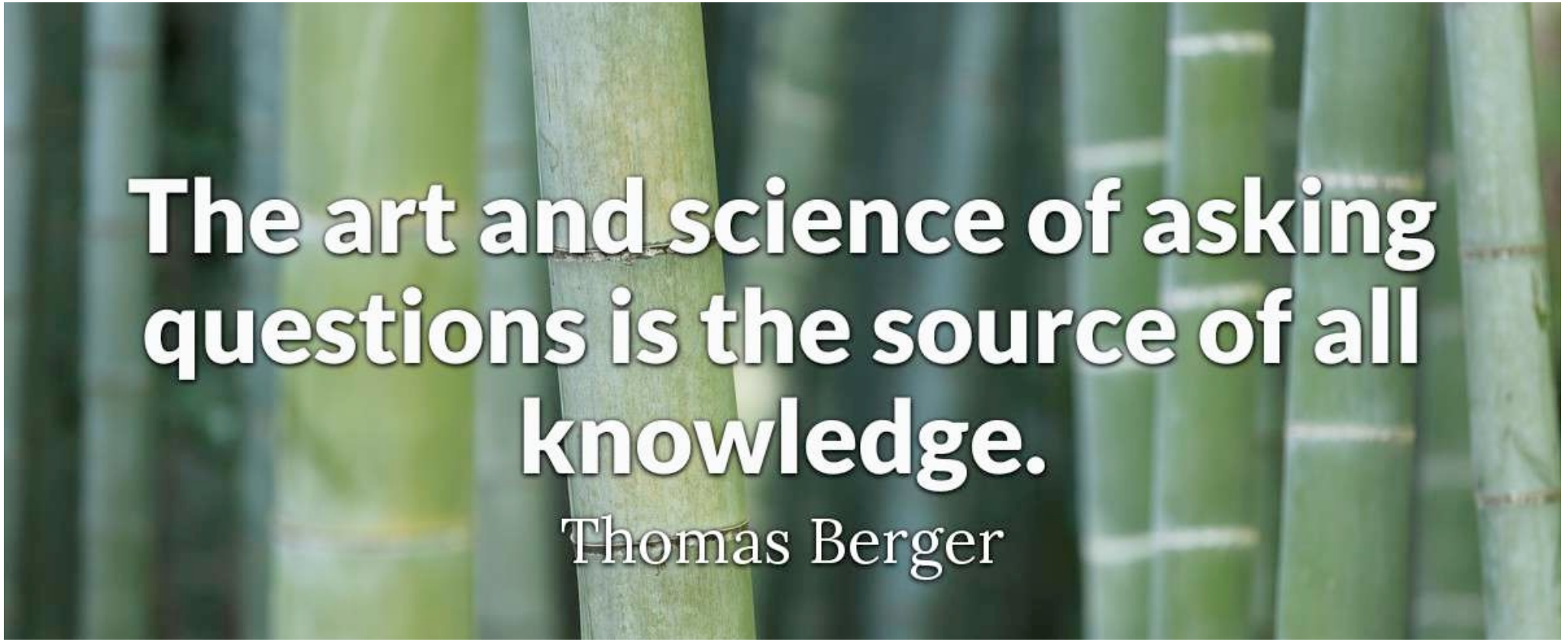

The art and science of asking questions is the source of all knowledge.
Thomas Berger

