4D QUANTITATIVE TRACKING OF CELL CYCLE PROGRESSION IN LIVE DROSOPHILA EMBRYOS

Wee Choo Puah, Martin Wasser
Division of Imaging Informatics
Bioinformatics Institute, A*STAR
30 Biopolis Street, Singapore 138671
E-mail: puahwc@bii.a-star.edu.sg

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Cell cycle progression is a highly regulated process and involves a continuous change in chromosomal phenotype, observable in 4-D live-cell imaging [1]. Abnormalities detected during cell cycle progression such as cell cycle division and/or mitosis duration can provide insights into gene function/s in cell cycle regulation. This continuous change, as well as any cell cycle abnormalities, is otherwise not accurately detected when using fixed samples. Using 4-D live-cell imaging, we quantify how this change in chromosomal phenotype is used to monitor cell cycle progression and how measurements in specific cell cycle phases can be used to detect cell cycle abnormalities between wildtype and maternal haploid (mh') embryos.

In order to monitor cell cycle progression, we track nuclei which are tagged with Histone2AvDGFP, a GFP tagged fusion protein in live Drosophila embryos. As nuclei do not move with constant velocity during mitosis and gastrulation, tracking of nucleus’ movement is a non-trivial problem. The tracking algorithm in our quantitative study of cell cycle progression is modified from Kalman Filter [2], with incorporation of actual nuclear velocity update after every time-step. This tracking approach overcomes the need for sufficient overlap of nuclei between time-frames and the presence of corresponding nuclei in the next time-frame in order for tracking to continue. Instead of processing selected nuclei, which requires some form of human interference, we track all visible nuclei observed in the 4-D time-lapse dataset. Trajectories of nuclei moving in an orchestrated manner in early Drosophila embryos, as well as their tracking accuracy will be shown. The features of tracked nuclei, including intensity and volume, are then extracted to monitor cell cycle dependent phenotypic differences, such as differences in ploidy, between wildtype and maternal haploid (mh') embryos.

REFERENCES: