Supporting Information

Correlation Between Desorption Force Measured by Atomic Force Microscopy (AFM) and Adsorption Free Energy Measured by Surface Plasmon Spectroscopy (SPR) for Peptide–Surface Interaction

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This supporting information contains (*i*) the data sheets of the parameters Q, K, and m determined by fitting equation (1) in the main manuscript to each of our peptide–SAM systems plots (i.e., the raw experimental data from SPR by plotting the changes in RU vs. peptide solution concentration) by non–linear regression using the Statistical Analysis Software program (SAS), and (*ii*) the resulting values from force–separation curves by AFM for F_{des}, and maximal separation distance corresponding to peptide–SAM systems presented.

The thermodynamic parameters (Q, K, and m) were then used to calculate ΔG^{o}_{ads} for each peptide–SAM combination. The following tables present these parameters for the designated peptide residues (TGTG–X–GTGT or TGTG–X–GTCT; X = L, V and D) on each of our modeled SAM surface (see

Tables 1-6). In these tables, ΔG^{o}_{ads} values present the experimentally measured standard state adsorption free energy for each peptide–SAM system, *m* is the proportionality constant between the peptide concentration in the bulk solution and the bulk shift in the SPR response, *K* is the effective equilibrium constant for the peptide adsorption reaction, and *Q* is amount of peptide adsorbed at surface saturation. The interested readers can obtain more detailed descriptions on analysis procedures in our previous publications on these studies.[1, 2]

The second part of this supporting information, which presents the force analyses for our AFM studies, is applied to measure the desorption–force for the TGTG–X–GTCT (X=L, V and D) peptide on selected surfaces. The following tables and figures present values and distributions of desorption force, F_{des}, and the maximal separation distance for designated peptide residues on each of our selected SAM surfaces (see Tables 7-9 and Figure 1-3) and material that are not amenable to being studied using SPR (Figure 4-5).

Table 1. Values of *m*, *Q*, *K* and ΔG^{o}_{ads} (mean ± 95% C.I.) for peptides TGTG–X–GTGT and TGTG–X–GTCT on SAM–OH surface with various functionalities (N=6).

–X–	ΔG^{o}_{ads} **(kcal/mole)**	***Q*** **(pg/mm^2)**	***K*** **(unitless)**	**m (RU/M)**
TGTG–X–GTGT				
–L–	–0.003 (0.001)	3.12 (0.57)	1.4 (0.4)	137594 (979)
–V–	–0.002 (0.001)	0.24 (0.11)	14.4 (2.3)	168000 (17000)
–D–	–0.003 (0.001)	0.15 (0.08)	23.7 (7.8)	171900 (18000)
TGTG–X–GTCT				
–L–	–0.002 (0.001)	0.16 (0.10)	35.8 (12)	137151 (1730)
–V–	–0.006 (0.002)	0.14 (0.05)	83.5 (13)	156927 (1453)
–D–	–0.003 (0.001)	0.33 (0.07)	15.7 (1.8)	177150 (40000)

Table 2. Values of *m*, *Q*, *K* and ΔG^{o}_{ads} (mean ± 95% C.I.) for peptides TGTG–X–GTGT and TGTG–X–GTCT on SAM–CH_3 (N=6).

–X–	**ΔG^{o}_{ads} (kcal/mole)**	***Q*** **(pg/mm^2)**	***K*** **(unitless)**	**m (RU/M)**
TGTG–X–GTGT				
–L–	–3.87 (0.7)	95 (22)	6951 (1115)	124311 (26569)
–V–	–4.40 (0.3)	79 (12)	19300 (9800)	177800 (3200)
–D–	–3.54 (0.6)	73 (20)	4940 (1980)	190200 (7400)
TGTG–X–GTCT				
–L–	–3.37 (0.3)	181 (19)	1794 (729)	148436 (22810)
–V–	–3.91 (0.25)	72 (19)	10705 (2703)	183129 (12480)
–D–	–3.38 (0.1)	107 (16)	2924 (850)	179516 (32354)

Table 3. Values of *m*, *Q*, *K* and ΔG^{o}_{ads} (mean ± 95% C.I.) for peptides TGTG–X–GTGT and TGTG–X–GTCT on SAM–$NHCOCH_3$ (N=6).

–X–	**ΔG^{o}_{ads} (kcal/mole)**	***Q*** **(pg/mm^2)**	***K*** **(unitless)**	**m (RU/M)**
TGTG–X–GTGT				
–L–	–1.04 (0.3)	163 (60)	28 (10)	118415 (6671)
–V–	–0.16 (0.1)	13 (7)	15 (10)	158334 (4223)
–D–	–1.93 (0.5)	25 (6)	1100 (500)	153823 (17388)
TGTG–X–GTCT				
–L–	–1.53 (0.2)	161 (49)	88.6 (40)	142309 (15420)
–V–	–0.38 (0.1)	2.18 (0.1)	399 (80)	192631 (31034)
–D–	–1.79 (0.5)	322 (48)	61 (4.4)	177028 (17590)

Table 4. Values of *m*, *Q*, *K* and ΔG^{o}_{ads} (mean ± 95% C.I.) for peptides TGTG–X–GTGT on SAM–OCH_2CF_3 (N=6).

–X–	**ΔG^{o}_{ads} (kcal/mole)**	***Q*** **(pg/mm^2)**	***K*** **(unitless)**	**m (RU/M)**
TGTG–X–GTGT				
–L–	–3.09 (0.31)	120 (42)	1121 (430)	133629 (9315)
–V–	–3.99 (0.22)	112 (36)	7253 (3273)	161699 (3136)
–D–	–3.59 (0.37)	134 (43)	2924 (1706)	165166 (11303)

Table 5. Values *of m*, *Q*, *K* and ΔG^{o}_{ads} (mean ± 95% C.I.) for peptides TGTG–X–GTGT on SAM–NH_2 (N=6).

–X–	ΔG^{o}_{ads} **(kcal/mole)**	***Q*** **(pg/mm^2)**	***K*** **(unitless)**	**m (RU/M)**
TGTG–X–GTGT				
–L–	–2.34 (0.80)	28 (12)	1683 (60)	137852 (18570)
–V–	–3.90 (0.12)	64 (24)	11686 (1281)	160172 (2118)
–D–	–3.75 (0.20)	53 (28)	9121 (6000)	171599 (10774)

Table 6. *Values* of *m*, *Q*, *K* and ΔG^{o}_{ads} (mean ± 95% C.I.) for peptides TGTG–X–GTGT on SAM–$(OCH_2CH_2)OH$ (N=6).

–X–	ΔG^{o}_{ads} **(kcal/mole)**	***Q*** **(pg/mm^2)**	***K*** **(unitless)**	**m (RU/M)**
TGTG–X–GTGT				
–L–	–0.40 (0.28)	42 (26)	20 (6)	135182 (1346)
–V–	–0.26 (0.06)	14 (5)	31 (13)	161803 (12948)
–D–	–0.44 (0.14)	55 (20)	23 (9)	165831 (4781)

Table 7. Desorption force *and* separation distance measurements for TGTG–L–GTCT on selected SAM–surfaces in PBS; pH=7.4 Mean (± 95% C.I.), N = 6. An asterisk (*) indicates negligible value. For desorption force, (*) especially represents the force lower than the sensitivity of our AFM measurement which is around 10 pN.

SAM	$\mathbf{F_{des}}$ **(pN)**	**Maximal separation (nm)**
SAM–OH	*	*
SAM–CH_3	100 (22)	32 (4)
SAM–OCH_2CF_3	77 (13)	33 (4)
SAM–$NHCOCH_3$	33 (8)	32 (5)
SAM–$(OCH_2CH_2)_3OH$	*	*
SAM–NH_2	58 (13)	33 (4)

Table 8. Desorption force and *separation* distance measurements for TGTG–V–GTCT on selected SAM–surfaces in PBS; pH=7.4 Mean (± 95% C.I.), N = 6. An asterisk (*) indicates negligible value. For desorption force, (*) especially represents the force lower than the sensitivity of our AFM measurement which is around 10 pN.

SAM	**F_{des} (pN)**	**Maximal separation (nm)**
SAM–OH	*	*
SAM–CH_3	107 (15)	25 (6)
SAM–OCH_2CF_3	90 (20)	26 (3)
SAM–$NHCOCH_3$	37 (8)	26 (3)
SAM–$(OCH_2CH_2)_3OH$	*	*
SAM–NH_2	85 (31)	23 (8)

Table 9. Desorption force and separation distance measurements for TGTG–D–GTCT on selected SAM–surfaces in PBS; pH=7.4 Mean (± 95% C.I.), N = 6. An asterisk (*) indicates negligible value. For desorption force, (*) especially represents the force lower than the sensitivity of our AFM measurement which is around 10 pN.

SAM	**F_{des} (pN)**	**Maximal separation (nm)**
SAM–OH	*	*
SAM–CH_3	98 (22)	28 (3)
SAM–OCH_2CF_3	98 (18)	26 (3)
SAM–$NHCOCH_3$	56 (14)	24 (6)
SAM–$(OCH_2CH_2)_3OH$	*	*
SAM–NH_2	94 (28)	23 (6)

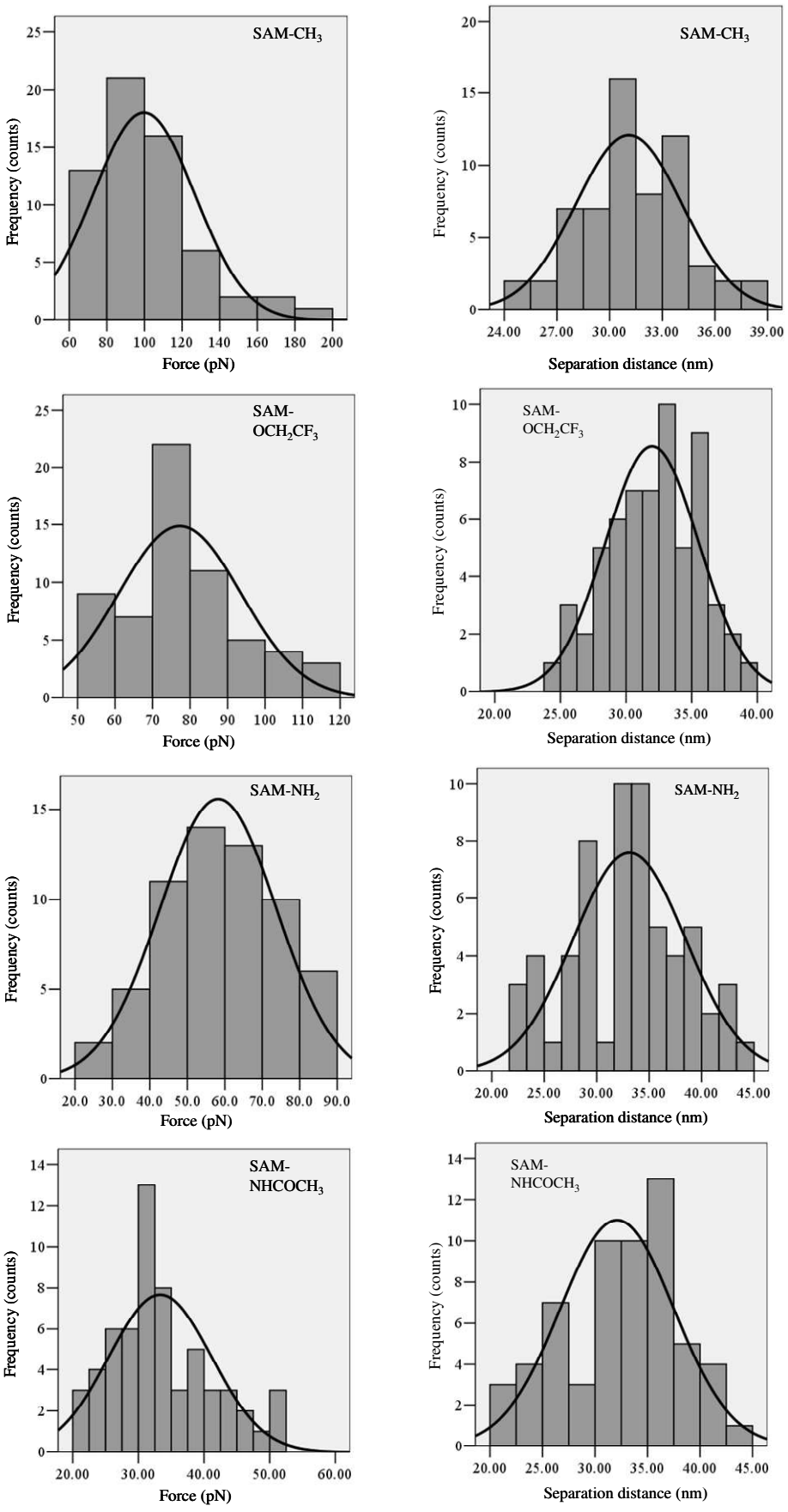

Figure 1. Distribution histogram of desorption force and separation distance from 60 measurements for TGTG–L–GTCT on selected SAM–surfaces in PBS.