# CASE STUDIES

# Regenerative medicine

Perdita irreversibile di tessuti e cellule

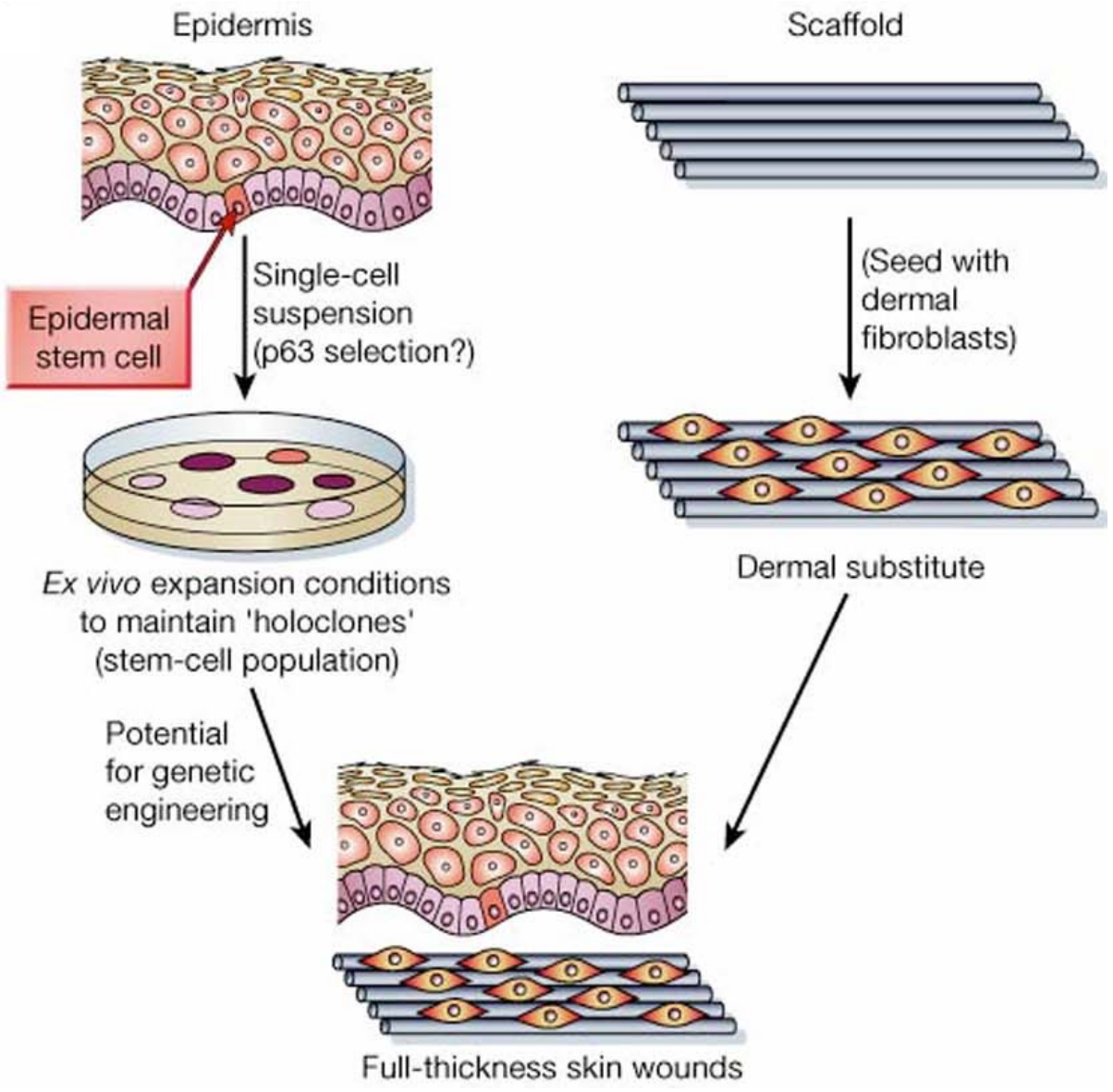
Infarto del miocardio
Ictus cerebrale
Diabete
m. Alzheimer

Anomalia irreversibile di tessuti e cellule

Malattie genetiche

# Regenerative medicine

1. *Ingegneria dei tessuti*
2. *Terapia cellulare*
3. *Terapia genica*




Bianco & Robey, 2001

Cell therapy of alpha-sarcoglycan null dystrophic mice through intra-arterial delivery of mesoangioblasts.

---

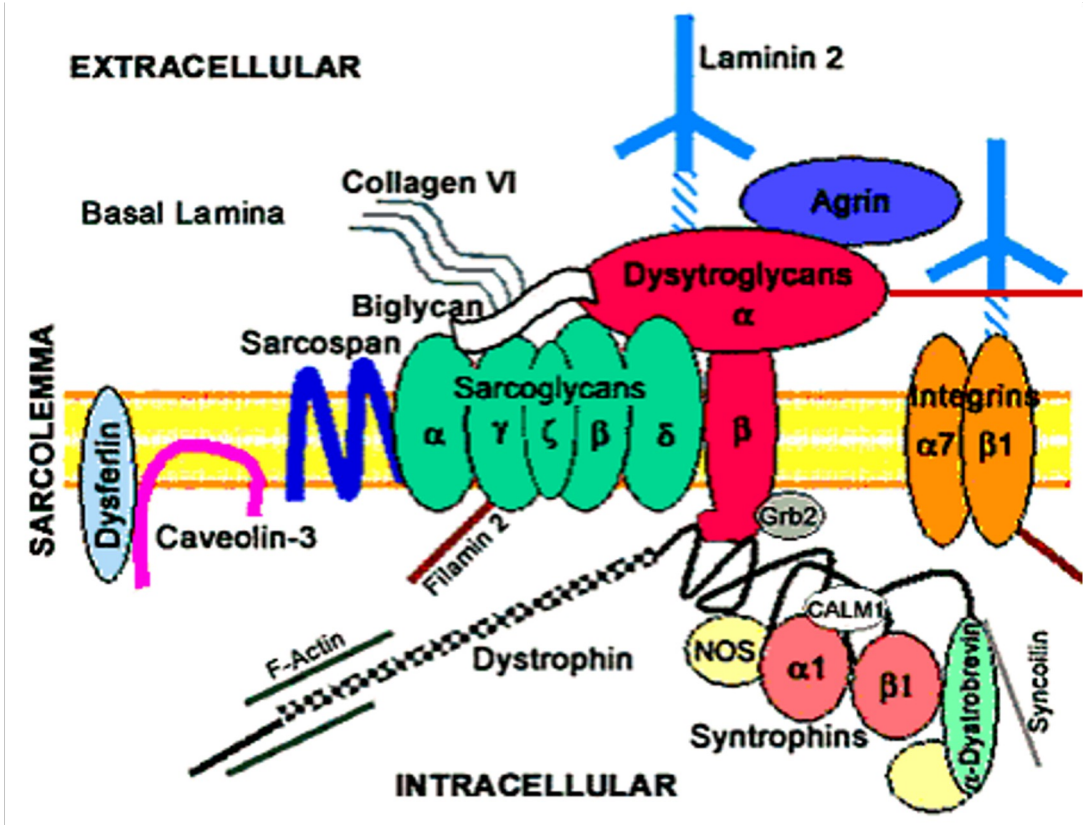
- knock out mice
- Cloned gene
- Genetically modified foetal cells

