



CloneSelect™

Imager

Hardware and Software v1.6

User Guide

This document is provided to customers who have purchased Molecular Devices equipment, software, reagents, and consumables to use in the operation of such Molecular Devices equipment, software, reagents, and consumables. This document is copyright protected and any reproduction of this document, in whole or any part, is strictly prohibited, except as Molecular Devices may authorize in writing.

Software that may be described in this document is furnished under a non-transferrable license. It is against the law to copy, modify, or distribute the software on any medium, except as specifically allowed in the license agreement. Furthermore, the license agreement may prohibit the software from being disassembled, reverse engineered, or decompiled for any purpose.

Portions of this document may make reference to other manufacturers and/or their products, which may contain parts whose names are registered as trademarks and/or function as trademarks of their respective owners. Any such usage is intended only to designate those manufacturers' products as supplied by Molecular Devices for incorporation into its equipment and does not imply any right and/or license to use or permit others to use such manufacturers' and/or their product names as trademarks.

Each product is shipped with documentation stating specifications and other technical information. Molecular Devices products are warranted to meet the stated specifications. Molecular Devices makes no other warranties or representations express or implied, including but not limited to, the fitness of this product for any particular purpose and assumes no responsibility or contingent liability, including indirect or consequential damages, for any use to which the purchaser may put the equipment described herein, or for any adverse circumstances arising therefrom. The sole obligation of Molecular Devices and the customer's sole remedy are limited to repair or replacement of the product in the event that the product fails to do as warranted.

For research use only. Not for use in diagnostic procedures.

The trademarks mentioned herein are the property of Molecular Devices, LLC or their respective owners. These trademarks may not be used in any type of promotion or advertising without the prior written permission of Molecular Devices, LLC.

Patents: <http://www.moleculardevices.com/patents>

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.
©2018 Molecular Devices, LLC.
All rights reserved.



Contents

Safety Information	5
Warnings, Cautions, Notes, and Tips	5
Symbols on Instrument Labels	6
Before Operating the Instrument	7
Electrical Safety	7
Chemical and Biological Safety	8
Moving Parts Safety	8
Cleaning and Maintenance Safety	9
Chapter 1: Introduction	11
Workflows	11
Before You Start	13
Powering On the System	13
Powering Off the System and the Software	13
Chapter 2: Instrument Overview	15
Status Lights	15
Microplate Holder	16
Leveling Feet	16
Setup	16
Using Barcodes	18
Chapter 3: Software Overview	19
Menu Options	20
Navigation Screen	21
Process Properties Screen	24
Chapter 4: Running Processes	25
Image Microplate	25
Review Results	40
Batch Microplate Imaging	84
E-Scope	85
Eject Plate Carrier	88

Chapter 5: Health and Safety	89
Chapter 6: Maintenance	91
Doing Preventive Maintenance	91
Cleaning the Instrument	92
Upgrading Software	92
Chapter 7: Troubleshooting	99
Troubleshooting Barcodes	99
Troubleshooting Well Detection Area	100
Viewing and Generating an Error Report	101
Obtaining Support	101
Appendix A: Technical Specifications	103
Instrument Dimensions	104
Appendix B: Compatible Plate Types	105
Appendix C: Well List File Format	107
Appendix D: Setting up Cell Number Estimation	109
Appendix E: CloneSelect Imager Data Viewer Software	111
Software Prerequisites	111
Installing CloneSelect Imager Data Viewer Software	112
Licensing CloneSelect Imager Data Viewer Software	112
Using CloneSelect Imager Data Viewer Software	112

Safety Information

Information about the safe use of the instrument includes an understanding of the user attention statements in this guide, the safety labels on the instrument, precautions to follow before you operate the instrument, and precautions to follow while you operate the instrument.

Read and observe all warnings, cautions, and instructions. The most important key to safety is to operate the instrument with care.



WARNING! If the instrument is used in a manner not specified by Molecular Devices, the protection provided by the equipment might be impaired.

Warnings, Cautions, Notes, and Tips

All warning symbols are framed within a yellow triangle. An exclamation mark is used for most warnings. Other symbols can warn of other types of hazards such as biohazard, electrical, or laser safety warnings as are described in the text of the warning. Follow the related safety information.

The following user attention statements can display in the text of Molecular Devices user documentation. Each statement implies the amount of observation or recommended procedure.



WARNING! A warning indicates a situation or operation that could cause personal injury if precautions are not followed.



CAUTION! A caution indicates a situation or operation that could cause damage to the instrument or loss of data if correct procedures are not followed.








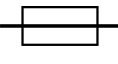



Note: A note calls attention to significant information.






Tip: A tip provides useful information or a shortcut, but is not essential to the completion of a procedure.

Symbols on Instrument Labels

Each safety label found on the instrument contains an alert symbol that indicates the type of potential safety hazard related to the label. The following table lists the alert symbols that can be found on Molecular Devices instruments.

Symbol	Indication
	Indicates a warning for a situation or operation that could cause personal injury if precautions are not followed. There are specific details written next to the warning symbol.
	Indicates a potential electrical-shock hazard from a high-voltage source. All safety instructions must be read and understood before proceeding with the installation, maintenance, and servicing of all modules. Power off the instrument and disconnect the power cord before you do maintenance procedures that require removal of a panel or cover or disassembly of an interior instrument component.
	Indicates a potential pinch hazard.
	Indicates power on. See Powering On the System on page 13 .
	Indicates power off. See Powering Off the System and the Software on page 13 .
	Indicates the location of a fuse.
	Indicates European technology conformity.
	Indicates certification by TUV Rheinland for compliance with US and Canadian product safety standards.
	Indicates compliance with TUV Rheinland product safety for Germany.

Symbol	Indication
	Indicates compliance with Chinese RoHS Pollution Control Requirements.
	Indicates compliance with the Waste Electrical and Electronic Equipment (WEEE) Directive of the European Union. You must not discard this electrical or electronic product or its components in domestic household waste or in the municipal waste collection system. For products under the requirement of the WEEE directive, contact your dealer or local Molecular Devices office for the procedures to facilitate the proper collection, treatment, recovery, recycling, and safe disposal of the device.
	Indicates the instrument manufacturer.

Before Operating the Instrument

Make sure that everyone involved with the operation of the instrument has:

- Received instruction in general safety practices for laboratories.
- Received instruction in specific safety practices for the instrument.
- Read and understood all Safety Data Sheets (SDS) for all materials being used.

Electrical Safety

To prevent electrically related injuries and property damage, inspect all electrical equipment before use and immediately report all electrical deficiencies. Contact Molecular Devices Technical Support to service equipment that requires the removal of covers or panels.

The instrument must be connected to a properly grounded power outlet to protect from the risk of electric shock. The main chassis of the instrument is grounded together with all related electrical components.

In the event of a liquid spillage into the main cavity of the instrument, disconnect the mains power supply before trying to clean up.

Do not remove the fixed covers, as there are no user-serviceable parts inside. All electrical work must be referred to Molecular Devices approved service personnel.

If the external covers on the instrument are removed, the power supply does not automatically stop.



WARNING! HIGH VOLTAGE Power off the instrument and disconnect the power cord before you do maintenance procedures that require removal of a panel or cover or disassembly of an interior instrument component.

Do not try to use the instrument until all covers are replaced.

To provide access for disconnecting power from the instrument, maintain a 66 cm (26 in.) minimum clearance area on the right side of the instrument.

To protect against fire hazard, replace the fuses only with the same type and rating as the original factory-installed fuses.

Chemical and Biological Safety

Normal operation of the instrument can involve the use of materials that are toxic, flammable, or otherwise biologically harmful. When you use such materials, observe the following precautions:

- Handle infectious samples based on good laboratory procedures and methods to prevent the spread of disease.
- Observe all cautionary information printed on the original containers of solutions before their use.
- Dispose of all waste solutions based on the waste disposal procedures of your facility.
- Operate the instrument in accordance with the instructions outlined in this guide, and take all the required precautions when using pathological, toxic, or radioactive materials.
- Splashing of liquids can occur. Take applicable safety precautions, such as using safety glasses and wearing protective clothing, when working with potentially hazardous liquids.
- Observe the applicable cautionary procedures as defined by your safety officer when using hazardous materials, flammable solvents, toxic, pathological, or radioactive materials in or near a powered-up instrument.



WARNING! Never use the instrument in an environment where potentially damaging liquids or gases are present.

Moving Parts Safety

The instrument contains moving parts that can cause injury. Under normal conditions, the instrument is designed to protect you from these moving parts.

To prevent injury:

- Never try to exchange labware, reagents, or tools while the instrument is operating.
- Never try to physically restrict the moving components of the instrument.



WARNING! Do not attempt to access the interior of the instrument unless specifically instructed to do so. The moving parts inside the instrument can cause injury. Do not operate the instrument with any covers or panels removed.



Note: Observe all warnings and cautions listed for all external devices attached to or in use during the operation of the instrument. See the applicable user guide for the operating and safety procedures of that device.

Cleaning and Maintenance Safety

Observe the cleaning procedures outlined in this guide for the instrument.

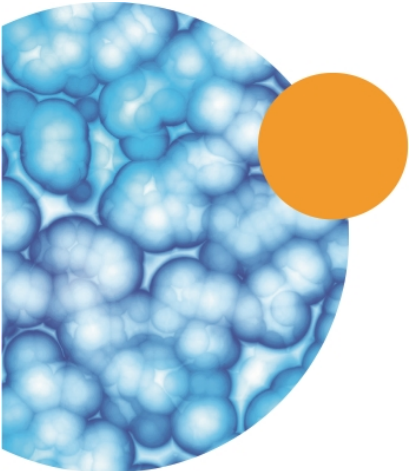
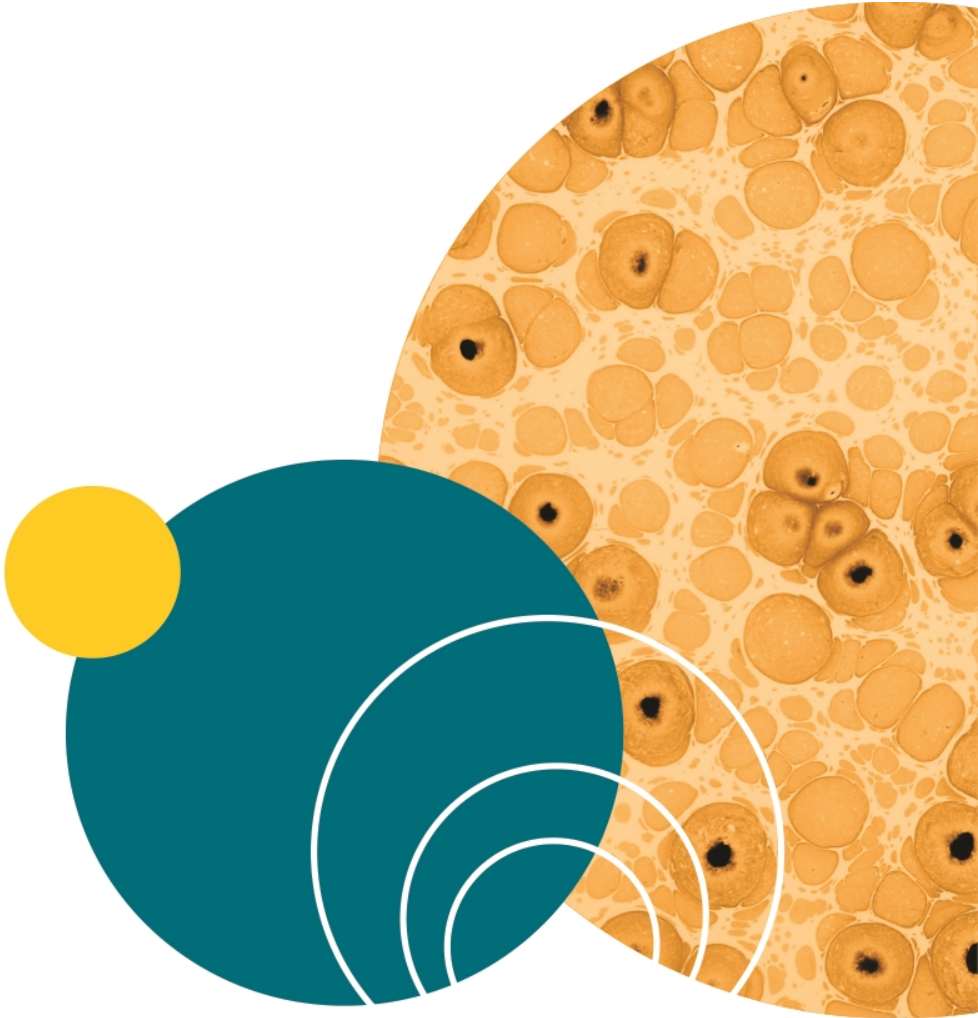
Do the following before you clean equipment that has been exposed to hazardous material:

- Contact the applicable Chemical and Biological Safety personnel.
- Review the Chemical and Biological Safety information contained in this guide.

Perform only the maintenance tasks described in this guide. Contact a Molecular Devices service engineer to inspect and perform a preventive maintenance service on the instrument each year.



WARNING! BIOHAZARD. It is your responsibility to decontaminate components of the instrument before you return parts to Molecular Devices for repair. Molecular Devices does not accept items that have not been decontaminated where it is applicable to do so. If parts are returned, they must be enclosed in a sealed plastic bag stating that the contents are safe to handle and are not contaminated.



The CloneSelect™ Imager system is an automated CCD camera-based system for imaging and confluence determination of cells growing in microplates.



Note: The CloneSelect Imager system is strictly for research use only and is not intended or recommended for the diagnosis of disease in humans or animals.



WARNING! If the instrument is used in a manner not specified in this guide, the protections provided by the equipment can be impaired.

Workflows

The CloneSelect Imager system uses the following workflow processes:

- [Imaging](#)
- [Analysis on page 12](#)
- [Reporting on page 12](#)
- [Verifying Monoclonality on page 12](#)

Imaging

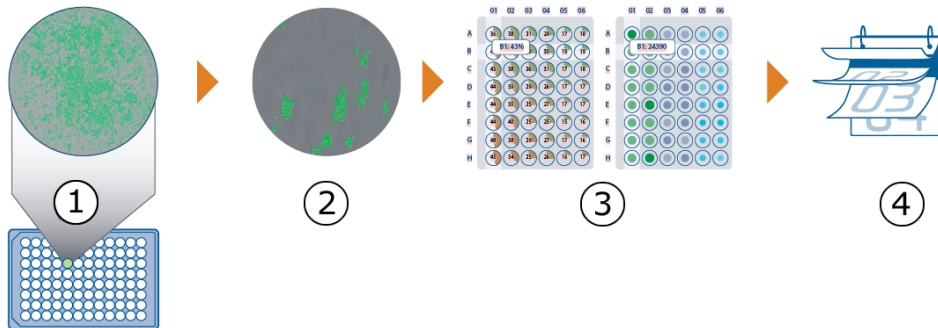
Use adherent or settled suspension cells in a microwell plate.



Figure 1-1: 96-well microplate and white light image

Analysis

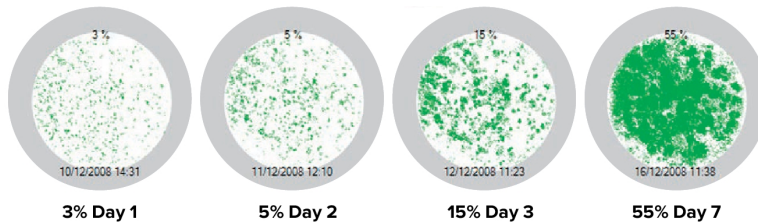
Display cell confluence and cell number estimation for each well and calculate growth curves.



Item	Description
1	Well selection
2	Cell distribution highlighted by software overlay
3	Cell confluence and cell number estimation for each well
4	Repeat over several days

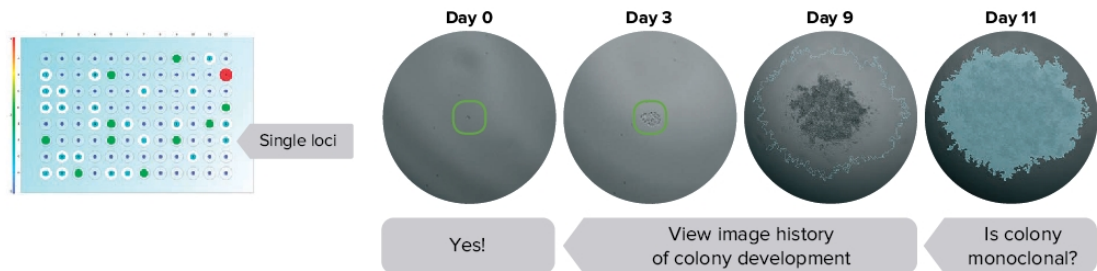
Reporting

Track and view growth of every cell line, and store plate data, including cell confluence, cell number estimation, and growth curve.



Verifying Monoclonality

After initial seeding, image every well, at any time point, using the loci of growth functionality to highlight those wells that contain a single colony. See [Loci Count on page 52](#) and [Monoclonality Workflow on page 57](#).



Before You Start



CAUTION! Before using the instrument, it is very important that you read this guide and understand all the safety instructions. Then follow the procedures in the [Powering On the System on page 13](#).

Powering On the System

Before continuing, do as instructed in [Before You Start on page 13](#).

To power on the CloneSelect Imager:

1. Turn on the CloneSelect Imager. See [Status Lights on page 15](#).
2. Wait for the Ready light to illuminate.
3. To warm up the instrument, leave the CloneSelect Imager in the **Ready** state for more than 5 minutes. When the unit is cold to the touch, wait for 30 minutes.
4. If the computer is powered off, power on the computer.
5. Double-click the desktop icon to start the CloneSelect Imager Software.



CAUTION! Do not start the software before the instrument is powered on and warmed up.

Rebooting the CloneSelect Imager

If you have to reboot the instrument:

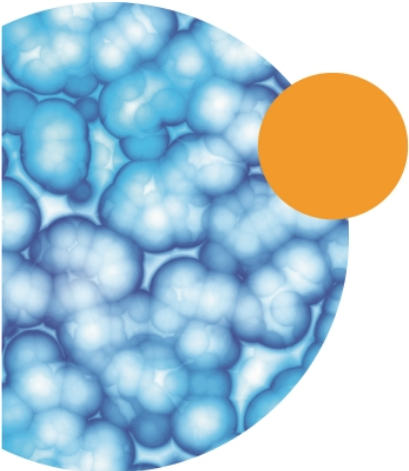
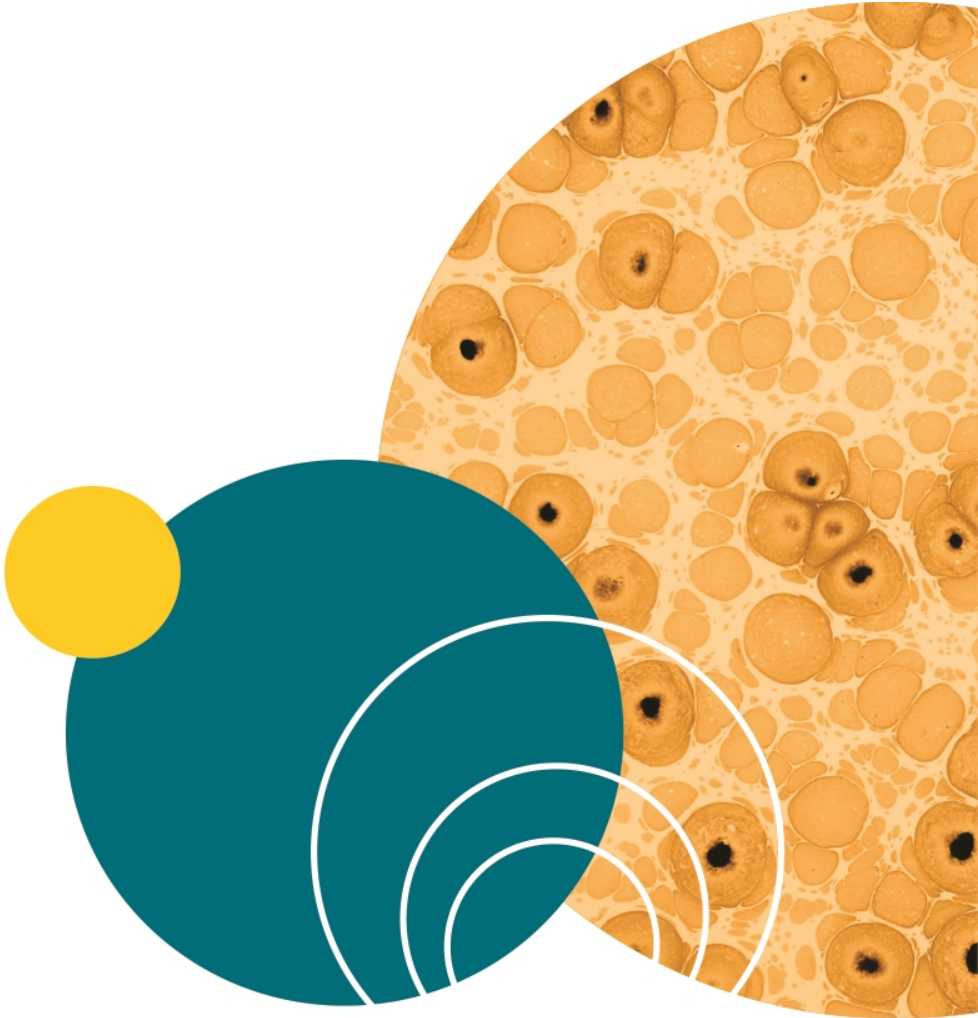
1. Power off the instrument. See [Powering Off the System and the Software](#).
2. Wait for 30 seconds.
3. Power on the instrument. See [Powering On the System on page 13](#).

When the **Ready** light illuminates, the instrument is ready to use.

Powering Off the System and the Software

To power off the CloneSelect Imager:

1. To exit the CloneSelect Imager Software, on the **Main Navigation** screen, select **File > Exit**.
2. To shutdown your computer, click **Start > Shut down**, and then wait for computer to power off completely.
3. To power off the instrument, on the left side of the instrument, toggle the power switch to off.



Chapter 2: Instrument Overview

2

The CloneSelect Imager system is used to image every well to count colony number, estimate colony area and track colony growth.

Before operating the instrument or doing maintenance operations, make sure that you are familiar with the safety information in this guide. See [Safety Information on page 5](#).



Table 2-1: CloneSelect Imager Overview

Item	Description
1	Power Connection Port and Power Switch
2	Status Indicator Lights
3	Microplate Holder

For technical specifications, see [Technical Specifications on page 103](#).

Status Lights

The status lights on the front of the instrument indicate the instrument activity.



Microplate Holder

The microplate holder consists of a glass plate surrounded by a vacuum bed seal. The microplate is placed on the microplate holder so that the skirt is over the vacuum bed seal. When the microplate holder retracts into the instrument for imaging, the microplate is pushed into the back right corner of the microplate holder into the correct position for imaging.

Before imaging, a vacuum is applied under the microplate that holds it down flat on the glass plate so that all the wells are in the same focal plane. This eliminates the need to autofocus on every well.



CAUTION! The plate skirt of the plate needs to be intact for the vacuum seal to work.

Leveling Feet



If needed, adjust the leveling feet by loosening the locking nuts and turning the feet until the instrument is level and stable. After you make adjustments, tighten the locking nuts.

Setup

Locating the CloneSelect Imager

Place the CloneSelect Imager on a stable, level surface. Level the instrument by loosening the locking nuts and turning the levelling feet to ensure that the CloneSelect Imager is stable and not able to move. After adjustment, tighten the locking nuts.

Computer



Note: Molecular Devices is responsible for installing the CloneSelect Imager Software during your initial instrument installation and configuration.

The CloneSelect Imager ships with a computer with special hardware and software to support the imaging function.



CAUTION! Do not attempt to use any other computer or change the operating system to operate the CloneSelect Imager.

The computer includes Windows 10. To connect to your network, security configuration is required. When that is done, ensure that no changes are made to the configuration of the private network connection to the CloneSelect Imager because that stops it operating.



CAUTION! Do not change the standard English settings. If these settings change, the performance of the system can be impacted.

For software upgrades, see [Upgrading Software on page 92](#).

Connecting the CloneSelect Imager

The connections to the CloneSelect Imager are on the connections panel on the left side of the instrument.

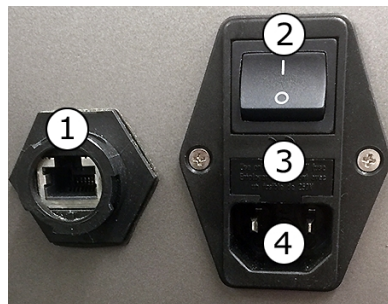


Table 2-2: Connections and Power Switch Panel

Item	Description
1	Ethernet port
2	On/Off switch
3	Fuse
4	Mains port

Connect the mains cables to the CloneSelect Imager, the computer and to the monitor before making the network connections.

The connection to the CloneSelect Imager is a private Gigabit Ethernet link and the connection must be made to the computer add-on card Gigabit Ethernet port.

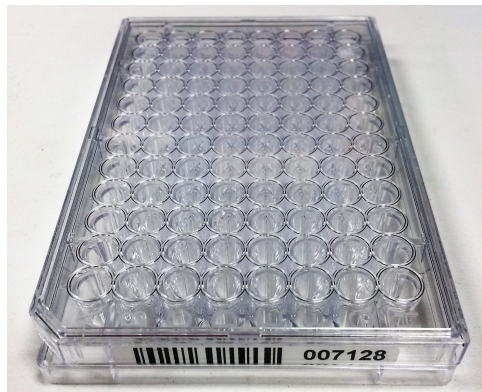
Using Barcodes

If you use barcodes, the following barcode parameters are required:

- Use a legible barcode of the following types: 2D barcodes with code 11, 39, 93 and 128



- Place the barcode consistently on one of the short sides of the plate.



For troubleshooting, see [Troubleshooting Barcodes on page 99](#).

The CloneSelect Imager Software controls the CloneSelect Imager instrument and is based around the concept of the instrument running processes. Using the default set of processes, you can modify and save them.

To start the software, from the Windows desktop, double-click the **CloneSelect Imager** icon.

When the software first starts, the Navigation screen displays the **New Process** tab (Figure 3-1). Processes are managed from the Navigation screen. Selecting a process takes you to the chosen process start-screen from where you can set the properties of the process and run it. See [Process Properties Screen](#) on page 24.

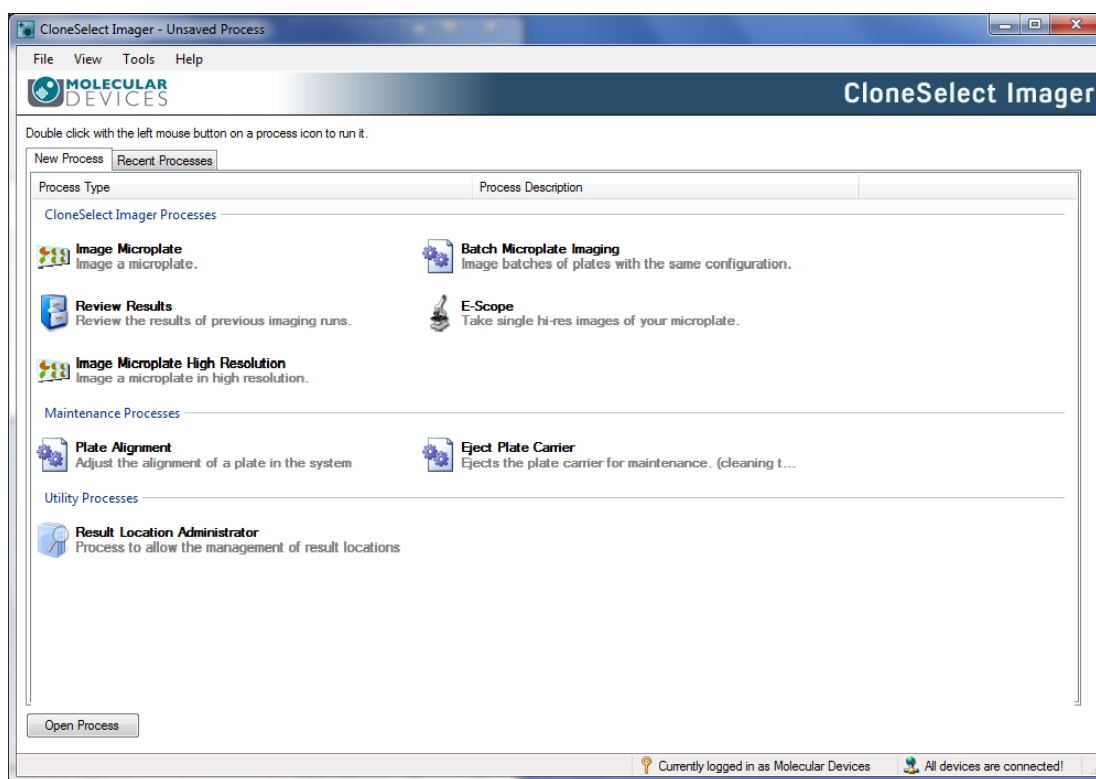


Figure 3-1: CloneSelect Imager Software Navigation Screen



Note: The options that appear on the start up screen depend on the features you purchased.

Menu Options

The available options in the menus at the top of the Navigation screen change depending on the screen view and selections.

File Menu

- **Open Process** allows you to open a previously saved process.
- **Save Process** saves the current process.
- **Save Process As** allows you to save the current process with a different name or location.
- **Close Process** closes the current process and returns you to the Process Selection Window.
- **Save As Template** saves the current process as a template that can be used for creating new processes.
- **Recent Processes** allows you to select and open a recent process.
- **Switch User** can be used in a multiple user environment to log off the current user so that a different user can log on.
- **Exit** closes the Navigation window and shuts down the software.

View Menu

- **Properties** displays the properties of the process that is being set before starting the process.
- **Progress** displays the progress of a running process.
- **Administrate Properties** lets you change the Properties view and default values.

Tools Menu

- **Configuration** displays the **Edit Configuration** dialog.



CAUTION! Changing the settings from this menu can cause the device to be inoperable. Proceed with caution and create a restore point before making any changes.

- **Prepare Error Report** starts the **Error Reporting Wizard** to create a data file containing the configuration and recent log files to help troubleshoot your problem.

Help

- **About** displays the version numbers of the software modules.
- **Online Support** displays the support web page. This option requires an active internet connection.
- **Help View** displays the online help file.

Navigation Screen

The Navigation screen is the CloneSelect Imager Software start-screen from which you select to run processes. Using the right-click option menu, you can view the process options as icons (Figure 3-2) or as a text list (Figure 3-3).



Note: Available processes will vary depending on your system configuration.

