# Cell Therapy for Cardiac Repair: tissue protection vs. cell replacement

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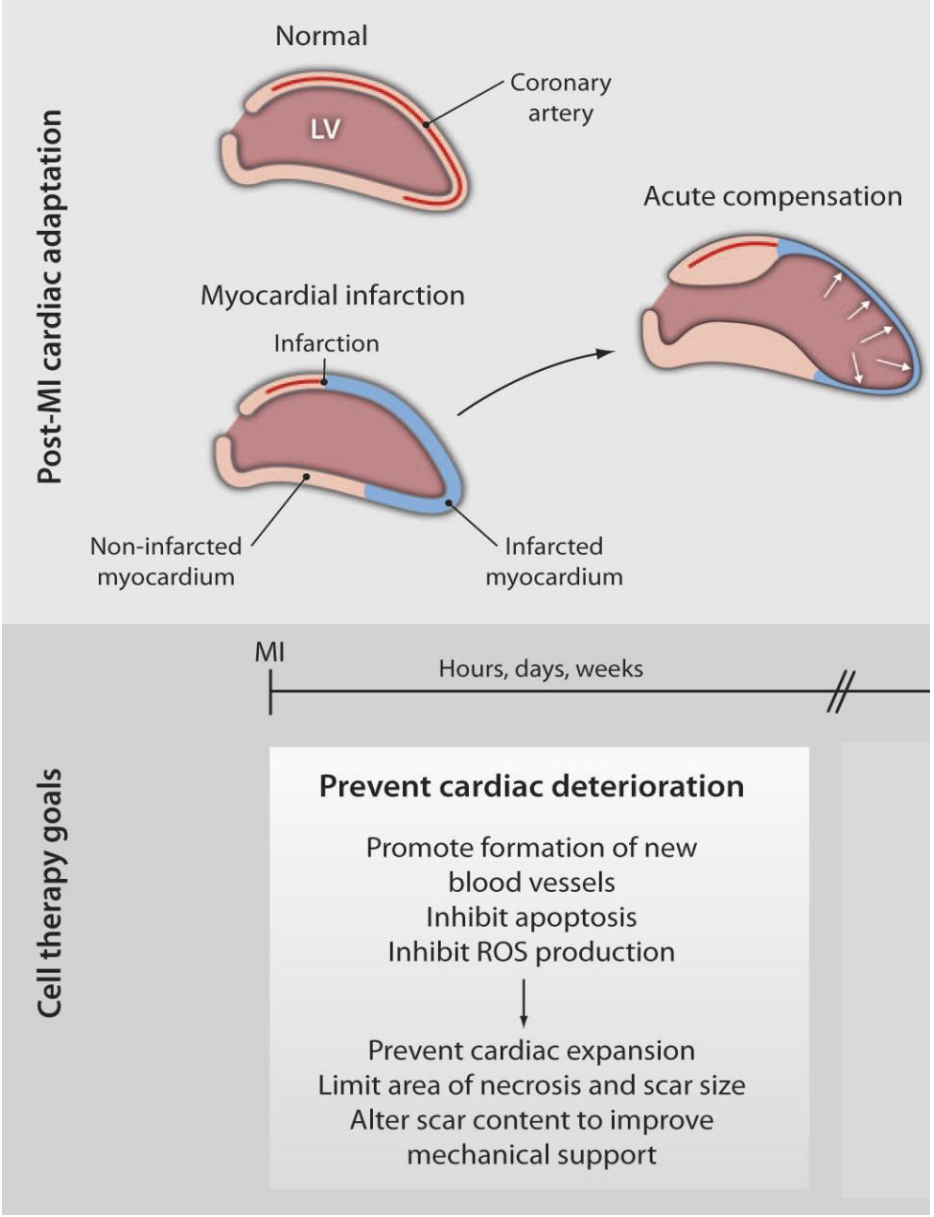
# Post-MI Adaptations & Goals For Cardiac Cell Repair
## (Complex Scenario)




**Mummery, Davis & Krieger, *Sci Transl Med,* 2010**

# Post-MI Adaptations & Goals For Cardiac Cell Repair
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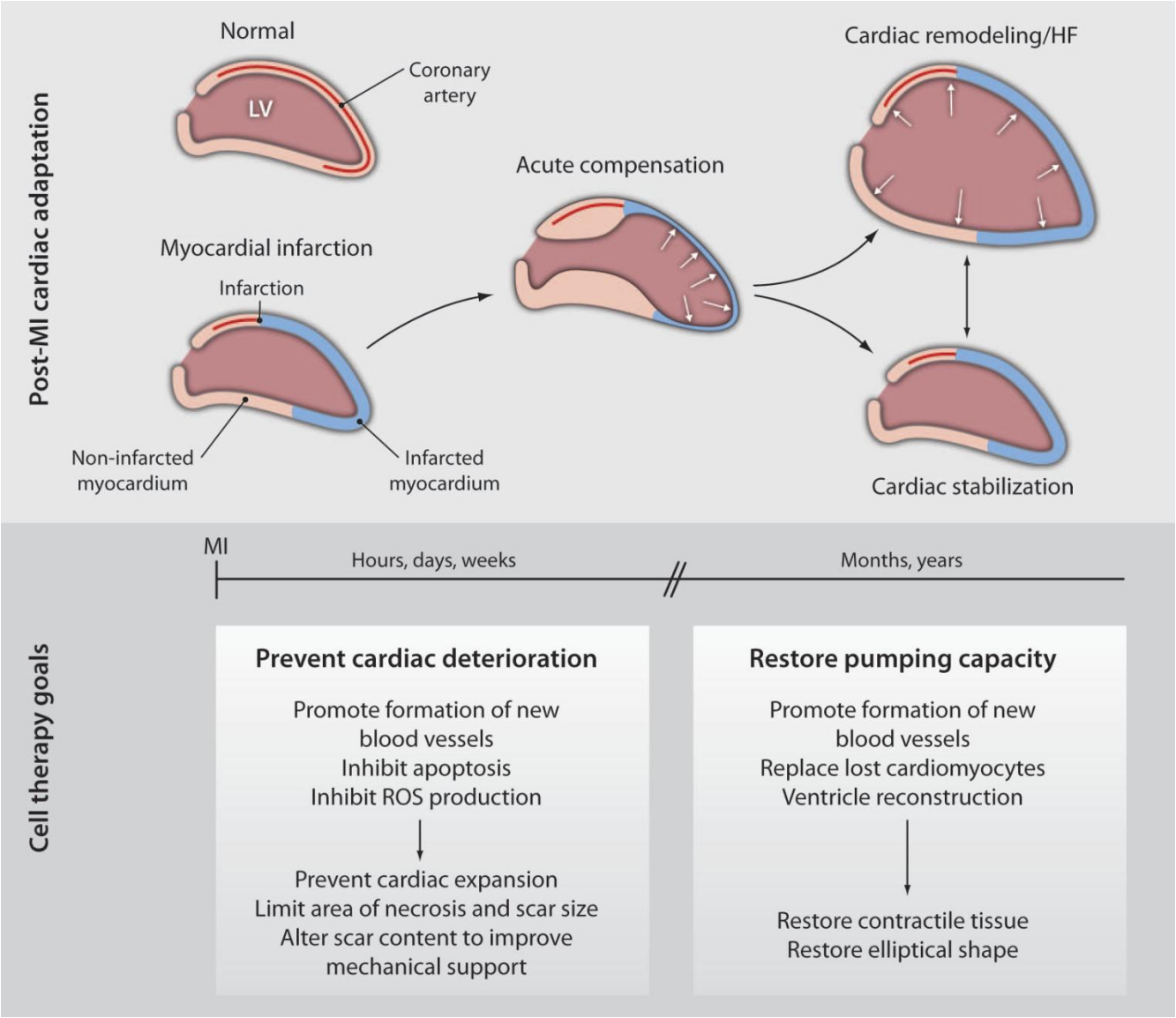
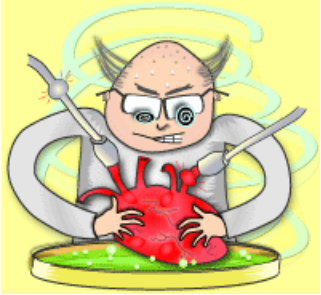



**Mummery, Davis & Krieger, *Sci Transl Med,* 2010**



# Biological Cardiac Repair

Cell replacement - “biological cardiomyoplasty”
Angiogenesis

1. The Naïve Approach

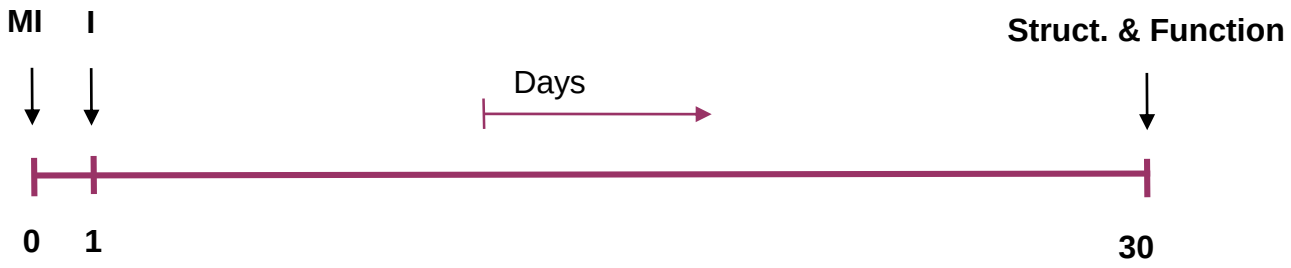
2. The “Awe” Approach (surprise)

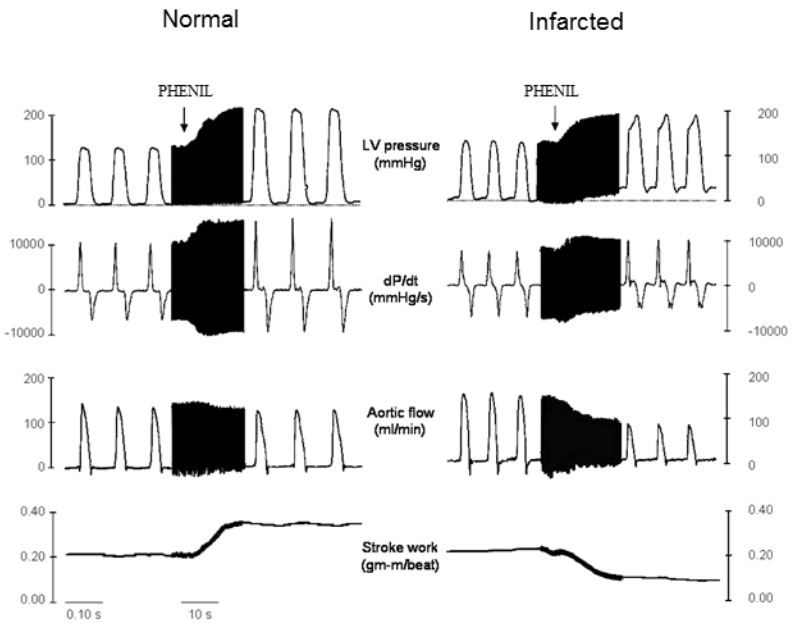
3. The Rational Approach

Evidence for Structural
&
Functional Benefit
(Mechanism of Action?)

Controlled
&
Rational Use

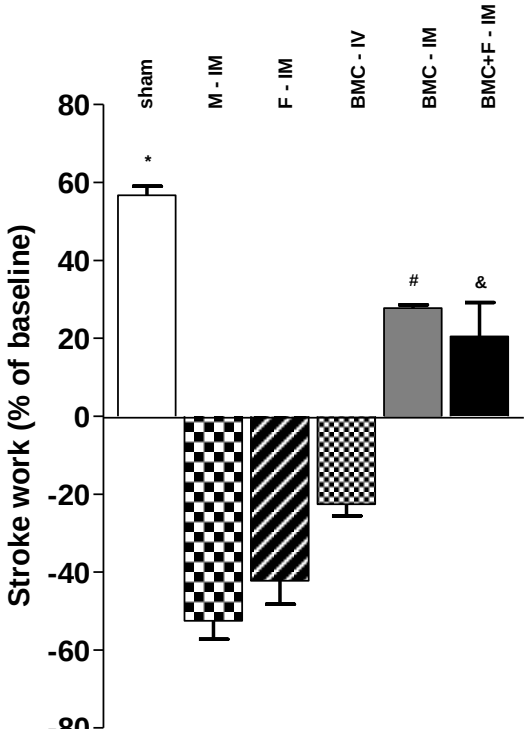
# ASC Transplantation & Cardiac Function Post-MI



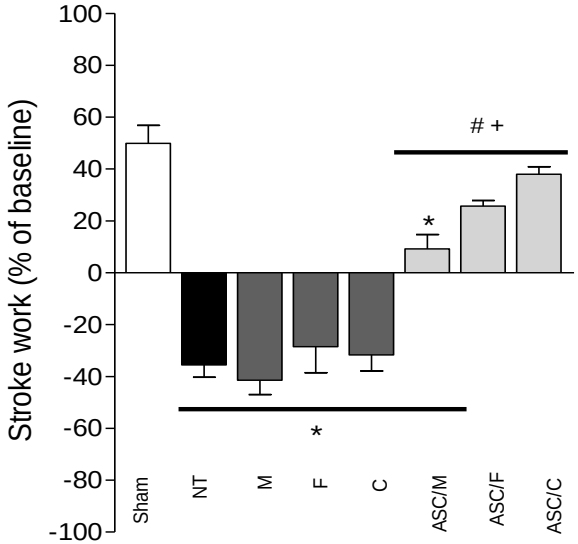

Danoviz et al, *PloS One*, 2010
Santos et al, *Can J Physiol Pharmacol,* 2010

# Prevention of Cardiac Function Deterioration Post-MI

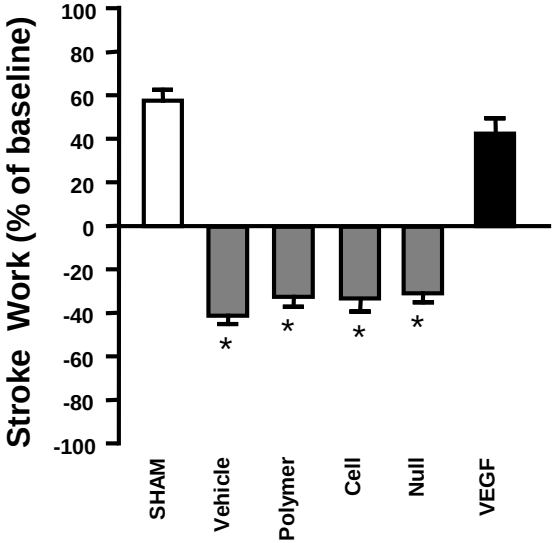



Nakamuta et al, *PLoS One*, 2009

BMCs
rASCs
Fibrobl.VEGF+




Danoviz et al, *PloS One*, 2010




Goncalves et al, *Gene Ther*, 2009

# Current Challenges

- Mechanism of action underlying **tissue protection**
- Validate and optimize the **tissue protection** approach in more suitable animal models (e.g. pigs)
- Devise strategies for the **cell replacement** approach

