

Supplementary Information

Application of a bivalent “click” approach to target tyrosyl-DNA phosphodiesterase 1 (TDP1)

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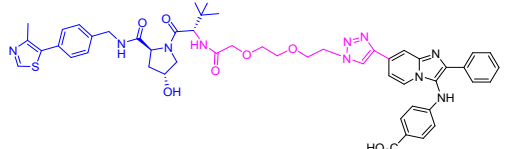
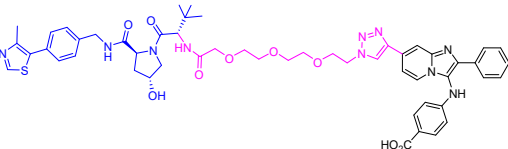
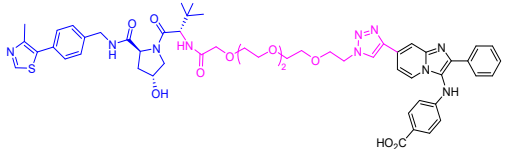
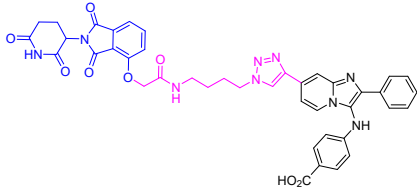
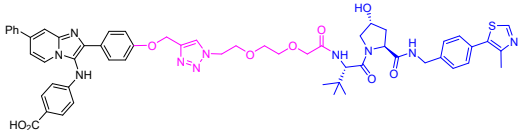
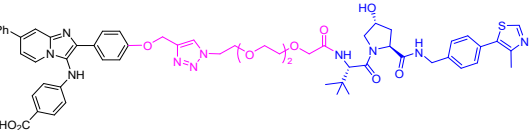
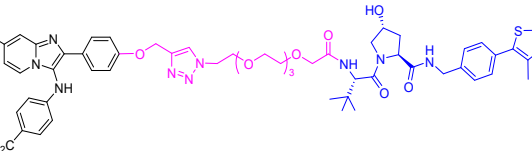
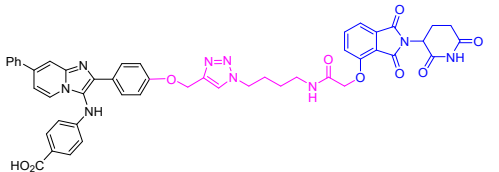
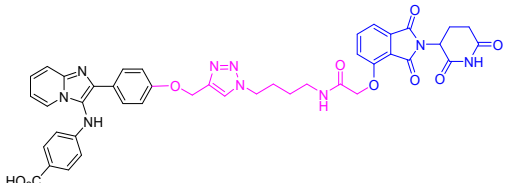
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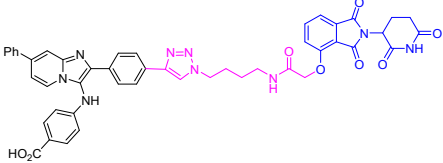
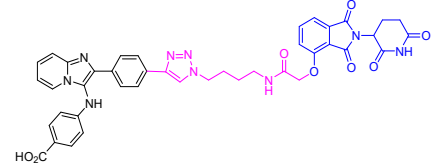
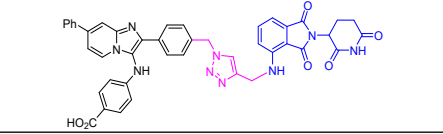
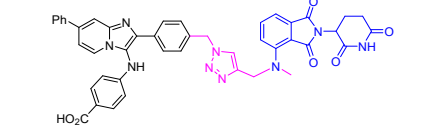
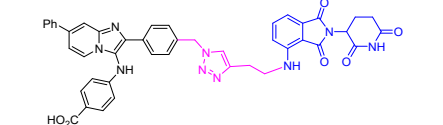
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I. Table S1. Structures of bivalent analogs **4a-4d** and **5a-5j**.

Compound	Structures ⁱ
4a	
4b	
4c	
4d	
5a	
5b	
5c	
5d	
5e	

5f	
5g	
5h	
5i	
5j	

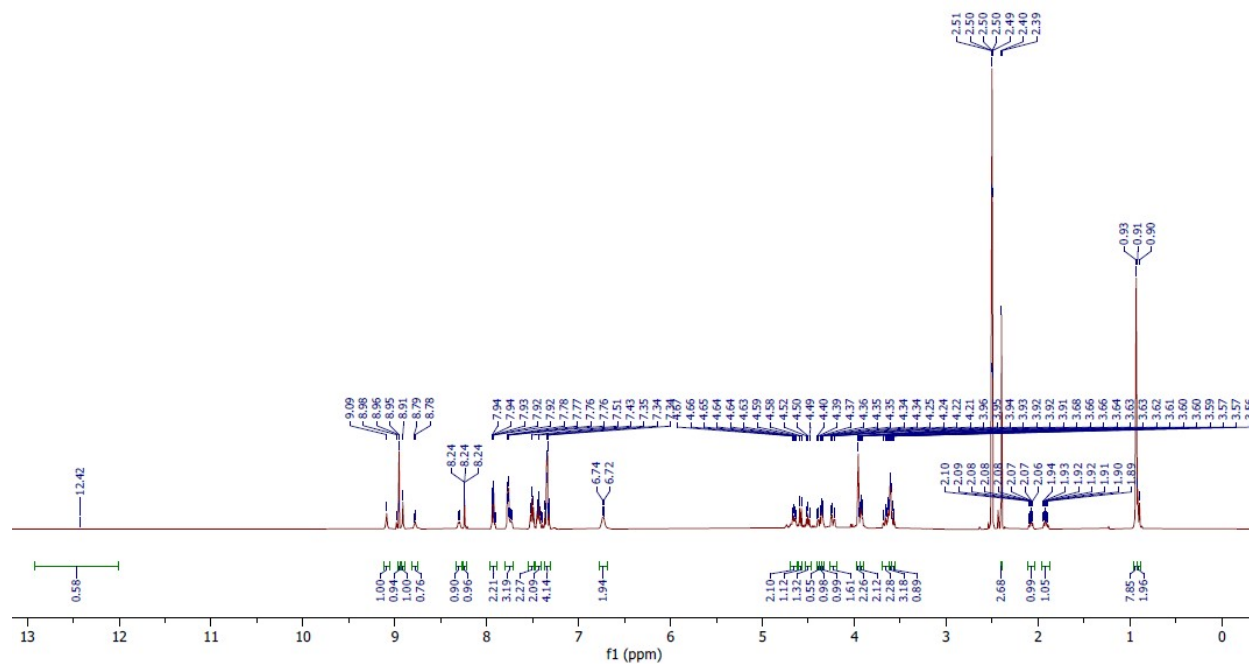
Note: TDP1 binders are in black, ligase recruiters are in blue and linker components are in purple.

Table S2. Cytotoxicity of compounds in cellular assays using MCF7 and HCT116 cell lines.

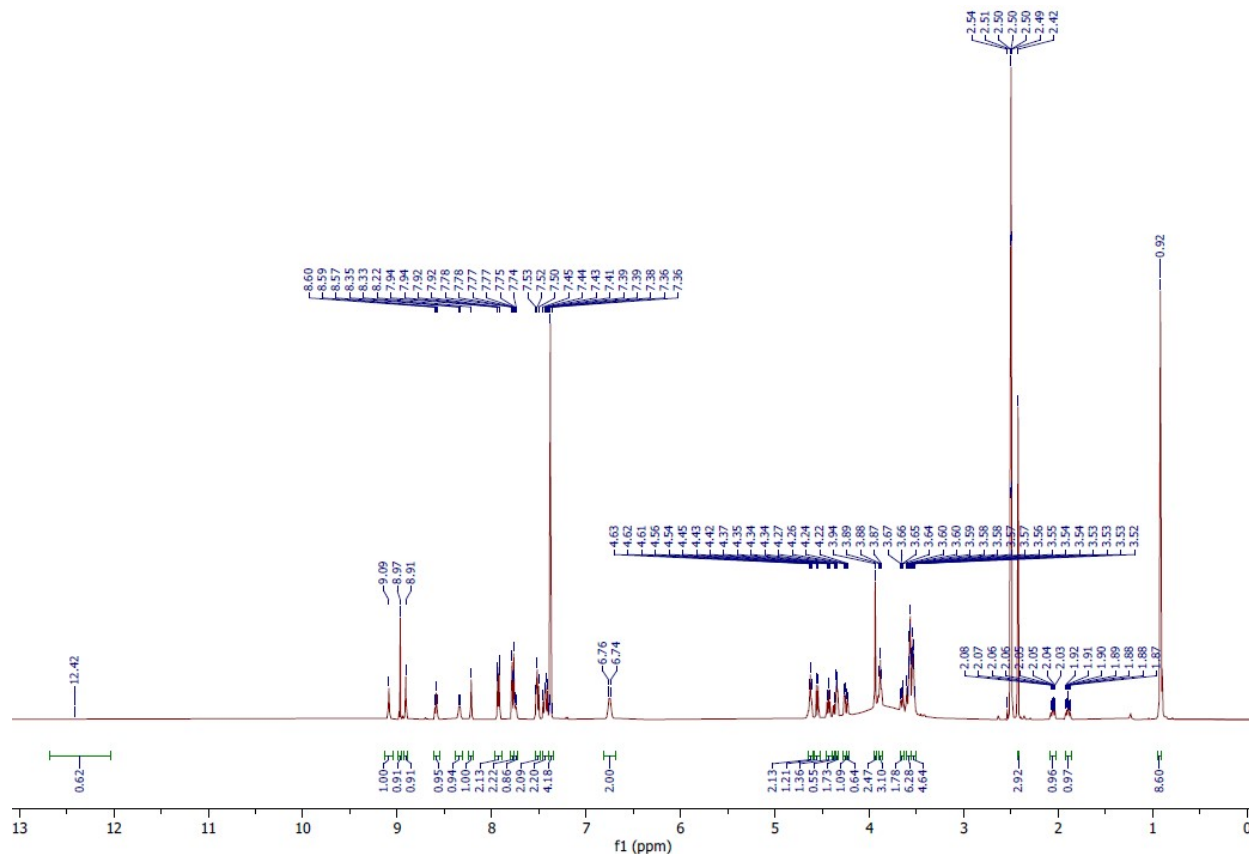
Compound	CC₅₀ (μM, MCF7)	CC₅₀ (μM, HCT116)
1b	110	50
5a	74	152
5h	105	193
5i	129	190
5j	108	151

