Incucyte Live-Cell Analysis Systems User Manual

Incucyte SX5
Incucyte S3
Incucyte SX1

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Preface

Welcome to the *Incucyte Live-Cell Analysis Systems User Manual*. The purpose of the *Incucyte Live-Cell Analysis Systems User Manual* is to answer your questions and guide you through the procedures necessary to use Incucyte efficiently and effectively.

Using the manual

You will find the *Incucyte Live-Cell Analysis Systems User Manual* easy to use. You can simply look up the topic that you need in the Table of Contents or the Index. Later, in this Preface, you will find a brief discussion of each chapter to further assist you in locating the information that you need.

Special information about the manual

The *Incucyte Live-Cell Analysis Systems User Manual* has a dual purpose design. It can be distributed electronically and then printed on an as-needed basis, or it can be viewed online in its fully interactive capacity. If you print the document, for best results, it is recommended that you print it on a duplex printer; however, single-sided printing also works. If you view the document online, a standard set of bookmarks appears in a frame on the left side of the document window for navigation through the document. For better viewing, decrease the size of the bookmark frame and use the magnification box to increase the magnification of the document to your viewing preference.

If you view the manual online, or print the manual using a single-sided printer, you might see a single blank page at the end of some chapters or appendices, or after a chapter page, appendix page, or section page. These blank pages have been added solely to ensure that the next chapter, appendix, or section begins on an odd-numbered page and in no way indicates that the manual is missing information.

Conventions used in the manual

The *Incucyte Live-Cell Analysis Systems User Manual* uses the following conventions:

- Information that can vary in a command—variable information—is indicated by alphanumeric characters enclosed in angle brackets; for example, <Vessel #>. Do not type the angle brackets when you specify the variable information.
- A new term, or a term that must be emphasized for clarity of procedures, is *italicized*.
- Page numbering is “online friendly.” Pages are numbered from 1 to x, *starting with the cover* and ending on the last page of the manual.

Although numbering begins on the cover page, this number is not visible on the cover page or front matter pages. Page numbers are visible beginning with the first page of the table of contents.

- This manual is intended for both print and online viewing.
  - If information appears in blue, it is a hyperlink. Table of Contents and Index entries are also hyperlinks. Click the hyperlink to advance to the referenced information.

Assumptions for the manual

The *Incucyte Live-Cell Analysis Systems User Manual* assumes that:

- You are familiar with Windows-based applications and basic Windows functions and navigational elements.
- References to any third-party standards or third-party software functions were current as of the release of this version of Incucyte, and might have already changed.

Organization of the manual

In addition to this Preface, the *Incucyte Live-Cell Analysis Systems User Manual* contains the following sections, chapters, and appendices:

- Section 1, “Scheduling and Acquiring Scans,” on page 13 provides detailed instructions for configuring and scheduling vessel scans in the Incucyte application. This section contains the following chapters:
  - Chapter 1, “Getting Started With Incucyte,” on page 15 explains how to start and log in to Incucyte and provides an overview of the major navigational elements for the application. It also explains how to log out of Incucyte.
  - Chapter 2, “Managing Scans,” on page 33 details all the procedures and considerations that are necessary for image acquisition, including configuring settings and scheduling acquisitions. It also details how to edit the acquisition settings for a vessel, and how to remove a vessel from the scanning schedule.
- Section 2, “Viewing Images,” on page 75 provides detailed instructions for selecting the vessel in the database that contains the images that you are analyzing and for viewing, comparing, and ultimately selecting the images in the vessel that you are analyzing. This section contains the following chapters:
  - Chapter 1, “Selecting a Vessel,” on page 77 details the Scanned Vessels window, which displays all the vessels that have ever been scanned on your Incucyte system by any user, as well as all the options that are available on the window for selecting the correct vessel.
  - Chapter 2, “Viewing and Working With Images,” on page 87 details the Vessel View window, which displays the scanned vessel that has been selected for image analysis, and all the options that are available on the window for viewing, comparing, and ultimately selecting the images that you are analyzing.
- Section 3, “Analyzing Images and Visualizing Results,” on page 111 details how you define or select your image analysis parameters, and then define your image analysis as well as how to visualize and investigate your analysis results. This section contains the following chapters:
  - Chapter 1, “Defining the Image Analysis,” on page 113 details the Launch Analysis wizard, which you use to define or select your image analysis parameters, and then launch the analysis of the images.

    - Chapter 2, “Visualizing the Analysis Results,” on page 135 details the various ways that you can visualize your analysis results so that you can access and interpret your data and identify the keys areas of your assay that worked well as well as the areas that need attention or improvement.
- Section 4, “Supporting Information,” on page 165 details the various tools and options that are available for supporting your image analysis. This sections contains the following appendices:
    - Appendix A, “Plate Map Editor,” on page 167 details the use of the Plate Map Editor, which you use for defining a custom plate map for image analysis.
- Section 5, “Incucyte Management,” on page 179 details the procedures and considerations that are necessary for managing various experiment components.
    - Appendix A, “Incucyte Management,” on page 181 details all the procedures and considerations that are necessary for managing experiment definitions, analysis definitions, and vessels for an Incucyte.
- Section 6, “Incucyte Administration and Security,” on page 215 details the various functions that are available for administering and securing your image data as well as the Incucyte system itself. This section contains the following appendices:
    - Appendix A, “Incucyte Data Archives,” on page 217 details Incucyte’s Archive function, which you use to manage your historical vessel data in Incucyte.
    - Appendix B, “Changing the Incucyte SX1 Objective,” on page 229 details how to change the objective for the Incucyte SX1 before you configure and schedule a vessel scan.
    - Appendix C, “Changing the Incucyte SX5 Optical Module,” on page 233 details how to change the optical module for the Incucyte SX5 before you configure and schedule a vessel scan.
    - Appendix D, “Incucyte Security,” on page 237 details all the functions that are available to an Incucyte administrator for managing Incucyte users and user workgroups.

Section 1 Scheduling and Acquiring Scans

Section Contents

Chapter 1
Getting Started With Incucyte

The Incucyte® Live-Cell Analysis Systems (referred to as Incucyte in this manual for brevity) enables observation and quantification of cell behavior via real-time, quantitative live-cell analysis. Incucyte is the most current software for your Incucyte Live-Cell Analysis Systems. The application provides enhanced user-friendly control of all aspects of imaging and analysis including powerful and intuitive image processing tools. This chapter explains how to start and log in to Incucyte and provides an overview of the major navigational elements for the application. It also explains how to log out of Incucyte.

This chapter covers the following topics:

Starting and Logging Into Incucyte

After you install Incucyte, a shortcut icon for the application and the Incucyte Plate Map Editor are placed on the desktop. An option for the application is also available from the Start menu (Start > All Programs > Incucyte > Incucyte). When you start Incucyte, you have the option of connecting to a device or opening an Incucyte archive.

Figure 1-1: Incucyte desktop icon and Plate Map Editor desktop icon

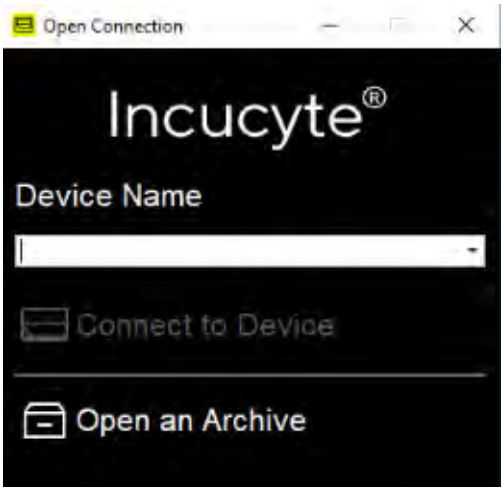
For detailed information about opening the Incucyte Plate Map Editor, see "Opening the Plate Map Editor" on page 169.For detailed information about opening an Incucyte archive, see "Opening an Archive" on page 227,

To start and log in to Incucyte

1. To start Incucyte, double-click the desktop icon, or select the option from the Start menu.

   The Incucyte Open Connection dialog box opens. The drop-down list that is displayed above these two connection options shows a history of device logins.

Figure 1-2: Incucyte Open Connection dialog box



2. On the Device Name drop-down list, select an instrument identifier, or enter the correct value.

The Incucyte device value is the instrument identifier that was assigned to your Incucyte system at installation, for example, IC50014. You can locate the device value for an Incucyte on the Application menu: Help > About. See "Common Features for an Incucyte Window" on page 21.

3. Click Connect to Device.

By default, Incucyte is set to connect to a device, so in lieu of clicking Connect to Device, you can just press [Enter].

The Login dialog box opens. If this is the first time that the selected device has been logged into, then the User Name field is blank; otherwise, the user name for the user who last logged into the selected device is displayed.

Figure 1-3: Incucyte Login dialog box

4. Enter your user name and password, and then click Connect or press [Enter].

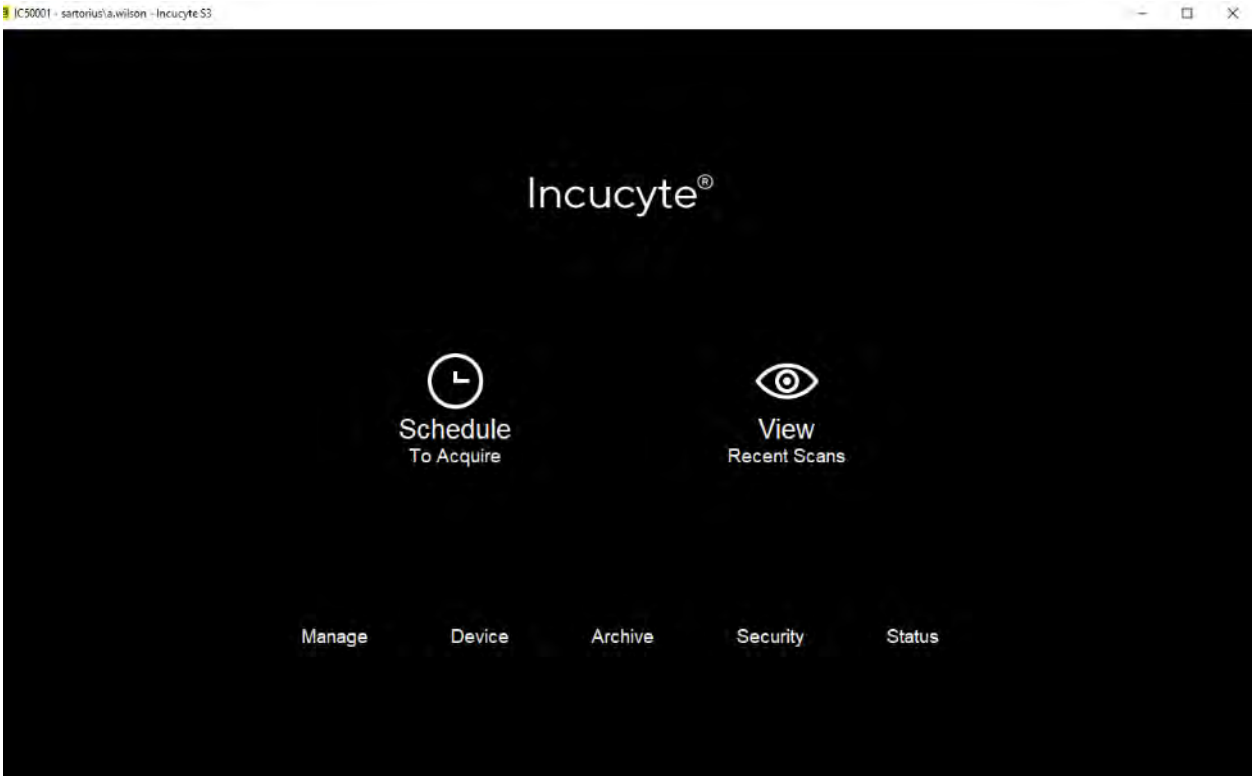
Sartorius establishes an Administrator user name and password with the purchase of the instrument. If you are the Administrator user, then you can create additional user accounts from the Security main menu option. When you create these additional user accounts, their credentials can be either Incucyte-specific or they can be Microsoft Windows domain credentials. See "Adding a User to Incucyte" on page 239.

The Incucyte main window opens. See "The Incucyte Main Window" on page 19.

The Incucyte Main Window

The Incucyte main window opens after you start and log in to Incucyte. The window is your starting point for accessing all the functionality that is available in Incucyte.

Figure 1-4: Incucyte main window



From top to bottom, the Incucyte main window has the following components: the title bar, the Scan icons, and the main menu.

Title bar

The following information is always displayed from left to right on the left side of the title bar: the Incucyte instrument name/ID, the domain that you are logged into and your User Name if you are a Domain user, or just your User Name if you are local user, and the instrument type. If applicable, additional information about the instrument (for example, a name/ID for a specific optical module) might be displayed after the instrument type. The standard Windows icons for minimizing, maximizing, and closing the window are displayed on the right side of the title bar.

The information that is displayed on the title bars of all other Incucyte windows depends on the version of Incucyte that you are logged into, the windows themselves, and the data that is displayed in the windows. For example, in addition to the user name and instrument type, the title bar of the Vessel View window displays the relevant assay name. The specific information that is displayed on the title bar of each unique Incucyte window is not detailed in this manual.

Scan icons

The two Scan icons—Schedule To Acquire and View Recent Scans—provide the links to the two primary functions of Incucyte: scheduling and acquiring scans, and then viewing and analyzing scans. The Schedule To Acquire icon provides the link to the scheduling and acquiring vessel scan functions. The View Recent Scans icon provides the link to the viewing scanned vessel functions. The Schedule and View options are also available in the menu that is displayed at the top of every Incucyte window, which is referred to as the *Incucyte Window menu*.

Main menu

The main menu on the Incucyte main window displays the top level of basic choices for Incucyte. The same options are also available from the menu that is displayed on the Incucyte Window menu.

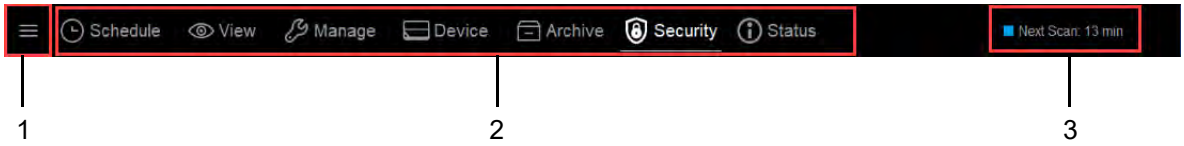
Option	Description
Manage	Opens the Manage window, which provides the options for managing critical components (analysis definitions and vessel types) for Incucyte.
Device	Opens the Device window, which provides the options for managing Incucyte support for scan diagnostics.
Archive	Opens the Archive window, which provides the options for managing your data archives. See Appendix A, “Incucyte Data Archives,” on page 217.
Security	Opens the Security functions, which you use for managing all your Incucyte users as well as specifying your Incucyte security settings. See Appendix D, “Incucyte Security,” on page 237. **Note:** Only Administrator users have access to Incucyte security functions.
Status	Opens the Status window, which provides options for viewing and managing the current status (system, temperature, archive, and analysis) for your Incucyte Live-Cell Analysis Systems as well as the corresponding system logs.

Common Features for an Incucyte Window

All Incucyte windows have the following features in common:

- (1) An Application menu.
- (2) An Incucyte window menu.
- (3) A Scheduling Status display.

Figure 1-5: Incucyte window common features



Application menu

The Application menu (Item #1 in Figure 1-5 above) is represented by three horizontal lines, referred to as a *Menu bar*, that are displayed in the top left corner of every Incucyte window. Click the Menu bar to expand the menu and display the following options:

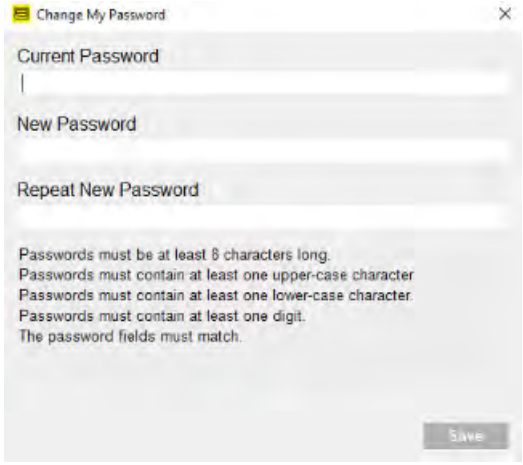
Option	Description
Connections	Provides options for opening (Open) and closing (Close) the connections to your Incucyte system. See “Starting and Logging Into Incucyte” on page 17 or “Logging out of Incucyte” on page 31.
Tools and Options	Provides the following options: • Plate Map Editor: Opens the Plate Map Editor. See Appendix A, “Plate Map Editor,” on page 167. • Open Graph: Opens a dialog box in which you can browse to and select a graph that was created in Incucyte. See Chapter 2, “Visualizing the Analysis Results,” on page 135. • Change My Password: If you are a local Incucyte user, then click this option to Open the Change My Password dialog box and change password. that you use to log in to Incucyte. See “To change your password as a local user” below. • Preferences: Opens the Preferences dialog box in which you can specify a variety of preferences for your Incucyte software installation. See “To set your Incucyte preferences” on page 23.
Help	Provides links to a variety of options that offer support for using Incucyte, including: • The user manual (User Manual) • Technical information about the instrument and software (Technical Notes) • A list of supported vessels (Supported Vessels) • General information about the installed software such as the current GUI version (About).
Exit	Logs you out of Incucyte and closes the Incucyte application. **Caution:** Because the Incucyte system is no longer active, your scans will not be acquired according to schedule.

To change your password as a local user

1. On the Menu bar, click Tools and Options > Change My Password.

   The Change My Password dialog box opens.

Figure 1-6: Change My Password dialog box



2. Change your password.

Option	Description
Current Password	The password that you are currently using to log in to Incucyte.
New Password	Passwords are alphanumeric and must have the following characteristics: • Contain at least 8 characters. • Contain at least one upper case character. • Contain at least one lower case character. • Contain at least one number. **Note:** This field is protected. You cannot copy the password from the Password field and then paste the password into the Confirm Password field.
Confirm Password	You must enter the password in this field *exactly* as you did in the New Password field.

3. Click Save.

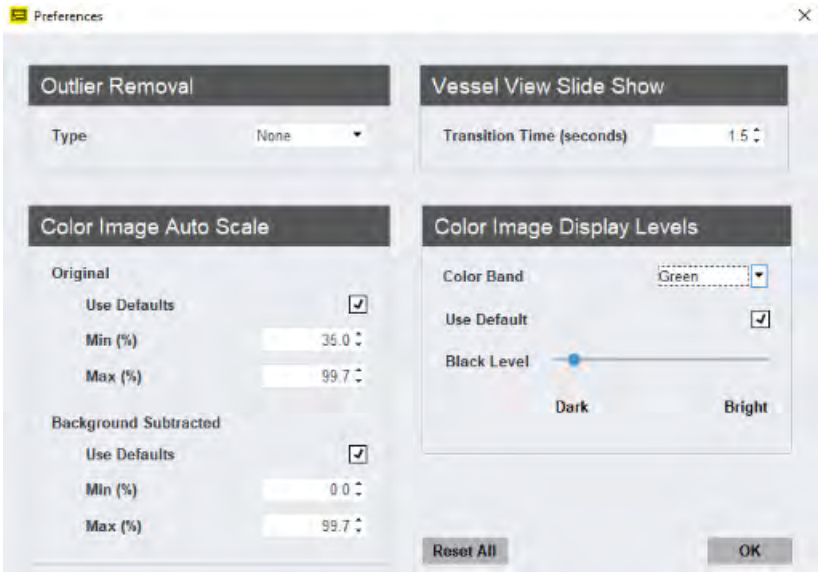
   The Change My Password dialog box closes. The currently active window/page remains open.

To set your Incucyte preferences

1. On the Menu bar, click Tools and Options > Preferences.

   The Preferences dialog box opens.

Figure 1-7: Preferences dialog box



You can hold your mouse pointer over any field on this dialog box, and then after the pointer changes from an arrow to an I-beam, a tooltip opens that describes the field.

2. Edit the preferences for your Incucyte system.

At any time, to reset all values in the Preferences dialog box to their default values in a single step, click Reset All. You also have the option of resetting selected values to their default values as described in the table below.

Option	Description
Outlier Removal	Type: This option facilitates the increasingly stringent removal of outlying metric data by discarding the metrics that are associated with poor image data outliers, thereby decreasing the standard deviation of the data. Select from one of three values: None (the default value), Standard, or Aggressive. **Note:** Increasing the stringency of this option from None to Standard or Aggressive does not necessarily improve the appearance of a Metric Time plot.
Vessel View Slide Show	Transition Time (seconds): The time, in seconds, between moving through the scan that were obtained for a vessel in the Vessel View window. See "The Vessel View Window" on page 89.

Option	Description

Color Image Auto Scale	When auto-scale is used for color image display, a histogram of the pixel values determines the Min and Max auto scale values.
	• Min (%): The specified value determines the percentage of pixels in the color channel that fall below the lowest range in the color map.
	• Max (%): The specified value determines the percentage of pixels in the color channel that fall below the highest range in the color map.
	• Original: Applicable for auto-scaling the originally acquired color images when background subtraction is *not* used in the analysis.
	  • Use Defaults. Selected by default.
	  • Min (%): Default value is 35.0.
	  • Max(%): Default value is 99.7.
	• Background Subtracted: Applicable for auto-scaling the originally acquired color images when background subtraction *is* used in the analysis.
	  • Use Defaults. Selected by default.
	  • Min (%): Default value is 0.0.
	  • Max(%): Default value is 99.7.

Note: Sartorius recommends that you use the up/down arrows to edit the Min/Max values as necessary. If you use the arrows to edit a Min value, a Max value, or both, then the Use Defaults option is automatically cleared. If you use your keyboard to edit a Min value or a Max value, then you must click in the field for the edited value to clear the Use Defaults option. If you select Use Defaults after editing either a Min value or a Max value, or both, then all edited values are automatically reset to their default values.	
Color Image Display Levels	On typical computer monitors, low fluorescent intensities that are near black (intensity = 0) are indistinguishable from black. This option is used to improve the visibility of dim objects by mapping the minimum fluorescent intensity to a display level that is set above zero (black). You can set this level independently for each different color band in an image. • Use Default: Selected by default. • Black Level: Set to a default value for each color band. Use the slider to change the default value for a selected color band.
Note: If you edit the default black level value for a selected color band, then the Use Default option is automatically cleared. If you edit the default black level value for a selected color band, and then click Use Default, the black level value is automatically reset to its default value.	

3. Click OK.

   The Preferences dialog box closes. The currently active window/page remains open.

Incucyte Window menu

The main menu on the Incucyte main window displays the top level of basic options for Incucyte. The same options are also available in the menu (Item #2 in Figure 1-5 on page 21) that is displayed at the top of every Incucyte window, referred to as the *Incucyte Window menu*. See “Main menu” on page 20.

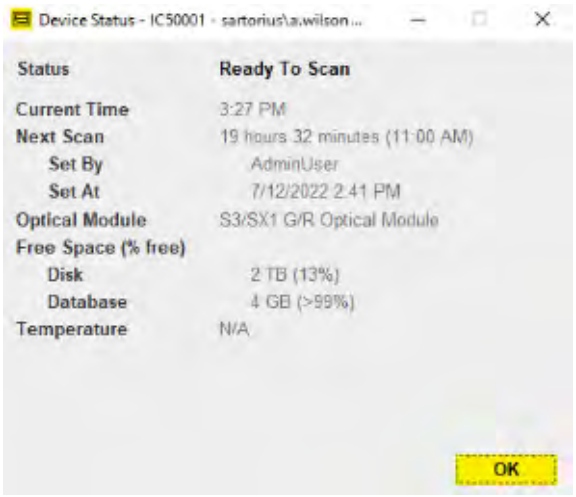
Scheduling Status

The Scheduling Status (Item #3 in Figure 1-5 on page 21) is color-coded to indicate a variety of statuses about your Incucyte system.

Color	Description
Yellow	Indicates a warning such as the drawer is open, the device is restarting or updating, the disk is running out of space, and so on.
Blue	Indicates the device is idle. The drawer is closed, no error state exists, and the device is not active. The text can vary, based on whether the instrument is processing data, sitting idle until the next scan, or sitting idle with no scans scheduled. If the device is idle waiting for the next scan, then “Next Scan” is displayed along with the time to the next scan overall. The selected vessel in the drawer has no relation to the time that is displayed. If no vessels are scheduled for scanning, then the message “No upcoming scans” is displayed.
Green	Indicates that the device is busy with some activity. Activities for which green is displayed include, but are not limited to, scanning, warming up, self test, and calibration. There is no relation with the selected vessel in the drawer.
Red	Indicates an error condition; however, there is no relation to the selected vessel in the drawer.

You can click anywhere on the Scheduling status display to open a Device Status message that displays general information about the device and when the next scan is to take place. The message has no relation to the selected vessel.

Figure 1-8: Device Status message



Working with Data Columns in an Incucyte Window

The following sections are intended to demonstrate, at a high level, how to work with and manage the data that is displayed in an Incucyte window or a wizard page. Not all data functions are available for all windows or pages, or even for all data columns, and not all functions are accessed or formatted identically; however, they are still similar enough that the following sections should provide a frame of reference for any approach that you select for working with and managing a data display.

Many windows or wizard pages in the different functional areas of Incucyte (Schedule, View, and so on) display the data in columns, where the columns are displayed in a default order and the data in the columns is ordered in ascending order based on the first column. For example, when the Vessel Selection page in the Add Vessel wizard first opens, the vessels are sorted in ascending order based on the Manufacturer. Regardless of the functional area, the majority of data columns in an Incucyte window or wizard page have the following features in common:

- A click and drag feature that you can use to change the order of the data columns, or change the width of the columns. See “To change the order of the data columns in an Incucyte window” below or “To change the widths of the columns in an Incucyte window” on page 27.
- A column context menu that has a multitude of options for sorting data, showing or hiding data columns, sizing columns, and so on. See “To use the options on the context menu for the data columns in an Incucyte window” on page 27.
- A search feature that you use to search for data that match specific criteria. See “To search for data in an Incucyte Window” on page 29.

Any changes that you make to a window or page display are persistent within the Incucyte session and thereafter across all sessions.

To change the order of the data columns in an Incucyte window

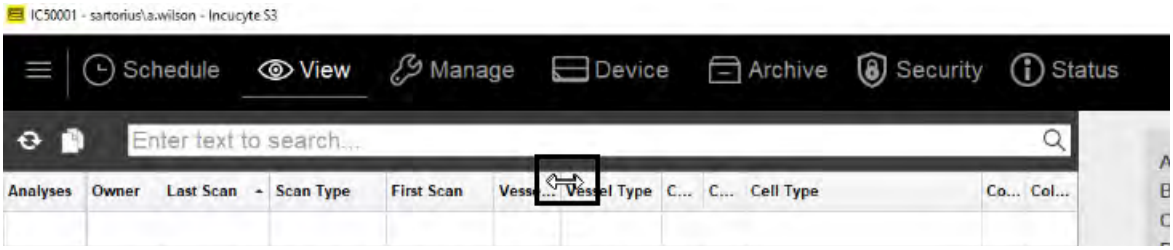
You can rearrange the column order in an Incucyte window or page by clicking and holding your mouse pointer on a column header, and then dragging the column to a new location.

Figure 1-9: Rearranging the columns in an Incucyte window

To change the widths of the columns in an Incucyte window

To change the width of a column in an Incucyte window or page, hold your mouse pointer on the right side of a column header until the pointer changes to a double-headed arrow, and then drag the column to its new width.

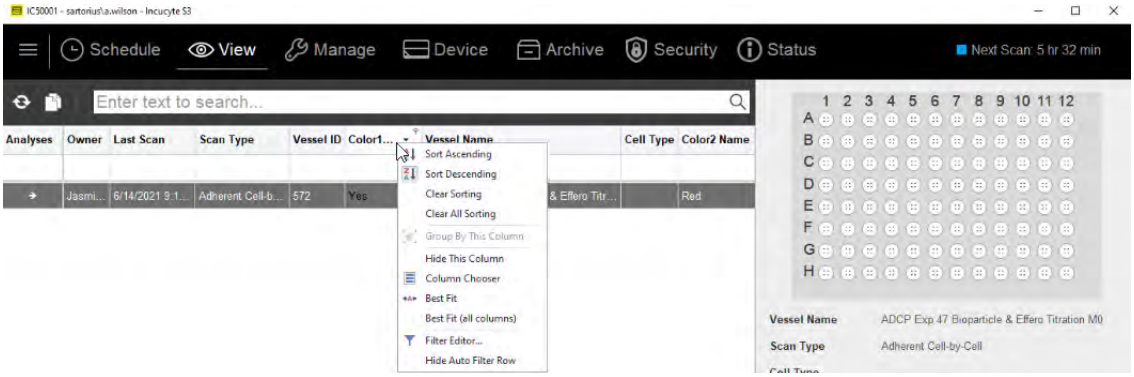
Figure 1-10: Changing the width of a column in an Incucyte window



To use the options on the context menu for the data columns in an Incucyte window

Right-click a data column in an Incucyte window or page to open a context menu that has a multitude of options for sorting data, grouping data, sizing columns, and so on.

Figure 1-11: Context menu for a data column in an Incucyte window



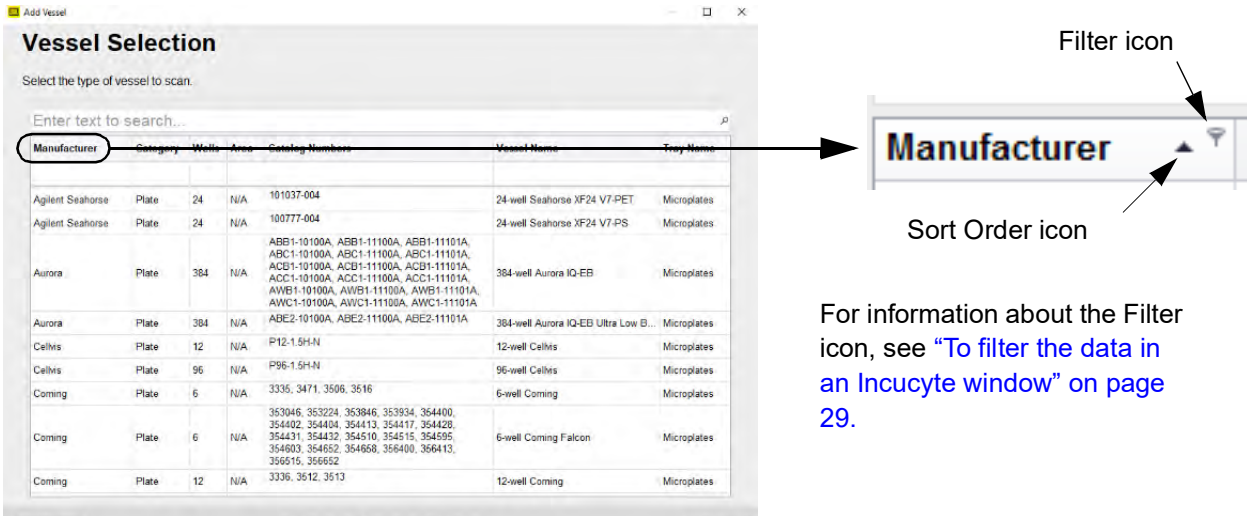
Option	Description
Sort Ascending	Display the data where the data in all columns is ordered based on the ascending sort of the selected column.
Sort Descending	Display the data in columns where the data in all columns is ordered based on the descending sort of the selected column.
Clear Sorting	Clear the sorting of the data in the selected column only.
Clear All Sorting	Clear the sorting of the data in all data columns.
Note: A column sorting option is also available from a Sort icon that is displayed in column headers. See "To sort the data in an Incucyte window" on page 28.	
Group By This column	Currently N/A.
Hide this column	Hide the selected column from the window or page display.

Option	Description
Column Chooser	Opens the Customization dialog box which lists all the columns by column header that are currently *not* displayed for the window or page. You use this option to select which columns to show in a window or page display, and which columns to hide from the display. • To display a column, double-click the column header to display the column in its default location. To display the column in a different location than it's default location, click and hold your mouse pointer on the column header, and then drag the column to its location.See "To change the order of the data columns in an Incucyte window" on page 26. • To hide a column, click and hold your mouse pointer on the column header in the display, and then drag the column to the Customization dialog box. When you are done selecting which columns to show and/or which to hide, click the X in the upper right corner of the Customization dialog box.
Best Fit	Incucyte automatically determines how much to widen or narrow the data column to fit the longest entry that is currently displayed in the column.
Best Fit (all columns)	Incucyte automatically determines how much to widen or narrow all data columns to fit the longest entry that is currently displayed in each column.
Filter Editor	Opens a Filter Editor dialog box, which provides options for filtering the data display based on filters that you specify. See "To filter the data in an Incucyte window" on page 29.
Hide/Show Auto Filter Row	Hides/shows the filter criteria, which is displayed in an automatically selected row at the top of the window or page, from the filtered data display.
Note: A filtering option is also available from a Filter icon that is displayed in column headers. See "To filter the data in an Incucyte window" on page 29.	

To sort the data in an Incucyte window

Many windows or wizard pages in Incucyte display the data in columns, where the data is ordered in ascending order based on the first column. For example, when the Vessel Selection page in the Add Vessel wizard first opens, the vessels are sorted in ascending order based on the Manufacturer. For such windows or pages, you can click once in a column header to display a Sort Order icon.

Figure 1-12: Add Vessel wizard, Vessel Selection page



For information about the Filter icon, see "To filter the data in an Incucyte window" on page 29.

The direction of the Sort Order icon indicates the sort order of the column data, where up indicates that the data is sorted in ascending order based on the data in the column and down indicates that the data is sorted in descending order. To change the sort order of the vessels to a descending sort based on a column, click the Sort Order icon for the column. To return the sort order to ascending order, click the Sort Order icon again.

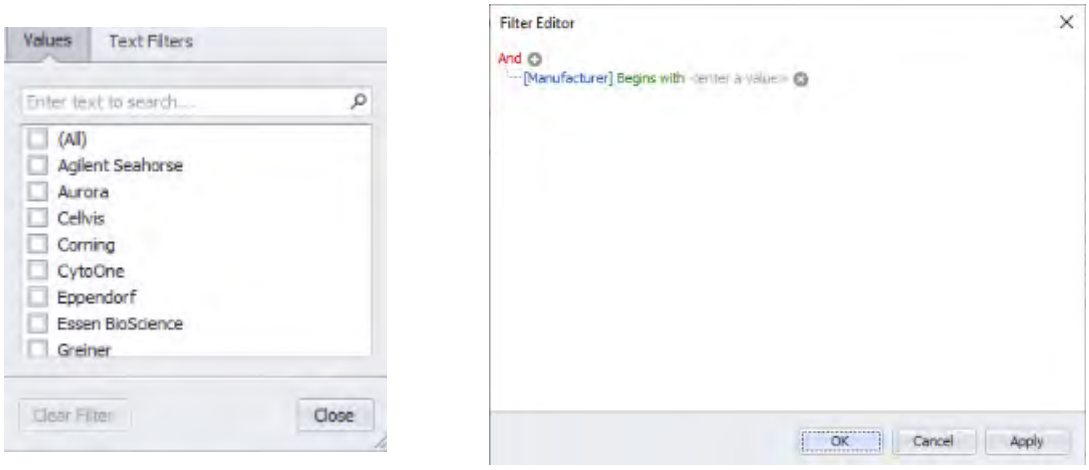
To filter the data in an Incucyte window

Filtering operates on the data that is currently displayed in a window or page and removes data from the display so that only a subset of the data that meets your criteria is displayed. Two options are available for accessing the data filtering function for a data column in an Incucyte window or page:

- A Filter icon that is displayed in the column header. To display this filter icon, hold your mouse pointer over a column header, and then click the icon to open the Filter Editor dialog box. See Figure 1-12 on page 28.
- The Filter Editor option on the data column context menu.

Both of these options open a variation of a Filter Editor dialog box in which you specify the criteria for filtering the data display.

Figure 1-13: Filter Editor dialog box via the column Filter icon (left) and the Filter Editor menu option (right)



A Filter list displays each unique value that has been entered for the field. Select a value on this list to filter the data by the value. For example, on the Vessel Selection page, to display only those vessel types that Corning manufactures, you can do either of the following:

- In the Manufacturer column, click the Filter icon and then on the Values tab of the Filter Editor dialog box, select Corning from the list of available values.
- In the Manufacturer column, right-click the column header and on the context menu that opens, select Filter Editor, and then set the filter as Manufacturer Equals Corning.

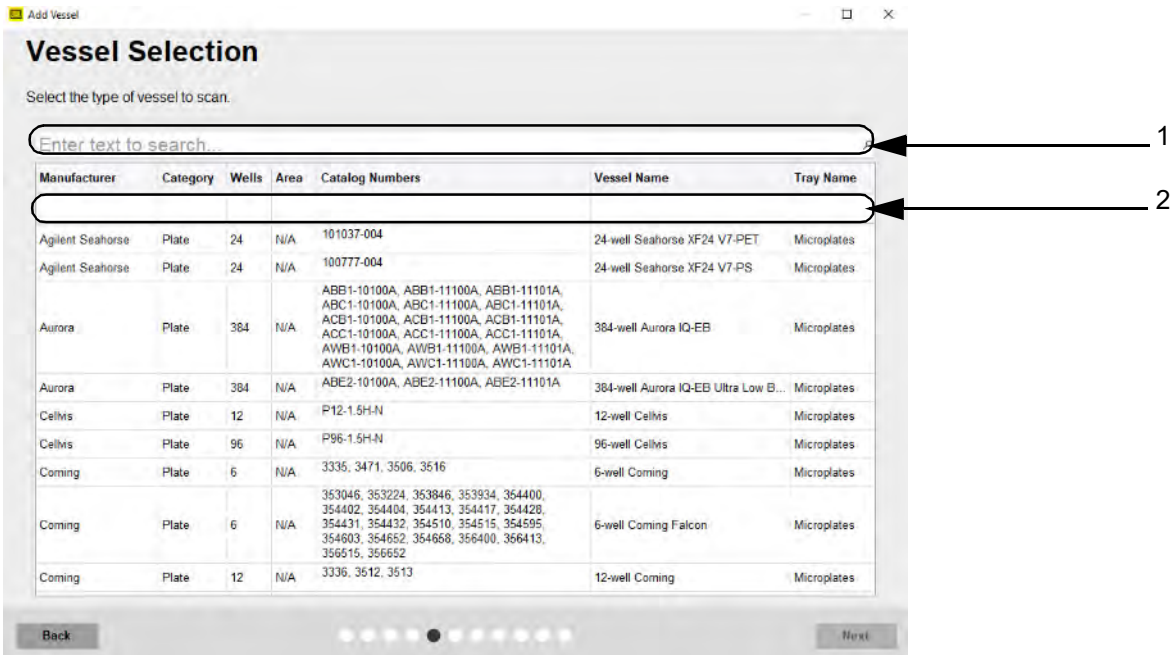
To search for data in an Incucyte Window

Searching starts from the data that is displayed and builds a new data list that matches your criteria. Many

Incucyte windows and wizard pages have an available Search function to assist you in locating specific data. If you can search the data in an Incucyte window or page, then a (1) blank Search field that spans all the columns is displayed at the top of the window or page, with the prompt “Enter text to search” displayed in the field. A (2) blank Search field is also displayed below the header for every column.*

**Unless noted otherwise, if a blank Search field is displayed beneath the header for a column, then the column can be searched.*

Figure 1-14: Add Vessel wizard, Vessel Type Search page



When you carry out a search for data in an Incucyte window, note the following caveats:

- To search across all columns, you enter the search string in the blank search field at the top of the page.
- To search based on a specific vessel property, for example, the Manufacturer column on the Vessel Selection page, you enter the search string in the blank field below the appropriate column header.
- The search is limited to the exact order of the characters in the string, but the string is not case-sensitive and it can appear anywhere in the search results. For example, if you search across all columns on the Vessel Selection page with **cor** as the search string, then the search results include vessels with a Manufacturer of **Cor**ning or a Vessel Name of 6-well **Cor**ning. The search string is highlighted in yellow in all the search results.
- To clear a search and reset the window or page to the default display of all data, clear the Search field, and then press [Enter].

Logging out of Incucyte

Unlike exiting an Incucyte (see “Exit” on page 21), when you log out of Incucyte, the Incucyte system remains active. This ensures you that your scans will be still be acquired according to schedule even if you are not logged in to the application.

To log out of Incucyte

1. From any window other than the Main window in Incucyte, click the Menu bar (the three vertical lines that are displayed in the top left corner of the window) to open a dropdown menu.
2. On the dropdown menu, select Connection.
3. On the Connection sub-menu, click Close.

Chapter 2
Managing Scans

The Incucyte software is divided into two major parts: acquiring scans and the viewing and analysis of the scans.

- Acquiring scans involves configuring vessel settings, scheduling acquisitions, acquiring images, and storing vessel data in a database. You can also edit the acquisition settings for a vessel and remove a vessel from the scanning schedule.
- Viewing and analysis consists of selecting, measuring, assessing, and managing the acquired images and vessel data.

This chapter details all the procedures and considerations that are necessary for acquiring images, including configuring vessel settings and scheduling acquisitions. It also details how to edit the acquisition settings for a vessel, and how to remove a vessel from the scanning schedule.

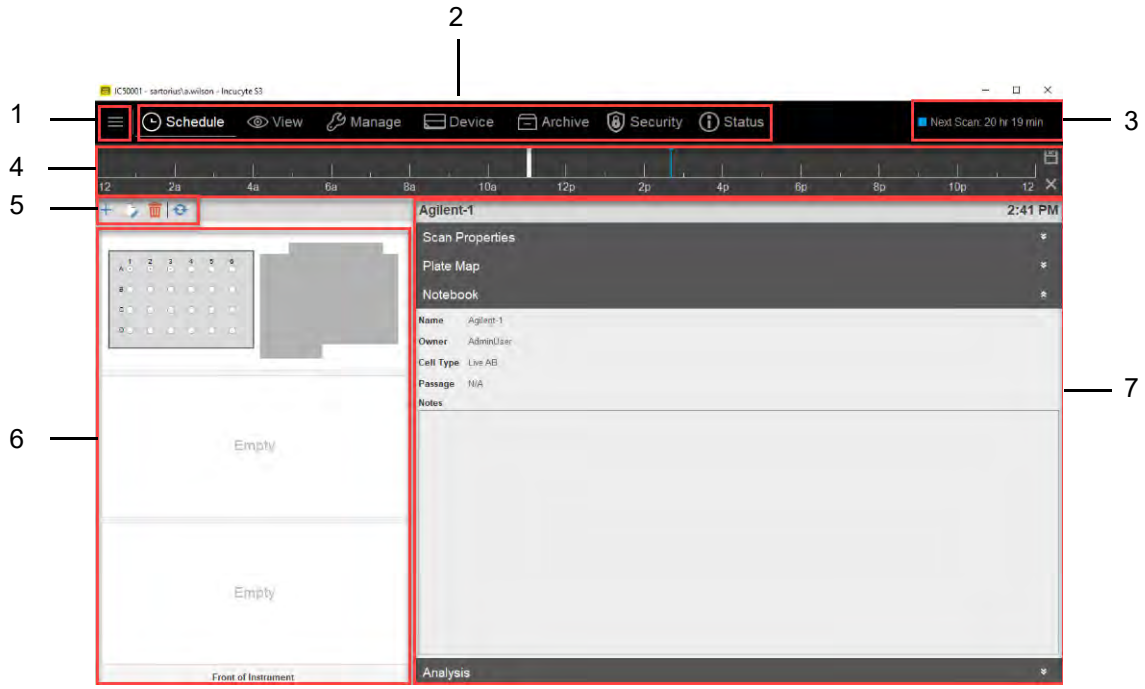
This chapter covers the following topics:

The Acquisition Window

You carry out all the necessary scheduling and configuration procedures for acquiring images in the Acquisition window. To open the Acquisition window, do one of the following:

- On the Incucyte main window, click Schedule To Acquire.
- On an Incucyte Window menu, click Schedule.

Figure 2-1: Acquisition window



The Acquisition window has the following layout:

	Window component	Description
1	Menu bar	Opens a dropdown menu with a variety of options. See “Application menu” on page 21.
2	Window menu	The Window menu displays all the same options that are available on the Incucyte main window. See “Incucyte Window menu” on page 24.
3	Scheduling status	Color-coded to indicate a variety of statuses about your Incucyte system. See “Scheduling Status” on page 25.
4	Vessel Schedule timeline	Displays the scanning schedules for all the vessels that are shown in the Vessel Drawer Setup pane. See “Vessel Schedule timeline” on page 36.
5	Vessel toolbar	Displays icons for carrying out specific tasks in the Acquisition window. See “Vessel toolbar” on page 36.
6	Vessel Drawer Setup pane	Indicates the positions in the Incucyte drawer in which a vessel has already been placed for scanning or where you can place a vessel for scanning. See “Vessel Drawer Setup pane” on page 36.

	Window component	Description
7	Vessel Information pane	Displays information for the vessel that is currently selected in the Vessel Drawer Setup pane. See “Vessel Information pane” on page 37.

Vessel Schedule timeline

The Vessel Schedule timeline displays the scanning schedules for all the vessels that are shown in the Vessel Drawer Setup pane. The schedule for the vessel that is currently selected in the Vessel Drawer Setup pane is shown in white bars. The schedules for the other vessels are shown in gray bars. A single blue line indicates the current day’s time. Options are available for viewing and adjusting the schedule for a selected vessel, combining vessel scanning schedules, scanning vessels on independent schedules (which is referred to as *per vessel scheduling*), saving a vessel scanning schedule, and deleting a vessel scanning schedule. See “Editing Vessel Information” on page 70.

Vessel toolbar

The Vessel toolbar displays icons for carrying out specific tasks in the Acquisition window. From left to right, these icons are as indicated below:

Icon	Description
	Launch Vessel Wizard icon: Opens the Add Vessel wizard. You use the Add Vessel wizard to configure the acquisition settings and the scanning schedule for a vessel. See “Configuring and Scheduling a Vessel Scan” on page 39.
	Edit selected vessel icon: Opens the Add Vessel wizard in Edit mode. You use this mode to edit specific properties for a vessel. See “Editing Vessel Information” on page 70.
	Remove selected vessel icon: Removes a selected vessel from the scanning schedule. See “Removing a Vessel from the Scanning Schedule” on page 72.
	Refresh schedule icon: Refreshes the Vessel Schedule timeline display. If you have scheduled new vessels for scanning, or made changes to an existing vessel scanning schedule, then you should click this icon.

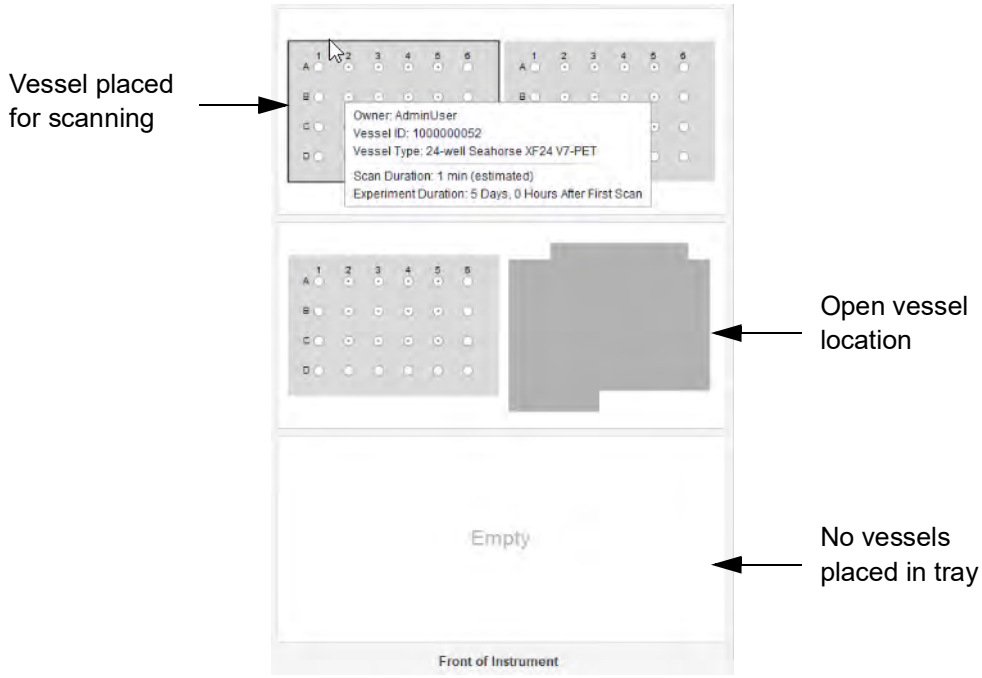
Vessel Drawer Setup pane

The Incucyte vessel drawer has three trays in which you can place vessels for scanning. The Vessel Drawer Setup pane indicates the tray locations in which a vessel has already been placed for scanning or where you can place a vessel for scanning. The pane depicts the three tray locations in the Incucyte drawer as if you are looking at the drawer from above, which is how you place vessels in the drawer.

- If a vessel has been placed in a tray location for scanning, then the location shows the vessel shape with the scan pattern.
- Any open vessel location that is in a partially filled tray is shown as a solid gray vessel shape.
- “Empty” is displayed for a tray that has all its vessel locations available.

You can hold your mouse pointer over a placed vessel to open a vessel tooltip that displays the following information about the vessel: Owner Name, Vessel ID, Vessel Type, Scan Duration, and Experiment Duration. See Figure 2-2 on page 37.

Figure 2-2: Vessel Drawer Setup pane



For the Incucyte SX5 and the Incucyte S3, all tray vessel locations are available. For the Incucyte SX1, only the front tray locations are available.

Vessel Information pane

The Vessel Information pane displays different information for the vessel that is currently selected in the Vessel Drawer Setup pane. Up to four different read-only tabs in the pane are available. Select an option on the pane (Scan Properties, Notebook, Plate Map, or Analysis) to open the corresponding tab. See Figure 2-3 on page 38.

- Scan Properties tab: Displays all the scan properties (Vessel Type, Scan Type, Image Channels, Objective, Scan Duration, Scan Schedule, and Experiment Duration) for the selected vessel. See “To specify the scan settings” on page 43.
- Plate Map tab: Applicable only if the vessel is a microplate and if a plate map has been has defined for the vessel. To open and maximize the Plate Map tab in its own window, click the tab once.
- Notebook tab: Displays all the information about the scheduled vessel, including the Vessel Name, the name of the Vessel Owner, the Cell Type, the Passage, the User Workgroup, and if applicable, the plate map for the vessel. See “To provide vessel information” on page 50.
- Analysis tab: If an analysis has been defined in the Add Vessel wizard for the selected vessel, then all the corresponding analysis information (analysis type, analysis definition, and if present, analysis notes) is displayed for the selected vessel. See “To set up the analysis” on page 52.

Figure 2-3: Vessel Information pane with Scan Properties tab for a selected vessel

Configuring and Scheduling a Vessel Scan

Acquisition involves configuring the acquisition settings, scheduling the vessel scan, acquiring images in specified vessel locations, and storing vessel data in a database.You use the Add Vessel wizard to configure the acquisition settings and scanning schedule for a single vessel at a time.

To demonstrate the full strength of the configuration and scheduling features for a scan in Incucyte and provide a frame of reference for any approach that you select for configuring and scheduling a vessel scan, the following procedures describe how to create a new vessel for a Standard scan with a scan schedule that is repeated every 24 hours.

If you are using the Incucyte SX1, then the objective that is currently in use is set as the default objective. If you require a different objective, then you must select it before you configure and schedule the vessel scan. If you configure and schedule a vessel scan for the Incucyte SX1, and then change the objective, the current schedule is immediately cleared and all scans for all vessels currently found in the schedule are immediately ended. See Appendix B, "Changing the Incucyte SX1 Objective," on page 229.

The procedures for configuring the acquisition settings and scheduling a vessel scan are applicable for *only a single vessel at a time*. You must repeat all the following procedures in their entirety for each vessel that you are scheduling:

- Setting the scanning frequency, which determines the number of times that the vessel is scanned, either on a scheduled 24 hour scan period or a single Scan Now option. See "To set the scanning frequency" on page 40.
- Creating the vessel, which determines how the vessel is created for scanning – as an entirely new vessel, by copying an existing vessel, by restoring a previously scanned vessel, or from an experimental definition. See "To create a new vessel" on page 41.
- Selecting the scan type, which determines the way that the vessel is scanned and the subsequent analysis options. You make your selection based on your assay and application. See "To select the scan type" on page 42.
- Specifying the scan settings, which consist of the image channels, objective, and if applicable, other settings based on the selected scan type. See "To specify the scan settings" on page 43.
- Selecting the vessel type that is be scanned, for example, a 96-well microplate or a flask.You select the vessel type, with the available choices pre-filtered based on the objective and scan type that you select for the scan settings. See "To select the vessel type" on page 45.
- Selecting the vessel location, which is the location in the vessel tray in the Incucyte drawer that the vessel is placed in for scanning. See "To select the vessel location" on page 46.
- Setting the scan pattern, which defines the locations in the vessel (wells, sectors, and so on) that you can select for image acquisition based on the selected vessel type. See "To set the scan pattern" on page 48.
- Providing vessel information, which requires that you name the vessel to identify it. You can also provide other optional information about the vessel such as the cell type that is being analyzed, the cell passage

number, or share the vessel with a user workgroup. If applicable, you can also create or import the plate map for your experiment. See “To provide vessel information” on page 50.

- Setting up the analysis, which determines the analysis type and associated analysis definition that you are carrying out for the vessel images. You can set up an analysis that is to launch automatically after the first image acquisition of the vessel, or you can defer the definition. See “To set up the analysis” on page 52.
- Setting the scanning schedule, which defines the frequency of vessel scanning during a 24-hour period. See “To set the scanning schedule for the Incucyte SX5 or Incucyte S3” on page 54 or “To set the scanning schedule for the Incucyte SX1” on page 57.
- Verifying the acquisition settings, which is a visual confirmation that the acquisition settings and scanning schedule have been correctly set. If so, you can add the vessel to the scanning schedule. See “To verify the vessel acquisition settings and schedule the scan” on page 59.

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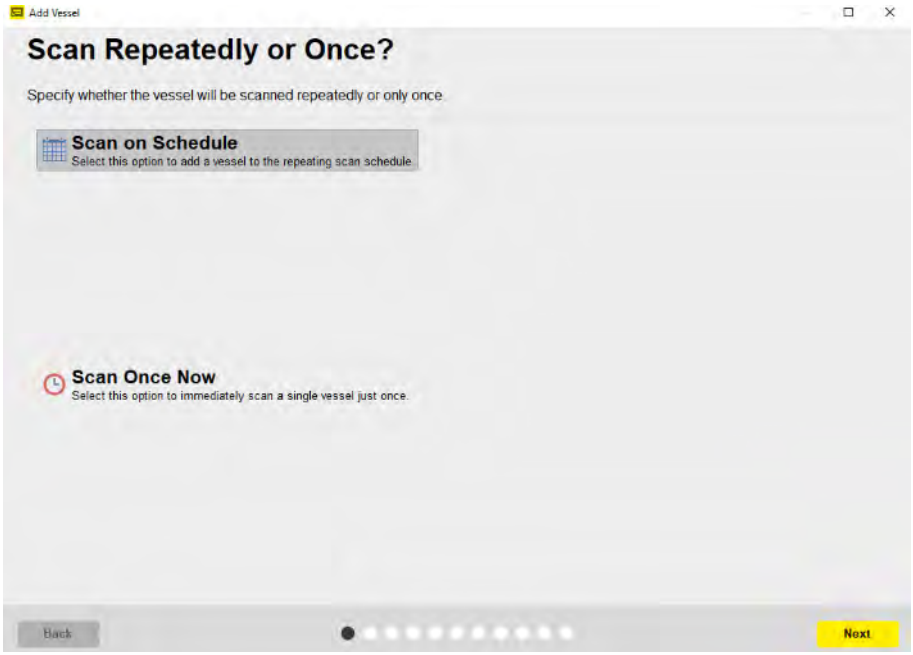
*The following procedures assume that the vessel has been shared with the **Everyone** user workgroup or that you are a member of the user workgroup for the vessel. To understand how user workgroups impact vessels and their data, see “Managing User Workgroups” on page 254.*

To set the scanning frequency

1. On the Acquisition window toolbar, click the Launch Add Vessel Wizard icon.

   The Add Vessel wizard opens. The Scanning Frequency page is the open page. Two options are available for scanning frequency: Scan on Schedule, which requires you to set up a scanning schedule for the vessel that is repeated every 24 hours until you end the scan (the default value), or Scan Once Now, which results in the vessel being scanned immediately one time after you specify all the scanning settings.

Figure 2-4: Add Vessel wizard, Scanning Frequency page

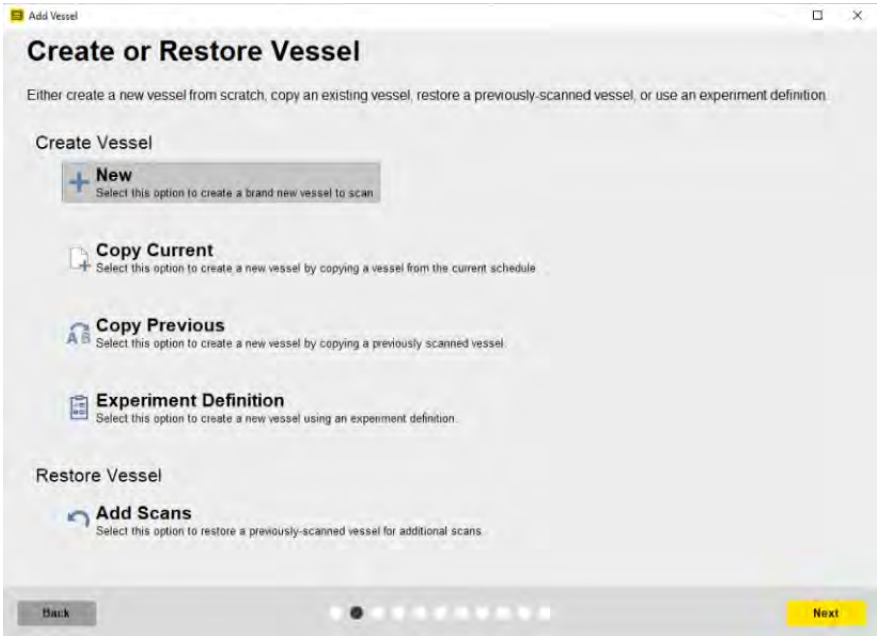


2. Leave Scan on Schedule selected.
3. Click Next.

   The Create Vessel page opens.
4. Continue to "To create a new vessel" below.

To create a new vessel

Figure 2-5: Add Vessel wizard, Create or Restore Vessel page



The Create or Restore Vessel page provides five options for creating the vessel that you are scanning.

Option	Description
Create Vessel	
New	Creates an entirely new vessel to scan.
Copy Current	Creates a new vessel by copying a vessel from the current schedule. The scan settings (objective, channels and other settings) and the vessel scan pattern are copied to the new vessel.
Copy Previous	Creates a new vessel by copying a vessel that was previously scanned. The scan settings (objective, channels and other settings) and the vessel scan pattern are copied to the new vessel.
Experiment Definition	Creates a new vessel based on the values that are specified in an *experiment definition*. • For detailed information about experiment definitions, see Appendix A, "Incucyte Management," on page 181. • For information about selecting an experiment definition to create a new vessel, see "To create the vessel based on an experiment definition" on page 60.

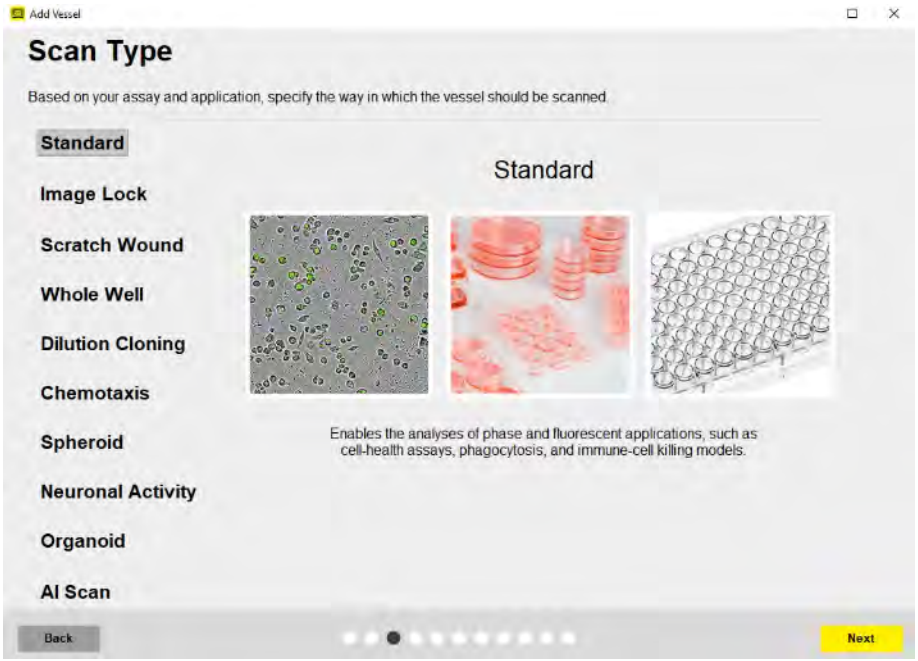
Option	Description
Restore Vessel	
Add Scans	Restores a previously scanned vessel for additional scanning. The new scans are added to the data for the previous scans for the vessel.

5. Under Create Vessel, leave New selected.
6. Click Next.

   The Scan Type page opens.
7. Continue to "To select the scan type" below.

To select the scan type

Figure 2-6: Add Vessel wizard, Scan Type page



The Scan Type page lists the different ways in which the vessel can be scanned. The physical capabilities of your Incucyte instrument and the version of the Incucyte software that you are running determine the scan types that are displayed on this page. You make your selection based on your assay and application. Standard is selected by default. If a scan type is functional for your Incucyte setup, then it is enabled on the Scan Type page; otherwise, if a scan type is not functional, then it is disabled (visible, but "grayed-out") on the Scan Type page.

