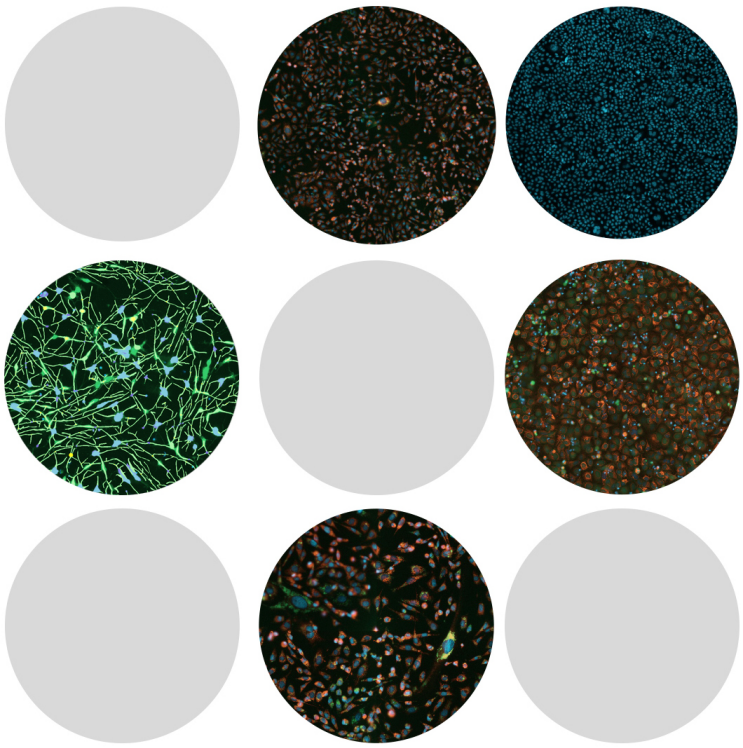

CellReporterXpress

Image Acquisition and Analysis Software

Version 2.0

User Guide

MOLECULAR DEVICES

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June 2018

Product manufactured by Molecular Devices, LLC.
3860 N. First Street, San Jose, California, 95134, United States of America.
Molecular Devices, LLC is ISO 9001 registered.

Contents

Chapter 1: CellReporterXpress Image Acquisition and Analysis Software

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The Molecular Devices® CellReporterXpress Image Acquisition and Analysis Software is the user interface for the Molecular Devices ImageXpress® Pico Automated Cell Imaging System.

The CellReporterXpress Software integrates image acquisition and analysis into a unified workflow. Along with the imaging device, the CellReporterXpress Software is part of a system that streamlines automated imaging to offer a simplified solution for scaling up microscopy. Its features include:

- A web-based interface that runs on many browsers, including those found on iPads and Android tablets.
- Over 25 available predefined experimental protocols.
- High-powered analysis tools equivalent to those found in desktop applications.
- Easy-to-manage data with no requirement to configure a database.
- A simplified user interface that is easy to learn and easy to use.

Obtaining Support

Molecular Devices is a leading worldwide manufacturer and distributor of analytical instrumentation, software, and reagents. We are committed to the quality of our products and to fully supporting our customers with the highest level of technical service.

Our Support website, www.moleculardevices.com/support, has a link to the Knowledge Base with technical notes, software upgrades, safety data sheets, and other resources. If you still need assistance, submit a request to Molecular Devices Technical Support.

Documentation

Review the product documentation on the Knowledge Base, including installation guides and user guides. In addition, online Help is available within the CellReporterXpress Software. Press **F1** to access Help for the active screen.

Technical Support

You can contact Molecular Devices Technical Support by phone or submit a support request through the Knowledge Base. To find regional support contact information, visit www.moleculardevices.com/contact.

You will need the instrument serial number and the software system ID.

Additional Resources

Web-based microscopy courses:

- www.leica-microsystems.com/science-lab
- www.ibiology.org/ibioeducation/taking-courses/ibiology-microscopy-short-course.html

The Molecular Probes Handbook offers advice on fluorescent probes and can help you determine if there are better stains available for your analysis:

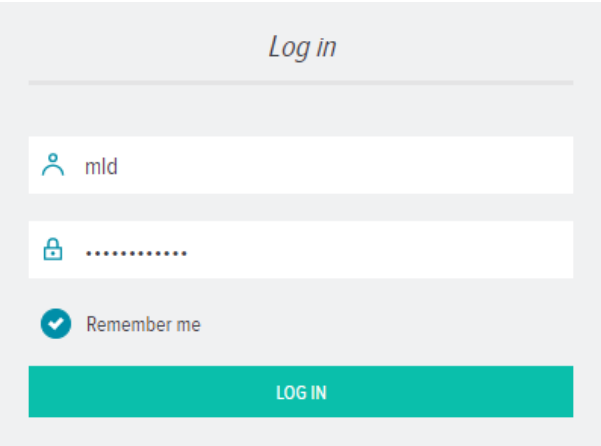
- www.lifetechnologies.com/us/en/home/references/molecular-probes-the-handbook.html

Logging In to the Software

To log in to the CellReporterXpress Software:

1. With a Windows computer using Google Chrome, do one of the following to display the CellReporterXpress Log In screen:
   - On the desktop, double-click  .
   - Click **Start > Molecular Devices > MD.CellReporterXpress**.
2. With a non-Windows computer, a Windows computer using Mozilla Firefox, or a tablet, do the following to display the CellReporterXpress Log In screen:
   a. Open a supported browser.
   b. In the Address bar, enter either the IP address or the host computer name along with the port being used by the remote client (by default, 80) in the following format:
   `http://address:port`
   For example, if the host computer is named CellReporterXpress, enter:
   `http://CellReporterXpress:80`
   Or, if the host computer's IP address is 10.133.30.151, enter:
   `http://10.133.30.151:80`

   Note: If you do not know the IP address or the name of the CellReporterXpress host computer, contact your IT professional.



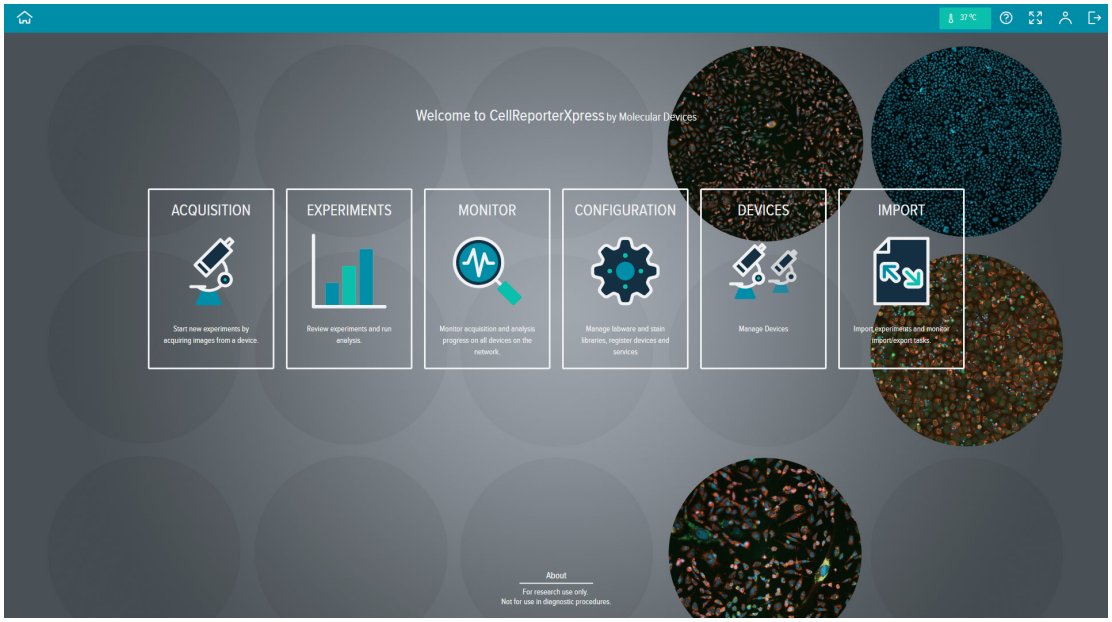
3. In the **Login** field, enter the Windows system user name.
4. In the **Password** field, enter the Windows system password.
5. Click **LOG IN**.

About This Guide

This guide is intended for the scientist using the CellReporterXpress Software. It is a PDF version of the Help that is integrated into the CellReporterXpress Software.

The information in this guide is subject to change without notice. Molecular Devices recommends that you review the guide on the Knowledge Base for the most up-to-date information.

Chapter 2: Home Page



The **Home** page is the first page displayed when you log in to the CellReporterXpress Software. It contains the following tiles that enable you to access the software modes:


Acquisition: Click to configure experiment settings and run experiments from supported instruments using protocols or templates. See Acquisition Mode on page 11 for details.


Experiments: Click to view images and analysis data collected in **Acquisition** mode and perform additional offline analysis. See Experiments Mode on page 69 for details.


Monitor: Click to view the progress and completion status of experiments run from **Acquisition** mode or **Experiments** mode. See Monitor Mode on page 137 for details.


Configuration: Click to set the systemwide options that affect all users of the CellReporterXpress Software. See Configuration Mode on page 139 for details.

 **Devices**: Click to manage and configure instruments for acquisition, including installing and calibrating objectives and filter cubes and controlling the temperature inside the instrument. See Devices Mode on page 151 for details.

 **Import**: Click to download and install the MD Import/Export Service, which enables you to import experiment data from temporary storage and export raw data from experiments. See Import Mode on page 163 for details.

Some of the tiles will be used often. Others will be used infrequently after you have set up the system.

The toolbar at the top of the CellReporterXpress window is always available. On the left are page navigation tabs, which are a "breadcrumb trail" indicating the path you used to get to the currently displayed page. To return to a previous page, click on that previously visited page tab.

The right side of the toolbar includes the following icons:

 **Help**: Opens the Help.

 **Full Screen**: Expands the software window to fill the entire screen of your computer or tablet.

 **User Preferences**: Enables you to specify the way the software looks and, for some functions, to specify the way the functions work. The options you set become your personal preferences and stay set every time you use the software. See Configuration Settings on page 167 for details.

 **Log out**: Exits the CellReporterXpress Software and returns to the Log In page.

The toolbar may also contain a toolbar notification for the temperature sensor:

Temperature: Click to open the temperature control panel. See Sensors on page 153 for details.

Chapter 3: Acquisition Mode

3

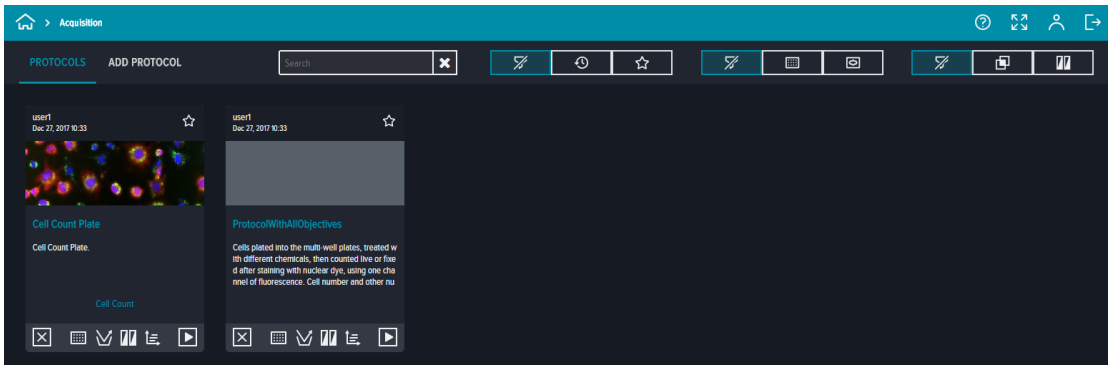
Use **Acquisition** mode to configure experiment settings and run experiments from supported instruments using protocols or templates.

On the **Home** page, click the  **Acquisition** tile to enter **Acquisition** mode. The **Protocol** library appears.

Protocol Library

A protocol enables you to reuse a specific configuration for an experiment. It includes no experiment data, only configuration settings. A typical protocol enables you to insert labware into the instrument, select the protocol, and then run the protocol to perform the experiment and collect data.

A protocol is created from a template. When you click **Add Protocol** in the Protocol library, the Template library appears. Each template guides you through the configuration process for a particular experiment. When you save a modified template, it becomes a protocol in the Protocol library. See Template Library on page 13 for details.



Each protocol in the library is displayed as a card. The cards are listed in alphabetical order by protocol name. Along with the protocol name and description, each card indicates the user who created the protocol, the date and time of creation, a  **Favorite** icon (that you can use to flag frequently used protocols), a  **Delete** icon, and a  **Run** icon.

In addition, each card contains icons to indicate protocol properties, including:

 **Locked**

 **Plate** or  **Slide**

 **Fluorescence** or  **Colorimetric**

 **Stitched** or  **Tiled**

 **Analysis On** or  **Analysis Off**

From the Protocol library, you can run a protocol, modify a protocol, add a protocol, or delete a protocol.

Running a Protocol

To run a protocol:

1. Click the card you want to run.
2. Click  **Run**. See Run Protocol on page 41 for details on running a plate protocol or Run Protocol on page 65 for details on running a slide protocol.

Modifying a Protocol

To modify a protocol:

1. Click the card you want to modify.
2. Go to each workflow step you want to modify and make changes as needed. See Plate Acquisition Workflow on page 16 or Slide Acquisition Workflow on page 44 for details.
3. Click  **Save Protocol**. See Run Protocol on page 41 for details on running a plate protocol or Run Protocol on page 65 for details on running a slide protocol.

Deleting a Protocol

To delete a protocol:

1. Click the card you want to delete.
2. Click  **Delete**.

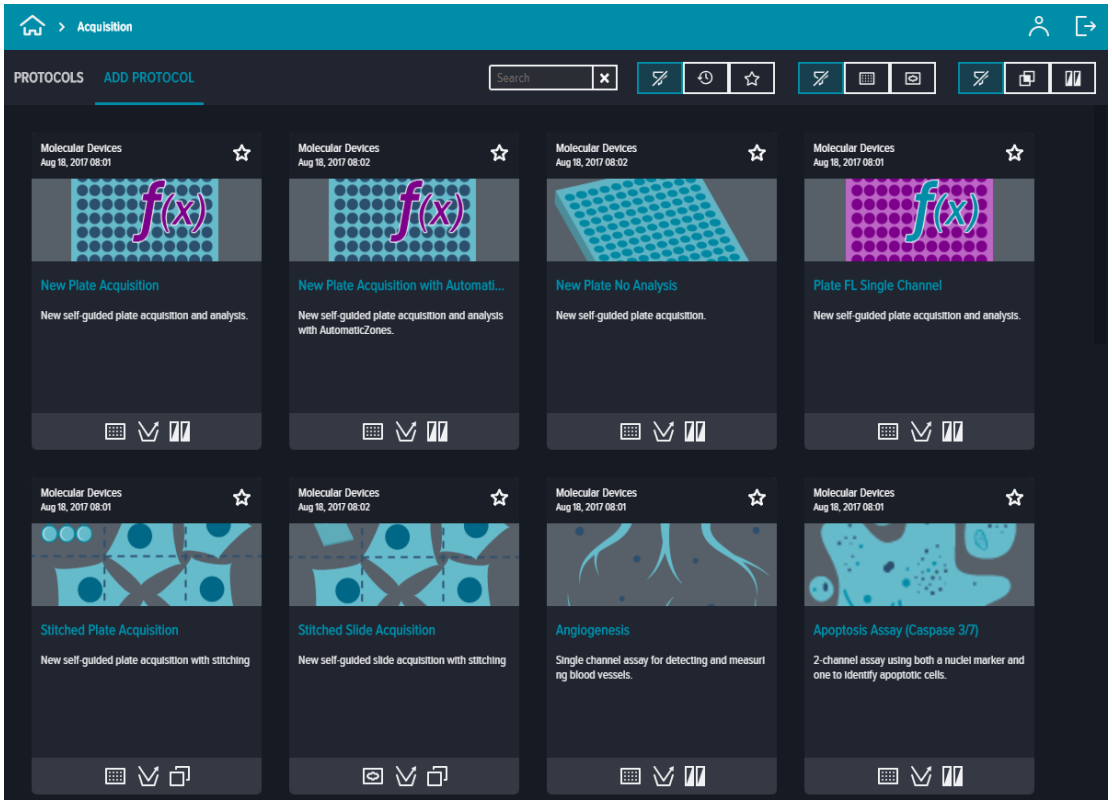
Adding a Protocol

To add a protocol, see Template Library on page 13.

Template Library

When you click **Add Protocol** in the Protocol library, the Template library appears. Each template guides you through the configuration process for a particular experiment. Most templates are designed for typical experiments and have restricted settings options. The restrictions help you focus on the most appropriate options for that experiment type. Two unrestricted templates (**New Plate Acquisition** and **New Slide Acquisition**) allow you to select from all the available experiment settings options.

Note: You cannot create new templates. When you save a template, it becomes a protocol. See Protocol Library on page 11 for details.



Each template in the library is displayed as a card. The cards are listed in alphabetical order by template name. Along with the template name, each card indicates the template description, a **Favorite** icon (that you can use to flag frequently used protocols). In addition, each card contains summary icons to indicate template settings, including:

Plate or **Slide**

Fluorescence or **Colorimetric**

Stitched or **Tiled**

There are two template types: Fluorescence templates and Transmitted Light (TL) templates.

Use Fluorescence templates to perform the following assays:

- **Angiogenesis Skeletonization**: Single channel analysis for detecting and measuring blood vessels.
- **Apoptosis**: Two-channel analysis using a nuclei marker and a marker to identify apoptotic cells.
- **Autophagy**: Two-channel assay for detecting autophagic granules.
- **Cell Count**: Single-channel assay for counting cells based on a nuclei stain.
- **Cell Differentiation**: Two-channel assay using a nuclei marker and a marker to identify differentiated cells.
- **Cell Scoring**: Two-channel assay for scoring cells based on a marker.
- **Endocytosis**: Two-channel assay for detecting endosomes.
- **Internalization**: Two-channel assay for detecting internalizing granules.
- **Live Cells**: Two-channel assay using a nuclei marker for all cells and a second marker to identify live cells.
- **Lysosomal Degradation**: Two-channel assay for detecting lysosomes.
- **Mitochondria**: Two-channel assay for detecting mitochondria.
- **Mitotic Index**: Two-channel assay using a nuclei marker and a second marker to identify mitotic cells.
- **Neurite Tracing**: Single-channel assay for measuring neurite outgrowth.
- **Phagocytosis**: Two-channel assay for detecting phagocytic vacuoles.
- **Pits and Vesicles**: Two-channel assay for detecting GPCR pits and vesicles.
- **Protein Expression Index**: Two-channel assay using both a nuclei marker and one to measure protein expression.
- **Translocation**: Two-channel assay for the quantification of cellular signaling events and intracellular trafficking.
- **Viral Infectivity**: Two-channel assay using both a nuclei marker and a marker to detect cells infected with a virus.

Use Transmitted Light (TL) templates to perform the following assays:

- **Cell Count, Beads**: Single-channel, transmitted light assay to find beads.
- **Cell Count, General**: Single-channel, transmitted light assay to find cells using a method optimized for best area coverage and a wide variety of cells.
- **Cell Count, Large Cells**: Single-channel, transmitted light assay to find larger cells, such as Hela.

- **Cell Count, Small Cells**: Single-channel, transmitted light assay to find smaller cells, such as CHO.
- **Cell Scoring, Beads**: Two-channel, transmitted light assay to find beads, then scoring for an additional fluorescence channel.
- **Cell Scoring, General**: Two-channel, transmitted light assay to find a range of cells, then scoring for an additional fluorescence channel.
- **Cell Scoring, Large Cells**: Two-channel, transmitted light assay to find larger cells, such as Hela, then scoring for an additional fluorescence channel.
- **Cell Scoring, Small Cells**: Two-channel, transmitted light assay to find smaller cells, such as CHO, then scoring for an additional fluorescence channel.

Search and Filters

To limit the number of visible cards on a page, use the **Search** field and filter icons at the top of the Protocol library and the Template library.

Using Search

Use search to find specific words in the titles and descriptions of protocols or templates.

To use search:

1. Click in the **Search** field.
2. Enter the word you want to search, and press **ENTER**.

Using Filters

Use the filter icons to control which template cards or protocol cards are shown. Active filter icons are highlighted. By default, **All** is the active filter. Filter options include:

Recent or **Favorites**

Plates or **Slides**

Stitched or **No stitching**

To use filter icons:

Click the filter icon you want to use. The icon is highlighted and only the cards that match the filter option are shown.

Plate Acquisition Workflow

Select a plate template to begin the plate acquisition workflow. The **New Plate Acquisition** template is an unrestricted template that allows you to select from all the available plate experiment settings options. Other plate templates may offer restricted settings options to help you focus on the most appropriate options for that experiment type.

The icons in the **Steps** pane on the left side of the page guide you through the plate experiment configuration process. The tools and controls in the pane on the right side of the page vary according to the step being configured and the experiment type.

Note: Depending on the selected template, some steps, tools, and options may not appear or may not be available. Use the **New Plate Acquisition** template to access all steps, tools, and options.

The plate acquisition workflow is as follows:


Acquisition Device is the first step for all acquisition workflows. In this step, you select the device for the acquisition and insert your experiment-ready labware into the instrument. See Acquisition Device on page 18 for details.


Acquisition Settings is the step where you set up image acquisition for the experiment, including the stains and objective to be used. You can preview the image capture and adjust channel identification color, histogram, focus, and exposure settings. See Acquisition Settings on page 19 for details.


Region Selection to Acquire is the step where you select the region of the well to be acquired. The page shows a representation of a round microplate well with a region selection overlay. You must select at least one region to run an experiment. See Region Selection to Acquire on page 28 for details.


Analysis Settings is the step where you set up image analysis for the experiment. The data generated based on these settings becomes the analysis data for your experiment available in **Experiments** mode. See Analysis Settings on page 29 for details.


Region Selection to Analyze is the step where you select the region of a well to be analyzed. You must select at least one region to run an experiment. See Region Selection to Analyze on page 35 for details.

 **Well Selection** is the step where you select the wells to be acquired. The **Well Selection** page shows a plate map for the plate selected in the **Acquisition Settings** step. You must select at least one well to run an experiment. See Well Selection on page 36 for details.

 **Time Series** is an optional step where you set up a time series for image acquisition. This enables you to acquire images at multiple time points. See Time Series on page 37 for details.

 **Save Protocol** is an optional step where you can save the protocol you have created. You will typically save a protocol only when you intend to run it frequently. After you save a protocol, it appears as a card in the Protocol library. See Save Protocol on page 40 for details.

 **Run Protocol** is the last step before you can run the experiment. In this step, you can review the acquisition settings and address any issues. When you run the experiment, the CellReporterXpress Software acquires images and analyzes the data according to your settings. When the experiment completes, the CellReporterXpress Software saves all acquired data as specified. Acquired data is then available for viewing and analysis in **Experiments** mode. See Run Protocol on page 41 for details.

Acquisition Device

 **Acquisition Device** is the first step for all acquisition workflows. In this step, you select the device for the acquisition and insert your experiment-ready labware into the instrument. See the *ImageXpress Pico User Guide* for details on inserting labware into the instrument.

The right side of the page includes the following icons:

 **Shutdown Device**: Prepares the software to power off the selected instrument.

 **Restart Device**: Restarts the selected instrument.

 **Open Plate Door**: Opens the stage door on the selected instrument so that you can insert or remove labware.

 **Close Plate Door**: Closes the stage door.

 **Set Up for Adjustment of Objective Collar**: Moves the objective turret so that you can adjust the correction collar on the selected objective.

 **Finish Adjustment of Objective Collar**: Moves the objective turret back into position after you adjust a correction collar.

The CellReporterXpress Software can control multiple instruments. If your software is configured to control multiple instruments, you may need to select the one you want to use.

 **Note:** If you click the  **Favorite** icon for a device, that device will be selected by default. If you have not specified a favorite device, the last device used will be selected by default.

To select an acquisition device:

On the **Acquisition Device** page, in the **Available Acquisition Devices** list, select the imaging device you want to use.

The selected device is highlighted in the Available Acquisition Devices list.

To continue to the next workflow step, click  **Acquisition Settings**. See Acquisition Settings on page 19 for details.

Acquisition Settings


Acquisition Settings is the step where you set up image acquisition for the experiment, including the stains and objective to be used. You can preview the image capture and adjust channel identification color, histogram, focus, and exposure settings.

The right side of the page includes the following icons:

Plate Format: Specifies the plate format for the acquisition.


Stains: Specifies the stains for the acquisition.

Objectives: Specifies the objective for the acquisition.


Comparison Mode: Captures two preview images, which enables you to compare the uniformity of the image quality or compare settings on two different phenotypes (such as positive and negative controls). See Snapping Well Comparison Previews on page 25 for details.

To configure plate acquisition settings:

1. On the **Acquisition Settings** page, in the **Tools** pane on the right, click 

Plate Format.
2. In the **Plate Format** pane, select the plate format you want to use.
3. Click  **Stains**.
4. In the **Stains** pane, select the stains you want to use.
5. Click  **Move Stain Up** and  **Move Stain Down** as needed to select the order in which the stains will be collected.

   Tip: For an acquisition with transmitted light, acquire the transmitted light stain first or last. If you plan to perform transmitted light segmentation, you will likely want to set the transmitted light stain to be collected first.

6. Click 

Objectives.
7. In the **Objectives** pane, select the objective you want to use.

As part of configuring acquisition settings, you may need to perform the following tasks:

- Adjusting a Correction Collar, see page 20
- Snapping a Preview of a Well, see page 22
- Snapping Well Comparison Previews, see page 25

To continue to the next workflow step, click  **Region Selection to Acquire**. See Region Selection to Acquire on page 28 for details.

Adjusting a Correction Collar

The 40x objective and 63x objective have application-optimized correction collars (CORR) to compensate for external influences such as well bottom thickness or coverslip thickness. The collars have a range of 0 mm to 2 mm correction. Changing this setting adjusts the distances between components inside the objective barrel. Image quality and resolution are very dependent on properly setting these collars.

The settings to be used depend on the well bottom thickness of the plate or the coverslip thickness on the slide on which the specimen is mounted. In general, set the correction collar for the physical thickness of the plate or slide that you are imaging. The physical thickness can be determined by the plate specifications from the plate manufacturer.

Note: Do not use a plate, slide, or coverslip with a thickness that is out of the range of the correction collar for the selected objective.

CAUTION! To prevent skin oils from damaging the optical coatings, Molecular Devices recommends that you wear powder-free disposable gloves when handling objectives and filter cubes.

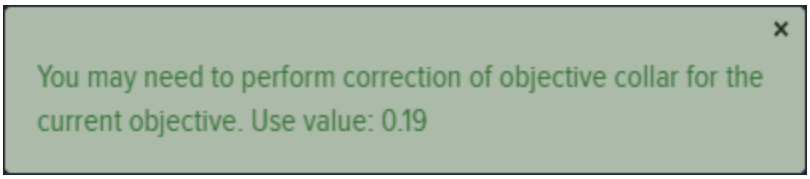
CAUTION! With the instrument power on, do not manually rotate the objective turret. Manually rotating the objective turret can damage the instrument.

You would typically adjust a correction collar as part of setting up an acquisition.

