Electronic Supplementary Material (ESI) for Biomaterials Science.

Supporting Information

Dynamic matrix potentiates mesenchymal stromal cells paracrine function *via* effective mechanical dose

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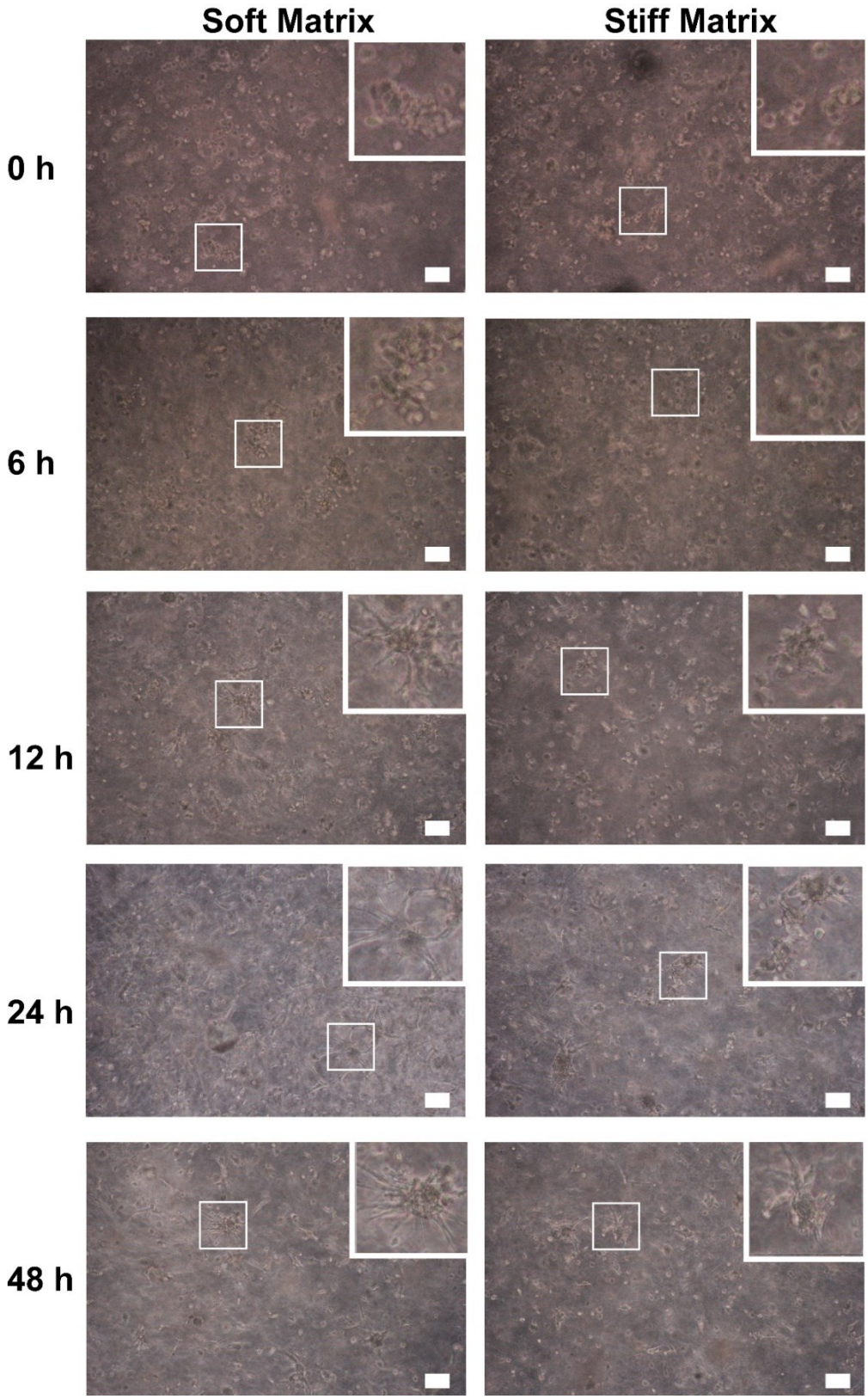

Figure S1. Spreading rate of MSCs in soft or stiffening hydrogel (scale bar = 100 μm).

