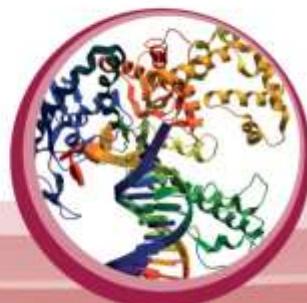
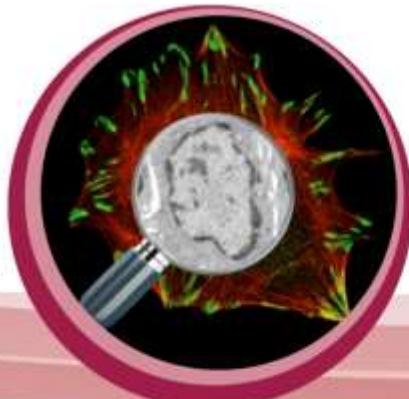


Using ImageJ/Fiji for microscope image processing and analysis

MMC 2023

Dr Kees Straatman

Advanced Imaging Facility
Core Biotechnology Services
College of Life Sciences
University of Leicester
krs5@le.ac.uk



Programme

- 1.30 pm
 - Introduction
 - Basics
 - Plugins
- Short break
 - Macros
- 4.30 pm end

Image analysis

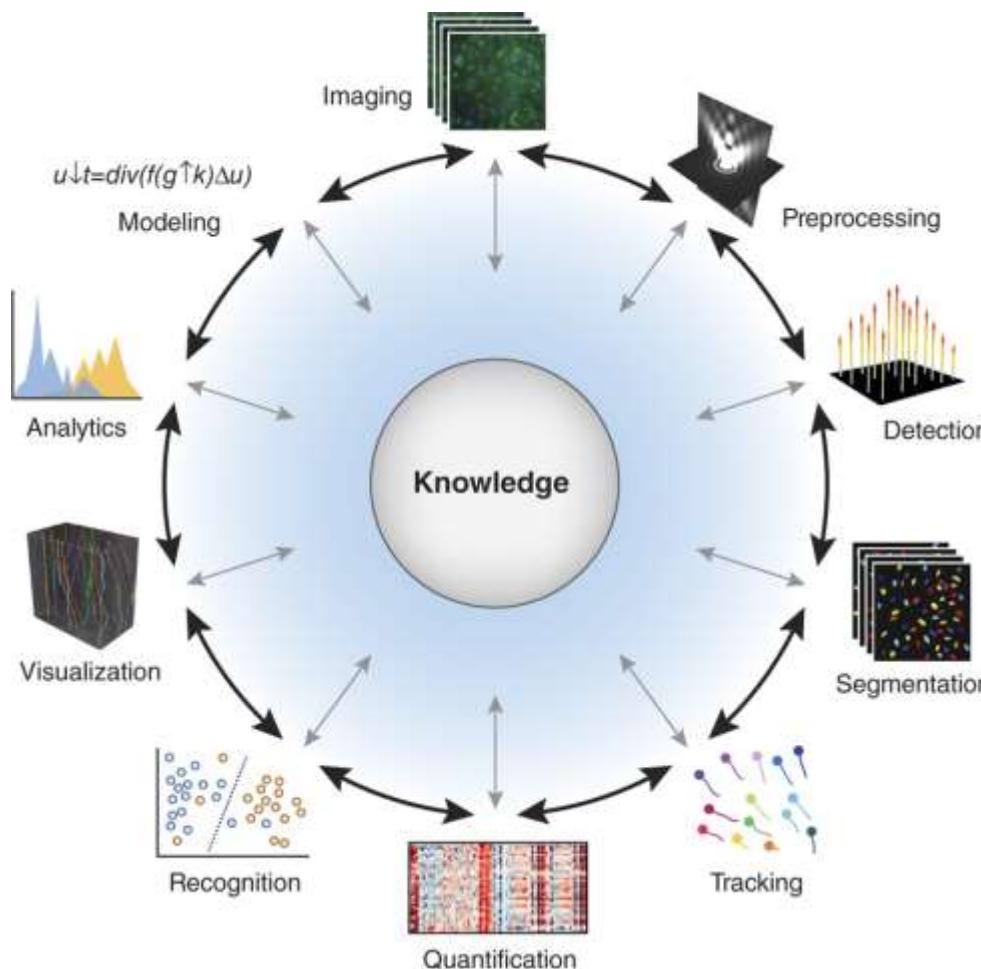
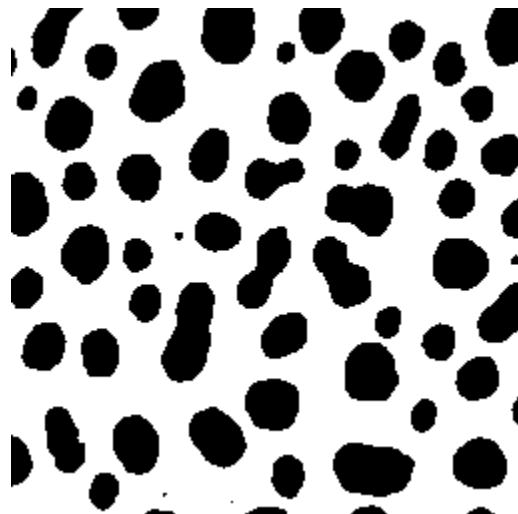
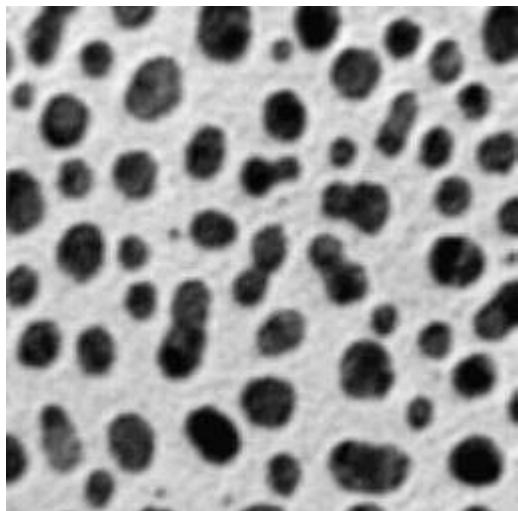


Image analysis

Image analysis is the extraction of meaningful information from digital images using digital image processing techniques.

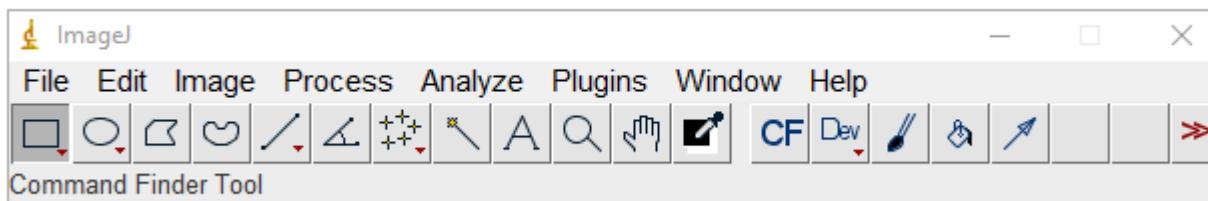


Results									
Label	Area	Min	Max	Perim.	Cke	AR	Round	Solidity	
1	blobs-1.gft	265	256	255	62.164	0.926	1.163	0.887	0.938
2	blobs-1.gft	81	256	255	32.305	0.971	1.204	0.630	0.926
3	blobs-1.gft	278	256	255	63.941	0.854	1.389	0.720	0.919
4	blobs-1.gft	231	256	255	55.698	0.836	1.141	0.877	0.941
5	blobs-1.gft	501	256	255	82.669	0.921	1.075	0.930	0.948
6	blobs-1.gft	669	256	255	98.320	0.858	1.337	0.748	0.852
7	blobs-1.gft	99	256	255	35.793	0.971	1.369	0.788	0.943
8	blobs-1.gft	226	256	255	55.698	0.924	1.142	0.876	0.940
9	blobs-1.gft	448	256	255	78.426	0.815	1.207	0.828	0.947
10	blobs-1.gft	401	256	255	87.497	0.626	2.498	0.400	0.890
11	blobs-1.gft	620	256	255	84.669	0.912	1.162	0.846	0.952
12	blobs-1.gft	425	256	255	77.941	0.891	1.451	0.609	0.953
13	blobs-1.gft	271	256	255	62.164	0.981	1.348	0.742	0.972
14	blobs-1.gft	159	256	255	46.284	0.933	1.225	0.816	0.930
15	blobs-1.gft	412	256	255	75.598	0.896	1.106	0.904	0.945
16	blobs-1.gft	426	256	255	87.497	0.699	1.010	0.553	0.869
17	blobs-1.gft	260	256	255	59.941	0.969	1.153	0.887	0.932
18	blobs-1.gft	289	256	255	62.770	0.922	1.131	0.884	0.930
19	blobs-1.gft	678	256	255	104.326	0.780	1.493	0.675	0.909
20	blobs-1.gft	361	256	255	76.184	0.921	1.222	0.818	0.945
21	blobs-1.gft	545	256	255	86.912	0.907	1.225	0.817	0.949
22	blobs-1.gft	610	256	255	112.669	0.656	2.748	0.364	0.848
23	blobs-1.gft	14	256	255	12.485	1.050	1.020	0.861	0.933
24	blobs-1.gft	641	256	255	106.569	0.709	1.936	0.517	0.804
25	blobs-1.gft	185	256	255	61.113	0.938	1.147	0.872	0.929

Introduction

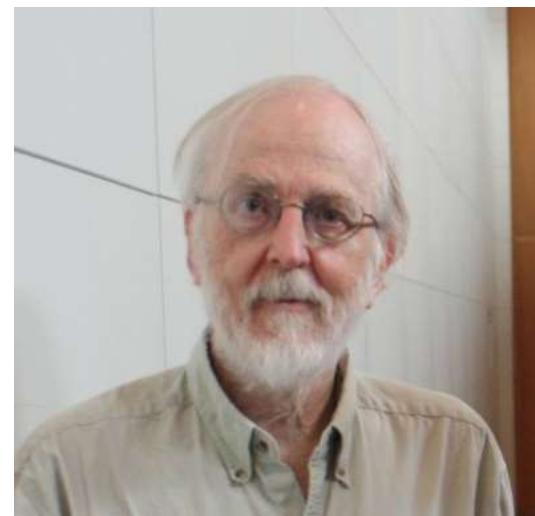
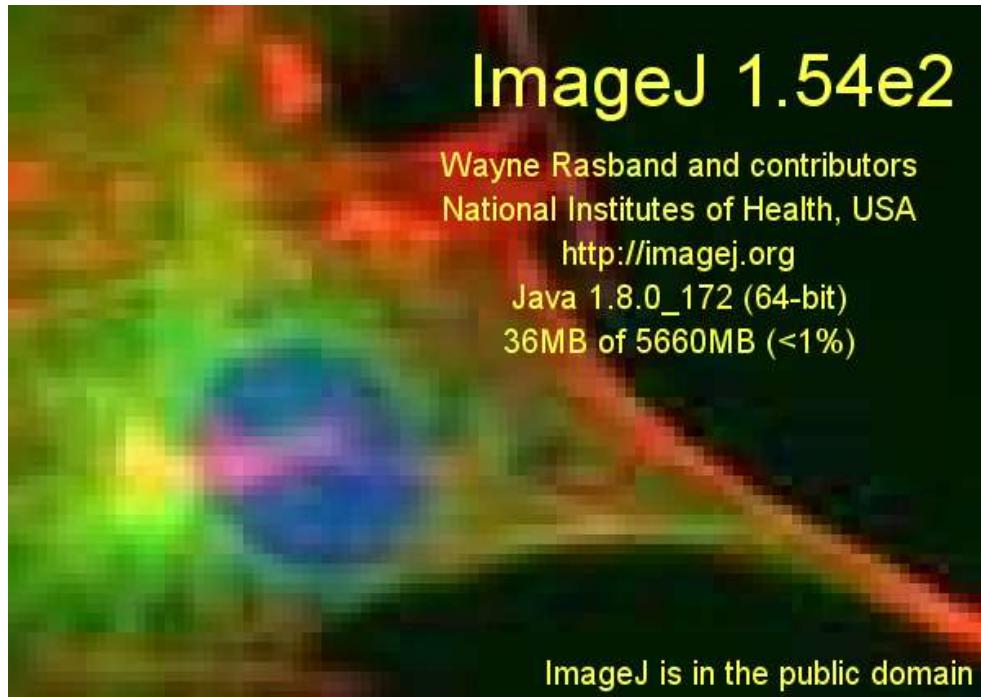
ImageJ

- It provides a Graphical User Interface (GUI) with extensive image analysis commands. Floating windows.



- Platform independent:
Mac OSX/ Linux/Windows: 64 bit version
- Open source (free!!).
- Active community → large number of solutions available or help to find solutions.

ImageJ



Wayne Rasband; Special volunteer
at National Institutes of Health

ImageJ

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Download

Platform Independent

To install ImageJ on a computer with Java pre-installed, or to upgrade to the latest full distribution (including macros, plugins and LUTs), download the [ZIP archive](#) (6MB) and extract the ImageJ directory. Use the *Help>Update ImageJ* command to upgrade to newer versions.

Mac OS X

Download [ImageJ bundled with Java 1.8.0_172](#) (may need to work around Path Randomization). Instructions. With M1 (ARM) Macs, download [ImageJ bundled with Zulu OpenJDK 13.0.6](#).

Linux

Download [ImageJ bundled with Java 1.8.0_172](#) (82MB). [Instructions](#).

Windows

Download [ImageJ bundled with 64-bit Java 1.8.0_172](#) (70MB). [Instructions](#).

Documentation

Tiago Ferreira's comprehensive [ImageJ User Guide](#) is available as an 8MB PDF document and as a ZIP archive. The online [JavaDoc API documentation](#) is also available as a ZIP archive.

Source Code

The ImageJ [Java source](#) consists of 132,000 lines of code in 348 files. It is available [online](#) and as [zip](#) archives.

Example Images

31 downloadable sample images and stacks are available in ImageJ's *File>Open Samples* submenu. These images, and more, are also available as a [8.2MB zip archive](#).

You can also browse the ImageJ download directory at imagej.nih.gov/ij/download/. Newer ImageJ distributions are available at <http://wsr.imagej.net/distros/>. Refer to the [release notes](#) for a list of new features and bug fixes.

ImageJ

Windows: download in ZIP format. Unpack in a folder you have access to, like 'My Documents' or a D-drive

You need access to the ImageJ installation folder:

- To install extensions (plugins/macros/scripts)
- To update ImageJ
- To change some of the *Options* settings

If you get the error:

'Unable to update the file "ImageJ.cfg C:\ Program Files\ImageJ\ImageJ.cfg (Access denied)'

ImageJ has been installed in a folder you don't have write permission for.

ImageJ

The functionality of ImageJ can be extended by using:

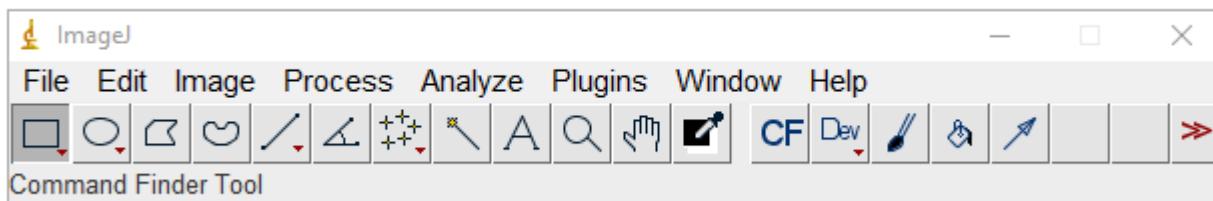
- Plugins: programmes that add functionality to ImageJ;
 - Written in Java.
- Macros: automations, add tools or document your analysis in ImageJ.
 - ImageJ's own scripting language.
- Scripts: JavaScript, BeanShell and Python.

ImageJ

NIH Image (for MAC) from before 1993 developed by Wayne Rasband. Windows version was called Scion Image.

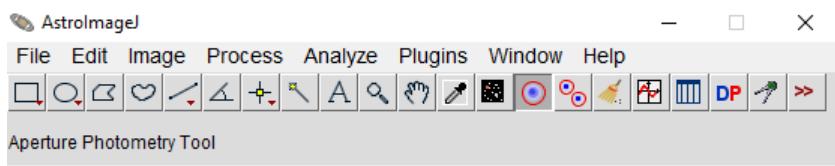
ImageJ since 1997

Schneider, CA, Rasband, WS, Eliceiri, KW (2012) NIH Image to ImageJ: 25 years of image analysis. *Nature Methods* 9, 671-675.



ImageJ

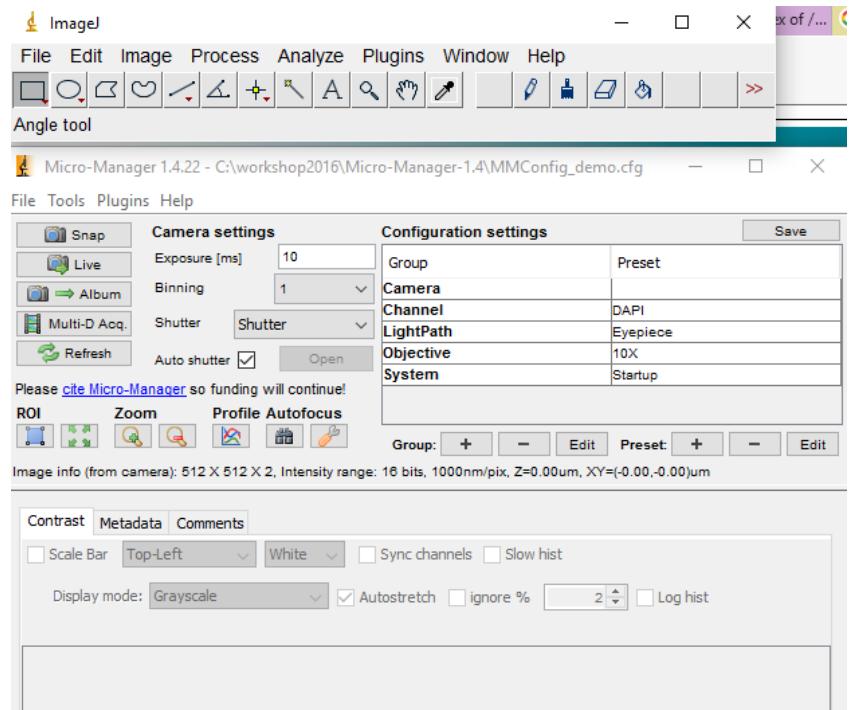
AstrolimageJ : Astronomy



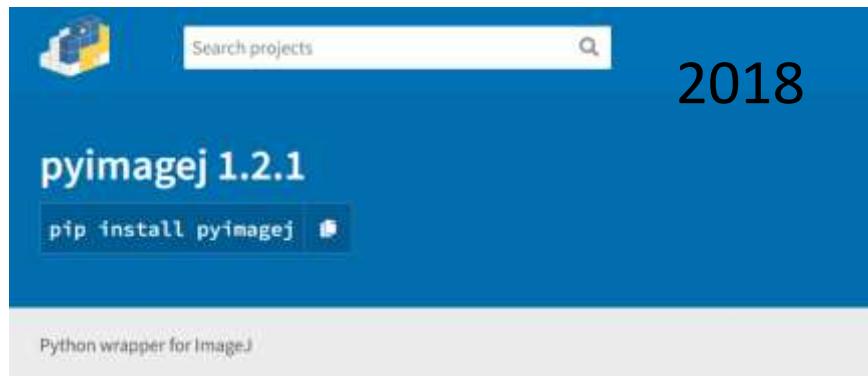
SalsaJ: Astronomy in the classroom



μManager: Image acquisition



ImageJ



Python wrapper for ImageJ

Navigation

Project description

Relevant history

Project description

PylImageJ: Python wrap

<https://ij.imjoy.io/>



ImagePy – 2017(?)

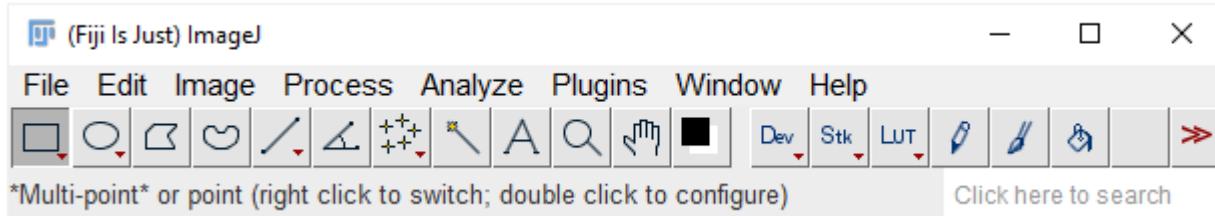


2020: ImageJ running in a web browser

Fiji

Fiji since 2007 (Fiji Is Just ImageJ); ImageJ distribution includes > 300+ plugins focussed on biological image analysis.

Schindelin et al. (2012) Fiji: an open-source platform for biological-image analysis. Nature Methods 9, 676–682.



ImageJ2 initiated in 2009

Rueden et al. (2017) ImageJ2: ImageJ for the next generation of scientific image data, BMC Bioinformatics 18:529-555



Fiji

Fiji = Fiji Is Just ImageJ

<http://fiji.sc/>

Fiji is an ImageJ distribution focussed on the life sciences:

- Image registration
- Image segmentation
- 3D reconstruction
- 3D Visualization

Includes 300+ plugins but hardly any macros and no macro tools.

Over the years Fiji has been built on top of the new developed
ImageJ2

Fiji

Download include:

ImageJ2 core

ImageJ1 as plugin (for backwards compatibility)

Many plugins (+ only few macros and scripts)

Java (JDK + JRE)

Java3D

Like ImageJ, Fiji is downloaded as ZIP and “extract”, not installed.

Caution: "Program Files" not recommended!

If you are installing Fiji on Windows, we strongly recommend that you store your Fiji.app directory somewhere in your user space (e.g., "C:\Users\[your name]\Fiji.app") rather than in "C:\Program Files" or other system-wide directory. If you move Fiji.app to such a directory, modern versions of Windows will deny Fiji write permission to its own directory structure, preventing it from being able to update.

Fiji

Contributors:

- Albert Cardona
- Barry DeZonia
- Benjamin Schmid
- Cornelius Sicker
- Curtis Rueden
- Dan White
- Erwin Frise
- Gabriel Landini
- Greg Jefferis
- Ignacio Arganda-Carreras
- Jean-Yves Tinevez
- Johannes Schindelin
- Kota Miura
- Larry Lindsey
- Mark Longair
- Mark Hiner
- Pavel Tomancak
- Rob Bryson-Richardson
- Stephan Preibisch
- Stephan Saalfeld
- Verena Kaynig
- Etc, etc

Fiji development during Hackathons.
Often in collaboration with third parties like:

- Knime
- Omero
- ImgLib2
- Cellprofiler
- Scifio

And/or on specific topics like:

- Metadata
- Performance and generality
- Multiview reconstruction

<https://imagej.net/people/>

<https://imagej.net/events/hackathons>

References

← → ⌂ ⌂ <https://imagej.nih.gov/ij/index.html>

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ImageJ

Image Processing and Analysis in

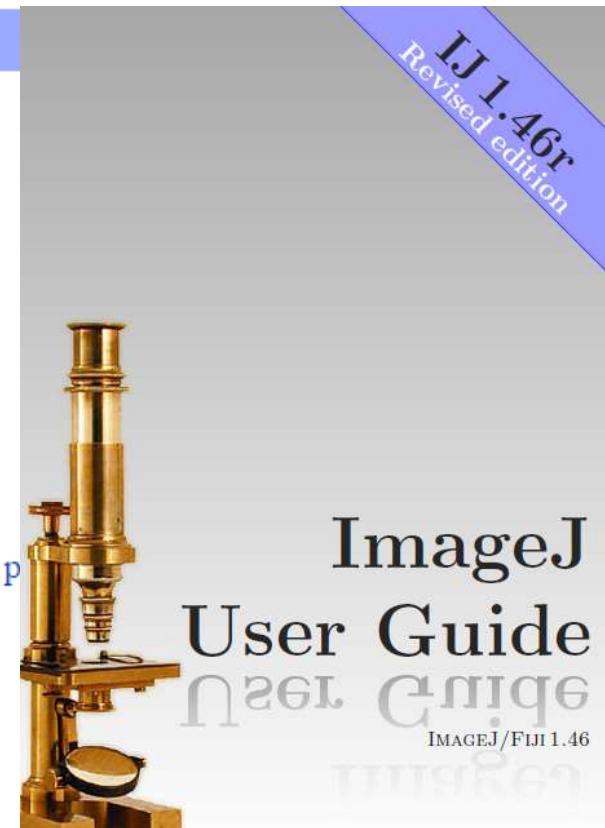
- [Features](#)
- [News](#)
- [Documentation](#)
- [Download](#)
- [Run ImageJ in Browser](#)
- [Plugins](#)
- [Developer Resources](#)
- [Mailing List](#)
- [Links](#)

Support is available on the main forum. [Disclaimer](#)

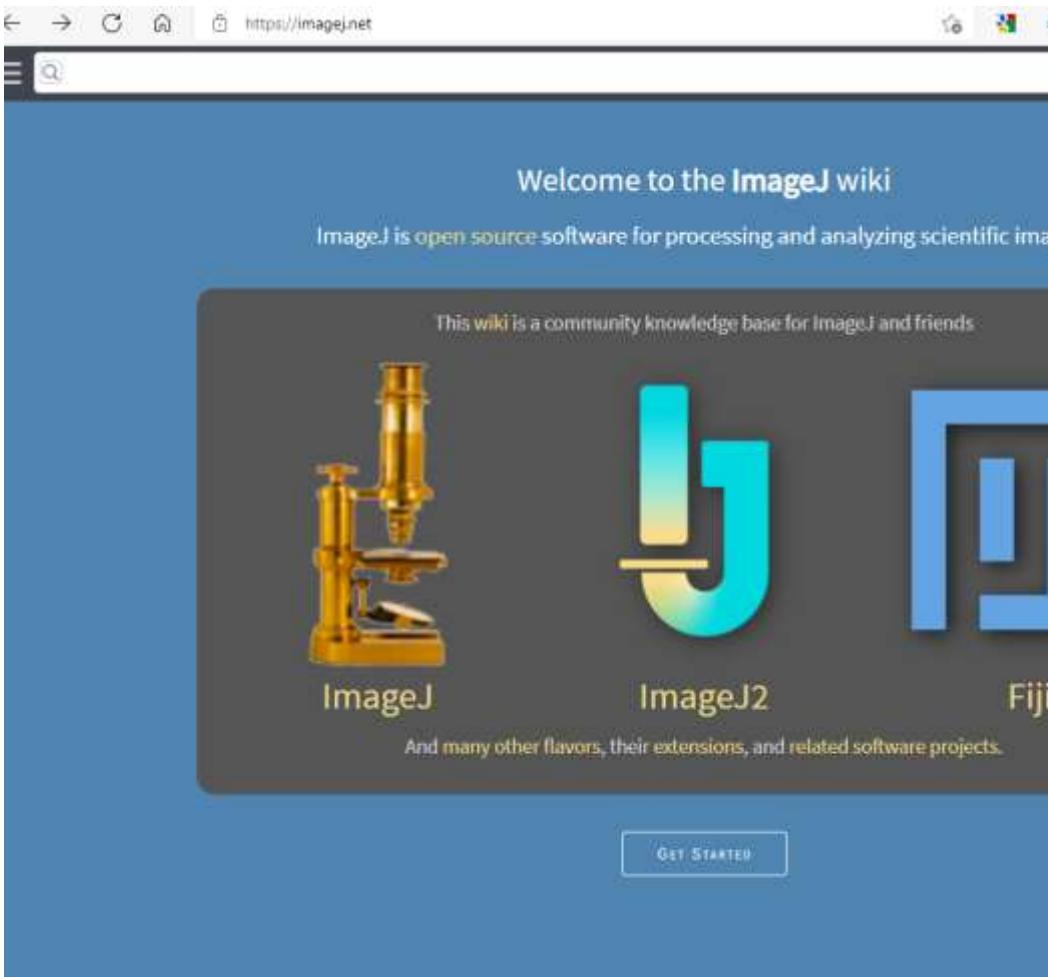
[home](#) | [docs](#) | [download](#) | [plugins](#) | [resources](#) | [list](#) | [links](#)

Documentation

- [Introduction](#)
- [Basic Concepts](#)
- [Installation](#)
- [*ImageJ User Guide* \(download PDF\)](#)
 - [User Interface and Tools](#)
 - [Menu Commands](#)
 - [Extending ImageJ](#)
 - [Keyboard Shortcuts](#)
- [Tutorials and Examples](#)
- [Image.sc Forum](#)
- [ImageJ Documentation Wiki](#)
- [Image Processing with ImageJ \(ebook or pdf\)](#)
- [ImageJ on Wikipedia](#)
- [Frequently Asked Questions](#)
- [Macro Language \(download PDF\)](#)
- [Complete Release Notes \(744K\)](#)



References



Welcome to the **ImageJ** wiki

ImageJ is open source software for processing and analyzing scientific images.

This [wiki](#) is a community knowledge base for ImageJ and friends

ImageJ **ImageJ2** **Fiji**

And many other flavors, their extensions, and related software projects.

GET STARTED

Start with the [ImageJ Tutorial Notebooks!](#)

And be sure to read over the [Development](#) pages.

Tutorials on this site

- [Analyze FRAP movies with a Jython script](#)
- [How to apply a common operation to a complete directory](#)
- [How to call a plugin with a range of parameters](#)
- [Correcting drift in FRAP experiments](#)
- [Downsample](#)
- [Edit LUT As Text](#)
- [Gabor Filter script](#)
- [Generate and exploit Kymographs](#)
- [The Hue Game](#)
- [ImageJ2 Python Scripts](#)
- [Linux command line tutorial](#)
- [Mamed](#)
- [Multithreaded Image Processing in Clojure](#)
- [Multithreaded Image Processing in JavaScript](#)
- [Open an image](#)
- [A Tutorial for using OpenCL in ImageJ](#)
- [Plasma Cloud](#)
- [RGB to CMYK](#)
- [Segmentation evaluation after border thinning - Script](#)
- [Segmentation evaluation metrics - Script](#)
- [Image Processing School Pilsen 2009 - Segmentation](#)
- [Stitch and Align a sequence of grid images Tutorial](#)
- [Super Sloppy Surface Reconstruction](#)

To add your tutorial to this list, place the page in the `/tutorials` folder.

See also

- [Presentations about ImageJ](#)
- [Fiji YouTube channel](#)
- [ImageJ videos and Fiji videos on YouTube](#)
- [Lecture BioImage Analysis 2020 videos on YouTube by Robert Haase](#)

<https://imagej.net/tutorials/index>

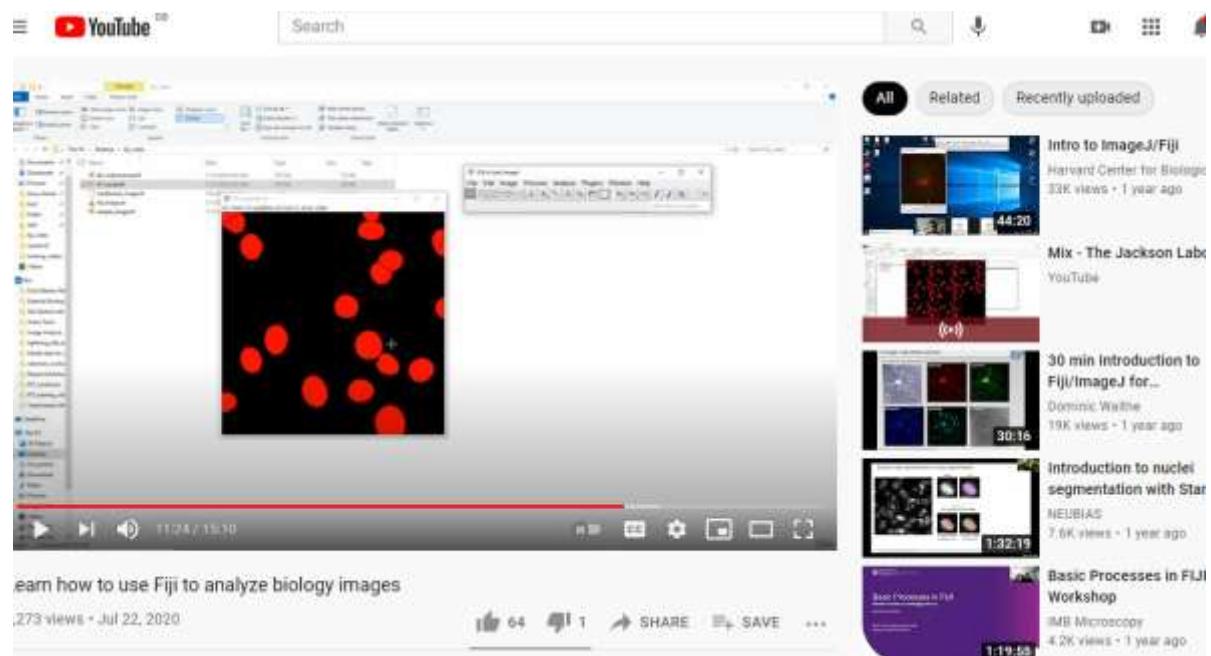
References

Analyzing fluorescence microscopy images with ImageJ

Peter Bankhead
Queen's University Belfast

May 2014

This work is made available in the hope it will help a few more people develop an interest in image analysis. If you have any comments, corrections or suggestions, please contact me at p.bankhead[at]qub.ac.uk, so that it might one day get better.



