

## **KB3003 (7,5 hp)** **Biochemistry II for chemistry and molecular biology majors**

Course Coordinator: Rob Daniels

Room: K439

### **Aim**

This Biochemistry II course is designed for chemistry and molecular biology majors that want to establish a strong foundation in the basic biochemistry that contributes to prokaryotic and eukaryotic cell homeostasis. It can also be taken as a free-standing course for students with general interest in biochemistry. The topics will include: nucleotides, sequencing reactions, amino acids, lipids, pH, transcription, translation, protein folding, enzyme kinetics, signal transduction and gene regulation. The course is organized over six weeks with lectures and lab practicals that are designed to complement the classroom curriculum. Students will be expected to discuss basic biochemical principles, perform lab experiments, and critically evaluate their findings. The main aim of this course is to strengthen the biochemical foundation of each student and provide exposure to practical applications of biochemistry.

### **Upon completion of the course you should be able to:**

- Define the basic chemical properties of nucleotides, amino acids, lipids, and buffers
- Explain DNA synthesis reactions and their application in modern sequencing
- Compare and contrast prokaryotic and eukaryotic gene organization with respect to transcription
- Explain how mRNAs are translated by ribosomes into polypeptides and proteins
- Describe enzyme functions (reaction mechanisms), enzyme kinetics and regulation
- Integrate the processes of signal transduction and gene regulation
- Design experiments to express/isolate proteins and analyze enzyme-substrate interactions

### **You will be expected to:**

- Attend the lectures and read the assigned literature
- Actively participate in the lab and discussions with other students and faculty
- Complete the laboratory exercises on time and present the results in a scientific manner
- Take a final exam consisting of short answer questions at the end of the course

### **Assessment**

1. Laboratory – Written lab reports, active participation in the discussions and lab.  
\*Attendance at all labs is a requirement to pass the course and the reports must be completed within two weeks of the final exam date to receive a grade.
2. Theory - Final exam consisting of short answer questions – Exam questions are written to assess the knowledge level regarding the key concepts outlined in the course and lecture intended learning outcomes (ILOs).

### **Instructor**

Dr. Rob Daniels, office: A423, e-mail: [robert.daniels@dbb.su.se](mailto:robert.daniels@dbb.su.se)

### **Required Course Literature (6<sup>th</sup> Ed is also applicable)**

Cox, Michael M., and David L. Nelson. (2017). *Lehninger - Principles of Biochemistry*. (7<sup>th</sup> Ed.). New York, NY: W. H. Freeman. (Cost ~800 SEK text + 80 SEK for eBook)

### **Recommended Complimentary Literature**

Lodish, Harvey, et al. (2013). *Molecular Cell Biology*. (7<sup>th</sup> Ed.). New York, NY: W. H. Freeman.

**COURSE SCHEDULE**

Date	Time	Room	Activity	Teacher	Reading Lehninger <sup>7</sup>
<b>15/2 Thur</b>	9-12:00	K439	Course Introduction Lecture 1: Properties of cellular molecules	RD	Ch.1 (p. 1-32) Ch.2 (p47-69)
<b>16/2 Fri</b>	9-12:00	K439	Lecture 2: DNA and RNA synthesis and the application to next generation sequencing	RD	Ch.8 (p. 281-99) Ch.9 (p. 281-99) Ch.25(1009-25) Ch.26(1057-81)
<b>19/2 Mon</b>	9-12:00	K439	<i>Lab Introduction</i>	RD/TAs	
<b>20/2 Tues</b>	9-12:00	K439	Lecture 3: Amino acids, translation, protein structure & folding	RD	Ch. 3 (75-88) Ch. 4 (115-150) Ch.27(1077-110)
<b>21/2 Wed</b>	9-12:00	K439	Lecture 4: Biochemistry of protein targeting	RD	
<b>22/2 Thur</b>	9-12:00	K439	Lecture 5: Protein function - ligand binding	RD	Ch. 5 (157-178)
<b>23/2 Fri</b>	9-12:00	K439	Lecture 6: Enzyme catalysis, inhibition and regulation	RD	Ch. 6 (187-225)
<b>*26/2 Mon</b>	9-10:00	K439	<i>Prelab and inoculate large cultures</i>	TAs	
	10-12:30	K439	Lecture 7: Prokaryotic & eukaryotic gene regulation	RD	Ch. 24(957-979) Ch. 28(1127-169)
	14-17:00	Lab	<i>(Induce culture, take samples, and final pellet)</i>	TAs	
<b>*27/2 Tues</b>	9-10:00	K439	<i>Prelab</i>	TAs	
	10-16:00	Lab	<i>(Protein purification and SDS PAGE)</i>		
<b>*28/2 Wed</b>	9-12:00	K439	Lecture 8: Signal transduction across membranes	RD	Ch. 12(437-474)
	13-16:00	Lab	<i>(Analyze SDS PAGE and Dialysis)</i>		
<b>1/3 Thur</b>	9-12:00	K439	Lecture 9: Biochemical analysis of influenza viral membrane proteins and RNAs	RD	
<b>*2/3 Fri</b>	9-10:00	K439	<i>Prelab</i>	TAs	
	10-16:00	Lab	<i>(Protein quantification and storage)</i>		
<b>*5/3 Mon</b>	9-10:00	K439	<i>Prelab</i>	TAs	
	10-17:00	Lab	<i>(Gene amplification by PCR and transformation)</i>		
<b>*6/3 Tues</b>	9-10:00	K439	<i>Prelab</i>	TAs	
	10-16:00	Lab	<i>(Clone identification)</i>		
<b>*7/3 Wed</b>	9-10:00	K439	<i>Prelab</i>	TAs	
	10-13:00	Lab	<i>(DNA isolation and sequencing)</i>		
<b>*8/3 Thur</b>	9-10:00	K439	<i>Prelab</i>	TAs	
	10-16:00	Lab	<i>(Enzyme analysis #1 titration pH optimum)</i>		
<b>*9/3 Fri</b>	9-10:00	K439	<i>Prelab</i>	TAs	
	10-16:00	Lab	<i>(Enzyme analysis #2 Km and Vmax titration)</i>		
<b>*12/3 Mon</b>	9-12:00	K439	<i>Lab help session sequence results, calculations</i>	TAs	
<b>13/3 Tues</b>	14-16:00	K439	Pre-exam help session	RD	
<b>19/3 Mon</b>	13-18:00	TBA	Final Exam		
		TBA	Re-Exam		
<b>4/4 Wed</b>			Lab Report Deadline**		

