Motion-blurring to calculate molecular dynamics from single molecule localisation data

Sohaib Abdul Rehman*[a], Leila Muresan[a], Martin O. Lenz[a], Maria Gomez-Lamarca[b], Steven F. Lee[c], Sarah Bray[b] and Kevin O'Holleran[a]
[a]Cambridge Advanced Imaging Centre, Anatomy Building, University of Cambridge,
[b]Department of Physiology, Development and Neuroscience, University of Cambridge
[c]Department of Chemistry, University of Cambridge
***E-mail: sa753@cam.ac.uk**

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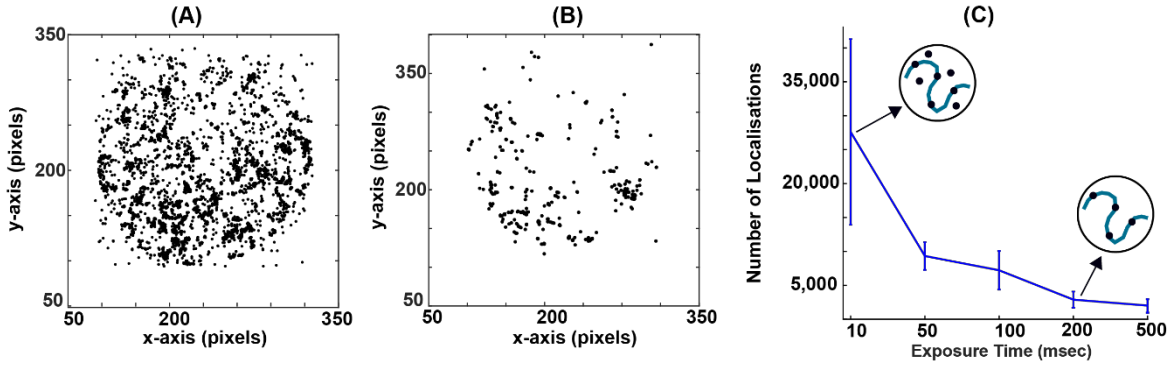
Abstract:

In localisation microscopy (LM), the diffraction limit of light is overcome by sparse activation of fluorophores in the field of view, which makes it an attractive tool to study molecular dynamics. Motion blurring, due to diffusion of protein molecules, have been used to selectively image different populations of proteins [1]. The technique works by counting the number of protein molecules localised over different exposure times. As fast-moving molecules are blurred over long exposure times, presence of fast and slow moving populations and their relative proportions can be estimated from the technique.

In this study, we carry out simulations to quantify the performance of the motion-blurring technique in the presence of multiple diffusion populations. We also apply the technique to study dynamics of a DNA-binding transcription factor (TF) for Notch pathway in Drosophila salivary glands. We show that the method can be used to detect the presence of freely diffusing and DNA-bound TF molecules and calculate their relative proportions [2]. The results signify the effectiveness of the technique to extract molecular dynamics from single molecule localisation microscopy data.



(A) and (B) show localised molecules over an exposure time of 10 msec and 50 msec respectively. (C) shows the decrease in the number of localised molecules with an increase in the exposure time.

References

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