Supporting Information

Exploring *N*-arylsulfonyl-L-proline scaffold as a platform for potent and selective αvβ1 integrin inhibitors

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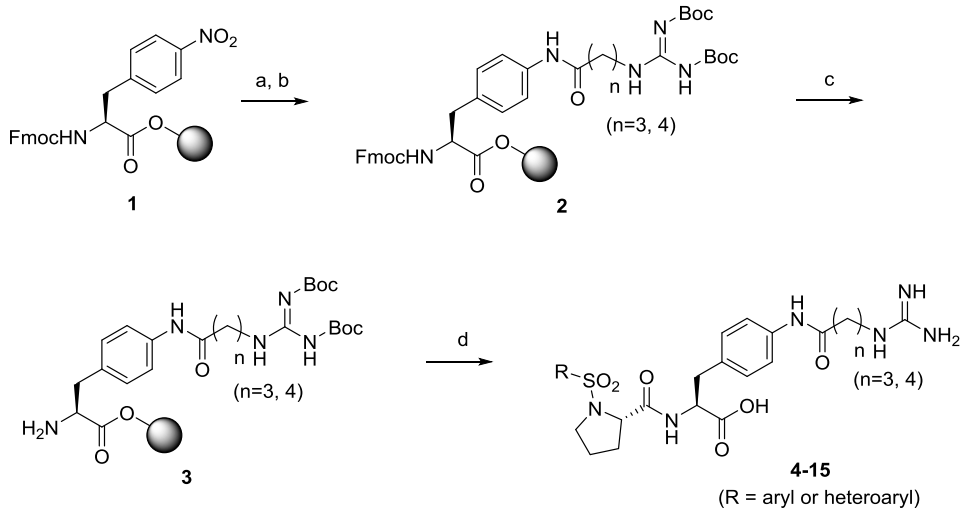
Syntheses of analogs 4-15



a. **Reduction of NO_2 to NH_2 on solid support**: To a slurry of Fmoc 4-nitrophenylalanine Wang resin **1** (10g, 0.55 meq/g) in DMF (30mL) was added aqueous 2M solution of $SnCl_2$ (13 mL) and heated at 80 ^oC for 5h. After cooling down to rt, the resin was washed with DMF extensively and finally washed with methanol and dried under vacuum to give the crude amine on resin.

b. **Synthesis of amide**: To a DCM swollen amino resin (10g, 0.55 meq/g assumed) was added N,N'-di-Boc-guanidino valeric (n=4) / N,N'-di-Boc-guanidino butanoic acid(n=3) (10g, 27.8 mmol) in DCM (25mL) , DIPEA (9.8 mL, 56 mmol), and HCTU (10.7g, 26 mmol) in DCM and shaken overnight. The resin was filtered and washed with DCM and DMF successively to give the N-Fmoc protected amide resin.

c. **Removal of Fmoc group**: The N-Fmoc protected amide resin (10g, 0.55 meq/g assumed) was treated with 20% 4-methylpiperidine in DMF (200 mL) for 5 min and repeated once to give the amino resin **3**. The resin was washed with DMF (x3), DCM (x3), and MeOH (x3) and dried under vacuum.

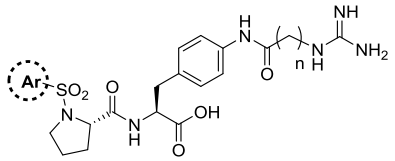
d. **(i) coupling to Fmoc-proline**: amino resin **3** (4g, 0.55 meq/g assumed) was treated with a solution of Fmoc-proline (3.7g,11 mmol), HCTU (4.5g, 10.8 mmol) and DIPEA (3.82 mL, 22 mmol) in DMF (11 mL) for 3h and washed with DMF (x3)
(ii) Removal of Fmoc group: The resin (4g, 0.55 meq/g assumed) was treated with 20% 4-methylpiperidine in DMF (100 mL) for 5 min and repeated once. The resin was washed with DMF (x3), DCM (x3), and MeOH (x3) and dried under vacuum.

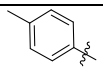
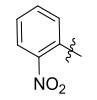
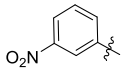
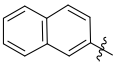
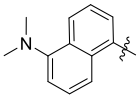
(iii) N-arylsulfonyl amide synthesis: the resin (0.4g, 0.55 meq/g assumed) in DMF (3mL) was treated with DIPEA (0.106 mL, 0.6 mmol) and arylsulfonyl chloride (0.3

mmol) for 1h and washed with DMF (x3), DCM (x3) and dried under vacuum

(iv) Deprotection and Cleavage: The resin was treated with a mixture of TFA:TIPS:H_2O (95:2.5:2.5) (10mL) for 3h and filtered. The filtrate was concentrated under a stream of N_2 and the product was obtained by Et_2O precipitation. The crude product was purified by RP-HPLC. HPLC analysis was performed using C4 Vydac column (4.6x250 mm), flow rate 0.9 ml/min, solvent A: 0.1% TFA in H_2O, solvent B: 0.1% in acetonitrile/H_2O (99/1), 254 (or 280 nm) detection. The mass spectrometry analysis was performed in Qtrap 3200 using positive ion mode.

Table S1. Characterization table of analogs 4-15.

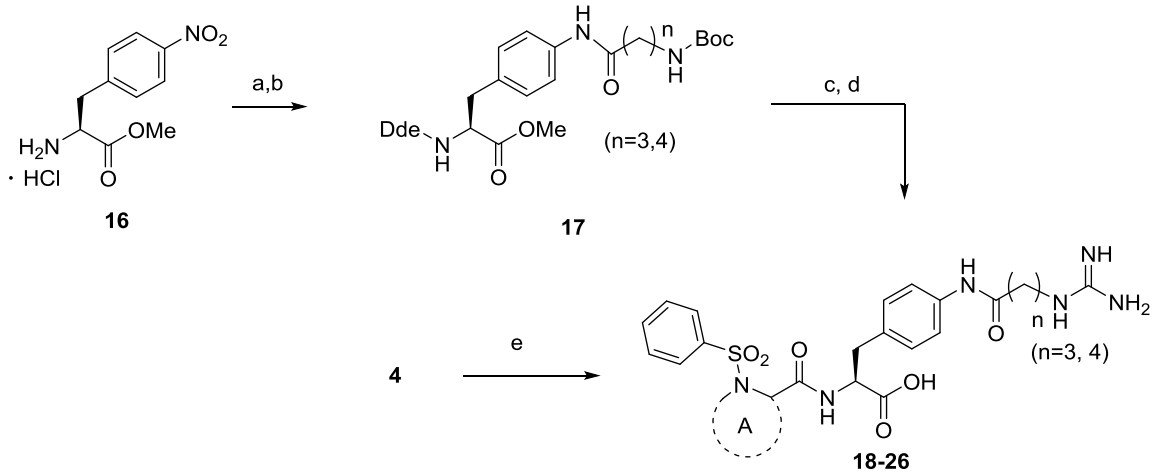


Compound	R	n	Retention time (min)	m/z
4		3	16.2	560.0 (MH+)
5		3	17.4	573.9 (MH+)
6		4	16.8	602.0 (MH+)
7		4	15.4	595.9 (MH+)
8		4	15.9	591.8 (MH+)
9		4	15.4	604.9 (MH+)
10		4	15.3	604.8 (MH+)
11		4	13.5	560.9 (MH+)
12		4	14.6	566.0 (MH+)
13		4	17.4	609.7 (MH+)
14		4	14.4	653.0 (MH+)
15		4	17.4	624.0 (MH+)

Molecular docking structure of c6 in αvβ1 integrin through homology modeling.

The modeling process for docking c6 was performed using MOE (molecular operating environment)[1] software package (Chemical Computing Group) according to the provided protocol by the Chemical Computing Group. The αvβ1 integrin protein model was constructed by alignment of the two published ligand-bound integrin protein structures (PDB ID: 1L5G, 4WK4) in Pymol. Average RMSD after alignment is 0.924 Å after 5 cycles. The initial ligand pdb structure was generated by Chem3D. After preparation of both the protein and the ligands in MOE, docking was performed with or without a pharmacophore model featuring carboxylate and guanidine in the structure as a guide to steer the docking process. Thus generated structures were checked by visual inspection.

Syntheses of analogs 18-26



a. **Dde protection of amine 16 and nitro reduction**: A mixture of amine **16** (1.3g, 5 mmol), DIPEA (0.87 mL, 5mmol), and 2-acetyldimedone (0.9 g, 5mmol) in MeOH (35 mL) was refluxed for 3h and cooled down. $SnCl_2 \cdot 2H_2O$ (5.6g, 25mmol) was added to the mixture and refluxed for additional 2h and cooled. The reaction mixture was poured into ice and neutralized with saturated $NaHCO_3$ solution to pH=8. Ethyl acetate was added and the mixture was filtered through a pad of Celite. The organic layer was washed with brine, dried over anhydrous Na_2SO_4, and concentrated to provide the amine as a yellow solid (1.74g, 97%) ESI-MS: 359.8 (MH^+) ^1H NMR (300 MHz, $CDCl_3$) δ ppm 1.04 (s, 6 H) 2.27 (s, 3 H) 2.33 (s, 2 H) 2.43 (s, 2 H) 2.96 (dd, J=13.94, 8.85 Hz, 1 H) 3.18 (dd, J=13.94, 4.71 Hz, 1 H) 3.63 (br. s, 2 H) 3.78 (s, 3 H) 4.53 (td, J=8.48, 4.90 Hz, 1 H) 6.61 (d, J=8.29 Hz, 2 H) 6.96 (d, J=8.29 Hz, 2 H) 13.89 (d, J=7.72 Hz, 1 H)

b. **Synthesis of amide 17**: To the amine (1.1 g, 3.1 mmol) in DMF (5.5 mL) was added Boc-5-aminovaleric acid (0.78 g, 3.6 mmol), HCTU (1.5 g, 3.6 mmol), and DIPEA (1.6 mL, 9.0 mmol). The mixture was stirred for 5 hours at rt. The mixture was diluted with ethyl acetate, washed with 10% aq. citric acid, washed with sat. aq. $NaHCO_3$, washed twice with water, washed with brine, dried over anhydrous Na_2SO_4, and concentrated to provide the crude **17** (n=4). (1.8g, quant.) ESI-MS: 559.0 (MH^+) ^1H NMR (300 MHz, $CDCl_3$) δ ppm 1.03 (s, 6 H) 1.44 (s, 9 H) 1.52 - 1.66 (m, 2 H) 1.68 - 1.83 (m, 2 H) 2.27 (s, 3 H) 2.31 - 2.50 (m, 6 H) 3.04 (dd, J=13.85, 8.76 Hz, 1 H) 3.13 - 3.31 (m, 3 H) 3.78 (s, 3 H) 4.58 (td, J=8.48, 4.90 Hz, 1 H) 4.67 (m, 1 H) 7.13 (d, J=8.29 Hz, 2 H) 7.49 (d, J=8.29 Hz, 2 H) 7.56 (br. s, 1 H) 13.93 (d, J=8.29 Hz, 1 H)

c. **Removal of Boc and bis-boc guanidylation**: 1.7 g (3.0 mmol) of crude Boc-amine **17** was stirred in dichloromethane (8 mL) and trifluoroacetic acid (8 mL) for

