

iBox[®] Scientia[™]

Small Animal Imaging System

Instruction Manual



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Introduction

The iBox Small Animal Imaging System is designed to automate research with one-touch, pre-set or user-defined PC controls for accurate, repeatable imaging of small animals.

The iBox system incorporates the light tight darkroom, BioLite and matched emission/excitation green fluorescent protein (GFP) and red fluorescent protein (RFP) filter sets.

The darkroom has a UV blocking window, built-in overhead 365nm UV, Visi-Blue™ 480nm and white light, five-position emission filter array, and a software controlled power lift.

The iBox system comes equipped with a warming plate with internal heating elements that keep the surface of the warming plate at a constant 37°C.

The BioLite offers an eight position filter wheel, six position intensity selector, and a direct lighting source using fiber optic bundles to tightly control the output spectrum for consistent, repeatable measurements.

The iBox offers a highly sensitive CCD camera for in vivo applications and supplies real time, live preview images.



**iBox Small Animal Imaging System
shown with the Automated BioLite MultiSpectral Source**

Components

The iBox Imaging System is comprised of the following equipment:

- Darkroom Cabinet
- Automated Filter Wheel with Emission Filters (GFP & RFP)
- CCD Camera and Lens options:
 - OptiChemi™ 615 with either 50mm f/1.2 Fixed or 25mm f/0.95 Fixed Lens
 - BioChemi™ 515 with the 12.5-75mm f/1.2 zoom lens
- Viewer Window
- Motorized Lift
- Epi Illumination
- BioLite MultiSpectral Source with Excitation Filters (GFP & RFP)
- Warming Plate
- VisionWorksLS Software

Check the packing slip for a complete equipment list. The system may additionally include a computer.

Darkroom Cabinet

The iBox Imaging System is manufactured of aluminum construction and fabricated to provide a light-tight chamber.

Darkroom Dimensions: 17.5W x 17.5D x 32H inches (445 x 445 x 813 mm)

Note: Camera cover dimensions additional

Emission and Excitation Filters

Green Florescent Protein (GFP) and Red Fluorescent Protein (RFP) emission and excitation filters are included. The darkroom can accommodate five emission filters in its filter wheel. The BioLite MultiSpectral Source is designed to accommodate up to eight excitation filters.

BioLite™ MultiSpectral Source

The BioLite Automated Multispectral Source is a 150-Watt quartz halogen visible light source designed for use with ferrule fiber optic bundles. It features a closed optical path to tightly control the output spectrum, allowing consistent repeatable measurements with superior signal to noise. The BioLite features a 6 position dimmer permitting variable intensity. The integrated filter wheel accommodates up to 8 excitation filters.

Warming Plate

The warming plate houses internal heating elements which connect inside the iBox and keep the surface of the warming plate at a constant 37°C.

CCD Camera and Lens

The CCD camera, housed in the top of darkroom, generates extremely fast optics and high sensitivity for capturing high resolution images. The camera's motorized lens is controlled by the VisionWorksLS software.

Viewer Window

The viewer in the darkroom door opens for safe viewing of samples. The acrylic window allows viewing without exposure to ultraviolet radiation.

Motorized Lift

The motorized lift, controlled by the software, can be finely adjusted within its 10 inch range.

Epi Illumination

Overhead lighting includes 365nm UV, Visi-Blue 480nm and white light. Lighting is controlled via the software interface.

Minimum Computer Requirements

Processor:	Pentium class, 1.6 GHz or higher
RAM:	2 GB (4 GB Preferred)
Hardware:	Uses 200 MB of hard disk space NOTE: For 21 CFR Part 11 support functionality, the hard drive partition must be formatted with NTFS
Display:	1024 x 768 and 16-bit or better; 24-bit or 32-bit color is strongly recommended
Operating System:	Windows Vista or Newer (32-bit or 64-bit)
Internet Browser:	Microsoft Internet Explorer 6.0 or later
Computer USB Ports:	Computer must come equipped with a minimum of four USB ports; additional peripherals (mouse, keyboard, printer, etc.) may require additional USB ports.

