Nuovi farmaci biologici per riparare cuori infranti

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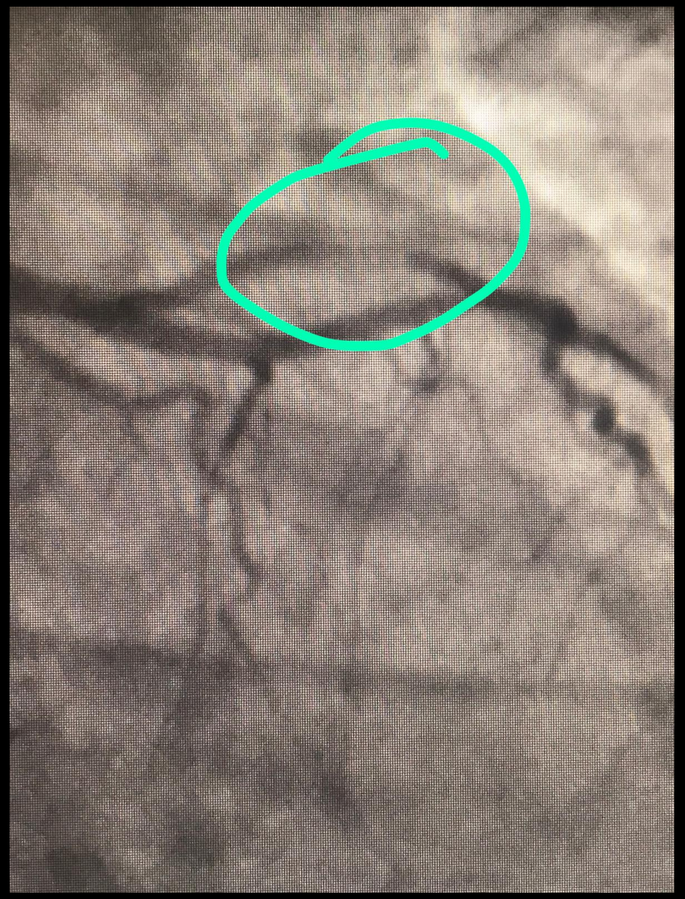
UNIVERSITÀ DEGLI STUDI DI TRIESTE

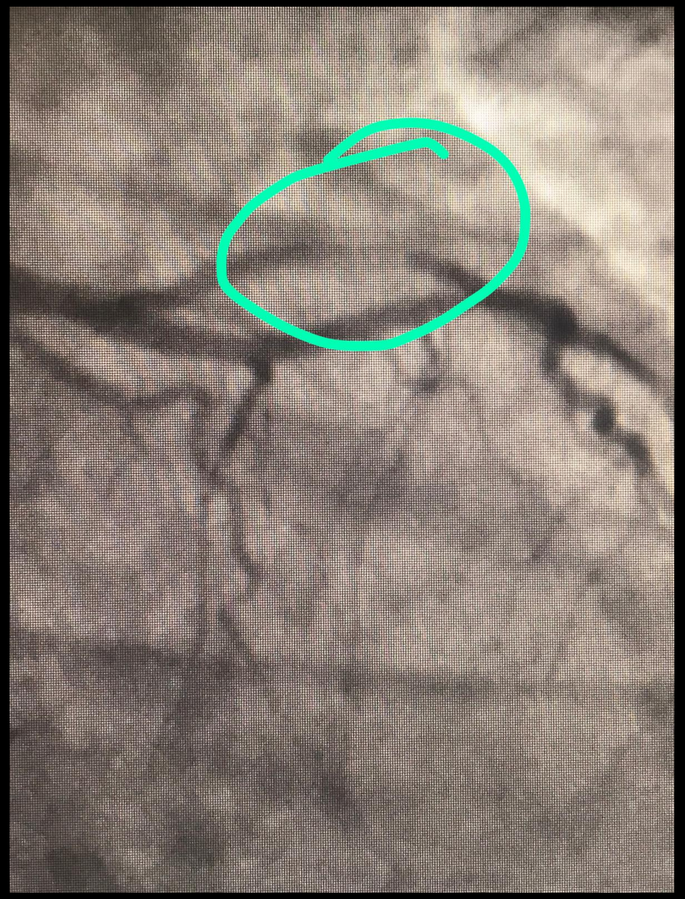
International Centre for Genetic Engineering and Biotechnology (ICGEB)

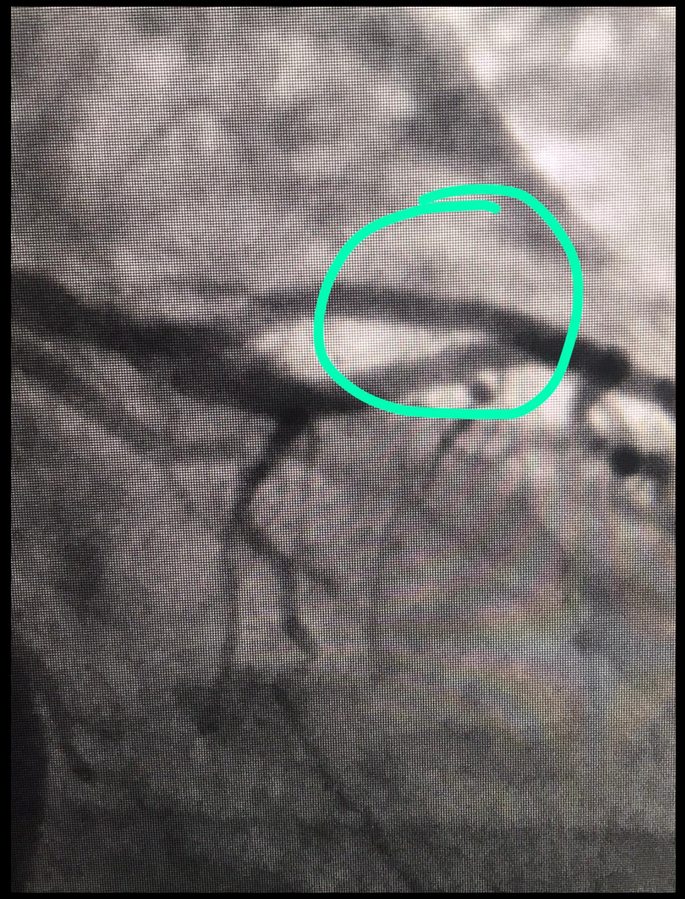
Una storia personale… uomo, 75 anni



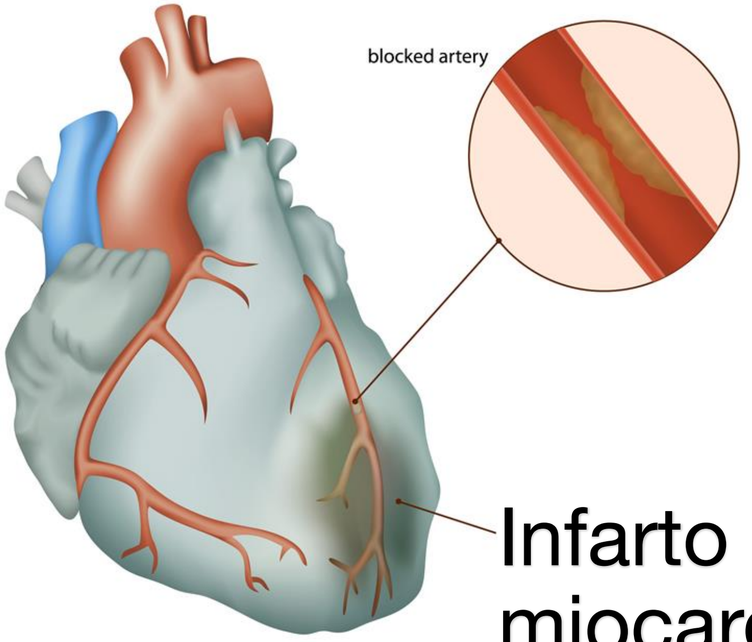
stenosi del 90%
di una arteria
coronaria

Una storia personale… uomo, 75 anni

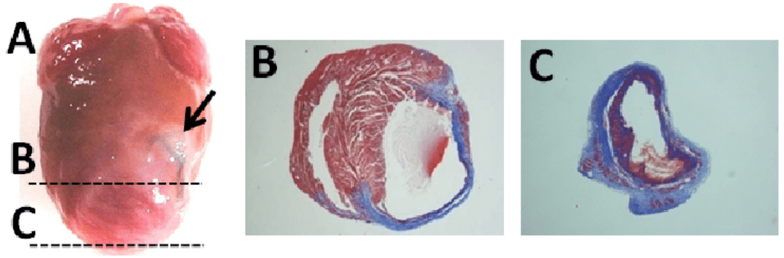




Cosa sarebbe successo se non rivascolarizzato in tempo?



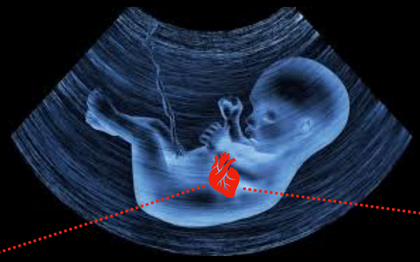
Infarto del miocardio

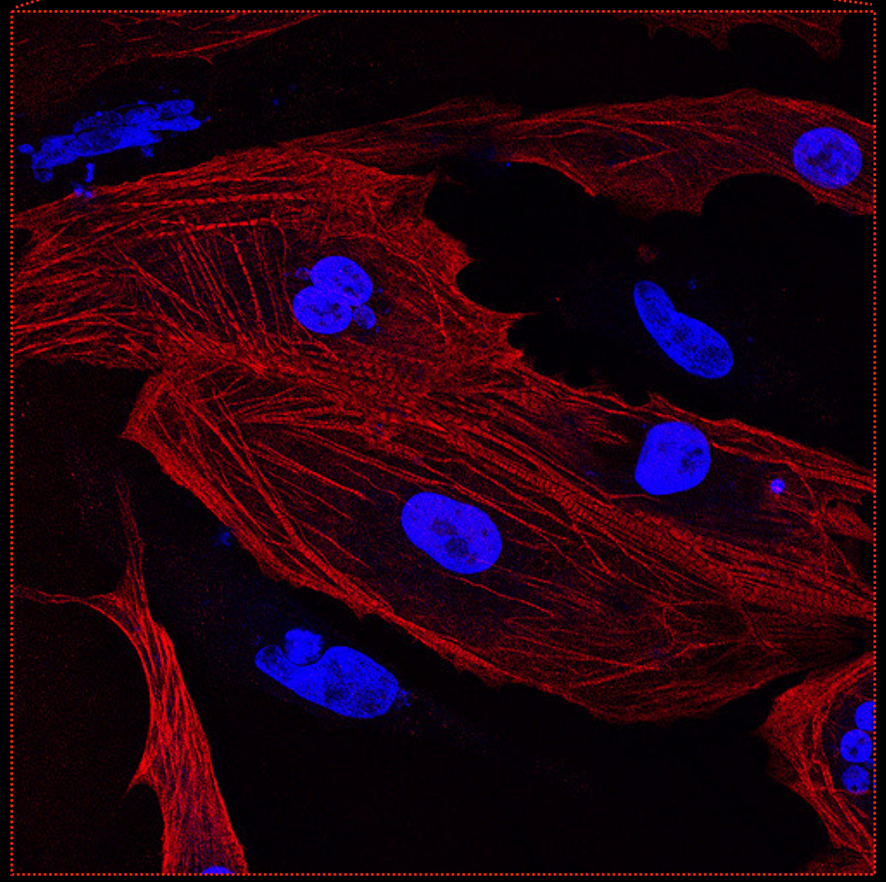


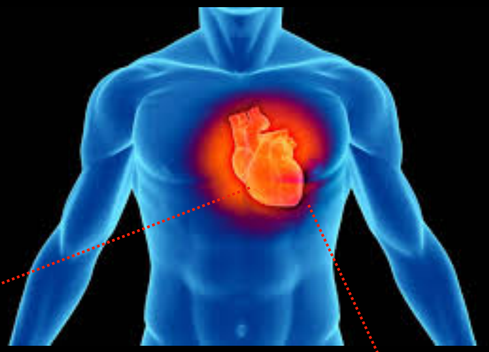
→ Perdita di tessuto contrattile

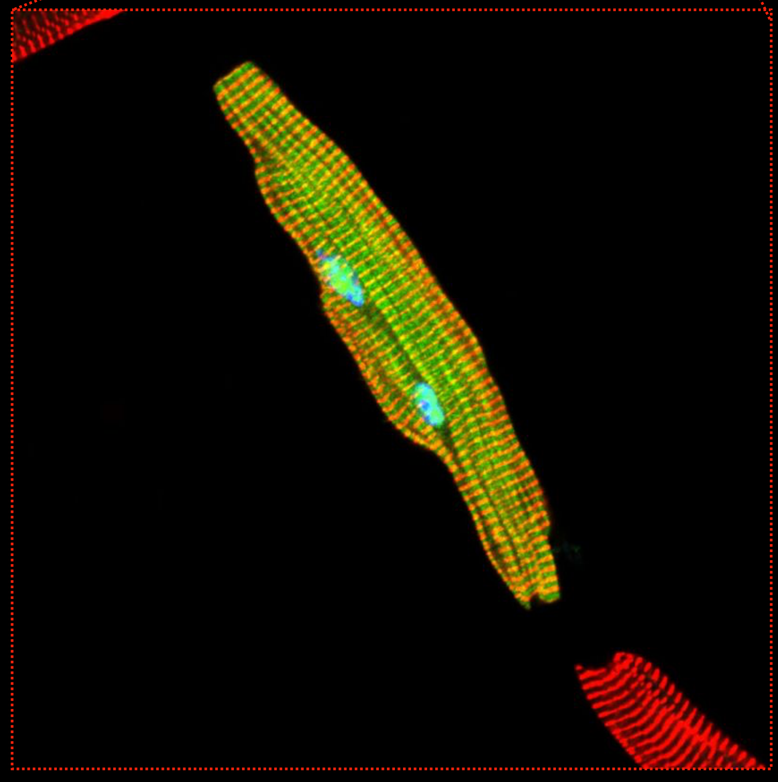
→ Scompenso cardiaco

Il cuore dei mammiferi perde la capacità di rigenerarsi alla nascita

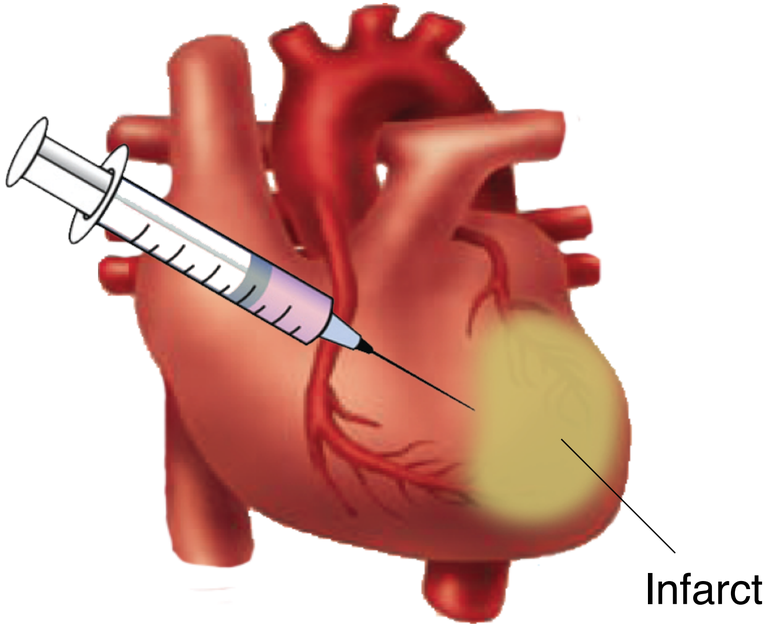








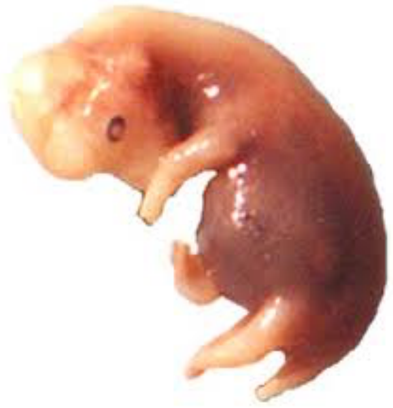
Il problema principale delle cardiopatie



2-4 miliardi di cardiomiociti muoiono in seguito ad un infarto del miocardio

Servono nuovi farmaci per ridurne la morte e/o promuoverne la rigenerazione

I cardiomiociti smettono di proliferare alla nascita: perchè?



