

Supplementary Information for

Grafting of Cationic Molecules to Hyaluronic Acid Improves Adsorption and Cartilage Lubrication

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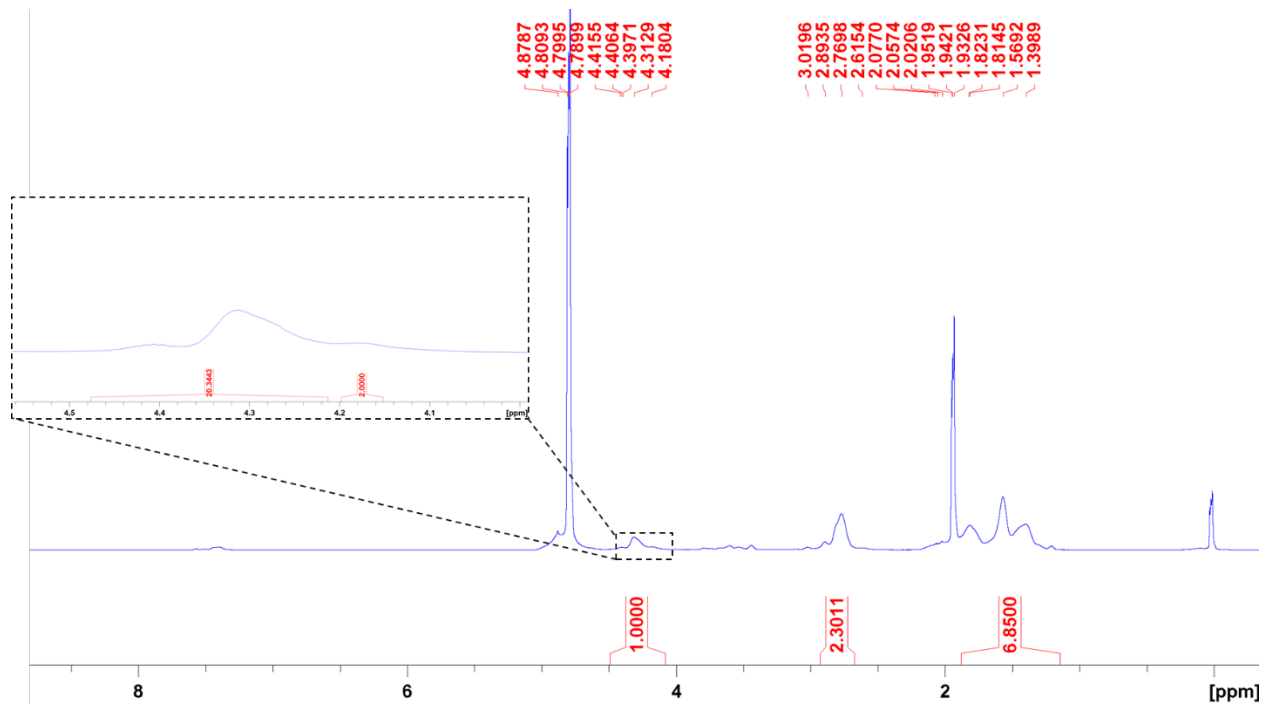


Figure S1. ¹H NMR Spectrum of fully deprotected core peptide recorded in D₂O at 25 °C.