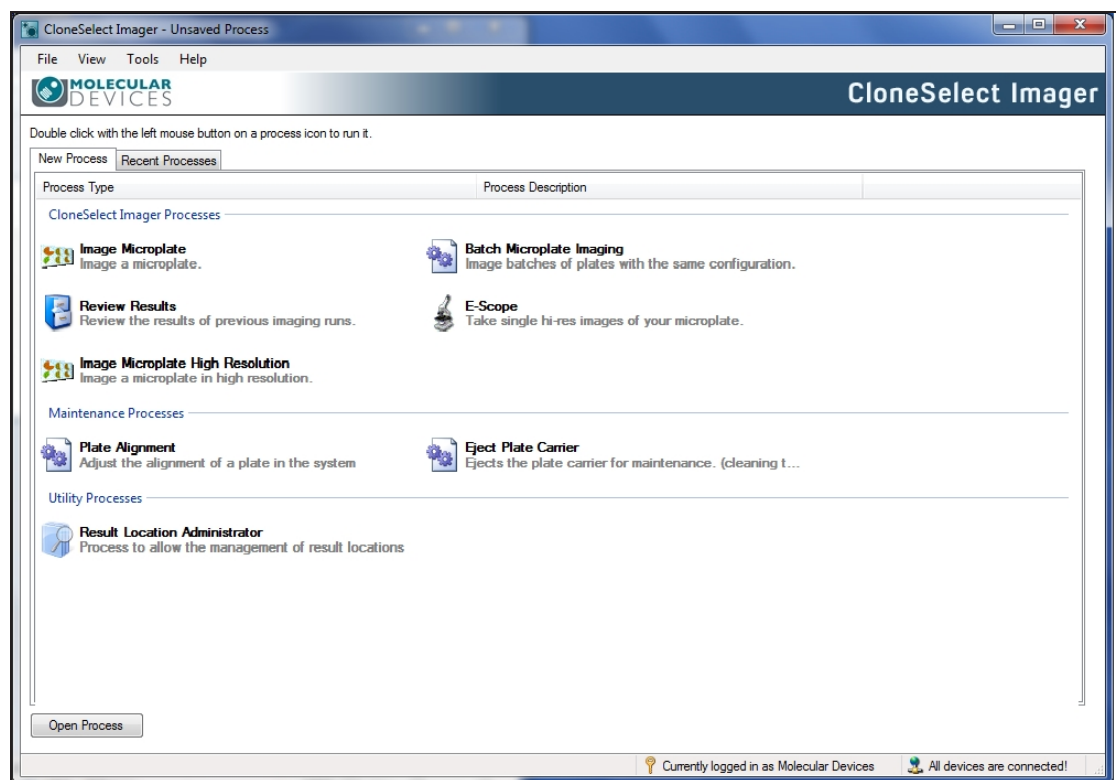


Figure 3-2: The Navigation screen with process options as icon view

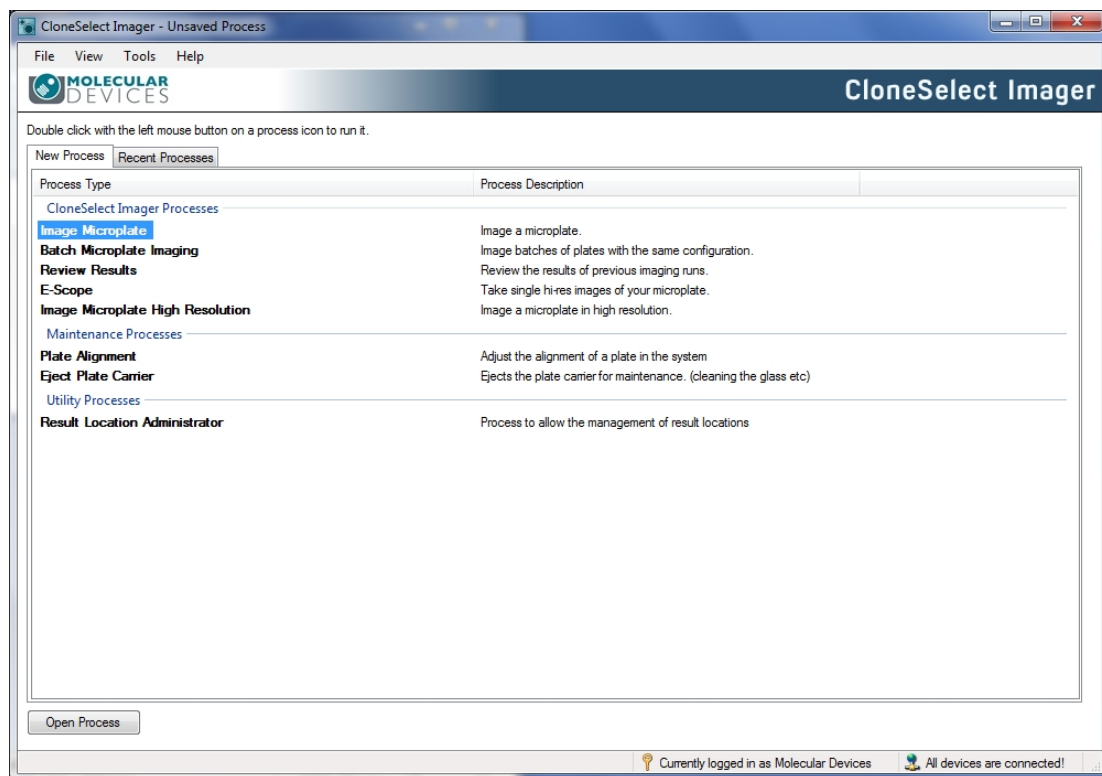


Figure 3-3: The Navigation screen with process options as text list view

The Navigation screen includes the following functionality:

- **Open Process** button displays the **Open** dialog where you can select and open a saved process file. Can also be opened using the **File > Open Process** menu option.
- **Connection Status** appears in the bottom right corner of the Navigation screen and displays the status of the Ethernet connection between the computer and the CloneSelect Imager.
- **New Process Tab** displays the available processes, which are grouped into the following categories:
 - **CloneSelect Imager Processes**
 - **Maintenance Processes**
 - **Utility Processes**
 - (Optional) **Automation Process**—Available only if optionally purchased to use to integrate the CloneSelect Imager with other devices, such as automated incubators or robotic arms.

See [New Process Options on page 23](#).

- **Recent Processes Tab** displays a list of recently opened processes. This is the same list of processes available from the **File > Recent Processes** menu option.

New Process Options

The **New Process** tab of the Navigation screen gives you a full set of options for creating and managing processes and system maintenance.

CloneSelect Imager Processes

CloneSelect Imager Processes include the following:

- **Image Microplate** does standard imaging of microplates, saving the images and results for later review.
- **Batch Microplate Imaging** does imaging of batches of plates with the same configuration without reviewing the focus and alignment between plates.
- **Review Results** displays results from previous runs including confluence, cell number estimation, and monoclonality analysis for reviewing.
- **E-Scope** does single high-resolution image of microplates. Images are not saved.
- (Optional) **Image Microplate High Resolution** does high-resolution imaging of microplates, saving the images and results for later review.



Note: The **Image Microplate High Resolution** option also includes a camera upgrade. For more information, contact Molecular Devices.

Maintenance Processes

Maintenance Processes include the following:

- **Plate Alignment** runs the procedures for calibrating the instrument.
- **Eject Plate Carrier** ejects the plate carrier for cleaning and removal of microplates.

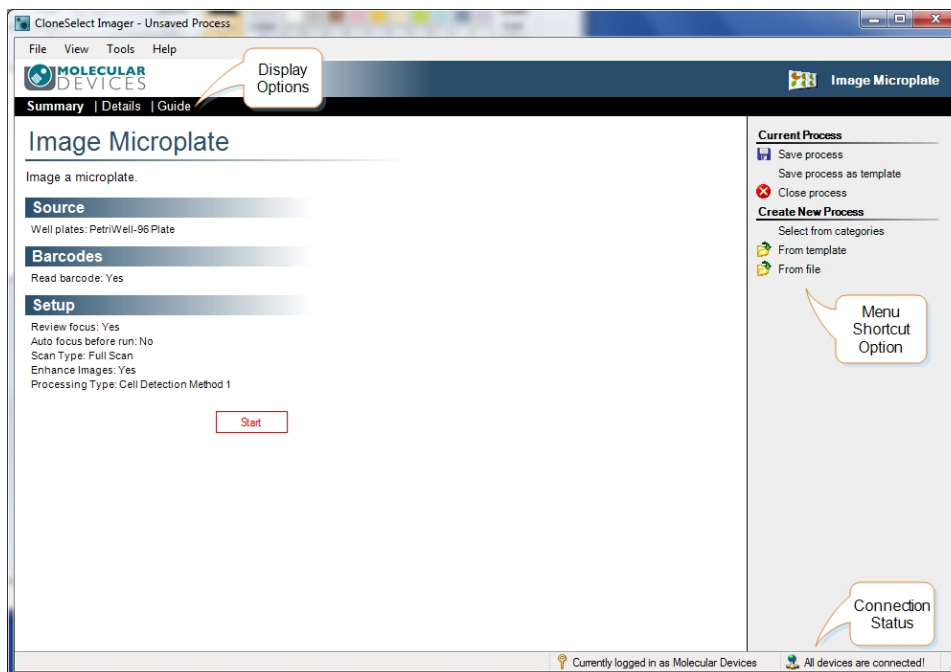
Utility Processes

Utility Processes include the following:

- **Result Location Administrator** lets you manage where the results get located.

Process Properties Screen

The process starts and ends on this **Summary** screen. Process properties can be edited using either of the three display options.



Display Options

There are three options for selecting the way in which the process settings can appear for editing:

- **Summary** is the default display option. The current settings for the process appear grouped in categories. Click on the category heading to open a screen where the relevant settings can be changed using drop-down menus.
- **Details** offers the current process settings displayed in drop-down menus from which they can be edited directly.
- **Guide** steps you through the setup process like a wizard with **Back** and **Next** buttons to scroll through the settings. When all the settings are done, you return to the **Summary** view.

Shortcuts

On the right side of the Process Properties screen there are shortcuts to frequently used menu options.

Connection Status

The status of the Ethernet connection between the computer and the CloneSelect Imager appears in the bottom right corner of the Process screen.

Chapter 4: Running Processes

This section explains in detail how to run the following processes on the CloneSelect Imager.

- [Image Microplate](#), see page 25
- [E-Scope](#), see page 85
- [Review Results](#), see page 40
- [Eject Plate Carrier](#) on page 88

Image Microplate

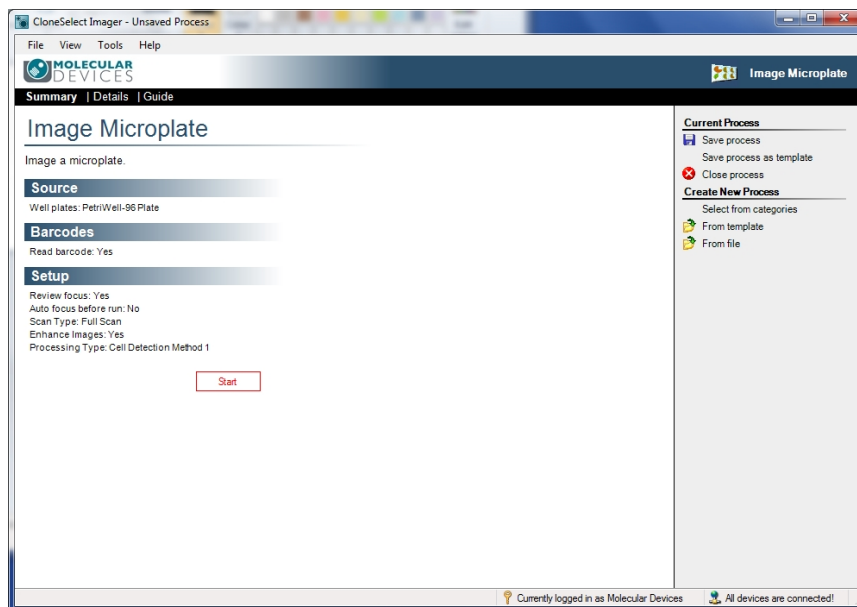
Image Microplate is the default process for imaging cells in microplates to determine their confluence and carry out further analysis, such as cell number estimation and loci counting. Image resolution in this process is 3.5 μm .

To enter an Image Microplate process, in the Navigation screen, click **Image Microplate**. See [Navigation Screen on page 21](#). Alternatively, you can open a previously saved Image Microplate-based process from the **Recent Processes** tab.

To set the properties for the Image Microplate process, the process screen opens in the Process Properties screen in the **Summary** view. See [Process Properties Screen on page 24](#). Change the screen view, as needed, and change properties as needed.

Alternatively, to create your new process, you can use the shortcuts menu on the right side of the screen to do any of the following **Create New Process** start tasks:

- **From template**—Starts the new process from a template file.
- **From file**—Starts the new process from a previously saved file.



The Image Microplate process involves the following tasks:

1. [Set Properties](#), see page 26
2. [Select Source](#), see page 28
3. [Load Plate](#), see page 29
4. [Focus](#), see page 30
5. [Alignment](#), see page 32
6. [Imaging](#), see page 33
7. [Results](#), see page 36
8. [Remove Plate](#), see page 38
9. [Finish](#), see page 39

Set Properties

Set the properties for the Image Microplate process you want to run.

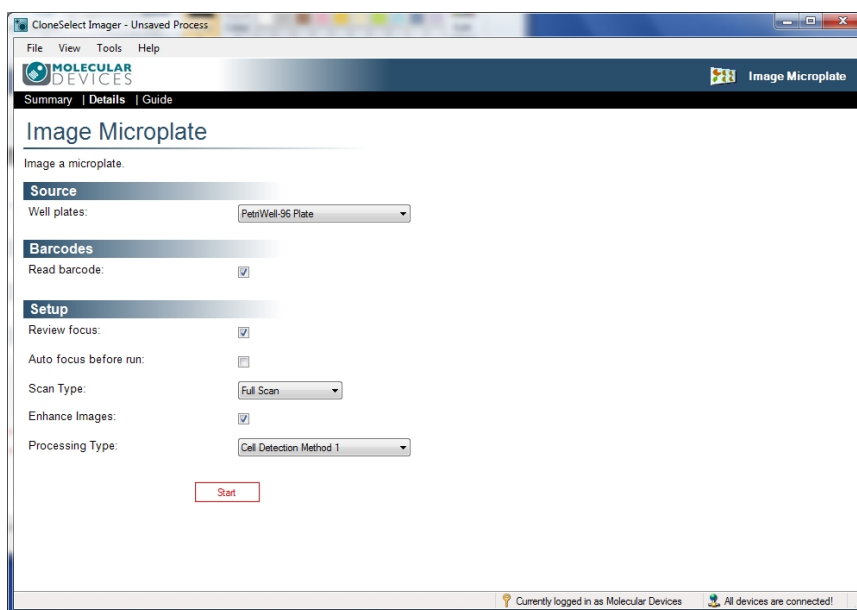


Figure 4-1: Process properties Details view

To set process properties, do the following:

1. On the main screen, select **Details**.
2. In the **Source** section, for **Well plates**, select your microplate from the list menu.



Note: To image a plate type that does not appear in the list, contact Molecular Devices. See [Obtaining Support](#) on page 101.

3. If you are using barcodes, in the **Barcodes** section, select the **Read barcode** check box.

*** Tip:** If barcode reading is enabled and the instrument fails to read the barcode, a dialog appears to let you manually enter the barcode. Also, if needed, you can edit the barcode or name within **Review Results**. See [Review Results on page 40](#).

4. In the **Setup** section, specify the following:

- **Review focus**—When enabled, before imaging a microplate, allows you to check the focus and brightness settings. Opens the **Focus** dialog. See [Focus on page 30](#).

*** Tip:** You can also save individual images from the **Focus** dialog.

- **Auto focus before run**—When enabled, before imaging the microplate, an auto-focus procedure runs on the instrument.

*** Tip:** If you are working with a sample that does not have wells with high contrast cells included, leaving this function disabled.

- **Scan type**—Select one of the following scan methods from the list menu:

- **Full Scan**—Images the entire area of each well in the microplate. This setting is recommended for monoclonality applications.
- **Partial Scan**—Images an area from the center of each well.

*** Tip:** This speeds up imaging. Suitable when the data from the center of each well is representative of the whole well.

- **Enhance Images**—When enabled, to enhance images for display, the background is flattened. This setting is recommended for monoclonality applications.

- **Processing Type**—Select one of the following cell and colony detection processing types from the list menu:

- **Cell Detection Method 1**—Formally know as *Typical Adherent Cell Confluence*, this algorithm is ideal for settled suspension cells and adherent cells with good contrast.
- **Cell Detection Method 2**—This algorithm is ideal for detecting cell samples when contrast is low, cells are clumped, or cells are at the edge of the well.
- **Cell Detection Method 3**—This algorithm is ideal for detecting colonies and distinguishing them from any artifacts around the edges of the colonies.
- **Very Low Contrast**

5. After you set the process properties, to enter the process, click **Start**. See [Select Source on page 28](#).

Select Source

The first step of the Image Microplate process is called **Select Source**. The sources you select are microplate wells. Wells selected for imaging are marked red, and those not selected for imaging are marked light blue.

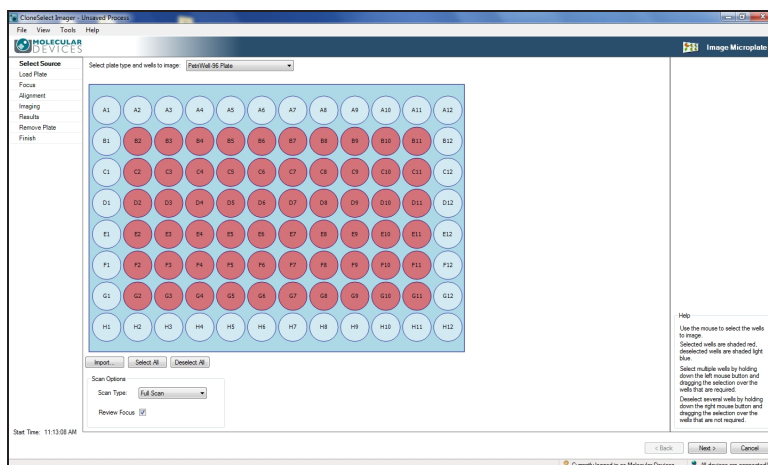


Figure 4-2: Select Source screen for selecting wells

To select source wells:

1. If you want to change the plate type, select your new microplate type from the list menu.
2. Select wells. By default, all of the wells are selected.
 - To select or deselect individual wells, left click on a well.
 - To select or deselect groups of wells, right-click and drag over the wells.
 - To deselect all wells, click **Deselect All**.
 - To select all wells, click **Select All**.
 - To select wells using a well list file, click **Import**, and then select your file containing your list of wells to image.

*** Tip:** For the format of the file, see [Well List File Format on page 107](#).

3. If you want to change the **Scan Options**:
 - In the **Scan Type** list menu, change your selection using the list menu.
 - If needed, change the **Review Focus** check box setting.
4. Do one of the following to continue:
 - To go to the next step, click **Next**. See [Load Plate on page 29](#).
 - To exit the process and return to the Image Microplate process properties screen, click **Cancel**. See [Set Properties on page 26](#).

Load Plate

The plate carrier automatically ejects and you are prompted to load the microplate.

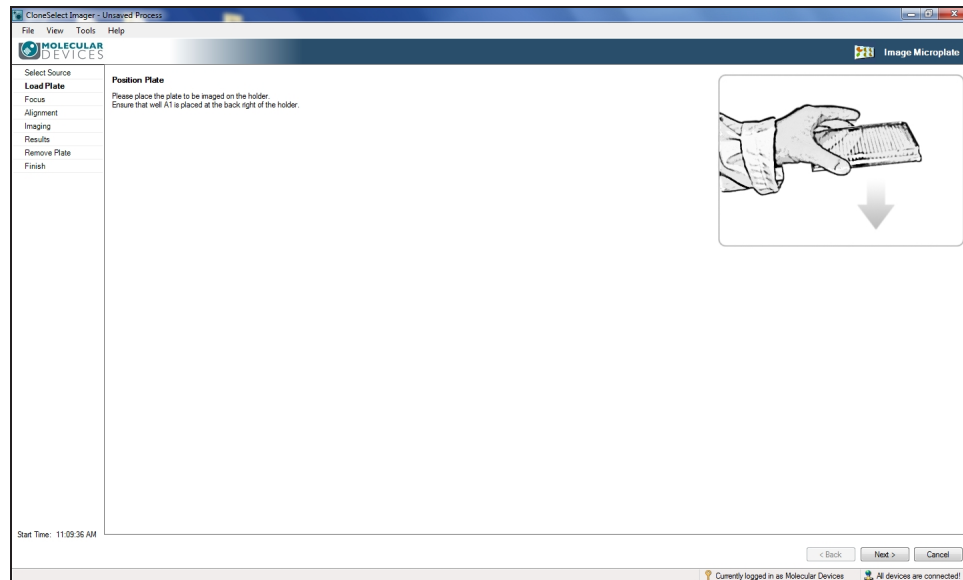


Figure 4-3: Load Plate screen

To load a microplate:

1. When the plate carrier ejects, place your microplate on the carrier with the A1 well in the back right corner.
2. Do one of the following to continue:
 - To go to the next step, click **Next**. See [Focus on page 30](#).
 - To exit the process and return to the Image Microplate process properties screen, click **Cancel**. See [Set Properties on page 26](#).

Focus

Use the **Focus** step to optimize the image view before the scan.

On this screen, you can do the following:

- Adjust Image Brightness on page 30
- Adjust Image Focus on page 31
- Save Image on page 31
- Change Processing Type on page 32
- Generate Confluence on page 32
- Continue to the Next Step on page 32

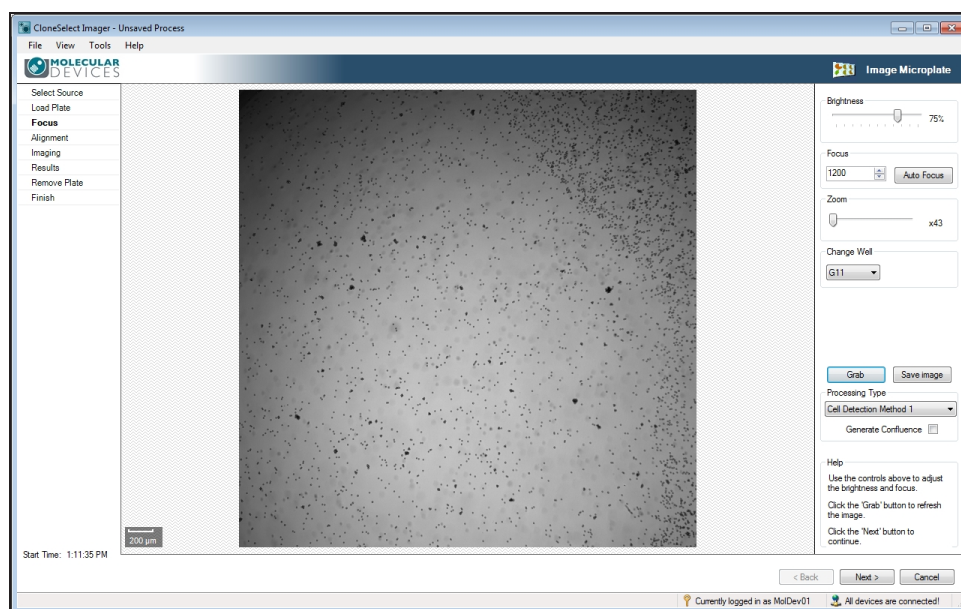


Figure 4-4: Focus screen

Adjust Image Brightness

The software automatically determines the optimal brightness setting for imaging, but you can manually adjust the **Brightness** setting.

To adjust image brightness:

1. Move the **Brightness** slider to the percentage value you want to use.
2. To update the image view using your new setting, click **Grab**.



Tip: Saturated pixels appear in red. Avoid making your image too bright.

3. Repeat steps 1 and 2 as needed.

Adjust Image Focus

The system stores the last focus setting used for each plate type. After imaging a plate type for the first time, the need for additional focus adjustments should be minimal.

Auto Focus

To adjust image focus:

1. Click **Auto Focus**.
2. To update the image view using your new focus setting, click **Grab**.
The changed focus setting becomes the default focus setting for the plate type in this process run.

*** Tip:** If there is debris on the bottom of the plate or on the glass plate carrier, the auto focus might focus on these instead of the cells, which requires manually focusing.

Manual Focus

To manually focus on the image:

1. In the **Focus** section of controls on the right side of the screen, click the up or down arrows to change the focus value, or type a new value in the field.
2. If needed, move the **Zoom** slider to look closer or farther at the image.
As you change the zoom level, the scale bar in the lower left of the image window automatically adjusts.

*** Tip:** When the zoom setting figure on the right of the slider is black, the system is zooming optically. When the figure turns red, the system is zooming digitally and you might see some pixelation of the image.

3. If needed, select a different well to focus on from the **Change Well** list menu.

*** Tip:** By default focusing is done on well A1, or if A1 is not being imaged, focusing is done on the first well of the plate to be imaged.

4. To update the image view using your new focus settings, click **Grab**.
The changed focus setting becomes the default focus setting for the plate type in this process run.

Save Image

Images can be saved in bmp, jpg or png format.

To save the current image used for focusing:

1. Click **Save Image**.
2. In the **Save File** dialog, type and name and click **Save**.

Change Processing Type

If needed, you can change the Processing Type you set in properties. See [Set Properties on page 26](#).

To change, in the **Processing Type** list menu, select a new cell and colony detection processing type.

Generate Confluence

When enabled, all of the objects within the well get identified and highlighted in green.

To enable, select the **Generate Confluence** check box.

Continue to the Next Step

To go to the next step, click **Next**. See [Alignment on page 32](#).

To exit the process and return to the Image Microplate process properties screen, click **Cancel**. See [Set Properties on page 26](#).

Alignment

Use the **Alignment** screen to check that the camera aligns properly with the wells of the microplate. Four images are taken over the north, south, east and west edges of well A1. The well edges should align with the middle of the box formed by the four red lines.

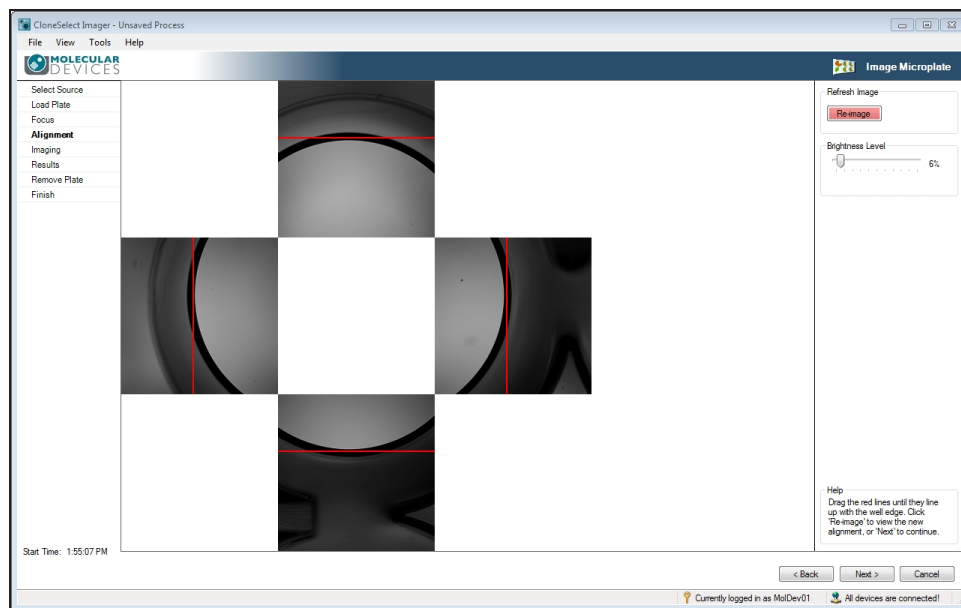


Figure 4-5: Alignment screen

To align the camera:

1. If the well edges do not fall in the middle of the box formed by the four red lines on screen, click and drag on the lines to correct the alignment.
2. If needed, you can also adjust the image brightness by moving the **Brightness** slider to the percentage value you want to use.



Note: Brightness changes only for the alignment image.

3. When the button is pink, click **Re-image** to recheck the alignment.

Continue to the Next Step

To go to the next step, click **Next**. See [Imaging on page 33](#).

To exit the process and return to the Image Microplate process properties screen, click **Cancel**. See [Set Properties on page 26](#).

Imaging

Imaging starts automatically.



Note: When the **Imaging** process finishes, you automatically go to the **Results** screen step.

The **Imaging** screen step includes the following tabs:

- [Confluence Tab on page 34](#)
- [View Image Tab on page 35](#)

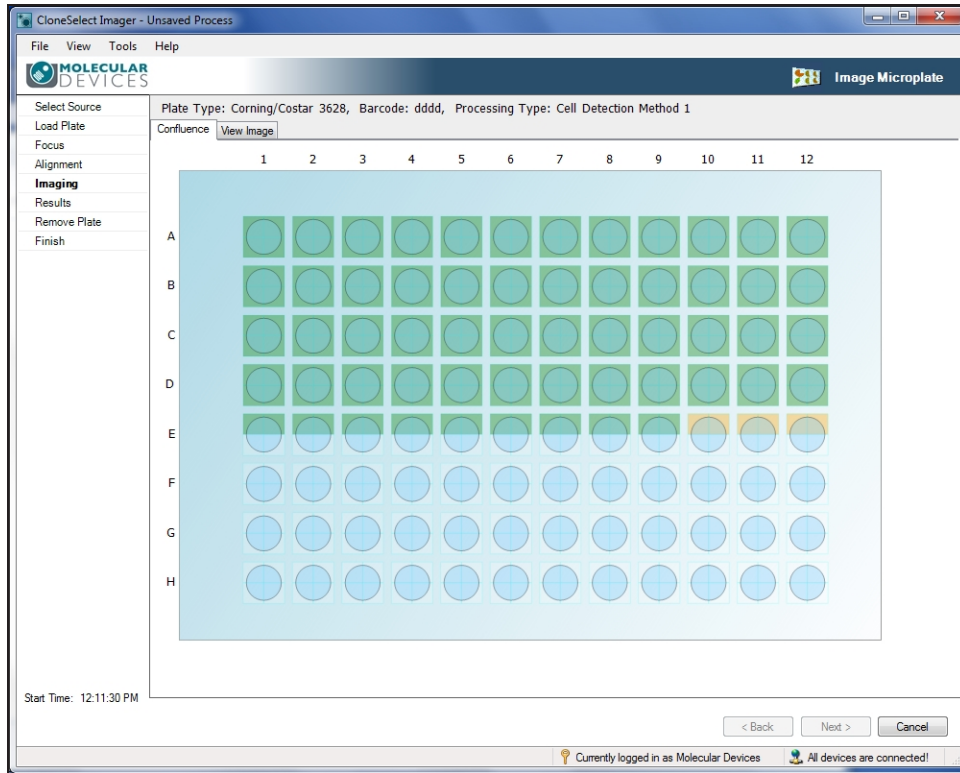


Figure 4-6: Imaging progress screen

Confluence Tab

Imaging runs in the **Confluence** tab and you can watch the process progress in a plate view. A schematic of the plate appears with the image frames superimposed in light blue. As images are captured and processed, the well frame changes color, light brown then to green. After a frame changes to green, you can click on it, and view the image in the **View Image** tab.

