Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics.

Supporting Information

Ligand Binding of Interleukin-8: A Comparison of Glycosaminoglycans and Acidic Peptides

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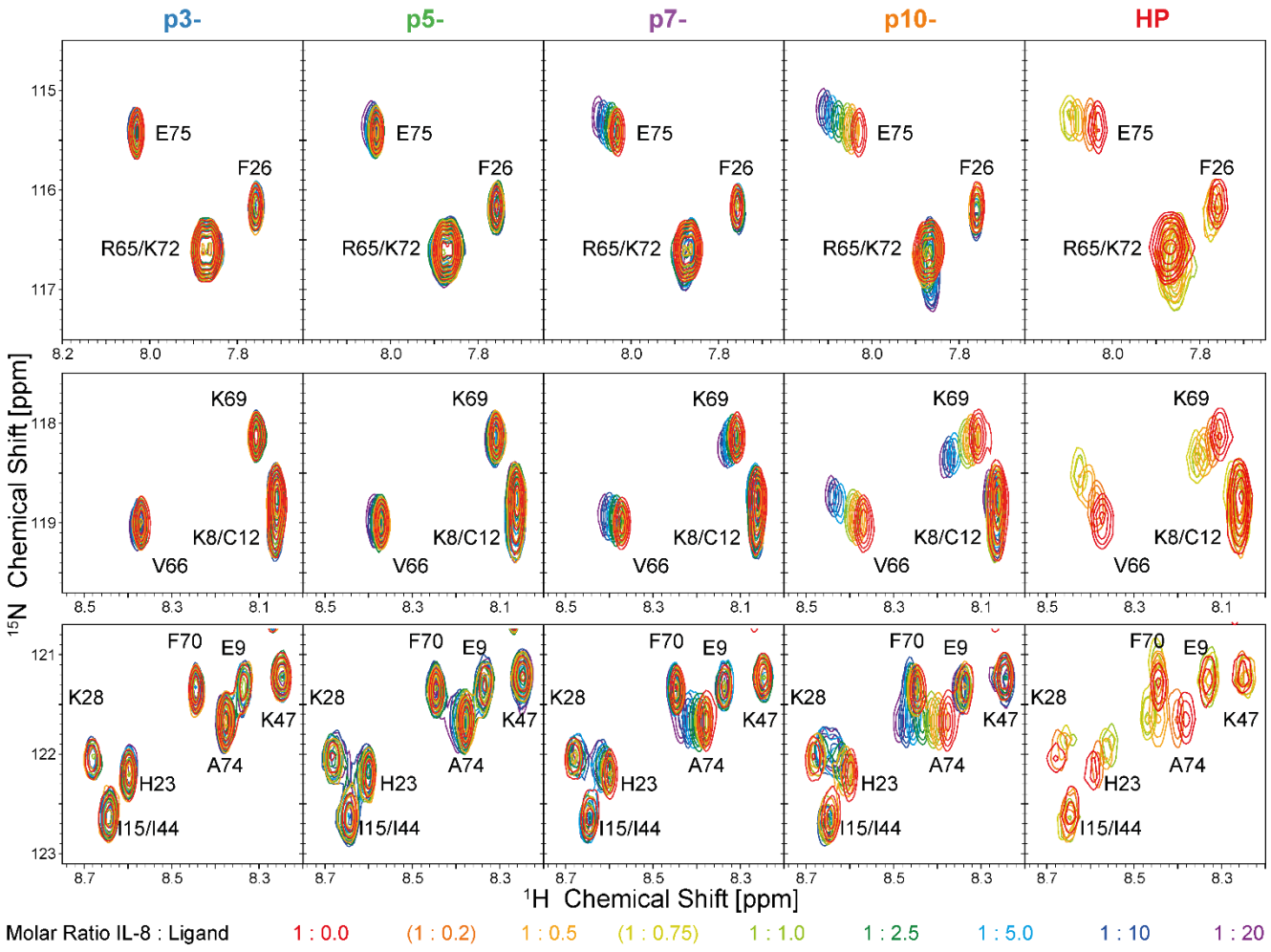
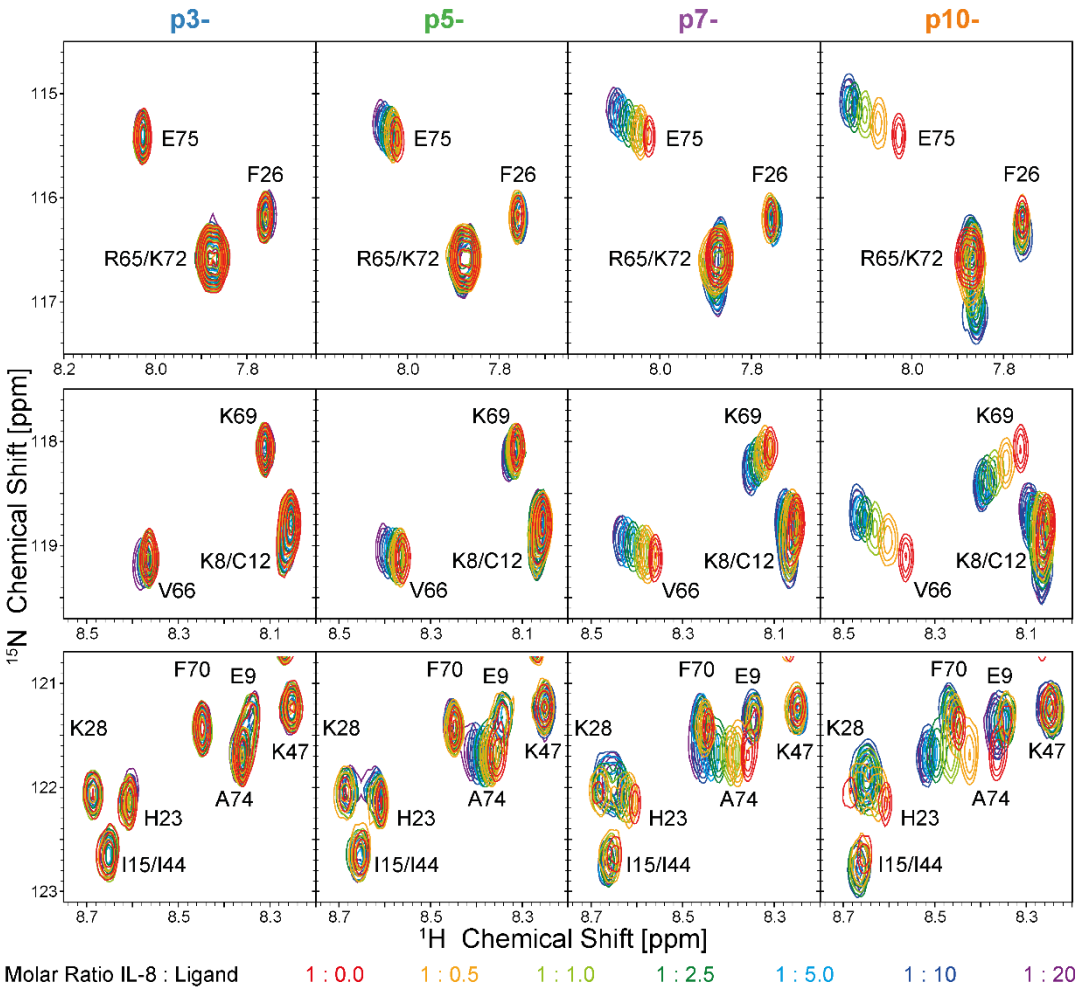
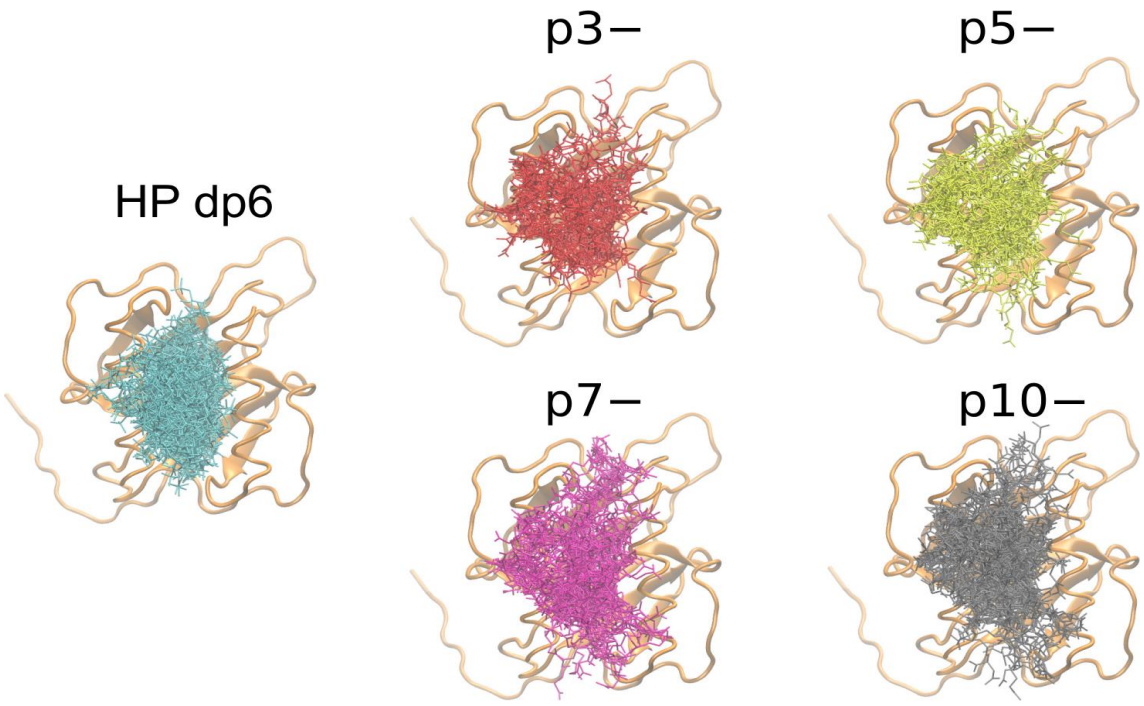


