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Supplementary Materials for

In vivo priming of human mesenchymal stem cells with hepatocyte growth factor–engineered mesenchymal stem cells promotes therapeutic potentialfor cardiac repair

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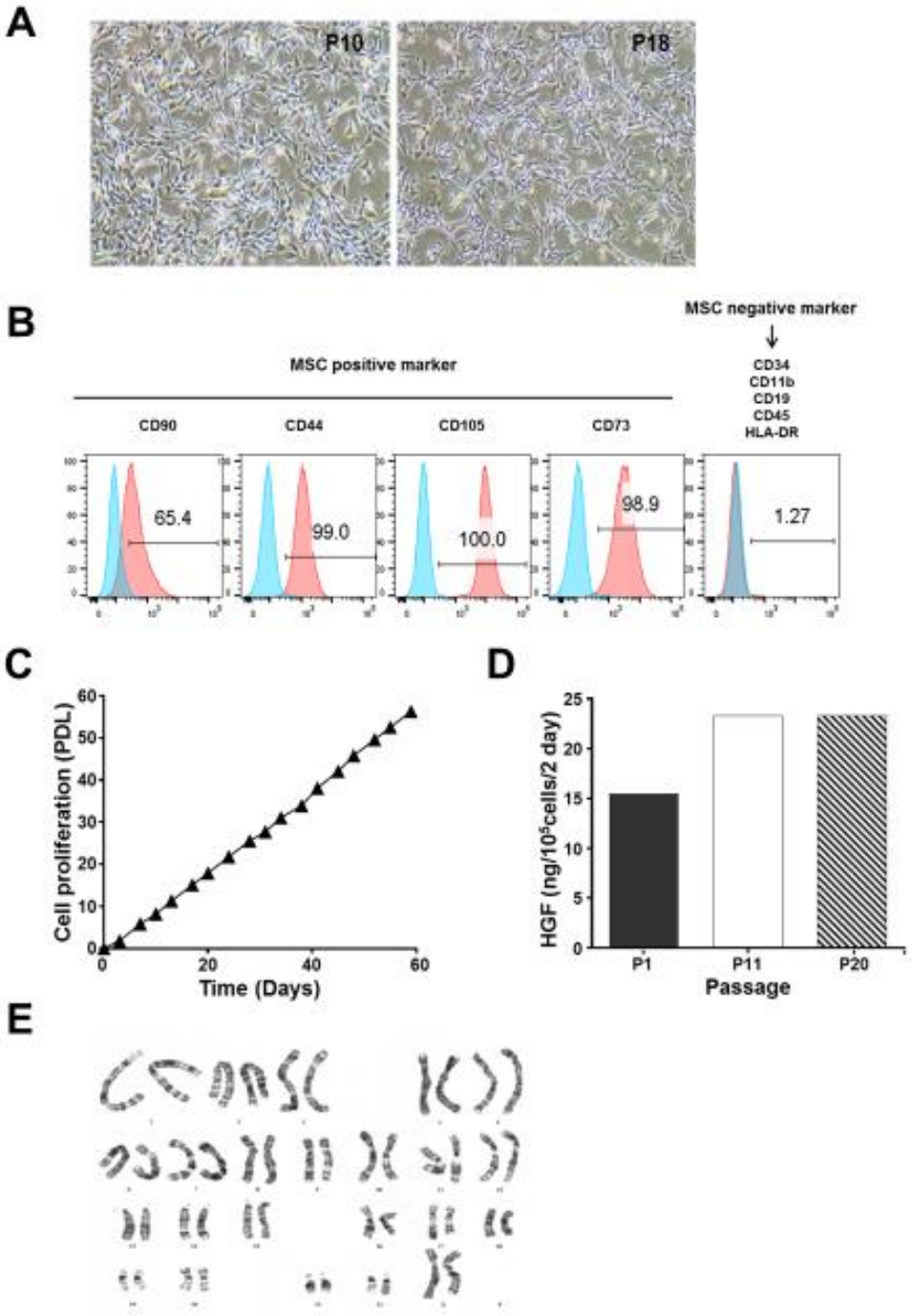


Fig. S1. Characterization of HGF-eMSC. (**A**) Morphology of human bone marrow derived MSCs. (**B**) Flow cytometry analyses show that HGF-eMSC express specific markers for MSCs such as CD90, CD44, CD105 and CD73. n=3 per group (**C**) Cell proliferation rate of HGF-eMSC. n=3 per group (**D**) HGF secretion of distinct passages of HGF-eMSC measured by ELISA. n=3 per group. (**E**) The results from karyotyping with HGF-MSCs. HGF-eMSC: Engineered hepatocyte growth factor expressing MSCs

