LINEAGE TRACING IN MACROSCOPIC INTESTINAL ADENO-CARCINOMAS ENABLED BY LIGHT SHEET MICROSCOPY

Ruth Sims[1,2], Filipe C. Lourenco[2], Martin O. Lenz[1], Leila Muresan[1], Gopi Shah[2], Dario Bressan[2], Stefanie Reichelt[2], Kevin O'Holleran[1]
[1]Cambridge Advanced Imaging Centre, University of Cambridge
[2]Cancer Research UK – Cambridge Institute
E-mail : rs803@cam.ac.uk

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Cancer stem cells have long been associated with therapeutic failure [1]. Studies of fluorescently labelled clones in the intestinal epithelium can be used to assess the proliferation, differentiation and migration of these stem cells [2]. Imaging clones inside bulky and disorganised intestinal tumours requires 3D fluorescent imaging at single cell resolution. Imaging these structures at depth requires optical clearing methods to increase the optical transmittance of the tissue whilst preserving its morphology [3]. Since mouse intestinal adeino-carcinomas can have diameters larger than a centimetre, the imaging speeds of point scanning techniques present a bottleneck in the experimental pipeline, limiting acquisition rates to 1 sample/day. The coupling of light sheet illumination with sCMOS detectors has enabled significant increases in rates of acquisition, in addition to other benefits [4]. We have exploited the high-acquisition rates offered by light sheet microscopy for high-throughput imaging of macroscopic, cleared samples. Further to this, we have optimised sample mounting to facilitate imaging invasive adeino-carcinomas with variable morphologies at depths which have eluded other light microscopy modalities.

We will demonstrate how a bespoke light sheet microscope was designed and built based on specific sample requirements, and further how this setup was integrated into an efficient experimental workflow to facilitate lineage tracing via the analysis of hundreds of specimens.

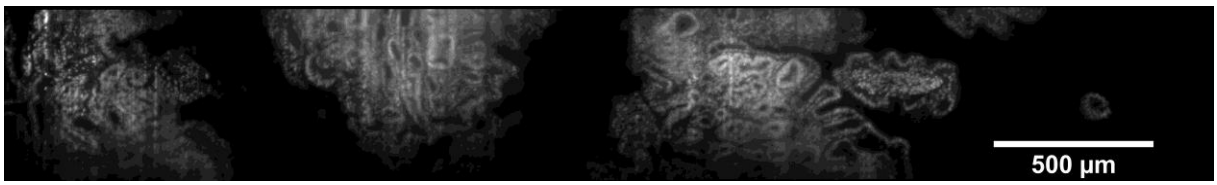

Figure 1: A single section of light sheet data-set of an invasive adeinocarcinoma (DAPI stained nuclei) demonstrating that single cell resolution can be achieved across large areas of dense tissue.

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