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Smart adaptive multi-sample imaging for multi-view light sheet microscopy – when the microscope decides what to look at

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Light sheet microscopy of early embryo developmental stages is challenging. A significant number of time-lapse acquisitions, started at the onset of development, need to be stopped due to suboptimal sample orientation, poor image quality, not-fertilised eggs or because the development process arrests due to the phototoxicity induced by the imaging process itself. To increase the likelihood of success, we devised a strategy based on simultaneous multi-sample imaging at a low frame rate coupled with smart, image-aware microscope control. The open source ClearControl framework allows flexible user-defined assembly of instructions, such as device-based instructions (e.g. image acquisition), GPU-accelerated image post-processing and analysis, adaptive instructions (e.g. auto focus) and smart instructions (e.g. sample selection).

The advantages of our strategy are twofold: first, it reduces phototoxic effects by minimizing sample exposure, and second, it allows the screening of several samples in parallel, until an operator-independent algorithm decides for a sample to image in more temporal detail and with higher signal-to-noise-ratio.

We implemented our multi-sample imaging strategy on a multi-view light sheet microscope – the XWingScope. FEP tubes with typically five embryos of *Drosophila melanogaster* in the early cleavage stage were mounted and serially imaged by using a motorised stage. The acquired volumetric images were analysed by searching for a rapid rise in an entropy based image quality metric, which is used to detect the formation of the syncytial blastoderm and to predict when the first cells will invaginate. The presented strategy allows to select a sample automatically, auto-focus it, and continue imaging to capture the beginning of gastrulation with increased temporal resolution. Further improvements, more sophisticated control and decision-making algorithms, and other applications are made possible by our highly modular and extensible framework.

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