

Supporting Information

Sulfonate Hydrogel-siRNA Conjugate Facilitates Osteogenic Differentiation of Mesenchymal Stem Cells by Controlled Gene Silencing and Activation of BMP Signaling

Soyon Kim¹, Jiabing Fan¹, Chung-Sung Lee¹, Chen Chen¹, and Min Lee^{1,2*}

¹Division of Advanced Prosthodontics, University of California, Los Angeles, USA

²Department of Bioengineering, University of California, Los Angeles, USA

*Corresponding author

Min Lee, PhD

UCLA School of Dentistry

10833 Le Conte Avenue, CHS 23-088F

Los Angeles, CA 90095-1668

Email: leemin@ucla.edu

Phone: +1-310-825-6674

Fax: +1-310-825-6345

Table S1. Primer information for qRT-PCR

Gene	Forward	Reverse
<i>GAPDH</i>	AGGTCGGTGTGAACGGATTTG	TGTAGACCATGTAGTTGAGGTCA
<i>Noggin</i>	GCCAGCACTATCTACACATCC	GCGT CTCGTTCAGATCCTTCTC
<i>Runx2</i>	CGGTCTCCTTCCAGGATGGT	GCTTCCGTCAAGCGTCAACA
<i>ALP</i>	GTTGCCAAGCTGGGAAGAACAC	CCCACCCGCTATTCCAAAC
<i>OCN</i>	GGGAGACAACAGGGAGGAAAC	CAGGCTTCCTGCCAGTACCT
<i>Gremlin</i>	CGCAGCCGCACCATCCTCAA	CGAGACCCGGCATTGAGC
<i>Chordin</i>	TCAAGTTACGCTTCTGTCTCCTA	AGGATGTTGCCTGTGGGATCT
<i>BMP-2</i>	CGGACTGCGGTCTCCTAA	GGGAAAGCAGCAACACTAGA
<i>Smad5</i>	CCAGCCGTGAAGCGATTGT	CTCCTCCATAGCACCCCTCT
<i>Id1</i>	AACGGCGAGATCAGTGCCTT	AACGGCGAGATCAGTGCCTT

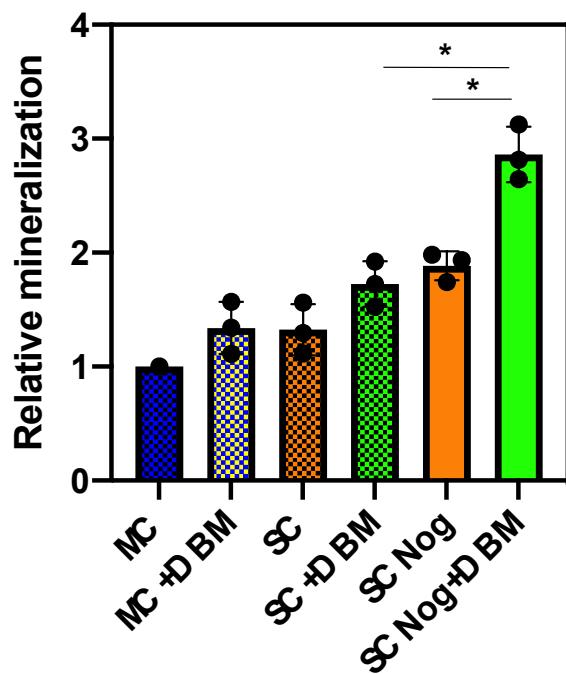


Figure S1. Relative mineralization of DBM incorporated hydrogels with sulfoacetic acid and noggin siRNA conjugation at day 21. * $p < 0.05$.

