

Supporting Information

Luminescent Alkyne-Bearing Terbium(III) Complexes and their Application to Bioorthogonal Protein Labeling

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Luminescence lifetime measurements

Lifetime measurements were performed on a Varian Cary Eclipse fluorescence spectrophotometer using a 1 cm path length quartz cuvette and the following settings: total decay time = 30 ms, number of flashes = 1, delay time = 0.1 ms, gate time = 0.1 ms, excitation slit width = 5 nm, emission slit width = 5 nm, number of cycles = 100. Samples were prepared at 10 μ M concentration in both H₂O and D₂O. Samples were excited at their respective λ_{max} : 287 nm for Tb-L¹ and 300 nm for “clicked” Tb-L¹. Emission intensity was recorded at 543 nm. The resulting luminescence decay curves were fitted to following equation using the SigmaPlot software:

$$I_t = I_0 * \exp(-t/\tau)$$

where I_t is the intensity at time t after the excitation flash, I_0 is the initial intensity at $t = 0$, and τ is the luminescence lifetime.

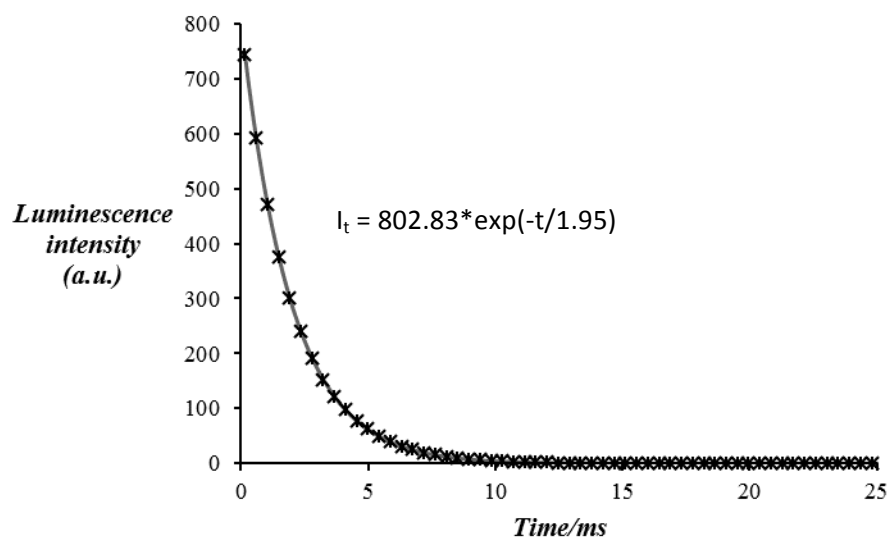


Figure S1. Luminescence decay for Tb-L¹ in H₂O; $\tau = 1.95$ ms. Experimental data points are marked by crosses (with only one in every four data points shown to aid clarity) and the fitted curve is shown as a solid gray line.

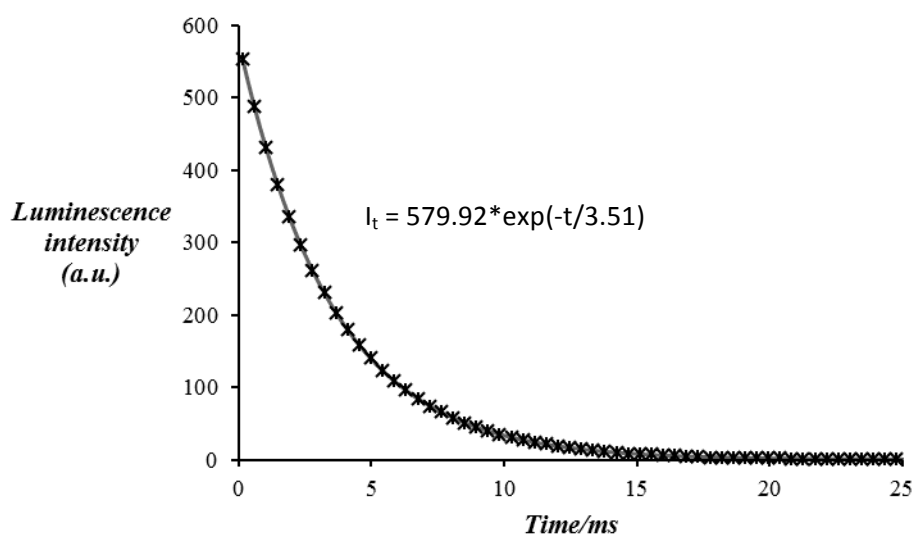


Figure S2. Luminescence decay for Tb-L¹ in D₂O; $\tau = 3.51$ ms. Experimental data points are marked by crosses (with only one in every four data points shown to aid clarity) and the fitted curve is shown as a solid gray line.

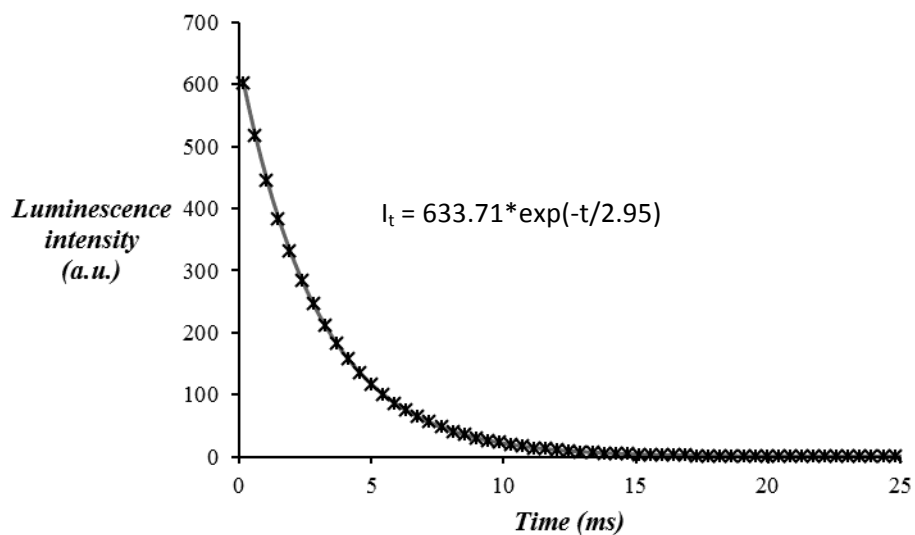


Figure S3. Luminescence decay for clicked Tb-L¹ in H₂O; $\tau = 2.95$ ms. Experimental data points are marked by crosses (with only one in every four data points shown to aid clarity) and the fitted curve is shown as a solid gray line.