*Science 2003 Jul 25*

# Muscular dystrophies

- Genetic disease
- Multiple genes involved; most common involving the dystrophin glyocprotein complex (DGC)
- typical trait is fiber necrosis

# Dystrophin-glycoprotein complex




defective

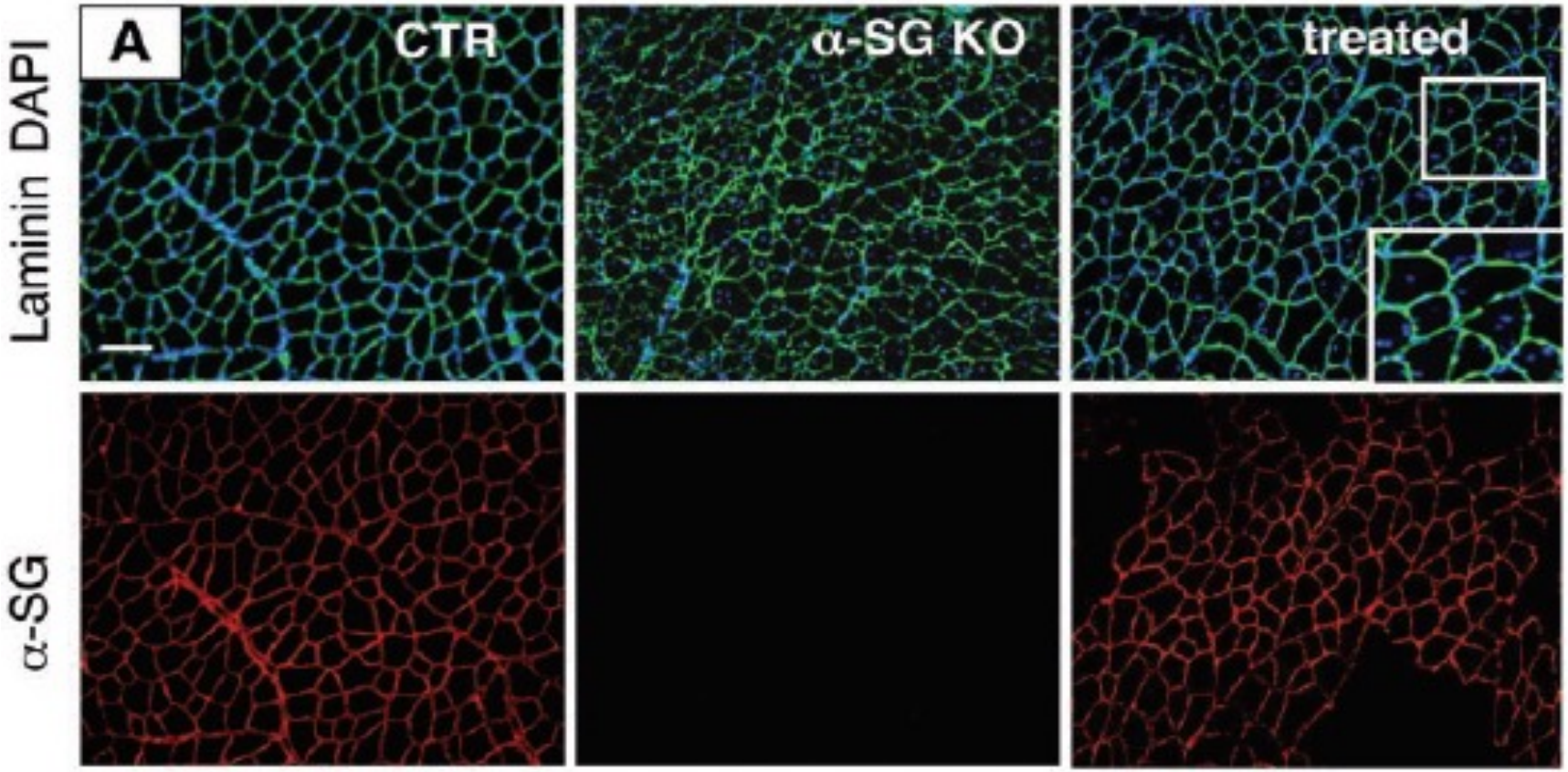
# Conventional therapy

| | |
|---|---|
| ➢ Stretching of muscles | ⇨ Does not cure |
| ➢ Corticosteroid treatment | ⇨ Side effectcs |
| ➢ Drugs to induce protein symthesis similar to the absent (utrophin) | ⇨ Mechanism validation to be assessed |