III. ¹H NMR Spectra of Compounds 4a – c and 5a – j.

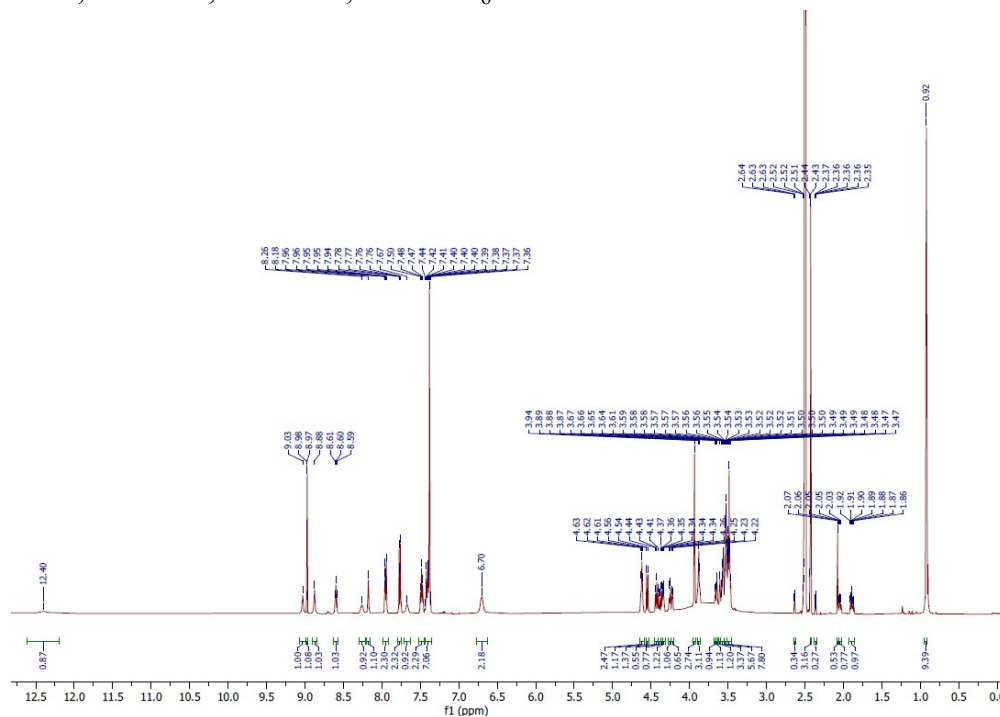
Compound 4a, ¹H NMR, 500 MHz, DMSO-d₆



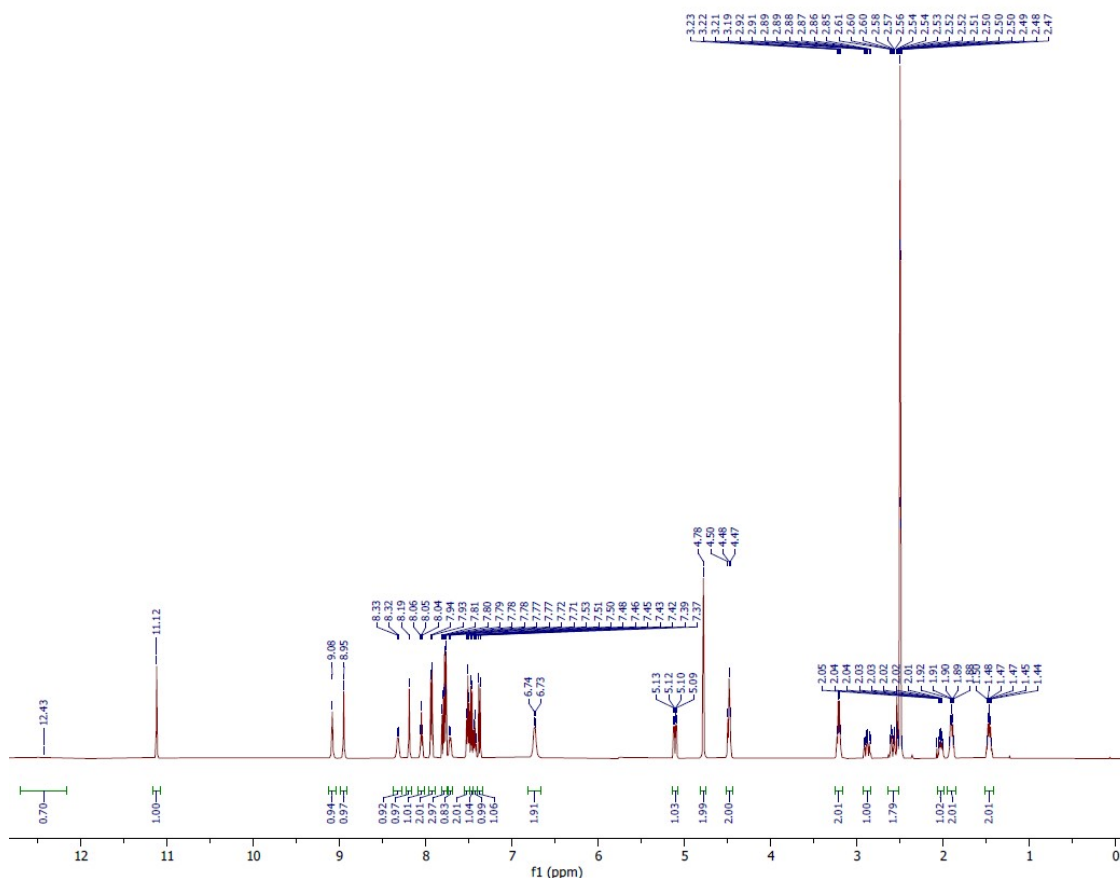
Compound 4b, ¹H NMR, 500 MHz, DMSO-d₆



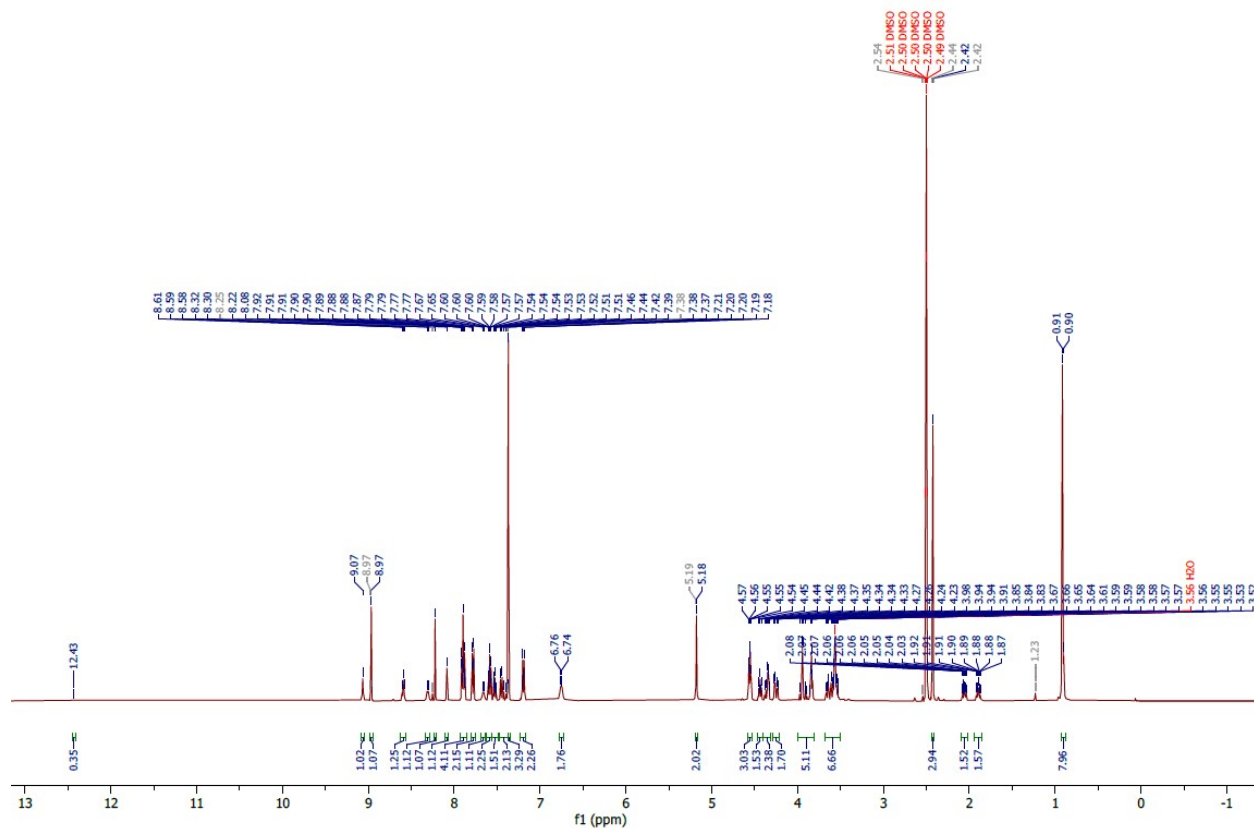
Compound 4c, ¹H NMR, 500 MHz, DMSO-d₆



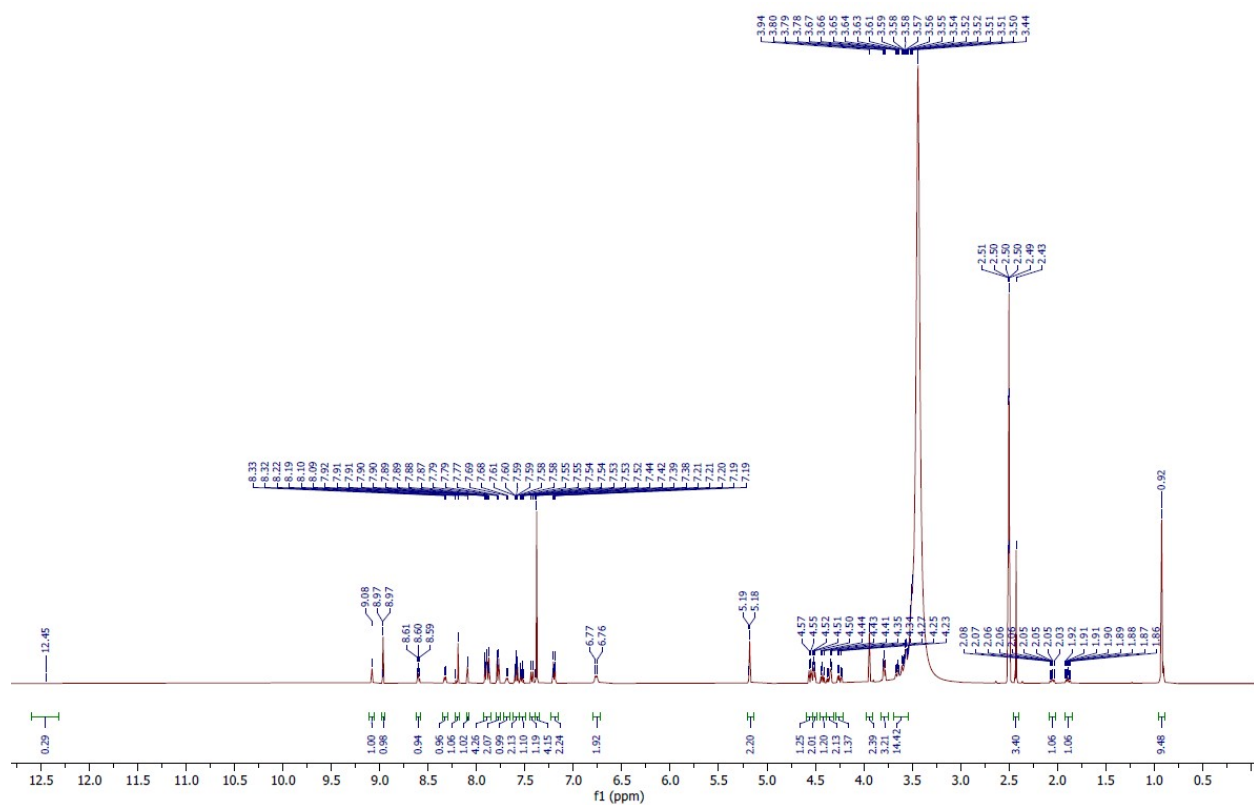
Compound 4d, ¹H NMR, 500 MHz, DMSO-d₆



