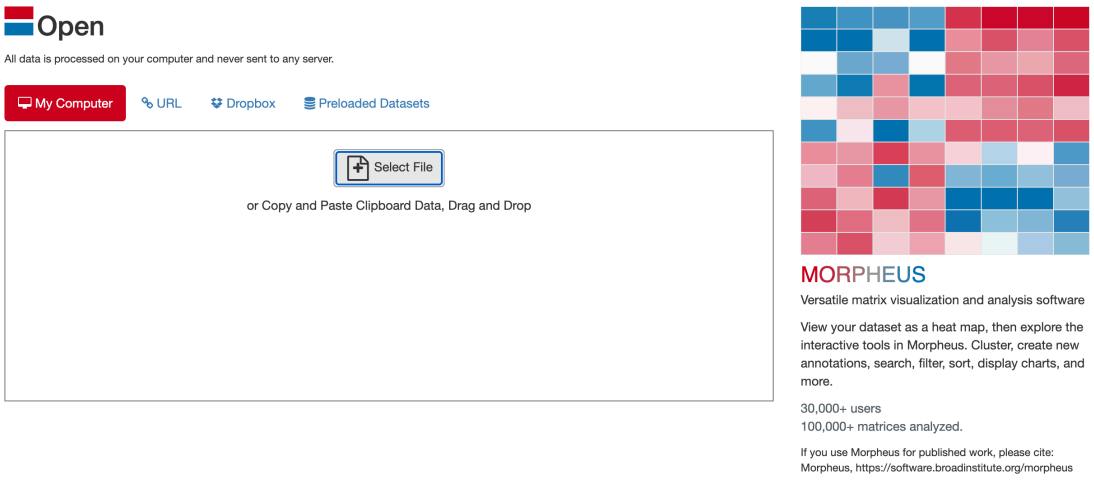
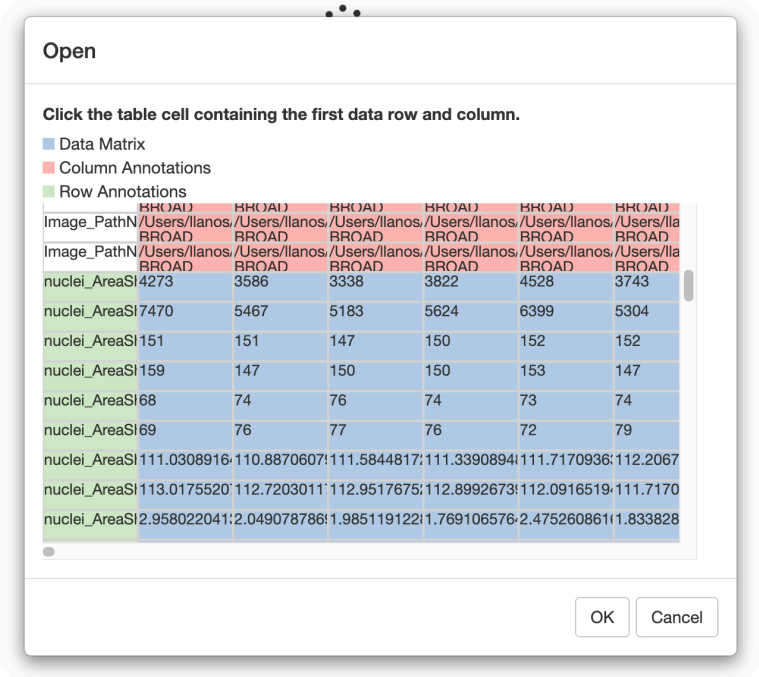
Guide for calculate the T-test value between cell lines in Morpheus.

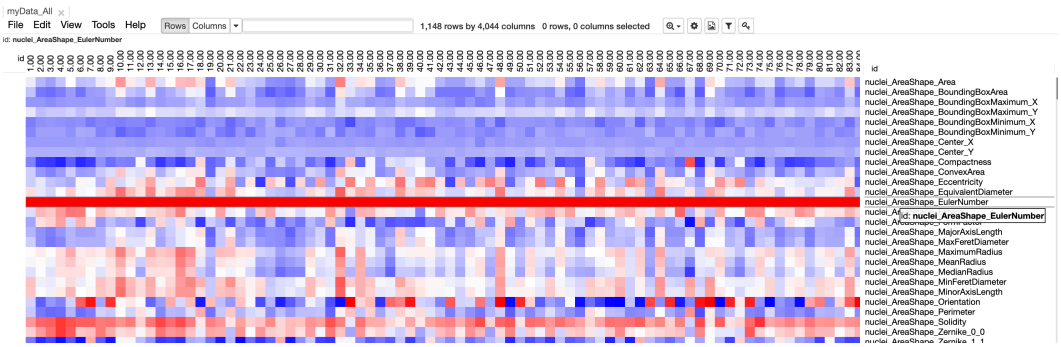
1- Load the .csv file obtained from Cellprofiler and previously edited with the python code.



