Cloning Club

Course code: 921244

1 Instructors

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2 Course Description

The course consists of two parts: a weekly discussion workshop and a following practical part. Participation in the practical part requires previous participation in the discussion workshop (but not vice-versa):

1. Weekly discussion workshop (8 events each 1 to 1.5 hours) starting Tuesday 30.8.2016; venue: meeting room 7 (Biomedicum Helsinki, 5th floor, except 27.09. and the last workshop on 25.10., which held in BM B136A); 1 credit

2. "Decentralized" lab course (at the participants schedule and venue or - for participants without own access to the necessary facilities - in the first two weeks of November in our lab); 1 credit

Important: Participants can team up into teams of two and share a cloning task! This is especially advisable for more complex (multi-step) cloning tasks!

Maximum amount of participants for the practical part: 16. No limit for the workshop.

3 Why this course structure?

Similar to the 2014 course, the goal is to teach practical molecular biology & genetic engineering. However, the course format has changed radically, as we realized that having a live person give didactic university lectures is now obsolete and we should adapt to this reality. A live person is only needed for interactive learning and to answer questions that are not answered by the online learning tools. In addition, digital natives learn at least as efficient when watching videos or listen to podcasts of lectures at their own pace and schedule compared to sitting in a class room and having to pay attention at someone else's demand.

Since we do not know in advance the participants' cloning tasks, the topics of the lectures are going to be determined only after participants have handed in their task description. However, we certainly will discuss: Restriction enzyme cloning, the Gateway system, gene synthesis, DNA sequencing, mutagenesis techniques, Gibson cloning (and competing technologies), Golden Gate Assembly, BioBricks.

4 Why do I have to become a cloning expert?

Over billions of years, evolution has shaped life's complex machinery. The collective knowledge that has built up by this process is vast and genetics is the key to that knowledge. While we need to understand life at the protein and organism level, it all starts with the DNA. Hence, genetic engineering is not only here to stay, but will become more and more important: we are just scratching the surface of its potential.

The overwhelming part of biomedical research starts with a DNA construct: expression vectors to transfect cells, shuttle plasmids to make viruses, constructs to generate transgenic animals, etc. Hence, the skill to generate a DNA construct is still needed until the arrival of that long promised, commerically available desktop device, which will split out any plasmid a few hours after you have fed it the plasmid's sequence.

A plethora of new cloning technologies and services have become available. In fact, it maybe today more difficult to clone than 25 years ago. 20 years ago, there was practically only one way to do clonings ("restriction enzyme cloning"), while nowadays the options are endless and you need to choose the right strategy to maximize success and speed.

5 Is this course right for me?

If you need to do cloning and you have not much previous experience, this is your couse. If you want to become a cloning expert, this might be your course.

Participants need a basic understanding of cloning: either from previous lectures/courses or by going thru online resources. Every participant should have an own cloning task related to her/his studies. In exceptional circumstances, we can also give out cloning tasks. During the the weekly discussion courses (8 occasions), we'll discuss the tasks and identify the appropriate methods. If there are local experts available for a specific task, we try to invite them for the discussion.

In the practical part, students are expected to carry out the cloning. We have found that arranging the practical part as a block course was demanding and stressfull for all participants and hence we want to decentralize the effort. Students will be able to perform the individual cloning steps at their own discretion and pace after the their task has been discussed at the weekly meeting.

With all practical questions concerning their cloning, students are encouraged to consult us for discussion and help any time during the semester. If students have no access to necessary cloning infrastructure in their own lab, they can carry out the practical part in the first two weeks of November within our premises. Since we know that specialized cloning reagents are not available in many labs, the course organizers will provide the necessary materials within the course budget. In the planning of the cloning, financial considerations will play a significant part since given an unlimited budget, one could always opt to outsource the complete cloning procedure.