Zebrafish/Mammiferi durante lo sviluppo
- proliferazione dei cardiomiociti
- rigenerazione cardiaca

- ambiente ipossico
- metabolismo glicolitico
- bassa pressione sanguigna

Mammiferi adulti
- uscita dei cardiomiociti dal ciclo cellulare
- assenza di rigenerazione cardiaca

- ambiente ricco di ossigeno
- metabolismo ossidativo
- alta pressione sanguigna

Programma genetico intrinseco?

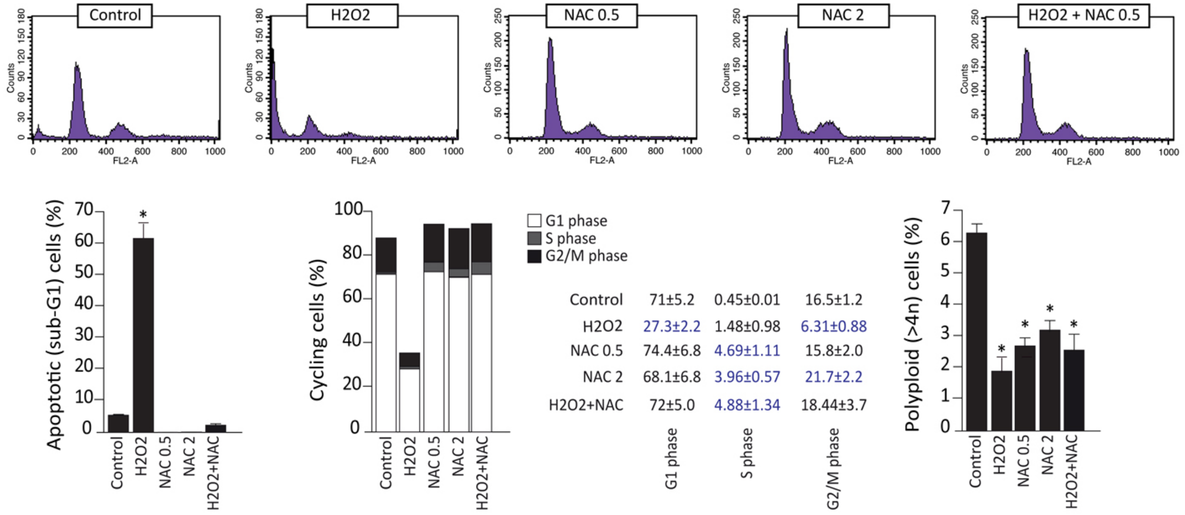
Controllo estrinseco?

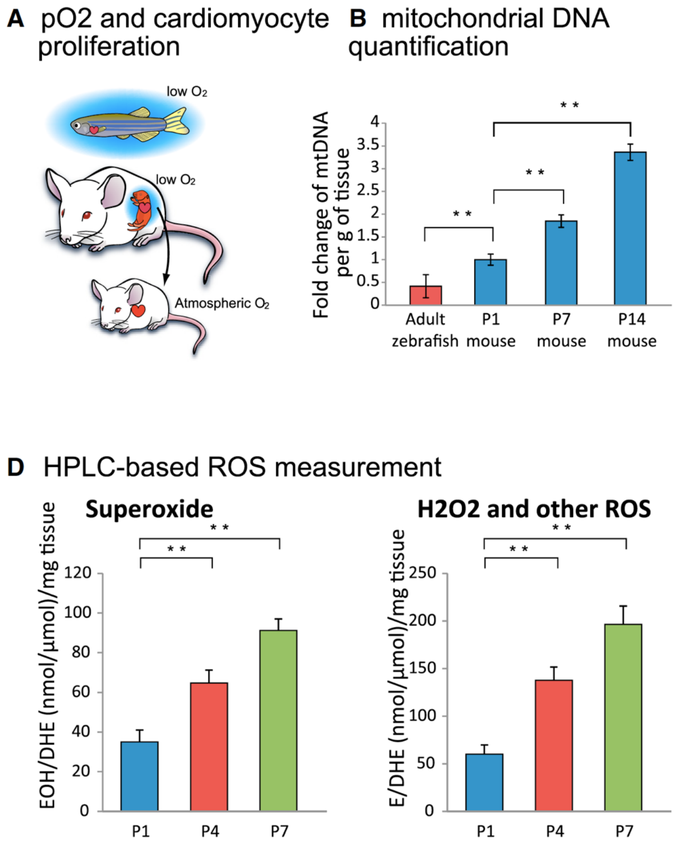
- forze meccaniche?
- shock iperossico?
- assenza di esposizione alla circolazione materna?

Zacchigna et al., Cardiovascular Research 2014

The Oxygen-Rich Postnatal Environment Induces Cardiomyocyte Cell-Cycle Arrest through DNA Damage Response

Bao N. Puente,[1,3,12] Wataru Kimura,[1,12] Shalini A. Muralidhar,[1] Jesung Moon,[3] James F. Amatruda,[1,2,3] Kate L. Phelps,[4] David Grinsfelder,[5] Beverly A. Rothermel,[1,2] Rui Chen,[1] Joseph A. Garcia,[1] Celio X. Santos,[7] SuWannee Thet,[1] Eiichiro Mori,[6] Michael T. Kinter,[8] Paul M. Rindler,[8] Serena Zacchigna,[9] Shibani Mukherjee,[6] David J. Chen,[6] Ahmed I. Mahmoud,[11] Mauro Giacca,[9] Peter S. Rabinovitch,[10] Asaithamby Aroumougame,[6] Ajay M. Shah,[7] Luke I. Szweda,[8] and Hesham A. Sadek[1,*]





Puente et al., Cell 2015

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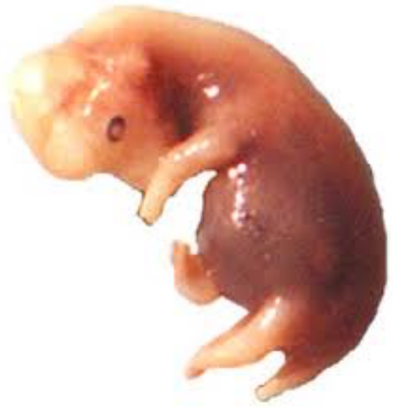

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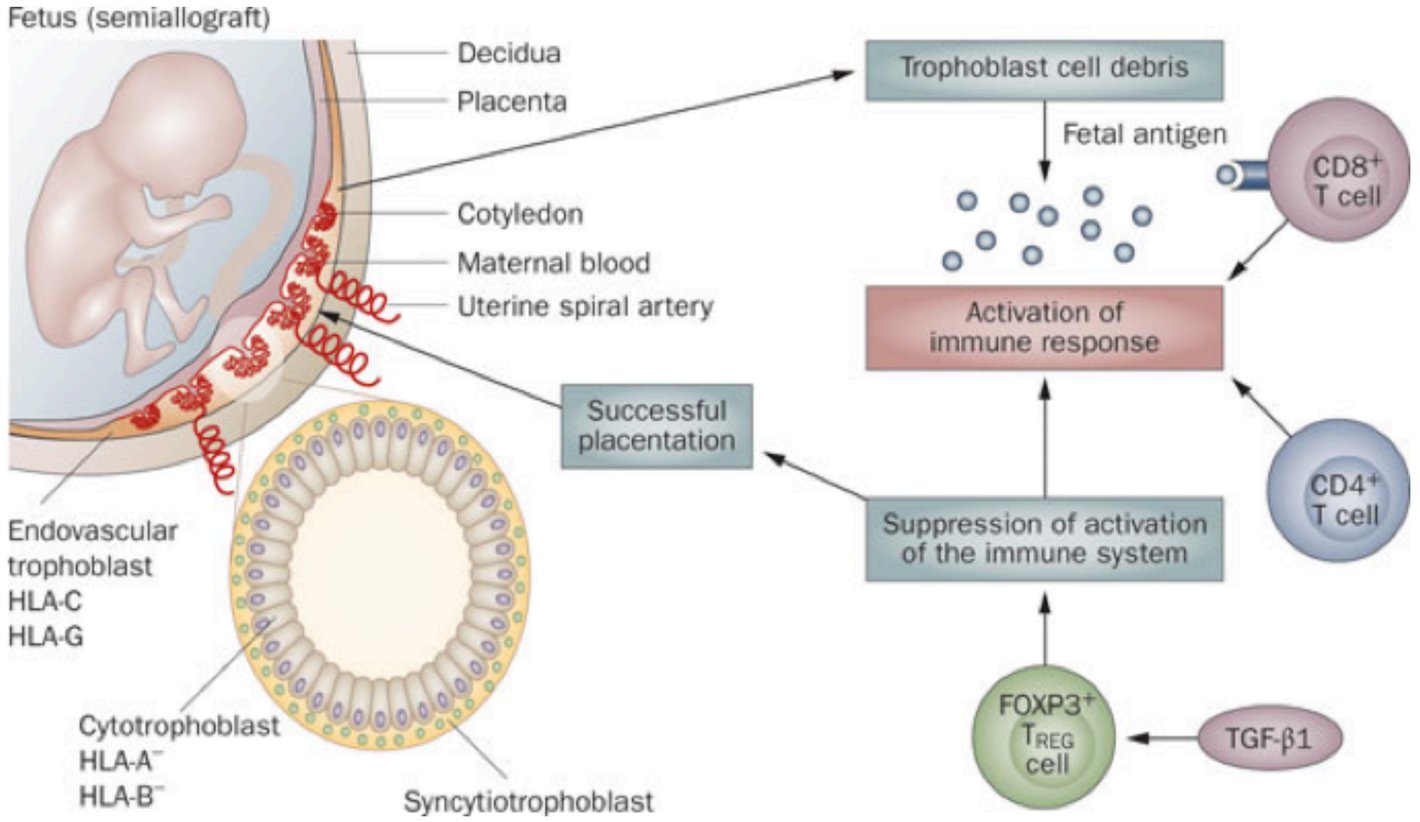
Zacchigna et al., Cardiovascular Research 2014

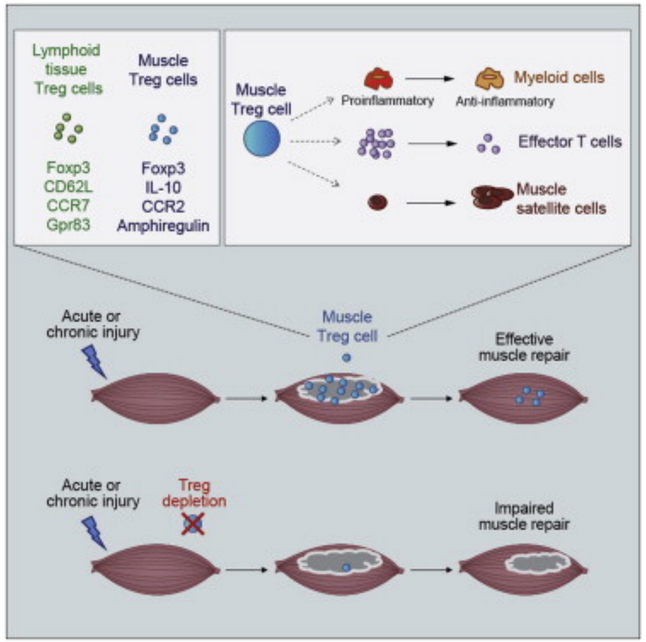
Le cellule T regolatorie (T-regs) sono espanse nel sangue materno in gravidanza per consentire la tolleranza nei confronti degli antigeni fetali

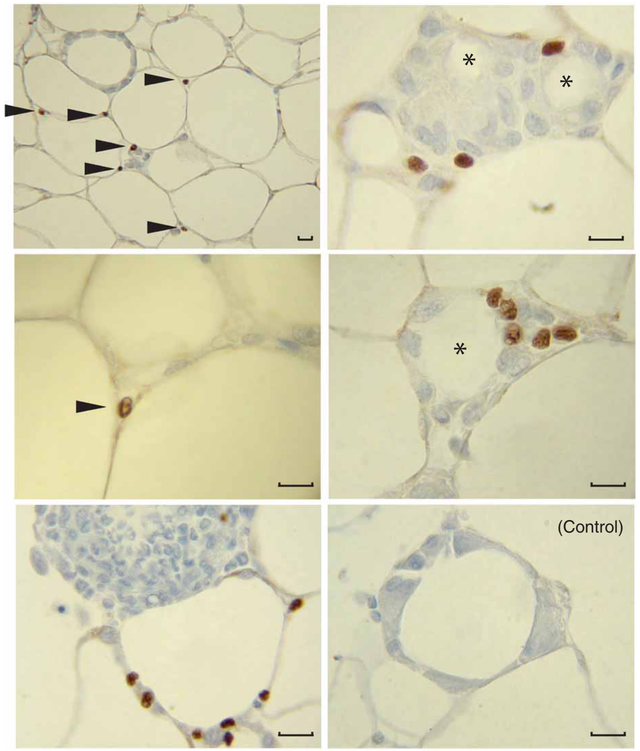


Funzioni extra-immunitarie delle Tregs

A Special Population of Regulatory T Cells Potentiates Muscle Repair

Dalia Burzyn,[1] Wilson Kuswanto,[1] Dmitriy Kolodin,[1] Jennifer L. Shadrach,[2,3] Massimiliano Cerletti,[2] Young Jang,[2] Esen Sefik,[1] Tze Guan Tan,[1] Amy J. Wagers,[2,3] Christophe Benoist,[1] and Diane Mathis[1,*]
[1]Microbiology and Immunobiology, Harvard Medical School, Boston, MA 02115, USA
[2]Stem Cell and Regenerative Biology, Harvard University, Cambridge, MA 02138, USA
[3]Howard Hughes Medical Institute, Chevy Chase, MD 20815, USA
*Correspondence: cbdm@hms.harvard.edu
http://dx.doi.org/10.1016/j.cell.2013.10.054





Lean, but not obese, fat is enriched for a unique population of regulatory T cells that affect metabolic parameters

Markus Feuerer[1,5], Laura Herrero[2,5], Daniela Cipolletta[1,4,5], Afia Naaz[2], Jamie Wong[1,5], Ali Nayer[2], Jongsoon Lee[2], Allison B Goldfine[3], Christophe Benoist[1,5], Steven Shoelson[2] & Diane Mathis[1,5]

VOLUME 15 | NUMBER 8 | AUGUST 2009 NATURE MEDICINE

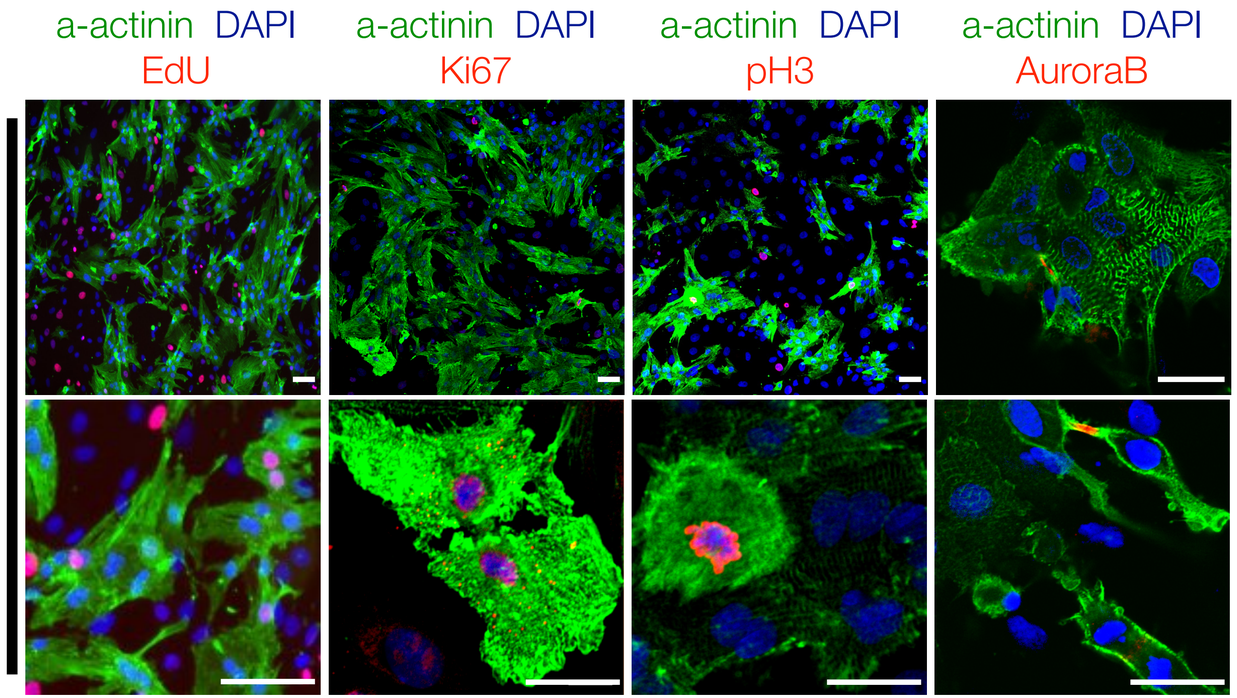
Tregs quale sorgente di fattori solubili responsabili della proliferazione dei cardiomiociti?