90 minutes. The mixture was concentrated to provide 2.6 g of crude amine trifluoroacetate salt. ESI-MS: 458.8 (MH^+) ^{1}H NMR (300 MHz, DMSO-d6) δ ppm 0.94 (s, 6 H) 1.47 - 1.69 (m, 4 H) 2.20 - 2.38 (m, 5 H) 2.71 - 2.86 (m, 2 H) 2.97 - 3.08 (m, 1 H) 3.09 - 3.19 (m, 1 H) 3.71 (s, 3 H) 4.99 (td, J=7.49, 5.37 Hz, 1 H) 7.07 (d, J=8.48 Hz, 2 H) 7.50 (d, J=8.48 Hz, 2 H) 7.65 (br. s, 3 H) 9.91 (s, 1 H) 13.53 (d, J=8.10 Hz, 1 H) To a mixture of a portion of amine trifluoroacetate salt (0.71 g, 1.2 mmol) in DMF (4 mL) was added N,N'-di-Boc-1H-pyrazole-1-carboxamidine (0.46 g, 1.5 mmol) and DIPEA (0.54 mL, 3.1 mmol). The mixture was stirred for 18 hours at rt. The mixture was diluted with ethyl acetate, washed with water twice, washed with brine, dried over anhydrous Na_2SO_4, and concentrated. The crude residue was purified by flash chromatography (40 g column silica, gradient elution 0 to 100 % ethyl acetate/hexanes) to provide the bis-Boc protected guanidine (0.47 g, 56%). ESI-MS: 701.3 (MH^+) ^{1}H NMR (300 MHz, $CDCl_3$) δ ppm 1.03 (s, 6 H) 1.47 (s, 9 H) 1.50 (s, 9 H) 1.57 - 1.85 (m, 4 H) 2.29 (s, 3 H) 2.37 - 2.47 (m, 2 H) 3.04 (dd, J=13.94, 8.85 Hz, 1 H) 3.25 (dd, J=13.94, 4.71 Hz, 1 H) 3.46 (q, J=6.66 Hz, 2 H) 3.78 (s, 3 H) 4.57 (td, J=8.43, 4.80 Hz, 1 H) 7.14 (d, J=8.48 Hz, 2 H) 7.47 (d, J=8.29 Hz, 2 H) 7.64 (s, 1 H) 8.40 (br. s, 1 H) 11.47 (s, 1 H) 13.92 (d, J=8.10 Hz, 1 H)

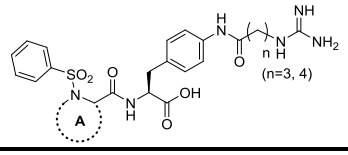
d. **Dde removal,coupling, and deprotection of Boc group**: The Dde amine (0.47g, 0.56 mmol) was stirred in DMF (2 mL) and 25% aq. hydrazine hydrate (0.2mL) for 90 minutes. The diluted with water and extracted with ethyl acetate 4 times. The combined organic layers were dried over anhydrous $MgSO_4$ and concentrated. The crude residue was purified by flash chromatography (40 g column silica, gradient elution 0 to 15 % MeOH/ethyl acetate) to provide the amine (0.30g, quant). ESI-MS: 536.7 (MH^+) ^{1}H NMR (300 MHz, $CDCl_3$) δ ppm 1.47 (s, 9 H) 1.50 (s, 9 H) 1.62 - 1.74 (m, 2 H) 1.74 - 1.85 (m, 2 H) 2.43 (t, J=6.78 Hz, 2 H) 2.78 - 2.88 (m, 1 H) 3.06 (dd, J=13.66, 4.62 Hz, 1 H) 3.47 (q, J=5.90 Hz, 2 H) 3.72 (s, 3 H) 7.14 (d, J=7.91 Hz, 2 H) 7.46 (d, J=7.91 Hz, 2 H) 7.62 (br. s, 1 H) 8.40 (br. s, 1 H) 11.48 (br. s, 1 H). To the resulting amine (5mmol) in DMF (9 mL) was added N-arylsulfonyl-L-proline (6mmol), HCTU (2.5g, 6mmol), DIPEA (2.6 mL, 15 mmol) and stirred for 18h. The mixture was diluted with ethyl acetate and washed with 10% citric acid, sat. $NaHCO_3$ solution, and brine. The organic phase was dried over Na_2SO_4, filtered and the solvent was removed in vacuo to give the crude mixture. The bis-Boc group was then removed by treatment with 50% TFA in DCM (20 mL) for 3 h. The volatiles were then removed and dissolved in THF:H_2O (4:1). Solid LiOH or 1M NaOH solution was added until the solution became basic. The mixture was stirred for 2h and neutralized with 1M HCl. The volatiles were removed under reduced pressure and the product was purified by RP-HPLC.

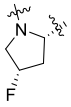
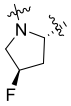
e. **Solid Phase Synthesis and Cleavage**: To the resin **4** (0.4g, 0.55 meq/g assumed) in DMF (5 mL) was added N-arylsulfonyl-L-proline (5 eq), HCTU (5eq), DIPEA (10) and agitated in 75 ^{o}C for 10 min. The resin was washed with DMF(x3) and DCM(x3). The product was cleaved from the resin by treatment with a cleavage cocktail (TFA:TIPS:H_2O=95:2.5:2.5) for 3h. The resin was filtered and the filtrate was concentrated by a stream of N_2 and the crude product was obtained by Et_2O precipitation and purified by RP-HPLC. HPLC analysis was performed using C4

Vydac column (4.6x250 mm), flow rate 0.9 ml/min, solvent A: 0.1% TFA in H_2O, solvent B: 0.1% in acetonitrile/H_2O (99/1), 254 (or 280 nm) detection. The mass spectrometry analysis was performed in Qtrap 3200 using positive ion mode.

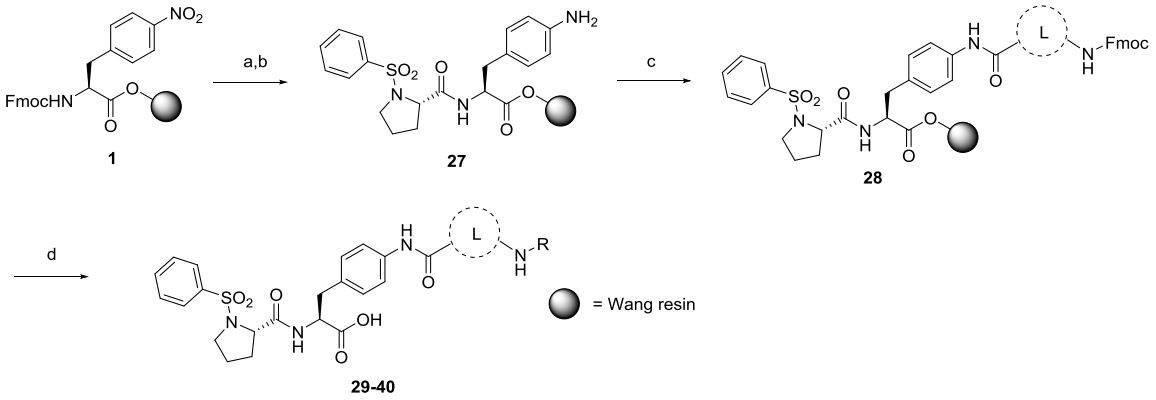
Table S2. Characterization table for compounds **18-26**.

* denotes the retention time obtained using Varian Microsorb 100-5 C18 (4.6x250 mm) under identical condition.



Compound	R	n	Retention time (min)	m/z
18		4	24.4*	573.9 (MH+)
19		3	22.8*	560.0 (MH+)
20		4	20.2 *	557.9 (MH+)
21		4	20.3*	545.9 (MH+)
22		4	16.7	578.0 (MH+)
23		4	16.4	606.0 (MH+)
24		4	14.8	577.9 (MH+)
25		4	14.6	578.0 (MH+)
26		4	22.4*	573.9 (MH+)

Syntheses of analogs 29-40



a. **(i) Removal of Fmoc group**: The N-Fmoc protected amide resin (10g, 0.55 meq/g assumed) was treated with 20% 4-methylpiperidine in DMF (200 mL) for 5 min and repeated once to give the amino resin. The resin was washed with DMF (x3), DCM (x3), and MeOH (x3) and dried under vacuum.

(ii) Synthesis of amide: amino resin (4g, 0.55 meq/g assumed) was treated with a solution of *N*-benzenesulfonyl-L-proline (2.8 g,11 mmol), HCTU (4.5g, 10.8 mmol) and DIPEA (3.82 mL, 22 mmol) in DMF (11 mL) for 3h and washed with DMF (x3).

b. **Synthesis of amine 27**: To a slurry of above resin in DMF (30mL) was added aqueous 2M solution of $SnCl_2$ (13 mL) and heated at 80 ^oC for 5h. After cooling down to rt, the resin was washed with DMF extensively and finally washed with methanol and dried under vacuum to give the crude amine on resin.

c. **coupling to Fmoc-amino acids**: amino resin **3** (0.5g, 0.55 meq/g assumed) was treated with a solution of Fmoc-amino acids (5eq), HCTU (5eq) and DIPEA (10 eq) in DMF (10 mL) for 3h and washed with DMF (x3)

d. **(i) Removal of Fmoc group**: The N-Fmoc protected resin (0.5g, 0.55 meq/g assumed) was treated with 20% 4-methylpiperidine in DMF (10 mL) for 5 min and repeated once to give the amino resin. The resin was washed with DMF (x3), DCM (x3), and MeOH (x3) and dried under vacuum.

(ii) Derivatization of amine

Cyclic guanidine (compounds 29-30): The amino resin (0.5g, 0.55 meq/g assumed) was treated with 2-(Methylthio)-2-imidazoline hydriodide (2eq) and DIPEA (5eq) in DMF (10 mL) for 1h at 60 ^oC. The resin was washed with DMF (x3), DCM (x3), and MeOH (x3) and dried under vacuum.

Cyclic guanidine (compound 31): The amino resin (0.5g, 0.55 meq/g assumed) was treated with 2-(Methylthio)-1,4,5,6-tetrahydropyrimidine hydroiodide (2eq) and DIPEA (5eq) in DMF (10 mL) for 1h at 60 ^{o}C. The resin was washed with DMF (x3), DCM (x3), and MeOH (x3) and dried under vacuum.

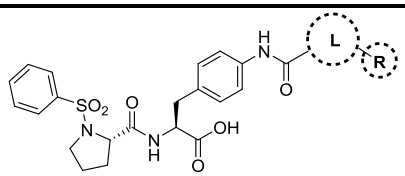
Aminopyridine (compounds 32-35): The amino resin (0.5g, 0.55 meq/g assumed) was treated with a solution of pyridine N-oxide (5eq), DIPEA (18.75 eq), PyBroP (6.5eq) at rt overnight. The resin was washed with DMF (x6), DCM (x6), and MeOH (x3) and dried under vacuum.

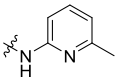
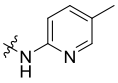
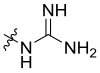
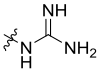
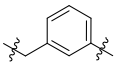
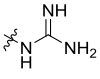
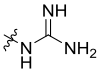
Guanidine (compounds 36-40): The amino resin (0.5g, 0.55 meq/g assumed) was treated with *N*,*N*'-di-Boc-1H-pyrazole-1-carboxamidine 2-(5eq) and DIPEA (5eq) in DMF (10 mL) for 1h at 60 ^{o}C. The resin was washed with DMF (x3), DCM (x3), and MeOH (x3) and dried under vacuum.

