

# **Topic 01 - Installation, configuration, help and updates**

#### **Solution 1.1: Installation**

Go to the ImageJ website at http://imagej.nih.gov/ij/.



Illustration 1: The ImageJ website http://imagej.nih.gov/ij/

Click on Download.

P	Platform Independent
	o install ImageJ 1.44 on a computer with Java
p	re-installed, or to upgrade to the latest full
	istribution (including macros, plugins and LUTs),
	ownload ij144.zip (3MB) and extract the ImageJ
	irectory. Use the <i>Help&gt;Update ImageJ</i> command to upgrade o the latest pre-release version.
N	Aac OS X
D	ownload ImageJ 1.44 (5.4MB) as a double-clickable Mac
	S X application. Includes ImageJ64, which uses Java 1.6
	n 64-bit mode on Intel Macs running OS X 10.5 or later.
(	Instructions)
L	inux
	ownload ImageJ 1.44 bundled with 32-bit Java (46MB) or
	ith 64-bit Java (40MB). Both versions include Java
	.6.0_20 from Sun and the ImageJ source code. Instructions)
(	instructions)
V	Vindows
D	ownload ImageJ 1.44 bundled with 32-bit Java 1.6.0_20
	28MB), with 64-bit Java 1.6.0_20 (24MB; requires 64-bit
W	indows) or without Java (3MB). (Instructions)

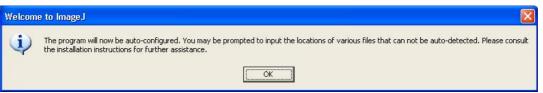
version for your system including the right java version, 32 or 64 bit.





Illustration 3: The system is 64bit and has 8GB of memory.

Look at the properties of My Computer in the windows explorer, to find out if your system is 64bit and how much memory it has. Download the ImageJ version for your system with the appropriate java included (32 or 64bit). Run the setup and answer the questions. At the end ImageJ will be started for the first time.



*Illustration 4: The first time ImageJ is started it auto-configures the thread and memory settings.* 



*Illustration 5: The configuration is* written into the file ImageJ.cfg.

Now the ImageJ-launcher window will be displayed.

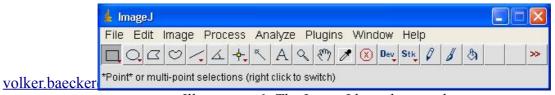
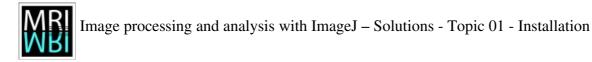


Illustration 6: The ImageJ launcher window.

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#### Solution 1.2: Memory-Configuration

Look at the system properties how much memory your machine has, then enter <sup>3</sup>/<sub>4</sub> of it in the memory options:

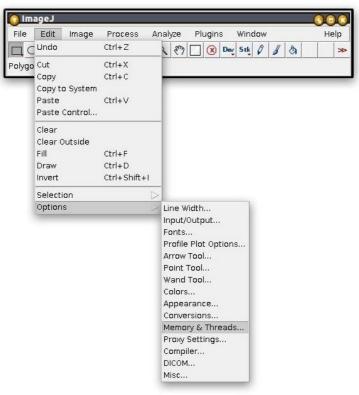


Illustration 7: Run Edit>Options>Memory & Threads...

🜔 Memory (32-bit)		00
Maximum memory:	2250	MB
Parallel threads for stacks:	2	
□Keep multiple undo buffe	rs	
<b>₩</b> Run garbage collector or	ı status	bar click
ОК	Cancel	Help

Illustration 8: The memory and threads settings. The machine has 3GB RAM and two cores.

If for example your machine has 3GB memory, you should enter  $\frac{3}{4} * 3000$ MB = 2250MB.



The content of the file ImageJ.cfg in the ImageJ base folder is:

jre\bin\javaw.exe -Xmx2250m -cp ij.jar ij.ImageJ

## Solution 1.3: Update

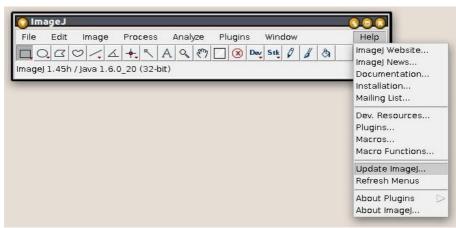


Illustration 9: The Help>Update ImageJ menu.



Illustration 10: You can either choose the current version, an older version or the daily build.

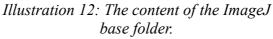




Illustration 11: The about-dialog shows the version information.

