Vall d'Hebron Hospital

ACORVH

ESC Council on Basic Cardiovascular Science

EUROPEAN SOCIETY OF CARDIOLOGY

CBCS/ESC Summer School on Cardiovascular Sciences
"From Basic Mechanisms to Clinical Application"

European Heart House / 12 – 16 June 2011

'Ischaemia and reperfusion'

Basic concepts- D Garcia Dorado (SP)

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Ischemia and Reperfusion. Basic concepts

1) Myocardial Ischemia
2) Reperfusion salvage and reperfusion injury
3) Basic molecular mechanisms
4) Integrative view
5) Translational perspective
6) Conclusion

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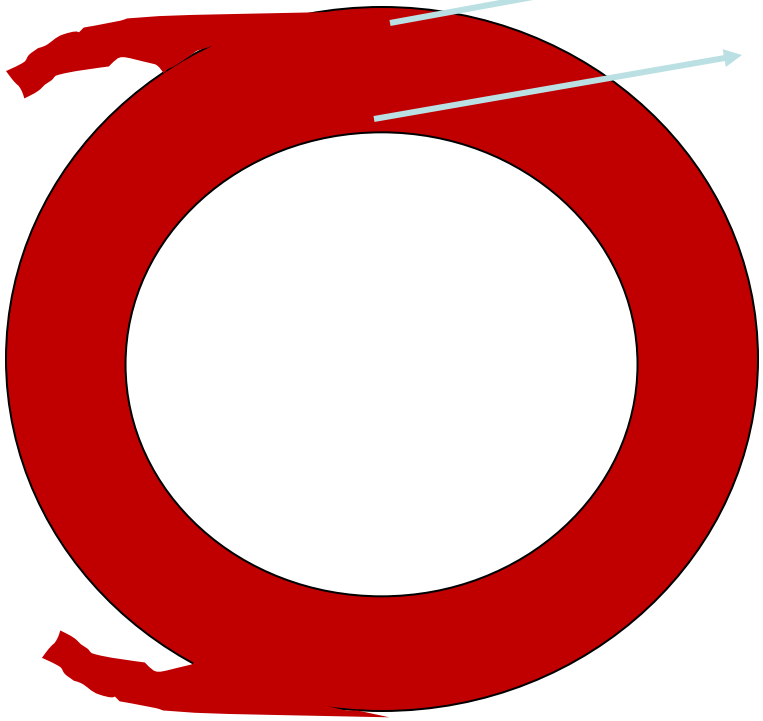
ISCHEMIA

Blood flow insufficient to sustain aerobic metabolism

ISCHEMIC HEART DISEASE

Estenosis /occlusion of epicardial coronary arteries



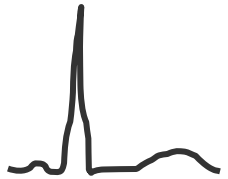
Subepicardial myocardium: less tissue pressure

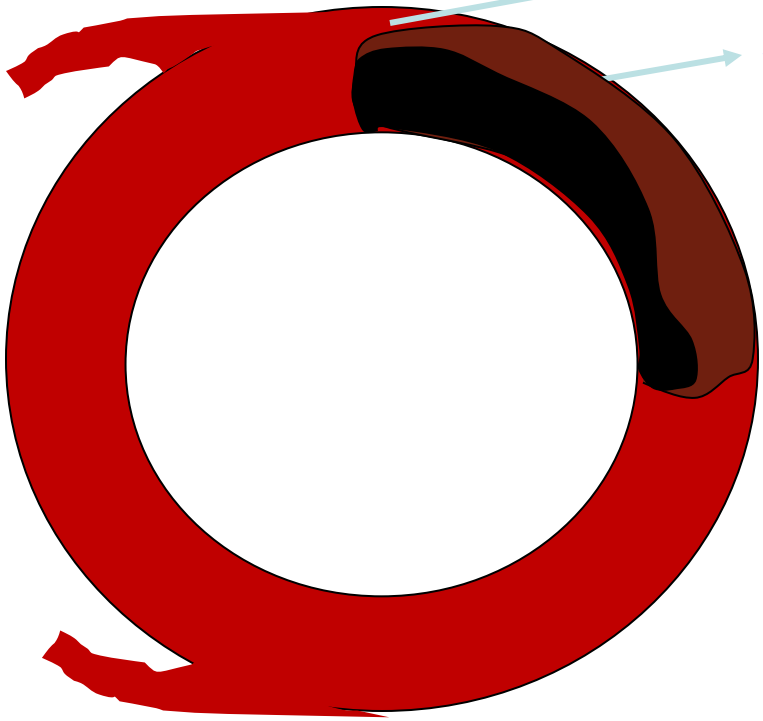
Subencardial layer: high tissue pressure

Normal

Arteriolar R (endo) < R(epi)

Flow (endo) = Flow (epi)





Subepicardial myocardium: less tissue pressure

Subencardial layer: high tissue pressure

Normal

Arteriolar R (endo) < R(epi)

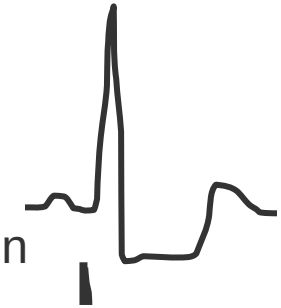
Flow (endo) = Flow (epi)

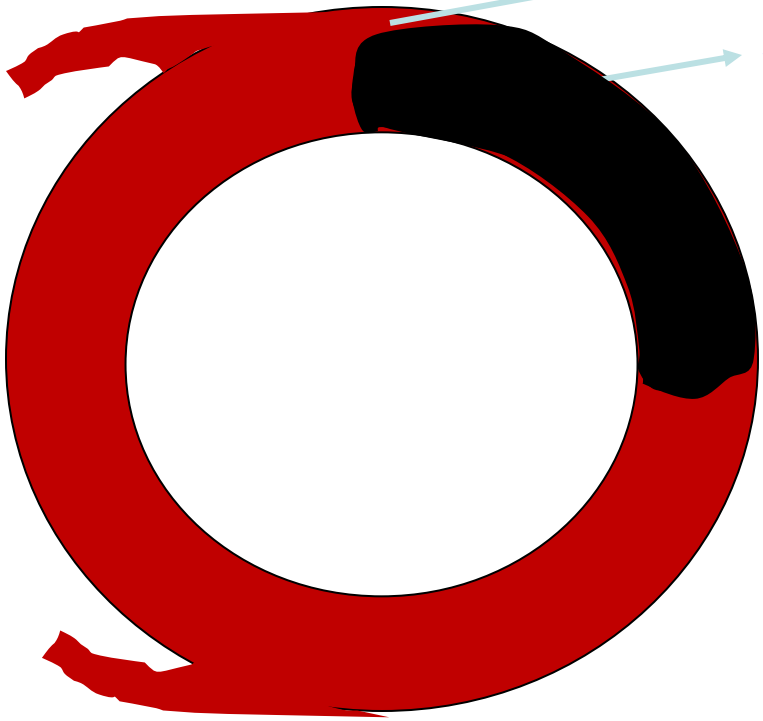
Hypoperfusion

Reactive vasodilation

R(endo) = R(epi)

F(endo) < F(epi) = 1 ml/gr/min





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Hypoperfusion

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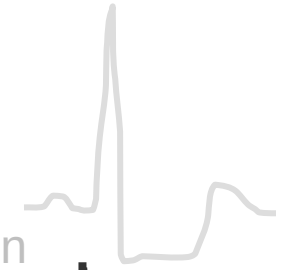
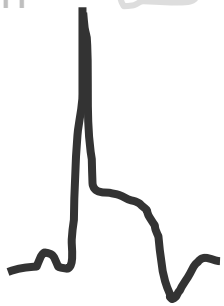
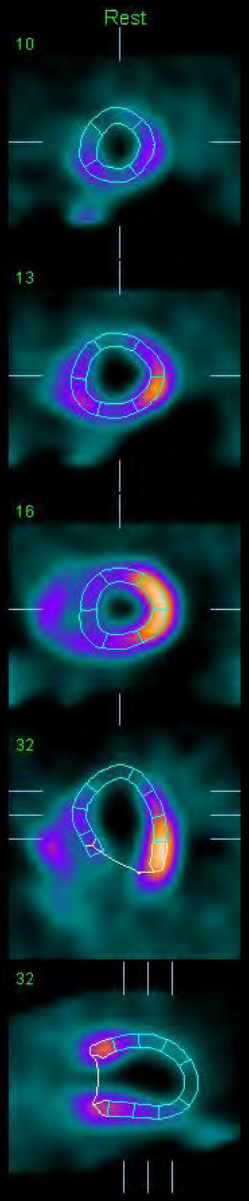
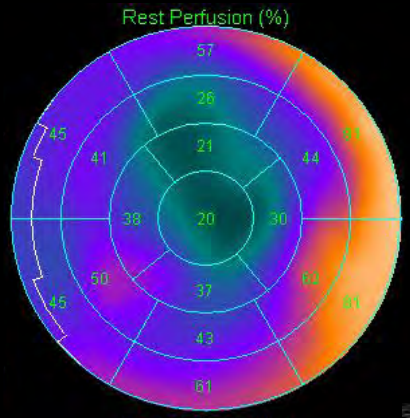


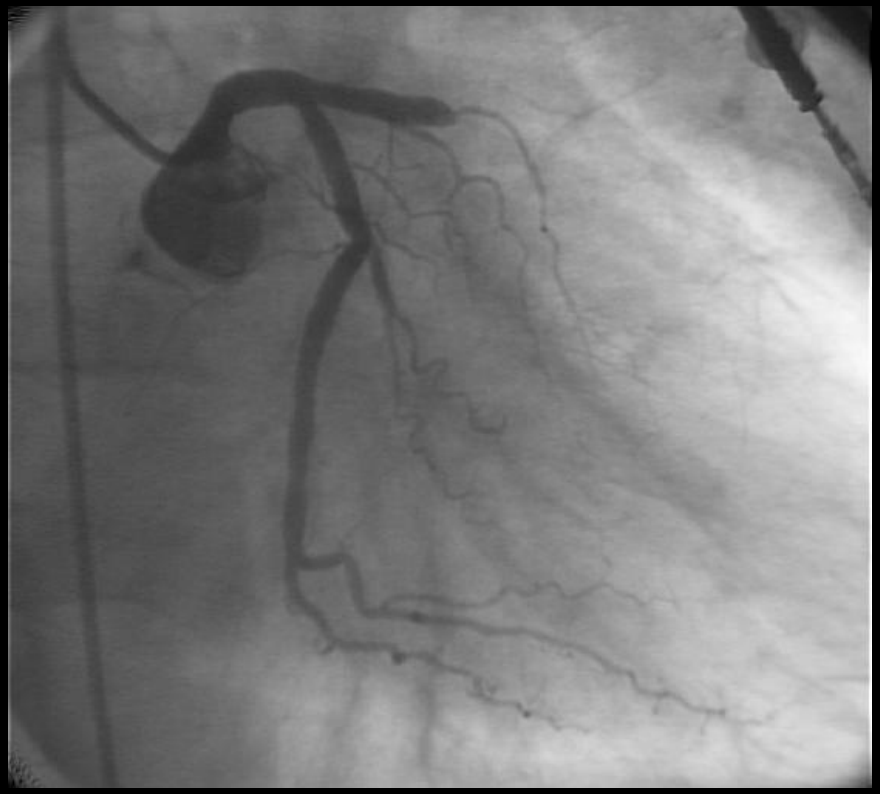
No flow

Flow(endo) = Flow(epi) = 0









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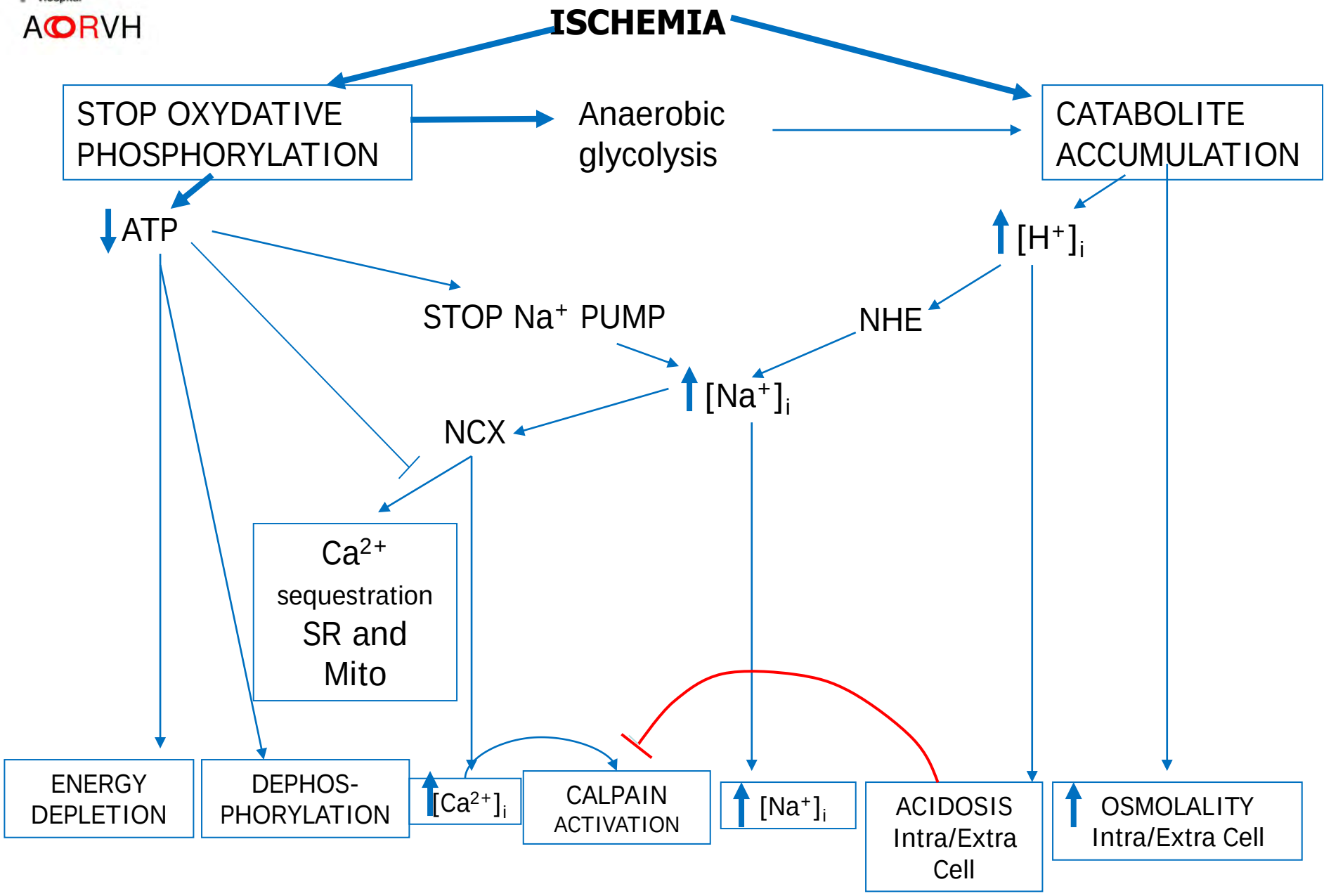
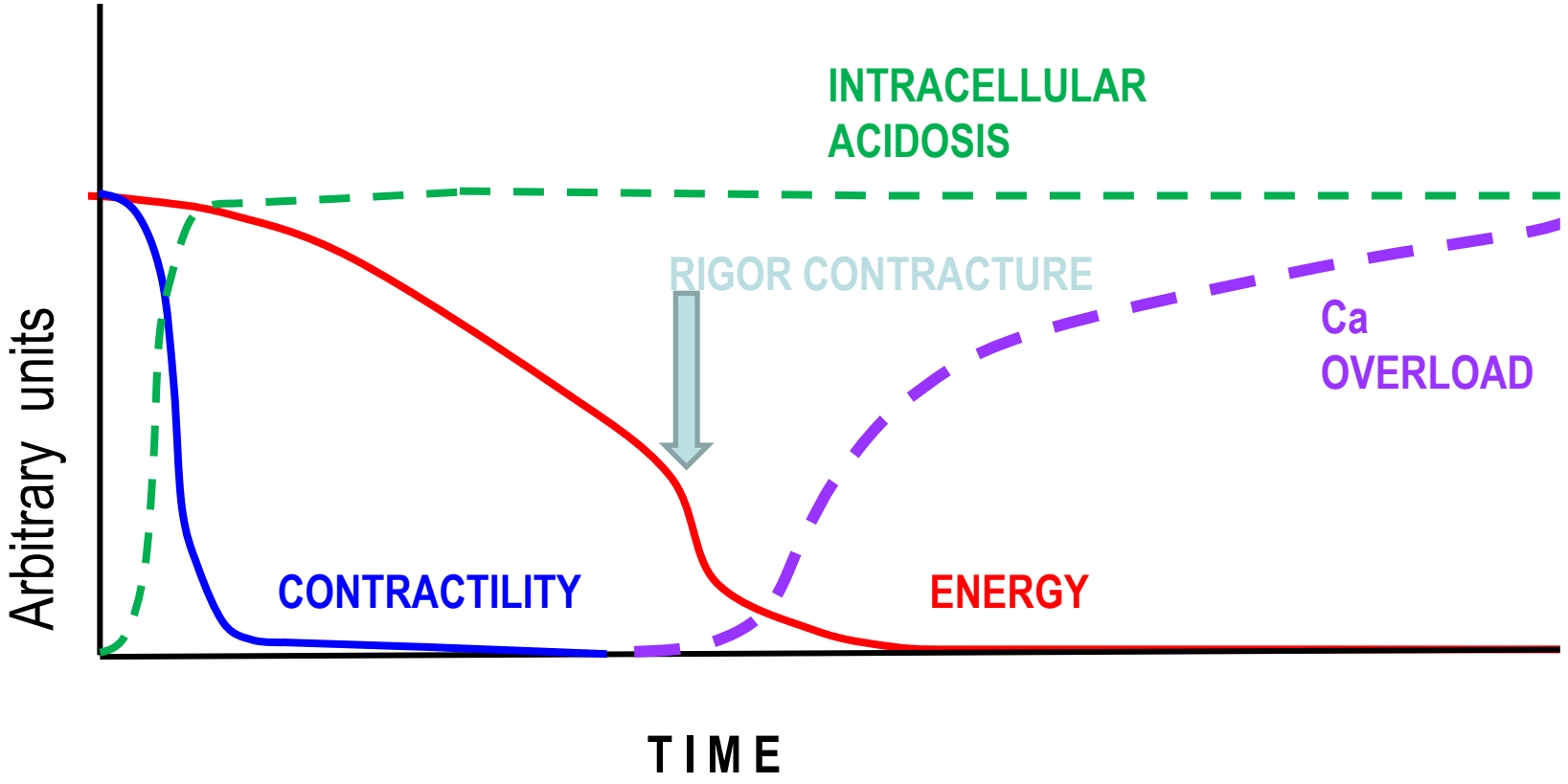
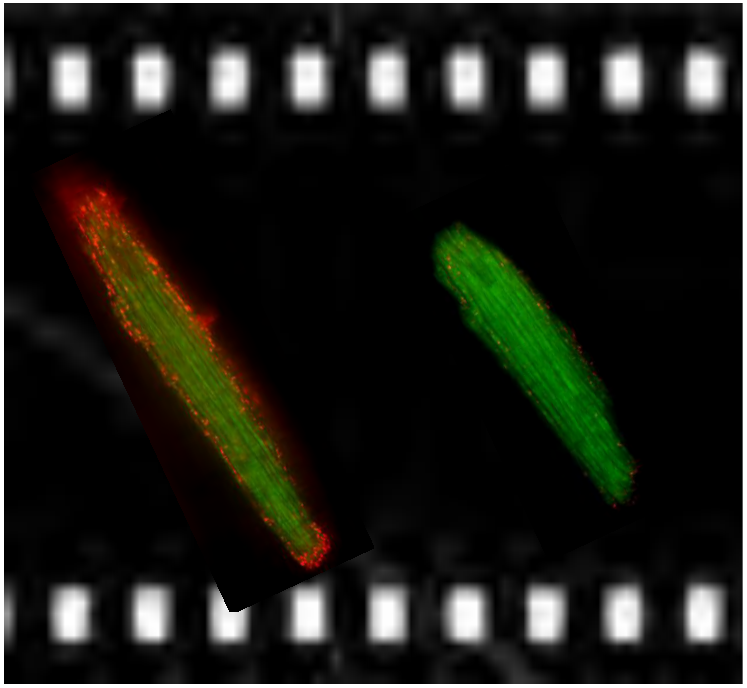

ISCHEMIA
STOP OXYDATIVE PHOSPHORYLATION
Anaerobic glycolysis
CATABOLITE ACCUMULATION
ATP
[H+]i
STOP Na+ PUMP
NHE
[Na+]i
NCX
Ca2+ sequestration SR and Mito
ENERGY DEPLETION
DEPHOS-PHORYLATION
[Ca2+]i
CALPAIN ACTIVATION
[Na+]i
ACIDOSIS Intra/Extra Cell
OSMOLALITY Intra/Extra Cell

