



# Welcome to the 10<sup>th</sup> MIC confocal microscopy course at the Dept. of Biomedicine, University of Bergen 4<sup>th</sup> of March - 7<sup>th</sup> of March 2014

The course focuses on the basic principles of confocal microscopy, the understanding of what a confocal microscope can do, and how to acquire the optimal images. We will also discuss what a digital image is made up of and introduce you to software for improvement of images and preparation/quantification for publishing. Some previous experience with confocal imaging is clearly an advantage but not a requirement.

We are happy to present this course to you and we hope it will be both useful and interesting!

You will all receive an invoice for the course fee of NOK 4500,-. This includes dinner the second evening (unfortunately, no discount can be made if people do not join), and lunch the 4<sup>th</sup> day.

#### Information regarding accommodation:

Haukeland hotel (55209200) and Regines Gjestehus (55979600) are both within a short walking distance, but practically all hotels in the city centre will be quite close to a bus stop for the lines 2, 3 and 12 that will take you to the bus stop Haukeland Nord. The bus takes approx. 10 minutes.

# Course program:

Day 1 (Tuesday 4 <sup>th</sup> of March)		Day 2 (Wednesday 5 <sup>th</sup> of March)		Day 3 (Thursday 6 <sup>th</sup> of March)		Day 4 (Friday 7 <sup>th</sup> of March)	
1000-1015	Welcome, intro, coffee	0900-1100	Software presentation and demonstration -	0900-1000	Imaris (H)	0830-1030	Workshop 4
1015-1130	Basics of confocal microscopy (H)		group B	1000-1015	Coffee break	1030-1045	Coffee break
1130-1215	Lunch	1100-1115	Coffee break	1015-1115	Image J / Fiji (E)	1045-1245	Workshop 5
1215-1330	Optimal image acquisition (E)	1115-1200	Sample preparation (M)	1115-1200	Do and don'ts in image processing (M)	1245-1330	Common lunch with evaluation (Seminar room 7 <sup>th</sup> floor)
1330-1415	Fluorochromes &	1200-1245	Lunch	1200-1245	Lunch		Workshop 6
	fluorescent proteins (H)	1245-1345	Photoshop CS4 / 5 (M)	1245-1445	Workshop 2		
1415-1430	Coffee break	1345-1400	Coffee break	1445-1500	Coffee break		
1430-1530	Image representation and processing (E)	1400-1600	Workshop 1	1500-1700	Workshop 3		
1530-1730	Software presentation and demonstration - group A						
		1800-	Common dinner				

H: Hege A Dale

M: Michaël Marie

E: Endy Spriet

#### The lectures:

# **Basics of confocal microscopy:**

We will compare conventional fluorescence microscopy and confocal microscopy, focusing on the basic principles of confocal imaging.

#### **Optimal image acquisition:**

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

#### Fluorochromes and fluorescent proteins (FPs):

How to combine different fluorochromes?

An overview will be given of the many different FPs, focusing on advantages and disadvantages of the most commonly used ones.

#### Sample preparation:

Optimal sample preparation is the most important prerequisite of successful confocal imaging. We will focus on different strategies of preparing your samples, potential problems and tricks to try when things get tough.

#### Imaging representation and processing:

A digital image can be processed in many ways, either to make it look better or to carry out measurements or quantifications. We will see how simple display adjustments change the perception of an image. We will learn how to use the histogram and lookup-table to optimize image representation. We will also briefly cover the role of human perception.

# Presentation of Leica/Zeiss software:

Introduction to the Leica or Zeiss hardware and software.

# **Presentation of Adobe® Photoshop® software:**

Introduction to the commercial Adobe® Photoshop® software.

#### **Presentation of Imaris software:**

Introduction to the commercial, scientific image visualization and analysis software Imaris.

# **Presentation of Fiji software:**

Introduction to the free, scientific image visualization, processing and analysis software Fiji.

# DO's and DON'Ts in image processing:

What is image processing and what is manipulation? Which guidelines do I have to follow when creating a figure panel for a publication? What is a "representative" image?

#### The workshops:

**General training** will be with Endy on the Leica confocal and Hege on the Zeiss confocal. We will go through the most common operating modes of the confocal microscopes, learn how to acquire a correct image and how to acquire Z-stacks, and if time allows it go through more specialized operation modes.

We will have two **confocal hands on** sessions where people are divided into groups of 3 people. On the second session you will be at the microscope mostly on your own and practice what they have recently learned. In the second session you can also bring your own samples.

**Software sessions on Photoshop, Imaris and Fiji** will be held at a PC-station with 4 PCs. We will give out some tasks for you to solve to learn simple image processing, and tip you on some useful freeware to download at "home".



In order to fit everyone into the schedule, all groups will have some free time at two different occasions during the four days.

We invite participants to bring their own samples to the course. The second confocal hands on session is dedicated to imaging of own samples.