Anesthesia Kit (Optional)

The optional anesthesia system is an optional component designed to safely anesthetize up to five small animals at a time inside the darkroom. The patented non-rebreathing technology safely prevents backflow of gases into the darkroom. Anesthesia units are available with either isoflurane or sevoflurane, as well as with or without oxygen regulators. Contact UVP for ordering information.



Software Installation

Software Installation

Installing VisionWorksLS Software

1. Insert the VisionWorksLS flash drive into the computer.
2. Navigate to the flash drive folder and open the folder.
3. Open the first folder found in the drive and then open the **Installation** folder.
4. Scroll to find the **VisionWorksLSSteup.exe** file and double click to run installation.
5. Click **OK**, **Next**, agree to "I accept terms of licensing agreement", then **Next**. Leave all options in their default settings. Then click **Next**, **Next**, **Install** and finally **Finish**.

Registering the Software

1. Double click the VisionWorksLS software icon on the desktop.
2. To activate the software, registration is required. To immediately activate the software online, choose **On-the-Fly activation**. Activation can also be done **using a smartphone**. Activation can also be done at REG.UVP.com using any other web connected device. A **Config ID** will be needed for this option and can be found by clicking **Next** after selecting **On the fly activation**. It will be displayed under **Configuration ID**.
3. Click **Next** to continue.
4. The **Already have an activation ID** option is useful when reloading the software after receiving an initial activation code.
5. Complete all required information on the form.
6. Fill out the Serial Number located on the box delivered with the flash drive. The number should be four sets of six numbers.
7. Once the form is completed, click on **Get Activation No.** and then click **Activate** once the Activation Number appears in the box.
8. If the computer is not connected to the Internet, click **Offline activation** to register the software. This allows the user to obtain the activation code and enter it at another time.
9. Click **Next** to continue.
10. Click the link provided and complete the form to obtain registration instructions. Click **Finish**.



Installing Camera Drivers

1. The software must be installed for the camera drivers to be installed. Open the VisionWorksLS software suite.
2. Plug the USB cable from the camera into the USB port. At this time, lens controller USB should also be plugged into the computer.
3. The computer should display **Found New Hardware**. The drivers will automatically install.

Note: The installation may cause a warning message stating that the drivers are “not verified by Microsoft”. Select **Continue Anyway** and the drivers will be installed correctly.

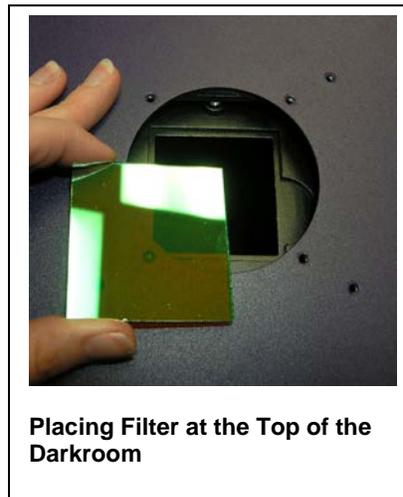
Hardware Installation

Darkroom Setup

1. Remove all the tape on the darkroom (filter wheel, doors, interior cables).
2. Detach the epi-light covers from the Velcro at the top of the darkroom and place the covers in a secure location.
3. Use a level to ensure that the darkroom is flat and make adjustments to the four feet of the darkroom if necessary.