To adjust a correction collar for a plate:

1. On the **Acquisition Settings** page, on the right side of the screen under **Tools**, click  **Plate Format**.
2. In the **Plate Format** list, select the plate format.
3. On the right side of the screen under **Tools**, click  **Objectives**.

4. In the **Objectives** list, select the objective.
   If a correction collar adjustment is required, the CellReporterXpress Software displays the recommended setting for the correction collar based on the thickness of the plate bottom, slide, or coverslip.



5. On the left side of the screen under **Steps**, click  **Acquisition Device**.

6. On the right side of the screen, click  **Set Up for Adjustment of Objective Collar**.

7. Click **OK**.
   The objective door opens.

8. If needed, loosen or remove the objective from the instrument by gently turning it counterclockwise.

9. Rotate the correction collar to its new setting.

> **Tip:** You might need a flashlight to see the markings for the graduated scale on the barrel and its current setting.

10. If you loosened or removed the objective, insert it back in its original slot in the turret or tighten it by gently turning it clockwise.

> **Note:** When installing the objective, take care to avoid changing the correction collar setting.

11. Close the objective door.

12. Click **OK**.

13. Test the correction collar setting by examining the image quality of some test snaps.

14. If the image quality is not satisfactory, repeat these steps to re-adjust the correction collar.

Snapping a Preview of a Well

You can create a preview of the acquisition by snapping an image. The preview uses the selected objective, wavelength, well, region, focus settings, and exposure settings.

To snap an image of a well:

1. Click  **Snap Image** to snap an initial preview image.

2. On the **Acquisition Settings** page, at the bottom of the screen on the  **Choose Well and Region to Acquire** tab, click  **Select Well**.

3. In the plate map, select the well for the preview. The instrument snaps a preview image of the newly selected well. The **A1** well is selected by default.

4. In the single well map, click and drag the  selection tool to select the region of the well for the preview, if needed. The instrument snaps a preview image of the newly selected region. The center area of the well is selected by default.

5. Click  **Focus/Exposure Settings**.

6. On the **Focus/Exposure Settings** tab, adjust the settings as needed for each channel.
   - Use the **Focus offset** controls to adjust the image sharpness. Click the **Mode** dropdown to set one of the following autofocus ranges:
     - **Normal**: Hardware and image autofocus mode using a short search range.
     - **Wide**: Hardware and image autofocus mode using a wide range.
     - **Super Wide**: Hardware and image autofocus mode designed for samples that don't sit directly on the well bottom.
     - **Plate and Well Bottom**: Hardware-only autofocus mode designed for speed.

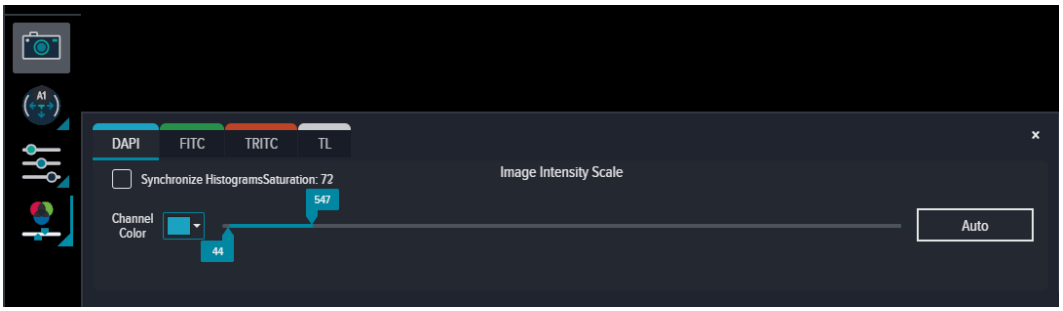
     After you set the focus mode, click **Auto** next to the **Focus Offset** controls to determine the focus offset between stains. This is particularly useful when using the **Plate and Well Bottom** focus mode.
   - Use the **Exposure** controls to adjust the exposure time, which affects image brightness and results in a broader dynamic range (that is, the difference between the brightest and dimmest object detected).

     When your image is in focus, click **Auto** to automatically determine the optimal exposure for the best dynamic range. The longer the exposure, the brighter the experiment image. Dimmer objects get brighter with longer exposure time, as does the background. Increased exposure time slows the experiment because each image takes longer and might lead to image saturation.

     Tip: You may want to use **Auto** with a known bright sample, such as a positive control.

7. Click  **Snap Image** again to refresh the preview.

8. Click  **Image Intensity Settings**.



9. On the **Image Intensity Settings** tab, adjust the settings as needed for each channel.
   - Use the **Channel Color** dropdown to change the identification color for the channel. The default channel color is based on the emission wavelength of the selected stain.
   - Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.
   - Select **Synchronize Histograms** to adjust the image intensity scale for all channels simultaneously.
10. Use the image viewer controls as needed to control the preview:

 : Toggle the available channels by clicking on the channel icons. "Hidden" channels are shown slightly dimmed.

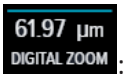
> **Tip:** When the image is overexposed, the channel icons indicate the overexposure  . Lower the **Exposure** value as needed.

 : Zoom in on the image.

 : Zoom out on the image after zooming in.

 : Reset the view zoom to the original image size.

 : View the image full screen.



: View the current zoom scale. **Digital Zoom** indicates that the image is zoomed in digitally (that is, not optically) and might not represent the actual quality of the image.

11. Repeat these steps as needed until you are satisfied with the quality of the preview.

> **Tip:** The **History** pane contains thumbnails of previous preview images. Click a thumbnail to show that preview image and revert the settings to the configuration used to acquire it.

To continue to the next workflow step, click  **Region Selection to Acquire**. See Region Selection to Acquire on page 28 for details.

Snapping Well Comparison Previews

You can create previews of the acquisition by snapping comparison images from two wells (or two regions of the same well). This enables you to compare the uniformity of the image quality or compare settings on two different phenotypes (such as positive and negative controls). As with snapping an image of a single well, the comparison images use the selected objective, wavelengths, well, region, focus settings, exposure settings, and histogram.

To snap well comparison images:

1. Click  **Snap Image** to snap an initial preview image.
2. On the **Acquisition Settings** page, in the **Tools** pane on the right, click  **Comparison Mode**.
3. At the bottom of the screen on the  **Choose Well and Region to Acquire** tab, click  **Select Well** on the left.
4. In the plate map, select the first well for the preview. The instrument snaps a preview image of the newly selected well. The **A1** well is selected by default.

5. Click  **Select Well** on the right.
6. In the plate map, select the second well for the preview. The instrument snaps a preview image of the newly selected well. The bottom right well is selected by default.

7. In the single well map on the left, click and drag the  selection tool to select the region of the first well for the preview, if needed. The instrument snaps a preview image of the newly selected region. The center area of the well is selected by default.
8. Repeat steps 6 and 7 in the single well map on the right to select the region of the second well for the preview, if needed.
9. Click  **Focus/Exposure Settings**.

10. On the **Focus/Exposure Settings** tab, adjust the settings as needed for each channel.
    - Use the **Focus offset** controls to adjust the image sharpness. Click the **Mode** dropdown to set one of the following autofocus ranges:
      - **Normal**: Hardware and image autofocus mode using a short search range.
      - **Wide**: Hardware and image autofocus mode using a wide range.
      - **Super Wide**: Hardware and image autofocus mode designed for samples that don't sit directly on the well bottom.
      - **Plate and Well Bottom**: Hardware-only autofocus mode designed for speed.

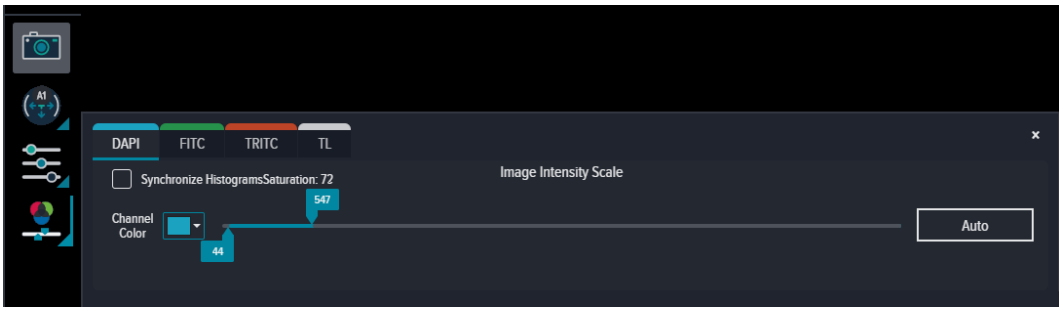
      After you set the focus mode, click **Auto** next to the **Focus Offset** controls to determine the focus offset between stains. This is particularly useful when using the **Plate and Well Bottom** focus mode.
    - Use the **Exposure** controls to adjust the exposure time, which affects image brightness and results in a broader dynamic range (that is, the difference between the brightest and dimmest object detected).

      When your image is in focus, click **Auto** to automatically determine the optimal exposure for the best dynamic range. The longer the exposure, the brighter the experiment image. Dimmer objects get brighter with longer exposure time, as does the background. Increased exposure time slows the experiment because each image takes longer and might lead to image saturation.

      Tip: You may want to use **Auto** with a known bright sample, such as a positive control.

11. Click  **Snap Image** again to refresh the preview.

12. Click  **Image Intensity Settings**.



13. On the **Image Intensity Settings** tab, adjust the settings as needed for each channel.
    - Use the **Channel Color** dropdown to change the identification color for the channel. The default channel color is based on the emission wavelength of the selected stain.
    - Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.
    - Select **Synchronize Histograms** to adjust the image intensity scale for all channels simultaneously.

14. Use the image viewer controls as needed to control the preview:

 : Toggle the available channels by clicking on the channel icons. "Hidden" channels are shown slightly dimmed.

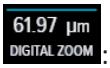
Tip: When the image is overexposed, the channel icons indicate the overexposure  . Lower the **Exposure** value as needed.

 : Zoom in on the image.

 : Zoom out on the image after zooming in.

 : Reset the view zoom to the original image size.

 : View the image full screen.



: View the current zoom scale. **Digital Zoom** indicates that the image is zoomed in digitally (that is, not optically) and might not represent the actual quality of the image.

15. Repeat these steps as needed until you are satisfied with the quality of the preview.

> **Tip:** The **History** pane contains thumbnails of previous preview images. Click a thumbnail to show that preview image and revert the settings to the configuration used to acquire it.

To continue to the next workflow step, click  **Region Selection to Acquire**. See Region Selection to Acquire on page 28 for details.

Region Selection to Acquire

 **Region Selection to Acquire** is the step where you select the region of the well to be acquired. The page shows a representation of a round microplate well with a region selection overlay. You must select at least one region to run an experiment.

You can change the region selection area by resizing and moving the overlay. If needed, you can set multiple region selection overlays.

> **Note:** Several factors (including the number of wells in your plate and the magnification of the objective) may prevent you from selecting some regions within certain wells.

The right side of the page includes the following icons:

 **From Center**: Adds a new acquisition region selection overlay in the center of the well or slide. You can control various elements of the acquisition region, including the percentage of the well or slide and the shape of the selection overlay.

 **Activate Edit Mode**: Activates the selection handles on the acquisition region selection overlay, enabling you to manually move and size it.

 **Add Acquisition Region**: Adds a new acquisition region selection overlay that you can size and move into position.

 **Delete Selected Region**: Removes the selected region overlay.

 **Clear All Regions**: Removes all the visible region overlays.

 **Toggle Actual Area to Capture**: Shows what the selected camera objective will snap based on the field of view for the lens. You may need to adjust the region selection or the objective selection based on this area.

To continue to the next workflow step, click  **Analysis Settings**. See Analysis Settings on page 29 for details.

Analysis Settings

 **Analysis Settings** is the step where you set up image analysis for the experiment. The data generated based on these settings becomes the analysis data for your experiment available in **Experiments** mode.

The right side of the page includes the following icons:

 **Choose Analysis**: Toggles analysis on or off and selects the analysis for the experiment.

 **Measurements**: Specifies the cell measurements included in the analysis.

 **Save Analysis**: Saves the analysis for use in future experiments.

 **Cell Info Mode**: Displays information on a selected cell.

 **Comparison Mode**: Captures two preview images, which enables you to compare the uniformity of the image quality.

Setting Up an Analysis

To set up an analysis:

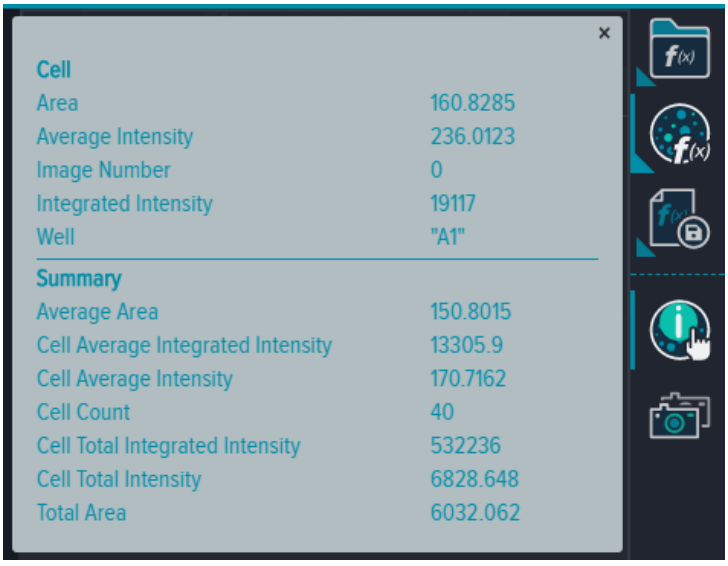
1. On the **Acquisition Analysis** page, in the **Tools** pane on the right, click  **Choose Analysis**.
2. Set **Analysis** to **On**.
3. Select the type of analysis.

   Note: The selected analysis appears at the top of the panel.

4. Click  **Measurements**.
5. In the **Measurements** pane, select the measurements for the analysis.

   Note: The recommended measurements for the analysis are set by default.

6. In the bottom pane, click  **Test Analysis** to calculate the summary measurements using the preview image.
7. If needed, click  **Cell Info Mode** to select a detected cell in the image preview and view cell information.



As part of configuring analysis settings, you may need to perform the following tasks:

- Testing the Analysis of a Well, see page 31
- Testing the Analysis of Comparison Images, see page 32
- Saving Analysis Settings, see page 34

To continue to the next workflow step, click  **Region Selection to Analyze**. See Region Selection to Analyze on page 35 for details.

Testing the Analysis of a Well

The preview represents the image quality to expect when you run your experiment.

To test the analysis of a well:

1. On the **Acquisition Analysis** page, in the bottom pane, click  **Test Analysis**.
2. Click  **Algorithm Input**.
3. On the **Algorithm Input** tab, adjust the settings as needed to optimize object detection. Settings vary depending on the selected analysis type. Setting options typically control intensity, minimum width, and maximum width for each stain used in the acquisition.
4. Click  **Test Analysis** to preview the analysis.

   Note: You should not need to adjust the image intensity settings, which use the Acquisition settings. If you do, click  **Image Intensity Scale** and adjust the settings as needed for each channel. Use the **Channel Color** dropdown to change the identification color for the channel. Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.

5. Click  **Choose Well and Area to Acquire**.
6. Click  **Select Well**.
7. In the plate map, select a different well.
   The CellReporterXpress Software runs a test analysis.

8. In the single well map, click and drag the  selection tool to select the region of the well for the preview, if needed. The center area of the well is selected by default.
   The CellReporterXpress Software runs a test analysis.
9. Click  **Algorithm Input**.
10. Click  **Test Analysis** to preview the analysis.
11. Repeat these steps as needed until you are satisfied with the quality of the preview.

After you are satisfied with the quality of the preview, you may want to save the analysis settings for later reuse. See Saving Analysis Settings on page 34 for details.

Testing the Analysis of Comparison Images

You can preview two images from different wells to compare the uniformity of the image quality.

To test the analysis of comparison images:

1. On the **Acquisition Analysis** page, in the **Tools** pane on the right, click  **Comparison Mode**.
2. In the bottom pane, click  **Test Analysis**.
3. Click  **Algorithm Input**.
4. On the **Algorithm Input** tab, adjust the settings as needed to optimize object detection. The settings affect both previews. Settings vary depending on the selected analysis type. Setting options typically control intensity, minimum width, and maximum width for each stain used in the acquisition.
5. On the left side of the pane, click the  **Choose Well and Area to Acquire** tab.
6. Click  **Select Well** on the left.

7. In the plate map, select the first well for the preview. The A1 well is selected by default.

8. Click 

Select Well on the right.

9. In the plate map, select the second well for the preview. The bottom right well is selected by default.

10. In the single well map on the left, click and drag the  selection tool to select the region of the first well for the preview, if needed. The center area of the well is selected by default.

11. Repeat the previous step in the single well map on the right to select the region of the second well for the preview, if needed.

12. Click  **Test Analysis** to preview the analysis.

Note: You should not need to adjust the image intensity settings, which use the Acquisition settings. If you do, click  **Image Intensity Scale** and adjust the settings as needed for each channel. Use the **Channel Color** dropdown to change the identification color for the channel. Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.

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13. Repeat these steps as needed until you are satisfied with the quality of the previews.

After you are satisfied with the quality of the preview, you may want to save the analysis settings for later reuse. See Saving Analysis Settings on page 34 for details.

Saving Analysis Settings

After you are satisfied with the quality of the preview in Testing the Analysis of a Well on page 31 or Testing the Analysis of Comparison Images on page 32, you may want to save the analysis settings for later reuse.

To save analysis settings:

1. On the **Acquisition Analysis** page, in the **Tools** pane on the right, click  **Save Analysis**.
2. In the **Save Analysis** pane, in the **Analysis Settings** field, enter a descriptive name.
3. (Optional) Add an avatar image by doing one of the following:
   - Click **Use Captured Picture**.
   - Click **Click to upload**, select an image file of your own, and click **Open**.
4. Click **Save**.

Region Selection to Analyze

Region Selection to Analyze is the step where you select the region of a well to be analyzed. You must select at least one region to run an experiment.

Note: Several factors (including the number of wells in your plate and the magnification of the objective) may prevent you from selecting some regions within certain wells.

The right side of the page includes the following icons:

From Center: Adds a new acquisition region selection overlay in the center of the well or slide. You can control various elements of the acquisition region, including the percentage of the well or slide and the shape of the selection overlay.

Activate Edit Mode: Activates the selection handles on the acquisition region selection overlay, enabling you to manually move and size it.

Add Analysis Region: Adds a new analysis region selection overlay that you can size and move into position.

Delete Selected Region: Removes the selected region overlay.

Clear All Regions: Removes all the visible region overlays.

To continue to the next workflow step, click  **Well Selection**. See Well Selection on page 36 for details.

Well Selection


Well Selection is the step where you select the wells to be acquired. The **Well Selection** page shows a plate map for the plate selected in the **Acquisition Settings** step. You must select at least one well to run an experiment.


Note: Several factors (including the number of wells in your plate and the magnification of the objective) may prevent you from selecting certain wells.

The right side of the page includes the following icons:


Select All: Selects all wells.


Clear All Regions: Removes all well selections.

By default, no well are selected.

Selecting a Group of Wells

In the plate map, click and drag to select a series of wells.

Selecting Individuals Wells

In the plate map, click a well to select it.

Deselecting Individuals Wells

In the plate map, click a selected well to deselect it.

To continue to the next workflow step, click 
Time Series. See Time Series on page 37 for details.

Time Series

Time Series is an optional step where you set up a time series for image acquisition. This enables you to acquire images at multiple time points.

Note: The CellReporterXpress Software does not support adjusting the temperature during a time series acquisition. If your acquisition requires this, you can perform a discontinuous time series by acquiring the first set of time points, adjusting the temperature as needed, and then acquiring the next set of time points. See Using Temperature Control on page 153 for details.

To set up a time series:

1. On the **Time Series** page, click the **On/Off** toggle switch as needed to activate the time series settings.
2. In the **Acquisition Order** section, click one of the following icons to indicate the order in which wells will be acquired for your time series:

All Wells: Performs a single time series on all selected wells. This option requires the least amount of time because only one time series is performed.

Per Column: Performs a complete time series on all selected wells in a column, then moves on to the next column. The leftmost column is acquired first. This option requires more time because the time series is repeated for each selected column.

Per Row: Performs a complete time series on all selected wells in a row, then moves on to the next row. The topmost row is acquired first. This option requires more time because the time series is repeated for each selected row.

Per Well: Performs a complete time series on each selected well, then moves on to the next well. This option requires the most time because the time series is repeated for each selected well.

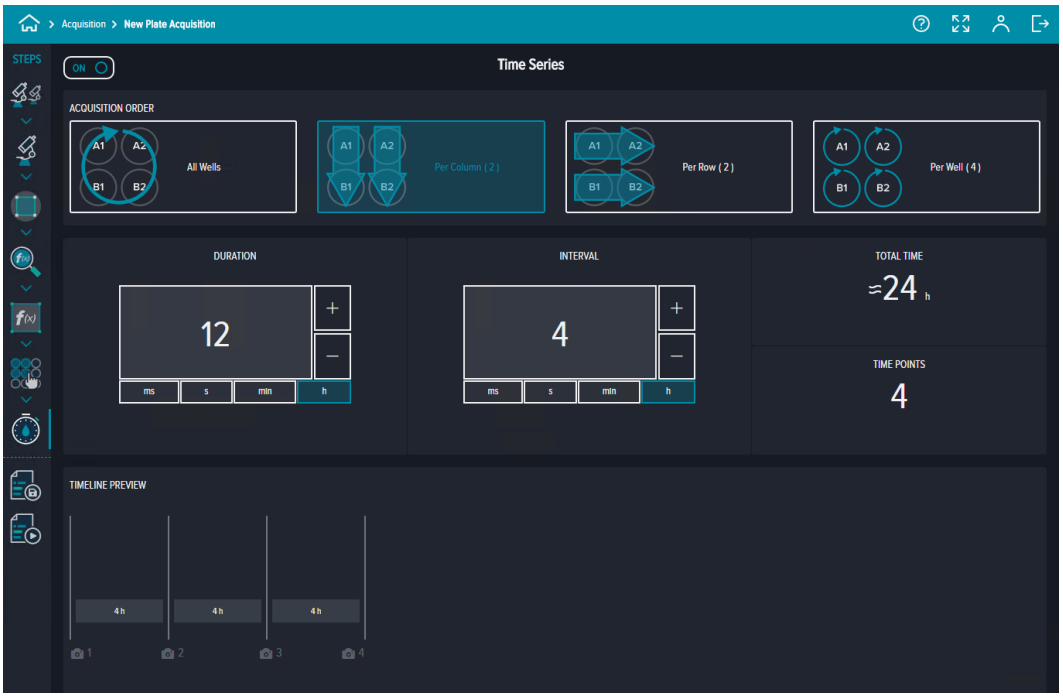
3. In the **Duration** section, do the following to specify the duration of your time series:
   a. Click in the value field and enter a value.
   b. Click the appropriate unit.
4. In the **Interval** section, do the following to specify the interval between the time points in your time series:
   a. Click in the value field and enter a value.
   b. Click the appropriate unit.

   The CellReporterXpress Software displays the recommended interval, which is based on several factors, including the following:

   - the number of wells and the number of regions per well
   - wavelength
   - exposure time
   - acquisition order

   For best results, specify at least the recommended interval. If you specify a lower interval, the system performs back-to-back acquisitions using the specified acquisition order. The actual interval will likely be greater than the specified interval.

The CellReporterXpress Software displays the total time required for the time series and the number of time points to be acquired for each selected well, along with a visual representation of the time series.



5. Review the time series settings.
6. If needed, repeat these steps to adjust the time series settings.

To continue to the next workflow step, click either  **Save Protocol** or  **Run Protocol**. See Save Protocol on page 40 or Run Protocol on page 41 for details.

Save Protocol


Save Protocol is an optional step where you can save the protocol you have created. You will typically save a protocol only when you intend to run it frequently. After you save a protocol, it appears as a card in the Protocol library.

Tip: Molecular Devices recommends creating protocols sparingly to avoid unnecessarily filling your Protocol library, which can make it difficult to find a protocol.

The right side of the page includes the following icon:


Lock Protocol: Manages the ability to modify the protocol. Note that users are not prevented from viewing or running a locked protocol.

To save a protocol:

1. On the **Save Protocol** screen, in the **Protocol Name** field, enter a name for the protocol.
2. (Optional) In the **Protocol Description** field, enter a description of the protocol.
3. Do the following to restrict other users from modifying the protocol:
   a. On the right side of the screen, click  **Lock Protocol**.
   b. Select the **Lock** checkbox to prevent other users from modifying the settings on the **Acquisition Device**, **Acquisition Settings**, **Analysis Settings**, and **Time Series** pages for the protocol.
   c. Select the **Lock region selection** checkbox to prevent other users from modifying the settings on the **Region Selection to Acquire** and **Region Selection to Analyze** pages.
   d. Select the **Lock well selection** checkbox to prevent other users from modifying the settings on the **Well Selection** page.
   e. To specify other users who can modify a locked protocol, click the **Add Editor** field and select users from the list.

   Tip: You can set default sharing permissions in  **Configuration Settings**. See Sharing Permissions on page 170 for details.
4. Click **Save Protocol**.

To continue to the next workflow step, click  **Run Protocol**. See Run Protocol on page 41 for details.

Run Protocol

 **Run Protocol** is the last step before you can run the experiment. In this step, you can review the acquisition settings and address any issues. When you run the experiment, the CellReporterXpress Software acquires images and analyzes the data according to your settings. When the experiment completes, the CellReporterXpress Software saves all acquired data as specified. Acquired data is then available for viewing and analysis in **Experiments** mode.

The right side of the page includes the following icons:

 **Experiment Details**: Displays acquisition and analysis settings and enables you to validate settings before running the experiment.

 **Storage**: Specifies image storage location during and after acquisition.

 **Public** and  **Private**: Manages the shared status of the experiment.

 **Open Plate Door**: Opens the sample door on the selected instrument so that you can insert or remove labware.

 **Close Plate Door**: Closes the sample door.

 **Run Experiment**: Runs the experiment using the specified acquisition and analysis settings. This icon becomes enabled when all the settings on the **Validation** tab in the **Experiment Details** pane are valid and an **Experiment Name** has been entered.

To run the experiment:

1. On the **Run Protocol** page, in the  **Experiment Details** pane, do the following:
   a. In the **Experiment Name** field, enter a name to identify the experiment in the Experiments library.
   b. (Optional) In the **Barcode** field, enter the barcode for the experiment labware.
   c. (Optional) In the **Experiment Description** field, enter a description of the experiment.
   d. In the **Validation** tab area, verify that all the required settings are valid. A  icon indicates a valid setting and a  icon indicates an invalid or missing setting. All acquisition settings must be valid to run the experiment. See Fixing Invalid Parameters on page 43 for details.
2. If you want to review the settings for image storage during and after acquisition, click  **Storage** and do the following:
   a. In the **Available Temporary Storage on Device** field, specify the computer for temporary image storage during acquisition. See the *ImageXpress Pico User Guide* for details on adding external temporary storage.
   b. In **Data Storage Settings** field, select a mapped folder for image storage after acquisition. See Data Storage on page 149 for details on registering external computers and mapping folders for image storage. Select the **Preserve Raw Images** checkbox to save TIFF images of the acquisition.
3. If you want to manage the shared status of the experiment to restrict other users from viewing it, click  **Public** and do the following:
   a. Select the **Private** checkbox.
   b. If you want to specify other users who can view a private experiment, click the **Share With** field and select users from the list.

