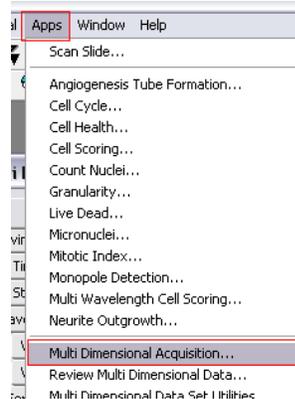




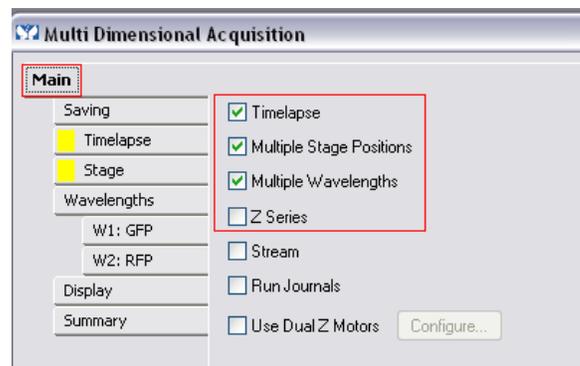
Multi Dimensional Acquisition & Data Review

1. Multi Dimensional Acquisition

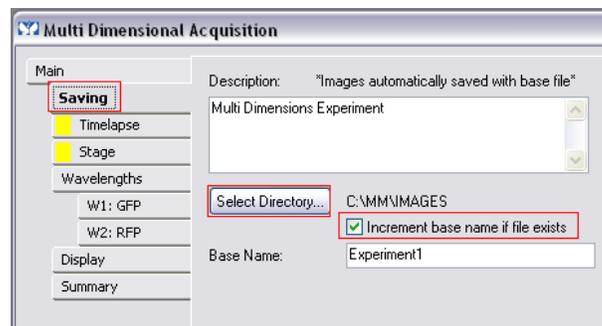
1.1. Call up “Multi Dimensional Acquisition” by click on “Apps” and “Multi Dimensional Acquisition”.



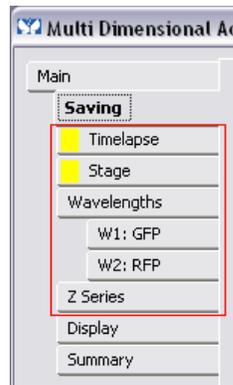
1.2. In the “Main” tab, choose the acquisition combinations you need to do.



1.3. Next go to “Saving” tab to define where you want to save your image. Click on “Select Directory” to choose the folder. Check the option “Increment base name if file exists” so that the file name would expand automatically.

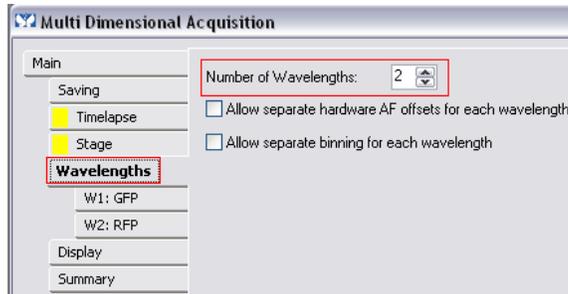


1.4. For the rest of acquisition related tab, define them one by one when applicable.

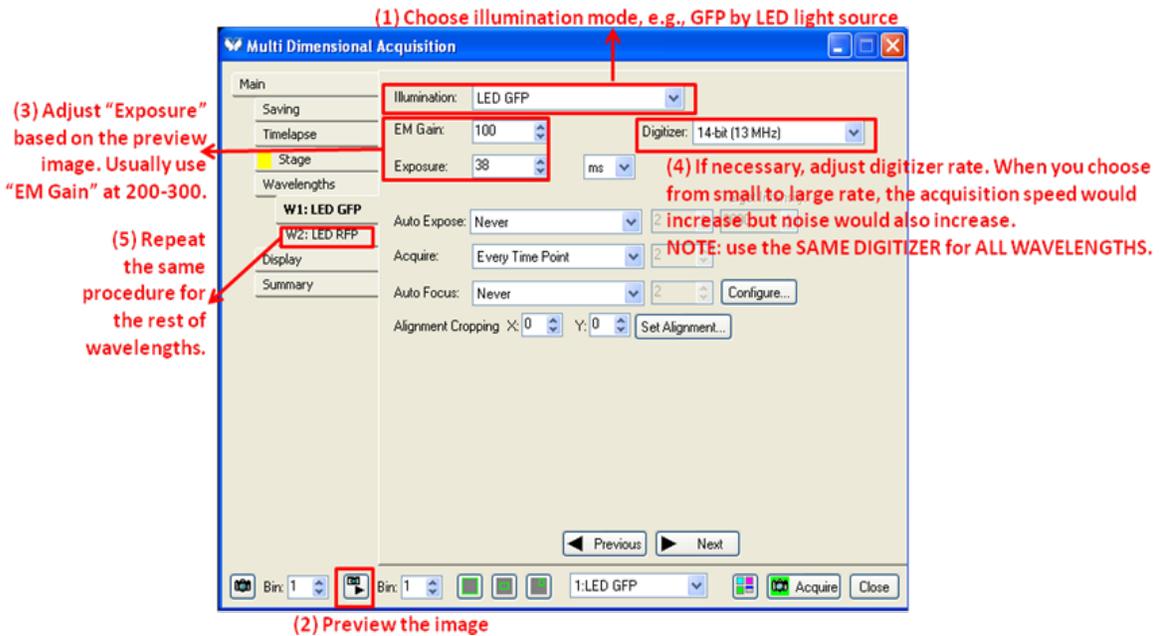


1.5. Wavelength

- a. First define wavelengths. Click on “Wavelengths” and type total wavelengths you want to observe, e.g., 2.



