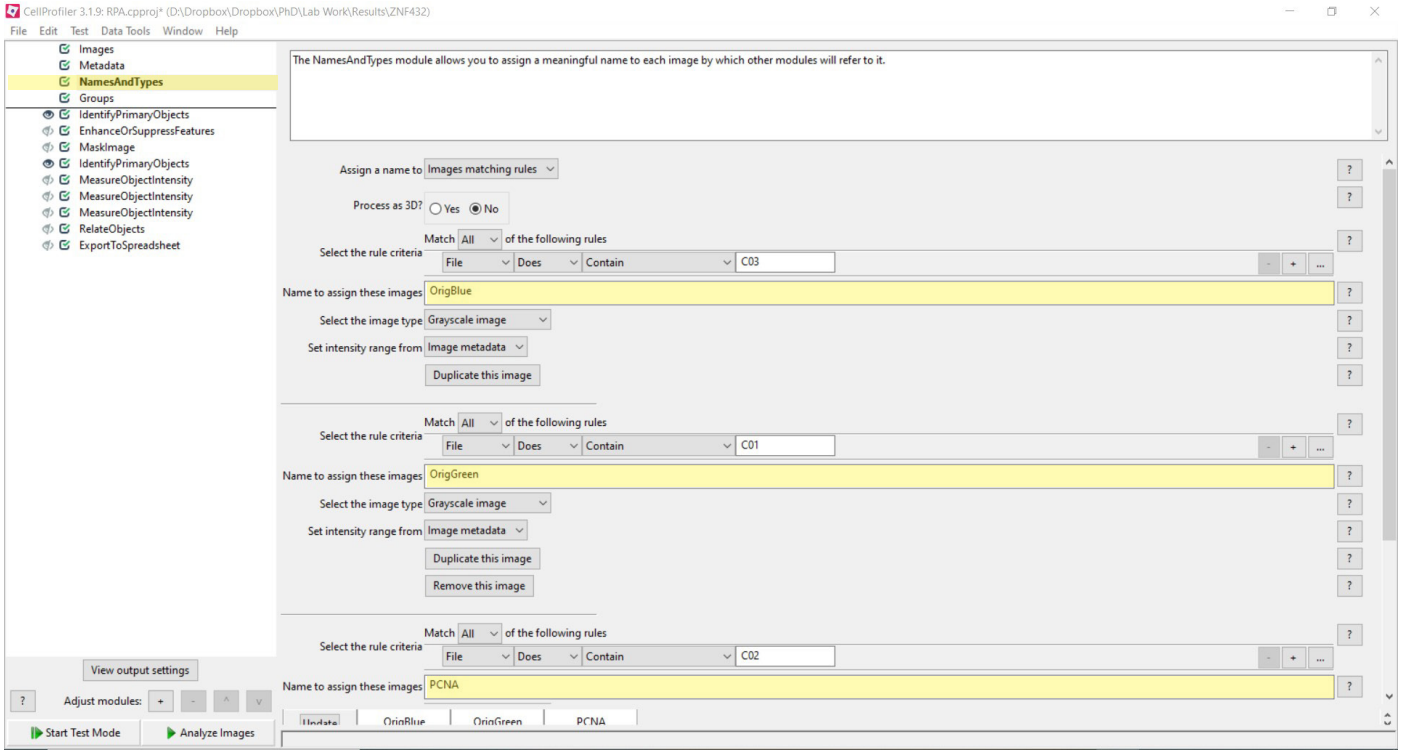
A)