Supplementary Figure S1. Different regions of ^{1}H-^{15}N HSQC NMR spectra of ^{15}N-labeled IL-8 (100 µM, red) in buffer with 50 mM NaCl in the presence of the ligands p3-, p5-, p7-, p10- and HP dp6 (from left to right). Different molar ratios of IL-8 to ligand are shown (red 0, orange 0.5, light green 1, dark green 2.5 light blue 5, dark blue 10 and purple 20 fold molar excess of the peptide ligands). For HP dp6 the final titration step was at equimolar ratio, hence two additional titrations at lower molar ratio were performed (dark orange 0.2 and yellow-green 0.75, in parentheses).

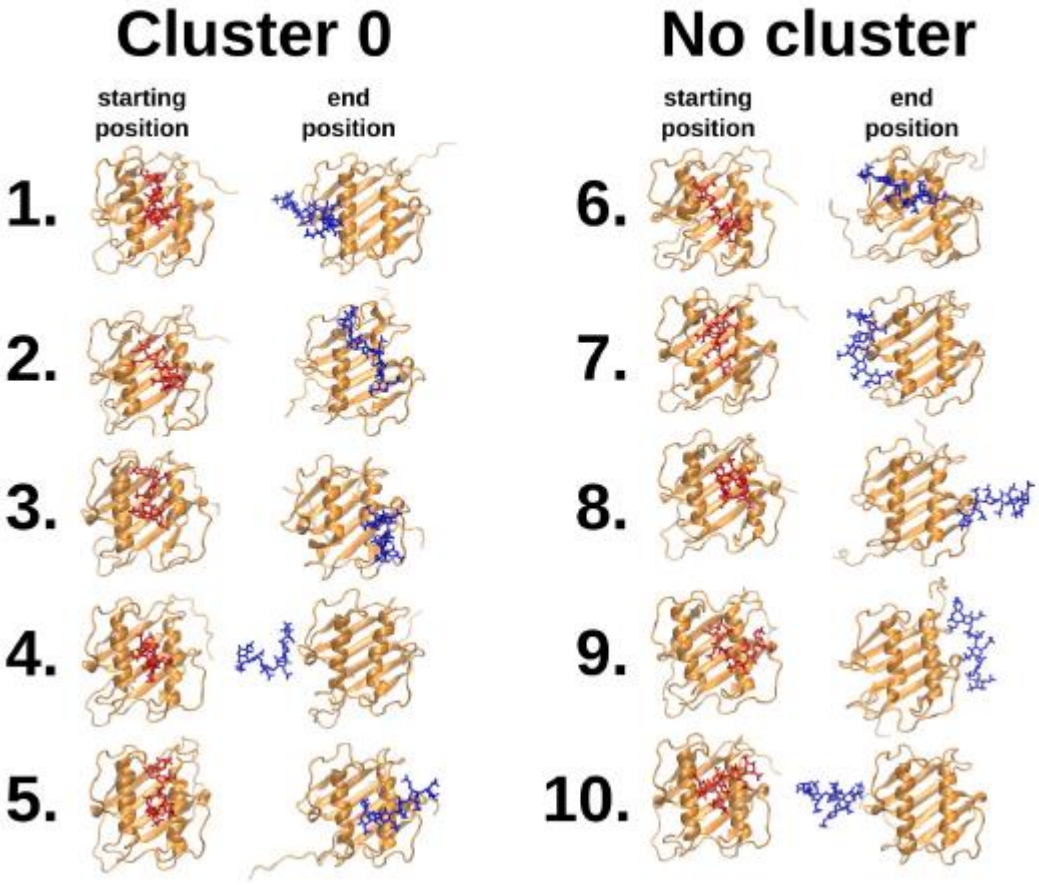


Supplementary Figure S2. Different regions of ^{1}H-^{15}N HSQC NMR spectra of ^{15}N-labeled IL-8 (100 µM, red) in buffer without NaCl in the presence of the ligands p3-, p5-, p7-, p10- (from left to right). Different molar ratios of IL-8 to acidic peptide ligands are shown (red 0, orange 0.5, light green 1, dark green 2.5 light blue 5, dark blue 10 and purple 20 fold molar excess of the peptide ligands).



Supplementary Figure S3. The 50 top-scoring docked poses for IL-8 complexed with HP dp6 (in cyan), p3– (in red), p5– (in yellow), p7– (in magenta), p10– (in grey). The homodimeric protein is shown in cartoon and the ligands in sticks.

