

## OPTOSPLIT II

Set up guide



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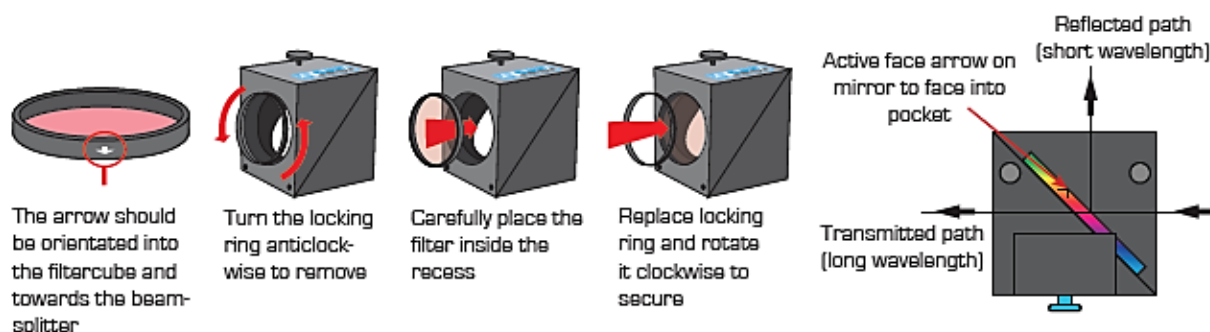
This guide details initial set up and installation of your OptoSplit II image splitter.

Each unit is serial numbered, calibrated and QC'd prior to delivery, therefore minimal setup is required providing you are familiar with the key controls. If you have any questions, please do not hesitate to get in touch.

Before using the Optosplit II image splitter you will first need to install the appropriate set of filters and beamsplitter for your application into the Cairn Filter Cube. A calibration cube with a 50% beamsplitter is also provided for alignment or spectrally neutral applications.

### Installing Filters into the Cairn Filter Cube

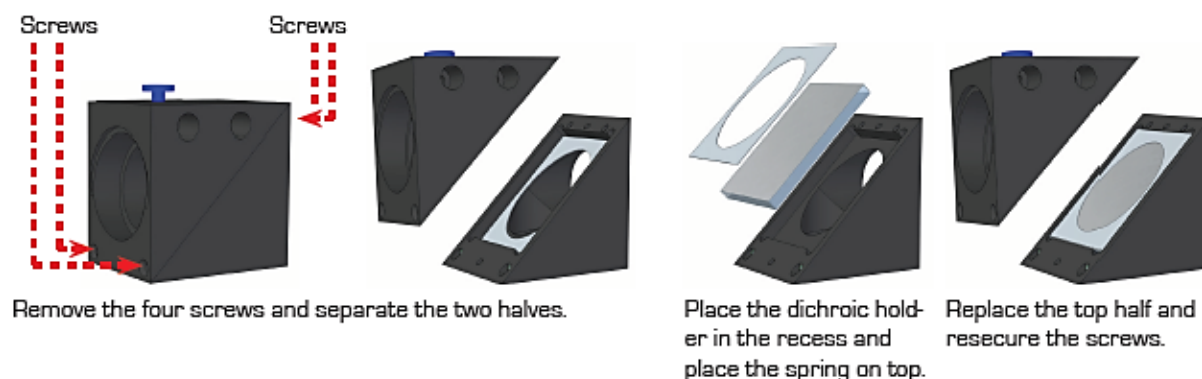
The Cairn Filter Cube has spaces for two 25mm filters. These are held in place by locking rings which can be removed using the tool provided. To fit a filter, remove the locking ring and place the filter orientated so that the arrow points into the Cairn Filter Cube and towards the beamsplitter (correct for Chroma mirrors, please refer to us if using filters from a different manufacturer). Then simply replace and tighten the locking ring.



\* Please note: care must be taken to avoid marks or dirt being transferred onto filters and mirrors.

