

Calibration Error Correction Strategy

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This process can be used to recalibrate an inaccurately calibrated series, or if you have made a lot of traces but mistakenly forgot to calibrate the series first.

This strategy is covered in Ch 9 of the [Reconstruct User Manual](#) (p. 97-98).

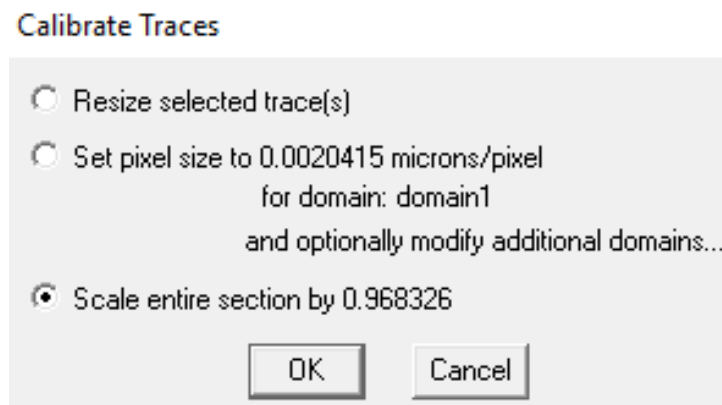
You should not use this strategy routinely -- it is only for correcting errors.

Always calibrate first!

This solution provides accurate quantification; however, it is not preferred because it simply applies a scale factor to your traces and domain images, rather than setting the correct pixel size in your domains from the start.

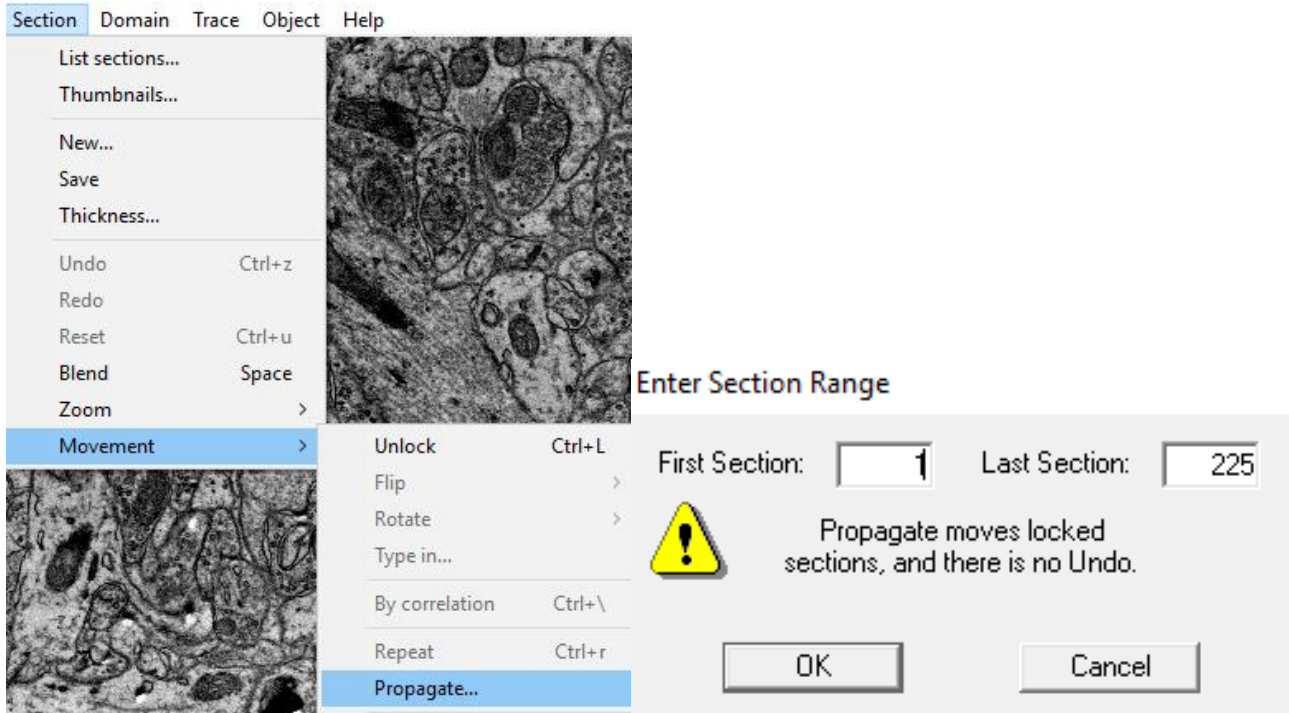
Read this entire protocol and make a backup of your trace files before starting.