6 Course Materials & Textbook

If you have a basic understanding of how cloning works, you are good to go. If not, or if you need to refresh your knowledge, there are a many introductory online lectures by universities, individuals or companies about cloning, most of which are available via YouTube. As a starting point:

- MIT Open courseware: http://ocw.mit.edu/courses/biology/7-01sc-fundamentals-of-biology-fall-2011/recombinant-dna/
- Shomus Biology: http://www.shomusbiology.com/recombinant-dna-technology.html

I did not find any good, up-to-date online textbooks available from the Helsinki University Library. The latest (4th) edition of the *Maniatis* (Molecular Cloning: A Laboratory Manual) is quite ok, but only available as “paper” copy at the Helsinki University Library Meilahti branch. The textbooks below are available online. The first one is fair, but already 8 years old; the second is newer, but only very few of the chapters are useful:

- An Introduction to Genetic Engineering (Desmond S. T. Nicholl, Cambridge University Press, 3rd Edition 2008), available online from http://biomolfa.edublogs.org/files/2009/04/genetics-engineering-08.pdf or http://www.e-bookspdf.org/download/introduction-to-genetic-engineering-nicholl-3rd-edition.html
- Comprehensive Biotechnology (Murray Moo-Young, Elsevier B.V., 2nd Edition 2011), available in electronic format via http://www.terkko.helsinki.fi

Additional course materials will be made available via the instructor's web pages.

7 Course Requirements

• SnapGene or Serial Cloner software (for the “in-silico” cloning of the DNA construct). A free one-month trial version of SnapGene can be downloaded from http://www.snapgene.com, Serial Cloner is

freeware. To complete their tasks, students may use alternative software if they can provide the results of their tasks in a sufficiently annotated, accessible format (e.g. GenBank). If you do not have access to a computer to which you can install these programs by yourself, please contact us well in advance to get the software installed or access to a computer with the software. Since the evaluation period for SnapGene is not long enough to cover the course period, we will try to get longer trial licenses from the software vendor (which they wer happy to do last time we asked). We also have two unused licenses that we can lend for the course period.

• One DNA engineering task per team. We recommend that each 2 participants will form a team. Team assignment can be made by the participants themselves in advance.

• Due to the limited course budget, specific reagents for your individual cloning project. If you are performing an individual cloning project, your laboratory has to pay for the specific reagents for the experiments, which are;

- cDNA: we get the sequence for your gene of interest from somewhere (most human and mouse cDNAs are available for a nominal fee of 10€ from the Genome Biology Unit in Viikki. We have also many common cDNAs availahble, please check with us before ordering!
- oligonucleotides for mutagenesis/PCR/sequencing (not common oligonucleotides) and the sequencing itself (13.93€/run if you do it here, but you may choose any other, more affordable services, e.g. the free introductory offer by ACGT Inc.)

The necessities of above reagents depends on your specific project, which we'll discuss during the workshop.

8 Course Objectives

The primary goal of this course is to introduce basic concepts and methods of molecular biology and genetic engineering, with emphasis more on practicalities of the engineering than on genetics. Knowledge in Molecular Biology will be helpful in understand the principle of various methodologies used in genetic engineering. Upon the completion of this course, you shall be able to:

1. Understand the basics of molecular cloning and genetic engineering
2. Know the most important methodologies in molecular cloning and genetic engineering
3. Design and construct a DNA construct

9 Grading Policy/Assignments

Workshop

1 credit (credit requirements: 2/3 participation and a successful cloning task performed “in silico”. The task can be either choosen by the team or given by the organizer. Each team needs to propose and design a genetic engineering task. For a successful course completion, teams need to hand in the documentation of their project. This does not have to be necessarily extensive, but rather like an extended/supplemental Materials & Methods section of a manuscript (sufficiently detailed for the reader to be able to replicate the cloning, including a map of the resulting construct).

Practical course

1 credit. The “in-cilico” and the practical cloning task should be the same for those that participate in both the discussion work shop and the practical course. Since these are real world clonings, there is no guarantee of success and hence success is not a requirement for the credit. However, 75% of cloning tasks were succesfully completed during our last course (and all remaining ones at least partially).