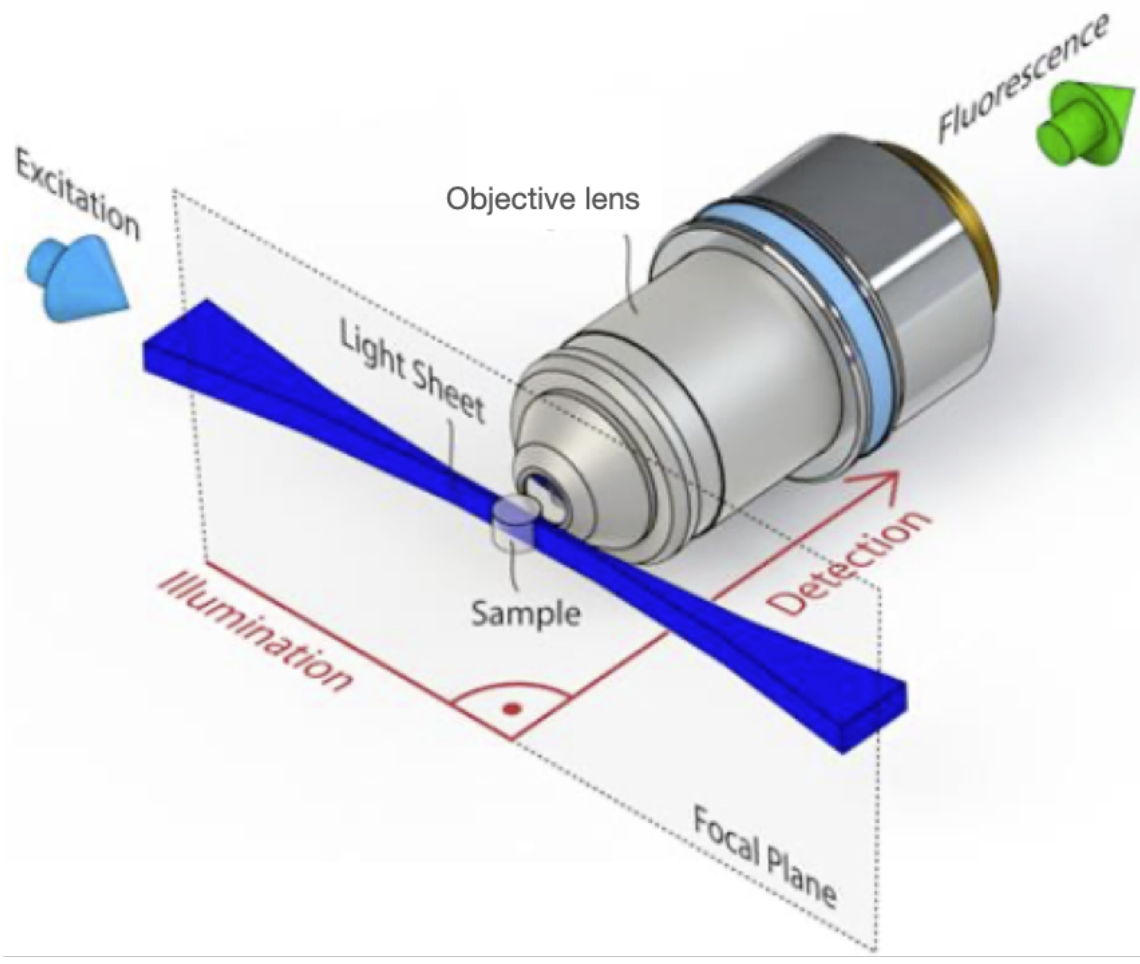


LIGHTSHEET MICROSCOPY

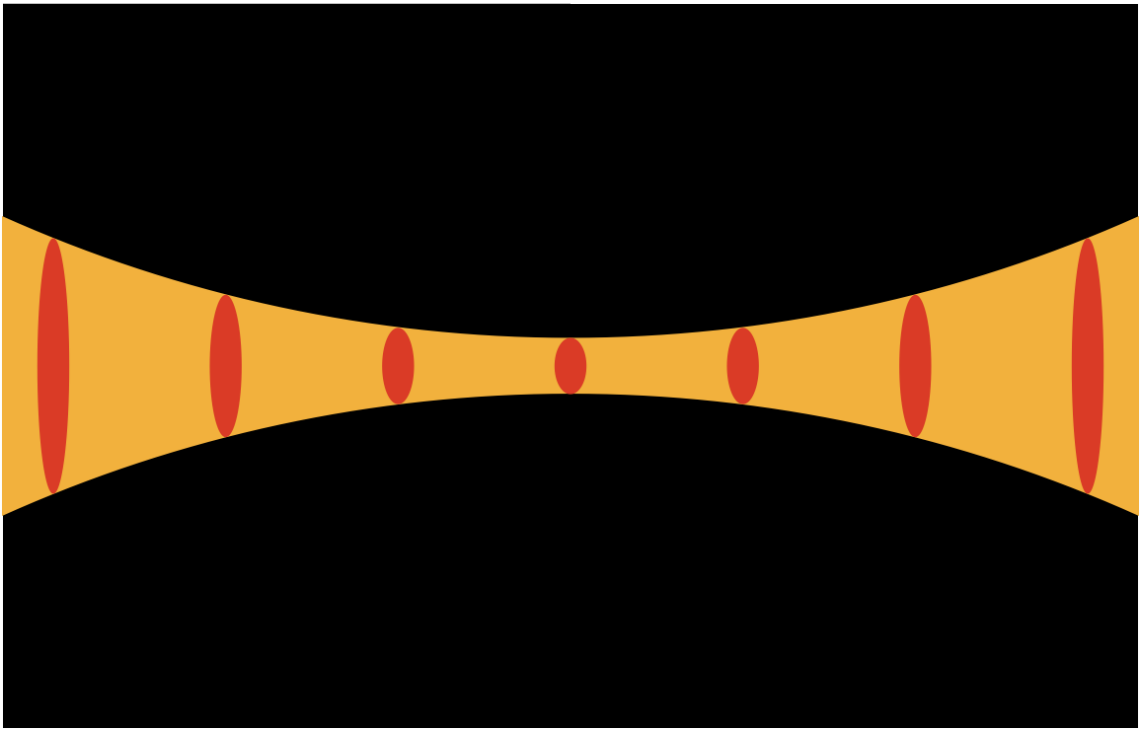
TODO: a few words about why lightsheet is good

Light sheet microscopy is a type of fluorescence microscopy used in cell biology due to its fast acquisition times and low photo-damage to the sample. This is achieved by selectively illuminating a slice of the sample using a sheet of light and detecting the emitted fluorescence