Supporting Information

Multi-modal hydrogel-based platform to deliver and monitor cardiac progenitor/stem cell engraftment

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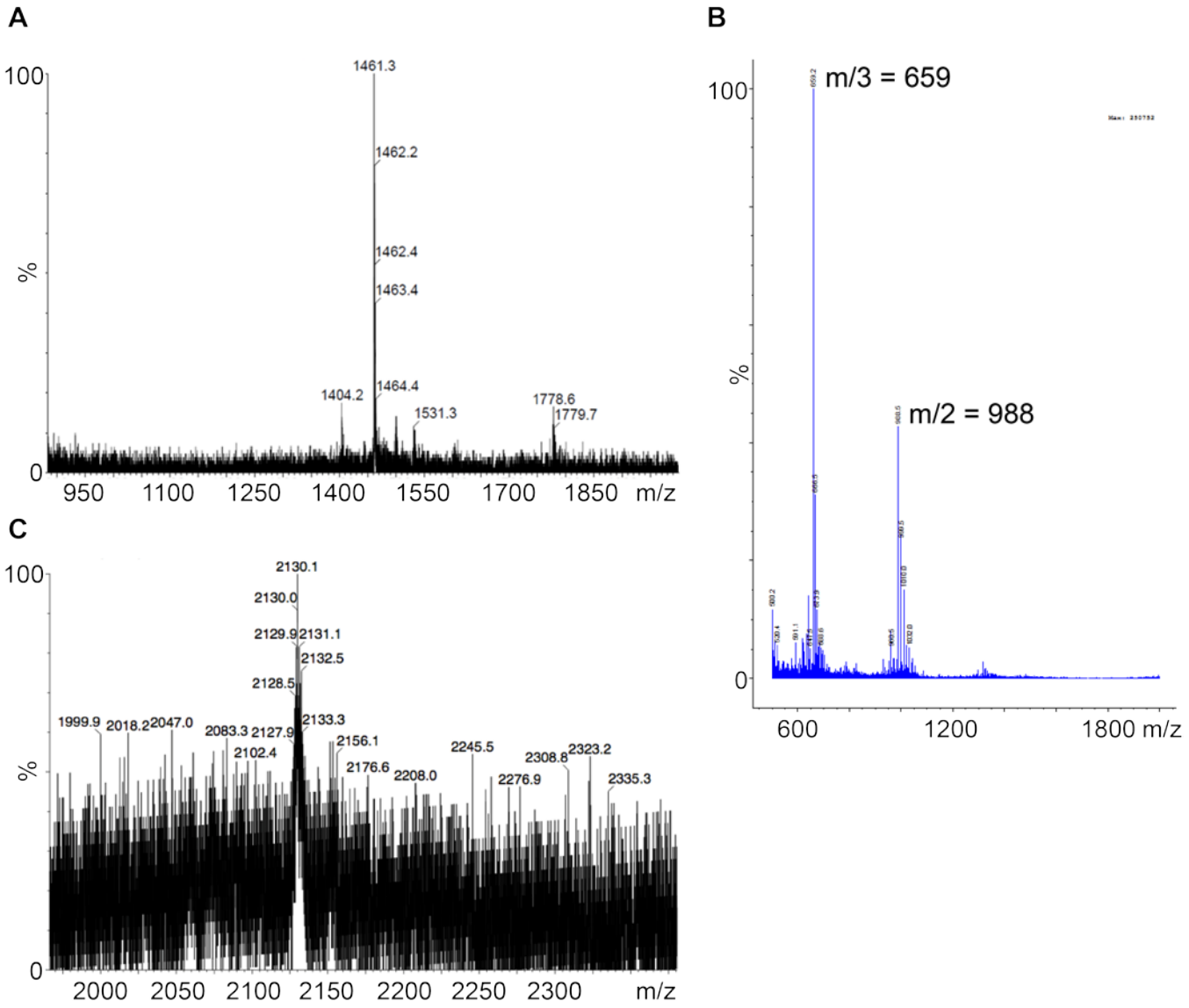
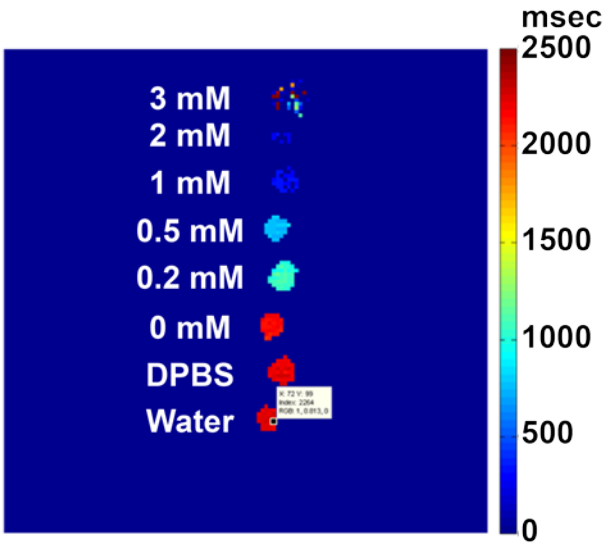
Supplementary Information



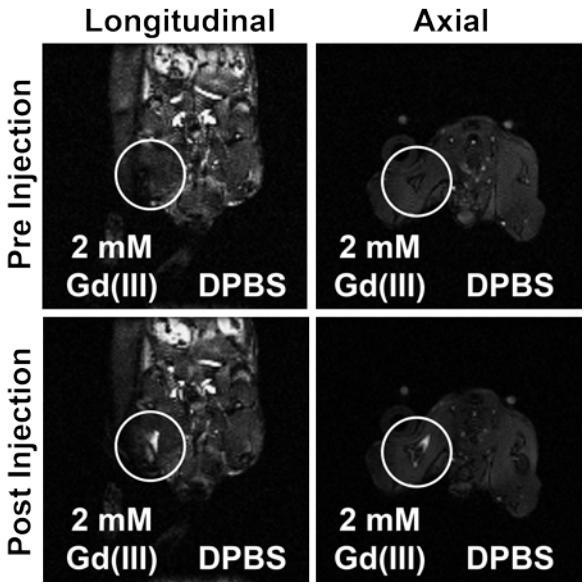
