



**MIC**  
Molecular Imaging Center



**UNIVERSITY OF OSLO**



**Nordic Caenorhabditis elegans Network**

**Program**  
**Course in Confocal Microscopy of *C. elegans***  
**20.-22. January 2010.**

<b>Wed 20.01.10</b>		<b>Thu 21.01.10</b>			<b>Fri 22.01.10</b>				
1000-1045	Introduction		0900-0945	Lab:IF 2. ab + mount live worms (A+B+C)	Live cell imaging (D+E+F meets 0830!)	0830-0915	Image processing (Endy)		
1015-1045	<i>C. elegans</i> presentation		0945-1030	Lab: IF 2. ab (D+E+F)	Live cell imaging (A+B+C)	0915-0930	<i>Break</i>		
1045-1130	Basic concepts of confocal imaging (Hege)		1030-1045	<i>Break</i>		0930-1130	Image processing workshop (A+B)	Workshop Zeiss II (C+D)	Scanning electron microscopy (E+F)
1130-1215	<i>Lunch</i>		1045-1130	Multi fluorescence labeling (Hege)		1130-1200	<i>Lunch</i>		
1215-1300	Optimization of image acquisition (Endy)		1130-1200	<i>Lunch</i>		1200-1400	Image processing workshop (E+F)	Workshop Zeiss II (A+B)	Scanning electron microscopy(C+D)
1300-1445	IF 1. ab (A+B+C)	Confocal basics (D+E+F)	1200-1330	Workshop Zeiss I (E+F)	Mount IF + radiation (A+B+C+D)	1400-1600	Image processing workshop (C+D)	Workshop Zeiss II (E+F)	Scanning electron microscopy(A+B)
1445-1500	<i>Break</i>		1330-1500	Workshop Zeiss I (A+B)	Mount IF + radiation (E+F)	1600-1630	Evaluation and diploma		
1500-1645	IF 1. ab (D+E+F)	Confocal basics (A+B+C)	1500-1630	Workshop Zeiss I (C+D)					

### ***Lectures:***

***C. elegans* presentation (Hilde Nilsen) 30 min**

**Basic concepts of confocal imaging (Hege Avsnes Dale) 45 min**

Confocal microscopes have three major differences from a conventional fluorescence microscopy; lasers, a scanner and a pinhole. The basic concepts of confocal imaging will be explained.

**Optimization of image acquisition (Endy Spriet) 45 min**

What is a digital image? Resolution in 3 dimensions, what limits the resolution of confocal images?  
How to acquire the optimal image?

**Multi fluorescence labeling (Hege Avsnes Dale) 45 min**

Principle of fluorescence and excitation and emission spectra of fluorochromes. How to combine different fluorochromes and avoiding pitfalls like cross talk (bleed through).

**Image processing (Endy Spriet) 45 min**

A confocal image can be processed in many ways, either to make it look better or to carry out measurements or quantifications. Software exist that cover most wishes and some options are included in the processing software supplied with the microscopes. More complicated operations are performed on specialized, licensed software, but there are also free downloads on the web that can perform a whole range of operations. Focus will be on the most common operations and prepare you for the workshop at the software workstations.

### ***Workshops:***

Most of the time students work in groups of 4, occasionally 6.

### **Lab work:**

The students will prepare their own samples. They will perform extraction of gonads from the *C. elegans* worms and immuno labeling with primary antibodies (1. ab) and secondary fluorescent antibodies (2. ab). In addition they will introduce DNA damage by UV irradiation on worms and prepare the worms for live cell imaging by mounting them in agar with anesthetic agent. You may also bring your own samples.

**Confocal basics on Zeiss (1 ¾ h):**

General training on the microscope. We will go through the software and how to set it up to acquire multi fluorescence images.

**Live cell imaging** (¾ h):

Introduction to live cell imaging and DIC imaging on the Leica SP5 confocal and/or NIKON fluorescence microscope. Group D, E and F needs to meet 830 Thursday morning to mount live worms.

**Workshop on Zeiss I** (1 ½ h):

Students will on their own try to set up the confocal and acquire images of their prepared samples (fixed) from the lab.

**Workshop on Zeiss II** (½ h mounting and 1 ½ h on the microscope):

Students will be introduced to live cell and DIC imaging on the Zeiss confocal. Prior to that they have to mount live worms.

**Image processing** (2 h):

The image processing will take place on a PC-work station. The students will be presented some tasks on image processing. They will process the images they have acquired during the course using programs like Zeiss LSM Examiner, ImageJ and Imaris.

**Scanning electron microscopy** (2 h):

An introduction to the scanning electron microscope (SEM). Make nice surface images of pre prepared of *C. elegans* worms. (This may be optional for the students...)

**Practical Information**

- Registration deadline is **December 1<sup>st</sup>**. Register by sending e-mail to Hilde Nilsen: [hilde.nilsen@biotek.uio.no](mailto:hilde.nilsen@biotek.uio.no)
- Maximum 12 participants can be accommodated. PhD students and post docs affiliated with the Nordforsk *C. elegans* network will be prioritised.
- Accepted participants will be notified by December 15<sup>th</sup>.
- Registration fee of 1000 NOK will be paid directly by Nordforsk for Nordforsk affiliated participants. External participants will have to pay (details will be given upon acceptance).
- Participants accepted to the course will be notified by December 15<sup>th</sup>.
- Participants must make their own travel arrangements.
- Rooms have been reserved at Rica Strand Hotel (<http://www.strandhotel.no>). Participants must book directly with the hotel using the reference number: 20091556.
- For more information, please contact <http://www.nordiccelegans.org/> or contact: Hilde Nilsen, University of Oslo, The Biotechnology Centre: [hilde.nilsen@biotek.uio.no](mailto:hilde.nilsen@biotek.uio.no)