signal using a dedicated objective orthogonal to the plane of the sheet.

- Project aim: computationally improve the quality of light sheet microscopy images.



SPATIALLY VARYING PSF



PROBLEM

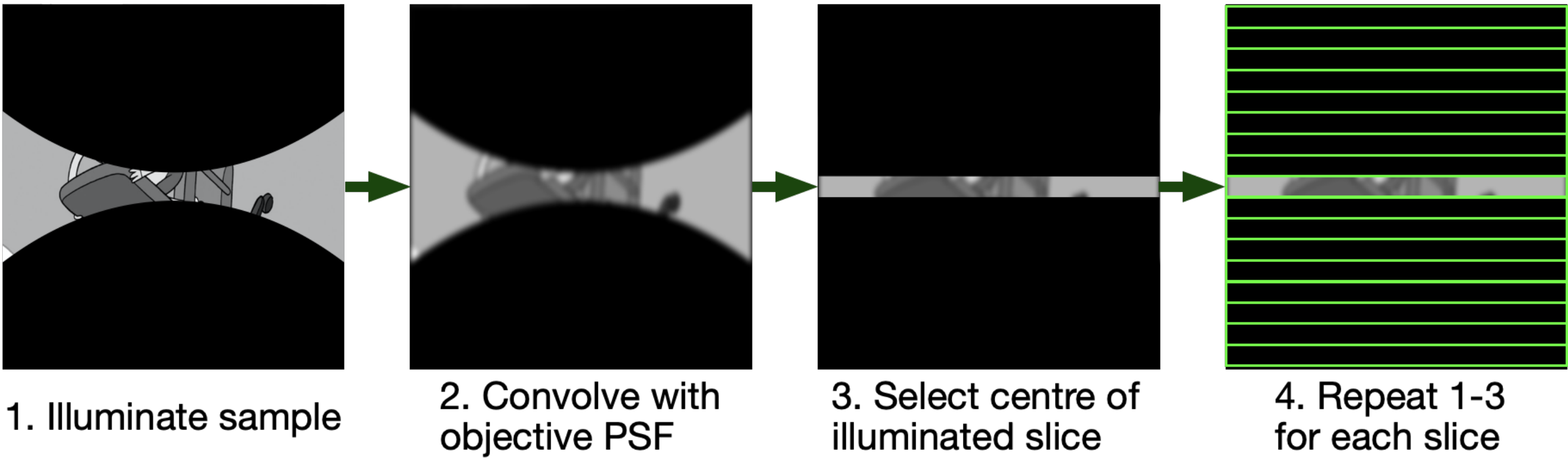
Due to the combination of light-sheet beam (yellow) and the objective PSF (red), the effective PSF of the system is spatially varying, and therefore standard deconvolution approaches are not applicable.

