PhD Open Days

A novel biomimetic platform for cartilage tissue engineering and in vitro disease modelling

BIOENGINEERING - Cell Therapies and Regenerative Medicine

KRISTIN SCHÜLER (kristin.schuler@tecnico.ulisboa.pt)

iBB Institute for Bioengineering and Biosciences, Tagus Park

Fig. 1. Hierarchical structure of cartilage with collagen arcades [6]

Motivation and Background

Osteoarthritis (OA) is the most common cartilage disease affecting over 600 million people worldwide with numbers yet increasing due to the aging population. However, disease mechanisms are not yet fully understood, and therapeutics demonstrate still very limited success in relieving patients' pain and reversing their immobility. Therefore, **relevant disease models** are urgently required to **understand the disease mechanisms and find effective drugs**. [1, 2]

In cartilage research, a variety of models are existing. However, due to the complexity of the situation *in vivo*, drastic simplifications are done and mostly only parts of the system get recapitulated in in-vitro models ranging from very simple 2D cell sheets with a single cell type to 3D tissue cultures trying to mimic the complex architecture of articular cartilage. [3, 4]

Yet, **a model able to mimic the native hierarchical structure of articular cartilage in all three layers** is yet to be developed, **especially in regard to the orientation and alignment of collagen type II fibres**, which make up around 95 % of the articular collagen, and 60 % of the dry-weight of cartilage tissue (Fig. 1). [5]
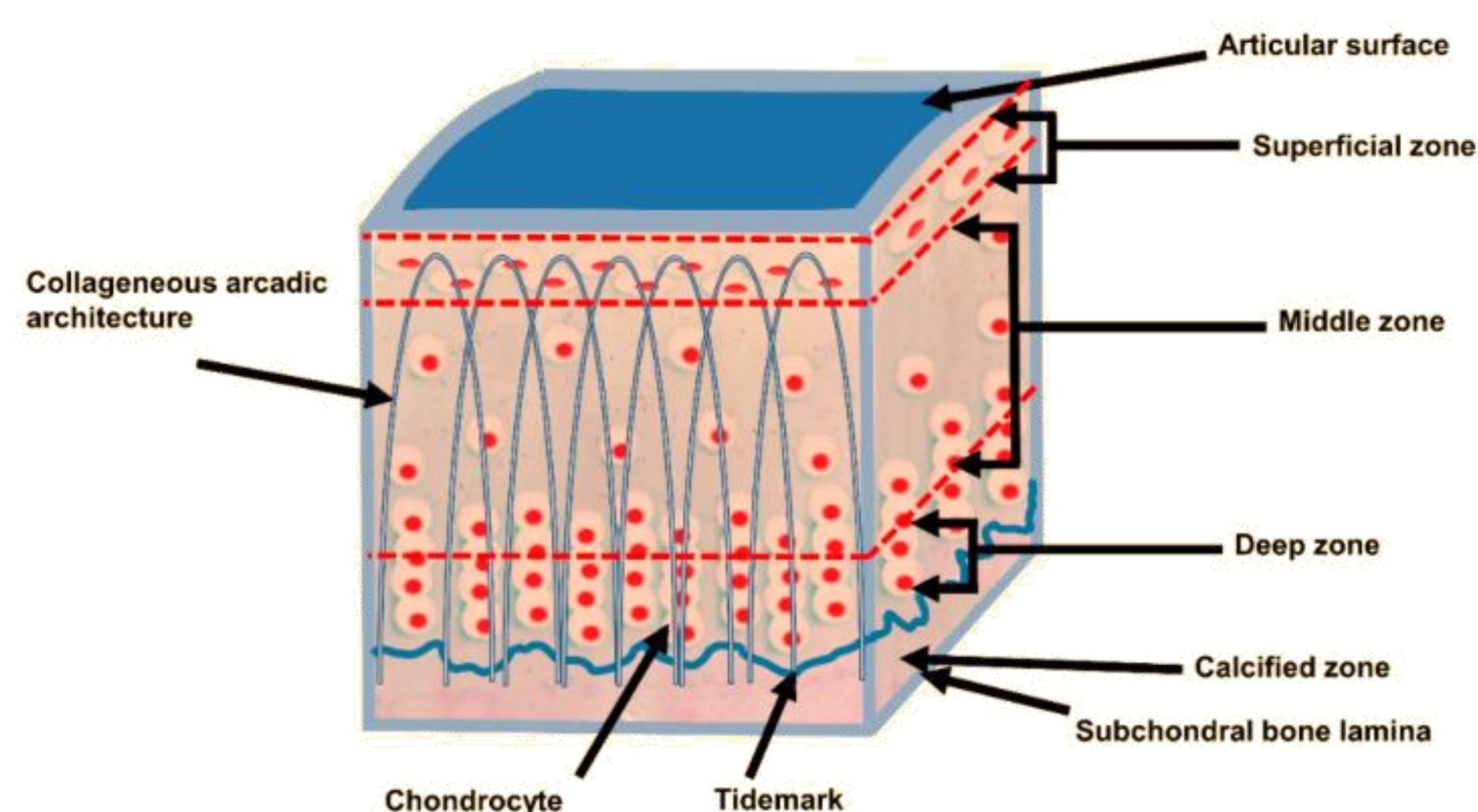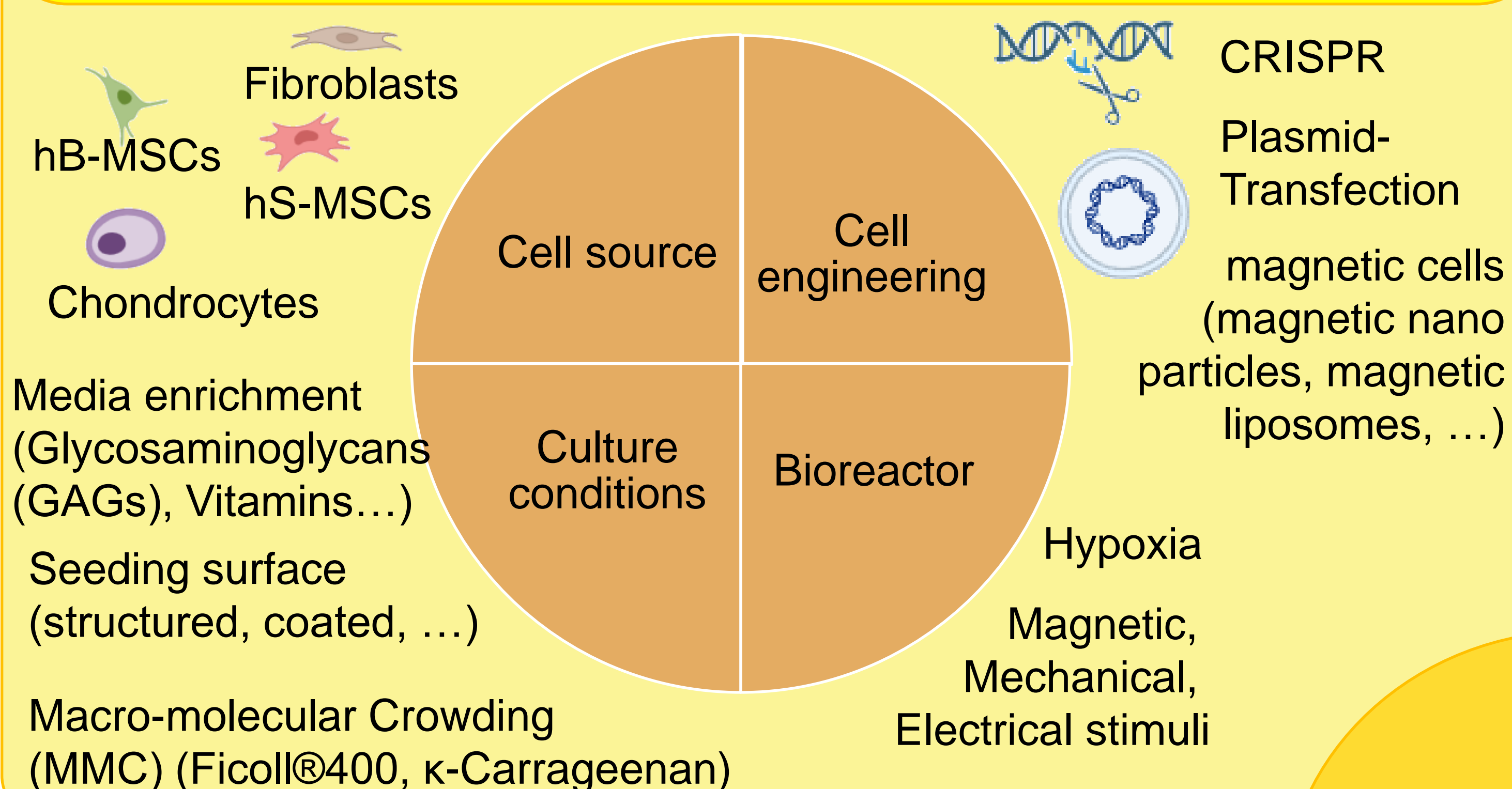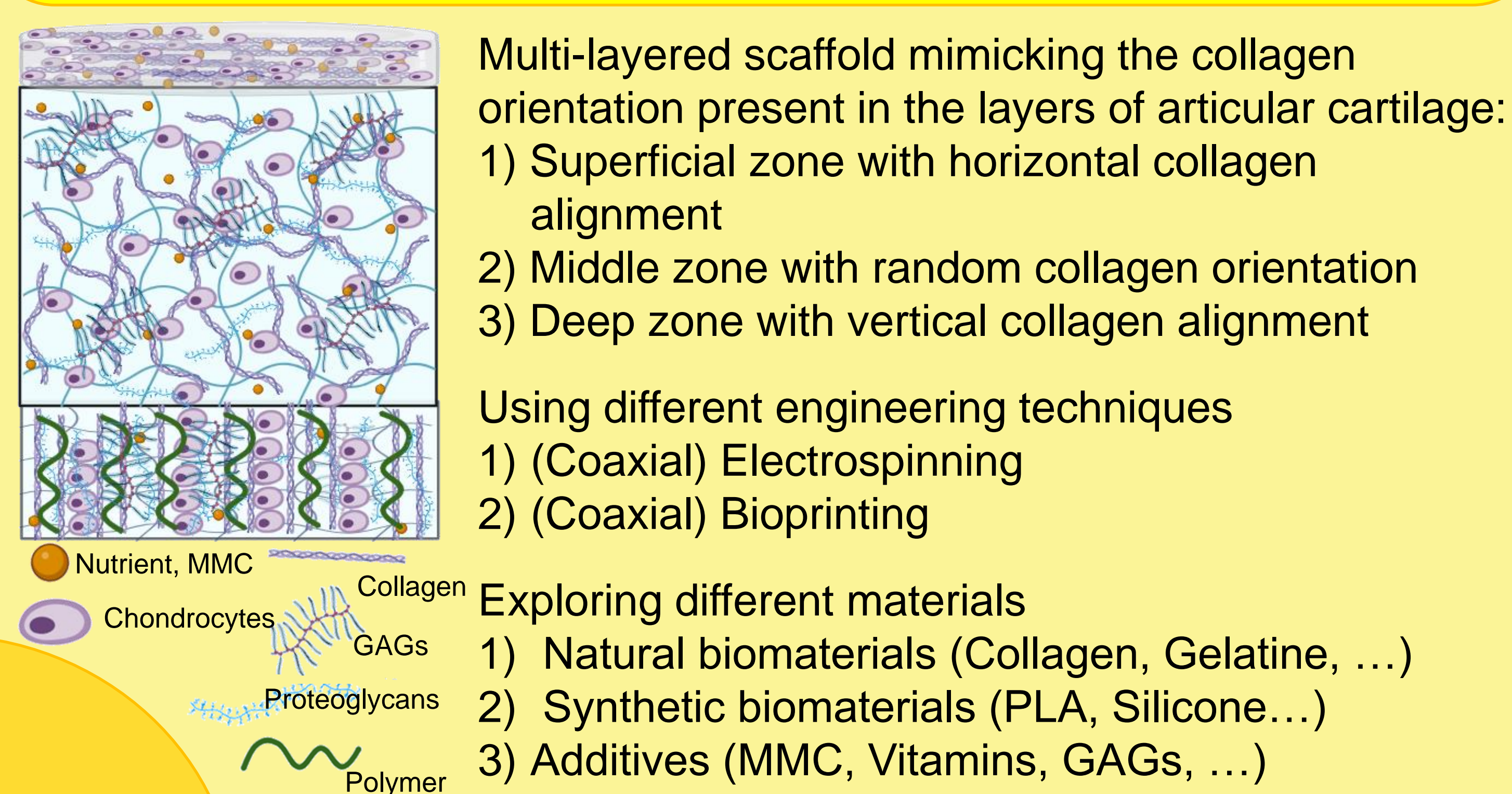
Concept