View Image Tab

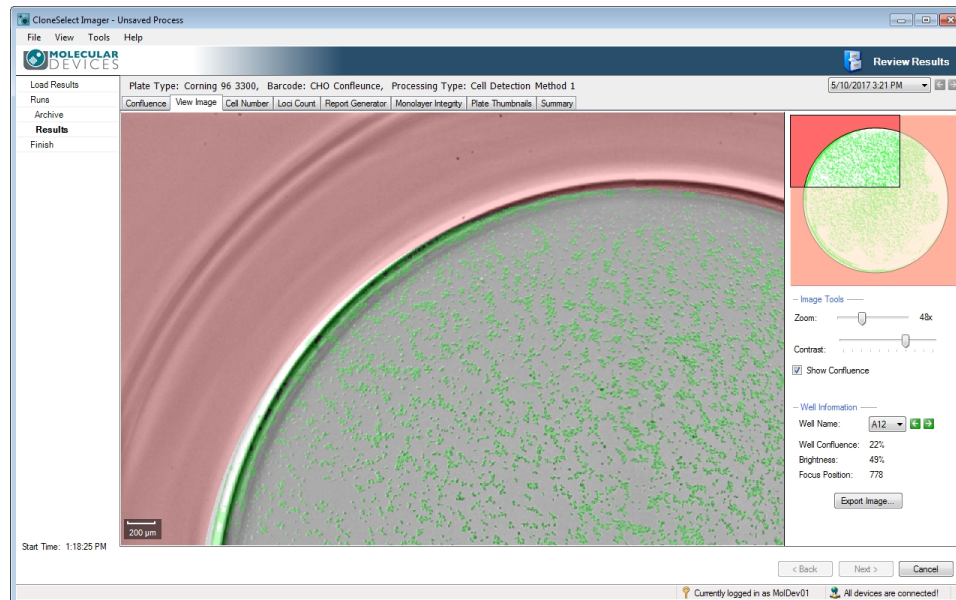


Figure 4-7: View Image screen

To view a frame image:

1. In the **Confluence** tab, after a frame changes to green, click on it.
2. In the **View Image** tab screen (Figure 4-7), you can use the following controls:
 - Well Schematic—Shows you which part of the well you are viewing.
 - Show confluence overlay—Click the check box to toggle off or on the green overlay.
 - Change well view—Click on the well name list menu to select a different well to view.
 - Export Image—Click to save the current image in bmp, jpg or png format. The overlay is not saved with the image.
 - Zoom—Move the **Zoom** slider to look closer or farther at the image. As you change the zoom level, the Scale Bar in the lower left of the image window automatically adjusts.

*** Tip:** The figure turns red when there is distorting image pixelation.

3. To return to the plate view, click the **Confluence** tab.

Continue to the Next Step

When the **Imaging** process finishes, you automatically go to the **Results** screen step. See [Results on page 36](#).

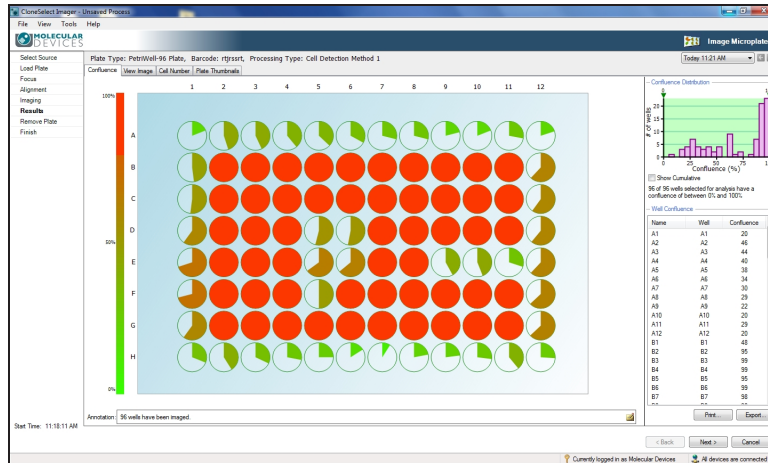
To exit the process and return to the **Image Microplate** process properties screen, click **Cancel**. See [Set Properties on page 26](#).

Results

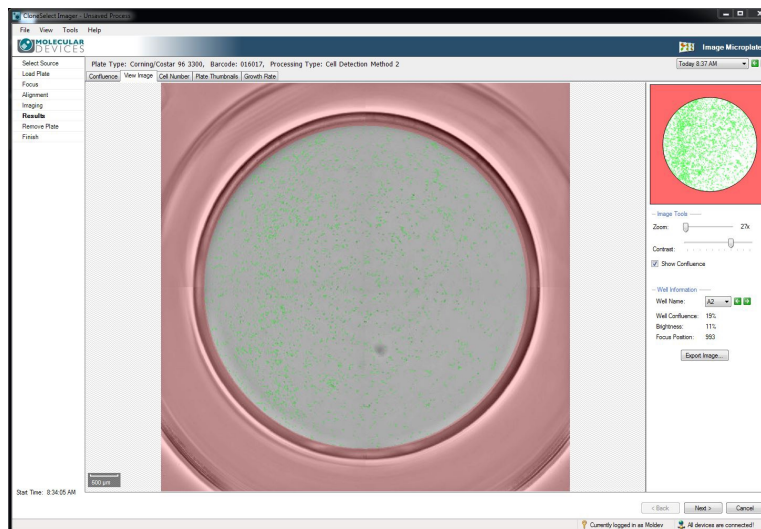
After an **Imaging** process completes, by default the **Results** screen for the **Image Microplate** process opens in the **Confluence** tab. The **Plate Type**, **Barcode** and **Processing Type** appear at the top of the screen for reference.

The **Results** screen for the **Image Microplate** process includes the following tabs:

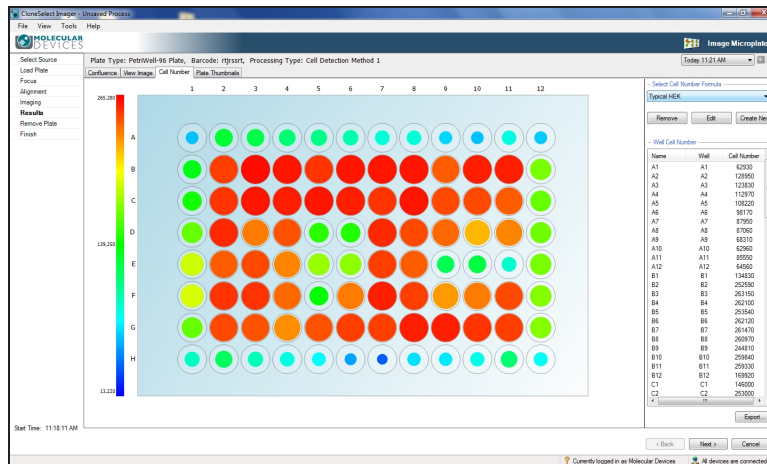
- **Confluence**—Shows the confluence level for each well, both graphically and as a list. Also see [Confluence on page 45](#).



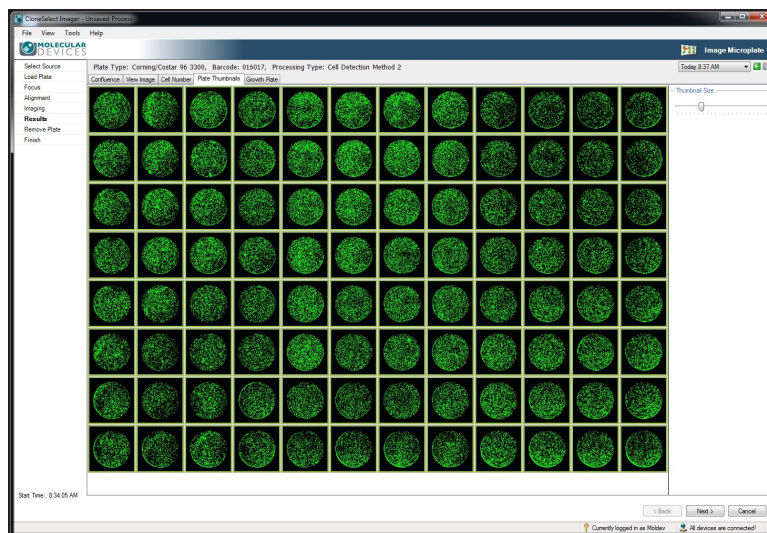
- **View Image**—Shows the image for a selected well. Also see [View Image on page 48](#).



- **Cell Number**—Shows the estimated number of cells in each well, both graphically and as a list. Also see [Cell Number](#) on page 49.



- **Plate Thumbnails**—Shows the confluence area for each well. Also see [Plate Thumbnails](#) on page 68.



✳ **Tip:** You can use these tabs and more for reviewing and analyzing the results in the **Review Results** process. See [Review Results](#) on page 40.

Continue to the Next Step

To continue to the next step, click **Next**. See [Remove Plate](#) on page 38.

To exit the process and return to the Image Microplate process properties screen, click **Cancel**. See [Set Properties](#) on page 26.

Remove Plate

In the **Remove Plate** screen step, the plate carrier automatically ejects, and you are prompted to remove the microplate from the plate carrier. You can choose to either repeat the imaging process with another plate, or continue to the end of the process.

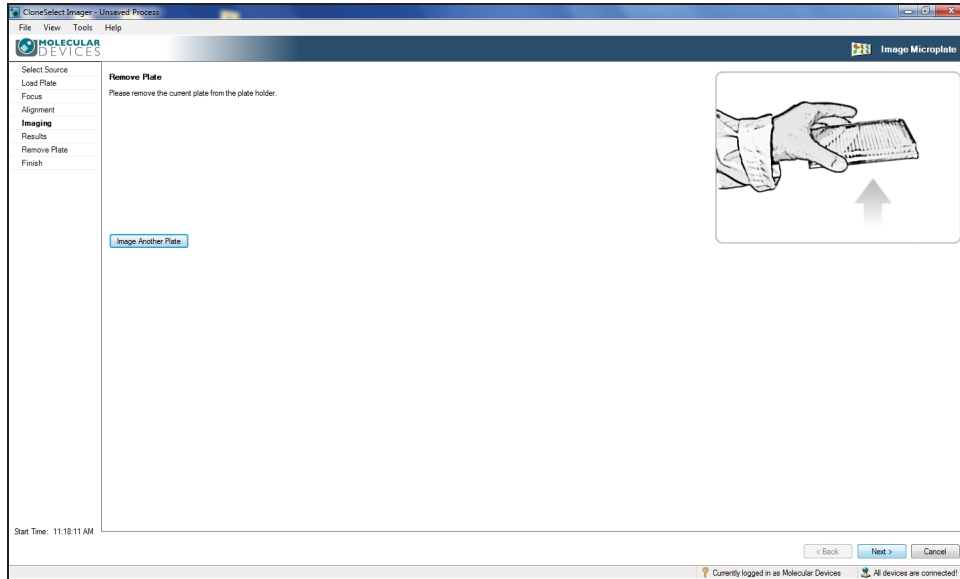


Figure 4-8: Remove Plate prompt screen

Image Another Plate

To image another plate:

- If you want to image another plate, click **Image Another Plate**.
The plate carrier remains in the ejected position, and the **Select Source** screen appears for you to continue. See [Select Source on page 28](#).

Continue to the Next Step

To continue to the next step, click **Next**. The plate carrier retracts, and the **Finish** screen appears. See [Finish on page 39](#).

To exit the process and return to the Image Microplate process properties screen, click **Cancel**. See [Set Properties on page 26](#).

Finish

When the **Image Microplate** process ends, the **Finish** screen appears.

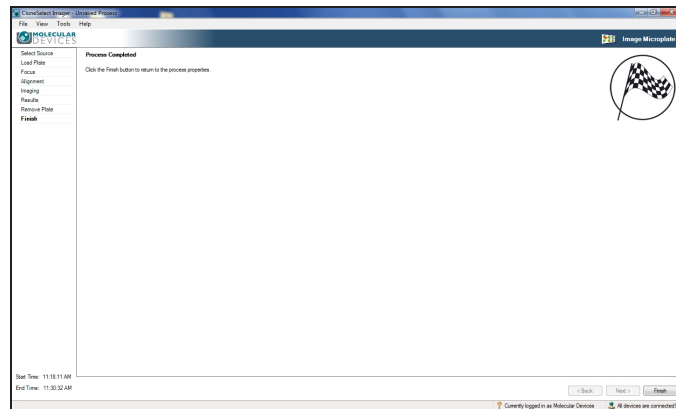


Figure 4-9: Image Microplate process Finish screen

To close the screen, click **Finish**. You return to the **Image Microplate** process **Summary** screen.

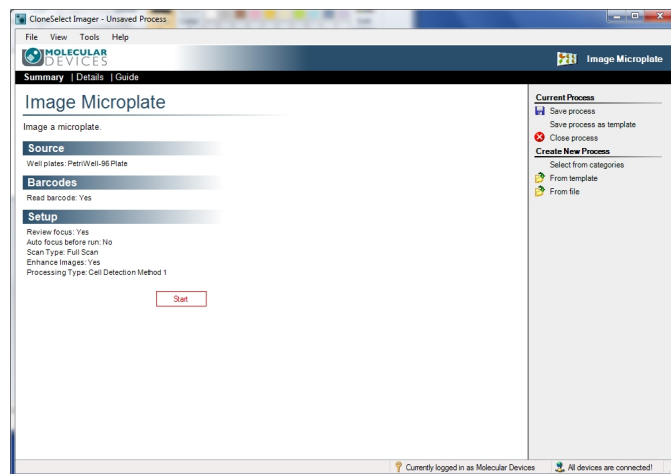


Figure 4-10: Image Microplate process properties Summary screen

In the **Image Microplate** process **Summary** screen, you can use the shortcut menu on the right side of the screen to do any of the following **Current Process** tasks:

- **Save process**—Saves the current process to the **Recent Processes** tab for future access.
- **Save process as template**—Saves the current process to the **Template** tab for future access.



Note: A saved template cannot be changed.

- **Close Process**—Closes the process and returns you to the **Navigation** screen. See [Navigation Screen on page 21](#).

Review Results

The **Review Results** process lets you review previous imaging runs to be reviewed and also to do further data processing, such as confluence, monitor cell growth, cell number estimation, monoclonality analysis and migration assay analysis.

*** Tip:** After an **Image Microplate** process completes, the results automatically appear in a **Results** process screen. See [Results on page 36](#). That screen includes a subset of the functionality that can be done using the **Review Results** process.

To review results:

1. From the **Navigation** screen, click on **Review Results** to start the process.
2. From the **Review Results** properties screen, click **Start**.

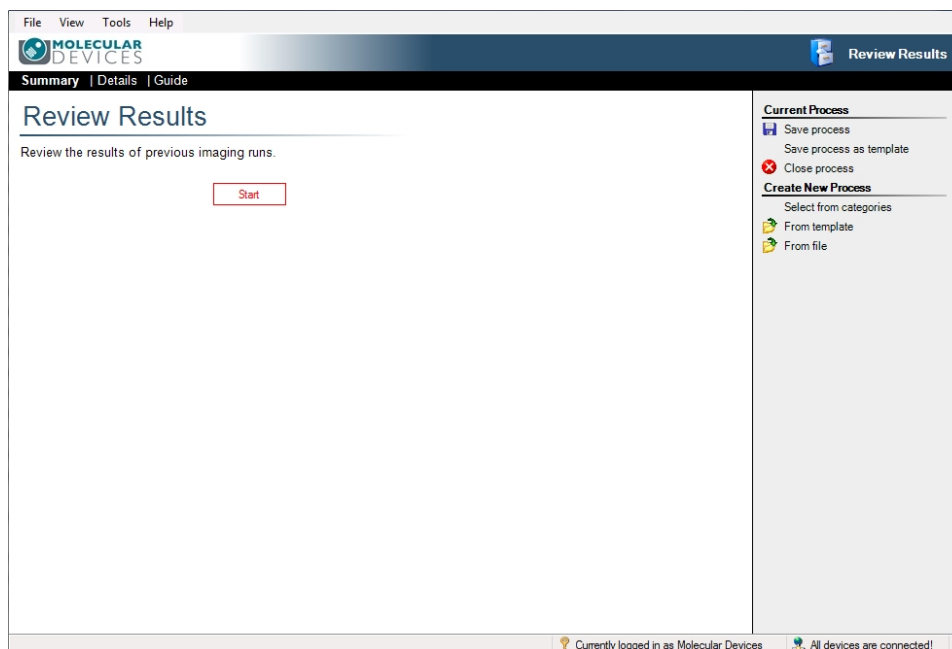


Figure 4-11: Review Results start screen

3. Continue in the **Load Results** screen. See [Load Results on page 41](#).

Load Results

The default location where imaging results are stored is the `C:\Image Archive` folder. All the image and data files for each individual run are stored in a folder named with the date and time stamp for the run, for example `2016-11-01T11-52-10`. Within the *Image Archive* folder, the run folders are automatically organized into separate folders for each month, for example `2016-10`.

The **Review Results** process, starts in the **Results Archive** tab, in the **Runs** list. Plates imaged on the system appear in a list.

*** Tip:** If free space on the disk is running low, the bar at the top of the window appears orange and a warning is displayed in the top right corner. If disk space is getting critically low the bar is colored red.

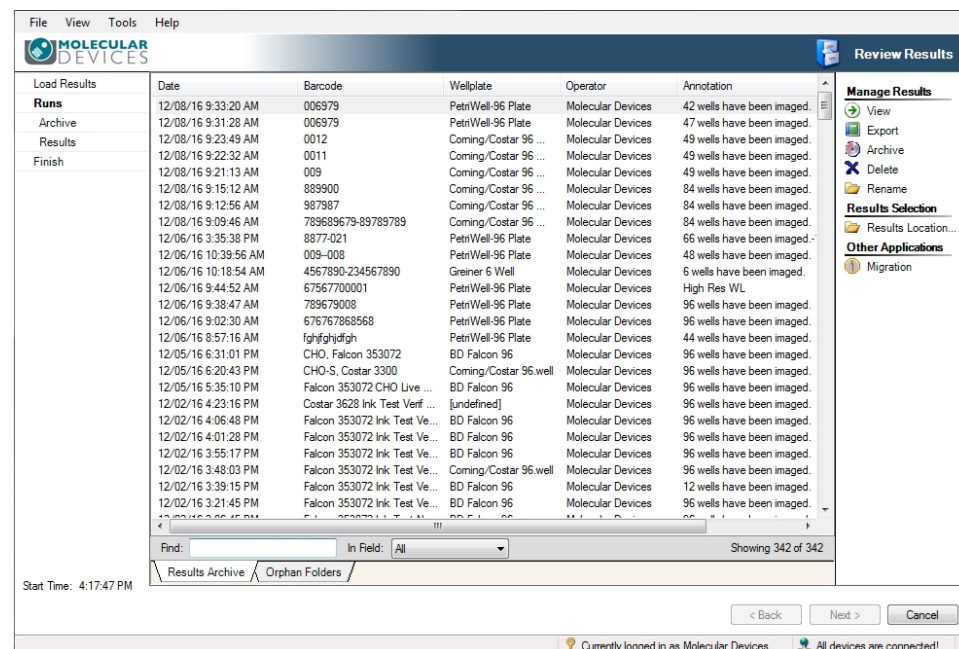


Figure 4-12: Review Results Runs screen

To load results:

- In the **Results Archive** list, double-click on the run date you want to view.

*** Tip:** To select multiple plates in the list, use the **<SHIFT>** and **<CTRL>** keys.

- Alternatively, you can use the following menu options on the right side of the screen:
 - **Manage Results**—See [Manage Results on page 42](#).
 - **Results Selection > Results Location**—When you save results data to a location other than the default *C:\Image Archive* folder location, click to select from that alternate location.
 - **Other Applications > Migration**—(Optional) You must purchase a license for this software module. See [Migration Analysis on page 77](#).

Manage Results

The right side of the **Review Results** process screen includes the **Manage Results** menu. In this menu you can do the following tasks:

- [View on page 42](#)
- [Export on page 43](#)
- [Archive on page 43](#)
- [Delete on page 43](#)
- [Rename on page 44](#)

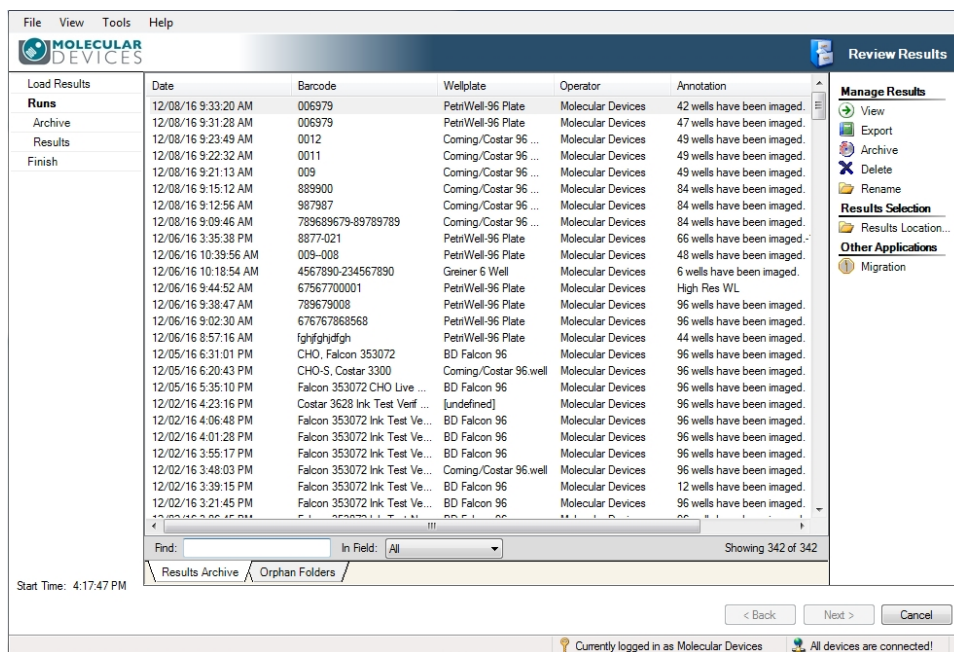


Figure 4-13: Review Results screen

View

This is another way to view from the **Results Archive** list.

- Select a run date from the **Results Archive** list, and then click **View**.

Export

Opens an export wizard dialog. Using the wizard you can select the results data you want to export.

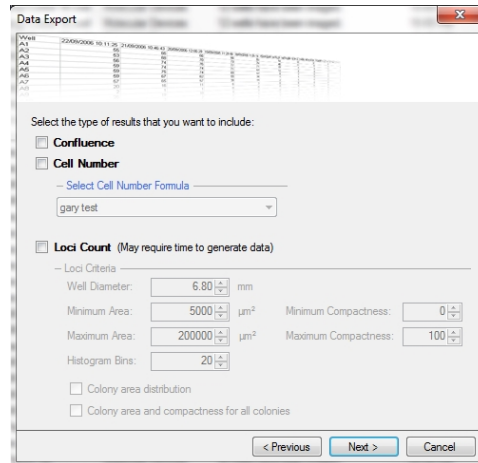


Figure 4-14: Data Export wizard dialog

Archive

Lets you archive to a network drive or disk of your choice the results data stored in the default *C:\Image Archive* folder location.

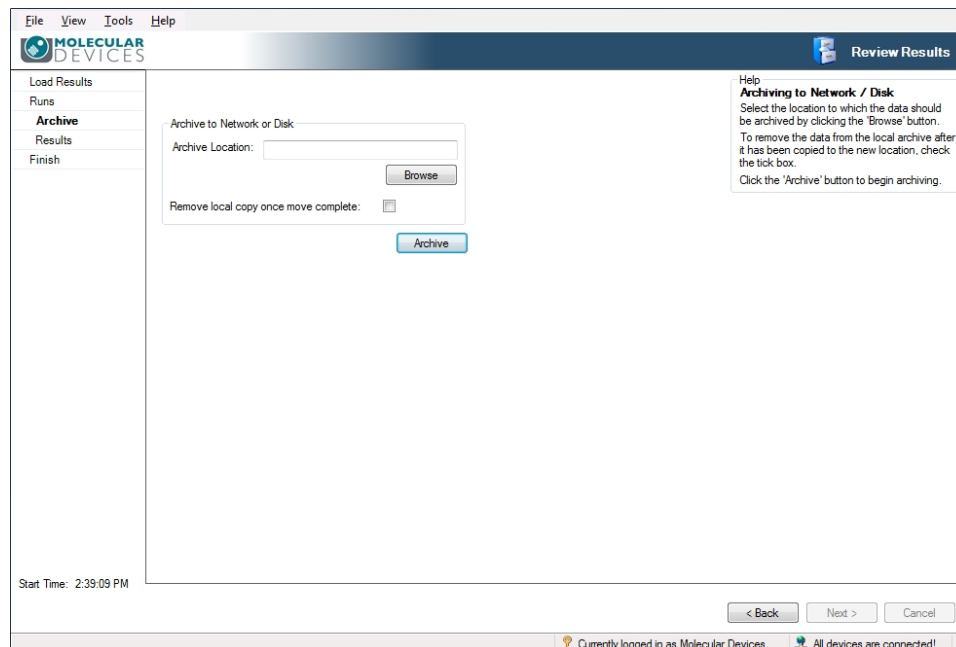


Figure 4-15: Archive screen

Delete

Deletes all the images and results for the selected data set.

Rename

Lets you rename the **Barcode** identification information.

*** Tip:** To do **Growth Rate** analysis, this information must be the same for multiple runs.

To rename a run in the list:

1. Select a run date from the **Results Archive** list, and then click **Rename**.
2. In the **Barcode** field, type new identification information, and then press **ENTER**.

Analyzing Results

When you load results, by default the **Results** screen for the **Review Results** process opens in the **Confluence** tab. The **Plate Type**, **Barcode** and **Processing Type** appear at the top of the screen for reference.

The **Results** screen for the **Review Results** process includes the following tabs:

- [Confluence on page 45](#)
- [View Image on page 48](#)
- [Cell Number on page 49](#)
- [Loci Count on page 52](#)
- [Plate Thumbnails on page 68](#)
- [Growth Rate on page 70](#)
- [Summary on page 75](#)

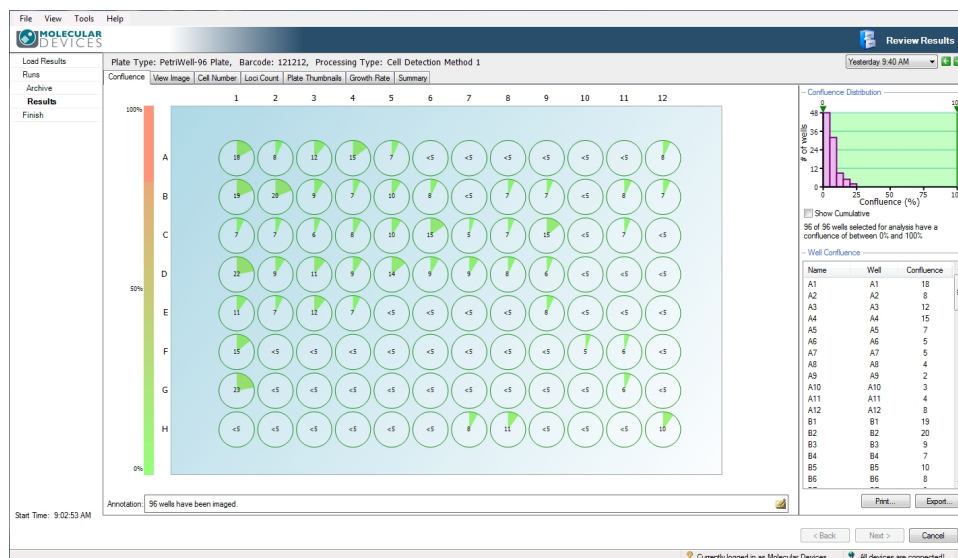


Figure 4-16: Results screen for the Review Results process

Confluence

The **Confluence** tab displays the confluence level for each well, both graphically and as a list.

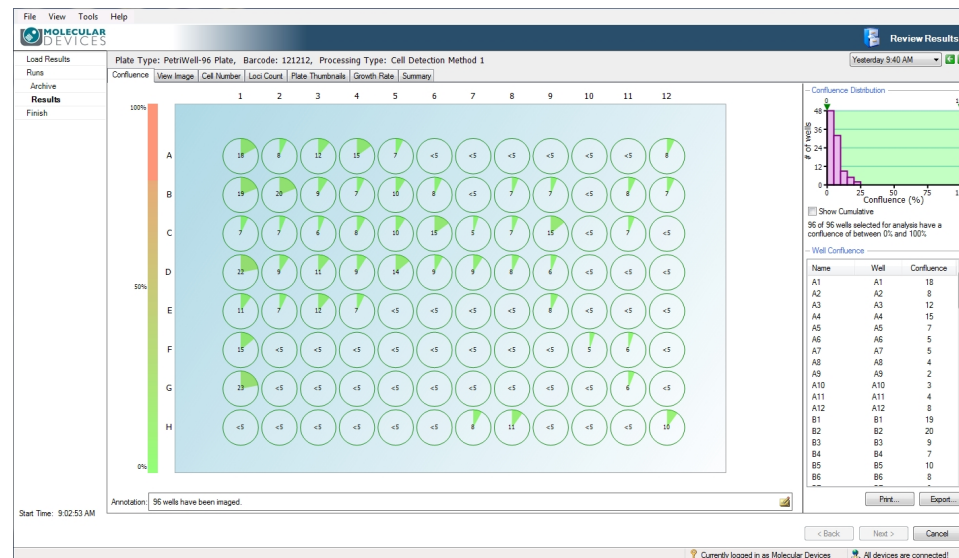


Figure 4-17: Confluence tab screen

Plate Confluence Overview

Each well is represented by a pie chart of the confluence level for that well. The pie charts are color-coded such that low confluence is green and high confluence is red with shades in between representing the intermediate levels.

To see the percentage of confluence for all the wells in the plate, hover the cursor over the pie chart area. Confluence lower than 5% appears as <5 and confluence above 80% appears as >80.

