

MusiJ

A front-end macro to simplify the use of the Jmusical plugin which implements Krishna Agarwal's MUSICAL

- MusiJ manual

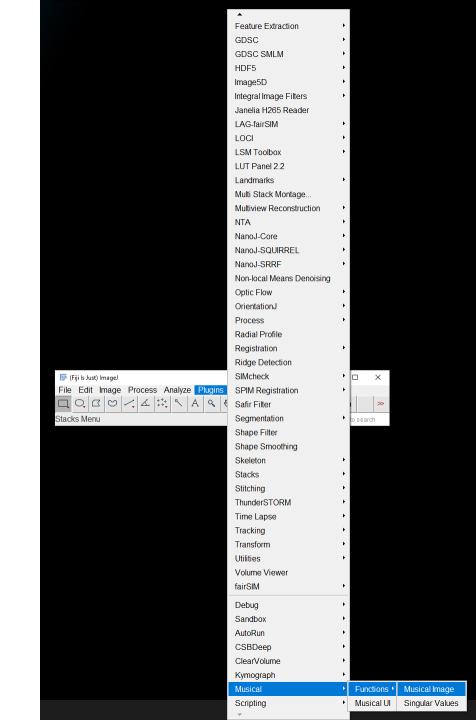
Seba Acuna and Florian Ströhl

Department of Physics and Technology



Make sure you have the plugin Musical for ImageJ installed.

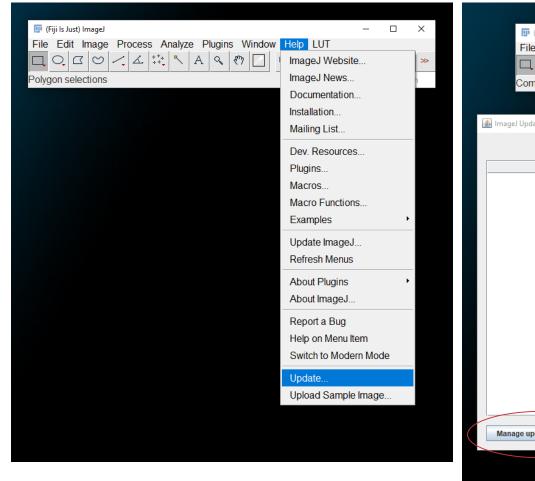
If you don't have it, follow the guide on the next page on how to install Musical.

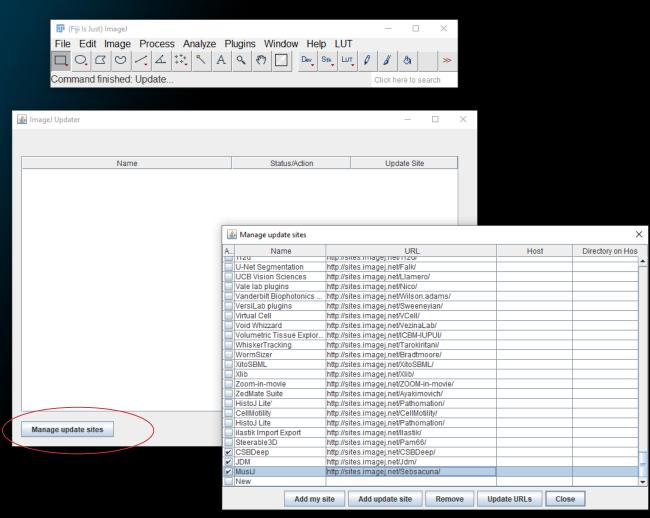


If you don't have Musical, go to «Help» -> «Update...» and in the ImageJ Updater window click on «Manage update sites».

Scroll all the way down and add the update site "http://sites.imagej.net/Sebsacuna/" with a suitable Name, e.g. "MusiJ".

