**Al-Farabi Kazakh National University**

**Faculty of Biology and Biotechnology**

**Department of Biotechnology**

**PROGRAM OF THE FINAL EXAM BY THE DISCIPLINE**

**«Е 2217» Enzymology**

Educational program «6В05103 Biotechnology»

**Fall semester 2023-2024 academic year**

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| Course | 3 |
| Semester | 5 |
| Number of credits | 5 |
| Seminars | 30 |
| IWSP | 7 |

**Almaty 2023**

The program of the final exam by the discipline «Enzymology» of the specialty «6В05103 Biotechnology» was compiled by senior teacher Orazova S.B.

Considered and recommended at the meeting of the department of Biotechnology

Protocol No \_\_ of «\_\_» \_\_\_\_\_\_\_\_\_\_\_\_\_, 2023

Head of Department \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Kistaubayeva A.S.

The final exam platform – IS Univer

The final exam form – written exam, standard form

The final exam type – online, synchronous exam

The final exam contains 3 questions. The first block includes questions to metabolism of main low molecular bio compounds. The answer is estimated at 33 points. The second block includes questions about methods and techniques of studying of metabolites. The answer is estimated at 33 points. The third block includes questions about practical applications of metabolomics, which will find this course valuable. The answer is estimated at 34 points.

**Organization of the written online exam:**

The process of passing a written exam by a student involves the automatic creation of an examination ticket for a student, to which it is necessary to form a written answer by directly entering text into the system.

Check the Internet connection on a computer device (monoblock, laptop, tablet). The device must be provided with charging during the entire time of the exam.

Open the Univer.kaznu web portal.kz via any browser, but preferably via Google Chrome.

Log in with your account. If he does not remember his username and password, he must contact his supervisor-adviser before the exam begins.

Go to the Bachelor tab. Then activate the Exam Schedule functionality.

5. If the exam time has come, the command to pass the written exam will appear (highlighted in red). This means that the student can click on the link and answer the exam questions.

The student should use the "Start Exam" function on their exam schedule viewing page.

• The Pass written exam function is active only after the start of the exam time.

• The pass written exam function is available to the student for the duration of the exam.

• The pass written exam function is active only for those students who have unclosed final statements (exam, Retake, Incomplete).

• The pass written exam function is closed after the allotted time for passing the exam.

In the Univer IP, a student cannot attach files. He is obliged to enter his answer in the answer field using the computer keyboard in online mode.

After clicking on the link “Take a written exam”, a window will open where the student will see the questions of his exam ticket.

After the end of the time, the system will not accept written answers. Therefore, it is recommended to periodically save the printed answers on the page until the time expires. The page displays a timer by which the student can navigate by time. If during the exam, the students lost Internet connection or the student accidentally closed the page, then he must reconnect or log in again. During the exam, the student can return to the system and continue to answer the questions of his ticket. The system will automatically save the text every 10 minutes. There is a "Save" button on the ticket answer page, the student can click it when he decides to complete the written exam. After the answer is saved, the file will be automatically checked for originality.

The duration of the exam – 120 minutes, 1 attempt.

**Key issues**

Section 1. Introduction to Enzymology

The history of the development of enzymology. The relationship of enzymology with other sciences. Purpose Tasks, methods of enzymology. Definition of the science of enzymology. The role of enzymes in the body and their capabilities in comparison with chemical catalysts. The significance of the achievements of enzymology for biochemistry, molecular biology, biotechnology, medicine.

The structure of enzymes. Simple and complex enzymes. Holoenzyme, apoenzyme, coenzymes: cofactors and prosthetic groups. Principles of spatial organization of the enzyme molecule.

Section 2. Coenzymes A variety of the chemical nature of coenzymes. The chemical nature of enzymes. The molecular structure of enzymes. Active and allosteric centers. Contact and catalytic sections of the active center. Proferments. Apoenzymes and prosthetic groups of complex enzymes. Coenzymes, cofactors and their role in the catalytic process. Classification of coenzymes. Vitamins and coenzymes. Metals in the role of coenzymes and cofactors, their role in the action of enzymes. Polyfunctionality of metals in the realization of enzyme activity. Mechanism of reactions involving kinases.

Section 3. Classification, nomenclature and mechanism of action of enzymes

Classification and nomenclature of enzymes. The enzyme cipher (CF or EC). The significance and disadvantages of the unified nomenclature system.

Specificity of the action of enzymes, types of specificity. The importance of group-specific enzymes for metabolism in the cell. Enzymes with absolute specificity. Examples, the value for the cell.

Kinetics of enzymatic reactions. Theories of catalysis. Distinctive features of enzymatic catalysis. The effectiveness of enzymes. Formation of enzyme-substrate complexes. The use of the binding energy of the enzyme with the substrate in catalysis; types of catalysis used in enzymatic reactions; functional groups of enzymes. The dependence of the reaction rate on the concentration of the substrate. The Michaelis–Menten theory. Constants of the rates of formation and decay of enzyme-substrate complexes (small constants). The numerical value of the Michaelis constant and its practical significance. Determination of the Michaelis constant and the maximum reaction rate by the Linuiver–Burke method. The concept of enzymatic activity. Methods of expression of enzymatic activity.

