Construction and analysis of a modular model of caspase activation in apoptosis

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[1] COB Colloquium, NYU

Introduction

Background: Apoptosis

Apoptosis is a conserved, highly regulated form of programmed cell death in multicellular organisms.

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- Dysregulation associated with pathological conditions
- Characteristic cell death morphology

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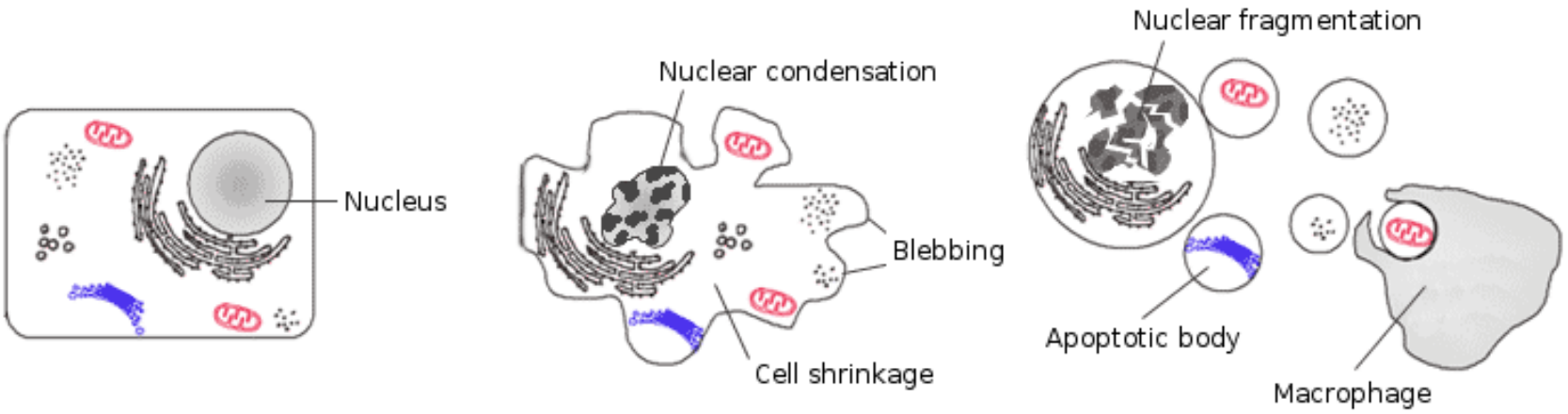
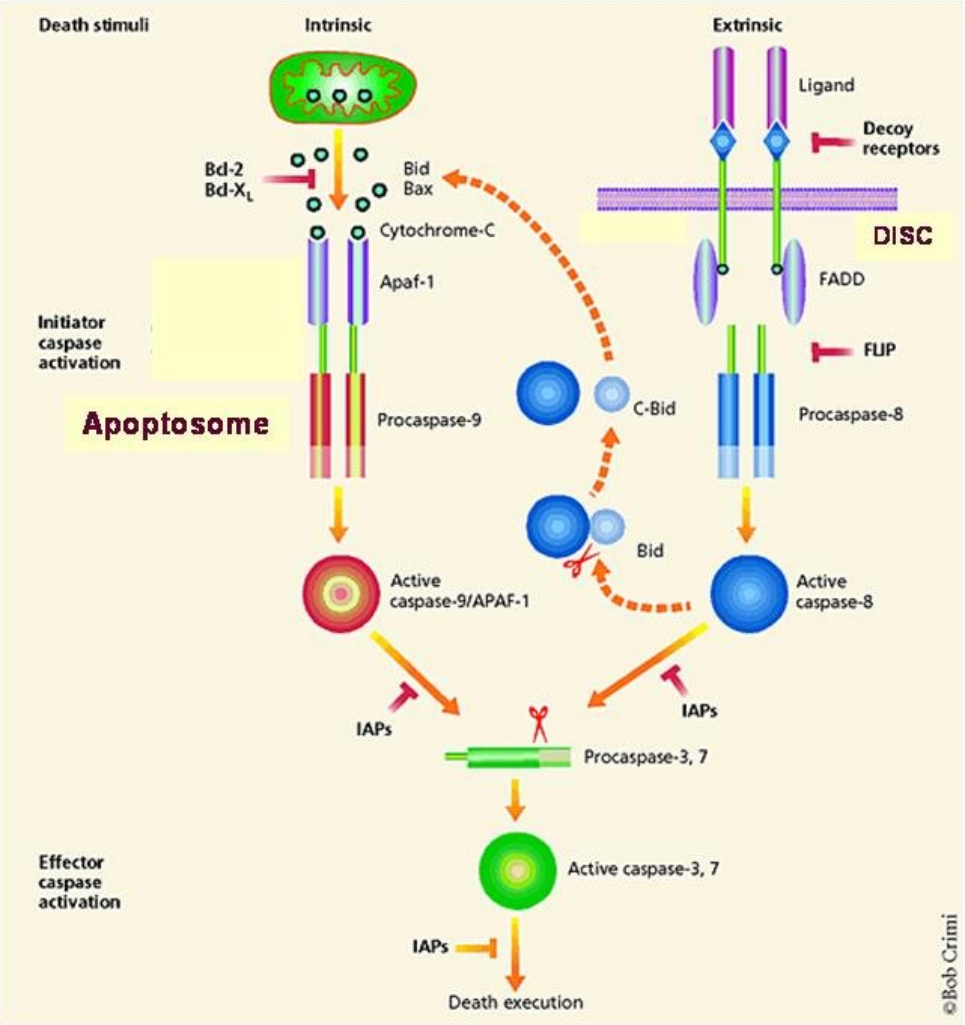


adapted from Wikipedia

Introduction

Background: Signaling pathways



Zheng and Flavell (2000)

Introduction

Approach

- Model construction by integration of previous models from the literature

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- Simplification of dynamics by steady-state abstraction of oligomerization kinetics

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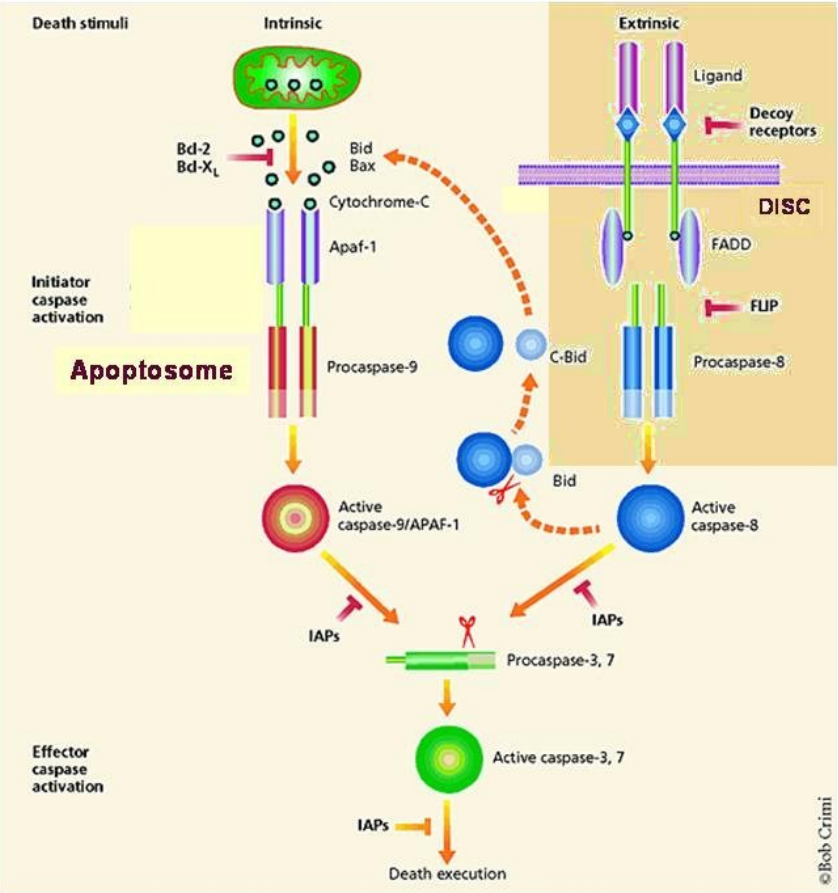
Approach

- Model construction by integration of previous models from the literature
- Simplification of dynamics by steady-state abstraction of oligomerization kinetics
- Linear regression to identify essential reactions
- Reduced models and validation

Model construction

Network integration: DISC subnetwork



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A MATHEMATICAL MODEL OF RECEPTOR-MEDIATED APOPTOSIS: DYING TO KNOW WHY *FASL* IS A TRIMER

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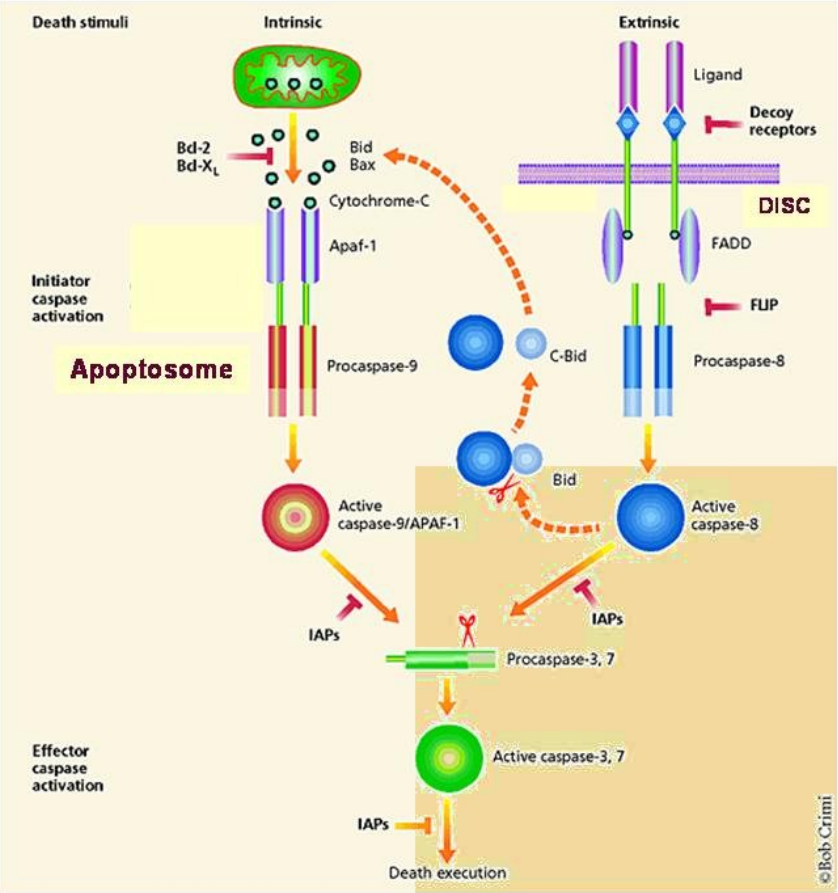
(Communicated by Yang Kuang)

Abstract. The scientific importance of understanding programmed cell death is undeniable; however, the complexity of death signal propagation and the formerly incomplete knowledge of apoptotic pathways has left this topic virtually untouched by mathematical modeling. In this paper, we use a mechanistic approach to frame the current understanding of receptor-mediated apoptosis with an immediate goal of isolating the role receptor trimerization plays in this process. Analysis and simulation suggest that if the death signal is to be successful at low-receptor, high-ligand concentration, Fas trimerization is unlikely to be the driving force in the signal propagation. However at high-receptor and low-ligand concentrations, the mathematical model illustrates how the ability of FasL to cluster three Fas receptors can be crucially important for downstream events that propagate the apoptotic signal.

1. **Introduction.** Apoptosis or programmed cell death (PCD) is a critical process in normal tissue development [1]. It is the primary mechanism through which cells are removed when malfunctions arise from cell stress, cell damage, or conflicting cell division signals [2]. Maintaining the balance between programmed cell death and cell survival is fundamentally important since disturbing this equilibrium can lead to a number of pathological disorders. The core component of the cell suicide

Model construction

Network integration: Extrinsic subnetwork



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Bistability Analyses of a Caspase Activation Model for Receptor-induced Apoptosis*

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Apoptosis is an important physiological process crucially involved in development and homeostasis of multicellular organisms. Although the major signaling pathways have been unraveled, a detailed mechanistic understanding of the complex underlying network remains elusive. We have translated here the current knowledge of the molecular mechanisms of the death-receptor-activated caspase cascade into a mathematical model. A reduction down to the apoptotic core machinery enables the application of analytical mathematical methods to evaluate the system behavior within a wide range of parameters. Using parameter values from the literature, the model reveals an unstable status of survival indicating the need for further control. Based on recent publications we tested one additional regulatory mechanism at the level of initiator caspase activation and demonstrated that the resulting system displays desired characteristics such as bistability. In addition, the results from our model studies allowed us to reconcile the fast kinetics of caspase 3 activation observed at the single cell level with the much slower kinetics found at the level of a cell population.

