Tissue engineering applied to muscle regeneration

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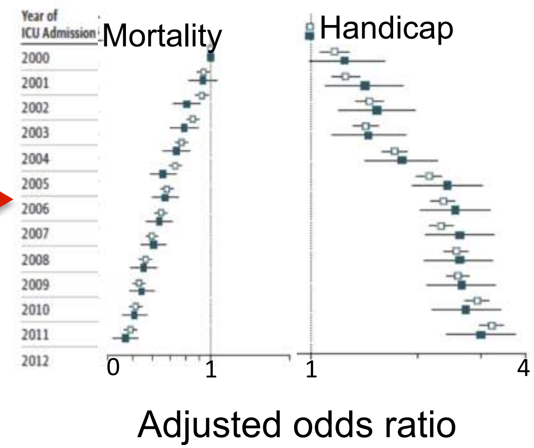
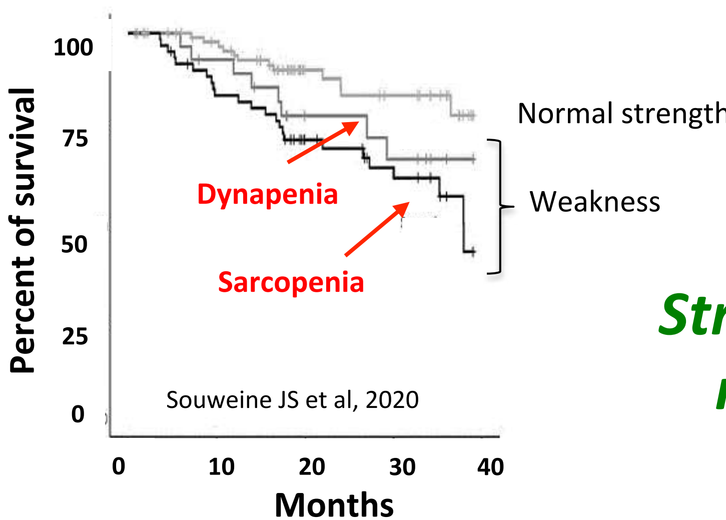
Example of a General Research Strategy which will need Skeletal Muscle Tissue Engineering

Critically ill patients:
Organ(s) dysfunction
➔Functional supplementation

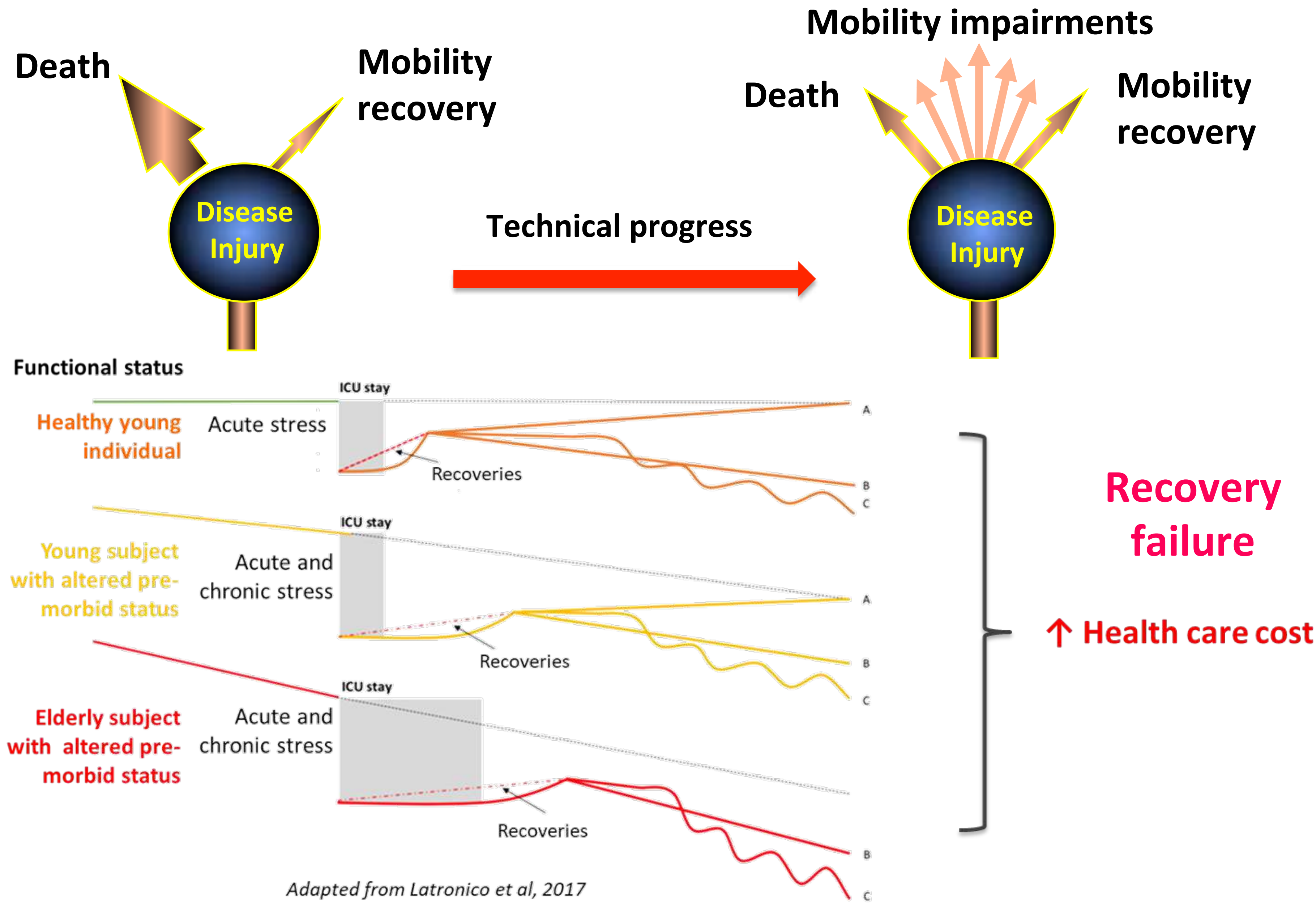
Acquired myopathy
(Weakness, motor disability, handicap..)





Strength and skeletal muscle mass as a survival factor

Example of a General Research Strategy which will need Skeletal Muscle Tissue Engineering



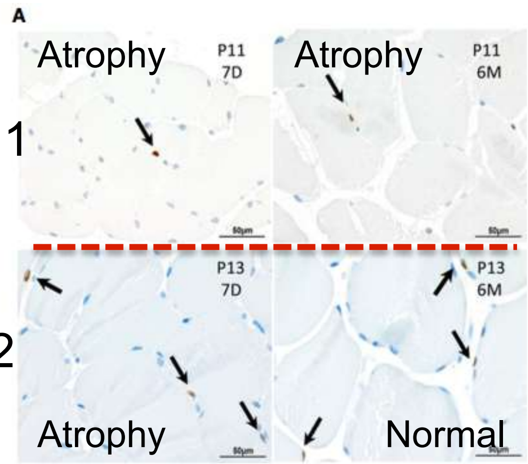
Adapted from Latronico et al, 2017

Example of a General Research Strategy which will need Skeletal Muscle Tissue Engineering

7 Days **6 Month**

Patient 1

Patient 2



Loss of muscle stem cell pool
Disability
Handicap

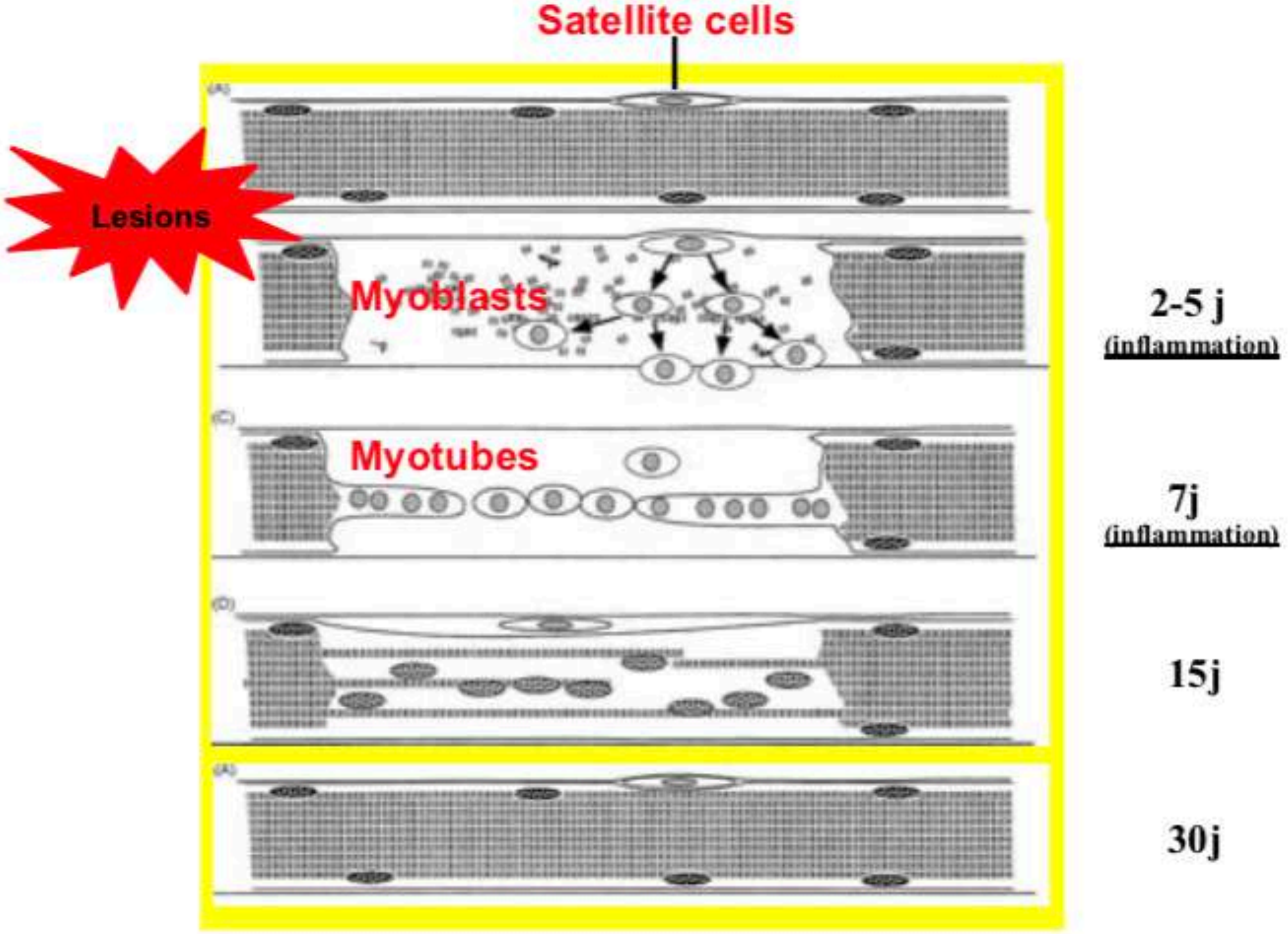
récupération

Dos Santos AJRCCM 2017

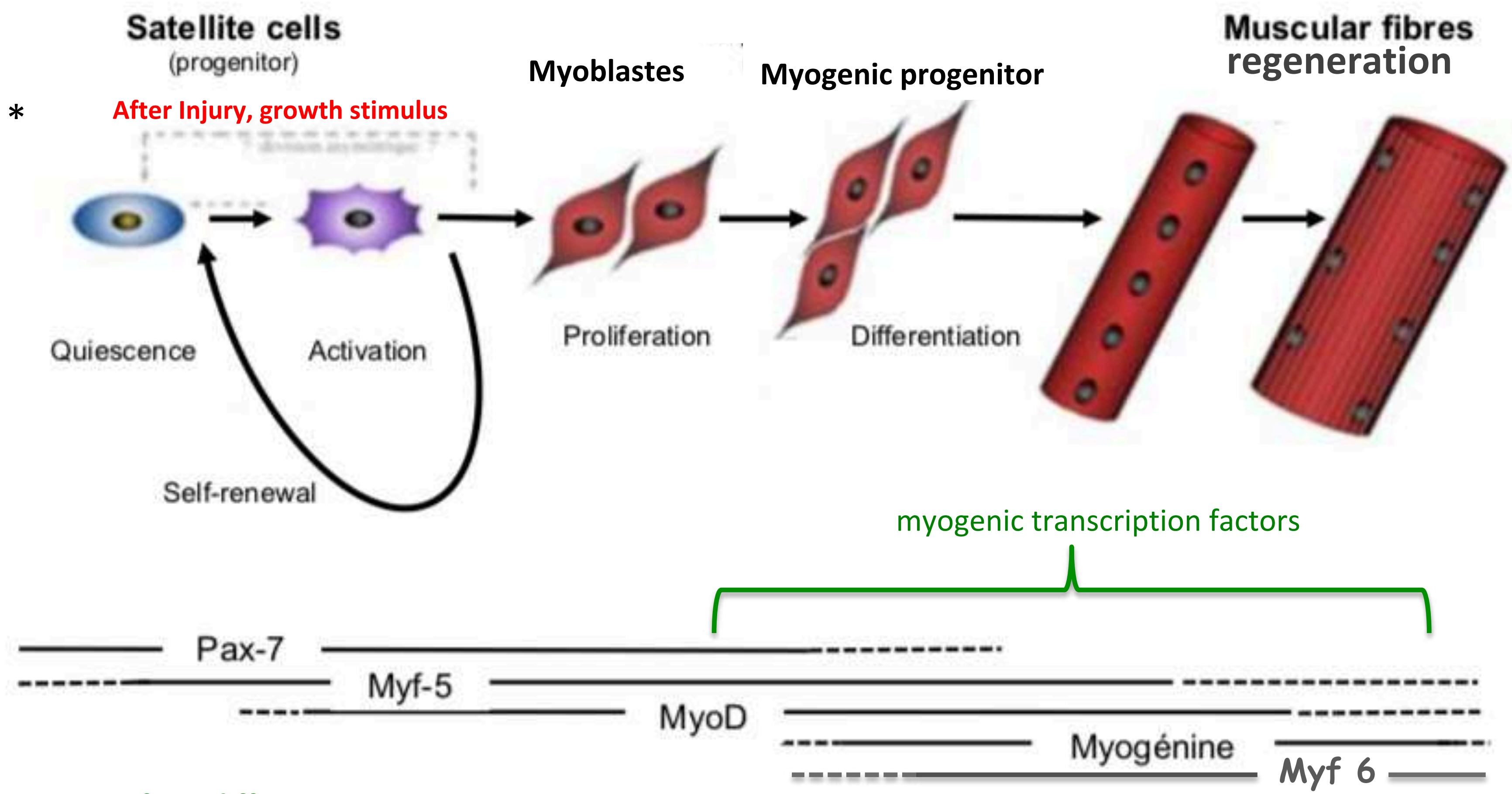
Two chief questions

1. What are the mechanisms underlying this heterogeneity?
2. How to restore mobility?

Satellite cell and muscle regeneration in vivo



Cell fate: A cascade of transcription factors controls regeneration



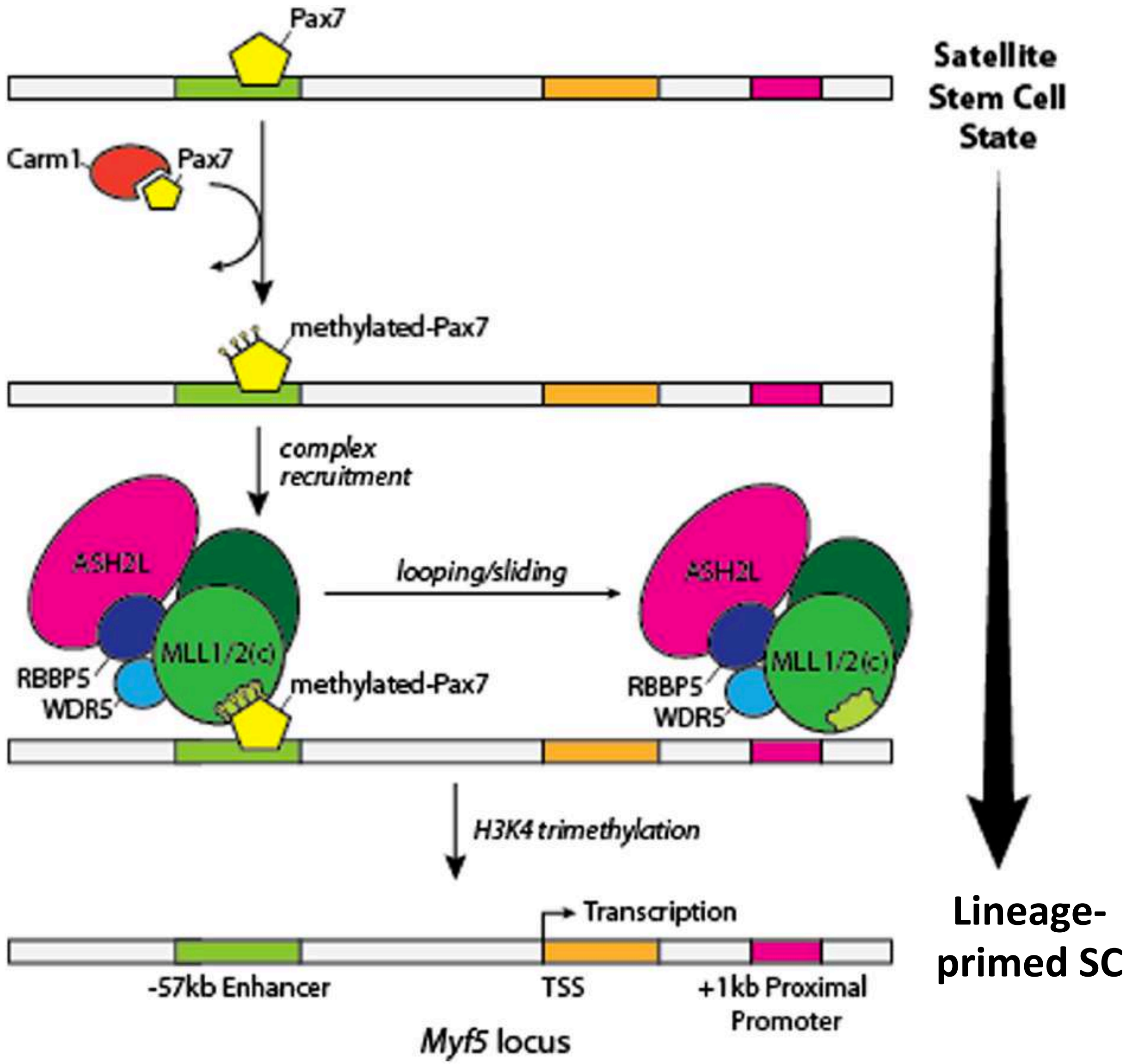
Different stages of SCs differentiation

Pax7+ Myf 5 - MyoD– (quiescence/sef-renewal)
Pax7+ Myf 5 + MyoD– (activation)
Pax7+ Myf 5 + MyoD+ (proliferation)
Pax7–MyoD+MyoG+ (differentiation)