## Solution 1.4: ImageJ folders





a)



File	Edit	Font	Macros	Debug		
// Ma	ndelbro	ot set di	Run Macro	c	Ctrl+R	-
// jerome.mutterer { // This macro draws		Evaluate Line Abort Macro Install Macros		Ctrl+Y Ctrl+I	m in by	
						// making a selectio
// fror	/ which illustrates t / from one macro t		Evaluate J	avaScript	Ctrl+J	ass arguments macro is
// ber	nchmai	rked.	Mandelbro	ot		
				ed with IJ :	L.31I and jav	a 1.3.1 jre :
		-350 Mł		.44 secs		
*.*			hz 45.			
			Mhz 1		s	
// Mai	cG4-40	0 Mbz	57	03 secs		

Illustration 13: Running a macro from the macro-editor.

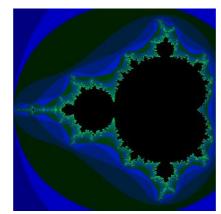


Illustration 14: The result of running the Mandelbrot-Macro.

b)



File Edit Image Process Analyze	Plugins Wir	ndow	Help
Developer Menu	Macros Shortcuts Utilities New Compile and Ru	.n	×
	3D Analyze	$\bigtriangleup$	
	Examples Filters Graphics Input-Output	-Macr -Scrip P Der	ts D mo
	LOCI LoG3D Macros		
	Process Segmentation Stacks		
	Tools	$\triangleright$	

c) Instead of running the macro, the text file is opened in the macro-editor.

## Solution 1.5: Opening images

a)

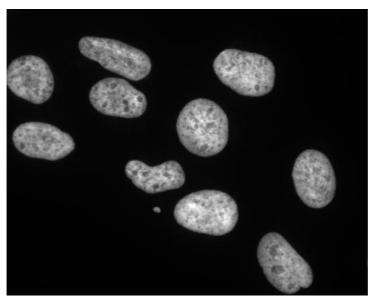


Illustration 15: The image A4 dapi1.tif.

b)



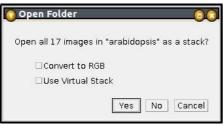


Illustration 16: The open folder dialog.



Illustration 17: The image-stack created from the images in the Arabidopsis folder.

Sequence	Option	S	00
Number of	images:	17	
Starting	image:	1	
Inc	rement:	1	
Scale	ima <b>ge</b> s:	100	%
File name co	ontains:		
or enter p	pattern:		
Conve	rt to RGE	3	
Sort n	ames nu	merica	ally
□Use vir	rtual sta	ck	
352 x 28	0×17(6	6.4MB)	
	ок с	ancel	Help

Illustration 18: A dialog that allows to import image-sequences.







d) The second time ImageJ adds -1 to the name to avoid confusion. For example opening the Image *A4 dapi 1.tif* twice, results in one image with the title *A4 dapi 1.tif* and another one with the title *A4 dapi 1-1.tif*.

e)

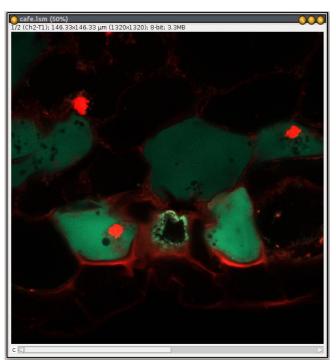


Illustration 19: The image cafe.lsm displayed in composite-mode.

f)



Stack viewing -		— Met	adata viewing	Information
View stack with:	Hyperstack		isplay metadata	Display metadata - Reads metadata that
Stack order:	XYCZT		isplay OME-XML metadata	may be contained within the file format and displays it. You can save it as a text file or
		D	isplay ROIs	copy it from the File and Edit menus specific to the "Original Metadata" window.
Dataset organi	zation	— Mer	nory management ——	<ul> <li>Readability depends upon the manner in which metadata is formatted in the data</li> </ul>
Group files with	n similar names	ΠU	se virtual stack	source. The metadata is formatted in the data
□Open files indiv	ridually	□ s	pecify range for each series	pressing "i" (Image > Show Info) when the imported image is active.
□Swap dimensio	ns	C	rop on import	imported image is active.
□Open all series				
□Concatenate s	eries when compa	tible Spli	t into separate windows	
Color options –		s	plit channels	
Color mode:	Default		plit focal planes	
□Autoscale		□S	plit timepoints	

Illustration 20: The Bio-Formats Import Options.

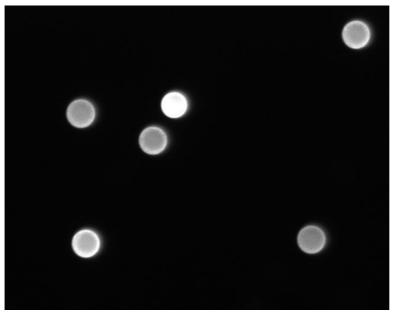


Illustration 21: The image FC green-40X sec.ics

g)



a)

Image processing and analysis with ImageJ – Solutions - Topic 01 - Installation

Miscellaneous O	ptions 🖂 🔀
Divide by zero value:	Infinity
Use pointer cu Hide "Process s	Stack?" dialog
	l key for shortcuts plugins to Misc. menu
Run single inst □Debug mode	

*Illustration 22: The misc-options dialog allows to activate the single-instance-listener.* 

In windows, select *open-with>Choose Program*... from the context menu of a tif-file. Browse to ImageJ.exe or select ImageJ Launcher if it appears already in the list. Select "Always use the selected program..." and press ok.

