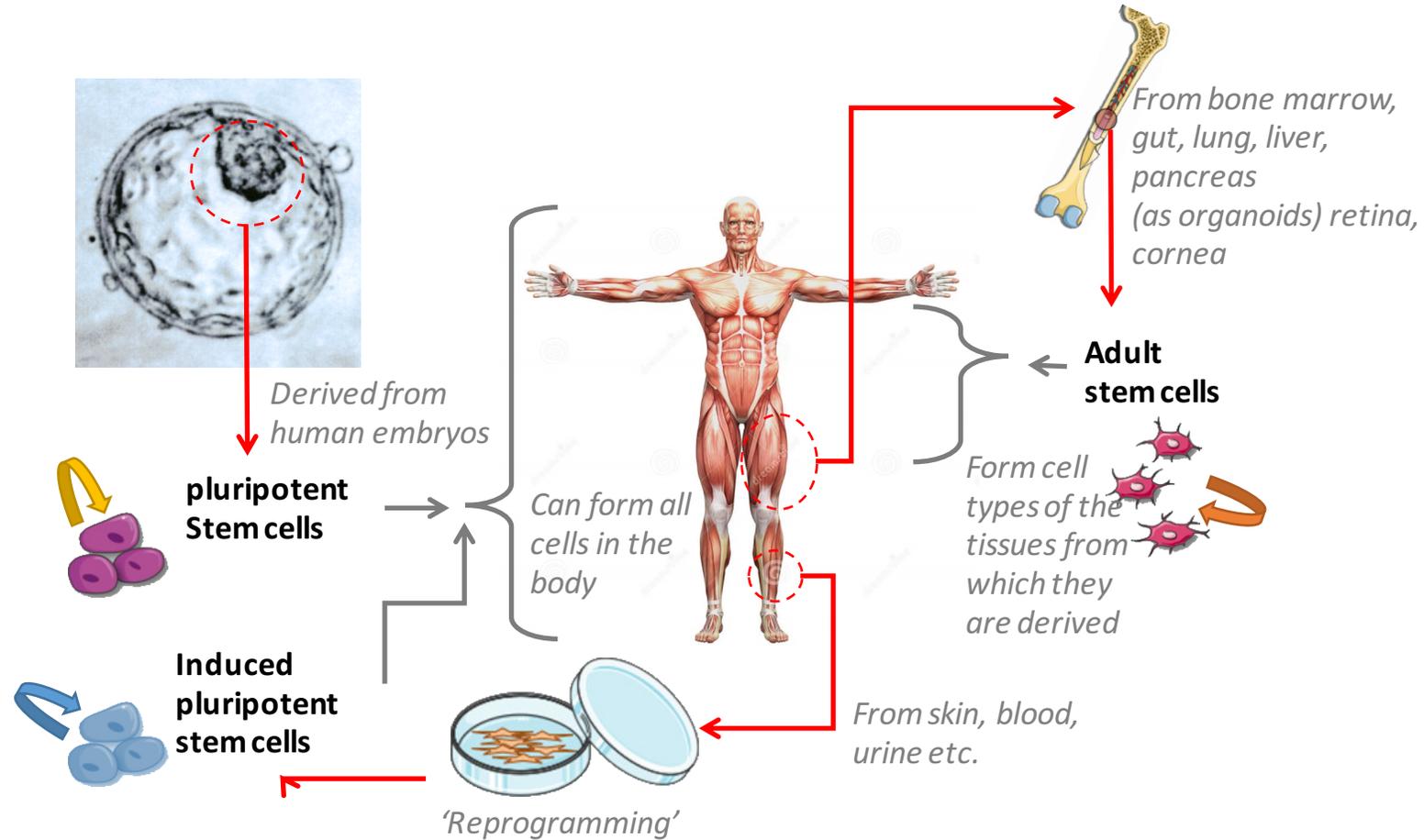


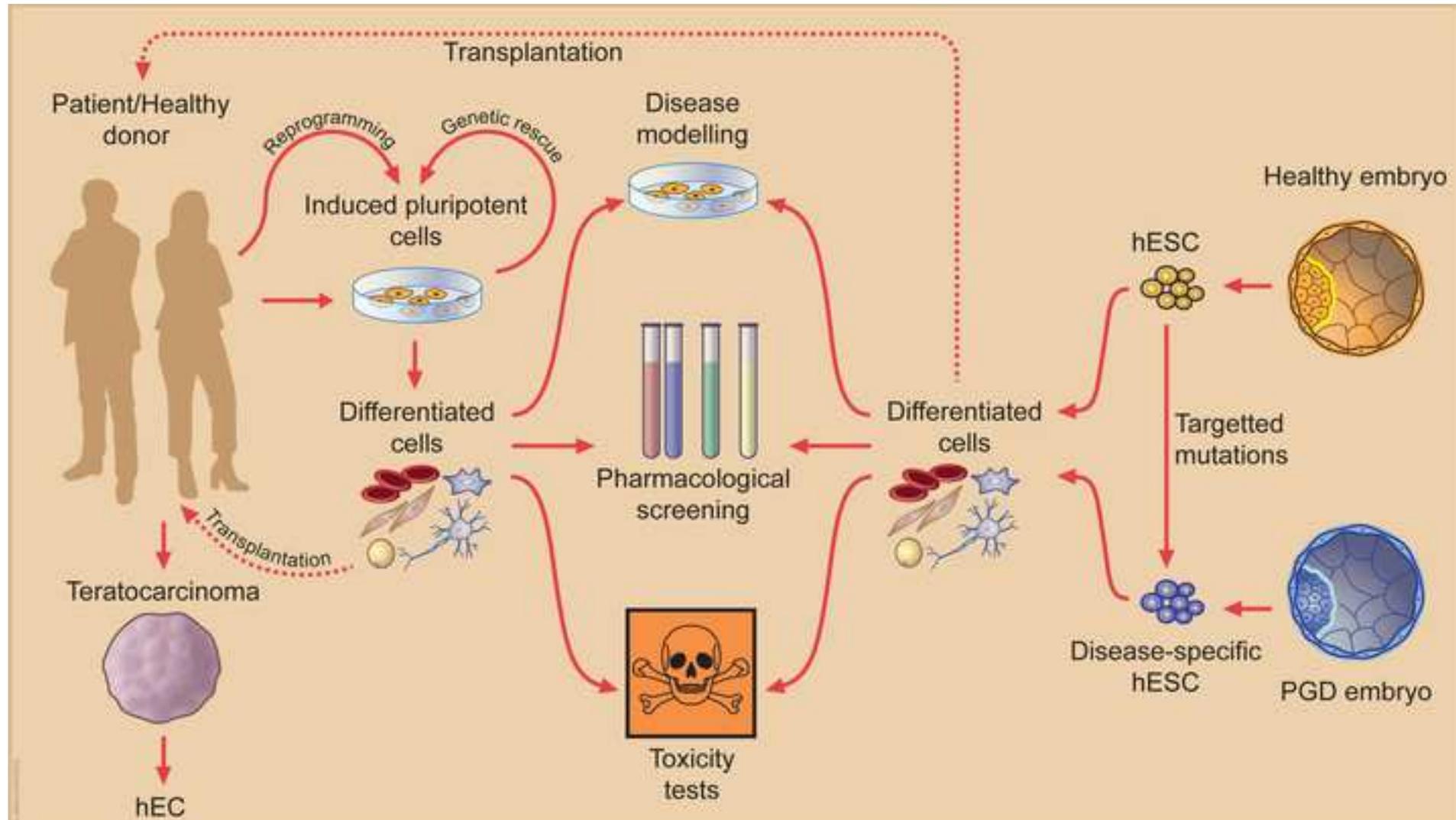
EMA workshop Organs on Chip 2017

Populating organs-on-chip with cells derived from human stem cells

WHICH HUMAN STEM CELLS ?



STEM CELL MODELS OF DISEASE



THE PROBLEM

- No drugs for many chronic diseases
- Existing drugs do not work in all patients
- Drug side effects are the 4th leading cause of death

WHY?

- Poor insight into *human* disease mechanisms
- Lack of personalized treatment prediction
- Animals are poorly predictive for *humans*

heart, immune system, brain, reproductive system, stomach....