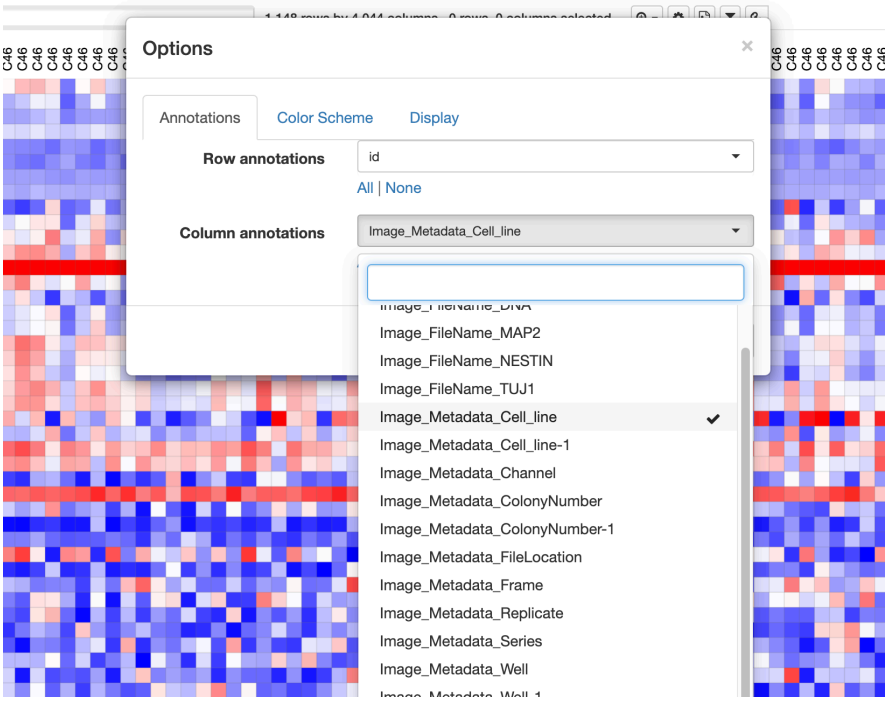
2- Select in red the metadata columns, which are the rows with the names starting in “Image_”. In green will be the first column with the features measured.



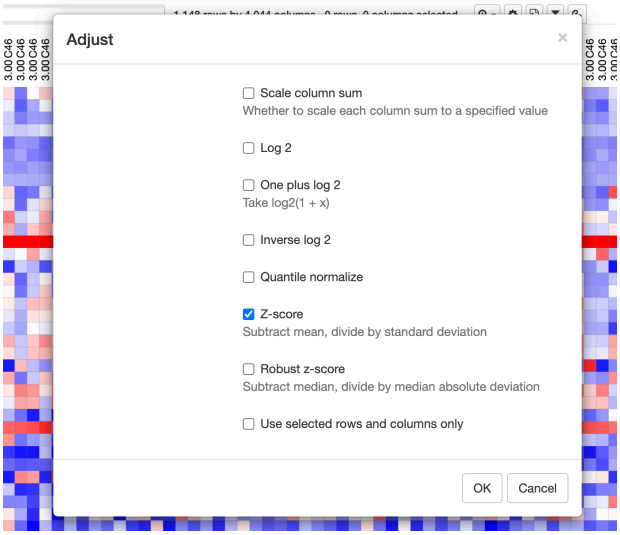
3- You will get something like the image below. Where the rows corresponds to the features and the columns correspond to each organid ID. Since this, doesn't give us enough information, we can display the data to show in the columns, like cell line, replicate, well, etc.



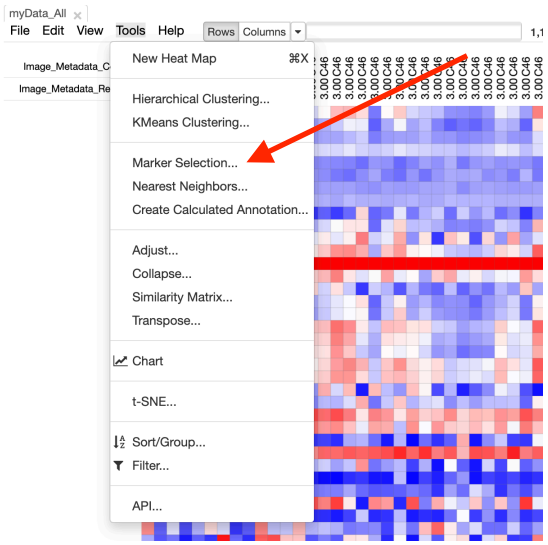
4- Select the gear and click in the tab "Annotations", then select any metadata that you are interesting in see difference. In this case, will be "Cell_line".