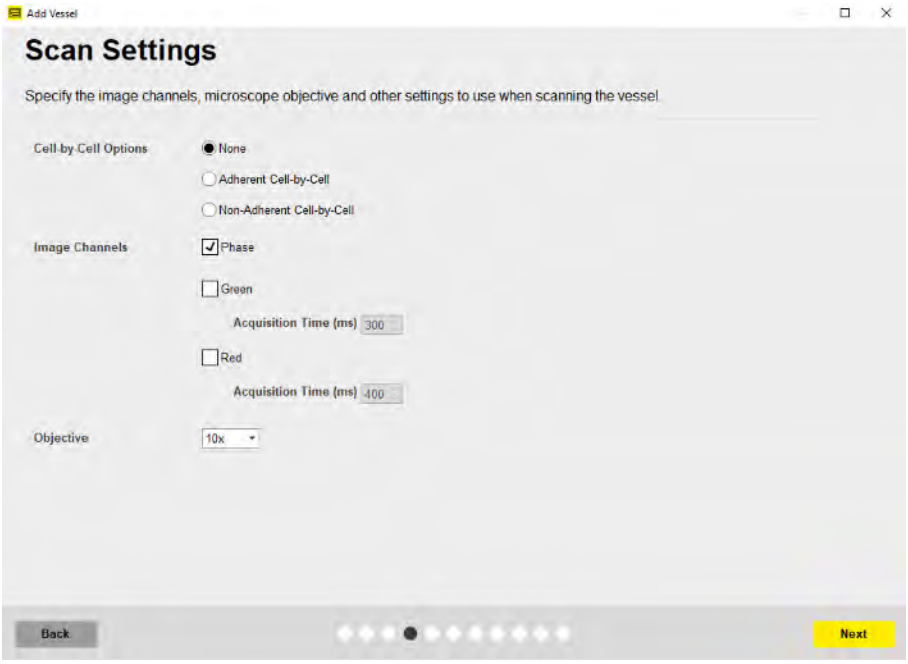
If a required scan type is not functional for you, then you must contact your Sartorius sales representative for assistance.

1. Leave Standard selected.
2. Click Next.

   The Scan Settings page opens.
3. Continue to "To specify the scan settings" below.

To specify the scan settings

Figure 2-7: Add Vessel wizard, Scan Settings page



You specify the Cell-by-Cell options, image channels, objective, and if applicable, other settings, based on the selected Scan Type on the Scan Settings page.

1. Specify the scan settings.

Option	Description
Cell-By-Cell Options	You can select only a single option. • None: Selected by default. • Adherent Cell-By-Cell: Requires the Phase image channel and a 10x or 20x objective. When this option is selected, then Phase is automatically selected and the objective is automatically set to 10x. You cannot clear the Phase selection but you can change the objective. • Non-Adherent Cell-By-Cell: Requires the Phase image channel and a 20x objective. When this option is selected, then Phase is automatically selected and the objective is automatically set to 20x and you cannot clear the Phase selection or change the objective.

Option	Description
Image Channels	Select the correct combination of channels: • Incucyte SX5: Phase, Green, Orange, and NIR. • Incucyte S3: Phase, Green, and Red or Phase, Orange, and NIR. • Incucyte SX1: Phase, Green, and Red. The acquisition time, which is comprised of a series of exposures, is the length of time that the cells are stimulated for fluorescence. The Acquisition Time options are set to the following default values: • Green: 300 ms • Red: 400 ms • Orange: 400 ms • NIR: 400 ms Sartorius has determined that these default values are the optimal values when scanning across a wide variety of fluorescent tags using the Incucyte Live-Cell Analysis Systems. Although you can modify these values, Sartorius does not recommend that you do so, unless you have the following considerations: • If phototoxicity is a problem, then you should lower the acquisition times. • If very faint objects are being imaged on a low background (objects and background together are less than 3 calibrated units (CU)), then it might be helpful to increase the acquisition times.
Objective	• Incucyte SX5 and Incucyte S3: The objective is set to a default value of 10x, which is suitable for the majority of vessel types and scan types. Other objectives (4x and 20x) are available for special scan types, for example, the Whole Well and Dilution cloning scan types, which require a 4x objective. • Incucyte SX1: The currently installed objective is set to the default objective. If you require a different objective, see Appendix B, “Changing the Incucyte SX1 Objective,” on page 229.

After you have scheduled this vessel for acquisition, you can always view a summary of the scan settings. In the Vessel Drawer Setup pane, select the vessel, and then in the Vessel Information pane, select Scan Properties. See “The Acquisition Window” on page 35.

2. Click Next.

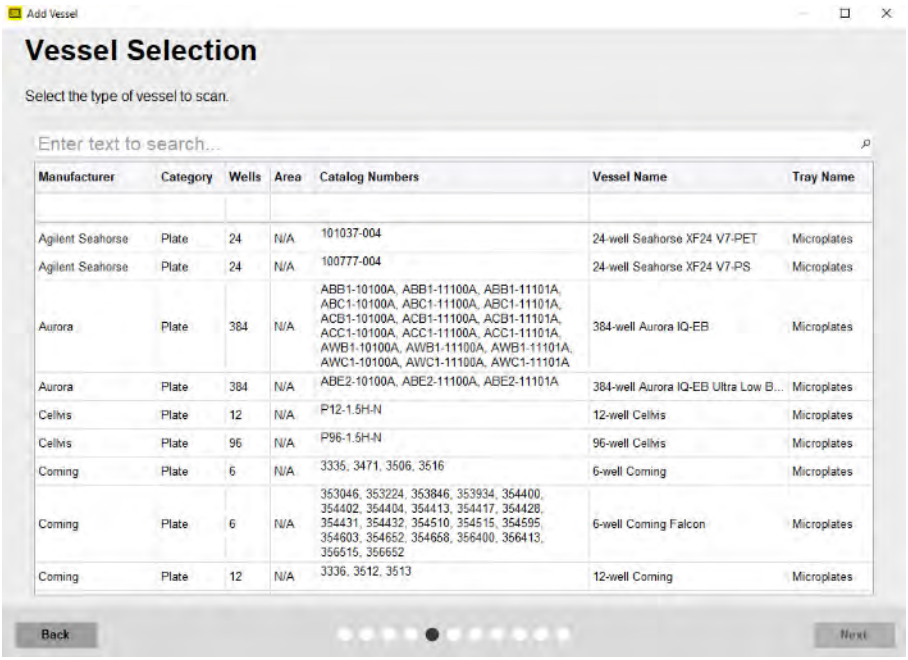
   The Vessel Selection page opens.

3. Continue to “To select the vessel type” on page 45.

To select the vessel type

The Vessel Selection page lists all the vessel types that the Incucyte system can support for your assay based on the objective and scan type. When the Vessel Selection page first opens, the vessels are sorted in ascending order based on the Manufacturer.

Figure 2-8: Add Vessel wizard, Vessel Type Search page



1. Select the appropriate vessel type.

You can filter and sort or search the data on the Vessel Type Search page using the standard Incucyte filter and sort or search functions. See ""To sort the data in an Incucyte window" on page 28 and "To search for data in an Incucyte Window" on page 29.

2. Click Next.

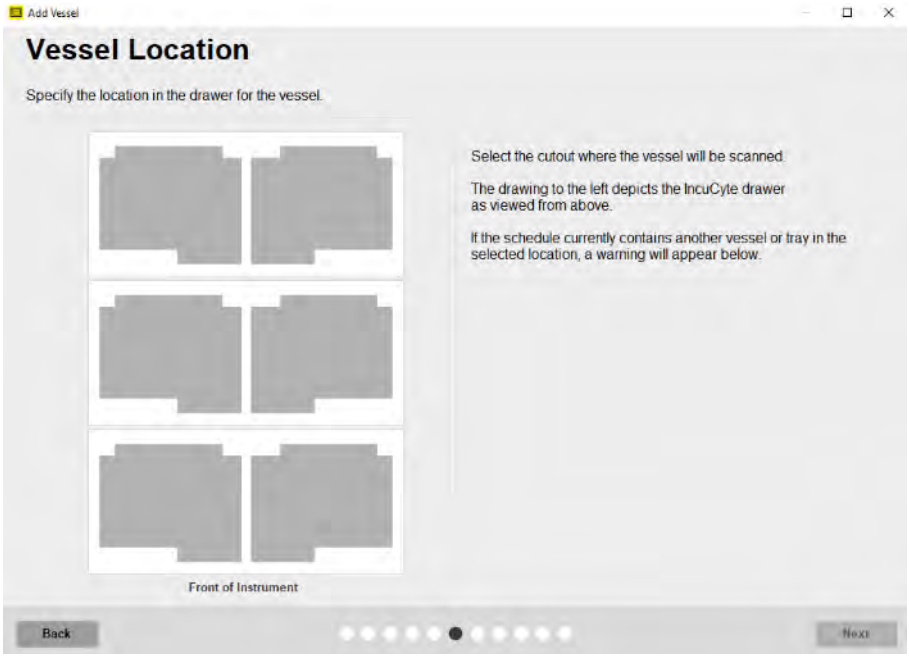
   The Vessel Location page opens.

You can also double-click a vessel to select it, and then open the Vessel Location page.

3. Continue to "To select the vessel location" on page 46.

To select the vessel location

Figure 2-9: Add Vessel wizard, Vessel Location page for a 96-well microplate



The pattern on the Vessel Location page and the shape of the location cutout are visual indicators of important information.

- The pattern on the Vessel Location page depicts the Incucyte drawer that is associated with your selected vessel type as viewed from above. Each “cutout” represents a location in a tray in which you can potentially place the vessel. For the Incucyte SX5 and the Incucyte S3, all six tray locations are available. For the Incucyte SX1, only the two front tray locations are available.
- The shape of the location cutout that is displayed on the page is dependent on the selected vessel type.
- The number of location cutouts that are available for vessel placement is dependent on your product type (Incucyte SX5 and Incucyte S3, or Incucyte SX1). For example:
  - If the selected vessel type is a Corning 96-well microplate, then the cutout is shaped like a 96-well microplate, and for the Incucyte SX5 and Incucyte S3, the maximum number of vessel locations is six, while for the Incucyte SX1, the maximum number of vessel locations is two.
  - If the selected vessel type is a Corning T-25 flask, then the cutout is shaped like a flask and for the Incucyte SX5 and Incucyte S3, the maximum number of vessel locations is increased to 12, while for the Incucyte SX1, the maximum number of vessel locations is increased to four.

1. Click the location in which you are placing the vessel.

   Two results are possible:

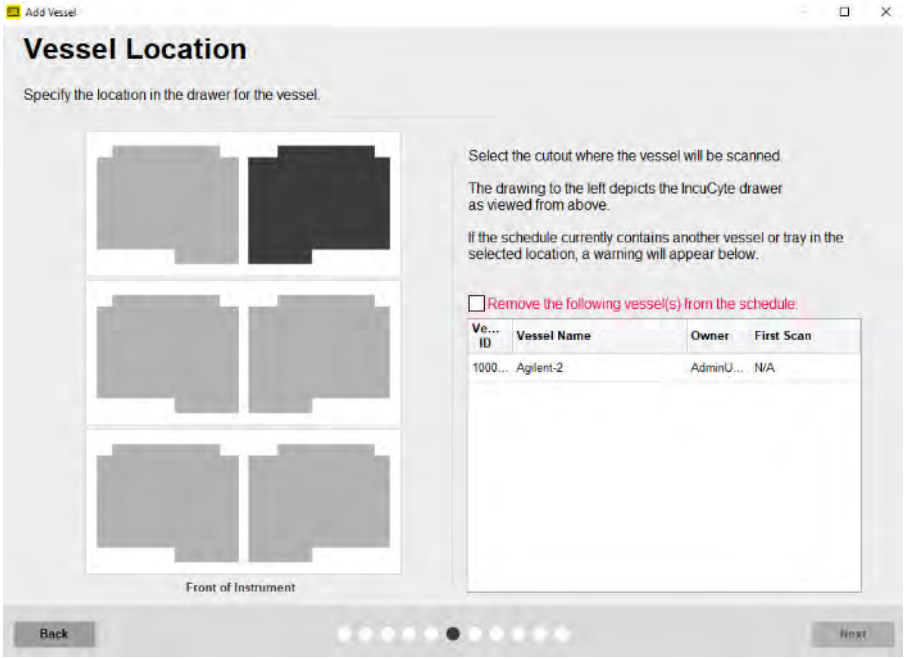
   - If any other schedules contain a vessel in the selected location(s), then a Vessel Schedule table opens. The table displays the following information for each affected vessel: the vessel ID and name, the vessel owner name, and when the first scan for the vessel was acquired. It also displays

an option to remove the previously scheduled vessels from the schedule. Continue to Step 2.

Before you continue Step 2, if you are scanning the vessel only once now, and another vessel already occupies the selected location, then based on the scanning schedule for the current vessel, you can remove the current vessel, add your vessel and carry out the scan, and then return the original vessel to its location without causing any conflicts with its scan schedule.

- If no other schedules contain a vessel in the selected location, then the location cutout is displayed in dark gray/black. Continue to Step 3.

Figure 2-10: Add Vessel wizard, Vessel Location page showing a currently occupied vessel location



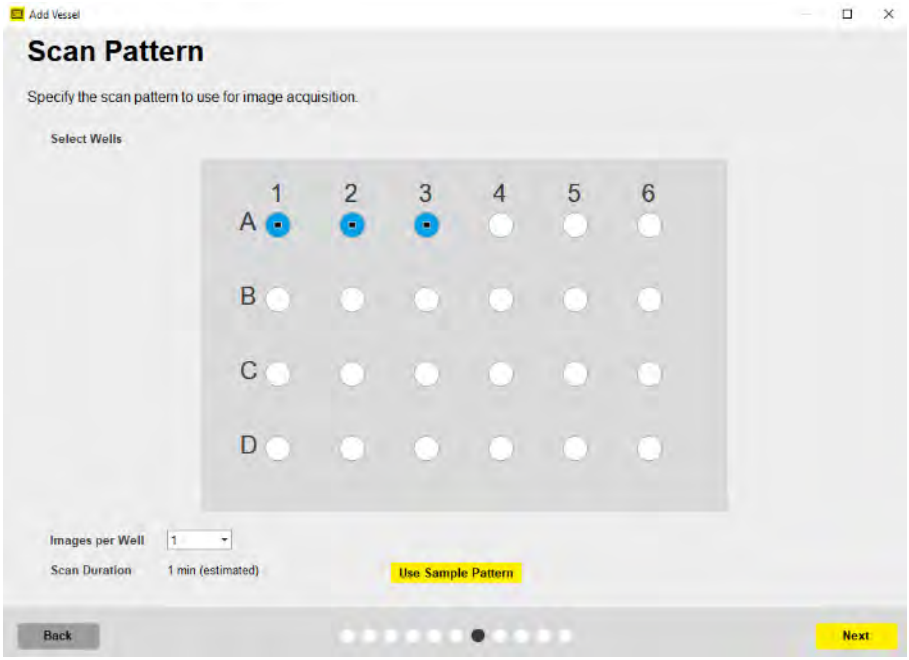
2. Do one of the following:
   - Select a different vessel location.
   - In the Vessel Schedule table, select the vessel or vessels that you are removing from the schedule, and then click “Remove the following vessel(s) from the schedule.”
3. Click Next.

   The Scan Pattern page opens.
4. Continue to “To set the scan pattern” on page 48.

To set the scan pattern

The Scan Pattern page displays a vessel map that mimics the shape of the selected vessel type and shows the locations in the vessel (wells or sectors) that you can select for image acquisition based on the selected vessel type. For example, for a 96-well microplate, you would select the *wells* that you are scanning and specify the number of images that you are acquiring in each well. For a flask, you would select the *sectors* that you are scanning and specify the number of images that you are acquiring in each sector. A solid blue fill pattern marks each location on the vessel map that you select for analysis. To set a scan pattern, you can use a sample scan pattern that is based on the manufacturer and selected vessel type, or you can set your own custom pattern.

Figure 2-11: Add Vessel wizard, Scan Pattern page with a Vessel map for a selected vessel type of Plate and three wells selected for analysis



1. Select the appropriate option.
   - To view and use a sample scan pattern, click Use Sample Pattern.

If a scan pattern has already been specified on the vessel map, then selecting the sample scan pattern overwrites the existing pattern.

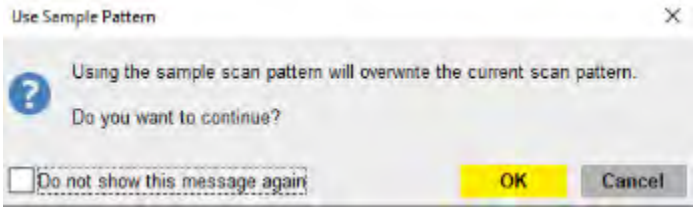
   - To set your own custom scan pattern for the vessel, you can do any or all of the following:
     - To select a single location in a vessel, for example, a well in a 96-well microplate, click once in the appropriate location in the vessel map.
     - To select multiple contiguous locations in a vessel, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.
     - To select all the locations in a vessel column in a single step, right-click a location in the

column, and on the context menu that opens, select Select this Column. Conversely, to clear all the selected locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Deselect this Column.

- To select all the locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Select this Row. Conversely, to clear all the selected locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Deselect this Row.
- To select all the locations in a vessel in a single step, right-click any location in the vessel, and on the context menu that opens, select Select All. Conversely, to clear all the selected locations in a column (including multiple contiguous locations), right-click any location in the vessel, and on the context menu that opens, click Deselect All.

To clear all the selected locations in the vessel map, or multiple contiguous locations, you can also press and hold the ALT key, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

Figure 2-12: Use Sample Pattern dialog box

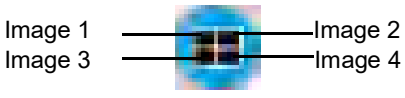


2. Specify the number of images that you are acquiring per vessel location (well, sector, or so on).

   The Scan Duration, which is the estimated amount of time required to scan the microplate, is displayed at the bottom of the Scan Pattern page. The value is based on the number of selected locations as well as the number of images that you are acquiring in each location. If you change one or both of these values, then the Scan Duration is updated accordingly.

If multiple images are acquired per vessel location, then the system considers the images ordered in rows from top to bottom and then, within the rows, from left to right. See Figure 2-13 below.

Figure 2-13: Four images acquired per vessel location



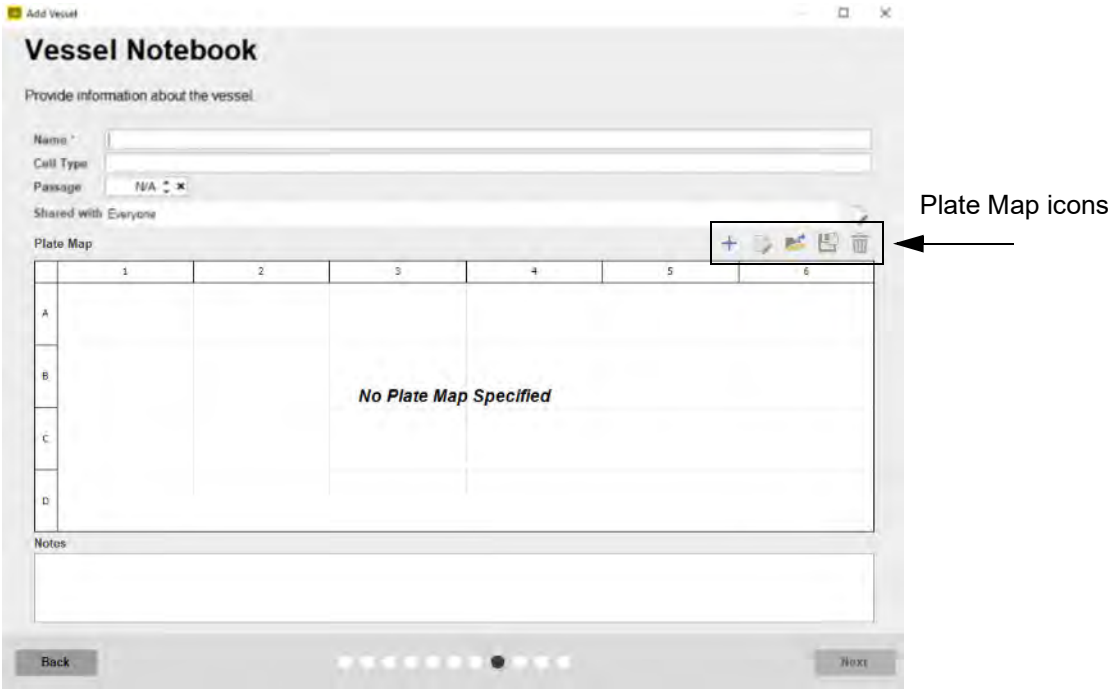
3. Click Next.

   The Notebook page opens.

4. Continue to “To provide vessel information” on page 50.

To provide vessel information

Figure 2-14: Add Vessel wizard, Vessel Notebook page for a microplate vessel type



You use the options on the Notebook page to provide information about the vessel. The Vessel Name is required. Cell Type and Passage are optional. You can also optionally share the vessel with a user workgroup, and, if applicable, you can create or import a plate map for your experiment.

1. Enter the vessel name.
2. Complete any optional information for the vessel, including the Cell Type, the Passage, the user workgroup, and if applicable, create or import the plate map. See:
   - “To share a vessel with a user workgroup” on page 51.
   - Appendix A, “Plate Map Editor,” on page 167.

Because a Plate Map is optional, and you are not limited to using the Plate Map Editor within the Add Vessel wizard, its details are not discussed here. Instead, only the icons that are available to access and use the editor are described.

Icon	Description
	Add plate map icon. Opens the Plate Map Editor which you use to create a new plate map for the vessel.
	Edit plate map icon. Opens the Plate Map Editor which you use to edit the plate map that has been assigned to the vessel.

Icon	Description
	Import plate map from disk icon. Opens the Open Plate Map dialog box which you use to browse to and select the plate map that is to be assigned to the vessel. An existing plate map is named as <Plate Map Name>.PlateMap.
	Save plate map to disk icon. Opens the Save Plate Map As dialog box in which you specify the name for the plate map that is assigned to vessel and the directory in which to save the plate map. The plate map is saved with a file extension of .PlateMap and you cannot change this.
	Remove plate map icon. Removes the plate map that has been assigned to the vessel. If the plate map has been previously saved to a specific directory such as one the Incucyte client or on the network, the plate map is *not* deleted from this directory.

3. Click Next.

   The Analysis Setup page opens.

4. Continue to "To set up the analysis" below.

To share a vessel with a user workgroup

Before you can share a vessel with a user workgroup, the User Workgroups setting must be turned on for the instrument. See "Specifying User Settings for an Incucyte" on page 248.

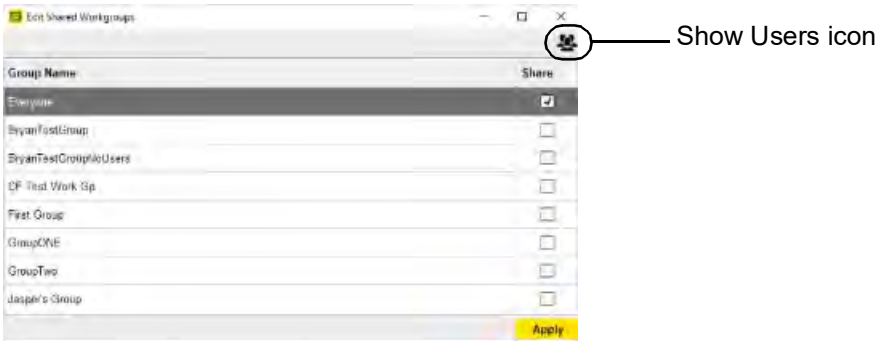
A *user workgroup* defines the users who are able to see a vessel and work with the vessel data. Sharing a vessel with a user workgroup is alway optional. By default, a vessel is initially shared with the **Everyone** user workgroup, which means that any user can view a vessel and work with its data. To limit the users who can view a vessel and work with its data, you must share the vessel with one or more specific workgroups.

1. At the far right of the Shared with field, click the Edit icon .

   The Edit Shared Workgroups dialog box opens. The dialog box lists all the user workgroups with which the vessel is already shared and the workgroups that are still available for sharing.

Before you share a vessel with a workgroup, you can view the users that are currently assigned to the workgroup. Click the Show Users icon to update the dialog box display with a list of current users for the selected workgroup.

Figure 2-15: Edit Shared Workgroups dialog box



2. Do one or both of the following:
   - To share the vessel with a user workgroup, click Share for the workgroup.

If the appropriate user workgroup is not available, contact your Incucyte administrator.

   - To stop the sharing of the vessel with a workgroup, clear the Share selection for the workgroup.

*If you clear all workgroup sharing for a vessel, then, by default, the workgroup is automatically reset to **Everyone**.*

3. Click Apply.

   The Edit Shared Workgroups dialog box closes and you return to the Vessel Notebook page. The selected user workgroup is displayed in the Shared with field.

There are two locations in the Incucyte software where you can view the workgroups with which a scanned vessel has been shared: the Scanned Vessels window and the Notebook tab on the Vessel Information window. (You can also change the workgroups for a scanned vessel on the Notebook tab.) See:

- “The Scanned Vessels Window” on page 79.
- “The Vessel Information window (via the Vessel View window)” on page 92.

To set up the analysis

You set up the analysis of the vessel images on the Analysis Setup page.

Figure 2-16: Add Vessel wizard, Analysis Setup page

You can set up an analysis that is launched automatically after the acquisition of the first image, or you can defer the setup. When you are setting up the analysis, you must understand the distinction between the two components of an analysis.

- Analysis Type: The selected scan type and within this selected scan type, the selected vessel type (such as a plate or flask) and the objective, determine the analysis types that are available.
- Analysis Definition: The specific analysis settings (parameters) that you select and define values for based on the selected analysis type.

Although the analysis definition is ultimately required to obtain the vessel data, you should consider the following *before* you decide to set up the analysis within the wizard or at a later date:

- An analysis definition does *not* currently exist in the Incucyte system that you can apply to your vessel because this is the first time that you have analyzed a vessel of this type (plate, flask, and so on) with these analysis settings (assay scan type, objective, cell type, fluorophore, and image channels). Therefore, you would set up the analysis at a later date. Leave Analysis Type set to <Defer analysis until later> and go to Step 2.
- An analysis definition currently exists in the system with the same analysis settings (assay scan type, objective, cell type, fluorophore, and image channels), but this analysis definition is not applicable for your vessel type. Therefore, you would set up the analysis at a later date. Leave Analysis Type set to <Defer analysis until later> and go to Step 2.
- An analysis definition has been created that aligns with the vessel type and analysis settings (assay scan type, objective, cell type, fluorophore, and image channels). Therefore, you can apply an analysis type and definition at the time of scheduling image acquisition for your vessel. Data is processed following each scan to provide analysis in real-time. Go to Step 1.

1. To set up the analysis, do the following:
   a. Select the analysis type.
   b. Select the analysis definition.

If you are an Admin user, then every analysis definition that was created on/imported to the Incucyte is displayed on the Analysis Definition drop-down list; otherwise, if you are not an Admin user, then only those definitions that have been shared with the ***Everyone*** *user workgroup or with a workgroup that you are a member of are displayed.*

   c. If applicable, enter the values for Spectral Unmixing.

Spectral unmixing options are displayed on the Analysis Setup page only if you selected more than one fluorescent channel (for example, Red and Green) for the analysis on the Scan Settings page. You can also complete spectral unmixing in the Vessel View window. See "Spectral Unmixing" on page 97 in the "The Vessel View Window."

   d. Optionally, enter notes for the analysis.

After you have scheduled this vessel for acquisition, you can always view a summary of the analysis settings. In the Vessel Drawer Setup pane, select the vessel, and then in the Vessel Information pane, select Analysis. See "Vessel Drawer Setup pane" on page 36.

2. Click Next.

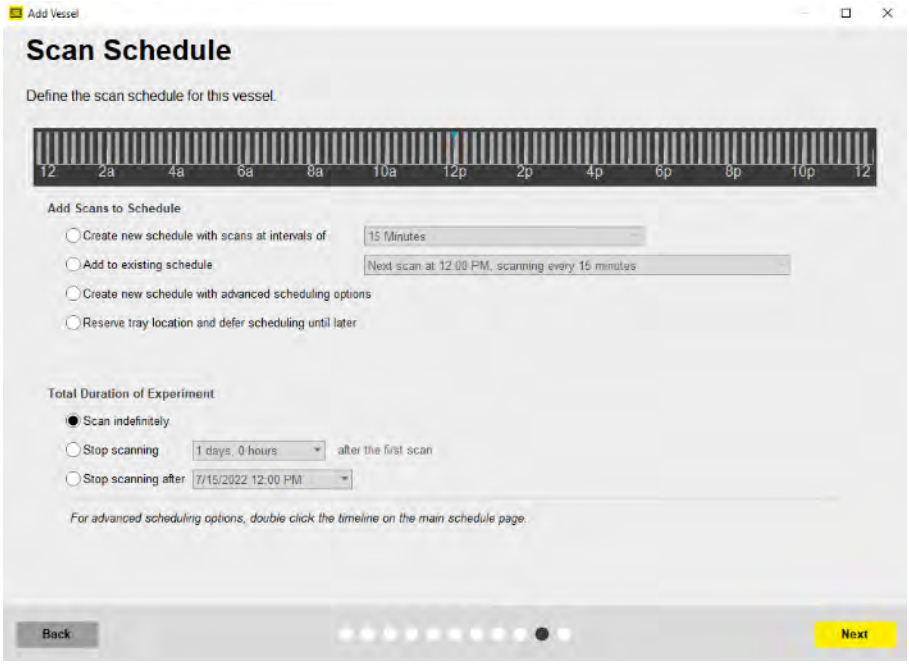
   The Scan Schedule page opens.

3. Continue to one of the following:

   - “To set the scanning schedule for the Incucyte SX5 or Incucyte S3” below.
   - “To set the scanning schedule for the Incucyte SX1” on page 57.

To set the scanning schedule for the Incucyte SX5 or Incucyte S3

Figure 2-17: Add Vessel wizard, Scan Schedule page for the Incucyte SX5 and Incucyte S3



The Scan Schedule page for the Incucyte SX5 and Incucyte S3 displays the scanning schedules for all the vessels that are currently scheduled for daily scans. The Vessel Schedule timeline that is displayed at the top of the page is identical to the Vessel Schedule timeline that is displayed at the top of Acquisition window. (See “Vessel Schedule timeline” on page 36.) The current time is indicated by a single blue line in the Vessel Schedule timeline. The scanning schedule for the vessel that you are scheduling is indicated by white bars in the timeline. The scanning schedule for each vessel that is currently scheduled in the vessel tray is shown as gray bars. The width of each schedule bar correlates to the amount of time that is required to scan the corresponding vessel. You have three options for setting the scan schedule for the vessel:

- You can set the scan schedule immediately for the vessel with at least one repeating scan. See “To set the scan schedule immediately with at least one repeating scan” on page 55.
- You can use the advanced scheduling option to set a custom scan schedule for the vessel. See “To use the advanced scheduling option to set a custom scan schedule for the vessel” on page 56.
- You can “reserve” the vessel tray and defer the scheduling for the vessel until a later time. See“To defer the vessel scheduling” on page 56.

To set the scan schedule immediately with at least one repeating scan

1. Select one of the following options:

Option	Description
Create new schedule with scans at intervals of	Select the scanning interval. White bars in the Vessel Schedule timeline indicate the schedule. **Note:** If this is the first time that a vessel is being scheduled for an Incucyte system, then the scan interval is set to a default value of 15 minutes; otherwise, it is set to the value that was selected for the last previously scheduled vessel. **Caution:** If you select an interval that results in overlapping scan times with the scanning schedules that have already been set for any of the other scheduled vessels in the vessel drawer, then a single red line marks each white bar in the timeline for the vessel that you are currently scheduling and the warning message "You have scan times that overlap" is displayed at the bottom of the Scan Schedule page. You must change the times of the scans so that no overlaps (shown in red in the timeline) occur before you can continue. *To do so, you must select a different value on the interval dropdown list.*
Add to existing schedule	Select an existing scanning schedule. The start times for the scanning schedules that have already been set for the other vessels in the vessel tray determine the next scan start time for the vessel that you are currently scheduling. When you select an existing schedule, the corresponding bars in the Vessel Schedule timeline are displayed in white instead of gray.

2. Specify the total duration of the experiment:

Option	Description
Scan indefinitely	The scanning schedule for the vessel is repeated every 24 hours until you manually end the scan.
Stop scanning (days/hours) after the first scan	Select the number of days/hours after the first scan at which the vessel scanning is automatically stopped.
Stop scanning after (date/time)	Select the date/time combination after which the vessel scanning is automatically stopped.

3. Click Next.

   The Summary page opens.

4. Continue to "To verify the vessel acquisition settings and schedule the scan" on page 59.

To use the advanced scheduling option to set a custom scan schedule for the vessel

You use the advanced scheduling option to set custom scan times for a vessel. When you set the custom scan times for a vessel, you do *not* have to specify a schedule that adheres to a regular repeating interval. For example, the schedule could be something as irregular as a scan every 11 minutes, then a scan every 25 minutes, and then finally, a scan every hour.

1. Select Create new schedule with advanced scheduling options.
2. For each scan time that are scheduling, click the Vessel Schedule timeline.
3. Specify the total duration of the experiment:

Option	Description
Scan indefinitely	The scanning schedule for the vessel is repeated every 24 hours until you manually end the scan.
Stop scanning (days/hours) after the first scan	Select the number of days/hours after the first scan at which the vessel scanning is automatically stopped.
Stop scanning after (date/time)	Select the date/time combination after which the vessel scanning is automatically stopped.

4. Click Next.

   The Summary page opens.

5. Continue to "To verify the vessel acquisition settings and schedule the scan" on page 59.

To defer the vessel scheduling

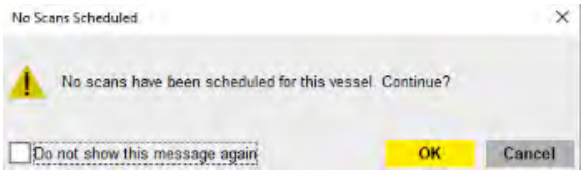
1. Select Reserve tray location and defer scheduling until later.
2. Leave Total Duration of Experiment set to the default value of Scan indefinitely.

Although all Experiment Duration options are enabled, selecting an option here is not applicable as you have not yet set the scan schedule.

3. Click Next.

   A No Scans Scheduled dialog box opens, indicating that no scans have been scheduled for the vessel and asking you if you want to continue.

Figure 2-18: No Scans Scheduled dialog box



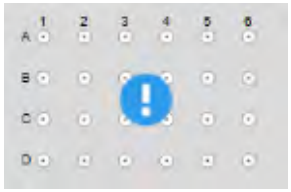
4. Click OK

   The No Scans Scheduled dialog box closes and the Summary page opens.

5. On the Summary page, click Add to Schedule.

   The Add Vessel wizard closes and you return to the Acquisition window. A blue exclamation point is displayed on top of the vessel image in the Vessel Drawer Setup pane, indicating that no scan schedule has been set up for the vessel.

Figure 2-19: No scan scheduled for vessel



6. Continue to "To verify the vessel acquisition settings and schedule the scan" on page 59.

To set the scanning schedule for the Incucyte SX1

The Scan Schedule page that opens for the Incucyte SX1 is different based on whether you are scheduling the first vessel for an experiment, or you are scheduling a subsequent vessel.

If you are scheduling the first vessel for an experiment, then the page displays three scheduling options:

- You can set the scan schedule immediately for the vessel with at least one repeating scan. See "To set the scan schedule immediately with at least one repeating scan" on page 55.
- You can use the advanced scheduling option to set a custom scan schedule for the vessel. See "To use the advanced scheduling option to set a custom scan schedule for the vessel" on page 56.

The option to reserve the vessel tray and defer the scheduling for the vessel until a later time is currently not applicable for the SX1.

See Figure 2-20 on page 58.

If you are scheduling a subsequent vessel for an experiment, then the only option is to add the subsequent vessel to a existing schedule. You cannot create a new schedule for the vessel and you cannot defer the scheduling of the vessel. See Figure 2-21 on page 58.

Regardless if you are scheduling a vessel the first time, or scheduling a subsequent vessel, the Vessel Schedule timeline that is displayed at the top of the page is identical to the Vessel Schedule timeline that is displayed at the top of Acquisition window. (See "Vessel Schedule timeline" on page 36.) The current time is indicated by a single blue line in the Vessel Schedule timeline. The scanning schedule for the vessel that you are scheduling is indicated by white bars in the timeline. The scanning schedule for each vessel that is currently scheduled in the vessel tray is shown as gray bars. The width of each schedule bar correlates to the amount of time that is required to scan the corresponding vessel.

Figure 2-20: Add Vessel wizard, Scan Schedule page for Incucyte SX1 (First scheduled vessel)

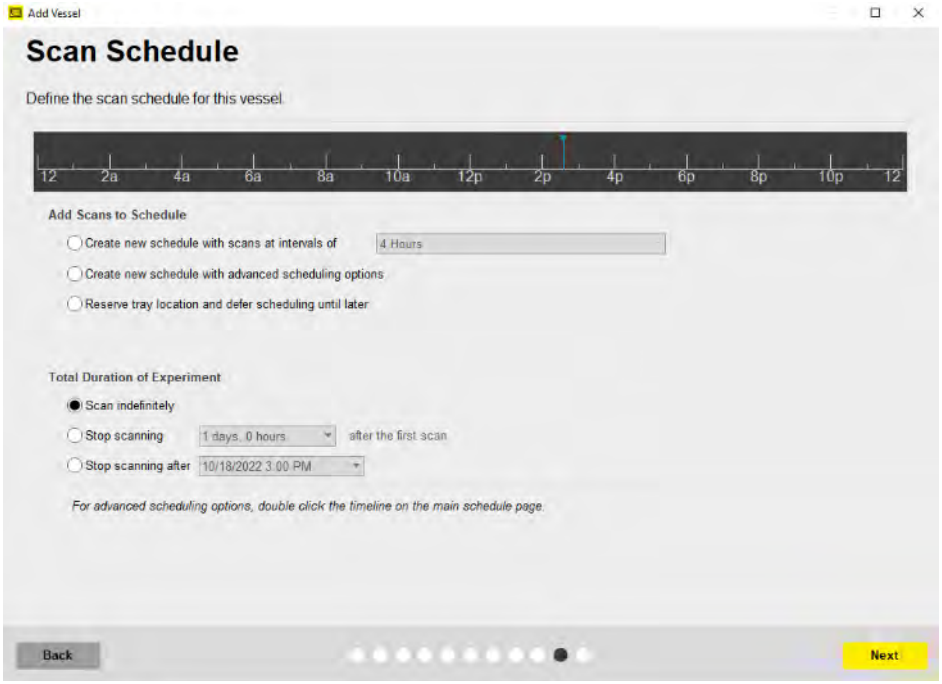
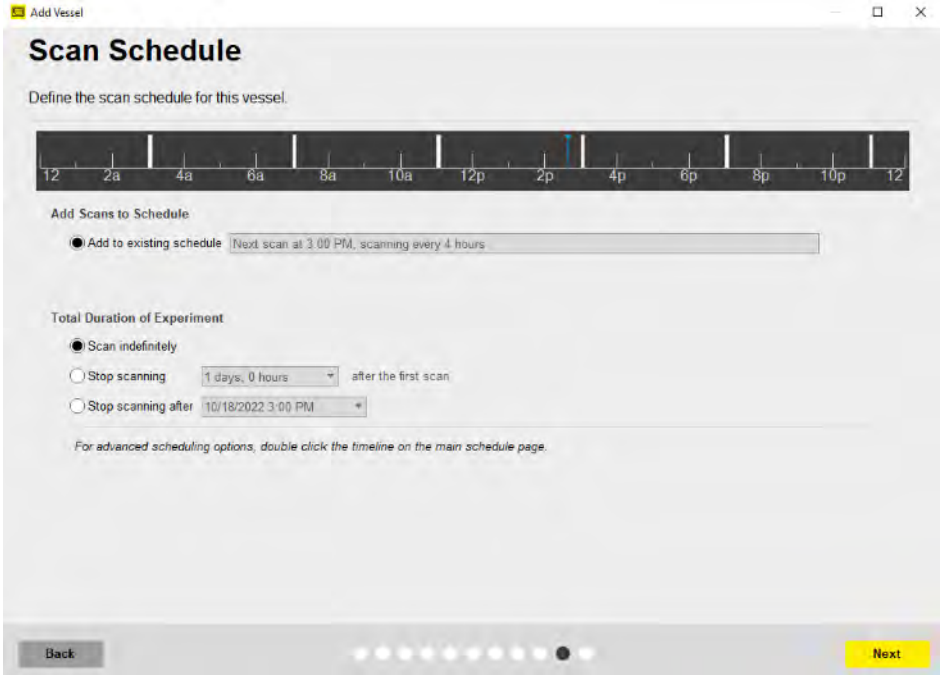
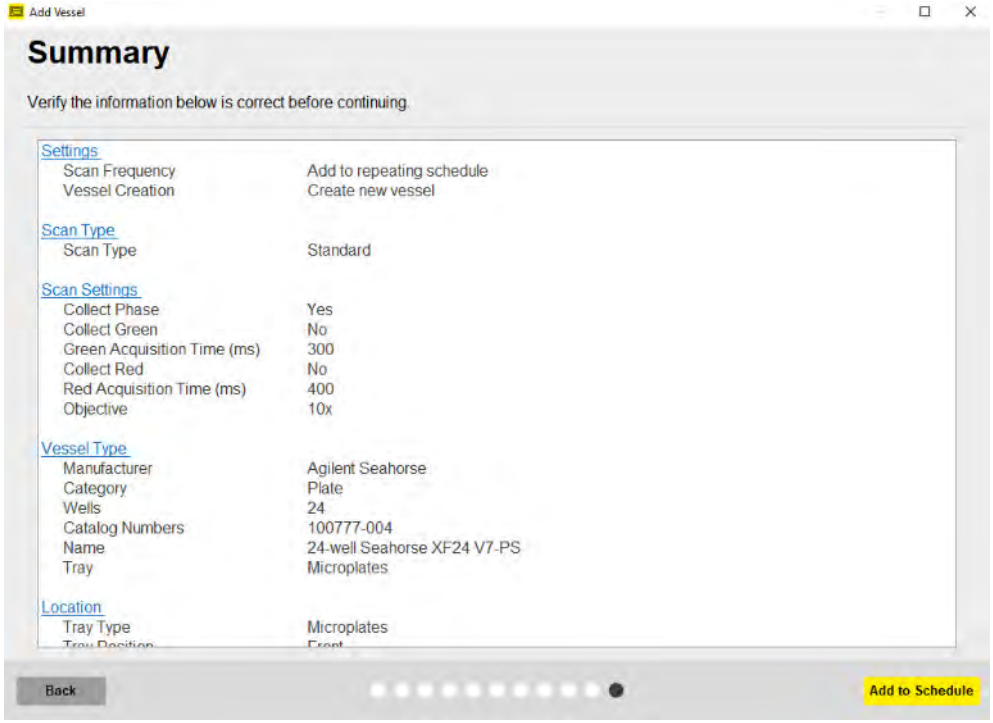


Figure 2-21: Add Vessel wizard, Scan Schedule page for Incucyte SX1 (Subsequently scheduled vessel)



To verify the vessel acquisition settings and schedule the scan

Figure 2-22: Add Vessel wizard, Summary page



The Vessel Summary page displays all the selections/settings that you have specified for the vessel, organized by wizard page. Each section heading corresponds to a wizard page, and is a hyperlink to the indicated page. Hold your mouse pointer anywhere on the page to display the scroll bar (a thin, gray vertical line) at the right side of the page, and then use the scroll bar to view all the information on the page.

1. Review the information for the scheduled vessel, and then do one of the following:
   - If all the selections/settings are correct for the vessel, then continue to Step 2.
   - If you must edit any of the selections or settings, then click the appropriate section header/page name to return to the page in the wizard. Edit the selections/settings as required, and then click Next to move through the wizard (making additional edits if applicable) until you return to the Summary page, and then continue to Step 2.

Any edit that you make to the selections/settings for a scheduled vessel might affect downstream selections/settings. For example, if you select a different vessel type, then you might have to edit the scan pattern for the vessel as well. As a result, you always have to move through the remaining pages on the wizard after you edit any selections/settings for the vessel.

2. Click Add to Schedule.

   A message opens, asking you to wait, and then the message and the Add Vessel wizard closes and the Acquisition window opens. The vessel is displayed and selected (outlined in gray) in the Vessel Drawer Setup pane. The Vessel Information pane displays the scan properties for the vessel.

3. You can now continue in Incucyte.

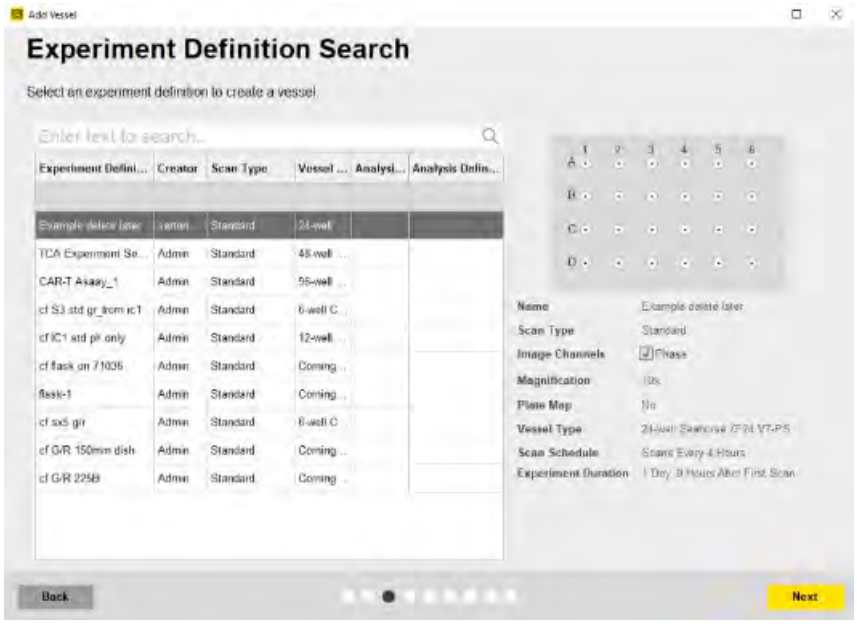
   - If you are satisfied with the vessel information and vessel schedule for all your scheduled vessels, then after the vessels are scanned, any and all options for working with vessel images, analyzing the images, and so on are available to you.
   - If you must make changes to the information and/or the schedule for a scheduled vessel *before* any scan of the vessel has occurred, then see "The Vessel Schedule Timeline and Editing a Vessel Schedule" on page 65 and "Editing Vessel Information" on page 70.
   - If you deferred the scheduling for the vessel, then to facilitate the scheduling of the vessel, you should schedule at least the first repeating scan for the vessel. See "Scheduling One Repeating Scan for a Vessel" on page 62.

Although you are not required to do so, Sartorius strongly recommends that you schedule at least one repeating scan for a vessel before carrying out any other actions for the vessel. This minimizes your chances of making errors when scheduling additional vessel scans.

To create the vessel based on an experiment definition

If you create a vessel based on an experiment definition, then the procedure is *identical* to the procedure for creating a new vessel with the following exception: After you click Experiment Definition on the Create or Restore Vessel page in the Add New Vessel wizard, the Experiment Definition Search page opens.

Figure 2-23: Add Vessel wizard, Experiment Definition Search page



The page lists the experiment definitions that are available to you for creating a new vessel based on *both* the following criteria:

- The physical capability of the Incucyte that you are using to carry out the experiment, for example, does the instrument have specific channels available such as the Orange channel, is there an optional module activated for the instrument, and so on.
- Your access to the experiment definitions that have been created on/imported into the Incucyte.
    - If you are an Admin user, then *every* experiment definition that has been created on/imported into the Incucyte is displayed on the Experiment Definition Search page.
    - If you are not an Admin user, but the experiment definition has been shared with the **Everyone** user workgroup, then the experiment definition is displayed on the Experiment Definition Search page.
    - If you are not an Admin user, or the experiment definition has not been shared with the **Everyone** user workgroup, then only those experiment definitions that have been shared with the user workgroups that you are a member of are displayed.

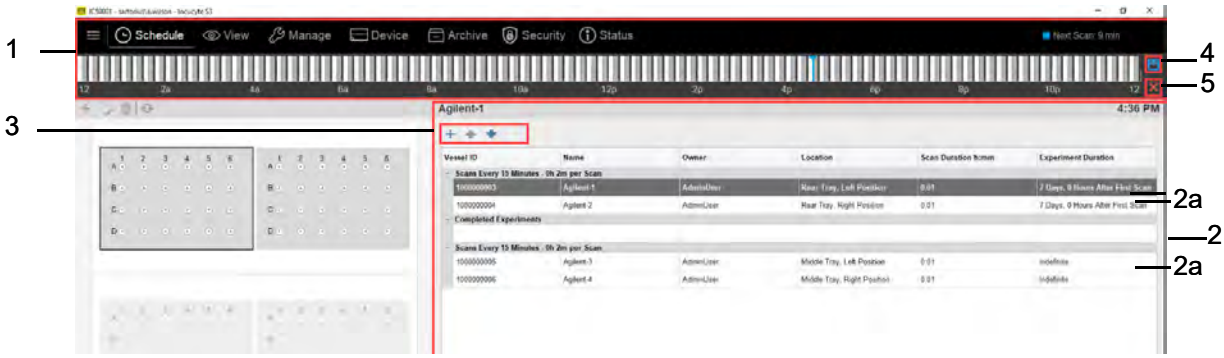
After you select an experiment definition, and then click Next, the wizard pages are identical to those for creating a new experiment vessel. See “To select the vessel location” on page 46.

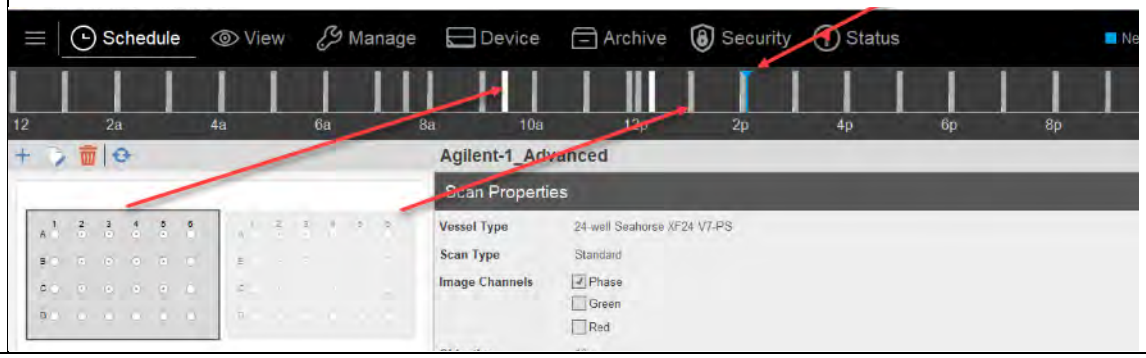
Scheduling One Repeating Scan for a Vessel

After you select any vessel in the Vessel Drawer Setup pane, and then double-click anywhere on the Vessel Schedule timeline on the Acquisition window, the Vessel Information pane is refreshed with the Vessel Schedule table which shows the current schedule information for all the vessels in the Vessel Drawer Setup pane.

After the Vessel Information pane is refreshed with the Vessel Schedule table, additional clicking on the Vessel Schedule timeline can result in a variety of actions being carried out to the vessels that are displayed in the Vessel Drawer Setup pane. To understand what these actions can accomplish and ensure that you properly schedule all your vessels, do NOT click anywhere else on the Vessel Schedule timeline until you read all the information in this section.

Figure 2-24: Acquisition window, Scheduling features for a vessel



Item	Description
1	The Vessel Schedule timeline. This timeline is identical to the Vessel Schedule timeline that is displayed at the top of Acquisition window. See "Vessel Schedule timeline" on page 36. The current time is indicated by a single blue line in the Vessel Schedule timeline. The scanning schedule for the vessel that is currently selected in the Vessel Schedule table is indicated by white bars in the timeline. The scanning schedules for all other vessels are shown as gray bars. The width of each schedule bar correlates to the amount of time that is required to scan the vessels in the corresponding scan group. *Figure 2-25: Vessel Schedule timeline showing schedule for selected vessel* 

Item	Description	
2	The Vessel Schedule table. The table displays the daily scan information (Vessel ID, Vessel Name, Vessel Owner, Vessel Location, Scan Duration in hours:minutes, and Experiment Duration in days/hours) for each vessel that is currently scheduled in the vessel tray, which is referred to as *per vessel scheduling*. Vessels with identical daily scanning schedules are organized in the Vessel Schedule table *by scan groups*. **Note:** When you select a vessel in the Vessel Drawer Setup pane, then the scanning information for the vessel is automatically selected in the Vessel Schedule table. If you select a vessel that is a member of a multi-vessel scan group, then all vessels that belong to the scan group are displayed in dark gray in the Vessel Drawer Setup pane. All other vessels are displayed in light gray in the pane.	
	2a	Scan groups. Vessels with identical daily scanning schedules are organized in the Vessel Schedule table by *scan groups*, with each scan group having its own unique combination of the number of scans per day and the amount of time (in minutes) required per scan. For example, in Figure 2-24 on page 62, two separate scan groups are displayed in the Vessel Schedule table, with two vessels in each scan group. If scanning has been completed for a vessel, then this vessel is displayed in the Completed Experiments scan group.
3	The Vessel Schedule toolbar. From left to right, the toolbar contains the following icons:	
		Create new scan group icon: Applicable only for a multi-vessel scan group.
		Move to previous scan group icon: Applicable only if two or more scan groups are displayed in the Vessel Schedule table.
		Move to next scan group icon: Applicable only if two or more scan groups are displayed in the Vessel Schedule table.
4	Save schedule changes icon: Click this icon to save the changes that you make to an existing vessel schedule.	
5	Cancel schedule changes icon: Click this icon to cancel any changes that you make to an existing vessel schedule.	

To schedule the first repeating scan for a vessel

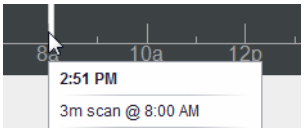
1. In the Vessel Drawer Setup pane, select the appropriate vessel, and then double-click the Vessel Schedule timeline.

   The Vessel Information pane is refreshed and the Vessel Schedule table is displayed. The information for the selected vessel is selected in the table.

2. Click once on the Vessel Schedule timeline to add the first repeating scan for the vessel.

   A white schedule bar is placed at the selected location. You can hold your mouse pointer over the bar to open a tooltip that displays the current time, the duration of the scheduled scan, and the time for which the scan is scheduled.

Figure 2-26: Tooltip for a scheduled scan for a vessel



3. Click the Save schedule changes icon.

   A message opens indicating that the schedule is being saved for the vessel. After the schedule is saved, the Vessel Information pane is refreshed to show the scan properties only for the newly scheduled vessel.

4. You can now edit the vessel schedule to complete the vessel schedule, including adding more scans, moving the vessel to a scan group and so on. You have two options for accessing the options to edit the schedule. You can:

   - Double-click the Vessel Schedule timeline.
   - Right-click the white schedule bar, and on the context menu that opens, click Edit Schedule.

   A message opens indicating that the Vessel Schedule table is being refreshed, and then after the message closes, the Vessel Information pane displays the vessel information only for the newly scheduled vessel.

5. Continue to “The Vessel Schedule Timeline and Editing a Vessel Schedule” on page 65.

The Vessel Schedule Timeline and Editing a Vessel Schedule

The Vessel Schedule timeline that is displayed at the top of the Acquisition window is the tool that you use for advanced scheduling and managing a vessel scan. Using the options on the timeline, you can:

- Add more scans one at a time to an existing schedule.
- Delete a single repeating scan from a scan group.
- Delete a single scan from multiple repeating scans for a scan group, or delete all the repeating scans in a scan group.
- Shift the scan time for a single repeating scan in a scan group.
- Shift the scan time for a single scan in a group of repeating scans for a scan group, or shift all the scan times for all the repeating scans in a scan group.
- Add scans at set intervals to a scan group.
- Delete all scans in all scan groups (regardless of the vessel that is selected) in a single step.

When you are editing a vessel schedule, you also have the option to place a vessel in a different scan group and/or rearrange the order of vessels in a scan group. See "To place a vessel in a different scan group and/or rearrange the order of vessels in a scan group" on page 68.

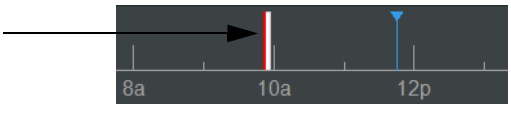
Multiple scan groups are available for the Incucyte SX5 and Incucyte S3. Only a single scan group for all vessels is available for the Incucyte SX1. If you did not schedule the vessel during the Add Vessel wizard, then for the Incucyte SX5 and Incucyte S3, you can use the options that are available on the Vessel Schedule timeline to move the vessel to an existing scan group that has the appropriate scanning schedule, or optionally, you can edit the vessel. You can only edit the vessel for the Incucyte SX1. See "Editing Vessel Information" on page 70.

To edit a vessel schedule using the Vessel Schedule timeline

When you are using the Vessel Schedule timeline to set and/or edit the scanning schedule for a vessel/scan group, note the following:

- If scan times overlap between vessels/scan groups, then a red line is displayed at the overlapping point in the Vessel Schedule timeline. You must change the time for at least one of the scans before you can save the vessel scanning schedule.

Figure 2-27: Overlapping scans indicated in the Vessel Schedule timeline



- At any time that you are scheduling vessel scans, you can click the Cancel schedule changes icon and undo any and all changes that you have made. The vessel schedules are returned to their starting values, vessels are returned to their original scan groups and listed in their original order within their

scan groups, and the Vessel Schedule table closes. The Acquisition window remains open.

1. In the Vessel Drawer Setup pane, select the vessel for which you are editing the schedule.

   After you select a vessel in the Vessel Drawer Setup pane, the vessel schedule is indicated by white schedule bars on the timeline. The schedule for all other vessels are indicated by gray schedule bars. The width of each schedule bar correlates to the amount of time that is required to scan the vessels in the corresponding scan group. The current time is indicated by a single blue line.

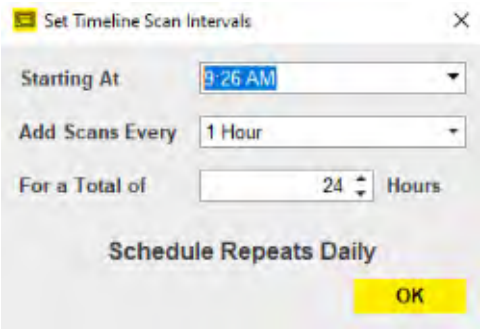
If two or more vessels are members of the same scan group, then when you select any vessel in the group, the color coding on the timeline does not change.

2. Double-click the Vessel Schedule timeline, or on the Vessel Schedule timeline, right-click any schedule bar for the vessel, and on the context menu that opens, click Edit Schedule.

   The Vessel Schedule table opens. The table displays all the schedules for all the vessels in the Vessel Drawer Setup pane that are currently scheduled for scanning, organized by scan group. The Delete schedule changes icon is enabled. The entry for the currently selected vessel is highlighted in the table, and white bars in the Vessel Schedule timeline indicate its corresponding scanning schedule. The Vessel Schedule toolbar is displayed above the Vessel Schedule table.

3. For the selected vessel, the following options are available to set and/or edit its scanning schedule using the Vessel Schedule timeline:

Option	Description
Add a single repeating scan one at a time to the existing schedule	On the Vessel Schedule Timeline, click once at each appropriate time point. A white schedule bar is displayed for every standalone scan that you add.
A *single* repeating scan has been scheduled for the vessel/scan group, or multiple repeating scans have been scheduled and you want to work with only *one* of the repeating scans.	• To delete a single repeating scan, right-click the schedule bar, and on the context menu that opens, select Scan > Delete. **Note:** If you delete a *single* repeating scan for the vessel/scan group, then no scan is scheduled for the vessels in the scan group. An exclamation point is displayed on all the affected vessels in the Vessel Drawer Setup pane. You can move the vessel to an existing scan group that has the correct scanning schedule or use the options that are available on the Vessel Schedule timeline to add a scan and scan intervals. • To adjust the start time for the repeating scan (start the scan earlier or later than its currently scheduled time), right-click the schedule bar, and on the context menu that opens, select Scan, and then select the time. For example, to start the scan 5 minutes earlier, select Scan >: -5 Min. **Tip:** For a *single* repeating scan, you can also click and drag the schedule bar to a different location on the Vessel Schedule timeline

Option	Description
Multiple repeating scans have been scheduled for the vessel/scan group and you want to work with all the scans at the same time	• To delete all the scheduled scans for the vessel, right-click any of the repeating schedule bars, and on the context menu that opens, select Scan Group > Delete All Scans. • To adjust the start times for all the repeating scans for the vessel (start the scans earlier or later than their currently scheduled times), right-click any of the schedule bars, and on the context menu that opens, select Scan Group, and then select the appropriate time. For example, to start all the repeating scans in the scan group 5 minutes earlier, select Scan Group > –5 Min. **Tip:** You can also click and drag any of the schedule bars to a different location on the Vessel Schedule timeline to adjust the start times for all the repeating scans.
Add another repeating scan to the vessel's scan group at set intervals	1. Right-click a schedule bar to open a context menu. 2. On the context menu, select Scan Group > Add Scans at Intervals. The Set Timeline Scan Intervals dialog box opens. **Tip:** You can also right-click anywhere in the open (black) space on the Vessel Schedule timeline to open a context menu with a Set Selected Scan Group Interval option. *Figure 2-28: Set Timeline Scan Intervals dialog box*  3. Set the scan interval values. • Starting At: Sets the time at which the scan is to start. • Add Scans Every: Sets the interval at which the scan is to be repeated. • For a Total of: Restricts the number of hours that the scan is to be repeated out of the 24 hour period. If you are not placing any restrictions on the scan, then leave the value set to the default value of 24 hours. 4. Click OK.

