Workshop – "Scientific Image Processing and Analysis"

Content:
The workshop – "Scientific Image Processing and Analysis" aims to teach natural scientists from different areas of life science how to handle and process digital images starting from e.g. microscopic image acquisition until the incorporation into the final publication figure. This includes important theory about high quality digital images in general (e.g. how to achieve good resolution) as well as a broad spectrum of methods for scientifically correct image processing and specific analytical purposes according to high scientific standards. You will learn how to extract most information from your images and how to quantify objects and intensities. Additionally, the workshop includes a lot of hands-on sessions and explains how to save time during repetitive image processing steps or how to build your publication figures in a way that preserves image quality and stores processing data. You will be able to revisit the learned material using the provided exercises and script also later on. The workshop content is generally of importance for scientists working with digital images and their analysis. Optional topics will be adjusted to meet the participants needs as good as possible if there is time left.
Furthermore, specific participant question regarding image processing or solutions for analysis issues can be personally discussed if communicated beforehand (e-mail with question and example images).

Specific Topics (among others):

- Basics about correct image acquisition
  - How to achieve good image resolution (sampling)
  - Image formats - which formats serve scientific images and which should be avoided
  - Metadata - information saved beyond the visible image
  - Information content of images - learn about bit-depth, color spaces and different image types (or: how much information can be saved in and retrieved from an image).
- Correct image adjustments avoiding alterations - contrast and brightness, image rotation, size changes, background subtraction methods.
- Use of different image filters to improve extractability and preparation for further analysis
- Image segmentation - How to extract specific objects of interest (e.g. cells positive for a certain marker stain)
- Automated object counting and tracking of moving objects (optional)
- Basic 3D reconstructions (e.g. microscopic z-stacks, or medical MRI-Data)
- Image Quantifications (selected topics depending on participants field of interest):
  - Measurements of areas, length, …
  - 3D object analysis (volume, surface, distances, angles…)
  - How to correctly measure intensities in images (e.g. fluorescence)
- Dimension scaling and intensity calibration of images
- Labeling of images and time series/movies (text, numbers, scale bars, calibration bars,…)
- Ethics in image handling, processing and publication - where are the limits?!
- How to efficiently prepare publication figures.

Aim:
The workshop should give scientists a better understanding about the Do's and Don'ts during digital image processing and insight in the methodology of extracting a multiplicity of information from their images. The participants will gain extensive knowledge about the possibilities they have to analyze their imaging data.

Target Group:
PhD Students and PostDocs which are working or plan to work with digital images. The workshop has a focus on microscopic images and life science applications but all the content is applicable for digital images of different origin in general. No previous software knowledge required.

Methodology
During the practical parts of the workshop we will use the professional software Fiji (customized ImageJ bundle). All necessary software will be provided and installed during the workshop. Software is free to use (open source).

Trainer:
Dr. Jan Brocher (www.biovoxxel.de)

2.5-day BioVoxxel Workshop
"Comprehensive Image Processing and Analysis"

Basics in Microscopy and Imaging

- Correct Illumination
- Signal-to-noise and background
- Pixels and voxels
- Resolution and its limit
- Imaging artifacts
- Correct image sampling

Digital Images

- Image formats
- Image compression and artifacts
- Metadata handling
- Bit-depth
- Human vision and digital images
- True-color and pseudo-color images

Introduction into the ImageJ/Fiji software

Scientifically Correct Image Adjustment

- The histogram
- Correct contrast adjustments
- Color spaces
- Image transformation (size, rotation,...)
- Background subtraction methods
- Image filters

Image Segmentation

- Thresholds (8-bit images)
- Create and modify binary images
- Feature extraction methods
- Thresholding true-color images
- Image segmentation decision tree

Higher Dimensional Images in Fiji

- 3D, 4D, 5D images
- Visualization of n-dimensions
- 3D-Reconstruction
- Montages

Counting and Tracking

- Computer assisted counting
- Automatic counting of objects applying different methods

Quantitative Image Analysis

- Prerequisites for image quantification
- Image scaling and calibration
- Area and length measurements
- Quantification in 3D (volume, surface,...)
- Measuring intensities

Customized Automation using Macros

- Introduction into the macro language
- Recording and writing small macros
- Batch application of macros

Specific Analyses Techniques *according to participants needs*, such as...

- Densiometry of Western blots
- Statistical co-localization analysis
- Automated tracking of moving objects
- 3D Segmentation (object extraction)
- Image Stitching

(selected topics from the above listed)

Image Annotation

- Labeling with overlays
- Time-stamps
- Scale bars
- Calibration bars

Publication Figures

- Documentation and ethics
- Image data integrity preservation
- Effective figure composition
  - FigureJ

2.5-day intensive workshop (~21 hours)

Extensive preparation for good microscopic image acquisition, image processing and extensive quantitative image analysis as well as publication figure preparation

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