YouTube

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Harvard Center for Biological
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Mix - The Jackson Lab
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Only online version 2016 at
[https://petebankhead.gitbooks.io/
imagej-intro/content/](https://petebankhead.gitbooks.io/imagej-intro/content/)

References

NIH NIH LISTSERV

Subscriber's Corner Email Lists Log In

 IMAGEJ Home Page

IMAGEJ@LIST.NIH.GOV

Latest Messages

Re: problem with scalebar in recent daily builds - more info	Commer, Michael <[log in to unmask]>	Thu, 22 Jun 2023 01:47:30 +0000	
Re: ArrayFindMaxima doesn't ignore NaN values	Herbie <[log in to unmask]>	Thu, 22 Jun 2023 00:03:50 +0200	
Re: ArrayFindMaxima doesn't ignore NaN values	Wayne Rasband <[log in to unmask]>	Wed, 21 Jun 2023 15:53:59 +0400	

IMAGEJ

Archive-Month: none This is a mailing list for users and developers of ImageJ, a public domain, Java-based and extensible image processing program for Linux, Macintosh and Windows. The

View Latest Messages

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image.sc

all categories ► fiji ► all ► any status ▾ Latest Top Categories

Topic Replies Views Activity

Illumination correction of fluorescence image using the BaSiC plugin in Fiji 2 34 11m

■ Usage & Issues fiji, plugin

Random ROI Multi Measure in batch omero execution 1 50 1h

■ Image Analysis fiji, macro, omero, batch-processing

Help with "Analyze particles" of challenging particles 5 40 2h

■ Image Analysis imagej, fiji, segmentation, analyze-particles, thresholding

QuPath update to 0.4.3/ Image registration 5 140 3h

fiji/warpy - Qupath project not opening in fiji

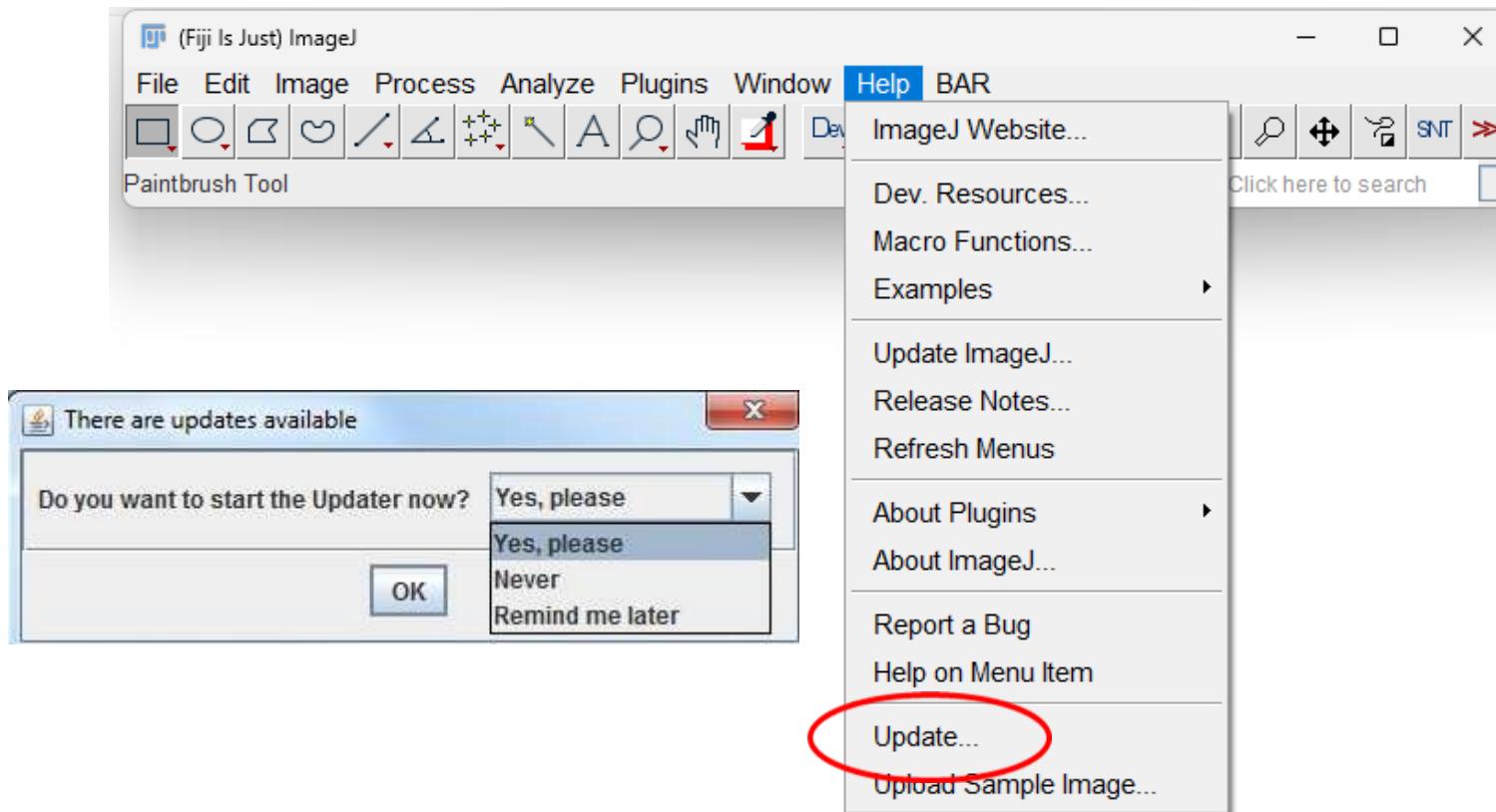
■ Image Analysis fiji, qupath, warpy, vs200

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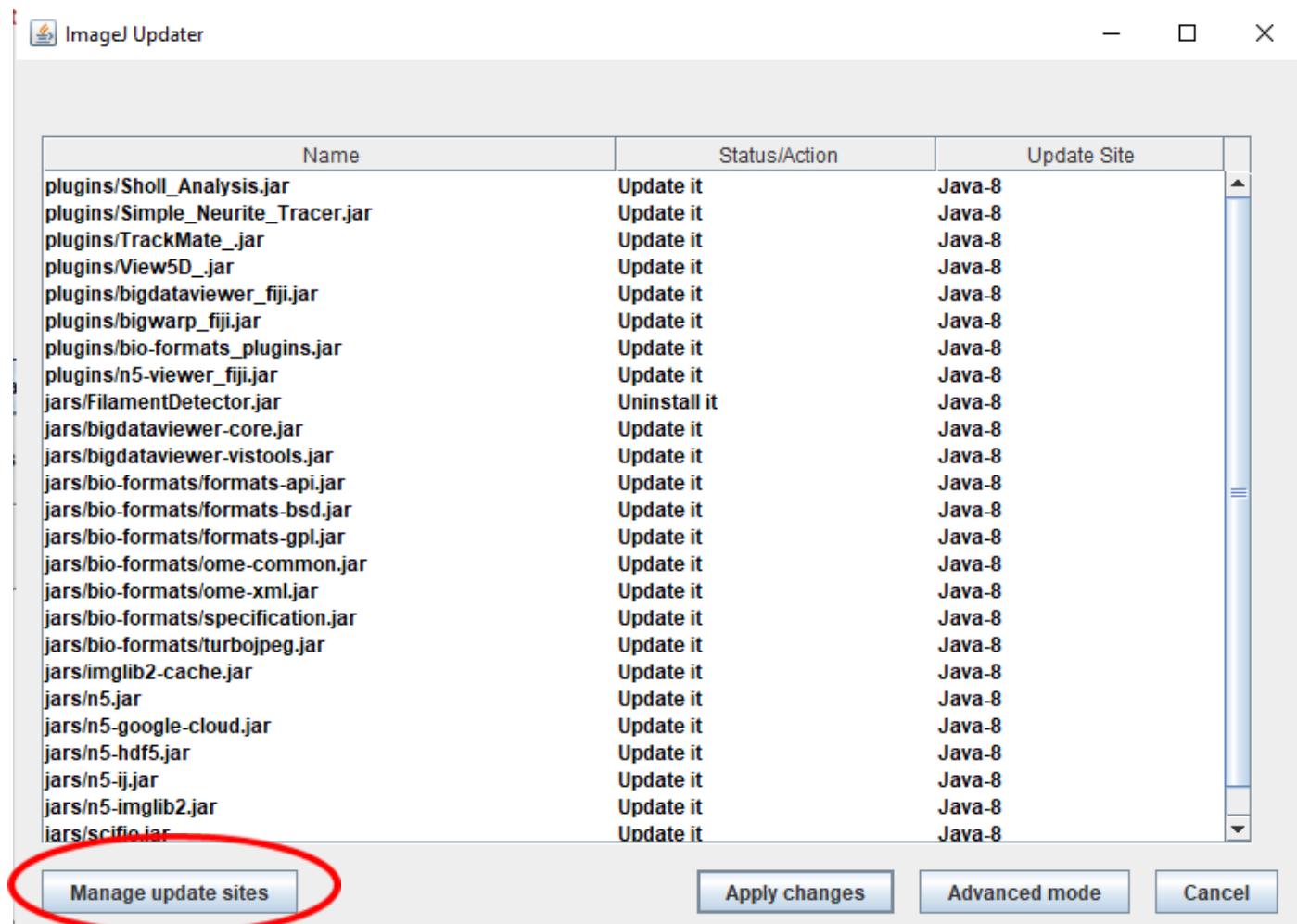
Fiji is an image processing package — a “batteries-included” distribution of ImageJ, bundling many plugins which facilitate scientific image analysis.

Update Fiji



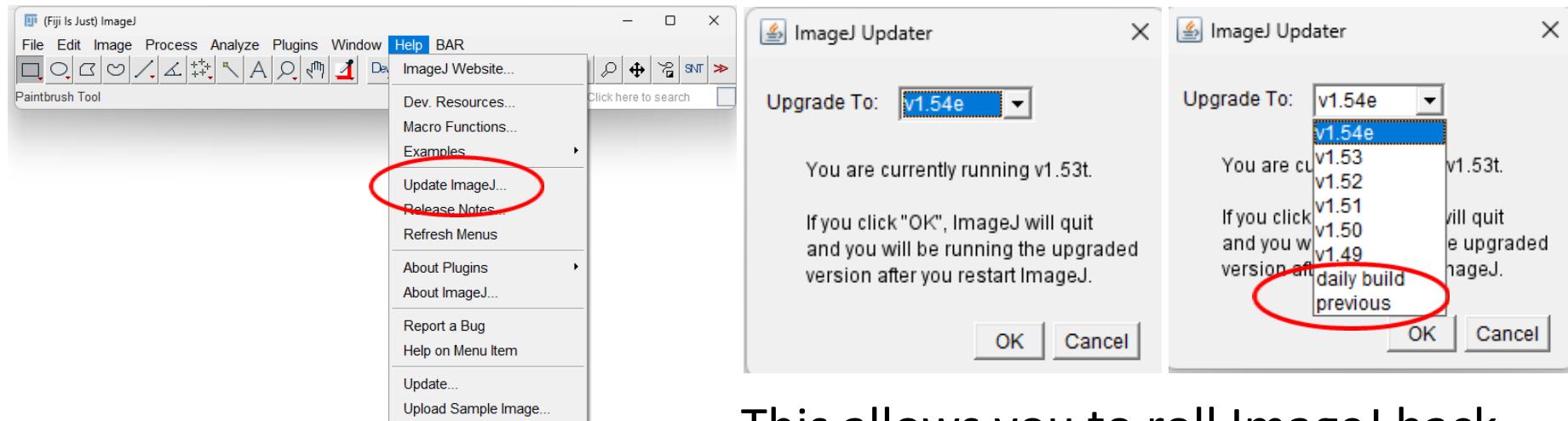
It is not possible to roll back to a previous version.

Update Fiji



Update ImageJ

ImageJ is continuously updated. This is done by Wayne Rasband. If you update Fiji, only major releases of ImageJ are included.



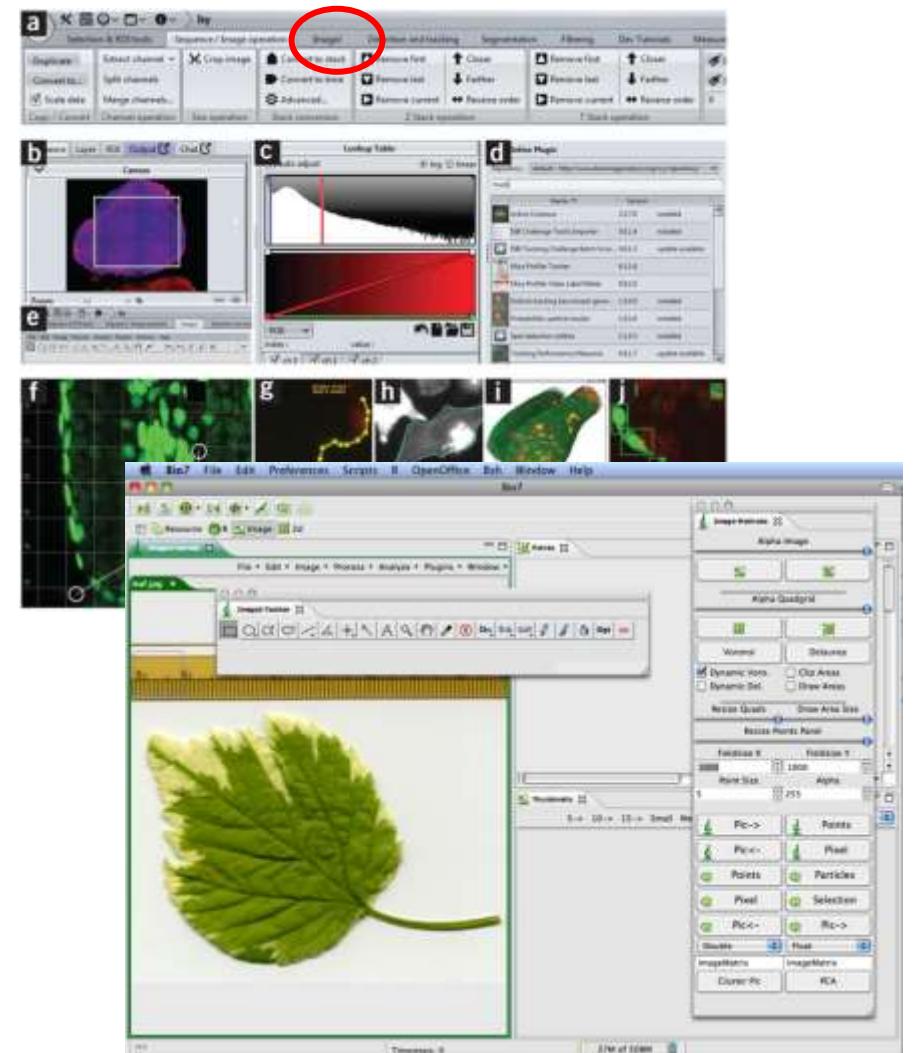
This allows you to roll ImageJ back to a previous version.

Be aware: ImageJ closes after update!

Other projects

Icy; created by the Quantitative Image Analysis Unit at Institut Pasteur

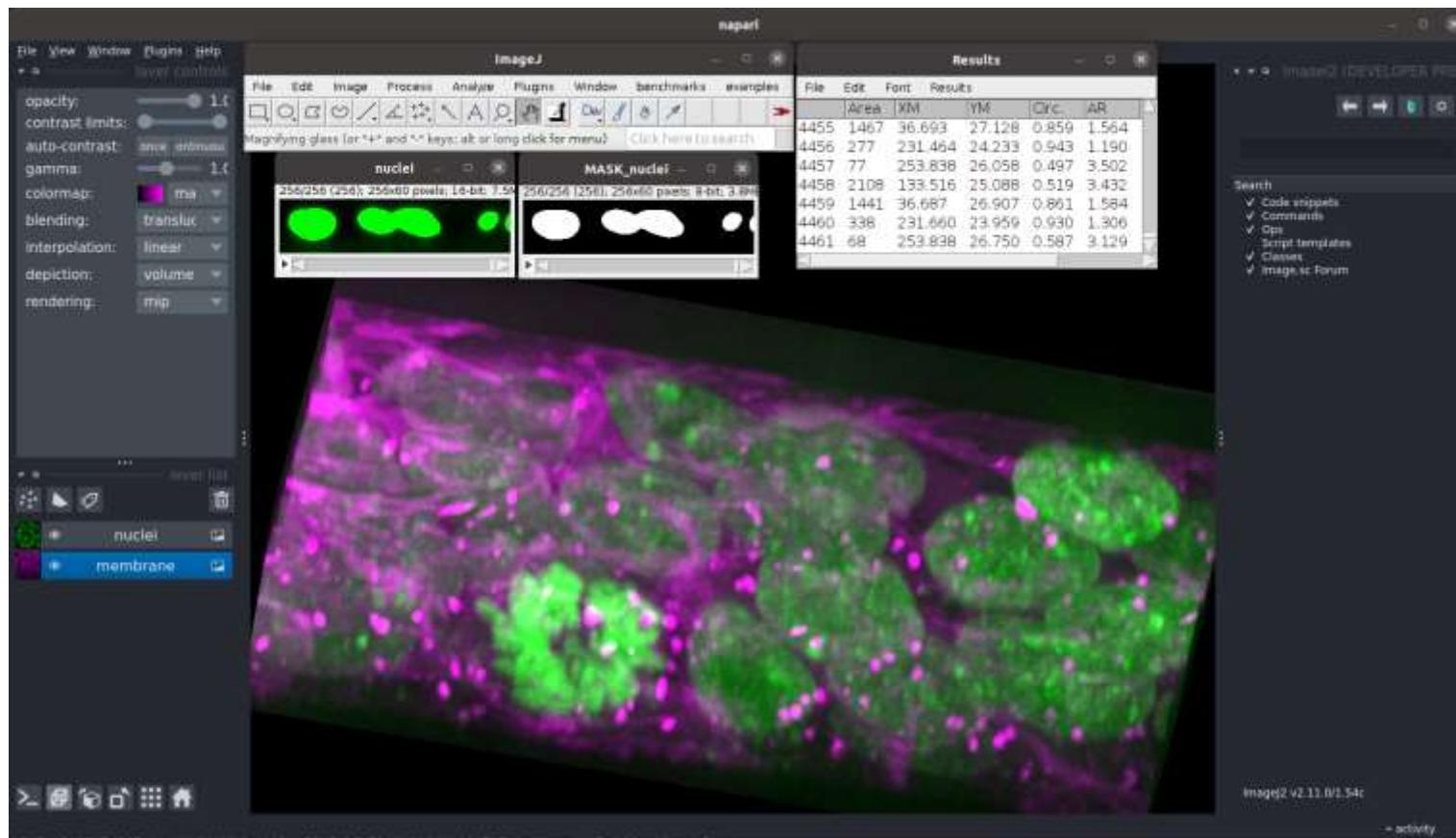
(<http://www.bioimageanalysis.org/>).



Bio7: ImageJ integrated in an ecological modelling environment including Statistical analysis (R).

Other projects

napari-imagej



Other projects

<https://forum.image.sc>

 All Topics	 AGAVE	 AICSIImageIO	 Aydin	 BIAFLOWS	 BIII
 BiofilmQ	 Bio-Formats	 BiolImageIO	 BoneJ	 BrainGlobe	 Cellpose
 CellProfiler	 CLIJ	 CytoMAP	 Cytomine	 DAIM	 DeepLabCut
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 MCMICRO	 MIA	 MIB	 μManager	 MoBIE	 MorphoGraphX
 MorphoNet	 napari	 NEUBIAS	 NFDI4BIOIMAGE	 OME	 OMERO
 OmeSliCC	 OpenIRIS	 OpenSPIM	 Orbit	 Piximi	 PYME
 Python-Microscope	 QUAREP-LiMi	 QuPath	 SCIFIO	 scikit-image	 sciview
 SR-Tesseler	 StarDist	 starfish	 TissUUmaps	 vedo	 webKnossos
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Mango

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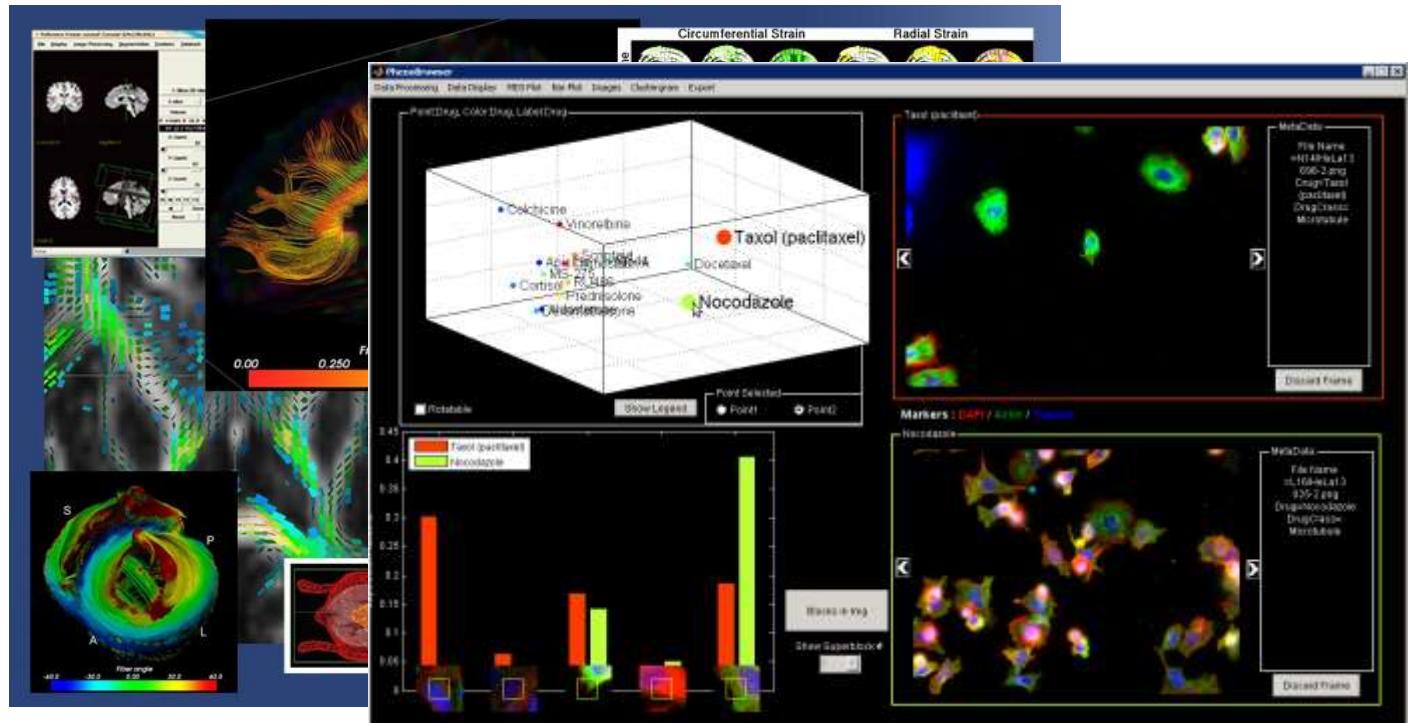
PickCells

Vaa3D

IMMI - Rapidminer 5 Image Processing Extension

MIPAV (Medical Image Processing, Analysis, and Visualization)

Etc



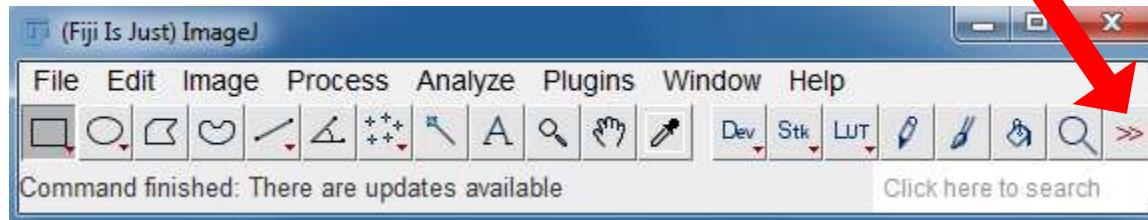
Some basics

User interface

Menu Bar →

Tool Bar →

Status Bar →



Menu items are written in Java. Use of menu items will be written in *Italic* in this workshop.

Tools: mostly macros that are added to the toolbar.

Tools are saved in subfolder\macros\tools.

Toolsets: a toolset can contain up to 12 macro tools, along with any number of ordinary macros.

Toolsets are saved in subfolder\macros\toolsets.

Toolsets

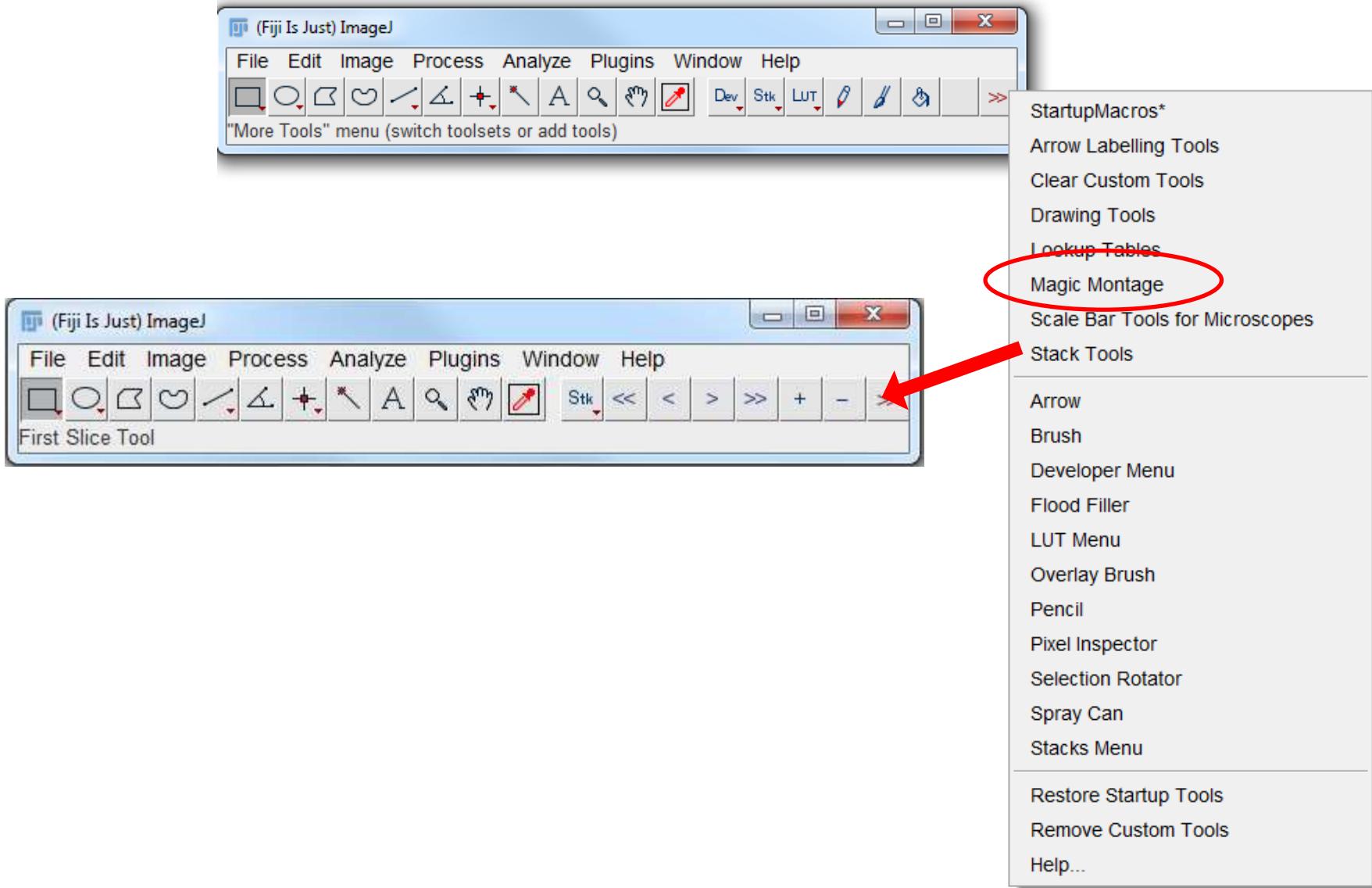
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Developer Resources

- Macro Language ([download PDF](#))
-
- Built-in Macro Functions
- Introduction to Macro Programming
- Macro Programming in ImageJ (Kota Miura) New
- Macros
 - Macros Directory
 - Examples
 - [Macro Tools](#)
 - [Toolsets](#)
- Scripting
 - JavaScript Examples
 - BeanShell (examples) New
 - [Python \(examples\)](#) New

Name	Last modified
Angiogenesis Analyze..>	2013-12-03 13:35
BioVoxxel Toolbox.txt	2013-04-28 21:35
Built-in Tools.txt	2012-07-14 14:44
Developer Tools.txt	2010-07-29 14:18
Dot Blot Analyzer.txt	2013-12-03 23:02
Drawing Tools.txt	2012-07-14 14:43
Dynamic_Stats_and_Hi..>	2010-07-29 14:18
Example Icons.txt	2013-12-03 12:16
Image, Stack and Tim..>	2013-12-03 23:06
Intersects_Method.txt	2014-07-15 13:01
LSM Transmission Deb..>	2010-07-29 14:18
Lookup Tables.txt	2013-12-03 13:45
Luts Macros and Tool..>	2013-12-15 18:22
MBF Tools.txt	2010-07-29 14:18
MagicMontage.pdf	2018-11-29 15:41
Magic Montage.txt	2018-11-29 15:41
Magic Montage2.txt	2010-07-29 14:18
Multi-Channels Image..>	2018-10-16 21:06
Original Drawing Too..>	2012-07-13 22:16
Plugins.txt	2008-10-21 12:25
Protein Array Analyz..>	2018-10-16 21:06
ROI Manager Tools.txt	2013-12-15 18:48
Rename and Save ROI ..>	2010-07-29 14:18

Toolsets



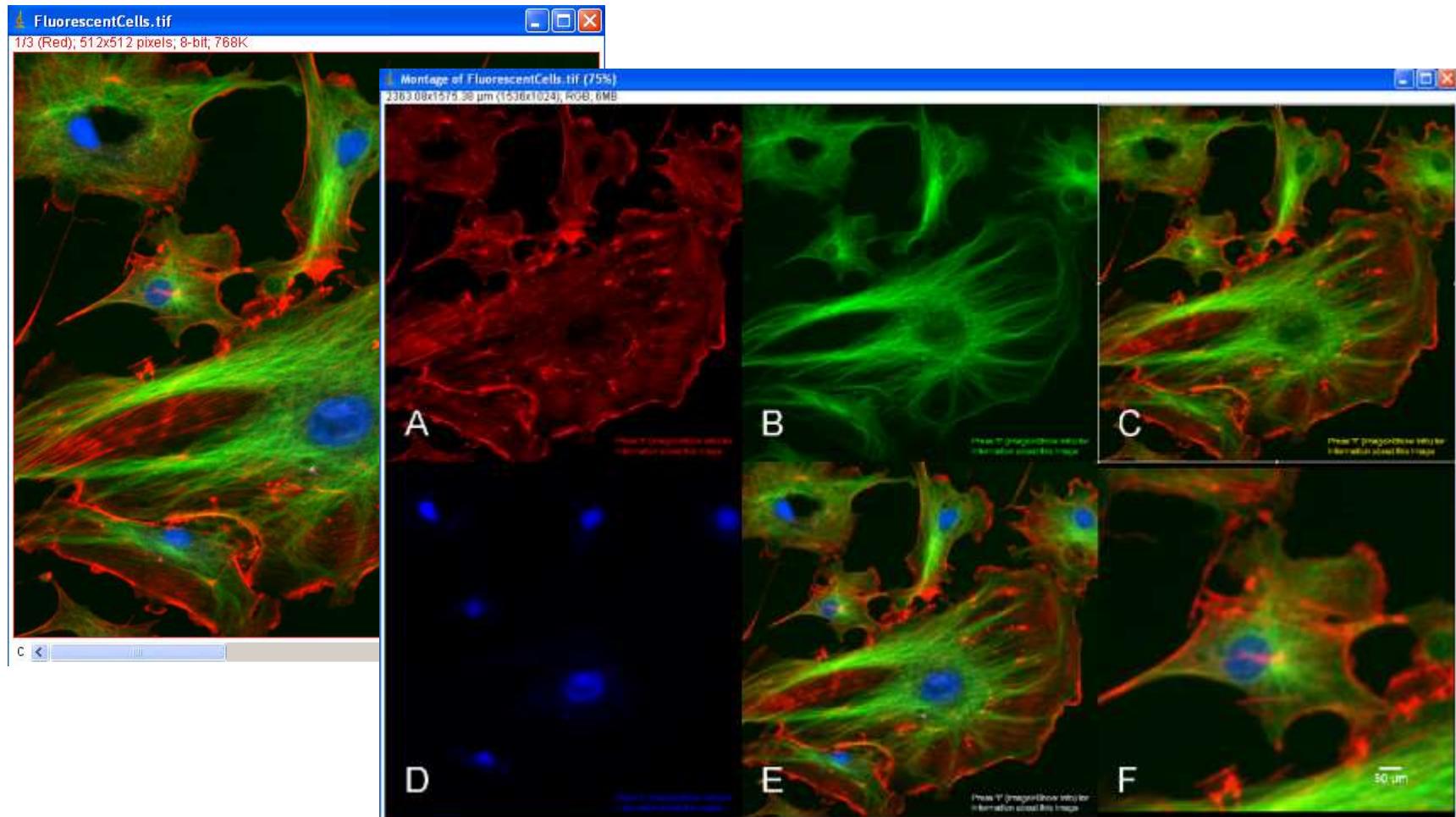
The image shows two windows of the Fiji software interface. The top window is titled '(Fiji Is Just) ImageJ' and displays a toolbar with various tools like selection, drawing, and measurement. A dropdown menu labeled "'More Tools' menu (switch toolsets or add tools)" is open, showing a list of toolsets. The bottom window is also titled '(Fiji Is Just) ImageJ' and shows a similar toolbar. A red arrow points from the 'Stack Tools' item in the dropdown menu to the '-' button in the toolbar of the bottom window, indicating how to switch toolsets.

- StartupMacros*
- Arrow Labelling Tools
- Clear Custom Tools
- Drawing Tools
- Lookup Tables
- Magic Montage**
- Scale Bar Tools for Microscopes
- Stack Tools

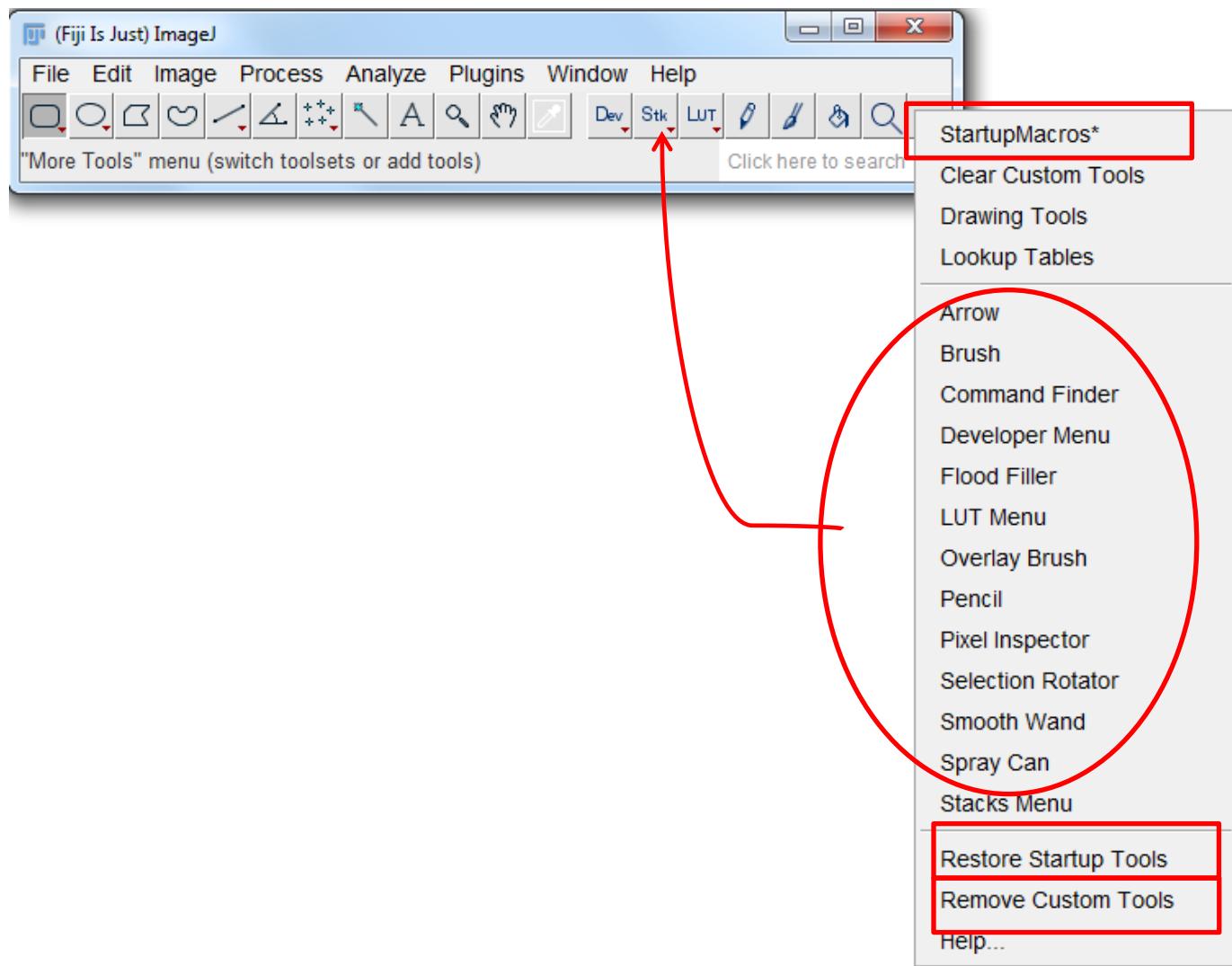
- Arrow
- Brush
- Developer Menu
- Flood Filler
- LUT Menu
- Overlay Brush
- Pencil
- Pixel Inspector
- Selection Rotator
- Spray Can
- Stacks Menu

- Restore Startup Tools
- Remove Custom Tools
- Help...

