

Supplementary Information

Design, characterization and evaluation of β -hairpin peptide hydrogels as support for osteoblast cell growth and bovine lactoferrin delivery

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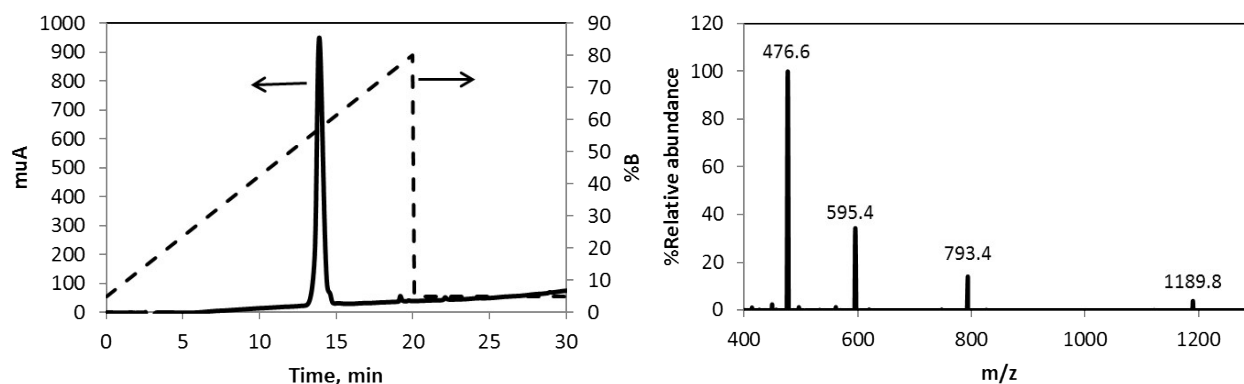


Figure S1. LCMS profile of purified peptide H4LMAX (t_R = 13.9 min, ca 98 % as analysed by peak area of RP-HPLC at 214 nm). ESI-MS spectrum of H4LMAX, mass spectra depict m/z ions found at the signal. Peak assignment: $[M + 2H]^{2+}$ obs. 1189.8 (calc. 1190.0); $[M + 3H]^{3+}$ obs. 793.4 (calc. 793.7); $[M + 4H]^{4+}$ obs. 595.4 (calc. 595.5); $[M + 5H]^{5+}$ obs. 476.6 (calc. 476.6).

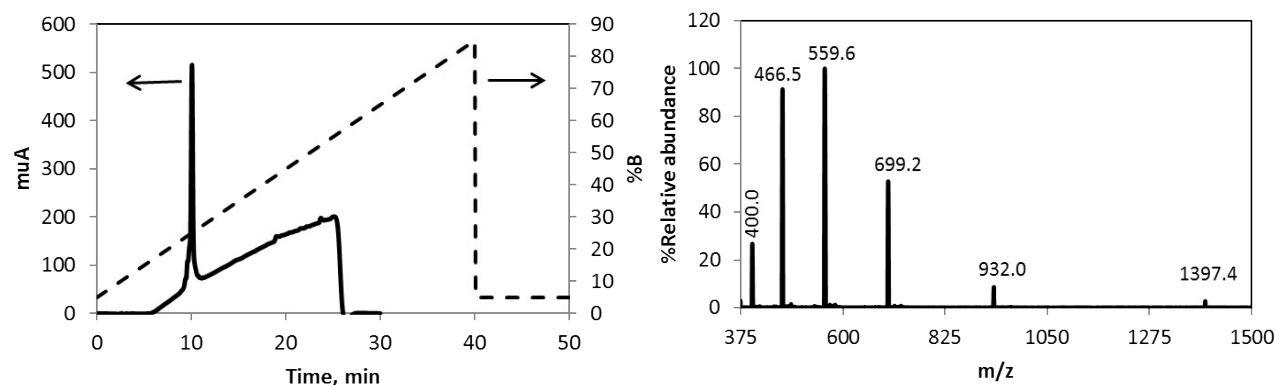


Figure S2. LCMS profile of purified peptide H4LMAX-RGDS (t_R = 10.1 min, ca 99% as analysed by peak area of RP-HPLC at 214 nm). ESI-MS spectrum of H4LMAX-RGDS, mass spectra depict m/z ions found at the signal. Peak assignment: $[M + 2H]^{2+}$ obs. 1397.4 (calc. 1397.7); $[M + 3H]^{3+}$ obs. 932.0 (calc. 932.2); $[M + 4H]^{4+}$ obs. 699.2 (calc. 699.4); $[M + 5H]^{5+}$ obs. 559.6 (calc. 559.7); $[M + 6H]^{6+}$ obs. 466.5 (calc. 466.6); $[M + 7H]^{7+}$ obs. 400.0 (calc. 400.1).