### Installing Dichroic Mirrors into the Cairn Filter Cube

To fit your dichroic mirror into the Cairn Filter Cube remove each of the four screws that secure the two halves together. You will then be able to gently pull the two halves apart. There is one steel locating rod in each half of the cube to ensure the correct alignment when it is reassembled. Once the two halves of the cube are separated, place the dichroic mirror into the rectangular recess, active face down. Then simply reassemble the cube and tighten the screws.

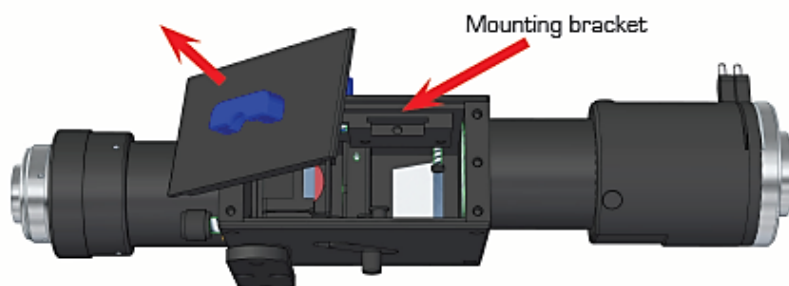


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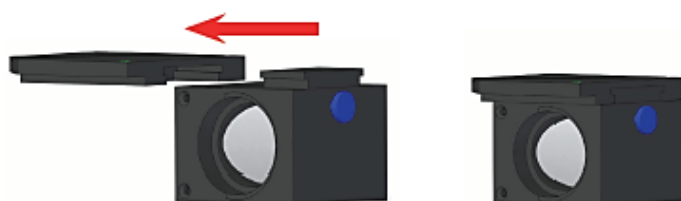
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### Inserting the Cairn Filter Cube

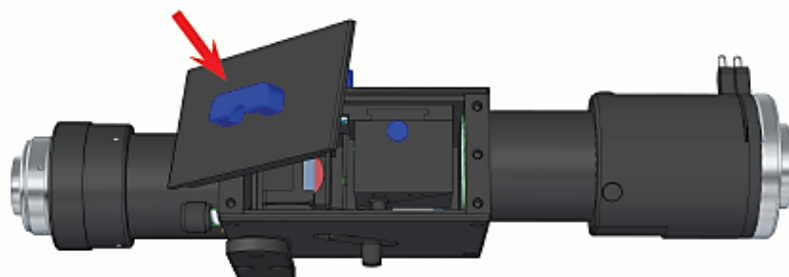
The Cairn filter cube is designed to be easily accessible in order to facilitate quick and easy changes of filter sets. Access to the filter cube mount is gained by removing the magnetic access panel using the handle to pull gently and firmly. The filter cube mount will now be visible. The mount attached to the rear of the Cairn Filter Cube is designed to mate with the bracket on the internal wall of the Optosplit II so that the small handle on top of the cube will be facing out. Once the Cairn Filter Cube is mounted successfully, replace the access panel using the handle.



Remove the access panel using the handle.



Mount the filter cube onto the bracket on the interior of the Optosplit II.



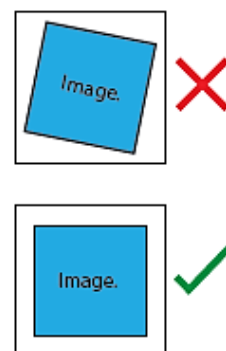
Replace the access panel using the handle.