Pax: Protein paired Box
MyoD: myogenic differentiation 1
Myf: Myogenic factor

Arginine méthyltransférase

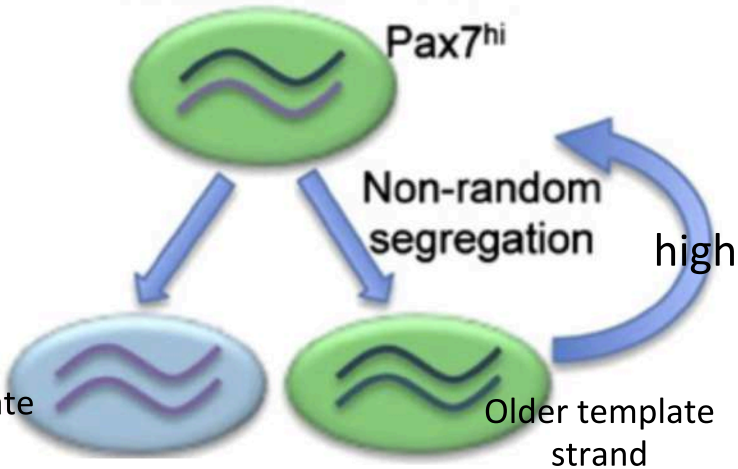
histone méthyltransférase complex



Cell Stem Cell 11, 333–345, September 7, 2012

Cell fate: A cascade of transcription factors controls regeneration

Génération de progéniteurs myogéniques et le maintien du pool de cellules souches



inheritance

Cam 1

Numb (Notch inhibitor)

MyoD

Pax7+Myf5+ Pax7+Myf5-

Genom protected

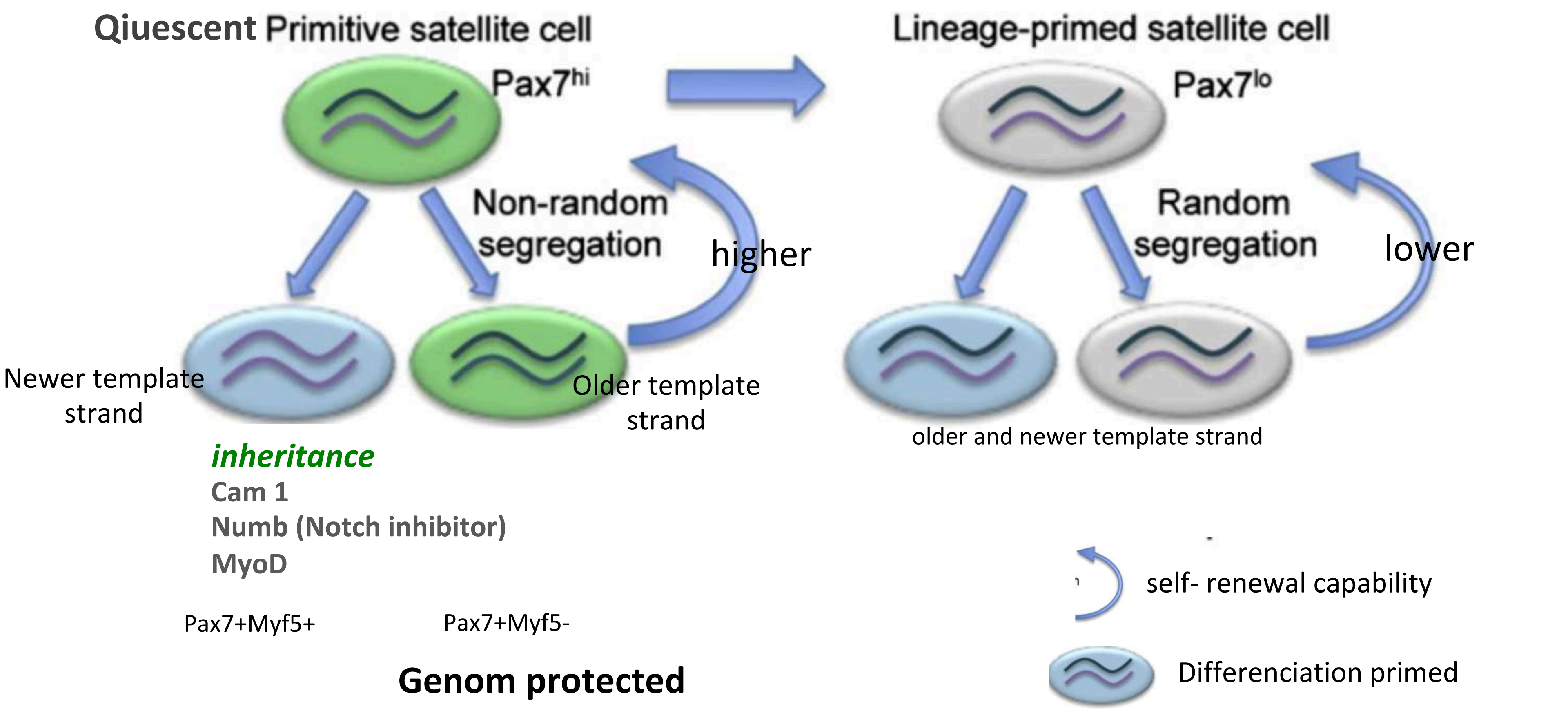

self- renewal capability


Differenciation primed

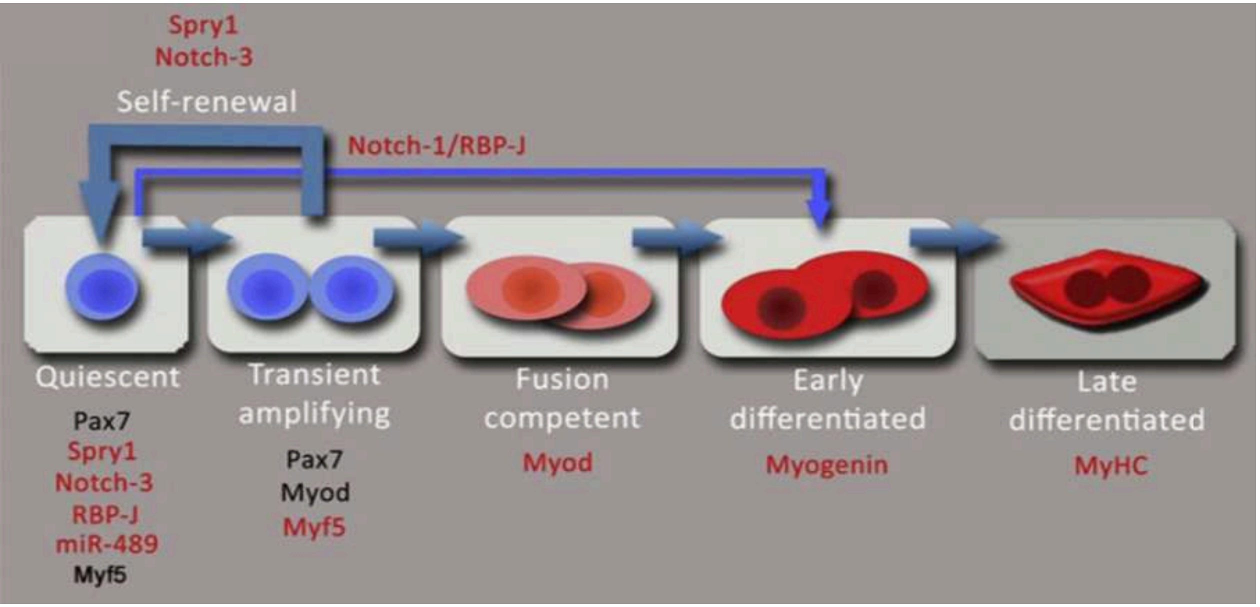
Cell fate: A cascade of transcription factors controls regeneration

Génération de progéniteurs myogéniques et le maintien du pool de cellules souches

Expansion du pool de cellules souches satellites



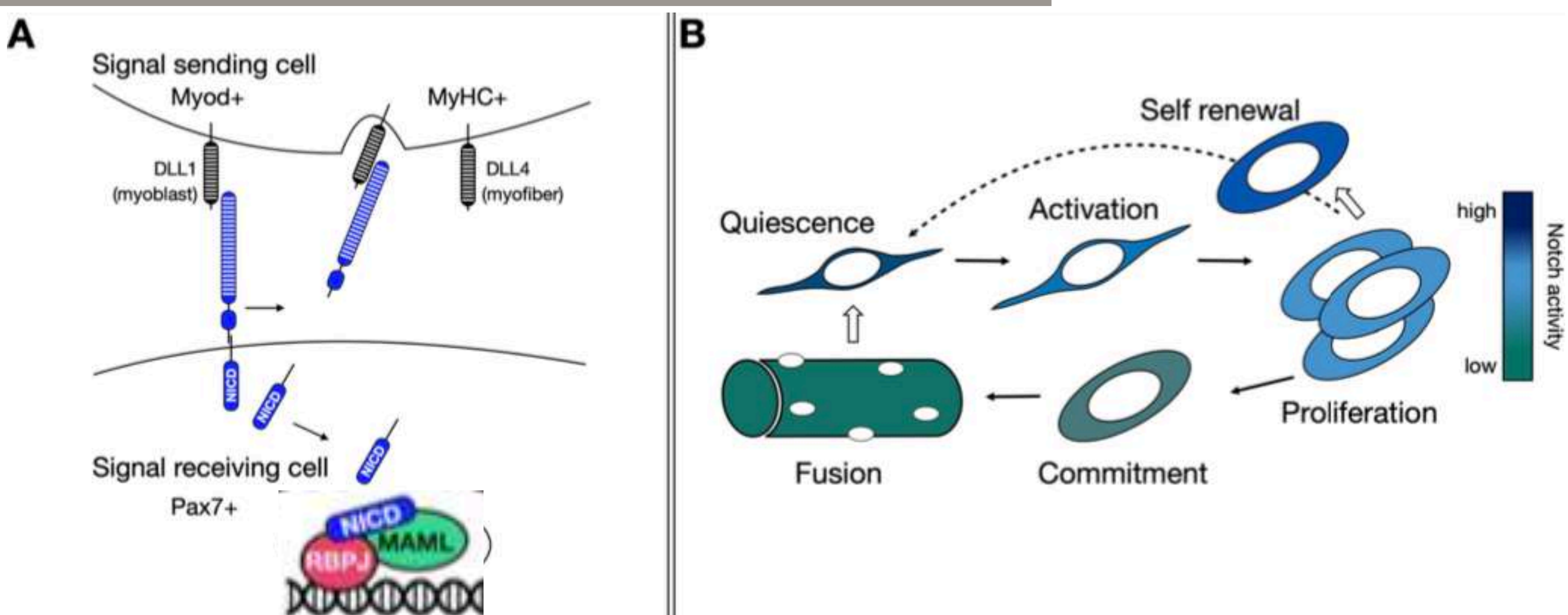
The Notch signaling pathway during myogenic progression and self-renewal



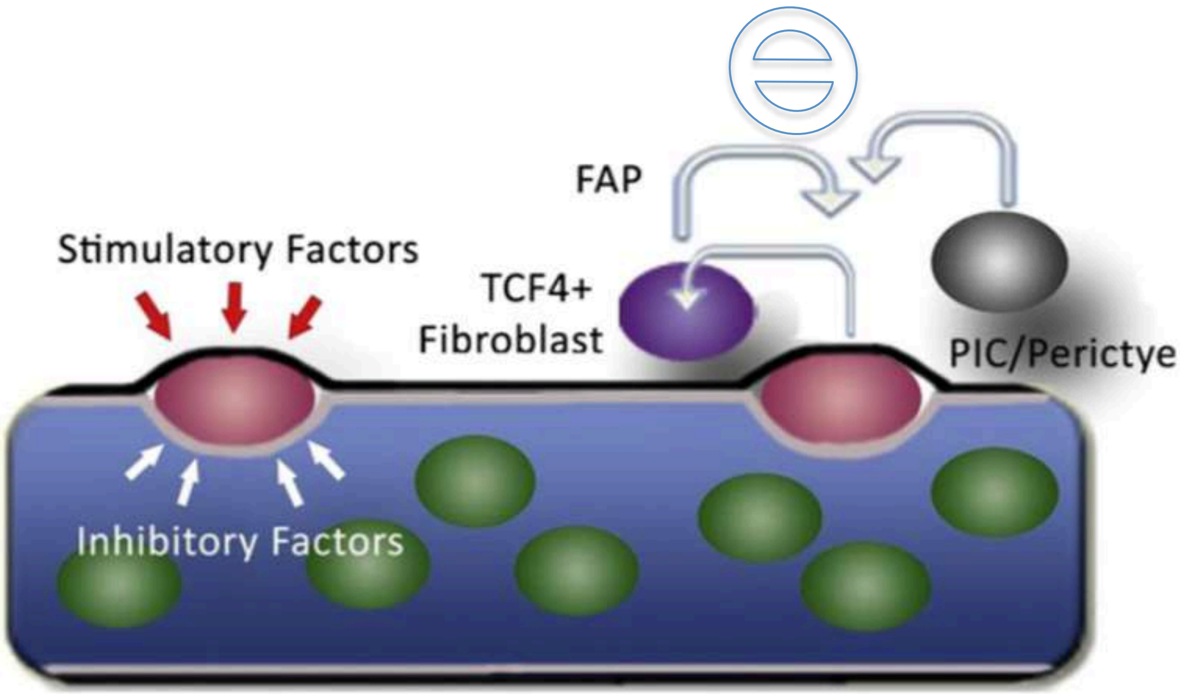
RBP-J : Recombination Signal Binding Protein For Immunoglobulin Kappa J Region

DLL1: Familles de ligands Notch
NIC: domaine intracellulaire de Notch.

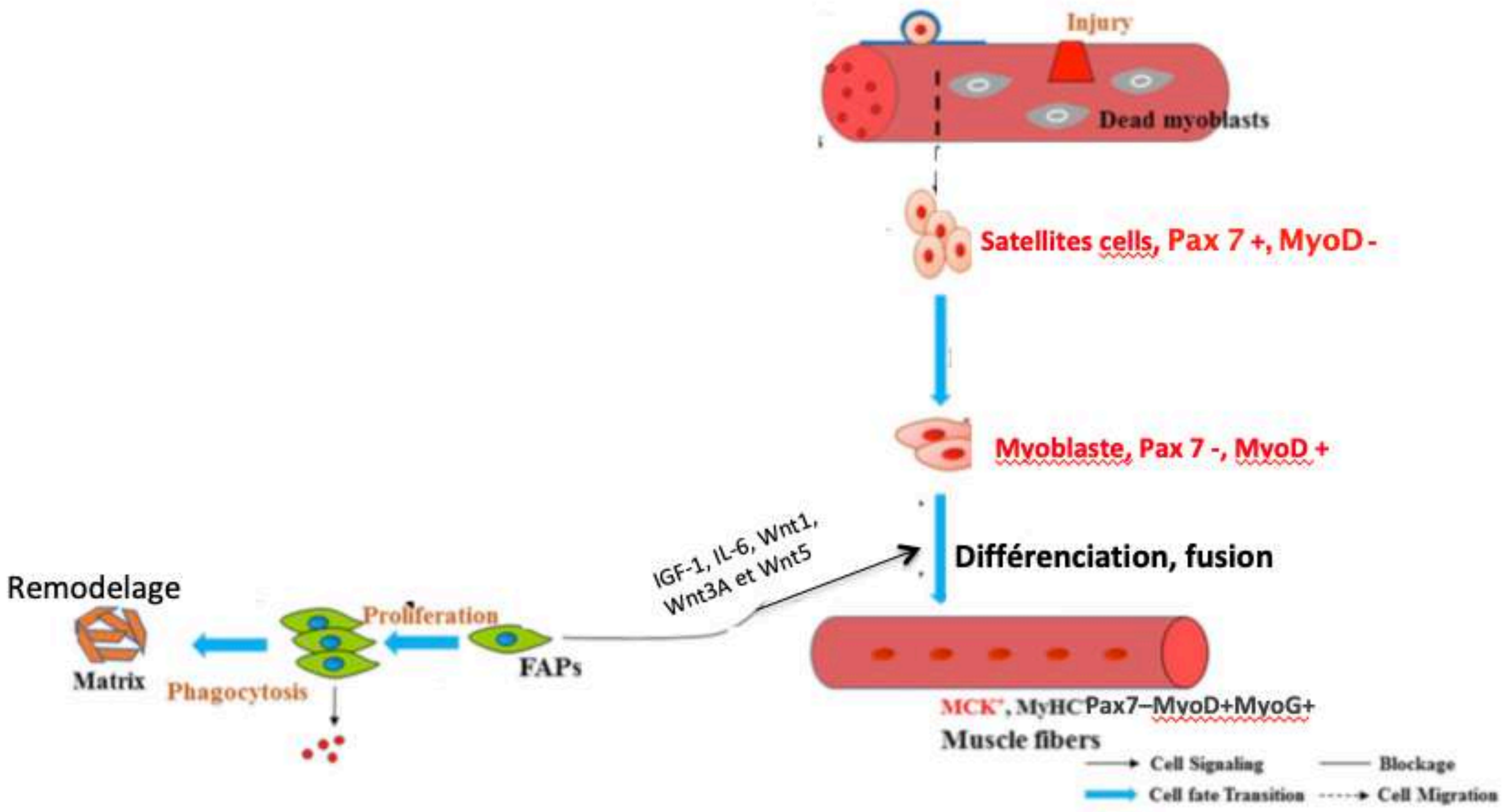
Spyr1: an inhibitor of growth factor signaling



