

Together through life sciences.

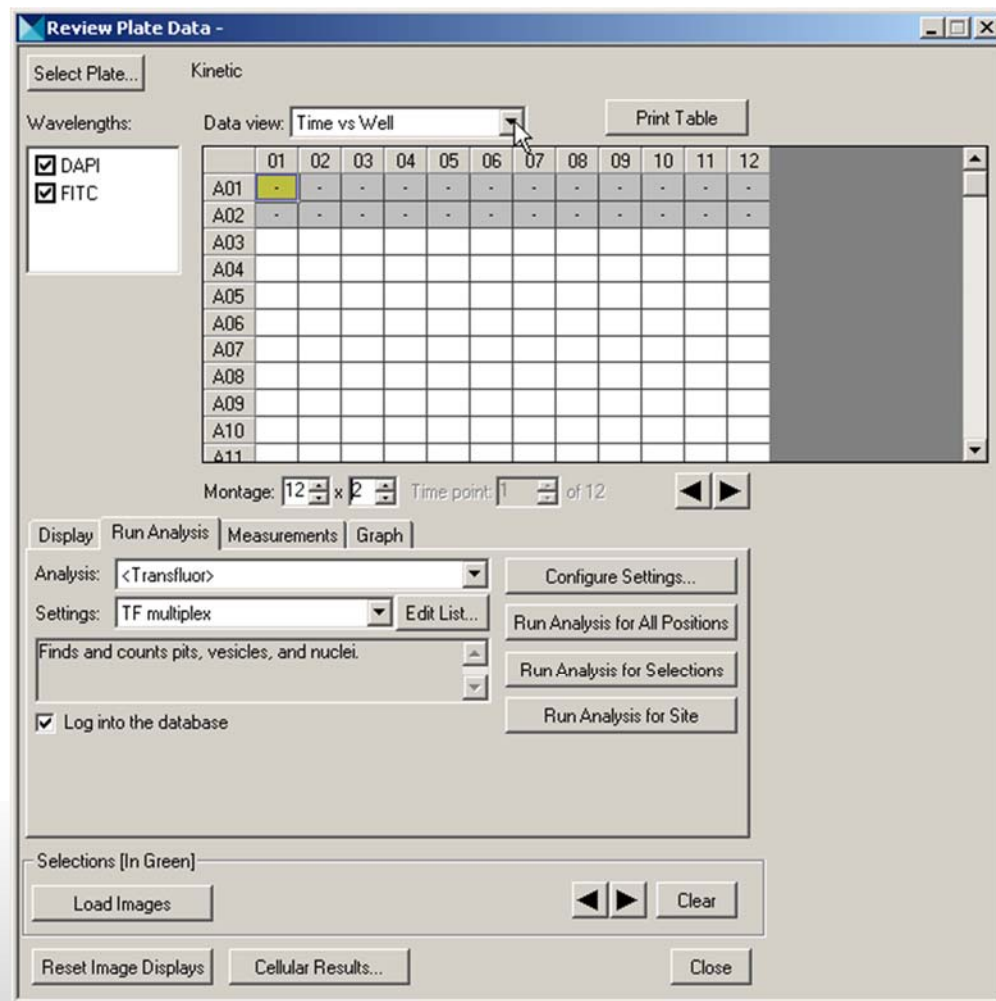


Multi Dimensional Motion Analysis

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April 29, 2013

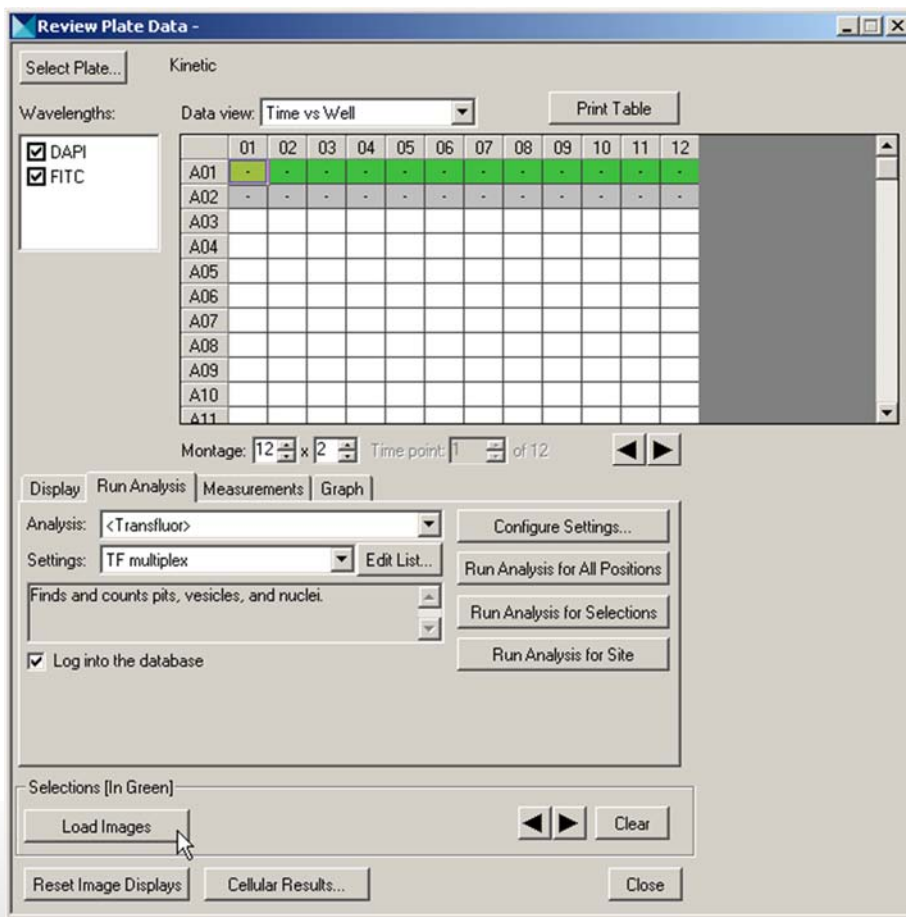
View time lapse / kinetic data in Review Plate Data

- Select the plate of interest
- Choose the “Time vs Well” data view
- The montage will show thumbnails of each time point
- To create a stack (for viewing as a movie), right-click on ONE WELL ONLY to select it.
- Click on “Load Images” to create the stacks.
- You may now view each timepoint as a plane in the stack.
- The stack can now be used for analysis with MDMA