Apoptosis is a genetically defined major form of programmed cell death enabling the organism to remove unwanted cells, *e.g.* during embryonal development and after immune responses, to select educated immune cells and to eliminate virally infected and transformed cells (1, 2). Enhanced or inhibited apoptotic cell death can be involved in severe pathological alterations, including developmental defects, autoimmune diseases, neurodegeneration, or cancer. Extrinsic and intrinsic apoptotic pathways can be distinguished, although partly employing overlapping signal transduction pathways. A hallmark of the ongoing

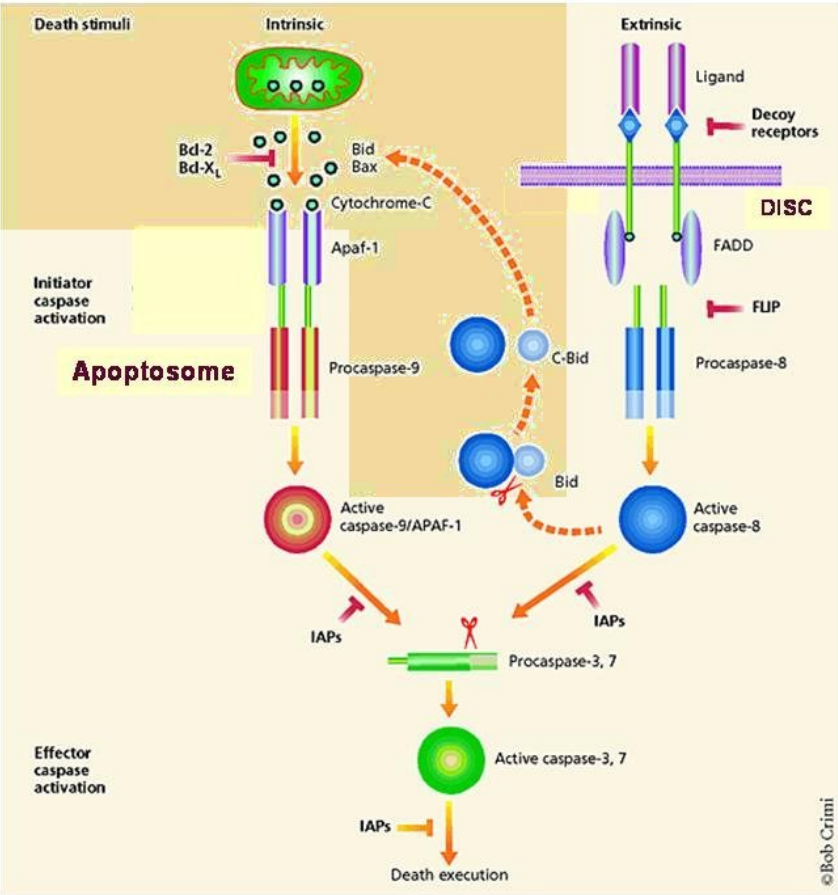
outline of the extrinsic pathway of apoptosis induction after death receptor stimulation is depicted in Fig. 1.

Mathematical modeling and systems theory can provide valuable tools to get insight into complex dynamical systems, to test hypotheses, and to identify weak points (5, 6). Previous modeling approaches in apoptosis focused on the extrinsically triggered pathways, resulting in complex models (7, 8). The model parameters were fitted to data derived from cell population studies showing caspase activation in a range from 30 min to several hours. These models can describe and nicely illustrate certain aspects of the signal transduction pathway. However, more recent experimental results performed at the single cell level show that the majority of caspases are activated within a very short time interval (<15 min) (9–12).

Obviously, the single cell level is relevant for a mechanistic understanding. With the focus on receptor-induced apoptosis, we used Monte Carlo methods to look for parameter domains that enable an appropriate description of apoptosis induction in a single cell (model based on Fig. 1, data not shown). The obtained results revealed an unexpected responsiveness of the system toward minute initiator caspase activation if required to act rapidly. This behavior of the model was caused by the caspase cascade that represents the main signaling route in so-called type I cells (13) (see Fig. 1, *yellow background*). We therefore translated the current picture of the extrinsically triggered caspase cascade in a very elementary form into a mathematical model enabling a thorough investigation through the application of analytical methods. Our results showed that within large parameter ranges, including values from the literature, this straightforward model structure is unable to appropriately describe the expected behavior that can be deduced from experimental data. We then showed a way of extending our model structure to reconcile these observed differences and presented a model now able to describe key

Model construction

Network integration: Coupling subnetwork



The Journal of Immunology

Effects of Bcl-2 Levels on Fas Signaling-Induced Caspase-3 Activation: Molecular Genetic Tests of Computational Model Predictions[1]

Fei Hua,[2]* Melanie G. Cornejo,* Michael H. Cardone,‡ Cynthia L. Stokes,§ and Douglas A. Lauffenburger*†

Fas-induced apoptosis is a critical process for normal immune system development and function. Although many molecular components in the Fas signaling pathway have been identified, a systematic understanding of how they work together to determine network dynamics and apoptosis itself has remained elusive. To address this, we generated a computational model for interpreting and predicting effects of pathway component properties. The model integrates current information concerning the signaling network downstream of Fas activation, through both type I and type II pathways, until activation of caspase-3. Unknown parameter values in the model were estimated using experimental data obtained from human Jurkat T cells. To elucidate critical signaling network properties, we examined the effects of altering the level of Bcl-2 on the kinetics of caspase-3 activation, using both overexpression and knockdown in the model and experimentally. Overexpression was used to distinguish among alternative hypotheses for inhibitory binding interactions of Bcl-2 with various components in the mitochondrial pathway. In comparing model simulations with experimental results, we find the best agreement when Bcl-2 blocks the release of cytochrome *c* by binding to both Bax and truncated Bid instead of Bax, truncated Bid, or Bid alone. Moreover, although Bcl-2 overexpression strongly reduces caspase-3 activation, Bcl-2 knockdown has a negligible effect, demonstrating a general model finding that varying the expression levels of signal molecules frequently has asymmetric effects on the outcome. Finally, we demonstrate that the relative dominance of type I vs type II pathways can be switched by varying particular signaling component levels without changing network structure. ***The Journal of Immunology,*** **2005, 175: 985–995.**

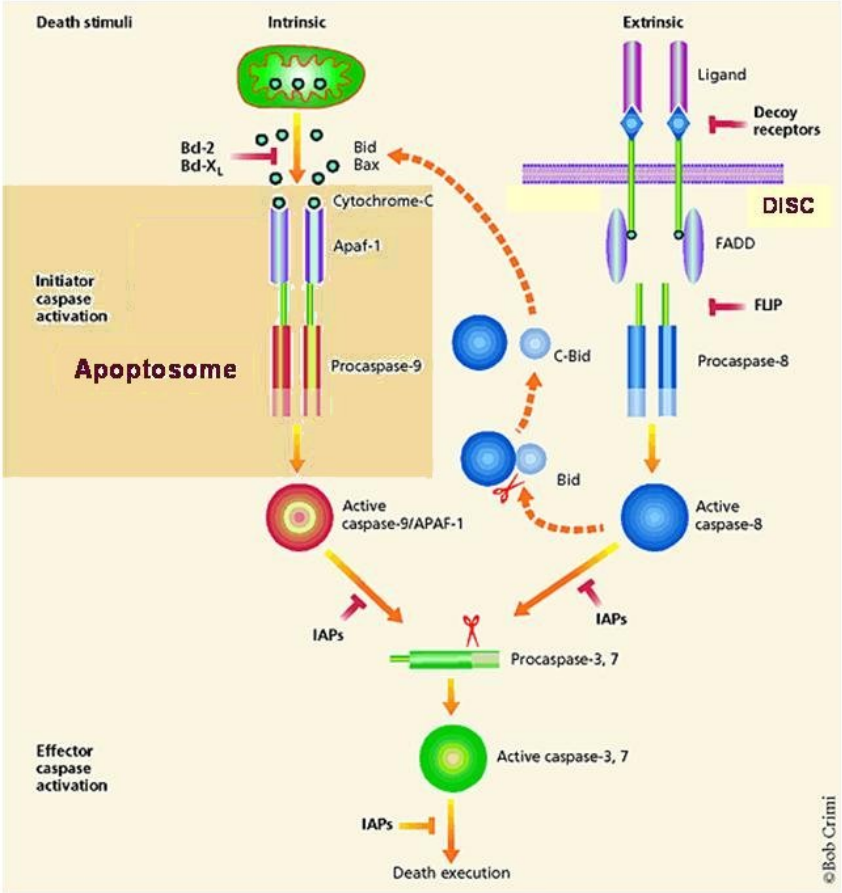
Apoptosis is an essential cellular event for maintaining homeostasis of the immune system and its normal function. Dysregulation of apoptosis can contribute to various autoimmune diseases and cancer (1, 2). One major mechanism for inducing apoptosis is through the activation of death receptors such as TNF, Fas (Apo-1/CD95), DR3 (TRAMP), DR4 (TRAIL-R1), and DR5 (TRAIL-R2) (3, 4). Among death receptors, the signaling pathways for Fas-induced apoptosis are the best characterized (5) (Fig. 1). Two pathways activated by Fas have been identified (6), and are referred to as type I and type II pathways. For both pathways, caspases, a family of cysteine proteases, are crucial for both the initiation and execution of apoptosis. The pathways diverge after activation of initiator caspases (e.g., caspase-8 and caspase-10) and converge at the end by activating executor caspases (e.g., caspase-3). In the type I pathway, initiator caspases cleave and activate executor caspases directly. In the type II pathway, also called the mitochondrial pathway, a more complex sig-

multiple steps along both signaling cascades (2, 4, 5, 7). For instance, FLIP blocks activation of initiator caspases, Bcl-2 prevents mitochondrial disruption, and X-linked inhibitor of apoptosis protein (XIAP)[3] inhibits downstream caspases (i.e., caspase-9 and caspase-3).

Despite the characterization of the components of these apoptotic pathways, we have an incomplete understanding of how all of the signaling molecules fit together into a single coherent network, and how the quantitative and dynamic aspects of the network function in relation to the final cellular outcome (i.e., apoptosis vs survival). Studies in other systems, such as the epidermal growth factor receptor (8), have shown that using computational models to study the dynamic behavior of an integrated signaling system can contribute significantly to understanding complex biological processes. In this study, we organize biochemical and biophysical knowledge about the Fas signaling pathway into a mechanistic mathematical model to advance our understanding of the integra-

Model construction

Network integration: Apoptosome subnetwork



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A mathematical model for apoptosome assembly: The optimal cytochrome *c*/Apaf-1 ratio

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Abstract

Apoptosis, a highly conserved form of cell suicide, is regulated by apoptotic signals and their transduction with caspases, a family of cystein proteases. Caspases are constantly expressed in the normal cells as inactive pro-enzymes. The activity of caspase is regulated by the proteolysis. Sequential proteolytic reactions of caspases are needed to execute apoptosis. Mitochondrial pathway is one of these apoptotic signal pathways, in which caspases are oligomerized into characteristic heptamer structure, called apoptosome, with caspase-9 that activate the effector caspases for apoptosis. To investigate the dynamics of signal transduction pathway regulated by oligomerization, we construct a mathematical model for Apaf-1 heptamer assembly process. The model first reveals that intermediate products can remain unconverted even after all assemble reactions are completed. The second result of the model is that the conversion efficiency of Apaf-1 heptamer assembly is maximized when the initial concentration of cytochrome *c* is equal to that of Apaf-1. When the concentration of cytochrome *c* is sufficiently larger or smaller than that of Apaf-1, the final Apaf-1 heptamer production is decreased, because intermediate Apaf-1 oligomers (tetramers and bigger oligomers), which themselves are unable to form active heptamer, accumulate too fast in the cells, choking a smooth production of Apaf-1 heptamer. Slow activation of Apaf-1 monomers and small oligomers increase the conversion efficiency. We also study the optimal number of subunits comprising an active oligomer that maximize the conversion efficiency in assembly process, and found that the tetramer is the optimum.

