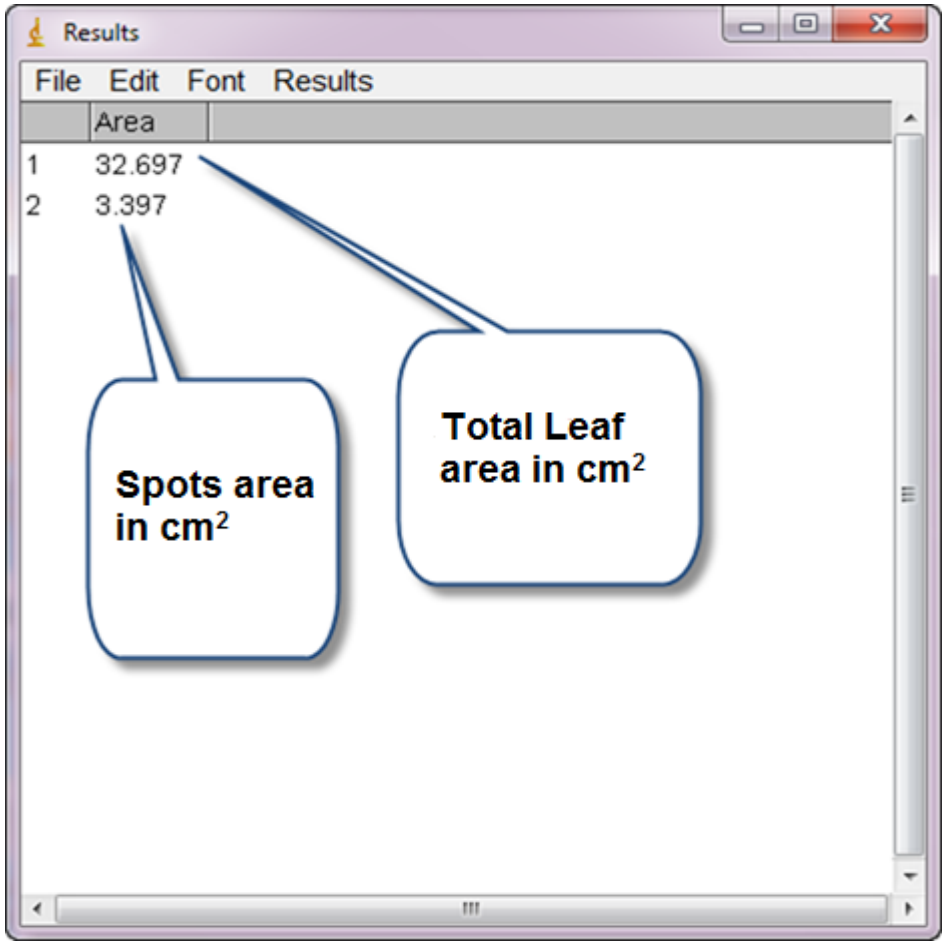
Estimating leaf spot percentage

❖ Go to **Analyze >> measure or (Ctrl+M) >>** the results will appears in the result window.