Collagen production enhancement and alignment
AIM: Cell-produced scaffold/dECM (bottom-up)

Fibroblasts
hB-MSCs
hS-MSCs
Chondrocytes

Cell source · Cell engineering · Culture conditions · Bioreactor

CRISPR
Plasmid-Transfection
magnetic cells (magnetic nano particles, magnetic liposomes, …)
Hypoxia
Magnetic, Mechanical, Electrical stimuli

Media enrichment (Glycosaminoglycans (GAGs), Vitamins…)

Seeding surface (structured, coated, …)

Macro-molecular Crowding (MMC) (Ficoll®400, κ-Carrageenan)

Collagen alignment – with scaffold:
AIM: Complex bio-mimetic 3D scaffold (top-down)

Nutrient, MMC
Chondrocytes
Collagen
GAGs
Proteoglycans
Polymer

Multi-layered scaffold mimicking the collagen orientation present in the layers of articular cartilage:
1) Superficial zone with horizontal collagen alignment
2) Middle zone with random collagen orientation
3) Deep zone with vertical collagen alignment

Using different engineering techniques
1) (Coaxial) Electrospinning
2) (Coaxial) Bioprinting

Exploring different materials
1) Natural biomaterials (Collagen, Gelatine, …)
2) Synthetic biomaterials (PLA, Silicone…)
3) Additives (MMC, Vitamins, GAGs, …)

Cartilage Tissue Engineering

Chondrogenic differentiation
AIM: Complex biomimetic culture condition

One bioreactor with multiple stimuli to test influence of
- O_2 tension
- dynamic culture (fluid stress)
- electrical stimulation
- co-culture (static/dynamic)
- hydrostatic / hydrodynamic pressure
- magnetic stimulation

Different cultures of cell-aligned or artificially aligned scaffolds to find the best conditions for chondrogenic differentiation

Disease modelling osteoarthritis
AIM: Proof of model

1) Disease induction with known cytokines
2) Study of the diseased tissue and differences to healthy tissue (knowledge gain)
3) Disease treatment with known OA drugs (reversibility of disease, proof of model)

