

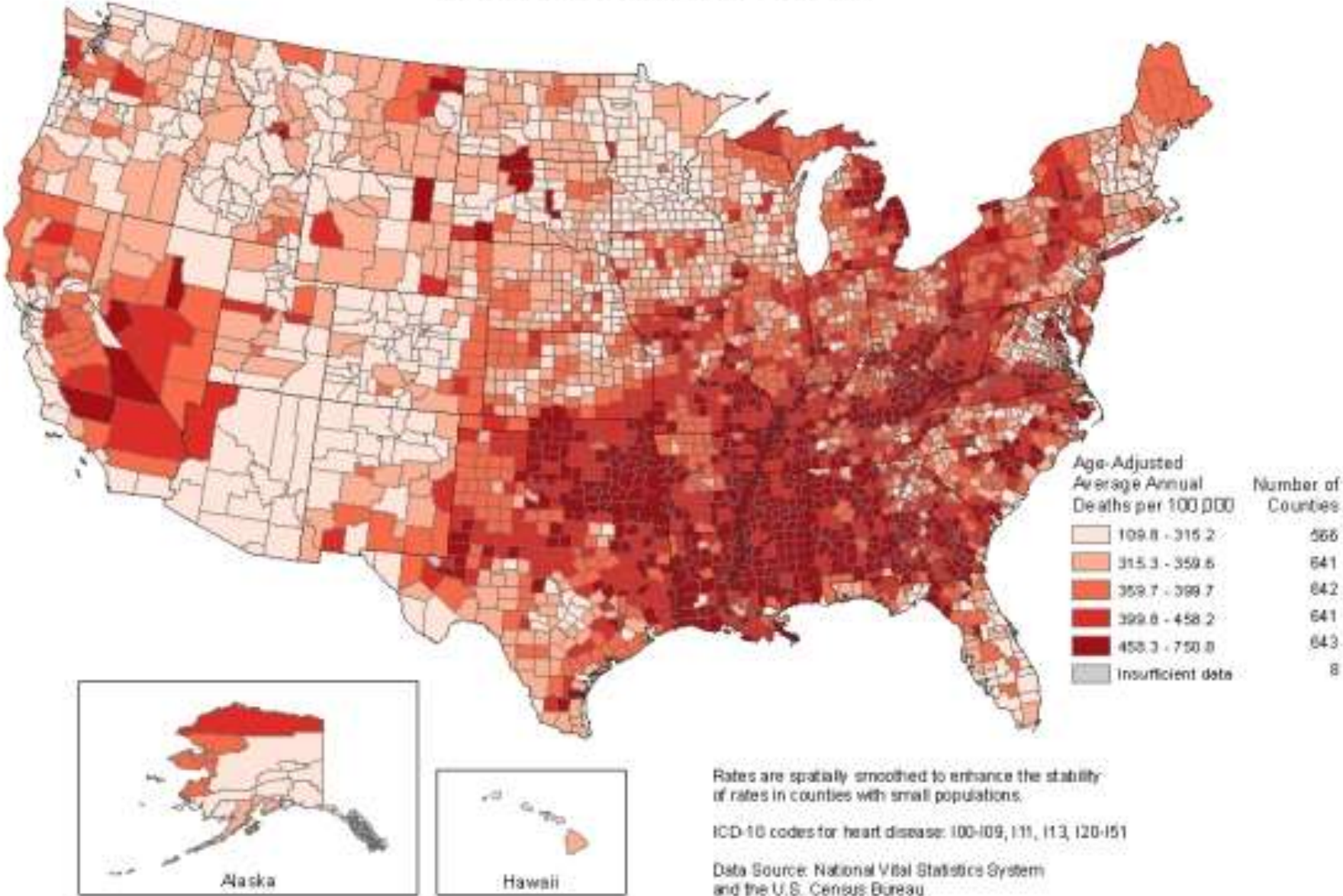
CATHEPSIN K INHIBITION
ALLEVIATES ANTIMYCIN-
INDUCED CARDIOMYOCYTE
APOPTOSIS

Dawn Anne Davison

Heart Disease

- “Heart Disease is the leading cause of death in the United States for the past 80 years”

Heart Disease Death Rates, 2007-2009 Adults Ages 35+, by County



Heart Disease Classifications

- Heart Failure
- Angina
- Coronary Artery Disease
- Cardiomyopathy
- Hypertension (high blood pressure)