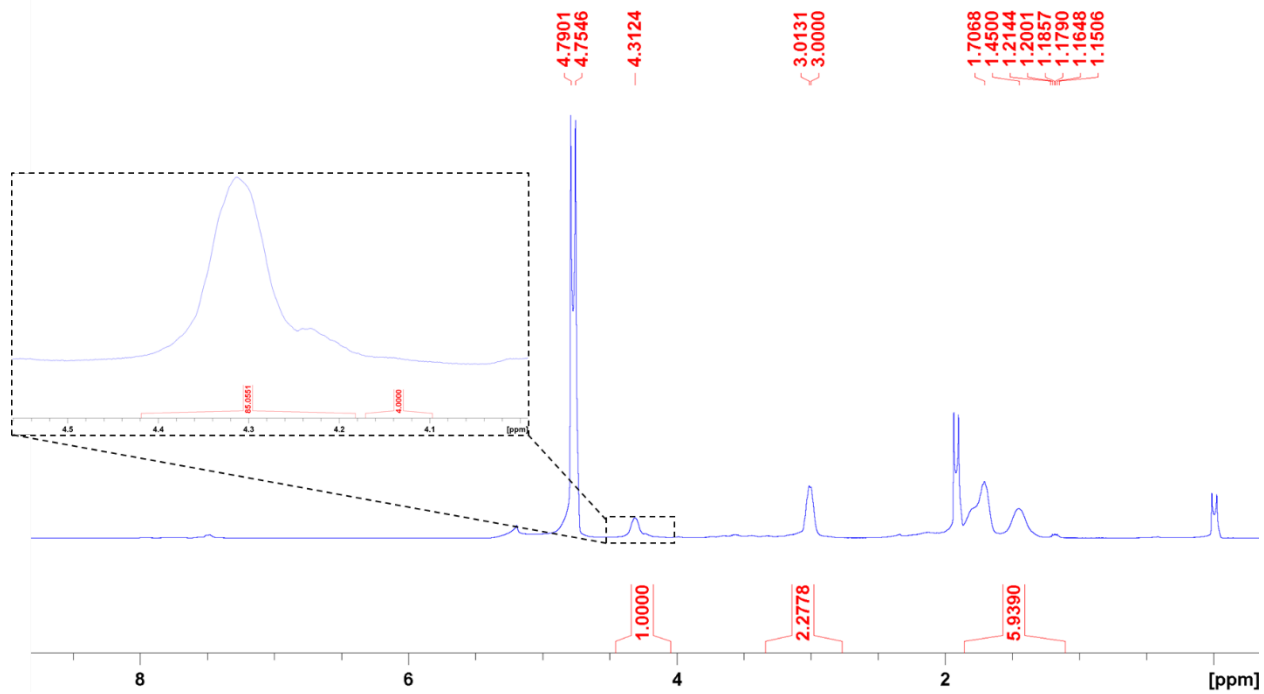


Figure S2. ¹H NMR Spectrum of fully deprotected G0 peptide recorded in D₂O at 25 °C.