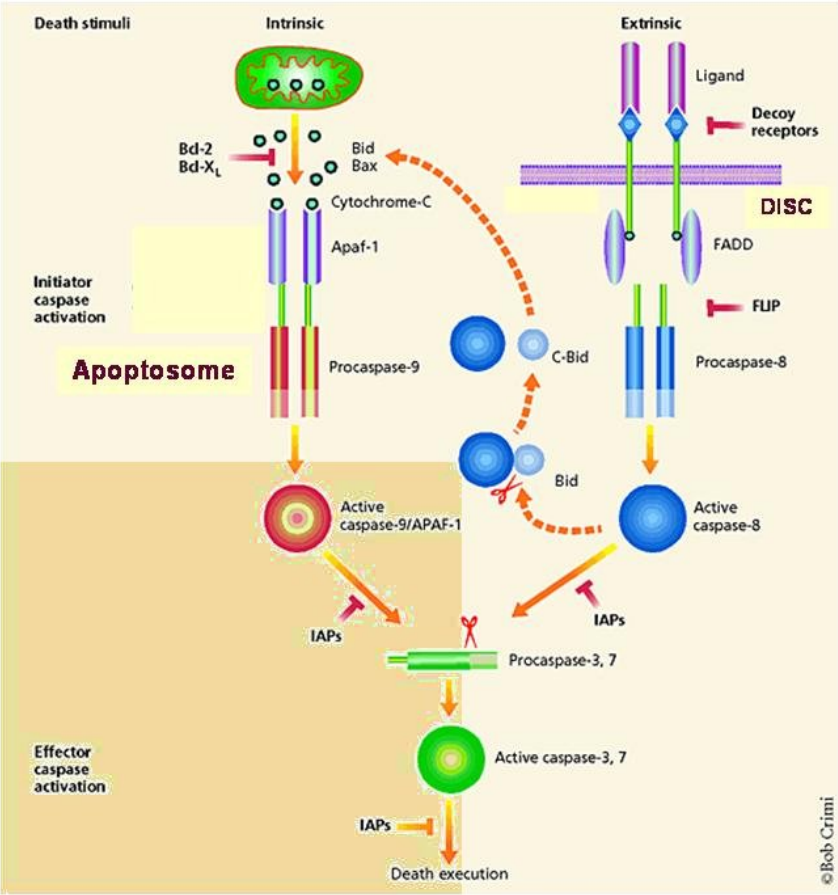
Keywords: Apoptosis; Apoptosome; Mitochondrial pathway; Cytochrome *c*; Apaf-1; Oligomerization

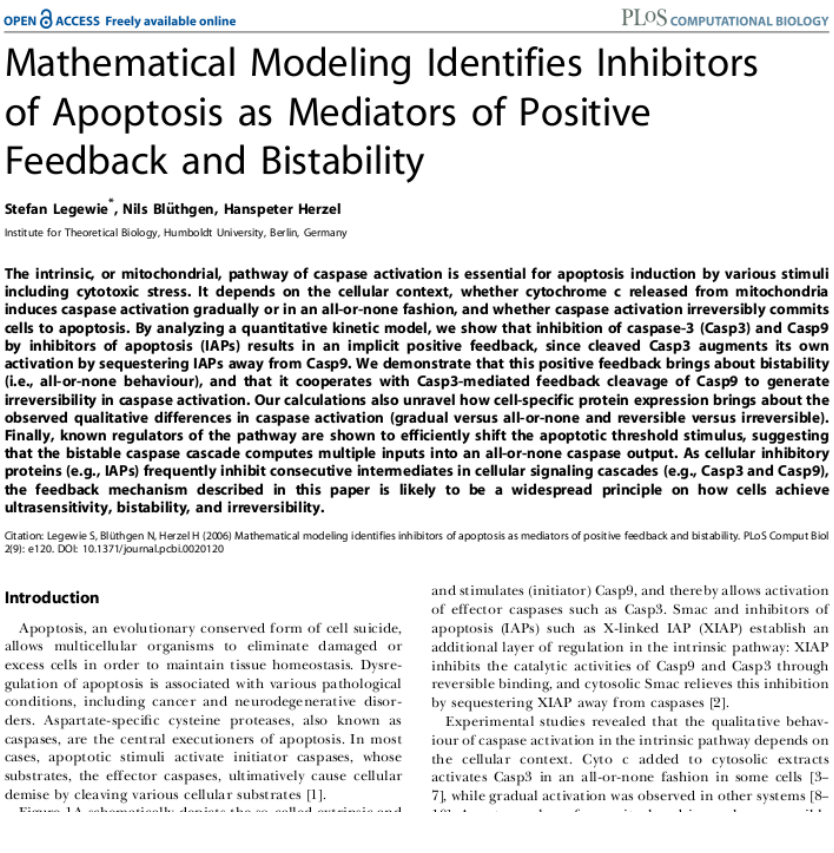
1. Introduction

Apoptosis is a highly conserved form of cell suicide, which plays a key role in controlling the cell number in multicellular organisms (Zimmermann et al., 2001). The apoptotic signals are transduced through the specific pathways. All of the apoptotic signal pathways are composed of the cascade of caspase reactions. Caspases are highly conserved cystein protease (Nunez et al., 1998) protein cleavage. Once caspase is cleaved into subunits, an active complex is produced from the large and the small subunits. The active complex of caspase can cleave the specific site of caspase and/or substrates in the downstream of the cascade. The apoptotic signal is transduced by this sequential activations of caspases. Caspases are divided into two classes by their functions in this cascade. One, called initiator caspase, takes part in the upstream of the cascade, which are activated by the receptor of the

Model construction

Network integration: Intrinsic subnetwork





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Mathematical Modeling Identifies Inhibitors of Apoptosis as Mediators of Positive Feedback and Bistability

Stefan Legewie*, Nils Blüthgen, Hanspeter Herzel
Institute for Theoretical Biology, Humboldt University, Berlin, Germany

The intrinsic, or mitochondrial, pathway of caspase activation is essential for apoptosis induction by various stimuli including cytotoxic stress. It depends on the cellular context, whether cytochrome c released from mitochondria induces caspase activation gradually or in an all-or-none fashion, and whether caspase activation irreversibly commits cells to apoptosis. By analyzing a quantitative kinetic model, we show that inhibition of caspase-3 (Casp3) and Casp9 by inhibitors of apoptosis (IAPs) results in an implicit positive feedback, since cleaved Casp3 augments its own activation by sequestering IAPs away from Casp9. We demonstrate that this positive feedback brings about bistability (i.e., all-or-none behaviour), and that it cooperates with Casp3-mediated feedback cleavage of Casp9 to generate irreversibility in caspase activation. Our calculations also unravel how cell-specific protein expression brings about the observed qualitative differences in caspase activation (gradual versus all-or-none and reversible versus irreversible). Finally, known regulators of the pathway are shown to efficiently shift the apoptotic threshold stimulus, suggesting that the bistable caspase cascade computes multiple inputs into an all-or-none caspase output. As cellular inhibitory proteins (e.g., IAPs) frequently inhibit consecutive intermediates in cellular signaling cascades (e.g., Casp3 and Casp9), the feedback mechanism described in this paper is likely to be a widespread principle on how cells achieve ultrasensitivity, bistability, and irreversibility.

Citation: Legewie S, Blüthgen N, Herzel H (2006) Mathematical modeling identifies inhibitors of apoptosis as mediators of positive feedback and bistability. PLoS Comput Biol 2(9): e120. DOI: 10.1371/journal.pcbi.0020120

Introduction

Apoptosis, an evolutionary conserved form of cell suicide, allows multicellular organisms to eliminate damaged or excess cells in order to maintain tissue homeostasis. Dysregulation of apoptosis is associated with various pathological conditions, including cancer and neurodegenerative disorders. Aspartate-specific cysteine proteases, also known as caspases, are the central executioners of apoptosis. In most cases, apoptotic stimuli activate initiator caspases, whose substrates, the effector caspases, ultimatively cause cellular demise by cleaving various cellular substrates [1].

and stimulates (initiator) Casp9, and thereby allows activation of effector caspases such as Casp3. Smac and inhibitors of apoptosis (IAPs) such as X-linked IAP (XIAP) establish an additional layer of regulation in the intrinsic pathway: XIAP inhibits the catalytic activities of Casp9 and Casp3 through reversible binding, and cytosolic Smac relieves this inhibition by sequestering XIAP away from caspases [2].

Experimental studies revealed that the qualitative behaviour of caspase activation in the intrinsic pathway depends on the cellular context. Cyto c added to cytosolic extracts activates Casp3 in an all-or-none fashion in some cells [3–7], while gradual activation was observed in other systems [8–

Model construction

Steady-state abstraction

For an oligomer X with intermediate structures $X_1, \ldots, X_n$ and dynamics

$$\frac{d\,[X]}{dt} = \underbrace{f\left([X], [X]_1, \ldots, [X]_n\right)}_{\text{synthesis}} - \underbrace{\mu\,[X]}_{\text{degradation}},$$

use the steady-state approximation $f \approx f_{ss} = \mu\,[X]_{ss}$.

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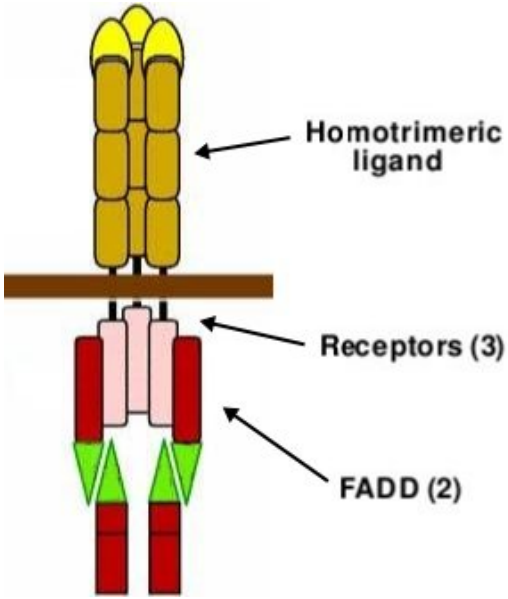
use the steady-state approximation $f \approx f_{ss} = \mu\,[X]_{ss}$. Fit μ by comparing full dynamics with reduced (first order) dynamics.

- This is not correct but is useful!
- Allows modularization of oligomerization kinetics

Model construction

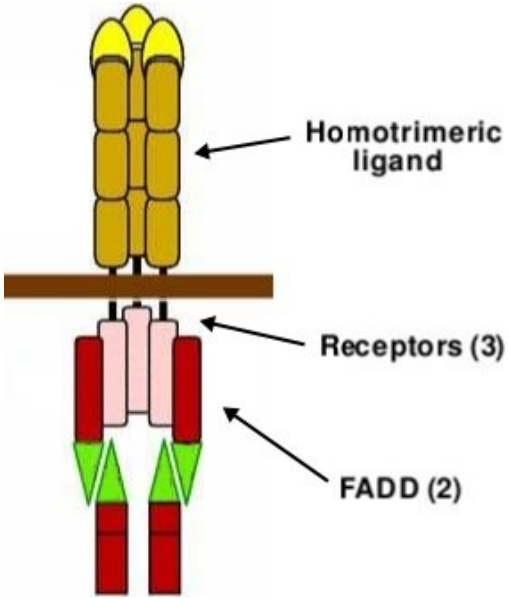
Steady-state abstraction: DISC module

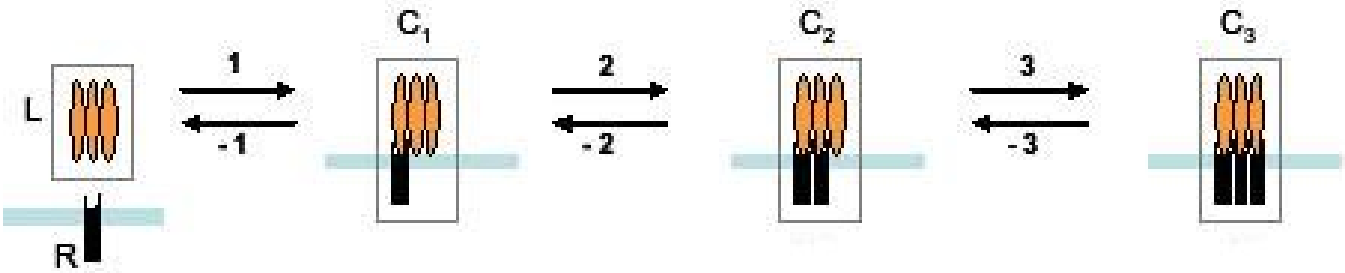


Lai and Jackson (2004), Aguda and Friedman (2008)

Model construction

Steady-state abstraction: DISC module

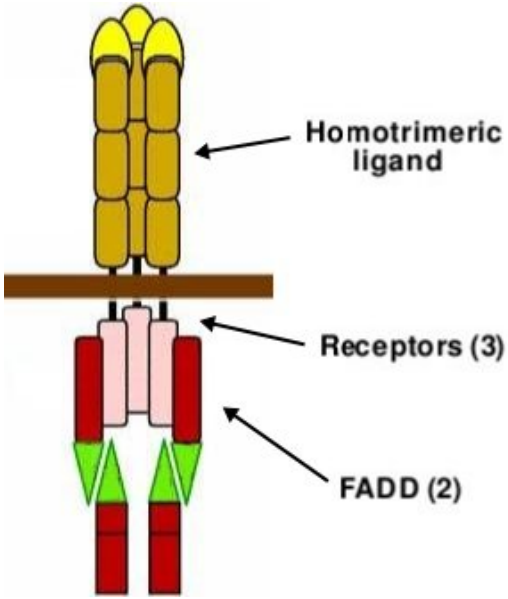


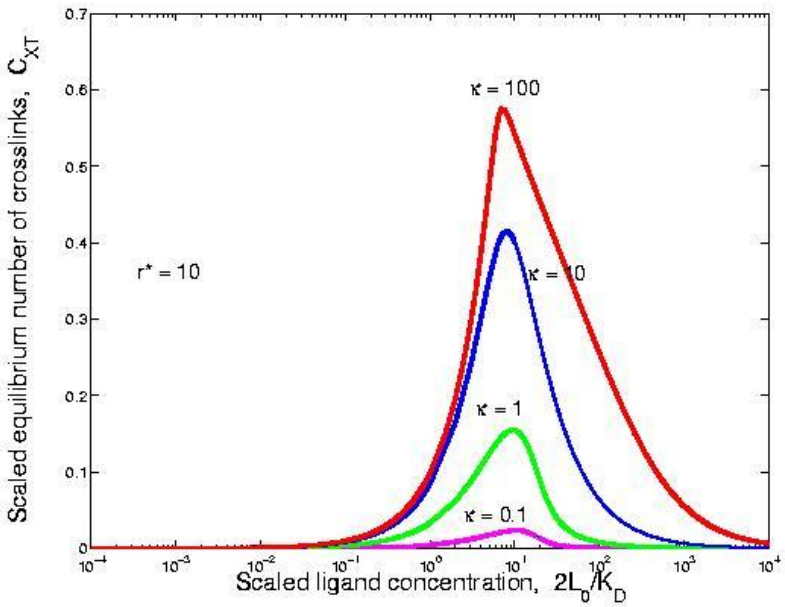


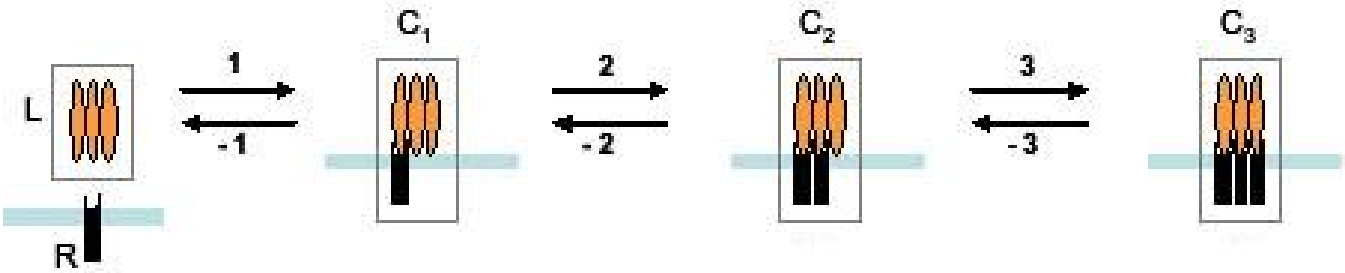
Lai and Jackson (2004), Aguda and Friedman (2008)

Model construction

Steady-state abstraction: DISC module







Lai and Jackson (2004), Aguda and Friedman (2008)

Model construction

Steady-state abstraction: MAC module

- Use tBid-Bax_2 as a functional surrogate of MAC, which releases Cyt*c* and Smac from mitochondria

Model construction

Steady-state abstraction: MAC module

- Use tBid-Bax$_2$ as a functional surrogate of MAC, which releases Cyt$c$ and Smac from mitochondria
- Analogously define a crosslinking model of formation:

$$\text{tBid} + \text{Bax} \underset{k_r}{\overset{2k_f}{\rightleftharpoons}} \text{tBid-Bax}, \quad \text{tBid-Bax} + \text{Bax} \underset{2k_r}{\overset{k_f}{\rightleftharpoons}} \text{tBid-Bax}_2$$

Model construction

Steady-state abstraction: MAC module

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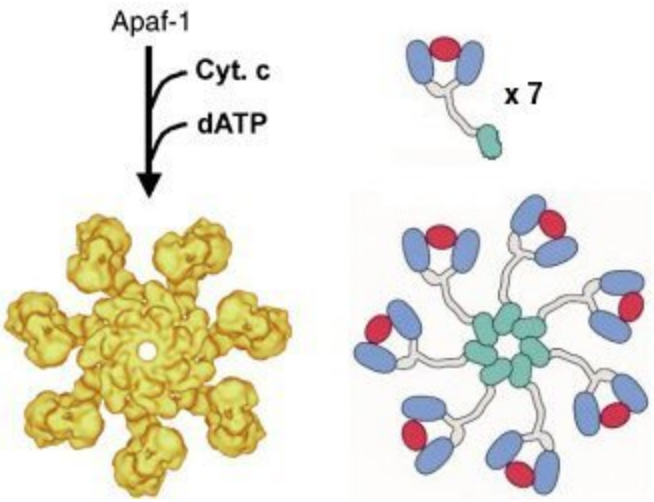
- Results are similar as for DISC

Model construction

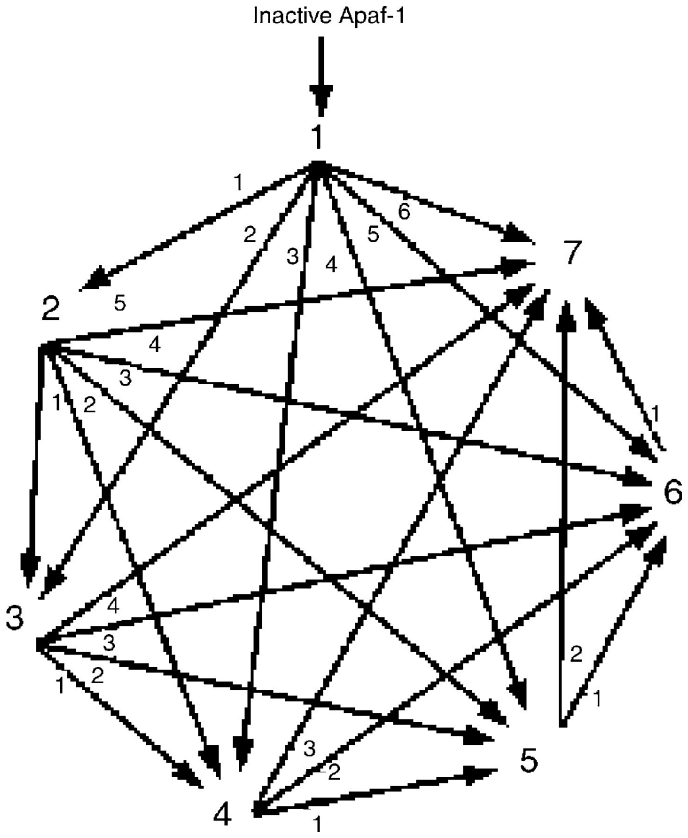
Steady-state abstraction: Apoptosome module



Acehan *et al.* (2002), Nakabayashi and Sasaki (2008)

Model construction

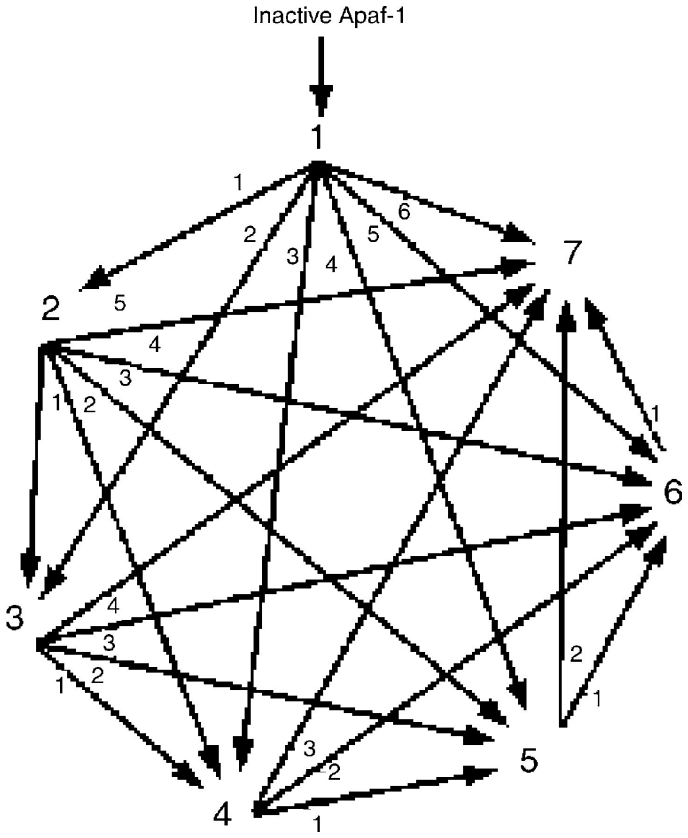
Steady-state abstraction: Apoptosome module

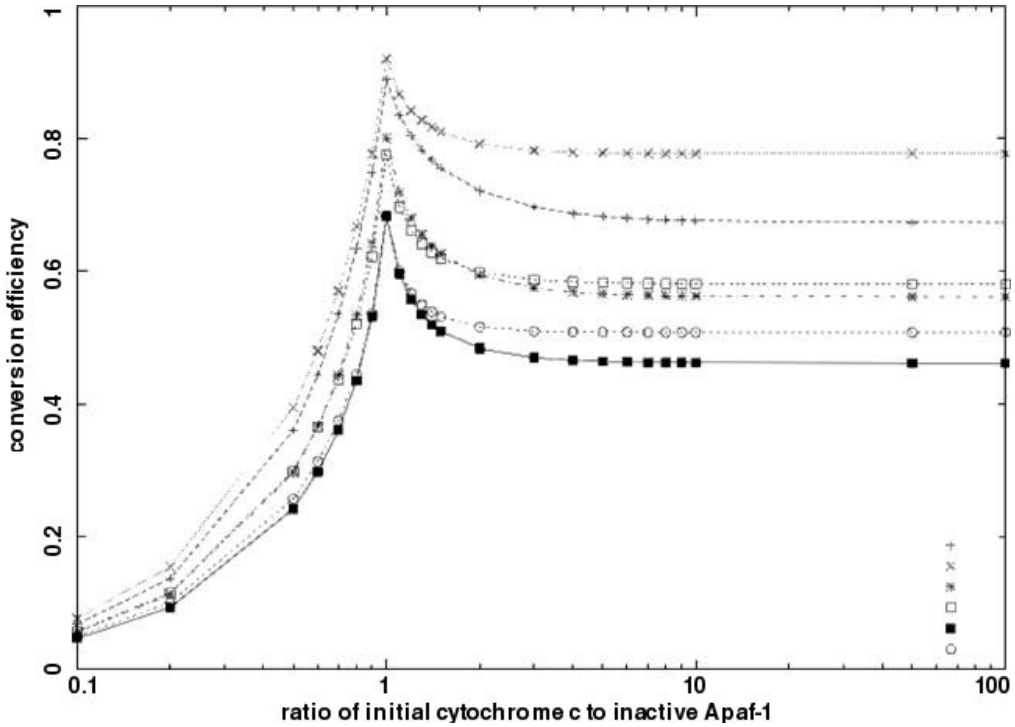


Acehan *et al.* (2002), Nakabayashi and Sasaki (2008)

Model construction

Steady-state abstraction: Apoptosome module

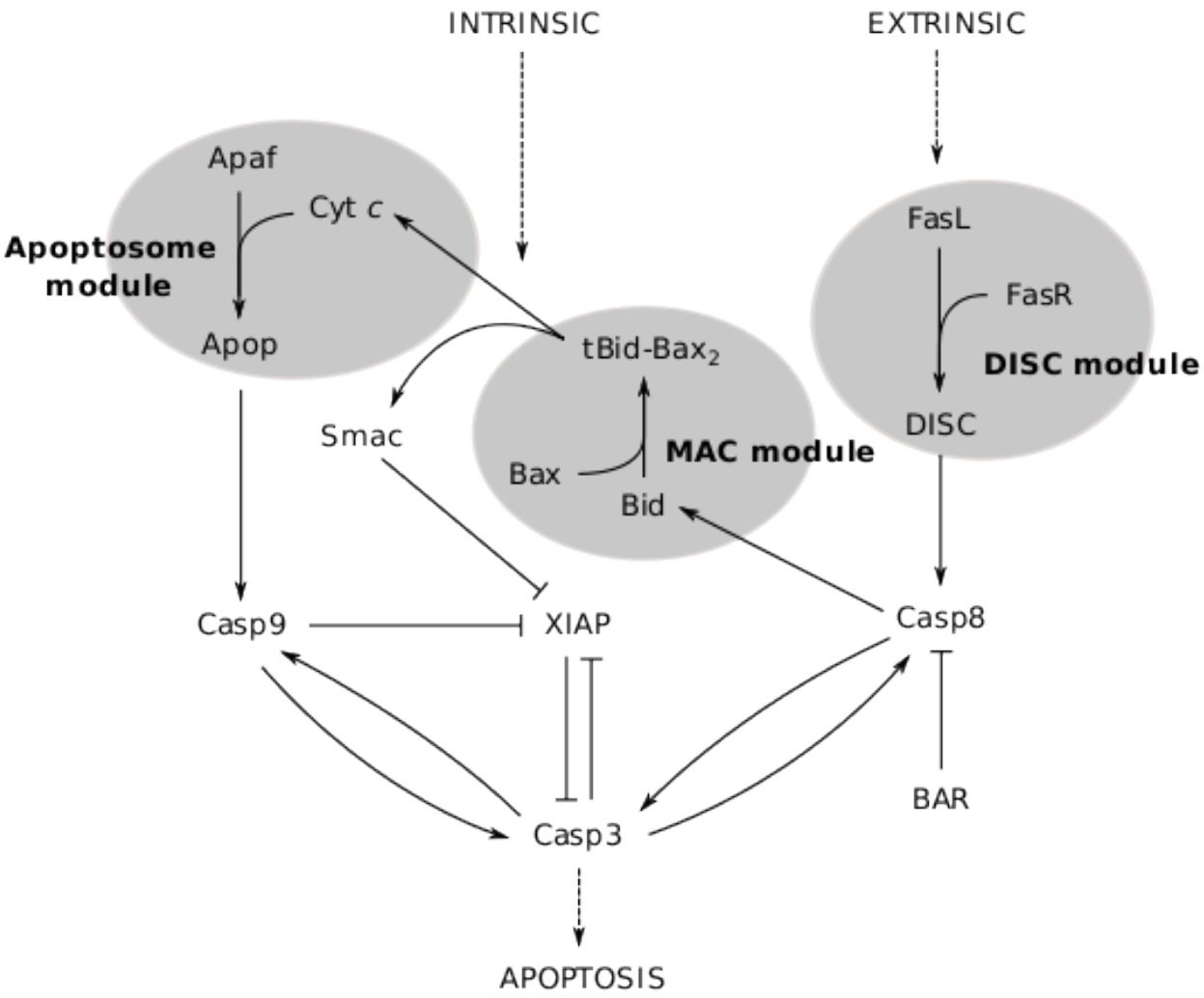




Acehan *et al.* (2002), Nakabayashi and Sasaki (2008)

Model construction

Full model: Reaction network



Model construction

Full model: Model species

Species	Description	Synthesis rate (nM/s)	Degradation rate (s^{-1})
DISC	DISC		8.807×10^{-3}
Casp8	procaspase-8	adjusted	6.5×10^{-5} [62]
Casp8*	caspase-8		9.667×10^{-5} [62]
Casp3	procaspase-3	adjusted	6.5×10^{-5} [62]
Casp3*	caspase-3		9.667×10^{-5} [62]
XIAP	XIAP	adjusted	1.933×10^{-4} [62]
Casp3*-XIAP	Casp3*-XIAP complex		2.883×10^{-4} [62]
BAR	BAR	1.111×10^{-3} ($[\mathrm{BAR}]_0 = 66.67$ nM [62])	1.667×10^{-5} [62]
Casp8*-BAR	Casp8*-BAR complex		1.933×10^{-4} [62]
Bid	Bid	4.168×10^{-4} ($[\mathrm{Bid}]_0 = 25$ nM [72,73])	1.667×10^{-5} (μ_{BAR})
tBid	truncated Bid		1.667×10^{-5} (μ_{Bid})
tBid-Bax$_2$	tBid-Bax$_2$ complex		0.0264
Cytc	cytochrome *c* (mitochondrial)	10^{-3} ($[\mathrm{Cytc}]_0 = 100$ nM [72,73])	10^{-5}
Cytc*	cytochrome *c* (cytosolic)		10^{-5}
Smac	Smac (mitochondrial)	0.0167 ($[\mathrm{Smac}]_0 = 100$ nM [72,73])	1.667×10^{-5} (μ_{BAR})
Smac*	Smac (cytosolic)		1.667×10^{-5} (μ_{Smac})
Smac*-XIAP	Smac-XIAP complex		1.933×10^{-4} ($\mu_{\mathrm{Casp8^*\text{-}BAR}}$)
Apop	apoptosome		1.487×10^{-5}
Casp9	procaspase-9	1.3×10^{-3} ($[\mathrm{Casp9}]_0 = 20$ nM [72,73])	6.5×10^{-5} (μ_{Casp8})
Casp9*	caspase-9		9.667×10^{-5} ($\mu_{\mathrm{Casp8^*}}$)
Casp9*-XIAP	Casp9*-XIAP complex		2.883×10^{-4} ($\mu_{\mathrm{Casp3^*\text{-}XIAP}}$)

Model construction

Full model: Model reactions

Number	Reaction	Forward rate ($nM^{-1}\,s^{-1}$)	Reverse rate (s^{-1})
DISC	$(\text{FasL}, \text{FasR}) \longrightarrow \text{DISC}$	f_{DISC}	
1	$\text{DISC} + \text{Casp8} \longrightarrow \text{DISC} + \text{Casp8}^*$	10^{-4} (k_2)	
2	$\text{Casp3}^* + \text{Casp8} \longrightarrow \text{Casp3}^* + \text{Casp8}^*$	10^{-4} [3]	
3	$\text{Casp8}^* + \text{Casp3} \longrightarrow \text{Casp8}^* + \text{Casp3}^*$	5.8×10^{-4} [3]	
4	$\text{Casp3}^* + \text{XIAP} \rightleftharpoons \text{Casp3}^*\text{-XIAP}$	3×10^{-3} [3]	0.035 [3]
5	$\text{Casp3}^* + \text{XIAP} \longrightarrow \text{Casp3}^*$	3×10^{-3} [3]	
6	$\text{Casp8}^* + \text{BAR} \rightleftharpoons \text{Casp8}^*\text{-BAR}$	5×10^{-3} [3]	0.035 [3]
7	$\text{Casp8}^* + \text{Bid} \longrightarrow \text{Casp8}^* + \text{tBid}$	5×10^{-4} (est. [4])	
MAC	$(\text{tBid}, \text{Bax}) \longrightarrow \text{tBid-Bax}_2$	$f_{\text{tBid-Bax}_2}$	
8	$\text{tBid-Bax}_2 + \text{Cytc} \longrightarrow \text{tBid-Bax}_2 + \text{Cytc}^*$	10^{-3} [4]	
9	$\text{tBid-Bax}_2 + \text{Smac} \longrightarrow \text{tBid-Bax}_2 + \text{Smac}^*$	10^{-3} [4]	
10	$\text{Smac}^* + \text{XIAP} \rightleftharpoons \text{Smac}^*\text{-XIAP}$	7×10^{-3} [4]	2.21×10^{-3} [4]
Apop	$(\text{Cytc}^*, \text{Apaf}) \longrightarrow \text{Apop}$	f_{Apop}	
11	$\text{Apop} + \text{Casp9} \longrightarrow \text{Apop} + \text{Casp9}^*$	2×10^{-4} (est. [8])	
12	$\text{Casp3}^* + \text{Casp9} \longrightarrow \text{Casp3}^* + \text{Casp9}^*$	2×10^{-4} [8]	
13	$\text{Casp9}^* + \text{Casp3} \longrightarrow \text{Casp9}^* + \text{Casp3}^*$	5×10^{-5} [8]	
14	$\text{Casp9}^* + \text{XIAP} \rightleftharpoons \text{Casp9}^*\text{-XIAP}$	1.06×10^{-4} [4]	10^{-3} [4]

Model construction

Full model: Dynamical equations

Differential equations	Reaction velocities
$d[\text{DISC}]/dt = \mu_{\text{DISC}}(f_{\text{DISC}}([\text{FasL}]_0, [\text{FasR}]_0; K_{\text{DISC}}) - [\text{DISC}])$	$v_1 = k_1 [\text{DISC}][\text{Casp8}]$
$d[\text{Casp8}]/dt = -v_1 - v_2 + \alpha_{\text{Casp8}} - \mu_{\text{Casp8}} [\text{Casp8}]$	$v_2 = k_2 [\text{Casp3}^*][\text{Casp8}]$
$d[\text{Casp8}^*]/dt = v_1 + v_2 - v_6 - \mu_{\text{Casp8}^*} [\text{Casp8}^*]$	$v_3 = k_3 [\text{Casp8}^*][\text{Casp3}]$
$d[\text{Casp3}]/dt = -v_3 - v_{13} + \alpha_{\text{Casp3}} - \mu_{\text{Casp3}} [\text{Casp3}]$	$v_4 = k_4 [\text{Casp3}^*][\text{XIAP}] - k_{-4} [\text{Casp3}^*\text{-XIAP}]$
$d[\text{Casp3}^*]/dt = v_3 - v_4 + v_{13} - \mu_{\text{Casp3}^*} [\text{Casp3}^*]$	$v_5 = k_5 [\text{Casp3}^*][\text{XIAP}]$
$d[\text{XIAP}]/dt = -v_4 - v_5 - v_{10} - v_{14} + \alpha_{\text{XIAP}} - \mu_{\text{XIAP}} [\text{XIAP}]$	$v_6 = k_6 [\text{Casp8}^*][\text{BAR}] - k_{-6} [\text{Casp8}^*\text{-BAR}]$
$d[\text{Casp3}^*\text{-XIAP}]/dt = v_4 - \mu_{\text{Casp3}^*\text{-XIAP}} [\text{Casp3}^*\text{-XIAP}]$	$v_7 = k_7 [\text{Casp8}^*][\text{Bid}]$
$d[\text{BAR}]/dt = -v_6 + \alpha_{\text{BAR}} - \mu_{\text{BAR}} [\text{BAR}]$	$v_8 = k_8 [\text{tBid-Bax}_2][\text{Cytc}]$
$d[\text{Casp8}^*\text{-BAR}]/dt = v_6 - \mu_{\text{Casp8}^*\text{-BAR}} [\text{Casp8}^*\text{-BAR}]$	$v_9 = k_9 [\text{tBid-Bax}_2][\text{Smac}]$
$d[\text{Bid}]/dt = -v_7 + \alpha_{\text{Bid}} - \mu_{\text{Bid}} [\text{Bid}]$	$v_{10} = k_{10} [\text{Smac}^*][\text{XIAP}] - k_{-10} [\text{Smac}^*\text{-XIAP}]$
$d[\text{tBid}]/dt = v_7 - \mu_{\text{tBid}} [\text{tBid}]$	$v_{11} = k_{11} [\text{Apop}][\text{Casp9}]$
$d[\text{tBid-Bax}_2]/dt = \mu_{\text{tBid-Bax}_2}$	$v_{12} = k_{12} [\text{Casp3}^*][\text{Casp9}]$
$(f_{\text{tBid-Bax}_2}([\text{tBid}], [\text{Bax}]_0; K_{\text{tBid-Bax}_2}) - [\text{tBid-Bax}_2])$	$v_{13} = k_{13} [\text{Casp9}^*][\text{Casp3}]$
$d[\text{Cytc}]/dt = -v_8 + \alpha_{\text{Cytc}} - \mu_{\text{Cytc}} [\text{Cytc}]$	$v_{14} = k_{14} [\text{Casp9}^*][\text{XIAP}] - k_{-14} [\text{Casp9}^*\text{-XIAP}]$
$d[\text{Cytc}^*]/dt = v_8 - \mu_{\text{Cytc}^*} [\text{Cytc}^*]$	
$d[\text{Smac}]/dt = -v_9 + \alpha_{\text{Smac}} - \mu_{\text{Smac}} [\text{Smac}]$	
$d[\text{Smac}^*]/dt = v_9 - v_{10} - \mu_{\text{Smac}^*} [\text{Smac}^*]$	
$d[\text{Smac}^*\text{-XIAP}]/dt = v_{10} - \mu_{\text{Smac}^*\text{-XIAP}} [\text{Smac}^*\text{-XIAP}]$	
$d[\text{Apop}]/dt = \mu_{\text{Apop}}(f_{\text{Apop}}([\text{Cytc}^*]/[\text{Apaf}]_0; \lambda_{\text{Apop}}) - [\text{Apop}])$	
$d[\text{Casp9}]/dt = -v_{11} - v_{12} + \alpha_{\text{Casp9}} - \mu_{\text{Casp9}} [\text{Casp9}]$	
$d[\text{Casp9}^*]/dt = v_{11} + v_{12} - v_{14} - \mu_{\text{Casp9}^*} [\text{Casp9}^*]$	
$d[\text{Casp9}^*\text{-XIAP}] = v_{14} - \mu_{\text{Casp9}^*\text{-XIAP}} [\text{Casp9}^*\text{-XIAP}]$	