Darkroom Filter Wheel Setup

1. Ensure that the darkroom is in a fixed and permanent location as filters may slide in movement.
2. Locate the filter wheel in the darkroom's camera well at the very top of the darkroom.
3. Each filter location in the darkroom is numbered for a corresponding filter. By touching the sides of the filters only, insert the filters into the proper location. (The numbers on the side of the filter facing you and right side up.)
 - First position #1 = Leave Empty
 - Second position #2 = GFP filter (Rectangular filter, 503nm-523nm)
 - Third position #3 = RFP filter (Rectangular filter, 580nm-630nm)
 - Fourth position #4 = User Choice (filter provided if ordered separately)
 - Fifth position #5 = User Choice (filter provided if ordered separately)



Cameras

High sensitivity, scientific grade CCD cameras are designed for use with UVP's iBox Scientia Imaging Systems. This section covers the components and steps required to install and run UVP's cameras. Every camera has slightly different installation instructions. The following cameras are covered in this manual:

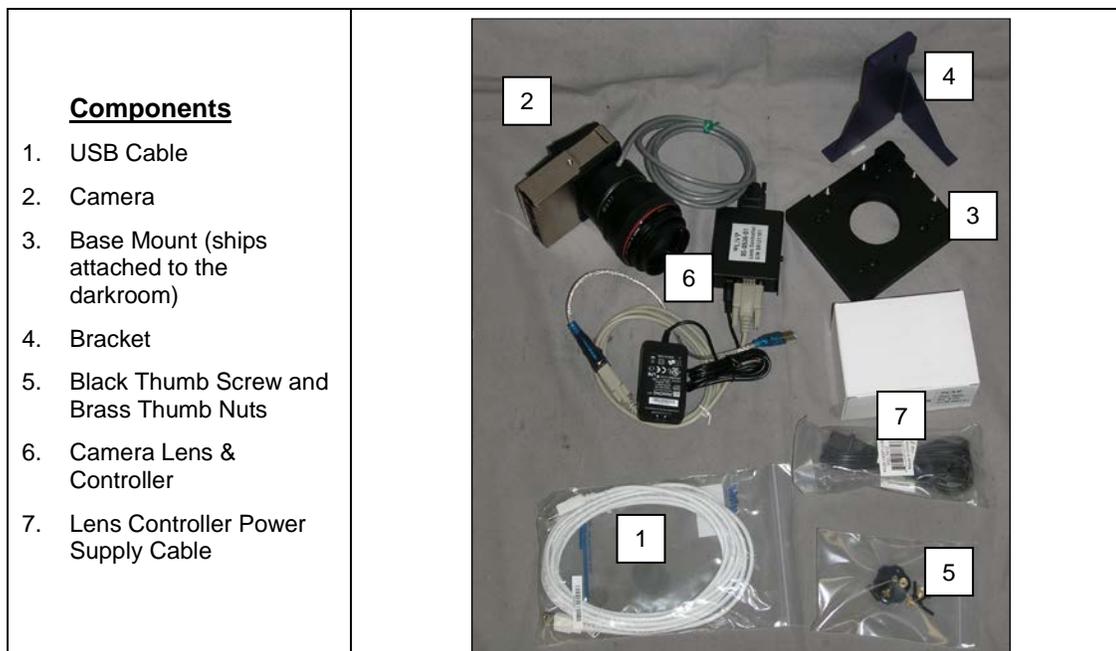
- BioChemi 515
- OptiChemi 615

Refer to the packing list for the camera included with the system.

Components

Note: The BioChemi 500 and OptiChemi 615 cameras use many of the same components and are installed in a similar manner. Therefore, the installation instructions herein will refer to both cameras interchangeably.

Note: Some components may not be shipped with all systems.



Hardware Installation

The photograph shows the parts required for assembly of the BioChemi 515 and OptiChemi 615 camera kits with the automated lens on the iBox Scientia Imaging System. The BioChemi 500 and OptiChemi 615 cameras will have a different appearance but will be installed in the same manner.

Note: The image above shows the 50mm f/1.2 lens (6). Another lens and/or lens controller may be shipped with the system.

1. Attach the camera bracket (4) to the base mount with the brass thumb nuts (5).
2. If not already removed, take the lens cap off the lens. Do not unscrew the lens cap, as it will simply pull straight off.
3. Place the camera/lens assembly in the hole in the base mount. Then, use the black thumb screw (5) to secure the camera in place, attaching it through the hole at the top of the bracket (4).

