

Process Description:

“**Comet detection**” identifies near-resolution sized comets using a combination of difference of Gaussian filter and watershed segmentation. For a description of the algorithm, see **Applegate et al. Quantitative image analysis software for the measurement of microtubule dynamics. J Struct Biol, 176, 168-184 (2011).**

Parameter Descriptions:

Input channels:

This allows you to select which channels you want to detect objects in. Select the channels by clicking on them in the "Available Input Channels" box and then clicking "Select>" to move them to the "Selected Channels" box. You can unselect a channel by clicking the "Delete" button.

Mask process to be used for detection: If any segmentation process has been performed on the movie, this drop-down menu allows to select the mask process which output should be used when detection comets.

Range of frames to detect: These values determine the range of frames to perform the comet detection on.

Band-pass filter parameters:

Low-pass Gaussian standard deviation: This value is used for the low-pass Gaussian filter applied to the image to eliminate high-frequency intensity fluctuations due to noise.

High-pass Gaussian standard deviation: This value is used for the high-pass Gaussian filter applied to the image to eliminate larger-scale variations in cell background.

Watershed segmentation parameters:

Minimum threshold: This value is used to compute the minimum threshold to detect comets during the watershed segmentation. It is expressed as a multiple of the image intensity standard deviations (averaged over 5 neighboring frames).

Threshold step size: This value is used as the step size between threshold levels during the watershed segmentation. It is expressed as a multiple of the image intensity standard deviations (averaged over 5 neighboring frames).

Process output:

The important output variable stored in the .mat file defined above is “movieInfo”, which contains the detected particle information:

For a movie with N frames, movieInfo is a structure array with N entries.

Every entry has the fields xCoord, yCoord and amp.

If there are M features in frame i, each one of these fields in movieInfo(i) will be an Mx2 array, where:

the first column is the value (e.g. x-coordinate in xCoord and intensity in amp),
and the second column is the standard deviation.