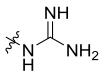
(iii) Deprotection and Cleavage: The resin was treated with a mixture of TFA:TIPS:H_2O (95:2.5:2.5) (10mL) for 3h and filtered. The filtrate was concentrated under a stream of N_2 and the product was obtained by Et_2O precipitation. The crude product was purified by RP-HPLC. HPLC analysis was performed using C4 Vydac column (4.6x250 mm), flow rate 0.9 ml/min, solvent A: 0.1% TFA in H_2O, solvent B: 0.1% in acetonitrile/H_2O (99/1), 254 (or 280 nm) detection. The mass spectrometry analysis was performed in Qtrap 3200 using positive ion mode.

Table S3. Characterization table for compounds **29-40**.

Retention time was obtained using 19x100mm Atlantis T3 OBD, detection: 254 nm, Solvent A: 0.1% TFA in H_2O, Solvent B': 0.1% TFA in 60% *i*-PrOH, 30% acetonitrile, 10% H_2O. **condition A**: flow rate: 10 ml/min, gradient: 5% to 70% solvent B' for 43 min, **condition B**: flow rate: 13 ml/min, gradient: 8% solvent B' for 5 min, 8% to 18% solvent B' for 1min, 18% to 24% solvent B' for 24 min. **condition C**: flow rate: 13 ml/min, gradient: 8% solvent B' for 5 min, 8% to 20% solvent B' for 1min, 20% to 26% solvent B' for 24 min. **condition D:** flow rate: 15 ml/min, gradient: 8% solvent B' for 3 min, 8% to 15% solvent B' for 1min, 15% to 24% solvent B' for 30 min, **condition E**: flow rate: 10 ml/min, gradient: 5% to 60% solvent B' for 30 min. **condition F**: flow rate: 15 ml/min, gradient: 5% to 100% solvent B' for 37 min, **condition G**: flow rate: 15 ml/min, gradient: 5% to 100% solvent B' for 35 min, **condition H**: flow rate: 10 ml/min, gradient: 5% to 70% solvent B' for 40 min



Compound	L	R	Retention time (min)	Condition	m/z
29	-$(CH_2)_4$-		24.3	A	585.9 (MH+)
30	-$(CH_2)_3$-		24.1	A	572.0 (MH+)
31	-$(CH_2)_4$-		18.6	B	598.0 (MH+)
32	-$(CH_2)_4$-		15.5	C	593.0 (MH+)
33	-$(CH_2)_3$-		19.2	D	580.0 (MH+)
34	-$(CH_2)_3$-		26.2	A	594.9 (MH+)
35	-$(CH_2)_3$-		23.6	E	594.9 (MH+)
36			16.2	F	579.9 (MH+)
37			15.9	F	579.9 (MH+)
38			15.9	G	594.0 (MH+)
39			11.8	G	586.0 (MH+)

40			31.3	H	637.9 (MH+)

Characterization data for key compounds 18, 19, 29, 33, and 37

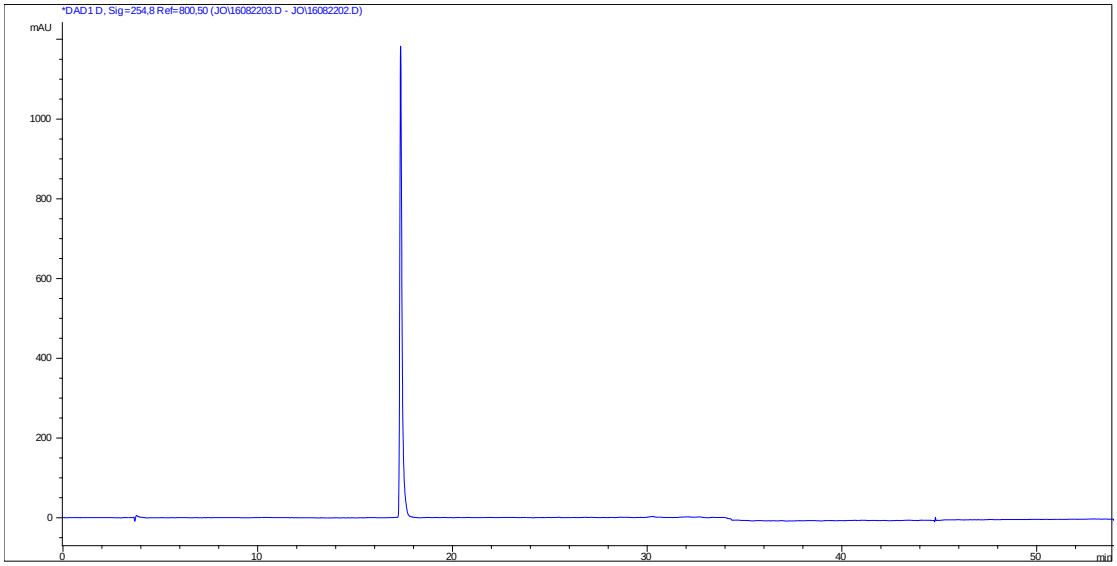
Compound 18:

1H NMR (900 MHz, DMSO-d6) δ ppm 1.12 (t, J=10.32 Hz, 2 H), 1.33 - 1.40 (m, 2 H), 1.42 - 1.51 (m, 3 H), 1.58 (quin, J=7.53 Hz, 2 H), 1.91 (d, J=13.20 Hz, 1 H), 2.31 (t, J=7.36 Hz, 2 H), 2.83 (dd, J=13.89, 9.90 Hz, 1 H), 3.01 (dd, J=13.96, 4.47 Hz, 1 H), 3.10 (q, J=6.60 Hz, 2 H), 3.19 - 3.25 (m, 1 H), 3.59 (d, J=11.69 Hz, 1 H), 4.22 (td, J=8.77, 4.75 Hz, 1 H), 4.49 (d, J=5.23 Hz, 1 H), 6.84 (br s, 2 H), 7.18 (d, J=8.39 Hz, 2 H), 7.35 (br s, 1 H), 7.40 (t, J=7.77 Hz, 2 H), 7.52 - 7.61 (m, 5 H), 8.10 (d, J=7.84 Hz, 1 H), 9.89 (s, 1 H), 12.73 (br s, 1 H).

^{13}C NMR (226 MHz, DMSO-d_6) δ ppm 18.74, 22.22, 23.66, 27.04, 28.14, 35.77, 35.80, 40.52, 42.46, 53.47, 54.10, 118.87, 126.76, 128.98, 129.37, 132.19, 132.55, 137.83, 139.42, 156.69, 169.80, 170.82, 172.80.

HRMS-ESI (m/z): [MH+] calcd for $C_{26}H_{35}N_6O_6S^+$, 559.2333; found, 559.2338.

Purity (>95%) by HPLC (Vydac C4 4.6x250mm, detection: 254 nm, Solvent A: 0.1% TFA in H_2O, Solvent B: 0.1% TFA in CH_3CN, 5% to 100% B over 30 min, 0.9mL/min



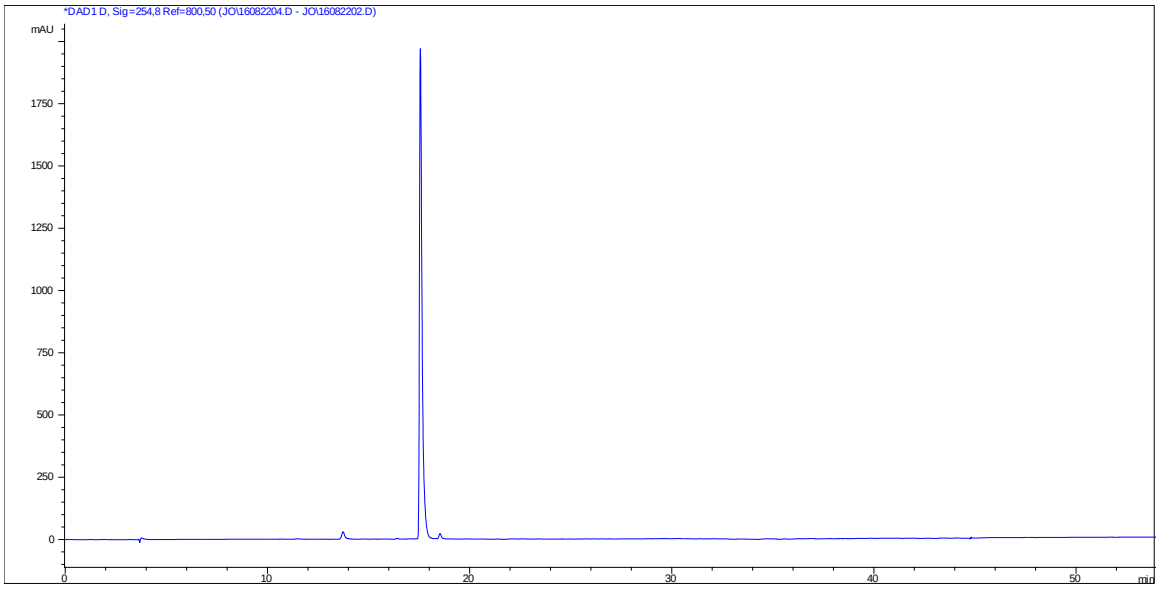
Compound 19:

^{1}H NMR (900 MHz, DMSO-d_6) δ ppm 1.13 (t, J=8.60 Hz, 2 H), 1.31 - 1.40 (m, 2 H), 1.44 (br. s., 1 H), 1.76 (t, J=6.95 Hz, 2 H), 1.91 (d, J=12.52 Hz, 1 H), 2.34 (t, J=7.08 Hz, 2 H), 2.83 (dd, J=13.41, 10.25 Hz, 1 H), 3.01 (dd, J=13.82, 3.78 Hz, 1 H), 3.10 – 3.14 (m, 2 H), 3.208 - 3.24 (m, 1 H), 3.59 (d, J=12.10 Hz, 1 H), 4.20 - 4.23 (m, 1 H), 4.47 - 4.50 (m, 1 H), 6.85 (br s, 1 H), 7.19 (d, J=8.25 Hz, 2 H), 7.38 - 7.43 (m, 2 H), 7.51 - 7.63 (m, 5 H), 8.10 (d, J=7.84 Hz, 1 H), 9.95 (s, 1 H), 12.73 (br s, 1 H).

^{13}C NMR (226 MHz, DMSO-d_6) δ ppm 18.74, 23.66, 24.32, 27.03, 32.91, 35.75, 40.31, 42.46, 53.47, 54.09, 118.96, 126.76, 128.97, 129.38, 132.30, 132.56, 137.72. 139.42, 156.73, 169.80, 170.42, 172.80.