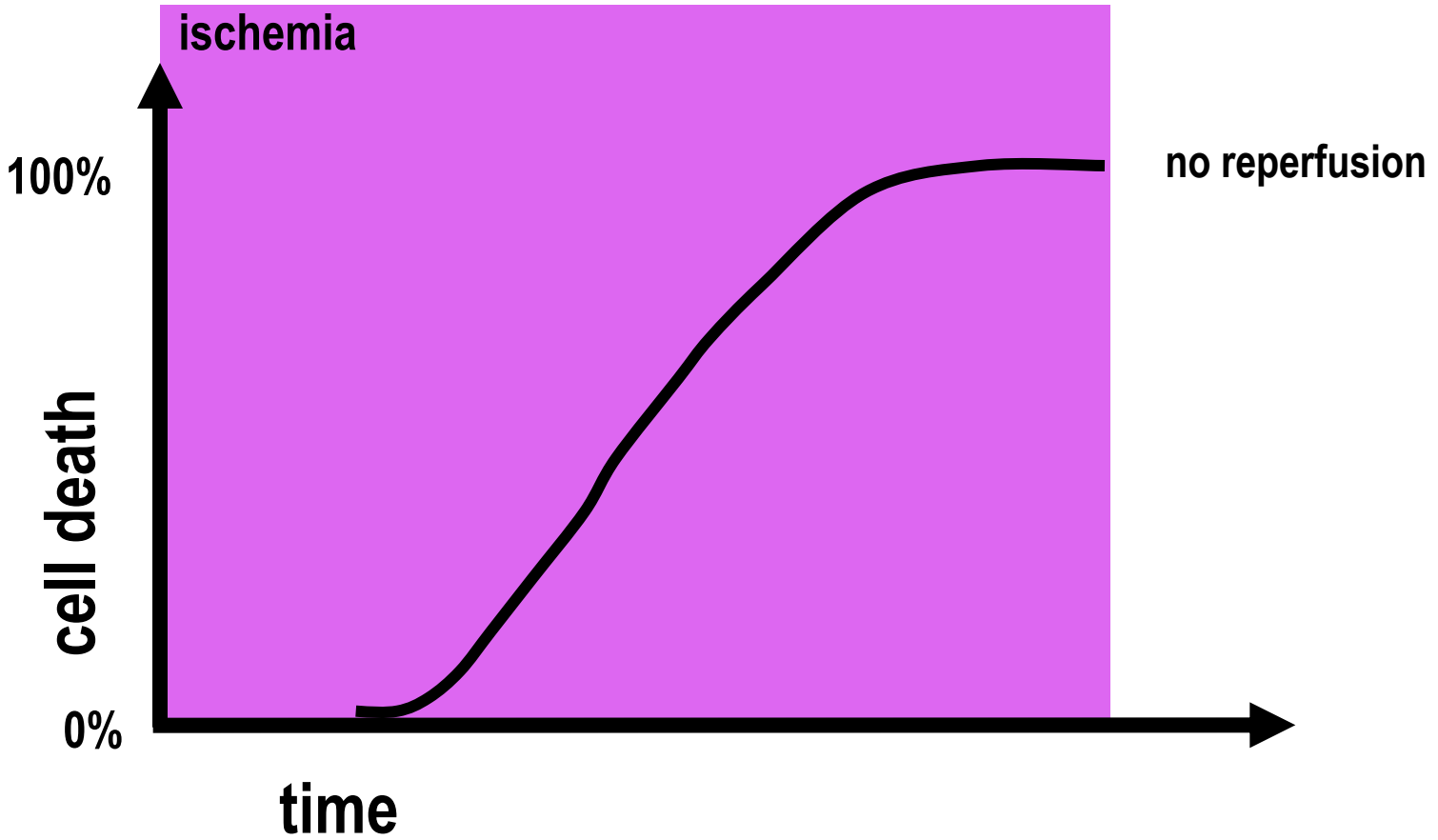
CHRONOLOGY OF ISCHEMIC CHANGES



Ischemic rigor



Normoxia **20 min Isch (anoxia pH 7.4)**



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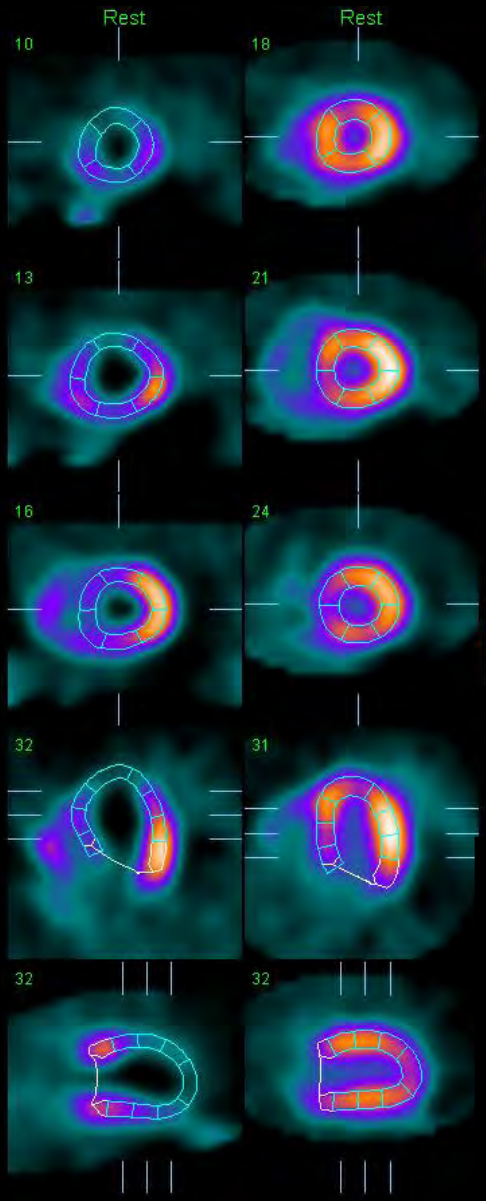
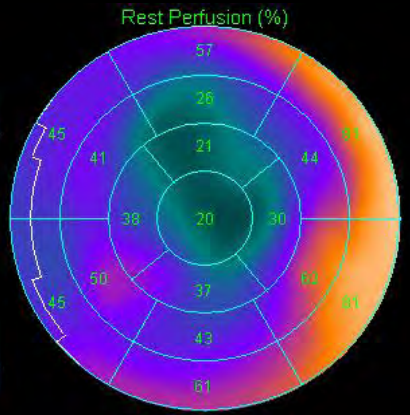
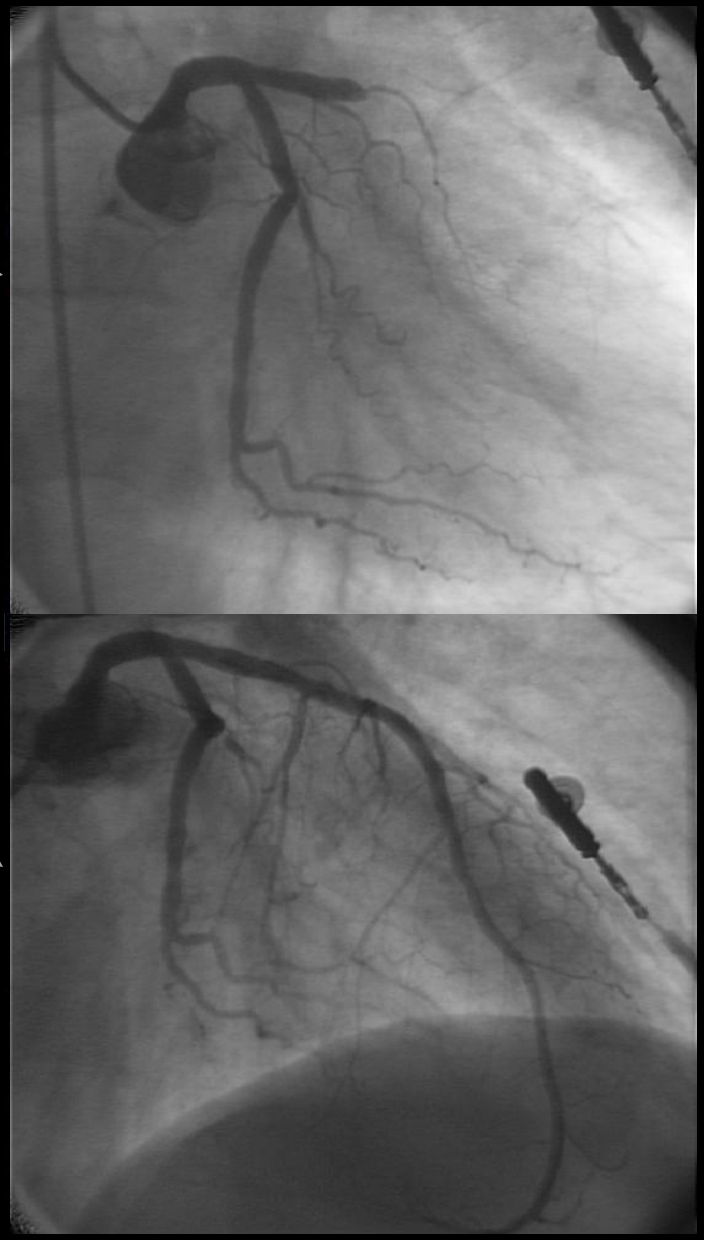
Ischemia and Reperfusion. Basic concepts

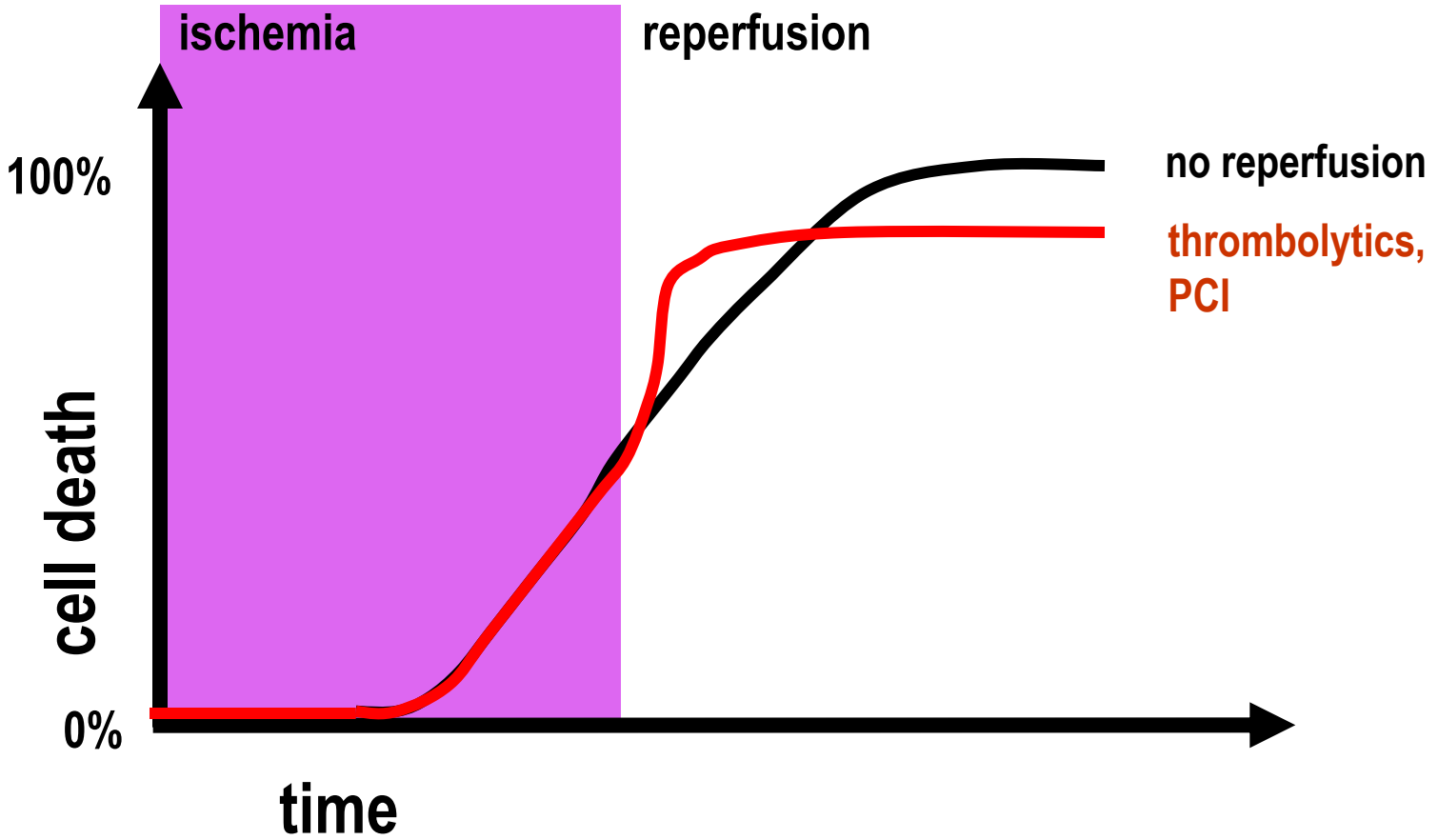
1) Myocardial Ischemia
2) **Reperfusion salvage and reperfusion injury**
3) Basic molecular mechanisms
4) Integrative view
5) Translational perspective
6) Conclusion


Rest
Rest
10
18
13
21
16
24
32
31
32
32


Rest Perfusion (%)
57
26
45
41
21
44
38
20
30
60
37
61
45
43
61
61


Rest Perfusion (%)
59
71
66
92
80
78
66
77
64
72
93
75
63
76
67





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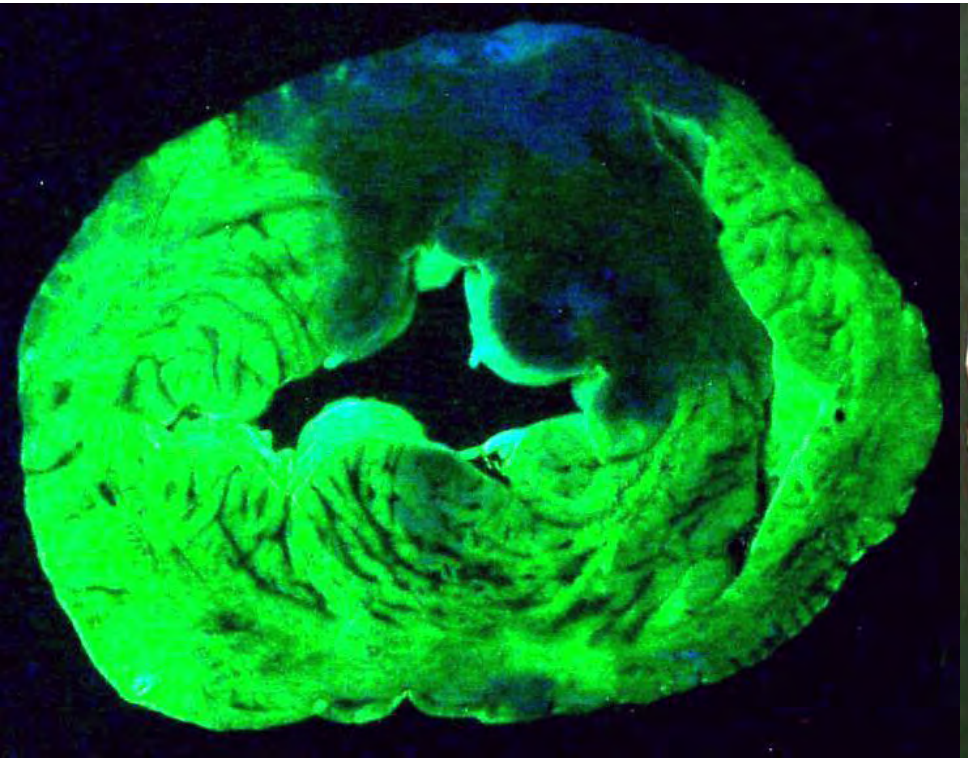
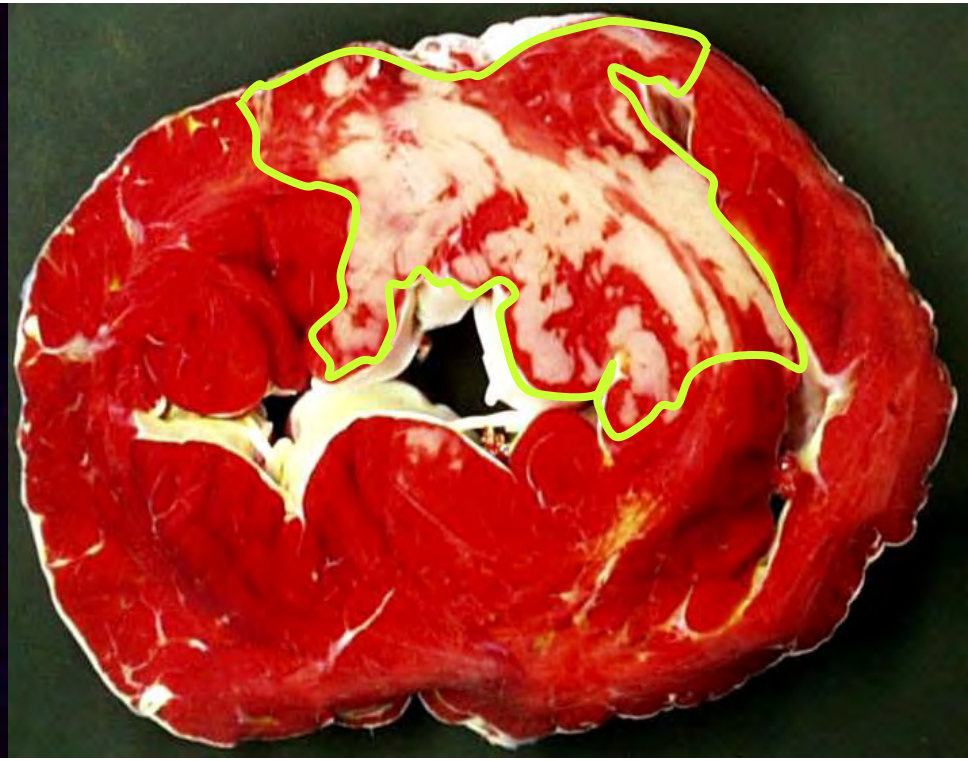
Myocardial infarctIon after reperfusion following 48 min of LAD occlusion



AREA AT RISK



INFARCT AREA

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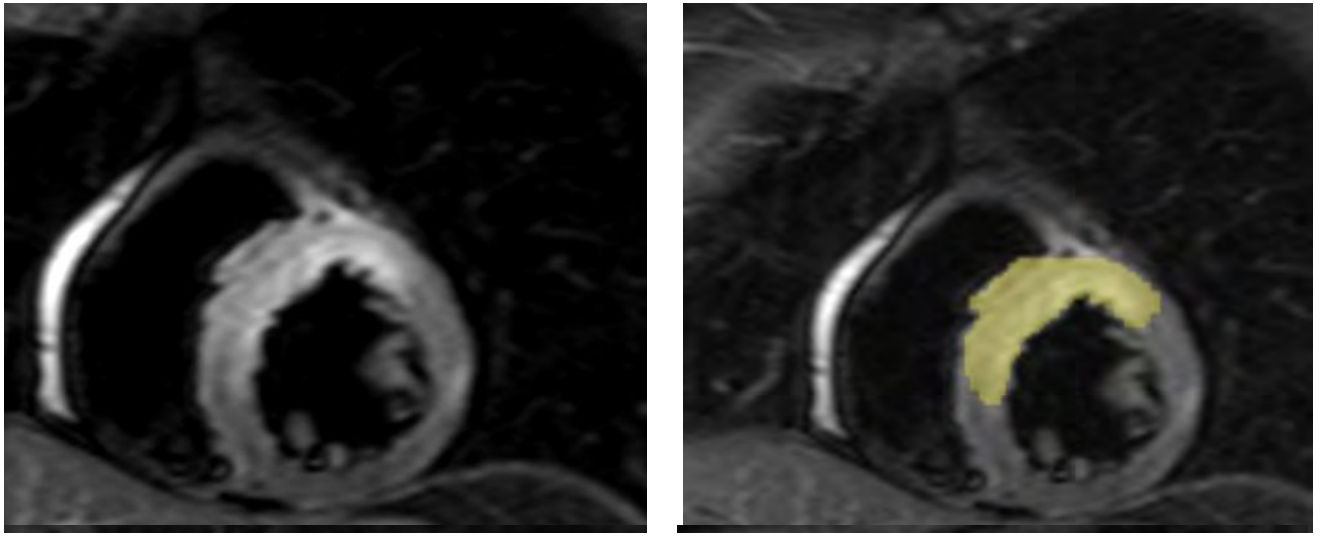
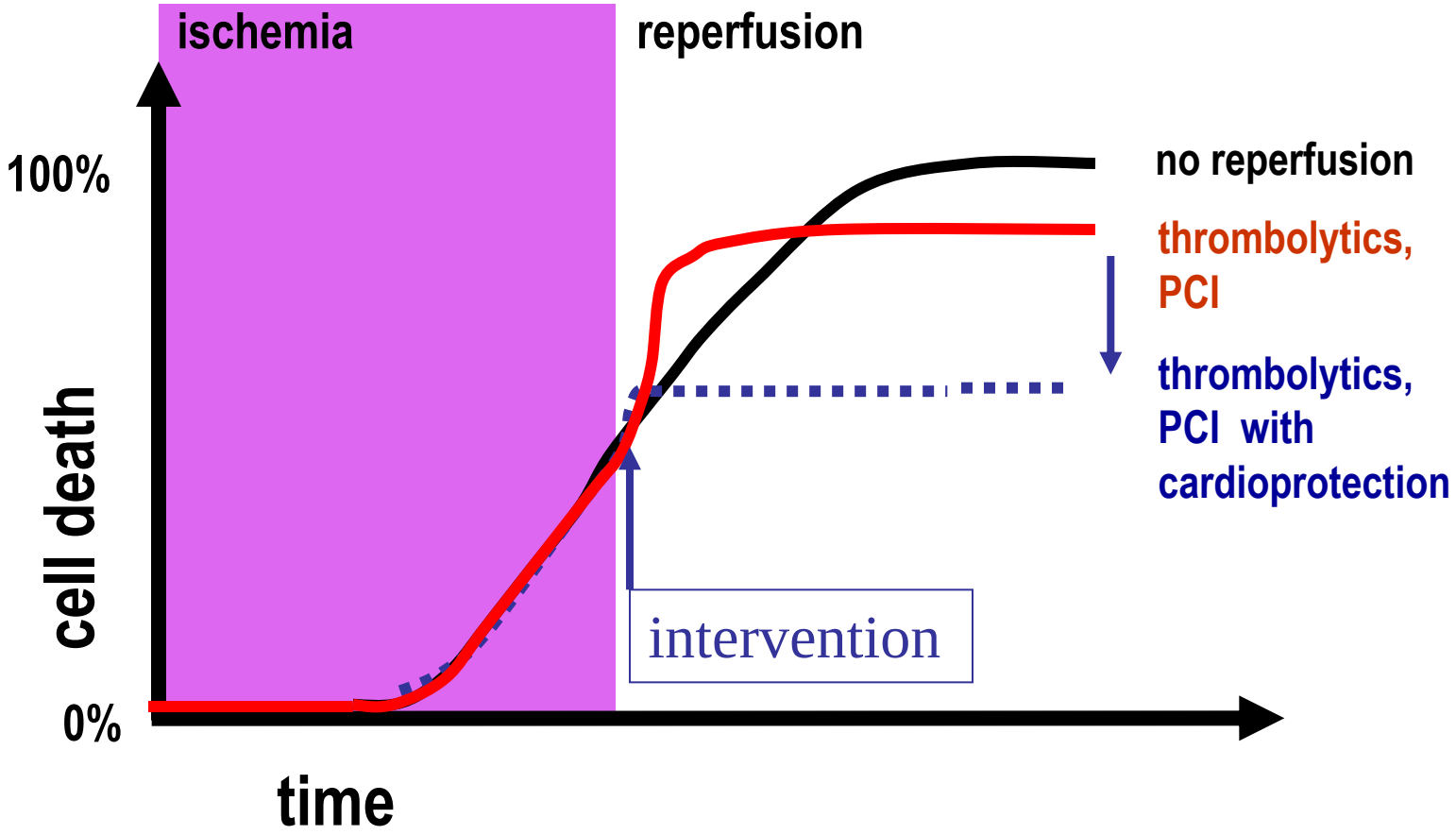


Reperfusion injury:

cell death preventable by interventions applied at the time of reperfusion



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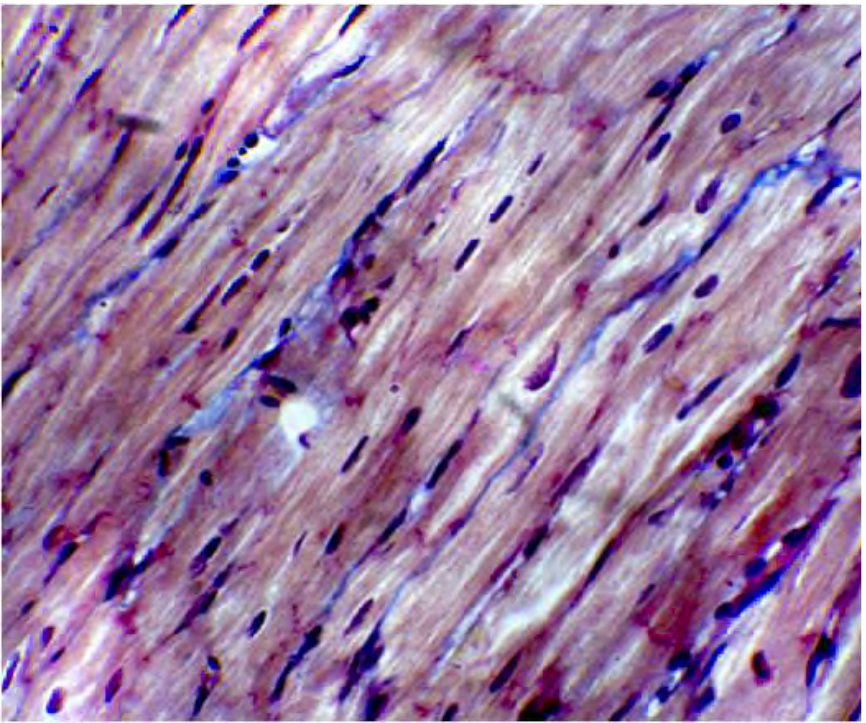
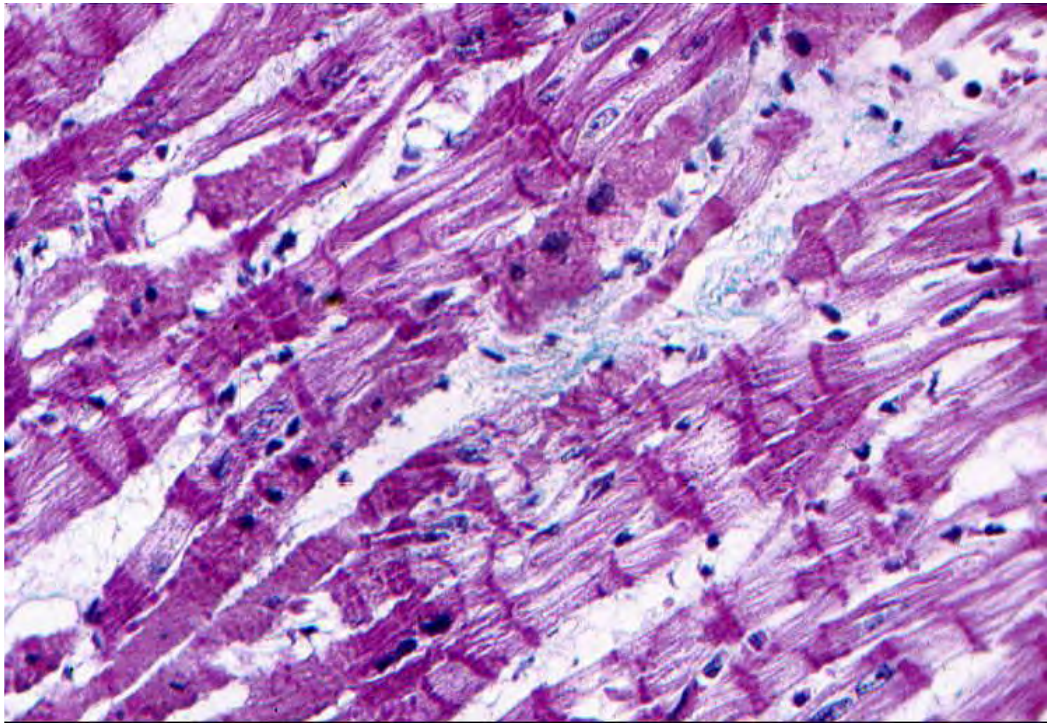
Ischemia and Reperfusion. Basic concepts

