BAYESIAN STATISTICAL PROCESS OF LOCALISATION MICROSCOPY IMAGES

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Localisation Microscopy (LM) and other super-resolution techniques[1] are increasingly being used to study biological processes at resolutions far greater than diffraction limited techniques. However, to decrease acquisition time, samples with higher densities of fluorophores are being imaged, causing point spread functions to overlap making it difficult to characterise the number of molecules present.

A two-stage algorithm which uses Bayesian Statistics[2] and Markov random fields[3] has been developed with the aim of overcoming this problem. The algorithm yields probabilistic binary classifications (signal or noise) of the pixels which can be used to de-noise the individual frames, as well as distributions of the two classifications. The first model treats all pixels as stochastically independent and the output of this is used to seed the second model which refines the classification using a spatial distribution.

The output of this algorithm has several different uses; to calculate the Signal-to-Noise ratio, to reduce the amount of memory needed by (on average) 94% and to count the number of molecules in a fixed area of the sample.

By removing the background noise, and calculating a distribution for the signal, the number of fluorophores within a sample can be calculated. The difference between consecutive frames and the output from the algorithm is used to determine the number of fluorophores within the sample.

References:

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