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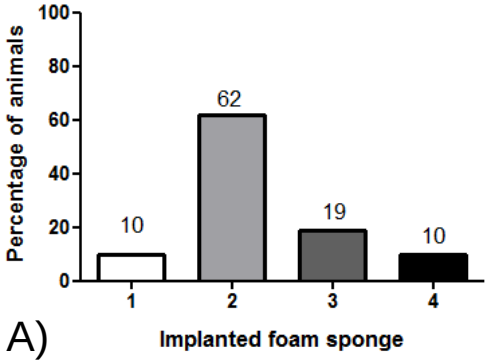
# Closed-artery Catheter-based Myocardial Infarction Method in Pigs (Sponge implantationin LCx)

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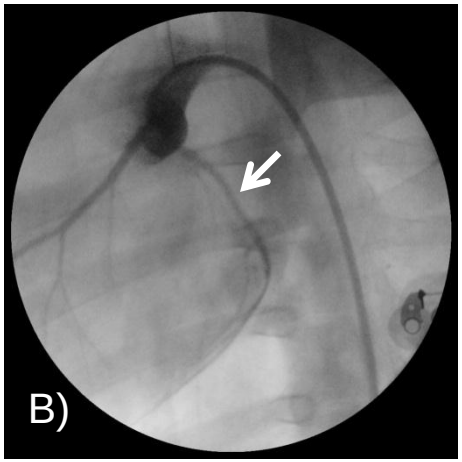
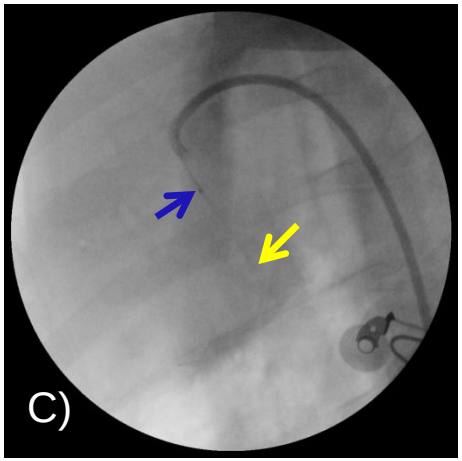
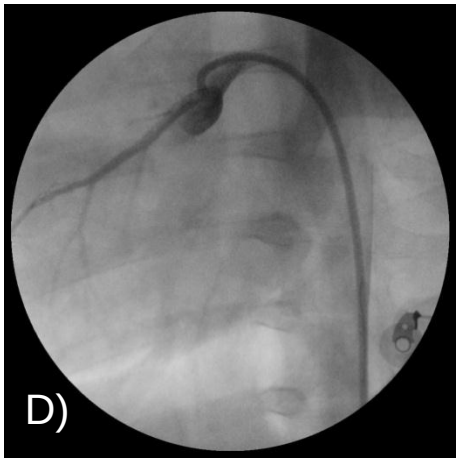
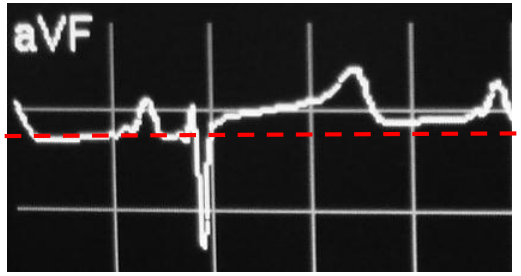


# Ischemia (short-time) and Necrosis (long-time) in Pigs with Occluded LCx

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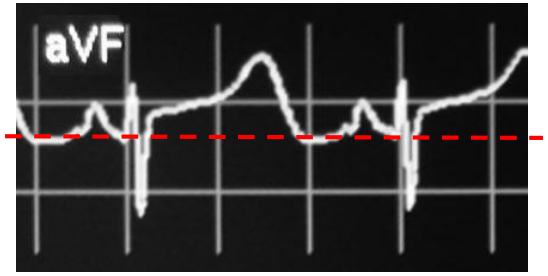


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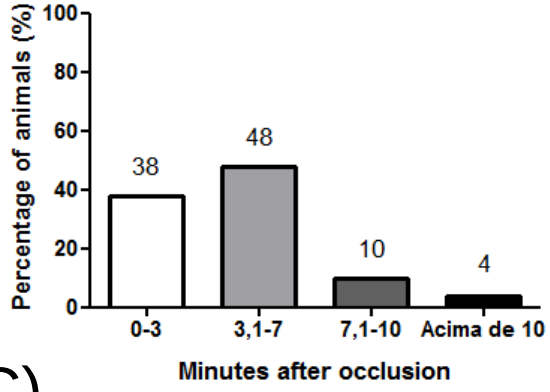


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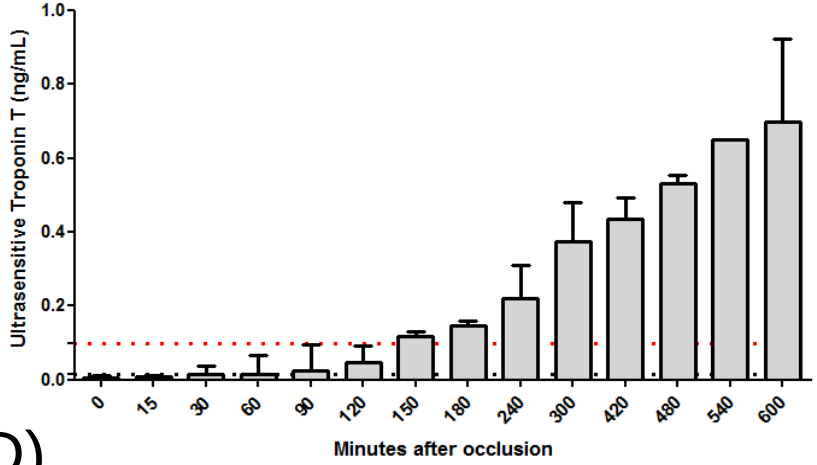


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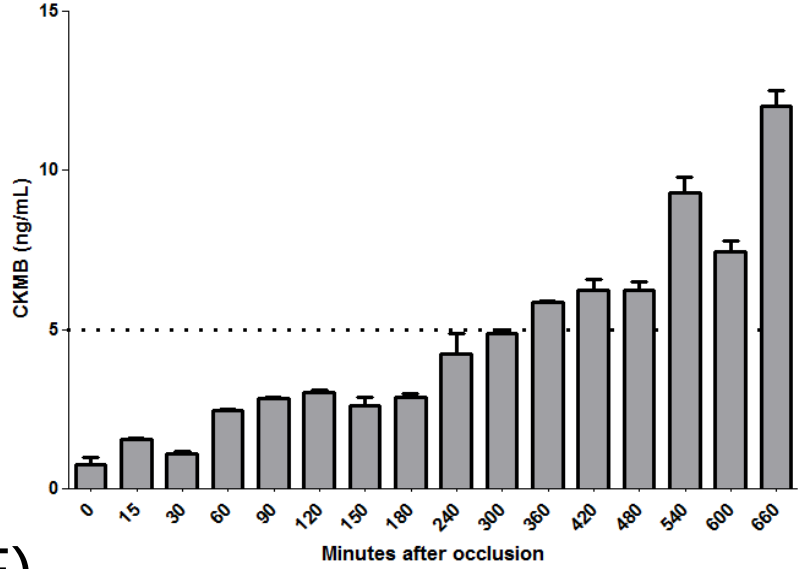
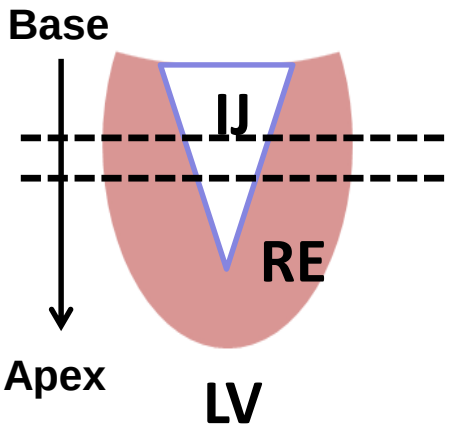



E)

# Anatomopathological Analysis 30 Days after LCx Occlusion

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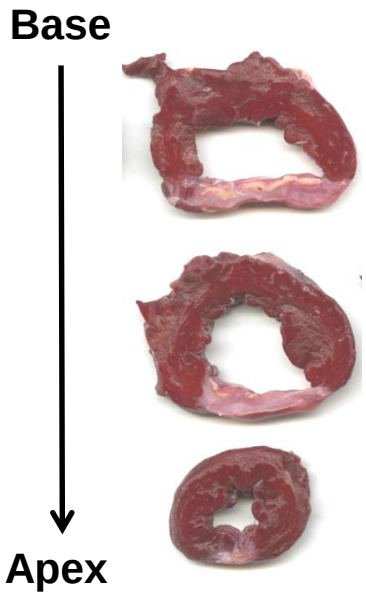


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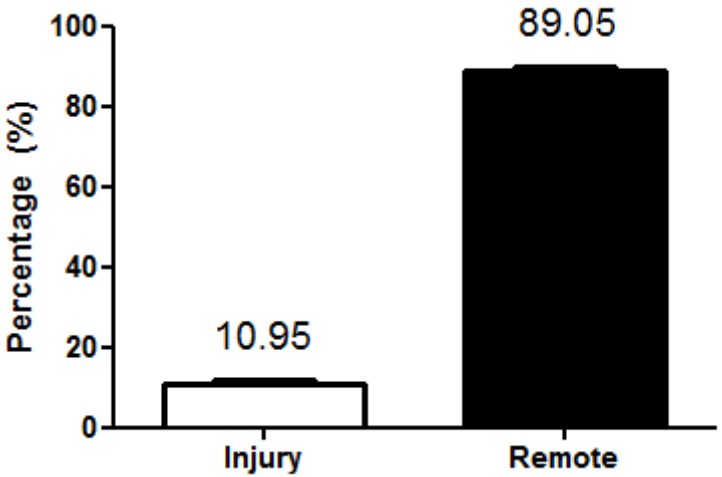


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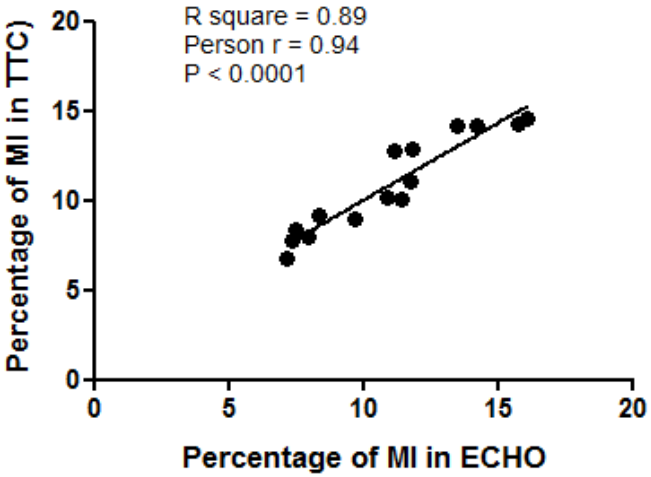


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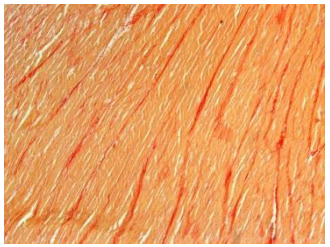

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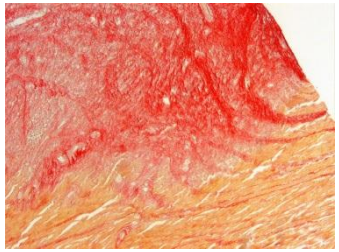
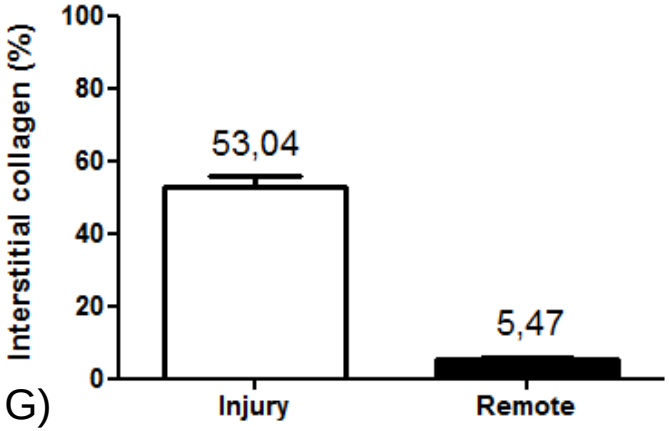





G)

# Summary/Pig MI Model:

- These data provide evidence for a suitable closed-artery catheter-based method to produce MI in pigs;
- The MIs affect about 11% of the LV and are not accompanied by overall cardiac dysfunction.