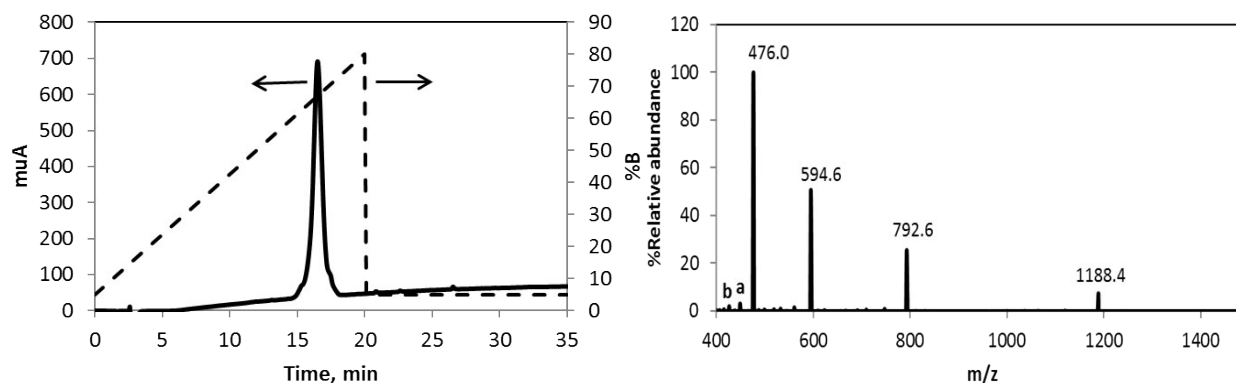


Figure S3. LCMS profile of purified peptide H2LRDMAX ($t_R = 16.5$ min, ca 94 % as analysed by peak area of RP-HPLC at 214 nm). ESI-MS spectrum of H2LRDMAX, mass spectra depict m/z ions found at the signal. Peak assignment: $[M + 2H]^{2+}$ obs. 1188.4 (calc. 1188.5); $[M + 3H]^{3+}$ obs. 792.6 (calc. 792.7); $[M + 4H]^{4+}$ obs. 595.5 (calc. 594.8); $[M + 5H]^{5+}$ obs. 476.0 (calc. 476.0). The signal labeled as (a) corresponds to the His N-terminus truncated peptide $[M + 5H]^{5+}$ obs. 448.5 (calc. 448.6). The signal labeled as (b) corresponds to the His-Leu N-terminally truncated peptide $[M + 5H]^{5+}$ obs. 425.9 (calc. 425.9).

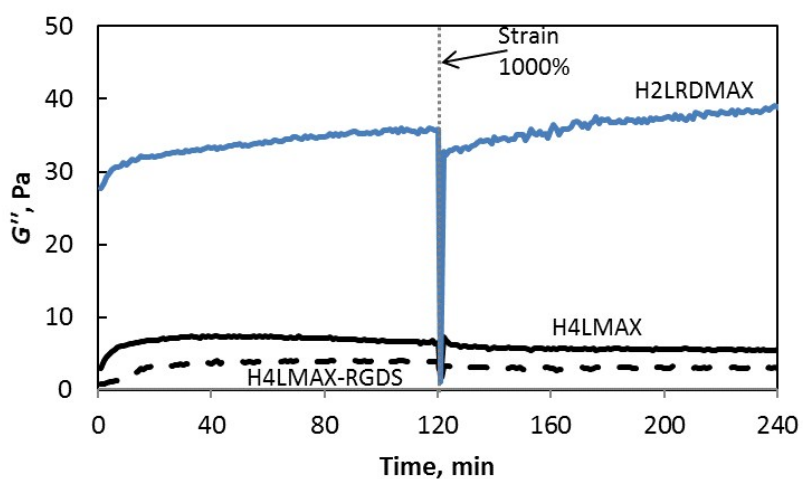


Figure S4. Loss modulus (G'') of 1 wt % peptide hydrogels in 50 mM Tris buffer pH 7.4 with 30 mM NaCl at 37 °C as a function of time (6 rad.s^{-1} , 0.2% strain).

