

Supporting Information

Injectable Hydrogel Mediated Delivery of Gene-Engineered Adipose-Derived Stem Cells for Enhanced Osteoarthritis Treatment

Wei Yu, Bin Hu, Kofi Oti Boakye-Yiadom, William Ho, Qijing Chen, Xiaoyang Xu*, and Xue-Qing Zhang*

Gene Name	Primer Sequence
GAPDH	forward, 5'-ACCACAGTCCATGCCATC-3' reverse, 5'-TCCACCACCCTGTTGCTGTA -3'
Collagen II	forward, 5'-TCCTAAGGGTGCCAATGGTGA-3' reverse, 5'-AGGACCAACTTTGCCTTGAGGAC-3'
Aggrecan	forward, 5'-TCCGCTGGTCTGATGGACAC-3' reverse, 5'-CCAGATCATCACTACGCAGTCCTC-3'
TNF- α	forward, 5'-CACGAAAGCATGATCCGAG-3' reverse, 5'-TCCTCCTTGTGGGACCGAT-3'

Table S1. Primer sequences used for qRT-PCR.

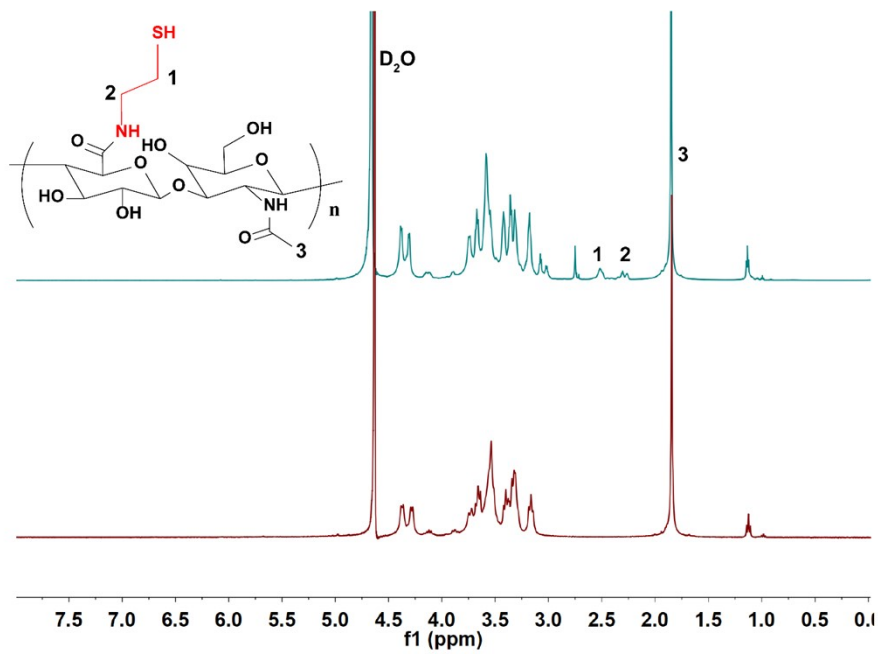


Fig. S1. The ¹H NMR spectra of HA and HA-SH.

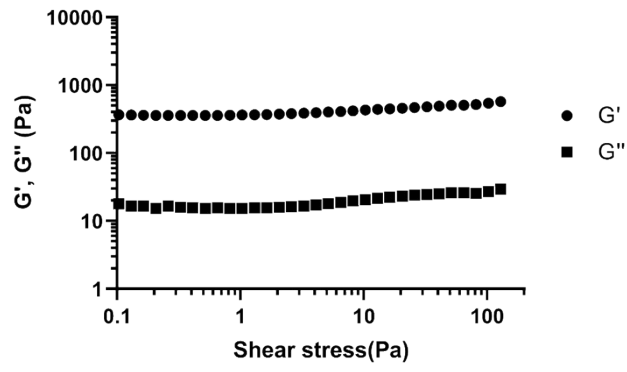


Fig. S2. The linear viscoelastic region of 2-HA-Col hydrogel at 37°C.

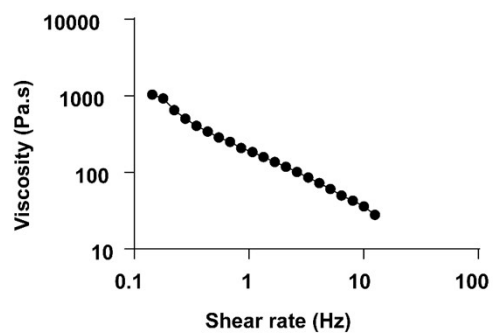


Fig. S3. The shear-thinning property of 2-HA-Col hydrogel at 37 °C.