To see coordinates and confluence for an individual well, hover the cursor over a single well.

To see an image of a well in the **View Image** tab, click on a well. See [View Image on page 48](#).

To copy the the overview to the clipboard, hover the cursor over a well, right-click, and then click **Copy to clipboard**.

Confluence Distribution

The bar chart at the top right of the window shows the number of wells with a given confluence level.

Dual Gates

The arrows pointing down at the top of the bar chart can be dragged across the chart. As both lower and upper arrows are dragged, the corresponding confluence data appears. Any wells that do not fall inside these lower and upper gates will be grayed out. The text shown below the chart updates to give the number of wells that fall within the selected confluence. For example, 10 out of 36 wells selected for analysis have a confluence of between xx % and xx %.

Show Cumulative

When you select **Show Cumulative**, the individual bars in the chart merge to show the confluence as continuous data.

Well Confluence

The confluence for each well appears in the **Well Confluence** list on the right.

Print

The **Print** option prints a schematic presentation of the confluence overview in the plate format. The confluence values appear within the wells and the plate details appear at the top of the overview.

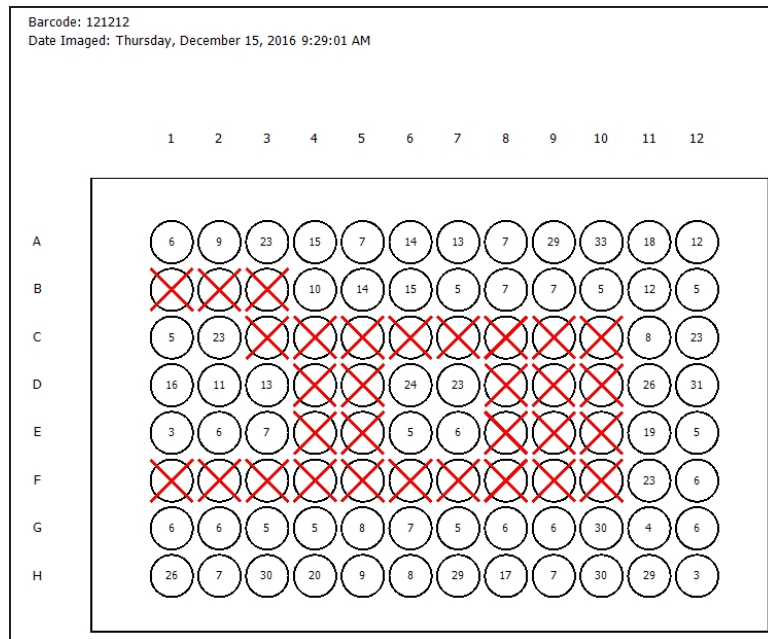


Figure 4-18: Print schematic presentation of the confluence overview

To print the list, click **Print**.

Export

The **Export** option lets you save the well confluence list as a **Comma Separated Values (CSV)**, **Excel**, or **XML** file.

The **Data Export** wizard guides you through the process of exporting the **Confluence** data. **Cell Number** and **Loci Count** data can also be exported.

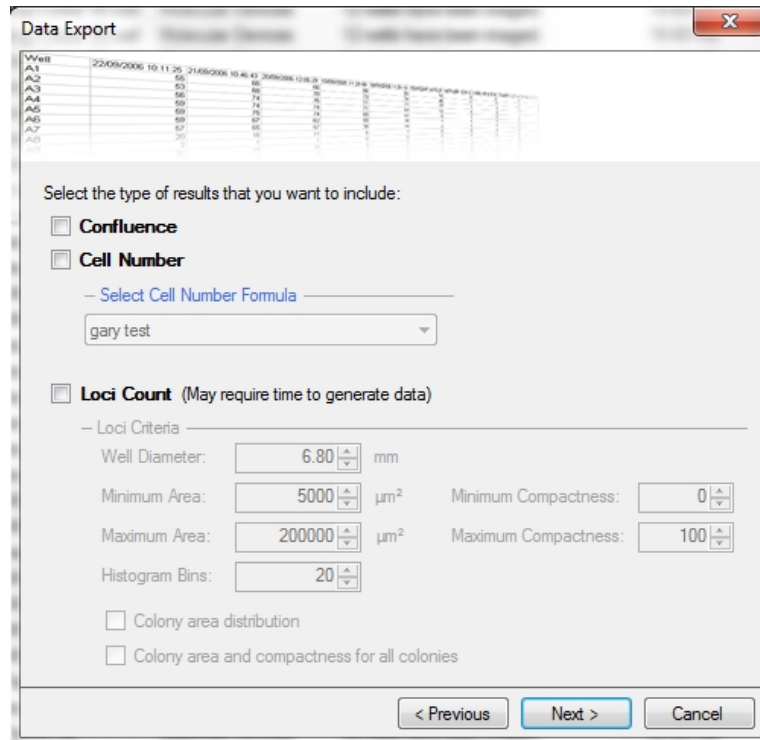


Figure 4-19: Data Export wizard dialog

The wizard provides the option to export the data for the following time points:

- The currently selected time
- The most recent time for each well plate
- The complete time series.

Annotation

By default the **Annotation** field shows the number of wells that have been imaged. You can edit this to enter some more meaningful annotation.



Note: This cannot be changed when viewing the data through a remote data viewer.

Cancel

If you click **Cancel**, you are prompted to confirm you want to end the process. Clicking **Yes** at the prompt takes you to the **Load Results** screen. See [Load Results on page 41](#).

To finish, see [Finish on page 83](#).

View Image

The **View Image** tab displays the images for a selected well. The whole well is displayed.

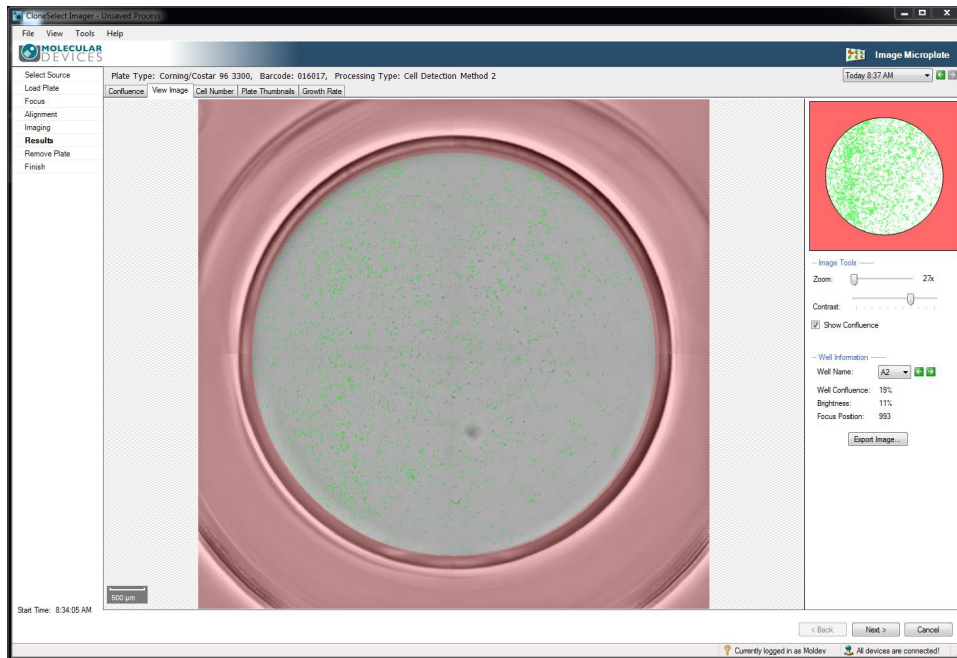


Figure 4-20: View Image tab screen

Well Schematic

By clicking on the image of the well currently being displayed, you can navigate to different areas of the well and see where you are in the well in the image schematic on the right. The confluence levels of the currently selected whole well are displayed below the schematic.

Imaging Tools

The following controls are available in the **Imaging Tools** section:

- **Zoom**—Move the slide either left or right to zoom out or in. The lowest magnification of the image is 18x and the highest is 144x. The figure turns red when there is distorting image pixelation. When zoomed into an image, the zoomed area will be displayed on the image thumbnail to the right.
- **Contrast**—Move the slider either left or right to improve the image contrast.
- **Show Confluence**—Select to turn the overlay on or off. The area of the image outside the well is overlaid in red and the regions of the well where colony growth is detected are overlaid in green.

Well Information

Well Information includes the following:

- **Well Name**—Use the drop down menu or the arrows to select a different well.
- **Well Confluence**—Shows the well confluence percentage for the selected well.
- **Focus Position**—Shows the focus point of the image when captured.
- **Brightness**—Shows as a percentage the brightness level used to capture the imaged.

Export Image

The **Export Image** option lets you save the currently displayed image in .bmp, .jpg or .png format. If **Show Confluence** is selected, the confluence overlay is displayed with the exported image. The zoomed position is saved within the image.

Cancel

If you click **Cancel**, you are prompted to confirm you want to end the process. Clicking **Yes** at the prompt takes you to the **Load Results** screen. See [Load Results on page 41](#).

To finish, see [Finish on page 83](#).

Cell Number

The **Cell Number** tab displays the estimated number of cells in each well, both graphically and as a list. The number of cells is estimated using a formula that can be created for each of your cell types from the confluence readings of a standard plate containing known numbers of cells. See [Setting up Cell Number Estimation on page 109](#) for a detailed procedure.

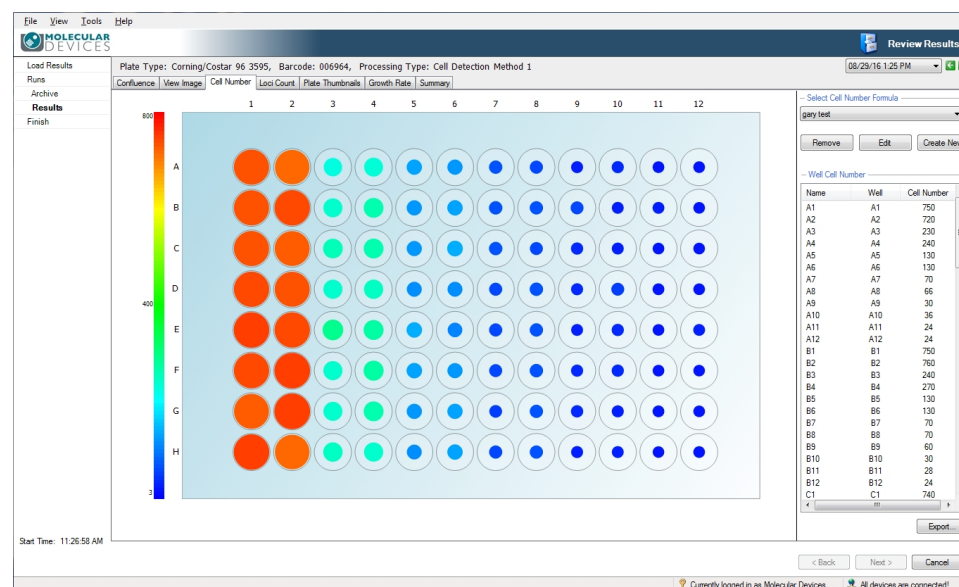


Figure 4-21: The cell number view

Each well contains a colored fill with the estimated cell number indicated by the color and size of the fill area, with a large red fill area representing a high cell number and a small blue fill area representing a low cell number. A color scale is shown to the left of the graphic. Hovering the cursor over a well displays a tool tip giving the well coordinate and the estimated cell number for that well.

Select Cell Number Formula

Use the **Select Cell Number Formula** menu to select from the created formulas for estimating the number of cells per well.

In the **Select Cell Number Formula** section, you can also do the following:

- **Remove**—Click **Remove** to delete the currently selected cell number formula from the menu list.
- **Edit**—Click **Edit** to change the currently selected cell number formula. See [Edit on page 50](#).
- **Create New**—Click **Create New** to create a new cell number estimation formula. See [Create New on page 50](#).

Edit

A graph displays the cell number against confluence for the formula.

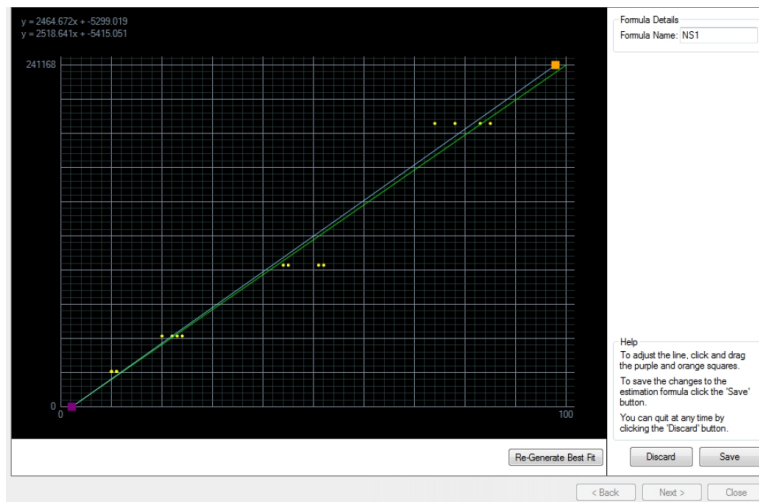


Figure 4-22: Edit Cell Number Formula screen

To edit, drag the handles at each end of the line. The formula is appears at the top left and the edited version appears immediately below it.

To automatically generate a best fitting line, click **Regenerate Best Fit**.

To change the formula name, in the **Formula Name** field, type a new name.

To cancel the edits and return to the **Cell Number** tab, click **Discard**.

To keep the edits and return to the **Cell Number** tab, click **Save**.

Create New

Ensure that the confluence results displayed in the Confluence tab are those for a standard plate containing known numbers of cells. The software automatically selects up to twelve wells with confluence values ranging between 10% and 80% and displays them at the bottom of the graph.

A screen to create a new cell number estimation formula appears. Ensure that the confluence results displayed in the Confluence tab are those for a standard plate containing known values of cells. The software automatically selects up to twelve wells with confluence values ranging between 10% and 80% and displays them on the left of the graph.

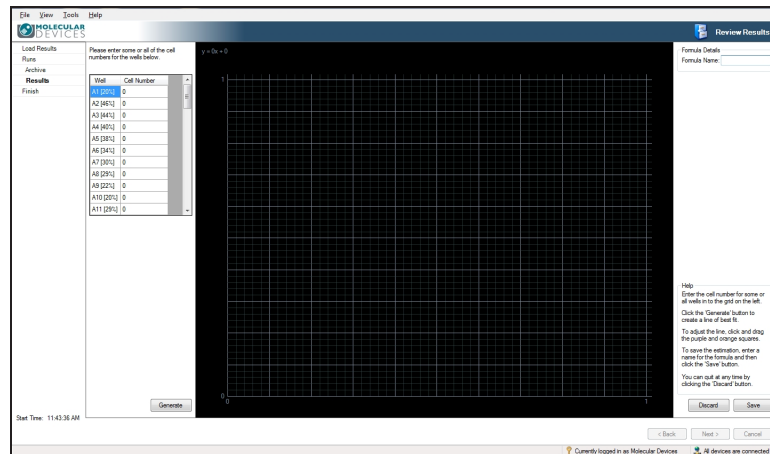


Figure 4-23: Create New Cell Number Formula screen

To name your formula, in the **Formula Name** field, type your formula name.

Enter the cell numbers for each well into the corresponding well number box to the left of the graph. After you enter all the relevant information, click **Generate**.

To cancel the edits and return to the **Cell Number** tab, click **Discard**.

To keep the edits and return to the **Cell Number** tab, click **Save**.

Well Cell Number

A list of estimated cell numbers for each well appears in the **Well Cell Number** list on the right.

To change the well name, in the **Name** column of the list, click the well name, pause, then click the well name again. When the edit field appears, type a new name.

Export

The **Export** option lets you save the well cell number list as a **Comma Separated Values (CSV)**, **Excel**, or **XML** file.

The **Data Export** wizard guides you through the process of exporting the **Cell Number** data. **Confluence** and **Loci Count** data can also be exported.

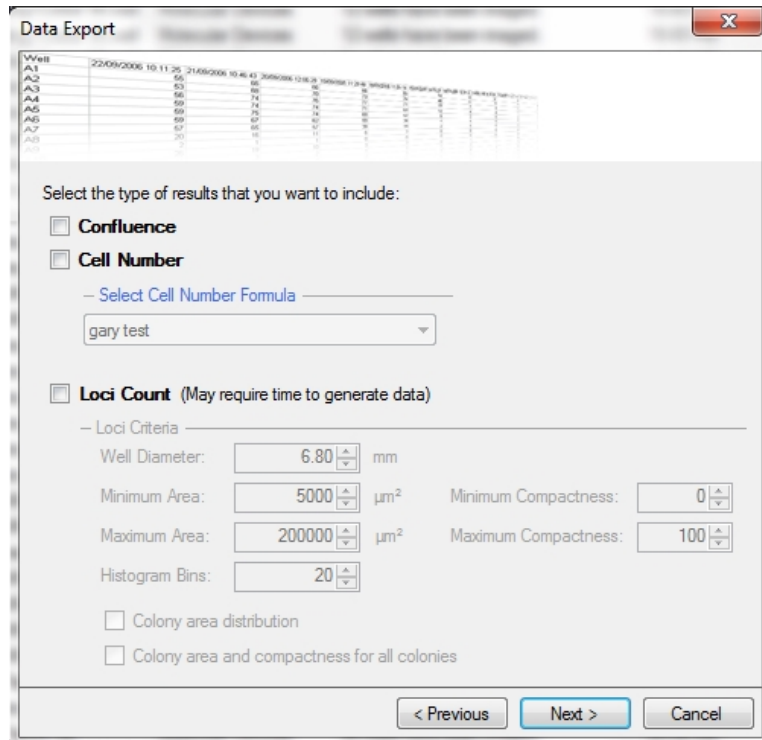


Figure 4-24: Data Export wizard dialog

Cancel

If you click **Cancel**, you are prompted to confirm you want to end the process. Clicking **Yes** at the prompt takes you to the **Load Results** screen. See [Load Results on page 41](#).

To finish, see [Finish on page 83](#).

Loci Count

For colony formation assays, use Loci Count to count the number and size (area) of colonies in each well.

Generated images can be processed using **Loci Count**. This detects and counts the number of loci of growth, for example, cell colonies for applications including monoclonality verification and colony forming assays. See [Monoclonality Workflow on page 57](#) and [Colony Forming Assay Data Workflow on page 60](#). If a plate is imaged multiple times during colony growth, the history of each well with one identified colony can be viewed for visual proof that the colony is derived from a single cell progenitor.

Plate View

In the **Loci Count** tab, the plate view displays each well with a figure indicating the number of cell colonies found in the well. The default plate view displays the **Loci Count** (in the **Display Statistic** section menu), however this can be changed to **Mean Loci Area**, which displays this value within each of the wells.

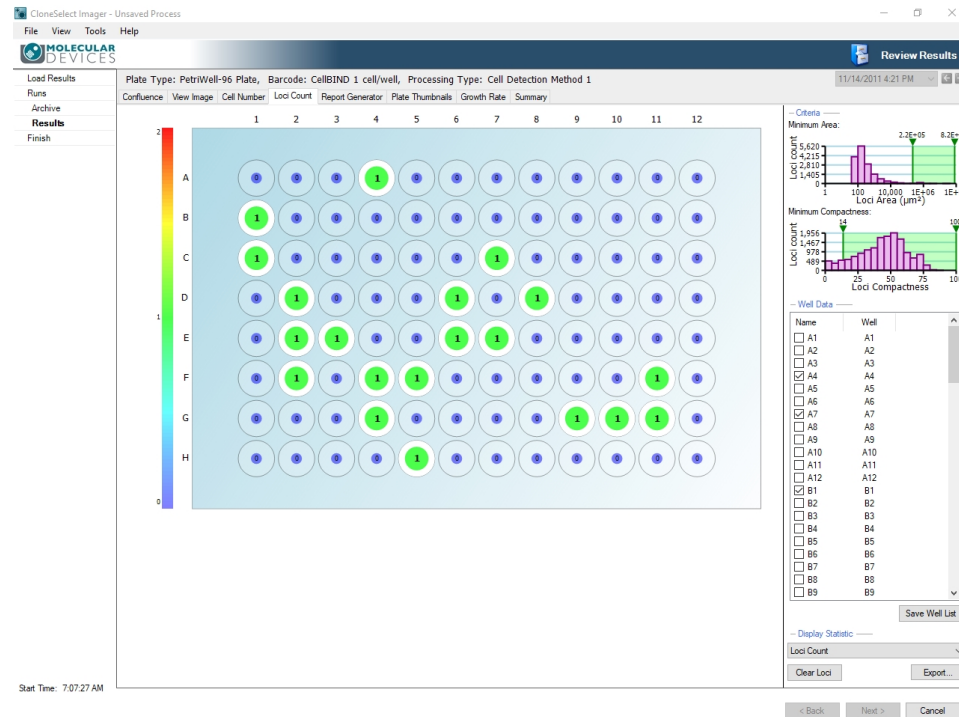


Figure 4-25: Loci Count in Plate View

Criteria

The **Criteria** section includes the following bar charts:

- **Minimum Area**—The loci count (frequency) is plotted against the loci area (μm^2) in a log scale. The bar chart has two, lower and upper, gates that can be moved accordingly to eliminate unwanted objects/debris within the well that artificially inflate the number of loci.
- **Minimum Compactness**—The loci count (frequency) is plotted against the loci compactness. The bar chart has two, lower and upper, gates that can be moved accordingly to eliminate irregularly shaped objects from the loci count. Compactness is the relation between the area and the perimeter, expressed as a ratio of the actual area and that of a perfect circle with the same perimeter.

Well Data

In the Plate View **Well Data** section, you can click on a well to view the image. You can also select well check boxes to save as a well list.

To save the selected wells in the list, click **Save Well List**. This file can be saved as a .csv file. The file includes the plate barcode, run date, operator and the selected wells.

Display Statistics

The **Display Statistics** menu lets you select to view as the overview either **Loci Count** or **Mean Loci Area**.

To clear all of the current loci data processing, click **Clear Loci**.

Export

The **Export** option lets you save the wells and the corresponding Loci Count list as a **Comma Separated Values (CSV)**, **Excel**, or **XML** file.

The **Data Export** wizard guides you through the process of exporting the **Loci Count** data. **Confluence** and **Cell Number** data can also be exported.

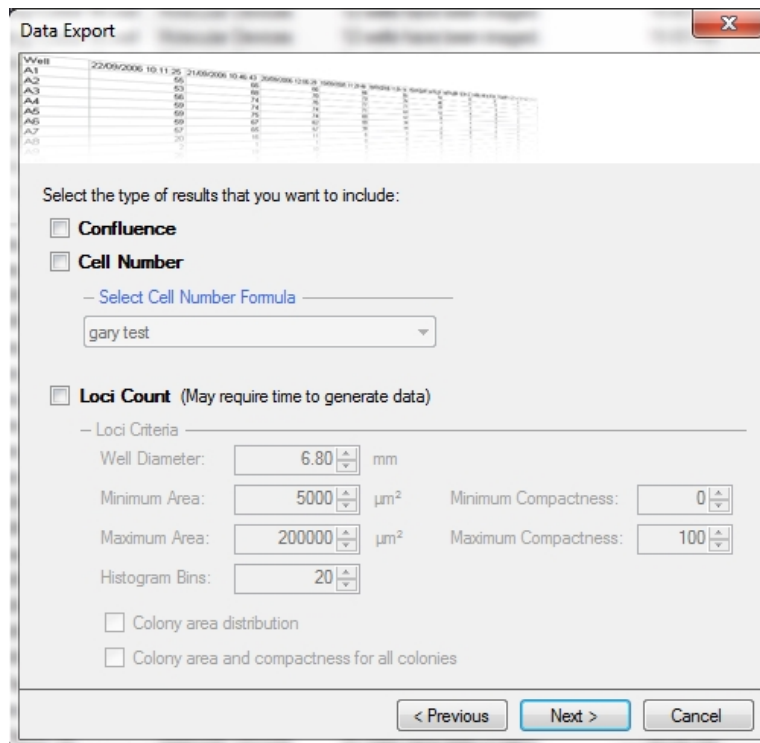


Figure 4-26: Data Export wizard dialog

The **Loci Count** criteria automatically populate from the gated **Minimum Area** and **Minimum Compactness** graphs. You can also export the following:

- **Histogram Bins**—This option is used if the Colony area distribution box is selected. This value sets the number of equi-sized bins for exporting loci frequency counts. The default value is set to 20 bins.
- **Colony area distribution**—When this box is selected, the colony area distribution data is divided into the number of bins specified in the **Histogram Bins** and displayed with any other exported data.



Note: Only wells selected in the **Well Data** list export.

- **Colony area and compactness for all colonies**—When this box is selected, a list of all colonies in all wells with the area and compactness for each colony exports.



Note: There is no filtering, so all the data is listed.

Well View

Clicking on the well of interest in the plate view of the **Loci Count** tab displays the image for the well with an overlay over areas of growth detected according to the **Minimum Area** and **Minimum Compactness** criteria.

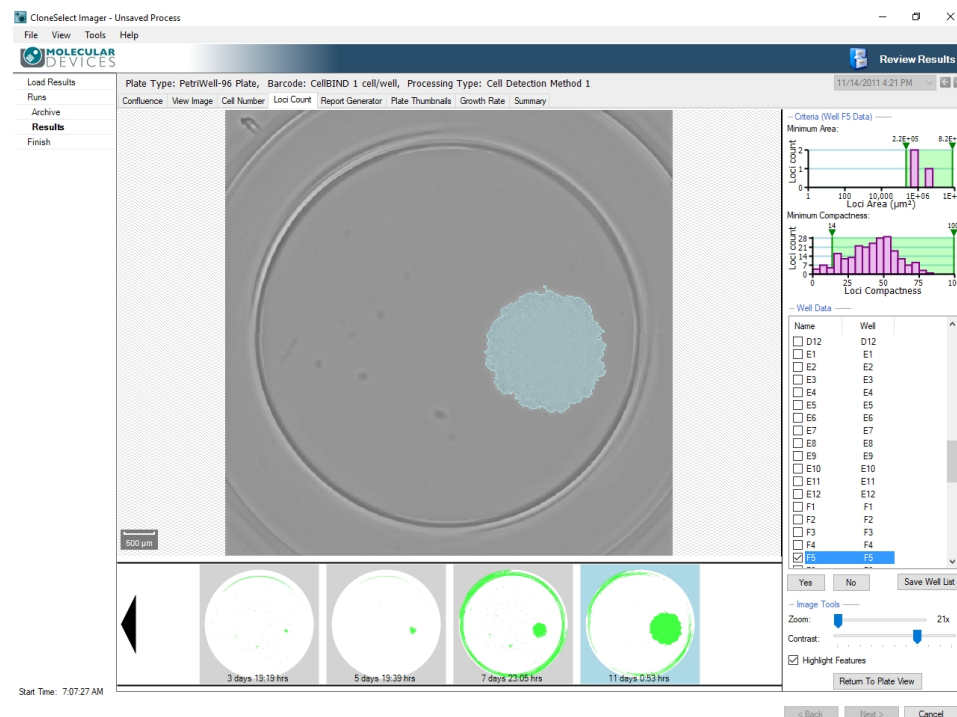


Figure 4-27: The Well View in Loci Number

If the microplate has been imaged at several time points a film strip of schematics of the well at the various time points appears across the bottom of the screen with loci colored in green. This provides a way of checking if a colony results from a single cell or from more than one cell.

To view the well image at that time point with the loci from the latest image outlined, click on any of the schematics thumbnails.

To save the sequence of images, right-click on the sequence, and select **Export image sequence**. The sequence can be saved as a .bmp, .jpg or .png file. The file includes the confluence percentage and time and day the image was acquired.

To print a monoclonality report, see [Report Generator on page 63](#).

Well Data

In the Well View **Well Data** section, to see if a colony is monoclonal, click on a well in the list. Move through the list and mark wells as monoclonal in the following ways:

- Click **Yes** button to mark a well as monoclonal and scroll to the next well image. Click **No** to scroll to the next well without marking the well as monoclonal.
- Click on a well to view its image and click on the check box to mark it as monoclonal.
- Use the keyboard up and down arrow keys to scroll through the wells and the spacebar to mark the wells.

To save the selected wells in the list, click **Save Well List**. This file can be saved as a .csv file. The file includes the plate barcode, run date, operator and the selected wells.

Imaging Tools

The following controls are available in the **Imaging Tools** section:

- **Zoom**—Move the slide either left or right to zoom out or in respectively. The lowest magnification of the image is 18x and the highest is 144x. When the figure turns red the system is zooming digitally and can cause some pixelation of the image. When zoomed into an image, the zoomed area will be displayed on the image thumbnail to the right.
- **Contrast**—Move the slider either left or right to improve the image contrast.
- **Highlight Features**—Select to mark features with different overlay colors.
- **Return To Plate View**—Click to return to the plate view within the Loci Count tab to view other wells. See [Plate View on page 53](#).

Cancel

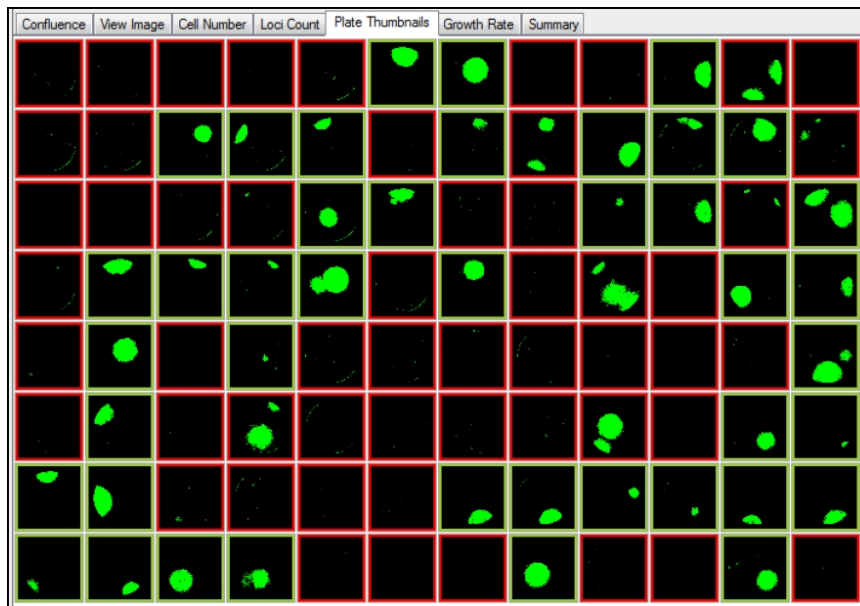
If you click **Cancel**, you are prompted to confirm you want to end the process. Clicking **Yes** at the prompt takes you to the **Load Results** screen. See [Load Results on page 41](#).

