Function: Normalization\_ionomycin()

Filename: Normalization\_ionomycin.R

Description: This function takes drug response file and ionomycin response file from a Calcium Imaging dataset as input and gives out a file with F/Fmax values for all the cells that meet threshold (mean+2st.dev) for both ionomycin and drug. For detailed explanations and variable listings see CaImagingFunctionsRDocumentations.docx.

How to use this function?

1. Install R from <https://cran.r-project.org/>
2. Open R interface 🡪 File 🡪 Change dir … : set to a folder where you will store function files and data files. You have to set this at the beginning of each session
3. Copy and paste Normalization.R, Threshold.R and Threshold\_iono.R files into the folder from step 2
4. In the R interface: File 🡪 Source R code🡪 select a file from step 3, repeat for all the three files
5. Type: Normalization\_ionomycin() in the R console and follow along with the prompt

Important considerations:

Make sure the input file is in .csv format with columns as Time/frames, <however many cells you have>, column with background intensity (obtained by selecting a dark area in the field of view in ImageJ).

Two sample input files are provided: one with ph5 response named “TestR.csv” and another with ionomycin response “ionotest.csv”.

“SampleCalculations.xlsx” shows all the steps that this function performs inside in excel for ease of understanding.