ARTICLE

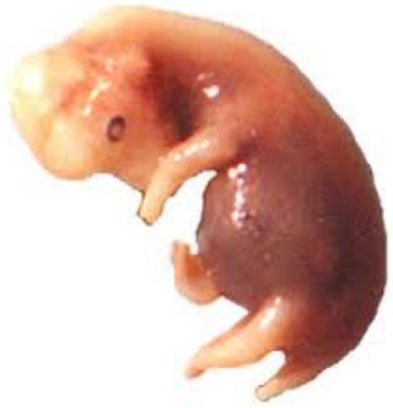
DOI: 10.1038/s41467-018-04908-z OPEN

Paracrine effect of regulatory T cells promotes cardiomyocyte proliferation during pregnancy and after myocardial infarction

Serena Zacchigna[1,2], Valentina Martinelli[3], Silvia Moimas[2,3], Andrea Colliva[1], Marco Anzini[2], Andrea Nordio[2], Alessia Costa[1], Michael Rehman[1], Simone Vodret[1], Cristina Pierro[1], Giulia Colussi[3], Lorena Zentilin[3], Maria Ines Gutierrez[3], Ellen Dirkx[3], Carlin Long[3], Gianfranco Sinagra[2], David Klatzmann[4,5] & Mauro Giacca[2,3]



I cardiomiociti smettono di proliferare alla nascita: perchè?



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- assenza di esposizione alla circolazione materna?

Zacchigna et al., Cardiovascular Research 2014

Lo scarico meccanico del cuore dopo impianto di LVAD stimola la proliferazione dei cardiomiociti umani

Human Ventricular Unloading Induces Cardiomyocyte Proliferation

Diana C. Canseco, PhD,* Wataru Kimura, PhD,* Sonia Garg, MD,* Shibani Mukherjee, PhD,†
Souparno Bhattacharya, MS,‡ Salim Abdisalaam, PhD,‡ Sandeep Das, MD,* Aroumougame Asaithamby, PhD,‡
Pradeep P.A. Mammen, MD,* Hesham A. Sadek, MD, PhD*

ABSTRACT

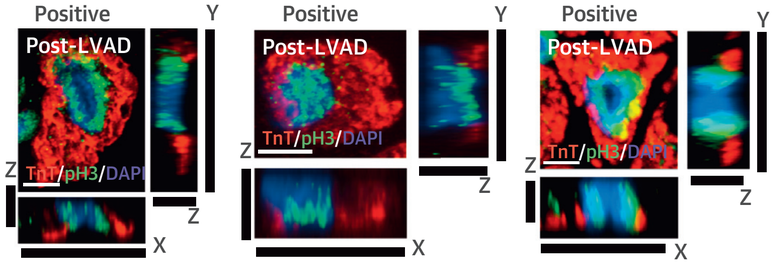
BACKGROUND The adult mammalian heart is incapable of meaningful regeneration after substantial cardiomyocyte loss, primarily due to the inability of adult cardiomyocytes to divide. Our group recently showed that mitochondria-mediated oxidative DNA damage is an important regulator of postnatal cardiomyocyte cell cycle arrest. However, it is not known whether mechanical load also plays a role in this process. We reasoned that the postnatal physiological increase in mechanical load contributes to the increase in mitochondrial content, with subsequent activation of DNA damage response (DDR) and permanent cell cycle arrest of cardiomyocytes.

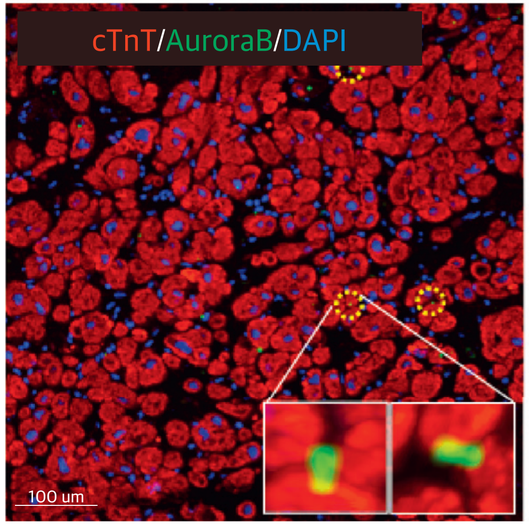
OBJECTIVES The purpose of this study was to test the effect of mechanical unloading on mitochondrial mass, DDR, and cardiomyocyte proliferation.

METHODS We examined the effect of human ventricular unloading after implantation of left ventricular assist devices (LVADs) on mitochondrial content, DDR, and cardiomyocyte proliferation in 10 matched left ventricular samples collected at the time of LVAD implantation (pre-LVAD) and at the time of explantation (post-LVAD).

RESULTS We found that post-LVAD hearts showed up to a 60% decrease in mitochondrial content and up to a 45% decrease in cardiomyocyte size compared with pre-LVAD hearts. Moreover, we quantified cardiomyocyte nuclear foci of phosphorylated ataxia telangiectasia mutated protein, an upstream regulator of the DDR pathway, and we found a significant decrease in the number of nuclear phosphorylated ataxia telangiectasia mutated foci in the post-LVAD hearts. Finally, we examined cardiomyocyte mitosis and cytokinesis and found a statistically significant increase in both phosphorylated histone H3–positive, and Aurora B–positive cardiomyocytes in the post-LVAD hearts. Importantly, these results were driven by statistical significance in hearts exposed to longer durations of mechanical unloading.

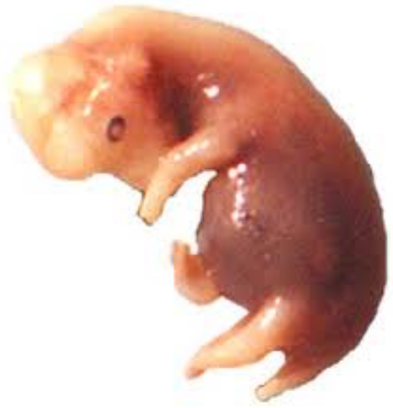
CONCLUSIONS Prolonged mechanical unloading induces adult human cardiomyocyte proliferation, possibly through prevention of mitochondria-mediated activation of DDR. (J Am Coll Cardiol 2015;65:892–900) © 2015 by the American College of Cardiology Foundation.





JACC VOL. 65, NO. 9, 2015
MARCH 10, 2015:892–900

I cardiomiociti smettono di proliferare alla nascita: è davvero un dogma?



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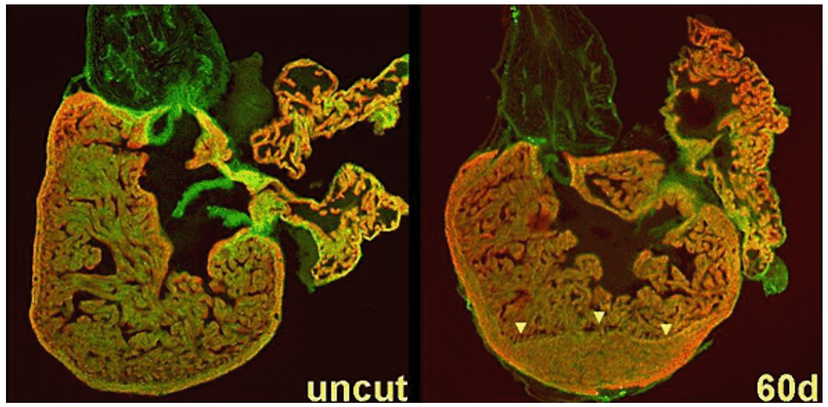
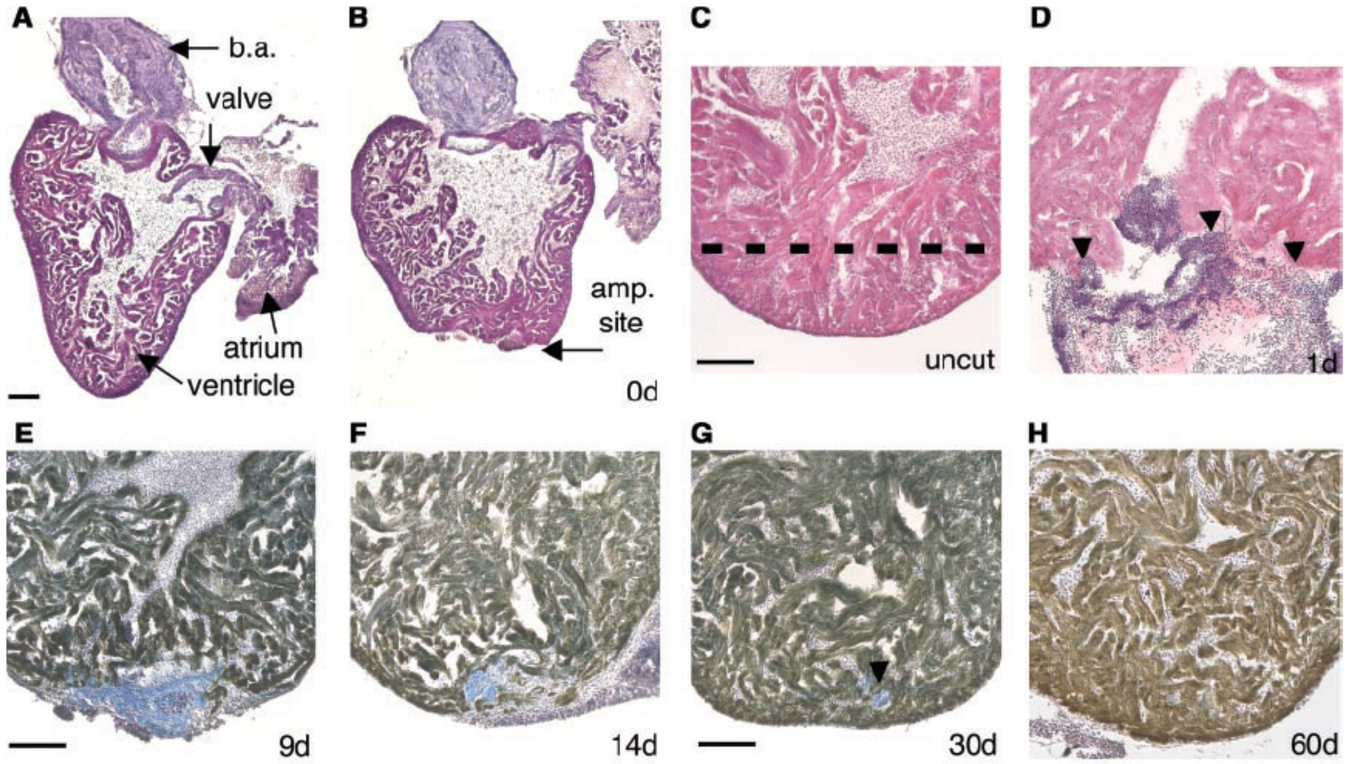
Zacchigna et al., Cardiovascular Research 2014

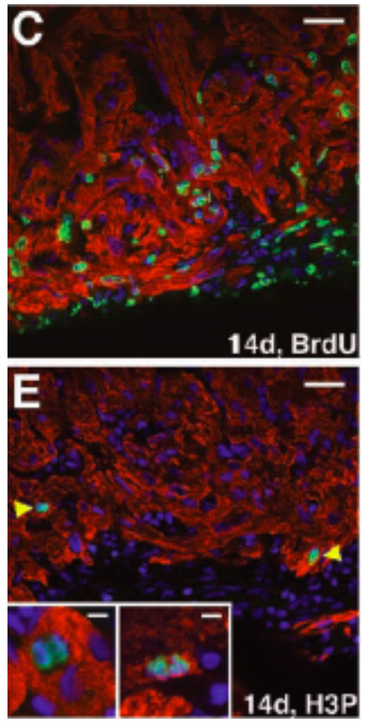
Heart Regeneration in Zebrafish

Kenneth D. Poss,* Lindsay G. Wilson, Mark T. Keating*

Cardiac injury in mammals and amphibians typically leads to scarring, with minimal regeneration of heart muscle. Here, we demonstrate histologically that zebrafish fully regenerate hearts within 2 months of 20% ventricular resection. Regeneration occurs through robust proliferation of cardiomyocytes localized at the leading epicardial edge of the new myocardium. The hearts of zebrafish with mutations in the Mps1 mitotic checkpoint kinase, a critical cell cycle regulator, failed to regenerate and formed scars. Thus, injury-induced cardiomyocyte proliferation in zebrafish can overcome scar formation, allowing cardiac muscle regeneration. These findings indicate that zebrafish will be useful for genetically dissecting the molecular mechanisms of cardiac regeneration.