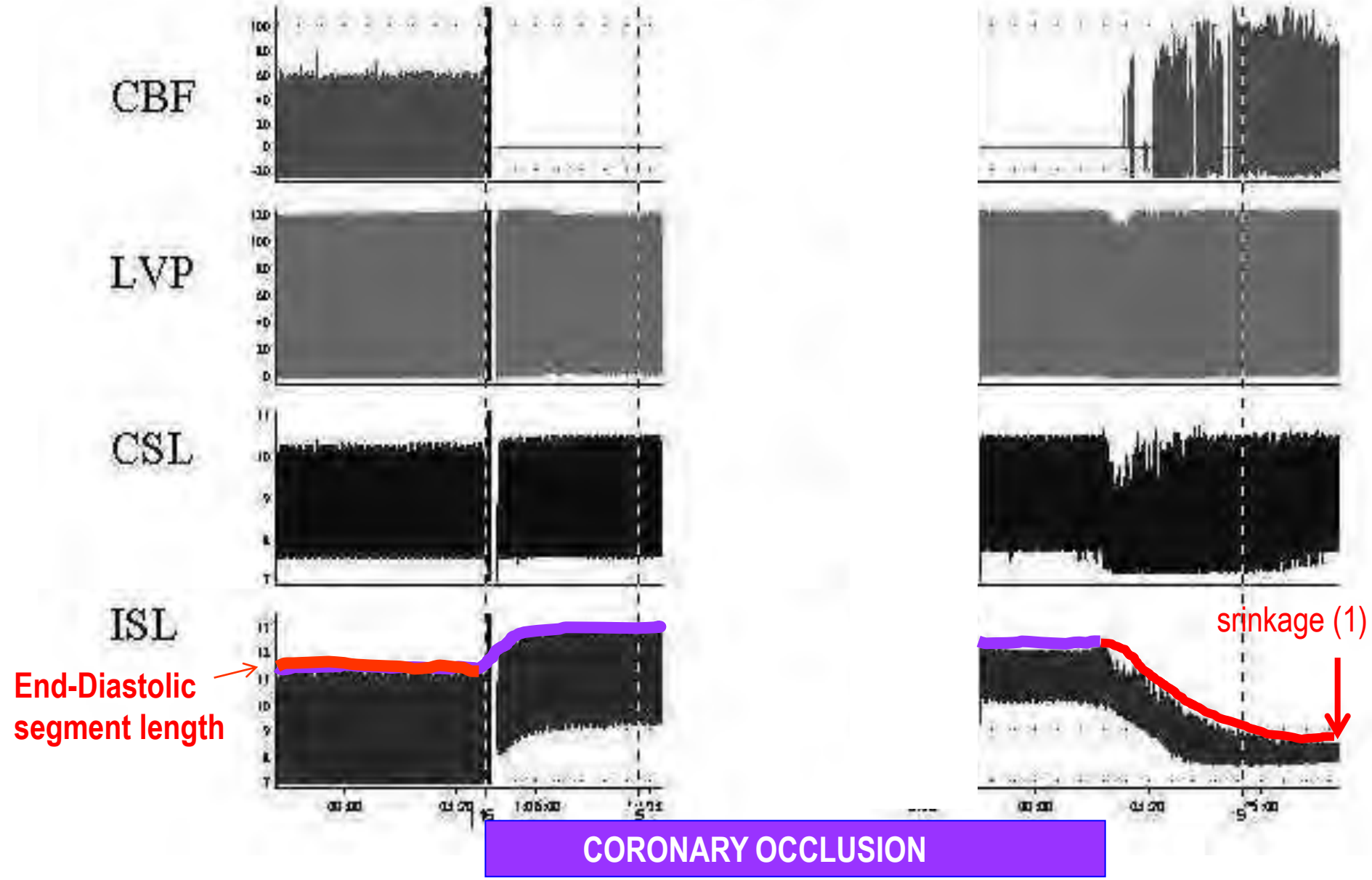
1) Myocardial Ischemia
2) Reperfusion salvage and reperfusion injury
3) **Basic molecular mechanisms**
4) Integrative view
5) Translational perspective
6) Conclusion

Post reperfusion necrosis: ***"Contraction band necrosis"***





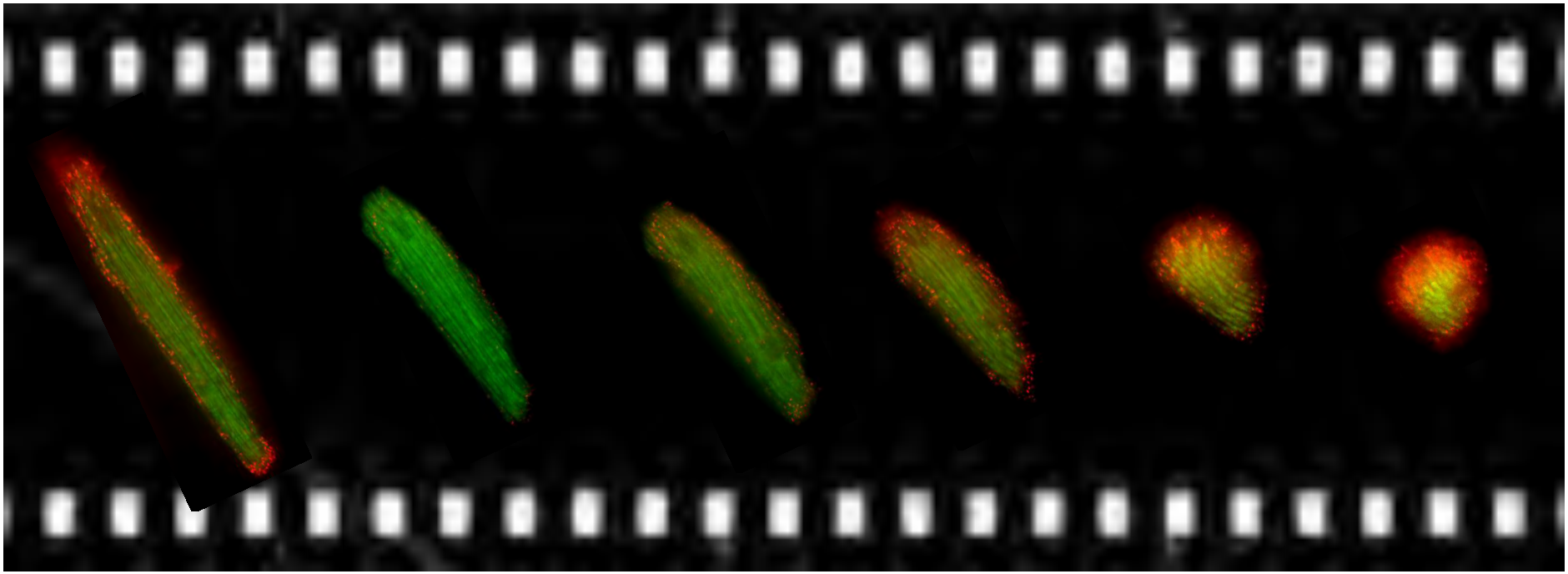
Reperfusion after 48 min coronary occlusion in the anesthesized pig



(1) Barrabés et al. Pflüger Arch 1998

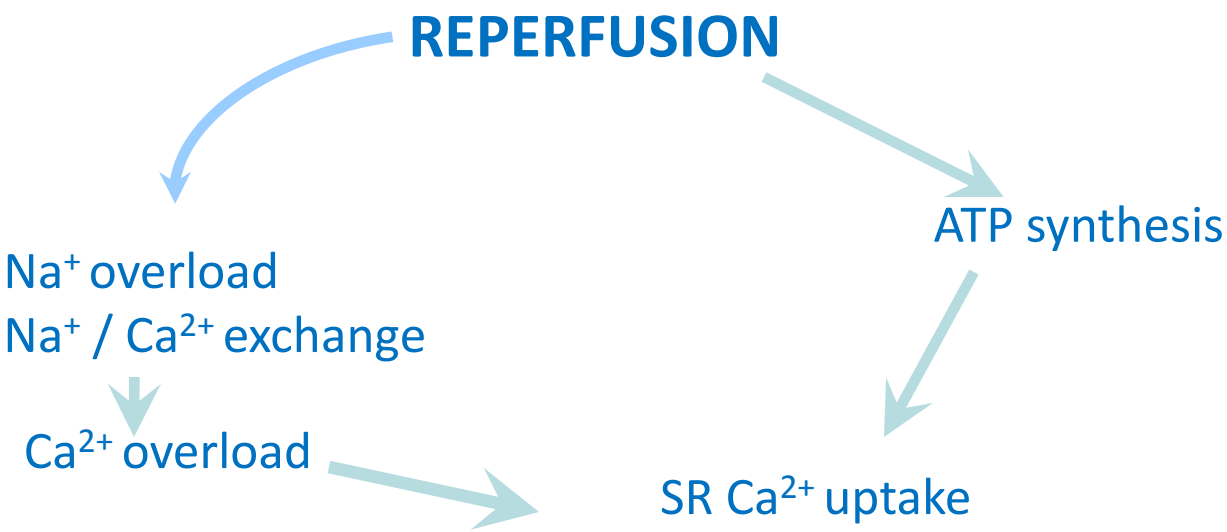
Ischemic rigor

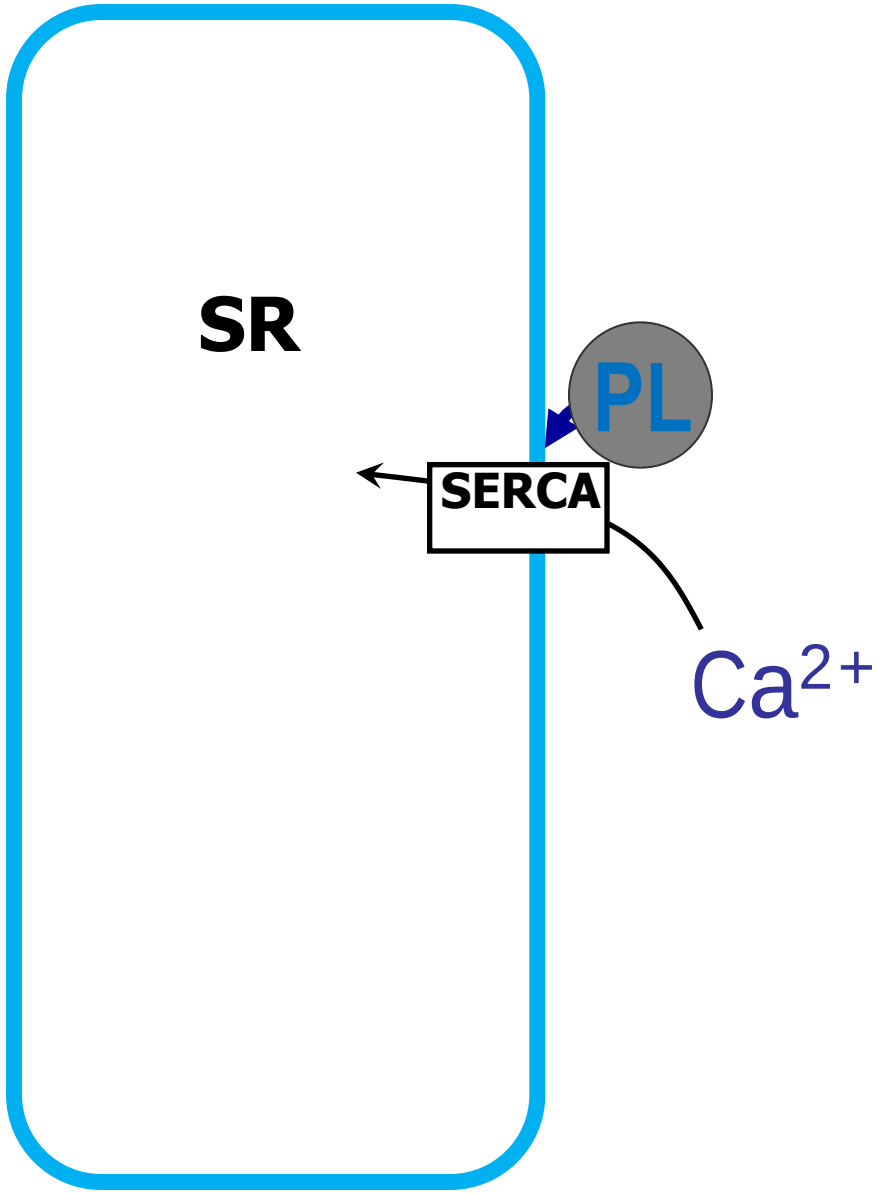
Hyper-contracture

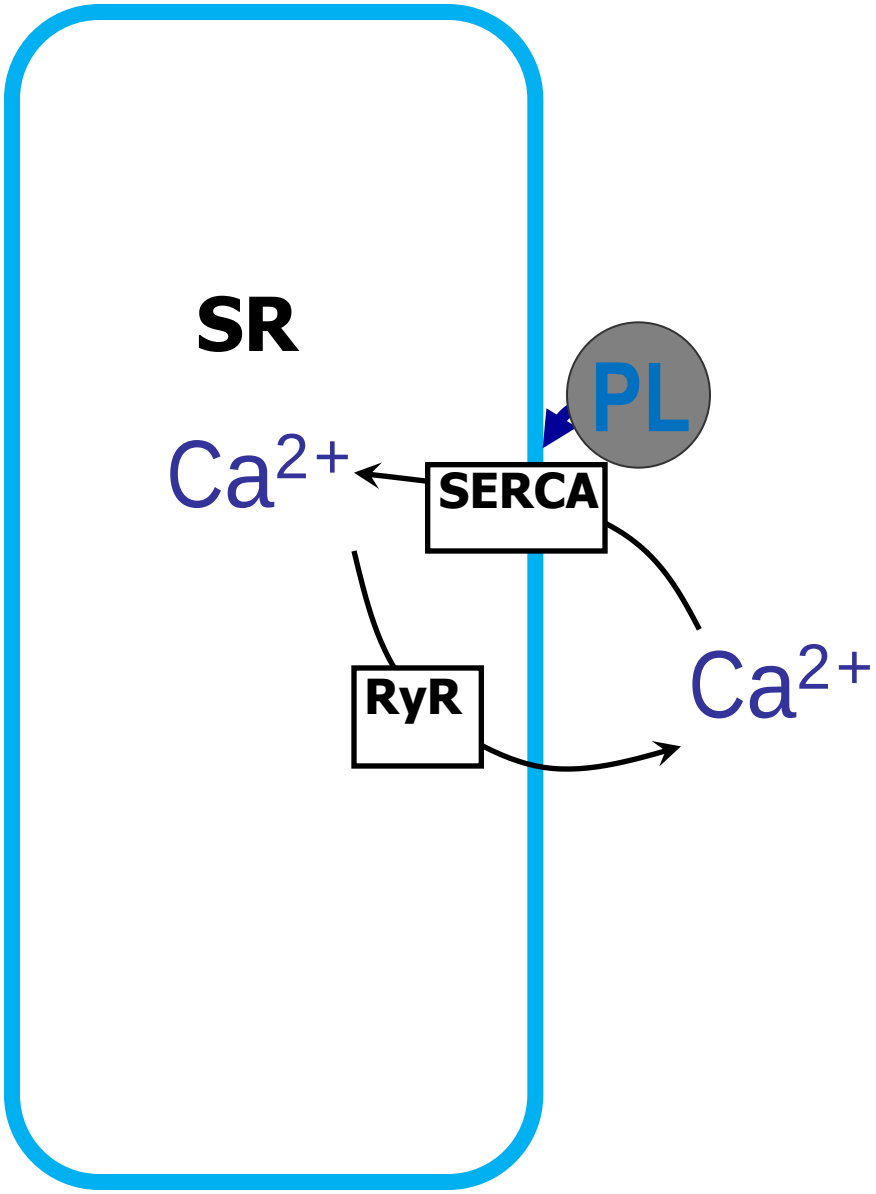


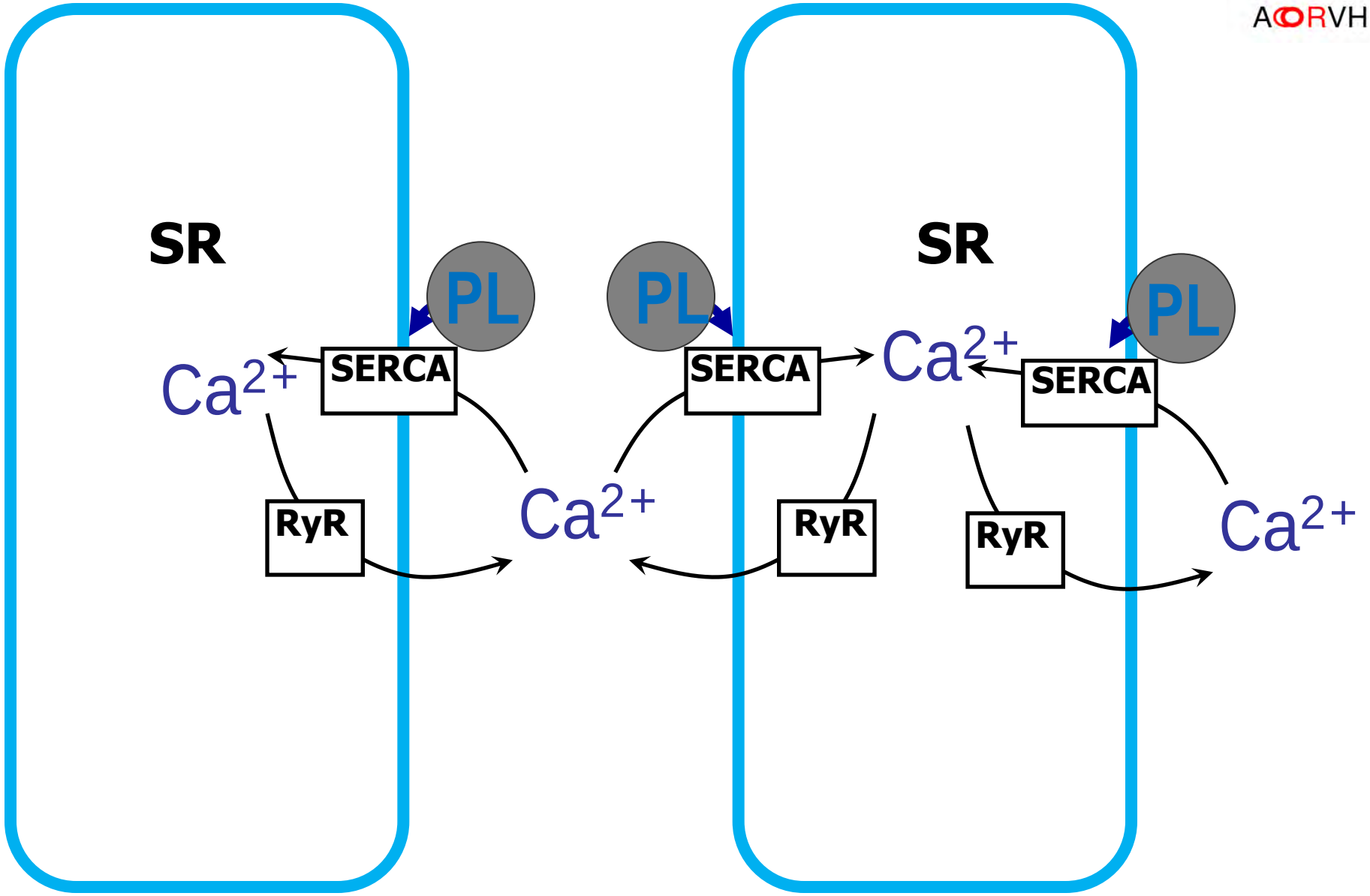
Normoxia **20 min Isch (anoxia pH 7.4)** **1” 60” 120” 180”**

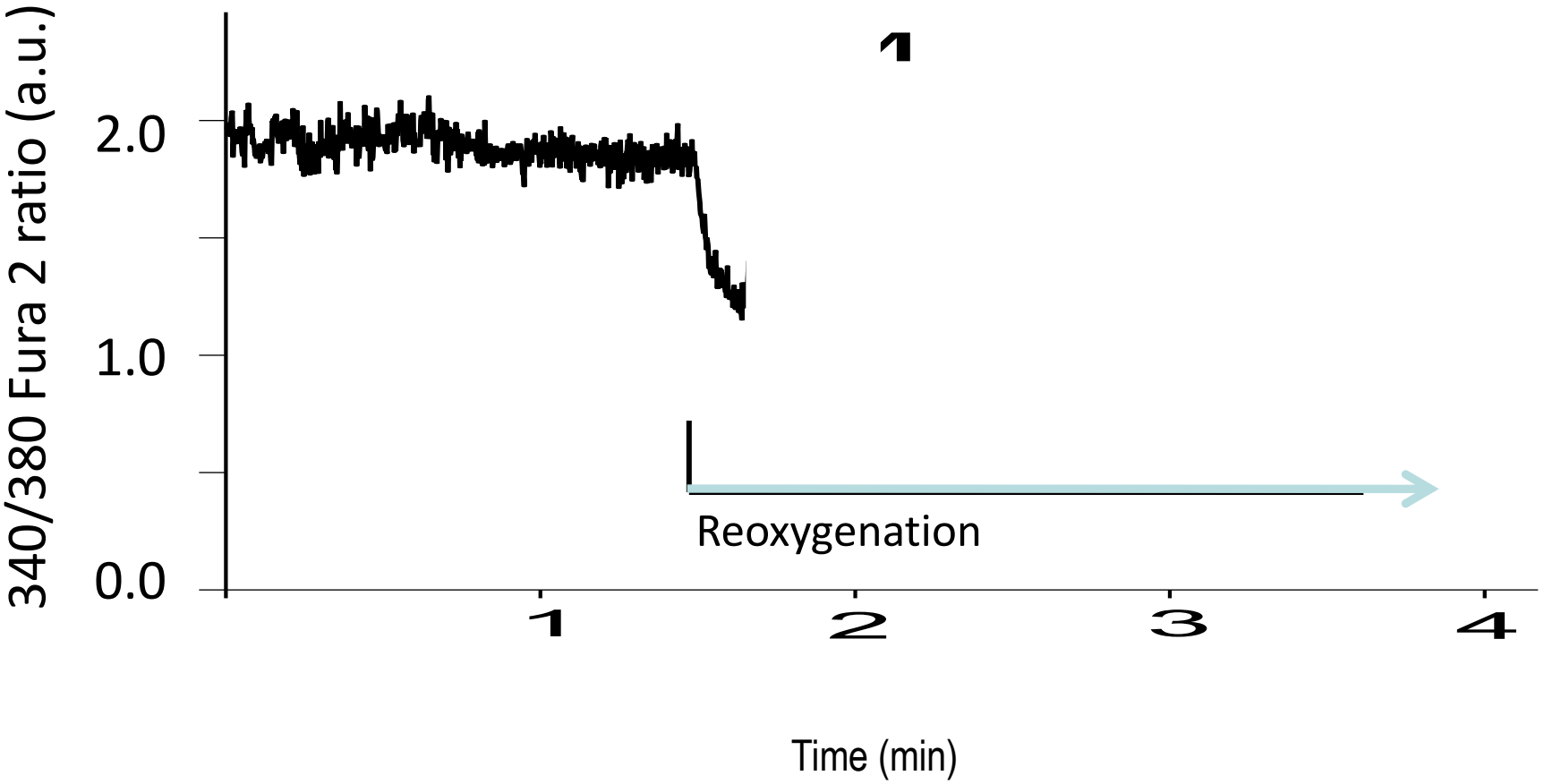
Reperfusion

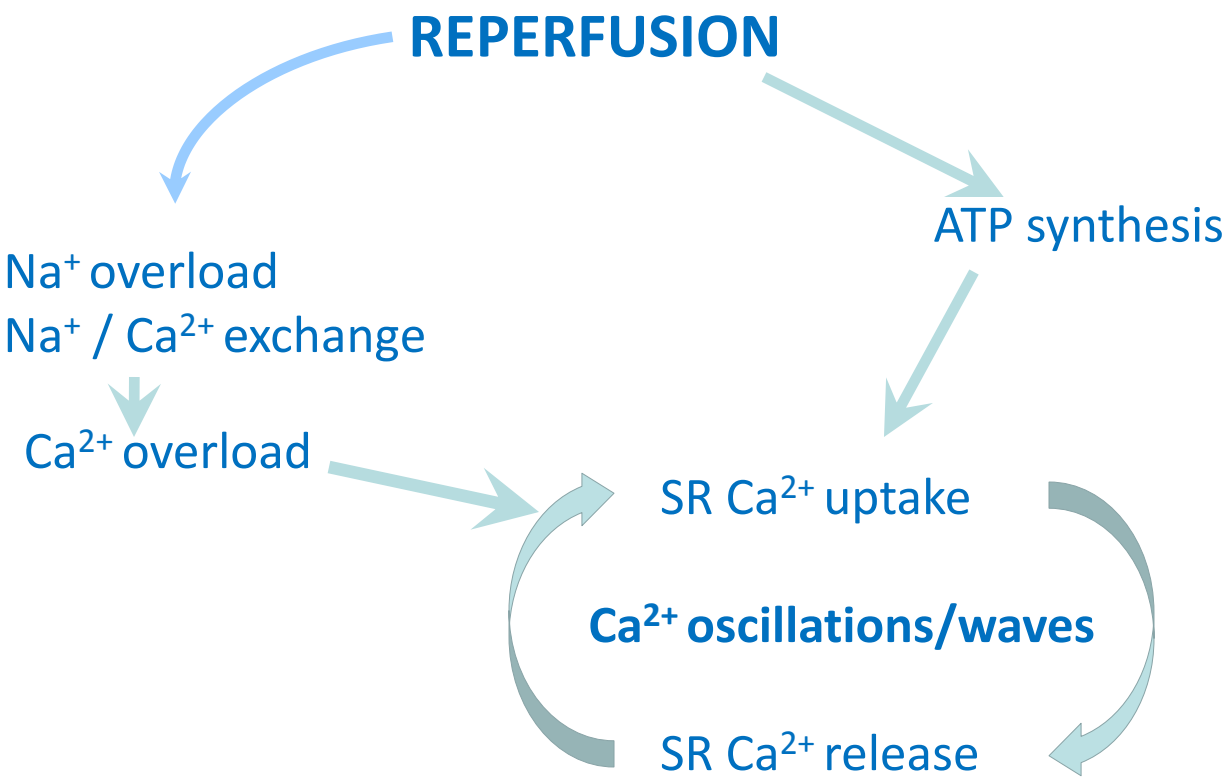

REPERFUSION
ATP synthesis
Na+ overload
Na+ / Ca2+ exchange
Ca2+ overload
SR Ca2+ uptake

