ZOE™ Fluorescent Cell Imager

Instruction Manual

Catalog #145-0031

BIO-RAD

Bio-Rad Technical Support

For help and technical advice, please contact the Bio-Rad Technical Support department. In the United States, the Technical Support department is open Monday–Friday, 5:00 AM–5:00 PM, Pacific time.

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Legal Notices

Bio-Rad reserves the right to modify its products and services at any time. This instruction manual is subject to change without notice.

Although prepared to ensure accuracy, Bio-Rad assumes no liability for errors, or for any damages resulting from the application or use of this information.

Bio-Rad Laboratories Resources

Bio-Rad provides many resources for scientists. Bio-Rad resources and how to locate what you need are listed below.

Bio-Rad resources.

Resource	How to Contact
Local Bio-Rad Laboratories representatives	Find local information and contacts on the Bio-Rad Laboratories website by selecting your country on the homepage (**www.bio-rad.com**). Find the nearest international office listed on the back of this manual
Technical support scientists	Bio-Rad's technical support scientists provide our customer with practical and expert solutions. To find local technical support on the phone, contact your nearest Bio-Rad office. For technical support in the United States and Canada, call 1-800-424-6723 (toll-free), and select the technical support option
Service support engineers	Maintenance and repairs should be carried out only by authorized service support engineers For service support in the United States and Canada, call 1-800-424-6723 (toll-free), and select the technical support option to request service support
Technical notes and literature	Go to the Bio-Rad website (**www.bio-rad.com**). Type a term in the Search box and select Documents tab to find links to literature

Warranty

The ZOE Fluorescent Cell Imager and associated accessories are covered by a standard Bio-Rad warranty. Contact your local Bio-Rad Laboratories office for the details of the warranty.

Safety Use Specifications and Compliance

A **Warning!** label in this manual warns you about sources of injury or harm, including risk of electrical shock.

Warning! Do not attempt to repair or remove the outer case of the ZOE inverted cell imager or other accessories. If you open this instrument, you put yourself at risk for harm to body or equipment from electrical shock.

This instrument has been tested and found to be in compliance with all applicable requirements of the following safety and electromagnetic standards.

Environmental conditions for safe operation of the ZOE Fluorescent Cell Imager.

Transient category	Installation category II
Operating power	100–240 V AC
Frequency	50/60 Hz
Electrical input	24 V DC, 100 W
Installation site	Indoor use only
Operating temperature	10–31°C
Maximum relative humidity	20–80%
Altitude	<2,000 m
Pollution degree	2
Degree of protection	IPX0

Note: Do not store or operate the unit near a sink as contact with water could cause electric shock.

Unit is heavy, do not store or operate it at the edge of a laboratory bench. Unit must be in upright position during operation.

Condensation will form on any object when the temperature of the object is at or below the dew point temperature of the air surrounding the object. If the instrument has been taken from a cold environment to a warmer environment, allow the instrument to equilibrate to above the dew point temperature before operating.

Do not tip the unit over while a sample is located on the sample stage. This could cause accidental spillage of sample into the ZOE Fluorescent Cell Imager's optical and electronic systems, causing electrical shock and rendering unit inoperable.

Do not operate if the glass layer on the LCD screen is broken; this could injure the operator.

To prevent injury from movement of motorized stage, do not put fingers into the stage opening while the unit is powered on.

The angle on LCD screen is adjustable, keep fingers away from the back of the screen when folding down the screen. The ZOE Fluorescent Cell Imager is factory calibrated; no further calibration is needed.

Do not connect the ZOE Fluorescent Cell Imager to a PC, it may render the device's operating system unusable.

Safety Compliance

This instrument has been tested and found to be in compliance with all applicable requirements of the following safety and electromagnetic standards:

- EN 61010-1:2010 — Electrical Equipment for Measurement, Control, and Laboratory Use
- UL Std No. 61010-1:2012 — Electrical Equipment for Measurement, Control, and Laboratory Use, Part 1: General Requirements
- CAN/CSA C22.2 No. 61010-1-12 — Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use, Part 1: General Requirements
- IEC 61010-1:2010 — Safety Requirements for Electrical Equipment for Measurement, Control, and Laboratory Use, Part 1 General Requirements
- IEC62471 Photobiological Safety of Lamps and Lamp Systems
- EN62471 Photobiological Safety of Lamps and Lamp Systems

Electromagnetic Compatibility (EMC)

- F.C.C. Title 47 Part 15B as a Class A digital device
- IEC/EN61326-1 Class A Electrical Equipment for Measurement, Control, and Laboratory Use — General Requirements

FCC Warning and Notes

- **Warning:** Changes or modifications to this unit not expressly approved by the party responsible for compliance could void the user's authority to operate the equipment
- **Note:** This equipment has been tested and found to comply with the limits for a Class A digital device, pursuant to part 15 of the FCC Rules. These limits are designed to provide reasonable protection against harmful interference when the equipment is operated in a commercial environment. This equipment generates, uses, and can radiate radio frequency energy and, if not installed and used in accordance with the instruction manual, may cause harmful interference to radio communications. Operation of this equipment in a residential area is likely to cause harmful interference, in which case the user will be required to correct the interference at the user's own expense
- **Note regarding FCC compliance:** Although this design of instrument has been tested and found to comply with Part 15, Subpart B, of the FCC Rules for a Class A digital device, please note that this compliance is voluntary, as the instrument qualifies as an "exempted device" under 47 CFR 15.103(c) in regard to the cited FCC regulations in effect at the time of manufacture
- **Note regarding Canadian EMC compliance:** Le present appareil numerique n'emet pas de bruits radioelectrique depassant les limites applicables aux appareils numeriques de classe A prescrites dans le reglement sur le brouillage radioelectrique edicte par le Ministere des Communications du Canada

Canada Warning and Notes

This ISM device complies with Canadian ICES-001.
Cet appareil ISM est conforme à la norme NMB-001 du Canada.

This Bio-Rad instrument is designed and certified to meet EN61010* and the EMC requirements of EN61326 (for Class A) safety standards. Certified products are safe to use when operated in accordance with the instruction manual. This instrument should not be modified or altered in any way. Alteration of this instrument will cause the following results:

- Void the manufacturer's warranty
- Void the EN61010 safety certification
- Create a potential safety hazard

Bio-Rad Laboratories is not responsible for any injury or damage caused by the use of this instrument for purposes other than those for which it is intended, or by modifications of the instrument not performed by Bio-Rad Laboratories or an authorized agent. We strongly recommend that you follow the safety specifications listed in this section and throughout this manual. Use only the supplied power cord in the instrument, making sure to choose the plug adaptor that corresponds to the electrical outlets in your region.

Table of Contents

1 Introduction

The ZOE Fluorescent Cell Imager is an inverted imaging system with brightfield and three fluorescent channels (emitting in blue, green, and red color), suitable for routine cell culture and imaging applications. All channels are fully integrated and optimized for most commonly used fluorescent proteins and dyes; no calibration or installation is needed. It is a stand-alone instrument and a PC is not needed to operate it.

