

Anti-colonization Effect of Au Surfaces with Self-Assembled Molecular Monolayers Functionalized with Antimicrobial Peptides on *S. Epidermidis*

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¹H NMR of *O,O'*-bis(tosyloxy)polyethylene glycols

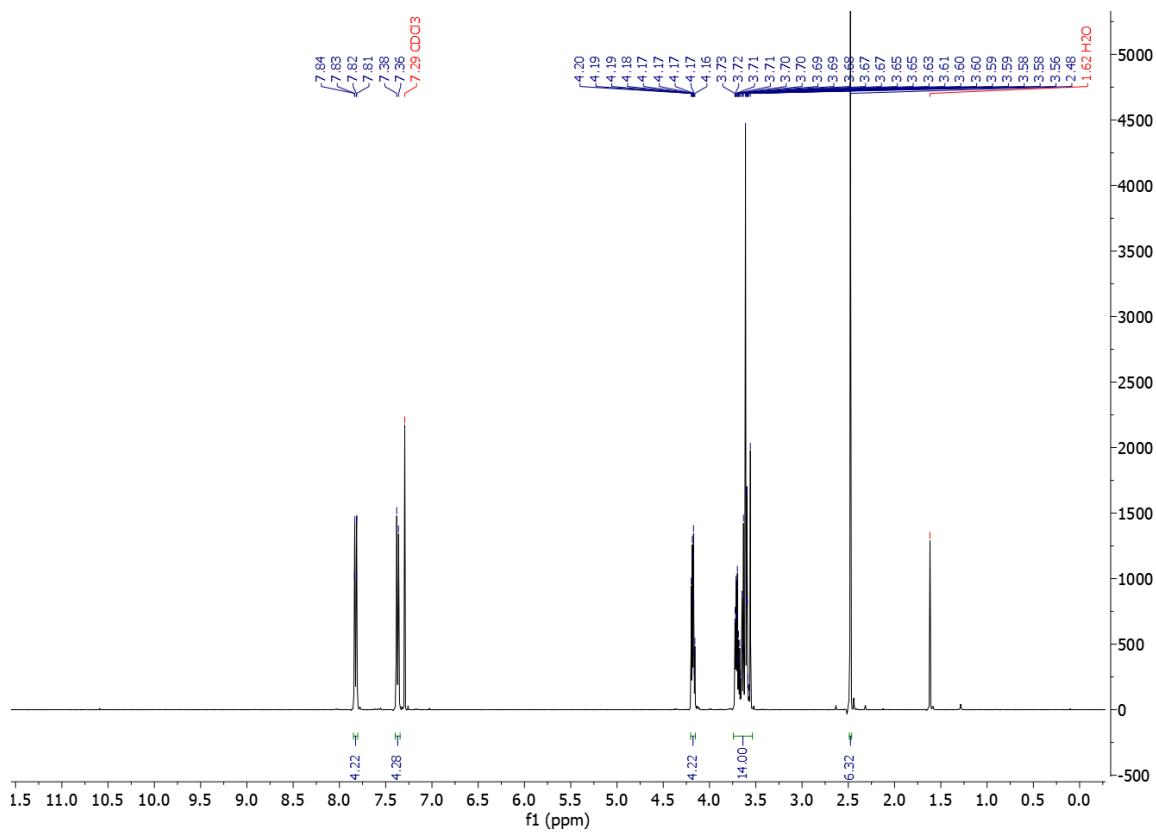


Figure S1. ¹H NMR of compound 3a.

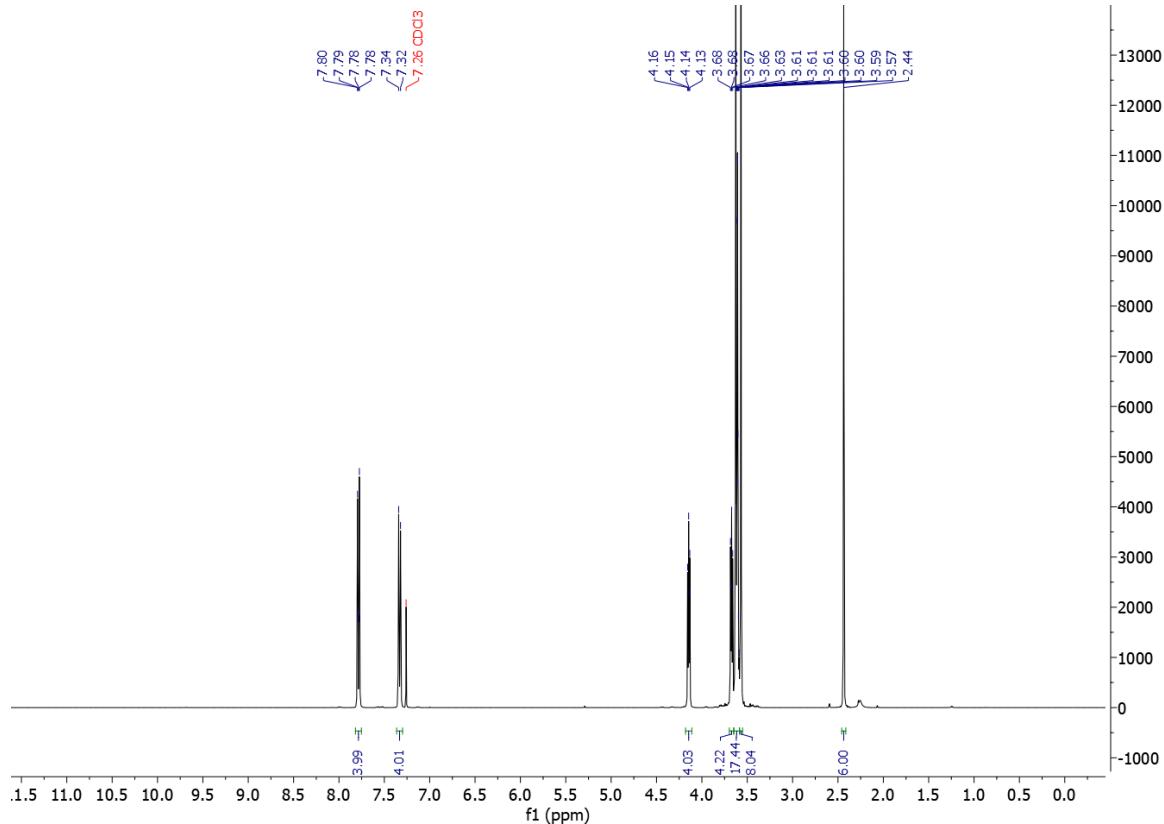


Figure S2. ¹H NMR of compound 3b.

¹H NMR of *O,O'*-bis(2-azidoethyl)polyethylene glycols

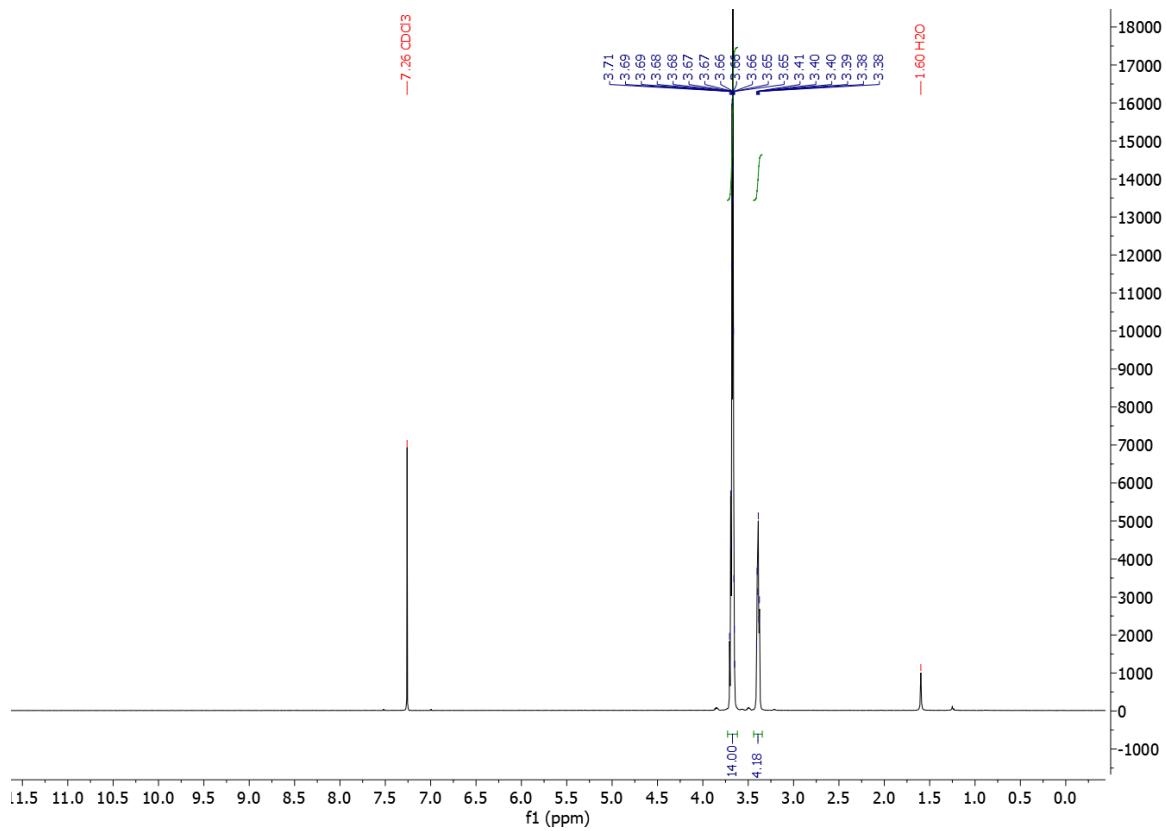


Figure S3. ^1H NMR of compound 4a.

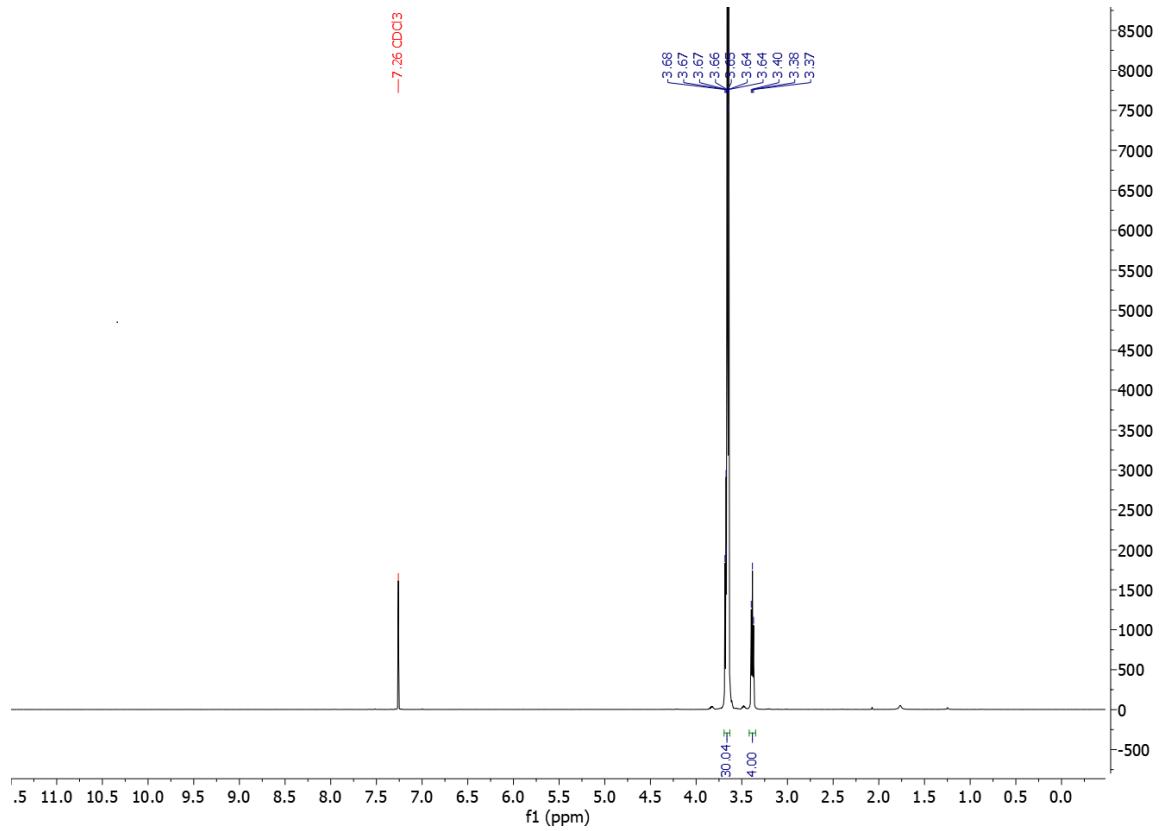


Figure S4. ^1H NMR of compound **4b**.