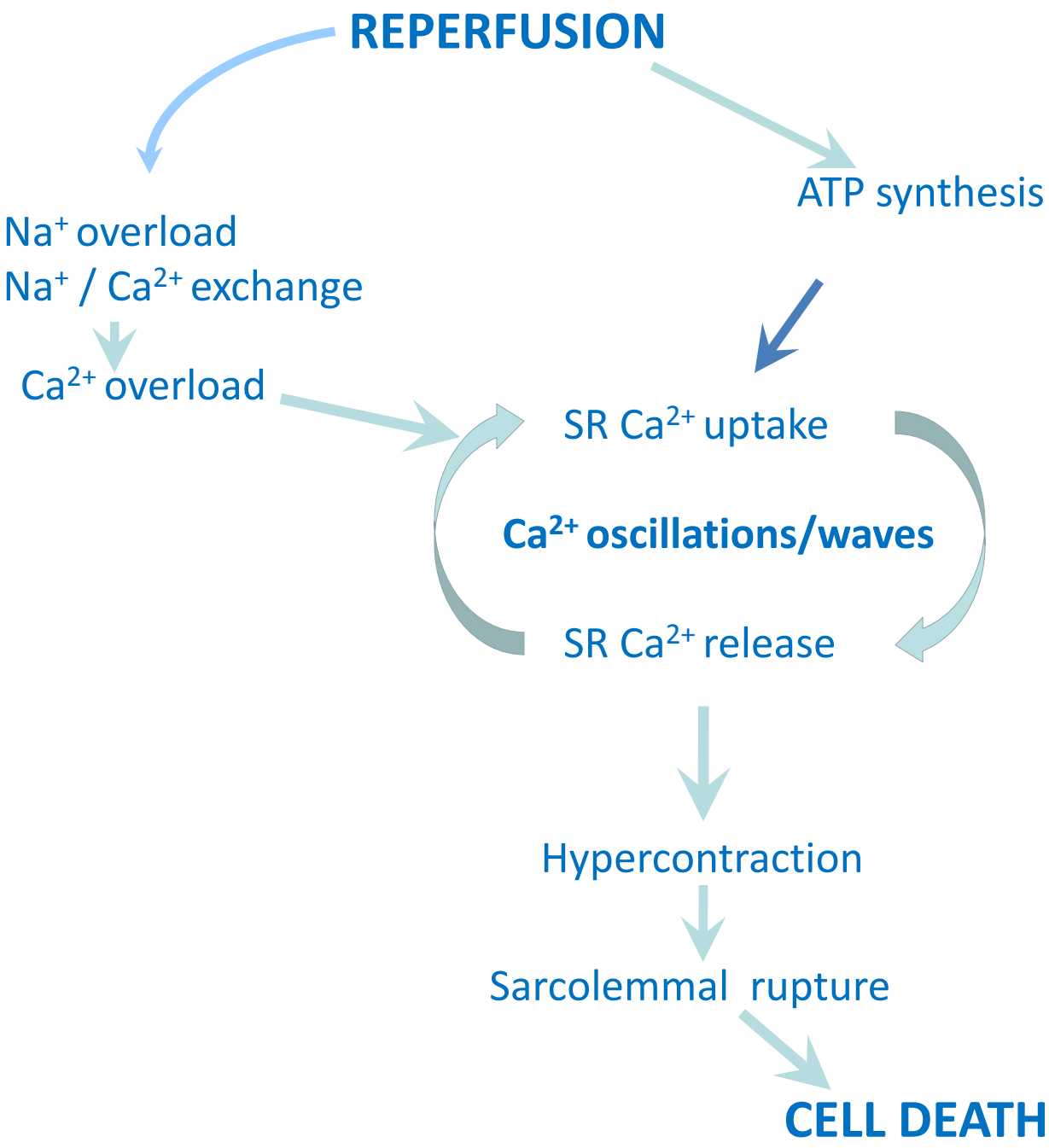

SR
PL
SERCA
Ca2+


SR
PL
Ca^{2+}
SERCA
RyR
Ca^{2+}


SR
PL
Ca2+
SERCA
RyR
Ca2+
PL
SERCA
RyR
SR
Ca2+
PL
SERCA
RyR
Ca2+


340/380 Fura 2 ratio (a.u.)
2.0
1.0
0.0
1
2
3
4
Reoxygenation
Time (min)


REPERFUSION
ATP synthesis
Na+ overload
Na+ / Ca2+ exchange
Ca2+ overload
SR Ca2+ uptake
Ca2+ oscillations/waves
SR Ca2+ release


REPERFUSION
ATP synthesis
Na+ overload
Na+ / Ca2+ exchange
Ca2+ overload
SR Ca2+ uptake
Ca2+ oscillations/waves
SR Ca2+ release
Hypercontraction
Sarcolemmal rupture
CELL DEATH

Mechanism of cell death during reperfusion: contration band necrosis

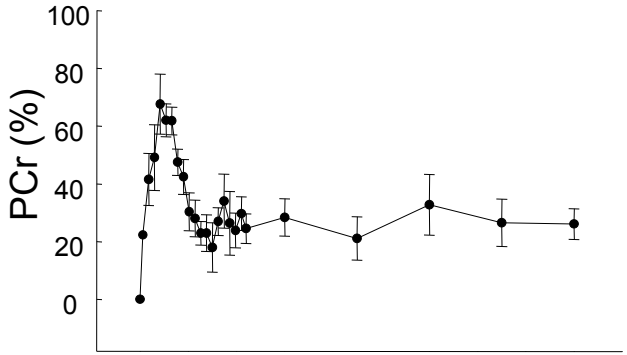
Energetic recovery of reperfused myocardium

40 min

Mechanism of cell death during reperfusion: contration band necrosis

Energetic recovery of reperfused myocardium

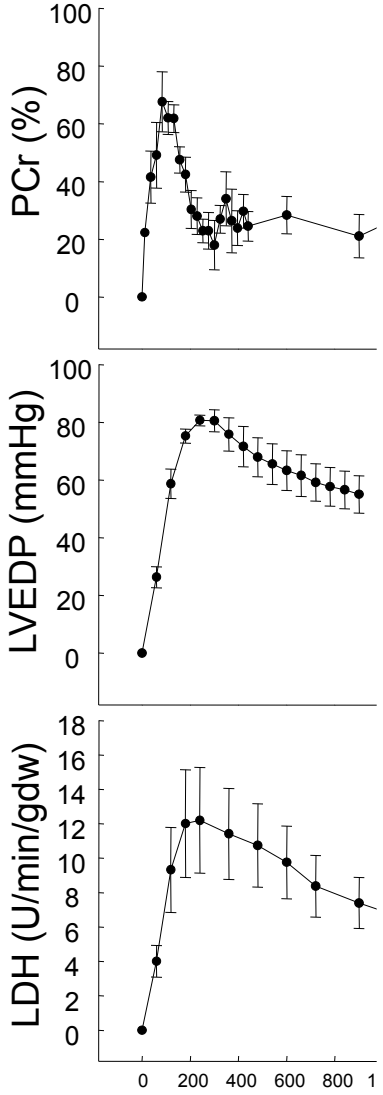
40 min



Mechanism of cell death during reperfusion: contration band necrosis

Energetic recovery of reperfused myocardium

40 min

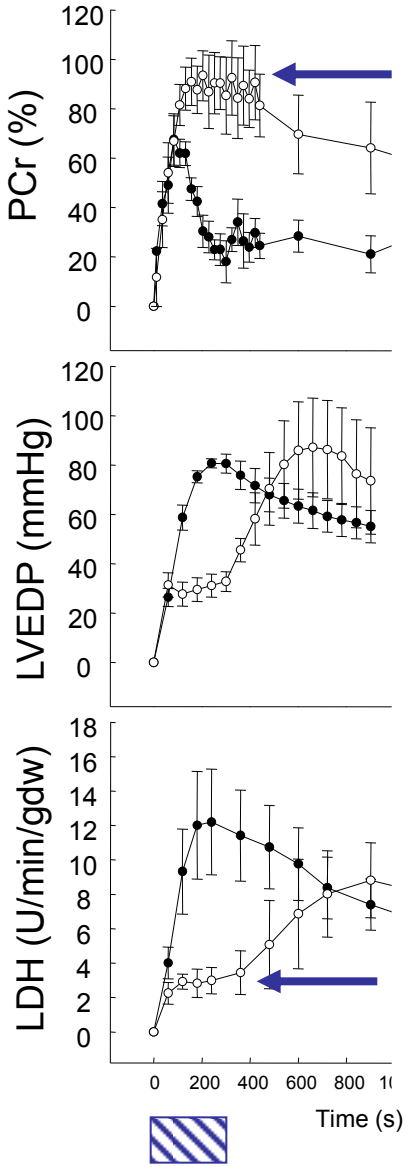


Mechanism of cell death during reperfusion: contration band necrosis

Energetic recovery of reperfused myocardium

40 min

BDM 20 mM 5 min



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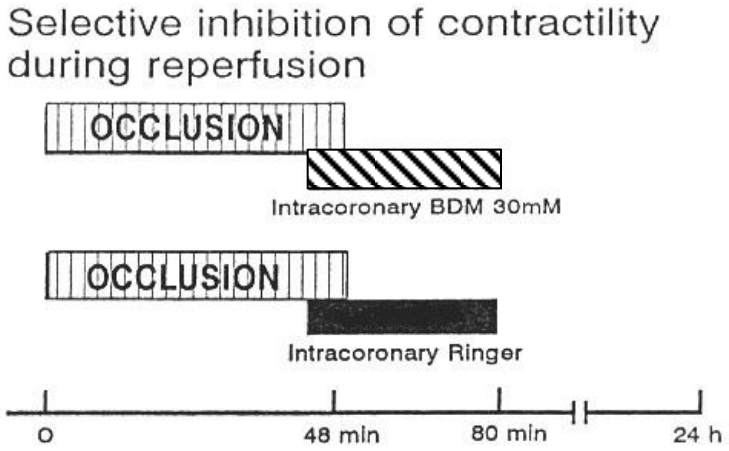
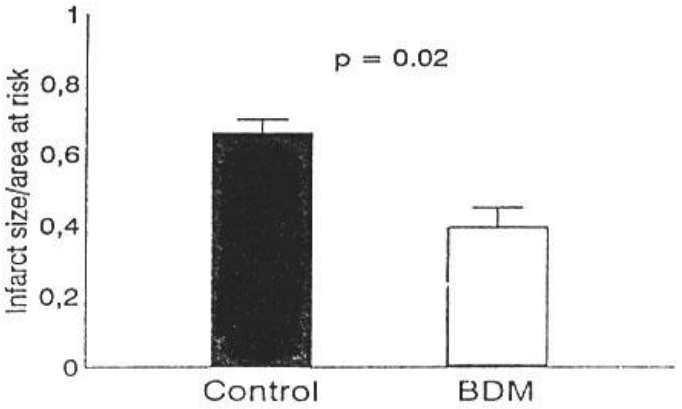
Selective Inhibition of the Contractile Apparatus

David Garcia-Dorado, MD; Pierre Théroux, MD; Juan M. Duran, MD; Julia Solares, MD; Joaquin Alonso, MD; Esther Sanz, MD; Roberto Munoz, MD; Jaime Elizaga, MD; Javier Botas, MD; Francisco Fernandez-Avilés, MD; Javier Soriano, MD; and Elena Esteban, MD

coronary artery occlusion in pigs





(*Circulation* 1992;85:1160–1174)

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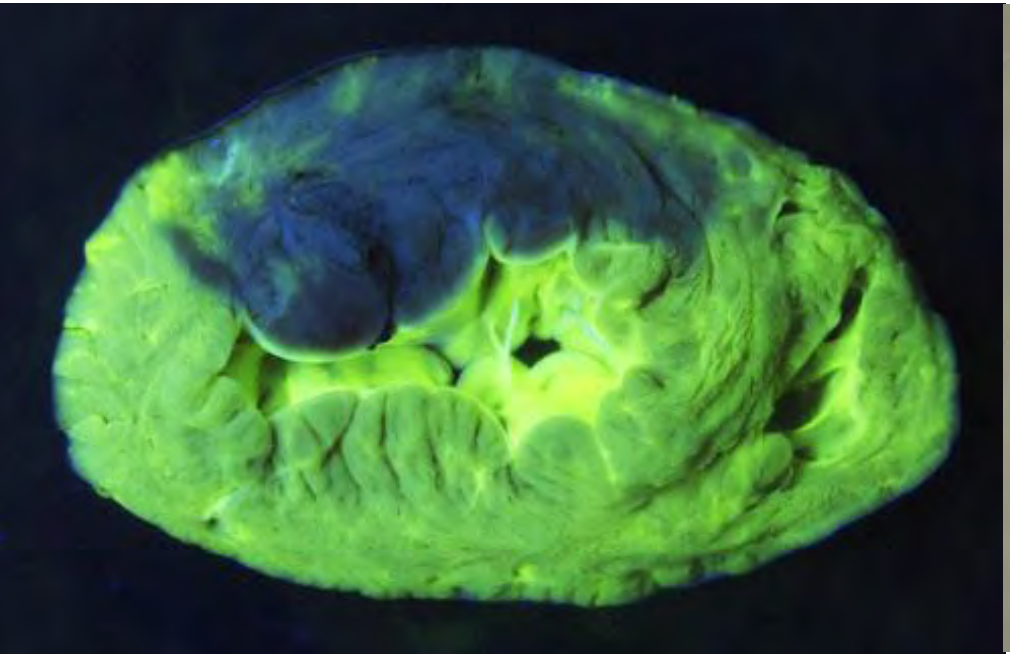
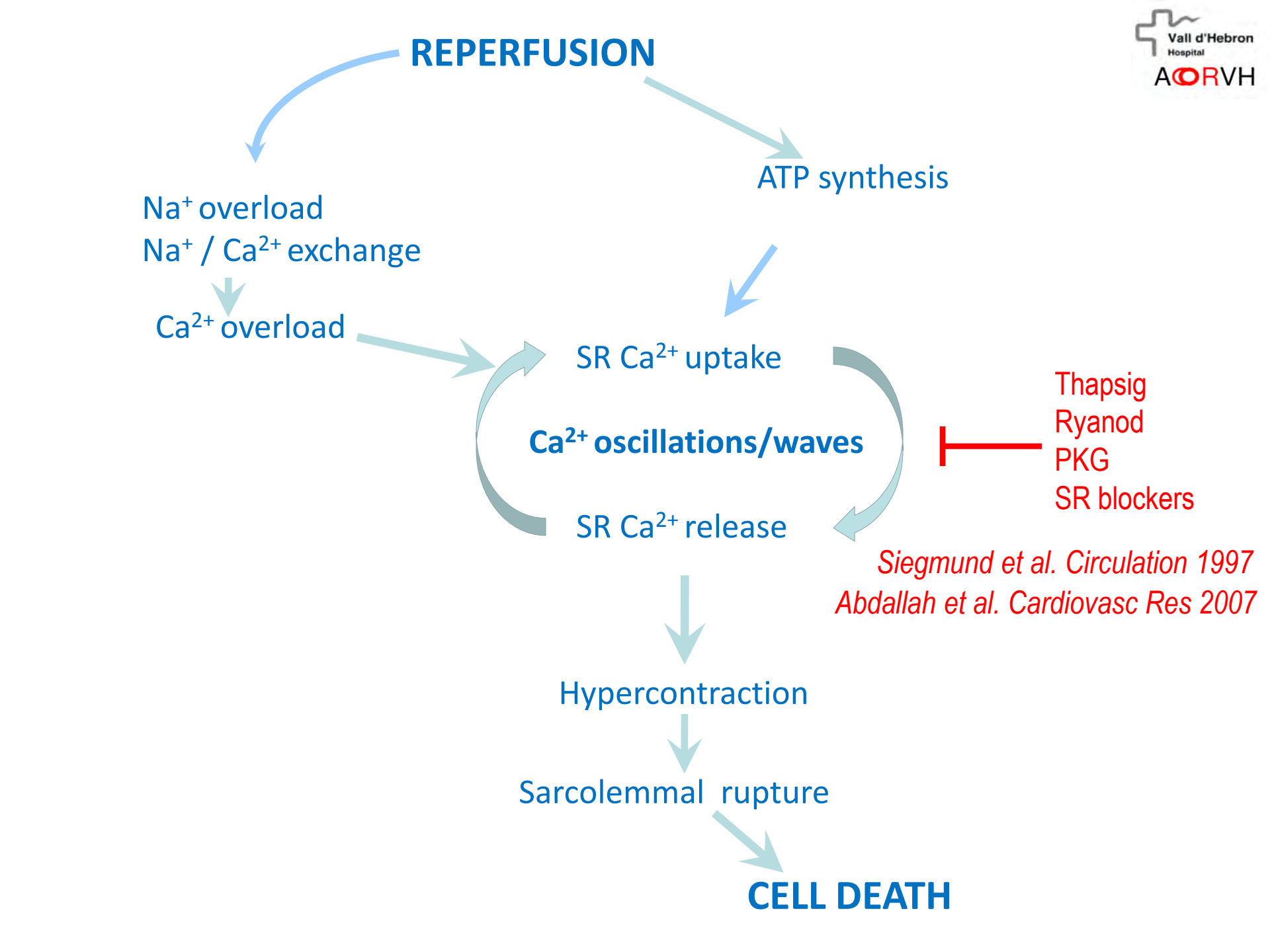
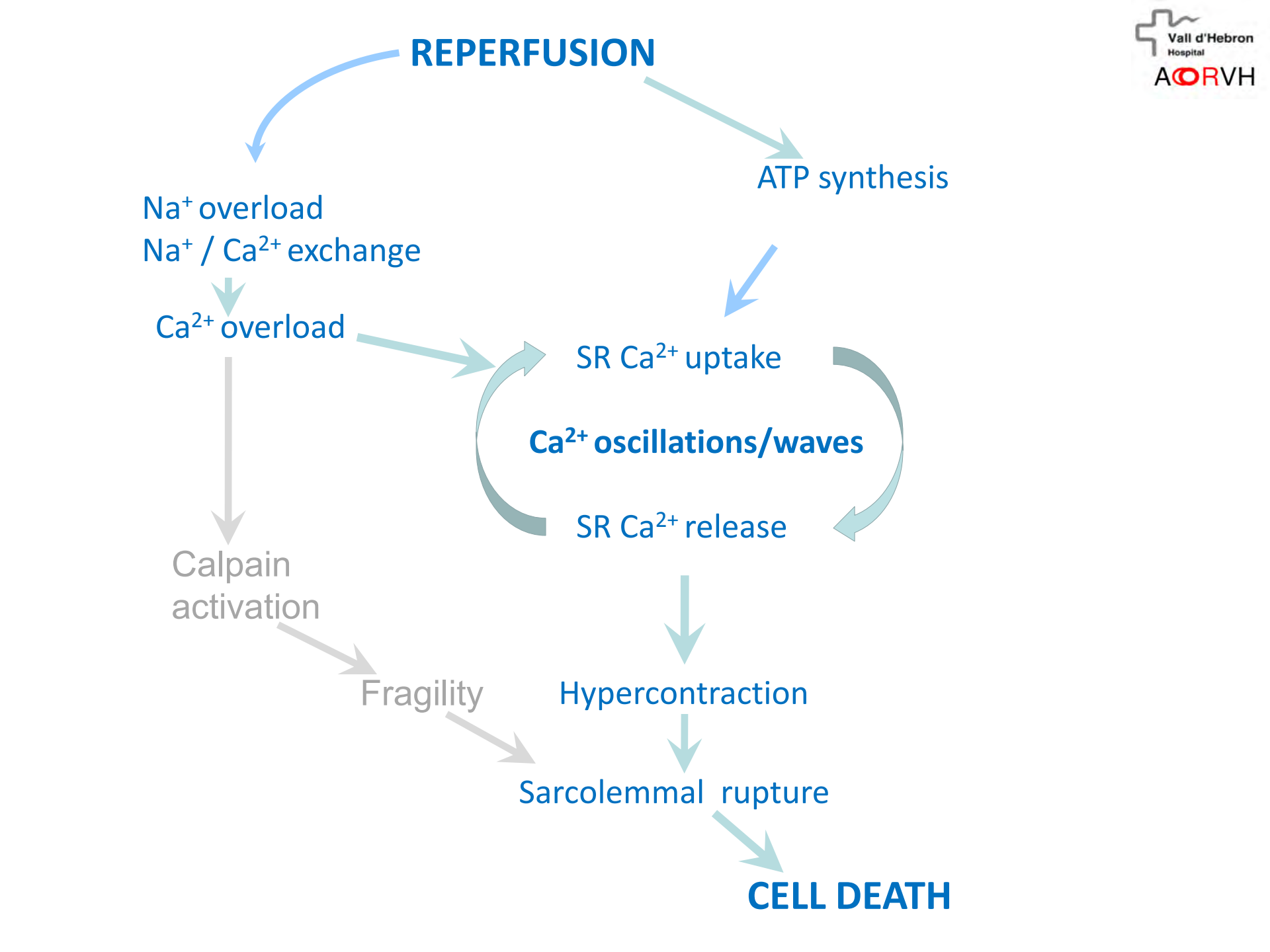


50 min LAD occlusion and four hours of reperfusion in the in situ pig heart.
SELECTIVE contractile blockade at the time of reperfusion
June 2011


REPERFUSION
ATP synthesis
Na+ overload
Na+ / Ca2+ exchange
Ca2+ overload
SR Ca2+ uptake
Ca2+ oscillations/waves
SR Ca2+ release
Thapsig
Ryanod
PKG
SR blockers
Siegmund et al. Circulation 1997
Abdallah et al. Cardiovasc Res 2007
Hypercontraction
Sarcolemmal rupture
CELL DEATH


REPERFUSION
ATP synthesis
Na+ overload
Na+ / Ca2+ exchange
Ca2+ overload
SR Ca2+ uptake
Ca2+ oscillations/waves
SR Ca2+ release
Calpain activation
Fragility
Hypercontraction
Sarcolemmal rupture
CELL DEATH

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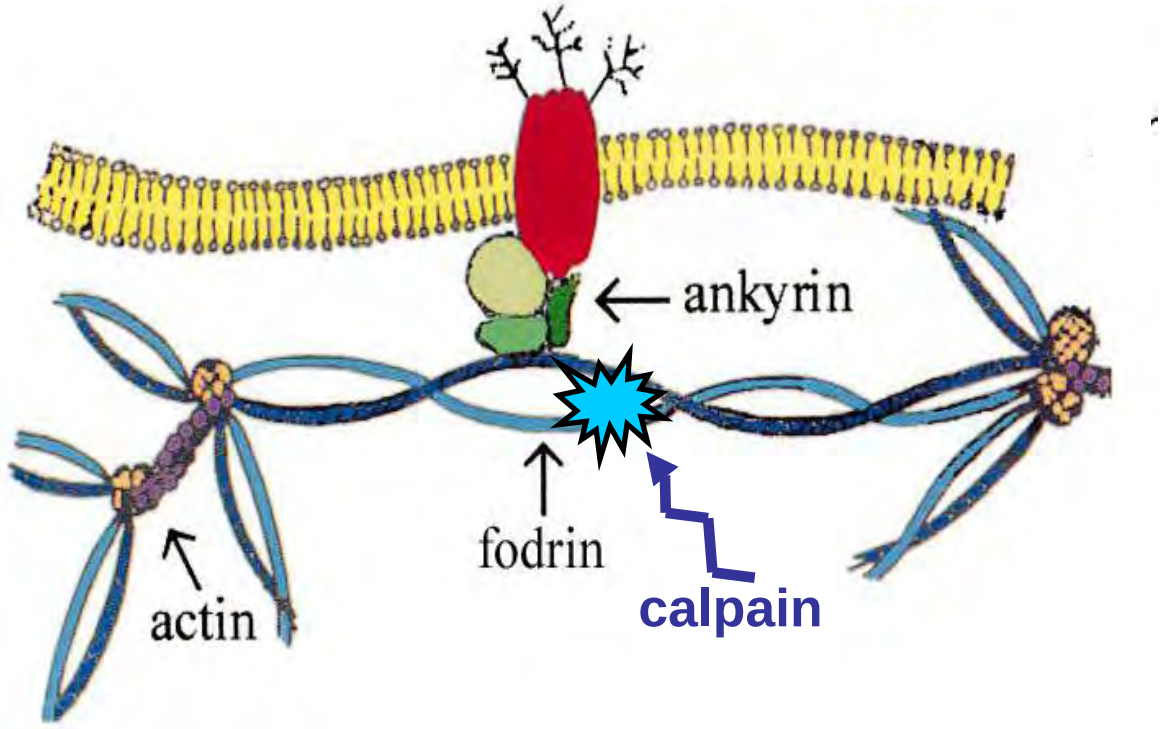
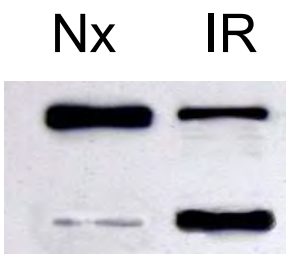
Na^+/K^+-ATPase





fodrin

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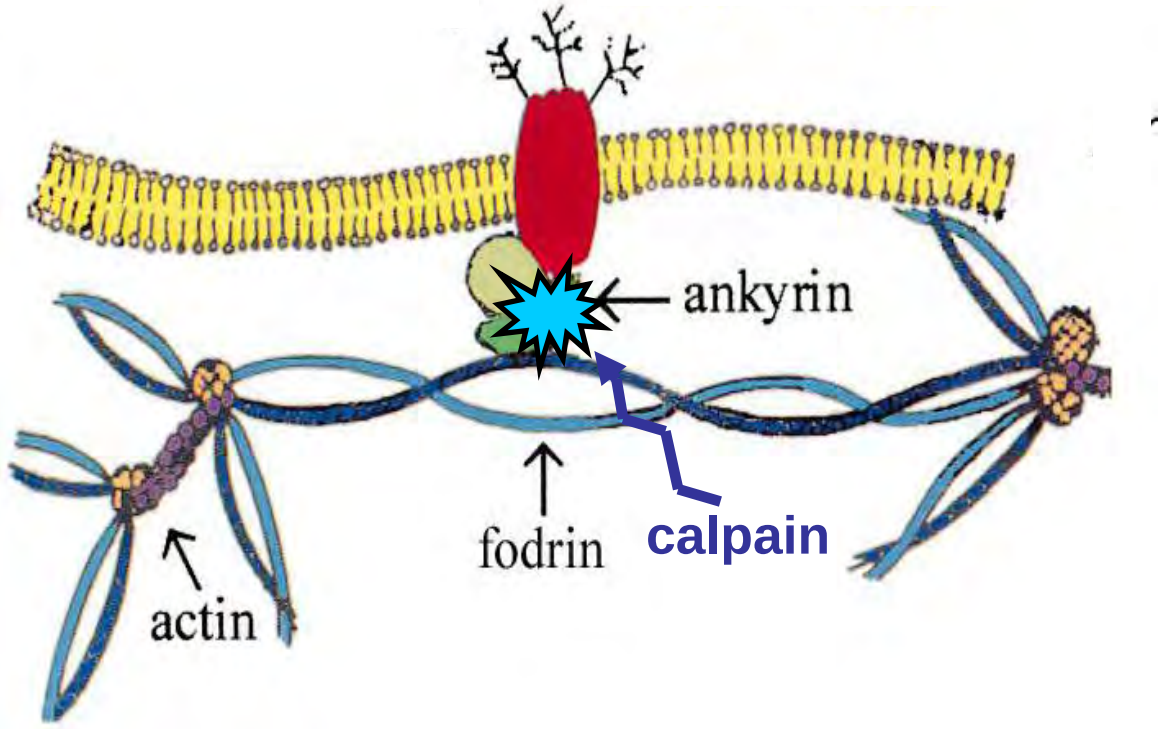
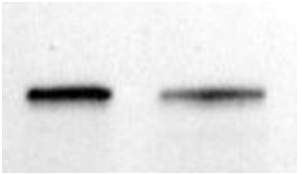
Na^+/K^+-ATPase