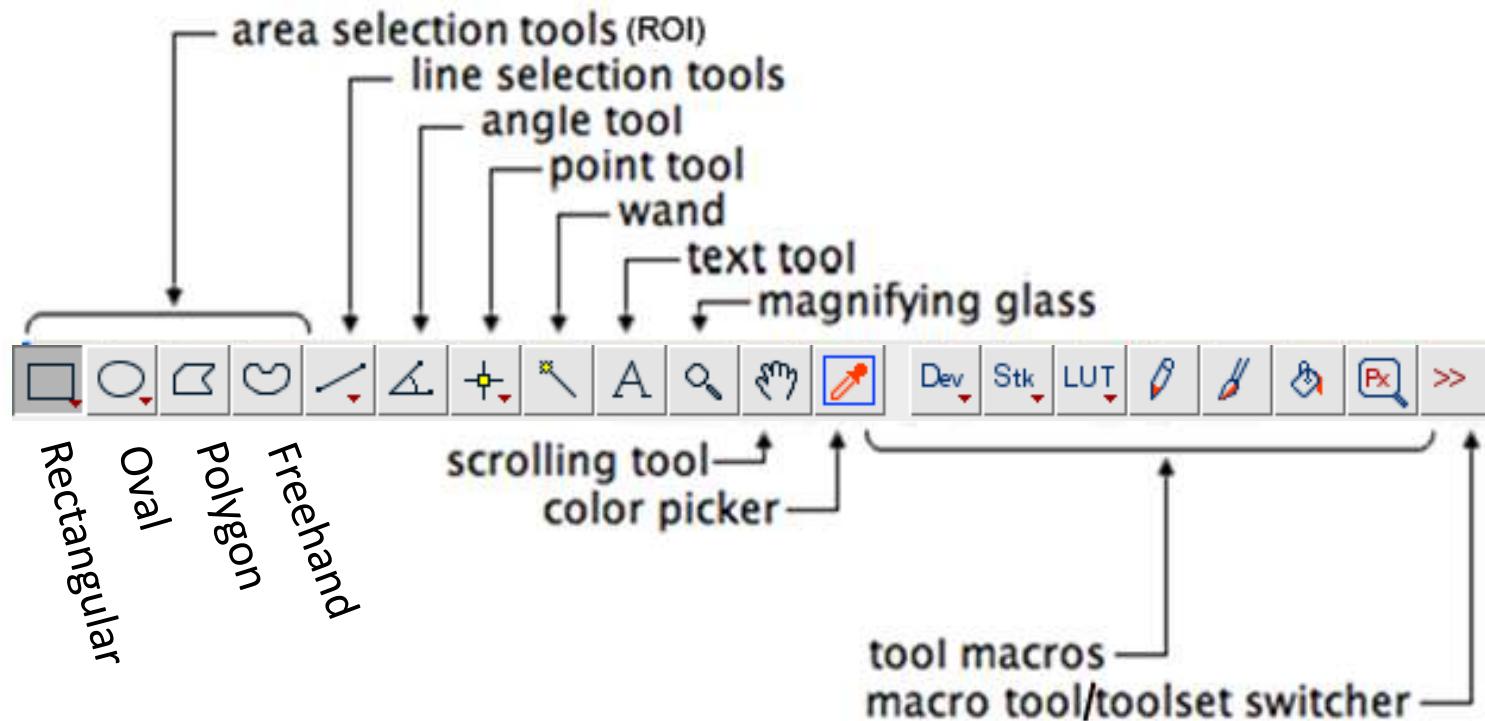
Toolsets



Toolsets



Toolbar



Right mouse click on  : access to different tools

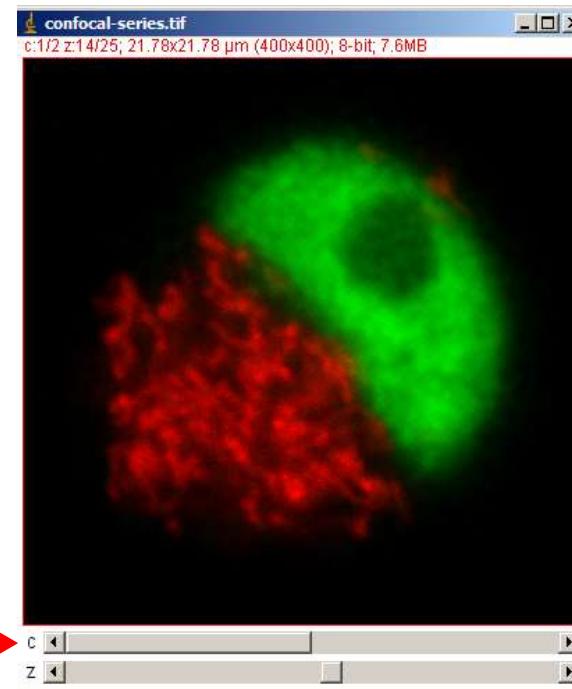
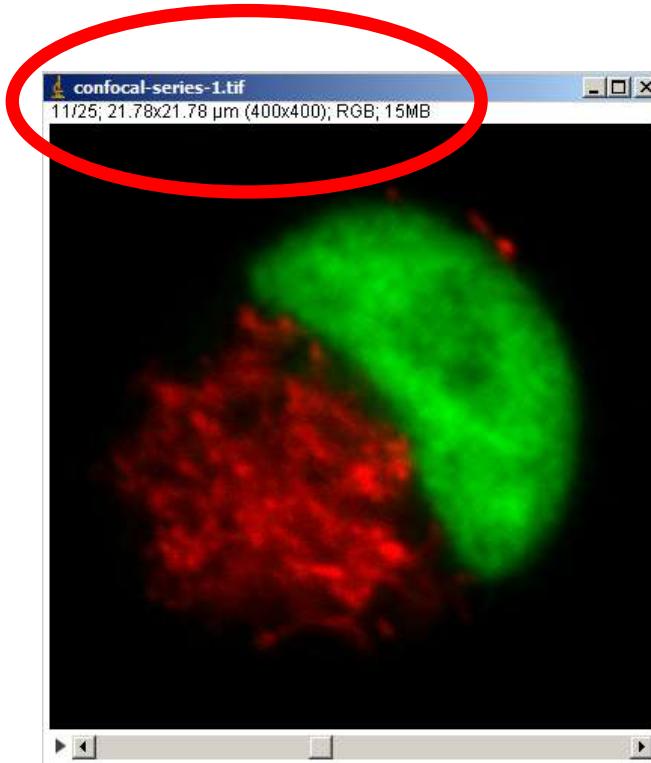
Double mouse click on some tools: access to options menu

Image types

- 8 bit: 256 grey levels
- 16 bit: 65536 grey levels
- 32 bit: floating point scale; can have any value
- RGB 24 bit: 8 bit Red + 8 bit Green + 8 bit Blue
- RGB 32 bit: as RGB 24 bit + additional eight bits coding for alpha channel transparency information
- Color composites
 - Each channel can be 8, 16 or 32 bit
 - Color channels kept separate
 - More than 3 channels can be merged

Image types

Composite image



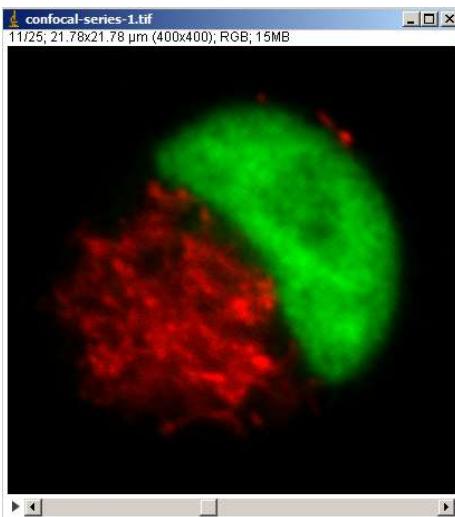
Advantage:

- Each channel is separate
- More than 3 channels possible
- Can work on 1 channel while all are visible

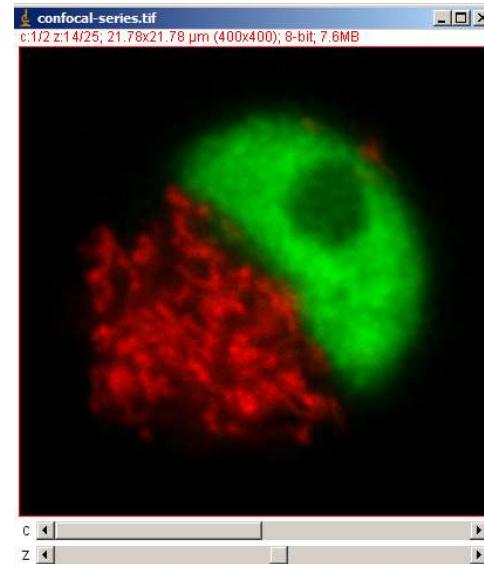
Image types

- Single image (x,y)
- Stack: one channel (x,y,z; x,y,t)
- Hyperstack: multidimensional extended stack (x, y, z, c and/or t)

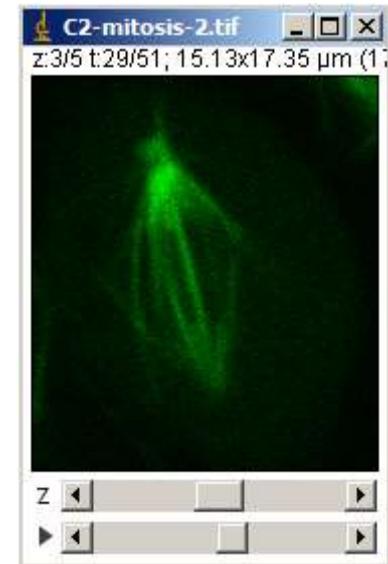
Stack



Hyperstack



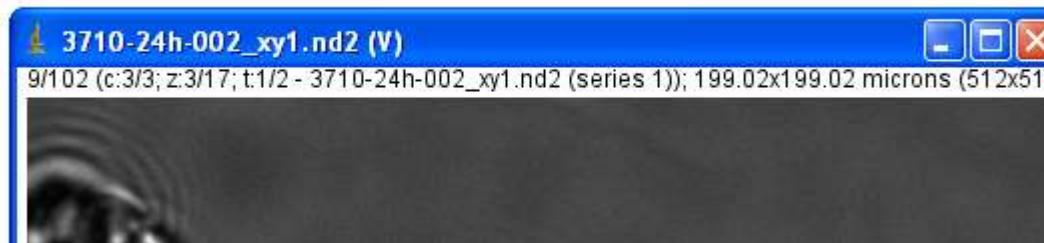
= Composite image



≠ Composite image

Image types

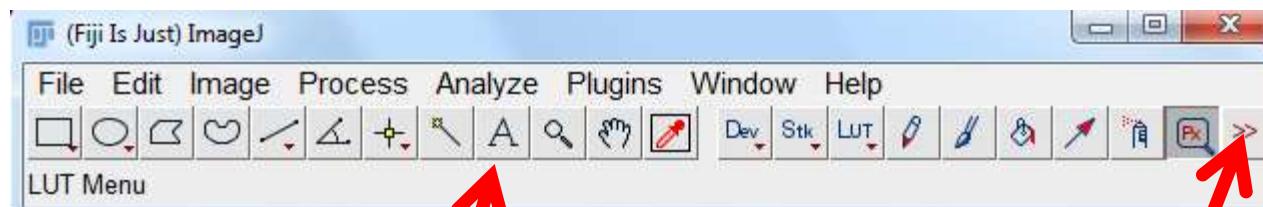
- Single image (x,y)
- Stack: one channel (x,y,z; x,y,t)
- Hyperstack: multidimensional extended stack (x, y, z, c and/or t)
- Virtual Stack: read-only disk-resident (virtual) stack (be aware, changes won't be saved; overlays won't be loaded!). This is useful for large image series.



File Formats

Default image support for BMP, DICOM, GIF, JPEG, PGM, PNG, TIFF and raw data.

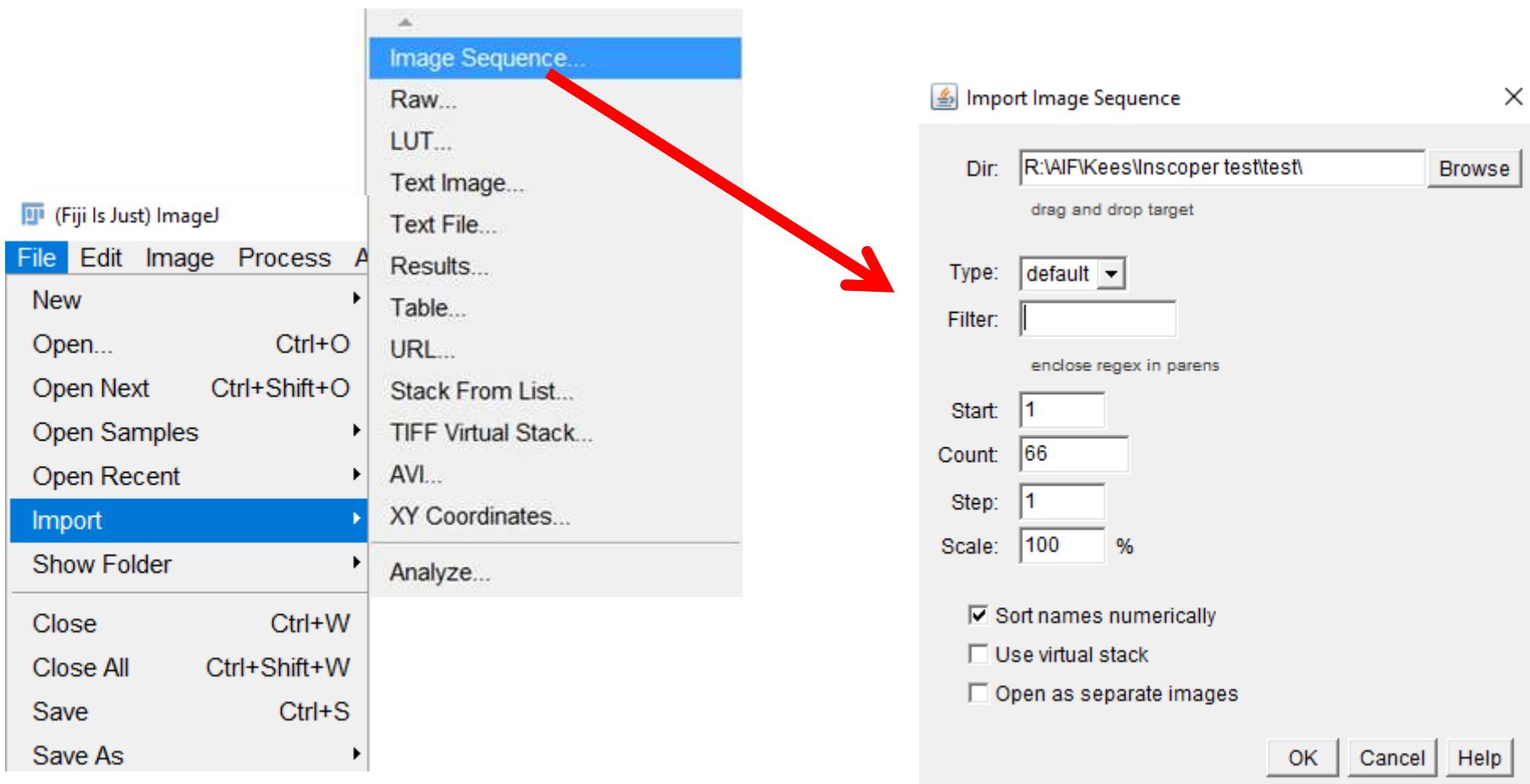
Open images using *File > Open*, drag-and-drop or *File > Import* (for image series):



Drag-and-drop;
Opens file.

Drag-and-drop;
opens TIF stacks
as virtual stack.

File Formats



File Formats

Default image support for BMP, DICOM, GIF, JPEG, PGM, PNG, TIFF and raw data.

Using **Bio-Formats library**. This plugin will allow you to open most microscope file formats directly via *File > Open* or *File> Import > Bio-Formats*.

However, it is possible that you have to open an image via Plugins > Bio-Formats

- reads >160 formats
- writes 14 formats

File Formats

Bio-Formats Import Options X

Stack viewing

View stack with: **Hyperstack** ▼

Stack order: **XYCZT** ▼

Metadata viewing

Display metadata
 Display OME-XML metadata
 Display ROIs

ROIs Import Mode: **ROI manager** ▼

Dataset organization

Group files with similar names
 Open files individually
 Swap dimensions
 Open all series
 Concatenate series when compatible
 Stitch tiles

Memory management

Use virtual stack
 Specify range for each series
 Crop on import

Split into separate windows

Split channels
 Split focal planes
 Split timepoints

Information

View stack with - The type of image viewer to use when displaying the dataset.

Possible choices are:

- **Metadata only** - Display no pixels, only metadata.
- **Standard ImageJ** - This option is deprecated (i.e. intended for use by old macros only). Please use *Hyperstack* instead.
- **Hyperstack** - Display the pixels in ImageJ's built-in 5D viewer.
- **Data Browser** - Display the pixels in the multidimensional Data Browser viewer. The Data Browser has some additional features on top of the normal ImageJ hyperstack.
- **Image5D** - Display the pixels in Joachim Walter's Image5D viewer. Requires the Image5D plugin.
- **View5D** - Display the pixels in Rainer Heintzmann's View5D

Option Information OK Cancel

File Formats

Latest development update; Latest stable update is automatic

Bio-Formats library

Active	Name	URL	Host	Directory on Host
<input checked="" type="checkbox"/>	ImageJ	http://update.imagej.net/		
<input checked="" type="checkbox"/>	Fiji	http://fiji.sc/update/		
<input type="checkbox"/>	3D ImageJ Suite	http://sites.imagej.net/Tboudier/		
<input type="checkbox"/>	Angiogenesis	http://sites.imagej.net/Angiogenesis/		
<input type="checkbox"/>	Archipelago	http://sites.imagej.net/Lindsey/		
<input type="checkbox"/>	BigDataViewer	http://sites.imagej.net/Pietzsch/		
<input checked="" type="checkbox"/>	Bio-Formats	http://sites.imagej.net/Bio-Formats/		
<input type="checkbox"/>	Biomedgroup	http://sites.imagej.net/Biomedgroup/		
<input type="checkbox"/>	BioVoxel	http://sites.imagej.net/BioVoxel/		
<input type="checkbox"/>	Cookbook	http://sites.imagej.net/Cookbook/		
<input type="checkbox"/>	CMCI-EMBL	http://sites.imagej.net/Miura/		
<input type="checkbox"/>	CMP-BIA tools	http://sites.imagej.net/CMP-BIA/		
<input type="checkbox"/>	FFMPEG	http://fiji.sc/~schindelin/ffmpeg-plugins/		
<input type="checkbox"/>	IBMP-CNRS	http://www-ibmp.u-strasbg.fr/fijiupdates/		
<input type="checkbox"/>	ImageJ Latex	http://sites.imagej.net/Yul.liuyu/		
<input type="checkbox"/>	IMCF Uni Basel	http://sites.imagej.net/UniBas-IMCF/		
<input type="checkbox"/>	LLTT	http://sites.imagej.net/Alex-krull/		
<input type="checkbox"/>	LOCI	http://sites.imagej.net/LOCI/		
<input type="checkbox"/>	MOSAIC ToolSuite	http://mosaic.mpi-cbg.de/Downloads/update/Fiji		
<input type="checkbox"/>	OpenSPIM	http://openspim.org/update/		
<input type="checkbox"/>	PHANTAST	http://sites.imagej.net/Nicjac/		
<input type="checkbox"/>	PTBIOPI	http://biop.epfl.ch/Fiji-Update/		
<input type="checkbox"/>	ScientificFig	http://sites.imagej.net/Aigouy/		
<input type="checkbox"/>	SCIFIO-dev	http://sites.imagej.net/SCIFIO/		
<input type="checkbox"/>	SLIM Curve	http://sites.imagej.net/SLIM-Curve/		
<input type="checkbox"/>	Stowers	http://research.stowers.org/ImageJplugins/update/		
<input type="checkbox"/>	TrackMate-dev	http://sites.imagej.net/TrackMate-dev/		

[Add my site](#) [Add](#) [Remove](#)

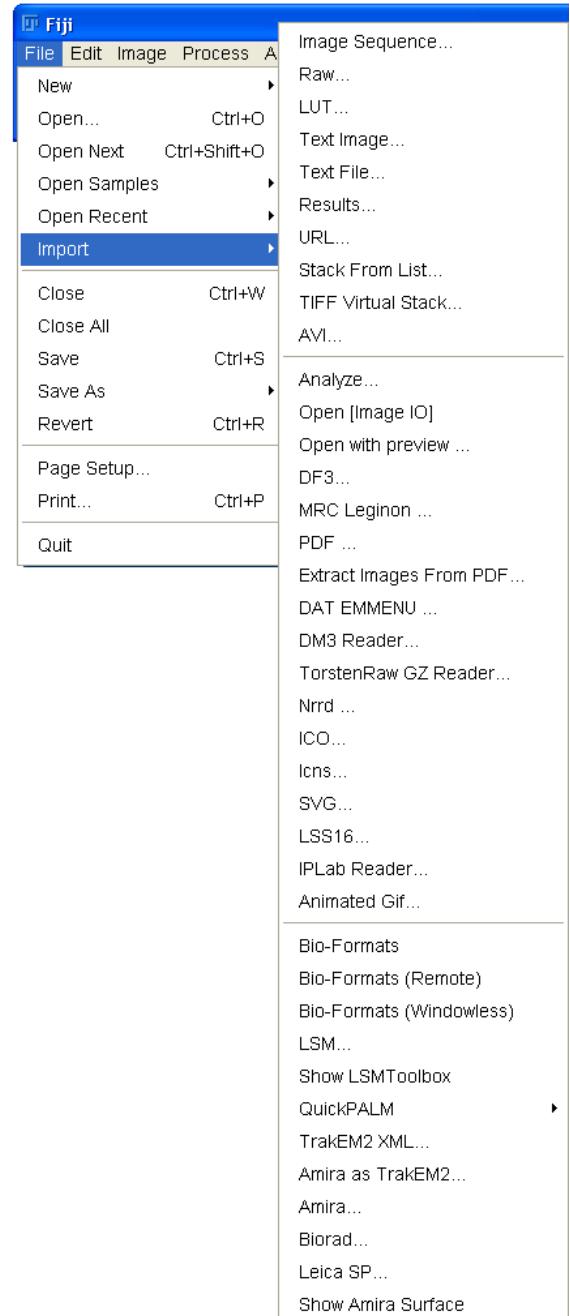
[https://bio-formats.readthedocs.io/en/v6.13.0/
supported-formats.html](https://bio-formats.readthedocs.io/en/v6.13.0/supported-formats.html)

Format	Extensions	Pixels	Metadata	Openness	Presence	Utility	Export	BSD	Multiple Images	Pyramid
3i SlideBook	.sld	▲	▼	▼	▲	▼	✗	✗	✓	✗
3i SlideBook 7	.sldy	▲	▲	▲	▲	▲	✗	✓	✗	✗
Andor Bio-Imaging Division (ABD) TIFF	.tif	▲	▲	■	▼	■	✗	✗	✓	✗
AIM	.aim	■	▼	▼	▼	▼	✗	✗	✗	✗
Alicona 3D	.al3d	▲	▲	▲	▼	■	✗	✗	✗	✗
Amersham Biosciences Gel	.gel	▲	■	■	▼	▼	✗	✗	✗	✗
Amira Mesh	.am, .amiramesh, .grey, .hx, .labels	▲	▼	▼	▼	▼	✗	✗	✗	✗
Amnis FlowSight	.cif	■	■	■	▼	▼	✗	✓	✓	✗

File Formats

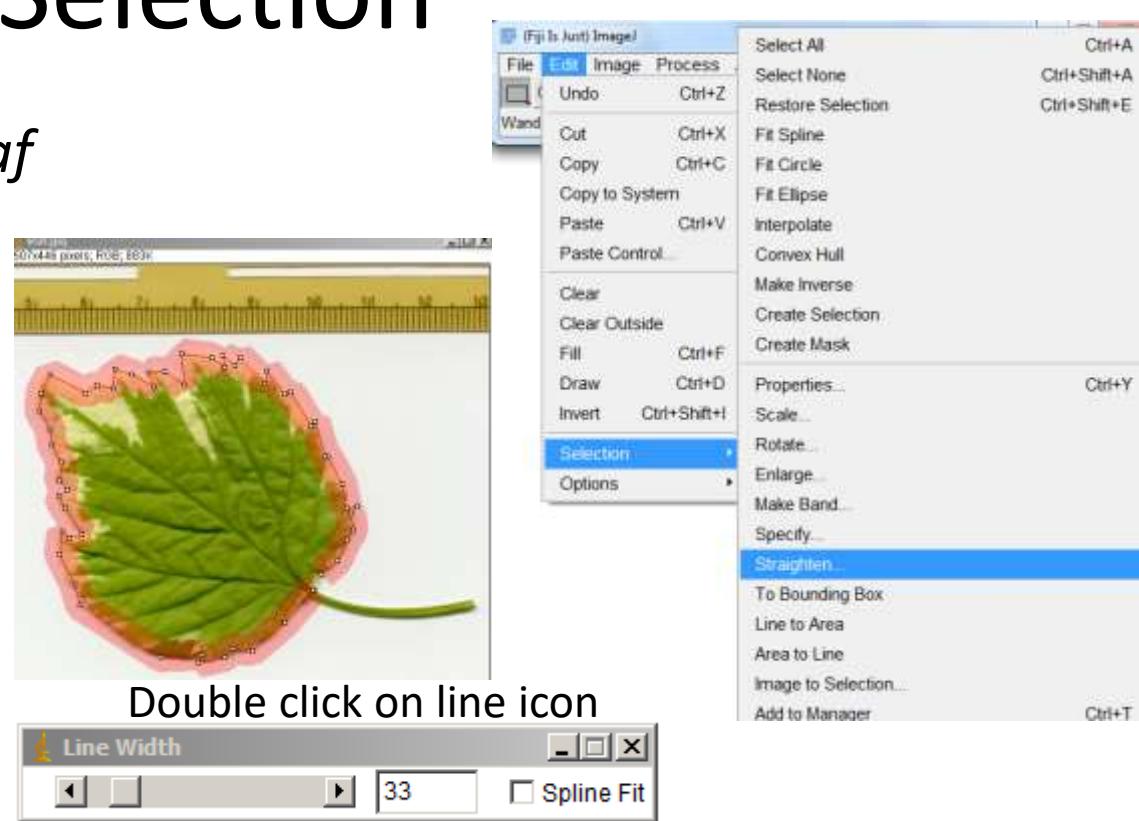
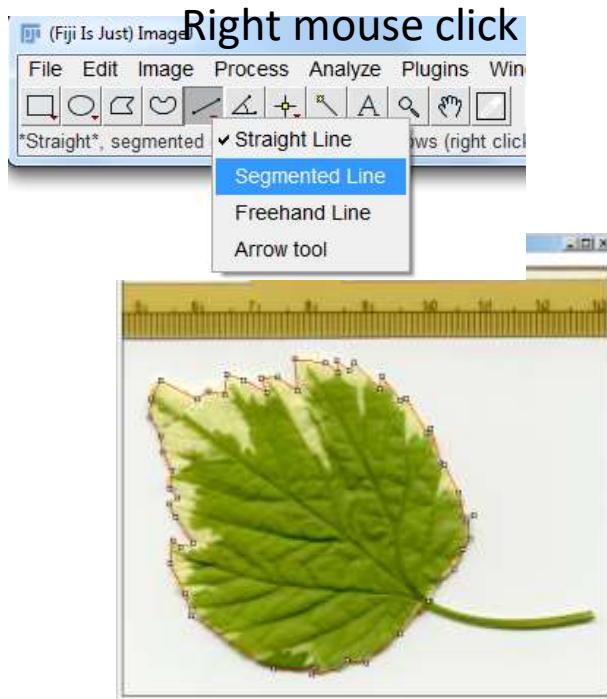
Fiji installs with more file formats supported.

If you miss a format or an image does not open correctly using Bio-Formats plugin there might be a plugin available.



Selection

File > Open Samples > Leaf



Change colour of selection line: *Edit > Options > Colors*

Restore selection line: *Edit > Selection > Restore Selection*



Brightness and Contrast

File > Open samples > Fluorescence Cells

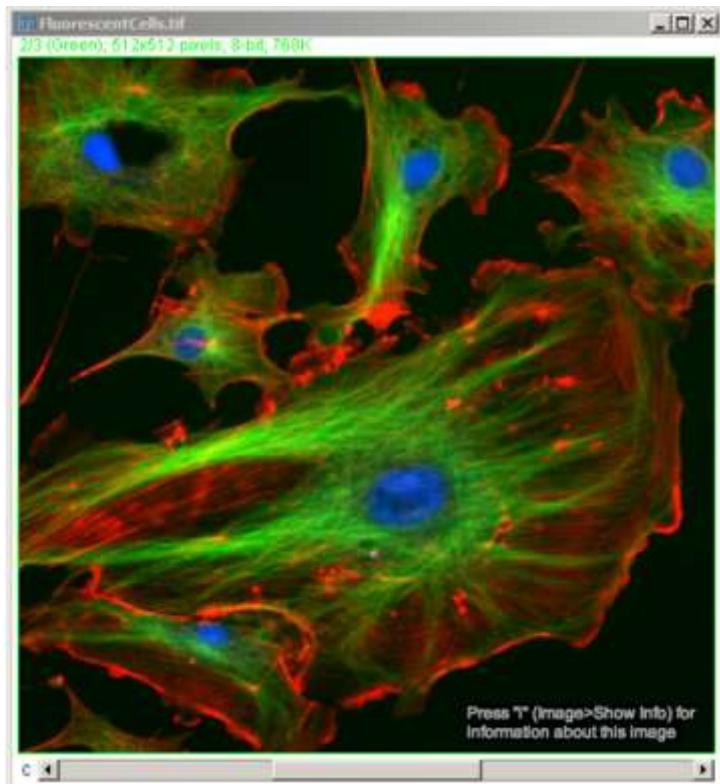
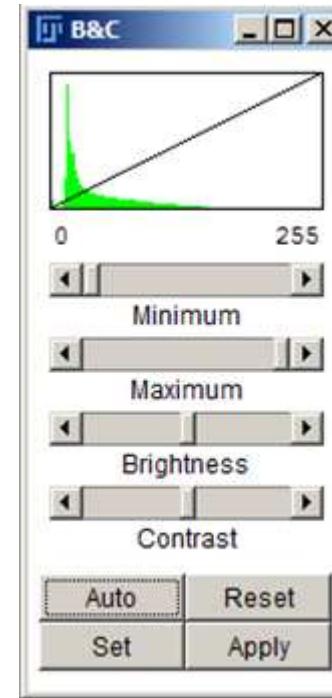


Image > Adjust > Brightness/Contrast



Composite: use slider to make different channels active

Brightness and Contrast

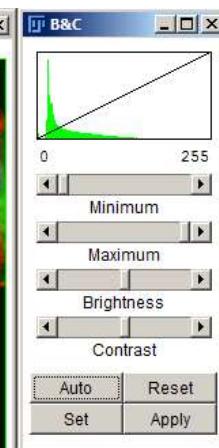
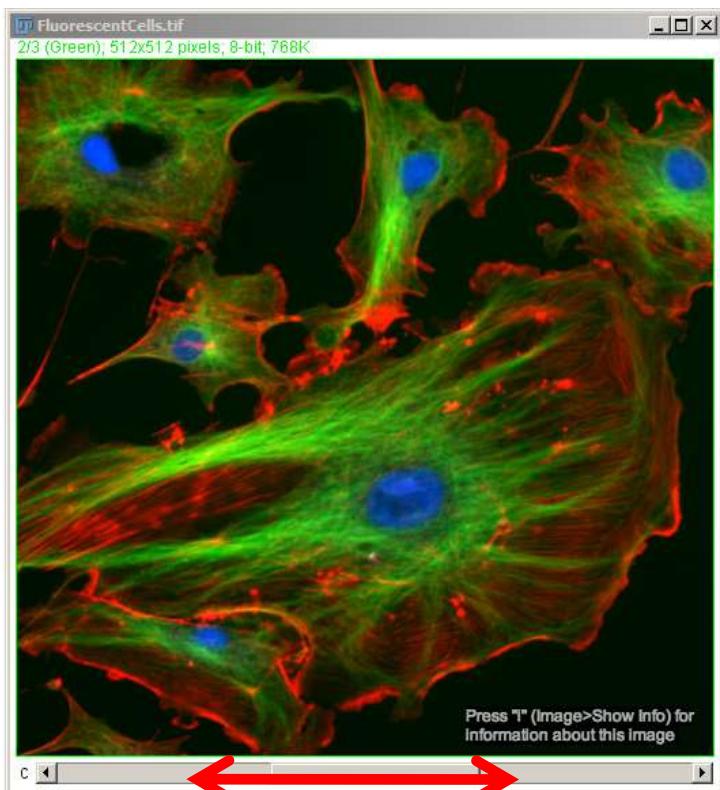
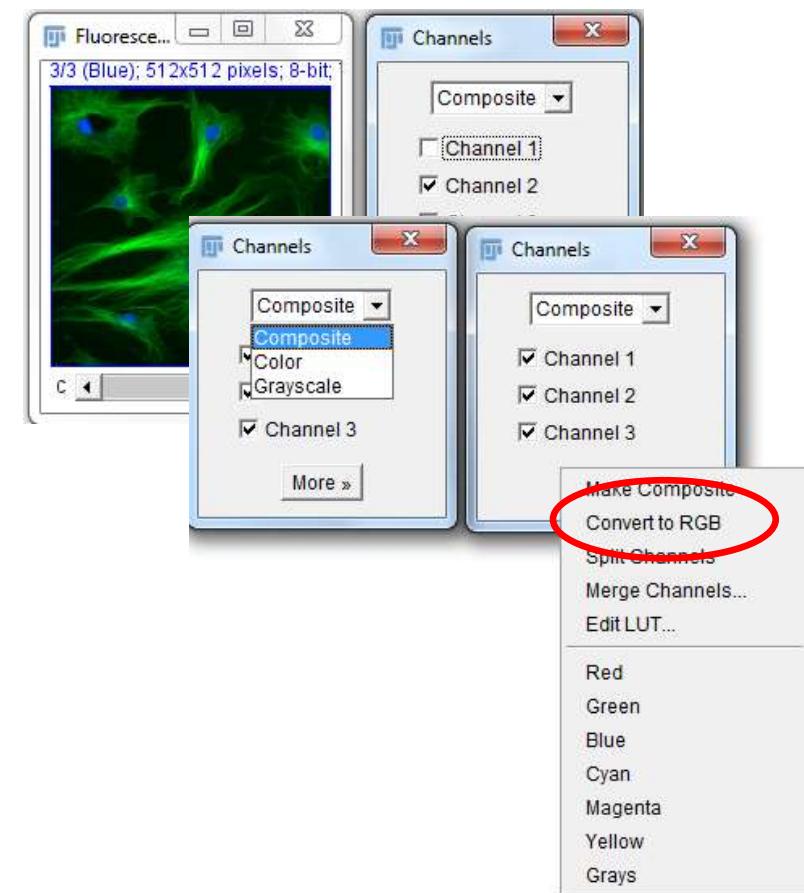


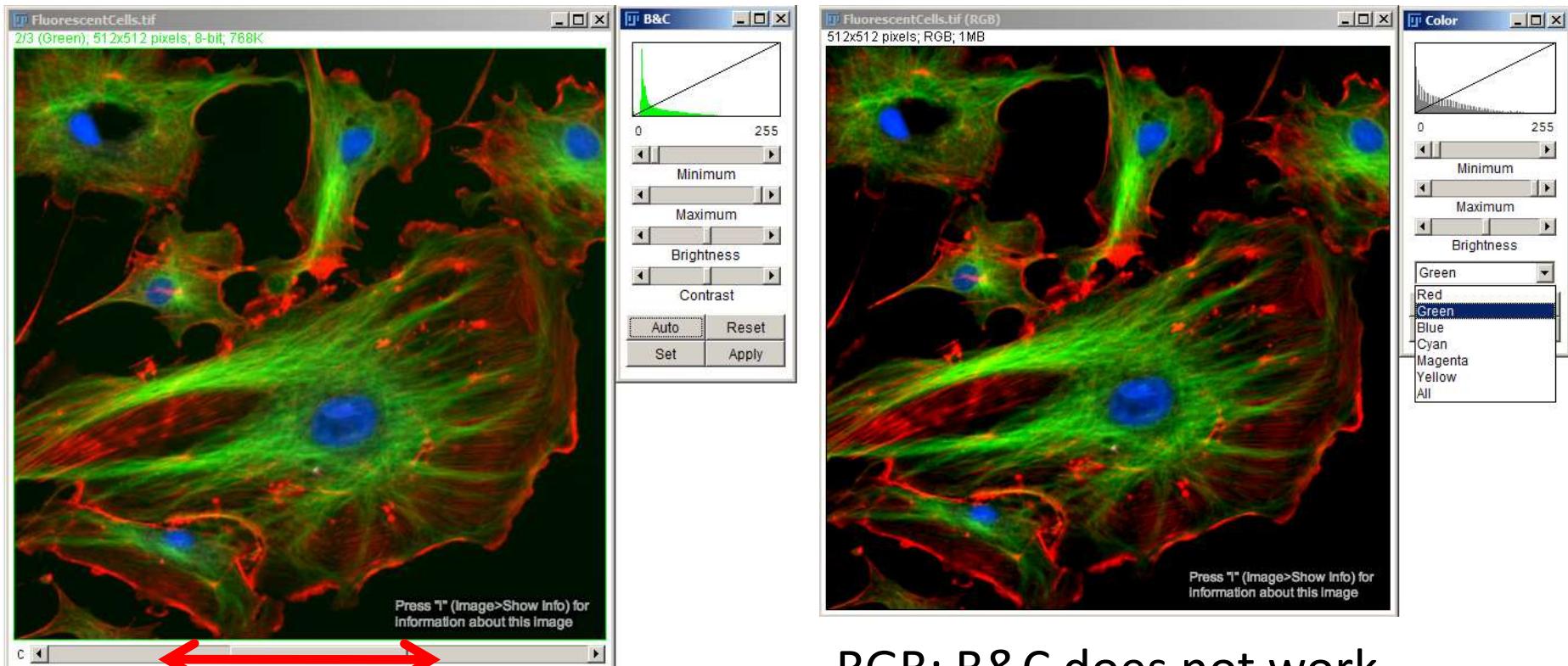
Image > Color > Channels Tools



Composite

Brightness and Contrast

Open sample Fluorescence Cells (400K)

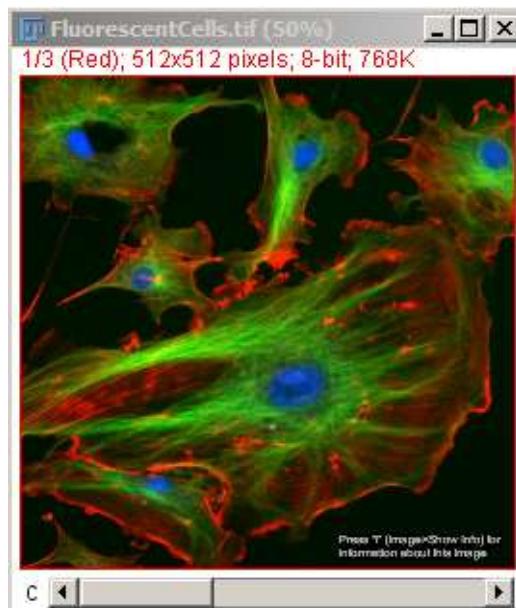


Composite

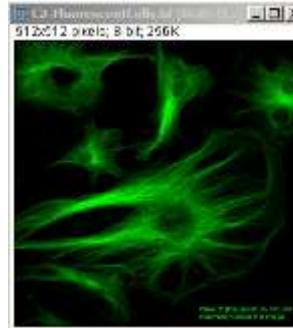
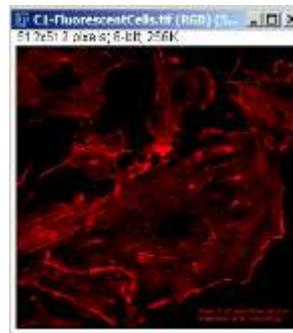
RGB: B&C does not work.
Use: *Image > Adjust > Color Balance*.