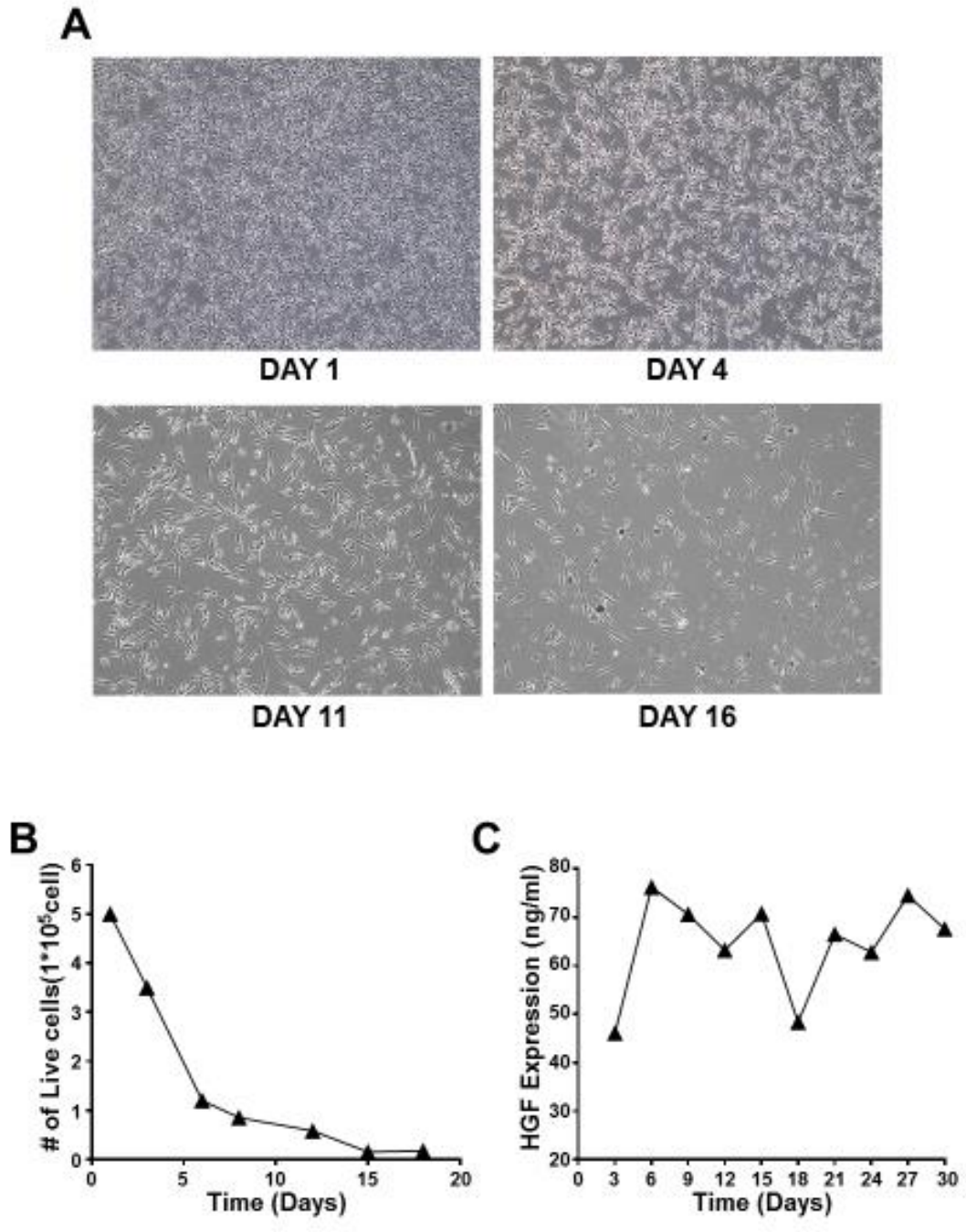


Fig. S2. Cellular characteristics of irradiated HGF-eMSC. (**A**) Morphology of irradiated HGF-eMSCs. (**B**) Cell proliferation rate of irradiated HGF-eMSC. n=3 per group. (C) HGF secretion of irradiated HGF-eMSC over times measured by ELISA. n=3 per group. HGF-eMSC: Engineered hepatocyte growth factor expressing MSCs.

