# **Design of Multienzyme systems. Practical course**

Title of the Academic Program	Master's Degree Programs in English "Medical and Biological Physics"	
Type of the course	core /mandatory	
Course period	Fall semester, 1 semester (16 weeks)	
Study credits	4 ECTS credits	
Duration	144 hours	
Language of instruction	English	
Academic requirements	<ul> <li>BSc degree in Biology, Physics, Biophysics, Chemistry, Biochemistry, Environmental Sciences or equivalent (transcript of records),</li> <li>good command of English (certificate or other official document)</li> </ul>	

### **Basic Information**

### **Course Description**

The course is devoted to the practical study of modern methods of enzyme inhibition-based analysis and its application in medical and environmental fields.

### **Special Features of the Course**

The course examines the basics of enzymatic analysis based on the inhibition of single-enzyme system, coupled enzyme reaction or multienzyme system. The main principles and mechanisms of action of toxicants on enzymes are considered within lab activities and seminars. Laboratory works are practical exercises in which students learn the basics of working with enzyme analysis, determine the sensitivity of enzyme systems of varying complexity to model toxicants and test samples, such as saliva, water, soil, snow, etc. At seminars, students discuss modern research in the field of enzymatic analysis. As a final work, students should compose a multi-enzyme system, determine its sensitivity to model substances and conduct a medical or environmental analysis.

### **Course Aim**

The aim of the course is to introduce students to enzyme inhibitionbased analysis for medical and environmental research.

### **Course Objectives**

1) to enable students to build up their knowledge and skills pertaining to the use of enzymes for solving topical issues related to the life sciences;

2) to give students an introduction of recent trends in the area of enzyme inhibition-based analysis;

3) to provide students with practical skills of designing monoenzymatic and coupled enzyme assays, and multienzyme systems.

# Learning Outcomes of the Course (module)<sup>1</sup>

By the end of the course students will be able:

1) to demonstrate basic methods of designing bioluminescent and multienzyme systems;

2) to interpret the obtained data after enzyme inhibition-based analysis;

3) to independently collect information from professional publications;

4) to apply successful laboratory practice principles in experiment planning, conducting and control.

### **Teaching and Learning Methods**

The course consists of 6 modules: an introduction, 4 labs and a final project. Teaching Methods: learner-centered teaching, classroom discussion, individual research projects, laboratory experiments, creating and giving a presentation.

## **Course Structure**

Learning Activities	Hours
Practice sessions	24
Seminars	12
Self-study Assignments	108
Final Exam (including preparation)	0
Total study hours	144

### **Course (module) Outline**

Wee	Practice	Seminars	Assignmen	Hour	
k	sessions		ts	S	
Chapter 1 «Introduction»					
1	Lab safety Rules and Precautions	Basis of enzyme inhibition-based analysis	-	1/1/0	
Chapter 2 «Single-enzyme systems»					
2	Single-enzyme systems:	Examples of using single-enzyme	Make a report with a	2/1/8	

preparation for	systems for medical	presentation	
-	research		
Freedomen			
Single-enzyme	Examples of using	Make a report	2/1/8
	single-enzyme		
	systems for medical	-	
,	and environmental		
	research	article	
Coupled enzyme		_	2/1/8
systems:	couple enzyme		
preparation for	systems for medical	-	
analysis,	and environmental		
procedure	research	article	
Coupled enzyme	Examples of using	Make a report	2/1/8
	couple enzyme		
	systems for medical	-	
	and environmental		
determination)	research	article	
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		_	2/1/8
systems:	_		
preparation for		-	
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procedure	research	article	
Multi-enzyme	Examples of using	Make a report	2/1/8
-	multi-enzyme		
	systems for medical	1 <b>±</b>	
	and environmental		
	research	article	
Chapter 5	nzumo inhibition hass	donalucios	
Chapter 5 «E.		-	2/0/8
	-		4/0/0
Enzyme inhibition-		about the	
U U		application of	
		the analysis.	
meandar research		Participation	
		· · · · · · · · · · · · · · · · · · ·	
		in the	
		in the discussion. Searching	2/0/8
	Coupled enzyme systems: preparation for analysis, procedure Coupled enzyme systems: analysis (EC <sub>20</sub> , EC <sub>50</sub> determination) <u>Chapter</u> Multi-enzyme systems: preparation for analysis, procedure Multi-enzyme systems: analysis (EC <sub>20</sub> , EC <sub>50</sub> determination)	preparation for analysis, procedureand environmental researchSingle-enzyme systems: analysis (EC20, EC50 determination)Examples of using single-enzyme systems for medical and environmental researchCoupled enzyme systems: preparation for analysis, (EC20, EC50 determination)Examples of using couple enzyme systems researchCoupled enzyme systems: and environmental researchExamples of using couple enzyme systems for medical and environmental researchCoupled enzyme systems: analysis (EC20, EC50 determination)Examples of using couple enzyme systems for medical and environmental researchMulti-enzyme systems: preparation for analysis, (EC20, EC50 determination)Examples of using researchMulti-enzyme systems: preparation for analysis, procedureExamples of using researchMulti-enzyme systems: analysis, (EC20, EC50 determination)Examples of using multi-enzyme systems for medical and environmental researchMulti-enzyme systems: analysis (EC20, EC50 determination)Examples of using multi-enzyme systems for medical and environmental researchMulti-enzyme systems: analysis 	preparation for analysis, procedureand environmental researchon the selected articleSingle-enzyme systems: analysis (EC20, EC50 determination)Examples of using single-enzyme systems for medical and environmental researchMake a report with a presentation on the selected articleCoupled enzyme systems: preparation for analysis, procedureExamples of using couple enzyme systems for medical and environmental esearchMake a report with a presentation on the selected articleCoupled enzyme systems: analysis, (EC20, EC50 determination)Examples of using couple enzyme systems for medical and environmental researchMake a report with a presentation on the selected articleCoupled enzyme systems: analysis (EC20, EC50 determination)Examples of using couple enzyme systems for medical and environmental researchMake a report with a presentation on the selected articleMulti-enzyme systems: analysis, (EC20, EC50 determination)Examples of using multi-enzyme systems for medical and environmental researchMake a report with a presentation on the selected articleMulti-enzyme systems: analysis (EC20, EC50 determination)Examples of using multi-enzyme systems for medical and environmental and environmental eselected articleMulti-enzyme systems: analysis (EC20, EC50 determination)Examples of using multi-enzyme systems for medical and environmental and environmental eselected articleMulti-enzyme systems: analysis (EC20, E

