Binding mechanism of oligopeptides on solid surface: Assessing the significance of single-molecule approach

Electronic Supplementary Material

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Content

S1. XPS data

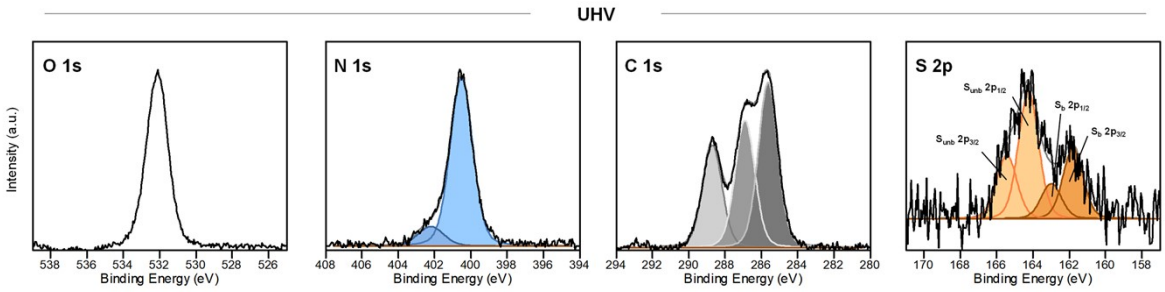


Figure S1. O 1s, N 1s, C 1s and S 2p XPS peaks recorded on gold surface after the adsorption of **C**$(KAAAA)_2$K**C** oligopeptide at high surface coverage in UHV using the ES-IBD method. Peaks are shown after the subtraction of the U 2 Tougaard baseline. In the S 2p peak, S_b and S_{unb} refer to bound thiolates and unbound thiols, respectively.

S2. PM-IRRAS data

PM-IRRAS spectra were recorded using a Nicolet 5700 spectrometer equipped with a nitrogen-cooled MCT wide-band detector. A ZnSe grid polarizer and ZnSe photoelastic modulator to modulate the incident beam between p and s polarization were placed prior to the sample. The spectrometer was interfaced to the UHV chamber via ZnSe windows. The reflected light was focused onto the detector at an optimal incident angle of 85°. All spectra were obtained after 1024 scans at 8 cm^{-1} resolution.

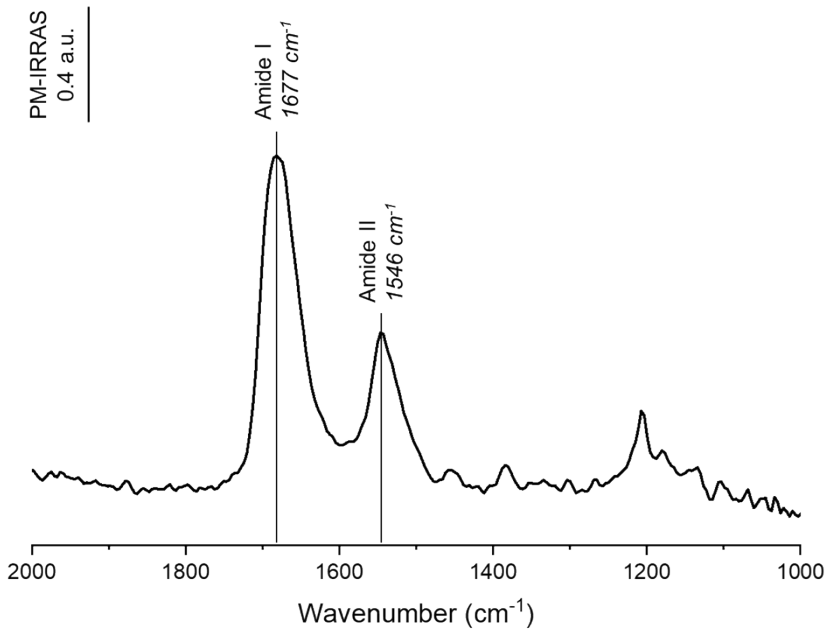


Figure S2. PM-IRRAS spectra recorded on Au(111) surface after the adsorption of $C(KAAAA)_2KC$ oligopeptide using ES-IBD.