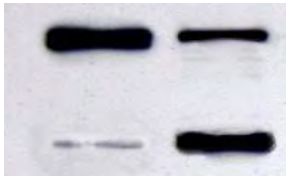


Nx IR

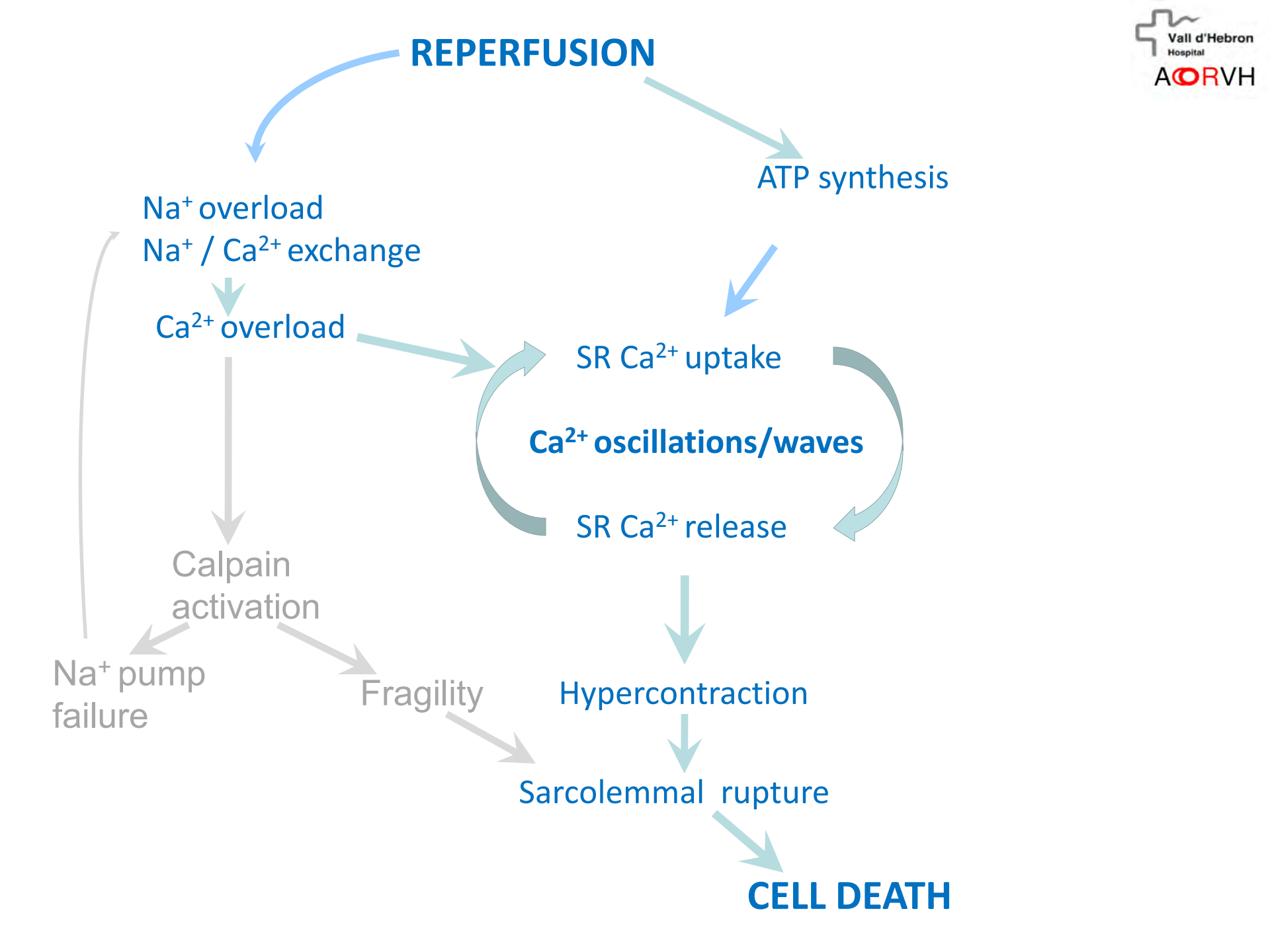


ankyrin

Nx IR



fodrin


REPERFUSION
ATP synthesis
Na+ overload
Na+ / Ca2+ exchange
Ca2+ overload
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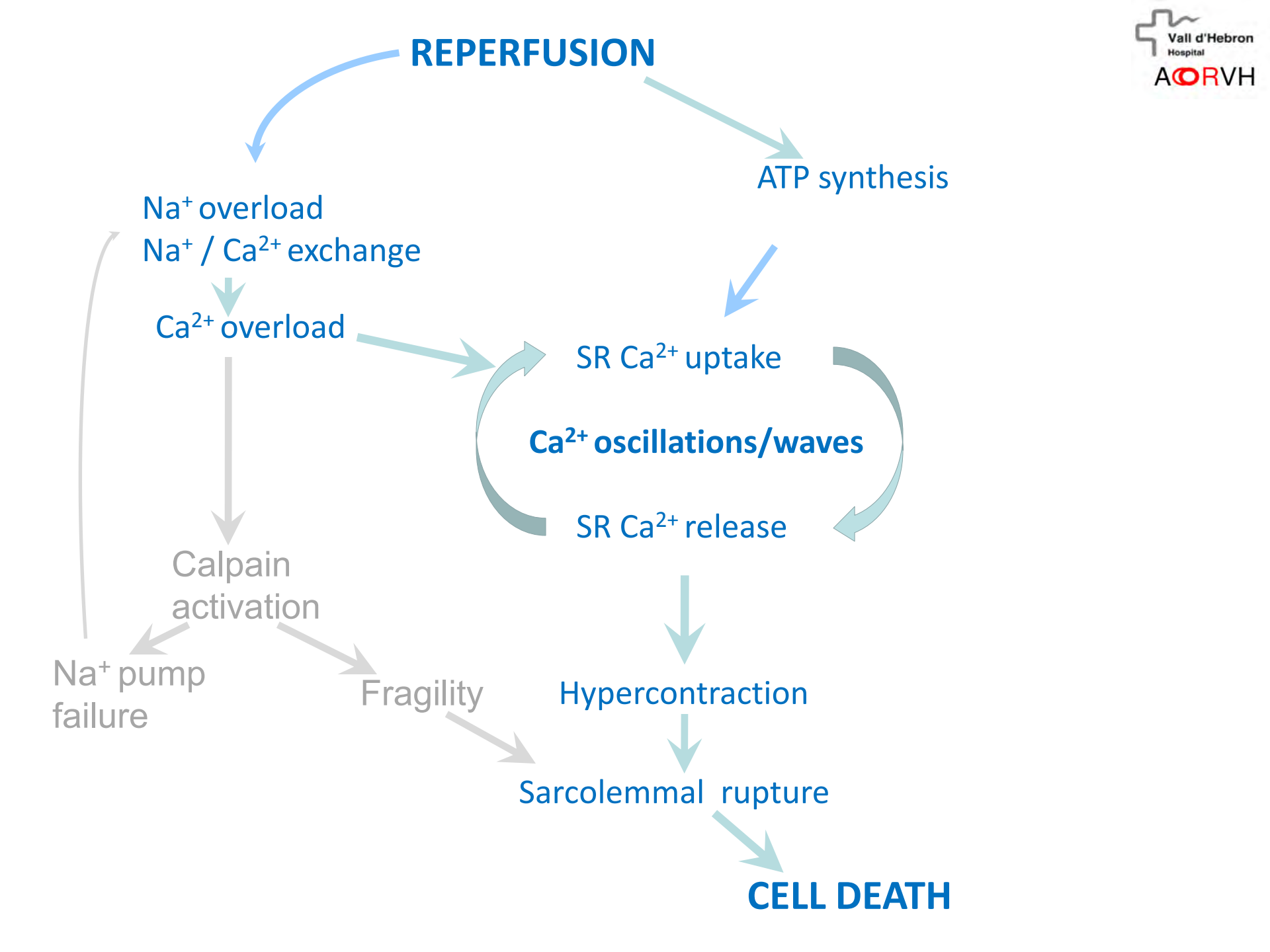
SR Ca2+ release
Calpain activation
Na+ pump failure
Fragility
Hypercontraction
Sarcolemmal rupture
CELL DEATH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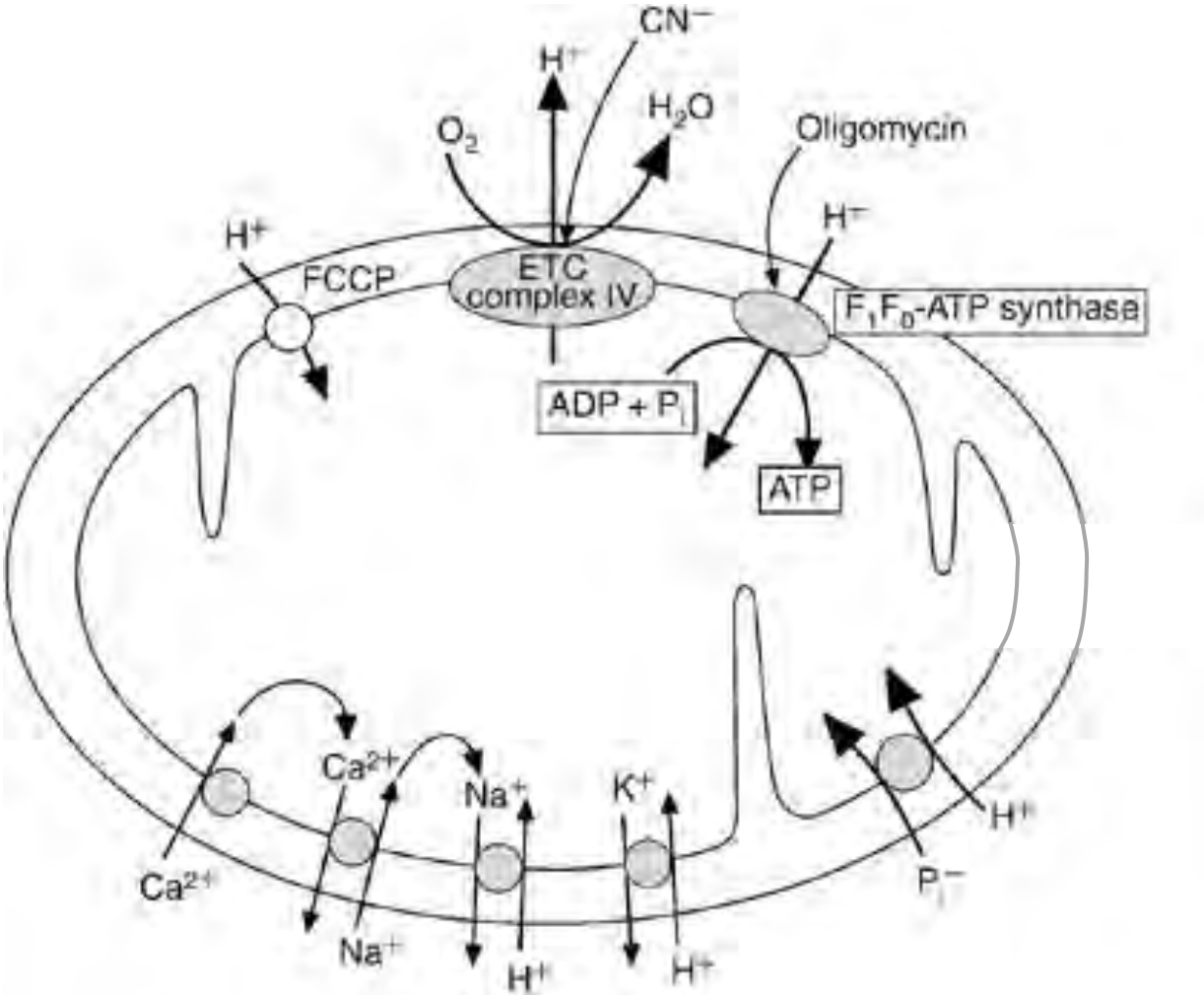
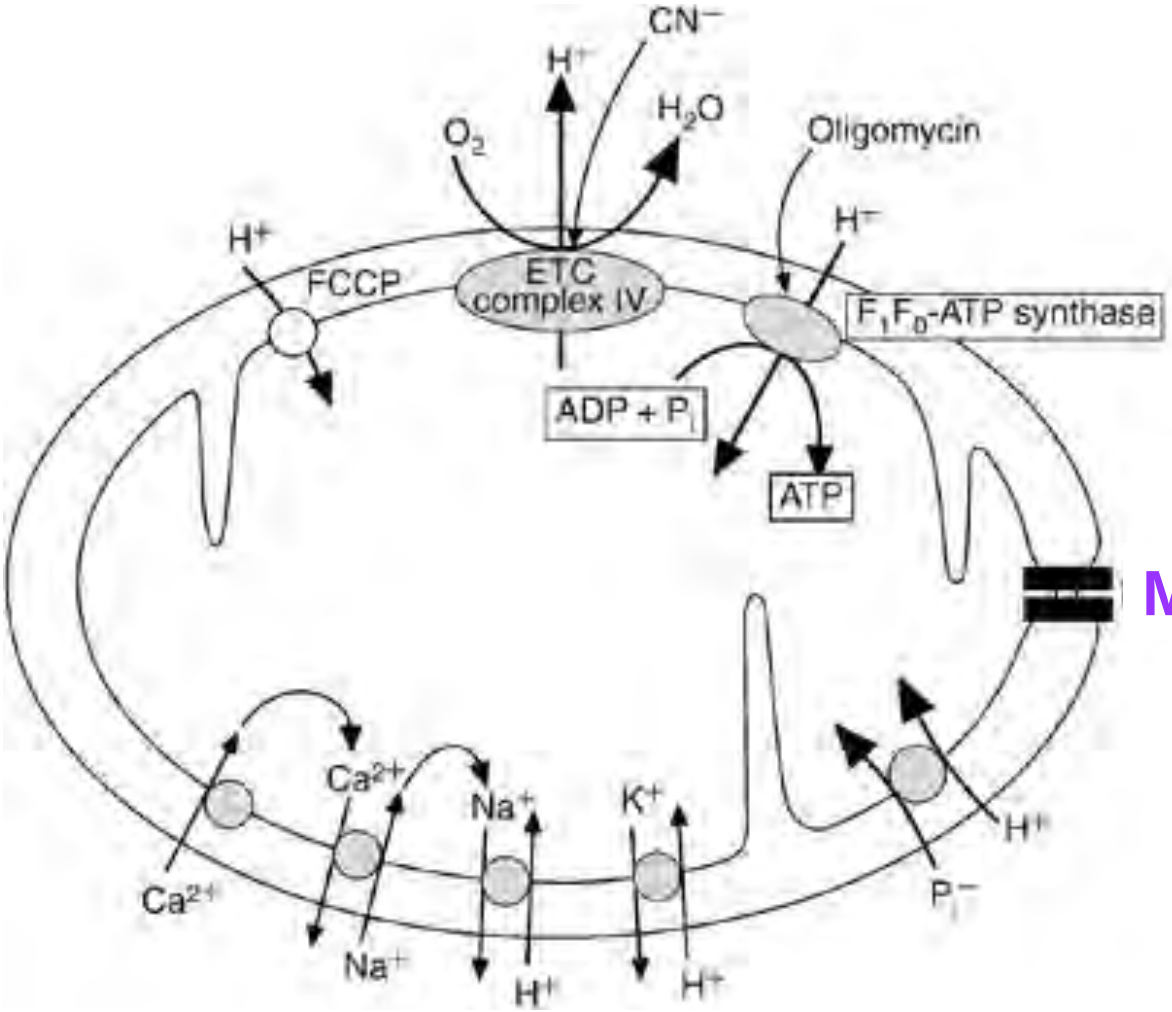
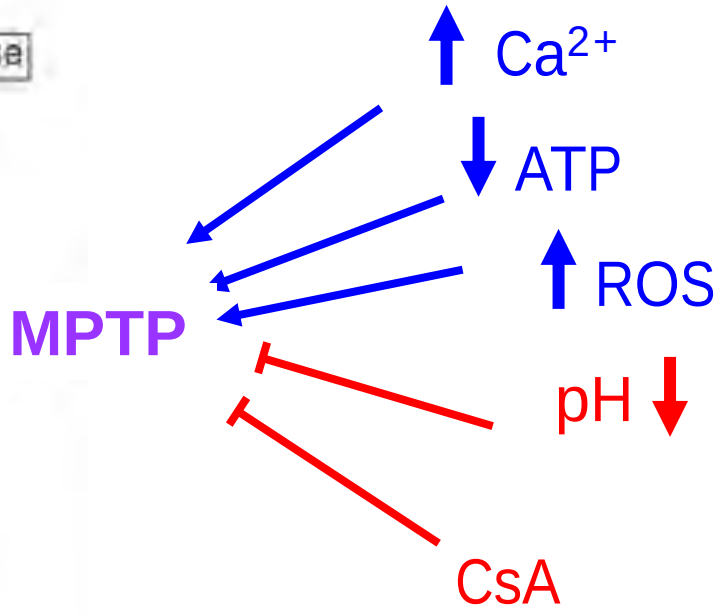


Balut C, Kidney International **73**, 226-232





Crompton
Halestrap
Griffiths
Bernerdi
DiLisa Weiss

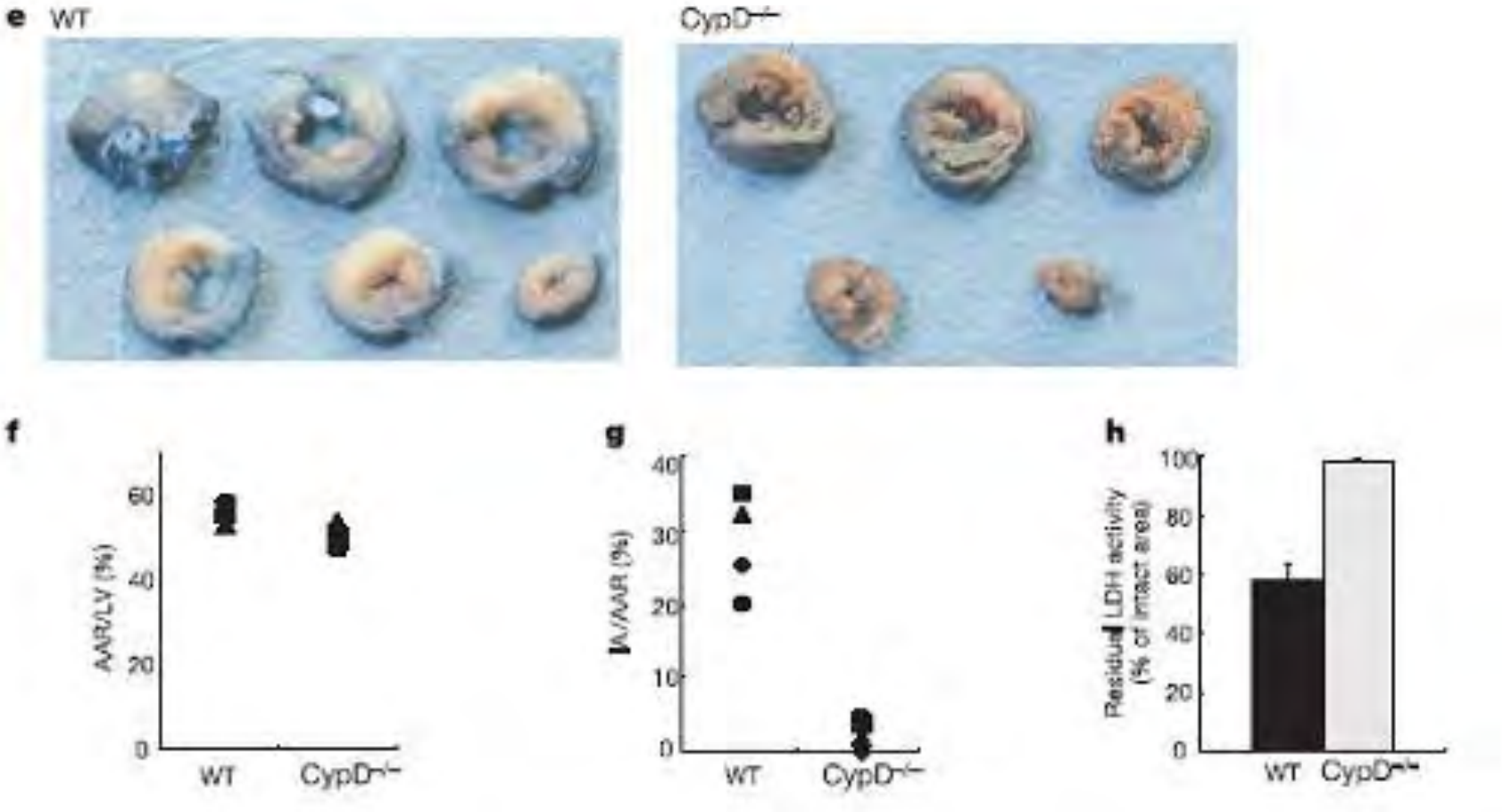
Balut C, Kidney International **73**, 226-232