Figure credit: Jörg Ritter, PhD thesis (2011)

A light-sheet microscope consists of two ob-  
jectives: TODO etc

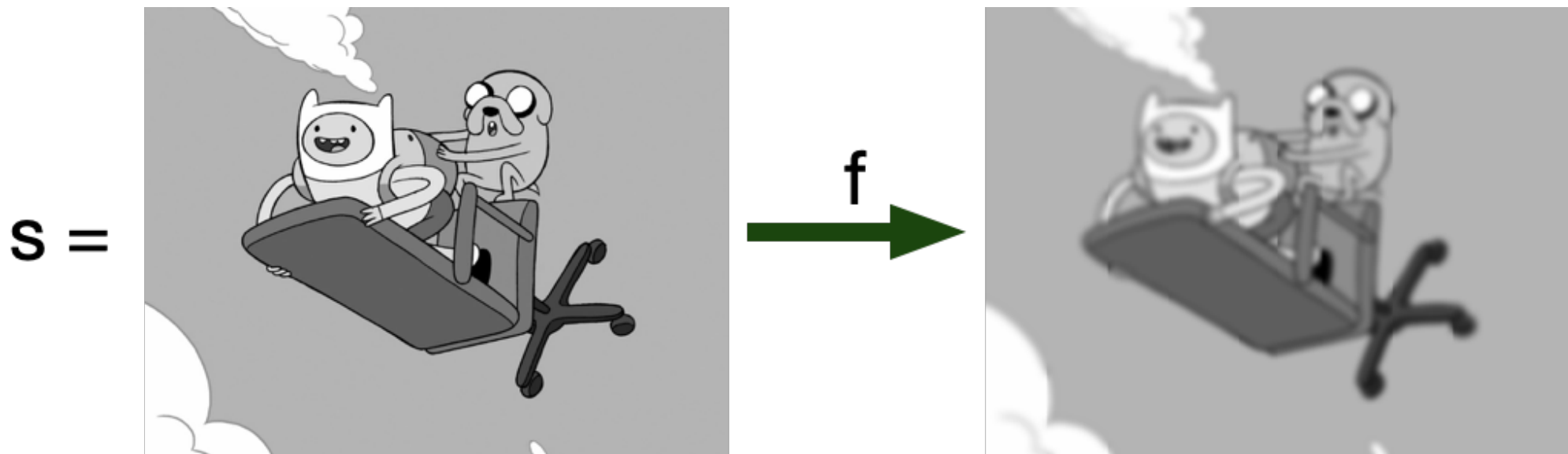
IMAGE FORMATION MODEL

The sample  $s$  illuminated at  $z = z_0$  by the light-sheet  $l$  and the photons are collected by an objective with PSF  $h$ :



$$f(x, y, z_0) = \iiint l_{avg_y}(u, v, w) s(u, v, w - z_0) h(x - u, y - v, w) du dv dw$$

where  $h$  is the objective PSF, calculated using (1) and  $l_{avg_y}$  is the light-sheet, calculated by averaging the beam PSF obtained in a similar way to  $h$ , without Zernike polynomials.