A cascade of transcription factors controls regeneration

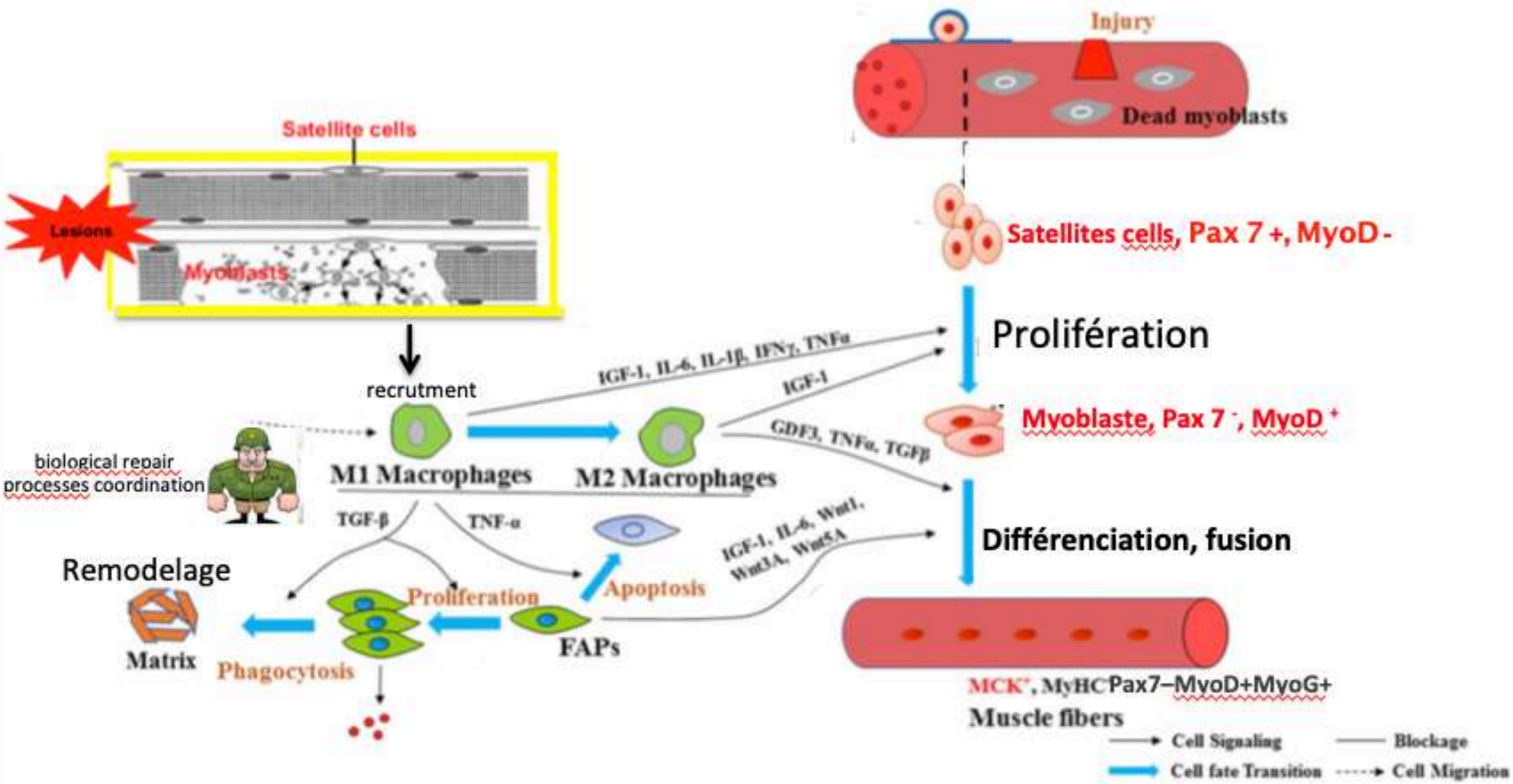


Cell microenvironment and muscle regeneration process



Journal of Muscle Research and Cell Motility (2019) 40:1–8

Cell microenvironment and muscle regeneration process



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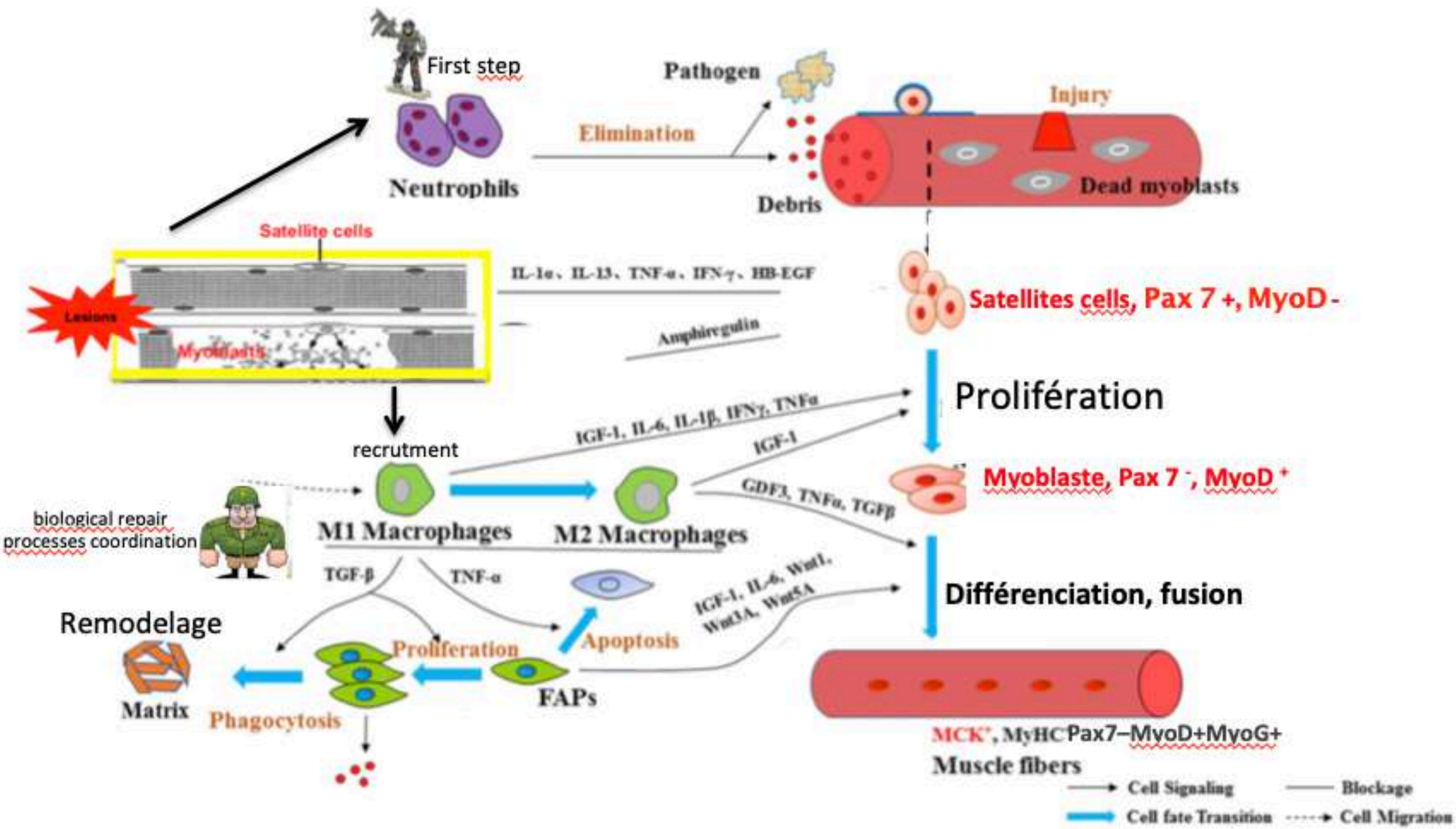

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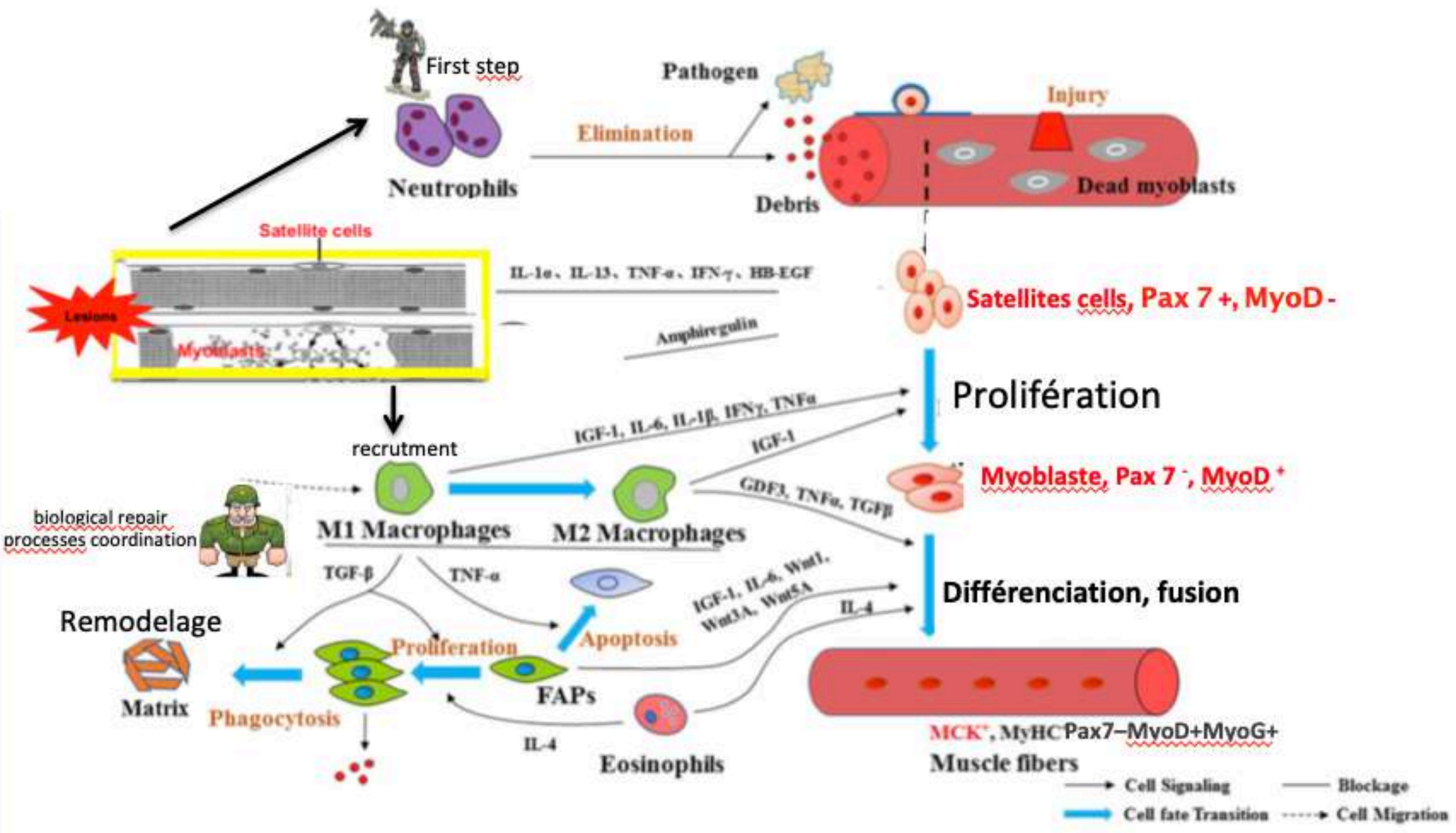

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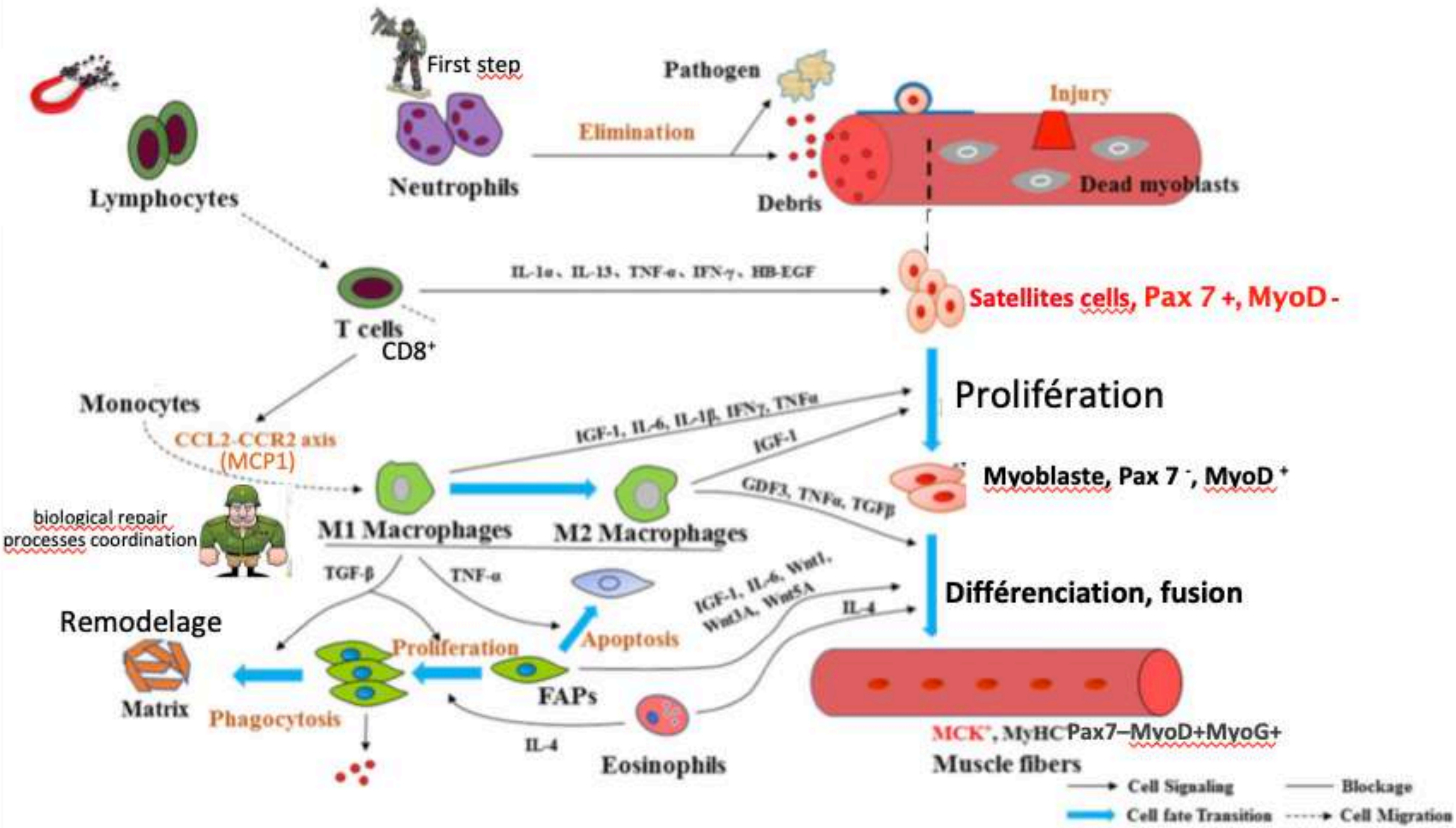

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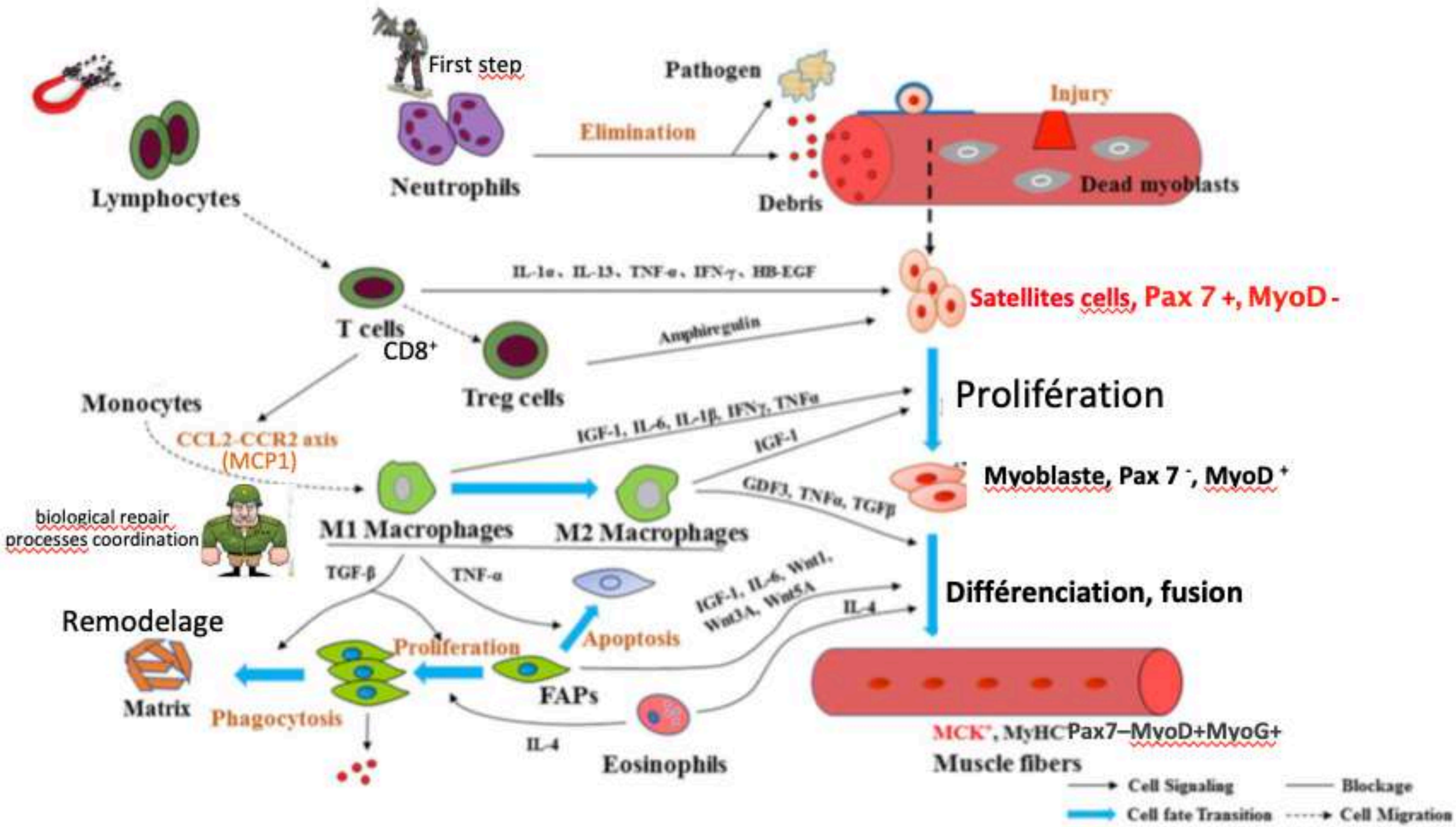

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Cell microenvironment and muscle regeneration process

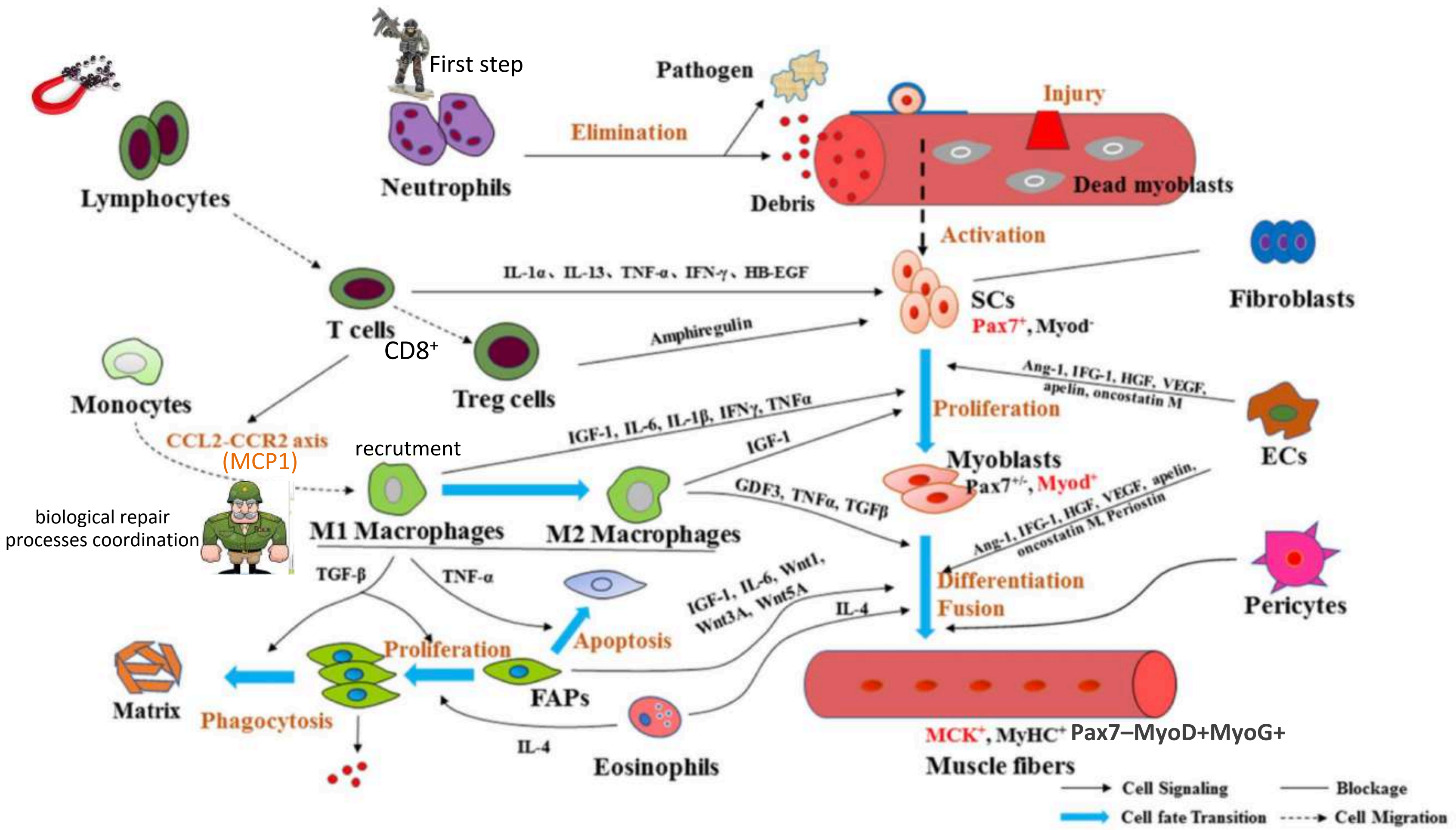

Fig. 1 Multiple *cell* types contribute to skeletal muscle regeneration. *SCs* satellite cells, *FAPs* fibro-adipogenic precursor cells, *ECs* endothelial cells