 **Tip:** You can set default sharing permissions in  **Configuration Settings**. See Sharing Permissions on page 170 for details.

4. If you have not already done so, do the following to insert your experiment-ready labware into the instrument:
   a. Click  **Open Plate Door** to open the stage door on the instrument.
   b. Insert your experiment-ready labware into the instrument. See the *ImageXpress Pico User Guide* for details.
   c. Click  **Close Plate Door** to close the stage door on the instrument.
5. Click  **Run Experiment** to run the experiment.

The **Monitor** page opens to display the progress of the running experiment. See Monitor Mode on page 137 for details.

Fixing Invalid Parameters

Invalid parameter settings are indicated by a  icon. Click the icon to display the reason for the invalid parameter.

To fix an invalid parameter:

1. Click the link next to the  icon to open the workflow step for the invalid parameter.
2. Address the issue.
3. Click  **Run Protocol** to return to **Run Protocol** page.

Slide Acquisition Workflow

Select a slide template to begin the slide acquisition workflow. The **New Slide Acquisition** template is an unrestricted template that allows you to select from all slide experiment settings options. Other slide templates may offer fewer or restricted options.

The icons in the **Steps** pane on the left side of the page guide you through the slide experiment configuration process. The tools and controls in the pane on the right side of the page vary according to the step being configured and the experiment type.

Note: Depending on the selected template, some steps, tools, and options may not appear or may not be available. Use the **New Slide Acquisition** template to access all steps, tools, and options.

The slide acquisition workflow is as follows:


Acquisition Device is the first step for all acquisition workflows. In this step, you select the device for the acquisition and insert your experiment-ready labware into the instrument. See Acquisition Device on page 46 for details.


Acquisition Settings is the step where you set up image acquisition for the experiment, including the stains and objective to be used. You can preview the image capture and adjust channel identification color, histogram, focus, and exposure settings. See Acquisition Settings on page 47 for details.


Region Selection to Acquire is the step where you select the region of the slide to be acquired. The page shows a representation of a slide. You must select at least one region to run an experiment. See Region Selection to Acquire on page 56 for details.


Analysis Settings is the step where you set up image analysis for the experiment. The data generated based on these settings becomes the analysis data for your experiment available in **Experiments** mode. This step is not included in the slide workflow for a colorimetric or stitched acquisition. See Analysis Settings on page 57 for details.


Region Selection to Analyze is the step where you select the region of a slide to be analyzed. You must select at least one region to run an experiment. This step is not included in the slide workflow for a colorimetric or stitched acquisition. See Region Selection to Analyze on page 63 for details.

 **Save Protocol** is an optional step where you can save the protocol you have created. You will typically save a protocol only when you intend to run it frequently. After you save a protocol, it appears as a card in the Protocol library. See Save Protocol on page 64 for details.

 **Run Protocol** is the last step before you can run the experiment. In this step, you can review the acquisition settings and address any issues. When you run the experiment, the CellReporterXpress Software acquires images and analyzes the data according to your settings. When the experiment completes, the CellReporterXpress Software saves all acquired data as specified. Acquired data is then available for viewing and analysis in **Experiments** mode. See Run Protocol on page 65 for details.

Acquisition Device

 **Acquisition Device** is the first step for all acquisition workflows. In this step, you select the device for the acquisition and insert your experiment-ready labware into the instrument. See the *ImageXpress Pico User Guide* for details on inserting labware into the instrument.

The right side of the page includes the following icons:

 **Shutdown Device**: Prepares the software to power off the selected instrument.

 **Restart Device**: Restarts the selected instrument.

 **Open Plate Door**: Opens the stage door on the selected instrument so that you can insert or remove labware.

 **Close Plate Door**: Closes the stage door.

 **Set Up for Adjustment of Objective Collar**: Moves the objective turret so that you can adjust the correction collar on the selected objective.

 **Finish Adjustment of Objective Collar**: Moves the objective turret back into position after you adjust a correction collar.

The CellReporterXpress Software can control multiple instruments. If your software is configured to control multiple instruments, you may need to select the one you want to use.

 **Note:** If you click the  **Favorite** icon for a device, that device will be selected by default. If you have not specified a favorite device, the last device used will be selected by default.

To select an acquisition device:

On the **Acquisition Device** page, in the **Available Acquisition Devices** list, select the imaging device you want to use.

The selected device is highlighted in the Available Acquisition Devices list.

To continue to the next workflow step, click  **Acquisition Settings**. See Acquisition Settings on page 47.

Acquisition Settings


Acquisition Settings is the step where you set up image acquisition for the experiment, including the stains and objective to be used. You can preview the image capture and adjust channel identification color, histogram, focus, and exposure settings.

The right side of the page includes the following icons:


Slide Format: Specifies the slide format for the acquisition.


Stains: Specifies the stains for the acquisition.


Objectives: Specifies the objective for the acquisition.


Comparison Mode: Captures two preview images, which enables you to compare the uniformity of the image quality or compare settings on two different phenotypes (such as positive and negative controls). See Snapping Slide Comparison Previews on page 53.

To configure slide acquisition settings:

1. On the **Acquisition Settings** page, in the **Tools** pane on the right, click  **Slide Format**.
2. In the **Slide Format** pane, select the slide format you want to use.
3. If your slide has a frosted area at one end, in the **Slide Frost Area** section, enter details on the size and position of the frosted area.
4. Click  **Stains**.
5. In the **Stains** pane, select the stains you want to use.
6. Click  **Move Stain Up** and  **Move Stain Down** as needed to select the order in which the stains will be collected.

> **Tip:** For an acquisition with transmitted light, acquire the transmitted light stain first or last. If you plan to perform transmitted light segmentation, you will likely want to set the transmitted light stain to be collected first.

7. Click  **Objectives**.
8. In the **Objectives** pane, select the objective you want to use.

As part of configuring acquisition settings, you may need to perform the following tasks:

- Adjusting a Correction Collar, see page 48
- Snapping a Preview of a Slide, see page 50
- Snapping Slide Comparison Previews, see page 53

To continue to the next workflow step, click  **Region Selection to Acquire**. See Region Selection to Acquire on page 56 for details.

Adjusting a Correction Collar

The 40x objective and 63x objective have application-optimized correction collars (CORR) to compensate for external influences such as well bottom thickness or coverslip thickness. The collars have a range of 0 mm to 2 mm correction. Changing this setting adjusts the distances between components inside the objective barrel. Image quality and resolution are very dependent on properly setting these collars.

The settings to be used depend on the well bottom thickness of the plate or the coverslip thickness on the slide on which the specimen is mounted. In general, set the correction collar for the physical thickness of the plate or slide that you are imaging. The physical thickness can be determined by the plate specifications from the plate manufacturer.

Note: Do not use a plate, slide, or coverslip with a thickness that is out of the range of the correction collar for the selected objective.

Observe the following when handling an objective:

CAUTION! To prevent skin oils from damaging the optical coatings, Molecular Devices recommends that you wear powder-free disposable gloves when handling objectives and filter cubes.

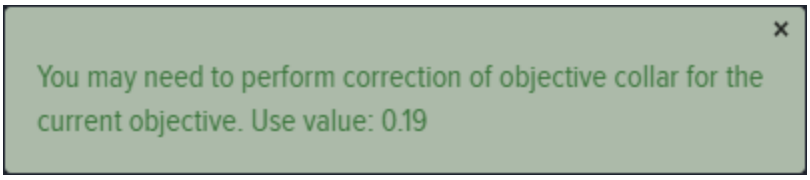
CAUTION! With the instrument power on, do not manually rotate the objective turret. Manually rotating the objective turret can damage the instrument.

You would typically adjust a correction collar as part of setting up an acquisition.

To adjust a correction collar for a slide:

1. On the **Acquisition Settings** page, on the right side of the screen under **Tools**, click  **Slide Format**.
2. In the **Slide Format** list, select the slide format.

3. On the right side of the screen under **Tools**, click  **Objectives**.
4. In the **Objectives** list, select the objective.
   If a correction collar adjustment is required, the CellReporterXpress Software displays the recommended setting for the correction collar based on the thickness of the plate bottom, slide, or coverslip.



5. On the left side of the screen under **Steps**, click  **Acquisition Device**.
6. On the right side of the screen, click  **Set Up for Adjustment of Objective Collar**.
7. Click **OK**.
   The objective door opens.
8. If needed, loosen or remove the objective from the instrument by gently turning it counterclockwise.
9. Rotate the correction collar to its new setting.

   Tip: You might need a flashlight to see the markings for the graduated scale on the barrel and its current setting.

10. If you loosened or removed the objective, insert it back in its original slot in the turret or tighten it by gently turning it clockwise.

    Note: When installing the objective, take care to avoid changing the correction collar setting.

11. Close the objective door.
12. Click **OK**.
13. Test the correction collar setting by examining the image quality of some test snaps.
14. If the image quality is not satisfactory, repeat these steps to re-adjust the correction collar.

Snapping a Preview of a Slide

You can create a preview of the acquisition by snapping an image. The preview uses the selected objective, wavelength, slide, region, focus settings, and exposure settings.

To snap an image of a well:

1. Click  **Snap Image** to snap an initial preview image.
2. On the **Acquisition Settings** page, at the bottom of the screen on the  **Choose Slide and Region to Acquire** tab, click  **Select Slide**.
3. In the slide holder map, select the slide for the preview. The instrument snaps a preview image of the newly selected slide. The **A1** slide is selected by default.

4. In the single slide map, click and drag the  selection tool to select the region of the slide for the preview, if needed. The instrument snaps a preview image of the newly selected region. The center area of the slide is selected by default.
5. Select **Include Overview** to include a low-resolution overview image of the entire slide in the single slide map. This can help you identify the best region for your experiment. The overview image is created using the planetary view camera with no objective.
6. Click  **Focus/Exposure Settings**.

7. On the **Focus/Exposure Settings** tab, adjust the settings as needed for each channel.
   - Use the **Focus offset** controls to adjust the image sharpness. Click the **Mode** dropdown to set one of the following autofocus ranges:
     - **Normal**: Hardware and image autofocus mode using a short search range.
     - **Wide**: Hardware and image autofocus mode using a wide range.
     - **Super Wide**: Hardware and image autofocus mode designed for samples that don't sit directly on the well bottom.
     - **Plate and Well Bottom**: Hardware-only autofocus mode designed for speed.

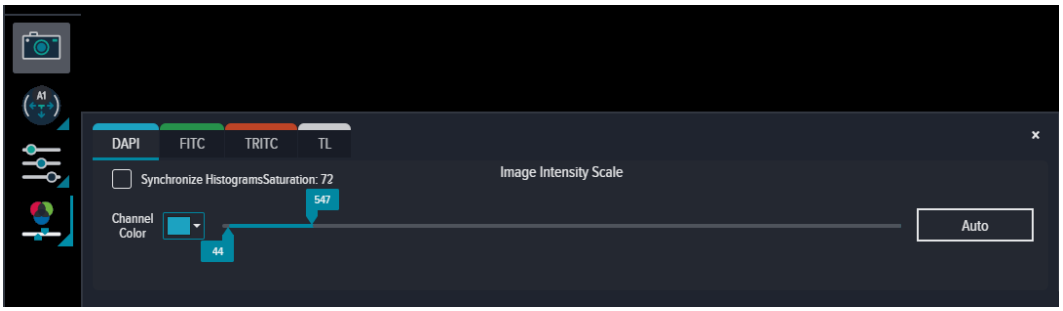
     After you set the focus mode, click **Auto** next to the **Focus Offset** controls to determine the focus offset between stains. This is particularly useful when using the **Plate and Well Bottom** focus mode.
   - Use the **Exposure** controls to adjust the exposure time, which affects image brightness and results in a broader dynamic range (that is, the difference between the brightest and dimmest object detected).

     When your image is in focus, click **Auto** to automatically determine the optimal exposure for the best dynamic range. The longer the exposure, the brighter the experiment image. Dimmer objects get brighter with longer exposure time, as does the background. Increased exposure time slows the experiment because each image takes longer and might lead to image saturation.

     Tip: You may want to use **Auto** with a known bright sample, such as a positive control.

8. Click  **Snap Image** again to refresh the preview.

9. Click  **Image Intensity Settings**.



10. On the **Image Intensity Settings** tab, adjust the settings as needed for each channel.
    - Use the **Channel Color** dropdown to change the identification color for the channel. The default channel color is based on the emission wavelength of the selected stain.
    - Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.
    - Select **Synchronize Histograms** to adjust the image intensity scale for all channels simultaneously.
11. Use the image viewer controls as needed to control the preview:

 : Toggle the available channels by clicking on the channel icons. "Hidden" channels are shown slightly dimmed.

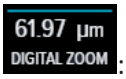
> **Tip:** When the image is overexposed, the channel icons indicate the overexposure  . Lower the **Exposure** value as needed.

 : Zoom in on the image.

 : Zoom out on the image after zooming in.

 : Reset the view zoom to the original image size.

 : View the image full screen.



: View the current zoom scale. **Digital Zoom** indicates that the image is zoomed in digitally (that is, not optically) and might not represent the actual quality of the image.

12. Repeat these steps as needed until you are satisfied with the quality of the preview.

> **Tip:** The **History** pane contains thumbnails of previous preview images. Click a thumbnail to show that preview image and revert the settings to the configuration used to acquire it.

To continue to the next workflow step, click  **Region Selection to Acquire**. See Region Selection to Acquire on page 56 for details.

Snapping Slide Comparison Previews

You can create previews of the acquisition by snapping comparison images from two slides (or two regions of the same slide) or compare settings on two different phenotypes (such as positive and negative controls). This enables you to compare the uniformity of the image quality. As with snapping an image of a single slide, the comparison images use the selected objective, wavelengths, well, region, focus settings, exposure settings, and histogram.

To snap slide comparison images:

1. Click  **Snap Image** to snap an initial preview image.
2. On the **Acquisition Settings** page, in the **Tools** pane on the right, click  **Comparison Mode**.
3. At the bottom of the screen on the  **Choose Slide and Region to Acquire** tab, click  **Select Slide** on the left.
4. In the slide holder map, select the first slide for the preview. The instrument snaps a preview image of the newly selected slide. The **A1** slide is selected by default.

5. Click  **Select Slide** on the right.
6. In the slide holder map, select the second slide for the preview. The instrument snaps a preview image of the newly selected slide.
7. In the single slide map on the left, click and drag the  selection tool to select the region of the first slide for the preview, if needed. The instrument snaps a preview image of the newly selected region. The center area of the slide is selected by default.
8. Select **Include Overview** to include a low-resolution overview image of the entire slide in the single slide map. This can help you identify the best region for your experiment. The overview image is created using the planetary view camera with no objective.

9. Repeat steps 7 and 8 in the single slide map on the right to select the region of the second slide for the preview, if needed.
10. Click  **Focus/Exposure Settings**.

11. On the **Focus/Exposure Settings** tab, adjust the settings as needed for each channel.
    - Use the **Focus offset** controls to adjust the image sharpness. Click the **Mode** dropdown to set one of the following autofocus ranges:
      - **Normal**: Hardware and image autofocus mode using a short search range.
      - **Wide**: Hardware and image autofocus mode using a wide range.
      - **Super Wide**: Hardware and image autofocus mode designed for samples that don't sit directly on the well bottom.
      - **Plate and Well Bottom**: Hardware-only autofocus mode designed for speed.

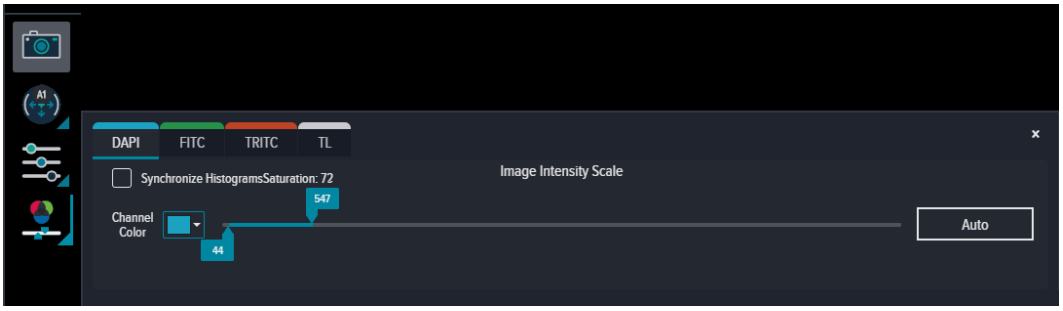
      After you set the focus mode, click **Auto** next to the **Focus Offset** controls to determine the focus offset between stains. This is particularly useful when using the **Plate and Well Bottom** focus mode.
    - Use the **Exposure** controls to adjust the exposure time, which affects image brightness and results in a broader dynamic range (that is, the difference between the brightest and dimmest object detected).

      When your image is in focus, click **Auto** to automatically determine the optimal exposure for the best dynamic range. The longer the exposure, the brighter the experiment image. Dimmer objects get brighter with longer exposure time, as does the background. Increased exposure time slows the experiment because each image takes longer and might lead to image saturation.

      Tip: You may want to use **Auto** with a known bright sample, such as a positive control.

12. Click  **Snap Image** again to refresh the preview.

13. Click  **Image Intensity Settings**.



14. On the **Image Intensity Settings** tab, adjust the settings as needed for each channel.
    - Use the **Channel Color** dropdown to change the identification color for the channel. The default channel color is based on the emission wavelength of the selected stain.
    - Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.
    - Select **Synchronize Histograms** to adjust the image intensity scale for all channels simultaneously.
15. Use the image viewer controls as needed to control the preview:

 : Toggle the available channels by clicking on the channel icons. "Hidden" channels are shown slightly dimmed.

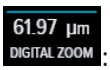
Tip: When the image is overexposed, the channel icons indicate the overexposure  . Lower the **Exposure** value as needed.

 : Zoom in on the image.

 : Zoom out on the image after zooming in.

 : Reset the view zoom to the original image size.

 : View the image full screen.



: View the current zoom scale. **Digital Zoom** indicates that the image is zoomed in digitally (that is, not optically) and might not represent the actual quality of the image.

16. Repeat these steps as needed until you are satisfied with the quality of the preview.

> **Tip:** The **History** pane contains thumbnails of previous preview images. Click a thumbnail to show that preview image and revert the settings to the configuration used to acquire it.

To continue to the next workflow step, click **Region Selection to Acquire**. See Region Selection to Acquire on page 56 for details.

Region Selection to Acquire

Region Selection to Acquire is the step where you select the region of the slide to be acquired. The page shows a representation of a slide. You must select at least one region to run an experiment.

By default, there are no slide region selections. You can add, resize, move, and delete region selection overlays. If needed, you can set multiple region selection overlays.

> **Note:** Several factors (including the configuration of your slide holder and the magnification of the objective) may prevent you from selecting some regions near the edges of the slide.

The right side of the page includes the following icons:

Add Acquisition Region: Adds a new acquisition region selection overlay that you can size and move into position.

Delete Selected Region: Removes the selected region overlay.

Clear All Regions: Removes all the visible region overlays.

Toggle Actual Area to Capture: Shows what the selected camera objective will snap based on the field of view for the lens. You may need to adjust the region selection or the objective selection based on this area.

To continue to the next workflow step for a colorimetric or stitched acquisition, click either **Save Protocol** or **Run Protocol**. See Save Protocol on page 64 or Run Protocol on page 65 for details.

To continue to the next workflow step for all other acquisitions, click  **Analysis Settings**. See Analysis Settings on page 57 for details.

Analysis Settings

 **Analysis Settings** is the step where you set up image analysis for the experiment. The data generated based on these settings becomes the analysis data for your experiment available in **Experiments** mode.

The right side of the page includes the following icons:

 **Choose Analysis**: Toggles analysis on or off and selects the analysis for the experiment.

 **Measurements**: Specifies the cell measurements included in the analysis.

 **Save Analysis**: Saves the analysis for use in future experiments.

 **Cell Info Mode**: Displays information on a selected cell.

 **Comparison Mode**: Captures two preview images, which enables you to compare the uniformity of the image quality.

Setting Up an Analysis

To set up an analysis:

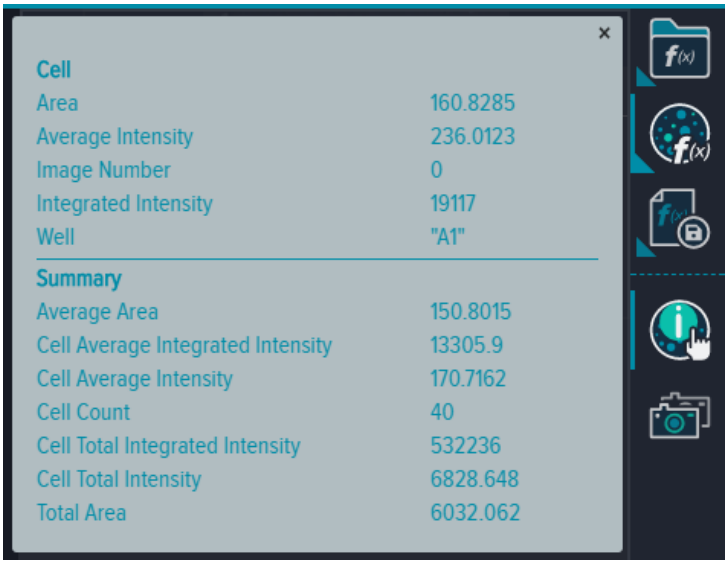
1. On the **Acquisition Analysis** page, in the **Tools** pane on the right, click  **Choose Analysis**.
2. Set **Analysis** to **On**.
3. Select the type of analysis.

   Note: The selected analysis appears at the top of the panel.

4. Click  **Measurements**.
5. In the **Measurements** pane, select the measurements for the analysis.

   Note: The recommended measurements for the analysis are set by default.

6. In the bottom pane, click  **Test Analysis** to calculate the summary measurements using the preview image.
7. If needed, click  **Cell Info Mode** to select a detected cell in the image preview and view cell information.



As part of configuring analysis settings, you may need to perform the following tasks:

- Testing the Analysis of a Region, see page 59
- Testing the Analysis of Comparison Images, see page 60
- Saving Analysis Settings, see page 62

To continue to the next workflow step, click  **Region Selection to Analyze**. See Region Selection to Analyze on page 63 for details.

Testing the Analysis of a Region

The preview represents the image quality to expect when you run your experiment.

To test the analysis of a region:

1. On the **Acquisition Analysis** page, in the bottom pane, click  **Test Analysis**.
2. Click  **Algorithm Input**.
3. On the **Algorithm Input** tab, adjust the settings as needed to optimize object detection. Settings vary depending on the selected analysis type. Setting options typically control intensity, minimum width, and maximum width for each stain used in the acquisition.
4. Click  **Test Analysis** to preview the analysis.

Note: You should not need to adjust the image intensity settings, which use the Acquisition settings. If you do, click  **Image Intensity Scale** and adjust the settings as needed for each channel. Use the **Channel Color** dropdown to change the identification color for the channel. Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.

5. Click  **Choose Position to Acquire**.
6. Click  **Select Slide**.
7. In the slide map, select a different region.
   The CellReporterXpress Software runs a test analysis.

8. In the single slide map, click and drag the  selection tool to select the region of the slide for the preview, if needed. The center area of the slide is selected by default.
   The CellReporterXpress Software runs a test analysis.
9. Click  **Algorithm Input**.
10. Click  **Test Analysis** to preview the analysis.
11. Repeat these steps as needed until you are satisfied with the quality of the preview.

After you are satisfied with the quality of the preview, you may want to save the analysis settings for later reuse. See Saving Analysis Settings on page 62 for details.

Testing the Analysis of Comparison Images

You can preview two images from different regions to compare the uniformity of the image quality.

To test the analysis of comparison images:

1. On the **Acquisition Analysis** page, in the **Tools** pane on the right, click  **Comparison Mode**.
2. In the bottom pane, click  **Test Analysis**.
3. Click  **Algorithm Input**.
4. On the **Algorithm Input** tab, adjust the settings as needed to optimize object detection. The settings affect both previews. Settings vary depending on the selected analysis type. Setting options typically control intensity, minimum width, and maximum width for each stain used in the acquisition.

5. On the left side of the pane, click the 

Choose Position to Acquire tab.
6. Click 

Select Slide on the left.
7. In the slide map, select the first slide for the preview. The A1 slide is selected by default.
8. Click 

Select Slide on the right.
9. In the slide map, select the second slide for the preview.
10. In the single slide map on the left, click and drag the  selection tool to select the region of the first slide for the preview, if needed. The center area of the slide is selected by default.
11. Repeat the previous step in the single slide map on the right to select the region of the second slide for the preview, if needed.
12. Click  **Test Analysis** to preview the analysis.

Note: You should not need to adjust the image intensity settings, which use the Acquisition settings. If you do, click  **Image Intensity Scale** and adjust the settings as needed for each channel. Use the **Channel Color** dropdown to change the identification color for the channel. Use the **Image Intensity Scale** controls to adjust the contrast between the dark and bright pixels in the image. Click **Auto** to automatically set the best contrast based on the current preview.

13. Repeat these steps as needed until you are satisfied with the quality of the previews.

After you are satisfied with the quality of the preview, you may want to save the analysis settings for later reuse. See Saving Analysis Settings on page 62 for details.

Saving Analysis Settings

After you are satisfied with the quality of the preview in Testing the Analysis of a Region on page 59 or Testing the Analysis of Comparison Images on page 60, you may want to save the analysis settings for later reuse.

To save analysis settings:

1. On the **Acquisition Analysis** page, in the **Tools** pane on the right, click  **Save Analysis**.
2. In the **Save Analysis** pane, in the **Analysis Settings** field, enter a descriptive name.
3. (Optional) Add an avatar image by doing one of the following:
   - Click **Use Captured Picture**.
   - Click **Click to upload**, select an image file of your own, and click **Open**.
4. Click **Save**.

Region Selection to Analyze

 **Region Selection to Analyze** is the step where you select the region of a slide to be analyzed. You must select at least one region to run an experiment.

The right side of the page includes the following icons:

 **From Center**: Adds a new acquisition region selection overlay in the center of the well or slide. You can control various elements of the acquisition region, including the percentage of the well or slide and the shape of the selection overlay.

 **Activate Edit Mode**: Activates the selection handles on the acquisition region selection overlay, enabling you to manually move and size it.

 **Add Analysis Region**: Adds a new analysis region selection overlay that you can size and move into position.

 **Delete Selected Region**: Removes the selected region overlay.

 **Clear All Regions**: Removes all the visible region overlays.

To continue to the next workflow step, click either  **Save Protocol** or  **Run Protocol**. See Save Protocol on page 64 or Run Protocol on page 65 for details.

Save Protocol

 **Save Protocol** is an optional step where you can save the protocol you have created. You will typically save a protocol only when you intend to run it frequently. After you save a protocol, it appears as a card in the Protocol library.

Tip: Molecular Devices recommends creating protocols sparingly to avoid unnecessarily filling your Protocol library, which can make it difficult to find a protocol.

The right side of the page includes the following icon:

 **Lock Protocol**: Manages the ability to modify the protocol. Note that users are not prevented from viewing or running a locked protocol.