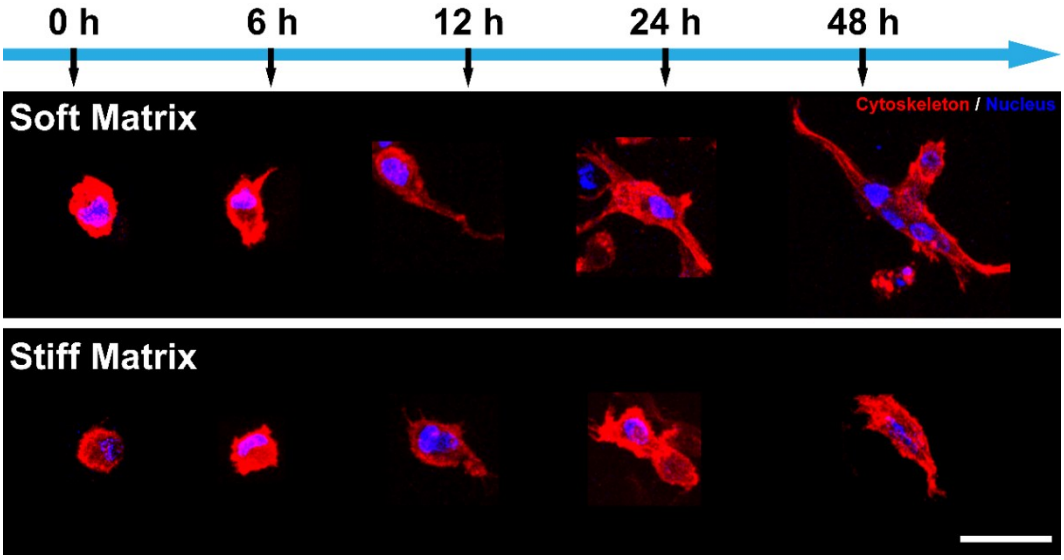

Figure S2. Spreading rate of MSCs in soft or stiffening hydrogel (red: cytoskeleton; blue: nuclei, scale bar = 20 μm).