Supplementary Figure 1. Mass Spectra of HPLC-purified synthesized peptides. (A) HBP, CGGGLRKKLGKAGGGC peptide expected mass is 1461. (B) CK(DOTA)GGGLRKKLGKAGGGC peptide total expected mass is 1976.35. The primary peak corresponds to expected m/3 = 659 and the secondary peak corresponds to expected m/2 = 988. (C) CK(DOTA-Tris)GGGLRKKLGKAGGGC peptide with Gd(III) expected mass is 2132.28, the sum of CK(DOTA-Tris)GGGLRKKLGKAGGGC peptide (MW 1975.05) and Gadolinium (MW 157.23).

Supplementary Table 1. pH of hydrogel solutions.

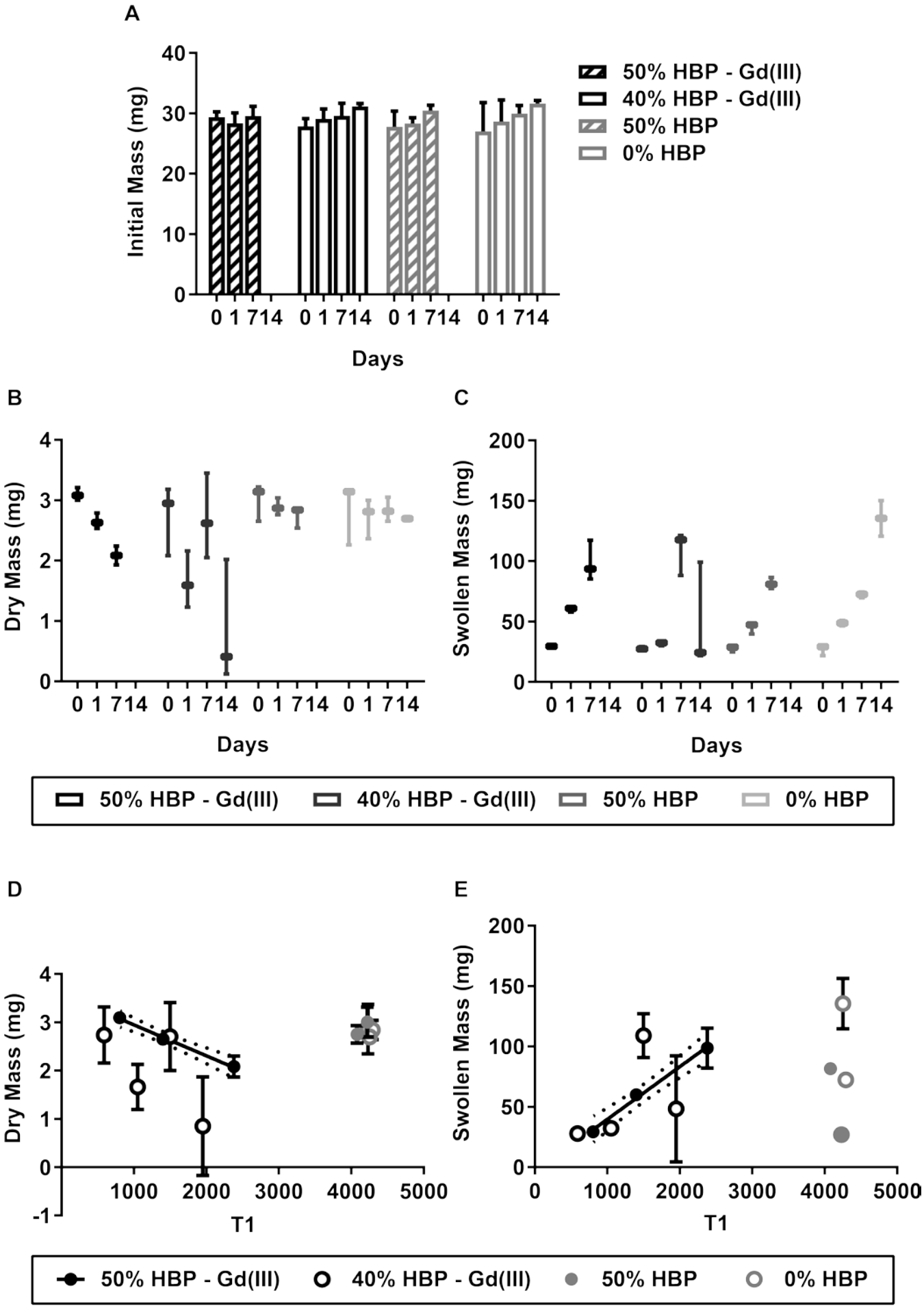
	Alone in DPBS solution	5% (w/v) crosslinked with	
		PEG dithiol	HBP
4 arm PEG-Acrylate	~6-7	~7	~8



