

Chapter 1

Installation, configuration, help and updates

1.1 Installation

Install ImageJ from the ImageJ website <http://imagej.nih.gov/ij/>:

- go to the download section
- select the version bundled with Java that corresponds to your system (win, mac, 32bit, 64bit), if you don't know, open the windows file-explorer and right-click on my computer, then select properties from the context menu. If your system is 64bit it will be mentioned on the dialog
- download the file and execute the installer
 - you need to have write access on the folder into which you install ImageJ
 - on windows you can for the purpose of this workshop install it somewhere under **My Documents**
- run ImageJ, the first time it is started it will configure itself depending on the available memory and the number of processors of the machine

1.2 Memory-Configuration

Configure the memory and threads settings of ImageJ. ImageJ is a java application. Java applications need to be told how much memory they can use. To work with ImageJ, the more memory available the better. The number of threads tells ImageJ how many parallel threads it should use when working with stacks. The number should correspond to the number of processors of the machine.



- go to the menu **Edit>Options>Memory & Threads**
- you should set the available memory to 3/4 of the available memory on your machine
- to find out how much memory your machine has, open the properties of **My Computer** and look for the amount of available RAM or click on **start** and in the search box type "ram" without quotes and click the "Show how much RAM is on this computer"
- if you assign too much memory to ImageJ it can happen that you can't start ImageJ anymore.
 - in this case you can edit the amount of memory in the file **ImageJ.cfg** in the base folder of the ImageJ installation

1.3 Update

Update ImageJ to the latest version. At the time this document is written the latest version is 1.48t. The numbers signify major version changes while the different letters indicate minor changes.

- you can do this from the menu **Help>Update ImageJ...**
- ImageJ will be closed and you have to restart it
- you can display version information about you installation from the menu **Help>About ImageJ...**

1.4 ImageJ folders

Open the folder into which you have installed ImageJ. It contains among others:

- a file **ij.jar** – this is the ImageJ program. An update replaces this file
 - a file **ImageJ.cfg** – this file contains the memory configuration for ImageJ
 - a folder **macros** – the folder contains ImageJ macros, i.e. is small programs written in the ImageJ macro language
 - a folder **plugins** – the folder contains plugins that add functionality to ImageJ – the plugins in this folder and its sub-folders usually appear in the plugins menu of ImageJ
 - to appear in the menu the filename must contain an underscore characters ("_") or end with **.ijm** for **imagej macro**.
- a) Enter the macros folder. Drag the macro **RandomOvals.txt** onto the ImageJ launcher window. The macro will be opened in a macro-editor. Run the macro from the menu **Macros>Run Macro** of the editor.



- b) Save the macro `RandomOvals.txt` under the name `RandomOvals.ijm` into the folder `ImageJ/plugins/Examples`. You can use the command `File>Save As...` from the macro-editor. Close all open windows in ImageJ. Call `Help>Refresh Menus` from the ImageJ launcher window. Now run the macro from the menu `Plugins>Examples>RandomOvals`.
- c) Hold down `shift` and click on `Plugins>Examples>RandomOvals`. What happens?



1.5 Opening images

There are two basic possibilities to open images. You can use the menu `File>Open...` or you can drag images from your file-explorer onto the ImageJ launcher window.

`File>Open...` will open a file-dialog in which you can browse and select one or more images. The keyboard-shortcut for `File>Open...` is `ctrl+o`. Note that the shortcut will only work when an ImageJ window is active. When there is an active image, `ctrl+shift+o` will open the next image in the folder of the currently active image and close the active image.

You can use drag and drop to open one or multiple images. If you drag a folder onto the ImageJ- launcher, you will be asked if you want to open all images in the folder separately or all images as one stack.

The command `File>Import>Image Sequence...` allows to import a series of images as a stack as well. This way you have more control about which images in a folder should be imported and in which way.

ImageJ opens a number of file-formats. For other file-formats specific image-reader plugins must be installed.

Finally you can configure your system to open images with a specific file-extension by default with ImageJ. In this case you might want to select `Run single instance listener` from the `Edit>Options>Misc` menu. Double clicking will open the image with ImageJ and all images will be opened in the same ImageJ instance (instead of running one ImageJ per image).

- a) Open the image `A4 dapi 1.tif` from the folder `01 - opening images`. Try the different possibilities. Try `ctrl+shift+o` to switch to the next image in the folder. Try to open multiple images in the same time using drag and drop and by using the file-dialog.
- b) Open the images in `arabidopsis` by dragging the folder onto the ImageJ-launcher. Open them using `File>Import>Image Sequence...`



- c) Drag and drop works with image urls (online links) as well. Search an image with the web- browser, for example `http://www.mri.cnrs.fr/datas/fichiers/115.jpg` and drag it onto the ImageJ-launcher.
- d) Open the same image twice. What happens to the title of the image when the same image is opened multiple times?

- e) Open the image `cafe.lsm`. The image is opened as a so called hyper-stack. The slider on the bottom of the window changes the channel that is displayed. Run `Image>Color>Channels Tool...` and select `Composite` instead of `Color` as display mode to see an overlay of both channels.
- f) Try to open the image `FC green-40X sec.ics`. You will get an error. ImageJ doesn't know how to open the ics-format. Use `Help>Plugins` to open the ImageJ plugins website in your web-browser. Search for `loci` and click on the link. On this page click on `Bio-Formats Downloads` and drag the link `loci.tools.jar` onto the ImageJ launcher. This will open a file-dialog. Click `ok` to save the plugin into the plugins-folder. Try to open the image again. Hint: Select `Autoscale` in the color options of the `Bio-Formats Import Options` to have the image displayed correctly.
- g) Activate the single instance listener `Edit>Options>Misc...` in ImageJ and configure ImageJ as default application for tif-images in your operating system.

1.6 The ImageJ website

You can access the different parts of the ImageJ website from the help menu of ImageJ.

- a) Go to the *Documentation* part of the website and have a look at the “ImageJ User Guide”. Read the explication of the `Unsharp mask` command. How does it work? Try the command on one of the images in the `01 - opening images/arabidopsis` folder.
- b) In the Documentation part have a look at the *Tutorials and Examples*. Where does the link with the title *ImageJ Workshop (manuscript, slides and exercises)* point to?



1.6. THE IMAGEJ WEBSITE

- c) ImageJ users can ask questions in the ImageJ mailing list. Go to the list-archive. In the search text-field type *colocalization*. Find the name of at least one colocalization plugin from the answer of the search.

- d) Go to the *plugins* part of the website. How many colocalization plugins can you find on this page?

- e) Go to the *macros* part of the website. Drag the link to the `CellSegmentation.txt` macro onto the ImageJ-launcher and run the macro from the macro-editor. Do you understand what it does?