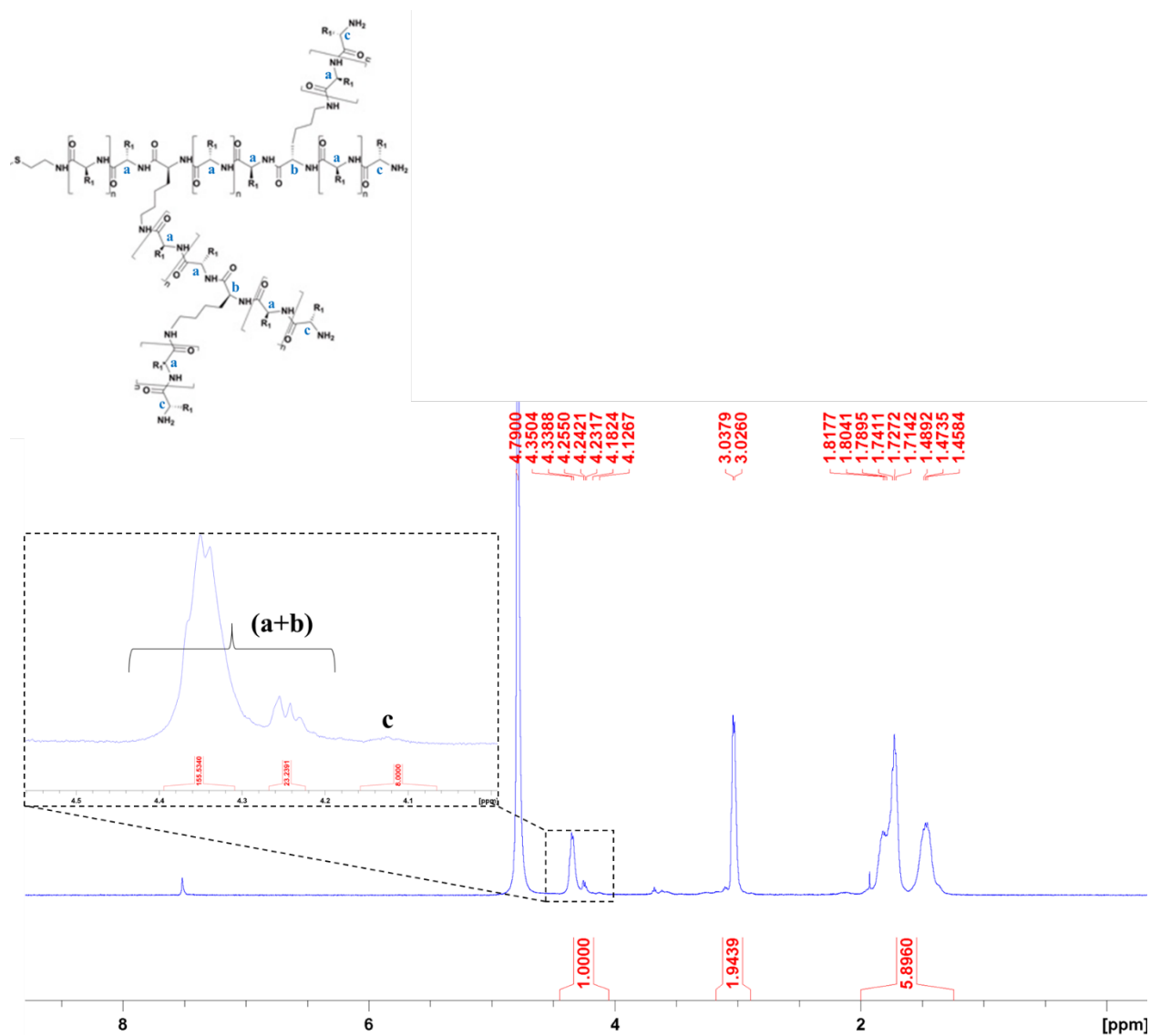


Figure S3. ¹H NMR Spectrum of fully deprotected G1 peptide recorded in D₂O at 25 °C.

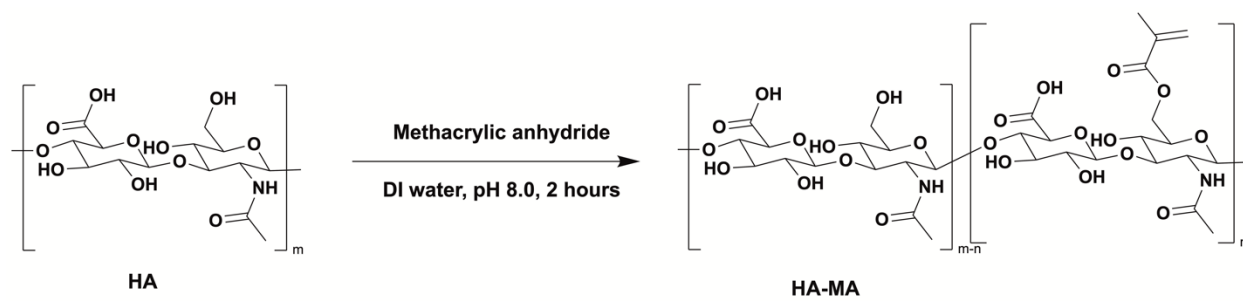


Figure S4. Synthesis of HA-MA from HA. Reaction scheme showing the incorporation of methacrylate groups in the HA by reacting HA with methacrylic anhydride.