Inter leukin-1β
Tumor Necrosis Factor (TNF)-α
TNF-α
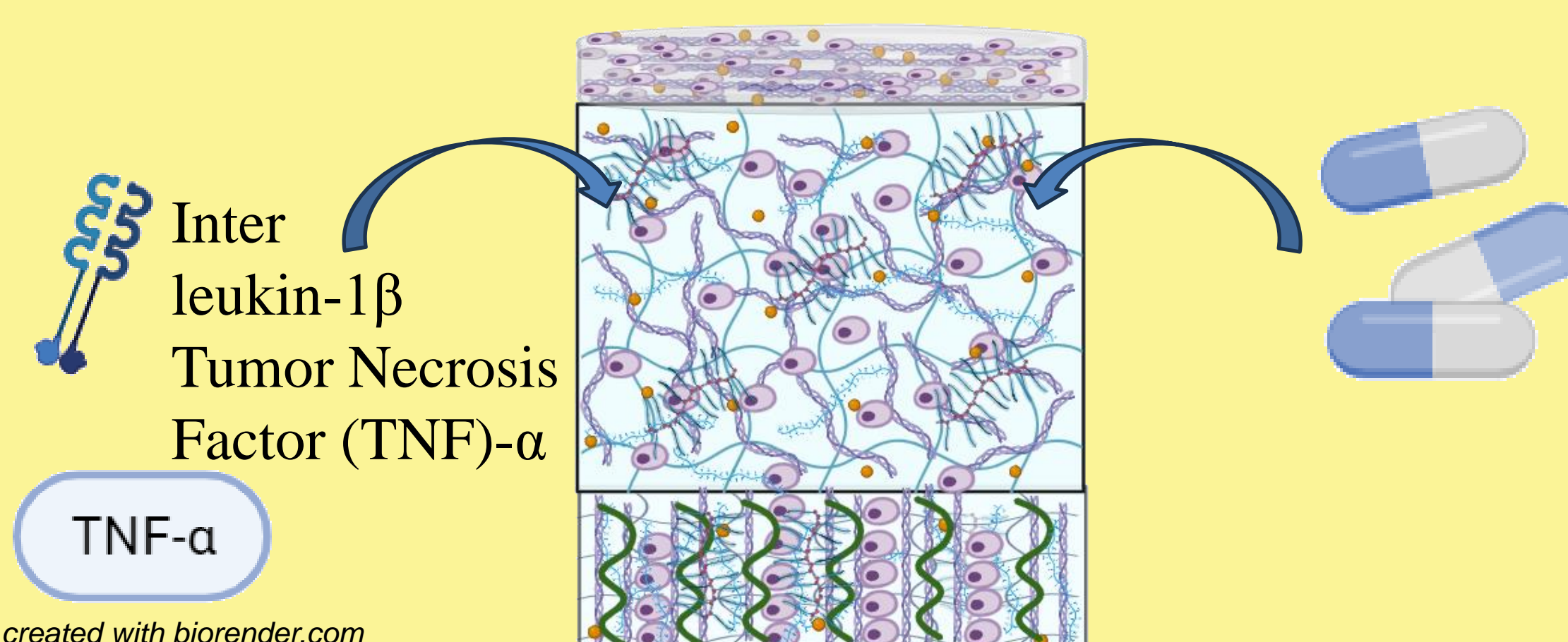
Images were created with biorender.com

Sources
[1] Neogi, T. (2013): The epidemiology and impact of pain in Osteoarthritis.
[2] I. Bartolotti *et al.* (2021): A Roadmap of In Vitro Models in Osteoarthritis: A Focus on Their Biological Relevance in Regenerative Medicine
[3] X. Niu *et al.* (2022): Integrated gradient tissue-engineered osteochondral scaffolds: Challenges, current efforts and future perspectives
[4] H. J. Samvelyan *et al.* (2020): Models of Osteoarthritis: Relevance and New Insights
[5] N. S. Gudmann, M. A. Karsdal (2016): Chapter 2 – Type II Collagen
[6] Joerg Eschweiler et al (2021): The Biomechanics of Cartilage—An Overview

Supervisor: Prof. Dr. Frederico Castelo Ferreira

Co-Supervisor: Dr. Paola Sanjuan-Alberte, Dr. João Carlos Silva

phdopendays.tecnico.ulisboa.pt