To finish, see [Finish on page 83](#).

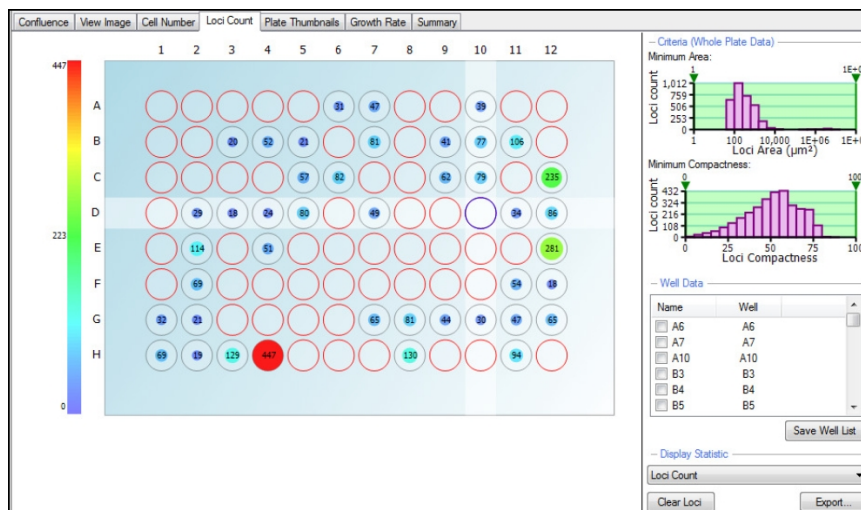
Monoclonality Workflow

To run the Loci Count process for a monoclonal dataset:

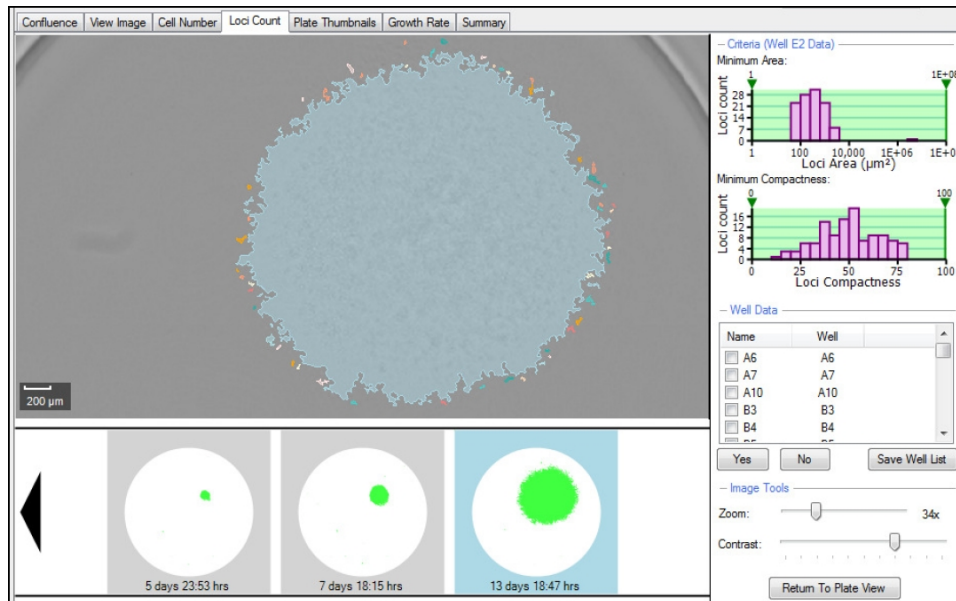
1. After imaging the plate of interest at several time points, open the latest data point in **Review Results**. The software automatically opens the data in the **Confluence** tab.
2. Click **Plate Thumbnails** to display all the imaged wells, and then to save processing time, exclude wells without at least 1 colony within the well. See [Plate Thumbnails](#) on page 68.



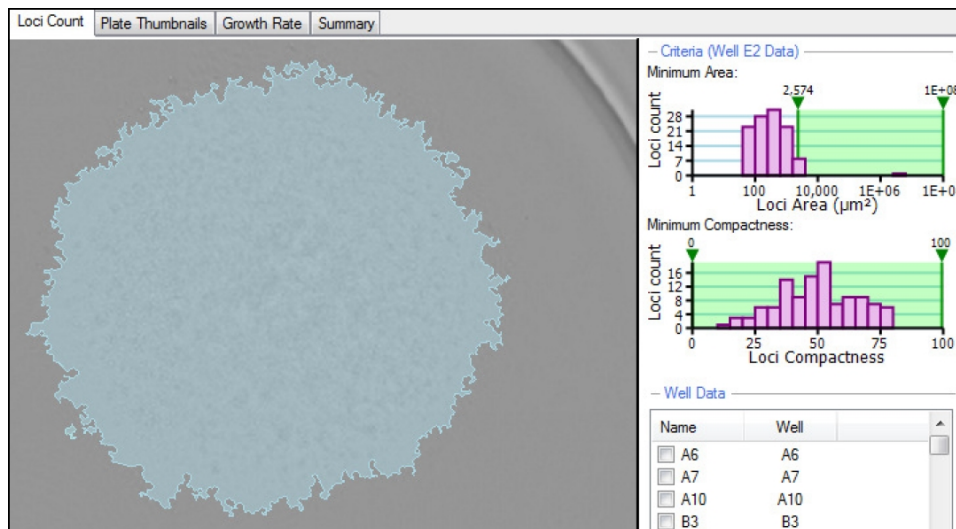
3. Click **Loci Count** and adjust the well diameter as needed. See [Loci Count](#) on page 52.
4. Click **Process** to produce the loci counts for the selected wells. The **Loci Count** tab screen changes to display an overview of all the selected wells and their corresponding loci count.



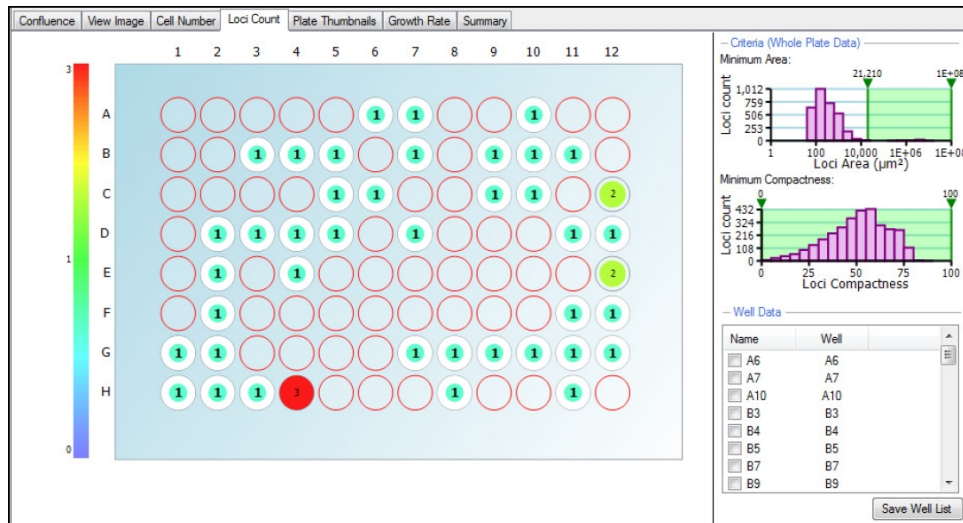
- Click on one of the wells to display the well image. The image shows the loci processing.



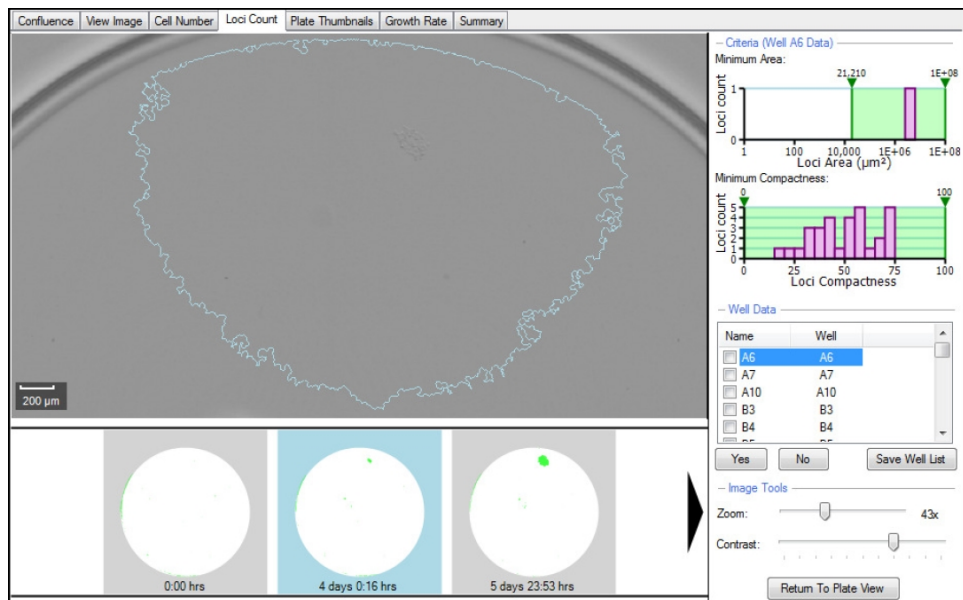
- Move the **Minimum Area** and **Minimum Compactness** lower and uppers sliders on the graphs to gate out any unwanted debris and misshaped colonies within the wells. Adjust to show only wells with 1 or 0 colonies.



- Click on **Return To Plate View** and check through some of the other wells to make sure all debris and misshapen colonies are excluded. The plate overview reflects the processing and displays the new loci count number in each of the wells.



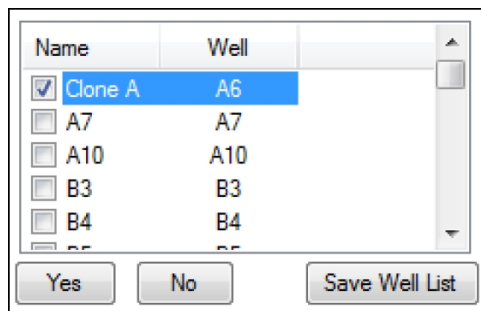
- Click through each well in the **Well Data** list. The well image appears and monoclonality can be determined by looking back through the series of well images and identifying if the colony originated from a single cell.



- If the colony did originate from a single cell, select the well by either selecting the check box or clicking **Yes**.



CAUTION! Do not change the well **Name**. This can cause report errors.



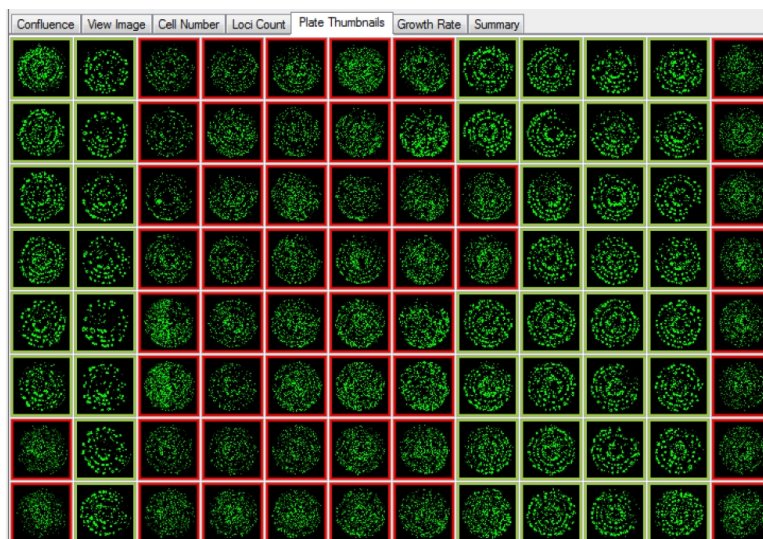
After all the wells have been selected, the list of selected wells containing a monoclonal colony can be saved and printed for further use.

To print a monoclonality report see [Report Generator on page 63](#).

Colony Forming Assay Data Workflow

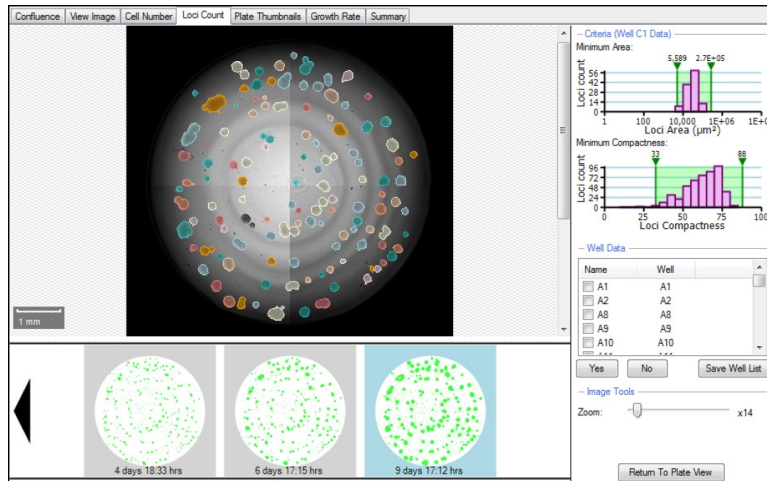
Analyzing colony forming assay data sets are very similar to gating monoclonality data, however the data export can be more detailed.

- After imaging the plate of interest at several time points, open the latest data point in **Review Results**. The software automatically opens the data in the **Confluence** tab.
- Click **Thumbnails** to display all the imaged wells, and then to save processing time, exclude wells without at least 1 colony within the well. See [Plate Thumbnails on page 68](#).



- Click **Loci Count** and adjust the well diameter as needed. See [Loci Count on page 52](#).

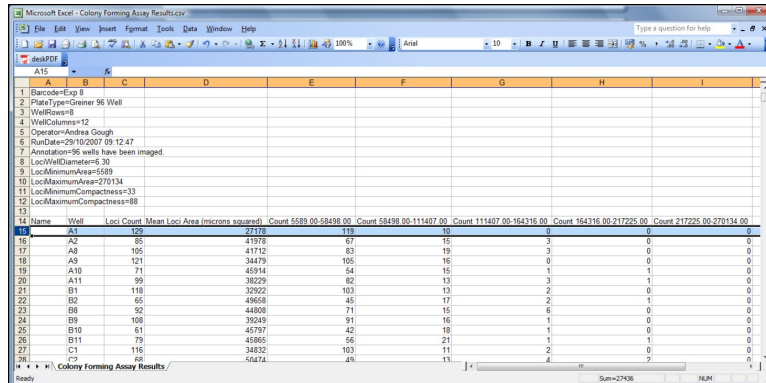
- Click **Process** to produce the loci counts for the selected wells. The **Loci Count** tab screen changes to display an overview of all the selected wells and their corresponding loci count.
- Move the Minimum Area and Minimum Compactness lower and upper sliders on the graphs to gate out any unwanted debris and misshaped colonies within the wells.



- When satisfied with the gating of the data and the loci count produced, the data can be exported. The **Well Data** list is used for accepting wells based on monoclonality so it is not necessary to select any of these wells. The data exports for all wells when exporting the data via the wizard.

- In the **Data Export** wizard, select **Loci Count**. The criteria automatically populate. The default number of bins is 20, but in the example it has been changed to 5 bins. Select **Colony area distribution** and **Colony area and compactness for all colonies** and then click **Next** to finish exporting the data.

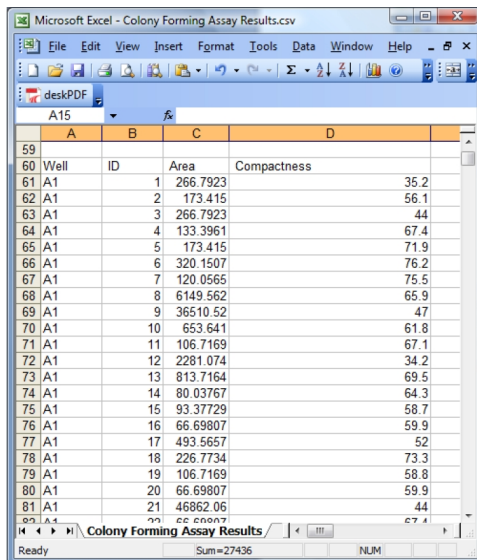
- Within the exported results produced, a summary of the experiment criteria appears in the first rows of the spreadsheet (top left).



- When the **Colony area distribution** is selected, the following information is produced for each well:

- Name (if changed within the software)
- Well number (eg A1)
- Loci Count (this is the total)
- Mean Loci Area
- Bins equally splitting the loci area range into equal groups so that each identified loci will fall into one of the corresponding groups based on its size. For example, out of the 129 identified loci within well A1, 119 of these fall into the first bin with a loci area range of $5589\mu\text{m}^2$ to $58498\mu\text{m}^2$.

When selecting the **Colony area and compactness for all colonies**, the area and compactness of each identified loci is displayed:



Report Generator



Note: Loci Count is included in CloneSelect Imager Software version 1.6 and newer, but is an optional software license in CloneSelect Imager Software version 1.5. Contact your Molecular Devices representative to purchase.

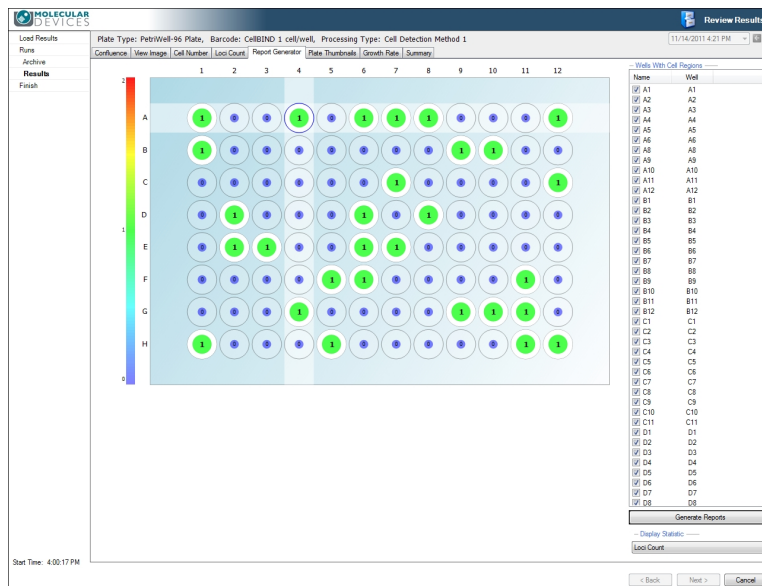
The **Report Generator** tab is an extension of the **Loci Count** functionality and as such is only available when the **Loci Count** license for CloneSelect Imager Software version 1.6 or newer is installed. Use these reports to provide evidence of monoclonality.



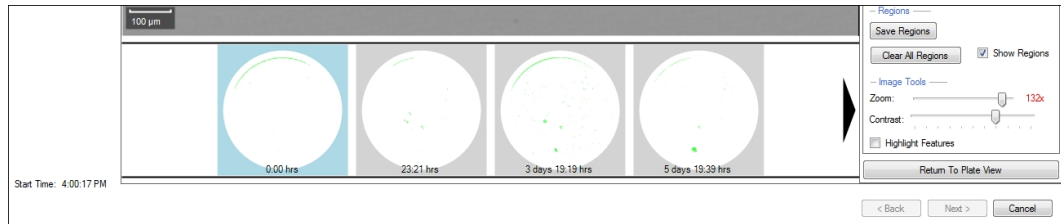
Note: If you see the following error message: “Report generation failed. Please contact technical support for assistance.” make sure that the DocumentAssemblerServiceSetup.exe is installed. Refer to the installation instructions in the *CloneSelect Imager Software Release Notes*.

To generate a monoclonality report:

1. Follow the procedure in [Monoclonality Workflow on page 57](#) to confirm the presence of a single cell.
2. When the **Loci Count** tab plate view shows a matching colony number to that observed in the **Plate Thumbnail** tab, preferably all ones and zeros, click the **Report Generator** tab.
3. Select a monoclonal well of interest.



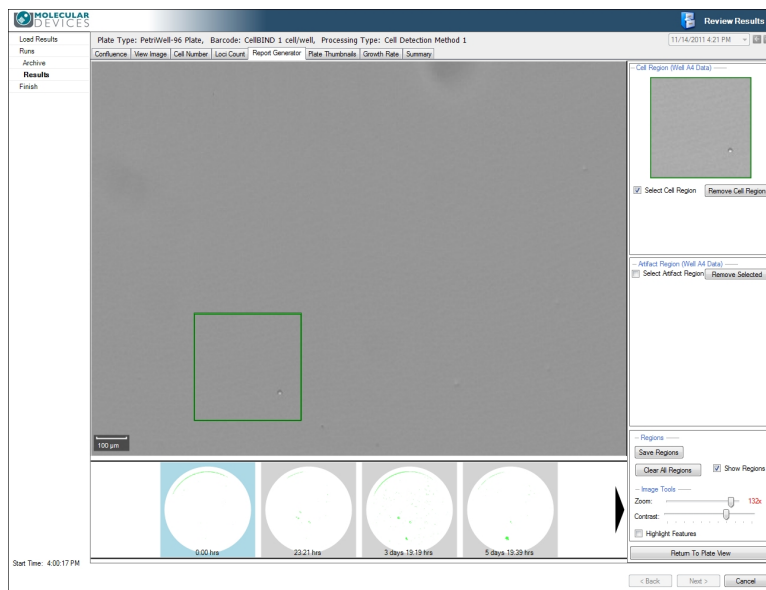
- In the timeline at the bottom of the window, select the **0.00 hrs** time point.



- In the upper right pane, under **Cell Region**, select the **Select Cell Region** check box to activate the cell region marker, move the green selector-box over the cell image and then click to apply the mark. Only one cell region can be marked and the green selector-box size is not adjustable.

The image shown in the right side **Cell Region** pane appears in your report.

*** Tip:** By default, the location of the first cell region remains fixed for all other time points. To adjust the location of the cell region box, click on a different time point thumbnail, select **Select Cell Region**, position the green box as needed, and click. Click on other time points and repeat the adjustment process as needed



*** Tip:** Deselect the **Select Cell Region** check box to end the marking process. Alternatively, cell region marking is automatically ended when you go to mark artifacts.

*** Tip:** To remove the cell region mark, click **Remove Cell Region**.

- To mark artifacts in the well, under **Artifact Region**, select the **Select Artifact Region** check box and then move the red selector-box over an artifact and click to apply the mark. Continue to move and click the red selector-box to mark up to 10 artifacts in the well. The selector-box size is not adjustable.

The images shown in the right side **Artifact Region** pane appear in your report.

*** Tip:** If you zoom in close and want to scroll around the well image during the artifact marking process, you must deselect the **Select Artifact Region** check box, click and drag to move within the cell view and select the **Select Artifact Region** check box again when you are ready to mark again.

*** Tip:** To remove an artifact region, on the right in the **Artifact Region** pane, select the artifact thumbnail that you want to remove and click **Remove Selected**. To remove multiple artifact regions, press <CTRL> while selecting the multiple artifact thumbnails, and then click **Remove Selected**.

- When you are finished marking regions, click **Save Regions**.

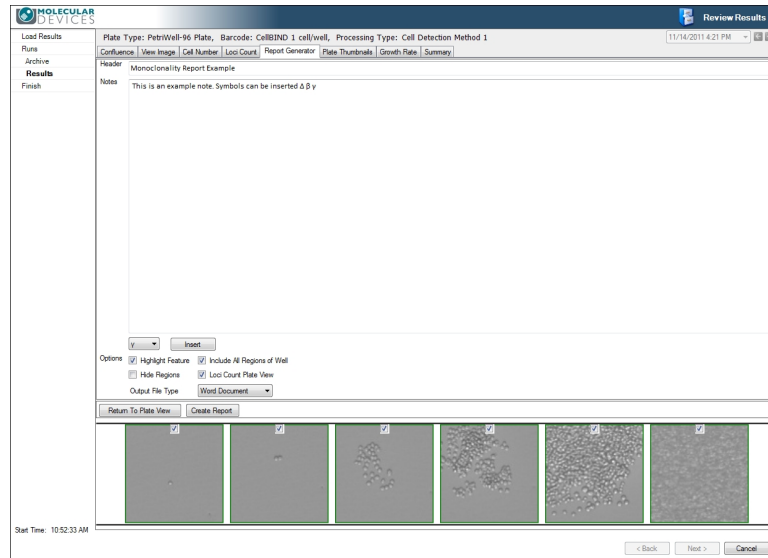
*** Tip:** The image **Contrast** setting is used in the generated report. If you batch generate reports, the last image **Contrast** setting is used in all of the reports.

- Click **Return To Plate View** to continue.
- In the right side pane, in the **Wells With Cell Regions** list, select the **Name** check box of the well of interest you marked with regions. Only wells with cell regions marked appear in the right side pane list.

*** Tip:** If you need reports for multiple wells of interest, select the **Name** check box for those too. Multiple well selections produce multiple reports with one well detailed per report.



10. Click **Generate Report** to open the report preparation screen.



11. In the report preparation screen you can adjust the following:

- **Header**—Type the title used in your report in this field.
- **Notes**—Type any additional information you need to include in your report in this field.

If you are generating multiple reports, whatever is typed here appears in all the reports.

To include symbols in your notes, use the drop-down list to select an available symbol and click **Insert**.

- **Options**—Select these check boxes as needed to determine how the content is presented in the report. Available options include:
 - **Highlight Feature**—When selected this enables the highlighting of the colony in your report.
 - **Hide Region**—When selected this excludes the cell and artifact region markings on the whole well image from your report.
 - **Include All Regions of Well**—When selected this includes all images from the well at single cell resolution.



Note: Generates a much longer report because all of the images are exported.

- **Loci Count Plate View**—When selected this includes the Loci Count Plate View in your report including well details and criteria settings.
- **Output File Type**—Enables you to select either **Word Document** output or **PDF Document** output for your report.

12. At the bottom of the screen, by default all of the time point thumbnails are selected to be included in the report. If needed, to exclude a time point thumbnail from your report, deselect the thumbnail check box.
13. To generate the report, click **Create Report**.
14. Specify where the report should be saved and click **OK**.
15. When the **Report generation success** message appears, click **OK**.
16. When finished, click **Return to Plate View**.

View Report

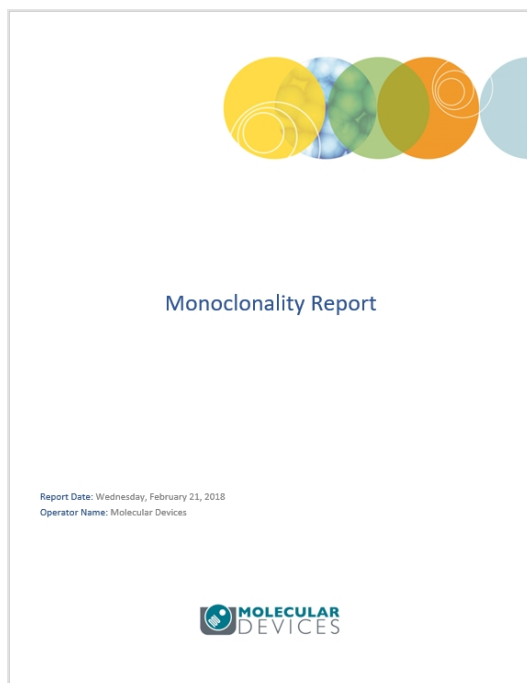
To view your report:

If you created a **Word Document** report, you must have a Word file format reader installed, or some Microsoft Word software installed, such as:

- Windows 10 users—Microsoft Word Viewer or Microsoft Word 2016
- Windows 7 users—Microsoft Word 2016

If you created a **PDF Document** report, you must have some PDF viewer software installed, such as Adobe Acrobat Reader.

Go to the location you specified when you clicked **Create Report** and double-click the file name.



*** Tip:** By default the Word document output is read-only. To edit a Word document, before opening the report, right-click on the file name, select **Properties** and deselect **Attributes > Read-only**.

Plate Thumbnails

The **Plate Thumbnails** tab shows the entire plate and the wells that have been imaged as thumbnails. The thumbnails include the confluence overlay of each of the imaged wells. Use this screen to select wells to be included or excluded from analysis, which helps to be focus only on wells of interest. Any wells included or excluded in the **Plate Thumbnails** tab automatically get included or excluded in the **Confluence**, **Cell Number**, **Loci Count**, and **Growth Rate** tabs as well.

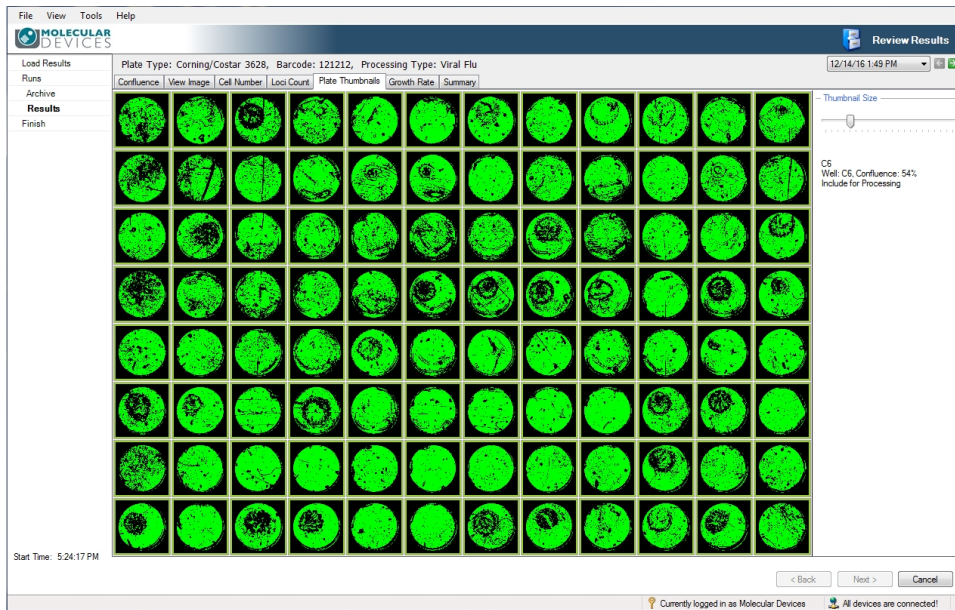


Figure 4-28: Plate Thumbnails tab screen

- A group well selection highlights in yellow.
- Excluded well selections highlight in red.
- Included well selections highlight in green.

