Biology of perivascular progenitor cells

Paolo Madeddu, MD
Chair Experimental Cardiovascular Medicine, Bristol Heart Institute, University of Bristol

CBCS Summer School on Cardiovascular Sciences

"Basic Mechanisms translated to the Clinic"

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Structure of presentation

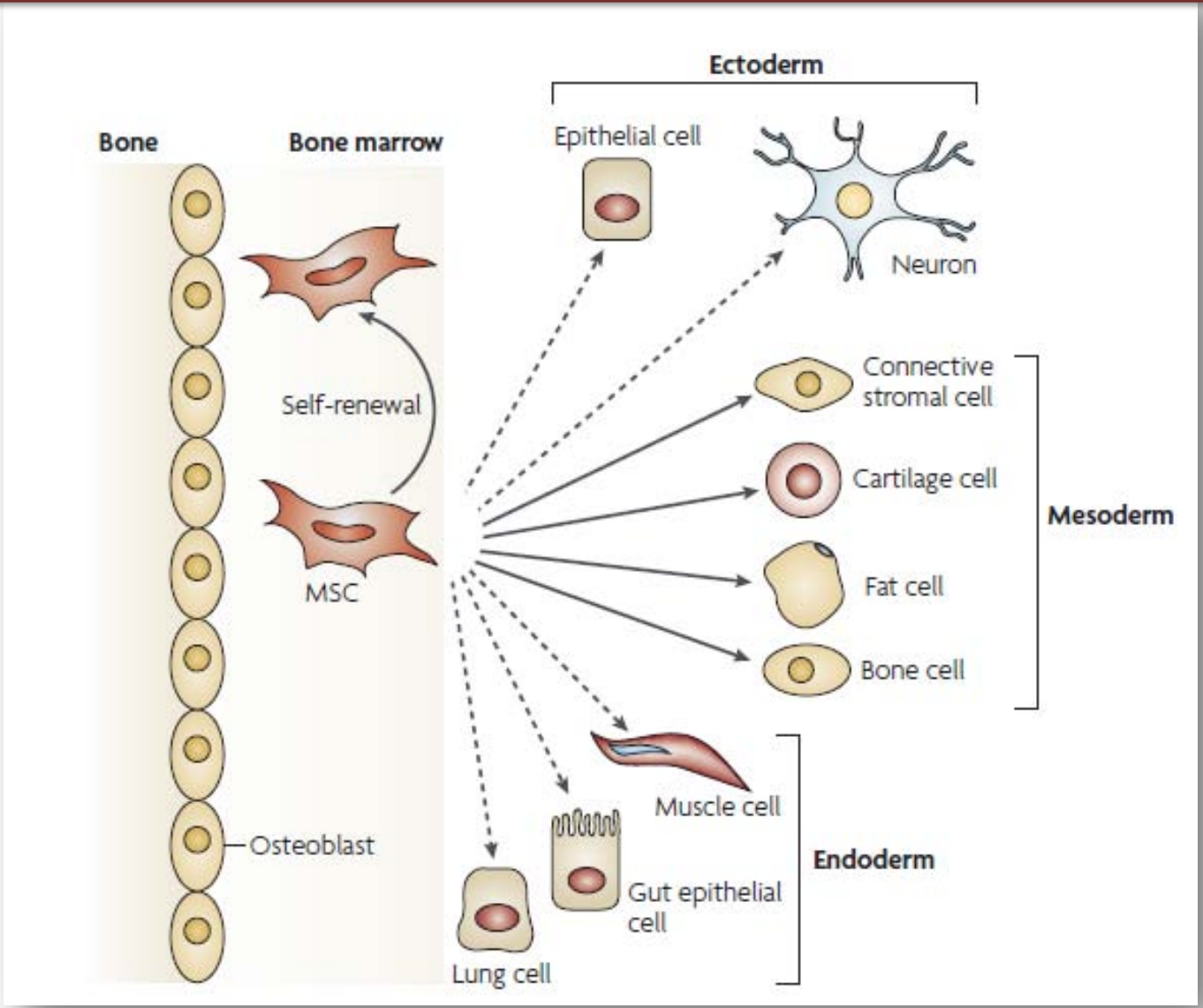
- Introducing mesenchymal stem cells (MSCs).
- Revisiting the identity/equivalence of perivascular MSCs, adventitial progenitor cells and pericytes.
- Illustrating the biology and therapeutic prospect of pericytes and adventitial progenitor cells.

The family of Mesenchymal Stem Cells (MSCs)

(ii) possessing self-renewal capacity and ability to differentiate *in vitro* into chondrogenic, osteogenic, adipogenic and myogenic lineages,

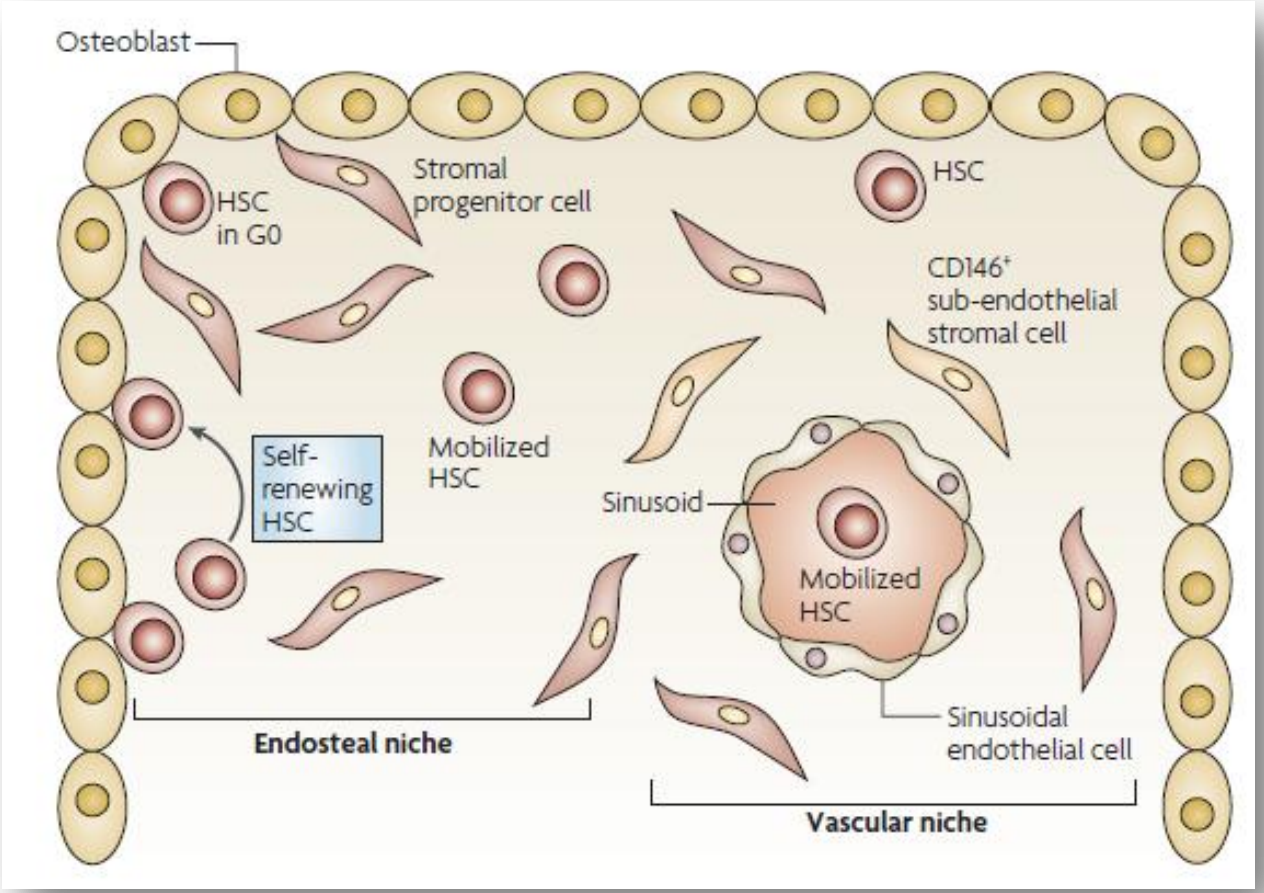
(iii) expressing CD73, CD90 and CD105 and being negative for CD34, CD11, CD19, CD45, CD79a, CD14, histocompatibility locus antigen HLA-DR.

The term MSC was coined by Caplan after Friedenstein's discovery of multipotent stromal cells endowed of self renewal and plasticity



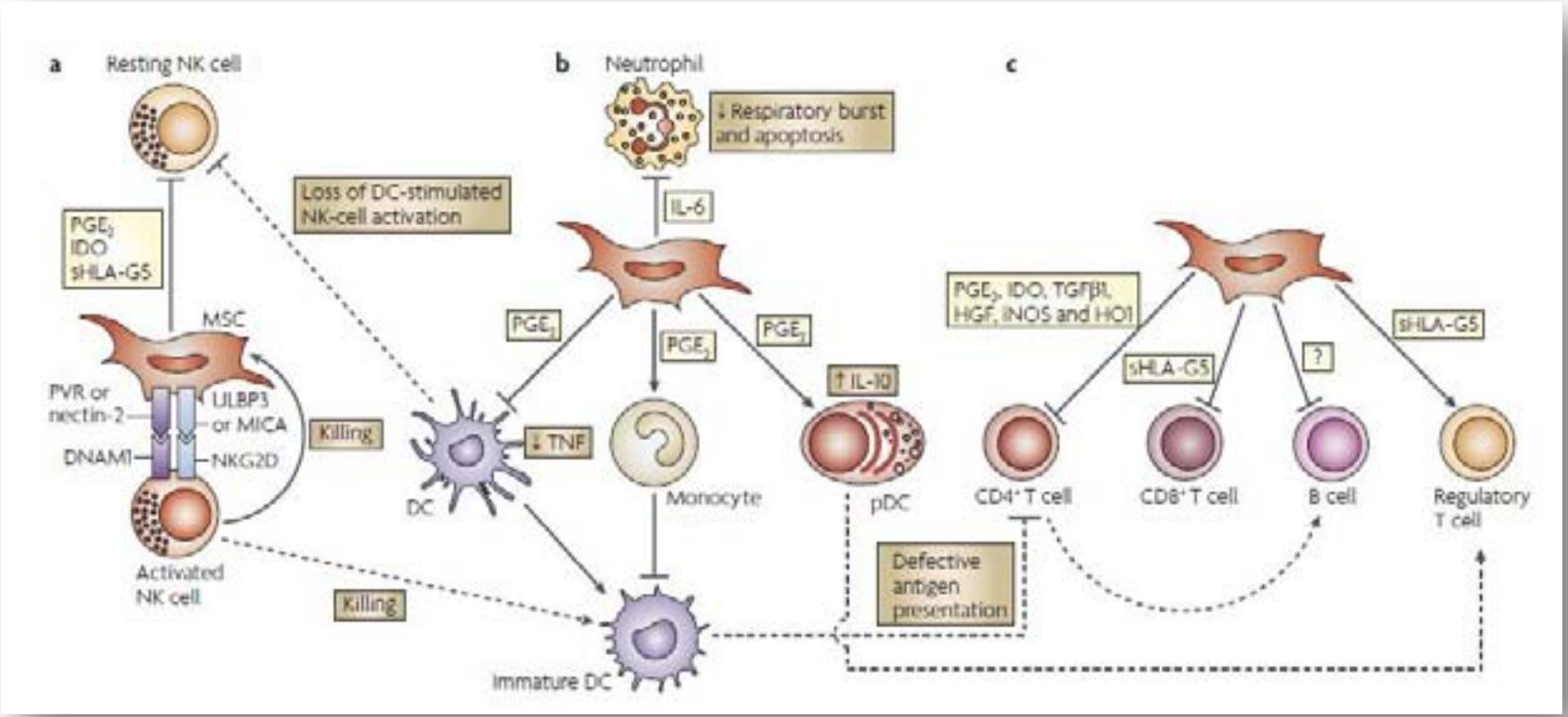
Uccelli et al Nature Rev Immunology, 2008

MSCs are part of the stromal cell pool that support the endosteal and vascular niches in bone marrow



