

Multi-dimensional Motion Analysis Guide

Overview

- Multi-dimensional Motion Analysis (MDMA) is used to track position of objects within a z-stack over time. Other common measurements such as object velocity can then be derived from this information.
- MDMA data can be presented in tabular form or by using the integrated graphing utilities. All data can be exported to Excel or text files.
- Individual objects in the field are automatically tracked and data is graphically presented on the output image.

ARTICLE #
T20102

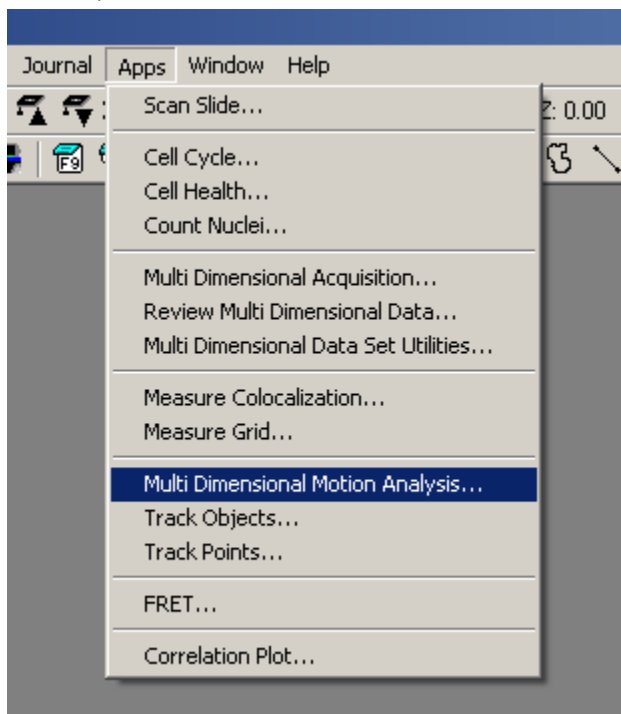
PRODUCTS
MetaMorph®

CREATED
26-January-09

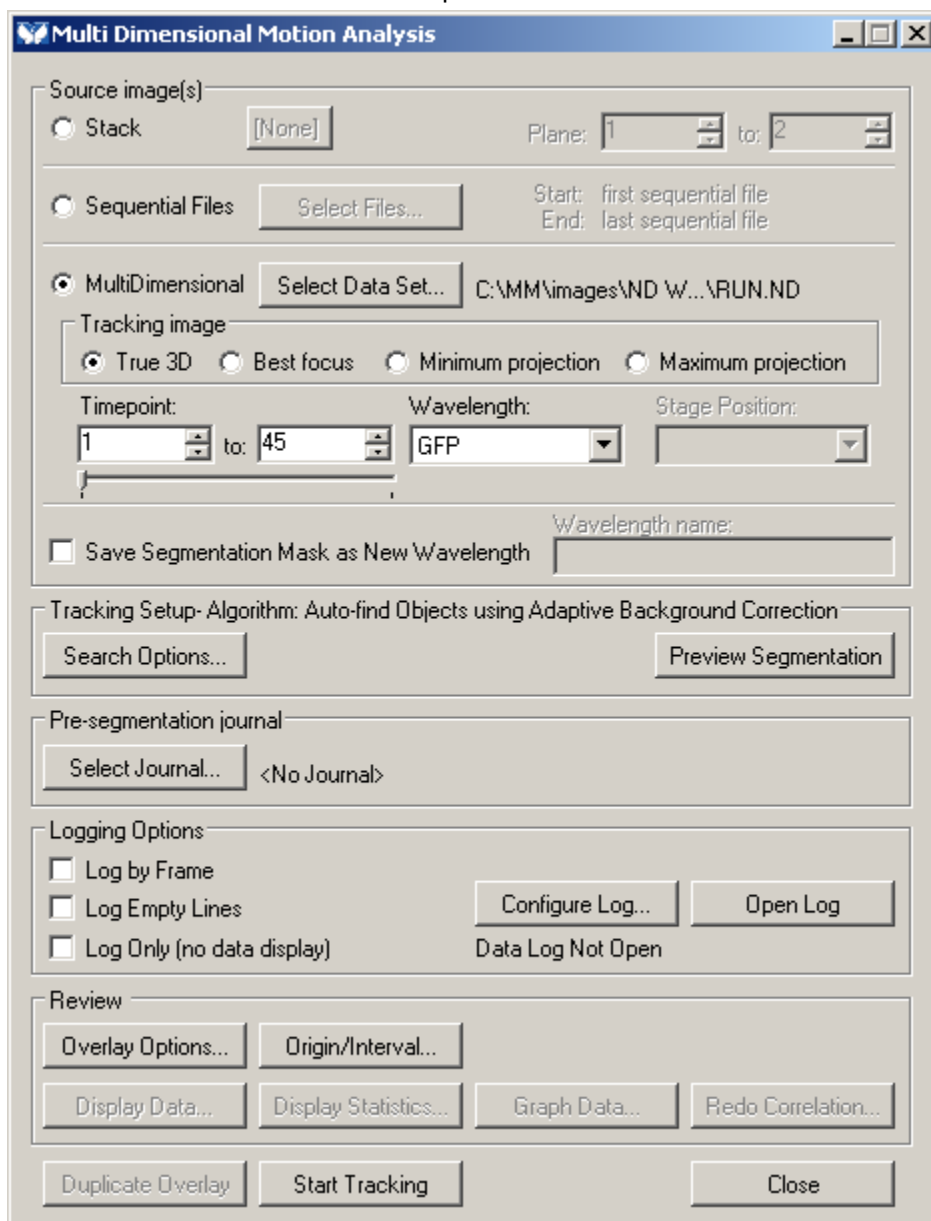
LAST UPDATED
30-January-09

Opening Interface

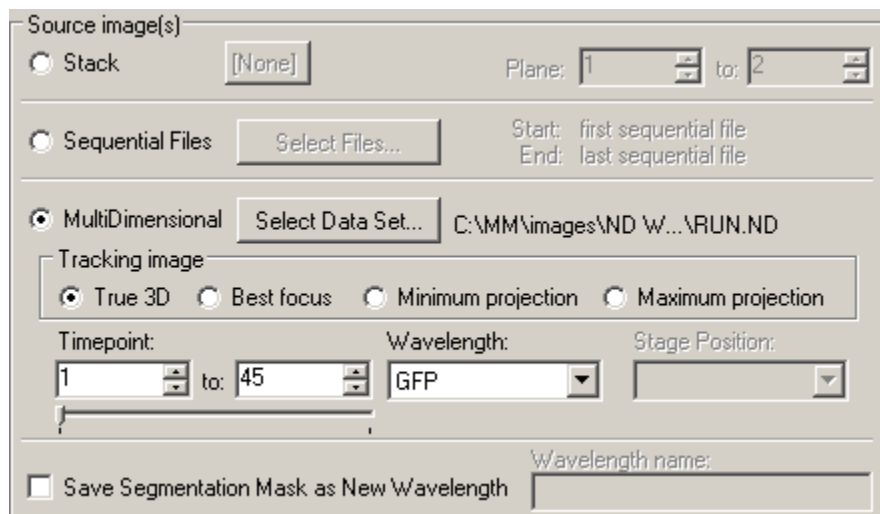
The MDMA application module is found under the Apps menus. If it is not present you may need to add it by using the “Drop-ins/Toolbars” button within the MetaMorph® Administrator application (see article T20103).



Once selected the MDMA interface opens.