# Mesoangioblasts

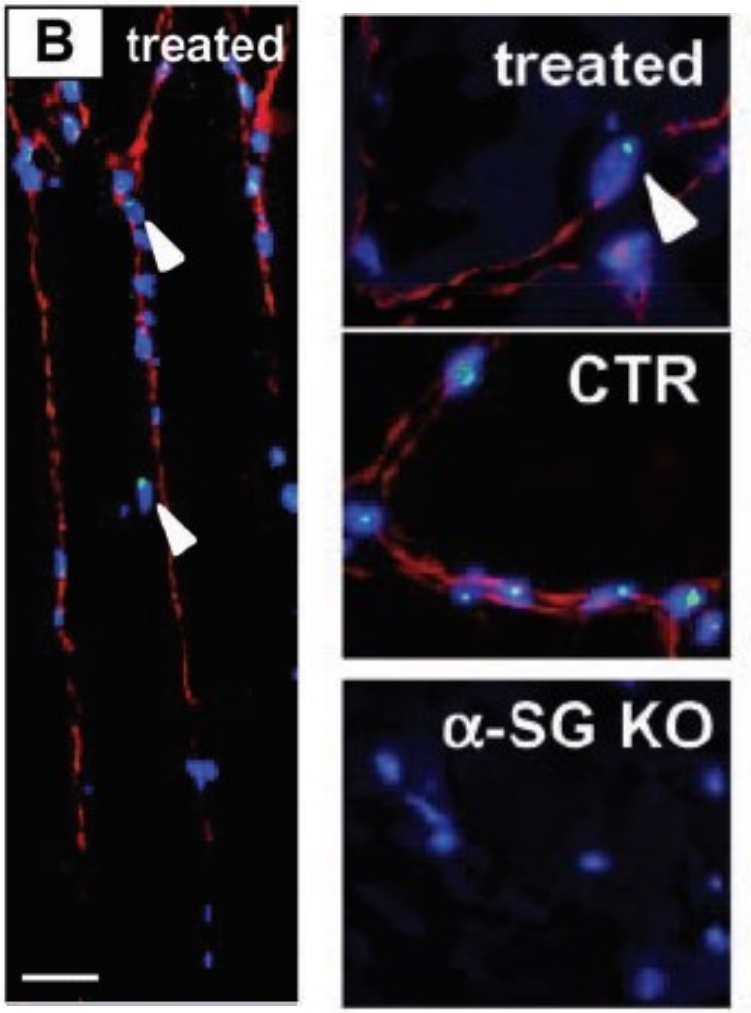
- Foetal stem cells associated with vascular system
- Highly proliferative
- Can move out of the vasculature in presence of inflammation
- Respond to mecrotic cytokines

alpha-sarcoglican expression after mesoangioblasts (10e5) **wt,** heterologous in female -/- mice. Analysis at 2 months.