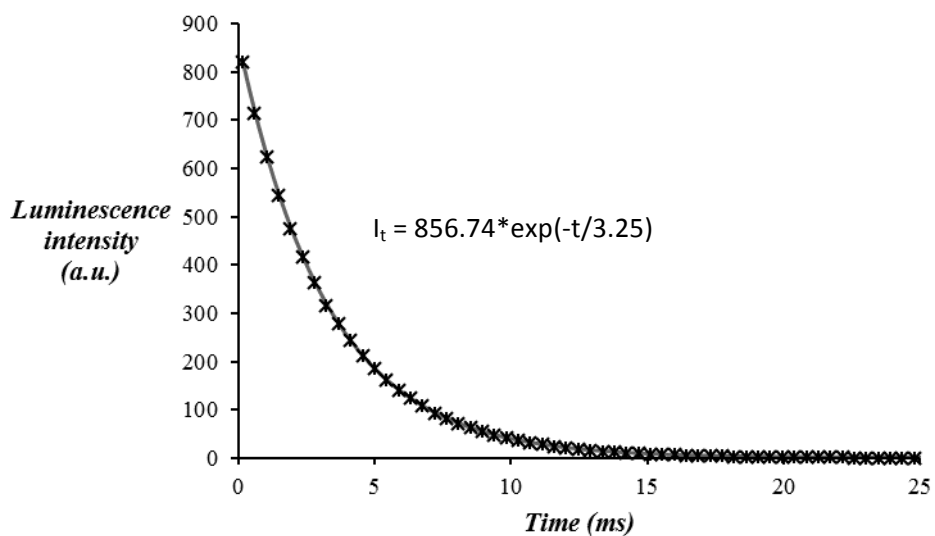


Figure S4. Luminescence decay for clicked Tb-L¹ in D₂O; $\tau = 3.25$ ms. Experimental data points are marked by crosses (with only one in every four data points shown to aid clarity) and the fitted curve is shown as a solid gray line.

Luminescence of labeled protein samples

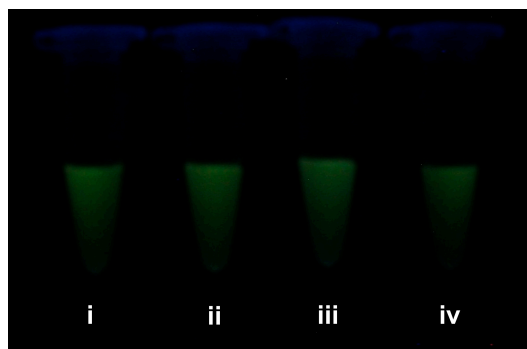


Figure S5. Photograph of spin-dialyzed solutions of labelled DEBP samples (*ca.* 10 μM in H_2O) irradiated with a common laboratory TLC lamp (254 nm): (i) DEBP-Q80AzF+Tb- L^1 , (ii) DEBP-Q80AzF+Tb- L^2 , (iii) DEBP-Q80AzMF+Tb- L^1 , (iv) DEBP-Q80AzMF+Tb- L^2 .

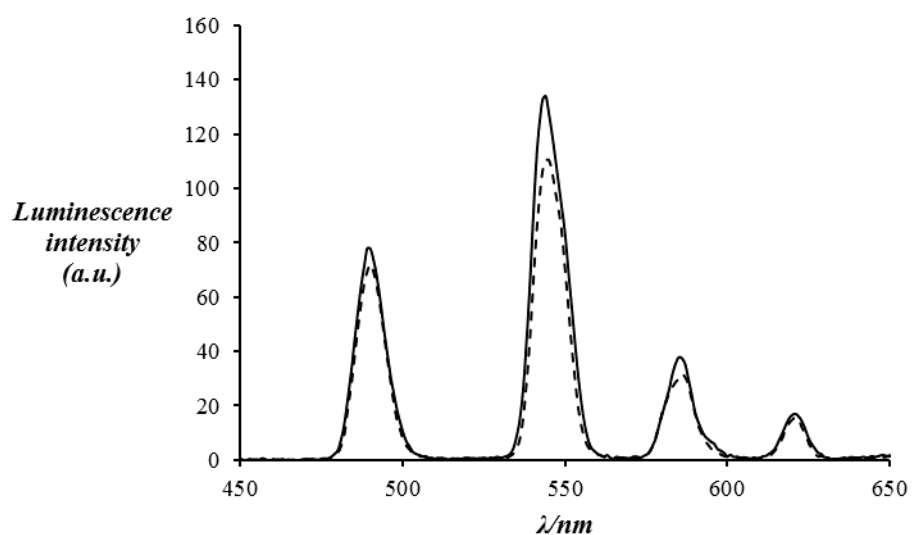


Figure S6. Luminescence emission spectra of DEBP-Q80AzF+Tb- L^1 (*ca.* 10 μM in H_2O) excited at 300 nm (dashed line) and DEBP-Q80AzF+Tb- L^2 (*ca.* 10 μM in H_2O) excited at 274 nm (solid line).

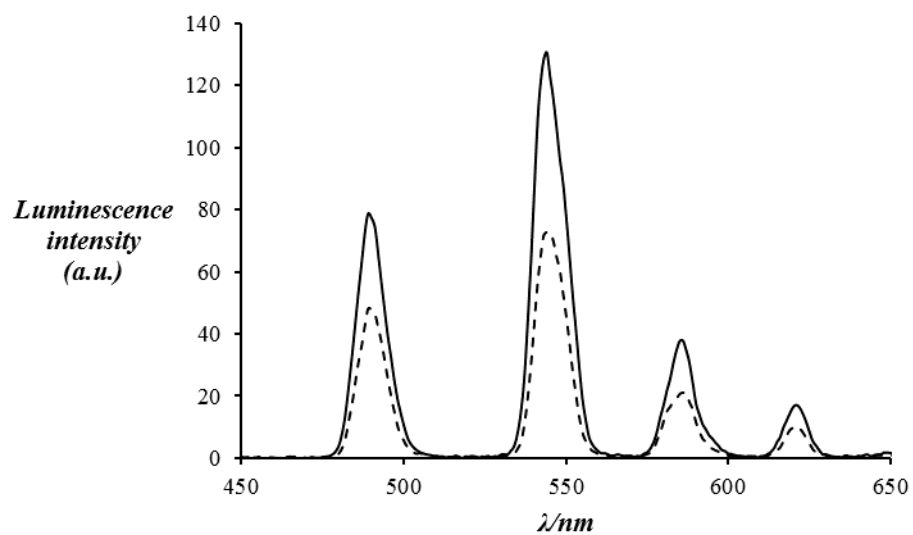


Figure S7. Luminescence emission spectra of DEBP-Q80AzMF+Tb-L¹ (ca. 10 μ M in H₂O) excited at 300 nm (dashed line) and DEBP-Q80AzMF+Tb-L² (ca. 10 μ M in H₂O) excited at 274 nm (solid line).