HRMS-ESI (m/z): [MH+] calcd for $C_{27}H_{37}N_6O_6S^+$, 573.2490; found, 573.2494

Purity (>95%) by HPLC (Vydac C4 4.6x250mm, detection: 254 nm, Solvent A: 0.1% TFA in H_2O, Solvent B: 0.1% TFA in CH_3CN, 5% to 100% B over 30 min, 0.9mL/min)



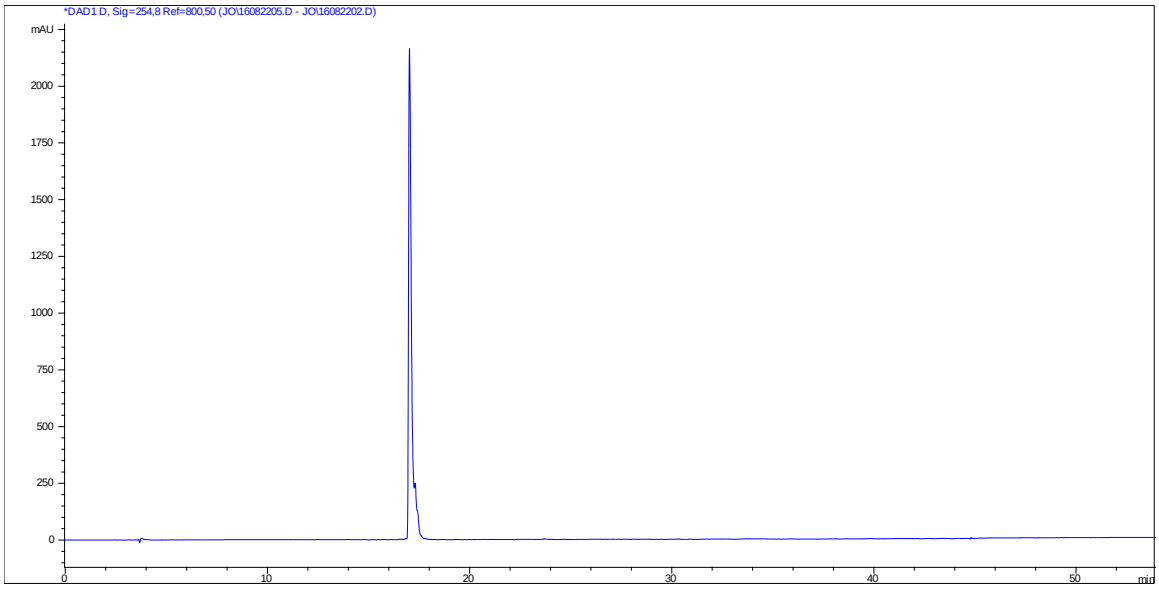
Compound 29:

^{1}H NMR (900 MHz, DMSO-d_6) δ ppm 1.44-1.48 (m, 1H), 1.50-1.54 (m, 2H), 1.55-1.64 (m, 3H), 1.65-1.69 (m, 2H), 2.34 (t, J=7.2 Hz, 2H), 2.96 (dd, J=8.64, 14.04 Hz, 1H), 3.06 (dd, J=4.95, 14.04 Hz, 1H), 3.15-3.18 (m, 4H), 3.37-3.39 (m, 2H), 4.28 (dd, J=2.7, 8.5 Hz, 1H), 4.44-4.47 (m, 1H), 7.20 (d, J=8.5 Hz, 2 H), 7.53 (d, J=8.5 Hz, 2H), 7.64 (t, J=7.8 Hz, 2H), 7.75 (t, J=7.4 Hz, 1H), 7.85 (d, J=7.3 Hz, 2H), 8.16 (d, J=8.0 Hz, 1H), 8.39 (t, J=3.87 Hz, 1 H), 9.91 (s, 1 H), 12.9 (br s, 1H).

^{13}C NMR (226 MHz, DMSO-d_6) δ ppm 25.42, 27.06, 31.59, 33.64, 39.00, 39.30, 45.19, 45.66, 52.23, 56.62, 64.39, 122.03, 130.61, 132.61, 132.70, 135.14, 136.43, 139.98, 140.99, 162.61, 174.02, 174.06, 175.82.

HRMS-ESI (m/z): [MH+] calcd for $C_{28}H_{37}N_6O_6S^+$, 585.2490; found, 585.2493

Purity (>95%) by HPLC (Vydac C4 4.6x250mm, detection: 254 nm, Solvent A: 0.1% TFA in H_2O, Solvent B: 0.1% TFA in CH_3CN, 5% to 100% B over 30 min, 0.9mL/min)



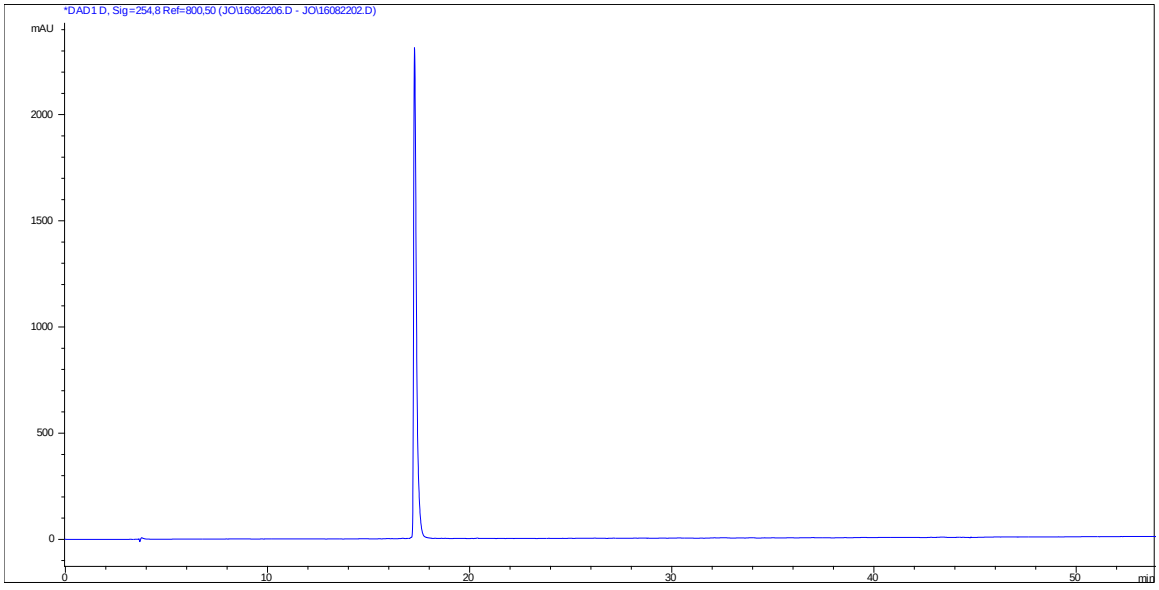
Compound 33:

^{1}H NMR (900 MHz, DMSO-d_6) δ ppm 1.44-1.47 (m, 1H), 1.56-1.61 (m, 1H), 1.64-1.68 (m, 2H), 1.92-1.95 (m, 2H), 2.46 (t, J=7.2 Hz, 2H), 2.95 (dd, J=8.64, 13.95 Hz, 1H), 3.06 (dd, J=4.77, 13.95 Hz, 1H), 3.37-3.39 (m, 2H), 4.18 (dd, J=2.61, 8.37 Hz, 1H), 4.45-4.47 (m, 1H), 6.86 (t, J=6.57, 1H), 7.07 (d, J=8.55 Hz, 1H), 7.20 (d, J=8.46 Hz, 2H), 7.52 (d, J=8.46 Hz, 2H), 7.64 (t, J=7.92 Hz, 1H), 7.84 (d, J=7.38 Hz, 1H), 7.90 (br s ,1H), 7.94 (d, J=5.22 Hz, 1H), 8.16 (d, J=8.01 Hz, 1H), 8.89 (br s, 1H).

^{13}C NMR (226 MHz, DMSO-d_6,) δ ppm 26.85, 27.06, 33.63, 36.32, 39.29, 44.31, 52.23, 56.61, 64.41, 115.08, 122.11, 130.61, 132.61, 132.71, 135.22, 136.43, 139.96, 140.89, 156.01, 173.58, 174.07, 175.82 (some aromatic peaks are missing due to the low sample amount)

HRMS-ESI (m/z): [MH+] calcd for $C_{29}H_{34}N_5O_6S^+$, 580.2224; found, 580.2233

Purity (>95%) by HPLC (Vydac C4 4.6x250mm, detection: 254 nm, Solvent A: 0.1% TFA in H_2O, Solvent B: 0.1% TFA in CH_3CN, 5% to 100% B over 30 min, 0.9mL/min)



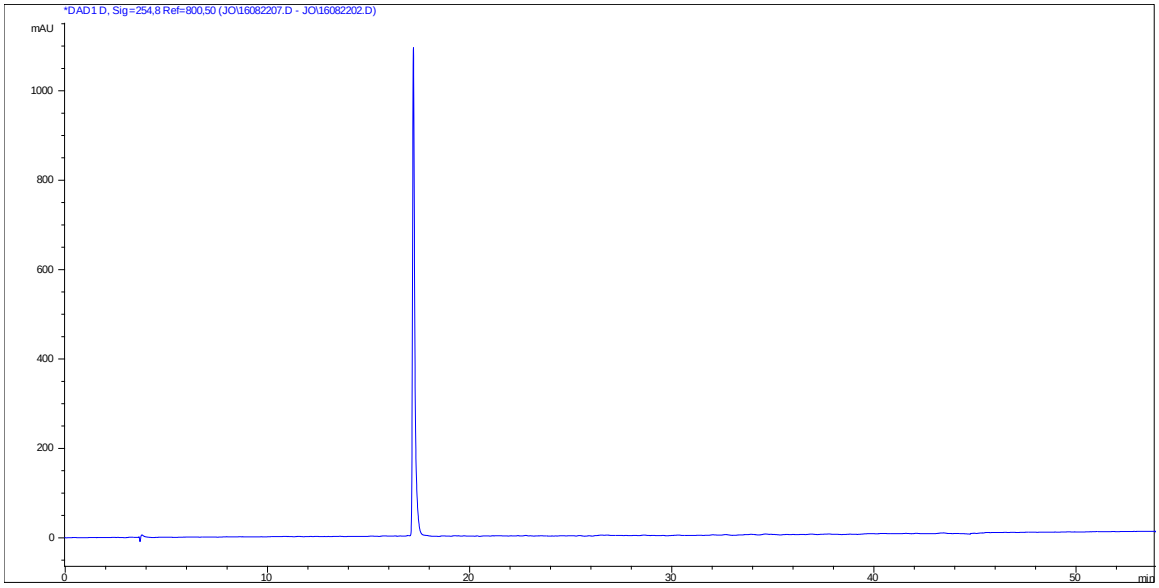
Compound 37:

^{1}H NMR (900 MHz, DMSO-d_6) δ ppm 1.46-1.49 (m, 1H), 1.58-1.61 (m, 1H), 1.67-1.7 (m, 2H), 1.90-1.96 (m, 2H), 3.01 (dd, J=8.73, 13.95 Hz, 1H), 3.11 (dd, J=4.86, 13.95 Hz, 1H), 3.35-3.41 (m ,1H), 4.20 (dd, J=2.61, 8.37 Hz, 1H), 4.49-4.51 (m, 1H), 7.29 (d, J=8.37 Hz, 2H), 7.47 (d, J=7.74, 1H), 7.6-7.7 (m, 5H), 7.7-7.75 (m, 3H), 7.85-7.89 (m, 3H), 7.91 (d, J= 7.83 Hz, 1H), 8.20 (d, J=8.01 Hz, 1H), 10.01 (s, 1H), 10.27 (s, 1H)