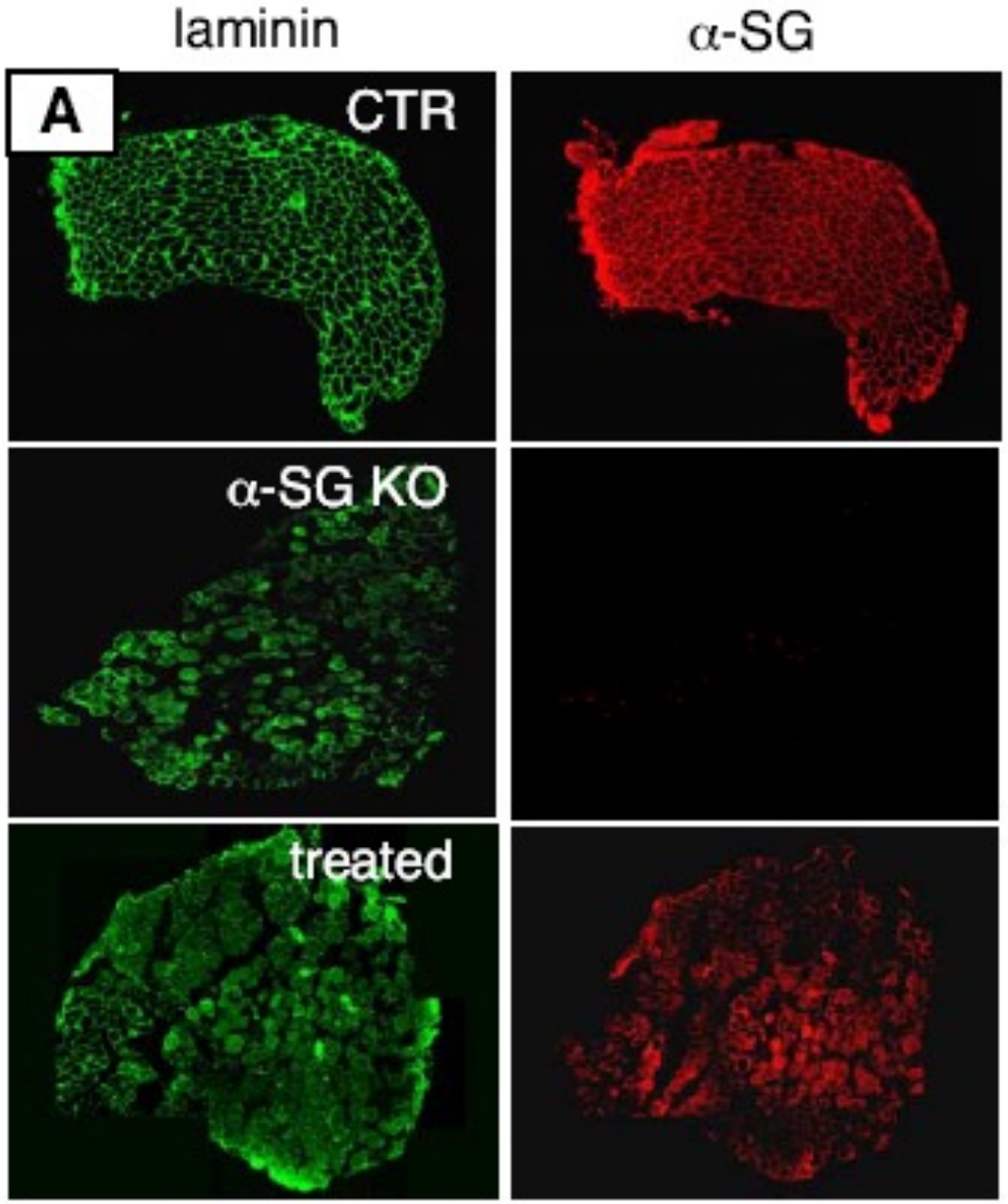

α-SG ab and tissue quality

# alpha-SG in mesoangioblasts



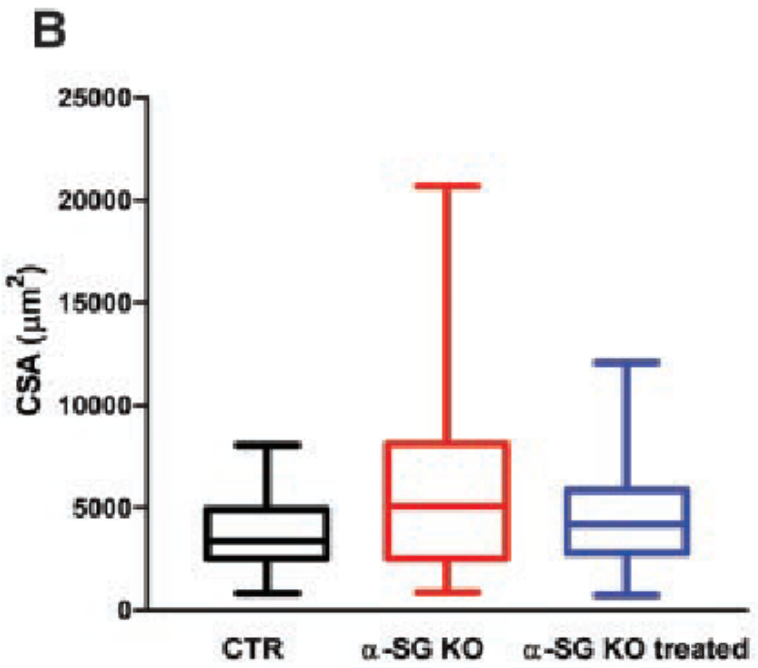

Fish (in blue) shows Y (arrow head), and α-SG positive (red)

3 injections at  40 days intervals; 5 x 10^5 mesoangioblasts male wt in female -/-. Assay 4 months after 1st injection




α-SG (in red), absent in diseases animals, is visible in treated mice

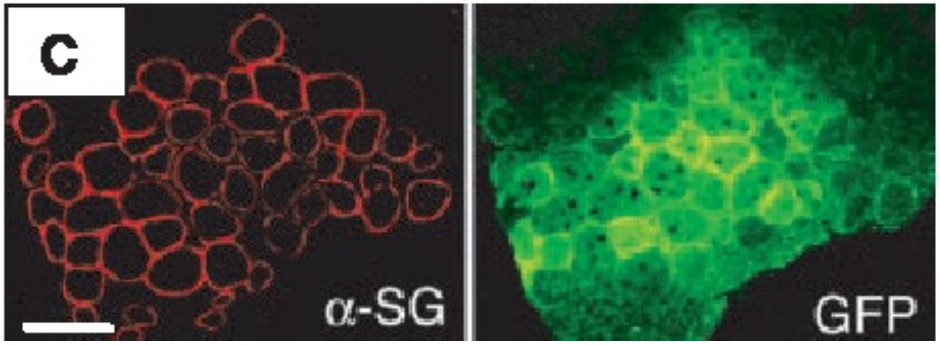
# Triple injection. Muscle functionality evaluated ex vivo