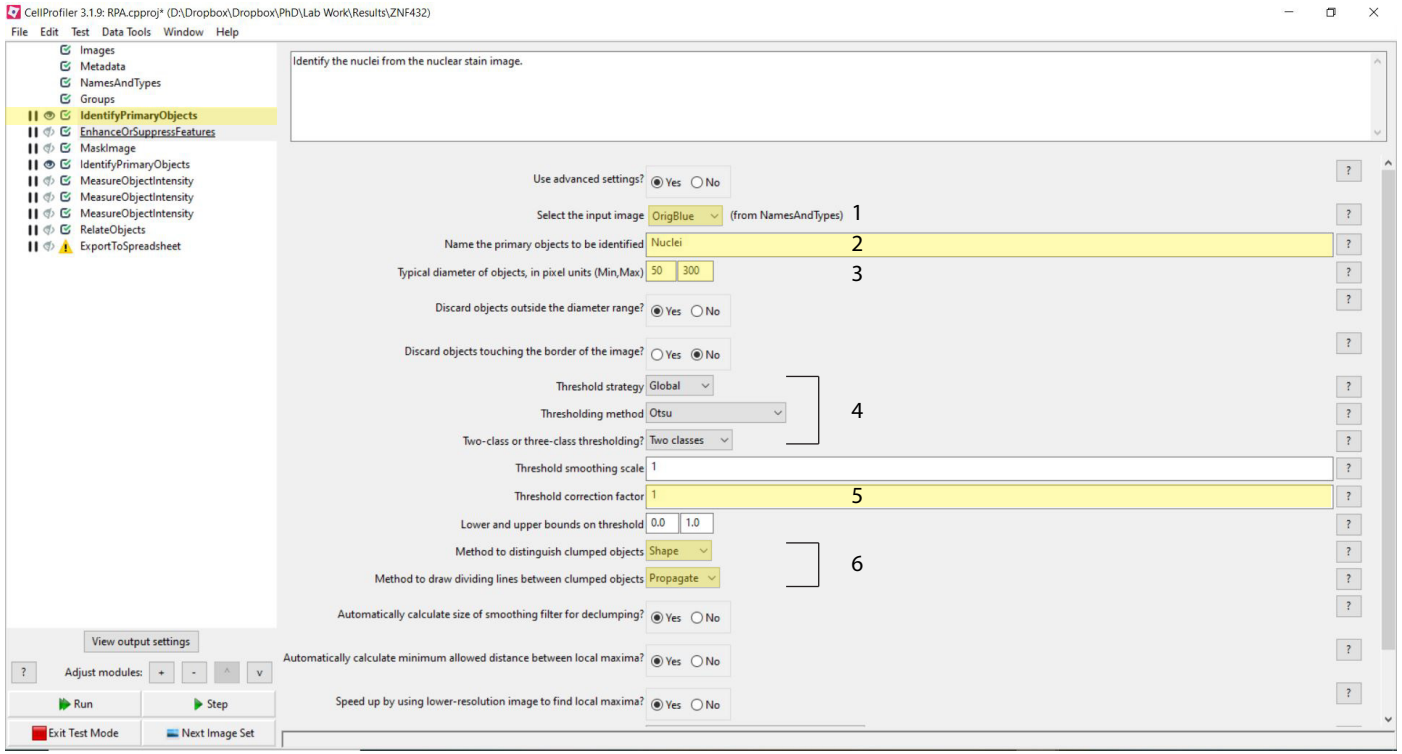


Representing the DAPI channel

Representing the BrdU channel

Representing the PCNA channel

B)



1) The channel used to identify the nucleus in this case is the DAPI

2) The label assigned the objects identified

3) The acceptable size range for the nuclei, which should be changed to best fit the cells in your images.