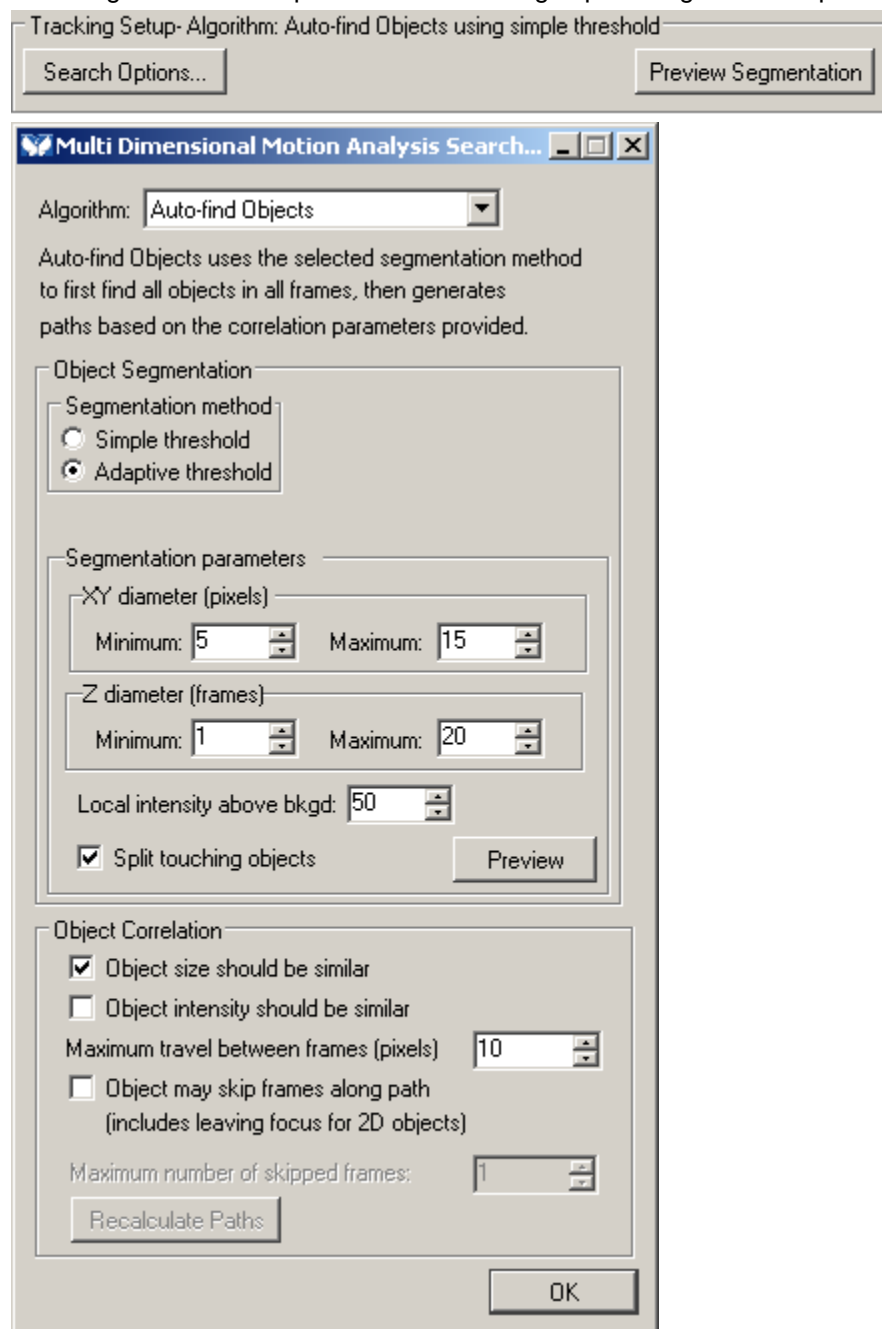


The first part of the interface is dedicated to selecting the image set you want to analyze.



- The “Stack” option is for analyzing single planes from a time series. This is useful for 2D analysis.
- “Sequential Files” works the same as the “Stack” option except you can select images saved sequentially from an application other than the MetaMorph® program.
- The “MultiDimensional” option along with the “True 3D” setting allows for object analysis across all spatial dimensions (x,y,z).
- The “Select Data Set...” button allows you to select a data set generated with the Multi-dimensional Data Acquisition (MDA) tools within the MetaMorph® program. This is the most common way to perform MDMA.
- If the data set contains multiple wavelengths, you need to use the drop down “Wavelength:” menu to select the appropriate channel. You also have the option of selecting a subset of time points for analysis. Selecting a subset can speed the analysis, especially useful if you are trying to determine the optimal segmentation settings.
- Finally, you can save the intermediate segmentation mask generated by the MDMA by activating the check box and providing a name in the text field.

Selecting the “Search Options...” button brings up the segmentation parameters interface.

