

LS620, LS560, LS460 Microscopes

Operator's Manual

With

Etaluma's Lumaview Software



LS560/LS460 (with Phase Contrast Accessory and Manual XY Stage, both optional and ordered separately)



LS620 (with Phase Contrast Accessory and Manual XY Stage, both optional and ordered separately)

For assistance, please call 760-298-2355 or email support@etaluma.com.

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This document is available for download at <https://etaluma.com/products/downloads>.
Any steps specific for a particular LS model are noted in italics.

I. SETTING UP THE LUMASCOPE MICROSCOPE

A. Items Included With Each LS Microscope

1. LS620 and LS560
 - Brightfield Illuminator (Unless you ordered Phase Accessory)
 - USB power/communication cable
 - Fluorescence shroud (black microplate lid)
 - External power supply/cord with country-specific plug
2. LS460
 - Brightfield
 - USB power/communication cable

B. Optional Accessories (purchased separately)

- Phase Contrast Accessory (Olympus)
- Manual XY Stage
- Labware Holders
 - Holder for 35 mm Petri dishes, fits inside Holder for 60 mm Petri dishes
 - Holder for 60 mm Petri dishes & Terasaki plates, SBS outer dimensions
 - Holder for microscope slides & 50 mm Petri dishes, SBS outer dimensions
 - Holder for 4 microscope slides in parallel, SBS outer dimensions
- Light Hood
- *Lumaquant*, image analysis software
- *Microvolution*, deconvolution software

C. Recommended Computer Specifications

All LS Microscopes with *Lumaquant*: Windows 10; Core i7 or better processor; 500 GB to 1 TB SSD hard drive, 4 Gb minimum RAM(8 Gb min. *Lumaquant*); single 4K monitor or two with 1080p HD resolution. Windows must have Service Pack 2.0 with .NET version 4.5.2.

Microvolution deconvolution plugin for ImageJ/Fiji: A GPU is required; the most powerful NVidia chip running CUDA that can be afforded is recommended for the best performance.

D. Connecting the Cables

1. **Wait to connect the USB cable from the LS Microscope to the computer until Lumaview is installed.**
2. (*LS620 and LS560 only*). For almost all fluorescence imaging applications, power from the computer via the USB cable to the LS Microscope will be sufficient. If higher levels of illumination are required,e.g., with very dim samples or for photo bleaching(FRAP), insert the

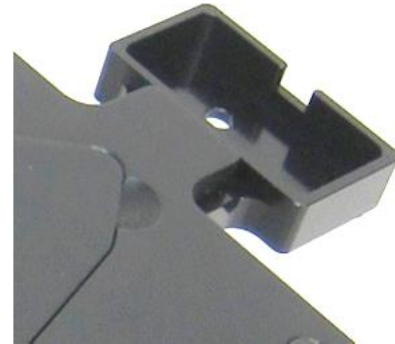
small round plug of the external power supply/cord into the round port labeled **SUPPLEMENTAL POWER** on the back of the LS Microscope and the electrical plug into an AC outlet. You should see an increase in the Illumination level beyond the 40% limit as indicated on the Illumination slider in Lumaview with USB only.

E. Phase Contrast

1. If using the Phase Contrast Accessory, ensure that the Phase Accessory adaptor is attached to the rear tab of the LS Microscope Deck. Attach the Phase Accessory by inserting the base of the arm into the Phase Accessory adaptor. Ensure it is fully seated and tighten the thumb screw on the bottom of the adaptor.



Phase setup attached to Lumascope



Phase accessory adaptor

2. Connect the free end of the Phase Accessory communication cable to the round port on the LS Microscope back panel (left side) labeled **External Lamp Power Out**. This allows the Phase Accessory to be controlled by Lumaview.
3. Insert the phase contrast Slider (pre-centered version included with Phase Accessory) into the slot going through the condenser. Of the 3 positions in the Slider, the phase ring is present in the middle position. A short lever on the front of the phase condenser controls the iris. The iris should be fully open (to the left) for phase contrast.



F. Brightfield

1. A flexible LED light source is provided if no Phase accessory is purchased. This illumination allows direct and many oblique illumination setups.



2. For optimal brightfield, use the Phase Accessory with the included Slider in an open position (no phase ring) and the iris closed down (fully right). The Phase Accessory, which provides collimated light, is directly connected to the LS Microscope, allowing it to be controlled by Lumaview. Transmitted light images will be in gray scale due to the monochromatic CMOS camera.
3. Ambient light in a typically lit area can often be used. The light should be as uniform as possible to prevent shadows or other dark or light areas. Overhead fluorescent lighting can cause ballast interference -- a striped pattern on the image is observed. Turn off the fluorescent lights or move the Lumascope to another room.

G. Installing Objectives

1. Remove the triangular insert in the deck. Unscrew the black cap (or previously installed objective) on top of the inside optics block and screw in the new objective. Do not overtighten. Return the removable insert to the deck.
3. When installing or changing an objective, carry out the insertion quickly to minimize dust falling onto the mirrored dichroic filter. A small amount of dust will not affect imaging. However, if dust is significant and it appears to affect quality of the live image or snapped images, contact Etaluma by email (support@etaluma.com) to discuss possible methods for cleaning. IMPORTANT: Using compressed air can result in an oily liquid being sprayed onto the dichroic, which may damage it.

H. About Lumaview

1. LS Microscopes are controlled by the Lumaview software program. The latest Lumaview version is downloadable from Etaluma's website and must be installed prior to connecting your computer for the first time to the LS Microscope.

2. Lumaview runs on Windows 10. Both desktop computers and laptops can be used, but the best visualization correlates with monitor resolution equal to the sensor resolution (up to 1900 x 1900 pixels for the LS620).
3. Note: The monitor does not affect image resolution unless the monitor is of such low quality that it affects your ability to judge focus.

The computer should have Windows .NET Framework 4.5.2 or higher installed; see <https://msdn.microsoft.com/en-us/library/bb822049%28v=vs.110%29.aspx> for more information. If your computer does not have .NET Framework 4.5.2 or higher installed, downloading Lumaview may automatically take you to the Microsoft .NET download page. Scroll down the page to find the correct download. This will not require a purchase. Contact Etaluma with any questions.