A. Heparin


Cluster 0
No cluster
starting position
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2.
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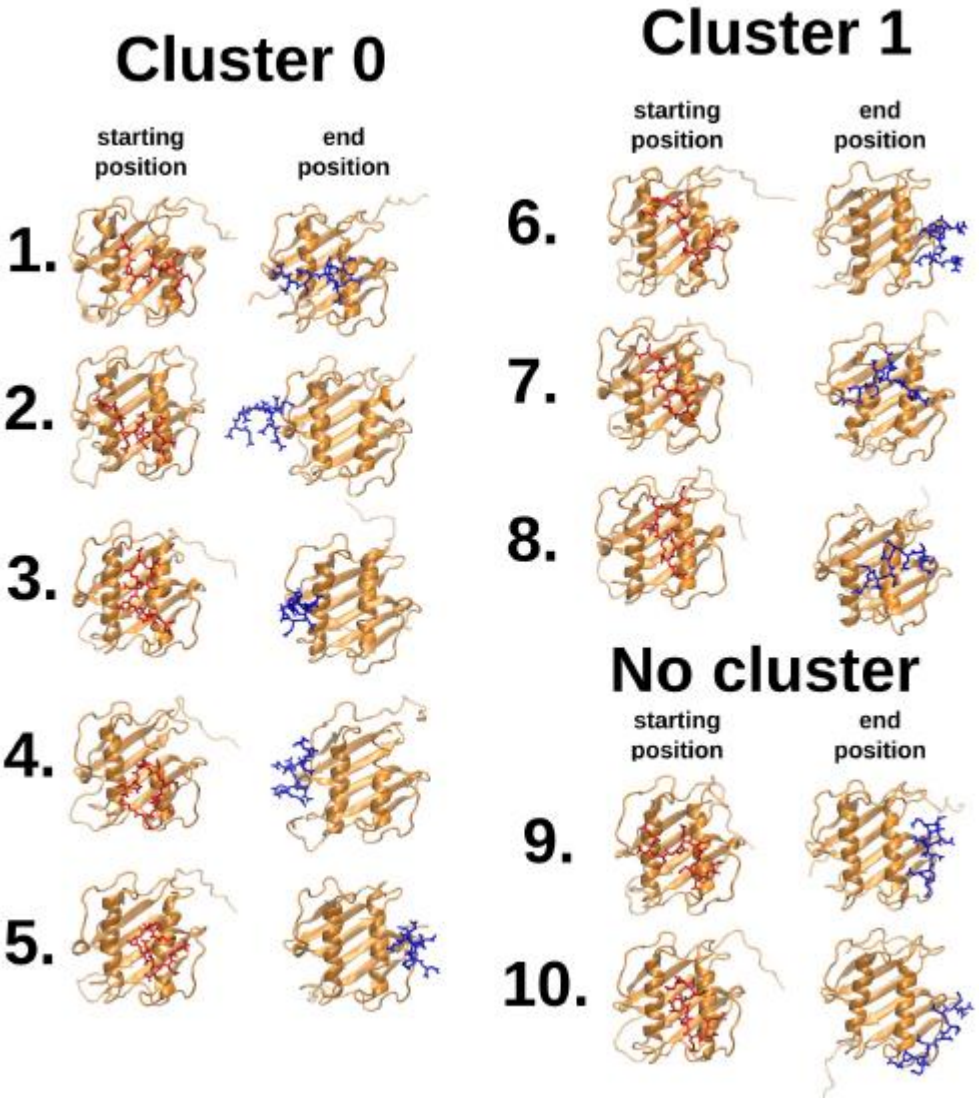
B. p10–


