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Supplementary data

Nanoclay-Functionalized 3D Nanofibrous Scaffolds Promote Bone Regeneration

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ALP and Alizarin Red S (ARS) staining

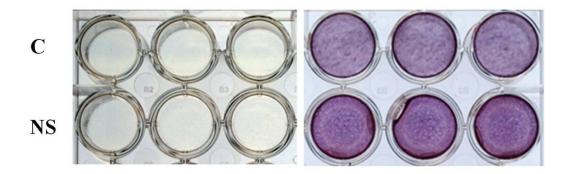


Figure S1. The effects of NS (50 μ g/mL) on the ALP activity (on day 7, left panel) and mineralization (on day 21, right panel) of MC3T3-E1 cells (mouse pre-osteoblast cells).

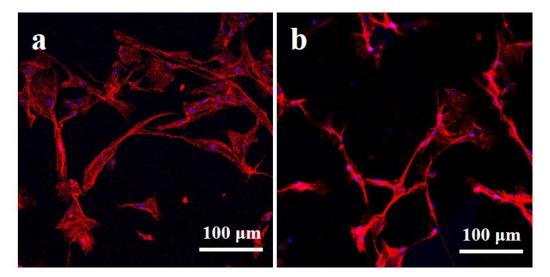


Figure S2. Confocal images of hMSCs cells on (a) GF and (b) GF/NS scaffolds.

Wnt/β-Catenin signaling related gene expression

The housekeeping gene glyceraldehyde 3-phosphate dehydrogenase (GAPDH) was used for normalization and the primer sequences of AXIN2 and β -Catenin genes for real time PCR were summarized in Table 1. At least three replicates were performed on each sample.

Genes	Forward primer sequences	Reverse primer sequences
GAPDH	5' TCAGCAATGCCTCCTGCAC 3'	5'TCTGGGTGGCAGTGATGC3
		,
AXIN2	5'CCCCAAAGCAGCGGTGC3'	5'GCGTGGACACCTGCCAG3'
β-Catenin	5'GCTACTGTTGGATTGATTCGAAATC	5'CCCTGCTCACGCAAAGT3'
	3'	

Table S1. Real-time PCR primer sequences.

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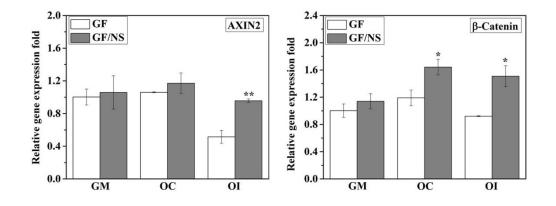


Figure S3. The relative gene expression levels of AXIN2 (left) and β -Catenin in hMSCs cultured on GF and GF/NS scaffolds in growth medium (GM), osteoconductive medium (OC) and osteoinductive medium (OI). Data are expressed as mean \pm SD (n = 3). *p < 0.05