The intuitive touch screen allows researchers to visualize their samples and capture cell images with the integrated digital complementary metal-oxide semiconductor (CMOS) camera. Images stored in the internal memory can be edited, overlaid into multicolor merges, and/or exported to a USB flash drive using one of the two USB ports. The integrated light shield removes the need for a dark room and allows visualization of fluorescence in ambient light on the bench, where researchers work with cells. The screen viewing angle can be adjusted easily and optimized to a user's body height.

Each LED light source provides thousands of hours of reliable illumination that are instantly ready after power on. They provide cool, even, and continuous illumination; their brightness can be adjusted by the user to reduce sample photobleaching.

Focusing is conducted manually using coaxial coarse/fine focus knobs located on the instrument base in an ergonomically deduced position, thus minimizing the strain on the user's hands.

The ZOE Cell Imager's 20x lens is mounted in a proprietary manner that results in a large field of view, one that is approximately equivalent to that of a 4x objective lens (0.70 mm^2). Using the pinch-to-zoom gesture, researchers can instantly change magnification while retaining resolution of 1 µm.

Thanks to the large field of view and a motorized stage (up to 6 mm of travel), a large sample area can be visualized rapidly. This attribute is useful when assessing transfection efficiency or cell confluency. The direction and speed of the stage's movement are controlled through the touch screen.

Instrument Overview

The front panel of the ZOE cell imager (Figure 1) includes:

- **Screen** — 10.1", high-resolution, color LCD touch screen with antiglare and antifingerprint coatings; screen angle can be adjusted to optimize viewing experience
- **Light shield** — allows use of fluorescent channels in ambient light
- **Sample stage** — positioned on top of the instrument base, sample stage is large enough to fit any of the commonly used cell culture vessels
- **Objective opening** — located in the center of the sample stage, objective lens is below the opening
- **USB port** — for connecting USB flash drive
- **Indicator light** — glows green when instrument is powered on. If light is not on, instrument is powered off

The right side panel (Figure 1) includes:

- **Power switch** — for turning the instrument on and off
- **Focus knobs** — for manually adjusting image focus; outer knob adjusts coarse focus, inner knob adjusts fine focus

The left side panel includes:

- **Focus knobs** — manual-adjust image focus; outer knob adjusts coarse focus, inner knob adjusts fine focus

The rear panel (Figure 1, inset) includes:

- **USB port** — for connecting USB flash drive
- **HDMI port** — for connecting a projector or external monitor via HDMI cable
- **Power inlet** — for connecting the imager to an electrical outlet via the supplied power cord (select the cable with appropriate plug adaptor for your region)

Note: Hold the power cord with its flat side facing down; push into the power inlet without blocking the locking mechanism. To remove, pull the locking mechanism to unlock the cable. Pulling the cable without releasing the locking mechanism can result in instrument damage.

Graphical User Interface (GUI)

The touch screen commands include four functions:

- **Position** — select the target sample area for cell imaging; view of the entire vessel is shown on the LCD screen with the center of the crosshair located above the objective lens (Figure 2)
- **Live** — view samples using brightfield (Figure 3) or any of the three fluorescent channels; use this mode for image acquisition
- **Gallery** — for accessing captured images stored in the internal memory
- **Merge** — for creating multicolor image overlays

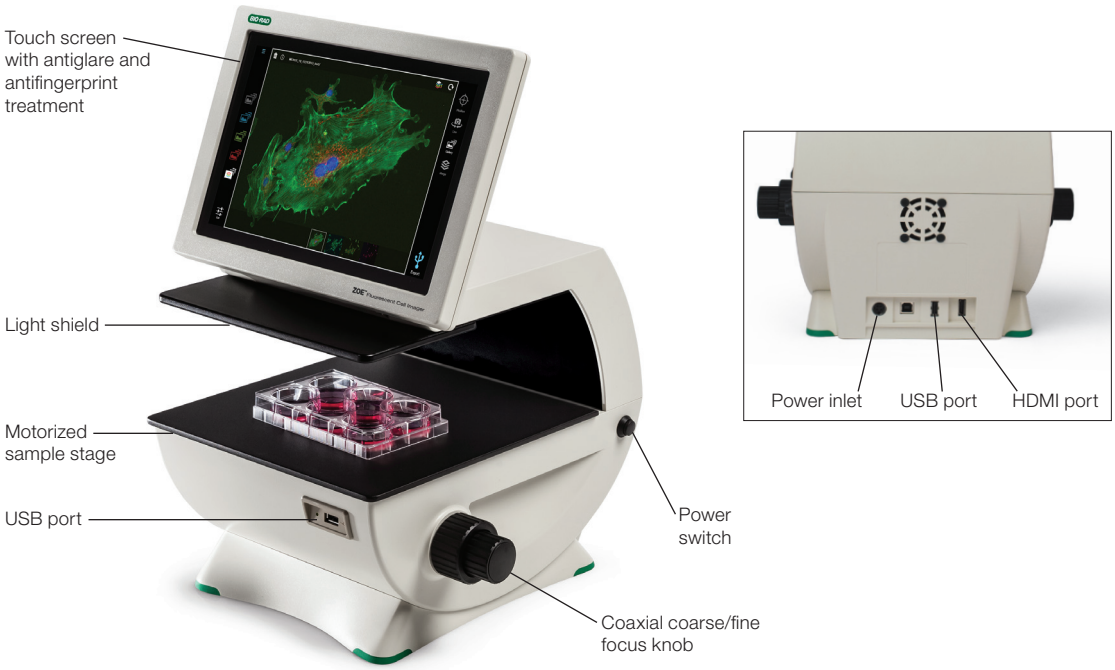


Fig. 1. Elements of the ZOE Fluorescent Cell Imager. Though not shown in full instrument image, both left and right sides have focus knobs. (They can be seen in inset image of the back of the instrument.)

Specifications

The specifications for the ZOE Fluorescent Cell Imager are shown in Table 1.

Table 1. ZOE Fluorescent Cell Imager specifications.

Color channels	Brightfield*, blue, green, and red fluorescence channels	
Illumination	UV LED for blue channel Blue LED for green channel Green LED for red channel	
Excitation	Blue channel Green channel Red channel	355/40 nm 480/17 nm 556/20 nm
Emission	Blue channel Green channel Red channel	433/36 nm 517/23 nm 615/61 nm
Numerical aperture	0.40	
Working distance	7 mm	
Objective	20x plan achromatic	
Display magnification	175x	
Digital zoom magnification	700x	
Motorized stage	6 mm travel in X, Y direction, touch screen control of travel speed and direction	

continues

Table 1. ZOE Fluorescent Cell Imager specifications. (cont.)

FOV	0.70 mm^2
Camera	Monochrome, 12 bit CMOS, 5 megapixels
Image format	JPEG, TIFF, RAW (8 and 12-bit)
Internal memory	16 GB
Focus	Coarse and fine, manual adjustment
Stand-alone instrument	Yes
LCD monitor	10.1", color, LCD, touch screen with antiglare and antifingerprint treatment, 1,280 x 800 pixels image resolution, 80–180° angle tilt range
Ports	2 USB ports
Dimensions	33 x 32 x 30 cm (13 x 12.6 x 11.6")
Weight	9 kg (19.7 lb)

* Brightfield channel is illuminated with green LEDs that improve image contrast by reducing chromatic aberration. Photobleaching due to this green light source is comparable to that encountered with a regular white light.