CHALLENGES TO TRANSLATION OF hPSCs

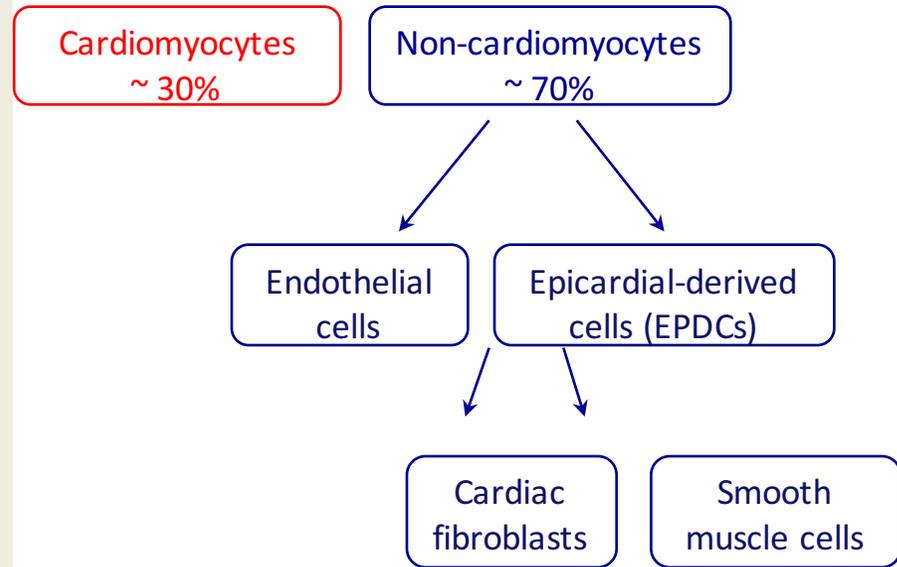
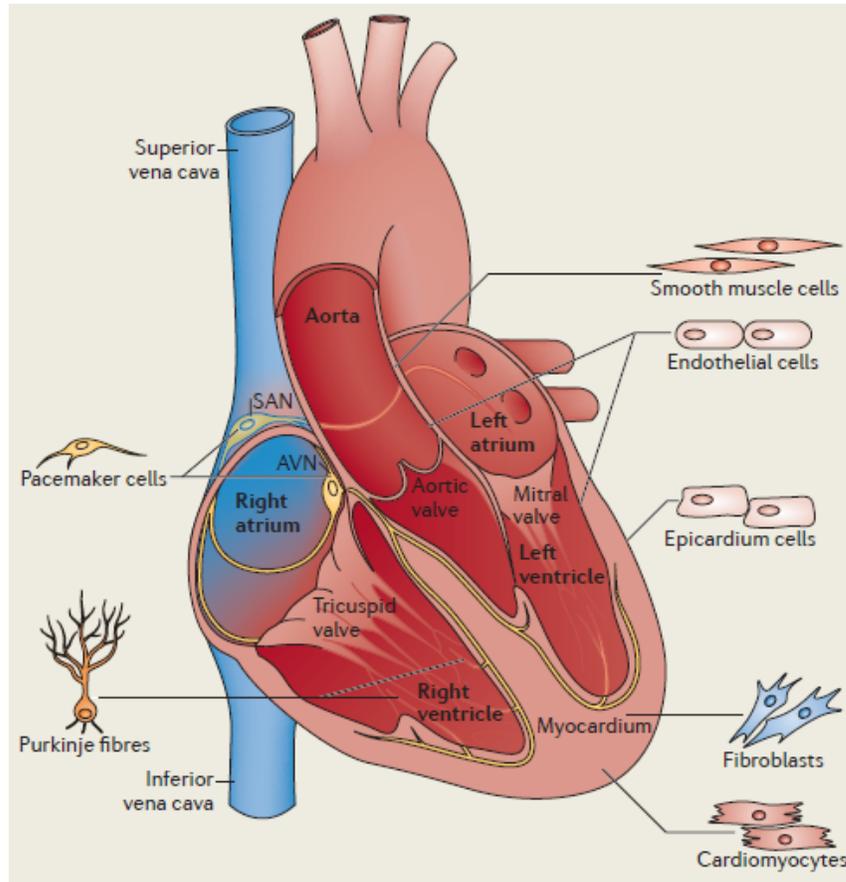
Line-to-line variability

Directed differentiation to functional cells

Maturation of differentiated cells

Genetic stability

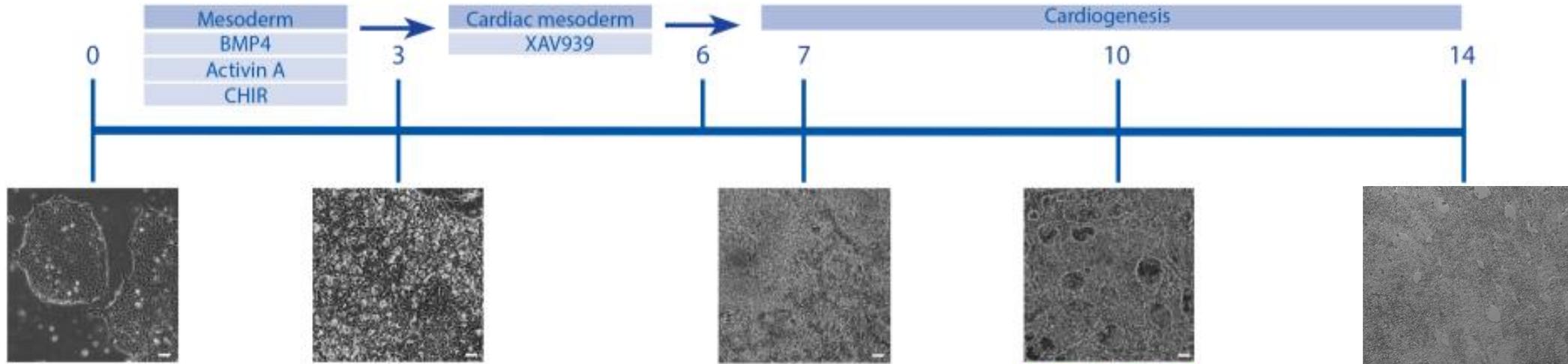
TISSUES ARE MULTICELLULAR : HEART AS EXAMPLE



Xin M. et al. Nat Rev 2013; Tirziu D. et al. Circulation 2010; Furtado MB. et al. Dev. 2016

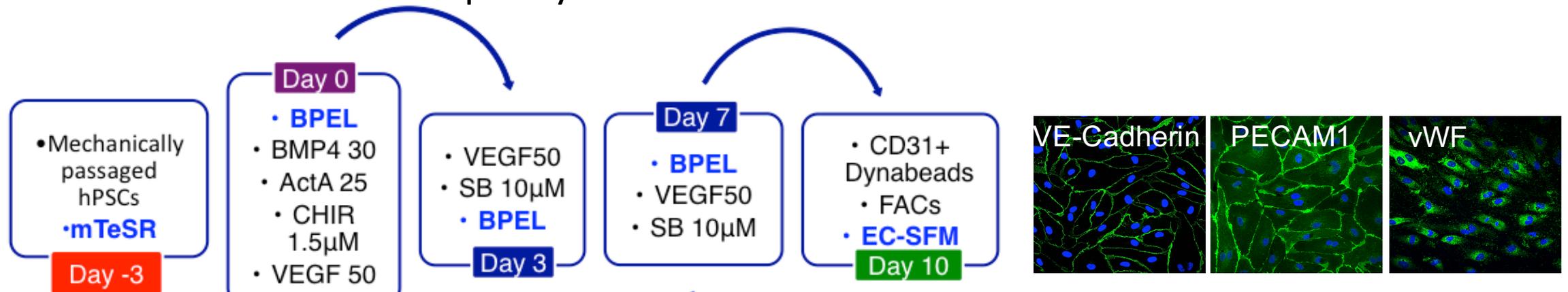
DIRECTING DIFFERENTIATION IN DEFINED CONDITIONS

Cardiomyocytes



Van den Berg et al 2015

Vascular endothelial cells and pericytes



Orlova et al., ATVB, 2013, Nat Protocols 2014

Are these cardiac and vascular cells functional?

CARDIOMYOCYTES FROM HPSC ARE ELECTRICALLY ACTIVE:

used in drug safety and toxicology

The Comprehensive in Vitro Proarrhythmia Assay (*CiPA*) initiative

human

QRS

QT

mouse

'QRS'

QT

0

0.5

1.0

0

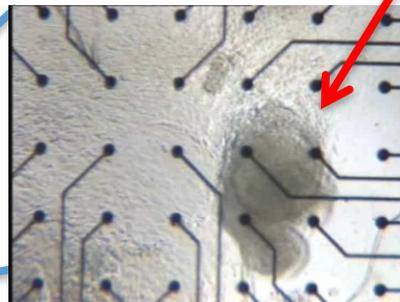
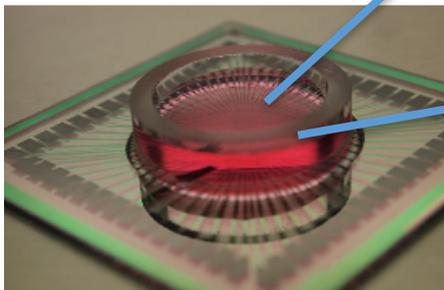
50