# pASC's Injection Protocol in Pigs

A

**Experimental Design:**

**Phase I**

- Examination
  *Basal Angiography
- MI induction

**Phase II**

- Examination
  * 1st Echo
- Cell injection

**Phase III**

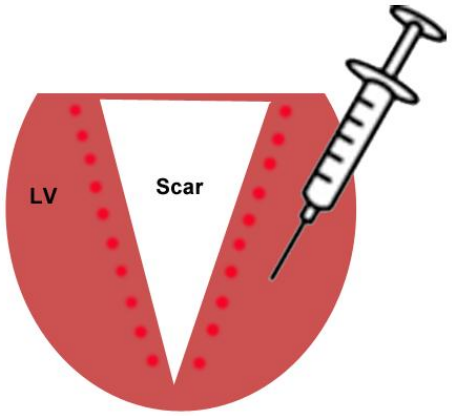
- Examination
  * 2nd Echo
  * Final Angiography
- Anatomopathology

1

0 — 30 — 60 days

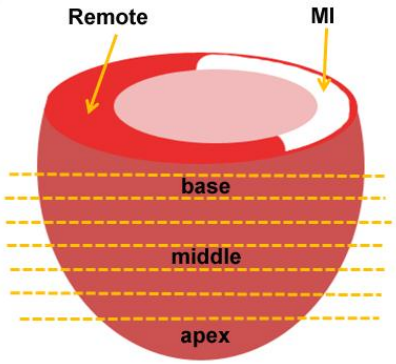
Enalapril Maleate (10 mg/day) + metoprolol succinate (25 mg/day)

B



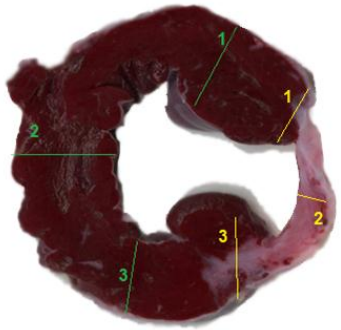

Posterior view

C

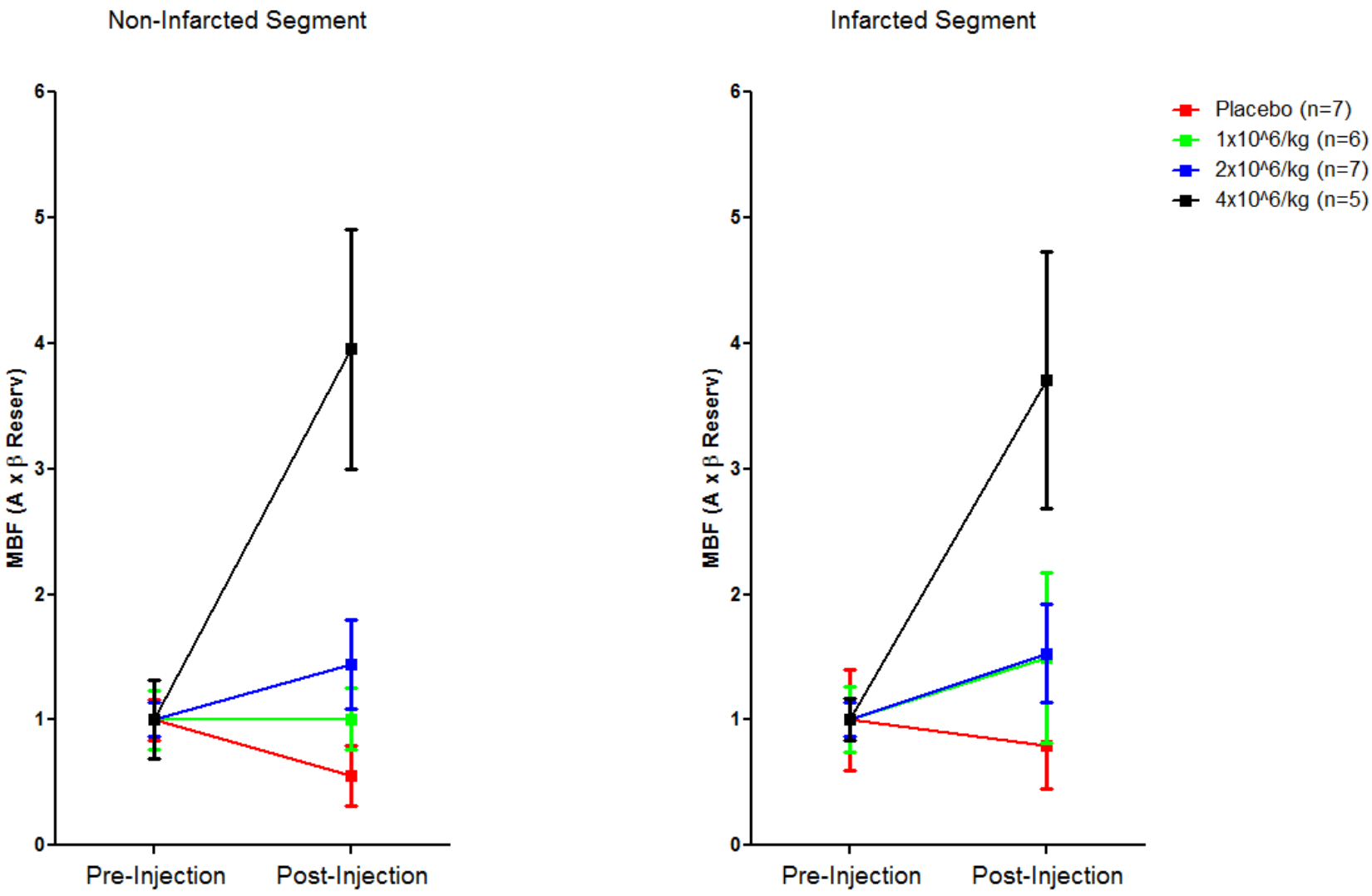



Anterior view

D



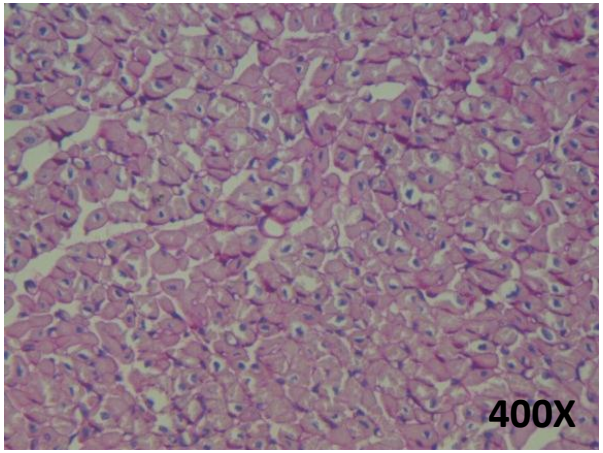
# Effect of ASC Transplantation on Myocardial blood flow Post-MI




Dariolli et al, unpubl.

# Effect of ASC Transplantation on Blood Vessel Number Post-MI (PAS staining)

A)

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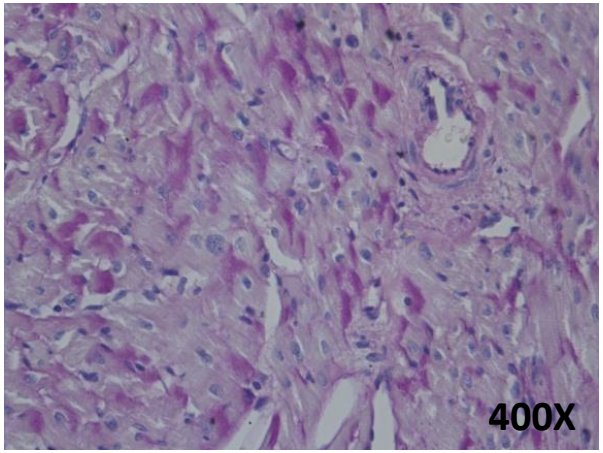


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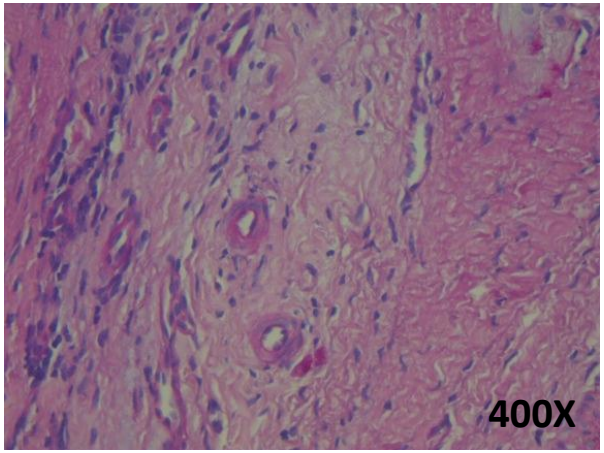
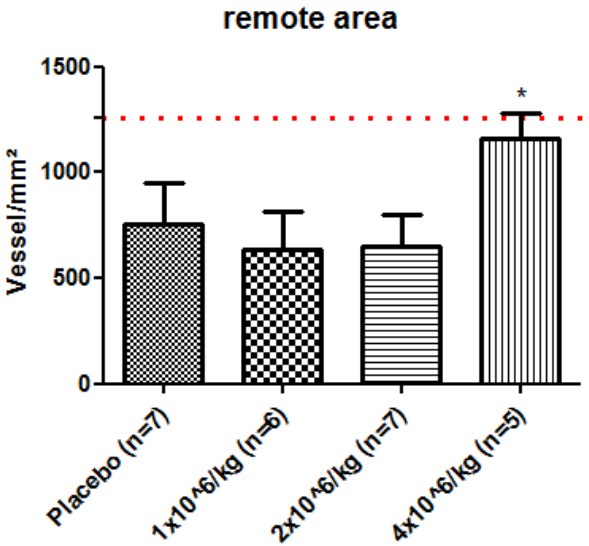
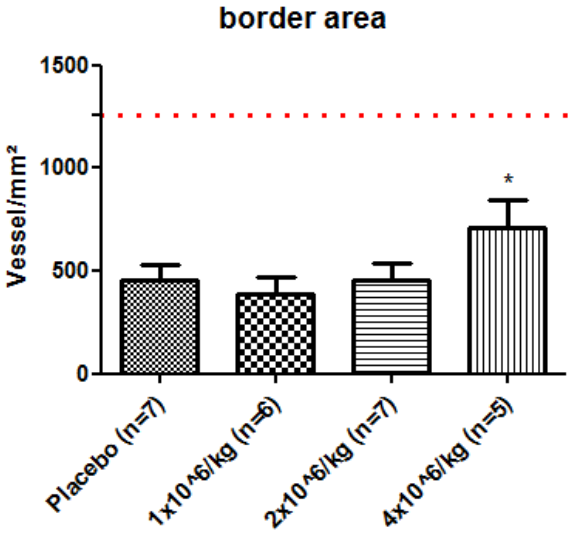
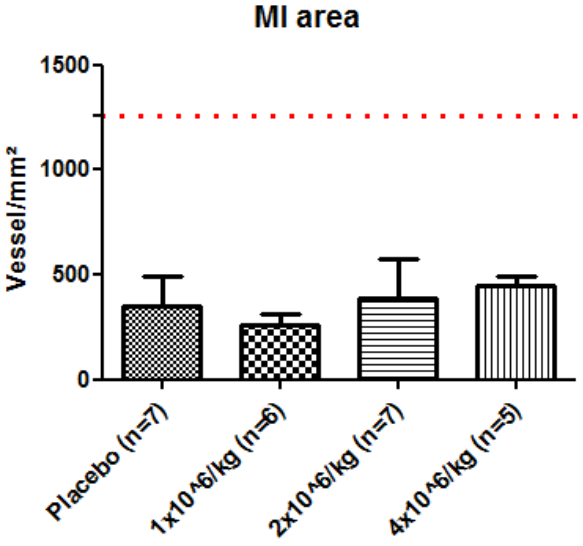


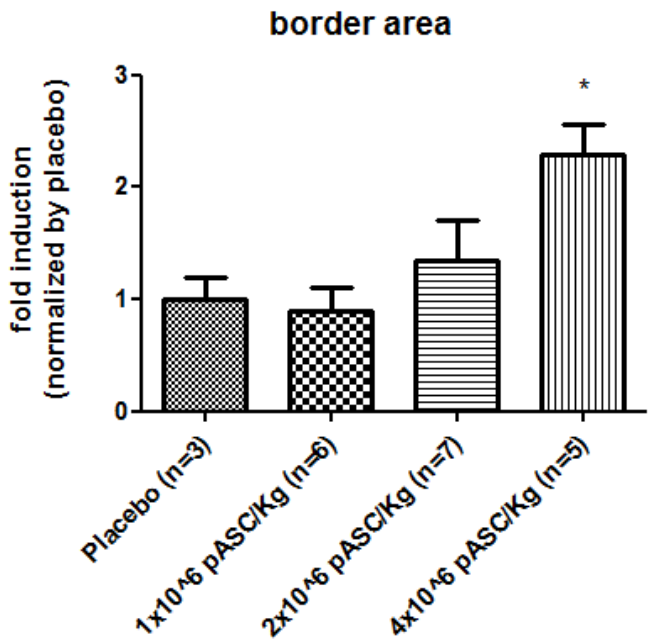



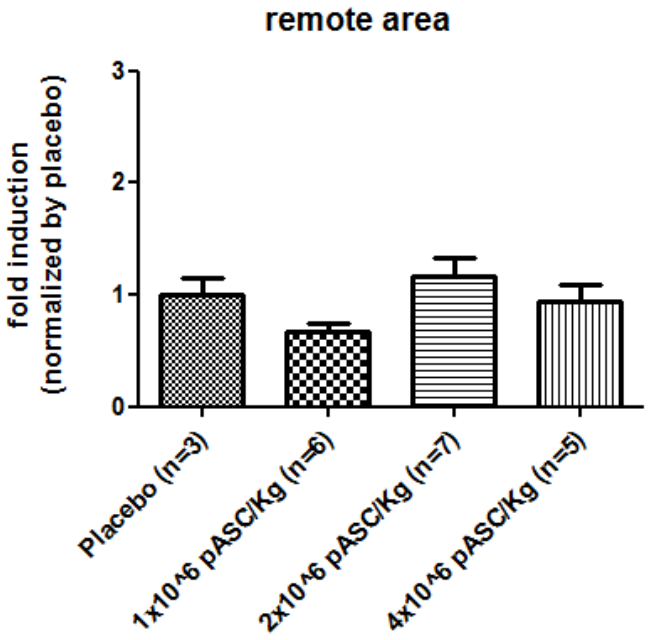



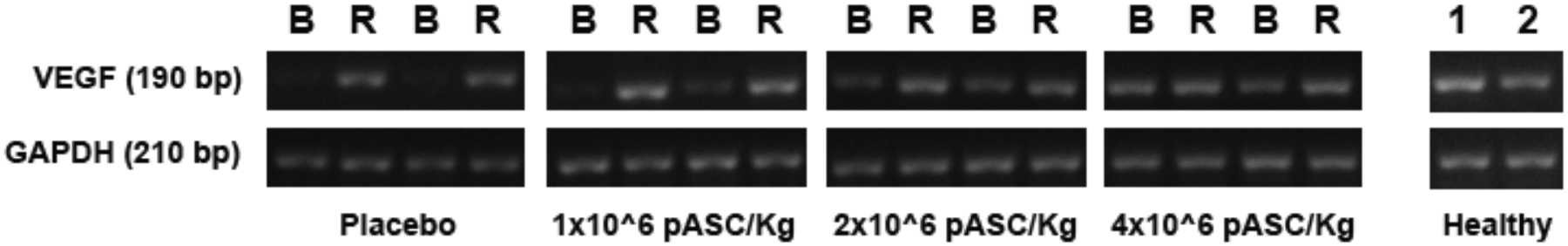
# Effect of ASC Transplantation on VEGF Expression Post-MI

