

Workshop III: Advanced Cell Cycle Analysis

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The objective of measuring cell proliferation is usually to obtain an estimate of the population growth rate, degree of cell cycle perturbation, and/or sub-compartment (phase) specific effects.

The objective of this workshop is to show and discuss the mechanics of cell cycle analysis by laser scanning cytometry (LSC). Human cells growing in either an optical culture dish or chamber slide will be stained prior to the workshop for DNA content, either cyclin A2 or cyclin B1, and an epitope that is phosphorylated more often in mitosis (in this case, phospho-S10-histone H3).

We will show:

- 1) Isolation of 2C → 4C interphase cells and quantification of the fractions of G1, S, G2.
- 2) Isolation of cells entering mitosis
- 3) Isolation of cells in mitosis; enumeration of the mitotic stages by chromatin condensation, cyclin expression, and morphology

Additionally, since we can measure the local density of cells on the slide, we will ask whether we can detect differences in the cell cycle phase distribution, and therefore estimate relative proliferation rate, as a function of cell contact.