Electronic Supplementary Material (ESI) for Nanoscale.

Supplementary Information

SERS nanostructures with engineered active peptides against an immune checkpoint protein

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Table S1 – Peptides characterization

Ligand	**Sequence**	**HPLC** t_R [min]	**ESI-MS** Expect.	**$[M+H]^+$** Found
L1	Ac-Cys-O_2oc-O_2oc-Trp-His-Arg-Ser-Tyr-Tyr-Thr-Trp-Asn-Leu-Asn-Thr-NH_2	20.3 [a]	2074.9	2074.8
L2	Ac-Trp-His-Arg-Ser-Tyr-Tyr-Thr-Trp-Asn-Leu-Asn-Thr-O_2oc-O_2oc-Cys-NH_2	20.6 [a]	2074.9	2074.8
P3	Lys-Lys-Lys-Gly-Gly-Trp-His-Arg-Ser-Tyr-Tyr-Thr-Trp-Asn-Leu-Asn-Thr-NH_2	17.2 [a]	2138.1	2138.9
P4	Lys-Lys-Lys-Gly-Gly-Thr-Arg-Trp-Ser-His-Tyr-Asn-Thr-Leu-Trp-Tyr-Asn-NH_2	17.8 [a]	2138.1	2138.2
L3	HS-PEG-Lys-Lys-Lys-Gly-Gly-Trp-His-Arg-Ser-Tyr-Tyr-Thr-Trp-Asn-Leu-Asn-Thr-NH_2	19.3 [b]	> 5000	-[c]
L4	HS-PEG-Lys-Lys-Lys-Gly-Gly-Thr-Arg-Trp-Ser-His-Tyr-Asn-Thr-Leu-Trp-Tyr-Asn-NH_2	18.9 [b]	> 5000	-[c]

[a] Elution conditions: 5-50% B in 30 min.
[b] Elution conditions: 20-60% B in 30 min; broad peaks centered at the t_R
[c] See Figure S1 and Figure S2.

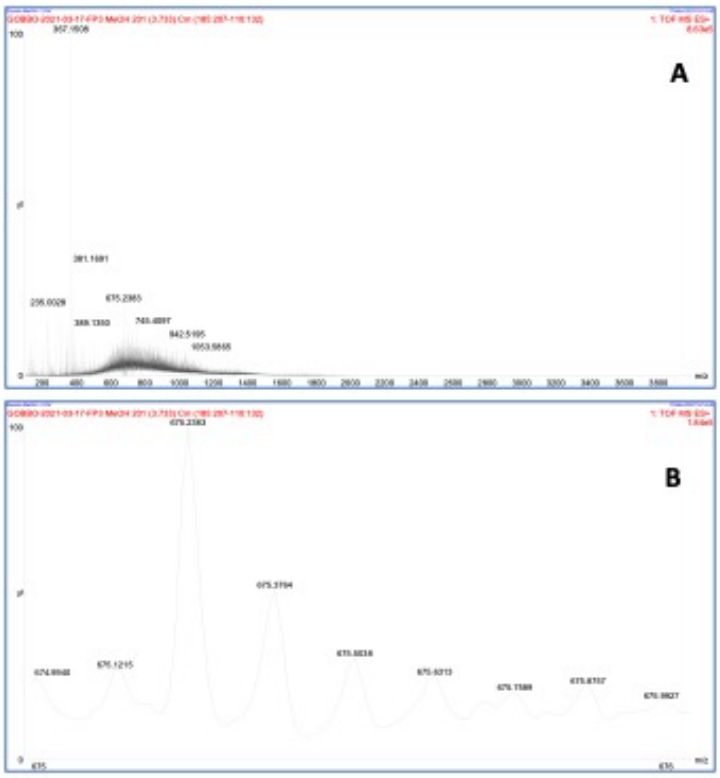

Figure S1. ESI(+)-Mass spectrum of the ligand 3. **A**: Full spectrum; **B**: Zoom of the region corresponding to $[M+8H]^{8+}$ ions

