-D-glucopyranoside;
4) methyl-β-D-glucopyranoside.

9. Point out glucuronic acid:

1) 

2) 

3) 

4) 

10. Select correct statements about transformation acyclic form of monosaccharide in cyclic form:

1) acetal is cyclic form of monosaccharide
2) carbon atom pass into sp^3-hybridization from sp^2-hybridization and becomes asymmetric
3) anomer forms of monosaccharide are created
4) acetal is acyclic form of monosaccharide

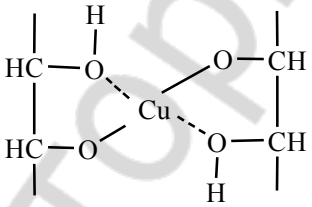
PRACTICAL PART

1. A qualitative test on the hydroxyl groups in the glucose molecule.

Definition of some hydroxyl groups in the monosaccharide composition is carried out with $Cu(OH)_2$. This reaction is the same that on the polyatomic alcohols.

$$CuSO_4 + 2NaOH \longrightarrow Cu(OH)_2\downarrow + Na_2SO_4$$

First forming sediment $Cu(OH)_2$ is dissolved when polyatomic alcohol is added. This is the evidence of some hydroxyl group presence in the compound.



Accomplishment: to 5 drops of glucose (54) solution add 2 drops of NaOH (21) and 2 drops of $CuSO_4$ (26).

Observed changes: _____

Conclusion: _____

_____

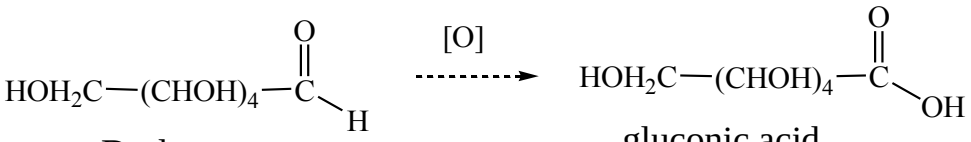
2. A qualitative test on the aldehyde group in the glucose molecule.

This reaction is carried out with the Fehling's reagent which is an alkaline solution of Cu^{2+} alcoholate with K-, Na-tartrates. Obtained chelate is stable and when heated the color doesn't change. However if it is heated at the aldose presence alcoholate will be hydrolyzed. And obtained $Cu(OH)_2$ oxidizes glucose.

$$2Cu(OH)_2 \longrightarrow [O] + H_2O + 2CuOH$$

$$2CuOH \longrightarrow H_2O + Cu_2O$$

Oxygen molecule oxidizes glucose and monosaccharide molecules are completely broken up into acids and oxoacids. The first intermediate of glucose oxidation is gluconic acid.



D-glucose gluconic acid

The Fehling's reaction is used to discover glucose in urine.

Accomplishment: pour 10–12 drops of glucose (54) solution in the test-tube and add 3 drops of the Fehling's reagent (55) and heat up.

Observed changes: _____

Conclusion: _____

_____

3. Comparison of reactions of glucose and formalin with Shiff's reagent.

This qualitative test is negative for monosaccharides because of cyclic hemiacetal structure that hasn't aldehyde group.

Accomplishment: in one test tube pour 5–7 drops of formalin (32), in another the same solution of glucose (54) and add in everyone on 2 drops Shiff's reagent (*). In a test tube with formalin — red violet color with glucose this reaction is negative.

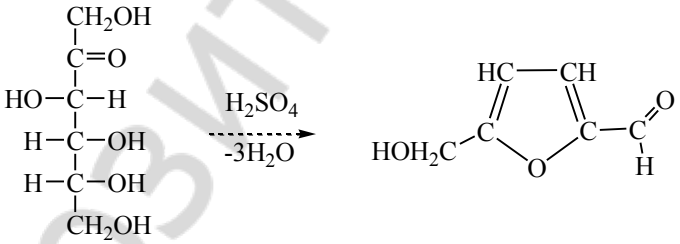
Observed changes: _____

Conclusion: _____

_____

4. The qualitative test on ketohexoses (the Selivanov's test).

The test is predicated on the oxymethylfurfural formation which is condensed with resorcinol forming complex compound of characteristic color.



D-fructose 5-hydroxymethylfurfural

Accomplishment: to 10 drops of fructose (56) solution add 2 drops of HCl* concentrated solution and 1 spatula of resorcinol* crystals. Heat up.

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 12
OLIGO- AND POLYSACCHARIDES

Objective: to develop knowledge of a structure, consider important chemical properties of homo- and heteropolysaccharides in view of their biological properties.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 199–208.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 207–216.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 85–91.

Problems for discussion:

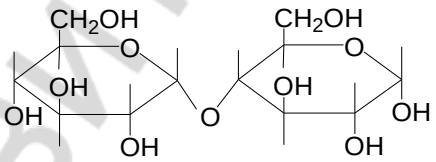
1. Classification of polysaccharides.
2. Disaccharides: maltose, cellobiose, lactose, lactulose, sucrose. Their structures and properties.
3. Starch: structure, biological role. Glycogen.
4. Cellulose: structure, biological role.
5. Dextrane as a source to obtain plasma substitutes.
6. Heteropolysaccharides.

Exercises

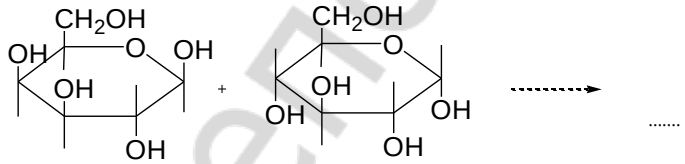
1. Classify the polysaccharides (reducing disaccharide, nonreducing disaccharide, homopolysaccharide, heteropolysaccharide)

sucrose	cellulose	starch	cellobiose	Lactose	maltose
dextrane	heparine	lactulose	chondroitin sulfate	hyaluronic acid	

2. Indicate the monosaccharide residuals of disaccharide. Call this disaccharide and type of glycoside bond.

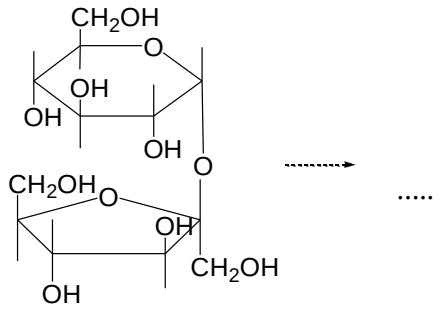


3. Write the reaction of lactose formation.



4. Write the formula of lactulose.

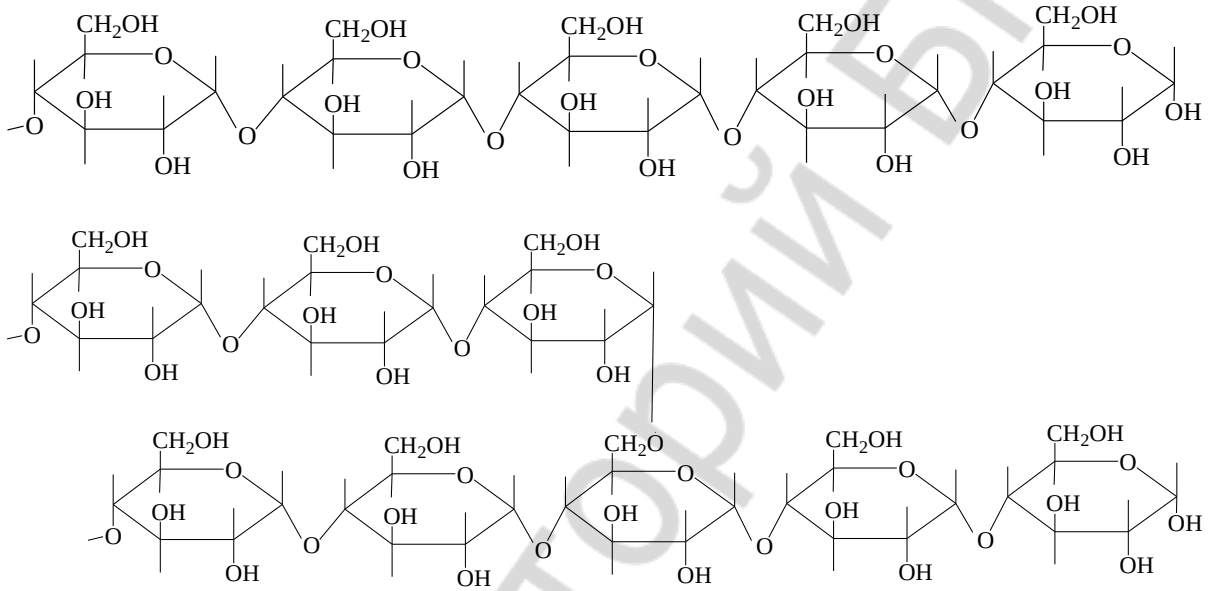
5. Complete the reaction of sucrose hydrolysis:



.....

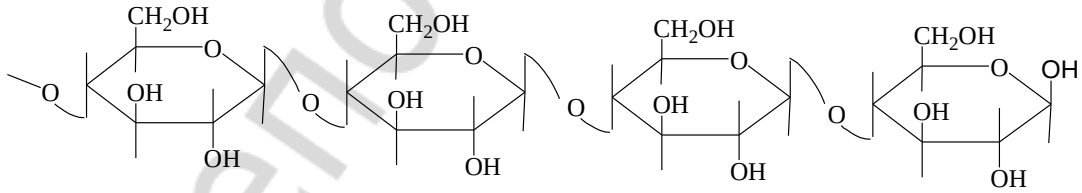
6. Starch consists of the following fractions:

At the amylose and amylopectine fragments indicate monomer, bond types between monosaccharide residuals.

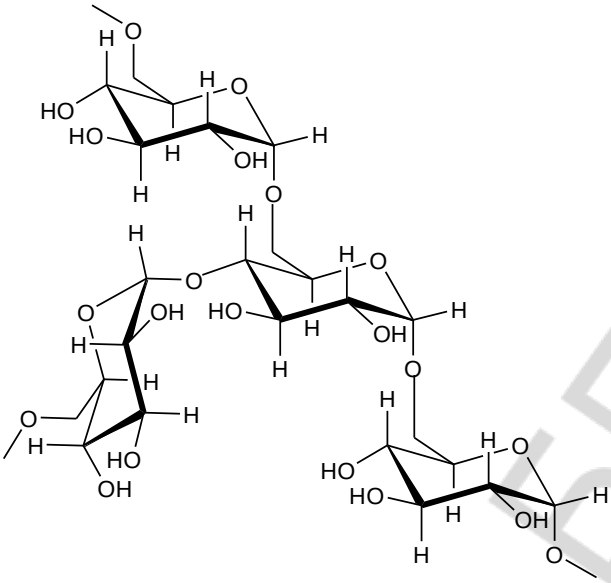


The end hydrolysis product of starch is …

7. Call mentioned below fragment of polysaccharide. Indicate monomer and bond types between monosaccharide residuals.

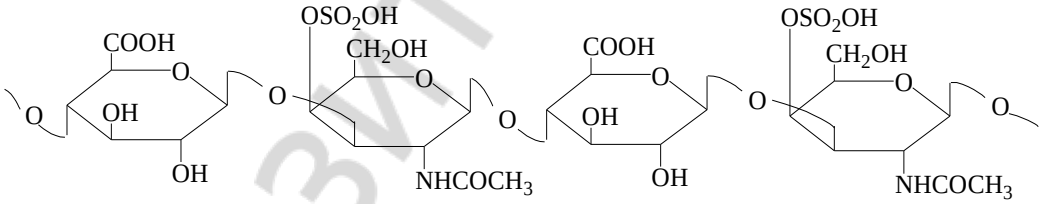


8. Analyze the fragment of homopolysaccharide. Indicate monomer of this polymer and type of bonds.



9. Write the fragment of hyaluronic acid (min. 4 monosaccharide residuals) consisting of disaccharide fragment which linked by β(1-4) glycoside linkage, D-glucuronic acid and N-acetyl-D-glucosamine bonded β (1-3) glycoside linkage.