Note: When looking at the darkroom from the front, the BioChemi 500 attaches to the bracket with the Black Thumb Screw at the front of the system facing the user. The USB and power cables will extend from the left of the darkroom.

The OptiChemi 615 cameras attach to the bracket with the Black Thumb Screw at the right of the system. The USB and power cables will extend from the front of the camera, facing the user.

BioLite MultiSpectral Light Source

1. Plug the power cord into the receptacle on the back of the unit and to a power outlet.
2. Connect the USB cable (if applicable) to the unit and leave the other end unplugged. The manual BioLite does not have USB connection.
3. Position the power source to the left of the darkroom.

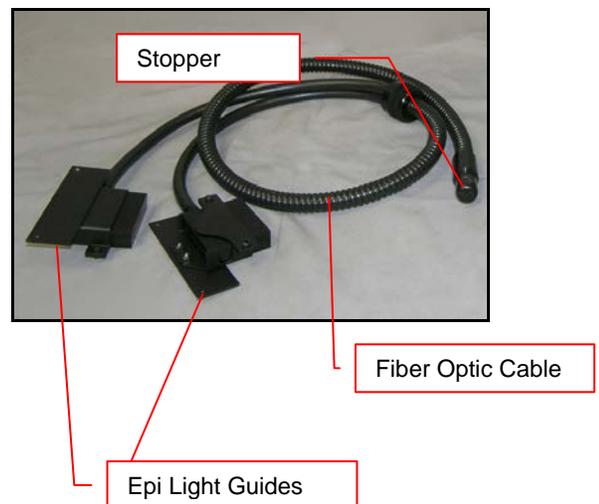


Manual BioLite (Manually Controlled)

1. Insert the filter cartridge into the filter port.
NOTE: Filters and their cartridges are directional. Make sure that the label is facing the front. Because of the intense light of the BioLite, only tempered interference filters should be used.



2. Remove the epi light covers at the top of the darkroom. They should be held in place by two strips of Velcro adhesive. There are two screws that protrude from the epi light fixtures, which will be used to secure the BioLite light guides.
3. Remove the top plug from the access port at the upper left side of the darkroom.
4. Thread the knurled (non-smooth) side of the fiber optic cable from the inside of the darkroom through the access port until the large stopper separating the smooth and rough side of the fiber optic cable can no longer be pushed through the opening.
5. The light guides should be on the inside of the darkroom. Make sure that the fiber optic cables extend from the light guide towards the front of the darkroom (towards you). Match the drilled holes in the light guides to the protruding screws in the epi-light structure. Use the thumbscrews provided to secure the light guides.



6. Insert the fiber optic bundle into the silver connector.



7. Attach the cable to the silver connector with the screw.



Automated BioLite (Software Controlled)

Filters will be installed in their black casing at the UVP factory.

1. To insert the filter/holder into the filter wheel, hold the assembly so that it is positioned vertically with the ridge to the top left as shown in the photo.
2. The filter/holder ridge is then positioned closest to the front of the unit.

Side of
Unit



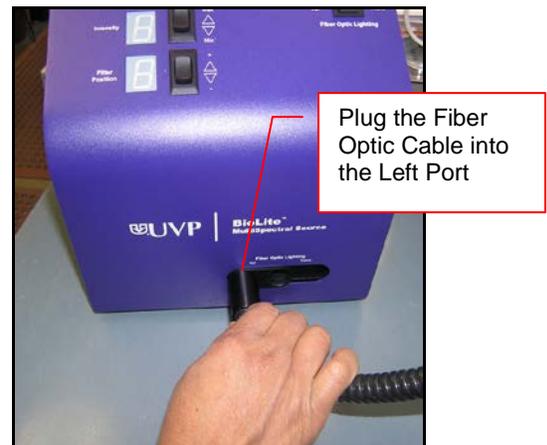
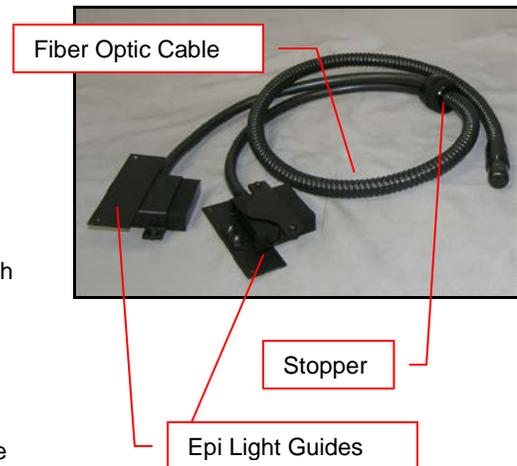
Ridge