Physico-chemical mechanisms of enzymatic catalysis. Active centers of enzymes, their topography. Methods for identifying functional groups of active centers. Physico-chemical mechanisms of enzymatic catalysis. The temperature optimum of the enzymatic reaction. Thermostable and thermolabile enzymes. Enzyme activity at low temperatures. The dependence of the reaction rate on the pH value of the solution. Effect of pH on the charge of ionogenic groups in protein molecules

Section 4. Regulation of the action of enzymes in a living organism

Regulation of the action of enzymes in a living organism. Levels of regulation of enzymatic activity. Allosteric enzymes. Non-covalent and covalent modification.

Induction and repression of enzyme biosynthesis. Activity of native enzymes. Specific factors that increase the activity of enzymes. Classification, mechanisms of action. The role of metal anions and cations in the activation of enzymes. Allosteric regulation of enzyme activity, the action of intermediate and final reaction products. Enzyme inhibitors: classification, mechanisms of action. Reversible and irreversible inhibitors. Inhibition constants. Competitive and allosteric inhibition of enzymes. Protein enzyme inhibitors. Covalent modification of the structure and activity of enzymes. Isoenzymes. Ribozymes.

Section 5. Methods of isolation and purification of enzymes

The basic principles of isolation and purification of enzymes. Classical methods. Extraction of enzymes from biological material. Acid treatment, heat treatment, fractionation with salts, organic solvents, selective adsorption method. Desalination with sephadex G-25. Ion exchange chromatography, gel filtration. Application of DEAE-cellulose for protein separation. Affinity chromatography, carriers and ligands. Ultracentrifugation, crystallization. Combining different methods for enzyme purification using the example of glucose-6-phosphate dehydrogenase of rat liver. Criteria for the purity of enzyme preparations.

Electrophoresis of proteins. Principles of electrophoresis. Native step electrophoresis. The physical principle of the method. Electrophoresis of proteins with DDS by Lammley. Universal staining of proteins in PAAG. Specific manifestation of proteins in PAAG. Specific manifestation of dehydrogenases. Detection of proteases. Isoelectrofocusing.

Kinetic parameters and physico-chemical properties of purified forms of enzymes. General rules for working with enzymes. Determination of the molecular weight of the native enzyme using the gel chromatographic method. Investigation of the catalytic properties of enzymes. Definition of the Michaelis constant. Conditions of the enzymatic reaction. Measurement of the rate of an enzymatic reaction.

Section 6. Enzymes in analytical chemistry

Methods for determining the activity of enzymes. Preparative enzymology. Methods of quantitative expression of enzymatic activity. Total activity, specific activity, molecular activity, activity of the catalytic center. Modern methods for determining the activity of enzymes: chemical, electrochemical, photometric, spectrophotometric, fluorimetric. The main absorption (spectrophotometric) methods for determining the activity of enzymes.

Section 7. Methods of obtaining and applying immobilized enzymes

Heterogeneous catalysts based on immobilized enzymes and cells. Methods of immobilization. Media. Immobilized cells of microorganisms. Immobilized enzymes: new properties. Industrial processes using immobilized enzymes and cells.

Section 8. Medical aspects of enzymology

The use of enzymes in medicine. The use of enzymes in medicine. The use of ribozymes in the treatment of viral infections and cancer. Immobilized enzymes and proteins as medicines. Therapy with immobilized enzymes. Enzyme immunoassay and its use in medicine. The main methods of ELISA. The use of ELISA.

Section 5. Synthesis and modification of organic compounds

Enzymatic conversion of cellulose into sugars. Cellulolytic microorganisms and enzymes. The mechanism of action of cellulases. The influence of the cellulose structure on the efficiency of its hydrolysis. Cellulase adsorption on cellulose and its role in catalysis. Fundamentals of biotechnology of enzymatic hydrolysis of cellulose.

Section 6. Biocatalytic methods of environmental protection

Creation of new technological processes with a minimum level of xenobiotic waste. Development of highly efficient processes of destruction of xenobiotics with the formation of products that are non-toxic to the environment. Features of the kinetics of biocatalytic processes. Adaptation of microorganisms to xenobiotics. Associations of microorganisms. Realization of "impossible" chemical reactions.

Section 7. Modern methods of designing enzymes with the necessary properties

Chemical modification. Combinatorial methods, genetic and protein engineering. Methods of directed evolution. Genetic engineering of enzymes.

**Information resources**

Literature: main, additional.