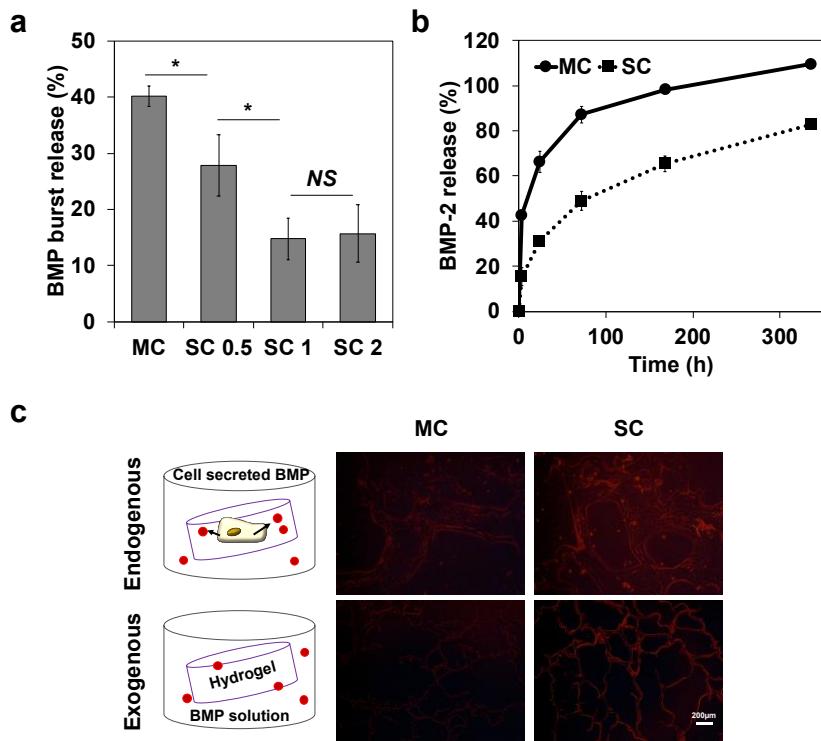


Figure S2. a) Initial burst release of BMP with different sulfoacetic concentrations. * $p < 0.05$. b) BMP-2 release profile of MC and SC hydrogels for 14 days. c) Exogenous and endogenous BMP sequestering images obtained by immunostaining.

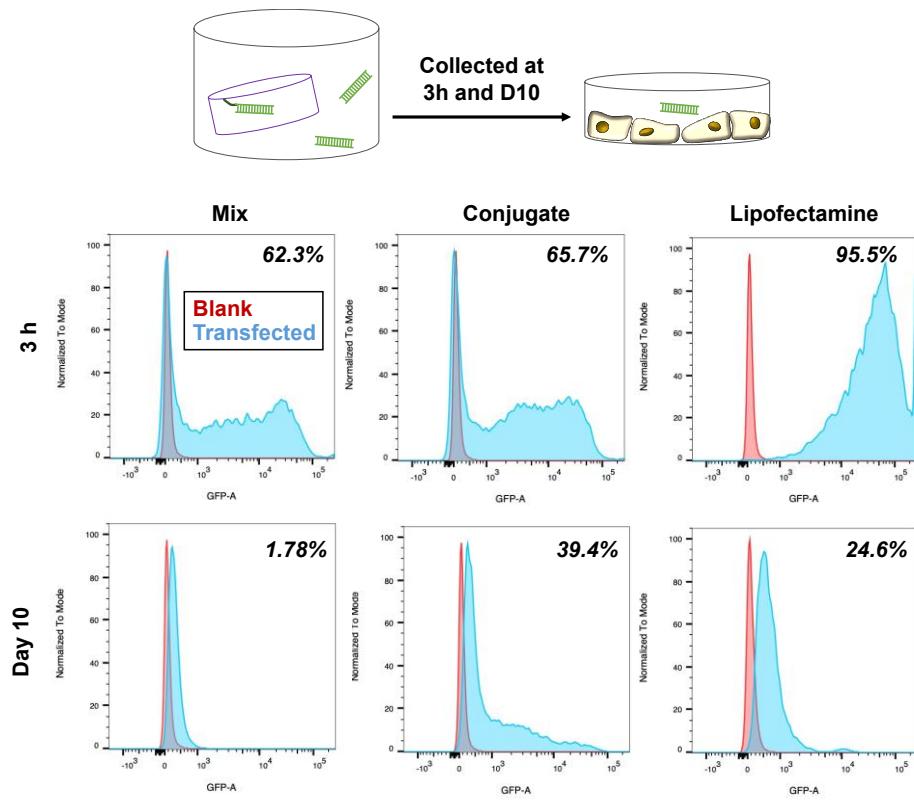


Figure S3. 2D transfection experiments by collecting the releasates of siRNA from hydrogels at 3 h and 10 days and flow cytometry histogram. Transfection results of siRNA delivered by lipofectamine was provided as a control.