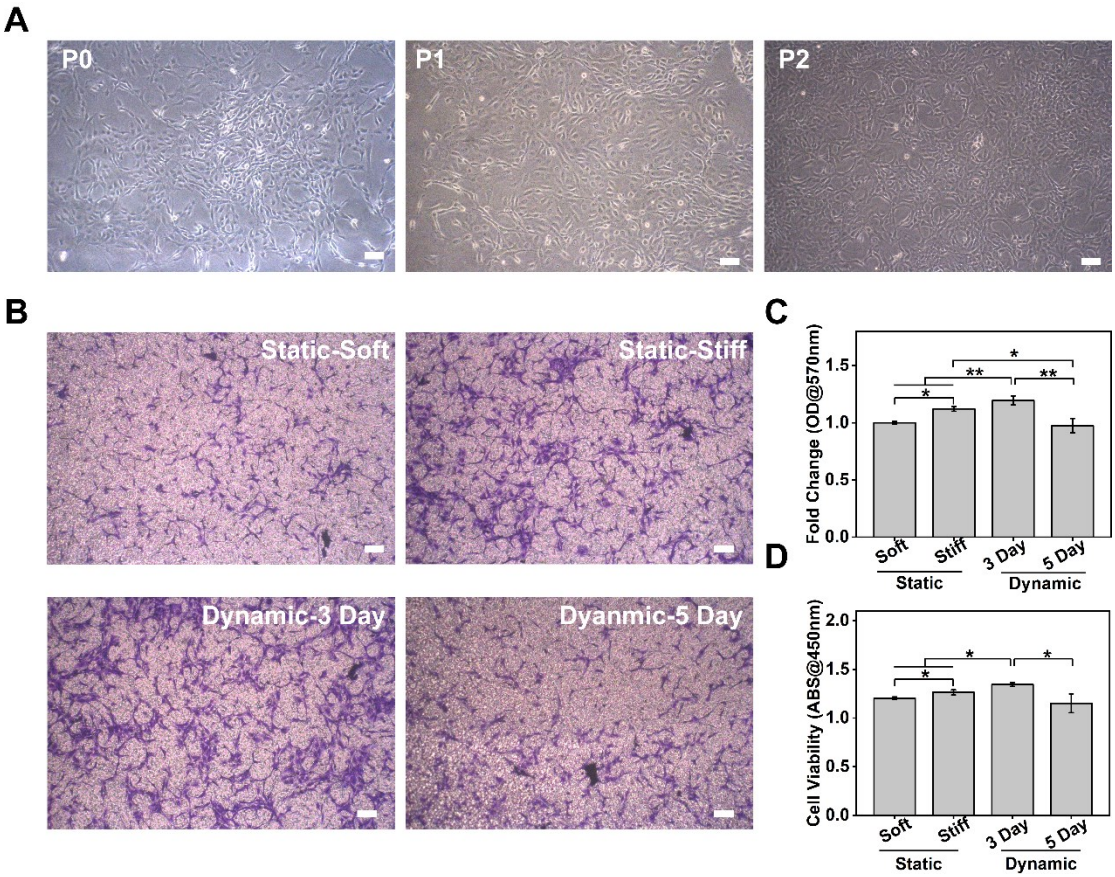

Figure S3. The effects of conditioned medium (CM) of MSCs on (A) The morphology of SD rat thoracic aorta endothelial cells; (B - D) the effects of conditioned medium (CM) of MSCs on SD rat thoracic aorta endothelial cells (B & C) migration and (D) proliferation. (Scale bar = 100 μm, n = 3, *$p < 0.05$, **$p < 0.01$) All data was shown as mean ± SD.

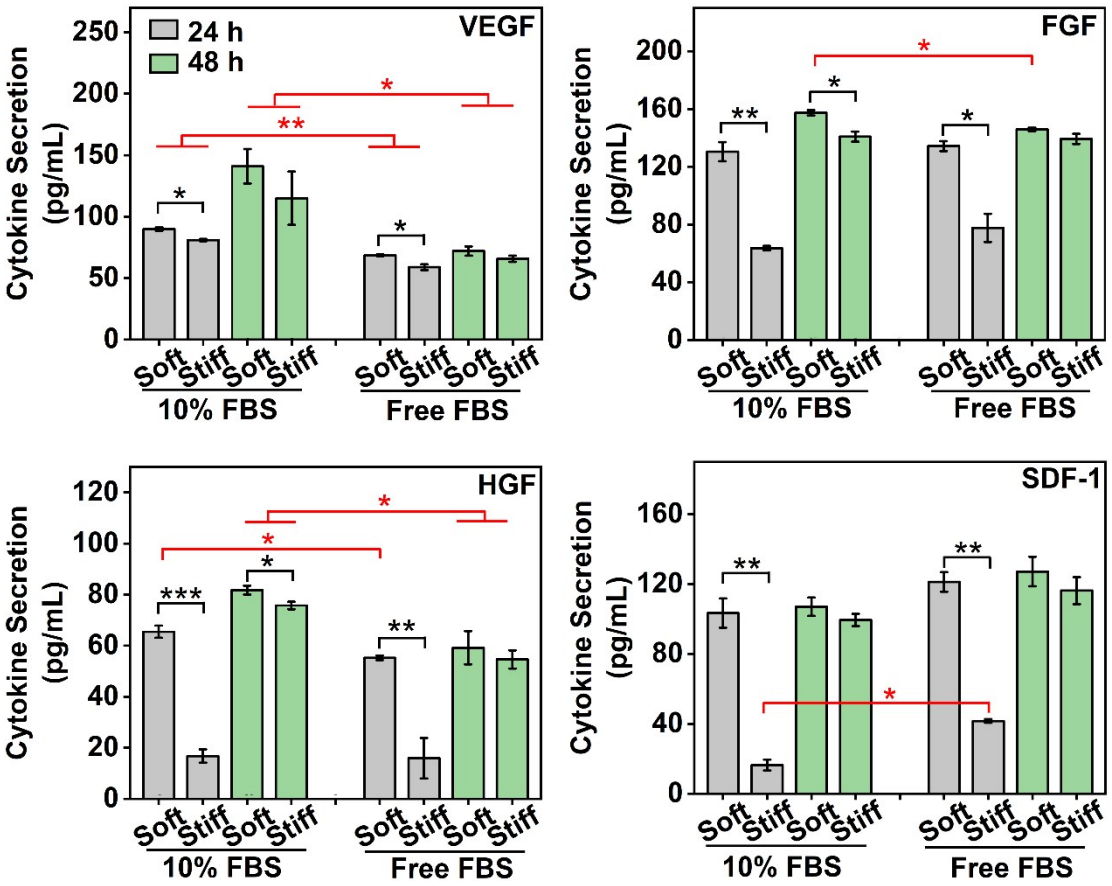

Figure S4. Paracrine factors of MSCs in soft or stiffening hydrogel after 24 h / 48 h under 10 % FBS medium or free FBS medium (n=3, *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$). All data was shown as mean ± SD.

