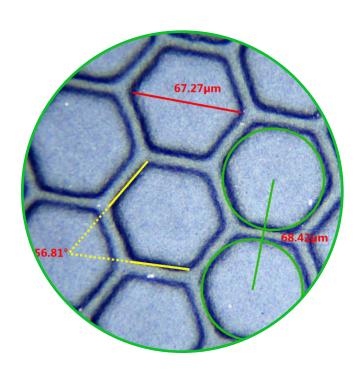


Calibrating and Measuring with Richter Optica DCM5.3 Microscope Camera







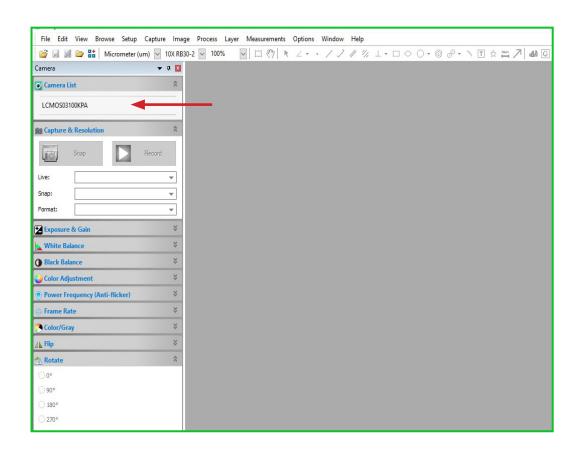
Items you will need to calibrate your camera

- Microscope
- DCM5.3 camera
- Stage micrometer
- PC or laptop. Measurement features do not work on MAC.



Getting Started

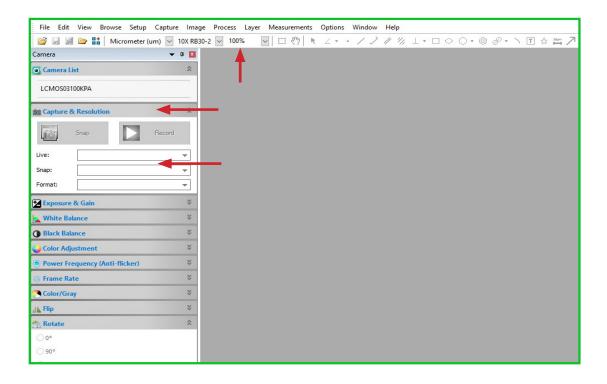
- Once your microscope, camera, and computer are connected and properly set up, launch the software.
- Place the stage micrometer on the microscope's stage and bring the stage micrometer into focus. Place the stage micrometer as straight as possible on the stage. This will assist in getting a proper calibration later.
- Check that you have selected the camera which is found on the tool bar to the left. The camera is recognized by the following model number: C3CMOS05100KPB. Once the camera is selected you will see a live video image of the stage micrometer ruler. If the camera is not selected you will not see an image.



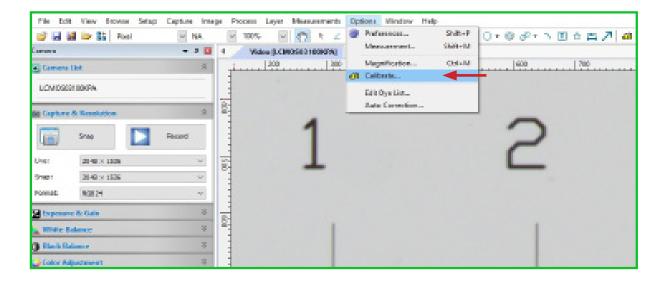




- On the Camera sidebar, click double arrows next to <u>Capture & Resolution</u> to expand that box. In the
 Capture & Resolution box you will see <u>Live</u> and <u>Snap</u> with double arrows next to them. Click on those
 dropdown arrows and set numbers to the max resolution available.
- On the toolbar look for a number with a <u>% sign</u> next to it. With the drop down arrow that is next to that number, select 100%.



