Application of Rényi entropy-based 3D electromagnetic centroids to segmentation of fluorescing objects in tissue sections

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1. Theoretical assumptions

- Extended Nijboer-Zernike Theory
- Multifractality

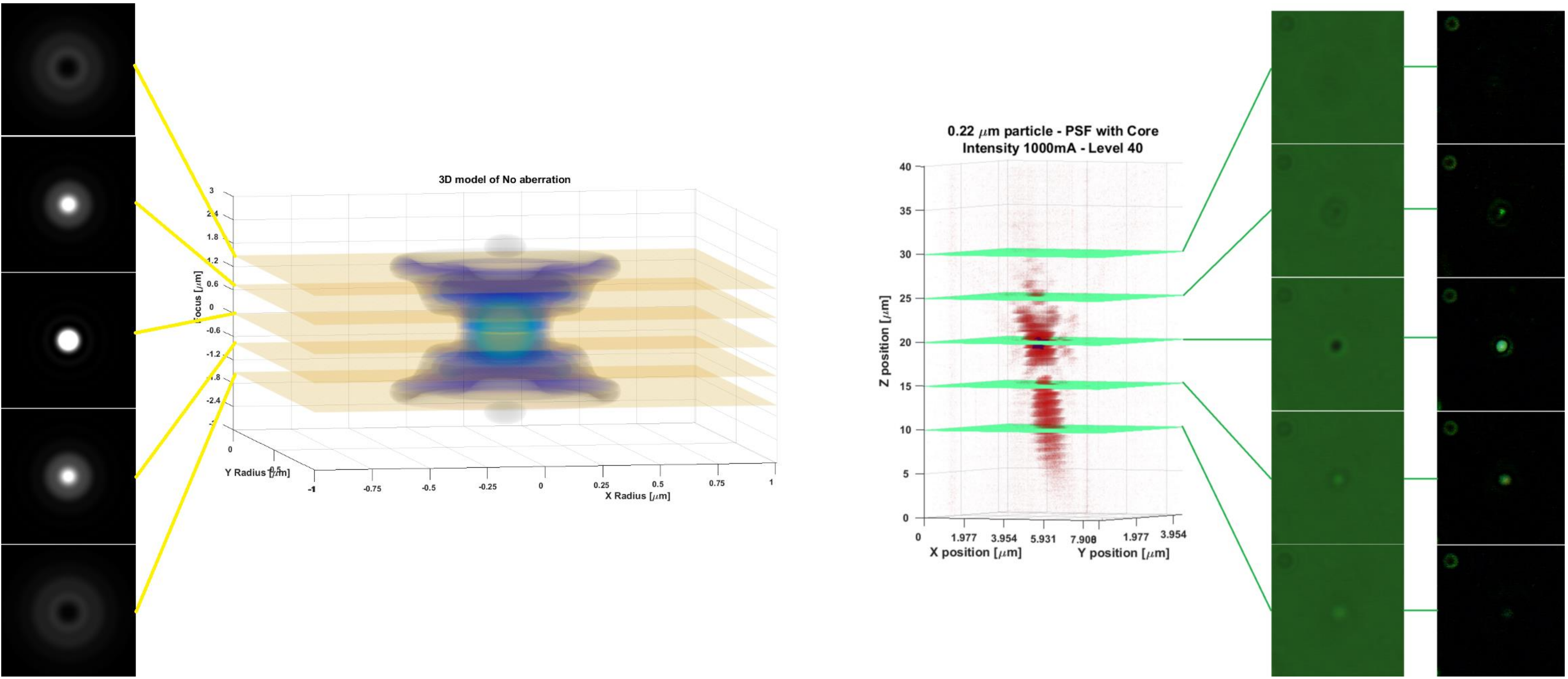
+

2. Technical solutions

- Small camera pixel
- Primary vice-bit camera signal
- Short z-step
- Strong light illumination

Rychtáriková et al., Ultramicroscopy, 2017.