I. Downloading and Installing Lumaview

1. To download Lumaview, go to <https://etaluma.com/products/downloads> (under the Resources tab). Click on Lumaview - ZIP link to start the download and save the folder when prompted. Go to your Downloads folder and click to extract, then open the Lumaview.zip file.

Alternatively, the same Lumaview.zip file can be copied from the flash drive that comes with the Lumascope Microscope. Verify that this file is the latest version as posted on the Etaluma website.

2. If your computer has an older version of Lumaview, installing the newest Lumaview will overwrite the older version but retain all user data and information.

If there is a need to revert to an older version Lumaview, it will be necessary to first uninstall the current version through Windows.

3. To install Lumaview, right click on the .zip file and Open with Windows Explorer. Double click on the .msi installer file to start the installation. If a Windows warning box about an unrecognized App appears, click More... then Run anyway.

You will be asked about the location; note that the default is a new Etaluma folder inside the Program Files (x86) folder.

During installation, a **Device Driver Installation Wizard will open. Click to continue and finish (twice) installing the two drivers.** Installation of Lumaview will then finish. After installation, a Lumaview shortcut (orange circle icon) convenient for launching the software will be present on the desktop.

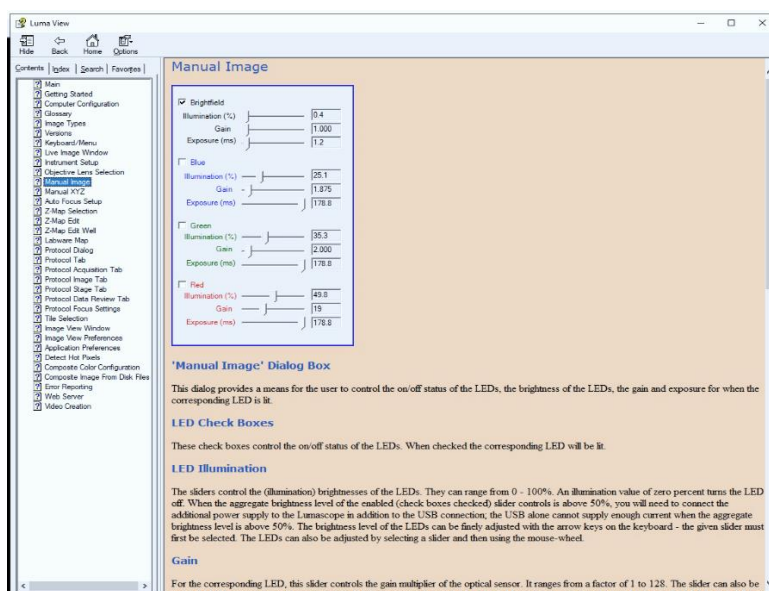
J. Connecting the LS Microscope

1. If connecting with a computer that has been off, turn the computer on. Make sure the computer is connected to its monitor(s). Insert the standard USB-A end of the supplied USB cable into a USB port on your computer and the other square USB-B end into the square port labeled **TO COMPUTER** on the back of the *LS620*, *LS560* and *LS460*. It is important to connect the LS Microscope directly to the computer USB port and not use a USB hub.

- If Lumaview has not been run previously, it is important that the LS Microscope and computer be connected using the USB cable before launching Lumaview. This is because Windows needs to load the USB drivers before Lumaview can run. After the first time, the program can be launched before connecting the LS Microscope and computer. In the latter case, only the Outer Main Window will open. Connecting the LS Microscope and computer with the USB cable will result in opening of the Live Image Window as well.

II. SOFTWARE: GETTING STARTED WITH LUMAVIEW

The following sections give instructions for getting started with Lumaview on the LS620, LS560 and LS460 and are intended as a Startup Manual only. Not all options are given and not all features may be explained in any particular section. The complete User Guide for Lumaview is contained in the Help Section of Lumaview. Click on Help in the Title Bar to open the pull down menu and click Contents, or simply Click F1 to open the Help Section. Becoming familiar with the complete Help Section is a must for using LS Microscopes optimally and taking advantage of the many features of these Microscopes and accompanying software.



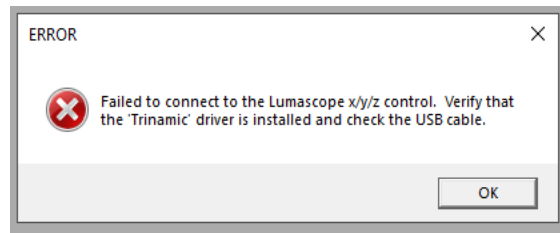
A. Starting Conditions

- Lumaview has been downloaded and installed on the computer.
- The LS Microscope is connected to the computer via the UBS cable.
- Sample to be imaged is present on the LS Microscope stage and objective is installed.
- LS620 or LS560*: Fluorescence shroud (black microplate lid) is over sample if imaging in fluorescence.

B. Launching Lumaview

Launch the Lumaview software from the desktop icon (or other chosen location) and allow the LS Microscope to be discovered. This may take many seconds the first time. Lumaview is installed with the Configuration set to a Lumascope model LS720 by default. The following ERROR message will appear indicating that the software has not found the

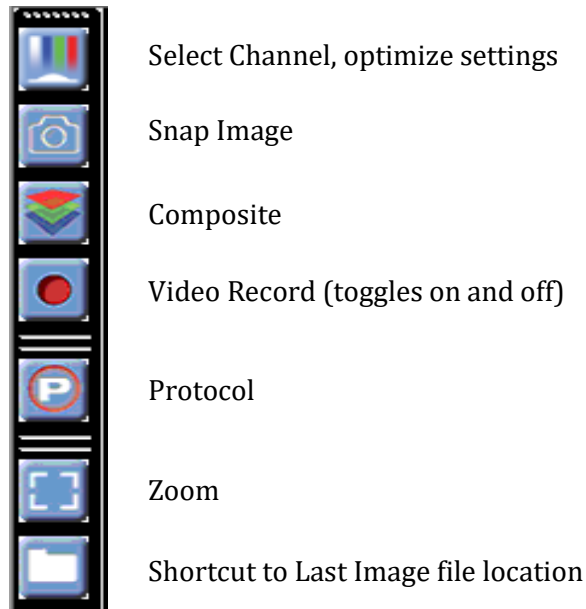
automated LS720 model and thus this will need to be changed to your respective model in the Configuration>Instrument Settings Menu seen below. Click OK, change the model selection and restart Lumaview.