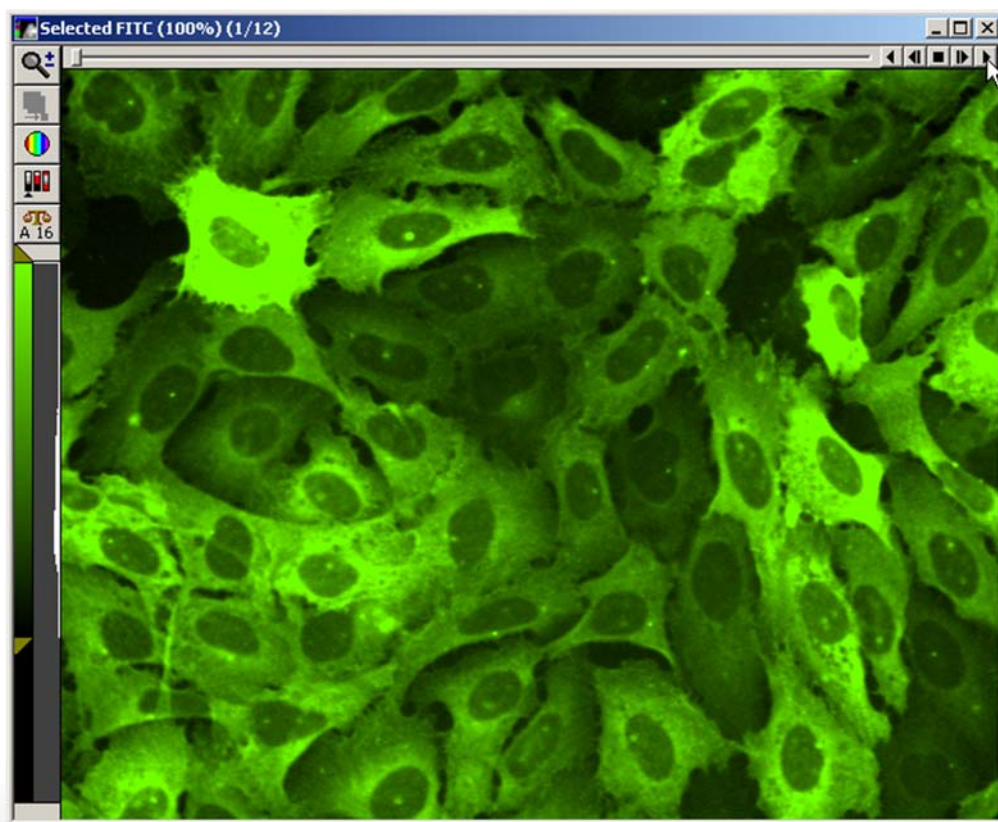
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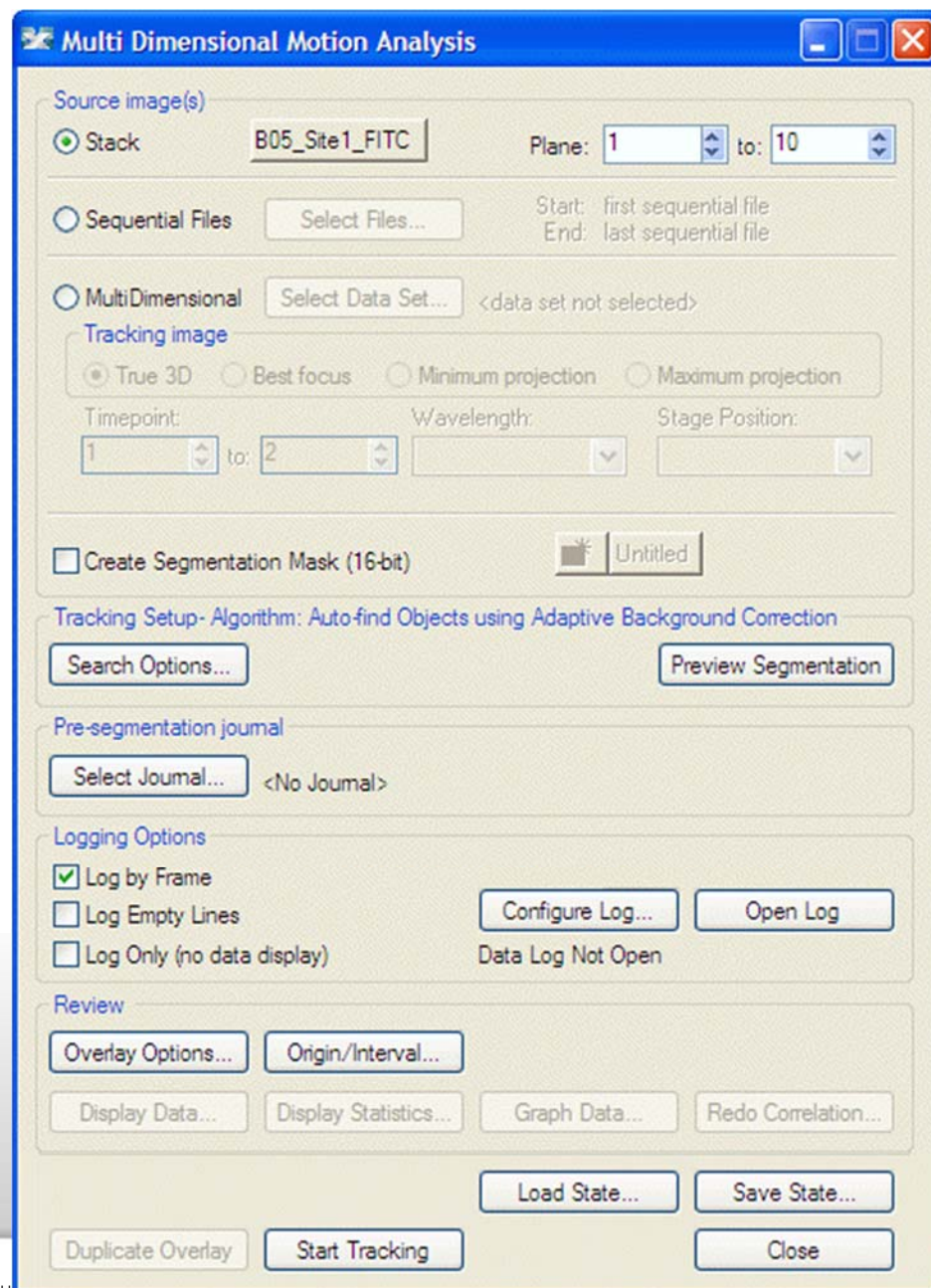
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Multi Dimensional Motion Analysis

- Use for analyzing motion of multiple objects over time
- Get data per cell per time point as well as summary data over entire time course



MDMA – Finding objects

Multi Dimensional Motion Analysis...