¹H NMR of O-(2-aminoethyl)-O'-(2-azidoethyl)polyethylene glycols

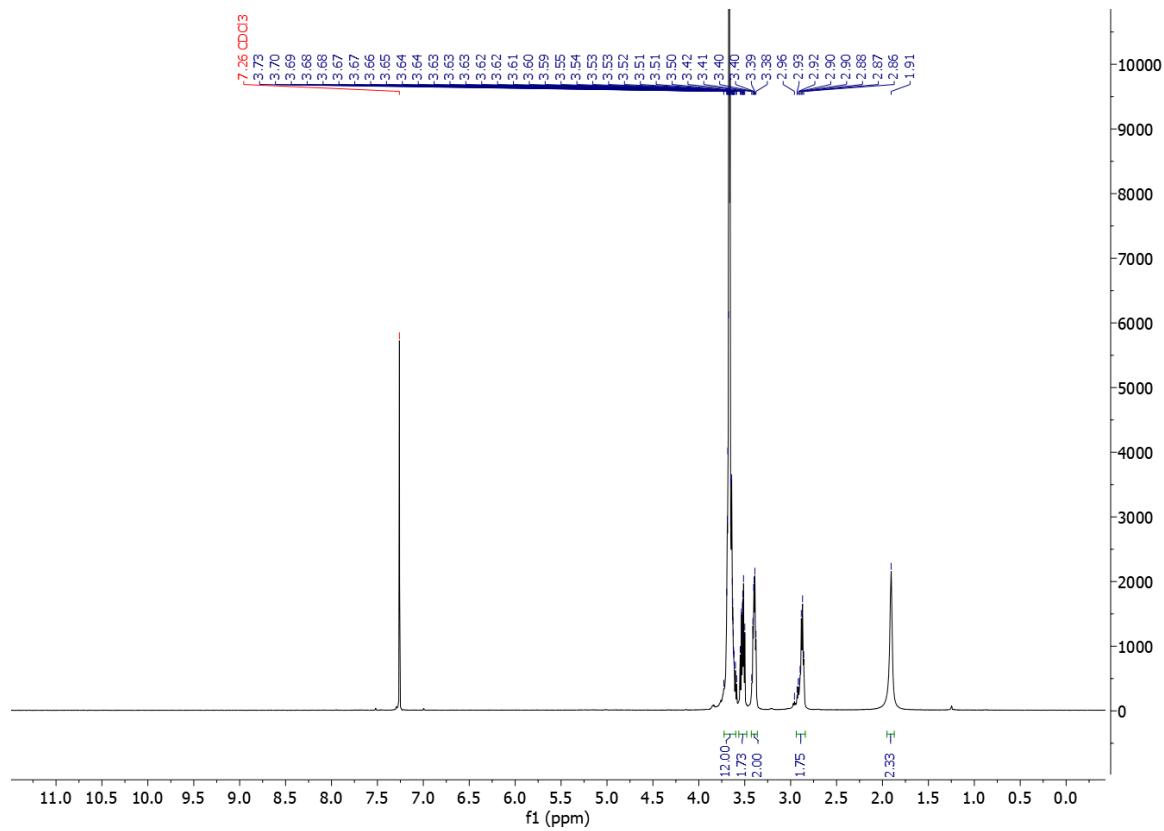


Figure S5. ^1H NMR of compound 5a.

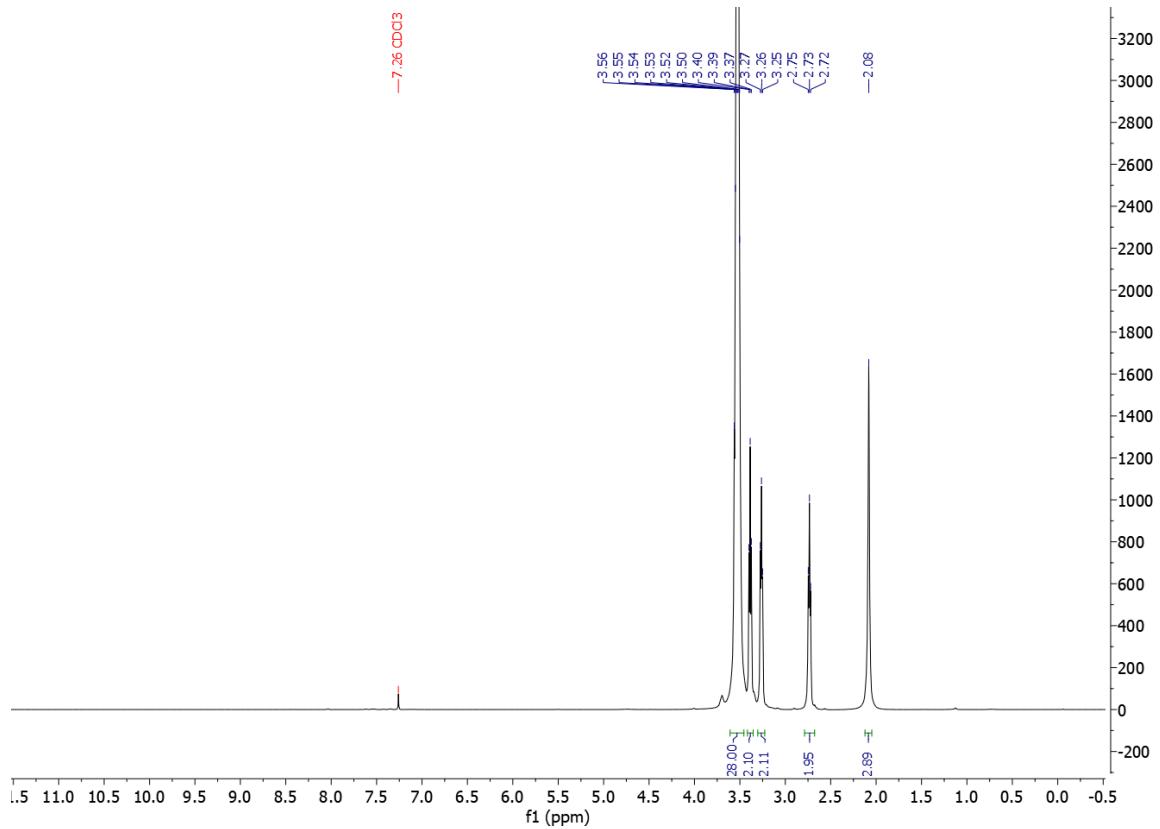


Figure S6. ^1H NMR of compound 5b.

¹H NMR of *O*-(2-azidoethyl)-*O*-[2-(diglycolyl-amino)ethyl]polyethylene glycols

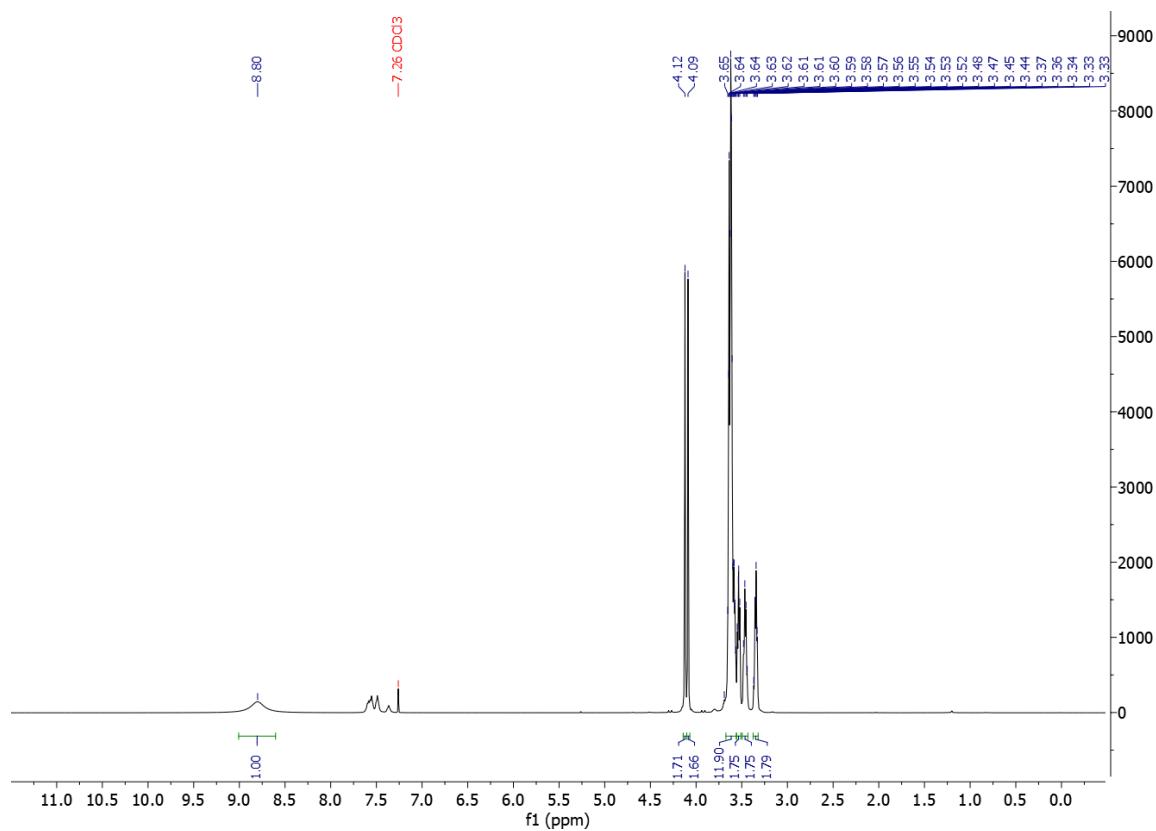


Figure S7. ¹H NMR of compound 6a.

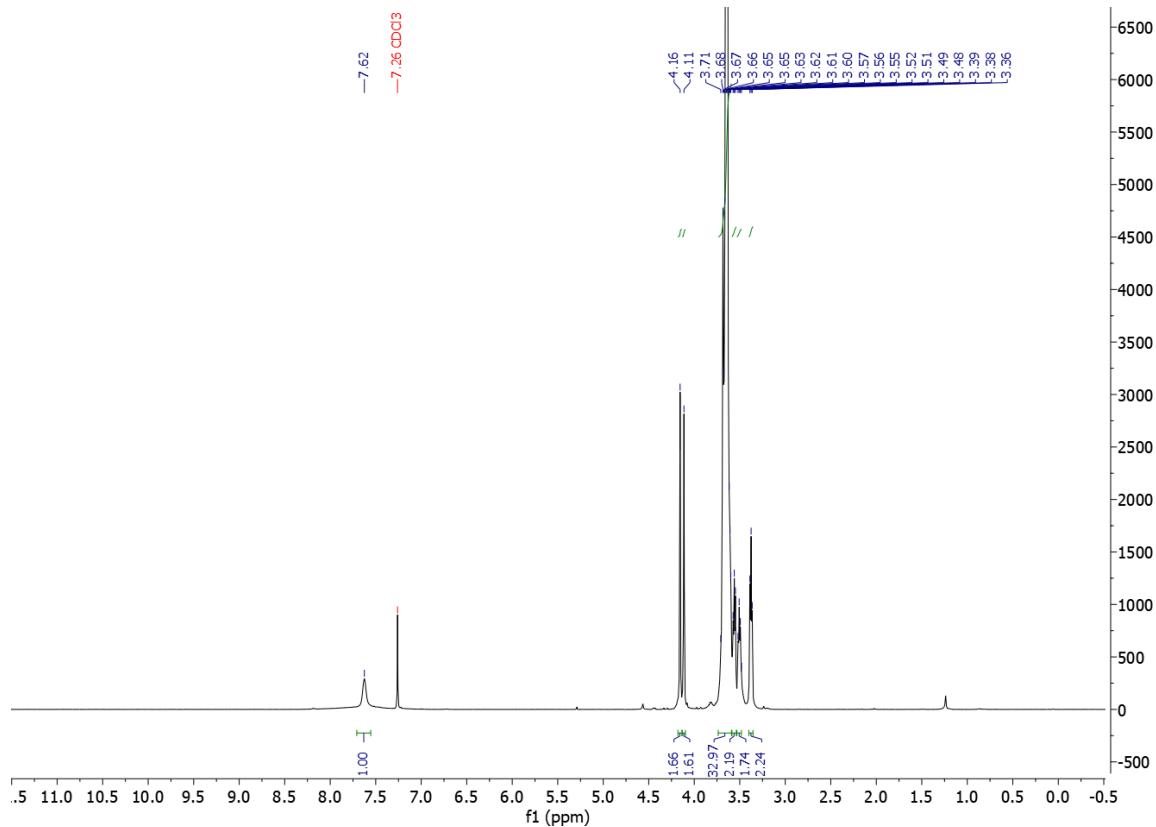


Figure S8. ¹H NMR of compound 6b.