Cardiac Cell Death

⦿ Problem

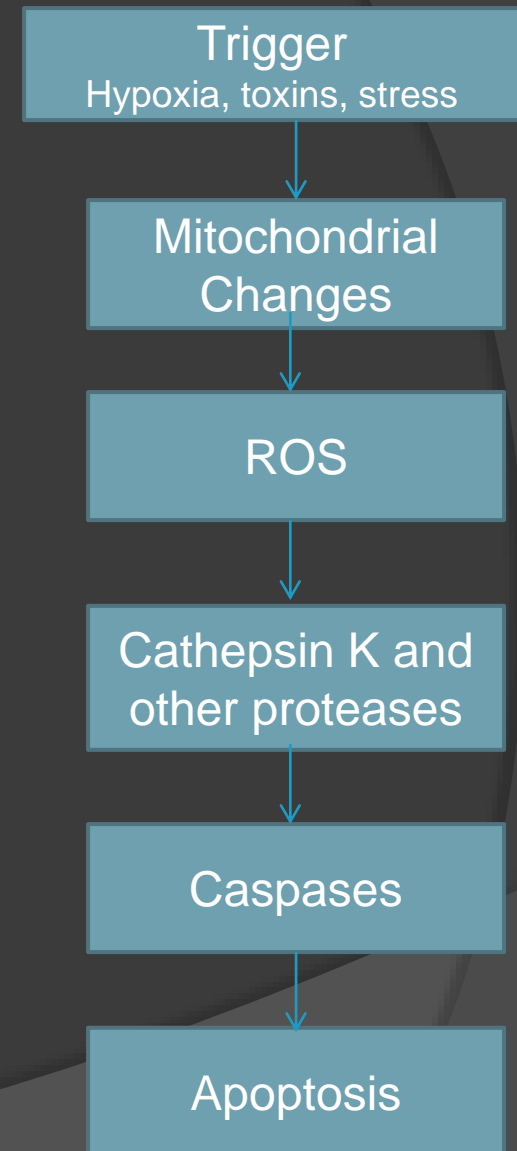
- Cardiomyocytes lack oxygen
 - Die by apoptosis