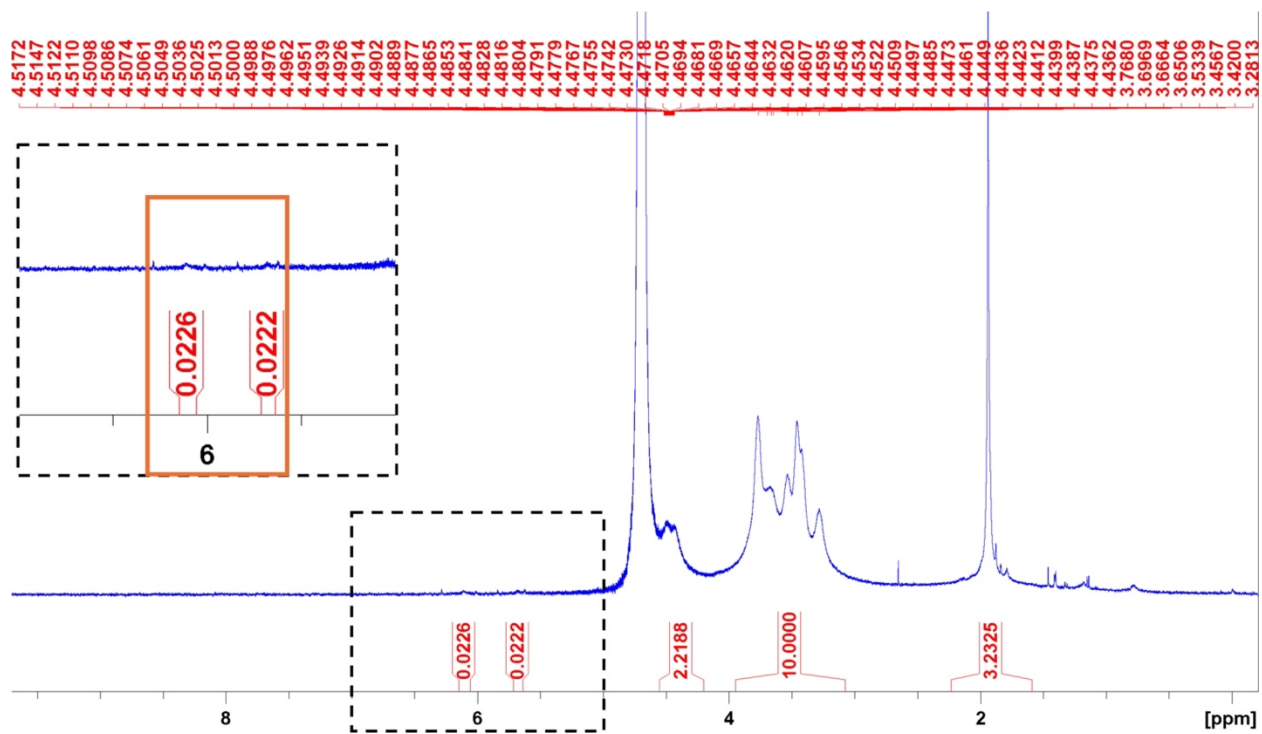


Figure S5. ¹H NMR Spectrum of HA-MA recorded in D₂O at 25 °C. The box shows an expansion of the region from 4.25-6.5 ppm with the methacrylate protons of the HA-MA marked by the orange box.

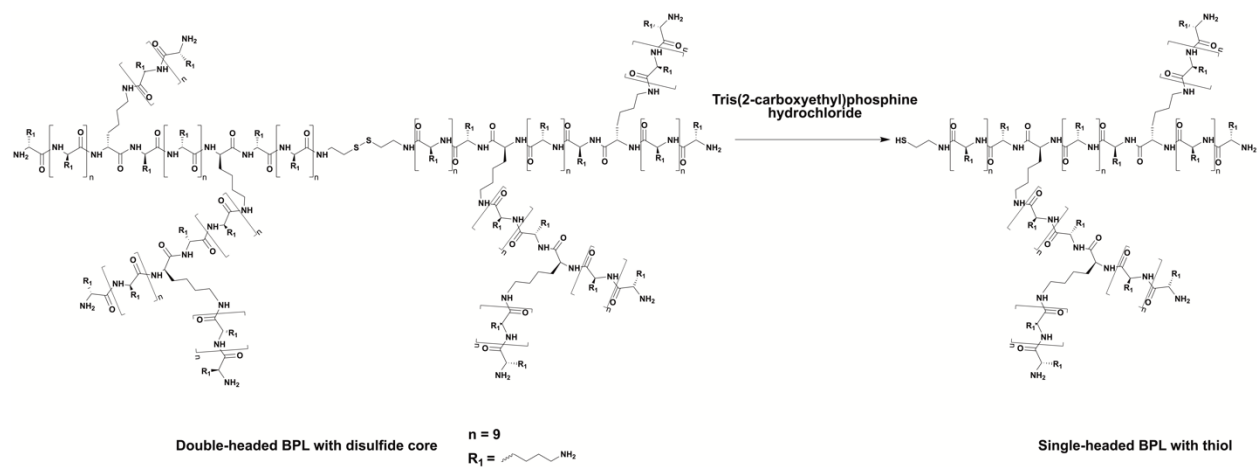


Figure S6. Reduction of disulfide bond in the double-headed BPL molecule.

