

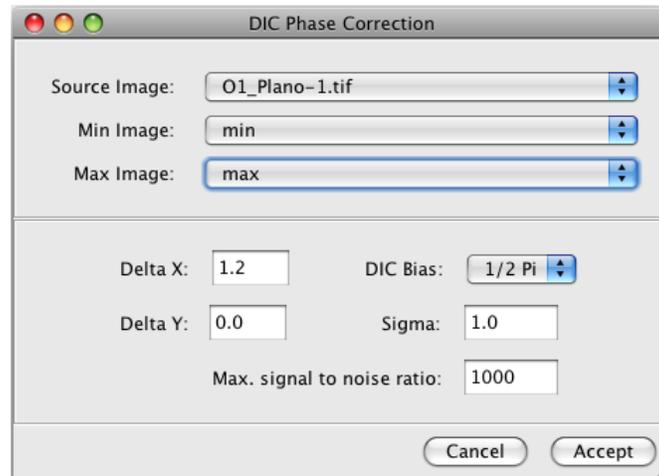
# NEURITE, QUICK REFERENCE MANUAL

## Average Background

Generates a new background image sequence from the active image sequence, where each frame of the background sequence is the average of all the previous frames on the sequence.

## DIC Phase Correction

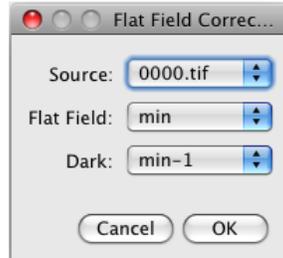
Corrects the shadow-cast effect of Differential Interference Contrast (DIC or Nomarsky) images. This operation is applied to the active image, but requires two other opened images: minimum and maximum light condition images. In the *plug-in* window you must choose other parameters such as Phase shift in X and Y directions in pixels, the selected bias of the DIC microscope and the signal to noise ratio (SNR) applied to the output image. For any image sequence you only need a one minimum and a one maximum image, and the process will be applied to whole sequence.



For a detailed description about this method and the significance of the parameters see Van Munster, E. B., Van Vliet, L. J., and Aten, J. A. (1997). *Reconstruction of optical path-length distributions from images obtained by a wide-field differential interference contrast microscope*. *Journal of Microscopy*, 188(2), 149.

## Flat Field Correction

Applies a flat field correction to an image or image sequence. This operation is applied on the active image, but needs two other opened images: an offset image and a flat image. The offset image can be obtained with the microscope light turned off, and a flat image can be a defocus image or a snapshot without specimen in the preparation. You must select each image from the *plug-in* window.



## Normalize Light

Normalizes the illumination along the sequence, using as a reference the current image displayed in the active sequence.

## Trim Borders

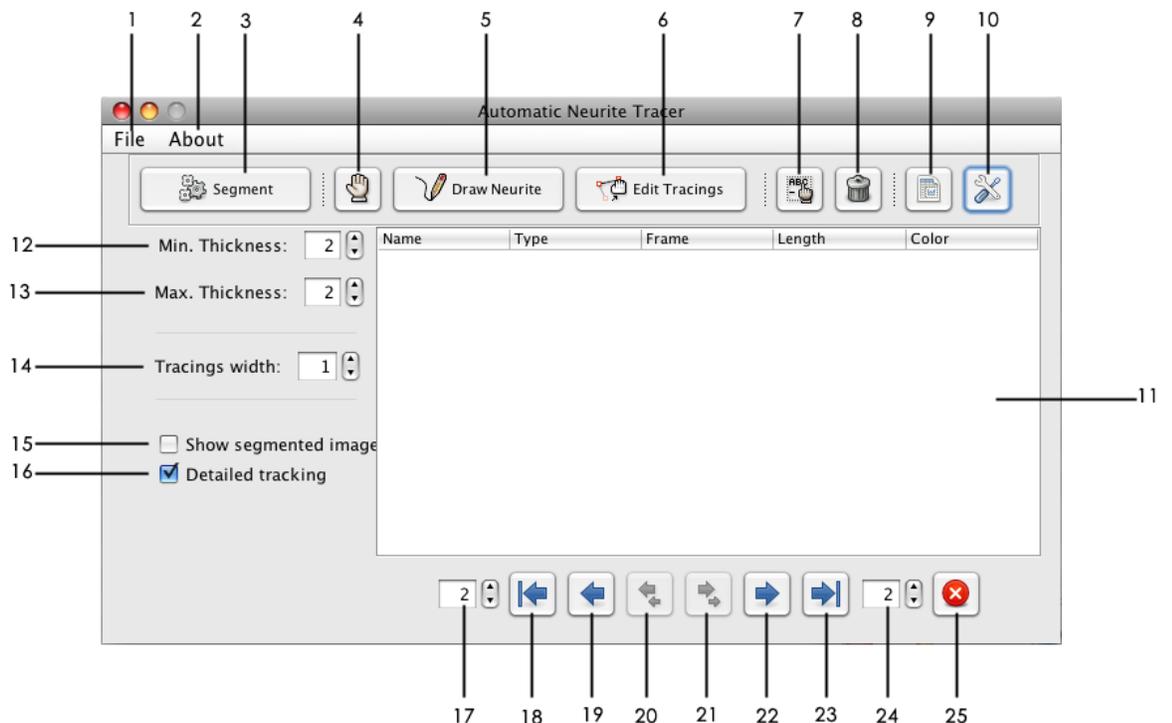
Automatically crops an image or image sequence if it has a dark frame. After cropping it displays the cropped image or image sequence in a new window.