Split channels

File > Open samples > Fluorescence Cells



Split
Channels



Merge
Channels

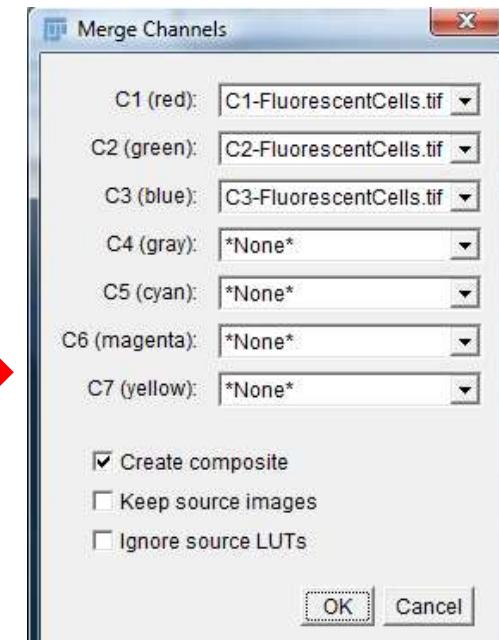
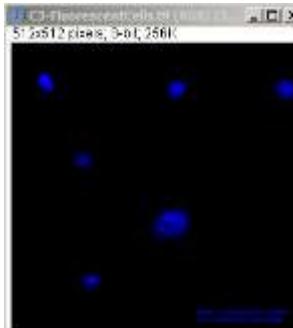


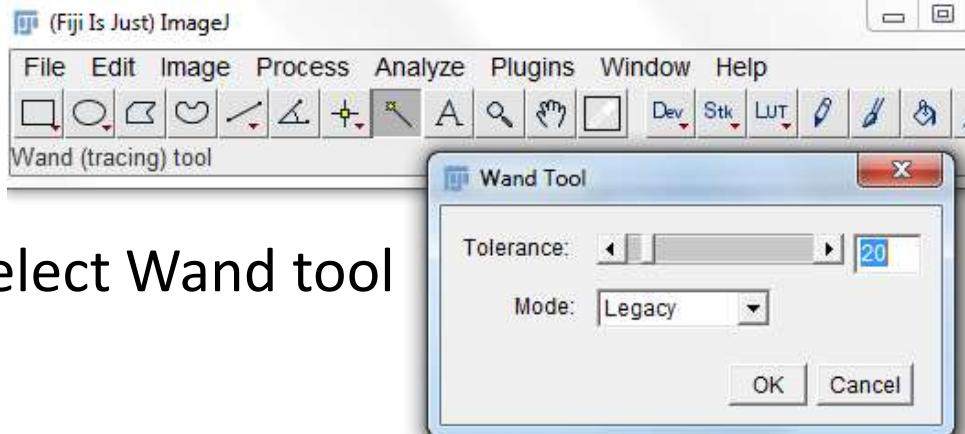
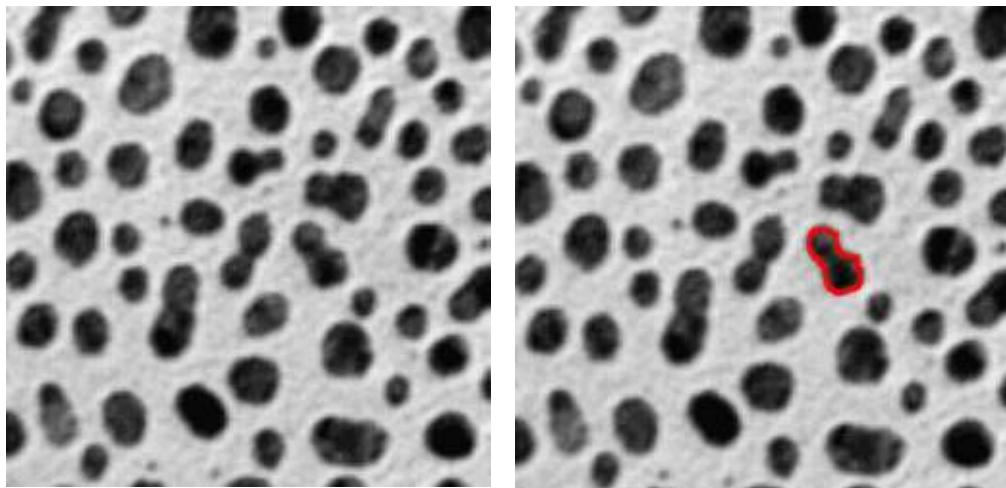
Image > Color > Split Channels



*Image > Color >
Merge Channels*

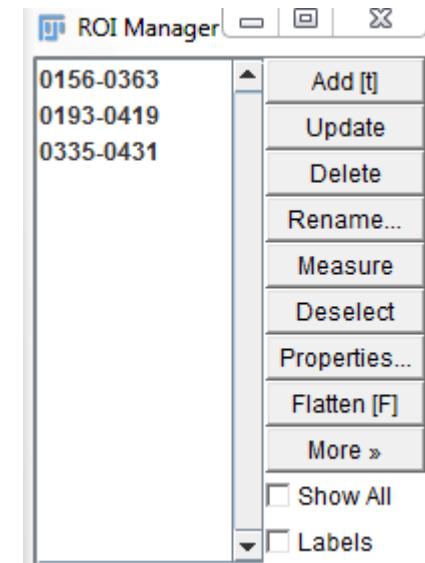
Selections

File > Open samples > Blobs

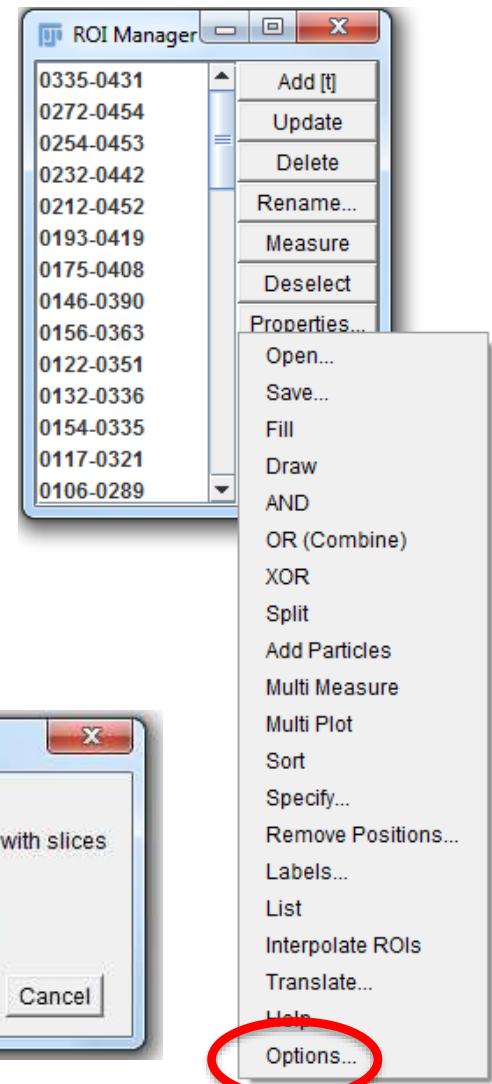
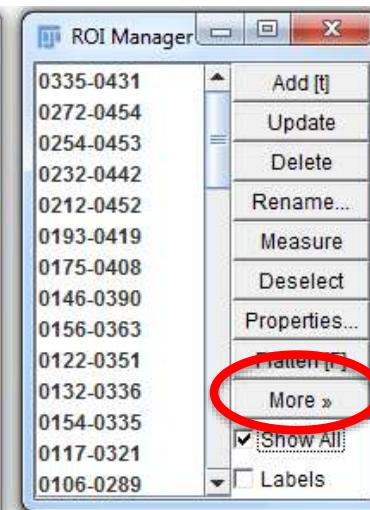
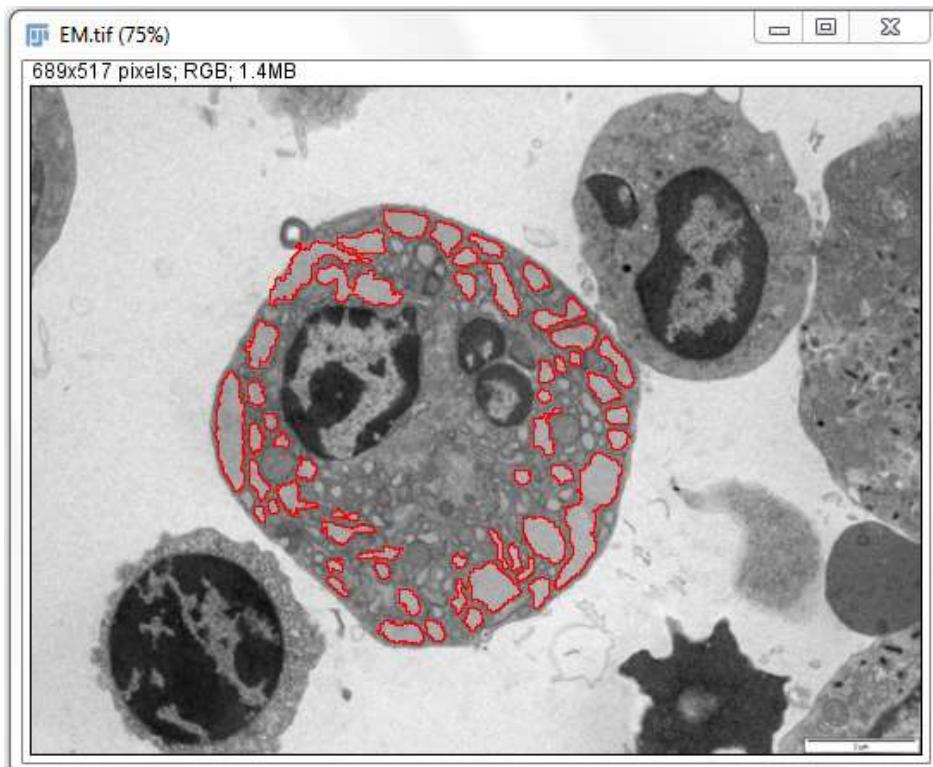


Select Wand tool

*Analyse > Tools >
ROI manager
(Short cut: t)*



Selections



Threshold

We create a binary image (black and white) to extract information

*File > Open Samples
> Blobs (25k)*

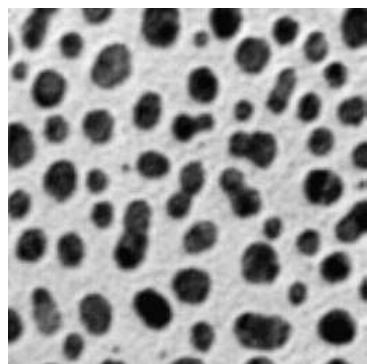
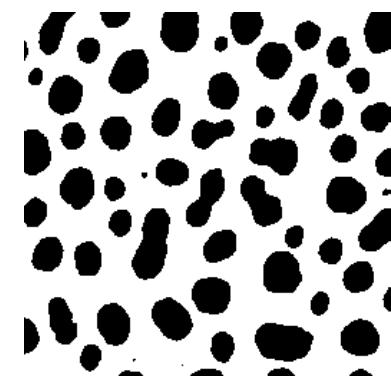
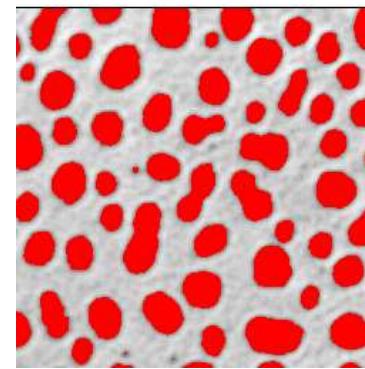
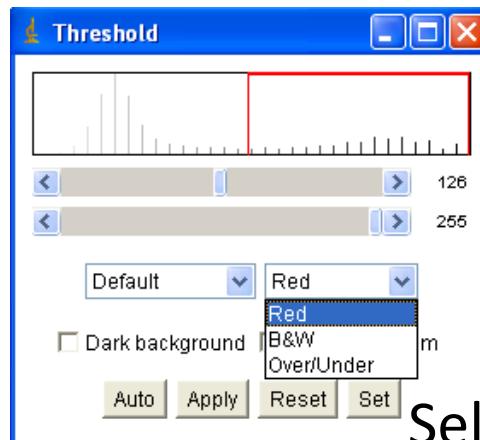


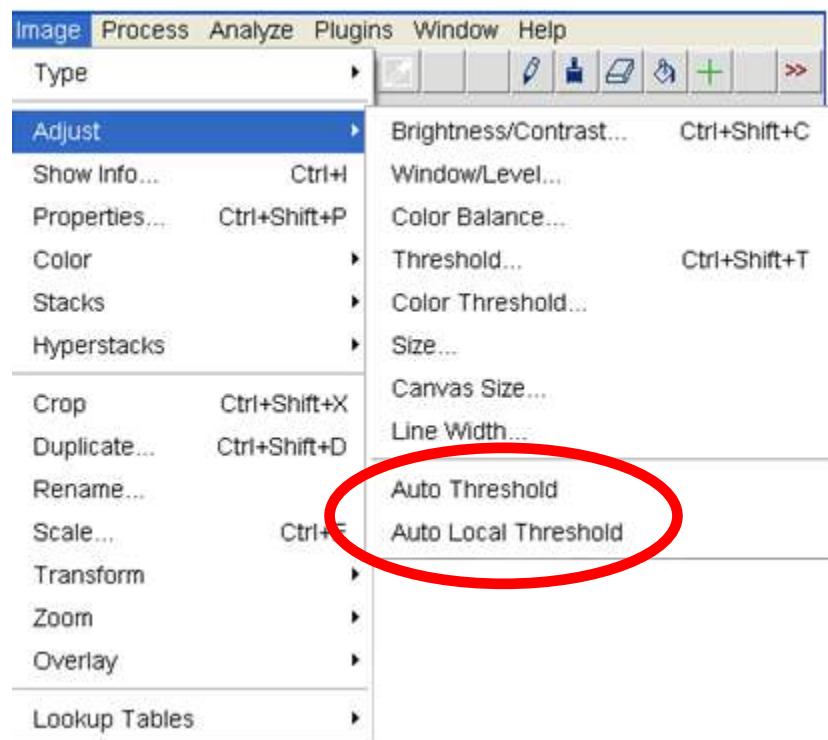
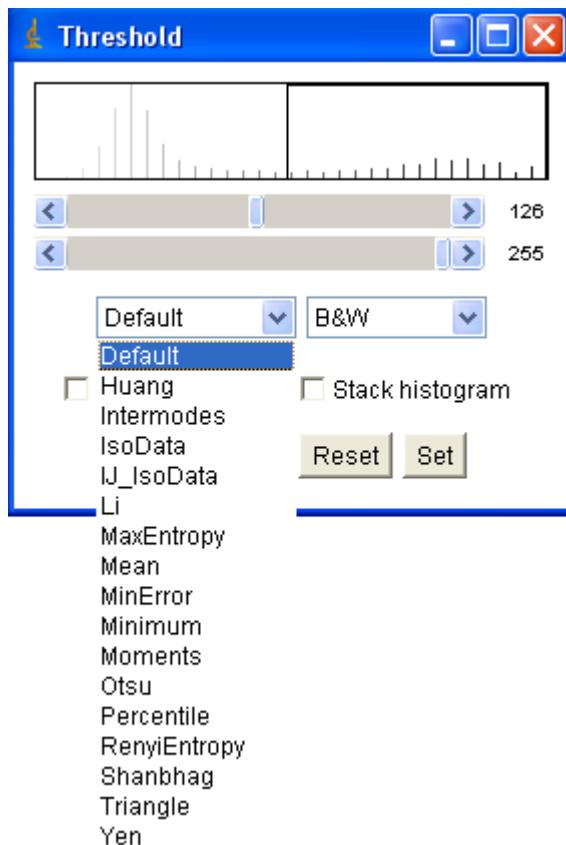
Image > Adjust > Threshold...



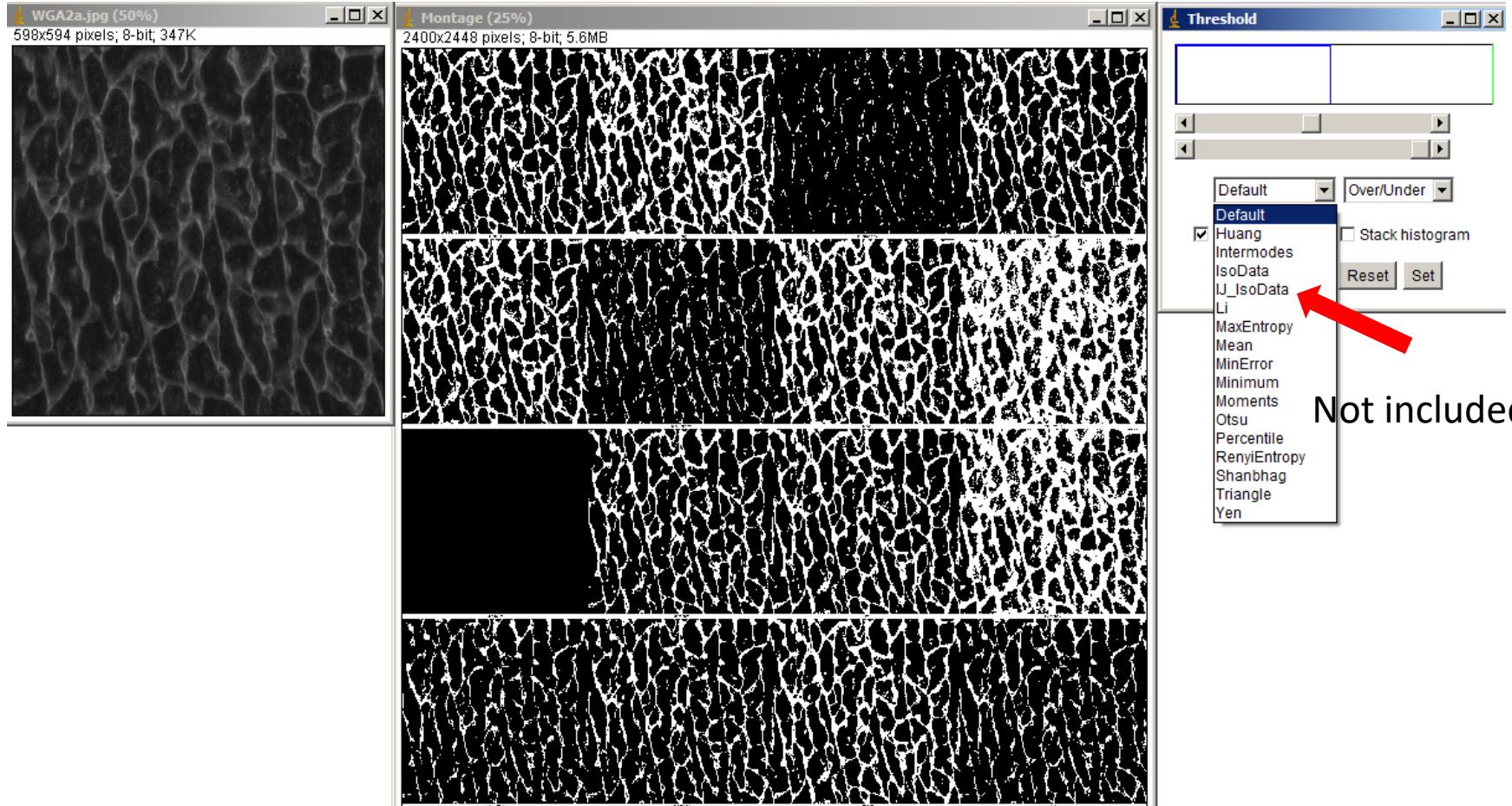
Select “Red”; You can
see what you select Select “Apply”

Historically, selected pixels have a value of 255 inside the selection and 0 outside, so black is 255 and white is 0 unless Black Background in *Process > Binary > Options* is checked.

Threshold



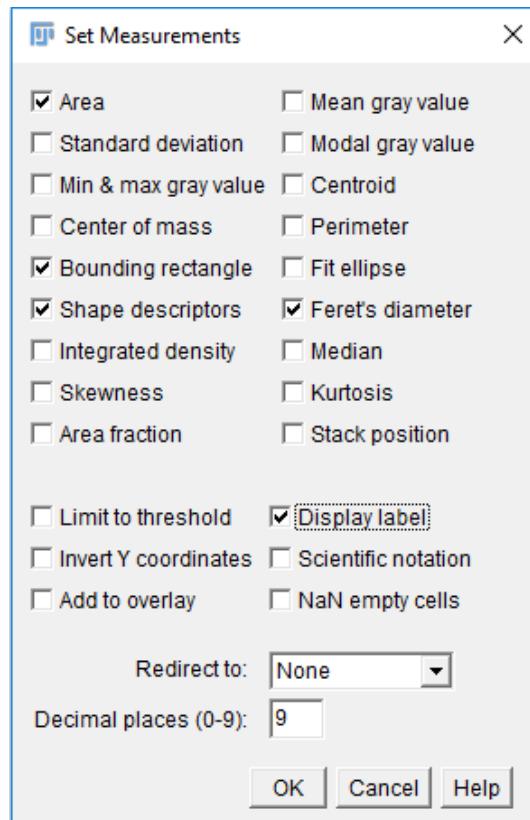
Threshold



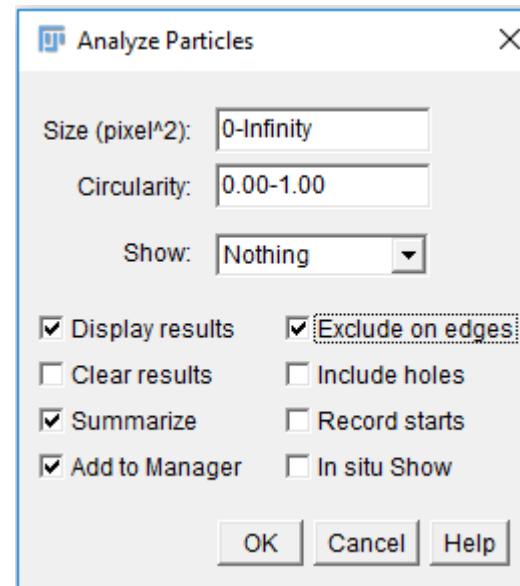
Auto Threshold

Measurements

Analyze > Set Measurements...



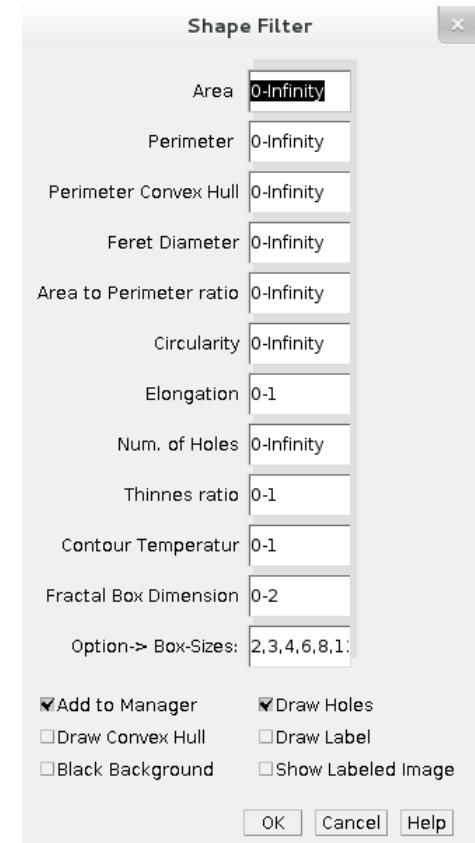
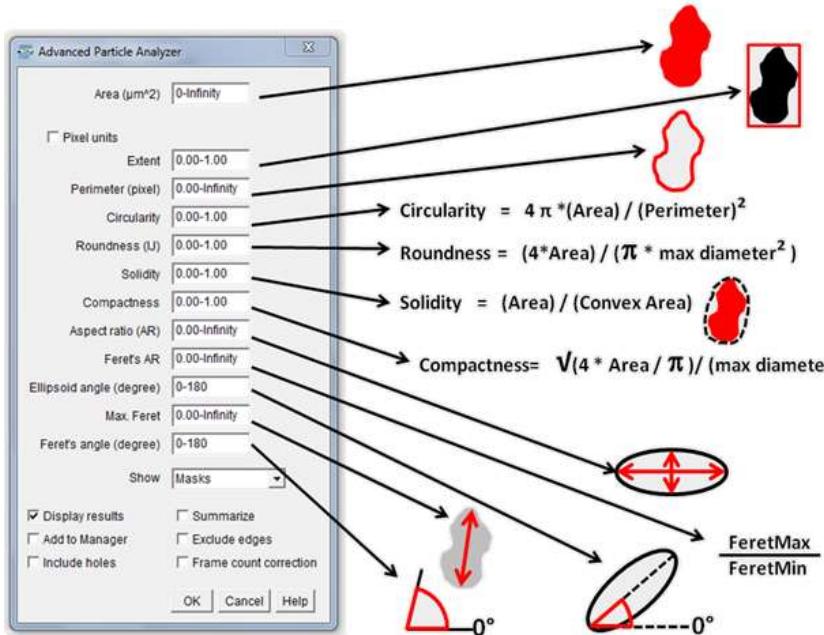
Analyze > Analyze Particles



Measurements

IJblob

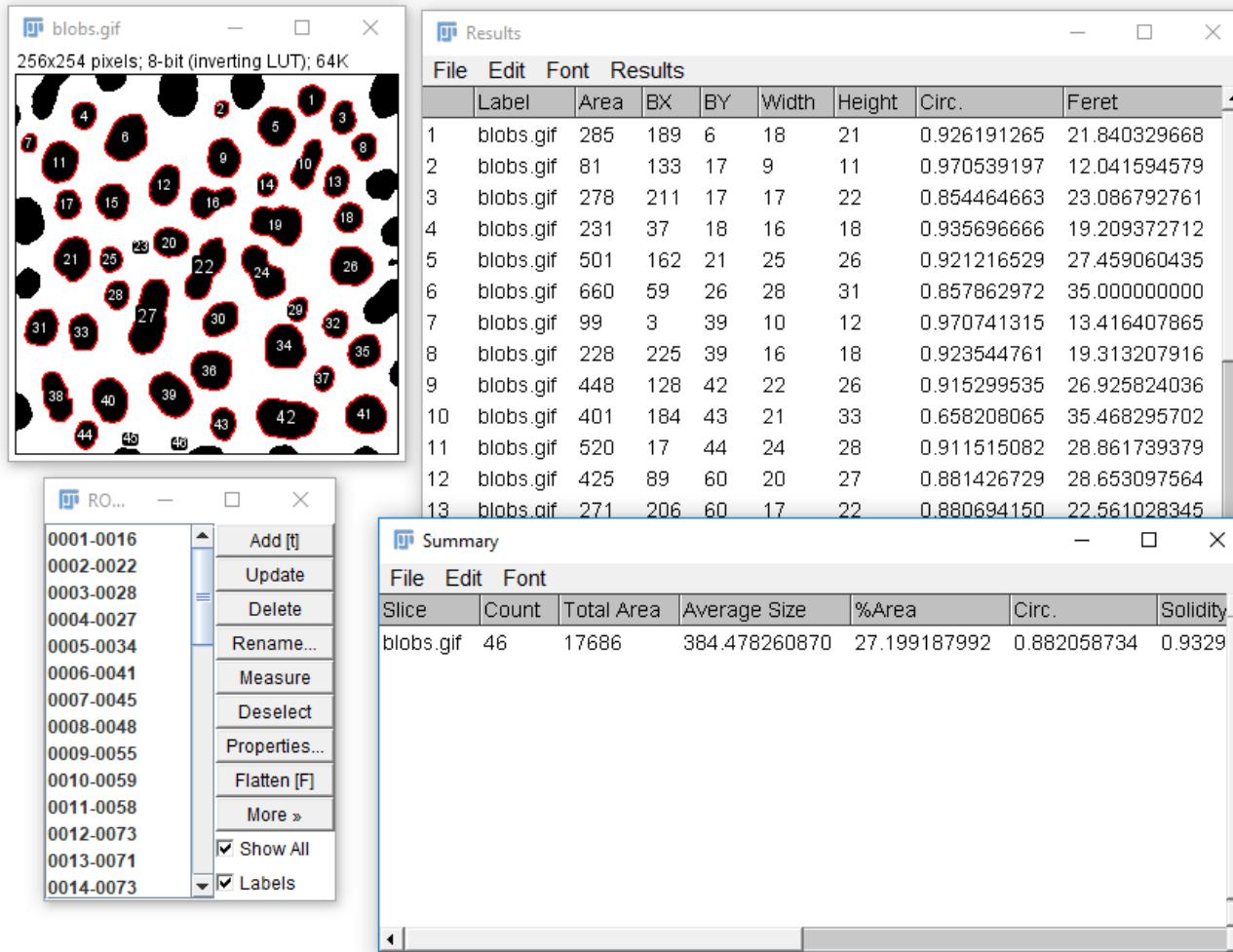
BioVoxel Toolset:
includes Advanced Particle Analyzer



<http://www.biovoxxel.de/macros.html>

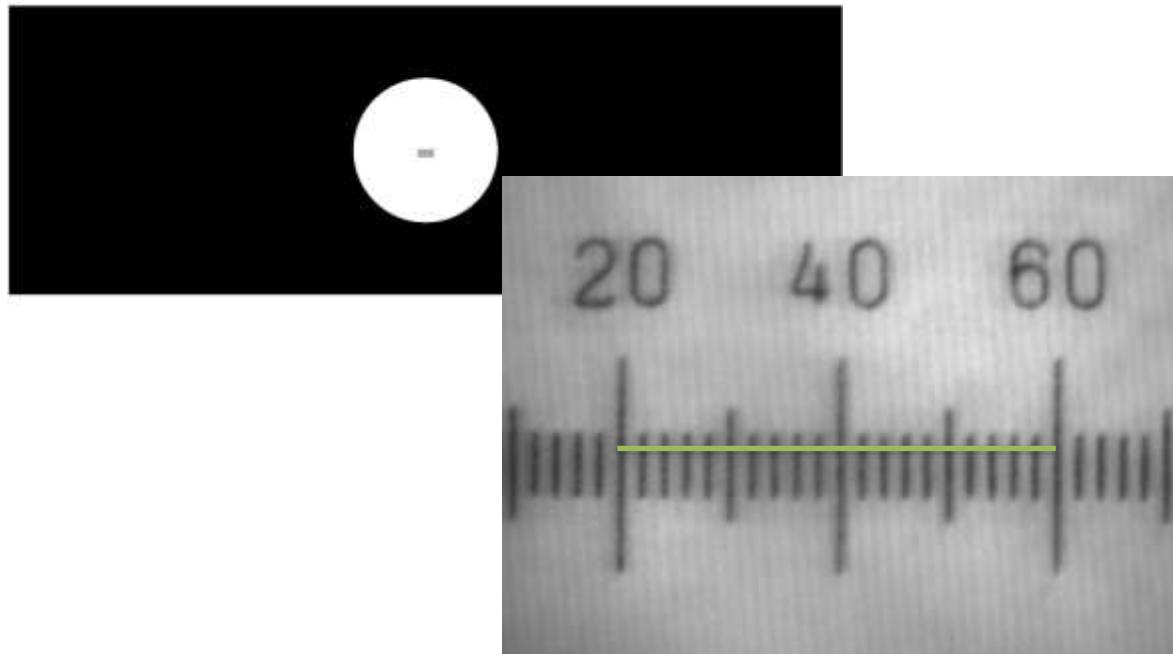
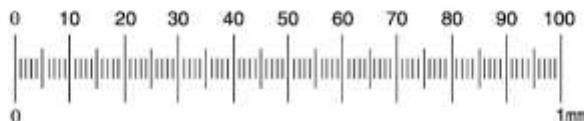
<https://code.google.com/p/ijblob/>

Measurements

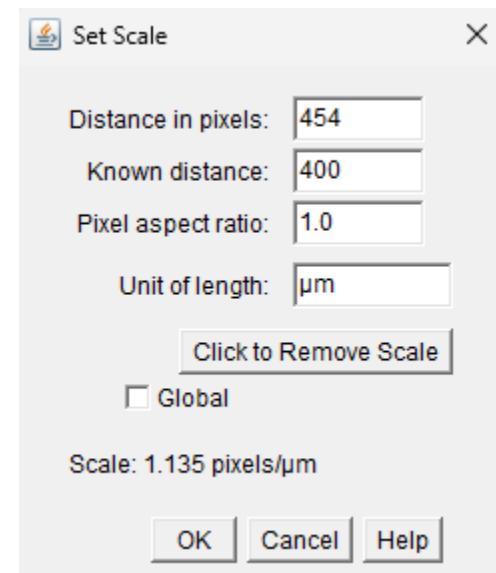


Set scale bar

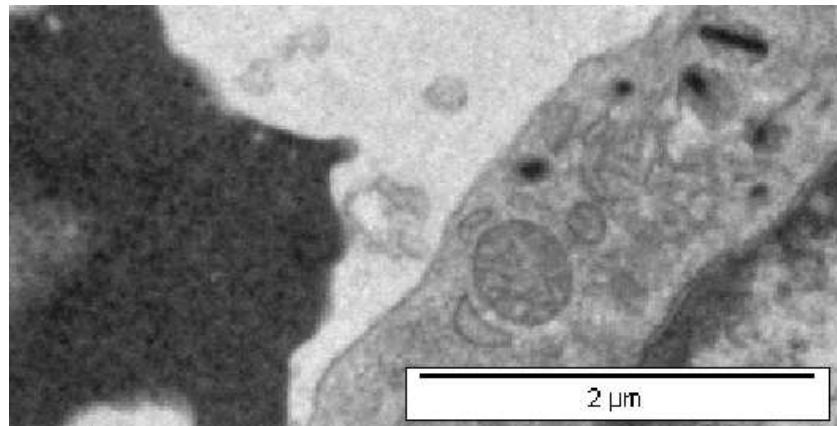
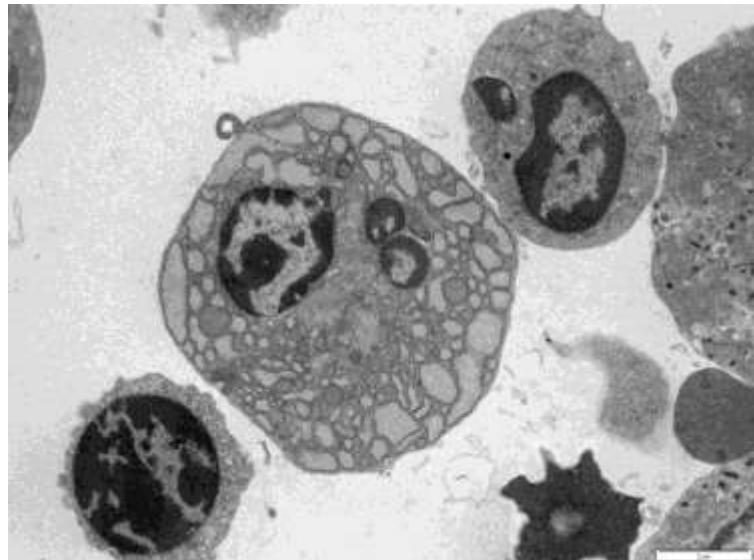
If you don't know the scale on a light microscope, use a stage graticule slide.



Analyze > Set Scale



Set scale bar

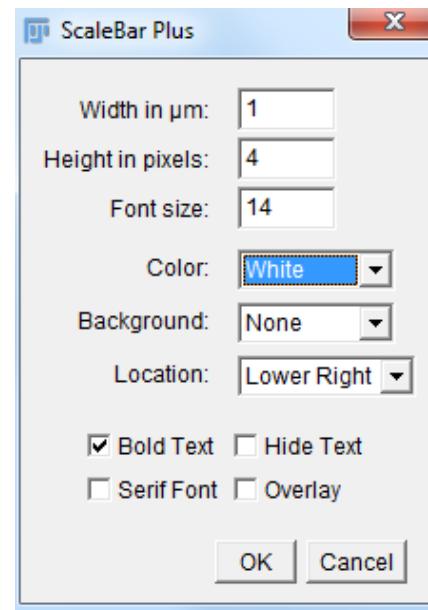


Make straight line on the scale bar

Set scale bar



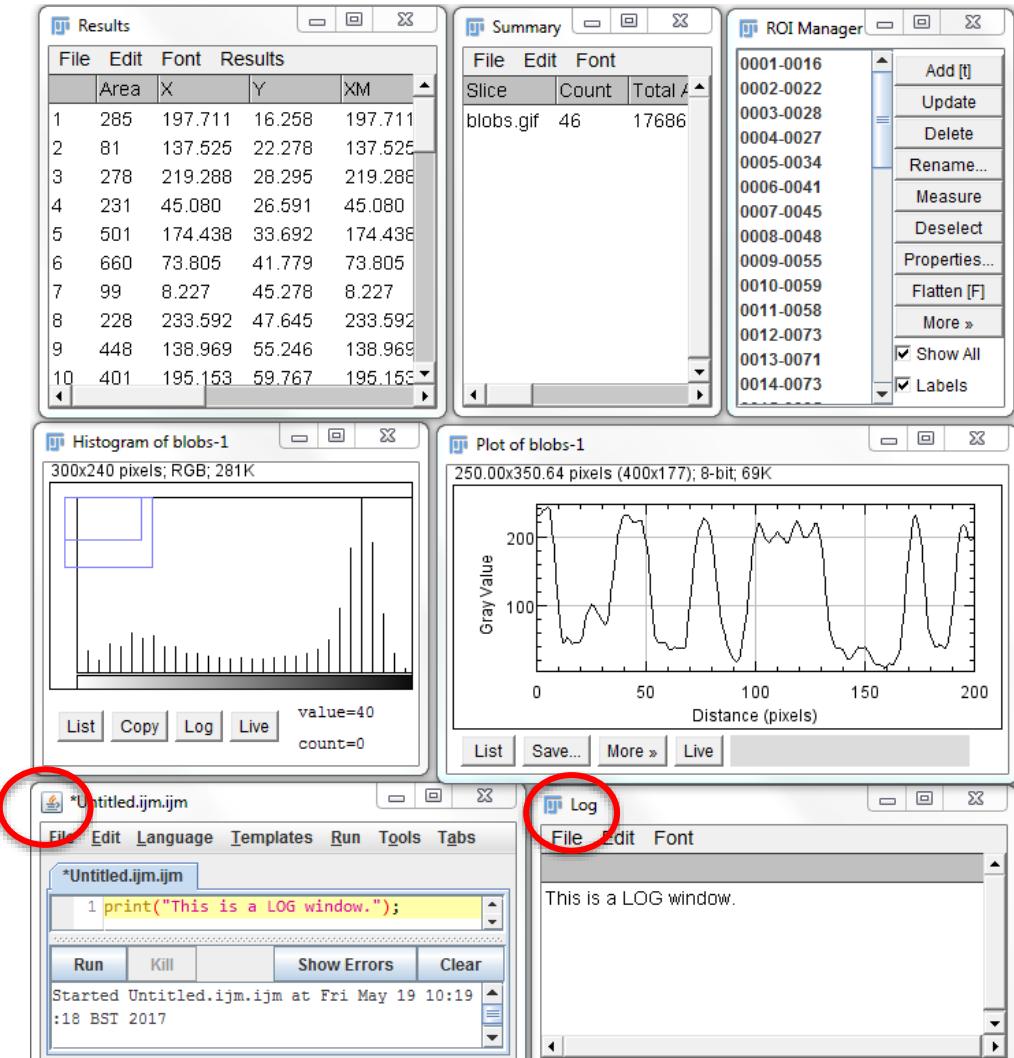
Analyze > Tools > Scale Bar...



