Methods in Biochemistry (KB 7014)

start at 10 am on 23-3-2021 – end 30-4-2021

AIM

'Methods in Biochemistry' will introduce students to the fundamental principles behind several basic and more advanced techniques commonly used in biochemical research. During the course, the strengths and weaknesses of each technique will be covered to explain which techniques are suitable for particular applications. Upon establishing this foundation, the students will be taught how to combine different methods to overcome problems associated with the individual techniques. The theoretical aspect of the course will be supplemented with a lab practical where the students will apply a number of these techniques themselves (unfortunately, due to Corona this year no practicals will be organized). In the end, the students should be able to design strategies to address complex biological questions using the techniques they have been exposed to from a theoretical and practical perspective.

Upon completion of the course you should be able to:

- Describe the principles behind a number of common biochemical techniques.
- Explain the strengths and weaknesses of a technique for particular applications.
- Combine different biochemical methods to address a complex biological question.
- Troubleshoot biochemical methods based on their scientific principles.
- Analyze generated data and communicate them in writing and orally.
- Read, communicate and critically evaluate course-related scientific literature.

You will be expected to:

- Attend and actively participate in the lectures and tutorials.
- Participate in discussions with other students and the faculty.
- Read the assigned literature and complete the assignments and lab reports on time.
- Present the lab practical results in writing and make the appropriate corrections (unfortunately, due to Corona this year no practicals will be organized).

COURSE CONTENT

-Cell homogenization and fractionation The organization and chemical composition of pro- and eukaryotic cells will be discussed in relationship to different homogenization methods, which are used to enrich for particular fractions from these cells.
-Materials on how to make buffers and solutions (incl. sterilization), pH, the metric system, statistics and making lab. notes are provided at the beginning of the course.

-'History of molecular biology' Students will be taught the history behind the main organisms used for molecular biology and how these are handled, genotyped, cultured and stored.

-Centrifugation The theoretical basis for multiple centrifugation techniques will be explained as well as their applications. These include: differential centrifugation, density gradient centrifugation and analytical ultracentrifugation.

-Recombinant DNA techniques Several modern DNA analysis methods will be covered ranging from DNA isolation and sequencing to PCR and a variety of molecular biology techniques for DNA manipulation. Significant time will be spent on: restriction enzyme digests and other reactions that modify DNA, PCR, primer design, sequencing methods, homology cloning, Gibson cloning, recombineering, Cas9-based genome editing, forwarded evolution etc.

-Protein production Several different eukaryotic and prokaryotic protein production platforms are available. These will be thoroughly explained from both a theoretical and need perspective based on the current protein production bottlenecks. Specifically, we will address how to choose an organism, the expression system, and the design of the target gene.

-Protein isolation using physiochemical properties The biochemical principles for a number of basic protein purification techniques will be covered. This will include the buffer systems and resins that support: affinity chromatography, ion exchange chromatography, hydrophobic interaction chromatography, and gel-filtration. We will also go over which of these are suitable for HPLC and why.

-Antibodies and their applications We will discuss the factors to consider for generating a polyclonal/monoclonal antibody, alternative binding approaches that can replace antibodies, and several applications such as: immuno-blotting, immuno-precipitations, and ELISAs.

-Mass spectrometry Basics of mass spectrometry, MALDI-TOF, MS/MS (incl. sequencing proteins), quantitative proteomics (*e.g.*, iTRAQ, iCAT, spectral counting), characterization of post-translational modifications using MS etc.

-Gel-electrophoresis and detection methods Different gel-electrophoresis techniques for DNA and protein will be explained as well as a number of methods for staining and detection.

-Microarrays DNA + protein arrays, RNA-seq, ribosome profiling etc. (if time allows)

Lectures and tutorials All the lectures are linked to tutorials, which are a significant part of the course. For the tutorials the students get assignments ('homework'), *i.e.*, they have to answer questions based on the lectures and articles they have to read. The assignments are handed in, checked (feedback) and graded before the tutorial (-: fail, +: good, ++: very good). In case the tutorial is graded with a 'fail', the tutorial has to be corrected until it is graded with at least a 'good'. To be able to finish the course all tutorials should have been graded with a 'good' or 'very good'. There is one 'special' tutorial: the cloning tutorial. In the cloning tutorial the students design constructs (primer design, plasmid design, they learn how to use different computer programs to facilitate this).