Two windows will open on your monitor: Live Image Window inside the larger Main Window. “Live Image Window” will be in the title bar when the Window is not maximized. When Live Image Window is maximized, [Live Image Window] transfers to the end of the Main Window title bar. The Live Image Window status bar at the bottom shows communication with the camera sensor via continuous display of several useful metrics: current frame rate, data transfer rate, number of frames collected thus far in the session, frame size, and session date and time start.

Note: A variety of Windows configuration errors can be displayed at this point. Please consult Appendix A.

The Live Image Windows includes a Left Toolbar with the following commands:



Select Channel, optimize settings

Snap Image

Composite

Video Record (toggles on and off)

Protocol

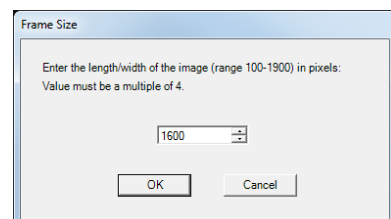
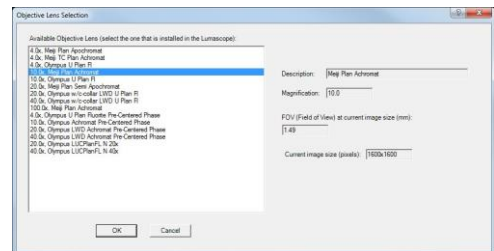
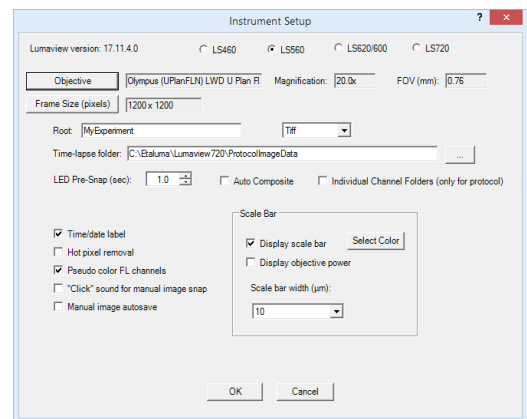
Zoom

Shortcut to Last Image file location

When Lumaview is open, the icon that resides in your task bar appears as a document with an arrow.

C. Instrument Setup

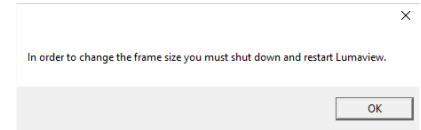
1. Click on Configuration in the File Menu bar to open the drop-down menu and select Instrument Setup to open its dialog box.
2. Check the Lumaview version to make sure it is the most recently posted version on the Etaluma website (www.etaluma.com/products/downloads/). Ensure that the LS Microscope being used is checked. If it is necessary to change the LS Microscope model number that is checked, you will be prompted to restart the program. Be sure to restart so that all functions work correctly.
3. Confirm the current model is selected.
4. Click the Objective button to open its dialog box (called Objective Lens Selection). Select the desired objective to highlight it. Note that other fields about the objective and image automatically fill based on magnification and a frame size of 1200 x 1200 pixels. Click OK. Once selected, the same information automatically fills in Instrument Setup.



5. Click the Frame Size button to open its dialog box. Using the pull-down menu, select frame size desired (frame size will be the number times itself because it is square).

Note: If larger Frame Sizes are used, there will be some darkening in the corners.
If faster frame rates are needed, use smaller frame sizes, e.g., 800 x 800 pixels.)

If the Frame Size is changed, there will be a prompt to restart Lumaview. Click OK to close the prompt and then shut down Lumaview. Upon restarting Lumaview, the frame size in the lower status bar will be the new size. If not restarted, the new frame size may show in Instrument Setup but it may not actually be different. Always check the lower left status bar of Live Image Window to see the actual frame size.



6. Continue to review and adjust the various settings in Instrument Setup. For Root, enter desired file name root for all images in this imaging session. Select the file format desired (Tiff is recommended).
7. The Time-lapse folder path default is given but can be changed if desired. Time-lapse is set up under Protocol as described below. Note that the folder name Lumaview720 is the current default name and is used for images snapped on the other Lumascope Microscopes as well unless a different folder is chosen.

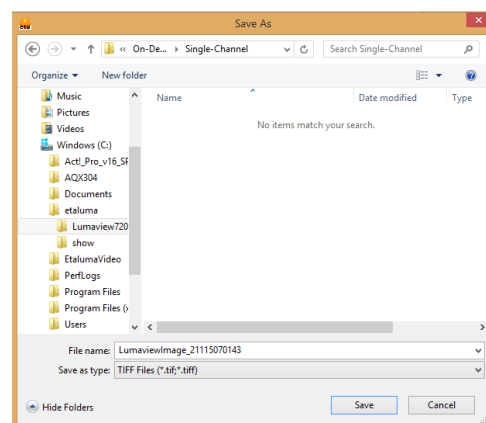
8. Select the time in seconds for each LED to be on before image snap. Recommended time for fluorescence is 0.6 seconds (do not go below this time). Check Auto Composite (*LS720 and LS620 only*) to have images in multiple channels automatically composited and saved in the Composites folder. Check Individual Channel Folders to have images from multiple channels in a Protocol saved in separate folders (e.g., for compiling time-lapse videos in single channels).

Of the next 6 features listed on the left, check those desired:

- Time/date label will be displayed in the lower left corner of each image.
- Hot pixel removal. Check if desired and then complete Instrument Setup. (To start this feature, click Utilities in the title bar and from the pull-down menu select Detect Hot Pixels to open its dialog box. Information on next steps is available in the Help section.)
- Pseudo color FL channels (*all LS Microscopes except LS460*) results in live and snapped fluorescent images colored with the channel color and according to signal intensity.
- “Click” sound for manual image snap results in a “camera shutter-like” sound whenever a manual image is snapped.
- Manual Image Autosave results in images being saved automatically without the Save As dialog box opening each time. When checked, each image is numbered according to DOYHHMMSSmm where DOY is day of year, HH is hour in 24 hour time, MM is minutes, SS is seconds, and mm is milliseconds.