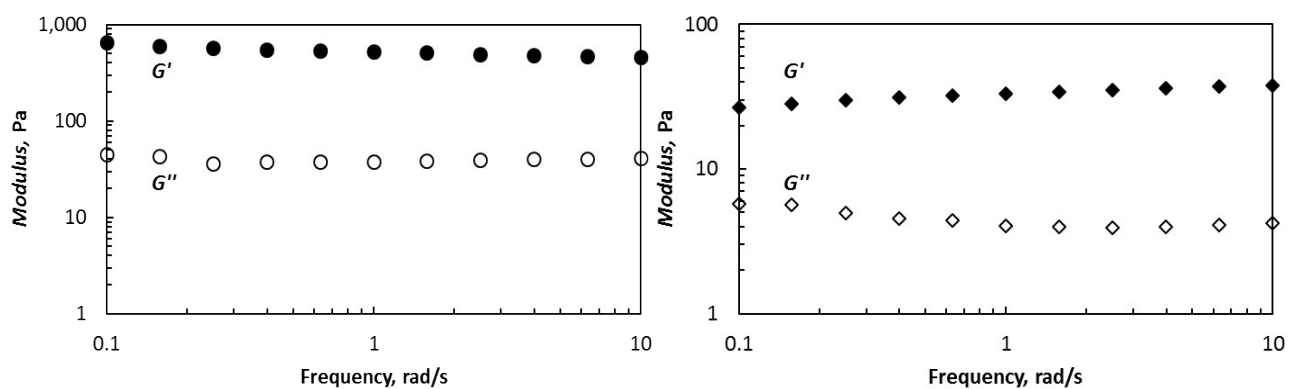


Figure S5. Frequency sweep (0.2 % strain) of 1 wt % hydrogel of H4LMAX-RGDS (left) and H2LRDMAX (right) in 50 mM Tris buffer pH 7.4 with 30 mM NaCl at 37 °C after first dynamic time sweep measurement in Fig S4. G' values are represented by filled symbols and G'' by open symbols.

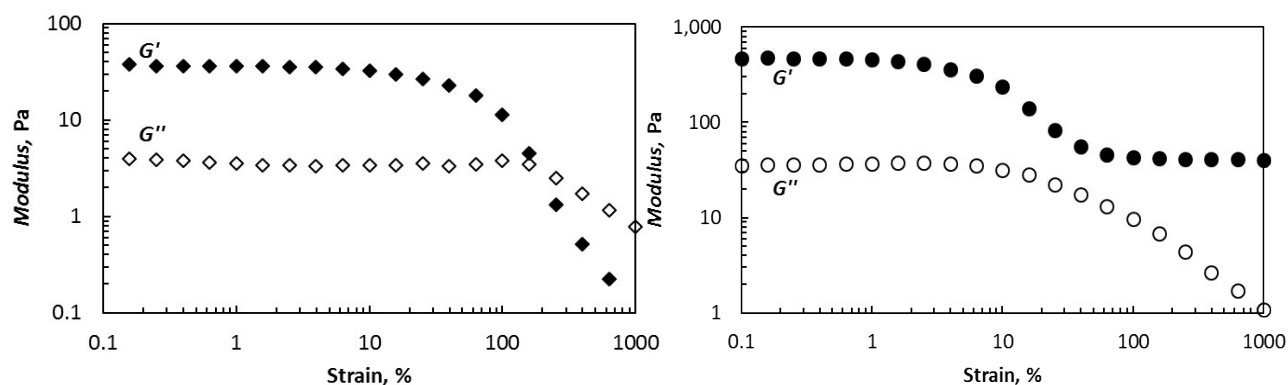


Figure S6. Strain sweep (6 rad.s⁻¹) of 1 wt % hydrogel of H4LMAX-RGDS (left) and H2LRDMAX (right) in 50 mM Tris buffer pH 7.4 with 30 mM NaCl at 37 °C after first dynamic time sweep measurement in Fig S4. G' values are represented by filled symbols and G'' by open symbols.

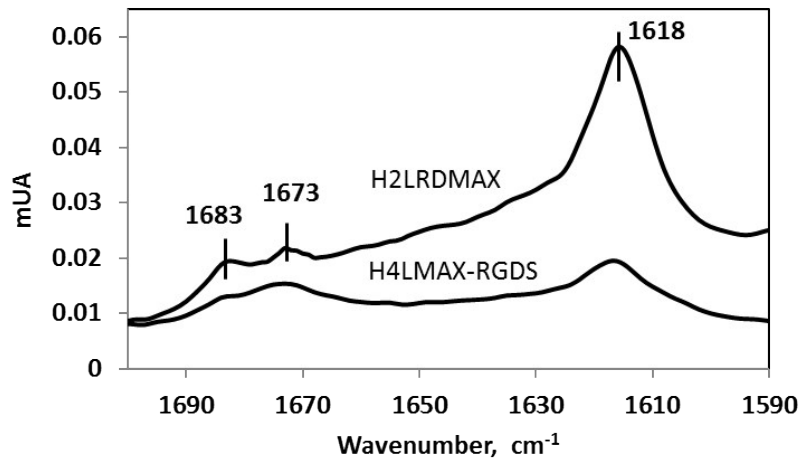


Figure S7. ATR-FTIR spectra of hydrogels at 1 wt % in D₂O with 50 mM Tris buffer pH 7.4 with 30 mM NaCl.