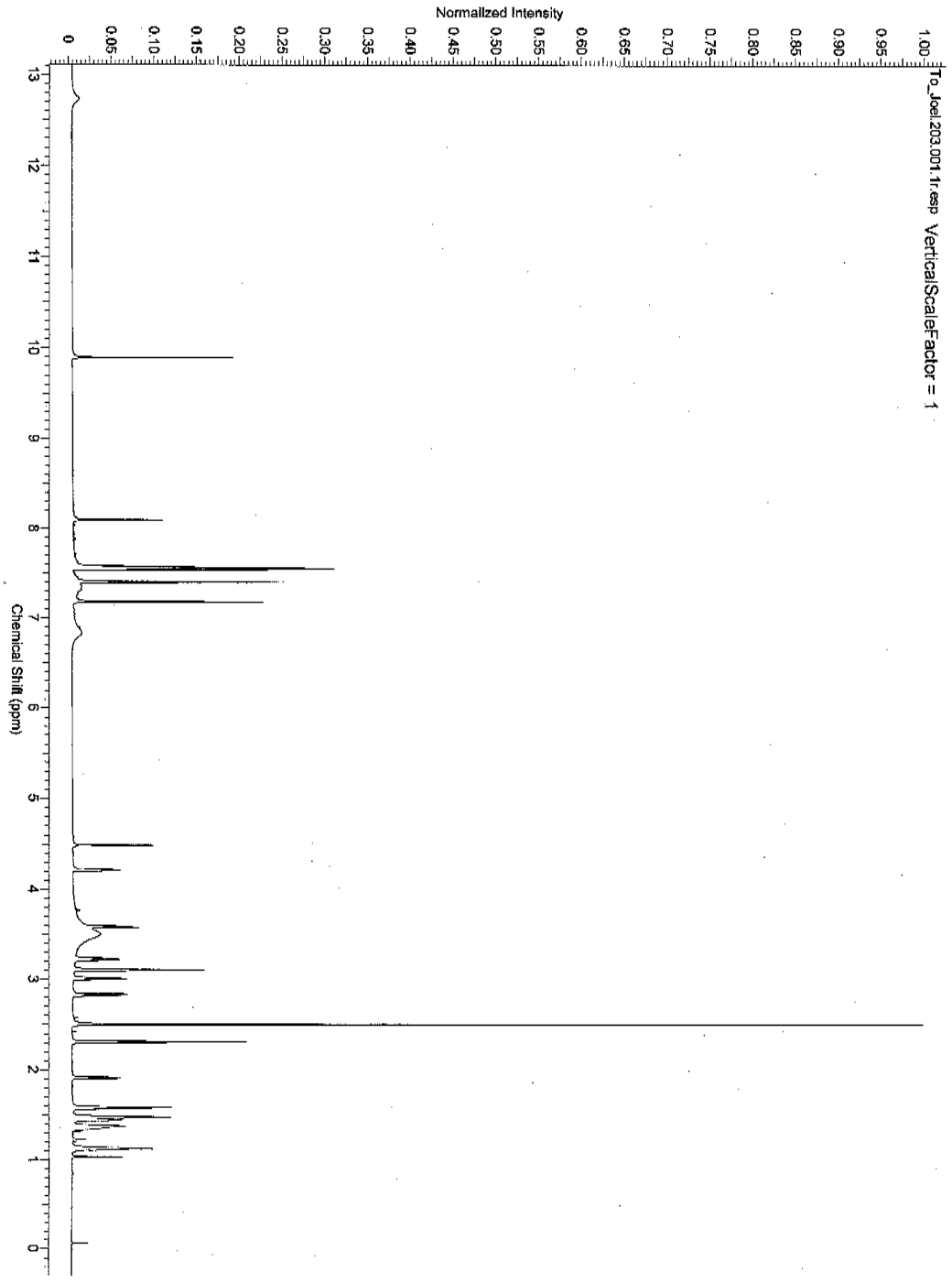
^{13}C NMR (226 MHz, DMSO-d_6) δ ppm 24.80, 31.38, 37.12, 49.9, 54.31, 62.15, 116.9, 121.08, 124.2, 126.3, 128.34, 130.33, 130.44, 130.7, 133.9, 134.2, 136.58, 137.1, 137.7, 138.4, 156.72, 165.33, 171.79, 173.53.

HRMS-ESI (m/z): [MH+] calcd for $C_{28}H_{31}N_6O_6S^+$, 579.2023; found, 579.2020.

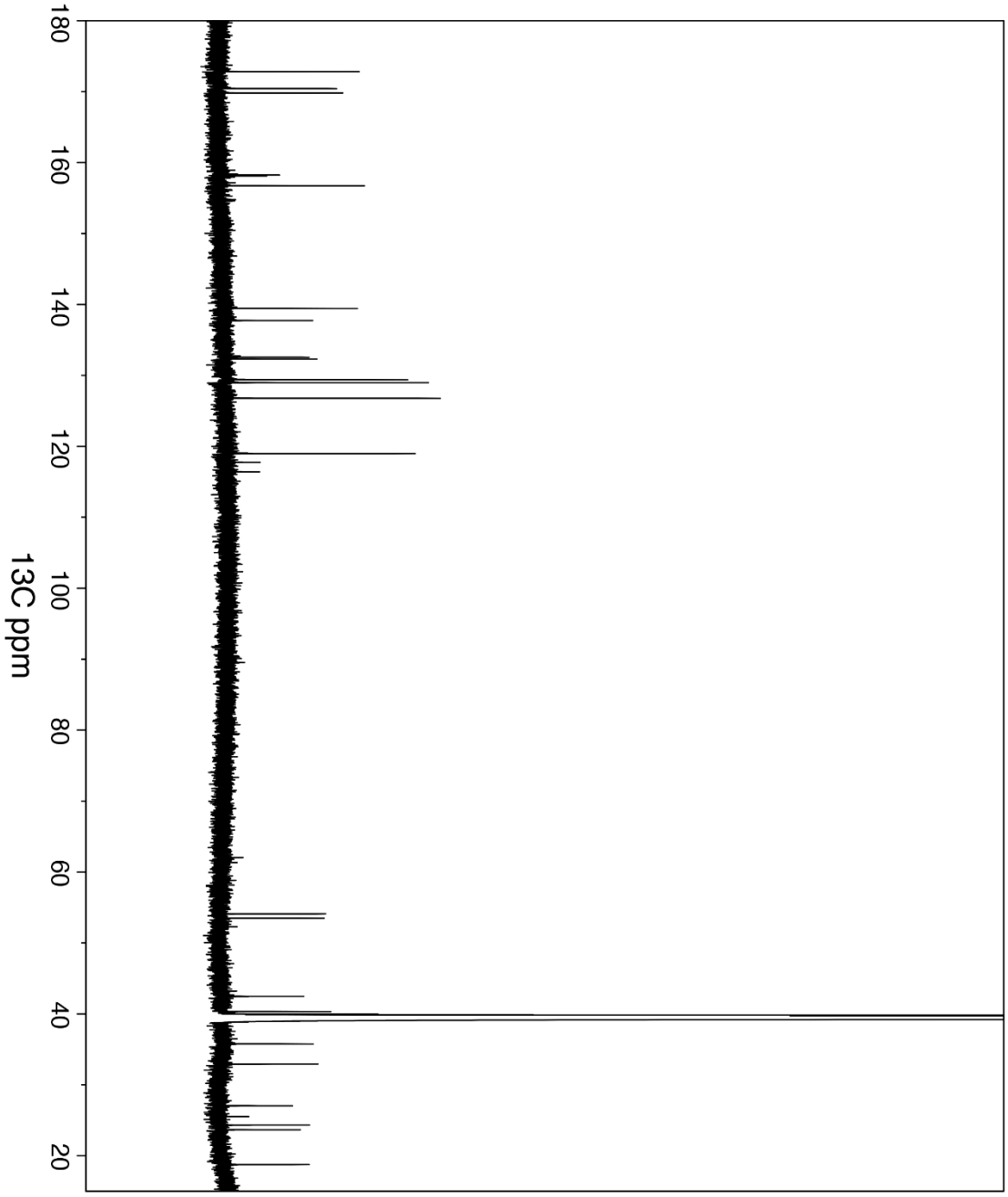
Purity (>95%) by HPLC (Vydac C4 4.6x250mm, detection: 254 nm, Solvent A: 0.1% TFA in H_2O, Solvent B: 0.1% TFA in CH_3CN, 5% to 100% B over 30 min, 0.9mL/min)



