

# Multi-parametric Analysis of the Apoptotic Decision Process in HT-29 Cells

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## ABSTRACT

Apoptosis, a specific, programmed sequence of events that results in a morphologically distinct type of cell death, is a critical process in all higher organisms and is an important factor in a number of human diseases, such as cancer and stroke. Apoptosis is controlled by a network of survival and death and signals that originate both inside and outside the cell. For example extracellular ligands, such as tumor necrosis factor (TNF) initiate the apoptotic process whereas insulin and epidermal growth factor (EGF), when used to costimulate cells under TNF treatment, are able to protect cells against death. We are interested in understanding the internal network through which cells decide to commit to apoptosis or to continue to proliferate. We have chosen as a model system the human colon carcinoma adherent cell line HT-29. We are implementing a number of high-throughput and quantitative techniques to gather molecular information about the death decision network in these cells. Flow cytometry is being used to

examine the extent of cell death and to monitor receptor behavior, while western blots are used to quantitatively measure the levels and phosphorylation states of a number of proteins known to be involved in the apoptosis signaling pathway. A cDNA microarray has been developed to measure the transcriptional levels of over 600 genes involved in apoptosis and proliferation. We are attempting to model information flow within the apoptosis regulatory cascade using a variety of methods including Markov chain analysis.

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