A Novel Cross-linkable, Microfiber-like Collagen Scaffold Supports Chondrocyte Differentiation and Growth

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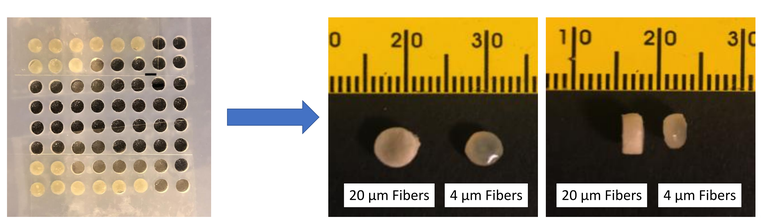
Objective

- We have created a **novel scaffold** composed of cross-linkable collagen fibers, which can be mixed with cells and **cross-linked into any shape desired**
- The fibrous scaffold **creates a 3D porous structure**: **Different fiber sizes can produce different pore sizes** and potentially influence tissue growth/differentiation
- The purpose of this study **was to identify the optimal fiber/pore size for promoting cartilage tissue growth from therapeutic mesenchymal stem cells (MSC)** based on in-vitro cultures

Methods

- Two UV light cross-linkable scaffolds of different fiber size (**thin 4µm and thick 20µm**) were used
- **Human-originated MSCs were encapsulated** in the scaffolds to form cylindrical (2.5 mm thick by 5 mm wide) samples
- **Cultivated in-vitro** with growth media - TGF-β added one week after encapsulation
- **Assessments were made (on days 1, 21, and 42)**

Fig.1 Gross appearance of samples at day 0 and day 42



Results

- **All samples remained in their cylindrical form** with no fragmentation [Fig.1]
- **Mechanical testing** demonstrated significant **increases in the compression properties** in samples of **both fiber sizes** from day 1 to 42
- **Histological analysis** showed **increased GAG production** in samples **of both fiber groups** [Fig 2]
- **Biochemical testing**, showed a significant (p=0.024) **increase of GAG in the 4µm fiber group** between day 21-42
- At day 42, the **average GAG content of 4µm fiber** group, was **almost 11 times higher** than the **20µm fiber** group (P=0.021)
- **qRT-PCR** showed a **more significant rise in both aggrecan** (P<0.002), **and collagen II** (P<0.009) **in the 4µm fiber** group compared to the 20 µm fiber group [Fig 3]

Fig. 2 Histological analysis showed improved ECM production with thinner fibers

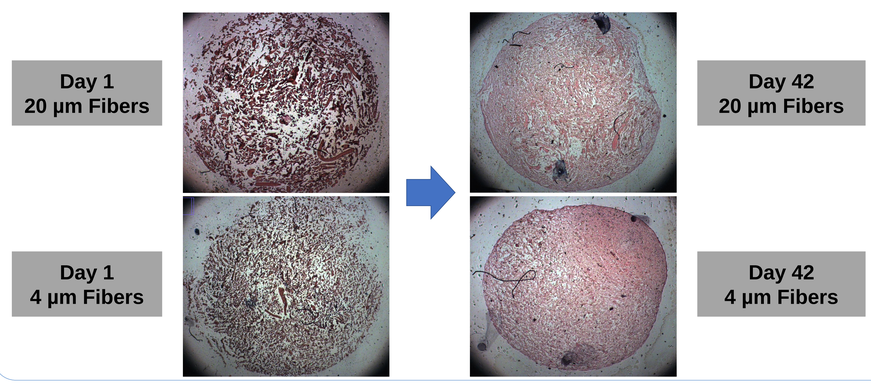
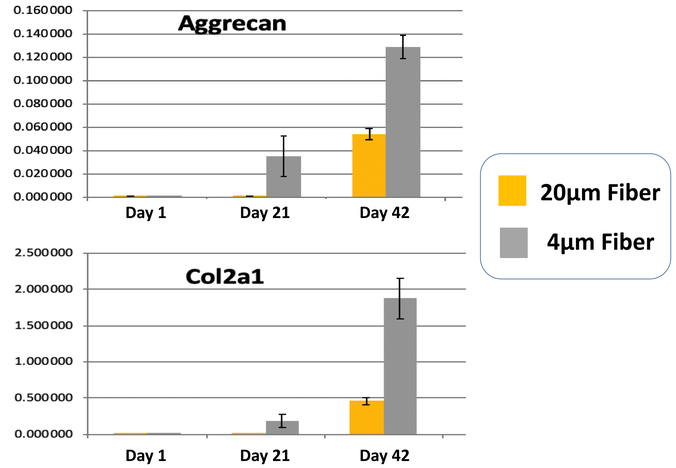


Fig. 3 qRT-PCR Gene expression showed enhanced Aggrecan and Collagen II production with thinner fibers



Conclusions

- **Thinner (4 µm) fibers create pores about the size of a chondrocyte** (about 20 µm) whereas larger fiber/pore size scaffolds (20 µm) create larger pores (about 100 µm)
- When MSCs are cultivated in chondrogenic media in-vitro, **thinner fibers promote chondrogenesis more strongly then thicker fibers**

Significance

- Cellular microenvironments that mimic the native state enhance chondral differentiation and may improve **future chondral defects repair solutions**