Processing images for papers & posters: CHEAT SHEET

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Open & Save

Recommended:
Plugins > Bio-Formats > Bio-Formats Importer
Alternative:
File > Open ... or ***Ctrl + O*** or ***Drag & Drop***

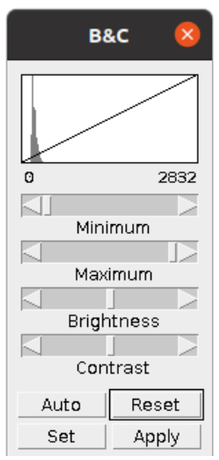
Review existing metadata:
Image > Properties... - *Ctrl + Shift + P*

File > Save As ...
TIFF: analysis, quantification
PNG: presentation, figure
AVI: movie, animation, GIF

TIP: Duplicate image before processing, keep raw image intact:
Image > Duplicate... - Ctrl + Shift + D

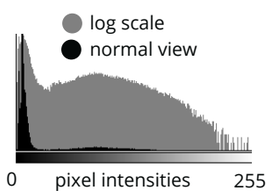
Brightness & Contrast

Image > Adjust > Brightness/Contrast...
Ctrl + Shift + C



- ❑ Auto: saturates the image by 0.35%
- ❑ Reset: to min & max or 0-255 for 8-bit
- ❑ Set: fixed values - use for comparisons
- ❑ Apply: histogram stretch using set min & max. **Use with caution!**

Check for problems with intensity sampling:
Analyze > Histogram... - ***Crtl + H***



Pixel count
Intensity range:
8-bit image: 0-255
16-bit image: 0-65535

TIP: For optimal acquisition refer to:
Jonkman et al. 2020 and Jost & Waters 2019

Image histogram examples

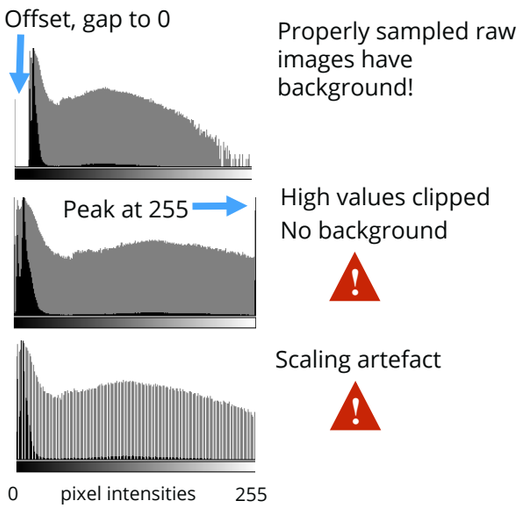


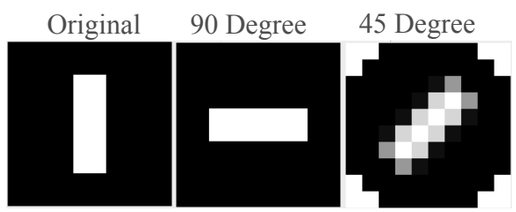
Image Processing

Gaussian blur:
Process > Filters > Gaussian Blur...

Projection:
Image > Stacks > Z Project...

Rotation & Resizing

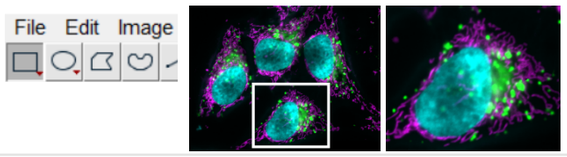
Image > Transform > Rotate 90 Degrees...
Image > Transform > Flip...
Rotation by multiples of 90 Degree can preserve data. Anything else interpolates values:



Save images in smaller format and size:
Plugins > Example > Downsample...
Avoid upsampling

