

## Guide to Trigger X-Cite® XLED1 with the optiMOS sCMOS camera

### Introduction

Hardware triggering uses high-speed Transistor-Transistor Logic (TTL) voltage pulses to synchronize multiple devices (camera, light source, microscope stage, etc.) in an imaging system. This hardware-based synchronization is often a necessary mode of operation for high-speed experiments including patch clamp electrophysiology, particle tracking and Ca<sup>2+</sup> imaging.

Triggering a traditional arc lamp system involves opening and closing of a physical shutter to allow or block light going through a microscope system. High speed pulsing of arc lamps requires specialized lamps and high-voltage power supplies, which lack stability. LEDs on the other hand can be switched ON and OFF almost instantaneously. This allows them to respond quickly in a triggering situation. When voltage is sent to the unit, it switches ON and when the voltage is stopped, it switches OFF without any delay.

A particularly useful feature of the QImaging optiMOS for synchronizing with LED sources is the "All Rows" exposure mode. This mode allows the optiMOS to run in a high-speed rolling shutter mode while avoiding rolling shutter artifacts,

such as uneven illumination across all the rows of an image or mixing of multiple color channels in a single image. These artifacts occur as a result of each row beginning exposure at a slightly later time than the previous row due to the mechanism of rolling shutter readout. All Rows mode avoids these artifacts by using the Expose Out TTL voltage pulses to trigger the light source to illuminate only when all the optiMOS' rows are exposing at the same instant in time. For this mode to function properly, especially at high frame rates, precise synchronization between the camera and a high speed light source is required. It is essential that the time required to turn the light on and off is very small relative to the exposure time of the camera. New high speed light sources such as the X-Cite® XLED1 from Excelitas Technologies are capable of shuttering the light on the order of microseconds, making it possible to both achieve a uniform global illumination in this All Rows Mode as well as rapidly switch between multiple excitation wavelengths.

This guide will demonstrate how to use hardware triggering with the optiMOS sCMOS camera from QImaging and the X-Cite XLED1 fluorescence light source from Excelitas Technologies.

### Required Items:



QImaging optiMOS  
sCMOS Camera



optiMOS Power Supply



X-Cite XLED1 Light Source



optiMOS PCIe PC Interface card



optiMOS Data Cable



optiMOS Trigger Cable



Computer



X-Cite XLED1 Light Source  
Trigger Cable

## Setup:



## Diagram:



## Connection Instructions



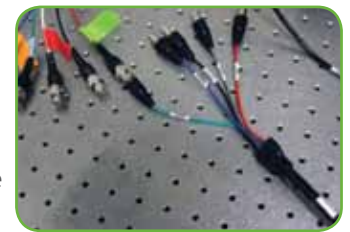
1. Connect the components as diagrammed above. A close-up of the connectors on the back of the optiMOS is shown below.



2. Connect the XLED1 trigger cable to the "Trigger Input" connector on the back of the XLED1 controller, as shown below.

The X-Cite XLED1 light source trigger cable splits into 5 separate lines. The blue, green, red, and white lines each correspond to one of 4 LEDs in the XLED1 light source, each with a different color/wavelength range. Each of the 4 different color LEDs can be used to excite a different fluorophore. Sending a TTL voltage pulse into a line in the XLED1 trigger cable causes the corresponding LED to switch on when the TTL pulse voltage is high and off when the TTL pulse voltage is low. The black line in the XLED1 trigger cable is a "global" line which triggers all lines/LEDs simultaneously. The light source must be set up for triggered operation. This procedure will be described in the next section.

For single color triggering, attach optiMOS trigger cable "Expose Out" line to the line on the XLED1 trigger cable corresponding to the specific LED color to be triggered. With this configuration, the optiMOS' Expose Out line will produce a high TTL pulse while the optiMOS is exposing, causing the specific color LED in the XLED1 light source to turn on. The Expose Out line will produce a low TTL signal at the end of exposure, causing the LED in the light source to turn off.



3. Set up external triggering on the XLED1 Display Module touchscreen or software panel by first pressing the Trigger button.

*Note: Even if the button looks greyed out and is labelled "triggered", it still needs to be pressed.*



4. Select the color channel to be triggered and cycle through the trigger modes using "Change Trigger" until triggering by an "External Source" (Ext) is chosen. Return to LED screen.

## Software Setup

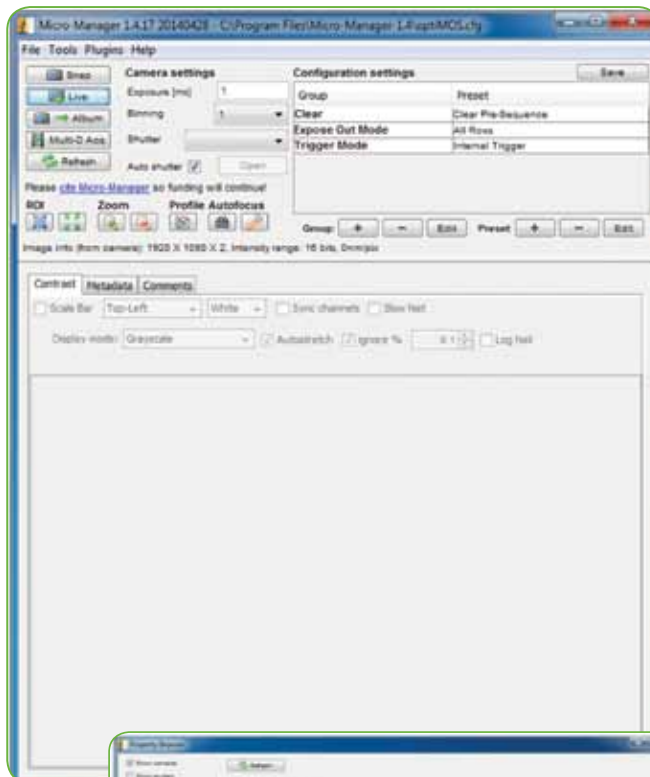
Install the latest version of the PVCAM driver, version 3.1.8 or higher. This can be found on the thumb drive included with the optiMOS or from the Software section of the QImaging website.