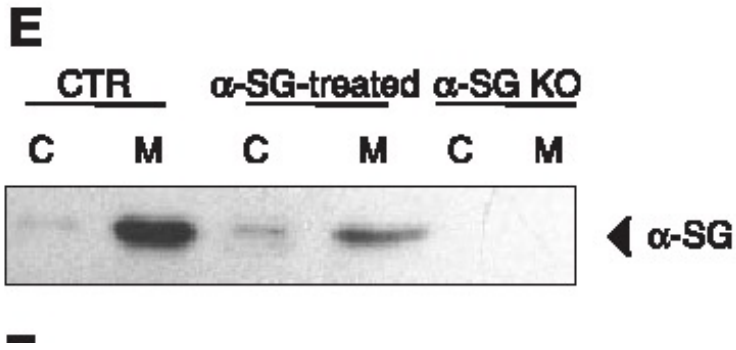



- Fibre section area

3 injections at 40 days intervals 5 x 10^5 autologous **mesoangioblasts**

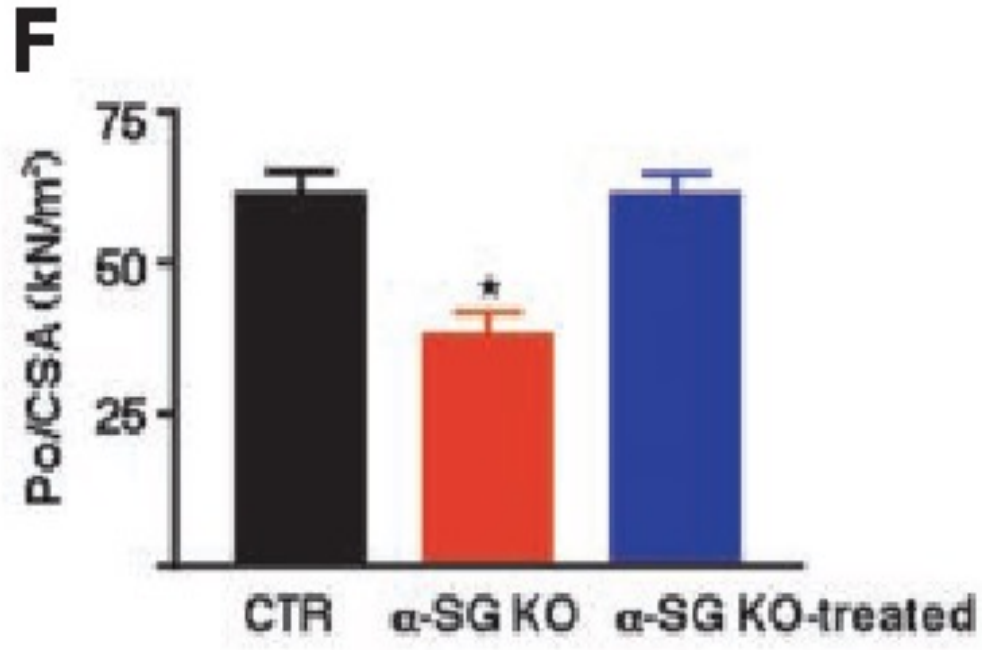
(from -/- mice aged **15d**) treated with lenti-PGK-SG-IRES-GFP