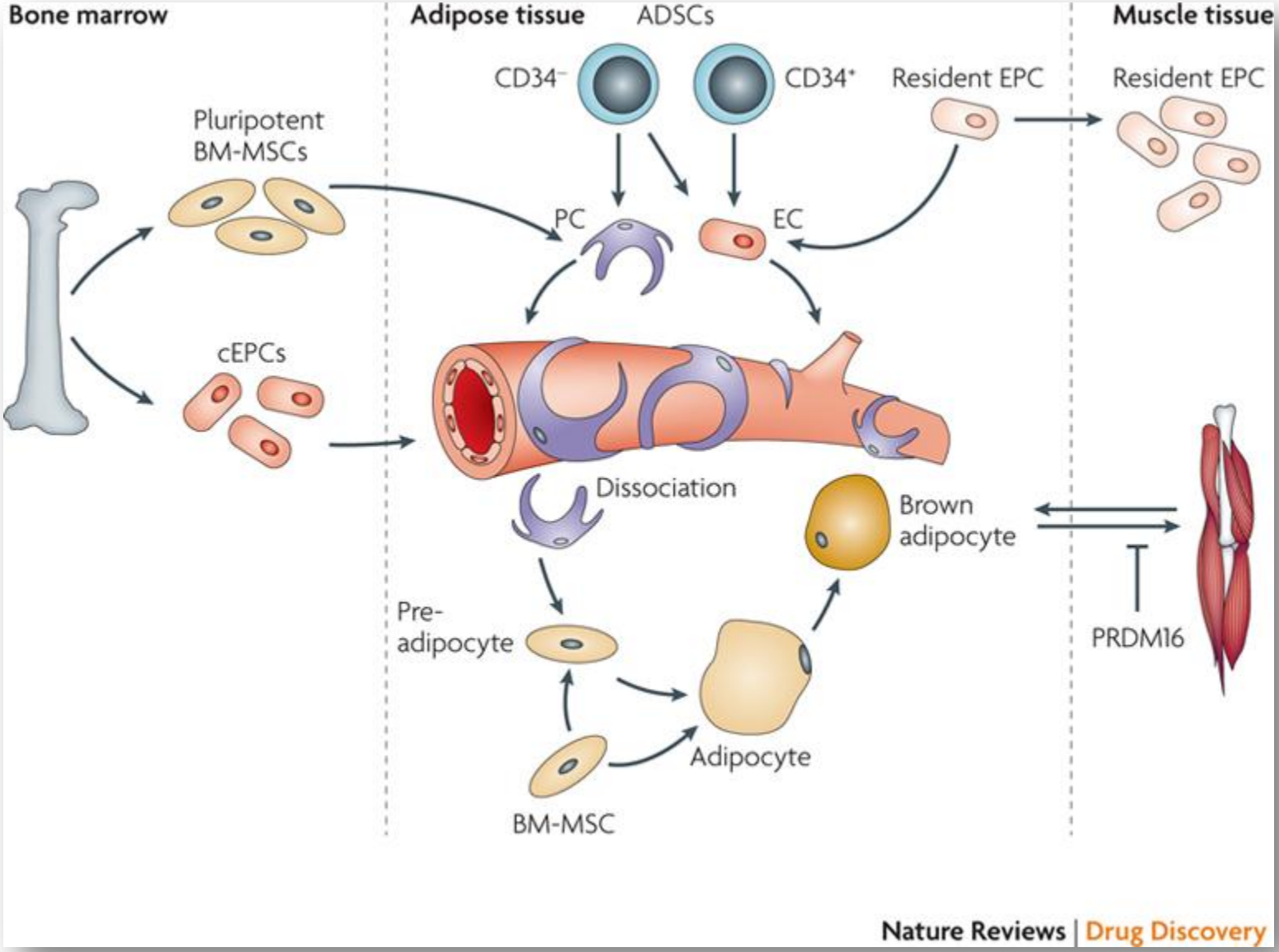
Uccelli et al Nature Rev Immunology, 2008

Interaction between MSCs and cells of acquired and innate immunity

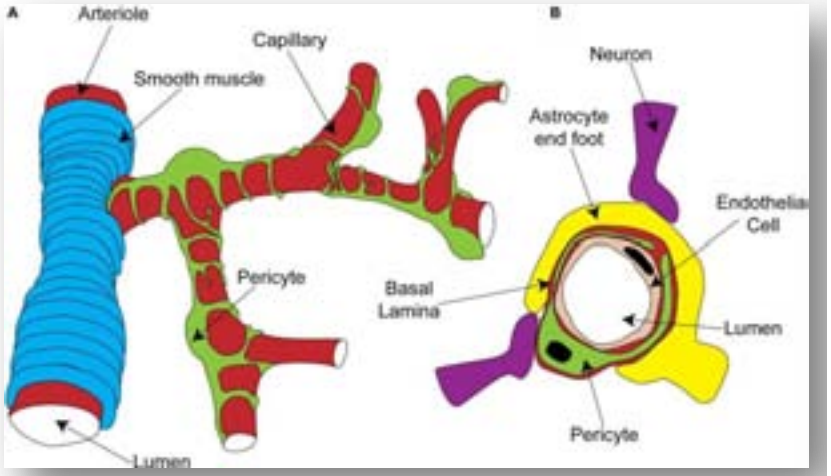


Uccelli et al Nature Rev Immunology, 2008

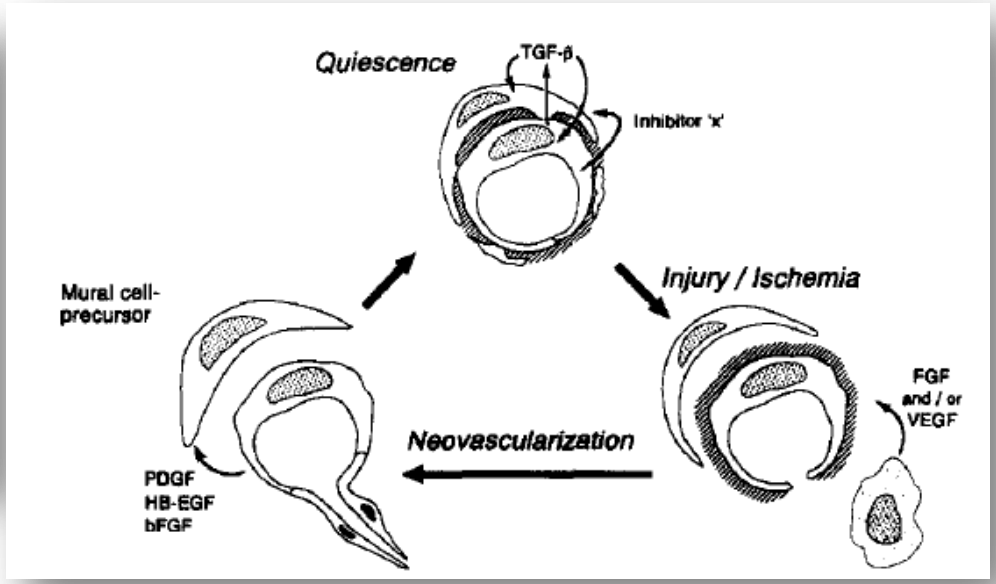
MSCs are abundantly present in adult tissues: the adipose tissue paradigma



Pericytes: stabilizers of the vasculature

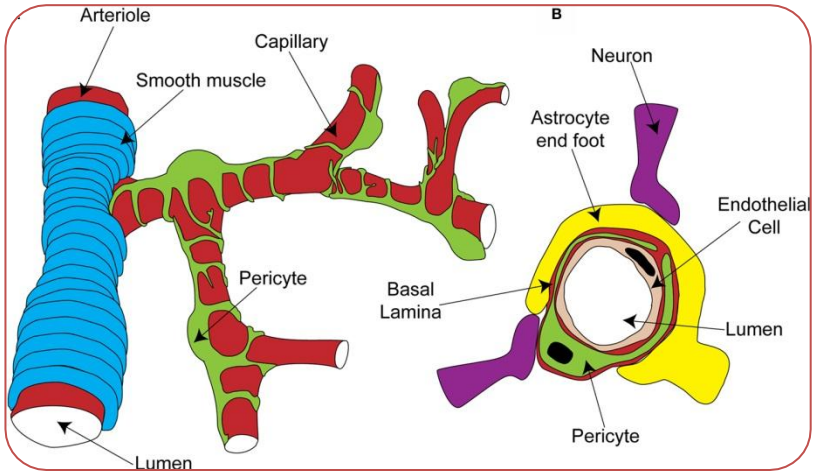


Hamilton et al Frontiers in Neuroenergetics 2010



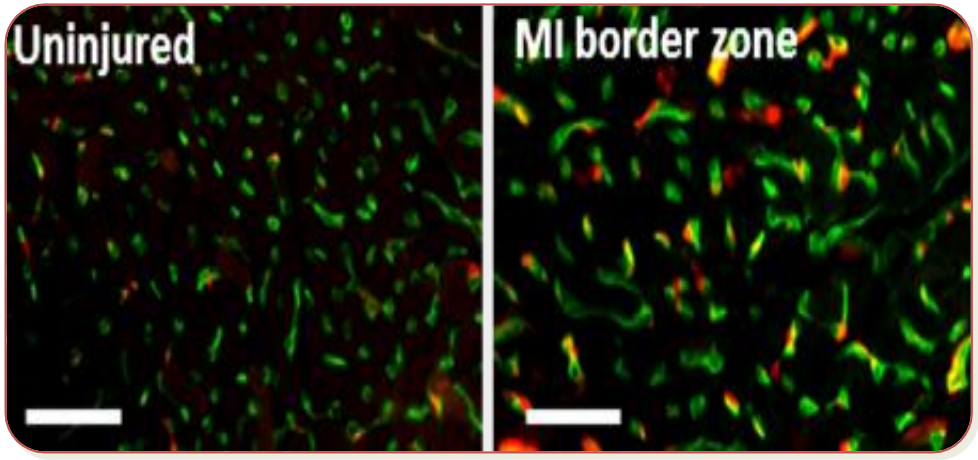
K.K. Hirschi, P.A. D'Amore / Cardiovascular Research 32 (1996) 687–698

Pericytes: stabilizers of the vasculature

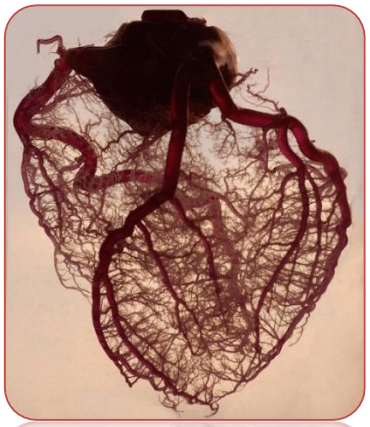


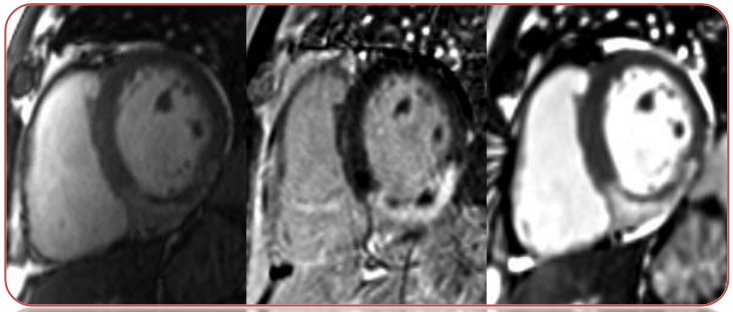
Hamilton et al Frontiers in Neuroenergetics 2010

