Day 1: Deconvolution, Data handling, and Visualization

08:30 Welcome + Installation of the Huygens software 08:40 - 09:50 Introduction

General introduction to microscopy Image Formation and Image Restoration

- What is image restoration?
- Deconvolution
- Point spread function

09:50 -10:10 Coffee break

10:10 - 11:10 Huygens and Batch processing

Preferences & Settings in Huygens How to start a deconvolution job in <u>Huygens (Professional)</u>, and how to schedule multiple deconvolution tasks with the <u>Batch Processor</u> HANDS-ON

11:10 -11:30 Converting and rescaling of image data

How to prepare images in Huygens, treat large image data, converting image dimensions, scaling, and <u>File formats</u>

11:30 – 11:50 Visualization Part I: 2D visualization

Twin Slicer and OrthoSlicer fine tricks HANDS-ON

11:50 - 12:15 Visualization Part II: 3D visualization Huygens MIP and SFP renderers

HANDS-ON: <u>Huygens Surface renderer</u> and <u>Movie Maker</u>

12:15 - 13:00 Lunch Break

13:00 – 14:00 General advice and Hands-on

What algorithm to choose, differences between microscope types, optimal parameter settings, deconvolution artifacts and how to prevent them HANDS-ON

14:00 – 14:20 Measuring and distilling PSF's

Distilling an experimental PSF from bead images, PSF quality HANDS-ON

14:20 – 14:40 Coffee Break

14:40 – 14:50 GPU accelerated deconvolution

14:50 – 16:00 STED & Airyscan processing HANDS-ON

Day 2: Huygens Restoration and Analysis

08:30 – 09:20 How to Optimize your Image acquisition

How to deal with image distortions and <u>acquisition pitfalls</u> Issues that will be addressed are for example: noise, blurring, bleaching, hot-pixels, spherical aberration, bleedthrough, drift.

09:20 – 09:40 Chromatic Aberration Corrector & Crosstalk Correction

Correcting chromatic aberration with the <u>Chromatic Aberration Corrector</u> HANDS-ON

09:40 – 10:00 Drift & movement correction - Object Stabilizer

Correcting unwanted movement and vibration with the <u>Object Stabilizer</u> HANDS-ON

10:00 - 10:20 Tile Stitching

Automatic vignetting, deconvolution and stitching in one workflow. HANDS-ON Stitcher

10:20 – 10:50 Coffee break

10:50 - 11:10 Quiz

Find and fix imaging issues HANDS-ON

11:10 – 11:40 Object Tracking

How to detect and analyze tracks of moving objects with the $\underline{\mbox{Object Tracker}}$ HANDS-ON

11:40 – 12:30 Colocalization Analysis

<u>Colocalization Analyzer</u> HANDS-ON

12:30 – 13:15 Lunch break

13:15 – 14:00 Measuring in 3D: Object Analysis Analyzing your microscopy images with the <u>Object Analyzer</u> HANDS-ON

14:00 – 14:30 Light-Sheet deconvolution & Fusion PRESENTATION

14:30 – 14:50 Coffee break

14:50 – 16:00 Working with your own data or demo images HANDS-ON



What?: 2-day workshop on optimizing microscopy imaging, image restoration, deconvolution and analysis for all your microscopy images

When?: 14 + 15th of May, 2019

Where?: Kopenhagen

For whom?:

For any researcher in Denmark working with fluorescence microscopy. In particular when you use one of the of the following imaging techniques:

- ✔ Widefield/epi-fluorescence
- ✓ Confocal or Multi-Photon
- ✓ Airyscan
- ✓ STÉD
- ✓ Spinning-disk
- ✓ Light-sheet

What will be covered?:

- ✓ Optimizing image acquisition for reliable image analysis
- ✓ Image restoration
- ✓ Deconvolution & Batch Processing
- ✓ Image visualization (2D and 3D)
- ✓ 3D Object Analysis
- ✓ 3D Colocalization
- ✓ Object Tracking (tracking moving objects)
- ✓ Hands-on: both with demo-data and your own data
- ✓ And much more!

Interested to join?:

Send an e-mail to Vita Solovyeva (vita@bmb.sdu.dk)



Huygens Professional User Interface



GATTAquant nanorulers (140nm) imaged with Zeiss Airyscan and deconvolved with Huygens. Courtesy: Dr. Christoffer Lagerholm (WIMM, University of Oxford)



Get the most from your microscopy data! Huygens image deconvolution, restoration, and analysis

Microscopy images show you no more than what the microscope is willing to tell you about the object being investigated. Artifacts like blurring, noise, drift, chromatic aberration and crosstalk all distort your image and affect the 'true' object distribution. Consequently, image interpretation and subsequent co-localization analysis and quantitative objects measurements are severely perturbed. Fortunately, most of these distortion can be measured. For example, the Point Spread Function (PSF), the basic building block from which your acquired image is constructed, can be calculated and used within the Huygens software to trace back this construction process to the original constituent object, thus restoring it. As a result, Huygens significantly improves image contrast and resolution, and decreases noise levels.

Typical improvements of 1.5x to 2x the optical resolution limit, and at least a 2x increase in contrast can be achieved with Huygens deconvolution. This allows researchers to obtain more reliable data, which facilitates the subsequent visualization, segmentation, and analysis of the imaged objects. The significant gain in contrast that can be obtained even permits faster image acquisition over longer periods of time with less bleaching and photo-toxicity issues.

With the new Huygens Batch Express option, deconvolution can be fully automatically performed on images from a wide range of microscope types, including widefield, confocal, Airyscan, spinning disk, multi-photon, 3D STED, and Light Sheet. Besides having the most advanced GPU-accelerated deconvolution algorithms currently available, Huygens also contains a complete set of restoration options to address other imaging problems like for example drift, crosstalk, and chromatic aberration. Additional colocalization and object analysis options may complement your imaging workflow.

Huygens is suited for beginners, advanced users, and multi-user facilities. Tests licenses can be requested on <u>www.svi.nl</u>