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Model parameters

- Reaction rates taken from appropriate model in the literature
- Rates appearing in more than one model are consistent
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Species	Initial concentration (nM) HeLa	Jurkat T	Parameter	Value
Casp8	216.67 [62]	33.33 [72, 73]	$[\mathrm{FasL}]_0$	2 nM [72, 73]
Casp3	35 [62]	200 [72, 73]	$[\mathrm{FasR}]_0$	10 nM [72,73]
XIAP	66.67 [62]	30 [72,73]	K_{DISC}	1.032 nM [72,73]
BAR	66.67 [62]		$[\mathrm{Bax}]_0$	83.33 nM [72,73]
Bid	25 [72, 73]		$K_{\mathrm{tBid\text{-}Bax_2}}$	100 nM [72, 73]
Cytc	100 [72, 73]		$[\mathrm{Apaf}]_0$	100 nM [72, 73]
Smac	100 [72, 73]		λ_{Apop}	1 [70]
Casp9	20 [72, 73]			

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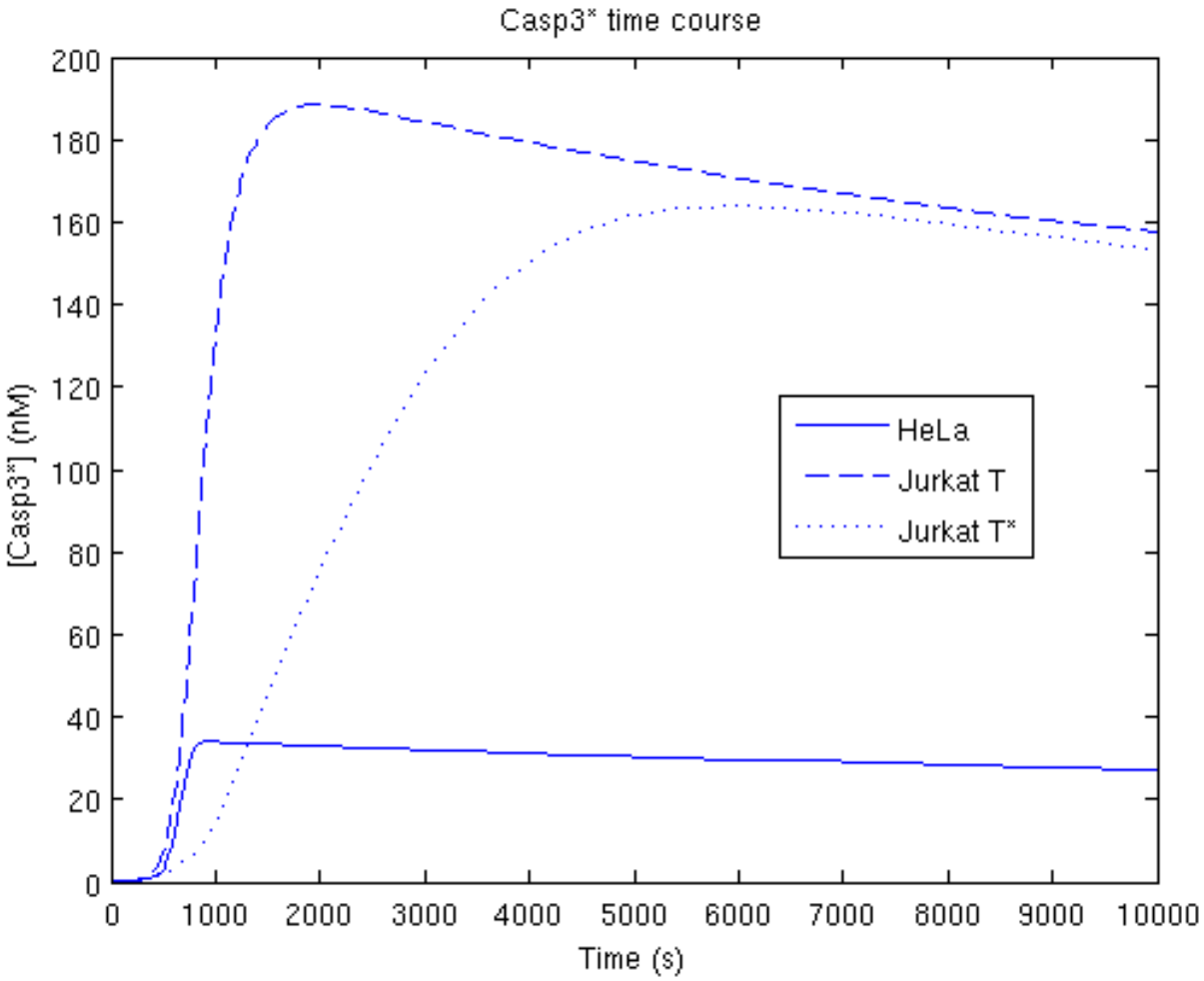
Casp9	20 [72, 73]			

- Philosophy: assume parameters are correct and study model solutions

Analysis and results

Characteristic time course



Analysis and results

Regression analysis and model reduction

- Define quantitative descriptors of caspase activation
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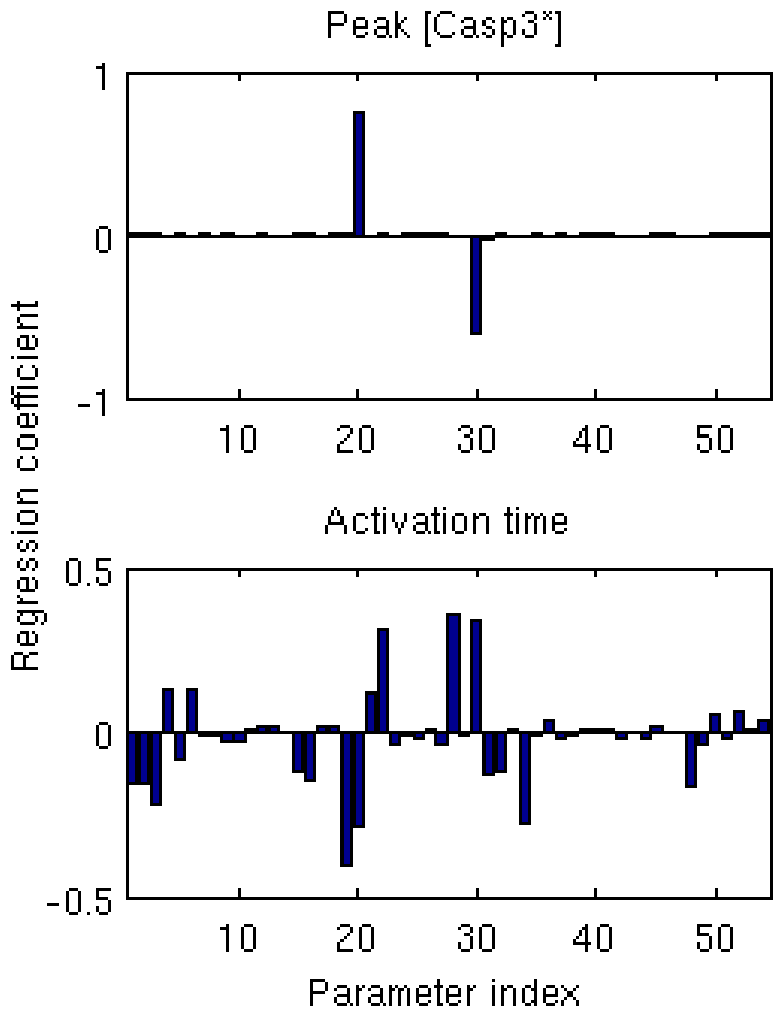
- Define quantitative descriptors of caspase activation
  1. Peak activation: maximum [Casp3*] over time course
  2. Activation time: time at which this peak is achieved
- Sensitivity analysis
  - Generate locally perturbed parameters around baseline values
  - Simulate to collect synthetic data
  - Multiple linear regression on standardized data:

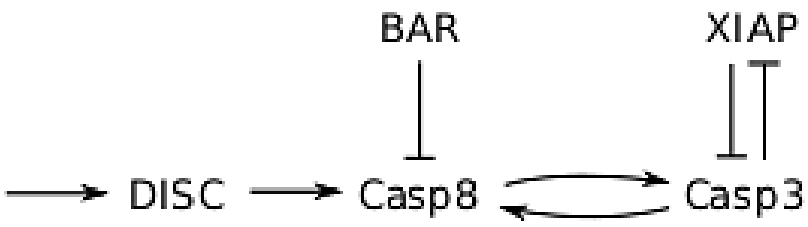
$$\mathbf{Y} = \begin{pmatrix} \mathbf{1} & \mathbf{X} \end{pmatrix} \mathbf{b}$$

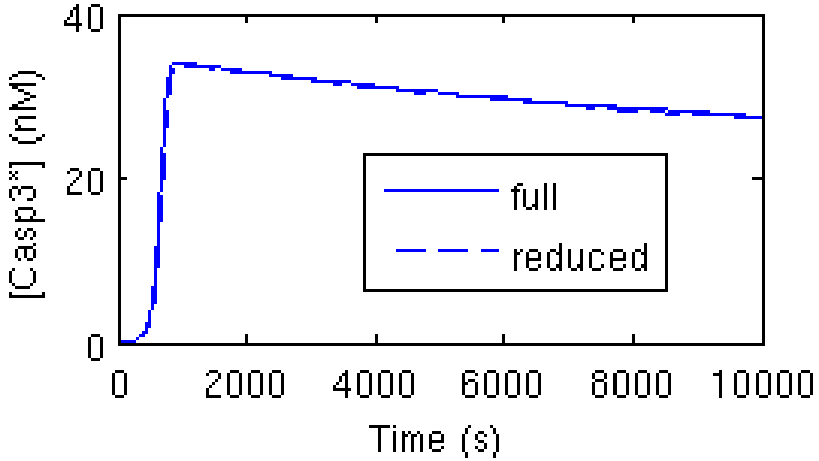
  - Model reduction based on identified key parameters