NG2+ pericytes in mouse heart



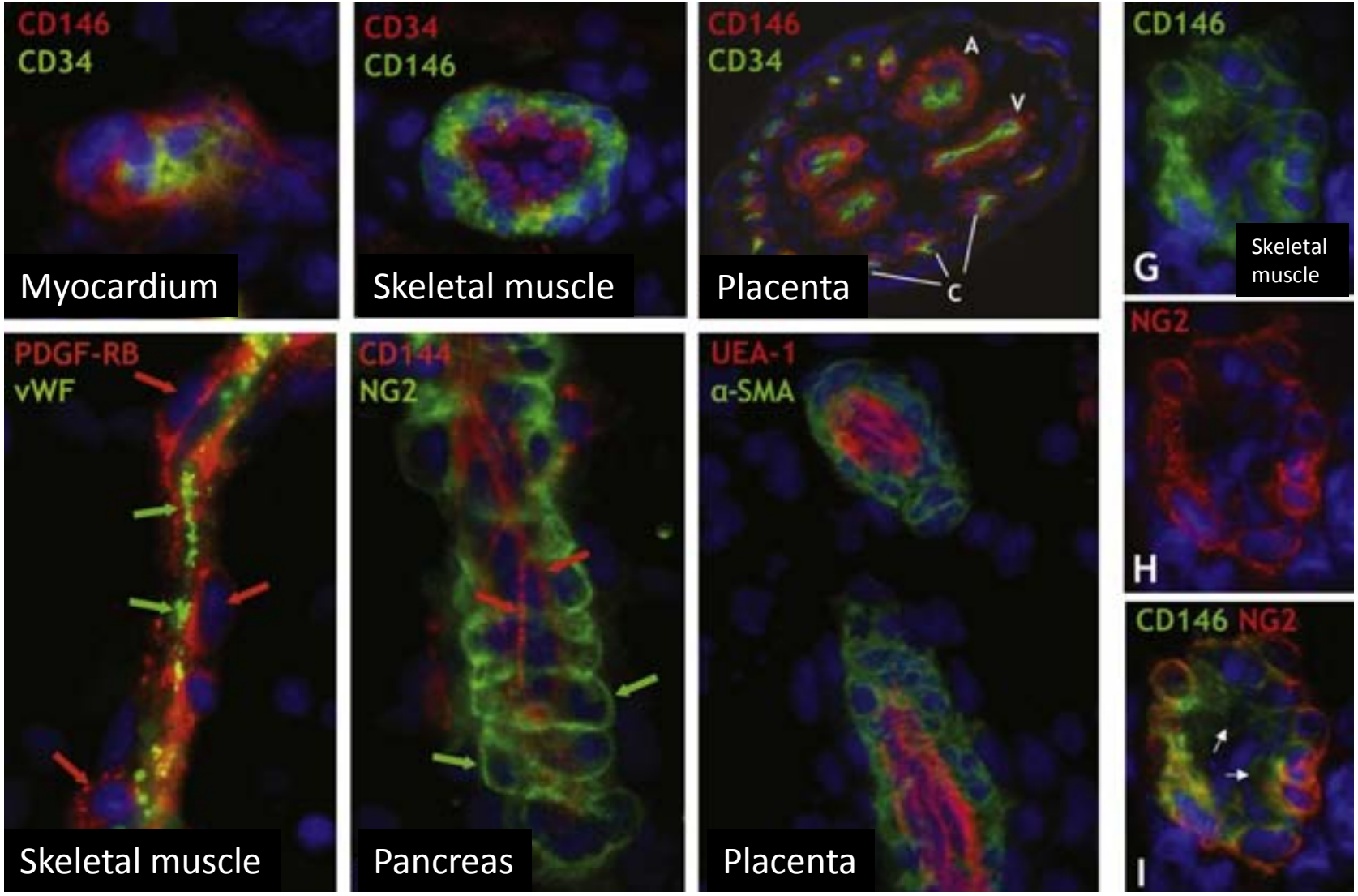
Mitchel and Madeddu, unpublished





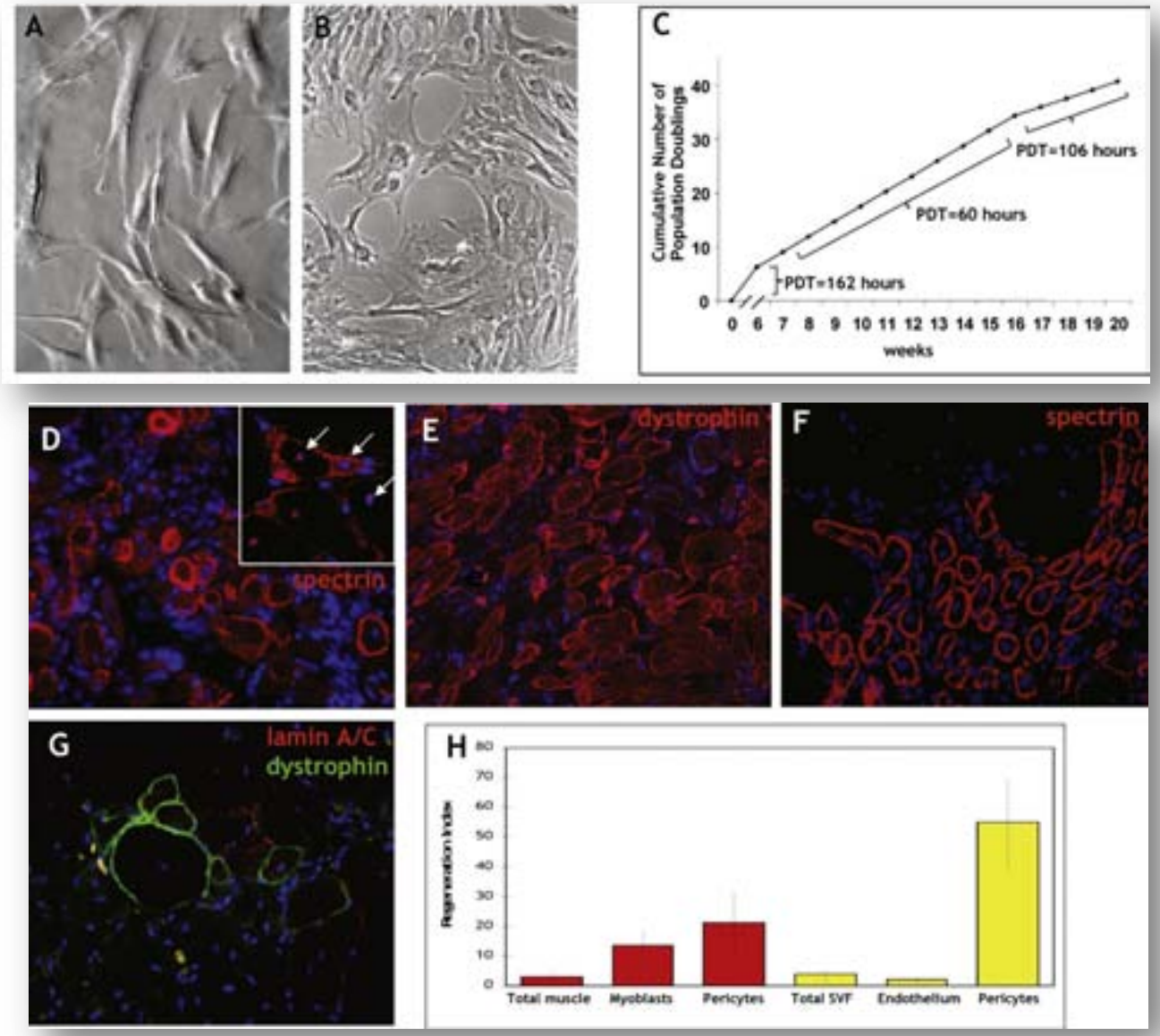
Berry et al Circ Cardiovasc Imaging 2010

Perivascular CD146+ pericytes are present in different organs

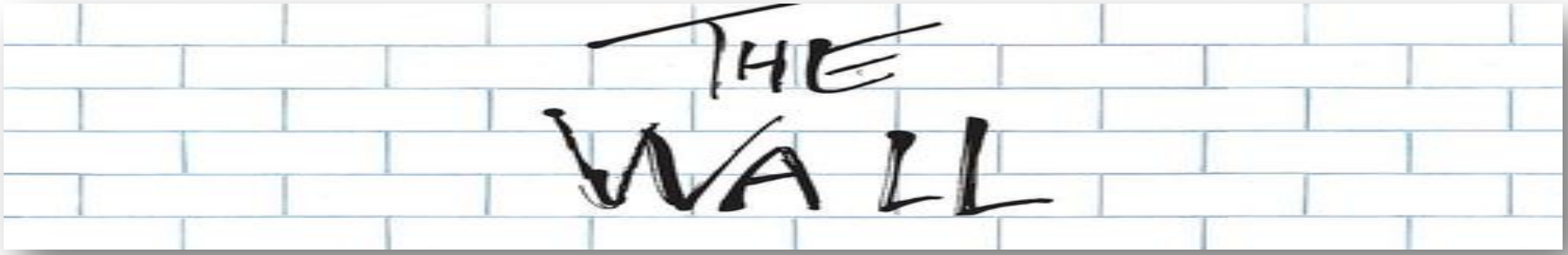


Crisan et al. Cell Stem Cell 2008

CD146+ Pericytes expanded in culture regenerate skeletal muscle in dystrophin-deficient mice



Crisan et al. Cell Stem Cell 2008

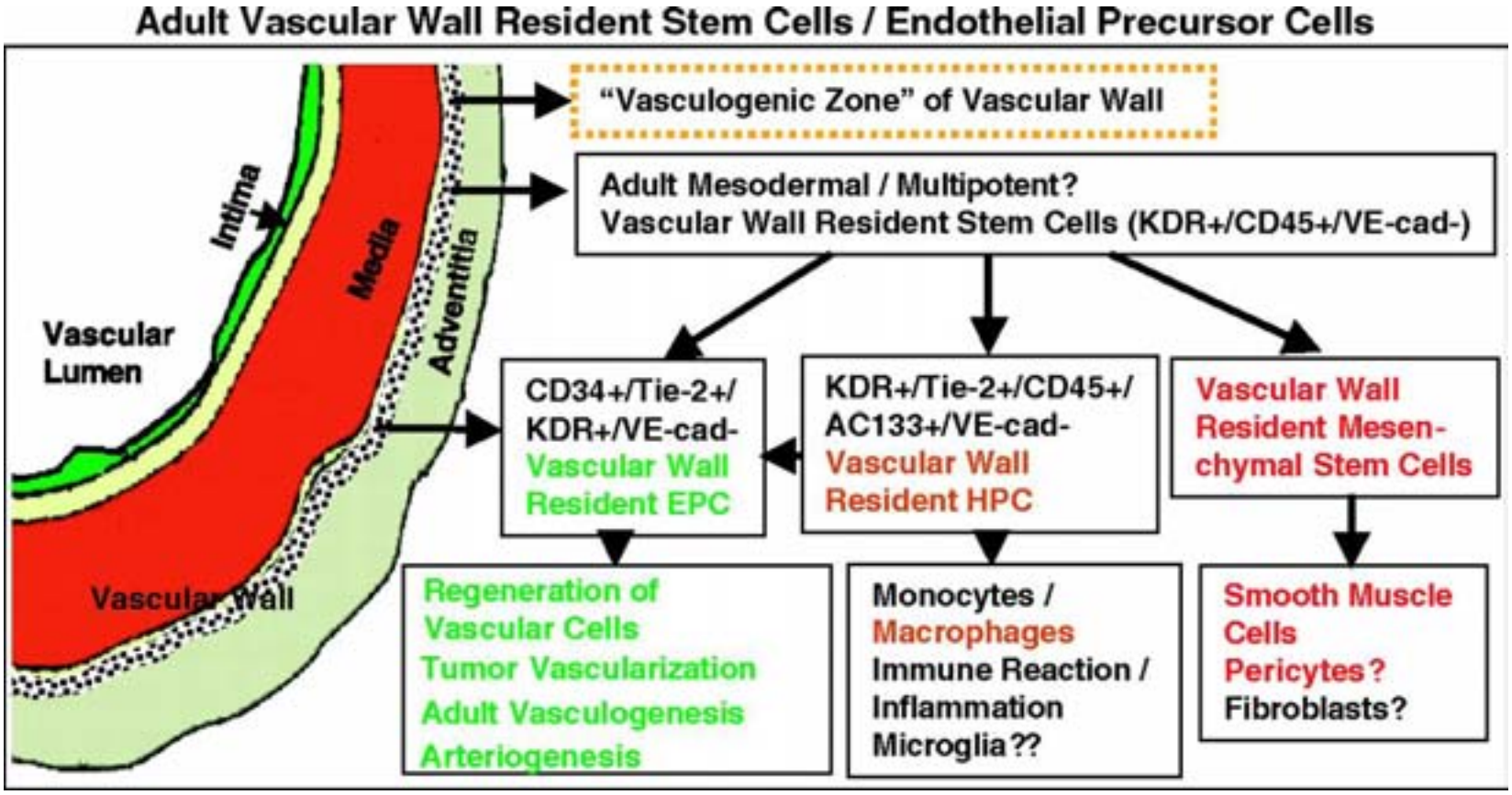


- **The presence of early progenitor cells, named mesoangioblasts because of their ability to differentiate into endothelial cells and other mesodermal lineages, has been already demonstrated in the embryonic dorsal aorta.**
- **Moreover, hematopoietic stem cells are generated from *hemogenic endothelium* in the embryonic aortic wall.**

Minasi MG, Riminucci M, De Angelis L, *et al*. The meso-angioblast: a multipotent, self-renewing cell that originates from the dorsal aorta and differentiates into most mesodermal tissues.

***Development*. 2002; 129: 2773–83.**

Hypothetical scheme of the `vasculogenic zone'



Zengin E et al. Development 2006;133:1543-1551

Table 1 Comparison between human pericytes and adventitial perivascular cells [21, 22]

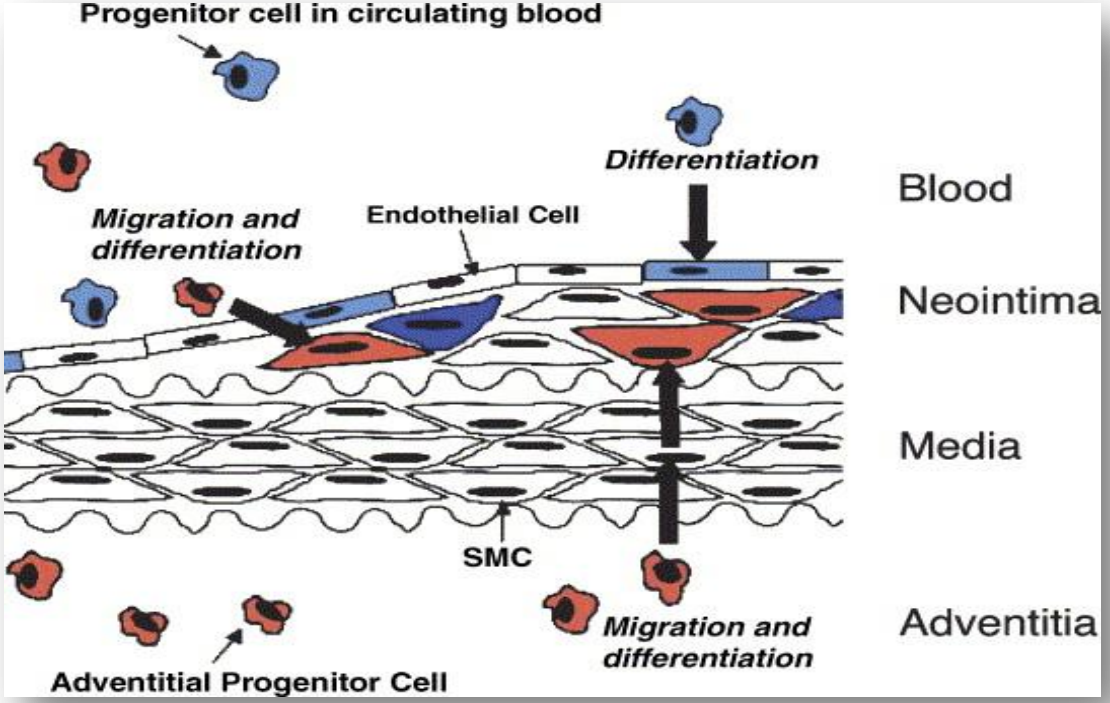
	Pericytes	Adventitial cells
Perivascular location	Capillaries and microvessels	Large vessels
Human tissue origin	Adult, foetal and embryonic skeletal muscle and pancreas, adult WAT, foetal skin, small intestine, brain, foetal and embryonic BM, term and mid-term placenta	Adult WAT, foetal skeletal muscle, lung and BM
FACS selection	CD146+CD34-CD56-CD45-	CD34+CD31-CD146-CD45-
Markers *in vitro*	CD146, NG2, PDGFRβ, αSMA, CD90, CD73, CD105, CD44, ALP, nestin, vimentin	CD34, CD90, CD73, CD105, CD44, vimentin
Markers *in vivo*	CD146, NG2, PDGFRβ, α SMA, CD90, CD73, CD105, CD44, ALP	CD34, CD90, CD73, CD105, CD44
Documented differentiation potential	Osteogenic, adipogenic, chondrogenic, myogenic	Osteogenic, adipogenic, chondrogenic, pericytic