Extended Nijboer-Zernike Theory



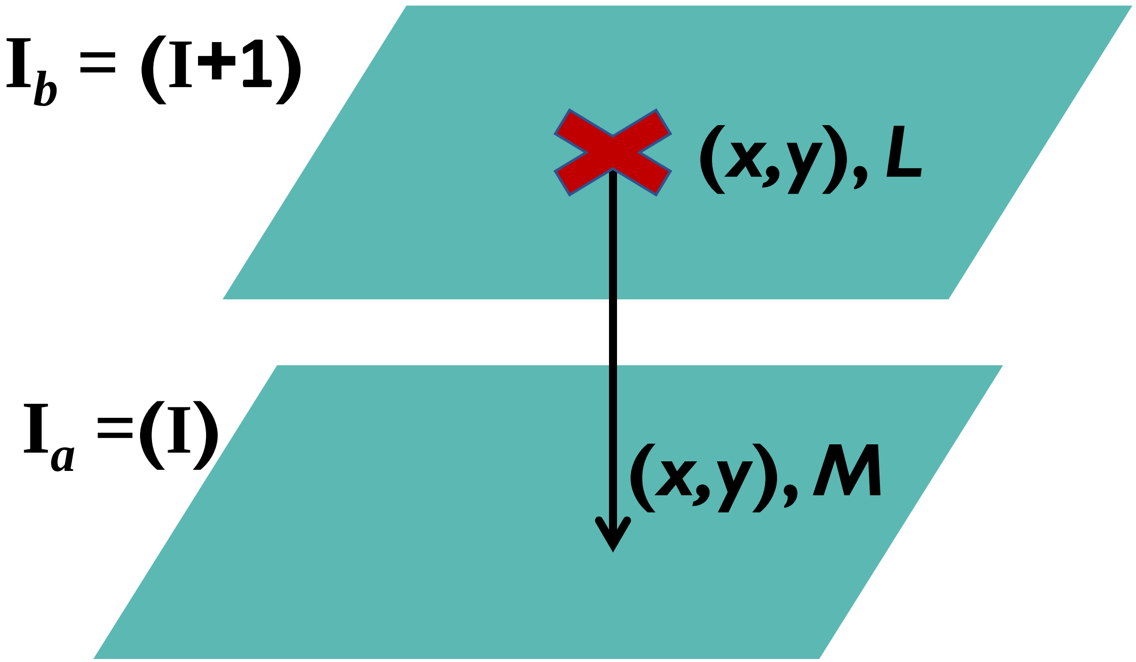
+ Theory of Electromagnetic Centroid

Volume electromagnetic centroid:

- intensity extreme
- the same intensity in two consecutive images

Rychtáriková et al., Ultramicroscopy, 2017.

Multifractality approach: Point Divergence Gain $\Omega_{\alpha}^{(L \to M)}$



Two consecutive z-stack images

Rychtáriková et al., Entropy, 2018.

Multifractality approach: Point Divergence Gain $\Omega_\alpha^{(L \to M)}$

Difference of two Rényi entropies:

$$\Omega_\alpha^{(L\to M)} = \boxed{\frac{1}{1-\alpha}\log_2 \sum_{i=1}^{j}\left(p_i^{(L\to M)}\right)^\alpha} - \boxed{\frac{1}{1-\alpha}\log_2 \sum_{i=1}^{j}(p_i)^\alpha}$$

$$\vdots$$

$$\Omega_\alpha^{(L\to M)} = \frac{1}{1-\alpha}\log_2\left[\frac{(n_L-1)^\alpha - n_L^\alpha + (n_M+1)^\alpha - n_M^\alpha}{C_\alpha} + 1\right]$$

Specific case $\alpha = 2$ (the Rényi collision entropy)

Taylor s.

$$\Omega_2^{(L\to M)} = \frac{1}{1-\alpha}\log_2\left[\frac{2}{C_2}(n_M - n_L + 1) + 1\right] \approx \mathrm{A}(n_M - n_L) + \mathrm{B}$$

i – value of intensity
M – pixel intensity in the first image (I)
L – pixel intensity in the following image (I+1)
j – number of intensities occupied in the image
p_i – probability of the occurrence of intensity i in the image

n_i – number of the occurrence of intensity i in the image
α – the Rényi dimensionless coefficient ($\alpha \geq 0, \alpha \neq 1$)
$C_\alpha = \sum_{i=1}^{j} n_i^\alpha$ – constant for intensity distribution of image (I)

Rychtáriková et al., Entropy, 2018.