NMR spectra of reported compounds

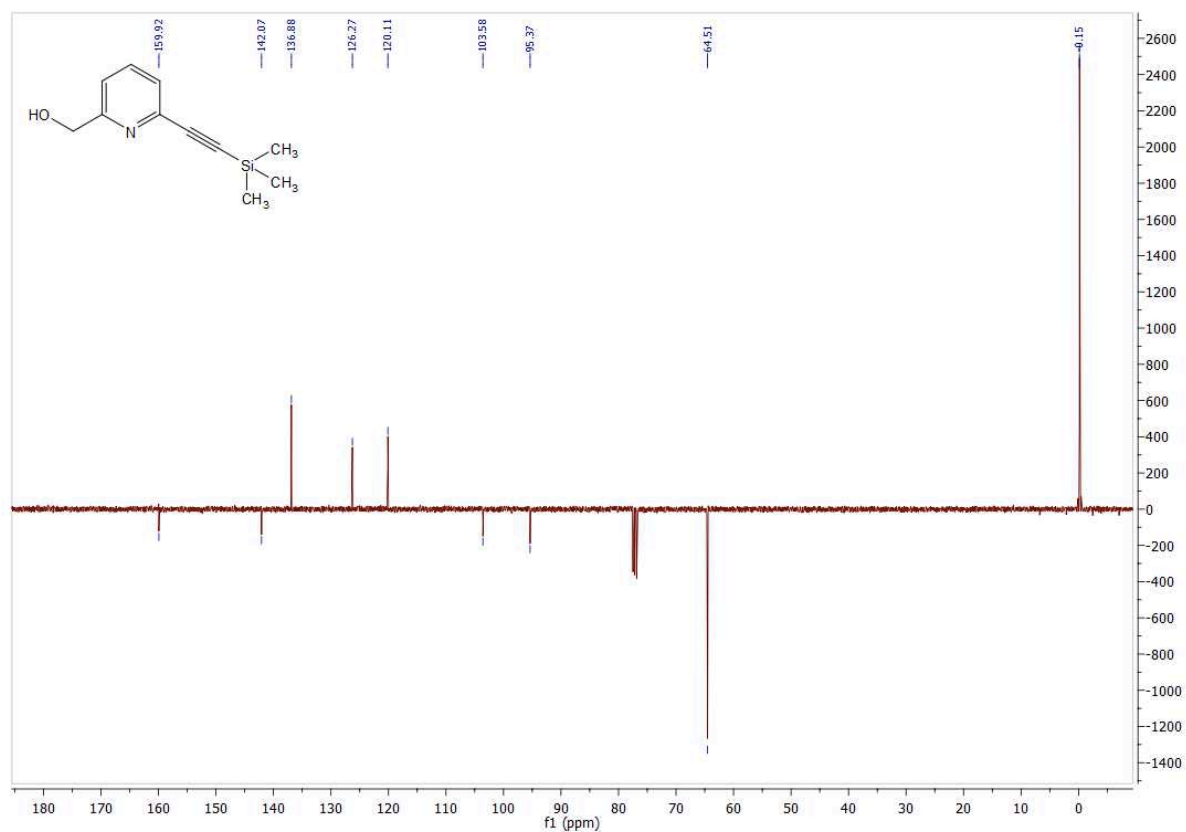
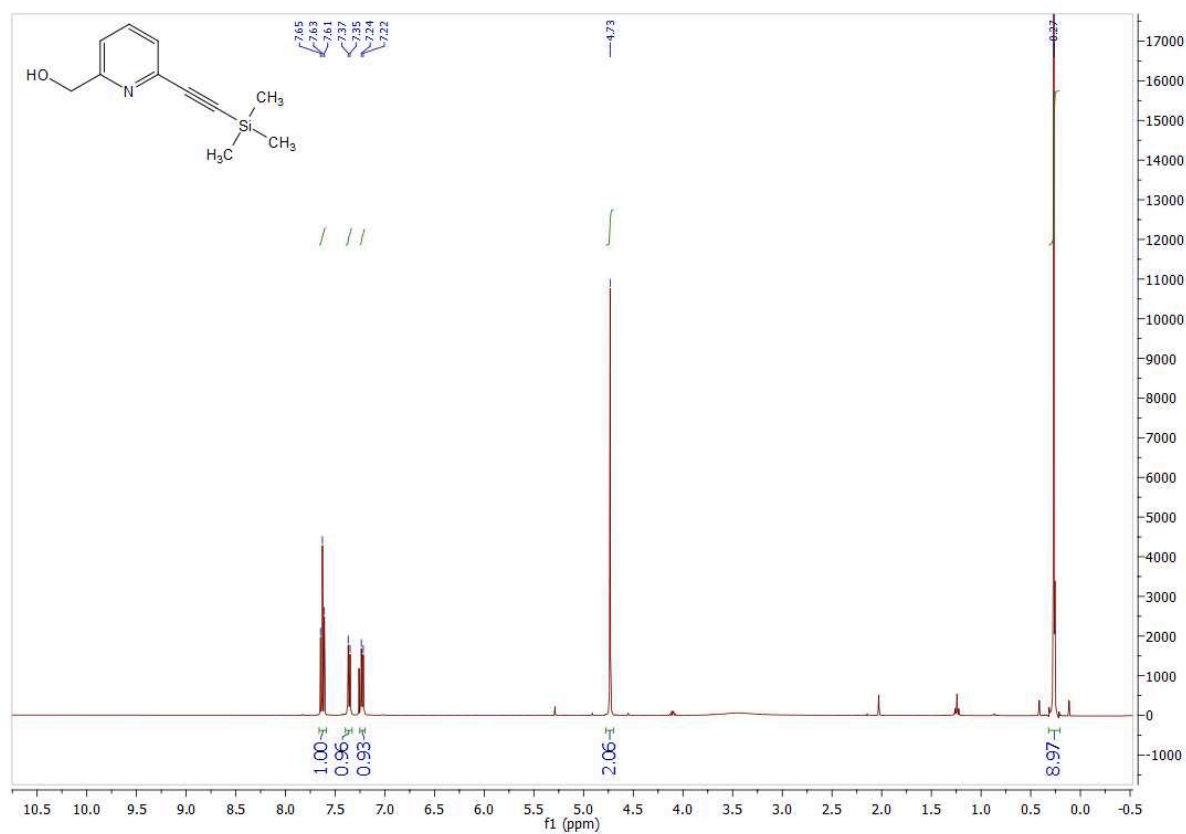


Figure S8. ¹H and ¹³C NMR spectra of **2** in CDCl₃.