Cluster 0
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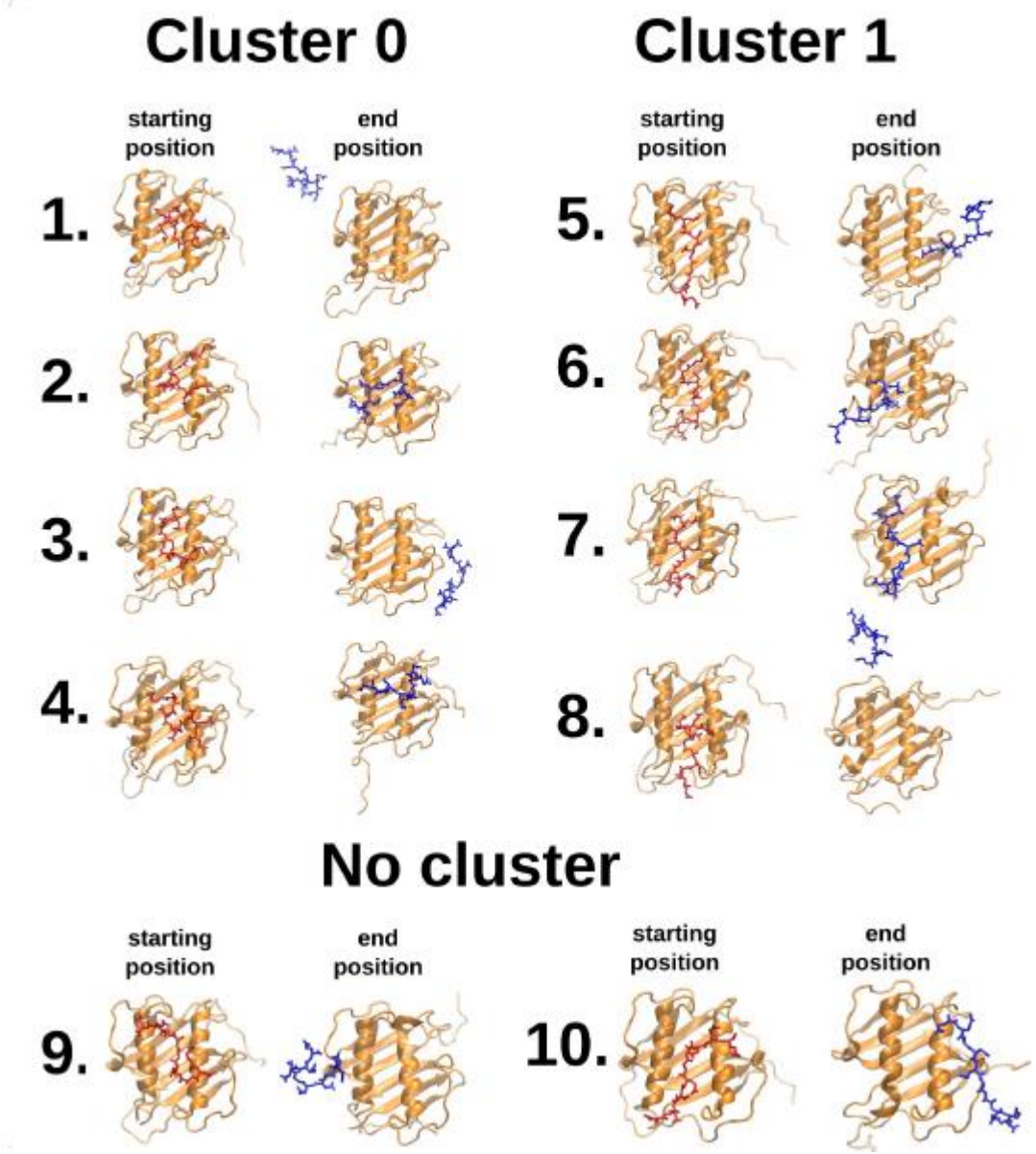
C. p7–