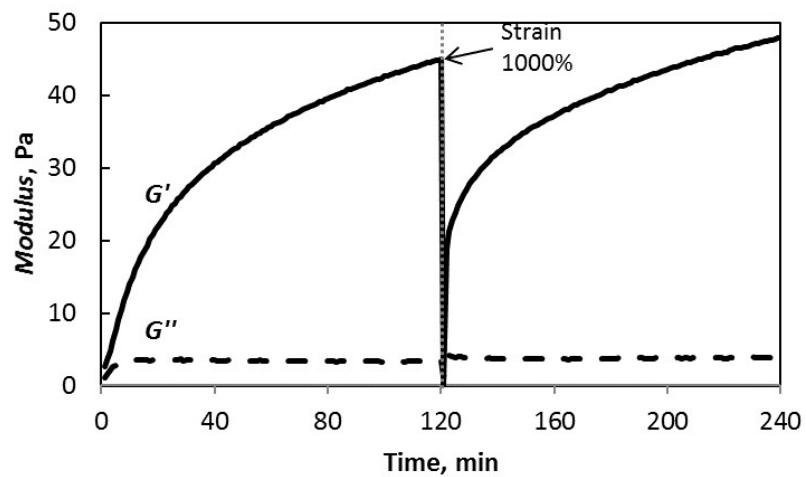


Figure S8. Storage (G') and loss modulus (G'') of 1 wt % H4LMAX-RGDS hydrogel containing 80 μ g of LF in 50 mM Tris buffer pH 7.4 with 30 mM NaCl at 37 °C as a function of time (6 rad.s⁻¹, 0.2% strain).

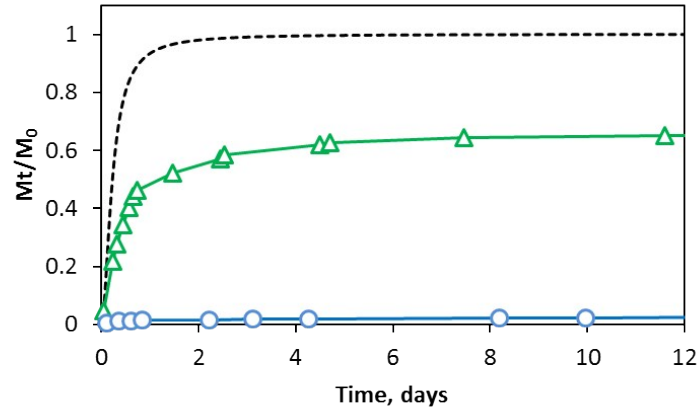


Figure S9. Cumulative LF release profiles from 1 wt % H4LMAX (blue open circles) and H4LRD MAX (green open triangles) hydrogels in 50 mM Tris buffer pH 7.4 with 150 mM NaCl at room temperature. The data points are connected with a solid line to guide the eye of the reader. Error bars are smaller than the symbol where not evident. The black dashed line represents a theoretical LF release scenario driven by the mass action law. Dissolution of the hydrogels was not observed during the experiments.

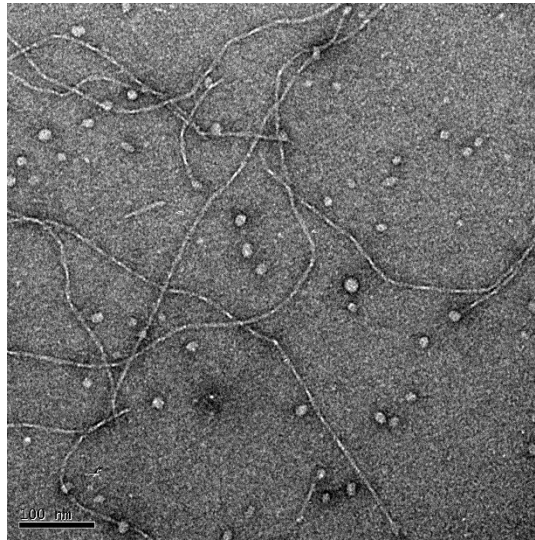


Figure S10. Transmission electron micrographs of a sample of H4LMAX-RGDS with LF 50-fold water diluted from a 1 wt % hydrogel in 50 mM Tris buffer pH 7.4 with 30 mM NaCl and 80 μ g of LF. The black scale bar corresponds to 100 nm.