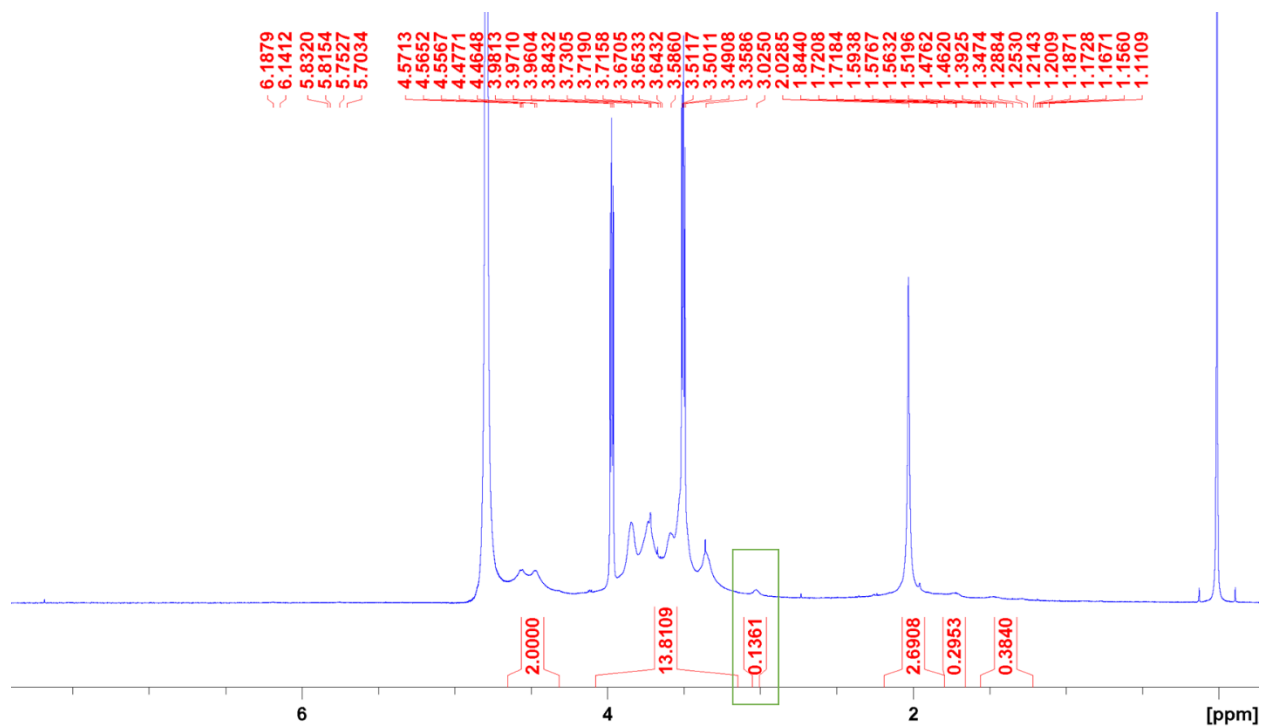


Figure S7. ^1H NMR Spectrum of HA-2BPL recorded in D_2O at $25\text{ }^\circ\text{C}$. Peak at 3.025 ppm (marked with a green colored box) indicates the appearance of methylene protons adjacent to NH_2 group ($-\text{CH}_2\text{CH}_2\text{CH}_2\text{CH}_2\text{NH}_2$) of lysine unit from fully deprotected BPL.

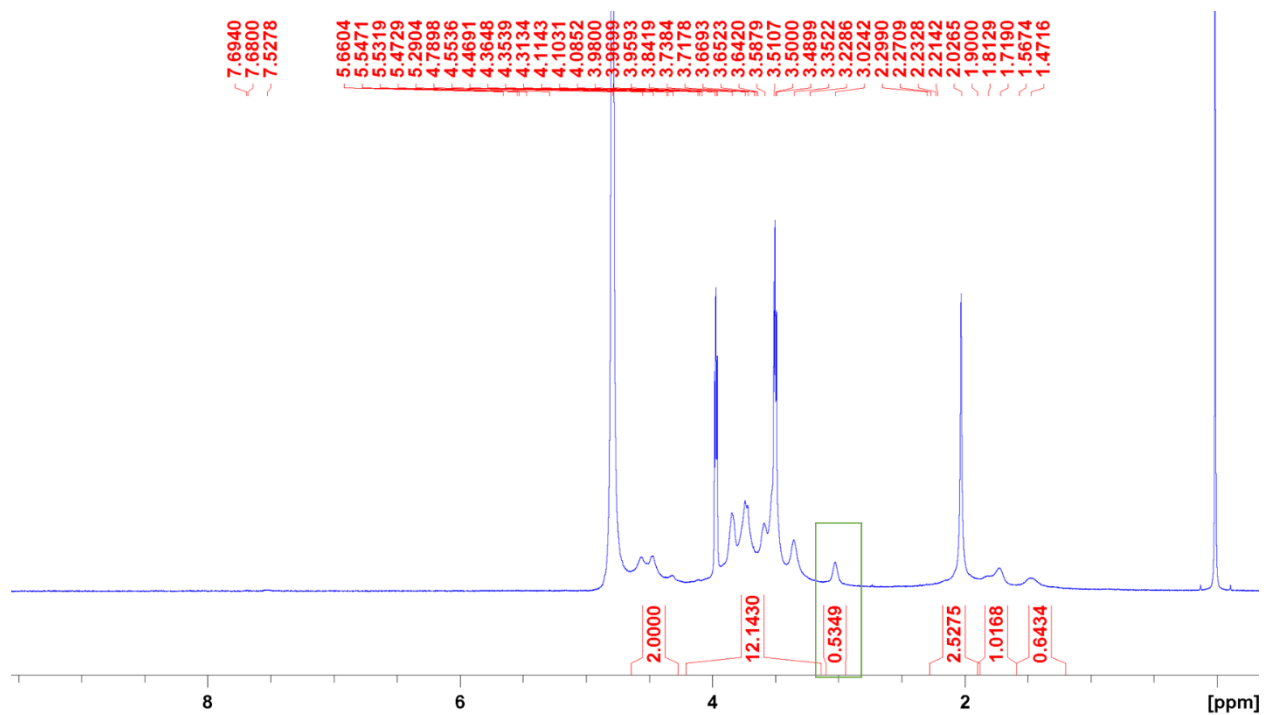


Figure S8. ^1H NMR Spectrum of HA-8BPL recorded in D_2O at 25 °C. Peak at 3.024 ppm (marked with a green colored box) indicates the appearance of methylene protons adjacent to NH_2 group ($-\text{CH}_2\text{CH}_2\text{CH}_2\text{CH}_2\text{NH}_2$) of lysine unit from fully deprotected BPL.