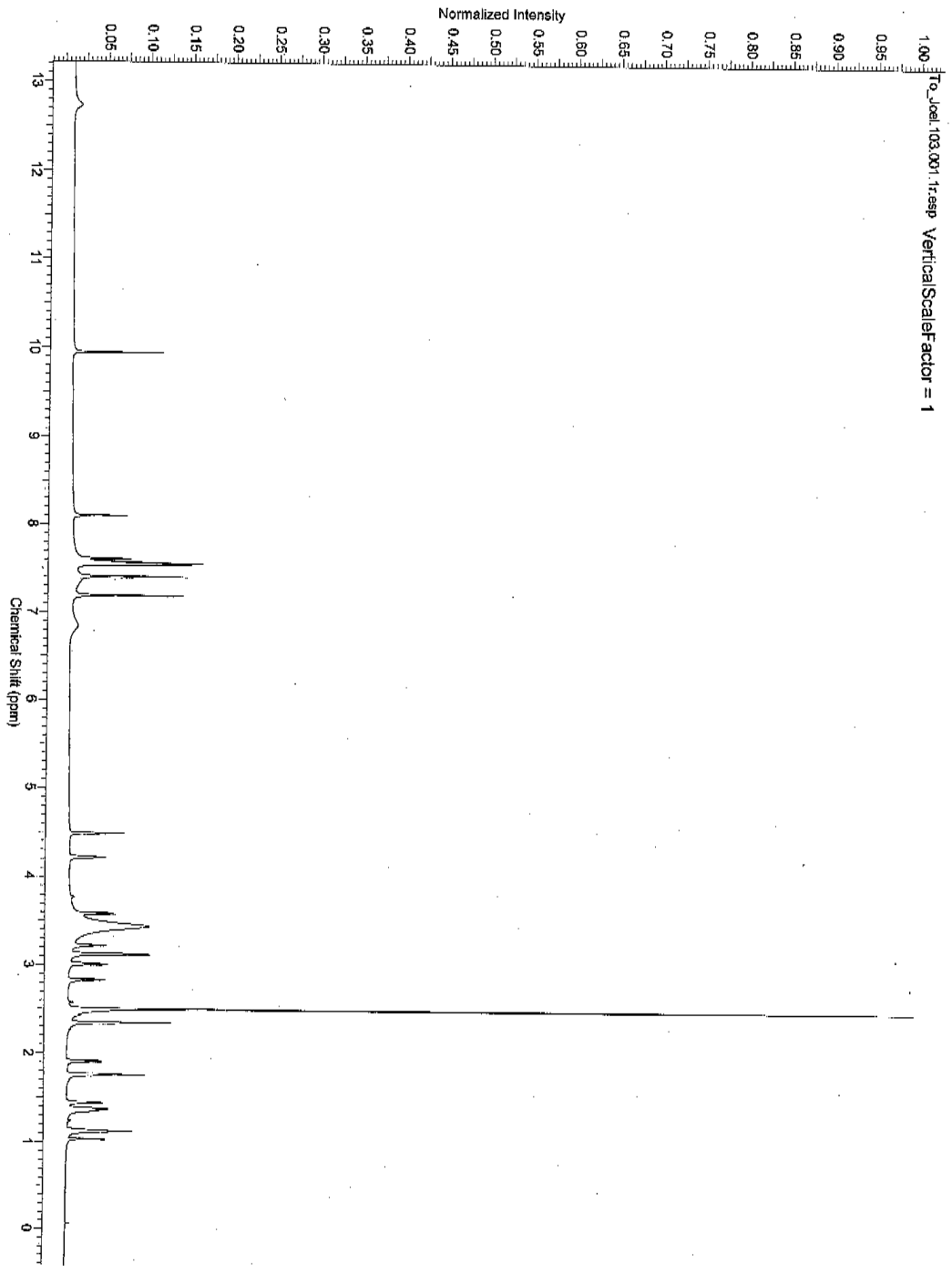
COPY of 1H NMR spectrum of compound 18



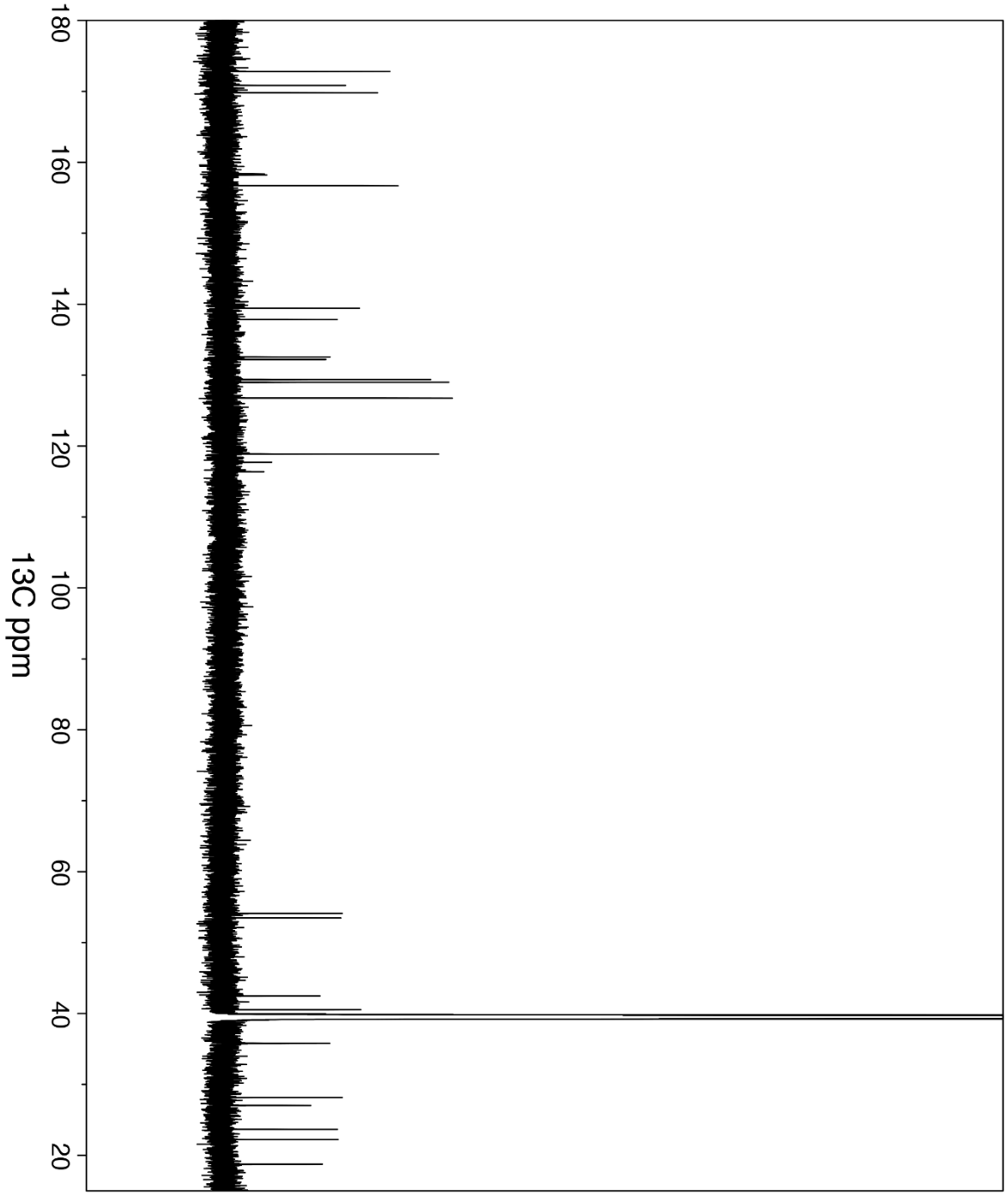
COPY of ^{13}C NMR spectrum of compound **18**



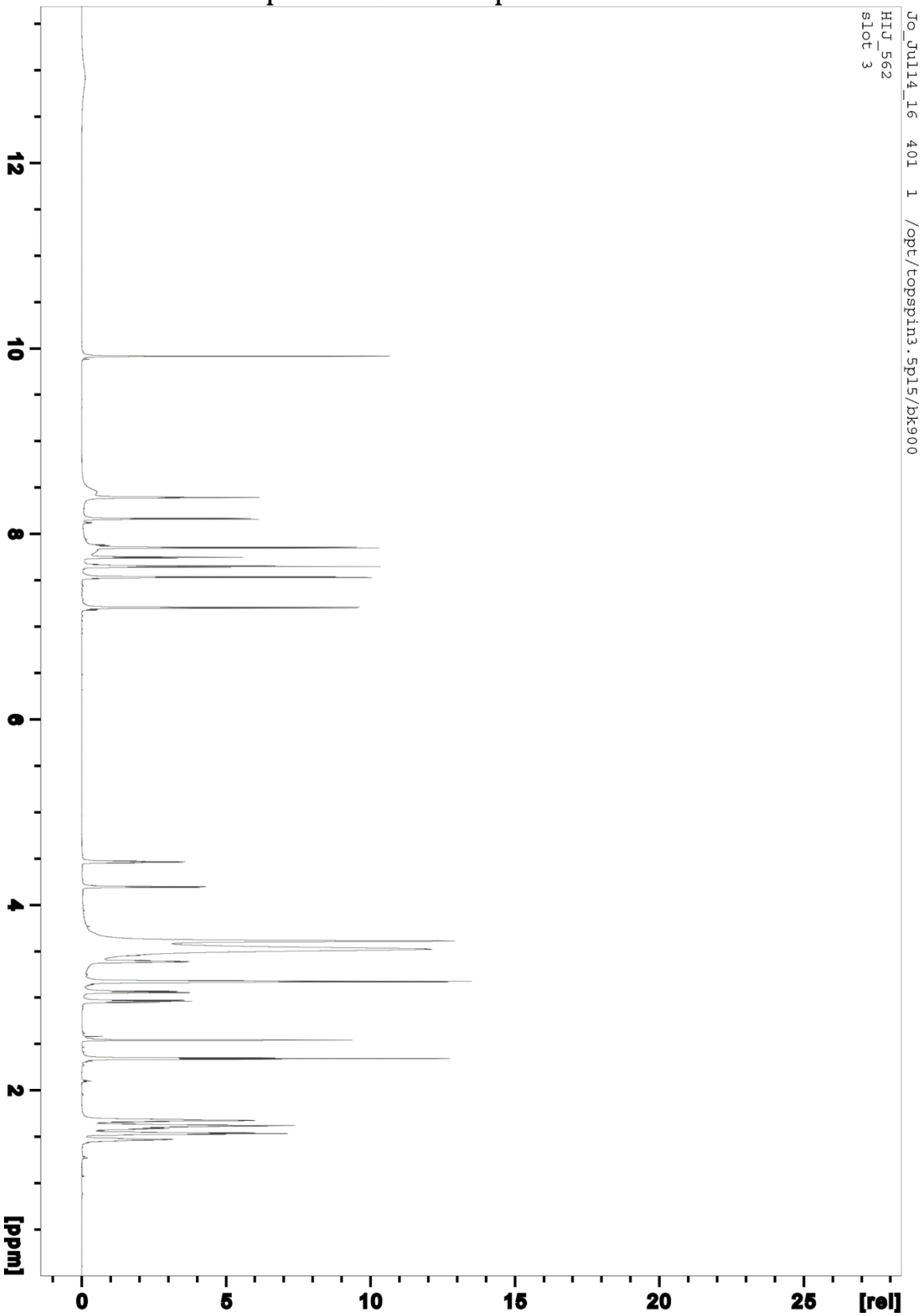
COPY of ^{1}H NMR spectrum of compound **19**



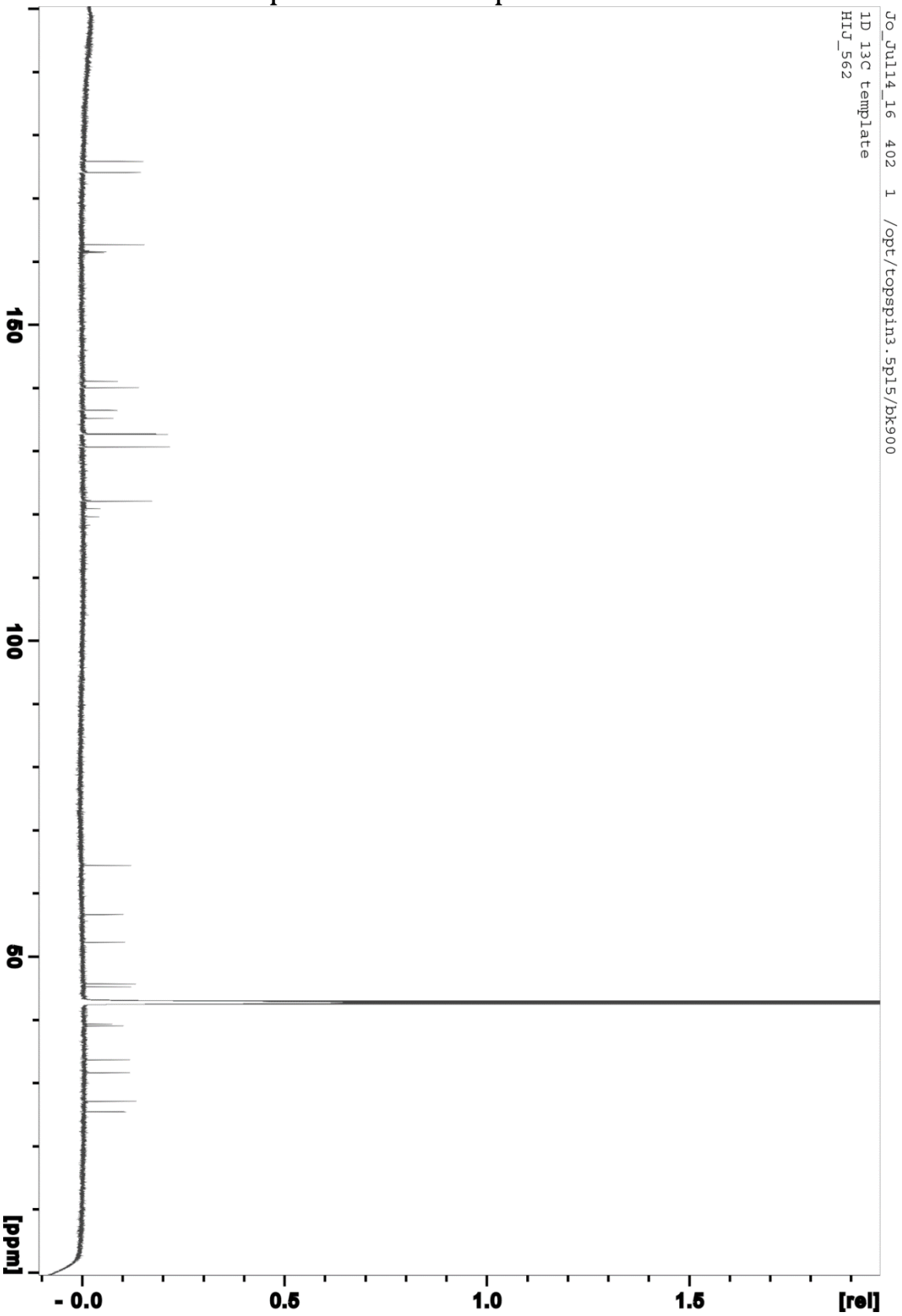
COPY of ^{13}C NMR spectrum of compound **19**