¹H NMR of azidopeptides

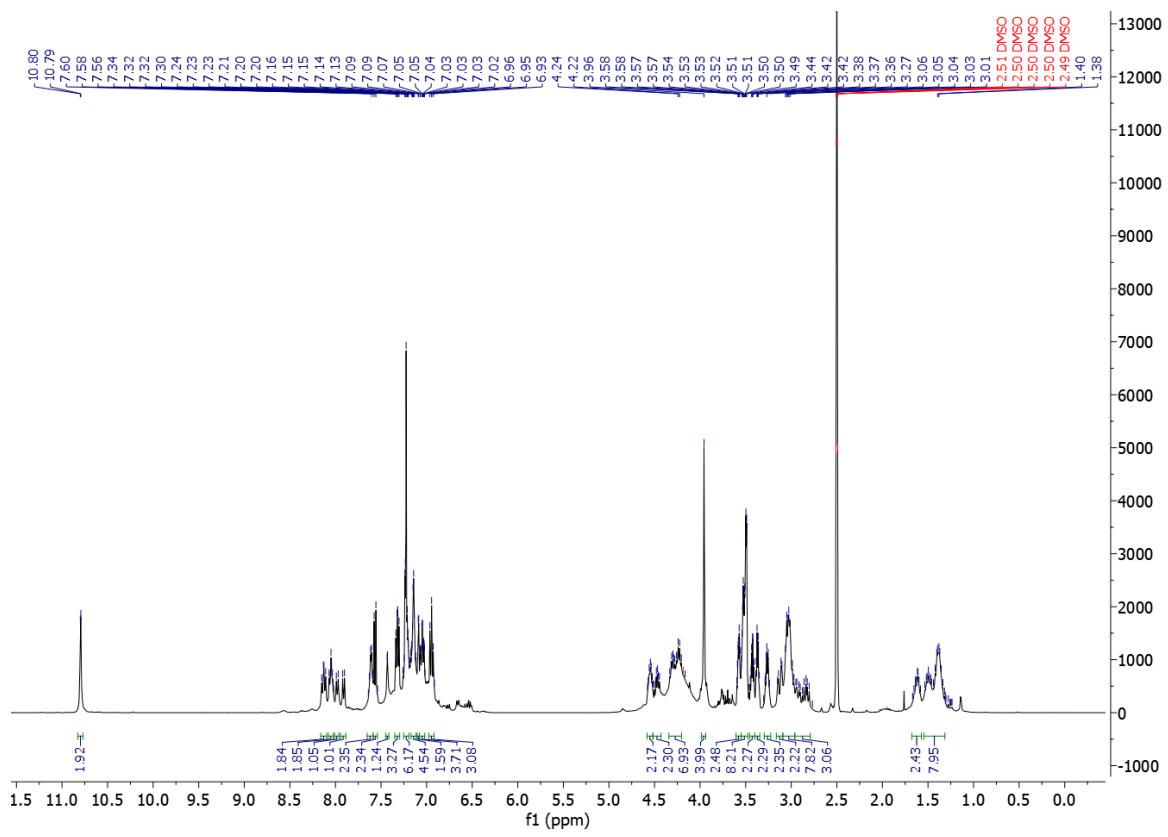


Figure S9. ^1H NMR of compound **1a**.

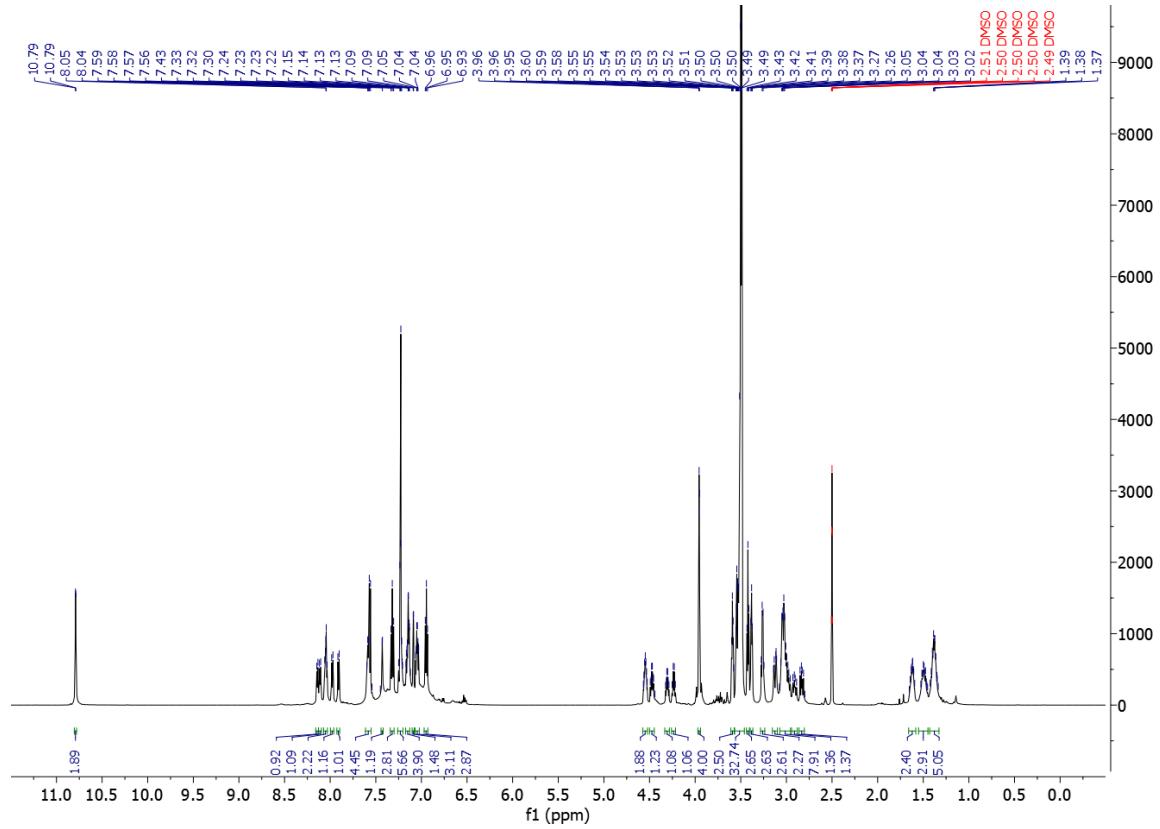


Figure S10. ^1H NMR of compound **1b**.

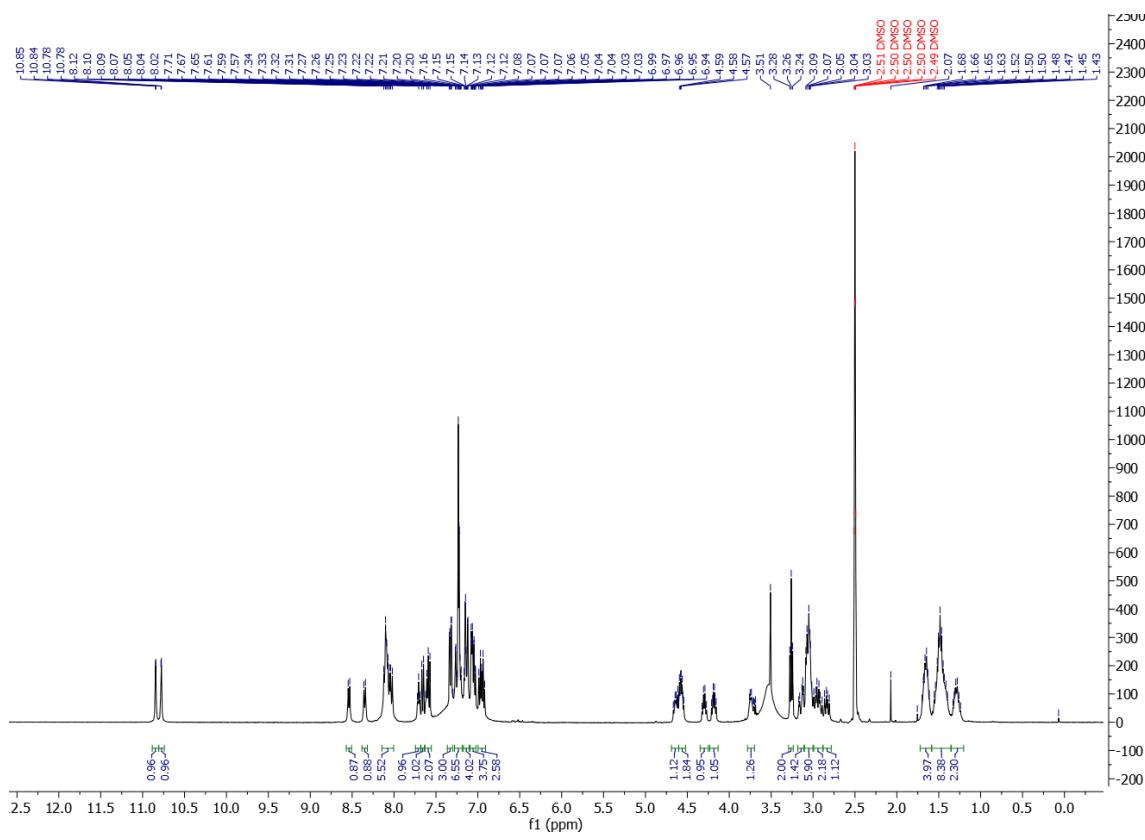


Figure S11. ^1H NMR of compound **1c**.

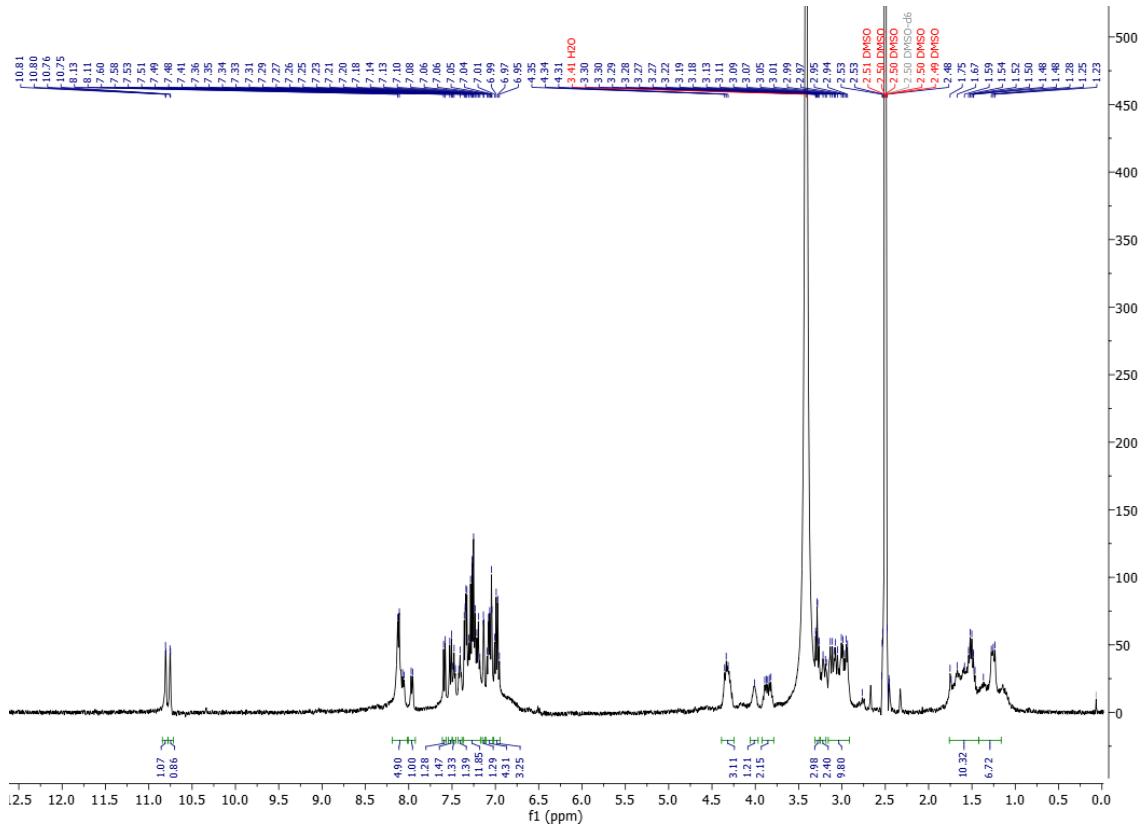


Figure S12. ^1H NMR of compound **1d**.

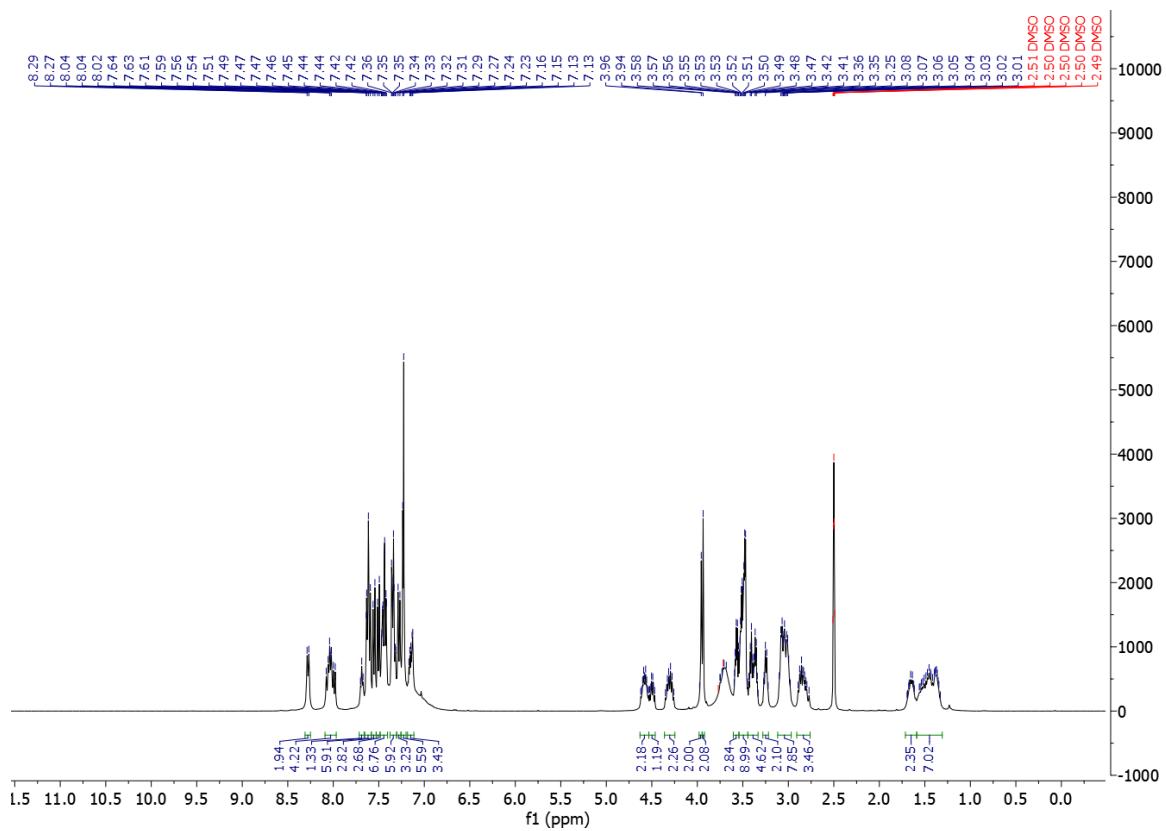


Figure S13. ^1H NMR of compound **2a**.

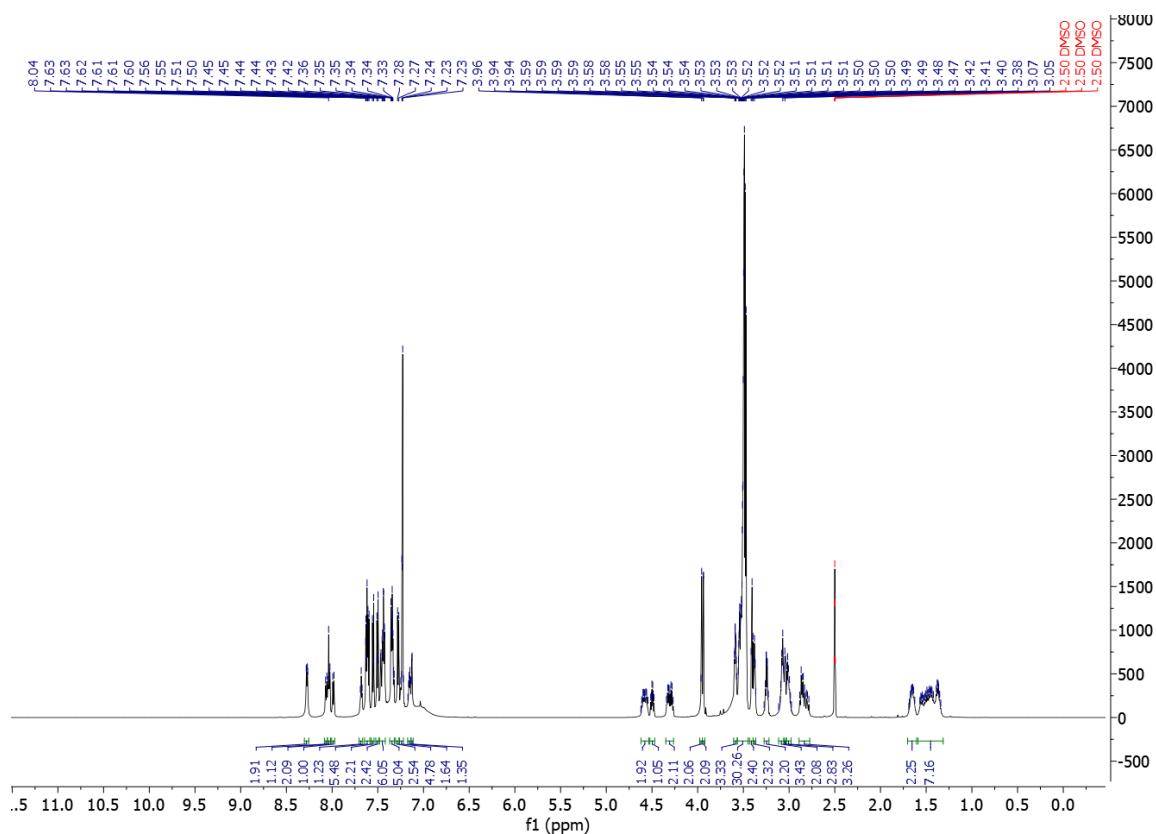


Figure S14. ^1H NMR of compound **2b**.

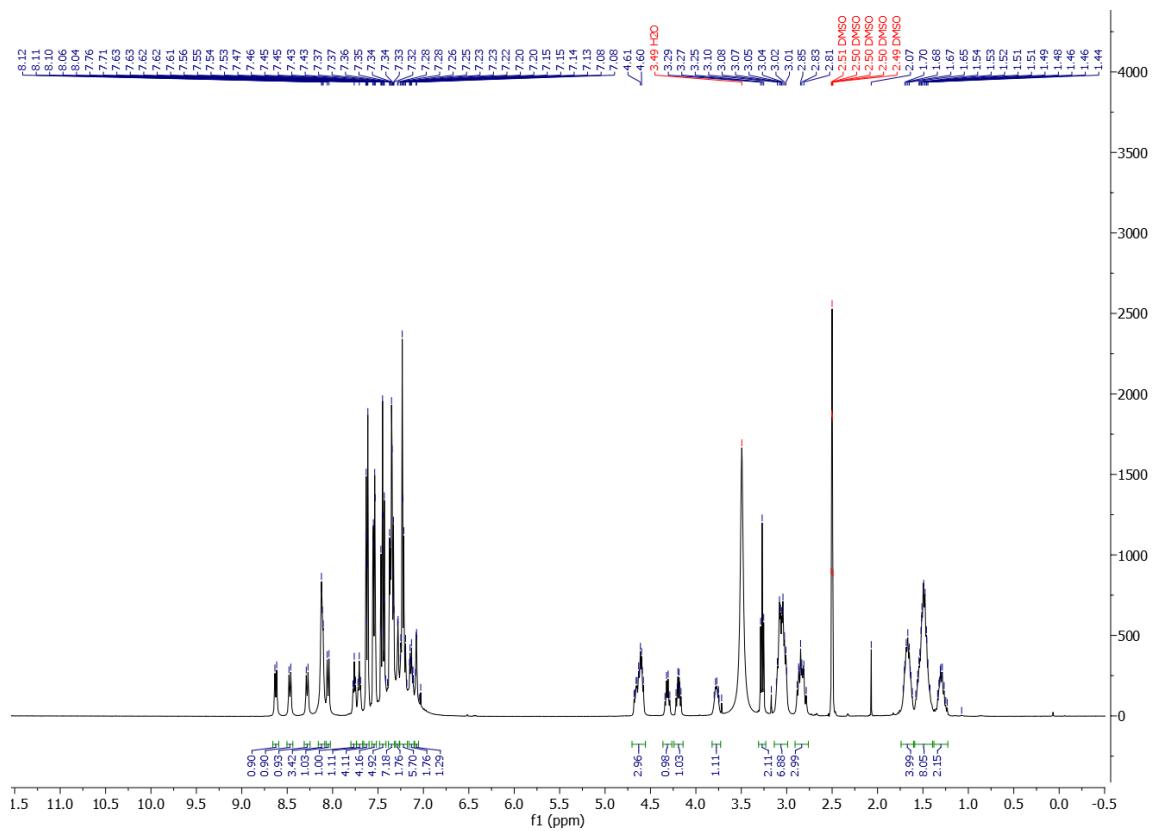


Figure S15. ^1H NMR of compound 2c.

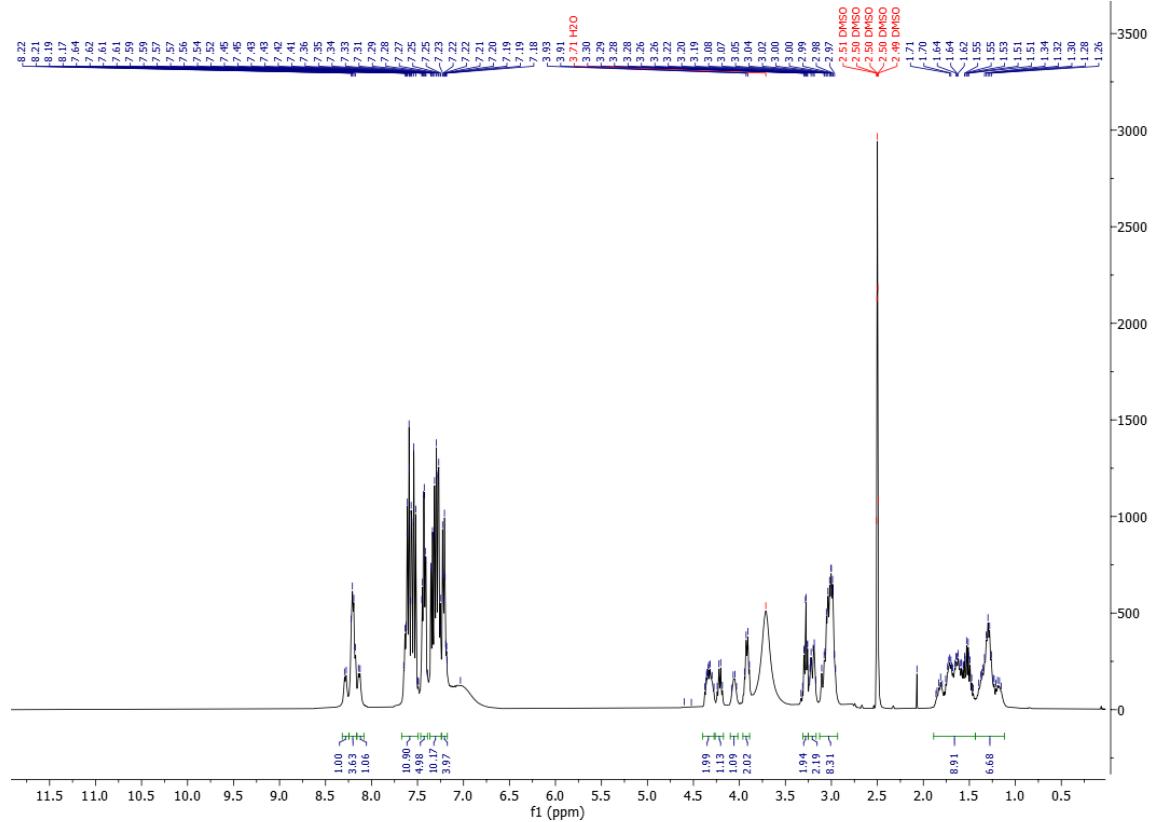


Figure S16. ^1H NMR of compound 2d.

Analytical HPLC of azidopeptides

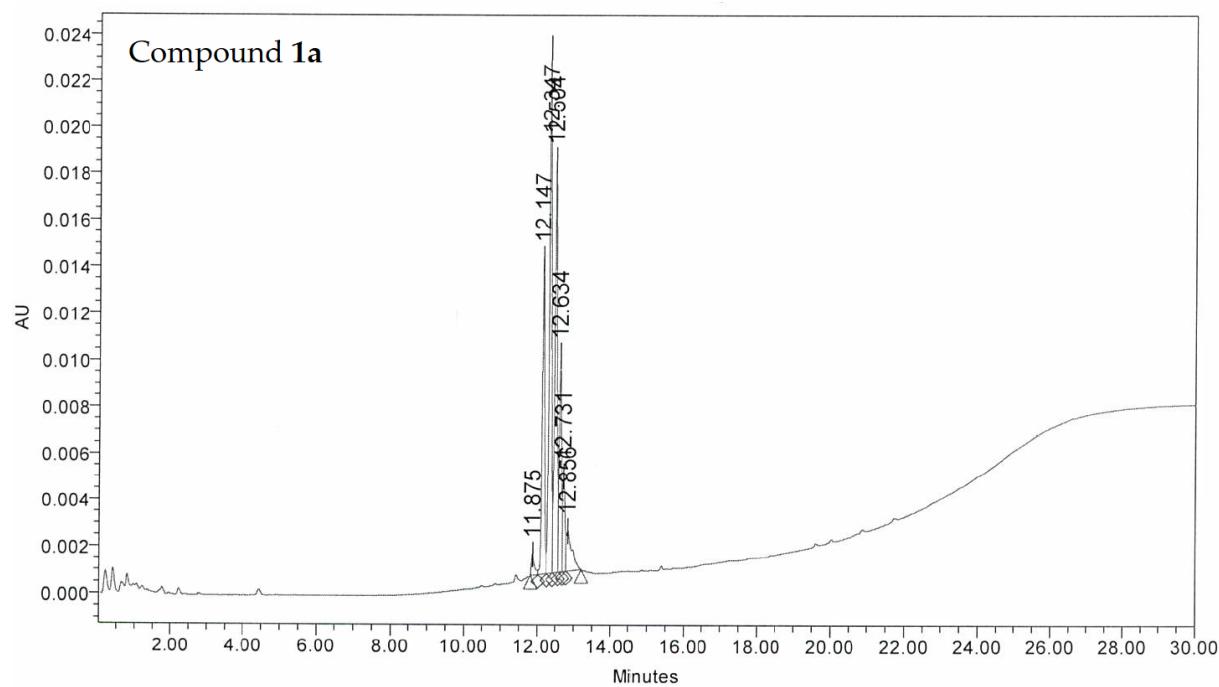


Figure S17. Analytical HPLC of compound 1a.

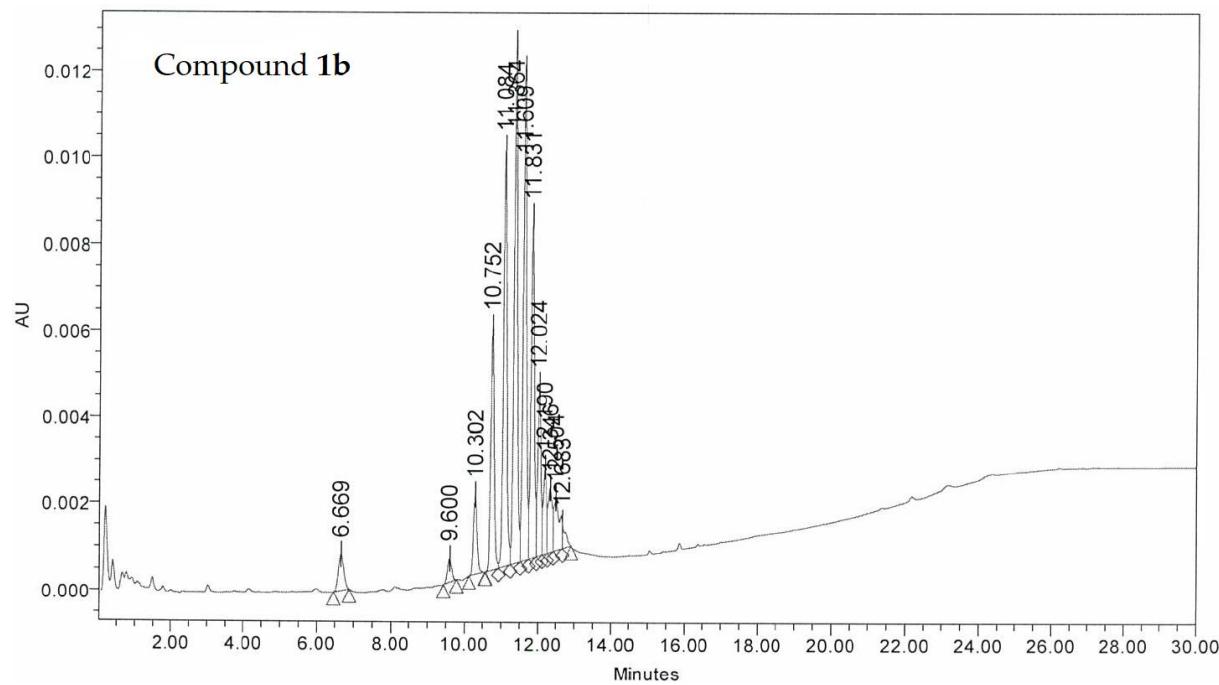


Figure S18. Analytical HPLC of compound 1b.

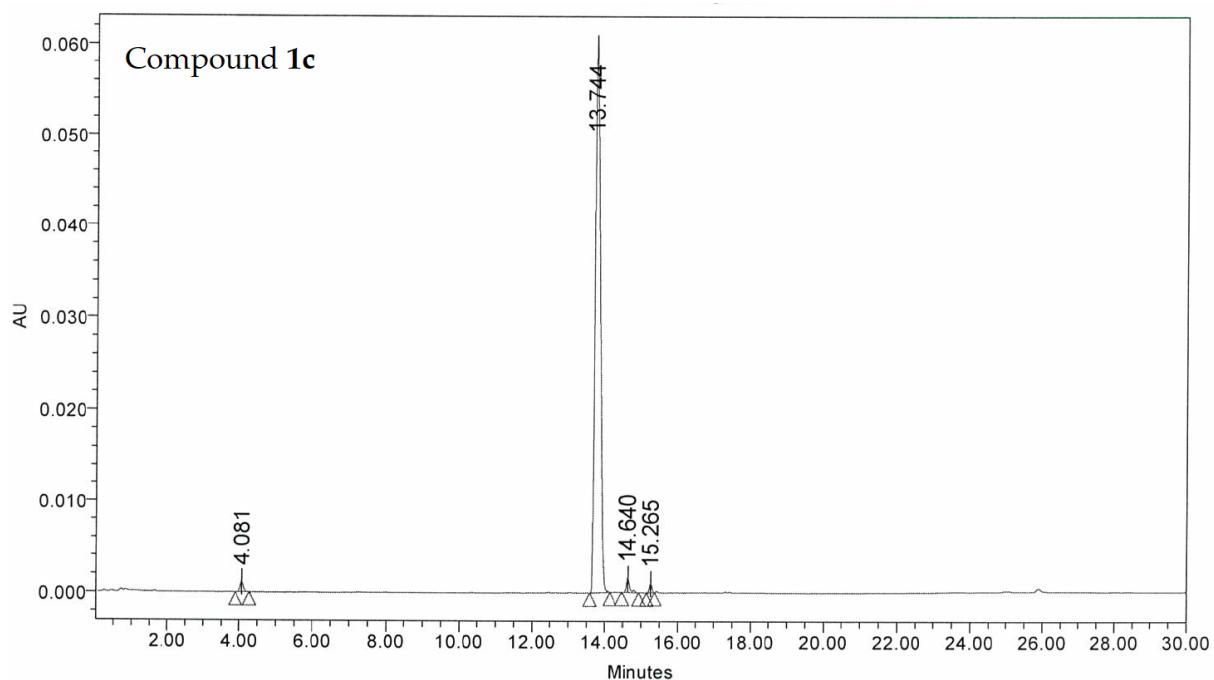


Figure S19. Analytical HPLC of compound 1c.

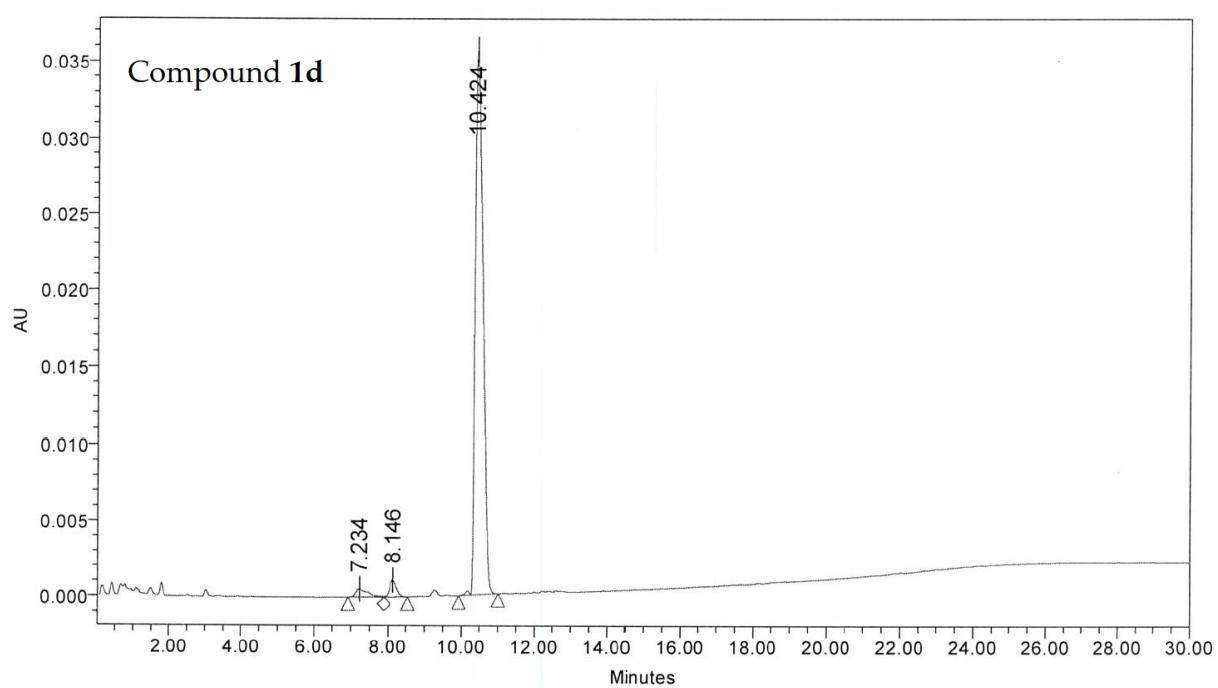


Figure S20. Analytical HPLC of compound 1d.