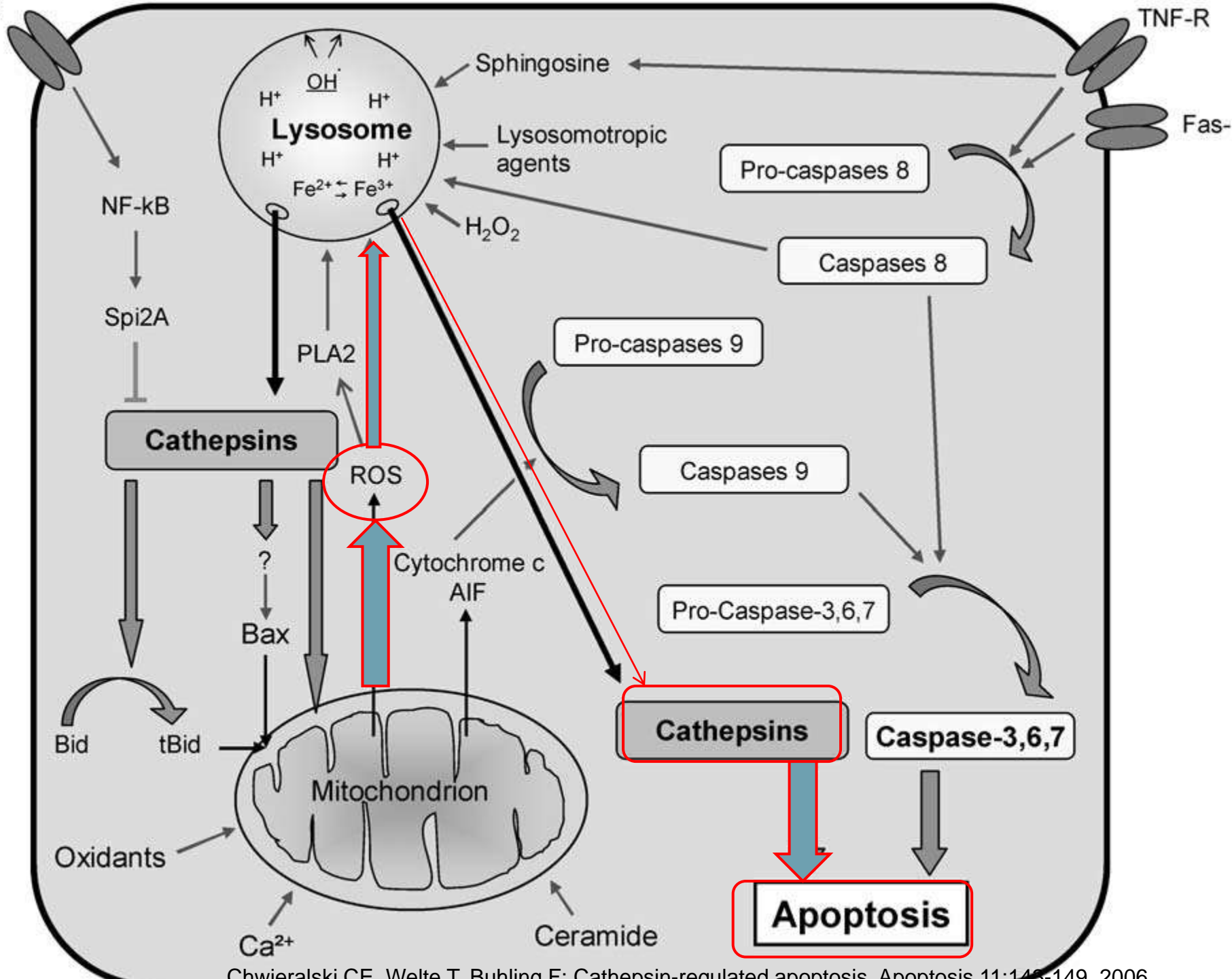
⦿ Result

- Irreversible fibrosis
- Inefficient contractile functions

Apoptosis

- ⦿ Programmed cell death
- ⦿ Mitochondrial Activation
 - Release of damaging Reactive Oxygen Species (ROS)
- ⦿ Apoptosis Markers
 - Bcl - 2
 - BAX
 - Cytochrome C





Cathepsin K (Cat K)

- A cysteine protease
- Released from lysosomes
- Potent collagenolytic activity
- Elevated in the dysfunctioning heart
- Cathepsin K knockout alleviates cardiac dysfunction
- Mechanism?

Hypothesis

- Cathepsin K *inhibition* prevents cardiomyocyte apoptosis

Methods

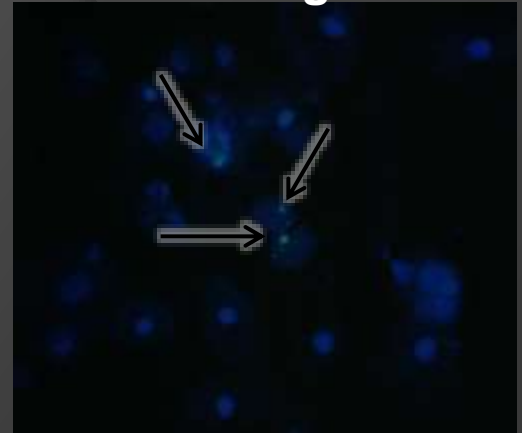
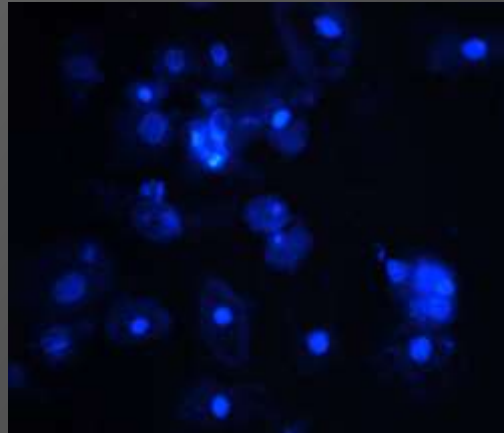
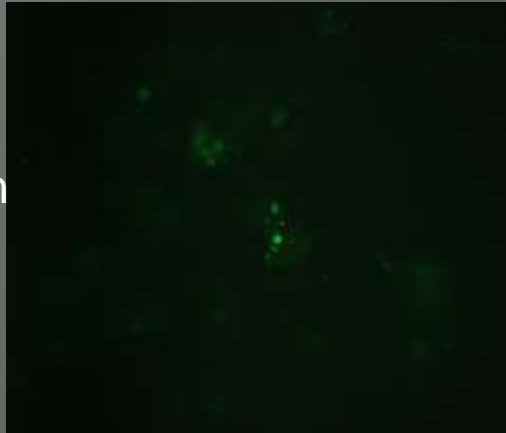
- H9c2 cardiomyocytes
- Antimycin A –inhibitor of respiratory chain in mitochondria
 - Generates reactive oxygen species
- Markers of apoptosis
 - Gel Electrophoresis and Western Blot
 - Primary antibodies against apoptosis markers
 - TUNEL staining
 - Lysosomes and Cat K dual staining (immunohistochemistry; confocal microscopy)

