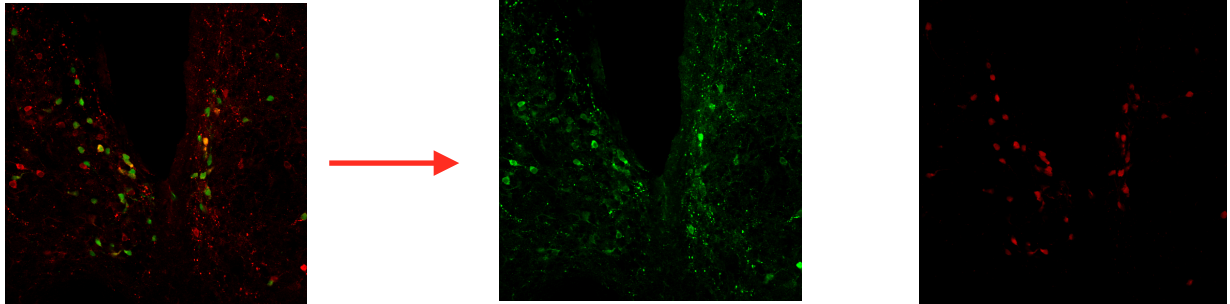


Determining Colocalised Objects Using Fiji (fiji.sc)

Open image in Fiji (fiji.sc)

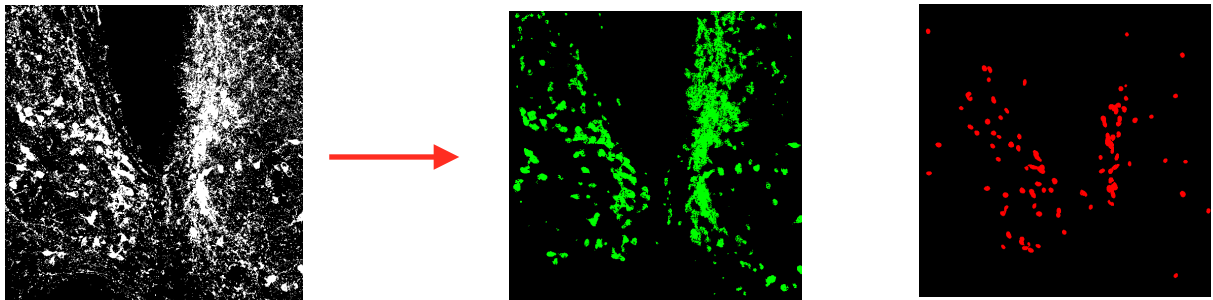
Split the colour channels using Image / Colour / Split Channels

If a stack of images and the whole series needs to be compressed into a single plane of focus, use Image / Stacks / Z project ... and generally use Maximum Intensity projection.



To reduce the amount of noise, threshold the image to try and isolate the regions of interest Image / Adjust / Threshold. Click the Apply button to binarise the image (the Look Up Table LUT will probably need to be inverted back to normal (i.e. 0=black and 255 = white) using Image / Look Up Tables / Invert LUT. Check if top of image window says 'Inverting LUT'.

Remove unwanted areas using Process / Noise / Remove Outliers ... this removes objects smaller than a defined size (check if they are bright or dark), experiment with the Radius option and use preview to check the results. Always compare the result to the original image to ensure the data is not being distorted.



Repeat for second channel and apply appropriate LUT colour.

To show the colocalised regions, either use Process / Image Calculator and the function AND to show the regions that occur in the same place in both images, then add the appropriate LUT colour.

Or, use Image / Colour / Merge Channels and combine the two images from the step above.