Multifractality approach

Point Divergence Gain Entropy I_α:

$$I_\alpha(\mathbf{I}_a;\mathbf{I}_b) = \sum_{i=1}^{n} \left|\Omega_\alpha^{a_i \to b_i}\right| = \sum_{L=1}^{j}\sum_{M=1}^{j} n_{lm}\left|\Omega_\alpha^{L\to M}\right|$$

Point Divergence Gain Entropy Density P_α:

$$P_\alpha(\mathbf{I}_a;\mathbf{I}_b) = \sum_{L=1}^{j}\sum_{M=1}^{j} X_{lm}\left|\Omega_\alpha^{L\to M}\right|$$

$$X_{lm} = 1,\ n_{lm} \geq 1$$
$$X_{lm} = 0,\ n_{lm} = 0$$

$\mathbf{I}_a = \{a_1, \dots, a_n\}$ and $\mathbf{I}_b = \{b, \dots, b_n\}$ – two consecutive one-dimensional data frames with pixel indices a_i and b_i, respectively.
n_{lm} – number of substitutions $l \to m$ at transformation $\mathbf{I}_a \to \mathbf{I}_b$

Rychtáriková et al., Entropy, 2018.

$\Omega_{\alpha}^{(L \to M)}$ in microscopy image processing

1. 3D segmentation

- Finding „volume electromagnetic centroid“
- Movements detection

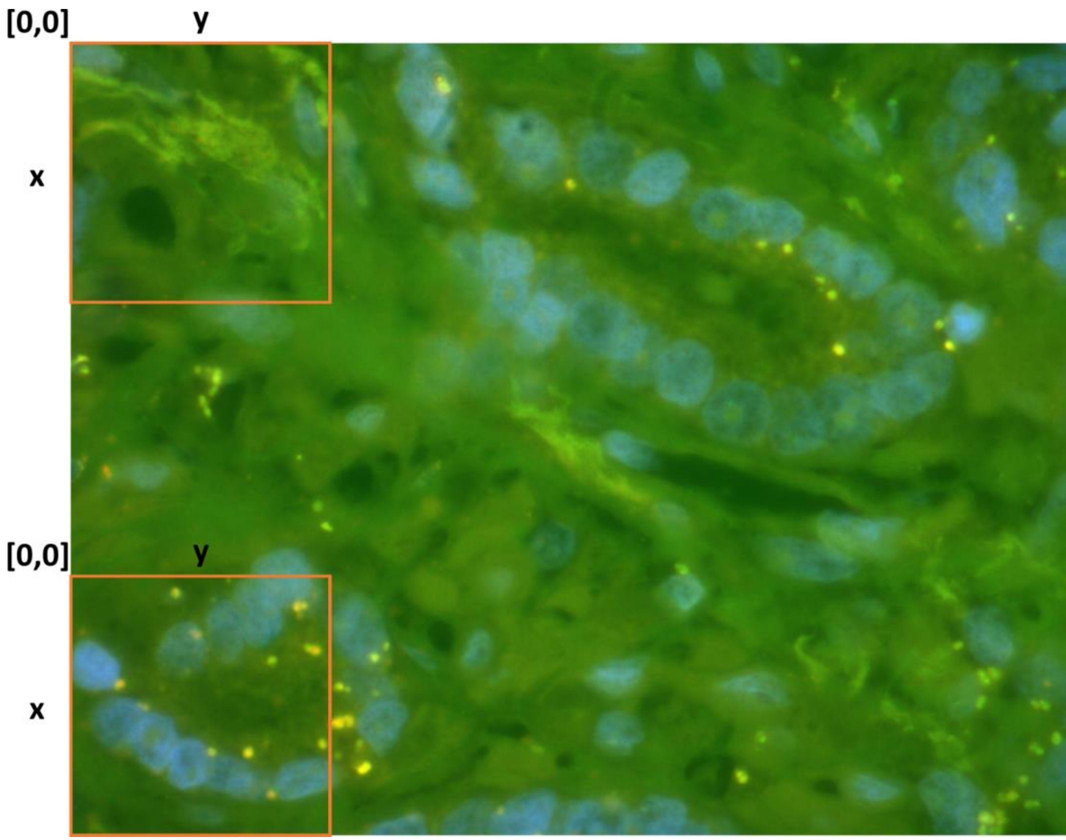
2. Image classification

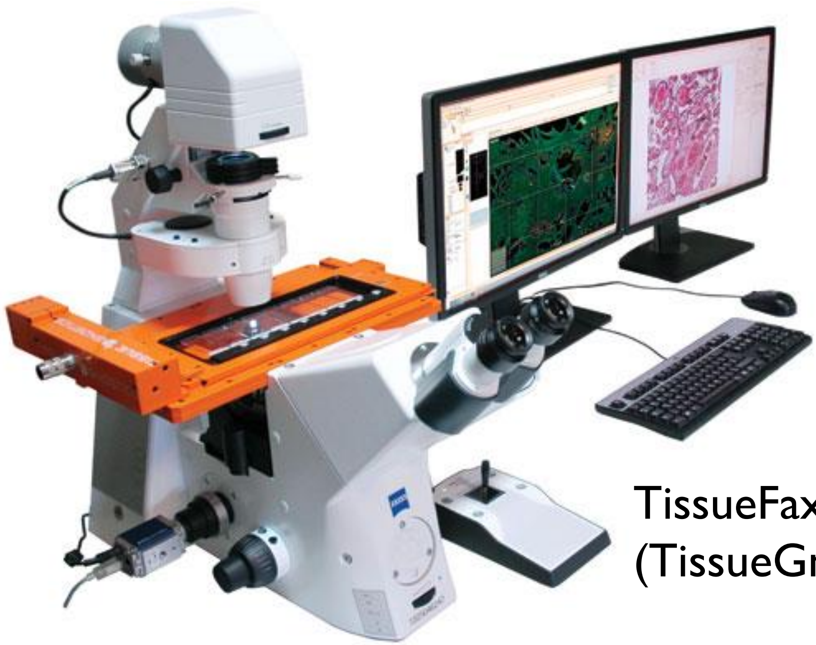
- Finding in-focus region/image

Application in fluorescence microscopy

Prostate cancer tissue section

- DAPI, red, and green autofluorescence
- Pixel size 328×328 nm^2
- Wide-field mode