Practical (unfortunately, due to Corona this year no practicals will be organized).

-Protein production practical Primer design, PCR, cloning, overexpression screening, monitoring, protein production, cell breakage, centrifugation, protein characterization (SDS-PAGE, immuno-blotting).

The students write a lab. report that is corrected (until the level of the reports is acceptable)(unfortunately, due to Corona this year no practicals will be organized).

Assessment
Written open book home exam, and the student have to complete all tutorials and lab. reports on time (unfortunately, due to Corona this year no practicals will be organized and therefore do not).

Final mark:
-Exam 100% (at least 50% of the points are needed).
-Tutorials should have been handed in and either be '+' or '++' plus the lab reports should be done (given that they have been handed in on time)

Practical matters

There will be no more than 15 students.

Setup

Due to Covid, the setup of the course has been changed quite a bit. There is a Athena link where we will put all course materials: **https://athena.itslearning.com** (if you are registered as a student at SU, you should have access to this link). I will send you an Email when new materials have been uploaded. Importantly, I have pre-recorded parts of some of the lectures. There will be also Zoom-based direct contact lectures to further discuss the content of the pre-recorded lectures. All tutorials will also be Zoom-based. Direct contact lectures and tutorials will be recorded and made available to you (I assume that I have your permission to do this). I will organize the course programme and set dates for handing in the tutorials answers based on the progress we are making. You send your tutorial answers to me by Email (degier@dbb.su.se, label your Email and file with answers with your name and the topic of the tutorial). I will send you information/updates using Email. Make sure that every day you are available between 10 and 12 in the morning. I have made a recurring Zoom link. Again, based on progress made we will plan our direct contact meetings (not all days there will be meetings, but since we do not know when meetings are needed make sure that you are available every day between 10 and 12!).

Zoom link
Jan-Willem de Gier is inviting you to a scheduled Zoom meeting.

Topic: Jan-Willem de Gier's Zoom Meeting
Time: This is a recurring meeting Meet anytime

Join Zoom Meeting
https://stockholmuniversity.zoom.us/j/66258570488

Meeting ID: 662 5857 0488

In case you cannot make contact through Zoom send an Email to degier@dbb.su.se.

The teachers:
-JWdG: Jan-Willem de Gier (degier@dbb.su.se)

Tutorials:
The students have to hand in the answers before the tutorials and they have to attend the tutorials. The teachers have a look at the answers before the tutorial. **A mark will be given; - (not ok),+ (ok),++ (good). The students have to hand in the answers two days (48 hrs) before the tutorial** (send your answers to me by Email as described above)**. Answers that are handed in too late won't be considered.**

PRACTICAL:

When the practicals start you can pick the students up at room K241 (unfortunately, due to Corona this year no practicals will be organized).

Assistants:
NA

Book + course materials:
Principles and Techniques of Biochemistry and Molecular Biology (8th edtion). Edited by Andreas Hofmannn and Samuel Clokie (Cambridge University Press). Hand-outs, articles etc. will be provided.

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Some golden rules:
-Be on time (10 o'clock we start).
-Lectures, tutorials and the practical (practical NA) are compulsory: i.e., you must attend them. Not attending seminars (lectures) and tutorials, and not handing in the assignments before the deadline will result in being excluded from doing the exam. If there is a problem you contact Jan-Willem de Gier (degier@dbb.su.se). Attendance of seminars (lectures), tutorial and the practical (practical NA) will be monitored using lists that have to be signed.
-If the score of a tutorial was '-' a corrected version of the assignement has to be handed in within 2 days after the tutorial. All tutorials should be '+' or '++' to be able to do the exam.
-Lab reports handed in after the deadlines won't be considered. No lab reports means no final mark.
-For being able to do the overexpression practical the molecular biology tutorial has to be passed (practical NA).
-All the students should have signed the SU papers stating what SU considers plagiarism and what happens if plagiarism is detected before they start with KB7014. If plagiarism is detected in first versions of lab reports and assignments this will also be reported to the university.
-Students make their own assignements and lab. report (practical NA) (this of course should not prevent students from having discussions etc.).