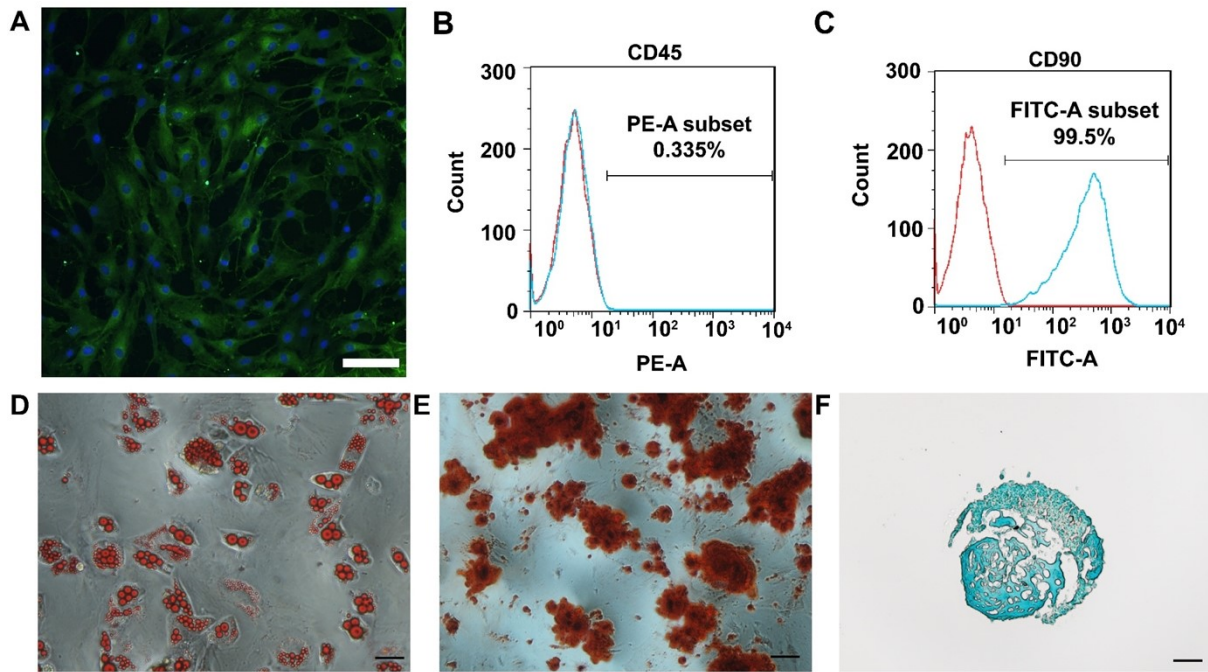


Fig. S4. Identification of the isolated ADSCs. A) Morphology of ADSCs with DiO staining the cell membrane and Hoechst staining the nuclei. Scale bar 200 μm . B, C) Phenotypic analysis via flow cytometry. D-F) Identification of multiple differentiation ability of ADSCs. The differentiation status of ADSCs into D) adipogenic, E) osteogenic, and F) chondrogenic lineages was identified by staining with oil red O, Alizarin red, and Alcian blue. Scale bar 200 μm .

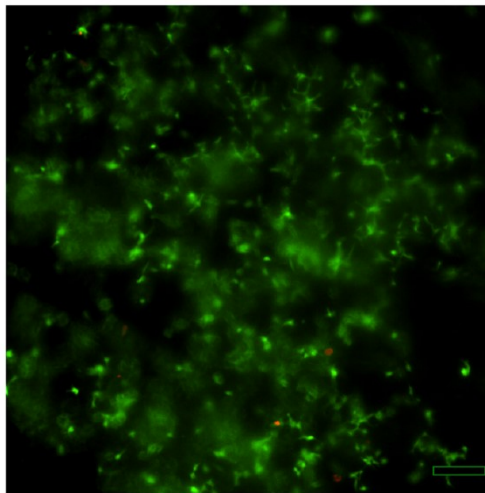


Fig. S5. Live/dead staining of ADSCs after 3 days of culture in 2-HA-Col hydrogel. Scale bar 200 μm .

