ACCELERATED DIFFUSION COEFFICIENT ESTIMATION IN FRAP USING DEEP NEURAL NETWORKS

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Fluorescence recovery after photobleaching (FRAP) is a powerful technique for estimation of diffusion coefficients. Briefly, using a confocal laser scanning microscope (CLSM), fluorescent particles are irreversibly photobleached in a well-defined bleach region. The recovery of fluorescence within the bleach region is modeled either as (i) a temporal evolution of concentration using the recovery curve i.e. the average intensity in the bleach region as a function of time, or (ii) as a spatio-temporal evolution using the full set of pixels in the microscopy images. In both cases, estimation of the diffusion coefficient and other parameters is typically performed using least squares, and sometimes maximum likelihood [1].

In previous work [2], we introduced a new and highly accurate numerical FRAP model based on spectral methods, for both recovery curve-based and pixel-based estimation. One downside of numerical models for FRAP is computational time, in particular when using multiple initial parameter guesses during fitting and when analyzing multiple data sets. In this work, for the first time, we utilize machine learning and specifically deep neural networks for very fast estimation of diffusion coefficients in FRAP. We use realistic simulated data generated from the model in [2] for training the networks. We present network architectures for both recovery curve-based and pixel-based estimation, based on combinations of convolutional layers and fully connected layers.

We validate that the neural networks perform well on a wide range of diffusion coefficients using simulated data, although they perform worse than least squares fitting which constitutes the benchmark. The networks are quite robust against a wide range of noise levels, to some extent due to the smoothing nature of convolutional layers. Further, estimation is orders of magnitude faster compared to least squares. Finally, we also confirm for two sodium fluorescein-sucrose-water solutions with different viscosities that the results on experimental data are in good agreement with conventional methods. This novel approach for FRAP analysis is useful for fast batch processing of many data sets and can also be used for providing good initial parameter guesses for least squares fitting, hence speeding up conventional FRAP analysis as well.

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