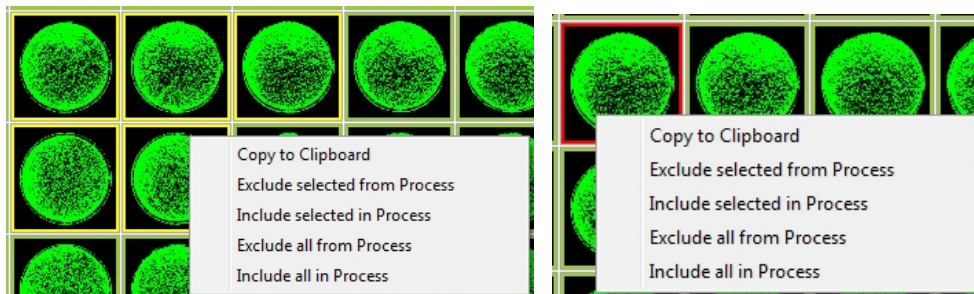


Figure 4-29: Group selected wells and excluded well with selection options menu

To specify what to do with each well for your analysis, do the following:

1. Hover the cursor over a well of interest.



Tip: To select a group of wells, press and hold **CTRL** and then select multiple wells. Release **CTRL** when you finish selecting.

2. Right-click on the well of interest, and then select one of the following from the options menu:
 - Copy to Clipboard
 - Exclude selected from Process
 - Include selected in Process
 - Exclude all from Process
 - Include all in Process

Cancel

If you click **Cancel**, you are prompted to confirm you want to end the process. Clicking **Yes** at the prompt takes you to the **Load Results** screen. See [Load Results on page 41](#).

To finish, see [Finish on page 83](#).

Growth Rate

If a microplate has been imaged previously, growth curves appear for all the wells in the plate. The system determines if the plate has been imaged by looking for the barcode in the stored results, so use barcoded microplates to enable this function. Named plates can also be used, but for the software to identify the series, the name must be typed exactly the same each time the plate is imaged. The ability to rename plates does risk the inclusion of mis-named plates in the growth curve data.

The captured images for the selected well appear in the well schematic below the graph.

To see the well coordinate for a point, hover the cursor over a data point.

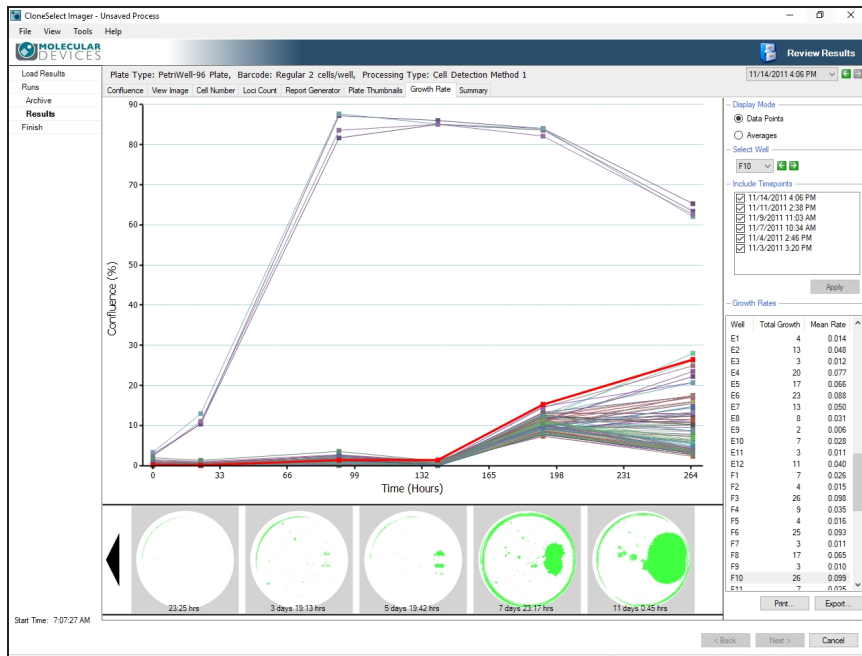


Figure 4-30: Growth Rate tab screen showing Data Points graph

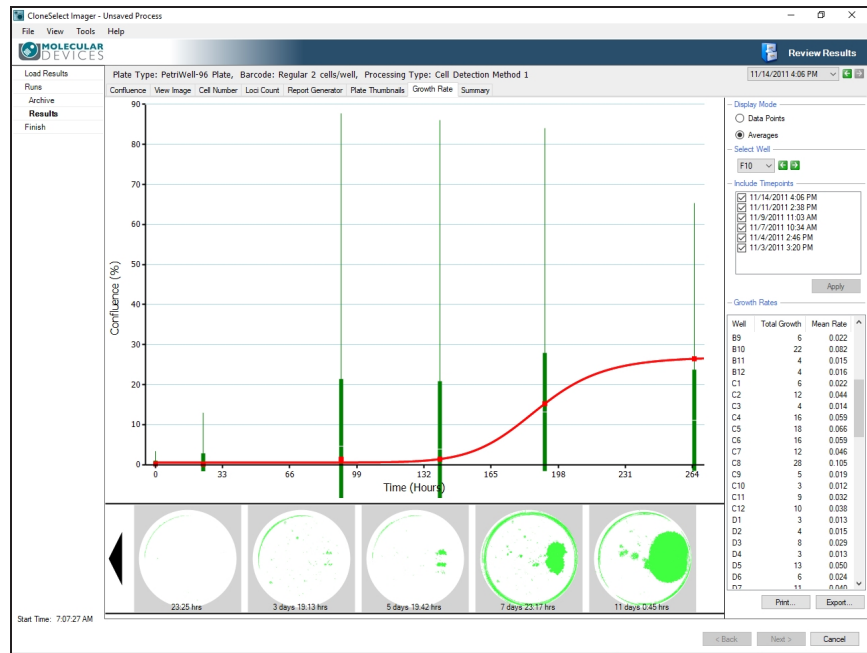


Figure 4-31: Growth Rate tab screen showing Averages graph

Display Mode

You can view the following growth rate graphs:

- **Data Points**—graphs all the well data points
- **Averages**—graphs the averages of the well data points.

The thick red line within both graphs shows the growth rate for the selected well.

Select Well

Use the menu or the arrows to select a different well.

Include TimePoints

Use the **Include Timepoints** list to select which time points get included or excluded from the growth rate output. All plate scans with the same barcode are automatically selected for inclusion in the growth rate data unless you deselect them here.

To change time point selections:

- Adjust the time point selections, and then click **Apply**.

Growth Rates

The **Growth Rates** list displays the **Well**, **Total Growth** and **Mean Rate** for each well in the data set.

Print

The **Print** option prints the growth curve graph.

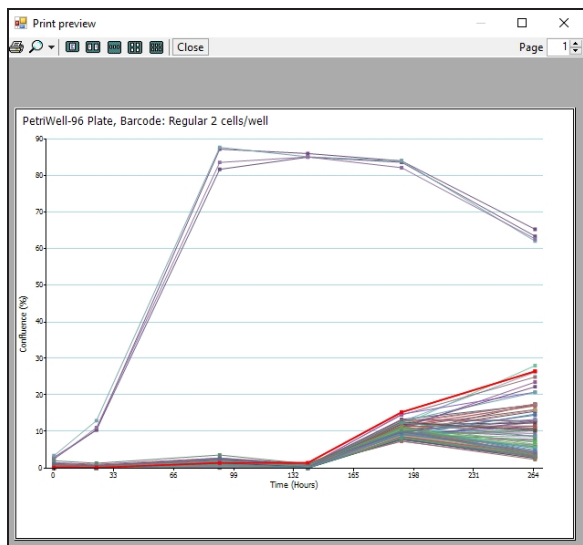


Figure 4-32: Growth Curve Graph Print Preview

Export

The **Export** option lets you save all the data used to construct the growth curve as a **Comma Separated Values (CSV)**, **Excel**, or **XML** file.

The **Data Export** wizard guides you through the process of exporting the **Confluence** data. **Cell Number** and **Loci Count** data can also be exported.

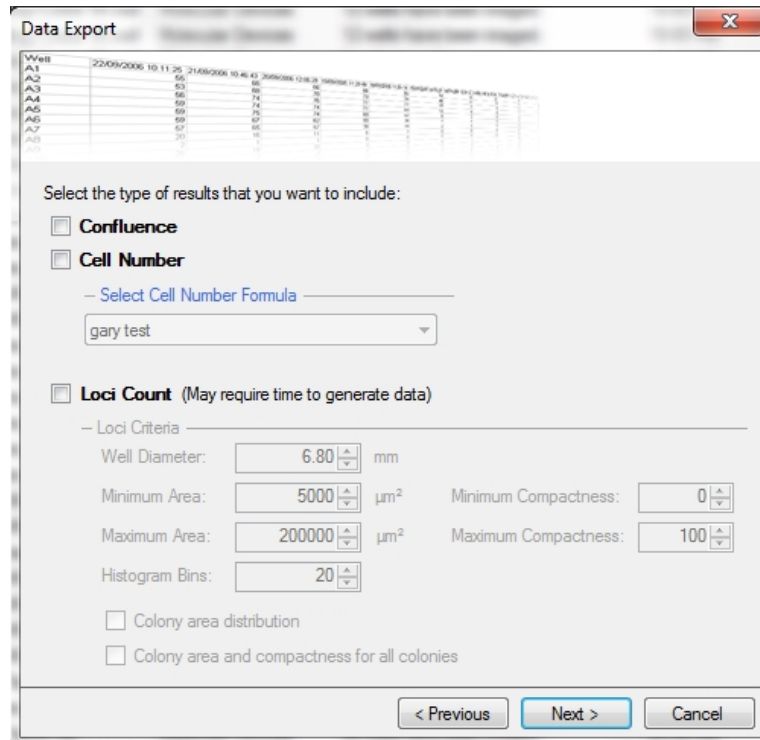


Figure 4-33: Data Export wizard dialog

The wizard provides the option to export the data for the following time points:

- The currently selected time
- The most recent time for each well plate
- The complete time series.

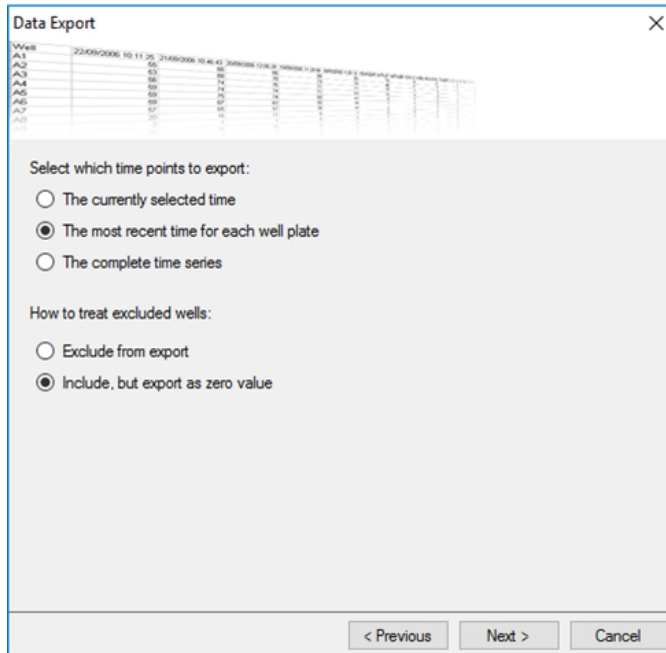


Figure 4-34: Data Export wizard with excluded wells options

When the option **Include, but export as zero value** is selected, the excluded wells are listed in the exported data with a zero value in the **Cell Number** column.

Well	Confluence	Cell Number	Loci Count
A1	0	0	0
A2	0	4	0
A3	0	0	0
A4	1	8	0
A5	1	9	0
A6	1	16	0
B1	1	9	0
B2	1	13	0
B3	1	9	0
B4	0	5	0
B5	2	19	0
B6	1	16	0

Figure 4-35: Example export data with Well A1 and Well A3 excluded

Cancel

If you click **Cancel**, you are prompted to confirm you want to end the process. Clicking **Yes** at the prompt takes you to the **Load Results** screen. See [Load Results on page 41](#).

To finish, see [Finish on page 83](#).

Summary

The **Summary** tab shows a summary of the confluence data, growth curve data and imaging credentials.

The table lists the **Date Imaged**, **Elapsed Time**, **Operator**, **Imaged on**, **Min Confluence**, **Mean Confluence** and **Max Confluence** for each time the plate was imaged.

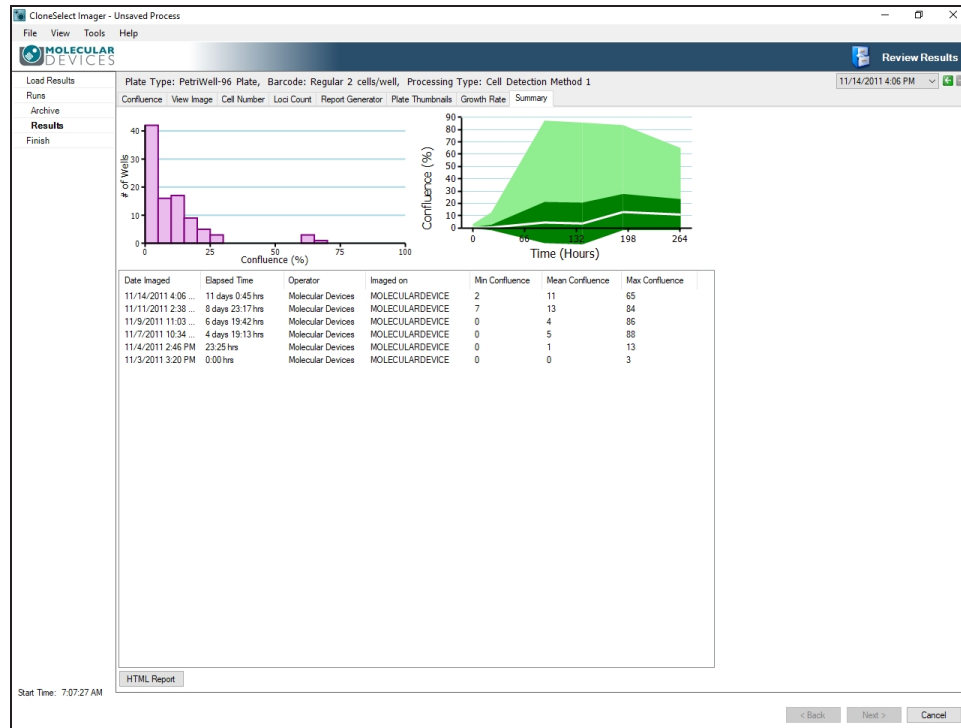


Figure 4-36: Summary tab screen

HTML Report

The **HTML Report** button lets you generate an HTML formatted report of the various information relating to the data that you select, such as confluence, growth rate, and cell number.

To create an HTML report:

1. In the **Review Results > Results > Summary** tab, click **HTML Report**.
2. In the **HTML Report Wizard** dialog, select the information you want included in the report.

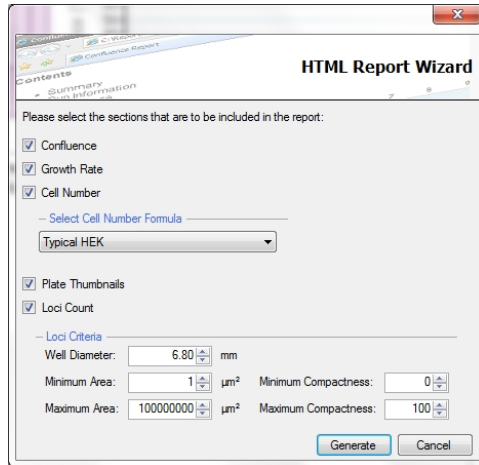


Figure 4-37: HTML Report Wizard dialog



Note: The loci data are automatically populated using the Minimum and Maximum Area and Minimum and Maximum Compactness data set in the **Loci Count** tab. See [Loci Count on page 52](#).

3. To save the report as an .html file, click **Generate**.

Cancel

If you click **Cancel**, you are prompted to confirm you want to end the process. Clicking **Yes** at the prompt takes you to the **Load Results** screen. See [Load Results on page 41](#).

To finish, see [Finish on page 83](#).

Migration Analysis



Note: (Optional) You must purchase a license for this software module.

Do Migration analysis on data that has been imaged over a number of time points.

To view the data for a migration assay:

- In the **Results Archive** list, double-click on the run date you want to use, and then click **Migration**.

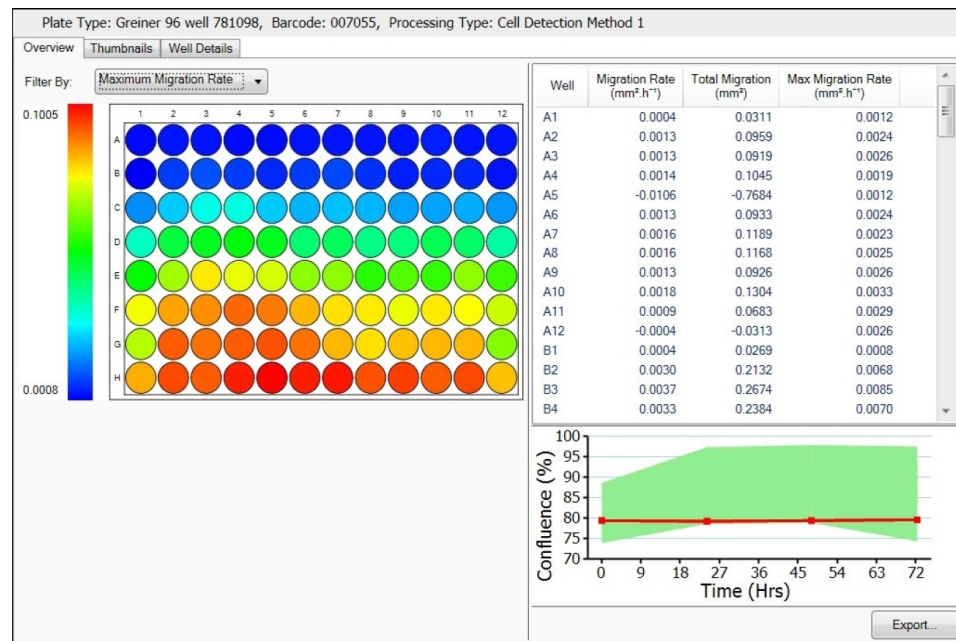


Figure 4-38: Migration analysis Overview screen

The Migration data analysis screen includes three tabs:

- [Overview on page 78](#)
- [Thumbnails on page 79](#)
- [Well Details on page 80](#)

For each data set analyzed, the **Plate Type**, the **Barcode**, and the **Processing Type** information appears at the top of the screen for reference.

Overview

The **Overview** tab provides an overview of the migration assay data. A heat map is generated and can be displayed using one of three **Filter By** menu options:

- **Migration Rate**—measures the rate of increase in cell movement (mm^2 per hour) over the entire time period.
- **Total Migration**—measures the increase in area of cell movement (mm^2) over the entire time period.
- **Maximum Migration Rate**—measures the largest rate of increase in cell movement (mm^2 per hour) between any two adjacent time points.

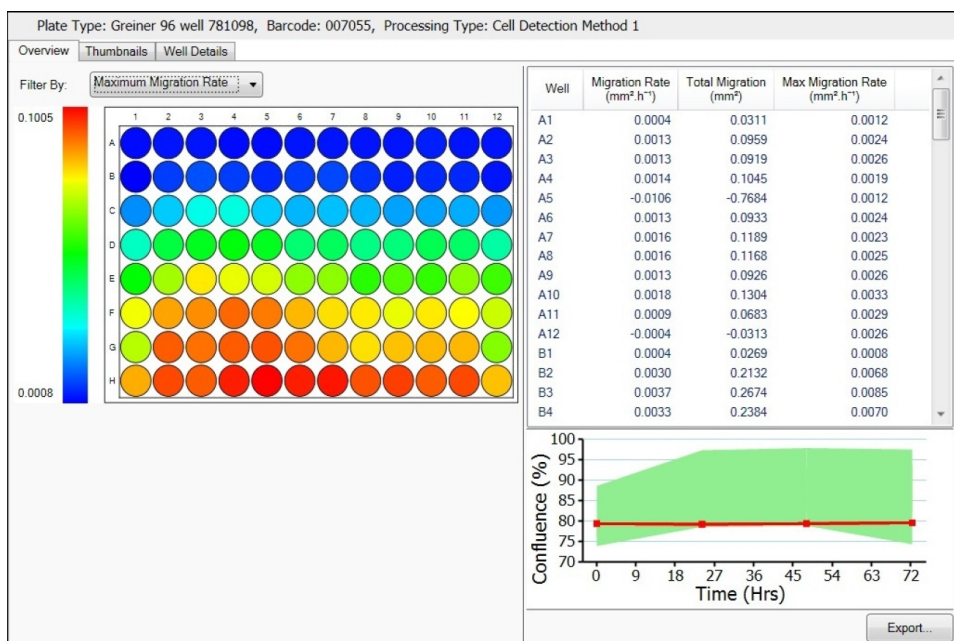


Figure 4-39: Migration analysis Overview screen

The table to the right of the heat map also displays these statistics for each well.

To reorder the data, click on any of the table headers.

To copy the heat map to the clipboard, right-click anywhere on the map and select **Copy to clipboard**.

To highlight the well on the information table and on the Confluence (%) graph, click on any well of interest.

To view a well in the **Well Details** tab, double-click on the well of interest.

Export

The **Export** option lets you save data for either **Selected Time Points** or for **All Time points**. You can also export the confluence data if required.

The **Data Export** wizard guides you through the process of exporting. The data can be exported as a **Comma Separated Values (CSV)**, **Excel**, or **XML** file.

Thumbnails

The **Thumbnails** tab provides an overview of each well image with wound detection, where no cells are detected.

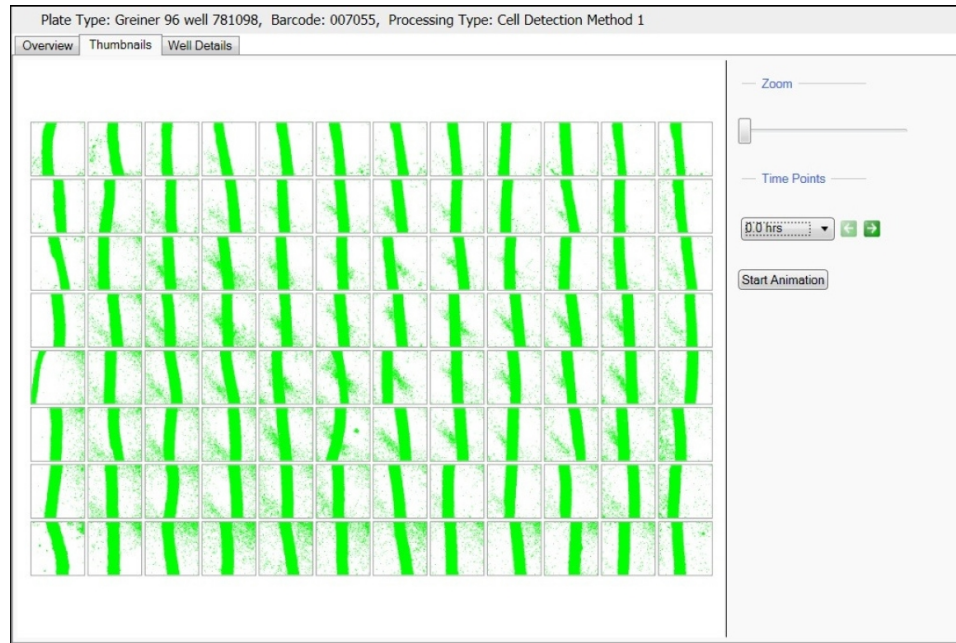


Figure 4-40: Migration analysis Thumbnails screen

To copy the thumbnails to the clipboard, right-click anywhere on the thumbnail map and select **Copy to clipboard**.

The following controls are available:

- **Zoom**—Move the slide either left or right to zoom out or for the whole thumbnail map.
- **Time Points**—Use the drop down menu or the arrows to select a different time point.



Note: If the time points available are restricted by the **End Time Point** option in the **Display Graph** section of the **Well Details** tab, only the selected time points appear here.

- **Start Animation**—To observe the migration taking place in the wells, click this button to animate the thumbnails from the starting time point to the final time point.



Note: If the time points available are restricted by the **End Time Point** option in the **Display Graph** section of the **Well Details** tab, only the available time points animate here.

Well Details

The **Well Details** tab provides all the details generated from individual wells. A whole well image appears center screen with time point thumbnails in the film strip below.

To display the enlarged image for a time point in the film strip below, click on any of the thumbnails.

By default **Well Details** opens in the **Display Image** view. You can also view as a graph by clicking **Display Graph** in the right side screen controls.

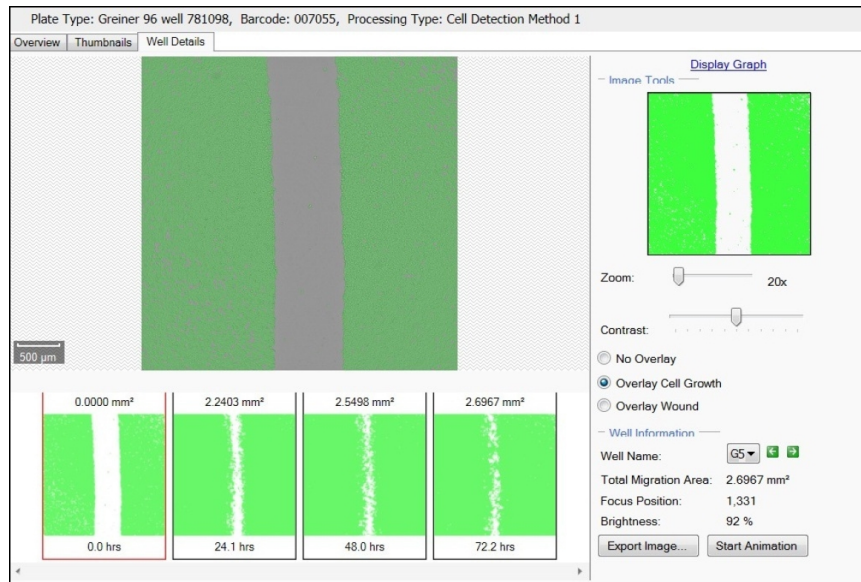


Figure 4-41: Migration analysis Well Details screen

Display Image

The **Display Image** controls include the following:

- **Image Tools**

Image Tools include the following:

- **Zoom**—Move the slide either left or right to zoom out or in. The lowest magnification of the image is 18x and the highest is 144x. When the figure turns red the system is zooming digitally and can cause some pixelation of the image. When zoomed into an image, the zoomed area will be displayed on the image thumbnail to the right.
- **Contrast**—Move the slider either left or right to improve the image contrast.
- **No Overlay**—Select to hide all overlay highlights.
- **Overlay Cell Growth**—Select to highlight in green all areas with cells.
- **Overlay Wound**—Select to highlight in green all areas without cells.

- **Well Information**

Well Information include the following:

- **Well Name**—Use the drop down menu or the arrows to select a different well.
- **Total Migration Area**—Shows as a value the measurement of the total cell growth in mm^2 over the entire time period for the selected well.
- **Focus Position**—Shows the focus position used for the microplate.
- **Brightness**—Shows the brightness value used for imaging.
- **Export Image**—Lets you save the currently displayed image in .bmp, .jpg, or .png format.
- **Start Animation**—To observe the migration taking place in the wells, click this button to animate the thumbnails from the starting time point to the final time point.



Note: If the time points available are restricted by the **End Time Point** option in the **Display Graph** section of the **Well Details** tab, only the available time points animate here.

Display Graph

The Display Graph view shows the information gathered from the individual wells and displays it in graph format. The graph appears center screen with Migration Area (mm^2) plotted against Time (hours). A time point thumbnail of the selected well, denoted by a red line on the graph, appears below the graph. Each thumbnail image in the film strip displays the total migration at that time point and time passed since the first time point in the series. When hovering over a data point on the graph for the selected well (denoted by a red line on the graph), the corresponding thumbnail will be highlighted in red.

To return to the image view, click **Display Image** in the right side screen controls.

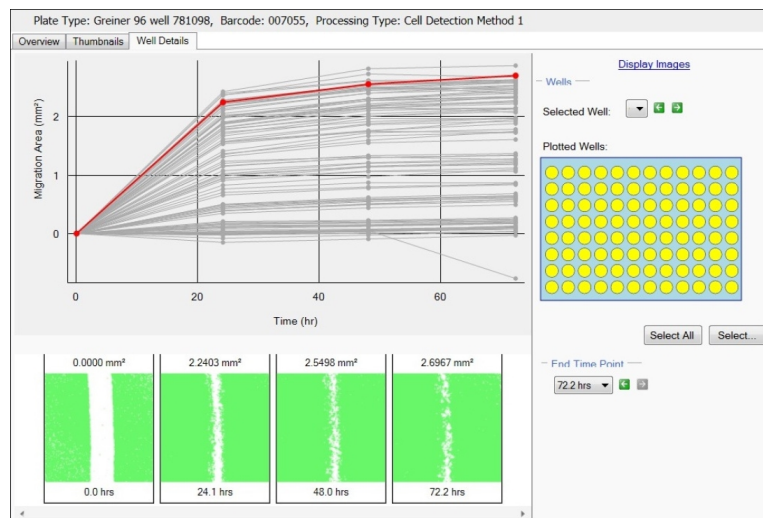


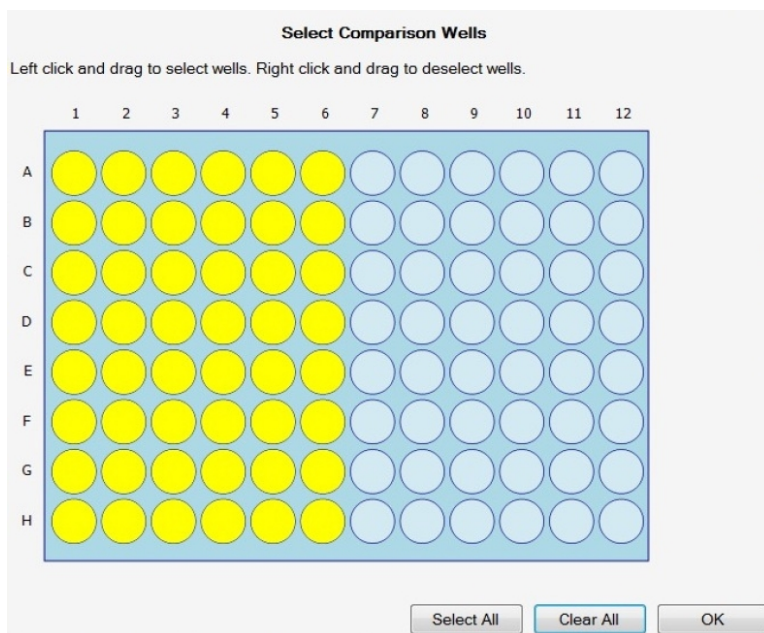
Figure 4-42: Migration analysis Well Details Display Graph screen

The **Display Graph** controls include the following:

- **Wells**

Wells include the following:

- **Selected Wells**—Use the drop down menu or the arrows to select the well currently displayed on the graph as a bold red line.
- **Plotted Wells**—Use to select the wells for plotting on the graph. The selection and deselection of these wells only apply to the graph and not the overview. Shows on a mini plate map all of the wells that were imaged and are plotted on the graph. When wells are selected they are yellow in the plate map. When wells are deselected they are blue in the plate map.