TUNEL

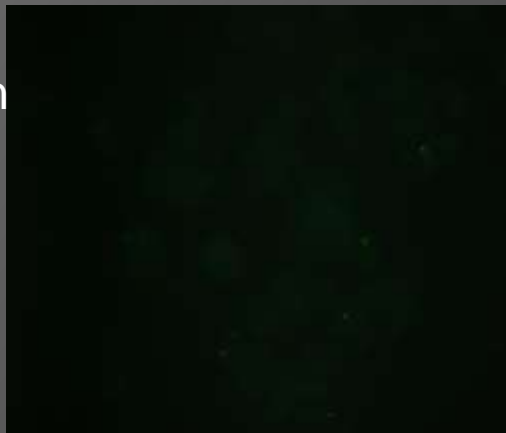
DAPI

Merge

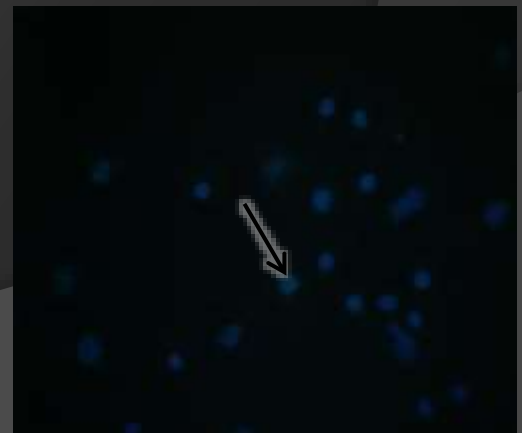
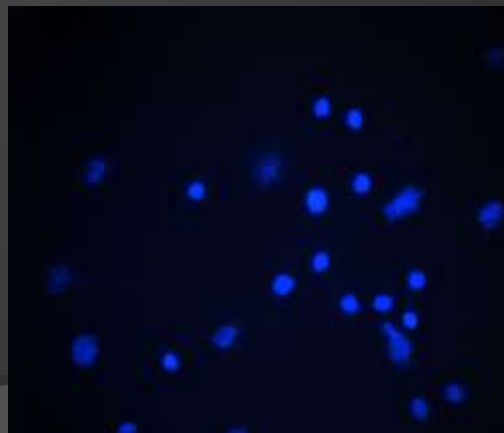
Antimycin