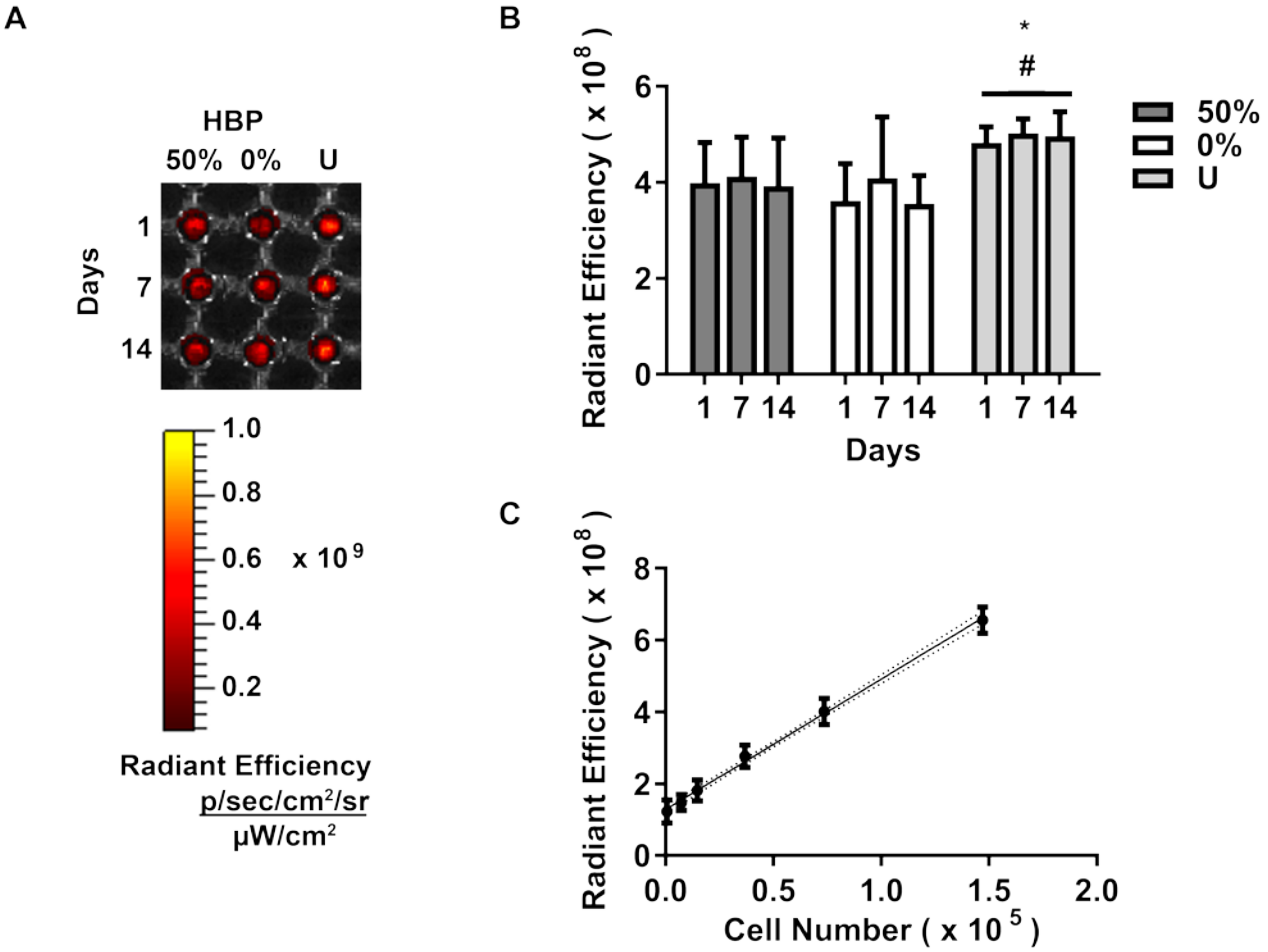
Supplementary Figure 2. Gd(III) concentration calibration with T1 values (msec).
T1 values of 50 µL10% (w/v) PEG gels with 3, 2, 1, 0.5 and 0.2 mM Gd(III)-HBP as well as DPBS and DI water controls.



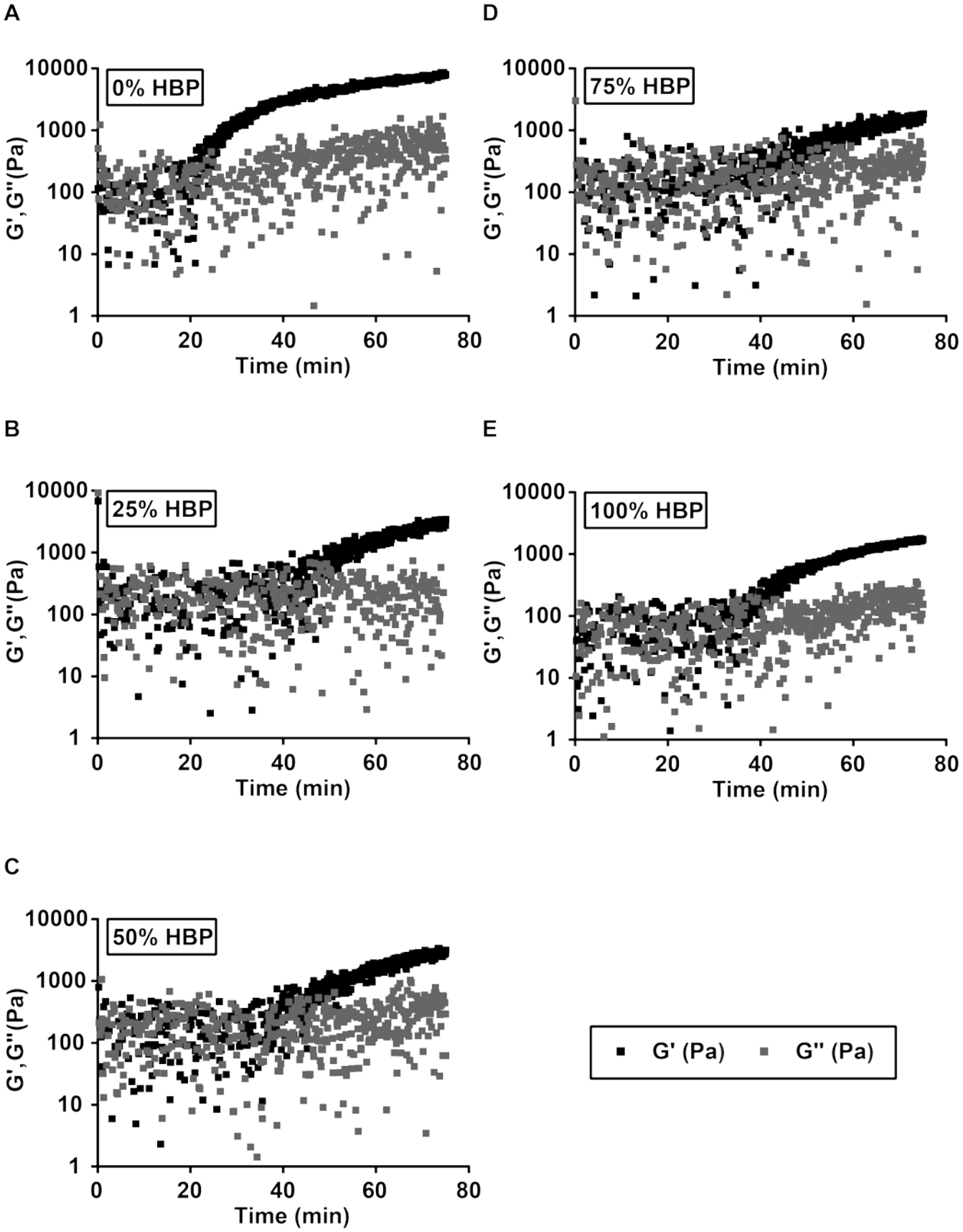
Supplementary Figure 3. T1 weighted images of 10% (w/v) PEG hydrogel crosslinked with 2 mM Gd(III)-HBP hydrogels in mouse hind limbs. Images before (top row) and after (bottom row) injection. 50 µL of 10% (w/v) PEG with 2 mM Gd(III)-HBP (right leg) and DPBS (left leg) were injected into mouse hind limbs. Longitudinal images are shown in the left two columns and axial images are shown in the right two columns.