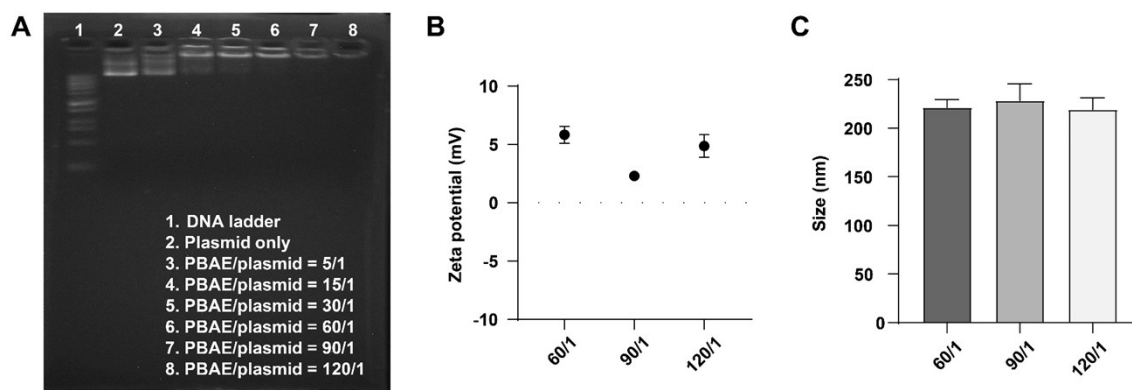


Fig. S6. A) Agarose gel electrophoresis retardation of the PBAE/pEGFP-TGF- β 1 nanocomplexes formulated at different polymer/plasmid weight ratios. The pEGFP-TGF- β 1 was effectively condensed when formulated with PBAE at or above a weight ratio of 60/1. B) Zeta potential and C) size of PBAE/pEGFP-TGF- β 1 nanocomplexes with different weight ratios. Data are presented as the mean \pm standard deviation ($n = 3$).

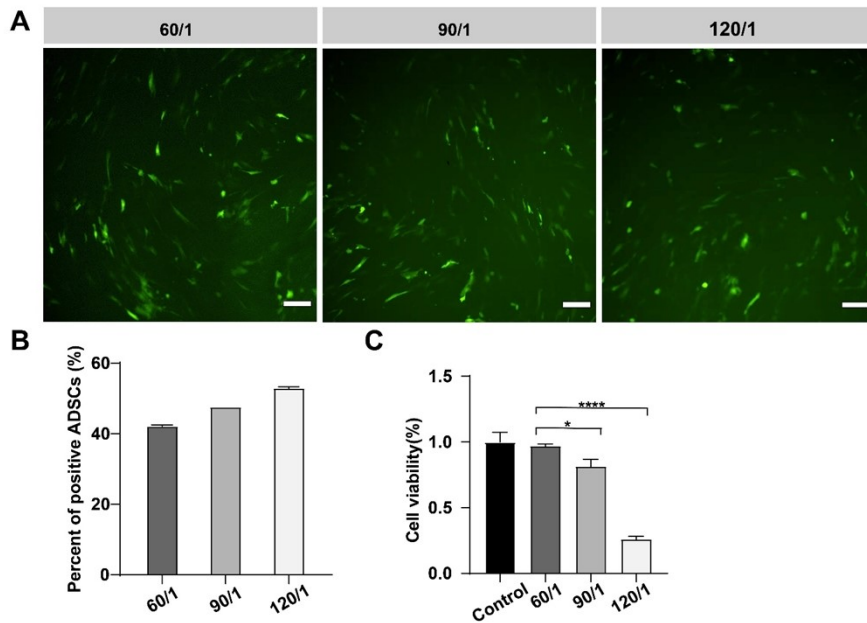


Fig. S7. A) The EGFP expression of ADSCs transfected with different PBAE/pEGFP-TGF-β1 nanocomplexes. The weight ratio of PBAE to pEGFP-TGF-β1 was increased from 60/1 to 120/1. Scale bar 200 μm. B) The percentage of EGFP positive cells increased with an increase in the polymer/plasmid weight ratio. C) Cell viability test of different PBAE/pEGFP-TGF-β1 nanocomplexes. Nanocomplexes formulated at above weight ratio of 120 resulted in a significant decrease in cell viability. Data are presented as the mean ± standard deviation ($n = 3$), * $p < 0.05$, **** $p < 0.0001$.

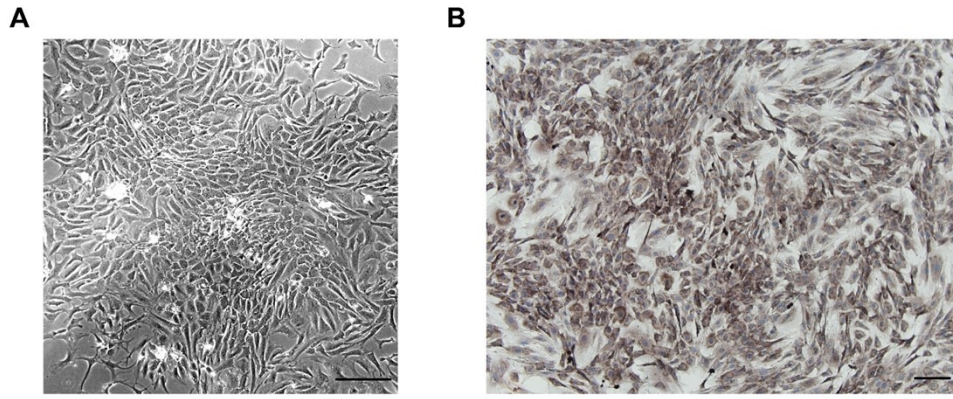


Fig. S8. A) Morphology of chondrocytes. B) Col II immunocytochemistry of chondrocytes. Scale bar 200 μm .



Fig. S9. The procedure of ACLT/MMx surgery.