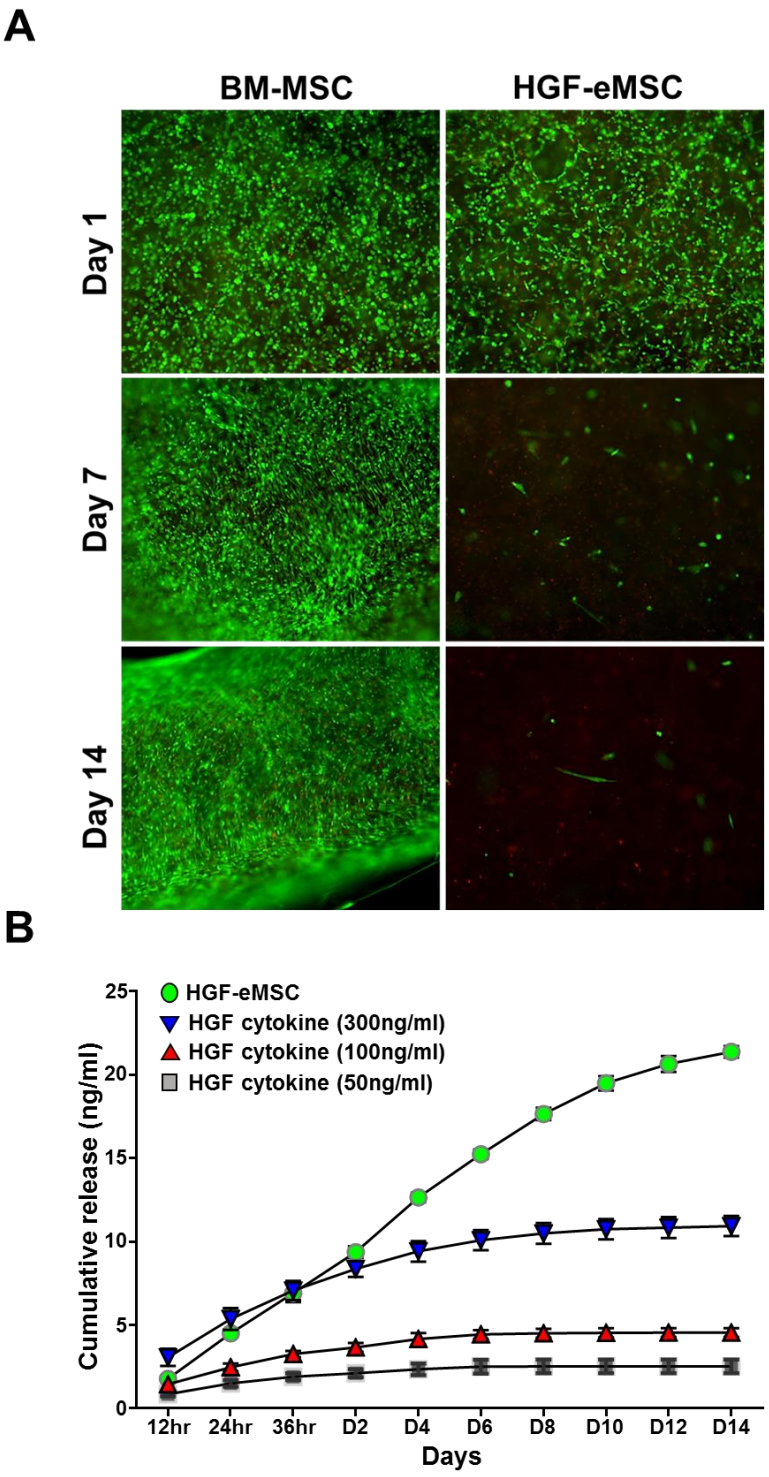


Fig. S3. Cellular behavior of irradiated HGF-eMSC within the hdECM patch. (**A**) The results from Live & Dead staining of HGF-eMSC within the hdECM patch indicating that irradiated HGF-eMSC are not remain viable within the the hdECM patch over time. (**B**) Cumulative release of HGF cytokine from irradiated HGF-eMSC encapsulated in hdECM.

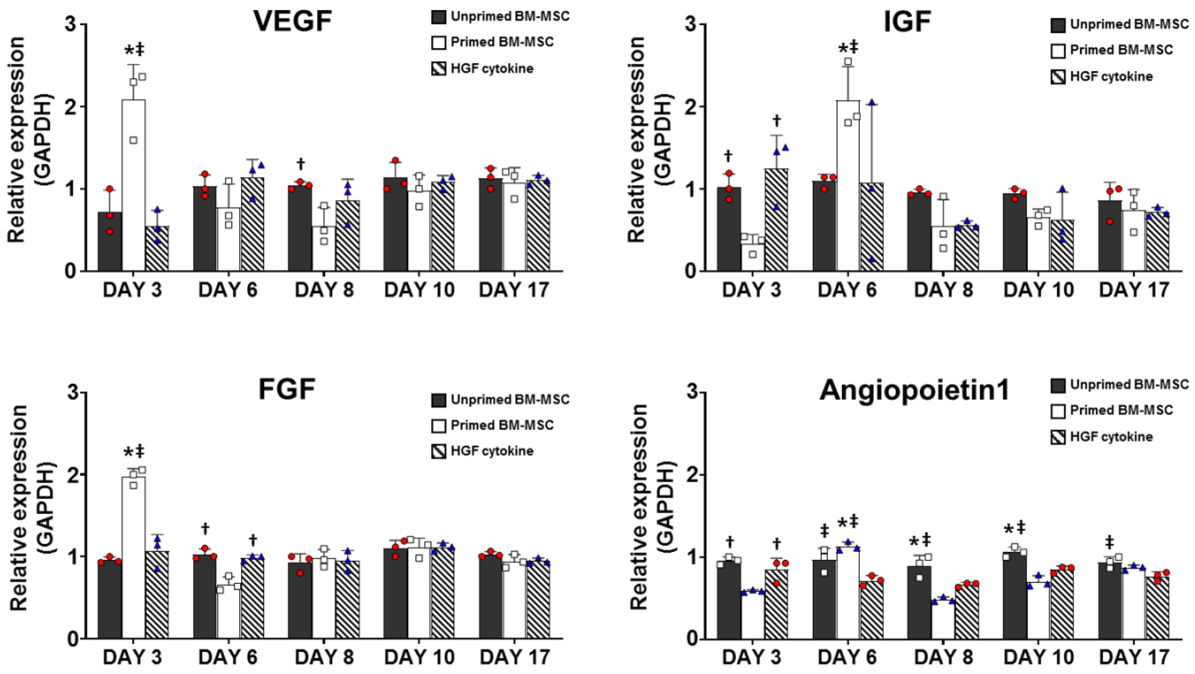


Fig. S4. Investigation of the duration of priming effect in BM-MSC by HGF-eMSC. To determine how long the effect of priming with HGFe-MSCs persists in BM-MSCs, the BM-MSCs and HGF-eMSCs were co-cultured for 3 days for priming purpose. After 3 days of co-culture, the BM-MSCs were continuously cultured and were harvested in different time points (day 3, 6, 8, 10 and 17). *p <0.05 compared to unprimed BM-MSC group, †p < 0.05 compared to primed BM-MSC group; ‡p < 0.05 compared to HGF Cytokine primed BM-MSC group.