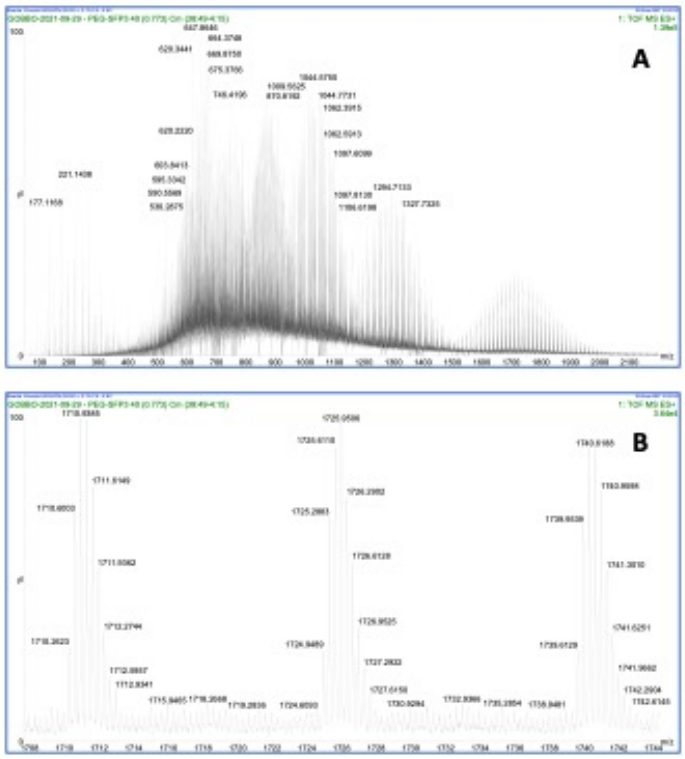

Figure S2. ESI(+)-Mass spectrum of L4. **A**: Full spectrum; **B**:Zoom of the region corresponding to $[M+3H]^{3+}$ ions.

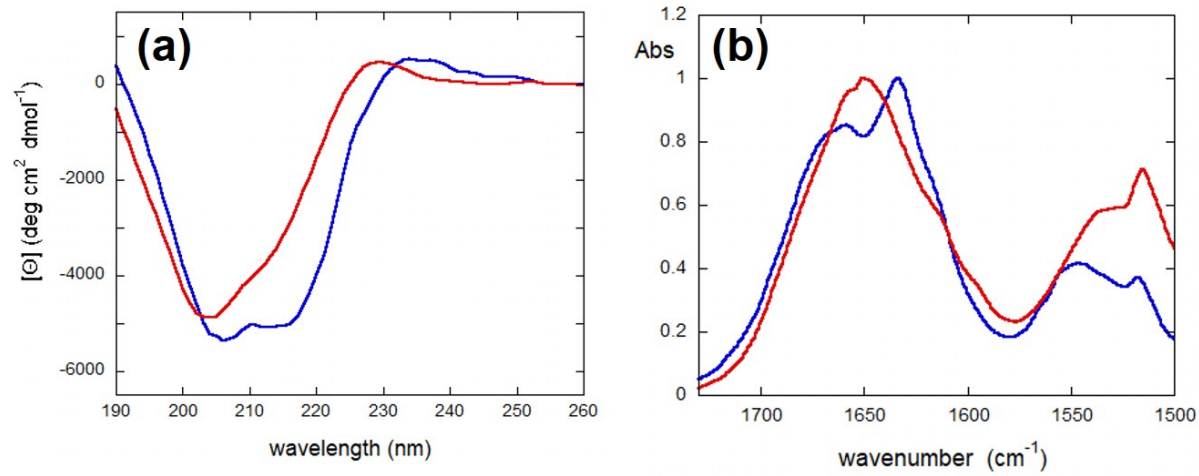

Figure S3. A) Far UV CD and B) FTIR spectra of CLP002 (blue line) and *sCLP002* (red line) in water. The molar ellipticity refers to the concentration of amino acid residues.

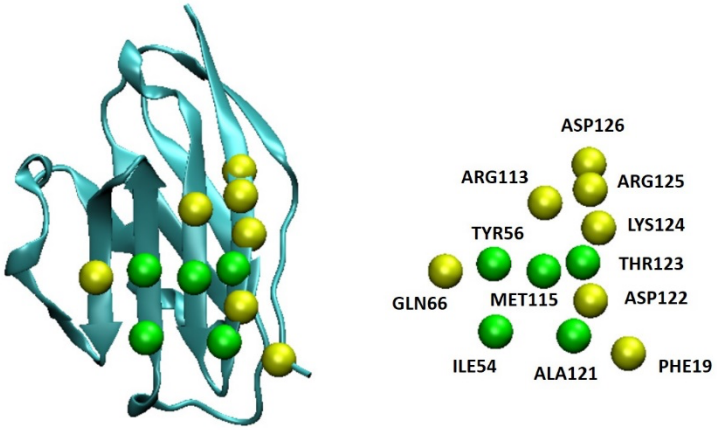

Figure S4. PD-L1 protein residues stabilizing the PD-1/PDL-1 complex. Residues belonging to the hydrophobic core[1] are in green.

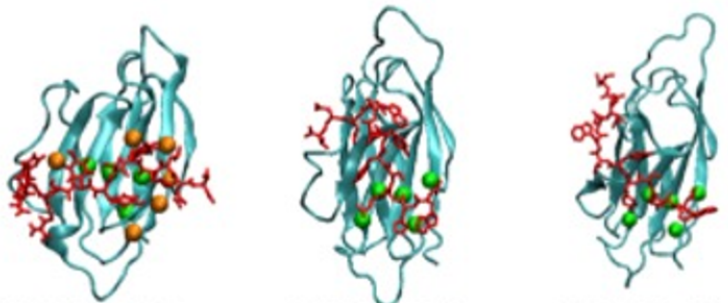

Figure S5. Structures of PD-L1 (light blue)/ CLP002 (red) adduct for the three replicas of the MD simulations. Green spheres are relative to the hydrophobic region of the protein.

References

1. K. M. Zak, R. Kitel, S. Przetocka, P. Golik, K. Guzik, B. Musielak, A. Domling, G. Dubin and T. A. Holak, *Structure*, 2015, **23**, 2341-2348.