A
Initial Mass (mg)
40
30
20
10
0
0 1 7 14 0 1 7 14 0 1 7 14 0 1 7 14
Days
50% HBP - Gd(III)
40% HBP - Gd(III)
50% HBP
0% HBP
B
Dry Mass (mg)
4
3
2
1
0
0 1 7 14 0 1 7 14 0 1 7 14 0 1 7 14
Days
C
Swollen Mass (mg)
200
150
100
50
0
0 1 7 14 0 1 7 14 0 1 7 14 0 1 7 14
Days
50% HBP - Gd(III)
40% HBP - Gd(III)
50% HBP
0% HBP
D
Dry Mass (mg)
4
3
2
1
0
-1
1000 2000 3000 4000 5000
T1
E
Swollen Mass (mg)
200
150
100
50
0
0 1000 2000 3000 4000 5000
T1
50% HBP - Gd(III)
40% HBP - Gd(III)
50% HBP
0% HBP

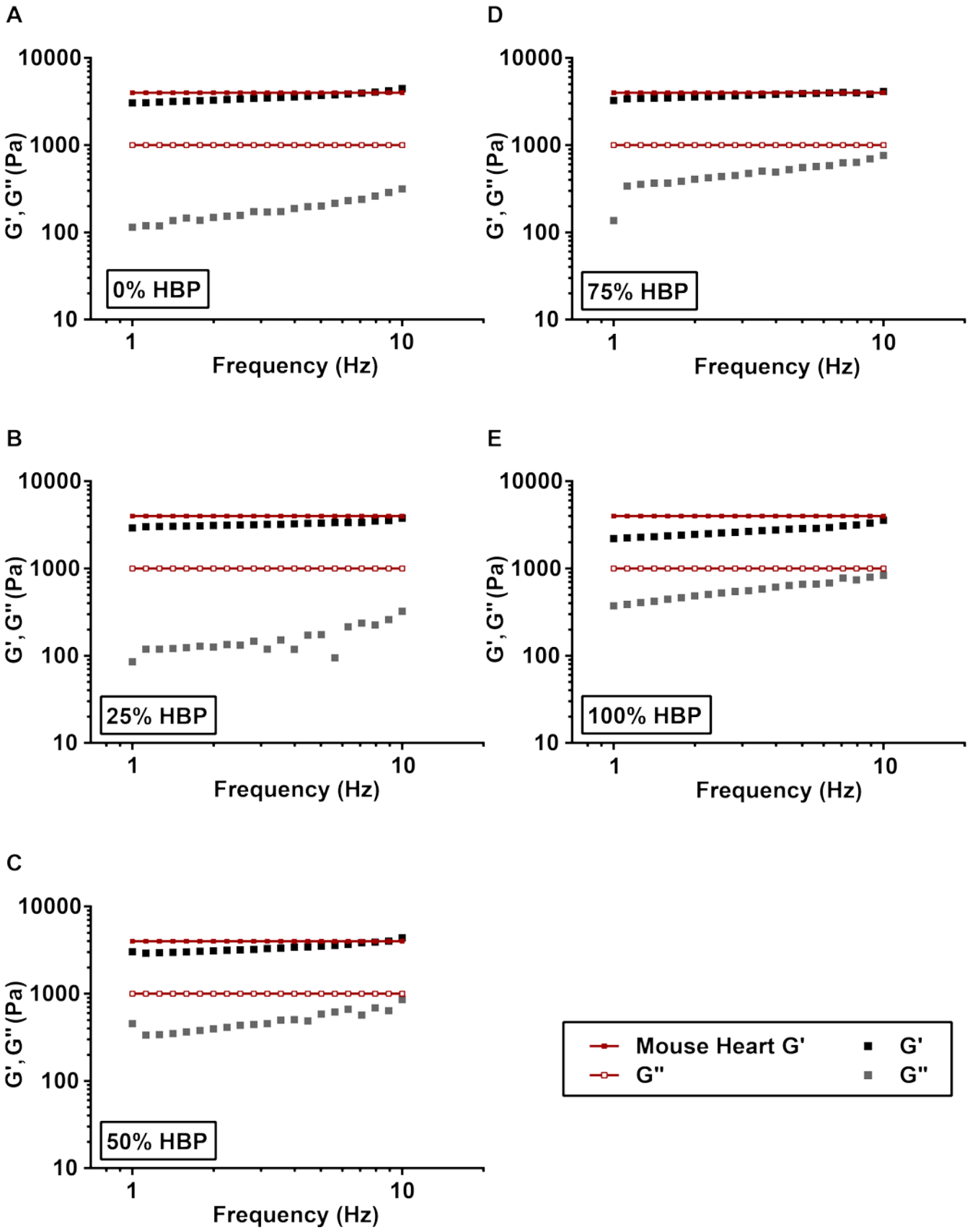
Supplementary Figure 4. Correlation of Gd(III)-containing hydrogel mass change and T1 *in vitro*. (A) Average initial hydrogel wet masses for the various time points and hydrogel formulations. No statistical difference between samples, verified by one-way ANOVA. (B) Dry mass and (C) Swollen (wet) mass distributions of samples over time. Bars designate mass ranges. (D) Representative dry mass correlation with T1 values (50% HBP – Gd(III) $r^2 = 0.9231$). (E) Representative swollen (wet) hydrogel mass correlation with T1 values (50% HBP – Gd(III) $r^2 = 0.9158$). T1 units are all in milliseconds. Error bars in (A), (D), and (E) denote one standard deviation. 95% CI are designated by black dotted lines and were included for experimental samples where r^2 of correlation was above 0.90.