Prelabs will occur in the classroom K439 prior to lab

\*-Lab component scheduled for this day (see lab schedule for details)

\*\*--Please email robertd@dbb.su.se to report any delays or problems with lab reports

TBA – To be announced

## Biochemistry II – Laboratory

Instructor: Rob Daniels

Rooms: K232, K242 & M320

### Aim

The lab module of this Biochemistry I course is designed to provide students with the opportunity to apply the class concepts in an investigative research project for one month that can potentially result in publishable biochemical data. The project will utilize several molecular biology and biochemical techniques in a supervised setting to reconstitute an enzymatic pathway. Each group will collect data on the enzymatic properties of their proteins; compare their values with other groups and with respect to literature to decide on the suitability of the data for publication. The isolated enzymes will then be combined in an attempt to reconstitute a catabolic process in vitro. The overarching goal of this lab is to strengthen the practical skills in biochemistry and expose each student to the scientific process.

### Upon completion of the lab portion of the course you should be able to:

- Define the goal of an experiment
- Describe the biochemical principles utilized in each experimental method (see list below)
- Explain the experimental results in a scientific manner
- Compare and contrast the results obtained by other groups
- Discuss and critically evaluate your work in reference to relevant literature
- Interpret your results to develop a hypothesis

### You will be expected to:

- Attend and actively participate in the lab with other students and teachers
- Explain the scientific basis behind the techniques used in the labs
- Complete the laboratory exercises on time and present the results from each part in a report

**Report 1:** Protein expression and purification

**Report 2:** Gene isolation and subcloning into an expression vector

**Report 3:** Enzyme property analysis

### Methods covered:

- Plasmid and primer design, transformation and selection.
- Polymerase chain reaction (PCR), cloning, agarose gel electrophoresis, and sequencing
- Inducible expression in *E. coli*, protein purification (affinity tag), dialysis, SDS-PAGE
- Protein quantification
- Determine enzymatic properties, pH optimum,  $K_m$ , and  $V_{max}$

### Laboratory Assistants

Henrik Östbye [henrik.ostbye@dbb.su.se](mailto:henrik.ostbye@dbb.su.se)

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Magdalena Rzepka [magdalena.rzepka@dbb.su.se](mailto:magdalena.rzepka@dbb.su.se)

## Lab schedule

- Labs will be in rooms K232, K242 and M320 -

Date	Time	Room	Lab	Activity	TA
19/2 Mon	9-12:00	K439		Lab introduction, how to write a lab report, and design primers	RD HÖ, RE, MR
*26/2 Mon	9-15:00	K232	#1- Protein expression, purification and quantification	Start large culture, induce expression, harvest the pellet & store	HÖ, RE, MR
*27/2 Tues	9-17:00	K232		Protein purification and SDS-PAGE	HÖ, RE, MR
28/2 Wed	13-16:00	K232		Analyze SDS-PAGE and dialysis	HÖ, RE, MR
*2/3 Fri	9-16:00	K232		Protein quantification and storage (Lab Report 1)	HÖ, RE, MR
*5/3 Mon	9-17:00	K232	#2 - Gene amplification & subcloning	Gene isolation by PCR & transformation	HÖ, RE, MR
*6/3 Tues	9-16:00	K232		Clone identification	HÖ, RE, MR
*7/3 Wed	9-13:00	K232		DNA isolation and preparation for sequencing	HÖ, RE, MR
*8/3 Thur	9-16:00	K232	#3 - Enzymatic analysis & properties	Enzyme analysis 1: Enzyme titration & pH optimum	HÖ, RE, MR
*9/3 Fri	9-17:00	K232		Enzyme analysis 2: Determine $K_m$ & $V_{max}$	HÖ, RE, MR
12/3 Mon	9-12:00	K439		Help session with sequencing results (Lab Report2) & enzyme calculations (Lab Report 3)	HÖ, RE, MR
4/4 Wed				Lab Report Deadline**	

\*Prelab before beginning at 9:00 in classroom K439

\*\* -Please email robertd@dbb.su.se to report any delays or problems with lab reports

***There are large time windows during incubations.  
Use this time to talk to your TAs and prepare the lab reports.***