To save a protocol:

1. On the **Save Protocol** screen, in the **Protocol Name** field, enter a name for the protocol.
2. (Optional) In the **Protocol Description** field, enter a description of the protocol.
3. Do the following to restrict other users from modifying the protocol:
   a. On the right side of the screen, click  **Lock Protocol**.
   b. Select the **Lock** checkbox to prevent other users from modifying the settings on the **Acquisition Device**, **Acquisition Settings**, **Analysis Settings**, and **Time Series** pages for the protocol.
   c. Select the **Lock region selection** checkbox to prevent other users from modifying the settings on the **Region Selection to Acquire** and **Region Selection to Analyze** pages.
   d. Select the **Lock well selection** checkbox to prevent other users from modifying the settings on the **Well Selection** page.
   e. To specify other users who can modify a locked protocol, click the **Add Editor** field and select users from the list.

   Tip: You can set default sharing permissions in  **Configuration Settings**. See Sharing Permissions on page 170 for details.
4. Click **Save Protocol**.

To continue to the next workflow step, click  **Run Protocol**. See Run Protocol on page 65 for details.

Run Protocol

 **Run Protocol** is the last step before you can run the experiment. In this step, you can review the acquisition settings and address any issues. When you run the experiment, the CellReporterXpress Software acquires images and analyzes the data according to your settings. When the experiment completes, the CellReporterXpress Software saves all acquired data as specified. Acquired data is then available for viewing and analysis in **Experiments** mode.

The right side of the page includes the following icons:

 **Experiment Details**: Displays acquisition and analysis settings and enables you to validate settings before running the experiment.

 **Storage**: Specifies image storage location during and after acquisition.

 **Public** and  **Private**: Manages the shared status of the experiment.

 **Open Plate Door**: Opens the sample door on the selected instrument so that you can insert or remove labware.

 **Close Plate Door**: Closes the sample door.

 **Run Experiment**: Runs the experiment using the specified acquisition and analysis settings. This icon becomes enabled when all the settings on the **Validation** tab in the **Experiment Details** pane are valid and an **Experiment Name** has been entered.

To run the experiment:

1. On the **Run Protocol** page, in the  **Experiment Details** pane, do the following:
   a. In the **Experiment Name** field, enter a name to identify the experiment in the Experiments library.
   b. (Optional) In the **Barcode** field, enter the barcode for the experiment labware.
   c. (Optional) In the **Experiment Description** field, enter a description of the experiment.
   d. In the **Validation** tab area, verify that all of the required settings are valid. A  icon indicates a valid setting and a  icon indicates an invalid or missing setting. All acquisition settings must be valid to run the experiment. See Fixing Invalid Parameters on page 67 for details.
2. If you want to review the settings for image storage during and after acquisition, click  **Storage** and do the following:
   a. In the **Available Temporary Storage on Device** field, specify the computer for temporary image storage during acquisition. See the *ImageXpress Pico User Guide* for details on adding external temporary storage.
   b. In **Data Storage Settings** field, select a mapped folder for image storage after acquisition. See Data Storage on page 149 for details on registering external computers and mapping folders for image storage. Select the **Preserve Raw Images** checkbox to save TIFF images of the acquisition.
3. If you want to manage the shared status of the experiment to restrict other users from viewing it, click  **Public** and do the following:
   a. Select the **Private** checkbox.
   b. If you want to specify other users who can view a private experiment, click the **Share With** field and select users from the list.

 **Tip:** You can set default sharing permissions in  **Configuration Settings**. See Sharing Permissions on page 170 for details.

4. If you have not already done so, do the following to insert your experiment-ready labware into the instrument:
   a. Click  **Open Plate Door** to open the stage door on the instrument.
   b. Insert your experiment-ready labware into the instrument. See the *ImageXpress Pico User Guide* for details.
   c. Click  **Close Plate Door** to close the stage door on the instrument.
5. Click  **Run Experiment** to run the experiment.

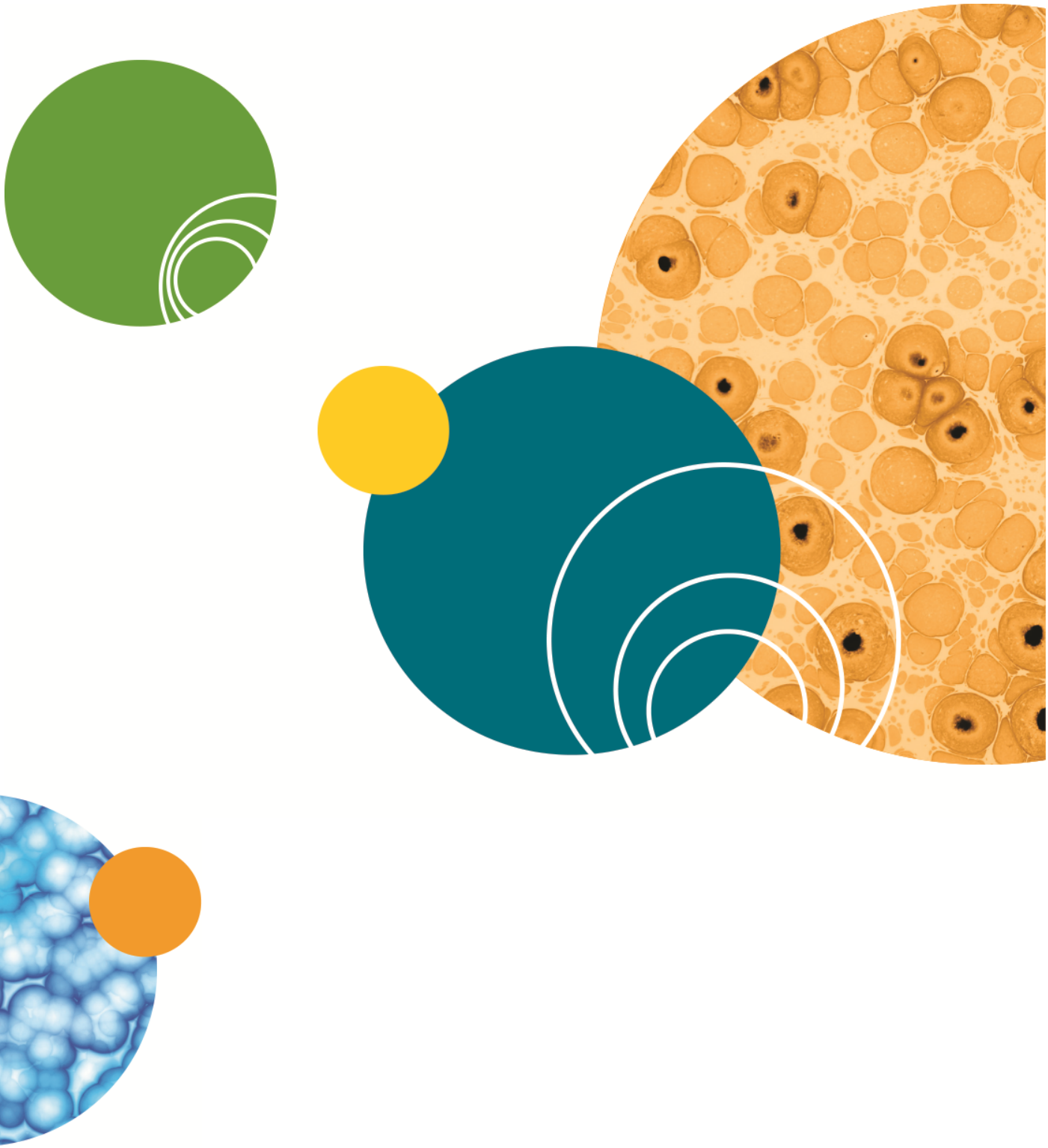
The **Monitor** page opens to display the progress of the running experiment. See Monitor Mode on page 137 for details.

Fixing Invalid Parameters

Invalid parameter settings are indicated by a  icon. Click the icon to display the reason for the invalid parameter.

To fix an invalid parameter:

1. Click the link next to the  icon to open the workflow step for the invalid parameter.
2. Address the issue.
3. Click  **Run Protocol** to return to **Run Protocol** page.



Chapter 4: Experiments Mode

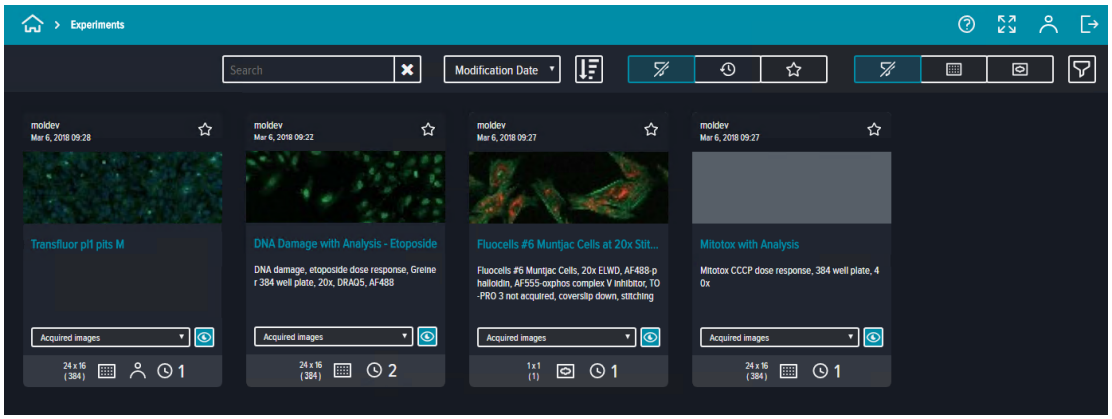
4

Use **Experiments** mode to view images and analysis data collected in **Acquisition** mode and perform additional offline analysis.

On the **Home** page, click the  **Experiments** tile to enter **Experiments** mode. The **Experiments** library opens.

Experiments Library

When you run a protocol in **Acquisition** mode, a card is created in the **Experiments** library. The card links all the processed images and any analysis data associated with the experiment run.



The experiment cards contain the experiment name and description, along with the name of the user who ran the experiment, the date and time of the experiment run, a  **Favorite** icon (that you can use to flag certain experiments), and a  **View** icon. In addition, each card contains icons to indicate experiment properties, including:

 **Geometry**

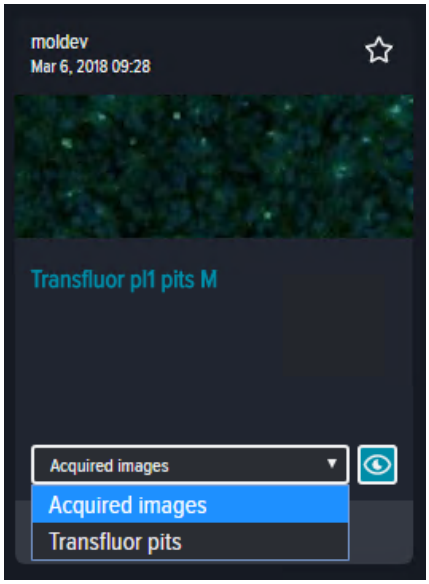
 **Plate** or  **Slide**

 **Shared**

 **Time Points**

Each experiment card includes the following functionality:

- Click a card to display the **Experiments** page, which shows the details for that experiment. From here, you can review analyses and acquisitions details.
- Click **View** (with the default value of **Acquired Images** selected) to display the **Thumbnail View** page for that experiment.
- Click the dropdown on the card to select acquired images or analysis data. The options vary depending on what was acquired during the experiment.



Search and Filters

To limit the number of visible cards on a page, use the **Search** field, the sort field, and the filter controls at the top of the Experiments library.

Search

Use search to find specific words in the titles and descriptions of experiments.

To use search:

1. Click in the **Search** field.
2. Enter the word you want to find then press **ENTER**.

Sort

Use sort to arrange the experiment tiles. You can sort in ascending or descending order based on the experiment name, creation date, or modification date.

To use sort:

1. Click the sort dropdown and select one of the following sort types:
   - **Creation Date**
   - **Modification Date**
   - **Name**
2. Click the sort order icon to specify one of the following sort orders:
   -  Sorts from A to Z or from earliest to latest.
   -  Sorts from Z to A or from latest to earliest.

Basic Filters

Use the filter icons to control which experiment cards are shown. Active filter icons are highlighted. By default,  **All** is the active filter. Filter options include:

-  **Recent** or  **Favorites**
-  **Plates** or  **Slides**

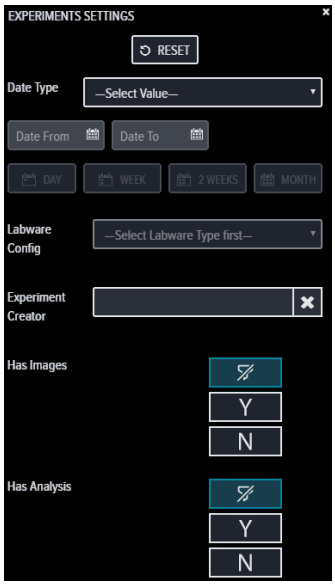
To use filter icons:

Select the filter icon you want to use. The icon is highlighted and only the cards matching the filter option are shown.

Complex Filters

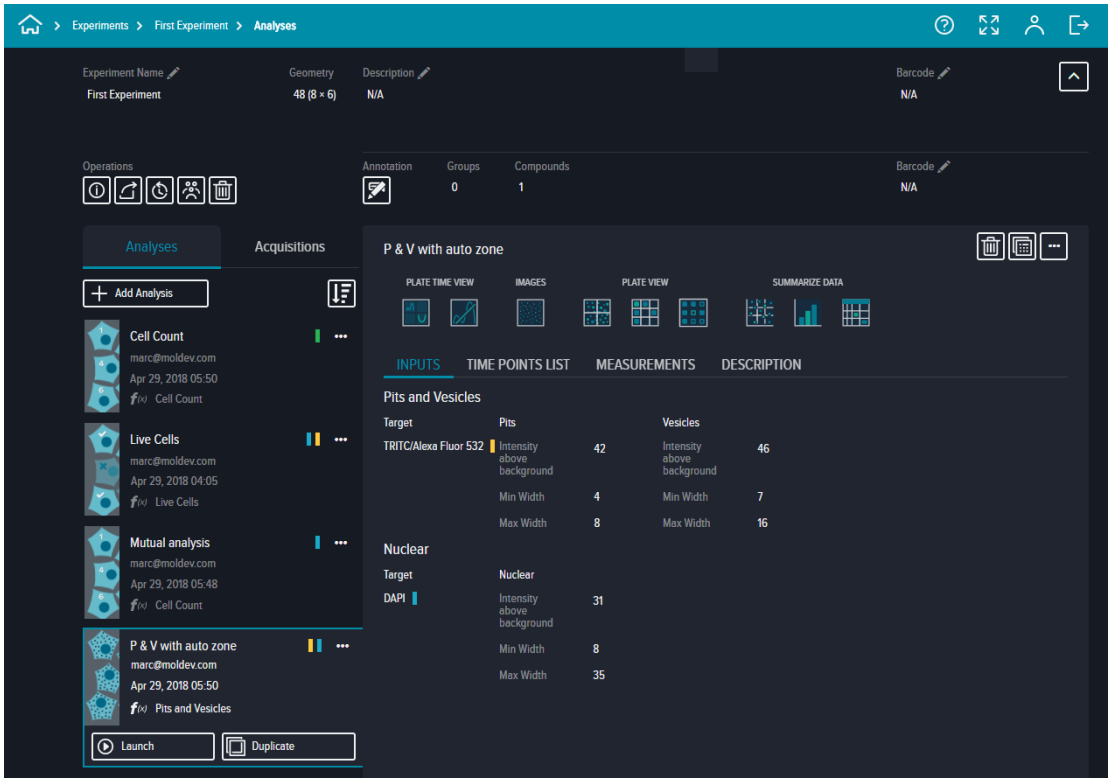
Click the **Filter** icon to create more complex filters. For example, you can filter for experiments created within a specific date range or filter for experiments created by one of three specified users.

Note: In order to use the **Labware Config** field, you must first select either **Plates** or **Slides** from the basic filter icons at the top of the page.



Experiments Page

The **Experiments** page shows the data for an experiment. From here, you can review general experiment details and access analyses and acquisitions details.

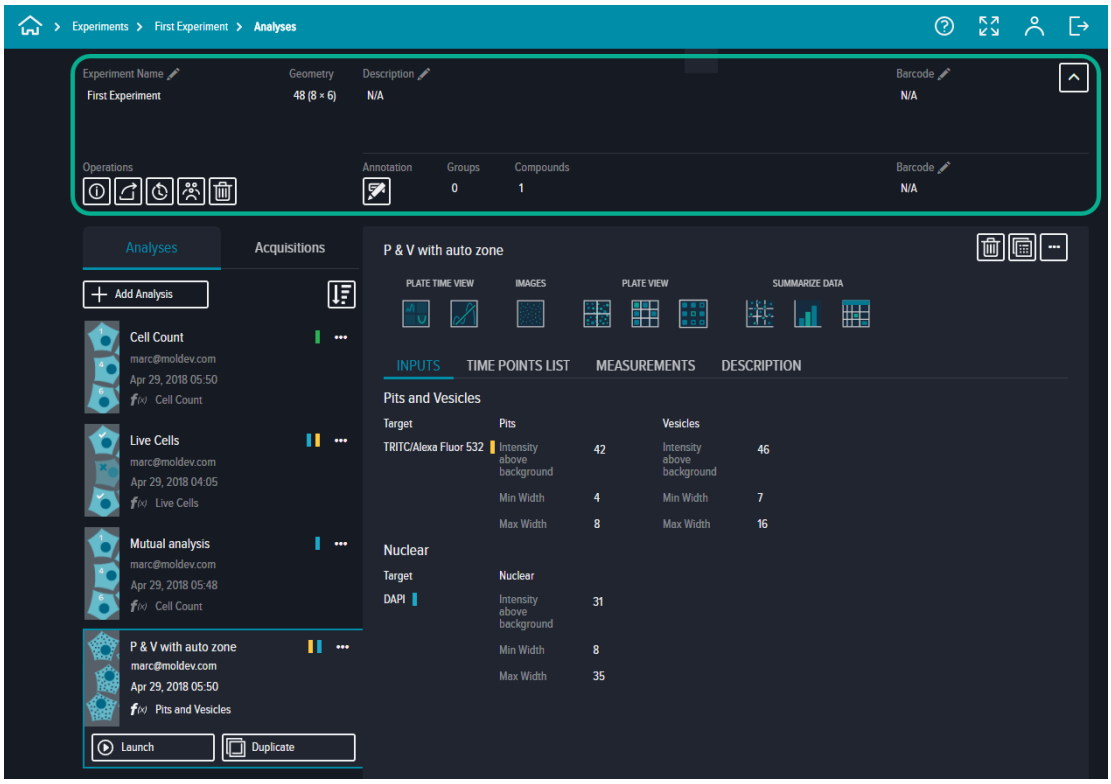


The screen is divided into three sections:

- The top section shows experiment details.
- The bottom left section contains tabs that show analyses and acquisitions for the experiment.
- The bottom right section contains details for the selected analysis or acquisition.

Experiment Details

The top section of the **Experiments** page shows details for the experiment.



The experiment details pane includes the following:

- **Experiment Name**: Indicates the name of the experiment. This field is editable.
- **Geometry**: Indicates the dimensions of the labware used for the experiment.
- **Description**: Indicates the description of the experiment. This field is editable.
- **Barcode**: Indicates the barcode of the plate for the experiment. This field is editable.
- **Operations**: Provides tools to manage the experiment, including exporting experiment images, importing times points, and changing sharing permissions. See Experiment Operations on page 75 for details.
- **Annotation**: Click **Edit Annotations** to open the **Annotations** page to upload or edit annotations. See Annotations on page 76 for details.
- **Groups**: Indicates the number of annotation groups currently in use.
- **Compounds**: Indicates the number of annotation compounds currently in use.
- **Barcode**: Indicates the barcode for the experiment annotations. This field is editable.

Note: The **Edit** icon indicates that a value can be edited. Click the value to edit it.

Experiment Operations

The **Experiments** page includes the following tools to help you manage the experiment:

 **Experiment Properties**: Displays properties including storage information and creation and modification details. You can upload an image avatar for the experiment.

 **Export Experiment Images**: Enables you to export images from the experiment as TIFF files. This function is available with a Windows computer only. The MD Import/Export Service is required for this function. See Import Mode on page 163 for details.

 **Import Time Point**: Enables you to import time point data from another acquisition. This enables you to recover data lost during an acquisition error. This function is available with a Windows computer only. The MD Import/Export Service is required for this function. See Import Mode on page 163 for details.

 **Public** or  **Private**: Indicates the shared status of the experiment. You can manage this status to restrict other users from viewing the experiment. See Restricting Others from Viewing an Experiment on page 75 for details.

 **Delete Experiment**: Deletes an experiment and all experiment data, including acquisition and analysis details.

Restricting Others from Viewing an Experiment

You can manage the shared status of an experiment to restrict other users from viewing it.

The shared status icon under **Operations** indicates the current shared status, either  **Public** or  **Private**.

 **Tip:** You can set default sharing permissions in  **Configuration Settings**. See Sharing Permissions on page 170 for details.

To restrict other users from viewing the experiment:

1. On the **Experiments** page, under **Operations**, click  **Public**.
2. Select the **Private** checkbox.
3. If you want to specify other users who can view a private experiment, click the **Share With** field and select users from the list.

Annotations

Click **Annotation** on the **Experiments** page to open the **Annotations** page. The **Annotations** page enables you to attach annotations to the experiment. You can manually enter annotations or import them from a CSV file. The annotation data is available as Measurements that can be used in heatmaps, bar graphs, scatter plots, and tables.

Tip: Positive Group and Negative Group values are recognized by the software and can be used later in relevant calculations, like Z Prime.

The right side of the page includes the following icons:

Assign Values: Assigns annotation values to selected wells or slides.

Edit Annotation Names: Adds annotation field names that you can assign to the default field names. Default field names are **group**, **compound**, and **concentration**.

Configure Display Annotations: Specifies which annotation data to show and the heatmap measurements to use.

Import/Export Annotations: Imports annotations from a CSV file and exports annotations to a CSV file. See Importing Annotations on page 77 for details.


Map Annotations: Assigns annotation field names to the default field names.

Selection Mode: Activates selection mode, which enables you to select wells or regions. Click and drag to select multiple wells. Click individual wells or regions to select and deselect them. The number on the icon indicates the number of selected wells or regions.


Deselect All: Deselect all selected wells.

Importing Annotations

You can import annotations from a CSV file. For best results, use one of the sample templates available in the *CellReporterXpress Software Help* and customize it to your needs.

To import annotations:

1. Click 

Upload Annotations.
2. In the **Import/Export Annotations** pane, click **Choose File**.
3. Browse to select the CSV file with your annotations.
4. In the **Import Mode** field, select one of the following options:
   - **Replace**: Overwrites current annotations.
   - **Keep Existing**: Adds to existing annotations.
5. In the **File Format** field, select either **Plate** or **Column**.
6. Click **Upload**.

Assigning Annotation Values

To assign values to selected wells or slides:

1. On the **Annotations** page, click  **Assign Values**.
2. Select the wells or slides you want to annotate.

   Click and drag to select a series of wells or slides. Click individual wells or slides to select and deselect them.
3. If needed, do one of the following:
   - Click  **Clear All Values** to clear all values in selected wells or slides.
   - Click  **Clear Values for Selected Wells** for a field to clear values for that field in selected wells or slides.
4. Enter annotation values as needed.
5. For a numeric value field, click  **Assign Series** if you want to assign a series of values. See Assigning a Series of Annotation Values to a Numeric Field on page 78 for details.
6. Do one of the following:
   - Click  **Apply All Values** to apply all values to selected wells or slides.
   - Click  **Apply Value to Selection** for a field to apply values for that field in selected wells or slides.

Assigning a Series of Annotation Values to a Numeric Field

For numeric value fields, you can assign a series of annotation values.

To assign a series:

1. On the **Annotations** page, click  **Assign Values**.
2. For a numeric value field, click  **Assign Series**.
3. In the **x-direction** field, enter the number of well or slides to repeat values in a horizontal direction.
4. In the **y-direction** field, enter the number of well or slides to repeat values in a vertical direction.

5. In the **Start From** section, select one of the following icons to indicate the order of the series:

 : Assigns the series from top to bottom, then left to right.

 : Assigns the series from top to bottom, then right to left.

 : Assigns the series from right to left, then bottom to top.

 : Assigns the series from right to left, then top to bottom.

 : Assigns the series from bottom to top, then right to left.

 : Assigns the series from bottom to top, then left to right.

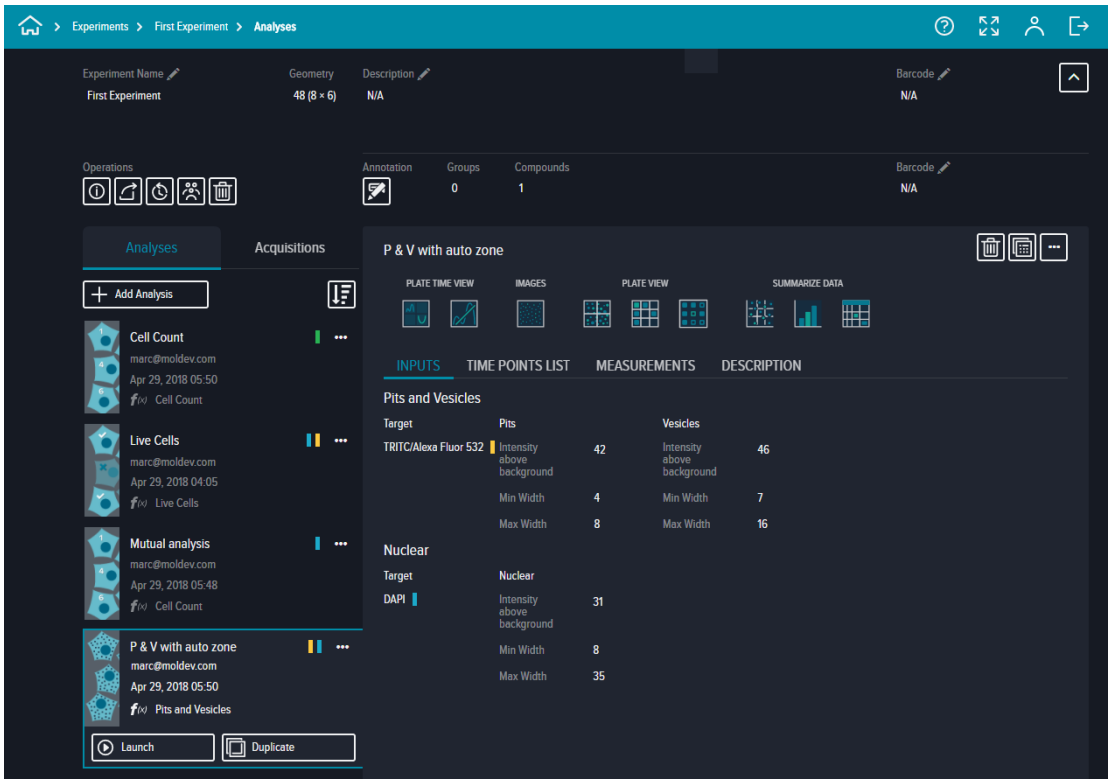
 : Assigns the series from left to right, then bottom to top.

 : Assigns the series from left to right, the top to bottom.

6. In the **Starting Value** field, enter the starting value for the series.
7. Click the **Step By** dropdown, and select the operator for the series.
8. In the **Step By** field, enter the step value for the series.
9. Click **OK**.