10. Call the residuals of monosaccharide at the chondroitin sulfate structure.



TEST CONTROL

1. Point out functional groups participated in bond formation between monosaccharide residues in nonreducing disaccharide:

1) two alcoholic OH-groups;
2) hemiacetal and alcoholic OH-groups;
3) two hemiacetal OH-groups;
4) aldehyde and alcoholic OH-group.

2. Which disaccharides could undergo mutarotation?

1) lactulose; 2) cellobiose; 3) sucrose; 4) lactose.

3. As a result of sucrose hydrolyses forms:

1) glucose and mannose;
2) galactose and glucose;
3) galactose and fructose;
4) glucose and fructose.

4. Point out characteristics and properties of dextran:

1) main type of glycoside bond between monosaccharide residue is α(1→6);
2) hydrolysis yield glucose;

3) bacterial metabolic product;
4) has plant origin.

5. Choose disaccharide(s) acid-catalyzed hydrolysis of which yields only glucose:
1) lactose; 2) lactulose; 3) maltose; 4) cellobiose; 5) sucrose.

6. Select sugar which refer to homopolysuccharides:
1) heparin; 2) starch; 3) dextran; 4) cellulose; 5) hyaluronic acid.

7. Invert sugar is hydrolysis product of:
1) cellobiose; 2) maltose; 3) lactose; 4) sucrose.

8. Chose the type of glycoside bond in lactose:
1) α (1-4); 2) α,β (1-2); 3) β (1-4); 4) α (1-3).

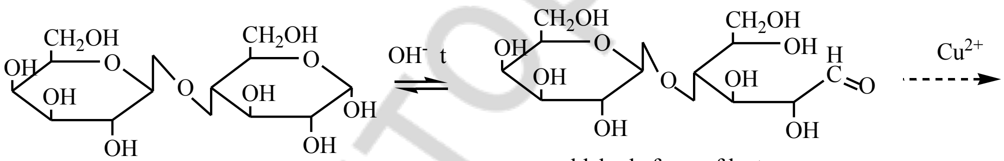
9. Chose the type of glycoside bond in lactulose:
1) α (1-2); 2) α (1-4); 3) β (1-4); 4) α (1-6).

10. Find characteristics and properties of cellulose:
1) monosuccharide residues link by α(1-4) glycoside bond;
2) hydrolysis yield glucose molecules;
3) monosuccharide residues link by β(1-4) glycoside bond;
4) produced by plants.

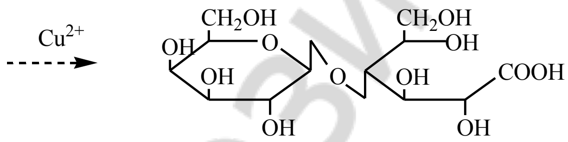
PRACTICAL PART

1. The Fehling's reaction with sucrose and lactose.

Lactose has free hemiacetal hydroxyl group and in alkaline medium when heated it can turn into tautomeric forms containing aldehyde groups that possess reducing properties. Unlike lactose (and maltose) sucrose hasn't free hemiacetal hydroxyl group and belong to unreducing disaccharides.



aldehyde form of lactose



lactonic acid

Accomplishment: take 2 test tubes. In the one test-tube pour 10 drops of sucrose solution (57), in another pour the same quantity of the lactose solution (58), add to each test-tube 3–4 drops of the Fehling's reagent (55) and carefully heat up.

Observed changes: ____

Conclusion: ____

2. The qualitative test on the starch.

Accomplishment: to 10–12 drops of starch solution (*) add 1 drop of the Lugol's solution (47). Fix the color change, heat up the solution and fix the changes.

Observed changes: ____

Conclusion: ____

Signature of teacher:

LABWORK № 13
STRUCTURE AND REACTIVITY OF AMINO ACIDS ACTING AS HETEROFUNCTIONAL COMPOUNDS

Objective: to discuss characteristics of amino acids as heterofunctional compounds acting as structural components of peptides and proteins; to form skills for carrying out qualitative reactions on the amino acids.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 211–217.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 217–224.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 92–98.

Problems for discussion:

1. Biogenic amino acids. Proteinogenic amino acids: classification, structures, stereochemistry.
2. Amphoteric properties of amino acids.
3. Reactions of amino acids on the carboxylic group.
4. Reactions of amino acids on the amino group.
5. Biologically important reactions of amino acids: deamination, transamination, decarboxylation, hydroxylation reactions.

Exercises

1. Write down proteinogenic amino acids at the mentioned below table with three letter code.

Hydrophobic AA (8)	**Hydrophilic AA**		
	With non-ionized radical (7)	*With negative ionized radical (2)*	*With positive ionized radical (3)*

Aliphatic AA (5)	
Hydroxy amino acids (2)	
Dicarbonic (acidic) AA (2)	
Amides of dicarbonic AA (2)	
Diaminomonocarbonic acids (2)	
S-containing AA (2)	
Aromatic AA (2)	
Heterocyclic AA (3)	

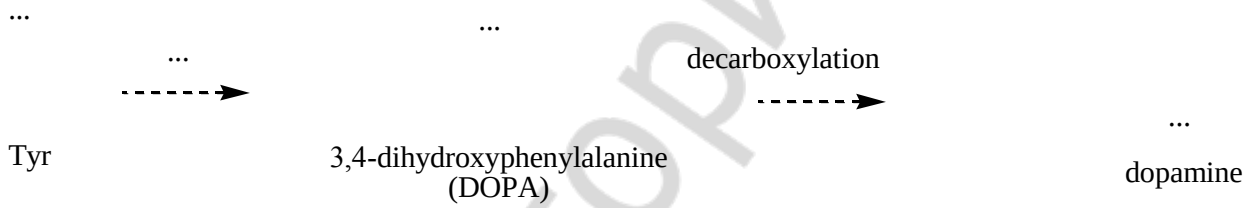
Designate (*) essential AA at the table.

2. Write down the formulas of aliphatic amino acids, designate chiral centers.

3. Write Fischer projection of L-valine.

4. Write down the formulas of aromatic amino acids.

Complete the scheme.



5. Write the structures of hydroxyl containing amino acids.

Write down the serine decarboxylation reaction. Call vitamin, which participates at that reaction as coenzyme.

6. Write the structures of S-containing amino acids.

Write the oxidation reaction of cysteine *in vivo*.

7. Write down the hydrophilic amino acids with negative ionized radical.

Write down the reaction of decarboxylation of Glu. Indicate the biological role of reaction product.

8. Write down the hydrophilic AA with positive ionized radical.

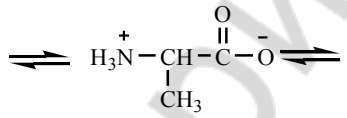
Write down the reaction of decarboxylation of His. Indicate the biological role of reaction product.

9. Write down the tryptophan formula. Indicate heterocycle in this AA.

10. Write down the hydrophilic AA containing amide group.

11. Complete the scheme:

pH 1,0 pH 6,0 pH 12,0

....  ...

12. Write down the formula of proline and fill in the scheme. Indicate the coenzyme of this reaction.

hydroxylation

... ...

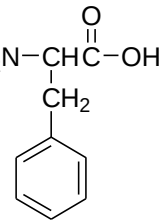
proline 4-hydroxyproline

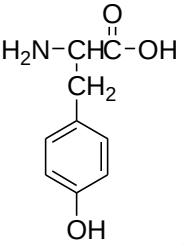
13. Write the reaction of transamination between L-alanine and α oxoglutaric acid. Indicate the coenzyme of this reaction.

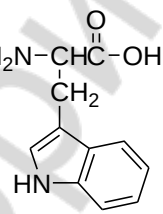
14. Write the scheme of oxidative deamination reaction of Glu *in vivo.*

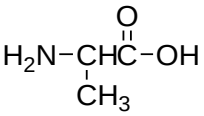
TEST CONTROL

1. Choose structural formulas of essential amino acids:

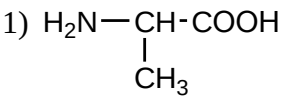
1) 

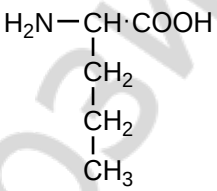
2) 

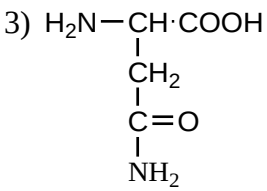
3) 

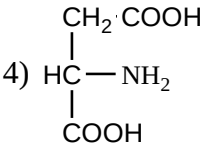
4) 

2. Choose structural formulas of proteinogenic amino acids:

1) 

2) 

3) 

4) 

3. Choose aromatic cycle containing amino acids:
1) Tyr; 2) Pro; 3) Thr; 4) His; 5) Trp.

4. Point out amino acids with ionogenic radical:
1) Asn; 2) Asp; 3) Arg; 4) Glu; 5) His.

5. Choose amino acids which exist in the form of four stereoisomers:
1) isoleucine;
2) threonine;
3) 4-hydroxyproline;
4) arginine.

6. Choose amino acids with two carboxylic groups:
1) Gln; 2) Ala; 3) Glu; 4) Asn; 5) Asp.

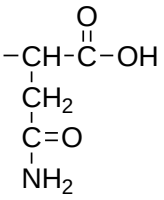
7. Which vitamin participate in reactions of prolin and lysine hydroxylation for connective tissue synthesis:

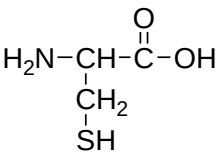
1) B_6; 2) C; 3) PP; 4) D.

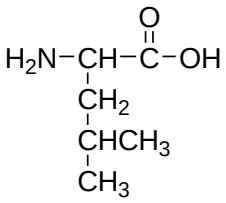
8. As a result of posttranslational modification is formed:

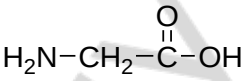
1) cysteine; 3) 5-hydroxylysine;
2) 4-hydroxyproline; 4) threonine.

9. Choose amino acids structures in following sequence: leucine, asparagine, cysteine, glycine:

a) 

b) 

c) 

d) 

1) c, a, b, d; 2) a, c, d, b; 3) a, b, c, d; 4) d, a, b, c.

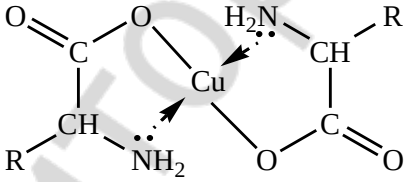
10. Select transamination reaction products of pyruvic acid and Glu:

1) Ala and 2-oxobutanedioic acid;
2) Gly and 2-oxopentanedioic;
3) Ala and 2-oxopentanedioic;
4) Asp and 2-oxopentanedioic.