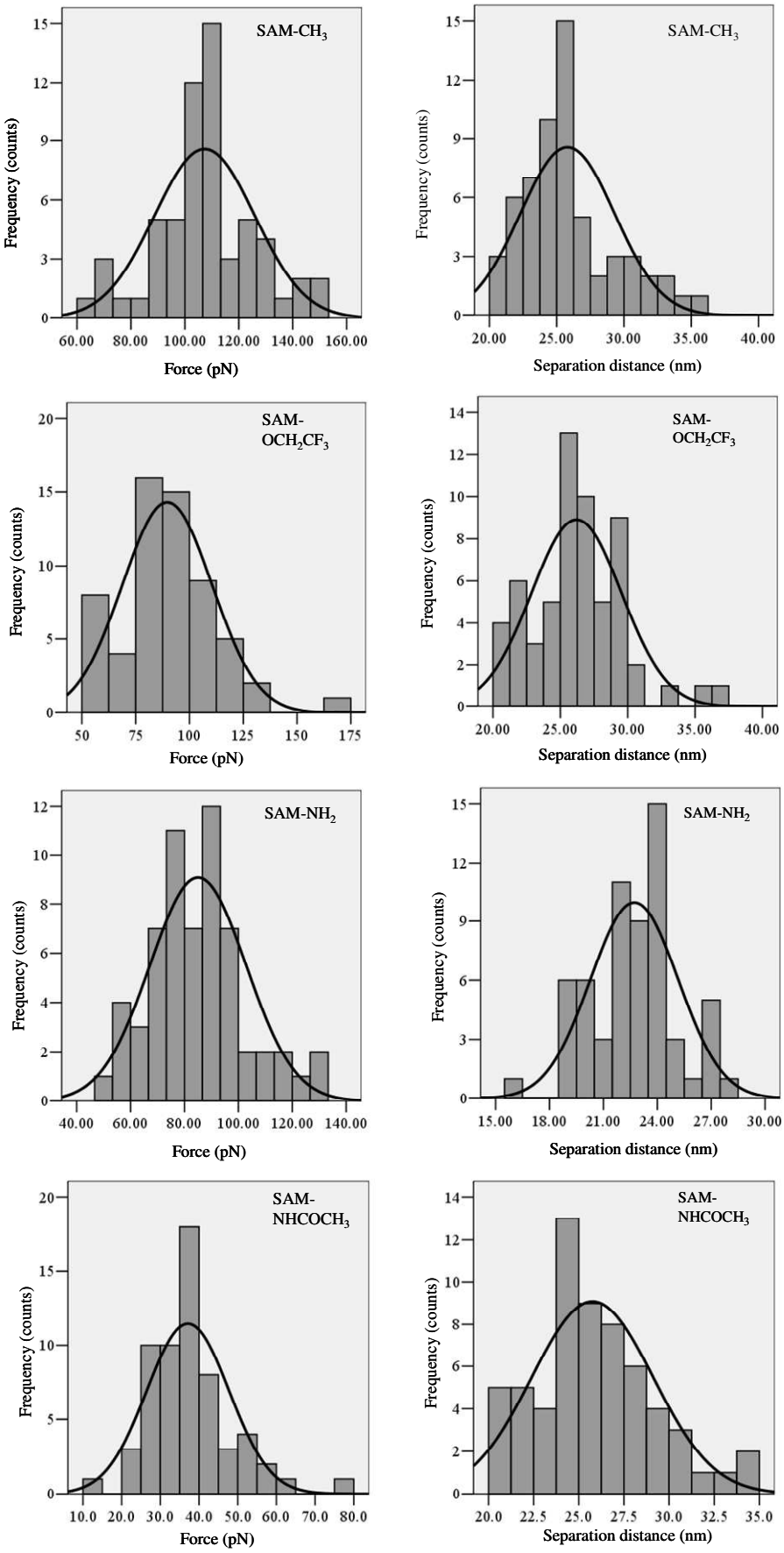

Figure 2. Distribution histogram of desorption force and separation distance from 60 measurements for TGTG–V–GTCT on selected SAM–surfaces in PBS.