Experiment Analysis Details

The bottom section of the **Experiments** page shows analysis and acquisition details for the experiment. The left pane in the bottom section contains two tabs: **Analyses** and **Acquisitions**. Click the **Analyses** tab to show analysis details for the experiment.



On the **Analyses** tab, each analysis for the experiment is listed. The following functions are available:

- **Launch**: Opens the analysis for the experiment and enables you to perform additional analysis to different time points (using the same protocol).
- **Duplicate**: Opens the analysis for the experiment and enables you to modify the protocol and perform additional analysis to different time points.

In addition, you can click  to save the analysis as a template or delete the analysis.

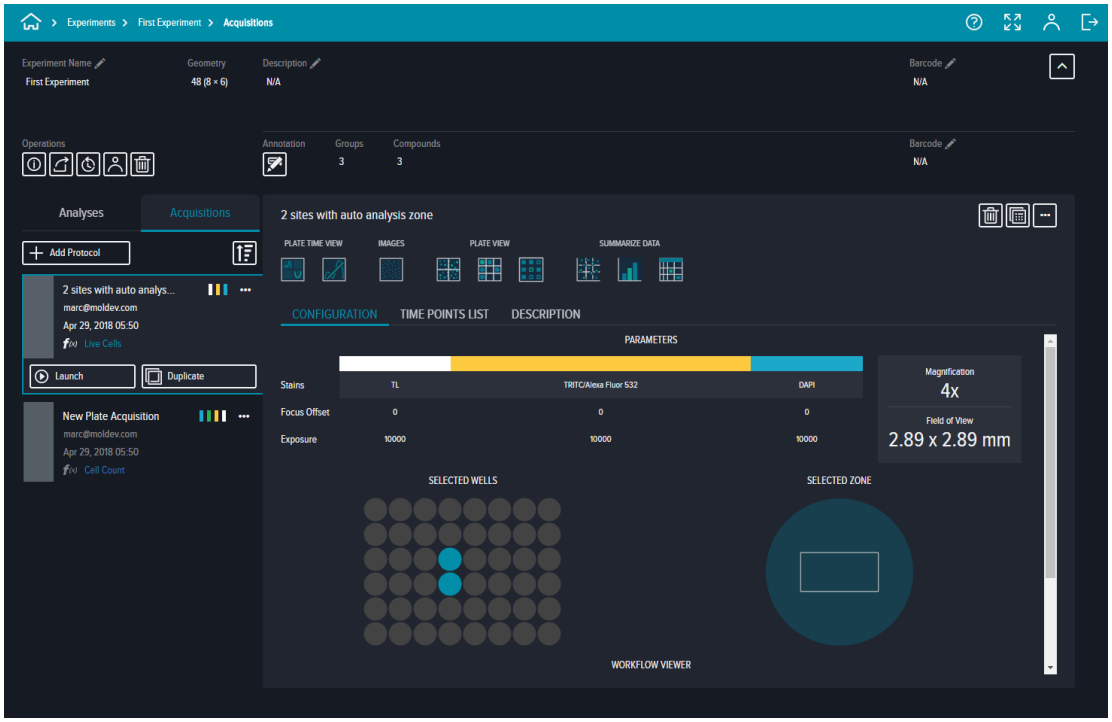
The section on the right shows analysis data on the following tabs:

- **Inputs**: Displays the parameters for the analysis set in **Acquisition** mode.
- **Time Points List**: Displays the timepoints for the acquisition and allows you to select specific timepoints to analyze.
- **Measurements**: Remove or export selected cell measurements.
- **Description**: View and edit the analysis name, description, and avatar.

You can also access various plate views or slide views. See Plate Views Overview on page 82 and Slide Views Overview on page 115 for details.

Experiment Acquisition Details

The bottom section of the **Experiments** page shows analysis and acquisition details for the experiment. The left pane in the bottom section contains two tabs: **Analyses** and **Acquisitions**. Click the **Acquisitions** tab to show acquisition details for the experiment.



On the **Acquisitions** tab, each acquisition for the experiment is listed. The following functions are available:

- **Launch**: Opens the acquisition for the experiment and enables you to perform additional acquisitions (using the same protocol).
- **Duplicate**: Opens the acquisition for the experiment and enables you to modify the protocol and perform additional acquisitions.

In addition, you can click  to save the protocol as a template or delete the acquisition.

The section on the right shows acquisition data on the following tabs:

- **Inputs**: Displays the parameters for the acquisition set in **Acquisition** mode.
- **Time Points List**: Displays the timepoints for the acquisition and allows you to get details on specific timepoints.
- **Description**: View and edit the analysis name, description, and avatar.

You can also access various plate views or slide views. See Plate Views Overview on page 82 and Slide Views Overview on page 115 for details.

Plate Views Overview

The following plate views are available:

Plate Views

 **Plate Time View**: Shows measurements over time for each well. See Plate Time View on page 84 for details.

 **Time Graph**: Shows measurements over time for selected wells. See Plate Time Graph on page 86 for details.

 **Thumbnail View**: Shows an overview of the well images in low resolution. See Plate Thumbnail View on page 87 for details.

 **Data View**: Shows a heatmap with up to four measurements displayed in the wells. See Plate Data View on page 89 for details.

 **Heatmap**: Shows a heatmap of one measurement. See Plate Heatmap on page 91 for details.

 **Images**: Shows high-resolution images for deep zoom viewing. See Plate Images on page 93 for details.

Summary Views

 **Scatter Plot**: Shows a scatter plot of two summary measurements. See Summary Scatter Plot on page 96 for details.

 **Stacked Bar**: Shows a histogram–style bar graph with up to two different measurements side by side. You can add a third measurement to subdivide each bar. See Summary Stacked Bar on page 98 for details.

 **Table**: Shows a table with summary well-level measurements. See Summary Table on page 101 for details.

Cellular Views

Note: You must select at least one well in a Plate view or a Summary view to enable the Cellular views.


Cell Level Density Heatmap: Shows a scatter plot-style graph of two measurements. See Cell Level Density Heatmap on page 104 for details.


Cell Level Stacked Bar: Shows a histogram–style bar graph with up to two different measurements side by side. You can add a third measurement to subdivide each bar. See Cell Level Stacked Bar on page 106 for details.

Cell Zoom Views

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.

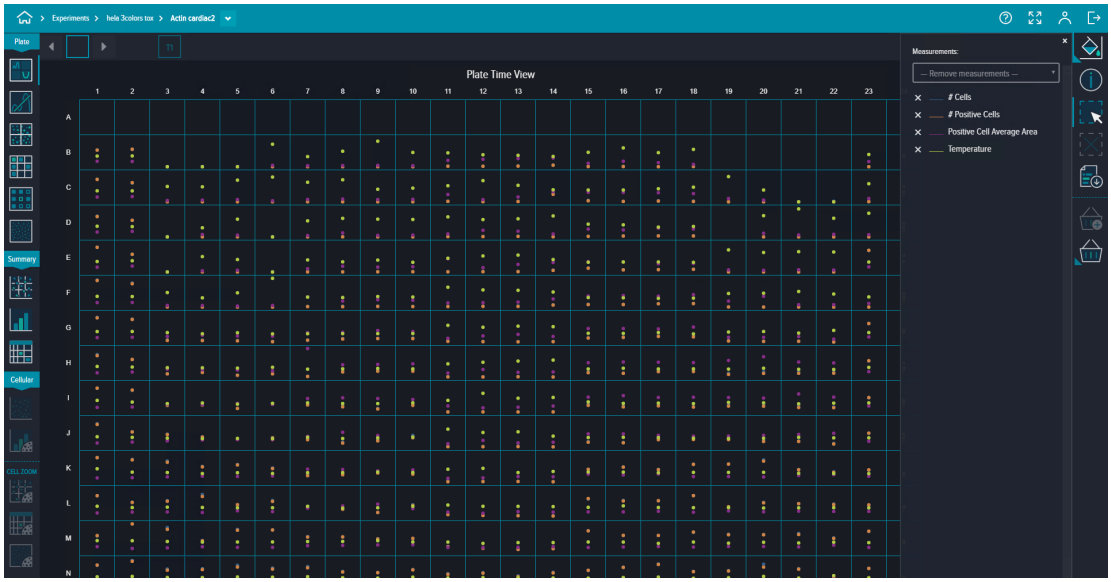

Scatter Mode: Shows a scatter plot graph of two measurements. See Cell Zoom Level Scatter Plot on page 108 for details.


Cell Level Table: Shows a table with cellular measurements. See Cell Zoom Level Table on page 110 for details.


Cell Level Images: Shows high-resolution images for deep zoom viewing of individual cells. See Cell Zoom Level Images on page 112 for details.

Plate Time View

The  **Plate Time View** shows measurements over time for each well.



The right side of the page includes the following icons:

 **Measurements**: Selects the measurements to show.

 **Well Info Mode**: Shows summary measurements for the selected well.

 **Selection Mode**: Activates selection mode, which enables you to select wells. Click and drag to select multiple wells. The number on the icon indicates the number of selected wells.

 **Deselect All**: Deselects all wells.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

 **Add to Quick List**: Saves selections to the Quick List for easy access.

 **Quick List**: Opens the Quick List where you can access saved selections.

Plate Time Graph

The  **Time Graph** shows measurements over time for selected wells.

Double-click on a well image to navigate to the **Images View** page for that well.

The right side of the page includes the following icons:

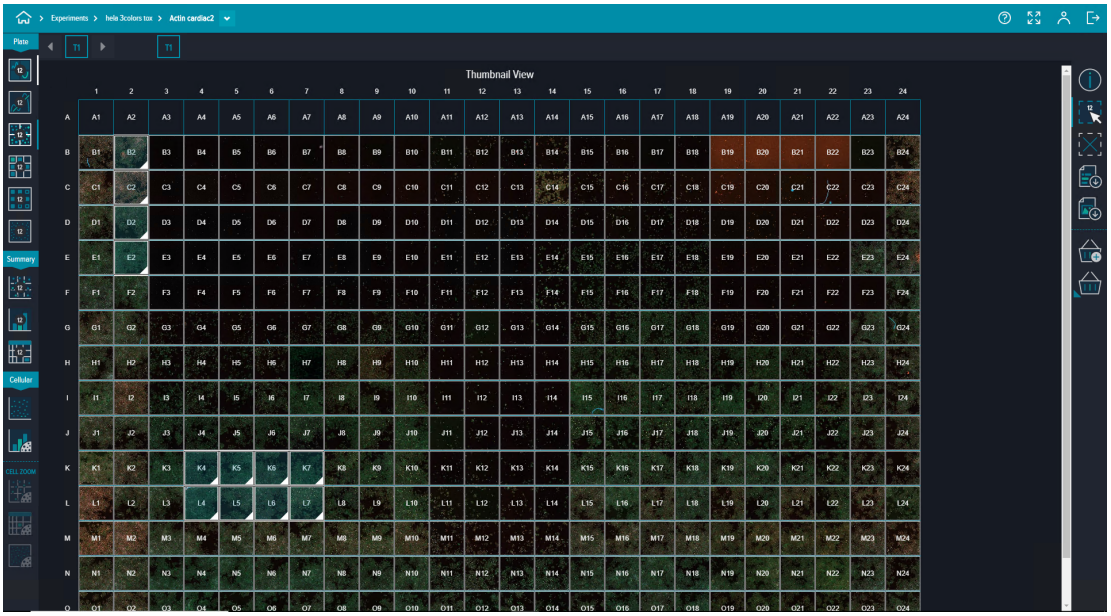
 **Measurements**: Selects the measurements to show.

 **Toggle Grid Lines**: Toggles the display of grid lines on the graph.

 **Toggle Value Labels**: Displays the numeric value for the selected measurement at each timepoint.

Plate Thumbnail View

The **Thumbnail View** shows an overview of the well images in low resolution. Double-click on a well image to view the **Image** for that well. See Plate Images on page 93 for details.



The right side of the page includes the following icons:

 **Well Info Mode**: Shows summary measurements for the selected well.

 **Selection Mode**: Activates selection mode, which enables you to select wells. Click and drag to select multiple wells. The number on the icon indicates the number of selected wells.

 **Deselect All**: Deselects all wells.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

 **Export Raw Images**: Exports TIFF images of the selected wells. This function is available with a Windows computer only. The MD Import/Export Service is required for this function. See Import Mode on page 163 for details.

 **Add to Quick List**: Saves selections to the Quick List for easy access.

 **Quick List**: Opens the Quick List where you can access saved selections.

Plate Data View

The  **Data View** shows a heatmap with up to four measurements displayed in the wells.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The right side of the page includes the following icons:


Measurements/Heatmap: Selects the measurements to show and the measurements to use for the heatmap.


Well Info Mode: Shows summary measurements for the selected well.


Selection Mode: Activates selection mode, which enables you to select wells. Click and drag to select multiple wells. The number on the icon indicates the number of selected wells.


Deselect All: Deselects all wells.


Export Current View to an Image: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

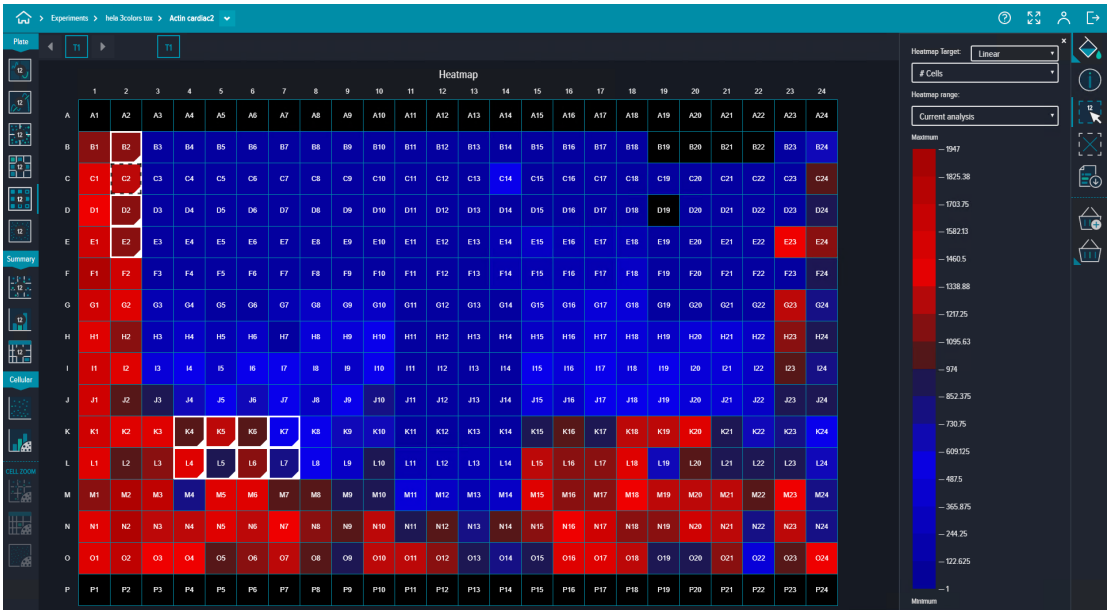

Add to Quick List: Saves selections to the Quick List for easy access.


Quick List: Opens the Quick List where you can access saved selections.

Plate Heatmap

The  **Heatmap** shows a heatmap of one measurement.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.



The right side of the page includes the following icons:


Color: Selects the measurement to use as the heatmap and the heatmap color scale to display.


Well Info Mode: Shows summary measurements for the selected well.


Selection Mode: Activates selection mode, which enables you to select wells. Click and drag to select multiple wells. The number on the icon indicates the number of selected wells.


Deselect All: Deselects all wells.


Export Current View to an Image: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

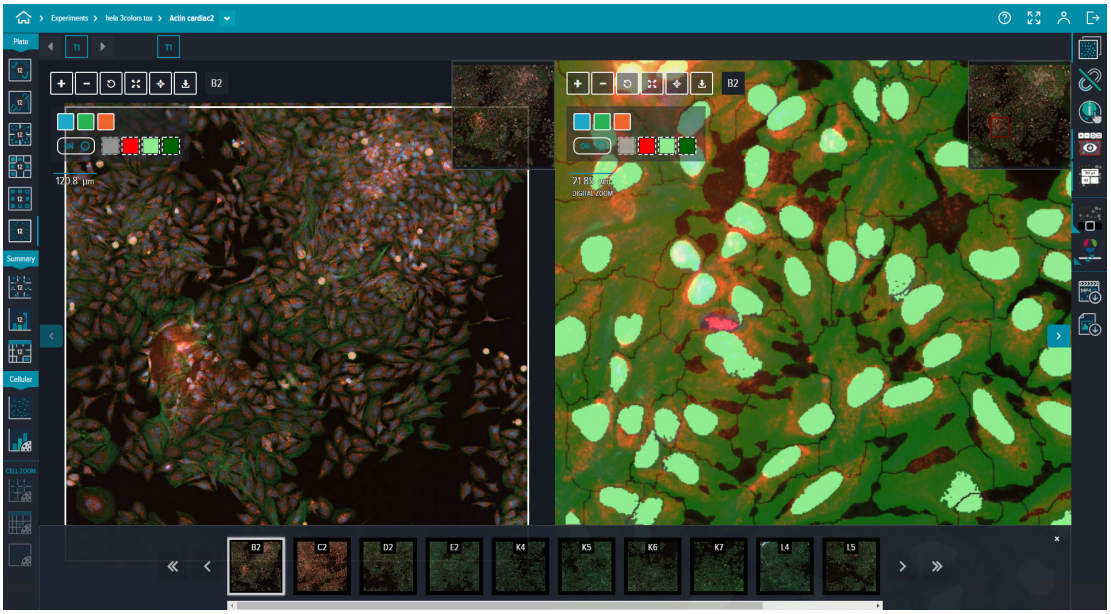

Add to Quick List: Saves selections to the Quick List for easy access.


Quick List: Opens the Quick List where you can access saved selections.

Plate Images

The **Images** show high-resolution images for deep zoom viewing.

Click on the image to zoom in on objects of interest. You can also use your mouse wheel to zoom in and out.



The right side of the page includes the following icons:

 **Show One Image / Show Two Images**: Toggles between a single image and two side-by-side images.

 **Link Images** or  **Unlink Images**: When showing two images, toggles between controlling images together or independently.

 **Cell Info Mode**: Shows cellular and summary measurements.

 **Show Navigation Maps**: Toggles the visualization tools in the top left corner and the mini-map in the upper right corner.

 **Show Scale and Zone**: Toggles the measurement scale and the well number.

 **Show Image Gallery**: Toggles the image gallery at the bottom of the screen.

 **Show Channel Settings**: Toggles the display scaling tools at the bottom of the screen.

Download MP4 Movie: Downloads an MP4 video of the current plate image over time. This function is not available with a tablet.

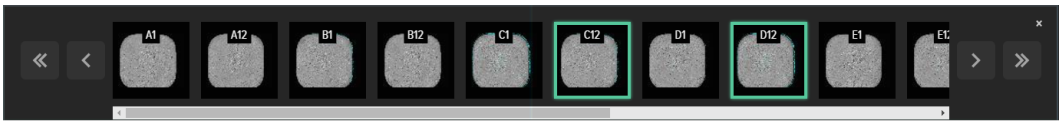
 **Export Raw Images**: Exports a TIFF image of the current plate image. This function is available with a Windows computer only. The MD Import/Export Service is required for this function. See Import Mode on page 163 for details.

Comparing Images

By default, a single image appears. You can display two views of the same image, which enables you to compare images.

To compare images:

1. Click  **Show Two Images**.
2. At the bottom of the page, in the image gallery, drag and drop image thumbnails to the comparison panes as needed.



3. To synchronize image zooming and changing positions in both panes, click  **Unlink Images**. The icon toggles to  **Link Images**.

Summary Scatter Plot

The **Scatter Plot** shows a scatter plot of two summary measurements. Use the mini map at the upper left as a guide while moving through data in the graph.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Selection Mode**: Activates selection mode, which enables you to select wells. Click and drag to select multiple wells. The number on the icon indicates the number of selected wells.

 **Deselect All**: Deselects all wells.

 **Show Selected Only**: Shows or hides the selected data or all the data.

 **Toggle Mini Map**: Shows or hides the small overview of the graph at the top left.

 **Toggle Grid Lines**: Toggles the display of grid lines on the graph.

 **Aggregation Function Line**: Not used in this version of the CellReporterXpress Software.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

 **Add to Quick List**: Saves selections to the Quick List for easy access.

 **Quick List**: Opens the Quick List where you can access saved selections.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.

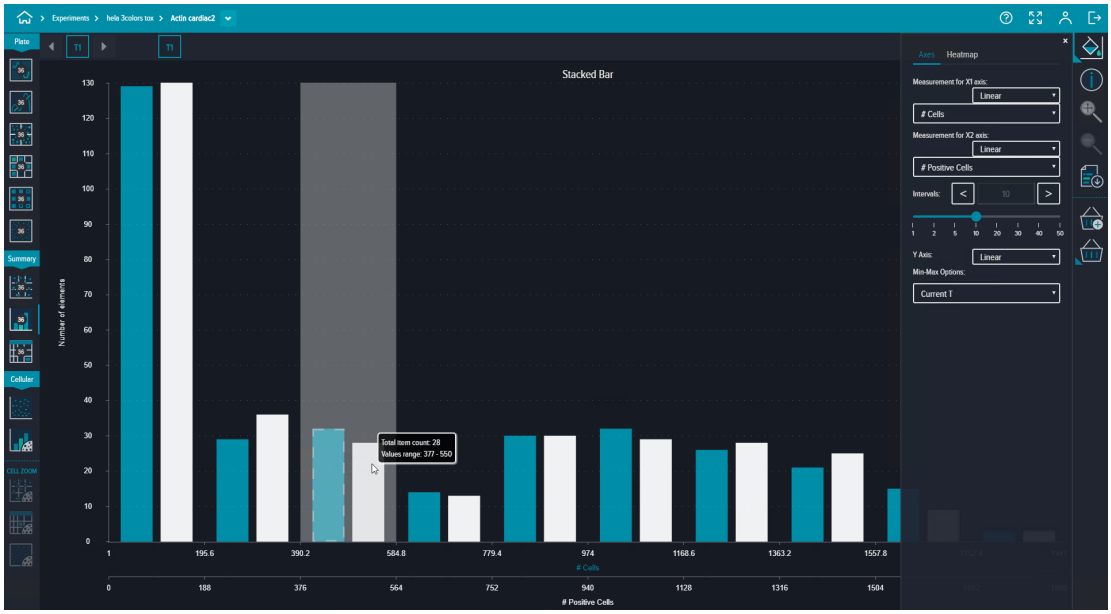
The following tabs are available:

- **Axes**: Specifies the measurements for two scatter plot axes.
- **Heatmap**: Specifies the heatmap coloring to the graph data.
- **Labels**: Specifies the label text next to the data in the graph.

Summary Stacked Bar

The **Stacked Bar** shows a histogram-style bar graph with up to two different measurements side by side. You can add a third measurement to subdivide each bar.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.



The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Well Info Mode**: Shows summary measurements for the selected well.

 **Zoom In**: When bins are selected, replots the selected bars only for a more granular view.

 **Zoom Out**: When bins are selected, replots the selected bars only for a more general view.

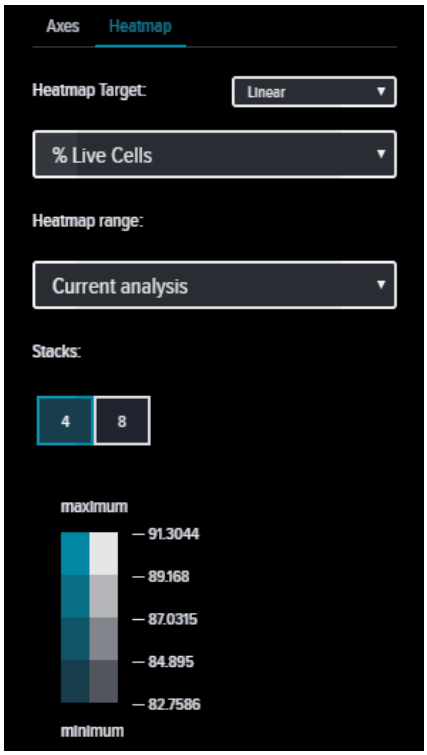
 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

 **Add to Quick List**: Saves selections to the Quick List for easy access.

 **Quick List**: Opens the Quick List where you can access saved selections.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.



The following tabs are available:

- **Axes**: Specifies the measurements for two scatter plot axes and intervals.
- **Heatmap**: Specifies the heatmap coloring to the graph data.

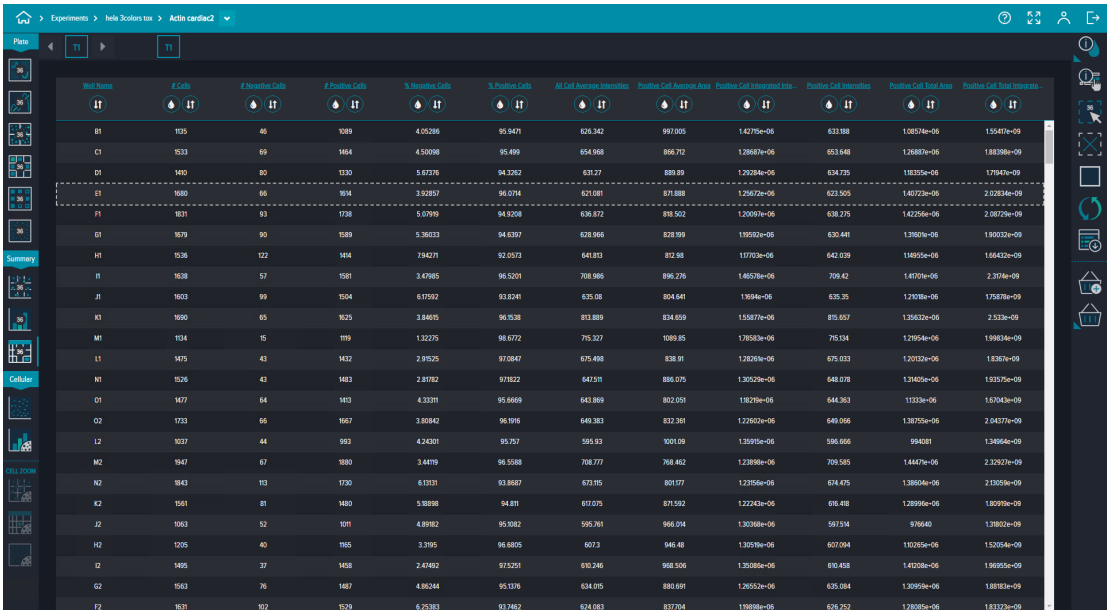
Stacks

Stacks show the range of values for each shade in the heatmap. The number of shades of each color can be adjusted to either **4** or **8**.

Summary Table

The  **Table** shows a table view of all well-level measurements.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.



The right side of the page includes the following icons:


Legend: Enables you to select a measurement and show the mapping between color and values. Provides information used when column heatmaps are active.


Well Info Mode: Shows summary measurements for the selected well.


Selection Mode: Activates selection mode, which enables you to select wells. Click and drag to select multiple wells. The number on the icon indicates the number of selected wells.


Deselect All: Deselects all wells.


Show Selected Only: Shows or hides the selected data or all the data.


Reset: Reverts the table to the default configuration.


Export: Downloads the currently configured table as a CSV file. This function is not available with a tablet.


