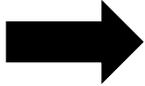


OVERVIEW OF THE CALIBRATION PROCESS

- A. Add and tighten all the objectives.
- B. Verify the camera to stage alignment.
- C. Calibrate lenses with calibration grids.
- D. Perform parcentric/parfocal calibration.**
- E. Check the accuracy of the calibration.



In this tutorial, we demonstrate parcentric/parfocal calibration for 100x, 63x, 40x, 20x, 10x, and 5x lenses:

PERFORMING A PARCENTRIC/PARFOCAL CALIBRATION

Don't move the stage at any time; don't use oil with oil objectives.

1. The 100x (highest power) objective is in position and the corresponding software lens is selected.
2. The top left corner of the 25 μm (small) grid is in view.
3. **Optional:** Increase the size of the cursor for a more accurate alignment for the **parcentric** calibration.

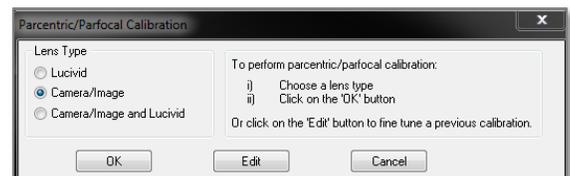
Use the **Options>General Preferences>Cursor** tab to increase the **Crosshair Size** (recommended: 75 px)

OR

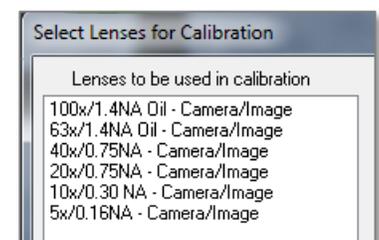
Hold down the **CTRL** key while pressing the **Up** or **Down** arrow keys.

4. Click **Tools>Parcentric/Parfocal calibration**.

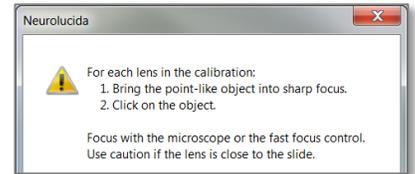
- a. In the **Parcentric/Parfocal Calibration** window, select **Camera/Image** under **Lens Type**.



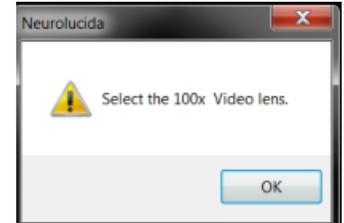
- b. In the **Select Lenses for Calibration** window, verify that the lenses are listed in descending order, from highest to lowest power then click **OK**.



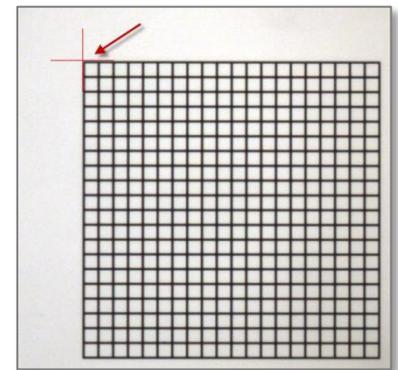
- An alert is displayed. This is a reminder that, for each lens, you need to click the *EXACT SAME POINT* on the grid, regardless of where it appears on the screen.



- Follow the prompt: Verify that the 100x objective (called video lens here) is in position.
- Bring the top left corner of the grid into focus.



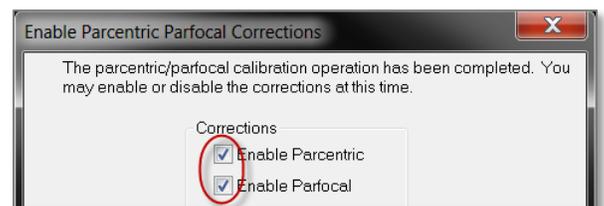
- Click the outer edge of the top left corner of the grid.
Align the crosshairs of the cursor with the edges of the grid as precisely as possible. You'll click the same point with the other objectives.



- The program prompts you to switch to the next highest power objective. Don't use oil.
Optional: Before switching objectives, adjust the light level on the microscope to see the grid clearly enough.
When you switch objectives on the microscope, the program automatically selects the corresponding software lens.
 - Bring the top left corner of the grid into focus.
 - Click the outer edge of the top left corner of the grid.
- Repeat step 9 for the remaining lenses (40x, 20x, 10x, 5x).

- Once all lenses have been calibrated, the **Enable Parcentric Parfocal Corrections** window is displayed. Check BOTH the **Enable Parcentric** box and the **Enable Parfocal** box.

Don't check ENABLE PARFOCAL if your microscope has an internal motorized Z axis.



The next step is to check the accuracy of the calibration.