1. Follow the normal calibration protocol: Place lines on cal grid > Trace > Calibrate > Enter New Length: type in the known scale length (4.63 um for 10 squares on a typical TEM calibration grid [Ted Pella Cat #607, [Diffraction grating replica](#), 2160 lines/mm]) > Enter it for each (multiple entries). Stop following the standard protocol when you get to this window:

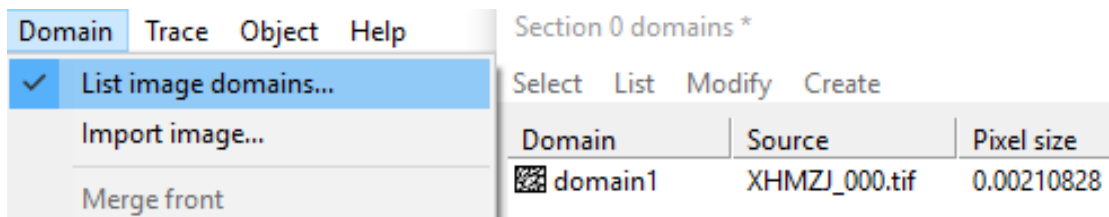


2. Deviating from the standard calibration protocol, select "Scale entire section by #####". That number in the window (0.968326 in the example window above) is the Scale Factor. ***Write down the scale factor now.*** You need to keep a record of that number, and Reconstruct will not show you that number again. After you have written it down, hit OK. For this section (the cal grid), the new scale factor will be multiplied across both images and traces so that they remain scaled to one another. This only works for the section you're on.

3. Immediately, as your next action, go to: Section > Movement > Propagate > DO NOT DO THE FIRST SECTION AGAIN. Don't re-do a section. Do, for example, 2-200 (if your cal grid is section 1) or 1-200 (if the cal grid is section 0). (Note: Dr. Harris prefers that ALL calibration grids be assigned section 0.) This must be done immediately next because it will propagate your last action.



4. Create a file for your series and write into that file a note indicating the procedure you applied here. Keep that file with the most up-to-date set of trace files. Indicate the scale factor that was applied. Although this fix gives the correct value, it doesn't change the pixel size -- so the pixel size listed in Reconstruct (Domain > List image domains) will be incorrect.



5. To find the actual pixel size, multiply: Scale Factor X the listed pixel size = new scaled pixel size. So the pixel size which is listed in Reconstruct after this "fix" will be incorrect, but you can calculate that value on your own by multiplying the Scale Factor by the listed pixel size, and indicate that information in your file associated with the series.

From the [Reconstruct User Manual](#) (p. 97-98):

Resize Traces, Set Pixel Size, or Scale Section

The end result of calibration can be one of three things: resizing the selected traces to the length specified, changing the pixel size of the domain in which the traces were drawn, or scaling all the transformations in the section (domains and traces). Which of these three operations will be performed is determined in the Calibrate Traces dialog (Fig. 9.2), presented after the lengths of all selected traces have been entered. Complete the calibration operation by choosing one of the options and selecting OK. No changes will be made if the Cancel button is selected.