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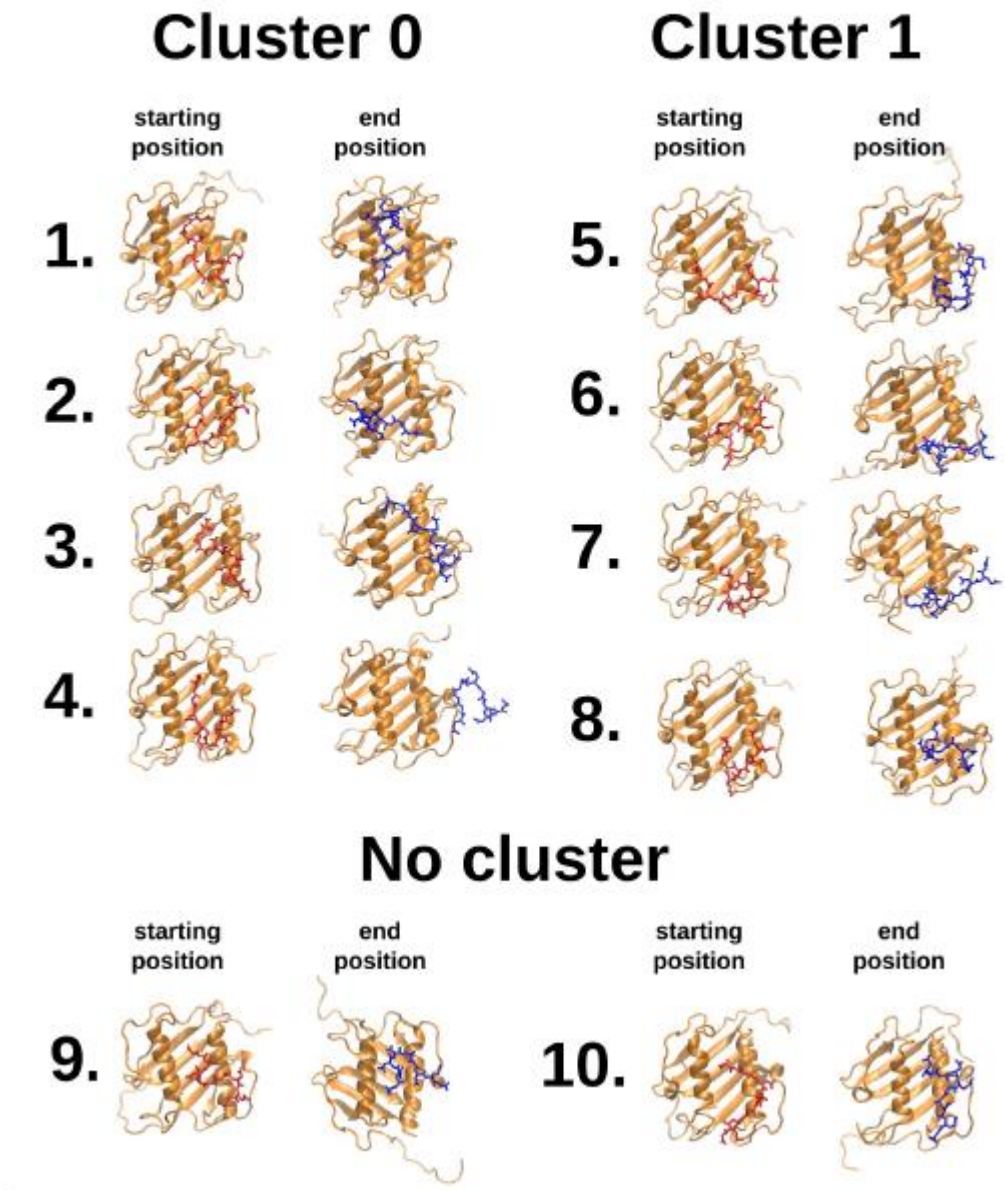
D. p5–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