100

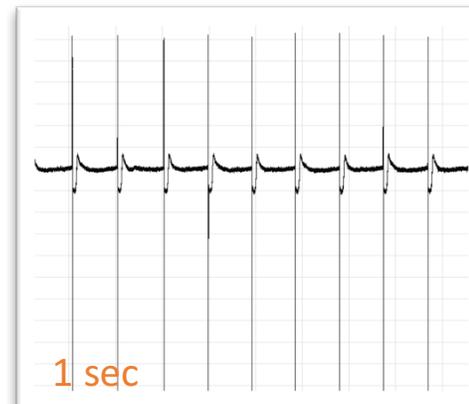
Electrocardiogram

"personal" heart cells

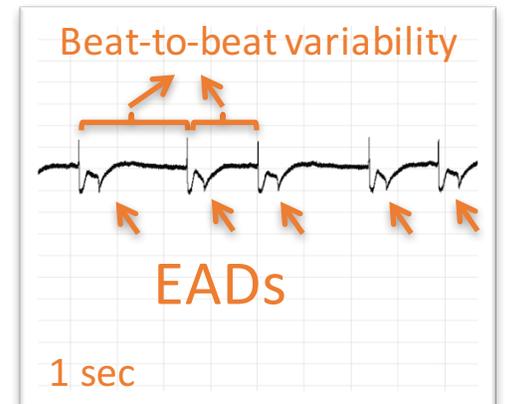
chip



Control

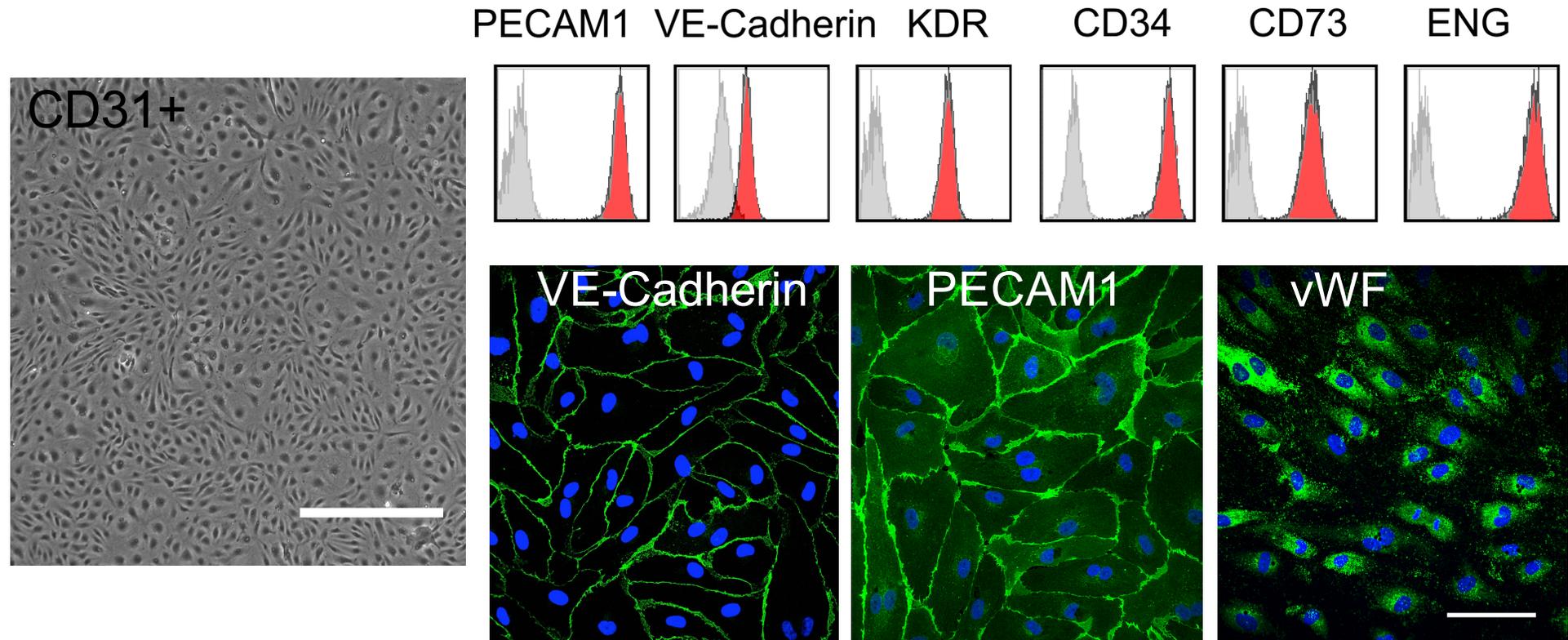


Dofetilide



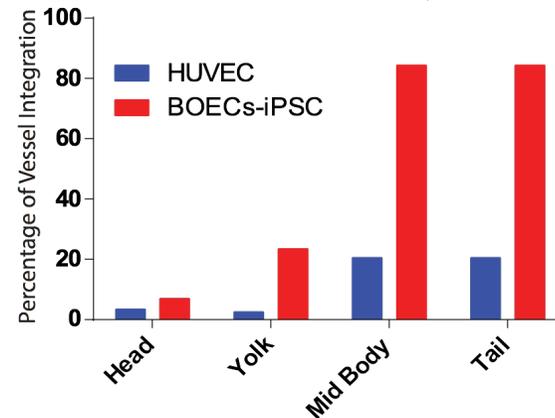
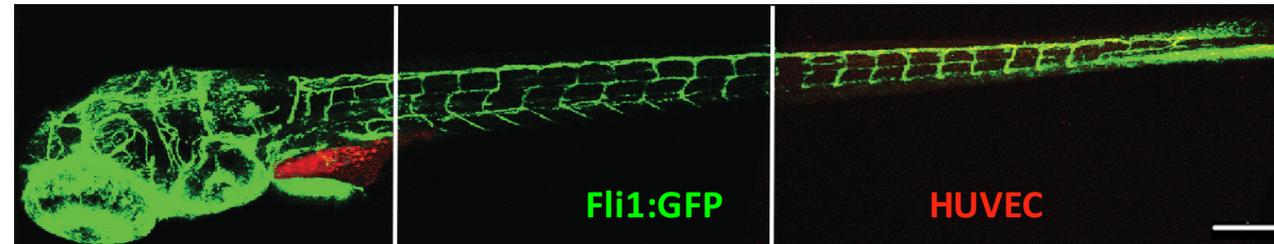
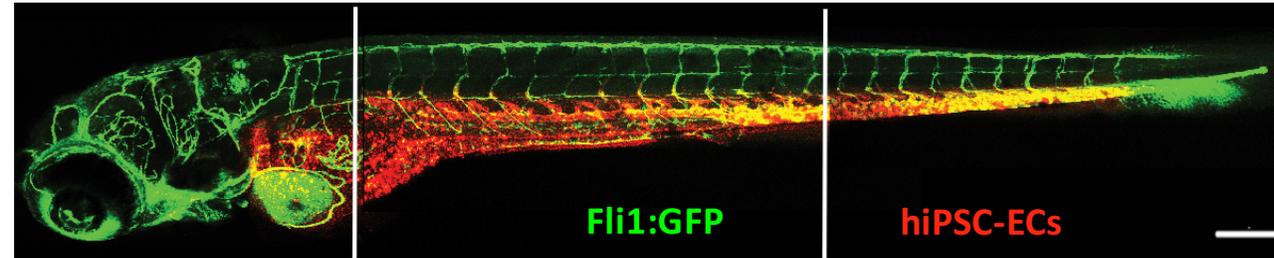
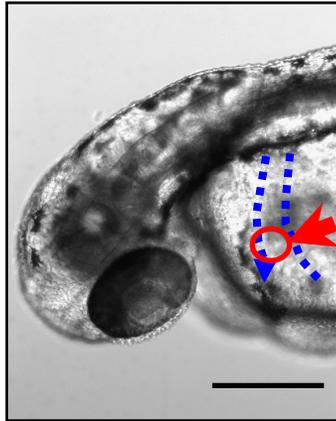
Induction of cellular arrhythmias