3. Slide the holder into the wheel slot.
4. Load the GFP filter in position number one.
5. Load the RFP filter in position number two.

NOTE: Do not force the filter assembly inside the BioLite wheel slot. First check to ensure that the orientation of the filter assembly is correct.



6. Remove the epi light covers at the top of the darkroom. They should be held in place by two strips of Velcro adhesive. There are two screws that protrude from the epi light fixtures, which will be used to secure the BioLite light guides.
7. Remove the top plug from the access port at the upper left side of the darkroom.
8. Thread the knurled (non-smooth) side of the fiber optic cable from the inside of the darkroom through the access port until the large stopper separating the smooth and rough side of the fiber optic cable can no longer be pushed through the opening.
9. The light guides should be on the inside of the darkroom. Make sure that the fiber optic cables extend from the light guide towards the front of the darkroom (towards you). Match the drilled holes in the light guides to the protruding screws in the epi-light structure. Use the thumbscrews provided to secure the light guides.
10. Remove the soft rubber cap from the tip of the fiber optic cable. Plug the cable into the left port in the BioLite unit.



Warming Plate Setup

1. Place the warming plate on the lift platform inside the darkroom. Plug the jumper cord into the **Accessory** outlet located in the upper back panel of the darkroom. Plug the other end into the warming plate.

NOTE: Ensure that the warming plate is plugged into the **Accessory** outlet. This **Accessory** outlet is a live outlet that has a constant supply of power.

2. Ensure that the warming plate on/off switch is turned on and that the warming plate is plugged into the **Accessory** plug of the darkroom.
3. The warming plate will show the temperature of the warming plate in the display. In several minutes, the warming plate will reach 37°C.

NOTE: The warming plate must be turned off or unplugged after use. Software does not control the function of the warming plate.

NOTE: The warming plate has an over limit switch that prevents the unit from exceeding 41°C. Should the unit reach 41°C, the plate must be returned to a temperature of 25°C before the unit reheats.

NOTE: Allow the warming plate to reach room temperature prior to operating. If the unit is extremely cold when switched on, the warming surface temperature can exceed the preset temperature indicated on the LCD display.

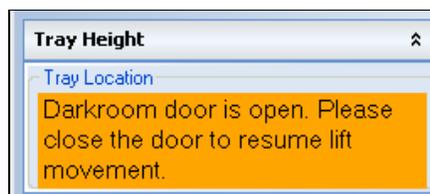
Operation

Using the Software

Caution: Do not place objects under the lift platform as these will suffer damage if lift is moved to the lowest position.

If the darkroom cabinet's automated lift platform is unexpectedly stopped by an obstruction, the software will send a warning message to the user requesting that the user check for the obstruction before raising or lowering the lift platform.

Note: The lift platform will only move if the darkroom door is closed. If the user attempts to change the height of the platform, a warning message from the software will appear in orange stating "Darkroom door is open. Please close the door to resume lift movement".

