Olympus BX-51 microscope calibration

Dec 07

| OBJECTIVE | OBJECTIVE | nivel length | | Calibration pixels/um | Calibration um/pixel |
|-----------------------|-----------|-------------------|-----|-----------------------|-------------------------|
| | OBJECTIVE | pixel length | um | pixeis/um | um/pixei |
| 10x objective | 10 | 517.0 | 700 | 0.739 | 1.354 |
| 40x objective NA=0.5 | 40 | 445.0 | 150 | 2.967 | 0.337 |
| 40x objective NA=1.0 | 40 | 441.5 | 150 | 2.944 | 0.340 |
| 60x objective NA=0.65 | 60 | 442.0 | 100 | 4.420 | 0.226 |
| 60x objective NA=1.25 | 60 | 443.0 | 100 | 4.430 | 0.226 |
| 100x objective | 100 | 368.0 | 50 | 7.360 | 0.136 |
| Binning = 2 | | Scale measurement | | | |

Note: Binning on the Sensys camera is set to 2.

Sensys KAF1401 cooled CCD replaced Oct 2009

This combines each set of 4 pixels into 1, reducing resolution by a factor of 2 (but increasing sensitivity by a similar amount) Image size is 656x517 [with binning set to 2]. Most users set binning to 2 with this camera.

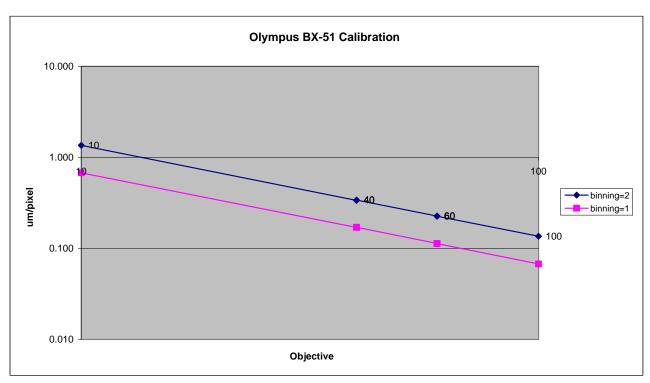
| | | | | Calibration | |
|-----------------------|-----------|-----------------|-----|-------------|----------|
| OBJECTIVE | OBJECTIVE | pixel length | um | pixels/um | um/pixel |
| 10x objective | 10 | 1257.0 | 850 | 1.479 | 0.676 |
| 40x objective NA=0.5 | 40 | 1175.6 | 200 | 5.878 | 0.170 |
| 40x objective NA=1.0 | 40 | 1174.0 | 200 | 5.870 | 0.170 |
| 60x objective NA=0.65 | 60 | 883.0 | 100 | 8.830 | 0.113 |
| 60x objective NA=1.25 | 60 | 885.0 | 100 | 8.850 | 0.113 |
| 100x objective | 100 | 739.0 | 50 | 14.780 | 0.068 |
| Binning = 1 | | Scale measureme | ent | | |

Note: Binning on the Sensys camera is set to 1.

Sensys KAF1401 cooled CCD replaced Oct 2009

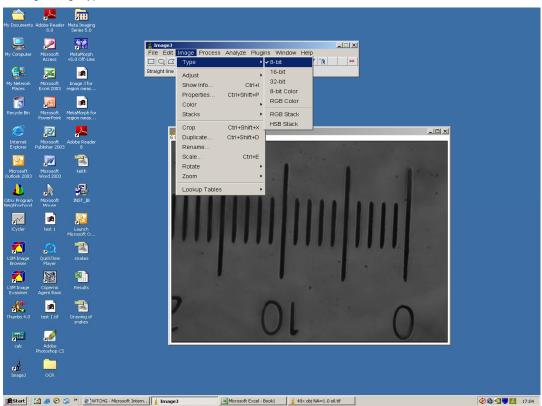
This is the maximum number of pixels - the camera is at maximum resolution Image size is 1312x1034 pixels [with binning set to 1]

Most users set camera binning to 2, you can check easily enough which setting was used by looking at the image size.

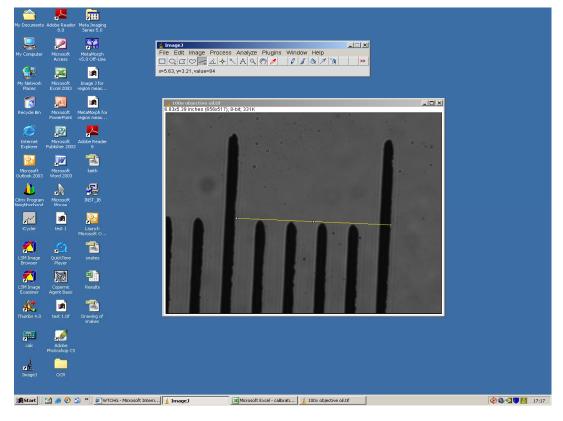


Calibrated using ImageJ

Use image, type, 8-bit (e.g. converted to 256 grey levels) Change image type from 36-bit RGB to 8-bit for set scale

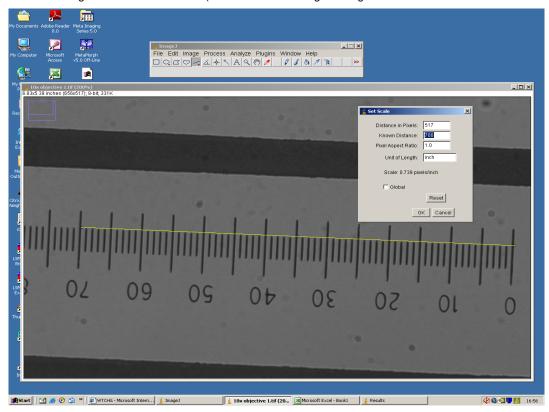


Use 'straight line' selection - the button is 'pushed in' [greyed out], right click for other options Draw a line with the mouse (left click down)



Use Analyze, Set scale to obtain the calibration

The scale length measured is 700 um (the scale is 1mm long and is graduated from 0 to 100 in units of ten)



You can use Analyze, Set Scale to input both values using the calibration data in this pdf file Select OK and the image will be calibrated for ImageJ area/length measurements in um etc..

See our Microscopy Web Site links for the freeware image analysis software ImageJ

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