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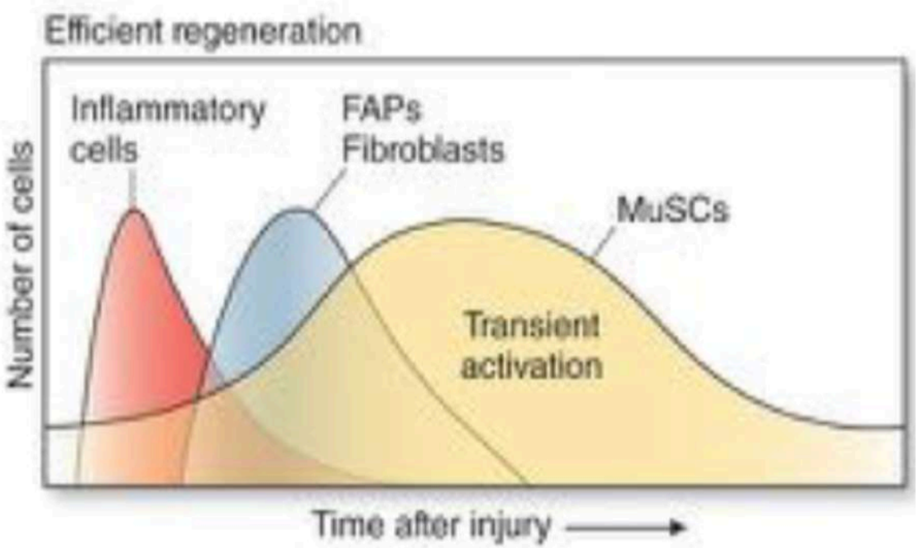
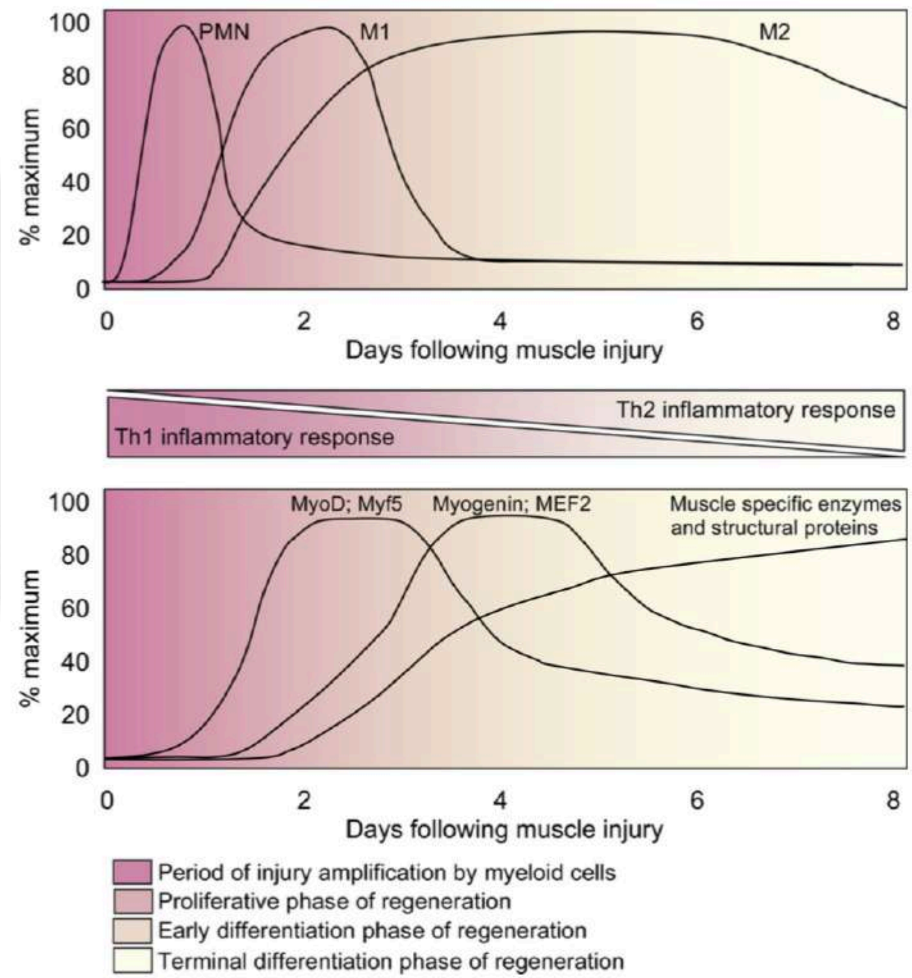
Cell microenvironment and muscle regeneration





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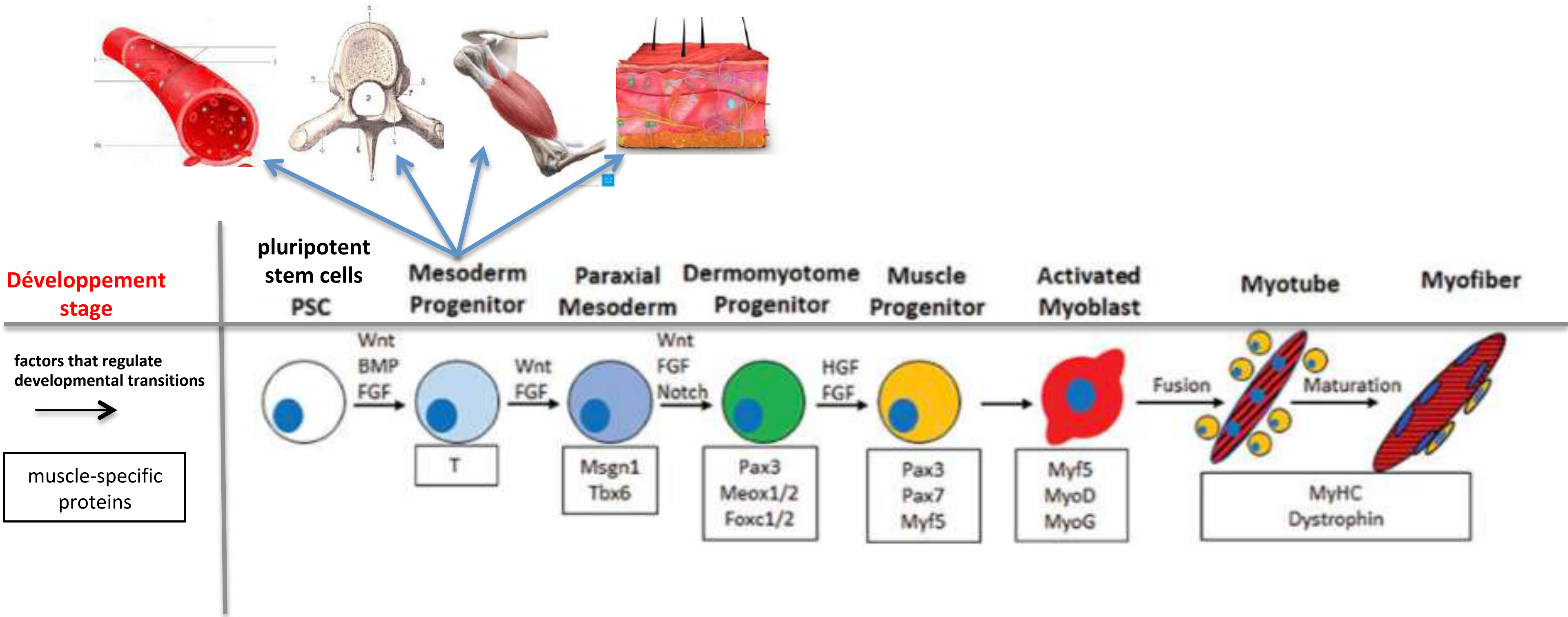
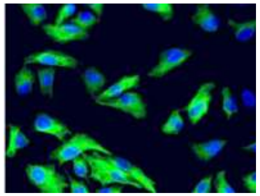
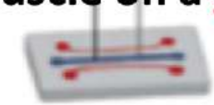


FGF: Fibroblast growth factor
BMP: Bone morphogenetic protein
TBx6 : T-Box Transcription Factor 6
Msgn1 : transcription factor mesogenin 1
Pax : Paired box protein
MEOX1: Mesenchyme Homeobox 1
FOX : FOX (forkhead box)

Why satellite cells become rapidly activated and lose engraft-ment ability following in vitro culture ?



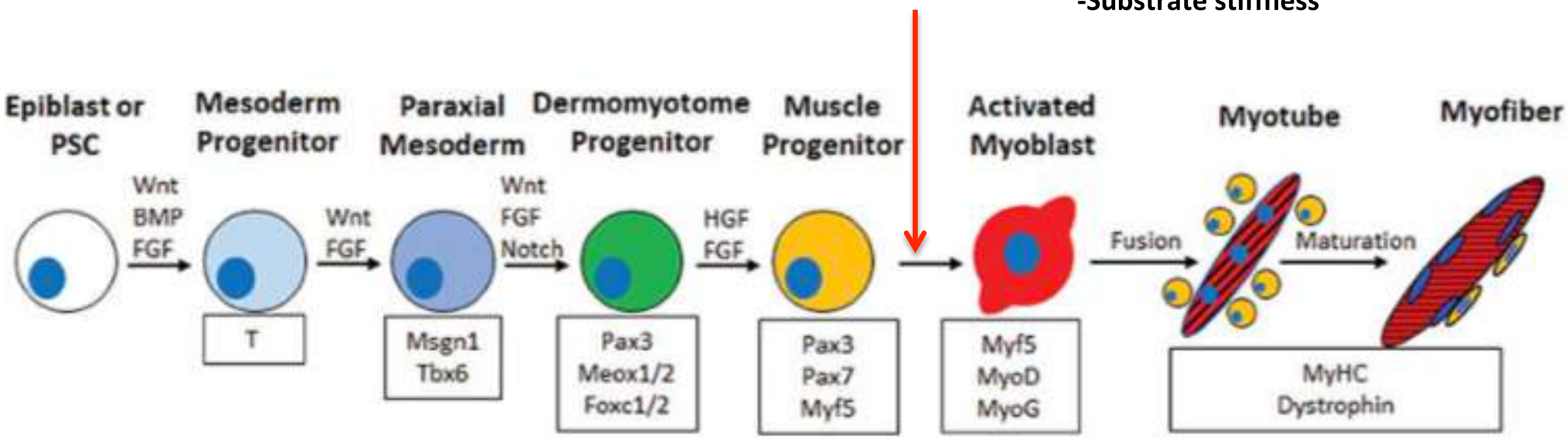
Muscle on a ship



Difficulties studies of skeletal muscle in vitro

-p38 MAPK activation (mitose)
-Metabolism
Glycolysis: ↗SC activation and MyoD ↗

-Substrate stiffness

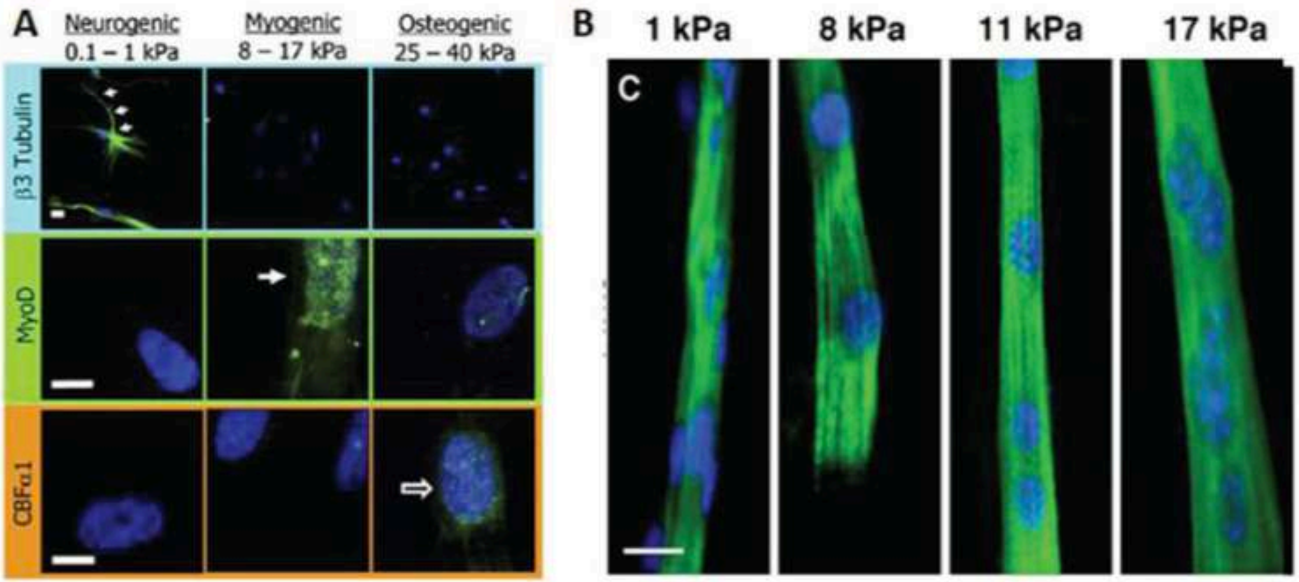


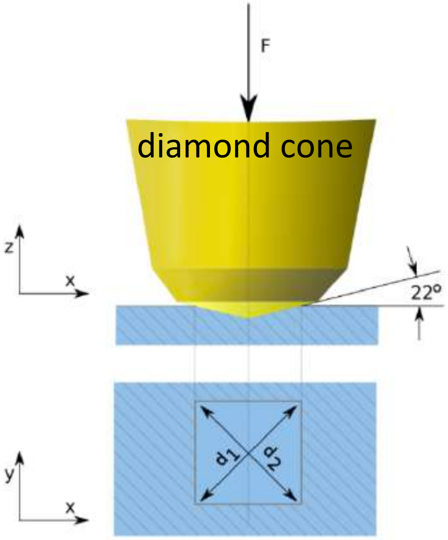
Pax 7 + Myf 5 + MyoD-

A subset of muscle progenitors does not differentiate but resides on the outside of the myofiber to contribute to future muscle regeneration events