Note: User interface controls for individual channels are labeled according to the emission colors (blue, green, and red).

System Components

Catalog #145-0031 includes:

- ZOE Fluorescent Cell Imager
- Power cord
- USB flash drive
- Instruction manual
- Quick guide

Unpacking the System Components

1. Unpack the ZOE Cell Imager carefully. Remove all packaging materials and store them for future use. Examine the instrument carefully for any damage incurred during transit. Ensure that all parts of the instrument listed above are included with the product. If any item is missing or damaged, contact your local Bio-Rad office.
2. Place the imager in an upright position on a dry, level surface.
3. Thanks to its light shield, the ZOE Cell Imager can be used in ambient light; dark room operation is not required. However, it is not designed to be operated in direct sunlight.
4. Insert the supplied power cord into the instrument with its flat side facing down and without blocking the locking jacket.

Note: To remove, pull the locking jacket to unlock the cable. Pulling the cable without unlocking this mechanism can result in instrument breakage.

5. Plug the power cord into the appropriate electrical outlet and the instrument will power on. The loading screen displays. After the loading sequence is completed the Position mode will appear onscreen.

Note: Turning the unit off by pressing the Power off button results in only partial shutoff. For complete power off remove power cord from electrical outlet.

2 Setting Up the Instrument

Menus on the touch screen can be customized. To change the default instrument settings, tap the icon located in the upper left corner of the screen (Figure 2) and select Settings.

1. In the Settings window that opens, tap the option you want to modify.
2. Make the desired modifications and tap to exit.

Options on the following menus can be modified.

Camera

The default values for Gain, Exposure time, LED Intensity, and Contrast can be changed. The instrument must be powered off and turned back on for new values to become active.

Preferences

The following options can be modified:

Image File Format — captured images are stored as JPEGs. Alternatives include TIFF and RAW, but the large files of these formats slow down the operating system.

Image File Name Format — default file name format is *Color_Serial number_Time stamp*. Alternatively, *Color_Prefix_Time stamp* allows you to use a custom prefix (for example, Hela).

Add scale bar to captured image — tap **Yes** to add a scale bar to captured images; tap **No** to disable the function.

Attach underlying single color images to merge — tap **Yes** to export underlying single-color images along with the merge file; tap **No** to disable the attach function during export to a USB flash drive.

Automatic export of image files to USB drive — enable automated image export to inserted USB flash drive. Images will be saved to both the USB drive and the internal memory.

Sleep Mode — determine how long the unit should stay fully on after a user interaction. In sleep mode certain functionalities are turned off to preserve power. The ZOE Cell Imager comes fully back on within 20 seconds when the screen is touched.

Date and Time

The date/time stamp is used to track images stored in the internal memory. The date and time should be set before using the imager. Resetting the date/time after the cell imager has been used will not affect already stored images.

System

The following information is available in this menu option:

- Legal information
- Android version
- Kernel version
- Vision board drive version
- Driver board firmware version
- Galileo software version
- Firmware update
- System test posting

Firmware update

To update firmware on the instrument:

1. Insert a USB flash drive with the firmware update file into USB port. The firmware file must be saved in the root directory of the USB flash drive
2. Tap **Firmware update** in the System option of the Settings menu.
3. When the instrument recognizes and validates the firmware file, it proceeds with the update.

Note: If the USB flash drive with firmware file is not inserted, a message prompting you to do so will be displayed. If the update file is not saved in the root directory, a “No update file present.” message displays.

4. Tap **Enter** to continue updating.
5. When the update is successfully completed, the ZOE Cell Imager automatically restarts.

Note: Three types of update files may be available: a Vision board driver update, Driver board firmware, and/or ZOE software.

If both the Vision board driver and ZOE software are updated at the same time, the Vision board driver will be updated first. When that is completed the instrument automatically restarts itself and repeats steps 2–5 to update the ZOE software.

If the Driver board firmware is also present, it and any other update files are automatically updated without repeating steps 2–5.

3 Viewing Cells; Capturing and Processing Cell Images

Tap the appropriate icon, located on the right side of the touch screen (Figure 2), to access the Position, Live, Gallery, or Merge functions.

Position Mode

The unit automatically goes into Position mode when it is powered on. The display area of the touch screen shows the top view of the sample stage. The red crosshair is always over the objective lens (Figure 2) and represents the actual imaging spot. Tap the **Center** icon to return stage to the home position (X, Y; 0, 0) at any time.

Note: If the stage returns to center while you are viewing a sample, the sample viewing area will change. To prevent accidental overlaying of images captured from different locations of the sample, all images previously selected for merge (multicolor images) are cleared whenever the stage is centered.

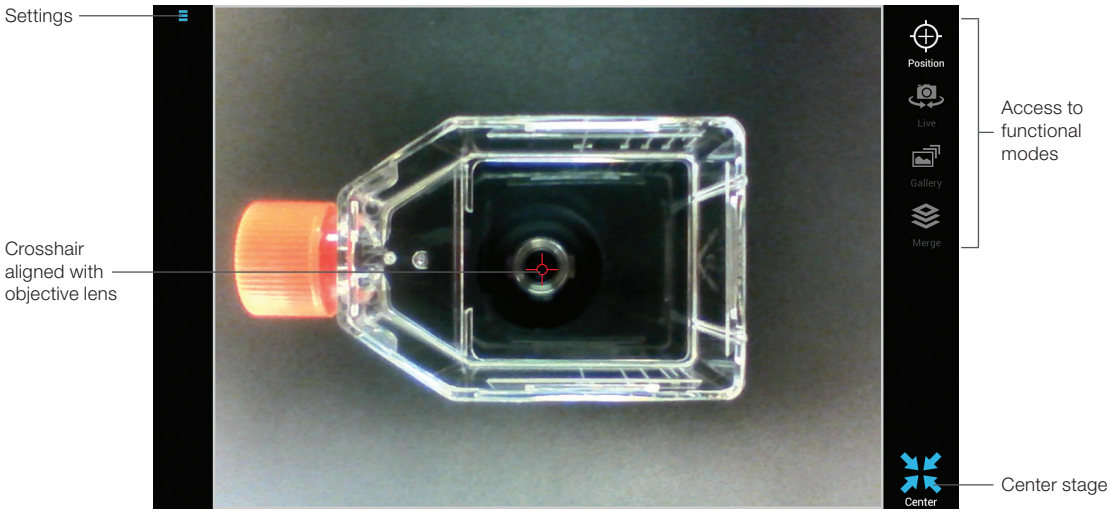


Fig. 2. Elements of the touch screen when the ZOE Fluorescent Cell Imager is powered on. White icon shows imager is in Position mode; other modes are grayed out.

Live Mode

The **Live** function is used for viewing samples illuminated in one of the four channels (brightfield or one of the three fluorescent channels) and for acquiring cell images.