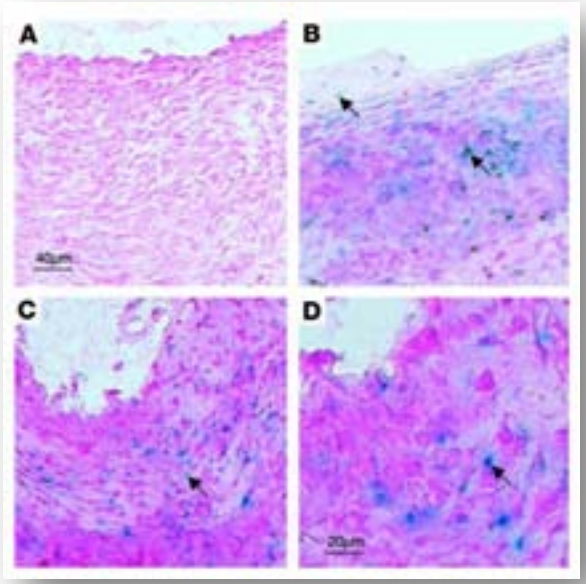
Crisan M, Corselli M, Chen WC, Péault B.
J Cell Mol Med. 2012

	Cell lIne identificative markers	References
Mesangioblasts	Mesenchymal:CD13$^+$, CD73$^+$, CD44$^+$, CD49b$^+$ Pericyte:NG2$^+$, CD105$^+$ Endothelial:Tie2$^+$, KDR$^-$, CD31$^-$, CD34$^-$, HMT: CD133$^-$, CD45$^-$	Morosetti, 2011, Acta Myol.
Cordblood MSC	Mesenchymal:CD71$^+$,CD73$^+$, CD80$^+$, CD105$^+$ Pericyte:NG2$^+$, ALP$^+$ Endothelial:CD146$^+$, CD34$^-$, HMT: CD45$^-$	Bosch, 2012, Stem Cell Dev
Humbelical cord perivascular cells	Mesenchymal: CD56+, CD71$^+$, CD73$^+$, CD90$^+$, CD105$^+$, CD44$^+$, desmin$^+$, Pericyte:RGS5$^+$ Endothelial: CD34$^-$	Bosch, 2012, Stem Cell Dev
Human foetal aorta VPCs	Mesenchymal: desmin$^+$ Pericyte:NG2$^+$ Endothelial:Tie2$^+$, KDR$^+$, CD31$^+$, CD34$^+$, HMT: CD133$^+$, CD45$^-$	Invernici, 2007, Am . J. Pathol. Invernici, 2008, Cytotechnology
Muscular and tissue pericytes	Mesenchymal:CD13$^+$, CD73$^+$, CD44$^+$, CD90$^+$, aSMA$^+$ ALP Pericyte:NG2$^+$, CD105$^+$, ALP$^+$, RGS5$^+$, PDGFR$^+$ Endothelial:CD146$^+$, CD31$^-$, CD34$^-$, HMT: CD45$^-$	Dellavalle, 2007, Nat. Cell Biol. Corselli, 2012, Stem Cells Dev Crisan, 2008, Cell Stem Cell Psaltis, 2011,J. Cardiovasc. Transl. Res.
Vascular wall resident multipotent SCs	Mesenchymal: CD73$^+$, CD44$^+$, CD90$^+$, desmin$^+$, aSMA$^-$, CD29$^+$ Pericyte:NG2$^{+/-}$, CD105$^+$, PDGFR$^+$ Endothelial:Tie2$^+$, KDR$^-$, CD31$^-$, CD34$^-$, CD146$^-$ HMT: CD133$^-$, CD45$^-$, CD68$^-$, CD19$^+$, CD29$^+$	Ergun, 2010, Antioxid Redox Signal Klein, 2011, Plos one
SVPs	Mesenchymal:CD90$^+$, CD44$^+$, desmin$^+$, CD13$^+$, CD73$^+$, CD29$^+$, CD49$^+$, aSMA$^-$, CD59$^+$ Pericyte:NG2$^+$, CD105$^+$, PDGFR$^+$, Endothelial:Tie2$^+$, CD146$^-$, CD31$^-$, CD34$^{-/+}$, HMT: CD133$^-$, CD45$^-$	Campagnolo, 2010, Circulation

Vono et al 2012

Adventitial Progenitor Cells Contribute to Arteriosclerosis

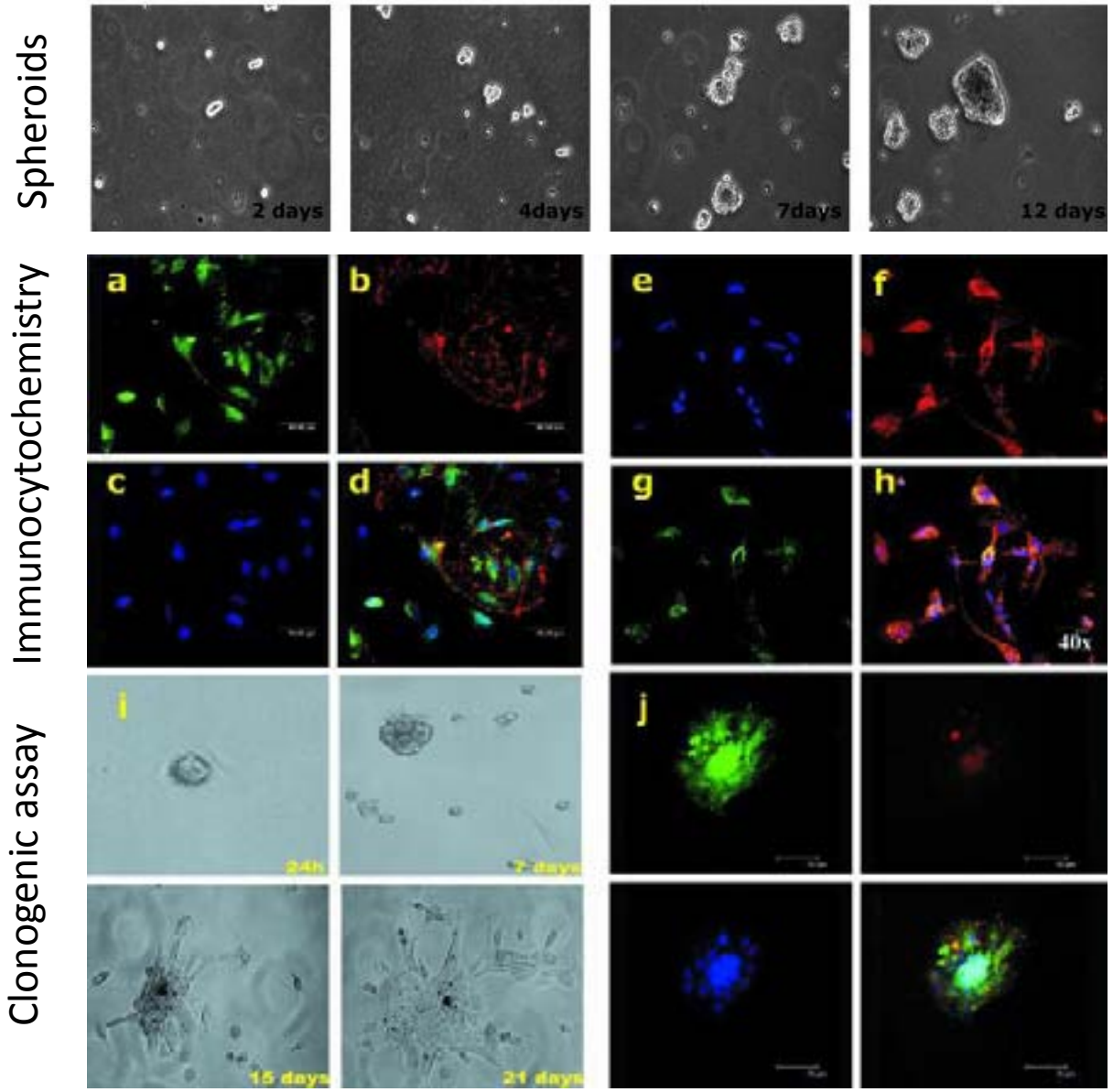




Hu et al. J Clin Invest. 2004

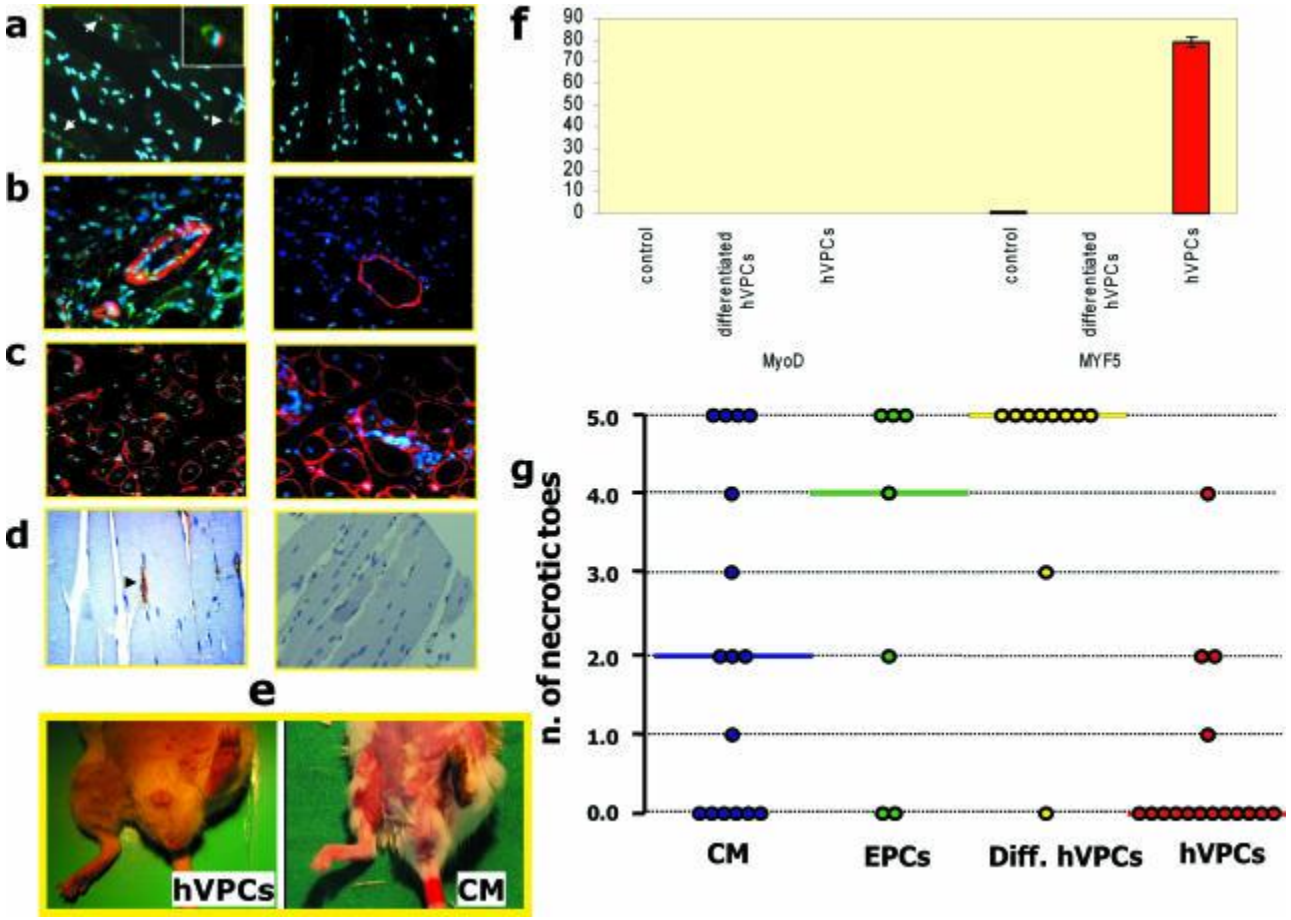
Evelyn Torsney , Yanhua Hu , Qingbo Xu
Trends in Cardiovascular Medicine Volume 15, Issue 2 2005 64 - 68

Human fetal aorta contains vascular progenitor cells capable of inducing vasculogenesis, angiogenesis, and myogenesis



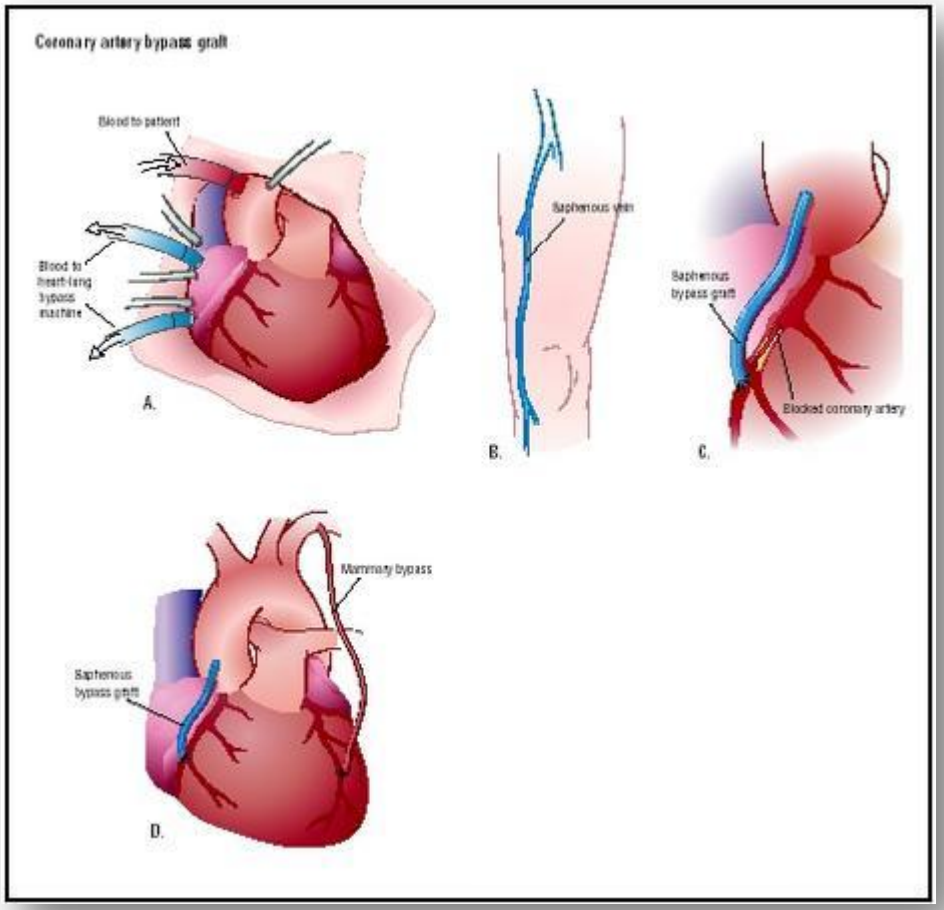
Invernici et al. Am J Pathol 2007

Human fetal aorta vascular progenitor cells transplantation in a murine model of peripheral ischemia