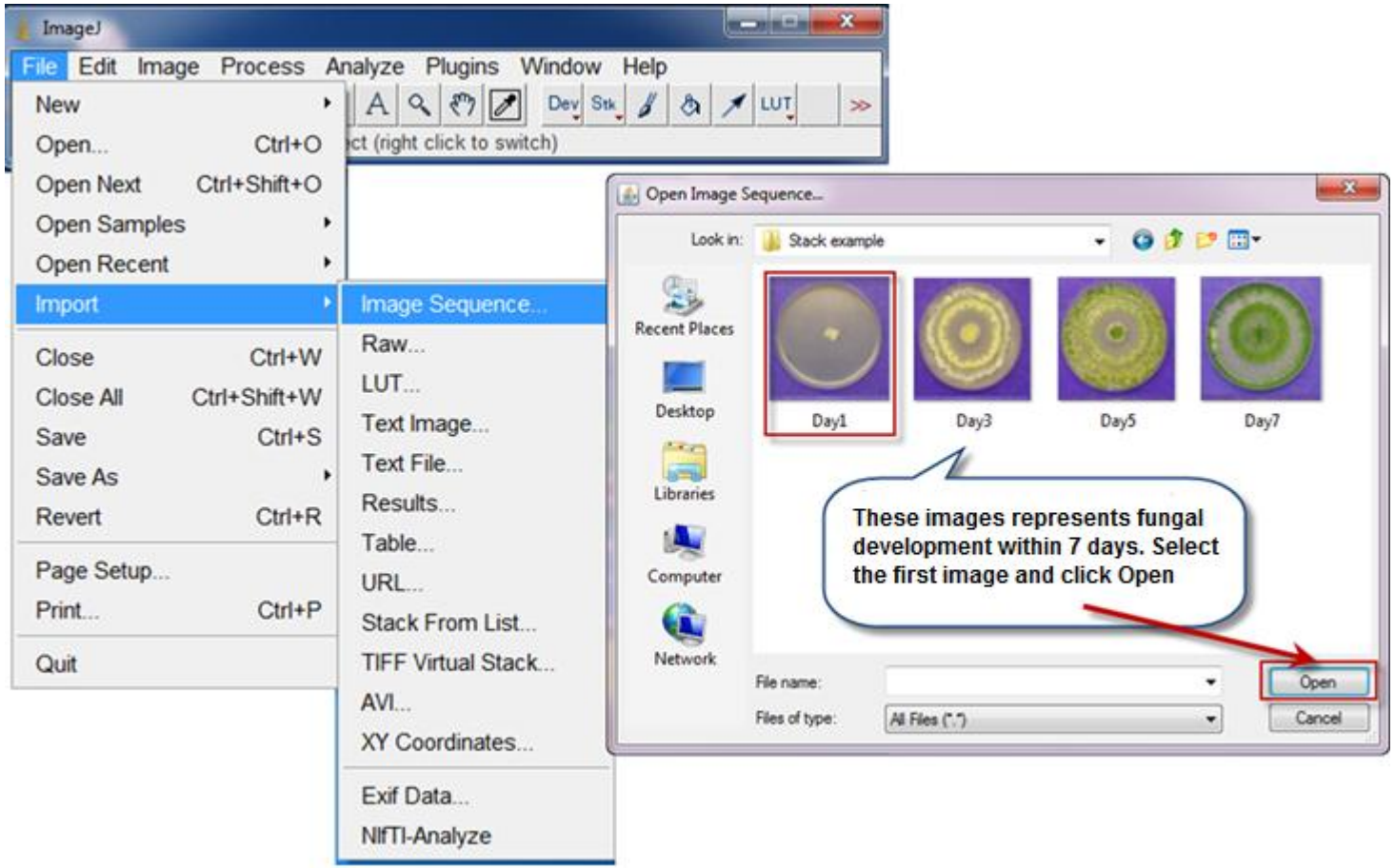


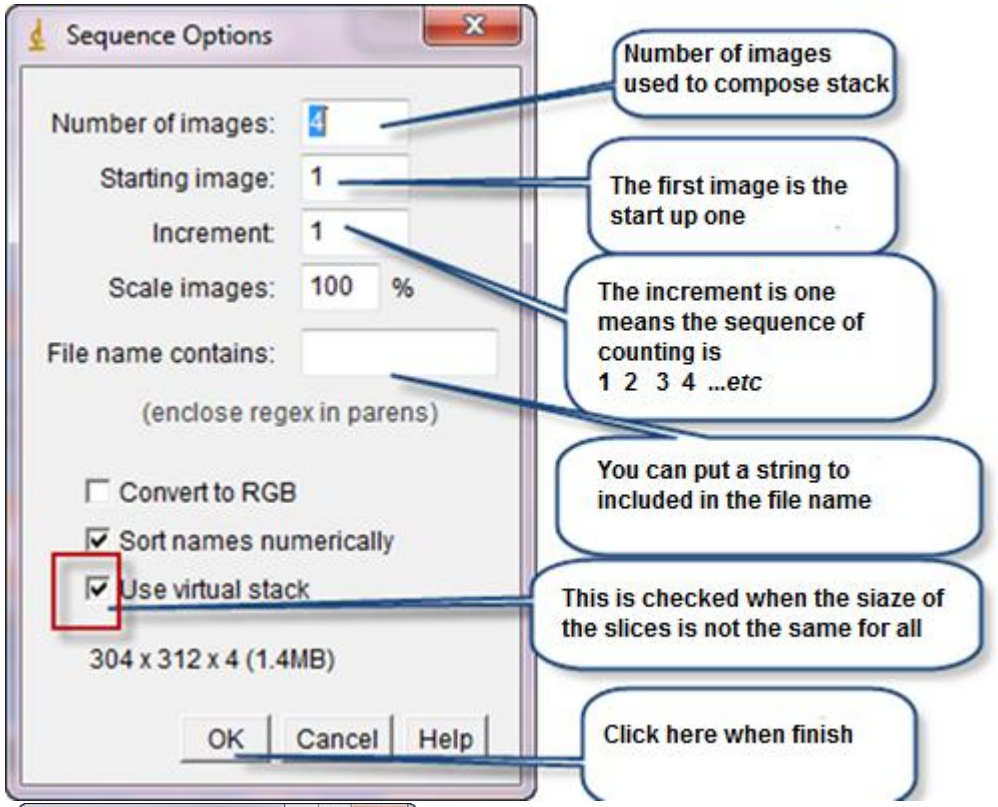
Damage % = (Spots area / Total leaf area) × 100
= (3.397 / 32.697) × 100 = 10.39 %