Algorithm: **Auto-find Objects**

Auto-find Objects uses the selected segmentation method to first find all objects in all frames, then generates paths based on the correlation parameters provided.

Object Segmentation

Segmentation method

☐ Simple threshold

☒ Adaptive threshold

☐ Current IMA settings

Open IMA...

Segmentation parameters

XY diameter (um)

Minimum: **12** Maximum: **28**

Z diameter (frames)

Minimum: **5** Maximum: **15**

Local intensity above bkgd: **40**

☒ Split touching objects

Preview

Object Correlation

☒ Object size should be similar

☒ Object intensity should be similar

Maximum travel between frames (um) **10**

☒ Object may skip frames along path
(includes leaving focus for 2D objects)

Maximum number of skipped frames: **1**

Recalculate Paths

OK

Multi Dimensional Motion Analysis...

Algorithm: **Template Match**

Template Matching uses an image convolution to compare each object's intensities with the values in the first or preceding image.

If object not found:

☐ Select Position ☐ Quit Object ☒ Skip Frame

☐ Use velocity for center of next search

Delay: **0** sec/object

☐ Update Template Each Frame

Minimum % for match: **50**

☐ Use Derivative of Image
(recommended for non-DIC images)

Multi Dimensional Motion Analysis...

Algorithm: **Threshold Result**

Threshold Result calculates the intensity centroid of the object above threshold and finds the nearest centroid in the next frame.

If object not found:

☐ Select Position ☐ Quit Object ☒ Skip Frame

☐ Use velocity for center of next search

Delay: **0** sec/object

Object size match requirement (as %): **50**

MDMA – Display and Other Options

ND Motion Analysis Origin/Time ...

Origin Options

☐ Top/left corner of image

☐ First point in track

☒ Corresponding point of first object

Time Interval Options

☒ Image creation time

☐ User defined Units:
 Interval: 1 Seconds

OK Cancel

Multi Dimensional Motion Analysis Data

	Object 392	Object 393	Object 394	Object 395
Display-	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>
Frame 1	522.45, 1251.30)	549.86, 1251.30)	378.81, 1260.98)	199.88, 1264.20)
Frame 2	522.45, 1251.30)	549.86, 1251.30)	380.43, 1262.59)	199.88, 1265.81)
Frame 3	520.84, 1249.69)	548.25, 1251.30)	378.81, 1259.36)	199.88, 1265.81)
Frame 4	522.45, 1249.69)	549.86, 1251.30)	380.43, 1259.36)	199.88, 1265.81)
Frame 5	522.45, 1246.46)	551.48, 1249.69)	377.20, 1260.98)	199.88, 1262.59)
Frame 6	520.84, 1248.08)	551.48, 1249.69)	378.81, 1257.75)	199.88, 1264.20)
Frame 7	522.45, 1244.85)	553.09, 1246.46)	377.20, 1256.14)	501.49, 1260.98)
Frame 8	522.45, 1243.24)	553.09, 1246.46)	378.81, 1256.14)	501.49, 1260.98)
Frame 9	519.23, 1241.63)	551.48, 1244.85)	378.81, 1256.14)	501.49, 1259.36)
Frame 10	522.45, 1241.63)	553.09, 1244.85)	375.59, 1254.53)	503.10, 1259.36)

Position Show All Tracks Edit Data...
☒ Highlight Selected Track Hide All Tracks Print Table Close

