

<b>Live Cell Imaging Facility Microscopy course</b> <b>Course #2870 (everything)</b> <b>Course #2871 (light and dark green)</b> <b>Public lectures (dark green). No registration required</b>		
<b>07.04</b>	<b>Topic: Detecting and avoiding bleed through, antibody staining and sample carriers</b>	
09:00-10:00	Welcome, introduction to the course structure, tour of the facility	
10:00-10:10	<i>Coffee and fruits break</i>	
10:10-11:10	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
11:10-12:10	The different types of fluorophores Spectral detectors, virtual filters and unmixing	Sylvie Le Guyader, KI
12:10-13:00	<i>Lunch</i>	
13:00-14:45	Bleed through video: peer review of quiz and discussion.	
14:45-15:00	<i>Coffee and buns break</i>	
15:00-16:00	Antibody staining troubleshooting	Edward Verwayen, Cell Signaling Technology
16:00-17:00	The different types of plates, dishes, chambers...	Tina Freisinger, Ibidi
<b>08.04</b>	<b>Topic: Microscopes and objectives</b>	
09:00-10:00	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
10:00-10:10	<i>Coffee and fruits break</i>	
10:10-12:10	Types of light microscope systems: upright/inverted, wide field/confocal Specifications of objectives Which objective for which application? Deconvolution	Sylvie Le Guyader, KI
12:10-13:00	<i>Lunch</i>	
13:00-14:00	Quiz on previous lectures	
14:00-14:45	Answers to quizz on previous lectures	
14:45-15:00	<i>Coffee and buns break</i>	
15:00-16:00	Workshop: Spectral Unmixing or STORM or student imaging challenge	
16:00-17:00	Workshop: Spectral Unmixing or STORM or student imaging challenge	
<b>11.04</b>	<b>Topic: Cameras and fluorophores</b>	
09:00-10:00	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
10:00-10:10	<i>Coffee and fruits break</i>	
10:10-11:10	High throughput imaging	Victoria Menendez Benito, KI
11:15-12:15	Fluorophore specifications (quantum yield, brightness...) How to judge what a 'good' fluorophore is	Vladana Vukojevic, KI
12:15-13:00	<i>Lunch</i>	
13:00-14:00	Quiz on previous lectures	
14:00-14:45	Answers to quizz on previous lectures	
14:45-15:00	<i>Coffee and buns break</i>	
15:00-16:00	Workshop: Spectral Unmixing or STORM or student imaging challenge	
16:00-17:00	Workshop: Spectral Unmixing or STORM or student imaging challenge	
<b>12.04</b>	<b>Topic: Resolution and contrast</b>	
09:00-10:00	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
10:00-10:10	<i>Coffee and fruits break</i>	
10:10-12:10	Resolution and contrast Aberrations and diffraction Point Spread Function Confocal aperture size. Airy unit, Rayleigh criterion, Abbe's limit and MTF curves What is the Nyquist sampling theory and how to use it	Kjell Carlsson, KTH
12:10-13:00	<i>Lunch</i>	
13:00-14:00	Quiz on previous lectures	
14:00-14:45	Answers to quizz on previous lectures	
14:45-15:00	<i>Coffee and buns break</i>	
15:00-16:00	Workshop: sample preparation or objectives and RI or student challenge	
16:00-17:00	Workshop: sample preparation or objectives and RI or student challenge	
<b>13.04</b>	<b>Topic: Confocal settings</b>	
09:00-10:00	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
10:00-10:10	<i>Coffee and fruits break</i>	
10:10-11:10	Camera and PMT properties. How to select the correct camera for your application	Colin Coates, Andor Technology