Option	Description
Delete all scans in all scan groups in a single step (the vessel that is selected is irrelevant)	1. Right-click any open (black) space on the Vessel Schedule timeline, and the context menu that opens, click Delete All Scans In All Scan Groups. • After all the scans in all the scan groups are deleted, a blue exclamation point is displayed on top of every vessel image in the Vessel Drawer Setup pane, indicating that no scans have been scheduled for any vessel. • The number of vessels in each can group remains the same, no information (scan name, scan pattern, and so on) is deleted for any vessel, no vessel is deleted from any scan group, and the scan groups themselves are *not* deleted. 2. You must now schedule at least one repeating scan for every vessel in every scan group.

4. After you have successfully scheduled all the appropriate vessels, on the Vessel Schedule timeline, click the Save schedule changes icon.

   The Vessel Schedule table closes. The Acquisition window remains open.

To place a vessel in a different scan group and/or rearrange the order of vessels in a scan group

1. To place a vessel in a different scan group and/or rearrange the order of vessels in a scan group, right-click the selected vessel, and on the context menu that opens, select the appropriate option.

You can also click the vessel entry in the Vessel Schedule table, and then click the appropriate icon on the Vessel Schedule toolbar.

Option	Description
Create New Scan Group	Places the vessel in a new scan group. The scan group is displayed as the last scan group in the list of scan groups in the Vessel Schedule table. You must schedule at least one repeating scan for this vessel/scan group.
Move to Previous Scan Group	Moves the vessel to the scan group that is displayed immediately before the vessel's current scan group in the Vessel Schedule table. The vessel is displayed as the last vessel in the scan group. You can select this option as many times as needed to move the vessel to a different scan group.
Move to Next Scan Group	Moves the vessel to the scan group that is displayed immediately after the vessel's current scan group in the Vessel Schedule table. The vessel is displayed as the last vessel in the scan group. You can select this option as many times as needed to move the vessel to a different scan group.
Move Before	Moves the selected vessel up one row at a time in the list of vessels that are displayed for its scan group. You can select this option as many times as needed to move the vessel to the correct location in its scan group. The vessels are scanned in the order in which they are displayed in their scan group.

Option	Description
Move After	Moves the selected vessel down one row at a time in the list of vessels that are displayed for its scan group. You can select this option as many times as needed to move the vessel to the correct location in its scan group. The vessels are scanned in the order in which they are displayed in their scan group.

If a scan group contains a single vessel, and you move this vessel to a different scan group, then the empty scan group that remains is automatically titled "Experiment Completed." The purpose of the title "Experiment Completed" is to serve as a visual reminder in the event that you inadvertently moved the vessel to a different scan group, you can always move the vessel back to its original scan group. If you did not move the vessel in error, then after you click the Save schedule changes icon, the Experiment Completed scan group is deleted from the Vessel Schedule table.

2. Click the Save schedule changes icon

   The Acquisition window remains open.

Editing Vessel Information

As long as the first scan has *not* started for a vessel, then you can edit any and all of the following information for the vessel: the scan settings, the scan pattern, the vessel information (the vessel notebook), the analysis definition, and/or the scan schedule. If scanning has already started for a vessel, then you can still edit the scanning schedule; however, you cannot edit any other vessel information. Also, if you did not schedule at least one repeating scan for the vessel while configuring the vessel in the Add Vessel wizard, then you can edit the vessel to set its scanning schedule.

To edit vessel information

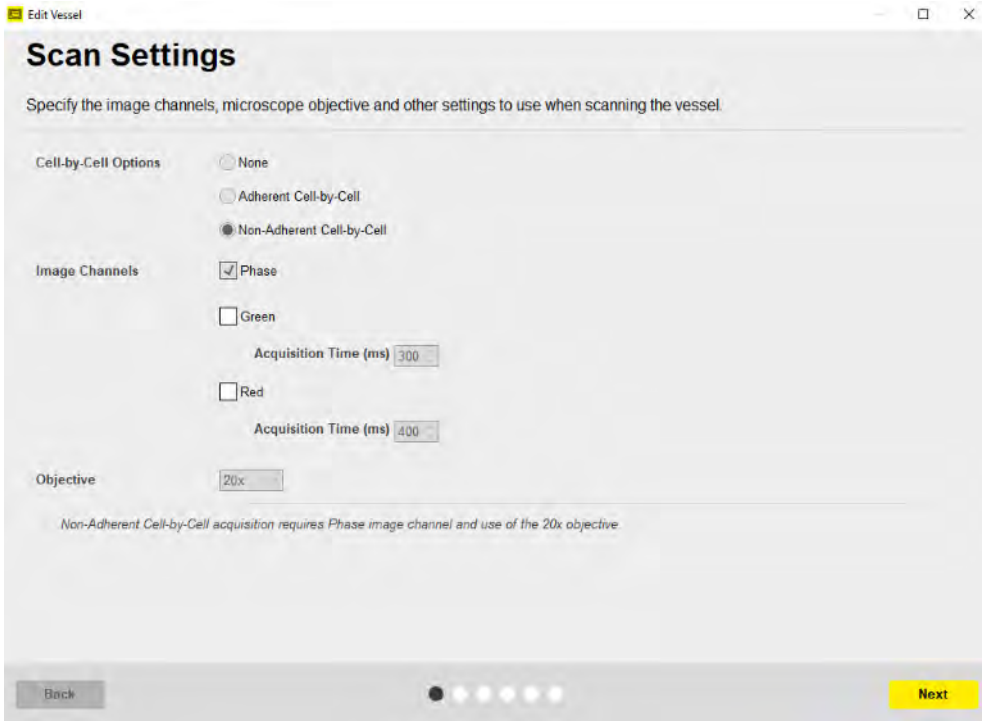
To prevent the selected vessel type from being invalidated, you cannot change the objective.

1. In the Vessel Drawer Setup pane, select the vessel that you are editing, and then on the Vessel toolbar, click the Edit selected vessel icon.

If the Edit Selected Vessel icon is not enabled, click the Refresh Schedule icon.

The Add Vessel wizard opens in Edit mode. The Scan Settings page is the open page.

Figure 2-29: Add Vessel wizard in Edit mode, Scan Settings page



2. Edit the selections/settings, and then click Next to move through the wizard (making additional edits as required) until the Summary page opens.

The Summary page that is display when you are editing vessel in formation is almost identical to the Summary page in the Add Vessel wizard, and you can interact with it the same way. *See "To verify the vessel acquisition settings and schedule the scan" on page 59.*

3. After you have made all the necessary edits for the vessel, click Save Changes.

   The Add Vessel wizard closes. The Acquisition window remains open with the vessel selected in the Vessel Drawer Setup pane.

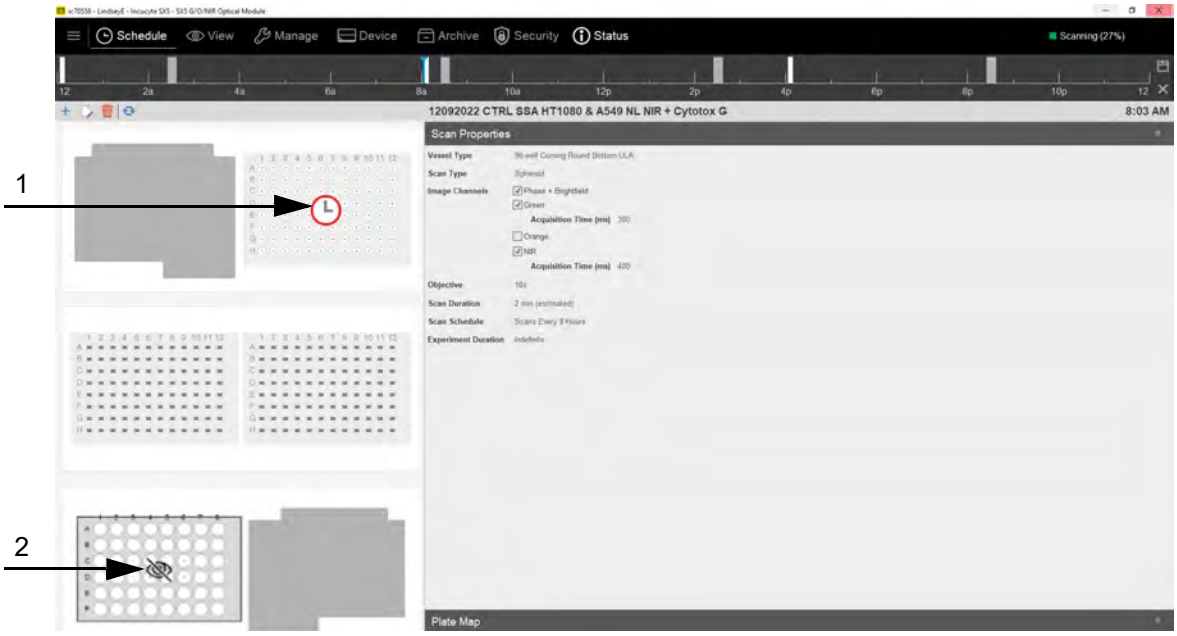
Removing a Vessel from the Scanning Schedule

When you remove a scheduled vessel, the vessel is no longer displayed in the Vessel Drawer Setup pane. You are *not* deleting the vessel and/or its associated data from the database. You can remove any scheduled vessel that is shared with the **Everyone** user workgroup, or if you are member of the user workgroup for the vessel, even if scans have already been acquired for the vessel.

- If the scanning duration for a vessel is complete, then a (1) Clock icon is displayed on the vessel image on the Acquisition window. If you select the vessel image, then “Completed” is displayed in the scan properties for the vessel.
- If you are not a member of the user workgroup for the scheduled vessel, then a (2) Not Viewable icon is displayed on the vessel image on the Acquisition window, and you cannot remove the scheduled vessel.

If you select the vessel image, then the Vessel ID and the user (login) name of the user who scheduled the vessel are displayed at the top of the Vessel Information pane, and you can contact the user about removing the scheduled vessel.To understand how user workgroups impact vessels and their data, see “Managing User Workgroups” on page 254.

Figure 2-30: Clock icon displayed on a scheduled vessel for which the scan has been completed and Non-viewable icon displayed on a scheduled vessel that you cannot remove from the schedule



If you can remove a *scheduled* vessel, note the following:

- You can remove only a single vessel at a time.
- If the vessel is the only vessel in its scan group, then its scanning schedule is also removed from the Vessel Schedule timeline.
- If the vessel is part of a scan group, then the scanning schedule for the scan group remains displayed

on the Vessel Schedule timeline.

- Removing a vessel does not impact any scans that have already been acquired for the vessel. Going forward, scans are simply not acquired for the vessel.

To remove a vessel from the scanning schedule, in the Vessel Drawer Setup pane, select the vessel, and then on the Vessel toolbar, click the Remove selected vessel icon.

To delete a vessel from the Incucyte database, you use the Delete function on the Vessel Delete tab on the Manage window. See Appendix A, "Incucyte Management," on page 181.

Section 2
Viewing Images

Section Contents

Chapter 1
Selecting a Vessel

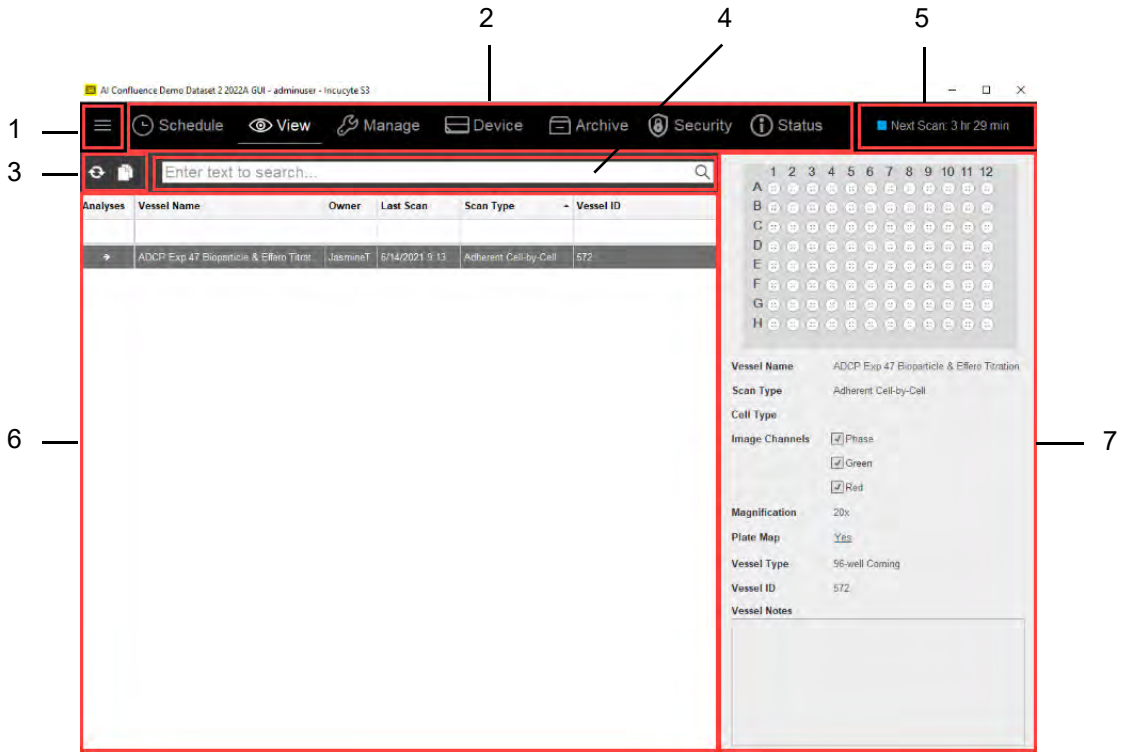
The first step in analyzing images is to select the vessel in the database that contains the images that you are analyzing. This chapter details the Scanned Vessels window, which displays all the vessels that have ever been scanned on your Incucyte system by any user, as well as all the options that are available on the window for selecting the correct vessel.

This chapter covers the following topics:

The Scanned Vessels Window

The Scanned Vessels window displays all the vessels that have ever been scanned on your Incucyte system by any user. To open the Scanned Vessels window, on the Incucyte main window, click View.

Figure 1-1: View window

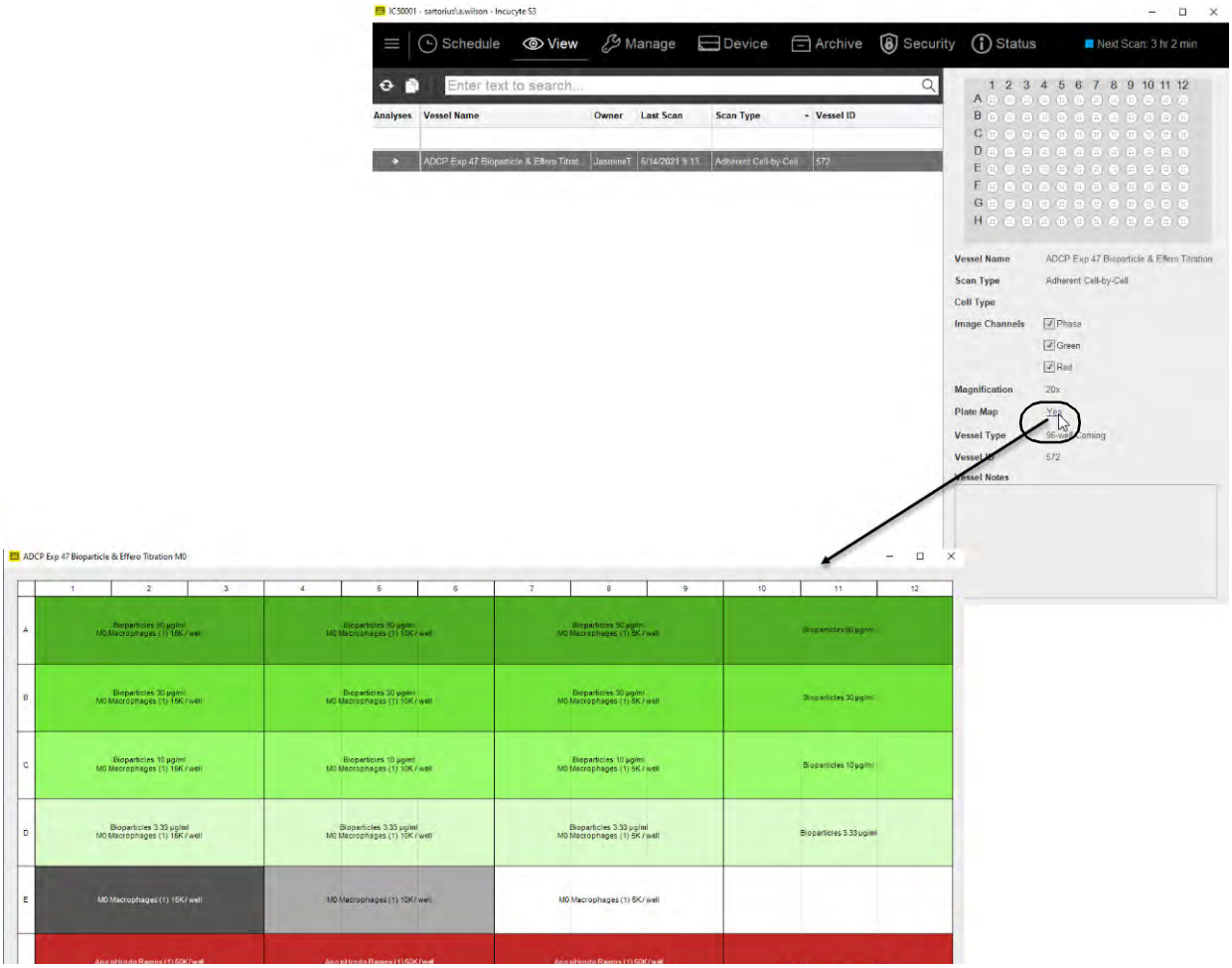


The Scanned Vessels window has the following layout:

	Window component	Description
1	Menu bar	Opens a dropdown menu with a variety of options. See “Application menu” on page 21.
2	Window menu	The menu is always displayed at the top of every window in Incucyte. The window menu displays all the same options that are available on the Incucyte main window. See “Incucyte Window menu” on page 24.
3	Scanned Vessels toolbar: Displays two icons for working with vessels in the Scanned Vessels window:	
		Refresh Vessels icon: Refreshes the display of the vessels in the Vessels pane.
		Copy Grid icon: Copies all the information that is currently displayed in the Vessels pane to your client’s clipboard. You can then use the standard Paste commands to paste the information in to a third-party application such as Microsoft Excel.

	Window component	Description
4	Search field	To search for a specific vessel or vessels across all columns, enter your search criteria in this field. See “To search for data in an Incucyte Window” on page 29.
5	Scheduling status	Color-coded to indicate a variety of statuses about your Incucyte system. See “Scheduling Status” on page 25.
6	Vessels pane	Displays all the vessels that have ever been scanned on the instrument by any user. When the Scanned Vessels window first opens, the vessels are sorted in the pane in reverse chronological based on the scan date/time stamp. An Expand/Collapse icon in the Analysis column indicates that the images that were acquired for the vessel have been analyzed. See “The Vessels Pane” on page 82. **Note:** The Vessels pane is not dynamically updated after a vessel is scanned. You must click the Refresh Vessel icon to update the display after a vessel is scanned.
7	Vessel Information pane	Displays the vessel acquisition map (the vessel locations for which images were acquired as well as the number of images that were acquired per vessel location) and all the information that was defined in the Add Vessel wizard for a selected vessel. If the vessel is a microplate and a plate map was defined for the vessel, then next to the plate map entry, you can click Yes to open a read-only display of the map. See Figure 1-2 on page 81.

Figure 1-2: Scanned Vessels window, Vessel Information pane for a plate map

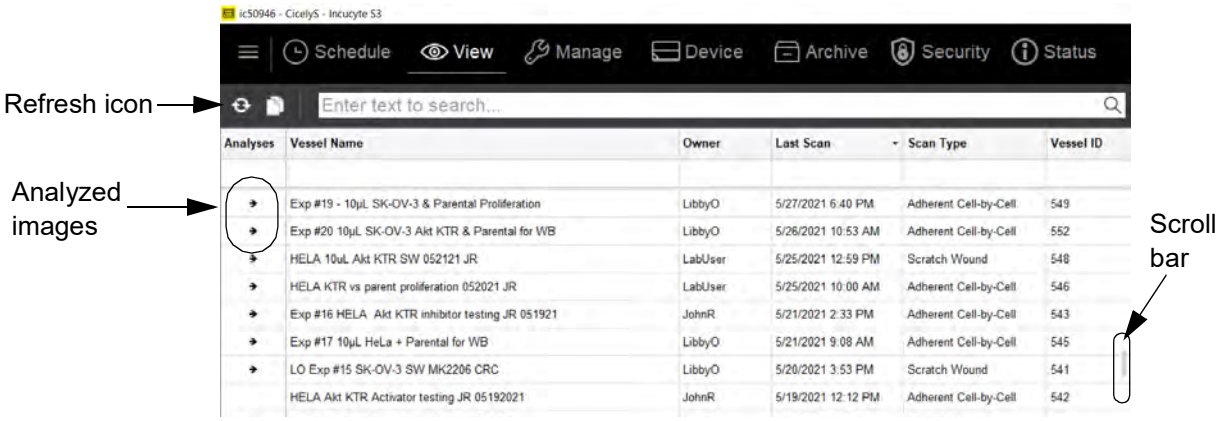


The Plate Map display is shown as offset from the Scanned Vessel window for clarity only. When the Plate Map display opens, it opens over the Scanned Vessels window. To close the Plate Map display and return to the Scanned Vessels window, click the Close (x) icon in the upper right corner of the display.

The Vessels Pane

The list of scanned vessels on the Scanned Vessels window is displayed in the Vessels pane. All the vessels that have been shared with the **Everyone** user workgroup and all the vessels that are shared in the user workgroups to which you have been assigned are displayed in the pane. When the window first opens, the vessels are sorted in the pane in reverse chronological order based on the scan date/time stamp. By default, the first vessel that is displayed in the list of vessels is selected. An Expand/Collapse icon in the Analysis column indicates that the images that were acquired for the vessel have been analyzed.

Figure 1-3: Scanned Vessels window, Vessels pane



Note the following about the Vessels pane:

- The Vessels pane is not dynamically refreshed after a vessel is scanned. To ensure that all scanned vessels are being displayed in the pane, you should periodically click the Refresh Vessels icon.
- You can use the Up and Down arrow keys on your client keyboard to scroll through the display, or you can use the Scroll bar on the right side of the pane.

If the scroll bar is not displayed, click once on the far right side of the pane to display it. The scroll bar is a thin, gray vertical line.

- In addition to the standard features that are available for most Incucyte windows (see "Working with Data Columns in an Incucyte Window" on page 26*), the following actions are available for the Vessels pane:
  - You can view the analysis definitions that are associated with a scanned vessel. See "To view the analysis definitions for a scanned vessel" on page 83.
  - You can open the Vessel View window for the selected vessel. See "To open the Vessel View window" on page 85.

Sort and filter functions are not available for the Analyses column in the Vessels pane.

To view the analysis definitions for a scanned vessel

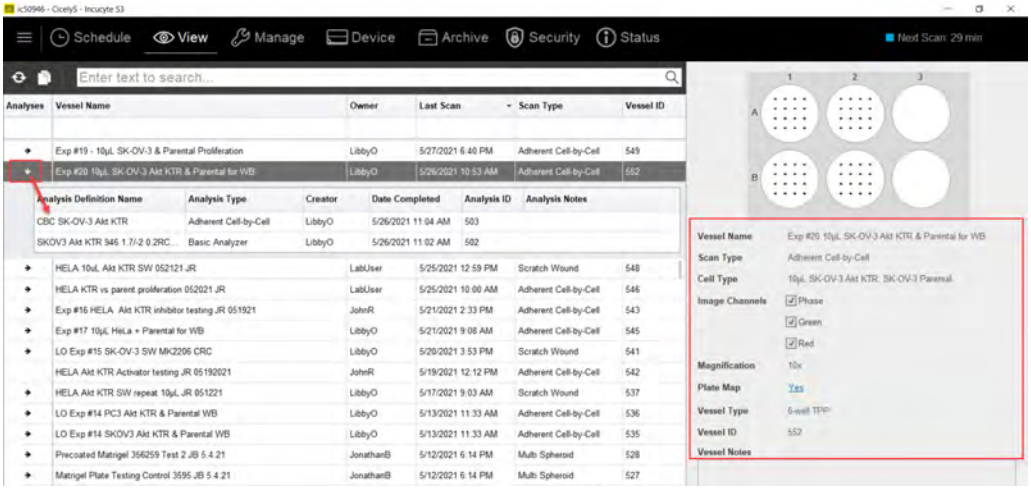
If the images have been analyzed for a scanned vessel, then to view the associated analyses (type, definition, and so on), click the Analysis arrow. The Analysis arrow is now displayed vertically, and an Analysis pane that displays the associated analysis information opens. Multiple analyses for a single vessel are allowed.

Figure 1-4: Analysis definitions associated with a scanned vessel

Although the Analysis pane is shown with columns, no sort functions are available.

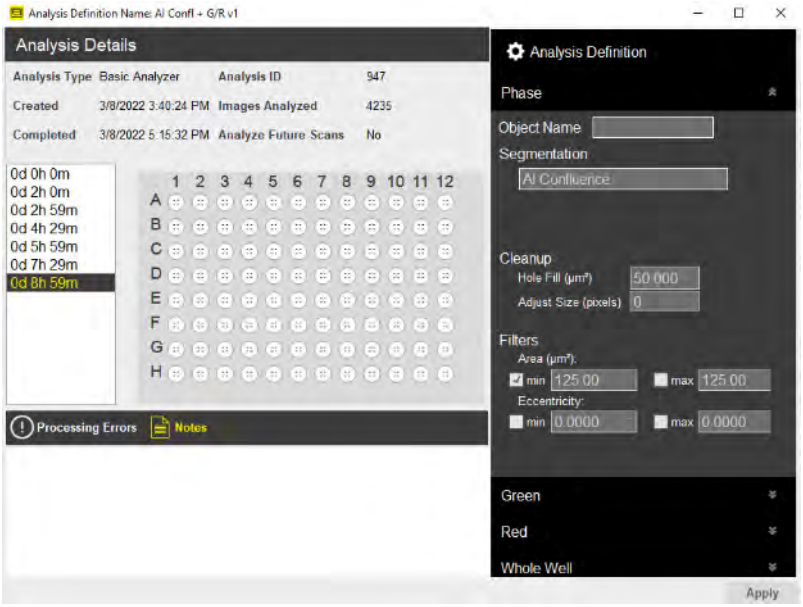
To update the Vessel Information pane so that the vessel information for only a specific analysis is displayed, select the analysis. The vessel graphic at the top of the Vessel Information pane is updated to show only those vessel locations for which the selected analysis is applicable. See Figure 1-5 below.

Figure 1-5: Vessel Information pane displaying analysis information



You can right-click any analysis for a selected vessel to open a context menu with the following analysis options:

Option	Description
Open Analysis	Opens the Vessel View window, which now displays the associated analysis masks that were applied to the images. See "Viewing the Analysis Masks" on page 137. **Tip:** You can also double-click an analysis to open the Vessel View window and view the analysis masks.

Option	Description
Graph Analysis Metrics	Opens the Graph Metrics window, which provides options for graphing the metrics that are associated with the analysis without having to open the vessel. See "The Graph Metrics Window" on page 138.
View Analysis Details	Opens the Analysis Details window, which details all the parameters that were specified in the Launch Analysis wizard for analyzing the images in the vessel. The parameters that are displayed depend on the analysis type, for example, Basic Analyzer, that was selected for the vessel. *Figure 1-6: Analysis Details window* 
Export Analysis Definition	Opens the Export Analysis Definition dialog box, which provides the options for exporting the full definition for the selected analysis. See "To work with an analysis definition (Context menu options)" on page 197. *Figure 1-7: Export Analysis Definition dialog box*  **Tip:** This option is particularly useful if your organization has two or more Incucyte systems. Instead of setting up the same definition on each of the instruments, you can set up the definition one time on one instrument, and then export this definition to all your other instruments.
Delete Analysis	Deletes the selected analysis for the vessel. **Warning:** If you delete a selected analysis for a scanned vessel, then only the analysis data is deleted. Neither the vessel nor its images are deleted from the Incucyte database.

To open the Vessel View window

The Vessel View window displays the scanned vessel that has been selected for image analysis. The window provides the options for viewing, comparing, and ultimately selecting the images that are to be analyzed. To open the Vessel View window for a scanned vessel, do one of the following in the Vessels pane in the Scanned Vessels window:

- Right-click the vessel, and on the context menu that opens, click Open Vessel.
- Double-click the vessel.

See “The Vessel View Window” on page 89.

Chapter 2
Viewing and Working With Images

After you have selected the vessel in the database that contains the images that you are analyzing, you can then view the images and adjust the image properties in a variety of ways to examine and compare the images. For example, you can set a range of minimum and maximum intensities to help you determine the areas of the image that have the highest intensities, you can turn channels on and off, and you can remove signal in a given channel (spectral unmixing). Based on all the information that you collect from this examination and comparison, you can then decide which vessel locations are appropriate for image analysis, and how you are analyzing the images. This chapter details the Vessel View window, which displays the scanned vessel that has been selected for image analysis, and all the options that are available on the window for viewing, comparing, and ultimately selecting the images that you are analyzing.

This chapter covers the following topics:

The Vessel View Window

The Vessel View window displays the scanned vessel that has been selected for image analysis. The window provides the options for viewing, comparing, and ultimately selecting the images in the vessel that you are analyzing. To open the Vessel View window for a scanned vessel, do one of the following in the Vessels pane:

- Right-click the vessel, and on the context menu that opens, click Open.
- Double-click the vessel.

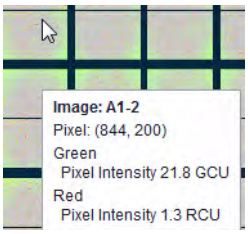
Figure 2-1: Vessel View window (96-well microplate)

The Vessel View window has the following layout:

	Window component	Description
1	Image toolbar	The Image toolbar contains options for viewing and working with the images that have been acquired for the scanned vessel. See "The Image toolbar" on page 95.
2	Analysis toolbar: The Analysis toolbar contains two icons.	
		Launch Analysis icon: Opens the Launch Analysis wizard, which you use to define the analysis for the images in the scanned vessel. See Chapter 1, "Defining the Image Analysis," on page 113.
		Graph Metrics icon: Opens the Graph Metrics window, which provides options for graphing the *scan* metrics for the vessel. See Chapter 2, "The Graph Metrics Window," on page 138.

	Window component	Description
3	Visualization toolbar: The Visualization toolbar contains two icons.	
		Vessel Information icon: Opens the Vessel Information window, which displays different information about the scanned vessel. See “The Vessel Information window (via the Vessel View window)” on page 92.
		Export Images and Movie icon: Launches the Image Export wizard. See “Exporting Images” on page 100.
3	Navigation toolbar	The Navigation toolbar contains options for navigating the Vessel View display, including zooming in, zooming out, and moving through the scans for the vessel. See “The Navigation toolbar” on page 99.
5	Measure toolbar: The Measure toolbar contains two icons:	
		Measure image features icon: Used to measure the distance between two points on an image. The unit of measure that is available, for example, microns, depends on the measured size of the image feature. To use the Measure image features function, click the Measure image features icon, and then drag your mouse pointer across an image in a vessel location to measure the distance from the starting point to the ending point. The value is displayed in the lower right corner of the Vessel View window. **Tip:** After you complete the measurement, you can click the Measure image features icon again to clear the measurement information that is displayed on the image. **Note:** When defining analysis parameters for vessel images, if Top-Hat color processing is selected, then you must use this feature to set the radius parameter.
		Legend icon: Toggles the display of a legend for the measurement scale. The scale is displayed in the lower left corner of the Vessel View window. The unit of measurement depends on the zoom level.

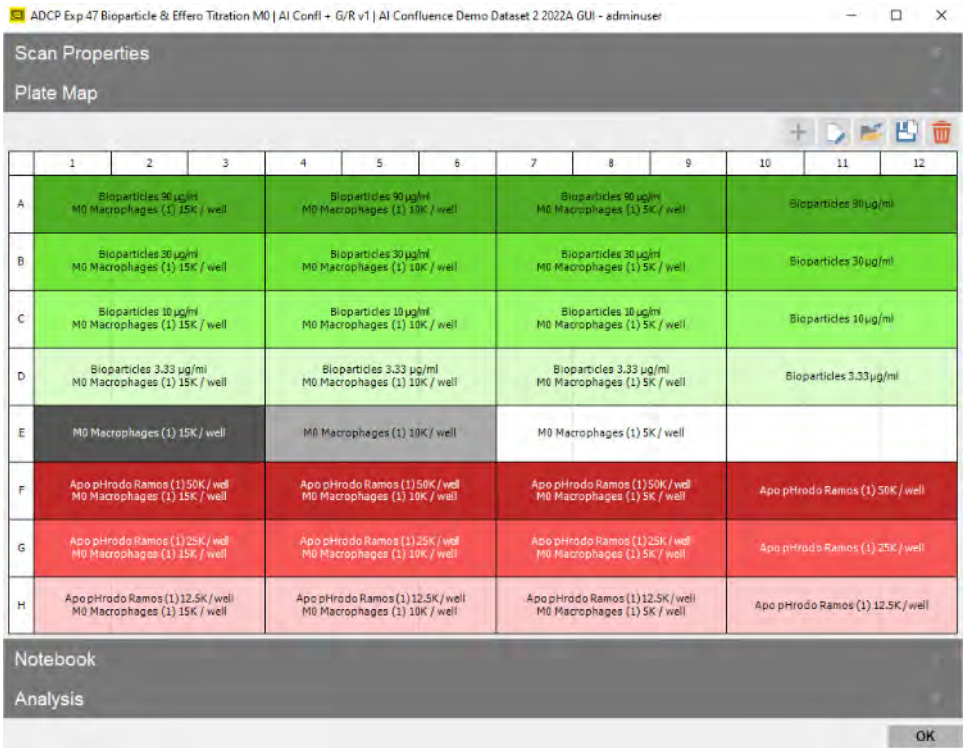
	Window component	Description

6	Vessel View display	Displays the acquisition map for the scanned vessel. The number of images that have been acquired for each vessel location (as specified in the Add Vessel wizard) is indicated by the number of individual squares in the vessel locations. For example, in Figure 2-2 below, one image per vessel location is indicated for Figure A while four images per vessel location is indicated for Figure B. The display is dynamically updated based on the scan time that is selected on the Vessel Scanning timeline. (See "To select a scan time for the Vessel View display" on page 95.)
		Figure 2-2: Comparing the number of images in a vessel location
		 
		
		Figure A. Figure B.
		The following options are available for an image site:
		• You can hold your mouse pointer over an image site to open a tooltip that displays information about the image.
		Figure 2-3: Image tooltip in the Vessel View window
		 
		
		• You can right-click an image site to open a context menu with the following options:
		  • Copy viewing area to clipboard: Copies the *viewing area* (the area in the Vessel View window where the vessel's contents are displayed) to your client's clipboard. You can then paste this image into a third-party application such as PowerPoint.
		  Note:: The viewing area does not include the title bar or the toolbars and is therefore less than the full area of the Vessel View window. Because you can zoom in and zoom out the Vessel View display, the contents that are displayed in the viewing area are dynamic. If the entire vessel is shown for the Vessel View display, then this is the viewing area that is copied to your client's clipboard. If you have zoomed in the display to show a limited portion of the vessel (for example, 6 wells out of a 96-well plate), then only this limited portion is copied to your client's clipboard.
		  • Export Image via Designer: Opens the Export Image wizard. See "Exporting Images" on page 100.
		  • Export Image as Stored: Opens the Export Image wizard. See See "Exporting Images" on page 100.

The Vessel Information window (via the Vessel View window)

The Vessel Information window has up to four different information tabs: Scan Properties, Plate Map, Notebook, and Analysis. When the window first opens, if the vessel is a microplate, then the Plate Map tab is the open tab; otherwise, the Notebook tab is the open tab. When the Vessel Information window first opens, the window is minimized and opens on top of the Vessel View window, but the Vessel View window is still visible.

Figure 2-4: Vessel Information window (from the Vessel View window), Plate Map tab



Vessel Information window, Plate Map tab

The Plate Map on the Vessel Information window tab is identical to the Plate Map tab from the Acquisition window with one exception: Unlike the Plate Map tab from the Acquisition window, the Plate Map tab from the Vessel View window is *not* read-only. Instead, all the functionality that is associated with the Plate Map Editor is available. See Appendix A, “Plate Map Editor,” on page 167.

Vessel Information window, Scan Properties tab

The Scan Properties tab is a read-only tab that is identical to the Scan Properties tab in the Vessel Information pane in the Acquisition window. See“Vessel Information pane” on page 37.

Vessel Information window, Notebook tab

The Notebook tab for the Vessel Information window is identical to the Notebook tab in the Vessel Information pane in the Acquisition window. (See “Vessel Information pane” on page 36.) The following functions are available on the Notebook tab:

- Editing vessel and/or analysis information. See “To edit vessel and/or analysis information” below.
- Adding vessel notes. See “To add vessel notes” below.
- Copying vessel notes. See “To copy vessel notes” on page 94.
- Changing the workgroups with which the vessel is shared. See “To share a vessel with a user workgroup” on page 51.

To edit vessel and/or analysis information

You can edit the vessel name, the cell type and/or the passage, and then click OK to save your edits. These values are displayed in the Vessel Information pane in the Scanned Vessels window. See “The Scanned Vessels Window” on page 79.

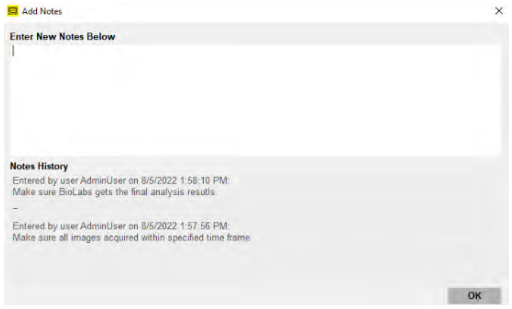
After you click OK to save your edits, the Vessel View window closes. Optionally, you can make your edits to the analysis information and then continue your work in any of the other views before clicking OK to save all your edits in all views in a single step.

To add vessel notes

1. Click the Add Note icon  .

   The Add Notes dialog box opens. Notes that have already been added to the vessel are displayed under Notes History with the name of the user who added the note and the date and time stamp of the addition in reverse chronological order.

Figure 2-5: Add Notes dialog box



2. Enter the notes where indicated in the dialog box, and then click OK to save the notes.

   The Add Notes dialog box closes and the Notebook tab remains open. The notes that you added are displayed last in the list of existing notes along with your user name and the date and time stamp of the added notes.

To copy vessel notes

You can copy *all* notes for the vessel in a single step to your clipboard, which makes them available for pasting into a third-party application such Microsoft Word. To copy all the notes for the vessel in a single step, click the Copy Notes icon  .

You cannot copy a single note at a time, or select multiple notes to copy. The Copy Notes function is all or nothing.

Vessel Information window, Analysis tab

The Analysis tab displays the read-only analysis details and analysis definition for every scan time for the vessel. The following options are also available on the Analysis tab.

- Viewing processing errors that are associated with the vessel analysis. See "To view analysis processing errors" below.
- Viewing, adding, or editing notes for the vessel analysis. See "To view, add, or edit analysis notes" below.

To view analysis processing errors

At the bottom of the Analysis Details tab, click Processing Errors to open the Processing Errors pane. If applicable, the pane displays any processing errors that are associated with the vessel analysis. If no processing errors are associated with the analysis, then the pane is blank.

To view, add, or edit analysis notes

The notes that you add here are unique to the Analysis tab and are not displayed on the Notebook tab. Analysis notes are displayed in the Vessel Information pane in the Scanned Vessel window. See "The Scanned Vessels Window" on page 79.

At the bottom of the Analysis Details tab, click Notes to open the Notes pane. If no notes have been added for the analysis, then the pane is blank; otherwise, the pane displays all the notes that have been previously added for the analysis.

- To add vessel analysis notes, click in the Notes pane, enter the notes, and then click OK to save the notes.
- To edit an existing note, use standard text editing functions such as highlighting, backspacing, and so on, and then click OK to save the edited notes.

After you click OK to save your edits, the Vessel View window closes. Optionally, you can make your edits to the analysis information and then continue your work in any of the other views before clicking OK to save all your edits in all views in a single step.

The Image toolbar

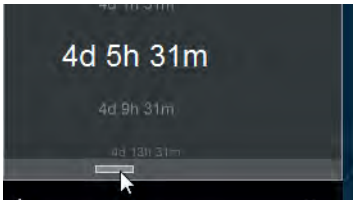
The Image toolbar on the Vessel View window contains options for viewing and working with the images that have been acquired for the scanned vessel. Click an icon on the toolbar to access the associated functions.

Icon	Description
	Scans icon: Opens the Vessel Scanning Timeline popup. See "To select a scan time for the Vessel View display" below.
	Layers icon: Opens the Image Layers popup. See "To work with the image channels" on page 96.
	Tools icon: Opens the Tools popup. See "To work with the Vessel View display" on page 98.
 	Expand/Collapse icon: Expands and collapses the Image toolbar. When the Vessel View window is first opened, the icon is pointing to the right, which indicates that the Image toolbar is collapsed. Click the icon to expand the Image toolbar. The Expand/Collapse icon is now pointing to the left. After you expand the Image toolbar, an Expand/Collapse icon is displayed to the right of each icon on the toolbar and is pointing up, indicating that the additional information/functions for each toolbar icon is collapsed. You can click each of these Expand/Collapse icons to access the same information that you would by directly clicking the Image toolbar icons.

To select a scan time for the Vessel View display

The Vessel Scanning Timeline popup displays all the time points at which the vessel was scanned (xd xh xm), with the scan times listed in ascending order, with the first scan time (0d 0h 0m) always at the top of the list.

Figure 2-6: Vessel Scanning Timeline popup



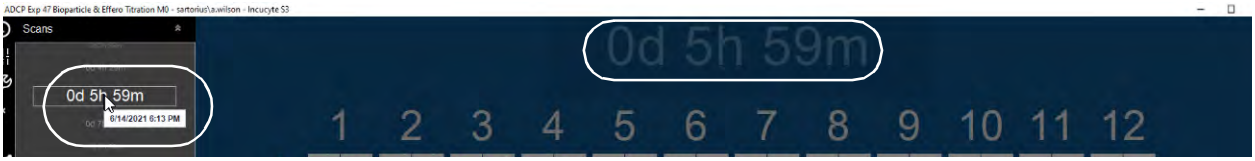
When the Vessel View window *first* opens, the results of the most recent scan for the vessel are displayed. To select a different scan time, do any of the following:

- Click the scan time in the Vessel Scanning Timeline popup.
- Click at the bottom of the Vessel Scanning Timeline popup to display a horizontal gray scroll bar, and then click and drag the scroll bar to move through and select a scan time.
- Click any scan time in the Vessel Scanning Timeline popup, and then use the Up and Down arrows on your client keyboard, or use your mouse scroll wheel.

- Use the Next Scan and/or Previous Scan icons on the Navigation toolbar. See “The Navigation toolbar” on page 99.

After you select a scan time, it is displayed in white and in a larger font than the other scan times in the Vessel Scanning Timeline popup. The Vessel View display is dynamically updated based on the selected scan time and the selected scan time is temporarily shown at the top of the display. You can hold your mouse pointer on the selected scan time in the popup to open a tooltip that displays the exact date and time that the scan occurred.

Figure 2-7: Vessel Scanning Timeline popup with tooltip

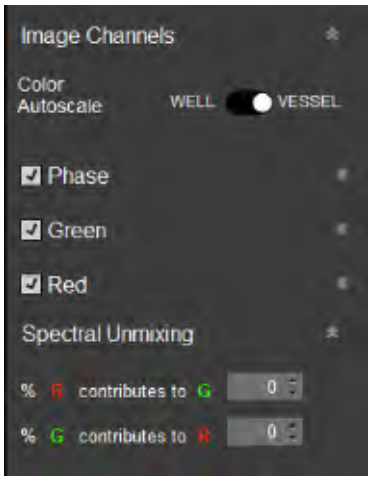


To work with the image channels

The Image Layers popup provides options for working with the channels that were used when acquiring the vessel images, including auto-scaling, turning on and/or turning off channels, adjusting the brightness and contrast for the Phase channel (if applicable), and spectral unmixing. See:

- “Color Autoscale” below.
- “Spectral Unmixing” on page 97.

Figure 2-8: Image Layers popup



Color Autoscale

Color Autoscale automatically sets a range of minimum and maximum intensities to account for changes in biological samples across different samples and time. When viewing a vessel in the Vessel View window:

- If Vessel (the default value) is selected, then an intensity range is automatically applied across the entire vessel based on the minimum and maximum brightness of the entire vessel. If you turn off Autoscale for a channel with Vessel selected, then you can manually adjust the intensity range for the channel.

- If Well is selected, then an intensity range is automatically applied *to each vessel location*. If you turn off Autoscale for a channel with Well selected, then the intensity range for the channel is automatically set for the entire vessel based on the intensity range for the image that is closest to the viewport. For example, if the vessel view is not zoomed for a 96-well microplate, then the intensity range for the channel would be set for the entire microplate based on the intensity range for the image that is in Well D6. If the vessel view is zoomed in for a 96-well microplate to Well A1, then the intensity range for the channel would be set for the entire microplate based on the intensity range for the image that is in Well A1.

For information about navigating the Vessel View display, including zooming in and zooming out, see "The Navigation toolbar" on page 99.

Spectral Unmixing

The Spectral Unmixing option is displayed only if more than one fluorescent channel has been used for image acquisition. For example, if a fluorophore was used that produces signal in both the Red and Green channels, then spectral unmixing might be necessary to account for signal that has been contributed from one of the given channels. In this example, green fluorophores typically do not require any spectral unmixing while red fluorophores require a small amount of adjustment (2% - 8%) to account for contribution in to the green channel. You must apply spectral unmixing values before you run an analysis job. To ensure the most appropriate values, you should make adjustments in small increments. After you change one or more spectral unmixing values, two icons are displayed for the Spectral Unmixing options on the Image Channel popup - a Save icon and an Undo icon. To save the changes that you make to the spectral unmixing values, you must always click the Save icon. At any time after you make an edit, but before you click Save, you can click the Undo icon to return the spectral unmixing values to their last *saved* values.

Figure 2-9: Spectral unmixing percentages edited

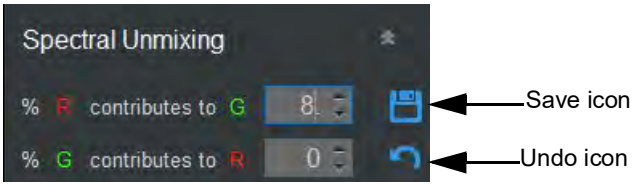
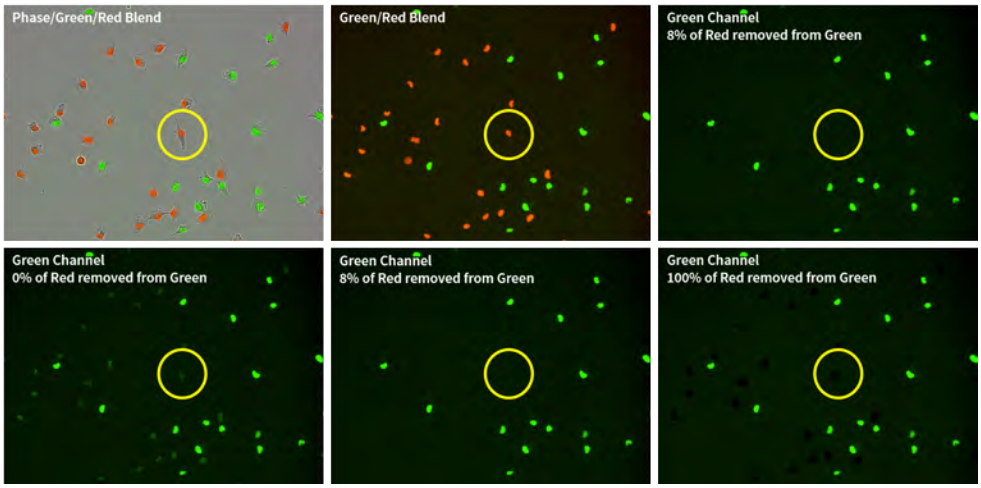


Figure 2-10 on page 98 is an example of correct and incorrect spectral unmixing. In this figure, the top row shows blended images of a mixed population of HT-1080 Nuclight Red and Nuclight Green cells. The bottom row demonstrates optimized spectral unmixing. Using the HT-1080 Nuclight Red cell that is circled in yellow as a guide, the red fluorophore is visible in the green channel with an average Green Calibrated Unit (GCU) of 8.2 and an average Red Calibrated Unit (RCU) of 29.2. Optimization of the red fluorophore in Incucyte showed a spectral unmixing adjustment for 8% of the red contributing to the green was required (GCU - (RCU x 0.08) = the amount of red signal that contributed to the green channel). If the amount of red signal contributing to the green channel is set too high, then over corrected pixels develop, giving the appearance of "pits" or "holes" in the images, which can affect assay metrics.

Figure 2-10: Spectral unmixing of a mixed population of HT-1080 cells expressing Nuclight Green or Red



To work with the Vessel View display

The Tools popup contains options for working with and changing the information that is shown for the Vessel View display.

Option	Description
Labels	Selected by default. Shows the vessel locations (for example, A1 for a well in a 96-well microplate) while panning and zooming the Vessel View display.
Image Metrics	Shows a snapshot of the image data for the vessel at the selected scan time. The value for the selected metric is displayed on each image. The data is dynamically updated as you select a different time in the Vessel Scanning Timeline. For a vessel that has not been analyzed, only Scan metrics are available. For an analyzed vessel, analysis-specific metrics are also available. Use the Transparency slider bar to adjust the opacity of the selected metric. **Note:** The RMS Row Shift and RMS Column Shift scan metrics are primarily for diagnostic use for the microscope functionality. Unlike the other scan metrics (Focus Position, Phase Exposure Time, and so on), they have no "biology-related" use. **Tip:** To change the color of the metric that is displayed, click the solid white circle that is displayed to the right of the Transparency slider bar to open the standard Windows Color dialog box, and then select a basic color, or define a custom color for the metric.
Packed View	Default. Removes the dead space between vessel locations to optimize the viewing area. **Note:** Sartorius recommends that you leave the Packed View option selected for the Vessel View window.

You can also right-click any vessel location in the Vessel View display to open a context menu with the following options:

Option	Description
Copy Viewing Area to Clipboard	Copies the *current* Vessel View display to your client's clipboard. You can then use the standard Paste commands to paste the copied display in to a third-party application such as Microsoft PowerPoint.

Option	Description
Export Image via Designer	See “To export images as displayed” in “Exporting Images” on page 100.
Export Image as Stored	See “To export images as stored” in “Exporting Images” on page 100.

The Navigation toolbar

The Navigation toolbar contains options for navigating the Vessel View display, including zooming in, zooming out, and moving through the scans for the vessel. When the Vessel View window first opens, the zoom level of the display is set to a starting, or *Home*, value of 100%, which means that the entire vessel is shown in the Vessel View display.

Icon	Description
	Zoom In icon: Click as needed to zoom in on the Vessel View display. **Tip:** To zoom in to the maximum extent possible on a vessel location, you can double-click the location.
	Zoom Out icon: Available only after at zooming in on the Vessel View display. Click as needed to zoom out the Vessel View display.
Tip: You can also use the scroll button on your mouse to zoom in on and zoom out the Vessel View display and you can also click and hold the mouse pointer anywhere on the Vessel View display, and then drag the display to move it to a different location.	
	Home icon: Available only after zooming in on or zooming out the Vessel View display. Click once to return the Vessel View display to its starting, or Home, value of 100%. (The entire vessel is shown in the Vessel View display.)
	Next Scan icon: Updates the Vessel View display to the next scan in the Vessel Scanning Timeline. Click as needed to move forward through the scans for the vessel.
	Previous Scan icon: Updates the Vessel View display to the previous scan in the Vessel Scanning Timeline. Click as needed to move backward through the scans for the vessel.
	Start/Stop Slide Show icon: Creates a self-running presentation of all the scans for the vessel. As the presentation runs, the corresponding date/time stamp for each scan is temporarily shown at the top of the Vessel View display. To stop the slide show at any time, click the Stop Slide Show icon.

Exporting Images

After you have acquired images in a vessel, you can export the images from one or more image sites at a time. You use the Image Export wizard to export the image. Two choices are available for exporting an image:

- As Displayed: Exports the data as it is currently displayed in the Vessel View window.You can layer image channels and masks to customize the image view, you can crop and scale the image, and you can apply an overlay. You typically select As Displayed if you are intending to use the image in reports or presentations, and/or sharing the image data, such as in an email. When you export the image as displayed, you can export the image as a series of individual files, or you can collate the images and export them as a single movie. See “To export images as displayed” below.
- As Stored: Exports the image and mask data as stored on the Incucyte system *prior* to displaying in the Vessel View window. This is typically referred to as the “raw” data. User-specified settings are not reflected in this type of export and the image components are exported to individual files. You typically select As Stored when you are importing the image in to third-party analysis tools or you are sending the image as part of an Incucyte support request. See “To export images as stored” on page 106.

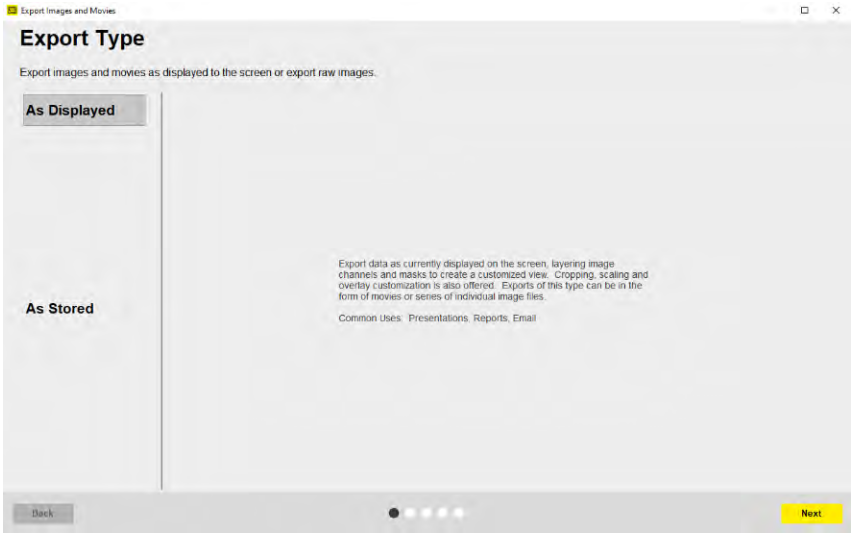
The Launch Analysis wizard uses the current display (zoom level, focus, and scan time) of the vessel in the Vessel View window. Before you open the wizard, optionally, you can set the zoom level of the Vessel View window to its Home value (100%) and you can confirm that the preferred scan time is selected on the Vessel Scanning timeline.

To export images as displayed

1. Open the vessel in the Vessel View window. See “To open the Vessel View window” on page 85.
2. On the Visualization toolbar, click the Export Images and Movie icon.

   The Image Export wizard opens. The Export Type page is the open page. As Displayed is selected.

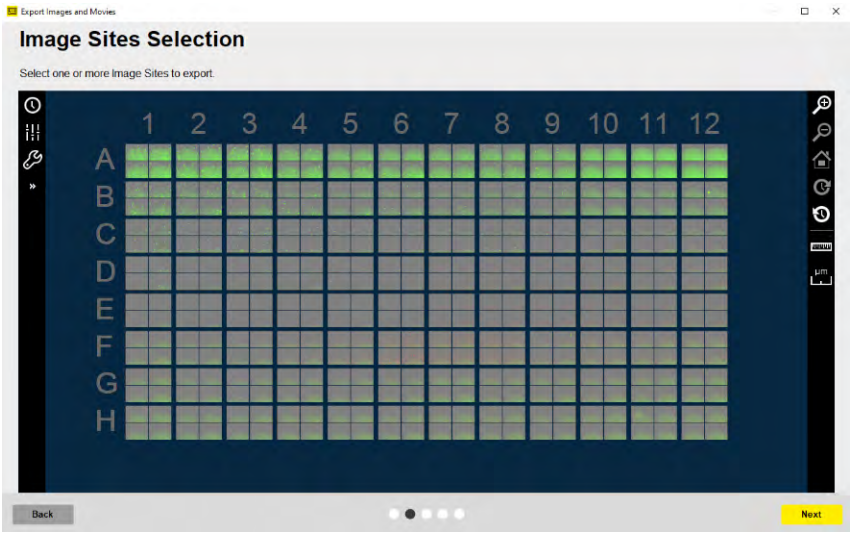
Figure 2-11: Image Export wizard, Export Type page



3. Leave As Displayed selected, and then click Next.

   The Image Site Selection page opens. The page shows the vessel as it was displayed (zoom level, focus, and scan time) in the Vessel View window at the time that you opened the wizard.

Figure 2-12: Image Export wizard, Image Site Selection page



All the options that are available on the Image Site Selection selection page are identical to that on the Vessel View window. With the exception of the spectral unmixing option, you can use all the options to help you select the images that you are exporting. For example, if you did not have the correct scan time selected in the Vessel View window before you opened the Image Export wizard, then you can open the Vessel Scanning timeline for the selected vessel and scroll through and select a different scan time, you can use the options on the Navigation toolbar to zoom in on the selected vessel location to confirm that it shows the biology of interest, and so on. See "The Vessel View Window" on page 89.

4. For each image site that you are exporting, click the site. (To clear an image selection, click it again.)

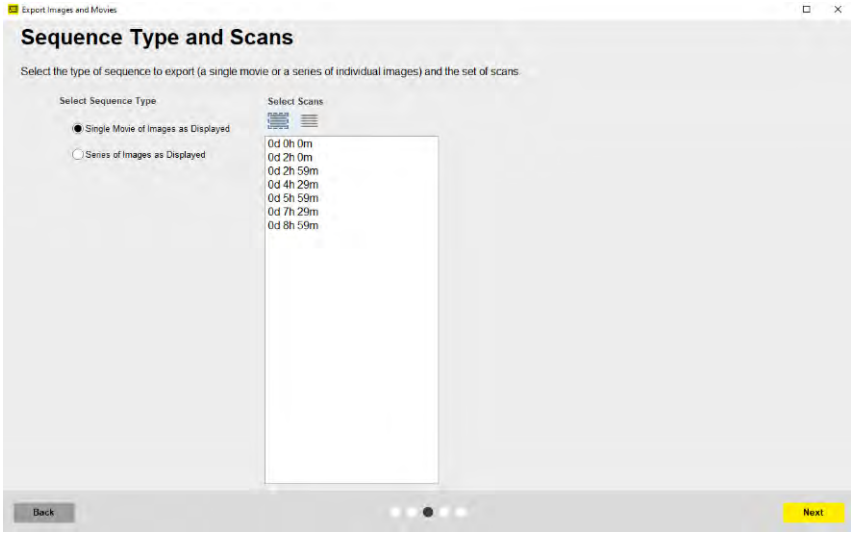
   As you hold the mouse pointer over a site, the pointer changes to a blue plus sign. After you click a site to select it, the pointer changes to a red X and the selected site/well is filled with a solid light blue color.

If multiple images have been acquired from a site or well, then you must select each image that you are exporting.

5. Click Next.

   The Sequence Type and Scans page opens. You use the options on this page to select the sequence type that you are exporting (a single movie or a series of individual images) and the specific scans for which you are exporting the images. The page displays the Vessel Scanning Timeline for the vessel. By default, single movie is selected. See Figure 2-13 on page 102.

Figure 2-13: Image Export wizard, Sequence Type and Scans page



6. To export the images as a movie, leave Single Movie of Images as Displayed selected; otherwise, to export the images as a series of individual files, select Series of Images as Displayed.

Although the option is "Series of Images," you can export a single image scan type.

7. Select the scans for the images that you are exporting. You can do any of the following:
   - To select all scans in a single step, under Select Scans, click the Select All Scans icon . To clear an individual scan, click the scan again.
   - In the Vessel Scanning timeline, do either one or both of the following:
     - To select an individual scan, click the scan. You can select multiple individual scans. To clear an individual scan, click it again.
     - To select a range of scans, click the first scan in the series of scans, press and hold the Shift key, and then click the last scan in the series of scans.
   - To clear all selected scans in a single step, under Select Scans, click the Clear All Scans icon .
8. Click Next.

   The Export Designer page opens. You use the options on this page to customize the movie/image files before exporting, including cropping the image, adding a legend, and so on. When the page first opens, default values are set for many of the options. For either export sequence, the Final Size is the final export size in pixels. If you selected multiple images with multiple scan times, then the first image selected at the first scan time selected is displayed. See Figure 2-14 on page 103.

If you selected Single Movie of Images as Displayed, then depending on the image size, a series of messages about Automatic Scaling and/or Export Size might open before the Export Design page, indicating that to automatically reduce the size of the exported movie, automatic scaling has been applied. The messages also prompt you to consider additional scaling and/or cropping to further reduce the size. Click OK to close these messages and continue to the Export Designer page.

Figure 2-14: Image Export wizard, Export Designer page

9. Optionally, select a different image and/or scan time to display.
10. Modify any of the design options as needed, including cropping and scaling the image. Note the following about the Export Design page:
    - After you make any edits, the display on the Export Designer page is dynamically updated.
    - You can click the Full Screen Preview icon to view a full, onscreen preview of the movie or image file that you are exporting at any time. To reset all values to their default values in a single step, click the Reset icon .
    - The image quality is set to approximately the highest value. Although not recommended, you can use the Quality slider bar to reduce the image quality, which also reduces the final file size.