Analysis and results

Regression analysis and model reduction: HeLa induced by FasL

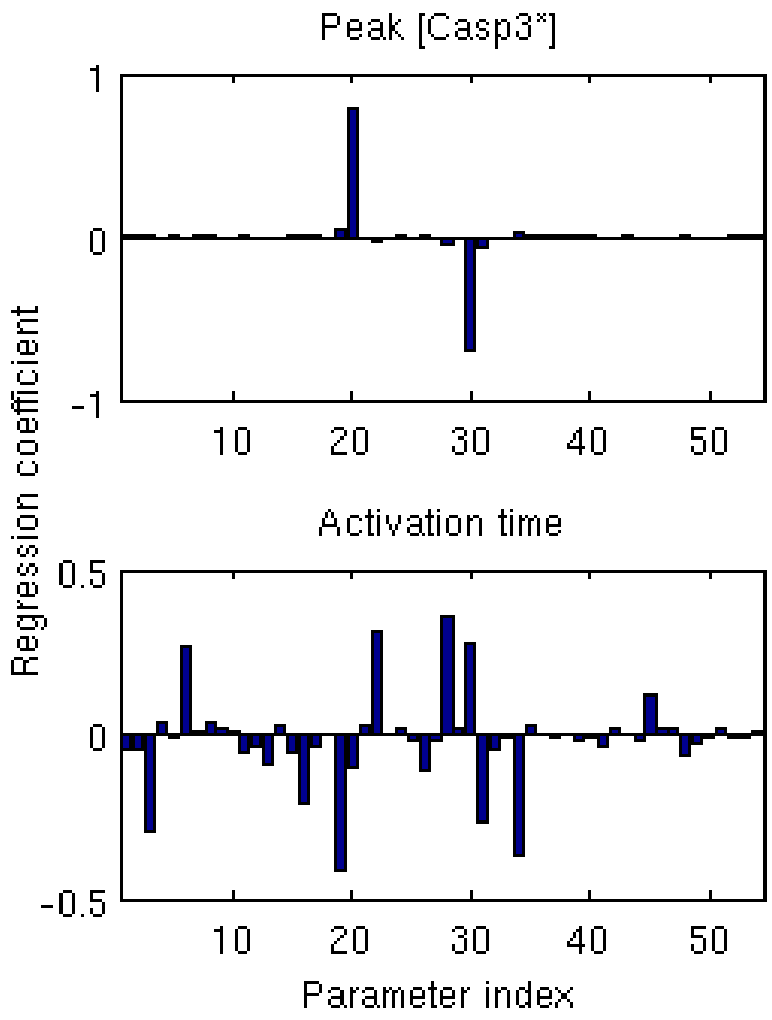


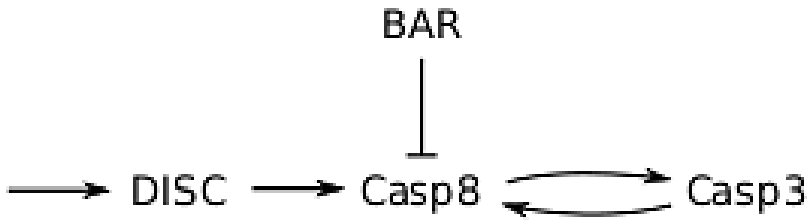


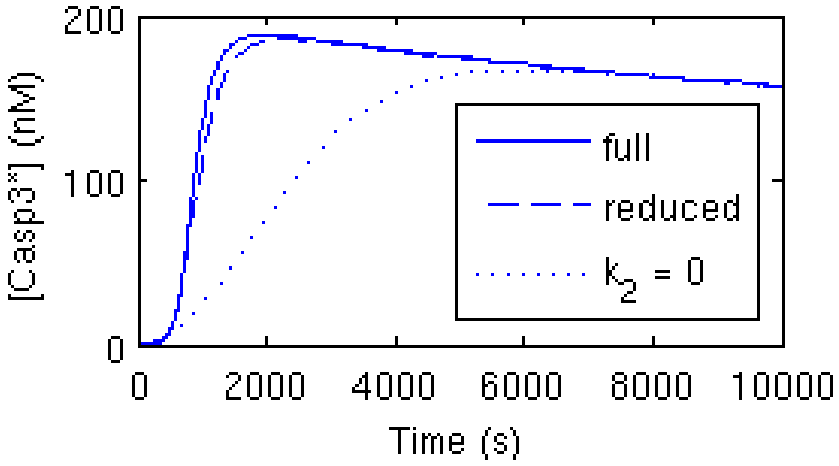


Analysis and results

Regression analysis and model reduction: Jurkat T induced by FasL







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Regression analysis and model reduction: Jurkat T* induced by low FasL

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Analysis and results

Regression analysis and model reduction: Jurkat T* induced by low FasL

- Interestingly, both baseline HeLa and Jurkat T parameters exhibit type I behavior
- Define new cell parameters Jurkat T* by turning off the reactions

$$\text{Casp3}^* + \text{Casp8} \longrightarrow \text{Casp3}^* + \text{Casp8}^*, \tag{2}$$

$$\text{Casp3}^* + \text{XIAP} \longrightarrow \text{Casp3}^*, \tag{5}$$

$$\text{Casp3}^* + \text{Casp9} \longrightarrow \text{Casp3}^* + \text{Casp9}^*, \tag{12}$$

i.e., $k_2 = k_5 = k_{12} = 0$

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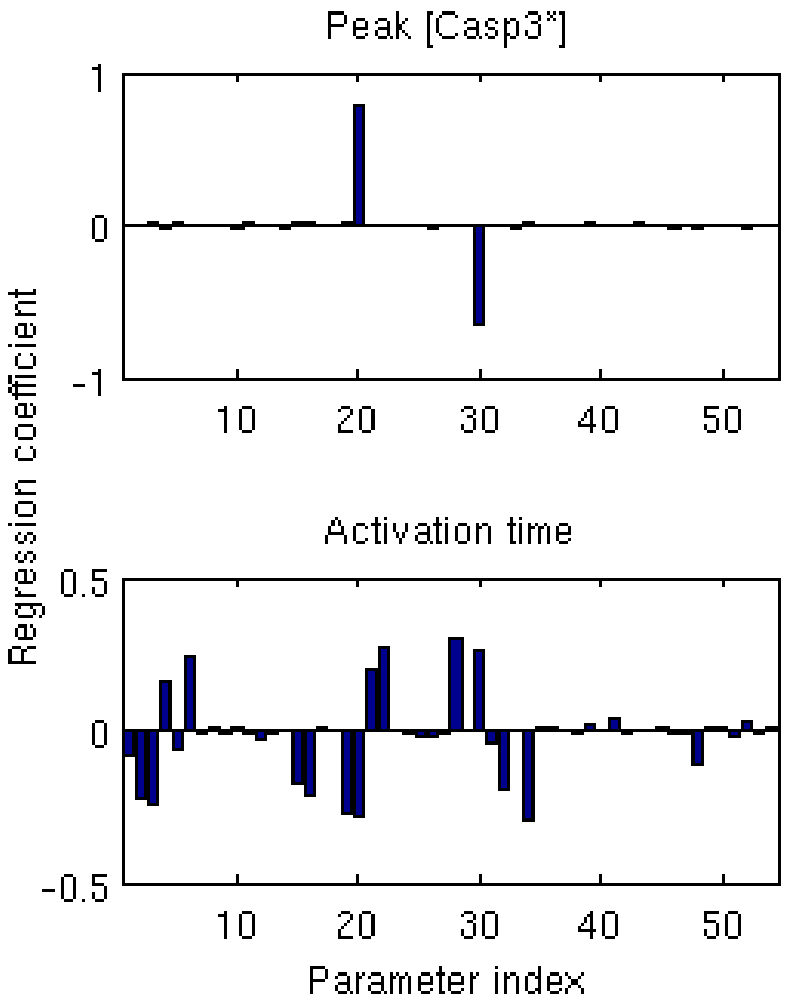

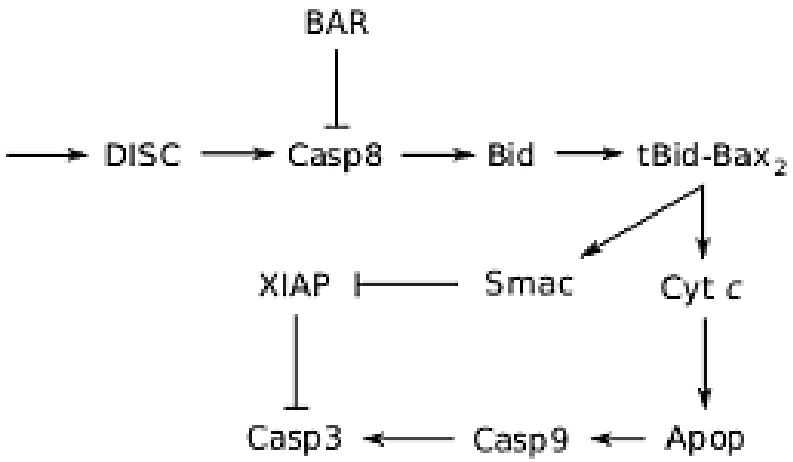

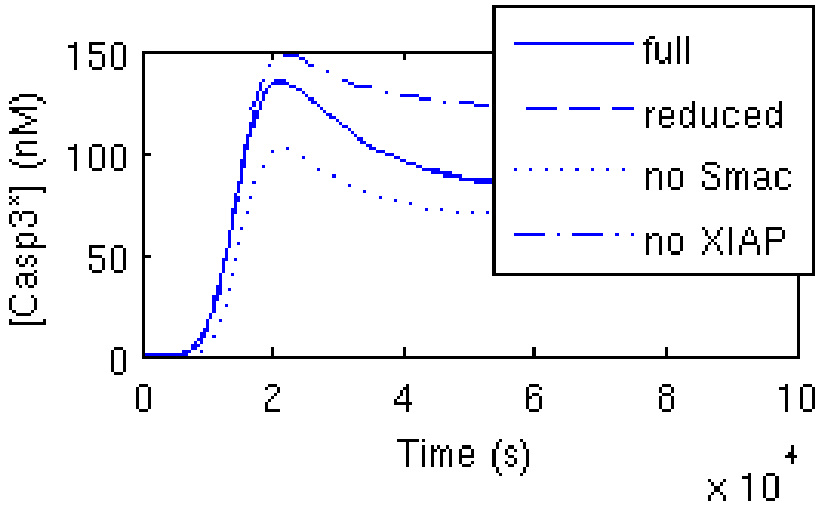

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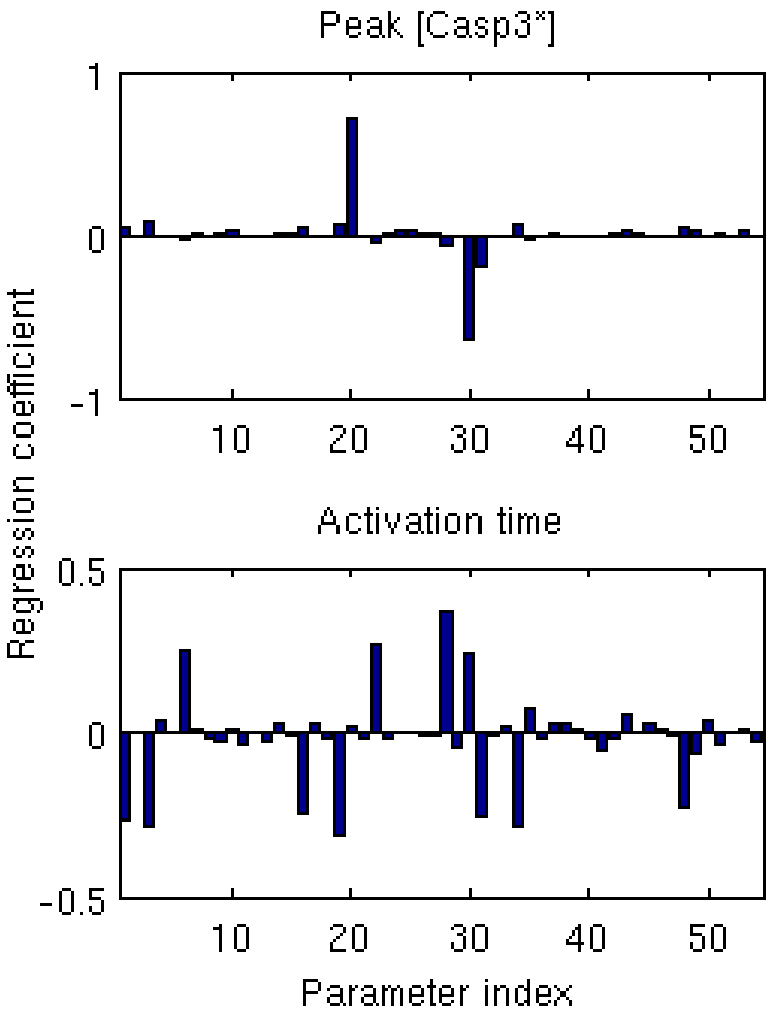
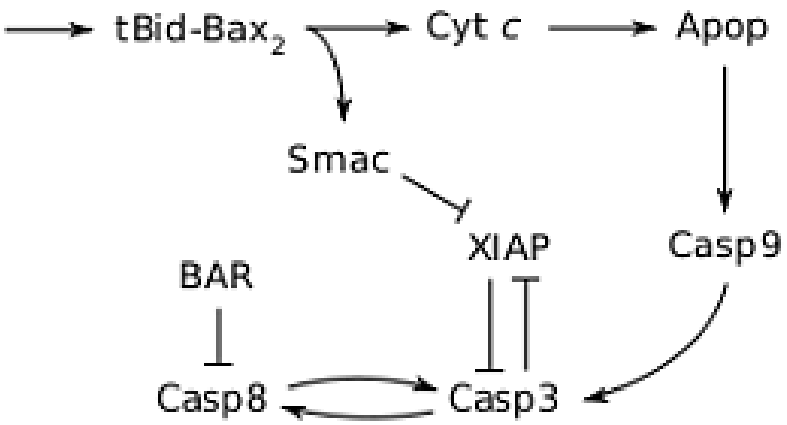
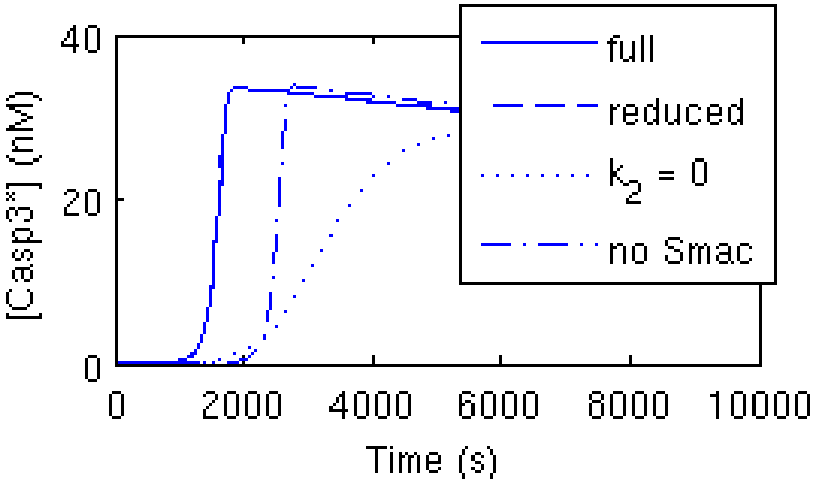
- Can also consider mitochondrial apoptosis through the intrinsic pathway
- Cell stress, DNA damage, cytotoxicity causes mitochondria permeabilization and release of Cyt*c* and Smac
- Functionally represent with input $[\text{tBid}]_0 > 0$