Thus each image number is unique and in numerical order.

Not checking Manual image autosave will result in the Save As dialog box opening every time the camera icon is clicked and the image has been snapped. In this case, the image file name desired must be entered before clicking Save.



- On the right side of Instrument Setup, checking Display scale bar results in a scale bar in the lower right corner of live and snapped images. If checked, click Select Color to choose color, check Display objective power if desired, and select Scale bar width.

After all settings have been selected, click OK to save the settings before exiting Instrument Setup

D. Finding a Live Image

- Install the desired objective and sample labware. Manual XY movement of SBS labware (microplate footprint) is facilitated by Etaluma's Manual XY Stage (optional). For labware that is not SBS, use a labware holder such as the Multi-Slide Holder for 1-4 microplate slides. Remember to update Objective in Instrument Setup as needed and click OK.
- Click on the Manual Image icon in the Left Toolbar (top icon) to open its dialog box. *LS560* dialog box contains only the Green and Brightfield controls; *LS460* dialog box contains only the Brightfield controls. If desired, move dialog boxes to the sides of the Live Image Window. If closed and then reopened, they will open in the same locations prior to closing.

The screenshot shows the 'Manual Image' dialog box with the following settings:

Channel	Mode	Illumination (%)	Gain	Exposure (ms)
Brightfield	Checked	14.9	2.000	178.8
Blue	Unchecked	25.1	1.875	178.8
Green	Unchecked	35.3	2.000	178.8
Red	Unchecked	49.8	19	178.8

- In Manual Image, select the channel to use. For fluorescence, the included Shroud (or other protection from light) must be placed over the labware. Start with **Exposure** at maximum and **Gain** as low as possible. Increase **Illumination** gradually to the desired brightness. For dim samples, if Illumination is at maximum, increasing the Gain can be used to increase signal but it will also increase the background. *LS620 and LS560*: Illumination can also be increased by using the external power supply/cord.
- For brightfield using ambient light, start with Gain as low as possible and adjust Exposure from 0 up, until a mid-gray tone is seen in the Live Image Window.

For brightfield using the Phase Contrast Accessory (use open position in Slider), check Brightfield in Manual Image and move the iris on the front of condenser to the right so only a low level of light is transmitted. Set Gain to minimum, Illumination (%) to 10-20%, and adjust Exposure from 0 up until a mid-gray tone is seen in Live Image Window.

- For phase contrast using the Phase Contrast Accessory (use appropriate Slider phase ring for objective), check Brightfield in Manual Image and turn the phase condenser iris (on front of condenser) to the left to the fully open position. Turn Exposure and Gain to very low. Increase Illumination (%) until a mid-gray tone is seen in the Live Image Window.

E. Focusing

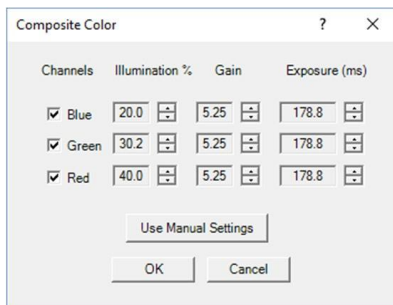
- Unless approximate sample focus level is known, it is best to initially raise objective to maximum height (without hitting sample) and then turn focus knob counter-clockwise until sample comes into focus. Ensure you are focusing on correct sample plane and not another surface, for example, back side of a microscope slide. If uncertain, place a similar but easy to focus sample on stage, focus, and then switch to new sample.
- To facilitate the focusing process, click on the Zoom icon on the Left Tool Bar (second from bottom). The Zoom feature toggles between displaying the entire image fitted to the Live Image Window and displaying the image matched 1:1 with the monitor resolution (pixel: pixel). If the monitor resolution in pixels is less than the frame size, the image will be enlarged (zoomed). The Live Image Window will now show a center portion of the entire image. Click the Zoom icon again to fit the image to the Live Image Window. If necessary, adjust Illumination and/or Gain as needed.

F. Snapping Images

- When the desired field has been illuminated and focused in Live Image Window, click the Camera icon in the Left Tool Bar (third from the top) to snap the image. If the Zoom function was used to focus, the image captured will include the entire field of view (FOV) observable with the Zoom off. If “Click” sound for manual image snap was checked in Instrument Setup, a shutter-like sound occurs when each image is snapped.
- If Manual Image Autosave was checked in Instrument Setup, images will be saved automatically using the time-based numbering system. If Manual Image Autosave was not checked, a Save As dialog box will open after the image was snapped. Images will be saved in the Etaluma folder formed automatically whenever Lumaview is downloaded and installed.
- Images are captured and saved as seen by the camera, i.e., from the bottom.
- To view the last image snapped, click the Folder icon in the Left Tool Bar (bottom icon) to launch Windows Explorer and open the most recently used Destination Folder.

G. Auto-Generation of Single Composite Images (*LS620 only*)

- Click the Composite icon in the Left Tool Bar (fourth from the top) to open its dialog box. Check the channels desired. To use values optimized previously in Manual Image (Section above), click Use Manual Settings to upload the settings. Values can also be entered manually.



- Click OK to start the snapping of individual images in progression. Each LED will turn on for the pre-snap interval and then flash when the image is snapped. If “Click” sound for

manual imagesnap was checked in Instrument Setup, the shutter-like sound will be heard for each image.

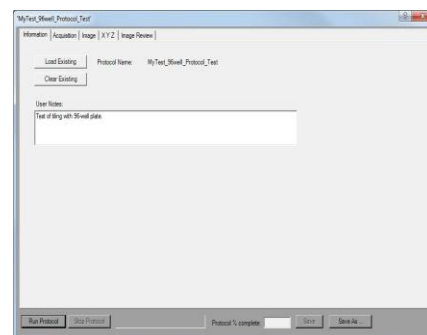
- The fluorescence composite image will appear automatically in the Main Window matched 1:1 with the monitor resolution (pixel:pixel) and with its Title Bar showing the full path where it is located.
- Composites including phase contrast or brightfield (*LS620, LS560*) *Note: this is a hack* ☺
 - In Instrument Setup, make sure Auto Composite is checked.
 - Configure a Protocol with the channels desired. In the Acquisition tab, select Time-Lapse and set both Interval and Duration to 1 sec. Click Run and after the images are snapped, click the Last File icon in the Left Tool Bar (lowest icon). The folder containing the individual images plus the Composite image will open.