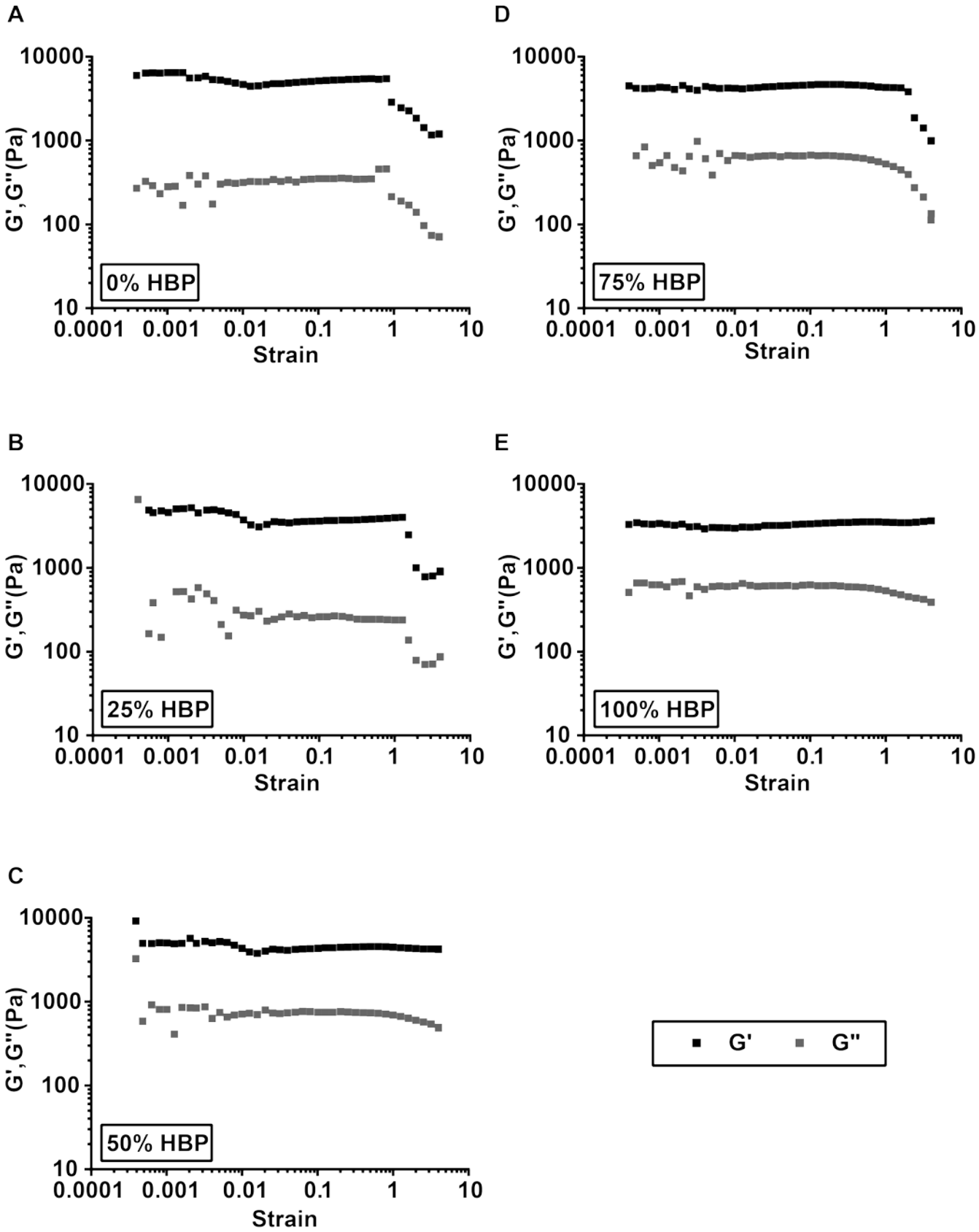
Supplementary Figure 5. Relative quantification by DiR membrane staining of initially seeded CSC-Luc2 for metabolic activity assay in 0% and 50% HBP hydrogels. (A) Image of radiant efficiency of representative initial seeded DiR labelled CSC-Luc2 in each hydrogel formulation and for each time point. (B) Mean radiant efficiency of all samples as relative quantification of initial cell density. Samples compared by two-way ANOVA, Tukey multiple comparison post hoc test *$p < 0.05$ significant difference of untransduced control samples with 50%, #$p < 0.01$ significant difference of untransduced control samples with 0%. (C) Linear correlation ($r^2 = 0.9716$) and 95% confidence intervals of DiR (radiant efficiency) signal (dotted lines) with cell number. Error bars in all panels represent one standard deviation.