Non-image windows

Windows besides image windows:

- Results
- Summary
- ROI manager
- Histogram
- Plots
- Editor
- Log
- Plugins
-



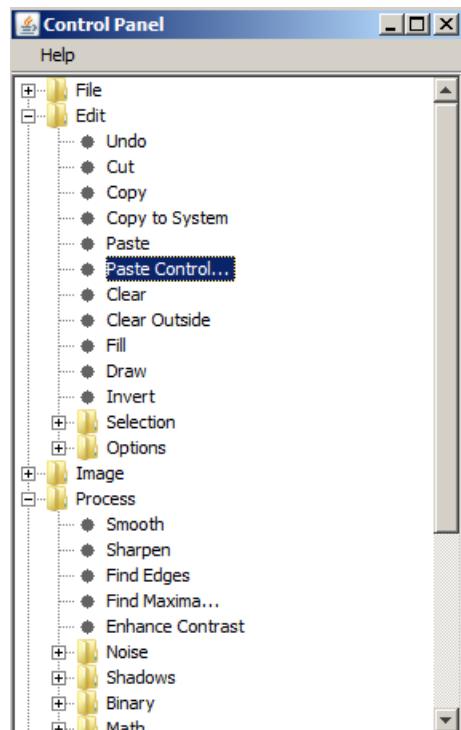
These windows have their own menus.

Find where you are looking for

Fiji search bar. Change appearance: *Edit > Options > Search Bar...*

Plugins > Utilities >

Control Panel



Find Command

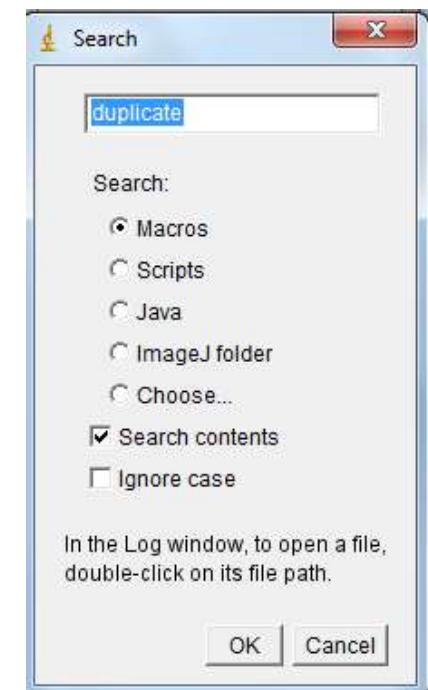
Command Finder

Command	Menu Path	Class	File
-11	Window		
-13	Window		
-33	Window		
-59	Window		
-7	Window		
-73	Window		
-79	Window		
-80	Window		
-About These Macros	Plugins>Examples>-Macros	ij.plugin.Macro_Runner("Example...")	
-About These Scripts	Plugins>Examples>-Scripts	ij.plugin.Macro_Runner("Example...")	
16 Colors	Image>Lookup Tables		
16-bit	Image>Type	ij.plugin.Converter("16-bit")	
3-3-2 RGB	Image>Lookup Tables	ij.plugin.LutLoader("3-3-2 RGB")	
32-bit	Image>Type	ij.plugin.Converter("32-bit")	
3D Project...	Image>Stacks	ij.plugin.Projector	
3D Viewer	Plugins>3D	ImageJ_3D_Viewer	D:\PRO...
5 Ramps	Image>Lookup Tables		
6 Shades	Image>Lookup Tables		

Close window after running command

Run Source Close

Search



Plugins

Plugins

If not included on update site plugins can be found at:

[ImageJ website](#)

Fiji Website

Google search for ImageJ plugins

Search on YouTube

Many institutional and personal websites (few examples)

- <http://bigwww.epfl.ch/algorithms.html>
- <http://www.mecourse.com/landinig/software/software.html>
- <http://www.sussex.ac.uk/gdsc/intranet/microscopy/imagej/findfoci>
- <http://research.stowers.org/imagejplugins/>
- <https://www.optinav.com/imagej-plugins>

Ask mailing list (first search archives!)



Plugins

<https://imagej.net/list-of-extensions>



Page hist

List of Extensions

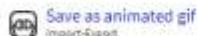
▼ Categories filter

Select All

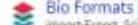
Select None

All Any

- | | | | | |
|---|---|--|--|---|
| <input checked="" type="checkbox"/> 3D | <input type="checkbox"/> Example Data | <input type="checkbox"/> Lisp | <input type="checkbox"/> Optic Flow | <input type="checkbox"/> Stacks |
| <input checked="" type="checkbox"/> Analysis | <input type="checkbox"/> Feature Extraction | <input type="checkbox"/> MATLAB | <input type="checkbox"/> Particle Analysis | <input type="checkbox"/> Stitching |
| <input checked="" type="checkbox"/> Automation | <input type="checkbox"/> Fiji | <input type="checkbox"/> Machine Learning | <input type="checkbox"/> Pattern Recognition | <input type="checkbox"/> Super-resolution |
| <input checked="" type="checkbox"/> Binary | <input type="checkbox"/> Filtering | <input type="checkbox"/> Mathematical Morphology | <input type="checkbox"/> Perfusion | <input type="checkbox"/> Tracking |
| <input checked="" type="checkbox"/> Colocalization | <input type="checkbox"/> Help | <input type="checkbox"/> Microscopy | <input type="checkbox"/> Photography | <input type="checkbox"/> TrakEM2 |
| <input checked="" type="checkbox"/> Color processing | <input type="checkbox"/> Image Annotation | <input type="checkbox"/> Microtubules | <input type="checkbox"/> Plotting | <input type="checkbox"/> Transform |
| <input checked="" type="checkbox"/> Cookbook | <input type="checkbox"/> ImageJ2 | <input type="checkbox"/> Montage | <input type="checkbox"/> Projection | <input type="checkbox"/> Tutorials |
| <input checked="" type="checkbox"/> Data | <input type="checkbox"/> ImageScience | <input type="checkbox"/> Neuroanatomy | <input type="checkbox"/> Registration | <input type="checkbox"/> Uncategorized |
| <input checked="" type="checkbox"/> Deconvolution | <input type="checkbox"/> ImgLib | <input type="checkbox"/> Noise | <input type="checkbox"/> SciJava | <input type="checkbox"/> Unmaintained |
| <input checked="" type="checkbox"/> Development | <input type="checkbox"/> Import-Export | <input type="checkbox"/> OME | <input type="checkbox"/> Scripting | <input type="checkbox"/> User interface |
| <input checked="" type="checkbox"/> Digital Volume Flattening | <input type="checkbox"/> Integral Image | <input type="checkbox"/> Object Detection | <input type="checkbox"/> Segmentation | <input type="checkbox"/> Visualization |
| <input checked="" type="checkbox"/> Digital Volume Unrolling | <input type="checkbox"/> Interactive | <input type="checkbox"/> Ops | <input type="checkbox"/> Skeleton | |



Import-Export



Import-Export, SciJava, OME



Import-Export



Import-Export



Import-Export



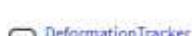
Import-Export



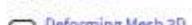
Import-Export



Import-Export



Uncategorized



Uncategorized



Uncategorized



Scripting



Uncategorized



Uncategorized



Uncategorized



Stacks



Uncategorized



Transform, Registration



Segmentation



Uncategorized



Uncategorized



Uncategorized



Tutorials



Color processing



Uncategorized



Analyze



Uncategorized



Image Annotation



Uncategorized



Uncategorized



Filters

Install plugin via update

Help > Update....

ImageJ Updater

Name	✓ Java-8
plugins/Sholl_Analysis.jar	U 2015-Conference
plugins/Simple_Neurite_Tracer.jar	U 3D ImageJ Suite
plugins/TrackMate_.jar	U 3Dscript
plugins/View5D_.jar	U ActogramJ
plugins/bigdataviewer_fiji.jar	U AIC Janelia - Course
plugins/bigwarp_fiji.jar	U Angiogenesis
plugins/bio-formats_plugins.jar	U AngioTool
plugins/n5-viewer_fiji.jar	U Archipelago
jars/FilamentDetector.jar	U AxoNet
jars/bigdataviewer-core.jar	U BACMMAN
jars/bigdataviewer-vistools.jar	U BAR
jars/bio-formats/formats-api.jar	U BaSiC
jars/bio-formats/formats-bsd.jar	U BigDataProcessor
jars/bio-formats/formats-gpl.jar	U BigDataViewer-Playground
jars/bio-formats/ome-common.jar	U BIG-EPFL
jars/bio-formats/ome-xml.jar	U BigStitcher
jars/bio-formats/specification.jar	U BigVolumeViewer Demo
jars/bio-formats/turbojpeg.jar	U Bio-Formats
jars/imglib2-cache.jar	U Biomat
jars/n5.jar	U Biomedaroun
jars/n5-google-cloud.jar	U
jars/n5-hdf5.jar	U
jars/n5-ij.jar	U
jars/n5-imglib2.jar	U
jars/scifio.jar	U

Manage update sites

Add my site Add update site Remove Update URLs Close

Apply changes Advanced mode Cancel

Update it Java-8
Update it Java-8

Overview of update sites + additional information:
<https://imagej.net/list-of-update-sites/>

Note

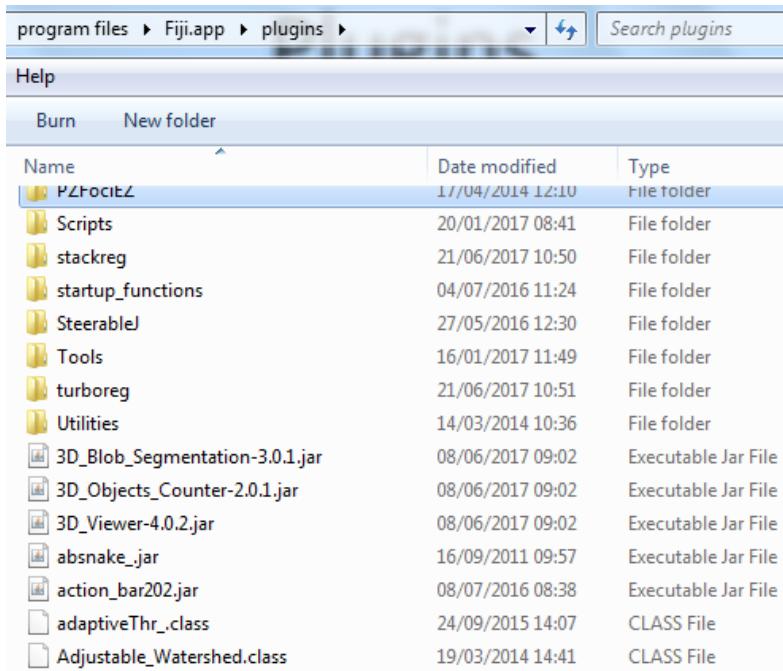
- All ImageJ plugins can be used in Fiji.
- You can have more than one ImageJ/Fiji installation on a computer.
- If you use Fiji, don't forget all those other nice plugins not included in Fiji update sites.

!!Don't forget to put a reference to the plugin in your publication!!

Plugins

If not via update site: download plugin and install in the subfolder 'plugins'. Restart Fiji.

- When *.class and *.jar files.
(*.jar files are *.zip files; collection of files.)



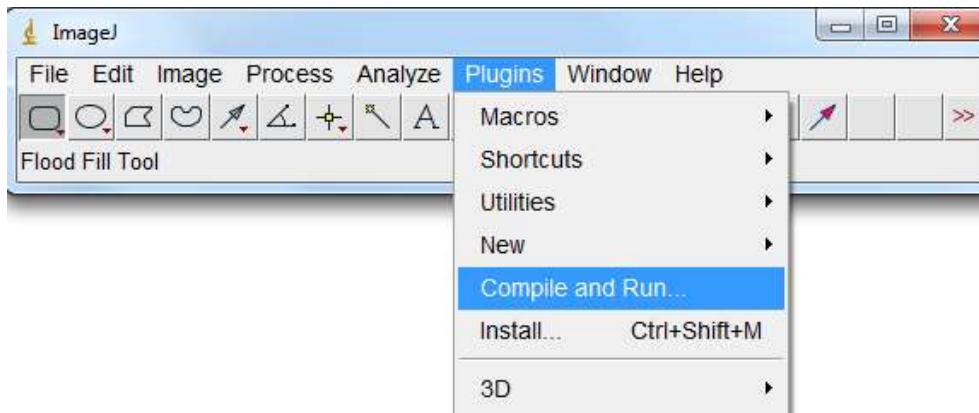
Name	Date modified	Type
PZFocus	11/04/2014 12:10	File folder
Scripts	20/01/2017 08:41	File folder
stackreg	21/06/2017 10:50	File folder
startup_functions	04/07/2016 11:24	File folder
Steerable	27/05/2016 12:30	File folder
Tools	16/01/2017 11:49	File folder
turboreg	21/06/2017 10:51	File folder
Utilities	14/03/2014 10:36	File folder
3D_Blob_Segmentation-3.0.1.jar	08/06/2017 09:02	Executable Jar File
3D_Objects_Counter-2.0.1.jar	08/06/2017 09:02	Executable Jar File
3D_Visualizer-4.0.2.jar	08/06/2017 09:02	Executable Jar File
absnake_.jar	16/09/2011 09:57	Executable Jar File
action_bar202.jar	08/07/2016 08:38	Executable Jar File
adaptiveThr_.class	24/09/2015 14:07	CLASS File
Adjustable_Watershed.class	19/03/2014 14:41	CLASS File

Plugins

If not via update site: download plugin and install in the subfolder 'plugins'. Restart Fiji.

- When *.class and *.jar files.
(*.jar files are *.zip files; collection of files.)

- If plugin file is *.java you have to use submenu: *Plugins > Compile and Run to create a *.jar file*; **this works only in ImageJ!** Take care capitals in name are maintained.

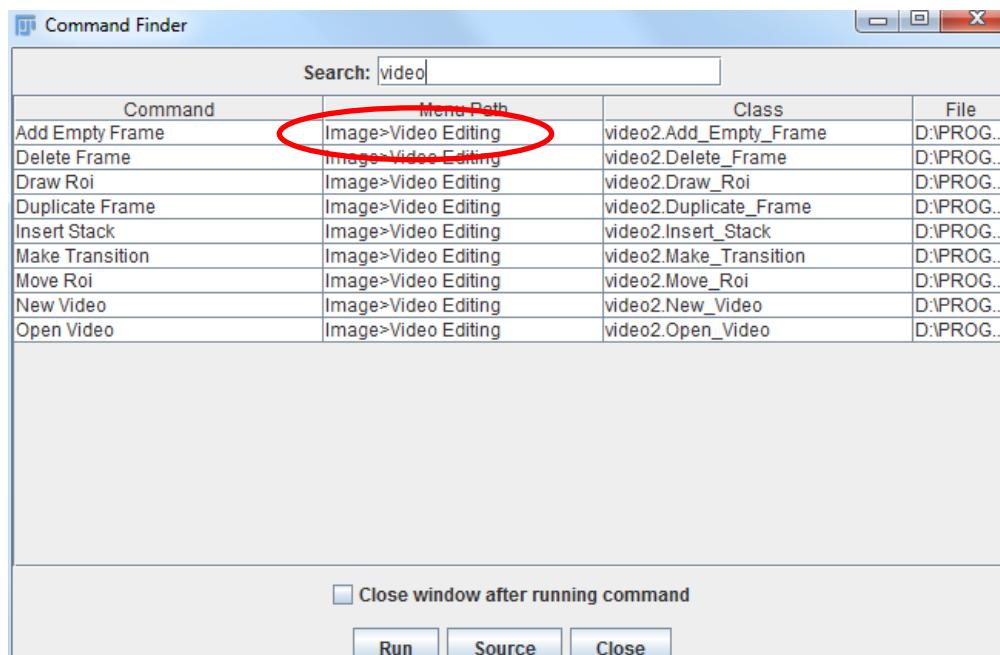


Plugins

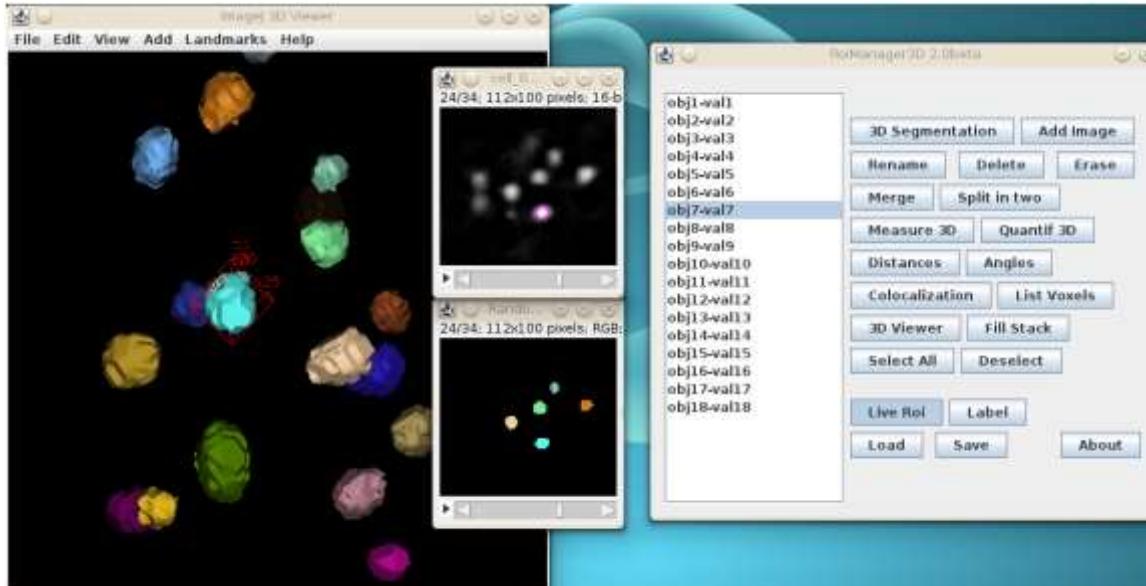
Plugins not always in Plugins menu, so how to find them?

For example: Video editing (Video_Editing.jar)

Use search bar or *Plugins > Utilities > Find Commands*



3D ROI manager

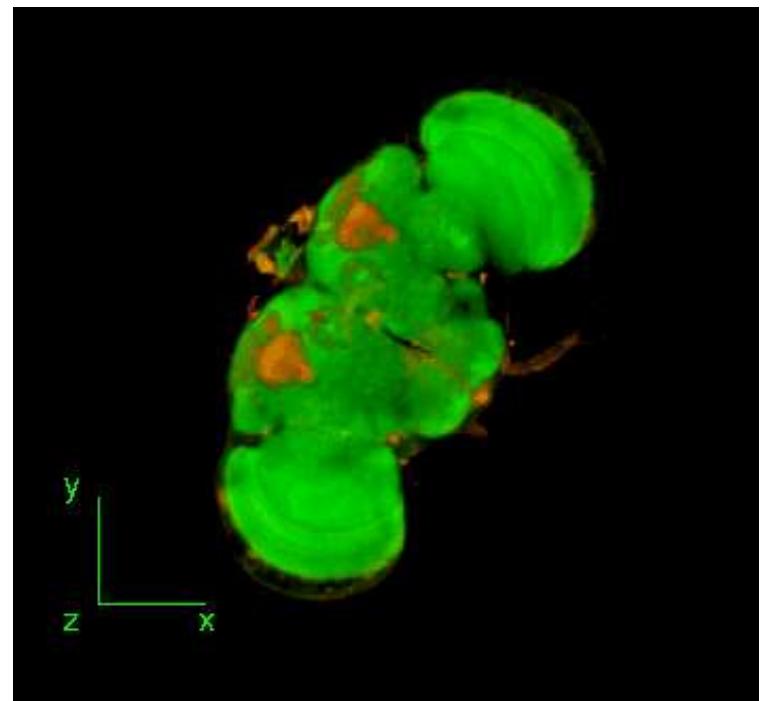


<https://mcib3d.frama.io/3d-suite-imagej/plugins/3DManager/3D-Manager/> part of 3D Suite ImageJ
<https://mcib3d.frama.io/3d-suite-imagej/>

3D viewer

3D Viewer: provides hardware-accelerated 3D visualization of image stacks as volumes, surfaces and orthoslices.

You can export into 3D formats that you can use in other programs like Blender.



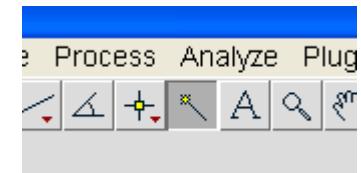
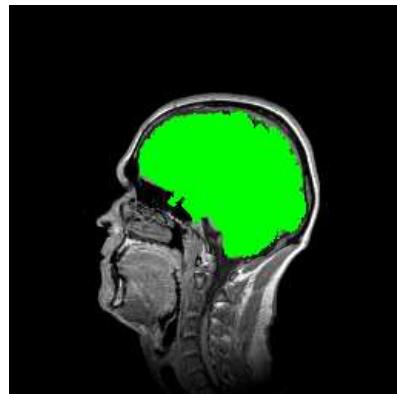
3D viewer

Sample T1-head.tif



Make RGB
(*Image > Type*)

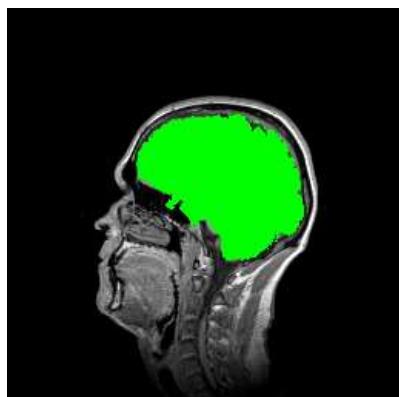
Select using wand tool and
add selection to ROI manager
(*Analyze > Tools > ROI Manager*)



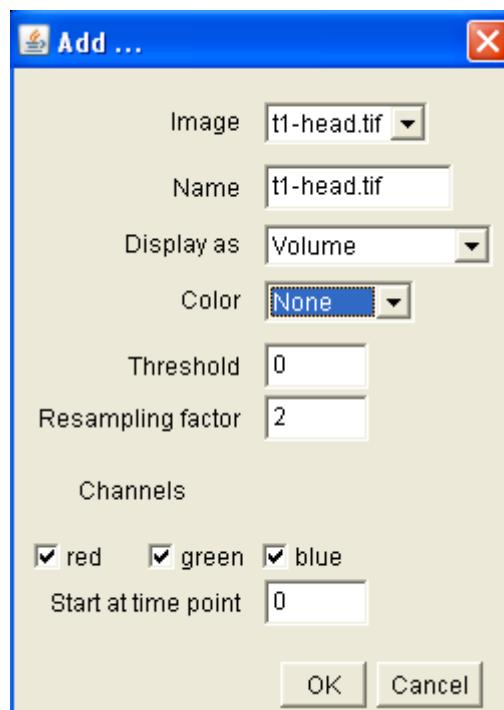
In ROI manager:
More >> Options > select
Associate “Show All” ROIs with
Slices.

Wand tool: Tolerance 40; Shift = add selection;
Alt = delete selection;
Ctrl + Shift + E = restore selection

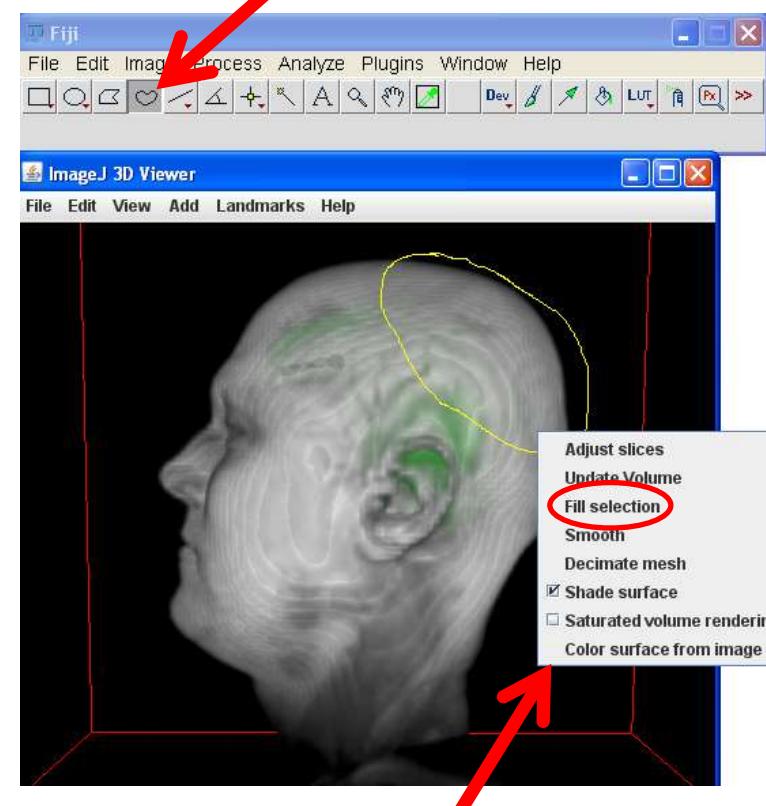
3D viewer



Plugins > 3D Viewer

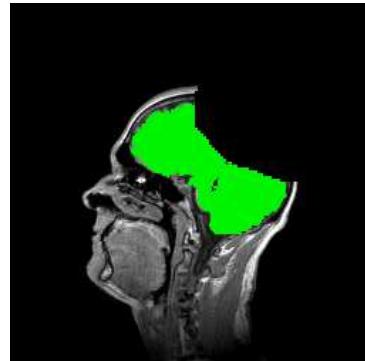
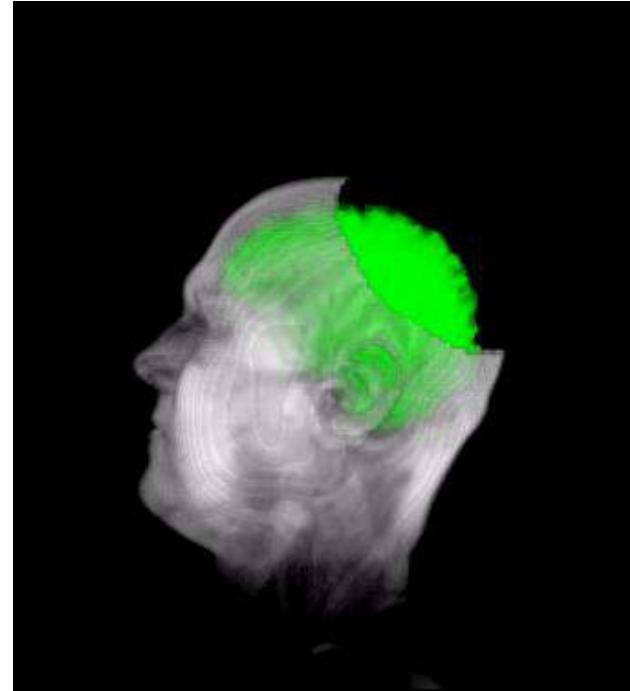
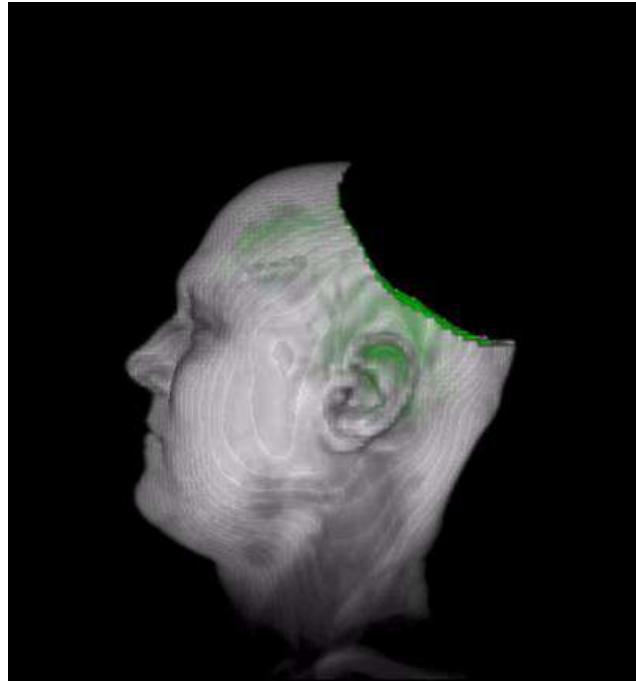


Freehand selection

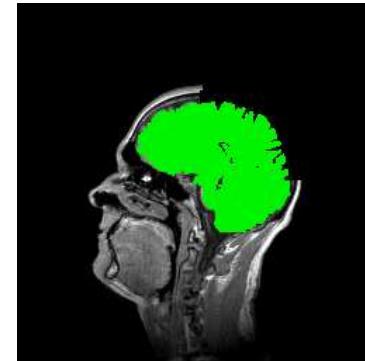


Right mouse click in selection

3D viewer

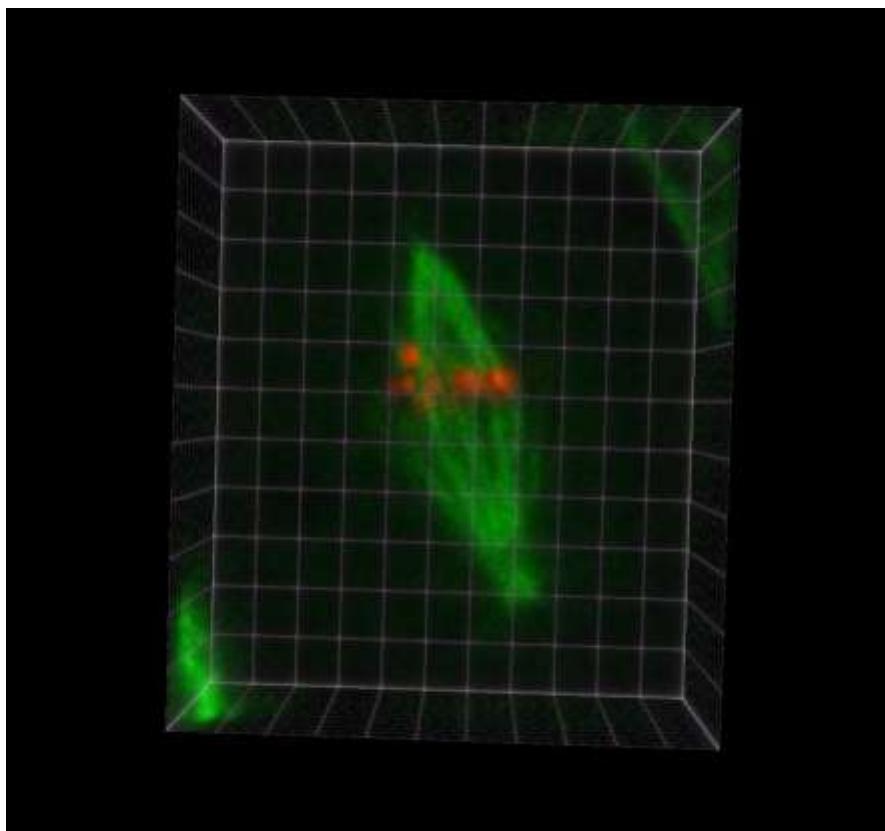


Use ROI manager
to restore ROIS



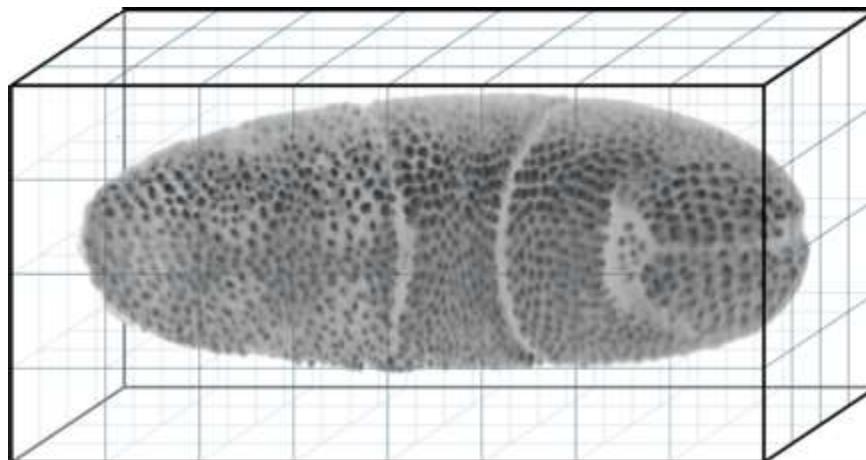
3D image display

ClearVolume



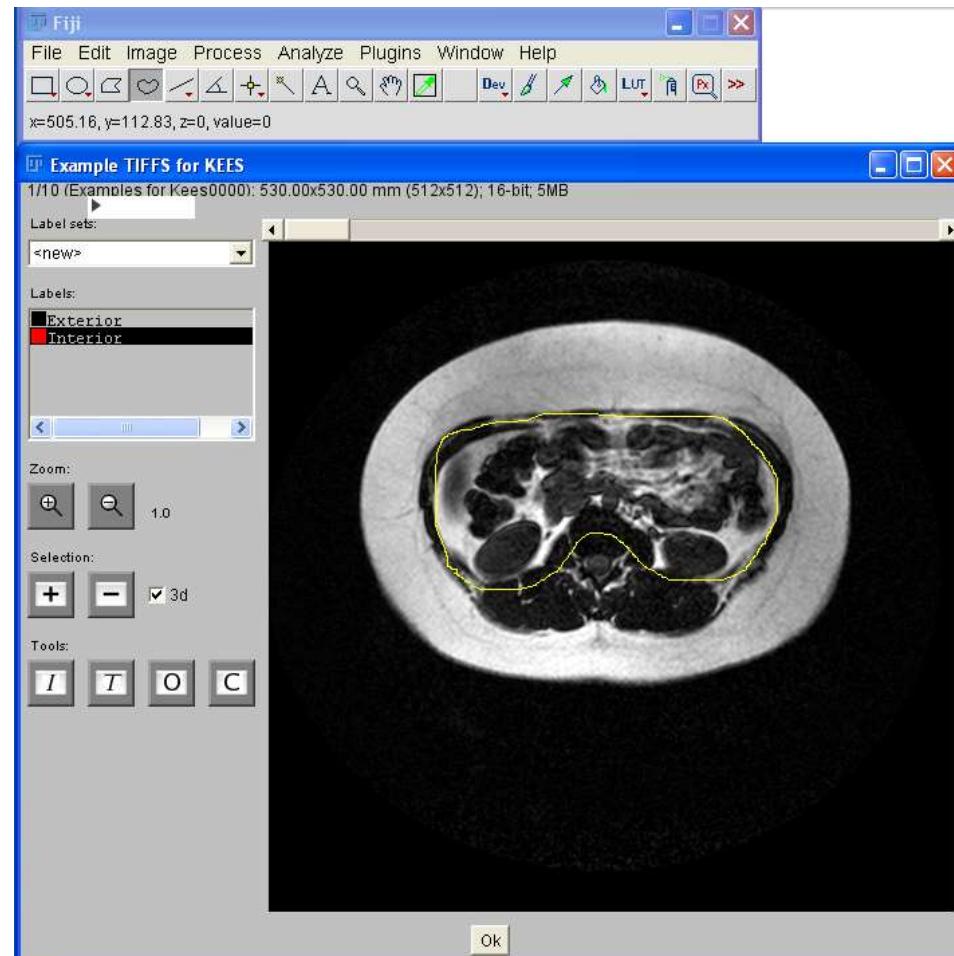
3D image display

BigDataViewer: browser for terabyte-sized multi-view image sequences



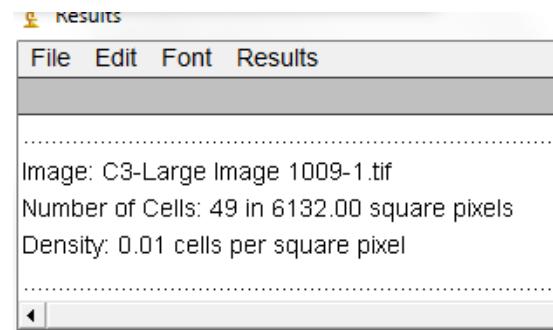
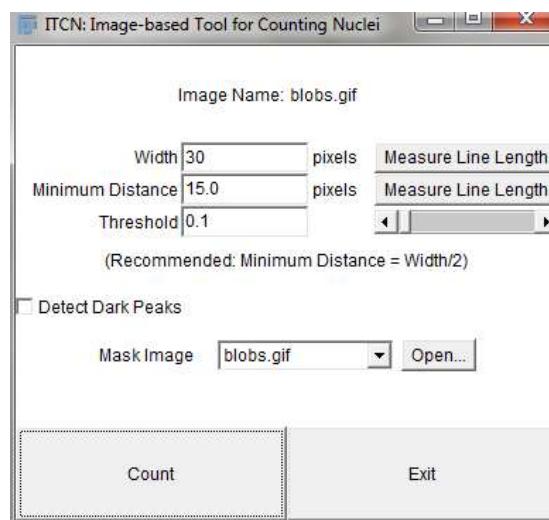
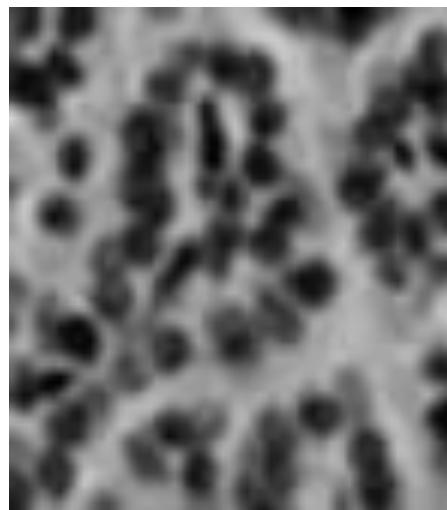
Segmentation editor

Allows 3D editing of segments. Make your selection on a few slices. Select Tools > / followed by Selection > + (with 3d selected).