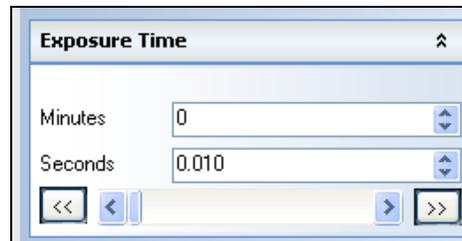
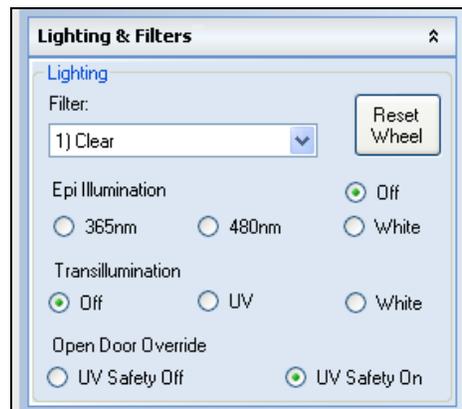
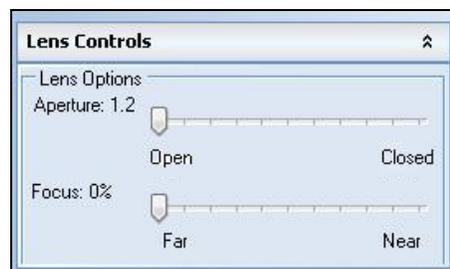


Capture White Light Image of Mouse/Rat

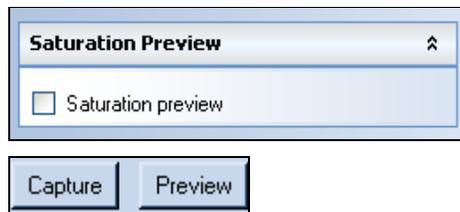
1. The mouse should be under anesthesia to prevent movement.
2. It is highly recommended that the mouse be hairless for imaging. The researcher may remove the hair on the target areas prior to imaging or purchase hairless mice at the beginning of the experiment.
3. Position the mouse on the warming plate so that the fluorescent target is closest to the camera. (For example, brain tumor with the mouse on its feet; pancreatic cancer with mouse on its back)
4. Set the **Aperture** to the smallest number possible (**1.2**).
5. Set the **Focus** so that the image is clear.
6. Turn on the **Epi Illumination White** lights or use the **Automated BioLite** plugin to set the **Intensity** six with no filter inserted into the slot.
7. Move the **Tray Location** to the **Top** position or type in the **Height %** desired. (Example: 50% is halfway up, 100% is at the top position)

NOTE: After setting the lift height, the software will return to the specified height even after the darkroom has been turned off. There is no need to readjust the lift height.

8. Set **Exposure time** to 50msec frame rate.
9. Select **Preview**.
10. If important areas of the mouse are not visible in preview (viewing area is too small), lower the lift.

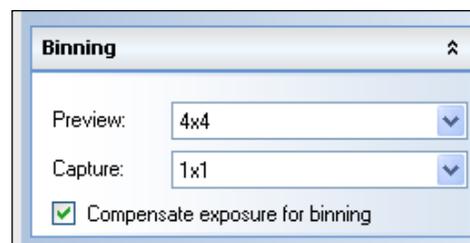


11. After adjusting the lift, ensure that the **Saturation preview** box is checked, adjust the focus and the exposure time so that the preview is not saturated (saturated areas appear red when the Saturation preview box is checked).
12. **Capture** the mouse image. The captured image can be saved to be used later in the **Composite Image** step.



Capture Fluorescent Image of Mouse/Rat

1. Select the appropriate BioLite filter and the Darkroom filter from that corresponds to the fluorescent protein in the mouse. (For example, if the mouse has a GFP fluorescent target, use the GFP BioLite filter and GFP Darkroom filter.)
2. Change the **Exposure time** to 1 second.
3. Ensure that **4X4 binning** is selected for preview and that **1X1 binning** is selected for capture.
4. Ensure that **Compensate exposure for binning** box is checked.
5. **Capture** the image. The captured image can be saved to be used later in the **Composite Image** step