Although not common, you might reduce the quality, for example, in the event that you simply want to share the perspective of what is occurring in a selected image site with a colleague and you want to reduce the file size to make it easier to share via email.

- If you are exporting a single movie, then you can increase the Frames per second value to play the movie at a faster rate. After you adjust the value, you can hold your mouse pointer over the Example Movie display to preview a *stock movie* that is played at the specified rate.

The preview is that of a stock movie display. It is not built from your selected image site.

11. Click Next.

    The File Information page opens. You specify the export settings such as the target folder on this page. The options on this page are different for a single movie (Figure 2-15) and individual image files (Figure 2-16).

Figure 2-15: Image Export wizard, File Information page (Single movie)

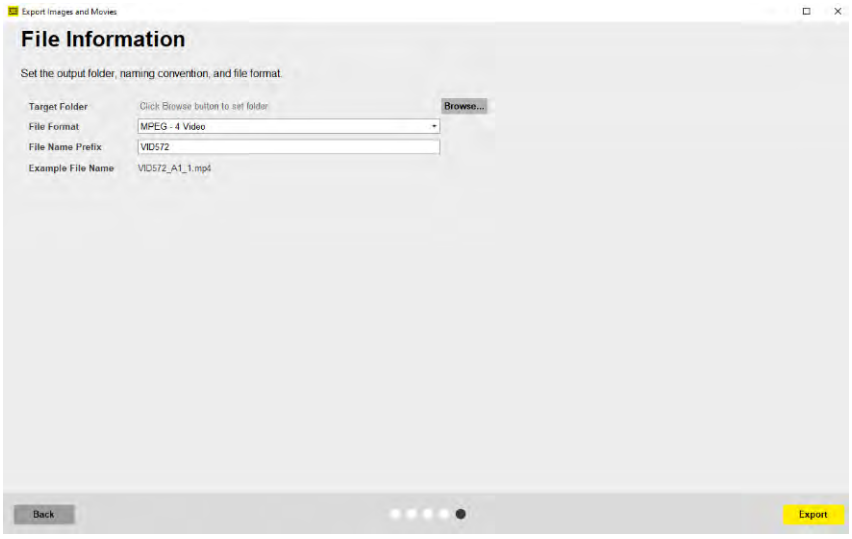
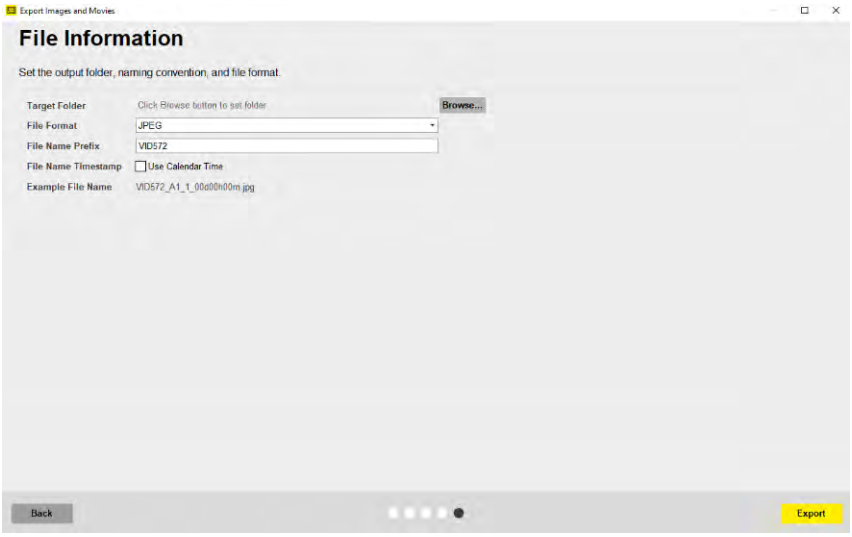


Figure 2-16: Image Export wizard, File Information page (Individual image files)



12. Specify the file information.

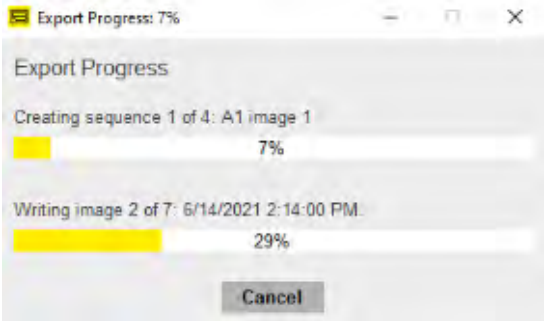
Option	Description
Target Folder	The folder in which the exported images are stored. Click Browse to open the Select a folder to export to dialog box, and then browse to and select the appropriate folder.
File Format	The default value for a single movie is MPEG-4 Video and for individual files is JPEG.

Option	Description
File Name Prefix	The exported file is automatically named according to the following: • For a single movie: <FileName Prefix>_<Site/Well ID>_<Image Number>_<File Format>, for example, VID2197_D7_1_.mp4. • For a series of individual images: <FileName Prefix>_<Site/Well ID>_<Image Number><Date/Timestamp><File Format>, where the File Name Timestamp option determines the timestamp. See "File Name Timestamp" below. **Note:** The default value for the file name prefix is the Vessel ID for the selected vessel. You can leave this value as-is, or you can edit it as necessary.
File Name Timestamp	Applicable only for individual image files. • Select Use Calendar Time to append the file name with the date and timestamp that the selected image was acquired, for example, VID2197_D7_1_2016y05m11d_21h36m.jpg • Clear Use Calendar to append the filename with the relative date and timestamp that the selected image was acquired, for example, VID2197_D7_1_.00d00h00m.jpg

13. Click Export.

    The Image Export wizard closes. You return to the Vessel View window. An Export Progress dialog box opens on top of the Vessel View window.

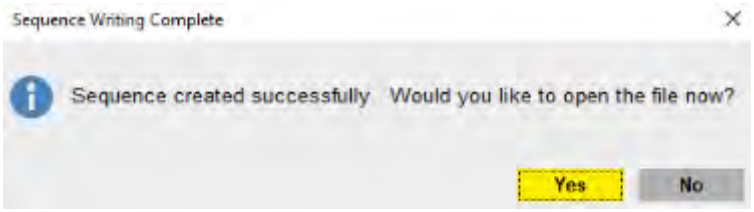
Figure 2-17: Export Progress dialog box



After the export progress is 100%, a Sequence Writing Complete dialog box opens.

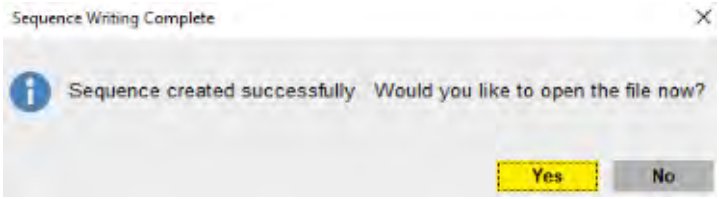
- If you exported a single movie, then the dialog box indicates that the sequence was successfully created, and asks you if you want to open the file.

Figure 2-18: Sequence Writing Complete dialog box



- If you exported individual image files, then the dialog box indicates the number of files that were created successfully and asks you if you would like to open the target folder. See Figure 2-19 on page 106.

Figure 2-19: Sequence Writing Complete dialog box



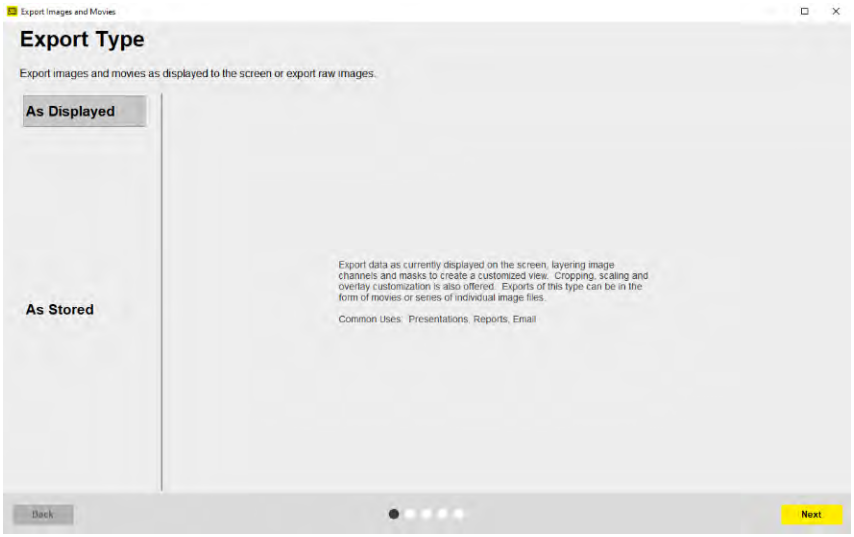
14. Do one of the following:
    - To close the wizard, return to the Vessel View window, and open the file or target folder, click Yes.
    - To close the wizard and return to the Vessel View window, click No.

To export images as stored

1. Open the vessel in the Vessel View window. See “To open the Vessel View window” on page 85.
2. On the Visualization toolbar, click the Export Images and Movie icon.

   The Image Export wizard opens. The Export Type page is the open page. As Displayed is selected.

Figure 2-20: Image Export wizard, Export Type page



3. Select As Stored, and then click Next.

   The Image Type Selection page opens. You specify the image type that are exporting on this page. See Figure 2-21 on page 107.

Figure 2-21: Image Export wizard, Image Type page

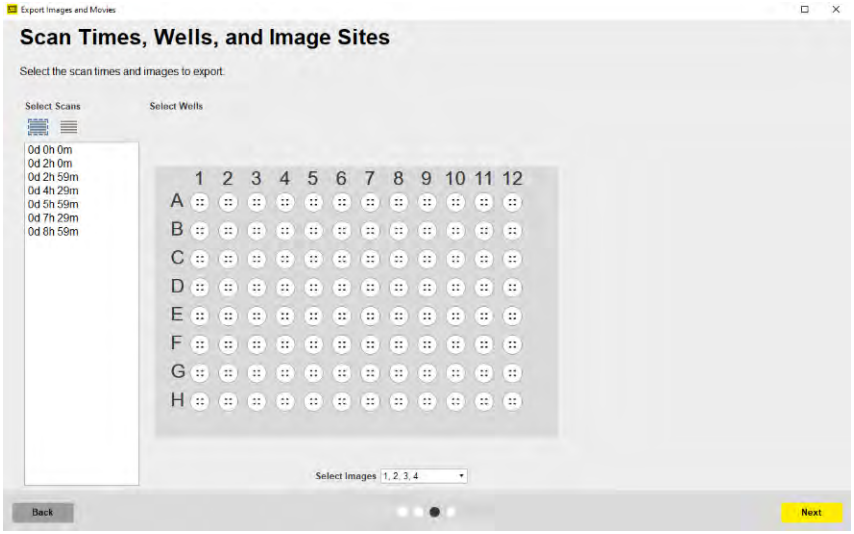
4. Select the image type that you are exporting.

Image Type	Description
Phase original	Raw 8-bit image
Green uncalibrated	Raw 16-bit image without calibration
Green calibrated	32-bit floating point image in calibrated units
Red uncalibrated	Raw 16-bit image without calibration
Red calibrated	32-bit floating point image in calibrated units

5. Click Next.

   The Scan Times, Wells, and Image Sites page opens. The page displays the Vessel Scanning Timeline for the vessel and the vessel acquisition map. You use the options on this page to select the vessel locations, the number of images, and the scans for which you are exporting the raw image data. See Figure 2-22 on page 108.

Figure 2-22: Image Export wizard, Scan Times, Wells, and Image Sites page

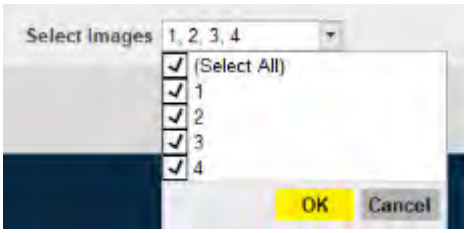


6. Do the following as indicated to select the scan times, image sites, and number of images per site for which you are exporting the raw data:

Option	Description

To select scan times	• To select all scans in a single step, under Select Scans, click the Select All Scans icon . To clear an individual scan, click it.
	• In the Vessel Scanning Timeline, do either one or both of the following:
	  • To select an individual scan, click the scan. You can select multiple, individual scans. To clear an individual scan, click it again.
	  • To select a range of scans, click the first scan in the series of scans, press and hold the Shift key, and then click the last scan in the series of scans.
	• To clear all selected scans in a single step, under Select Scans, click the Clear All Scans icon .

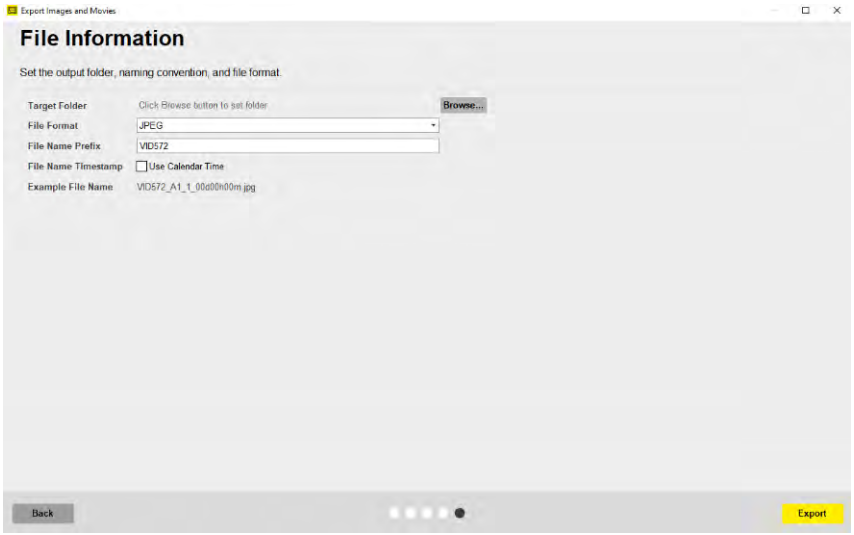
To select wells/ image sites	• To select a single location in a vessel, for example, a well in a 96-well microplate, click once in the appropriate location in the vessel map. • To select multiple contiguous locations in a vessel, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them. • To select all the locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Select this Column. Conversely, to clear all the selected locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Deselect this Column. • To select all the locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Select this Row. Conversely, to clear all the selected locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Deselect this Row. **Tip:** To clear all the selected locations in the vessel map, or multiple contiguous locations, you can also press and hold the ALT key, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

Option	Description
To select which images to export	Applicable only if more than a single image has been acquired per vessel location. By default, all images are selected for export. To select different images to export, open the Select Images dropdown list, clear the selections for the images that you are *not* exporting, and then click OK. *Figure 2-23: Select Images list (4 images per vessel location)*  **Note:** Remember, if multiple images were acquired for image sites, then the system considers the images ordered in rows from top to bottom, and then, within the rows, from left to right. See Figure 2-13 on page 49.

7. Click Next.

   The File Information page opens. You specify the export settings such as the target folder on this page.

Figure 2-24: Image Export wizard, File Information page



8. Specify the file information.

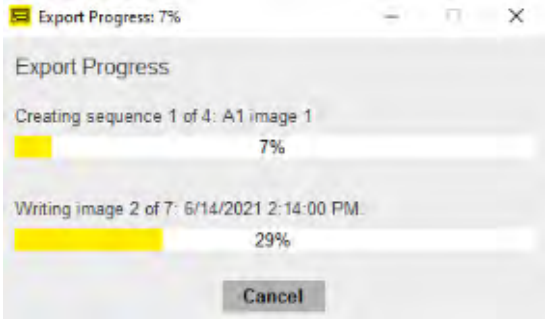
Option	Description
Target Folder	The folder in which the exported images are stored. Click Browse to open the Select a folder to export to dialog box, and then browse to and select the appropriate folder.
File Format	The default value is dependent on the image type that is selected for export.

Option	Description
File Name Prefix	The exported file is automatically named according to the following: <FileName Prefix>_<Site/Well ID>_<Image Number><Date/Timestamp><File Format>, where the File Name Timestamp option determines the timestamp. See "File Name Timestamp" below. **Note:** The default value for the file name prefix is the Vessel ID for the selected vessel. You can leave this value as-is, or you can edit it as necessary.
File Name Timestamp	• To append the file name with the date and timestamp that the selected image was acquired, select Use Calendar Time. For example: VID2197_D7_1_2016y05m11d_21h36m.tif • To append the filename with the relative date and timestamp that the selected image was acquired, which is always 00d00h00m, clear Use Calendar Time. For example: VID2197_D7_1_.00d00h00m.tif

9. Click Export.

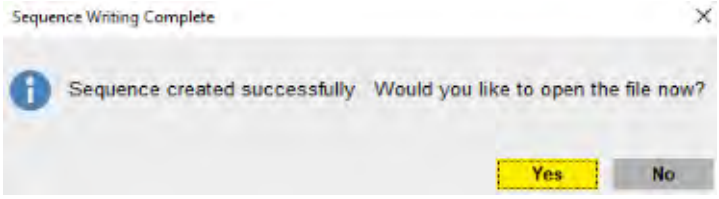
   The Image Export wizard closes. You return to the Vessel View window. An Export Progress dialog box opens on top of the Vessel View window.

Figure 2-25: Export Progress dialog box



After the export progress is 100%, a Sequence Writing Complete dialog box opens. The dialog box indicates the number of files that were created successfully and asks you if you would like to open the target folder.

Figure 2-26: Sequence Writing Complete dialog box



10. Do one of the following:
    - To close the wizard and return to the Vessel View page, click No.
    - To close the wizard and return to the Vessel View page and open the target folder, click Yes.

Section 3
Analyzing Images and Visualizing Results

Section Contents

Chapter 1
Defining the Image Analysis

After viewing and accessing your vessel images, your next step is to select the images that are representative of the biology that is occurring in your vessel and define the analysis parameters. The parameters that you define based on these representative images are then applied to analyze all images within in the vessel. This chapter details the Launch Analysis wizard, which you use to define the values for your image analysis settings (parameters), and then launch the analysis of the images.

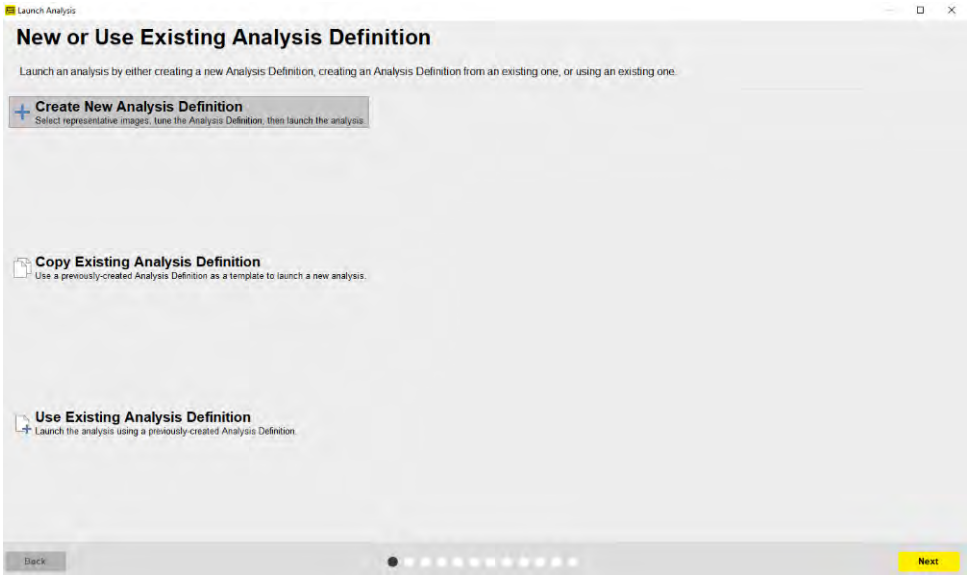
This chapter covers the following topics:

After you have defined an image analysis, you manage it on the Analysis Definitions tab on the Manage window, which includes editing it, renaming it, deleting it, and so on. For detailed information about managing an analysis definition, see Appendix A, “Incucyte Management,” on page 181.

Launch Analysis Wizard Overview

You use the Launch Analysis wizard to analyze vessel images.

Figure 1-1: Launch Analysis wizard



Three options are available in the wizard for analyzing vessel images: You can create a new analysis definition for analyzing vessel images, you can copy and edit an existing analysis definition, or you can use an existing analysis definition as-is.

- To create a new analysis definition (the default value), see“Setting up a New Analysis” on page 116.
- To copy and modify an existing analysis definition. See “Copying an Existing Analysis Definition” on page 130.
- To use an existing analysis definition, see “Using an Existing Analysis Definition” on page 132.

The following sections specifically detail how to create an analysis definition. After analysis definitions are created, they must be managed. Managing existing analysis definitions consists of viewing and sorting definitions, importing existing definitions, deleting definitions, and so on. You manage analysis definitions on the Analysis Definitions tab on the Manage window. See “Managing Analysis Definitions” on page 197.

Setting up a New Analysis

The following procedure provides high-level information for setting up a completely new analysis for images. For setting up a new analysis by copying and then editing an existing analysis definition, see "Copying an Existing Analysis Definition" on page 130.

Setting up a new analysis consists of the following:

- Indicating whether to create a new analysis definition, or use an existing definition. See "To create a new analysis definition" on page 116.
- Selecting the analysis type, which is dependent on the scan type. See "To select the Analysis Type" on page 118.
- Selecting the image channels that you are analyzing. See "To select the image channels for analysis" on page 119.
- Selecting the images that best represent the biology of interest that is occurring in the vessel. See "To select the images for analysis" on page 120.
- Refining the analysis settings for each of the image channels as appropriate. See "To define the analysis settings (parameters)" on page 121.
- Selecting the scan times and images that you are analyzing. See "To select the scan times and wells/sectors/image sites" on page 123.
- Save and apply the analysis definition. See "To save and apply the analysis definition" on page 125.
- Confirming the analysis definition, and then launching the analysis. See "To review the analysis summary information" on page 127.

For the following procedures, the vessel is assumed to be a 96-well microplate and the analysis type is Basic Analyzer. The majority of information, however, is "vessel-agnostic" and offers a frame of reference for setting up a new analysis for any scan type with any analysis type.

The Launch Analysis wizard uses the current display (zoom level, focus, and scan time) of the vessel in the Vessel View window. Before you open the wizard, optionally, you can set the zoom level of the Vessel View window to its Home value (100%) and you can confirm that the preferred scan time is selected on the Vessel Scanning Timeline.

To create a new analysis definition

1. Open the vessel in the Vessel View window. See "To open the Vessel View window" on page 85.
2. On the Analysis toolbar, click the Launch Analysis icon.

   The Launch Analysis wizard opens. The New or Use Existing Analysis Definition page is open. Create New Analysis Definition is selected. See Figure 1-3 on page 117.

If the acquired images were multi-channel, and you have not specified any spectral unmixing values, then the No Spectral Unmixing dialog box opens (see Figure 1-2 below), warning you that you are about to launch an analysis with no spectral unmixing defined and asking you if you want to continue. Click Yes or No as appropriate. If you click No, then dialog box closes and the Vessel View window remains open. For information about spectral unmixing, see "Spectral Unmixing" on page 97.

Figure 1-2: No Spectral Unmixing dialog box

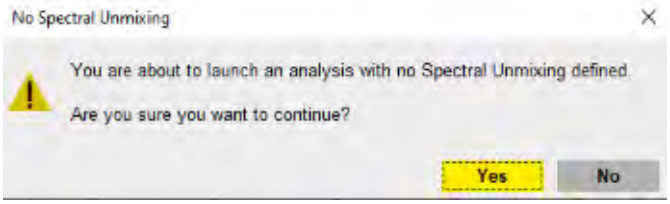
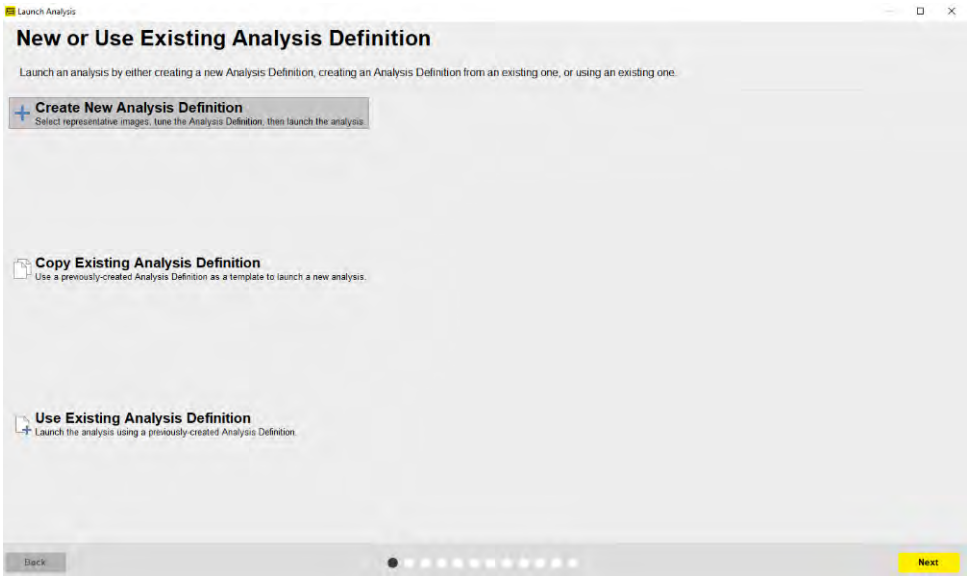


Figure 1-3: Launch Analysis wizard, New or Use Existing Analysis Definition page



3. Leave Create New Analysis Definition selected, and then click Next.

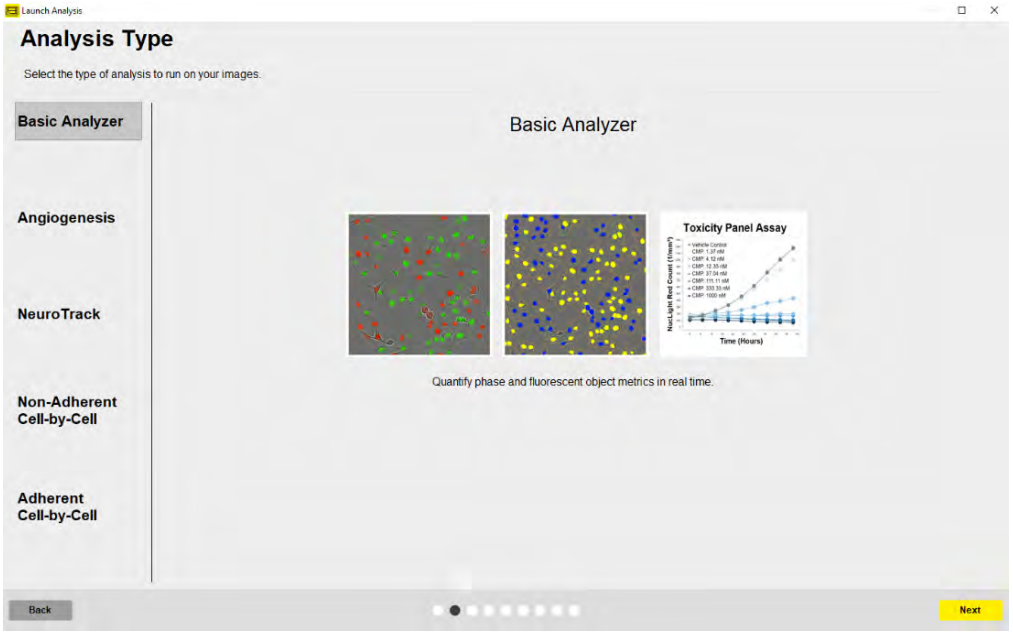
   The Analysis Type page opens.

4. Continue to "To select the Analysis Type" on page 118.

To select the Analysis Type

The Analysis Type page displays the analysis types that are available are based on the scan type and the image channels that were collected during scanning. Basic Analyzer is always available.

Figure 1-4: Launch Analysis wizard, Analysis Type page



1. Select the analysis type (In this example, Basic Analyzer is selected).
2. Click Next.

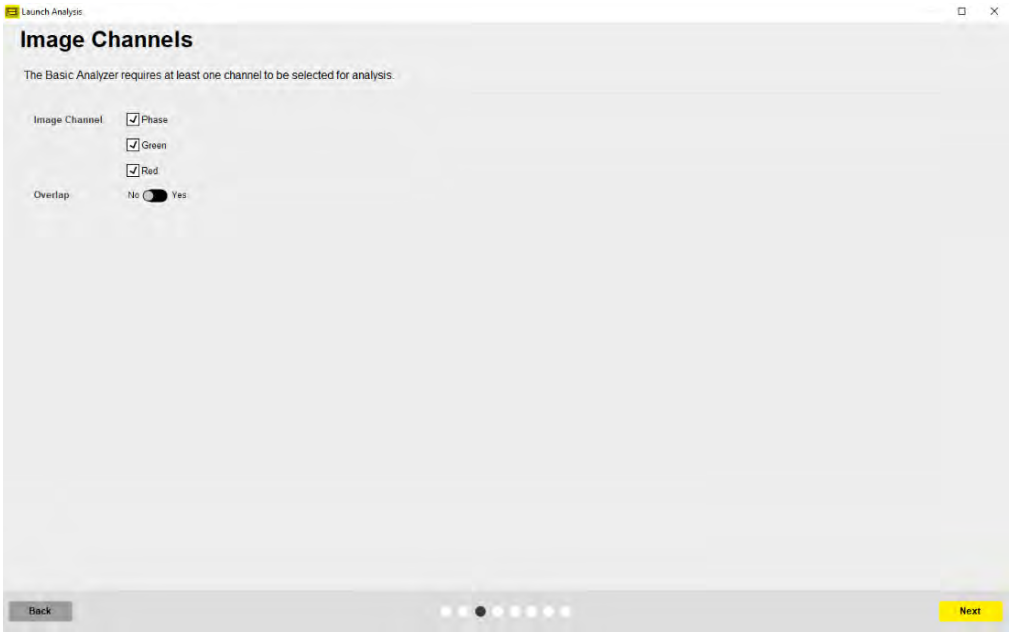
   The Image Channels page opens.
3. Continue to “To select the image channels for analysis” on page 119.

To select the image channels for analysis

The Image Channels page displays all the possible image channels (Phase, Red, and Green) for the analysis. By default, all the channels for which images were acquired are selected.

Overlap is applicable for co-incident object analysis (red and green objects overlap) and is not discussed here.

Figure 1-5: Launch Analysis wizard, Image Channels page



1. Select the channels for analysis. When selecting the channels, note the following:
   - To limit your analysis to a green object and/or red object count, leave Green and/or Red selected as appropriate, and clear Phase.
   - To include the Phase metric in your analysis, leave Phase selected.

In this example, all three channels are selected.

2. Click Next.

   The Image Set Selection page opens.

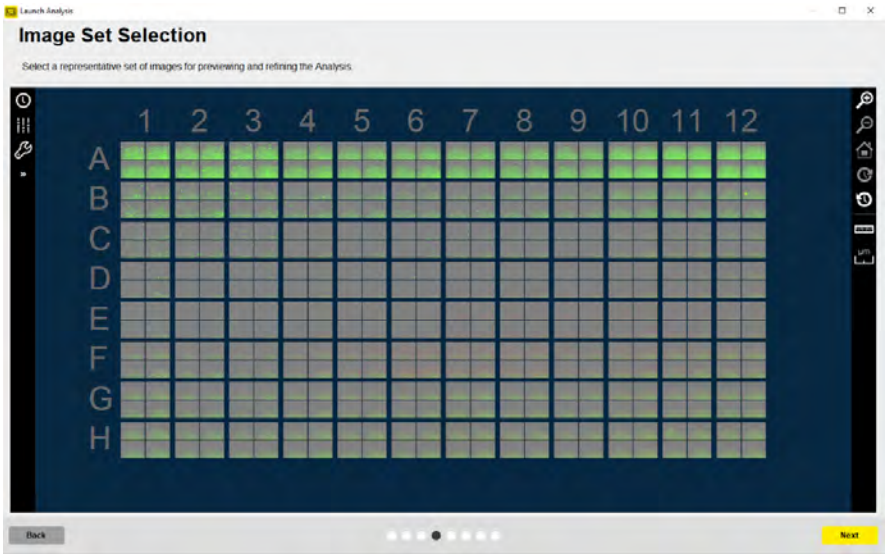
3. Continue to "To select the images for analysis" on page 120.

To select the images for analysis

You use the options on the Image Set Selection page to select the images that best represent the biology of interest that is occurring in the vessel so that you can preview and refine the analysis before applying it to all the images that you are analyzing. When selecting these representative images, note the following:

- All the options that are available on the Image Set Selection selection page are identical to that on the Vessel View page. With the exception of the spectral unmixing option, you can use all the options to help you select the images that you are analyzing. For example, if you did not have the correct scan time selected in the Vessel View window before you opened the Launch Analysis wizard, then you can open the Vessel Scanning Timeline for the selected vessel and scroll through and select a different scan time, you can use the options on the Navigation toolbar to zoom in on the selected vessel location to confirm that it shows the biology of interest, and so on. See “The Vessel View Window” on page 89.
- Typically, two to four images are adequate for defining the analysis; however, if the biology is significantly different across samples, then you can select more images. Ideally, regardless of the number of images, one of these images should be a control image.
- The images need not have been acquired at the same scan time. You can select images that have been acquired at different scan times. Select the appropriate scan time on the Vessel Scanning Timeline popup to update the vessel display with the images that were acquired at the selected time.

Figure 1-6: Launch Analysis wizard, Image Set Selection page



1. To select an image site for analysis, click the site.

   As you hold your mouse pointer over a site, the mouse pointer changes to a blue plus sign. After you click a site to select it, the pointer changes to a red X and the selected site is filled with a solid light blue color. A message is also briefly displayed at the top of the page that indicates which images have been added to the analysis, for example, “Images from A4-2 added.”

To clear an image selection, click it again.

2. Click Next.

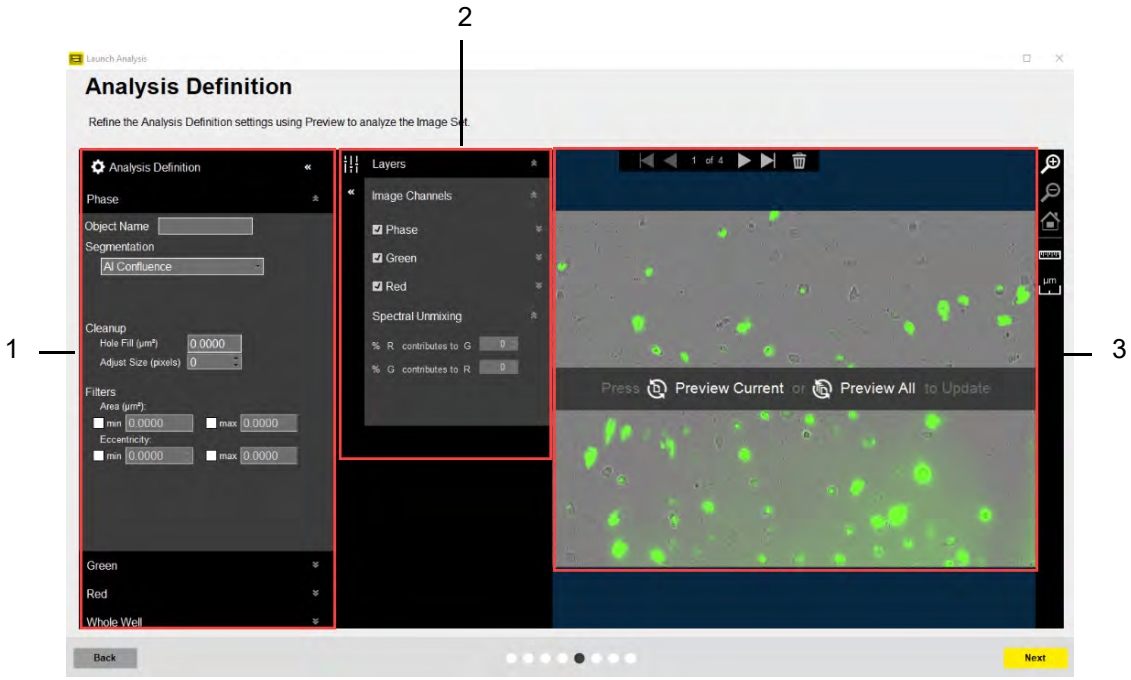
   The Analysis Definition page opens.

3. Continue to "To define the analysis settings (parameters)" below.

To define the analysis settings (parameters)

You use the options on the Analysis Definition page to initially define the analysis settings (parameters) based on the images that you just selected, and then iteratively, preview and refine the analysis settings until you are satisfied that the definition can be applied to all the images that you select for analysis.

Figure 1-7: Launch Analysis wizard, Analysis Definition page



The Analysis Definition page has the following layout:

	Window component	Description
1	Analysis Definition pane	Displays various settings that are specific to the analysis type that you used to set up the analysis. Whole Well is always displayed but it is applicable only for whole well imaging, dilution cloning, and chemotaxis. **Tip:** When the Analysis Definition page first opens, options that are expanded are indicated either by a double-headed arrow that points to the left, or a double-headed arrow that points up. Options that are collapsed are indicated either by a double-headed arrow that points right or a double-headed arrow that points down. You can click these expand/collapse icons as needed to optimize the page display for your work.

	Window component	Description
2	Layers pane	Displays the Image Channel layers that will be used to test the analysis definition. For example, in Figure 1-7 on page 121, three image channels (Phase, Green, and Red) were selected for analysis. You can turn on or off a layer to help define the analysis settings. **Note:** The spectral unmixing values that are displayed here are read-only. You cannot adjust spectral unmixing values in the Launch Analysis wizard. **Note:** Changes made in the image channel, for example, autoscale or contrast, aid in evaluating the analysis mask. Changing these values does not affect the analysis definition.
3	Image pane	Displays the images that were selected for creating the analysis definition. A navigation bar at the top of the pane indicates 1 out of <n> total images. For example, in Figure 1-7 on page 121, four total images were selected for the analysis definition, so the navigation bar shows 1 of 4. You use the Preview options that are displayed in the Image pane to preview the image that is currently selected (as shown in the Navigation bar), and to Preview All selected images, which updates the analysis parameters and filters across all the image channels for all the images that were selected for testing the analysis definition. **Tip:** At any time, you can hold your mouse pointer over any object in an image to open a tooltip that displays information about the object.

When refining the analysis parameters for an image channel:

- You should define the analysis settings for the image channels in the following order: Phase, Green, and Red.
- Prior to changing any settings, you use the default values that are provided for the analysis definition.
- Ideally, you should define, and then refine only a single analysis setting at a time.

1. Leave the first image that you selected for defining the analysis settings displayed in the Image pane, or use the Navigation toolbar at the top of the pane to select a different image.
2. Refine the analysis definition settings for each of the image channels as appropriate based on your selected analysis type.
3. Click Preview All.

   The analysis definition settings are applied to all the images that you selected.
4. Use the Navigation toolbar at the top of the Image pane to move to each image and then toggle your masks on and off to confirm that the analysis settings that you have defined are correctly mapping your objects of interest in each image. Make adjustments where necessary. If you make any adjustments, then you can preview a single image or preview all images as appropriate. When previewing your images, note the following:
   - If you decide not to include the image during the analysis definition, you can click the Remove Image icon  at the top of the Image pane to remove the image.
   - If you decide that you must include more or different images during the analysis definition, click Back once to return to the Image Set Selection page, select the additional image or images, and then click Next to return to the Analysis Definition page.

5. Click Next.

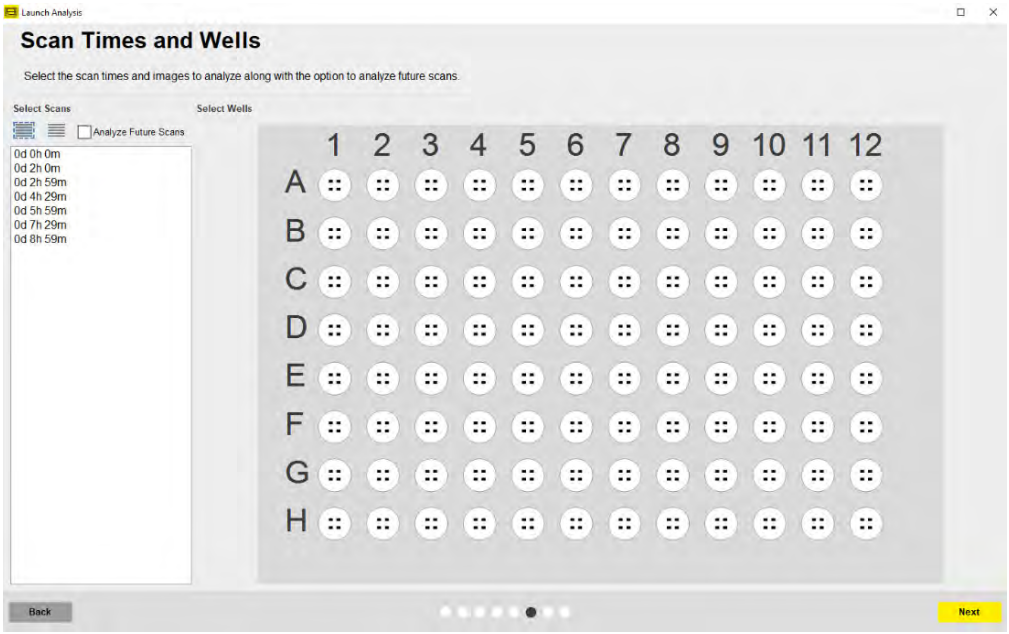
Next is available only after you have previewed all your selected images.

The Scan Times and Wells page opens. Continue to "To select the scan times and wells/sectors/image sites" below.

To select the scan times and wells/sectors/image sites

You use the options on the Scan Times and Wells page to select the scan times and images that you are analyzing. You also indicate whether to analyze futures scans for the vessel according to the currently defined parameters.

Figure 1-8: Scan Times and Wells page



1. Select the scan times and image sites for which you are analyzing the data.

Option	Description

To select scan times	• To select all scans in a single step, under Select Scans, click the Select All Scans icon . To clear an individual scan, click it.
	• In the Vessel Scanning Timeline, do either one or both of the following:
	  • To select an individual scan, click the scan. You can select multiple, individual scans. To clear an individual scan, click it again.
	  • To select a range of scans, click the first scan in the series of scans, press and hold the Shift key, and then click the last scan in the series of scans.
	• To clear all selected scans in a single step, under Select Scans, click the Clear All Scans icon .

Option	Description
To select wells/sectors/ image sites	• To select a single location in a vessel, for example, a well in a 96-well microplate, click once in the appropriate location in the vessel map. • To select multiple contiguous locations in a vessel, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them. • To select all the locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Select this Column. Conversely, to clear all the selected locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Deselect this Column. • To select all the locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Select this Row. Conversely, to clear all the selected locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Deselect this Row. **Tip:** To clear all the selected locations in the vessel map, or multiple contiguous locations, you can also press and hold the ALT key, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

2. Optionally, to run the defined analysis on all future scans of this vessel (the images are analyzed in real time), select Analyze Future Scans.

If you select Analyze Future Scans, then the vessel can remain in the Incucyte instrument. If the vessel has been removed, then you can use the Restore function to scan it at a later time.

3. Click Next.

   The Save and Apply Analysis Definition page opens.

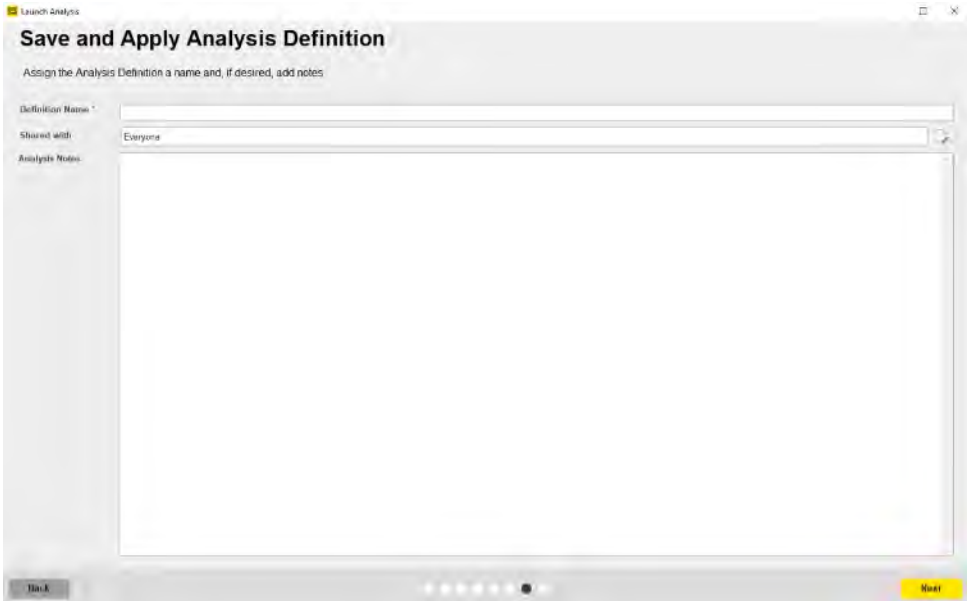
4. Continue to "To save and apply the analysis definition" on page 125.

To save and apply the analysis definition

If you are following this procedure when working with an existing analysis definition, then the Analysis Notes page replaces the Save and Apply Analysis Definition page. The Definition Name is read-only. You can enter only analysis notes on this page.

By default, a new analysis definition is shared with the **Everyone** user workgroup.

Figure 1-9: Launch Analysis wizard, Save and Apply Analysis Definition page



1. In the Definition Name field, enter the name for the analysis definition.

Enter a name that is descriptive for the experiment/assay that you are carrying out, for example, HT-1080 Apoptosis The name does not need to include any user identification (the name of the user who is defining the assay). The user name is automatically associated with the assay and is displayed in the Analysis tooltip in the Vessel View window. See Figure 1-4 on page 83.

2. The Shared with field displays the user workgroups with which the analysis definition is currently shared. Optionally, select one or more different user workgroups with which to share the definition. See “To share an analysis definition with a user workgroup” on page 126.
3. Optionally, in the Analysis Notes field, enter pertinent notes about the analysis.
4. Click Next.

   The Summary page opens.
5. Continue to “To review the analysis summary information” on page 127.

To share an analysis definition with a user workgroup

Before you can share an analysis definition with a user workgroup, the User Workgroups setting must be turned on for the instrument. See "Specifying User Settings for an Incucyte" on page 248.

A *user workgroup* defines the users who are able to see and work with an analysis definition. Sharing an analysis definition with a user workgroup is alway optional. To limit the users who can view and work with an analysis definition, you must share the definition with one or more specific workgroups.*

1. At the far right of the Shared with field, click the Edit icon .

   The Edit Shared Workgroups dialog box opens. The dialog box lists all the user workgroups with which the definition is currently shared and the workgroups with which you can still share the definition.

**Before you share an analysis definition with a workgroup, you can view the users that are currently assigned to the workgroup. Click the Show Users icon to update the dialog box display with a list of current users for the selected workgroup.*

Figure 1-10: Edit Shared Workgroups dialog box

2. Do one or both of the following:

   - To share an analysis definition with a user workgroup, click Share for the workgroup.

If the appropriate user workgroup is not available, contact your Incucyte administrator.

   - To remove a user workgroup from the vessel, clear the Share selection for the workgroup.

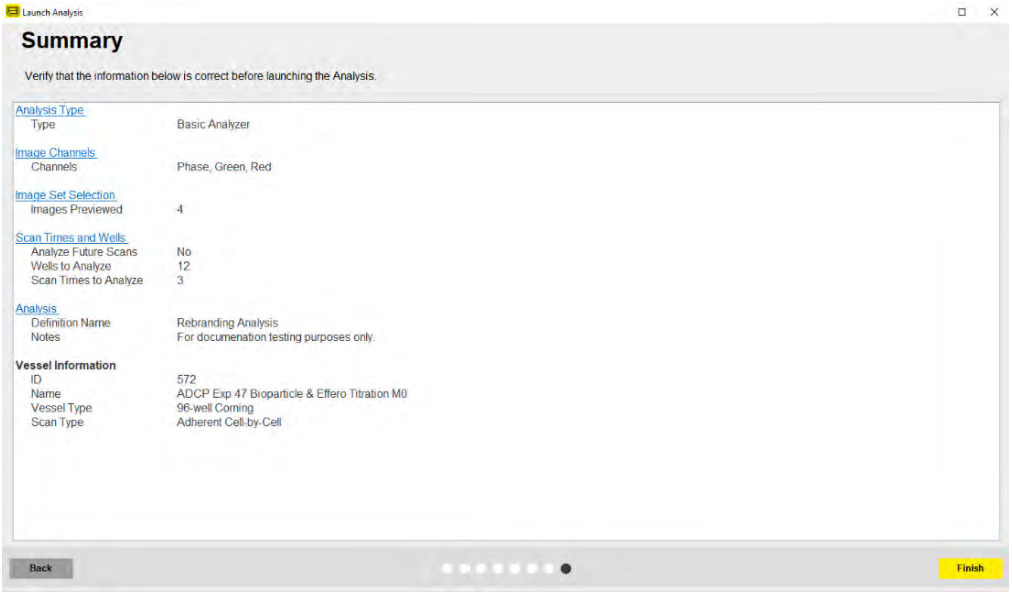
*If you clear all workgroup assignments for a definition, then, by default, the assignment is automatically reset to **Everyone**.*

3. Click Apply.

   The Edit Shared Workgroups dialog box closes and you return to the Vessel Notebook page. The selected user workgroup is displayed in the Shared with field.

To review the analysis summary information

Figure 1-11: Launch Analysis wizard, Summary page



The Summary page displays all the selections/settings that you have specified for the image analysis, organized by wizard page. Each section heading corresponds to a wizard page, and is a hyperlink to the indicated page.

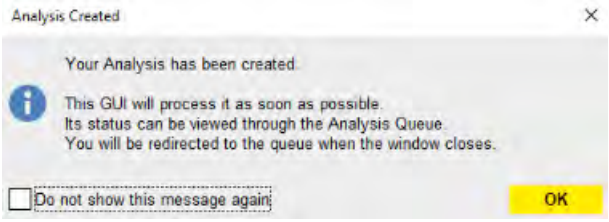
1. Review the information for the planned analysis, and then do one of the following:
   - If all the selections/settings are correct for the analysis, then continue to Step 2.
   - If you must edit any of the selections or settings, then click a section header/page name to return to the page in the wizard. Edit the selections/settings as appropriate, and then click Next to move through the wizard (making additional edits as required) until you return to the Summary page, and then continue to Step 2.

Any edit that you make to the selections/settings for a scheduled vessel might affect downstream selections/settings. For example, if you select a different analysis type, then you must edit the analysis parameters for an image channel. As a result, you always have to move through the remaining pages on the wizard after you edit any selections/settings for the vessel.

2. Click Finish.

   The analysis is launched and an Analysis Created dialog box opens. The dialog box indicates that the analysis has been created and that it will be processed as soon as possible. It also indicates that you can view the status through the Analysis Queue and that you will be directed to the queue after you close the dialog box. See Figure 1-12 on page 128.

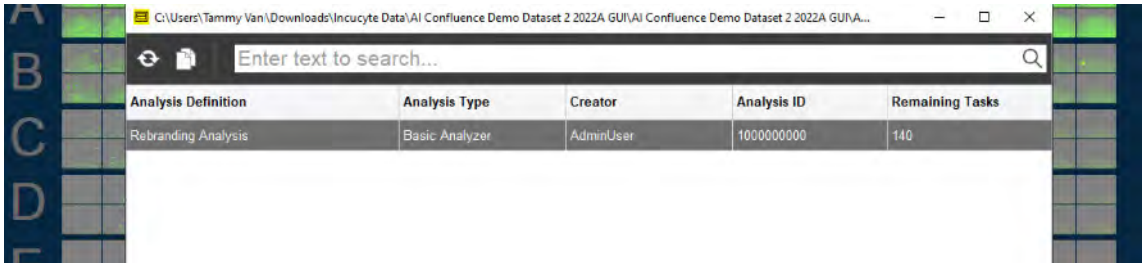
Figure 1-12: Analysis Created dialog box



3. Click OK.

   The Analysis Created dialog box closes and the Analysis Queue opens on top of the Vessel View window. The queue displays all the analyses that are currently taking place in your Incucyte system in reverse chronological order based on their launch dates and times.

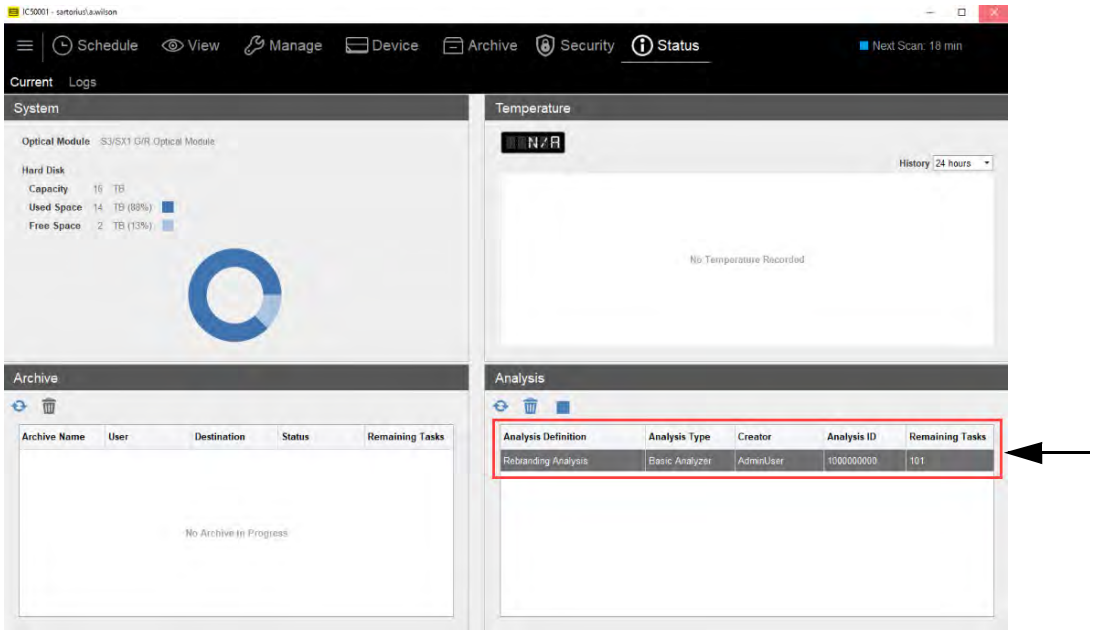
Figure 1-13: Analysis Queue



You can now view information about the analysis in two locations in the Incucyte GUI.

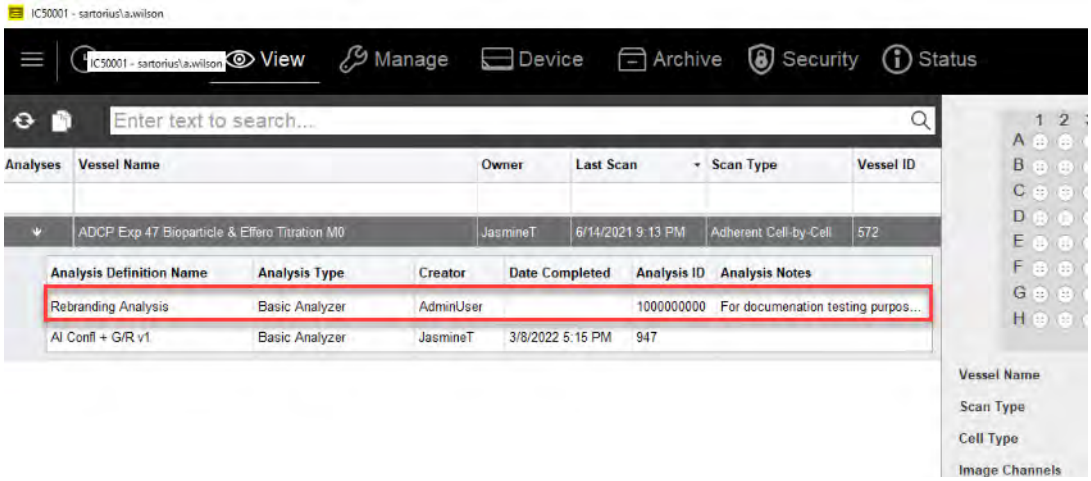
- You can view the information that is displayed in the Analysis Status dialog box on the Status page. On the main menu, click Status and view this information in the Analysis (lower right) pane on the Status window.

Figure 1-14: Status window



- You can open the Scanned Vessels window, and then open the Analysis pane for the vessel. While the analysis is in progress, the pane displays the analysis without a Date Completed time stamp. After the analysis is completed, if you refresh the Analysis pane, a Date Completed time stamp is displayed.

Figure 1-15: Analysis pane showing all the analyses (in progress and completed) for a selected vessel



Copying an Existing Analysis Definition

If an existing analysis definition is similar to the new definition that you are creating, then you can copy the existing definition, which is considered to be a template, and then edit it as appropriate to create a new definition. When you select an existing analysis definition to copy, you must select one that has the required channels and objective. (To add or remove channels in the analysis, you must create an entirely new definition and because the objective is set during acquisition, you cannot select an objective in the launch analysis workflow.)

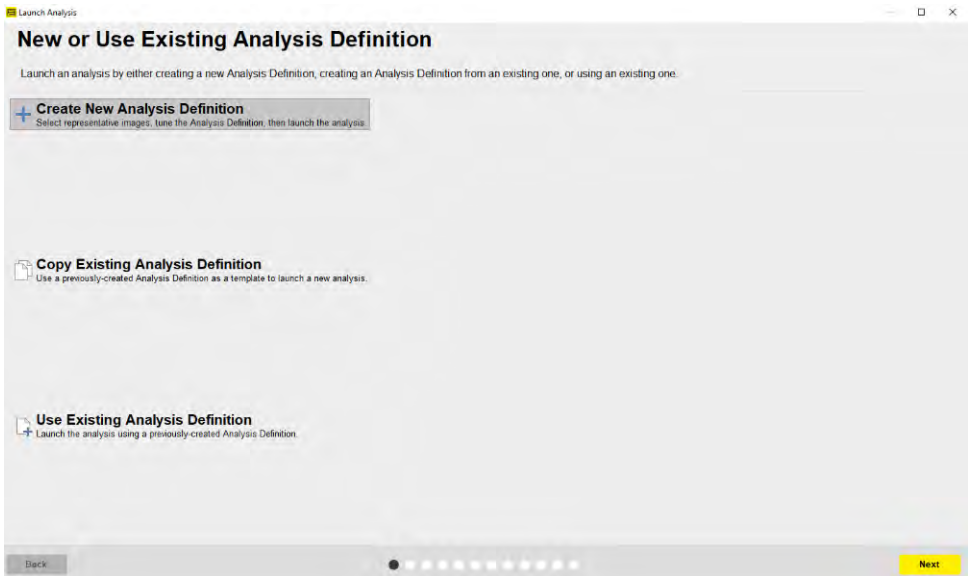
For detailed information about creating a new analysis definition, see "Setting up a New Analysis" on page 116.

To copy an existing analysis definition

1. Open the appropriate vessel in the Vessel View window. See "To open the Vessel View window" on page 85.
2. On the Analysis toolbar, click the Launch Analysis icon.

   The Launch Analysis wizard opens. The New or Use Existing Analysis Definition page is open. Create New Analysis Definition is selected.

Figure 1-16: Launch Analysis wizard, New or Use Existing Analysis Definition page



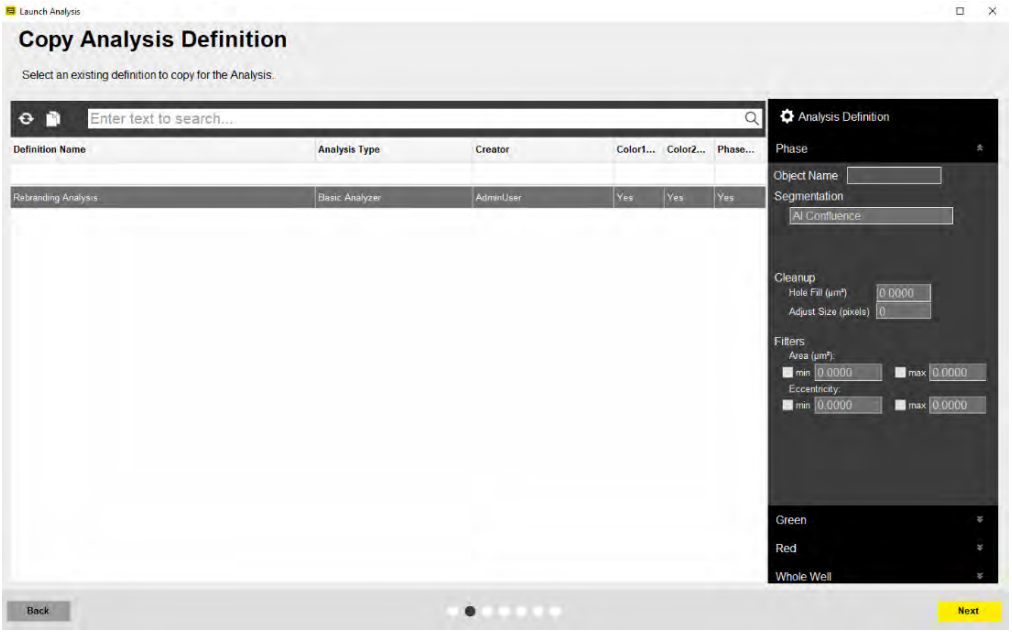
3. Select Copy Existing Analysis Definition, and then click Next.

   The Copy Analysis Definition page opens. The page lists all the analysis definitions in the Incucyte database that are available for copying based on your user type and the workgroups with which the definition is currently shared. See Figure 1-17 on page 131.

