INDIAN INSTITUTE OF TECHNOLOGY GUWAHATI
PhD-17 SHORT ABSTRACT OF THESIS

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Thesis Title: Improved localisation and reconstruction in a single molecule localisation microscope to enable super-resolution imaging with lowered resource pre-requisites.

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SHORT ABSTRACT

Super-resolution microscopy techniques overcome the diffraction limit of conventional optical microscopes, enabling the visualisation of nanometre-scale structures using visible light. Among these, Single Molecule Localisation Microscopy (SMLM) including techniques like PALM, STORM, and PAINT allows super-resolution imaging using minimally modified fluorescence microscopes. By temporally separating the stochastic emission of fluorophores and localising them computationally, SMLM reconstructs images with nanometre-level precision.

Despite its effectiveness, SMLM faces several challenges. Image acquisition often requires thousands of frames, leading to long acquisition times, large datasets, and high computational demands. Weak fluorescence signals and detection noise complicate accurate emitter localisation, while overlapping signals in densely labelled regions introduce reconstruction artifacts. Additionally, prolonged imaging increases susceptibility to sample drift, further degrading image quality.

This thesis addresses these limitations with the goal of improving the accessibility of SMLM, particularly in low-resource settings. It presents a lightweight, modular, Python-based framework for emitter localisation and image reconstruction, integrated into a user-friendly graphical interface. The work introduces real-time localisation during acquisition, an adaptive thresholding method for improved signal detection in low SNR conditions, a convolutional neural network for resolving overlapping emitters, and a cross-correlation-based approach for drift correction during imaging.