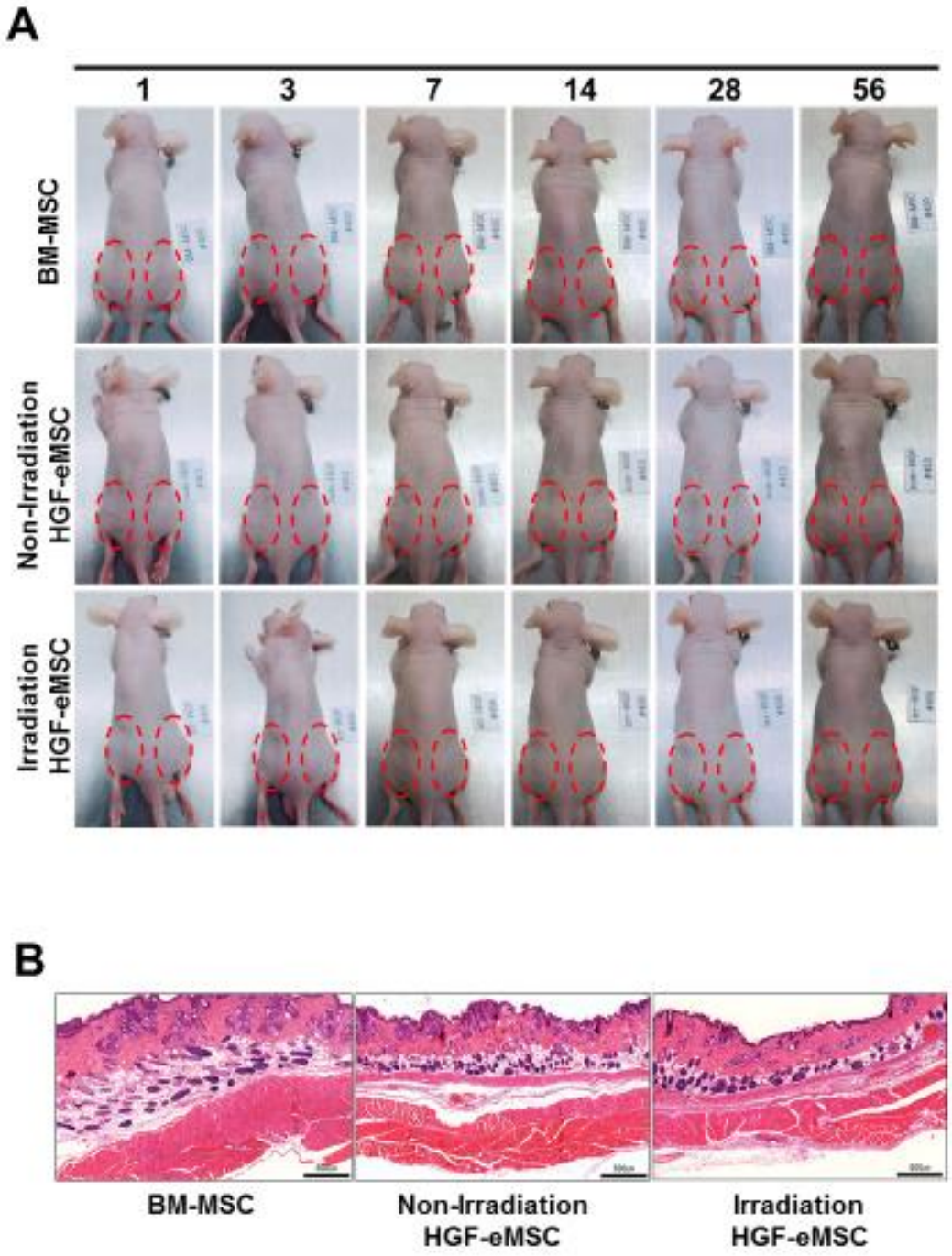


Fig. S5. Examination of the tumorigenicity of HGF-eMSCs. Subcutaneous implantation of different types of MSCs into the nude mice did not form the teratoma up to 56 days from their treatments. (**A**) Representative images of nude mice receiving different types MSCs ($1*10^6$ cells/100ul per red circle). (**B**) Representative images of Hematoxylin-and-eosin staining. Scale bars: 50μm.