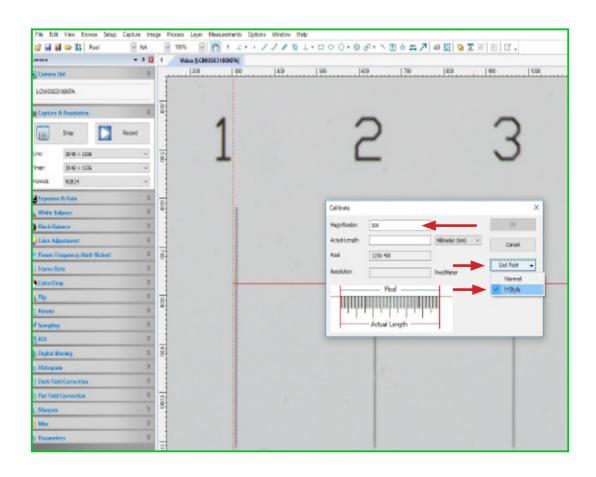
- On the toolbar, go to Options > Calibrate. Bring the stage micrometer ruler into focus if needed.
- Note: Your stage micrometer ruler will have a known distance. In this example we are using a stage micrometer ruler that is 25mm (25,000um) long.





- In the <u>Calibration Box</u>, select your <u>Magnification</u> via the drop down arrow. You may type in a different magnification number if your magnification is not listed. (Note: the Magnification is determined by the magnification of the eyepieces and the objective. In this example we are using 10x eyepieces with a 2x objective for a total magnification of 20x.)
- You will need to calibrate each objective being used.
- Click on <u>End Point</u> and select <u>H-style</u>.

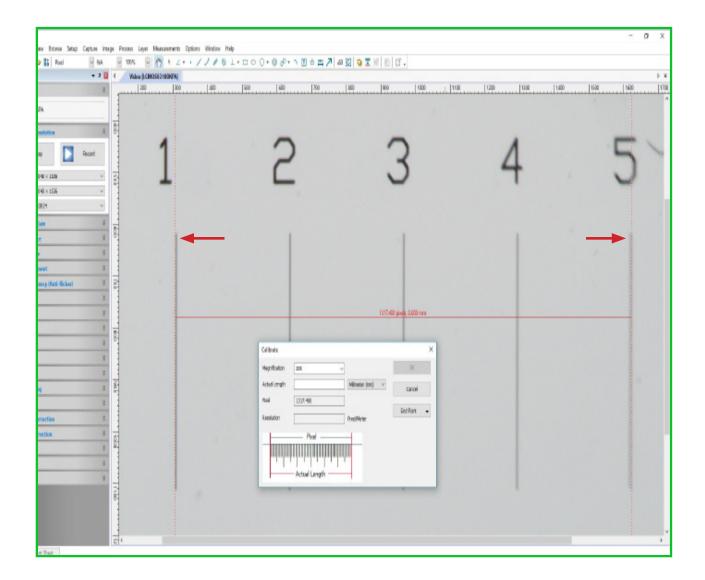






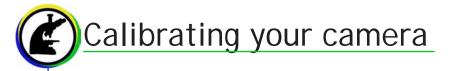


• Now align the left red <u>H-line</u> (<u>Horizontal Line</u>) to the stage micrometer's <u>one</u> (1) line then move the right red H-line to the furthest major line seen (5). (Moving H-lines: Do this by moving the pointer track symbol (represented by a pointer hand) over to where the horizontal dotted redline and vertical solid red line meets or just over the red horizontal line itself. The symbol will change to either a double sided arrow or to cardinal arrows, at this point you can move the vertical or horizontal line.

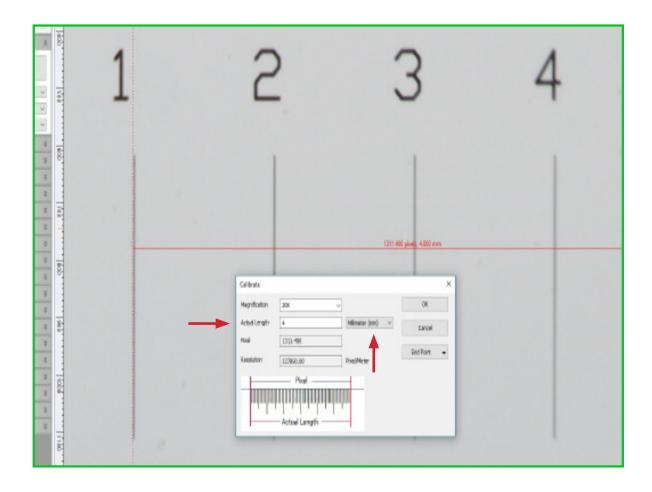


- Notes: In the example, we are using a 1mm long with 100 division stage micrometer. 1mm = 1000um (micron).
- Depending on your camera and it's C-mount adapter, you should see most, if not all, of the stage micrometer ruler. Place the right H-line on the farthest last hash line and count from the left red H-line to the other and add total.