- **End Time Point**—Use the drop down menu or the arrows to select an end time point. When the end time point is selected the graph shows only the time points before and including the end point selected.



Note: When changing the **End Time Point**, only the selected time points appear on the **Thumbnails** tab. Data on the **Overview** tab is also recalculated using the selected time points.

Finish

When you finish the **Review Results** process, you return to the **Review Results** process start screen. See [Review Results on page 40](#).

To finish the **Review Results** process from a results tab:

1. In any review results tab, click **Cancel**. See [Analyzing Results on page 44](#).
2. In the **Load Results** screen, click **Cancel**. See [Load Results on page 41](#).
3. In the **Finish** screen, click **Finish**.

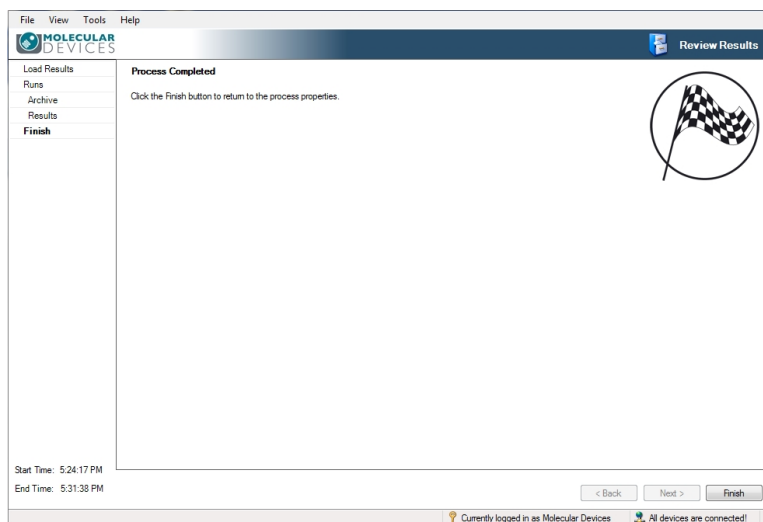


Figure 4-43: Review Results process Finish screen

Moving Images

You can move acquired images from the local system computer. The default location to which imaging results save is `c:\image archive\`. For more imaging results details, see [Load Results on page 41](#).

Batch Microplate Imaging

Batch Microplate Imaging enables you to image multiple plates with the same configuration without reviewing the focus and alignment between plates.

To enter a Batch Microplate Imaging process, in the Navigation screen, click **Batch Microplate Imaging**. See [Navigation Screen on page 21](#).

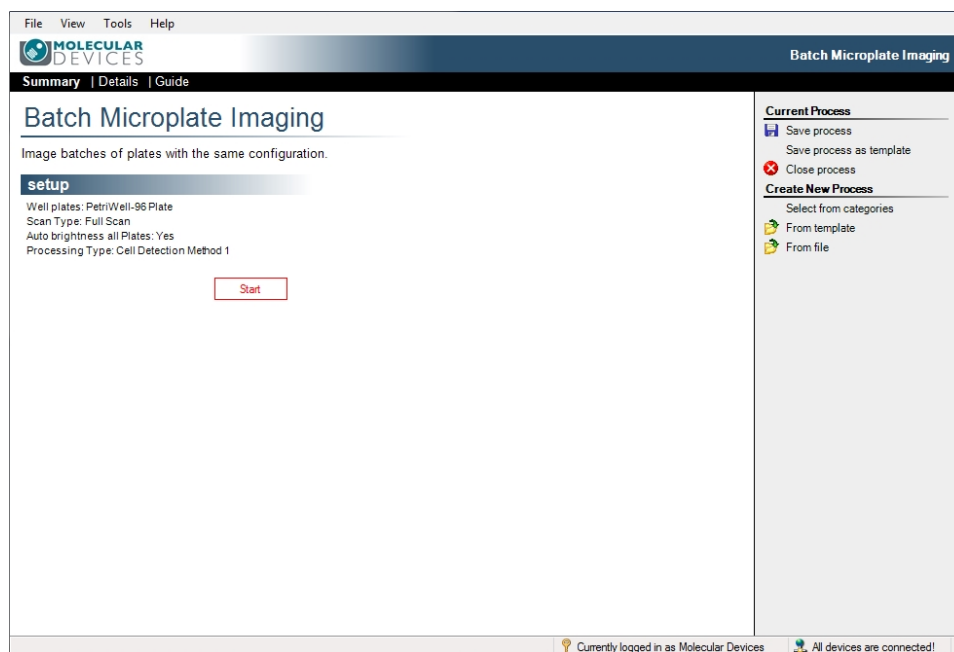


Figure 4-44: Batch Microplate Imaging Summary screen

To set the properties for the Batch Microplate Imaging process, the process screen opens in the Process Properties screen in the **Summary** view. Change the screen view, as needed, and change properties as needed. See [Process Properties Screen on page 24](#). Alternatively, you can click the **Setup** header, and then select from the available properties.

The **Auto brightness all Plates** option automatically selects the appropriate brightness for the image.

This process automatically looks for barcodes on plates, but if not detected, you are prompted to manually enter a plate barcode or plate name.

To begin the process, click **Start**. The first step of the Batch Microplate Imaging process is called **Select Source**. This and the remaining process steps are the same as the Image Microplate process. See [Image Microplate on page 25](#).

E-Scope

The **E-Scope** process lets you view cells at a high resolution (1.85 μm) without performing any image analysis. Images are not saved.

Frames to be imaged can be freely selected by clicking with the mouse on the screen.

To enter an **E-Scope** process, in the Navigation screen, click **E-Scope**. See [Navigation Screen on page 21](#).

How to Image Using E-Scope



Note: Images are not saved.

To image using E-Scope:

1. To set up the properties screen, click the **Source** header, and then select from the available plates.

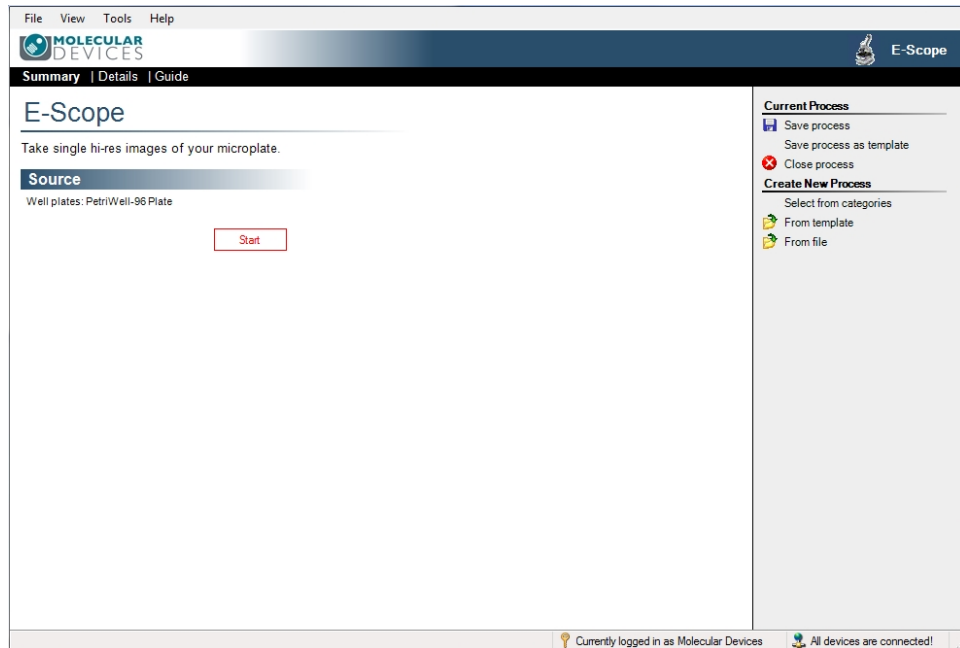


Figure 4-45: E-Scope Summary screen

2. To begin the process, click **Start**.
3. When the plate carrier ejects, place your microplate on the carrier with the A1 well in the back right corner, and then click **Next to Load Plate**.

- Use the **Alignment** screen to check that the camera aligns properly with the wells of the microplate. Four images are taken over the north, south, east and west edges of well A1. The well edges should align with the middle of the box formed by the four red lines. Adjust as needed.

- If the well edges do not fall in the middle of the box formed by the four red lines on screen, click and drag on the lines to correct the alignment.
- If needed, you can also adjust the image brightness by moving the **Brightness** slider to the percentage value you want to use.



Note: Brightness changes only for the alignment image.

- When the button is pink, click **Re-image** to recheck the alignment.
 - To finish Alignment and continue, click **Next**.
- In the **Select Location** screen, in the displayed schematic of the microplate, to view a real-time image, click on one of the available image frames.

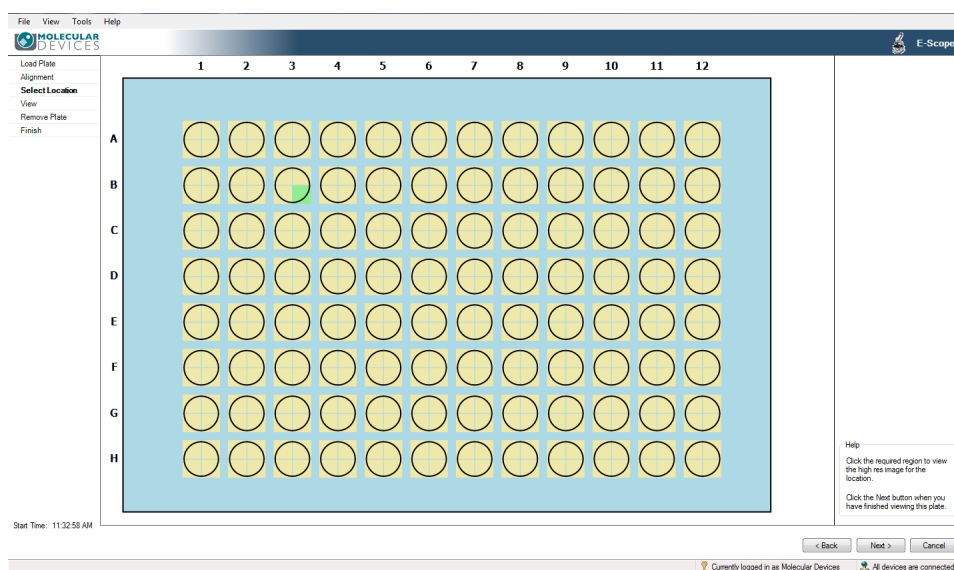


Figure 4-46: E-Scope Select Location screen

6. The **View** screen opens automatically with the selected image frame.

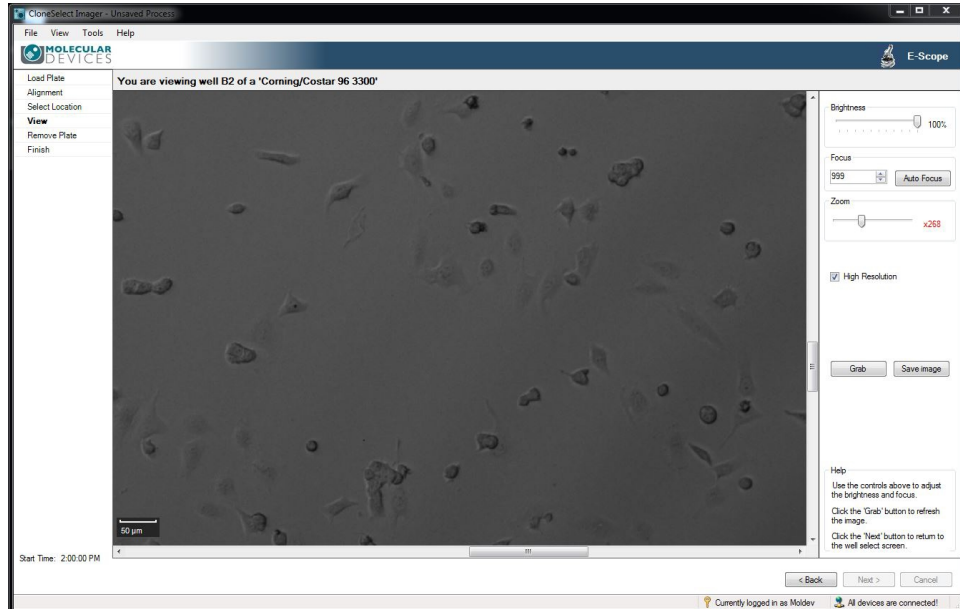


Figure 4-47: E-Scope View screen

You can do any of the following in the **View** screen:

- **Brightness**—Move the slider either left or right to improve the image brightness.
- **Focus**—Click the up or down arrows to change the focus value, or type a new value in the field.
- **AutoFocus**—Click to automatically adjust the image focus.
- **Zoom**—Move the **Zoom** slider to look closer or farther at the image. As you change the zoom level, the Scale Bar in the lower left of the image window automatically adjusts.

*** Tip:** The figure turns red when there is distorting image pixelation.

- **High Resolution**—Select to use high resolution imaging (1.85 µm), deselect to use standard resolution imaging (3.7 µm).
 - **Grab**—Click to apply the changes you made while viewing the image.
 - **Save Image**—Click to save the current image in bmp, jpg or png format.
7. To select another image or well, click **Back** to go back to the **Select Location** screen. See step 5.
 8. To continue, click **Next**.
 9. When the plate carrier ejects, remove your plate, and then click **Next**.
 10. In the **Finish** screen, click **Finish** to return to the E-Scope process start screen. See step 1.

Eject Plate Carrier

The **Eject Plate Carrier** process is one of the **Maintenance Processes** on the Navigation screen. See [Navigation Screen on page 21](#). It enables a way to eject the plate carrier for cleaning or if a microplate has been left in the instrument.

To eject the plate carrier:

1. To enter the **Eject Plate Carrier** process, in the Navigation screen, click **Eject Plate Carrier**.
2. In the **Eject Plate Carrier** start screen, click **Start**.
3. Clean the plate carrier or remove a microplate.
4. To close the plate carrier and continue, click **Next**.
5. In the **Finish** screen, click **Finish** to return to the Eject Plate Carrier process start screen.

Transport and Storage

The CloneSelect Imager must be:

- Stored and transported in temperatures within the range -25°C to $+55^{\circ}\text{C}$.
- Transported with the original packing foam inserts in place to protect the objective and objective holder.

Lifting Points

The CloneSelect Imager is intended for bench top operation. Care should be taken when moving the CloneSelect Imager. It is recommended that 2 people lift it.

External Covers



WARNING! If any of the external covers on the CloneSelect Imager are removed the power supply is not automatically interrupted. If it is necessary to remove any of the external covers you must ensure that the power is switched off first and do not attempt to use the robot until the covers are replaced.

Electrical Safety



WARNING! HIGH VOLTAGE. The CloneSelect Imager must be connected to a properly earthed power outlet to protect users from the risk of electric shock. The main chassis of the machine is earthed together with all associated electrical components. Do not remove any of the fixed covers, as there are no user serviceable parts inside. All internal work should be referred to Molecular Devices approved service personnel.

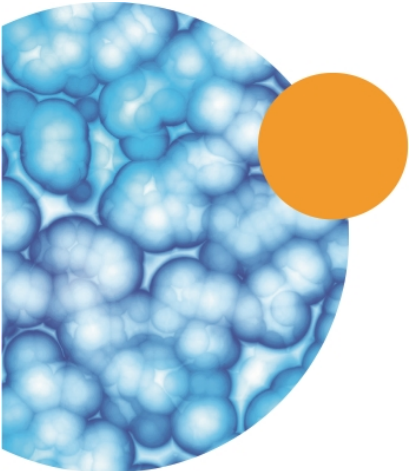
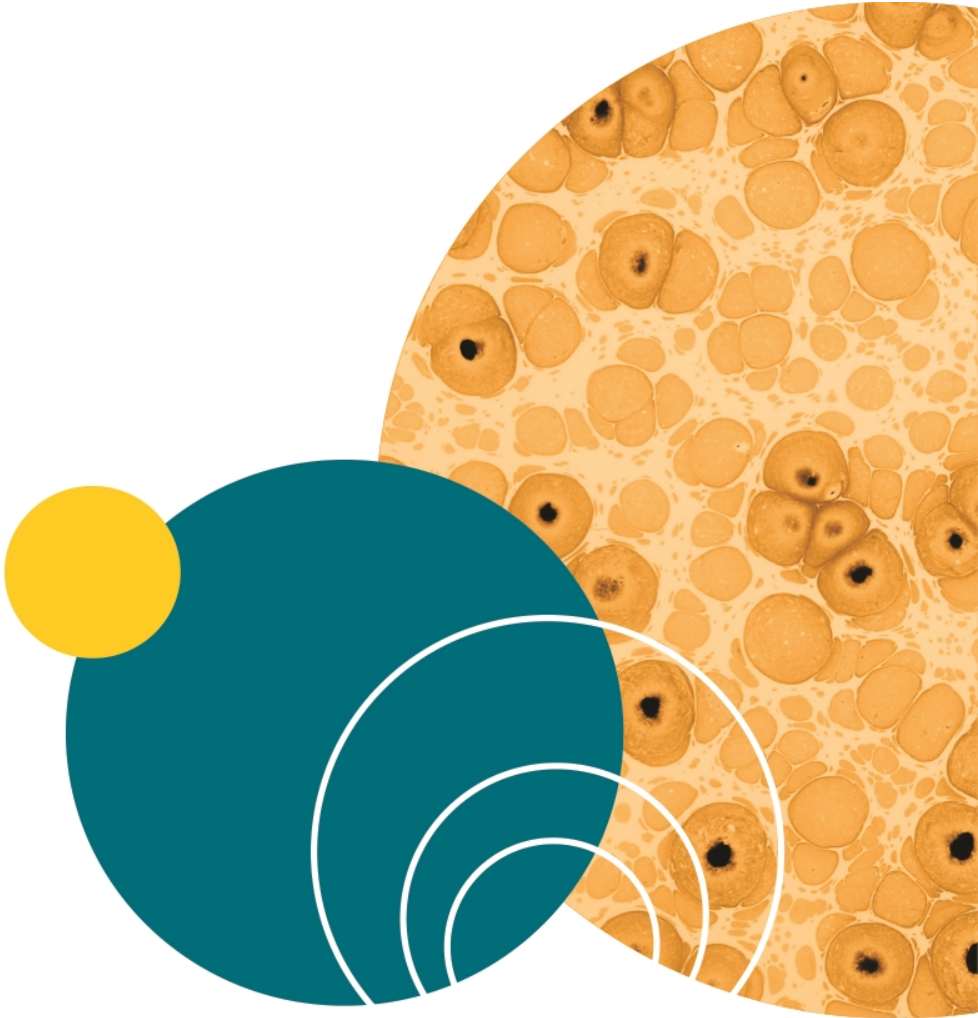
Drive Safety



WARNING! PINCH HAZARD. There is a potential pinch hazard with the drive mechanism. Please ensure that you do not attempt to load the plate until the drive has fully extended.

Noise levels

During normal operation the level of airborne noise emitted by the robot will not exceed 70 db measured at 1 meter.



Do only the maintenance described in this guide. Maintenance procedures other than those specified in this guide must be done by qualified Molecular Devices personnel only. See [Obtaining Support on page 101](#).

Maintenance procedures that you can do to ensure optimal operation of the instrument include:

- [Doing Preventive Maintenance](#)
- [Cleaning the Instrument on page 92](#)



WARNING! Service or maintenance procedures other than those specified in this guide can be done only by Molecular Devices qualified personnel. When service is required, contact Molecular Devices Technical Support.

Doing Preventive Maintenance

You are responsible for doing daily and weekly maintenance. Also, Molecular Devices strongly recommends that a complete instrument preventative maintenance service be done annually by Molecular Devices.

General Precautions

- All waste must be disposed of according to local regulations.



WARNING! Do not use in explosive environments.

Daily Maintenance

- Keep the CloneSelect Imager instrument dirt-free and dust-free.
- Clean the top and bottom of the removable glass plate holder using a lint-free cloth sprayed with 80% ethanol.

Weekly Maintenance

- Check operation of equipment.
- Check for damage.

Annually Maintenance

A complete instrument maintenance should be done annually by an approved service engineer. To obtain a maintenance contract or schedule a service visit, contact your Molecular Devices representative or technical support www.moleculardevices.com/service-support.

Cleaning the Instrument

Clean the instrument using 80% ethanol or dilute detergent and a lint-free cloth.



CAUTION! Do not use organic solvents and abrasive cleaners because they can damage the cover.

Apply cleaning solution using a suitable lint-free cloth.



CAUTION! Do not pour cleaning solution directly on to the instrument or other objects.

Upgrading Software



Note: Molecular Devices is responsible for installing the CloneSelect Imager Software during your initial instrument installation and configuration.

The CloneSelect Imager Software controls the CloneSelect Imager instrument and is based around the concept of the instrument running processes.

The following procedures are also located in the *Software Release Notes*. The following procedures are required to upgrade the CloneSelect Imager Software:

- [Computer System Requirements](#)
- [Backing Up the Configuration File on page 93](#)
- [Uninstalling the Current the Software on page 94](#)
- [Installing Microsoft .Net 4.5.2 on page 94](#)
- [Installing the CloneSelect Imager Software Upgrade on page 95](#)
- [Obtaining a License for the Software on page 95](#)

Computer System Requirements

The CloneSelect Imager Software requires the following computer specifications:

Table 6-1: Minimum Computer System Requirements

Item	Description
Operating system	Windows 10, 64-bit Windows 7, 32-bit (x86)
Memory	4 GB RAM
Data Connection	10/100 Ethernet port
Camera Connection	USB 2.0 port



CAUTION! Do Not replace or upgrade the computer operating the system with one of your own computers. The computer supplied with your system includes hardware components specifically configured to control your instrument.

Backing Up the Configuration File

Before removing the previous installation of the software, you must back up the configuration in case you need to reinstall it later.

The recommended process is two steps:

1. [Add a Windows System Restore Point](#)
2. [Back up CloneSelect Imager Software](#)

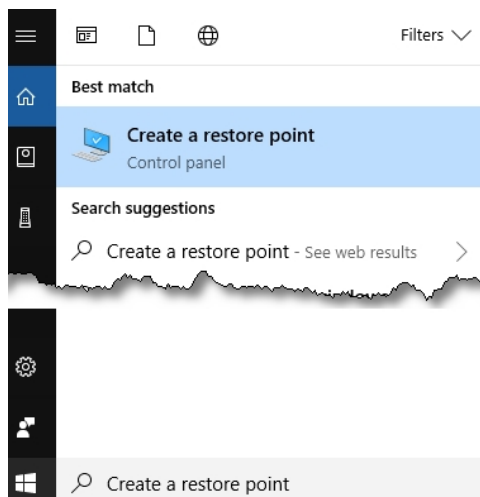
Add a Windows System Restore Point

If you are using the Windows 7 operating system, do the following:

1. Click **Start > Control Panel > System and Security**, and then click the **System** link.
From the **Start** menu, you can also get to the **System** screen by typing **SYSTEM** in the **Search** field, and then click **System** from the results list.
2. In the left panel, click the **System Protection** link.
3. In the **System Properties** dialog, click the **System Protection** tab and then click **Create**.
4. In the **Create a Restore Point** dialog, choose a name that you will remember, such as the current date or the name of a program you are about to install.
5. When the message **The restore point was created successfully** appears, click **Close**.
6. In any other related open dialogs, click **Close** to finish.

If you are using the Windows 10 operating system, do the following:

1. In the **Start** search field, type **CREATE A RESTORE POINT**, and select **Control panel**.



2. In the **System Properties** dialog, in the **System Protection** tab, select the main system drive, and click **Create**.
3. Type a description to identify the restore point.
4. Click **Create**.
5. When the message The restore point was created successfully appears, click **Close**.
6. In any other related open dialogs, click **Close** to finish.

Back up CloneSelect Imager Software

To back up your current configuration:

1. Click **Start > All Programs > Molecular Devices > CloneSelect > Configuration Manager**.
2. In the **Fusion Configuration Manager**, click **Backup To File**.
3. In the **Backup Fusion Configuration** dialog, type the version number of the currently installed software and the date and then click **OK**.
4. In the **Save Configuration File** dialog, locate and select the folder where you want to save the backup file, type a **File name**, and then click **Save**.

Uninstalling the Current the Software

To uninstall the current software:

1. Click **Start > Control Panel**.
2. Under **Programs**, click **Uninstall a program**.
3. From the list of programs, click CloneSelect Imager.
4. Click **Uninstall**.
5. Follow the on-screen instructions to finish uninstalling the software.

Installing Microsoft .Net 4.5.2

Windows 10 computers should already have Microsoft .Net 4.6 or newer installed. If your system has Microsoft .Net 4.5.2 or newer installed, skip the following procedure.

To install Microsoft .Net 4.5.2 software:

1. Download the Microsoft .Net 4.5.2 software from the Microsoft website.
2. Run the installer and follow the on-screen instructions to finish installing the software.



CAUTION! If you have a custom software configuration, contact Molecular Devices Technical Support to determine if you can upgrade your software. See [Installing Microsoft .Net 4.5.2 on page 94](#).

Installing the CloneSelect Imager Software Upgrade

To install the software upgrade:

1. Download the upgrade installation files to the system computer.
2. Double click the **CloneSelect Imager v.1.6.x.msi** file to start the installation program.
3. Follow the on-screen instructions to finish installing the software upgrade.
4. Select **Setup Type: Typical**.
5. Contact Molecular Devices Technical Support for the upgrade license. See .

Obtaining a License for the Software

The first time that you start the your Molecular Devices software after completing the installation, the software prompts you for a license.

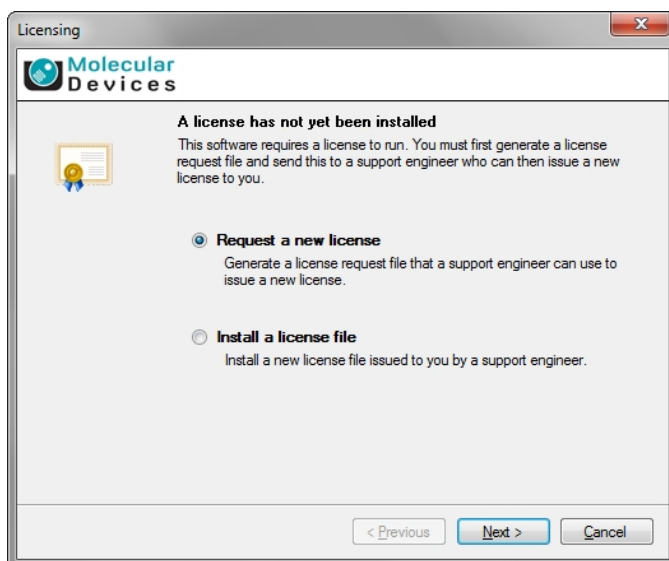
Obtaining a software license is a two-part process involving the following:

- [Requesting a Software License on page 95](#)
- [Installing a Software License on page 97](#)

Requesting a Software License

To request a new software license:

1. In the license message dialog, click **Yes**.
2. In the license request wizard, select **Request a new license**, and then click **Next**.



3. Enter the requested information, and then click **Next**.

Licensing

Molecular Devices

Please provide the following details. This will help our support engineer to create a license for you.

Registered User Name*: User

Company/Institute Name*: Company

Instrument Serial Number (optional):

Notes/Comments:

*These fields are required

< Previous Next > Cancel

4. To save the request file to a location on your computer for attaching it to an email, click **Save**.

Licensing

Molecular Devices

Click the 'save' button to store the license request into a file. Take note of where you save the file to so that you can locate it later.

Save...

< Previous Next > Cancel

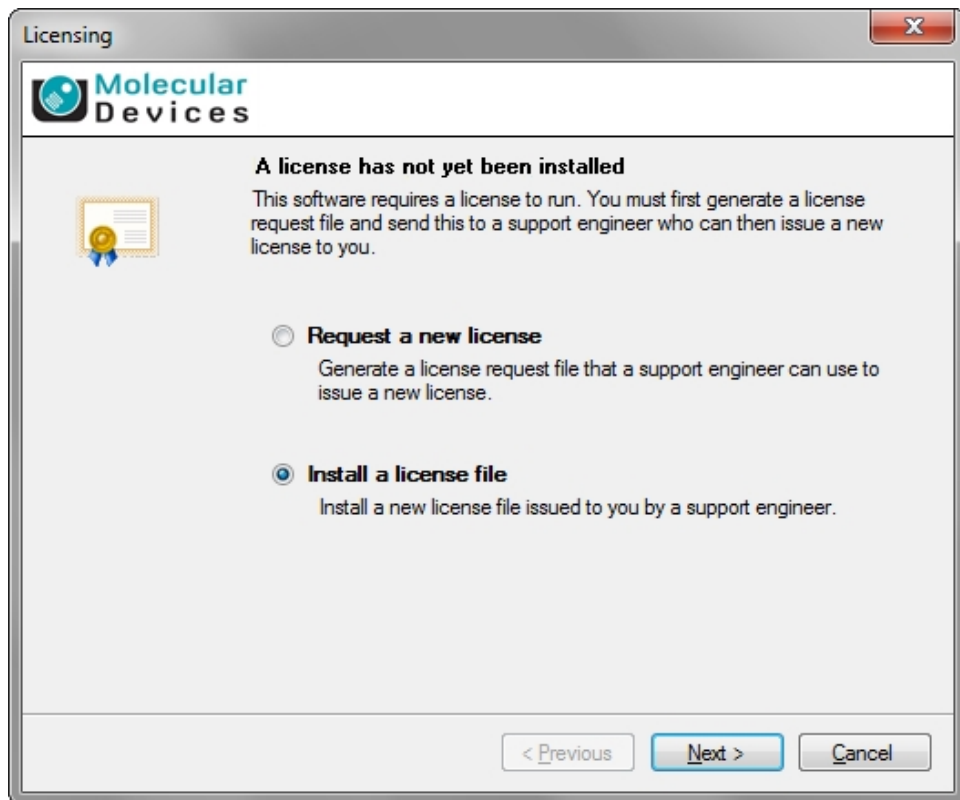
5. In the **Save the request to a file** dialog, save the file in a location where you can easily find it.
6. To close the wizard, click **Finish**.
7. Change the **LicenseRequest.req** file name to a **LicenseRequest.txt** file, and then email it to Molecular Devices Technical Support. See www.moleculardevices.com/service-support.
8. After you receive the license file, save it on the system computer where it is easily accessible, and then install the license.