Click on ((Close)) and in the ImageJ Updater window on ((Apply changes))

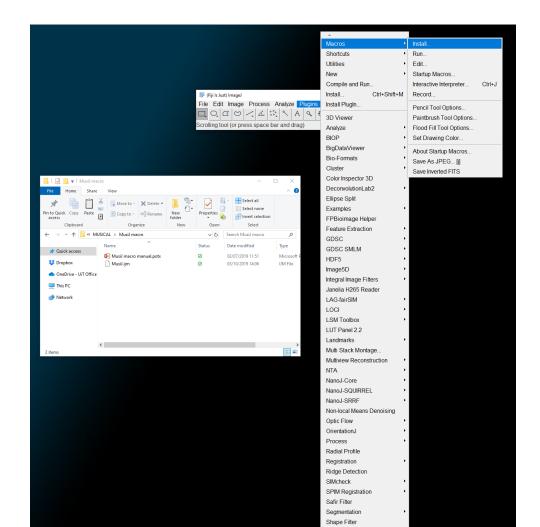


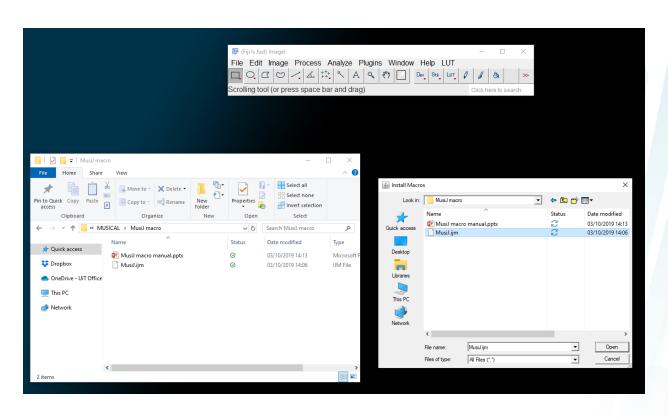


You have Musical for ImageJ, now you can proceed with installing the MusiJ macro.

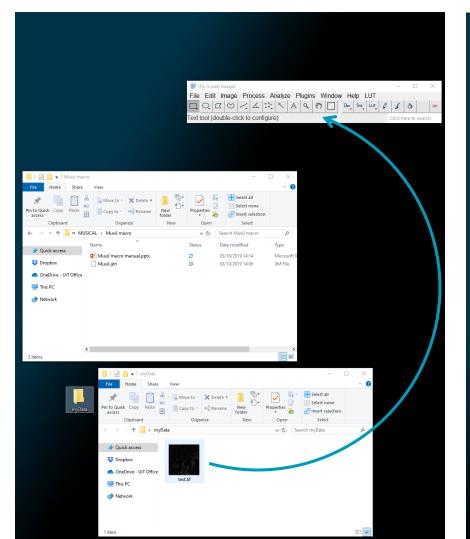
Go to ((Plugins)), ((Macros)), ((Install...)) and the ((Install Macros)) window pops up.

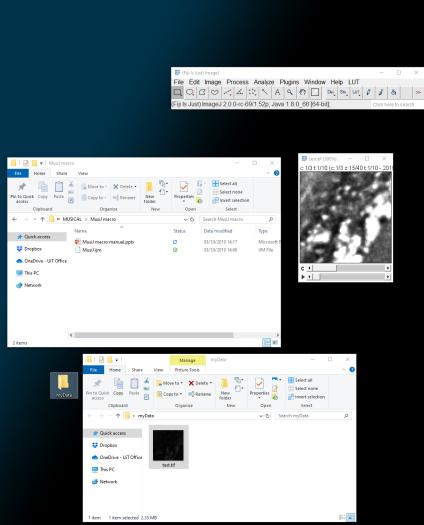
Navigate to the folder with "MusiJ.ijm", select it, and click "Open". Now you can access MusiJ from the Macro list.





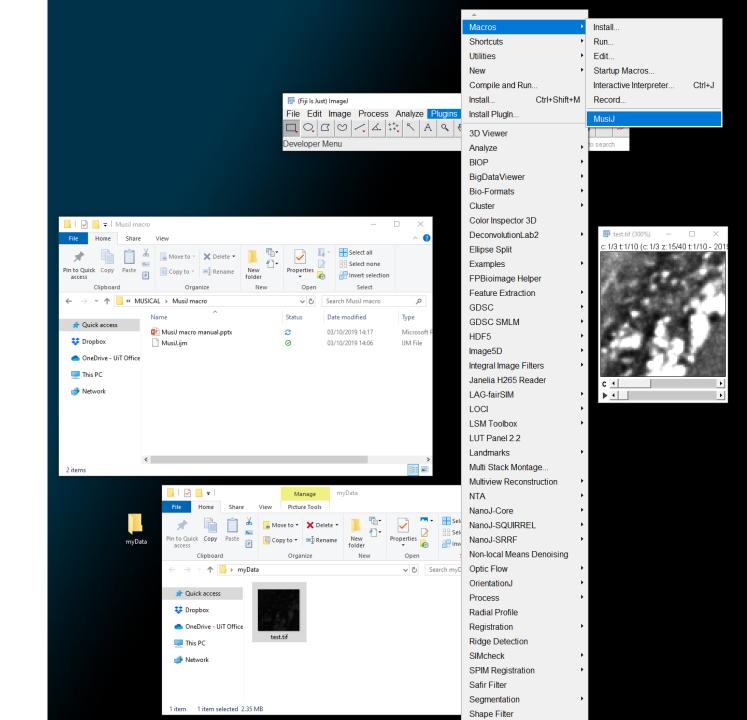
As an example, I'll use the image called (test.tif) from my folder (myData). Drag and drop the image into ImageJ to open it.