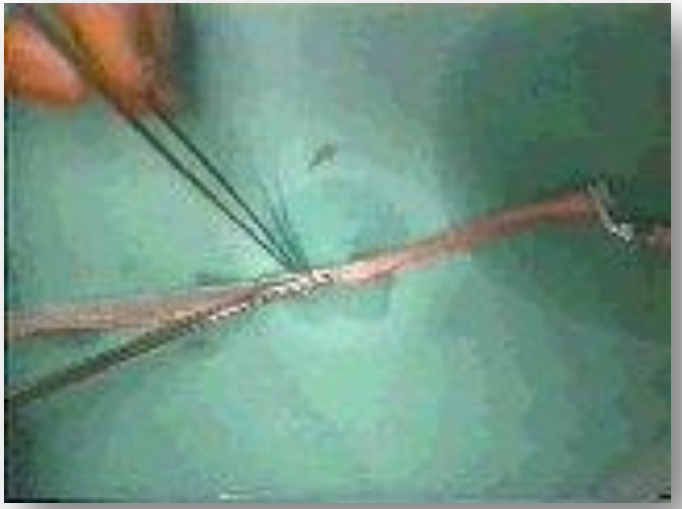


Invernici et al. Am J Pathol 2007

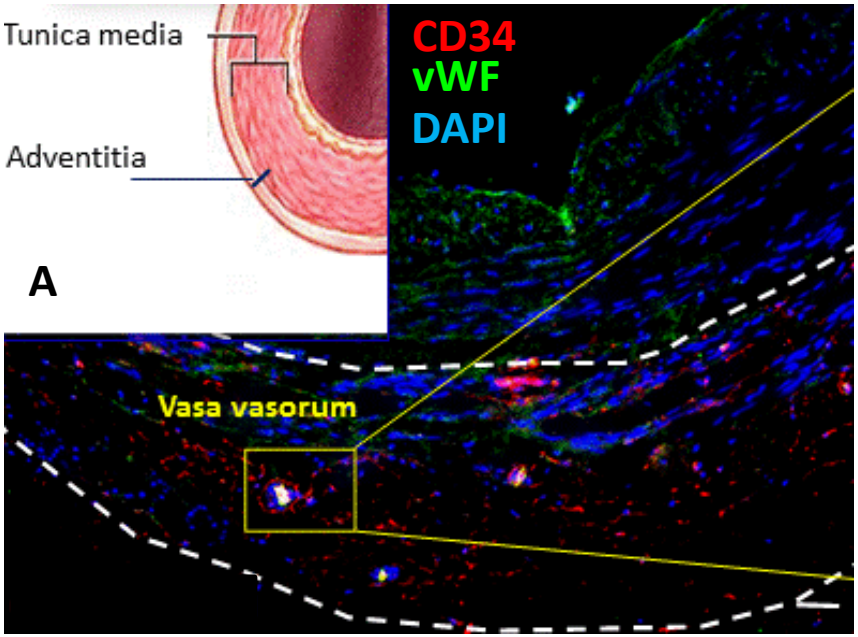
A convenient source of autologous progenitor cells

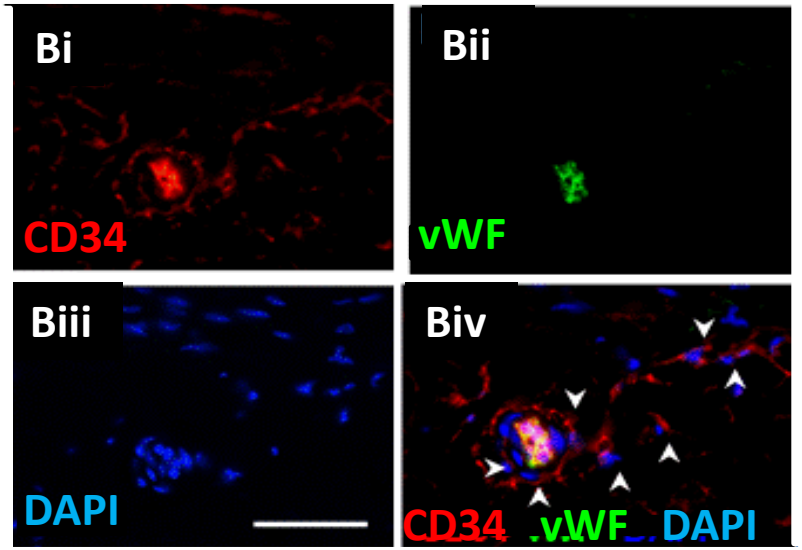


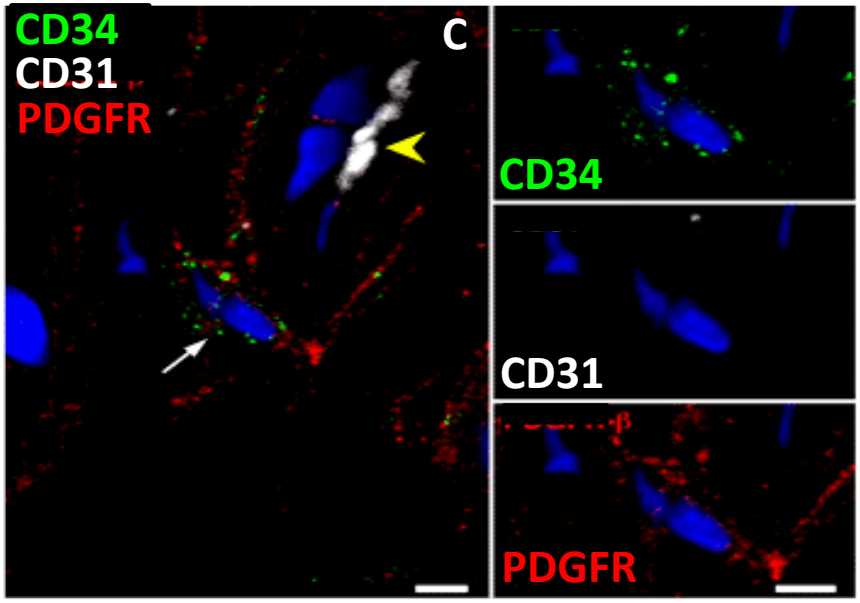
Vein leftover

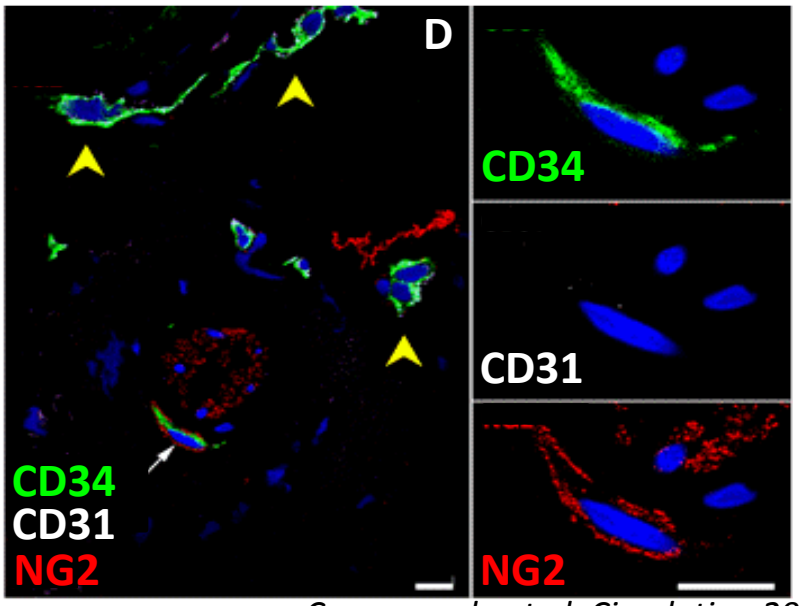


Localisation of SVPs in human saphenous vein adventitia



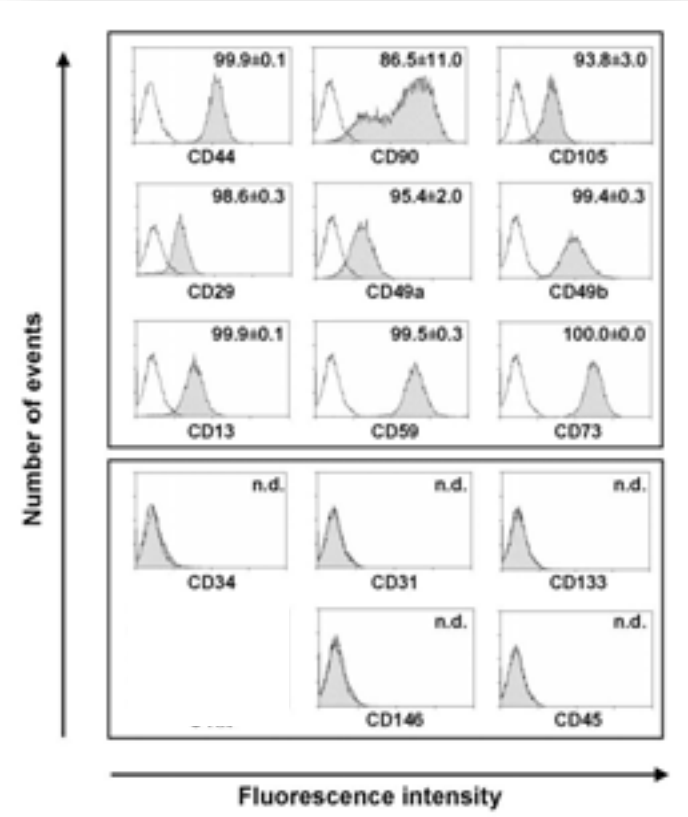


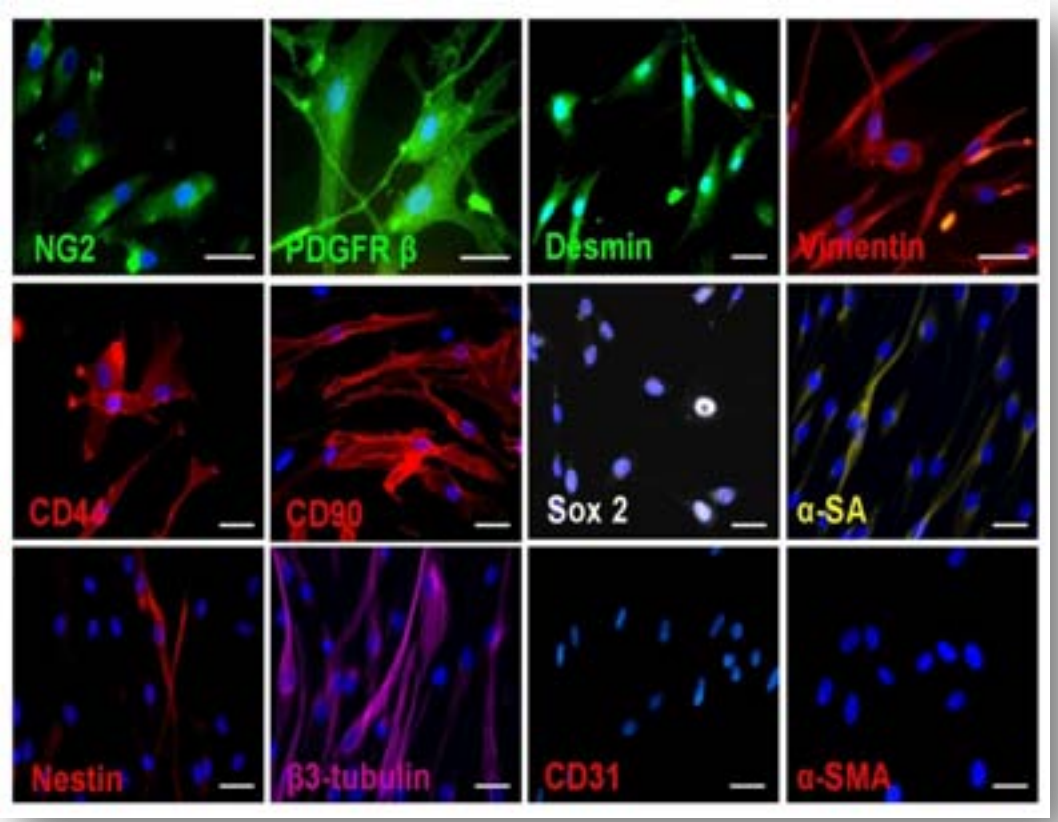




Campagnolo et al. Circulation 2010

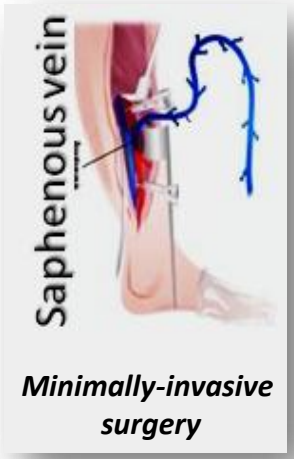
Expansion and characterization of SVPs from polyclonal preparations