Cell microenvironment and muscle regeneration

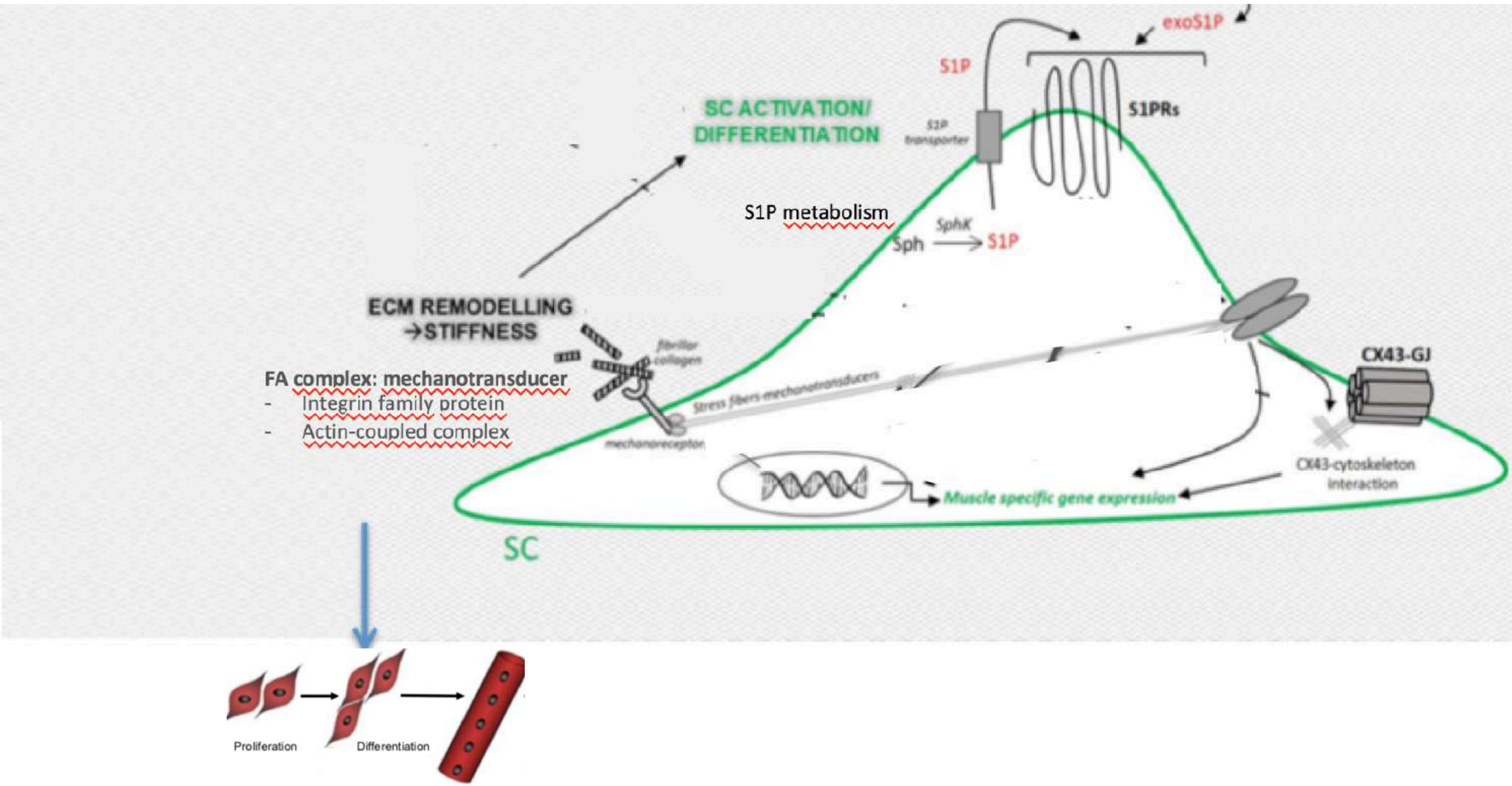
Substrate stiffness



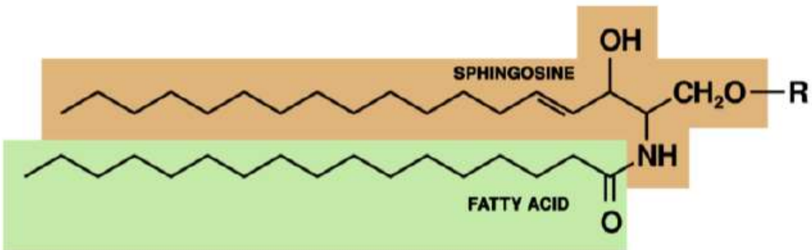


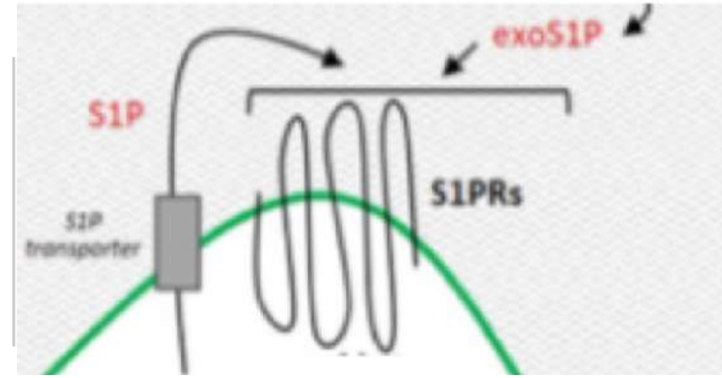
Cbfa1: Core-binding factor alpha: Cbfa1

Mechanotransduction and cell fate



Sphingosine 1-Phosphate (S1P)/ S1P Receptor Signaling and Mechanotransduction

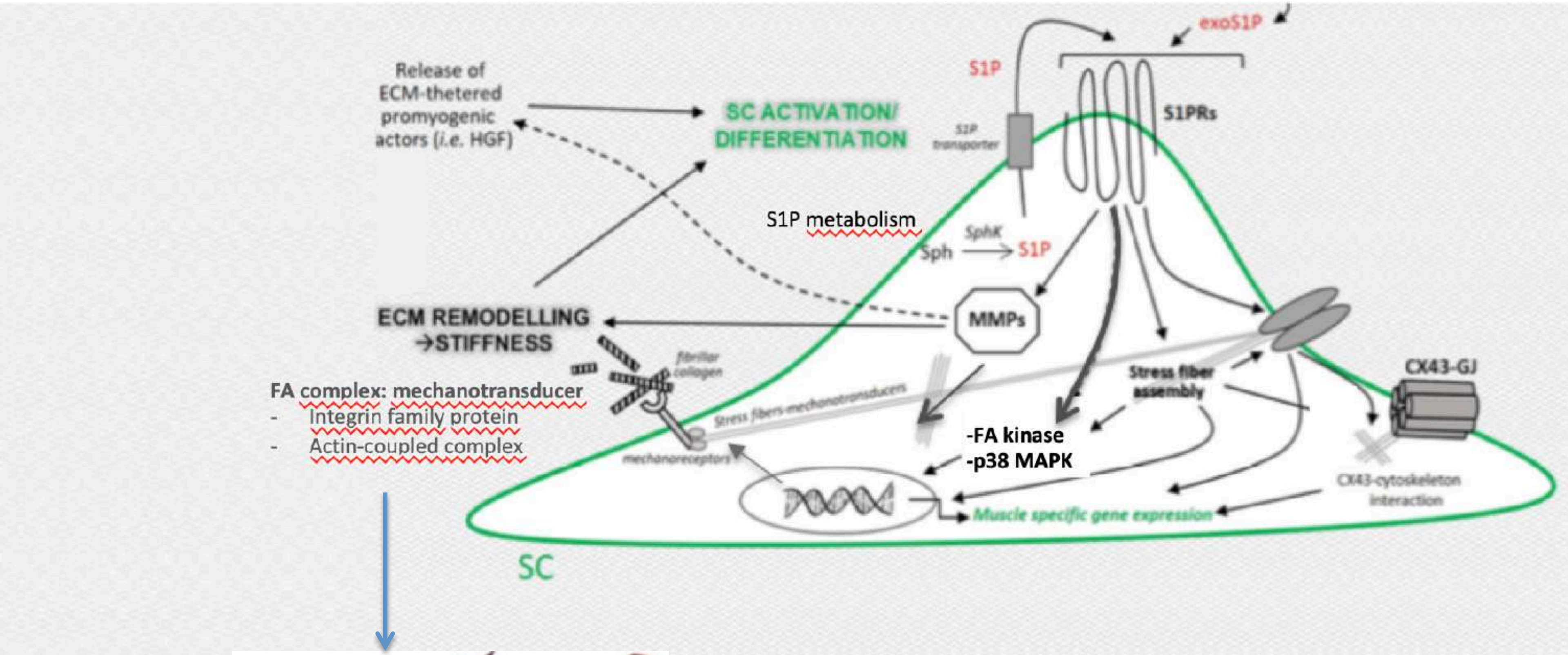


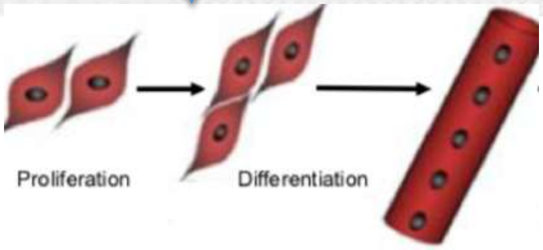


Structural components of cell membrane
signal transduction, cell growth, differentiation,

Dynamic changes in S1P metabolism: endogenous mechanisms of tissue repair/ regeneration

Sphingosine 1-Phosphate (S1P)/ S1P Receptor Signaling and Mechanotransduction





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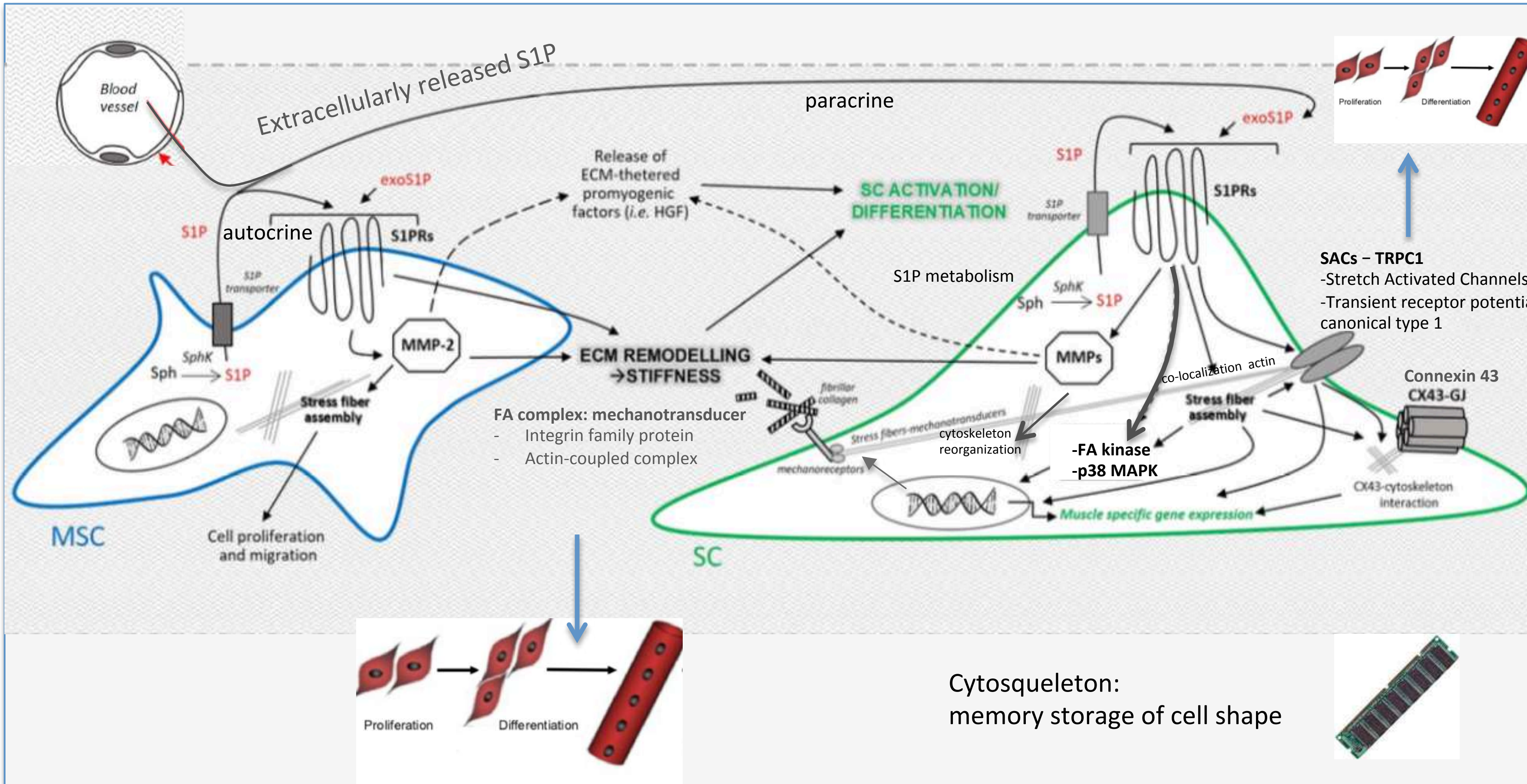
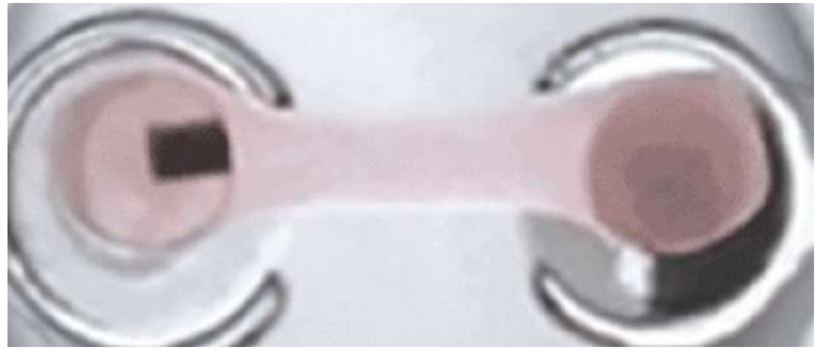
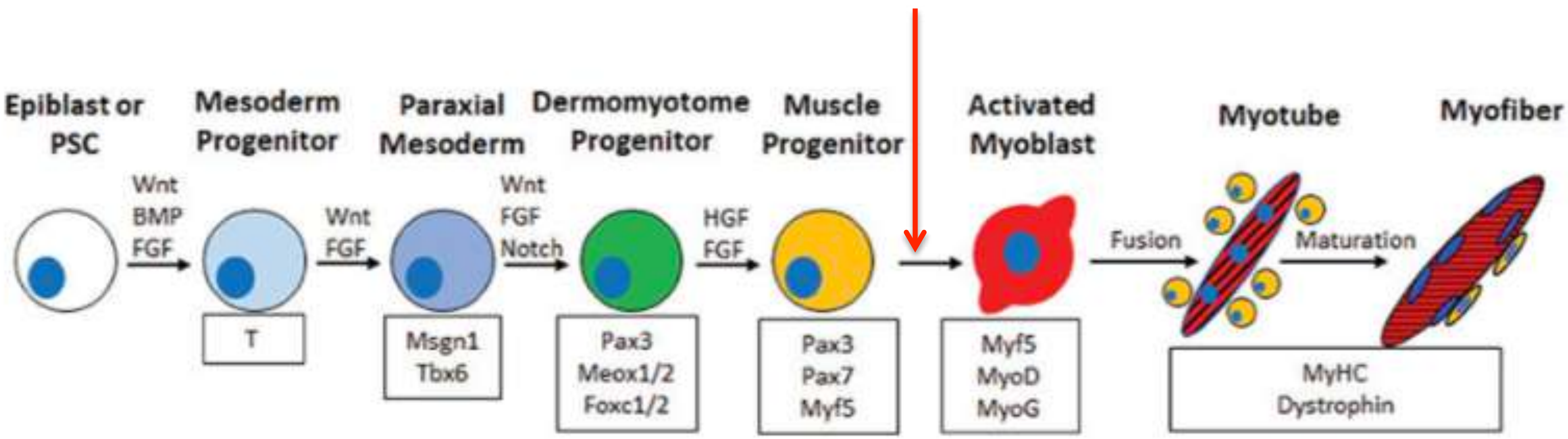


Int. J. Mol. Sci. **2019**, *20*, 5545

Muscle on a ship



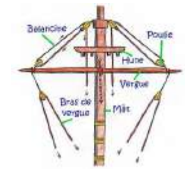
Material-controlled release of S1P ?

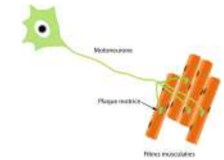


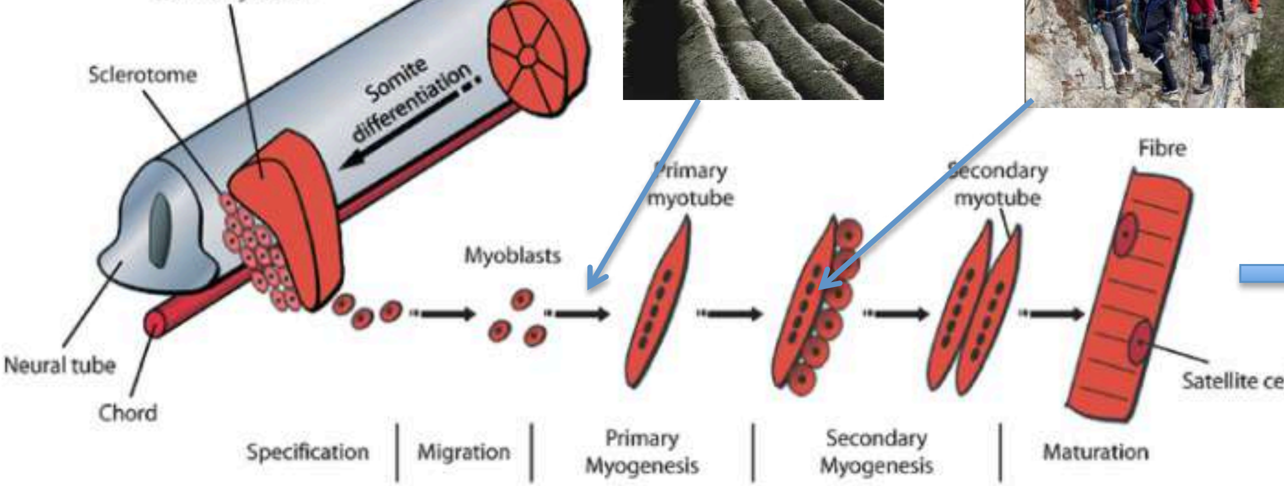
Skeletal muscle tissue engineering:
To reproduce in vitro the environment of Muscle Progenitor

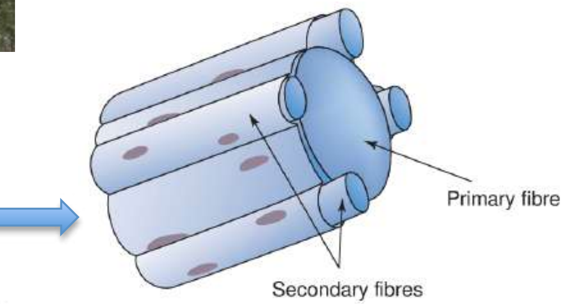
Basal lamina

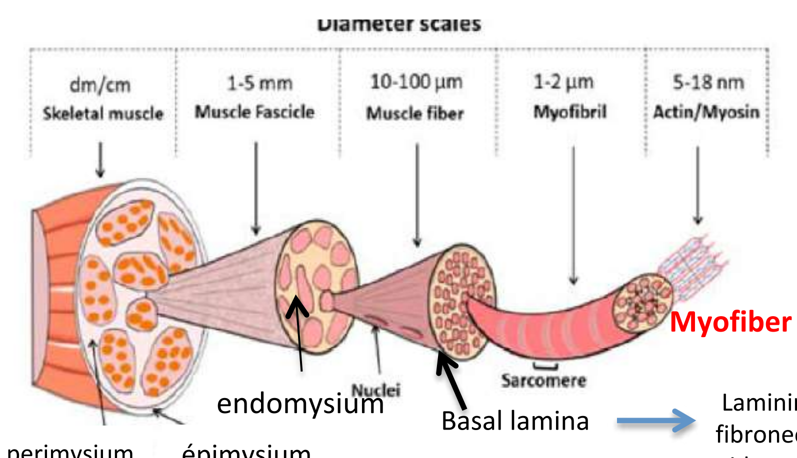
Primary myotube

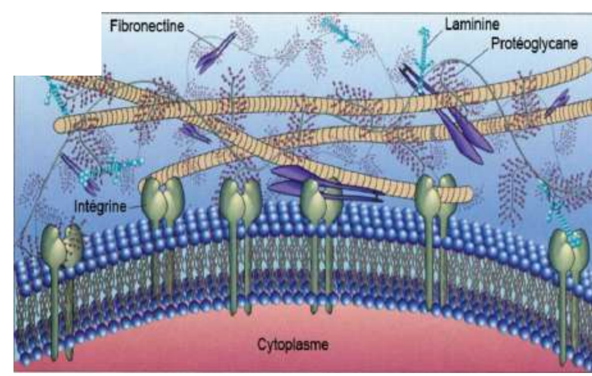








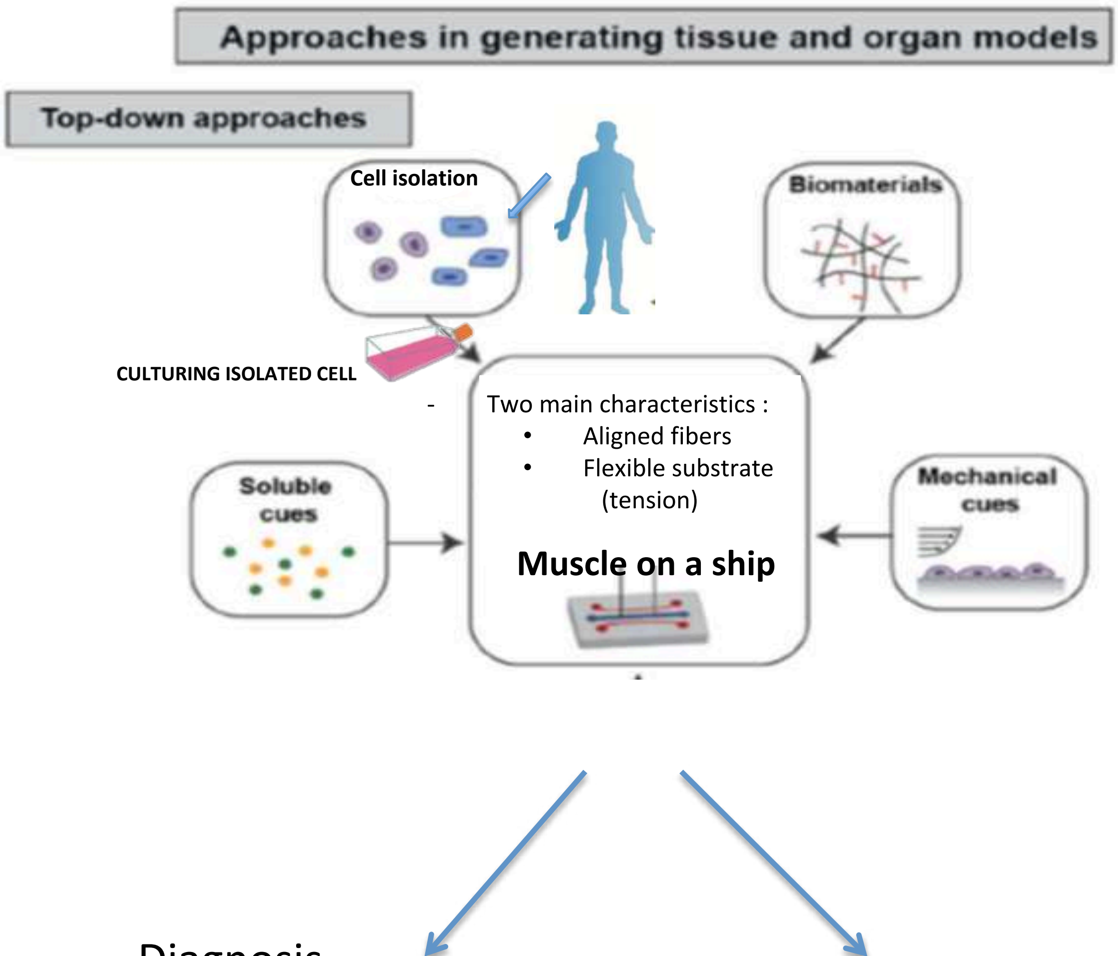




Laminin-rich matrix, collagen IV, fibronectin, nidogen/entactin, perlecan (protéoglycan)