PRACTICAL PART

1. Reactions of amino acids with copper salts.

Amino acids as the amphoteric compounds form water soluble chelated compounds with copper ions.

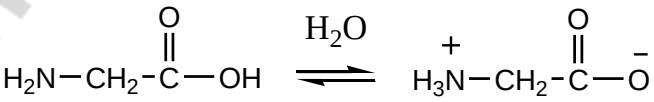


Accomplishment: add 1 copper (II) sulfate crystal (3) and 1 sodium acetate crystal (42) to 10 drops of 1 % glycine (6) solution. Shake the test-tube.

Observed changes: _____

Conclusion: _____

2. Glycine has neutral medium.



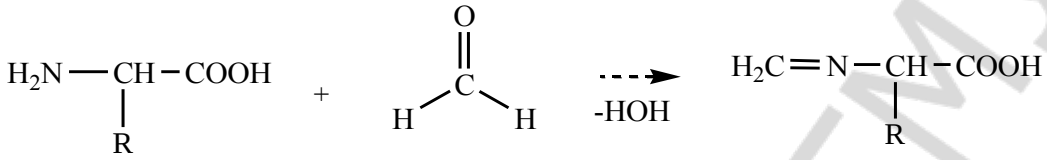
Accomplishment: add 1 drop of 0,2 % methyl red indicator* solution to 5 drops of 1 % glycine (6) solution.

Observed changes: _____

Conclusion: _____

3. Reactions of amino acid with formaldehyde.

Formaldehyde is able to react in the A_N reaction (nucleophilic addition with the following water elimination) with amines and amino acids. At the same time the amino group of amino acid transforms into the methylenamino group (substituted imine). A free carboxylic group of the methylenamino acid causes pH medium change (the medium becomes acidic). It may be proved by the indicator color change.



This reaction is the basis of amino acid quantitative detection in the biological substrates (formalin titration with alkali according to the Serensen method).

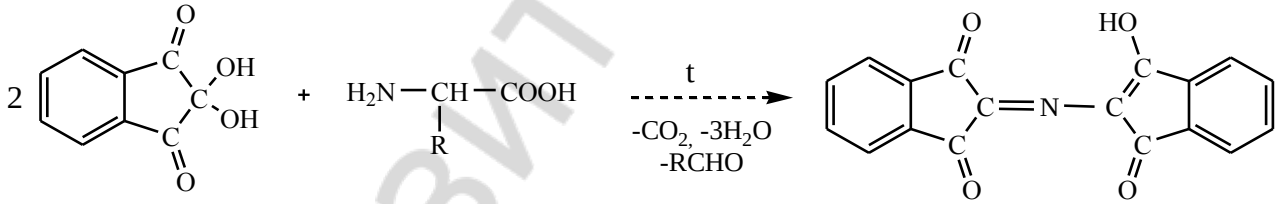
Accomplishment: add 1 drop of 0,2 % methyl red indicator* solution to 5 drops of 40 % formaldehyde solution (32). Carefully add (with glass stick) 1 % NaOH solution* to neutral medium of solution (fix color change). Then add 3 drops of 1 % glycine (6) solution (fix color change again).

Observed changes: _____

Conclusion: _____

4. Ninhydrin reaction.

This reaction is characterized for amino groups of free amino acids and α-amino groups of amino acids that are the part of peptide and protein structure. A ninhydrin reaction is used for α-amino acid detection in the biological liquids under consideration.



ninhydrine reaction product

Accomplishment: add 2 drops of 0,1 % ninhydrin* solution to 5 drops of 1 % glycine (6) solution. Heat the mixture to boil.

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 14
PEPTIDES. THE LEVELS OF PROTEIN ORGANIZATION

Objective: to form knowledge about organization levels of protein molecules, stereochemical features of peptide bond and types of interactions in protein molecule formation.

Recommended literature:

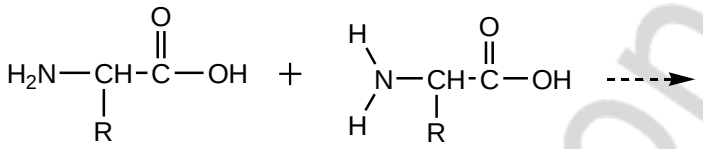
1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 211–224.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 224–228.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 99–103.

Problems for discussion:

1. Peptides: structure and functions. Glutathione, aspartam, insulin.
2. Peptide bond.
3. Proteins. Primary structure of peptides and proteins.
4. Artificial peptide synthesis.
5. Secondary structure of proteins.
6. Tertiary and quaternary structures of proteins. Hemoglobin.
7. Denaturation of proteins.

Exercises

1. Write down the reaction of dipeptide formation.



2. Describe the formation reaction of the following peptides and indicate their charge:

a) Ala-Thr

b) Glu-His

c) Asp-Pro-Met

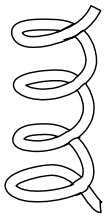
d) aspartyl asparaginyl leucine

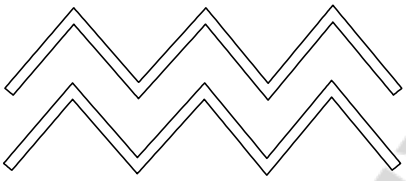
3. Write down the formula of glutathione.

4. Call the type of the secondary protein structure:

They are stabilized with …

Complete the pictures with bonds stabilizing secondary protein structure.





5. Tertiary structure is stabilized with …

Indicate the type of interaction between AA at the polypeptide chain.

Phe and Ala	Arg and Glu	Ile and Val	Cys and Cys
Ser and Gln	Tyr and Thr	Asp and Lys	His and Ser
Trp and Leu	Glu and His	Asn and Ser	Met and Ala

6. ***Denaturation*** — is …

TEST CONTROL

1. Indicate amino acids which participate in ion bonds formation in tertiary structure of protein:

1) Asn; 2) Arg; 3) Cys; 4) Asp; 5) Glu.

2. Indicate amino acids which participate in hydrophobic interactions in tertiary structure of protein:

1) arginine; 2) isoleucine; 3) phenylalanine; 4) thryptophan; 5) asparaginic acid.

3. Choose correct statements:

1) proteins are polymers of proteinogenic amino acids;
2) secondary protein structure is stabilized by ionic bonds;
3) N-end and C-end presents in polypeptide chains;
4) proteins-shaperones participates in tertiary protein structure formation.

4. Indicate amino acids which participate in hydrogen bonds formation in tertiary structure of protein:

1) glutamine; 2) phenylalanine; 3) tyrosine; 4) proline; 5) serine.

5. In physiological conditionals positive charge has:

1) His-Val; 2) Thr-Lys; 3) Arg-Ser; 4) Ile-Tyr; 5) Cys-Arg.

6. Aspartame is dipeptide consisting of asparaginic acid and residue of methyl ether of:
1) glycine; 2) phenylalanine; 3) glutamine; 4) tyrosine.

7. Point out correct statement(s) about peptide bond:
1) carbon, nitrogen and oxygen atoms are in sp^2-hybridisation;
2) lone pair of electrons enter in conjugation with p-electrons of double bond;
3) rotation is capable around peptide bind;
4) carbon, nitrogen and oxygen atoms are in the same plane.

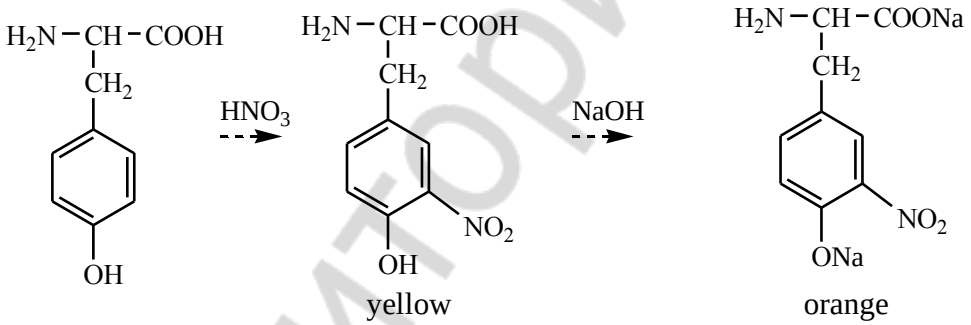
8. Peptide bonds in proteins and peptides are detected by reaction:
1) biuretic; 2) xanthoproteinic; 3) decarboxylation; 4) deamination.

9. In physiological conditionals negative charge has:
1) Asp-Phe; 2) Gln-Trp; 3) Glu-Thr; 4) Ile-Asp; 5) Asn-Pro.

10. C-end amino acid of glutathione is:
1) Glu; 2) Gly; 3) Cys; 4) Gln; 5) Ser.

PRACTICAL PART

1. Xantoproteinic reaction proves the presence of aromatic and heterocyclic α-amino acids such as tryptophane, phenylalanine, tyrosine, histidine in protein structure. When reacted HNO_3 concentrated solution with protein solution nitro-compound is formed. When alkali is added to protein solution the ionization of phenol OH-group occurs.



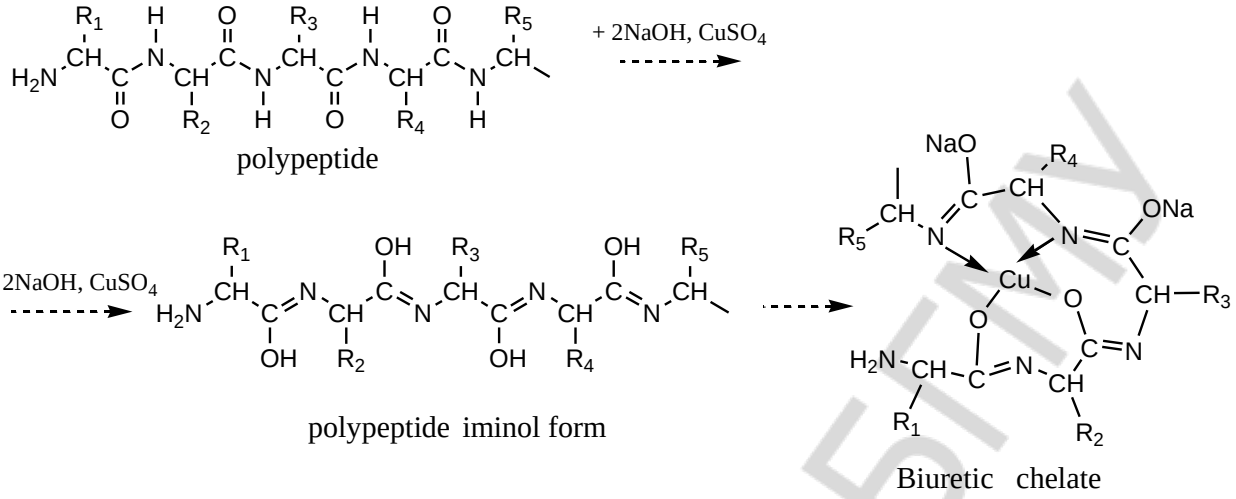
Accomplishment: to 10 drops of protein solution* add drop by drop concentrated solution of HNO_3* to form sediment (of what color?). Then heat carefully this test-tube (fix the change of color). Add some NaOH (21) solution (fix the change of color again).

