## **MULTISPLIT V2** Quick start guide July 2018

The MultiSplit is calibrated and QC'd prior to delivery, however some alignment is required upon initial setup using the calibration cubes provided (containing a 50% mirror). A detailed setup guide is also supplied; however, the following simplified instructions are useful for initial alignment.

1. Ensure all filter cubes are removed from the unit.

2. Using brightfield illumination, focus on an image of a graticule and divert the microscope output to the camera.

3. Close the adjustable aperture down to ensure one, small rectangle is present within the field of view. This single image is only reflected by the internal MultiSplit mirrors and is therefore by default the longest wavelength channel once filter cubes are in place. This image is transmitted through two dichroic mirrors and referred to as  $\mathbf{T}$ .



No cubes in the unit Image TT

4. Locate the horizontal and vertical image separation controls at the front of the unit (labelled on the lid). When turned **anti-clockwise**, TT should move toward the **bottom right corner**. If this is not the case, rotate your camera until this orientation is achieved.





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5. Remove the magnetic door cover and insert the first calibration cube into the position labelled Horizontal Split - Cube 1, which will generate your second image. This is reflected by the first filter cube, and transmitted by the second, hence the nomenclature **RT**.



**Cube 1 inserted** 

6. Adjust the position of RT using the horizontal (H) and vertical (V) controls on the magnetic door cover until it reaches the bottom left hand corner.

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7. Next insert Cube 2, which will cause image **TR** to appear.



**Cube 2 inserted** 



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8. Cube 2 is mounted internally on a carrier, with the adjustment controls mounted on the top plate. Adjust the position of TR using the horizontal and vertical adjustment knobs associated with Cube 2.



9. Now insert the final cube into Cube 3 position on the back side panel of the unit, which will cause the final image, **RR** to appear.



10. Cube 3 is housed in a carrier similar to Cube 2, with the H&V adjustments also on the lid to allow alignment of this image in the top left corner.

Final adjustment of all controls can now be carried out for more precise image positioning.

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When using long pass dichroic mirrors, the four images correspond to the following channels in order of descending wavelength: TT: (1) Longest channel RT: (2) TR: (3)

RR: (4) Shortest channel

For advice regarding dichroic mirrors and emission filter positions, please contact us with details of the four-way imaging you with to achieve and we are happy to advise accordingly.

## Multi-depth Imaging

The MultiSplit has two auxiliary drop-in positions for each channel, accessible from the outer panels of the unit. Blank covers are provided as standard; however, additional accessory kits can be provided with holders for a) Neutral Density filters or b) Corrector lenses. Neutral Density filters are useful if one or more channels are disproportionately brighter than another's. Corrector lenses serve a dual purpose for chromatic correction, or to deliberately shift the focal plane for simultaneous multi-depth imaging.

Our corrector lens kit provides a series of weak crown glass lenses and their optimal position for each channel is indicated below. For precise focal plane shifting, we have a spreadsheet available to estimate the best focal length lens (if available). Please contact us for this further detail and we will be happy to advise for your application.









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