D)



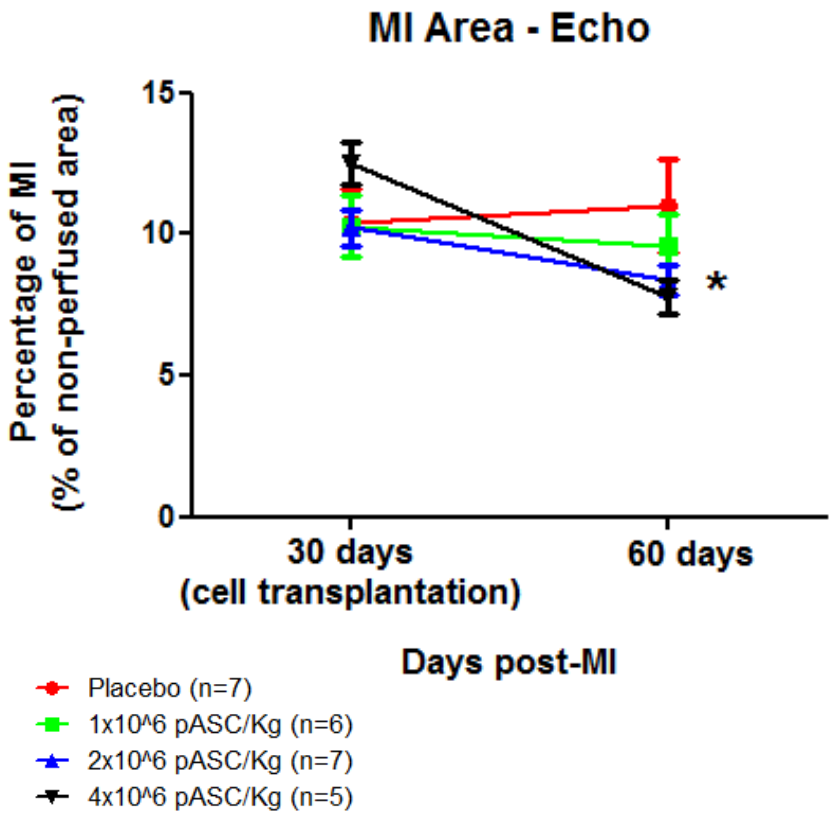




E)



# Effect of ASC Transplantation on Echocardiographic MI Area

A)

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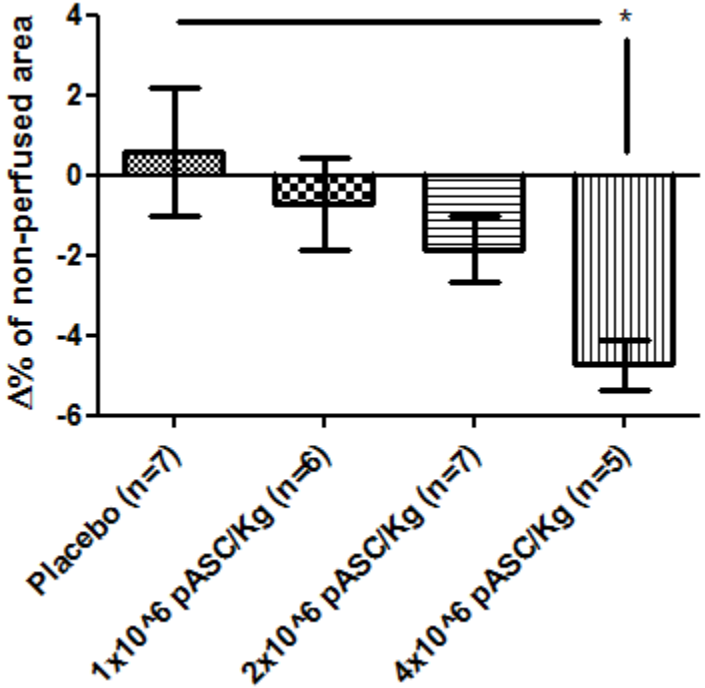


# Effect of ASC Transplantation on MI Area (TTC staining)

C)

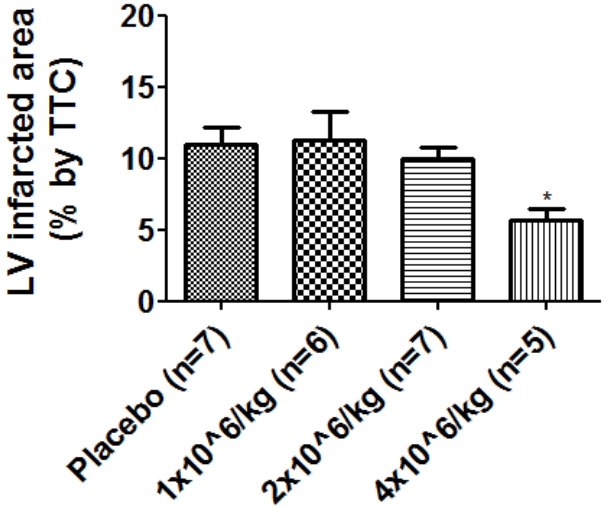
Placebo (n=7)

1x10^6/kg (n=6)

2x10^6/kg (n=7)

4x10^6/kg (n=5)

D)

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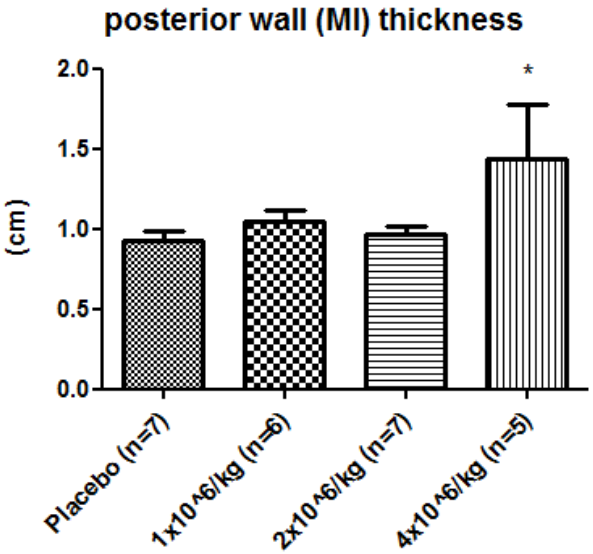


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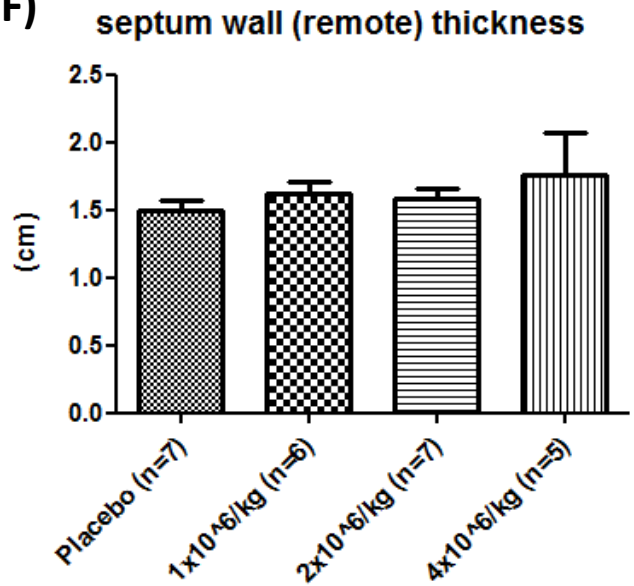
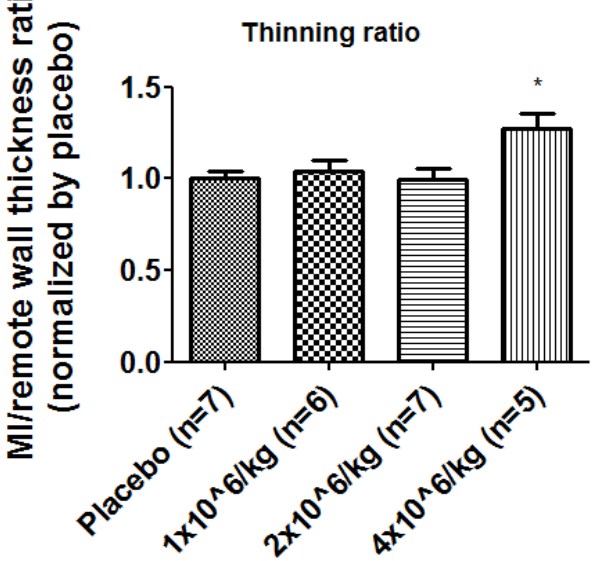
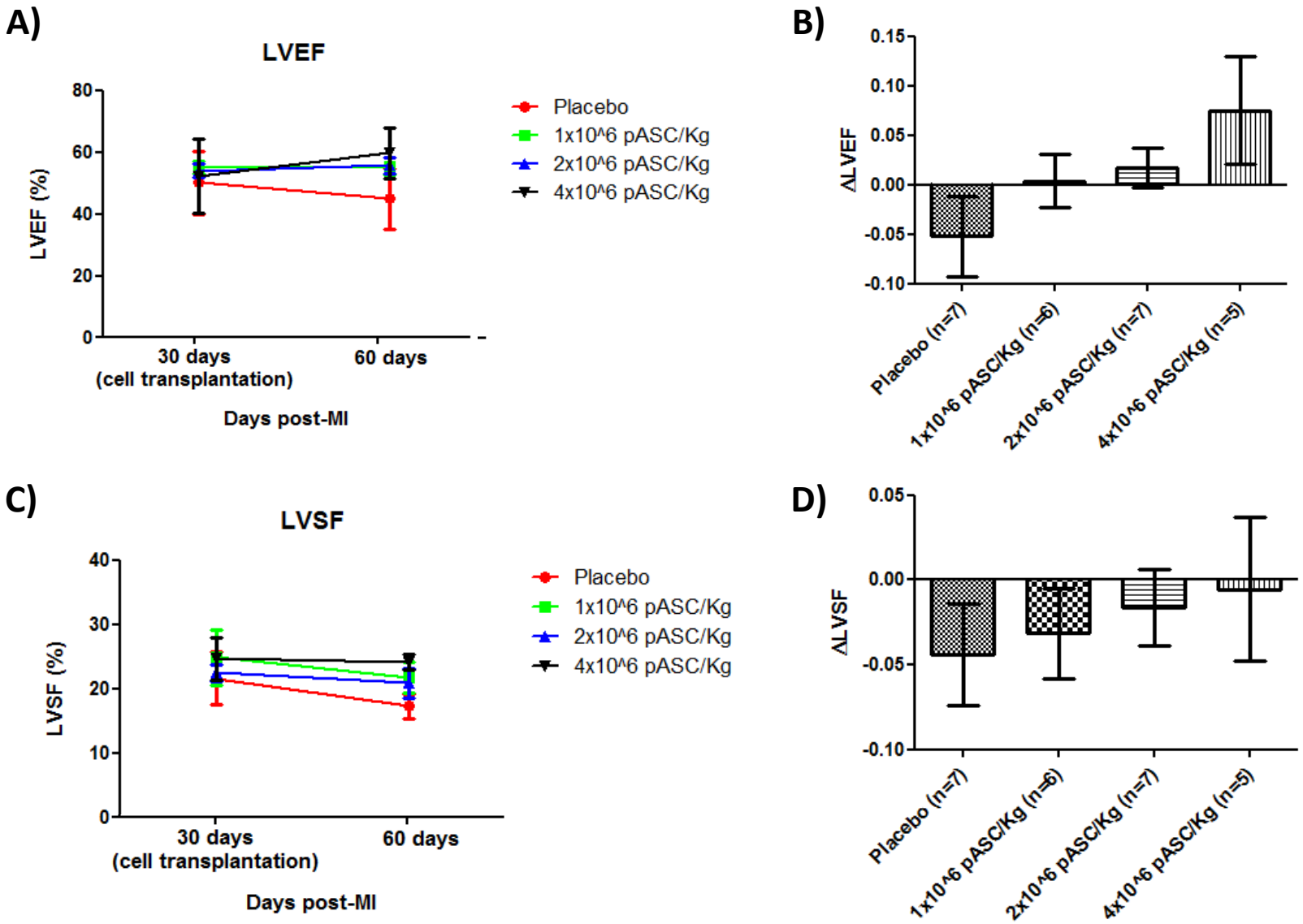



G)