Vall d'Hebron Hospital

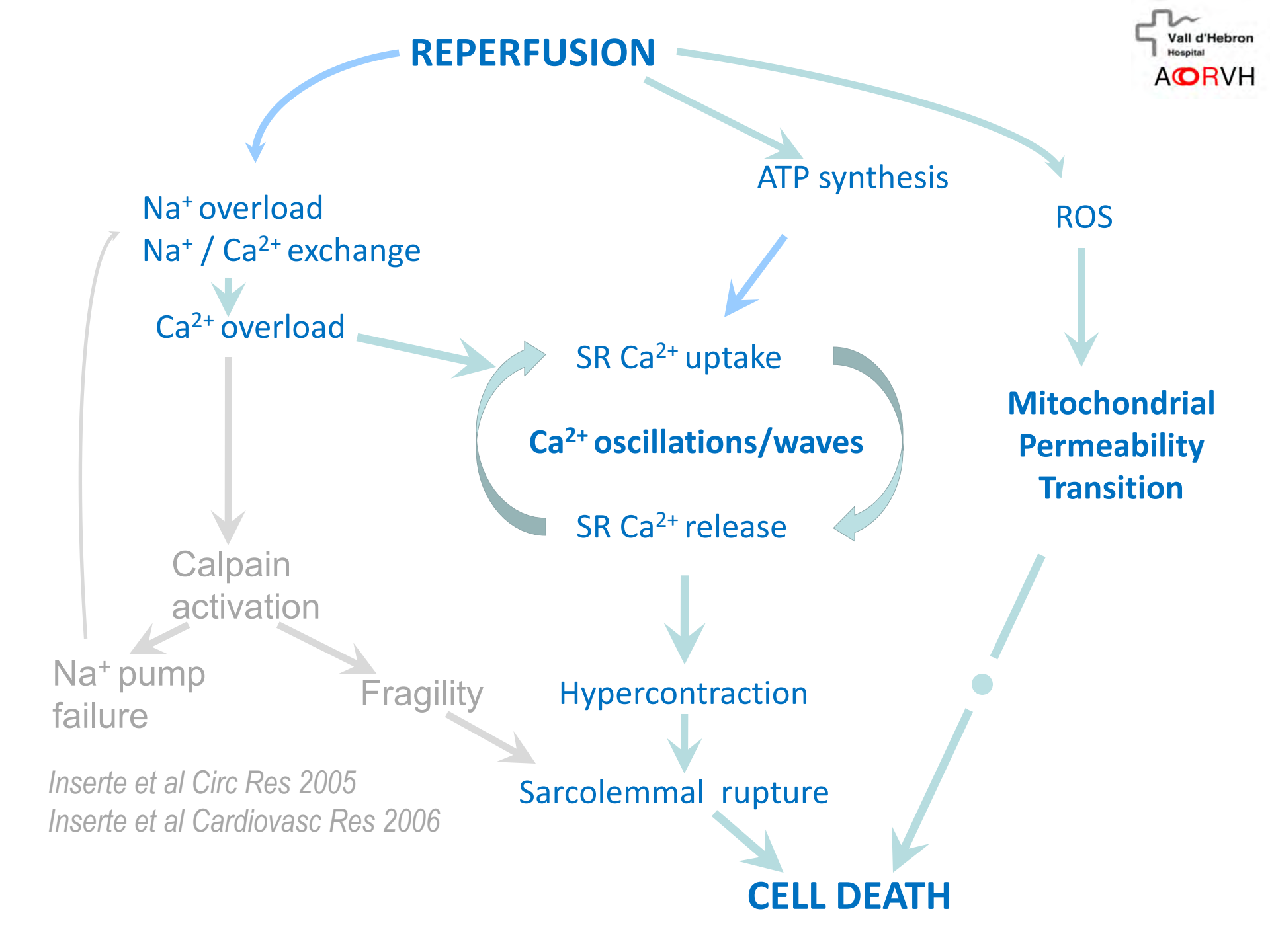
ACORVH

Loss of cyclophilin D reveals a critical role for mitochondrial permeability transition in cell death

Christopher P. Baines[1], Robert A. Kaiser[1], Nicole H. Purcell[1], N. Scott Blair[1], Hanna Osinska[1], Michael A. Hambleton[2], Eric W. Brunskill[3], M. Richard Sayen[4], Roberta A. Gottlieb[4], Gerald W. Dorn II[3], Jeffrey Robbins[1] & Jeffery D. Molkentin[1]



Nature 2005; 434:655 and 658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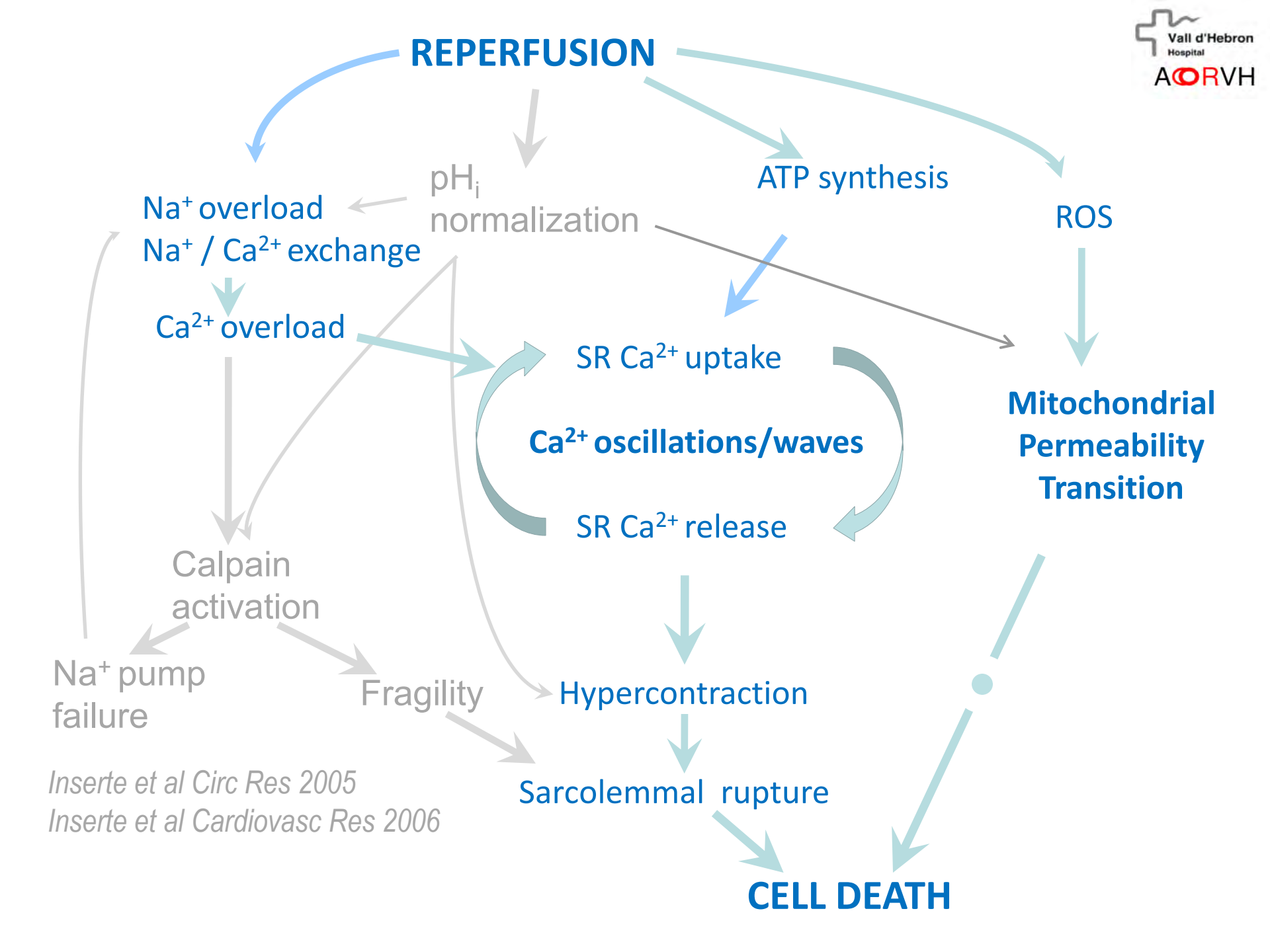
Sarcolemmal rupture
CELL DEATH
Inserte et al Circ Res 2005
Inserte et al Cardiovasc Res 2006


REPERFUSION
Vall d'Hebron Hospital
ACORVH
pH$_i$ normalization
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Inserte et al Circ Res 2005
Inserte et al Cardiovasc Res 2006

Vall d'Hebron Hospital
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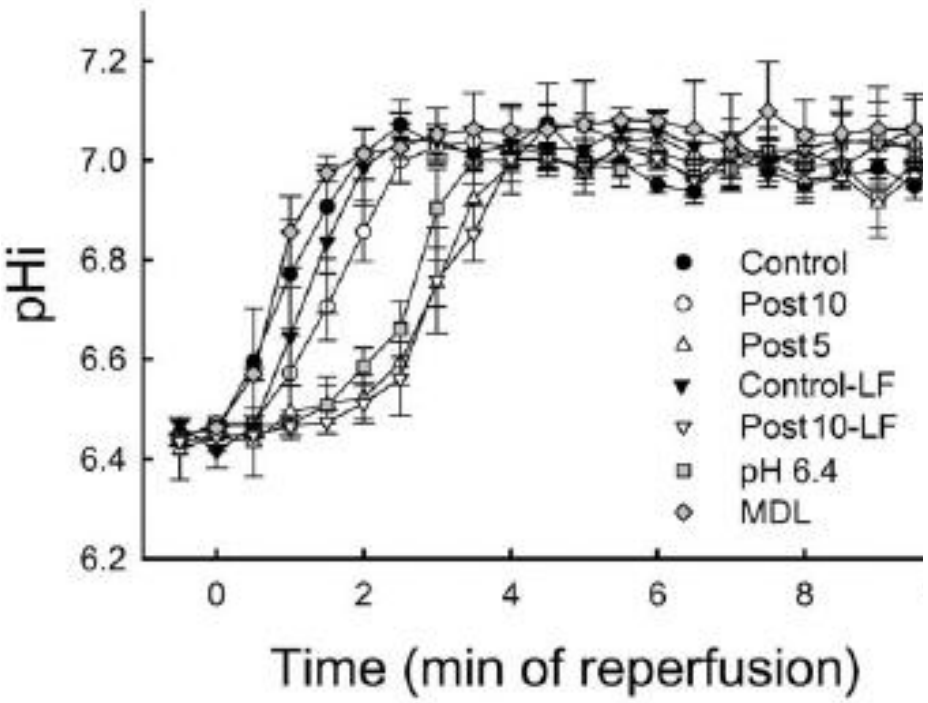
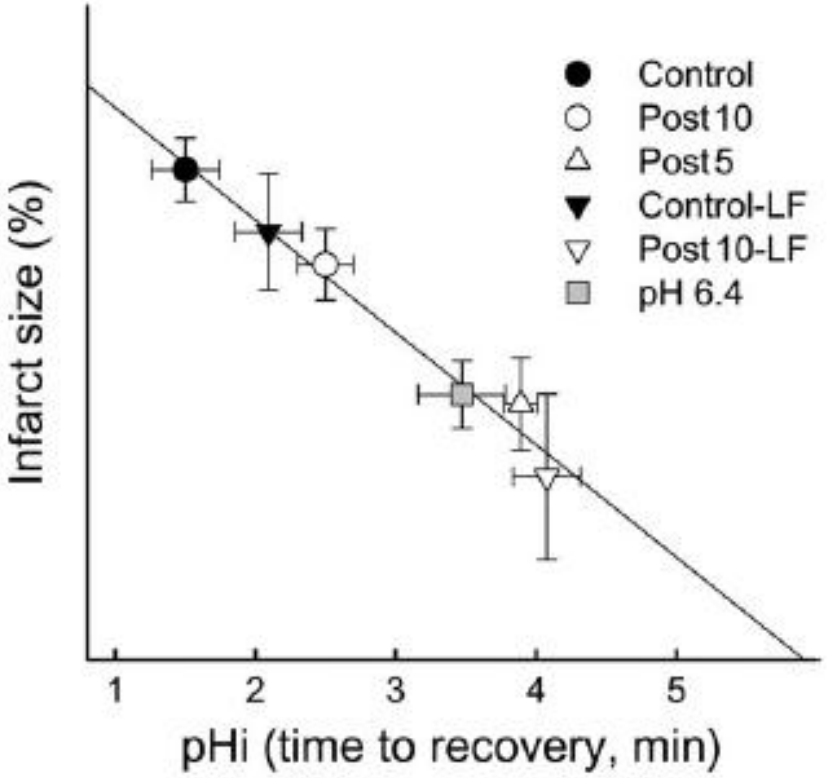
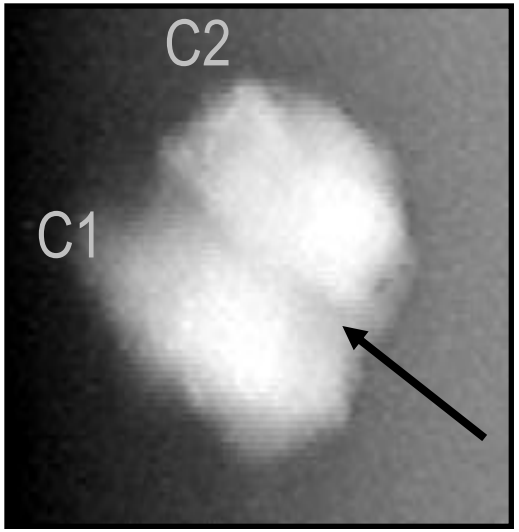
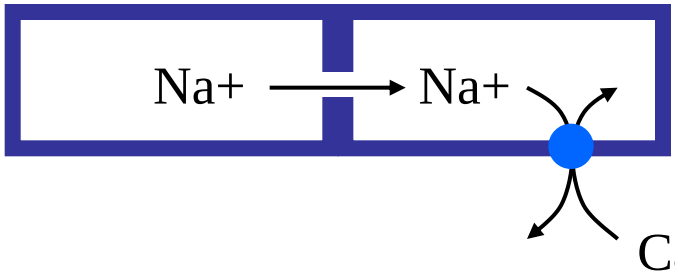
Delay in pHi recovery explains postconditioning protection

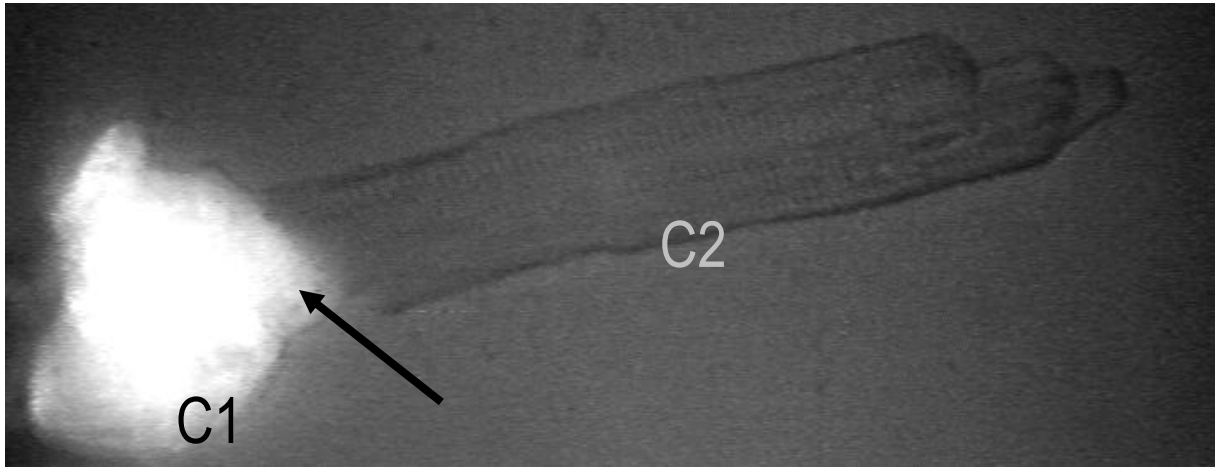




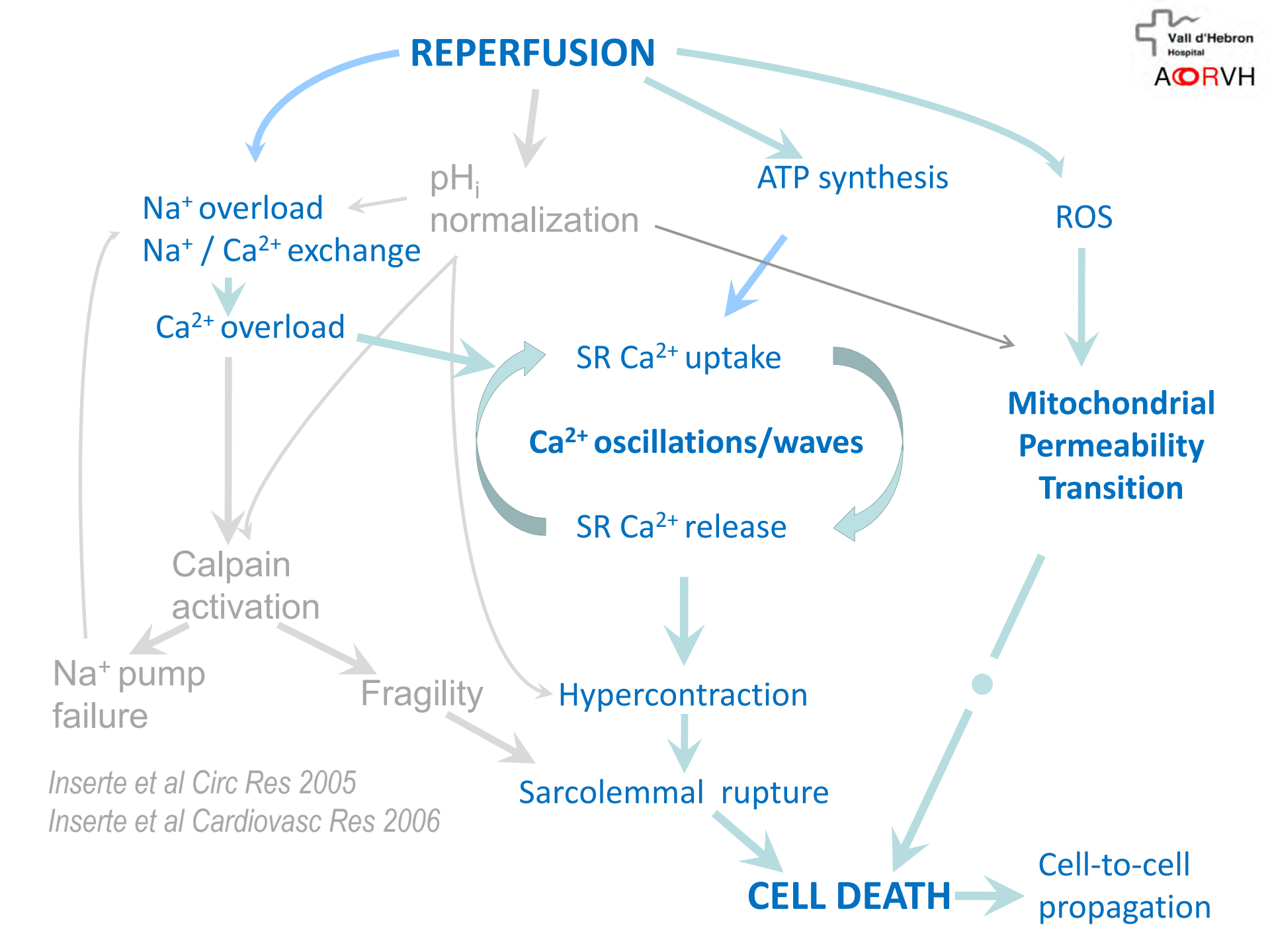
Inserte et al. Cardiovasc Res 2008







Garcia-Dorado et al. Circulation1997
Ruiz-Meana et al. Circ Res 1999


REPERFUSION
Vall d'Hebron Hospital
ACORVH
pHi normalization
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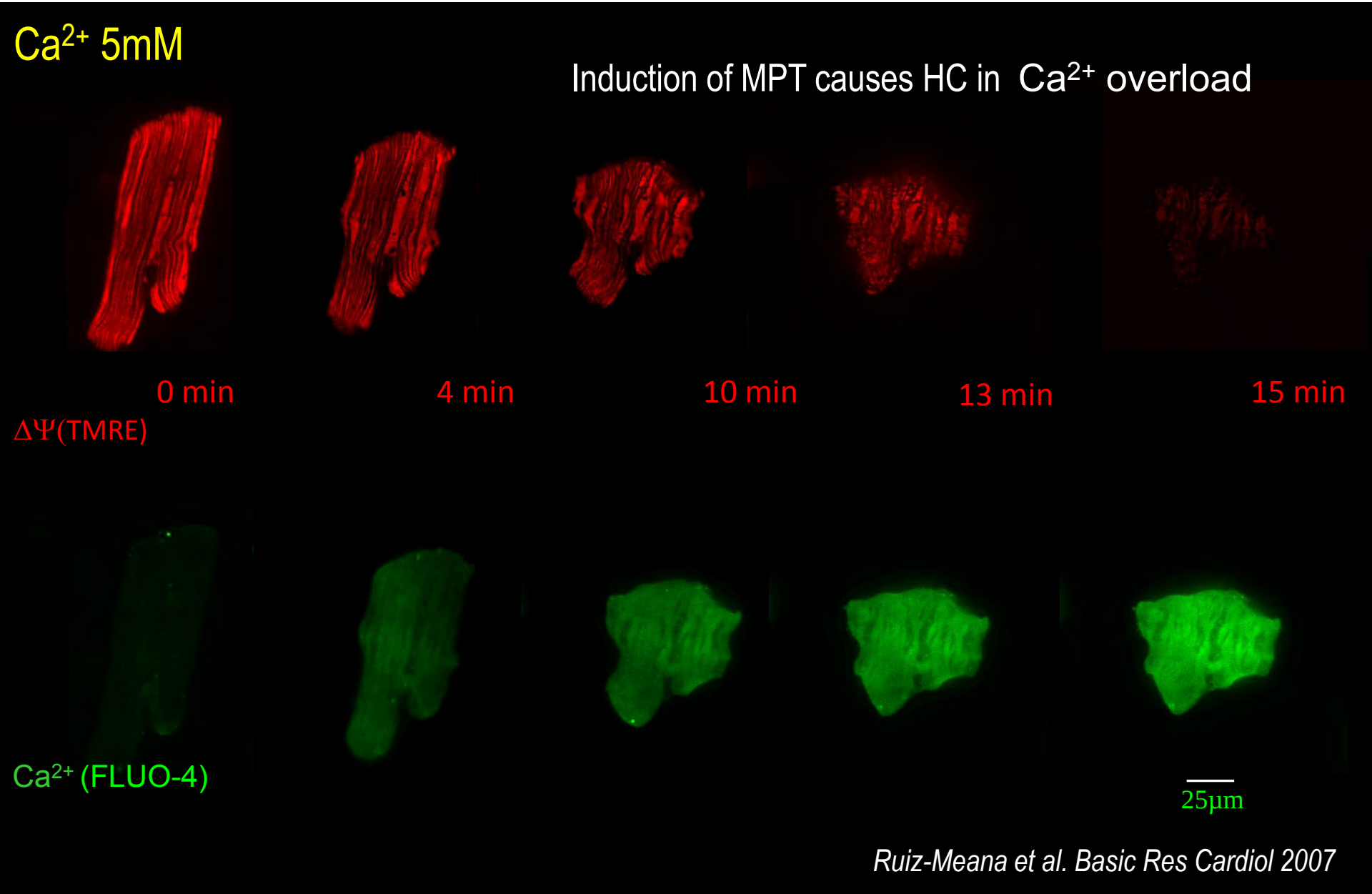
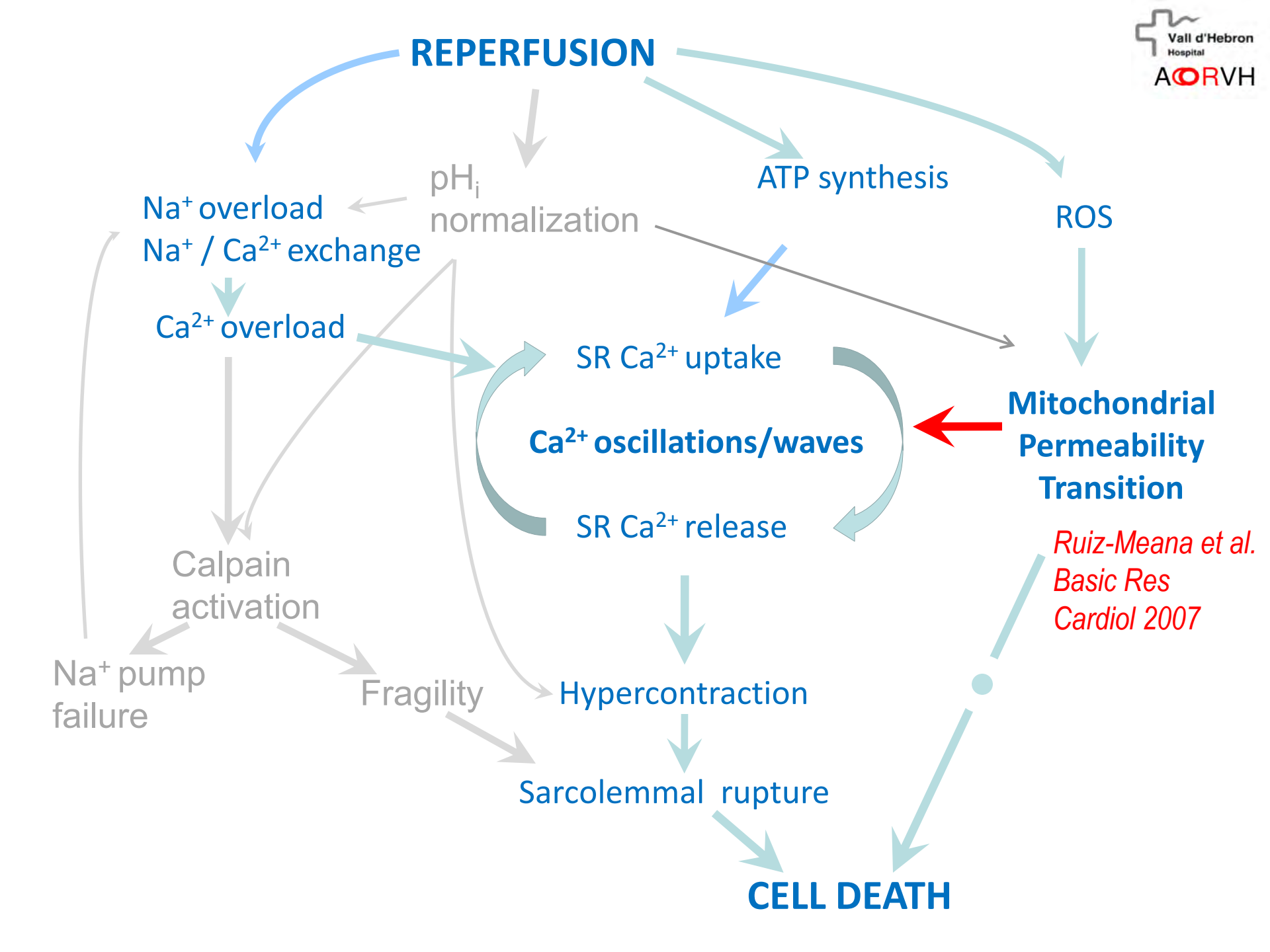
Sarcolemmal rupture
CELL DEATH
Cell-to-cell propagation
Inserte et al Circ Res 2005
Inserte et al Cardiovasc Res 2006

