

- 68036\_screenshot\_1.mp4
  - 2.7.1 (Activating 488 nm laser and selecting Live mode) 00:03-00:10
  - 2.7.2 (Adjusting focus and illumination settings) 00:11-00:20
  - 2.8.1 (Initiating acquisition recording clicking in Start) 00:21-00:30
- 68036\_screenshot\_2.mp4
  - 3.1.1 (Opening the microscopy software and selecting Quantification mode) 00:03-00:05
  - 3.1.2 (Displaying fluorescence spike analysis before and after capsaicin stimulus) 00:06-00:09
  - 3.2.1 (Drawing an ROI over a responsive cell, ensuring coverage of the entire cell area) 00:10-01:05
  - 3.3.1 (Navigating to the export option and saving the data as a CSV file) 01:06-01:21
- 68036\_screenshot\_3.mp4
  - 3.4.1 (Opening the FIJI software and displaying the home interface) 00:00-00:01
  - 3.5.1 (Navigating to File > Import > Bio-Formats and selecting the image file) 00:02-00:16
  - 3.6.1 (Using the Magnifying Glass tool to zoom into the image) 00:17-00:20
  - 3.6.2 to 3.7.1 (Drawing an ROI around a responsive cell using the Freehand Selection tool and Adding an ROI to the ROI Manager) 00:21-00:55
  - 3.8.1 (Selecting Analyze > Set Measurements in main menu > Check Mean Gray Values) 00:56-01:03
  - 3.8.2 (Navigating to ROI Manager and selecting all ROIs) 01:04-01:12
  - 3.9.1 (Selecting More > Multi Measure in the ROI Manager. A new window displaying fluorescence intensity measurements across multiple frames appears) 01:13-01:21
  - 3.10.1 (Selecting File > Save As and saving the fluorescence intensity data as a CSV file.) 01:22-01:43
- 68036\_screenshot\_4.mp4
  - 3.12.1 (Opening Google Sheets and uploading the CSV file) 00:00-00:19
  - 3.13.1 (Calculating normalization for each time point) 00:20-01:14
  - 3.14.1 (Calculating  $F_0$  for each ROI) 01:20-01:33
  - 3.15.1 (Calculating  $F(t)$  for each ROI) 01:41-02:01
  - 3.16.1 (Calculating  $\Delta F/F$  for each ROI) 02:06-02:58 - 03:03-03:13 - 03:18-03:40
  - 3.17.1 (Pasting data into GraphPad) 03:47-03:49 - 04:01-04:03 - 04:08-04:10
  - 3.18.1 (Choosing scatter plot graph) 04:15-04:29 - 04:36-04:39