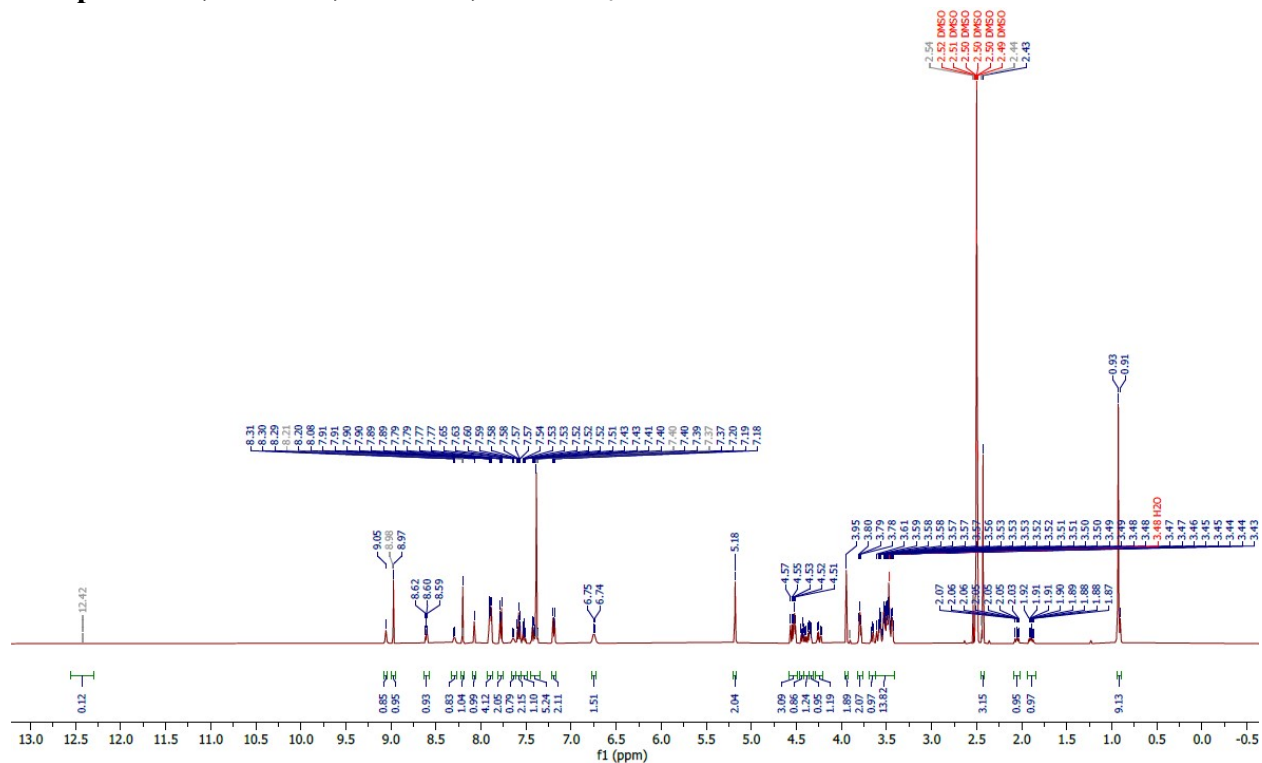
Compound 5a, ¹H NMR, 500 MHz, DMSO-d₆



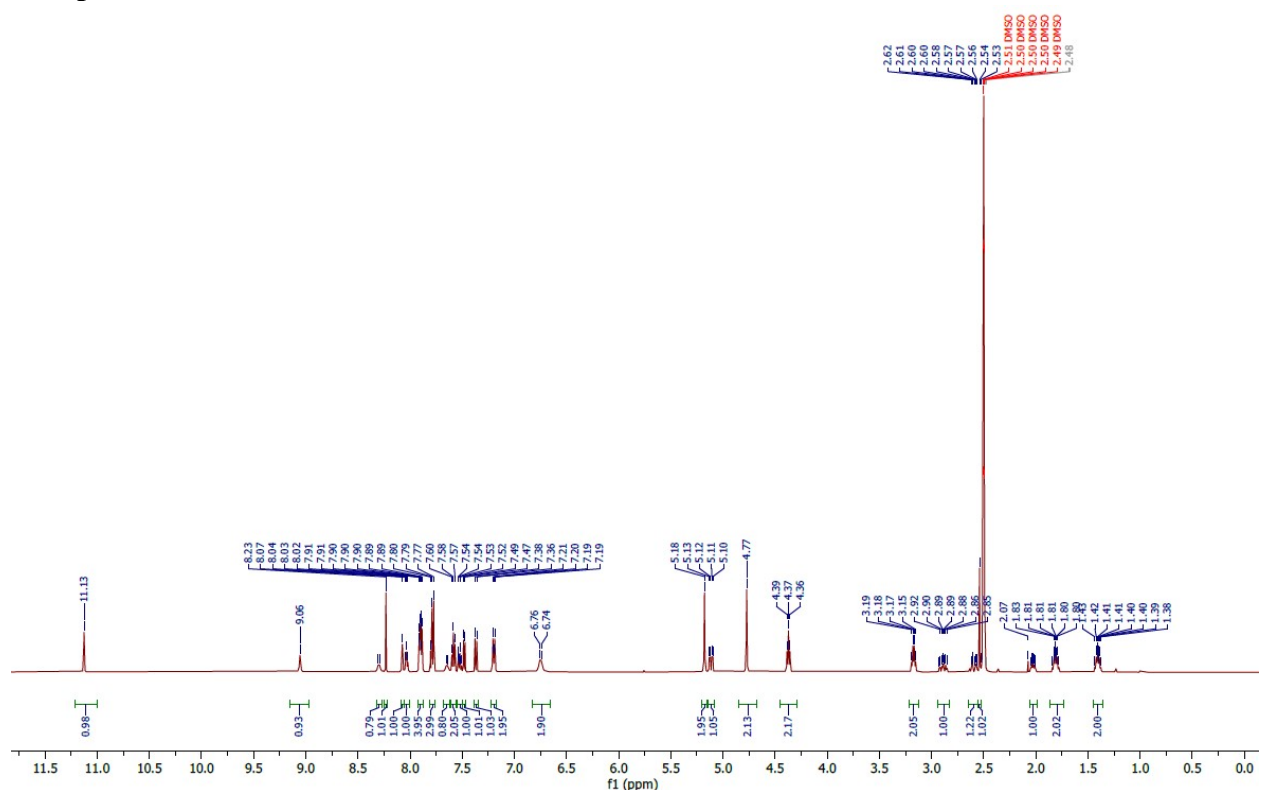
Compound 5b, ¹H NMR, 500 MHz, DMSO-d₆



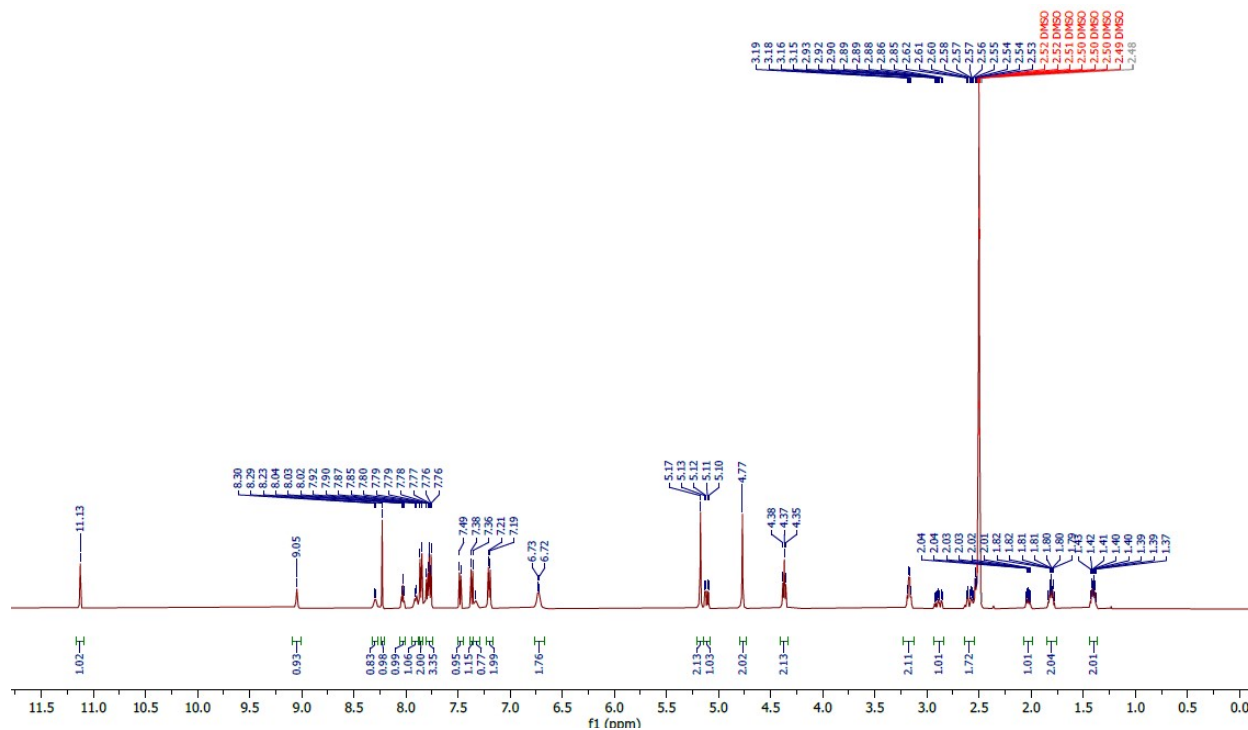
Compound 5c, ¹H NMR, 500 MHz, DMSO-d₆



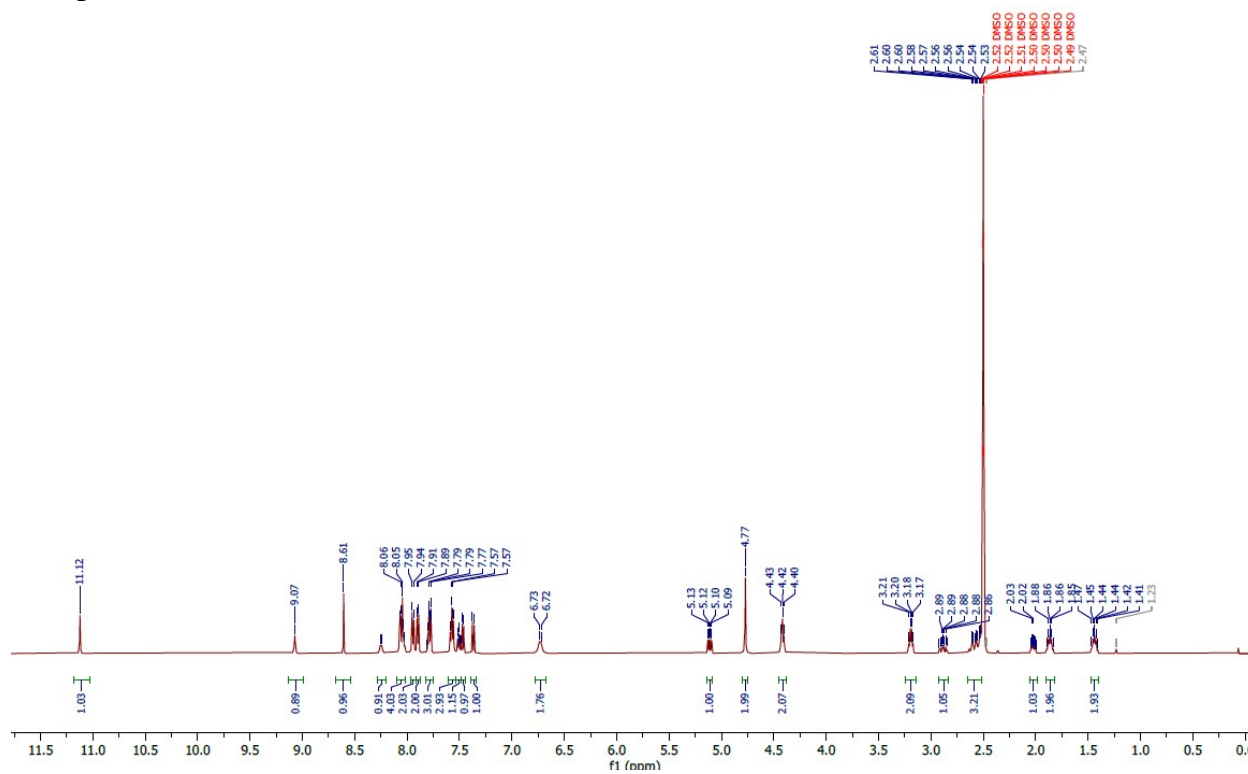
Compound 5d, ¹H NMR, 500 MHz, DMSO-d₆



Compound 5e, ¹H NMR, 500 MHz, DMSO-d₆



Compound 5f, ¹H NMR, 500 MHz, DMSO-d₆