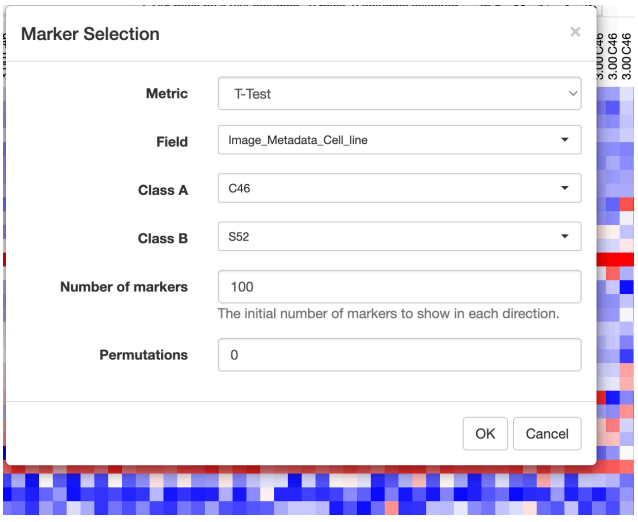


5- In Tools, go to adjust to normalize the scales of the data, then everything will be in a range between 0 to 1. Tools->Adjust->Z-score

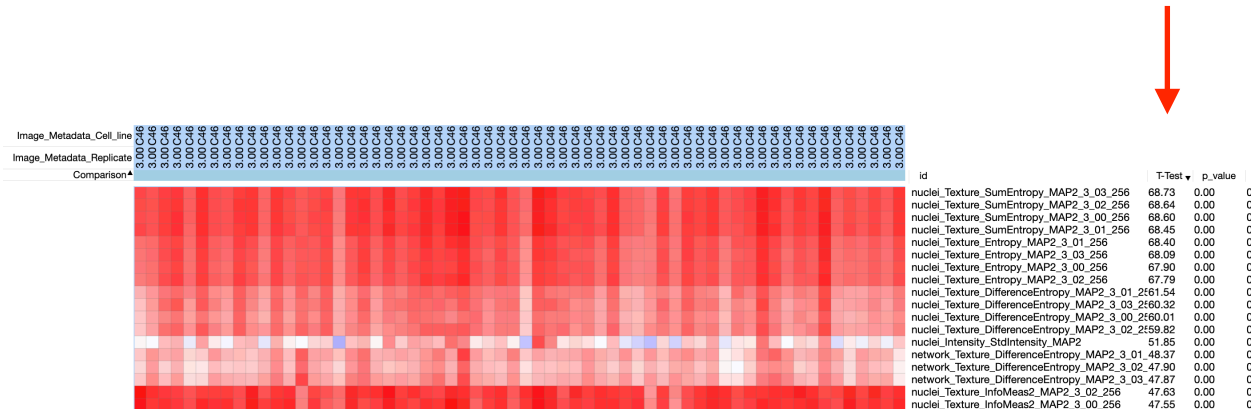


6-No we can calculate the T-test values for the groups desired.
In tools, go to Marker Selection. In the Metric you should select T-test, the field, is the metadata that you want to compare. In this case “cell_line”.
Class A will be the cell line of one group (e.g: WT). Could be one or as many as you want in that group.
Class B, the second group, e.g, mutant.





7- The result will be displayed like the image below. Then, you can sort the T-test values by the absolute values, which will give you the features with more difference. If is positive will be a difference in the same direction and if is negative will be a difference in an opposite direction. All the p_values are pretty low, then many of all the features measured are possible to use for the PCA.



id	T-Test	p_value	
nuclei_Texture_SumEntropy_MAP2_3_03_256	68.73	0.00	0
nuclei_Texture_SumEntropy_MAP2_3_02_256	68.64	0.00	0
nuclei_Texture_SumEntropy_MAP2_3_00_256	68.60	0.00	0
nuclei_Texture_SumEntropy_MAP2_3_01_256	68.45	0.00	0
nuclei_Texture_Entropy_MAP2_3_01_256	68.40	0.00	0
nuclei_Texture_Entropy_MAP2_3_03_256	68.09	0.00	0
nuclei_Texture_Entropy_MAP2_3_00_256	67.90	0.00	0
nuclei_Texture_Entropy_MAP2_3_02_256	67.79	0.00	0
nuclei_Texture_DifferenceEntropy_MAP2_3_01_25	61.54	0.00	0
nuclei_Texture_DifferenceEntropy_MAP2_3_03_25	60.32	0.00	0
nuclei_Texture_DifferenceEntropy_MAP2_3_00_25	60.01	0.00	0
nuclei_Texture_DifferenceEntropy_MAP2_3_02_25	59.82	0.00	0
nuclei_Intensity_StdIntensity_MAP2	51.85	0.00	0
network_Texture_DifferenceEntropy_MAP2_3_01_	48.37	0.00	0
network_Texture_DifferenceEntropy_MAP2_3_02_	47.90	0.00	0
network_Texture_DifferenceEntropy_MAP2_3_03_	47.87	0.00	0
nuclei_Texture_InfoMeas2_MAP2_3_02_256	47.63	0.00	0
nuclei_Texture_InfoMeas2_MAP2_3_00_256	47.55	0.00	0

8- To zoom out, go to the lens and select Fit to the Windows.