This workflow briefly describes necessary procedures to explore and reproduce the statistical, graphical and imaging-based (examples only) analyses underpinning the article "Reticular adhesions are a distinct class of cell-matrix adhesions that mediate attachment during mitosis" by Lock et al, published in Nature Cell Biology, 2018.

This workflow requires the following free software tools: 1. R (https://www.r-project.org) e.g. v3.5.1
RStudio (https://www.rstudio.com/products/rstudio/download/) e.g. v1.1.453
3. Knime (https://www.knime.com/downloads) e.g. v3.6.0 Knime community nodes including Knime Image Processing & HCS Tools Go to github repository following links defined in Download respoitory to desired location Lock et al article Unzip knime workflows: Move knime workflows to Reticular_Adhesion_Arp23_Inhibitor_Combining_All_Exp_Image_Quant_Outputs.zip local knime workspace Reticular_Adhesion_Arp23_Inhibitor_Image_Analysis_Exp1.zip Update input image file path locations in 'Image Reader' node e.g.: LOAD knime 'Image Analysis' workflow .../Source_Data/Knime_Image_Analysis/Arp2-3 Inhibition studies/Input Data/Arp23 Inhibition Images/Exp1 Combined/Exp1_666_Composite_004.tif (Load all images in folder) Update 96 well plate layout file path locations in 'Load Layout' node e.g.: .../Source_Data/Knime_Image_Analysis/Arp2-3 Update 'Table Writer' node (inside Data Export Inhibition studies/Input Data/Arp23 Inhibition metanode) output file path location: this will define the Images/Plate_Layouts/Arp2-3 Inhibitor Exp1.xlsx location of the quantitative data file derived from image analysis (e.g. of Arp2/3 inhibition experiment 1), e.g. .../Source_Data/Knime_Image_Analysis/Arp2-3 **RUN** knime 'Image Analysis' workflow Inhibition studies/Input Data/Table Repeat for image data sets from Exp2 & Exp3 Outputs/Arp2-3_Exp1_Table_All_Combined.table (changing input and output paths/names as This will be the location of the file imported into Exp1 appropriate) import node of the 'Combining Data' knime workflow. Update input data table path locations in 'Table Reader' node (within 'Import' metanode), e.g.: LOAD knime 'Combining Data' workflow .../Source Data/Knime Image Analysis/Arp2-3 Inhibition studies/Input Data/Table Outputs/Arp2-3_Exp1_Table_All_Combined.table (Repeat for Exp2 and Exp3 'Import' metanodes) Update output .csv file path location in 'CSV Writer' node, e.g.: .../Source Data/Knime Image Analysis/Arp2-3 **RUN** knime 'Combining Data' workflow Inhibition studies/Output Data/Arp2-3 Three Experiment Copy resulting data into sheet 'SuppFig5C' of excel file: Integrated Data for R.csv "Supplementary Table 1 Statistics Source Data.xlsx" LOAD R markdown file Update: > data_input_path <- ".../Source_Data"</pre> "Statistical Analysis Code to match location path of "Supplementary Table 1 Lock et al NCB 2018.Rmd" Statistics Source Data.xlsx" **RUN** R markdown file Graphical outputs will be saved to:

"data_input_path/Graphical_Outputs/"
An HTML summary file (preview) will be saved to:
 "data_input_path/Statistical_Analysis_
 Code Lock et al NCB 2018.nb.html"

(required packages will load automatically)