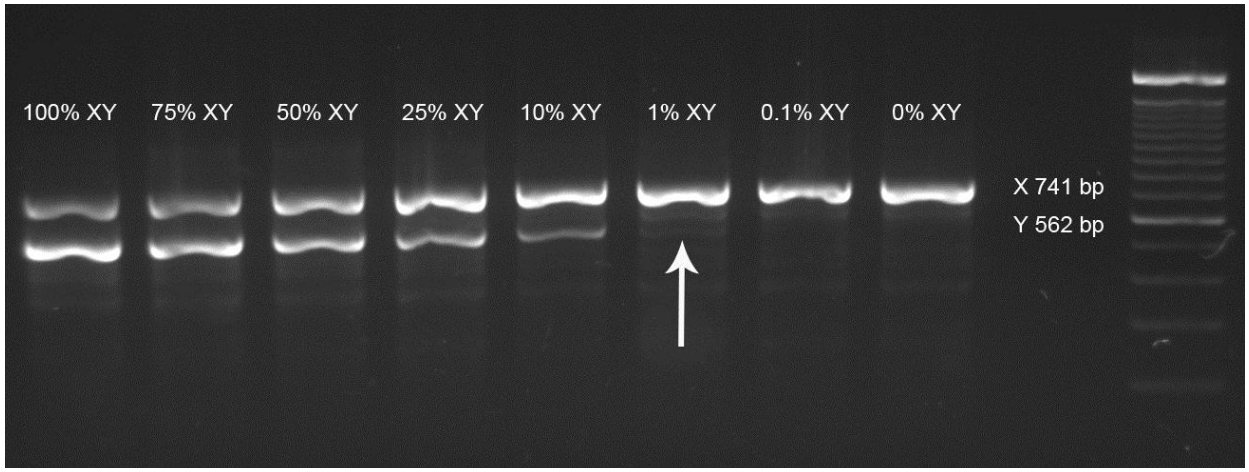


# Effect of ASC Transplantation on Overall Cardiac Function Post-MI

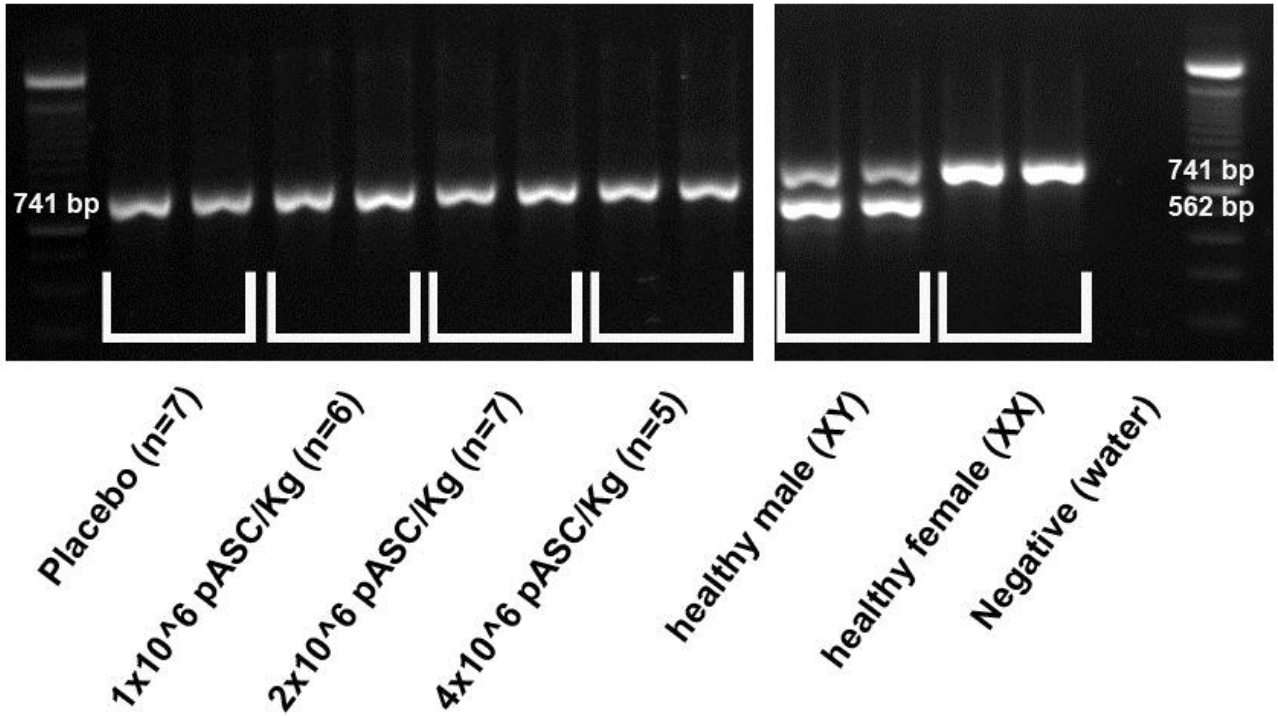


# Estimated ASC in LV 30 Days Post-transplantation by Direct Injection (pig SRY detection)

A)




B)



# Summary/pASC Transplantation:

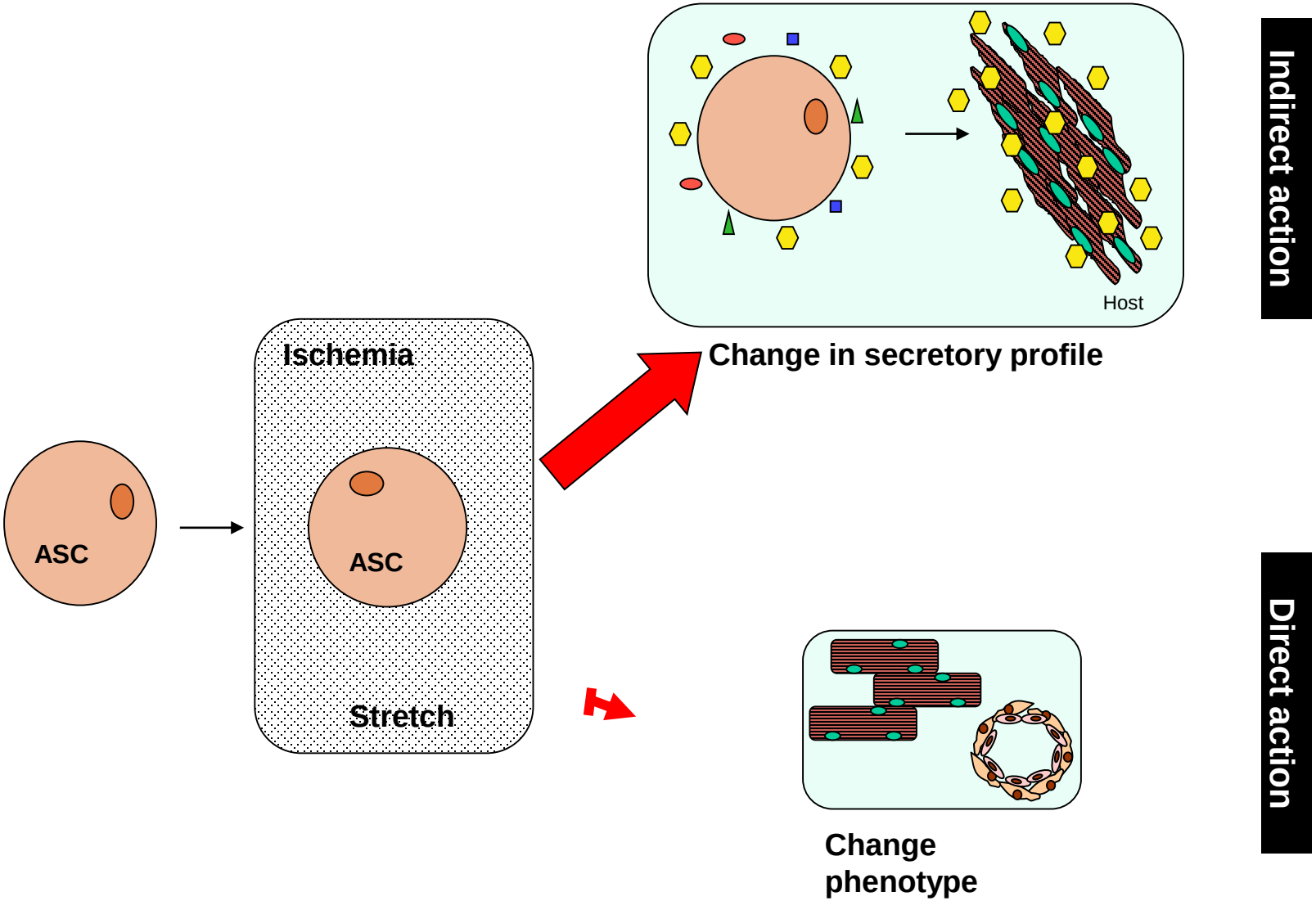
- These data shows that pASC can ameliorate cardiac deterioration Post-MI in pigs;
- These effects are associated with an increase angiovasculogenesis and cardiac perfusion.

# Current Challenges

- Mechanism of action underlying tissue protection
- Validate and optimize the tissue protection approach in more suitable animal models (e.g. pigs)
- Devise strategies for the cell replacement approach

# Prevention of Cardiac Deterioration Post-MI:

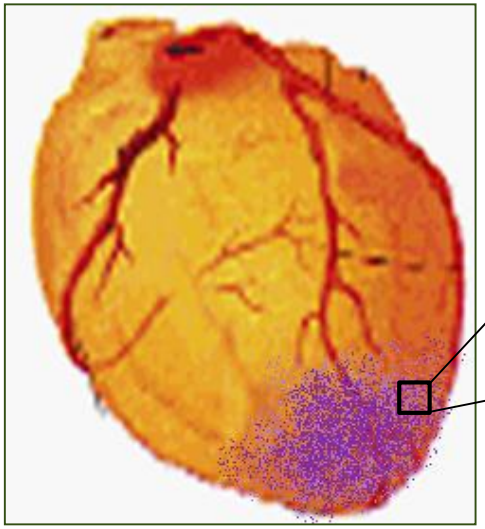
## What are the Molecular Mechanisms?



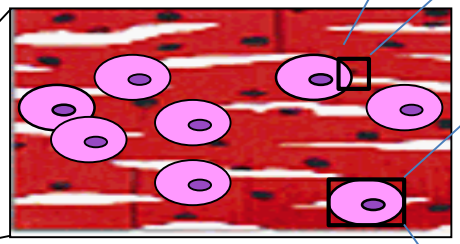
# Scenarios for the Role of Transplanted ASCs in the Ischemic Myocardium

1- Cardiac microenvironment affects the transplanted ASCs (ASC stimulation):

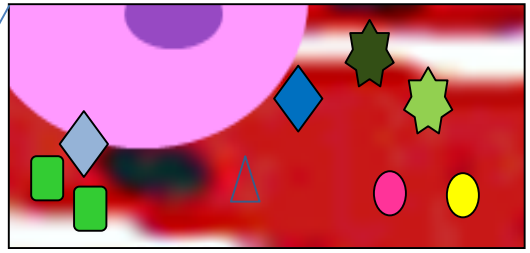
2 -The secretome affects the cardiac microenvironment (secretome therapy)