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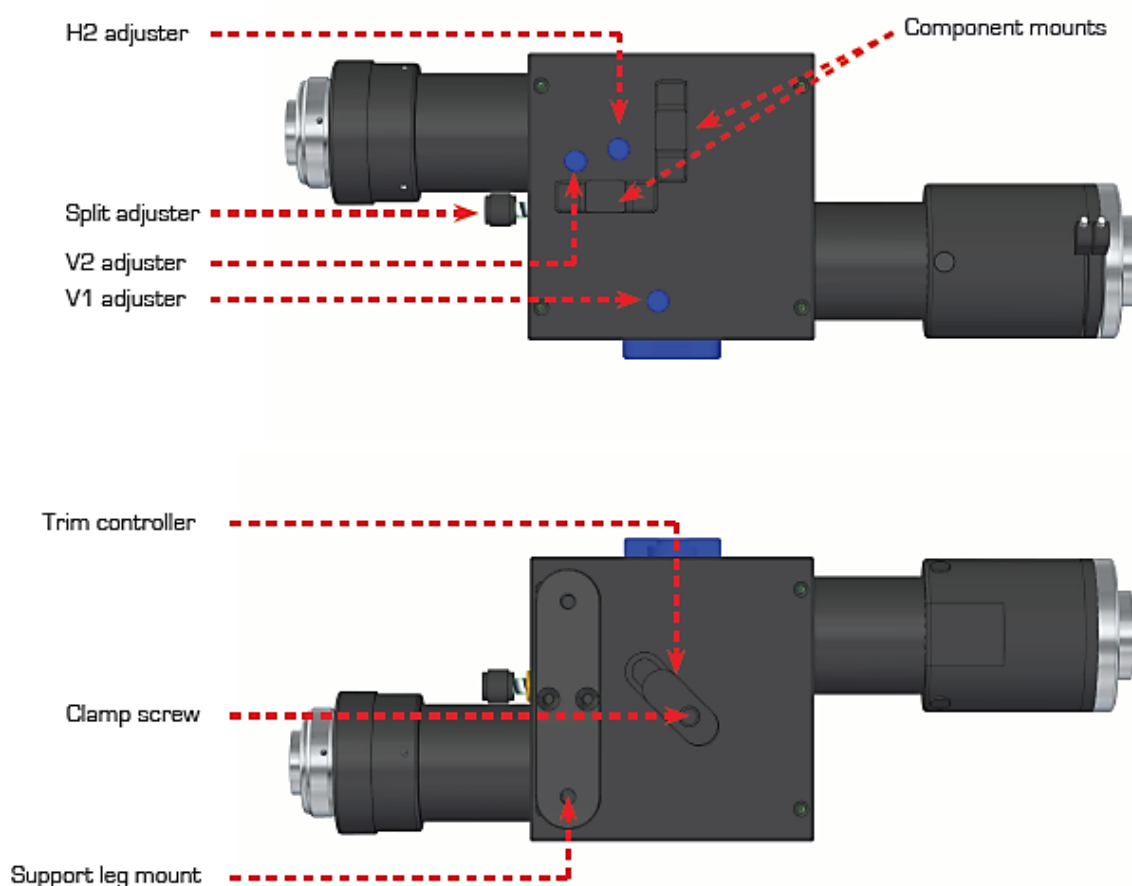
### Installing the Optosplit II in the Light Path

Having fitted the camera to the output port of the Optosplit II it should be possible to adjust the image to see a sharp picture of the aperture edges with the sample in focus. The image should line up with the edges of the aperture and should not have any rotation or slant in either direction. If your software can display a rectangular ROI then the image of the aperture can be aligned to this.



### Optosplit II: Key controls

The Optosplit II uses a single control for adjusting image separation (Split Adjuster) and allows for different sized camera chips to be used. There are additional controls for refining the ROI, and centering the image.

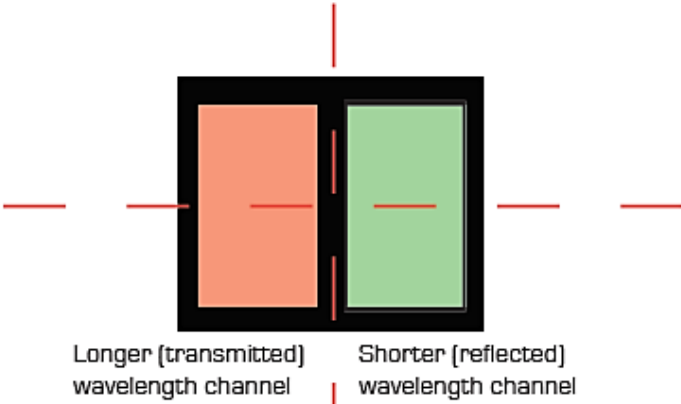


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Optosplit II: Key controls table