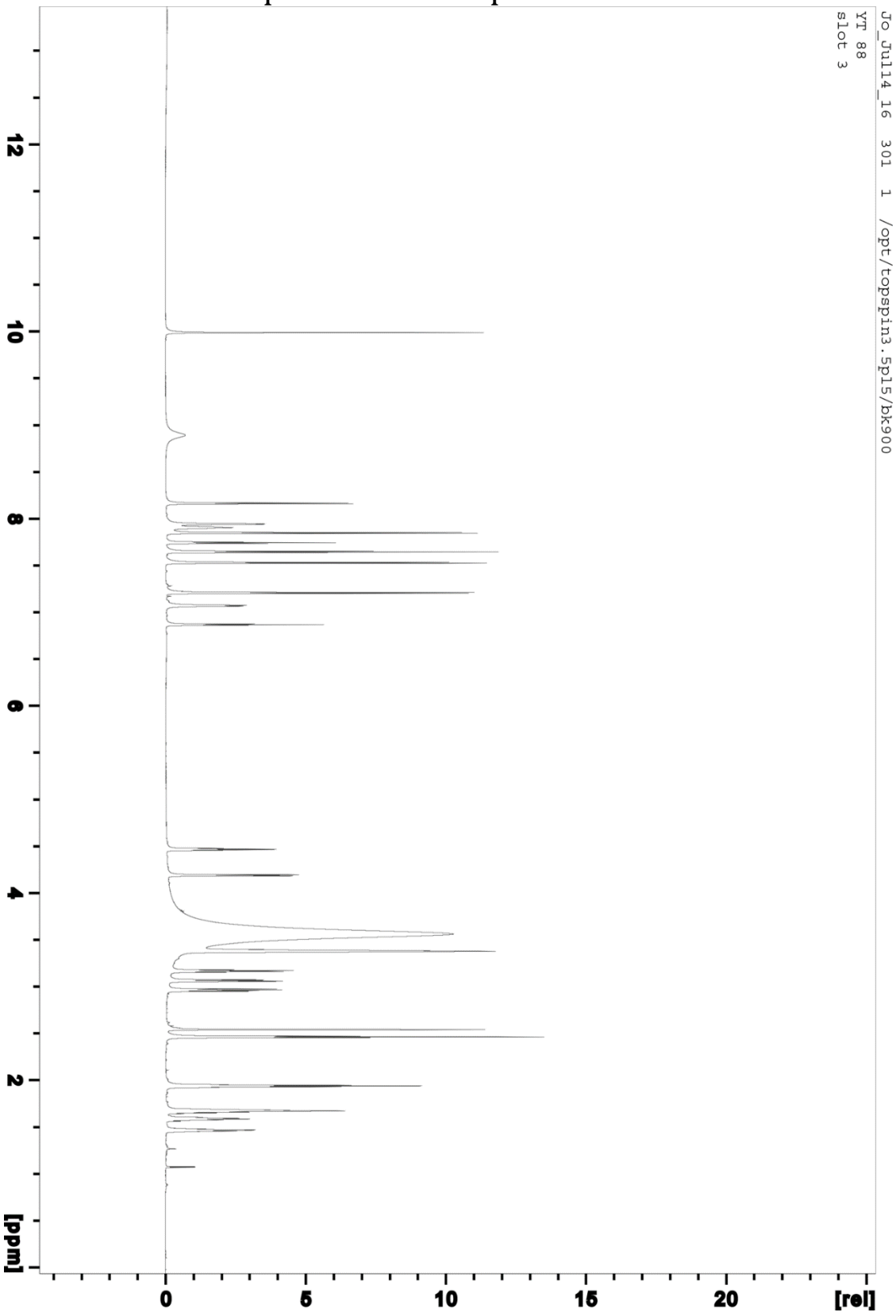
COPY of ^{1}H NMR spectrum of compound **29**



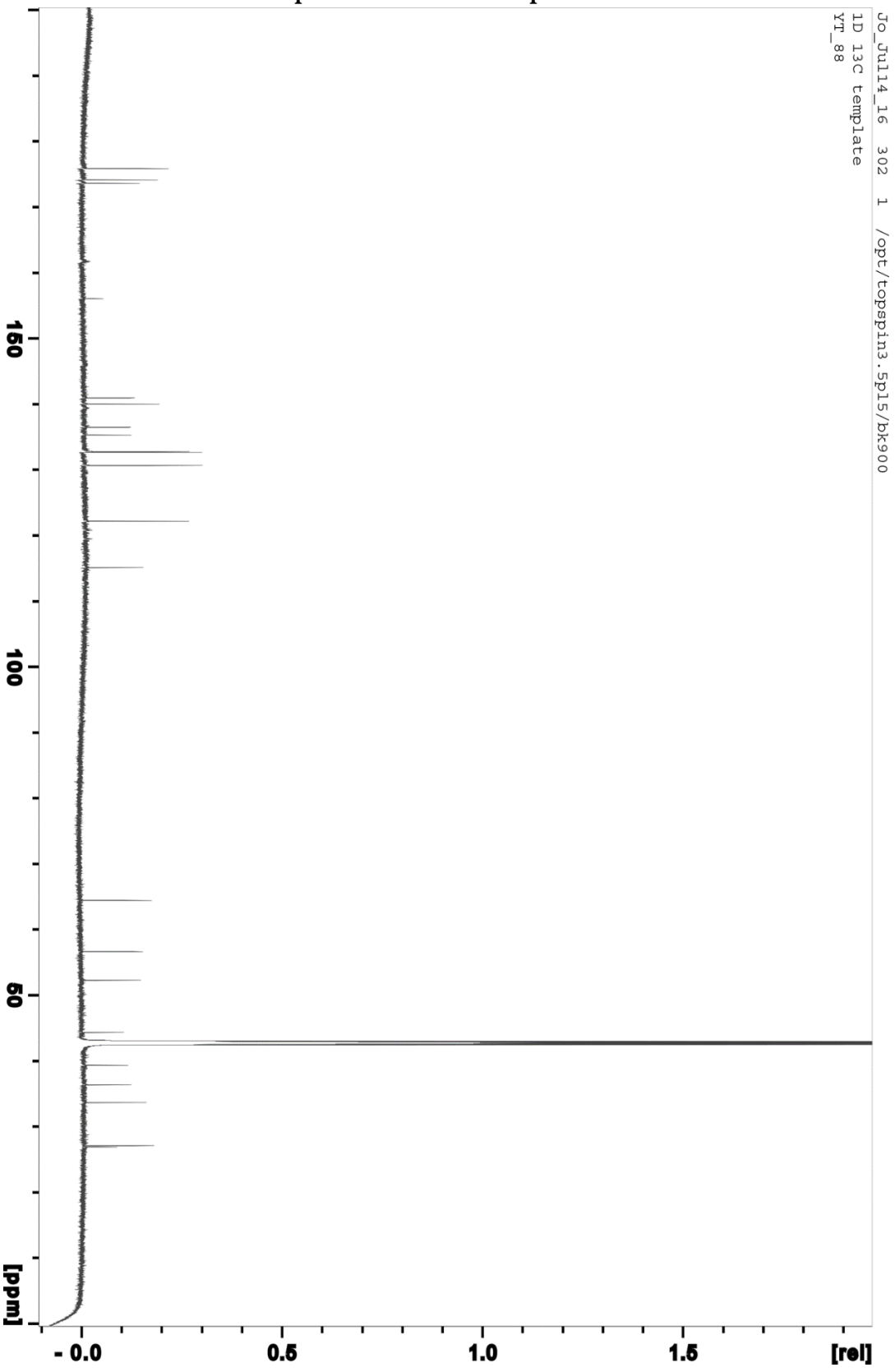
COPY of ^{13}C NMR spectrum of compound **29**



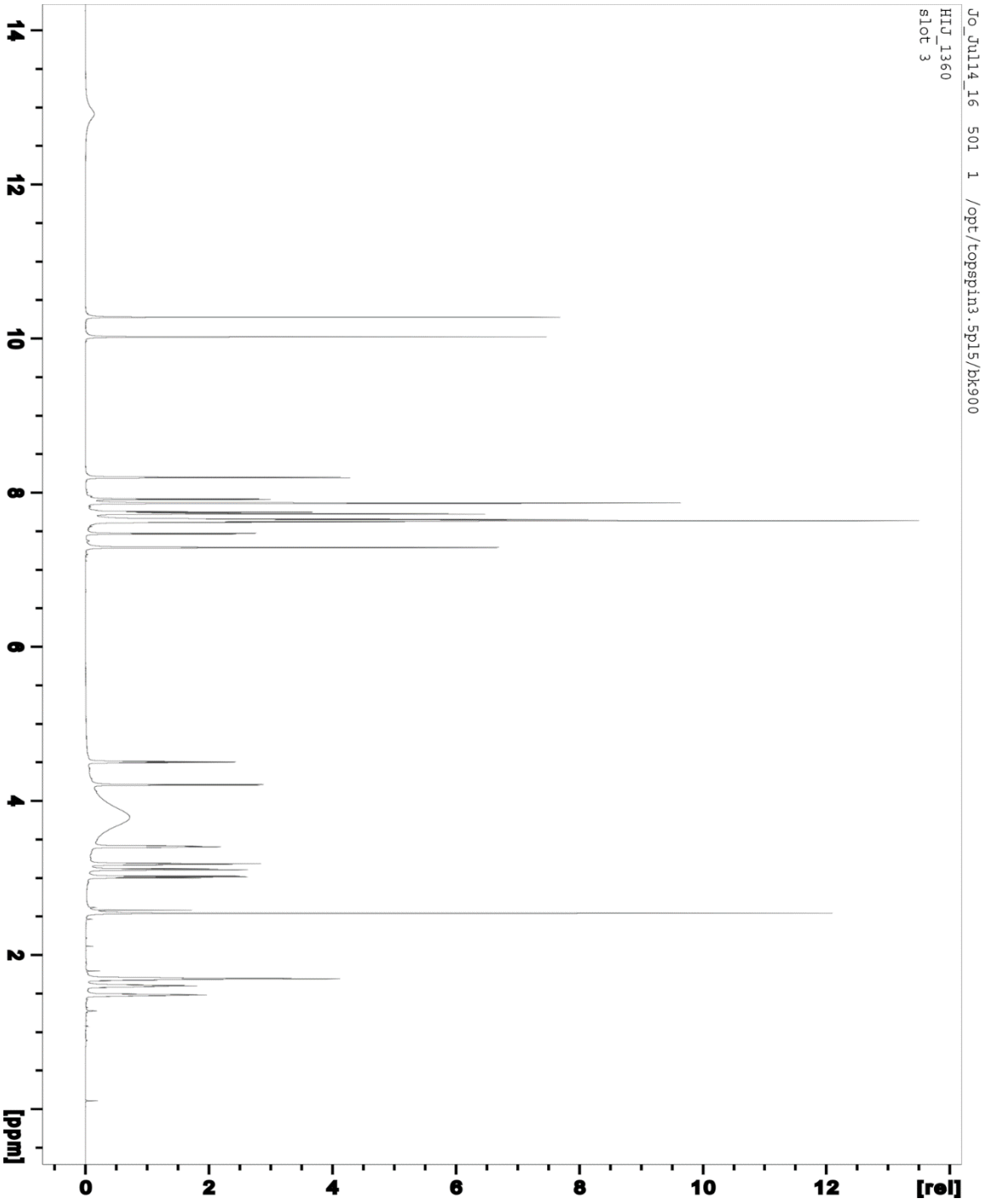
COPY of ^{1}H NMR spectrum of compound **33**