Add to Quick List: Saves selections to the Quick List for easy access.

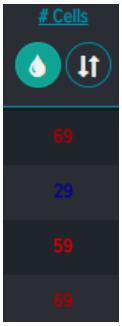

Quick List: Opens the Quick List where you can access saved selections.

Changing Columns

To change the column headings and associated data, click the heading name and then select from the menu options.

Adding Heatmap Coloring

To add heatmap coloring to a column of data, click  .



Sorting Data

To sort rows by the values in the column, click  .

- Click once to sort lowest to highest.
- Click again to sort highest to lowest.
- Click a third time to deactivate sorting for the column.

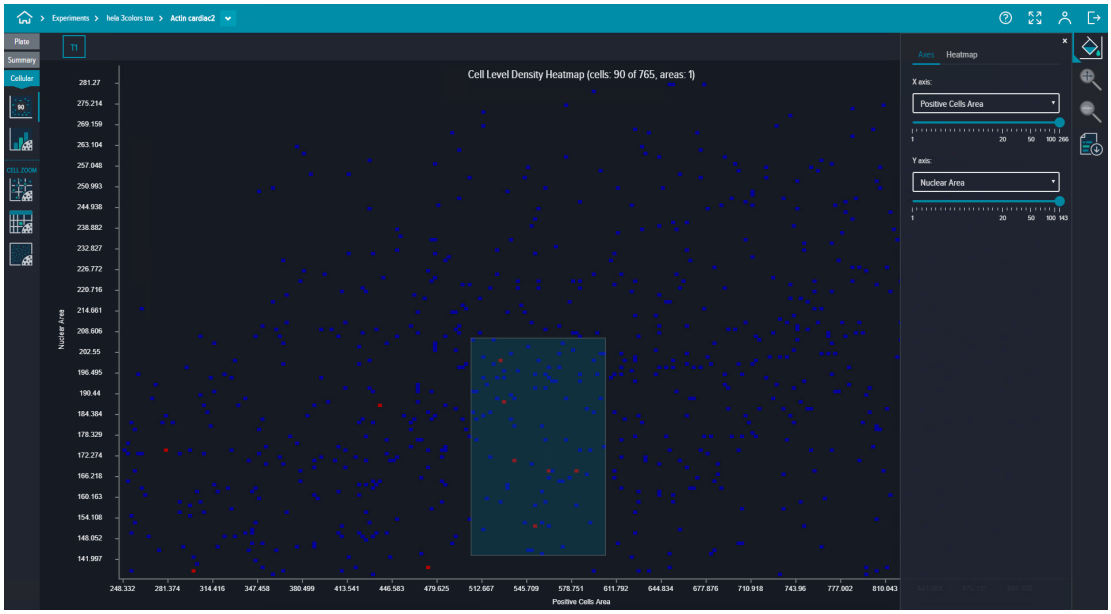
Cell Level Density Heatmap

The **Cell Level Density Heatmap** shows a scatter plot-style graph of two measurements. Each spot represents all the cells with similar measurements. The heatmap color of the spot is based on the cell count for the spot.

Note: You must select at least one well in a Plate view or a Summary view to enable the Cellular views.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The page heading indicates the number of selected cells (out of the total number of cells plotted on the graph) and the number of selected wells.



The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Zoom In**: When bins are selected, replots the selected bars only for a more granular view.

 **Zoom Out**: When bins are selected, replots the selected bars only for a more general view.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.

The following tabs are available:

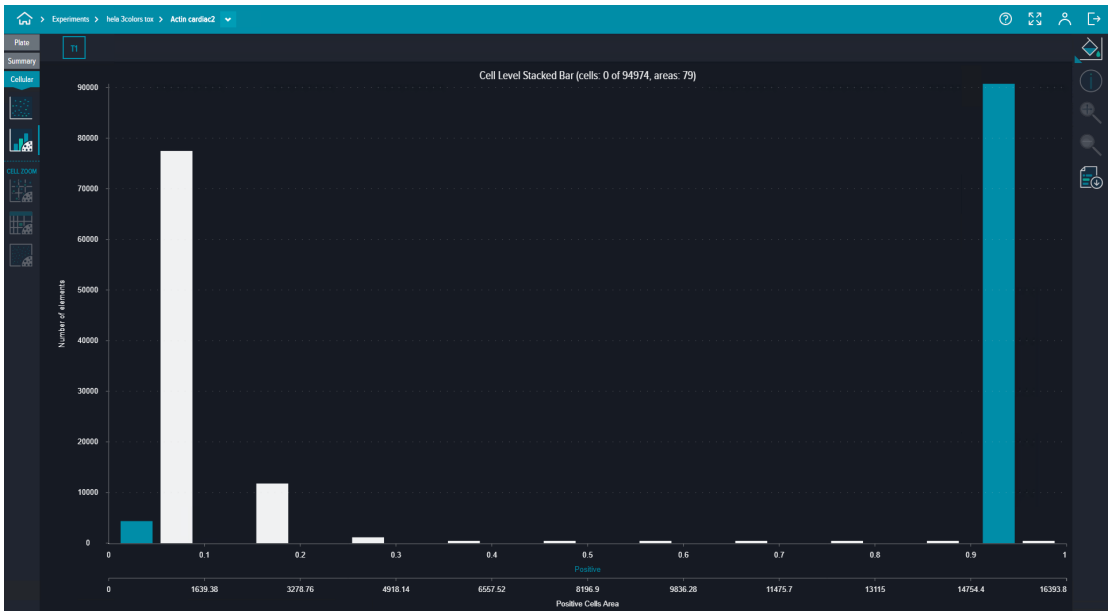
- **Axes**: Specifies the measurements for two scatter plot axes and intervals.
- **Heatmap**: Specifies the heatmap coloring to the graph data.

Cell Level Stacked Bar

The **Cell Level Stacked Bar** shows a histogram–style bar graph with up to two different measurements side by side. You can add a third measurement to subdivide each bar.

Note: You must select at least one well in a Plate view or a Summary view to enable the Cellular views.

The page heading indicates the number of selected cells (out of the total number of cells plotted on the graph) and the number of selected wells.



The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Cell Info**: Shows summary measurements for the selected well.

 **Zoom In**: When bins are selected, replots the selected bars only for a more granular view.

 **Zoom Out**: When bins are selected, replots the selected bars only for a more general view.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.

The following tabs are available:

- **Axes**: Specifies the measurements for two scatter plot axes and intervals.
- **Heatmap**: Specifies the heatmap coloring to the graph data.

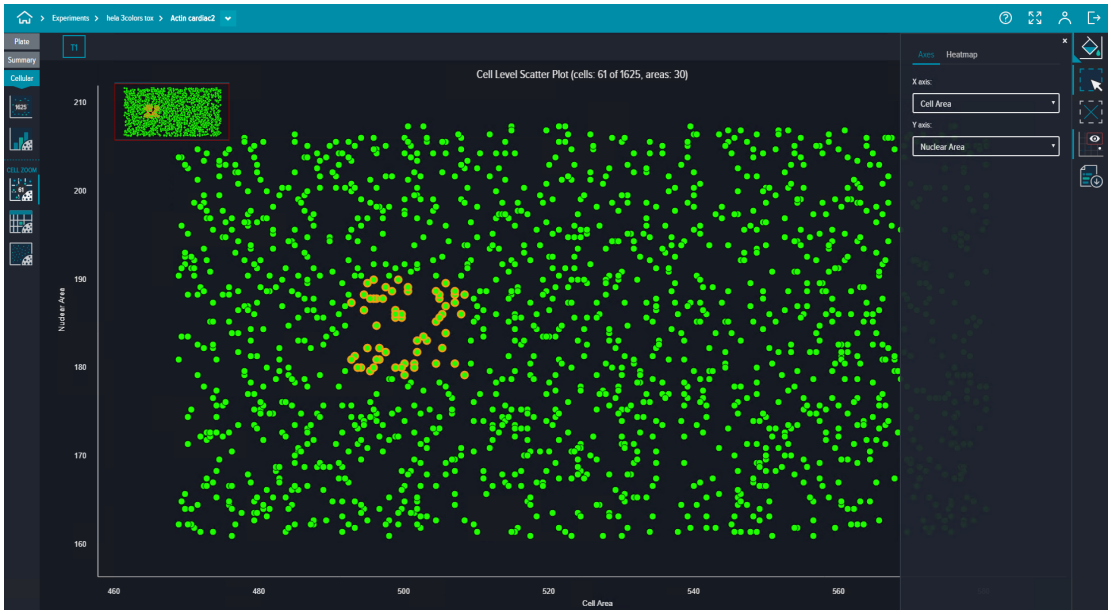
Cell Zoom Level Scatter Plot

The  **Scatter Mode** shows a scatter plot of two measurements.

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The page heading indicates the number of selected cells (out of the total number of cells plotted on the graph) and the number of selected wells.



The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Selection Mode**: Activates selection mode, which enables you to select wells. Click and drag to select multiple wells. The number on the icon indicates the number of selected wells.

 **Deselect All**: Deselects all wells.

 **Toggle Mini Map**: Shows or hides the small overview of the graph at the top left.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.

The following tabs are available:

- **Axes**: Specifies the measurements for two scatter plot axes.
- **Heatmap**: Specifies the heatmap coloring to the graph data.

Cell Zoom Level Table

The **Cell Level Table** shows a table with cell-level measurements.

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The right side of the page includes the following icons:


Legend: Enables you to select a measurement and show the mapping between color and values. Provides information used when column heatmaps are active.


Reset: Reverts the table to the default configuration.


Well Info: Shows the summary measurements for the last row selected.

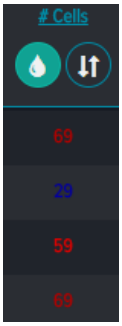

Export: Downloads the currently configured table as a CSV file. This function is not available with a tablet.

Changing Columns

To change the column headings and associated data, click the heading name and then select from the menu options.

Adding Heatmap Coloring

To add heatmap coloring to a column of data, click  .



Sorting Data

To sort rows by the values in the column, click  .

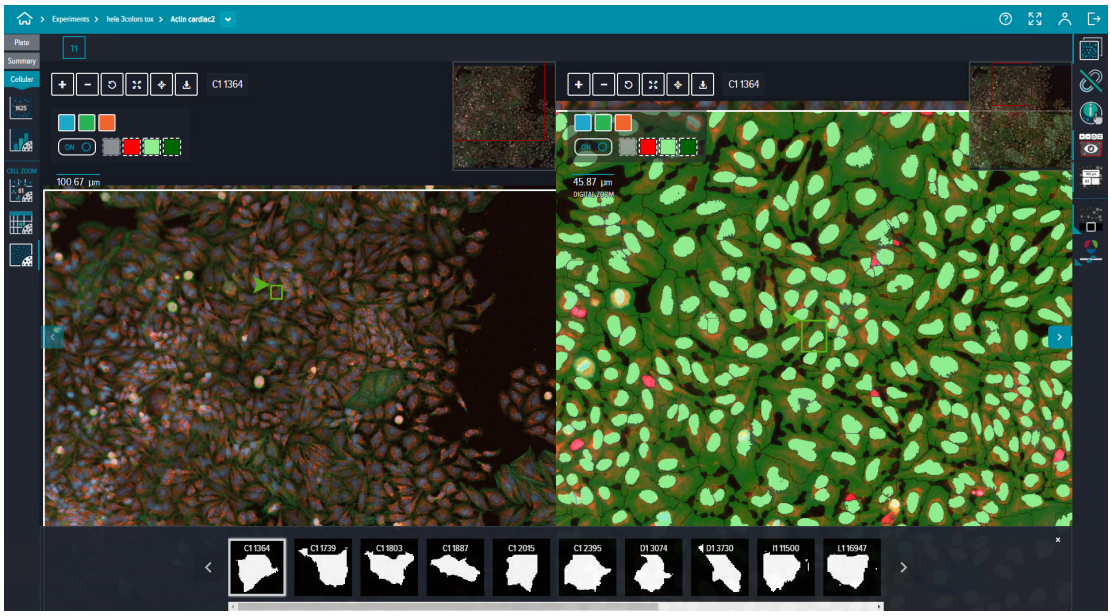
- Click once to sort lowest to highest.
- Click again to sort highest to lowest.
- Click a third time to deactivate sorting for the column.

Cell Zoom Level Images

The **Cell Level Images** page shows high-resolution images for deep zoom viewing. At the bottom of the page, zoom-level segments appear.

Click on the image to zoom in on objects of interest. You can also use your mouse wheel to zoom in and out.

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.



The right side of the page includes the following icons:

 **Show One Image / Show Two Images**: Toggles between a single image and two side-by-side images.

 **Link Images** or  **Unlink Images**: When showing two images, toggles between controlling images together or independently.

 **Cell Info Mode**: Shows cellular and summary measurements.

 **Show Navigation Maps**: Toggles the visualization tools in the top left corner and the mini-map in the upper right corner.

 **Show Scale and Zone**: Toggles the measurement scale and the well number.

 **Show Image Gallery**: Toggles the image gallery at the bottom of the screen.

 **Show Channel Settings**: Toggles the display scaling tools at the bottom of the screen.

Comparing Images

By default, a single image appears. You can display two views of the same image, which enables you to compare images.

To compare images:

1. Click  **Show Two Images**.
2. At the bottom of the page, in the image gallery, drag and drop image thumbnails to the comparison panes as needed.

3. To synchronize image zooming and changing positions in both panes, click  **Unlink Images**. The icon toggles to  **Link Images**.

Slide Views Overview

The following slide data views are available:

Slide Views


Thumbnail View: Shows an overview of the slide images in low resolution. See Slide Thumbnail View on page 117 for details.


Images: Shows high-resolution images for deep zoom viewing. See Slide Images on page 119 for details.

Summary Views


Table: Shows a table with summary slide region-level measurements. See Summary Table on page 122 for details.

Cellular Views

Note: You must select at least one region in a Slide view or a Summary view to enable the Cellular views.


Cell Level Density Heatmap: Shows a scatter plot-style graph of two measurements. See Cell Level Density Heatmap on page 125 for details.


Cell Level Stacked Bar: Shows a histogram-style bar graph with up to two different measurements side by side. You can add a third measurement to subdivide each bar. See Cell Level Stacked Bar on page 127 for details.

Cell Zoom Views

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.

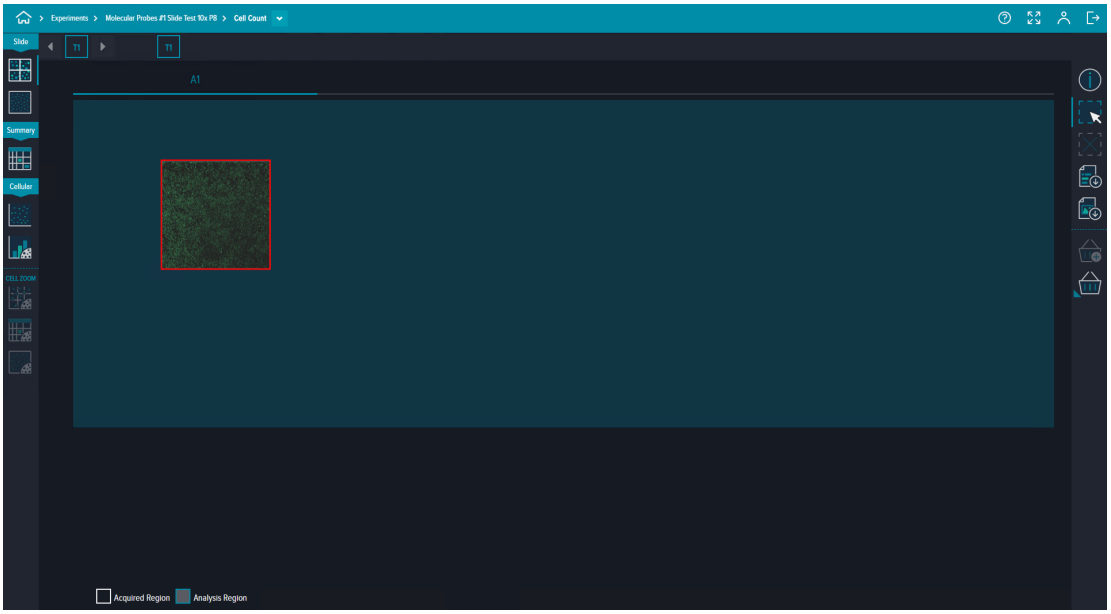

Scatter Mode: Shows a scatter plot graph of two measurements. See Cell Zoom Level Scatter Plot on page 129 for details.


Cell Level Table: Shows a table with cellular measurements. See Cell Zoom Level Table on page 131 for details.


Cell Level Images: Shows high-resolution images for deep zoom viewing of individual cells. See Cell Zoom Level Images on page 133 for details.

Slide Thumbnail View

The **Thumbnail View** shows an overview of the slide images in low resolution. Double-click on a region to view the **Image** for that region. See Slide Images on page 119 for details.



The right side of the page includes the following icons:


Well Info: Shows summary measurements for the selected region.


Selection Mode: Activates selection mode, which enables you to select regions. Press **SHIFT** and click to select multiple regions. The number on the icon indicates the number of selected regions.


Deselect All: Deselects all regions.


Export Current View to an Image: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.


Export Raw Images: Exports TIFF images of the selected regions. This function is available with a Windows computer only. The MD Import/Export Service is required for this function. See Import Mode on page 163 for details.

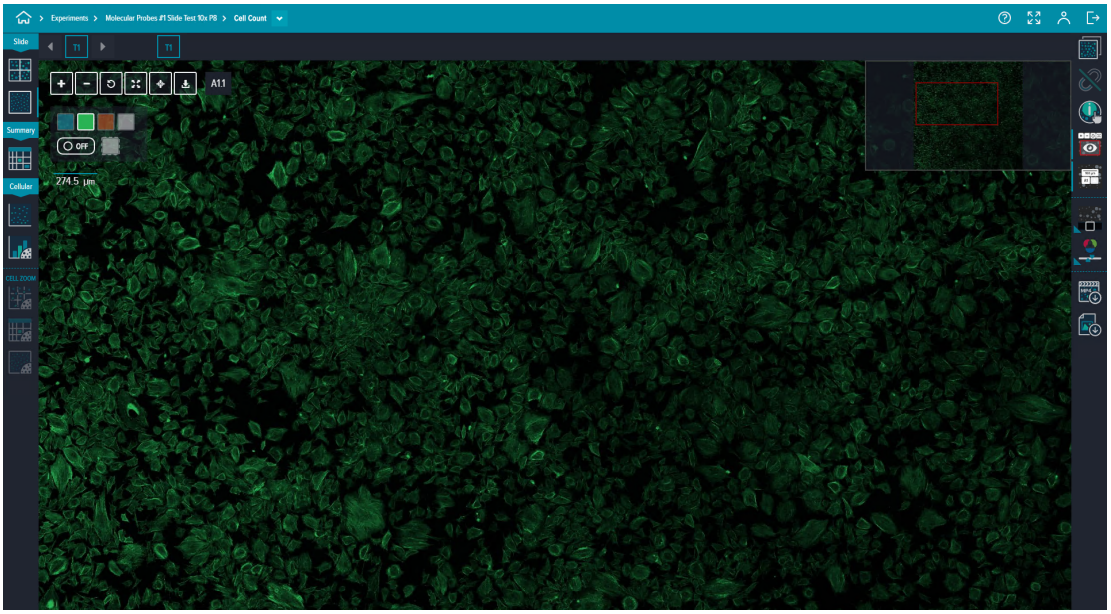

Add to Quick List: Saves selections to the Quick List for easy access.


Quick List: Opens the Quick List where you can access saved selections.

Slide Images

The  **Images** show high-resolution images for deep zoom viewing.

Click on the image to zoom in on objects of interest. You can also use your mouse wheel to zoom in and out.



The right side of the page includes the following icons:

 **Show One Image / Show Two Images**: Toggles between a single image and two side-by-side images.

 **Link Images** or  **Unlink Images**: When showing two images, toggles between controlling images together or independently.

 **Cell Info Mode**: Shows cellular and summary measurements.

 **Show Navigation Maps**: Toggles the visualization tools in the top left corner and the mini-map in the upper right corner.

 **Show Scale and Zone**: Toggles the measurement scale and the slide number.

 **Show Image Gallery**: Toggles the image gallery at the bottom of the screen.

 **Show Channel Settings**: Toggles the display scaling tools at the bottom of the screen.

Download MP4 Movie: Downloads an MP4 video of the selected slide image over time. This function is not available with a tablet.

 **Export Raw Images**: Exports TIFF images of the current slide image. This function is available with a Windows computer only. The MD Import/Export Service is required for this function. See Import Mode on page 163 for details.

Comparing Images

By default, a single image appears. You can display two views of the same image, which enables you to compare images.

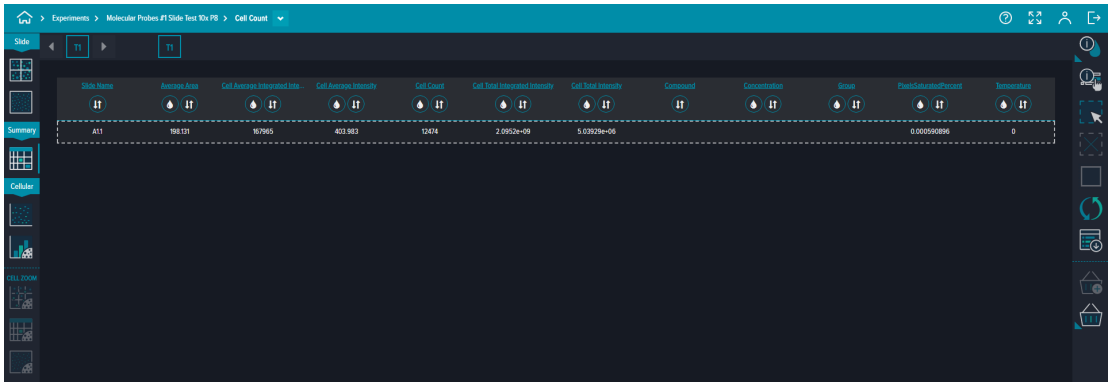
To compare images:

1. Click  **Show Two Images**.
2. At the bottom of the page, in the image gallery, drag and drop image thumbnails to the comparison panes as needed.
3. To synchronize image zooming and changing positions in both panes, click  **Unlink Images**. The icon toggles to  **Link Images**.

Summary Table

The  **Table** shows a table view of all region-level measurements.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.



The right side of the page includes the following icons:

 **Legend**: Enables you to select a measurement and show the mapping between color and values. Provides information used when column heatmaps are active.

 **Region Info Mode**: Shows the summary measurements for the last row selected.

 **Selection Mode**: Activates selection mode, which enables you to select regions. Press **SHIFT** and click to select multiple regions. The number on the icon indicates the number of selected regions.

 **Deselect All**: Deselects all regions.

 **Show Selected Only**: Shows or hides the selected data or all the data.

 **Reset**: Reverts the table to the default configuration.

 **Export**: Downloads the currently configured table as a CSV file. This function is not available with a tablet.

 **Add to Quick List**: Saves selections to the Quick List for easy access.

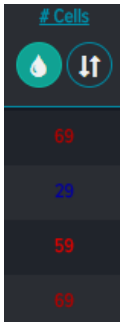
 **Quick List**: Opens the Quick List where you can access saved selections.

Changing Columns

To change the column headings and associated data, click the heading name and then select from the menu options.

Adding Heatmap Coloring

To add heatmap coloring to a column of data, click  .



Sorting Data

To sort rows by the values in the column, click  .

- Click once to sort lowest to highest.
- Click again to sort highest to lowest.
- Click a third time to deactivate sorting for the column.

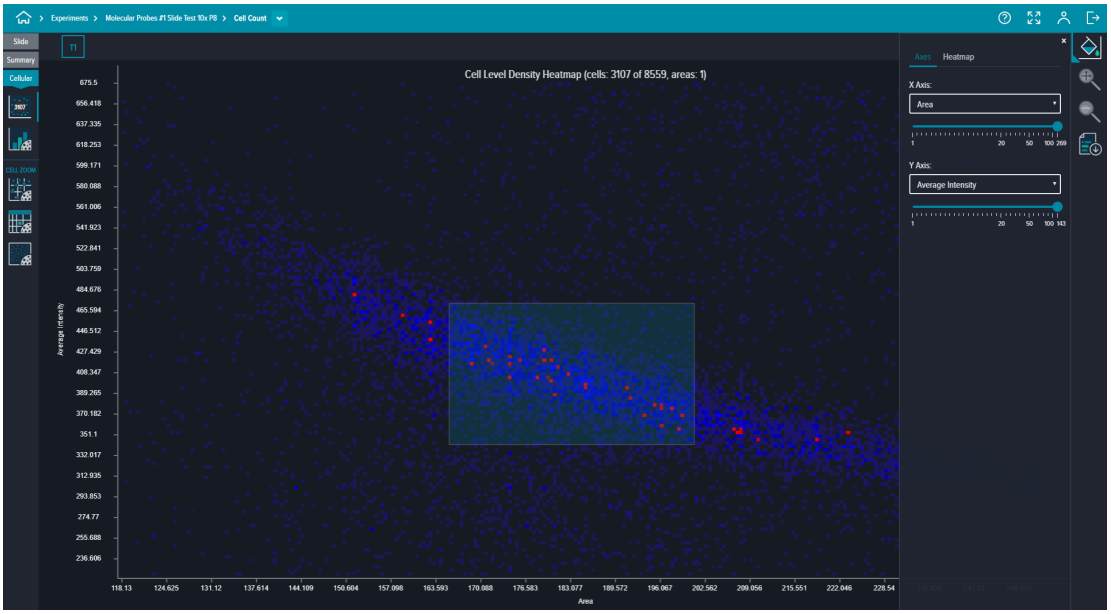
Cell Level Density Heatmap

The  **Cell Level Density Heatmap** shows a scatter plot-style graph of two measurements. Each spot represents all the cells with similar measurements. The heatmap color of the spot is based on the cell count for the spot.

Note: You must select at least one region in a Slide view or a Summary view to enable the Cellular views.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The page heading indicates the number of selected cells (out of the total number of cells plotted on the graph) and the number of selected regions.



The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Zoom In**: When bins are selected, replots the selected bars only for a more granular view.

 **Zoom Out**: When bins are selected, replots the selected bars only for a more general view.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.