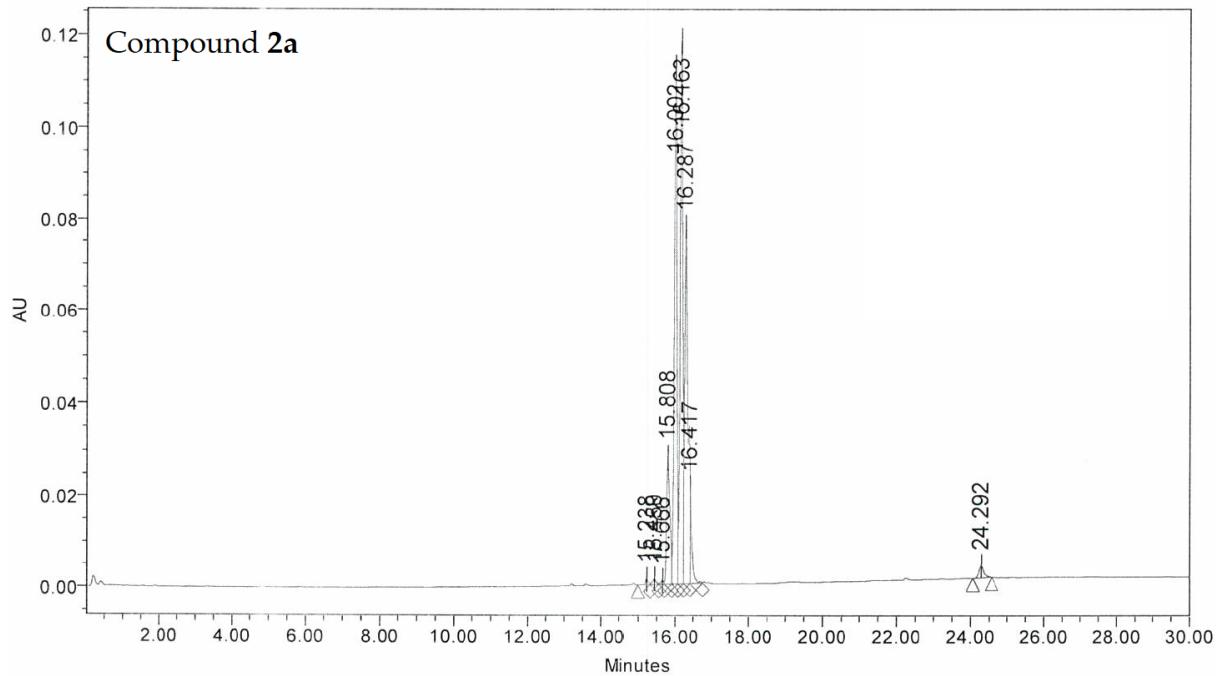


Figure S21. Analytical HPLC of compound 2a.

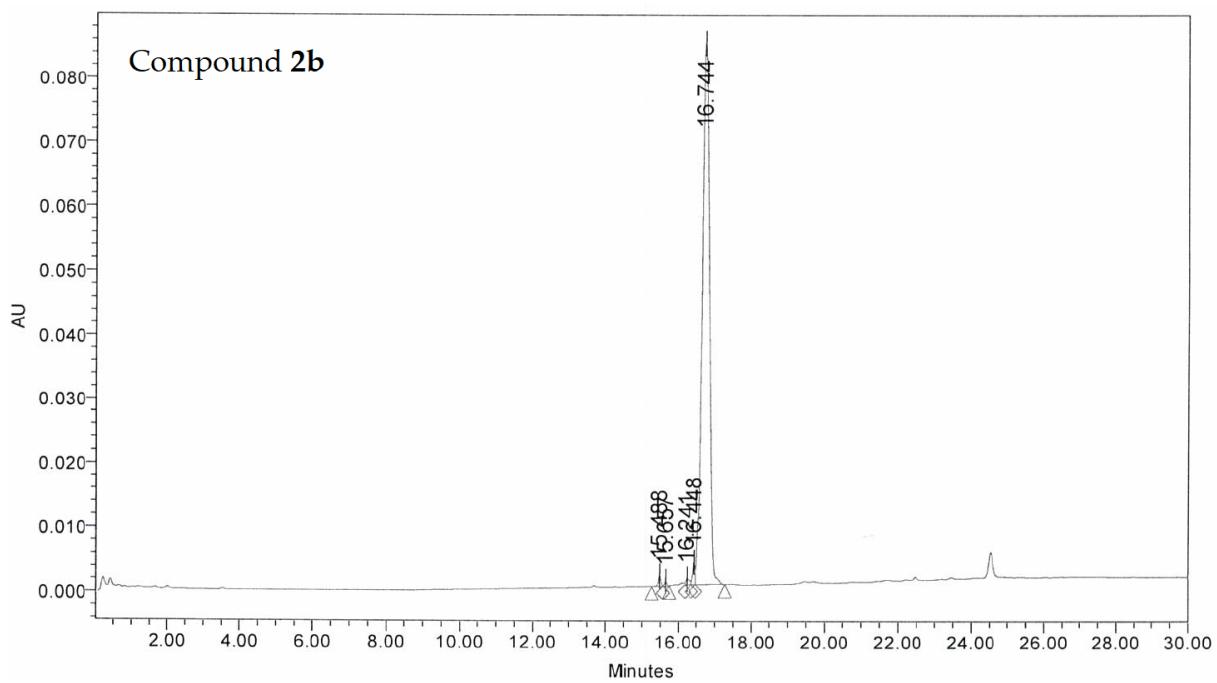


Figure S22. Analytical HPLC of compound 2b.

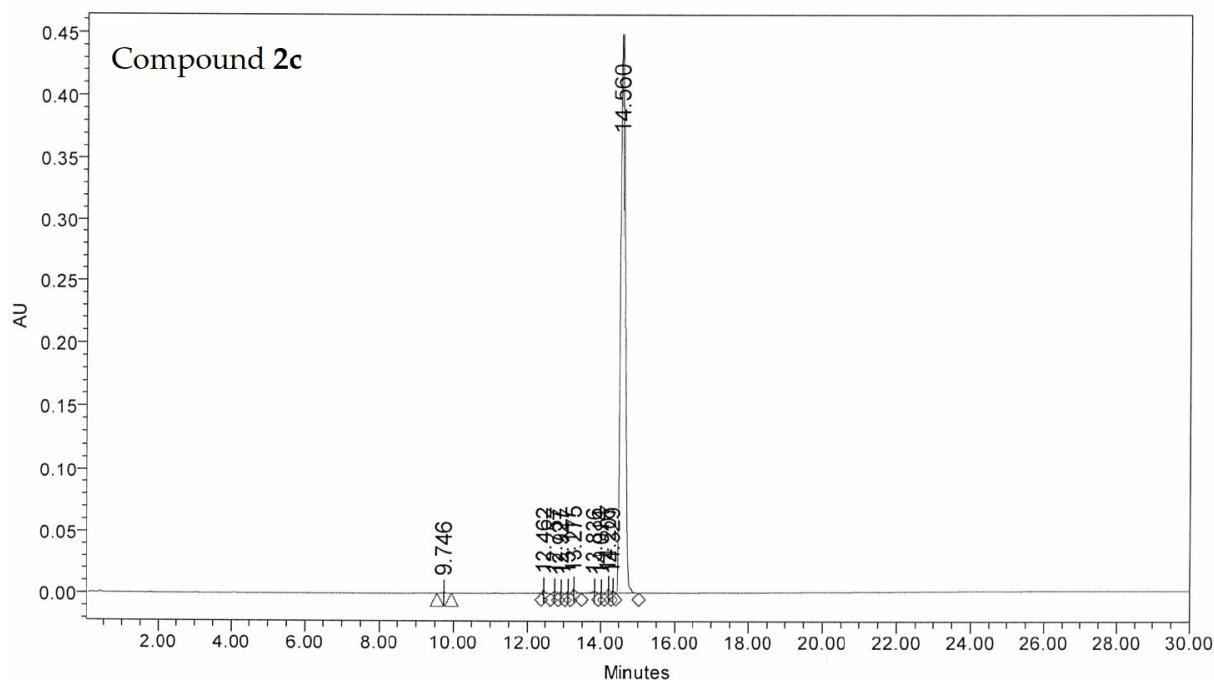


Figure S23. Analytical HPLC of compound 2c.

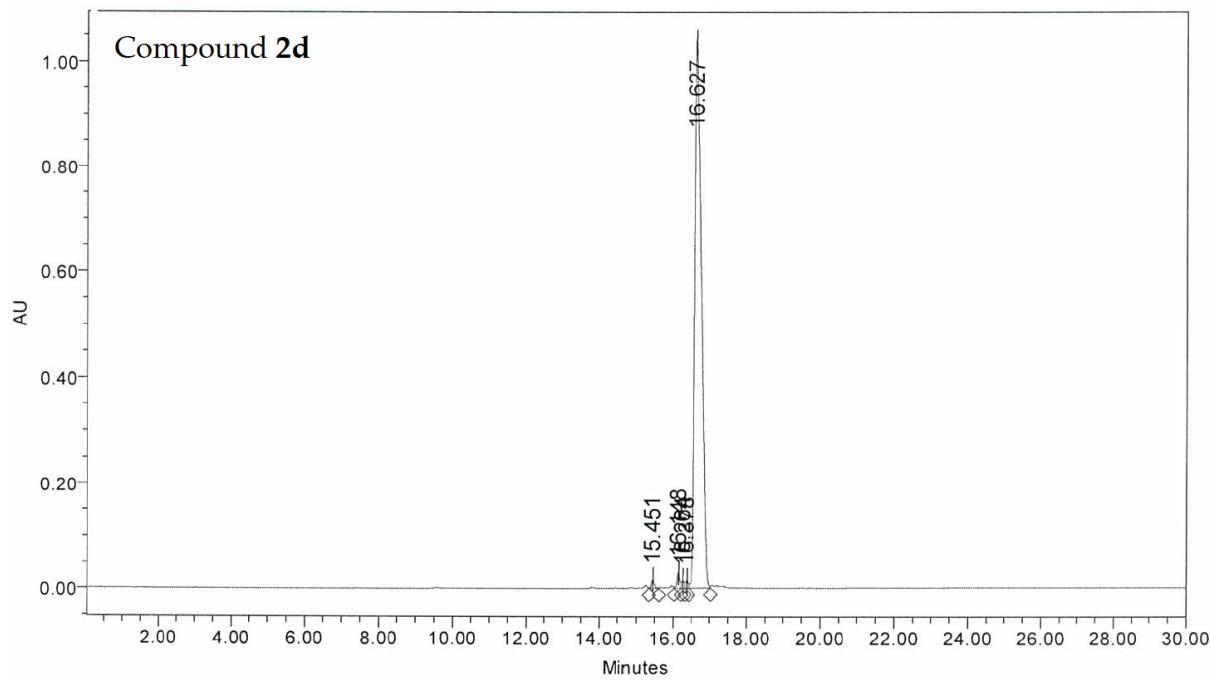


Figure S24. Analytical HPLC of compound 2d.