S3. DFT complementary results

Table S1. Fragments of the oligopeptide after geometry optimization; these fragments are adsorbed on the Au(111) surface.

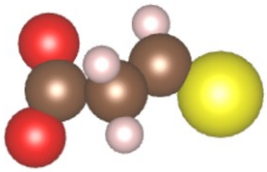
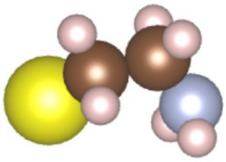
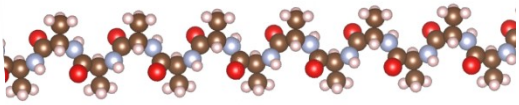
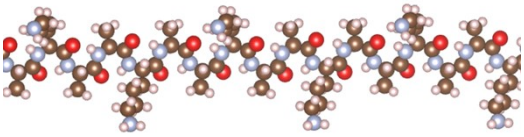
Fragment formula and chemical states of the moieties	**Geometry after optimization**
COO^--CH_2-CH_2-S^- $-S^-$, $-COO^-$	
S^--CH_2-CH_2-NH_2 $-S^-$, $-NH_2$	
$(CO\text{-}NH\text{-}CH\text{-}CH_3)_n$ $(Ala)_n$	
$((CO\text{-}NH\text{-}CH\text{-}CH_3)_2\text{-}CO\text{-}NH\text{-}CH\text{-}(C_4H_{10}N))_n$ $(Ala\text{-}Ala\text{-}Lys)_n$	

Table S2. Geometrical details of adsorbed fragments on Au(111)

Fragment formula	**Geometrical details**
$COO\text{-}CH_2\text{-}CH_2\text{-}S$	O-Au = 2.38 ± 0.02 Å (on top adsorption) S-Au = 2.50 ±0.02 Å (bridge adsorption)
$S\text{-}CH_2\text{-}CH_2\text{-}NH_2$	S-Au = 2.52 ± 0.09 Å (bridge adsorption)
$(CO\text{-}NH\text{-}CH\text{-}CH_3)_n$	O -- Au = 2.7 Å N -- Au = 3.3 Å
$((CO\text{-}NH\text{-}CH\text{-}CH_3)_2\text{-}CO\text{-}NH\text{-}CH\text{-}(C_4H_{10}N))_n$	O -- Au = 3.0 Å N -- Au = 3.5 Å

S4. Force spectroscopy

S4.1. Biotin/Streptavidin control experiment

Streptavidin immobilization was achieved following a procedure based on previous work [1]. A 200 µL droplet of a 0.1 mg/mL streptavidin (Sigma-Aldrich) in 15 mM NaCl solution was added on a freshly cleaved mica sheet and incubated for 30 minutes. Then, the sample was thoroughly rinsed with a phosphate buffer, and if not mounted immediately, stored at 4°C for no more than 2 weeks.

The biotin/streptavidin system was used as reference to ensure good functionalization of the tip by the protocol used. The results showed on Fig. S3 allow to validate the functionalization protocol because (i) the F-D curves exhibit a predominance of single-event interactions and (ii) the force spectra plotted in function of the loading rate exhibit a typical Bell-Evans profile, thoroughly described in the literature. The fitting of the data with the Bell-Evans model enables the estimation of the following kinetic and thermodynamic parameters: x_t=0.33 nm and k_{off}=2.604 s^{-1}. The estimated value of k_{off} is in excellent agreement with the literature [2].

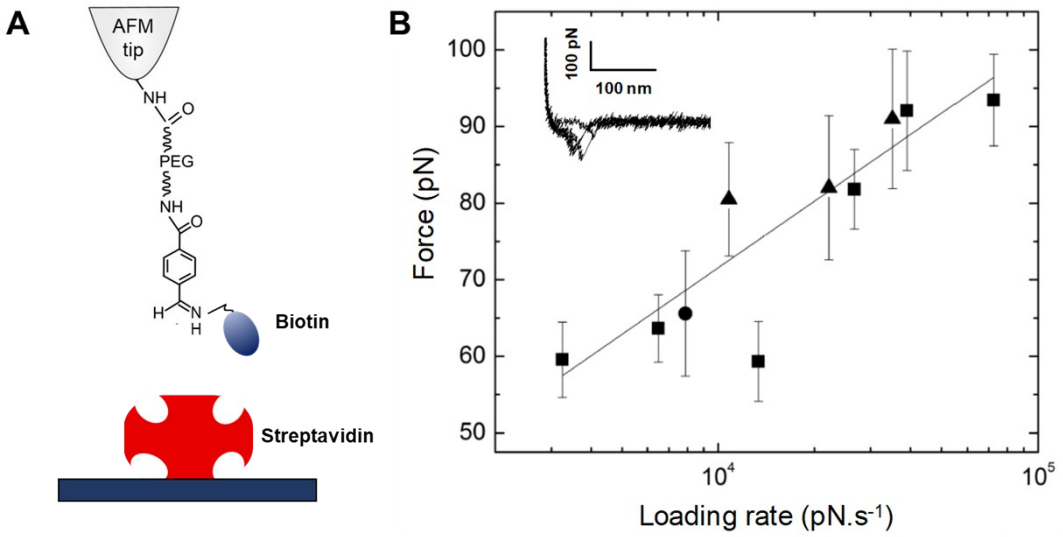


Figure S3. (A) Illustration of the biotin-streptavidin system with prior PEG linker functionalization. As biotin possesses its own linker, this additional step is not mandatory. The subsequent experiments were carried out with and without the coupling agent. (B) Force spectra plotted as a function of the loading rate for the biotin-streptavidin interaction. The fitting yields estimated parameters of x_t = 0.33 nm and k_{off} =2.604 s^{-1}.