The following tabs are available:

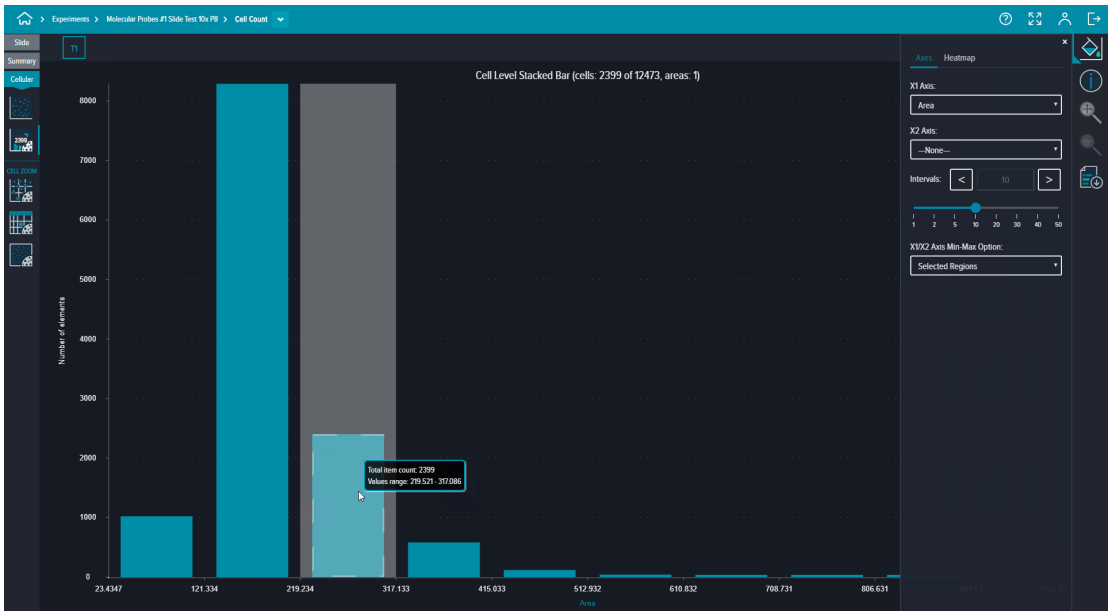
- **Axes**: Specifies the measurements for two scatter plot axes and intervals.
- **Heatmap**: Specifies the heatmap coloring to the graph data.

Cell Level Stacked Bar

The **Cell Level Stacked Bar** shows a histogram-style bar graph with up to two different measurements side by side. You can add a third measurement to subdivide each bar.

Note: You must select at least one region in a Slide view or a Summary view to enable the Cellular views.

The page heading indicates the number of selected cells (out of the total number of cells plotted on the graph) and the number of selected regions.



The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Cell Info**: Shows summary measurements for the selected region.

 **Zoom In**: When bins are selected, replots the selected bars only for a more granular view.

 **Zoom Out**: When bins are selected, replots the selected bars only for a more general view.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.

The following tabs are available:

- **Axes**: Specifies the measurements for two scatter plot axes and intervals.
- **Heatmap**: Specifies the heatmap coloring to the graph data.

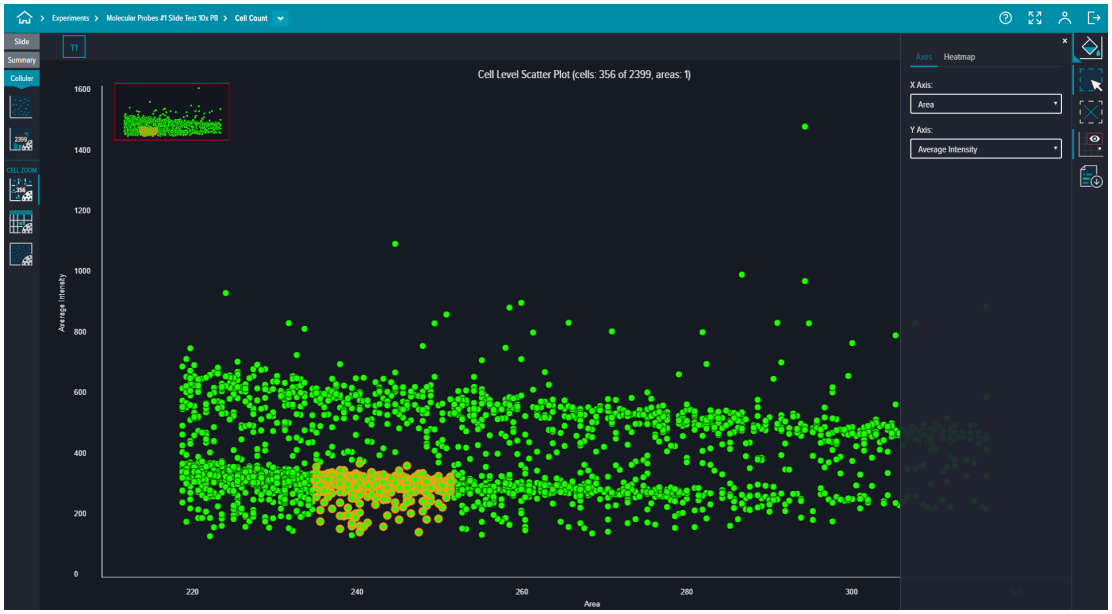
Cell Zoom Level Scatter Plot

The  **Scatter Mode** shows a scatter plot of two measurements.

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The page heading indicates the number of selected cells (out of the total number of cells plotted on the graph) and the number of selected regions.



The right side of the page includes the following icons:

 **Axes/Color**: Selects the measurements to show and the measurements to use for the heatmap.

 **Selection Mode**: Activates selection mode, which enables you to select regions. Press **SHIFT** and click to select multiple regions. The number on the icon indicates the number of selected regions.

 **Deselect All**: Deselects all regions.

 **Toggle Mini Map**: Shows or hides the small overview of the graph at the top left.

 **Export Current View to an Image**: Downloads a JPEG image of the page view, including selections. This function is not available with a tablet.

Axes/Color Pane

Use the  **Axes/Color** pane to specify the data that appears in the graph.

The following tabs are available:

- **Axes**: Specifies the measurements for two scatter plot axes.
- **Heatmap**: Specifies the heatmap coloring to the graph data.

Cell Zoom Level Table

The **Cell Level Table** shows a table with cell-level measurements.

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.

Tip: The heatmap color scheme can be changed in **Configuration Settings**. See Color Scheme on page 168 for details.

The right side of the page includes the following icons:


Legend: Enables you to select a measurement and show the mapping between color and values. Provides information used when column heatmaps are active.


Reset: Reverts the table to the default configuration.


Well Info: Shows the summary measurements for the last row selected.

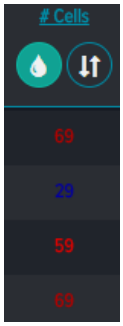

Export: Downloads the currently configured table as a CSV file. This function is not available with a tablet.

Changing Columns

To change the column headings and associated data, click the heading name and then select from the menu options.

Adding Heatmap Coloring

To add heatmap coloring to a column of data, click  .



Sorting Data

To sort rows by the values in the column, click  .

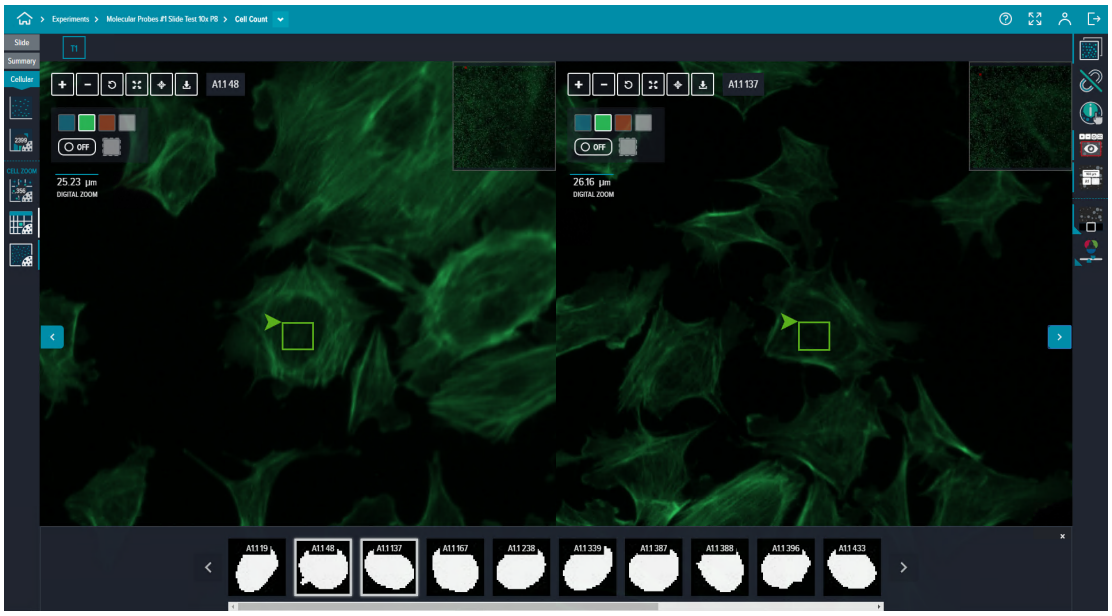
- Click once to sort lowest to highest.
- Click again to sort highest to lowest.
- Click a third time to deactivate sorting for the column.

Cell Zoom Level Images

The **Cell Level Images** page shows high-resolution images for deep zoom viewing. At the bottom of the page, zoom-level segments appear.

Click on the image to zoom in on objects of interest. You can also use your mouse wheel to zoom in and out.

Note: You must select at least one bin in a Cellular view to enable the Cell Zoom views.



The right side of the page includes the following icons:

 **Show One Image / Show Two Images**: Toggles between a single image and two side-by-side images.

 **Link Images** or  **Unlink Images**: When showing two images, toggles between controlling images together or independently.

 **Cell Info Mode**: Shows cellular and summary measurements.

 **Show Navigation Maps**: Toggles the visualization tools in the top left corner and the mini-map in the upper right corner.

 **Show Scale and Zone**: Toggles the measurement scale and the well number.

 **Show Image Gallery**: Toggles the image gallery at the bottom of the screen.

 **Show Channel Settings**: Toggles the display scaling tools at the bottom of the screen.

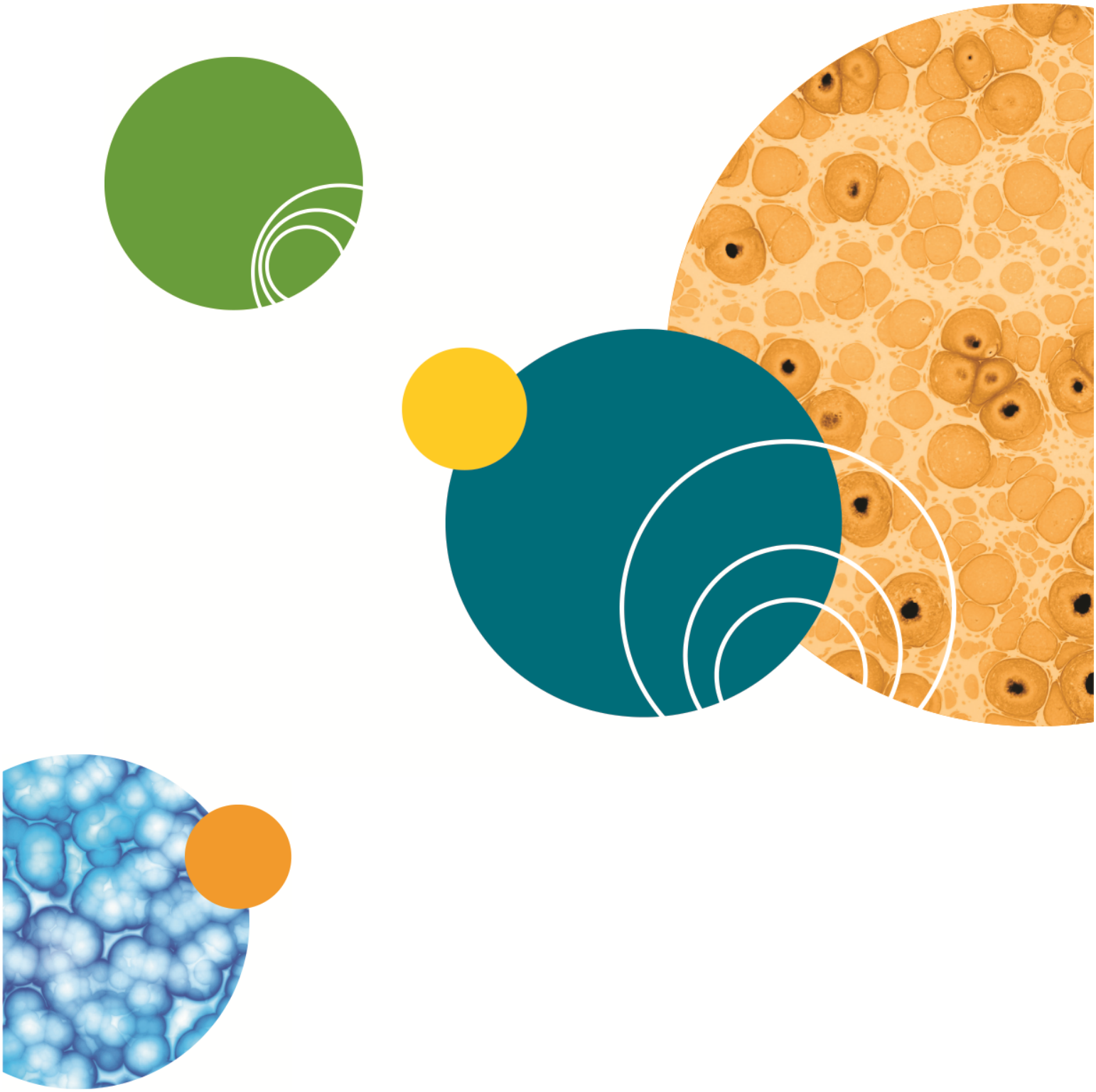
Comparing Images

By default, a single image appears. You can display two views of the same image, which enables you to compare images.

To compare images:

1. Click  **Show Two Images**.
2. At the bottom of the page, in the image gallery, drag and drop image thumbnails to the comparison panes as needed.

3. To synchronize image zooming and changing positions in both panes, click  **Unlink Images**. The icon toggles to  **Link Images**.

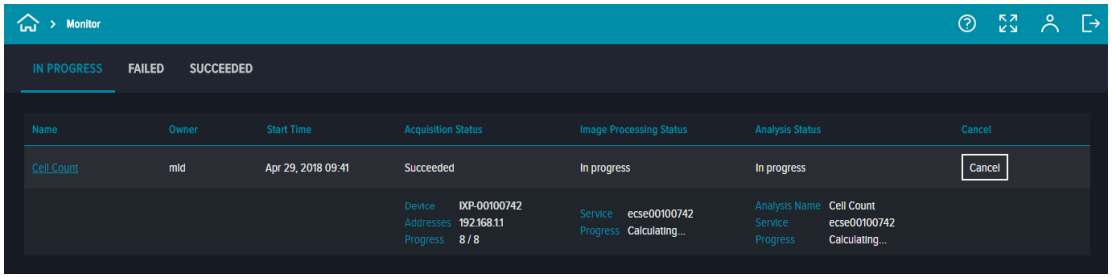


Chapter 5: Monitor Mode

5

Use **Monitor** mode to view the progress and completion status of experiments run from **Acquisition** mode or **Experiments** mode.

On the **Home** page, click the **Monitor** tile to enter **Monitor** mode.



The following tabs are available:

- **In Progress**: Displays details on currently running experiments.
- **Failed**: Displays details on failed experiments. An error message may appear to describe the reason for the failure. Failed experiment details remain listed on the **Failed** tab until you delete them.
- **Succeeded**: Displays details on successful experiments. Successful experiment details remain listed in this tab until you delete them.

Viewing the Analysis for an Experiment

To view the analysis for an experiment:

On the **Monitor** page, on any tab, click an experiment name in the **Name** field to view images and analysis data on the **Experiments** page.

Canceling a Running Experiment

To cancel a running experiment:

On the **Monitor** page, on the **In Progress** tab, click **Cancel**.

The experiment details move to the **Failed** tab and a card for the failed experiment is created on the **Experiments** page.

Responding to a Failed Experiment

There can be many reasons that an experiment fails. If the reason is caused by an issue with the CellReporterXpress Software or your network, the instrument may continue performing acquisition. In this case, the status light remains yellow. If you are using external temporary storage, you may be able to import the data for the failed experiment into the CellReporterXpress Software.

Note: If you are using internal temporary storage, images from a failed experiment cannot be imported.

To respond to a failed experiment:

1. On the **Monitor** page, on the **Failed** tab, review the error message.
2. Restart the instrument. See the *ImageXpress Pico User Guide* for details on restarting your instrument.
3. Check all network connections.
4. Confirm that you have enough temporary storage for the experiment. See the *ImageXpress Pico User Guide* for details on adding external temporary storage.
5. Retry running the experiment.

Deleting the Details of a Failed Experiment

To delete the details of a failed experiment:

1. On the **Monitor** page, on the **Failed** tab, select the **Delete** checkbox for each failed experiment you want to delete.
2. Click **Delete**.

Note: Only the details of the status of the failed experiment are deleted. The card for the failed experiment remains in **Experiments** mode.

Deleting the Details of a Successful Experiment

To delete the details of a successful experiment:

1. On the **Monitor** page, on the **Succeeded** tab, select the **Delete** checkbox for each successful experiment you want to delete.
2. Click **Delete**.

Note: Only the details of the status of the successful experiment are deleted. The card for the successful experiment remains in **Experiments** mode.

Chapter 6: Configuration Mode

6

Use **Configuration** mode to set the systemwide options that affect all users of the CellReporterXpress Software.

On the **Home** page, click the  **Configuration** tile to enter **Configuration** mode.

The left side of the page includes the following icons:

 **Stain Library**: Specifies the stain definitions available to all users of the CellReporterXpress Software. You can add, edit, and delete stain definitions as needed. See Stain Library on page 140 for details.

 **Labware Library**: Specifies the plate and slide holder configurations available to all users. A default library of plate and slide holder configurations is provided. You can add plate and slide holder configurations as needed. See Labware Library on page 141 for details.

 **Devices**: Specifies the imaging devices available to all users. You can add imaging devices as needed. See Devices on page 144 for details.

 **Image Analysis Computers**: Specifies the registered computers that are available for image analysis. This can include the host computer and (in a server configuration) any networked computers that have been registered to perform external image analysis. See Image Analysis Computers on page 147 for details.

 **Data Storage**: Specifies the registered computers and mapped folders that are available for image storage. This can include the host computer and (in a server configuration) any networked computers that have been registered to perform external image storage. See Data Storage on page 149 for details.

Stain Library

The  **Stain Library** page specifies the stain definitions available to all users of the CellReporterXpress Software. You can add, edit, and delete stain definitions as needed.

Adding a Stain Definition

To add a stain definition to the library:

1. On the **Stain Library** page, click **Add Stain**.
2. In the **Group Name** field, enter the stain-equivalent filter name.
3. In the **Stain Name** field, enter the dye name.
4. In the **Color** field, click the dropdown and select a representative display color.
5. In the **Excitation** field, enter the center excitation wavelength for the new stain. This value determines the LEDs used to illuminate the sample.
6. In the **Emission** field, enter the center emission wavelength for the new stain. This value determines which filter cube is used for detection.
7. In the **Edit** field, click  **OK**.

Editing a Stain Definition

To edit a stain definition in the library:

1. On the **Stain Library** page, in the row for the stain you want to edit, click  **Edit**.
2. Make the changes as needed.
3. In the **Edit** field, click  **OK**.

Deleting a Stain Definition

To delete a stain definition from the library:

1. On the **Stain Library** page, in the row of the stain you want to delete, click  **Delete**.
2. Click **OK**.

Labware Library

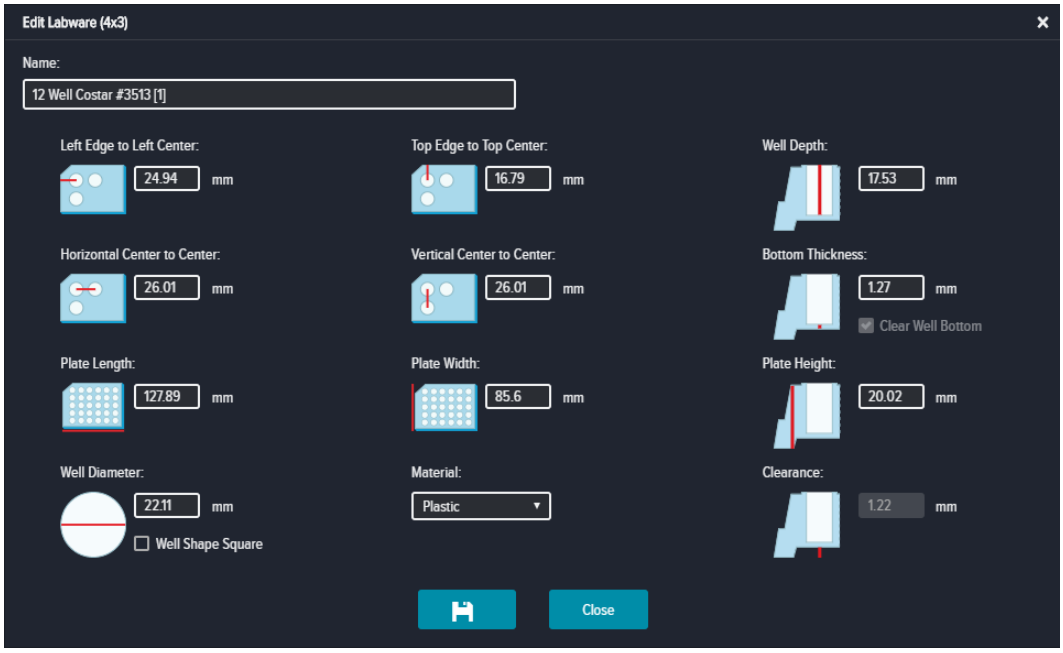
The  **Labware Library** page specifies the plate and slide holder configurations available to all users. A default library of plate and slide holder configurations is provided. You can add plate and slide holder configurations as needed.

Adding a Plate Configuration

To add a plate configuration to the library:

1. On the **Labware Library** page, click the **Plates** tab.
2. In the row of a plate configuration that is similar to the one you want to add, click  **Duplicate**.
3. In the new row, click  **Measure Plate Dimensions**.
4. Click the **Select the instrument** dropdown and select the instrument you are using.
5. Click **Open Door**.
6. Insert the plate for the new configuration.
7. Click **Close Door**.
8. Click **Measure Plate Dimensions**.
   The instrument measures the well depth and bottom thickness.
9. Click **Finish**.

10. Click **Edit**.



11. In the **Name** field, enter a name for the new plate configuration.
12. Edit the specifications for the plate configuration as needed.
13. Click **Save**.

Adding a Slide Holder Configuration

To add a slide holder configuration to the library:

1. On the **Labware Library** page, click the **Slides** tab.
2. In the row of a slide holder configuration that is similar to the one that you want to add, click **Duplicate**.
3. In the new row, click **Edit**.

4. In the **Name** field, enter a name for the new slide holder configuration.
5. Edit the specifications for the slide holder configuration as needed.
6. Click **Save**.

Deleting a Configuration

You can delete a plate or slide holder configuration that you have added to the library.

Note: You cannot delete a default configuration.

To delete a configuration from the library:

1. On the **Labware Library** page, click the tab (either **Plates** or **Slides**) for the configuration you want to delete.
2. In the row of the configuration you want to delete, click **Delete**.
3. Click **OK**.

Devices

The  **Devices** page specifies the imaging devices available to all users. You can add imaging devices as needed using one of the following connections:

- **Direct Connection**: A direct Ethernet connection between the imaging device and the host computer running the CellReporterXpress Software in a standalone configuration.
- **Remote Connection**: A network Ethernet connection between the imaging device and the host computer running the CellReporterXpress Software in a network configuration or a server configuration.

See the *CellReporterXpress Installation Guide* for details on setting up the various configurations.

The Available Acquisition Devices list shows all registered imaging devices and their current status, which can be one of the following:

 : Indicates online status.

 : Indicates offline status.

 : Indicates busy status.

 : Indicates error status.

The tile for each device contains a  **Favorite** icon. Click  **Favorite** to indicate a frequently used device. Note that the favorite setting affects your login only.

Registering a Directly Connected Imaging Device

To register a directly connected imaging device:

1. Confirm that the imaging device is connected to the host computer running the CellReporterXpress Software using the **LAN1** port on the back of the device.
2. On the **Devices** page, click **LAN1**.

The imaging device appears in the **Available Acquisition Devices** list.

Tip: Molecular Devices recommends that you do not directly connect an imaging device to the host computer using the LAN2 port unless advised to do so by Molecular Devices Technical Support.

Registering a Remotely Connected Imaging Device Using Autodiscovery

To register a remotely connected imaging device using autodiscovery:

1. Confirm that the imaging device is connected to the network using the **LAN2** port on the back of the device.
2. Confirm that the host computer running the CellReporterXpress Software is connected to the network.

Within a few minutes, the imaging device appears in the **Available Acquisition Devices** list. It may initially indicate offline status, but should change to online status shortly after it appears.

If the imaging device does not appear in the **Available Acquisition Devices** list, your computer or your network may be set up to block autodiscovery. In this case, try to register the imaging device using manual discovery.

Registering a Remotely Connected Imaging Device Using Manual Discovery

Your computer or your network may be set up to block autodiscovery. In that case, you can register a remotely connected imaging device using manual discovery.

To register a remotely connected imaging device using manual discovery:

1. Confirm that the imaging device is connected to the network using the **LAN2** port on the back of the device.
2. Confirm that the host computer running the CellReporterXpress Software is connected to the network.
3. In the **Remote Connection** field, enter the name or the IP address of the imaging device you want to add.

   Note: The imaging device name begins with IXP- followed by the serial number, which is on the back of the instrument. See the *ImageXpress Pico User Guide* for details on locating the serial number.

   The device IP address is determined by your network. Contact your network administrator for details.

4. Click **Register Device**.

The remotely connected imaging device appears in the **Available Acquisition Devices** list.

If the imaging device does not appear in the **Available Acquisition Devices** list, contact your network administrator.

Removing a Registered Imaging Device

To remove an imaging device from the Available Acquisition Devices list:

1. In the **Available Acquisition Devices** list, locate the instrument you want to delete and click **Delete**.
2. Click **OK**.

Image Analysis Computers

The  **Image Analysis Computers** page specifies the registered computers that are available for image analysis. This can include the host computer and (in a server configuration) any networked computers that have been registered to perform external image analysis. The CellReporterXpress Software determines which registered computer will be used for each analysis.

In a server configuration, any registered computer running MD.AnalysisService can perform image analysis. See the *CellReporterXpress IT Configuration Guide* for details on setting up a server configuration.

All registered computers appear in the Registered Image Analysis Computers list with one of the following status indicators:

 : Indicates online status.

 : Indicates offline status.

A registered computer may indicate offline status due to an issue with the network, the firewall, or the MD.AnalysisService (that is, it is not present or not started).

Registering a Computer for Image Analysis

To register a computer for image analysis:

1. On the **Image Analysis Computers** page, in the **Add Image Analysis Computer** field, enter the PC name or the IP address of the computer you want to register.
2. Click  **Add Image Analysis Computer**.

The computer appears in the **Registered Image Analysis Computers** list.

Removing a Registered Computer

You can remove a computer from the Registered Image Analysis Computers list, which prevents it from being used to perform image analysis. After you remove a registered computer, the MD.AnalysisService remains on that computer.

To remove a registered computer:

1. On the **Image Analysis Computers** page, in row for the registered computer you want to remove, click  **Delete**.
2. Click **OK**.

Restarting the Analysis Service

If a registered computer indicates offline status or an error occurs when testing analysis, you may need to restart the MD.AnalysisService on that computer.

To restart the analysis service on a registered computer:

1. On the **Image Analysis Computers** page, in row for the registered computer with the analysis service you want to restart, click  **Restart**.
2. Click **OK**.

Data Storage

The  **Data Storage** page specifies the registered computers and mapped folders that are available for image storage. This can include the host computer and (in a server configuration) any networked computers that have been registered to perform external image storage. You can select the registered computer and mapped folder to be used for storage when you run a protocol.