Observed changes: _____

Conclusion: _____

2. Biuretic reaction determines the peptide bond in the solution of analysed compound. Complex compound of Cu with protein peptide group is formed as a result of biuretic reaction. Commonly peptide bond is presented in amide (or keto-form) in peptides and protein, but in alkaline medium it turns to iminol (enol) form.

Biuretic reaction proceeds in such way:



Accomplishment: to 5 drops of protein solution* add 5 drops of NaOH (21) concentrated solution and then by degrees on the test-tube side pour 2–3 drops of 2 % solution of $CuSO_4$ (26).

Observed changes: _____

Conclusion: _____

_____

3. Precipitation of proteins with sulfosalicylic acid.

It is the example of inreversible protein precipitation. Proteins can not be soluble in the same solvent. Inreversible reactions are protein precipitation reactions with heavy metals, mineral (inorganic) and organic acids, alkaloid reagents and when boiled.

Accomplishment: pour 5 drops of 20 % sulfosalicylic acid* solution to 10 drops of protein solution*. Solution turbidity occurs.

Observed changes: _____

Conclusion: _____

_____

4. Precipitation of proteins with dehydrating agents (alcohol or acetone).

It is the example of reversible protein precipitation. It's called graining that means precipitation process with the concentrated salt solutions (NaCl, $(NH_4)_2SO_4$, $MgSO_4$) or denaturants (alcohol, acetone). Hydration of protein polar group decreases and charge disappearance leads to aggregation and precipitation of proteins. Obtained precipitate can be dissolved with dilution or dialysis that's why it is the reversible precepitation.

Accomplishment: to 10 drops of protein solution* pour 5 drop of acetone*. Solution turbidity occurs.

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 15
NUCLEOSIDES. NUCLEOTIDES. NUCLEIC ACIDS

Objective: to form knowledge about structure and properties of purine and pyrimidine bases, nucleosides and nucleotides, nucleic acids; to develop skills to carring out of qualititative reactions on structural components of nucleotides.

Recommended literature:

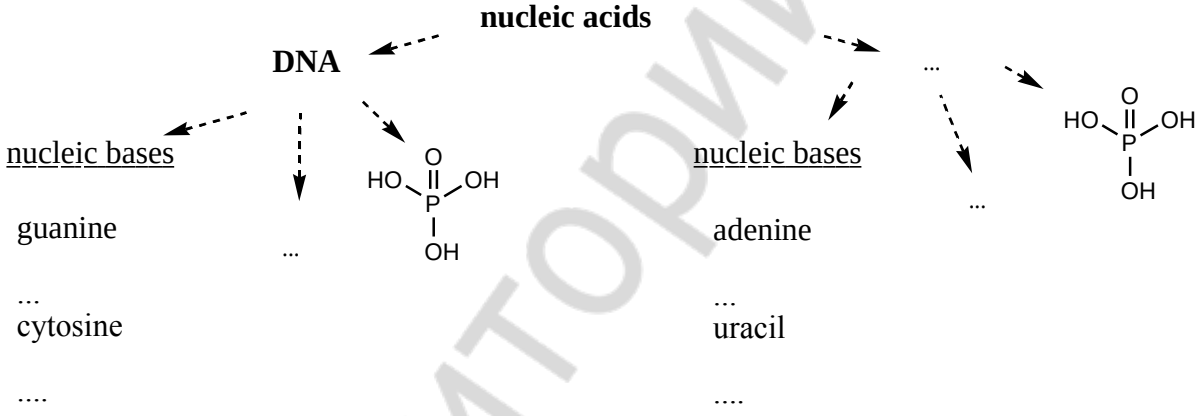
1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 225–237.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 246–256.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 103–108.

Problems for discussion:

1. Structural components of nucleic acids: heterocyclic bases, pentoses.
2. Nucleosides, nucleotides: their structure and properties.
3. Primary structure of DNA and RNA.
4. Secondary structure of DNA.
5. Nucleotide derivatives: cyclic AMP, cyclic GMP, ATP. NAD^+ coenzyme.

Exercises

1. Complete the table.



2. Write down the pyrimidine, number its atoms. Then write uracil, thymine and cytosine at the lactam and lactim tautomeric forms.

pyrimidine	uracil	thymine	cytosine

3. Write down the purine, number its atoms. Then write adenine and guanine at the lactam and lactim tautomeric forms.

purine	adenine	guanine

4. Write the structural formulas showing the hydrogen bonds in complementary base pairs of DNA:

a) thymine – adenine

b) cytosine – guanine

5. Write the formulas of the following nucleosides:

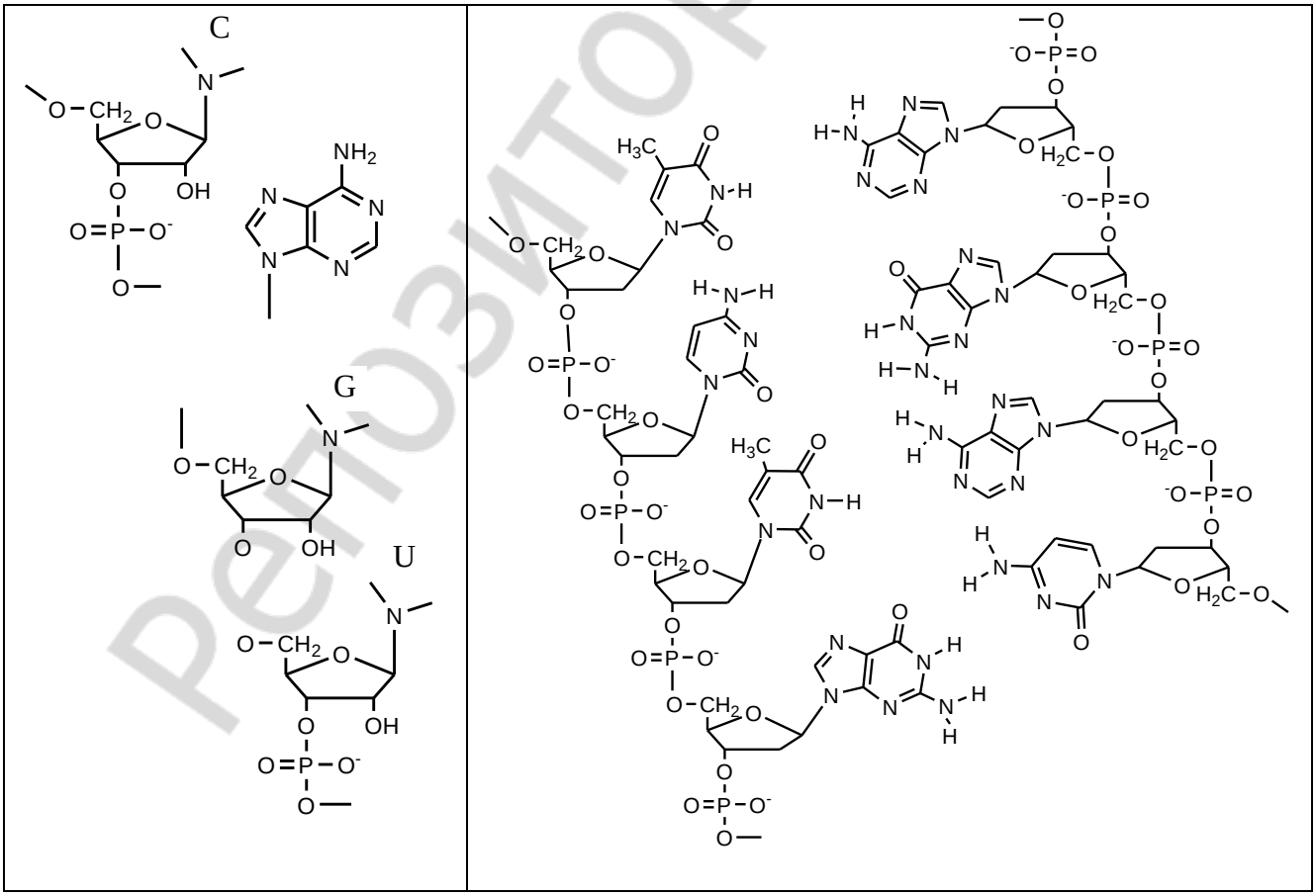
a) guanosine

b) thymidine

6. Write the formulas of the following nucleotides:

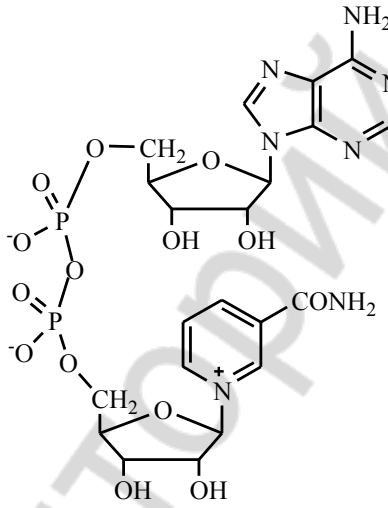
adenosine- 5′- monophosphate	deoxycytidine-5′-monophosphate
UDP	guanosine-3`-monophosphate

7. Restore the structure of 4-nucletide fragment of RNA (on the left) and designate the hydrogen bonds between chains of DNA (on the right).



8. Draw ATP molecule, indicate the bond types.

9. Analyze the formula of NAD^+. Mark the structural components of this molecule. Indicate its biological role.



TEST CONTROL

1. Point out types of tautomerism which characterize cytosine:
1) lactim-lactam; 3) amino-imine;
2) keto-enol; 4) cyclo-oxo.

2. Select products of deoxyadenosine-5'-monophosphate alkaline hydrolyses:
1) deoxyribose; 3) phosphate;
2) adenine; 4) deoxyadenosine

3. Choose nitrogen bases included in RNA:
1) 2-amino-6-hydroxypurine;
2) 2,4-dihydroxy-5-methylpyrimidine;
3) 6-aminopurine;
4) 4-amino-2-hydroxypyrimidine;
5) 2,4-dihydroxypyrimidine.

4. Which type of bond take place between amide of nicotine acid and ribose residue in coenzyme NAD^+:

1) anhydride bond; 3) O-glycoside bond;
2) N-glycoside bond; 4) amide bond.

5. Select products of thymidine-5'-monophosphate acidic hydrolyses (pH 1):

1) thymine; 4) thymidine;
2) ribose; 5) phosphoric acid.
3) deoxyribose;

6. How many ester bonds in adenosine-3',5'-cyclophosphate:

1) 1; 2) 2; 3) 3; 4) 4.

7. Point out type of tautomerism which characterize uridine:

1) keto-enol; 3) amino-imine;
2) cyclo-oxo; 4) lactim-lactam.

8. Which type of bonds presents in nucleotide structure:

1) ester and anhydride;
2) ester and N-glycoside;
3) anhydride and ether;
4) phosphodiester and N-glycoside.

9. How many high-energy bonds in adenosine-5'-triphosphate:

1) 3; 2) 2; 3) 1; 4) 4.

10. Which type of bonds presents in GTP molecule between second and third phosphoric acid residues:

1) anhydride; 3) thioester;
2) ester; 4) hydrogen.