   - If you are an Admin user, then every analysis definition that was created on/imported to the Incucyte is displayed on the page.

- If you are not an Admin user, then only those definitions that have been shared with the **Everyone** user workgroup or with a workgroup that you are a member of are displayed.

Figure 1-17: Launch Analysis wizard, Copy Analysis Definition page



4. Select the analysis definition that you are using as your template.

All the analysis parameters for the selected definition are displayed in read-only mode in the Analysis Definition (right) pane on the page. Before continuing, make sure to confirm that the selected analysis definition has the correct objective and image channels.

The Copy Analysis Definition page has the standard Incucyte filter and sort functions that you have encountered on other wizard pages, for example, on the Vessel Type Search page in the Add Vessel wizard. You can use these features to help you quickly search for and select the appropriate analysis. See "Working with Data Columns in an Incucyte Window" on page 26.

5. Click Next.

   The Image Set Selection window opens. With the following exceptions, the remainder of the procedure is now identical to that when creating a new analysis definition.

   - On the Save and Apply Analysis Definition page, the Definition Name field is pre-populated with the name of the copied analysis definition appended with the word "Copy," for example, CytoToxGreen_Copy. You can edit this value.
   - On the Summary page, the first heading is Copy Existing Analysis Definition.

6. Continue to "To select the images for analysis" on page 120.

Using an Existing Analysis Definition

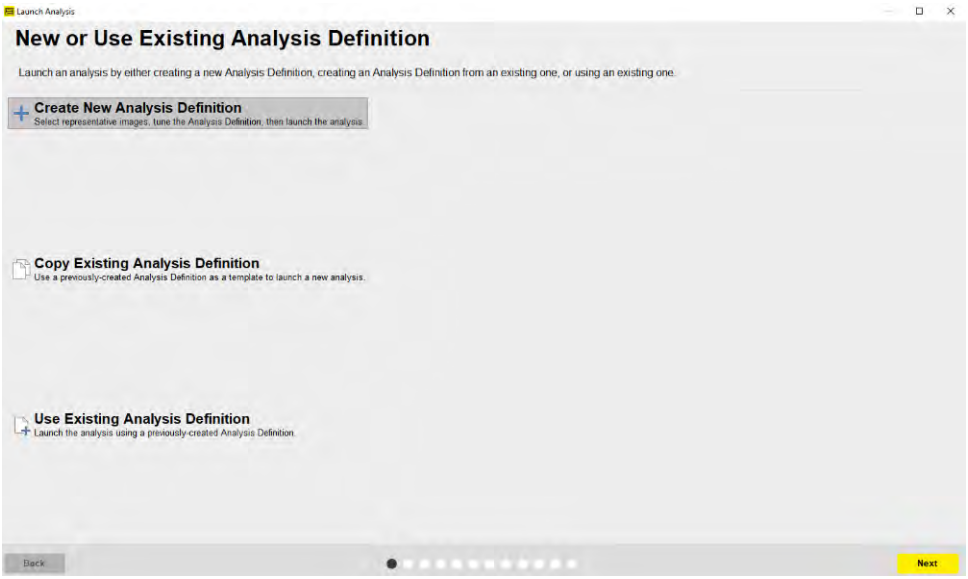
Because you cannot edit any of the analysis parameters for an existing analysis definition, you must use one that is defined for your scan type, vessel type, objective, and all the color channels and the phase that you used for your analysis.

To use an existing analysis definition

1. Open the appropriate vessel in the Vessel View window. See “To open the Vessel View window” on page 85.
2. On the Analysis toolbar, click the Launch Analysis icon.

   The Launch Analysis wizard opens. The New or Use Existing Analysis Definition page is open.

Figure 1-18: Launch Analysis wizard, New or Use Existing Analysis Definition page



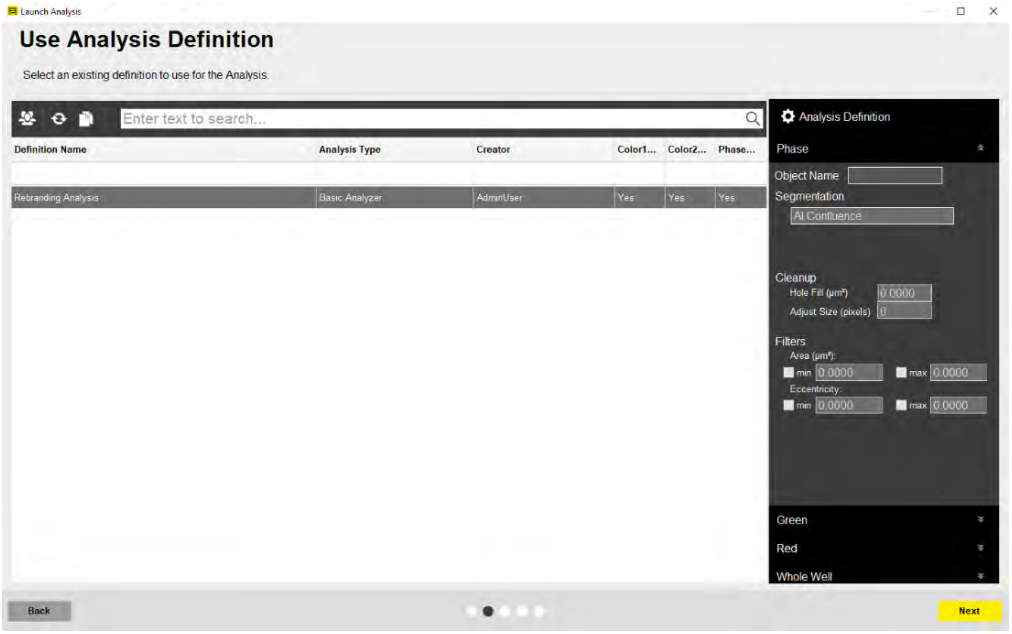
3. Select Use Existing Analysis Definition.

   The Use Analysis Definition page opens. The page lists all the current analysis definitions in the Incucyte database based on *both* the following caveats:

   - The definitions are applicable for your scan type and vessel type (microplate, flask, or so on) and that use the same objective and at least one of the same channels for image acquisition.
   - Your user type and the workgroups with which the definition is shared.
     - If you are an Admin user, then every analysis definition that was created on/imported to the Incucyte and that meets the first caveat is displayed on the page.
     - If you are not an Admin user, then only those definitions that have been shared with the **Everyone** user workgroup or with a workgroup that you are a member of are displayed and that meet the first caveat are displayed.

   See Figure 1-19 on page 133.

Figure 1-19: Launch Analysis wizard, Use Analysis Definition page



4. Select the correct analysis definition.

All the analysis parameters for the image channels are displayed in read-only mode in the Analysis Definition (right) pane on the page. Before you select an analysis definition, make sure to view the relevant information for each image channel to confirm that values are appropriate for your analysis.

The Analysis Definition page has the standard Incucyte filter and sort functions that you have encountered on other wizard pages, for example, on the Vessel Type Search page in the Add Vessel wizard. You can use these features to help you quickly search for and select the appropriate analysis. See "Working with Data Columns in an Incucyte Window" on page 26.

5. Click Next.

   The Scan Times and Wells page opens. With the following exceptions, the remainder of the procedure is now identical to that when creating a new analysis definition.

   - The Analysis Notes page replaces the Save and Apply Analysis Definition page. The Definition Name is read-only. You can enter only analysis notes on this page.
   - On the Summary page, the first heading is Use Existing Analysis Definition.

6. Continue to "To select the scan times and wells/sectors/image sites" on page 123.

Chapter 2
Visualizing the Analysis Results

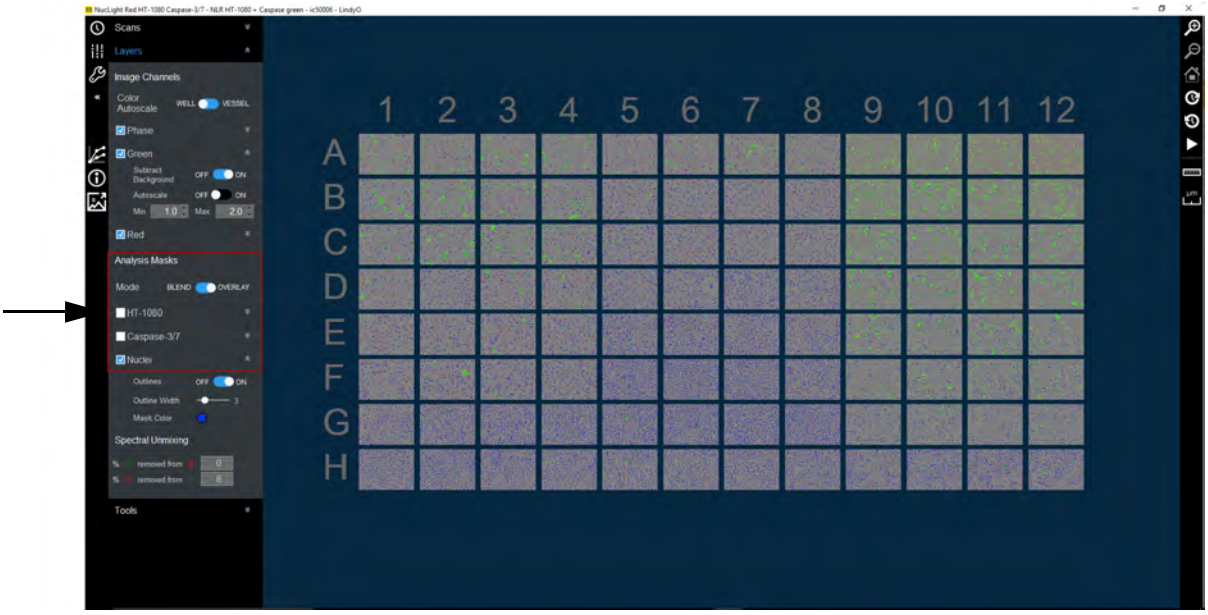
After your analysis is complete, the Incucyte software provides several ways for the visualization and investigation of your results, including the Vessel View with Analysis window and the graphing of your analysis results. This chapter details the various ways that you can visualize your analysis results so that you can not only access and interpret your data and identify the keys areas of your assay that worked well, but also identify the areas of your assay that require attention or improvement.

This chapter covers the following topics:

Viewing the Analysis Masks

After your vessel data has been analyzed, you can open the Vessel View window, which now displays the associated analysis masks that were applied to the images. (See “Open Analysis” on page 83.) To view the analysis masks, open the Image Channels popup (see “The Image toolbar” on page 95), and then in the Analysis Masks pane, toggle on and off the analysis masks as appropriate.

Figure 2-1: Vessel View window with the associated analysis, toggling image masks on and off

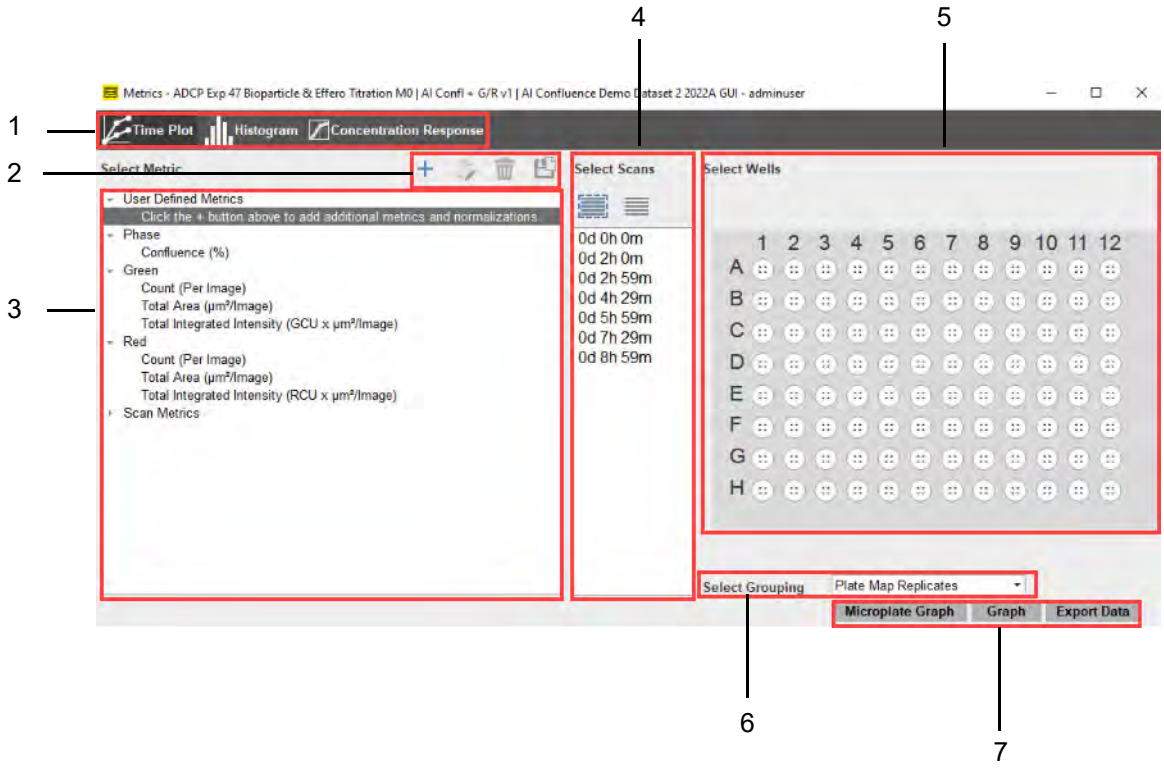


The Graph Metrics Window

The Graph Metrics window is the starting point for graphing your analysis results. The window provides all the functionality for graphing the analysis results for a microplate (Microplate Graph) or vessel (Vessel Graph), and for exporting the analysis results (Export Data). You can graph the results for a single metric at a time, where the metric is a pre-defined metric or a user-defined metric. To open the Graph Metric window, do one of the following:

- On the Vessel View window that shows the analysis masks, click the Graph Metrics icon. See "Viewing the Analysis Masks" on page 137.
- In the Vessel View window, right-click the analysis and on the context menu that opens, click Graph Analysis Metrics. See "To view the analysis definitions for a scanned vessel" on page 83.

Figure 2-2: Graph Metrics window



The Graph Metrics window has the following layout:

	Window component	Description
1	Graph menu	Displays the three types of graphs that you can plot for analysis results: a Time Plot (the default value), a Histogram, or a Concentration Response curve.

	Window component	Description
2	Metric menu: Displays options for working with a user-defined metric.	
		Create Metric: Opens the Create Metric dialog box, which contains all the options for defining a new metric for the analysis. See "To define user metrics" on page 140.
		Edit Metric: Opens the Edit Metric dialog box, which displays all the values for the currently selected user-defined metric. You can edit these values as needed and save the edited metric. **Note:** This action does not change any data for the currently selected analysis. If the metric was linked to the analysis definition, this action also does not affect the data for any other analysis that used the same analysis definition.
		Delete Metric: Deletes the currently selected user-defined metric from the analysis. **Note:** This action does not delete any data for the current analysis. If the metric was linked to the analysis definition, this action also does not affect the data for any other analysis that used the same analysis definition. The metric is simply not available going forward for any other analyses.
		Apply Metric to Definition: Saves and links the currently selected user-defined metric to the analysis definition that was used to generate the current data. This user-defined metric will now always be available when you use the same analysis definition for any other vessel.
3	Metrics pane	Always displays the pre-defined Analysis and Scan metrics that can be plotted for your analysis. Scan metrics are always displayed for any vessel. The analysis metrics that are available for graphing are specific to the channels that were used to acquire the images. The analysis metrics are grouped by image channel (click the Expand/Collapse icon next to a grouping to view the metrics in the grouping), and the name for each analysis metric is preceded by the object name that was specified in the analysis definition for the channel. If any user variables have been defined for the analysis, then these metrics are displayed under the heading of User Defined Metrics. **Note:** If you opened the Graphic Metrics window for a vessel from the Vessel View window (no analysis), then only the Scan metrics for the vessel are available for graphing. See Graph Metrics icon on page 89.
4	Scan pane	Displays the options for selecting the scans for which you are graphing the metrics.
5	Vessel pane	Displays the acquisition map for the scanned vessel. The number of images that have been acquired for each vessel location (as specified in the Add Vessel wizard) is indicated by the number of individual solid black squares in the vessel locations.

	Window component	Description
6	Select Grouping dropdown list	Displays the options for specifying how to group the data points on the Y axis. The default values are the following: • If the vessel is a microplate and a plate map has been specified, then the data is grouped by Plate Map Replicates. • If the vessel is a microplate and a plate map has *not* been specified, then the default grouping is "None." • If the vessel is any vessel other than a microplate, then the default grouping is "None." Other options are All, Columns, and Rows. **Note:** For information about exporting graph data, "Exporting Graph Data" on page 162.
7	Analysis Data options	• Microplate Graph: Option to graph the analysis results specifically for a microplate. • Vessel Graph: Option to graph the analysis results for any vessel type, including a microplate. • Export Data: Option to export selected analysis results to your client clipboard or to a text file.

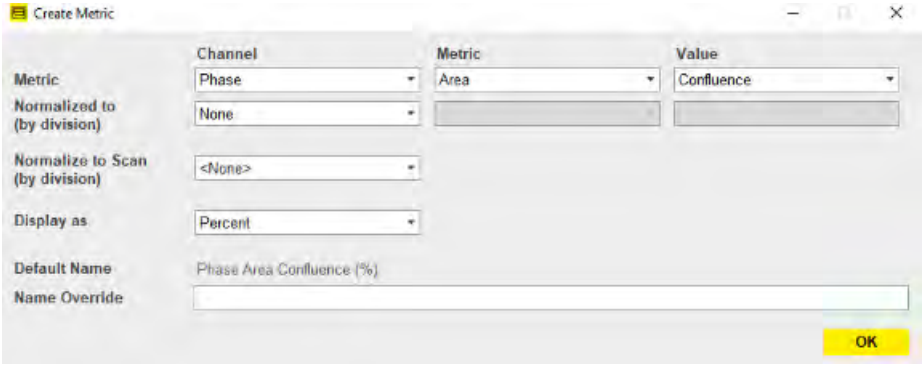
To define user metrics

Basic Analyzer, Cell-by-Cell, and Cell-by-Cell Classification analyses support the creation of user-defined metrics for the purpose of normalizing data. You can define user metrics for these analyses that are based on division calculations to normalize data to another metric, to a scan time, or to both. For example, for an apoptotic event, you might normalize to another metric, where the metric Green/Area/Confluence is normalized to the metric Phase/Area/Confluence. To account for changes in seeding density, you might normalize to a scan time, where the scan time that the data is normalized to is the starting scan time.

1. On the Metric menu, click the Create Metric icon.

   The Create Metric dialog box opens.

Figure 2-3: Create Metric dialog box



2. Enter the values—Channel, Metric, and Value—for the metric that you are normalizing.

The selected Metric determines the available options for Value.

3. Do one of the following:
   - To normalize to another metric, enter the metric value—Channel, Metric, and Value—for Normalized to (by division).
   - To normalize to a scan time, on the Normalize to Scan dropdown list, select the scan time.
   - To normalize to both another metric and a scan time, enter the metric value—Channel, Metric, and Value—for Normalized to (by division) *and* on the Normalize to Scan dropdown list, select the scan time.
4. For Display as, leave the default value as-is, or select another value from the list of available values.

The Display as value determines how the normalized values are displayed in the Metric graph. The normalization options that you have selected determine the available Display as values. For example, if you have selected Normalize to Scan, then the two available values for Display as are Ratio and Percent.

5. The default name for the metric indicates the division calculation that is being carried out, for example, when normalizing to a metric, Green Area/Phase Area. You can leave the default name as-is, or optionally, in the Name Override field, enter a different name for the metric.
6. Click OK.

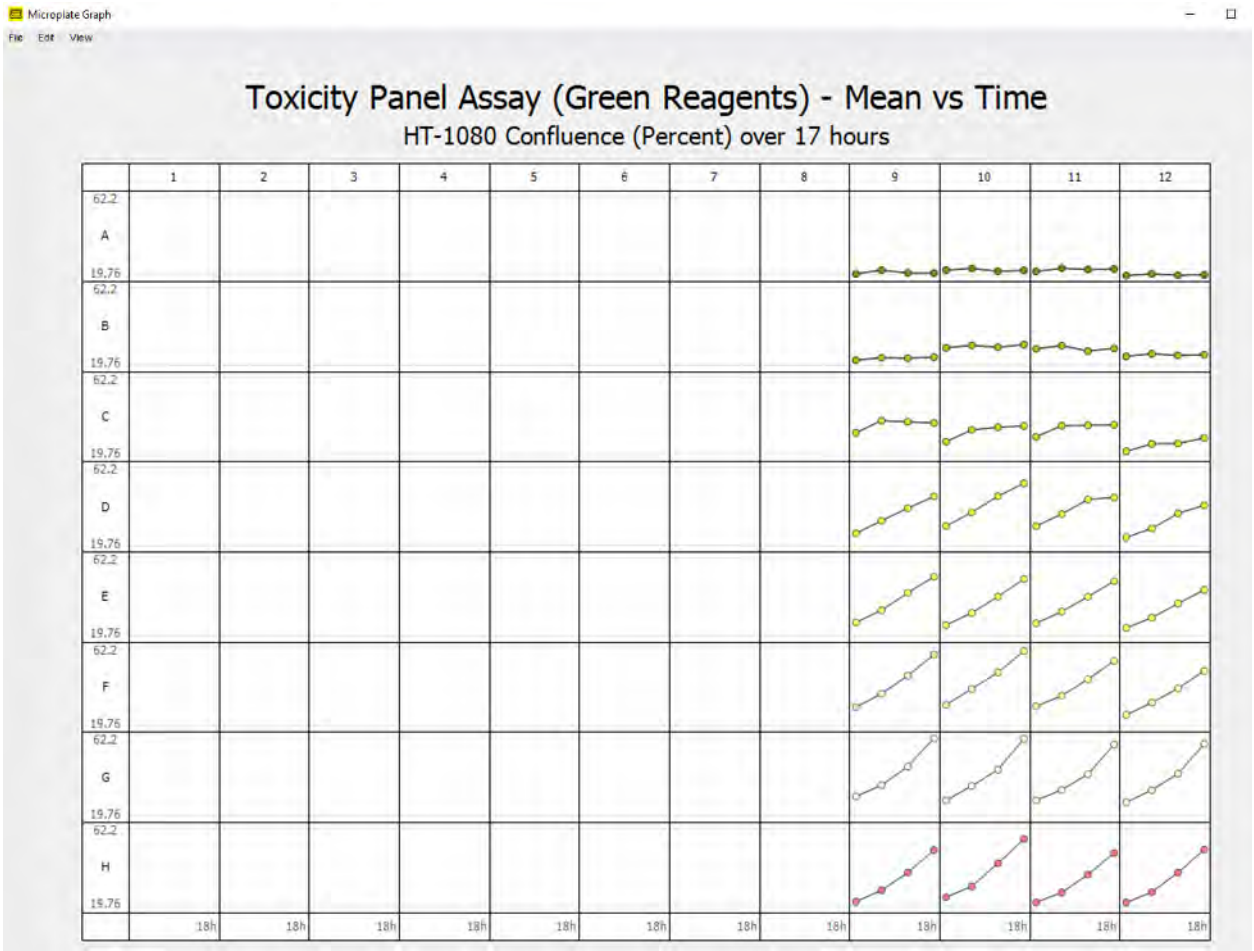
   The Create Metric dialog box closes. The metric is displayed in the Metrics pane under the heading User Defined Metrics.
7. Optionally, to save and link the user-defined metric to the analysis definition that was used to generate the current data, on the Metric menu, click the Apply Metric icon.

   This user-defined metric is now always be available when you use the same analysis definition for any other vessel.
8. You can now graph this user-defined metric. Continue to one of the following:
   - “Graphing the Analysis Results for a Microplate” on page 142.
   - “Graphing the Analysis Results for Any Vessel Type” on page 146.
   - “Graphing Analysis Results as a Histogram” on page 151.

Graphing the Analysis Results for a Microplate

You use the Microplate Graph option to graph the analysis results specifically for a microplate. The graph that is produced is displayed in the same layout as the selected microplate, with an individual data point plotted for each scan time that was selected for graphing in every well that was selected for graphing. If a plate map was specified, then the colors that are displayed in the graph correspond to the colors that were defined in the plate map. After the default graph is generated and displayed, options are available for customizing the graph, including options that are required for sharing the graph in presentations or reports.

Figure 2-4: Microplate graph example



In this example, we assume a 96-well plate for which a plate map was specified. Four scan times were selected for graphing and image data in Wells 9 through 10 in Rows 1 through 8 was selected for graphing.

To graph the analysis results for a microplate

1. Open the Graph Metrics window. See "The Graph Metrics Window" on page 138.
2. On the Graph menu, leave Time Plot selected.

3. In the Metrics pane, select the metric for which you are graphing the analysis results.

You might have to click the Expand/Collapse icon next to a grouping to expand the grouping and view the available metrics.

4. Select the scan times and images that you are graphing.

Option	Description

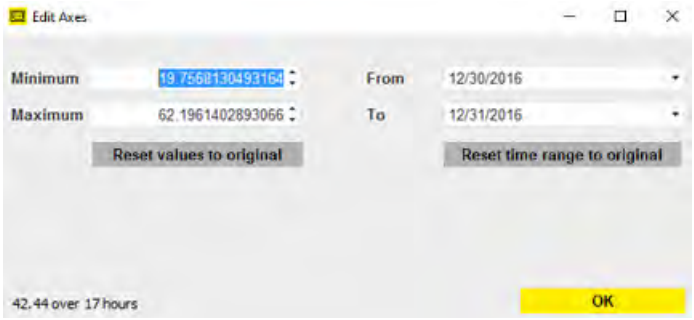
To select scan times	• To select all scans in a single step, under Select Scans, click the Select All Scans icon . To clear an individual scan, click it.
	• In the Vessel Scanning Timeline, do either one or both of the following:
	  • To select an individual scan, click the scan. You can select multiple, individual scans. To clear an individual scan, click it again.
	  • To select a range of scans, click the first scan in the series of scans, press and hold the Shift key, and then click the last scan in the series of scans.
	• To clear all selected scans in a single step, under Select Scans, click the Clear All Scans icon .

To select wells/image sites	• To select a single location in a vessel, for example, a well in a 96-well microplate, click once in the appropriate location in the vessel map. • To select multiple contiguous locations in a vessel, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them. • To select all the locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Select this Column. Conversely, to clear all the selected locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Deselect this Column. • To select all the locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Select this Row. Conversely, to clear all the selected locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Deselect this Row. **Tip:** To clear all the selected locations in the vessel map, or multiple contiguous locations, you can also press and hold the ALT key, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

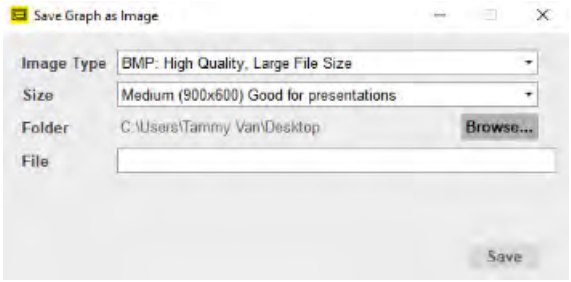
5. On the Select Grouping dropdown list, select the grouping option.
6. Click Microplate Graph.

   The Microplate Graph is generated and displayed on-screen in its own window. A menu (File, Edit, View) is displayed at the top of the graph window and a default title is displayed above the graph. The display is color-coded according to the colors that were specified in the plate map.

7. Optionally, after the microplate graph is generated, do any of the following to customize the graph display:

Option	Description
Edit	
Titles and Subtitles	Opens the Edit Titles dialog box. You use the options on this dialog box to edit the default title for the graph and/or add your own custom subtitles. You can also change the maximum font size of the title and subtitles. *Figure 2-5: Edit Titles dialog box* 
Axis Scales	Opens the Edit Axes dialog box. You use the options on this dialog box to edit the system-generated values for the X and Y axes. You can always click Reset values to original and/or Reset time range to original to reset the values to their system- generated values. *Figure 2-6: Edit Axes dialog box* 
View	
Logarithmic Y-Axis	Click to toggle the display of the Y axis between a decimal scale and a logarithmic scale.
Show Time Axis	Selected by default. Click to toggle the display of the X axis on and off.
Show Plate Map Colors	Available only for a microplate for which a Plate Map is specified. Selected by default. The data points are plotted in the colors as specified in the Plate Map Editor. Click to toggle the color display on and off. If you toggle this option off, then all the data points are displayed in blue.
Marker Size	The size of the data points in the graph. Available options are Small, Medium (the default value), and Large. As you select a different size, the graph display is dynamically updated.

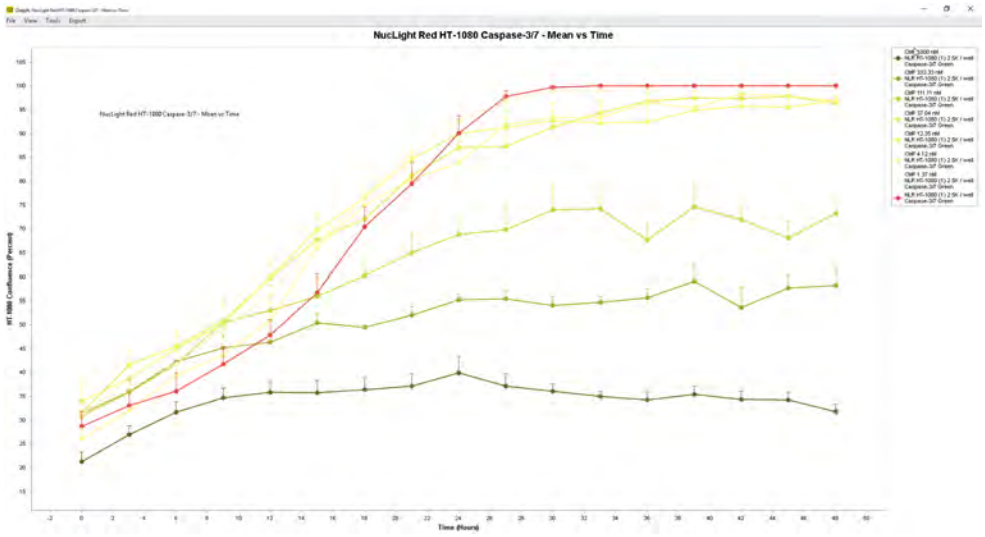
8. Optionally, open the File menu, and then do any or all of the following as appropriate:

Option	Description
Save As Image	Opens the Save Graph as Image dialog box. You use the options on this dialog box to save the displayed graph as image in a specific format (BMP, JPEG, PNG, or TIFF) and with a specific size. To save the file, you must select the location in which to save the image file, and you must name the file. **Note:** The default values are: • Image Type: BMP, High Quality, Large File Size • Size: Medium (900 x 600) Good for presentations • Folder: C:Users\<User Name>\Desktop *Figure 2-7: Save Graph as Image dialog box* 
Print	Opens the standard Windows Print dialog box for printing the graph.
Print Preview	Opens a Print Preview dialog box in which you can view what the graph looks like before it is printed, and make adjustments to the graph (such as zooming in on an area, or changing the number of pages across which you are printing the graph) before printing the graph.

Graphing the Analysis Results for Any Vessel Type

You use the Graph option to graph the analysis results for any vessel type, including a microplate for which a plate map was specified. The graph that is generated is a standard time plot graph, with the selected scan time (Time) on the X axis and the selected metric on the Y axis. If the range of scan times is less than or equal to 96 hours, then, by default, the time is displayed in hours. If the range of scan times is greater than 96 hours, then, by default, the time is displayed in days. After the default graph is generated and displayed, options are available for customizing the graph, including options that are appropriate for sharing the graph in presentations or reports. You can graph a single metric for the analysis results for any vessel type, or you can graph multiple single metrics for the analysis results for any vessel type, and then combine the individual metric graphs in to a single multi-metric graph, which is referred to as the Graph on Graph option.

Figure 2-8: Graph example



The following procedure assumes a 96-well plate for which a plate map was specified. The majority of information, however, is "vessel-agnostic" and offers a frame of reference for graphing data for any vessel type.

To graph the analysis results for any vessel type (Single metric)

1. Open the Graph Metrics window. See "The Graph Metrics Window" on page 138.
2. On the Graph menu, leave Time Plot selected.
3. In the Metrics pane, select the metric for which you are graphing the analysis results.

You might have to click the Expand/Collapse icon next to a grouping to expand the grouping and view the available metrics.

4. Select the scan times and images that you are graphing.

Option	Description

To select scan times	• To select all scans in a single step, under Select Scans, click the Select All Scans icon  . To clear an individual scan, click it.
	• In the Vessel Scanning Timeline, do either one or both of the following:
	  • To select an individual scan, click the scan. You can select multiple, individual scans. To clear an individual scan, click it again.
	  • To select a range of scans, click the first scan in the series of scans, press and hold the Shift key, and then click the last scan in the series of scans.
	• To clear all selected scans in a single step, under Select Scans, click the Clear All Scans icon  .
	Note: For an endpoint analysis, select a single time point to generate the corresponding bar graph. See Figure 2-9 on page 148.

To select wells/image sites	• To select a single location in a vessel, for example, a well in a 96-well microplate, click once in the appropriate location in the vessel map. • To select multiple contiguous locations in a vessel, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them. • To select all the locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Select this Column. Conversely, to clear all the selected locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Deselect this Column. • To select all the locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Select this Row. Conversely, to clear all the selected locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Deselect this Row. **Tip:** To clear all the selected locations in the vessel map, or multiple contiguous locations, you can also press and hold the ALT key, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

5. On the Select Grouping dropdown list, select the option for grouping the data.

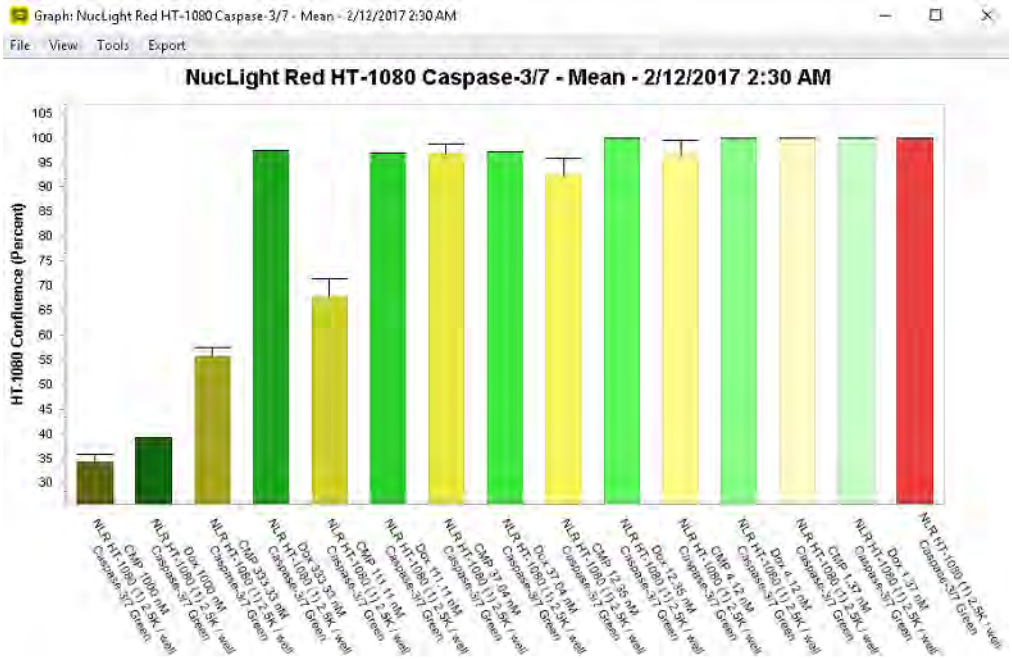
If the vessel is a microplate and a plate map has been specified, then by default, Plate Map Replicates is selected and this is the recommended value. If a plate map has not been specified or the vessel is any other vessel type, then None is the default value and you can select another value (All, Rows or Columns). In this example, we assume that Plate Map Replicates is selected.

6. Click Graph.

   The Graph is generated and displayed on-screen in its own Graph window. If the vessel is a microplate and a plate map has been specified, then information from the plate map (colors, concentrations, and so on) is carried over for the legend. You can hold your mouse pointer over any data point in the graph to open a tooltip that displays information about the data point. See Figure 2-9 on page 148.

7. Optionally, after the time plot graph is generated, options are available for customizing the graph display and exporting the graph data. See “Customizing a Time Plot Graph, Histogram, or a Concentration Response Curve” on page 157 and/or “Exporting Graph Data” on page 162.

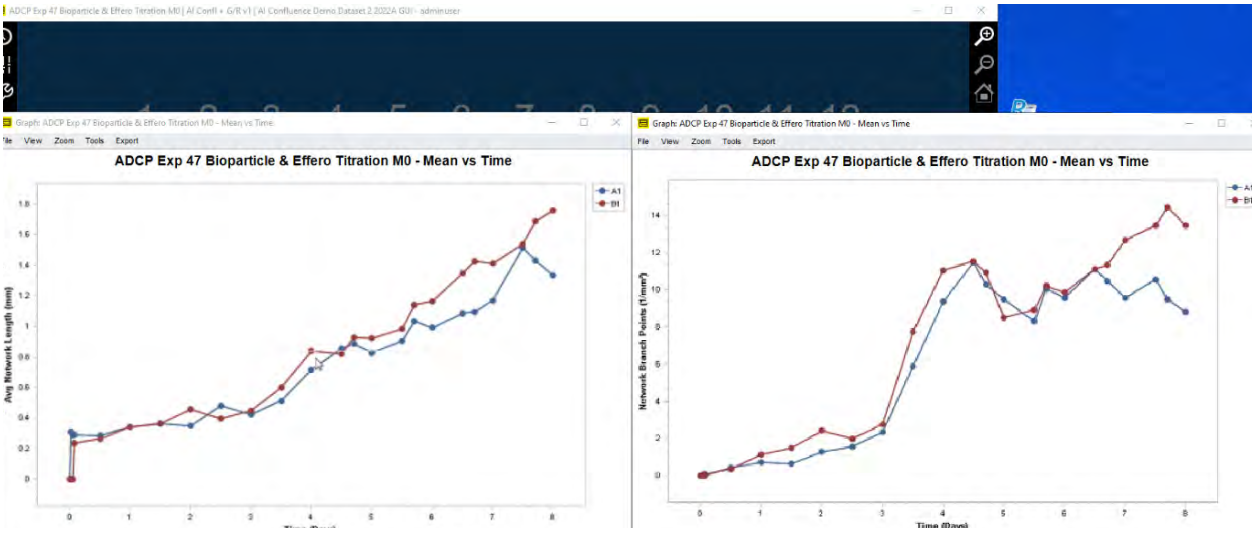
Figure 2-9: Endpoint analysis graph (Single time point and Plate Map Replicates selected)



To combine multiple metrics into a single graph (Graph on Graph)

1. For *each* metric that you are including in the multi-metric graph, graph the individual analysis results. See "To graph the analysis results for any vessel type (Single metric)" on page 146.
2. After you have graphed all the individual analysis results for each metric that you are including in the multi-metric graph, use standard Windows functions to bring all the individual metric graphs to the front of the screen display. For example, Figure 2-10 below shows two individual metric graphs brought to the front of the screen display.

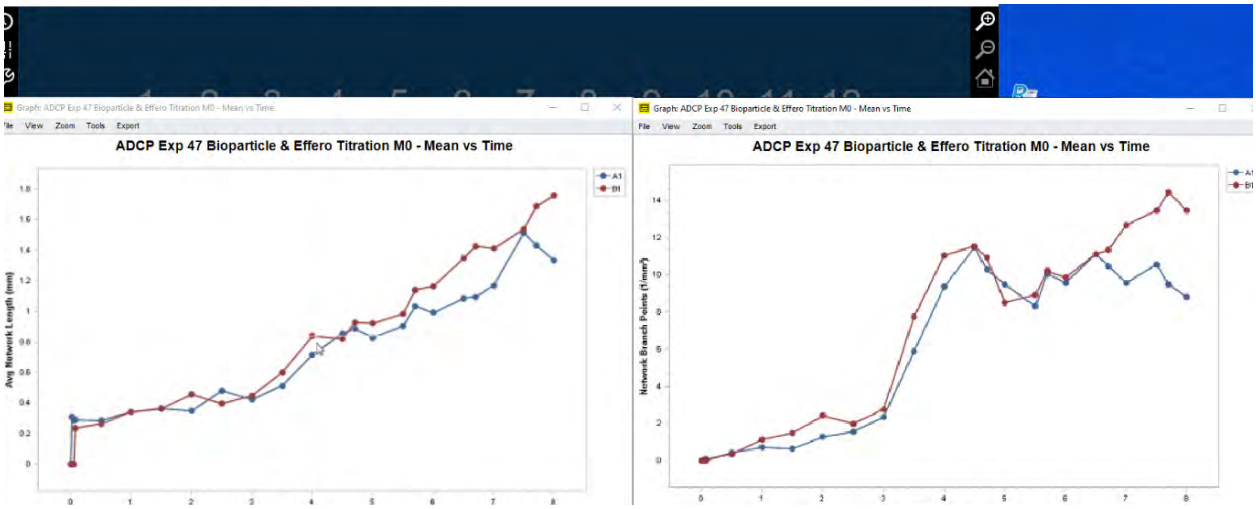
Figure 2-10: Graphs of analysis results for individual metrics brought to the front of the screen display



3. Determine which of the individual metric graphs should be the *destination* graph. The destination graph is the graph onto which you are dropping the other individual metric graphs.
4. For *each* individual metric graph that you are dropping onto the destination graph, do the following:
   a. On the graph's main menu, click View > Drag and Drop > Graph on Graph.

      A faint 2 x 2 grid is displayed on the metric graph. (In Figure 2-11 below, the grid is displayed on the left metric graph.)

Figure 2-11: 2 x 2 grid on left metric graph

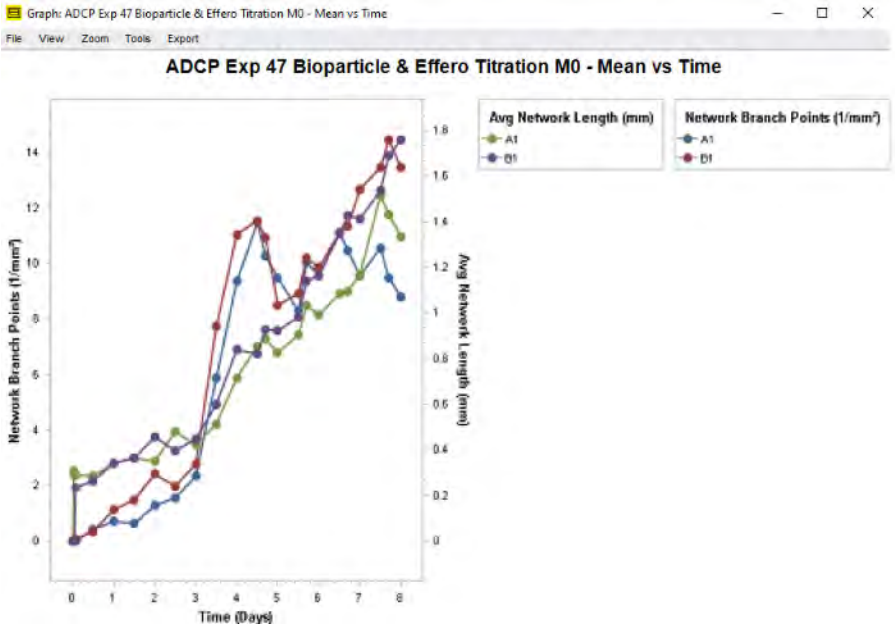


   b. Click and hold the *right* mouse button on the metric graph, drag the graph onto the destination graph, and then release the mouse button.

      The two individual metric graphs are combined into a single metric graph with the following characteristics:

      - The graph window is set to a default size. You must use standard Windows functionality to manually resize the graph window. The destination graph size is adjusted accordingly.
      - By default, the individual graph legends are displayed to the right and outside of the combined graph. You can manually adjust the position of each graph legend. See "To customize the graph display (Non-default properties)" on page 159.
      - If two more or more graphs use the same legend colors, then when the graphs are combined, the software automatically modifies the colors for one of the graphs so that the different data points can be distinguished from each other. For example, as shown in Figure 2-11 above, both individual metric graphs used the colors blue and red for their legends. After the graphs were combined into a single graph, as shown in Figure 2-12 on page 150, the software changed the legend for one of the graphs from blue and red to green and purple. You can manually adjust the options (visibility, back color, font, and/or color) for any legend. See "To customize the graph display (Non-default properties)" on page 159.

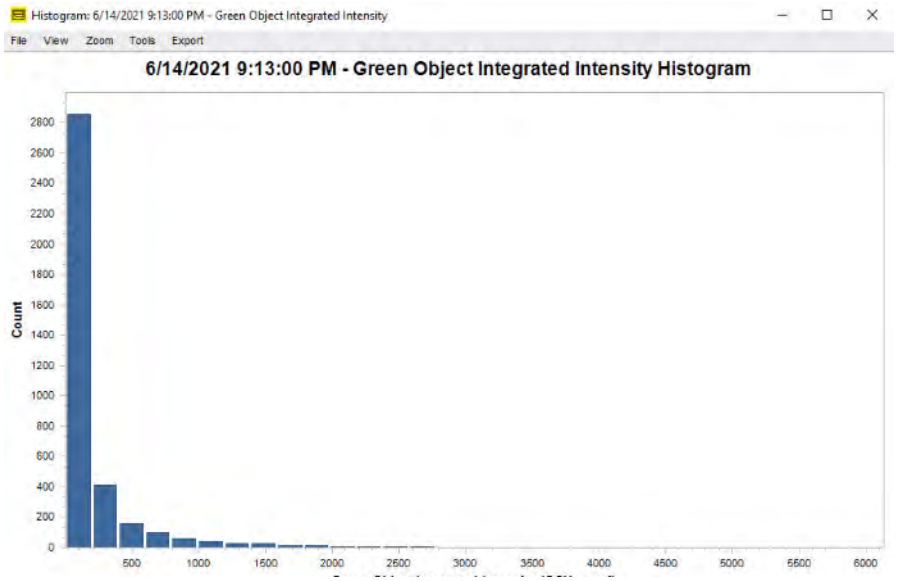
Figure 2-12: Graph on Graph example



Graphing Analysis Results as a Histogram

In addition to the common time plot, you can also graph your analysis results as a histogram. Typically, you graph your analysis results as a histogram if you are interested in determining the intensity of a fluorophore. For example, for a transfection, how consistent is the expression level based on your selected fluorophore?

Figure 2-13: Histogram example



To graph analysis results as a histogram

1. Open the Graph Metrics window. See "The Graph Metrics Window" on page 138.
2. On the Graph menu, click Histogram.
3. In the Metrics pane, select the metric for which you are graphing the analysis results.

You might have to click the Expand/Collapse icon next to a grouping to expand the grouping and view the available metrics.

4. To select an individual scan, click the scan. To clear an individual scan, click it again.

You can select only a single scan time for a histogram. You cannot select a range of scans and the Select All and Deselect All icons are not enabled.

5. Select the wells/image sites.
   - To select a single location in a vessel, for example, a well in a 96-well microplate, click once in the appropriate location in the vessel map.
   - To select multiple contiguous locations in a vessel, click and hold the left mouse button, and then

drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

- To select all the locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Select this Column. Conversely, to clear all the selected locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Deselect this Column.
- To select all the locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Select this Row. Conversely, to clear all the selected locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Deselect this Row.

To clear all the selected locations in the vessel map, or multiple contiguous locations, you can also press and hold the ALT key, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

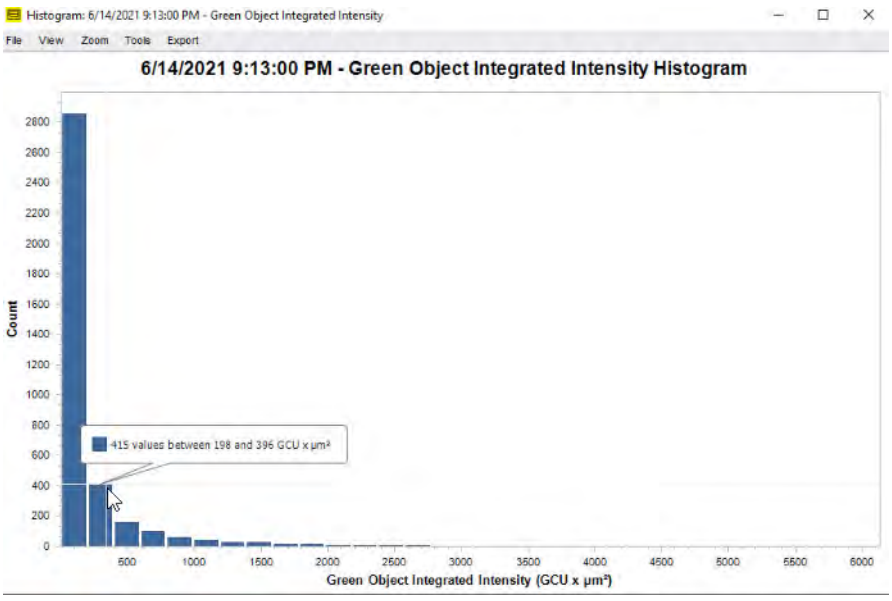
6. Leave AutoScale set to On, or optionally, turn AutoScale off, and then manually adjust the values for Min, Max and/or Bins.

After you select the wells/image sites that you are analyzing, the values for Min, Max and Bins are automatically generated. Although you can manually adjust these values, Sartorius recommends that you at least first graph the data according to these system-generated values.

For finer resolution of the data, you can increase the number of bins.

7. Optionally, after the histogram is generated:
   - You can hold your mouse pointer over any histogram ban to open a tooltip that indicates the number of values that are found within the selected bin as well as the minimum and maximum values for the bin. See Figure 2-14 on page 153.
   - You can customize the graph display and export the graph data. See:
     - “Customizing a Time Plot Graph, Histogram, or a Concentration Response Curve” on page 157.
     - “Exporting Graph Data” on page 162.

Figure 2-14: Histogram with bin tooltip



Graphing Analysis Results as a Concentration Response Curve

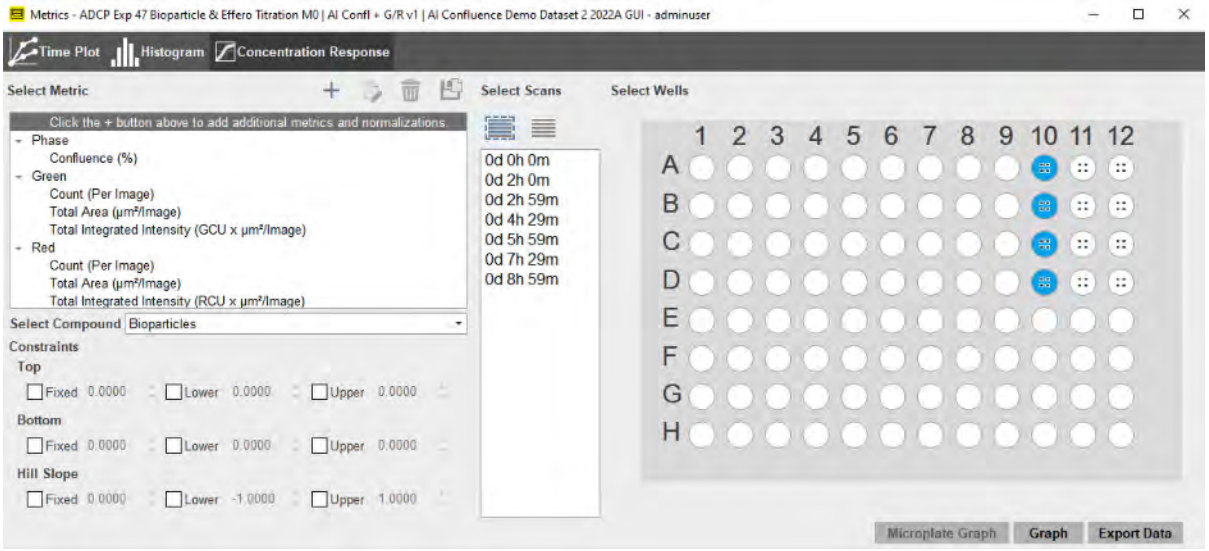
In addition to the common time plot and histogram, you can also graph your analysis results as a concentration response curve. Typically, you graph your analysis results as a concentration response curve if you are interested in determining the EC_{50} value of your compound or treatment. To graph analysis results as a concentration response curve, your plate must have an associated Plate Map that consists of at least one treatment at three different concentrations. See Appendix A, “Plate Map Editor,” on page 167.

To graph analysis results as a concentration response curve

1. Open the Graph Metrics window. See “The Graph Metrics Window” on page 138.
2. On the Graph menu, click Concentration Response.

   The Graph Metrics window is refreshed with a Select Compound dropdown list and a Constraints panel.

Figure 2-15: Graph Metrics window for an EC_{50} analysis



3. In the Metrics pane, select the metric for which you are graphing the analysis results.

You might have to click the Expand/Collapse icon next to a grouping to expand the grouping and view the available metrics.

4. On the Select Compound dropdown list, select the compound for which you are generating the concentration response curve.

   After you select the compound, all the wells that can be graphed are automatically selected based on the corresponding Plate Map. You can leave all the wells selected, or you can deselect wells of your choosing, for example, outliers.

   - To clear a single well, click the well.
   - To clear all the selected locations in a vessel column in a single step, right-click a location in the column, and on the context menu that opens, select Deselect this Column.
   - To clear all the selected locations in a vessel row in a single step, right-click a location in the row, and on the context menu that opens, select Deselect this Row.

To clear all the selected locations in the vessel map, or multiple contiguous locations, you can also press and hold the ALT key, click and hold the left mouse button, and then drag the mouse pointer over the appropriate locations in the vessel map. A box is formed around the locations as you are selecting them.

5. Select the scan time or times that you are graphing.
   - For a kinetic EC_{50}, select the full time course or a subset of time points.
   - For a single time point EC_{50}, select the specific acquisition time.

For a shortcut to generate a single EC_{50} time point graph, you can first generate the kinetic EC_{50} graph, and then click a point on the kinetic graph to generate the corresponding single time point graph.

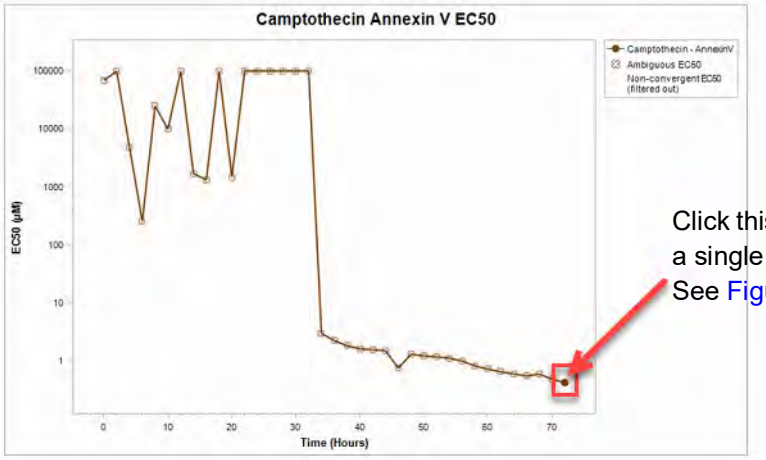
6. Click Graph.

   The Concentration Response Curve is generated and displayed on-screen in its own window. An rSq value and an EC_{50} value are generated and displayed for the graph. A menu (File, Edit, View) is displayed at the top of the graph window and a default title is displayed above the graph.

   - For a sample kinetic EC_{50} graph, see Figure 2-16 on page 156.
   - For a sample single time point EC_{50} graph, see Figure 2-17 on page 156.

Figure 2-17 on page 156 was generated using the above mentioned shortcut by clicking the single time point indicated in Figure 2-16 on page 156.

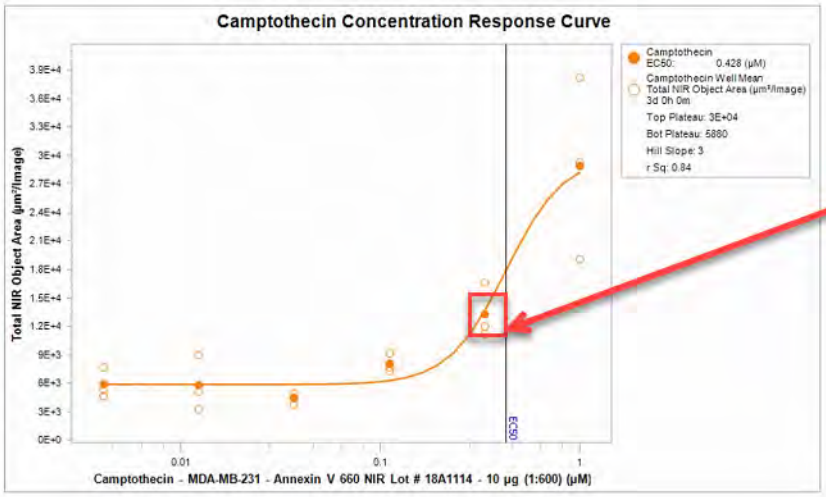
Figure 2-16: Kinetic EC_{50} graph



Click this time point to generate a single time point EC_{50} graph. See Figure 2-17 below.

In a kinetic EC_{50} graph, ambiguous data is marked with an open "x." Non-convergent data is filtered and is not displayed on the graph.

Figure 2-17: Single time point EC_{50} graph



Click any time point to open the Vessel View window and view images for the time point.

7. Optionally, after the concentration response curve is generated:
   - If the response does not have the expected fit, then based on the data expectations, you can apply constraints to modify the fit, and then generate the graph again.
   - You can customize the graph display and export the graph data. See:
     - "Customizing a Time Plot Graph, Histogram, or a Concentration Response Curve" on page 157.
     - "Exporting Graph Data" on page 162.

Customizing a Time Plot Graph, Histogram, or a Concentration Response Curve

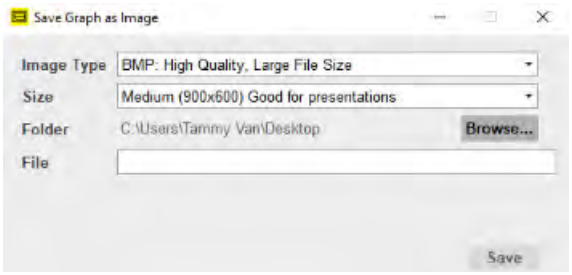
Optionally, after you generate a time plot graph, a histogram, or a concentration response curve for your analysis data, options are available for working with the graph, customizing the graph display and/or exporting the graph data. See:

- “To save the graph as an image file and/or print the graph” below.
- “To edit the default graph display” on page 157.
- “To save graph as an image to a third-party application or the raw data to a document” on page 158.
- “To customize the graph display (Non-default properties)” on page 159.

To save the graph as an image file and/or print the graph

1. On the Graph window menu, click File.
2. Under File, click one of the following:
   - Save as Image. Opens the Save Graph as Image dialog box. You use the options on this dialog box to save the displayed graph as image in a specific format (BMP, JPEG, PNG, or TIFF) and with a specific size. To save the file, you must select the location in which to save the image file, and you must name the file.

Figure 2-18: Save Graph as Image dialog box



   - Print. Opens the standard Windows printing dialog box for printing the graph.

To edit the default graph display

As you edit the default display for the graph in the Graph window, the graph is dynamically updated based on the edits that you make.

To customize the non-default properties for a graph, see “To customize the graph display (Non-default properties)” on page 159.

1. On the Graph window menu, click View.

2. Under View, use any of the following options to edit the default graph display:

Option	Description
Time Plot graph	
Error Bars	• No Error Bars: By default, errors bars are displayed on a metric graph. Select No Error Bars to remove the error bars from the graph display.
	• Standard Error/Standard Deviation: If Error Bars are turned on, then by default, the standard error is displayed for the error bars. Optionally, you can elect to display the standard deviation instead.
	• Full Bars/Top Bars Only/Bottom Bars Only: If Error Bars are turned on, then by default, full bars are displayed. Optionally, you can elect to display only top bars or only bottom bars.
Logarithmic Y-axis	Use a base-10 log scale for the Y axis.
X Axis Display	• Hours - If the range of scan times is less than or equal to 96 hours, then, by default, the time is displayed in hours.
	• Days - If the range of scan times is greater than 96 hours, then, by default, the time is displayed in days.
	• Date/Time - Select this option to display both the date and the time for the range of scan times.
Histogram	
Y Axis Display	• Count: The default value. The total number of objects that are contained within a bin. • Percent: The percentage of all objects that are contained within a bin.
Concentration Response	
Constraints	Opens the Constraints dialog box, which displays the specified constraints (limits) for the Top plateau, the Bottom plateau and the Hill Slope as you have defined in the Graph Metrics window.
Error Bars	See Error Bars above.
Well Mean Points	Selected by default. Clear this option to remove the individual well means from the graph view.
Logarithmic Y-axis	See Logarithmic Y-axis above.

To save graph as an image to a third-party application or the raw data to a document

If you are intending to use the graph in a presentation or a report, then you can save the graph as an image to a third-party application such as Microsoft Word or PowerPoint. If you are archiving the raw data outside Incucyte, or carrying out a statistical analysis of the raw data outside Incucyte, then you can save the raw data to a third-party application such as Microsoft Excel.

1. Make sure that the correct third-party application is open.
2. On the Graph window menu, click View.
3. Under View, select Drag and Drop, and then select one of the following options:
   - Image to Document. (The default value.)
   - Raw Data to Document.

4. Right-click the image, and then while holding down the right mouse key, drag the image into the open document.

   A context menu opens with the following options: Move Here and Copy Here.

5. On the context menu, select the appropriate option.

   - Move Here. A one-time “copy” of the selected image/data is placed in to the opened document. To copy the image again, you must repeat Step 3 through Step 5.
   - Copy Here. The information is copied to your client clipboard, which allows for pasting the copied information an unlimited number of times in to one or more open documents.