#### Solution 1.6: The ImageJ website

	Unsharp Mask	
	Radius (Sigma) 1.0 pixels Mask Weight (0.1-0.9) 0.60 Preview	
	Cancel	
and the second second second second second	edges by subtracting a blurred version of the image (th	e
unsharp mask) from the	original.	
Unsharp masking subtra	cts a blurred copy of the image and rescales the image	
	Lts a biurred copy of the image and rescales the image	to
	t of large (low-frequency) structures as in the input image	
obtain the same contra	.,	ge.
obtain the same contras This is equivalent to add	t of large (low-frequency) structures as in the input ima ing a high-pass filtered image and thus sharpens the in	ige. nage.
obtain the same contras This is equivalent to ado <b>Radius</b> The standard o	t of large (low-frequency) structures as in the input ima ing a high-pass filtered image and thus sharpens the in eviation ( $\sigma$ blur radius, cf. Gaussian Blur†) of the Gau	ige. nage. Issian
obtain the same contras This is equivalent to ado <b>Radius</b> The standard of blur that is subtracted.	t of large (low-frequency) structures as in the input ima- ing a high-pass filtered image and thus sharpens the im- eviation ( $\sigma$ blur radius, cf. Gaussian Blur $\uparrow$ ) of the Gau- ncreasing the Gaussian blur radius will increase contras	ige. nage. Issian t.
obtain the same contras This is equivalent to ado <b>Radius</b> The standard of blur that is subtracted. <b>Mask Weight</b> Determ	t of large (low-frequency) structures as in the input ima- ing a high-pass filtered image and thus sharpens the im- eviation ( $\sigma$ blur radius, cf. Gaussian Blur†) of the Gau- ncreasing the Gaussian blur radius will increase contras- mes the strength of filtering, whereby <i>MaskWeight</i> = 1 m	nge. nage. Issian t. would
obtain the same contras This is equivalent to add <b>Radius</b> The standard of blur that is subtracted. <b>Mask Weight</b> Determ be an infinite weight of	t of large (low-frequency) structures as in the input ima- ing a high-pass filtered image and thus sharpens the im- eviation ( $\sigma$ blur radius, cf. Gaussian Blur†) of the Gau- ncreasing the Gaussian blur radius will increase contrass hes the strength of filtering, whereby <i>MaskWeight</i> = 1 whe he high-pass filtered image that is added. Increasing the	nge. nage. Issian t. would
obtain the same contras This is equivalent to add <b>Radius</b> The standard of blur that is subtracted. <b>Mask Weight</b> Determ be an infinite weight of	t of large (low-frequency) structures as in the input ima- ing a high-pass filtered image and thus sharpens the im- eviation ( $\sigma$ blur radius, cf. Gaussian Blur†) of the Gau- ncreasing the Gaussian blur radius will increase contras- mes the strength of filtering, whereby <i>MaskWeight</i> = 1 m	nge. nage. Issian t. would
obtain the same contras This is equivalent to add <b>Radius</b> The standard of blur that is subtracted. <b>Mask Weight</b> Determ be an infinite weight of Weight value will provid	t of large (low-frequency) structures as in the input ima- ing a high-pass filtered image and thus sharpens the im- eviation ( $\sigma$ blur radius, cf. Gaussian Blur†) of the Gau- ncreasing the Gaussian blur radius will increase contrass hes the strength of filtering, whereby <i>MaskWeight</i> = 1 whe he high-pass filtered image that is added. Increasing the	ige. nage. issian t. would e <i>Mas</i> i





Illustration 24: The image before application of the unsharp mask filter.



Illustration 25: The image after application of the unsharp mask filter.

- b) Yes, the author's name is Volker Baecker.
- c) Colocalization\_Analysis, JACoP, ...
- d) I find nine plugins that have to do with colocalization:
  - Colocalization
  - Colocalization Finder
  - RG2B Colocalization
  - JACoP (Just Another Colocalization Plugin)
  - FRET and Colocalization Analyzer
  - IntraCell
  - Find Colocalized Pixels in RGB Channels



- CDA (Confined Displacement Algorithm for Colocalization)
- Colocalization Colormap
- e) It downloads an image cells.tif and counts the number of cells in the image.

File Edit Font								
Slice	Count	Total Area	Average Size	Area Fraction	Mean	Mode	IntDen	
cells-1.tif	72	14029.241	194.851	12.7	255	255	49686.894	