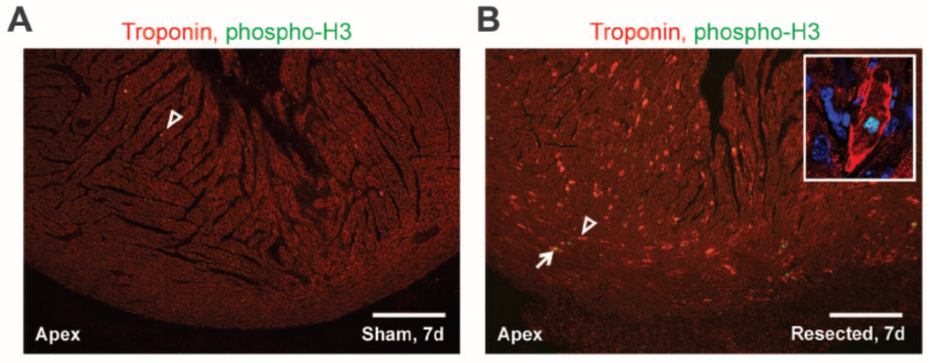


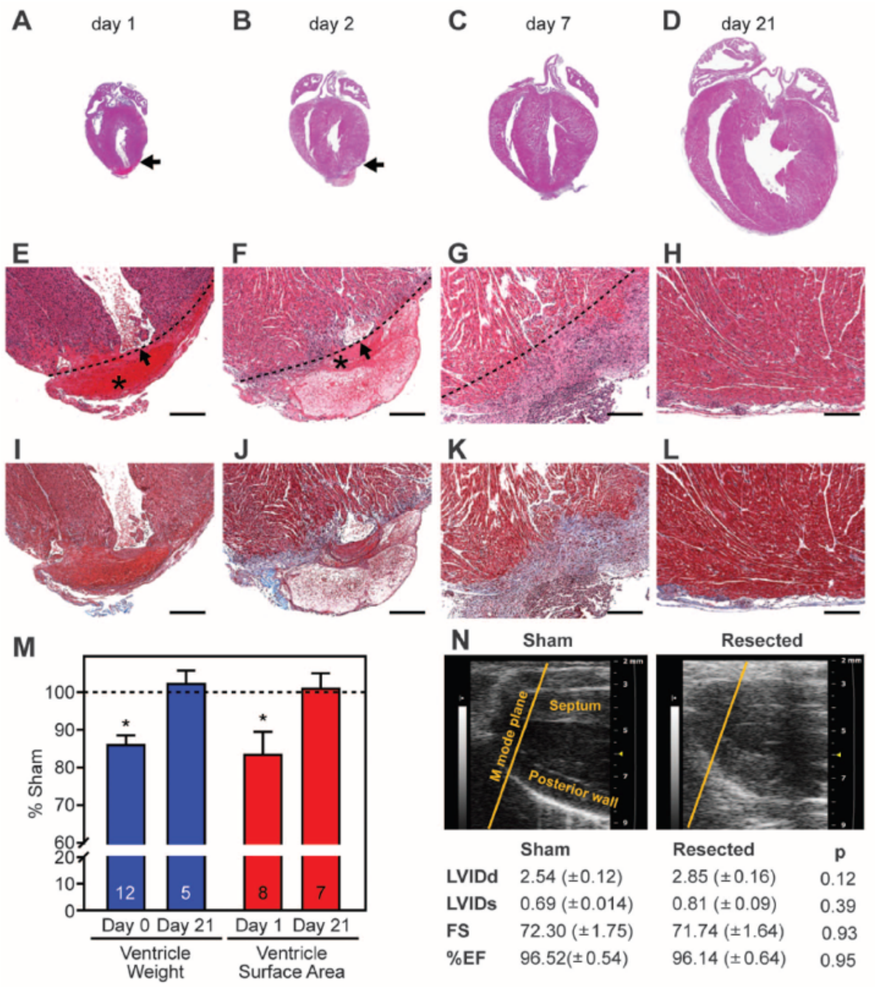




Transient Regenerative Potential of the Neonatal Mouse Heart

Enzo R. Porrello,[1] Ahmed I. Mahmoud,[2] Emma Simpson,[3] Joseph A. Hill,[1,2] James A. Richardson,[1,3]
Eric N. Olson,[1]* Hesham A. Sadek[2]*



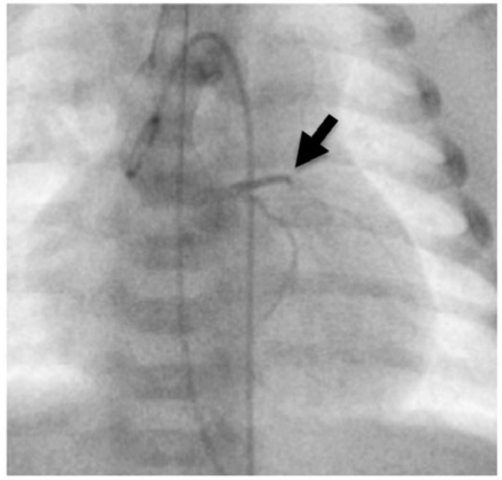


Caso clinico

- Bambino nato a termine (39 settimane), parto eutocico, ossigenazione arteria ombelicale ok
- Alla nascita compare cianosi severa, ridotta saturazione di ossigeno
- ECG: segni di ischemia acuta
- Echocardiografia: severa disfunzione ventricolo sinistro
- Aumento dei livelli di BNP, Troponina T and CK nel sangue

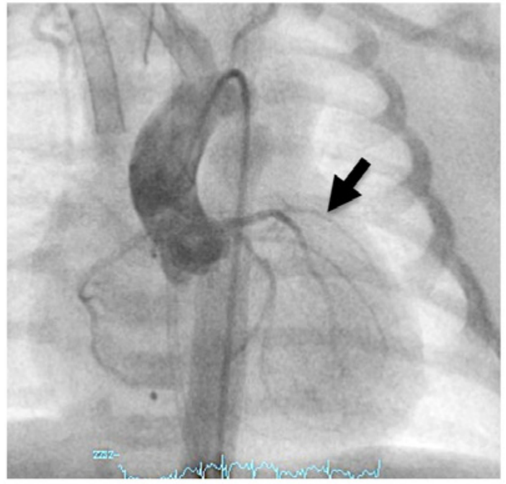
- Angiografia coronarica

Haubner et al., Circulation Research 2016



Occlusione completa della arteria coronaria discendente anteriore

- Trombolisi a 28 ore dall'inizio dei sintomi



Completa riapertura dell'arteria dopo 3 giorni

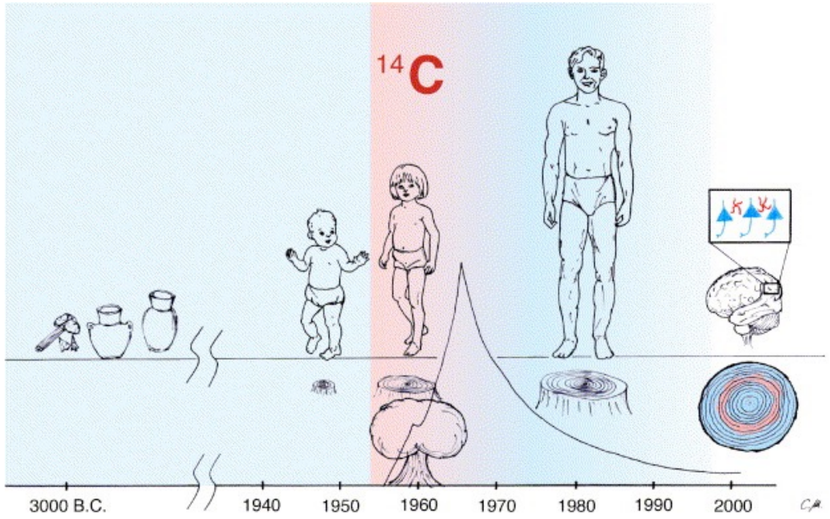
Segni persistenti di danno miocardico evidenti in echocardiogragia, ECG e analisi ematiche

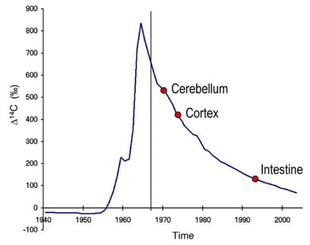
Diagnosi: occlusione completa della DS per oltre 20 ore, IMA massivo

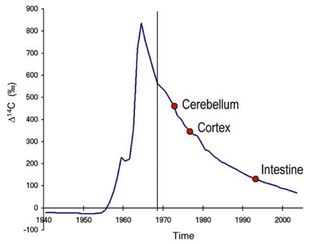
Evoluzione del piccolo paziente?

1. Recupero completo della funzione cardiaca a 45 giorni
2. Segni di disfunzione cardiaca persistente a visite periodiche di controllo
3. Scompenso cardiaco a 1 anno
4. Morte a 2 mesi

Esperimenti basati sul C14 indicano che il cuore umano rinnova il 50% dei suoi cardiomiociti in una vita media





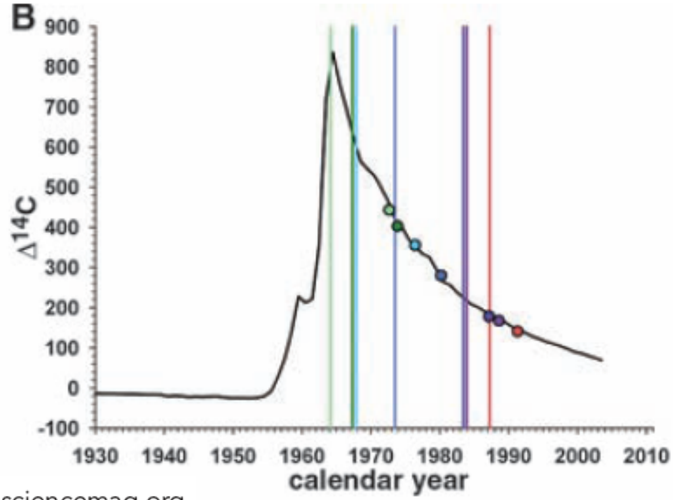


Evidence for Cardiomyocyte Renewal in Humans

Olaf Bergmann,[1]* Ratan D. Bhardwaj,[1]* Samuel Bernard,[2] Sofia Zdunek,[1] Fanie Barnabé-Heider,[1] Stuart Walsh,[3] Joel Zupicich,[1] Kanar Alkass,[4] Bruce A. Buchholz,[5] Henrik Druid,[4] Stefan Jovinge,[3,6] Jonas Frisén[1]†

A 25-year-old heart replaces about 1% of all cardiomyocytes over a year; a 75-year-old about half that.

Fewer than 50% of cardiomyocytes are exchanged during a normal life span.



Treatments and drugs

By Mayo Clinic staff

return to home MayoClinic.com

Heart failure is a chronic disease needing lifelong management. However, with treatment, signs and symptoms of heart failure can improve and the heart sometimes becomes stronger. Doctors sometimes can correct heart failure by treating the underlying cause. For example, repairing a heart valve or controlling a fast heart rhythm may reverse heart failure. But for most people, the treatment of heart failure involves a balance of the right medications, and in some cases, devices that help the heart beat and contract properly.

Medications

Doctors usually treat heart failure with a combination of medications. Depending on your symptoms, you might take one or more of these drugs. They include:

- **Angiotensin-converting enzyme (ACE) inhibitors.** These drugs help people with heart failure live longer and feel better. ACE inhibitors are a type of vasodilator, a drug that widens blood vessels to lower blood pressure, improve blood flow and decrease the workload on the heart. Examples include enalapril (Vasotec), lisinopril (Prinivil, Zestril) and captopril (Capoten).
- **Angiotensin II receptor blockers (ARBs).** These drugs, which include losartan (Cozaar) and valsartan (Diovan), have many of the same benefits as ACE inhibitors. They may be an alternative for people who can't tolerate ACE inhibitors.
- **Digoxin (Lanoxin).** This drug, also referred to as digitalis, increases the strength of your heart muscle contractions. It also tends to slow the heartbeat. Digoxin reduces heart failure symptoms and improves your ability to live with the condition.
- **Beta blockers.** This class of drugs slows your heart rate and reduces blood pressure. Examples include carvedilol (Coreg), metoprolol (Lopressor) and bisoprolol (Zebeta). These medicines also reduce the risk of some abnormal heart rhythms. Beta blockers may reduce signs and symptoms of heart failure and improve heart function.
- **Diuretics.** Often called water pills, diuretics make you urinate more frequently and keep fluid from collecting in your body. Commonly prescribed diuretics for heart failure include bumetanide (Bumex) and furosemide (Lasix). The drugs also decrease fluid in your lungs, so you can breathe more easily. Because diuretics make your body lose potassium and magnesium, your doctor may also prescribe supplements of these minerals. If you're taking a diuretic, your doctor will likely monitor levels of potassium and magnesium in your blood through regular blood tests.
- **Aldosterone antagonists.** These drugs include spironolactone (Aldactone) and eplerenone (Inspra). They're primarily potassium-sparing diuretics, but they have additional properties that help the heart work better, may reverse scarring of the heart and may help people with severe heart failure live longer. Unlike some other diuretics, spironolactone can raise the level of potassium in your blood to dangerous levels, so talk to your doctor if increased potassium is a concern.

Terapia standard per lo scompenso cardiaco

Atkinson AB & Robertson JI. 1979. Captopril in the treatment of clinical hypertension and cardiac failure. Lancet 2, 836-9 — ‘70s

Gottlieb SS et al. 1993. Hemodynamic and neurohormonal effects of tghe angiotensin II antagonist Losartan in patients with congestive heart failure. Circulation 88, 1602-1609 — ‘90s

Whiting AJ. 1918. On the comparative value of the digitalis series of remedies in the heart failure of auricular fibrillation and the changes in the clinical features of mitral stenosis after fibrillation of the auricle. Proc R Soc Med 11, 1-52 — ‘10s

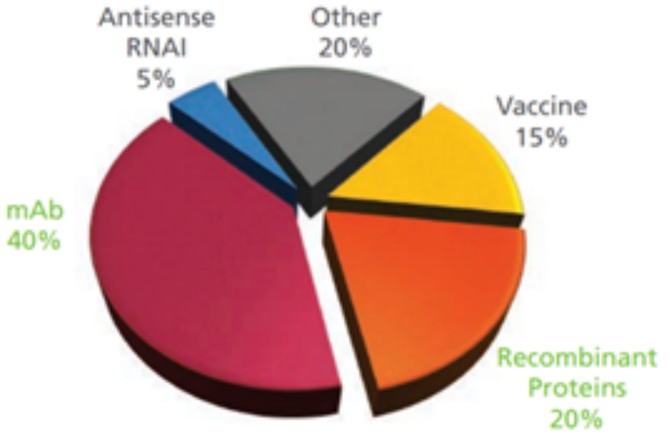
Swedberg K et al. 1979. Prolongation of survival in congestive cardiomyopathy by beta-receptor blockade. Lancet 30, 1374-6 — ‘70s

Marvin HM. 1927. Digitalis and diuretics in heart failure with regular rhythm, with espcial reference to the importance of etiologic classification of heart disease. J Clin Invest 3, 521-39 — ‘20s

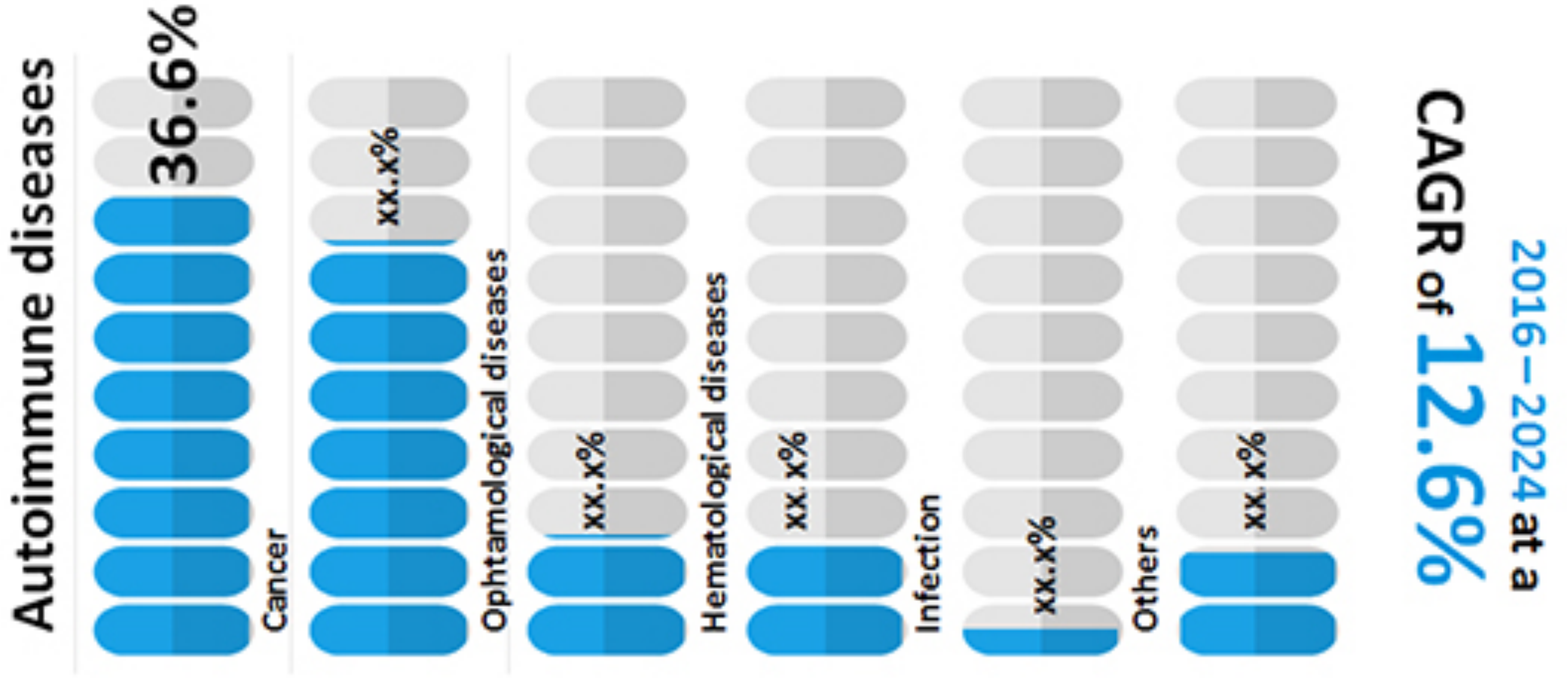
Goldberger E. 1965. Aldosterone and the edema of congestive heart failure. Am J Cardiol 15, 274 — ‘60s

LCZ696? SGLT2 inhibitors?