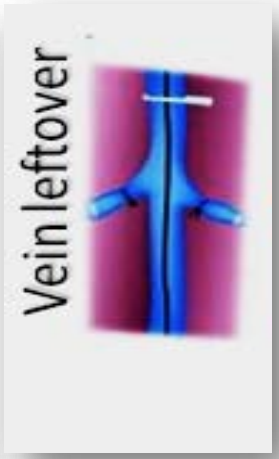


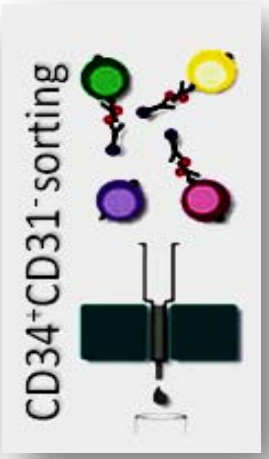


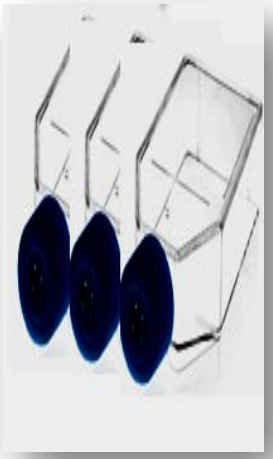
Campagnolo et al. Circulation 2010

Current standard operating protocol



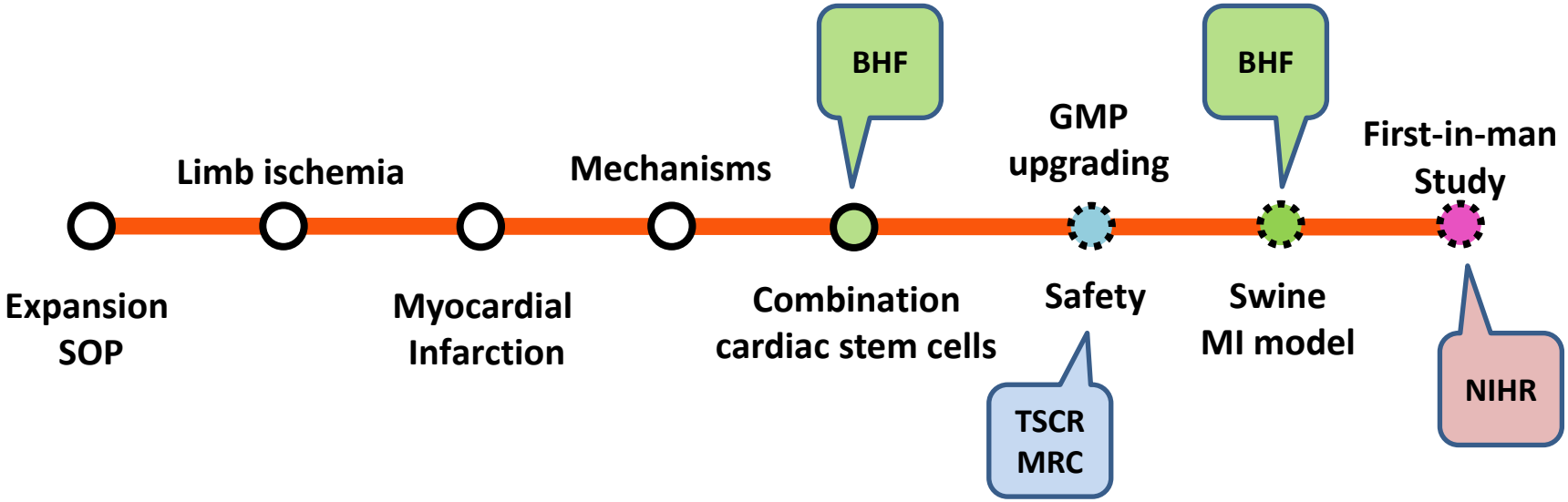




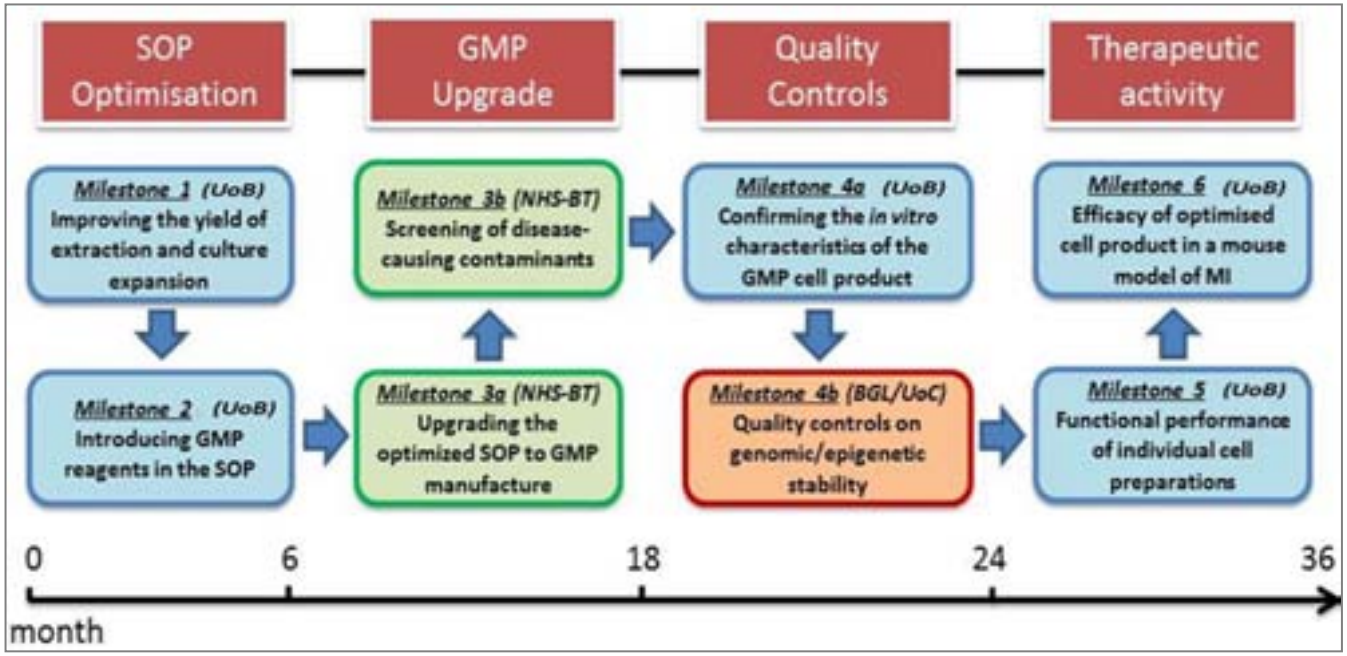


- Current SOP allowed successful expansion in 63% of 35 tested lines, which reached the therapeutic target of 30-50 million viable SVPs at passage 8 (P8) in ~10 weeks.
- Functional tests in 15 SV pericyte (SVP) lines, of which 10 derived from leftovers of coronary artery bypass grafts (CABG-SVP) and 5 from wastes of varicose SV from subjects with no evidence of coronary disease (NC-SVP)

The roadmap to first-in-man clinical trial

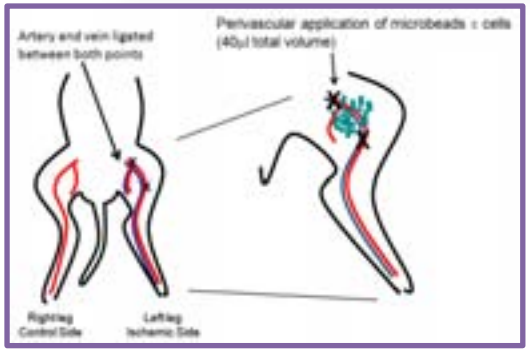


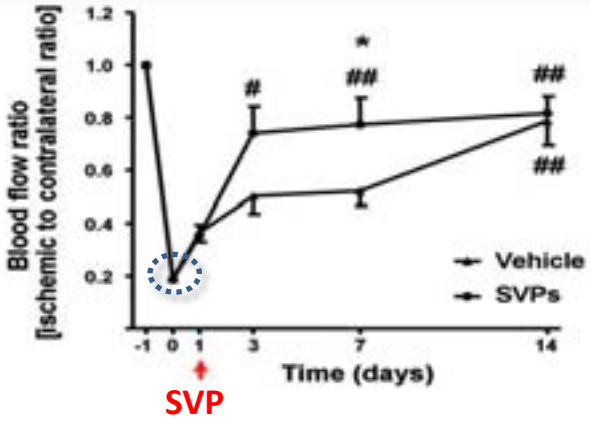
GMP upgrade and quality controls
NHS-BT , NHS-BLG and Cambridge Cancer UK

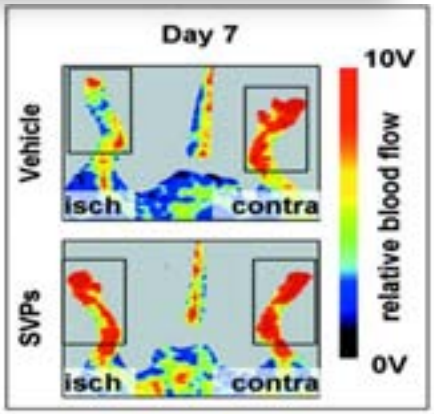


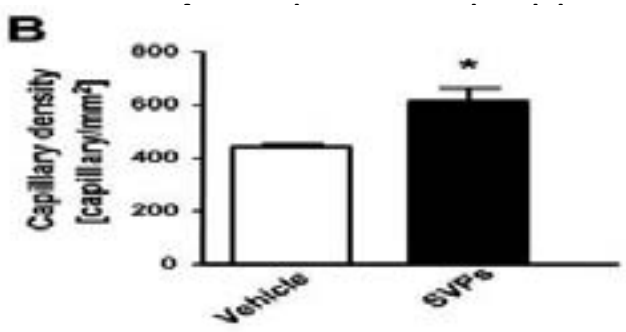
SVPs transplantation in a limb ischaemia model

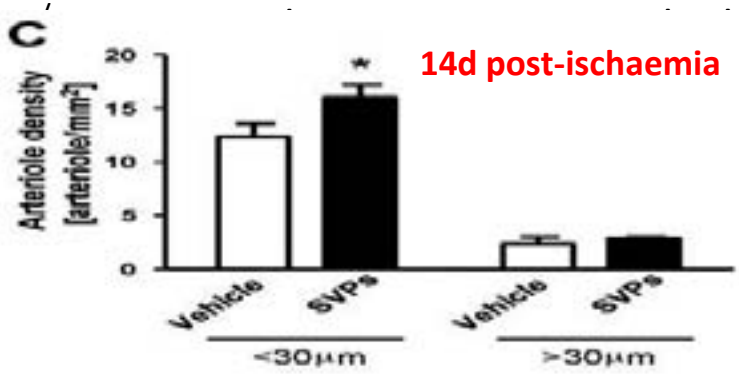
A







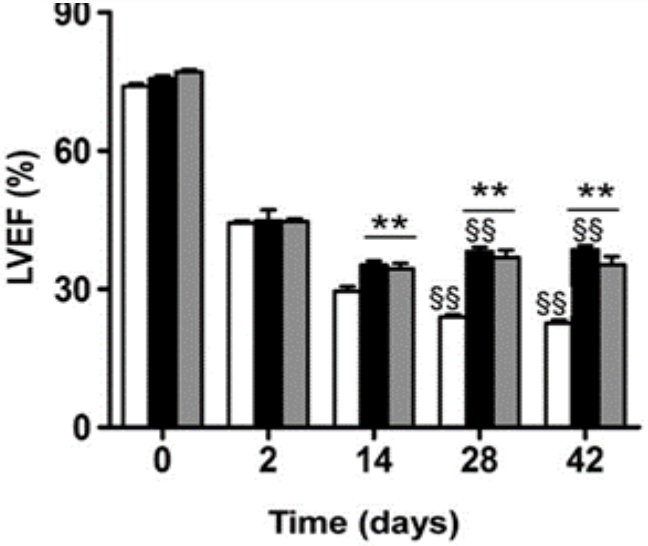


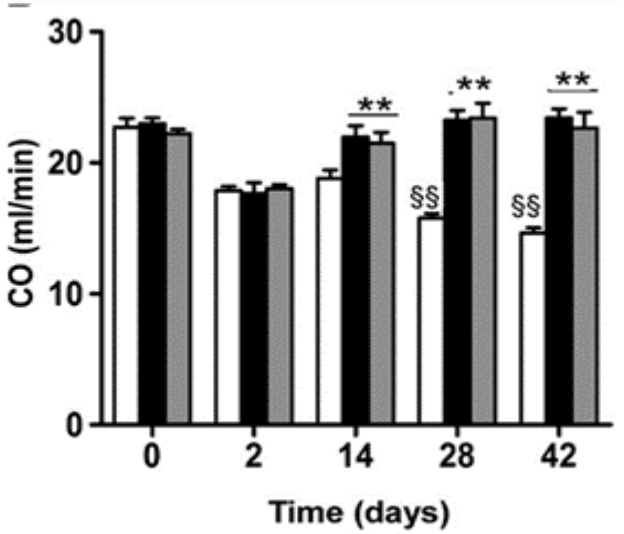


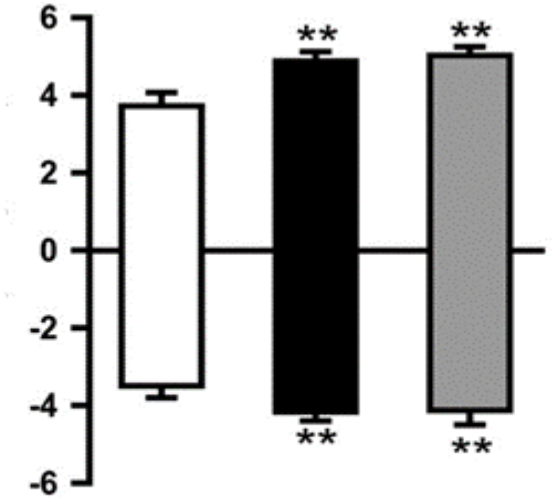
14d post-ischaemia

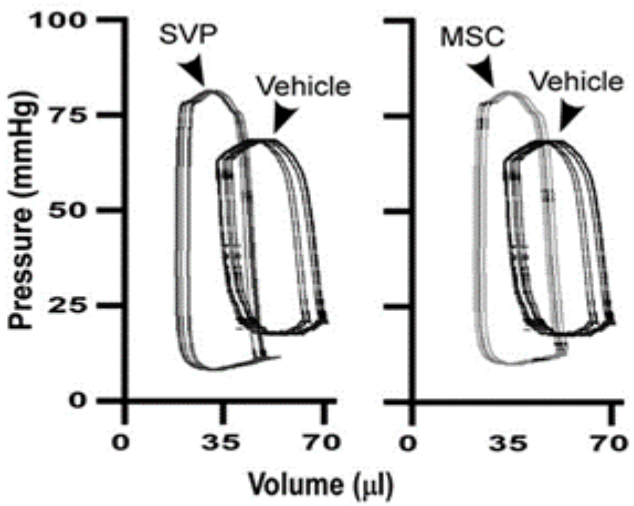
Campagnolo P et al. Circulation 2010;121:1735-1745