Go to ((Plugins)), ((Macros)), ((MusiJ))

This will open the MusiJ dialog box to process the last image you used (i.e. the one you just opened).



It is useful to check the «Singular Values» first to determine the correct thresholds for all of the image's colour channels. Simply select «Singular Values» and tell the software the parameters of you microscope. For singular value computation, the thresholds are not used. Parameters are:

Parameters are:

C1 – CX = number of colour channels

Batch size = how many raw frames are taken at a time for computation of 1 Musical frame **Slide by** = sliding window computation, e.g. for a values of 5 computes Musical of frames 1-X, then 6-(X+5), etc.

Pixel size = either camera pixel size or pixel size on the sample (in this case Magnification == 1).

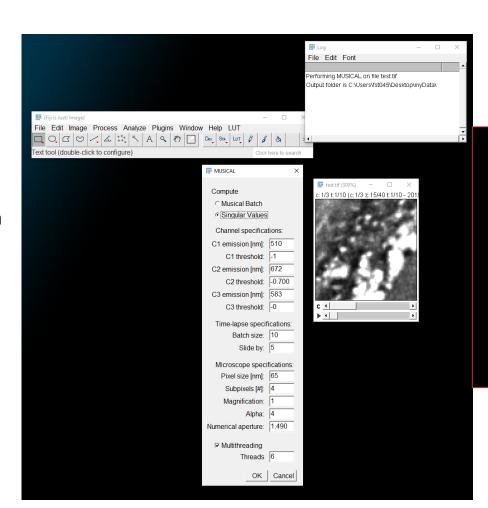
Subpixels = the factor of how many pixels more the Musical image has

Magnification = microscope magnification or 1 (see above)

Alpha = the alpha factor of the Musical algorithm

Numerical Aperture = NA of the microscope

Multithreading can be activated with X **Threads** if supported by the PC.



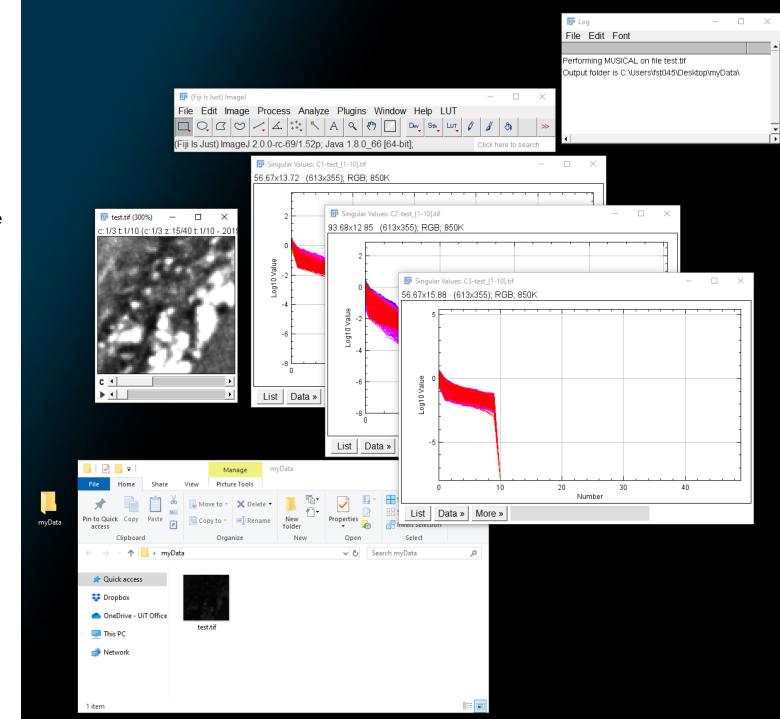


Output from «Singular Values» is display but not saved by default.