Farmaci biologici



Mercato globale degli anticorpi monoclonali per settore di applicazione (2016)



Farmaci biologici per le malattie cardiache

Proteine ricombinanti
Anticorpi monoclonali

ARTICLE

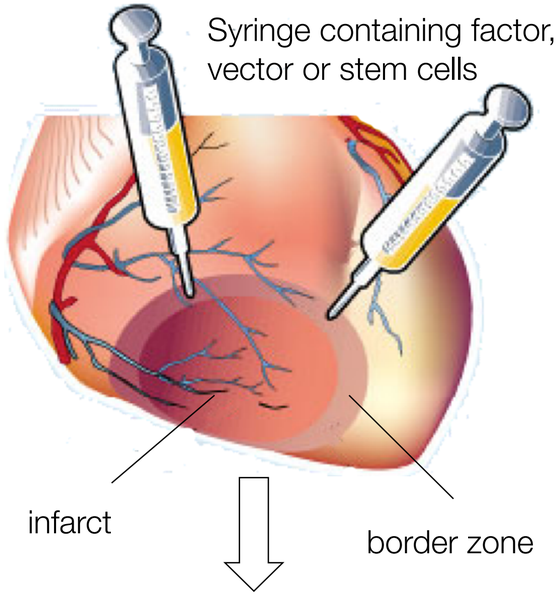
https://doi.org/10.1038/s41467-021-27622-9 OPEN

Bone morphogenetic protein 1.3 inhibition decreases scar formation and supports cardiomyocyte survival after myocardial infarction

Slobodan Vukicevic[1,10], Andrea Colliva[2,3,10], Vera Kufner[1], Valentina Martinelli[4], Silvia Moimas[4], Simone Vodret[2], Viktorija Rumenovic[1], Milan Milosevic[5], Boris Brkljacic[6], Diana Delic-Brkljacic[7], Ricardo Correa[2], Mauro Giacca[3,4,8], Manuel Maglione[9], Tatjana Bordukalo-Niksic[1], Ivo Dumic-Cule[1,11] & Serena Zacchigna[2,3,11]✉

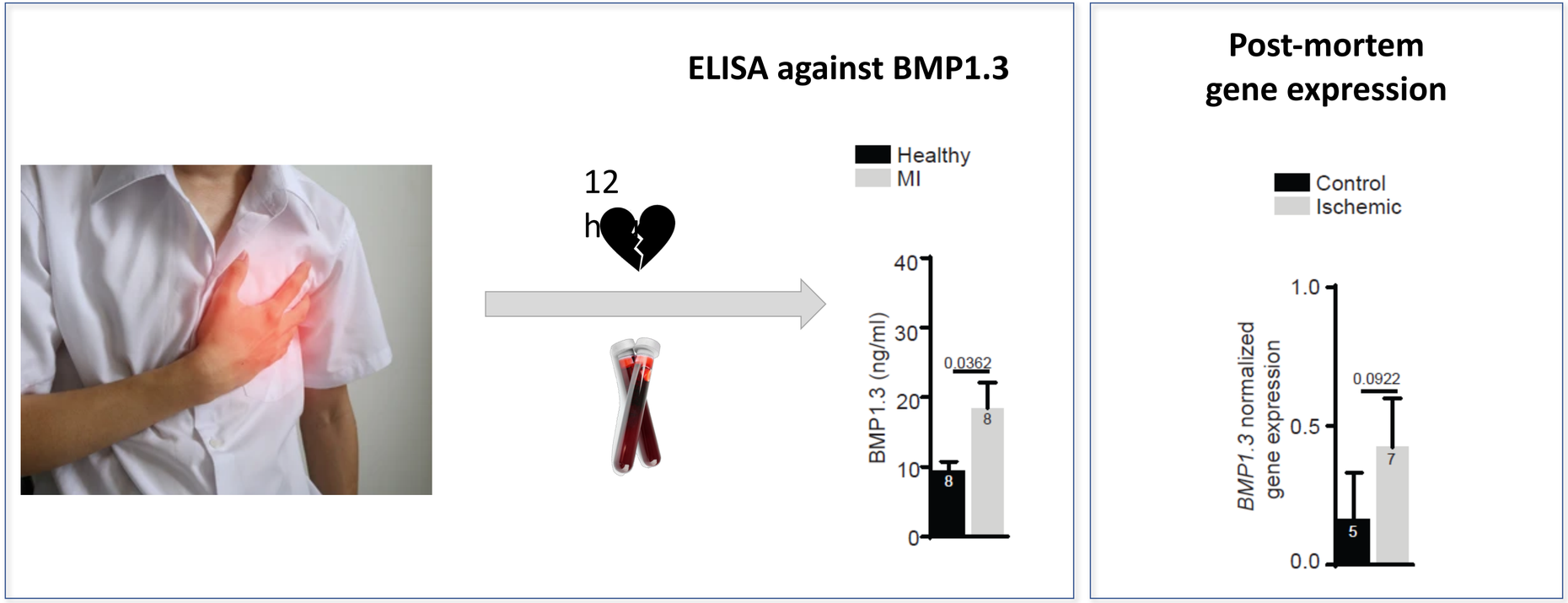
Terapia Genica
Vettori virali, microRNA

Terapia cellulare



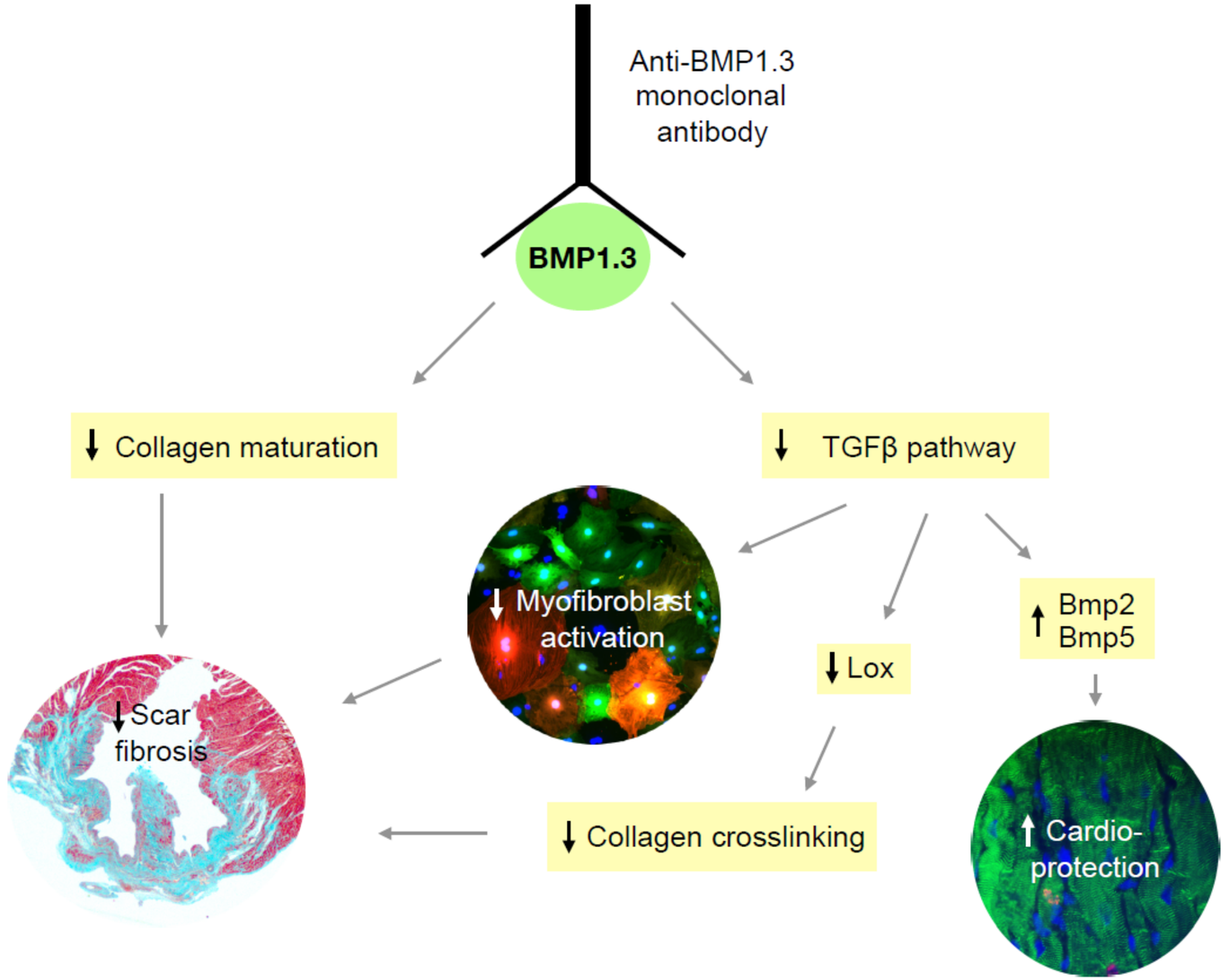
Neoangiogenesi Cardioprotezione Rigenerazione cardiaca

I livelli di BMP1.3 sono aumentati nel sangue dei pazienti con infarto acuto del miocardio



Vukicevic et al. 2022. Nat Comm, Jan 10;13(1)

Doppio meccanismo di azione di un nuovo anticorpo monoclonale anti-BMP1.3



Vukicevic et al. 2022. Nat Comm, Jan 10;13(1)

SVEUČILIŠTE U ZAGREBU MEDICINSKI FAKULTET

Prof. Slobodan Vukicevic, MD, PhD
University of Zagreb School of Medicine, Croatia

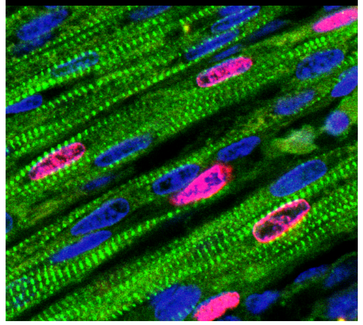
Farmaci biologici per le malattie cardiache

Proteine ricombinanti

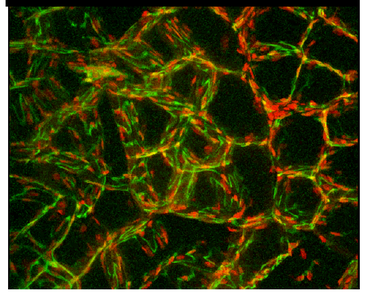
Terapia genica

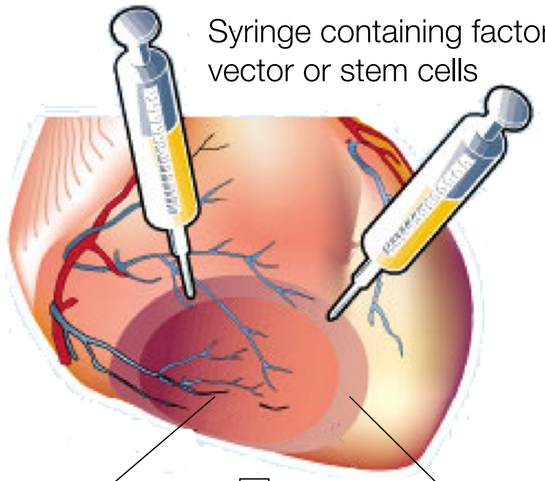
Terapia cellulare

microRNAs per la rigenerazione cardiaca

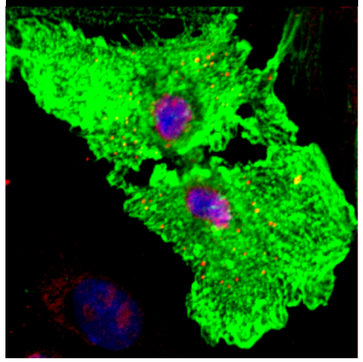


Vettori AAV per angiogenesi e cardioprotezione

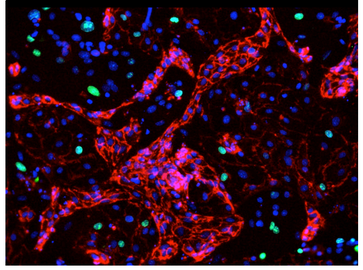




Tregs per stimolare la proliferazione dei CM

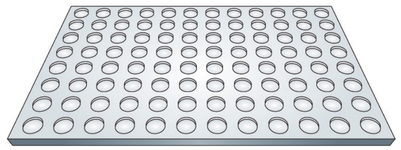


Cellule endoteliali per la rivascolarizzazione



Neoangiogenesi
Cardioprotezione
Rigenerazione cardiaca

Screening di microRNA per la proliferazione dei cardiomiociti

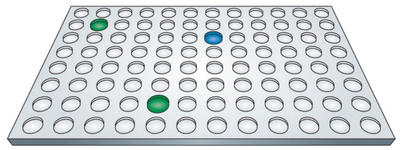


microRNA mimics
arrayed on 96-well plates
(988 mature sequences)

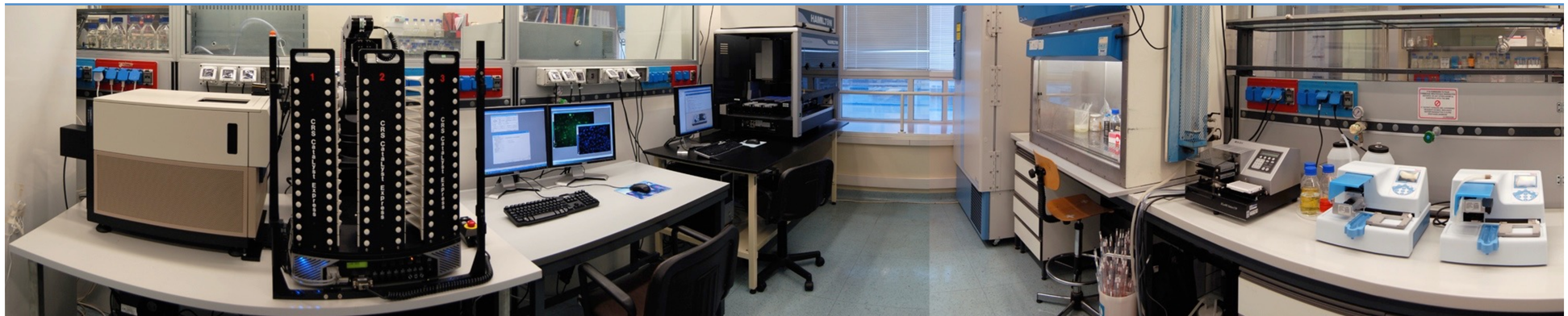
0h — add transfection reagent
add cardiomyocytes isolated from newborn rats