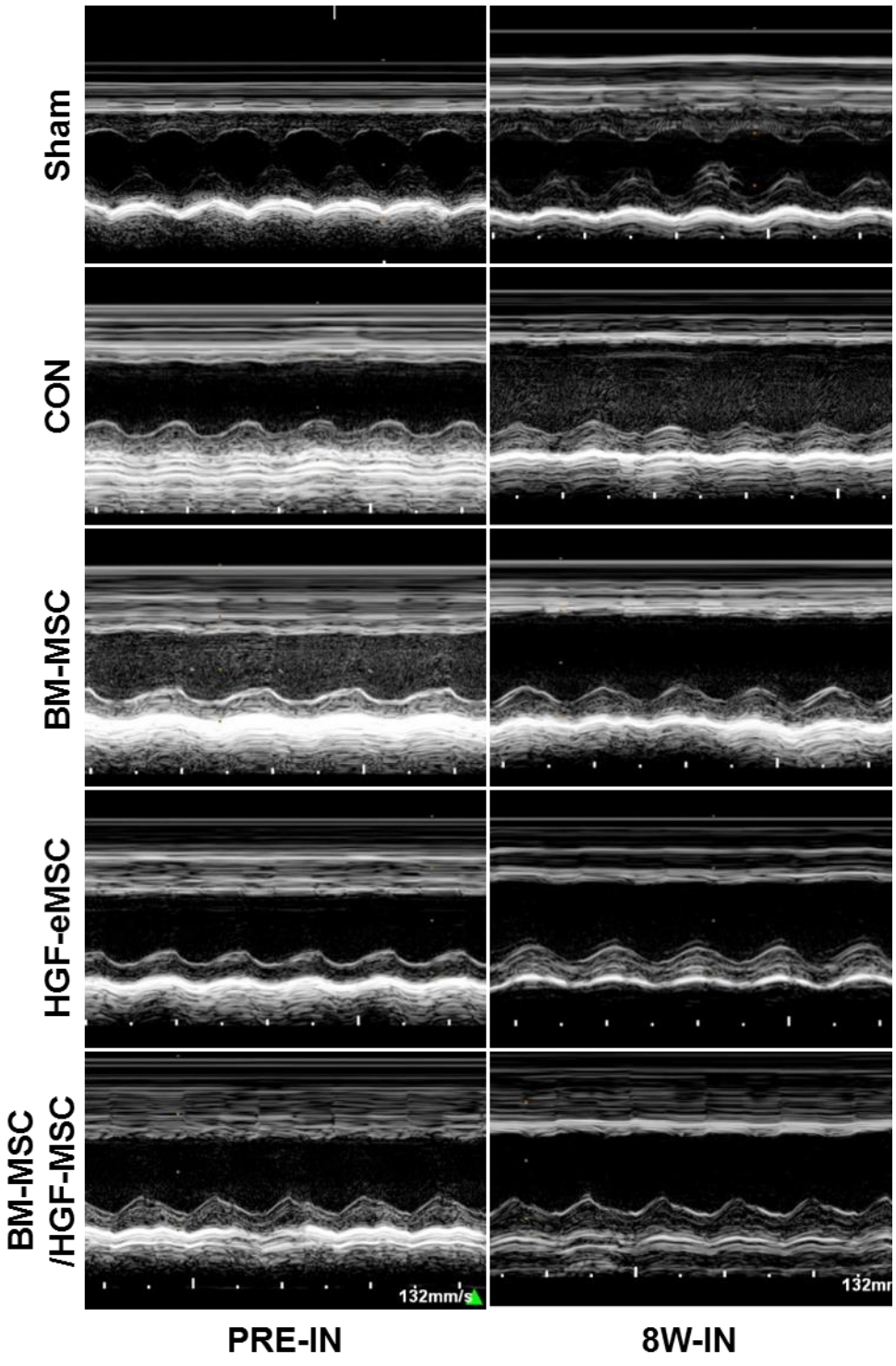


Fig. S6. Representative echo images of all experimental groups at 8 weeks after interventions.