PSF MODEL

TODO We calculate the objective PSF  $h$  by using a model that includes defocus etc etc.  
TODO: reference to paper

$$h(x, y, z) = \left| \iint g_\sigma * p(\kappa_x, \kappa_y) e^{2i\pi z \sqrt{(n/\lambda)^2 - \kappa_x^2 - \kappa_y^2}} e^{2i\pi(\kappa_x x + \kappa_y y)} d\kappa_x d\kappa_y \right|^2 \quad (1)$$

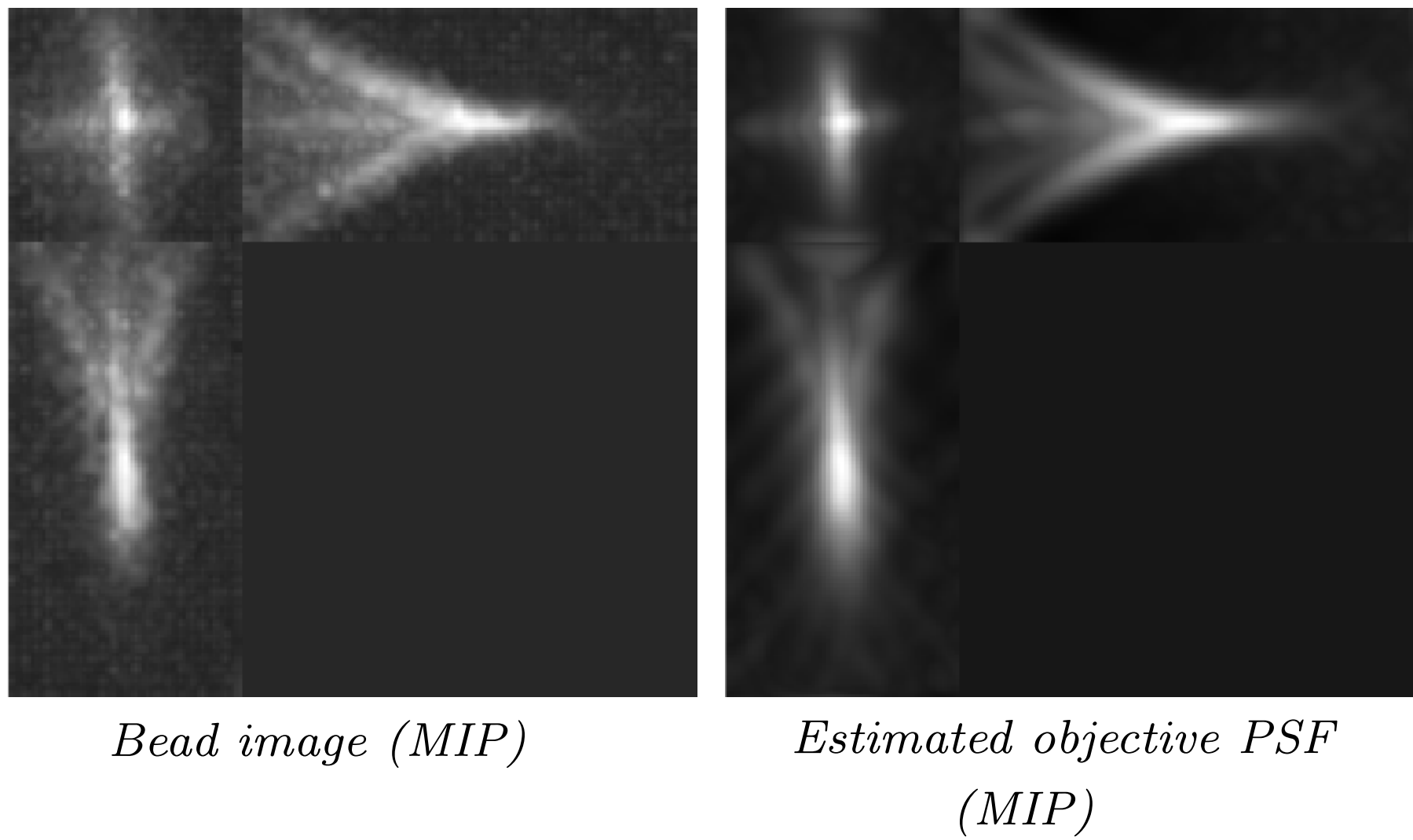
with the pupil function  $p$  defined

$$p(\kappa_x, \kappa_y) = \begin{cases} e^{2i\pi \sum_{j=1}^{15} c_j Z_j(\kappa_x, \kappa_y)} & \text{for } \rho = \sqrt{\kappa_x^2 + \kappa_y^2} \leq NA/\lambda, \\ 0, & \text{otherwise.} \end{cases} \quad (2)$$

where the phase of the pupil function is computed by least-squares fitting of the coefficients of the first 15 Zernike polynomials using an image of a bead.