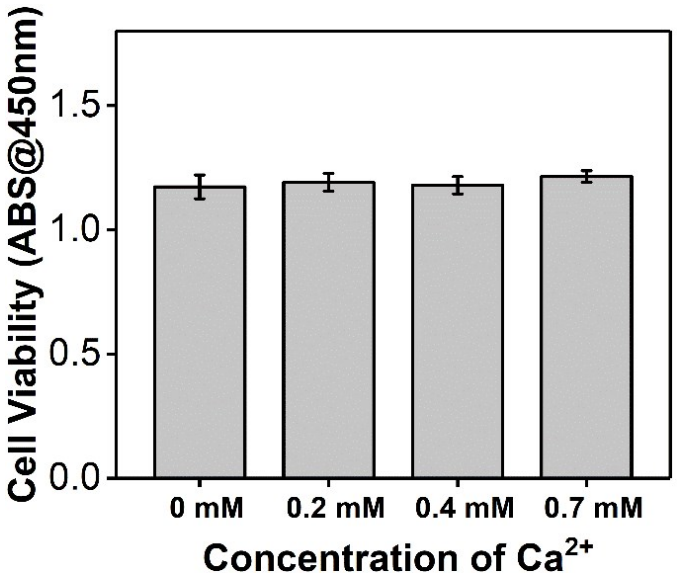

Figure S5. Cell viability of MSCs which treated with different Ca^{2+}. All data was shown as mean ± SD.

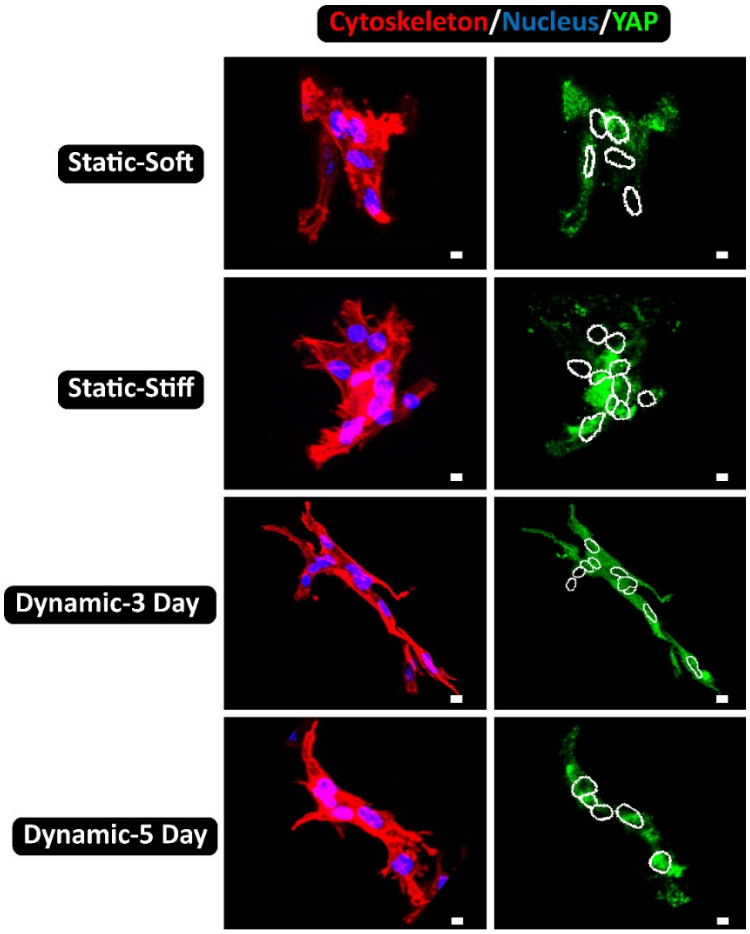

Figure S6. The morphology or YAP location of cluster cells in hydrogel.

