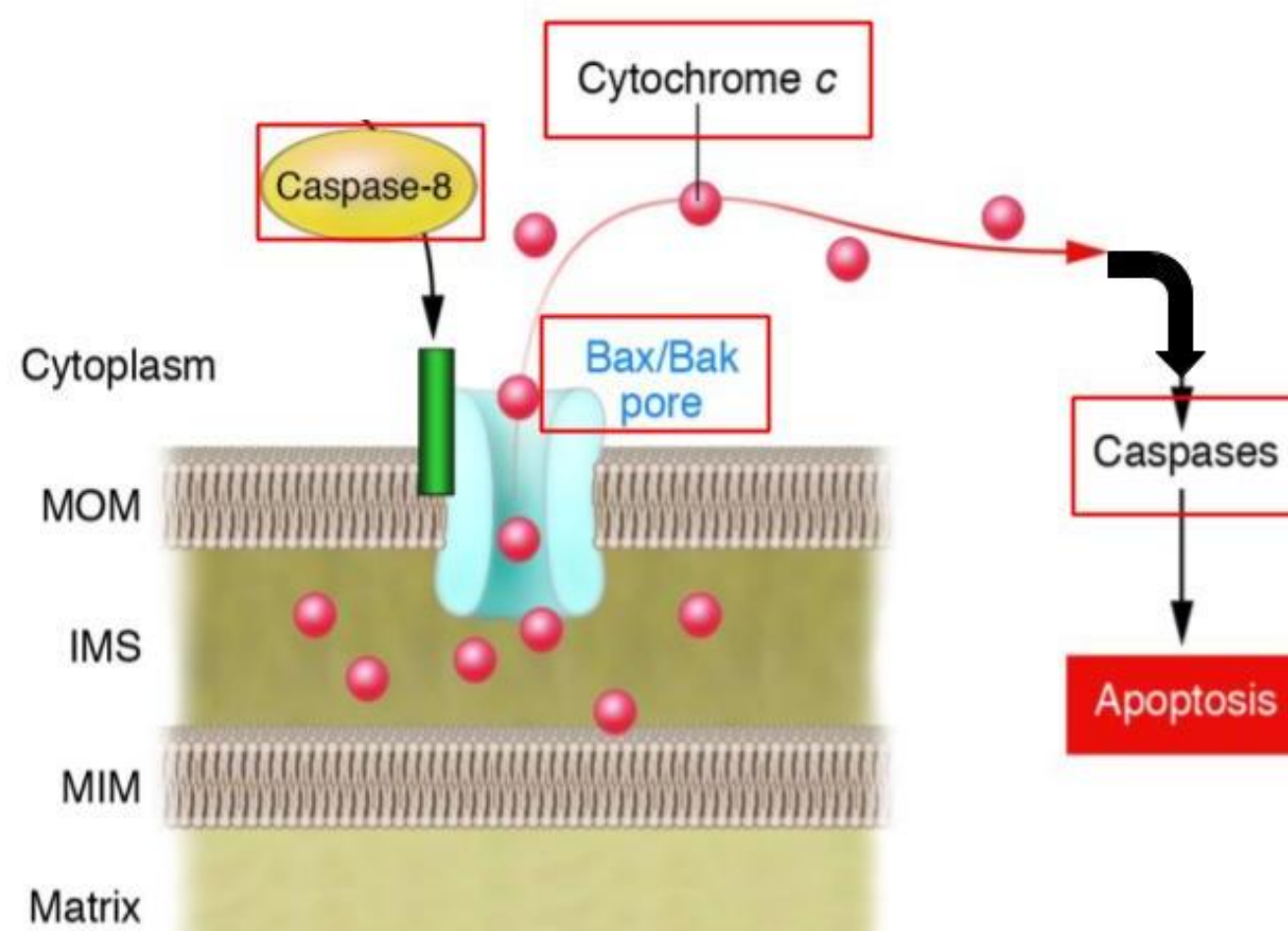


## Background

- Cell death caused by myocardial infarction leads to Heart Disease which is the leading cause of death in US
- Identifying a gene's role in cell death and its mechanism will lead to new treatment options
- 3 possible mechanisms for death through mitochondria