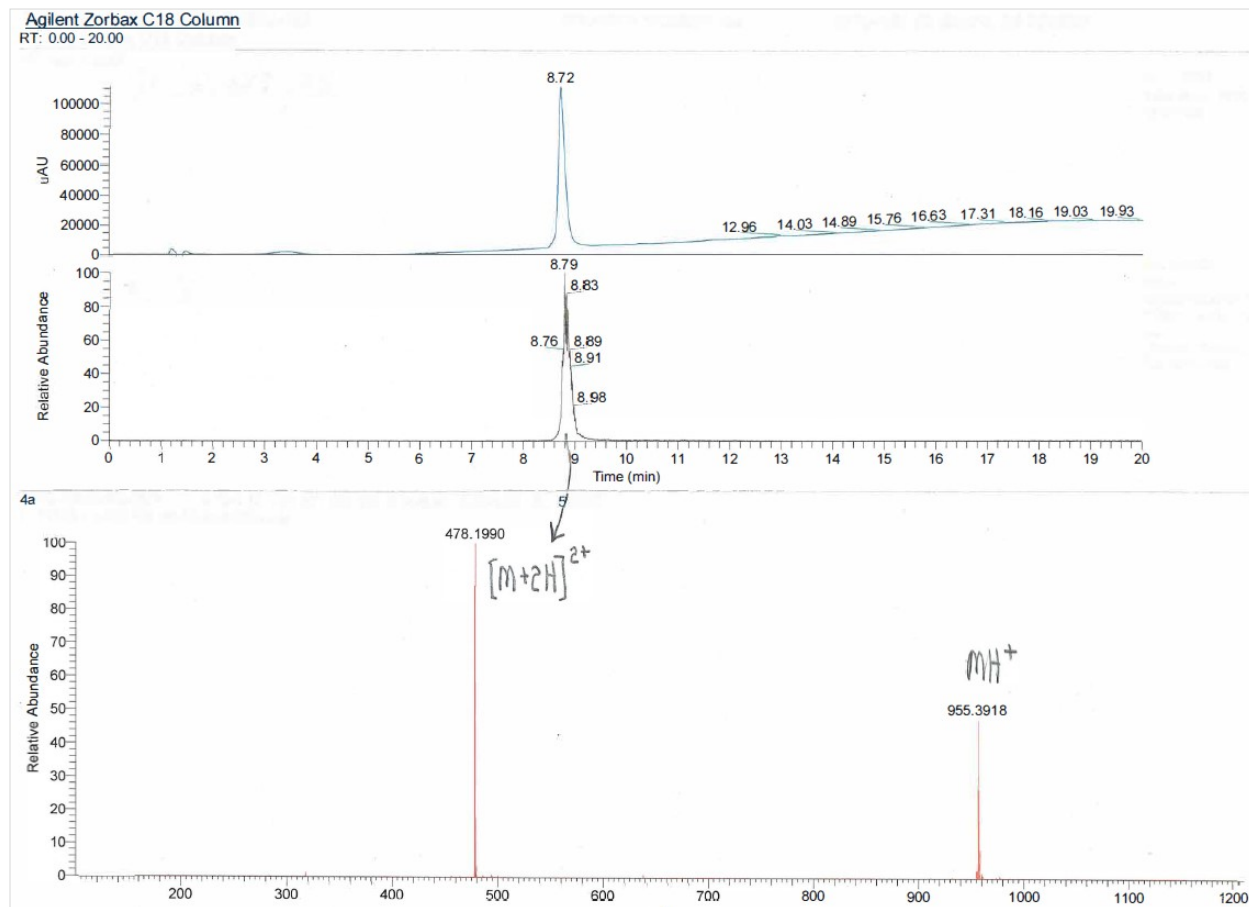


IV. HRMS of Compounds 4a – d and 5a – j.

Compound 4a

HRMS (ESI) m/z calcd. for $C_{50}H_{55}N_{10}O_8S$ (MH^+), 955.3920; found, 955.3918. ($\Delta = 0.2$ ppm).

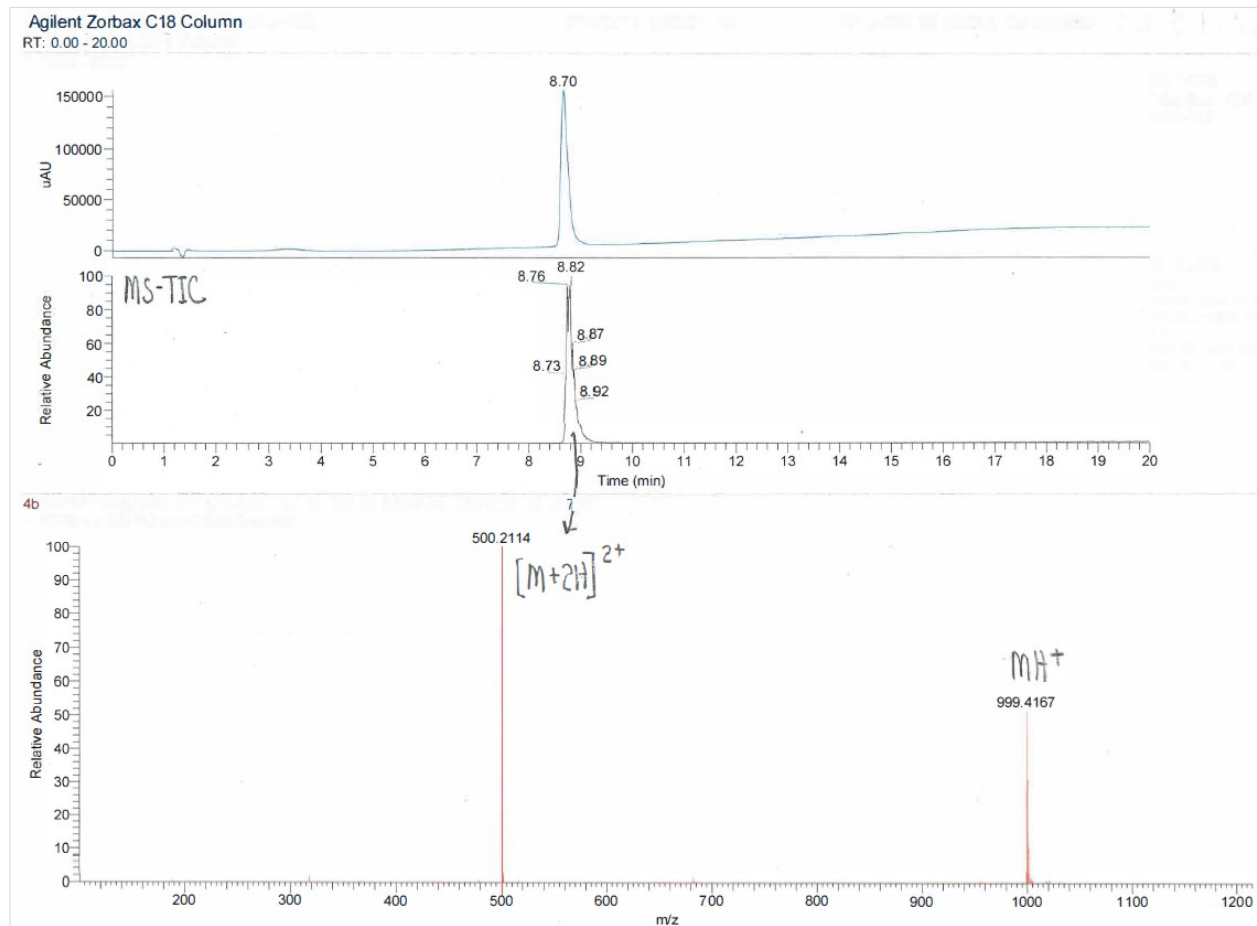
HRMS (ESI) m/z calcd. for $C_{50}H_{56}N_{10}O_8S$ (MH_2) $^{2+}$, 478.1996; found, 478.1990. ($\Delta = 1.3$ ppm).