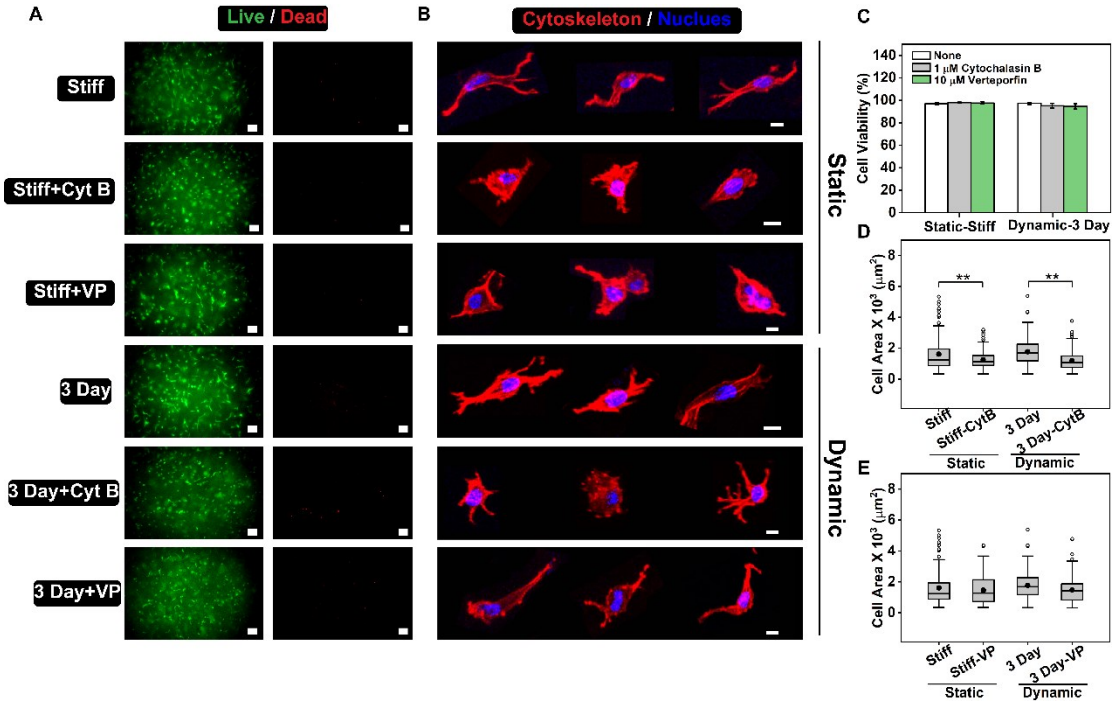

Figure S7. Cell morphology and viability of MSCs cultured in SA hydrogels for 7 days with inhibitors treatment. (A) Live / dead staining of MSCs (scale bar = 100 μm, green - live cells, red - dead cells). (B) The cytoskeleton staining of MSCs cultured in hydrogels (scale bar = 5 μm, red - cytoskeleton, blue - nucleus). (C) Cell viability of MSCs (calculated 5 scopes in every sample). (D & E) Cell shape index of single MSC (n = 60, $^{**}p < 0.01$). All data was shown as mean ± SD.