To customize the graph display (Non-default properties)

You use the Graph Customization tool to customize the non-default properties of the graph display.

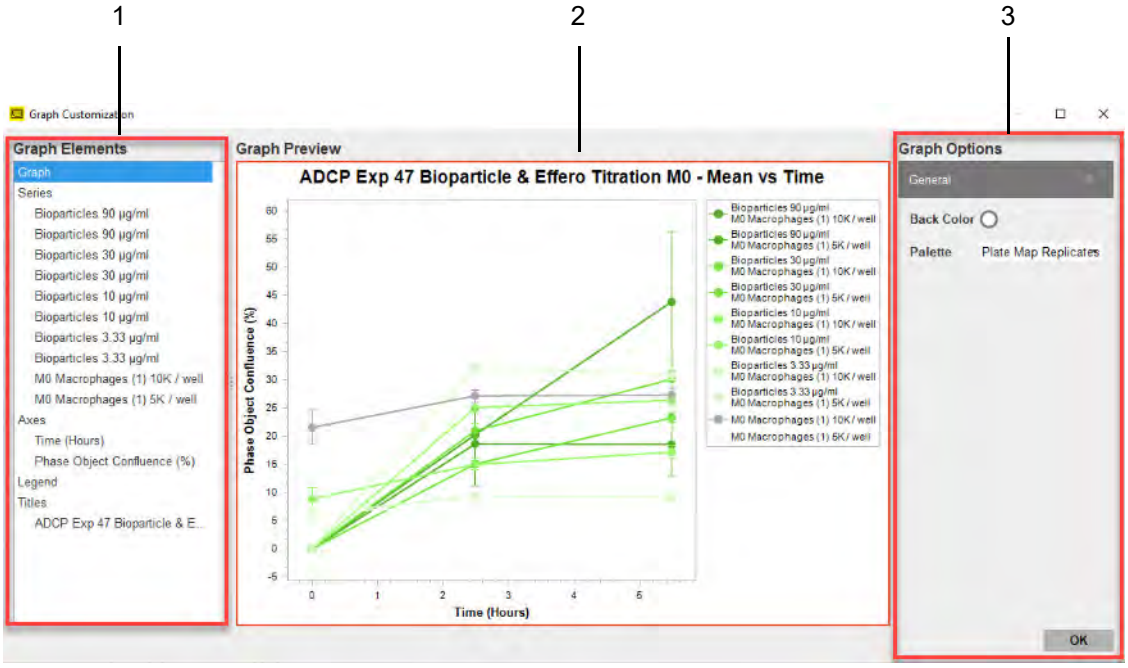
To customize the default properties for a graph, see “To edit the default graph display” on page 157.

1. On the Graph window menu, click Tools.
2. Under Tools, click Customize Graph.

   The Graph Customization dialog box opens.

You can also right-click the graph, and on the context menu that opens, click Customize Graph.

Figure 2-19: Graph Customization dialog box, Time Plot graph shown



The Graph Customization dialog box has the following layout:

	Window component	Description
1	Graph Elements pane	Displays all the graph elements for which you can customize the properties.
2	Graph Preview pane	Displays the graph and the graph legend according to the values that are currently set for its elements.
3	Graph Options pane	Displays the values that you can edit when customizing a selected graph element. As you customize a graph element, the graph and graph legend display are dynamically updated in the Graph Preview pane.

3. For each graph element that you are customizing, do the following:
   - In the Graph Elements pane, select the element that you are customizing.

     The Graph Options pane displays the values that you can edit for the selected graph element.
   - In the Graph Options pane, edit the values for the selected element accordingly.
4. After you have edited all the necessary graph elements, click OK.

   The Graph Customization dialog box closes. The Graph window remain open and the display is dynamically updated according to the edits that you made.

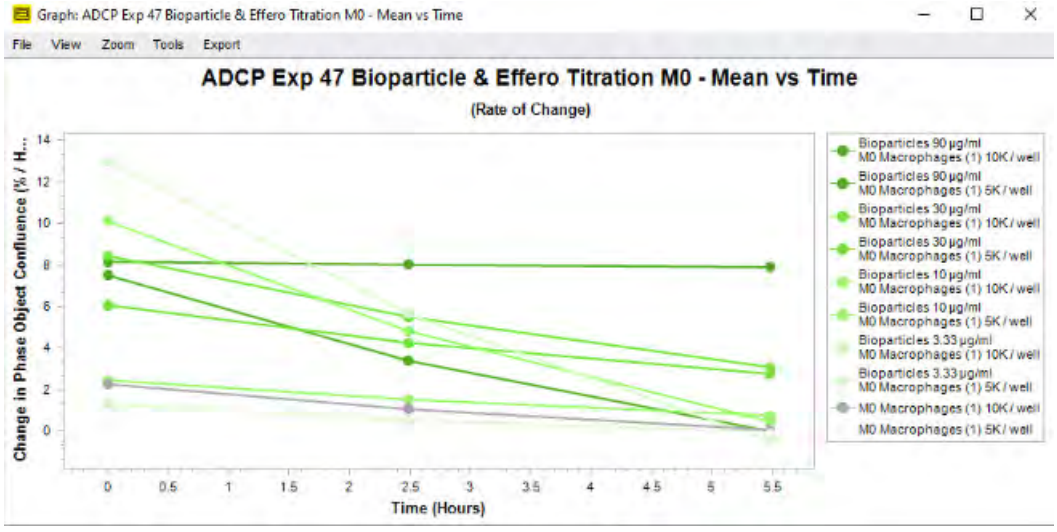
To generate a Rate of Change graph

After you generate a time plot graph, you have the option of use the Rate of Change tool to generate a corresponding Rate of Change graph, which provides an estimated rate of change for each point on the graph.

1. On the Graph window menu, click Tools.
2. Under Tools, click Estimate Rate of Change.

   The Rate of Change graph is automatically generated.

Figure 2-20: Rate of Change graph



3. Optionally, after the Rate of Change graph is generated, you can customize the graph display and export the graph data. See:
   - “Customizing a Time Plot Graph, Histogram, or a Concentration Response Curve” on page 157.
   - “Exporting Graph Data” on page 162.

Exporting Graph Data

You can export the data that was used to generate a graph to either a text file or your client clipboard.

- If you export the graph data to a text file, then you can use any text editor such as Notepad to open the file and work with the data in the file.
- If you export the graph data to your client clipboard, then you have the option of archiving your data and/or carrying out additional statistical analysis of your data outside the Incucyte software. For example, you can import the data in to a third-party spreadsheet program such as Microsoft Excel for analysis.

When you export the graph data, by default, each scan is exported as a single row and all the rows are contained in a single table for any Grouping option other than None. To customize the layout of the exported data, you must select None as the Grouping option.

Remember, you can acquire one or more images per vessel location. Because the default value is to export multiple images as a single data point, then you must customize the layout of the exported data to export every image as an individual data point.

To export graph data for any grouping option other than None

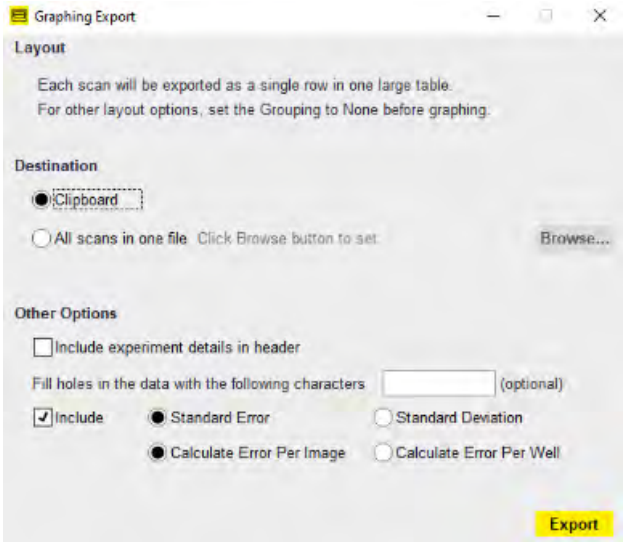
1. Do Step 2 through Step 5 of "To graph the analysis results for any vessel type (Single metric)" on page 146.

For Step 5, make sure that you select any option other than None for the Grouping option.

2. On the Graph Metrics window, click Export Data.

   The Graphing Export dialog box opens.

Figure 2-21: Graphing Export dialog box



3. To select the export destination, do one of the following:
   - To export the data to your client's clipboard, leave the default value of Clipboard selected.
   - To export the data to a single file:
     a. Select All scans in one file.
     b. Click Browse to open the Export Metrics As dialog box, and then browse to and select the destination folder.
     c. Name the file that you are exporting. (The file type is text and you cannot change this.)
     d. Click Save.
4. Optionally, under Other Options, indicate how you are modifying the data before exporting it.
5. Click Export.

To export graph data for the None grouping option

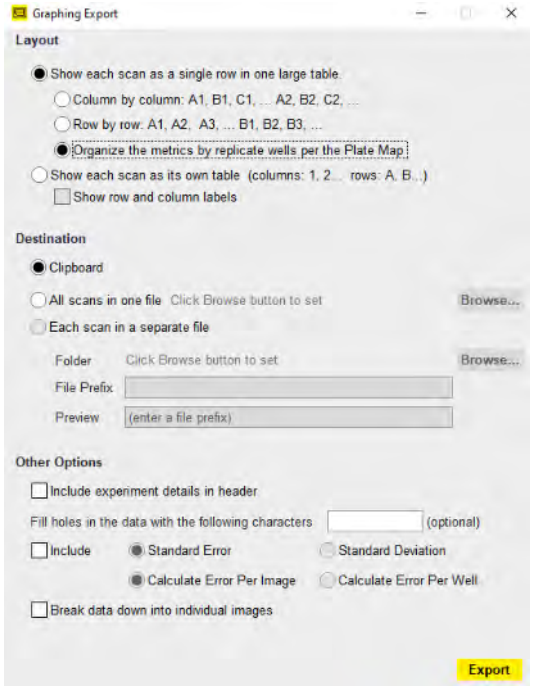
1. Do Step 2 through Step 5 of "To graph the analysis results for any vessel type (Single metric)" on page 146.

For Step 5, make sure that you select None for the Grouping option.

2. On the Graph Metrics window, click Export Data.

   The Graphing Export dialog box opens.

Figure 2-22: Graphing Export dialog box - Slide #18 note



3. Under Layout, select the layout for the exported data. Note the following about the layout options:
   - By default, Show each scan as a single row in a single table is selected.
   - If the vessel is a microplate for which a plate map has been specified, then, by default, Organize the metrics by replicate wells per the Plate Map is selected for the single table scan and this is the recommended value.
   - If you are storing each scan in its own separate file, then you *must* select Show each scan as its own table.
4. To select the export destination, do one of the following:
   - To export the data to your client's clipboard, select Clipboard.
   - To export each scan to its own separate file:
     a. Select Each scan in a separate file.

This option is available only if Show each scan as its own table is selected.

     b. Click Browse to open the Export Metrics As dialog box, and then browse to and select the destination folder.
     c. Name the file that you are exporting. (The file type is text and you cannot change this.)
     d. Go to Step 5.
   - To export the data to a single file:
     a. Leave the default value of All scans in one file selected.
     b. Click Browse to open the Export Metrics As dialog box, and then browse to and select the destination folder.
     c. Name the file that you are exporting. (The file type is text and you cannot change this.)
     d. Go to Step 6.
5. Optionally, in the File Prefix field, enter the prefix that you are adding to each individual file name.

   As you enter the prefix, an example file name is displayed in the Preview field. Each individual file is named according to the following format: <Prefix>_<yyyy>y<mm>m<dd>d<scan time>.txt.
6. Optionally, under Other Options, indicate how to modify the data before it is exported.

Remember, you can acquire one or more images per vessel location. By default, multiple images are exported as a single data point. If you select Break down data into individual images, then every image is exported as an individual data point.

7. Click Export.

Section 4
Supporting Information

Section Contents

Appendix A
Plate Map Editor

To assist in experimental design and data analysis, the Incucyte software includes a Plate Map Editor. You use the Plate Map Editor to define a custom plate map for any microplate-based experiment, ranging from 6-well to 384-well formats. You can define and save a plate map at any point before or during an experiment, or after the experiment has been completed. This appendix details the use of the Plate Map Editor for defining a custom plate map.

This appendix covers the following topics:

Appendix A
Plate Map Editor

Opening the Plate Map Editor

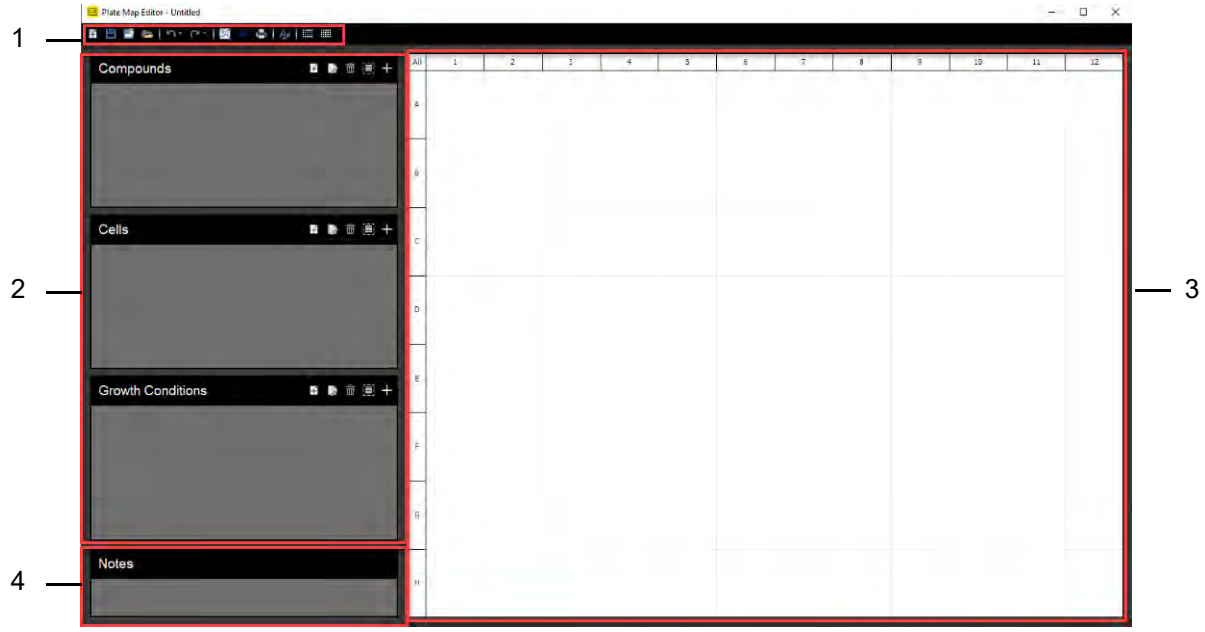
When you install the Incucyte software, the Plate Map Editor is also installed as separate, standalone application. A Plate Map Editor shortcut icon is automatically placed on the client desktop. Although the Plate Map Editor is installed as a standalone application, it is still accessible from within the Incucyte software. As a result, four different access points are available for the Plate Map Editor:

- From the Menu bar: Tools and Options > Plate Map Editor. See “Application menu” on page 21.
- From the Notebook page in the Add Vessel wizard. See “To provide vessel information” on page 50.
- From the Vessel Information window that is opened by clicking the Vessel Information icon on the Visualization toolbar for the Vessel View window. See “The Vessel Information window (via the Vessel View window)” on page 92.
- From the Plate Map Editor desktop icon  .

Continue to “Plate Map Editor Layout” on page 170.

Plate Map Editor Layout

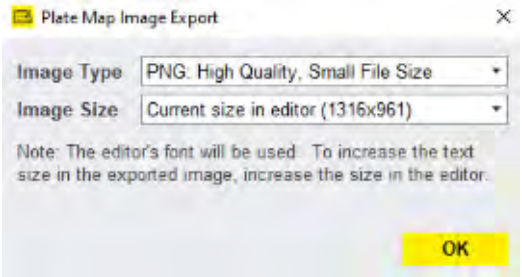
Figure A-1: Plate Map Editor



The Plate Map Editor has the following layout:

	Window component	Description
1	Main menu	Displays the icons that you use to access specific Plate Map Editor functions. See "Plate Map Editor menu bar icons" on page 171.
2	Well Items panes	Before you can specify the experimental settings for any well on a plate, you must first define the appropriate items that can be added to the plate wells: • Compounds: Any treatment to a well that requires either concentration or dilution to be specified. • Cells: Specifies the type, passage, and seeding density for the cells that are present in the plate wells. • Growth Conditions: Any addition such as growth medium where the concentration of a reagent does not vary or is not required to be specified on the plate map. See "Well Items panes icons" on page 172.
3	Microplate pane	Displays the format for the selected microplate, and ultimately, your custom plate map for the microplate. When the Plate Map Editor first opens, by default, the format is set to that of a 96-well plate. You can use the New Plate Map icon to select a different format. You use the pane options to select the wells when defining your custom plate map.
4	Notes pane	Optional. Enter any additional information that is appropriate or required for the plate map.

Plate Map Editor menu bar icons

Icon	Description
	New Plate map icon: Readies the Microplate pane for adding a new plate map with a format other than a 96-well plate.
	Save Plate Map to File icon: Opens the Save Plate Map As dialog box, which provides the options for saving the currently displayed plate map to a file. You must specify the name of the file and the location in which to save the file. The file is saved with the type of .PlateMap and you cannot change this.
	Save As icon: Saves the currently displayed plate map with a different name and/or in a different location. **Tip:** Useful if you simply need to edit an existing plate map to create a new plate map and you do not want to override the existing plate map.
	Open From File icon: Opens the Open Plate Map dialog box in which you can browse to and select an existing plate map file (*.PlateMap) to load in to the Plate Map Editor.
	Undo Last Action icon: Undoes the last action or “n” number of actions that were carried out for the plate map.
	Redo Last Action icon: Enabled only if one or more actions have been undone for the current session of the Plate Map Editor. Redoes the last action or “n” number of actions that were carried out for the plate map.
	Export as Image icon: Opens the Plate Map Image Export dialog box, which provides the options for exporting the currently displayed plate map as a read-only image. You must specify the image type and the image size before you can export the image. *Figure A-2: Plate Map Image Export dialog box*  **Note:** When you export the plate map as an image, the font that is displayed on the plate map is exported as shown (family, size, and style). To change any properties of the font before you export the image, you must use the font editor that is embedded in the Plate Map Editor. See the Set Font for Display, Print, and Export icon.

Icon	Description
	Print Preview icon: Opens a Print Preview dialog box in which you can view what the plate map looks like before it is printed, and make adjustments to the map (such as changing the number of pages across which you are printing the graph) before printing the map.
	Print icon: Opens the standard Windows Print dialog box for printing the currently displayed plate map.
	Set Font for Display, Print, and Export icon: Opens the standard Windows Font dialog box, in which you can adjust the font for any text that is displayed on the plate map. The selected font (family, size, and style) is also the font that is used when printing the plate map or exporting the plate map to an image.
	Keep Current Selection icon: Select the appropriate well or wells, and then click this icon to apply all subsequent actions that you carry out in the Plate Map Editor only to the selected well or wells. **Note:** This icon is a toggle. You can click it as needed to turn it on or off.
	Deselect All Wells icon: Clears all currently selected wells in a single step.

Well Items panes icons

Icon	Description
	Add New <Well Item> icon: Opens the Add/Edit <Well Item> dialog box, in which you can add the indicated new well item (Compound, Cell Type, or Growth Condition).
	Edit Selected <Well Item> icon: Opens the Add/Edit <Well Item> dialog box, in which you can edit the indicated well item (Compound, Cell Type, or Growth Condition).
	Delete Selected <Well Item> icon: Deletes the selected well item from all affected wells in the plate map. **Tip:** This deletion affects only the currently displayed plate map. It does not affect any other plate maps that might have used the same well item.
	Select All Wells Containing <Well Item> icon: Selects all wells in the plate map to which the currently selected well item has been added.
	Add <Well Item> to Plate icon: Enabled only after selecting one or more wells in the plate map. Adds the currently selected well item to all the wells that are selected in the plate map.

Defining a New Plate Map with the Plate Map Editor

The following procedure describes, at a high level, how to define a new plate map with the Plate Map Editor. Note that multiple approaches are appropriate when you use the Plate Map Editor to define a plate map. The following procedure is for example purposes only and is designed solely to provide a frame of reference. You can select whichever approach best suits your working needs.

The following procedure is written for defining a new plate map. To define a plate map by editing an existing map, see "Editing an Existing Plate Map" on page 177.

To define a plate map with the Plate Map Editor

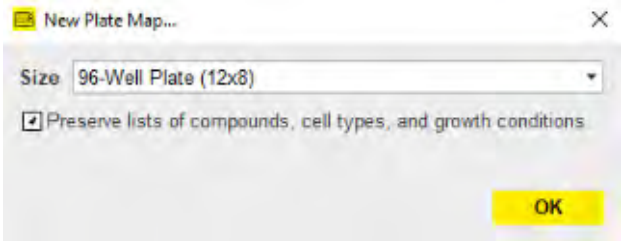
1. Open the Plate Map Editor. See "Opening the Plate Map Editor" on page 169.
2. Do one of the following:
   - If the plate map is for a 96-well plate, then continue to Step 3.

Remember, when the Plate Map Editor first opens, the format is set to that of a 96-well plate

   - If the plate map is a format other than a 96-well plate, then on the main menu, click the New Plate Map icon, and on the New Plate Map dialog box, select a different microplate size, and click OK. Continue to Step 3.

If applicable, to ensure that all previously added well items remain intact, leave Preserve lists of components, cell types, and growth conditions selected.

Figure A-3: New Plate Map dialog box

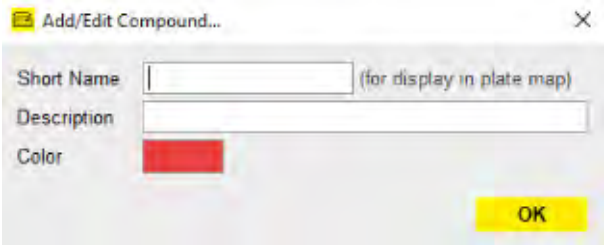


3. Define the well items. To add a well item, do the following:

   a. In the appropriate Well Item pane, click the Create <Well Item> icon. For example, to add a new compound, in the Compounds Well Item pane, click the Add New Compound icon.

      The Add/Edit <Well Item> dialog box opens.

Figure A-4: Add/Edit Compound dialog box



   b. Enter the information for the new well item.

Option	Description
Short Name	Required. A short, descriptive name for the well item, for example, for a new compound, CMP. This is the name that is displayed on the selected wells in the plate map.
Description	Optional. Additional information about the added well item. This information is not displayed on the plate map.
Color	Indicates the color in which to display the wells in the plate map that contain the added well item. You can leave the automatically selected color as-is, or you can click the color icon to open the Choose a Color dialog box and select a different color.

   c. Click OK.

      The Add/Edit <Well Item> dialog box closes. The well item is displayed in the <Well Item> pane.

4. In the Microplate pane, select the wells to which you are adding a well item.

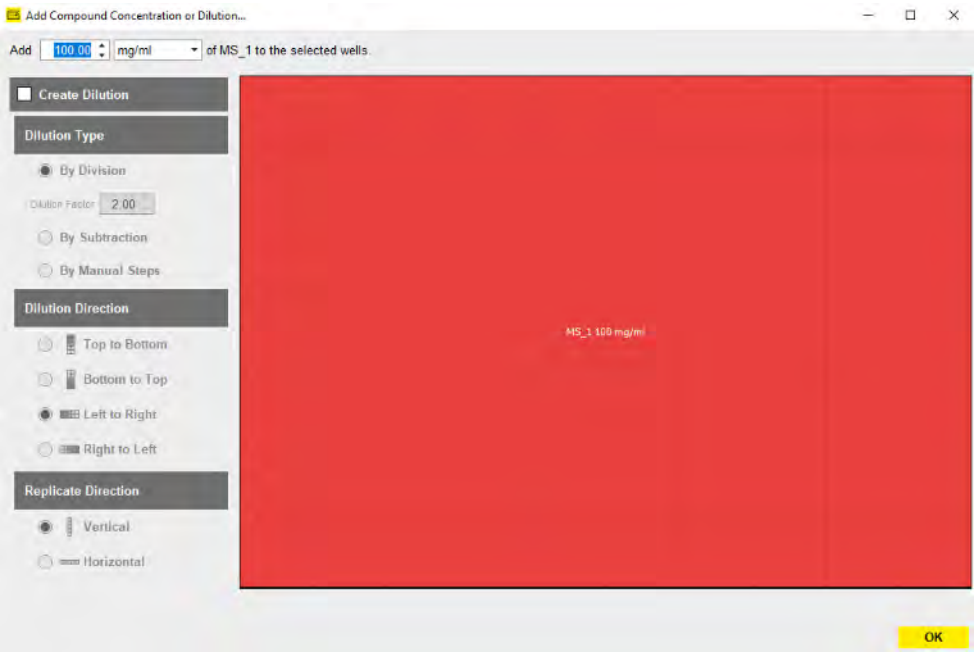
   - To select a single well, click the well. To clear the selection, click the well again.
   - To select multiple contiguous wells, click and hold the left mouse button, and then drag the mouse pointer over the appropriate wells. A box is formed around the wells as you select them. To clear all the wells in the selection, repeat this step. To clear individual wells in the selected area, click the wells.
   - To select all wells in the microplate a single step, in the upper left corner of the Microplate pane, click All. To clear all selected wells, click All again.

To clear all selected wells in a single step, you can also click the Deselect All Wells icon on the main menu for the Plate Map Editor.

   - To select all wells in a single row or column in a single step, click the appropriate row or column header. To clear the selection, click the header again. To clear individual wells in the row or column, click the wells.

5. To add a well item to the selected wells, do the following:
   a. In the appropriate Well Items pane, select the well item.
   b. Click the Add <Well Item> icon. The well item determines the next step.
      - If the well item is a compound, then the Add Compound Or Dilution dialog box opens. (See Figure A-5 below.) Go to Step 6.
      - If the well item is a cell type, then the Adding <Cells> to plate dialog box opens. (See Figure A-6 on page 176.) Go to Step 7.
      - If the well item is a growth condition, then the item is added to the selected wells and the plate map is dynamically updated with the well item and associated information. Go to Step 8.

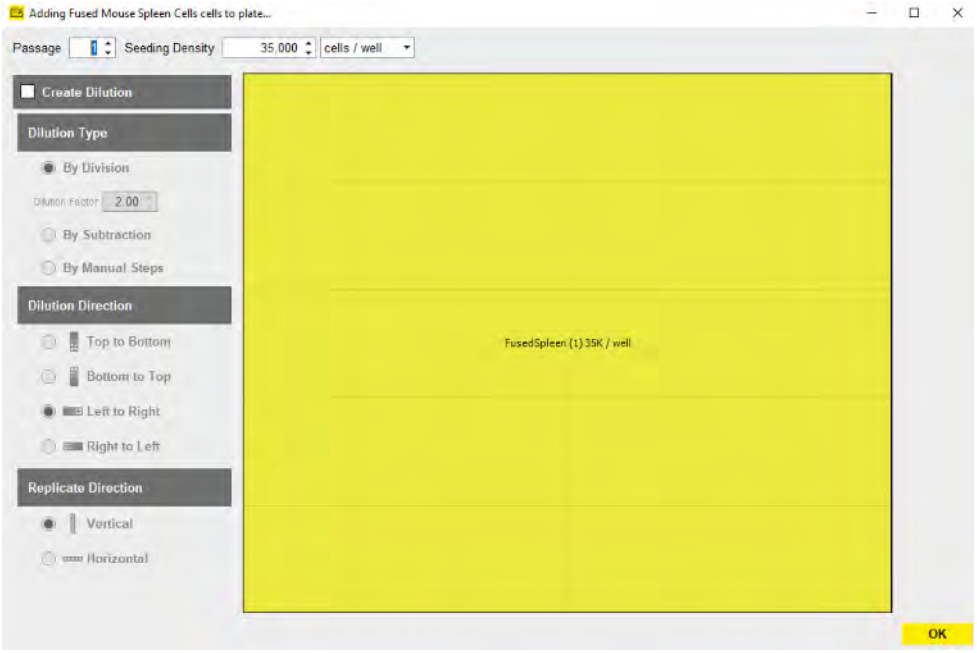
Figure A-5: Add Compound Concentration or Dilution dialog box



6. In the Add Compound Concentration or Dilution dialog box, do the following:
   a. Indicate the concentration of the compound that you are adding to the selected wells.
   b. If the compound is a dilution series, select Create Dilution, and then specify the dilution parameters (Dilution Type, Dilution Direction, and Dilution Replicate Direction).
   c. Click OK to close the dialog box.

      The plate map is dynamically updated with the well item and associated information.
   d. Continue to Step 8.

Figure A-6: Adding <Cells> to plate dialog box



7. In the Adding <Cells> to plate dialog box, do the following:
   a. Indicate the passage and seeding density of the cells that you added to the selected wells.
   b. If the cells are a dilution series, then select Create Dilution, and then specify the appropriate dilution parameters (Dilution Type, Dilution Direction, and Dilution Replicate Direction).
   c. Click OK to close the dialog box.

      The plate map is dynamically updated with the well item and associated information.
   d. Continue to Step 8.
8. Repeat Step 3 through Step 5 as needed to add all the necessary well items and define the plate map.
9. After you have defined the necessary plate map, on the Plate Map main menu, click the Save to File icon to save the plate map.

You must specify the name of the file and the location in which to save the file. The file is saved with the type of .PlateMap and you cannot change this.

Editing an Existing Plate Map

Instead of defining an entirely new plate, you can define a new plate map by editing an existing plate map. When you edit an existing plate map, you have the option of changing the label for a selected well or group of wells, changing the color for a selected well or group of wells, or clearing all the information for a selected well or group of wells, and then defining all the well options (well item, label, and color) again.

To edit an existing plate map

1. On the Plate Map Editor main menu, click the Open from File icon.

   The Open Plate Map dialog box opens.

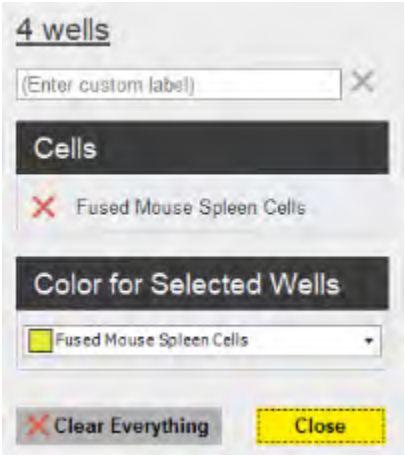
2. In the Open Plate Map dialog box, browse to and select the appropriate plate map file.

   The selected plate map is loaded in to the Plate Map Editor. All the information for the selected plate map (well items, dilutions, concentrations, colors, and so on) are displayed.

3. To edit the plate map, select the well or group of wells that you are editing (see Step 4 of "To define a plate map with the Plate Map Editor" on page 173), and then right-click anywhere in the selection.

   An <n> wells dialog box opens. The dialog box indicates the number of wells that have been selected for editing, the well items that can be edited for the selected wells, and the current color for the selected wells.

Figure A-7: <n> wells dialog box



4. Edit the selected wells, and then click Close.

   You can enter a custom label for the wells, remove one or more items from the selected wells, change the color of the selected wells, or click Clear Everything to clear all current well specifications for the selected wells. The Plate Map Editor display is dynamically updated as you make your edits.

   After you close the dialog box, if you removed one or more well items from the selected wells, or cleared everything, then optionally, you can define new information for the wells, including adding new well items. See "Defining a New Plate Map with the Plate Map Editor" on page 173.

5. After you have defined the necessary plate map, on the Plate Map main menu, click the Save As icon to save the new plate map.

To ensure that you do not override the existing plate map, you must click the Save As icon.

You must specify the name of the file and the location in which to save the file. The file is saved with the type of .PlateMap and you cannot change this.

Section 5
Incucyte Management

Section Contents

Appendix A
Incucyte Management

To facilitate the comprehension of using Incucyte, all the procedures that, at a minimum, are necessary for acquiring scans and viewing and analyzing the scans have been presented in this manual in a specific order. There are, however, additional functions that are available in Incucyte for managing your experiments and data that you can carry out in the order that best suits your working requirements. These functions, such as Experiment Definitions, can help you streamline some of your work in Incucyte while other functions such as Vessel Delete can help you maintain a “clean” working environment by deleting data that has no value or does not need to be archived. All these additional functions are available from the Manage window. This appendix details all the procedures and considerations that are necessary for managing experiment definitions, analysis definitions, and vessels for an Incucyte.

This appendix covers the following topics:

Appendix A
Incucyte Management

Overview of the Incucyte Manage Window

All the Incucyte management functions that are discussed in this section are accessed from the Incucyte Manage window. To open this window, you have two options:

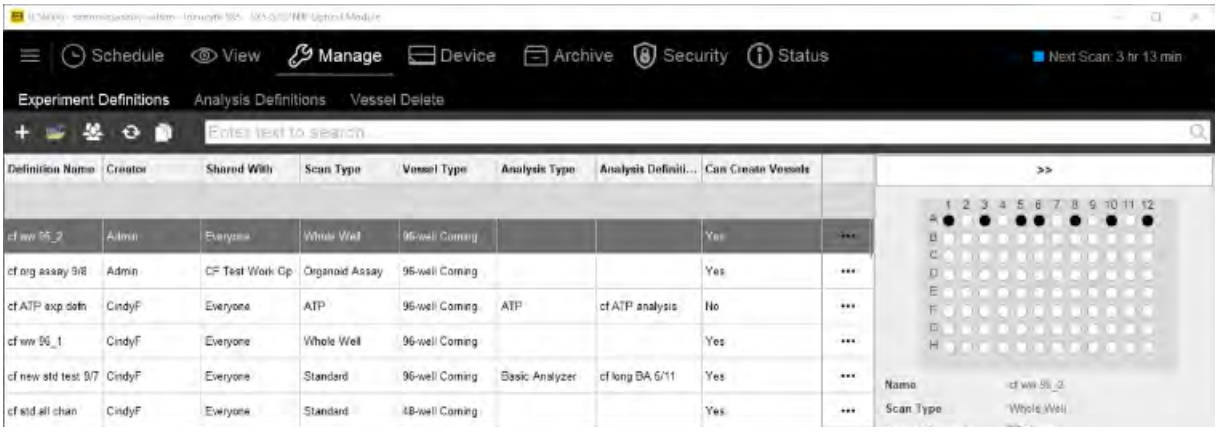
- On the Incucyte main window, click Manage.
- On any Incucyte Window menu, click Manage.

The window has three tabs:

- Experiment Definitions (opened by default). See “Experiment Definitions tab” below.
- Analysis Definitions. See “Analysis Definitions tab” on page 184.
- Vessel Delete. See “Vessel Delete tab” on page 185.

Experiment Definitions tab

Figure A-1: Incucyte Manage window, Experiment Definitions tab



The Experiment Definitions tab displays all the experiment definitions that been created for/imported into the Incucyte and to which you have access based on one of the following criteria being met:

- You are an Admin user.
- The experiment definition has been shared with the **Everyone** user workgroup.
- You are a member of the workgroup with which the definition has been shared.

*If you are not an Admin user, the experiment definition has not been shared with the **Everyone** user workgroup, or you are not a member of the user workgroup with which the experiment definition has been shared, then the definition is not displayed on the Experiment Definitions tab.*

The following columns are displayed for an experiment definition on the Experiment Definitions tab:

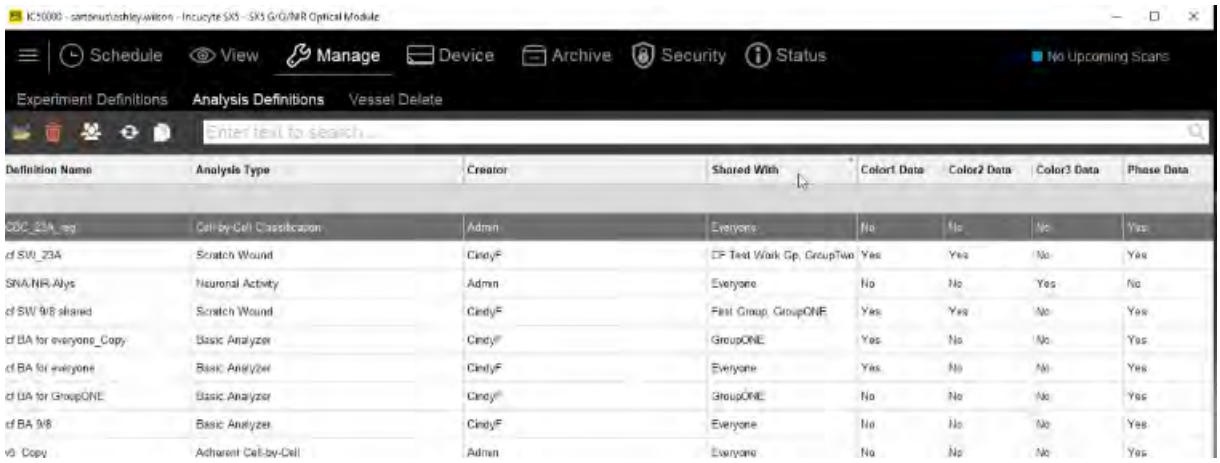
Column	Description
Definition Name	The name of the experiment definition.

Column	Description
Creator	The user who created the experiment definition. Also referred to as the definition owner.
Shared With	Indicates the user workgroups with which the experiment definition has been shared. When you are creating a new definition, or copying and editing an existing definition, you can leave the value set to the default value of **Everyone**, or you can select one or more specific workgroups with which to share the definition.
The next four data columns indicate some of the *template variables* for an experiment definition. See "Managing Experiment Definitions" on page 186.	
Scan Type	The applicable scan type for the definition.
Vessel Type	The applicable vessel type for the definition.
Analysis Type	When you create an experiment definition, you can assign an analysis for the definition. This column indicates the analysis type for the assigned analysis.
Analysis Definition	When you create an experiment definition, you can assign an analysis for the definition.This column identifies the analysis definition for the assigned analysis.
Can Create Vessels	Indicates whether the definition can be used to create a vessel. Values are: • Yes • No **Note:** There are many reasons as to why an existing experiment definition cannot currently be used to create a vessel. For example, a user might have created an experiment definition that specified the Orange phase channel, and then the Orange phase channel capability was subsequently removed from the Incucyte. In another example, your organization might have been beta testing a particular Incucyte module and created an experiment definition based on the module. If your organization chose not to purchase the module, then the module is de-activated for the Incucyte, so you can no longer use the definition.

For detailed information about working with experiment definitions on the Experiment Definitions tab, see "Managing Experiment Definitions" on page 186.

Analysis Definitions tab

Figure A-2: Incucyte Manage window, Analysis Definitions tab



The Analysis Definitions tab displays all the analysis definitions that been created for/imported into the Incucyte and to which you have access based on one of the following criteria being met:

- You are an Admin user.
- The experiment definition has been shared with the **Everyone** user workgroup.
- You are a member of the workgroup with which the definition has been shared.

---

*If you are not an Admin user, the experiment definition has not been shared with the **Everyone** user workgroup, or you are not a member of the user workgroup with which the experiment definition has been shared, then the definition is not displayed on the Experiment Definitions tab.*

For detailed information about working with analysis definitions on the Analysis Definitions tab, see "Managing Analysis Definitions" on page 197.

Vessel Delete tab

Figure A-3: Incucyte Manage window, Vessel Delete tab

The tab indicates the Archived status (Yes or No) for every displayed vessel, with one of the following criteria determining the vessels that are displayed on the tab:

- If you are an Admin user, then *all* the vessels that have ever been scanned on the Incucyte are displayed on the tab, regardless of the user workgroups with which a vessel has been shared.
- If you are not an Admin user, then all the vessels that have been shared with the **Everyone** user workgroup and all the vessels that have been shared with the workgroups that you are member of are displayed on the tab.

---

*If you are not an Admin user, the vessel has not been shared with the **Everyone** user workgroup, or you are not a member of the user workgroup with which the vessel has been shared, then the vessel is not displayed on the Vessel Delete tab.*

As long as the vessel is not in the current scan schedule, then you can delete any vessel that is displayed on the tab at any time, regardless of its archive status.

For detailed information about working with vessels on the Vessel Delete tab, see "Managing Vessels" on page 210.

Managing Experiment Definitions

To carry out an experiment in Incucyte, at a minimum, you must define values for the following experiment variables: the scan type, the scan settings (objective, channel, and other settings), the vessel type, and the vessel scan pattern. Optionally, you can also specify a scan schedule, and if the vessel type is a microplate, then you can also add or import a plate map for the experiment. If multiple experiments use the same or similar values for these variables, then instead of defining variable values repeatedly for each experiment, you can create and apply an *experiment definition*. An experiment definition is a template of these experiment variables and their values. If an experiment uses values that are identical to those in an existing experiment definition, then you can apply the definition as-is. If an experiment uses values that are similar to those in an existing experiment definition, you can copy the definition and edit it, and then apply the definition. Managing experiment definitions consists of:

- Creating new experiment definitions."To create a new experiment definition" below.
- Working with experiment definitions, either from the context menu or the toolbar. See:
  - "To work with an experiment definition (Context menu options)" on page 189
  - "To work with an experiment definition (Toolbar options)" on page 193.

After carrying out any of the following procedures that result in a change in the Experiment Definitions tab display, for example, renaming a definition, you might have to click the Refresh Experiment Definitions icon to correctly update the display on the Experiment Definitions tab.

To create a new experiment definition

To demonstrate all the features for creating a new experiment definition and provide a frame of reference for any approach that you select for creating an experiment definition, the following procedure describes how to create an entirely new scan.

When you create a new experiment definition, you can create only those definitions that the Incucyte is physically capable of running. This caveat, in turn, determines the options that are available in the Add New Experiment Definition wizard. For example, if the Incucyte does not have an Orange image channel, then Orange is not available as a selection for an image channel on the Scan Settings page in the Add New Experiment wizard.

1. Open the Manage window and ensure that the Experiment Definitions tab is the open tab.

   From left to right, the following icons are displayed at the top of the tab.

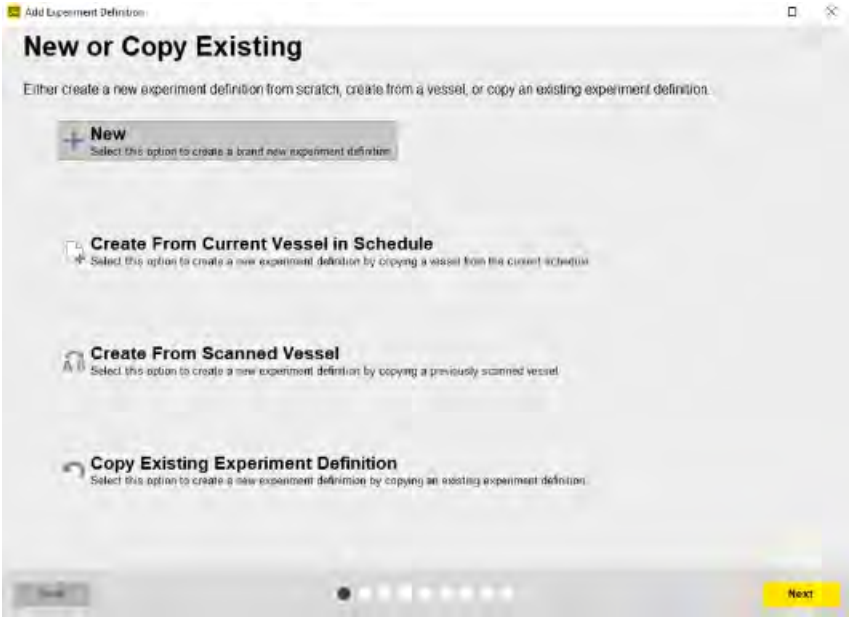
Icon	Description
	Launch Add Experiment Definition wizard icon: Opens the Add Experiment Definition wizard. You use the wizard to create a new experiment definition, create an experiment definition from an existing vessel (a scheduled vessel or a scanned vessel), or copy an existing experiment definition that you can then edit.
	Import Experiment Definition icon: Opens the Import Experiment Definition dialog box. You use the options on this dialog box to import a definition into the Incucyte. See "To work with an experiment definition (Toolbar options)" on page 193.

Icon	Description
	Group by Creator icon: Click this icon to group all the experiment definitions by their respective creators. See "To work with an experiment definition (Toolbar options)" on page 193.
	Refresh Experiment Definitions icon: Click this icon to update the display on the Experiment Definitions tab.
	Copy grid to clipboard: Click this icon to copy the contents that are currently displayed on the Experiment Definitions tab to your client's clipboard. You can now paste this information into a third-party application such as Microsoft Excel for further sorting, evaluation, and so on.

2. Click the Add New Experiment Definition wizard icon.

   The Add New Experiment Definition wizard opens. The New or Copy Existing page is the open page.

Figure A-4: Add Experiment Definition wizard, New or Existing page



Option	Description
New	Selected by default. Select this option to create a new experiment definition.
Create From Current Vessel in Schedule	Select this option to create a new experiment definition by copying a vessel from the current schedule. The scan settings (objective, channels and other settings) and the vessel scan pattern are copied to the new experiment definition.
Create From Scanned Vessel	Select this option to create a new experiment definition by copying a previously scanned vessel. The scan settings (objective, channels and other settings) and the vessel scan pattern are copied to the new experiment definition.
Copy Existing Experiment Definition	Select this option to create a new experiment definition by copying an existing experiment definition and then editing it as appropriate.

3. Ensure that New is selected.
4. Click Next to move through the next six pages in the Add Experiment Definition wizard—Scan Type, Scan Settings, Vessel Selection, Scan Pattern, Experiment Notebook, and Analysis Setup and enter the necessary information. These pages are identical to the pages in the Add Vessel wizard. See:
   - “To select the scan type” on page 42.
   - “To specify the scan settings” on page 43.
   - “To select the vessel type” on page 45.
   - “To set the scan pattern” on page 48.
   - “To provide vessel information” on page 50.

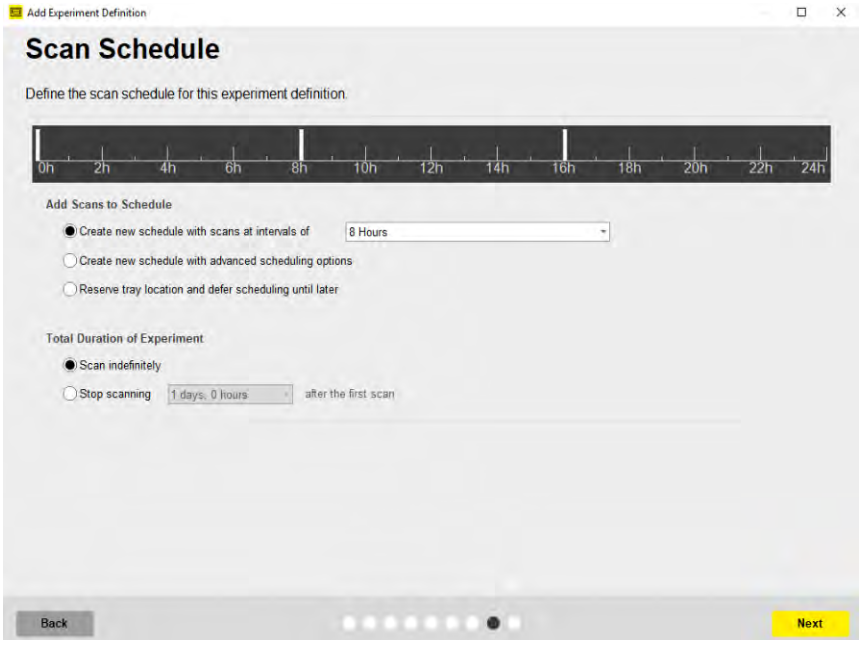
   The Name field on the Experiment Notebook page is specifically for the name of the experiment definition. If you share the definition with one or more user workgroup, then only Admin users and the users who are assigned to the workgroups can view and work with the definition on the Experiment Definitions tab.

   - “To set up the analysis” on page 52.

   After you click Next on the Analysis Setup page, the Scan Schedule page opens. Because you are creating a template and not applying the information to any currently “live” vessel, the page is different than the Scan Schedule page in the Add Vessel wizard.

   - Add Scans to Schedule: Only three options are available: Create new schedule with scans at intervals of, Create new schedule with advanced scheduling options, and Reserve tray location and defer scheduling until later.
   - Total Duration of Experiment: Only two options are available: Scan indefinitely and Stop scanning.

Figure A-5: Add Experiment Definition wizard, Scan Schedule page



5. After you set the scan schedule, click Next.

   The Summary page opens. The page displays all the selections/settings that you have specified for the experiment definition, organized by wizard page. The page functions identically to the Summary page in the Add Vessel wizard, where each section heading corresponds to a wizard page, and is a hyperlink to the indicated page. See "To verify the vessel acquisition settings and schedule the scan" on page 59.

6. When you are satisfied with all the information for the experiment definition, click Finish.

   The Add Experiment Definition wizard closes, and the Manage window, Experiment Definitions tab remains open. The new definition is displayed on the tab.

After an experiment definition is created, the only editing that you can carry out for the definition is changing the workgroups with which the definition is shared or renaming the definition. See "To work with an experiment definition (Context menu options)" below.To "edit" an experiment definition, you must create the definition from an existing vessel and then edit the definition, or copy an existing definition, and then edit the copied definition. See the options listed Step 2 in "To create a new experiment definition" on page 186.

To work with an experiment definition (Context menu options)

The following options for working with an experiment definition are available from the context menu for an experiment definition that is displayed on the Experiment Definitions tab:

- Changing the workgroups with which the experiment definition is shared.
- Renaming an experiment definition.
- Exporting an experiment definition.
- Deleting an experiment definition.

Because an experiment definition is a template, if you create a new vessel using an experiment definition, the definition is now considered to be a part of the vessel information. Changing the definition or deleting the definition has no effect on any experiments that were carried out previously using the "original" definition.

1. Open the Manage window and ensure that the Experiment Definitions tab is the open tab.
2. For the appropriate experiment definition, click the context menu icon (...) that is displayed at the far right of the definition.

   The context menu for the definition opens.

Figure A-6: Manage window, Experiment Definitions tab

3. Continue to one of the following:
   - “To change the workgroups with which an experiment definition is shared” below.
   - “To rename an experiment definition” on page 191.
   - “To export an experiment definition” on page 191.
   - “To delete an experiment definition” on page 192.

To change the workgroups with which an experiment definition is shared

A *user workgroup* defines the users who are able to see an experiment definition and work with the definition. Sharing an experiment definition with a user workgroup is alway optional. To limit the users who can view and work with an experiment definition, you must share the definition with one or more specific user workgroups.

Before you can share an experiment definition with a user workgroup, the User Workgroups setting must be turned on for the instrument. See “Specifying User Settings for an Incucyte” on page 248.

1. On the context menu, click Share with.

   The Edit Shared Workgroups dialog box opens. The dialog box lists all the user workgroups with which you can share the definition.

Before you share an experiment definition with a workgroup, you can view the users that are currently assigned to the workgroup. Click the Show Users icon to update the dialog box display with a list of current users for the selected workgroup.

Figure A-7: Edit Shared Workgroups dialog box

2. Do one or both of the following:
   - To share the definition with a specific user workgroup, click Share for the workgroup. You can select multiple workgroups.

If the appropriate user workgroup is not available, contact your Incucyte administrator.

   - To stop sharing the definition with a workgroup, clear the Share selection for the workgroup.

*If you clear all workgroups with which the definition is shared, then, by default, the definition is shared with the **Everyone** workgroup.*

3. Click Apply.

   The Edit Shared Workgroups dialog box closes. The Experimental Definitions tab remains open.The selected user workgroup is displayed in the Shared with field.

To rename an experiment definition

1. On the context menu, click Rename.

   The Experiment Definition Name dialog box opens. The current name for the definition is selected in the Name field.

Figure A-8: Experiment Definition Name dialog box

2. Edit the name of the definition.
3. Click Rename.

   The Experiment Definition Name dialog box closes. The new name for the definition is now displayed on the Experiment Definitions tab.

To export an experiment definition

The Export option is useful if your organization has two or more Incucyte systems. Instead of setting up the same experiment definition on each of the instruments, you can set up the definition one time on one instrument, and then export this definition so that it is available to import to all your other instruments.

1. On the context menu, click Export.

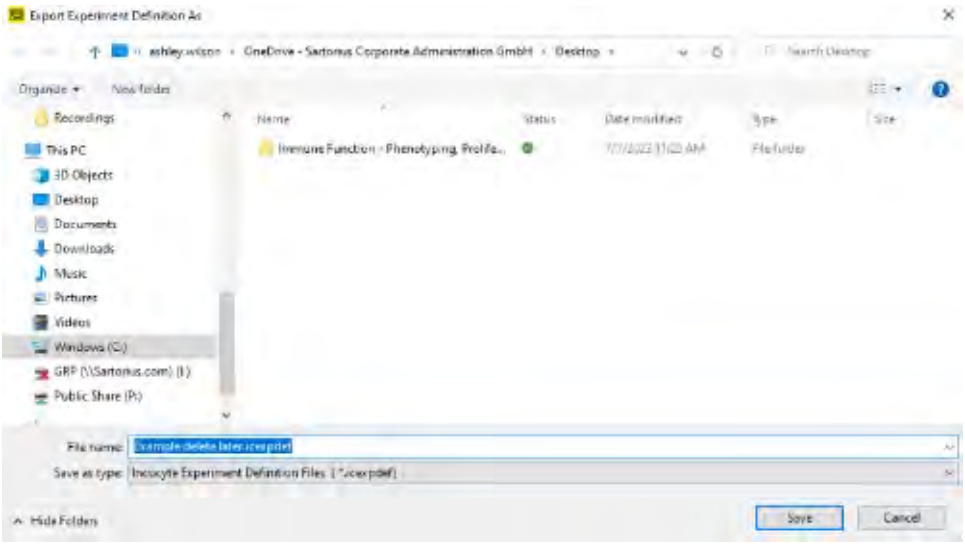
   The Export Experiment Definition dialog box opens. The dialog box shows the name of the selected definition and the current directory path (Filename) to the definition. See Figure A-9 on page 192.

Figure A-9: Export Experiment Definition dialog box

2. Click Browse.

   A second Export Experiment Definition As dialog box opens. The File name field displays the current name of the selected definition. The Save as type field displays Incucyte Experiment Definition field (*.icexpdef) and you cannot edit this.

Figure A-10: Export Experiment Definition As dialog box



3. Optionally, edit the name of the definition that you are exporting.
4. Browse to and select the folder in which to export the definition.
5. Click Save.

   The second Export Experiment Definition dialog box closes. The first Export Experiment Definition dialog box remains open.

6. Click Export.

   The first Export Experiment Definition dialog box closes. The definition is exported to the selected location. The Experiment Definitions tab remains open.

To delete an experiment definition

1. On the context menu, click Delete.

   The Experiment Definition Delete Confirmation dialog box opens, asking you if you are that you want to delete the experiment definition. See Figure A-11 on page 193.

Figure A-11: Experiment Definition Delete Confirmation dialog box

2. Click Yes.

   The Experiment Definition Delete Confirmation dialog box closes. The definition is immediately deleted and is no longer displayed on the Experiment Definitions tab.

To work with an experiment definition (Toolbar options)

The following options for working with an experiment definition are available from the toolbar on the Experiment Definitions tab:

- Importing an experiment definition.
- Organizing the experiment definitions by their creators (owners).

1. Open the Manage window and ensure that the Experiment Definitions tab is the open tab.
2. Continue to one of the following:
   - "To import an experiment definition" below.
   - "To group experiment definitions by creator" on page 195.

To import an experiment definition

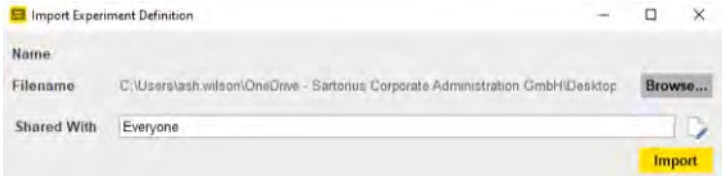
The Import option is useful if your organization has two or more Incucyte systems. If an experiment definition already exists on an Incucyte (Incucyte #1) and you want to use this definition on another Incucyte (Incucyte, #2), then instead of creating the experiment definition on Incucyte #2, you can export the definition on Incucyte #1 to a directory of your choice, and then import this definition to Incucyte #2.

You cannot import an experiment definition that is named the same as an existing definition. You must first rename either the definition that you are importing or the existing definition.

1. On the Experiment Definitions toolbar, click the Import Experiment Definition icon.

   The Import Experiment Definition dialog box opens. When the dialog box first opens, by default, the imported definition is to be shared with the **Everyone** user workgroup.

Figure A-12: Import Experiment Definition dialog box

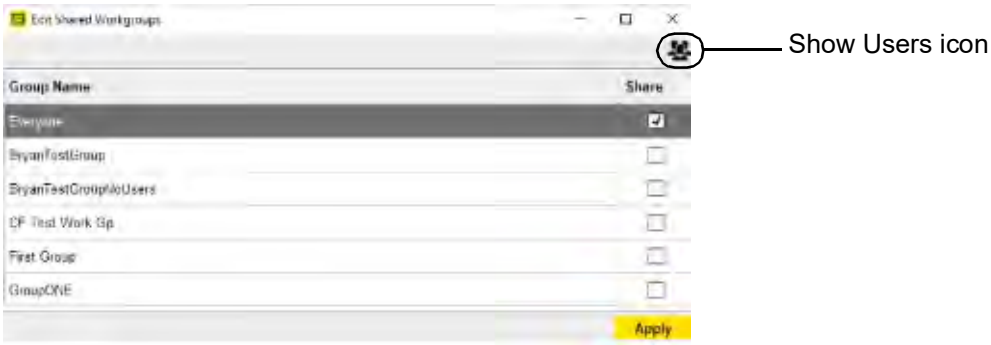


1. If the imported definition is to be shared with the Everyone workgroup, then continue to Step 5; otherwise, to share the imported definition with selected workgroups, continue to Step 2.
2. At the far right of the Shared with field, click the Edit icon .

   The Edit Shared Workgroups dialog box opens. The dialog box lists all the user workgroups with which the definition is already shared and the workgroups that are available for sharing.

Before you share an experiment definition with a workgroup, you can view the users that are currently assigned to the workgroup. Select the workgroup, and then click the Show Users icon to update the dialog box display with a list of users who are currently assigned to the selected workgroup.

Figure A-13: Edit Shared Workgroups dialog box



3. Do one or both of the following:
   - To share an experiment definition with a user workgroup, click Share for the workgroup.

If the appropriate user workgroup is not available, contact your Incucyte administrator.

   - To end the sharing of a definition with a workgroup, clear the Share selection for the workgroup.

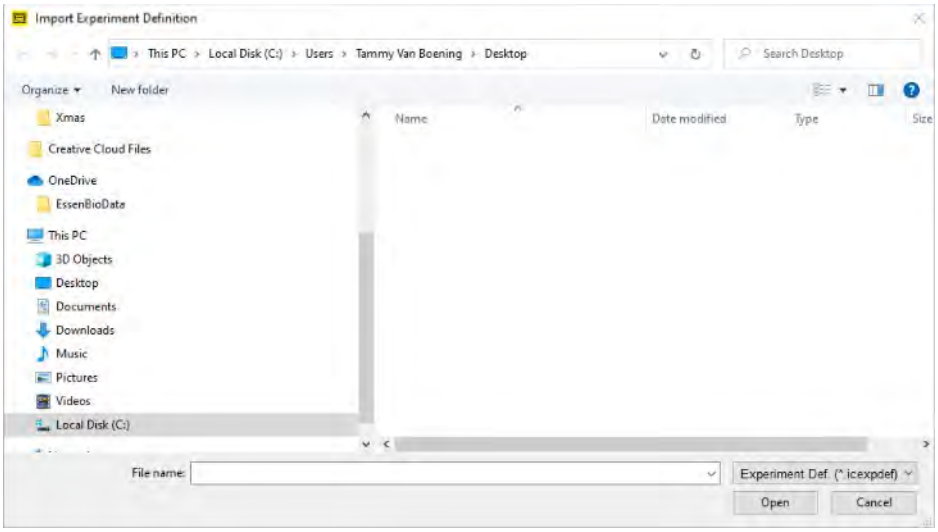
*If you clear all workgroup assignments for a vessel, then, by default, the assignment is automatically reset to **Everyone**.*

4. Click Apply.

   The Edit Shared Workgroups dialog box closes. The Import Experiment Definition dialog box remains open. The changed workgroup information is displayed in the Shared with field.
5. Click Browse.

   A second Import Experiment Definition dialog box opens. See Figure A-14 on page 195.

Figure A-14: Import Experiment Definition dialog box



6. Browse to and select the experiment definition file that you are importing.

   The definition name is displayed in the Filename field.

---

*Remember, an experiment definition file has the proprietary extension of *.icexpdef.*

7. Click Open.

   The second Import Experiment Definition dialog box closes. The first Import Experiment Definition dialog box remains open.

8. Click Import.

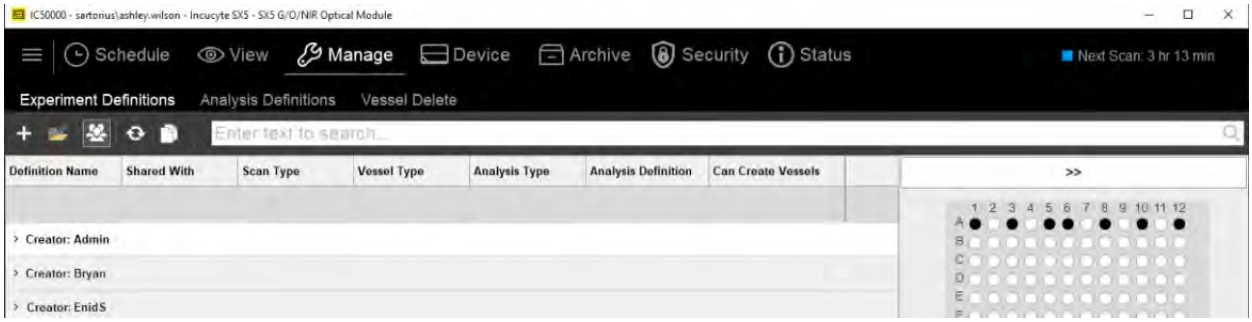
   The experiment definition is imported from the file that you selected in Step 6 into the Incucyte database. The Import Experiment Definition dialog box closes and the imported definition is displayed on the Experiment Definitions tab.