Compound 4b

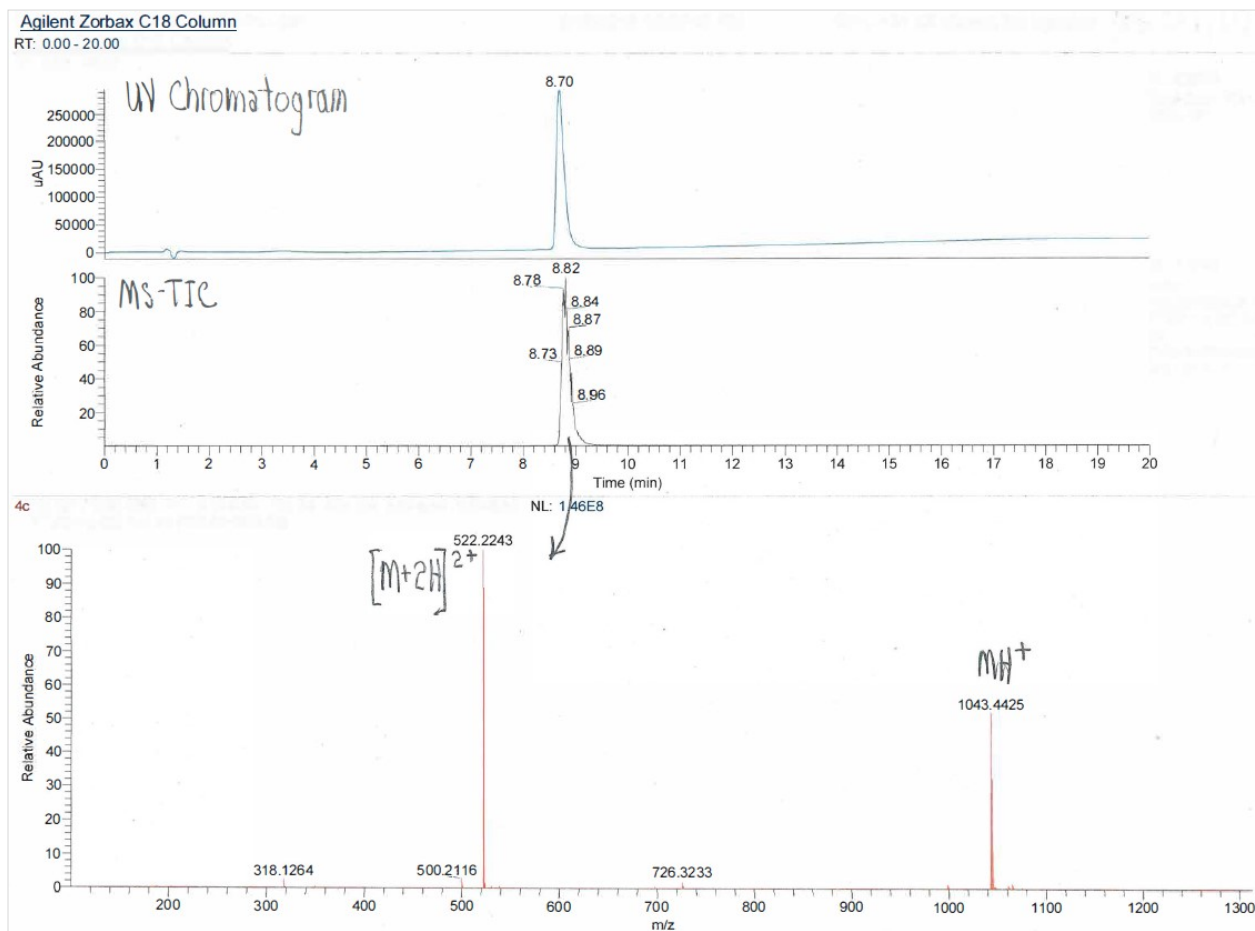
HRMS (ESI) m/z calcd. for $C_{52}H_{59}N_{10}O_9S$ (MH^+), 999.4182; found, 999.4167. ($\Delta = 1.5$ ppm).

HRMS (ESI) m/z calcd. for $C_{52}H_{60}N_{10}O_9S$ (MH_2) $^{2+}$, 500.2127; found, 500.2114. ($\Delta = 2.6$ ppm).



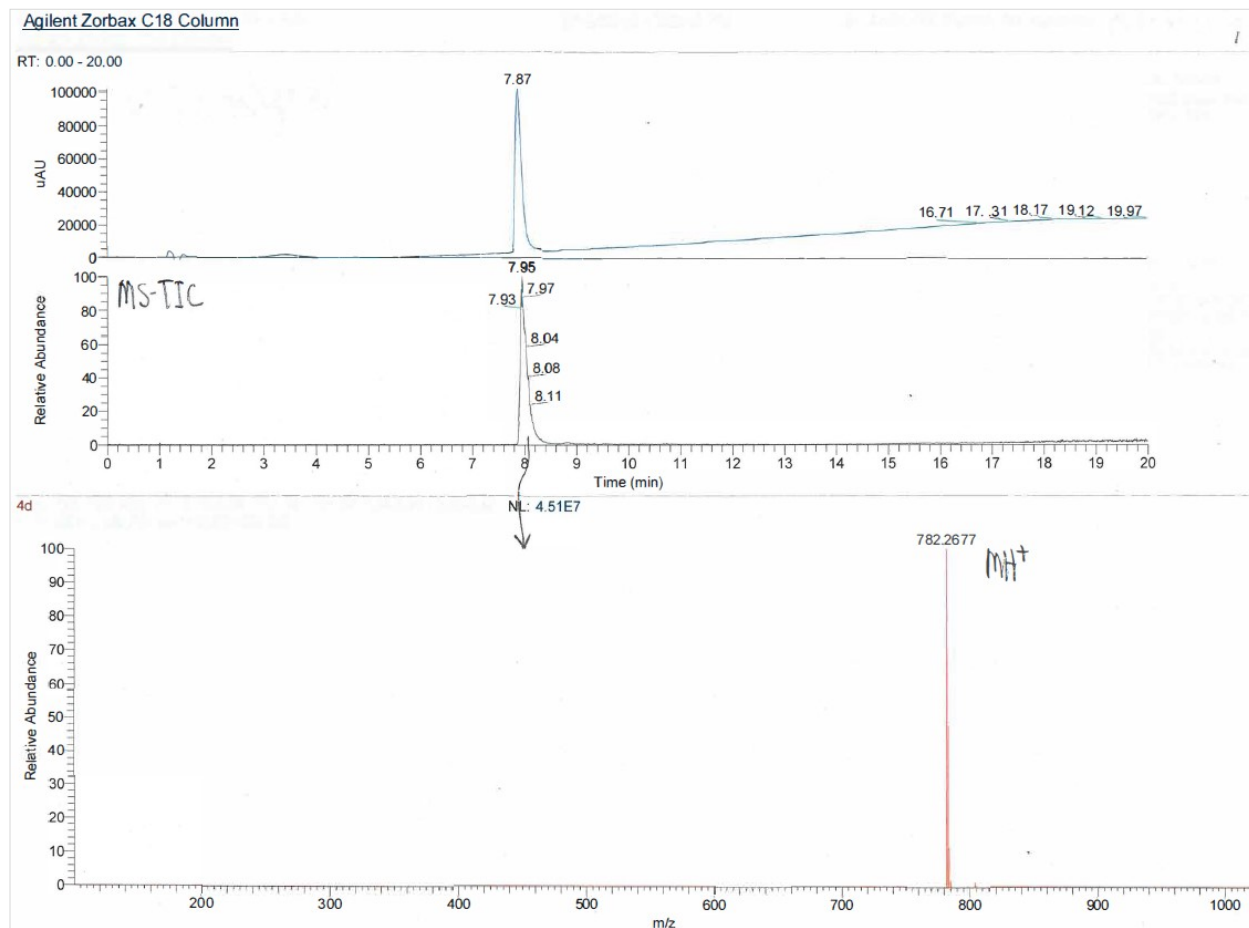
Compound 4c

HRMS (ESI) m/z calcd. for $C_{54}H_{63}N_{10}O_{10}S$ (MH^+), 1043.4444; found, 1043.4425. ($\Delta = 1.8$ ppm).
HRMS (ESI) m/z calcd. for $C_{54}H_{64}N_{10}O_{10}S$ (MH_2) $^{2+}$, 522.2258; found, 522.2243. ($\Delta = 2.9$ ppm).



Compound 4d

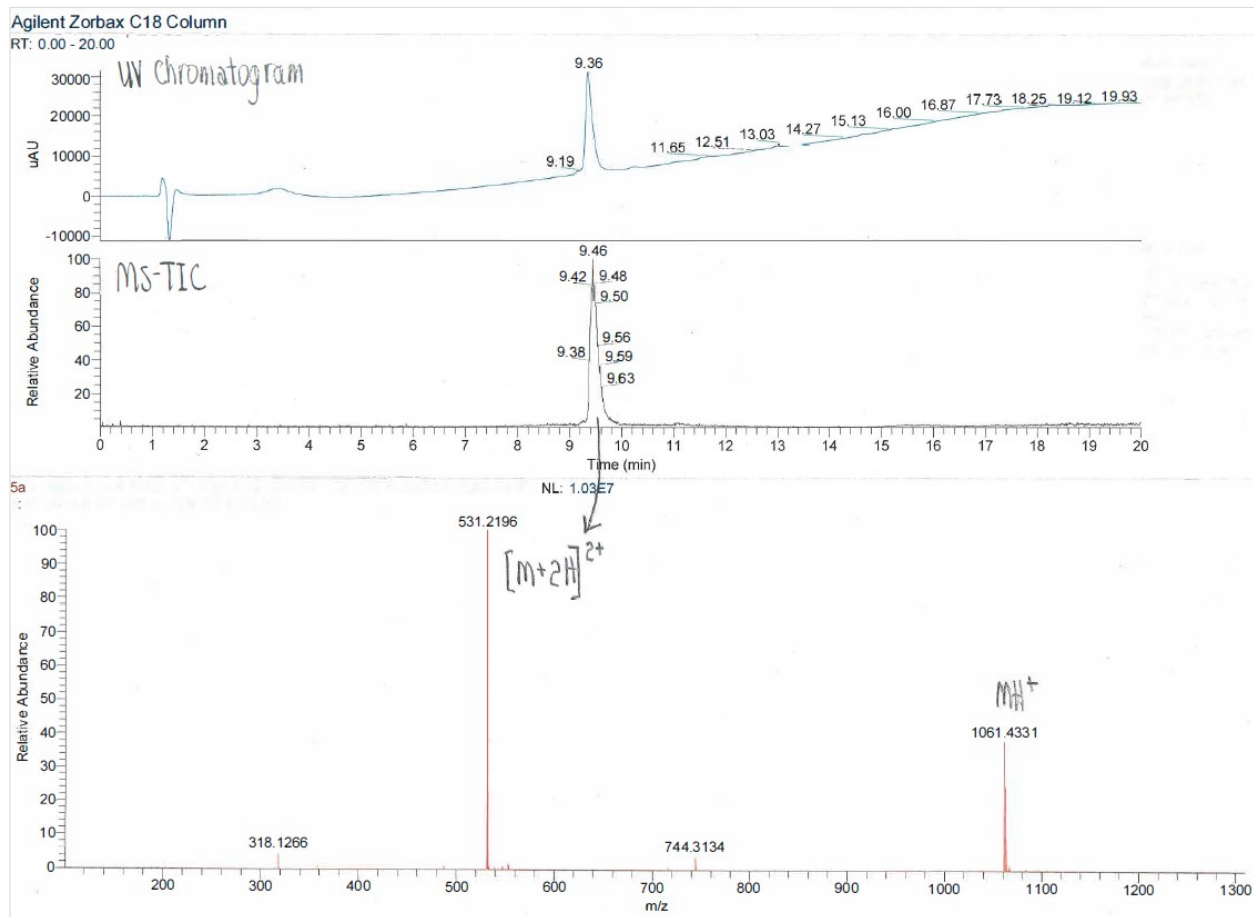
HRMS (ESI) m/z calcd. for $C_{41}H_{36}N_9O_8$ (MH^+), 782.2681; found, 782.2677. ($\Delta = 0.5$ ppm).



Compound 5a

HRMS (ESI) m/z calcd. for $C_{57}H_{61}N_{10}O_9S$ (MH^+), 1061.4338; found, 1061.4331. ($\Delta = 0.7$ ppm).