	based analysis for environmental research		about the application of the analysis. Participation in the discussion.		
Chapter 6 «Final project_»					
12- 15	Final project	Final project	Make a presentation for the final project defense.	7/2/36	
16	-	Presentation of the final project	Make a presentation for the final project defense.	0/3/8	

## **Course Instructor, Contact information**



Mrs Elizaveta M. Kolosova Junior research associate, the Laboratory of Bioluminescent Biotechnology e-mail: EKolosova@sfu-kras.ru Tel.: +7 (391) 206-23-07

### Assessment

The course assignments include:

- Labs (preparation, execution, progress report), each successfully completed lab 8 points;
- Seminars: presentations, discussions max 20 points;
- Final project max 40 points.

To pass the course a student should have more than 85 points.

## **Attendance Policy**

Students are expected to attend all courses and course activities for which they are registered. Any class meeting missed, regardless of cause, reduces the opportunity of learning and may adversely affect a student's achievement in the course. Students are required to attend at least 90% of the class meetings in order to receive credit for the course. An accurate record of attendance will be kept for each course. If a student misses one third or more of a class session, the student will be counted absent.

### Web page of the course

The webpage of the course is available through E-learning SibFU web site: www.e.sfu-kras.ru. You must be logged in to access this course. Course Guide and all accompanying materials are also available at the course webpage.

#### **Core reading**

The books:

- Nelson D. L. and Cox M. M. Lehninger Principles of Biochemistry. 5th edition. (http://archive.org/)
- Lee J. Bioluminescence, the Nature of the Light. 2016. (http://athenaeum.libs.uga.edu/)
- Shimomura O. Bioluminescence: Chemical Principles and Methods, Revised Edition. 2012. (http://ebookcentral.proquest.com/lib/krasu-ebooks/)
- Lakowicz J. R. Energy Transfer. In Principles of Fluorescence Spectroscopy. 3rd edition. 2006. Springer. ISBN: 978-0-387-46312-4 (Online) (http://link.springer.com)

Journal articles:

- Esimbekova E.N., Torgashina I.G., Kalyabina V.P., Kratasyuk V.A. Enzymatic Biotesting: Scientific Basis and Application // Contemporary Problems of Ecology, 2021, Vol. 14, No. 3, pp. 290–304. DOI: 10.1134/S1995425521030069
  - Sutormin O. S., Sukovataya I. E., Pande S., Kratasyuk V. A.Effect of viscosity on efficiency of enzyme catalysis of bacterial luciferase coupled with lactate dehydrogenase and NAD (P) H: FMN-Oxidoreductase //Molecular Catalysis. – 2018. – V. 458. – P. 60-66.
  - Kolosova E.M., Sutormin O.S., Stepanova L.V., Shpedt A.A., Rimatskaya N.V., Sukovataya I.E., KratasyukV.A. Bioluminescent enzyme inhibition-based assay for the prediction of toxicity of pollutants in urban soils // Environmental Technology & Innovation. – 2021. – V. 24. – P. 101842.
  - Kalyabina V. P., Esimbekova E. N., Torgashina I. G., Kopylova K. V., Kratasyuk V. A. Principles for construction of bioluminescent enzyme biotests for analysis of complex media // Doklady Biochemistry and Biophysics. – Pleiades Publishing, 2019. – V. 485. – №. 1. – P. 107-110.

#### **Facilities, Equipment and Software**

A student should bring their laptop or print learning materials in advance. Learning materials are provided to students in either Adobe PDF, Microsoft Office, Google files or compatible formats.

Laboratory work is carried out in specially equipped rooms. The laboratory room should contain the following equipment and reagents: a spectrophotometer, a luminometer, a pH meter, an analytical balance, pipettes, labware, enzymes (e.g. NAD(P)H:FMN- oxidoreductase, bacterial luciferase, dehydrogenases, tripsin), their substrates and other required reagents and salts.