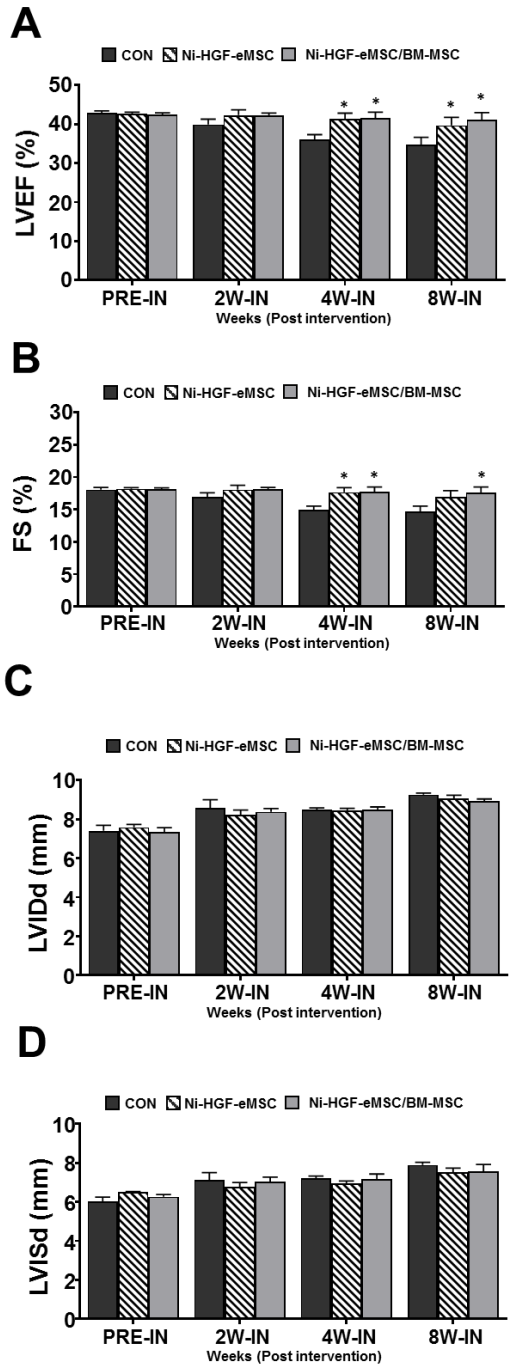
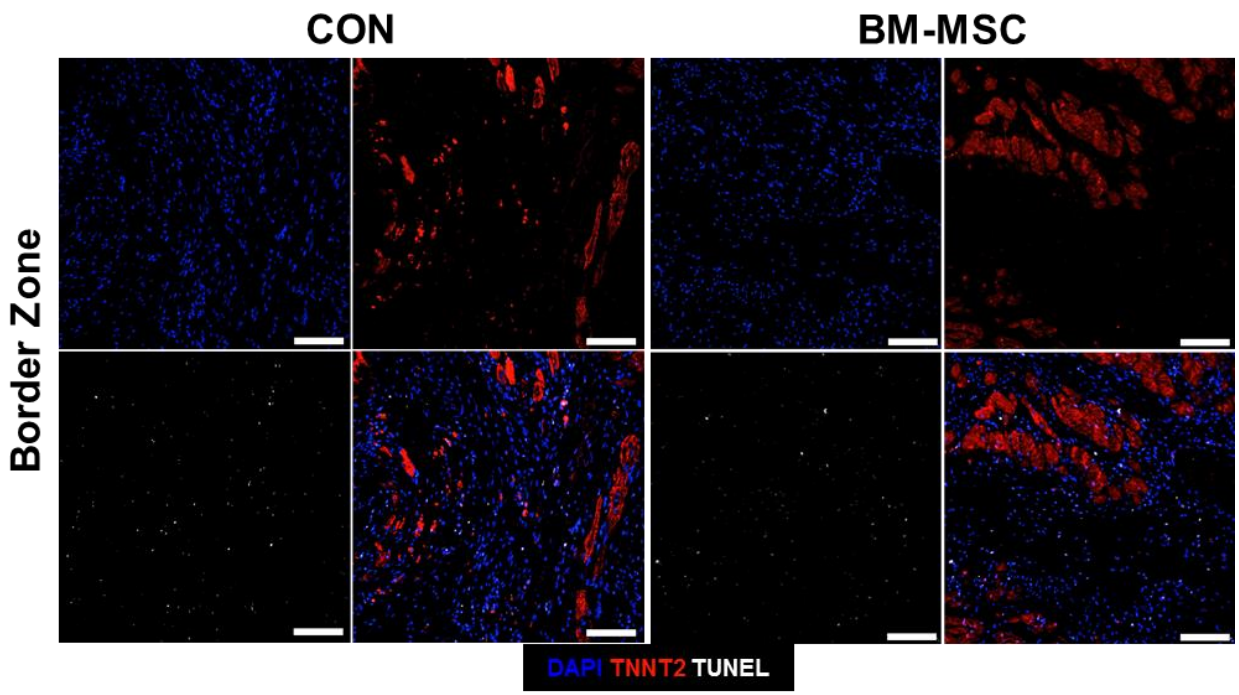
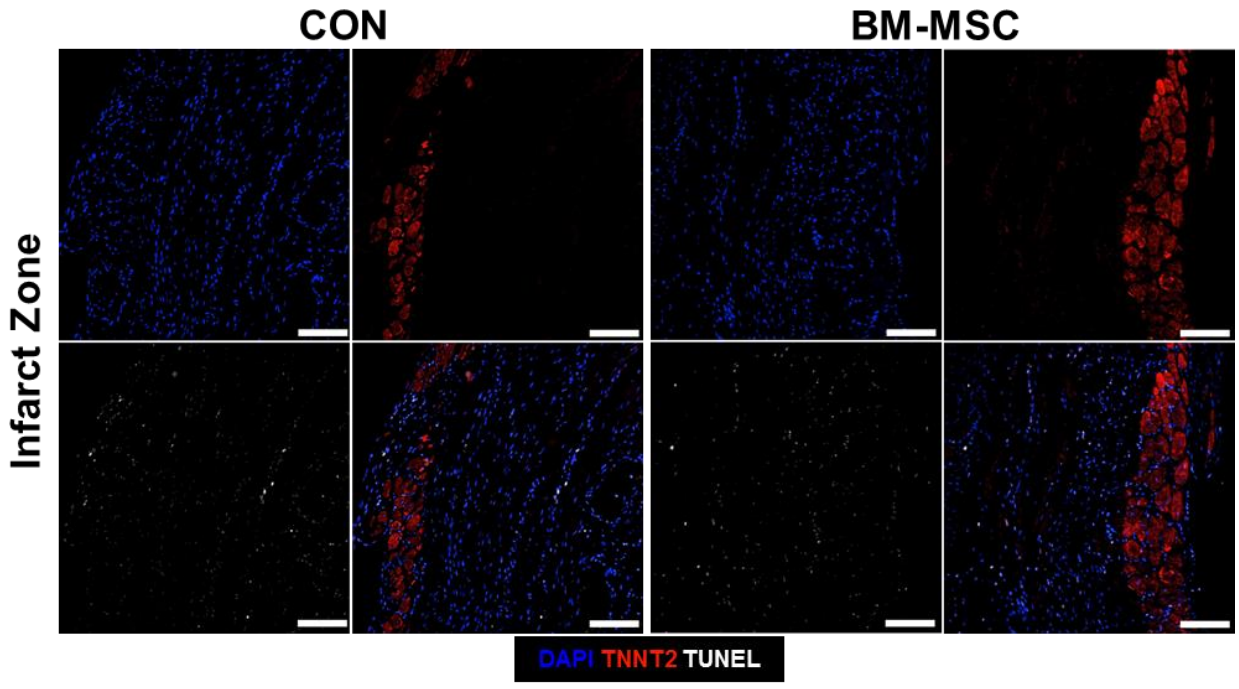