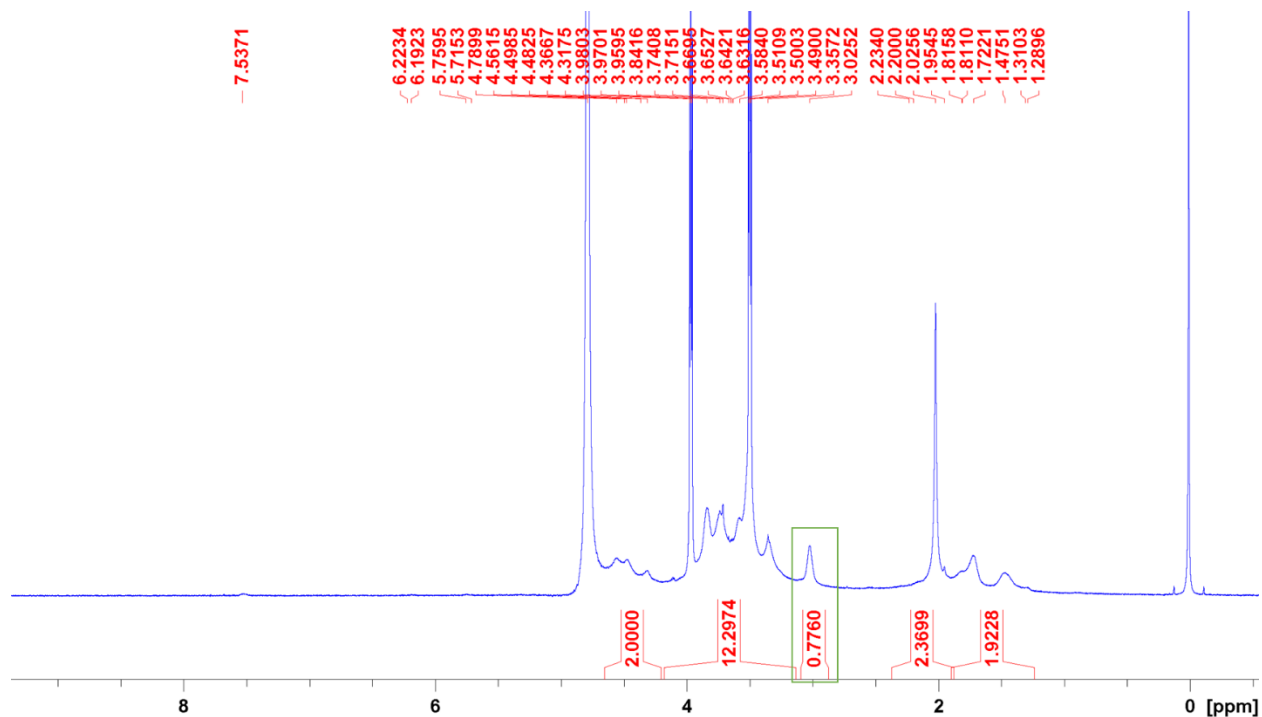


Figure S9. ^1H NMR Spectrum of HA-12BPL recorded in D_2O at 25 °C. Peak at 3.025 ppm (marked with a green colored box) indicates the appearance of methylene protons adjacent to NH_2 group ($-\text{CH}_2\text{CH}_2\text{CH}_2\text{CH}_2\text{NH}_2$) of lysine unit from fully deprotected BPL.