ENDOTHELIAL CELLS FROM HPSC



VASCULAR COMPETENCE OF hPSC-ENDOTHELIAL CELLS IN ZEBRAFISH

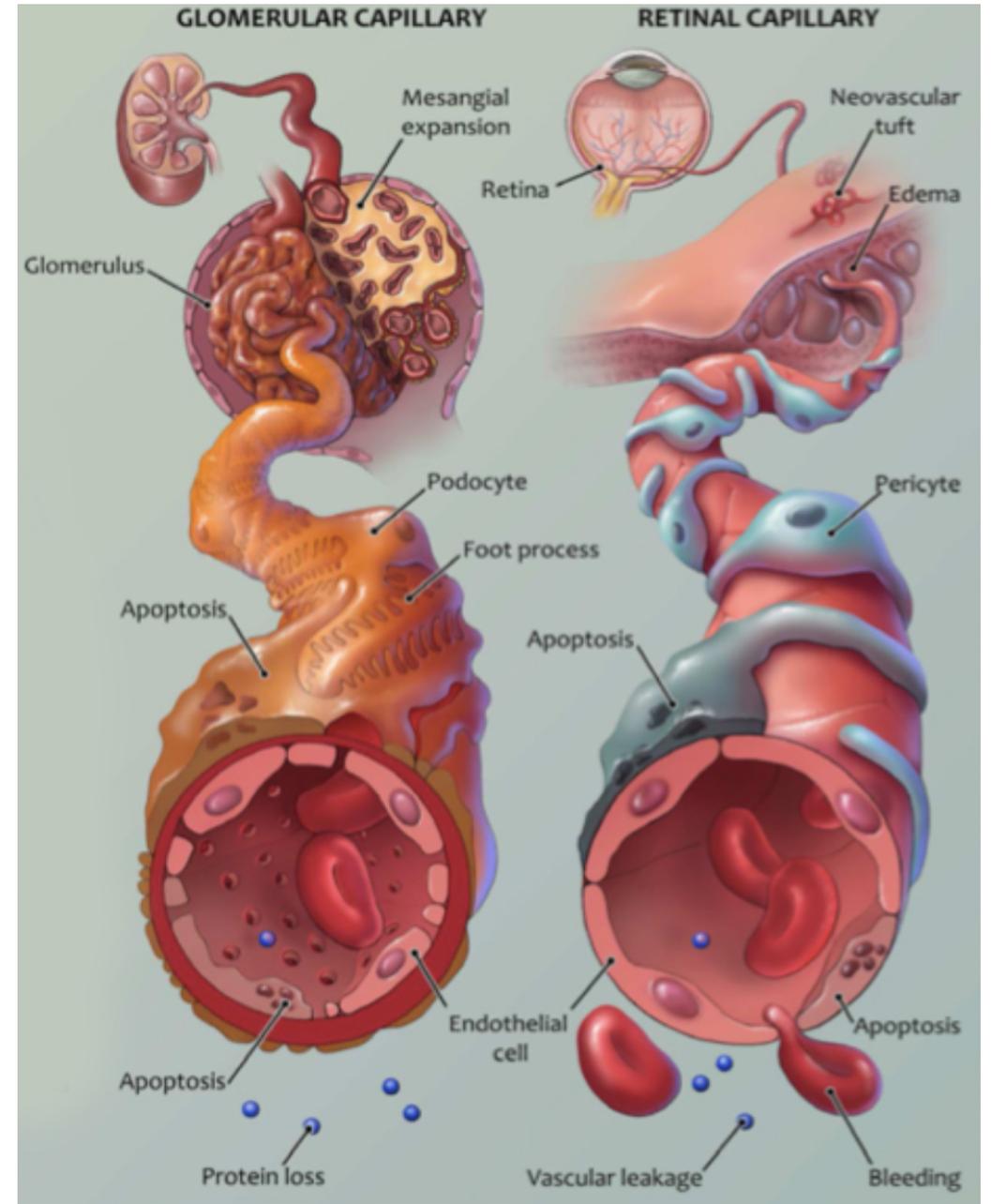
48 hpf transplantation
(duct of Cuvier)
~400 hu ECs



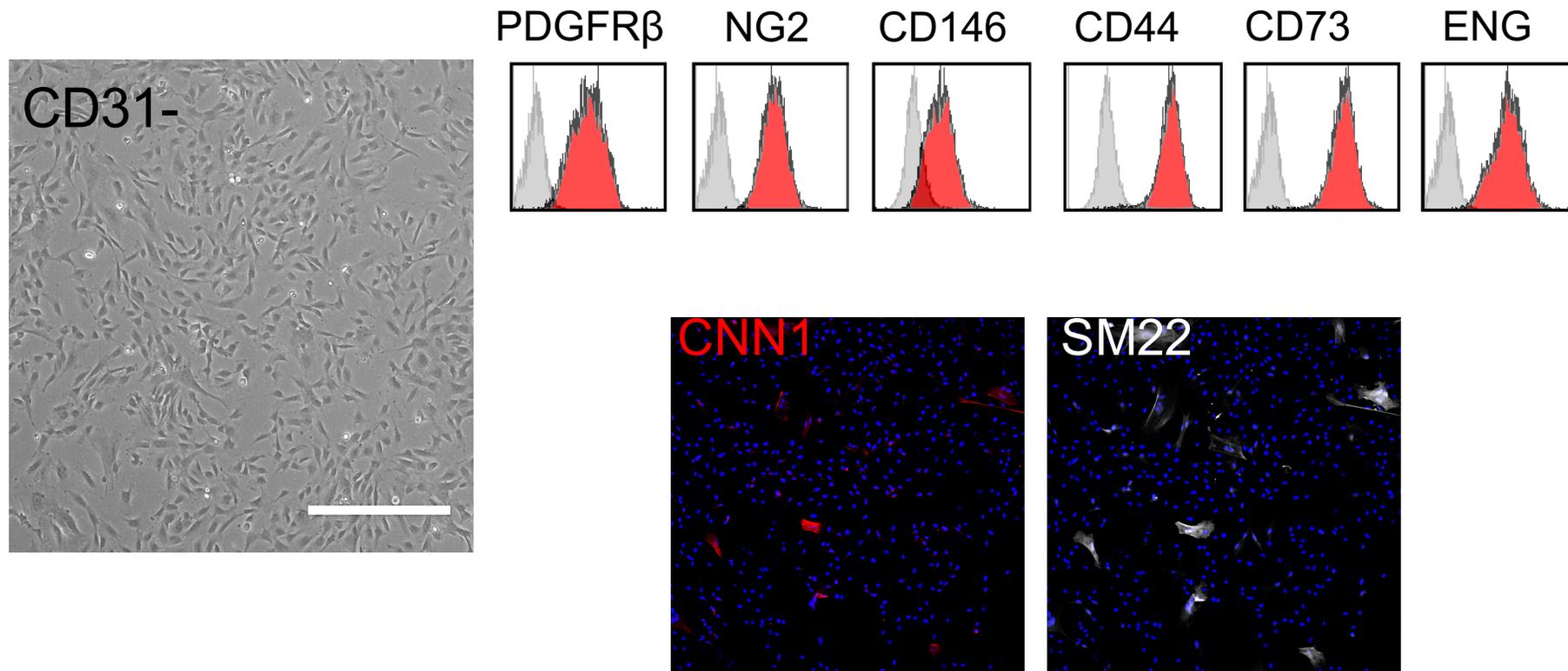
hiPSC-derived ECs integrate with much higher efficiency than adult ECs, such as human umbilical vein endothelial cells (HUVEC).

PERICYTES/SMOOTH MUSCLE CELLS COVER VESSELS TO CREATE STABILITY

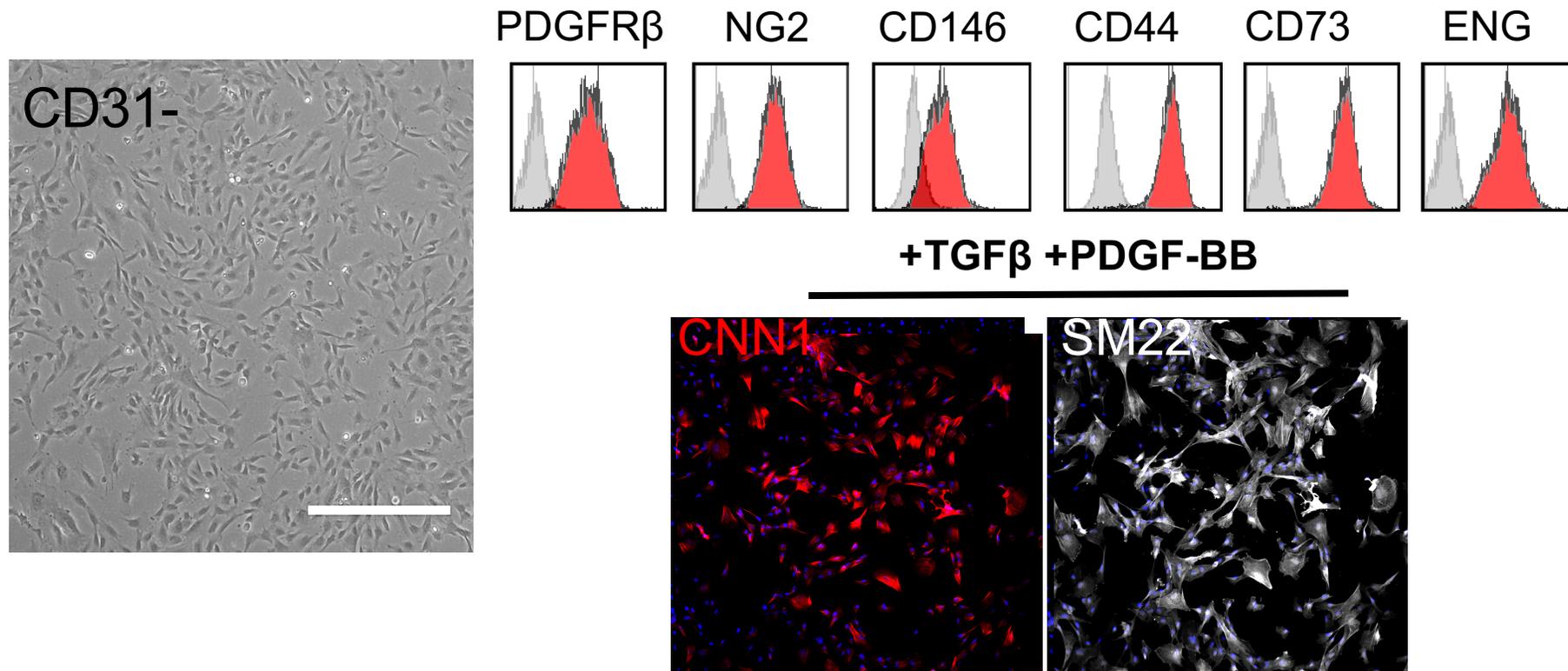
Each vessels has its own type of pericyte