PRACTICAL PART

1. Phosphoric acid detection in products of nucleoprotein hydrolysis (hydrolyzates).

$$H_3PO_4 + 12\ (NH_4)_2MoO_4 + 21\ HNO_3 \rightarrow (NH_4)_3PO_4 \cdot 12MoO_3 + 21NH_4NO_3 + 12\ H_2O$$

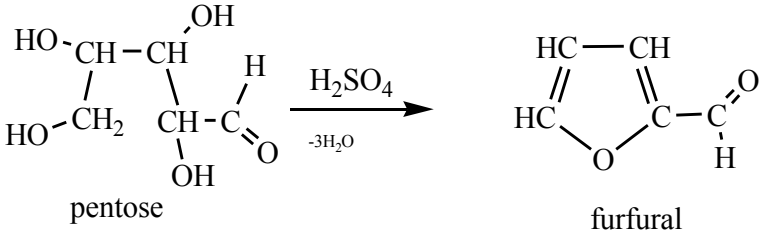
Accomplishment: add 5 drops of molybdenic reagent* to 3–5 drops of yeast hydrolyzate* and boil some minutes.

Observed changes: _____

Conclusion: _____

2. Pentose detection in products of nucleoprotein hydrolysis (the Bial's test).

When reacted with H_2SO_4 concentrated solution or dilute HCl pentoses are dehydrated to form furfural which is condensed with orcinol.



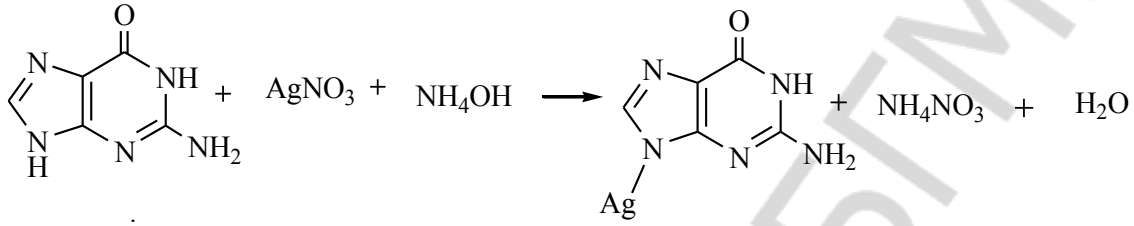
pentose furfural

Accomplishment: add 10 drops of the Bial's reagent* (orcinol solution in HCl with $FeCl_3$) to 10 drops of yeast hydrolyzate* and boil 1–2 minutes.

Observed changes: _____

Conclusion: _____

3. Purine base detection in products of nucleoprotein hydrolysis.



guanine

Accomplishment: add 1 drop of concentrated solution of ammonia and 5 drops of 1 % solution of $AgNO_3$* to 5 drops of yeast hydrolyzate*. Leave the test-tube for 3–5 minutes without mixing.

Observed changes: _____

Conclusion: _____

Signature of teacher:

LABWORK № 16
LIPIDS. LIPID PEROXIDATION

Objective: to develop knowledge about the saponifiable lipids.

Recommended literature:

1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 238–247.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 173–182.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 108–116.

Problems for discussion:

1. Classification of lipids, their biological role.
2. Fatty acids, their structure, properties and nomenclature. Alcohols which form fats and lipids.
3. Waxes, their composition and role.
4. Triacylglycerols, their structure, nomenclature, properties.
5. Phospholipids, their structure, nomenclature, physicochemical properties.
6. Sphingolipids, biological role.
7. The lipid peroxidation of cell membranes. Antioxidants.

Exercises

1. Write the molecular and stick formulas of fatty acids. Give their names according to ω-nomenclature.

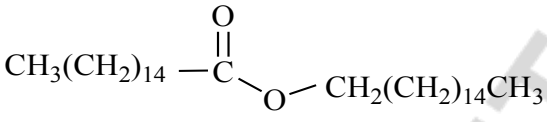
Stearic acid
Palmitic acid
Oleic acid
Linoleic acid
Linolenic acid

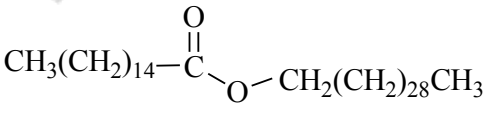
Arachidonic acid

2. Write the formulas of the following alcohols.

glycerol	ethanolamine
2-aminooctadecen-4-diol-1,3 (sphingosine)	
serine	choline

3. Analyze the mentioned below formulas of waxes.

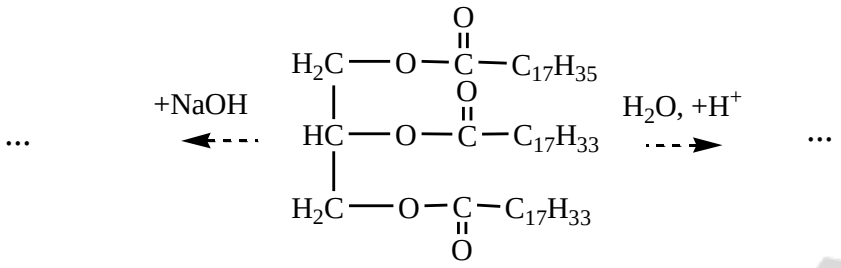




4. Write a structural formulas of the following triglycerides:

1-linoleoyl 2-palmitoyl 3-stearoylglycerol	1,3-dioleoyl-2-linoleoylglycerol

5. Write the hydrolysis reactions of fat. What is the soaps?



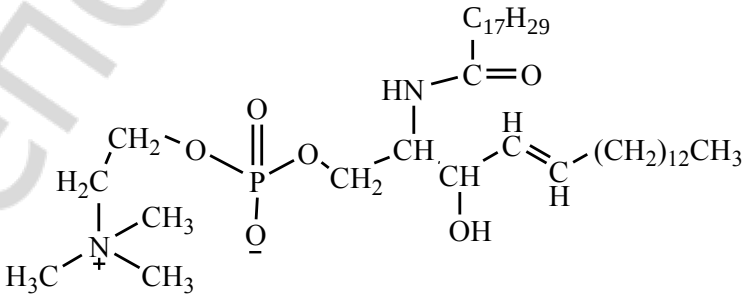
6. Draw the structural formulas of the following compounds. Mark the hydrophobic tails and hydrophilic head.

a) 1-stearoyl-2-oleoylphosphatidylserine

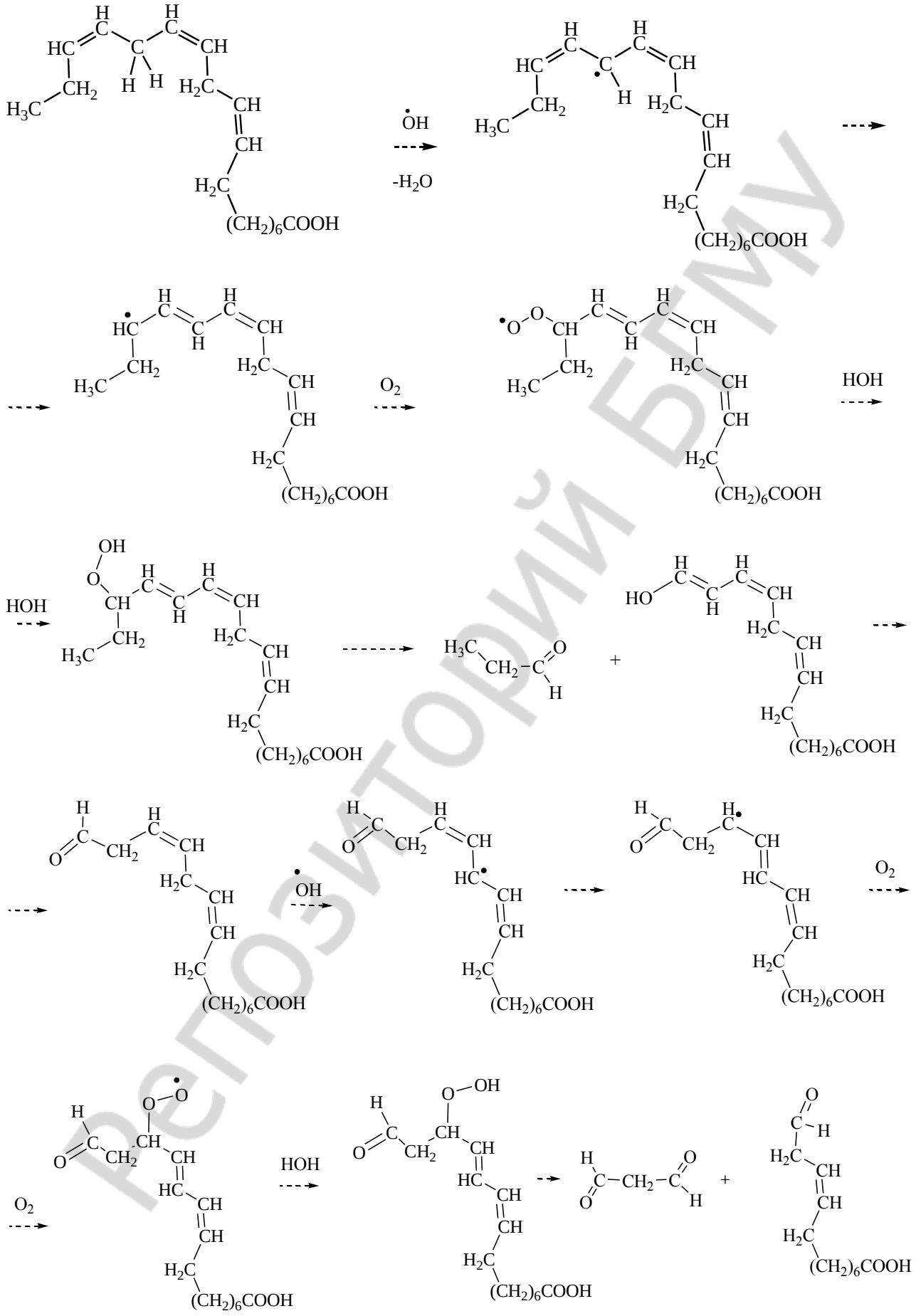
b) 1-stearoyl-2-linoleoylphosphatidylcholine

c) 1-palmitoyl-2-arachidonoylphosphatidylethanolamine

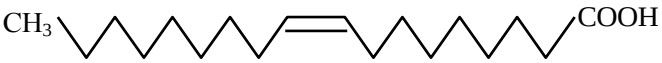
7. Designate well-know fragments at the formula of sphingomyeline.



8. Analyse the mentioned below scheme peroxidation of linolenic acid.



TEST CONTROL

1. Indicate name of the following structure: 

1) linoleic acid; 3) oleic acid;
2) arachidonic acid; 4) stearic acid.

2. Choose simple lipid:
1) myricylpalmitate;
2) trioleoylglycerol;
3) 1-palmytoil-2-oleoylphospatidylcholine;
4) dipalmitoylphosphatidylserine.

3. Point out correct statements about unsaturated fatty acids including in lipids structure:
1) has conjugated doubl bonds;
2) even number of atoms;
3) it is monocarboxylic acids;
4) has branched carbon chain;
5) it is usually cis-isomeres.

4. ω-Nomenclature name of linoleic acid is:
1) 20:4 **ω** 6; 2) 18:3 **ω** 3; 3) 18:1 **ω** 9; 4) 18:2 **ω** 6.

5. Choose complex lipid:
1) myricylpalmitate;
2) 1-srearoyl-2-oleoylphosphatidylinositol;
3) 1-palmitoyl-2-oleoylphosphatidylcholine;
4) tristearoylglycerol.

6. Select alcohols which are a part of lipids composition:
1) propantriol-1,2,3; 3) 2-aminooctadecen-4-diol-1,3;
2) ethanol; 4) inositol.