- b. Then go to “W1” and “W2” to define acquisition conditions for each wavelength accordingly.



1.6. Z Stack

(1) Click on "Z Series"

(2) Get the live preview image

(3) Uncheck the option "Range around Current"

(4) Adjust the focus knob until you see one end of the sample, then click on "Set Top To Current"

(5) Turn the focus knob the other way around, until you see the other end of the image, then click on "Set Bottom To Current".

(6) Define "Step Size" you want the Z motor to move at each step when taking the Z stack. Then "Number of Steps" would be calculated automatically. Vice versa.

(7) Choose the "Loop order" you preferred. 2nd option would take less time to complete the whole experiment.

1.7. Multiple Stage Positions

(1) Click on "Stage"

(2) Get the live preview image

(3) Move the joy stick while looking at the live image, until you find one of your region of interest

(4) If you would use ZDC, turn the focus knob to find the best focus, then click on "Find Offset" button. You shall hear one beep sound coming from the microscope.

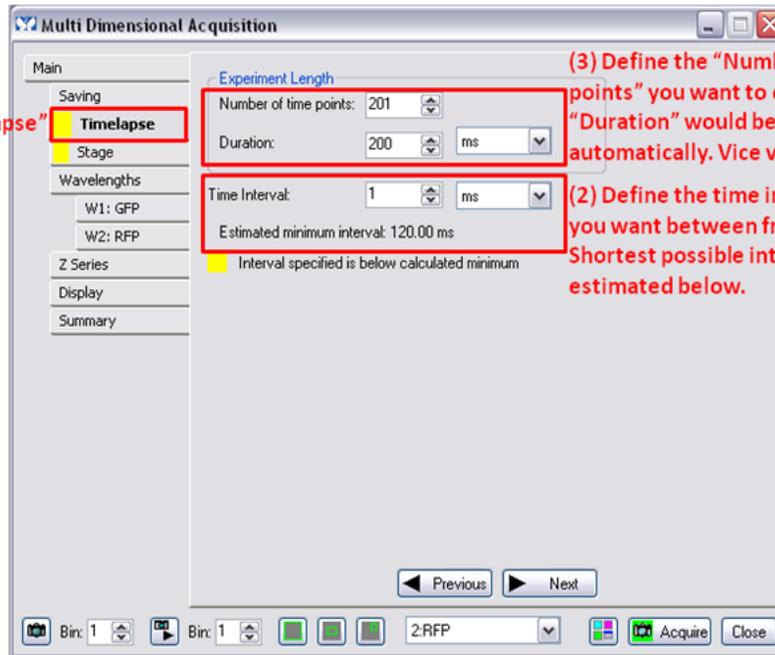
(5) Click on the "Add" button to register current stage position, i.e., its X-, Y-, Z-position and ZDC offset.

(6) Move joy stick again until you find next region of interest, then repeat Step (4) and (5).



1.8. Timelapse

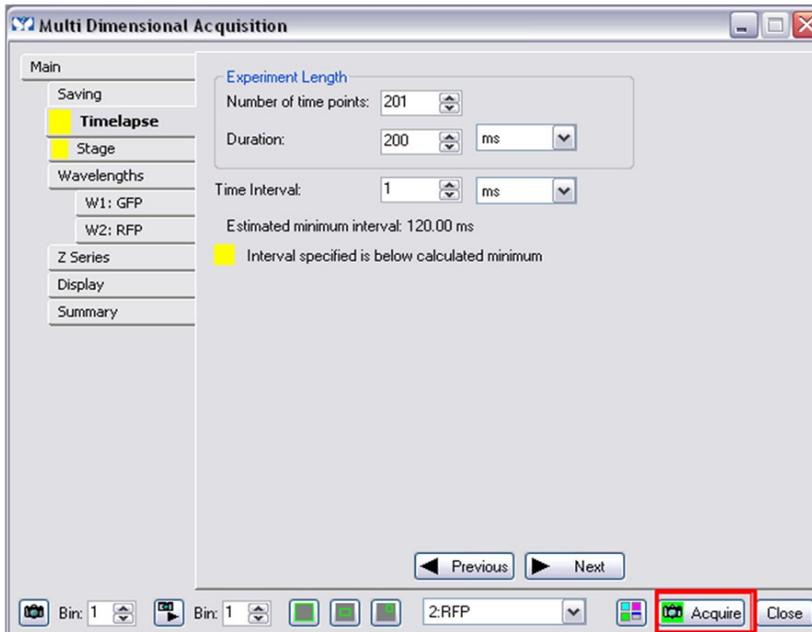
(1) Click on "Timelapse"



(3) Define the "Number of time points" you want to capture, then "Duration" would be calculated automatically. Vice versa.