Induction of MPT in Ca^{2+} overloaded cells causes hypercontracture:



Ruiz-Meana et al. Basic Res Cardiol 2007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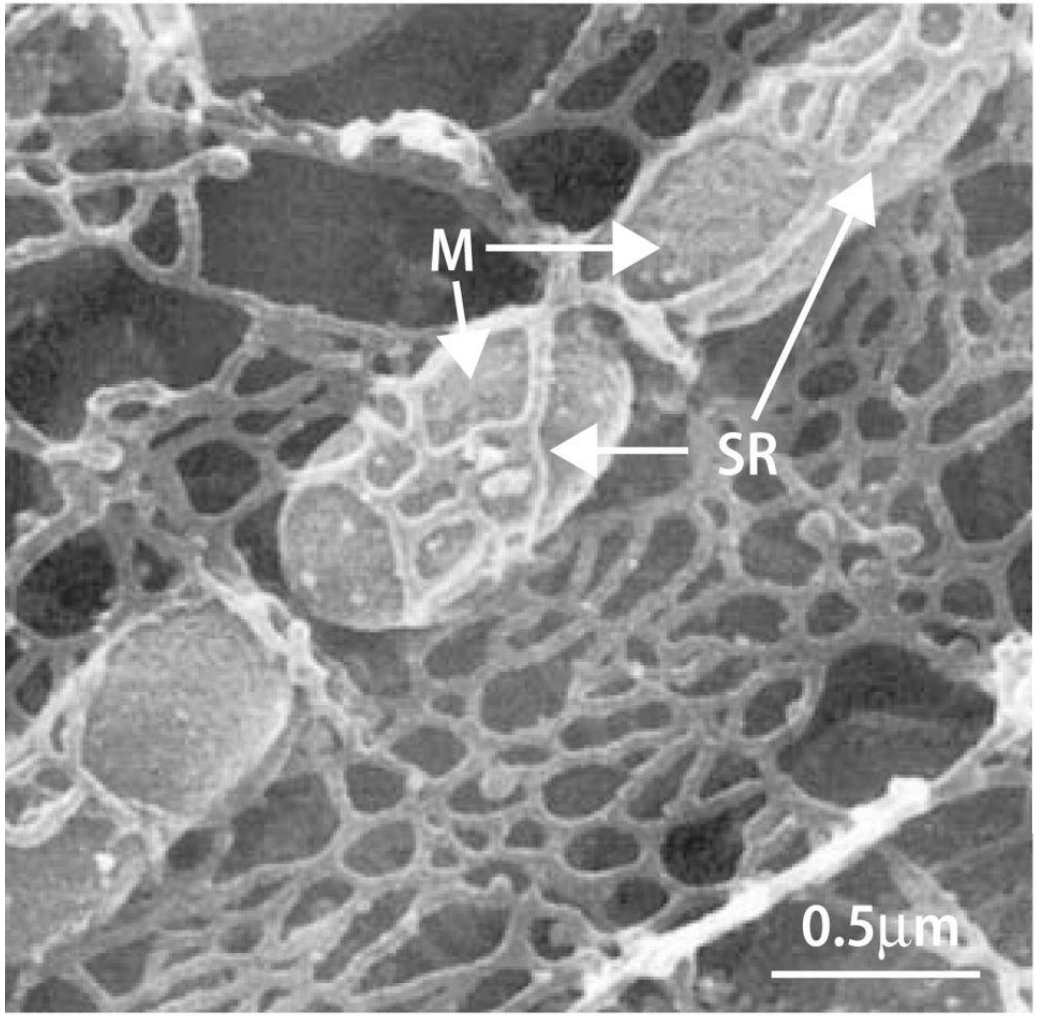
CELL DEATH

SR-mitochondria connection through Ca^{2+} microdomains



Yoshikane H. J Submicrosc Cytol 1986

Transfer and Tunneling of Ca^{2+} from Sarcoplasmic Reticulum to Mitochondria in Skeletal Muscle*

Received for publication, May 6, 2005, and in revised form, September 6, 2005 Published, JBC Papers in Press, October 10, 2005, DOI 10.1074/jbc.M505024200

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J Biol Chem 2006;281:1547

Physical Coupling Supports the Local Ca^{2+} Transfer between Sarcoplasmic Reticulum Subdomains and the Mitochondria in Heart Muscle*[S]

Received for publication, May 2, 2008, and in revised form, August 19, 2008 Published, JBC Papers in Press, September 12, 2008, DOI 10.1074/jbc.M803385200

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From the Department of Pathology, Anatomy and Cell Biology, Thomas Jefferson University, Philadelphia, Pennsylvania 19107

J Biol Chem 2008;283:32771

Mitofusin 2 tethers endoplasmic reticulum to mitochondria

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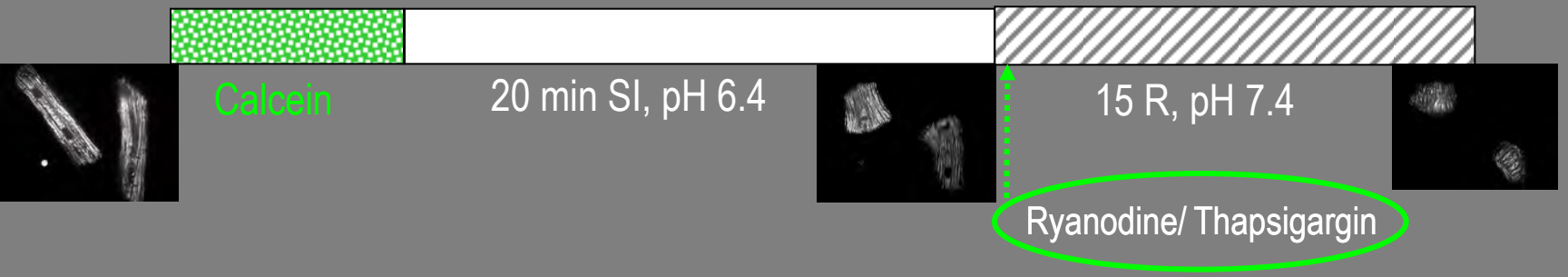
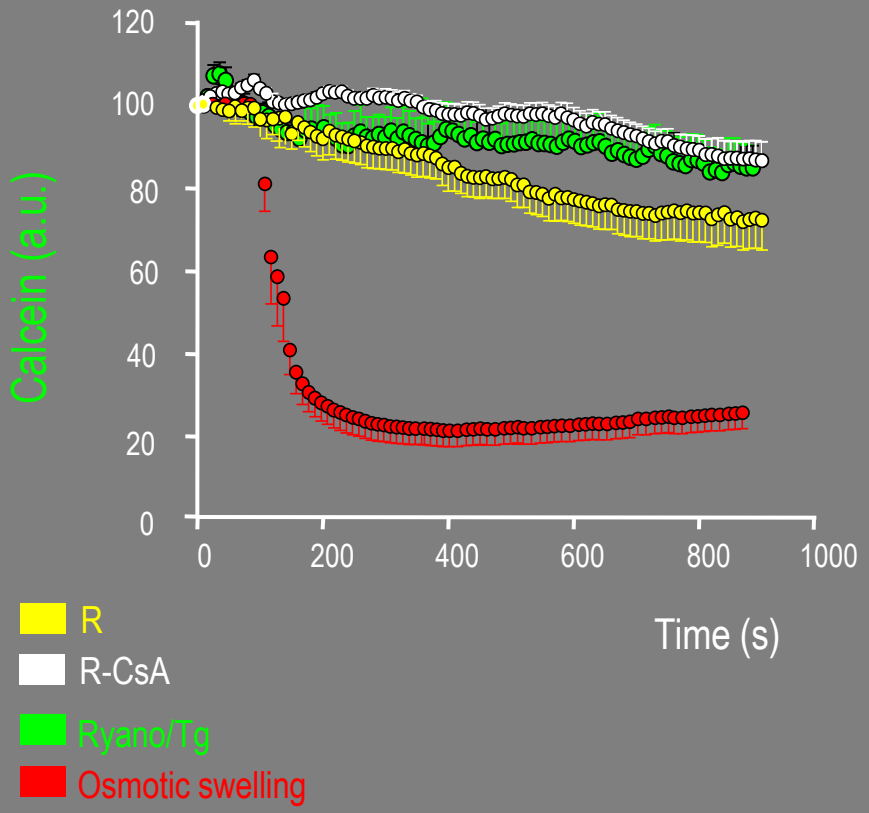
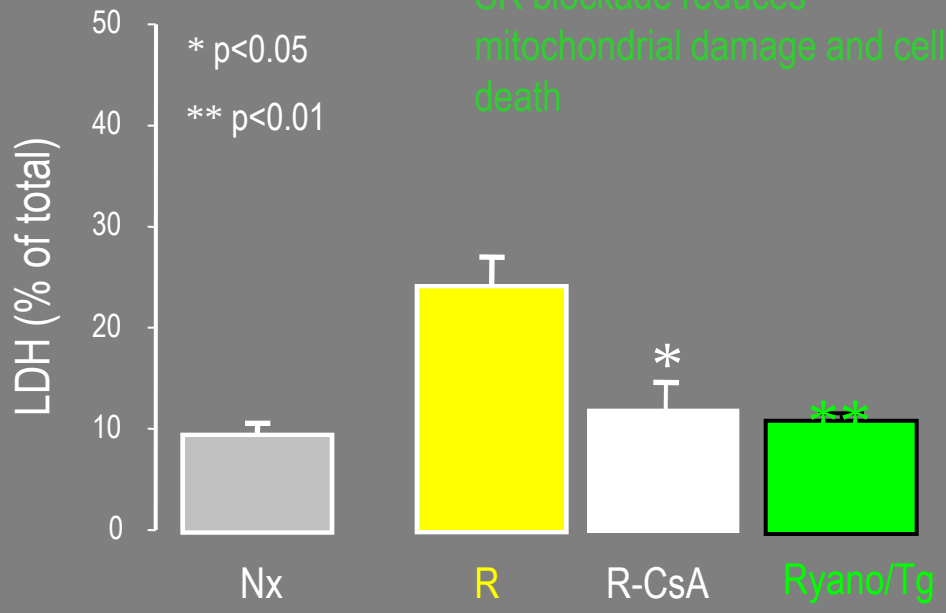
Nature 2008;456:605

SR blockade prevents MPT in reperfused myocytes

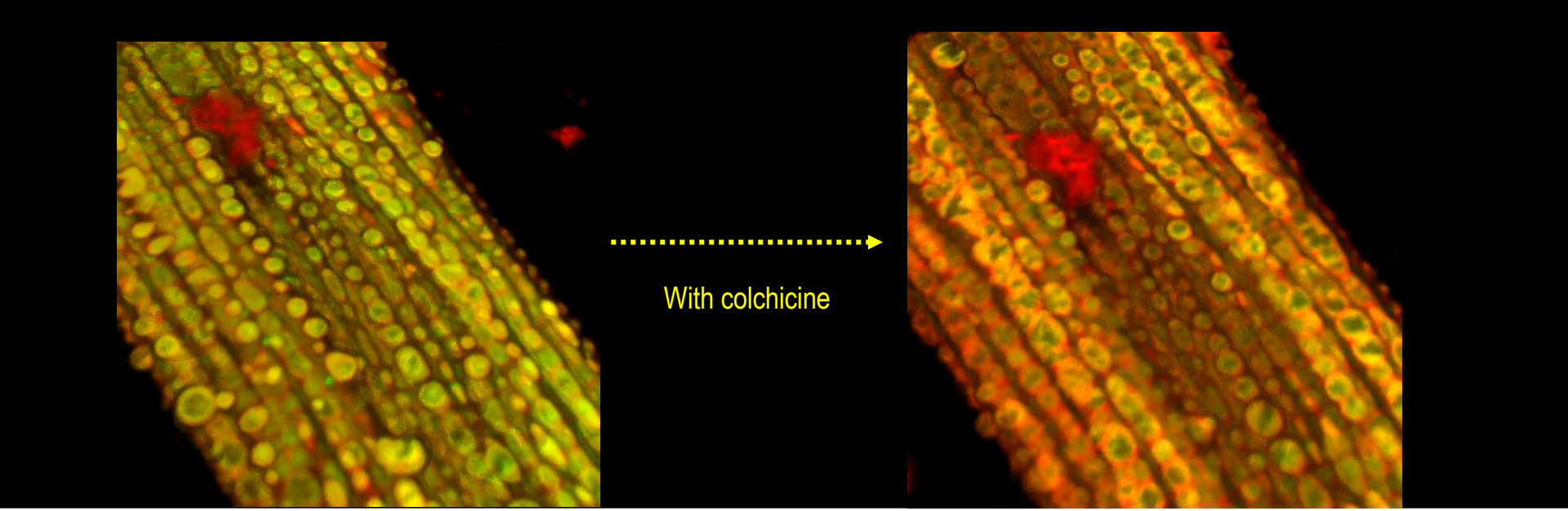


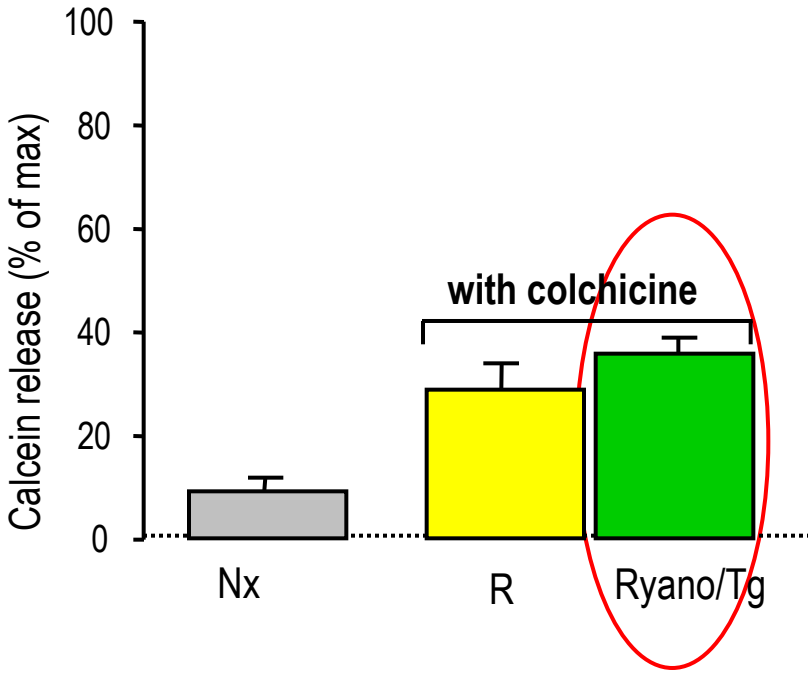


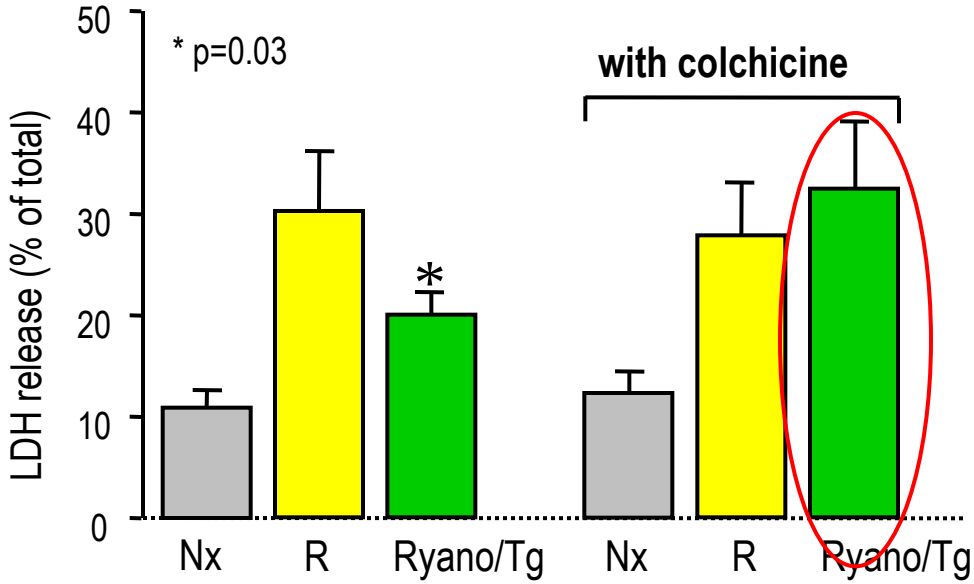
SR blockade reduces mitochondrial damage and cell death



Ruiz-Meana et al. Am J Physiol 2009







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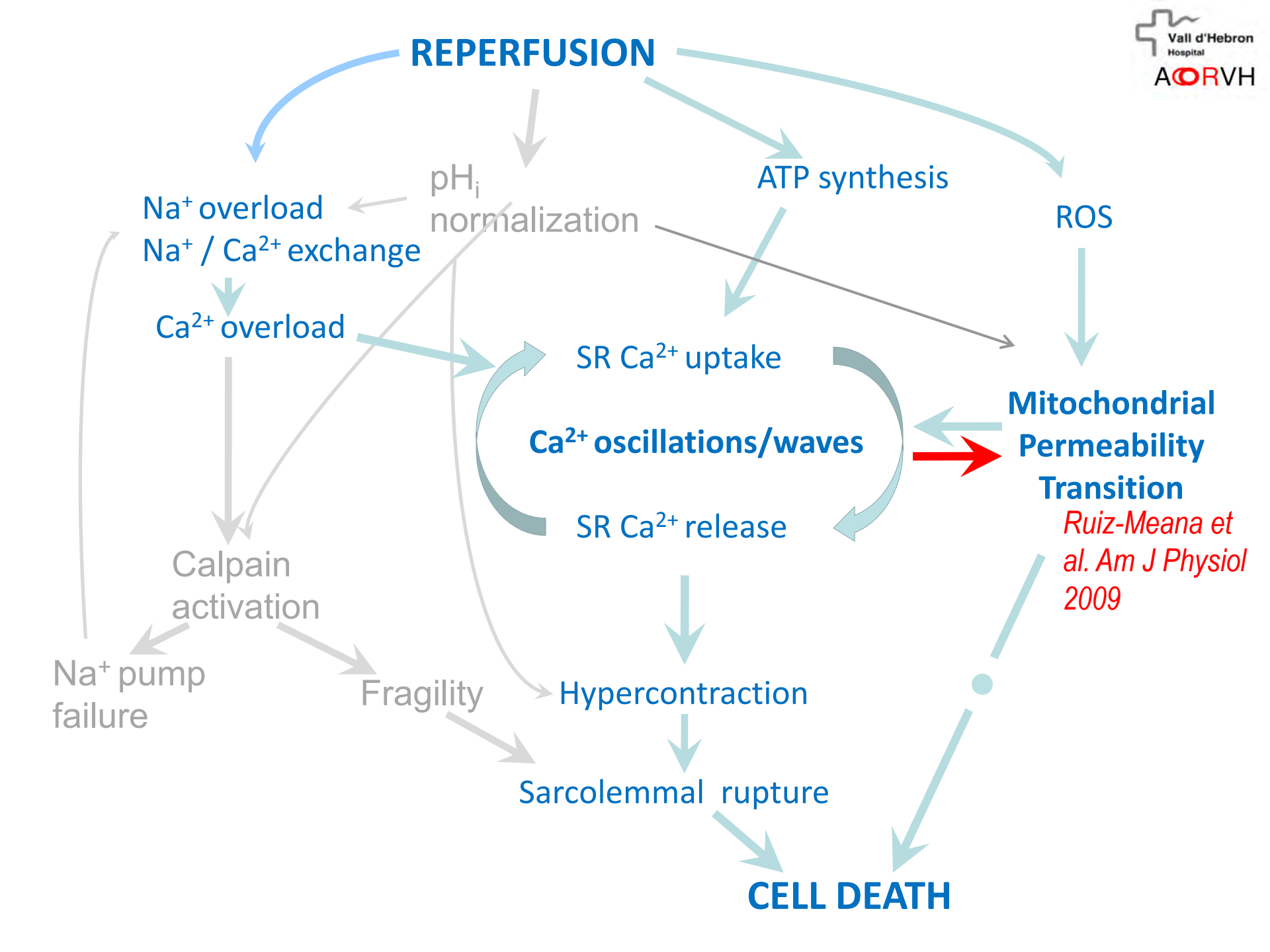
REPERFUSION
Vall d'Hebron Hospital
ACORVH
pH$_i$ normalization
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Ruiz-Meana et al. Am J Physiol 2009
Calpain activation
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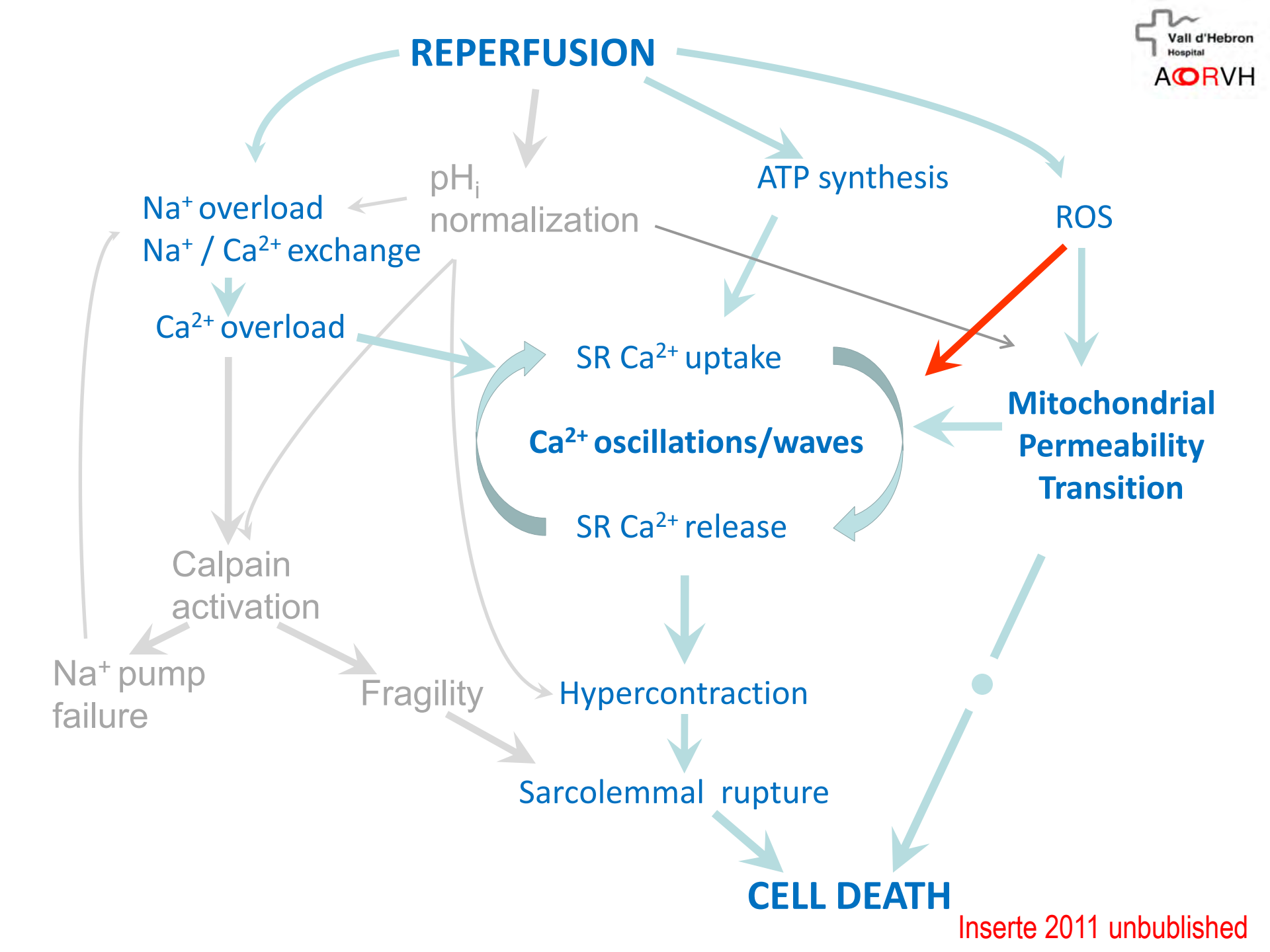
CELL DEATH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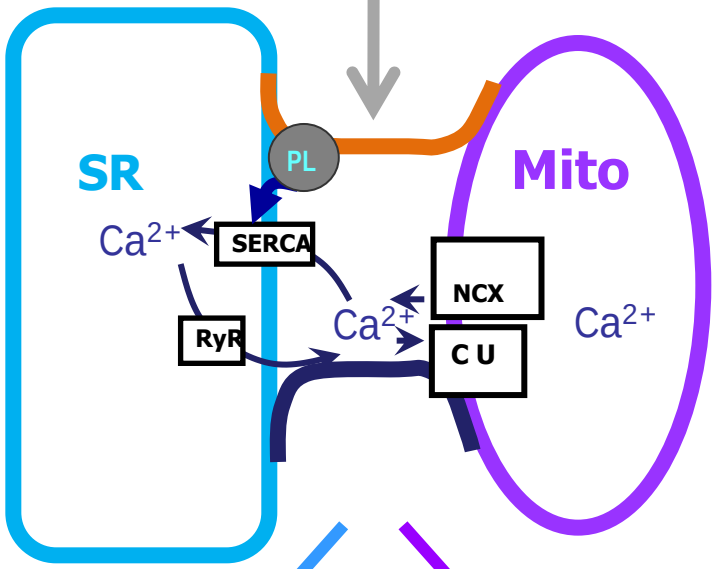
CELL DEATH
Inserte 2011 unbublished