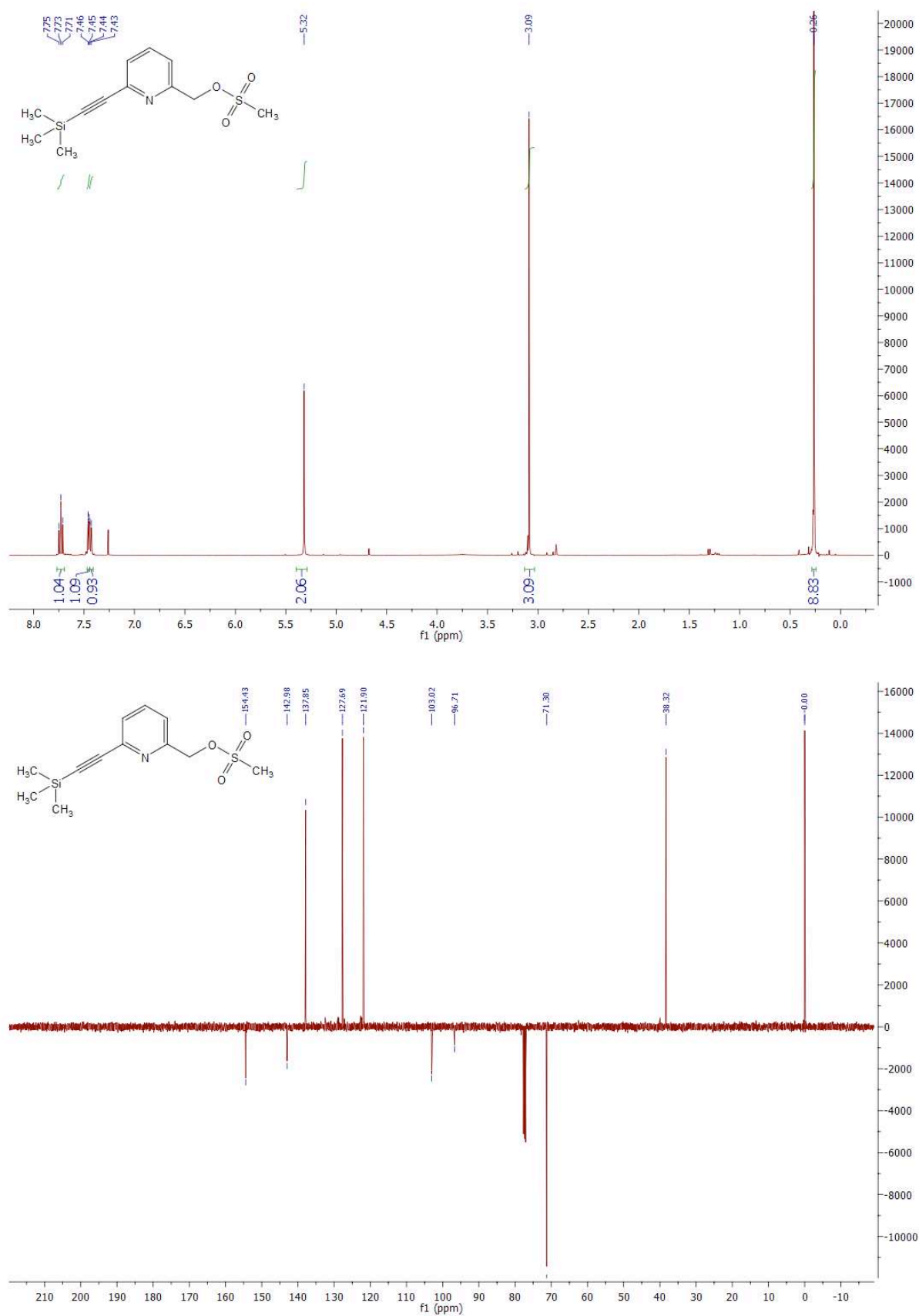


Figure S9. ¹H and ¹³C NMR spectra of **3** in CDCl₃.

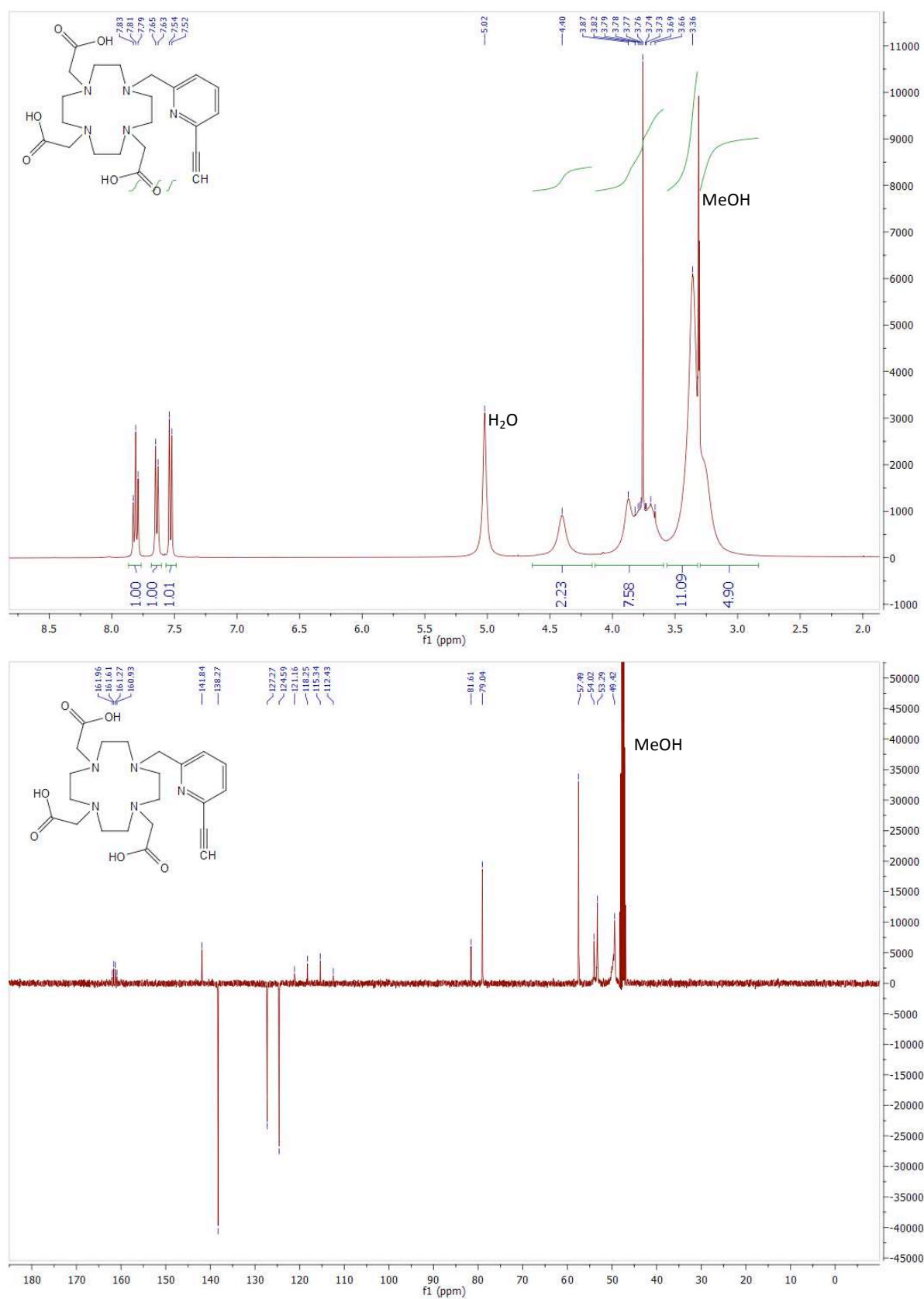


Figure S10. ¹H and ¹³C NMR spectra of H₃L¹ in MeOD.