Fig. S7. Effects of nonirradiated HGF-eMSC for priming BM-MSCs and their therapeutic potential for MI. (**A**) Left ventricular ejection fraction (**B**) Fractional shorting (**C**) Left ventricular internal Diastolic dimension and (**D**) Left ventricular internal systolic dimension. **p* <0.05 compared to Control group, n= 5 per each group.


CON
BM-MSC
Border Zone
DAPI TNNT2 TUNEL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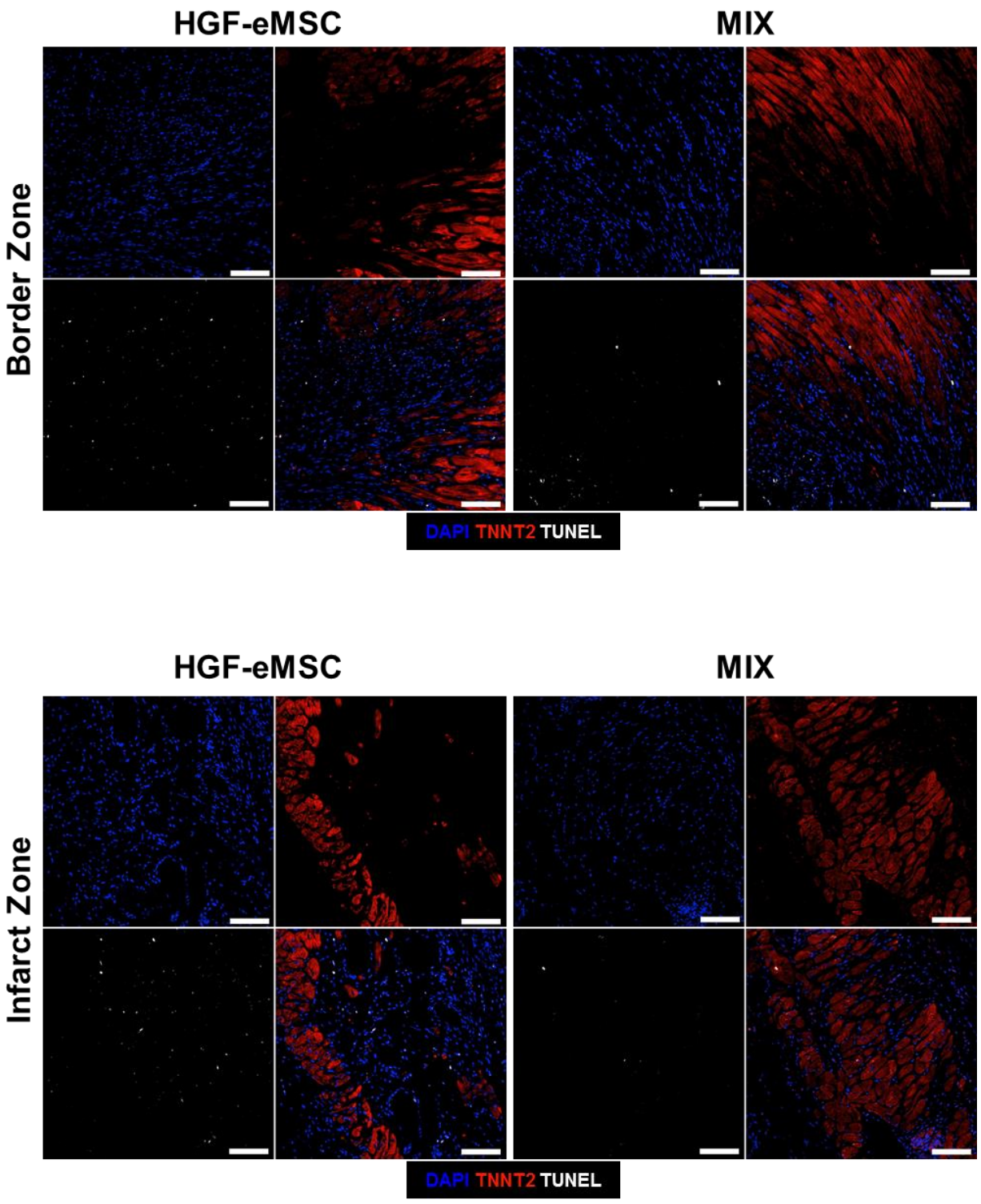

Fig. S8. Representative immunofluorescence staining images with cardiac-specific maker proteins. TNNT2 (red), TUNEL signal (grey), and DAPI for nucleus (blue) on cardiac tissues harvested at 8 weeks after MI. Scale bars: 50μm.