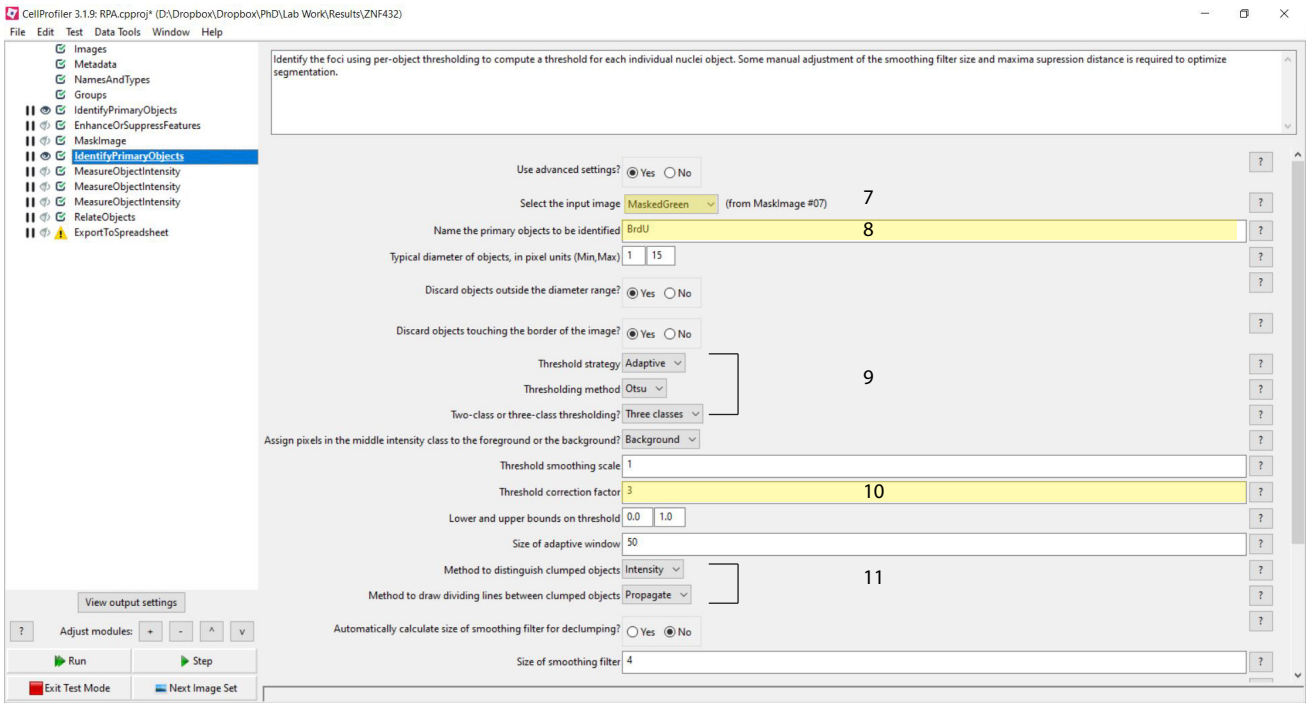
4) The thresholding strategy

5) The thresholding correction factor, i.e. how stringent the the thresholding strategy will be. This is the setting that will be most altered to fit cell shape and distribution.

6) How the cells are differentiated from each other by the program

O'Sullivan, Mersaoui et al. Supplemental Fig 2

A)



7) The channel used to identify the foci is the processed BrdU channel.

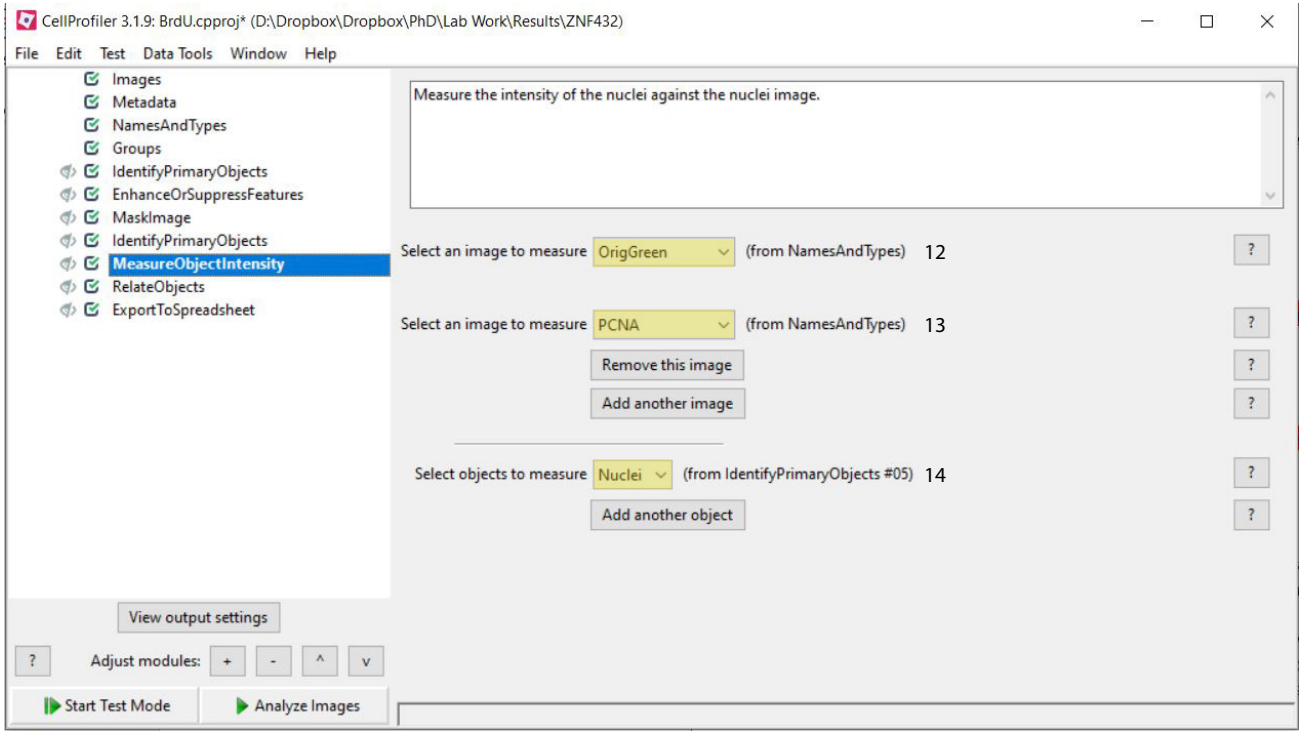
8) The name that will be given to data resulting from this section of analysis, which can be changed without issue.