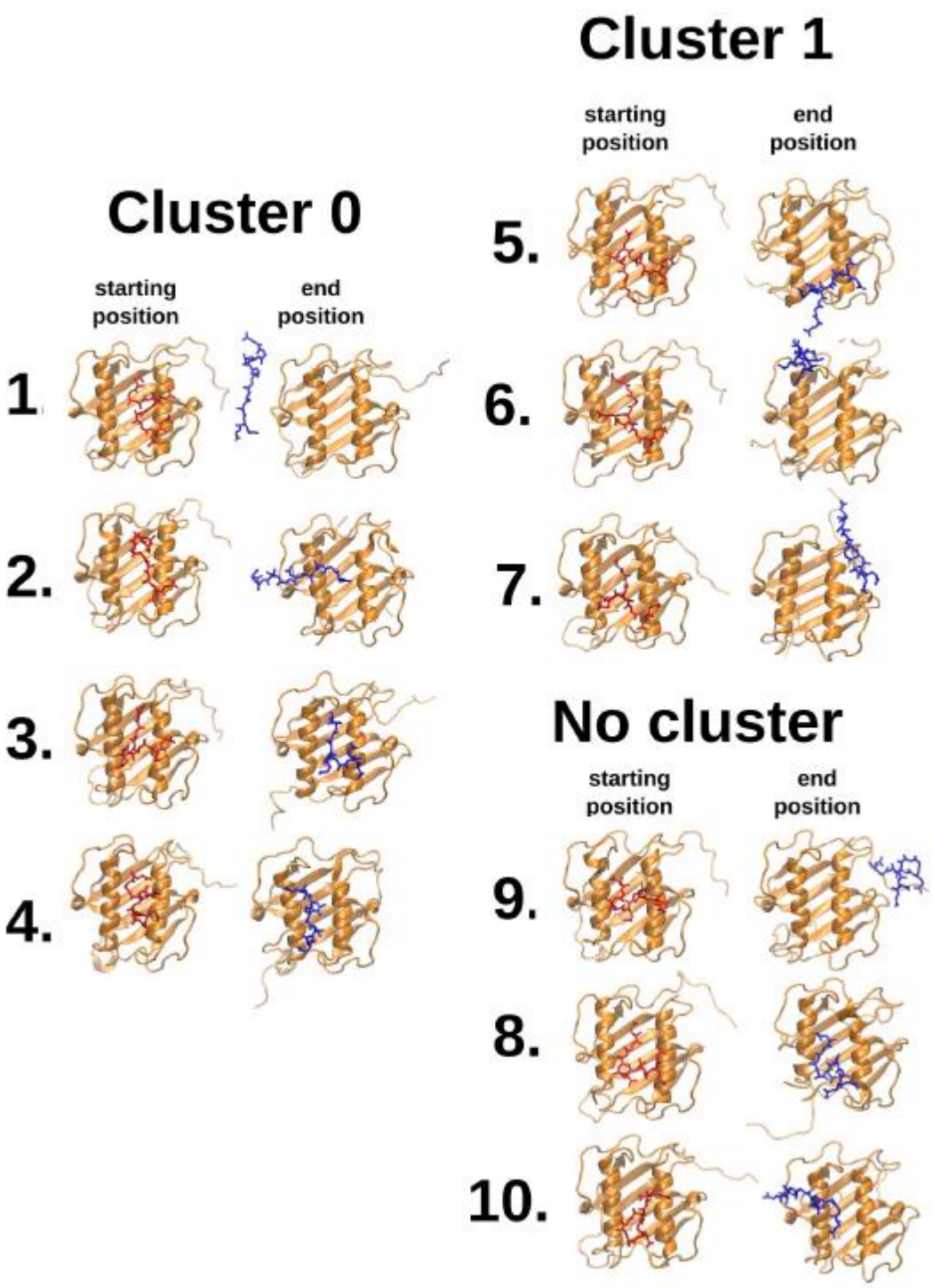
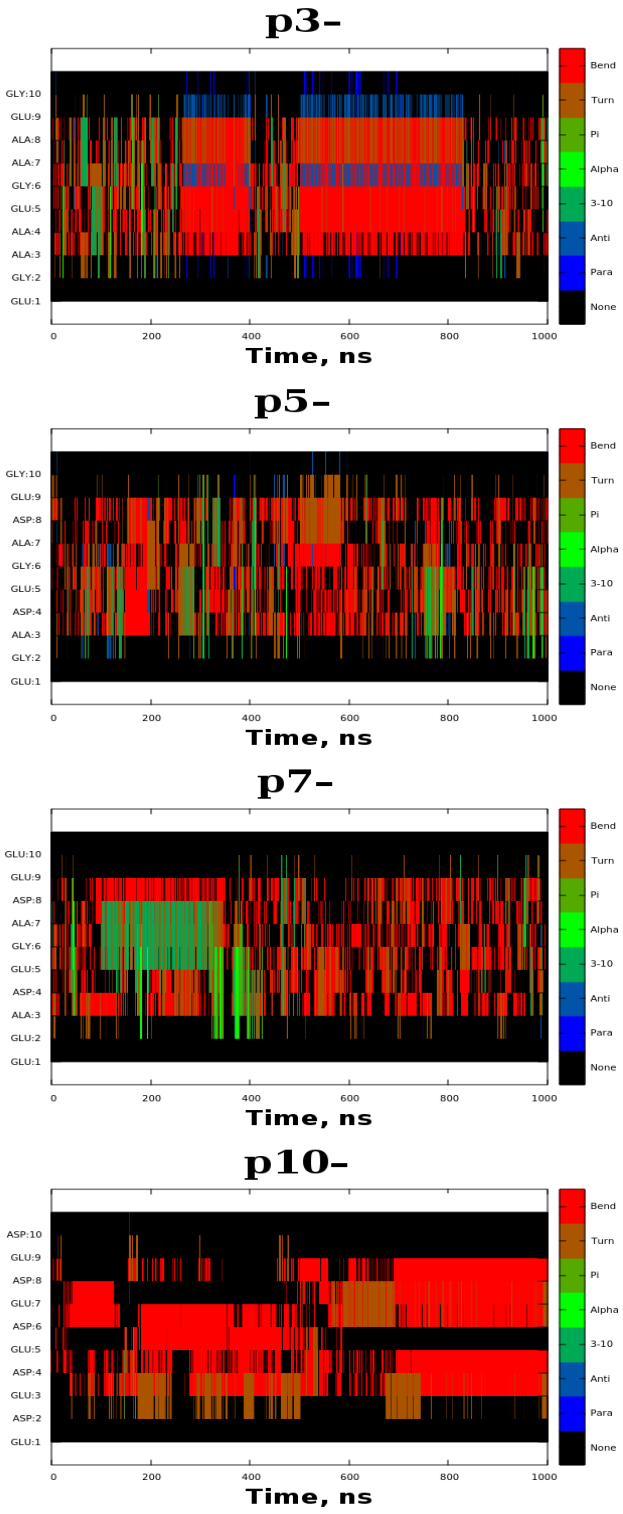
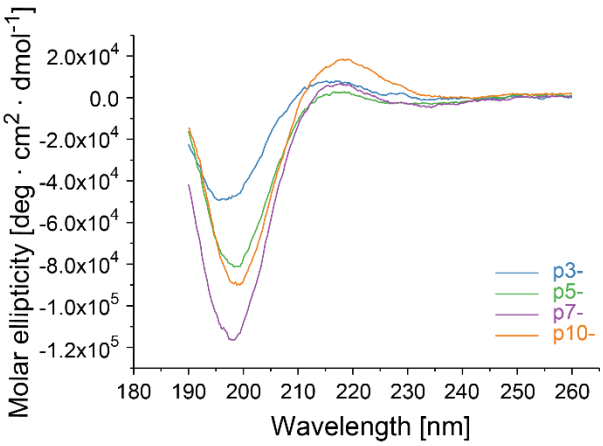
E. p3–