COPY of 13C NMR spectrum of compound 33



COPY of ^{1}H NMR spectrum of compound **37**



COPY of ^{13}C NMR spectrum of compound **37**

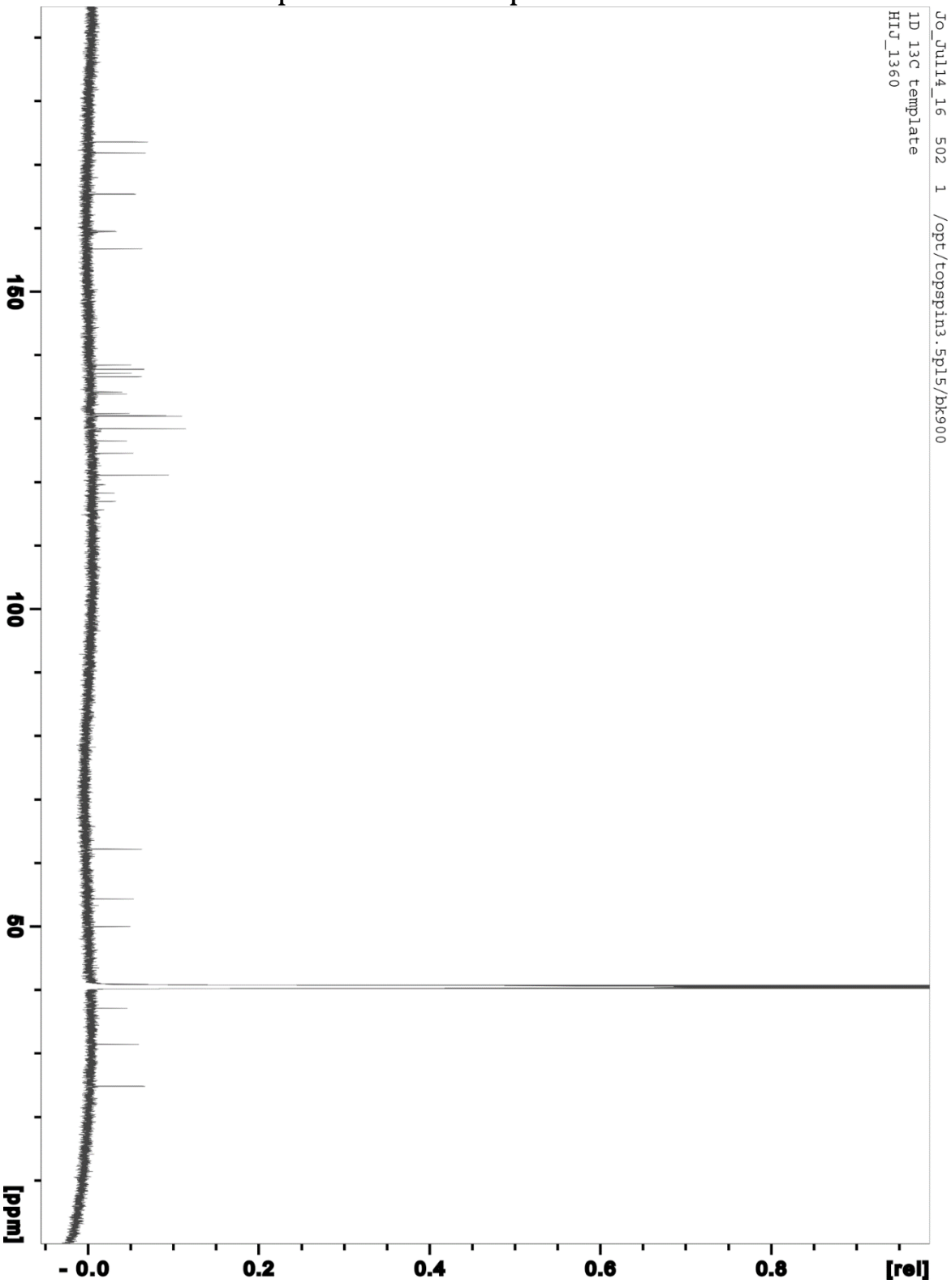


Figure S1. IC_{50} curve of c6, 18, 19, 29, 33, 37 against αvβ1 integrin

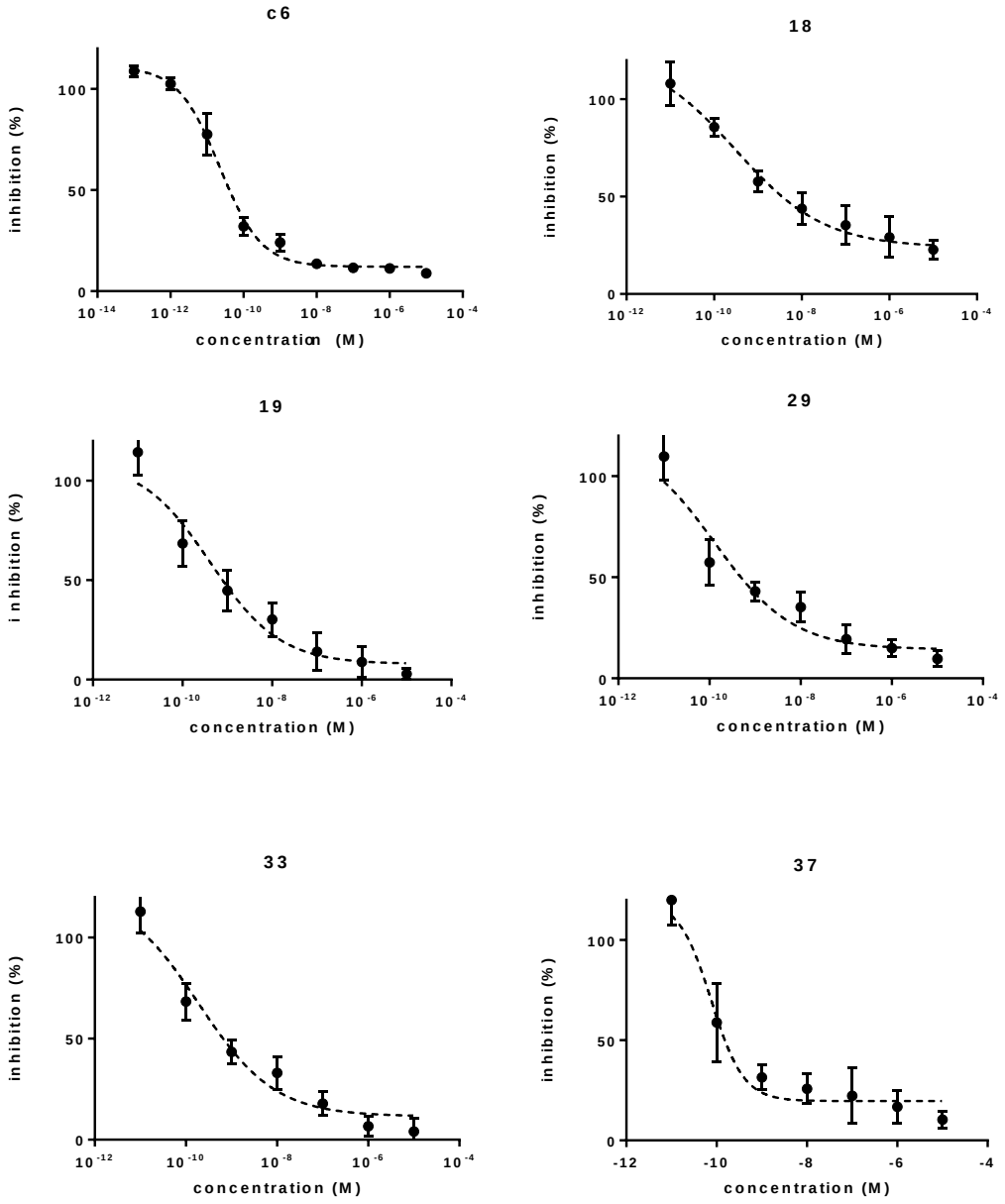
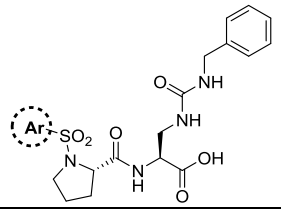
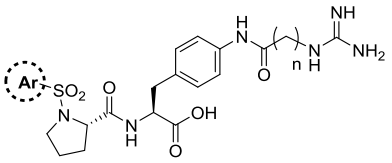
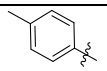
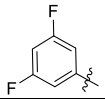
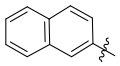
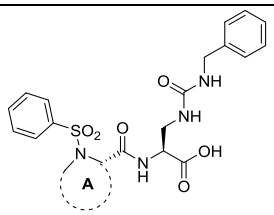
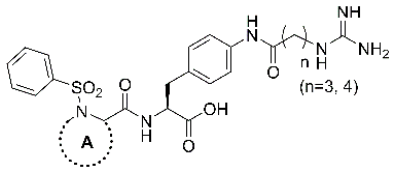
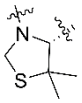


Table S4. pIC_{50} data of c6, 18, 19, 29, 33, 37 for RGD integrins

	IC50 (nM)				
	αvβ1	αvβ3	αvβ5	αvβ8	α5β1
18	9.5±0.7	5.4±0.3	6.8±0.5	5.7±0.4	5.5±0.2
19	9.6±0.6	5.7±0.3	5.8±0.4	5.6±0.3	5.5±0.2
29	9.5±0.2	<5	6.7±0.2	<5	<5
33	9.7±0.3	<5	<5	6.4±0.2	<5
37	10.2±0.3	6.8±0.2	<5	7.4±0.2	7.2±0.2

Table S5. IC_{50} comparison between selected α2β1 and αvβ1 integrin inhibitors

	α2β1 inhibitor[2,3] (Human platelet adhesion assay)	αvβ1 inhibitor
Structure		
Ar =	IC_{50} (nM)[2]	IC_{50} (nM)
	13	0.63 (n=4)
	36	630 (n=3)
	35	40 (n=4)
	50	2500 (n=4)
Structure		
	IC_{50} (nM)[3]	IC_{50} (nM)
	67	0.63 (n=4)
	60	800
	17	16
	12	2000

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3. Miller, M. W.; Basra, S.; Kulp, D. W.; Billings, P. C.; Choi, S.; Beavers, M. P.; McCarty, O. J.; Zou, Z.; Kahn, M. L.; Bennett, J. S.; DeGrado, W. F. Small-molecule inhibitors of integrin alpha2beta1 that prevent pathological thrombus formation via an allosteric mechanism. *Proc. Natl. Acad. Sci. U. S. A.* **2009,** *106,* 719-724.