Model-driven exploration of apoptosis or necroptosis cell-death decisions.

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***Short Abstract* — Programmed necrosis (a.k.a. necroptosis) has recently emerged as a programmed cell-death (PCD) alternative to apoptosis. Death receptor mediated signaling can induce either apoptotic or necroptotic cell death, through shared biochemical network machinery, and thus represents an ideal system to understand molecular mechanisms associated with cell-decision processes. In this work we show how novel mathematical modeling approaches can be combined with experimental data to explore multiple mechanistic hypotheses about apoptosis or necroptosis outcomes. We test multiple model execution hypotheses and find viable mechanisms that are validated through experiments. In particular we find that the core decision-making machinery seems to relate to the Rip1-Caspase 8-Bid set of interactions.**

***Keywords* — Apoptosis, necroptosis, programmed cell death, ordinary differential equations, model feature extraction, global sensitivity, algebraic geometry, parameter space sampling.**

I. Background

Apoptosis and necroptosis are the two main forms of programmed cell death (PCD), a genetically programmed and tightly conserved mechanism for cell removal in higher organisms.[1,2] Dysregulation in necroptosis signaling has been identified as an important contributor to pathologies of a number of human diseases such as myocardial infarct, inflammatory bowel diseases, stroke, and neurodegeneration. In this work, we build cell-death decision-making models, in tandem with experiments, to explain cell deatg outcomes across a range of conditions. Our models explore the competing interactions that lead to clear unambiguous outcomes: cells either implode (apoptosis) or explode (necroptosis).[3,4] Apoptosis and necroptosis mechanisms share upstream signaling components but diverge prior to commitment to the final death outcome, thus providing rich environment to explore how multiple complex protein interactions lead to clear cell decisions.[1]

II. Approach

We build mathematical models that explore the apoptosis-necroptosis cell-death decision based on experimentally identified regulatory proteins and their interactions (Figure 1). The experiments have been designed to preferentially undergo either apoptosis or necroptosis, which allows us to collect precise mechanistic information from multiple biological conditions. We build and calibrate multiple model topologies to experimental data, using our PySB models-as-programs framework,[6] to explore multiple explanations to experimental conditions. Our models are based on Myeloid Progenitor Cells (MPCs) or Jurkat cells due to their tight control of cell-death mechanisms.[5] We analyze our models using novel algebraic geometry[7] and statistical global sensitivity[8] tools to extract mechanistic information about how molecular interactions lead to PCD outcomes.

III. Results

Our findings suggest that Bid, Caspase-8, and Rip1 can inhibit or modulate Rip1-driven necroptosis or apoptosis. These outcomes can be explained based on relative protein concentrations. We find that the best fits to experimental data occur when a regulatory step for Bid is accentuated and is sensitive to the relative concentrations and interactions with Rip1 and Caspase-8. We also find plausible mechanisms that indicate that some cells could be biased to necroptosis or apoptosis under a range of protein concentrations. The models guided experimental tests that served to narrow down our understanding of the cell-death decision mechanisms.

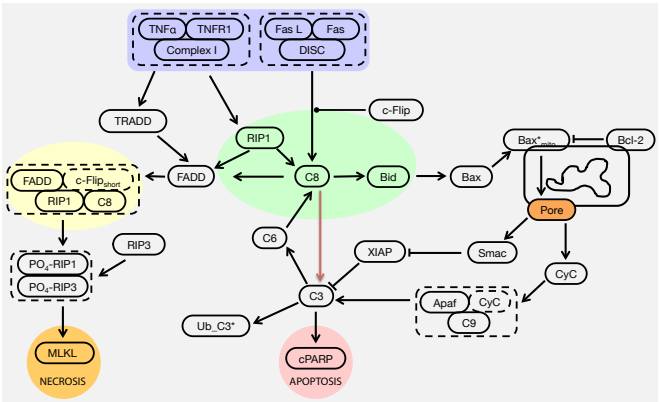

Figure 1: *Cartoon depiction of ANRM.* Initial signal leads to *DISC* and *Complex 1* formation (blue). We hypothesize the main decision node involves Rip1, Caspase-8 (C8) and Bid (green). If the signal proceeds to the right, it will undergo Bax- or Bak-dependent pore formation and eventual apoptosis, measured by cleaved PARP (cPARP) (red circle). Alternatively the signal could go through the necrosome (yellow) and lead to necrosis through MLKL activation (orange).

1. Green, D. R., Oberst, A., Dillon, C. P., Weinlich, R. & Salvesen, G. RIPK-dependent necrosis and its regulation by caspases: a mystery in five acts. *Molecular Cell* **44,** 9–16 (2011).
2. Vandenabeele, P., Galluzzi, L., Vanden Berghe, T. & Kroemer, G. Molecular mechanisms of necroptosis: an ordered cellular explosion. *Nat Rev Mol Cell Biol* **11,** 700–714 (2010).
3. Elmore, S. Apoptosis: A Review of Programmed Cell Death. *Toxicologic Path.* **35,** 495–516 (2007).
4. Wu, W., Liu, P. & Li, J. Necroptosis: an emerging form of programmed cell death. *Critical Reviews in Oncology/Hematology* (2012).
5. Zinkel, S. S. Proapoptotic BID is required for myeloid homeostasis and tumor suppression. *Genes \& Development* **17,** 229–239 (2003).
6. Lopez, C. F., Muhlich, J. L., Bachman, J. A. & Sorger, P. K. Programming biological models in Python using PySB. *Molecular Systems Biology* **9,** – (2013).
7. Radulescu, O., Gorban, A. N., Zinovyev, A. & Noel, V. Reduction of dynamical biochemical reactions networks in computational biology. *Front. Gene.* **3,** (2012).
8. Saltelli, A. Making best use of model evaluations to compute sensitivity indices. *Computer Physics Communications* **145,** 280–297 (2002).

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