

Figures

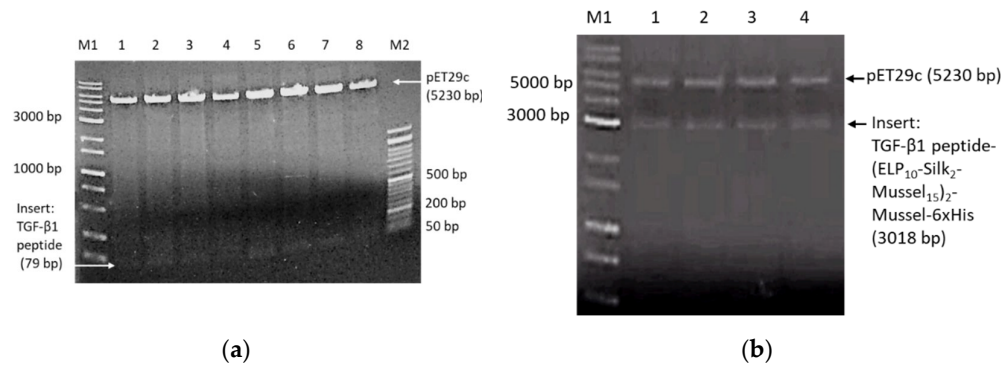


Figure S1. Synthesis of the gene "TGF-β1 peptide-(ELP₁₀-Silk₂-Mussel₁₅)₂-Mussel-6xHis". a) Electrophoresis in 1% w/v agarose gel of pET29c and "TGF-β1 peptide" insert after digestion of the recombinant plasmid "pET29c-TGF-β1 peptide" with NdeI and XhoI. Lanes 1-8: plasmids from different colonies that were screened. After DNA sequencing, it was verified that all colonies contained plasmids with the desired insert. b) Assembly of the gene "TGF-β1 peptide-(ELP₁₀-Silk₂-Mussel₁₅)₂-Mussel-6xHis" by PRe-RDL. Lanes 1-4: plasmids from different colonies that were screened by digestion with NdeI and XhoI. M1: 1 kb DNA ladder, M2: 50 bp DNA ladder.

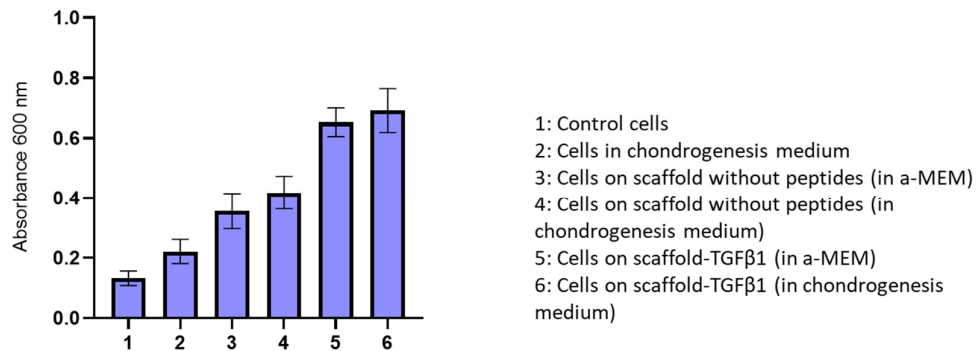


Figure S2. Quantification of Alcian Blue staining at 21 days. Optical absorption was measured at 600 nm with a microplate reader (Biotek Plate Reader).

Tables

Table S1. Nucleotide and amino acid sequences of the building blocks.

Building block	Nucleotide sequence (5' to 3')	Amino acid sequence
ELP	GTGCCGGGCGTTGGTGTTCGGGC GTCGGTGTCCCGGTAAAGGCGTT CCGGGTGTGGTGTTCGGGTGTT GGC	VPGVGVPGVGVPKGVP VGVPVG
Silk	GTGGGCGCGGGTGCCGGCAGCGG TGCAGGCGCTGGTTCTGGCGCTGG CGCGGGTTCGGTGCCGGTGCAG GCTCCGGCGCCGGCGCTGGTTCTG GC	V(GAGAGS) ₅ G
Mussel	GCGAAACCGAGCTATCCGCCGAC CTATAAA	AKPSYPPTYK
Fibronectin (RGD) peptide	TATGCGGTGACCGGTGCTGGTGAT AGCCCGGCCTCTAGCGGC	YAVTGRGDSPASSG
Laminin peptide	TATCATTACGTGACCATTACGCTG GATTACAGCAA	YHYVTITLDLQQ
Heparin-binding peptide	TATCCGACCCAGCGTGC GCGCTAT CAATGGGTGCGTTGCAACCCG	YPTQRARYQWVRCNP
TGF- β 1 peptide	TATTACGTGGGCCGTAAACCGAA A	YYVGRKPK
6xHis tag	CATCACCATCACCATCAC	HHHHHH

Table S2. Primer sequences for the synthesis of the DNA sequence encoding the “TGF- β 1 peptide” by PCR.