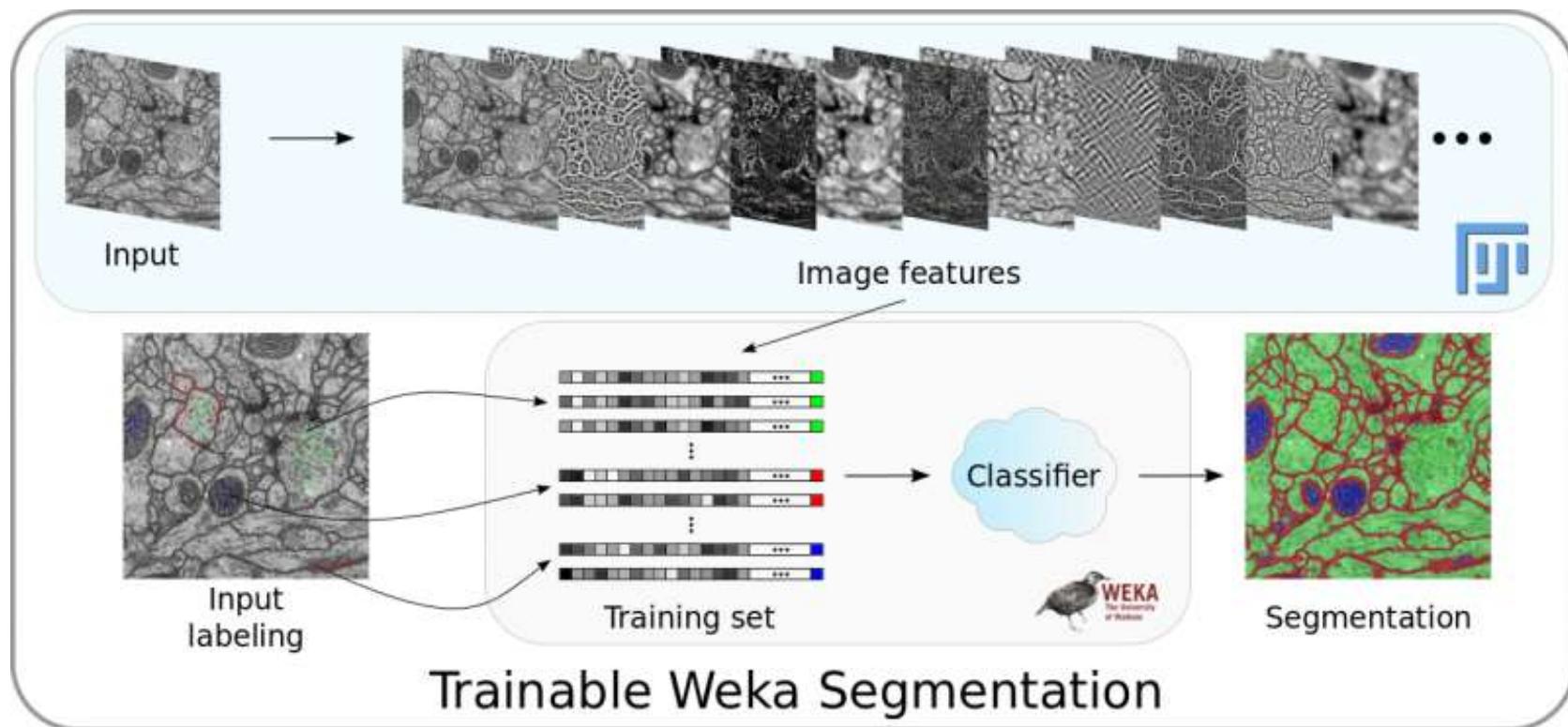


Segmentation

Automatic Nuclei Counter



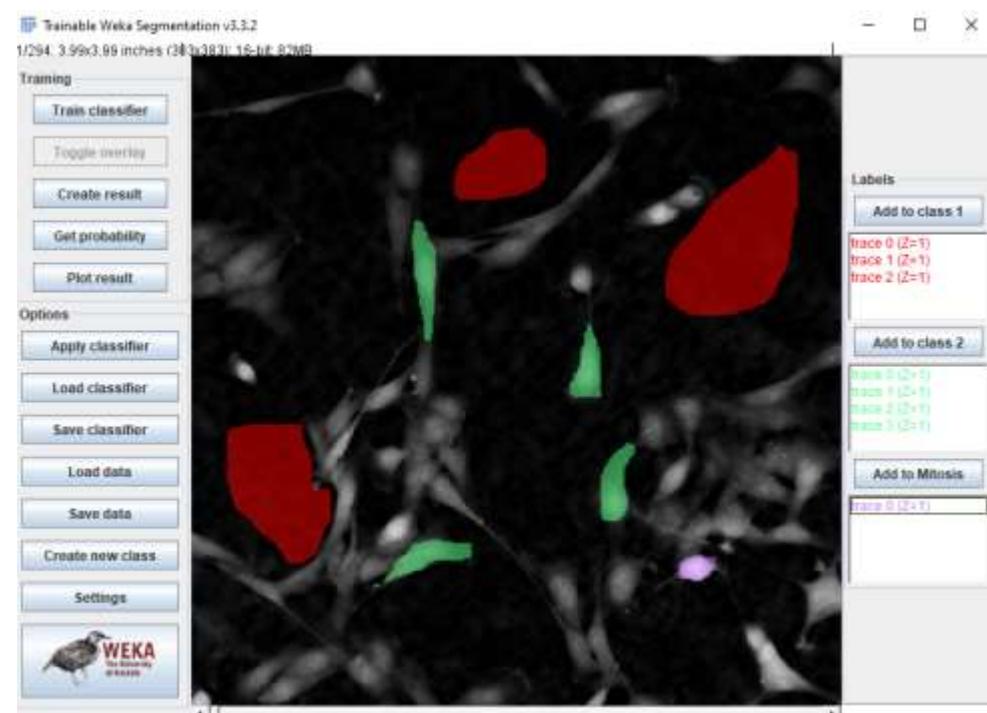
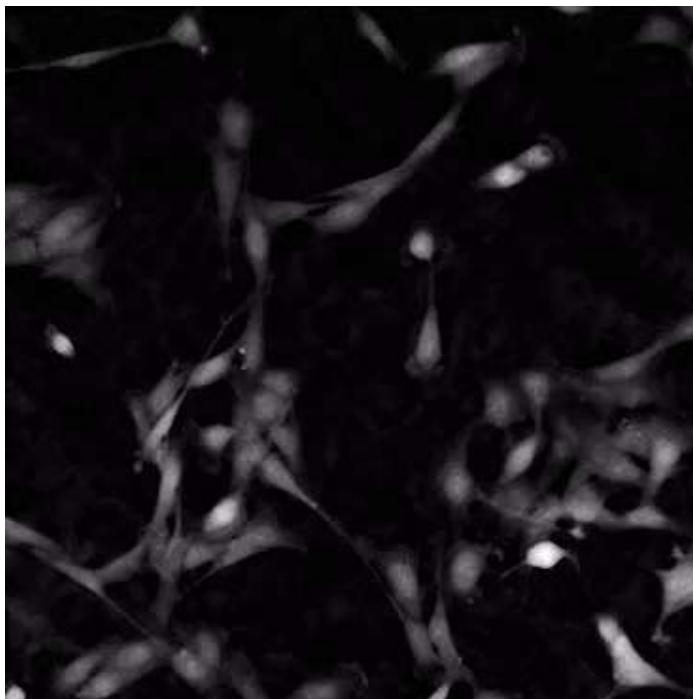
Segmentation



Uses a collection of machine learning algorithms.

Segmentation

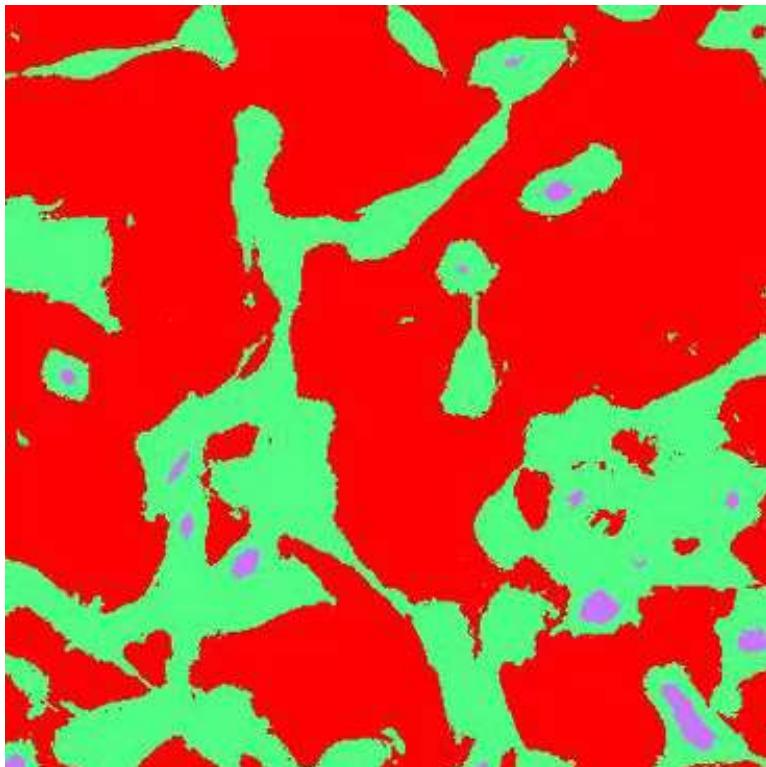
Advanced Weka Segmentation:



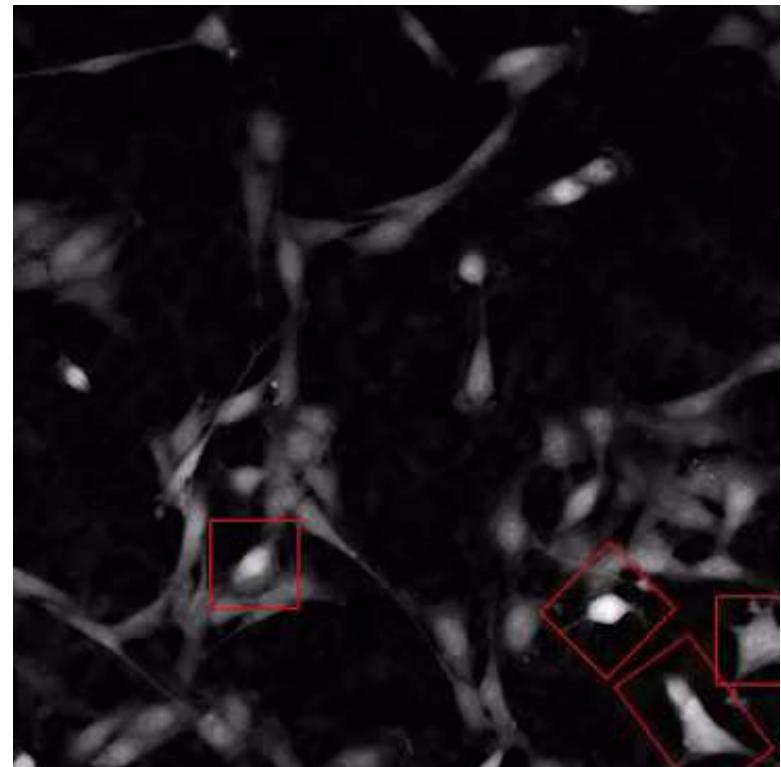
Train classifier

Segmentation

Advanced Weka Segmentation:

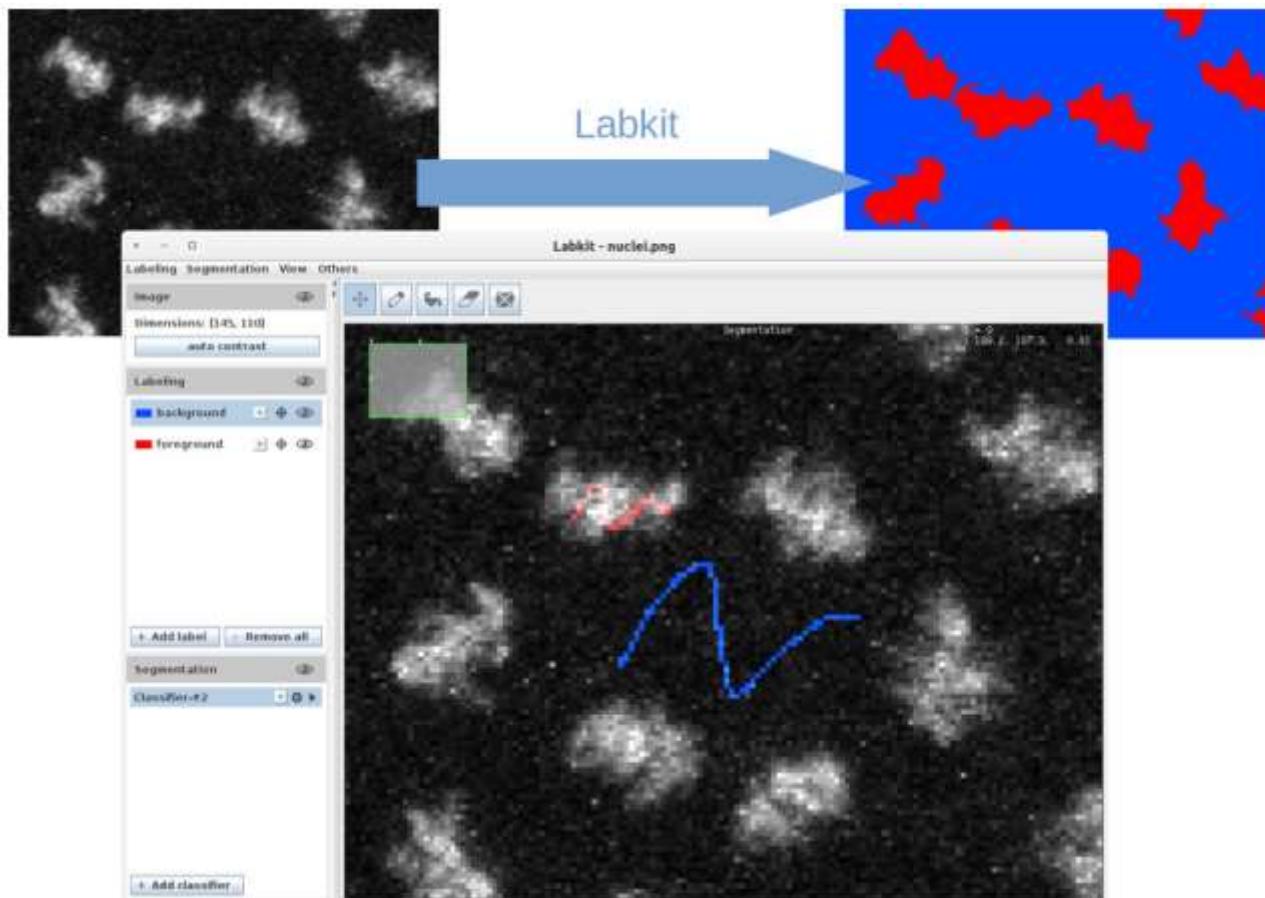


Create results



Extract information

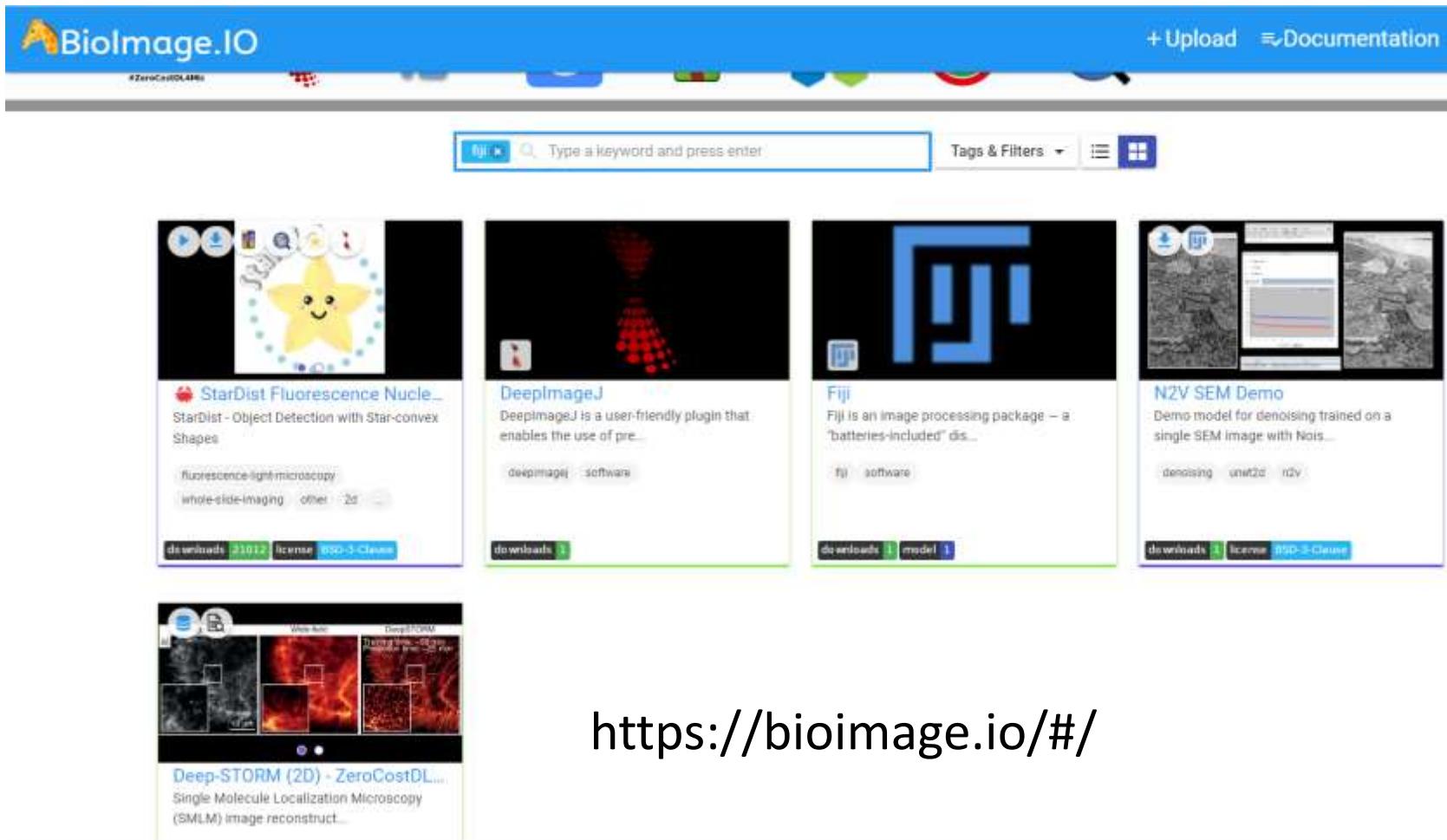
Segmentation



Segmentation



Segmentation



The screenshot shows the BioImage.IO platform interface. At the top, there's a navigation bar with a search bar, a 'Tags & Filters' dropdown, and a grid icon. Below the header, there are four main card-based components showcasing different segmentation projects:

- StarDist Fluorescence Nucleus Segmentation**: A card featuring a cartoon star with a smiling face. It includes a brief description: "StarDist - Object Detection with Star-convex Shapes", and tags: "fluorescence-light-microscopy", "whole-slide-imaging", "other", and "2d". It has a "downloads" button (21012) and a "license" button (BSD-3-Clause).
- DeepImageJ**: A card showing a red 3D reconstruction of a cell structure. It includes a brief description: "DeepImageJ is a user-friendly plugin that enables the use of pre..." and tags: "deepimagej", "software". It has a "downloads" button (1).
- Fiji**: A card featuring a blue logo consisting of a stylized letter 'f'. It includes a brief description: "Fiji is an image processing package — a 'batteries-included' dis..." and tags: "fiji", "software". It has a "downloads" button (1) and a "model" button (1).
- N2V SEM Demo**: A card showing a grayscale SEM image with a denoising interface. It includes a brief description: "Demo model for denoising trained on a single SEM image with Nois..." and tags: "denoising", "unet3d", "rdv". It has a "downloads" button (2) and a "license" button (BSD-3-Clause).

At the bottom left, there's a preview of another project, "Deep-STORM (2D) - ZeroCostDL", which shows a 2D reconstruction of a complex biological structure.

At the bottom center, there's a large URL: <https://bioimage.io/#/>

Segmentation

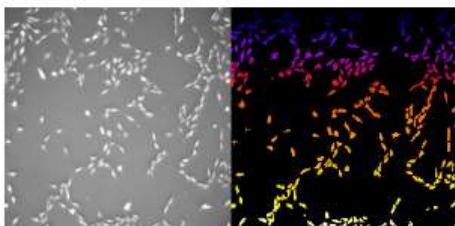
DeepImageJ is a user-friendly plugin that enables the use of a variety of pre-trained neural networks in [ImageJ](#) and [Fiji](#). The plugin bridges the gap between developers of deep-learning models and end-users in life-science applications. It favors the sharing of trained models across research groups and could have a broad impact in a variety of imaging domains. DeepImageJ does not require any deep learning expertise or any computer programmer skills.

DeepImageJ has been updated to DeepImageJ 3.0. The format of the pre-trained models are compatible with the format of the Bioimage Model Zoo. Contact us if you have any question!

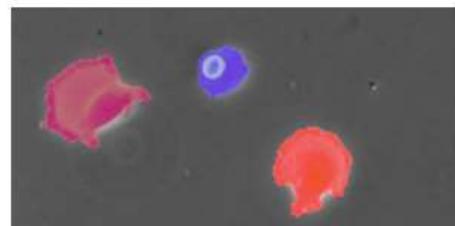
[!\[\]\(245e28142faa1131d188be32c80447b7_img.jpg\) Install DeepImageJ](#)[Try it yourself](#)[BioImage Model Zoo !\[\]\(474c03fc578d834e747854d13094d4fe_img.jpg\)](#)[Other models](#)

version 3.0

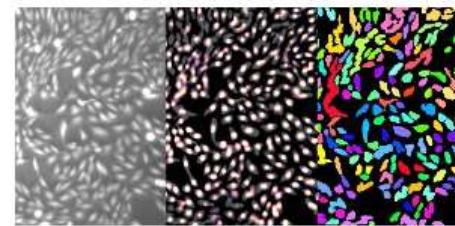
DeepImageJ trained models



U-Net Pancreatic Segmentation

[Binary Segmentation](#) [Phase Contrast](#)*Data: Cell Tracking Challenge
Training: deepImageJ & I. Arganda-Carreras*

U-Net Glioblastoma Segmentation

[Binary Segmentation](#) [Phase Contrast](#)*Data: Cell Tracking Challenge
Training: João Luis Soares Lopes (EPFL)*

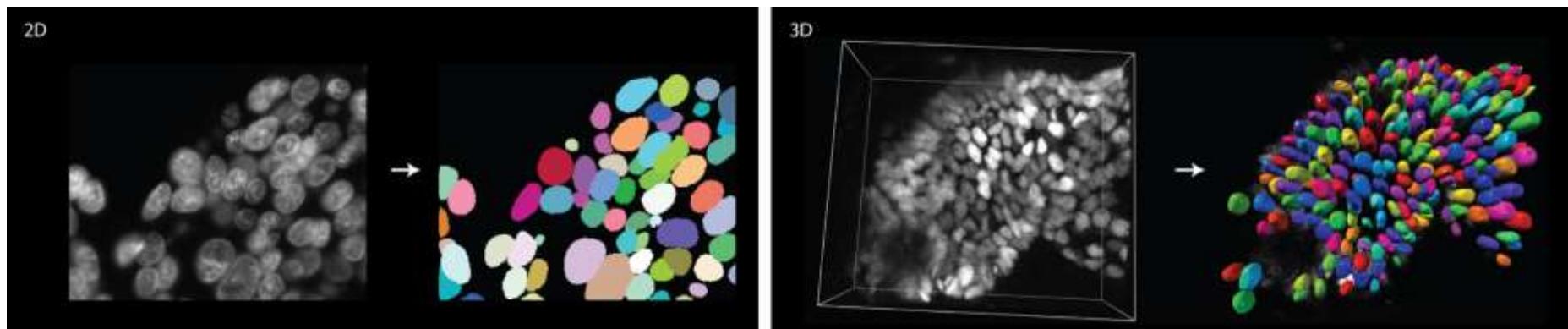
MU-Lux (CTC) PhC-C2DL-PSC cells

[Instance Segmentation](#) [Phase Contrast](#)*F. Lux & P. Matula, arXiv, 2020*

A first selection of state-of-the-art models from various groups has been made available in the [BioImage Model Zoo](#). Beyond its direct use, we expect DeepImageJ to contribute to the spread and validation of deep learning models in life-science applications.

Segmentation

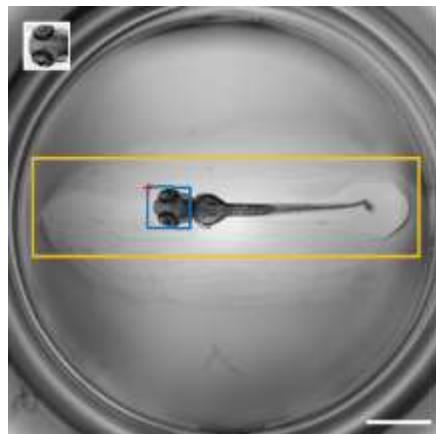
StarDist: a trained model to detect star-convex objects in 2D and 3D images.



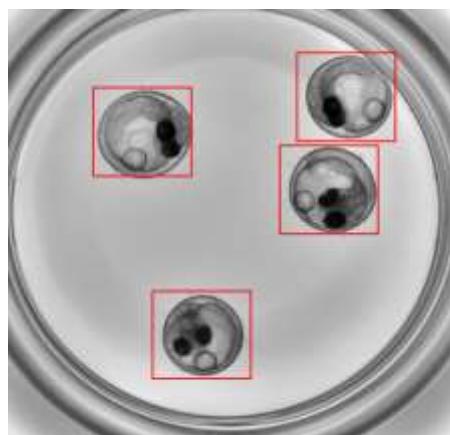
<https://github.com/stardist/stardist/>

Feature Extraction

Multi-Template Matching



Zebrafish head detection



Medaka larvae detections

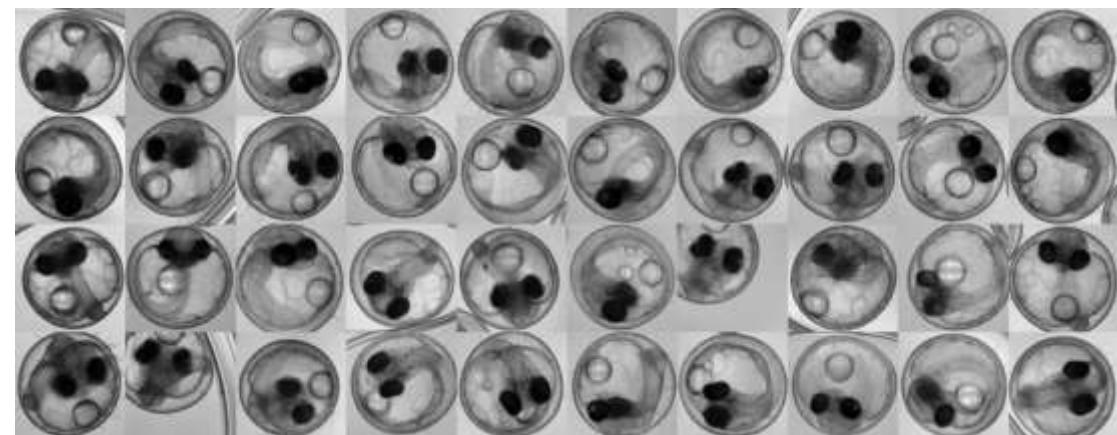
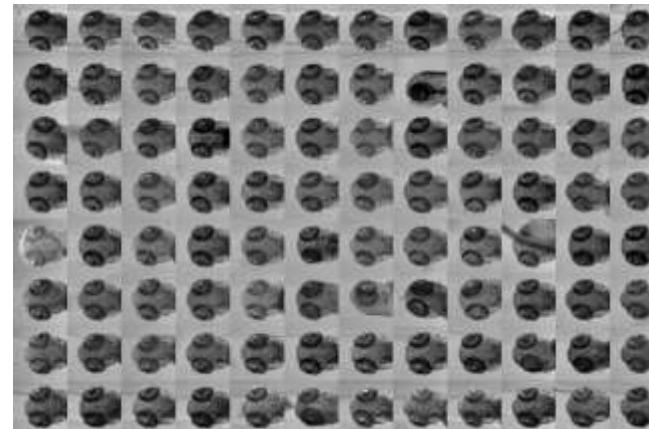
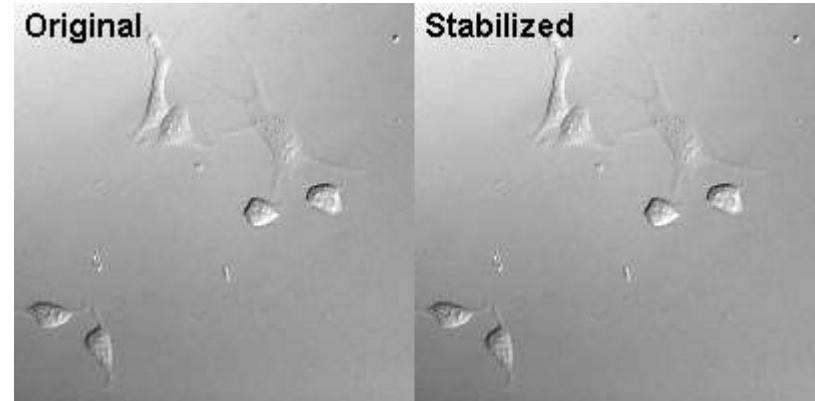


Image stabilization

Image Stabilizer: stabilizes jittery image stacks and supports both grayscale and colour images.



http://fiji.sc/wiki/index.php/Image_Stabilizer

Alternatives (for example):

StackReg + TurboReg:

Register Virtual Stack Slices

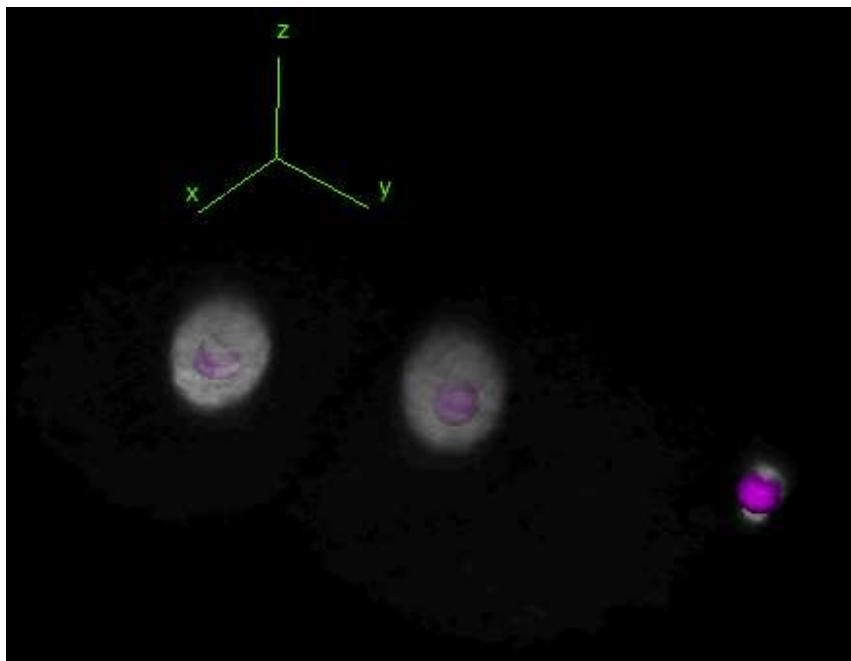
See also:

Linear Stack Alignment with SIFT

Elastic Alignment and Montage

Particle tracking

TrackMate: automatic particle tracking in 2D and 3D.



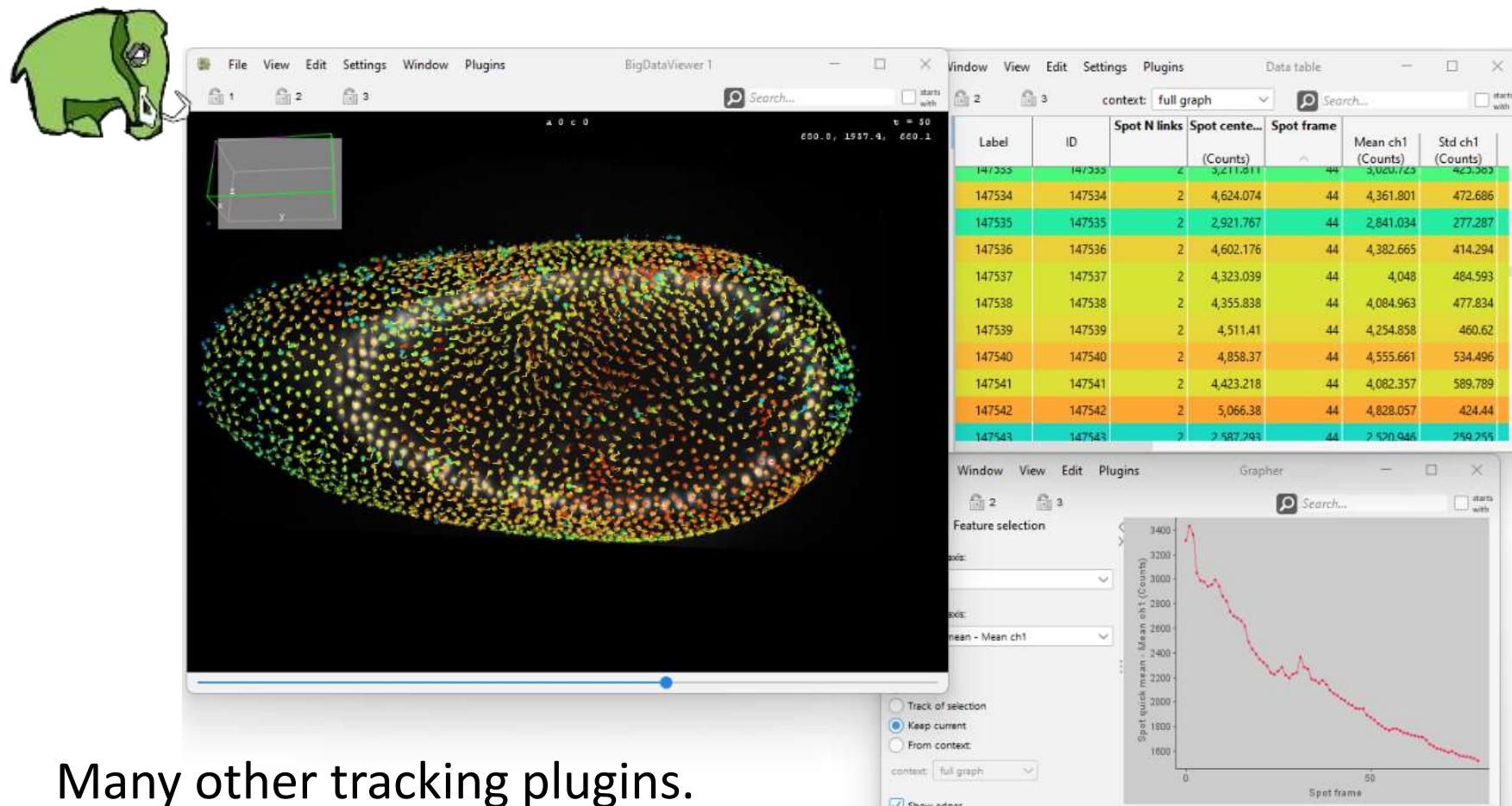
Fiji Update

<input type="checkbox"/> Tissue Analyzer	https://sites.imagej.net/TA/
<input type="checkbox"/> TomancakLab	https://sites.imagej.net/TomancakLab/
<input checked="" type="checkbox"/> TrackMate-Cellpose	https://sites.imagej.net/TrackMate-Cellpose/
<input type="checkbox"/> TrackMate-ExTrack	https://sites.imagej.net/TrackMate-ExTrack/
<input type="checkbox"/> TrackMate-Helper	https://sites.imagej.net/TrackMate-Helper/
<input type="checkbox"/> TrackMate-Ilastik	https://sites.imagej.net/TrackMate-Ilastik/
<input type="checkbox"/> TrackMate-MorphoLibJ	https://sites.imagej.net/TrackMate-MorphoLibJ/
<input type="checkbox"/> TrackMate-Oneat	https://sites.imagej.net/TrackMate-Oneat/
<input type="checkbox"/> TrackMate-StarDist	https://sites.imagej.net/TrackMate-StarDist/
<input type="checkbox"/> TrackMate-Weka	https://sites.imagej.net/TrackMate-Weka/
<input type="checkbox"/> TrackMateCSVImporter	https://sites.imagej.net/TrackMateCSVImporter/
<input type="checkbox"/> TraJClassifier	https://sites.imagej.net/TraJClassifier/
<input type="checkbox"/> Tr2d	https://sites.imagej.net/Tr2d/

The first 2 hours of a *C.elegans* embryo development, followed in 3D over time using TrackMate (strain: [AZ212](#)).