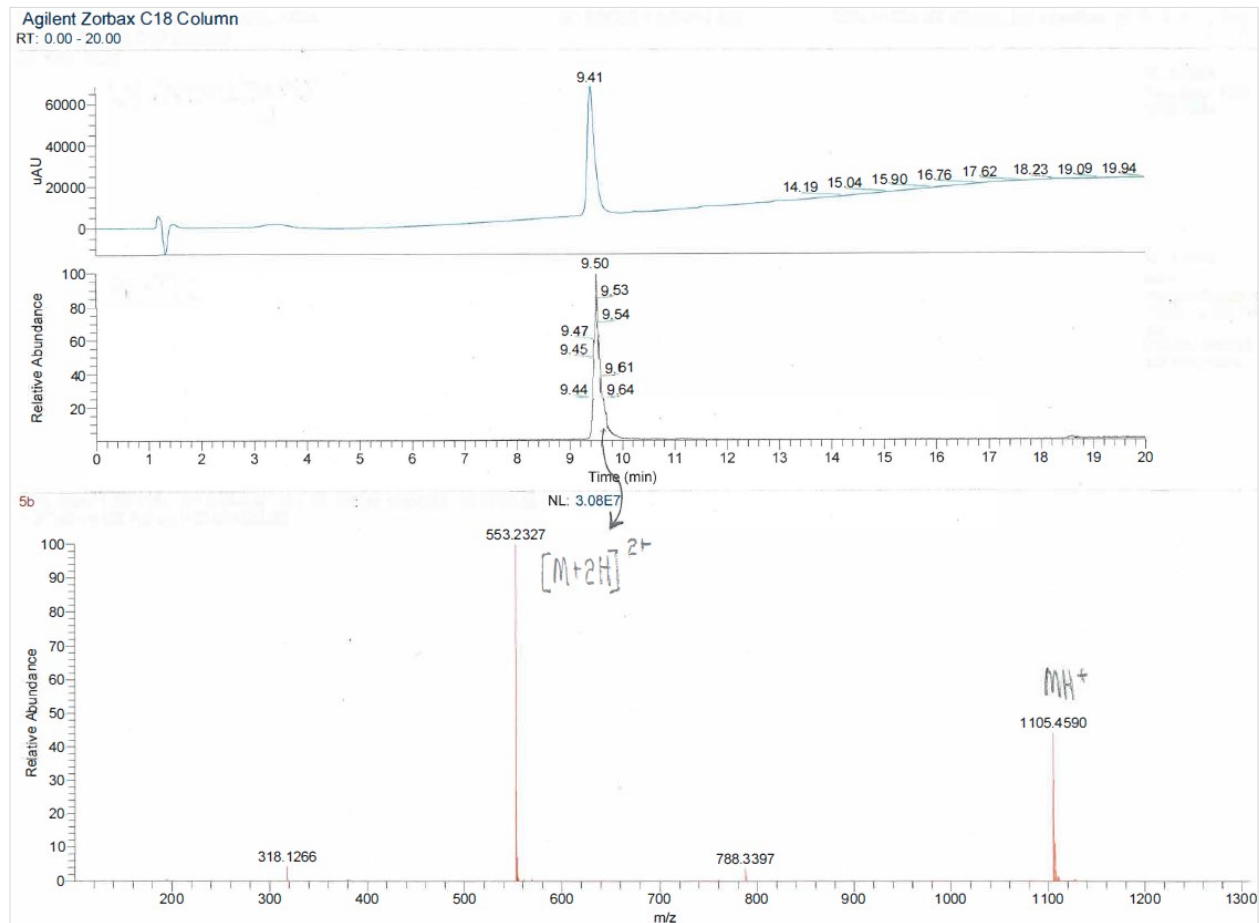
HRMS (ESI) m/z calcd. for $C_{57}H_{62}N_{10}O_9S$ (MH_2)²⁺, 531.2205; found, 531.2196. ($\Delta = 1.7$ ppm).



Compound 5b

HRMS (ESI) m/z calcd. for $C_{59}H_{65}N_{10}O_{10}S$ (MH^+), 1105.4600; found, 1105.2327. ($\Delta = 0.9$ ppm).

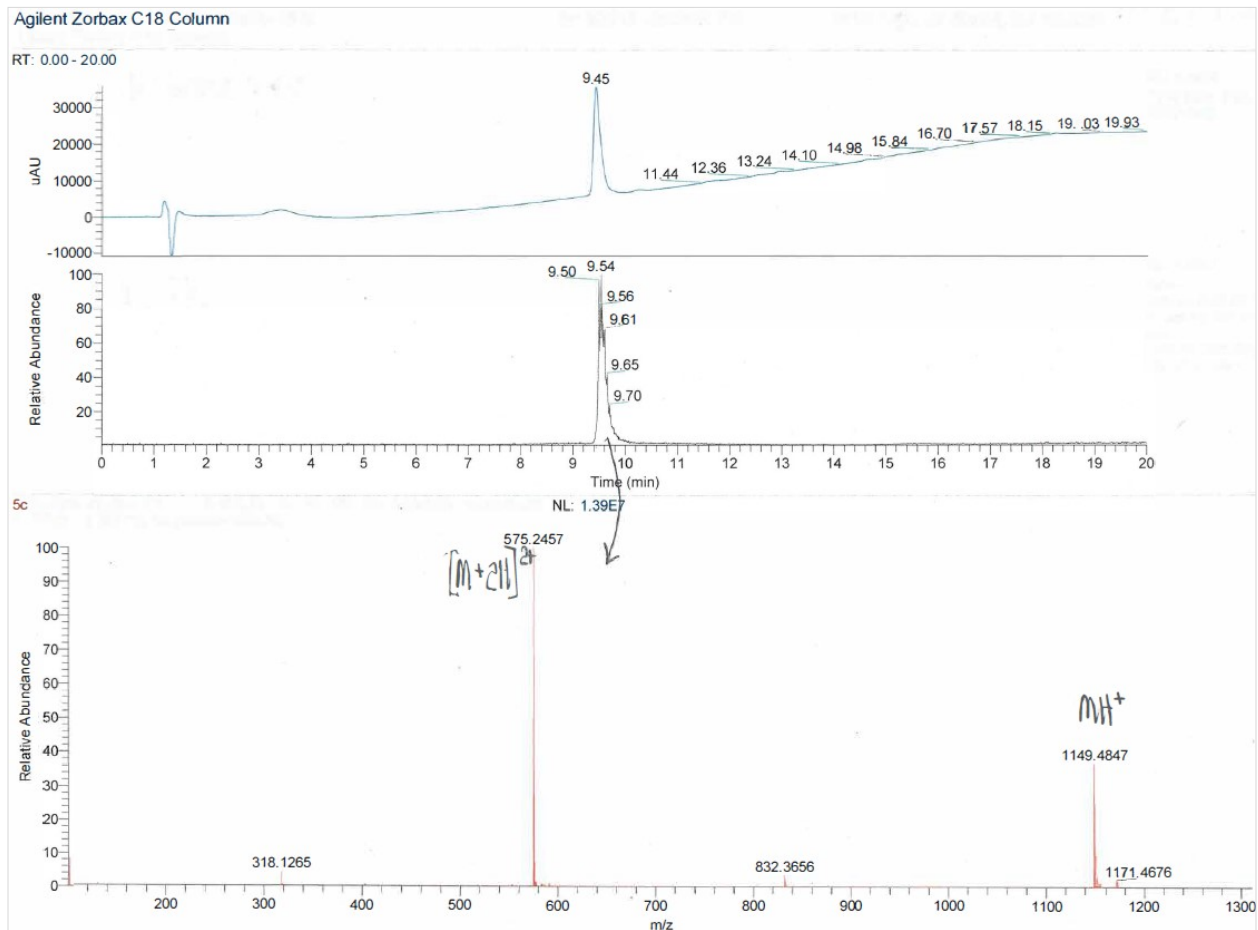
HRMS (ESI) m/z calcd. for $C_{59}H_{66}N_{10}O_{10}S$ (MH_2) $^{2+}$, 553.2337; found, 553.2327. ($\Delta = 1.8$ ppm).



Compound 5c

HRMS (ESI) m/z calcd. for $C_{61}H_{69}N_{10}O_{11}S$ (MH^+), 1149.4863; found, 1149.4847. ($\Delta = 1.4$ ppm).

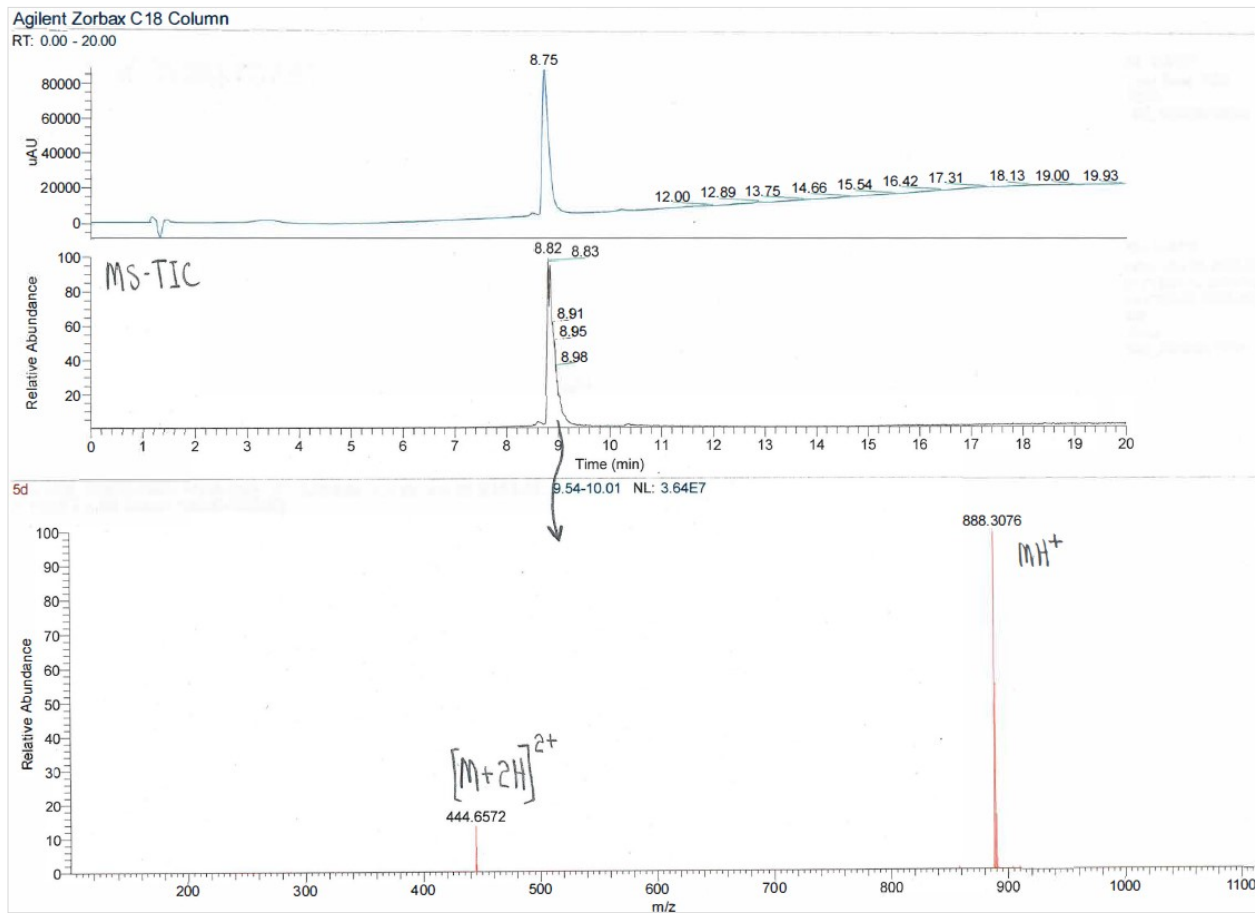
HRMS (ESI) m/z calcd. for $C_{61}H_{70}N_{10}O_{11}S$ (MH_2)²⁺, 575.2468; found, 575.2457. ($\Delta = 1.9$ ppm).