Motorized Stage and Field of View

The ZOE Cell Imager's motorized stage, with up to 6 mm of travel, enables rapid visualization of a large sample area. Direction and speed of the stage movement are controlled through the touch screen, and display magnification can be instantly increased using the pinch-to-zoom gesture. X and Y coordinates (Figure 3) are updated as the stage moves. Use the **Center** icon (Figure 3) to return the stage to the Home position (X,Y: 0,0). Tap the Stage/Zoom lock to lock the sample position and magnification during acquisition of images for multicolor merge. When done, tap the lock again to unlock.

Note: Because you might need to refocus when you switch to a different channel, the Stage/Zoom lock does not lock the focus.

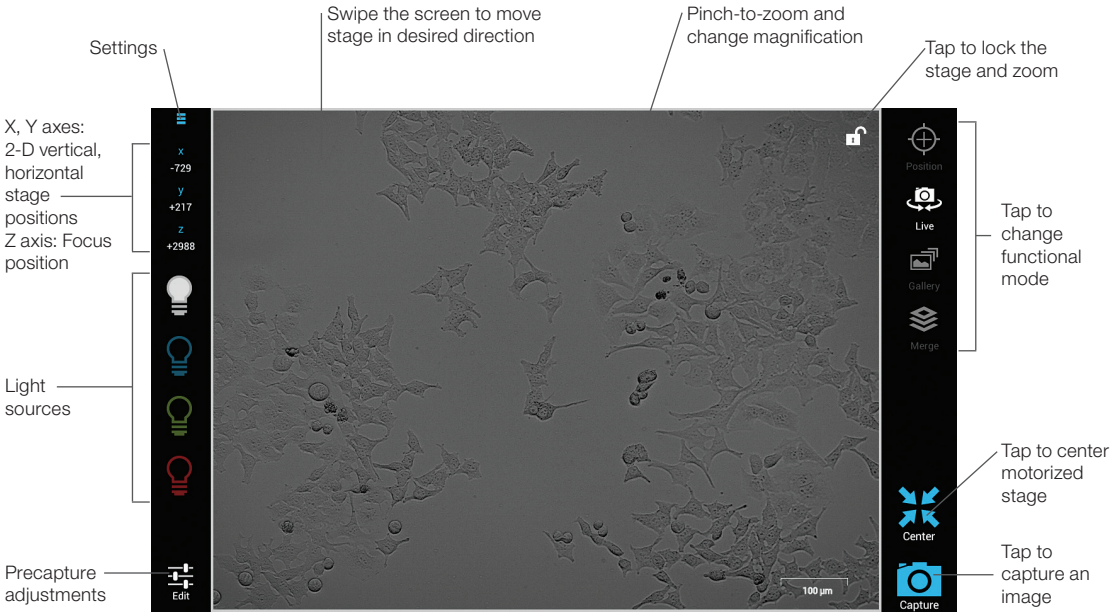


Fig. 3. User controls available in Live mode.

Using the Brightfield Channel

A control panel located on the left side of the screen contains controls for switching on and off light sources for the individual imaging channels (Figure 3). For improved image contrast, a ring of green LEDs is the light source in the brightfield channel. ZOE uses a monochrome camera and the resulting brightfield images are black and white.

Tap the **illumination control icon** (Figure 4) to turn the LED light source on, tap again to turn it off.

Fig. 4. Brightfield channel light source on/off.

Note: Photobleaching due to this green light source is comparable to that of a regular white light. The wavelength used does not cause the red fluorophore to fluoresce or photobleach.

Focus position indicator on the left side of the touch screen shows the Z axis position number. The number is updated during focusing (Figure 3). Knowing the approximate Z number for the tissue culture vessels (Table 2) you use can speed up focusing.

Table 2. Z axis position range of tissue culture vessels.

Vessel Type	Z Axis Position
Microscope slide	720 (± 100)
6-well plate	2,000 (± 100)
12-well plate	2,100 (± 100)
24-well plate	1,900 (± 100)
48-well plate	2,000 (± 100)
96-well plate	3,500 (± 200)
T25 flask	1,800 (± 200)
T75 flask	2,500 (± 200)
T225 tissue culture flask	3,200 (± 200)
Small petri dish	1,200 (± 200)
Large petri dish	1,100 (± 200)

Optimizing Imaging Parameters in Brightfield Channel

Use the Edit menu to adjust these precapture parameters (Figure 5): Gain, LED Intensity, and Contrast values; Exposure time (msec); and quadrant illumination. Drag the slider bar to adjust a parameter. Tap the – and + controls at either end of each slider bar for fine adjustments.

- **When searching for sample** — increase **Gain** value for brighter signal; reduce **Exposure** time for faster frame rate
- **Once the sample is identified and ready to be captured** — lower **Gain** value to reduce background noise; increase **Exposure** time to regain signal intensity
- **To further increase brightness** — start by increasing **LED Intensity** value; follow by increasing **Exposure** time if needed
- **To reduce nonspecific background signal** — increase **Contrast** value

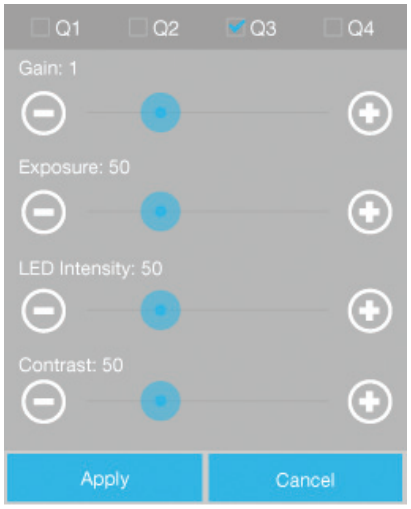


Fig. 5. Edit controls in the brightfield channel.

Improving Image Contrast of Brightfield Channel

A ring of green LEDs is used as a light source in the brightfield channel. Using a green light source reduces chromatic aberration and results in improved image contrast compared to white light.

The **Q1**, **Q2**, **Q3**, and **Q4 checkboxes** at the top of the brightfield channel edit controls panel (Figure 5) toggle quadrants of the ring of LEDs on and off. This control can be used to achieve phase-like image contrast. Lighting only one quadrant shifts the direction of the light, resulting in oblique illumination. The combination of on and off quadrants that provide the best imaging performance depends on the sample.

1. Start by turning off three quadrants, leaving only one quadrant lit. To turn off quadrants, tap the checkboxes and remove the checkmarks. For example, leave on Q2 and turn off Q1, Q3, and Q4.

   Note: If you want to optimize contrast by turning on two quadrants, they should be adjacent quadrants. For example, leave on Q1 and Q2, or Q2 and Q3, etc.
2. The imaging area becomes significantly darker when only one quadrant is lit. To compensate for loss of illumination from the unlit quadrants, increase the Gain slightly and then adjust the LED intensity and, if needed, the Exposure time.
3. Increase the Contrast level as needed.
4. The quadrant that provides the best contrast varies depending on cell culture vessel type and sample. Try turning on a different quadrant than the one you started with and see if this results in better image quality.
5. Tap **Apply** to accept the settings or **Cancel** to exit the dialog box without saving the changes.