Particle tracking

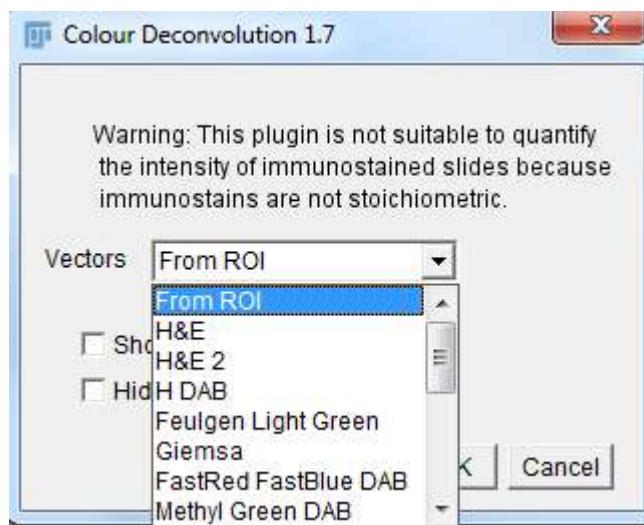
Mastodon: dealing with big data



Many other tracking plugins.

Immunohistochemistry

Image > Color > Colour Deconvolution

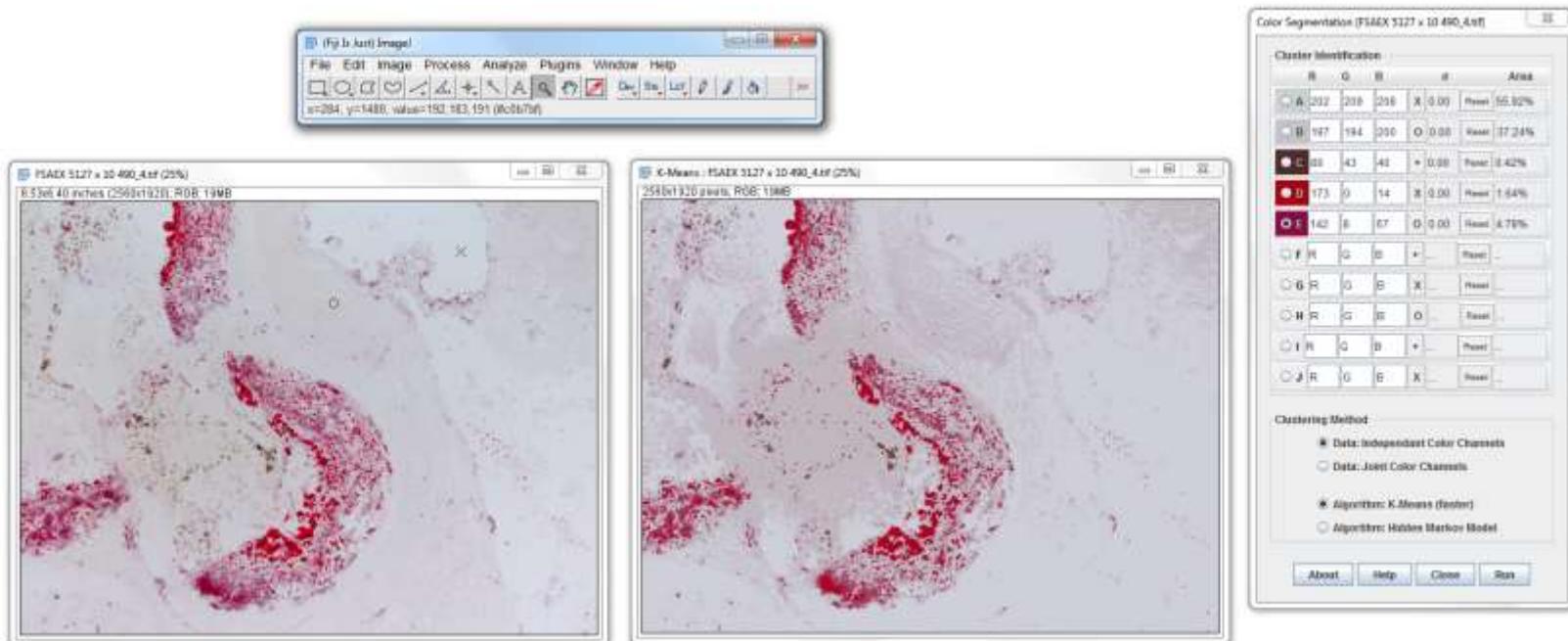


Gabriel Landini

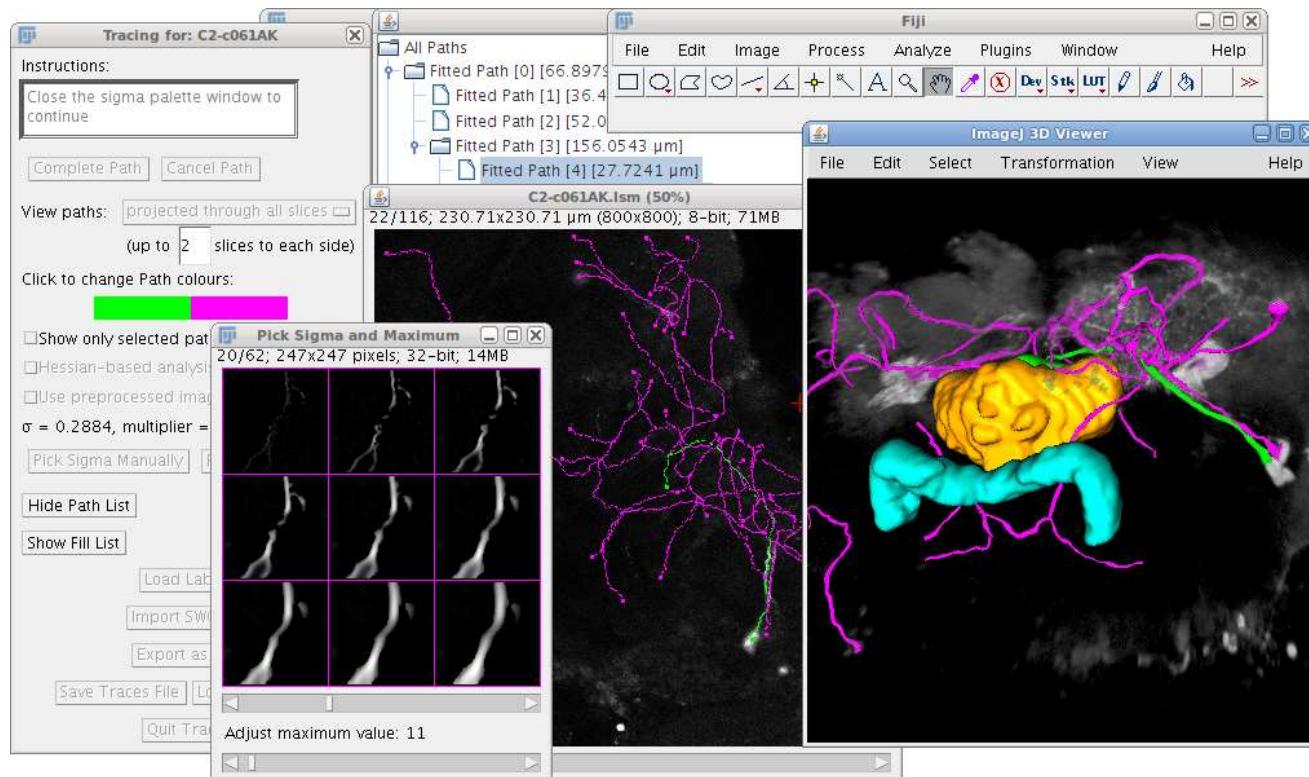
Haematoxylin and Eosin (H&E) x2
Haematoxylin and DAB (H DAB)
Feulgen Light Green
Giemsa
Fast Red, Fast Blue and DAB
Methyl green and DAB
Haematoxylin, Eosin and DAB (H&E DAB)
Haematoxylin and AEC (H AEC)
Azan-Mallory
Masson Trichrome
Alcian blue & Haematoxylin
Haematoxylin and Periodic Acid - Schiff (PAS)
Brilliant blue
RGB subtractive
CMY subtractive
User values entered by hand
Values from rectangular ROIs

Immunohistochemistry

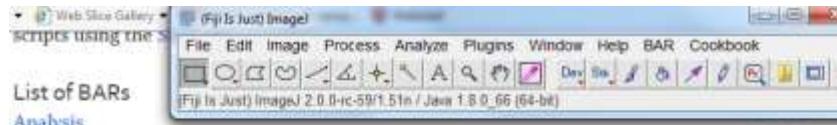
Color Segmentation



SNT: semi-automatic tracing of neurons or other tube-like structures (e.g. blood vessels) through 3D image stacks.



Bar (Broadly Applicable Routines)



Annotation

Combine Orthogonal Views, Cumulative Z Project, ROI Color Coder

Data Analysis

Create Boxplot, Create Polar Plot, Distribution Plotter, Find Peaks, Fit Polynomial, Interactive Plotting

Segmentation

Shen-Castan Edge Detector, Apply Threshold To ROI, Clear Thresholded Pixels, Remove Isolated Pixels, Threshold From Background, Wipe Background

Snippets, BAR lib and Tutorials

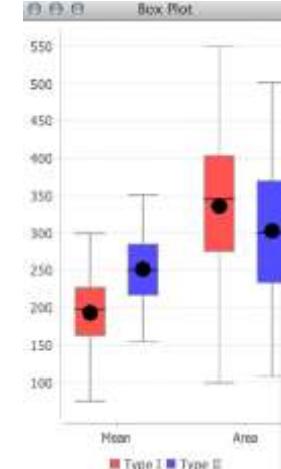
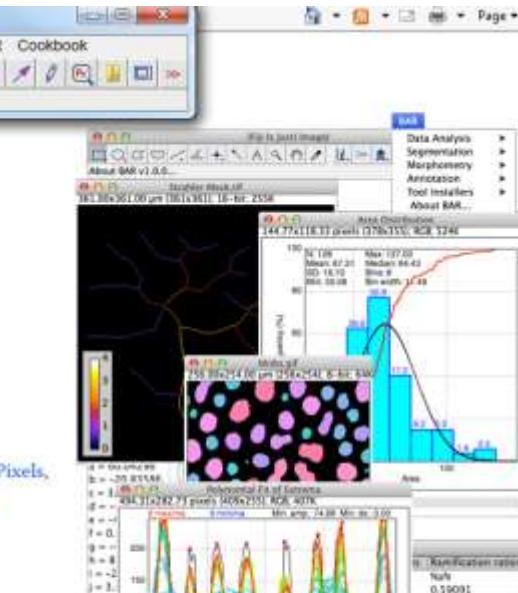
Described in Scripting BARs

Tools and Toolsets

Calibration Menu, List Folder Menu, Segment Profile, Shortcuts Menu, ROI Manager Tools, Toolset Creator

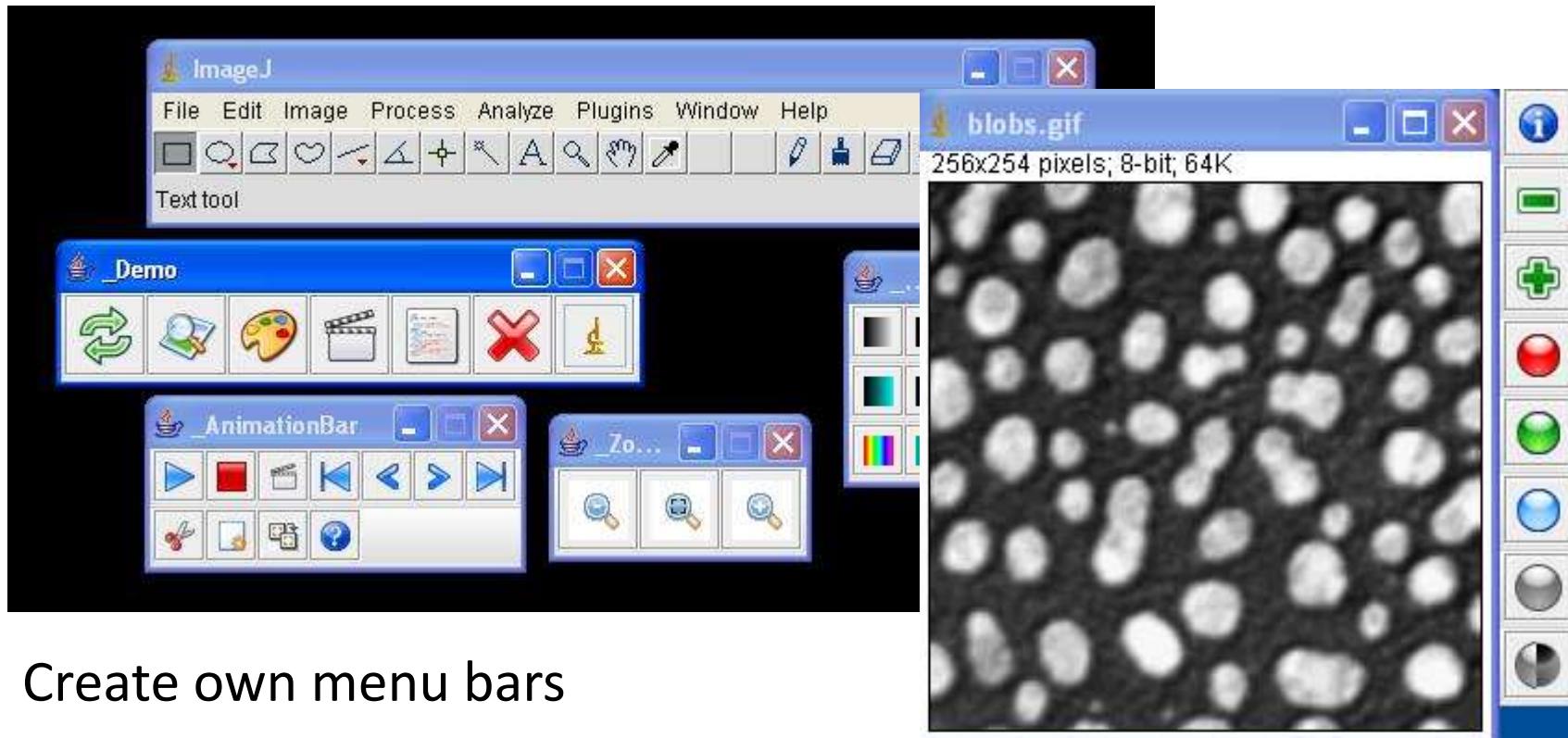
Utilities

Commander



Manage update sites				
A...	Name	URL	Host	Directory on Host
<input checked="" type="checkbox"/>	ImageJ	http://update.imagej.net/		
<input checked="" type="checkbox"/>	Fiji	http://fiji.sc/update/		
<input type="checkbox"/>	3D ImageJ Suite	http://sites.imagej.net/Tboudier/		
<input type="checkbox"/>	Angiogenesis	http://sites.imagej.net/Angiogenesis/		
<input type="checkbox"/>	Archipelago	http://sites.imagej.net/Lindsey/		
<input type="checkbox"/>	BAR	http://sites.imagej.net/Tiago/		
<input type="checkbox"/>	BigDataViewer	http://sites.imagej.net/Pietzsch/		
<input checked="" type="checkbox"/>	Bio-Formats	http://sites.imagej.net/Bio-Formats/		
<input type="checkbox"/>	Biomedgroup	http://sites.imagej.net/Biomedgroup/		
<input type="checkbox"/>	BioVoxel	http://sites.imagej.net/BioVoxel/		
<input type="checkbox"/>	Cookbook	http://sites.imagej.net/Cookbook/		
<input type="checkbox"/>	CMCI-EMBL	http://sites.imagej.net/Miura/		
<input type="checkbox"/>	CMP-BIA tools	http://sites.imagej.net/CMP-BIA/		
<input type="checkbox"/>	FFMPEG	http://fiji.sc/~schindelin/ffmpeg-plugins/		
<input type="checkbox"/>	GDSC	http://sites.imagej.net/GDSC/		

Action bar



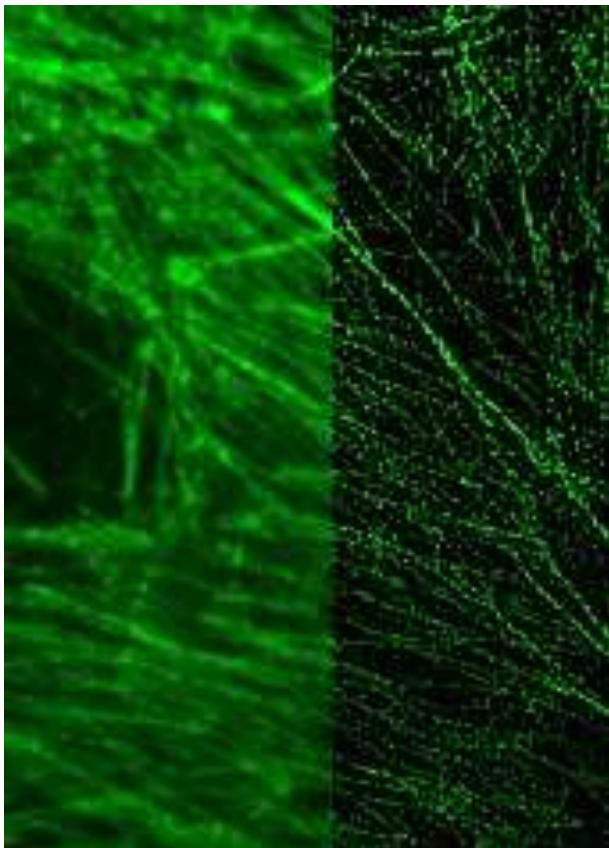
Create own menu bars

Alt-click action bar removes menu

https://figshare.com/articles/dataset/Custom_toolbars_and_mini_applications_with_Action_Bar/3397603

Super-Resolution plugins

Super-Resolution Radial Fluctuations (NanoJ-SRRF plugin),



Original

SRRF

Fifty images per channel were taken on the VisiTech confocal laser microscope with an exposure time of 100 ms using the 100x/1.47 NA objective.

SRRF calculations were performed using the free available ImageJ plugin nanoJ-SRRF.

No special microscope or sample preparation required!

Macros

ImageJ macro

A macro is a program that automates a series of ImageJ commands.

ImageJ has a special and relatively easy macro language.

It allows user interaction if needed.

Macros are saved as *.txt or *.ijm and are normally installed in the ImageJ subfolder “macros”.

What can a macro do for you?

Automated or semi-automated analysis of large data sets.

Automation or semi-automated of repetitive tasks.

Consistency in analysis.

Document analysis steps.

Speed up your work.

Add tools to the toolbar.

You can write simple to very complex macros.

Functions

[home](#) | [news](#) | [docs](#) | [download](#) | [plugins](#) | [resources](#) | [list](#) | [links](#)

Developer Resources

- [Macro Language \(download PDF\)](#)
- [Built-in Macro Functions](#) **(circled)**
- [Introduction to Macro Programming](#) New
- [Macros](#)
- [Macros Directory](#)
- [Examples](#)
- [Macro Tools](#)
- [Toolsets](#)
- [Scripting](#)
 - [Examples](#)
- [API Documentation \(v1.45s\)](#)
- [Browsable Source](#)
 - [Online Source](#)
 - [Source Code Archive](#)
- [Git Version Control Repository](#)
- [Writing ImageJ Plugins - A Tutorial](#)
- [Programming Tutorials \(Albert Cardona\)](#)
- [Imaging Book \(Burger and Burge\)](#)
- [UML Class Diagram \(320KB Jpeg\)](#)
- [Configuration File \(IJ_Props.txt\)](#)

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Built-in Macro Functions

[A][B][C][D][E][F][G][H][I][J][K][L][M][N][O][P] [Print List](#)
[Q][R][S][T][U][V][W][X][Y][Z]

A [[Top](#)]

abs(n)

Returns the absolute value of *n*.

acos(n)

Returns the inverse cosine (in radians) of *n*.

Array Functions

These functions operate on arrays. Refer to the [ArrayFunctions](#) macro for examples.

Array.concat(array1,array2) - Returns a new array created by joining two or more arrays or values ([examples](#)).

Array.copy(array) - Returns a copy of *array*.

Array.deleteValue(array, value) - Returns a version of *array* where all numeric or string elements in the array that contain *value* have been deleted ([examples](#)). Requires 1.52o.

Array.deleteIndex(array, index) - Returns a version of *array* where the element with the specified index has been deleted. Requires 1.52o.

Array.fill(array, value) - Assigns the specified numeric value to each element of *array*.

Array.filter(array, filter) Returns an array containing the elements of 'array' that contain 'filter'. Enclose the filter in parans to do regular expression matching. Requires 1.53f.

Array.findMaxima(array, tolerance) - Returns an array holding the peak positions (sorted with descending strength). 'Tolerance' is the minimum amplitude difference needed to separate two peaks. With v1.51n and later, there is an optional 'edgeMode' argument: 0=include edges, 1=exclude edges(default), 2=circular array. [Examples](#).

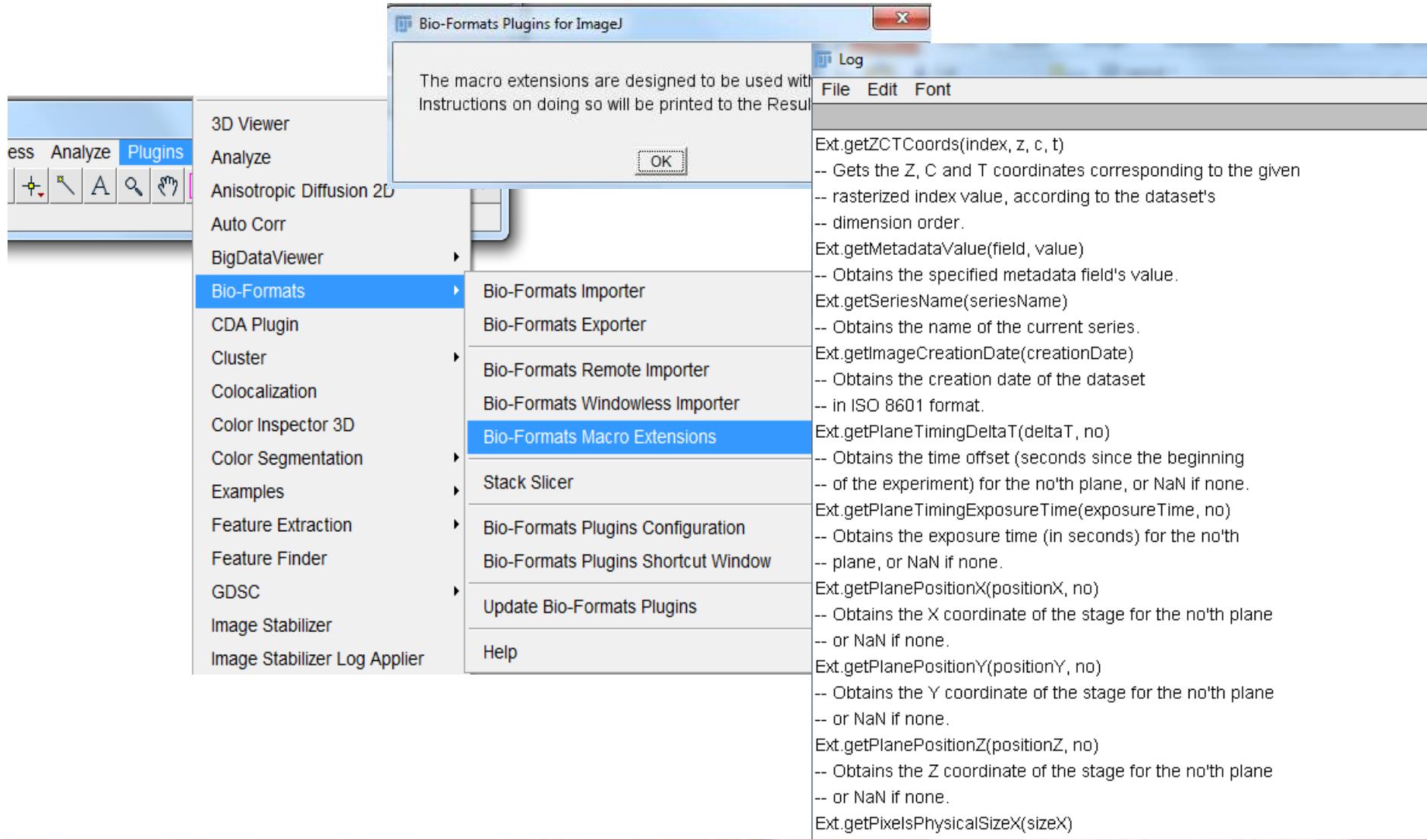
Array.findMinima(array, tolerance) - Returns an array holding the minima positions.

Macro extensions

Macro extensions are extra functions provided by a plugin that can be used in a macro. For example by:

- Bio-Formats plugin
- PDF Macro extension
- Serial Macro Extensions
- LSM Toolbox
- CLIJ2 - a GPU-accelerated image processing library

Macro extensions



The macro extensions are designed to be used with
Instructions on doing so will be printed to the Result

OK

Log

File Edit Font

- Ext.getZCTCoords(index, z, c, t)
-- Gets the Z, C and T coordinates corresponding to the given
-- rasterized index value, according to the dataset's
-- dimension order.
- Ext.getMetadataValue(field, value)
-- Obtains the specified metadata field's value.
- Ext.getSeriesName(seriesName)
-- Obtains the name of the current series.
- Ext.getImageCreationDate(creationDate)
-- Obtains the creation date of the dataset
-- in ISO 8601 format.
- Ext.getPlaneTimingDeltaT(deltaT, no)
-- Obtains the time offset (seconds since the beginning
-- of the experiment) for the no'th plane, or NaN if none.
- Ext.getPlaneTimingExposureTime(exposureTime, no)
-- Obtains the exposure time (in seconds) for the no'th
-- plane, or NaN if none.
- Ext.getPlanePositionX(positionX, no)
-- Obtains the X coordinate of the stage for the no'th plane
-- or NaN if none.
- Ext.getPlanePositionY(positionY, no)
-- Obtains the Y coordinate of the stage for the no'th plane
-- or NaN if none.
- Ext.getPlanePositionZ(positionZ, no)
-- Obtains the Z coordinate of the stage for the no'th plane
-- or NaN if none.
- Ext.getPixelsPhysicalSizeX(sizeX)

Resources

ImageJ website

[home](#) | [news](#) | [docs](#) | [download](#) | [plugins](#) | [resources](#) | [list](#) | [links](#)

Developer Resources

- [Macro Language \(download PDF\)](#)
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- Programming Tutorials (Albert Cardona)
- Imaging Book (Burger and Burge)
- UML Class Diagram (320KB Jpeg)
- Configuration File (IJ_Props.txt)

Index of /ij/macros

Name	Last modified	Size	Description
 Parent Directory		-	
 bsh/	14-Nov-2012 13:20	-	
 class/	29-Jul-2010 14:18	-	
 data/	29-Jul-2010 14:18	-	
 examples/	26-Nov-2012 22:58	-	
 images/	29-Jul-2010 14:18	-	
 js/	05-Jun-2012 14:20	-	
 misc/	12-Nov-2012 22:10	-	
 tools/	03-Mar-2012 11:07	-	
 toolsets/	18-Oct-2012 17:01	-	
 MeasureStack.txt	29-Jul-2010 14:18	193	
 ConvertSigned16BitToUnsigned.txt	29-Jul-2010 14:18	194	
 Factorials.txt	29-Jul-2010 14:18	246	
 ConvertImageToStack.txt	29-Jul-2010 14:18	266	
 OpenStackUsingURLs.txt	21-Apr-2011 23:27	274	
 build.xml	29-Jul-2010 14:18	275	
 CopyResultsToClipboard.txt	29-Jul-2010 14:18	277	
 Measure_Stack.txt	24-Jun-2012 18:31	279	
 FullRange16BitInvert.txt	29-Jul-2010 14:18	285	
 DeleteEdgeRois.txt	28-Feb-2011 15:00	286	
 ImageRotator.txt	29-Jul-2010 14:18	289	
 SplineFitForStraighten.txt	31-Oct-2011 12:13	289	
 SumAreas.txt	29-Jul-2010 14:18	308	
 About Startup Macros	29-Jul-2010 14:18	315	
 SineCosineTable.txt	08-Sep-2011 22:04	316	

Resources

There are 1000s of macro's available. Check their code to see how things are done.

Ask the mailing list.

Macro editors

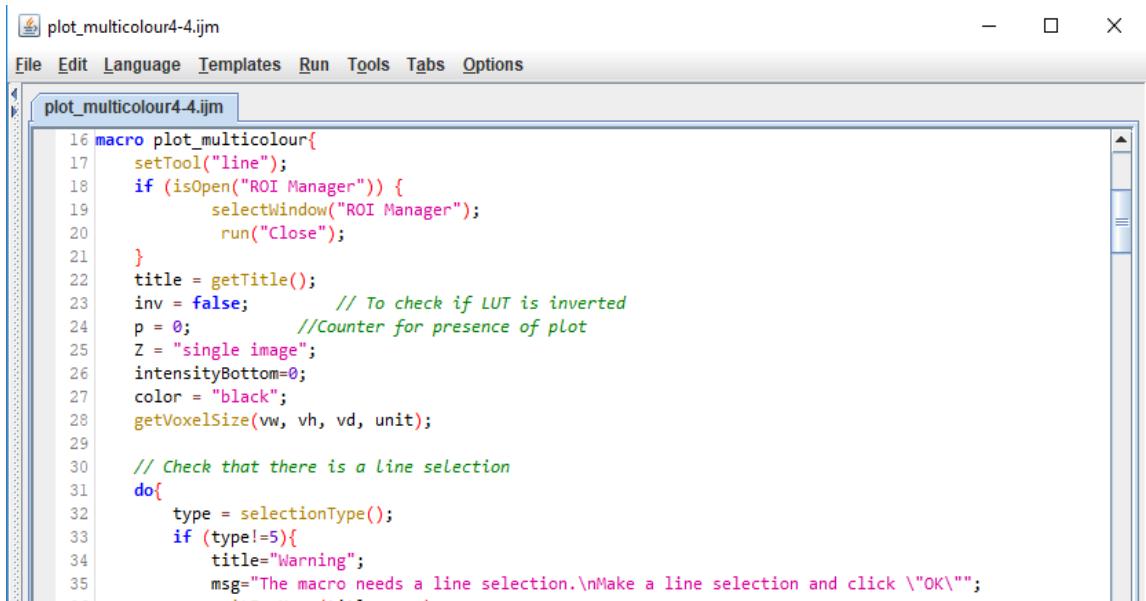
Simple text editor like Notepad

Fiji includes an editor:

File > New > Text Window OR

File > New > Script OR

Plugins > New > Macro



The screenshot shows a window titled "plot_multicolour4-4.ijm" containing a text editor with the following code:

```
16 macro plot_multicolour{
17     setTool("line");
18     if (isOpen("ROI Manager")) {
19         selectWindow("ROI Manager");
20         run("Close");
21     }
22     title = getTitle();
23     inv = false;          // To check if LUT is inverted
24     p = 0;                //Counter for presence of plot
25     Z = "single image";
26     intensityBottom=0;
27     color = "black";
28     getVoxelSize(vw, vh, vd, unit);
29
// Check that there is a line selection
30     do{
31         type = selectionType();
32         if (type!=5){
33             title="Warning";
34             msg="The macro needs a line selection.\nMake a line selection and click \"OK\"";
35             . . .
36         }
37     }while(type!=5);
38
39     . . .
40 }
```

Macro editors



The screenshot shows the homepage of the JIPipe website. At the top, there is a navigation bar with links for Home, Download, Examples, Tutorials, Documentation, Citation, and Community. The main title "JIPipe" is displayed prominently with a stylized logo consisting of two interlocking arrows forming a "J" shape. Below the title, the tagline "Macro programming for everyone!" is visible. A descriptive text block states: "JIPipe is a visual programming language for [ImageJ](#) that allows you to create macros **without any programming** by just creating a flowchart!". It also mentions the "Latest JIPipe version: Version 1.80.0" with a "Read more" link. At the bottom, there are three calls-to-action: "Download now" (in green), "Learn how to use it", and "Show examples". The footer includes the "nature methods" logo and links for "Read the publication" and "Supplementary information".

JIPipe is a visual programming language for [ImageJ](#) that allows you to create macros **without any programming** by just creating a flowchart!