Compound 5d

HRMS (ESI) m/z calcd. for $C_{48}H_{42}N_9O_9$ (MH^+), 888.3100; found, 888.3076. ($\Delta = 2.7$ ppm).

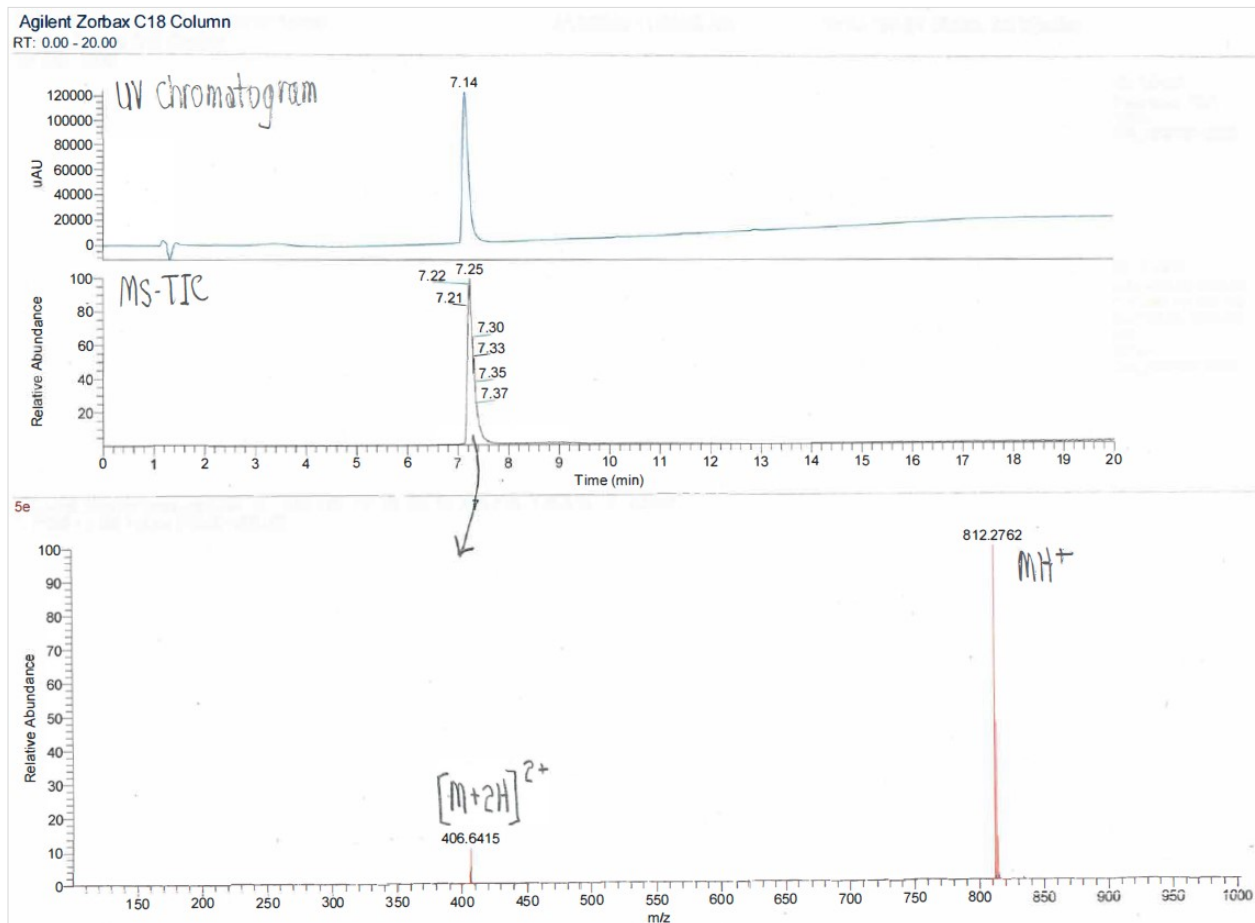
HRMS (ESI) m/z calcd. for $C_{48}H_{43}N_9O_9$ (MH_2) $^{2+}$, 444.6586; found, 444.6572. ($\Delta = 3.1$ ppm).



Compound 5e

HRMS (ESI) m/z calcd. for $C_{42}H_{38}N_9O_9$ (MH^+), 812.2787; found, 812.2762. ($\Delta = 3.1$ ppm).

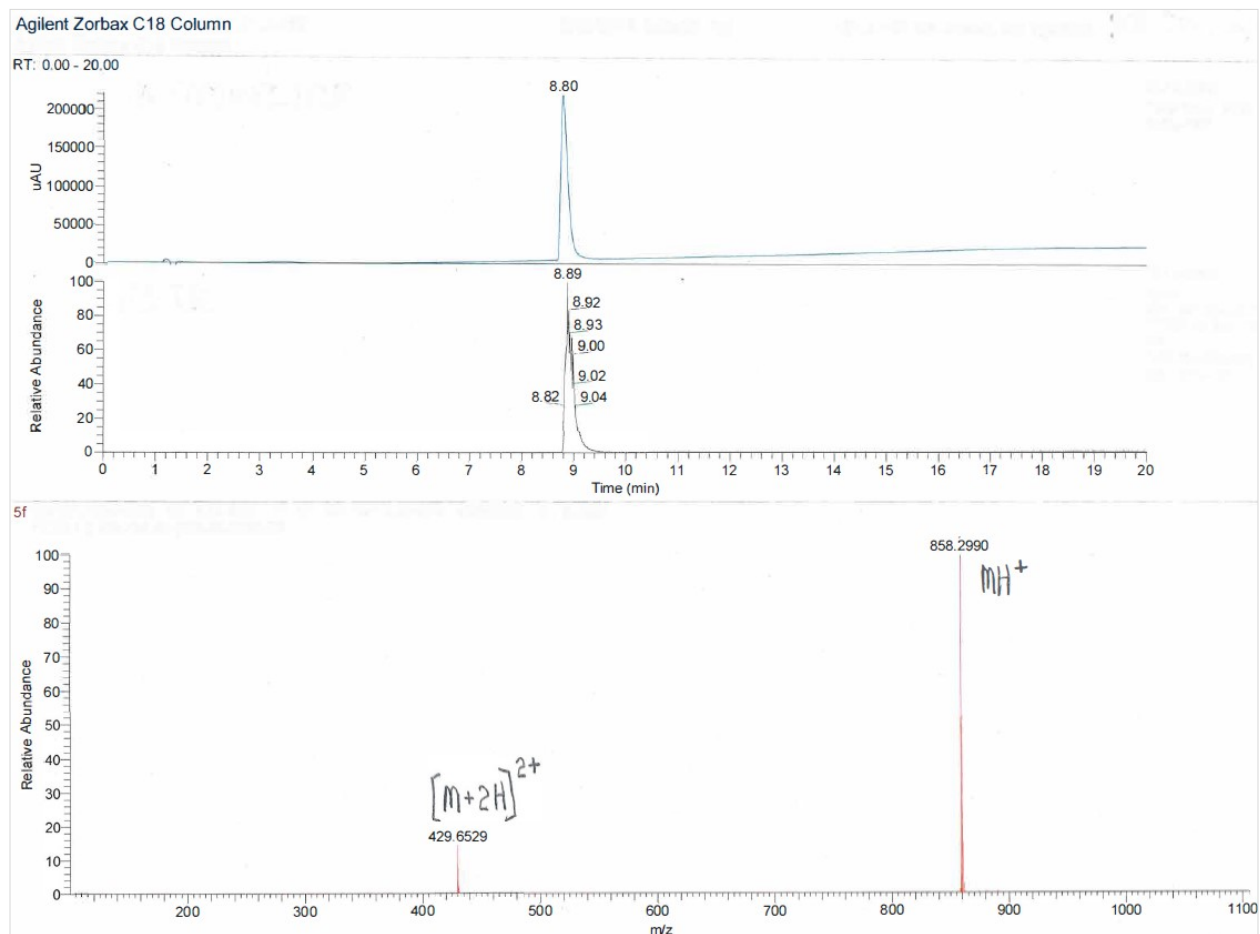
HRMS (ESI) m/z calcd. for $C_{42}H_{39}N_9O_9$ (MH_2) $^{2+}$, 406.6430; found, 406.6415. ($\Delta = 3.7$ ppm).



Compound 5f

HRMS (ESI) m/z calcd. for $C_{47}H_{40}N_9O_8$ (MH^+), 858.2994; found, 858.2990. ($\Delta = 0.5$ ppm).

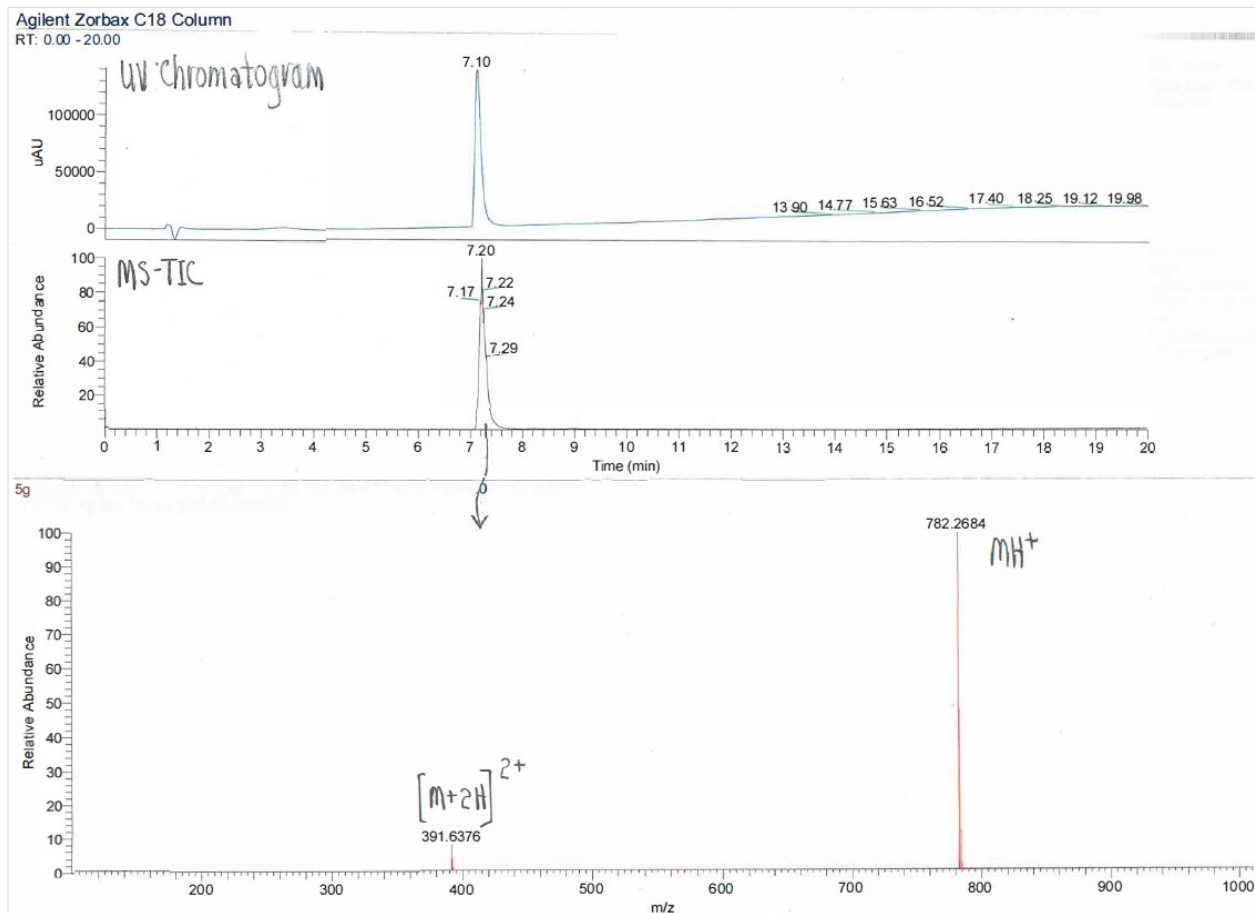
HRMS (ESI) m/z calcd. for $C_{47}H_{41}N_9O_8$ (MH_2^{2+}), 429.6534; found, 429.6529. ($\Delta = 1.2$ ppm).



