

Stereoscope Fluorescence Protocol

1) System Startup

- Please note the sign-up policy. You must inform the facility at least 24 hours beforehand if you can't come; otherwise, you will receive a charge for unused time. The facility will allow for extenuating circumstances (cells dying, sick day, etc.) if you inform us in a timely fashion.
- **Follow each step** of the startup poster
- **Constants** Log into the computer with you user account.



^{ce} **Open** cellSens software **b**. You should see the startup screen when the software has successfully started.

2) Focusing and scaling

- *Focus on your sample using the fast/slow wheel on the right of the microscope.*
- *Choose the appropriate fluorescent wheel from the filter turret.*
 - 1: DAPI
 - 2: FITC
 - 3: TRITC
 - For Cy5 or CFP please contact CORE staff
- *^{constant}* When switching from your eyes to the software, pull out the camera slider.
- The Designate your lens selection and zoom factor on both the microscope and in the software.

Lens:		Zoom Factor:	N.
D 0.63x 🔁 1x D 2x Đ	0.63x mag changer 🚦 1x mag changer	2x mag changer	5x • 🛋

* This step is critical for scaling. Please contact CORE staff if you need the 0.63x

3) Acquire your first image

@ Select the monochrome chip in the Camera Control window:



Ŧ Set up an exposure time.



Adjust the focus.

Click Live:

- Set the exposure time. •
 - Set it manually or automatically using the auto-exposure:

Exposure: 62.5 ms				
Manual • Automatic	SFL			
62.5 ms	: ►●◀ ⊘ 🔒			

٠ If you used the auto exposure setting, turn it off.

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	Black Balance on ROI Calibrates the black color on the image manual						
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Image Process Measure

Crop to New Image...

Calibrate Image...

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Calibrate Intensity...

Combine Channels.

Combine Frames...

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Duplicate

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Adjust Display

Pseudo Color..

Mode

Geometry Burn In Info

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Combine Channels

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Image_15

Image_14

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Acquire the image. P

4) Adding a pseudo-color

- Go to the image menu
 - Click "Combine ٠ Channels"
 - Assign the image ٠ name to the appropriate channel color
 - Click "Apply" ٠
 - Your overlay or ٠ individual image will appear in a new tab.
 - Save the image. ٠

Combine Color Imag Ctrl+2 Image Process Measure Tools Adjust Display Pseudo Color. Mode

> Geometry Burn In Info

- Adding a pseudo-color Go to the image menu P
- Click "Burn in Info" Ŧ