52h — add EdU

72h

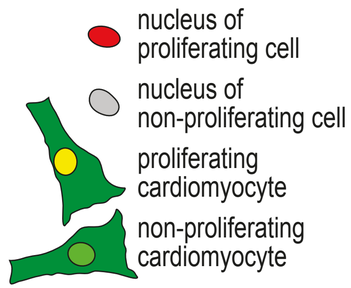


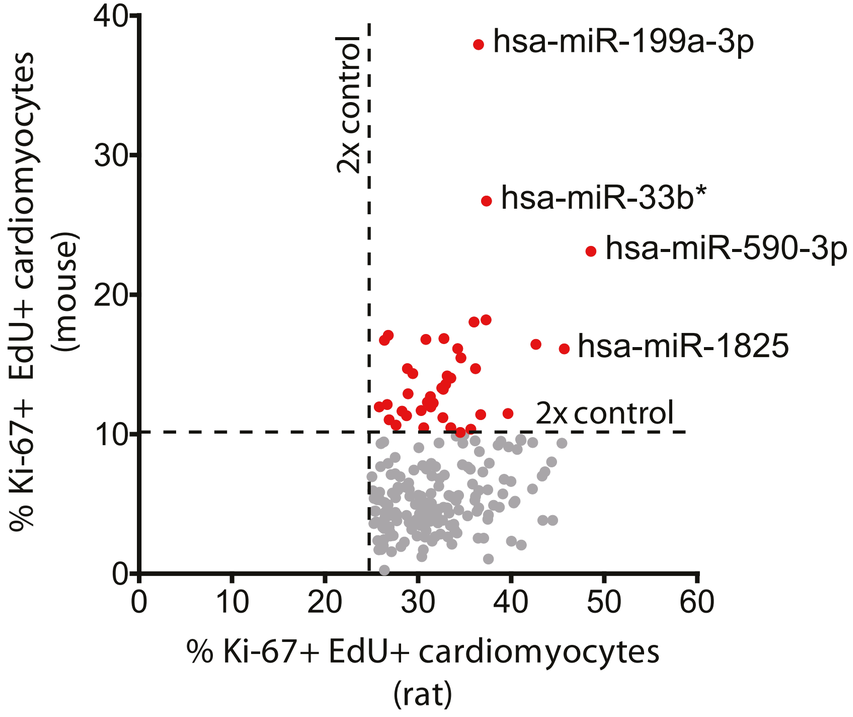
cell fixation and fluorescence staining
(Hoechst, alpha-actinin, Ki-67 and EdU)

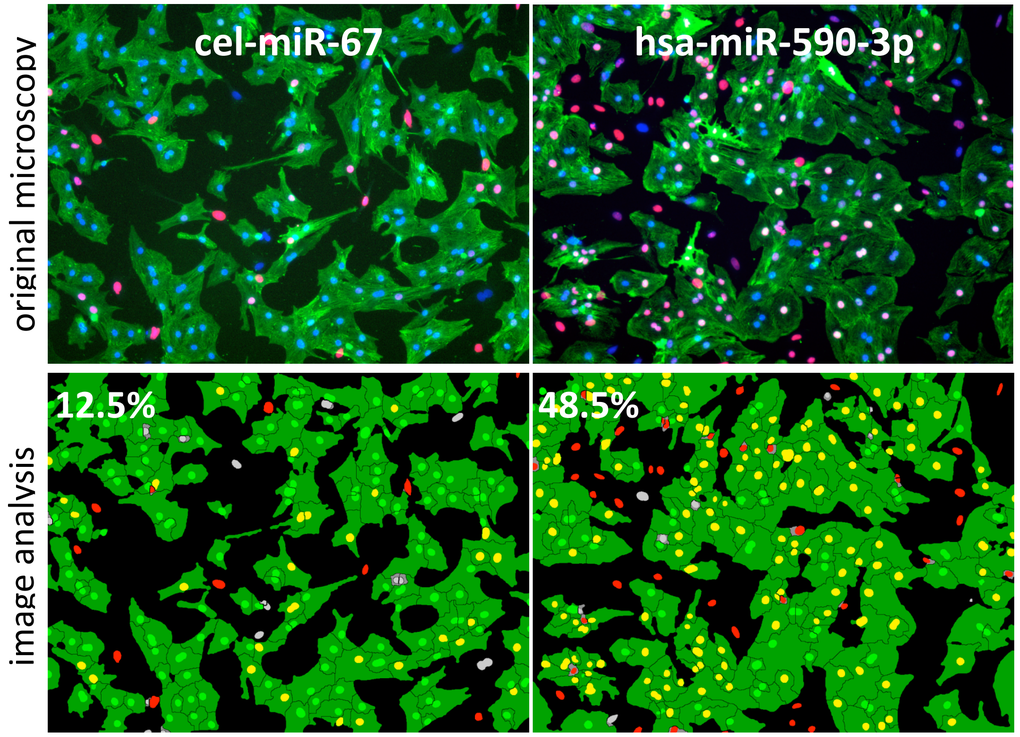


40 miRNA umano stimolano la proliferazione dei cardiomiociti

Hoechst
a-actinin
EdU



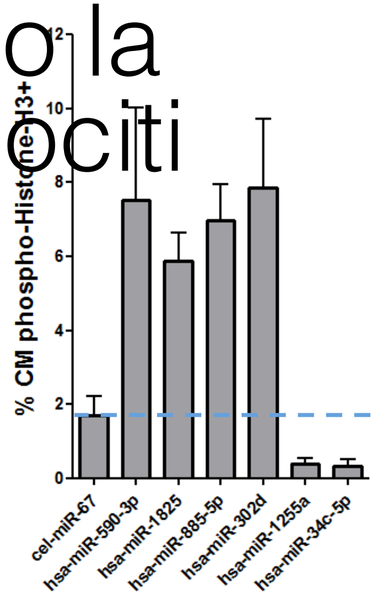


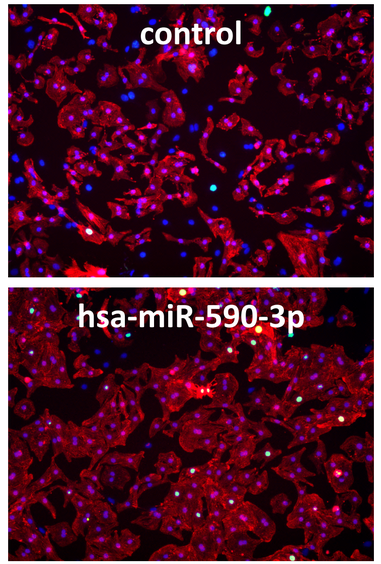


Eulalio et al. 2012. Nature 492, 376

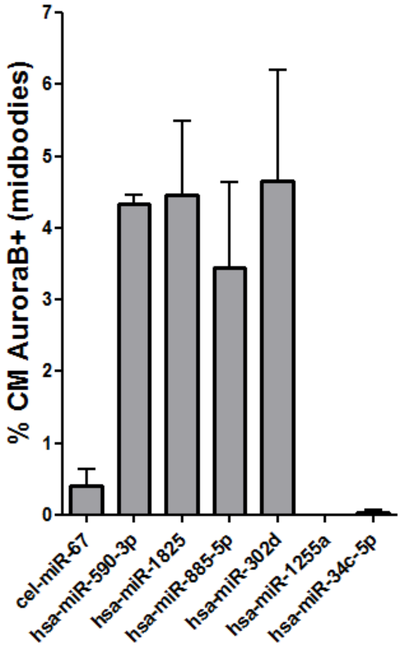
40 miRNA umano stimolano la proliferazione dei cardiomiociti