Stacks – Creating stack

- To create stack we need two or more images in the same size and bit-depth. Now let us create time series stack depending on sequence of images stored in folder.
- The first step is going to the file menu >> import >> Image Sequence.
- The browsing widow will appear >> brows to the folder and open it >> select the first image >> open
- A new widow will appear >> Specify if the information in the boxes are correct (edit if needed) >> Give a name to the file >> ok.
- The stack will be created >> file >> save.

Stacks – Creating stack



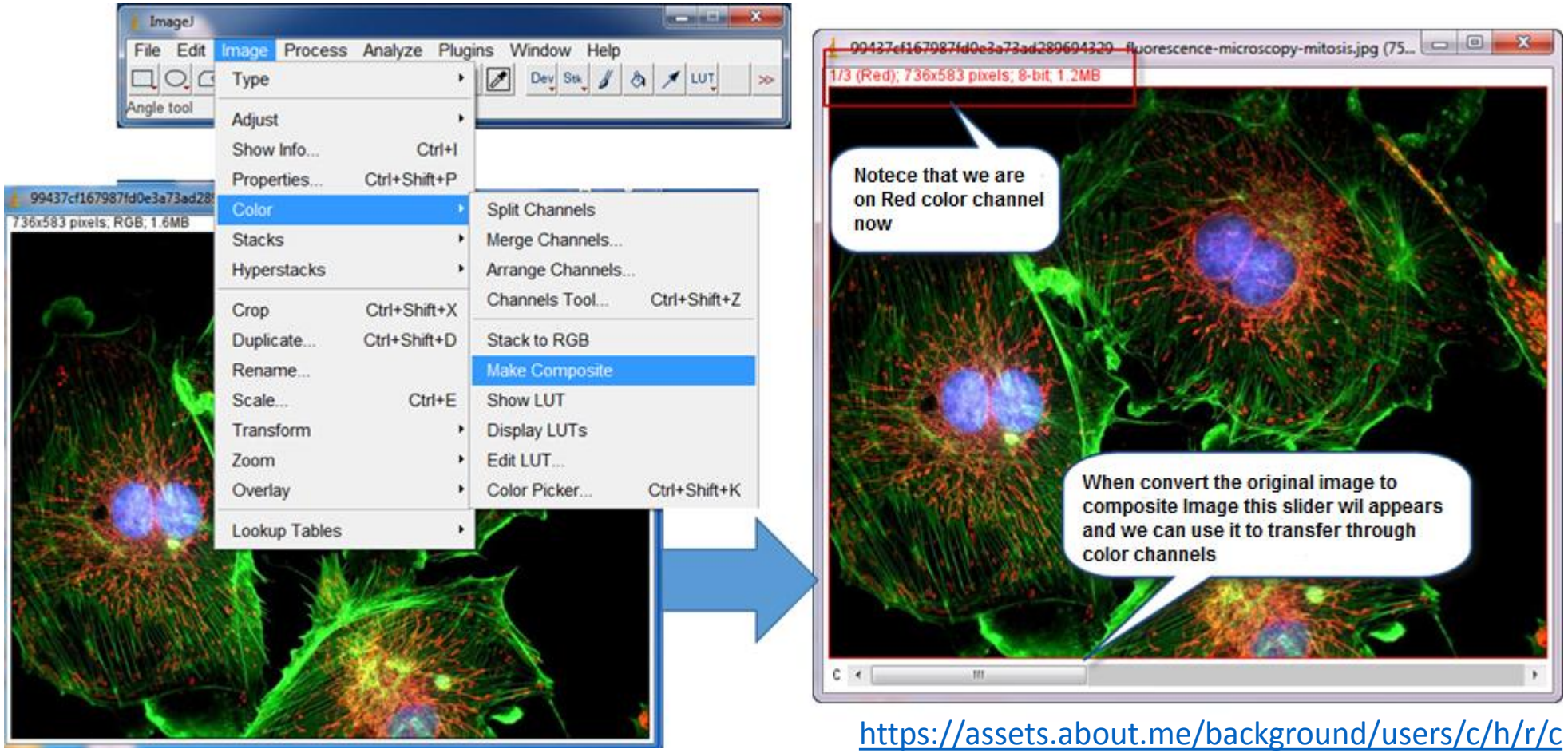


Stacks – Creating stack

- To create spectral stack we need a number of images equal to the number of spectra that we want to analyze (Red, Green, Blue for example). This technique used mostly for fluorescent microscope image analysis as we need to separate channels to isolate objects for analyze according to the colors that they got.
- If the images were separated previously (Red, Green and blue), so just we need to stack them as we learned before; if not, we have to separate channels as below:
  - Open fluorescent image >> go to Image menu >> Make Composite >> a slide bar will appear in the bottom of image frame.
  - Image menu >> Split Channels >> we'll got three images (one for each channel) >> Save them in a folder.

Stacks – Creating stack

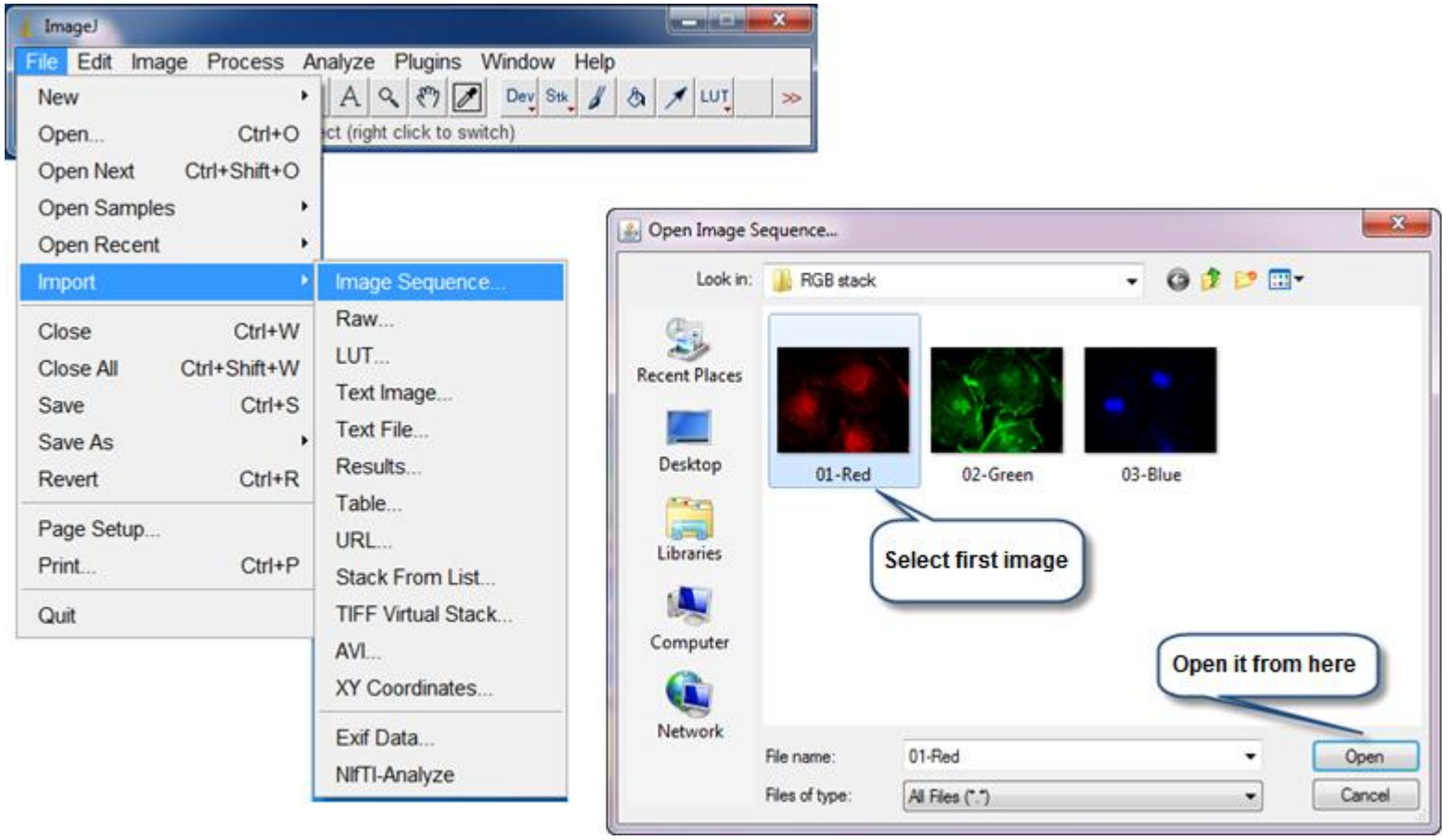
❖If the images were separated previously (Red, Green and blue), so just we need to stack