The parameters of the model are given by the experimental setup used to acquire the image:

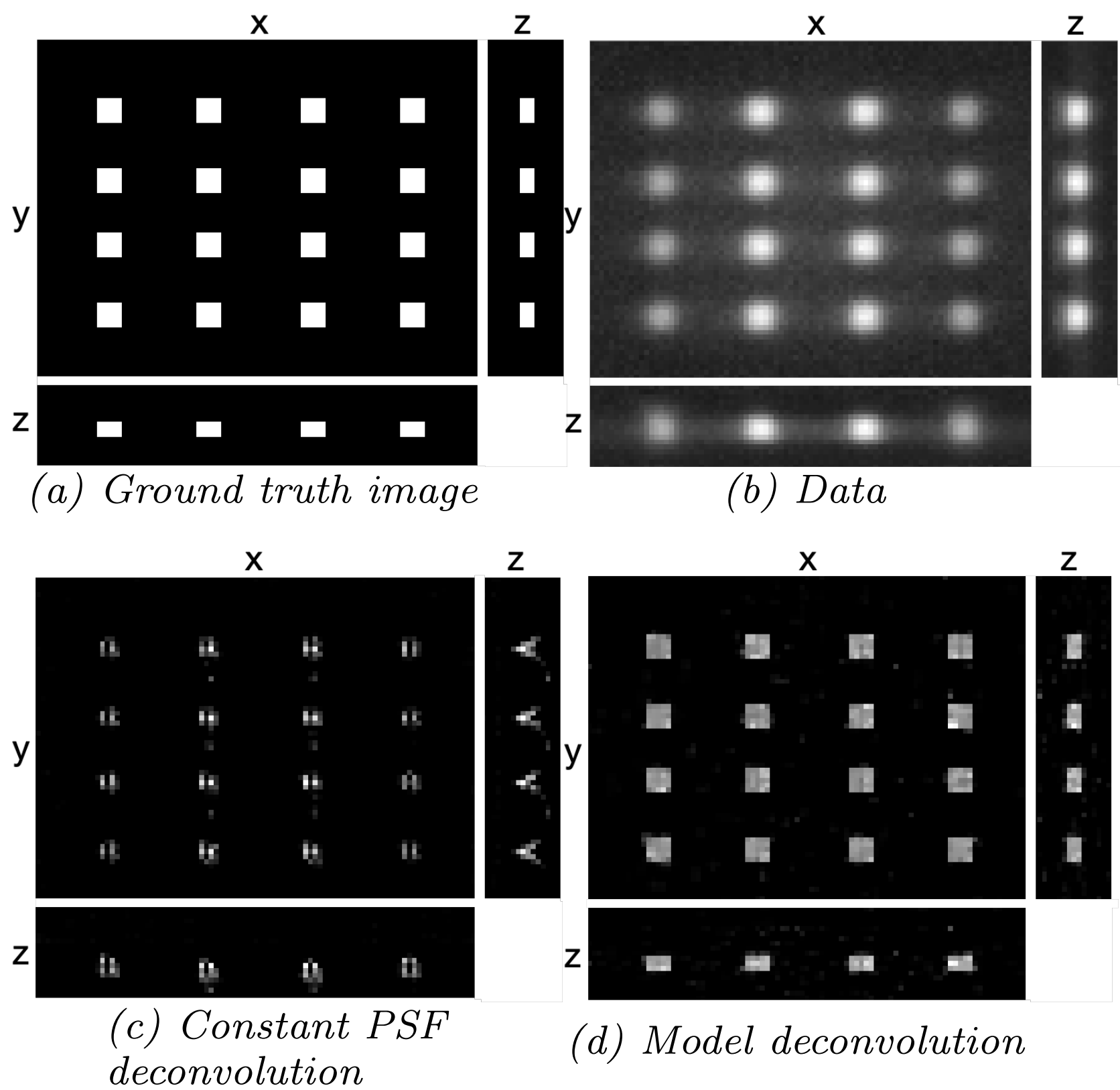
- $n$  - refractive index
- $\lambda$  - wave length
- NA - numerical aperture
- $g_\sigma$  Gaussian blur to take into account other properties not accounted for in our model