Multi Dimensional Motion Analysis Overlay Options

Data Point Markers

Size: 7

Color: Alternating

Type: Cross

☐ Fill Circle Markers

Display Mode

☐ All Points

☒ Points on Current Plane

Object Tracks

☒ Display Track Path

☐ Limit Path Displayed
 Frames before current: 5
 Frames after current: 5

☒ Display Track Number

☐ Display Track Pattern
 Color: Red

OK Cancel

MDMA – Measurements

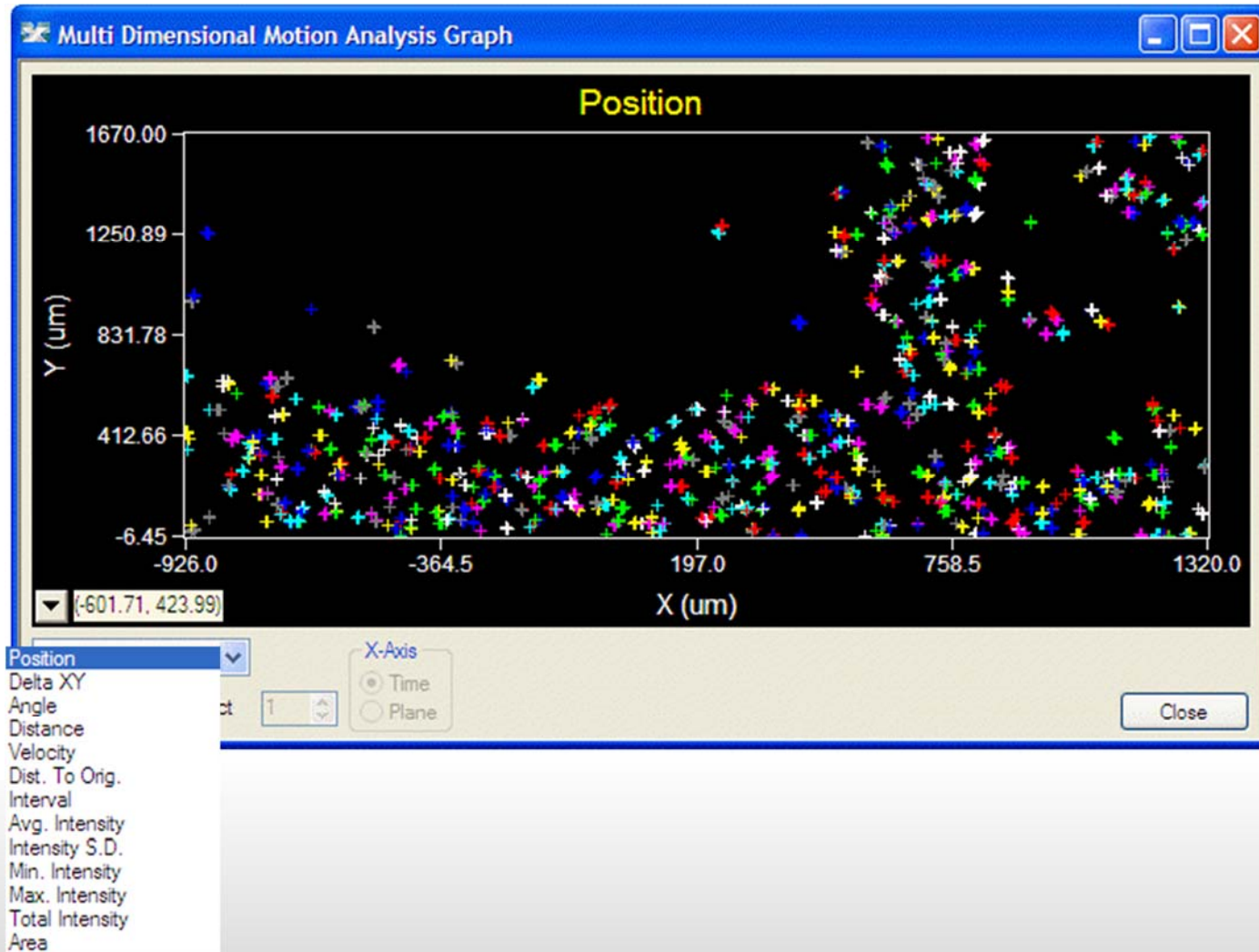
- Cell-by-cell, per time point

- ✓ Object #
- ✓ Frame #
- ✓ X
- ✓ Y
- ✓ Z
- ✓ Distance
- ✓ Time Interval
- ✓ Velocity
- ✓ Absolute Angle
- ✓ Distance To Origin
- ✓ Delta X
- ✓ Delta Y
- ✓ Delta Z
- ✓ Average Intensity
- ✓ Intensity S.D.
- ✓ Minimum Intensity
- ✓ Maximum Intensity
- ✓ Total Intensity
- ✓ Area