Supportive scaffold to spatiotemporally regulate muscle progenitor migration and fate

Skeletal muscle tissue engineering (SMTE)



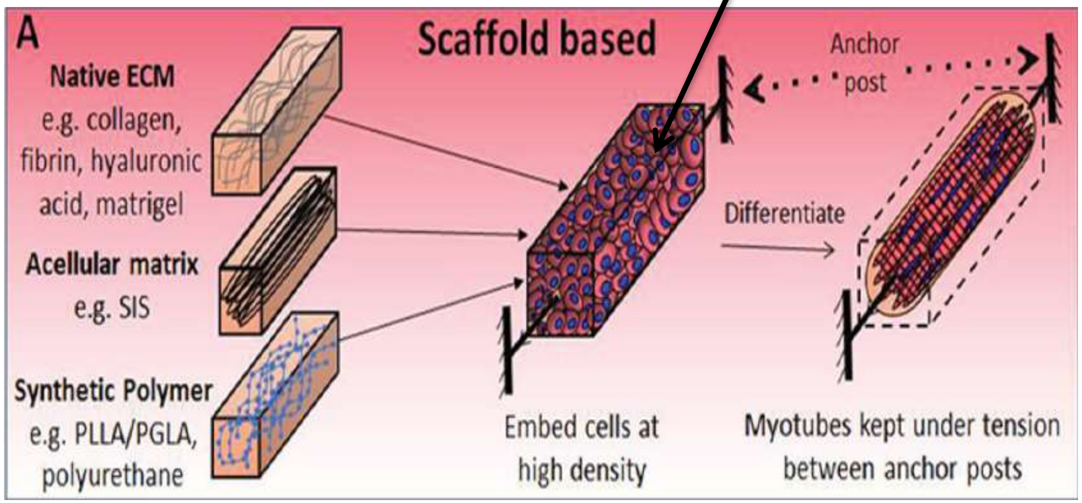
Diagnosis
Screening

Treatment

Skeletal muscle tissue engineering:
How To reproduce in vitro the environment of Muscle Progenitor ?

Hydrogel-based scaffolds: Hydrophilic polymer

constant bulk tension in a nylon support structure

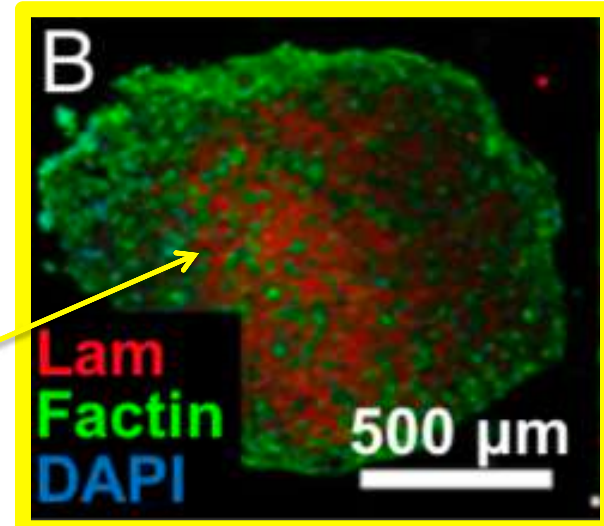


Hydrogel-based scaffold for in vitro engineering

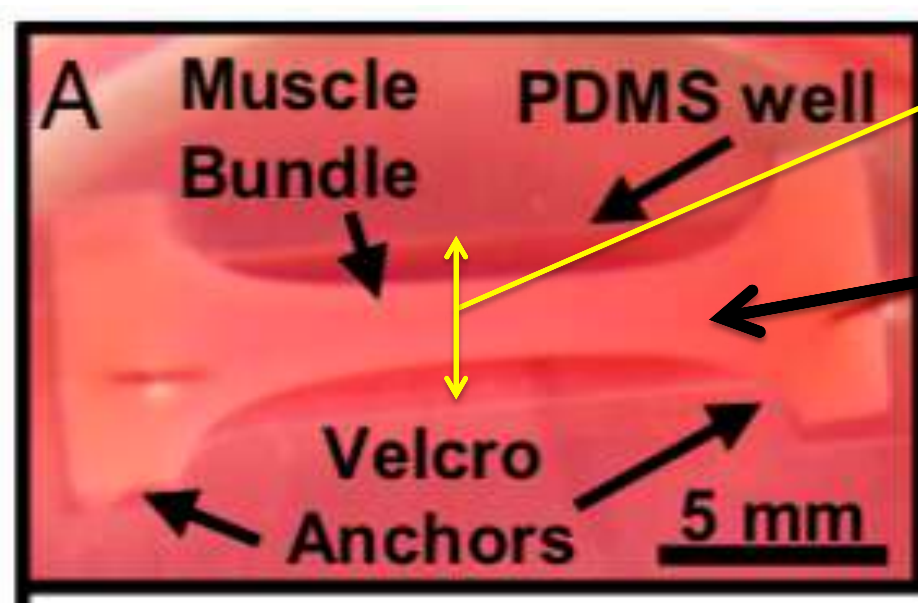
- Wide variety of hydrogels : collagen, fibrin, gelatin, alginate and polymers.
- Ease to functionalize them with adhesion peptides and conductive polymers.
- Muscle tissue-like stiffness which improves myotube differentiation[2]
- Possibility to create a volume up to 1mm of diameter[3]

1: Khodabukus and al. « In Vitro Tissue-Engineered Skeletal Muscle Models for Studying Muscle Physiology and Disease. » *Advanced healthcare materials* 2018
2: Engler and al. « Myotubes differentiate optimally on substrates with tissue-like stiffness: pathological implications for soft or stiff microenvironments. » J Cell Biol. 2004.
3: Chen and al. « Engineering multi-layered skeletal muscle tissue by using 3D microgrooved collagen scaffolds. » Biomaterials 2015;

Skeletal muscle tissue engineering:
How To reproduce in vitro the environment of Muscle Progenitor



Hydrogel-based scaffolds



Cell/hydrogel mixture/growth medium/ fibrinogen/ Matrigel

Polymerized at 37 °C

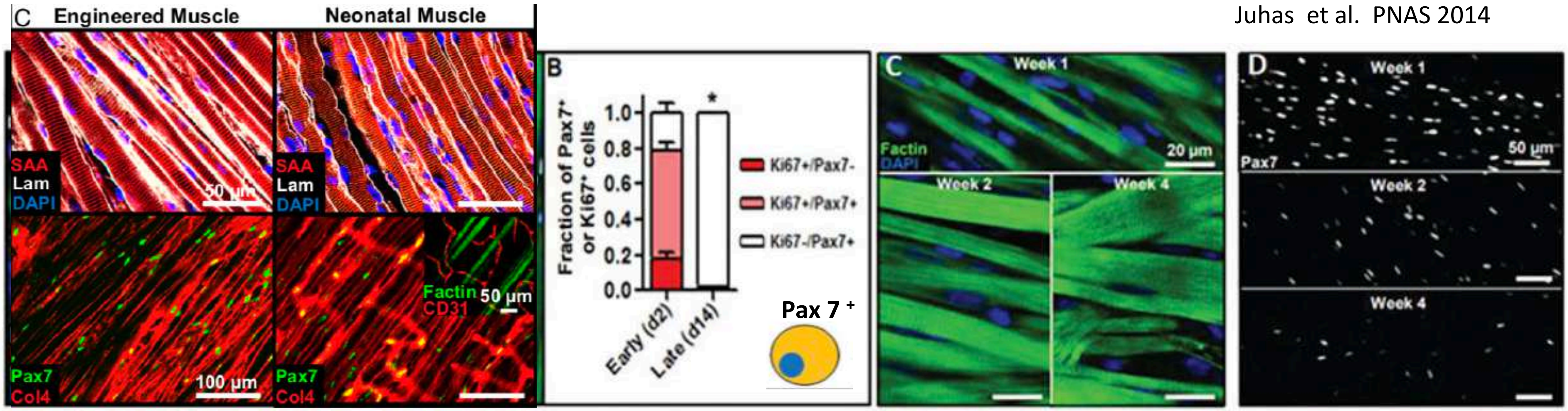
cultured

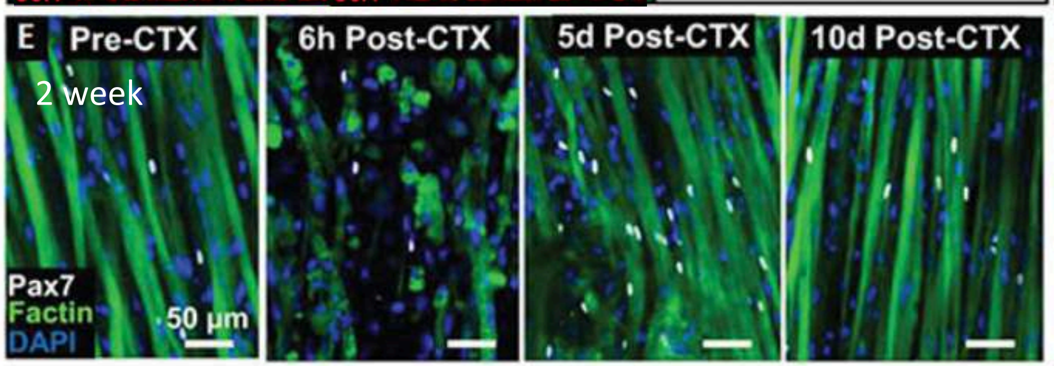
Matrigel resembles the laminin/collagen IV-rich basement membrane extracellular environment found in many tissues and is used by cell biologists as a substrate

Skeletal muscle tissue engineering:
Testing the Ability of satellite cells to maintain functionality in vitro

SCs from neonatal rat muscles and expanded for 2 d before tissue fabrication

Juhas et al. PNAS 2014





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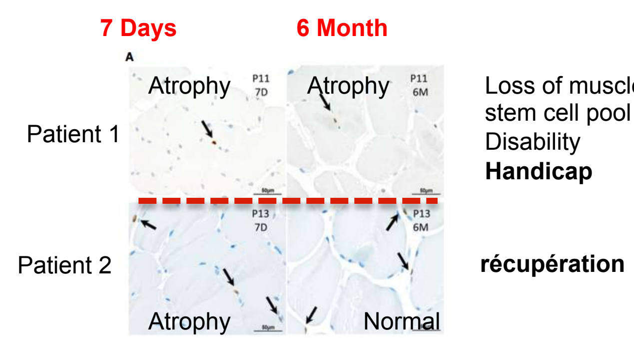
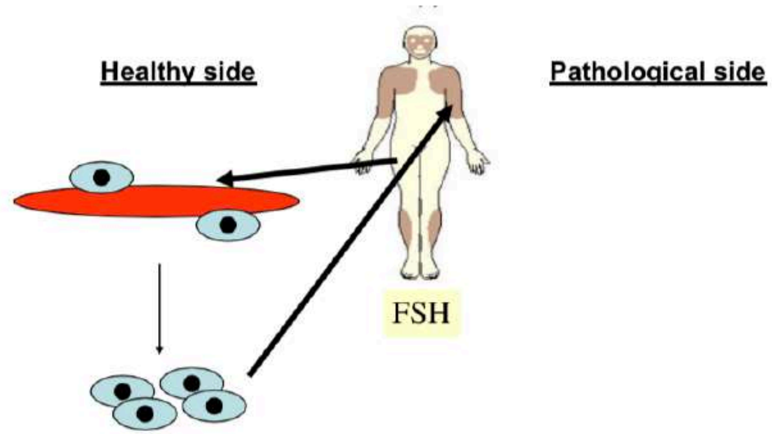
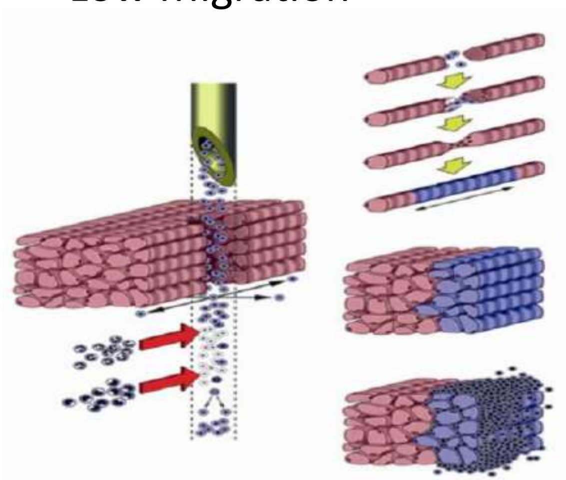


Dos Santos AJRCCM 2017

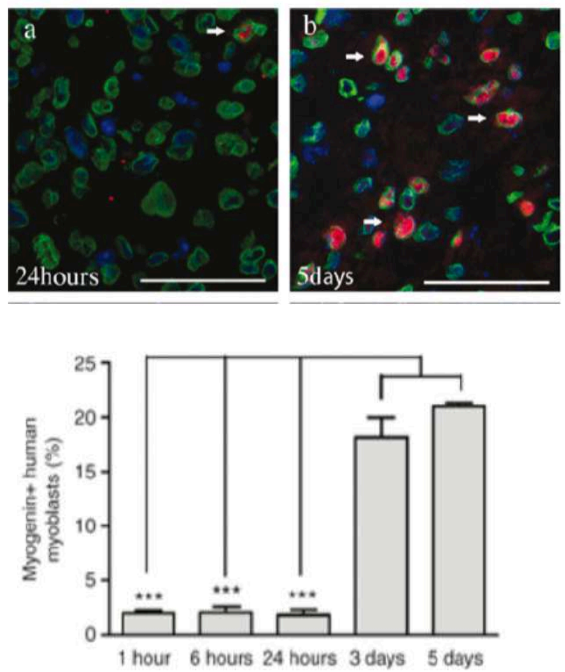
Skeletal muscle tissue engineering:
A potential vector for SCs implantation…



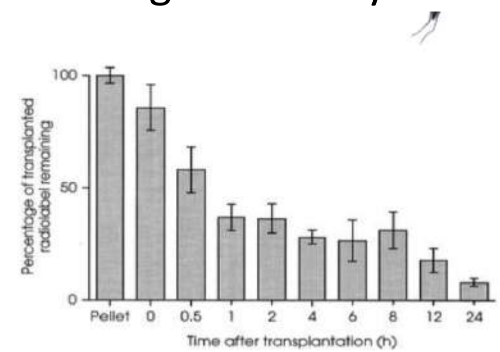
Low Migration



Early differenciation

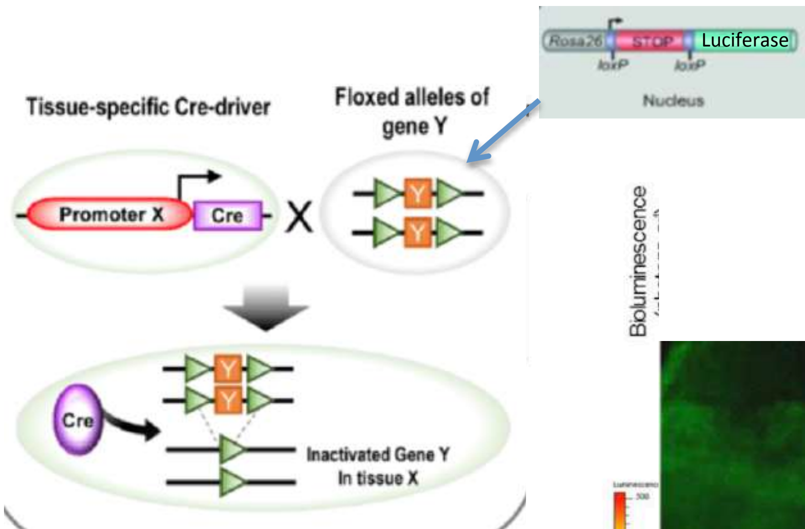


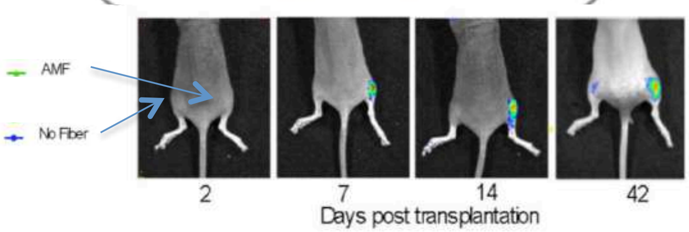
High mortality

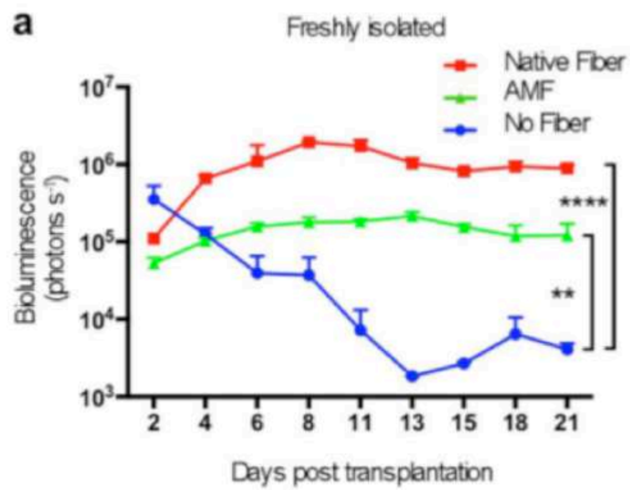


Skeletal muscle tissue engineering:
A potential vector for SCs implantation…

Pax7CreER and the ROSA26LuSEAP mouse strains to genetically label MuSCs with the Luciferase reporter