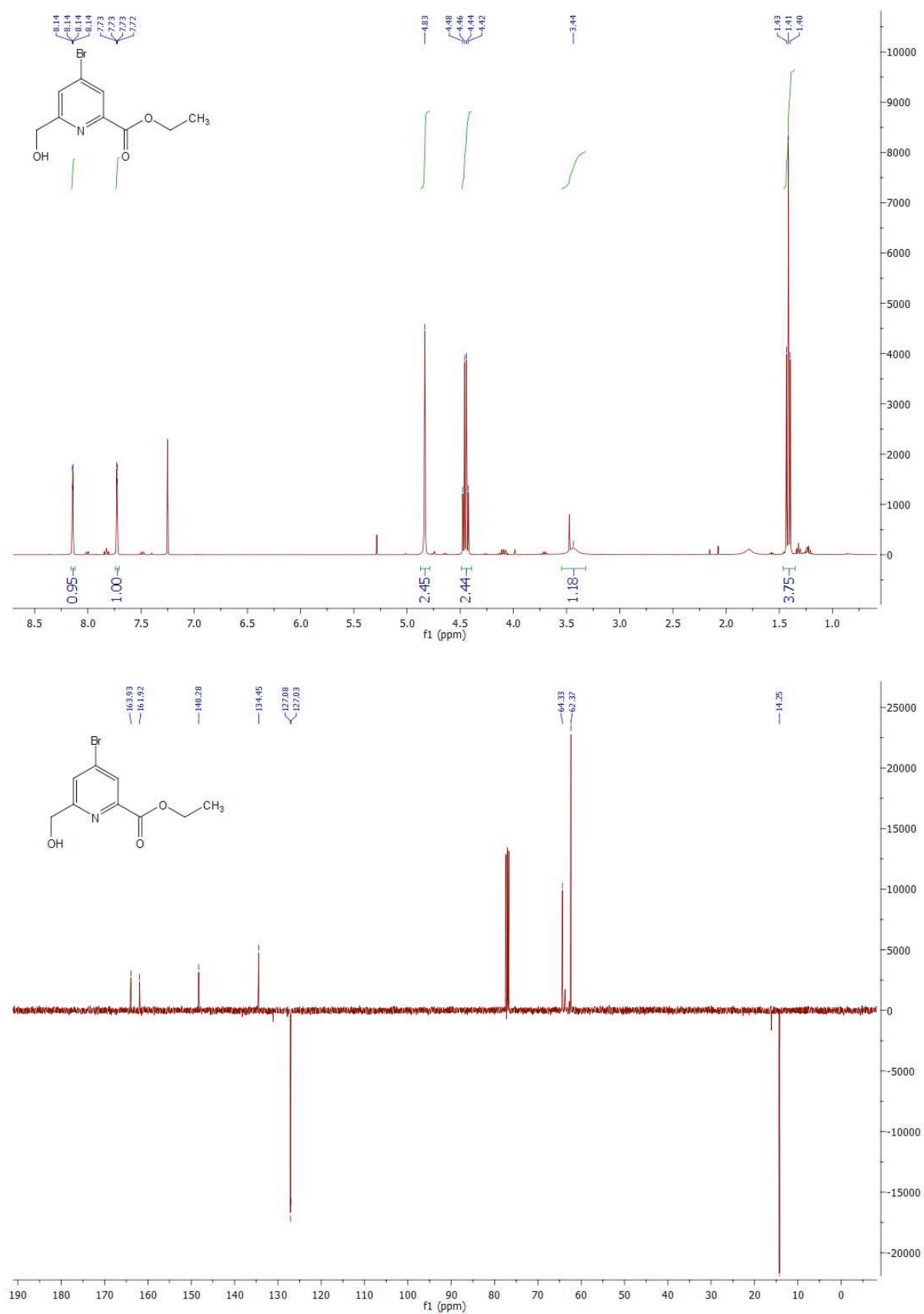


Figure S11. ¹H and ¹³C NMR spectra of **5** in CDCl₃.