Control	Description
V1	Vertical adjustment for the shorter wavelength image
V2	Vertical adjustment for the longer wavelength image
H2	Horizontal adjustment for the longer wavelength image
Split	Separates the two images horizontally. Always turn ANTI-CLOCKWISE for optimal image separation
Component mounts	Drop-in positions used to insert a variety of different components into the light path of either the shorter [reflected] wavelength or longer [transmitted] wavelength image. These include: <ul style="list-style-type: none"><li>• Shutter plate: used to physically block one of the channels when utilising 'single-channel' mode</li><li>• Neutral density [ND] filter: useful when one image is disproportionately brighter than the other</li><li>• Corrector lens: used to reduce chromatic aberration and ensure both pathways are focussed on the camera sensor</li></ul>
Trim	Corrects vignetting, which is characterised by one image appearing darker towards one edge. See the 'Image Optimisation' section.

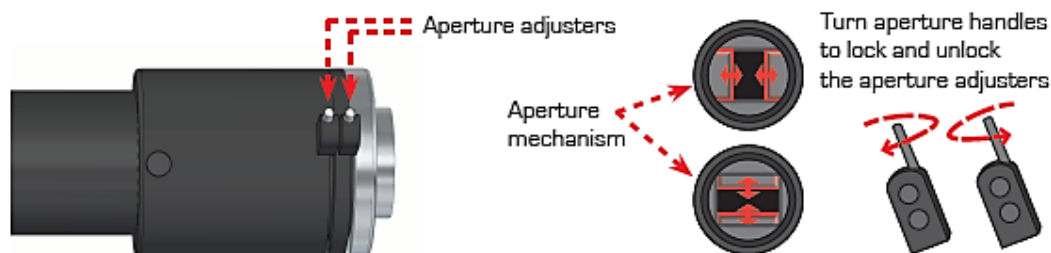


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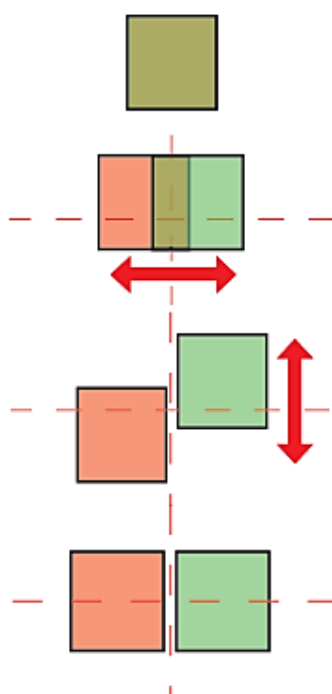
### Adjusting and Locking the Aperture

The Optosplit II is supplied with an adjustable rectangular aperture that allows the user to determine the ROI both vertically and horizontally.



### Adjusting the Position of the Images

Although the camera can be sensibly orientated in any of four rotated planes at 90 degrees to the microscope, we would recommend that it is set with a horizontal split and the V1 control adjusting the vertical height of the right hand side channel. This is the convention used in this manual and will assist troubleshooting. When the camera is mounted correctly, the split adjuster and the aperture control are the only controls that will be frequently adjusted. The remaining adjusters on the body of the Optosplit II should remain untouched unless the filter set has become misaligned. It may be easier to use the calibration cube for initial set up so that both channels display the same information.



Here we see the two superimposed images. Although your image may be monochromatic, colours have been used here to define the two images.

Turning the Split control anti-clockwise will separate the two images along the horizontal axis. Only small adjustments will be required when using the horizontal image separation adjustment.

Should the left and right images be at different vertical heights then adjust the images using the V1 and V2 adjuster. Turning V1 will alter the position of the shorter wavelength image (right channel), and V2 will alter the position of the longer wavelength image (left channel).

When the spectrally resolved images are side by side on the camera chip you are ready to record. When carrying out experiments the aperture should be set to mask the region of interest tightly so that the two images are located as closely as possible on the camera chip.