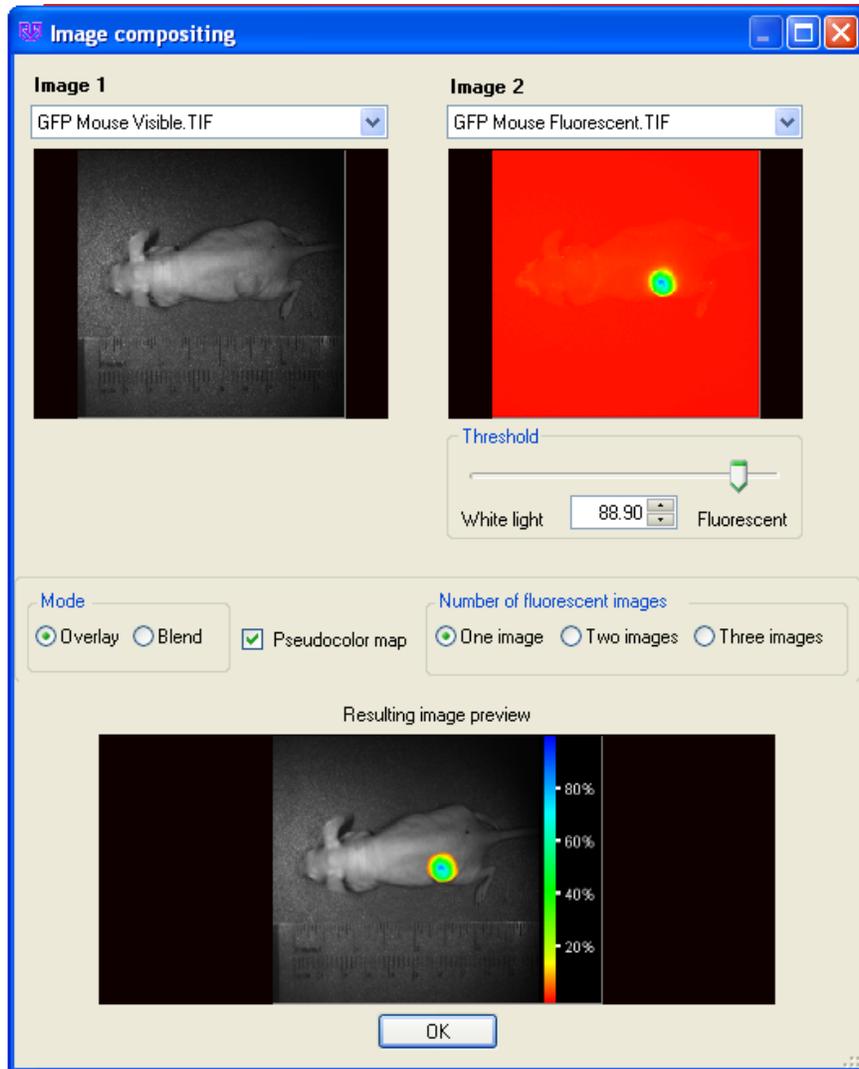
If the target is saturated (is showing red or yellow areas), reduce the exposure time to half [1sec->500msec->250sec...].

If the captured image shows a flat dark image, increase the exposure time in steps of 20 seconds [1sec->20sec->40sec->1min...] until yellow areas appear. Once yellow appears reduce by 20 seconds.

If the captured image shows a bright image looking no different from the white light image, the fluorescent signal might not be strong enough to be observed. (Fluorescent proteins can take 48 hours to express. Implanted, in-depth, fluorescent tissue can take one week to grow to a discernible size. In these two cases, one might just have to wait another day to image again.)