REPERFUSION

Ca^{2+} overload ATP pH correction ROS



Ca^{2+} oscillations ⇄ MPT

CELL DEATH

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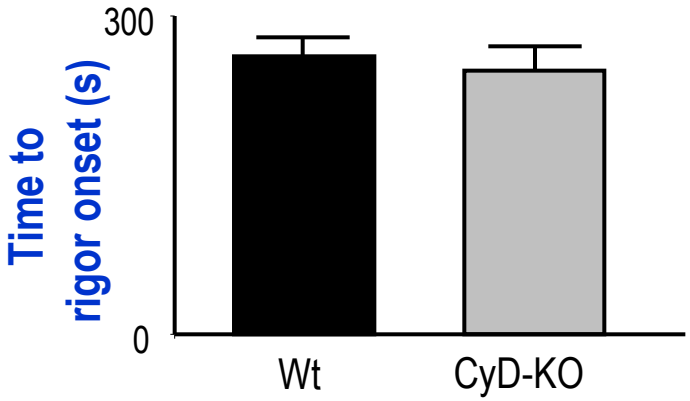
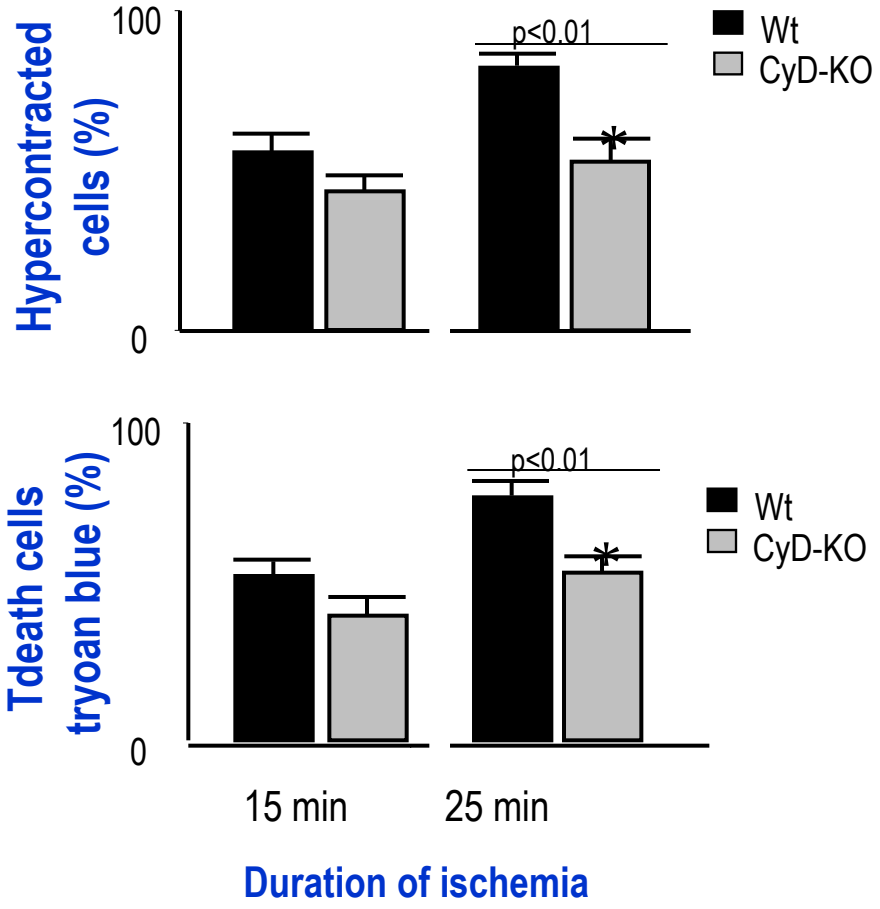
Ischemia and Reperfusion. Basic concepts

1) Myocardial Ischemia
2) Reperfusion salvage and reperfusion injury
3) Basic molecular mechanisms
4) **Integrative view**
5) Translational perspective
6) Conclusion

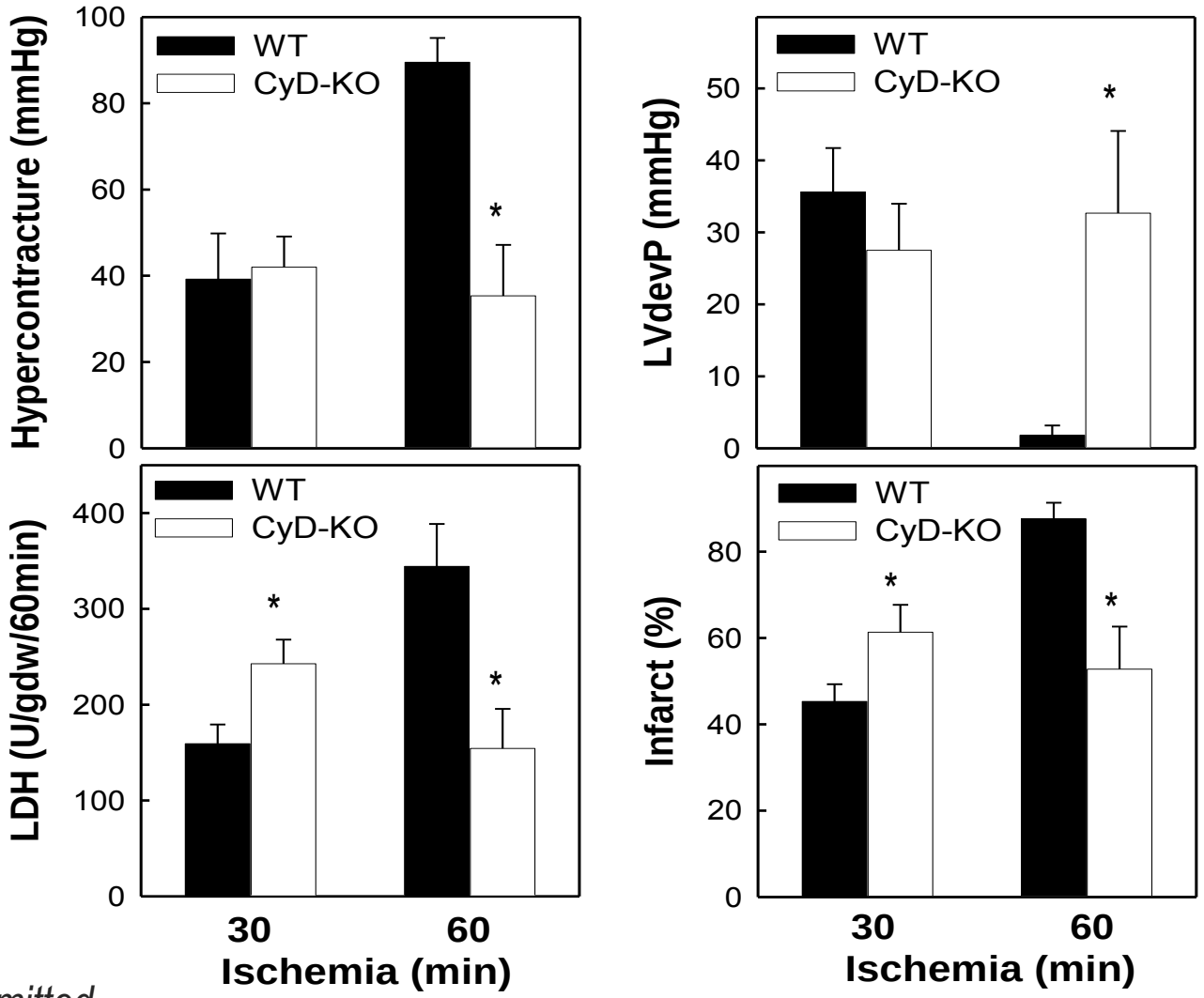
Effect of CyD abblation on **hypercontracture and cell death** during reperfusion after short (15 min) or more prolonged ischemia in **adult cardiomyocytes**





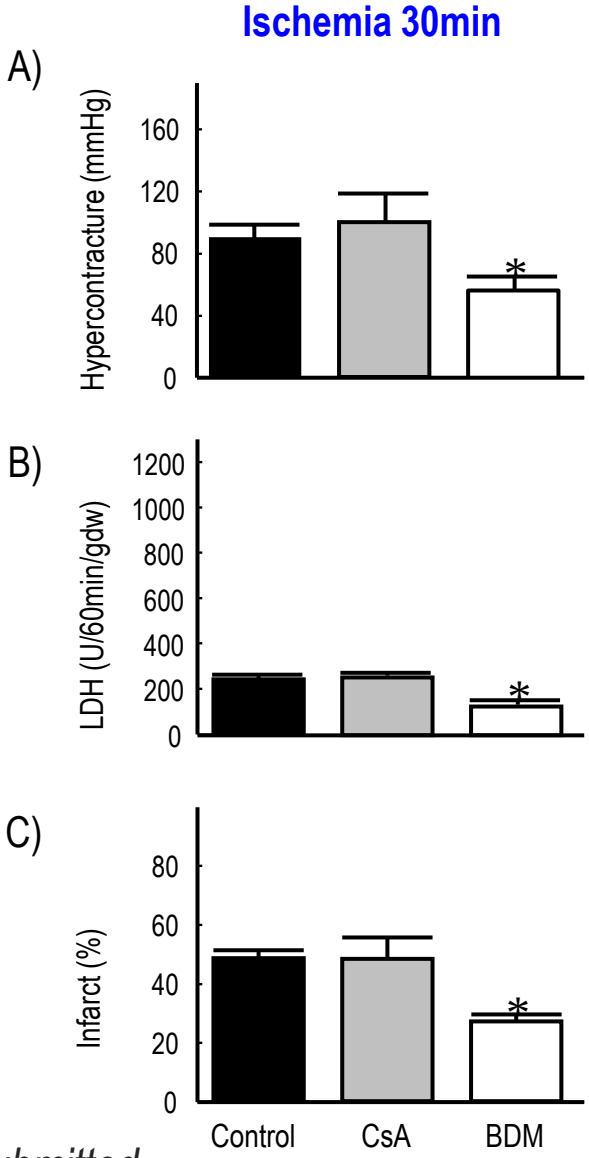
Ruiz-Meana 2010, submitted

Effect of CyD abblation on **hypercontracture, functional recovery and cell death** during reperfusion after short (30 min) or more prolonged (60 min) ischemia in **perfused mice hearts**



Ruiz-Meana 2010, submitted

Effect of **MPT inhibition and contractile blockade** infarct size after short (30 min) or more prolonged (50 min) ischemia in **perfused rat hearts**



Ruiz-Meana 2010, submitted

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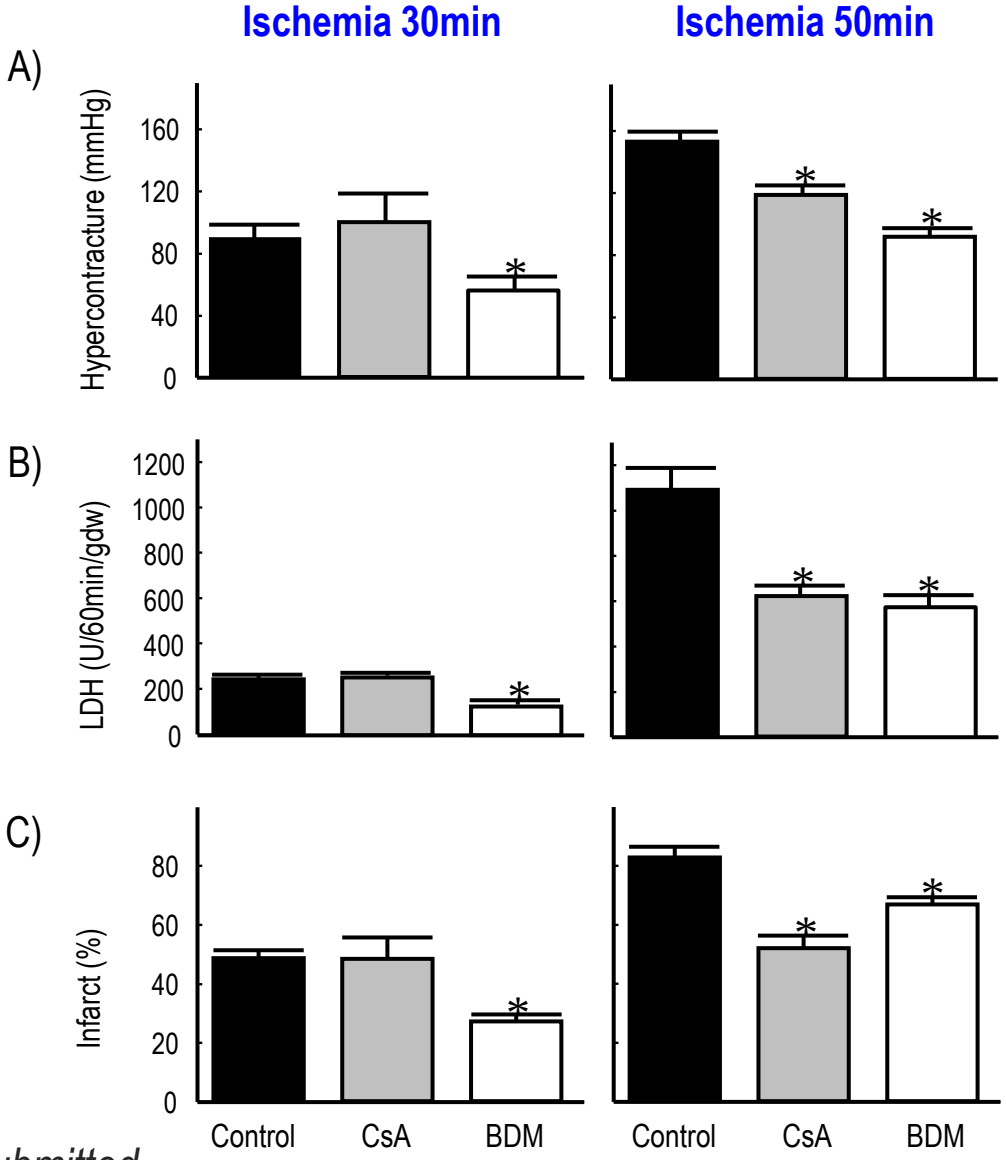

Ruiz-Meana 2010, submitted

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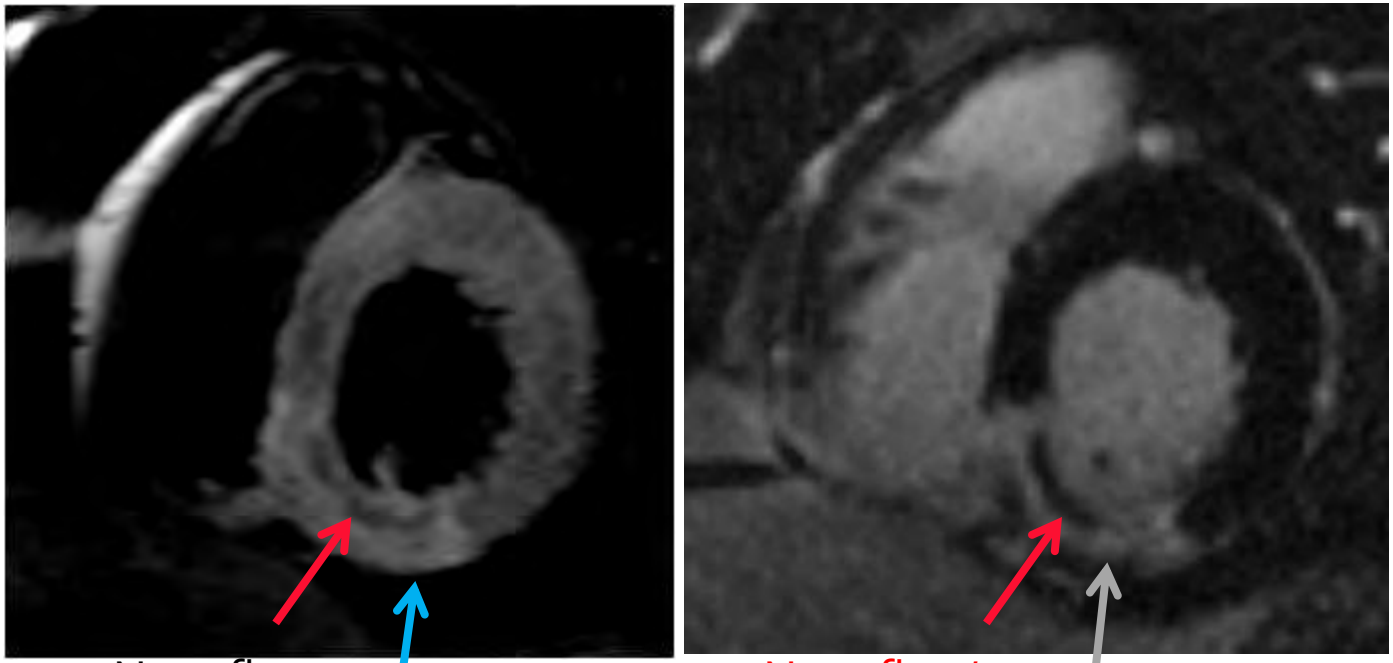
T2-STIR

Late enhancement



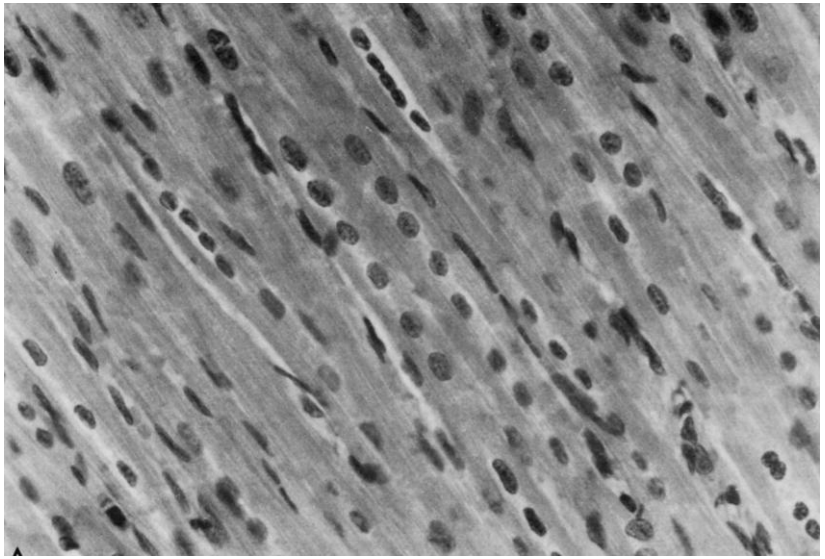
No-reflow

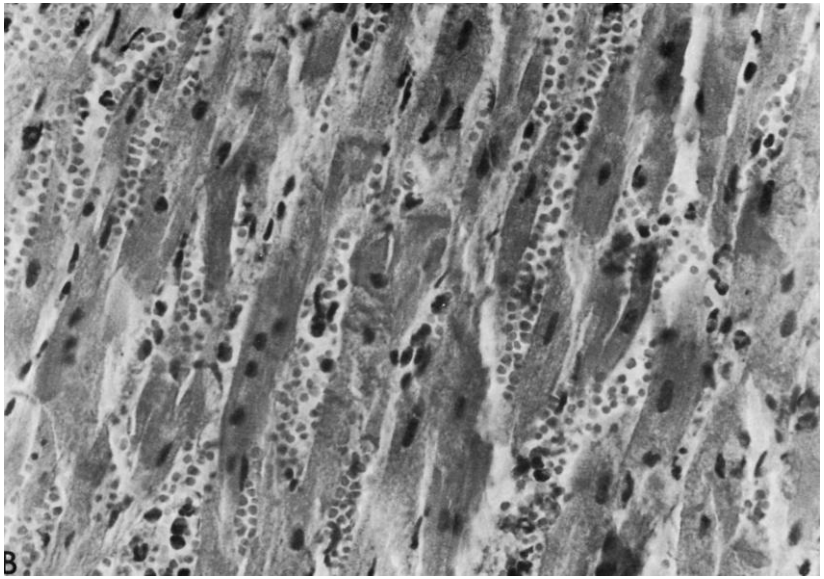
Edema

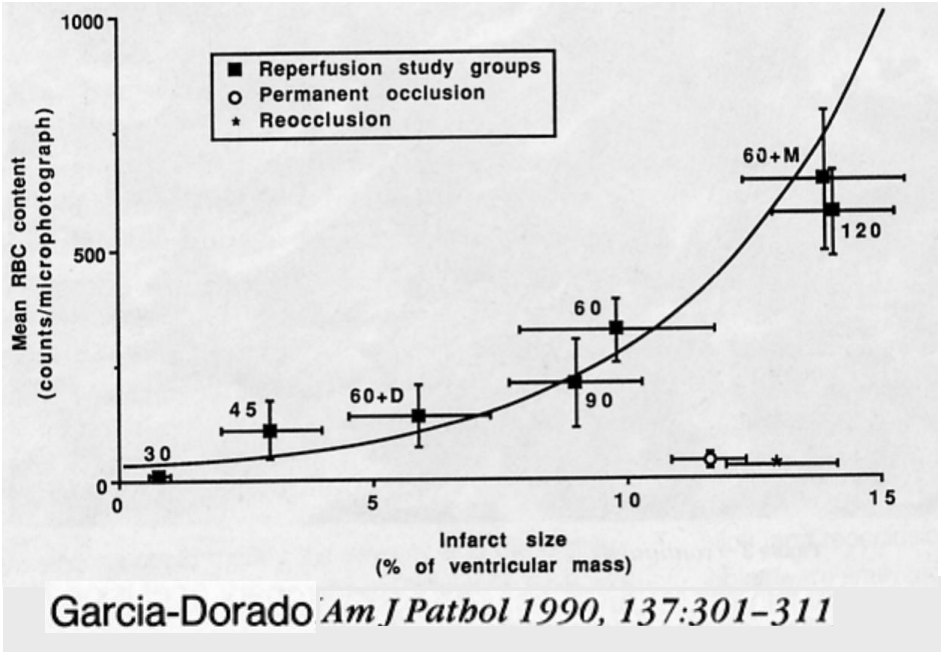
No-reflow/ hemorrage

Infarct

Mechanism of cell death during reperfusion: non-cardiomyocyte cells







Garcia-Dorado *Am J Pathol 1990, 137:301–311*

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The effect of STEMI, and CAD, on survival and quality of life is mediated by cell death causing contractile failure, LV remodeling, and arrhythmias

There is one opportunity for patients with to scape an adverse evolution: EARLY reperfusion

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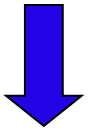
Results of current reperfusion therapy

Very few pts arrive early enough to PCI as to abort MI

During the initial 6h of ischemia myocardial salvage is larger than in most other animal species

Even patients arriving early end-up with significant infarcts



Interventions that would enhance myocardial salvage when applied at the time of reperfusion strongly needed

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Conclusion

1) The extent of ardiomyocyte death secondary to transient coronary occlusion depends mainly on duration of ischemia and residual flow (incomplete occlusion, collaterals), and progresses from endo to epicardium

2) During reperfusion, myocardial cell death occurs mainly during the first minutes after reflow, as necrosis, not apoptosis, that may propagate throug gap junctions

3) Part of cell death may be prevented by interventions applied at the time of reperfusion (lethal reperfusion injury)

4) Altered Ca2+ handling leading to protease activation and hypercontracture, and mitochondrial permeability transition are main, interconnected determinants of cell death whose relative importance appears to depend on the severity of prior ischemic insult. Both depend on on pH normalization

5) No-reflow is mostly a consequence and a marker rather than a cause of large infarcts

6) Prevention of reperfusion injury in patients appears feasible an constitues the next frontier in the treatment of patients with STEMI

Marisol Ruiz-Meana

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Esperanza Agulló, ScD
Luis Agulló, ScD, PhD
Ignasi Barba, ScD, PhD
Jose A. Barrabés, MD, PhD
Mª Ángeles Carmona, ScD, PhD
Celia Fernández-Sanz, ScD
Angeles García, Technician
Alejandra González, ScD
Víctor Hernando, ScD
Javier Inserte, ScD, PhD
Diego López, ScD, PhD
Marcos Poncela, ScD
Elisabet Miro-Casas, ScD
Adoración Quiroga, Technician
Angeles Rojas, Technician
Antonio Rodríguez-Sinovas, DVM
Giuliana Ríos, Assistant
Marisol Ruiz-Meana, DVM, PhD
José Antonio Sánchez, ScD
Carmen Sartorio, ScD, PhD
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