In a server configuration, any registered computer running MD.LocationService can be used for image storage. See the *CellReporterXpress Installation Guide* for details on setting up a server configuration.

All registered computers appear in the List of Registered Storage Computers and all mapped folders appear in the List of Mapped Folders with one of the following status indicators:

 : Indicates online status.

 : Indicates offline status.

A registered computer or mapped folder may indicate offline status due to an issue with the network, the firewall, or the MD.LocationService (that is, it is not present or not started).

Registering a Computer for Image Storage

To register a computer for image storage:

1. On the **Data Storage** page, in the **Add Data Storage Computer** field, enter the PC name or the IP address of the computer you want to register.
2. Click  **Add Data Storage**.

The computer appears in the **List of Registered Storage Computers**.

Mapping a Folder for Image Storage

If a registered computer is running the MD.LocationService, the following folder is mapped by default and appears in the List of Mapped Folders:

```
C:\ProgramData\Molecular Devices\MD.LocationService\Data
```

You can map other folders on a registered computer running the MD.LocationService.

To map a folder for image storage:

1. On the **Data Storage** page, in the **Map Folder on Storage Computer** field, enter the full path of the folder you want to map.
2. Click  **Map Existing Folder on Storage Computer**.

The mapped folder appears in the **List of Mapped Folders**.

Removing a Registered Computer

You can remove a computer from the List of Registered Storage Computers, which prevents it from being used for image storage. After you remove a registered computer, the MD.LocationService remains on that computer.

To remove a registered computer:

1. On the **Data Storage** page, in row for the registered computer you want to remove, click  **Delete**.
2. Click **OK**.

Removing a Mapped Folder

You can remove a mapped folder from the List of Mapped Folders, which prevents it from being used for image storage. After you remove a mapped folder, the folder and the images it contains remain on the computer.

To remove a mapped folder:

1. On the **Data Storage** page, in row for the mapped you want to remove, click  **Delete**.
2. Click **OK**.

Chapter 7: Devices Mode

Use **Devices** mode to manage and configure instruments for acquisition, including installing and calibrating objectives and filter cubes and controlling the temperature inside the instrument.

On the **Home** page, click the  **Devices** tile to enter **Devices** mode.

The right side of the page includes the following icons:

 **Shutdown Device**: Prepares the software to power off the selected instrument.

 **Restart Device**: Restarts the selected instrument.

 **Open Plate Door**: Opens the stage door on the selected instrument so that you can insert or remove labware.

 **Close Plate Door**: Closes the stage door.

The **Devices** page shows the imaging devices available to all users in the **Available Acquisition Devices** list. Click  **Show Device Options** to display the **Info** tab, which shows details for the selected device. From there, you can select other tabs, which enable you to do the following:

- Control the temperature inside the instrument and set a toolbar notification to help you monitor the temperature.
- Exchange and calibrate objectives.
- Exchange and calibrate filter cubes.

See Devices on page 144 in **Configuration** mode for details on adding an imaging device to the **Available Acquisition Devices** list.

Info

The **Info** tab displays details about your instrument, including the following:

- Device Name
- Serial Number
- Device Model
- Connected on
- Version
- Free Space
- IP
- MAC
- Number of installed objectives
- Number installed filter cubes

Sensors

The **Sensors** tab enables you to do the following:

- Review current temperature inside the instrument.
- Set a target value to regulate the temperature inside the instrument at 6°C (11°F) above ambient within a range of 25°C to 40°C (77°F to 104°F).
- Set a toolbar notification to help you monitor temperature inside the instrument.

Using Temperature Control

You can regulate the temperature inside the instrument at 6°C (11°F) above ambient within a range of 25°C to 40°C (77°F to 104°F). When you set a target temperature and activate temperature control, the instrument warms the air to the target temperature. The current temperature inside the instrument is shown on the **Sensors** tab and on the temperature toolbar notification (if enabled).

Notes:

- For best results, allow the instrument to reach the target temperature before inserting the sample.
- Molecular Devices recommends that the ambient room temperature be between 20°C and 24°C (68°F and 75°F) when using temperature control.
- The temperature sensor detects the temperature inside the instrument, not the temperature of the samples in the plate.
- You may want to use a seal or lid on the sample to prevent evaporation and maintain uniform temperature.
- Once warmed, it may take longer for the temperature inside the instrument to cool than it took to warm it.
- The CellReporterXpress Software does not support adjusting the temperature during a time series acquisition. If your acquisition requires this, you can perform a discontinuous time series by acquiring the first set of time points, adjusting the temperature as needed, and then acquiring the next set of time points. See Time Series on page 37 for details.

Starting Temperature Control

To start temperature control:

1. On the **Devices** page, click **Show Device Options** to expand the details for the device where you want to start temperature control.
2. Click the **Sensors** tab.
3. In the **Temperature** row, under **Component State**, in the **Target** field, enter a target temperature value in degrees Celsius.

   Note: The target temperature value must be within the range of 25°C to 40°C (77°F to 104°F).

4. Click **Start Regulation**.

The indicator in the **State** field enables to show that temperature control is on.

Modifying Temperature Control

When temperature control is on, you can set a new temperature.

To start temperature control:

1. On the **Devices** page, click **Show Device Options** to expand the details for the device where you want to set a new temperature.
2. Click the **Sensors** tab.
3. In the **Temperature** row, under **Component State**, in the **Target** field, enter a new target temperature value in degrees Celsius.

   Note: The target temperature value must be within the range of 25°C to 40°C (77°F to 104°F).

4. Click **Start Regulation**.

The indicator in the **State** field remains enabled.

Stopping Temperature Control

To stop temperature control:

1. On the **Devices** page, click  **Show Device Options** to expand the details for the device where you want to start temperature control.
2. Click the **Sensors** tab.
3. In the **Temperature** row, under **Component State**, click  **Stop Regulation**.

Setting a Toolbar Notification for Temperature

You can enable a toolbar notification to help you monitor temperature conditions. The toolbar notification appears at the top of the CellReporterXpress window. It changes color if the temperature is within the range you set (green) or outside of it (blue).

You can click on a toolbar notification to open the temperature control panel.

To set a toolbar notification for temperature:

1. On the **Devices** page, click  **Show Device Options** to expand the details for the device where you want to set a target temperature.
2. Click the **Sensors** tab.
3. In the **Temperature** row, under **Notification** Settings, in the **Min** field, enter the lower limit value for the temperature range in degrees Celsius.
4. In the **Max** field, enter the upper limit value for the temperature range in degrees Celsius.

Note: The lower limit and upper limit values must be within the range of 25°C to 40°C (77°F to 104°F).

5. Click  **Start/Stop Notification**.

Objectives

The **Objectives** tab contains a tile for each objective slot in your instrument. Each tile shows the registered objective for that slot and the calibration state of the objective. From here, you can install an objective and calibrate an objective.

Installing an Objective

Before installing an objective, review the following:

- Access only the user-serviceable components inside the enclosure as described in the procedure. Avoid contact with other components as they can be damaged or knocked out of alignment.
- To prevent dust from collecting inside the instrument, keep all access doors closed unless you are performing maintenance tasks.
- Ensure that all components and access doors are closed before starting the instrument.

In addition, observe the following when handling an objective:

CAUTION! To prevent skin oils from damaging the optical coatings, Molecular Devices recommends that you wear powder-free disposable gloves when handling objectives and filter cubes.

CAUTION! With the instrument power on, do not manually rotate the objective turret. Manually rotating the objective turret can damage the instrument.

Molecular Devices precalibrates the objectives to specific slots in the turret. You must install the objectives as follows:

Slot	Objective Magnification	Color Band
1	4x	Red
2	10x	Yellow
3	20x	Green
4	empty	n/a
5	40x or 63x	Light Blue or Dark Blue
6	empty	n/a

Note: Depending on how your ImageXpress Pico System is configured, you may not have all the objectives.

Note: The 40x objective and the 63x objective cannot be installed in the instrument simultaneously.

To install an objective:

1. On the **Devices** page, click **Show Device Options** to expand the details for the device where you want to install an objective.
2. Click the **Objectives** tab.
3. In the tile for the objective you want to install, click **Component Exchange**.
4. Click the **Choose Objective** dropdown and select the objective you want to install.
5. Click **Open Maintenance Door**.
6. If an objective is already installed in the slot, remove it from the instrument by gently turning it counterclockwise.

   CAUTION! When not installed in the instrument, an objective should always be stored in its case.

7. Install the objective in the slot by gently turning it clockwise.

   Note: When installing the objective, take care to avoid changing the correction collar setting.

8. Close the maintenance door.
9. In the CellReporterXpress Software, click **Close Maintenance Door**.
10. Click **Close**.

After you install a new objective, you may need to calibrate it. See Calibrating an Objective for details.

CAUTION! Retain the objective case for future storage needs. When not installed in the instrument, an objective should always be stored in its case.

Calibrating an Objective

After you install a new objective, you may need to calibrate it. **Molecular Devices precalibrates the objectives included with the initial purchase of the instrument. You must calibrate any objectives purchased after that time.**

A calibration kit, which ships with any after-sales objective purchase, includes the following items:

- Slide holder
- Stage micrometer slide
- Pink plastic slide
- Red plastic slide
- Bead slide
- Blank glass slide

To calibrate an objective:

1. On the **Devices** page, click  **Show Device Options** to expand the details for the device where you want to install an objective.
2. Click the **Objectives** tab.
3. In the tile for the objective you want to calibrate, click **Objective Calibration**.
4. Follow the on-screen instructions in the wizard to complete the calibration.

Filters

The **Filters** tab contains a tile for each filter cube slot in your instrument. Each tile shows the registered filter cube for that slot and the calibration state of the filter cube. From here, you can install a filter cube or calibrate a filter cube.

Installing a Filter Cube

Before installing a filter cube, review the following:

- Access only the user-serviceable components inside the enclosure as described in the procedure. Avoid contact with other components as they can be damaged or knocked out of alignment.
- To prevent dust from collecting inside the instrument, keep all access doors closed unless you are performing maintenance tasks.
- Ensure that all components and access doors are closed before starting the instrument.

In addition, observe the following when handling a filter cube:

CAUTION! To prevent skin oils from damaging the optical coatings, Molecular Devices recommends that you wear powder-free disposable gloves when handling objectives and filter cubes.

Molecular Devices precalibrates the filter cubes to specific slots in the turret. You must install the filter cubes as follows:

Slot	Filter Cube
1	DAPI
2	FITC
3	TRITC
4	Cy5
5	empty
6	empty

Note: Depending on how your ImageXpress Pico System is configured, you may not have all the filter cubes.

To install a filter cube:

1. On the **Devices** page, click  **Show Device Options** to expand the details for the device where you want to install an objective.
2. Click the **Filters** tab.
3. In the tile for the filter cube you want to install, click **Component Exchange**.
4. Click the **Choose Filter** dropdown and select the filter cube you want to install.
5. Click **Open Maintenance Door**.
6. If needed, slightly rotate the filter cube turret by hand to get direct access to the filter cube slot.
7. If a filter cube is already installed in the slot, remove it from the instrument by gently pulling it toward you.

    **CAUTION!** When not installed in the instrument, a filter cube should always be stored in its original packaging.

8. Install the filter cube in the slot by gently pushing it into the slot.

    **Tip:** The filter cube should "snap" into place.

9. Close the maintenance door.
10. In the CellReporterXpress Software, click **Close Maintenance Door**.
11. Click **Close**.

After you install a filter, you may need to calibrate it. See Calibrating a Filter Cube for details.

 **CAUTION!** Retain the packing material from the filter cube for future storage needs. When not installed in the instrument, a filter cube should always be stored properly.

Calibrating a Filter Cube

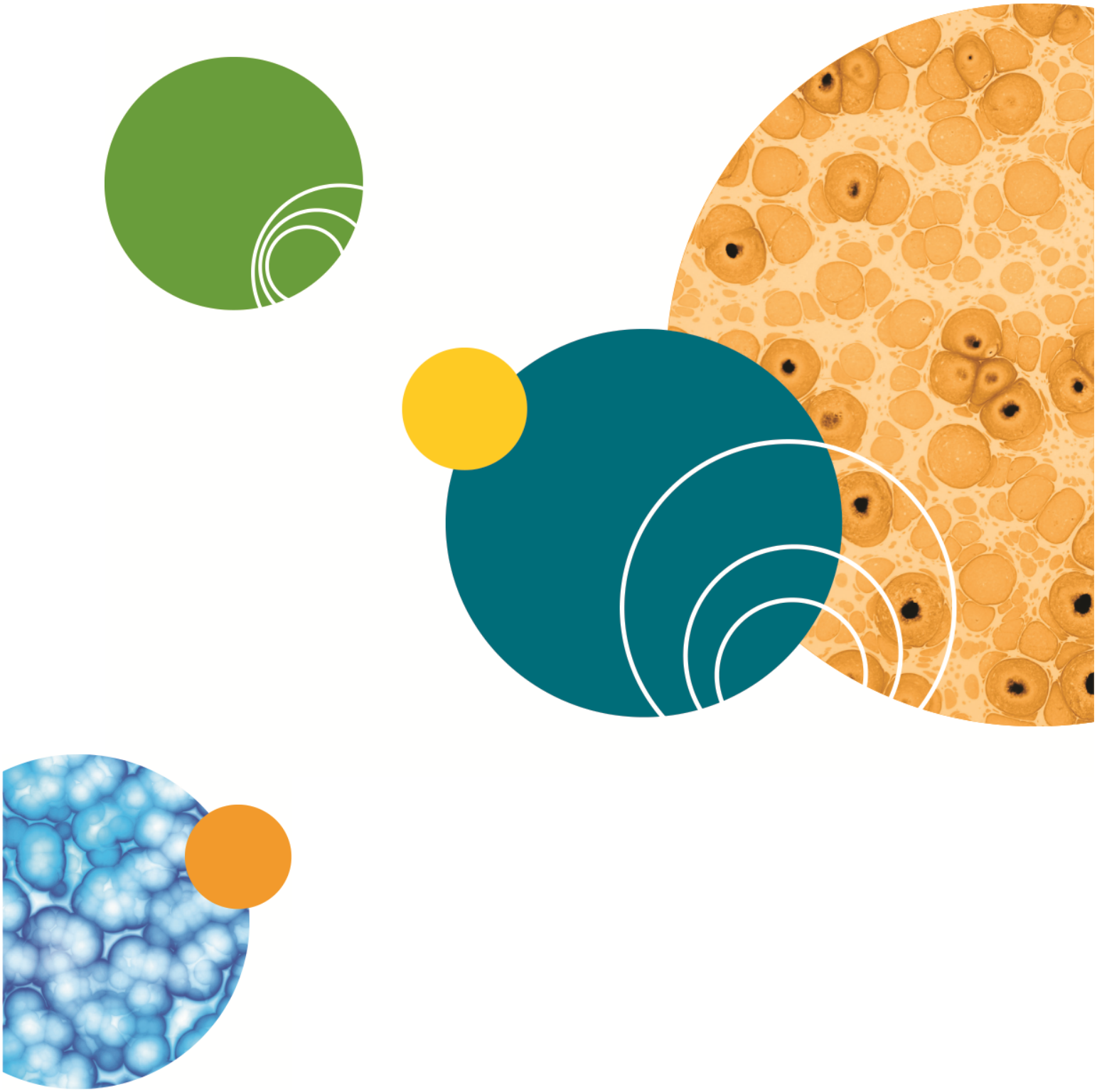
After you install a new filter cube, you may need to calibrate it. **Molecular Devices precalibrates the filter cubes included with the initial purchase of the instrument. You must calibrate any filter cubes purchased after that time.**

A calibration kit, which ships with any after-sales filter cube purchase, includes the following items:

- Slide holder
- Pink plastic slide
- Red plastic slide
- Blank glass slide
- Bead slide

To calibrate a filter cube:

1. On the **Devices** page, click  **Show Device Options** to expand the details for the device where you want to install an objective.
2. Click the **Filters** tab.
3. In the tile for the filter cube you want to calibrate, click **Filter Cube Calibration**.
4. Follow the on-screen instructions in the wizard to complete the calibration.



Chapter 8: Import Mode

8

Use **Import** mode to download and install the MD Import/Export Service, which enables you to import experiment data from temporary storage and export raw data from experiments.

On the **Home** page, click the  **Import** tile to enter **Import** mode.

Note: Import mode functions are available for Windows computers only.

Downloading the Import/Export Service

The MD Import/Export Service is required to import experiment data from temporary storage and export raw data from experiments. When the service is installed, the  **MD Import/Export Service** icon appears in the system tray at the bottom right of the screen.

Note: The MD Import/Export Service is available for Windows computers only.

To download the MD Import/Export Service:

1. On the **Import** page, click **Download Import/Export Service**.
2. After the download completes, navigate to your **Downloads** folder (if needed) and run **Imex.exe**.
3. Click **Install**.
4. If a Windows Security Alert appears noting that "Windows Firewall has blocked some features of this app", do the following:
   a. Select the **Domain Networks** checkbox.
   b. Select the **Private Networks** checkbox.
   c. Deselect the **Public Networks** checkbox.
   d. Click **Allow Access**.
5. Click **Finish**.

Exporting Raw Images

You can export raw images from an experiment.

Note: If you export raw images of different wells or regions from the same experiment to the same folder, the newly exported raw images are appended to the previously exported images.

If you export raw images of the same wells or regions from the same experiment to the same folder, the newly exported raw images overwrite the previously exported images.

To export raw images:

1. In the **Export Experiment** dialog, browse to select the folder to store the raw images.
2. Click **Export**.

The **Import/Export Monitor** page opens to display the progress of the import. The **Import/Export Monitor** page is similar to the **Monitor** page. See Monitor Mode on page 137 for details.

Importing Experiment Data from External Temporary Storage

If an experiment fails due to an issue with the CellReporterXpress Software or your network, the instrument may continue performing acquisition. In this case, the status light remains yellow. If you are using external temporary storage, you may be able to import the data for the failed experiment into the CellReporterXpress Software.

Note: If you are using internal temporary storage, images from a failed experiment cannot be imported.

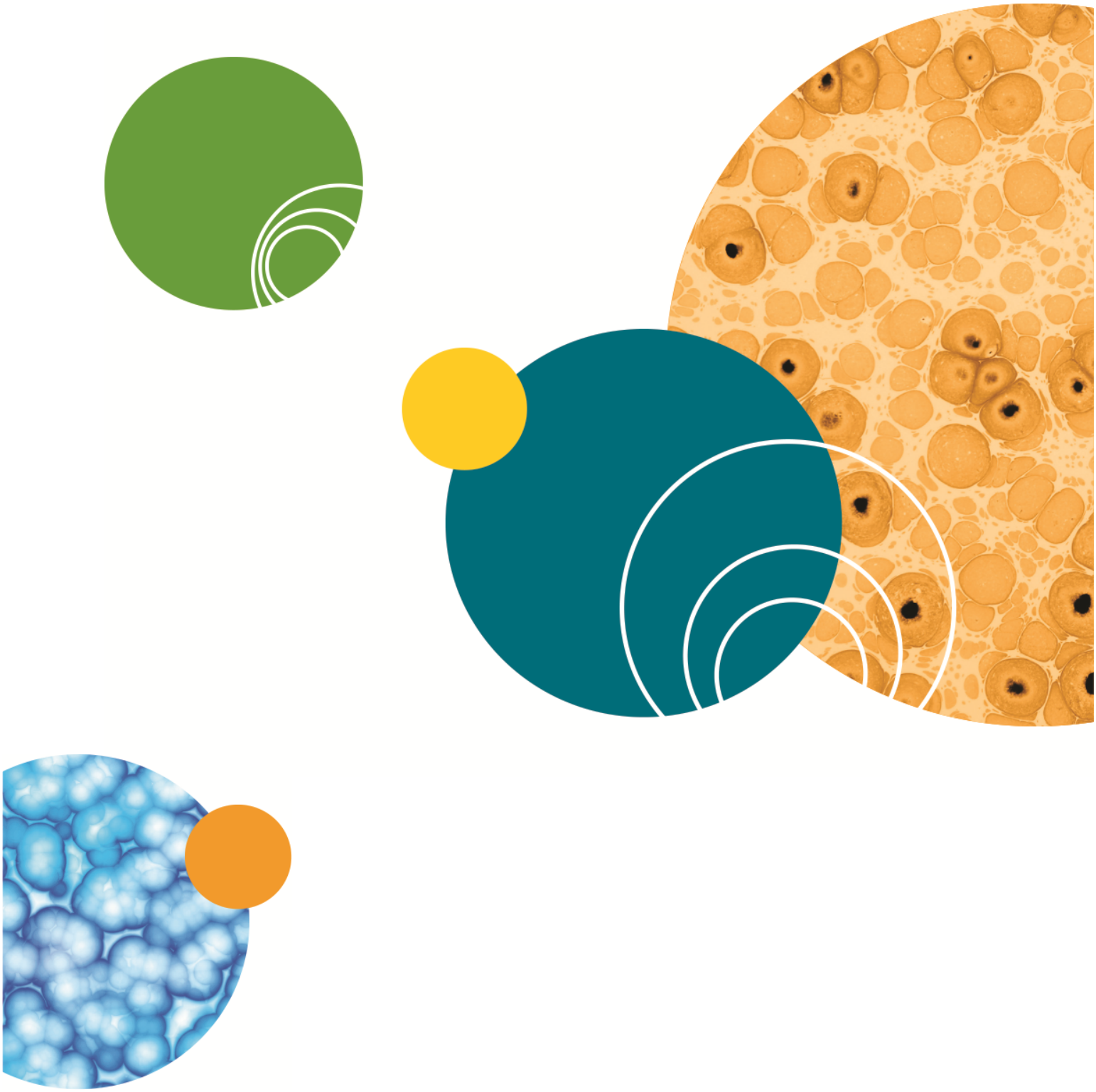
To import experiment data from external temporary storage:

1. On the **Import** page, click **Import New Experiment**.
2. In the **Select Experiment to Import** dialog, in the **File Explorer** section, browse to select a folder that contains an experiment.
3. In the **Choose Experiment** section, click **Scan**.

   Note: You can scan a folder at any level. You can even scan a logical drive. However, scanning a large drive can take a long time.

4. Select the experiment you want to import.
5. Click **Next**.
6. In the **Enter Experiment Details** dialog, enter or edit the experiment details as needed.
7. Click **Next**.
8. Select the storage location for the imported experiment data.
9. Click **Import**.

The **Import/Export Monitor** page opens to display the progress of the import. The **Import/Export Monitor** page is similar to the **Monitor** page. See Monitor Mode on page 137 for details.



Chapter 9: Configuration Settings

9

Use **Configuration Settings** to customize the CellReporterXpress Software interface for your login only.

In the toolbar, click  **View Preferences** to display the **Configuration Settings** page.

The left side of the page includes the following icons:

 **Themes**: Specifies the color scheme for the CellReporterXpress user interface. See Themes on page 167 for details.

 **Vocabulary**: Specifies the language for the CellReporterXpress user interface. See Vocabulary on page 168 for details.

 **Color Scheme**: Specifies the color scheme used for heatmaps in **Experiments** mode. See Color Scheme on page 168 for details.

 **Stains**: Specifies the stain definitions available in the CellReporterXpress Software. You can edit certain details of the stain definitions. See Stains on page 169 for details.

 **Sharing Permissions**: Specifies the default sharing permissions for protocols and experiments. You can modify this default setting for each protocol and experiment to set permissions individually. See Sharing Permissions on page 170 for details.

 **Miscellaneous**: Specifies various CellReporterXpress Software preferences, including image preferences and timeouts. See Miscellaneous on page 171 for details.

Themes

The  **Themes** settings specify the color scheme for the CellReporterXpress user interface. Your setting affects your login only.

Vocabulary

The **Vocabulary** settings specify the language for the CellReporterXpress user interface. Your setting affects your login only.

English Technical is the only vocabulary setting available with the current version of the CellReporterXpress Software.

Color Scheme

The **Color Scheme** settings specify the color scheme used for heatmaps in **Experiments** mode. Your setting affects your login only.

Stains

The **Stains** settings specify the stain definitions available in the CellReporterXpress Software. You can edit certain details of the stain definitions. Your edits affect your login only. All edited stain definitions display a **Restore Original** icon in the leftmost column.

Note: To add stain definitions to the library or edit the stain definition details of the stains available to all users, go to the **Stain Library** page in **Configuration** mode. See Stain Library on page 140 for details.

Editing a Stain Definition for Your Login

To edit a stain for your login:

1. On the **Stains** page, in the row for the stain you want to edit, click **Edit**.
2. Make the changes as needed.
3. In the **Edit** field, click **Apply**.

Restoring a Stain Definition to Its Systemwide Setting

To restore a stain definition to its systemwide setting:

On the **Stains** page, in the row for the stain you want to restore, click **Restore Original**.

Sharing Permissions

The  **Sharing Permissions** settings specify the default sharing permissions for protocols and experiments. You can modify this default setting for each protocol and experiment to set permissions individually. Your settings affect your login only. By default, sharing permissions for protocols and experiments are unlocked and unrestricted.

Sharing permissions function differently for protocols and experiments:

- A locked protocol can be viewed and run by all other users, but only specified users can modify it.
- A private experiment can only be viewed by specified users.

Setting Default Protocol Sharing Permissions

The default protocol sharing permission is unlocked.

To set your default protocol sharing permission to share protocols only with specific users:

1. On the **Sharing Permissions** page, under **Default Protocol Sharing Permissions**, click  **Unlocked**.
2. Do the following to assign users permissions to modify the protocol:
   a. Click in the **Share with** field.
   b. In the dropdown, click user names as needed to assign permissions.

Setting Default Experiment Sharing Permissions

The default experiment sharing permission is unlocked.

To set your default experiment sharing permission to share experiments only with specific users:

1. On the **Sharing Permissions** page, under **Default Experiment Sharing Permissions**, click  **Unlocked**.
2. Do the following to assign users permissions to view the experiment:
   a. Click in the **Share with** field.
   b. In the dropdown, click user names as needed to assign permissions.

Miscellaneous

The  **Miscellaneous** settings specify various CellReporterXpress Software preferences, including image preferences and timeouts. Your settings affect your login only.

The following settings are available:

- **Deep Zoom Images Preferences**: Specifies the image type (either **PNG** or **JPG**) for acquired images. If you select **JPG**, set the level of quality to be used. Higher quality means less compression and larger files, which affects the time required to open, redraw, and transfer acquired images.
- **Snap Image Preferences**: Specifies the image type (either **PNG** or **JPG**) and image resolution for preview images. If you select **JPG**, set the level of quality to be used. Higher quality means less compression and larger files, which affects the time required to open preview images.
- **Session Timeout**: Specifies the amount of time of inactivity before a session times out and logs off. Note that acquisitions (including time series acquisitions) continue after the session logs off. The default setting is 30 minutes.
- **Numeric Data Significant Figures**: Specifies the number of significant figures shown for measurements when measured analysis values appear. This setting also affects the number of significant digits saved when exporting measurements. The default setting is 6 figures.
- **Sensor Reading Decimal Places**: Not used in this version of the CellReporterXpress Software.
- **Storage Unit (byte) Decimal Places**: Specifies how many decimal digits appear in a data storage value. The default setting is 2 decimal digits.
- **Length Unit (mm) Decimal Places**: Specifies how many decimal digits appear in a data storage value. The default setting is 2 decimal digits.
- **Reached Max. Timepoints**: Specifies the maximum number of time points in a single acquisition.

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