S4.2. Oligopeptide/gold control experiments

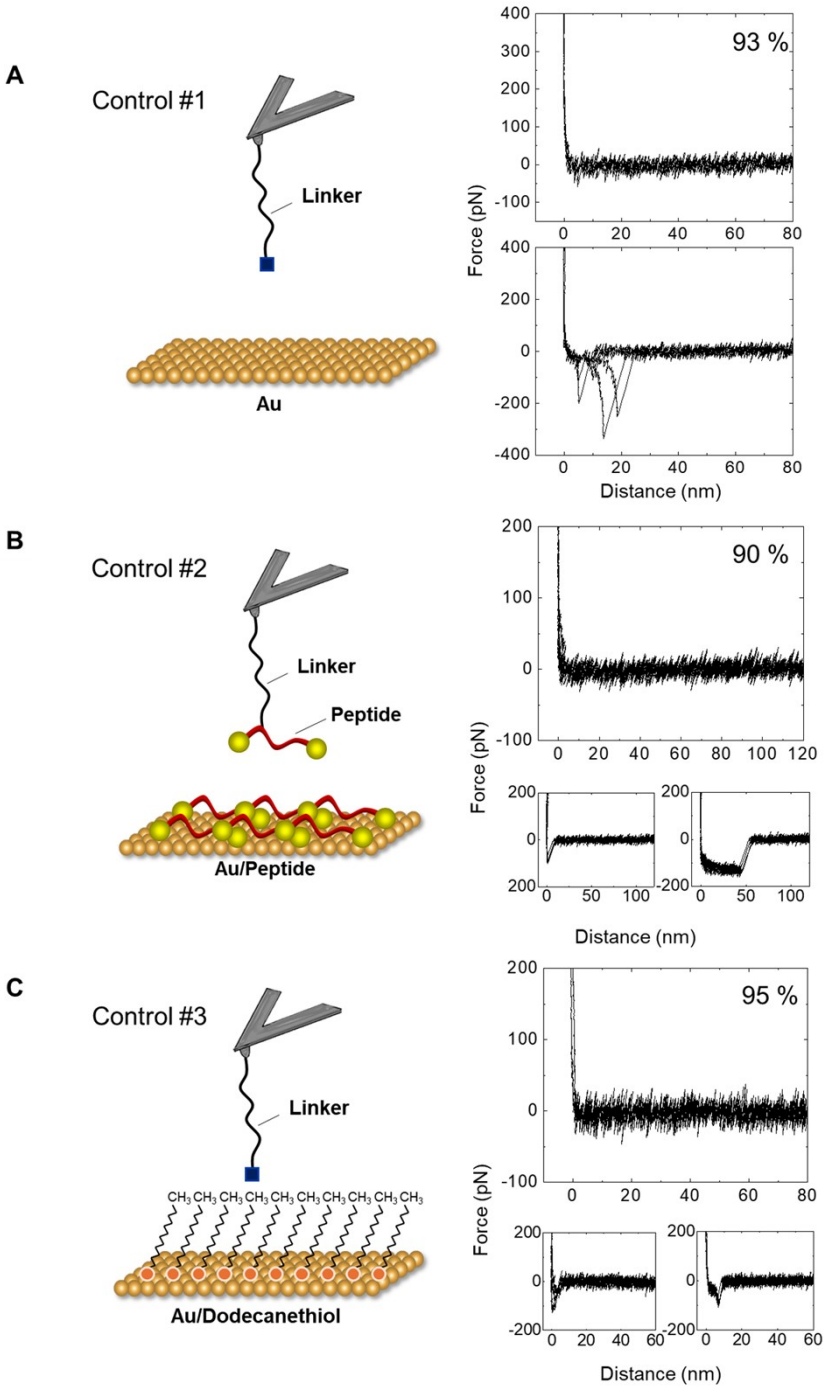


Figure S4. Scheme depicting the control tests (left) and their corresponding typical force-distance curves (right). (A) Control #1: PEG-gold (n=222), (B) Control #2: peptide-peptide (n=983) and (C) Control #3: PEG-dodecanethiol (n=300).

S5. Estimator of free energy from the Jarzynski equality

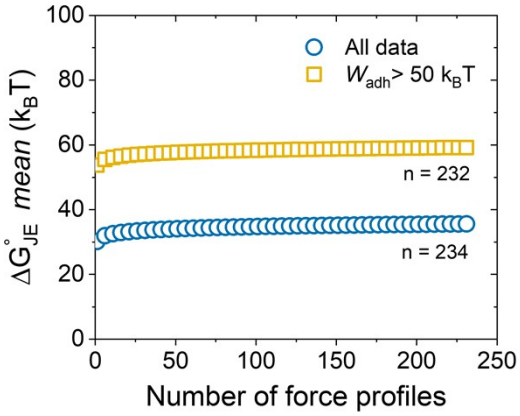


Figure S5. Convergence of the ΔG^{JE} obtained from a mean estimation in function of the number of force profiles used for **C**$(KAAAA)_2$**KC** oligopeptide. In blue all W_{adh} were used (n=234) and in yellow $W_{adh} < 50\ k_BT$ were excluded from the estimation (n=232).

