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Primed Track: Reliable Volumetric Single-cell Tracking and Lineage Tracing of Living Specimen with Dual-labeling Approaches

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Abstract Text

Mammalian embryonic development starts with a single fertilised zygote that develops into a blastocyst embryo consisting of three cell types that evolve into either embryonic or extra-embryonic tissues. Lineage tracing of these cells can provide important information about the molecular and cellular dynamics contributing to fate allocation during early development. While global labelling techniques allow for visualisation of all cells at the same time, lineage tracing of cells over several divisions can become complicated due to embryo movement and rotation as well as increasing cell densities. Here, we use green-to-red photoconvertible proteins for both global and sparse labelling of cells of interest in the developing murine embryo. We use primed conversion to achieve precise photoconversion of single nuclei in 4-cell stage embryos followed by volumetric live imaging to capture development up to the blastocyst stage. We developed an image analysis pipeline, called primed Track, that uses the dual labelling strategy for both straightforward segmentation and registration of all cells in the embryo as well as correction of rotational and spatial drift. Together, this strategy allows for reliable and fast tracking and lineage tracing of individual cells, even over increased imaging time intervals that result in a major reduction in data volume, all essential conditions for volumetric long-term imaging techniques.