To group experiment definitions by creator

1. On the Experiment Definitions toolbar, click the Group by creator icon.

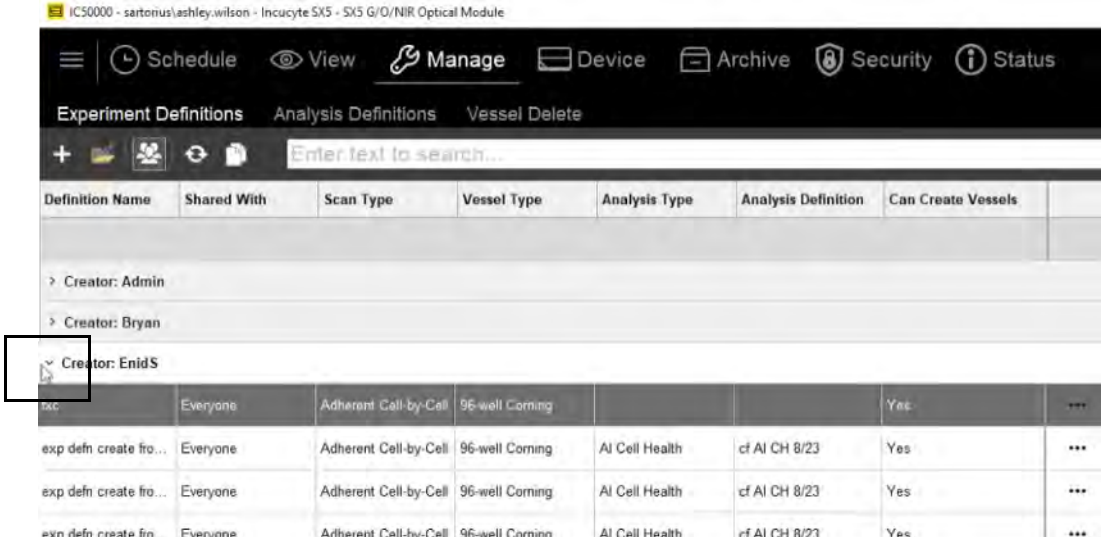
   The display on the Experiment Descriptions tab is updated to show the definitions grouped in individual sub-tabs. The sub-tabs are ordered alphabetically by creator (user) name and are collapsed. See Figure A-15 on page 196.

Figure A-15: Experiment Definitions tab, Grouped by Creator display



2. Optionally, to view all the experiment definitions that were created by a specific user, expand the appropriate creator sub-tab.

Figure A-16: Experiment Definitions tab, Grouped by Creator sub-tab expanded



Managing Analysis Definitions

As discussed in “To set up the analysis” on page 52, an analysis has two parts: an analysis type and an analysis definition. You manage the analyses that have been defined for an Incucyte on the Analysis Definitions tab.

Managing analysis definitions consists of:

- Working with an analysis definition using options on the analysis definition context menu. See “To work with an analysis definition (Context menu options)” below.
- Working with analysis definitions using options on the toolbar of the Analysis Definitions tab. See “To work with an analysis definition (Toolbar options)” below.

After carrying out any of the following procedures that result in a change in the Analysis Definitions tab display, for example, renaming a definition, you might have to click the Refresh Analysis Definitions icon to correctly update the display on the Analysis Definitions tab.

To work with an analysis definition (Context menu options)

The following options for working with an analysis definition are available from the context menu for an analysis definition that is displayed on the Analysis Definitions tab:

- Changing the workgroups with which the analysis definition is shared.
- Editing the analysis definition.
- Renaming an analysis definition.
- Saving the analysis definition as a new definition.
- Saving a protected (read-only) copy of an analysis definition.
- Exporting an analysis definition.
- Deleting an analysis definition.

Because an experiment definition is a template, if you create a new vessel using an experiment definition that has an associated analysis definition, then the resulting analysis is considered to be part of the vessel information. Any changes that you make to the analysis definition, such as renaming the analysis definition, or deleting the analysis definition, have no effect on any experiments that were carried out previously using the “original” analysis definition.

1. Open the Manage window and ensure that the Analysis Definitions tab is the open tab.
2. Right-click the appropriate analysis definition.

   The context menu for the definition opens. See Figure A-17 on page 198.

Figure A-17: Analysis Definition context menu

3. Continue to one of the following:
   - "To change the workgroups with which an analysis definition is shared" below.
   - "To edit an analysis definition" on page 199.
   - "To rename an analysis definition" on page 203.
   - "To save an analysis definition as a new definition" on page 203.
   - "To save a protected copy of an analysis definition" on page 204.
   - "To export an analysis definition" on page 204.
   - "To delete an analysis definition" on page 205.

To change the workgroups with which an analysis definition is shared

A *user workgroup* defines the users who are able to see an analysis definition and work with the definition. Sharing an analysis definition with a user workgroup is alway optional. To limit the users who can view and work with an analysis definition, you must share the definition with one or more specific user workgroups.

Before you can share an analysis definition with a user workgroup, the User Workgroups setting must be turned on for the instrument. See "Specifying User Settings for an Incucyte" on page 248.

1. On the context menu, click Share with.

   The Edit Shared Workgroups dialog box opens. The dialog box lists all the user workgroups with which you can share the definition. See Figure A-18 on page 199.

Before you share an analysis definition with a workgroup, you can view the users that are currently assigned to the workgroup. Click the Show Users icon to update the dialog box display with a list of current users for the selected workgroup.

Figure A-18: Edit Shared Workgroups dialog box

2. Do one or both of the following:
   - To share the definition with a specific user workgroup, click Share for the workgroup. You can select multiple workgroups.

If the appropriate user workgroup is not available, contact your Incucyte administrator.

   - To stop sharing the definition with a workgroup, clear the Share selection for the workgroup.

*If you clear all workgroups with which the definition is shared, then, by default, the definition is shared with the **Everyone** workgroup.*

3. Click Apply.

   The Edit Shared Workgroups dialog box closes and the Manage window, Analysis Definitions tab remains open. The selected user workgroup is displayed in the Shared with field.

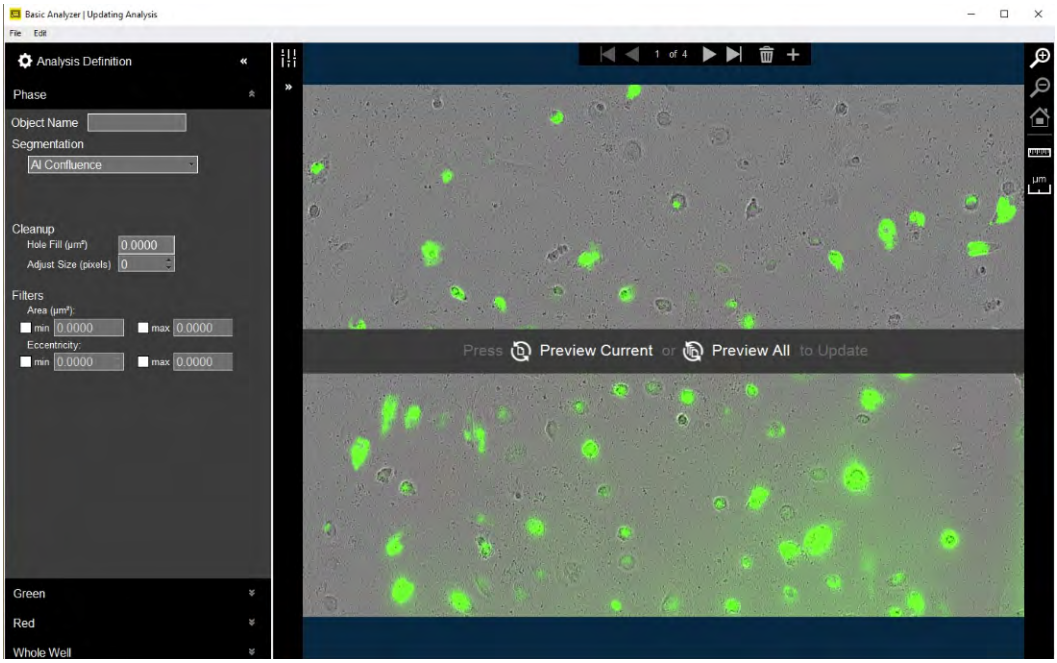
To edit an analysis definition

You have two options for editing an analysis definition: you can add more images to the definition, and/or you can add more metrics and/or normalizations to the definition.

1. On the context menu, click Edit.

   The <Analysis Type> | Updating Analysis window opens. The layout and window options are identical to the Analysis Definition page in the Launch Analysis wizard with one exception: Two menu options, File and Edit, are displayed at the top of the window. See Figure A-19 on page 200.

Figure A-19: <Analysis Type> | Updating Analysis window



If you selected the wrong definition to edit, then click File > Open to open the Analysis Definition Search dialog box and select the correct definition. The Analysis Definition Search dialog box is identical in layout to and has the same features as the Use Analysis Definition page that opens in the Launch Analysis wizard when you are copying an existing analysis definition. The same caveats also determine the analysis definitions that are displayed in the dialog box. See "To use an existing analysis definition" on page 132.

2. Edit the analysis definition as appropriate. You can:
   - Edit the definition parameters, including adding more parameters to the definition, removing parameters from the definition, and/or changing parameter values. See "To define the analysis settings (parameters)" on page 121.
   - Add images. See "To add images when editing an analysis definition" on page 201.
   - Edit the analysis metrics. See "To add metrics and/or normalizations when editing an analysis definition" on page 202.

Although you are not required to do so, Sartorius strongly recommends that you preview any changes that you make to the definition before you save it.

3. After you edit the analysis definition, at minimum, you must save the changes, but other options are also available. All the following options are available on the File menu.

Option	Description
Save	Saves the edits that you made to the definition. The definition is saved with the same name and in the same location.

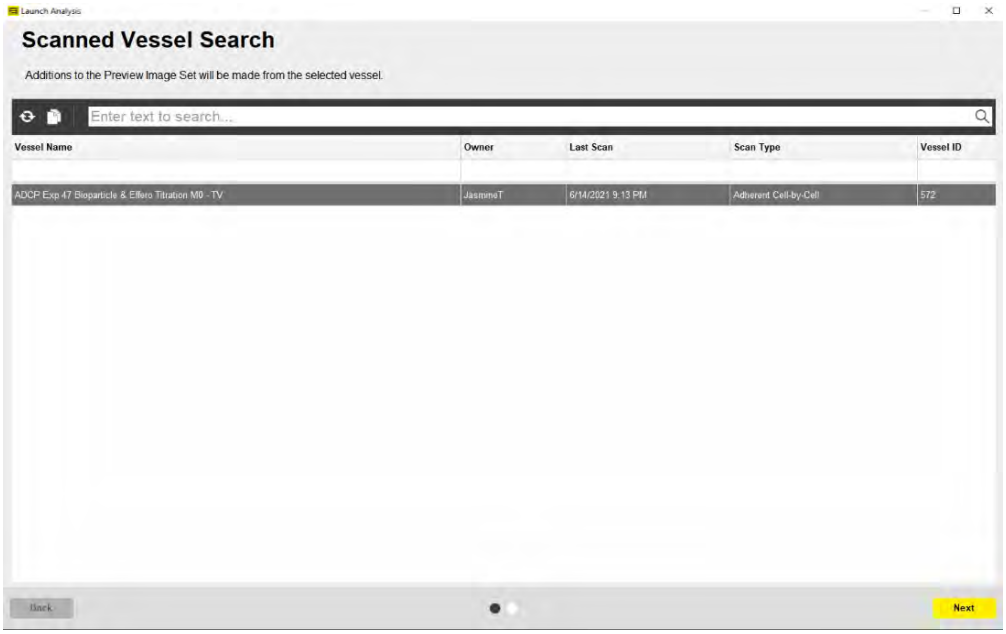
Option	Description
Save As	Save the analysis definition as a new definition (a different name is required) in the same location as the original definition. *Figure A-20: Analysis Definition Name dialog box (Save As)* 
Save Protected Copy	Save the analysis definition as a read-only definition (a different name is required) in the same location as the original definition. *Figure A-21: Analysis Definition Name dialog box (Save Protected Copy)* 

To add images when editing an analysis definition

1. On the Edit menu, click Add Image(s).

   An abbreviated Launch Analysis wizard opens. The Scanned Vessel Search page is the open page. The page lists all the vessels that have been scanned on the Incucyte and to which you have access.

Figure A-22: Scanned Vessel Search page



2. To add more images to the analysis definition, select the appropriate vessel.

The standard Incucyte filter and sort and search functions are available for searching for a specific vessel. See "Working with Data Columns in an Incucyte Window" on page 26.

3. Click Next.

   The Image Set Selection page opens. The page is identical in layout to and has the same functions as Image Set Selection page as the Image Set Selection page that opens in the Launch Analysis wizard when you are creating a new analysis definition. See "To select the images for analysis" on page 120.

4. After you have selected the necessary additional images, click Finish.

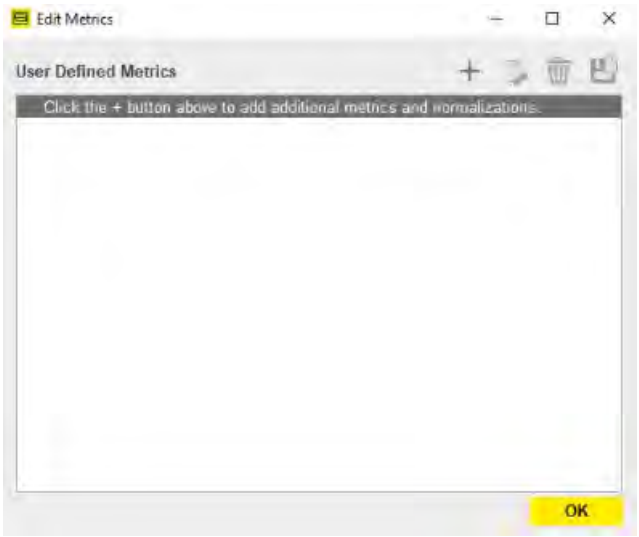
   The Launch Analysis wizard closes. You return to the <Analysis Type> | Updating Analysis window.

To add metrics and/or normalizations when editing an analysis definition

1. On the Edit menu, click Edit Metrics.

   The Edit Metrics dialog box opens. The icons that are displayed at the top of the dialog box are identical in layout and function to the icons that are displayed at the top of the Graph Metrics window. (See "The Graph Metrics Window" on page 138.) When the dialog box first opens, the only active icon is the Create Metric icon.

Figure A-23: Edit Metrics dialog box



2. Click the Create Metric icon.

   The Create Metric dialog box opens. The dialog box contains all the options for defining a new metric for the analysis and it is identical in layout to and has the same features as the Create Metric dialog box that opens from other areas of the Incucyte software. See "To define user metrics" on page 140.

After you define and save your first user metric, the remaining icons on the Create Metric dialog box are enabled.

3. After you create all the necessary metrics for the analysis definition, click OK on the Edit Metrics dialog box.

   The Edit Metrics dialog box closes. You return to the <Analysis Type> | Updating Analysis window.

To rename an analysis definition

1. On the context menu, click Rename.

   The Analysis Definition Name dialog box opens. The current name for the definition is selected in the Name field.

Figure A-24: Analysis Definition Name dialog box

2. Edit the name of the definition.
3. Click Rename.

   The Analysis Definition Name dialog box closes. The new name for the definition is now displayed on the Analysis Definitions tab.

To save an analysis definition as a new definition

The Save As command creates a new analysis definition while preserving the original definition,

1. On the context menu, click Save As.

   The Analysis Definition Name dialog box opens. The current name for the definition is selected in the Name field. The Save As command is displayed on the dialog box.

Figure A-25: Analysis Definition Name dialog box (Save As)

2. Enter a new name for the analysis definition.
3. Click Save.

   The Analysis Definition Name dialog box closes. The new analysis definition is created and saved in the same location as the original definition. The Analysis Definitions tab remains open. An entry for the new definition is displayed on the Analysis Definitions tab.

To save a protected copy of an analysis definition

The Save Protected Copy command creates a read-only version of the definition while preserving the original definition.

1. On the context menu, click Save Protected Copy.

   The Analysis Definition Name dialog box opens. The current name for the definition is selected in the Name field. The Save as Read-Only command is displayed on the dialog box.

Figure A-26: Analysis Definition Name dialog box (Save Protected Copy)

2. Enter a new name for the analysis definition.
3. Click Save As Read-Only.

   The Analysis Definition Name dialog box closes. A read-only copy of the analysis definition is created and saved in the same location as the original definition. The Analysis Definitions tab remains open. An entry for the new definition is displayed on the Analysis Definitions tab.

To export an analysis definition

The Export option is particularly useful if your organization has two or more Incucyte systems. Instead of setting up the same analysis definition on each of the instruments, you can set up the definition one time on one instrument, and then export this definition so that it is available to import to all your other instruments.

1. On the context menu, click Export.

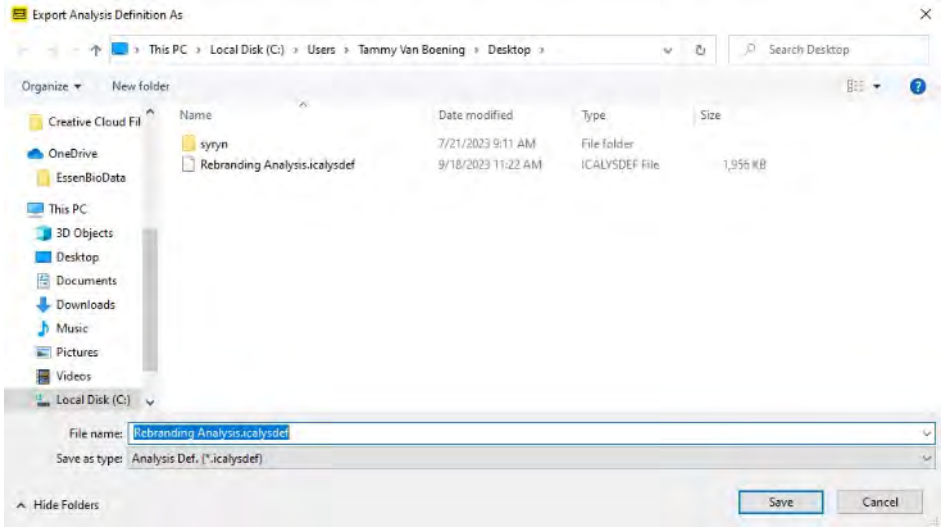
   The Export Analysis Definition dialog box opens. The dialog box shows the name of the selected definition as well as the current directory path (Filename) to the definition.

Figure A-27: Export Analysis Definition dialog box

2. Click Browse.

   A second Export Analysis Definition As dialog box opens. The File name field displays the current name of the selected definition. The Save as type field displays Incucyte Analysis Definition field (*.icalysdef) and you cannot edit this. See Figure A-28 on page 205.

Figure A-28: Export Analysis Definition As dialog box



3. Optionally, edit the name of the definition that you are exporting.
4. Browse to and select the folder in which to export the definition.
5. Click Save.

   The second Export Analysis Definition dialog box closes. The first Export Analysis Definition dialog box remains open.
6. Click Export.

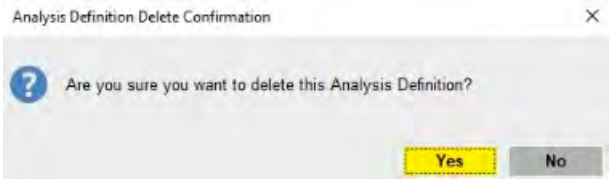
   The first Export Analysis Definition dialog box closes. The definition is exported to the selected location. The Analysis Definitions tab remains open.

To delete an analysis definition

1. On the context menu, click Delete.

   An Analysis Definition Delete Confirmation dialog box opens, asking you if you are sure that you want to delete the definition.

Figure A-29: Analysis Definition Delete Confirmation dialog box



2. Click Yes.

   The Analysis Definition Delete Confirmation dialog box closes, and the definition is deleted. The analysis definition is on longer displayed on the Analysis Definitions tab.

To work with an analysis definition (Toolbar options)

The following options for working with an analysis definition are available from the toolbar on the Analysis Definitions tab:

- Importing an analysis definition.
- Organizing the displayed definitions by their creators (owners).

1. Open the Manage window and ensure that the Analysis Definitions tab is the open tab.

   From left to right, the following icons are displayed at the top of the tab.

Icon	Description
	Import Analysis Definition icon: Opens the Import Analysis Definition dialog box. You use the options on this dialog box to import a definition into the Incucyte.
	Group by Creator icon: Click this icon to group all the analysis definitions by their respective creators.
	Refresh Analysis Definitions icon: Click this icon to update the display on the Analysis Definitions tab.
	Copy grid to clipboard: Click this icon to copy the contents that are currently displayed on the Analysis Definitions tab to your client's clipboard. You can now paste this information into a third-party application such as Microsoft Excel for further sorting, evaluation, and so on.

2. Continue to one of the following procedures:
   - "To import an analysis definition" on page 206.
   - "To group analysis definitions by creator" on page 208.

To import an analysis definition

The Import option is useful if your organization has two or more Incucyte systems. If an analysis definition already exists on an Incucyte (Incucyte #1) and you want to use this definition on another Incucyte (Incucyte, #2), then instead of creating the analysis definition on Incucyte #2, you can export the definition on Incucyte #1 to a directory of your choice, and then import this definition to Incucyte #2.

You cannot import an analysis definition that is named the same as an existing definition. You must rename either the definition that you are importing or the existing definition and then you can carry out the import.

1. On the Analysis Definitions toolbar, click the Import Analysis Definition icon.

   The Import Analysis Definition dialog box opens. See Figure A-30 on page 207.

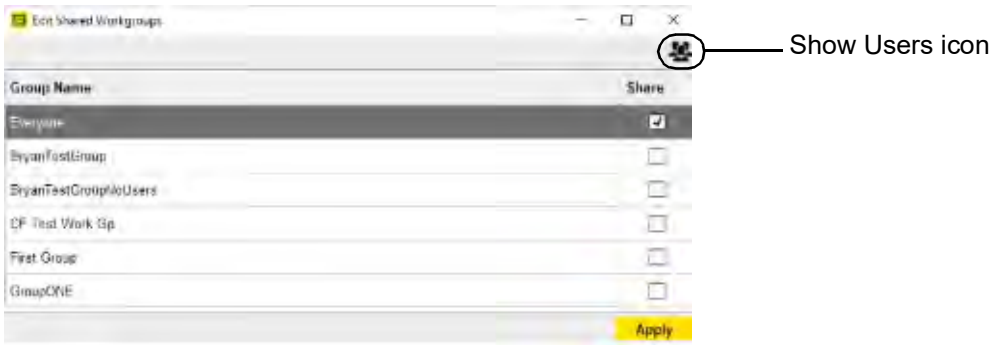
Figure A-30: Import Analysis Definition dialog box

1. If the imported definition is to be shared with the **Everyone** workgroup, then continue to Step 5; otherwise, to share the imported definition with selected workgroups, continue to Step 2.
2. At the far right of the Shared with field, click the Edit icon .

   The Edit Shared Workgroups dialog box opens. The dialog box lists all the user workgroups with which the definition is already shared and the workgroups that are available for sharing.

Before you share an analysis definition with a workgroup, you can view the users that are currently assigned to the workgroup. Select the workgroup, and then click the Show Users icon to update the dialog box display with a list of users who are currently assigned to the selected workgroup.

Figure A-31: Edit Shared Workgroups dialog box



3. Do one or both of the following:
   - To share an experiment definition with a user workgroup, click Share for the workgroup.

If the appropriate user workgroup is not available, contact your Incucyte administrator.

   - To end the sharing of a definition with a workgroup, clear the Share selection for the workgroup.

*If you clear all workgroup assignments for a vessel, then, by default, the assignment is automatically reset to **Everyone**.*

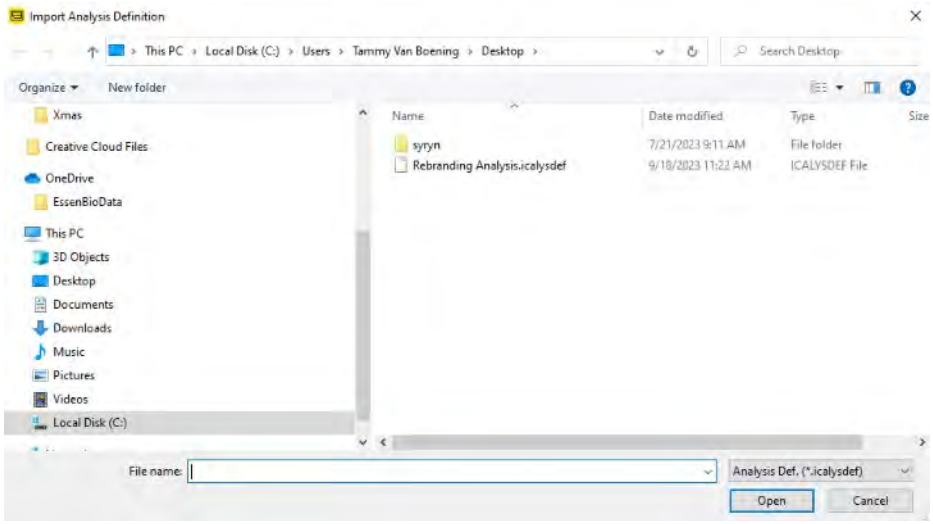
4. Click Apply.

   The Edit Shared Workgroups dialog box closes. The Import Analysis Definition dialog box remains open. The changed workgroup information is displayed in the Shared with field.

5. Click Browse.

   A second Import Analysis Definition dialog box opens.

Figure A-32: Import Experiment Definition dialog box



6. Browse to and select the analysis definition file that you are importing.

   The definition name is displayed in the Filename field.

---

*Remember, an analysis definition file has the proprietary extension of *.icalysdef.*

7. Click Open.

   The second Import Analysis Definition dialog box closes. The first Import Analysis Definition dialog box remains open.

8. Click Import.

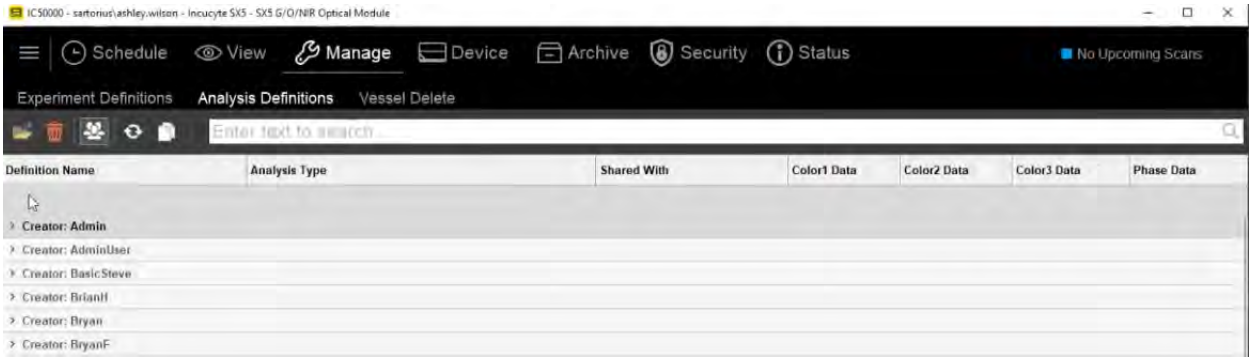
   The selected analysis definition is imported from the file that you specified in Step 6 into the Incucyte database. The Import Analysis Definition dialog box closes and the imported definition is displayed on the Analysis Definitions tab.

To group analysis definitions by creator

1. On the Analysis Definitions toolbar, click the Group by creator icon.

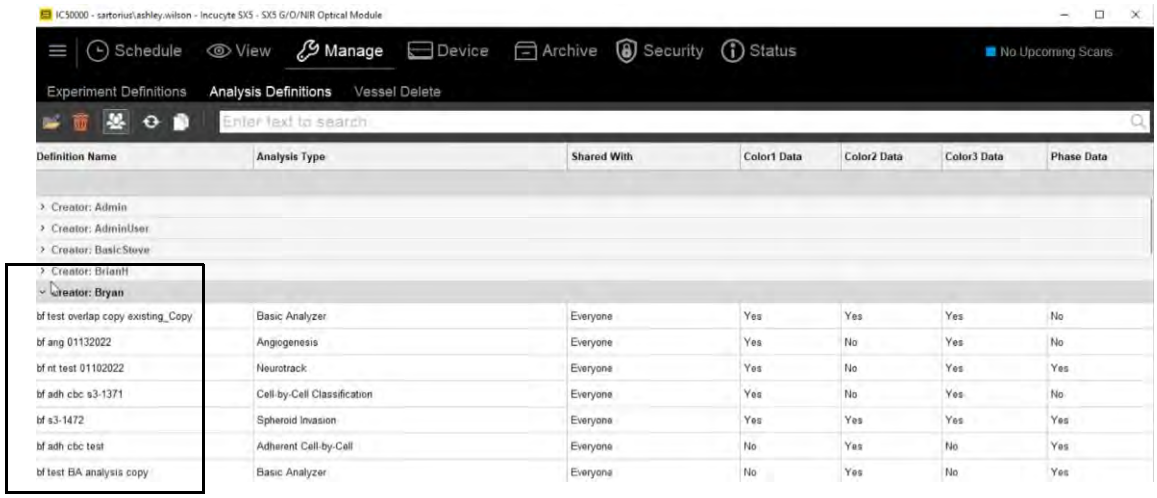
   The display on the Analysis Descriptions tab is updated to show the definitions grouped in individual sub-tabs. The sub-tabs are ordered alphabetically by creator (user) name and are collapsed. See Figure A-33 on page 209.

Figure A-33: Analysis Definitions tab, Grouped by Creator display



2. Optionally, to view all the analysis definitions that were created by a specific user, expand the appropriate creator sub-tab.

Figure A-34: Analysis Definitions tab, Grouped by Creator sub-tab expanded



Managing Vessels

You manage the vessels that have been scanned on an Incucyte on the Vessel Delete tab. To ensure that a user does not inadvertently delete experiment data, the information that is displayed on the tab and the management options that are available to you have been intentionally limited to the minimum that is required for managing the vessels on an Incucyte. Managing vessels consists of the following: Deleting one or more vessels and grouping vessels by owner.

To manage vessels on an Incucyte

1. Open the Manage window and ensure that the Vessel Delete tab is the open tab.

   The tab indicates the Archived status (Yes or No) for every displayed vessel, with one of the following criteria determining the vessels that are displayed on the tab:

   - If you are an Admin user, then *all* the vessels that have ever been scanned on the Incucyte are displayed on the tab, regardless of the user workgroups with which a vessel has been shared.
   - If you are not an Admin user, then all the vessels that have been shared with the **Everyone** user workgroup and all the vessels that have been shared with the workgroups that you are member of are displayed on the tab.

*If you are not an Admin user, the vessel has not been shared with the **Everyone** user workgroup, or you are not a member of the user workgroup with which the vessel has been shared, then the vessel is not displayed on the Vessel Delete tab.*

From left to right, the following icons are displayed at the top of the tab.

Icon	Description
	Delete Vessels icon: After you select one or more vessels for deletion, click this icon to delete the vessels from the Incucyte database.
	Select all rows: Click this icon to select all rows that are currently displayed on the Vessel Delete tab.
	Deselect all rows: Click this icon to clear the selections for all rows that are currently selected on the Vessel Delete tab.
	Group by Creator icon: Click this icon to group all the experiment definitions by their respective creators. See
	Refresh Vessels icon: Click this icon to update the display on the Vessel Delete tab.
	Copy grid to clipboard: Click this icon to copy the contents that are currently displayed on the Experiment Definitions tab to your client's clipboard. You can now paste this information into a third-party application such as Microsoft Excel for further sorting, evaluation, and so on.

2. Continue to one of the following:
   - "To delete a scanned vessel" below.
   - "To group vessels by owner" on page 213.

To delete a scanned vessel

As long as a vessel is not in the current scan schedule, you can delete any vessel that is displayed on the tab for you at any time, regardless of its archive status. If a vessel is in the current scan schedule, then before you can delete the vessel, you must remove the vessel from the scan schedule.

1. Select the vessels that you are deleting. You can do any of the following:
   - For each vessel that you are deleting, click the Select option for the vessel.
   - To select all vessels for deletion in a single step, click the Select all rows icon.

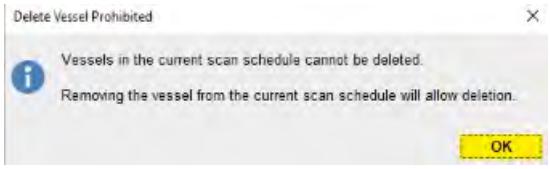
If you decide that one or more vessels should not be selected for deletion, then you can individually clear the Select option for each vessel, or to clear the selection for all vessels in a single step, click the Deselect all rows icon.

2. Click the Delete vessels icon.

   The icon turns red and one of the following happens:

   - If any of the selected vessels is in the current scan schedule, then a Delete Vessel Prohibited dialog box opens. The dialog box indicates that vessels in the current scan schedule cannot be deleted and instructs you to remove the vessels from the current scan schedule before you can continue. After you remove the affected vessels from the scan schedule (see "The Acquisition Window" on page 35), then you can continue with vessel deletion. See "To delete vessels (No vessels in current scan schedule)" on page 212.

Figure A-35: Delete Vessel Prohibited dialog box

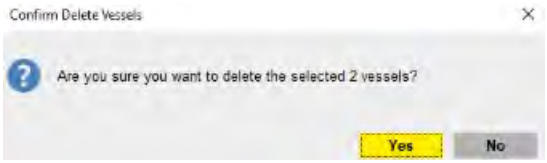


   - If none of the selected vessels are in the current scan schedule, then depending on the archive statuses of the selected vessels, one of three Confirm Vessel delete dialog box opens. Continue "To delete vessels (No vessels in current scan schedule)" on page 212.

To delete vessels (No vessels in current scan schedule)

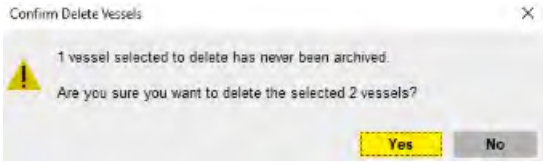
- If the data for all the selected vessels has been archived, then the Confirm Delete Vessels dialog box asks you if you want to continue. Go to Step 3.

Figure A-36: Confirm Vessel Delete dialog box, all vessels have archived data



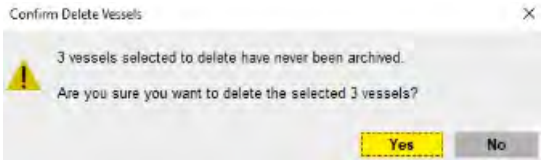
- If the data for some of the vessels, but not all, has been archived, then the Confirm Delete Vessel dialog box indicates the number of vessels for which the data has not been archived and asking you if you want to continue. To continue, go to Step 3; otherwise, go to Step 1.

Figure A-37: Confirm Vessel Delete dialog box, mix of archived and non-archived data



- If none of the data for the selected vessels has been archived, then the Confirm Delete Vessel dialog box indicating this, and asking you if you want to continue. To continue, go to Step 3; otherwise, go to Step 1.

Figure A-38: Confirm Vessel Delete dialog box, mix of archived and non-archived data



1. Click No.

   The Confirm Delete Vessels dialog box closes. The Vessel Delete Tab remains opens.

2. Individually clear the Select option for each vessel that you are not deleting, and or to delete all selected vessels in a single step, click the Deselect all rows icon.

   - If you deselect all vessels, then the procedure is complete for you.
   - If some vessels remain selected for deletion, then Click the Delete vessels icon again and after the Confirm Delete Vessel dialog box opens, continue to Step 3.

3. Click Yes.

   The Vessel Delete Confirmation dialog box closes. The selected vessels are deleted from the Incucyte database and are no longer displayed on the Vessel Delete tab.

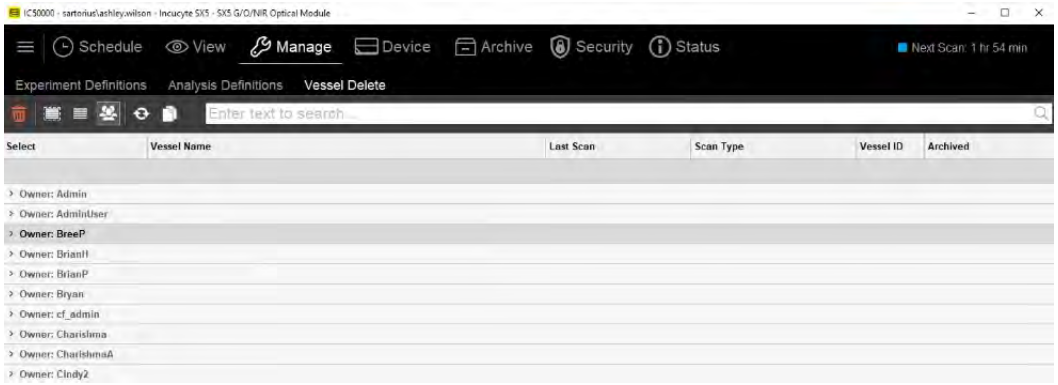
After you delete one or more vessels, you might have to click the Refresh Vessels icon to correctly update the Vessel Delete tab display.

To group vessels by owner

1. On the Vessel Delete toolbar, click the Group by owner icon.

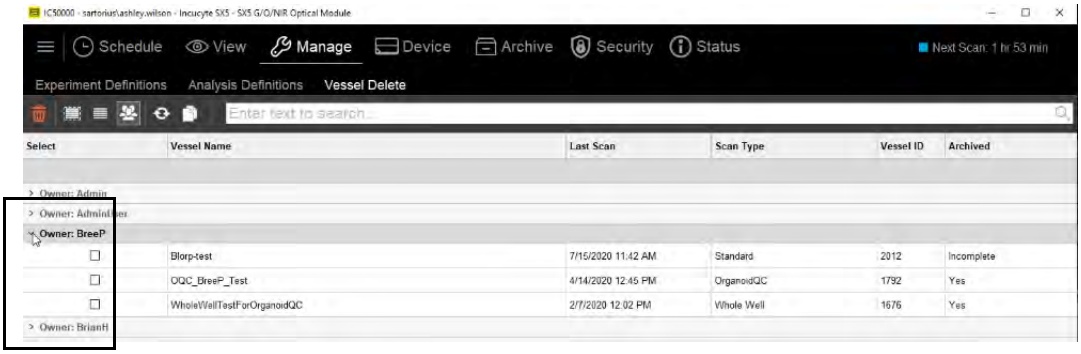
   The display on the Vessel Delete tab is updated to show the vessels grouped in individual sub-tabs. The sub-tabs are ordered alphabetically by owner name and are collapsed.

Figure A-39: Vessel Delete tab, Grouped by owner display



2. Optionally, to view all the vessels that were scanned by a specific owner, expand the appropriate owner sub-tab.

Figure A-40: Vessel Delete tab, Grouped by Owner sub-tab expanded



Appendix A
Incucyte Management

Section 6
Incucyte Administration and Security

Section Contents

Appendix A
Incucyte Data Archives

Vessel image data, plate maps, analyses, and all associated vessel data are stored on the Incucyte controller. Although the associated hard drive is approximately 16 TB, eventually, you will have to clear space to continue acquiring and processing vessel data. To clear space while still maintaining historical records of all your vessel data, you can archive your data. With the Archive data management function that is available in Incucyte, you can save historical vessel data to a location other than the hard drive on the Incucyte controller. After you have archived this information, you can then delete the vessel and its associated data from the controller's hard drive, which frees up the space that you need to continue acquiring and processing new vessel data. This appendix details Incucyte's Archive function.

This appendix covers the following topics:

Appendix A
Incucyte Data Archives

Archiving Overview

An archive contains vessel images and their associated analyses. A single archive can consist of images and the associated analyses for one or more vessels. When you archive vessel data, you must specify the location in which to store the archive. Two locations are available for archiving data in the Incucyte system: My Computer and Storage Attached to Incucyte Controller.

- My Computer: If you select “My Computer” as your archiving location, then you can save the archives either to a local drive on the Incucyte client, or to a network drive. If continuous access to the archives is anticipated, then local archives are especially advantageous for you; however, you must be aware of the following caveats:
  - Archiving to My Computer can be significantly slower than archiving to Storage Attached to Incucyte Controller.
  - Archiving to My Computer is more susceptible to interruptions (for example, system reboots) than archiving to Storage Attached to Incucyte Controller.
  - Archiving to My Computer requires that your Incucyte client be on for the duration of the archiving process.
  - Archiving to My Computer is, overall, a generally less efficient process than archiving to Storage Attached to Incucyte Controller.
- Storage Attached to Incucyte Controller: If you select “Storage Attached to Incucyte Controller” as your archiving location, then the archives are stored on a dedicated storage device that is directly attached to the Incucyte controller either through an eSATA (the fastest and preferred method) or through a USB port. Incucyte-attached archives are immune to network failures, computer reboots, or logoffs; however, you can access Incucyte-attached archives only if you remove the storage from the Incucyte controller and connect it directly to a local computer. As a result, if continuous access to the archives is anticipated, then you should archive to My Computer.

An optimized Incucyte Archive drive is available for purchase from Sartorius.

Because archiving is for historical data management purposes, immediate access to the data is typically not a priority. As a result, if you select “Storage Attached to Incucyte Controller” as your archiving location, then note the following caveats:

- You cannot open the archives while the storage device is attached to the Incucyte controller.
- Sartorius recommends that you detach and reattach the external storage device only when absolutely necessary. Ideally, the storage device should remain connected until it is completely full.
- If you must detach the storage device, then before doing so, you must open the Status page and verify that archives are not currently in the queue or are being processed.
- You must not power off or unplug the storage device during the archiving process.

Appendix A
Incucyte Data Archives

Although regular archiving is important for data management, before you archive any vessel data, note the following caveats:

- Although you can analyze image data that is contained within an archive, Sartorius strongly recommends that you do *not* archive the data for vessels that are being actively evaluated. Instead, the vessels should remain on the controller until all evaluation is complete and only then should you archive the data.
- After you archive the data for a vessel, and then delete the vessel from the Incucyte controller, you cannot retrieve the vessel.
- Because of the volume of data that can be involved (the number of scans, for example, scans every hour, the number of channels scanned, the associated analysis, for example, Basic or Angiogenesis, and so on), archives are more likely to be written successfully when the archive size is kept small. Moreover, not only are smaller archives more easily transported and shared, but also they are less likely to be interrupted by system issues (system reboots, logoffs, Windows updates and so on). As a result, Sartorius recommends that you archive data on at least on a monthly basis, if not more frequently.

Creating a New Archive

You use the Archive function to create a new vessel archive. You can archive any vessel that has ever been scanned by any user on your Incucyte instrument. You are not limited to archiving the vessels that only you own. When you create an Incucyte archive, it runs as an independent task, which means that, if applicable, you can exit the Incucyte software during the archiving process.

If you elect to archive to My Computer, you can still exit the Incucyte software, but remember, the Incucyte client must remain on for the duration of the archiving process.

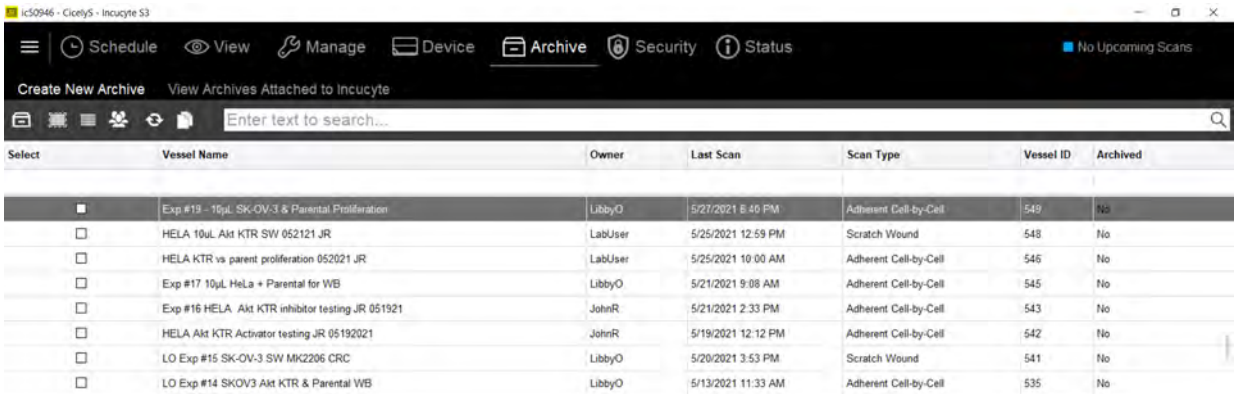
Because of the volume of data that is involved in an archive, before you create a new archive, you should review the vessels that you are archiving and their associated analyses, and delete any unnecessary data. See "The Vessel View Window" on page 89.

To create a new archive

1. On the Incucyte menu, click Archive.

   The Archive window opens. By default, the Create New Archive tab is open. The window displays all the vessels that have ever been scanned by any user on your Incucyte instrument.

Figure A-1: Archive window, Create New Archive selected



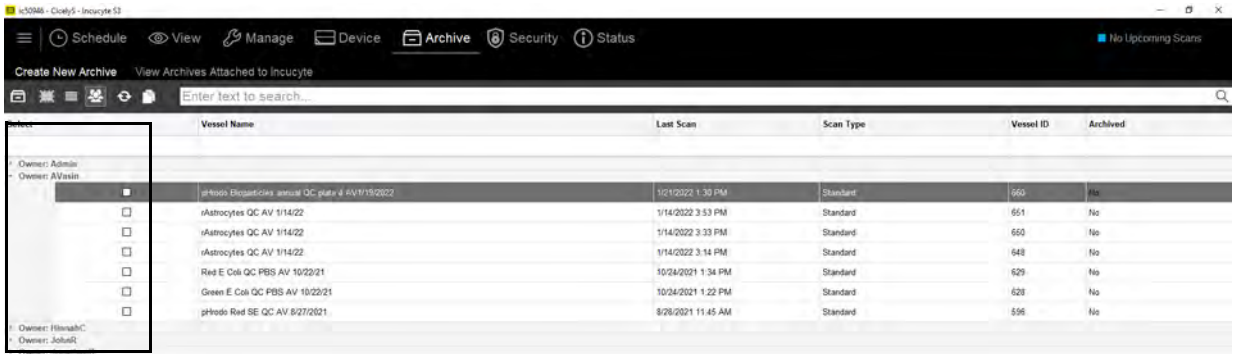
Because the Archive window displays the vessels as they are scanned in real-time, you should periodically click the Refresh icon to update the display with the most recently scanned vessels.

At any time, you can click the Copy icon to copy all the information that is currently displayed in the Archive window to your client clipboard, and then you can paste this information into a third-party application such as Microsoft Excel.

2. Select the vessels that you are archiving. When selecting the vessels that you are archiving, note the following options that are available for searching for and selecting the appropriate vessels:

Icon	Description
	Select all icon: Click this icon to select all vessels for archiving.
N/A	To select one or more vessels at a time for archiving, in the Select column, click the blank check box that is displayed for a vessel. If the vessel is not to be archived, then click the check box again to clear the selection.
	Clear All icon: Click this icon to clear all currently selected vessels in a single step.
	Group by Owner icon: Click this icon to group all the vessels by the owner (user). See Figure A-2 below. **Tip:** This option is particularly useful for the logged-in user, who can now immediately see the list of vessels that have been scanned, and then archive them as appropriate.
	Refresh Grid data icon: Click this icon to update the display on the Archive window with the most recently scanned vessels.
	Copy grid to clipboard : Click this icon to copy the contents that are currently displayed on the Archive window to your client's clipboard. You can now paste this information into a third-party application such as Microsoft Excel for further sorting, evaluation, and so on.

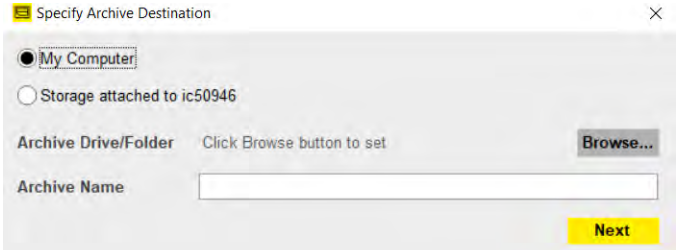
Figure A-2: Archive page, Vessels grouped by owner



3. After you have selected all the vessels that you are archiving, click the Archive Vessel icon  .

   The Specify Archive Destination dialog box opens. By default, My Computer is selected.

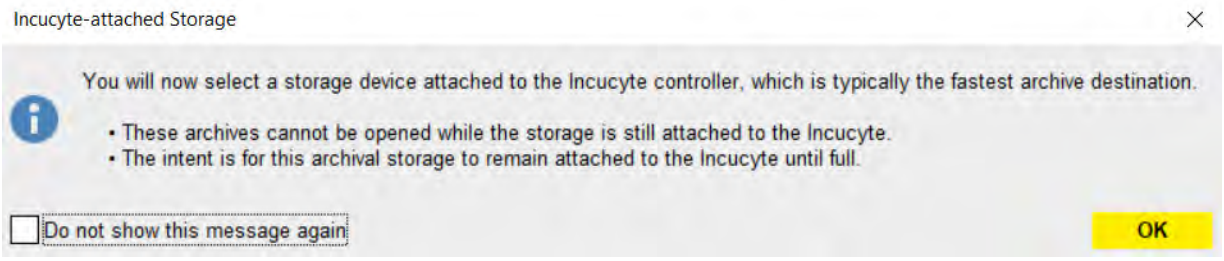
Figure A-3: Specify Archive Destination dialog box



4. Leave My Computer selected, or select Storage Attached to Incucyte.
5. Click Browse to open the Select a folder dialog box, and then browse to and select the drive and folder for the archive.

If you selected Storage Attached to Incucyte, then an Incucyte-attached Storage message opens with a list of caveats about selecting this option. You must click OK to close this message before you can continue to the next step. See Figure A-4 below.

Figure A-4: Incucyte-attached Storage message

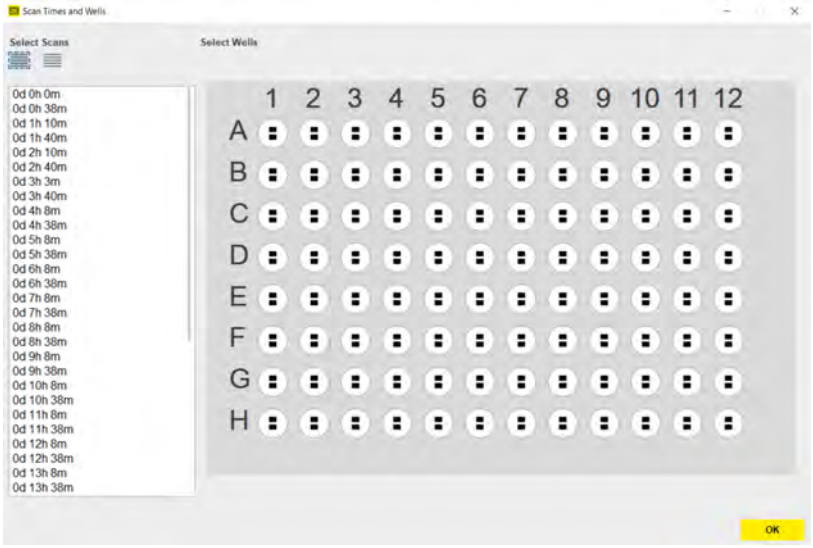


6. Enter a name for the archive.
7. Click Next.

   Two results are possible:

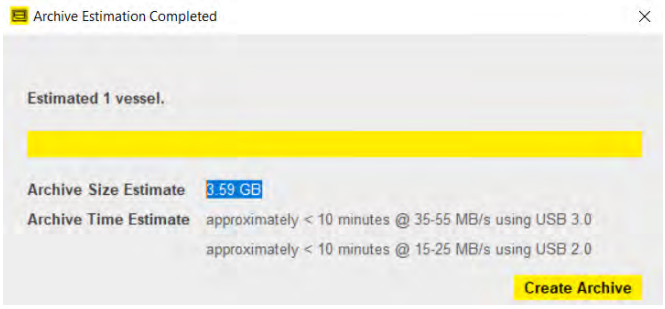
   - If you selected only a single vessel for archiving, then the Scan Times and Wells dialog box opens. Go to Step 8.

Figure A-5: Scan Times and Wells dialog box



   - If you selected more than one vessel for archiving, then the Estimating Archive dialog box opens. The dialog box displays the estimates for the archive size and the time required to archive the selected data. (See Figure A-6 on page 224.) After the estimates are completed, Create Archive is enabled on the dialog box. Go to Step 10.

Figure A-6: Estimating Archive dialog box



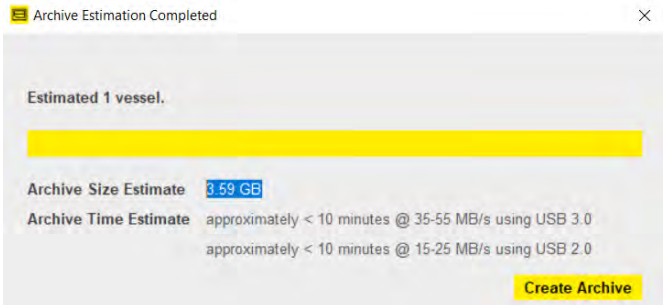
8. Use the options on the Scan Times and Wells dialog box to select the scan times and vessel locations that you are archiving.

The Scan Times and Wells dialog box contains the same standard options for selecting scan times and vessel locations that you have encountered previously in the Incucyte software, such as when you define an analysis or export an image. Refer to one of these sections in the manual if you need a refresher about selecting scan times and/or vessel locations.

9. Click OK.

   The Scan Times and Wells dialog box closes. The Estimating Archive dialog box opens. The dialog box displays the estimates of both the archive size and the time required to archive the selected data. After the estimates are completed, Create Archive is enabled on the dialog box.

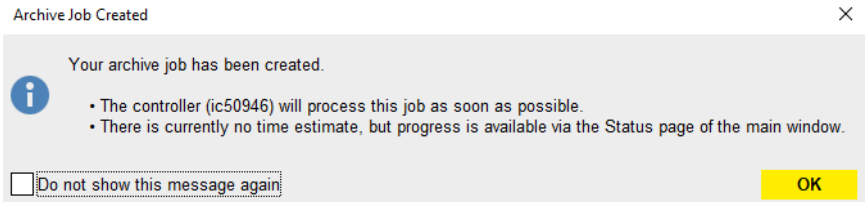
Figure A-7: Estimating Archive dialog box



10. Click Create Archive.

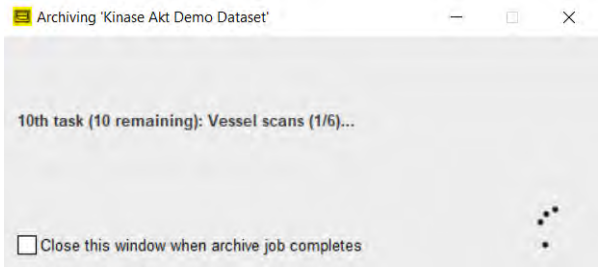
    An Archive Job Created message opens. The message informs you that the archive job has been created and that controller will process the job as soon as possible. A separate Archiving status message also opens, indicating the status of archiving the selected data. See Figure A-9 on page 225.

Figure A-8: Archive Job Created message



To keep the Preparing Archive message from opening again for this and all future instances of Incucyte, click Do not show this message again.

Figure A-9: Archiving status message



11. In the Archive Created message, click OK.

    The Archive Created message box closes.

12. Optionally, in the Archiving status message, click Close this window when archive job completes, or wait until the message indicates that the archive job is complete, and then in the upper right corner of the message, click the “X” to close it.

Viewing a List of the Archives Attached to Incucyte

Because you cannot open archives while the storage is attached to the Incucyte controller and you should detach and reattach the external storage device only when absolutely necessary, a View Archives Attached to Incucyte function is provided in the Incucyte software. You use this function to view a list of all the archives that are currently stored on the dedicated device that is attached to the Incucyte controller without having to detach the device. You can view the archives that are attached to your Incucyte controller at any time.

To open an archive that is attached to the Incucyte storage device, see "Opening an Archive" on page 227.

To view a list of the archives attached to Incucyte

1. On the Incucyte menu, click Archive.

   The Archive window opens. By default, the Create New Archive tab is open. See Figure A-1 on page 221.

2. Click View Archives Attached to Incucyte.

   The View Archives Attached to Incucyte tab opens. The tab displays a list of archives, sorted by archive name, that are stored on the dedicated device that is attached to your Incucyte controller. Note the following about the window:

   - The standard Incucyte filter and sort and search functions are available for searching for a specific archive. See "Working with Data Columns in an Incucyte Window" on page 26.
   - Because the Archive window displays the vessels as they are scanned in real-time, you should periodically click the Refresh archives icon  to update the display with the most recently scanned vessels.
   - At any time, you can click the Copy icon  to copy the information that is currently displayed in the Archive window to your client clipboard, and then you can paste this information in to a third-party application such as Microsoft Excel for further sorting, evaluation, and so on.

Opening an Archive

An archive is saved with a file extension of .icarch. You can open and view any archive that has been created for your Incucyte system, whether the archive is saved to My Computer or Storage Attached to Incucyte. You have two options for opening an archive: You can open an archive during the start up of Incucyte, or you can open an archive with a connection after you have opened and logged in to Incucyte.

To view an archive that has been saved to Storage Attached to Incucyte, you must first detach the storage device from your Incucyte system, and then attach the device to your Incucyte client so that the Incucyte software can recognize the device.

To open an archive

1. Do one of the following:
   - To open an archive when you start Incucyte, launch Incucyte.
   - To open an archive with a connection, do the following:
     a. From any window with a Menu bar in Incucyte, click the Menu bar to open a dropdown menu.
     b. On the dropdown menu, select Connection.

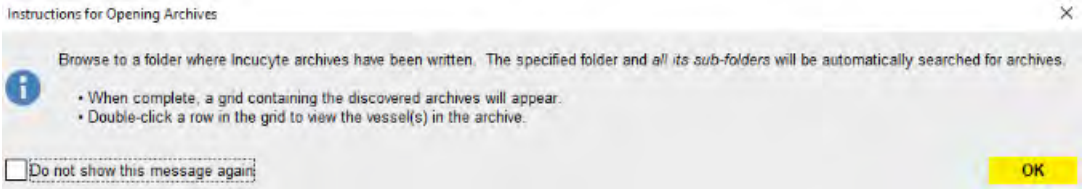
   The Incucyte Open Connection dialog box opens.

Figure A-10: Incucyte Open Connection dialog box

2. Click Open an Archive.

   An Instructions for Opening Archives message opens. The message details how to access an Incucyte archive.

Figure A-11: Instructions for Opening Archives message



To keep the Instructions for Opening an Archive message from opening again for this and all future instances of Incucyte, click Do not show this message again.

3. Optionally, read the message to ensure that you understand how to access an archive, and then click OK.

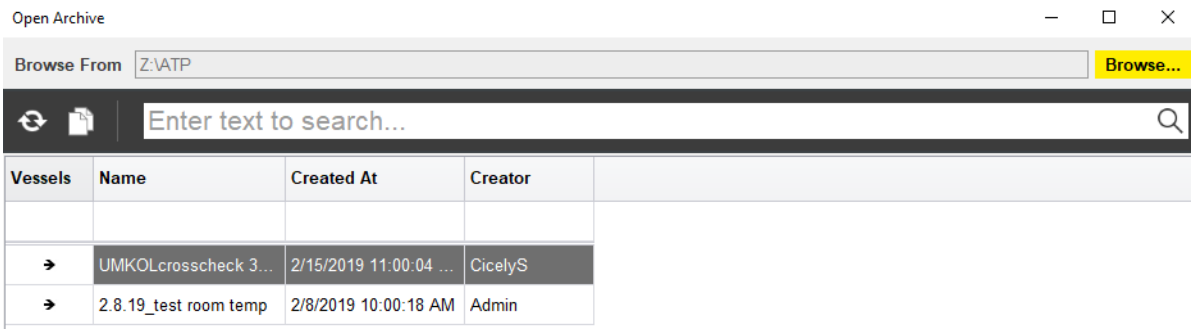
   The message closes and the Select Folder dialog box opens.

4. In the Select Folder dialog box, browse to and select the appropriate folder.

   The Open Archive window opens. The window displays a list of all the archives that are contained in the selected folder. Note the following about the window:

   - The standard Incucyte filter and sort and search functions are available for searching for a specific archive. See "Working with Data Columns in an Incucyte Window" on page 26.
   - You can click the Expand/Collapse icon that is displayed in the Vessels column for an archive to expand the archive and view a list of all the vessels that are contained in the archive.
   - Because the Archive window displays the vessels as they are scanned in real-time, you should periodically click the Refresh archives icon to update the display with the most recently scanned vessels.
   - At any time, you can click the Copy icon to copy the information that is currently displayed in the Archive window to your client clipboard, and then you can paste this information in to a third-party application such as Microsoft Excel for further sorting, evaluation, and so on.

Figure A-12: Open Archive window



After you open an archived vessel, the Vessel View window, which has the exact same features as the Vessel View window for an unarchived vessel, opens. For full details about this window and its available functions, see "The Vessel View Window" on page 89.