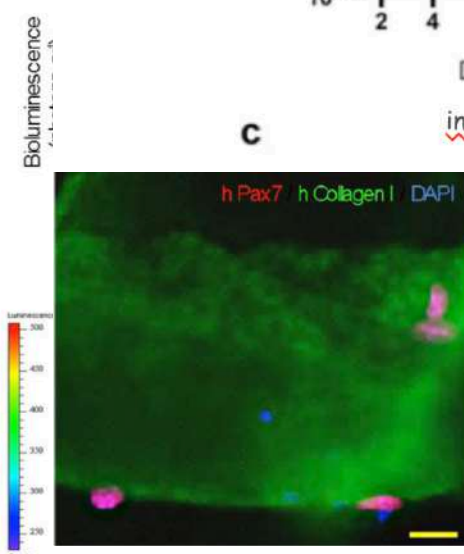


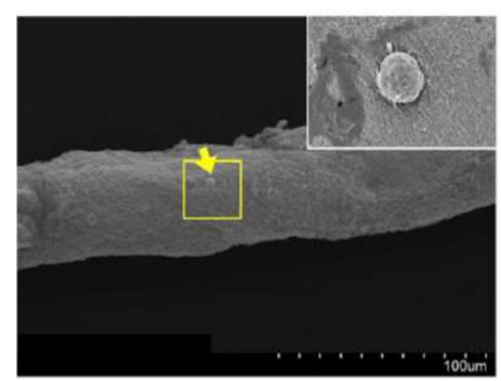




into pre- injured TA muscles

c

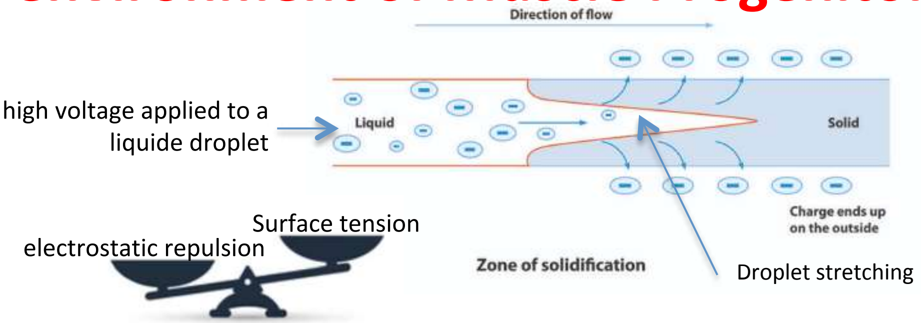


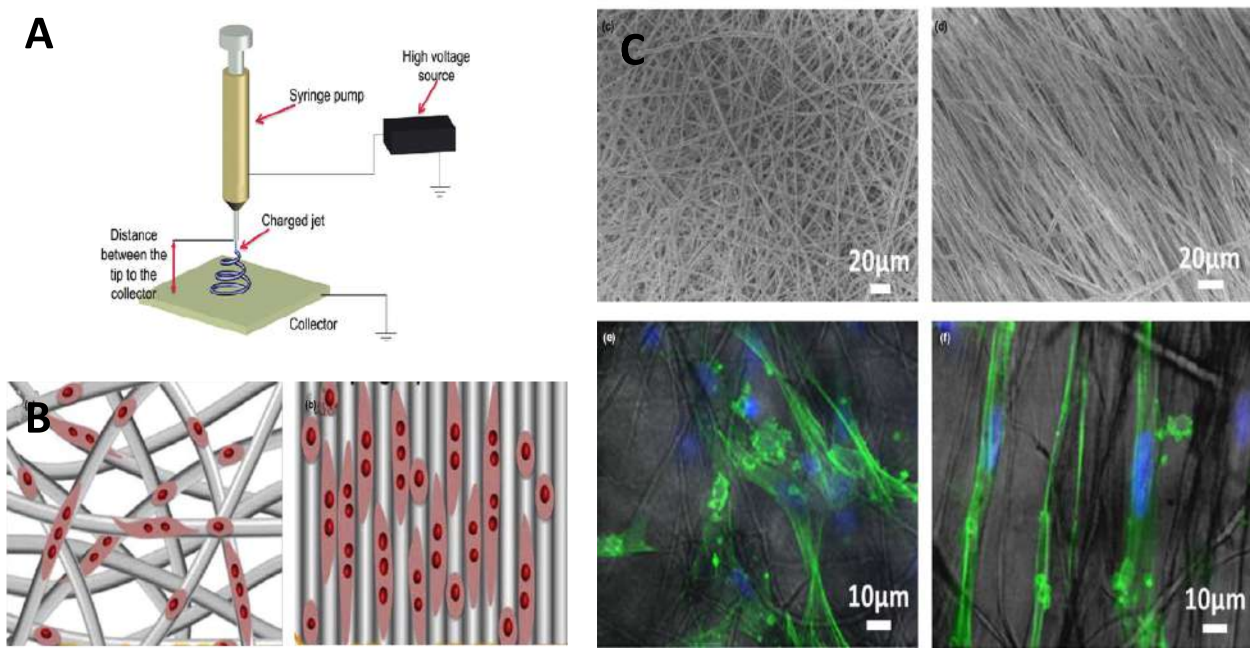


Nat Biotechnol. 2016 July ; 34(7): 752–759.

Skeletal muscle tissue engineering:
How To reproduce in vitro the environment of Muscle Progenitor

Electrospun scaffolds





Scheme of A) electrospinning technique B) random and aligned fibers
C) SEM micrographs of PCL scaffolds (Poly lactic-co glycolic acid)

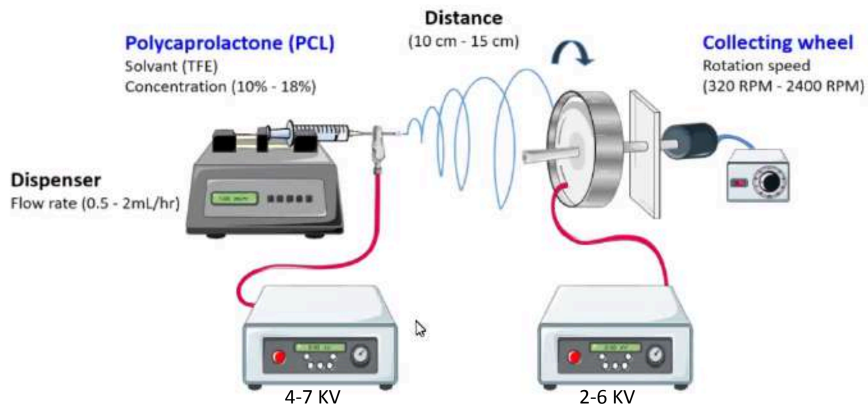
- Wide variety of biodegradable polymers (PCL, PLGA) with High molecular cohesion

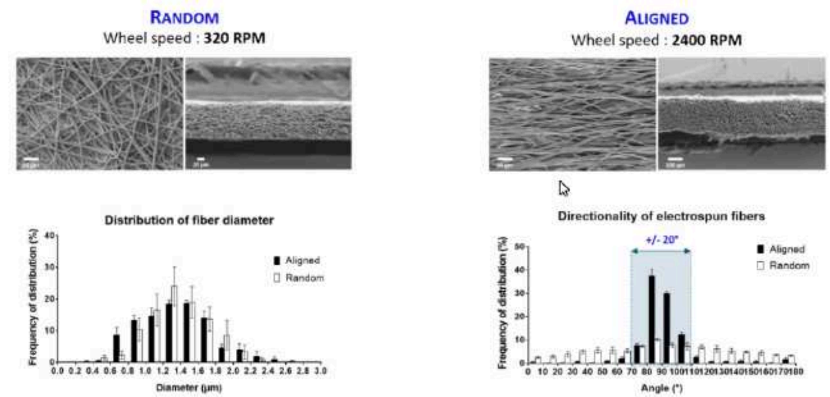
Already used for fabrication of controlled drug delivery systems (in-situ gels, microspheres, implants, nanoparticles….)

- Easy to produce aligned fibers and to regulate the diameter

- Long and difficult process to produce large scaffolds

Skeletal muscle tissue engineering:
How To reproduce in vitro the environment of Muscle Progenitor





⇒ Similar **fiber diameter** distribution

⇒ Aligned scaffold

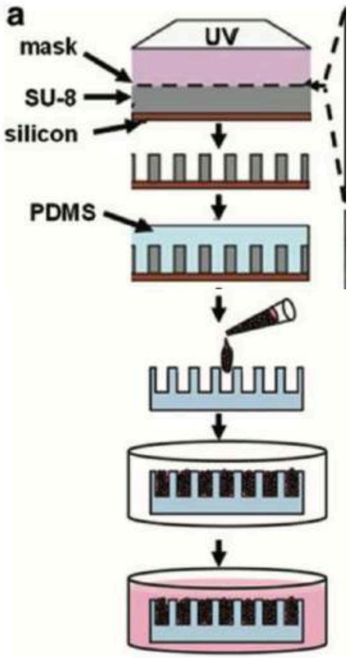
88 % of fibers oriented at same angle (± 20°)

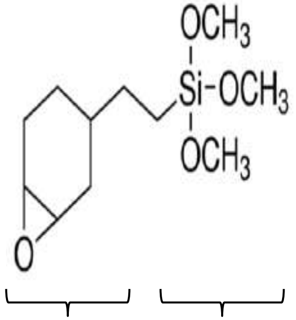
18

Skeletal muscle tissue engineering:
How To reproduce in vitro the environment of Muscle Progenitor

A Silicon wafer coated with SU-8

a negative photosensitive resin commonly used in the manufacture of micro-systems





- Hybrid component

- High reactivity and high conversion rate

- Organic part can be easily photopolymerized

- Inorganic part has a special affinity with substrates like glass or silicone

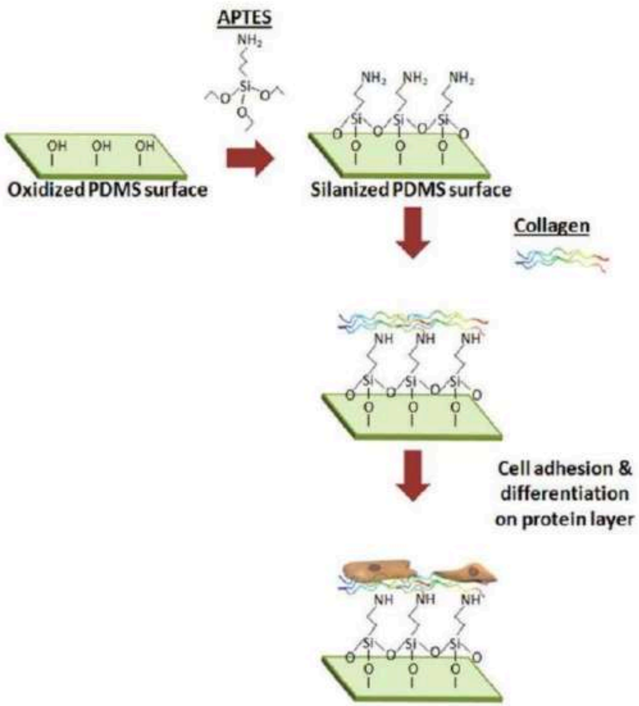
Skeletal muscle tissue engineering:
How To reproduce in vitro the environment of Muscle Progenitor

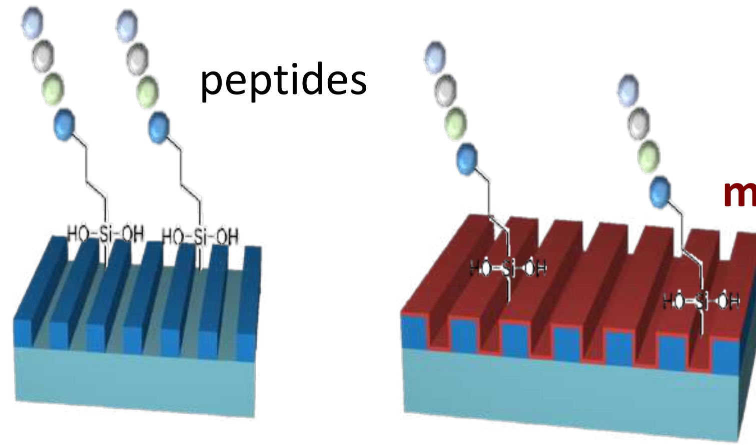
A Silicon wafer coated with SU-8

How to bio functionalize the silicon ? ….

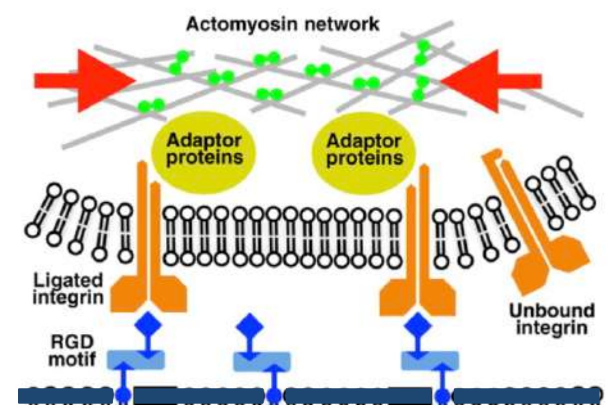
MEC mimic: peptid RGD
(tripeptide arginine, glycine, acide aspartique)

Alkoxysilans: APTES





matrigel



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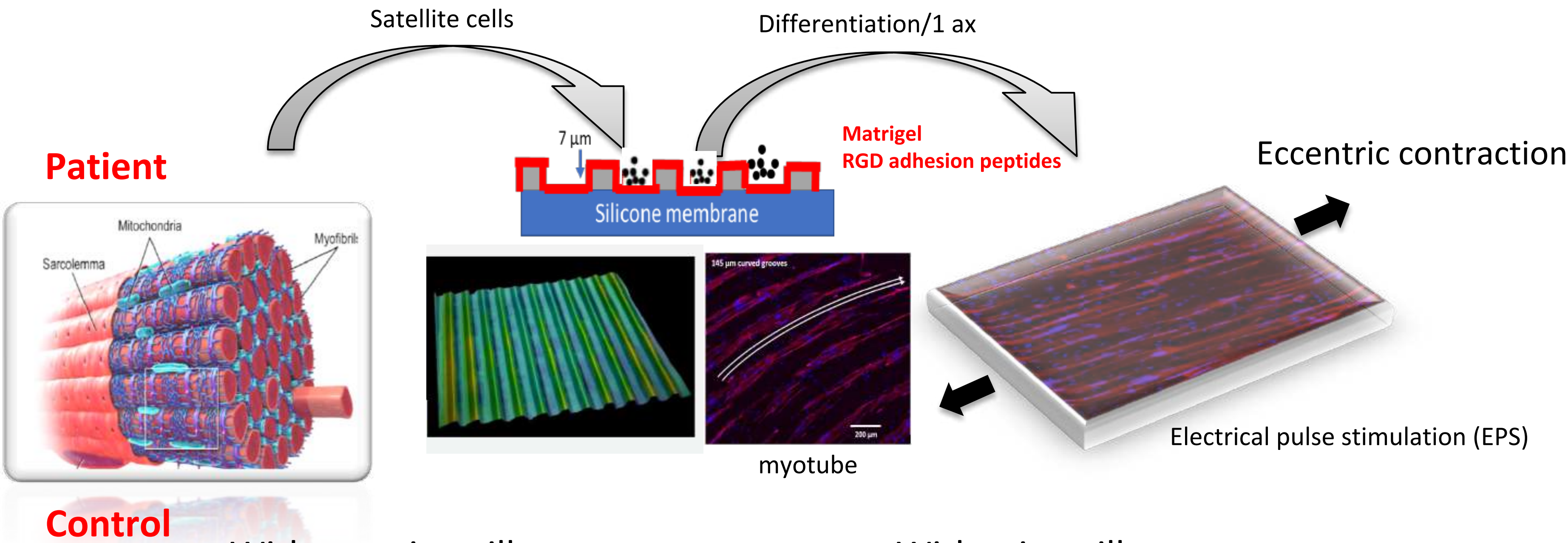
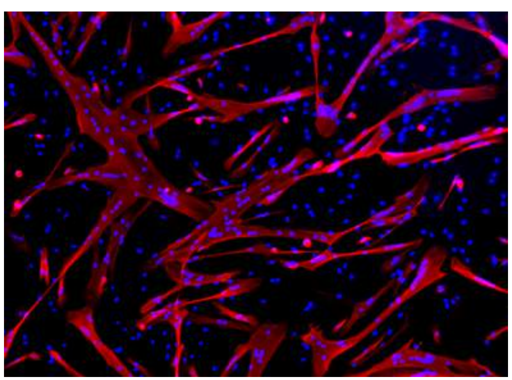
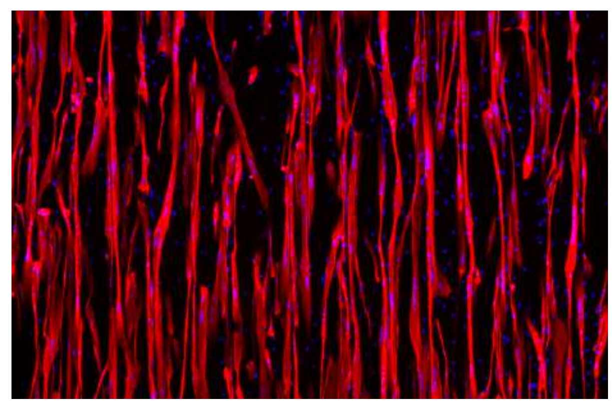