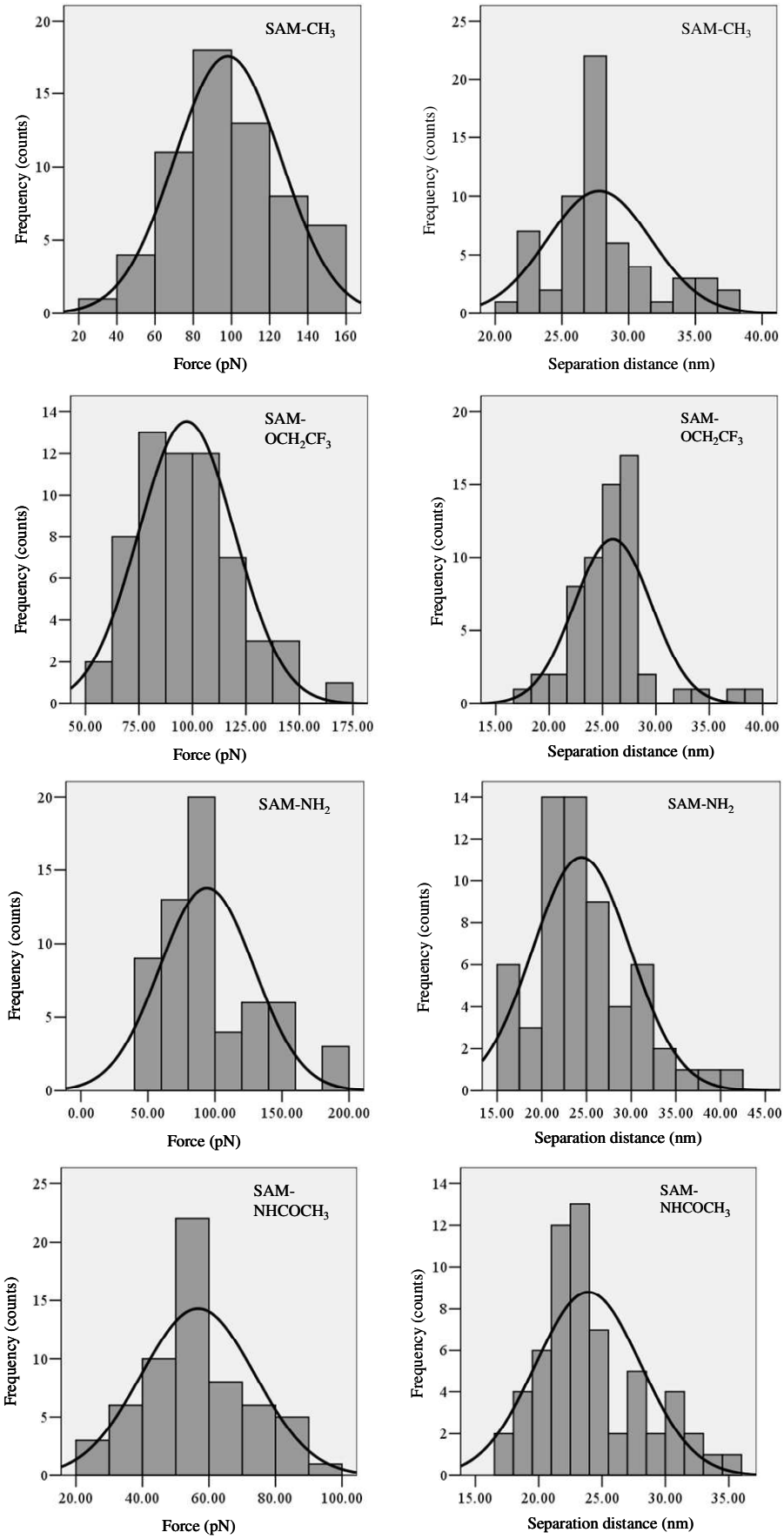

Figure 3. Distribution histogram of desorption force and separation distance from 60 measurements for TGTG–D–GTCT on selected SAM–surfaces in PBS.