H. Live Video

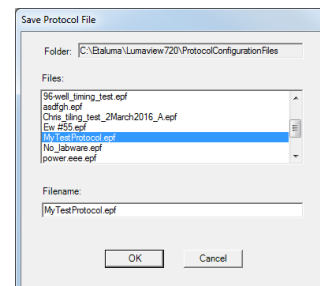
- When a desired field has been illuminated and focused in Live Image Window, click the Video Record icon in the Left Tool Bar (fifth from the top) to open its dialog box. Select parameters requested and also those in Instrument Setup such as file format. Images captured will be saved to the Video folder inside the Manual folder.
- Click OK and image capture will begin. To stop image capture, click the Record Video icon in the Left Tool Bar again.

I. Writing and Using Protocols

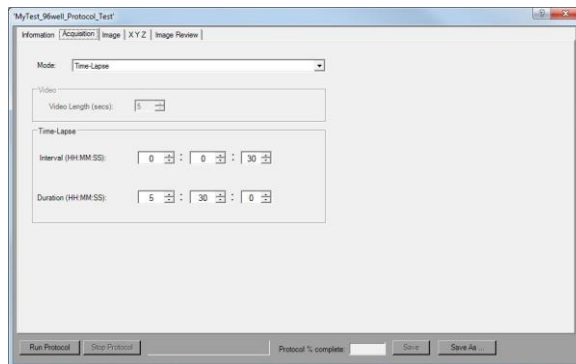
- To configure an automated microscopy run, including time-lapse on the manual LS620, LS560, and LS460, the Protocol feature is utilized. Click the “P” icon in the Left Tool Bar.



- Information and values will be added on the various tabs. Be sure to click Save before leaving any Protocol.
- In the first tab named Information, click the Load Existing button to bring up a previously saved Protocol. If the desired Protocol is present, click OK. Click the Clear Existing button if values have been entered on any of the tabs and saving is not desired or a previous Protocol is displayed and you want to start a new one. If unsaved entries are being cleared, a Save Protocol File dialog box will open asking if you want to save the current Protocol. If Yes, click OK.
- If not previously saved, enter the new desired Protocol name in the Filename field and click OK. Or click on an existing File name to overwrite and a File Overwrite Alert will open; click Yes. You are now ready to create a new Protocol.

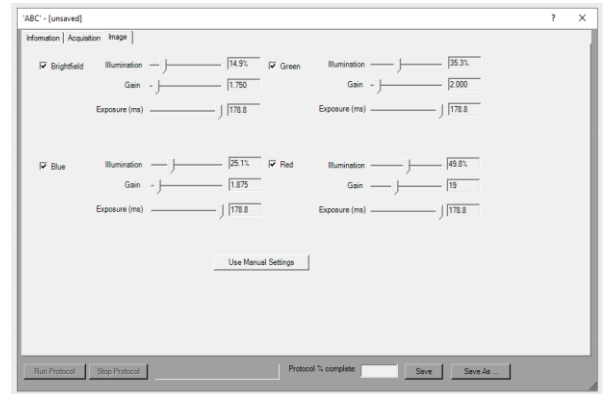


- On the Information tab, if you enter User Notes, they will become part of the new Protocol. You can also enter User Notes for a previously saved Protocol in this tab.
- Click on the Acquisition tab to select Time-Lapse Mode. Enter the Interval and Duration information.



IMPORTANT: Before initiating a time-lapse run, it is critical to change your computer power and Windows update settings so they cannot interfere with scheduled image captures. See Appendix C for details.

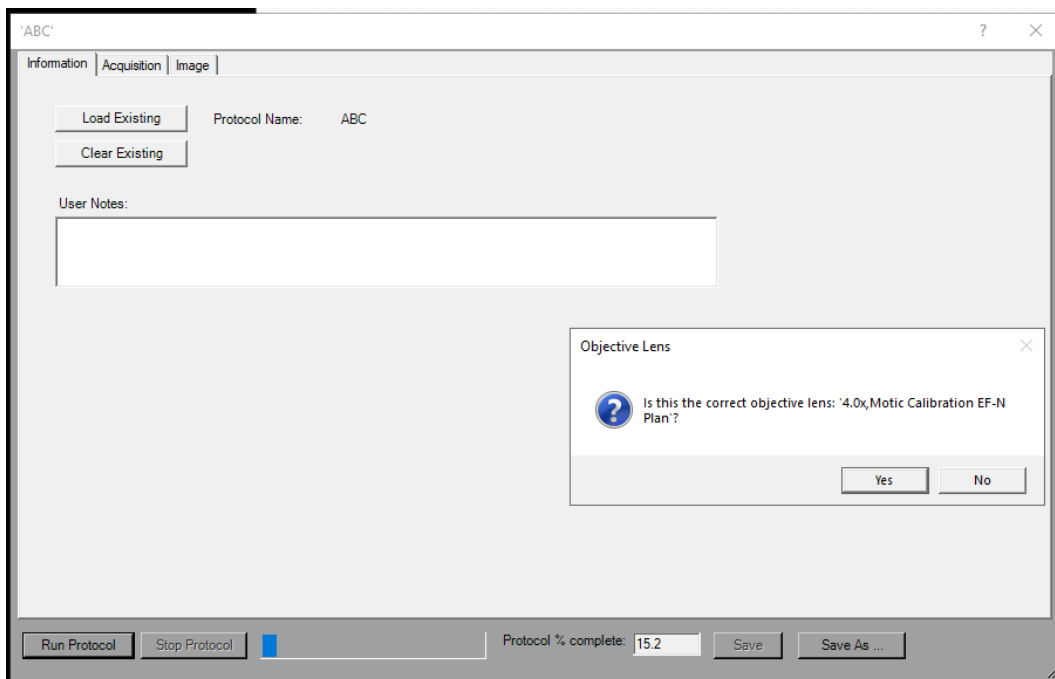
- Click on the Image tab to select the channels to be used in the Protocol and adjust their settings. If settings were previously determined in Manual Image, transfer the values into the current Protocol by clicking the “Use Manual Settings” button. The desired values can also be entered manually. Note that this dialog box cannot be used to optimize the image settings because it is not interactive with Live Image Window.