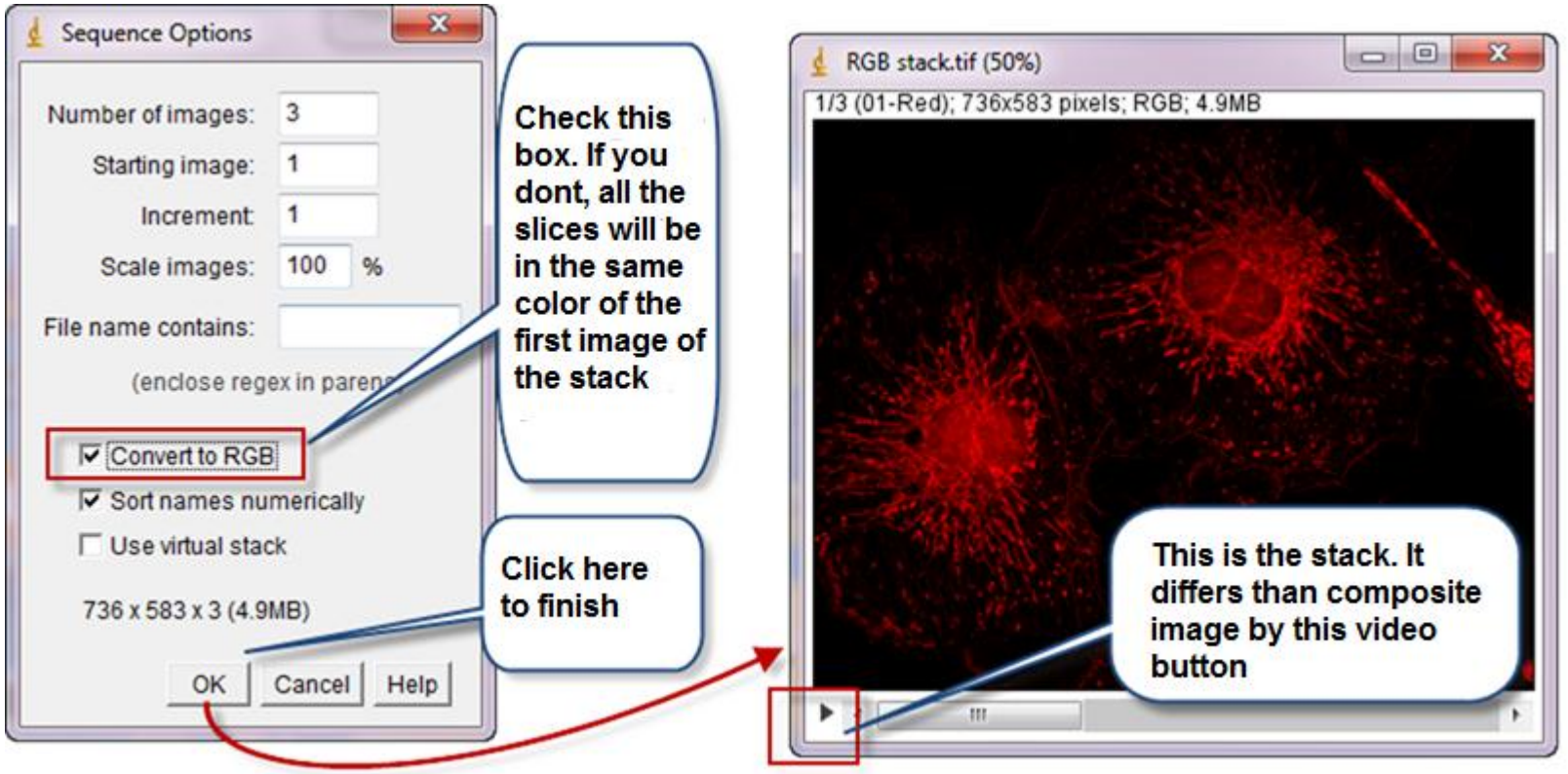
https://assets.about.me/background/users/c/h/r/christineleonswisher_1420341687_62.jpg