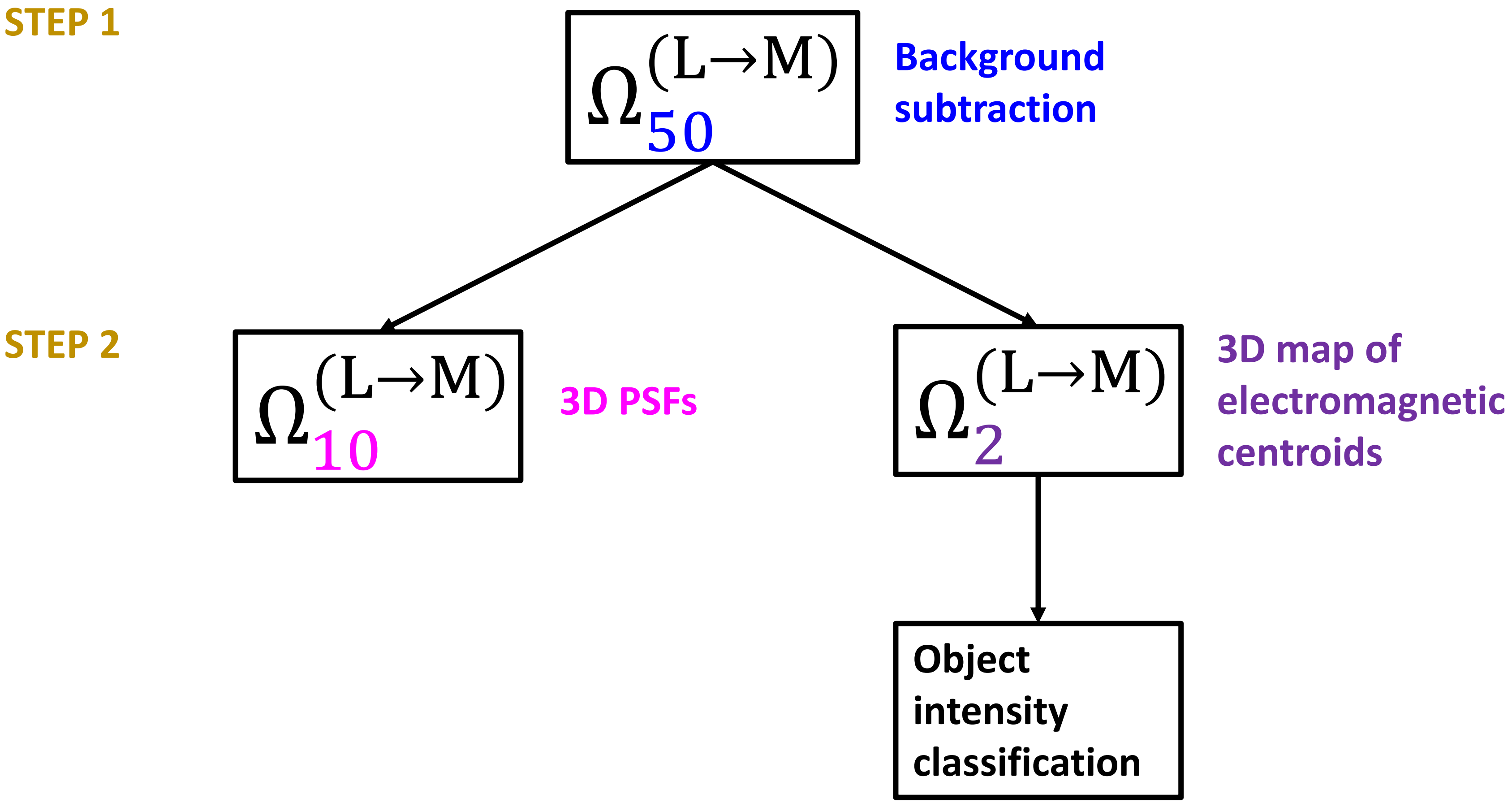


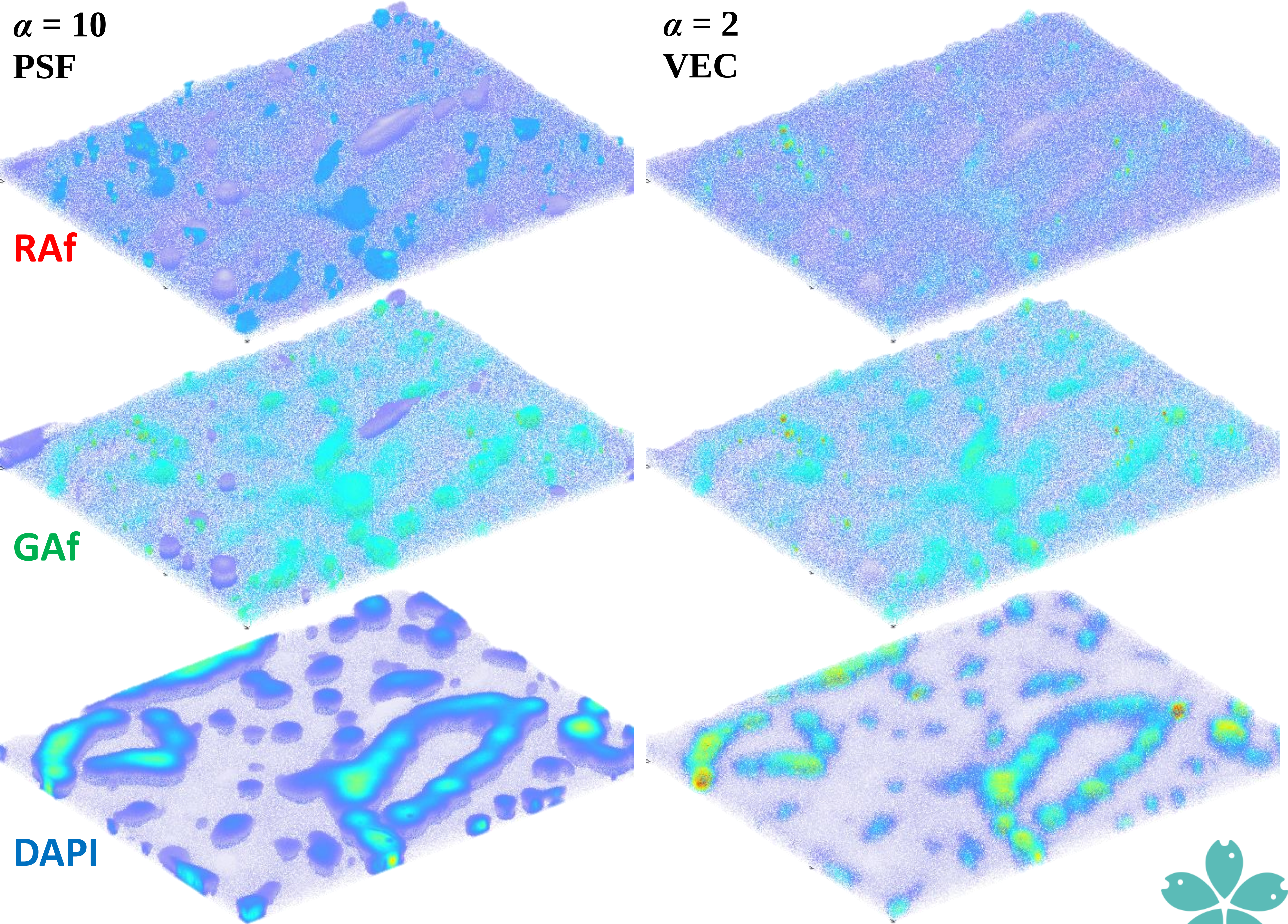


TissueFaxs-PLUS-Confocal fluorescence microscope (TissueGnostics, Vienna, AT)