7. Vitamin E is native antioxidant because of presence in its structure:
1) amino group; 3) phenol hydroxyl;
2) alcoholic hydroxyl; 4) thiol group.

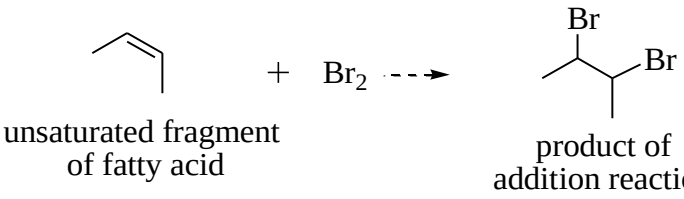
8. ω-Nomenclature name of arachidonic acid is:
1) 20:4 **ω** 6; 2) 20:4 **ω** 3; 3) 18:1 **ω** 6; 4) 18:2 **ω** 6.

9. Choose reserve lipids:
1) 1,2-dioleoyl-3-linolenoylglycerol;
2) 1-oleoyl-2-steariylphosphatidylcholine;
3) 1-oleoyl-2-stearoylphosphatidylinositol;
4) 1,3-dioleoyl-2-stearoylglycerol.

10. Point out type of chemical bond in phosphatidylserine between phospatidic acid and serine?
1) ester bond; 3) O-glycoside bond;
2) anhydride bond; 4) amid bond.

PRACTICAL PART

1. Qualitative reactions on the unsaturated acids which form fats.



unsaturated fragment of fatty acid — product of addition reaction

Accomplishment: to 1 drop of fat* add some drops of bromine water*. Shake the test-tube.

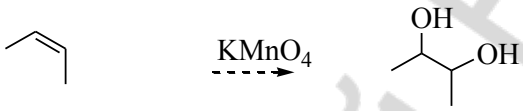
Observed changes: _____

Conclusion: _____

_____

2. Oxidation reaction with potassium permanganate.

Oxidation occurs in the double bond location.



unsaturated fragment of fatty acid — product of oxidation reaction

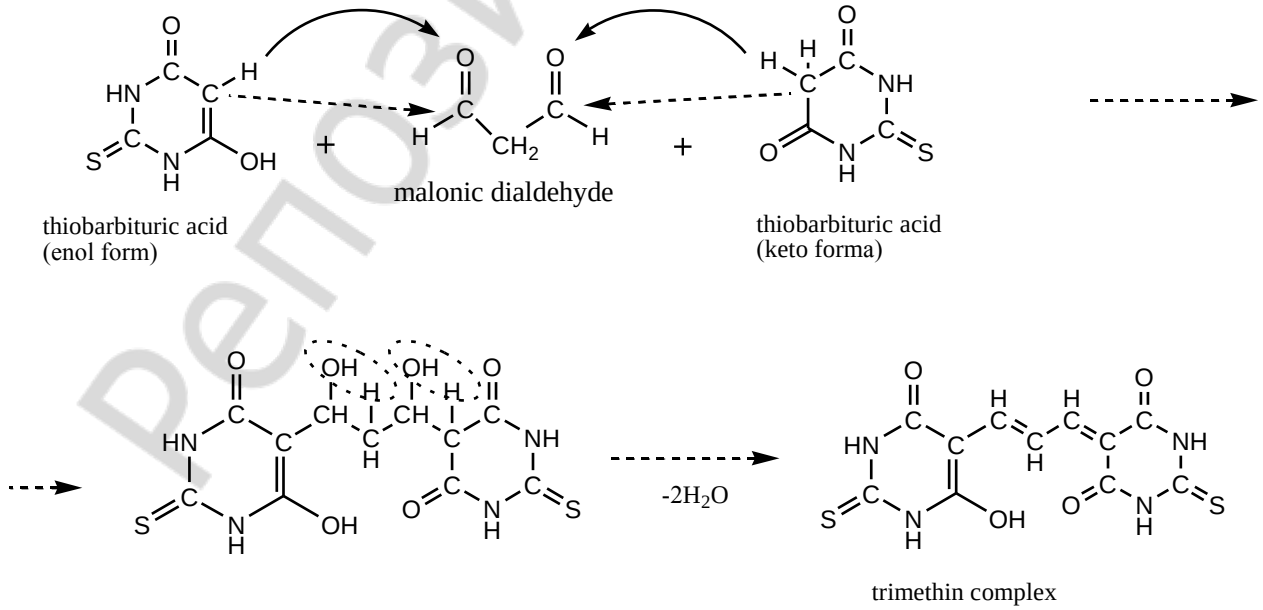
Accomplishment: to 3 drop of fat* pour 3 drops of $KMnO_4$ solution (14) and 2 drops of Na_2CO_3 (43). Shake the test-tube.

Observed changes: _____

Conclusion: _____

3. Malonic dialdehyde detection in the vegetable oil peroxidation products.

The model of lipid peroxidation is rancidification. One of the lipid peroxidation products is malonic dialdehyde which can be formed from ω-3 unsaturated fatty acid hydroperoxides. To detect the malonic dialdehyde the reaction with thiobarbituric acid is used which goes according to the nucleofilic addition mechanism.



thiobarbituric acid (enol form) — thiobarbituric acid (keto forma) — trimethin complex

Accomplishment: in a test-tube № 1 pour 10 drops of a fresh sunflower-seed oil* solution, in a test-tube № 2 pour 10 drops long time stored on the light (in conditions of oxygen access) sun-

flower-seed oil, in a test-tube № 3 pour 10 drops of margarine* solution (oils and margarine are dissolved in heptane-chloroform mixture in the volume ratio 1:1). Then in each of the test-tubes add on 10 drops of the TBA-reagent (0,8 % solution of thiobarbituric acid in an ice acetic acid)*. Test-tubes with a reaction mixture shake up, close with foil, place into boiling water bath. In 15 minutes take out the test-tubes and visually estimate color intensity of solutions.

Observed changes: _____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 17
STEROIDS. ALKALOIDS

Objective: to develop knowledge of a stereochemical structure, consider important properties of steroids and alkaloids and develop skills to carry out qualitative reactions on steroids.

Recommended literature:

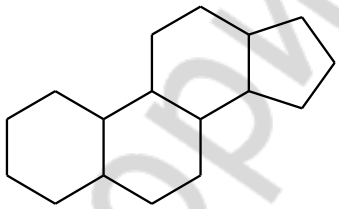
1. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2006. P. 251–258.
2. *Zurabyan, S. E.* Fundamentals of Bioorganic Chemistry / S. E. Zurabyan. 2012. P. 185–191.
3. *Ryneiskaya, O. N.* Bioorganic chemistry. Course of lecture / O. N. Ryneiskaya, I. V. Ramanouski. 2015. P. 116–122.

Problems for discussion:

1. Steroids: their structure, nomenclature and classification.
2. Stereochemistry of steroids. 5α and 5β series of steroids.
3. Sex hormones: estrane and androstane derivatives.
4. Pregnane derivatives: corticosteroids and progestins.
5. Cholic acid. Bile acids.
6. Cholesterol. Biological importance.
7. Vitamins D_2 and D_3.
8. Alkaloids.

Exercises

1. Number carbon atom of gonane.



2. Draw the conformations of 5α and 5β steroids.

3. Draw the basic classes of steroids:

estrane	androstane	pregnane

cholane	cholestane

4. Write the structural formulas of estrogens:

estr-1,3,5(10)- triene-3,17-diol (estradiol)	3-hydroxyestr-1,3,5(10)- trien-17-one (estrone)

5. Write the structural formulas of androgens:

17-hydroxyandrost-4-ene-3-one (testosteron)	3-hydroxy-5α-androstan-17-one (androsterone)

6. Write the structural formulas of pregnane derivatives:

17, 21-dihydroxypregn-4-ene-3,11,20-trione (cortisone)	17, 11, 21-trihydroxypregn-4-ene-3,20-dione (cortisol)

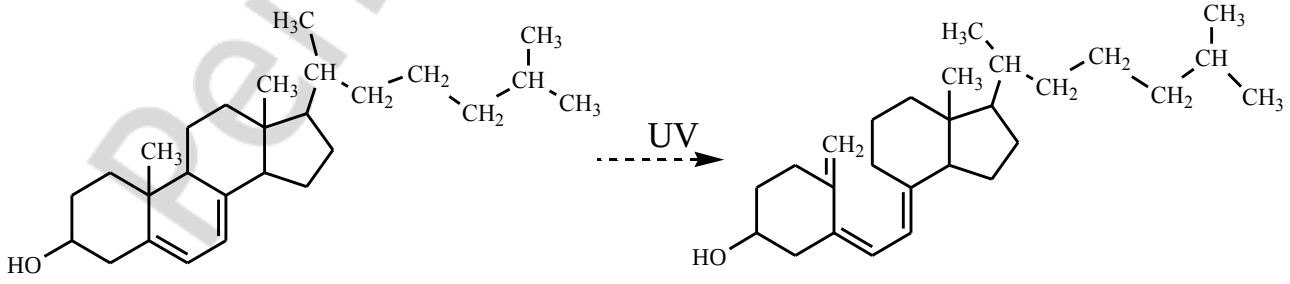
pregn-4-ene-3,20-dione (progesterone)

7. Write the structural formulas of bile acids:

3, 7, 12-trihydroxy-5β-cholan-24-oic acid	3, 12-dihydroxy-5β-cholan-24-oic acid

8. Write the formula of cholesterol (cholest-5-en-3β-ol).

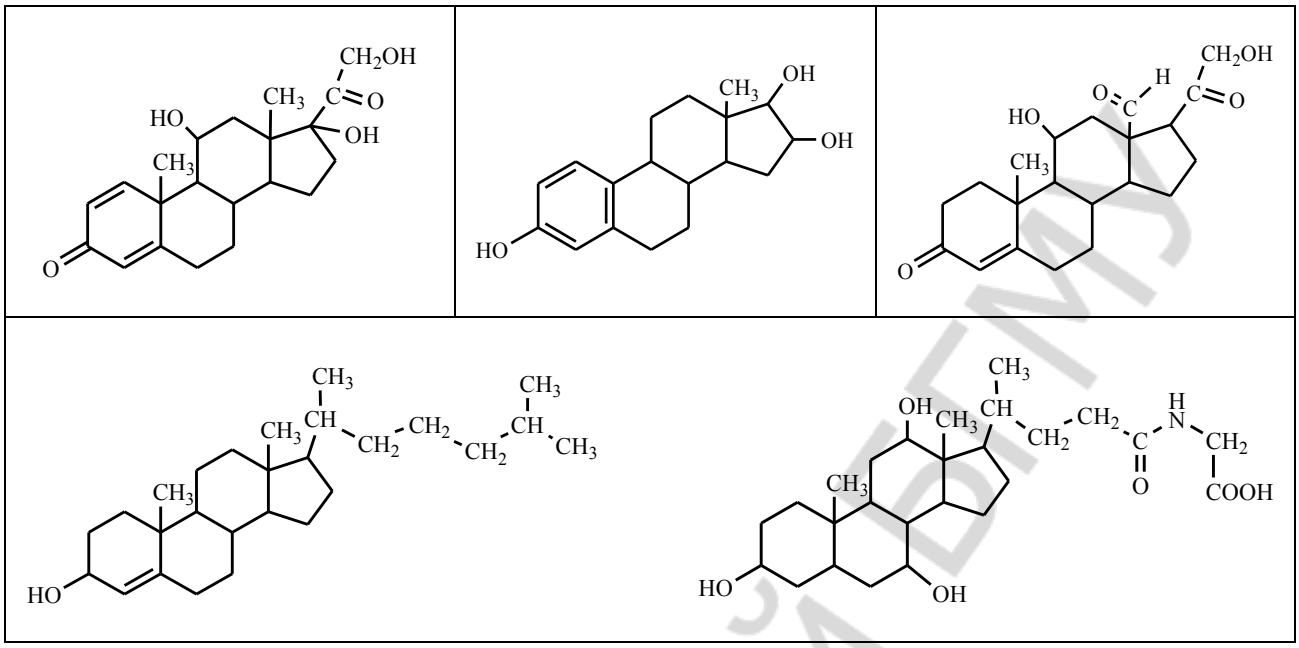
9. Vitamine D_3 is formed from7-dehydrocholesterol in skin with ultraviolet. Explain the peculiarity of this reaction.