11:10-12:10	typical workflow of how to set a confocal When do we care about resolution?	Sylvie Le Guyader, KI
12:10-13:00	Lunch	
13:00-14:00	Quizz on previous lectures	
14:00-14:45	Answers to quizz on previous lectures	
14:45-15:00	Coffee and buns break	
15:00-16:00	Workshop: sample preparation or objectives and RI or student challenge	
16:00-17:00	Workshop: sample preparation or objectives and RI or student challenge	
<b>14.04</b>	<b>Topic: xyz automation and fast imaging, intro to multiphoton microscopy</b>	
09:00-10:00	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
10:00-10:10	Coffee and fruits break	
10:10-12:10	How to find the area of interest in a large sample with minimum bleaching? Hardware versus software autofocus, focus surface, how to avoid focus drift Options for fast imaging Saturation, underexposure, bit depth and fluorophore saturation (video) Dealing with autofluorescence (sudan black, bleaching before staining)	Sylvie Le Guyader, KI
12:10-13:00	Lunch	
13:00-14:00	MetroloJ analysis of refraction index mismatch workshop	
14:00-14:45	MetroloJ analysis of refraction index mismatch workshop	
14:45-15:00	Coffee and buns break	
15:00-16:00	Workshop. Koehlering or spinning disk versus resonant or student challenge	
16:00-17:00	Workshop. Koehlering or spinning disk versus resonant or student challenge	
<b>15.04</b>	<b>Topic: Volume imaging</b>	
09:00-10:00	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
10:00-10:10	Coffee and fruits break	
10:15-11:15	The problem of imaging a fluorescent volume: Refractive index matching and scale distortion	Sylvie Le Guyader, KI
11:15-12:00	introduction to light sheet microscopy	Stephan Werk, LaVision Biotech
12:10-13:00	lunch	
13:00-14:45	introduction to 2P: Widefield vs confocal vs multiphoton including NDDs, clearing. Two photon microscopy theory, practice, advantages and limitations. Intravital imaging. SHG, THG	Sylvie Le Guyader, KI
	Review of the exam questions so far	
14:45-15:00	Coffee and buns break	
15:00-16:00	Workshop. Koehlering or spinning disk versus resonant or student challenge	
16:00-17:00	Workshop. Koehlering or spinning disk versus resonant or student challenge	
<b>18.04</b>	<b>Topic: Statistics in imaging</b>	
09:00-10:00	2 students presents their imaging challenge (5 min presentation, 25 min brain storming)	
10:00-10:10	Coffee and fruits break	
10:10-10:40	Go through colin's lecture about cameras	Sylvie Le Guyader, KI
10:40-12:10	Statistics in imaging	Tessma Mesfin Kassaye, KI
12:10-13:00	Lunch	
13:00-14:00	2P workshop 8 students or exam questions of the previous day	
14:00-14:45	2P workshop 8 students or exam questions of the previous day	
14:45-15:00	Coffee and buns break	
15:00-16:00	Workshop: light sheet or student challenge or self assessment	
16:00-17:00	Workshop: light sheet or student challenge or self assessment	
<b>19.04</b>	<b>Topic: Tissue processing, live cell imaging, colocalization</b>	
09:00-10:00	Tissue processing for imaging	Jan Mulder, SciLife
10:00-10:10	Coffee and fruits break	
10:10-11:10	Imaging live cells: how to assess the cell's health, how to reduce light toxicity	Sylvie Le Guyader, KI
11:10-12:10	Colocalization	Jeremy Adler, UU
12:10-13:00	Lunch	
13:00-14:00	Quizz on previous lectures	
14:00-14:45	Answers to quizz on previous lectures	
14:45-15:00	Coffee and buns break	
15:00-16:00	Workshop: light sheet or student challenge or self assessment	
16:00-17:00	Workshop: light sheet or student challenge or self assessment	
<b>20.04</b>	<b>Topic: Image processing and quantitative analysis</b>	
09:00-10:00	Presentation of ImageJ/FIJI. Image analysis strategy: what to think about before you start imaging	Sylvie Le Guyader, KI

10:00-10:10	<i>Coffee and fruits break</i>	Carolina Wälhby
10:10-12:10	Cell Profiler workshop	
12:10-13:00	<i>Lunch</i>	
13:00-17:00	Cell Profiler workshop	
<b>21.04</b>	<b>Topic: Data handling, data management</b>	
09:30-10:00	Super-resolution overview	Sylvie Le Guyader, KI
10:00-10:10	<i>Coffee and fruits break</i>	
10:10-12:10	Awareness about potential problems in image processing Recommendations by major scientific journals about modifying images for publication The different image formats The safe way of saving images and backing them up Recommendation for data management How to format images for publishing or poster printing Trends and future challenges in light microscopy	Sylvie Le Guyader, KI
12:10-13:00	<i>Lunch</i>	
13:00-14:00	Vernissage workshop: microscopy images troubleshooting	
14:00-14:45	exam questions	
14:45-15:00	<i>Coffee and buns break</i>	
15:00-16:00	exam questions	
16:00-17:00	zen intro	
<b>22.04</b>	<b>Topic: visualizing protein-protein interactions, examination</b>	
09:00-10:00	FRET and FLIM	Arne Lindqvist, KI
10:00-10:15	PLA, ICS, ICCS, BIFC	Sylvie Le Guyader, KI
10:15-10:30	Conclusion and feedback	
10:30-10:45	Microscopy as a source of inspiration: the 50 cents microscope	Manu Prakash (YouTube)
10:45-?	Examination and fruits	

## Presentation of the teachers at the Live Cell Imaging facility course 2016

### **Carolina Wählby**



Carolina is a Professor in Quantitative Microscopy at the Centre for Image Analysis, Dept. of Information Technology and SciLifeLab at Uppsala University. Her lab develops advanced methods and software tools to quantify and mine the rich information present in microscopy images. She will run a one day workshop in Image Analysis.