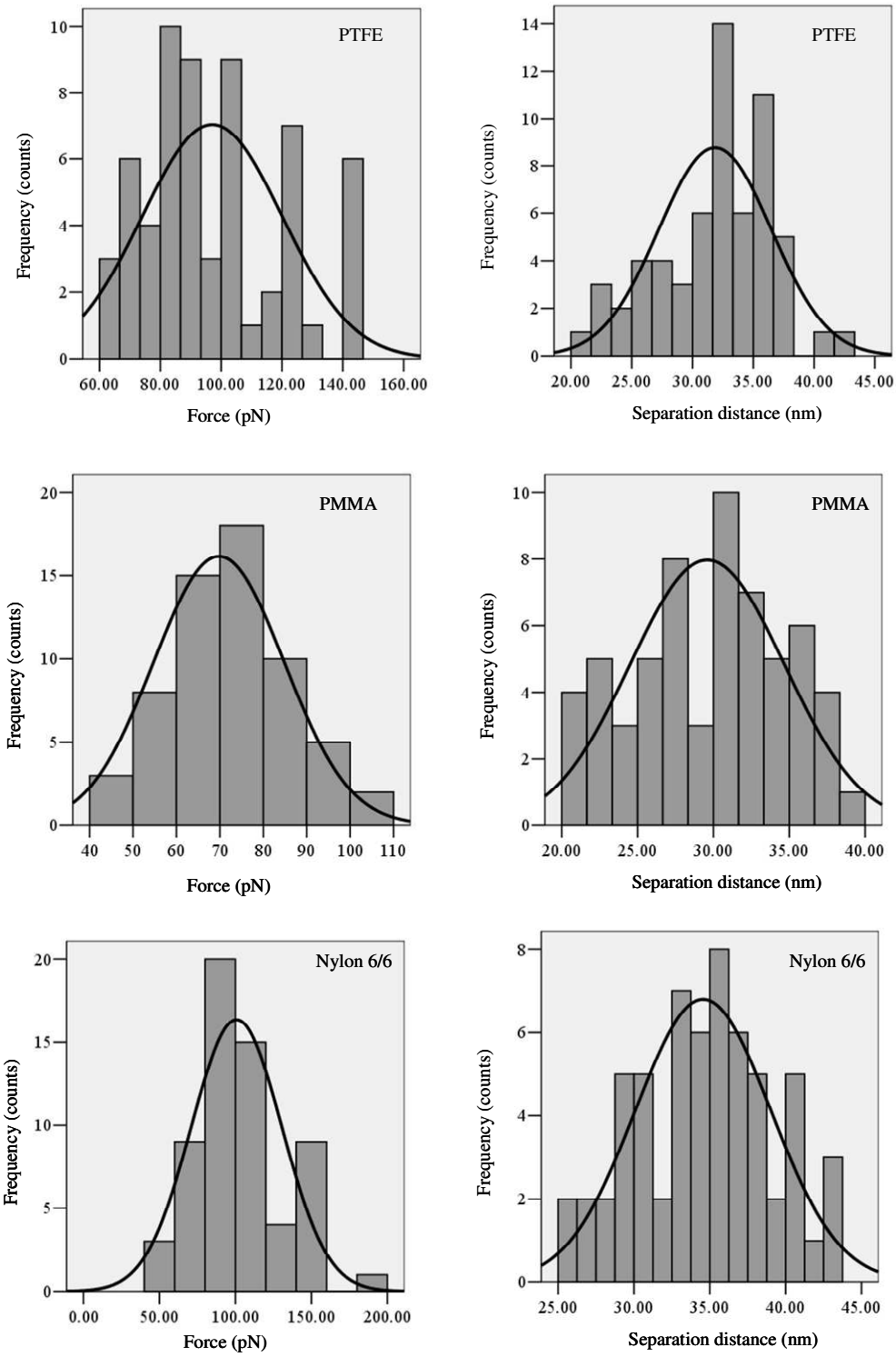

Figure 4. Distribution histogram of desorption force and separation distance from 60 measurements for TGTG–L–GTCT on selected polymer sheet in PBS.