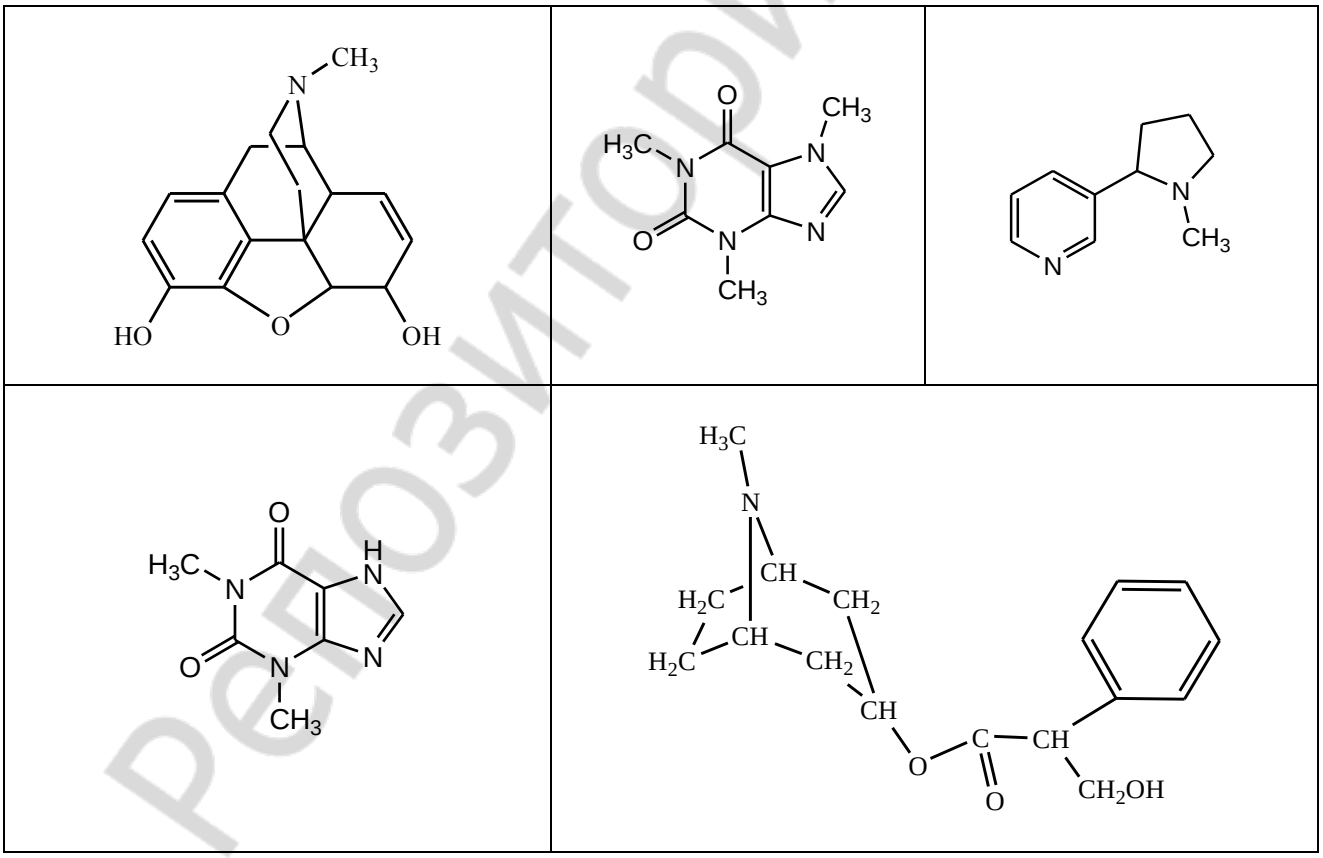
7-dehydrocholesterol

cholecalciferol

10. Define the group of the following steroids (according to structural classification — estrange, androstane, pregnane, cholane, cholestane).



11. Define the trivial names of the following alkaloids:



TEST CONTROL

1. Structural base of steroids is:
1) phenanthrene;
2) gonane;
3) perhydrophenanthrene cyclopentane;
4) pyrrole.

2. Structure of steroids is characterized:
1) plane structure;
2) non-plane structure;
3) gonane has chiral centers;
4) gonane has no chiral centers.

3. The parent structures of the sex hormones are:
1) androstane; 2) pregnane; 3) cholane; 4) estrange.

4. Bile acids contain:
1) androstane; 2) pregnane; 3) cholestane; 4) cholane.

5. Select the correct statements about estradiol:
1) it contains oxo group at 3 carbon atom;
2) it contains hydroxyl group at 3 carbon atom;
3) it has basic properties;
4) it has acidic properties.

6. Select the correct statements about cholesterol:
1) it has oxo-group at 3 carbon atom;
2) it is base to form sex hormone;
3) it is base to form bile acids;
4) it is a component of biological membranes.

7. Select the correct statements about bile acids:
1) they are cholestane derivatives;
2) they are formed by liver;
3) they are cholane derivatives;
4) it is a components of biological membranes.

8. Select the correct statements about steroids:
1) they have hydrophilic properties;
2) they have hydrophobic properties;
3) purine is the base of steroids;
4) gonane is the base of steroids.

9. Select the correct statements about alkaloids:
1) they are formed by plants;
2) alkaloids have basic properties;
3) alkaloids have acidic properties;
4) they are formed by animals.

10. Caffeine contains the following cycle:
1) isoquinoline; 2) xanthine; 3) phenanthrene; 4) indole.

PRACTICAL PART

1. Color reaction on the cholesterol.

Accomplishment: in the dry test-tube pour 1 drop of $FeCl_3$ solution in acetic acid* and 5–8 drops of concentrated solution of H_2SO_4*. Carefully shake the test-tube and add 5 drops of cholesterol solution in acetic acid*.

Observed changes: _____

_____

Conclusion: _____

_____

2. Reactions to discover alkaloids.

Accomplishment: on an object-plate add 3 drops of investigated solution* at distance of 2 cm from each other. To the first drop add 1 drop of the Lugol's solution (47), to the second add 1 drop of 1 % picric acid solution*, to the third add 1 drop of a phosphomolybdic acid.

Observed changes: _____

_____

Conclusion: _____

_____

Signature of teacher:

LABWORK № 18
CONCLUDING TEST "BIOPOLYMERS AND THEIR STRUCTURAL COMPONENTS"

Remind the program material from the theme № 9 to № 17.

Recommended literature:
Study the literature from the theme № 9 to № 17.

Questions to the test control:

1. Oxidation reactions of hydroxy acids *in vivo*.
2. Reduction reactions of oxo acids *in vivo*.
3. Decarboxylation reactions of hydroxyl and oxo acids *in vivo*. Biogenic amines.
4. Decomposition reaction of citric acid at heating.
5. Formation reaction of citric acid from oxaloacetic acid and acetyl coenzyme A.
6. Dehydration reaction of citric acid *in vivo*.
7. π-Diastereomers of butenedioic acid. Hydration reaction of fumaric acid.
8. Specific reactions of α-, β-, γ-hydroxy and amino acids.
9. Tautomerism. Keto-enol and lactim-lactam tautomerism.
10. Ketone bodies, their biological role.
11. Formation of acetylsalicylic acid.
12. p-Aminobenzoic acid, their derivatives. Modern anesthetics.
13. Fatty acids. Conformational structure. ω-Nomenclature of unsaturated fatty acids.
14. Triacylglycerols: structures, nomenclature, biological role. Hydrolysis of triacylglycerols.
15. Phospholipids as amphiphilic molecules. Structures, nomenclature, biological role. Hydrolysis of phospholipids.
16. Cyclo-oxo tautomerism of monosaccharides. The Fischer projection formulas and Haworth formulas of glucose and galactose. Conformations of monosaccharide cyclic forms. Glycosides.
17. Oxidation of monosaccharides. Biological role of glycuronic acids.
18. Ascorbic acid as water soluble antioxidant.
19. Reducing and nonreducing disaccharides. Structure, biological role.
20. Polysaccharides: structure, biological role.
21. Proteinogenic amino acids. Structure, nomenclature, acid-basic properties, bipolar structure. Stereoisomerism.
22. Biologically important reactions of α-amino acids: deamination, hydroxylation, decarboxylation, transamination reactions.
23. Peptides: structures, nomenclature, biological role. Representatives of peptides and their biological significance (glutathione, neuropeptides, insulin).
24. Nucleic bases: structures, tautomeric forms, biological role.
25. Complementary pairs of nucleic bases. Hydrogen bonds.
26. Nucleosides, nucleotides. Structure, biological role. Hydrolysis.
27. ATP, cyclo-AMP.
28. Steroids: classification, stereoisomerism. 5α- and β-steroids. Cholesterol: structure and biological role.
29. Estrogens and androgens: structure and biological role. Corticosteroids.
30. Bile acids: structure and biological role.

It is necessary to know formulas of the following compounds:

1. glycerol	14. uric acid	28. ethanolamine	42. dextran
2. pyruvic acid	15. hypoxanthine	29. D-glucose	43. uracil
3. oxaloacetic acid	16. xanthine	30. D-ribose	44. thymine
4. lactic acid	17. acetylsalicylic acid	31. D-deoxyribose	45. cytosine
5. malic acid	18. novocaine	32. D-fructose	46. adenine
6. acetyl coenzyme A	19. ultracaine	33.D-galatose	47. guanine
7. citric acid	20. lidocaine	34. ascorbic acid	48.fatty acids
8. fumaric acid	21.palmitic acid	35. sucrose	49. proteinogenic amino acids (20), their names and three letter codes
9. maleic acid	22. oleic acid	36. maltose	
10. β-hydroxybutyric acid	23. linoleic acid	37. lactose	
	24. linolenic acid	38. lactulose	
11. β-oxobutyric acid	25. arachidonic acid	39. starch	50.parent structures of steroids
12. barbituric acid	26. choline	40. glycogen	
13.phenobarbital	27. inositol	41. cellulose	

QUESTIONS TO THE BIOORGANIC CHEMISTRY EXAM

1. Conformations. Newman projections. Types of strains. Energetic characteristic of eclipsed, gauche and staggered conformations (butane). Conformational structure of hydrocarbon radicals of fatty acids (palmitic and stearic acids).

2. Cyclohexane conformations. Types of strains (angle, torsion, Van-der-Waals). Inversion of cycle. 1,3-diaxial interaction.

