

Particle Tracking in Multiple Confocal Volumes
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ABSTRACT

Nanoscale biomolecules are subject to Brownian motion. Hence, particle motion tracking is an important tool to characterise and localize these structures in microscopy. Here we show that fluorescent beads can be tracked in multiple confocal volumes simultaneously using a spatial light modulator, improving the accuracy of particle characterisation and quantification of diffusion.

A digital micromirror device (DMD), a type of spatial light modulator, has enabled the projection of a 720 spot illumination pattern, allowing parallel measurements of the diffusion coefficient of fluorescent beads to be taken ¹. Measurements have been taken on different viscosities, spot sizes and shapes, spot spacings and bead sizes. The diffusion coefficient is measured by locating the coordinates of the beads in each confocal volume using the Imagej plugin 'Trackmate' ². Currently, the process of machine learning is being used to adapt spot sizes to optimize the system.

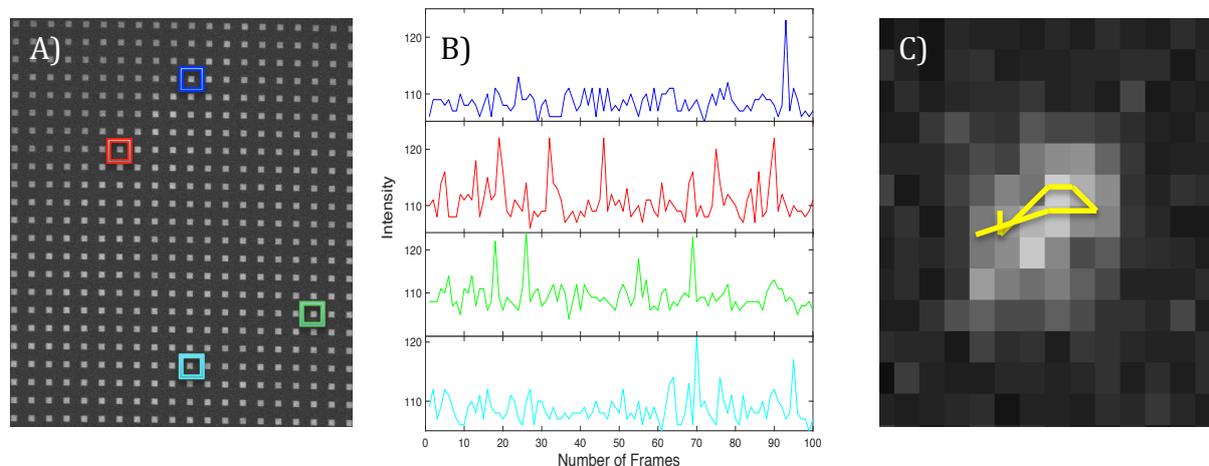


Figure 1: A) The pattern of the DMD showing the 10 pixels x 10 pixels in focus (white) spots and out of focus (dark background) planes with a spacing of 20 pixels between the spots. B) A graph of intensity of each of the four spots (inside the blue, red, green and cyan boxes in Figure 1A) against the number of frames taken. C) Track (yellow) of a fluorescent bead in the focused plane.

References

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