PERICYTES AND SMOOTH MUSCLE CELLS FROM hPSCs



PERICYTES AND SMOOTH MUSCLE CELLS FROM hPSCs

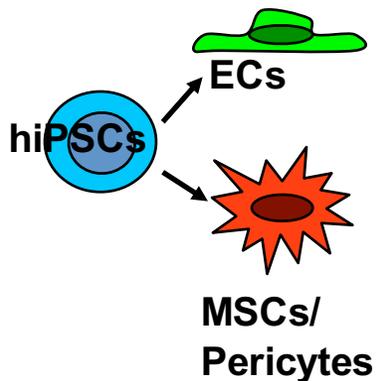
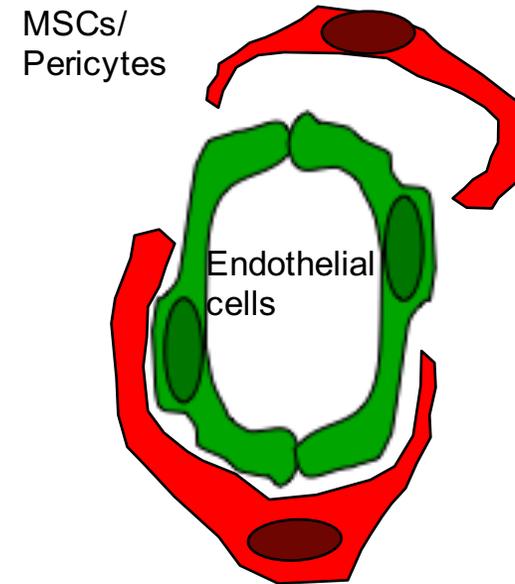
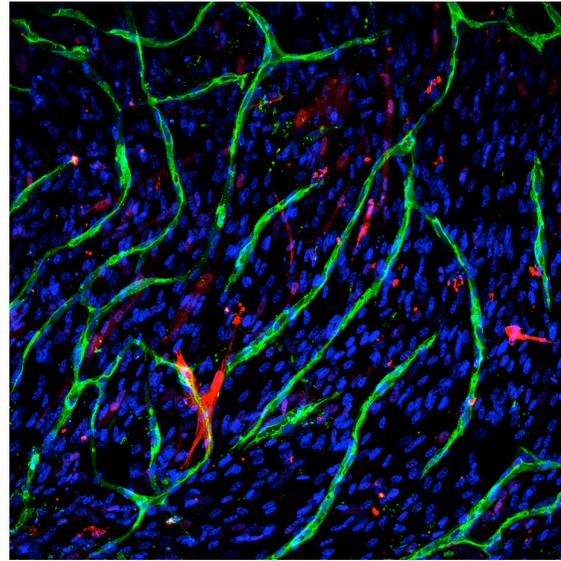
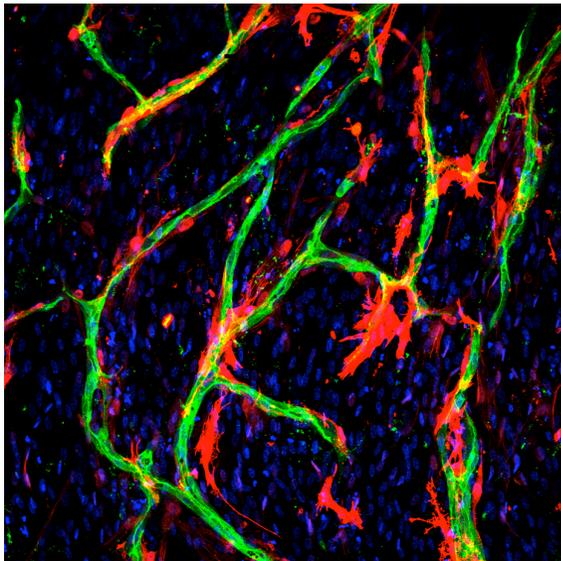


CO-CULTURES OF hPSC ENDOTHELIAL CELLS AND PERICYTES

day 7

DAPT

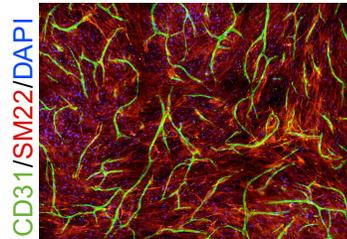
PECAM1/SM22/DAPI



CTRL or diseased



CTRL or diseased

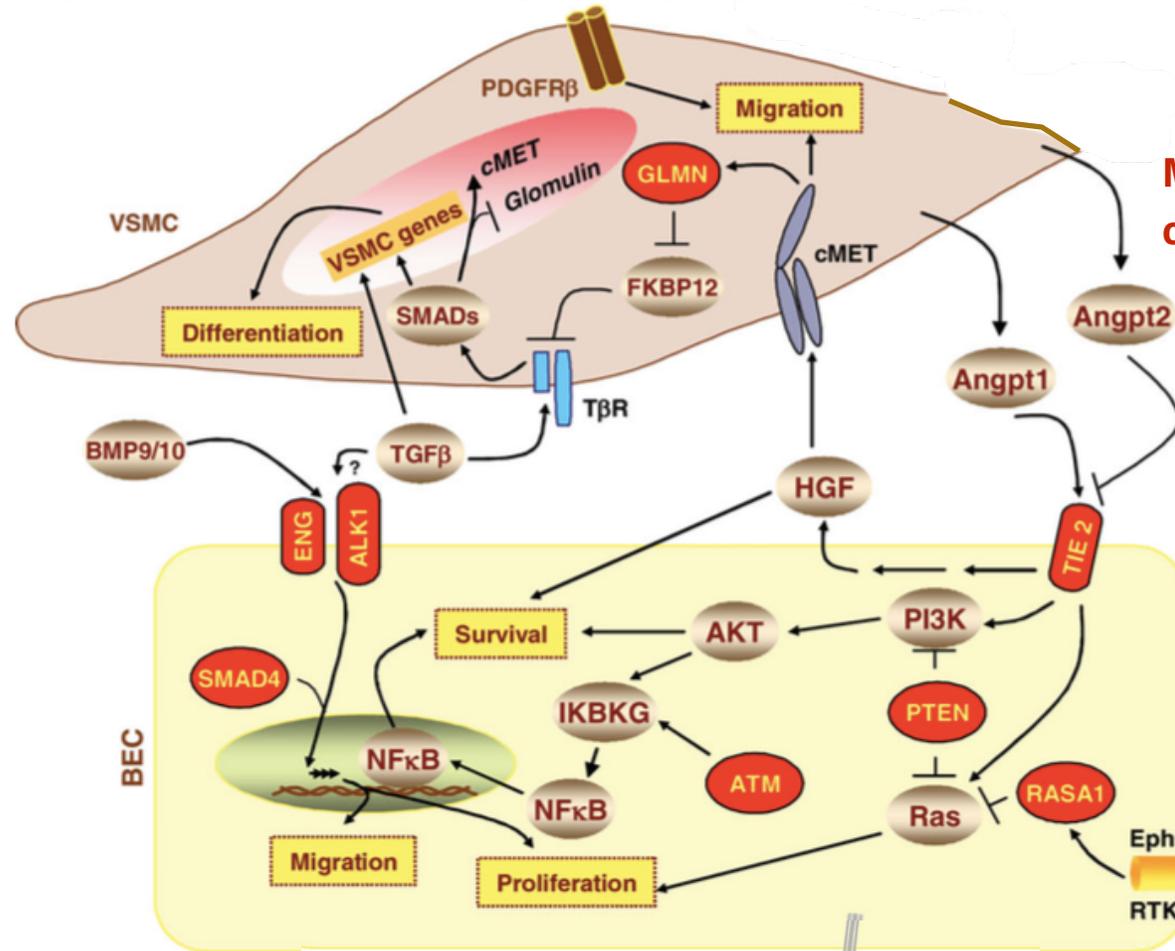


DAPT
(γ -secretase inhibitor)