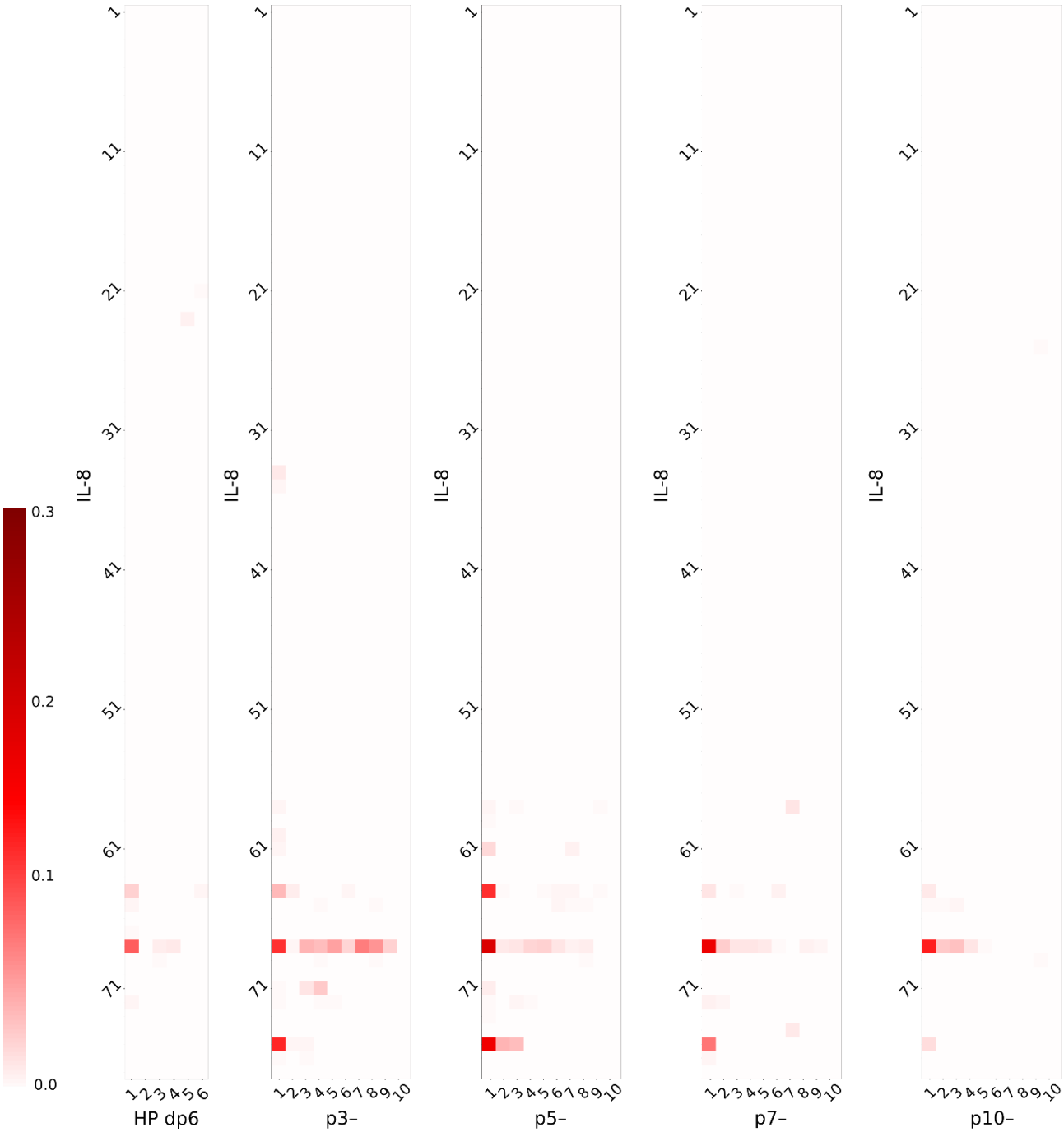
Supplementary Figure 4. The initial and final poses of the 10 MD simulations of IL-8 complexed with: A) HP, B) p10–, C) p7–, D) p5– and E) p3–. The protein is shown in cartoon representation and the ligands in stick representation, colored according to MD simulation step (initial pose in red, final pose in blue).