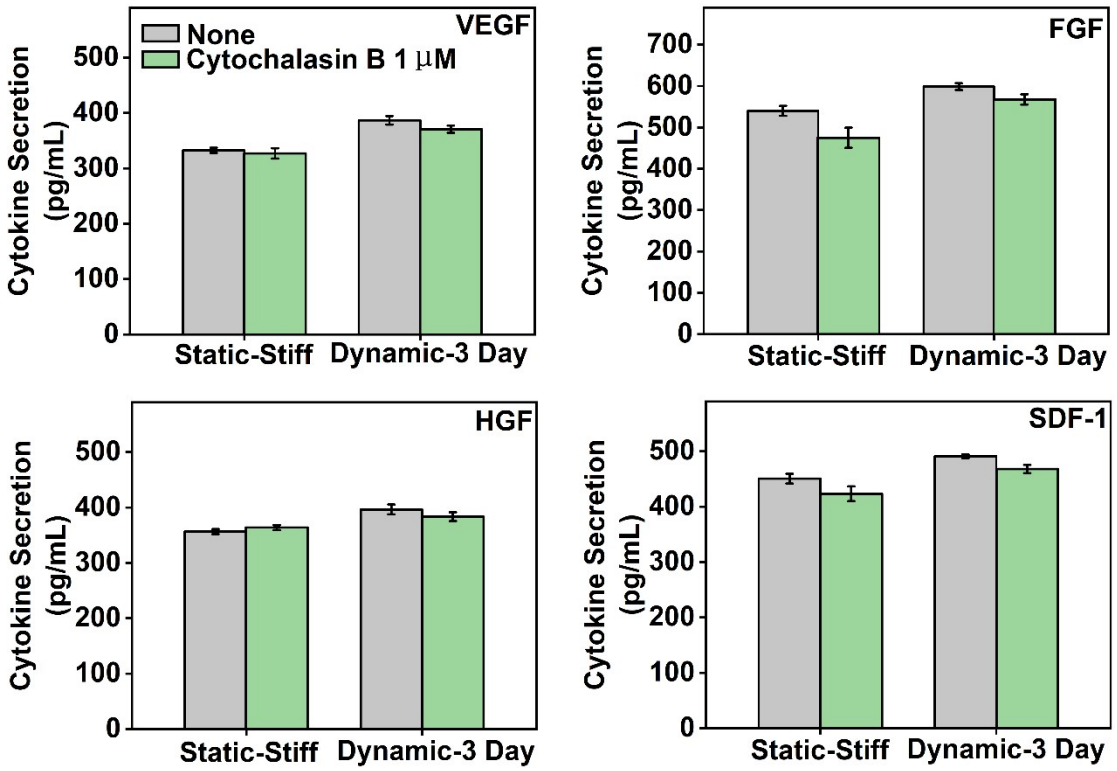

Figure S8. Inhibited the polymerization of F-actin cannot enough change the paracrine cytokines of MSCs in hydrogels (n = 3). All data was shown as mean ± SD.

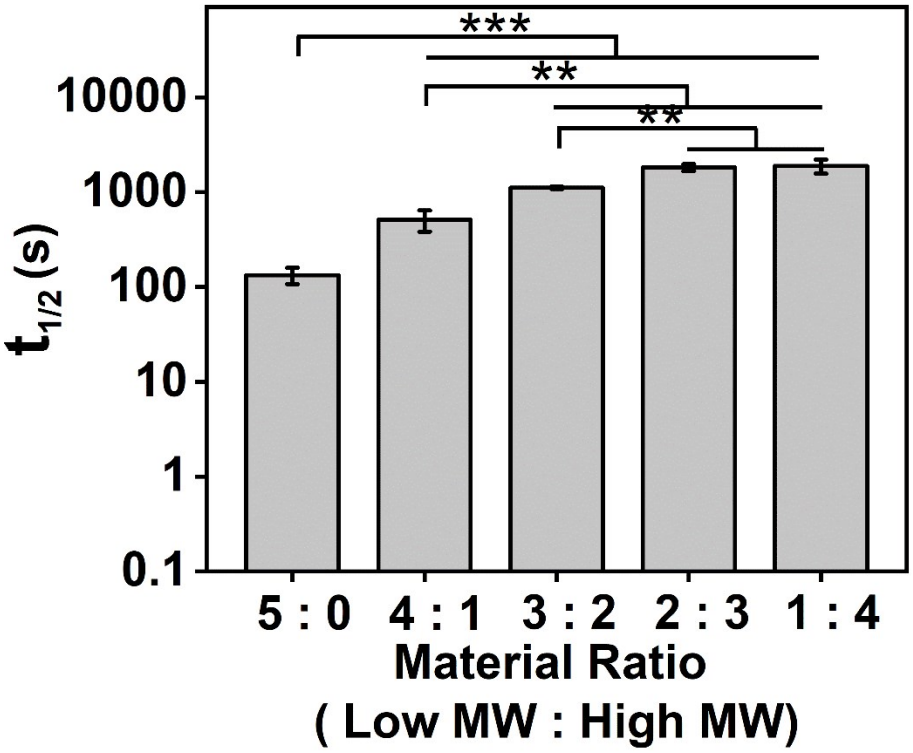

Figure S9. MW of SA can affect the stress relaxation of SA hydrogels (n = 3, $^{**}p < 0.01$, $^{***}p < 0.001$). All data was shown as mean ± SD.