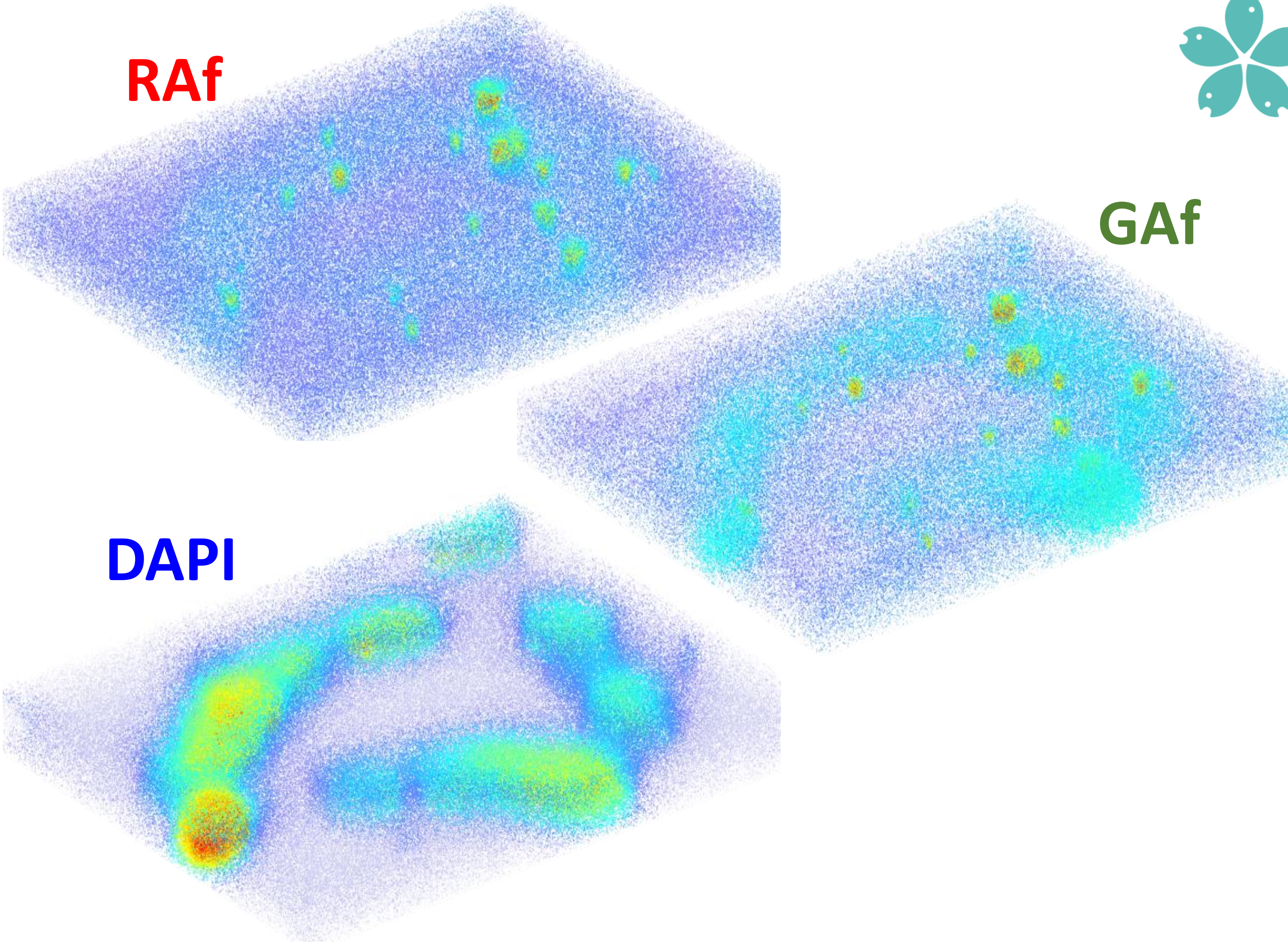
Rychtáriková et al., Arxiv 1709.03894, 2017.