## Neurite Tracings

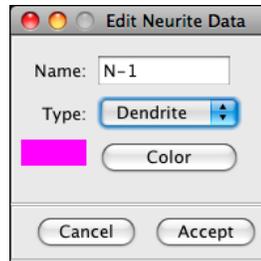
Opens a *plug-in* containing all the tools needed for automatic or semiautomatic neurite tracing and quantification. A detailed view is show below.

- 1. File Menu:** Contains different options for open, save and export the tracings.
  - **Open Tracings:** Opens a tracings file.
  - **Save Tracings:** Save an opened tracing file with the current modifications.
  - **Save Tracings File:** Creates a new tracing file.
  - **Save Trace Coordinates:** Creates a new file containing all the x, y coordinates of the traces.

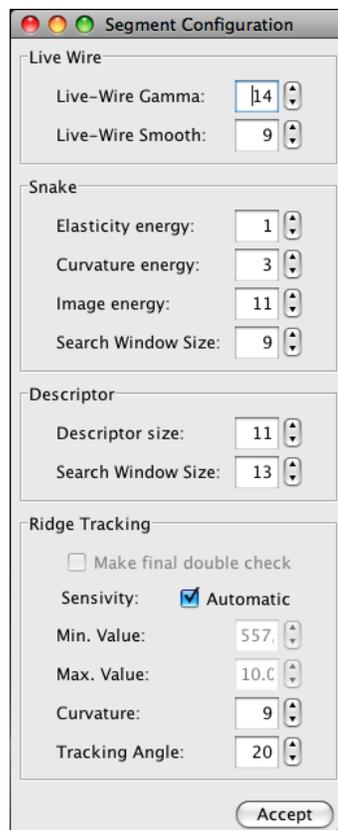
- **Export Lengths to Excel:** Exports the current trace lengths to an Excel file. Data are ordered by frame and by tracing name.
- **Export Trace to a Binary Image:** Exports the traces, to a new sequence. This is useful to made a Scholl analysis with other ImageJ *plug-in*.
- **Exit:** exits this *plug-in*.



2. **About Menu:** show the current version of this NEURITE *plug-in*.
3. **Segment Button:** Performs an eigen analysis on current frame.
4. **Automatic/Manual Button:** Switches between automatic and semiautomatic neurite tracing along the sequence.
5. **Draw Neurite Button:** Allows tracing the neurites in a semiautomatic way.
6. **Edit Tracings Button:** Allows to edit the traces on the current image. The traces must be made by the user or by NEURITE in an automatic way.
7. **Edit Info Button:** Opens a pop up window to edit the information (name, type and color) of the selected neurite in the “results window” (11).



8. **Delete Info Button:** Deletes the selected neurite(s) in the “results window”. This operation also erases the corresponding traces from the image or sequence.
9. **Save Info Button:** Exports the information of all the neurite traces that appear in the “results window” to an Excel spreadsheet.
10. **Parameters Button:** Opens a parameters window. These parameters are used to tune the automatic neurite tracing and tracking along the sequence.



11. **Results Window:** Shows the; name, type, length and selected color of each traced neurite.

- 12. Minimum scale parameter:** Fixes the value in pixels corresponding to the neurite present in the current active image or sequence. This parameter is used in the eigen analysis made with the “Segmentation Button”
- 13. Maximum scale parameter:** Fixes the value in pixels corresponding to the neurites present in the current active image or sequence. This parameter is used in the eigen analysis made with the “Segmentation Button”.
- 14.** Width in pixels of the traces in the current image or image sequence.
- 15.** Shows the eigen-image, of the active image or the current image in the sequence.
- 16.** Makes a detailed tracking of the neurite, when an automatic process is made.
- 17.** Number of images to trace neurites in automatic mode in backwards manner along the sequence.
- 18.** Starts the automatic tracing along the sequence backwards along the images selected in (17).
- 19.** Starts the automatic tracing backwards only to the next image.
- 20.** Performs an eigen analysis on the previous image of the sequence, allowing semiautomatic neurite tracing. To facilitate the process of neurite characterization, a selection of the previously characterized neurites shown in the neurites “results window”, is required.
- 21.** Performs an eigen analysis on the next image of the sequence, allowing de semiautomatic neurite tracing. To facilitate the process of neurite characterization, a selection of the previously characterized neurites shown in the neurites “results window”, is required.
- 22.** Starts the automatic forward tracing along the number of selected images only to the next image.
- 23.** Starts the automatic forward of images selected in (24).
- 24.** Number of images to trace in automatic forward manner.
- 25. Stop:** Stops the automatic neurite tracing along the sequence.