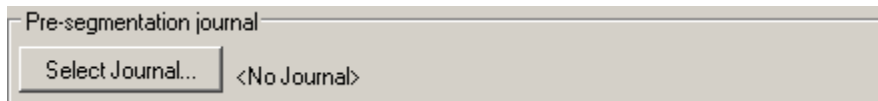


- There are three different methods for identifying objects, however, “Auto-find Objects” will be used most often.
- Most applications will benefit from selecting the “Adaptive threshold” option as this helps segment objects even in the presence of uneven background and other image artifacts.
- The segmentation parameters are used to optimally segment and identify your objects of interest. As you adjust the parameters it is convenient to use the “Preview” button to get an indication of the

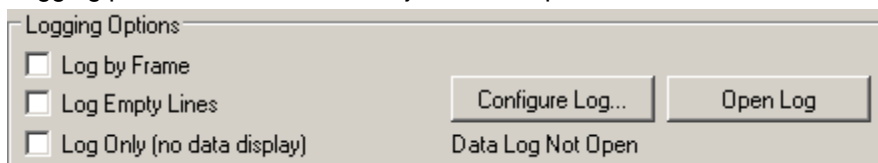
accuracy of segmentation. By using the image measurement tools, you can get an estimate of the min and max diameter of your objects in x, y and z. In addition, you can identify the pixel intensity difference between the object and its local background by hovering the pointer over a pixel of interest and reading the intensity information in the status bar of the MetaMorph® program.

- The “Object Correlation” options allow you to specify additional cues for identifying the moving objects.

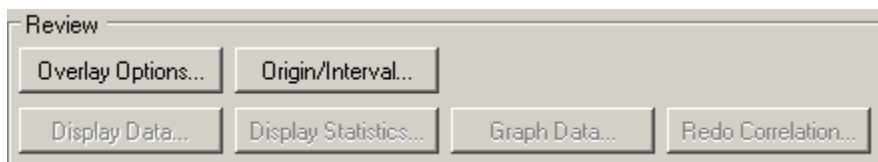
A journal can be selected to run prior to image segmentation. This can be used to process the image in such a way to improve the segmentation algorithm.



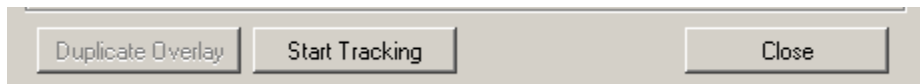
Logging parameters of tracked objects is setup in this section.



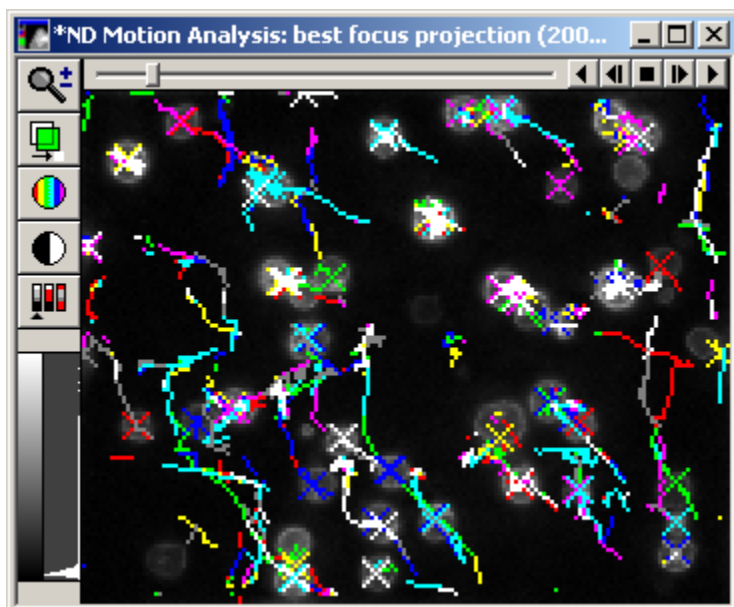
The “Review” section allows you to configure the display of different tracking data on the in output image as well as graphically. It is often useful to limit the amount of data presented to make it easier to identify tracks of interest.



Finally, the actually tracking analysis is started by pressing the “Start Tracking” button.



Once the analysis is complete you are presented with a new image.



- Each track is presented as a line and a different color. The track attributes can be modified using the "Overlay Options..." button in the "Review" section of the dialog.
- By dragging the scroll bar thumb at the top of the image, you can see how the individual objects move over time by observing the progression of the "X" marker along the track.
- The object tracks to display can be modified by using the "Display Data..." button.

The “Graph Data” button can be use the correlate two parameters as in this example.