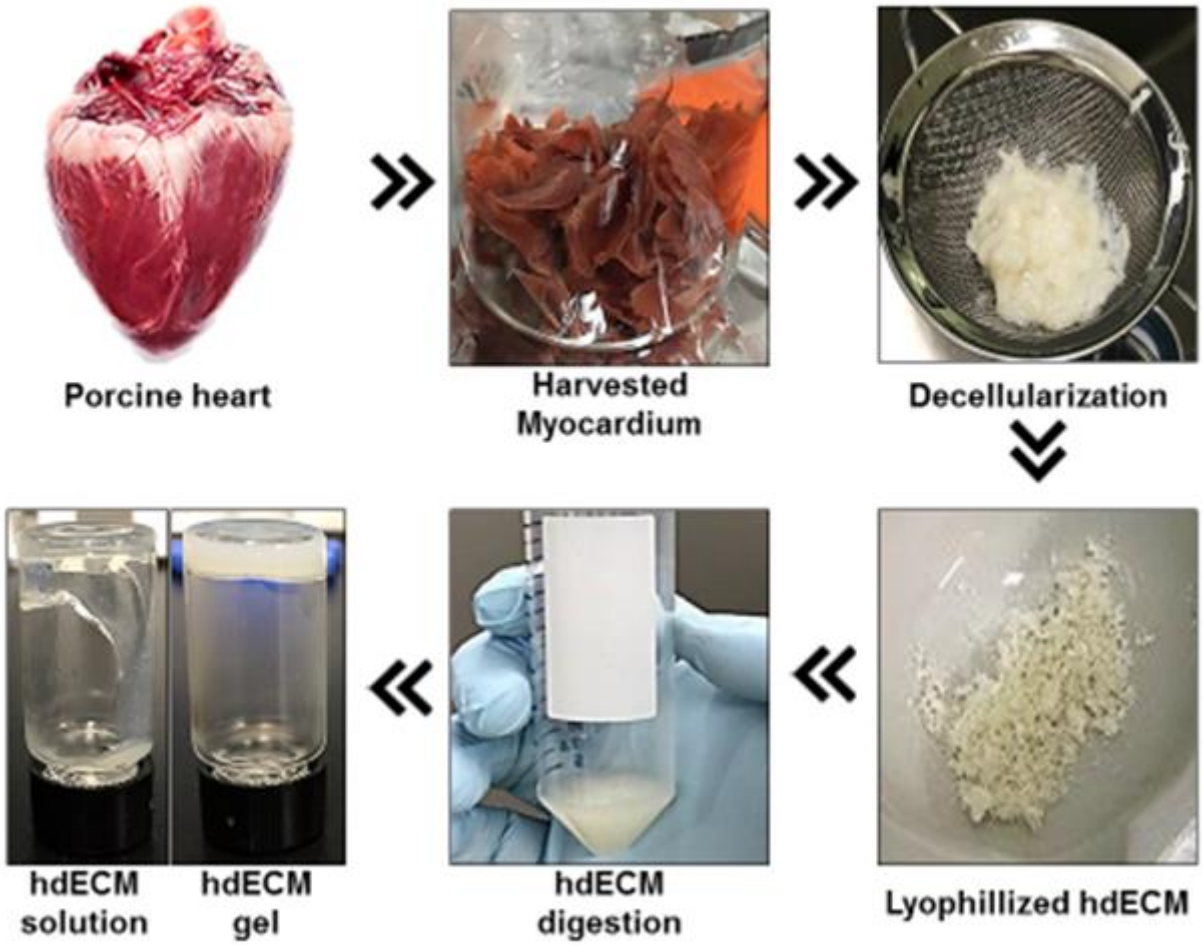

Fig. S9. Schematic illustration demonstrating the procedures for generating decellularized pig hdECM. Photo credit: Sanskrita Das, Pohang University of Science and Technology.

Table S1. Primer sequences used for qRT-PCR analysis.

Gene	Primers	
	Forward	**Reverse**
GAPDH	AGTGCCAGCCTCGTCTCATA	GTAACCAGGCGTCCGATACG
VEGF	ACGAAAGCGCAAGAAATCCC	CTCCAGGGCATTAGACAGCA
HGF	CGACAGTGTTTCCCTTCTCG	ATTGAGAACCTGTTTGCGTTTCT
FGF	AGAAGAGCGACCCTCACATCA	CGGTTAGCACACACTCCTTTG
IGF	AGTTGGTGGATGCTCTCCAGT	CACTCATCCACGATTCCTGTC
Angiopoietin-1	CTCGCTGCCATTCTGACTCAC	GACAGTTGCCATCGTGTTCTG