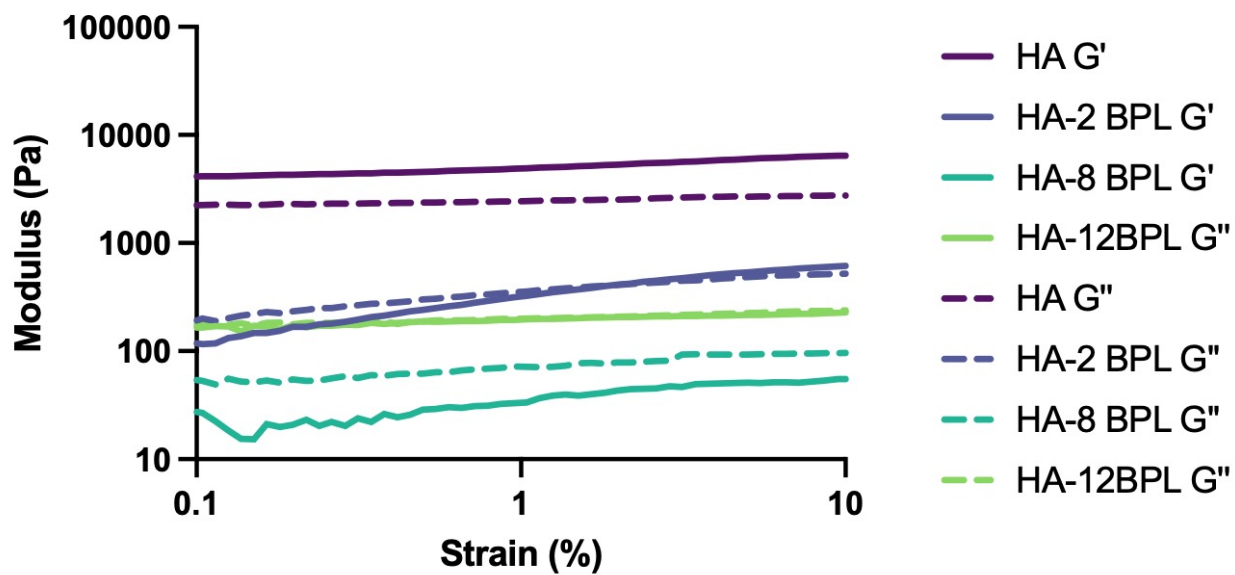


Figure S10. Strain sweep of HA and HA-BPL molecules.

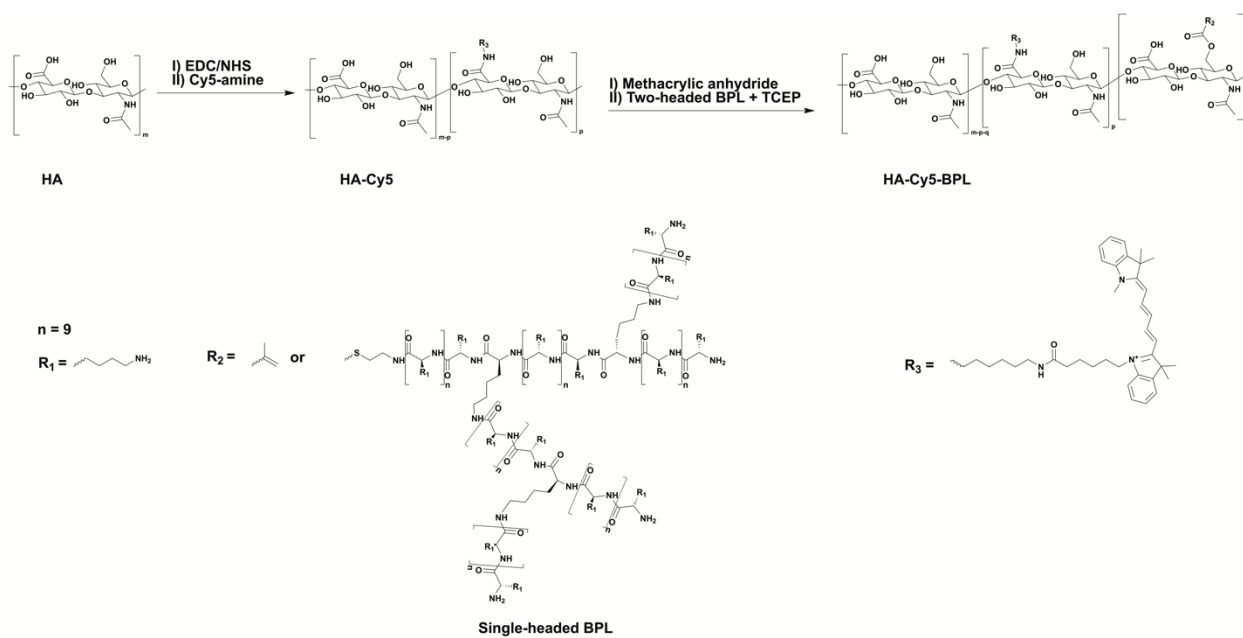


Figure S11. Synthesis of Cy5 containing HA-BPL molecules.