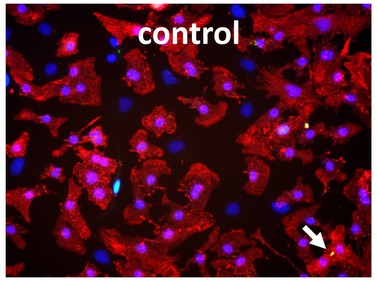
phosphoH3 positivity

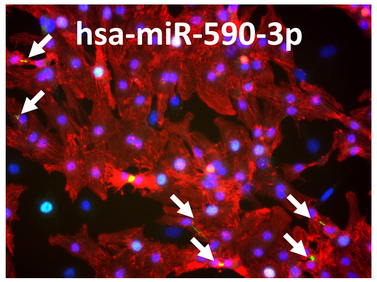




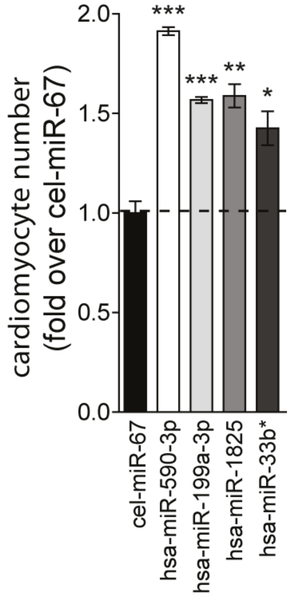
Aurora B midbody localization

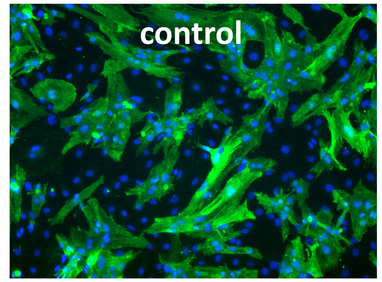


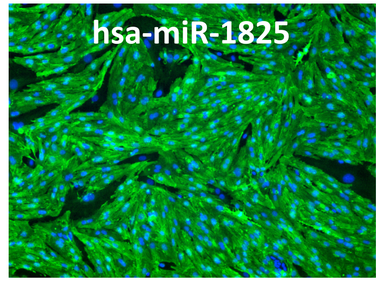




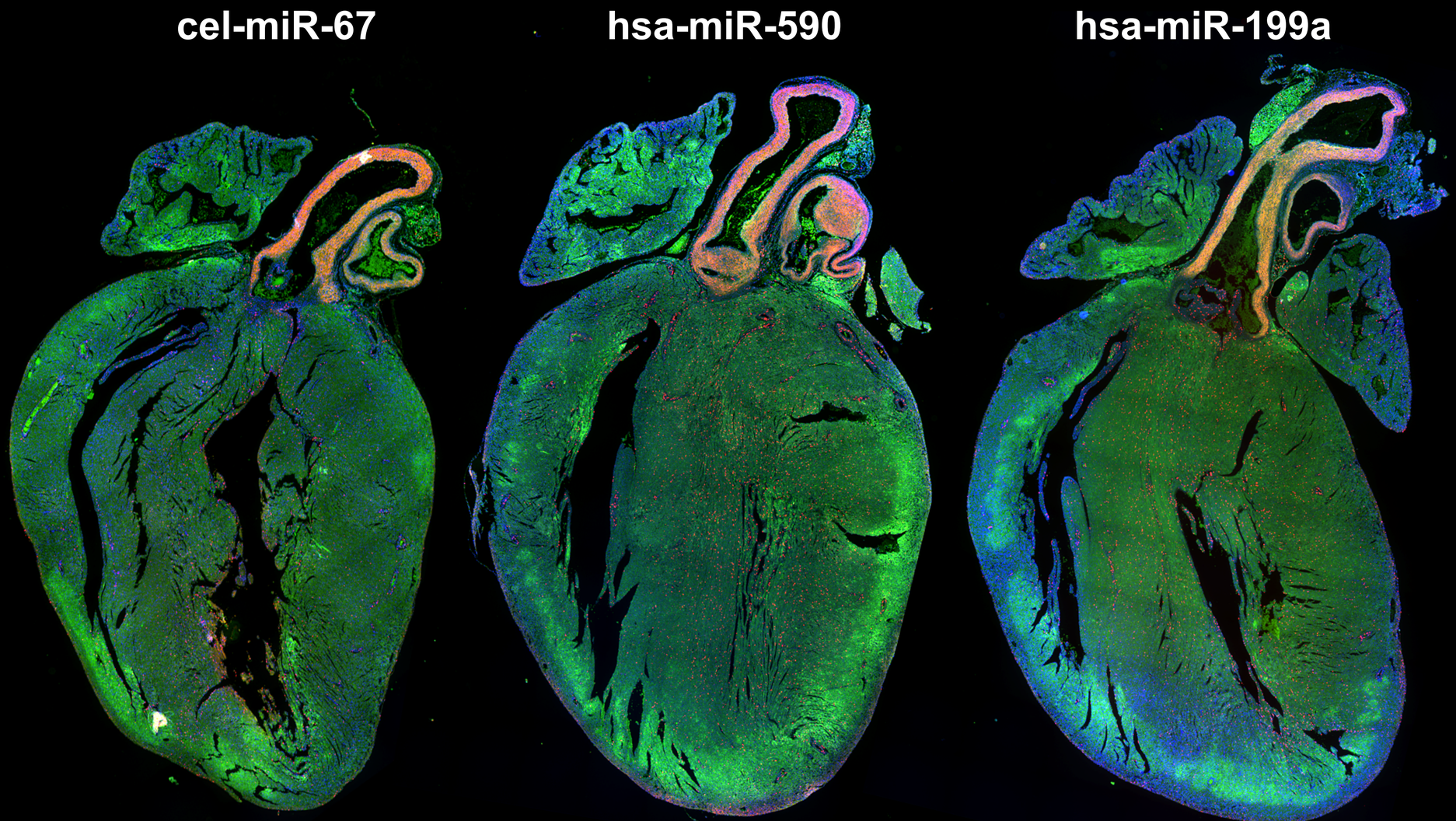
Increase in cell number







I miRNA aumentano la proliferazione dei cardiomiociti in vivo



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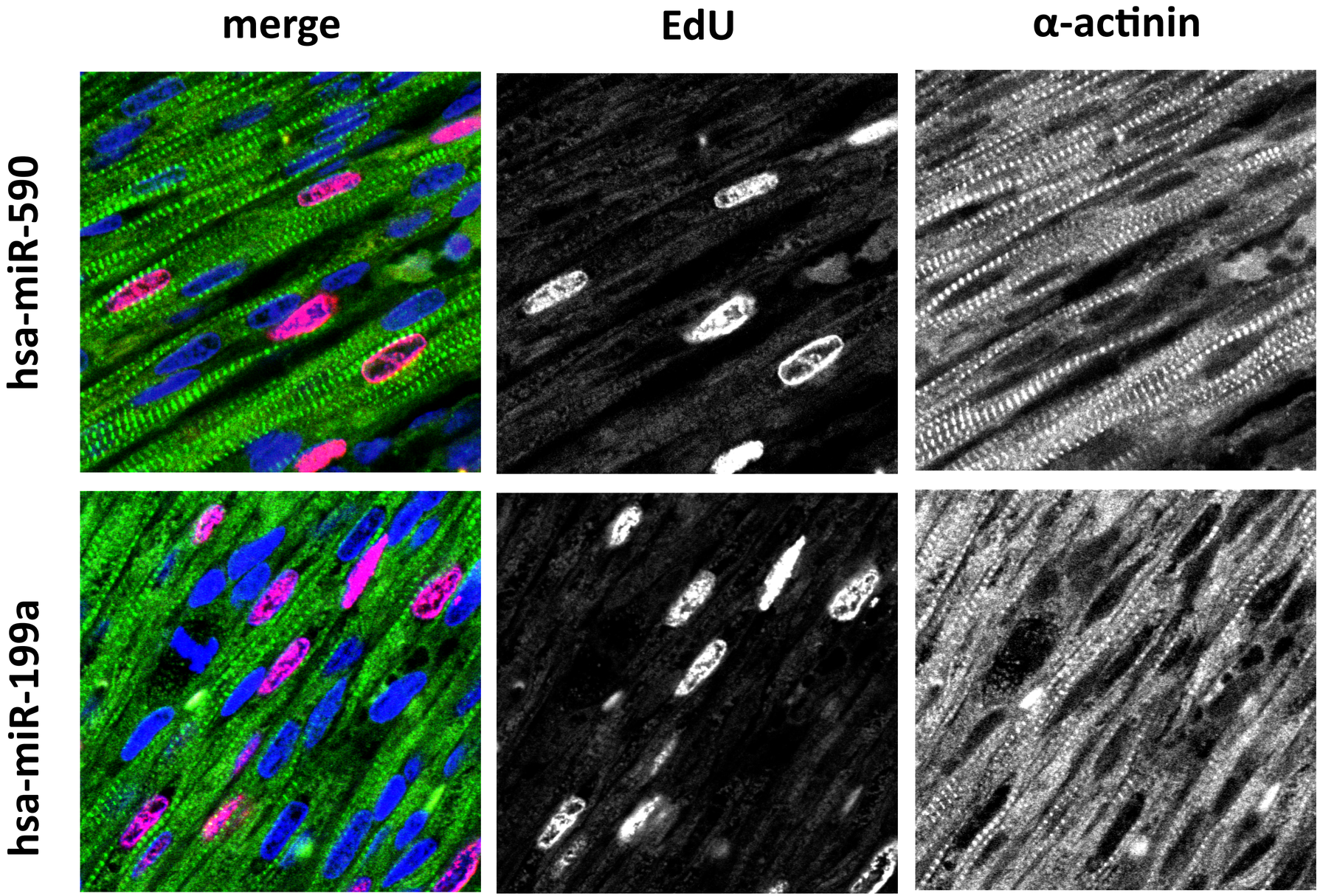
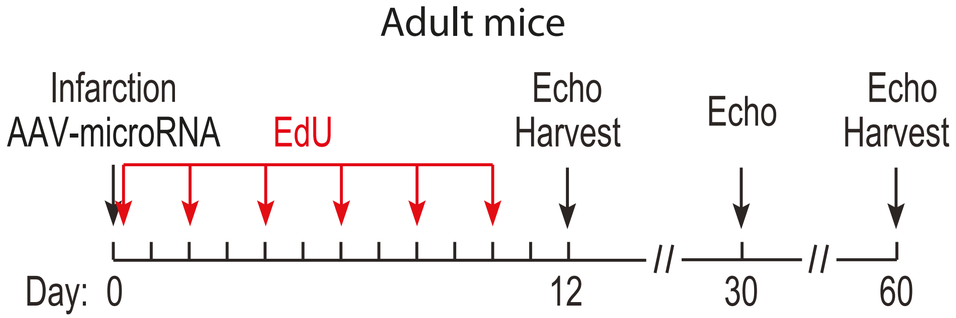
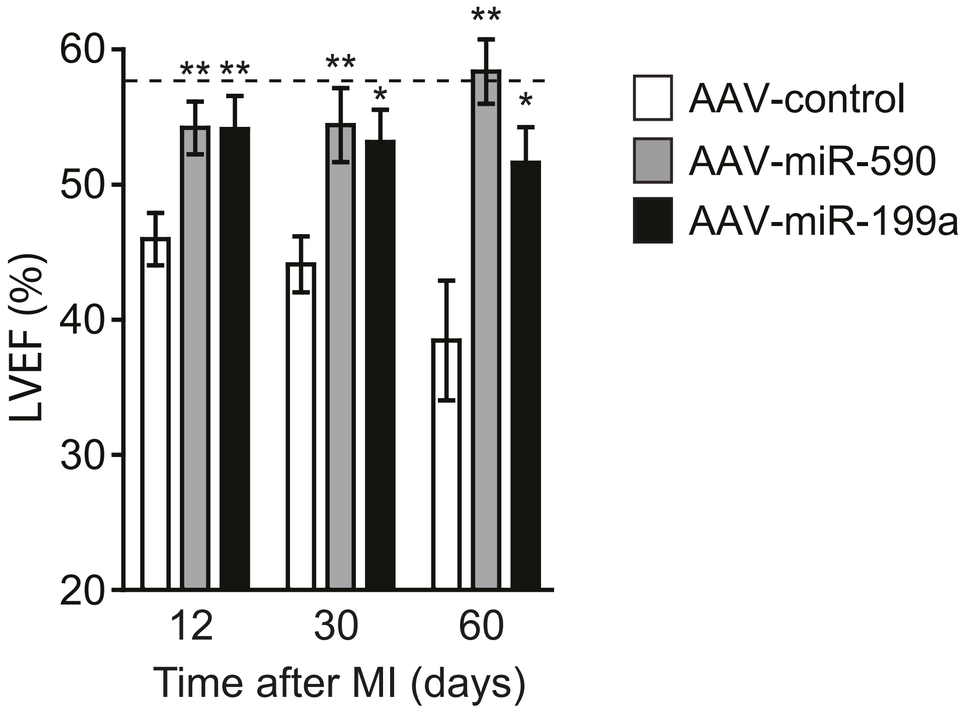
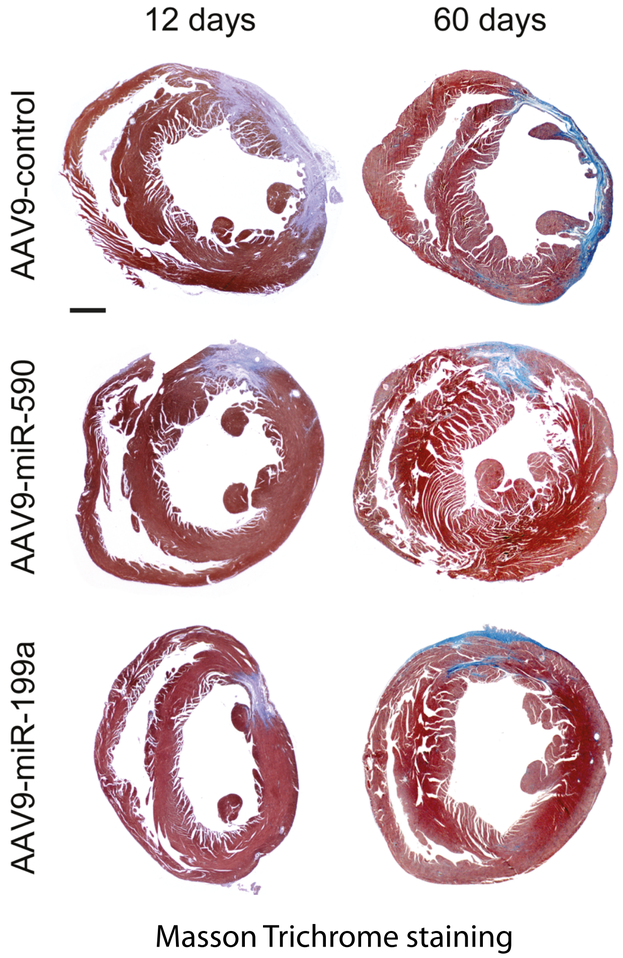


miR-590 e miR-199a preservano la funzione cardiaca e riducono la cicatrice dell'infarto



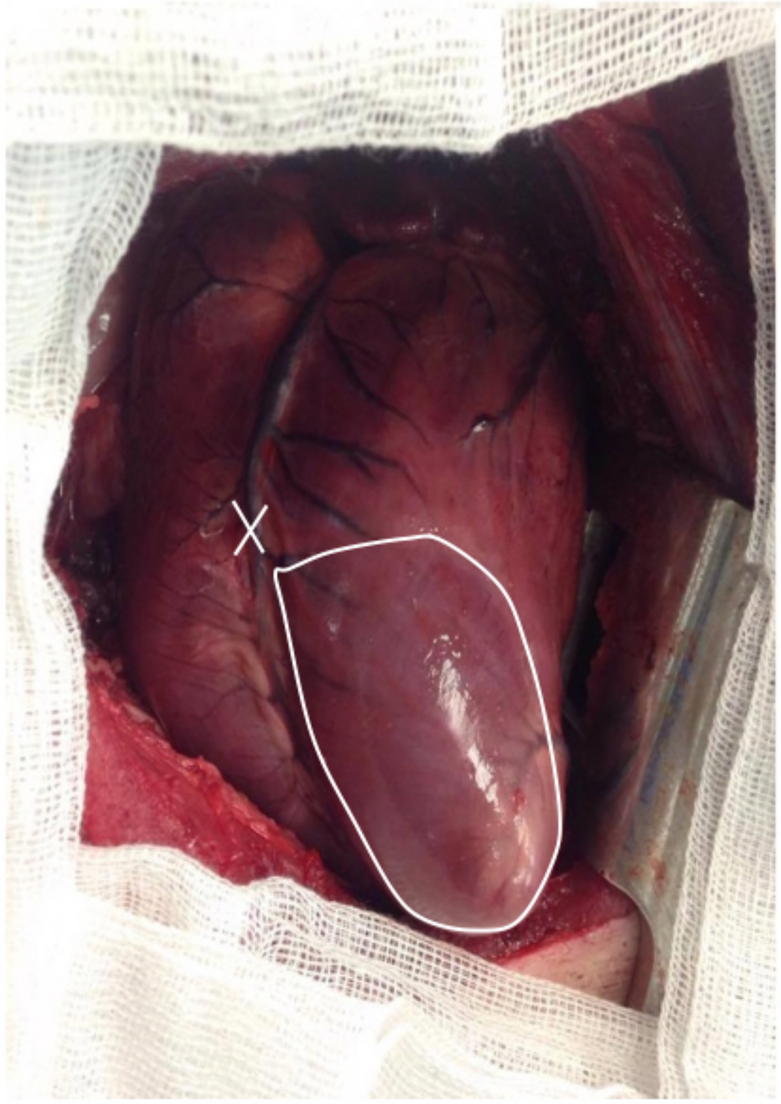




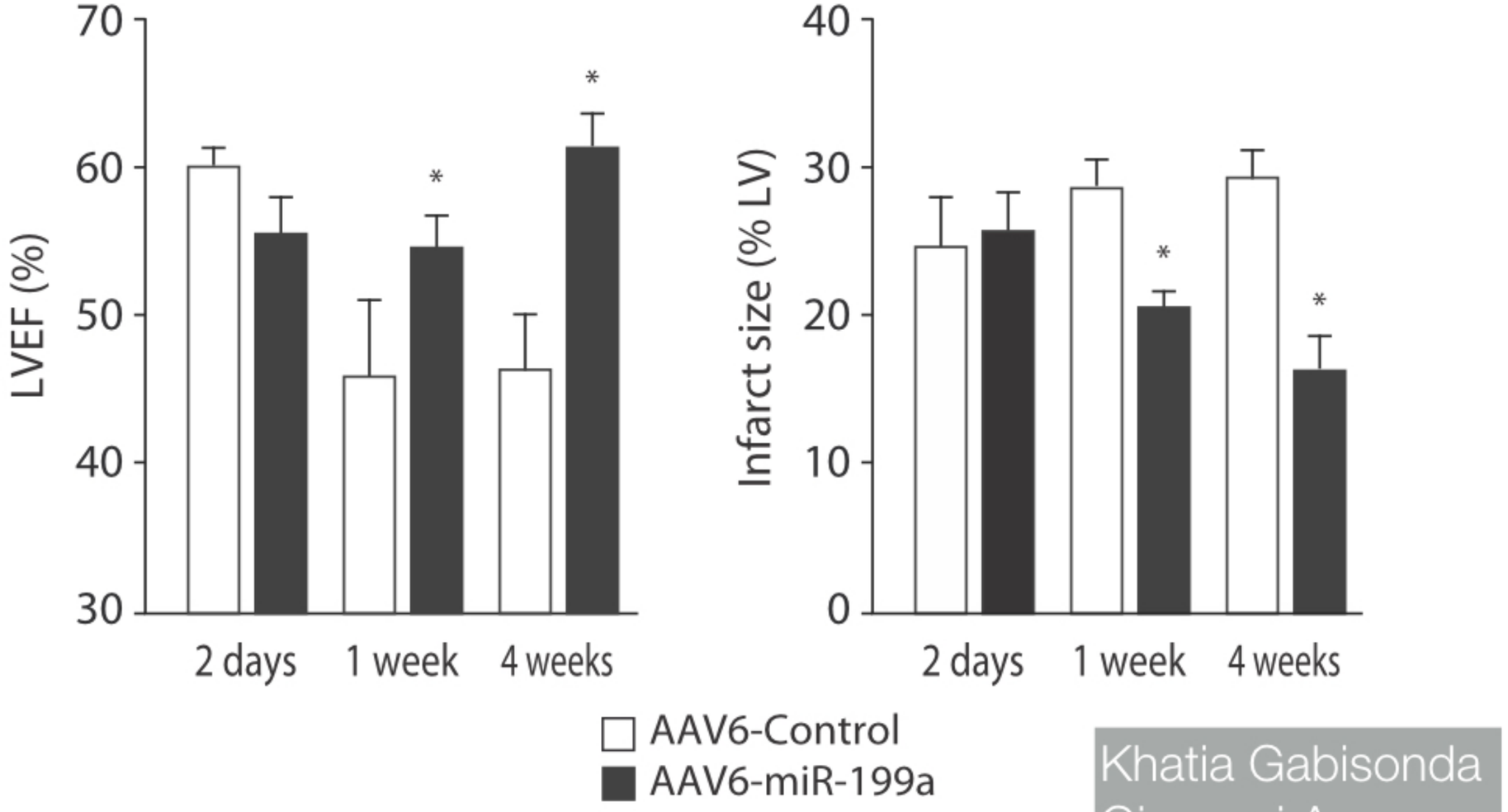
Eulalio et al. 2012. Nature 492, 376

3-6 months old farm pig

LAD Occlusion after first Diagonal branch for 90 minutes, followed by Reperfusion

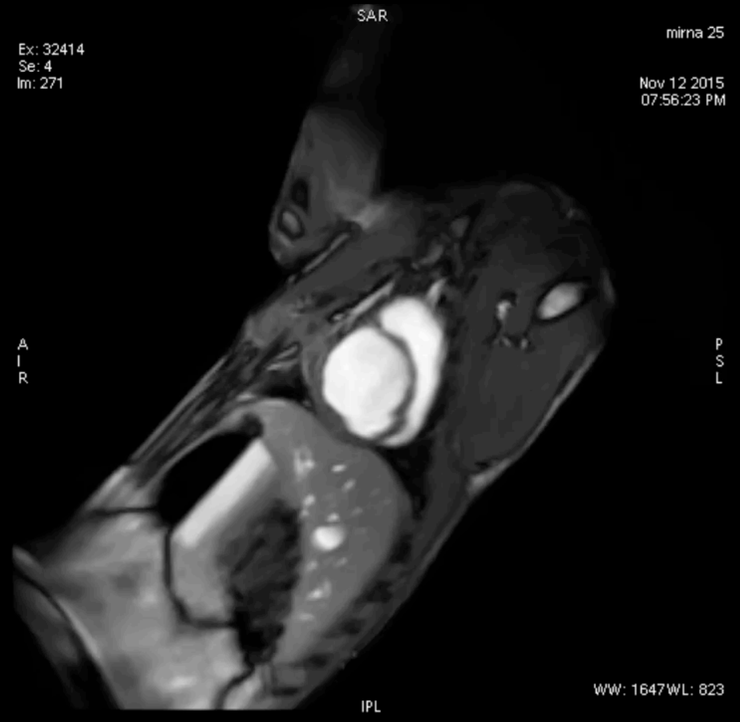


AAV6-miR-199a reduces infarct size and improves cardiac function after MI in pigs