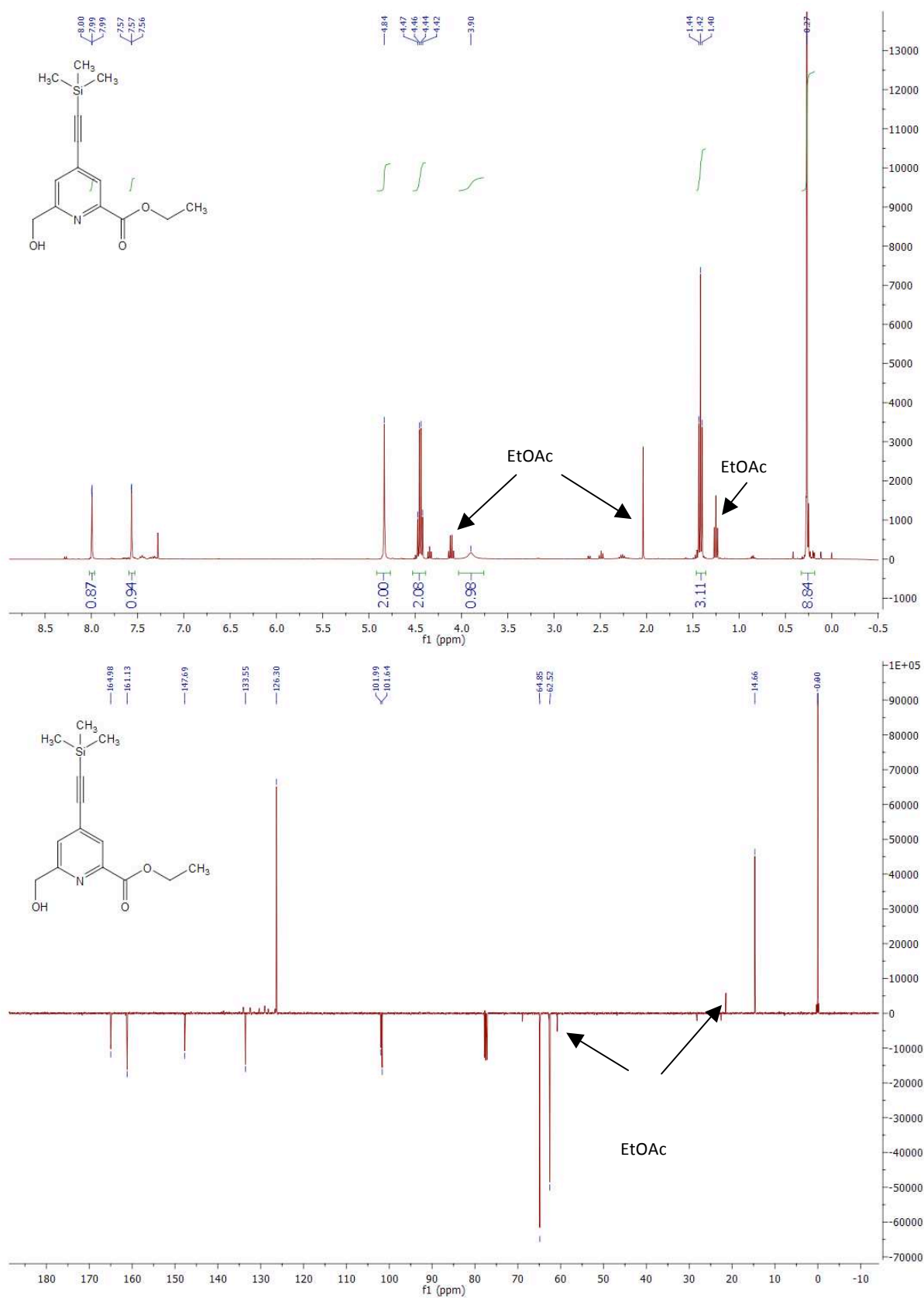


Figure S12. ¹H and ¹³C NMR spectra of **6** in CDCl₃.

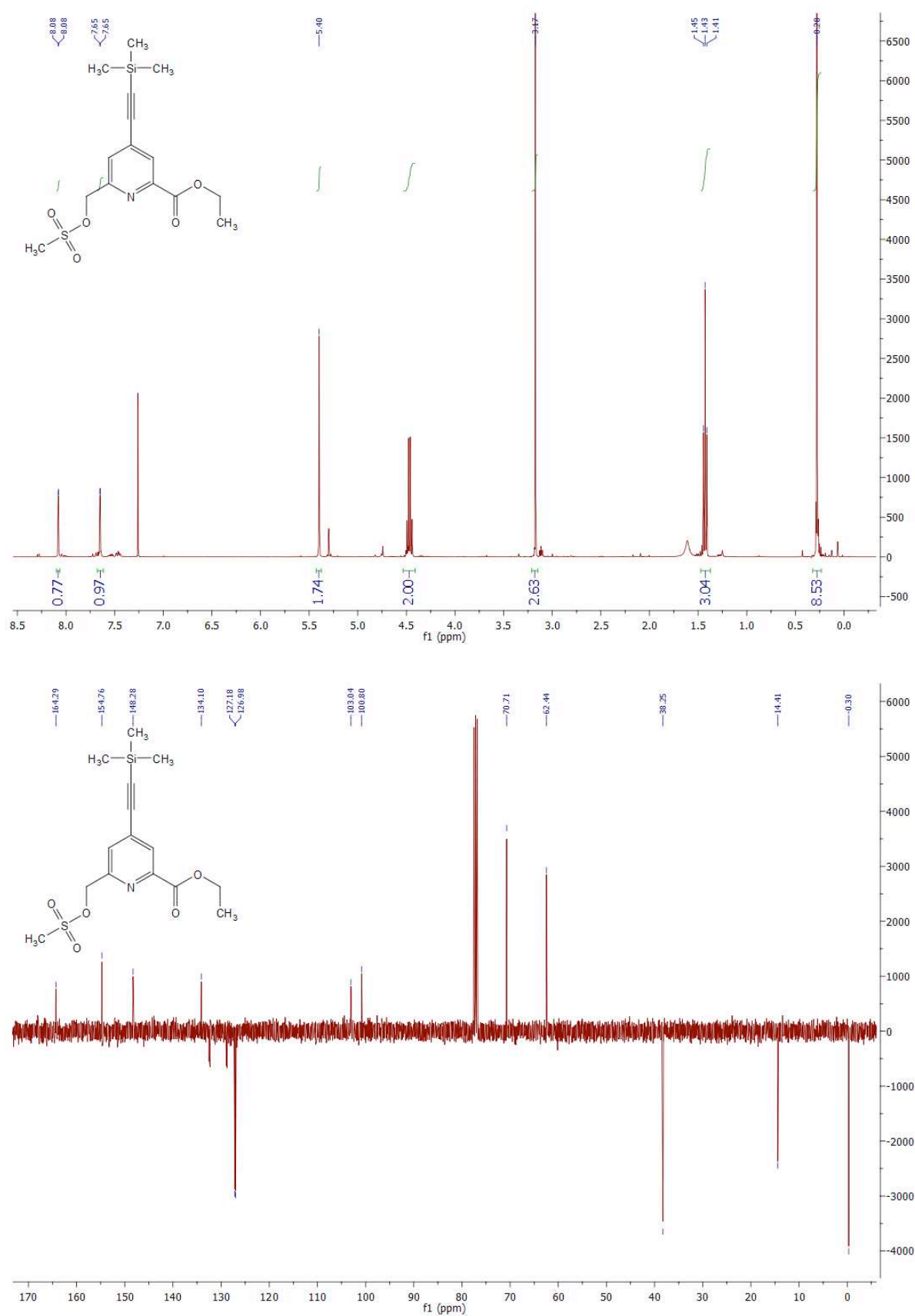


Figure S13. ¹H and ¹³C NMR spectra of 7 in CDCl₃.

