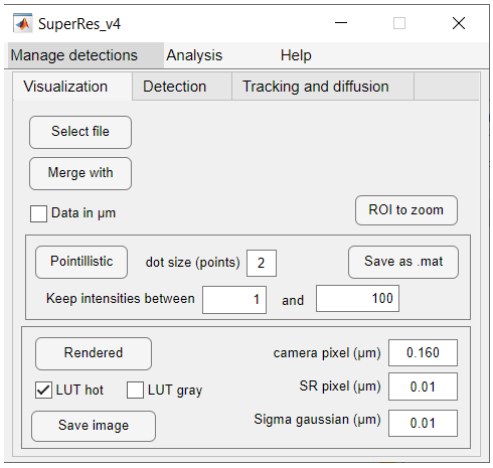
SuperRes_v4_App

Menu for detection, tracking, analysis and visualization of data generated by single-molecule super-resolution microscopy (PALM, STORM, sptPALM, uPAINT…).



Tabs

Visualization:

Creates pointillistic and rendered images from SMLM data (.mat files). The algorithm that creates the rendered image is based on PALMvis.m scripts (Pasteur Institute, Paris).

The Pointillistic image shows the position of detections, for one data set or two (Merge). The Rendered image is the interpretation of the coordinates considering a Gaussian distribution of intensity, with variance equal to Sigma Gaussian and a super-resolved pixel size (SR pixel). These two parameters are determined by the localization precision of the acquisition.

Detection:

Provides coordinates and data about single molecule detections in a movie in .tif format. Based on Multiple-target tracing (MTT) algorithm (Arnaud et al 2008).

Tracking and diffusion:

When the acquisition is obtained from living cells, it is possible to track the positions of each molecule by creating trajectories based on detections. Trajectories then can be used to analyze lateral diffusion as described in Renner et al, (2017).

Menus

- **Manage detections**

This menu offers GUIs to:

- verify detections (**Check quality**),
- correct the shift of the microscope stage (**Correct drift**)
- correct the shift between acquisitions done on two different channels (**Correct color shift**).
- correct for multiple detections of the same fluorophore (**Correct multiple detections**).

Finally, the option **Concatenate data** allows to create a single detection file from several acquisitions made sequentially on the same sample.

• **Analysis**:

Menu for launching the
- **Segmentation** of SMLM data, (Paupiah et al 2023)
- **Clustering** analysis, (Paupiah et al 2023)
- **Colocalization** analysis.

These analyzes work on detections obtained on fixed samples.

• **Help**: no need to describe this!

How to name the files?

Please pay attention to the naming of the files!

Most of the analysis steps are done automatically, often linking different files. In order to link them, the programs use "identifiers": series of characters that identify a type of file. Typically names are:

Movie of detections: name-number_identifier.tif

- name: invariant part (i.e. identifies the experiment)

- number: numbering of image acquisition

- identifier: labeled object, molecule, etc.

Examples: control-01_kv2.tif movie of the detections, number 1, in control conditions, of the molecule kv2.

control-01_ank.tif movie of the detections, number 1, in control conditions, of the molecule ank.

Image for localization: name-number_identifier-identifiermask.tif

Examples: control-01_kv2-mask.tif localization file "mask" for the movie of the detections number 1, in control conditions, corresponding to the molecule kv2.

If the program does not recognize files, it will display an error message or it will not execute the analysis properly…

References:

Arnaud et al 2008, Nat Methods. 2008 5:687-94. doi: 10.1038/nmeth.1233.

Paupiah et al. 2023, Biol Imaging e14, doi: 10.1017/S2633903X23000156.

Renner et al. 2017, Biophys J. 113:2452-2463. doi: 10.1016/j.bpj.2017.09.017.