Create a Composite Image of a Mouse/Rat

1. Open the **Image Action Tab** and click onto the **Multi-Image Actions Menu Button**.
2. The **Image Compositing** window appears.
3. Select the number of images to make the composite. One RFP tumor, for example, will need two images to create the composite.—a white light (or visible) image and a fluorescent image. (A Pseudocolor may be added to the fluorescent image to create a colorful Pseudocolor map as shown below.)
4. Use the drop down menu to open the saved image of the white light mouse or rat created using page 16 instructions for capturing a white light image.
5. Use the drop down menu to open the saved image of the florescent mouse or rat created using page 17 instructions for capturing a fluorescent image.
6. Select the **Mode** desired for your application. (Overlay is more pixilated, whereas Blend is smoothed out)
7. Select the **Pseudocolor map** checkbox to view the legend of the colors depicted in the image.
8. Select the desired florescent vs. white light combination by moving the **Threshold** slider for the image.



Service Procedures

Return Procedure

A **Returned Goods Authorization (RGA)** number must be obtained from UVP Customer Service before returning any product.

Replacement Parts and Accessories

Replacement parts and accessories part numbers are shown below. To order accessories or replacement parts, contact UVP's offices listed under Technical Support.

Part Description	Part Number
Bulb, Type EKE 150 Watt Halogen (for BioLite)	34-0088-01
Power Cord, 100V/115V	46-0023-38
Power Cord, 230V	46-0023-39
Thermal printer, digital archive quality 256-grayscale (Mitsubishi)	89-0069-06 (115V)
Thermal printer, digital archive quality 256-grayscale (Mitsubishi)	89-0069-07 (230V)
Thermal printer, digital archive quality 256-grayscale (Sony for Europe)	89-0069-15 (230V)
Thermal paper, Mitsubishi (4 rolls – 800 images)	89-0038-01
Thermal paper, Sony high gloss (5 rolls – 1000 images)	89-0174-01
Thermal paper, Sony glossy (5 rolls – 1000 images)	89-0031-01

Replacement Instructions

Bulb Replacement for the BioLite

The Type EKE 150 watt halogen bulb has a standard life of 200 hours. It must be replaced with the same type bulb only. The UVP part number is 34-0088-01. The replacement procedure is as follows:

1. After allowing the unit sufficient time to cool after last use (at least a half of an hour), unplug the BioLite from the wall. Then, use a wrench to open the bulb access port on the bottom of the BioLite.
2. Once the nut and access port cover have been removed, take out the bulb using the mechanism built into the bulb holder.
3. Grasp the chrome wire to the left of the bulb, and push it in the direction of the base of the bulb.
4. Return the bulb removal mechanism to its original downward, position.
5. Insert the replacement EKE halogen 150 watt bulb, making sure it fully seats.
6. Replace cover and secure with nut before plugging unit back in or turning it on.



Service Procedures

Return Procedure

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Troubleshooting

No Power to the Darkroom

1. Recheck main power cord connections to the darkroom.
2. Check fuses, located at the back of the unit, near the power port. A flat-head screwdriver will be required. Turn the fuseholder cap counterclockwise and the fuse holder will pop out. Inspect the thin wire within the glass fuse to see if there is a break in the wire. If so, replace the fuse(s). If fuses are blowing repeatedly, contact UVP Technical Support for additional troubleshooting.

Technical Support

UVP offers expert technical support on all UVP products. If there are any questions about product use, operation or repair, contact UVP's offices at the locations below.

NOTE: A Returned Goods Authorization (RGA) number must be obtained from UVP's Customer Service prior to returning any product.

If you are in North America, South America, East Asia or Australia:	If you are in Europe, Africa, the Middle East or Western Asia:
Call (800) 452-6788 or (909) 946-3197 , and ask for Technical Support during regular business days, between 7:00 am and 5:00 pm, PST.	Call +44(0) 1223-420022 , and ask for Customer Service during regular business days between 9:00 am and 5:30 pm.
E-mail your message to: info@uvp.com or techsupport@uvp.com	E-mail your message to: uvp@uvp.co.uk
Fax Technical Support at (909) 946-3597	Fax Customer Service at +44(0) 1223-420561
Write to: UVP, LLC. 2066 W. 11 th Street, Upland, CA 91786 USA	Write to: Ultra-Violet Products Ltd. Unit 1, Trinity Hall Farm Estate, Nuffield Road, Cambridge CB4 1TG UK

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