Contact angle data

Table S1. Contact Angle of **1a-d** and **2a-d** covalently linked to an Au surface. Control is Au surface treated with peptide, without the CuAAC reaction

No.	$\theta(M)$ [deg]	$\theta(L)$ [deg]	$\theta(R)$ [deg]	Fit-Er [um]	Method	Volum [ul]	BD [mm]
Au_1a_1	45.6 ± 0.06	45.5	45.6	0.92	T-1	0.211	1.349
Au_1a_2	52.1 ± 0.41	52.5	51.7	1.61	T-1	0.301	1.446
Au_1a_3	51.8 ± 0.66	51.1	52.4	2.28	T-1	0.249	1.368
Au_1a_4	48.2 ± 0.92	47.3	49.1	1.28	T-1	0.309	1.502
Au_1a_5	48.0 ± 1.10	46.9	49.1	1.06	T-1	0.260	1.422
Au_1b_1	56.2 ± 0.96	55.3	57.2	0.97	T-1	0.288	1.372
Au_1b_2	52.7 ± 0.39	53.1	52.3	1.16	T-1	0.269	1.372
Au_1b_3	50.6 ± 1.17	51.8	49.5	1.14	T-1	0.266	1.405
Au_1b_4	50.3 ± 0.86	49.4	51.1	0.88	T-1	0.277	1.420
Au_1b_5	51.0 ± 0.41	50.6	51.4	0.86	T-1	0.318	1.475
Au_1c_1	44.9 ± 0.60	44.3	45.5	1.10	T-1	0.280	1.514
Au_1c_2	50.5 ± 0.43	50.0	50.9	0.93	T-1	0.263	1.395
Au_1c_3	49.6 ± 0.29	49.9	49.3	1.76	T-1	0.286	1.491
Au_1c_4	54.2 ± 0.09	54.2	54.1	1.24	T-1	0.252	1.333
Au_1c_5	50.8 ± 0.23	50.6	51.0	1.15	T-1	0.264	1.383
Au_1d_1	46.5 ± 0.09	46.4	46.6	0.82	T-1	0.284	1.488
Au_1d_2	50.5 ± 0.28	50.8	50.3	1.39	T-1	0.315	1.438
Au_1d_3	49.1 ± 0.63	48.4	49.7	0.94	T-1	0.265	1.417
Au_1d_4	52.3 ± 1.26	51.1	53.6	1.54	T-1	0.168	1.184
Au_1d_5	49.6 ± 0.18	49.8	49.4	0.92	T-1	0.233	1.350
Au_2a_1	56.2 ± 0.22	56.0	56.4	0.98	T-1	0.317	1.411
Au_2a_2	50.6 ± 0.42	50.2	51.0	1.25	T-1	0.285	1.439
Au_2a_3	54.9 ± 0.92	55.9	54.0	1.10	T-1	0.304	1.413
Au_2a_4	54.7 ± 0.76	55.5	54.0	0.82	T-1	0.275	1.369
Au_2a_5	56.8 ± 1.09	57.8	55.7	0.91	T-1	0.295	1.379
Au_2b_1	52.7	52.6	52.7	1.42	T-1	0.282	1.405
Au_2b_2	53.9 ± 0.67	54.5	53.2	1.19	T-1	0.321	1.451
Au_2b_3	50.4 ± 0.95	49.5	51.4	1.18	T-1	0.285	1.429
Au_2b_4	53.7 ± 0.54	53.2	54.2	0.74	T-1	0.342	1.479
Au_2b_5	56.9 ± 0.65	56.3	57.6	0.65	T-1	0.275	1.343
Au_2c_1	57.2 ± 0.93	56.3	58.1	1.24	T-1	0.283	1.347
Au_2c_2	56.3 ± 0.59	55.7	56.9	1.08	T-1	0.335	1.442
Au_2c_3	52.6 ± 0.25	52.9	52.4	1.41	T-1	0.309	1.452
Au_2c_4	50.0 ± 0.26	50.3	49.7	1.09	T-1	0.294	1.452
Au_2c_5	58.4 ± 1.00	57.4	59.4	1.05	T-1	0.282	1.345
Au_2d_1	52.4 ± 0.71	51.7	53.2	1.15	T-1	0.251	1.347
Au_2d_2	50.8 ± 0.72	50.1	51.5	0.77	T-1	0.288	1.429
Au_2d_3	56.9 ± 0.11	56.8	57.0	0.64	T-1	0.267	1.322
Au_2d_4	56.4 ± 0.18	56.2	56.6	1.00	T-1	0.298	1.385
Au_2d_5	53.3 ± 0.25	53.1	53.6	0.67	T-1	0.288	1.402
Au_Control_1	36.1 ± 0.63	35.5	36.8	1.02	T-1	0.225	1.541
Au_Control_2	40.0 ± 1.75	38.3	41.8	0.76	T-1	0.250	1.510
Au_Control_3	41.4 ± 1.20	40.2	42.6	1.04	T-1	0.234	1.473
Au_Control_4	37.7 ± 1.03	38.7	36.7	1.46	T-1	0.303	1.643
Au_Control_5	42.8 ± 0.27	42.5	43.0	0.92	T-1	0.208	1.372

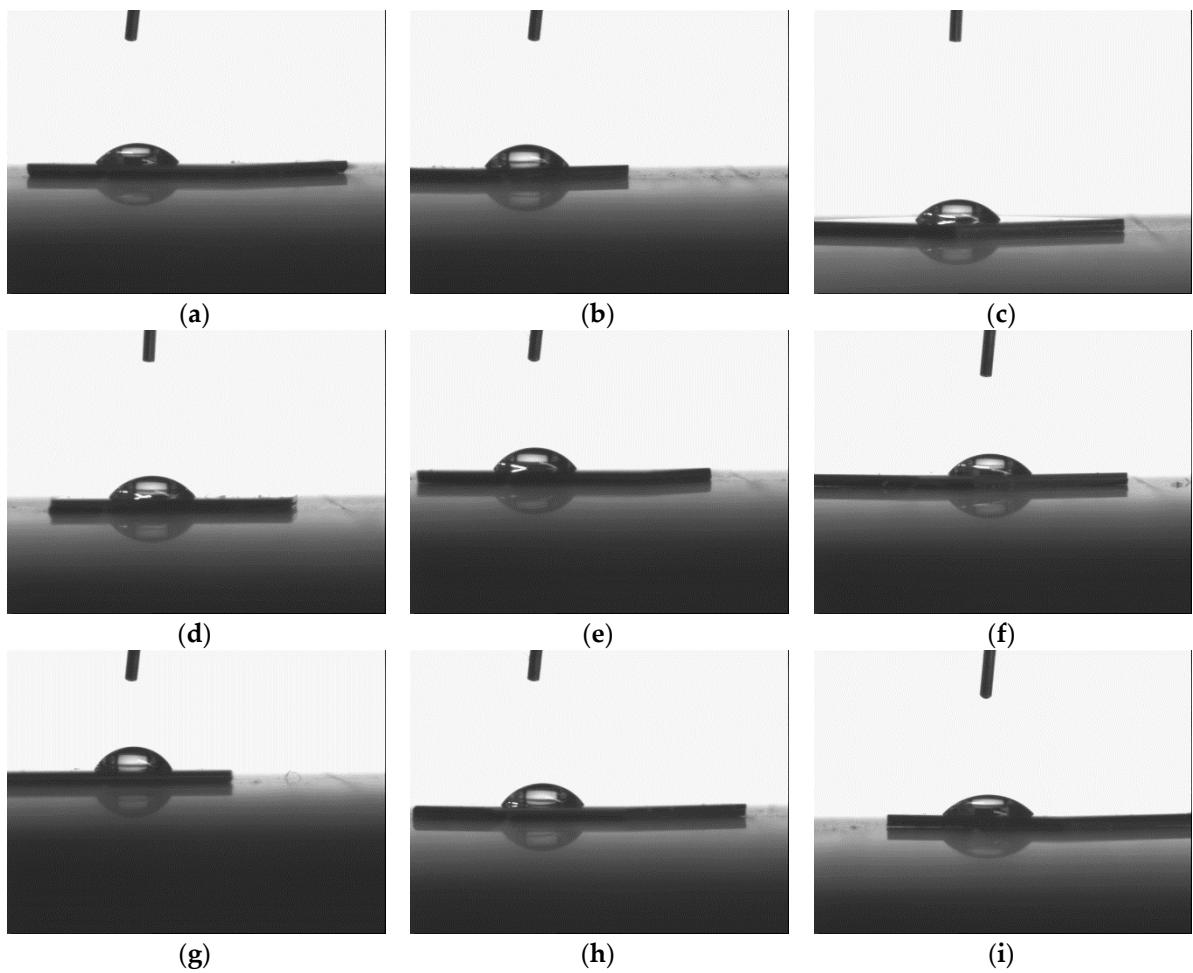


Figure S25. Contact angle images of **1a–d** and **2a–d** covalently linked to an Au surface. Control is Au surface treated with peptide, without the CuAAC reaction: (a) **1a** on Au surface; (b) **1b** on Au surface; (c) **1c** on Au surface; (d) **1d** on Au surface; (e) **2a** on Au surface; (f) **2b** on Au surface; (g) **2c** on Au surface; (h) **2d** on Au surface; (i) Au surface as a control group.

ToF-SIMS of series 1 on Au surfaces

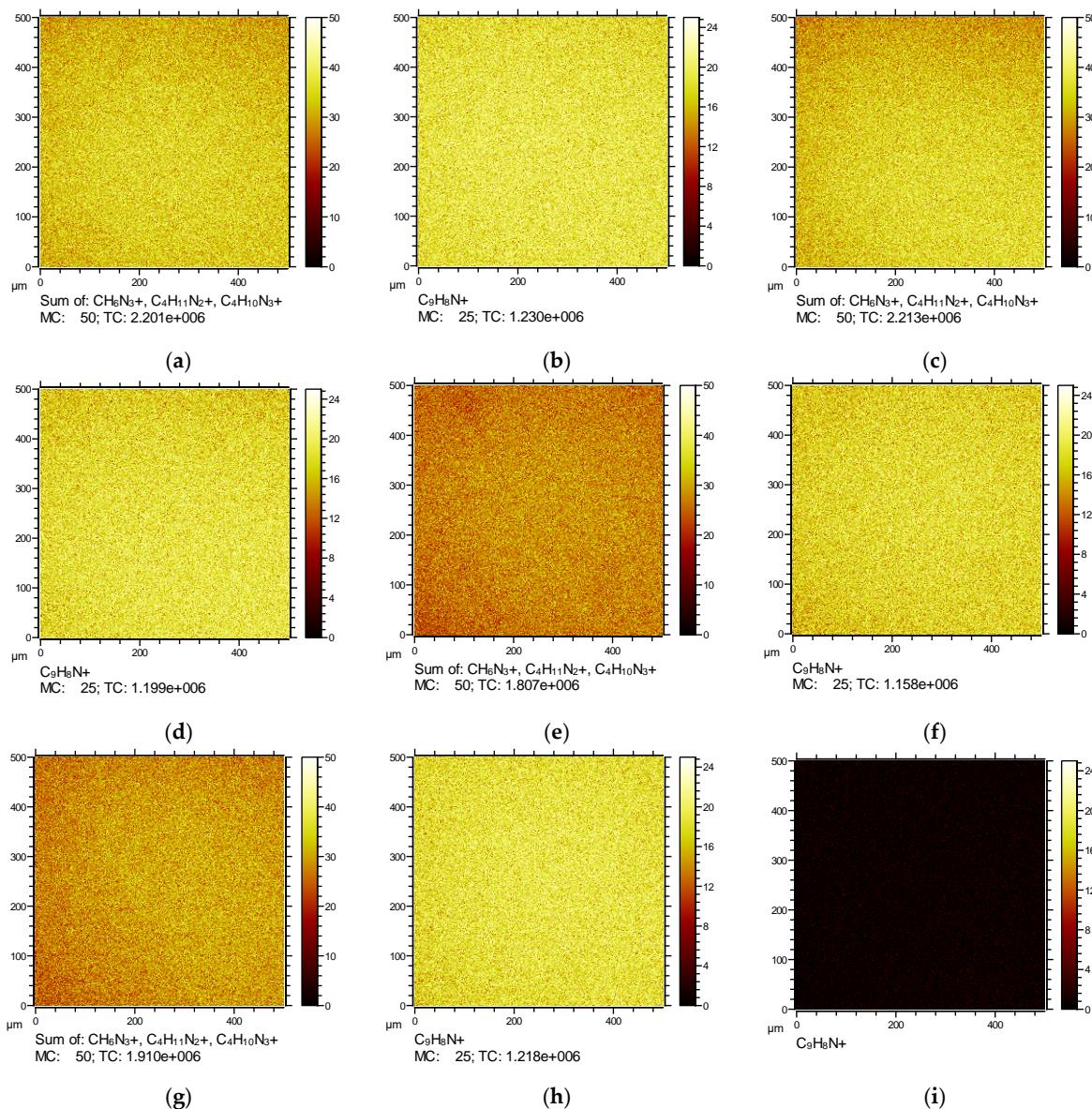


Figure S26. ToF-SIMS images of series 1 containing tryptophan: (a) **1a** surface of arginine residues with ion intensities for CH_6N_3^+ , $\text{C}_4\text{H}_{11}\text{N}_2^+$, and $\text{C}_4\text{H}_{10}\text{N}_3^+$; (b) **1a** surface of tryptophan residues with ion intensity for $\text{C}_9\text{H}_8\text{N}^+$; (c) **1b** surface of arginine residues with ion intensities for CH_6N_3^+ , $\text{C}_4\text{H}_{11}\text{N}_2^+$, and $\text{C}_4\text{H}_{10}\text{N}_3^+$; (d) **1b** surface of tryptophan residues with ion intensity for $\text{C}_9\text{H}_8\text{N}^+$; (e) **1c** surface of arginine residues with ion intensities for CH_6N_3^+ , $\text{C}_4\text{H}_{11}\text{N}_2^+$, and $\text{C}_4\text{H}_{10}\text{N}_3^+$; (f) **1c** surface with tryptophan residues with ion intensity for $\text{C}_9\text{H}_8\text{N}^+$; (g) **1d** surface of arginine residues with ion intensities for CH_6N_3^+ , $\text{C}_4\text{H}_{11}\text{N}_2^+$, and $\text{C}_4\text{H}_{10}\text{N}_3^+$; (h) **1d** surface of tryptophan residues with ion intensity for $\text{C}_9\text{H}_8\text{N}^+$; (i) image of the control Au surface observed at tryptophan-specific ion $\text{C}_9\text{H}_8\text{N}^+$.