If optimization of any of the Channel settings has not been done or is still needed, click on the Manual Image icon on the Left Toolbar (top icon) to open its dialog box and fine tune the settings with a representative sample. When settings are satisfactory, click the “Use Manual Settings” button in the Image tab to upload them into the Protocol.

J. Run Protocol

After entering all information and saving the Protocol, it is ready to be run.



K. Placing the Lumascope inside an Incubator

1. Prior to imaging in an incubator, place the Lumascope Microscope inside for at least 6 hours and preferably overnight to allow thermal equilibration. Before transferring to the incubator, disinfect the LS Microscope using any of the suggested procedures in Appendix B. Condensation on the microscope, including on the objective, will form in the first minutes but will evaporate as thermal equilibrium is achieved. Focus levels may change during this equilibration as materials expand.
2. Pass the communication and optional power cable through the incubator port, which is often located in the upper back wall. A plug or filtered stopper may be present. Every LS Microscope comes with a foam plug that can be used to seal around the cable(s) going through the port. Or the existing plug can be adapted. Make sure there is enough slack in the cables inside the incubator for sliding the shelf out. Place the computer to be used on a flat and stable surface near to the incubator and within the 3 M USB cable provided.

WARNING: Do not extend the USB cable or use another USB cable other than that supplied, particularly a longer one, as communication issues will arise.

3. Place the sample on the microscope, adjust focus and close the incubator door. Check the focus after 60 min to confirm it is still good. If the focus has changed, refocus and repeat. For higher magnification objectives (40x and above), a focus drop of a few microns may occur, which can be compensated for by over-focusing to a height above the focal plane equal to the expected drop. In this case, the focus will drop to the correct level within a few minutes.
4. The system is now ready for live cell imaging.

L. Creating Videos from Time-Lapse and Live Video

Several options are available for compiling videos from sets of time-lapse and live video images:

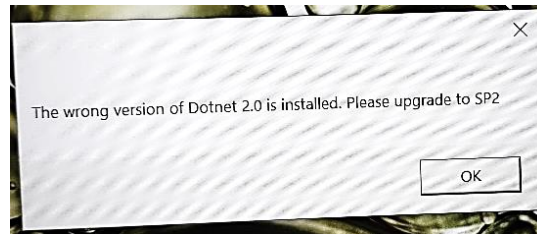
- A) The optional Lumaquant software offered by Etaluma (see Lumaquant Startup Guide for compiling images into video). The Lumaquant Startup Guide can be downloaded from Etaluma's website (www.etaluma.com/products/downloads/).
- B) Other video compiling software such as ImageJ, Fiji, Windows Video Editor, iMovie.

Appendix A

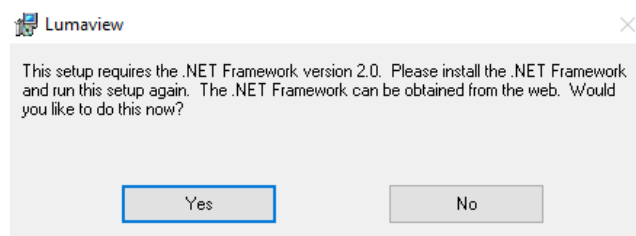
Windows Configuration Troubleshooting and Driver Conflicts (may require Administrator status)

You may encounter installation error messages such as the following:

Dotnet Service Pack 2 Upgrade:



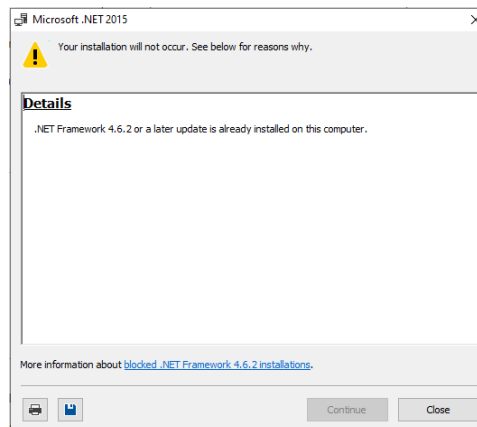
OR



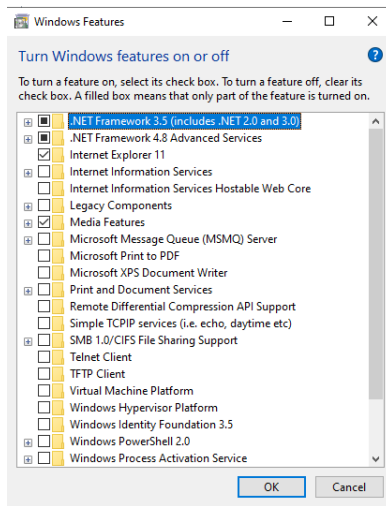
This error which will be presented as one of these two messages and can be solved by going to the Microsoft download page here:

<https://www.microsoft.com/en-us/download/details.aspx?id=21>. Searching for "Dotnet SP2 upgrade download" should yield a Microsoft download page in your country if the above link is inappropriate. After downloading and installing (should be automatic), restart Lumaview to confirm it has been upgraded successfully.

Occasionally the upgrade will already be present but needs to be enabled and you will see this message when you try to upgrade.



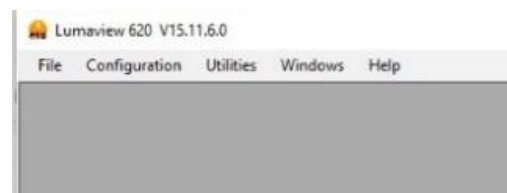
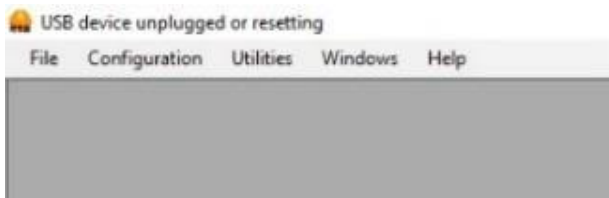
In this case, it must be enabled in Windows features as seen below.



