Package Description:

U-Track identifies and tracks objects in light microscopy time-lapse sequences. It is the MATLAB implementation of the tracking algorithm described in the paper "Robust single particle tracking in live cell time-lapse sequences" by **Jaqaman et al.**, **Nature Methods 5: 695-702 (2008)**.

U-Track consists of 3 steps:

- (1) **Detection**: Identifies the objects in each frame and returns information such as their positions and intensities.
- (2) Tracking: Takes in the objects identified in the first step and constructs their trajectories over the course of the movie.
- (3) Track analysis: Takes in the tracks computed in the second step and runs application-specific trajectory classification.

Tracking applications

While the tracking algorithm described in the Nature Methods paper is general, the currently supplied code is implemented for three tracking applications:

- 1. Sub-resolution objects, such as single molecules or small molecular aggregates that are smaller than the diffraction limit.
 - See Jaqaman et al. Cytoskeletal control of CD36 diffusion promotes its receptor and signaling function Cell 146: 593 606. (2011).
- 2. Microtubule plus-ends marked with labeled +TIP proteins such as EB1 or EB3. See Applegate et al. Quantitative image analysis software for the measurement of microtubule dynamics. J Struct Biol, 176, 168-184 (2011)
- 3. Labeled nuclei of migrating cells e.g. using histone markers
 See Ng et al. Substrate stiffness regulates cadherin-dependent collective migration
 through myosin-II contractility. J Cell Biol, 199, 545-563 (2012)

The detection, tracking and track analysis results will be saved in the analysis directory where movieData has been set up.

All steps require the definition of various parameters, done by clicking on the "Setting" buttons.

All steps also come with routines for visualizing the results, done by clicking on the "**Result**" buttons. Visualization is enabled only when there are results to view.

If analyzing multiple movies in a batch job, make sure to check "Run all movies."

To rerun detection and/or tracking on a previously analyzed dataset, check "Force Run" if necessary.

Command line

The code is written in a modular fashion and the detection and tracking steps are independent of each other.

To make full use of the software's flexibility, call the detection and tracking modules from the command line using "scriptDetectGeneral" and "scriptTrackGeneral." With this, you can use your own code to detect the objects of interest, e.g. if the objects are not sub-resolution features, and then you can call the tracker using "scriptTrackGeneral" to link the objects between frames. You can also write your own cost functions to optimize the tracker for a particular data type. See the non-GUI README for more details.