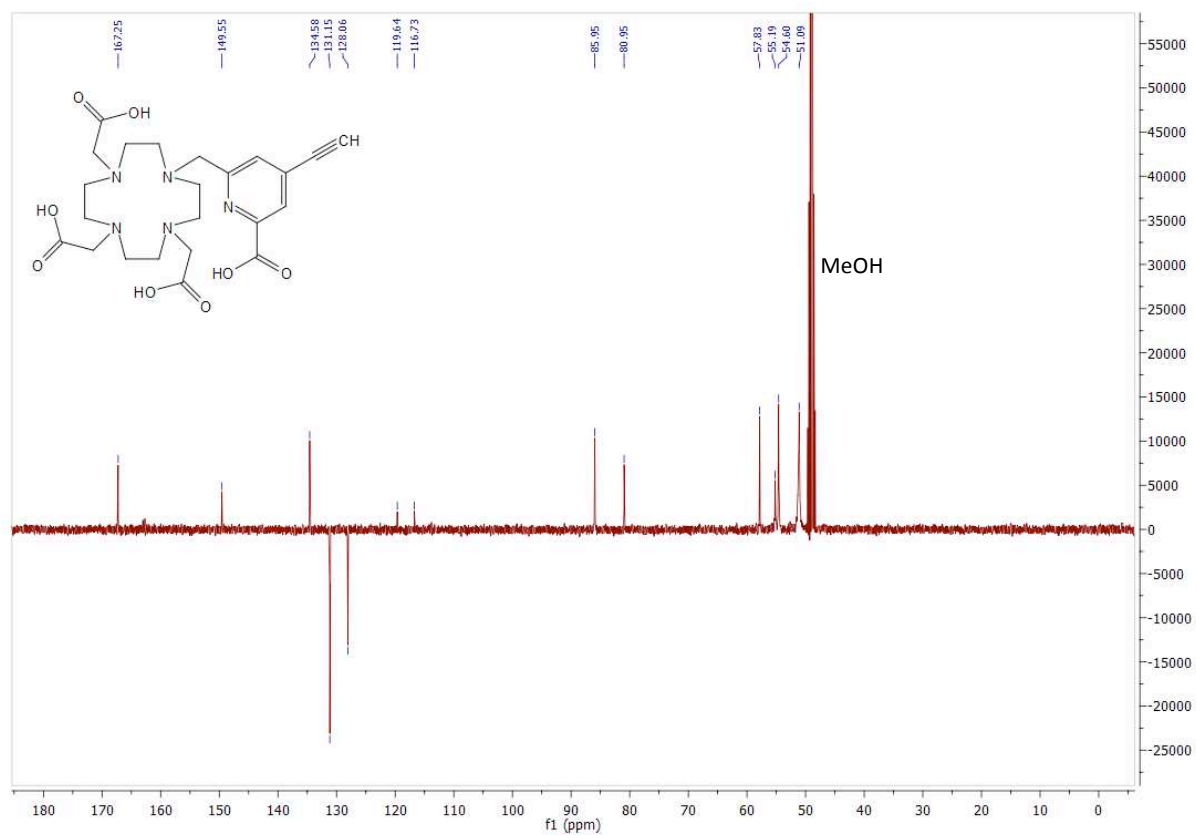
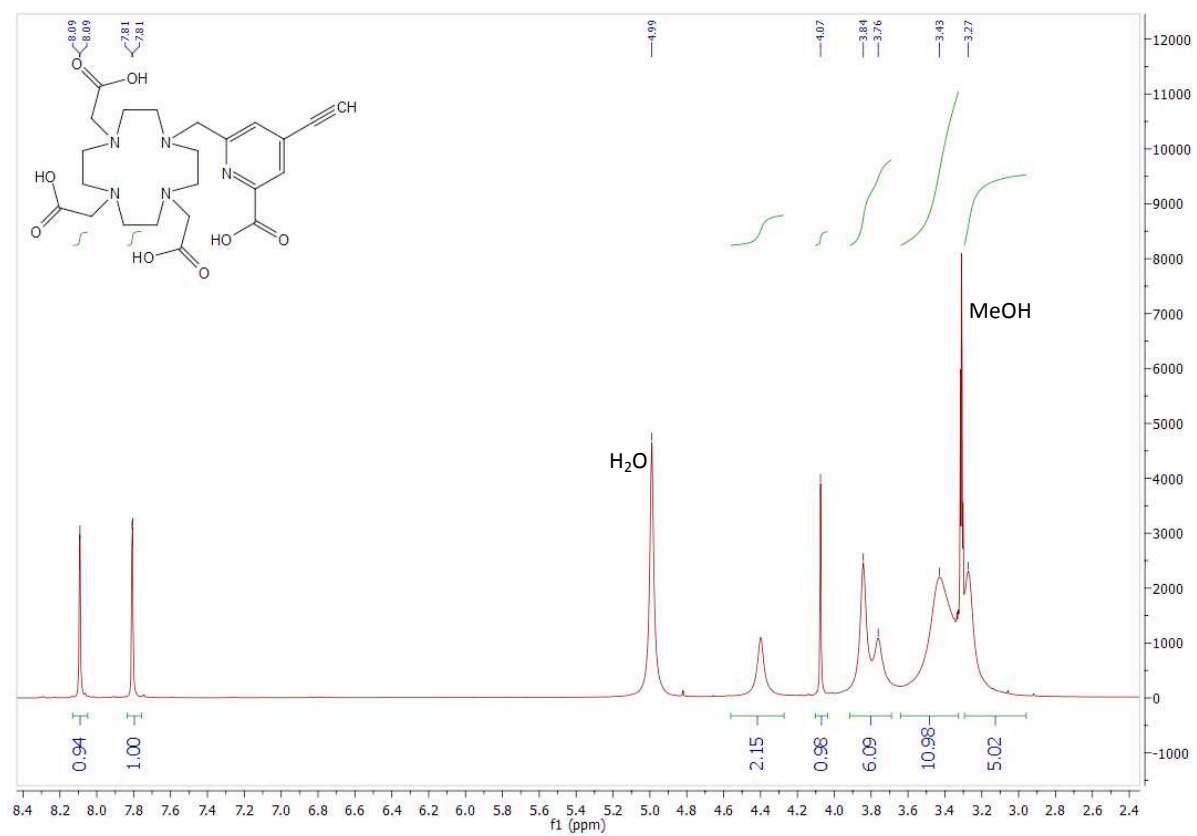


Figure S14. ¹H and ¹³C NMR spectra of H₄L² in MeOD.

LC traces for ligands

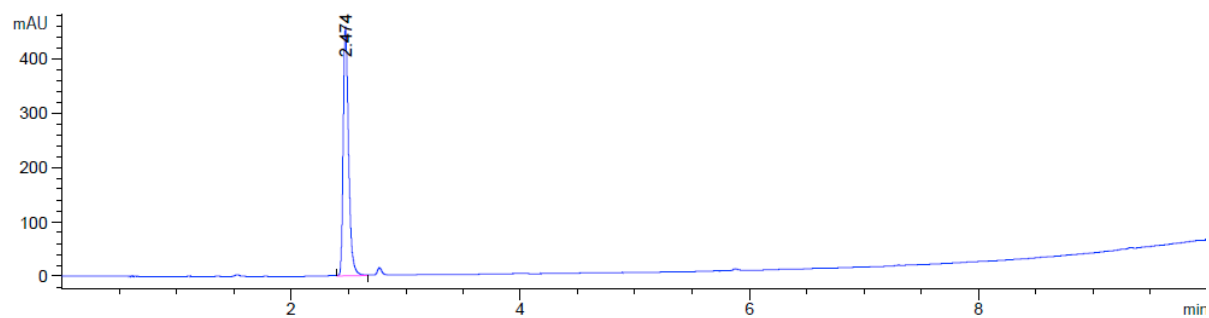


Figure S15. LC trace for H₃L¹.