1. Punekar N.S. Enzymes: Catalysis, Kinetics and Mechanisms. 2018. P.560

2. Bisswanger H. Practical Enzymology. 2011. P.435

3. Copeland R.A. Enzymes: A Practical Introduction to Structure, Mechanism, and Data Analysis. 2012. P.341

4. Cornish-Bowden A. Fundamentals of enzyme kinetics, 4th edn. Wiley-Blackwell, Weinheim. 2012. P.462

5. Bugg T.D.H. Introduction to enzyme and coenzyme chemistry, 3rd edn. Wiley, Hoboken. 2012. P.521

Professional scientific databases

1. MATLAB Student Version Release R2016a; Simulink, Symbolic Math and nine other toolboxes– available from MathWorks

Internet resources

1. Enzyme nomenclature database https://enzyme.expasy.org/

2. The Ligand Chemical Database for Enzyme Reactions (LIGAND) https://libguides.library.albany.edu/c.php?g=537010&p=3677113

3. KEGG http://www.genome.jp/kegg/

4. http://elibrary.kaznu.kz/ru

**RUBRICTOR FOR CRITERIAL ASSESSMENT OF FINAL CONTROL**

**Discipline:** «Е 2217» Enzymology  **Form:** written exam, standard form, offline  **Platform:** IS Univer

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| **№** | **Score**  **Criterion** | **DESCRIPTORS** | | | | |
| **«Excellent»** | **«Good»** | **«Satisfactory»** | **«Unsatisfactory»** | |
| **90-100 % (30-33 points)** | **70-89 % (25-29 points)** | **50-69 % (20-24 points)** | **25-49 % (15-19 points)** | **0-24 % (0-14 points)** |
| **Question 1**  **33 points**  **Question 2**  **33 points** | Knowledge and understanding of the theory and concept of the course | The answer contains an exhaustive disclosure of the question, a detailed argumentation of each conclusion and statement, is built logically and consistently, supported by examples | The answer contains a full, but not exhaustive coverage of the issue, a shortened argumentation of the main provisions, allows violation of the logic and sequence of the presentation of the material. Stylistic errors and inaccurate use of terms are allowed in the response. | The answer contains incomplete coverage of the issue, the main provisions are superficially reasoned, violations of logic and sequence of presentation of the material are allowed in the presentation, theoretical provisions are not illustrated with practical examples | The answer contains incorrect coverage of the question posed, erroneous argumentation, factual errors, incorrect conclusions. | Ignorance of basic concepts.  Violation of the Rules of the final control |
| **№** | **Score**  **Criterion** | **«Excellent»** | **«Good»** | **«Satisfactory»** | **«Unsatisfactory»** | |
| **90-100 % (30-34 points)** | **70-89 % (25-29 points)** | **50-69 % (20-24 points)** | **25-49 % (15-19 points)** | **0-24 % (0-14 points)** |
| **Question 3**  **34 points** | Knowledge and application of the chosen methodology and technology to specific practical tasks | Full completion of the task, a detailed, reasoned answer to the question, followed by the solution of a practical problem.  Consistent, logical and correct justification of scientific provisions and the applied methodology and technology, literacy, compliance with the norms of scientific language, 1-2 inaccuracies in the presentation of the material are allowed, which do not affect the conclusions that are generally correct, visualization of the results of the justification, if necessary. | Partial completion of the task, incomplete, sometimes reasoned answer to a question with an incomplete solution of a practical problem; illiterate use of the norms of scientific language.  3-4 inaccuracies in the use of conceptual material, minor errors in generalizations and conclusions that do not affect a good overall level of task performance are allowed. | The material is presented in fragments, with a violation of logical sequence, factual and semantic inaccuracies are allowed, theoretical knowledge of the course is used superficially.  Conclusions on the applicability of sound scientific provisions are vague and unconvincing, there are stylistic and grammatical errors, as well as inaccuracies in the processing of the results of a practical solution | An irrational method of solving a task or an insufficiently thought-out response plan; inability to solve tasks, perform tasks in general; assumption of more than 4 errors and shortcomings; the presence of gross errors; conceptual material and argumentation are poorly used. | The task has not been completed, there are no answers to the questions posed, materials and analysis tools have not been used.  Violation of the Rules of the final control. |

**Formula for calculating the final grade:**

Final grade = score for question 1 + score for question 2 + score for question 3

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| Additional Information: |
| |  |  |  |  | | --- | --- | --- | --- | | **Letter Grade** | **Grade Point Value** | **Percentage** | **Conventional Grade** | | A | 4,0 | 95-100 | Excellent | | A- | 3,67 | 90-94 | | B+ | 3,33 | 85-89 | Good | | B | 3,0 | 80-84 | | B- | 2,67 | 75-79 | | C+ | 2,33 | 70-74 | | C | 2,0 | 65-69 | Satisfactory | | C- | 1,67 | 60-64 | | D+ | 1,33 | 55-59 | | D | 1,0 | 50-54 | | FX | 0,5 | 25-49 | Failure | | F | 0 | 0-24 | | I (Incomplete) | - | - | Incomplete (shall not be taken into account when calculating GPA) | | AU (Audit) | - | - | Audit (shall not be taken into account when calculating GPA) | | Cert. | - | 30-60 50-100 | "Certification" (shall not be taken into account when calculating GPA) | | Uncert. | - | 0-29 0-49 | "Uncertification" (shall not be taken into account when calculating GPA) | | R-difference | - | - | "Discipline difference on curriculum" (shall not be taken into account when calculating GPA) | |
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