Using the Fluorescent Channels

The ZOE Cell Imager has three fluorescent channels: blue, green, and red. Channels are named according to the color they emit.

A control panel located on the left side of the screen (Figure 3) contains controls for switching on and off light sources for the individual imaging channels. The viewing area shows the sample illuminated with the selected light source; the displayed sample is pseudo-colored in the appropriate emission color (Figure 8). Tap the illumination control (Figure 6) to turn the LED light source on, tap it again to turn it off.

Fig. 6. Light source on/off controls. A, Blue channel light source on/off; **B**, green channel light source on/off; **C**, red channel light source on/off.

Focus — position indicator on the left side of the display area shows the Z axis position number and is updated during focusing (Figure 4). Noting the approximate Z number for the tissue culture vessels you use can speed up focusing (Table 2).

Optimizing Imaging Parameters for Fluorescence Imaging

Use the Edit menu to adjust these precapture parameters (Figure 7): Gain, LED Intensity, and Contrast values; Exposure time (msec); and quadrant illumination. Drag the slider bar to adjust a parameter. Tap the – and + controls at either end of each slider bar for fine adjustments.

- **When searching for sample** — increase the **Gain** value for brighter signal; reduce the **Exposure** time to obtain a faster frame rate
- **Once the sample is identified and ready to be captured** — lower the **Gain** value to reduce the background noise; increase the **Exposure** time to regain signal intensity as needed
- **To further increase brightness** — start by increasing **LED intensity** value; follow by increasing **Exposure** time if needed

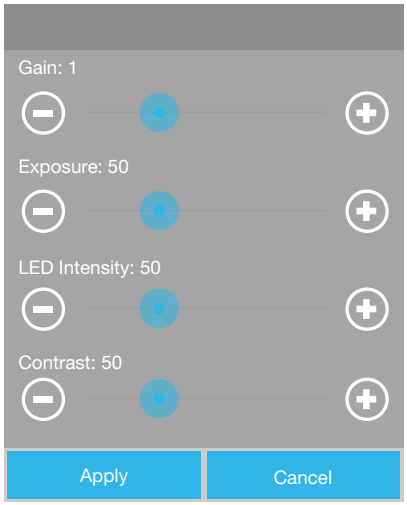


Fig. 7. Edit controls in fluorescent channels.

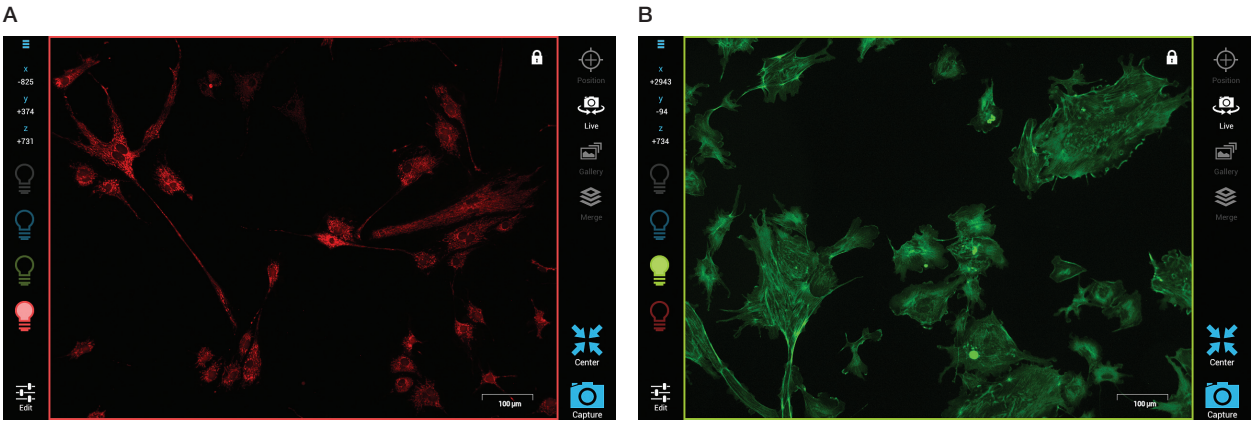


Fig. 8. Samples pseudo-colored in the appropriate emission color in Live mode. A, BPAE cells, mitochondria stained with MitoTracker red CMXRos; **B**, BPAE cells, F-actin stained with Alexa Fluor 488 phalloidin.

Image Capture

To capture the image of the sample displayed in the viewing area, tap the **Capture** (Figure 3) icon. The captured image will remain displayed on the screen (action mode will change from **Live** to **Gallery**) and the image file will automatically be saved to the internal memory. Captured images can be edited or used in multicolor image merges. For more details go to **Capturing Images for Multicolor Merge**.

Gallery Mode

All captured images can be accessed in the **Gallery** mode. They are stored in image folders by color: Brightfield, Blue, Green, or Red. Multicolor merges, created using the captured images, are stored in the Merge folder.

The internal memory can store approximately:

- JPEG ~2,500 files
- TIFF ~1,500 files
- 8-bit RAW ~400 files
- 12-bit RAW ~800 files

Note: Storing smaller numbers of images files (fewer than 1,000 JPEG files) will ensure rapid functioning of the cell imager and will also lengthen the life span of the internal memory.

Viewing Captured Images

Controls for opening the individual folders are located in the control panel at the left side of the screen (Figure 10). To open, tap a folder (Figure 9) and the most recently viewed image from that folder will open. The viewing area will be black if a gallery folder is empty.

Fig. 9. Five image folders are accessible in Gallery mode. A, folder with brightfield images; **B**, folder with images from the blue channel; **C**, folder with images from the green channel; **D,** folder with images from red channel; **E**, folder with multicolor merged images.

Images in every folder are organized according to the time of capture. To move to the next image, swipe the viewing area to the right. Swiping the viewing area to the left opens the previously captured image.

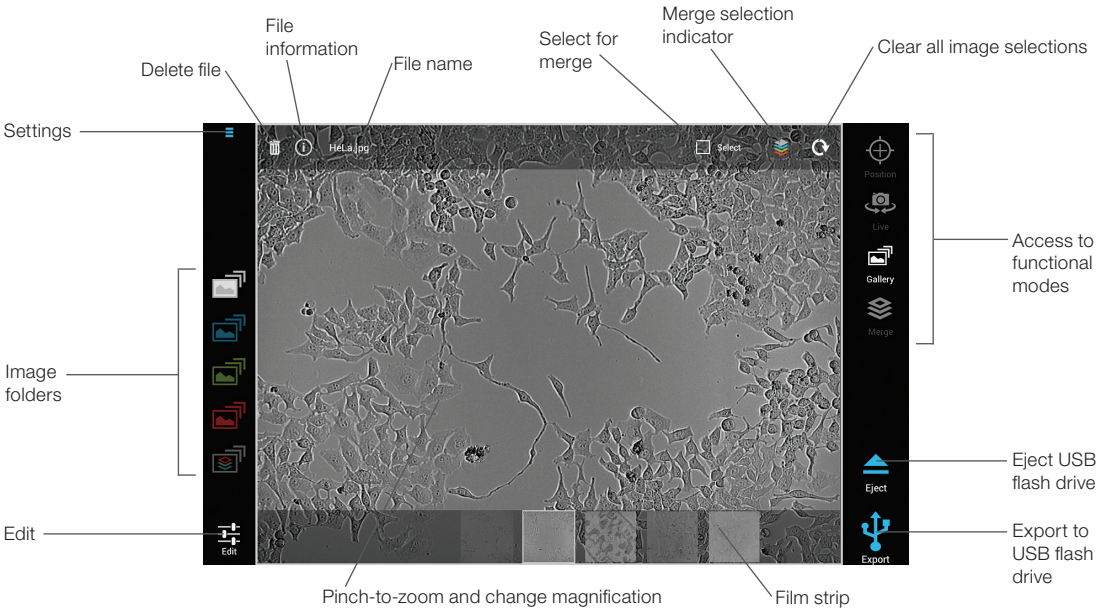


Fig. 10. Gallery mode. Selected folder is illuminated. In this case, the selected folder contains images captured using the brightfield channel.