SVPs transplantation in a acute MI model





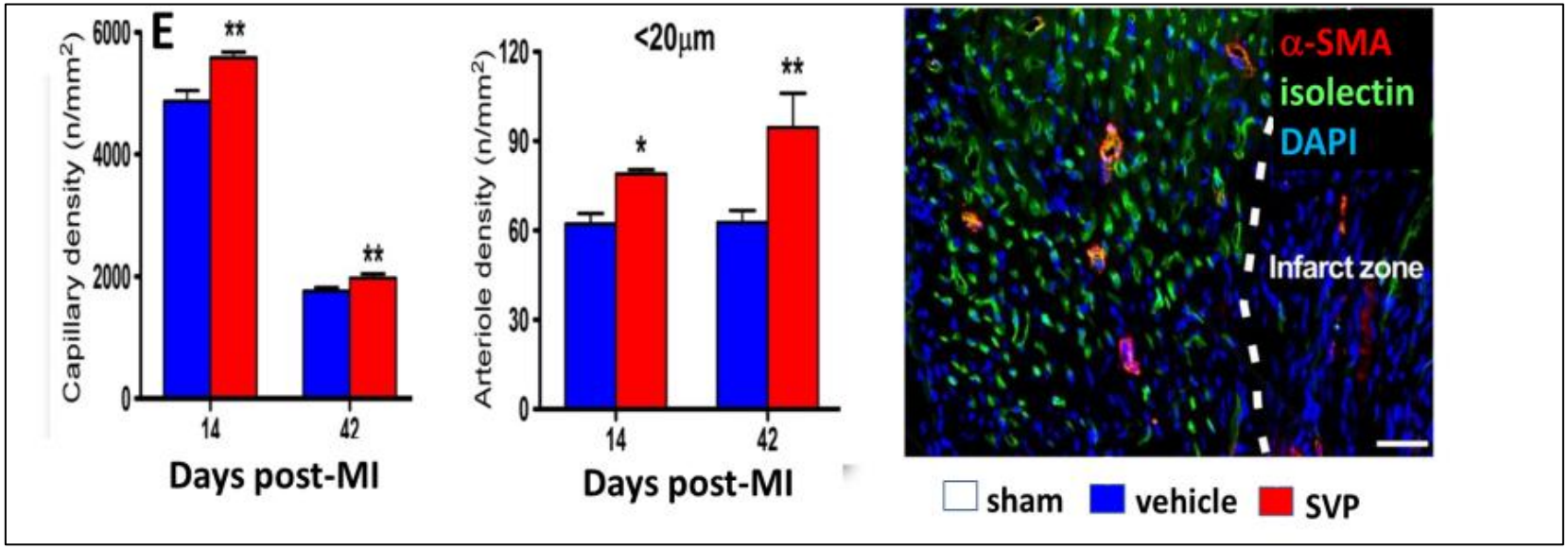


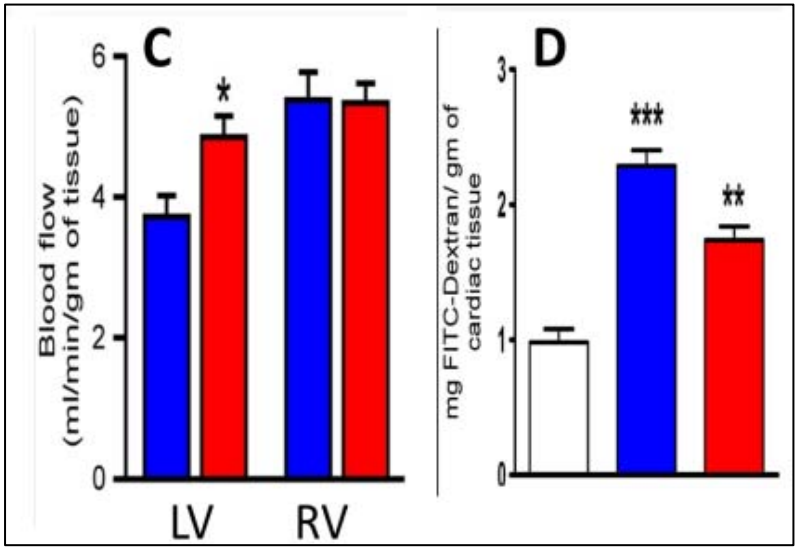


Vehicle SVP MSC

Katare R et al. Circulation Research 2011;109:894-906

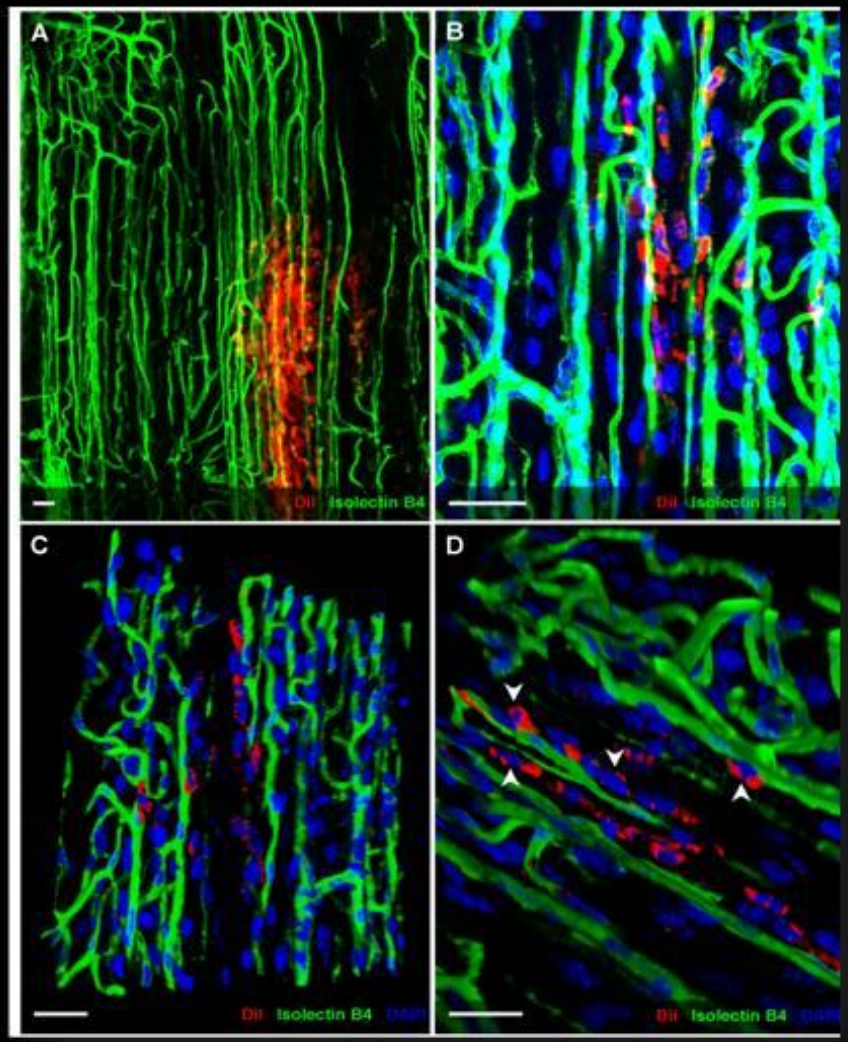
SVPs transplantation Improves neovascularization

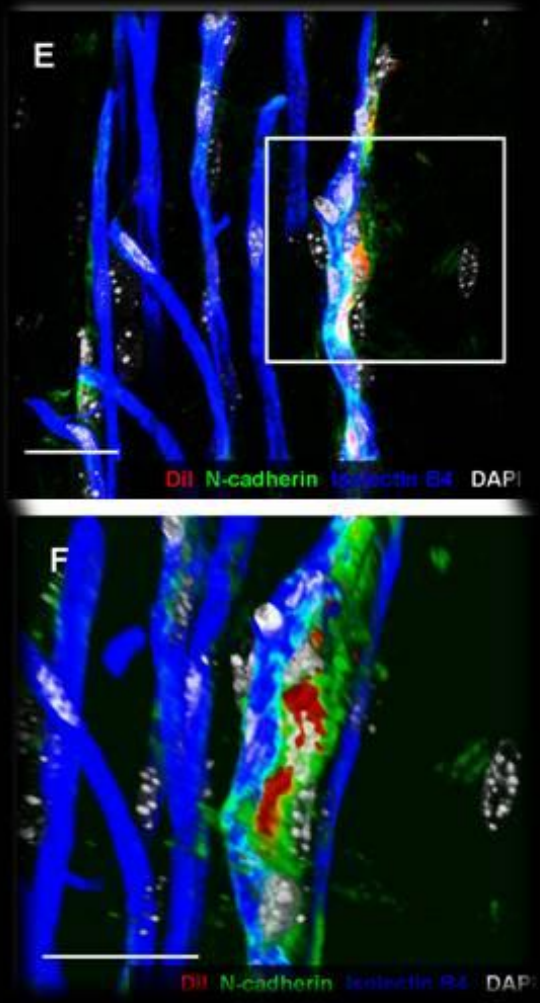




Katare et al. Circ Res 2011

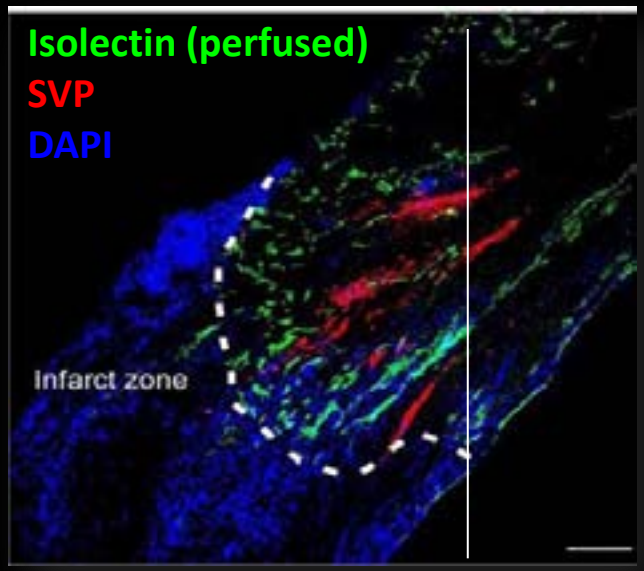
Tracking cell engraftment in the ischaemic limb

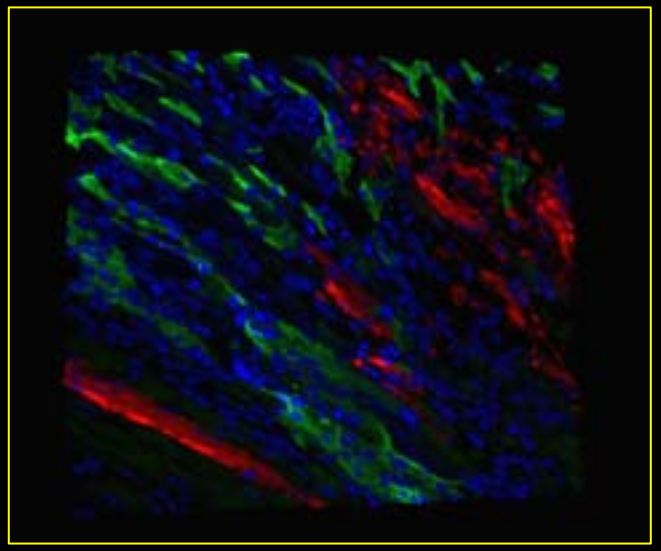


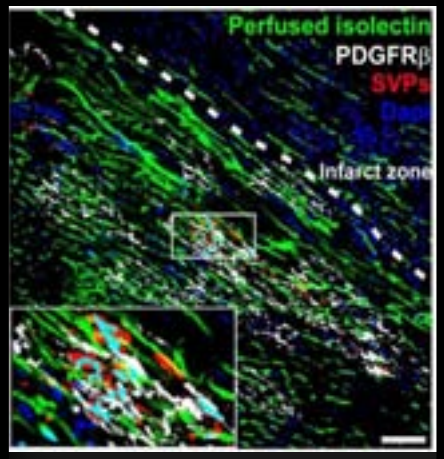


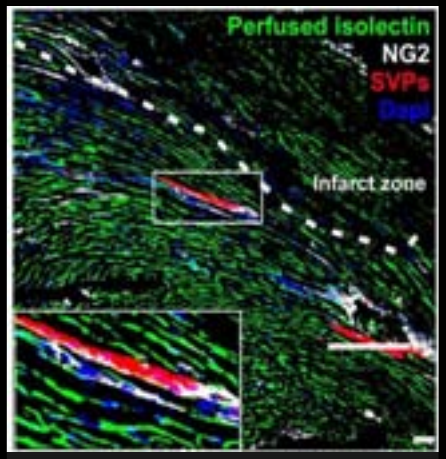
Campagnolo et al. Circulation 2010

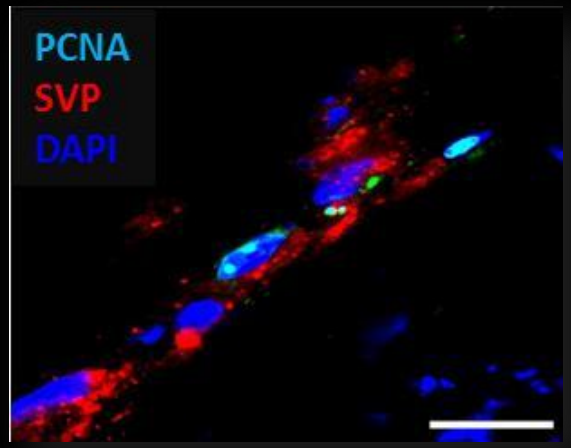
Tracking cell engraftment in the infarcted heart