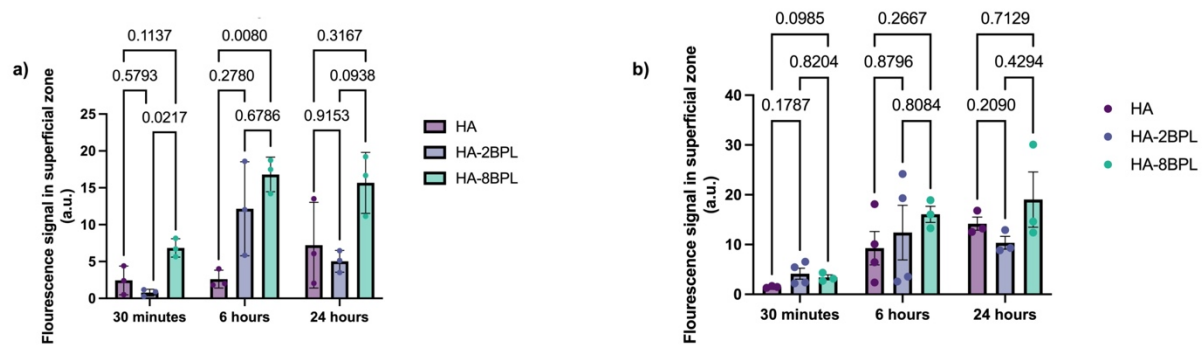


Figure S12. Adsorption of HA-BPL molecules to cartilage as a function of time. a) Penetration of HA BPL molecules in healthy cartilage explants. b) Penetration of HA BPL molecules in OA-mimetic cartilage explants.

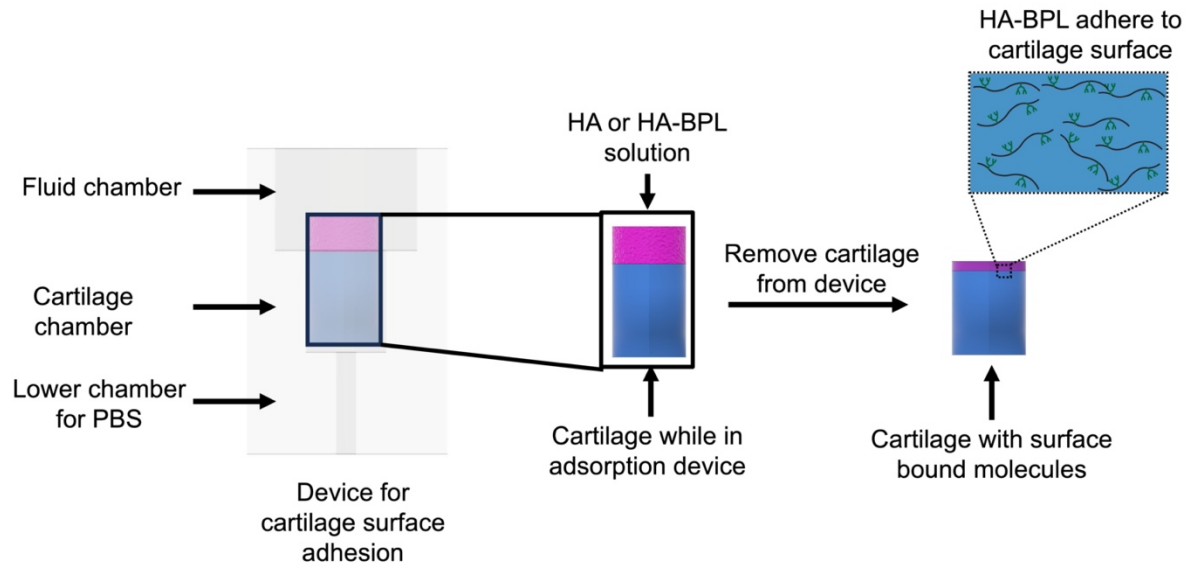


Figure S13. Schematic of device used to assess adsorption of HA and HA-BPL molecules to cartilage.

Table S1. Molecular characteristics of different generations

	Number of terminal lysine residues	Total number of branches/arms	^a DP _{branch}	^b DP _{total}
Core	2	2	10	20
G0	4	6	10	60
G1	8	14	10	140

^{a,b} Theoretical estimation of average number of lysine repeating units per branch (DP_{branch}) and the total number average degree of polymerization (DP_{total})

^a Yields determined gravimetrically after precipitation, filtration, and freeze-drying of the polypeptides.

Table S2. Results of the ^1H NMR characterization of BPL molecules

^1H NMR Integrals					
	$I(\text{H}_a) + I(\text{H}_b)$	$I(\text{H}_c)$	$[I(\text{H}_a) + I(\text{H}_b) + I(\text{H}_c)]/I(\text{H}_c)$	$\text{DP}_{\text{branch}}$	DP_{total}
Core	20.3	2	11.15	~11.1	22
G0	85.0	4	22.25	~11.1	89
G1	178.76	8	23.34	~11.6	187

$I(\text{H}_a)$ = Integral for methine protons of the terminal lysine units from the core peptide

$I(\text{H}_b)$ = Integral for methine protons of the terminal lysine units from the G0 peptide

$I(\text{H}_c)$ = Integral for methine protons of the terminal lysine units from the G1 peptide