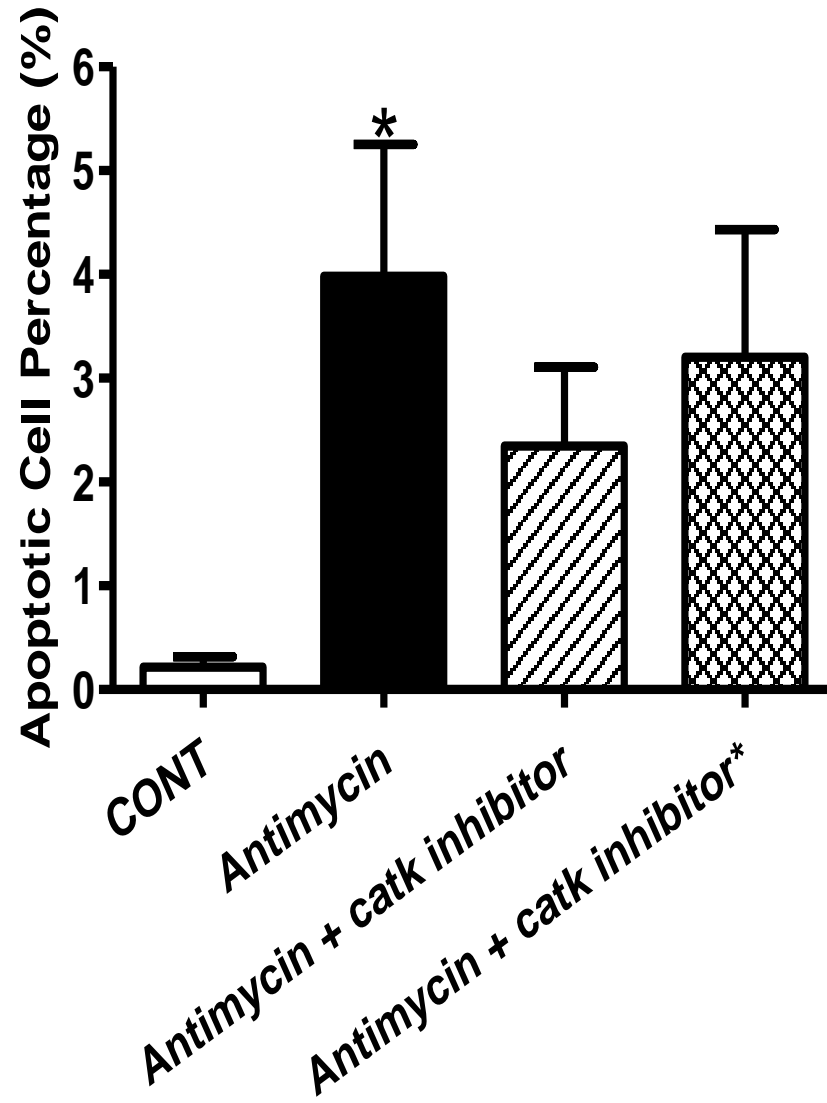


Antimycin
+ catk
inhibitor
10 nM



Antimycin
+ catk
inhibitor
100 nM



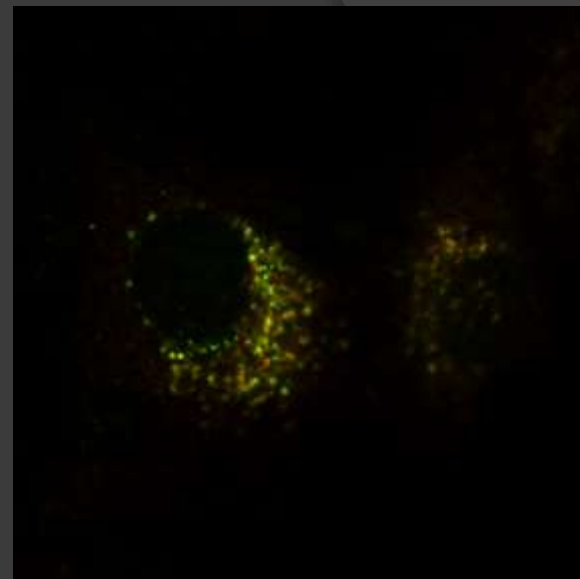
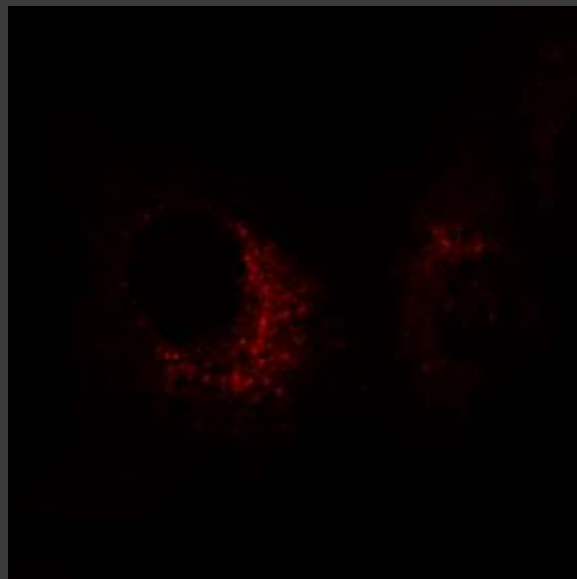
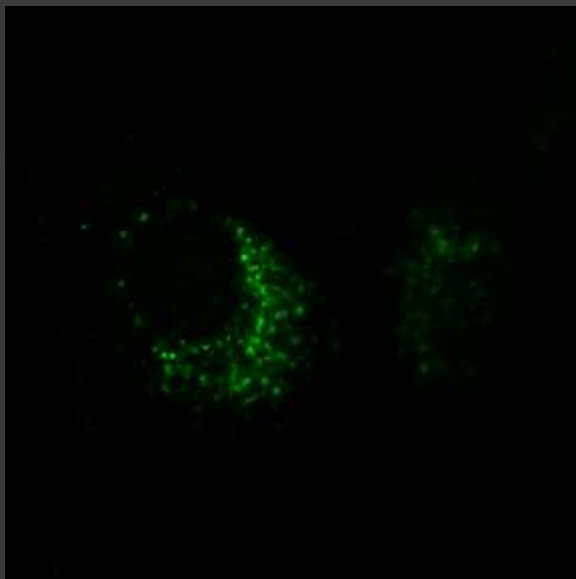