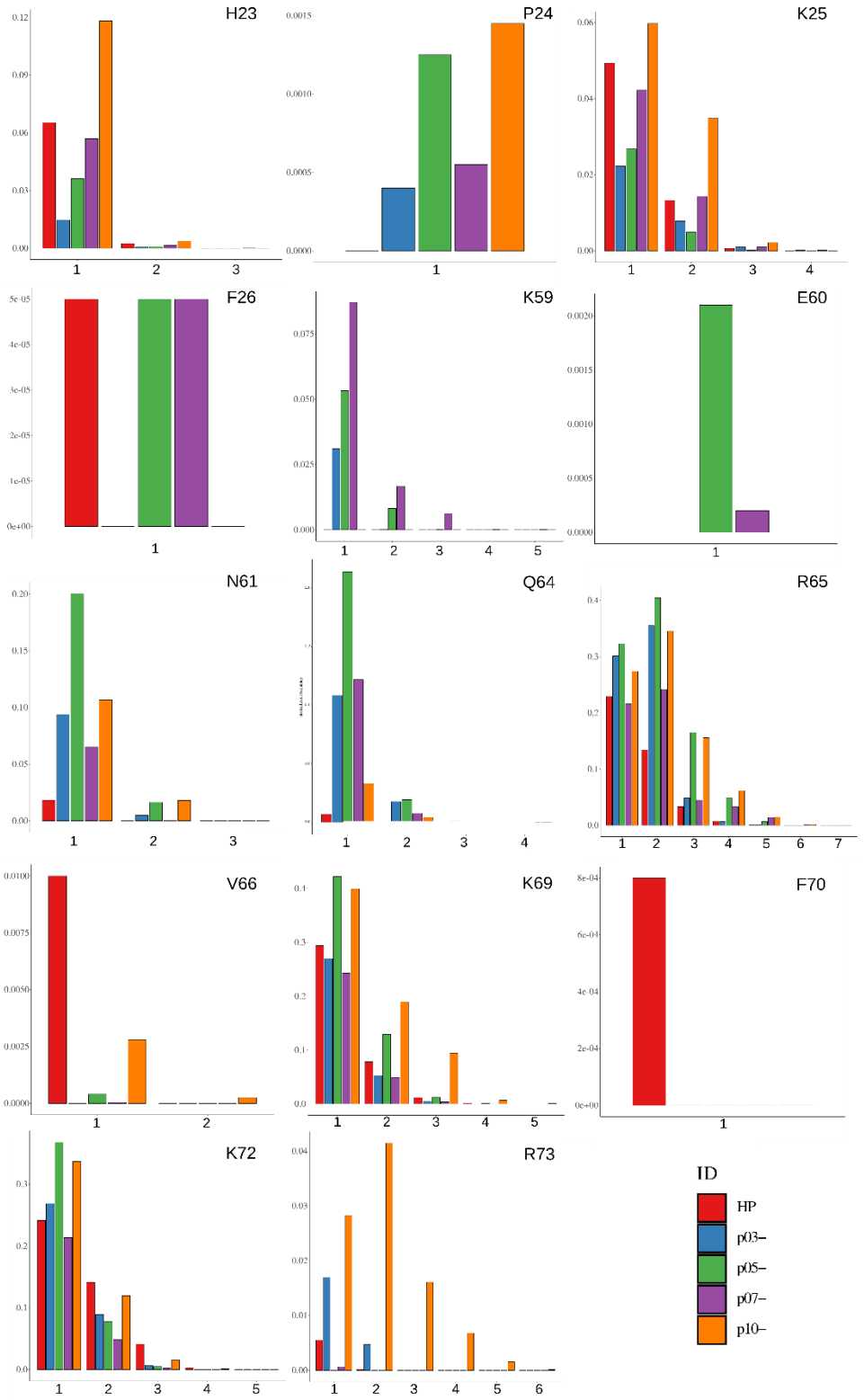
Supplementary Figure S5. Secondary structure propensities for the unbound acidic peptide ligand residues in the 1 μs MD simulation.



Supplementary Figure S6. Circular dichroism spectra of the four peptides indicating random coil structure for each peptide in buffer solution.



Supplementary Figure S7. Heatmap of hydrogen bond interactions between IL-8 (hydrogen atom acceptor) and HP dp6, p3–, p5–, p7– and p10–, averaged over IL-8 dimer subunits and across replicates of MD simulations. Protein residue numbers are shown on the x-axis, ligand residue numbers are shown on the y-axis. Color intensity corresponds to the frequency of the H-bond, i.e. the average fraction of MD trajectory frames in which the bond was formed.



Supplementary Figure S8. Frequency of H-bonds per frame for 14 IL-8 residues with most H-bonds as donors. Since the frequency was obtained by data normalization for all frames (including the ones where no H-bonds were formed), y-axes have different scales for each IL-8 residue. X-axis shows the number of H-bonds established by frame.