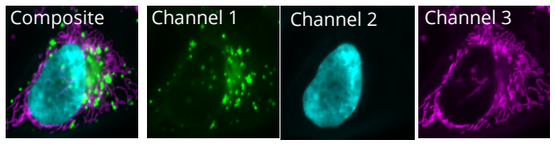
Cropping

Draw selection in toolbar
Image > Crop... - ***Ctrl + Shift + X***
Image > Duplicate... - Ctrl + Shift + D



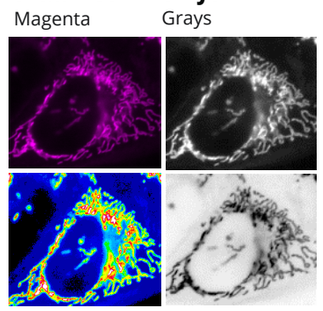
Color

To adjust color, split composite image to separate channels:
Image > Color > Split

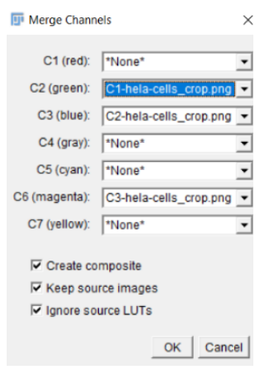


Change LUTs to preset color/colorscheme:
Image > Lookup Tables...
[pick color e.g grays, magenta, 16 colors]

Invert for better visibility:
Edit > Invert... - ***Ctrl + Shift + I***



Merge channels with predefined LUT:
Image > Color > Merge Channels...



- ❑ *tick 'ignore source LUTs' to merge with custom LUTs*
- ❑ *tick 'make composite' to retain bit-depth*

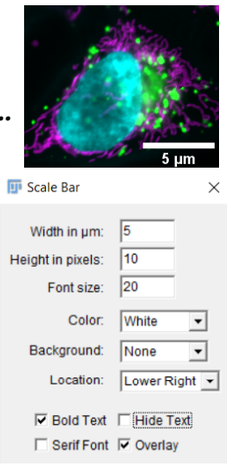
TIP: In composite images, switch between channels for viewing details:
Image > Color > Channels Tool

Create your own LUTs:
Image > Color > Edit LUT...

TIP: Test color blind safety: Most common form color blindness: Deuteranopia. Test general visibility in grayscale or "Monochromacy".
Required: RGB image.
Image > Color > Stack to RGB...
Image > Color > Simulate Color Blindness...

Annotate

Set scale:
Analyze > Set Scale...
Analyze > Tools > Scale Bar...



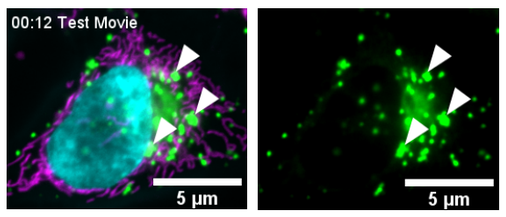
- ❑ Width: 1/5/10 steps
- ❑ Color: highest contrast to image, if necessary add background
- ❑ Overlay: separate layer from image (lost in png)
- ❑ Hide text and add later if resolution of image too low

TIP: check visibility of scale bar & annotation, alternatively add thin bar and annotate later

Add text and time labels:
Image > Stacks > Label...

Image > Annotate > Arrow...
arrows can be moved/rotated/shortened

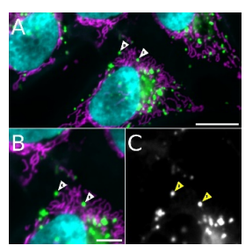
TIP: Overlay may be turned on/off:
Image > Overlay > Hide/Show Overlay



TIP manage multiple ROIs and Labels:
Analyze > Tools > ROI Manager

Layout

Create multi-image panels with annotations for publications:
FigureJ https://imagej.net/FigureJ
ScientiFig
https://grr.gred-clermont.fr/labmirouse/software/



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Example image used: HeLa Cells - ***File > Open Samples > HeLa Cells***