- After adding up the total between the red H-lines, input the number into the <u>Actual Length</u>. For this example, it is <u>4</u>. (Because our left red H-line was at 1 and our right H-line was on the 5th hash mark, and each hash mark is 1mm). Change the measurement standard that is located next to the Actual Length box to Millimeter.
- Note: Make sure to be consistent in your red H-line placements. If the left H-line is placed on the left side of the dividing line, place the right H-line on the left side of its dividing line as well.



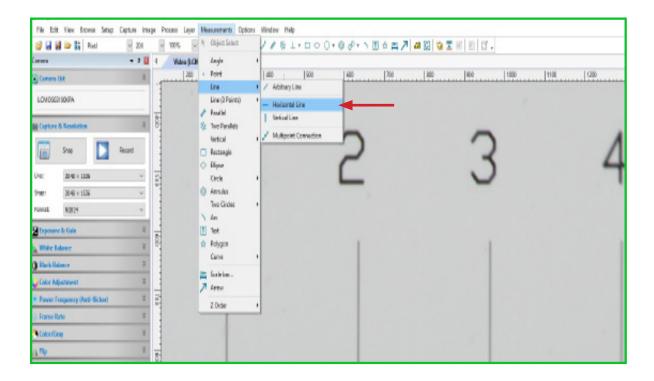
- Once all input fields in the Calibration Box are completed and you are satisfied with the H-line placements, click OK.*
- Note: *Once a calibration is entered it cannot be deleted from the computer, ensure your measurements are correct before clicking OK.
- Repeat calibration steps for each objective being calibrated. (i.e. Switch to next objective, set H-lines on the desired hash marks located on the stage micrometer, count, input data, save, repeat.)

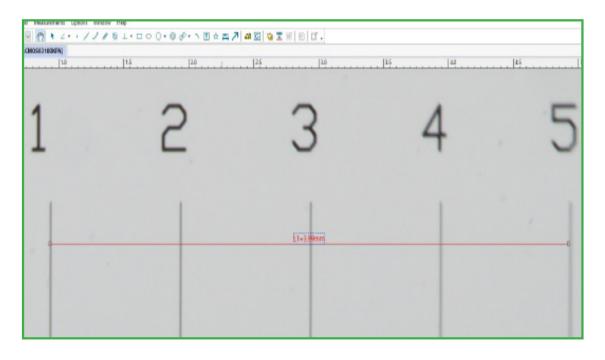




Calibration Checks

- On the toolbar, go to Measurements > Line > select Horizontal Line.
- Left mouse click at the stage micrometer's one line and drag line to the same point as where the right red H-line was before and left click. The number should read correctly +/- 1 micron.



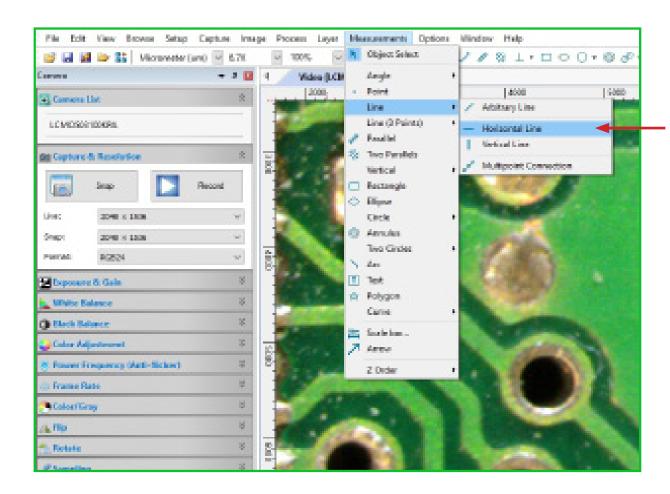




Measuring with your camera

- Now that you have performed the calibration you are ready to make measurements. (If using a Stereo Microscope, ensure the microscope has detents, also known as click stops. This feature is necessary for ensuring accurate measuring with a Stereo Microscope as it allows for a precise magnification to be determined.
- Place your specimen on the stage and bring the specimen into focus.
- We will be working with a standard horizontal line. Select the Measurements > Line > Horizontal Line.
- The software offers many options for performing measurements. They are located in the toolbar as well as under the Measurements drop down.

Helpful Tip:
If using a Stereo
Microscope, ensure the
microscope has detents,
also known as click stops. This
feature is necessary for ensuring accurate measuring with a
Stereo Microscope as it allows
for precise magnification
to be determined.