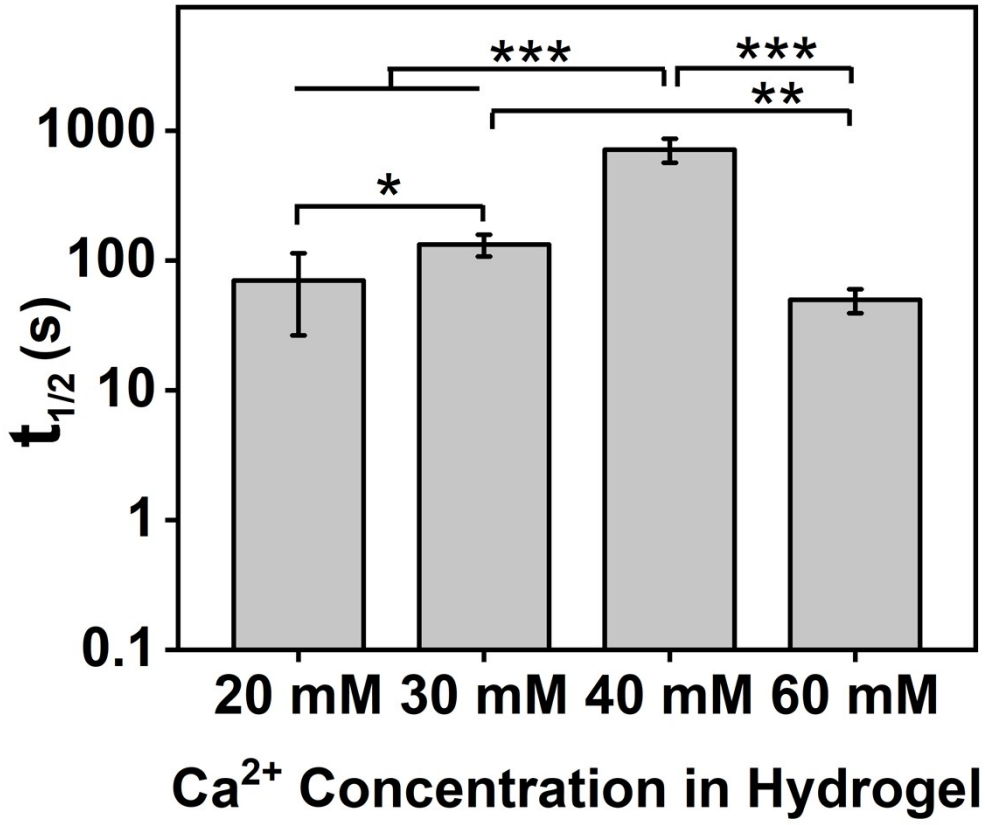

Figure S10. Correlation between Ca^{2+} concentration and stress relaxation (n = 3, *p < 0.05, **p < 0.01, ***p < 0.001). All data was shown as mean ± SD.

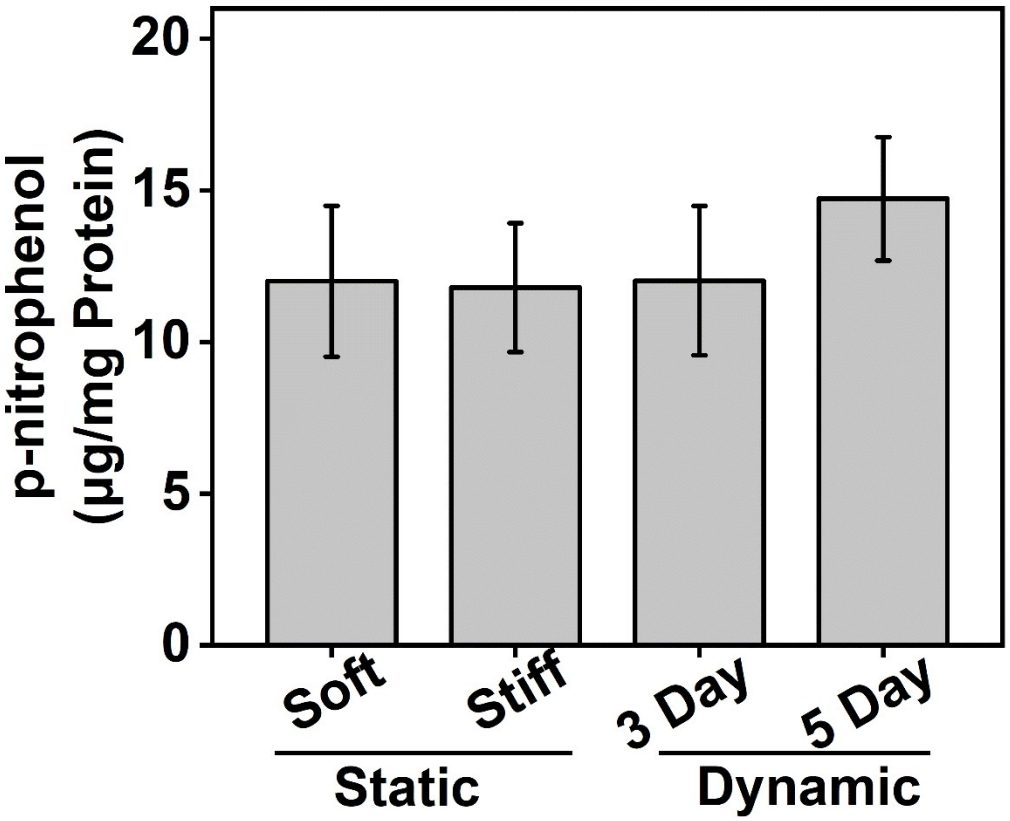

Figure S11. The ALP expression of MSCs in hydrogels after culturing 7 days (n = 5). All data was shown as mean ± SD.