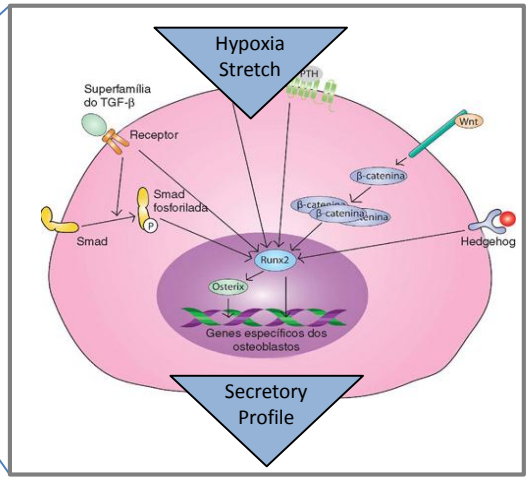
Ischemic Heart



Histological illustration of ASC into myocardium

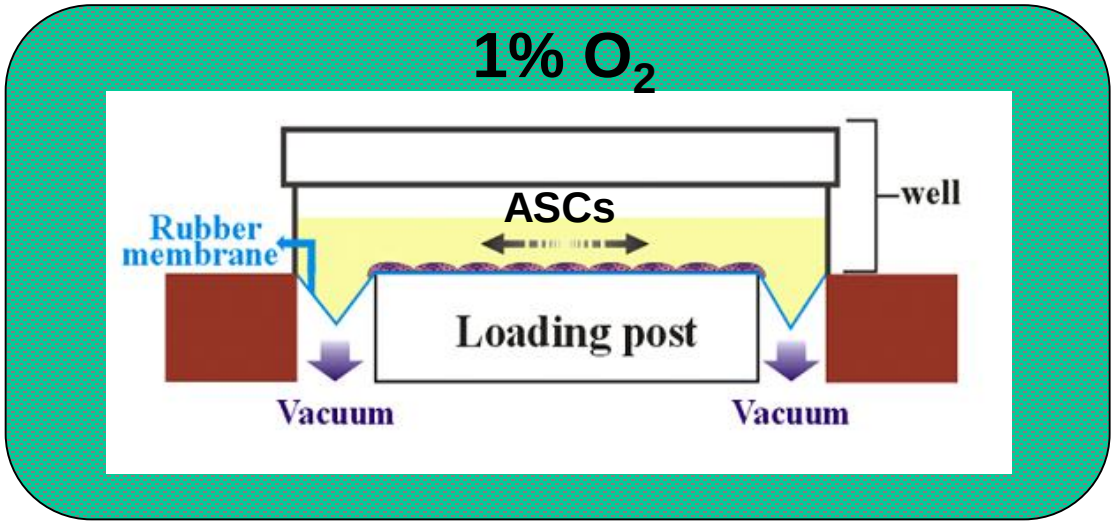


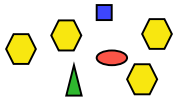
Scenario 1




Scenario 2

# Experimental Model

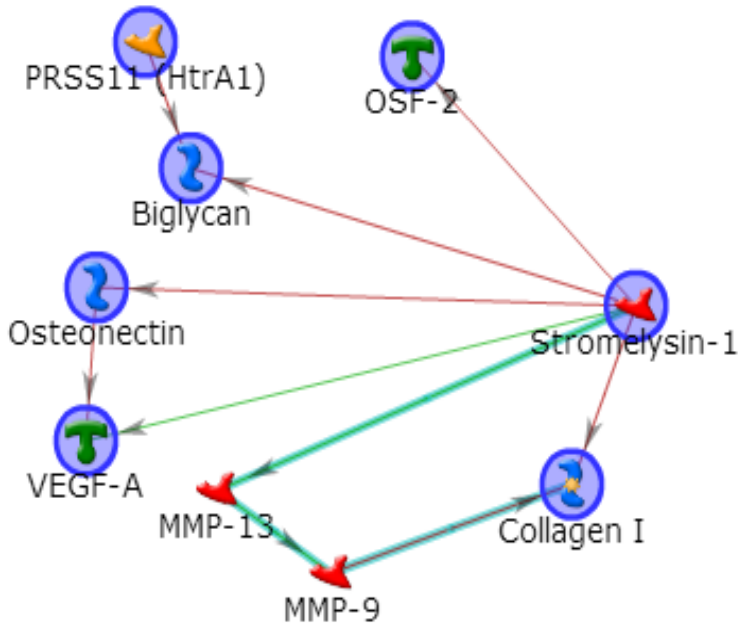


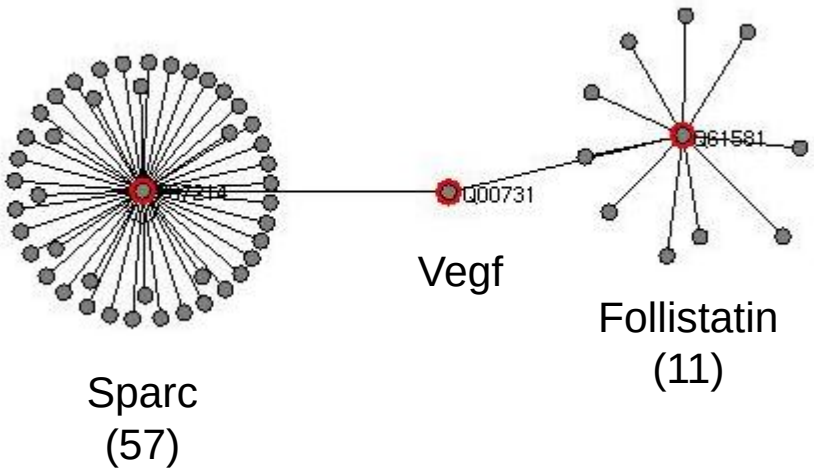



Secretory profile
ESI Q-TOF Analysis

# Interactome Analysis: Direct Interactions with VEGF

**Stromelysin** (MMP-3) inhibts **collagen** I & activates **VEGF**



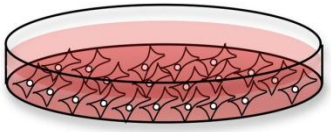

↑ **VEGF**

↓ **Colágeno I**

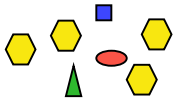
# Biological Assays

Step 1

ASCs

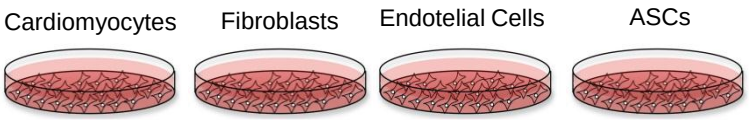


**Stretch + Hypoxia *(24hrs)***



Secretory profile analysis

Step 2




**VEGF stimulation/ inhibition**

Modulation of SPARC production

Step 3

SPARC-*null* ASCs injection

Cardiac Function Assessment

Nakamuta et al, unpublished

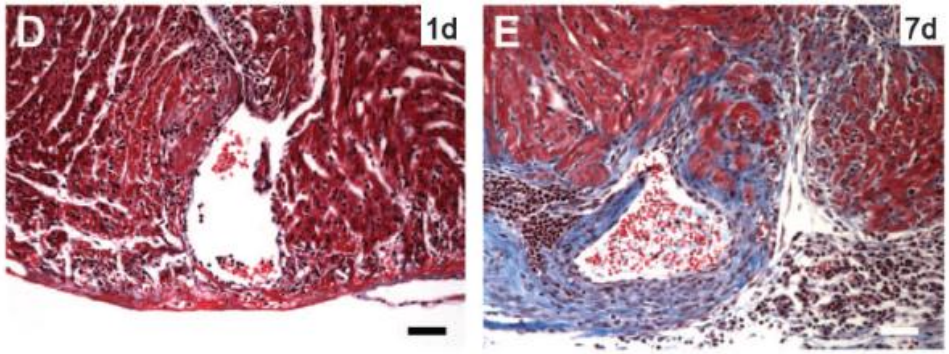
# Current Challenges

- Mechanism of action underlying tissue protection
- Validate and optimize the tissue protection approach in more suitable animal models (e.g. pigs)
- Devise strategies for the cell replacement approach
  - Post-natal LV injury model
  - iPS-derived cardiomyocytes

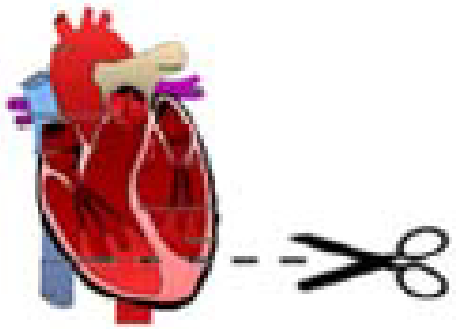
# Current Challenges

- Mechanism of action underlying tissue protection
- Validate and optimize the tissue protection approach in more suitable animal models (e.g. pigs)
- Devise strategies for the cell replacement approach
  - Post-natal LV injury model
  - iPS-derived crdiomyocytes

# Cardiac Regeneration: **Post-natal** LV Resection Model in Rats

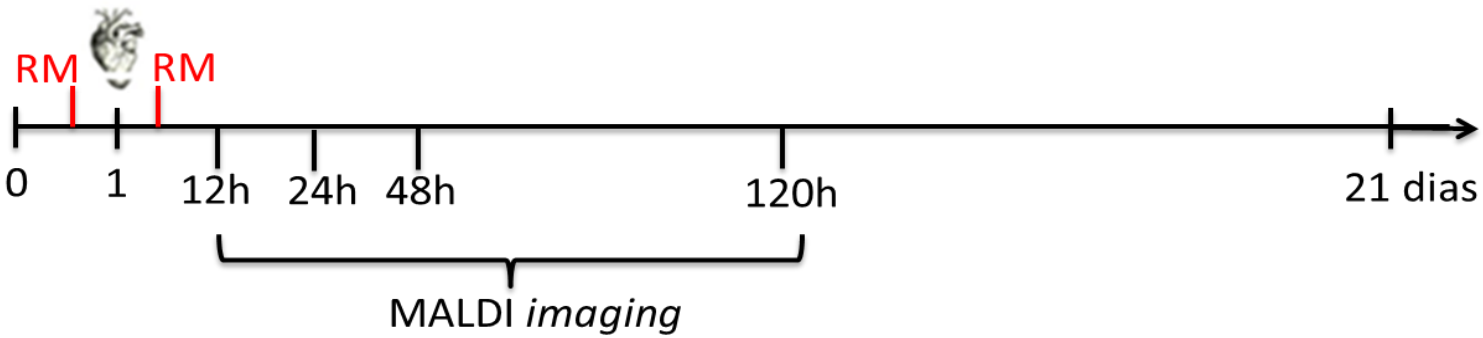


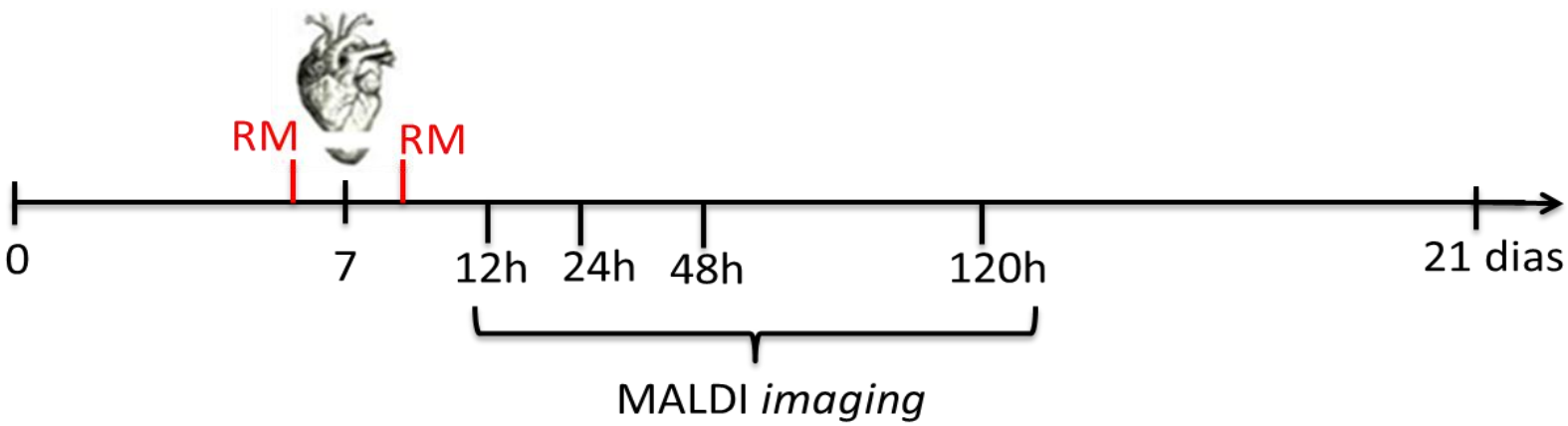

**Porrello et al, *Science,* 2011**



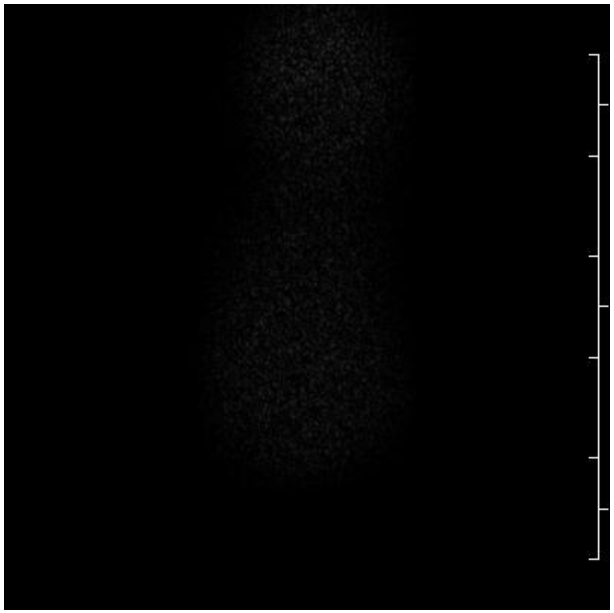
15% LV ressection

# Cardiac LV Ressection Protocol in Rats

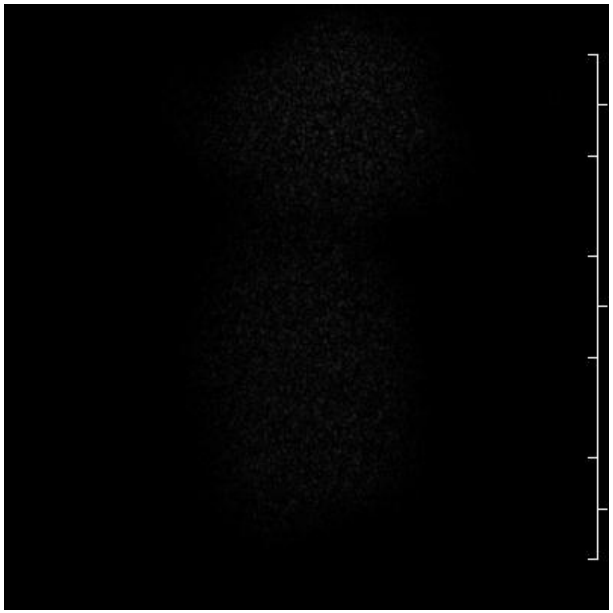


# LV Cardiac Resection in 1 & 7 day old Rats

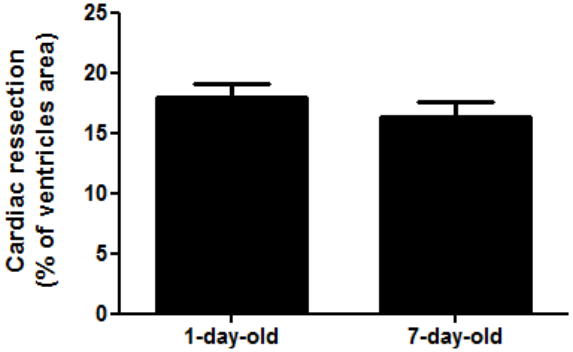


Pre-Ressec.

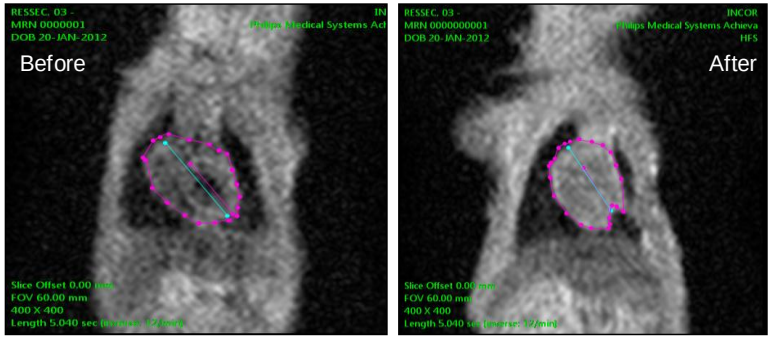


Post-Ressec.