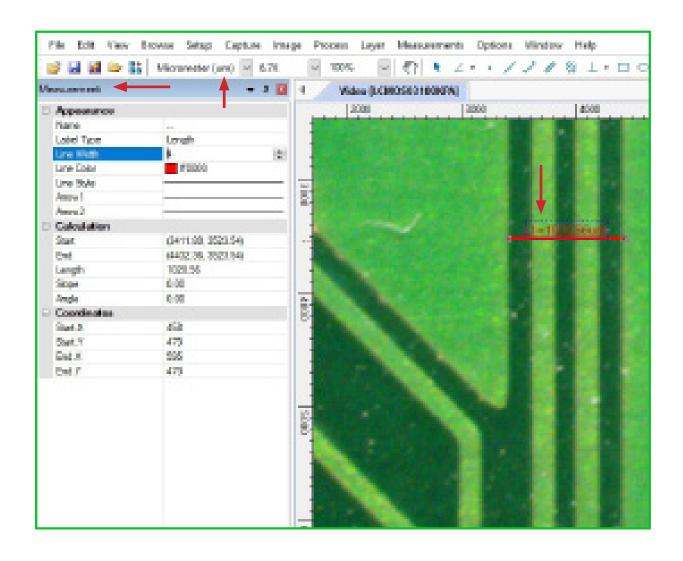




Measuring with your camera

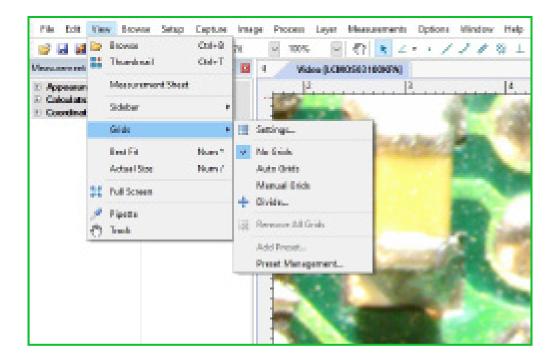


- A cross hair will appear. Click on the beginning point of where you intend to measure. A red line will appear. Move the line to the end point of your measurement and click to establish the end point. A line labeled "L1", indicating this is "Line One", with a measurement will be visible.
- Now that you have performed a measurement, you can make adjustments to the measurement line. To the left you will notice the tool bar has changed from Camera to Measurement.
- The features in this toolbar allow you to adjust the Name, Label Type, Line Width, Line Color, Line Style and add arrows using the Arrow 1 and Arrow 2 features.
- Click on the feature you would like to adjust and a drop down box will appear to the right. Select the adjustment you would like to make and the measurement line will reflect those changes.
- Just above the Measurement toolbar is a drop down box to select the unit of measurement. In the image below it is set to Micrometer.



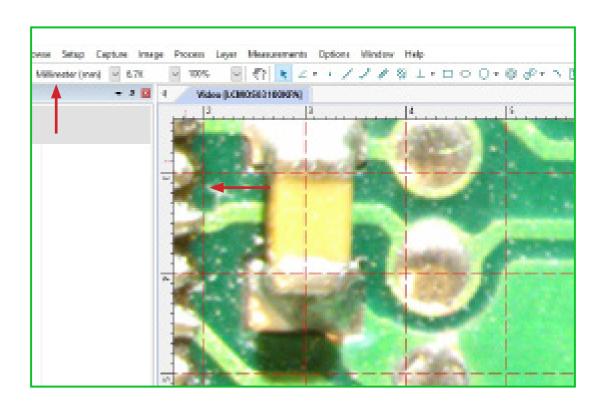


- Select View > Grids from the toolbar.
- Select Settings to make changes to the appearance of the Grid style. You can change the Line Style and Color. A dash line will be used in these images.
- Select from the different Grid options of No Grids, Auto Grids, Manual Grids and Divide.
- No Grids can be used to remove any grids you have used or to indicate no grids are to be placed over your image.

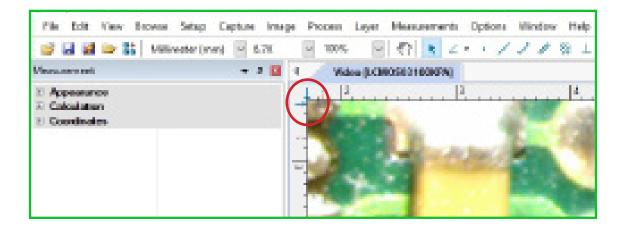




• Auto Grids establishes a grid overlay with measurements based on the unit of measure selected.