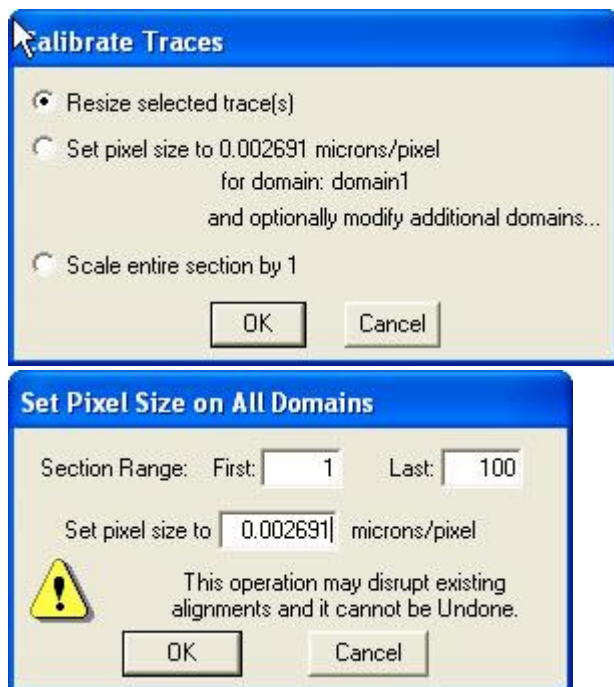


Figure 9.2. The Calibrate Traces dialog (left). Three options are provided for adjusting the section data based on the entered trace lengths. When the second option is chosen a subsequent dialog (right) allows for applying the pixel size adjustment to all domains in the series.

The "Resize selected trace(s)" option will only modify the selected traces. The transformations of the traces are scaled so that the traces will be redrawn with the specified new lengths. Each trace is scaled independently rather than to an average rescaling value as in the other options.

The domain on which the selected traces are centered is listed in the second option. If the calibration traces are not centered on a domain, then this option will be disabled. The correct pixel size of the domain will be set by selecting the second option. This pixel size value is computed by transforming the selected traces into the domain and computing their lengths on the image in pixels. The sum of the entered lengths (in units) is divided by the

sum of lengths in pixels to get the correct pixel size for the calibration grid. This pixel size can be subsequently applied to all domains as discussed in the next section.

Selecting the second option will only change the pixel size of domains, not of traces in the section. If the section has lots of traces and domains it may be advantageous to choose the final option instead. This option computes a scaling factor based on the ratio of the sum of the entered lengths divided by the sum of the current lengths. The scaling factor is applied to the transformations of all domains and traces in the section, effecting the calibration by scaling rather than setting the pixel size.

It is desirable to have the pixel size parameter accurate for later reference, since the scaling factor will not be visible to the user. So the second option should be used in the Calibrate Traces dialog for the initial calibration. With this second option existing traces will no longer be correct and existing alignments between domains will be disrupted. This is not a problem if calibration is carried out before any domain movements, section alignments, or tracing. However, the calibration traces will be incorrectly sized after a pixel size adjustment. These calibration traces may be simply deleted. To verify the calibration, redraw the calibration traces and check their length to see that they match the known lengths in the calibration image.

Apply the Calibration to Other Sections

The pixel size set by the "Set pixel size..." option can be subsequently extended to other domains in the series. When this option is chosen the calibration automatically opens the "Set Pixel Size on All Domains" dialog box (Fig. 9.2). This dialog displays the same pixel size value as in the Calibrate Traces dialog and allows a range of sections to be selected. All domains in the included sections will be set to the pixel size value.

Note: This operation cannot be undone when executed over many sections.

The rescaling performed by the third option of the Calibrate Traces dialog can be applied to other sections in the series by using the Repeat or Propagate... commands from the Movement submenu. To rescale one other section, unlock the section and select Repeat (or Ctrl-R). To rescale a range of sections, use Propagate... and specify the range of sections to modify. Be careful not to rescale the current section again, because this section was already rescaled by the Calibrate Traces dialog.

If the first calibration option "Resize selected trace(s)" was chosen in the Calibrate Traces dialog, then no multi-section application of the calibration is possible.