Lysosome

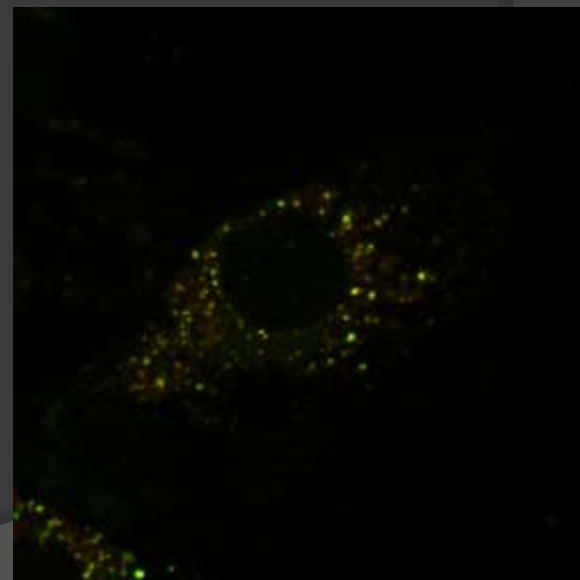
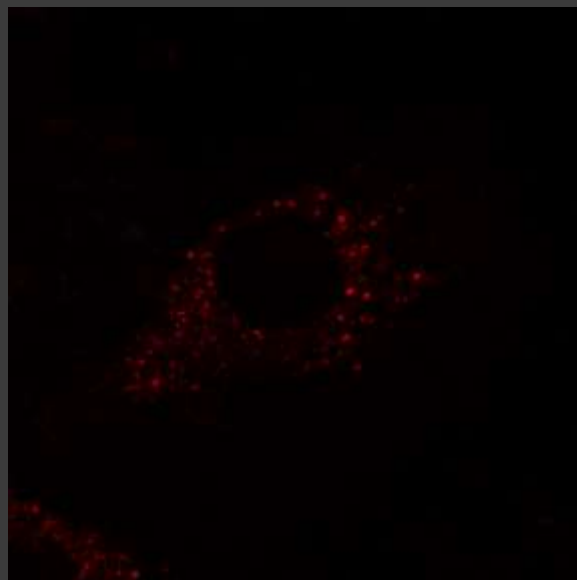
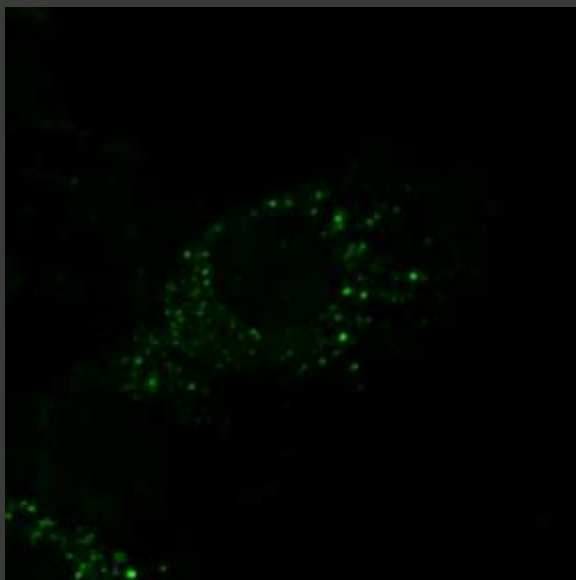
Cathepsin K

Merge

CONT



Antimycin 24 h

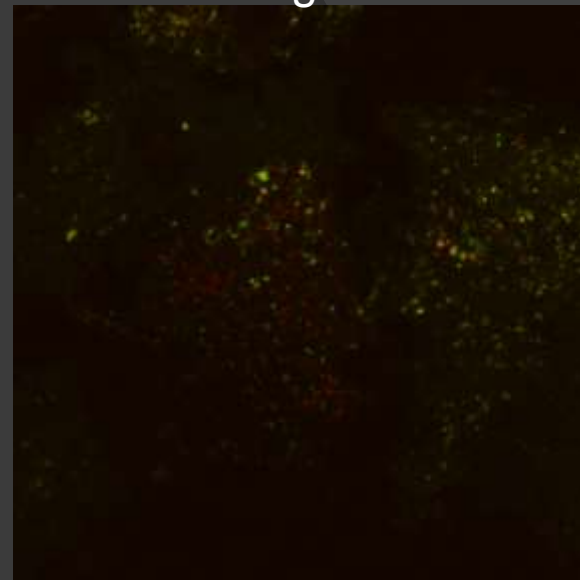
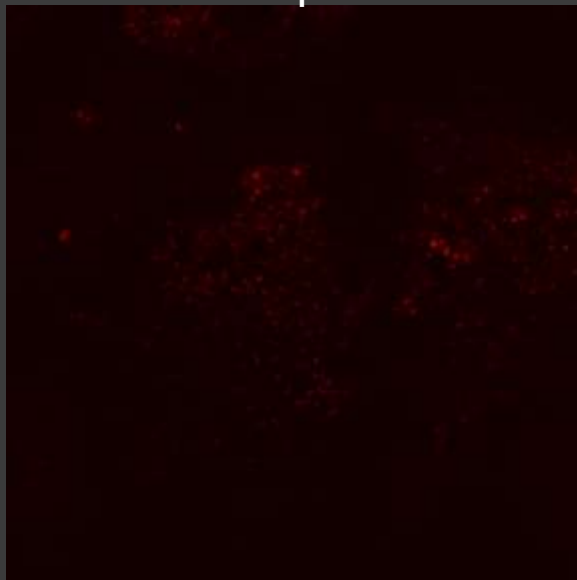
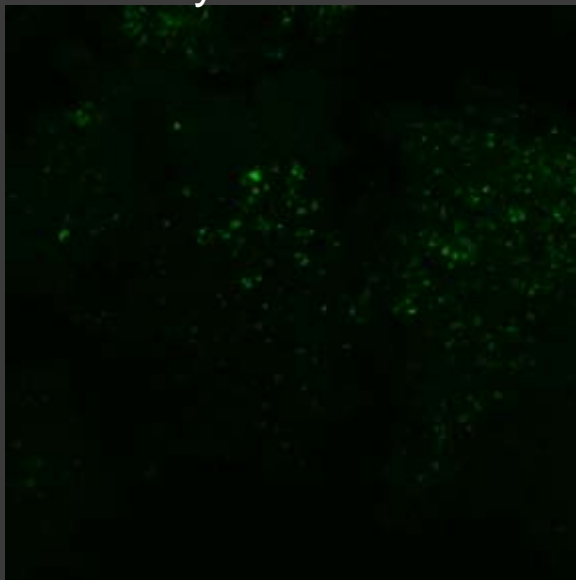