..... **NOTCH**

CADASIL: genetic disease caused by defective Notch signalling

GENETIC CAUSES OF VASCULAR MALFORMATIONS



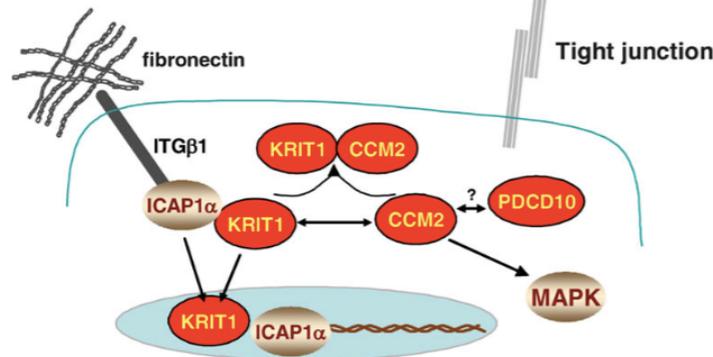
Mutations causing arterial, venous and capillary malformations:

Hereditary hemorrhagic telangiectasia (ENG, ALK1, SMAD4)

Cutaneomucosal venous malformation (TIE2)

Capillary malformation-arteriovenous malformation (RASA1)

Ataxia-telangiectasia (ATM)

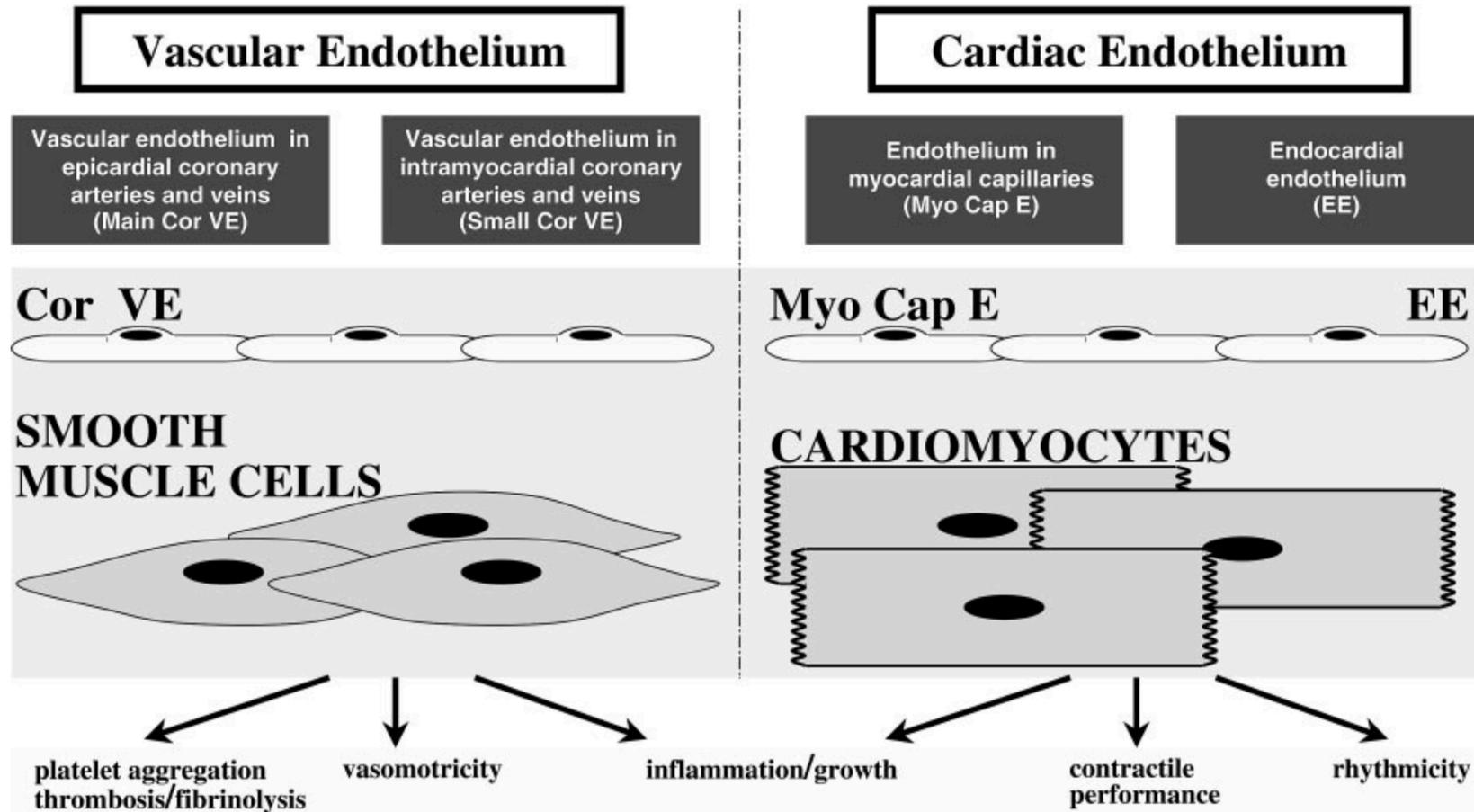


Cerebral cavernous (or capillary) malformation (KRIT1, CCM3, PDCD10)

Cross talk between cardiac myocytes and cardiac endothelial cells

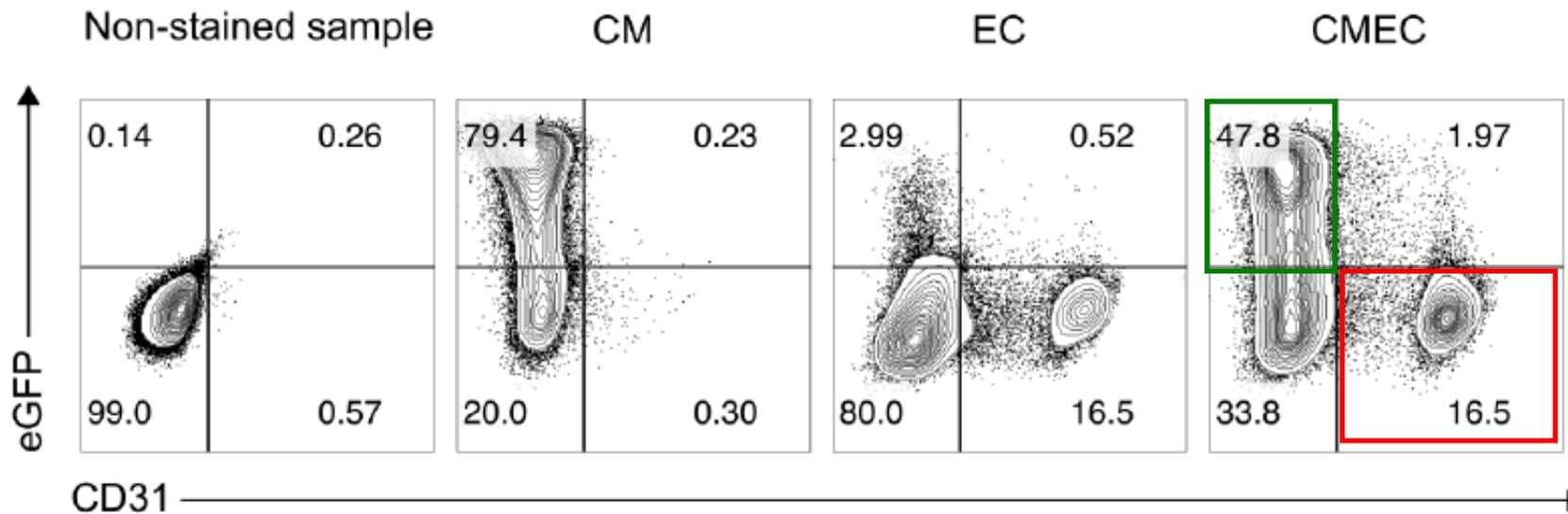
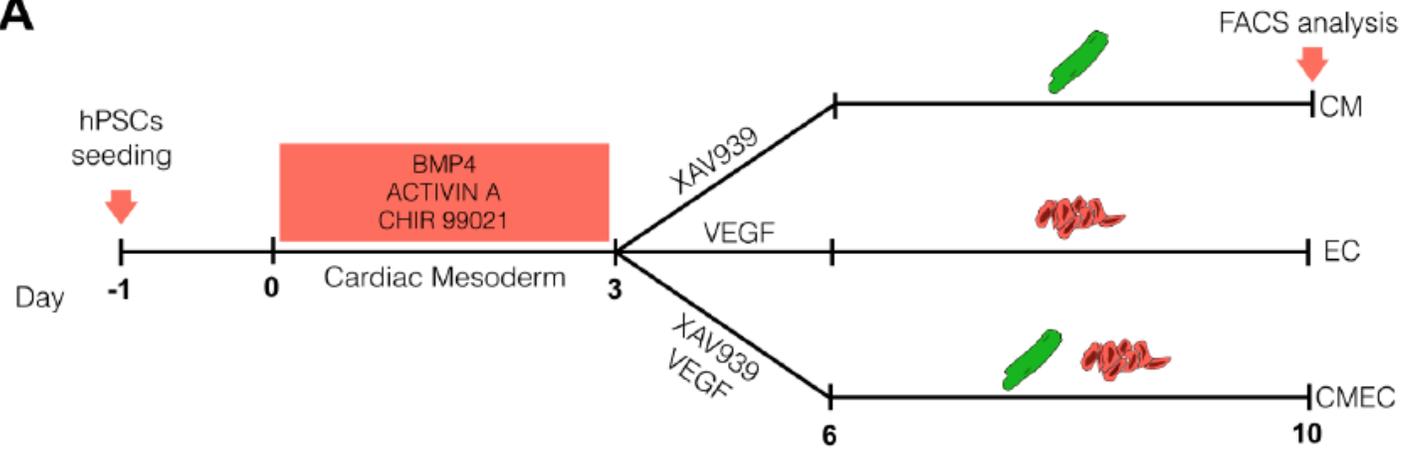
Cardiac endothelial cells