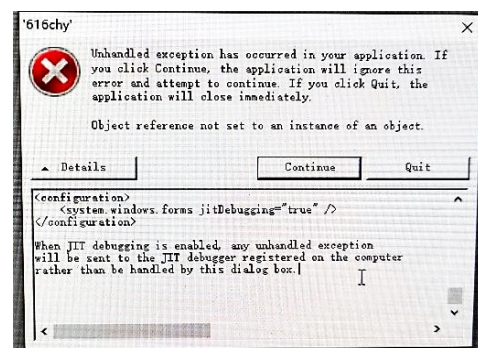
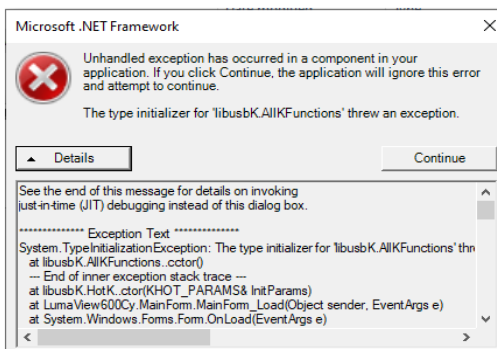
Driver Conflicts

Occasionally conflicts can occur between the drivers used by Lumaview and other Windows drivers, preventing communication between the LS Microscope and the connected computer. Often this occurs after a Windows Update or after installing another instrument software, especially camera-based instruments.

Generally this results in a blank Lumaview screen and one of the following messages in the upper left corner of Lumaview is observed.



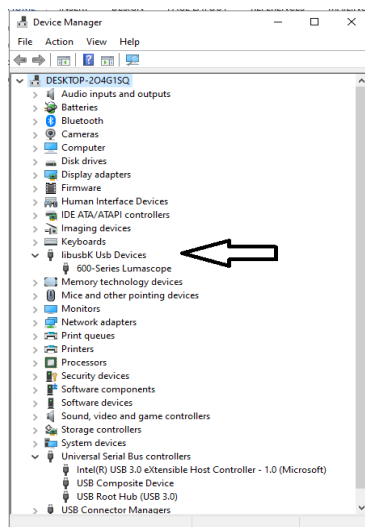
These error messages may also be presented:



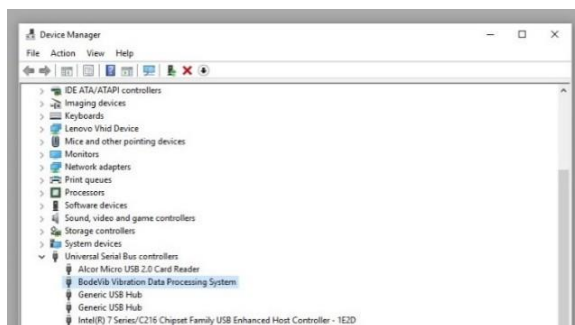
Restarting Lumaview, unplugging and replugging in the USB cable, or restarting the PC and relaunching Lumaview may resolve the issue. If not, removing an interfering driver and updating it with the

Lumascope driver may be required and is described below.

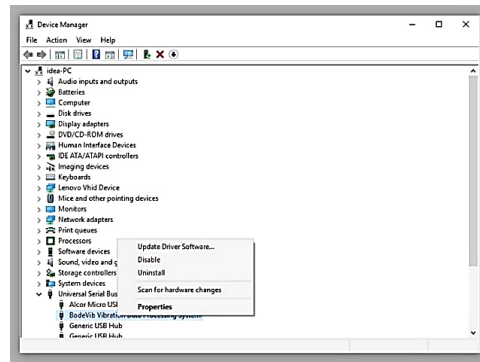
1. Open the Device Manager.
2. With the computer connected to the LS Microscope, in Device Manager click the View tab. In the pull-down menu, click Show hidden devices. Verify that libusbK Usb Devices is present in the list. If it is, then it is most likely not a Driver issue.



3. If “libusbK Usb Devices” is not present in the list, you will need to Repair the current version of Lumaview which re-installs the correct drivers. From the download location on your computer (e.g., in the Downloads folder), double click the Lumaview.msi file to start the install. The Lumaview Setup Wizard dialog box will open; select Repair Lumaview720 and click Finish. The installation will proceed, including the opening twice of the Device Driver Installation Wizard (click Continue and Finish each time).
4. Disconnect and then reconnect the USB cable from the computer to the LS Microscope. Repeat Step 2 (above) to verify libusbK Usb Devices is now present.
5. If this does not result in the driver appearing in the Device Manager then in the same Device Manager dialog box, scroll to the bottom of the device list and click on Universal SerialBus controllers to expand the list. Look for specific controller names (not the nonspecific such as USB Root Hub or Standard Open HCD Host Controller). Examples of conflicting drivers identified thus far that might be there include: BodeVib, Holdtescs, Cypress, NDTech.



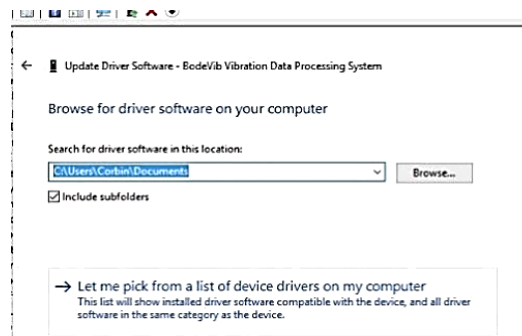
6. It may be necessary to unplug the LS Microscope (via the USB cable) from the computer and look for any names in the list where the name font color has turned a lighter gray. Reconnect the LS Microscope and verify that the one that turned lighter gray is now darker gray again. This is the driver that conflicts with the LS Microscope drivers.
7. Right click on the conflicting driver, in this example BodeVib Vibration Data Processing System, and in the pop-up menu, click on Update Driver Software.