### **Jeremy Adler**



Jeremy works mainly at the core imaging facility (BioVis) at Uppsala U on microscopy & image analysis and also at the BMC in Ingela Parmryd's group on live imaging and colocalization measurements. He will tell the students how to avoid the potential dangers associated with colocalization.

### **Victoria Menendez-Benito**



Victoria is a group leader at the Department of Biosciences and Medical Nutrition at KI. Her group focusses on the role of centrosomes in asymmetric cell division. For this they screen yeast libraries using fluorescence imaging and image analysis. She will share with students many tips about high throughput image-based screens.

### **Kjell Carlsson**



Kjell is a Professor at the KTH School of engineering sciences where he teaches Applied Physics including microscopy and photography. He will help students understand the physics behind imaging.

### **Sylvie Le Guyader**



Sylvie started the Live Cell Imaging facility at KI in 2008 after 10 years of research using microscopy. Her experience was enriched by training hundreds of users with a wide panel of samples. She now runs the facility as well as the intensive microscopy course.

### **Tobias Nyberg**



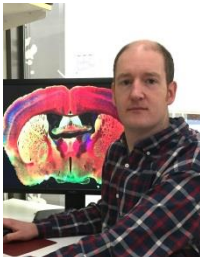
Tobias is a researcher at the KTH School of Technology of Health. He also works half time at the Live Cell Imaging facility. He will run workshops about imaging speed and refraction index mismatch.

### **Gabriela Imreh**



Gabriela used microscopy a lot in her research at KI. She now works full time at the Live Cell Imaging facility. She will run several workshops on sample preparation, spectral unmixing and give loads of tips and tricks.

### **Jan Mulder**



Jan is the group leader of the Fluorescence Tissue Profiling facility at KI/Scilife. The aim of the group is to identify proteins involved in brain development, normal brain physiology and pathophysiology of brain disorders. For this they use a vast antibody collection and perform multiplex fluorescence imaging. Jan will give a lecture on how to prepare tissue before imaging.

### **Vladana Vukojević**



Vladana is a group leader at the KI Department of Clinical Neuroscience. Her lab studies how opioid receptor-mediated signaling is perturbed by abuse of drugs and alcohol. For this they use functional Fluorescence Microscopy Imaging (fFMI), a multiplexed approach combining high-resolution fluorescence microscopy imaging and fluorescence correlation spectroscopy. Vladana will give a lecture on fluorophores.

### **Arne Lindqvist**



Arne is a group leader at the KI department of Cell and Molecular Biology. His group focuses on the regulation of cell division that they study using fluorescence microscopy and FRET sensors. He will give a lecture about how to quantify protein-protein interaction using the FRET technique.

### **Tessma Mesfin Kassaye**



Messfin is a Lecturer and Statistical Consultant at the KI Department of Learning, Informatics, Management and Ethics and that you are involved in teaching, assignments and research in the field of medical statistics. He will teach the students about statistics applied to microscopy images.

### **Marie Andersson**



Marie has used STORM in her research. She now works at Bergman Labora, the company that represents Nikon in Sweden. She will run a STORM workshop on the facility equipment.

### **Colin Coates**

Colin is Product Manager at Andor, a company well-known for their scientific cameras. Colin will give a lecture on how to understand camera specifications and what to look out for when imaging fluorescence with a camera.

### **Edward Verwayen**

Edward is a Key Account Manager for Cell Signaling Technology, a company that specializes in antibody production and testing especially in the domain of cancer research.

### **Tina Freisinger**

Tina is Application Specialist at Ibidi, a company that produces many types of sample carriers such as plates, dishes, perfusion chambers, multiwell slides... She will present the type of special carriers that are very useful to image live cells in different conditions but that researchers are often not aware exist.

### **Stephan Werk LaVision Biotech**

Stephan has professional experience in marine sciences and applied physics. He represents LaVision Biotech in the Nordic region and specializes in the Multiphoton and Light Sheet microscope systems. He will give a lecture about Light Sheet microscopy as well as run a workshop on a demo system. Students are welcome to bring their own samples.