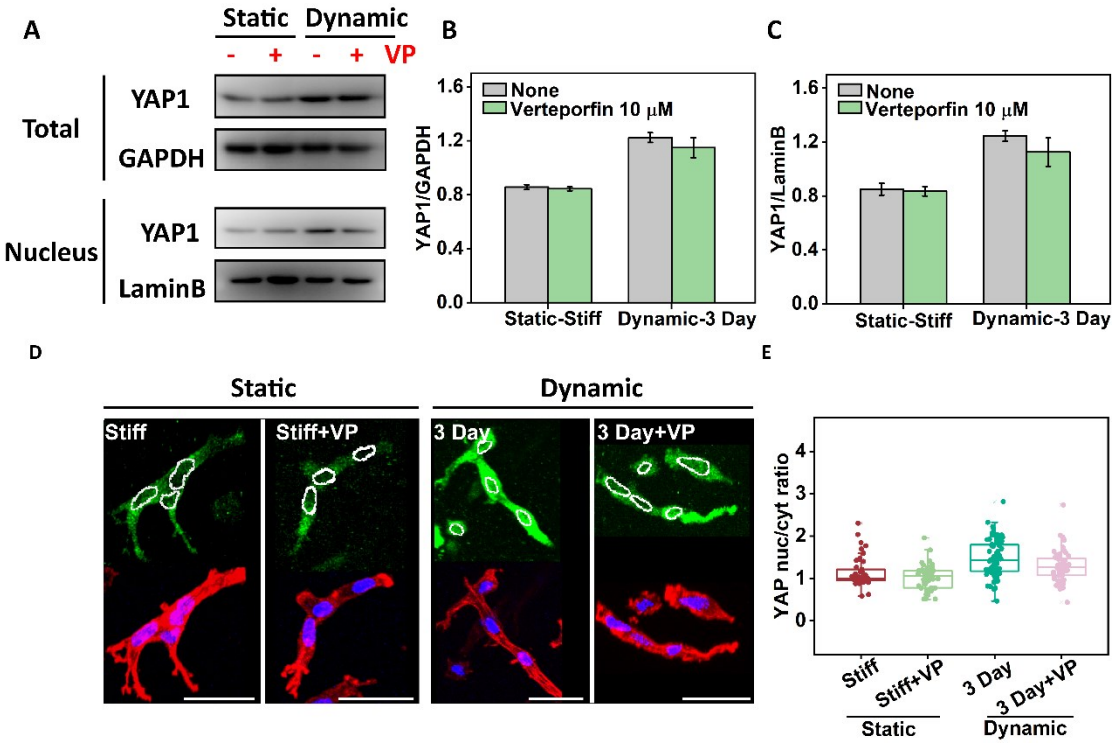

Figure S12. The effects of verteporfin on the expression and activation of YAP. (A-C) Western blotting analysis of the expression of YAP in MSCs in hydrogel (n = 3). (D-E) Immunofluorescent staining of YAP (green), cytoskeleton (red) and nuclei (blue) in MSCs in hydrogel (n =60 single MSCs, scale bar = 30 μm).

Table S1. The sequence of primers used in RT-PCR.

Gene	5’-3’	Primers
VEGFA	Sense	5’-CTTCAAGCCGTCCTGTGTGC-3'
	Anti-sense	5'-GGCTCACAGTGATTTTCTGGCT-3'
FGF	Sense	5’-AAGAGCGACCCACACGTCAAA-3'
	Anti-sense	5’-CTGCCCAGTTCGTTTCAGTGC-3'
HGF	Sense	5'-CCCCCATGAACACAGCTTTTTG-3'
	Anti-sense	5'-GCTTTCACCGTTGCAGGTCA-3'
IGF	Sense	5’-CTGGGACTTCTGAGTCTTGG-3'
	Anti-sense	5’-GGGCATTGTGGATGAGTG-3'
SDF-1	Sense	5’-ATGCCCCTGCCGATTCTTT-3'
	Anti-sense	5'-GTTGTTGCTTTTCAGCCTTGC-3'
GAPDH	Sense	5’-TGCGTTGTTGCGGTCCAC-3’
	Anti-sense	5’-GGCATTGCTCTCAATGACAA-3’