RECONSTRUCTION

Let  $\hat{f}$  be the image data and  $f(s)$  the result of applying the forward model to the sample  $s$ . To recover  $s$ , we solve:

$$\text{Find } \hat{s} \in \text{argmin}_s \left\{ \|\hat{f} - f(s)\|_{L_2} + \lambda TV(s) \right\}$$

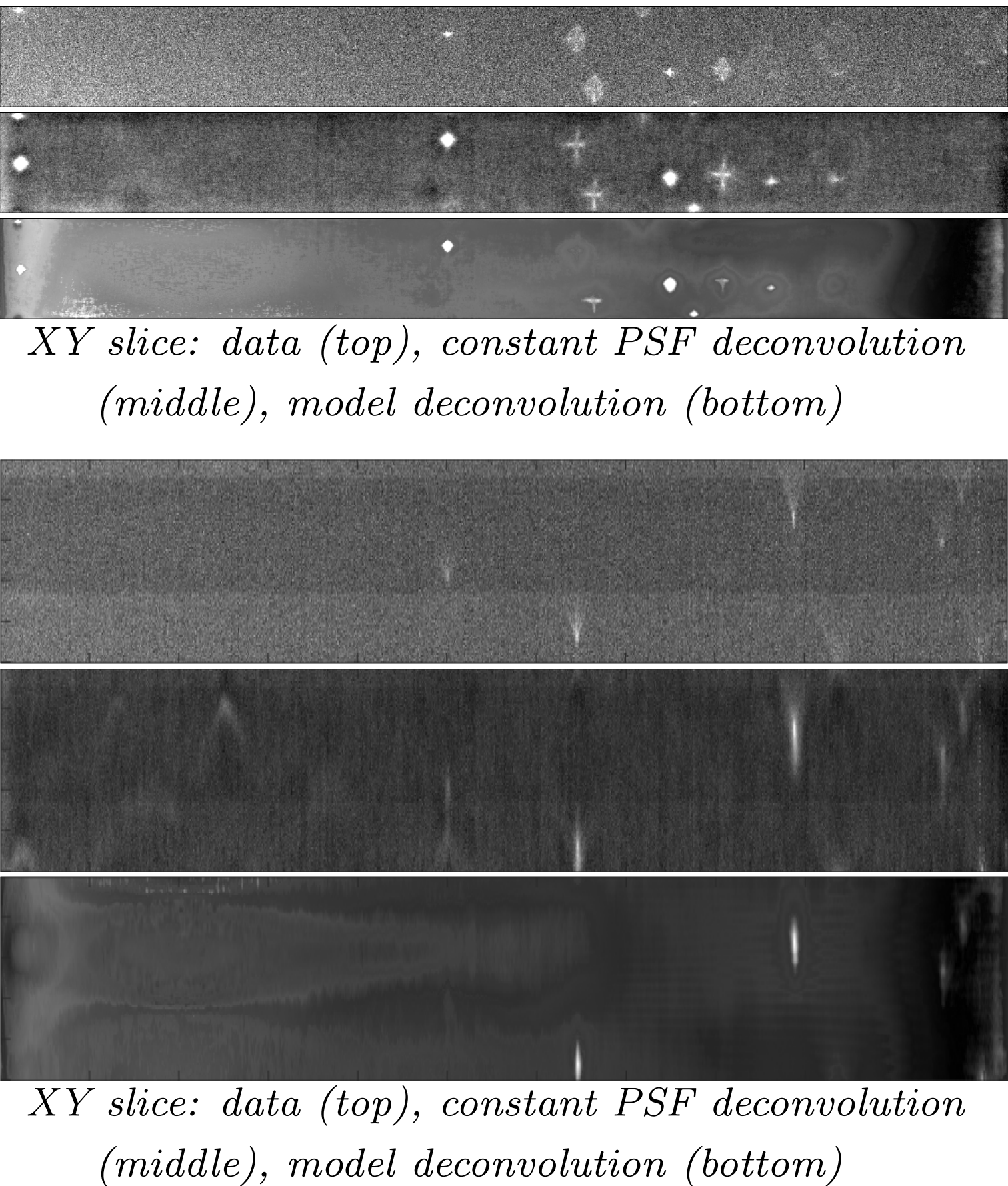


- To solve the optimisation problem, we apply a version of the Primal Dual Hybrid Gradient (PDHG) algorithm TODO:reference
- In the figure on the left, we show an example of a simulated sample (a) to which we apply the forward model with Gaussian noise (b) and the result of deconvolving using only a spatially varying PSF (c) and the full forward model (d). All images are shown using maximum intensity projection.