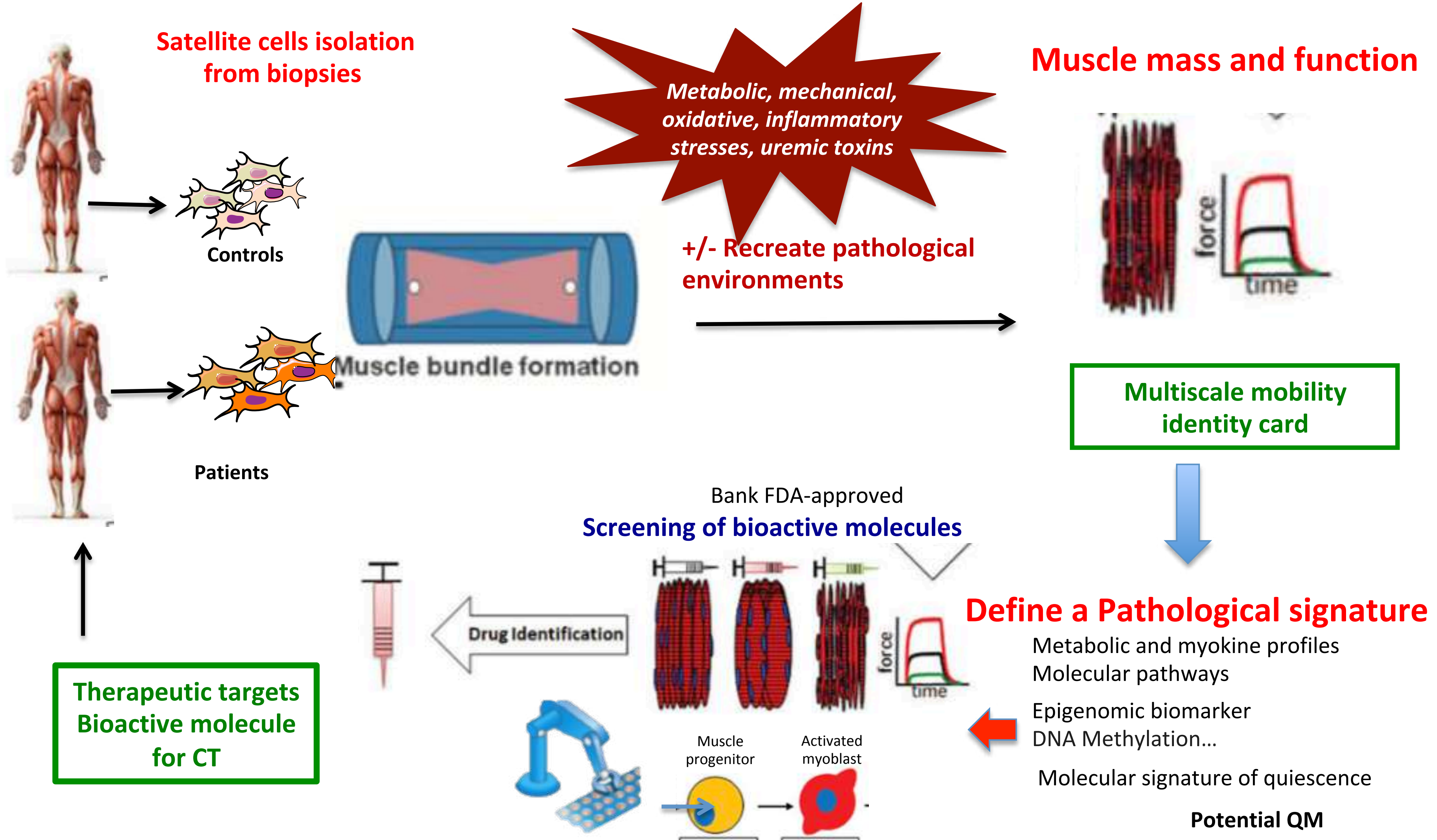
Without microsillon



With microsillon

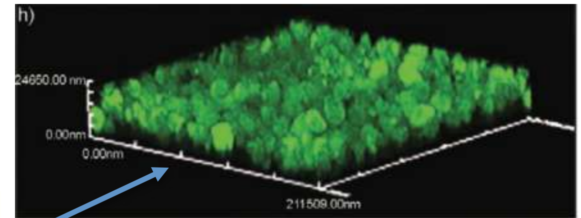


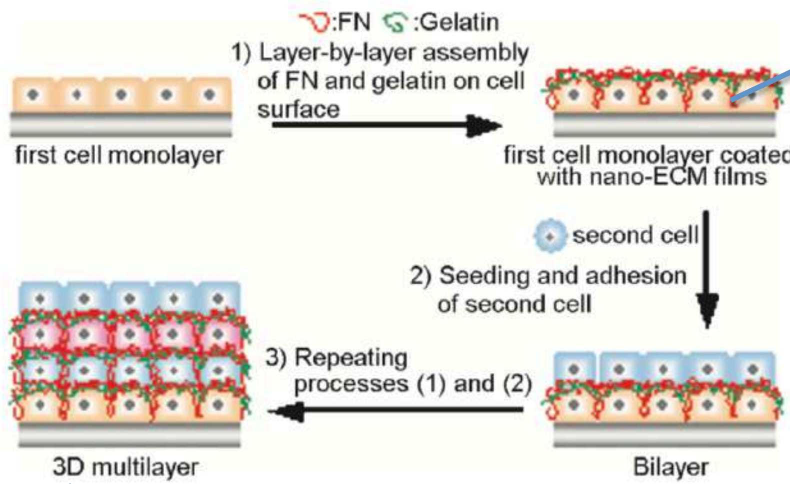
Interrest of Skeletal muscle tissue engineering (SMTE)

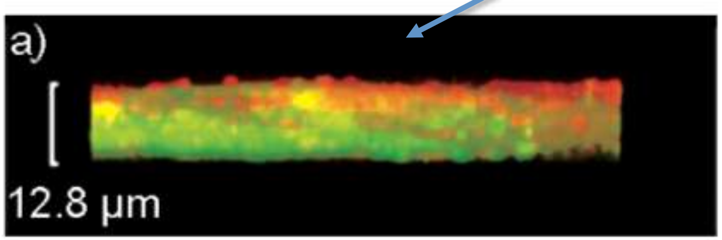


Skeletal muscle tissue engineering: From 2D to 3D

human umbilical artery smooth-muscle cells







human umbilical vascular endothelial cells (HUVEC)

human umbilical artery smooth-muscle cells

Accelerated implant perfusion and anastomosis with host vasculature in vivo

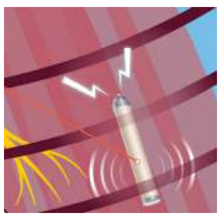
Bionic Initiative@Montpellier

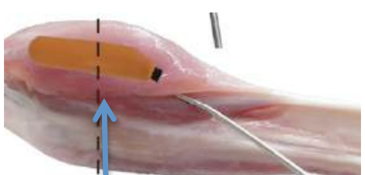
3D Muscle Tissue Engineering

Robotics – Electronics – Mechanics

Enhanced muscles

Intra-muscular sensors



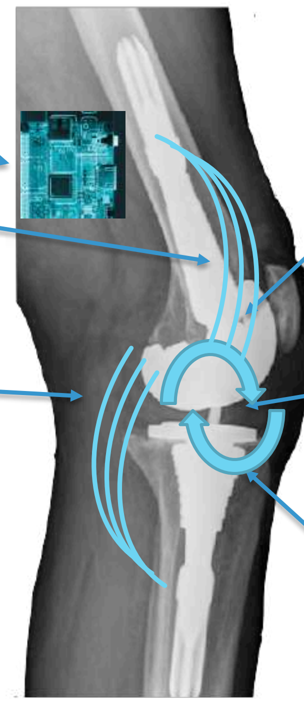


3D Muscle Tissue Engineering

-Vascularised

-Inervated

IA



Bio-mechanical design
motorisation

Biology printed meniscus

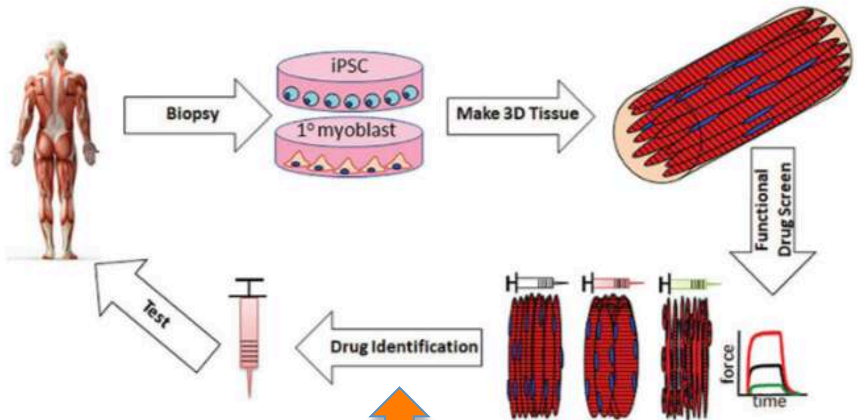
Knee artificial actuation
and integrated sensing

- *passive or active*?
- *what technology*?

LIRMM

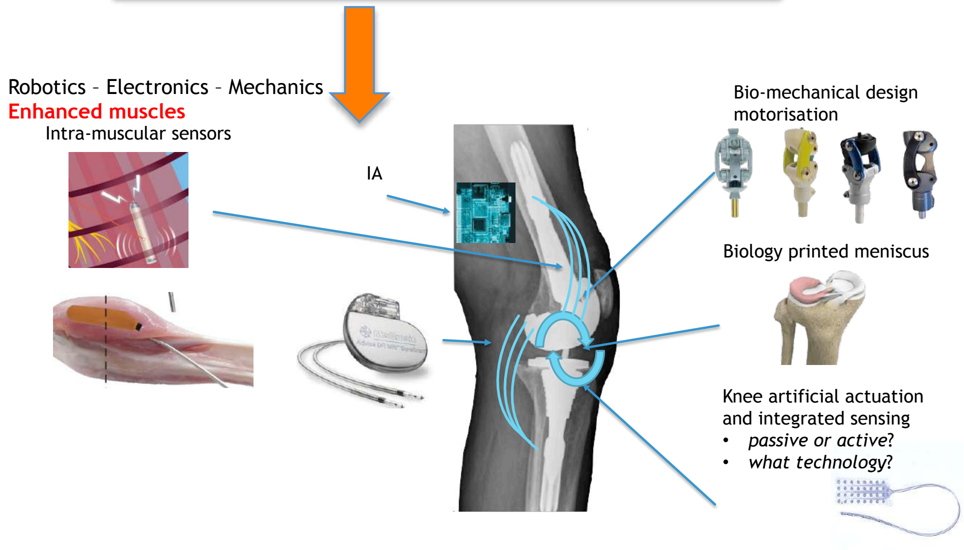
Laboratoire d'Informatique, de Robotique et de Microélectronique de Montpellier

To resume……



présent

In vitro tissue engineering



Futur…..long

5th SUNRiSE WEBINAR

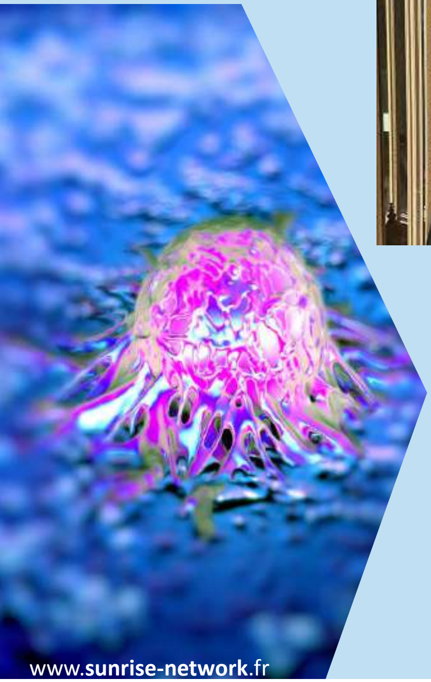
Monday, *October 11th, 2022*

11:00pm (CET) / Login: https://umontpellier-fr.zoom.us/j/96047043057

STEM CELLS: Between philosophy and biology

Dr Lucie Laplane, PhD

University Paris 1 Panthéon-Sorbonne
Institut Gustave Roussy
Arizona State University, USA



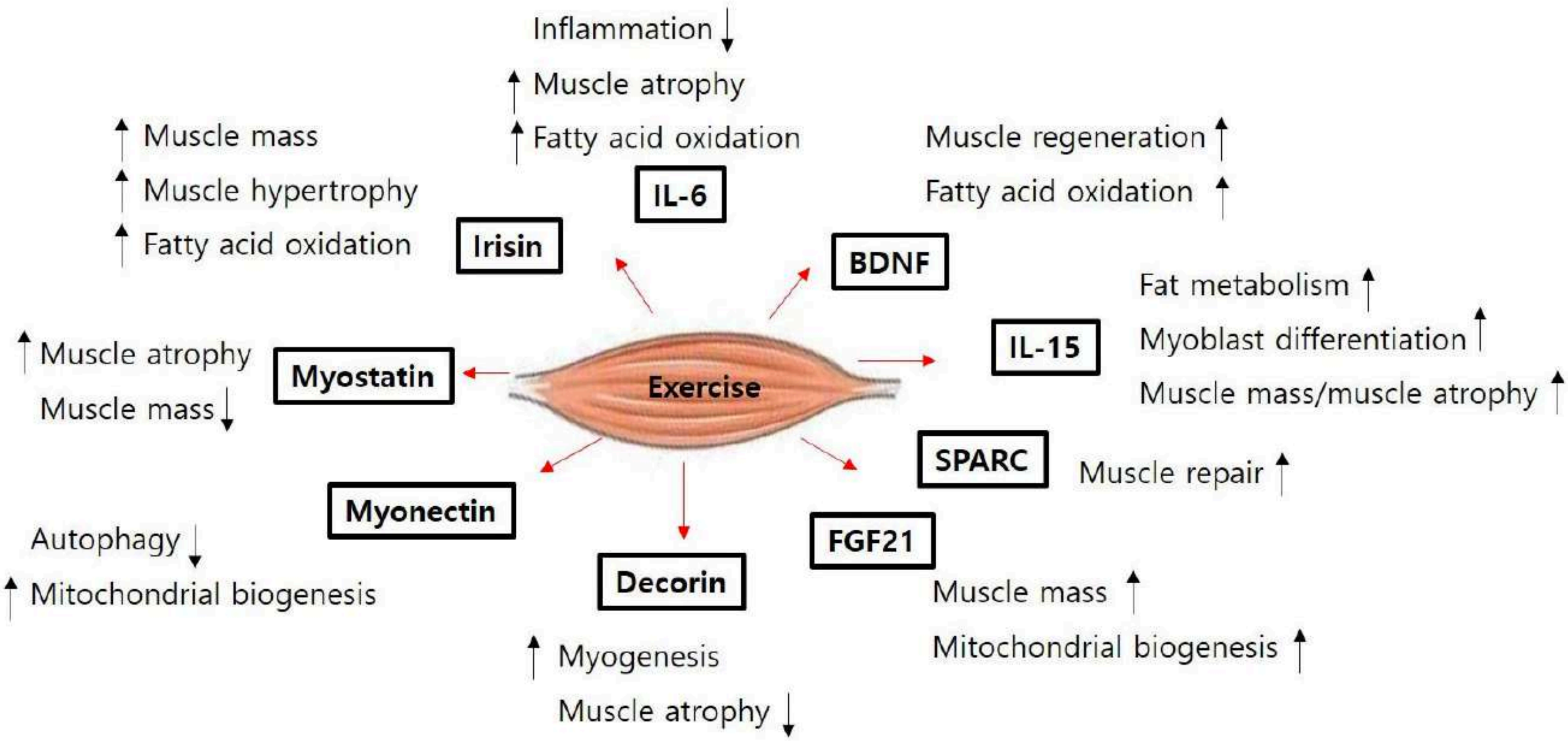
Research topic

Stem cells play a critical role in the development, daily renewal, and reparation/regeneration of tissues. They are also involved in various diseases in particular cancers. Yet, there is a lot that remains to be understood about them and the traditional view is being increasingly debated. This is both a biological and a philosophical issue. Mixing both approaches, I will discuss the following questions: (1) What kind of property is stemness? I will show that stemness can be four types of properties depending on tissues and contexts (2) Does it matter? I will highlight practical consequences of each type of stemness for cancer treatment (3) Is stemness stable? I will review empirical data questioning stemness stability and suggesting that some cancers could be associated with a switch in stemness property. (4) Is stem cell a unified biological category? I will end with a perspective on how to handle the debate on stemness natural kind by mixing philosophy, experimental biology and phylogenetic analyses.

Additional informatrion

Role of Myokines in Regulating Skeletal Muscle Mass and Function

Autocrine and paracrine effets



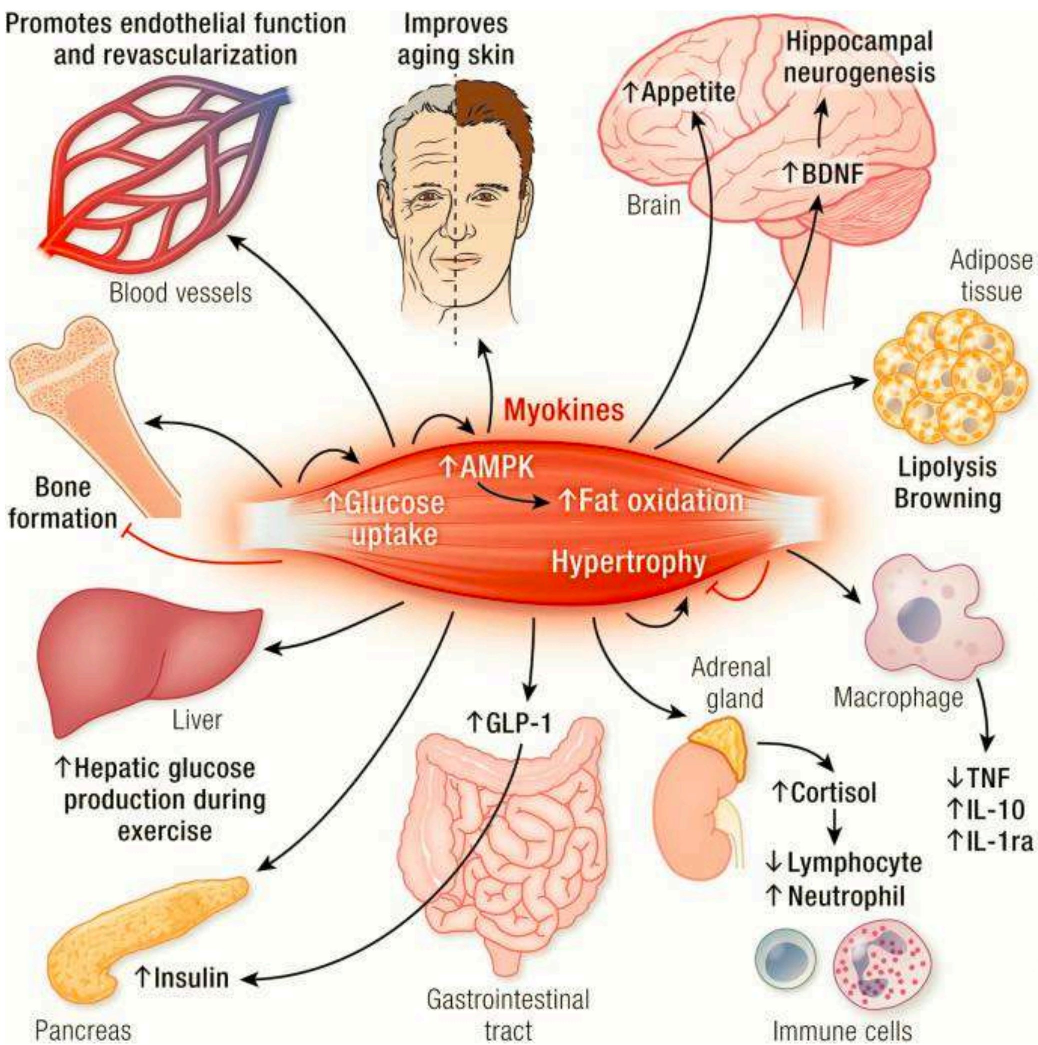
BDNF: Brain-Derived Neurotrophic Factor

SPARC: Secreted Protein Acidic and Rich in Cysteine

Front. Physiol., 30 January 2019
Sec. Striated Muscle Physiology
https://doi.org/10.3389/fphys.2019.00042

Muscle–Organ Crosstalk: The Emerging Roles of Myokines

endocrine effets



Exercice !!!

exercise-associated anti-inflammatory effects

Endocr Rev. 2020 Aug; 41(4): 594–609. PMC7288608