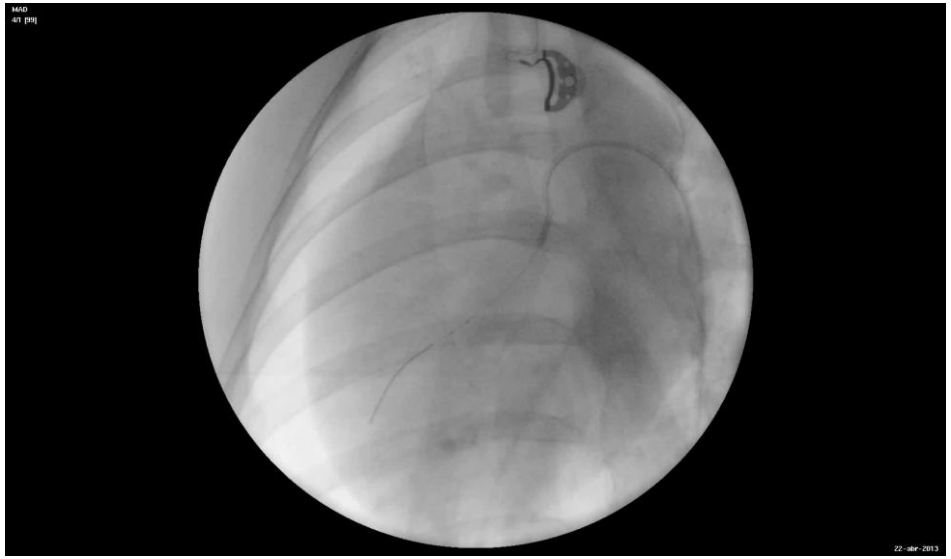




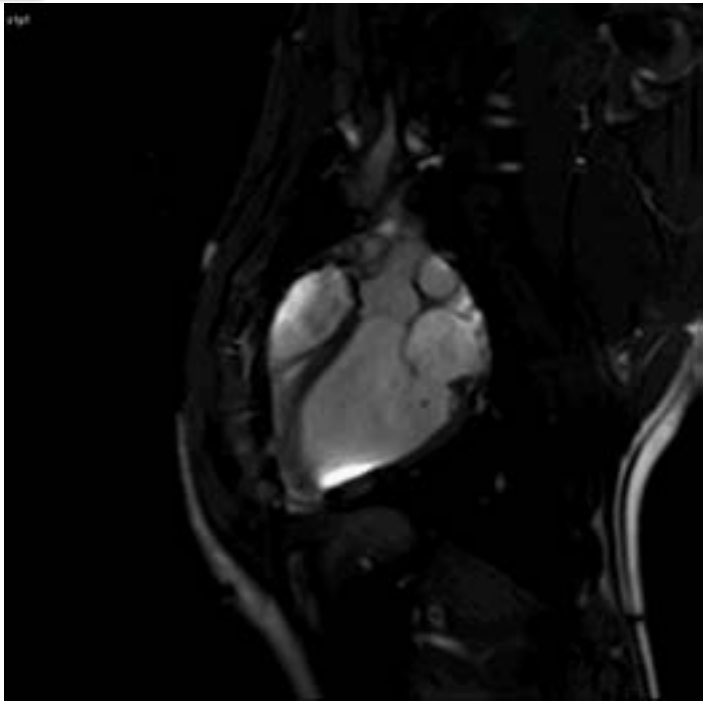
et al. Circ Res 2011

Pig Model

Balloon-induced MI

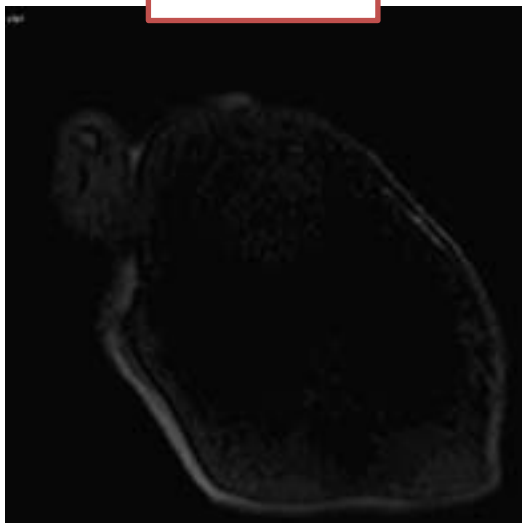


Cardiac MRI

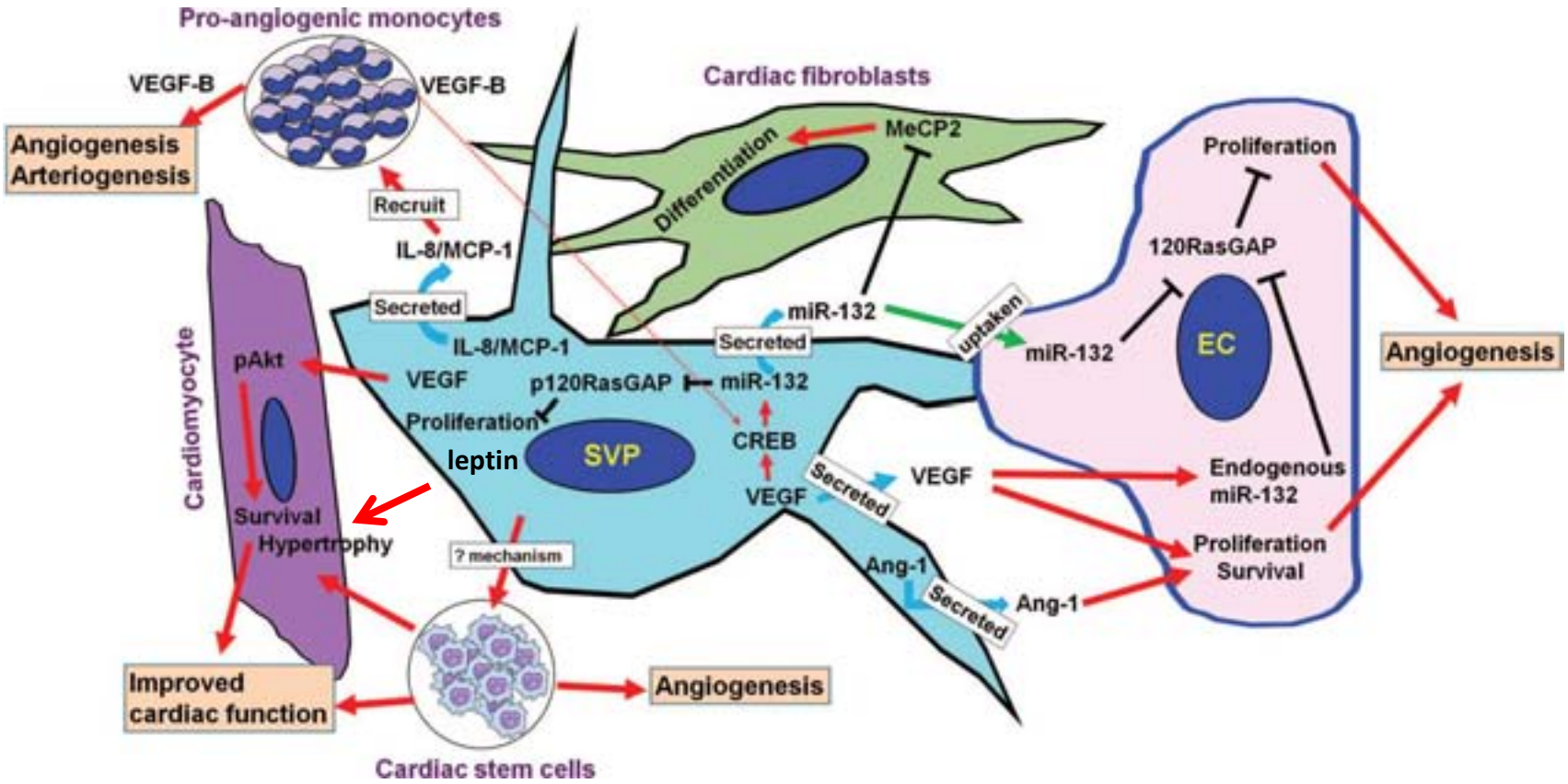


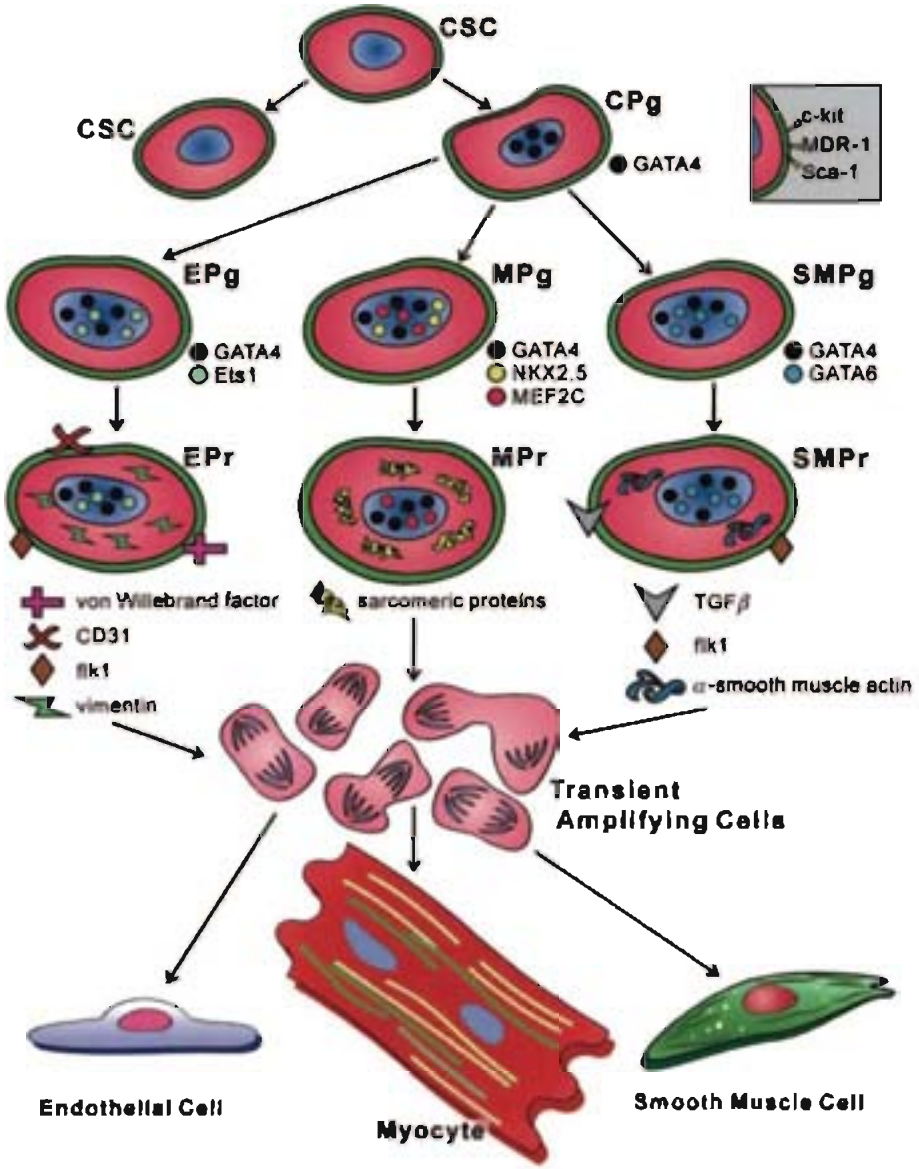
Cell Injection

Perfusion



Mechanisms of therapeutic action




CSC
CSC
CPg
GATA4
c-kit
MDR-1
Sca-1
EPg
GATA4
Ets1
MPg
GATA4
NKX2.5
MEF2C
SMPg
GATA4
GATA6
EPr
MPr
SMPr
von Willebrand factor
CD31
flk1
vimentin
sarcomeric proteins
TGFβ
flk1
α-smooth muscle actin
Transient
Amplifying Cells
Endothelial Cell
Myocyte
Smooth Muscle Cell

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