Lysosome

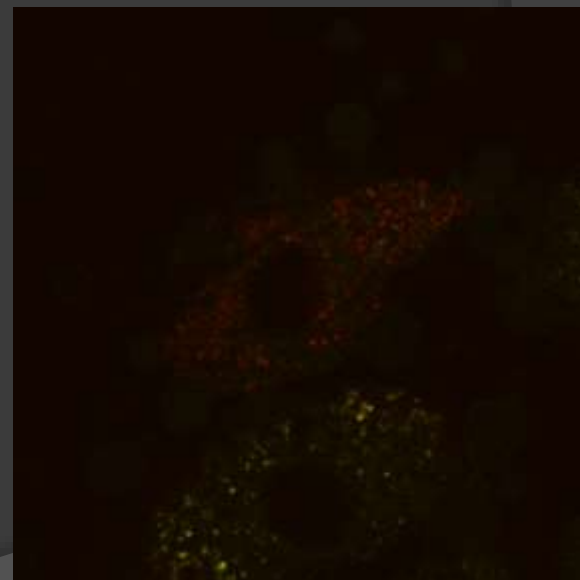
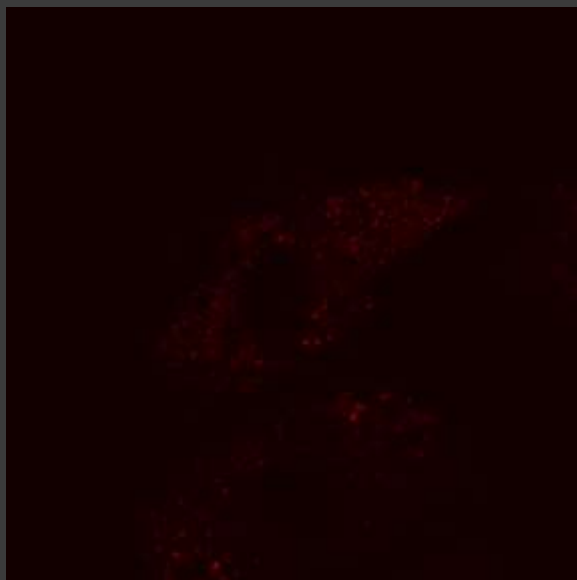
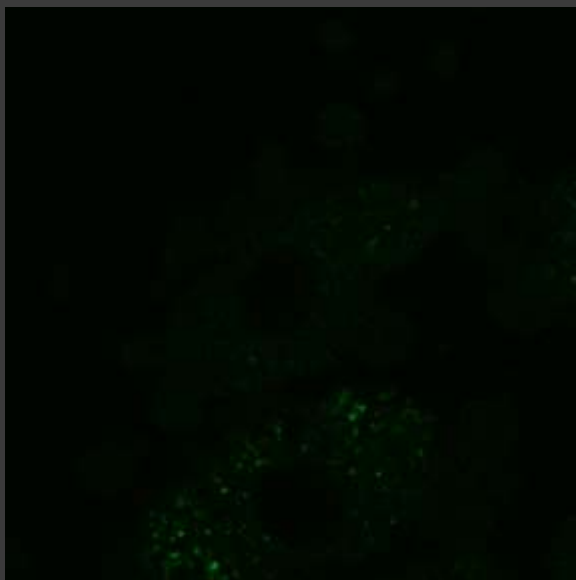
Cathepsin K