Outline of calculation



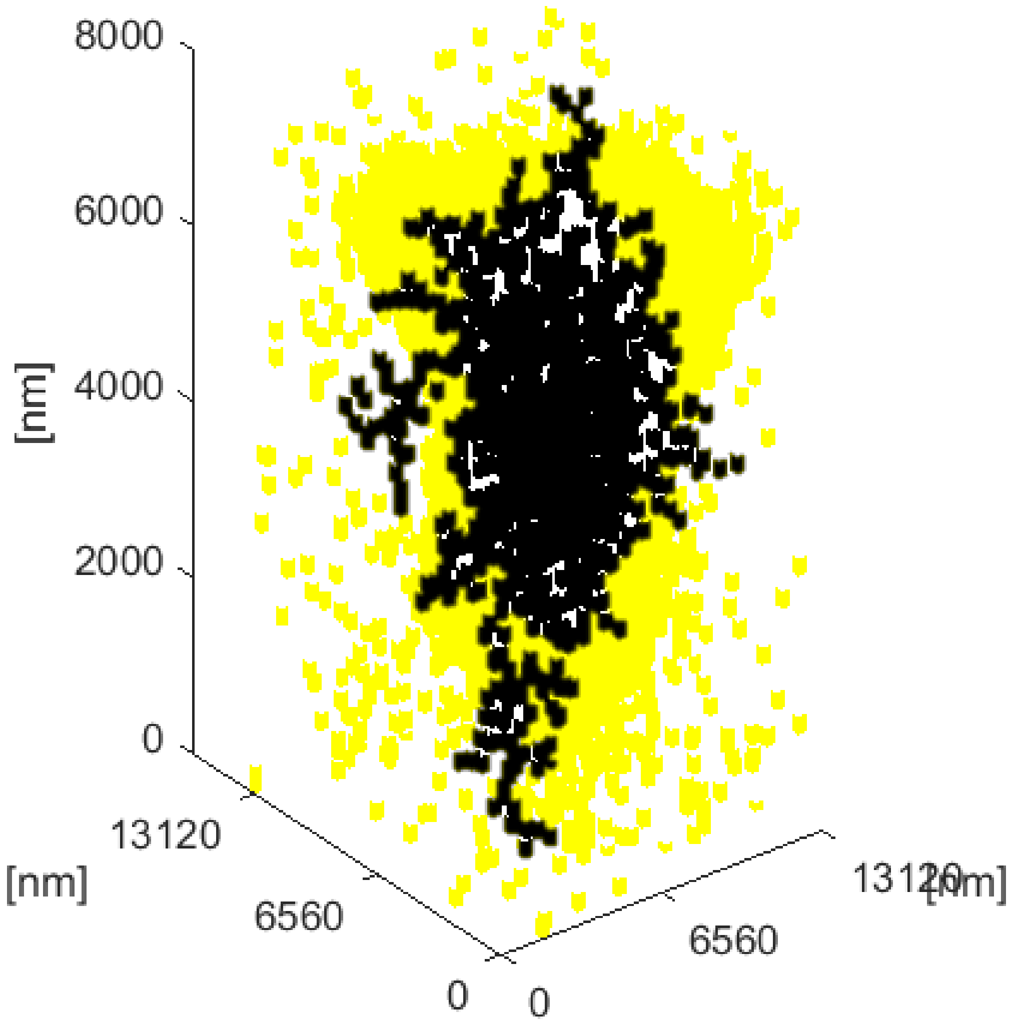


α = 10
PSF
α = 2
VEC
RAf
GAf
DAPI


RAf
GAf
DAPI

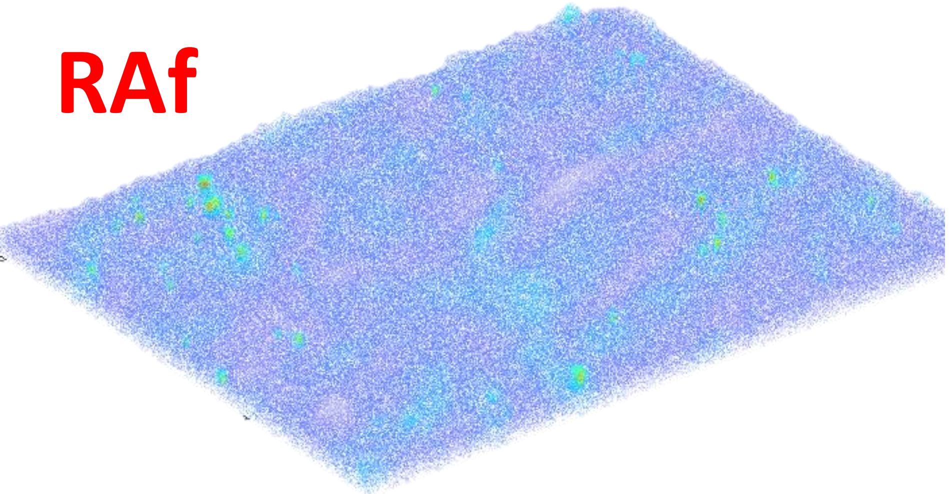
PSFs

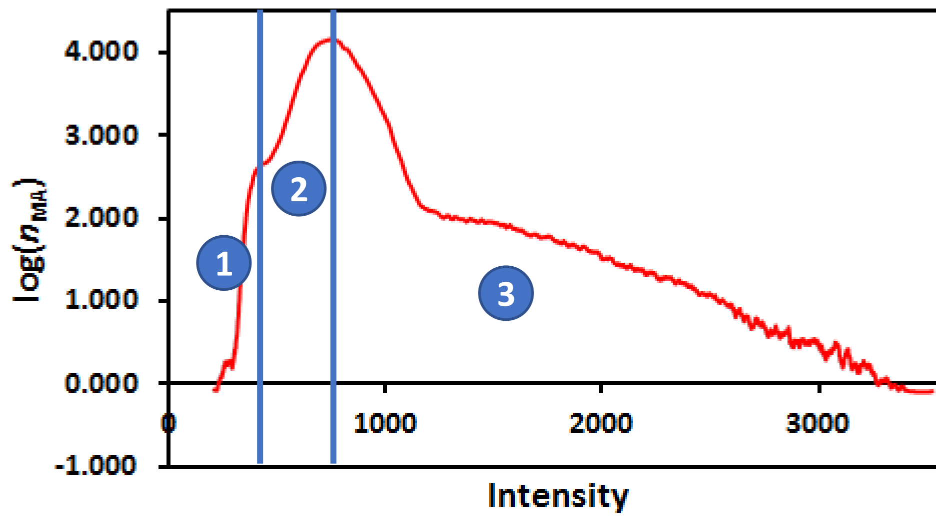
RAf



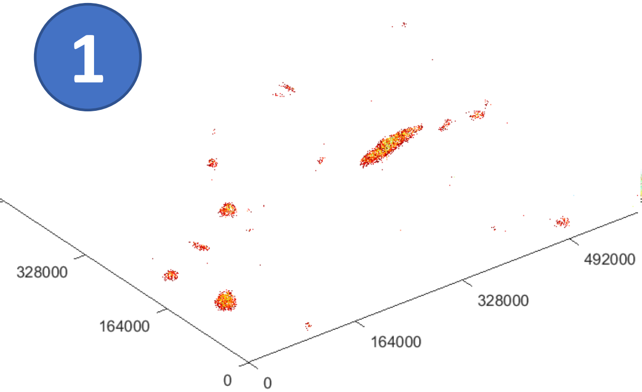
Intensity classification