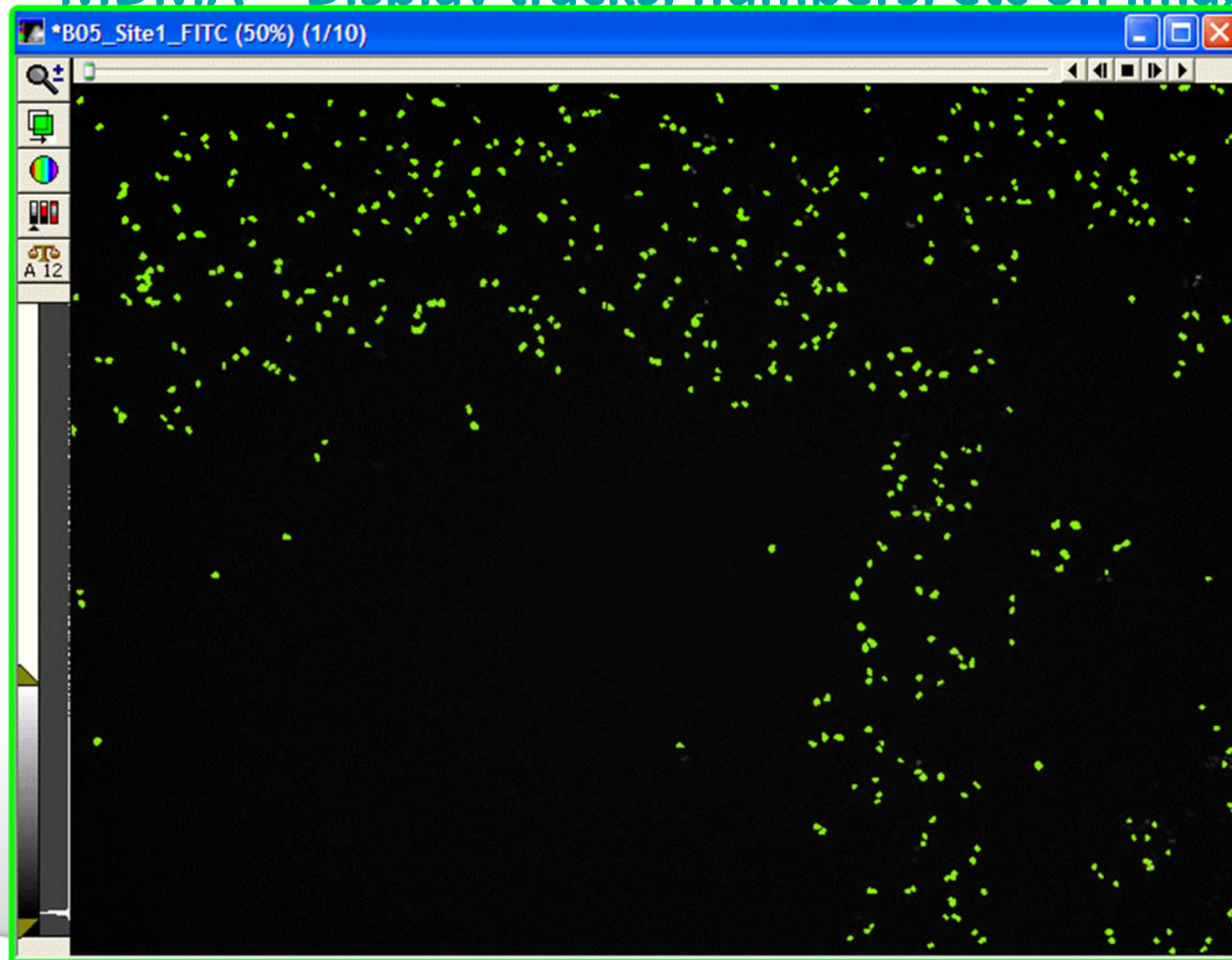
- Cell-by-cell, statistics over time course

- ✓ Mean X
- ✓ Mean Y
- ✓ Mean Z
- ✓ Mean Distance
- ✓ STD Distance
- ✓ Mean Angle
- ✓ Mean Angle Vector
- ✓ Angular Deviation
- ✓ Mean Velocity
- ✓ STD Velocity
- ✓ Mean Area
- ✓ STD Area

MDMA –Graphing



MDMA – Display tracks, numbers, etc on image



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