Follow the guidelines outlined in respective publications on the choice of ideal threshold.

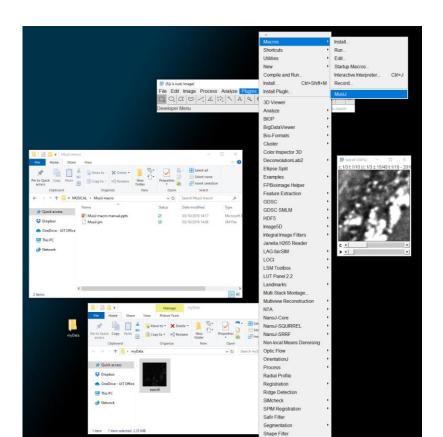
A good starting point is to use a value around the shoulder.

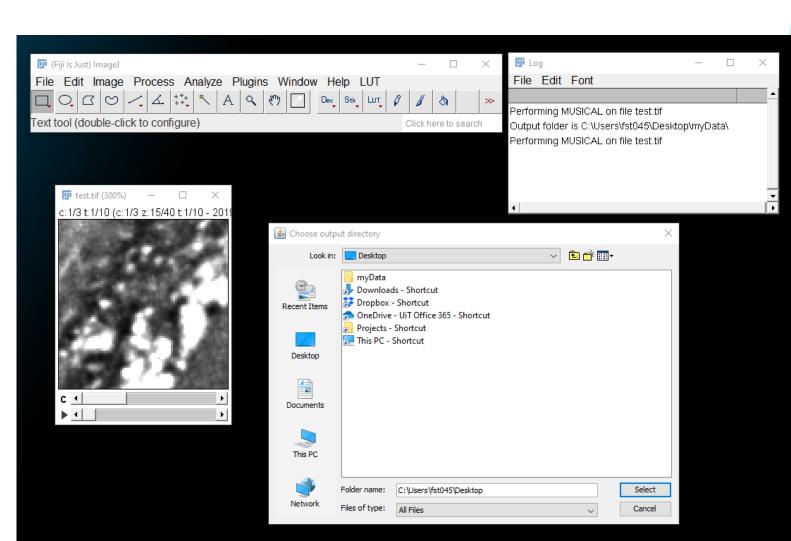
Note this down so it can be plugged into the macro.



If you run the macro directly again without open the image anew, then macro asks you where you would like to safe the result. This occurs everytime the macro is not sure about the save-location of the later produced Musical image.

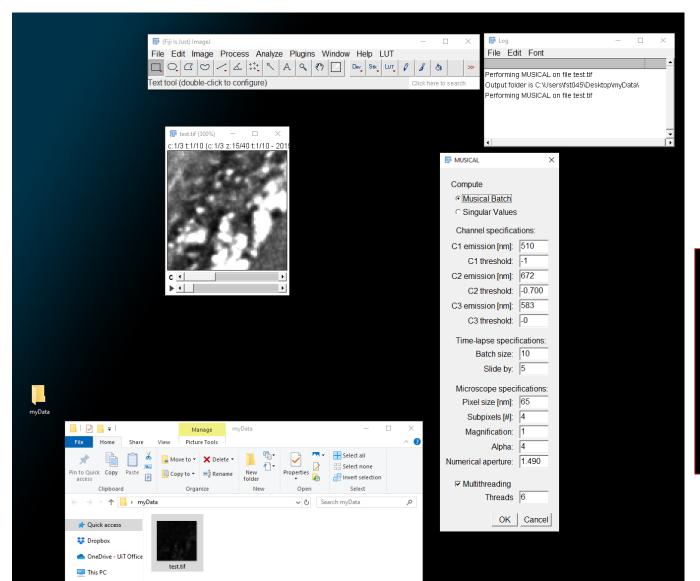
Chosing a location will promt the selection window again (see next page).





Now select «Musical Batch» to do some batch processing of your date. It also works with non-batch data.

Make sure to enter the values correctly. A handy trick for larger reconstruction tasts is given in the end of this manual.