RAf





1



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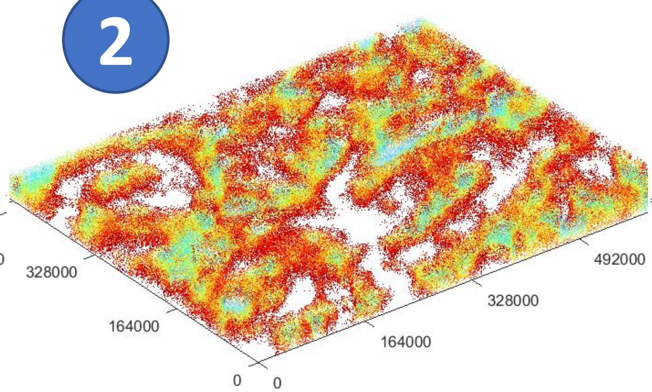

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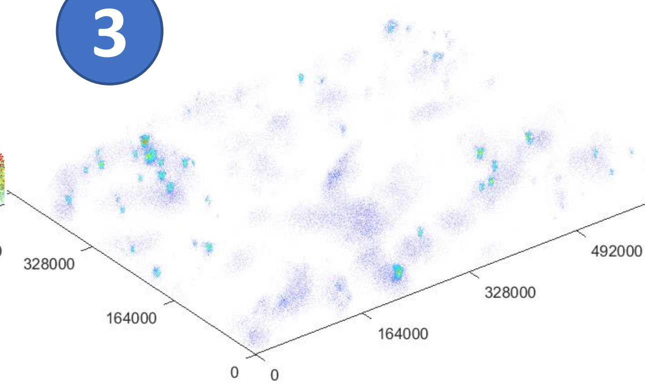

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