1. Form the myocardial microvasculature, which supplies oxygen and free fatty acids to cardiomyocytes
2. Release paracrine factors that regulate cardiomyocyte metabolism, survival and contractile function

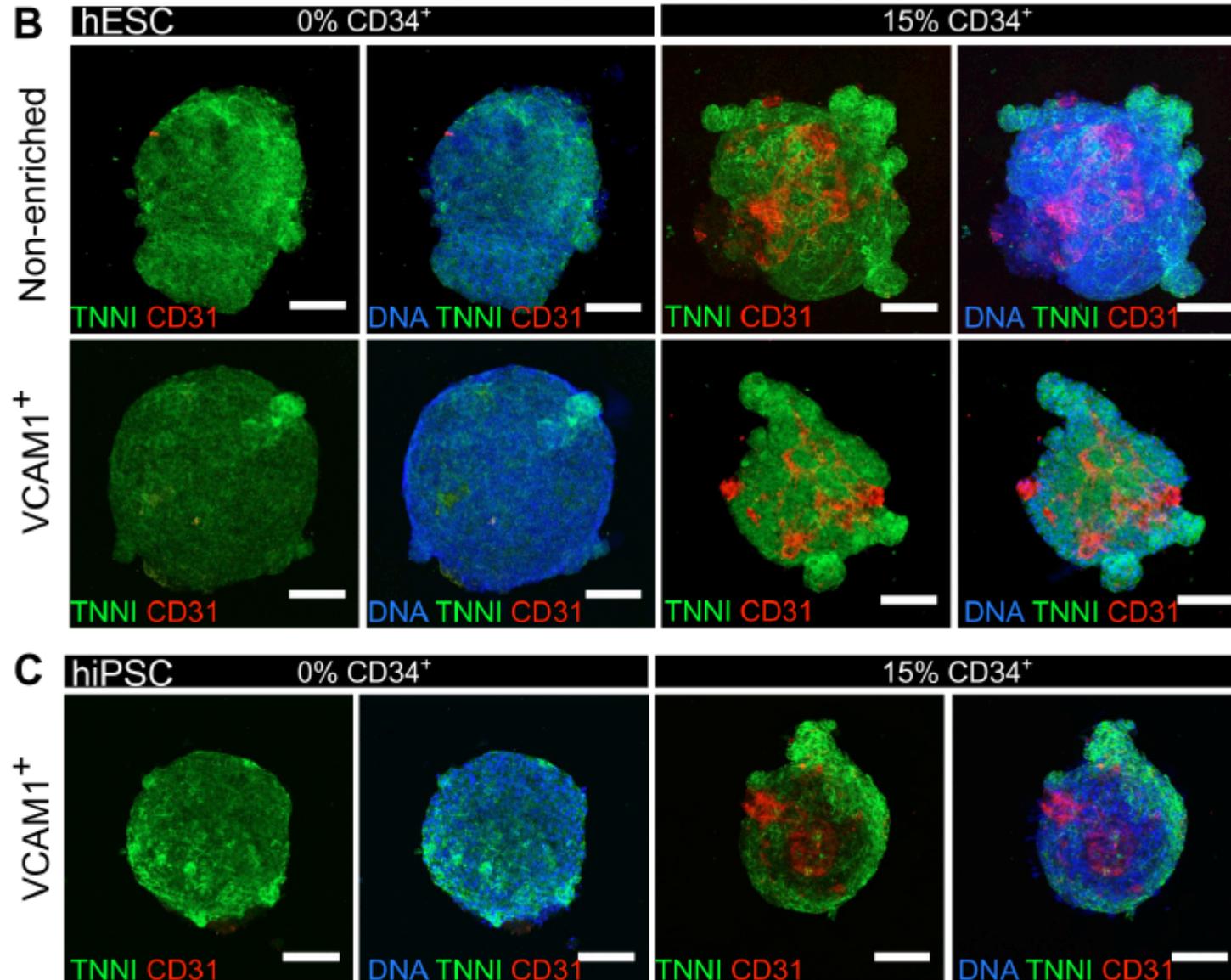


Simultaneous differentiation of hPSC into cardiomyocytes and endothelial cells from cardiac mesoderm

A



Cardiomyocytes form 3D microtissues alone or in combination with endothelial cells

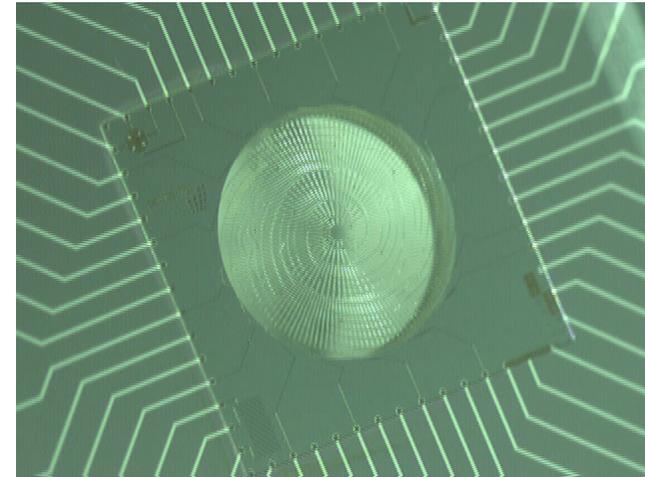


EXAMPLES OF CARDIOVASCULAR DISEASES MODELLED ON CHIP

- **CARDIAC DISEASE:**

- Heart-on-chip

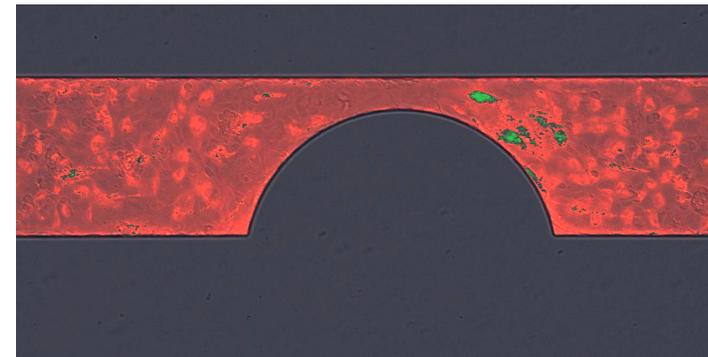
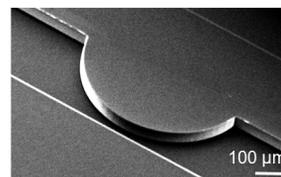
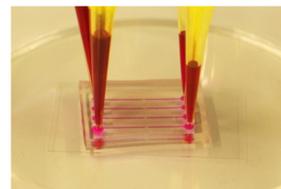
- Heart failure (genetic)
- Myocardial infarction (somatic: lack of oxygen)