Stacks – Creating stack

❖ Now we have to stack them: Go to file menu >> Import >> Image sequence >> brows >> open the first image >> in the appeared window be sure to check “Convert to RGB” box >> OK

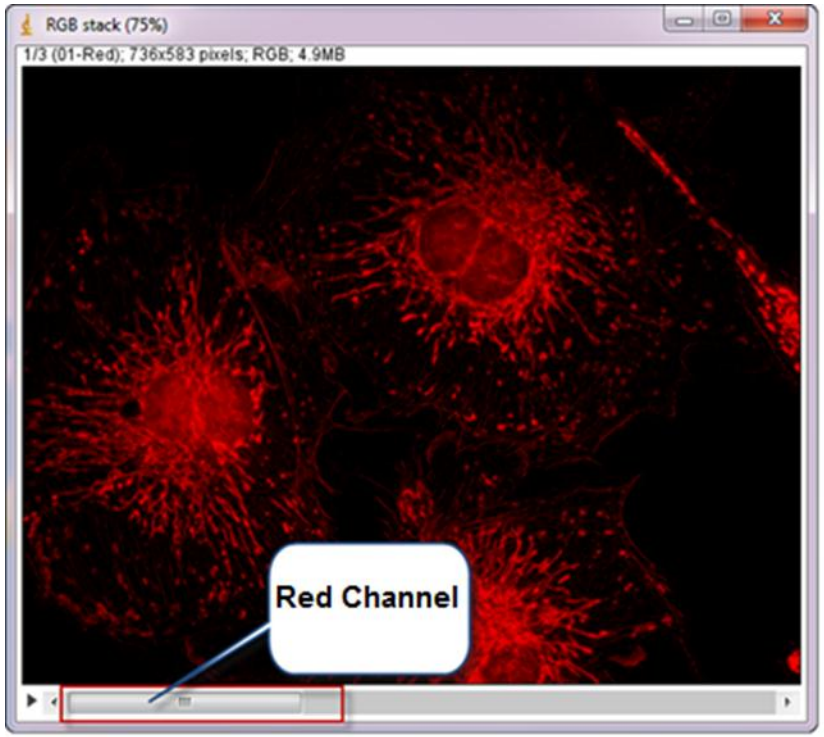


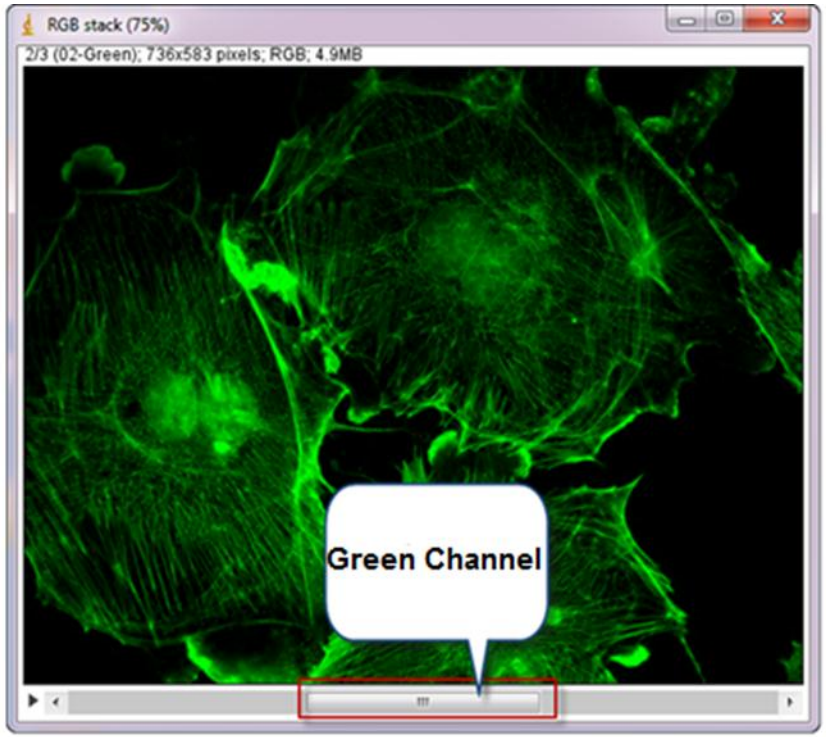
Stacks – Creating stack

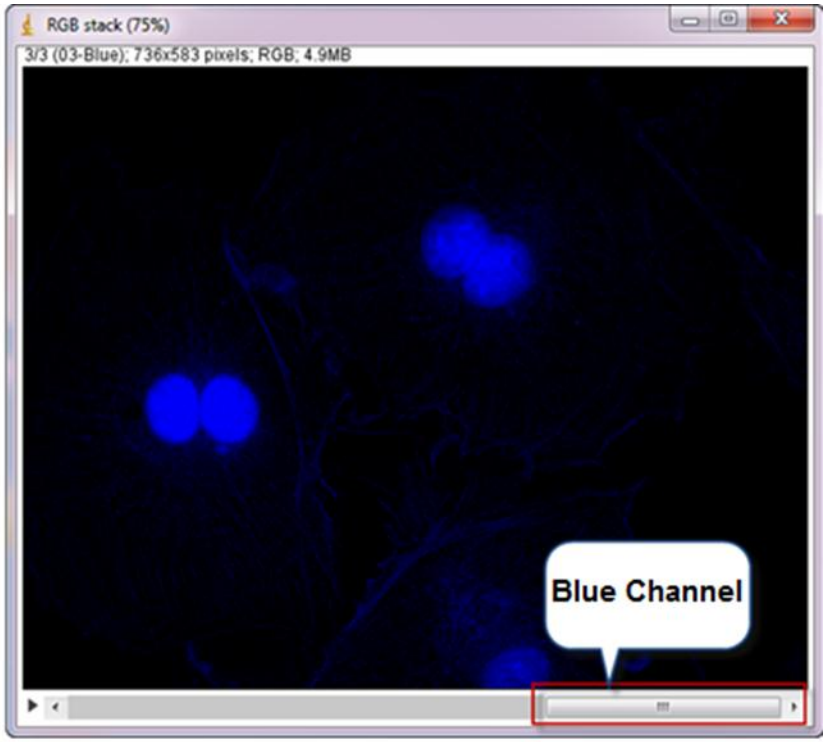


Stacks – Creating stack

❖ The stack will be created and when we move the slide we can explore the channel slides.