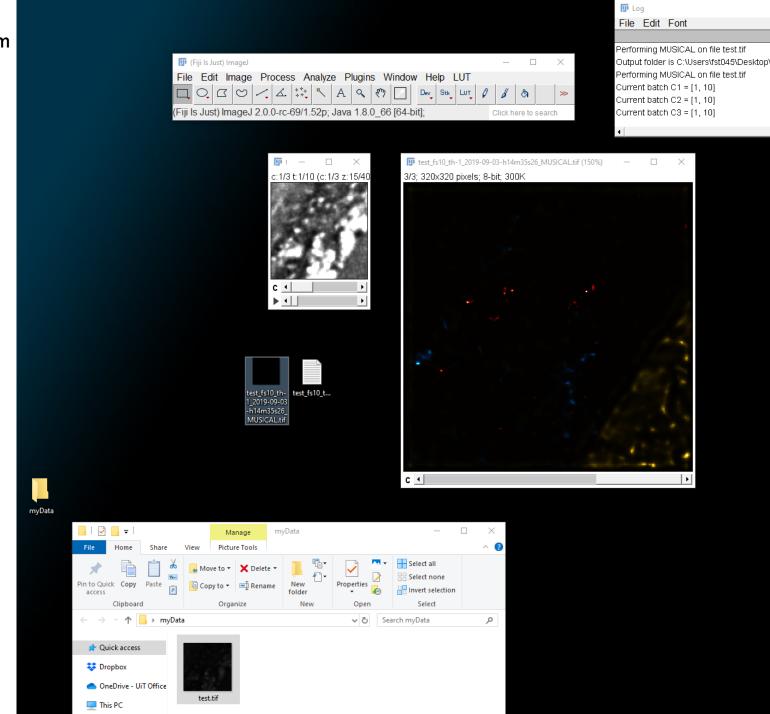




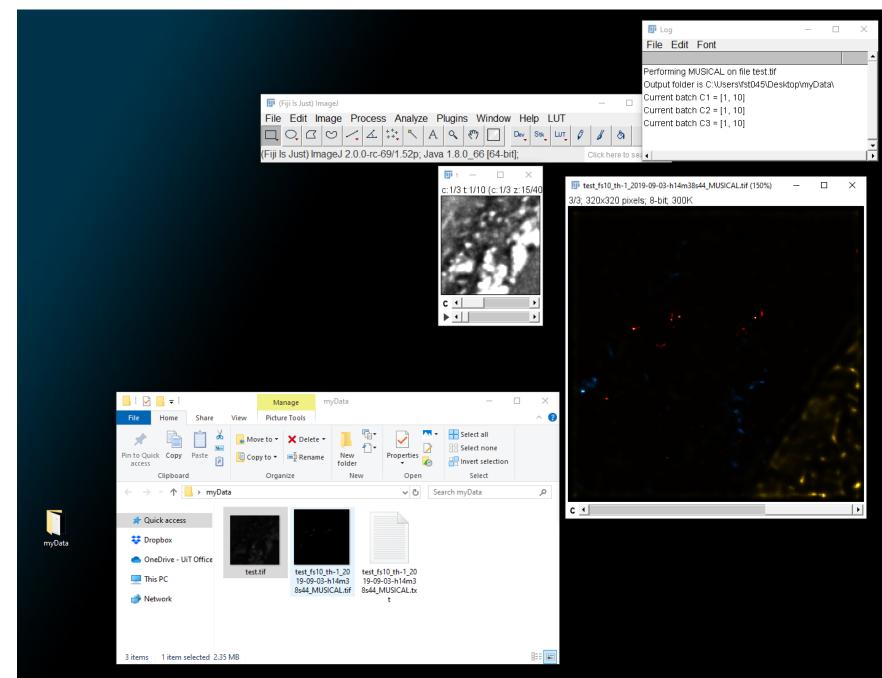
MusiJ calculates the images and saves them on the specified save location.

This was the Desktop in the example.

It also saves a parameter file.