The **film strip** (Figure 10) of all images stored in a particular folder is located at the bottom part of the screen, tap anywhere in the viewing area to hide it; tap again to bring it back. A thick border surrounds the thumbnail corresponding to the currently opened image. Tapping another window on the film strip will open that image.

Use the pinch-to-zoom gesture to magnify the image to view more details.

Edit — brightness and contrast can be adjusted by dragging the slider bar. For fine adjustments tap the + and – controls.

Image file Information — tapping the ⓘ icon opens a menu with information such as file type and time stamp.

Delete — tapping the 🗑 icon brings up a “Are you sure you want to permanently delete <file name>?” message. Select **OK** to delete or **Cancel** to cancel deletion.

Note: If you delete a merged image, the single-color image files used to create the merge will not be deleted.

File name — the file is automatically named at the time of capture based on the selected naming convention. To change a file name, tap the file name twice. A virtual keyboard is displayed. When new name is typed, tap **Enter** on the virtual keyboard; this hides the keyboard and records the new file name.

Export — tap to export the currently opened image onto the inserted USB flash drive. The export icon is active (blue) only when a USB flash drive is inserted in the USB port. To export all images in the instrument memory, tap ☰ and select **Export all**. To select and export multiple images tap ☰ and select **Image management**. For more information about Image management, see (page 15).

Eject — after inserting a USB flash drive, the Eject icon appears on the screen. When the image export is completed, tap **Eject** to safely remove the USB flash drive.

Image Merge Controls

For more details on creating merged images go to **Capturing Images for Multicolor Merge**.

Select for merge — tap the checkbox icon (Figure 11) to select an image for use in creating a multicolor merge, selected images are automatically uploaded into the Merge mode.

Note: Only one image per color channel can be used. To replace a selected image with a new one, tap the **Select for merge** control in the new image. This automatically overwrites the previous selection in the particular color.

Merge selection indicator — this icon (Figure 12) shows the color of images selected to be used in a merge. Tap the icon to move to the selected image in that particular captured image folder.

Clear – tap to clear all existing merge selections.

Fig. 11. Select for merge control checkbox. A, in an unselected image in the gallery, checkbox is dark (off); **B**, when the image is selected, checkbox turns green, white checkmark appears (on).

Fig. 12. Merge selection indicator showing merged colors. Images in the blue and green galleries are selected.

Delete All Images

To access Delete all images control, tap the Settings icon in the upper left corner of the user interface. To delete all images stored in the internal memory, select **Delete all images**.

Image Management

Image management is used to select image files using criteria such as channel options and time periods. After the image files are selected, you can export or delete them.

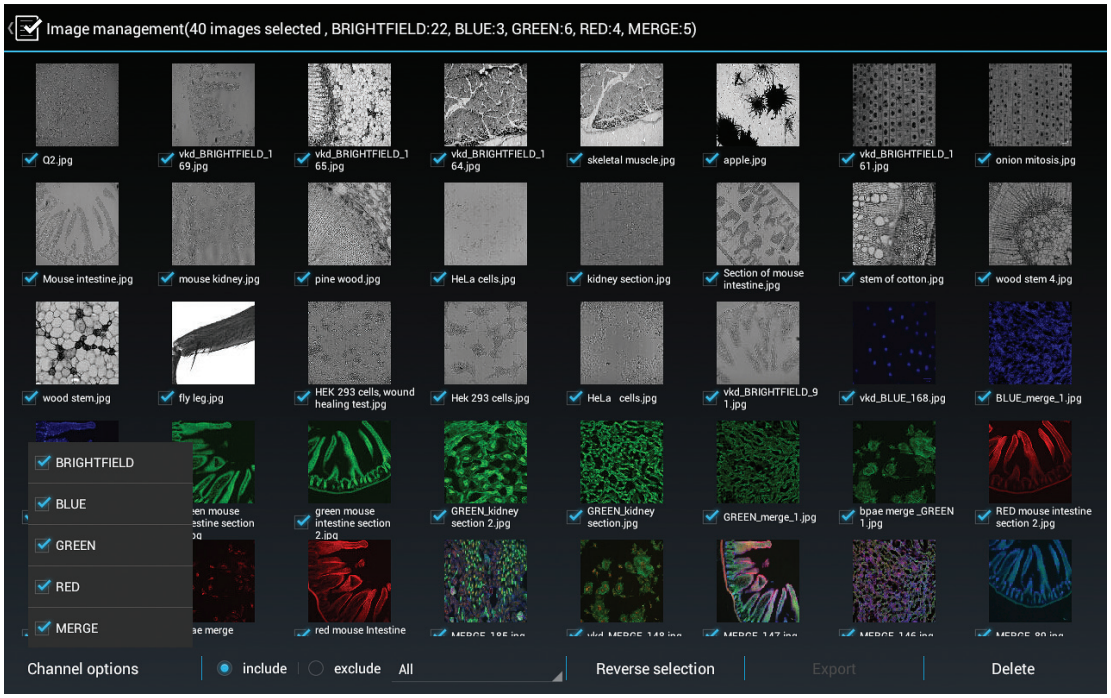


Fig. 13. Image management screen. In this example, channel options have been selected, a time period has been defined, and the Include option chosen. Those parameters determined these 40 images for selection.

Selecting Image Files

To use Image management, tap and select **Image management**. This enables you to simplify image selection by filtering according to:

- Channel options
- Include/exclude option
- Time period options

When Image management displays the image files that match your criteria, a checkmark appears in the checkbox associated with the selected image file. You can clear image file checkboxes manually by tapping the checkmarks.

Channel Options

Choose one or more channel options by tapping **Channel options** and then tapping the checkbox next to the channel option:

- Brightfield
- Blue
- Green
- Red
- Merge

Include/Exclude

Use **include** or **exclude** with the time period option. Tap the **include** radio button to select only image files taken during the time period you select. For example, after you select the channel options, tap the **include** radio button and then tap a time period such as **Last 7 days**. Only image files taken in the last seven days will appear.