A



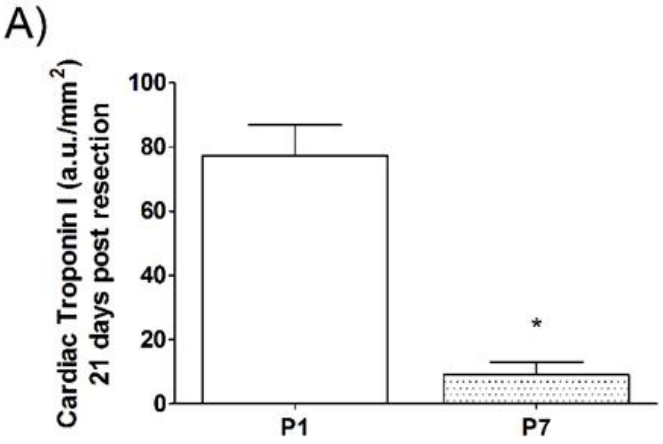

B

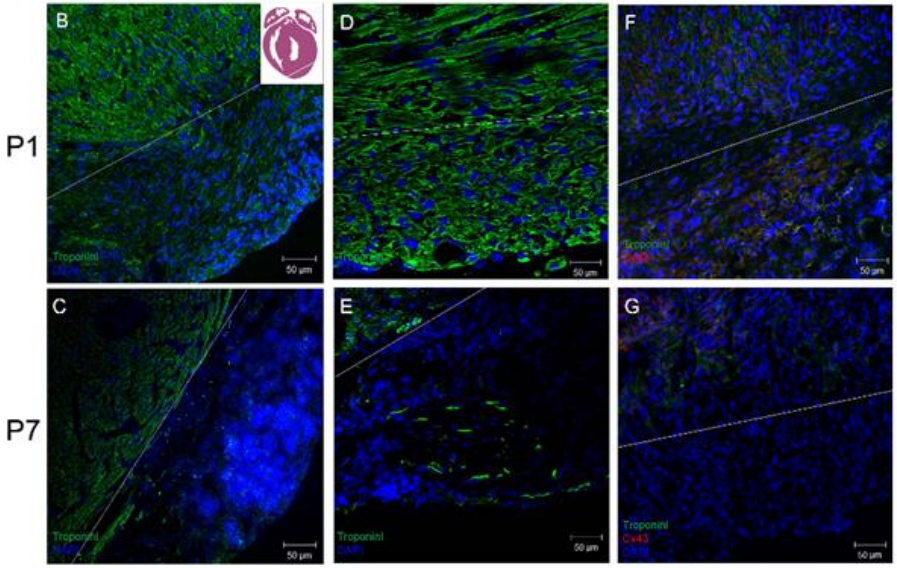


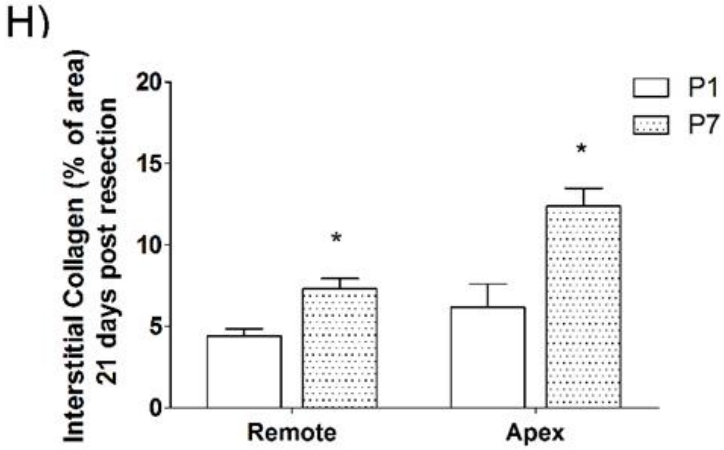

Zogbi et al, unpublished

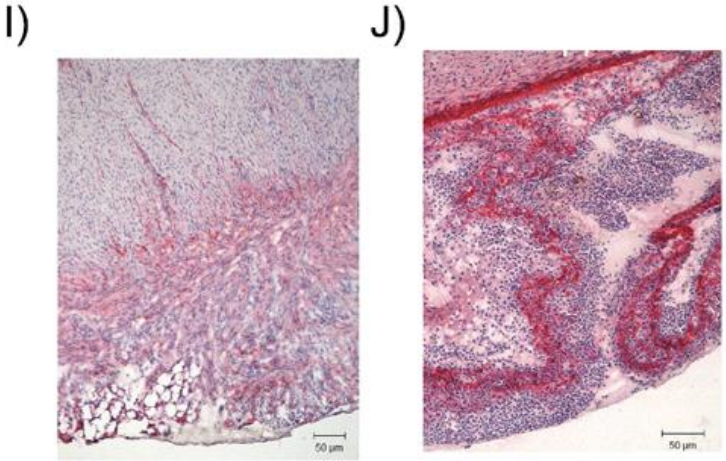
# Cardiomyocyte Regeneration & Collagen Deposition 21 days Post-LV Injury in Rats

Troponin I Immune & Connexin 43 Fluorescence



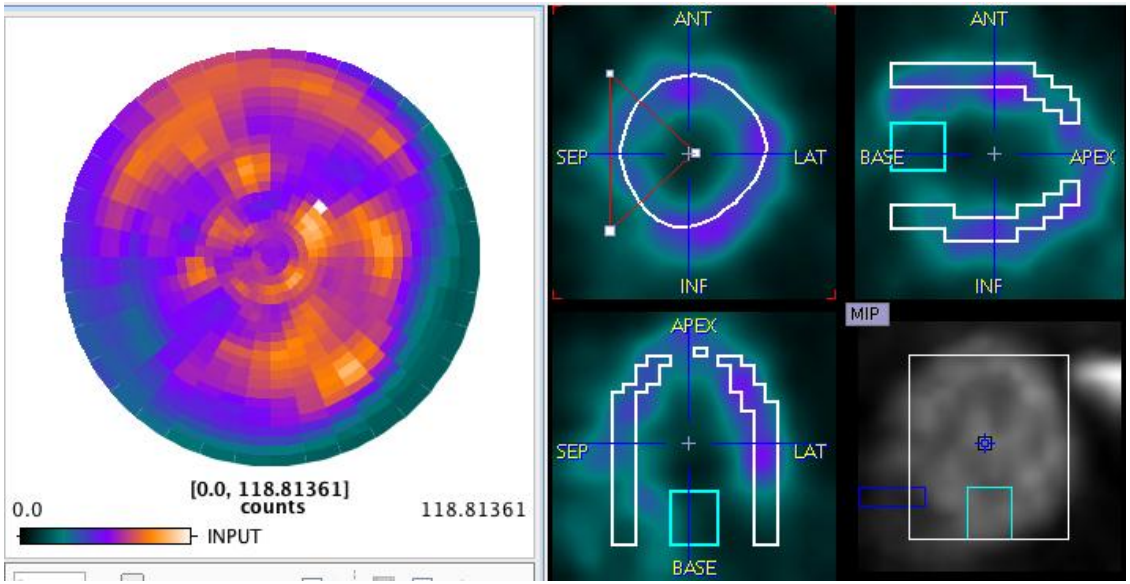




Picrosirius red staining

**Zogbi et al, unpubl.**

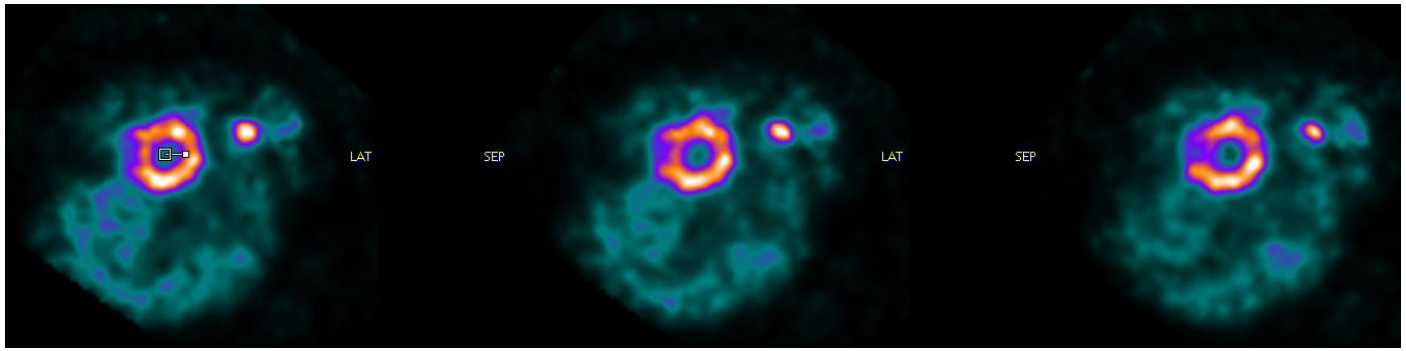
# Myocardial Perfusion in 1 & 7 day old Rats LV Injury