9)The threshold strategy to identify and differentiate individual foci.

10) The threshold level, this will be changed to increase or decrease the number of objects identified as foci.

11) How the foci are differentiated from each other by the program

B)



12) The unprocessed channel whose intensity will be measured, in this instance the green BrdU channel.

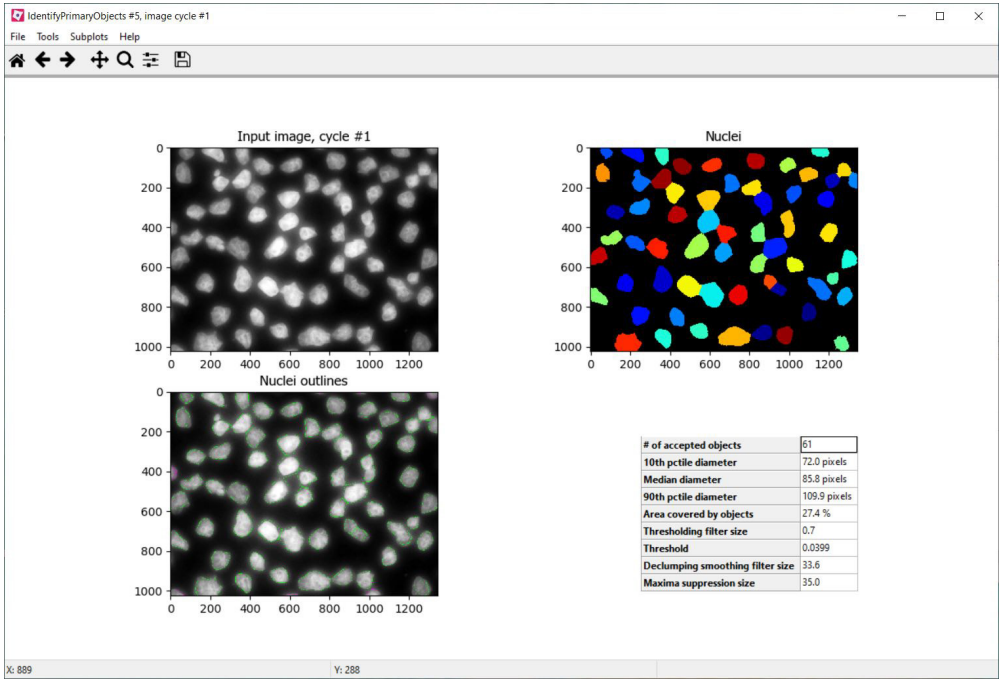
13) The unprocessed channel whose intensity will be measured, in this instance the red PCNA channel.

14) The area that is to be measured, either nuclei or foci.