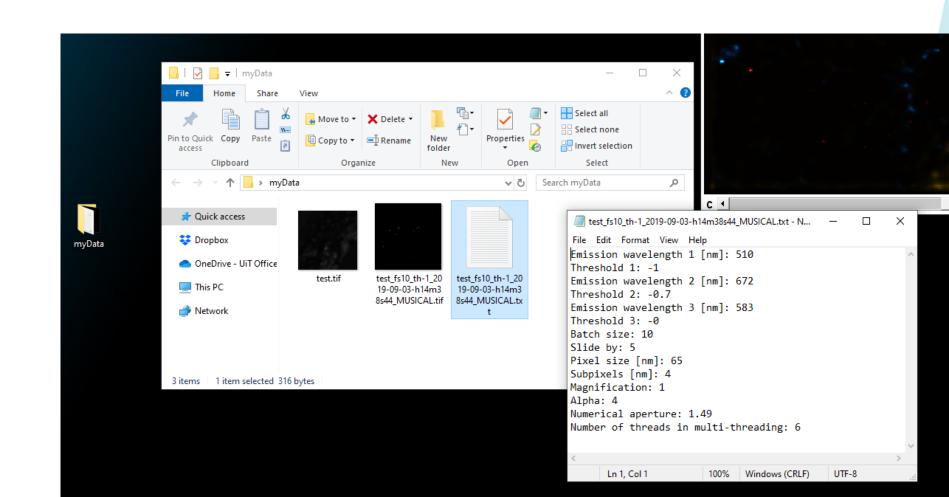


If MusiJ knows about the file location of the raw data, it will save the output in the same folder.



All the parameters used during the computation are saved in the log file.

A small portion (batch size «fs» and threshold «th» of the first colour) is also directly written into the file name. This makes finding a certain Musical image easier.

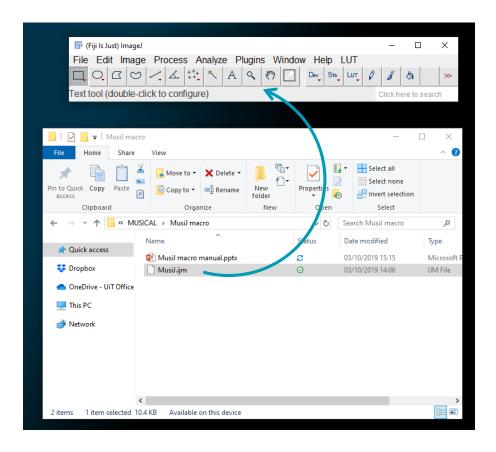


Pro Tip (1)

When a lot of similar data sets are to be processed (i.e. same microscope, colours, and thresholds), then the default values in the macro can be modified. For this, drag/drop the MusiJ.ijm file into ImageJ and edit the «default values» accordingly.

Click (File) and (Save). Now, either use the macro directly by clicking (Run), or follow the guidelines outlined above and load

the modified macro again to the macro tab of ImageJ.



```
ile Edit Language Templates Run Tools Tabs Options
            3 // This macro simplifies the use of JMusical on large data sets comprising multiple colours and
             4 // time-lapse recordings.
            6 // Author: Florian Ströhl, 16/01-2019
            8 // Modified by Ida Opstad and Florian Ströhl, 03/10-2019, to add more information in the file name.
            10 // ====== default settings
           11 // when processing lots of similar data sets, it is convenient to change the default settings:
            12 default_colours = newArray(510, 672, 583, 440, 440);
            13 default_thresholds = newArray(-1, -0.7, -0,5, -1,3, -1.0);
            14 default luts = newArray("Red Hot", "Cyan Hot", "Yellow Hot", "Green", "Magenta Hot");
            15 default_batchsize = 10;
            16 default slideby = 5;
            17 default pixelsize = 65;
            18 default_subpixels = 4;
            19 default magnification = 1;
           20 default alpha = 4.0;
           21 default numericalAperture = 1.49;
            22 default multithreadingOption = true;
           23 default threads = 6;
            26 // ===== Do not edit below unless you know what you are doing
           29 // ====== get image file information
            30 img = getTitle();
           31 img without extension = getTitleStripExtension();
           32 dir = getDirectory("image");
          Run Batch Kill persistent
                                                                                                Show Errors
                                                                                                            Clear
```

Pro Tip (2)

Add

```
//Run MusiJ
macro "JMusical Batch Action Tool -
C000D16D21D27D31D32D33D34D35D36D3aD3bD41D49D4bD4cD59D5aD5bD5cD60D61D62D63D64D65D66D67D68D69D6aD6bD71D73D81D83D91D93Da1Da3DadDaeDb2Db4DbcDbeDbfDc2Dc4DccDcdDceDcfDd2Dd3Dd4Dd5Dd6Dd7Dd8Dd9DdaDdbDdcDddDdeC000C111C222C333C444C555C666C777C888C999CaaaCbbbCcccCdddCeeeCfff" {
    runMacro("MusiJ.ijm");
}
```

III (Fiji Is Just) ImageJ

Edit Image Process Analyze Plugins Window

To the «StartupMacros.fiji.ijm» file in the macro folder of your Fiji directory to add quick-access button to Fiji.