3. Configuration of organic compounds. Stereoisomerism. Fischer projections. Relative configuration and D, L-convention. Glyceraldehyde as the configurational standart.

4. Stereoisomerism of molecules with one chiral centre (lactic acid as an example). Enantiomers. Optic activity. Racemic mixtures.

5. Absolute configuration of stereoisomers. R, S-convention. Relationship of spatial structure with biological activity.

6. Electronic effects in organic molecules (inductive and mesomeric), their role in the reactivity centers in the molecule. Electron donors and withdrawers.

7. Conjugation (π,π- and p,π-conjugations). Conjugated systems with open chain (butadiene-1,3).

8. Conjugated systems with close chain. Aromaticity, criteries of aromaticity, Huchel's rule (benzene, naphtaline, phenantrene).

9. Heterocyclic aromatic compounds (pyrrol, pyridine). Pyrrol and pyridine nitrogen atoms. Π-excess and π-deficient aromatic systems.

10. Acidity and basicity of organic compounds; Brensted and Lewis theories.

11. Acidic properties of organic compounds (alcohols, phenols, thiols, carboxylic acids, amides). Factors of anion stability.

12. Basic properties of organic compounds (alcohols, ethers, thioesters, amines). Comparing of basic properties of aliphatic and aromatic amines; salt formation.

13. Classification of organic reactions (substitution, addition, elimination, isomerisation, red-ox, acid-basic interaction).

14. Classification of organic reactions on the mechanism of covalent bond cleavage (radical and ionic). Electronic and spatial structure of free radicals, carbocations and carboanions.

15. Oxidation reactions of organic compounds (alcohols, thiols, phenols). Antioxidants (2,3-dimercaptopropanol, ascorbic acid, phenols and others).

16. Radical substitution reactions. Propane chlorination as an example of free radical substitution. Initiators of radical reactions. Antioxidants.

17. Electrophilic addition reactions of alkenes. Hydration reactions of alkenes. Acidic catalys. Markovnikov's rule.

18. Electrophilic substitution reactions of aromatic hydrocarbons. Substituent effects in the aromatic ring on the reactivity of aromatic hydrocarbons. O-, p-, m-orientants.

19. Alkylation reactions of aromatic compounds. Role of catalyst in the electrophile (Lewis acid, acidic catalys in the alkylation with alkenes and alcohols)

20. Electronic and spatial structure of the carbonyl group. Comparative reactivity of aldehydes and ketones.

21. Oxidation and reduction reactions of carbonyl compounds. Visual tests on the aldehyde group (silver mirror test, Trommer test). Reduction reactions *in vivo*, NADH as a hydride ion donor.

22. Nucleophilic addition reactions of aldehydes and ketones; addition of water and alcohols.

23. Addition of amines to carbonyl compounds, mechanism. Schiff's bases.

24. Electronic and spatial structure of the carboxylic group. Acidic properties of the carboxylic acids: mono-, dicarboxylic, aliphatic saturated, aliphatic unsaturated, aromatic carboxylic acids.

25. Nucleophilic substitution at sp^2-hybridized carbon atom in the carboxylic group: esterification reaction. Properties of esters, hydrolysis.

26. Polyfunctional compounds and their characteristics. Polyols: ethylene glycol, glycerol, inositol, xylitol, sorbitol. Visual test on the diol fragment.

27. Dicarboxylic acids and their properties. Decarboxylation reactions and anhydride formation.

28. Diatomic phenols: hydroquinone, resorcinol, catechol. Oxidation of diatomic phenols. Phenols as antioxidants. Adrenaline.

29. Heterofunctional compounds and their characteristics. Intramolecular and intermolecular reactions of nucleophilic substitution in the amino acids and hydroxy acids. Elimination reactions.

30. Citric acid (2-hydroxypropane-1,2,3-tricarboxylic acid). Decomposition reactions. Citrates. «Citrated blood».

31. Oxo acids (pyruvic acid, acetoacetic acid, oxaloacetic acid, α-ketoglutaric acid). Acidic properties and reactivity (A_N and S_N-mechanisms) of oxo acids. Transamination reactions of α-oxo acids.

32. Keto-enol tautomerism. Reactions on the enol fragment.

33. β-Hydroxy butyric acid, β-oxo butyric acid, acetone as representatives of *ketone bodies*, their biological and diagnostic significance (visual tests on the acetone).

34. Anesthesin and novocain as ester of p-aminobenzoic acid. Novocain chloride. Modern anesthetics: lidocaine, ultracaine.

35. Salicylic acid, salicylates, acetylsalicylic acid.

36. Sulfa drugs. Medicine applications.

37. Nicotinic acid and its amide as vitamin PP. NAD^+, NADH.

38. Barbituric acid. Tautomerism of barbituric acid. Phenobarbital.

39. Hydroxypurines: hypoxanthine, xanthine, uric acid. Tautomerism of hydroxypurines.

40. Alkaloids, their biological role and using in medicine. Nicotine. Morphine. N-methylated xanthines (caffeine, theophylline, theobromine).

41. Properties of fatty acids. Saturated and unsaturated fatty acids.

42. Lipids. Properties. Triacylglycerols: structures, biological role.

43. Phospholipids as amphiphilic molecules.

44. Carbohydrates. Classification, biological properties. Monosaccharides. D, L-stereochemical rows.

45. Tautomeric forms of monosaccharides: open chain and cyclic forms. The Fischer projection formulas and Haworth formulas of glucose and galactose. Conformations of cyclic forms of glucose.

46. Ring-chain tautomerism of fructose. Furanoses and pyranoses; α- and β-anomers.

47. Structure and tautomeric forms of important representatives of pentoses (ribose and deoxyribose). Their biological role.

48. Nucleophilic substitution at the anomeric centre in the cyclic forms of monosaccharides. O- and N-glycosides. Hydrolysis of glycosides.

49. Oxidation of monosaccharides. Biological role of glycuronic acids.

50. Ascorbic acid as water soluble antioxidant.

51. Reducing disaccharides (maltose, lactose, cellobiose). Structure, ring-chain tautomerism.

52. Lactose: structure, ring-chain tautomerism. Reducing properties. Hydrolysis. Role of oligosaccharides of lactose group in the nonpathogenic intestinal flora necessary for normal digestion. Lactulose.

53. Sucrose as representative of nonreducing disaccharides (the Haworth formula). Hydrolysis of sucrose. Invert sugar.

54. Starch. Structure (amylose and amylopectin), properties, hydrolysis reactions. Biological role.

55. Cellulose. Structure, properties, application, role in nutrition.

56. Glycogen is reserve homopolysaccharide of animals and human (the Haworth structure). Biological significance of branched structure of glycogen.

57. Dextran as representative of bacterial origin homopolysaccharides. The Haworth structure. Partial hydrolysis products of dextran and their medical application.

58. Proteinogenic amino acids. Structure, nomenclature, acid-basic properties, bipolar structure. Stereoisomerism of natural α-amino acids with one and two chiral centers.

59. Biologically important reactions of α-amino acids. Deamination reactions (oxidative and non-oxidative). Hydroxylation reactions (phenylalanine — tyrosine, tryptophane — 5-hydroxy-tryptophane, proline — 4-hydroxyproline).

60. Decarboxylation reaction of α-amino acids — way to formation of biogenic amines and bioregulators (colamine, histamine, γ-amino butyric acid).

61. Peptides. Electronic and spatial structure of peptide bond.

62. Representatives of peptides and their biological significance (glutathione, neuropeptides, insulin).

63. Proteins. Organization levels of protein molecules and types of interactions in the stabilization. Primary, secondary (α-helix and β-conformation) and tertiary protein structures.

64. Haemoglobin, structure, properties.

65. Pyridine and purine heterocyclic bases, their aromaticity as reason of high stability.

66. Nucleotides. Structure of mononucleotides forming nucleic acids. Nomenclature. Hydrolysis of nucleotides.

67. Primary structure of nucleic acids. Phosphodiester bond. Ribonucleic and deoxyribonucleic acid. Nucleotide composition of RNA and DNA. Hydrolysis of nucleic acids.

68. Cyclic 3`,5`-AMP as neurotransmitter.

69. Steroids: classification, stereoisomerism. 5α- and β-steroids. Cholesterol: structure and biological role.

70. Estrogens and androgens: structure and biological role. Corticosteroids.

71. Bile acids: structure and biological role.

ANSWERS TO TESTS

Labwork № 1. Classification and nomenclature of organic compounds

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
2	2	4	4	1	1	3	2	1, 3, 4	3

Labwork № 2. Chemical bond structure and atom effects in the organic molecules

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
2, 3, 4	2, 4	3, 4	3	2	2, 3, 4	3	4	1, 2	A3 B2 C3 D4

Labwork № 3. Stereoisomerism, its role for biological activity demonstration

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
4	1, 3, 4	4	2	1, 3	4	1, 2	1,3	A2 B1 C4 D3	2

Labwork № 4. Hydrocarbons

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
1, 2, 3, 5	2, 4	1, 2	3	1, 4	2	1, 4	2	3, 5	2

Labwork № 5. Monofunctional hydrocarbon derivatives

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
2	4	1, 3, 4	1, 3	3, 4, 1, 2	2	2, 4	4	2	A3 B2 C1 D4

Labwork № 6. Biologically important reactions of aldehydes and ketones

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
2, 3	A2 B4 C1 D3	2	3	1, 3	1, 2	1, 3, 4	4	2	2

Labwork № 7. Carboxylic acid and their derivatives

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
1, 4, 3, 2	A4 B2 C3 D1	1	1, 2, 4	1, 3, 4, 5	2	1, 2, 4	2, 3, 4	3, 4	1, 4

Labwork № 9. Poly- and heterofunctional compounds

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
2	3	3	2	1, 3, 4	1, 2, 4	1	2, 3, 4	3, 4	1

Labwork № 10. Biological important heterocyclic compounds. Alkaloids

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
4	3	3, 4	1, 3	1, 3	2, 3	2	2, 3, 4	1	4

Labwork № 11. Carbohydrates. Monosaccharides

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
1, 2, 4	1, 2	3	2	2	2	4	3, 4	1	2, 4

Labwork № 12. Oligo- and polysaccharides

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
3	1, 2, 4	4	1, 2, 3	3, 4	2, 3, 4	4	3	3	2, 3, 4

Labwork № 13. Structure and reactivity of amino acids

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
1, 3	1, 3, 4	1, 4, 5	2, 3, 4, 5	1, 2, 3	3, 5	2	2, 3	1	3

Labwork № 14. Peptides. The levels of protein organization

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
2, 4, 5	2, 3, 4	1, 3, 4	1, 3, 5	1, 2, 3, 5	2	1, 2, 4	1	1, 3, 4	2

Labwork № 15. Nucleosides. Nucleotides. Nucleic acids

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
1, 3	3, 4	1, 3, 4	2	1, 3, 5	2	4	2	2	1

Labwork № 16. Lipids. Structure, properties. Lipid peroxidation

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
3	1, 2	2, 3, 5	4	2, 3	1, 3, 4	3	1	1, 4	1

Labwork № 17. Steroids. Alkaloids

Test 1	Test 2	Test 3	Test 4	Test 5	Test 6	Test 7	Test 8	Test 9	Test 10
2, 3	2, 3	1, 2, 4	4	2, 4	3, 4	2, 3	2, 4	1, 2	2

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Ринейская Ольга Николаевна
Ермоленко Елена Михайловна

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