## Apoptosis

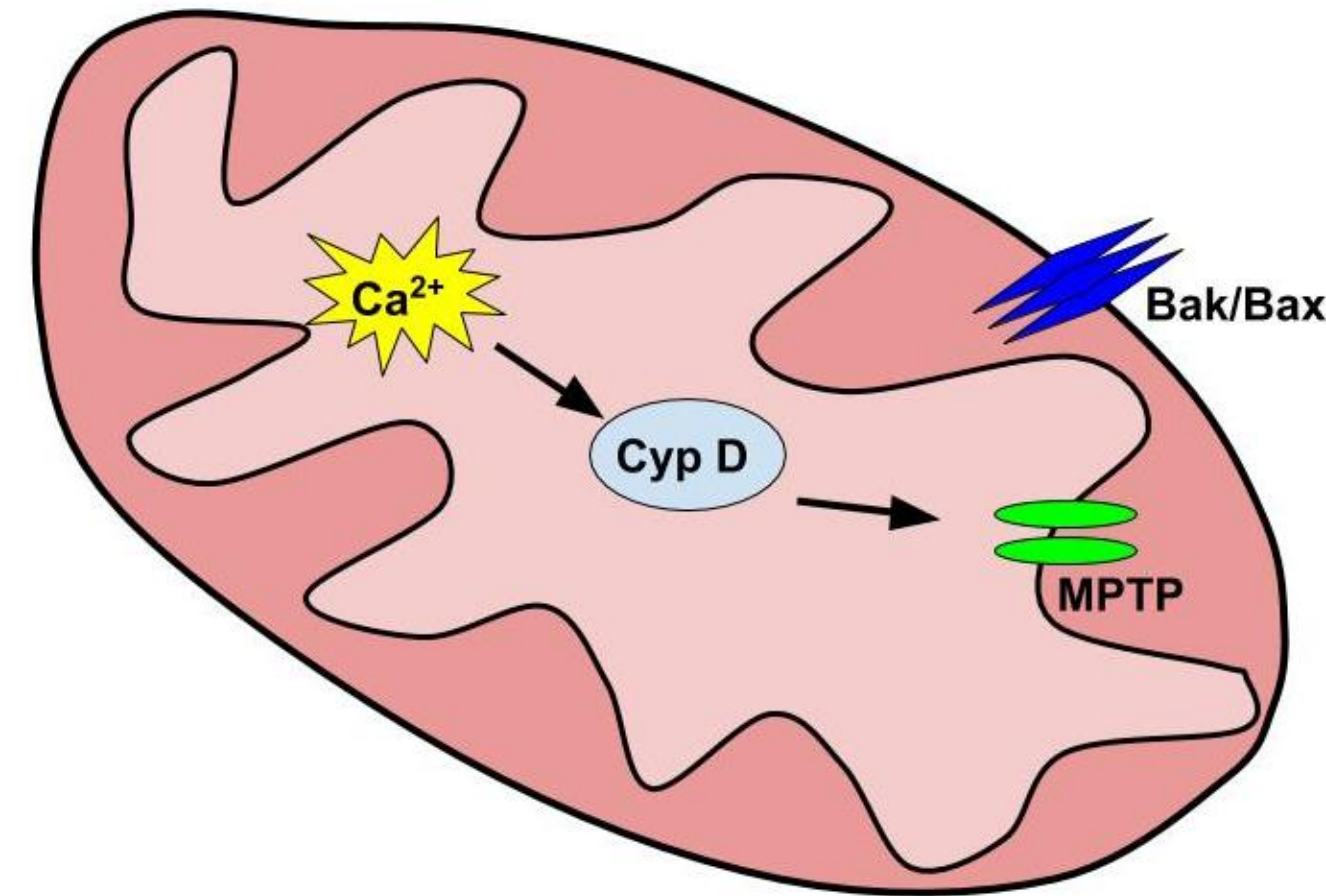


**Fig. 1.** The releasing of Cytochrome c through the Bax/Bak pore triggers the organized caspase cascade and results in efficient cell death. MOM is the mitochondrial outer membrane, IMS is the intermembrane space, and MIM is the mitochondrial inner membrane. Apoptosis is scene in regulatory and developmental cell death and is a normal, healthy process.