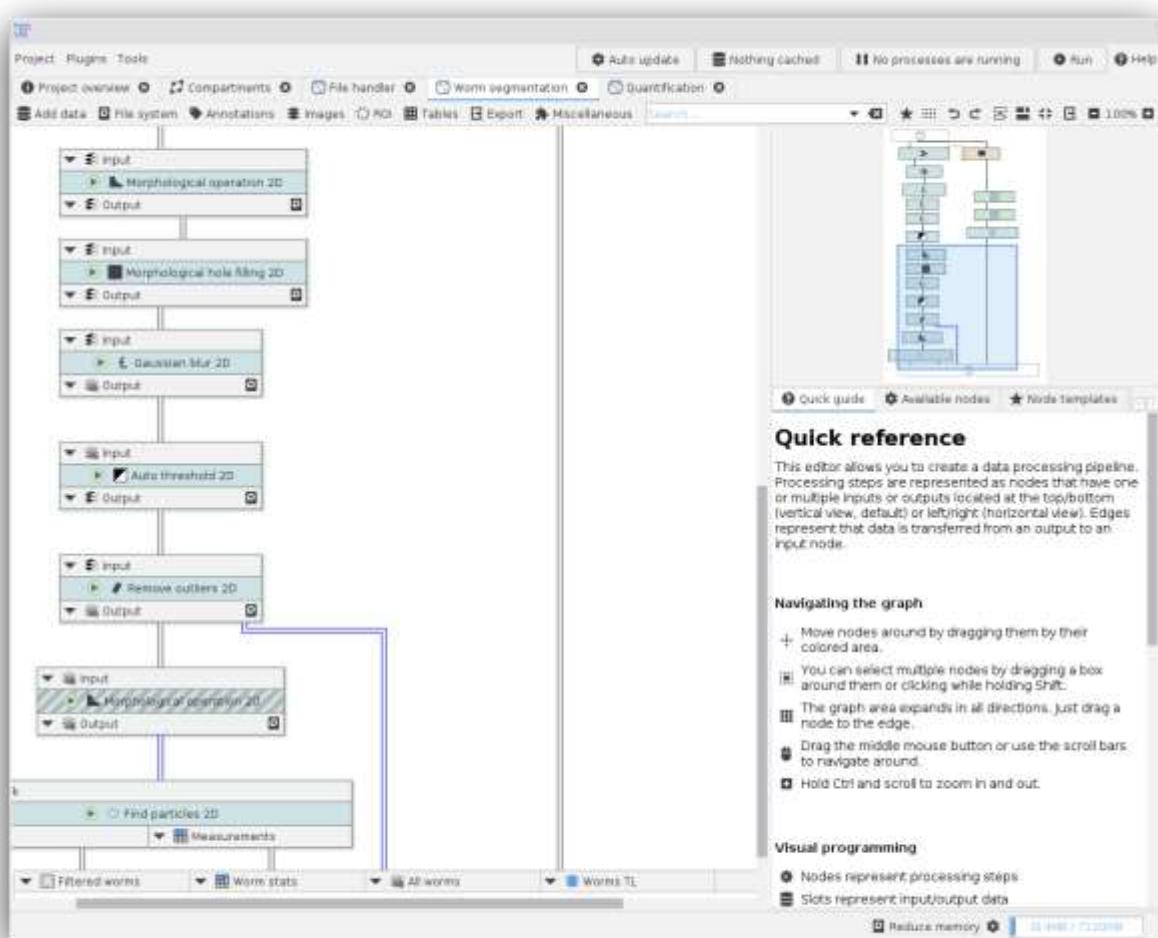
Latest JIPipe version: Version 1.80.0 [Read more](#)

[Download now](#) [Learn how to use it](#) [Show examples](#)

nature methods [Read the publication](#) [Supplementary information](#)

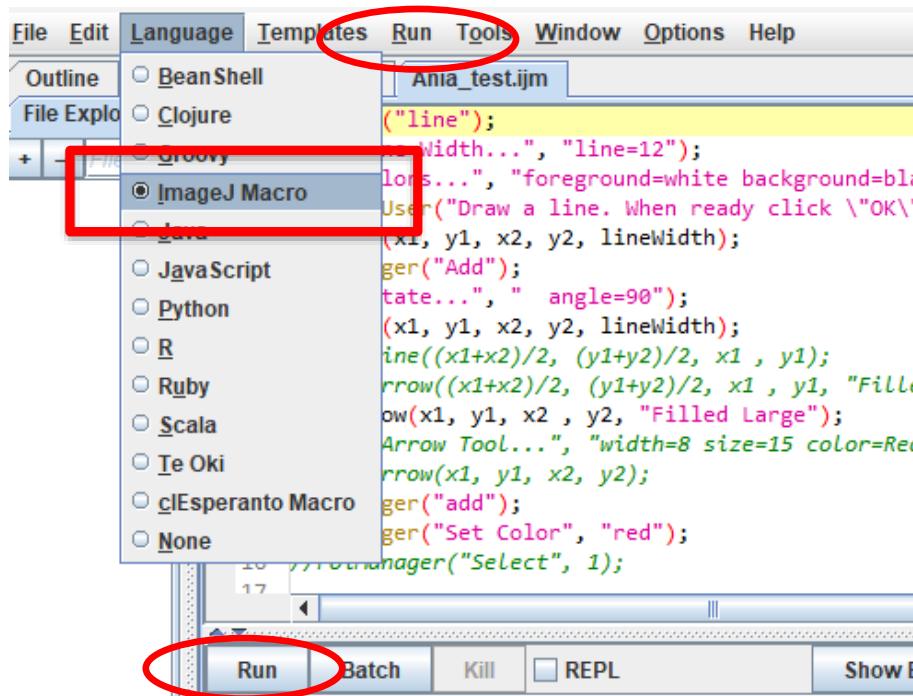
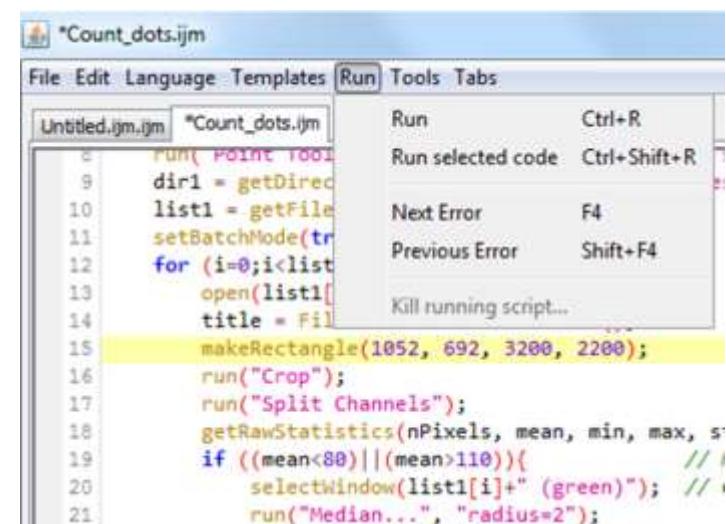
JIPipe

Plugin that adds
macro visual
programming to Fiji



Run a macro

Using *File > Open* from the editor menu will open a macro in the Text Window. If under language “IJ1 Macro” is selected, select “Run” from the menu.

The screenshot shows the Text Window with the menu bar File, Edit, Language, Templates, Run, Tools, and Tabs. The Run menu is open, showing the following options:

Run	Ctrl+R
Run selected code	Ctrl+Shift+R
Next Error	F4
Previous Error	Shift+F4
Kill running script...	

The main text area contains a portion of the same ImageJ macro script, with line 15 highlighted in yellow:

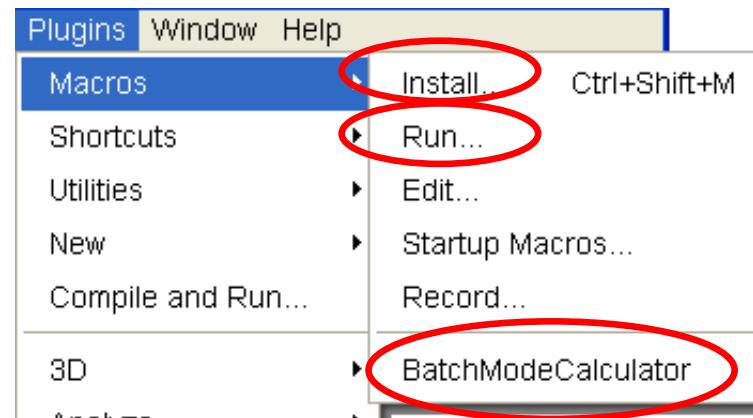
```

  8   run("Point Tool");
  9   dir1 = getDirec
10   list1 = getFile
11   setBatchMode(true);
12   for (i=0;i<list1.length();
13       open(list1[i]);
14       title = Fil
15       makeRectangle(1052, 692, 3200, 2200);
16       run("Crop");
17       run("Split Channels");
18       getRawStatistics(nPixels, mean, min, max, s
19       if ((mean<80)|| (mean>110)){
20           selectWindow(list1[i]+ (green));
21           run("Median...", "radius=2");
  
```

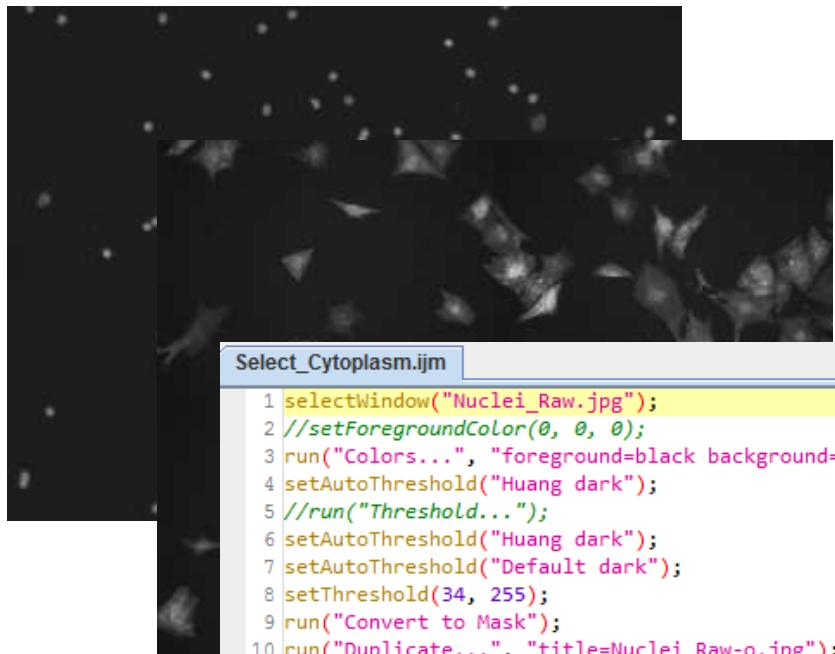
Run a macro

Using *Plugins > Macros > Install* and the macro will appear in the dropdown menu ready to be activated.

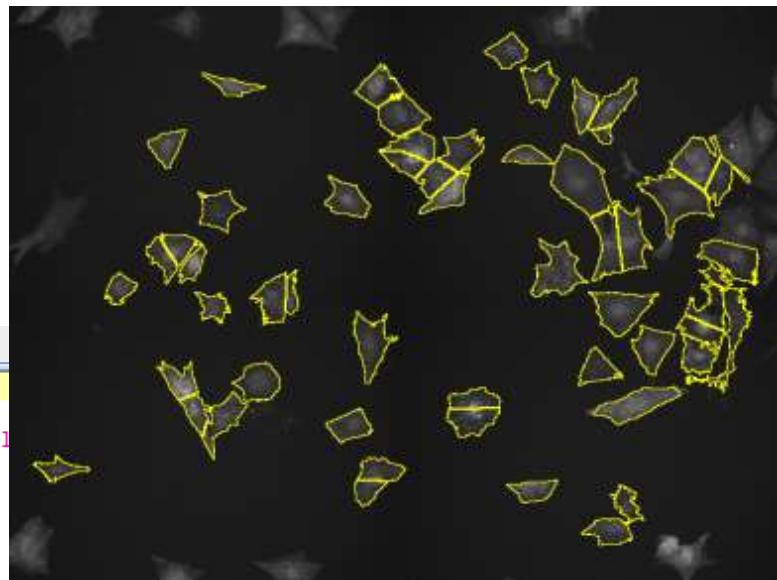
Using *Plugins > Macros > Run* and select the macro.



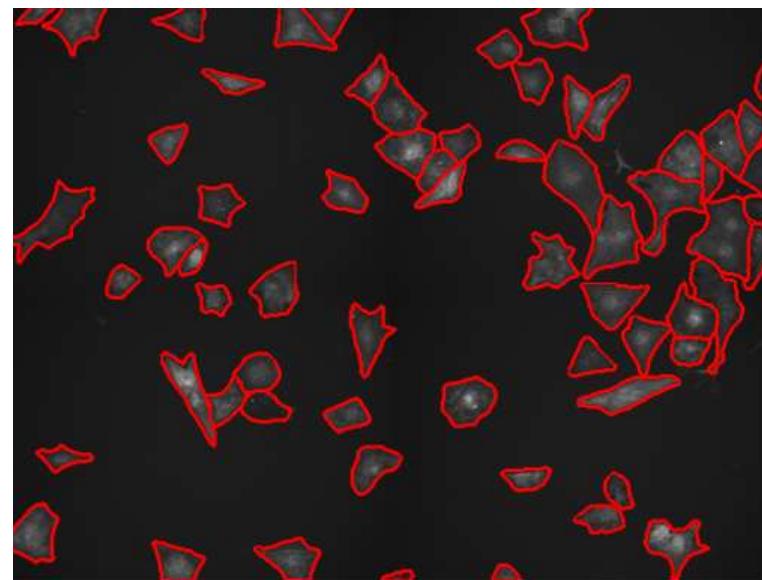
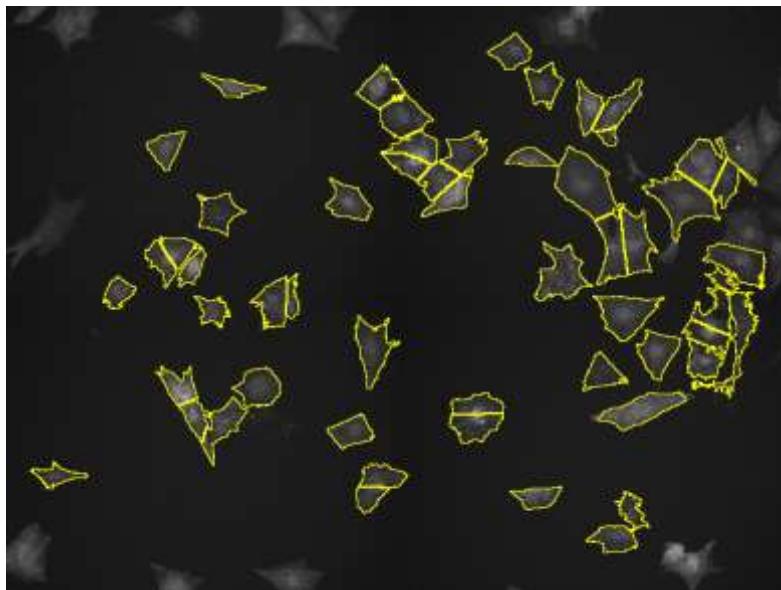
Example



```
Select_Cytoplasm.ijm
1 selectWindow("Nuclei_Raw.jpg");
2 //setForegroundColor(0, 0, 0);
3 run("Colors...", "foreground=black background=white selection");
4 setAutoThreshold("Huang dark");
5 //run("Threshold...");
6 setAutoThreshold("Huang dark");
7 setAutoThreshold("Default dark");
8 setThreshold(34, 255);
9 run("Convert to Mask");
10 run("Duplicate...", "title=Nuclei_Raw-o.jpg");
11 run("Watershed");
12 run("Voronoi");
13 setThreshold(0, 0);
14 run("Convert to Mask");
15 run("Analyze Particles...", "size=0-Infinity circularity=0.00-1");
16 run("Duplicate...", "title=Cytoplasm_Raw-o.jpg");
17 selectWindow("Cytoplasm_Raw.jpg");
18 //run("Threshold...");
19 setAutoThreshold("Default dark");
20 setThreshold(35, 255);
21 run("Convert to Mask");wait(1000);
22 selectWindow("Cytoplasm_Raw.jpg")
23 roiManager("Show All without labels");
24 roiManager("Set Color", "white");
```



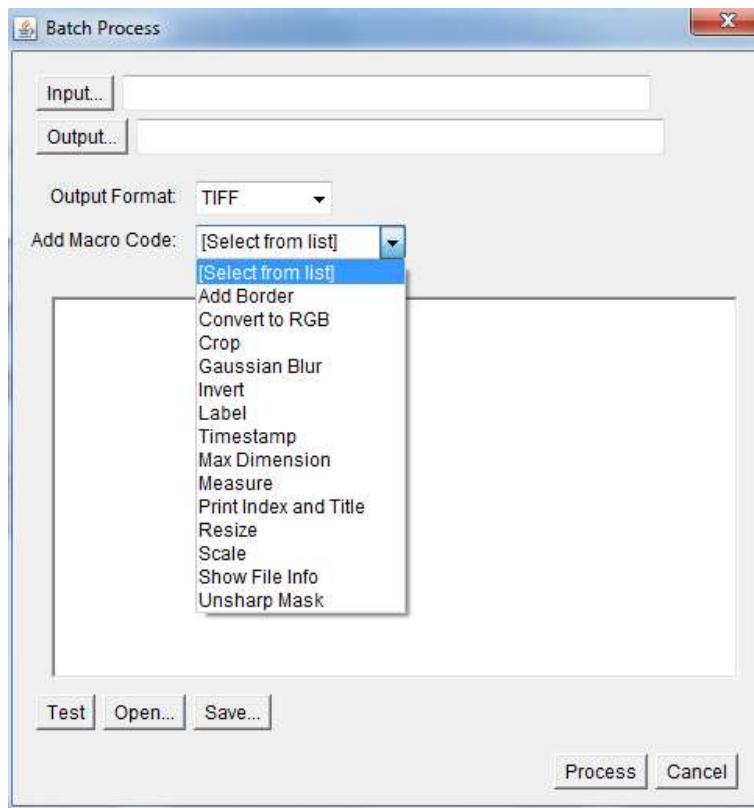
Example



Cellpose; <https://www.cellpose.org/>

Batch

- *Process > Batch > Macro:* allows to run commands on a dataset without user intervention.



Select an action
from the dropdown
menu

or

simple macro code
can be added from
the Macro recorder.

Macro recorder

Records a sequence of commands.

Also very useful to see how to pass parameters on to a plugin or macro.

IT RECORDS MANY OF YOUR ACTIONS/CLICKS!

However, the Recorder window is a text area. It is possible to change or delete commands that are not wanted in the macro.

Create a results folder

Create a results folder to save the results of the macro

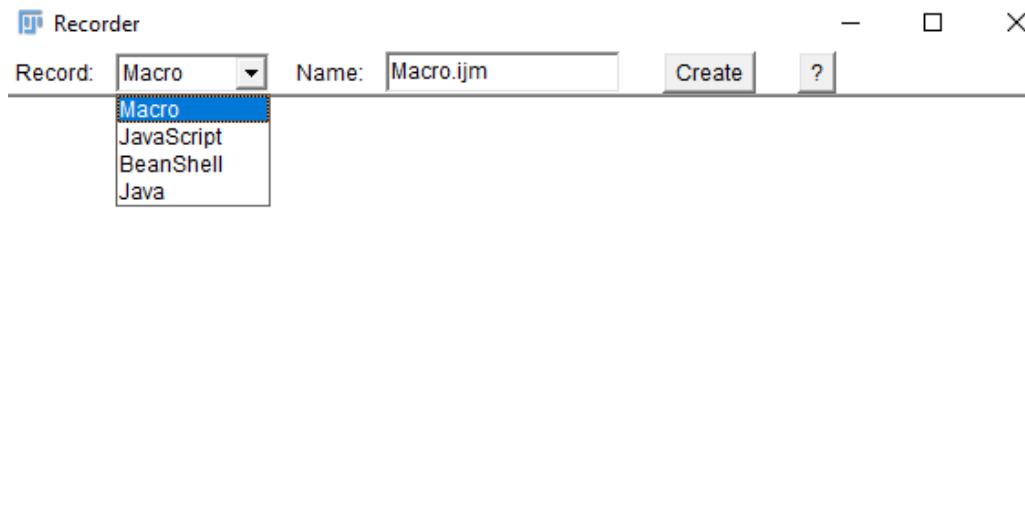
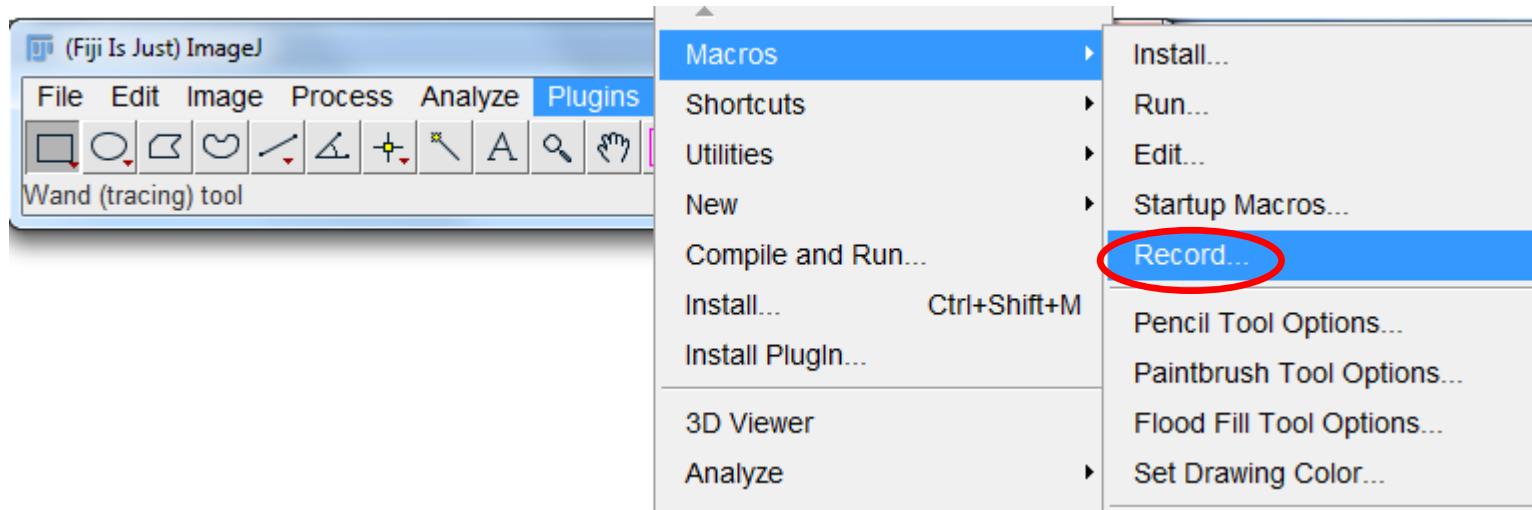
Create some macro code

Close all windows

Open the recorder (*Plugins > Macros > Recorder*)

Remember ALMOST EVERYTHING you do within ImageJ will be recorded! So, follow exactly!!

Macro recorder



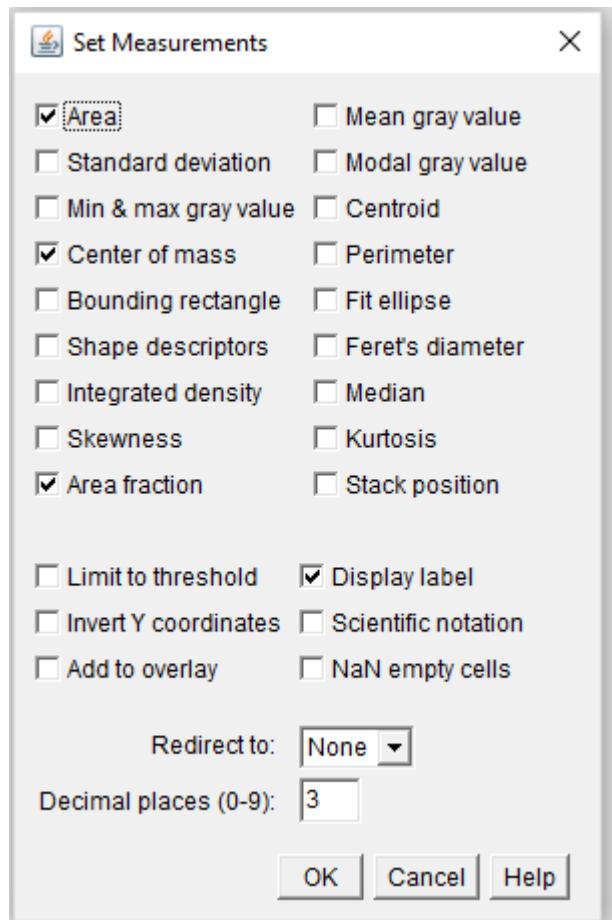
Create some macro code

File > Open Samples > Blobs(25k)

Image > Adjust > Threshold - Apply

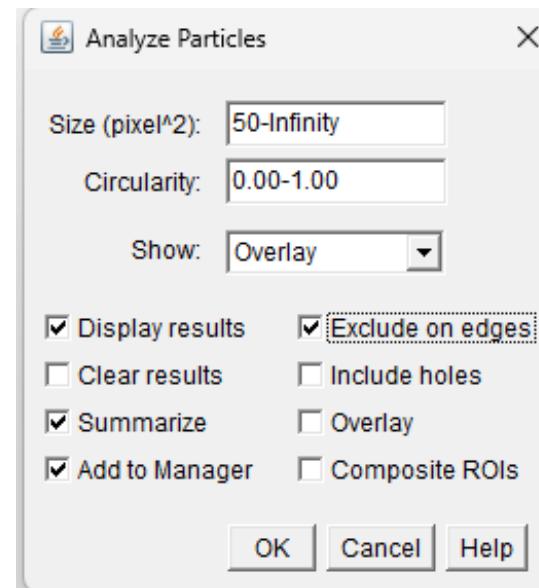
Analyze > Set Measurements

Create some macro code



Select “OK”

Analyze > Analyze Particles

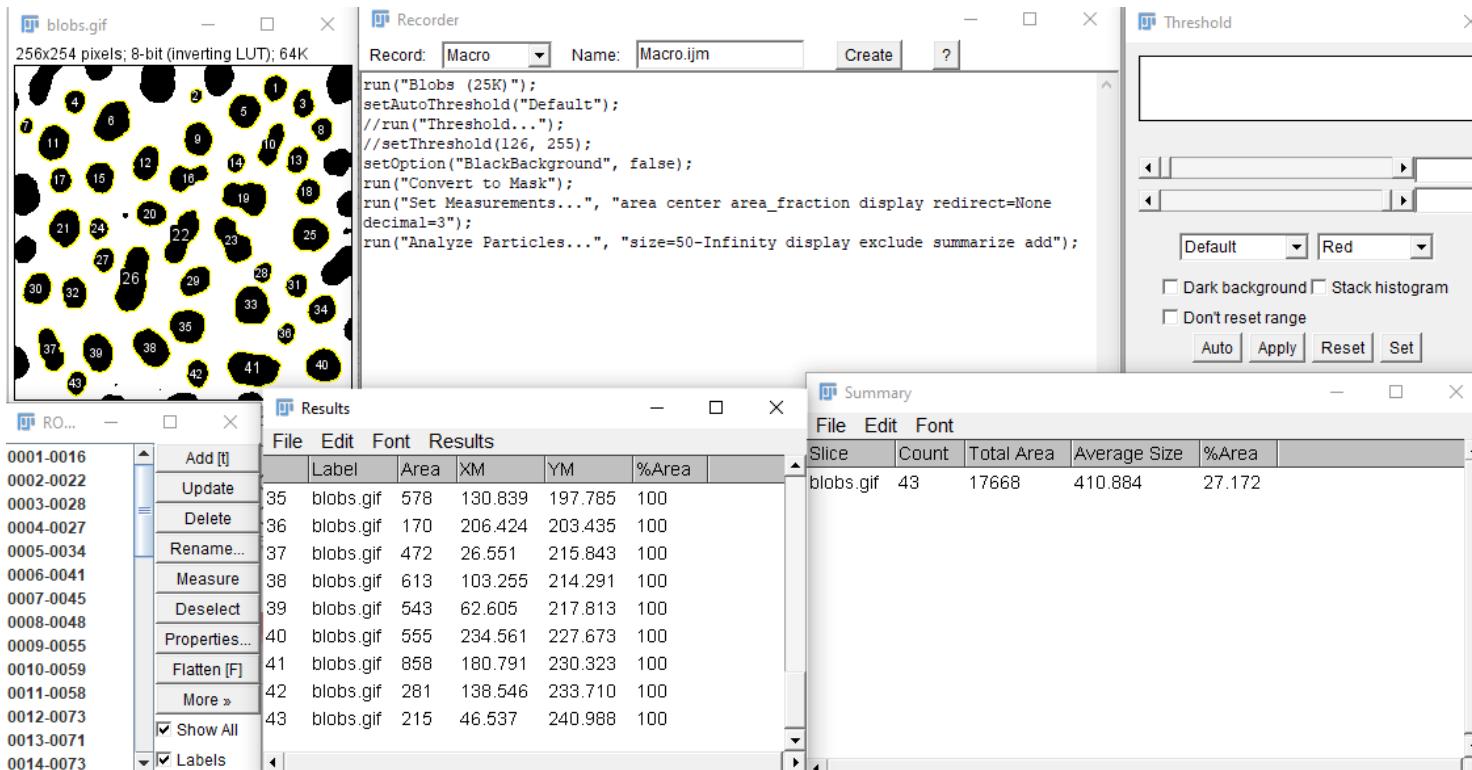


Select “OK”

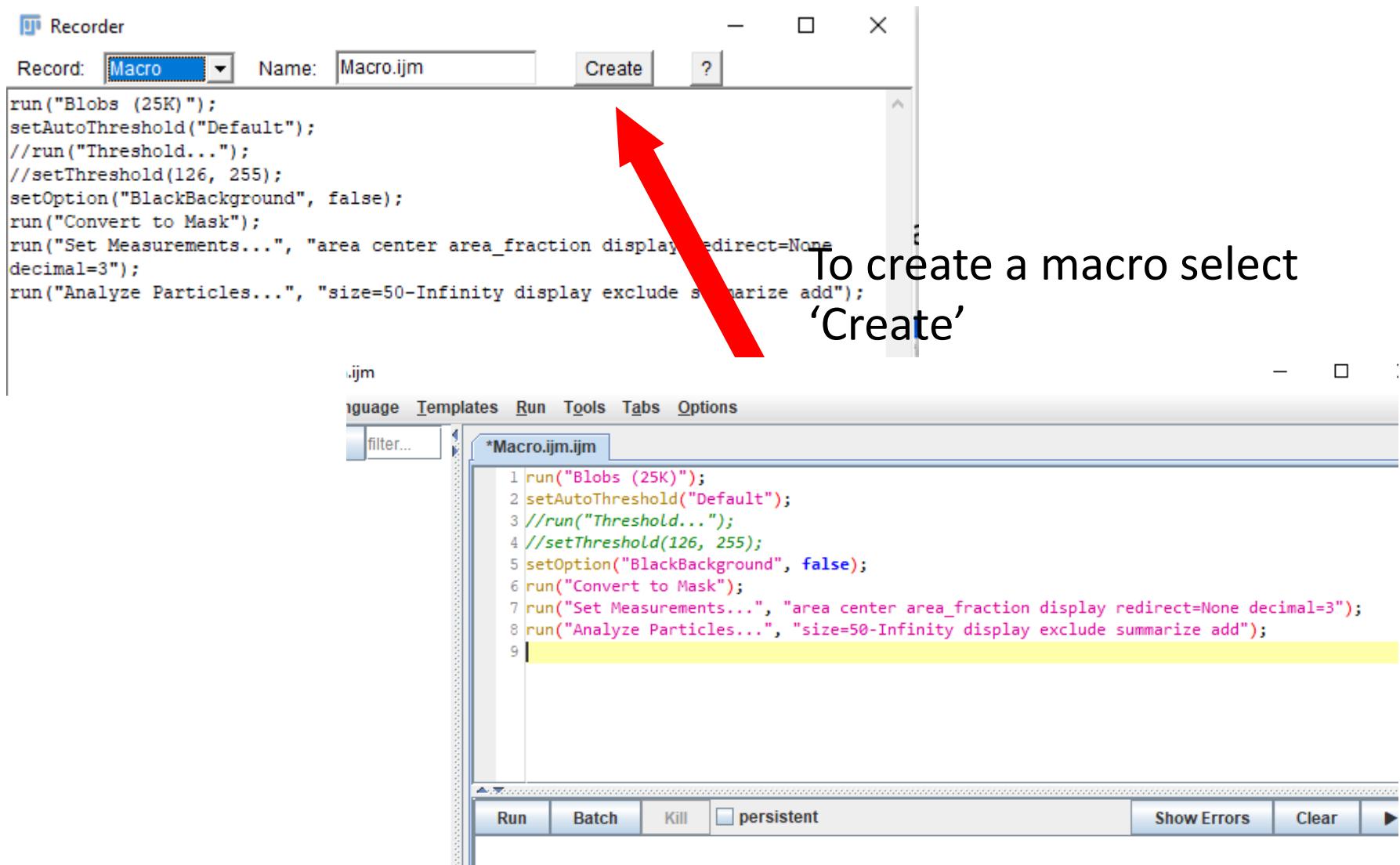
Don’t do anything else!

Create some macro code

Don't do anything else!

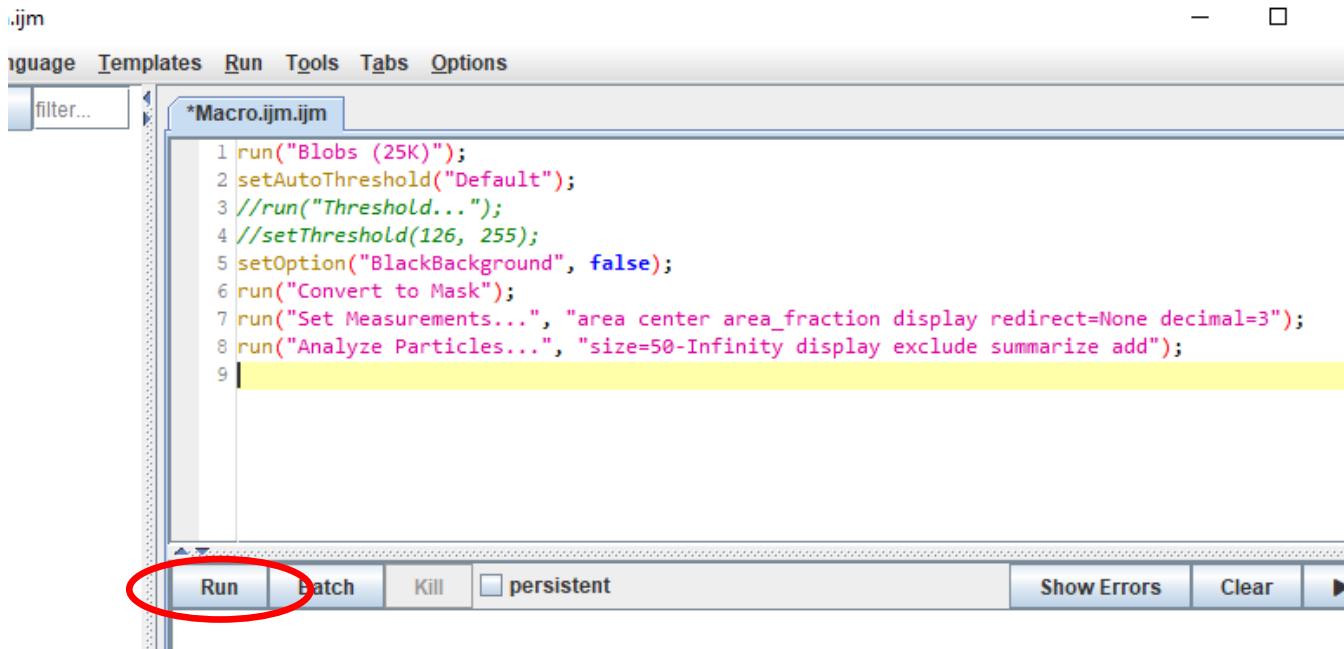


Macro recorder



Macro recorder

Close all windows except the macro editor



The screenshot shows the ImageJ macro editor window titled ".ijm". The menu bar includes File, Language, Templates, Run, Tools, Tabs, Options, and Help. A toolbar below the menu has buttons for Run, Batch, Kill, persistent (with a checked checkbox), Show Errors, Clear, and a right-pointing arrow. The main text area contains a recorded macro script:

```
1 run("Blobs (25K)");
2 setAutoThreshold("Default");
3 //run("Threshold...");
4 //setThreshold(126, 255);
5 setOption("BlackBackground", false);
6 run("Convert to Mask");
7 run("Set Measurements...", "area center area_fraction display redirect=None decimal=3");
8 run("Analyze Particles...", "size=50-Infinity display exclude summarize add");
9
```

The "Run" button in the toolbar is circled in red.

Create some test images

File > Open Samples > Blobs(25k)

Image > Duplicate... - OK

Image > Duplicate... - OK

Image > Duplicate... - OK

Save these files to a folder

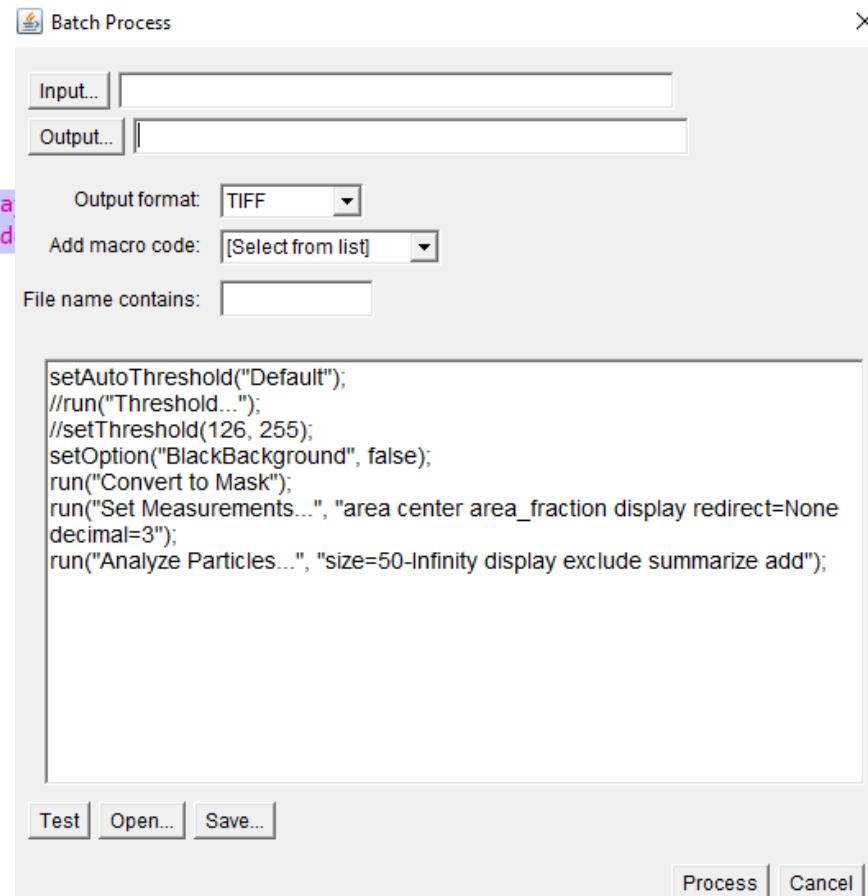
Create some macro code

File Edit Tools Help Options

Macro.ijm.ijm

```
1 run("Blobs (25K)");
2 setAutoThreshold("Default");
3 //run("Threshold... ");
4 //setThreshold(126, 255);
5 setOption("BlackBackground", false);
6 run("Convert to Mask");
7 run("Set Measurements...", "area center area_fraction display redirect=none decimal=3");
8 run("Analyze Particles...", "size=50-Infinity display exclude summarize add");
9
```

Select the code **without** the first line and copy it.



Open Batch process *Process > Batch > Macro...* and paste the code into the window.

Create some macro code

Select as “Input” the Blobs folder.

Select as “Output” a different folder (up to you).

Hit ‘Process’.

Check the output folder.

You notice that the Results, Summary table and ROI manager are not automatically saved.

