Gamma-Norm Image
Colocalization Analysis (GICA)
Fiji Plugin (Release Candidate 2)

Fabian Humpert∗ & IdirYahiatène†

Biomolecular Photonics
Department of Experimental Physics
Bielefeld University, Germany
Head: Prof. Dr. Thomas Huser‡

If you encounter any bugs or questions please feel free to contact the developers.

Installation Instructions (Fiji 32bit)

Remark:
The installation of GICA is straightforward assuming a running and up-to-date version of Fiji. Otherwise Fiji can be downloaded from [http://fiji.sc/wiki/index.php/Downloads](http://fiji.sc/wiki/index.php/Downloads) as freeware. In this version GICA needs to run under Windows XP/Vista/7 (64/32 bit) using the 32bit version of Fiji.

Installation:
1. Extract GICA.zip to the Fiji main directory.

2. Restart Fiji or refresh the menus.

Three new entries will appear under the Analyze>Colocalization menu:

GICA This module only works on a certain region of interest (ROI). So to use this a ROI has to be chosen prior to the execution of this module. If no ROI is chosen the entire image is processed. GICA is supposed to run with standard settings. After executing the calculation GICA outputs a results table containing all relevant information on this specific ROI.

∗fhumpert@physik.uni-bielefeld.de
†iyahiate@physik.uni-bielefeld.de
‡thomas.huser@physik.uni-bielefeld.de
**Gamma-Norm Topology** This module calculates a whole topology map indicating areas with high numbers of colocalizations in order to have an orientation on the sample. The user may fine tune the resolution of the reconstructed image and the number of iterations to be used (10-20 as initial values are a good choice although the number of iterations is automatically computed). The method outputs the plain topology map (where the pixel values represent the respective Gamma-Norms) and additionally an overlay of the topology map and the original image (Therefore the overlay is cropped to fit the reconstructed topology map dimensions).

**Gamma-Norm (fast)** This sub-menu holds experimental modules for slightly less accurate but faster colocalization analysis.

**GICA (headless)** This module is intended for scripting purposes only! All parameters are set automatically without interaction. In a Fiji script this module can be executed multiple times to compute 3D stacks of images. As long as single images are used **GICA** or **Gamma-Norm Coloc. Topology** should be the method of choice!

**fast GICA** This module is a fast alternative to the standard **GICA** module. Here the Analysis of a ROI is not repeated in $n$ iterations. Rather, the pixel-data is split into $n$ sections. This allows for a significantly faster analysis while providing a less accurate but still reasonable Gamma-Norm.

**fast Coloc. Topology** This module generates a Gamma-Norm Topology using the fast sectioning algorithm described above.

**Preprocessing:**
GICA processes only RGB images for now. Therefore, convert your images to RGB in Fiji, using **Image>Color>Stack to RGB** or the **Convert to RGB** option of the **Channels Tool**... to be found in the same menu.