

## **MetaXpress® 6 Software Guide**

Enabling the Multi-Well Acquisition Feature

DEVICES

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#### **Chapter Purpose**

The purpose of this chapter is to guide the user through enabling the **Multi-Well** acquisition feature available on the **Sites to Visit** tab in **Plate Acquisition Setup**.

This feature allows four wells of a multi well plate to be captured in a single image with a low magnification objective. This feature is used by pairing a 4X objective with a 1536 well plate or 2X objective with a 384 well plate.

The primary advantage of multi-well acquisition is speed. Using this feature, a 1536 well plate can be acquired in the same time as a 384 well plate and a 384 well plate can be acquired in the same time as a 96 well plate.





#### How Does Multi-Well Acquisition Work?

The camera's larger field of view enables the capture of 4 wells in one image

- MetaXpress automatically parses (splits) the image into the 4 corresponding wells
- Individual wells are saved and viewed as single images in **Review Plate Data**







- 1. Open Plate Acquisition Setup
  - In the main toolbar click on



#### OR

- Under the Screening menu, select Plate Acquisition Setup
- 2. Click on the **Configure** tab





- 3. Select the Objective and Camera tab
- 4. Select the appropriate **Magnification** from the drop-down menu
  - For 1536 well plates, select 4X
  - For 384 well plates, select **2X**
- 5. Set **Camera Binning** to **1** (maximum resolution)
- 6. If **Gain** options are present, set to **Low**



- 7. Select the Plate tab
- 8. Select the appropriate **Plate Type** from the drop-down menu









- 9. Select the appropriate wells to be imaged in the **Plate Section** 
  - Due to the camera's larger field of view, blocks of 4 wells (2 x 2) will be imaged whether or not they are selected (green)
  - Deselected wells (gray) will not be saved after parsing
  - Molecular Devices recommends to plate samples and select wells in adjacent columns and rows





Objective and Camera- 4X S Flu	Site Options	Custom field of view (%):	Well size: 2 mm <sup>2</sup>
Plate- Corning 1536-well Black-	Single site	×: 50 🗇 Y: 50 🜩	Number of sites: 1
Sites to Visit- multi-well	Adaptive acquisition	Ste /mage size: 1 /3 x 1 /3 mm	127.11% Well Coverage
Acquisition	Multi-well	Site/inage size. 1.45 x 1.45 min	
Autofocus	Acquires a single site in	4 (2 x 2) wells simultaneously	
Wavelengths	reducing plate acquisiti	on time	
W1 DAPI	Not for use with fluidics	or device/camera journal events	
W2 FITC			
Display			

- 10. Select the Sites to Visit tab
- 11. Select Multi-well under the Site Options section
  - Upon selecting Multi-well, the well view automatically changes to single site view



- To reduce the image size, enable Custom field of view (%) and adjust the X and Y values
  - This % change in size will be applied to each well after parsing
  - Custom field of view can be used to avoid acquiring the well edge





Objective and Camera- 4X SF	Autofocus options
Plate- 1536 Well Plate	Enable laser-based focusing
Sites to Visit- multi-well	Enable image-based focusing (for acquisition or laser recovery)
Acquisition	Acquisition options
Autofocus	Acquire Time Series
Wavelengths	Acquire Z Series
W1 DAPI	
W2 FITC	
Display	
	Run Journals During Acquisition
	Analyze Images After Acquisition
	Perform shading correction Directory C:\

- 13. Select the **Acquisition** tab
- 14. Enable the appropriate Autofocus options
  - Enable laser-based focusing
  - If required, Enable image-based focusing
- 15. Enable the appropriate **Acquisition options**





Objective and Camera- 4X S Flu	Laser-based Focusing
Plate- Corning 1536-well Black-	Configure Laser Settings
Sites to Visit- multi-well	Well to well autofocus Focus on plate bottom, then offset by bottom thickness
Autofocus	Image-based Focusing
Wavelengths	Algorithm: Standard Binning: 2 A Custom exposure times
W1 DAPI	
W2 FITC	Allow image-based focusing for recovery from laser-based well bottom failures
Display	
	Initial well for finding sample First well acquired
	Number of wells to attempt initial find sample 1

- 16. Select the **Autofocus** tab
- 17. Set Well to well autofocus to Focus to Plate bottom then offset by bottom thickness
  - Selecting either of the other choices will result in a red box displayed on the Autofocus tab





- 18. Select the appropriate **W** tab (wavelength)
- 19. Configure the wavelength settings as you would for acquisition
  - i. Select correct **Illumination Setting** from the drop-down menu
  - ii. Calculate focus offset
  - iii. Determine exposure time
- 20. Repeat steps 18-19 for subsequent wavelengths
- 21. Complete the rest of acquisition setup as you would normally





#### Limitations of Multi-Well Crop

- Not compatible with fluidics. If Multi-well is selected on the Sites to Visit tab, a red box will appear on the Fluidics tab.
- Some journals that control the camera or hardware devices are incompatible. Refer to accompanying documentation for details or contact Technical Support.
- On the **Timelapse** tab, when selecting the **Perform time series for** option, multi-well crop is not compatible with **One row then the next** and **One column then the next** choices.





#### Support Resources

- F1 / HELP within MetaXpress® Software
- Support and Knowledge Base: <u>http://mdc.custhelp.com/</u>
- User Forum: <a href="http://metamorph.moleculardevices.com/forum/">http://metamorph.moleculardevices.com/forum/</a>
- Request Support: <u>http://mdc.custhelp.com/app/ask</u>
- Technical Support can also be reached by telephone:
  - 1 (800) 635-5577
  - Select options for Tech Support → Cellular Imaging Products → ImageXpress Instruments





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