Installing a Software License

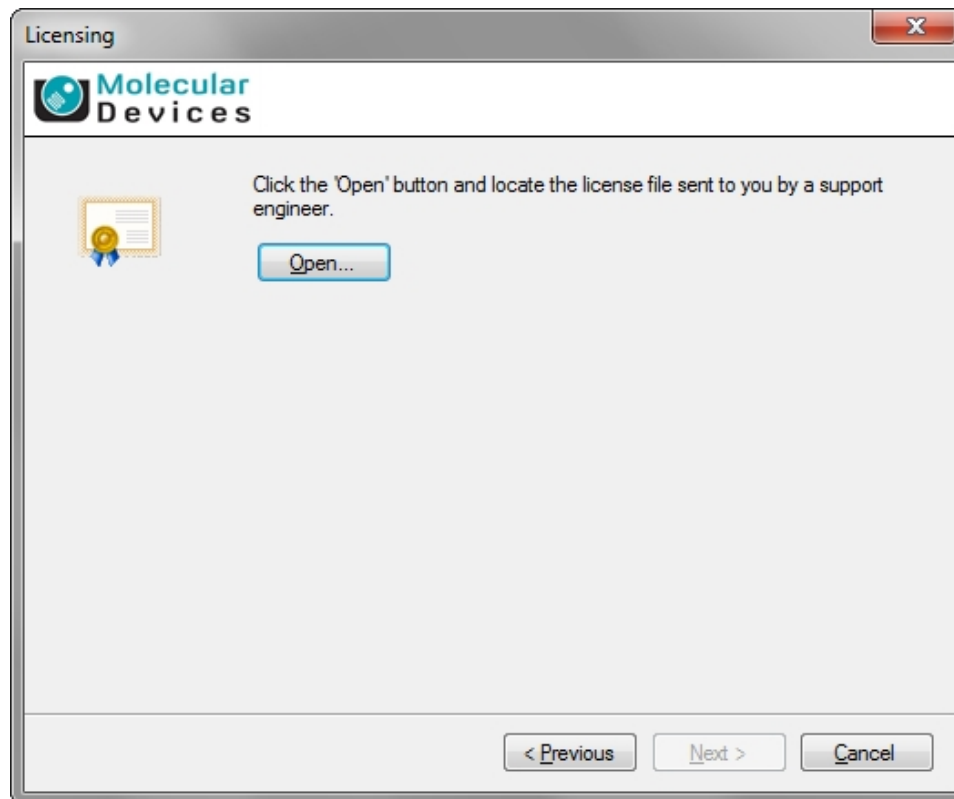
After you receive your license from Molecular Devices Technical Support, do the following:

To install a software license:

1. From the computer desktop, double-click the CloneSelect Imager icon to start the software.
2. In the **Licensing** dialog, select **Install a license file** and then click **Next**.



3. Click **Open**.



4. In the **Select the License File to install** dialog, locate and select the license file you previously saved, and then click **Open**.
5. After the license file automatically installs, click **Finish**.

This section explains how to resolve issues that you might experience when you work with the CloneSelect Imager and software.

Troubleshooting Barcodes

If you are using barcodes and they are not detected correctly, try the following:

- Rotate the plate 180 degrees.



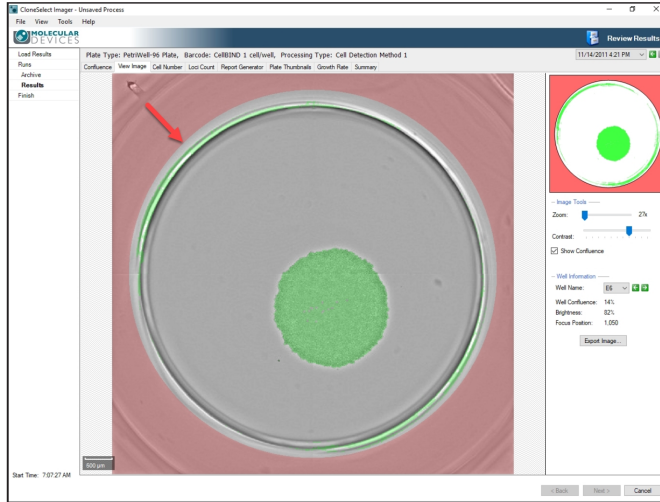
Tip: The instrument scans from either the front or the back, but only one sensor is active. Contact Molecular Devices Technical Support if you need to change the scan direction on your instrument.

- If one out of 10 plates fails to be read, then this might be a placement issue that can be fixed with a small change in the plate configuration.
- Add more white space at beginning or end.
- Restrict numbers to a maximum of six (6) digits.
- Verify that the codes meet the requirements. See [Using Barcodes on page 18](#).

If detection is still a problem, contact Molecular Devices Technical Support www.moleculardevices.com/service-support.

Troubleshooting Well Detection Area

If the detection area is outside the well, change the detection area by changing the plate parameters.



To change the well detection area plate parameters:

1. In the software select **Tools > Configuration**.
2. When the warning appears, click **Yes**.
3. In the **Edit Configuration** dialog, select **Consumables**.
4. Expand the **Microplates** tree.
5. Locate your plate and modify the following values as needed:
 - a. **WellBottomDiameter** changes the size of the red overlay. Decreasing this number makes the outcut in the red overlay smaller.
 - b. **A01** offset shifts the hole up, down, left, or right.
 - c. **Pitch** changes the distance between one well and the next, center to center.
 - d. To finish, click **Close**.

Well Dimensions		
WellA1CentreLengthOffset	13.63	Enter A01 Column offset in mm
WellA1CentreWidthOffset	10	Enter A01 Row offset in mm
WellBottom	Flat	Select Flat for flat bottom, Round for U bottom plates
WellBottomDiameter	6.3	Enter Well Bottom diameter in mm
WellBottomThickness	0.8	Enter thickness of the well bottom in mm*
WellColumnPitch	9	Enter the distance (in mm) from well to well. (9 for 96 well, 4.5 for 384)
WellColumns	12	Enter 12 for 96 well plate, 24 for 384 well plate
WellDepth	11	Enter the well depth in mm
WellPolygonPoints	Genetix.Fus	-
WellRowPitch	9	Enter the distance (in mm) from well to well. (9 for 96 well, 4.5 for 384)
WellRows	8	Enter 8 for 96 well plate, 16 for 384 well plate
WellShape	Round	Select 'Square' or 'Round' depending on the shape of the well
WellTopDiameter	6.5	Enter Top well diameter in mm

If detection area is still a problem, contact Molecular Devices Technical Support www.moleculardevices.com/service-support.

Viewing and Generating an Error Report

When your instrument is in an error state, you can view and generate an error report. If you need Molecular Devices Technical Support help, this report is required.

To generate an error report:

1. Select **Tools > Configuration**.
2. Select **System Log**.
3. From the **System Log** list, select the most recent Log.
4. From the **Event Severity** section, select the **Error** and **Fatal** check boxes.
5. To review the details, highlight the results lines.
6. To generate a report of the results listed, select **Tools > Prepare Error report**.
7. When the **Error Reporting Wizard** dialog appears, click **Next**.
8. Specify a location to save the error logs.
9. When requested, send the generated file to Molecular Devices Technical Support .

Obtaining Support

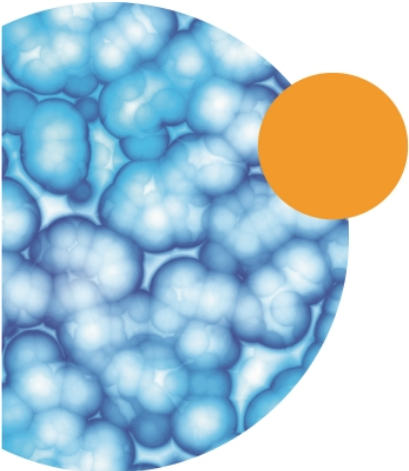
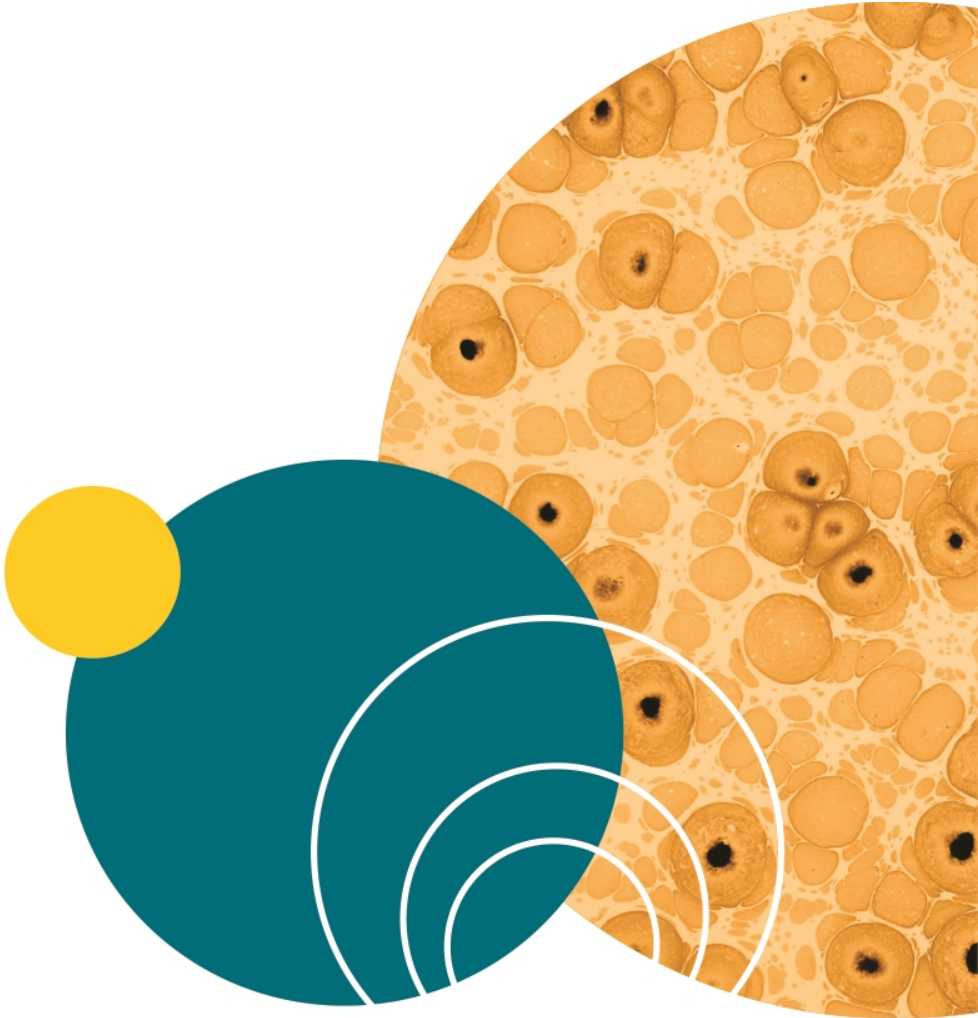
Molecular Devices is a leading worldwide manufacturer and distributor of analytical instrumentation, software, and reagents. We are committed to the quality of our products and to fully supporting our customers with the highest level of technical service.

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

Please have your instrument serial number or Work Order number, and your software version number available when you call.



WARNING! BIOHAZARD. It is your responsibility to decontaminate components of the instrument before you return parts to Molecular Devices for repair. Molecular Devices does not accept items that have not been decontaminated where it is applicable to do so. If parts are returned, they must be enclosed in a sealed plastic bag stating that the contents are safe to handle and are not contaminated.



Appendix A: Technical Specifications



The following tables list the technical specifications for the CloneSelect Imager.

Table A-1: Dimensions (Total assembled)

Item	Description
Size	575 mm (width) x 720 mm (depth) x 438 mm (height) Excluding ancillary equipment
Weight	45 kg

Table A-2: Operating Environment (Indoor use only)

Item	Description
Temperature	10°C to 40°C
Humidity	20% to 80% non-condensing
Altitude	Up to 2000 M
Mains supply	± 10% Rated Voltage
Transient overvoltage	Installation Category (Overvoltage category) II
Rated pollution	Pollution degree 2

Table A-3: Electrical Supply

Item	Description
Voltage	100 to 200 VAC, 50 to 60 Hz, single phase
Power	100 VA
Connections	IEC input
Fuses	F1 - F3A

Instrument Dimensions

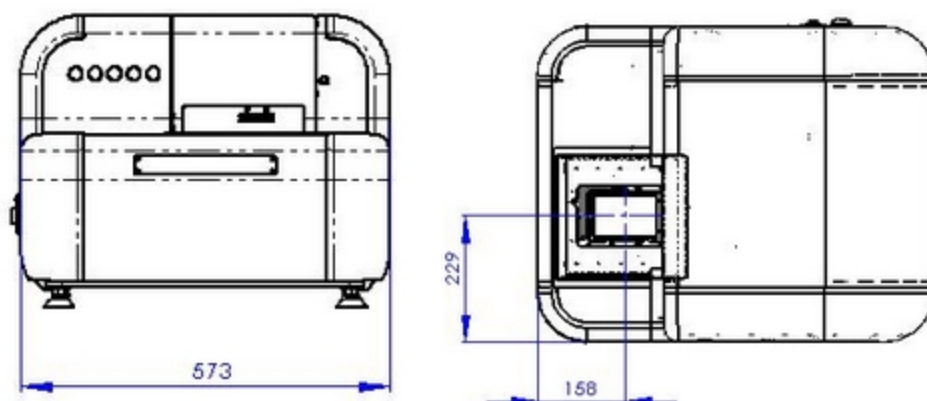


Figure A-1: The front and top views of the instrument.

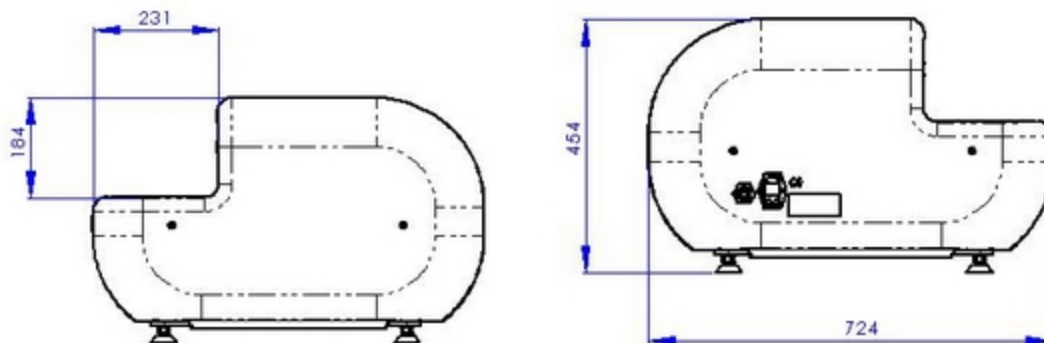


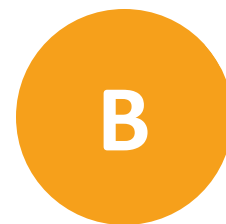
Figure A-2: The right side* and left side views of the instrument.

* The vertical measurement is from the top of the glass in the plate carrier to the top of the instrument



Note: All dimensions are in millimeters.

Appendix B: Compatible Plate Types

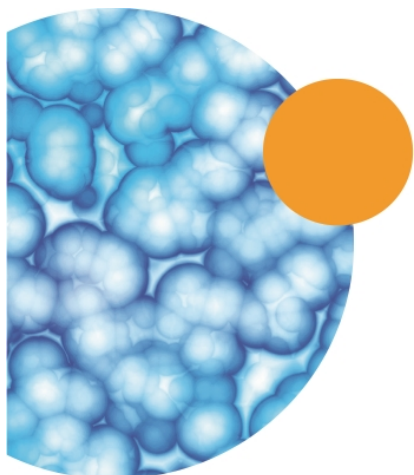
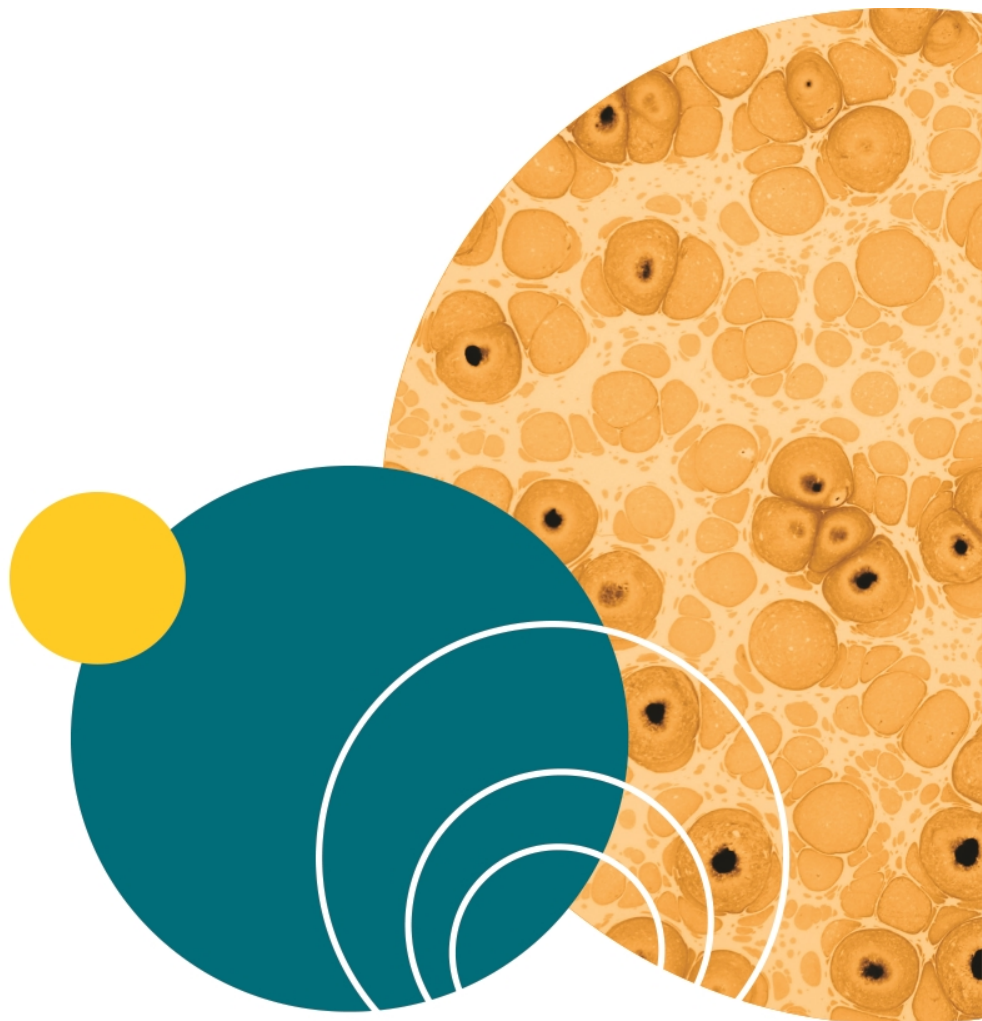


The standard CloneSelect Imager is configured to use the following plate types:

Manufacturer	Catalogue number	Well format
Corning / Costar	3300, 3595, 3474	96
	3524	24
	3513	12
	3516	6
Greiner	781182, 781091, 781098	384
	657185	6



Note: See the Knowledge Base for more plate compatibility information.
www.moleculardevices.com/service-support



Appendix C: Well List File Format



Note: You can only have one Well List File per Plate Type.

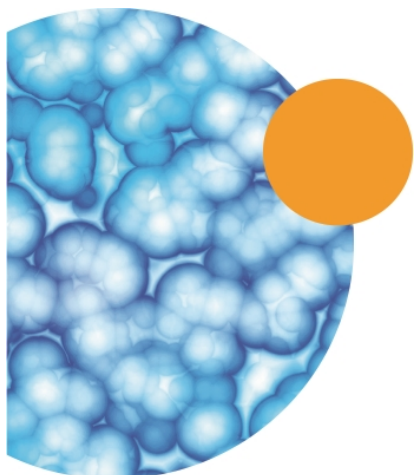
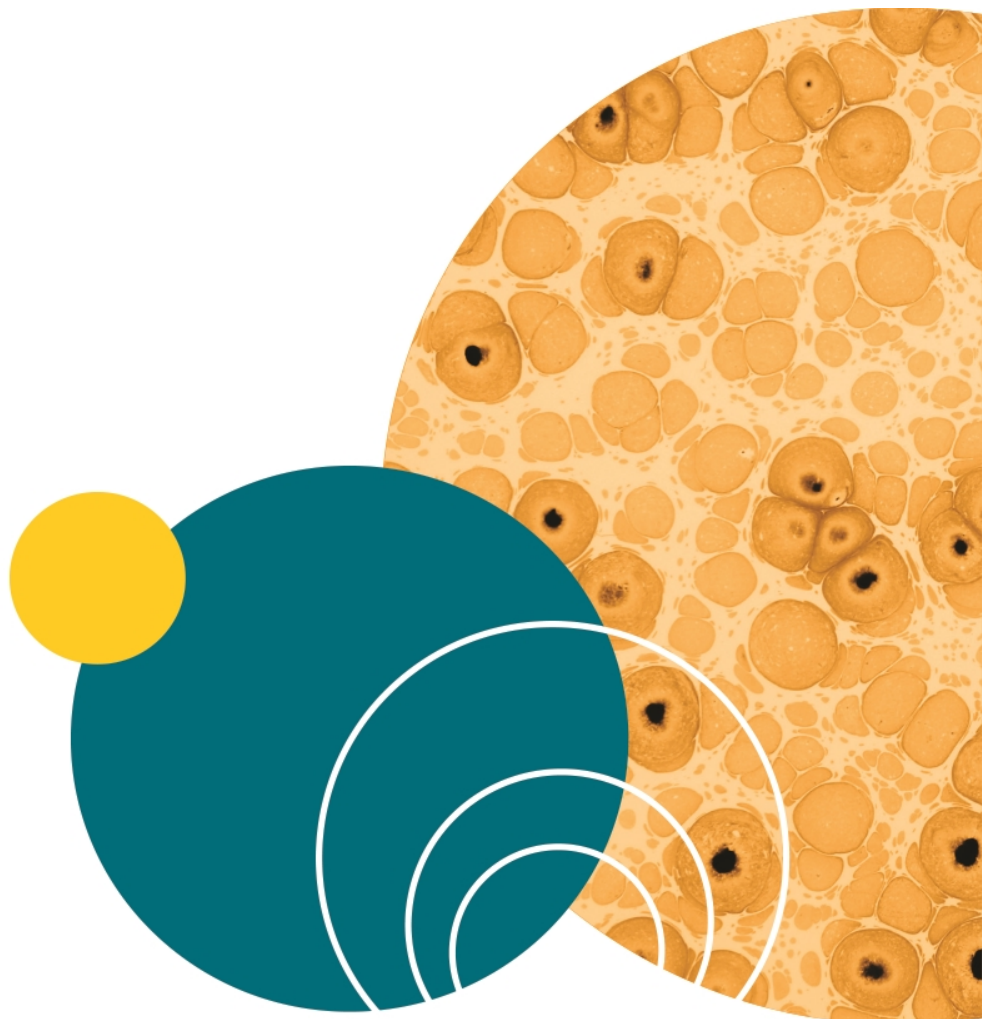
Use the following format for your Well List File:

```
Molecular Devices 96 well
A6
B5
E8
```

and so on.

To create a Well List File:

1. As the first line, enter the Plate Type name.
2. On a new line, enter the name of a well to image.
3. Repeat step 3 until all the wells you want to image are listed.
4. Save as a text file (.txt).
5. Use this file as your **Import** file during an Image Microplate process from the **Select Source** screen. See [Select Source on page 28](#).



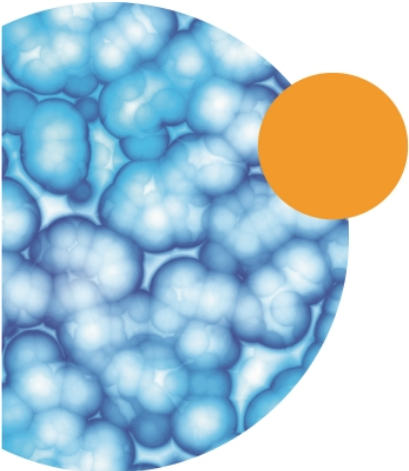
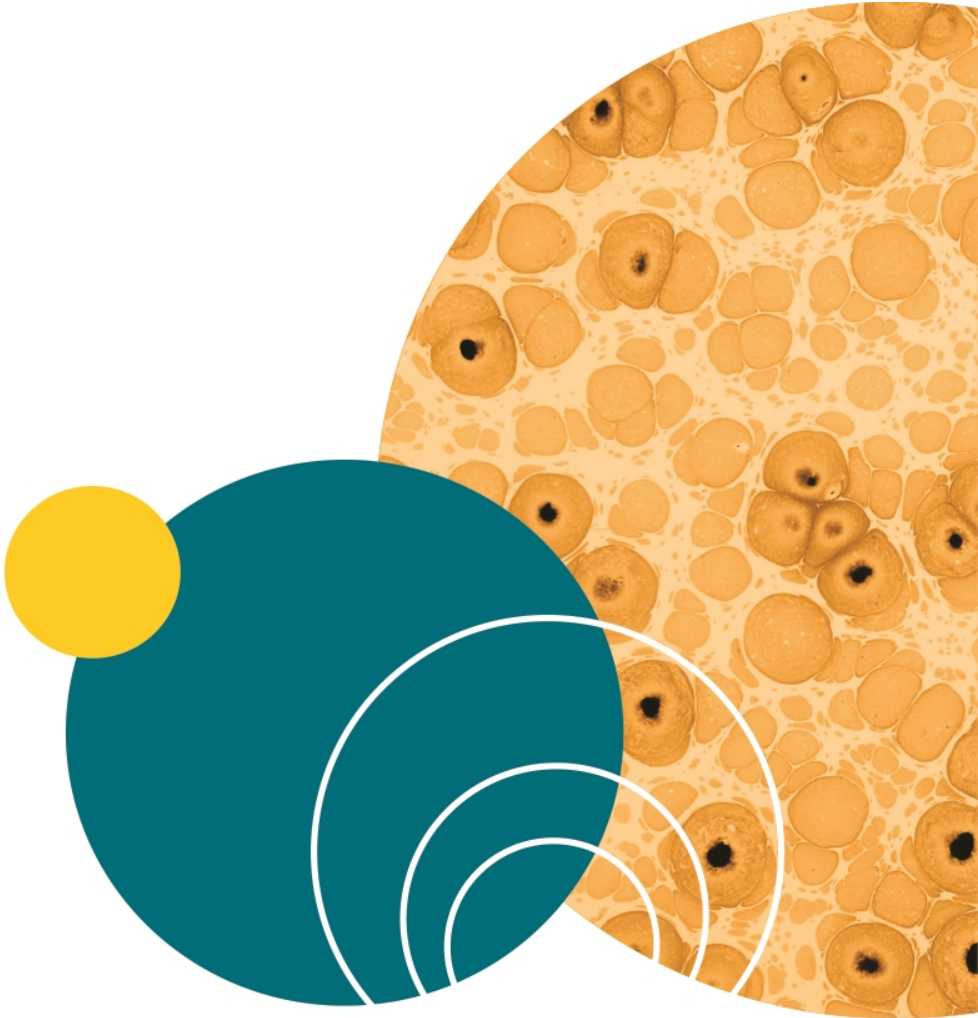
Appendix D: Setting up Cell Number Estimation



D

When creating a new [cell number estimation formula](#) you need to set up a standard plate that has a known number of cells in a range of wells.

Make a dilution series of a cell suspension on which a cell count has been performed (e.g. using trypan blue). Dispense an aliquot of each dilution into a well and incubate for a few hours to allow the cells to attach. Then image the plate and follow the instructions to create a new [cell number estimation formula](#).



Appendix E: CloneSelect Imager Data Viewer Software

E



Note: The CloneSelect Imager Data Viewer Software is optional. Contact Molecular Devices Technical Support to purchasing.

The CloneSelect Imager Data Viewer Software enables you to view results generated by the CloneSelect Imager instrument on a computer that is remote from the instrument.

You can either access a copy of the results files or you can access the folder on the instrument computer using Windows folder-sharing if the two computers are networked together. The default folder location is C:\Image archive.



Tip: Any time you install and use the CloneSelect Imager Software on a computer that is detached from the instrument, it functions as a data viewer.

Software Prerequisites

The CloneSelect Imager Data Viewer Software requires the installation of Microsoft .NET™ Framework 4.5.2 or newer. Your computer might already have it installed.

To check for .NET Framework 4.5.2:

1. Go to the **Control Panel** and click **Add/Remove Programs**.
2. Look in the list for **Microsoft.Net 4.5.2** or newer.



Tip: Windows 10 computers should already have Microsoft .Net 4.6 or newer installed.

3. If Microsoft .Net 4.6 or newer is installed, continue to [Installing CloneSelect Imager Data Viewer Software](#), otherwise install it now. See [Installing Microsoft .Net 4.5.2 on page 94](#).

Installing CloneSelect Imager Data Viewer Software

After verifying that the .NET Framework 3.5 SP1 is installed, you can install the CloneSelect Imager Data Viewer Software. See [Software Prerequisites](#).

To install the software:

1. Log in to the Molecular Devices Support website, go to the [CloneSelect™ Imager System Software Download Page](#), and in the table, click the **DataViewer > Installer** link.
2. Double-click **CloneSelect Imager Data Viewer v.x.x.x**, where x.x.x is the software version number, to start the installation.
To start the installation, if you are using the *Recovery DVD*, double-click **Setup_CloneSelectImager_Viewer.exe**.
3. Follow the on-screen instructions.

Licensing CloneSelect Imager Data Viewer Software

When the CloneSelect Imager Data Viewer Software is installed, you must license it.

The first time that you start the CloneSelect Imager Software after completing the installation, the software prompts you for a license. See [Obtaining a License for the Software on page 95](#).

Using CloneSelect Imager Data Viewer Software

When the CloneSelect Imager Data Viewer Software starts, the main process screen appears.

To view the results, locate where they are stored. On the right side of the screen, click **Results Location** and browse for the location of the results. The location can be a local folder on the computer, a CD-ROM or DVD, or, if the two computers are networked together, it can be a shared folder on the instrument computer.

After you select the location, the software catalogs all of the results and displays a list. You can then view the results the same way as you do on the instrument computer.

Contact Us

Phone: [+1-800-635-5577](tel:+18006355577)
Web: moleculardevices.com
Email: info@moldev.com

Visit our website for a current listing of worldwide distributors.