O'Sullivan, Mersaoui et al. Supplemental Fig 3

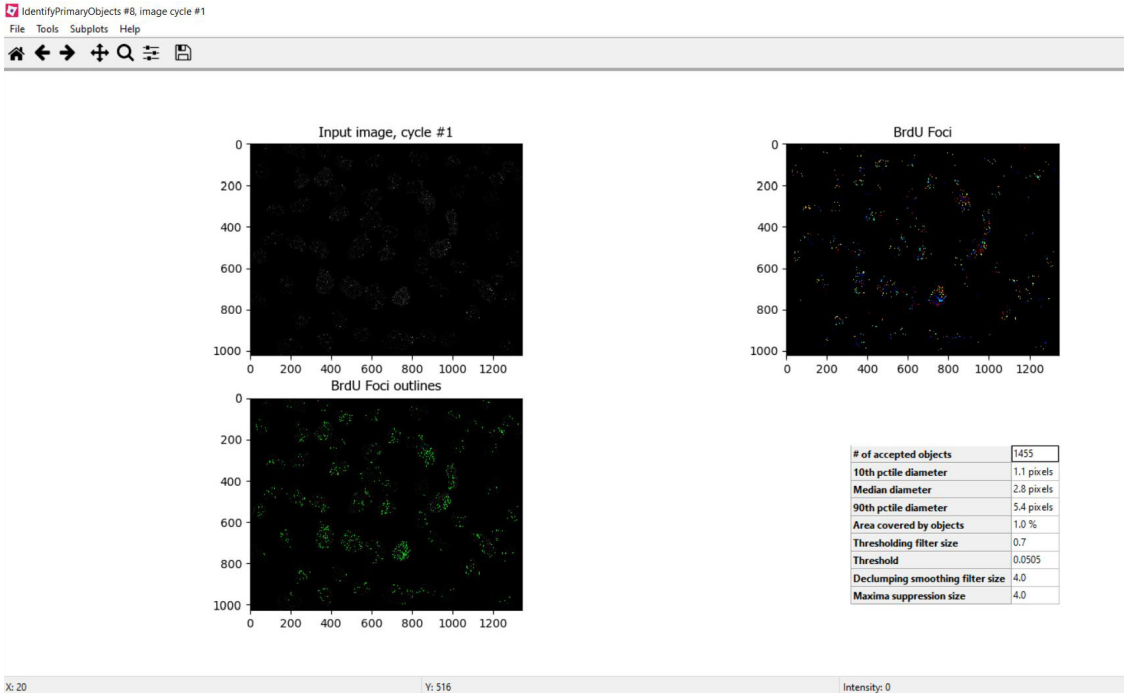
A)

Example of nuclei identification

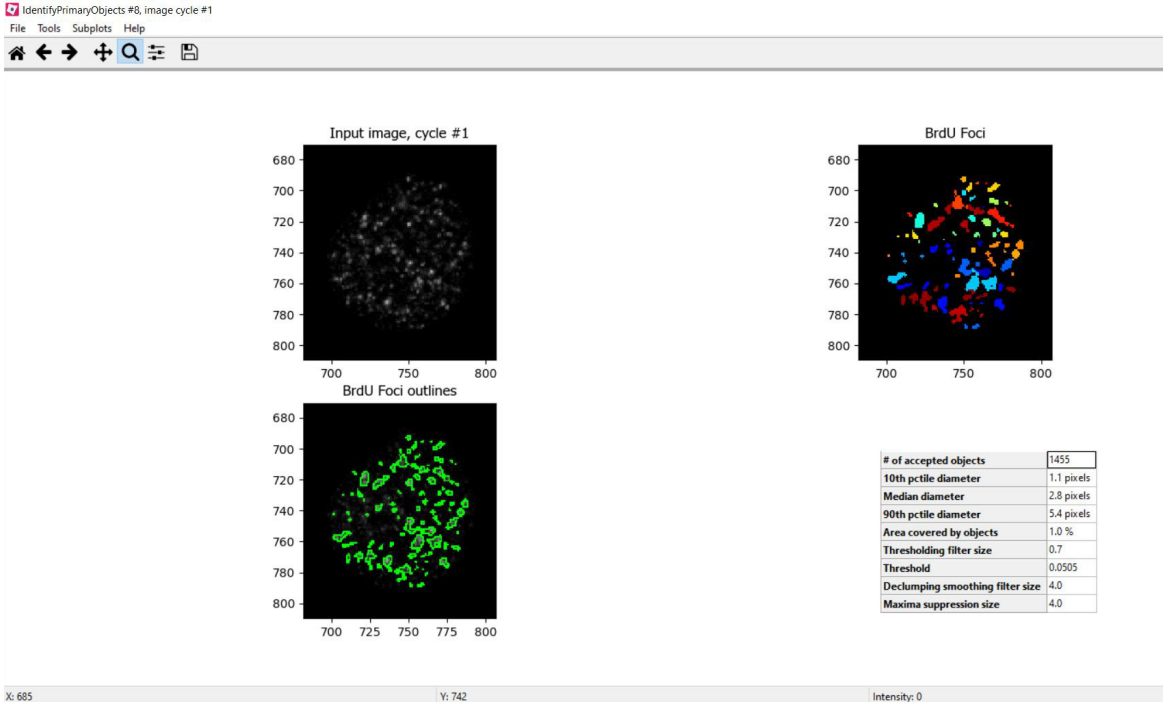


B)