RESULTS

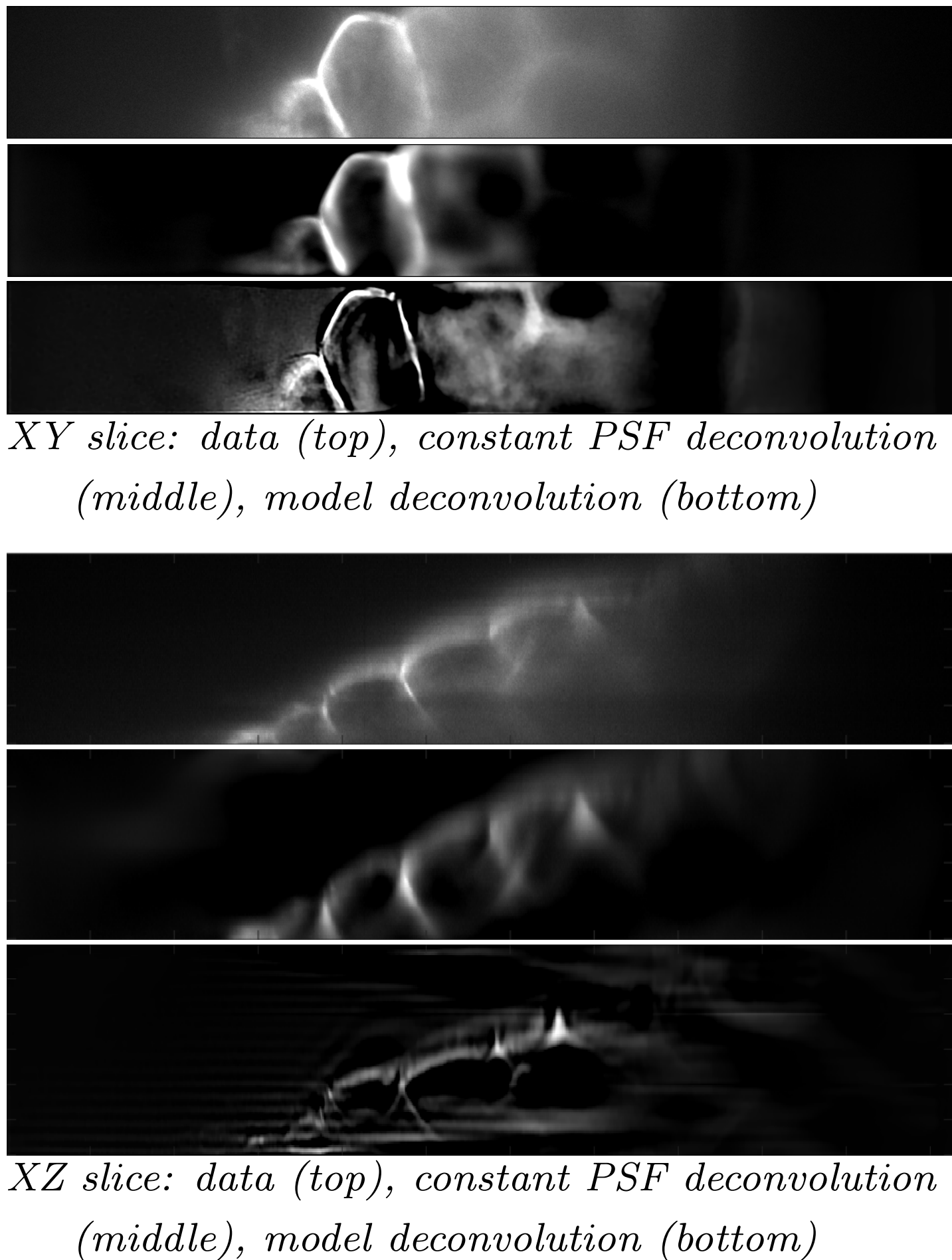
BEADS

- We apply the proposed method to a sample of beads in agarose.
- The dimensions of the sample are 1127 x 111 x 100 pixels.
- 



MARCHANTIA

- We apply the proposed method to a sample of Marchantia plant.
- The dimensions of the sample are 1127 x 155 x 100 pixels.
- 



REFERENCES

ACKNOWLEDGMENTS

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