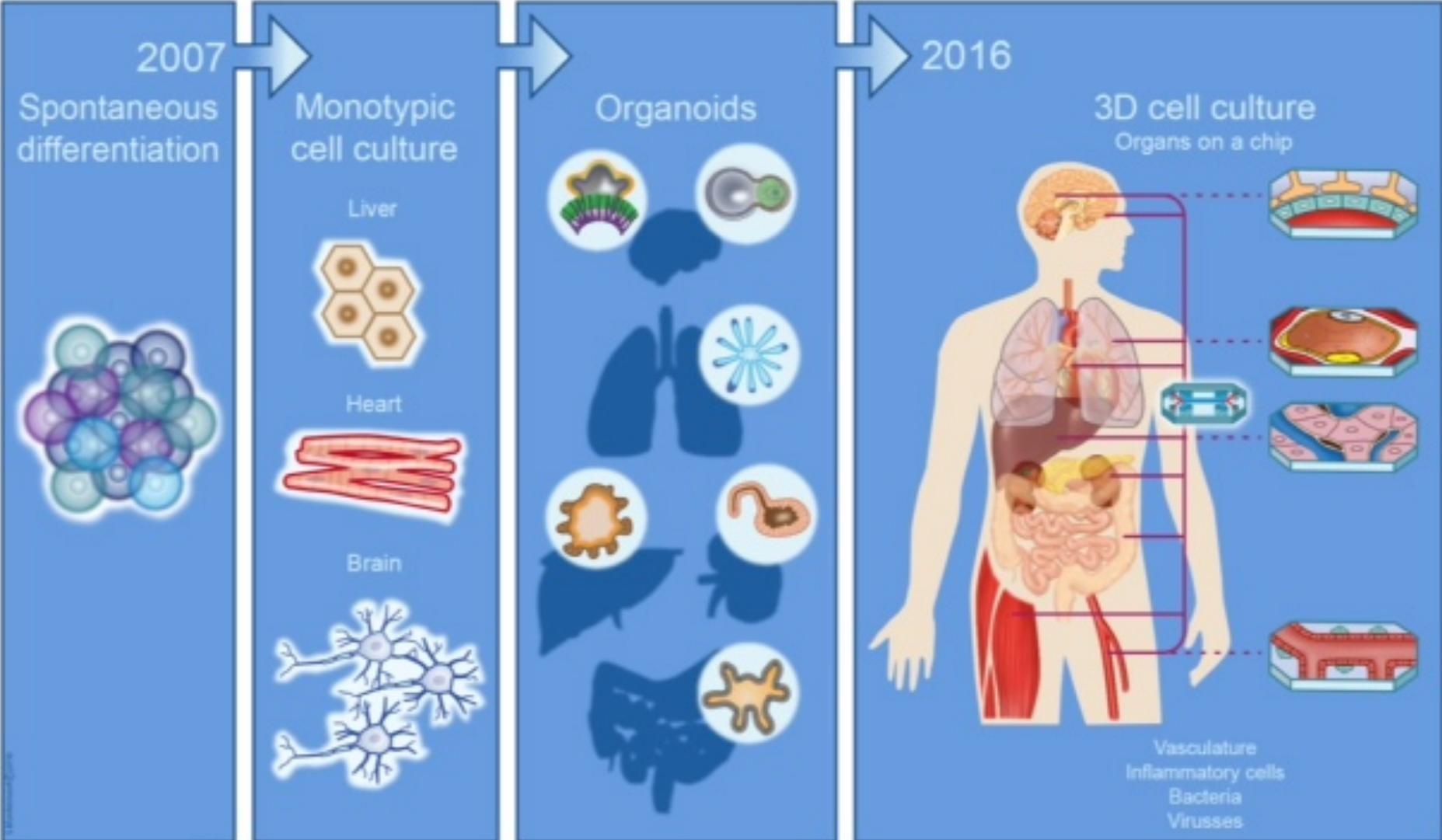
- **VASCULAR DISEASE:**

- Vessels-on-chip

- Atherosclerosis
- Thrombosis
- Blood Brain Barrier
- Vasculitis
- Vascular dementia



Evolution of hiPSC in disease modelling



Disease modelling

Neural diseases: Isogenic pairs of ALS hiPSC : drug repurposing

Intrinsic Membrane Hyperexcitability of Amyotrophic Lateral Sclerosis Patient-Derived Motor Neurons

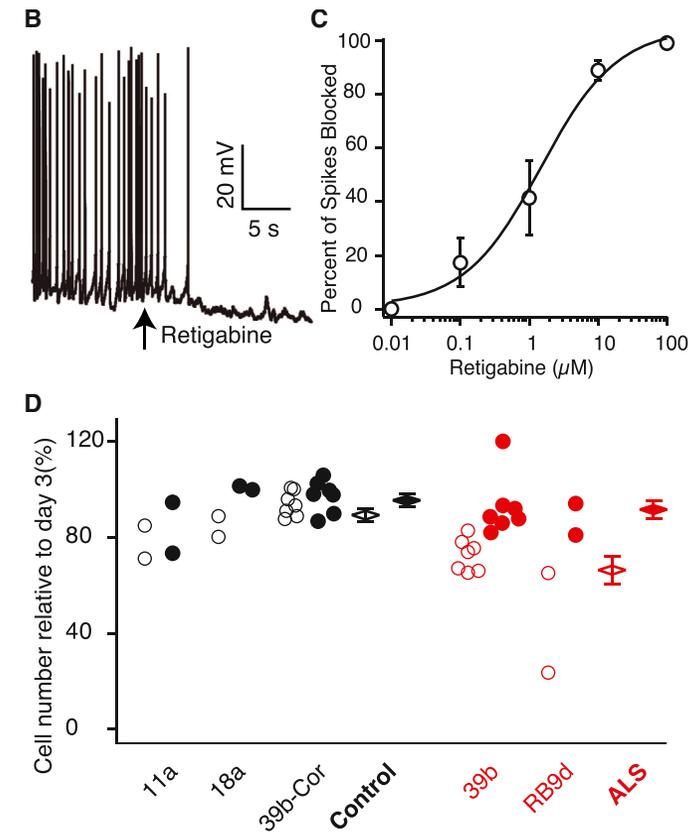
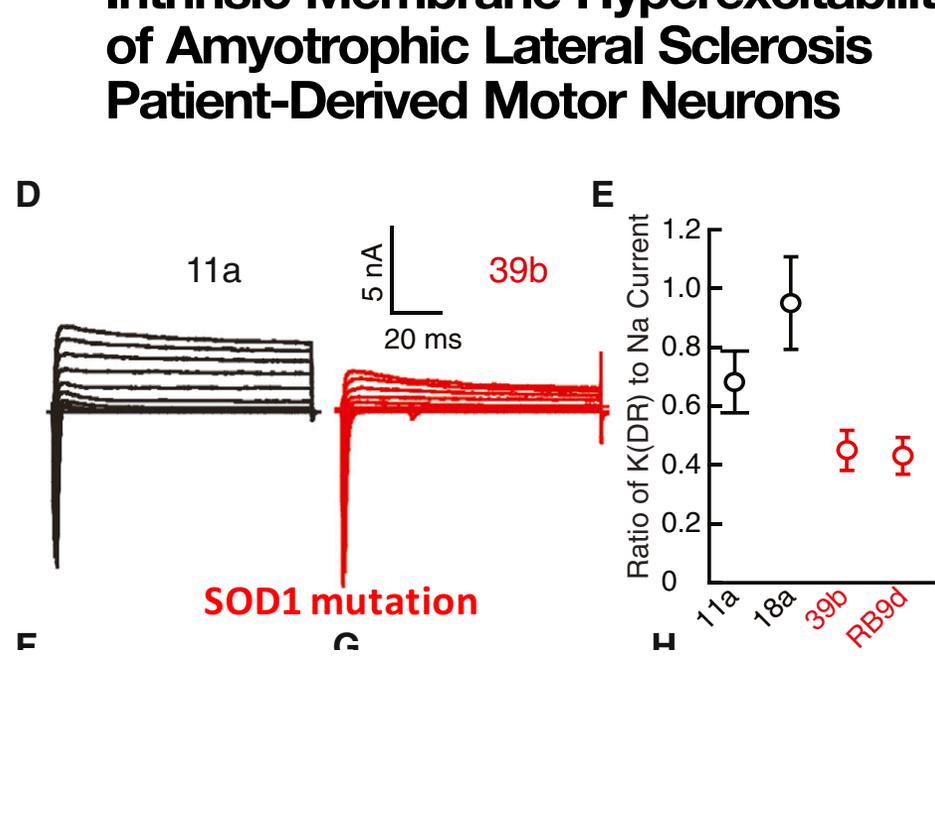


Figure 3. Retigabine Reduces Motor Neuron Excitability and Increases Survival

hPSC FOR ORGAN-ON-CHIP MODELS OF DISEASE, DRUG DISCOVERY AND SAFETY PHARAMCOLOGY

- isogenic hPSC lines with human disease mutations with functionally relevant phenotypes
- documented informed consent, genome sequence, donor medical history including drug responses
- “missing link” for GWAS through precision genetic engineering
- testing of drug combinations at different doses for effectiveness and toxicity in patients
- High-throughput bioassays including Organ-on-Chip and 3D formats to develop new drug treatments that delay or reverse symptoms of disease

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(per 15 Sept 2017 Ncardia)

Non-exec. board Galapagos



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