## Literature Cited

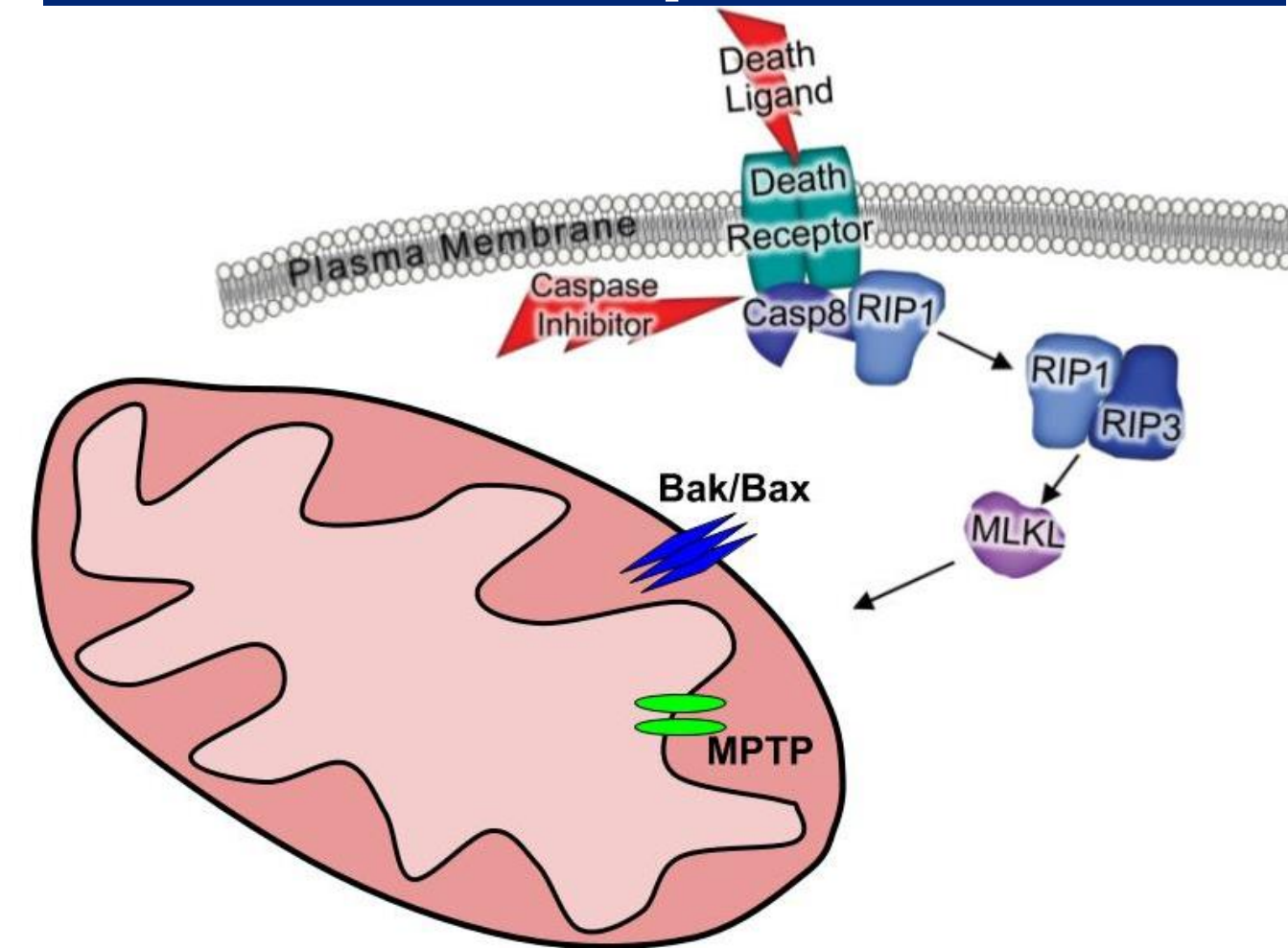
- <sup>1</sup>Karch, J., and J.D. Molkenin. 2015. Regulated Necrotic Cell Death. *Circulation Research* 116: 1800–1809.
- <sup>2</sup>Teng, Y., X. Ren, H. Li, A. Shull, J.Kim, and J.K. Cowell. 2016. Mitochondrial ATAD3A combines with GRP78 to regulate the WASF3 metastasis-promoting protein. *Oncogene* 35: 333-343.
- <sup>2</sup>Xie, H., J. Hu, H. Pan, Y. Lou, P. Lv, and Y. Chen. 2014. Adenovirus vector-mediated FAM176A overexpression induces cell death in human H1299 non-small lung cancer cells. *BMB Reports* 47: 104-109

## Necrosis



**Fig. 2.** Calcium overload in mitochondria results from high stress events like ischemia. This sequence of events results in mitochondrial dysfunction and adverse cell death. MPTP is the mitochondrial permeability transition pore and Cyp D represents activated cyclophilin D. Necrosis is a harmful form of cell death with negative effects on the surrounding areas.

