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# *Quantitative microscopy for characterizing material microstructures and mass transport*

To understand porous material microstructures and their mass transport properties, and ultimately to learn how to tailor materials for specific purposes, quantitative characterization using statistical models and methods is key. In this talk, we give three examples of quantitative methods in microscopy for this purpose.

First, we describe how single particle tracking (SPT) based on confocal laser scanning microscopy data can be used to estimate local diffusion coefficients, sizes, and concentrations of fluorescent nanoparticles in liquid suspension. Estimation is performed using simulation-based inference, specifically Approximate Bayesian Computation (ABC). The method is widely applicable for characterization of natural and man-made nanoparticles, including biological fluids like blood. We also demonstrate good performance on a validation case [1].

Second, we describe a new numerical model based on spectral-domain numerical methods for modeling of data from fluorescence recovery after photobleaching (FRAP) experiments. FRAP is a widely used technique for estimation of both local diffusion coefficients and binding kinetics i.e. association and disassociation rate constants, to understand heterogeneity, obstruction effects, structural dynamics, and interactions with a matrix in soft matter. This new FRAP model is one of the most comprehensive to date, and we show good agreement with previous literature on a validation case [2].

Third, we present a new method for segmentation of image data acquired using focused ion beam scanning electron microscope (FIB-SEM) nanotomography. FIB-SEM is a powerful technique for 3D imaging of nanoporous microstructures, allowing for substantially higher spatial resolution than e.g. X-ray tomography. A 3D data set is acquired in a serial fashion where in-between each image, a slice of the sample is milled away using the ion beam of the FIB to reveal a new planar cross-section, which is imaged using the SEM. The technique is challenging due to e.g. the fact that it produces a stack of 2.5D rather than 2D images, with subsurface pore information shining through. We propose a random forest classifier combined with scale-space feature extraction, yielding good agreement with manual segmentations on three data sets of porous polymer films [3].

*References*

[1] M Röding, M Billeter. Massively parallel approximate Bayesian computation for estimating nanoparticle diffusion coefficients, sizes and concentrations using confocal laser scanning microscopy. *Journal of Microscopy*, 2018, 271, 174–182.

[2] M Röding, L Lacroix, A Krona, T Gebäck, N Lorén. A highly accurate pixel-based FRAP model based on spectral-domain numerical methods. *Biophysical Journal*, 2019, DOI: 10.1016/j.bpj.2019.02.023.

[3] M Röding, C Fager, A Olsson, C von Corswant, E Olsson, N Lorén. Three-dimensional reconstruction of microporous polymer films from FIB-SEM nanotomography data using random forests. Submitted to *Microscopy and Microanalysis*.