IF and WB show GFP and α-SG

# Assay of muscular force in treated mice

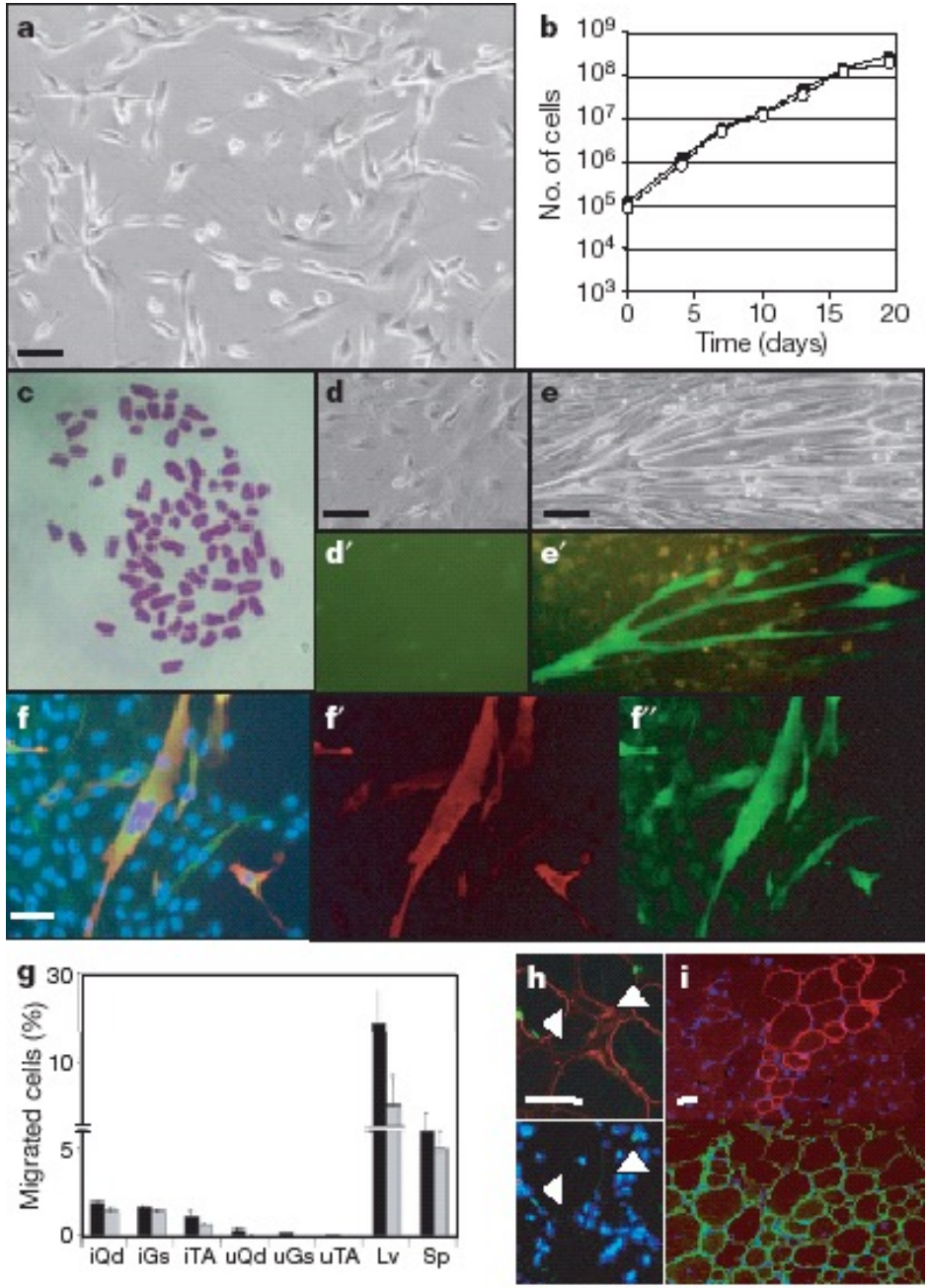


# Mesoangioblast stem cells ameliorate muscle function in dystrophic dogs

Maurilio Sampaolesi[1,2*], Stephane Blot[3*], Giuseppe D'Antona[2], Nicolas Granger[3], Rossana Tonlorenzi[1], Anna Innocenzi[1], Paolo Mognol[4], Jean-Laurent Thibaud[3], Beatriz G. Galvez[1], Ines Barthélémy[3], Laura Perani[1], Sara Mantero[4], Maria Guttinger[5], Orietta Pansarasa[2], Chiara Rinaldi[2], M. Gabriella Cusella De Angelis[2], Yvan Torrente[6], Claudio Bordignon[1], Roberto Bottinelli[2] & Giulio Cossu[1,5,7]

*Sanpaolesi Nature 2006*

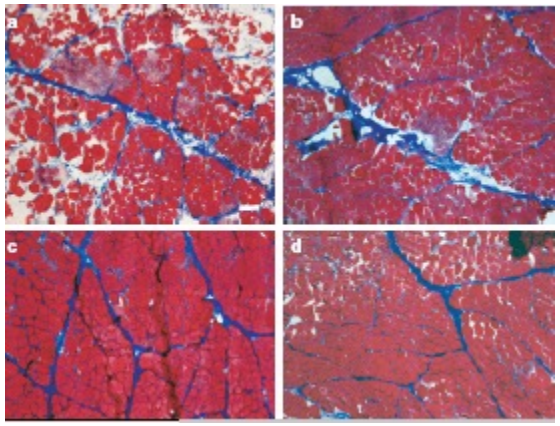
# Characterization of dog mesoangioblasts in vitro and in mice