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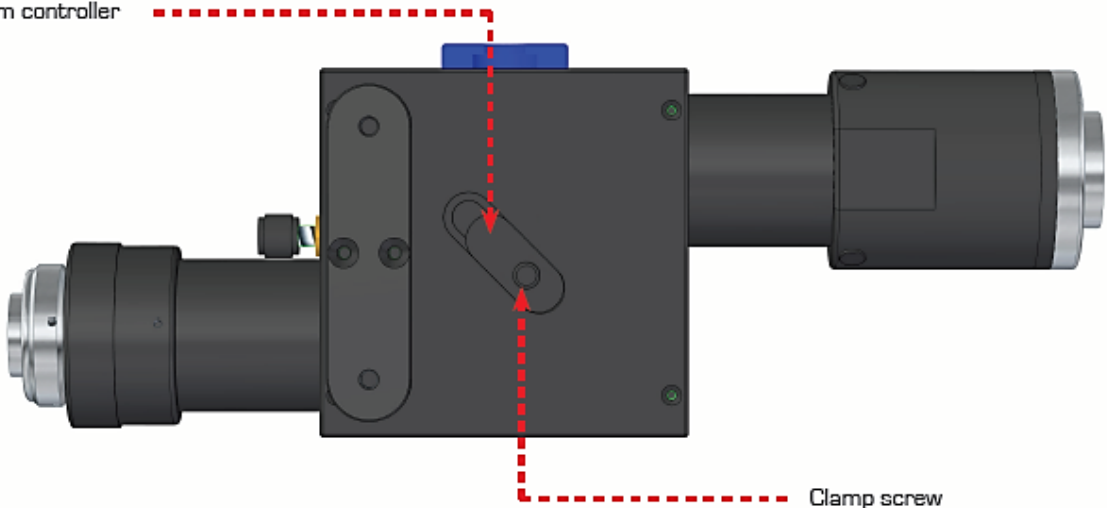
### Image Optimisation (Trim Adjustment)

Vignetting is when the image appears darker towards one edge. This effect can be easily corrected by adjusting the trim control. To adjust the trim control, loosen the clamp screw on the underside of the Optosplit II by a quarter of a turn in order to free the slider which controls the trim control. Gently move the slider until the vignetting is eliminated. If the slider is moved too far then the vignetting will become apparent on the other image and will need to be adjusted in the opposite direction until both images are the same intensity. It is important to remember to re-tighten the clamp screw once the adjustment is complete. If the Optosplit II has been configured into single channel (non-split mode) and Trim adjustment is required to compensate for vignetting being observed, it is likely the slider will need to be at the extreme of its range of movement to either end.

Vignetting is a darkening of the image towards one edge.



Trim controller



Clamp screw

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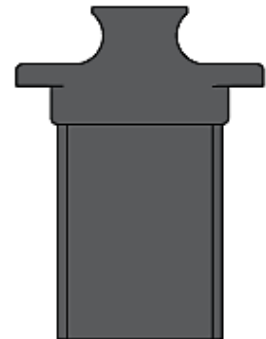
### Single channel (non-split) mode

When dual splitting is not required, the OptoSplit II unit can be used in bypass mode, allowing the unit to remain in situ on the microscope whilst utilising the full camera chip to generate one image. There are several ways this can be achieved:

#### A: Removing the Cairn filter cube

Suitable if no emission filters are required in the light path

- Remove the Cairn filter cube
- Centre the image using the Split control
- Open the aperture
- Adjust the Trim control to remove any vignetting



Cairn shutter plate

#### B: Blocking the longer wavelength path

Suitable if only the shortest wavelength channel is required

- Leave the Cairn filter cube in place and block the long wavelength path using the shutter plate provided in the auxillary holder position. Most dichroic mirrors have long-pass characteristics, in which case the longest wavelength will be the transmitted channel.
- Centre the image using the Split control
- Open the aperture
- Adjust the Trim control to remove any vignetting

#### C: Blocking the shorter wavelength path

Suitable if only the longest wavelength channel is required

- Follow the steps for option B, but block the short wavelength path (reflected channel) with the shutter plate instead.