Primer name	Sequence (5'-3')
TGF- β 1 peptide forward	GACTGTCATATGGAAAGCCTGTTGCCGGTGAGGAGCGGT GTGGGCTATTACGTGGGCCGTAAACCGAAA
TGF- β 1 peptide reverse	AACCACCTCGAGCTGAAGAGCTGTGGTCTGGTGCCTTTCC GTTTACGGCCCACGTAATA

Table S3. Composition of PCR reaction for the synthesis of the DNA sequence encoding the “TGF- β 1 peptide”.

Reagent	Final concentration
Forward primer	500 nM
Reverse primer	500 nM
Thermopol Buffer	1X (contains 2mM MgSO ₄)
MgSO ₄	10 mM
dNTP mix	3 mM each
Deep Vent DNA polymerase	6 U
Sterile ddH ₂ O	Up to 50 μ L

Table S4. Conditions of PCR reaction for the synthesis of the DNA sequence encoding the “TGF- β 1 peptide”.

Stage	Temperature (°C)	Duration (min)	Number of cycles
Initial denaturation	95	3	1
Denaturation	95	1	20
Annealing	65	1	
	59	1	
Extension	72	1	
Final extension	72	5	1
Hold	4	∞	

Table S5. Primer sequences for real-time PCR.

Primer	Nucleotide sequence (5'to3')
<i>GAPDH</i> forward	GCACCGTCAAGGCTGAGAAC
<i>GAPDH</i> reverse	TGGTGAAGACGCCAGTGGA
<i>RPLPO</i> forward	AATGTGGGCTCCAAGCAGAT
<i>RPLPO</i> reverse	TGAGGCAGCAGTTTCTCCAG
<i>SOX9</i> forward	GGCAAGCTCTGGAGACTTCTG
<i>SOX9</i> reverse	CCCGTTCTTCACCGACTTCC
<i>COL2A1</i> forward	AGAATCCATCTGAGAATATGC
<i>COL2A1</i> reverse	CCTCTTACTGCTATACCTTTAC

<i>ACAN</i> forward	GGAAGGGAGGGGAACCATTG
<i>ACAN</i> reverse	TGATGGCTGTCCACTGACAC
<i>TGFBR1A</i> forward	AGCAGCAGACAATAAAGACAATGG
<i>TGFBR1A</i> reverse	TGTGAAGATGGGCAAGACCG
<i>TGFBR2</i> forward	CTGTAATGCAGTGGGAGAAGTAAAA
<i>TGFBR2</i> reverse	AATTTCTGGTCGCCCTCGAT
<i>COL1A1</i> forward	CAGTGTGGCCCAGAAGAACT
<i>COL1A1</i> reverse	CCGCCATACTCGAACTGGAAT
<i>Osteocalcin</i> forward	CGGTGCAGAGTCCAGCAAA
<i>Osteocalcin</i> reverse	GGTAGCGCCTGGGTCTCTTC
<i>MMP9</i> forward	TTCCAGTACCGAGAGAAAGCCTAT
<i>MMP9</i> reverse	GGTCACGTAGCCCACTTGGT
<i>MMP13</i> forward	AAGGAGCATGGCGACTTCT
<i>MMP13</i> reverse	TGGCCCAGGAGGAAAAGC
<i>COL10A1</i> forward	AGAATCCATCTGAGAATATGC
<i>COL10A1</i> reverse	CCTCTTACTGCTATACCTTTAC
<i>PCNA</i> forward	TTAAATTGTCACAGACAAGTAATGTCTG
<i>PCNA</i> reverse	TGGCTTTTGTAAGAAGTTCAGGTAC
<i>BAX</i> forward	GGGCCCACCAGCTCTGA
<i>BAX</i> reverse	CCTGCTCGATCCTGGATGA
<i>BCL2</i> forward	CTTGACAGAGGATCATGCTGTAC
<i>BCL2</i> reverse	GGATGCTTTATTTCATGAGGC
<i>IL1b</i> forward	ATCACTGAACTGCACGCTCC
<i>IL1b</i> reverse	TTGTTCTCCATATCCTGTCCC
<i>TNFa</i> forward	CCTCTCTCAATCAGCCCTCTG
<i>TNFa</i> reverse	GAGGACCTGGGAGTAGATGAG

Sequences



Nucleotide sequence (2952 bp):

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Amino acid sequence (982 aa):

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