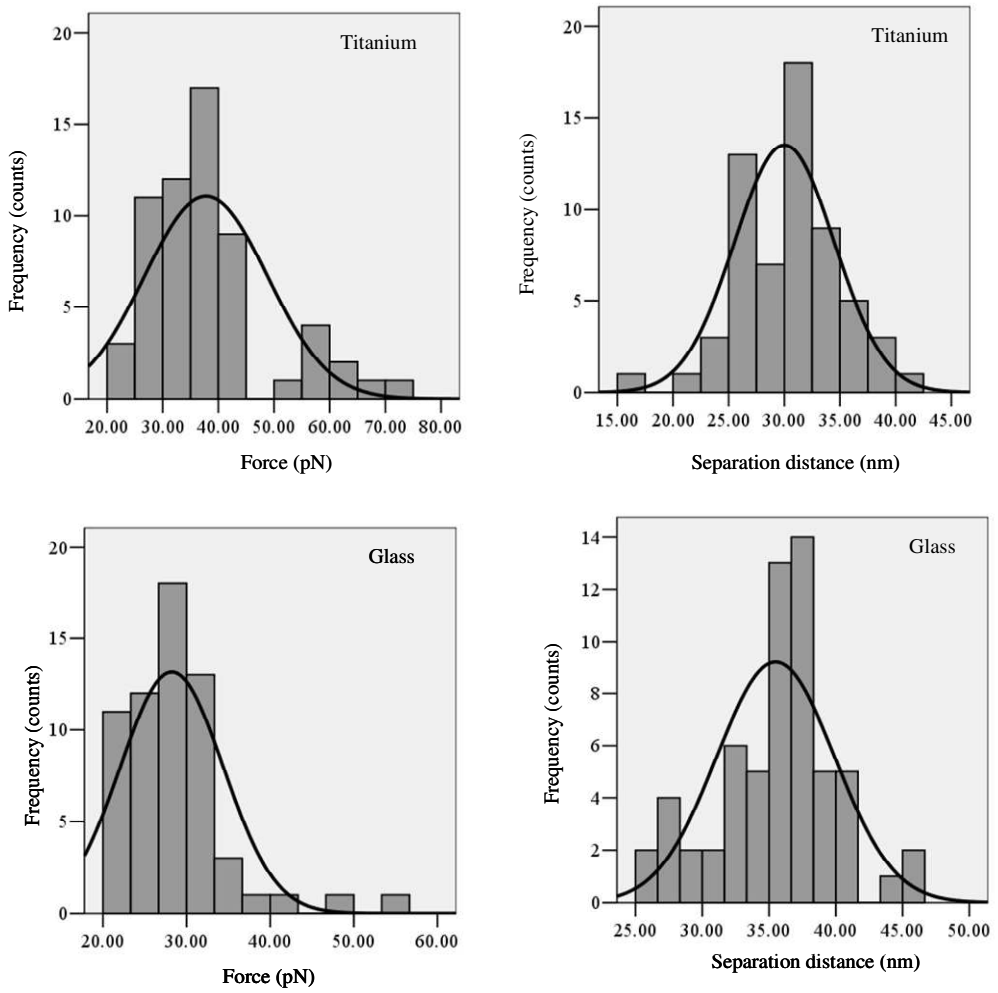

Figure 5. Distribution histogram of desorption force and separation distance from 60 measurements for TGTG–L–GTCT on titanium and glass surfaces in PBS.

REFERENCES:

1. Wei, Y.; Latour, R. A., Determination of the adsorption free energy for peptide–surface interactions by SPR spectroscopy. *Langmuir* **2008,** 24, (13), 6721–9.
2. Wei, Y.; Latour, R. A., Benchmark Experimental Data Set and Assessment of Adsorption Free Energy for Peptide−Surface Interactions. *Langmuir* **2009,** 25, (10), 5637–5646.