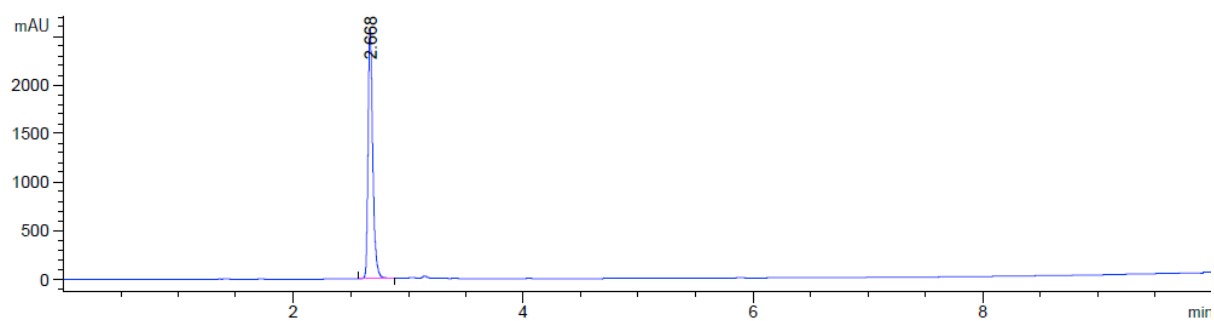


Figure S16. LC trace for H₄L².

LC traces for terbium(III) complexes

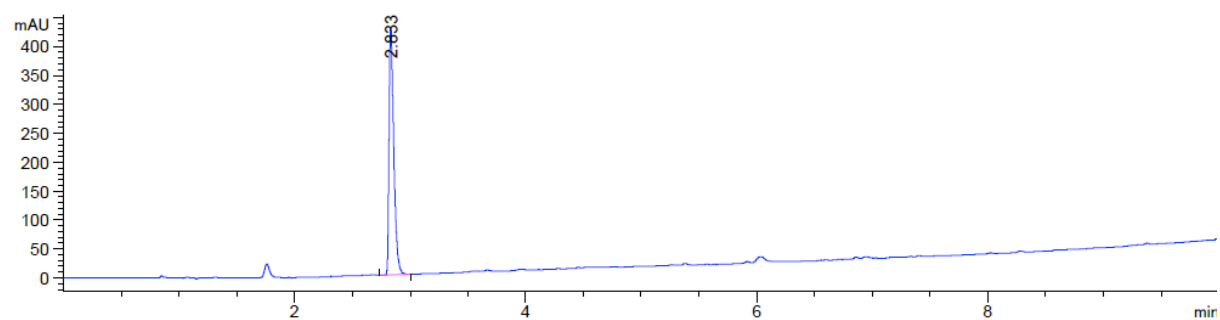


Figure S17. LC trace for Tb-L¹.

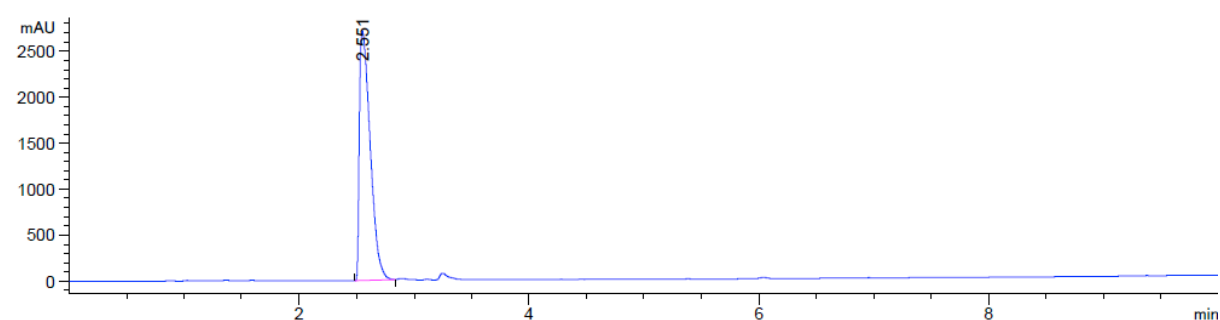


Figure S18. LC trace for Tb-L².

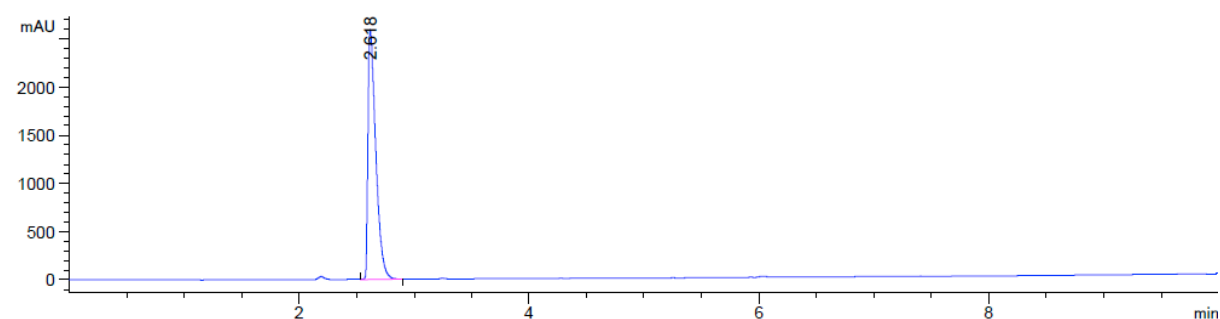


Figure S19. LC trace for clicked Tb-L¹.

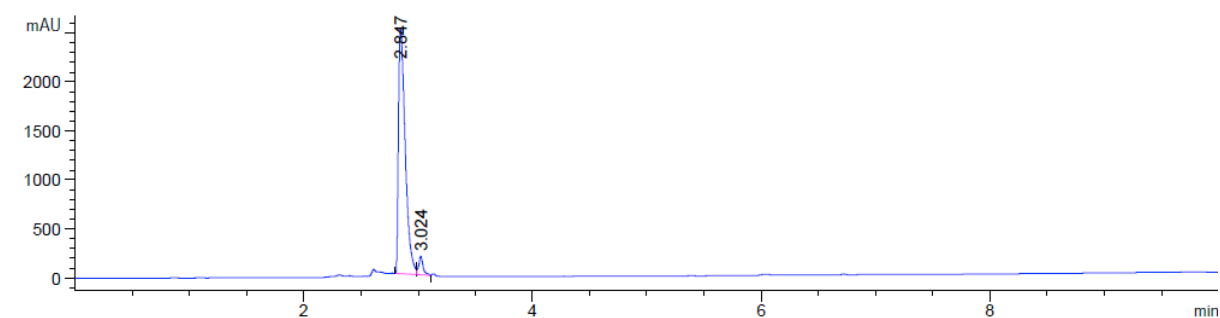


Figure S20. LC trace for clicked Tb-L².