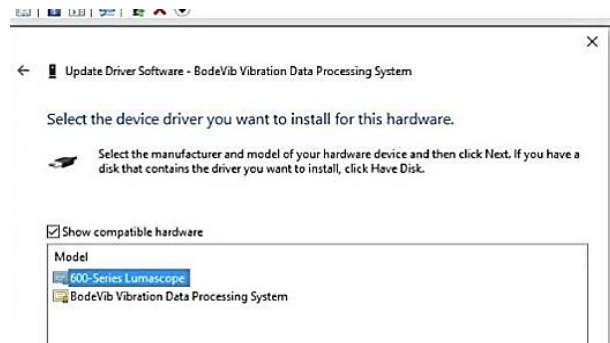
8. In the next dialog box that appears, click on Browse my computer for driver software.



9. In the next dialog box that appears, select Let me pick from a list of device drivers.



10. The next dialog box that appears will show the choices for the hardware devices. The Show compatible hardware box should be checked. Select 600-Series Lumascope and click Next. Windows should now install the LS Microscope driver over the conflicting driver.



11. When installation is complete, disconnect the LS Microscope from the computer and then reconnect it. These steps may need to be performed on every USB port that will be used with the LS Microscope.

NOTE: Older versions of Windows may ask you to restart the computer before driver is functional.

Appendix B

How to Disinfect Lumascope Microscopes

Etaluma's microscopes are revolutionary in their ability to be operated within cell culture incubators, providing the most stable and robust long term live cell imaging. Placing the LS Microscope in the incubator often includes a disinfection step before installing. In this Tech Note, we discuss the methods recommended.

Wiping Down with Alcohol

Ethyl and isopropyl alcohols are the two most widely used alcohols because of their biocidal activity. Alcohols work through the disruption of cellular membranes, solubilization of lipids, and denaturation of proteins. These processes require water so the alcohols must be diluted to 60-90% in water to be effective.

These alcohols are effective against lipid-containing viruses and a broad spectrum of bacterial species, but are ineffective against spore-forming bacteria. They also evaporate rapidly, which makes extended contact times difficult to achieve unless the items are immersed.

As mentioned above, the optimum bactericidal concentration for ethanol and isopropanol is in the range of 60% to 90% (typically 70%) by volume. Alcohols are generally regarded as being non-corrosive.

Wiping Down with Bleach

Chlorine compounds are good disinfectants, have a broad spectrum of antimicrobial activity, and are inexpensive and fast acting. Hypochlorites, the most widely used of the chlorine disinfectants, are available in liquid (e.g., sodium hypochlorite such as in household bleach) and solid (e.g., calcium hypochlorite, sodium dichloroisocyanurate) forms. Household bleach has an available chlorine content of 5.25%, or 52,500 ppm. Because of its oxidizing power, it loses potency quickly and **should be made fresh** and used within the same day it is prepared.

Exposure to Hydrogen Peroxide Vapor (HPV)

Hydrogen Peroxide Vapor (HPV) is another chemical that is effective in removing biological agents from the surfaces of equipment and other difficult-to-sterilize surfaces. The ability of vapor to reach a wide variety of desired areas means it is effective in sterilizing pass-through chambers and devices used in hospitals and manufacturing settings.

HPV's ability to decontaminate cell culture CO₂ incubators without the use of heat offers significant advantages in research laboratories in which costly down time must be avoided. The combination of a seven-minute HPV fog in the chamber with circulation by the incubator airflow blower, followed by exposure to narrow-bandwidth ultraviolet light, provides an effective antimicrobial disinfection. Further, it reaches all incubator walls, shelves, reservoirs, air plenums, sensors, and other interior components, and it leaves only small amounts of sterile water droplets as a residual.

For questions, contact Technical Support at support@etaluma.com.

Setting up Time-Lapse in Windows10

Power Settings

1. Navigate to *Settings* (click on the Windows icon on the lower left of your screen and then the gear icon). Choose *System*, then *Power & Sleep*. On the right side column click on [Additional Power Settings](#). In the left column of the Control Panel dialog, click on [Create a power plan](#). Choose the High Performance Plan as the template for your custom plan. Give your plan a name and click "Create".
2. Next, click on [Change plan settings](#) next to your plan. Set Display and Sleep settings to Never and brightness as desired. Next, click [Change advanced power settings](#). Scroll to "Hard Disk" and click on the "+" keys to open the selection. Click on blue writing and change the time to "Never". Scroll to "Sleep" and expand the selections. Make sure every item is selected as "Never", "Off" or "Disabled". Under "USB Settings", open "USB Selective suspend setting" and make sure it is set to "disabled". Press Apply to, then OK.
3. Now you have a custom power plan for running experiments on your LS Microscope. Before starting an experiment, be sure the computer is fully charged and plugged in to an outlet. When an experiment is finished, navigate to *Settings* -> *System* -> *Power and Sleep*. Click on [Additional power settings](#) and choose your usual power plan if desired.
4. Note: if you do not wish to create a custom power plan, you may simply change the advanced settings for your regular plan as described in step 2. above.

Connectivity Settings

A) Network connection NOT required (recommended configuration):

Disconnect the **Ethernet cable** (if wired connection is used) and turn on **Airplane Mode**.

Navigate to *Settings* -> *Network & Internet* -> *Airplane Mode* (left side) -> **ON**.

Airplane Mode will prevent all wireless communications (Wi-Fi, Bluetooth and cellular) between the computer and other computers and devices.

Note: after turning on Airplane mode, do not reconnect to your network, or Wi-Fi will be re-enabled, with Bluetooth and cellular remaining off.

B) Network connection required:

1. Navigate to *Settings* -> *Network & Internet* -> *Wi-Fi*.
2. Click on [Manage known networks](#), then select your network(s) and choose the following:
"Make this PC discoverable" (if an option) – **OFF**
"Metered connection" – **ON**

Note: Windows will download only critical updates to your computer when you are on a metered connection.

Windows Update Settings

1. Navigate to *Settings* -> *Update & Security* -> *Advanced Options*
2. Under "Choose how updates are installed" -> uncheck the checkbox next to "Enabling this policy will automatically download updates...".
3. Next, Click on [Delivery Optimization](#) and choose "Allow Downloads from other PCs" - OFF.

Your computer settings are now complete. You may start an experiment in Lumaview.