## Core-shell pure collagen threads extruded from highly concentrated solutions promote colonization and differentiation of C3H10T1/2 cells

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## Supplementary Data.



**Figure SI-1**: Protocol of cell culture on collagen threads under tension. 1) 6-well plate bases are covered with silicon gel (SYLGARD) and silk sutures are pinned with needles. 2) Threads are put under tension between the two sutures and 3) C3H10 T1/2 cells are added. 4) After 3 weeks of culture, 5) extremities are cut and threads are observed by different techniques.



**Figure SI-2**: SEM images of threads surface at A) 15 mg/mL, B) 30 mg/mL, C) 60 mg/mL at day 1 and 14. A. appears flatter than B. or C. where a more pronounced roughness is observed. TOP scale bars =  $25 \ \mu$ m, BOTTOM scale bars =  $5 \ \mu$ m.



**Figure SI-3**: SHG imaging of the full thickness of collagen threads at 60 mg/mL after 2 weeks in PBS 5X. A. and B. correspond to transverse images acquired at the depths -1- and - 2- indicated by the arrowheads in the cross-section reconstruction C. At the surface, clusters of fibrillary collagen are observed (A-1-). (B-2-) shows different textures defining two main regions seen in (C): the shell and the core separated by an area where the SHG signal is almost zero (black dashed line). Scale bars = 100  $\mu$ m.



**Figure SI-4:** Examples of C3H10T1/2 cell orientation assessment by means of Orientation J distribution analysis of actin filaments observed with 2PEF microscopy on 30mg/mL (left) and 60 mg/mL (right) threads. (Top) Hybrid images presenting the original Alexa Fluor 488 Phalloidin signal on one side and the local orientation as indicated by the circular color wheel (insert) on the other side. (Bottom) Histograms of orientations obtained for each specific example over the entire displayed image. Scale bars =  $50\mu m$ .



**Figure SI-5:** In situ hybridization on collagen threads cross-sections at 30 and 60 mg/mL after 3 weeks of culture. Tenomodulin (Tnmd) presents the strongest signal in both threads with collagen (ColIa1) coming right after. The myogenic gene (MyoD) is not detected. Scale bar =  $100 \mu m$ .



**Figure SI-6:** Scleraxis, collagen Ia1 and tenomodulin gene expression on 30 mg/mL collagen threads, effect of cell culture time at 1-, 2- and 3-weeks culture. (\*P < 0.05, \*\*P < 0.001, \*\*\*P < 0.001).



**Figure SI-7:** TEM images of cross-sections of collagen threads after 2 weeks in PBS 5X and after medium change to DMEM at  $37^{\circ}$ C for 2 weeks: at 30 mg/mL (A) and at 60 mg (B). Top scale bars = 500 nm, bottom scale bars = 200 nm.

TABLE SI-1		
Name	Direction	Sequence
	(F or R)	
36B4	F	ACCT CCTT CTTC CAGG CTTT
36B4	R	CTCC CACC TTGT CTCC AGTC
Col1a1	F	TGGA GAGA GCAT GACC GATG
Col1a1	R	GAGC CCTC GCTT CCGT ACT
Scx	F	CCTT CTGC CTCA GCAA CCAG
Scx	R	GGTC CAAA GTGG GGCT CTCC GTGA CT
Tnmd	F	AACA CTTC TGGC CCGA GGTA T
Tnmd	R	AAGT GTGC TCCA TGTC ATAG GTTT T

Table-S1: Table of primers used for the RT-q-PCR