

NutriChip Image Processor: A novel way of extracting fluorescent spots in microscopy images

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Objectives

Extracting fluorescent spots [1] in microscopy images. The resulting mask provides access to measurements (intensity, segmented blob sizes, ...) that can be linked with a biological event [2] (e.g. TLR2 expression).

We introduce a novel segmentation algorithm fit for the purpose.

- Tested on images from **stimulated (SG group)** and **unstimulated (NCG group)** Caco-2 cells.
- Compared against state-of-the-art methods on the problem of (a) **classification** and (b) **localization**.

Scope

The NutriChip project proposes to study the impact of dairy products ingestion by human through the use of a Lab-on-Chip platform.

Fluorescently stained biomarkers such as the toll-like receptors 2 and 4 (TLR2-4) are used to get a measurement of the cell immune response.

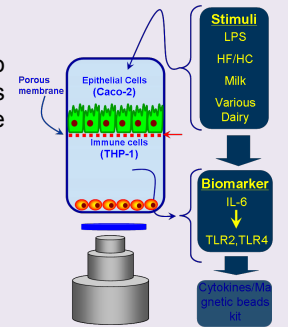


Image courtesy of Qasem Alramadan, EPFL

Novel algorithm

Local thresholding algorithm:

- Sweeps thresholding values.
- Extracts blobs of pixels of limited size with a local maxima (fluorescent spot).

- Filters out pixels with low local SNR.

$$LSNR = \frac{I \cdot B}{I - I \cdot B}$$

Segmentation mask examples

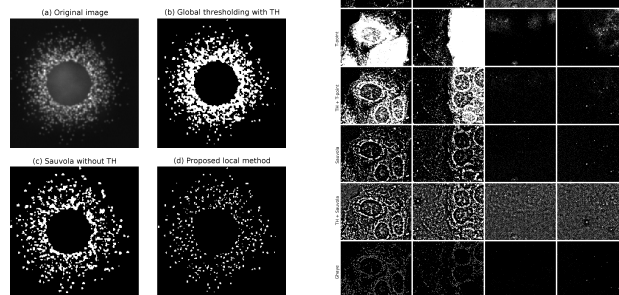
Comparison against state-of-the-art methods.

Pre-filtering:

- Top-Hat (TH)

Thresholding:

- **Global:** Otsu, T-point
- **Local:** Sauvola



On synthetic SG images

On real SG and NCG images

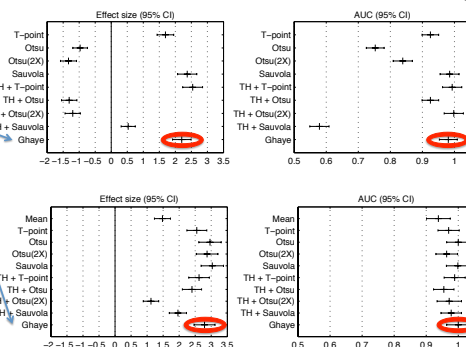
Comparison results

For classification (SG vs. NCG)

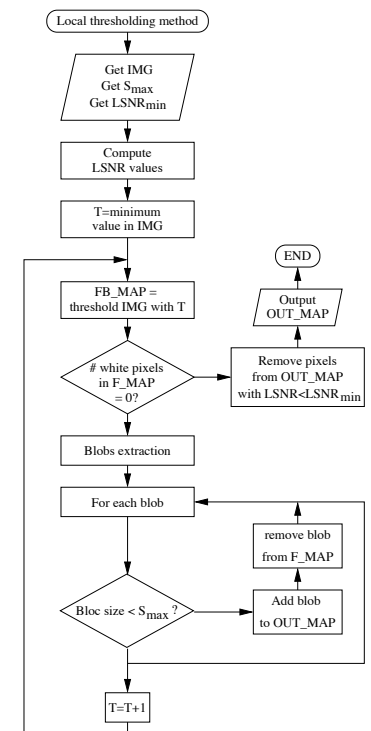
- Number of segmented pixels
- Intensity of the segmented pixels
- **Recommended methods:** TH+T-point, Sauvola, Ghaye.

For localization:

- ... while recovering ~60% of the spots
- Ghaye extracts the greater amount of blobs (~375 blobs).
- Limited blob sizes (13 to 25 pixels).



Algorithm flowchart



Conclusion

- ✓ Novel method for spot extraction.
- ✓ Provides useful masks for image classification.
- ✓ Recovers more blobs than any other methods when the blob size is limited.

References

1. J. Ghaye, M.A. Kamat, L. Corbino-Giunta, P. Silacci, G. Vergères, G. De Micheli, S. Carrara, "Image Thresholding Techniques for Localization of Sub-Resolution Fluorescent Biomarkers", *submitted to Cytometry Part A*.
2. J. Ghaye, G. De Micheli, S. Carrara, "Quantification of Sub-resolution Sized Targets in Cell Fluorescence Imaging", *BioCAS 2012 IEEE*, pp. 268-271.