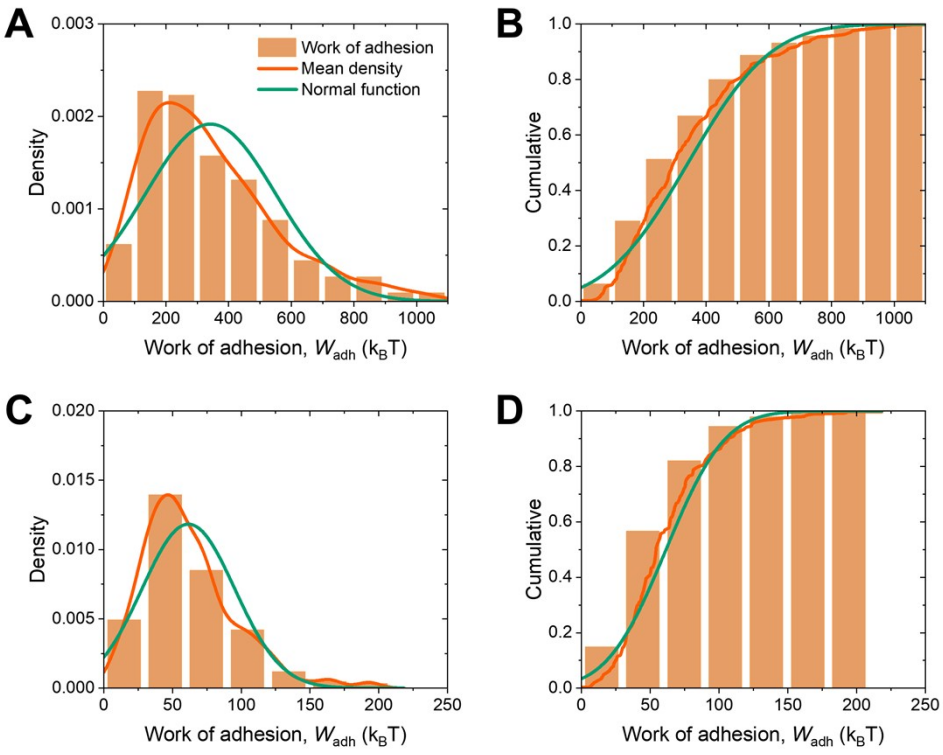


Figure S6. Frequency histograms of the work of adhesion for **C**$(KAAAA)_2$**KC** (A) and **G**$(KAAAA)_2$**KG** (C) oligopeptides extracted from n=234 F-D curves, fitted with a normal function (green curve). The nonparametric density estimation is also shown (red curve). Corresponding cumulative frequency histogram of the work of adhesion (n=234) fitted with a normal function (green curve) for **C**$(KAAAA)_2$**KC** (B) and **G**$(KAAAA)_2$**KG** (D) oligopeptides.

S6. AFM height imaging

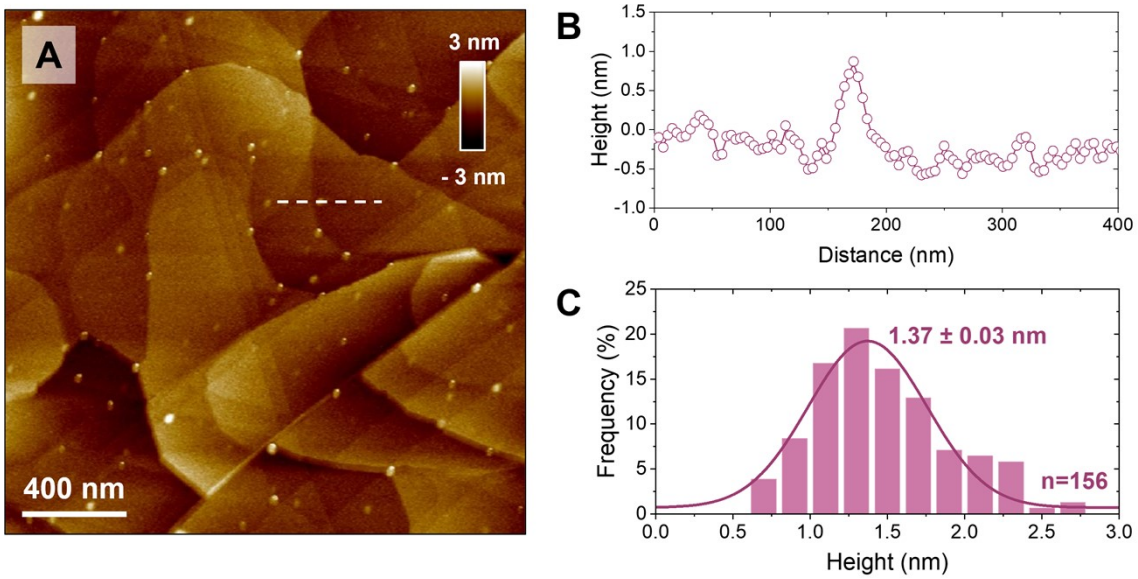


Figure S7. (A) Representative AFM height image recorded in aqueous solution after the adsorption of **G**$(KAAAA)_2$**KG** oligopeptide on gold surface. (B) Scan line taken at the location indicated in the inset of panel A by a dashed line. (C) Histogram of individual oligopeptide heights.

References

1. Ebner, A., et al., *Functionalization of probe tips and supports for single-molecule recognition force microscopy.* Top Curr Chem, 2008. **285**: p. 29-76.
2. Yuan, C., et al., *Energy Landscape of Streptavidin−Biotin Complexes Measured by Atomic Force Microscopy.* Biochemistry, 2000. **39**(33): p. 10219-10223.