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Determining the orientation of a radiating dipole through fluorescence microscopy

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Fluorescence microscopy is an imaging technique capable of resolving very tiny objects which are not within the resolution range of the normal eye. This technique uses the fluorescence properties of the object in order to get direct knowledge of the particle object and indirect knowledge of the substance in which the object is embedded [1]. In our work, a fluorophore is used as a probe object within a thin polymer layer. The emission from the fluorophore is imaged in a 4f-type imaging geometry onto a sensitive sCMOS camera. In order to determine the orientation, a particular phase pattern is loaded onto a spatial light modulator placed at the Fourier plane of the 4f imaging geometry [2]. A given phase pattern enables us to shape the initial electric field into a different one at the image plane. Information about the radiating dipole is then deduced from the intensity pattern of the shaped field. It consists of the orientation and the depth of the emitter, where the orientation is given by the azimuthal and polar angle. Application of this method is widely expanded in biological sciences [3].

References

- [1] Backlund MP, Lew MD, Backer AS, Sahl SJ, Grover G, Agrawal A, Piestun R, Moerner WE. Simultaneous, accurate measurement of the 3D position and orientation of single molecules. *Proceedings of the National Academy of Sciences*. 2012 Nov 20;109(47):19087-92.
- [2] Pavani SR, Thompson MA, Biteen JS, Lord SJ, Liu N, Twieg RJ, Piestun R, Moerner WE. Three-dimensional, single-molecule fluorescence imaging beyond the diffraction limit by using a double-helix point spread function. *Proceedings of the National Academy of Sciences*. 2009 Mar 3;106(9):2995-9.
- [3] Backer AS, Moerner WE. Extending single-molecule microscopy using optical Fourier processing. *The Journal of Physical Chemistry B*. 2014 May 12;118(28):8313-29.

Apply to be considered for a student award (Yes / No)?

Yes

Level for award (Hons, MSc, PhD, N/A)?

MSc

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Would you like to submit a short paper for the Conference Proceedings (Yes / No)?

No

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