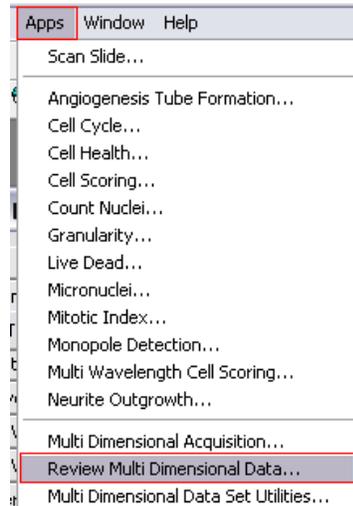
(2) Define the time interval you want between frames. Shortest possible interval is estimated below.

1.9. Click on "Acquire" to start the multi dimensional acquisition.

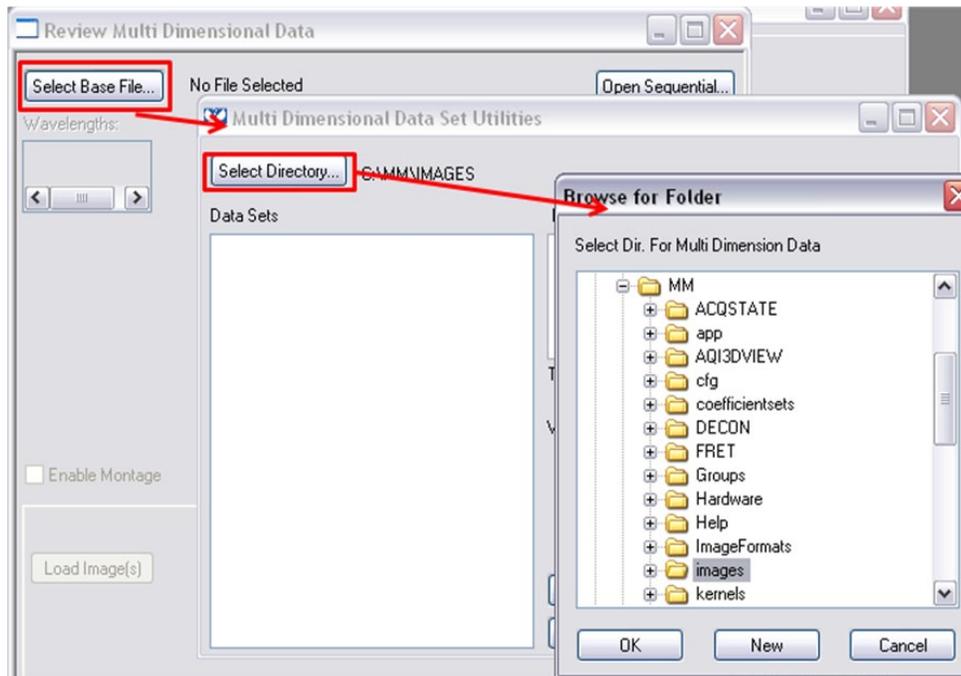


2. Data Review

2.1. Go to “Apps” -> “Review Multi Dimensional Data”



2.2. In the window pop-up, click on “Select Base File”. In the new window pop-up, click on “Select Directory” and choose the folder where you save your multi dimensional image files. Then click “OK”.



2.3. All the images saved inside that folder would be listed under “Data Sets”. Choose the image file you want to review and click on “View”.



2.4. Combine multi-color and load the timelapse file as a movie.

(5) Mouse right-click on this cell. Then all frames would be selected. Otherwise right-click on each individual frames you want one by one to load. Selected frame would be marked with a “X”.

(1) Select the colors you want to combine

(2) Choose “Display” tab

(3) Select “Color Composite”.

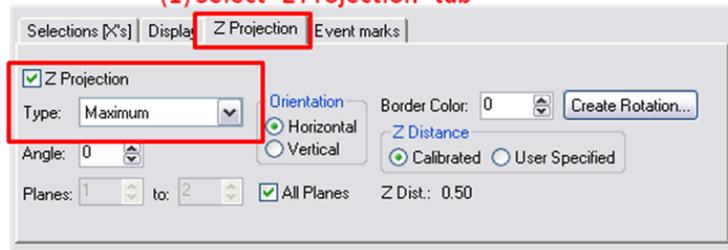
(4) Assign the pseudo color you want to give to each wavelength accordingly. For example, wavelength “CSU RFP” is given red.

(6) Select “Selections [X’s]” tab

(7) Click “Load Image (s). Movie with multi-color would be loaded

2.5. Load multi-color Z-stack timelapse

(1) Select "Z Projection" tab



(2) Tick "Z Projection" and choose "Maximum" as the "Type" for fluorescence images. If it's transmission image, choose "Minimum" as the "Type".

(3) Then repeat Step (2.4) to load the image.