ToF-SIMS of series 2 on Au surfaces

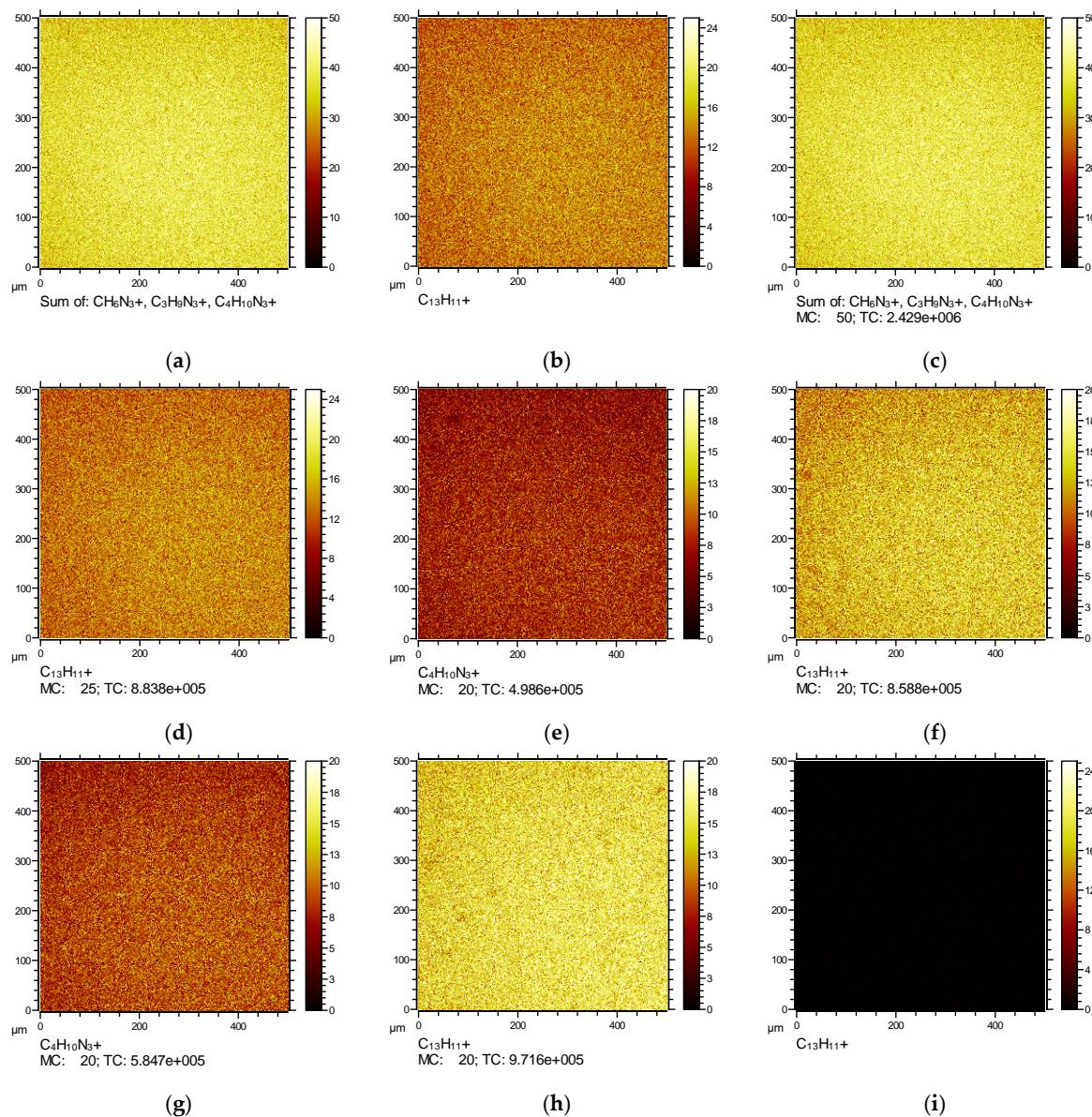


Figure S27. ToF-SIMS images of series 2 containing biphenylalanine: (a) **2a** surface of arginine residues with ion intensities for CH_6N_3^+ , $\text{C}_3\text{H}_9\text{N}_3^+$, and $\text{C}_4\text{H}_{10}\text{N}_3^+$; (b) **2a** surface of biphenylalanine residues with ion intensity for $\text{C}_{13}\text{H}_{11}^+$; (c) **2b** surface of arginine residues with ion intensities for CH_6N_3^+ , $\text{C}_3\text{H}_9\text{N}_3^+$, and $\text{C}_4\text{H}_{10}\text{N}_3^+$; (d) **2b** surface of biphenylalanine residues with ion intensity for $\text{C}_{13}\text{H}_{11}^+$; (e) **2c** surface of arginine residues with ion intensity for $\text{C}_4\text{H}_{10}\text{N}_3^+$; (f) **2c** surface of biphenylalanine residues with ion intensity for $\text{C}_{13}\text{H}_{11}^+$; (g) **2d** surface of arginine residues with ion intensity for $\text{C}_4\text{H}_{10}\text{N}_3^+$; (h) **2d** surface of biphenylalanine residues with ion intensity for $\text{C}_{13}\text{H}_{11}^+$; (i) image of the control Au surface observed at biphenylalanine-specific ion $\text{C}_{13}\text{H}_{11}^+$.

Certika data

Table S2. Certika data of **1a-d** and **2a-d** covalently linked to Au surface. Au surface without a peptide was used as a control group.

Sample	Replicate #1	Replicate #2	Average surface	Average per surface
Au_1a_1	7.4	7.6	7.5	
Au_1a_2	6.4	6.6	6.5	
Au_1a_3	7.3	7.3	7.3	
Au_1a_4	7.3	7.3	7.3	
Au_1a_5	7.2	7.0	7.1	7.13
Au_1b_1	7.9	7.9	7.9	
Au_1b_2	8.1	8.2	8.1	
Au_1b_3	7.4	7.5	7.5	
Au_1b_4	7.1	6.9	7.0	
Au_1b_5	6.8	6.9	6.9	
Au_1b_6	7.5	7.4	7.4	7.47
Au_1c_1	8.9	9.0	8.9	
Au_1c_2	6.9	7.1	7.0	
Au_1c_3	6.5	6.5	6.5	
Au_1c_4	6.4	6.3	6.4	
Au_1c_5	6.4	6.5	6.4	7.03
Au_1d_1	13.7	17.2	15.4	
Au_1d_2	6.3	6.4	6.4	
Au_1d_3	10.1	10.3	10.2	
Au_1d_4	8.7	8.5	8.6	
Au_1d_5	10.5	10.3	10.4	10.19
Au_2a_1	8.7	8.4	8.6	
Au_2a_2	7.9	8.1	8.0	
Au_2a_3	8.9	8.7	8.8	
Au_2a_4	6.9	7.4	7.2	
Au_2a_5	8.7	8.7	8.7	
Au_2a_6	10.9	11.1	11.0	8.70
Au_2b_1	14.9	14.3	14.6	
Au_2b_2	7.9	8.2	8.0	
Au_2b_3	8.4	8.5	8.5	
Au_2b_4	11.1	11.0	11.0	
Au_2b_5	11.0	10.7	10.9	
Au_2b_6	10.3	10.5	10.4	10.60
Au_2c_1	6.8	6.8	6.8	
Au_2c_2	9.1	9.2	9.2	
Au_2c_3	7.9	7.7	7.8	
Au_2c_4	6.9	6.6	6.7	
Au_2c_5	6.6	6.6	6.6	
Au_2c_6	8.5	8.3	8.4	7.60
Au_2d_1	15.6	15.4	15.5	
Au_2d_2	13.8	14.4	14.1	
Au_2d_3	15.0	13.0	14.0	
Au_2d_4	17.9	?	17.9	
Au_2d_5	17.9	17.4	17.7	
Au_2d_6	15.1	14.9	15.0	15.69
Au_Control_1	5.8	5.7	5.7	
Au_Control_2	6.1	6.1	6.1	
Au_Control_3	7.1	7.0	7.1	
Au_Control_4	5.5	5.4	5.4	
Au_Control_5	5.5	5.6	5.6	
Au_Control_6	5.6	5.7	5.6	5.93

Antimicrobial activity of the peptides

Table S3. Overview of MIC values for **1a-d** and **2a-d** screened against *S. aureus*, *S. epidermidis*, *E. coli*, and *P. aeruginosa*.

MIC in µg/mL																MIC in µg/mL									
		1a			1b			1c			1d			2a			2b			2c			2d		
Strain	Tech Rep	BR1	BR2	BR3	BR1	BR2	BR3	BR1	BR2	BR3	BR1	BR2	BR3	BR1	BR2	BR3	BR1	BR2	BR3	BR1	BR2	BR3	BR1	BR2	BR3
<i>S. aureus</i> ATCC 9144	1	64	64	64	128	128	128	32	32	32	8	16	8	8	8	8	16	32	16	8	4	8	8	4	4
	2	64	64	64	128	128	128	32	32	32	8	16	8	8	8	8	16	32	32	8	8	8	4	4	8
	3	64	64	64	128	128	128	32	32	32	8	16	16	8	8	8	16	32	32	8	4	8	4	4	4
<i>S. epidermidis</i> 1457	1	32	32	32	128	128	64	16	16	16	8	8	16	4	8	8	16	16	16	8	4	8	4	2	2
	2	32	32	32	128	128	128	16	16	16	8	8	16	4	8	8	16	16	16	4	4	8	2	2	2
	3	32	32	32	128	128	64	16	16	16	8	8	16	4	8	8	16	16	16	4	4	8	2	2	2
<i>E. coli</i> ATCC 25922	1	128	256	256	>256	>256	>256	32	64	32	64	64	64	64	64	128	128	128	8	8	16	64	64	64	
	2	128	256	256	>256	>256	>256	64	64	32	64	64	64	64	64	128	128	128	8	8	16	64	64	32	
	3	128	256	256	>256	>256	>256	64	64	32	64	64	64	64	64	128	128	128	8	16	8	64	64	32	
<i>P. aeruginosa</i> ATCC 27853	1	256	256	256	>256	>256	>256	64	64	32	128	256	128	64	64	64	128	256	256	8	8	16	256	256	256
	2	256	256	256	>256	>256	>256	64	64	32	128	256	128	64	64	64	128	256	256	16	16	16	256	256	256
	3	256	256	256	>256	>256	>256	64	64	32	128	256	128	64	64	64	128	256	256	16	8	16	256	256	256