Tap the **exclude** radio button to exclude image files that were taken during the time period you select. For example, after you select the channel options, tap the **exclude** radio button and then tap a time period such as **Last 24 hours**. No image files taken in the last 24 hours are selected.

Note: The radio buttons turn blue when active.

Time Period Options

Choose from one of the time periods:

- Last 24 hours
- Last 7 days
- Last 30 days
- Last 3 months
- All

Reverse Selection

Tapping **Reverse selection** reverses your selection — previously selected image files will be unselected and image files previously excluded based on your criteria will now be selected. Checkmarks will reverse accordingly. For example, if, based on the criteria you selected, you have 200 selected image files and three unselected, when you tap **Reverse selection** you will have three selected image files and 200 unselected image files.

Exporting Image Files

You can export one or more image files directly from the Image management area. To export image files, insert a USB flash drive in one of the two USB ports, located at the front and back of the instrument. Select the image files you wish to export and tap **Export**. A message appears warning you to ensure that the USB flash drive remains attached during export. Tap **Continue**. The export may take a few minutes to complete, depending on how many image files you selected. When the image export has completed, tap  and select **Eject USB flash drive** to safely remove the USB flash drive. Wait until the USB flash drive has been unmounted before removing it.

Deleting Image Files

Important! If you delete image files, they are completely removed from the instrument's internal memory.

You can delete one or more image files from the internal memory directly from the Image management area. Select the image files you wish to delete and tap **Delete**. A warning message appears asking if you want to permanently delete all selected images. If you are certain of your selection, click **Continue**.

Merge Mode

In this mode previously selected single-color images are overlaid into a multicolor image (Figure 14). For more details on capturing and selecting images for merge, go to **Capturing Images for Multicolor Merge**.

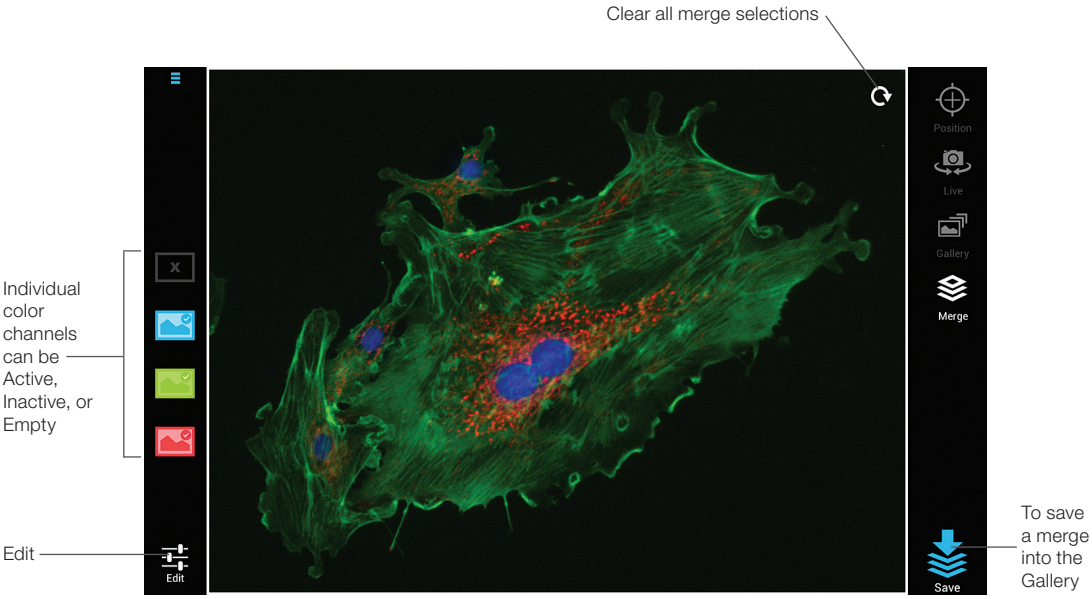


Fig. 14. Merge mode.

Image controls located on the left side of the screen change their appearance depending on whether an image from that color was selected (Figure 15).

Fig. 15. Color channel icons in Merge mode. A, active: color channel with selected image, the image is displayed; **B**, inactive: color channel with selected image, the image is hidden; **C**, empty: color channel for which no image was selected.

Tapping an active channel (image is displayed) hides the particular color from display, tapping it again makes it visible again.

Edit — tap the color for which you wish to adjust brightness and contrast. Drag the slider bar or tap the + and – controls for fine adjustments (Figure 16).

Note: Brightfield images cannot be edited in Merge mode. Any edits to a brightfield image must be made in Gallery mode and saved. Edits made and saved in Gallery mode to an image selected in Merge mode will also display in Merge.

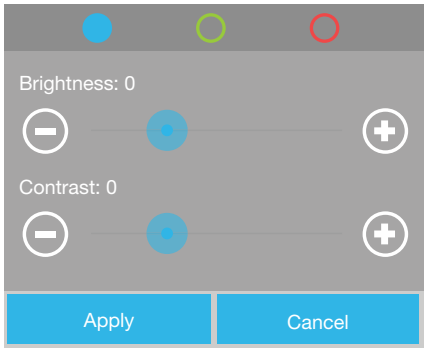


Fig. 16. Edit dialog window in Merge mode.

Save — when satisfied with the multicolor image tap the **Save** icon (Figure 14). The merged image, along with links to the underlying single-color images, is saved to the Merge folder in the Gallery (Figure 10).

Note: When creating a merge, only color channels that are active at the time of saving (Figure 15A) are included. If a color is inactive and thus not displayed (Figure 15B), it will not be included.

Clear — use to clear all selections (Figure 14). This control is also available in Gallery mode.

Note: To empty a single channel, go to the individual image folder in the **Gallery** and tap the active **Select** icon (Figure 11). The icon will go to its off state and the particular color channel in Merge mode will be empty (Figure 15C). To replace an image, return to Gallery mode and tap the **Select** icon in the image you wish to use instead.

Note: When the stage is centered, a different field of view is displayed. To prevent users from accidentally merging images with different fields of view, all image selections for merge are cleared when the stage is centered.

Capturing Images for Multicolor Merge

Using the ZOE Fluorescent Cell Imager's onboard software, images from up to four channels can be overlaid into a multicolor image merge. When capturing images that will be overlaid into a merge, all images should come from the same area of the sample and must be of the same magnification. To prevent inadvertently swiping the screen and moving the stage or changing the magnification, use the Stage/Zoom lock. After you have selected the sample area and level of zoom in the channel you wish to use, tap the Stage/Zoom lock. When done capturing images for the merge, tap the lock icon again to enable stage movement and magnification changes.