## Necroptosis

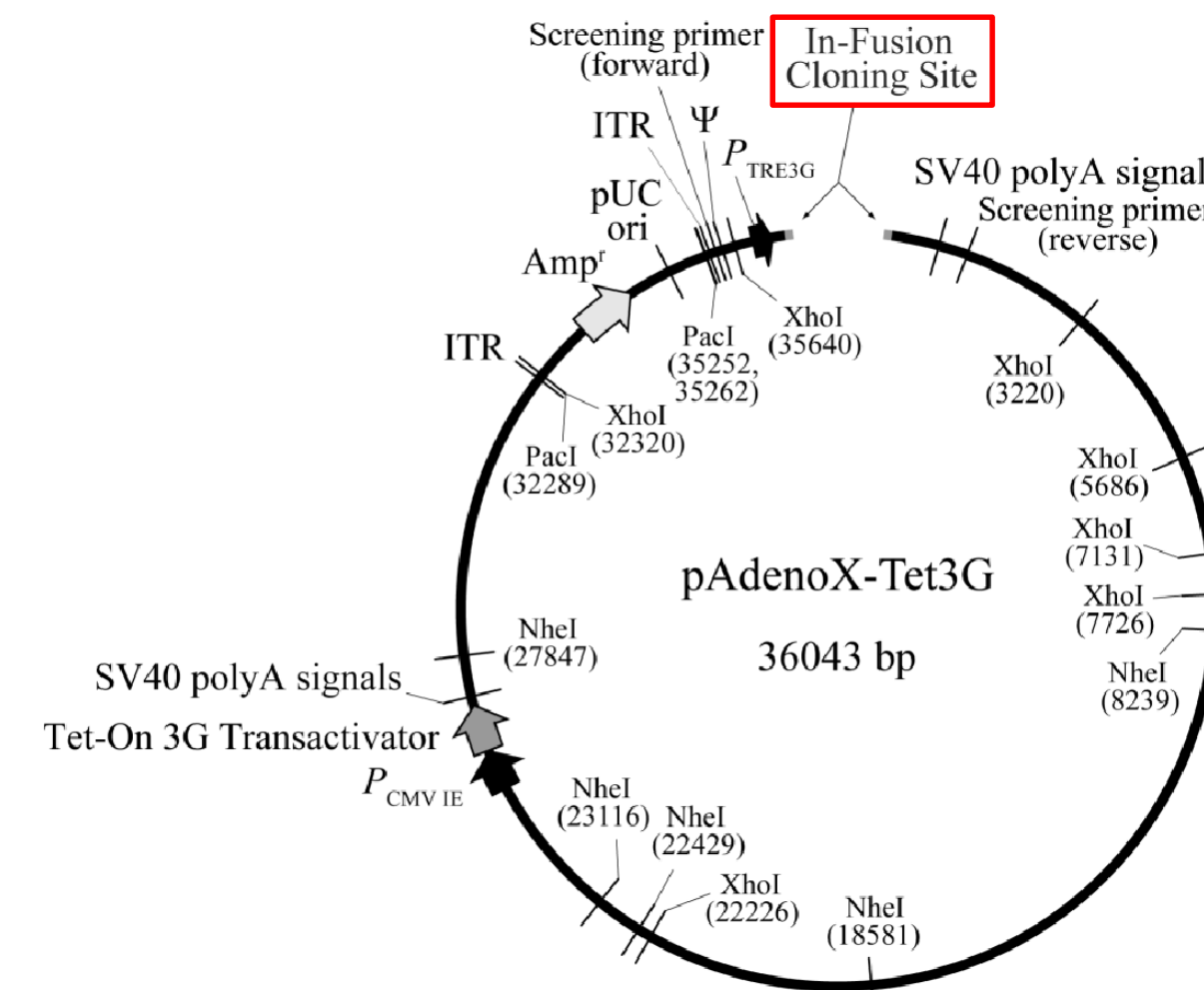


**Fig. 3.** Representation of necroptosis as modified from image from Karch et al. (2015). This pathway is triggered by inhibition of the Caspase 8 molecule which is needed for apoptosis. The mitochondria then undergoes MPTP formation and necrosis ensues.<sup>1</sup> MLKL is the mixed lineage kinase like complex. The result, similar to necrosis, is harmful to surrounding tissues.

## Approach

- Screen for potential genes
- Amplify cDNA of genes
- Insert into Inducible Vector
- Recombinant Virus
- Apply to Cells
- Assay for Death

## Inducible Vector



**Fig. 4.** Inducible Vector used to regulate gene expression. Highlighted site represents where chosen gene is inserted. This vector was then inserted into an AdenoX viral vehicle.

## Results

	No Virus	Virus
Induced	Healthy	<b>Death</b>
Not Induced	Healthy	Healthy

**Table 1.** Expected results based on gene delivery via virus and the induction of the gene

	No Virus	Virus
Induced	Healthy	<b>Death</b>
Not Induced	Healthy	<b>Death</b>

**Table 2.** Observed Resulted based on gene delivery via virus and the induction of the gene. Note the unexpected death in the “Not Induced Virus” cell.

## Discussion

- Possible explanation for results is death caused by viral toxicity– not the gene
- Xie et al. (2014) outline methods of similar experiment with improved results
- ATAD3A overexpression cancer cells suggest anti-apoptotic characteristics<sup>2</sup>
- FAM176A triggers apoptotic cell death when overexpressed<sup>1</sup>

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