## Single Photon Emission Tomography (SPECT)




ANT

SEP

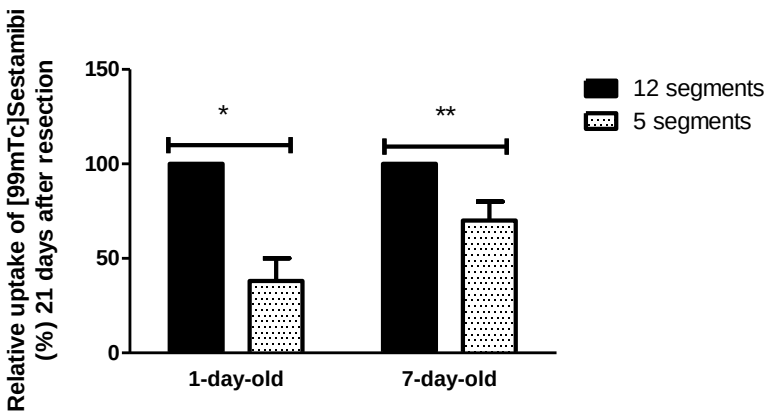



LAT

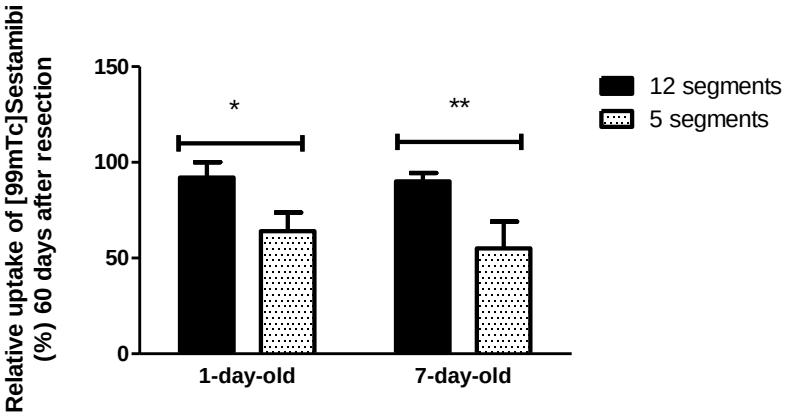
INF

# Myocardial Perfusion in 1 & 7 day old LV Injury Rats

## Single Photon Emission Tomography (SPECT)



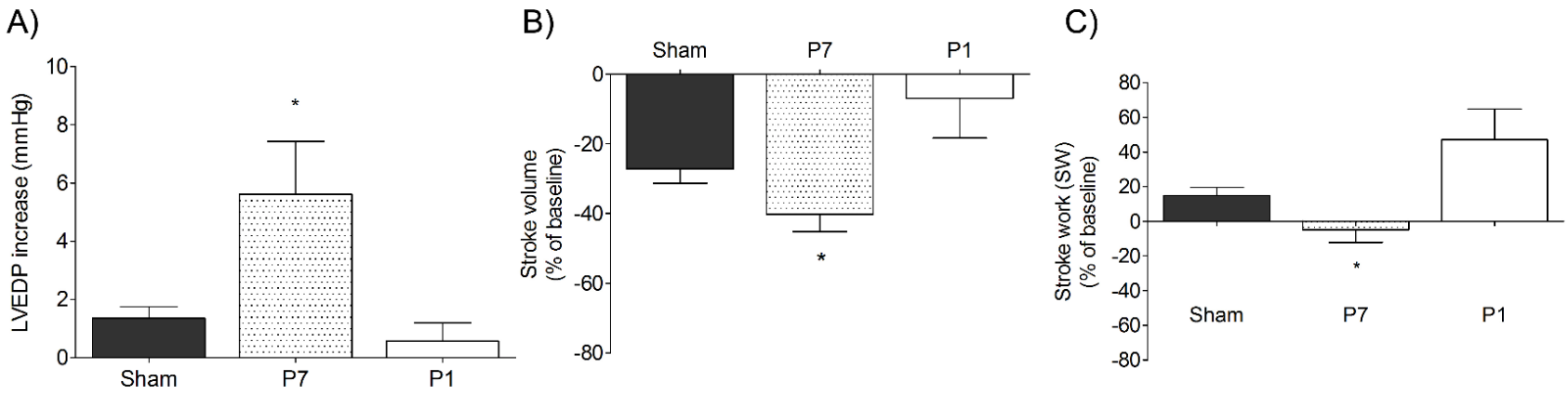

21 days post-resection



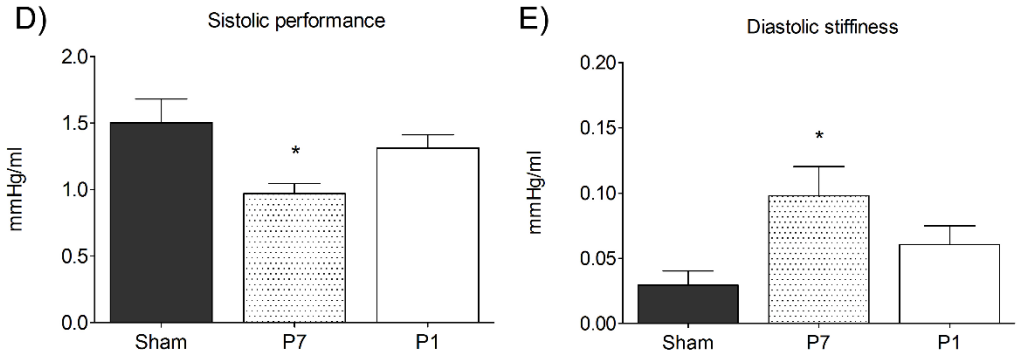

60 days post-resection

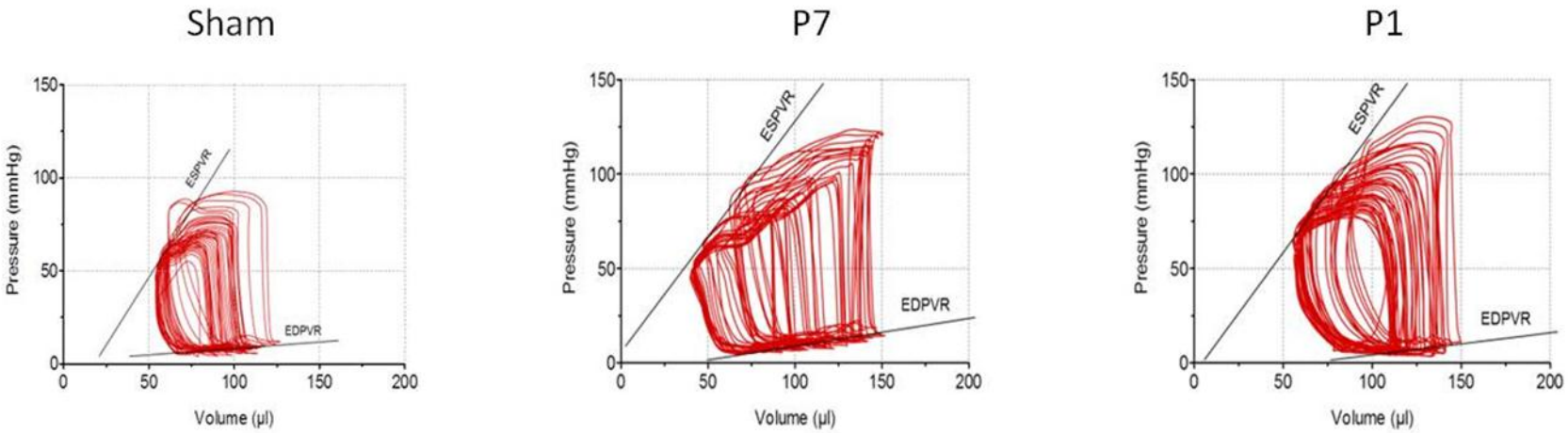
**Zogbi et al, unpublished**

# Afterload Hemodynamic Stress to Assess Cardiac Performance in Post-natal LV Injury Rats

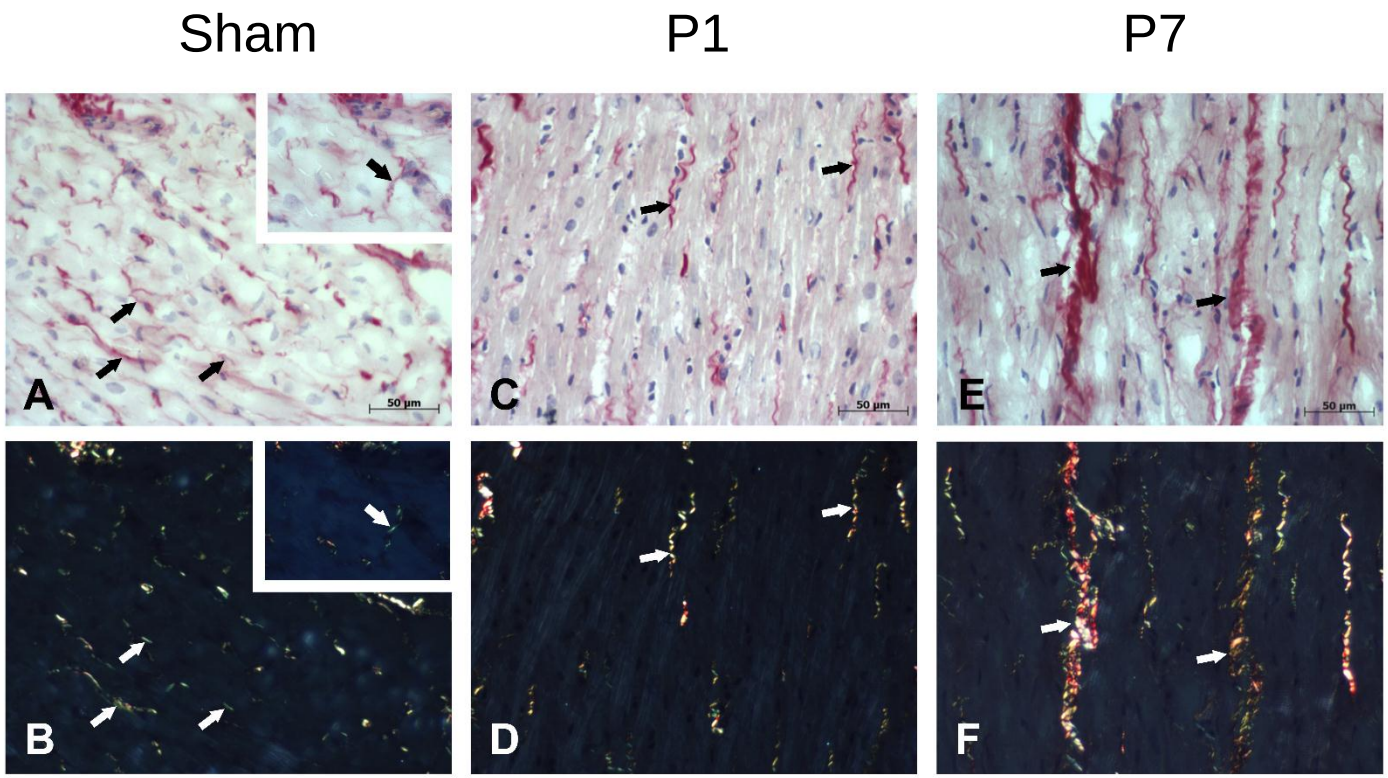


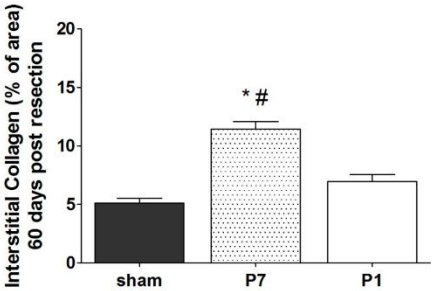
# Preload Hemodynamic Stress to Assess Cardiac Performance in Post-natal LV Injury Rats



# Tissue Collagen Deposition in Post-natal LV Injury Rats
## (HE & Picrosirius under polarized light)



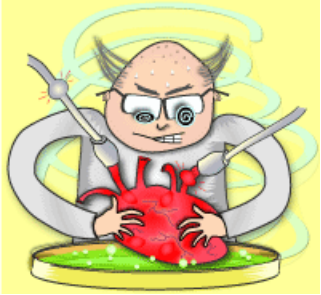



# Summary/Rat Apex Resection:

- Data show evidence that the rat displays early cardiomyocyte neoformation in response to apex resection;
- The overall response leads to long-term preservation of cardiac function despite hypoperfusion, highlighting issues in this complex response that must be taken into account when exploring future therapeutic approaches based on this response.

# Biological Cardiac Repair: Current Challenges

- Mechanism of action underlying tissue protection
- Validate and optimize the tissue protection approach in more suitable animal models (e.g. pigs)
- Devise strategies for the cell replacement approach
  - Post-natal LV injury model
  - iPS-derived cardiomyocytes



# Biological Cardiac Repair

Cell replacement - “biological cardiomyoplasty”
Angiogenesis

1. The Naïve Approach

2. The “Awe” Approach (surprise)

3. The Rational Approach

Evidence for Structural
&
Functional Benefit
(Mechanism of Action?)

Controlled
&
Rational Use

# Biological Cardiac Repair
# (cell replacement vs. tissue protection)

- Minimize cardiac structural and functional damage post-MI by seeding progenitor cells (e.g. adipocyte derived mesenchymal stem cells), **carriers of factors** that may influence a variety of processes (e.g. neoangiogenesis & anti-scar);

- Improve cardiac contractility post-MI by **replacement of lost cells** by newly seeded cardiomyocytes or precursors.

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**Vinicius Bassaneze**
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**Rafael Dariolli**
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**Camila Zogbi**
**Ana Elisa TS Carvalho**
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**Hematologist**
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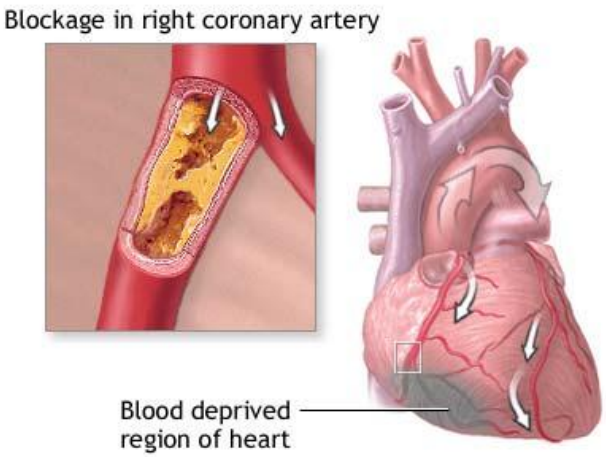
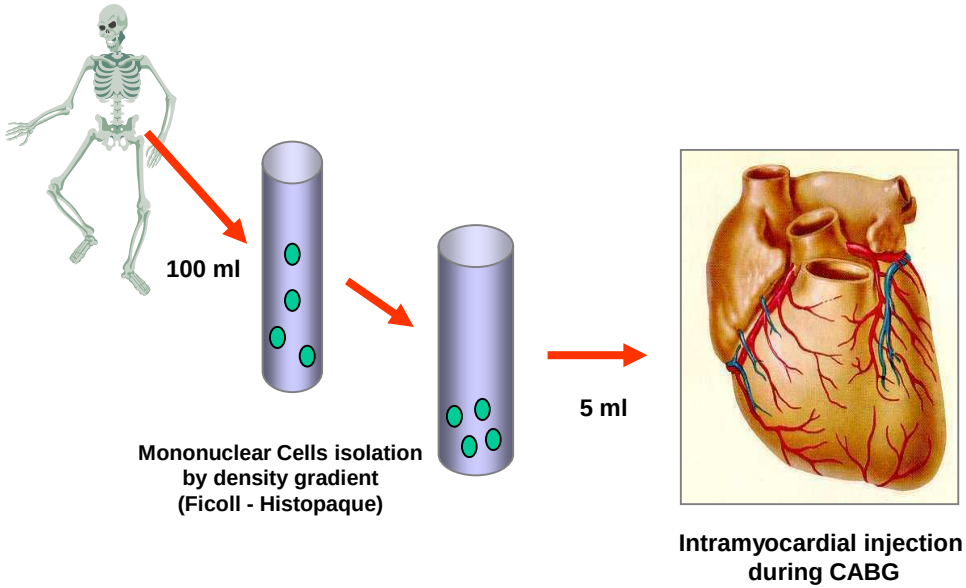
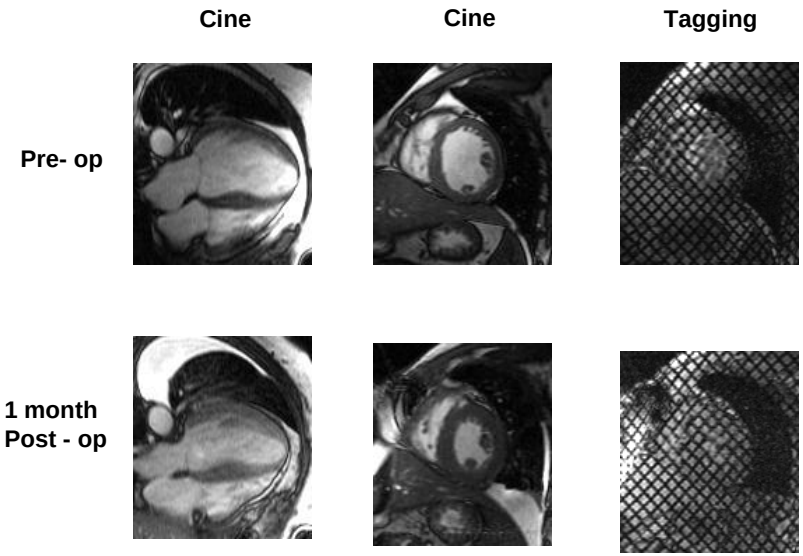
# Coronary Artery Disease

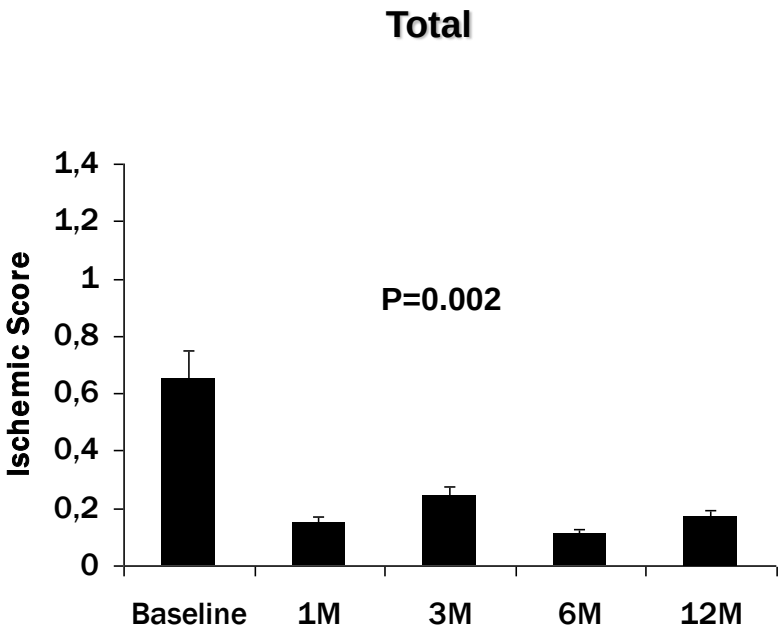


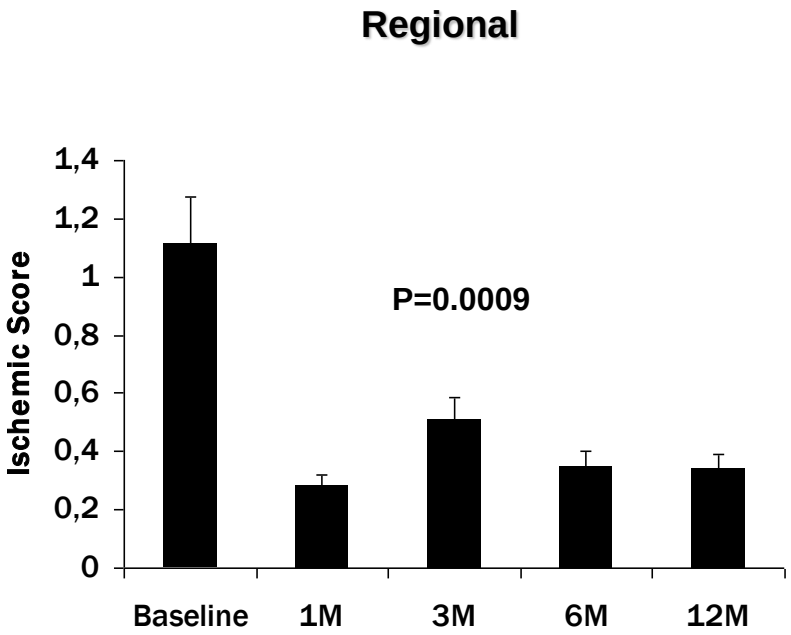




**Therapy:** Angioplasty
CABG
Biologic Repair



# Total and Regional LV Ischemic Score Assessed by MRI
# &
# BMC Injection




Gowdac LH, et al. *Clinics* 2008

# Efficacy – Controlled Double Blind Randomized Trials:

1. BMSC in Chronic CAD (Incomplete CABG) (N=140, 1:1)

2. BMSC in Chronic CAD (TMLR) (N=50, 1:1)