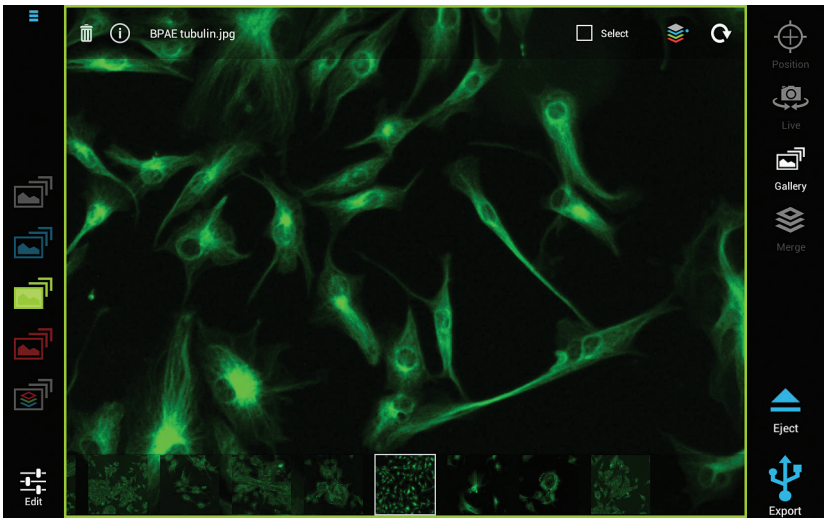
Note: Images of different magnification can not be overlaid; an error message will be displayed.

To capture images for a multicolor merge:

1. Go to Live mode.
2. Turn on the light source you wish to use.
3. Adjust settings in Edit if desired and tap **Capture**.
4. The active mode switches to the Gallery mode and the just-captured image is displayed. If satisfied with the image, tap **Select for merge** (Figure 17A) in the upper right corner of the screen. The selected image is uploaded into the Merge mode**.** The merge selection indicator is automatically updated to show all colors that are currently selected (Figure 17B).
5. Return to the Live mode and repeat steps 3 and 4 for the remaining channels that you wish to use in the merge.
6. Go to the Merge mode where all selected images are automatically overlaid (see section **Merge mode** for more details).
7. Optimize brightness and contrast in Edit if desired. Tap **Save** to save the merge as a single-image file that can be accessed in the Merge folder in Gallery mode. From there it can be exported to a USB flash drive along with the underlying single-color image files.

Note: Only one image per color channel can be used. To replace a selected image with a new one, tap the **Select for merge** control in the new image. This automatically overwrites the previous selection in that color. To clear all previous merge selections tap the **Clear** control.

A



B

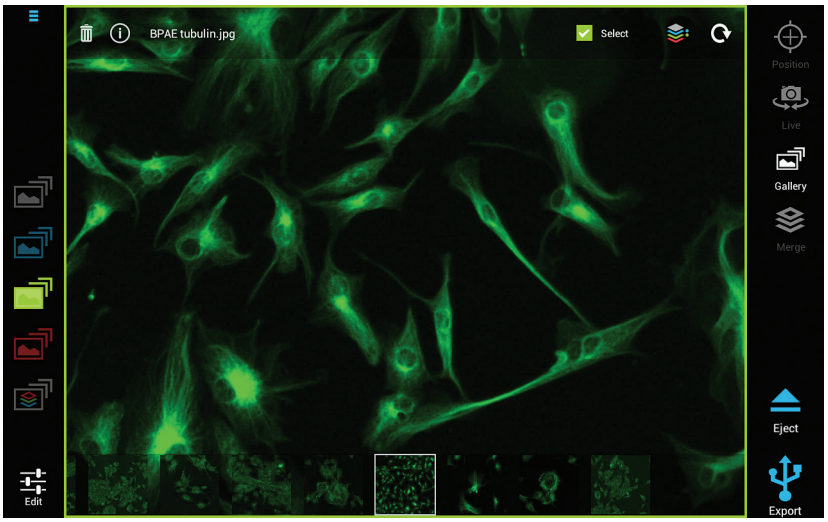


Fig. 17. Selecting an image for merge. A, the displayed file, BPAE tubulin.jpg, file has not been selected for merge. The merge selection indicator (upper right side of screen) shows that an image from the blue folder has been selected for a merge. **B**, the displayed image (BPAE tubulin.jpg) file has now been selected for merge. The merge selection indicator shows that images from both the blue and the green folders have been selected and are uploaded into the merge mode.

4 Maintenance and Troubleshooting

The ZOE™ Fluorescent Cell Imager requires little maintenance for proper operation. However, with long and constant use, the cell imager will need to be cleaned and some other maintenance performed.

Cleaning the ZOE Cell Imager

The instrument should be cleaned on a regular schedule to remove any debris or dirt that might interfere with proper function.

Cleaning the Instrument Body

Always turn off the instrument and disconnect the power cable before cleaning the case. Use a soft, lint-free cloth and deionized water to wipe down the outer case. Avoid wetting the power switch or the power jack while cleaning the case. To **decontaminate the instrument body**, wipe it with a soft, lint-free cloth and 70% alcohol to clean the outer case. These cleaning instructions apply only to the outer case and not to the LCD screen.

Warning! Never pour or spray water or other solutions directly on the instrument. Wet components can cause electrical shock when the instrument is plugged in.

If you use a 10% bleach solution to clean or decontaminate the instrument, it may leave a residue of bleach crystals that over time could scratch the surface. If bleach comes in contact with the LCD screen, wipe down the screen with a damp cloth to remove any traces of bleach.

Do not spill liquids inside the cell imager. Do not overfill cell culture vessels or tip such a vessel when placing it on the sample stage. This could render the cell imager not functional.

Cleaning the LCD Screen

Always turn off the instrument and disconnect the power cable before cleaning the LCD screen. Use a soft, lint-free cloth lightly moistened with 70% isopropyl alcohol. Cleaning the screen with excessive force can damage it. Wipe the screen dry immediately.

Warning! Do not use abrasive detergents or rough material because they may scratch the control panel and display. Do not use bleach or water for cleaning the LCD screen as both leave residues that make the screen appear hazy. If water must be used, it should be distilled water.

Cleaning the Objective Lens

1. Raise the objective lens to its highest focus position.
2. Unplug the instrument.
3. Put a drop of recommended lens cleaner on lens tissues or cotton swab and gently wipe the objective lens clean and dry.

Recommended Lens Cleaning Fluid

- Lens Cleaner, Zeiss, #490133
- Sparkle Optical Lens Cleaner, AmScope, #50104

Recommended Lens tissue

- Commercial Grade Lens Tissue, Edmund Scientific Co., #52105
- Cotton-Tipped Swab Applicators, Edmund Scientific Co., #56926

Warning! Do not put fingers into the stage hole or attempt to clean the objective lens while moving the motorized stage.

Battery

The ZOE Fluorescent Cell Imager uses a 3 V lithium coin cell battery with a 10-year expected lifetime to maintain the clock setting. If the date on the instrument changes without user input, it may be an indication that the battery is getting weak. Should this occur, contact Bio-Rad technical support for assistance with battery replacement.

Warning! Do not attempt to change the battery. Contact the Bio-Rad technical support team.

For the State of California (U.S.) only.

Perchlorate material — special handling may apply. See **www.dtsc.ca.gov/hazardouswaste/perchlorate** for more information.

Perchlorate material: lithium battery contains perchlorate.

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Ordering Information

Catalog #	Description
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145-0031	**ZOE Fluorescent Cell Imager**, 120–240 V, includes instrument, power supply, USB flash drive, instruction manual, quick guide

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Laboratories, Inc.

Life Science Group

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