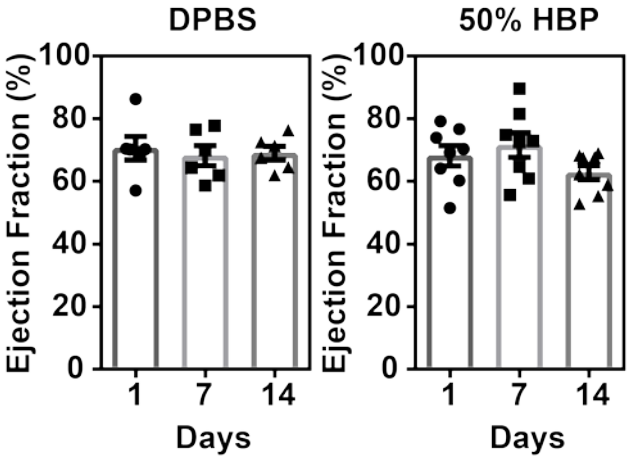
Supplementary Figure 6. Representative rheological time sweeps. The storage moduli (G') and loss moduli (G") of (A) 0% (B) 25%, (C) 50%, (D) 75%, and (E) 100% HBP hydrogels were measured at 1 Hz and 0.0001 (0.01%) strain at 37 °C. Black squares denote storage moduli (G') and gray squares denote loss moduli (G"). The point of gelation was determined as the point at which the storage modulus exceeds the loss modulus. Y-axis units are Pa and the x-axis units are minutes.



Supplementary Figure 7. Representative rheological frequency sweeps. G' and G" of (A) 0%, (B) 25%, (C) 50%, (D) 75% and (E) 100% HBP hydrogels compared with literature values of healthy murine heart tissue (G': 4000 Pa, G": 1000 Pa)[40] for 1-10 Hz (ω = 6.3–62.83 rad/s) at a fixed strain amplitude of 0.01 (1%) and 37 °C. G', storage moduli (black squares) and G", loss moduli (gray or open squares) for hydrogels (black, gray) and mouse heart tissue (red). Y-axis units are Pa and the x-axis units are Hz.



Supplementary Figure 8. Representative rheological strain sweeps. G' and G" of (A) 0%, (B) 25%, (C) 50%, (D) 75%, and (E) 100% HBP hydrogels from 0.004-4.00 (0.4-400%) strain were performed at an angular frequency of 8 Hz (52.3 rad/s) and 37 °C ensuring the inclusion of the physiologically relevant range of strains (Healthy: 0.15-0.4 (15 – 40%), Infarcted: 0.03-0.25 (3 – 25%))[41–43]. G', storage moduli (black squares) and G", loss moduli (gray squares). Y-axis units are Pa and the x-axis units are strain.



Supplementary Figure 9. Cardiac Magnetic Resonance (CMR) assessment of heart function. The left ventricle ejection fraction (LVEF) in mice 1, 7, and 14 days after intramyocardial injections of DPBS (left) or 50% HBP hydrogel (right) was measured. LVEF was not significantly different over 14 days after intramyocardial injections between the DPBS and 50% HBP hydrogel injected animals.