Appendix B
Changing the Incucyte SX1 Objective

If you are using the Incucyte SX1, then when you are specifying the settings for a vessel scan, the currently installed objective is set as the default objective. If you require a different objective, then you must select it *before* you configure and schedule a vessel scan. You must have Administrator privileges to change the objective.

If you configure and schedule a vessel scan for the Incucyte SX1, and then change the objective, the current schedule is immediately cleared and all scans for all vessels currently found in the schedule are immediately ended.

This appendix covers the following topics:

Appendix B
Changing the Incucyte SX1 Objective

Changing the Incucyte SX1 Objective

If you are using the Incucyte SX1, then when you are specifying the settings for a vessel scan, the currently installed objective is set as the default objective. If you require a different objective, then you must select it *before* you configure and schedule a vessel scan. You must have Administrator privileges to change the objective.

If you configure and schedule a vessel scan for the Incucyte SX1, and then change the objective, the current schedule is immediately cleared and all scans for all vessels currently found in the schedule are immediately ended.

To select the objective for the Incucyte SX1

1. On the Incucyte main window, click Device. See Figure 1-4 on page 19.

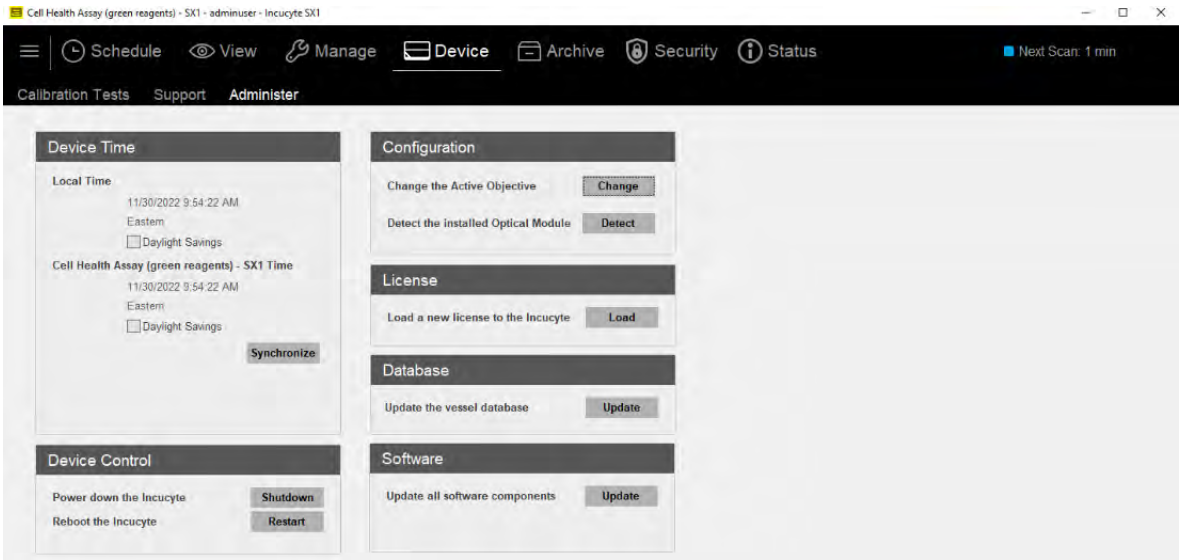
   The Device menu opens. The Calibration Tests tab is the open tab.

You can also click Device in the menu that is displayed at the top of every window in Incucyte.

2. Click Administer.

   The Administer tab opens.

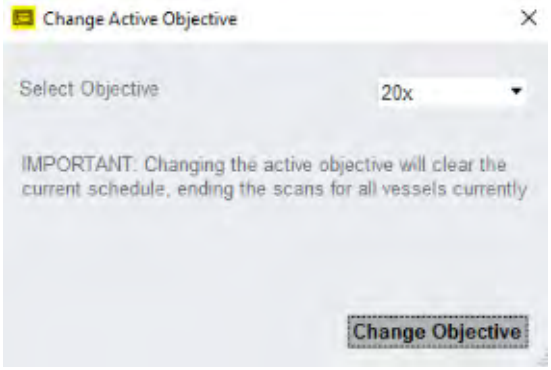
Figure B-1: Incucyte SX1 Administer tab



3. Under Configuration, click Change for Change the Active Objective.

   The Change Active Objective dialog box opens.

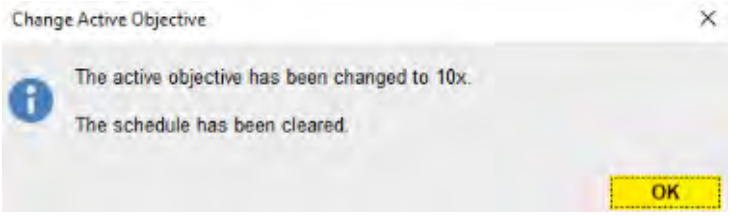
Figure B-2: Change Active Objective dialog box



4. On the Select Objective dropdown list, select the new objective, and then click change Objective.

   A Change Active Objective message opens on top of the Change Active Objective dialog box. The message states that the objective has been changed, and that the schedule has been cleared.

Figure B-3: Change Active Objective message



5. Click OK.

   Both the Change Active Objective dialog box and message close. You can now configure and schedule a vessel scan. See “Configuring and Scheduling a Vessel Scan” on page 39.

Appendix C
Changing the Incucyte SX5 Optical Module

If you are using the Incucyte SX5, and you require a different optical module, then you must select it *before* you configure and schedule a vessel scan. You must have Administrator privileges to change the optical module.

If you configure and schedule a vessel scan for the Incucyte SX5, and then change the optical module, the current schedule is immediately cleared and all scans for all vessels currently found in the schedule are immediately ended.

This appendix covers the following topics:

Appendix C
Changing the Incucyte SX5 Optical Module

Changing the Incucyte SX5 Optical Module

If you are using the Incucyte SX5, and you require a different optical module, then you must select it *before* you configure and schedule a vessel scan. You must have Administrator privileges to change the optical module.

If you configure and schedule a vessel scan for the Incucyte SX5, and then change the optical module, the current schedule is immediately cleared and all scans for all vessels currently found in the schedule are immediately ended.

To change the Incucyte SX5 optical module

1. On the Incucyte main window, click Device. See Figure 1-4 on page 19.

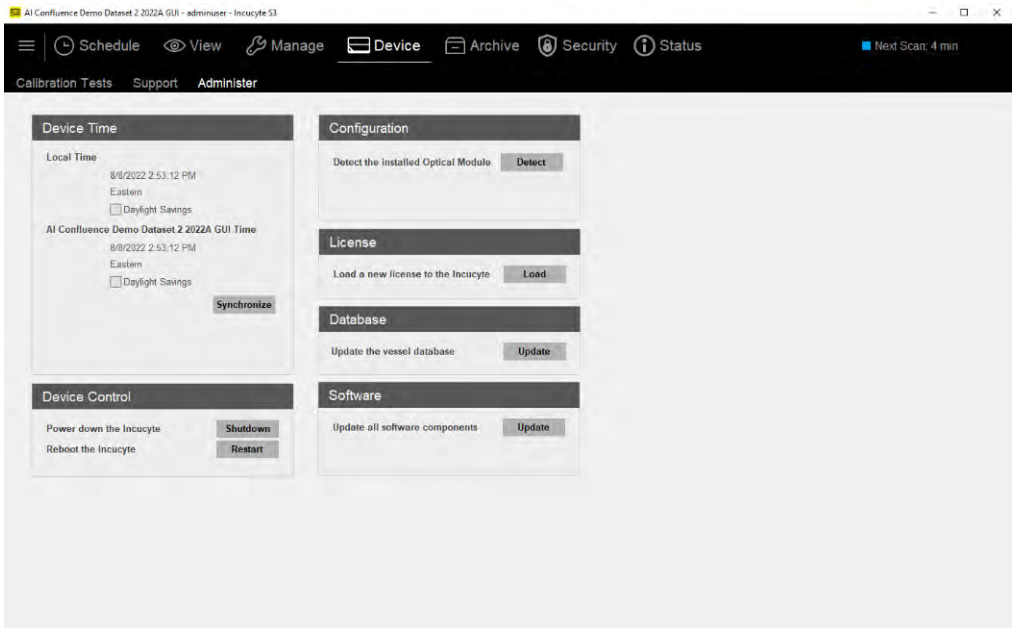
   The Device menu opens. The Calibration Tests tab is the open tab.

You can also click Device in the menu that is displayed at the top of every window in Incucyte.

2. Click Administer.

   The Administer tab opens.

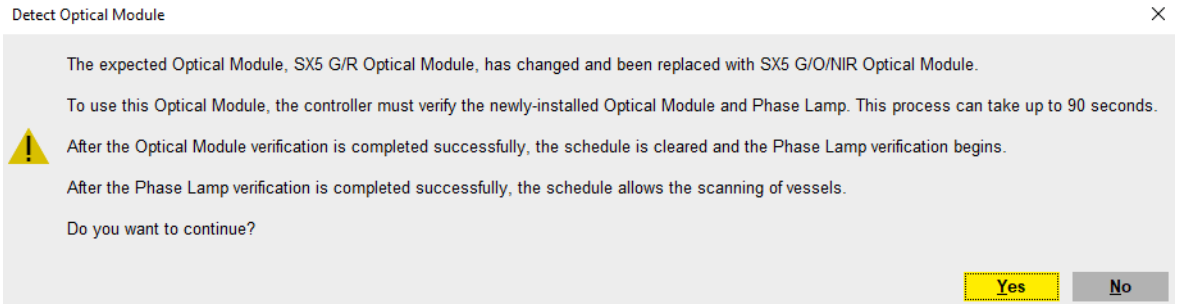
Figure C-1: Incucyte SX5 Administer tab



3. For the Configuration option, click Detect.

   A Detect Optical Module message opens, informing you that detection verification of the optical module and phase lamp is going to take place, and asking you if you want to continue.

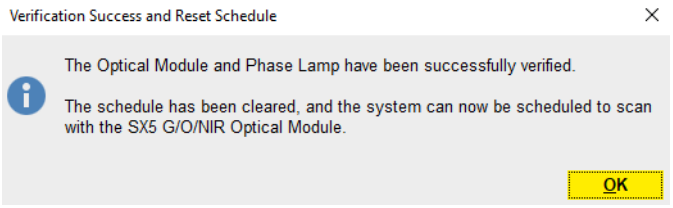
Figure C-2: Detect Optical Module message



4. Click Yes.

   A status message opens, indicating that the detection verification process has begun. After the process has successfully completed, the status message closes and a Verification Success and Reset Schedule message opens.

Figure C-3: Verification Success and Reset Schedule message



5. Click OK.

   You can now configure and schedule a vessel scan. See “Configuring and Scheduling a Vessel Scan” on page 39.

Appendix D
Incucyte Security

Users are the people who log into Incucyte, whether they are adding and reviewing content, or just using Incucyte in a read-only capacity. Remember, when Incucyte is first installed, it is installed with a default Administrator user. As the Incucyte administrator, you must manage all the necessary users for your Incucyte installations, which includes adding new users, and editing users, including editing user permissions, and inactivating users. You mus also manage user workgroups, which define the users who can view specific vessels and the vessels' data, experiment definitions, and analysis definitions. This appendix details all the procedures and considerations that are necessary for managing users and user workgroups to ensure the security of an Incucyte and its data.

This appendix covers the following topics:

Appendix D
Incucyte Security

Adding a User to Incucyte

When you add a user, you must specify the *user type* and the *user permission level*. The user type is either User or Domain User, where:

- User: A user must log in to Incucyte with credentials (user name and password) that are specific for the instrument. This type of user is referred to as a *local user*. See "To add a local user to an Incucyte" on page 244.
- Domain User: A user can log in to Incucyte with his/her Microsoft Windows credentials (user name and password). These credentials, therefore, are not specific to the instrument. As the Incucyte administrator, you can make it optional for a domain user to log in using domain credentials, or you can make it required. See "To add a Domain user to an Incucyte" on page 245.

The permission level determines the access that a user has to Incucyte and the actions that a user can therefore carry out, such as read-only access (the user cannot carry out any actions in Incucyte), or Access to Schedule (the user can create a new vessel to scan on schedule). Table D-1 below details the user permission levels in Incucyte, organized by UI location.

The user permission level of Operator is available only in the 21 CFR Part 11 compliant version of Incucyte software. See the Incucyte 21 CFR Part 11 Software Guide for further details.

Table D-1: User Permission Levels

		Permission Level				
UI	**Action/Access**	**Admin**	**Standard**	**Limited**	**Guest**	**Operator**
Launch	Access to Schedule	X	X			X
	Access to View	X	X	X	X	X
	Access to Manage	X	X	X		X
	Access to Device	X	X	X		X
	Access to Archive	X	X	X		X
	Access to Security	X				
	Access to Status	X	X	X	X	
Application Menu	Change Password*	X	X	X		X
*Change password is the only Application Menu option that has a restriction for a Guest User. All other menu options have no such restriction.						
Schedule	Create New Vessel to Scan	X	X			X
	Edit Any Unscanned Vessel	X	X			
	Edit Experiment End For Any Vessel	X	X			
	Remove Any Vessel from Repeating Schedule	X	X			
	Create New Scan Group for Any Vessel	X	X			
	Modify Any Scan Group	X	X			

Table D-1: User Permission Levels (Continued)

		Permission Level				
UI	**Action/Access**	**Admin**	**Standard**	**Limited**	**Guest**	**Operator**
View	Open Any Vessel	X	X	X	X	X
	Open Any Analysis	X	X	X	X	X
	Graph Metrics for Any Analysis	X	X	X	X	X
	View Details for Any Analysis	X	X	X	X	X
	Export Definition for Any Analysis	X	X	X		
	Delete Analysis Created by You	X	X	X		
	Delete Any Analysis	X				
Manage	Edit Experiment Definition Created by You	X	X	X		
	Edit Any Experiment Definition	X				
	Rename Experiment Definition Created by You	X	X	X		
	Rename Any Experiment Definition	X				
	Create Copy of Any Experiment Definition	X	X	X		
	Create Protected Copy of Any Experiment Definition	X	X	X		
	Import or Export Experiment Definition	X	X	X		
	Delete Experiment Definition Created by You	X	X	X		
	Delete Any Experiment Definition	X				
	Edit Analysis Definition Created by You	X	X	X		
	Edit Any Analysis Definition	X				
	Rename Analysis Definition Created by You	X	X	X		
	Rename Any Analysis Definition	X				
	Create Copy of Any Analysis Definition	X	X	X		
	Create Protected Copy of Any Analysis Definition	X	X	X		

Table D-1: User Permission Levels (Continued)

		Permission Level				
UI	**Action/Access**	**Admin**	**Standard**	**Limited**	**Guest**	**Operator**
	Import or Export Analysis Definition	X	X	X		
	Delete Analysis Definition Created by You	X	X	X		
	Delete Any Analysis Definition	X				
	Delete Any Vessel Created by You	X	X			
	Delete Any Vessel	X				
Device	Perform and View Calibrations	X				
	Export Setup Files or Logs	X	X	X		X
	Export Databases	X	X	X		X
	Export Scan Diagnostics	X	X	X		X
	View Administrator Page	X	X	X		
	Synchronize Device Time	X	X	X		
	Shut Down or Restart Incucyte	X	X	X		
	Perform Hardware or Software Changes	X				
Archive	View Create Archive Page	X	X	X		
	Create Local Archive for Any Vessel	X	X	X		
	Create Device-Attached Archive for Any Vessel	X	X	X		
	View Archives Attached to Incucyte Page	X	X	X		
	Delete Device-Attached Archive Created by You	X	X	X		
	Delete Any Device-Attached Archive	X				
Security	View, create or edit users	X				
	Change Local User Password	X				
	Change login settings	X				

Table D-1: User Permission Levels (Continued)

		Permission Level				
UI	**Action/Access**	**Admin**	**Standard**	**Limited**	**Guest**	**Operator**
Status	View Status Page	X	X	X	X	X
	Delete In Progress Archive Created by You	X	X	X		
	Delete Any In Progress Archive	X				
	Delete In Progress Analysis Created by You	X	X	X		
	Delete Any In Progress Analysis	X				
	Add Service Log Entry	X	X	X		
	Export Log Entries	X	X	X		
Vessel View	View Any Scan Time	X	X	X	X	X
	Modify Image Channel Options (On/Off, Scaling, Brightness, Contrast)	X	X	X	X	X
	Modify Spectral Unmixing Values for Any Vessel	X	X	X		
	Launch New Analysis for Any Vessel	X	X	X		
	Go to Scan Metrics	X	X	X	X	X
	Go to Scan Metrics with All Diagnostic Metrics	X				
	Go to Vessel Information	X	X	X	X	X
	Launch Image and Movie Export	X	X	X		X
Launch Analysis	Create Analysis	X	X	X		
Analysis View	Go to Edit User-Defined Metrics for any Analysis	X	X	X		
	Launch Classification for Any Cell-by-Cell Analysis	X	X	X		

Table D-1: User Permission Levels (Continued)

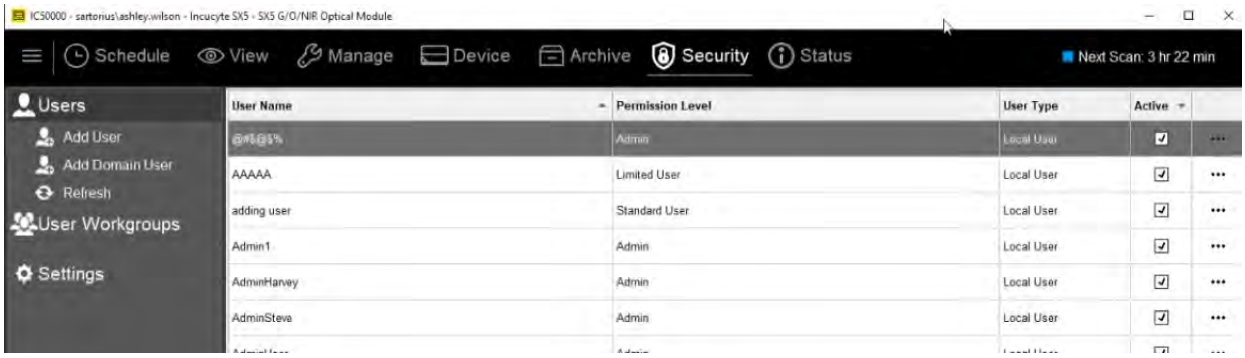
		Permission Level				
UI	**Action/Access**	**Admin**	**Standard**	**Limited**	**Guest**	**Operator**
Metrics	View Time Plot Metrics for Any Analysis	X	X	X	X	X
	Create or Edit User-Defined Time Plot Metric for Any Analysis	X	X	X		
	Delete Any User-Defined Time Plot Metric for Any Analysis	X	X	X		
	Apply Any User-Defined Time Plot Metrics to Analysis Definition Created by You	X	X	X		
	Apply Any User-Defined Time Plot Metrics to Any Analysis Definition	X				
	Save or Load Generated Graphs to or from Disk	X	X	X		X
	Export Metric Data	X	X	X		X
Vessel/Analysis Information	Create, Edit, or Delete Plate Map for Any Vessel	X	X	X		
	Import or Export Plate Map for Any Vessel	X	X	X		
	Create or Modify Details/ Notes for Any Vessel	X	X	X		X
	Create or Modify Details/ Notes for Any Analysis	X	X	X		X

To add a local user to an Incucyte

1. On the Incucyte main menu, click Security.

   The Users window opens. The window lists all the users (local and Domain) that have been added to the instrument.

Figure D-1: Users window



2. Click Add User.

   The window is updated with an Add User pane.

Figure D-2: Users window with Add User pane

3. Enter the information for the new user.

Option	Description
User name	User names for local user accounts cannot contain the following characters: a space, or any of the following: \ < > " & ? ;

Option	Description
Password	Passwords are alphanumeric and must have the following characteristics: • Contain at least 8 characters. • Contain at least one upper case character. • Contain at least one lower case character. • Contain at least one digit (number).
Confirm Password	You must enter the password in this field exactly as you did in the Password field. **Note:** This field is protected. You cannot copy the password from the Password field, and then paste the password into this field.
Caution: No option is available for recovering a user's password if the user loses/forgets it. Make sure that the user is aware that the password should be stored in a safe and recoverable location; otherwise, the only option that is available is to reset (edit) the user's password. See "Editing a User for an Incucyte" on page 251.	
Permission Level	Select the appropriate permission level for the user. See Table D-1 on page 239 for a detailed list of the access/actions provided to a permission level.
User Workgroups	Optional. Select the user workgroups to assign to the user. The user can view all vessels and their data in the assigned workgroups. Users cannot view any vessels and their data in the user workgroups that you do not assign to them.

4. Click Save.

   The local user is added by default as an Active user.

After you add a new user, click Refresh to refresh the display and ensure that the user was added successfully.

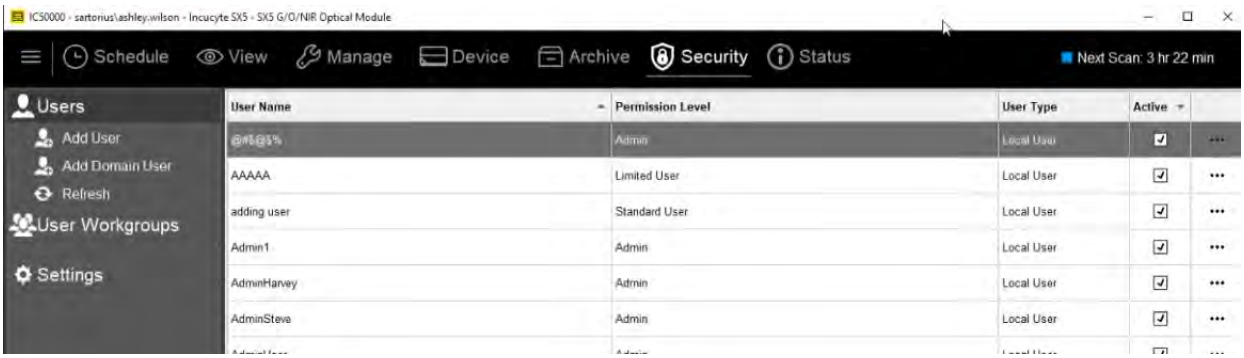
To add a Domain user to an Incucyte

Even though a Domain user uses Windows domain credentials to log in to Incucyte, you must still add the user to Incucyte before they can log in to the instrument. If a Domain user is to have access to multiple Incucytes within a domain, then you must add the Domain user to every applicable Incucyte.

1. On the Incucyte main menu, click Security.

   The Users window opens. The window lists all the users (local and Domain) that have been added to the instrument.

Figure D-3: Users window

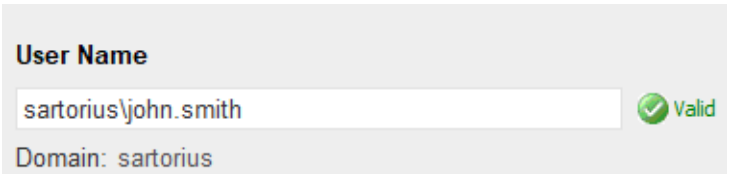


2. Click Domain User.

   The window is updated with an Add Domain User pane.

Figure D-4: Users window with a Domain User pane

3. Enter the information for the new user.

Option	Description
User name	Enter the user name for the domain user, making sure to specify the correct domain in the correct format; for example, sartorius\john.smith. **Note:** If you enter the user name correctly, then a “Valid” indicator is displayed next to the name. *Figure D-5: Valid indicator for a Domain User name* 
Domain	After you enter the domain user name, the domain is automatically parsed from the entered user name and displayed as read-only information in this field.
Permission Level	Select the appropriate permission level for the user. See Table D-1 on page 239 for a detailed list of the access/actions provided to a permission level.
User Workgroups	Optional. Select the user workgroups to assign to the user. The user can view all vessels and their data in the assigned workgroups. Users cannot view any vessels and their data in the user workgroups that you do not assign to them.

4. Click Save.

   The domain user is added by default as an Active user.

After you add a new user, click Refresh to refresh the display and ensure that the user was added successfully.

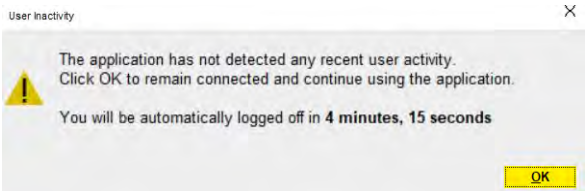
If you add Domain users to Incucyte, and they do not have local user accounts, then you must enable at least one of the available Windows Authentication settings, otherwise, your users are not able to log in to Incucyte. See "Specifying User Settings for an Incucyte" on page 248.

Specifying User Settings for an Incucyte

You have the option of specifying two settings—User Inactivity Timer and Automatic Logoff Countdown—that all apply to all users (local and Domain) that have been added to an instrument. You can also specify values for Windows Authentication settings that apply only to Domain Users for the instrument. Finally, if you intend to use User Workgroups to control user access to specific vessels and their data, you must turn on the User Workgroups setting.

Option	Description
Windows Authentication - These values apply only to the Domain users that have been added to an instrument.	
• Enable associated Windows domain accounts to login to the Incucyte using their Windows domain user name and passwords • Require users to log in with Window domain user names when connecting to the device	• By default, turned Off. If turned On, then users can use their Windows domain credentials to log in to Incucyte, but they are not required to do so. As a result, if the user has a local user account, then the user can still log in to Incucyte. • If enabled, then a Domain user can use only Domain credentials to log in to Incucyte, even if the user has a local account.
User Inactivity Timer - This value applies to all users (local and Domain) that have been added to an instrument.	
Specify how long a user may be Inactive before warning of automatic logoff	The default value is None. The value that you specify determines how long a logged in user can be Inactive on an instrument before a User Inactivity message opens, indicating the amount of time that remains before the user is automatically logged off the instrument. Allowed inactivity values range from a low of 1 minute to a high of 5 hours, or Never. See Figure D-6 below.
Automatic Logoff Countdown - This value applies to all users (local and Domain) that have been added to an instrument.	
Specify how long an inactive user has to respond before being automatically logged off.	The default value is None. Works in conjunction with the User Inactivity Timer value. The dynamic countdown value that is displayed in the User Inactivity message indicates how long a user has to respond to the message before being automatically logged off. Countdown values range from a low of 1 minute to a high of 5 hours, or Never.
User Workgroups - This value applies to all users (local and Domain) that have been added to an instrument.	
User Workgroups	The default value is Off. Administrators can control user access to vessels and their data, experiment definitions, and analysis definitions. Vessels and definitions that shared with a user workgroup are viewable only to those users who are members of the workgroup. See “Managing User Workgroups” on page 254.

Figure D-6: User Inactivity message



To specify user settings for an Incucyte

To ensure that at least one user always has access to the Security page, the following caveats are applicable before you can turn on the Require users to log in with Domain names setting:

- A Domain user with Admin permissions must already exist. See “To add a Domain user to an Incucyte” on page 245.
- The Require users to log in with a Domain name setting is enabled only *after* you turn on and save the Enable associated Windows domain accounts setting.

1. On the Incucyte main menu, click Security.

   The Users window opens. The window lists all the users (local and Domain) that have been added to the instrument. See Figure D-1 on page 244.

2. Click Settings.

   The Settings window opens. The window displays the following user settings:

   - Windows Authentication. These values apply only to the Domain users that have been added to the instrument. By default, these values are turned off.
   - User Inactivity Timer and Automatic Logoff Countdown and their current values. By default, the values are initially set to Never.
   - User Workgroups. Allows for the assignment of users to workgroups. If a vessel, experiment definition, or analysis definition is shared with one or more user workgroups, then only those users who are assigned to these workgroups can view and work with the shared vessels, experiment definitions, and analysis definitions.

Figure D-7: Settings window

3. Do any or all of the following:
    - Turn on or turn off Enable associated Windows domain accounts.
    - Turn on or turn off Require users to log in with Domain names.

If you turn off Require users to log in with Domain names, Enable associated Windows domain accounts remains turned on.

    - Specify values for the User Inactivity Timer and/or Automatic Logoff Countdown.
    - Turn on or turn off User Workgroups.
4. Click Save.

Editing a User for an Incucyte

You can edit the user permission level, the user workgroups, and the user status (Active or Inactive) for a local user or a Domain user for an Incucyte. Because a status edit is instrument specific, if a Domain user has access to multiple Incucytes within a domain, and the user is to have an Inactive status across all instruments in the Domain, you must edit the status on every applicable Incucyte. You can also edit the password for a local user.

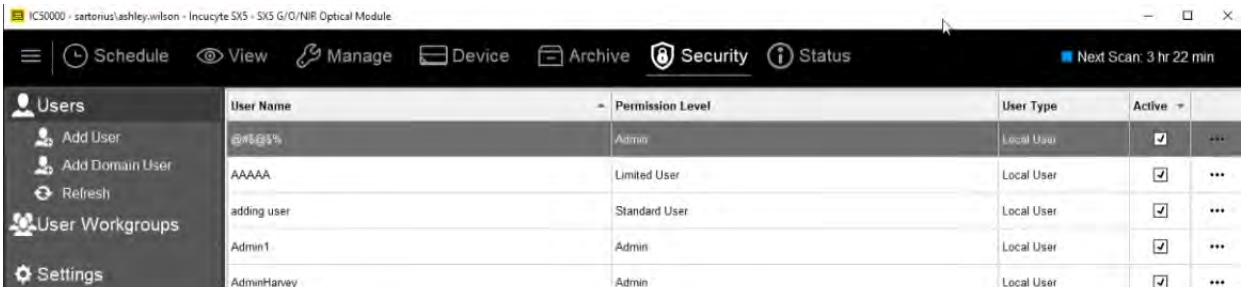
Because scans and analyses are tied to a specific owner and the user name is displayed for the owner, you cannot edit the user name for a user. To edit a user name, you must set up an entirely new user. See "Adding a User to Incucyte" on page 239.

To edit a user for an Incucyte

1. On the Incucyte main menu, click Security.

   The Users window opens. The window lists all the users (local and Domain) that have been added to the instrument.

Figure D-8: Users window

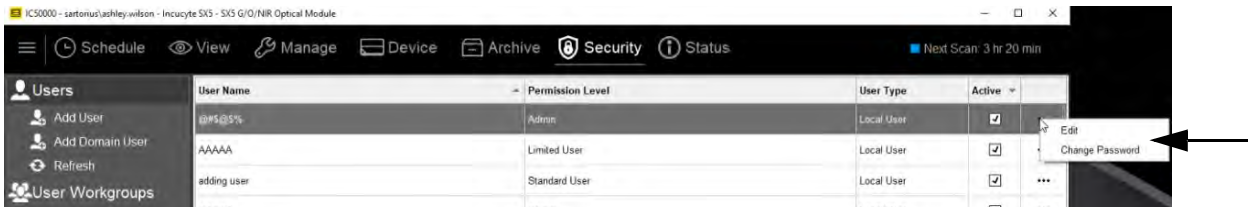


The standard Incucyte filter and sort functions are available on the Users window to assist you in searching for a specific user. See "To sort the data in an Incucyte window" on page 28.

2. To the far right of the entry for the applicable user, click the Ellipsis (. . .) icon.

   A drop-down menu opens with two options: Edit and Change Password.

Figure D-9: User drop-down menu



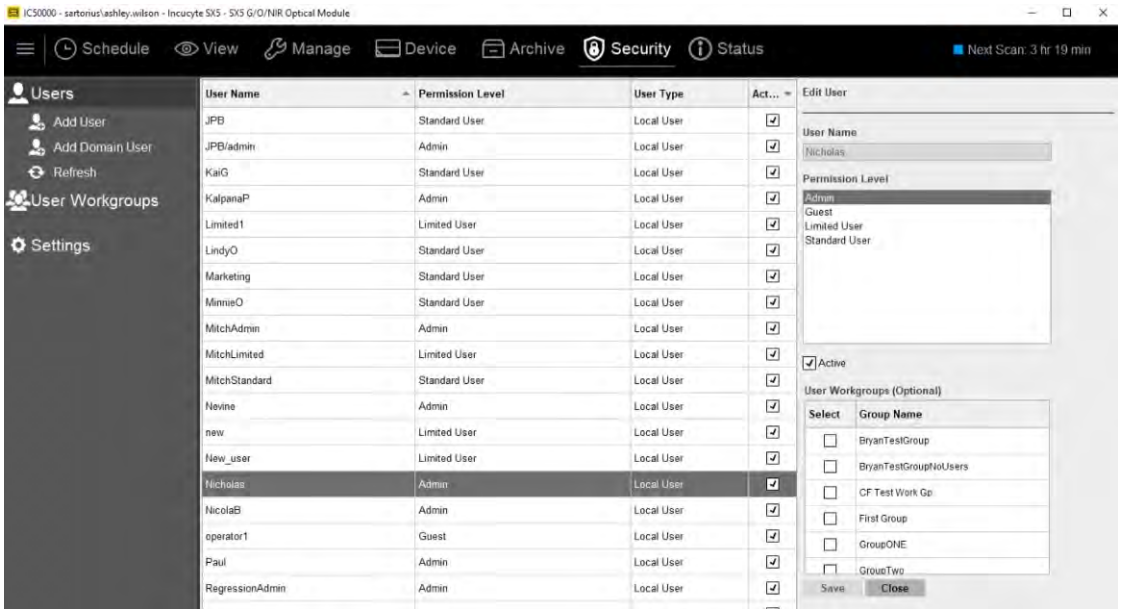
3. Continue to one of the following:
   - "To edit the permission level, user workgroups, or user status" on page 252.
   - "To edit the password for a local user" on page 253.

To edit the permission level, user workgroups, or user status

1. Click Edit.

   The Users window is refreshed with an Edit User pane. The current information for the selected user (name, permission level, and status) is displayed in the pane.

Figure D-10: Edit User pane



2. Edit any or all of the current information for the user.

Option	Description
Permission Level	Select the appropriate permission level for the user. See Table D-1 on page 239 for a detailed list of the access/actions provided to a permission level.
Status	By default, a user is added as an Active user. To inactivate a user, clear the Active checkbox. Conversely, to active an inactive user, select Active. **Tip:** Remember, If a Domain user has access to multiple Incucyte instruments within a domain, and the user is to have an Inactive status across all instruments in the Domain, then you must edit the status on every applicable Incucyte.
User Workgroup	Optional. To assign a user workgroup to the user, click Select for the workgroup. You can assign multiple user workgroups to a user. To remove a user workgroup assignment, clear the Select option for the workgroup.

3. Click Save.

   The Edit User pane closes. The Users window remains open.

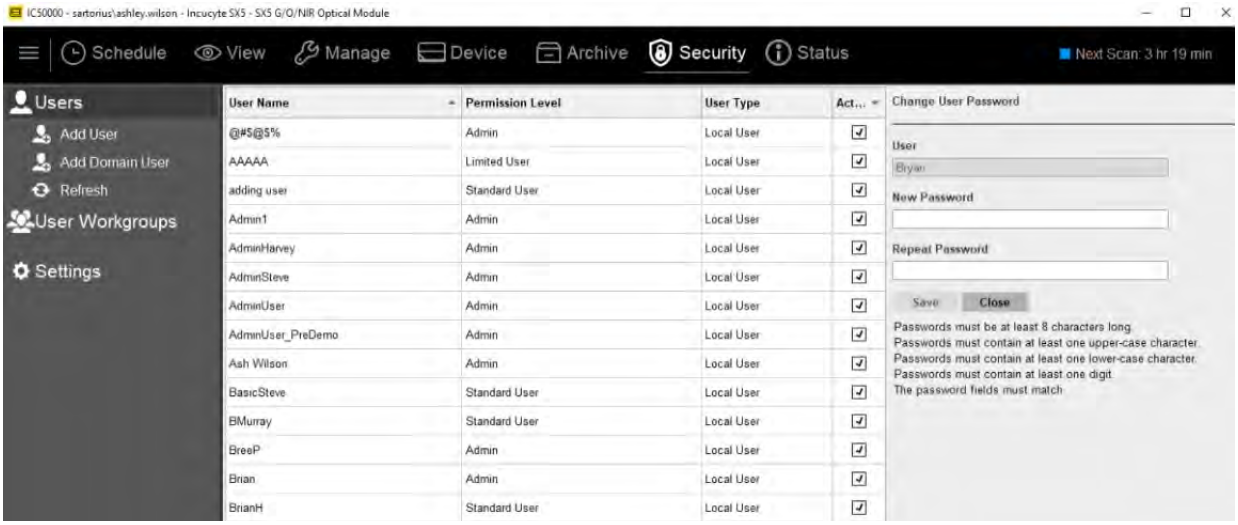
After you edit a user, click Refresh to refresh the display and ensure that your changes have been saved.

To edit the password for a local user

1. Click Change Password.

   The User window is refreshed with a Change User Password pane. The password displays the current user name for the selected user, but you cannot edit the value here.

Figure D-11: Change User Password pane



2. Edit the user's password.

Option	Description
New Password	Passwords are alphanumeric and must have the following characteristics: • Contain at least 8 characters. • Contain at least one upper case character. • Contain at least one lower case character. • Contain at least one digit (number). **Tip:** As you enter the new password, and you fulfill the first of any of its requirements, the requirements list is dynamically refreshed to indicate which requirements that the password has met (a green checkmark) and those that it has not (a red x). Only after the password meets all the requirements is the Save button enabled.
Repeat Password	You must enter the password in this field exactly as you did in the Password field. **Note:** This field is protected. You cannot copy the password from the Password field, and then paste the password into this field.

3. Click Save.

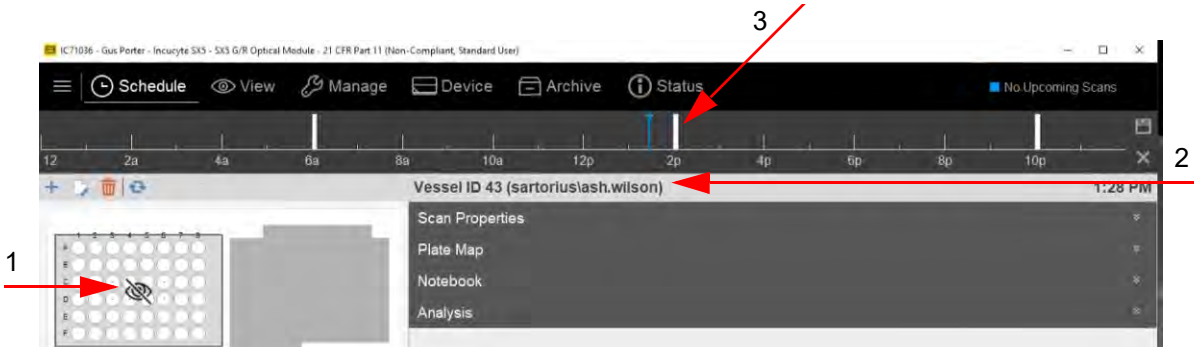
   The Change User Password pane closes. The Users window remains open.

Managing User Workgroups

A *user workgroup* defines the users who can view specific vessels and the vessels' data. You create and organize a user workgroup based on common vessel characteristics that you determine such as your organizational structure (for example, the name of the lab that uses the vessels) or the name of the study that uses the vessels (for example, CAR-T assay). After you create and name a user workgroup, you can assign users to the workgroup, and you can share vessels with the workgroup. Only those users who are assigned to the user workgroup can view and work with the following data for the vessels that are shared with the workgroup:

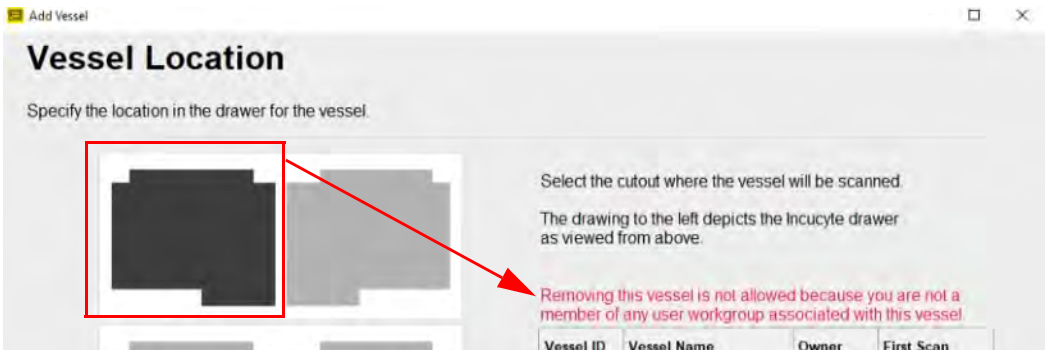
- The complete schedule information for the vessel on the Acquisition window. If you are not an Admin user, if the vessel has not been shared with the **Everyone** user workgroup, or if you are not a member of the workgroup with which the vessel has been shared, then on the Acquisition window, (1) a "Not Viewable" icon is displayed on the vessel image in the Vessel Drawer Setup pane, (2) only the Vessel ID and the user (login) name of the user who scheduled the vessel are displayed at the top of the Vessel Information pane, and (3) the vessel schedule is displayed in the Vessel Schedule timeline; however, you cannot edit the schedule and you cannot remove or delete the scheduled vessel.

Figure D-12: Acquisition window with non-viewable vessel schedule



- The location of the vessel on the Vessel Location page in the Add Vessel wizard. If you are not an Admin user, if the vessel has not been shared with the **Everyone** user workgroup, or if you are not a member of the workgroup with which the vessel has been shared, then you cannot remove the vessel from its location on the Vessel Location page in the Add Vessel wizard. Instead, a warning is displayed on the page stating that "Removing the vessel is not allowed because you are not a member of any user workgroup associated with this vessel."

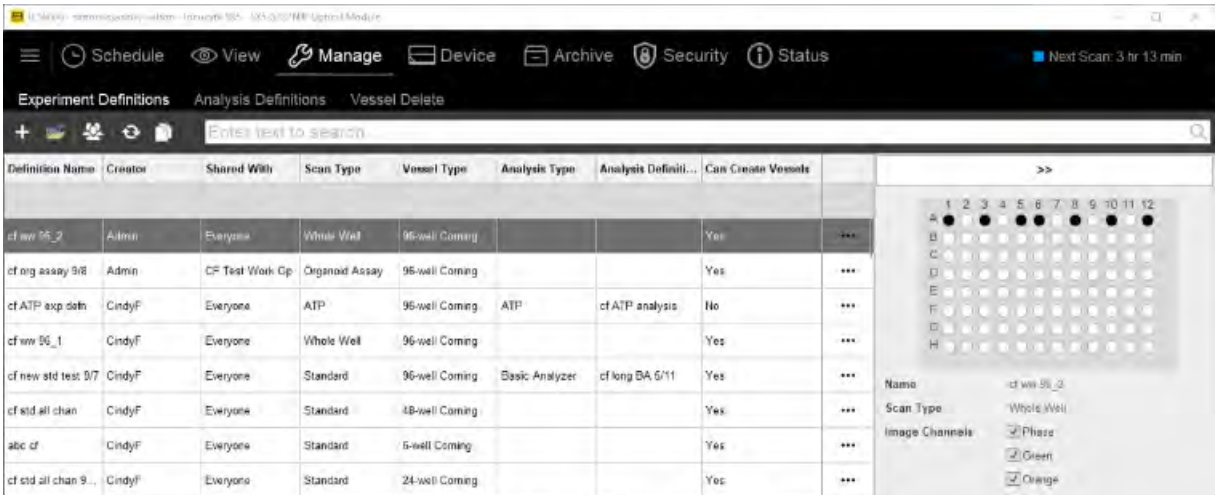
Figure D-13: Vessel Location page, scheduled vessel in a workgroup that you are not a member of



You can also use workgroups to limit a user's access to the following items in the Incucyte software:

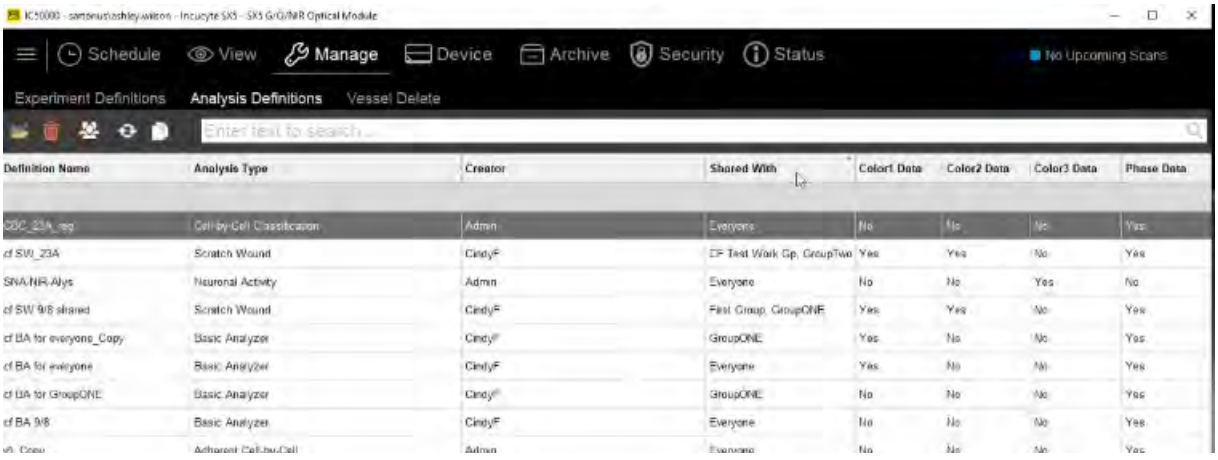
- An experiment definition. If the user is not an Admin user, the experiment definition has not been shared with the **Everyone** user workgroup, or the user is not a member of the workgroup with which the definition has been shared, then the experiment definition is not displayed for the user on the Experiment Definitions tab on the Manage window. The experiment definition is also not displayed on the Experiment Definitions Search page in the Add Vessel wizard.

Figure D-14: Manage window, Experiment Definitions tab



- An analysis definition. If the user is not an Admin user, the analysis definition has not been shared with the **Everyone** user workgroup, or the user is not a member of the workgroup with which the definition has been shared, then the analysis definition is not displayed for the user on the Analysis Definitions tab on the Manage window. The analysis definition is also not displayed on the Analysis Setup page in the Add Vessel wizard.

Figure D-15: Manage window, Experiment Definitions tab



You can assign a user to one or more user workgroups. The permission levels for the user determine the actions that the user can carry out for a vessel and its data whether the **Everyone** user workgroup is assigned to the vessel or the user is a member of the user workgroup with which the vessel is shared. Only

Admin users can manage user workgroups. Managing user workgroups consists of the following activities:

- Creating user workgroups. See “To create a user workgroup” on page 256.
- Editing user workgroups. See “To edit a user workgroup” on page 258.
- Deleting user workgroups.See “To delete a user workgroup” on page 260.

Optionally, you can share a vessel with a user workgroup at the time that you are configuring and scheduling a vessel scan. See “To provide vessel information” on page 50.

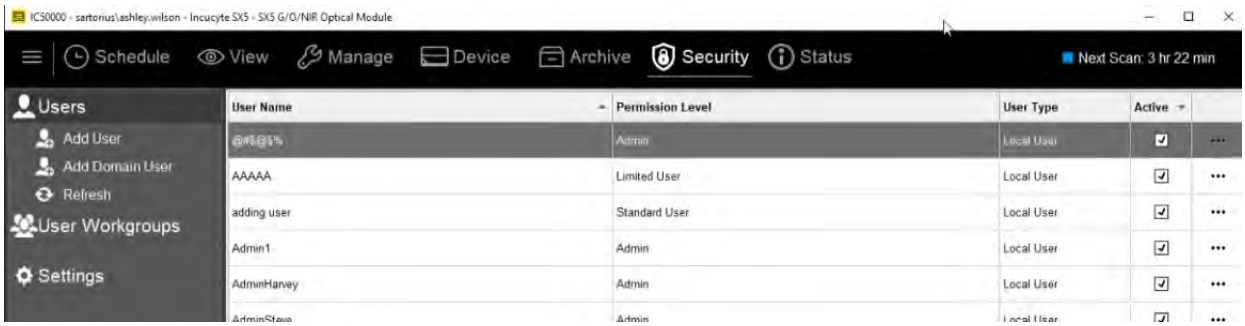
To create a user workgroup

Before you create a user workgroup, make sure that the User Workgroups setting is turned on for the instrument. See “Specifying User Settings for an Incucyte” on page 248.

1. On the Incucyte main menu, click Security.

   The Users window opens.

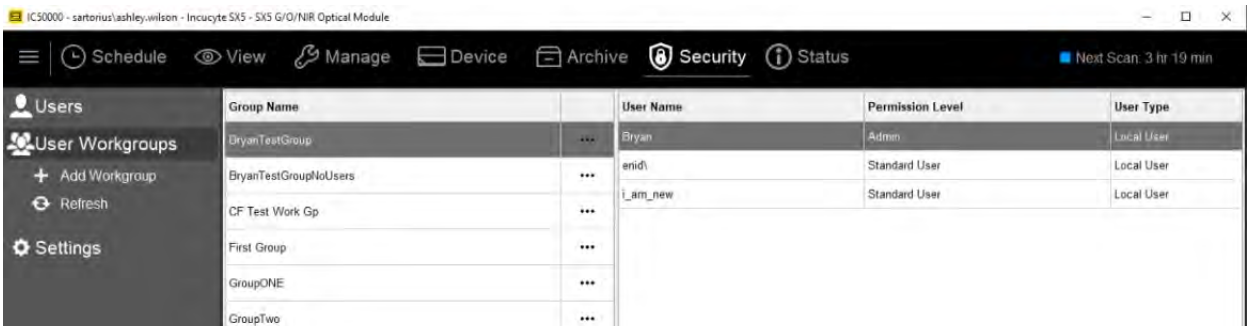
Figure D-16: Users window



2. Click User Workgroups.

   The User Workgroups window opens. The window is divided into two panes. The Group Name (left) pane lists all the user workgroups that are currently defined for the Incucyte. After you select a workgroup in the left pane, the right pane lists all the users who have been assigned to the selected user workgroup. If no users have been assigned to the selected user workgroup, then the message “No users are associated with the selected user workgroup” is displayed.

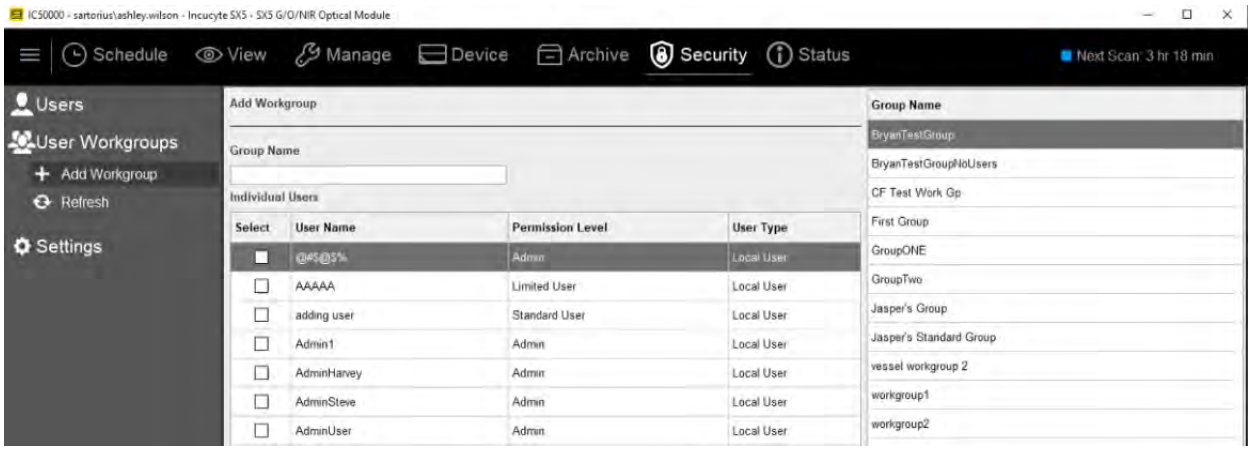
Figure D-17: User Workgroups window



3. Click Add Workgroup.

   The Add Workgroup window opens. The window is divided into two panes. The Add Workgroup (left) pane lists all the users for the Incucyte whom you can add to a workgroup. The Group Name field at the top of the pane is blank. The Group Name (right) pane lists all the user workgroups that are currently defined for the Incucyte. See

Figure D-18: Add Workgroup window



4. In the Group Name field, enter the name for the new user workgroup.
5. To add users to the user workgroup, click Select for each user that you are adding to the workgroup.

The standard Incucyte filter and sort functions are available to assist you in searching for the users whom you are adding to the user workgroup. See "Working with Data Columns in an Incucyte Window" on page 26.

6. Click Save.

   A Success message opens, indicating that the workgroup was created successfully.

Figure D-19: Success message

7. Click OK.

   The Success message closes. The newly added user workgroup is displayed in the Group Name pane.

After you add a user workgroup, click User Workgroups, and then click Refresh to refresh the display on the User Workgroups window and ensure that your changes have been saved.

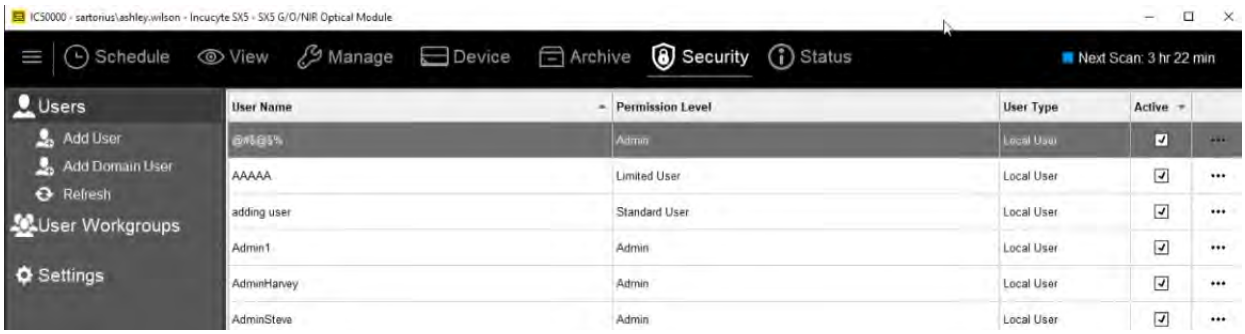
To edit a user workgroup

You can edit the name of a user workgroup and you can edit the users (add or remove) for a workgroup.

1. On the Incucyte main menu, click Security.

   The Users window opens.

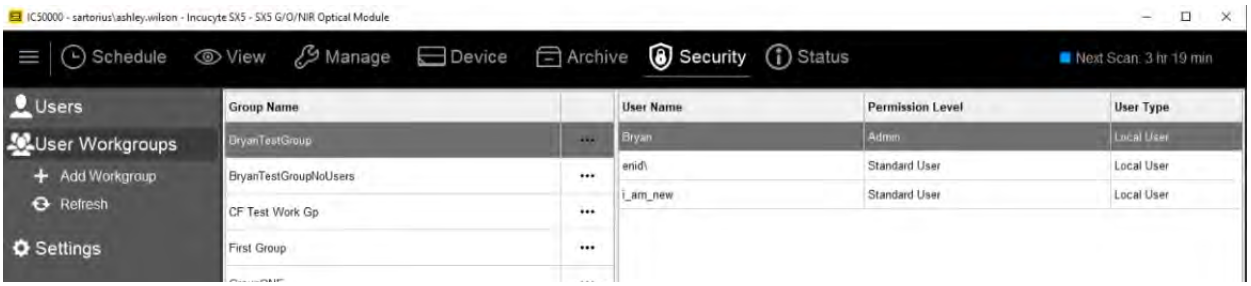
Figure D-20: Users window



2. Click User Workgroups.

   The User Workgroups window opens.

Figure D-21: User Workgroups window

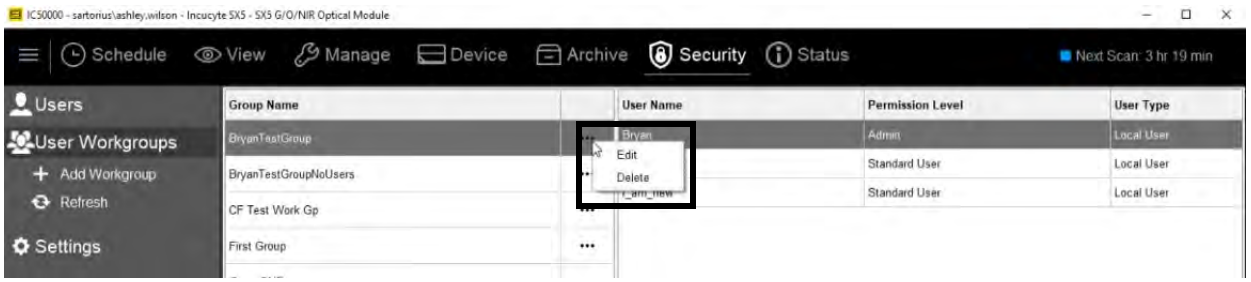


The standard Incucyte filter and sort functions are available to assist you in searching for the user workgroup that you are editing. See "Working with Data Columns in an Incucyte Window" on page 26.

3. To the far right of the entry for the workgroup that you are editing, click the Ellipsis (. . .) icon.

   A drop-down menu opens with two options: Edit and Delete.

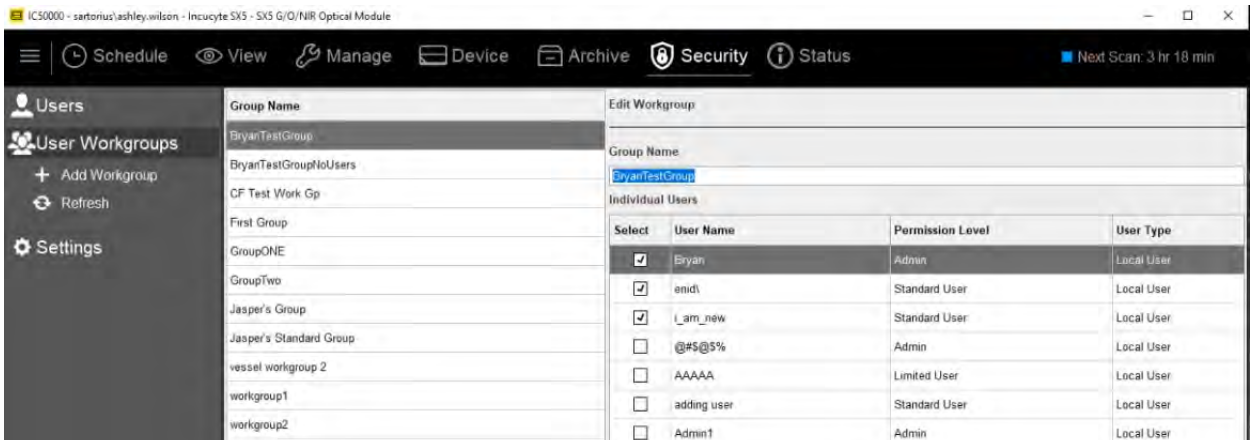
Figure D-22: User Workgroups window with drop-down menu for selected user workgroup



4. Click Edit.

   The Edit Workgroup window opens. The Group Name (left) pane lists all the user workgroups for the Incucyte. The workgroup that you are currently editing is selected in the pane. The right pane first displays all the users who are currently assigned to the workgroup (a check mark is displayed in the Selected column) and then all the users that are available for adding to the group.

Figure D-23: Edit Workgroup window



5. Do any or all of the following:

   - Edit the group name.
   - To add a user to the group, click Select for the user.
   - To remove a user from the group, clear the check mark in the Selected column for the user.

If you delete a user from a user workgroup, the user is deleted only for the selected workgroup. The user is not deleted from any other workgroups to which he/she is assigned.

6. Click Save.

   A Success message opens, indicating that the workgroup was updated successfully.

Figure D-24: Success message

7. Click OK.

   The Success message closes. The newly edited user workgroup remains selected in the Group Name pane.

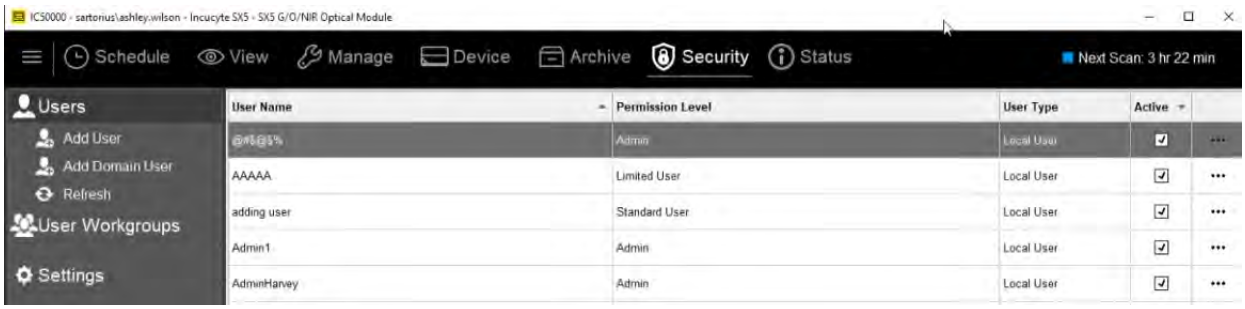
To delete a user workgroup

You can delete a user workgroup with or without assigned users.

1. On the Incucyte main menu, click Security.

   The Users window opens.

Figure D-25: Users window



2. Click User Workgroups.

   The User Workgroups window opens.

Figure D-26: User Workgroups window

The standard Incucyte filter and sort functions are available to assist you in searching for the user workgroup that you are deleting. See "Working with Data Columns in an Incucyte Window" on page 26.

3. To the far right of the entry for the workgroup that you are deleting, click the Ellipsis (. . .) icon.

   A drop-down menu opens with two options: Edit and Delete.

Figure D-27: User Workgroups window with drop-down menu for selected user workgroup

4. Click Delete.

   A Delete Workgroup message opens, asking you if you are sure that you want to delete the selected workgroup.

Figure D-28: Delete Workgroup dialog box

5. Click Yes.

   The Delete Workgroup dialog box closes. You return to the User Workgroups window. The deleted user workgroup is no longer displayed in the Group Name pane.

Appendix D
Incucyte Security

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