Analysis and results

Regression analysis and model reduction: HeLa induced by tBid







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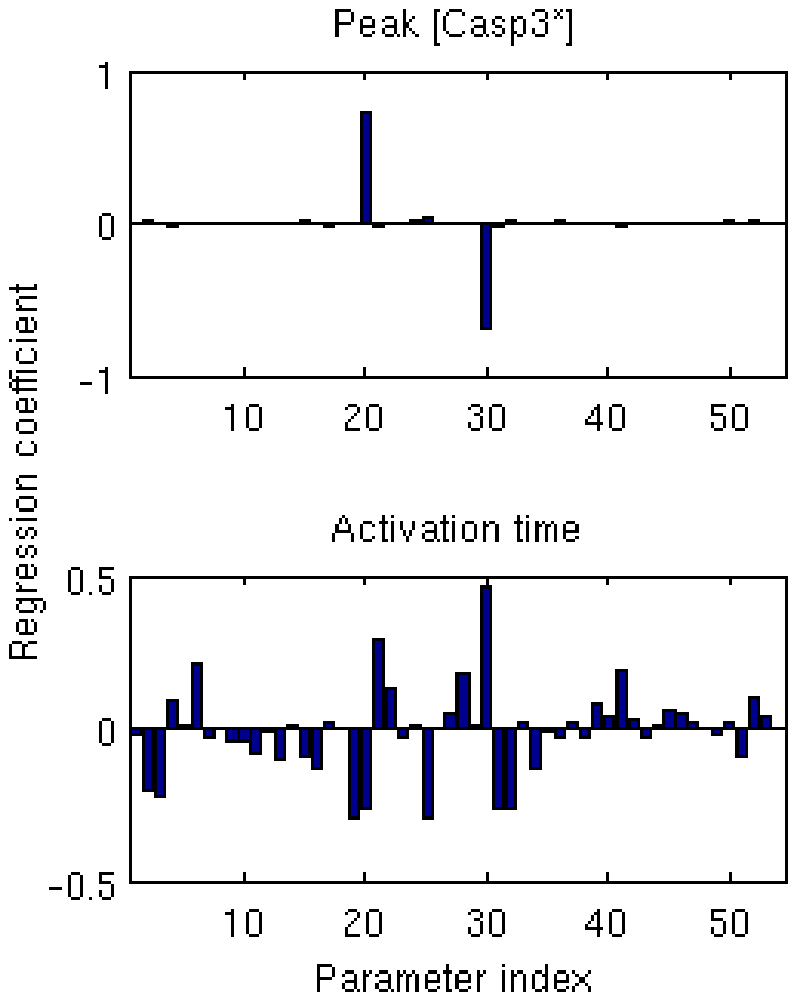
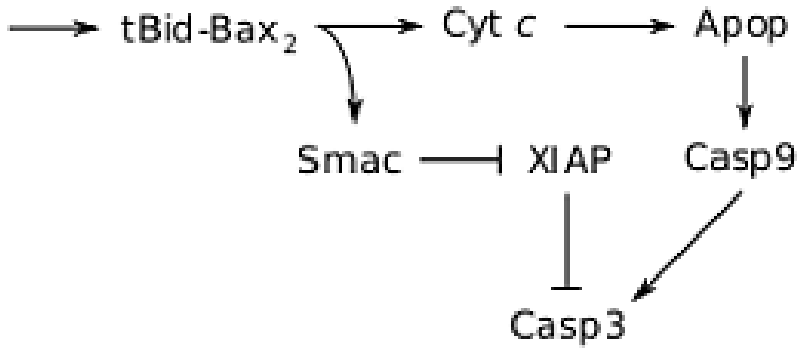
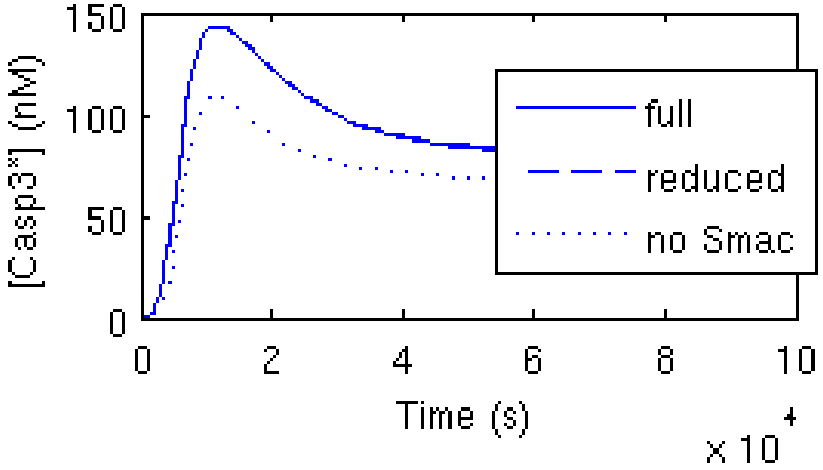
Regression analysis and model reduction: Jurkat T* induced by tBid







Analysis and results

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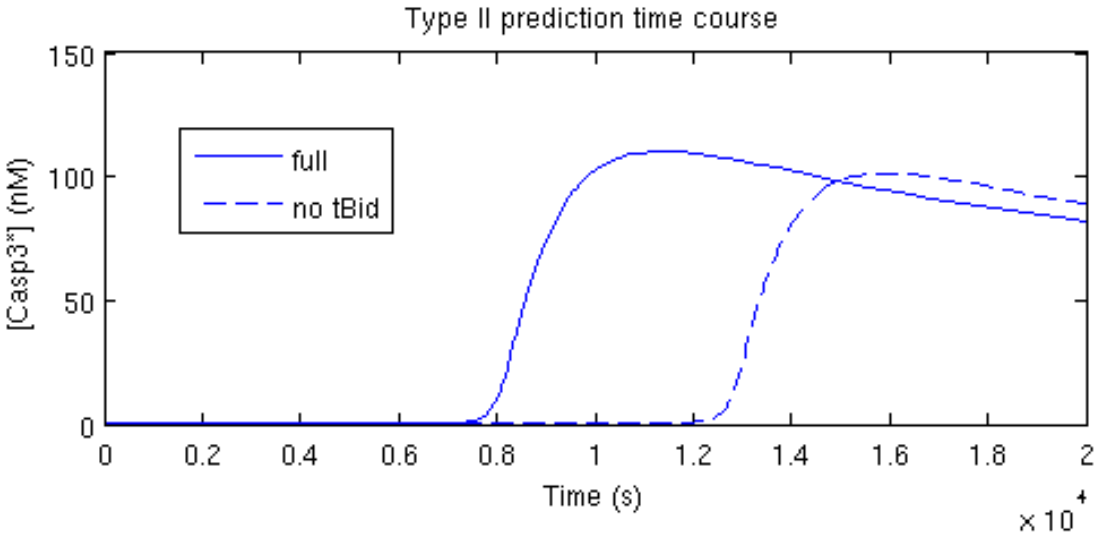
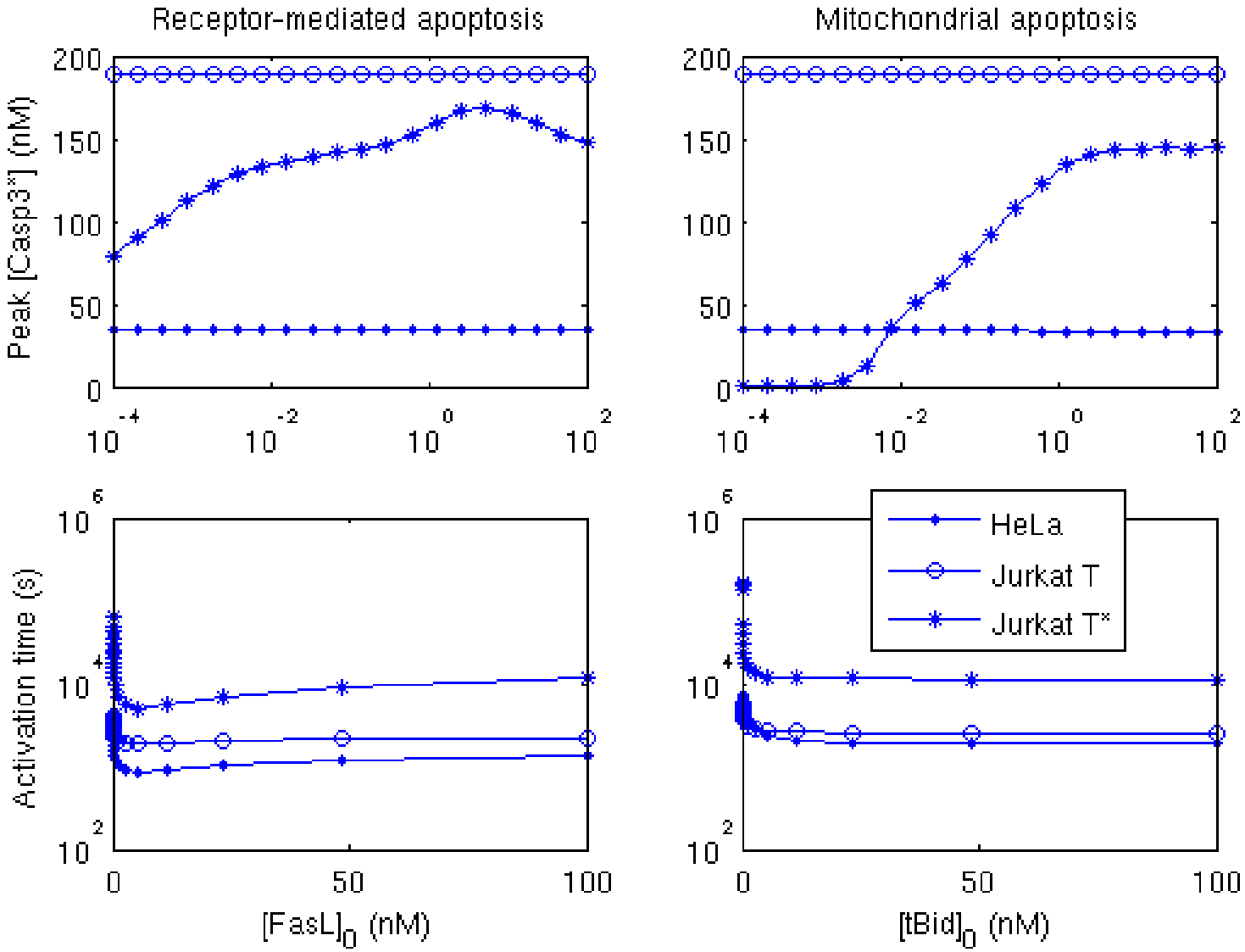


Analysis and results

Activation thresholds



Conclusion

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- Remarked on stability and caspase activation thresholds

Conclusion
Future directions

Methodology

Biology

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- Study transition of apoptotic behavior over cell parameters
- Include regulators or delays for potential model stability: bistability and the point of no return

Conclusion

Acknowledgments

- Heather Harrington (Imperial)
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- Profs. Baltazar Aguda and Chiu-Yen Kao (OSU)
- Prof. Jaroslav Stark (Imperial)
- MBI Summer School at OSU

Conclusion

References

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