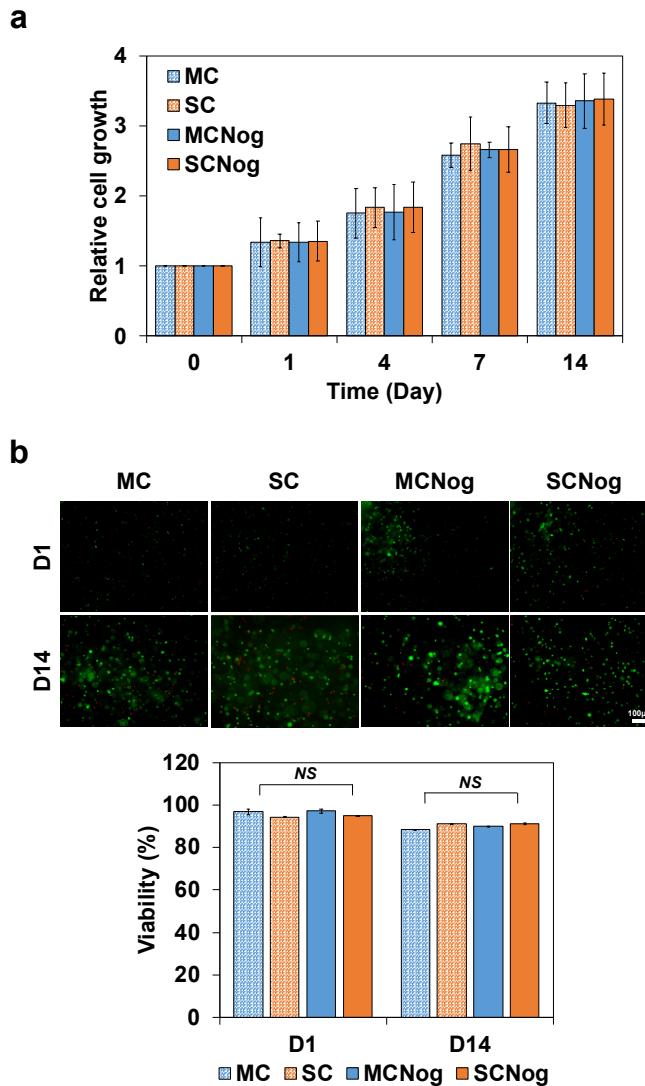


Figure S4. a) Relative cell growth of BMSCs encapsulated in hydrogel-siRNA conjugate measured by alamarBlue assay. b) Representative live/dead images of BMSCs encapsulated in hydrogel-siRNA conjugate and the quantified viability analyzed by ImageJ. NS = not significant.

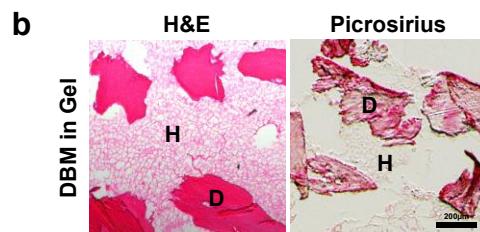
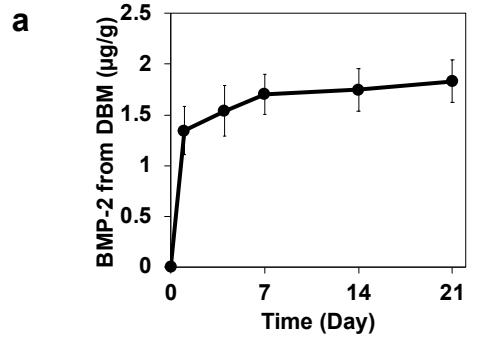


Figure S5. (a) BMP-2 release profile from DBM for 14 days quantified by BMP-2 ELISA. (b) Histology of DBM incorporated hydrogel with H&E and picosirius staining. H indicated hydrogel and D indicated DBM. H&E staining exhibited the localization of DBM and the red color stained from picosirius staining pointed out the existence of collagen in DBM, while hydrogel part was not stained.