Compound 5g

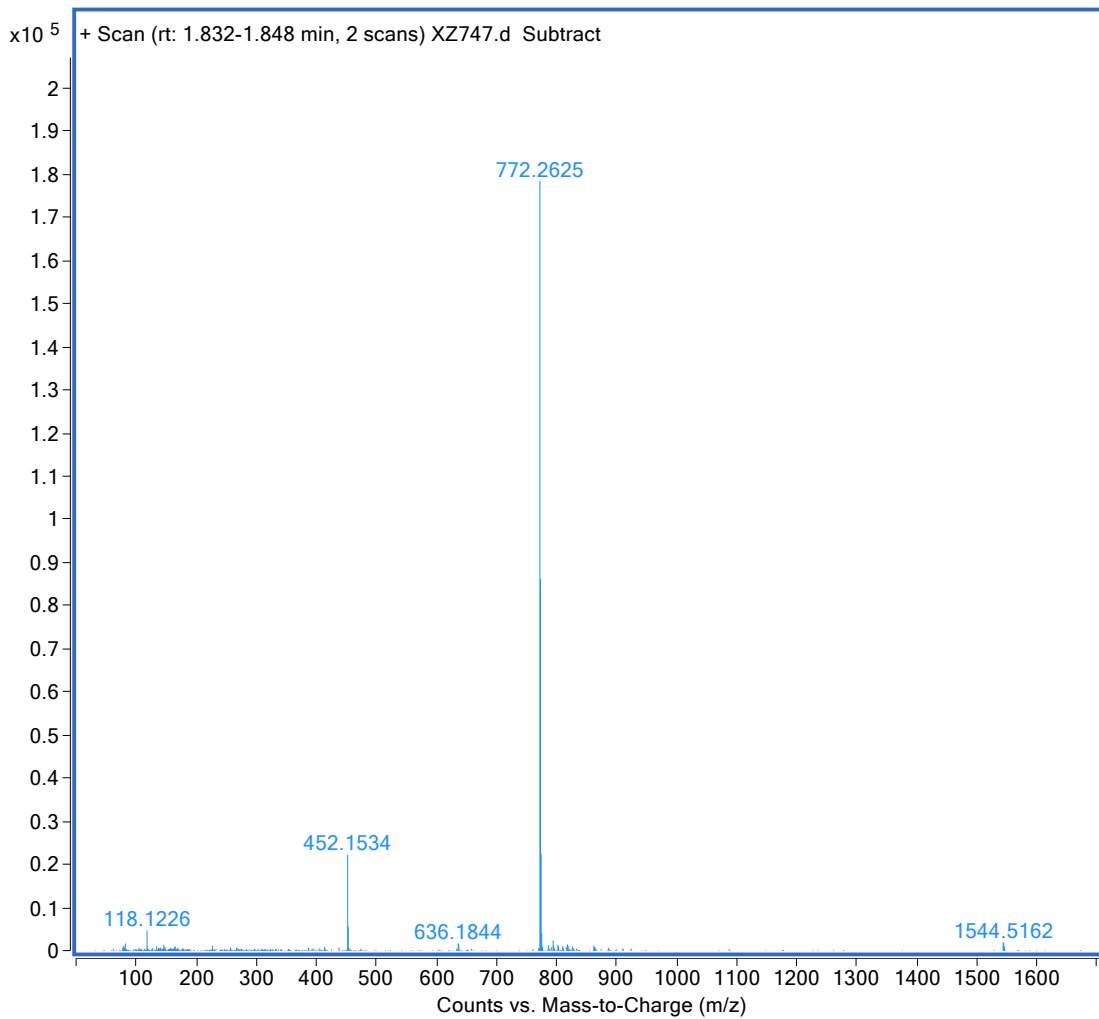
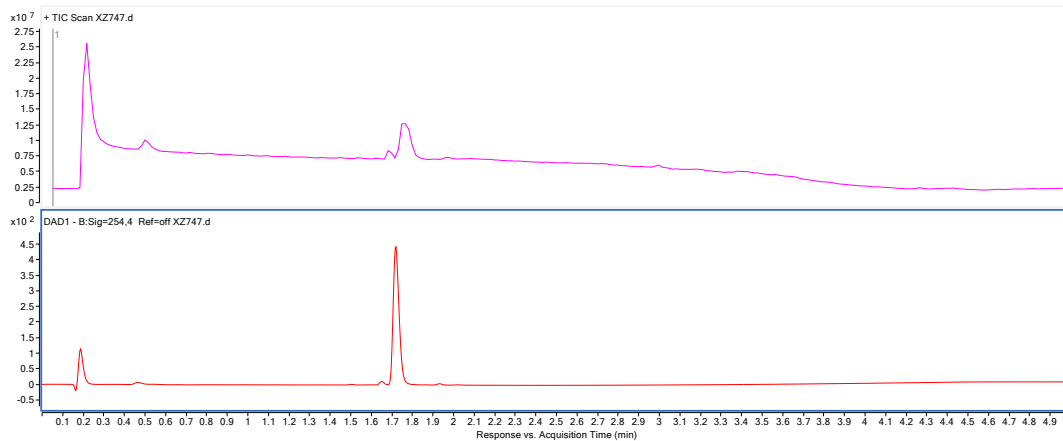
HRMS (ESI) m/z calcd. for $C_{41}H_{36}N_9O_8$ (MH^+), 782.2681; found, 782.2684. ($\Delta = 0.4$ ppm).

HRMS (ESI) m/z calcd. for $C_{41}H_{37}N_9O_9$ (MH_2) $^{2+}$, 391.6377; found, 391.6376. ($\Delta = 0.3$ ppm).



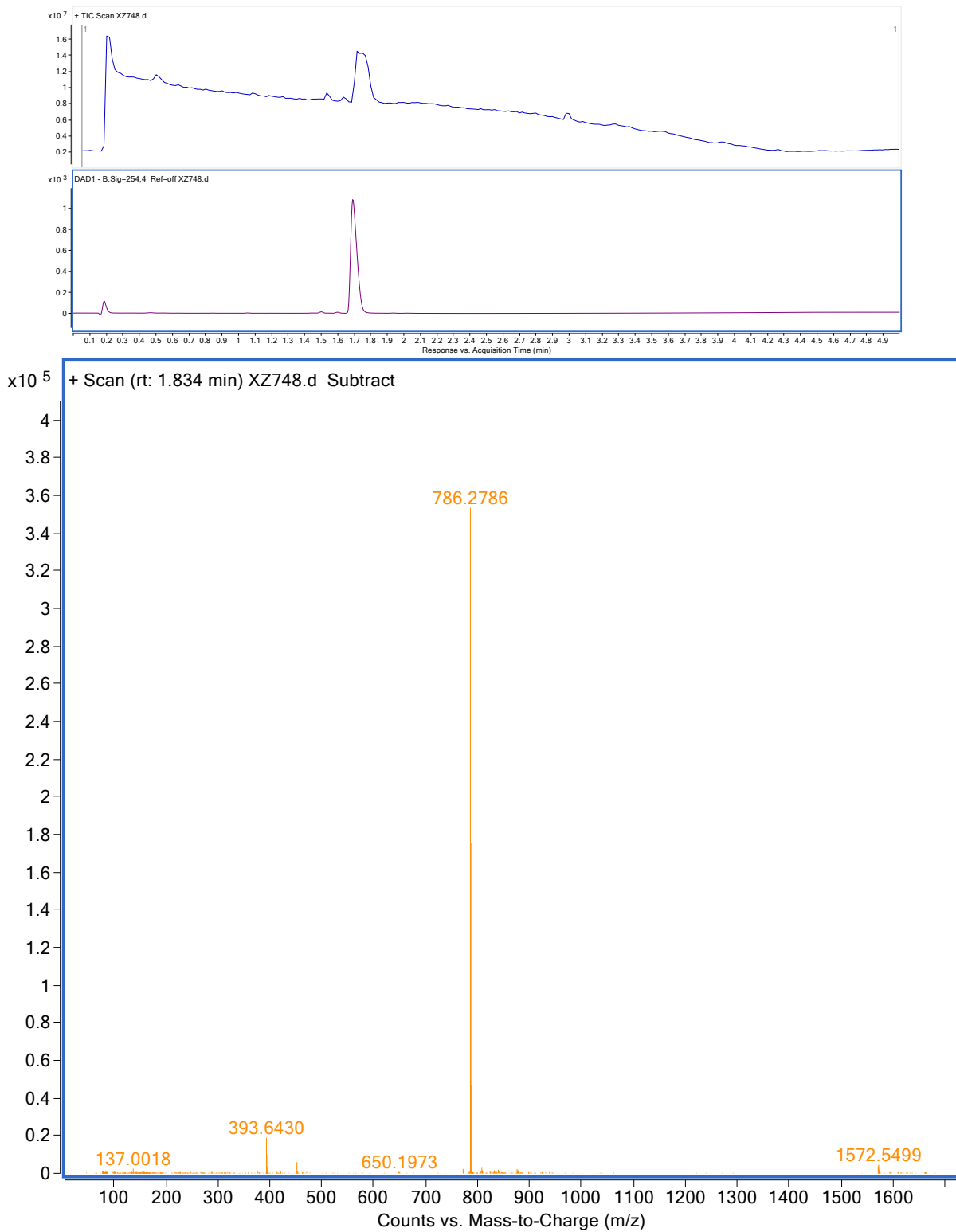
Compound 5h

HRMS (ESI) m/z calcd. for $C_{43}H_{34}N_9O_6$ (MH^+), 772.2627; found, 772.2625. ($\Delta = 0.2$ ppm).



Compound 5i

HRMS (ESI) m/z calcd. for $C_{44}H_{36}N_9O_6$ (MH^+), 786.2783; found, 786.2786. ($\Delta = -0.37$ ppm).



Compound 5j

HRMS (ESI) m/z calcd. for $C_{44}H_{36}N_9O_6$ (MH^+), 786.2783; found, 786.2782. ($\Delta = 0.14$ ppm).

