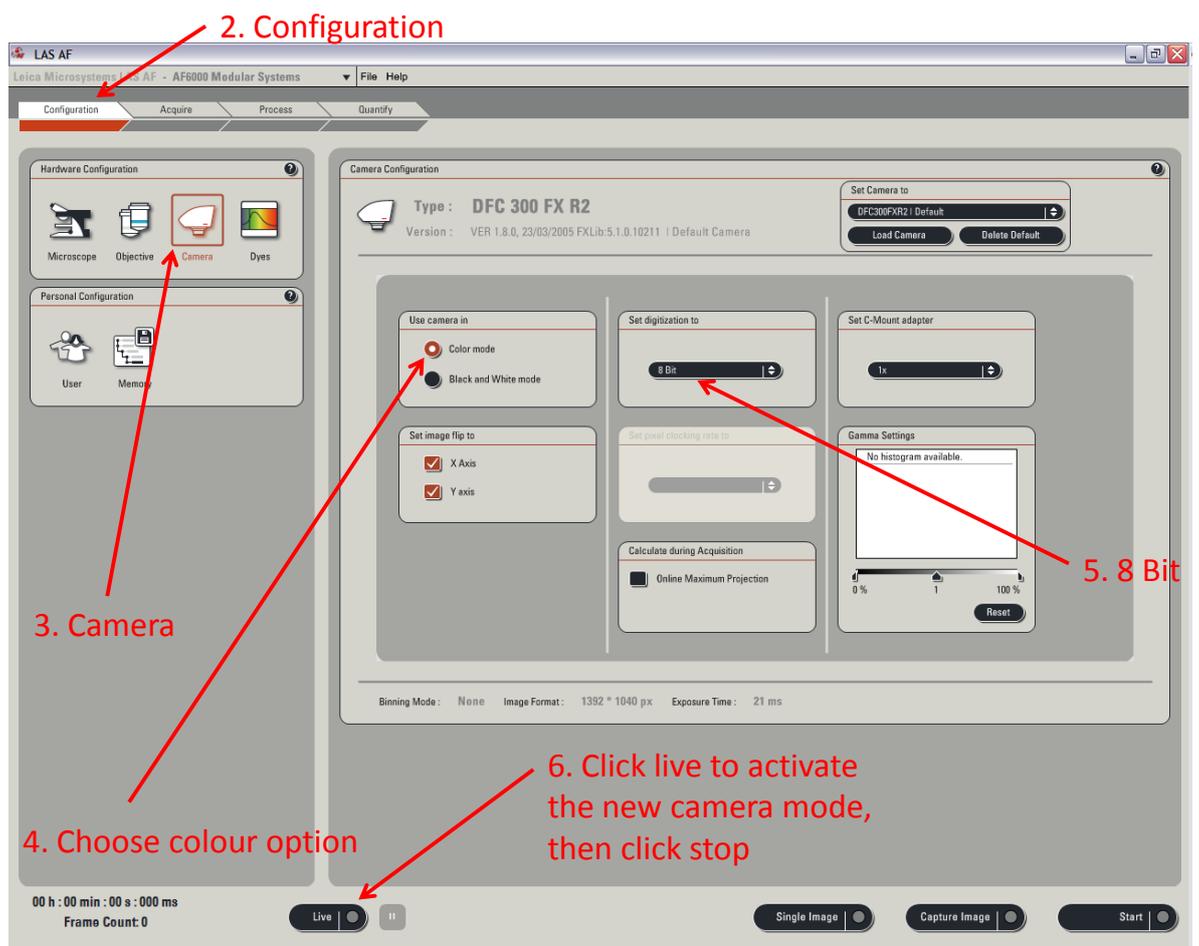


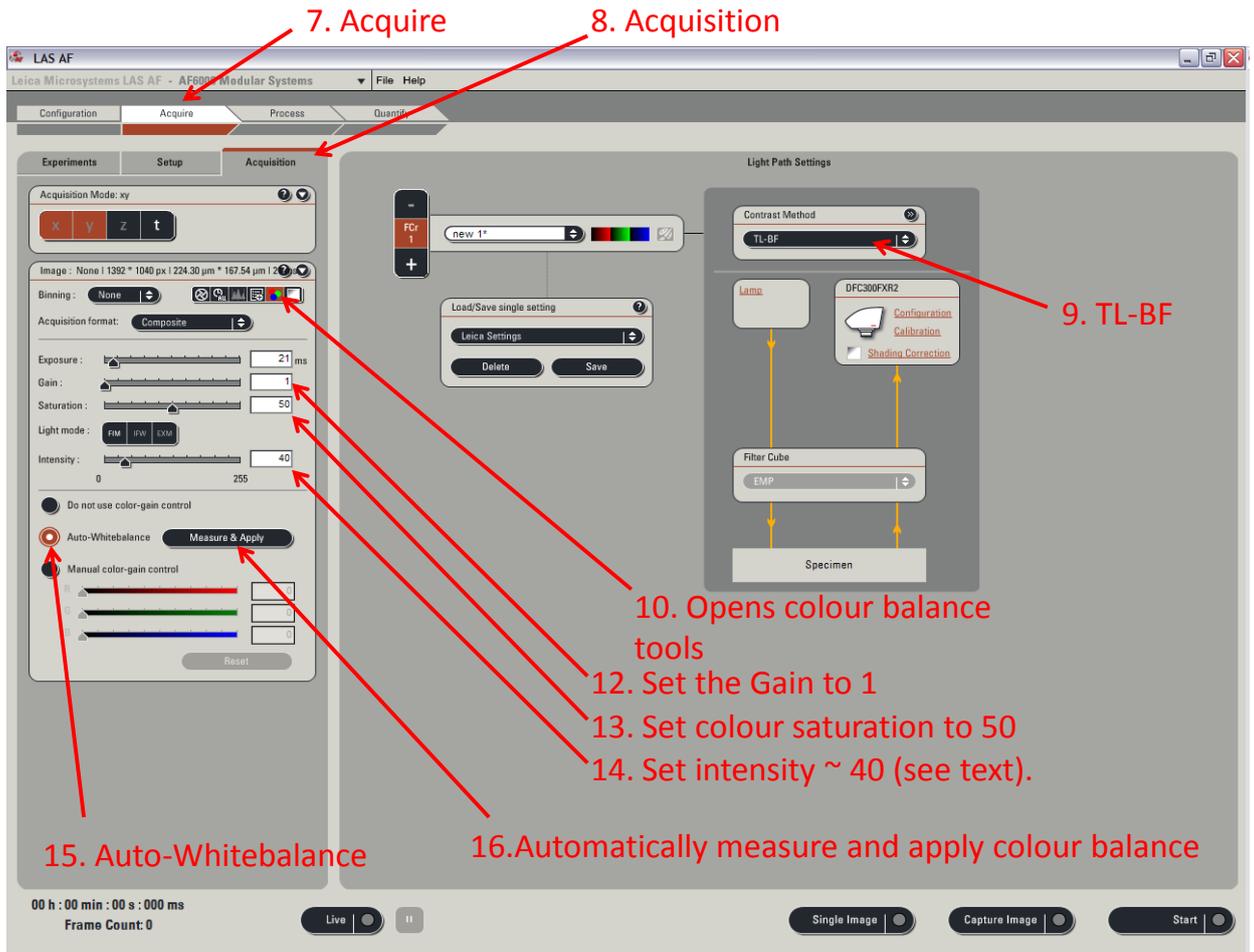
DMI4000 LASAF: Operating in colour mode.

The instructions below assume that you know how to set Kohler illumination and have been trained on the microscope.

When using Colour mode you will need to put the neutral density (ND) filter into the transmitted light path (cover the hole in the condenser) to reduce light levels. The ND filter should normally be found resting on top of the condenser.

1. Start LASAF and at the first checkpoint make sure that DefaultDynamicWidefield is listed as the configuration. If it is click OK.
2. Once LASAF has started select the “configuration” tab (top LHS).
3. Select the “camera” sub-tab (top LHS).
4. Select camera click “colour” (if “Colour” is greyed out go to Troubleshooting 1.)
5. Choose 8 bit
6. Click “live” then, after it has gone live, click “Stop” (if you do not do this the camera may not actually go into colour mode).





7. Select the "Acquire" tab.
8. Select the "Acquisition" sub-tab.
9. Select "contrast method" TL-BF
10. Click the button which has red, green and blue dots – it opens an extension to the main control panel.
11. Move the specimen to an area on the slide that does not have any specimen/staining.
12. Set the gain to 1
13. Set the saturation to 50.
14. Ideally make sure that the Intensity is set to at least 40 (with some high magnification/NA lenses you may need to go just below 40).
15. Adjust the exposure until the brightness looks ok (not too bright or dark). The exposure time will be very short.
16. Select the Auto-White balance button
17. Click Measure and Apply. When it finishes the image should look mid grey.
18. Move the specimen to an area of interest.
19. To make small adjustments to the brightness alter the exposure **not** the intensity. Changing the intensity will also change the colour balance which will then need to be reset (as above).
20. The colour balance will need to be reset (as above) if the lens is changed.

Troubleshooting.

1. If the "colour" option is greyed out under "Configuration -> camera" tab, go to the "Acquire -> Acquisition" tab, and set "Binning" to None. Then click "live" followed by "stop" to activate the new setting. It should now be possible to change the camera to colour mode.
2. If you need to turn the intensity up way above 40 the phase contrast ring may be set. To get a pure brightfield image turn the silver ring (in the front of the condenser) to position 6 or 2.
3. If there are strange out of focus spots in the image, clean the specimen (above and below), adjust the lever (condenser aperture iris) in the front of the condenser to a higher number or Ph. Ask for help in setting this in future.
4. If nothing seems to be working, close the software, logout, power down all components except the fluorescence lamp (if it is on) and then restart.

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