# NFκB Mediates Inflammation but not Survival in Individual Cells Treated with TRAIL

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***Short Abstract* —NFκB translocates to the nucleus following treatment with death ligands, where it mediates transcriptional activation of inflammatory and survival genes. To investigate the role of NFκB in mediating life-death decisions of individual cells treated with TRAIL (TNF-Related Apoptosis Inducing Ligand), we followed the fate of the surviving fraction of cells following treatment. We found that these cells transiently upregulated pathways associated with inflammation and survival. However, blocking NFκB signaling prevented inflammatory pathway activation but did not prevent survival.**

***Keywords* — TRAIL, apoptosis, cancer, cell-to-cell variability, NFκB, inflammation**

## I. Introduction

TRAIL holds promise as an anti-cancer agent due to its relative toxicity toward tumor cells compared with normal tissue, but many tumors are only partially sensitive to TRAIL [1]. Partial sensitivity to chemotherapy has often been ascribed to genetically distinct subpopulations or to cell cycle effects. Nongenetic variability and its effects on cell behavior have received recent attention in diverse systems from bacteria to mammalian cells; while much literature exists on the determinants of TRAIL sensitivity in different cancers, the contribution of nongenetic variability to partial TRAIL sensitivity has only begun to be explored [2].

TRAIL induces apoptosis via the extrinsic, or receptor-based, cell death pathway. However, in certain contexts TRAIL can also activate pro-survival pathways, such as the transcription factor NFκB [3]. NFκB is often presumed to mediate life-death decisions in individual cells treated with death ligands. However, some cell types exhibit NFκB activation following TRAIL or Fas stimulation, even in cells which go on to die [4]. In this study, we investigated whether NFκB mediates the survival of individual cells which escape apoptosis following treatment with death ligands.

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## II. Results

Cells which survived an initial treatment with TRAIL were examined for their ability to undergo apoptosis in response to a second treatment with TRAIL. We previously observed that these cells were transiently resistant to a second TRAIL treatment, but regained sensitivity over the course of several days. Periodic doses of TRAIL sustained resistance, suggesting a contribution from TRAIL-induced survival pathways [5].

NFκB nuclear translocation was apparent in subpopulations of cells following TRAIL-treatment; however, blocking NFκB signaling did not prevent transient or sustained resistance of survivor cells. NFκB did mediate expression of inflammatory cytokines and other wound response genes, as determined by gene expression microarray analysis, and was also partially responsible for transient phenotypic changes observed in survivor cells. One of the genes upregulated in survivor cells independently of NFκB was c-FLIP, an anti-apoptotic protein localized to the death-inducing signaling complex (DISC). DISC signaling was impaired in survivor cells, contributing to TRAIL resistance in an NFκB-independent manner.

## III. Conclusions

TRAIL is a potent mediator of apoptosis in cancer cells, but can also activate inflammation and metastasis in cell types which are resistant to apoptosis. In this study we demonstrate that the surviving sub-fraction of a cell population treated with TRAIL exhibits activation and expression of inflammatory pathways and genes. Some of these pathways are mediated by NFκB; however, our data calls into question the prevailing belief that NFκB is a mediator of life-death decisions in TRAIL-treated cells, and suggests instead a contribution from other pathways.

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