Example of foci identification

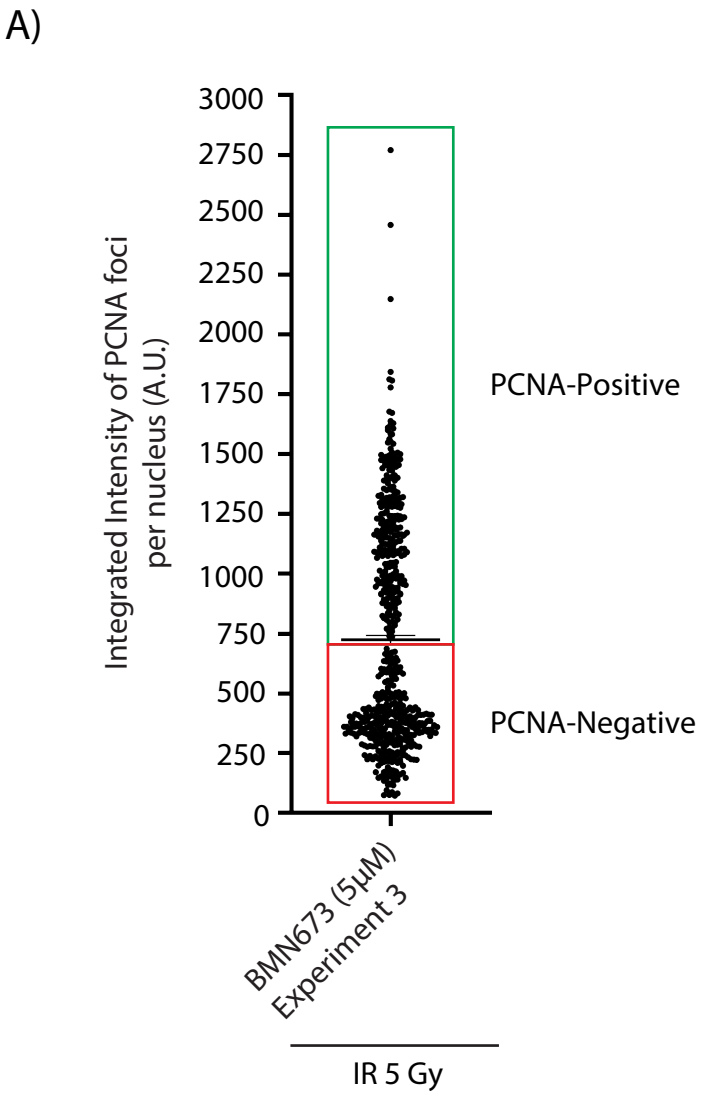


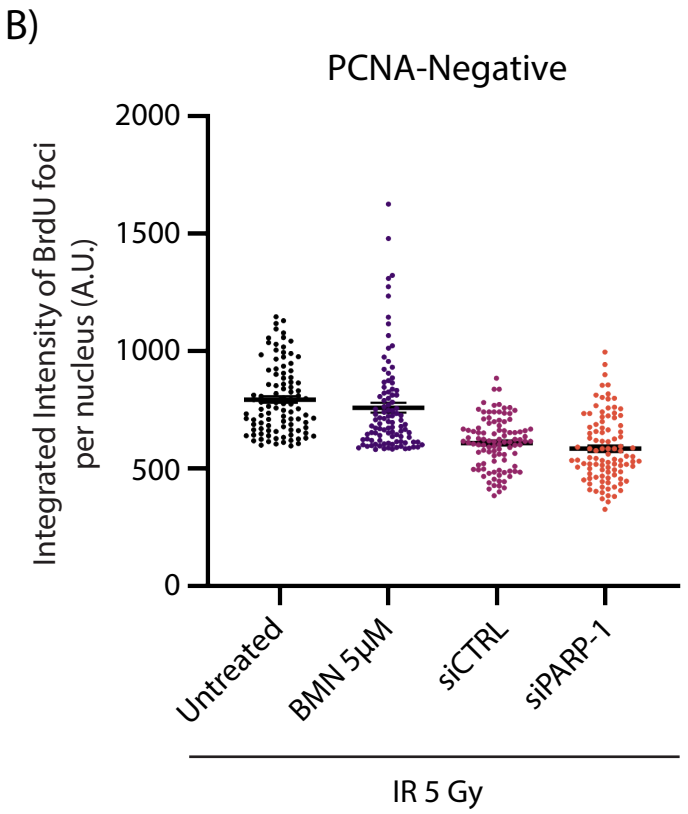
C)



Zoom-in of foci identification

O'Sullivan, Mersaoui et al. Supplemental Fig 4





O'Sullivan, Mersaoui et al. Supplemental Fig 5