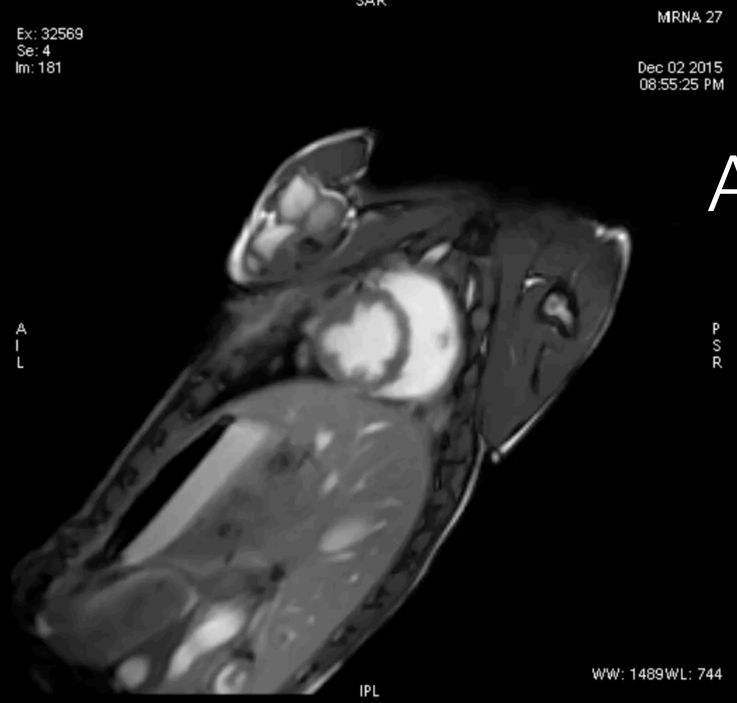


Khatia Gabisonda
Giovanni Aquaro
Fabio Recchia

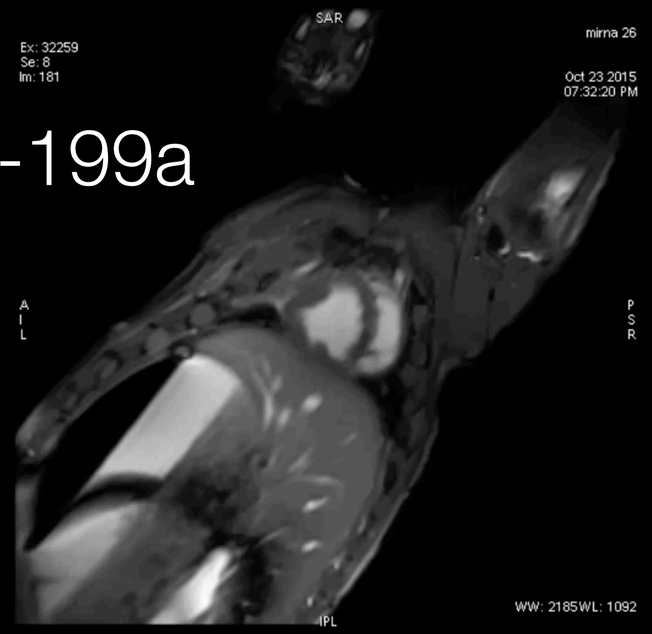
4 weeks after MI



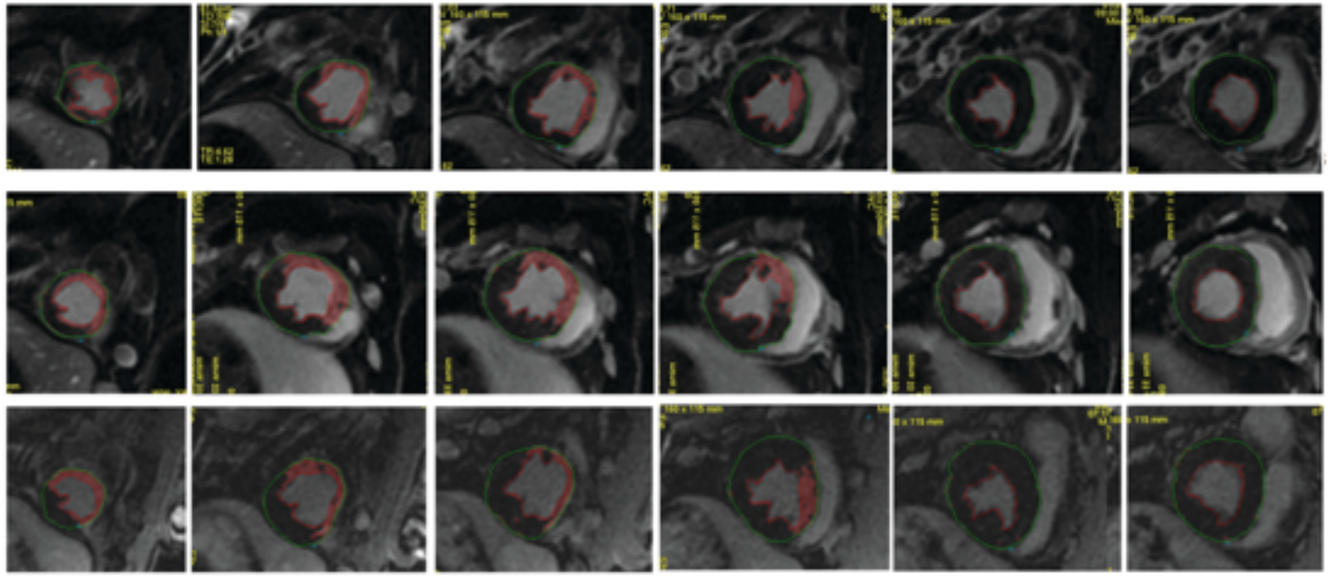
MI
Control



MI
AAV6-miR-199a



AAV6-Control

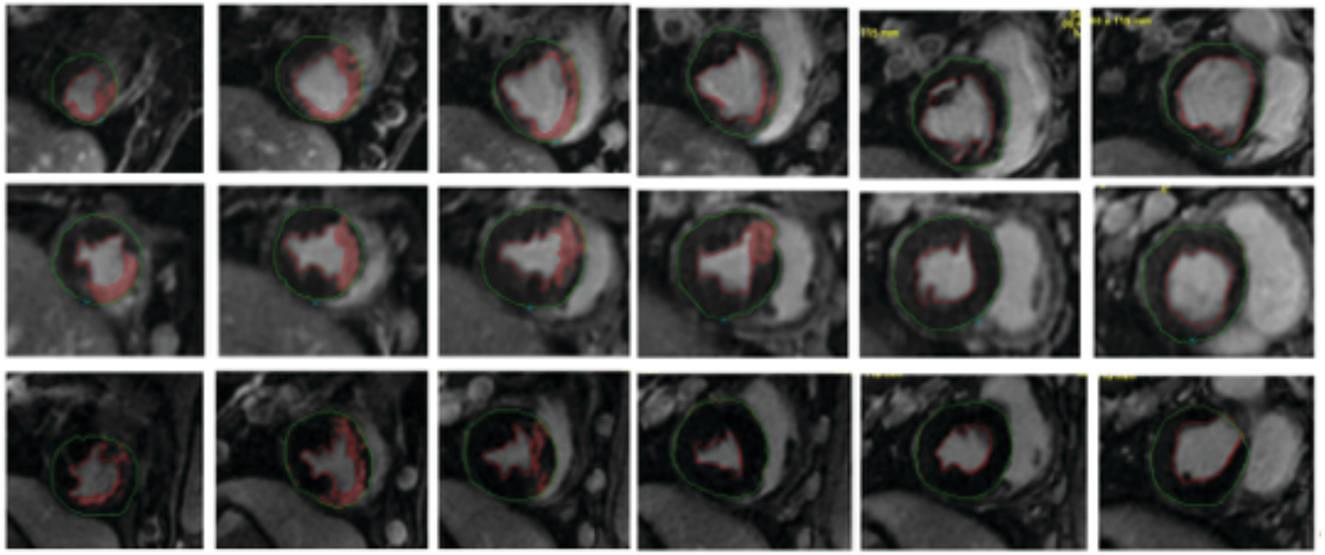


week 1

week 4

week 8

AAV6-miR-199a



week 1

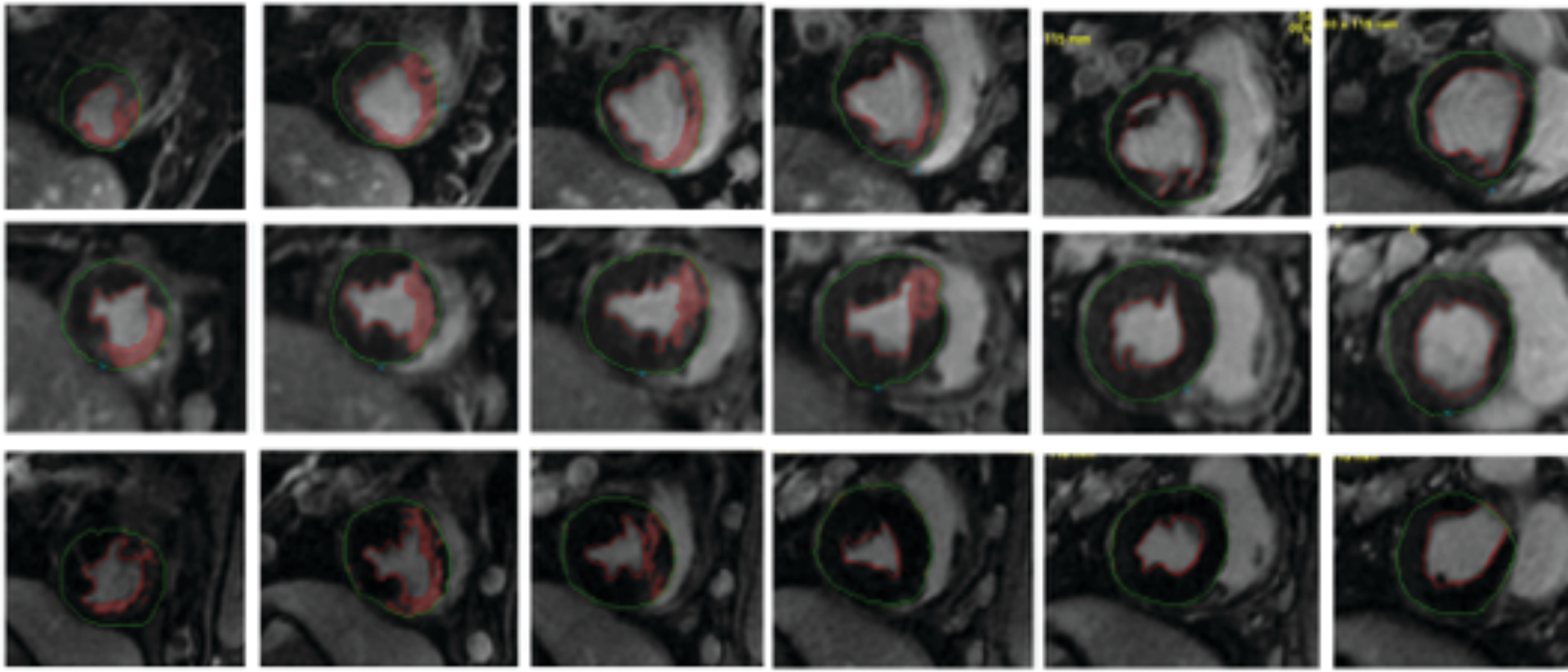
week 4

week 8

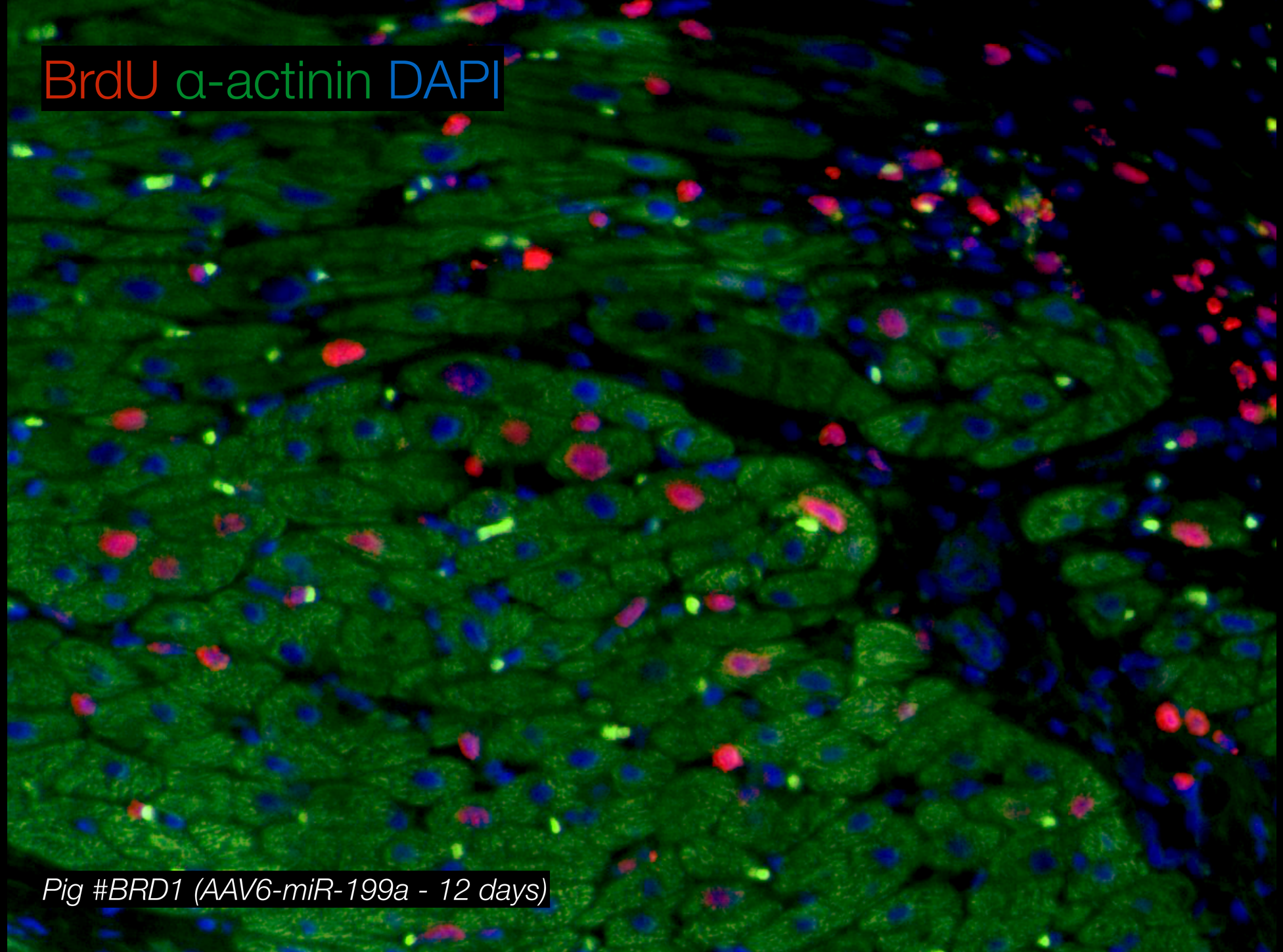
Infarct/fibrosis area

Gabisonia et al. 2019. Nature, 569:418

AAV6-miR-199a



Infarct/fibrosis area


BrdU α-actinin DAPI
Pig #BRD1 (AAV6-miR-199a - 12 days)