Install the high-speed PCI express card with Serial Light connector into an open PCIe slot with 4 channels or higher. Close the computer. Connect the data cable from the optiMOS to the high speed PCI express card. Turn on the computer and the optiMOS.



- All Rows—Global Illumination Mode. Expose Out signal voltage is high only while all rows are exposing, i.e. from when the last row begins exposing until the first row ends exposing.

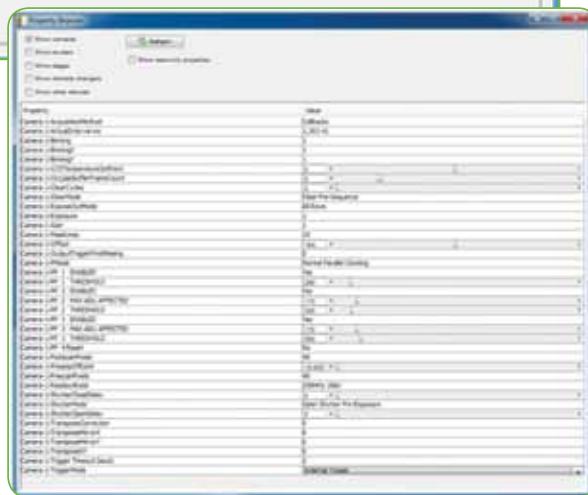
These settings can be added to the Micro-manager front panel for quick availability by clicking the “+” button next to “Group”. Alternatively, these settings can be adjusted under the Device/Property Browser available under “Tools”.



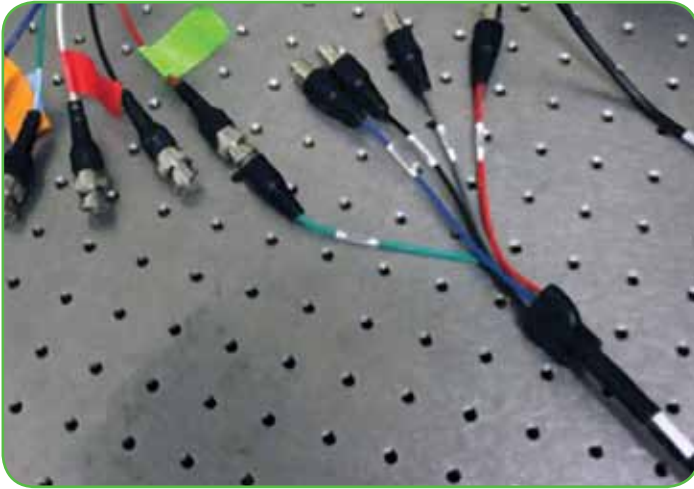
## optiMOS Triggering Modes

The optiMOS should be run in Clear Pre-Sequence mode for the highest speed performance. There are also three available Expose Out modes as well as three available Trigger modes. The Expose Out modes select the TTL pulse behavior of the Expose Out line on the optiMOS trigger cable. The three modes are First Row, Any Row, and All Rows. The All Rows behavior is diagrammed below:

All Rows (Global Illumination Mode) Vertical axis is Row number, Horizontal axis is Time:

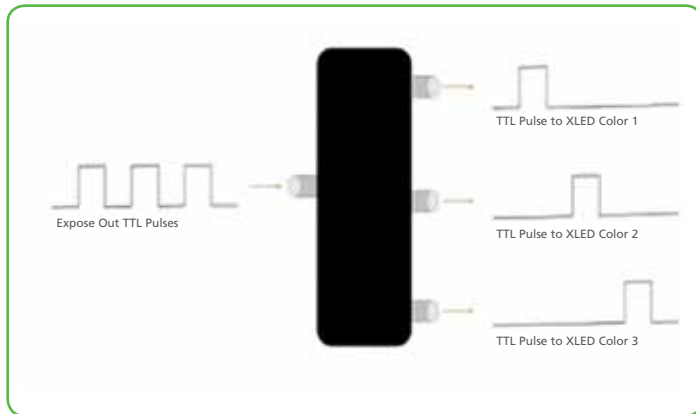


## Multicolor Triggering



For alternating multicolor triggering, a trigger box is needed to send the TTL pulse from the Expose Out line on the optiMOS trigger cable to different color lines on the XLED1 light source trigger cable in a sequential manner. A trigger box is available from QImaging. This sends trigger pulses to multiple ports in a sequence, then attach the lines to be triggered on XLED1 trigger cable to each of the trigger box's output ports.

### Trigger Box Diagram:



- External triggering in the XLED1 Display Module touchscreen or software panel can be set up similarly to the single color triggering setup. First, press the Trigger button.

*Note: Even if the button looks greyed out and is labelled "triggered", it still needs to be pressed.*



- Select each color channel to be triggered and cycle through the trigger modes using "Change Trigger" until triggering by an "External Source" (Ext) is chosen. Repeat this for all color channels to be triggered. Return to LED screen.

For further details about triggering, please refer to XLED1 user guide (<http://www.excelitas.com/Pages/Product/X-Cite-XLED1.aspx>)

*Acknowledgements: We would like to thank QImaging ([www.qimaging.com](http://www.qimaging.com)) for their help in editing this Technical Note.*

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