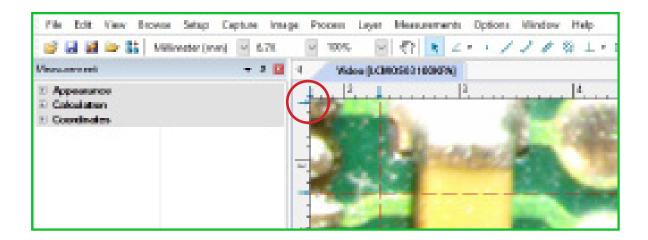


- Manual Grids allows the user to set the location of the individual grid lines.
- Two blue arrows will appear at the top left hand corner of the image.
- Move the two arrows horizontally or vertically to establish the grid lines.

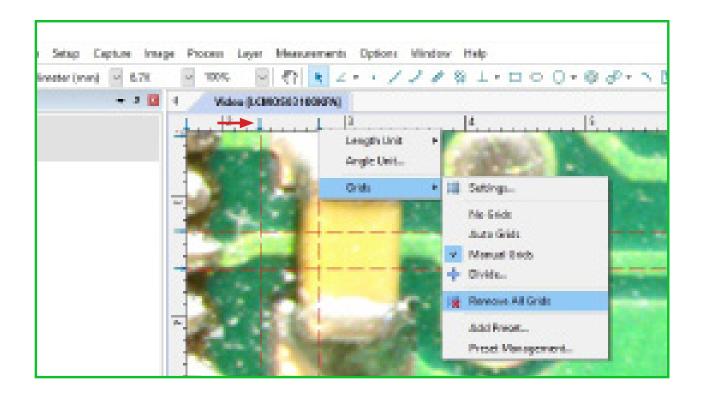




• New <u>blue arrows</u> will appear in the top left hand corner. Use the new blue arrows to establish additional lines.

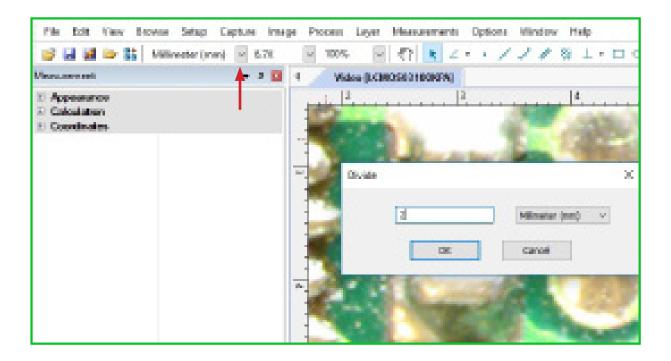


• To remove grid lines, right click any blue arrows, select Grids > Remove All Grids. This function will remove any grid lines you have created and reset the Manual Grid feature allowing you to start a new Manual Grid.

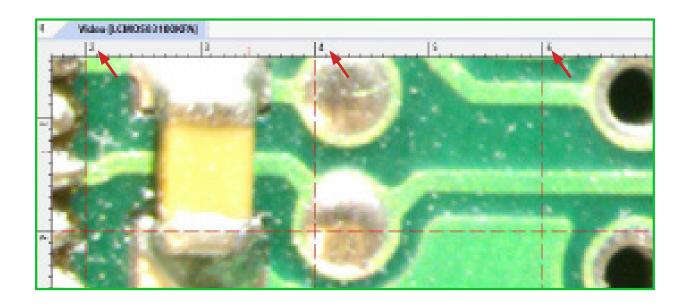




- The Divide feature allows the user to establish a grid overlay per the users specified measurements.
- Select View > Grids > Divide. A box will appear to input the desired divisions.
- In the below image a 2 has been input to designate a grid that will have a grid line placed every 2 millimeters. The unit of measurement can be adjusted by clicking the <u>down arrow</u> to the right of the unit of measurement.
- Click "OK".



• A grid with lines set at 2 millimeter divisions will now be overlaid on the image.





• To remove any of the Grids created, select View > Grids > No Grids.



Quick Trouble Shooting Guide



Problem Solution

| No live image displayed | Beam splitter is not in the proper setting. Check the beam splitter setting. |
|--|---|
| | Camera is not selected. Select the camera on the tool bar to the left. |
| Live image is dark | Adjust the illumination on the microscope Use the white balance option on the tool bar to the left. |
| Live image is upside down or backwards | Use the Rotate and Flip features on the tool bar to the left. |
| Dust or dirt is visible in the live image. | Clean the sample Clean the objective Clean the eyepieces Clean the C-mount Clean the lens on the camera |

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