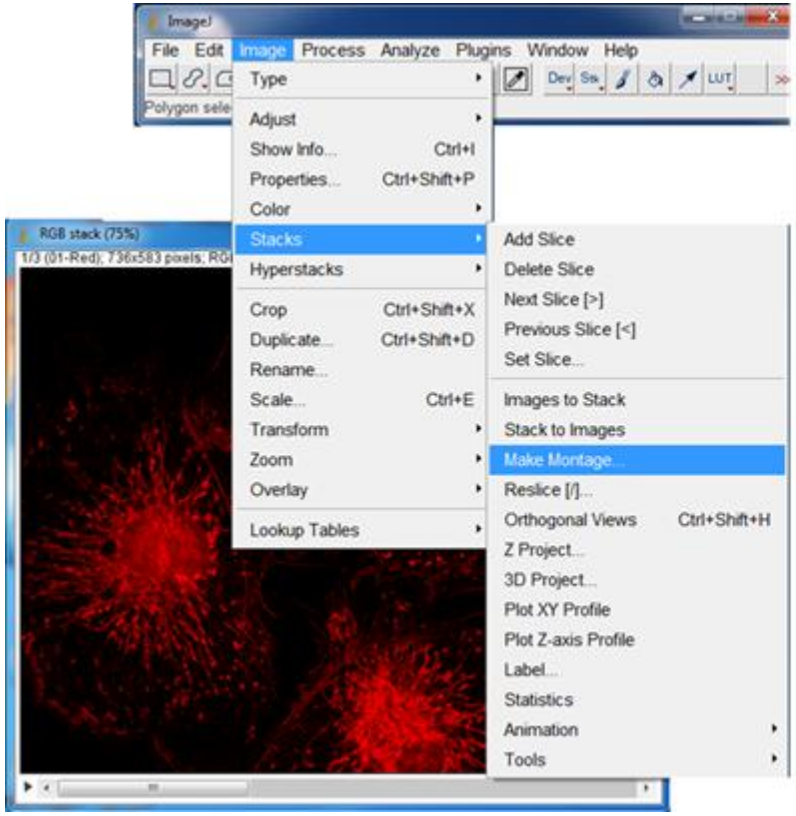


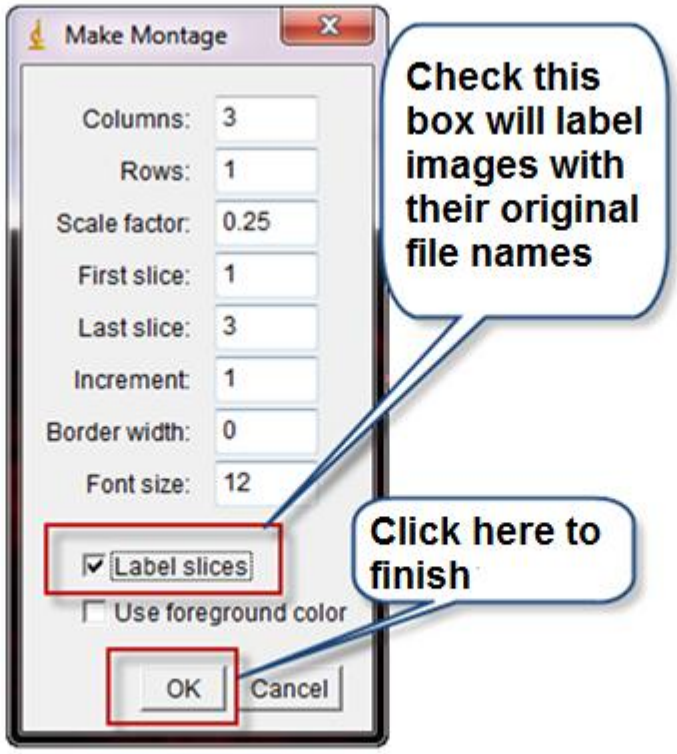


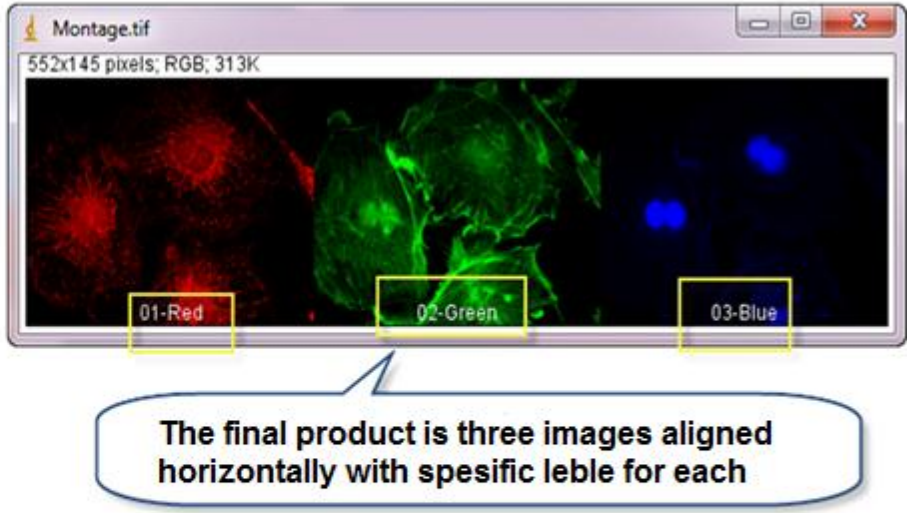
Stacks – Creating stack

❖ The stack slices can be prepared for printing or publishing in scientific papers using “Make montage” option as below:

- Go to Image menu >> stacks >> Make montage >> New window will appear >> Check “Label slices” check box >> OK.







Stacks – Example for time series measurements

In this example we'll calculate the area of the fungal radial growth to study the growth development by the time and estimating the growth rate.

❖ Open stack that we created before: Go to file menu >> Open >> Brows and open stack file.