Merge

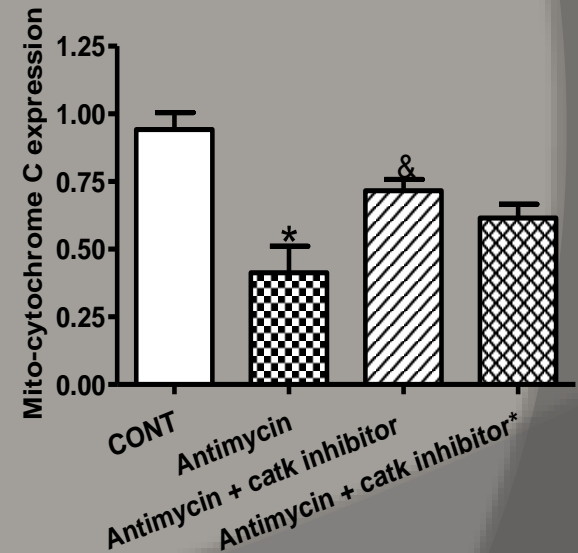
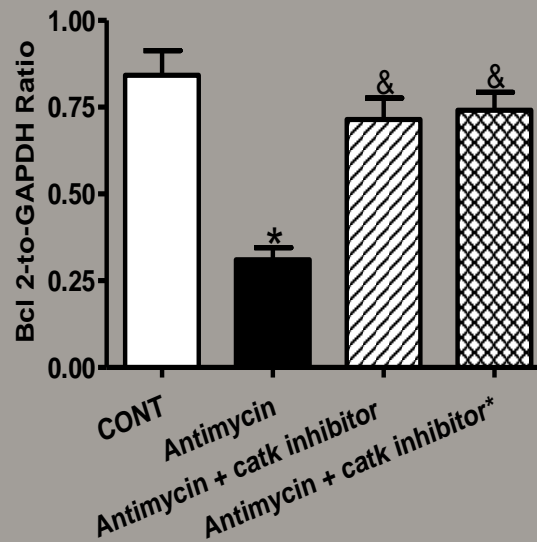
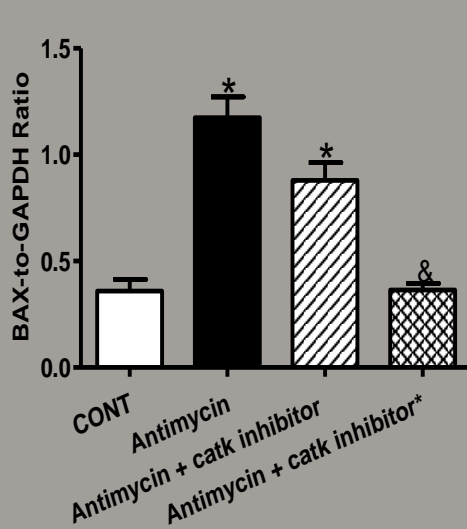
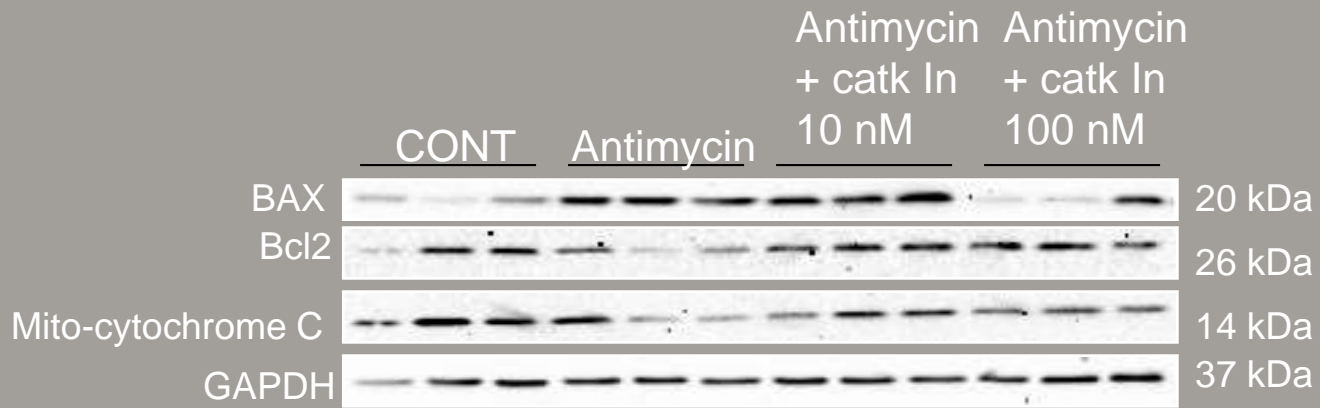
Antimycin 36 h



Antimycin 36 h







Conclusion

- Cathepsin K is involved in oxidative stress-induced cardiomyocyte apoptosis
- Pharmacological inhibition of Cathepsin K attenuates cardiomyocyte apoptosis
- **Significance**
 - Potential target for treatment of cardiovascular diseases
 - Cat K inhibitor is currently in clinical trials

References

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Thanks!

- ◎ Yinan Hua
- ◎ Dr. Sreejayan Nair