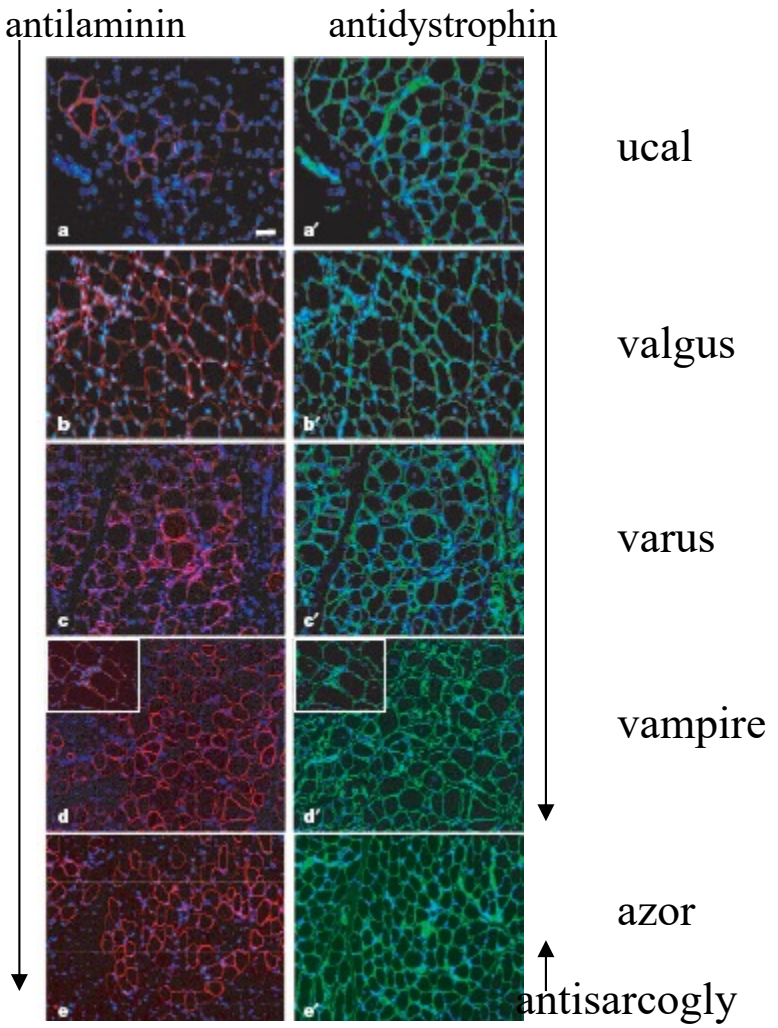

Dog mesangio
A-morphology
B-proliferation
C-karyotype
D-F transduction microdystro an GFP lenti
G-migration into skeletal muscle
H-histology in scid-mdx mice (laminin, dystro)

*Sanpaolesi Nature 2006*

# Dogs (duchenne model) after intraartherial delivery of heterologous wt mesangioblasts

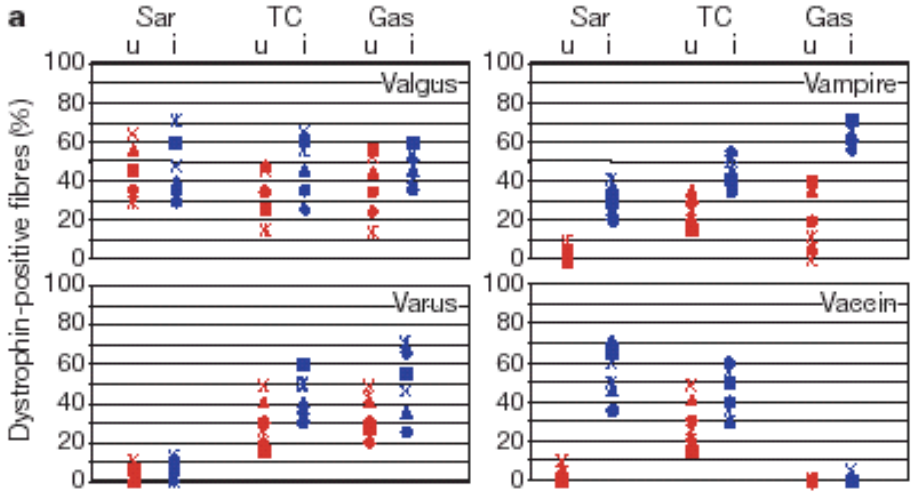


Histo (d-cured; a-c variable ill phehotype)



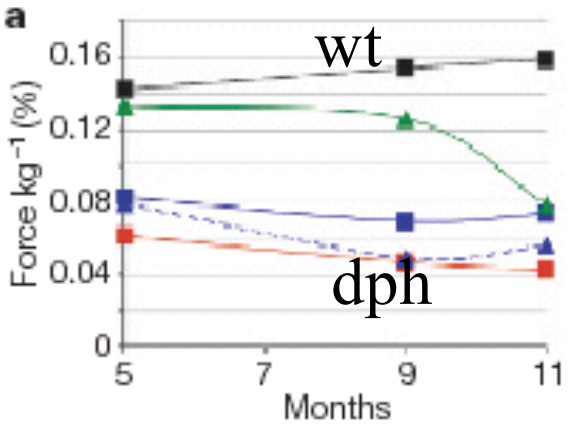

Immune histo on muscle

# Quantitative analysis of dystrophin content in tissue from treated dogs

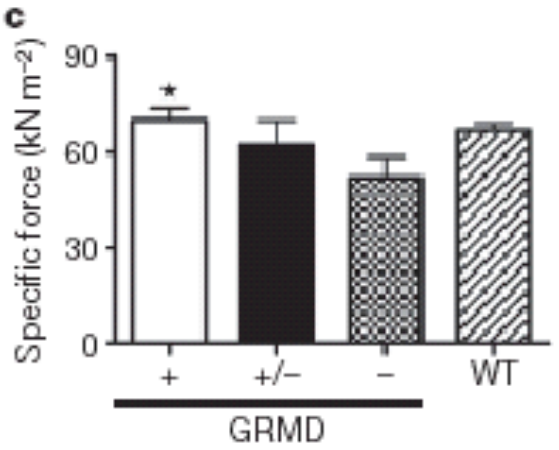


# Physiology of treated dogs

Tetanic force




Fiber counting



http://www.telethon.it/comunicazione/cossu/Cossu.MP3