

Supplementary Information

Picolinic acid based acyclic bifunctional chelating agent and its methionine conjugate as potential SPECT imaging agents: Syntheses and preclinical evaluation

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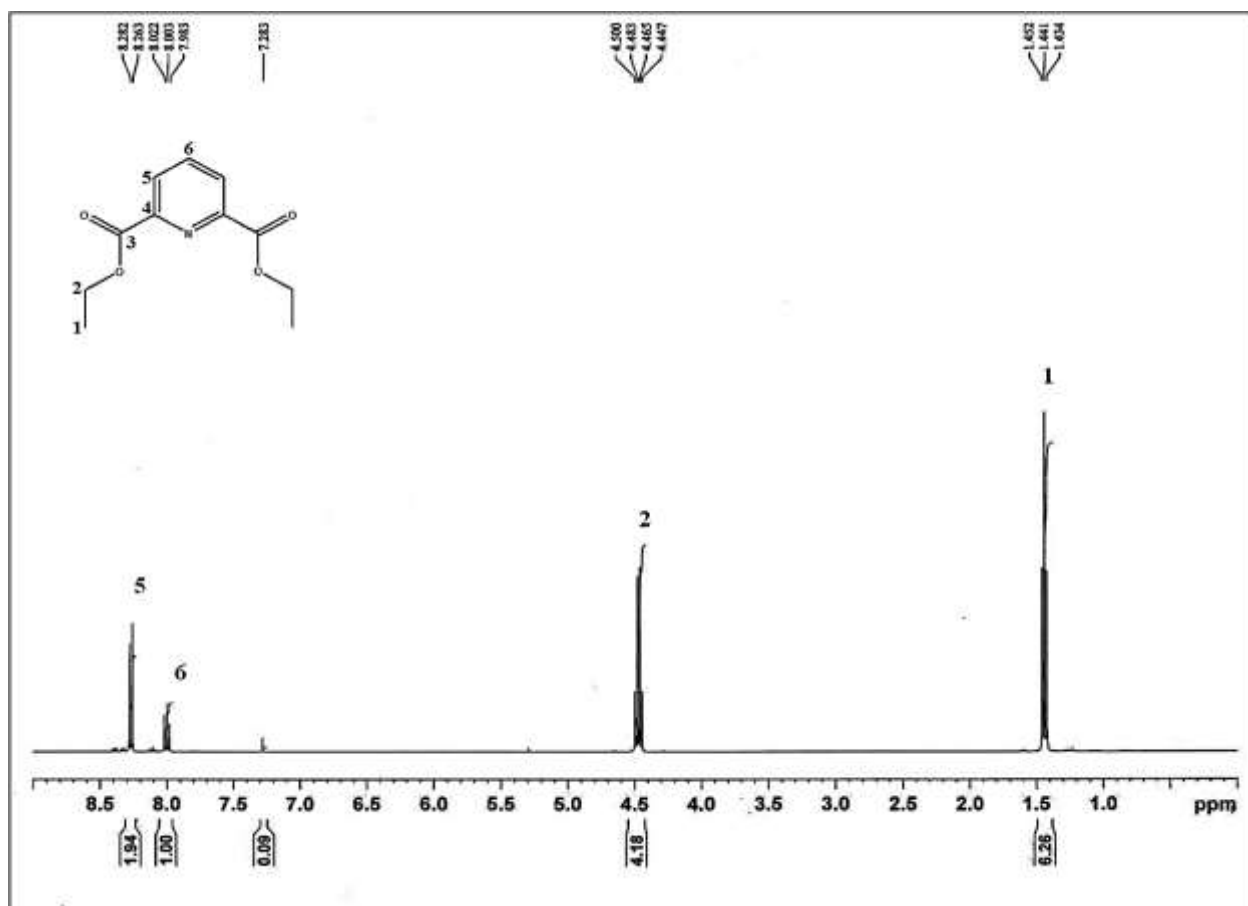


Figure S1. ¹H NMR spectrum of diethyl pyridine-2,6-di carboxylate (2)

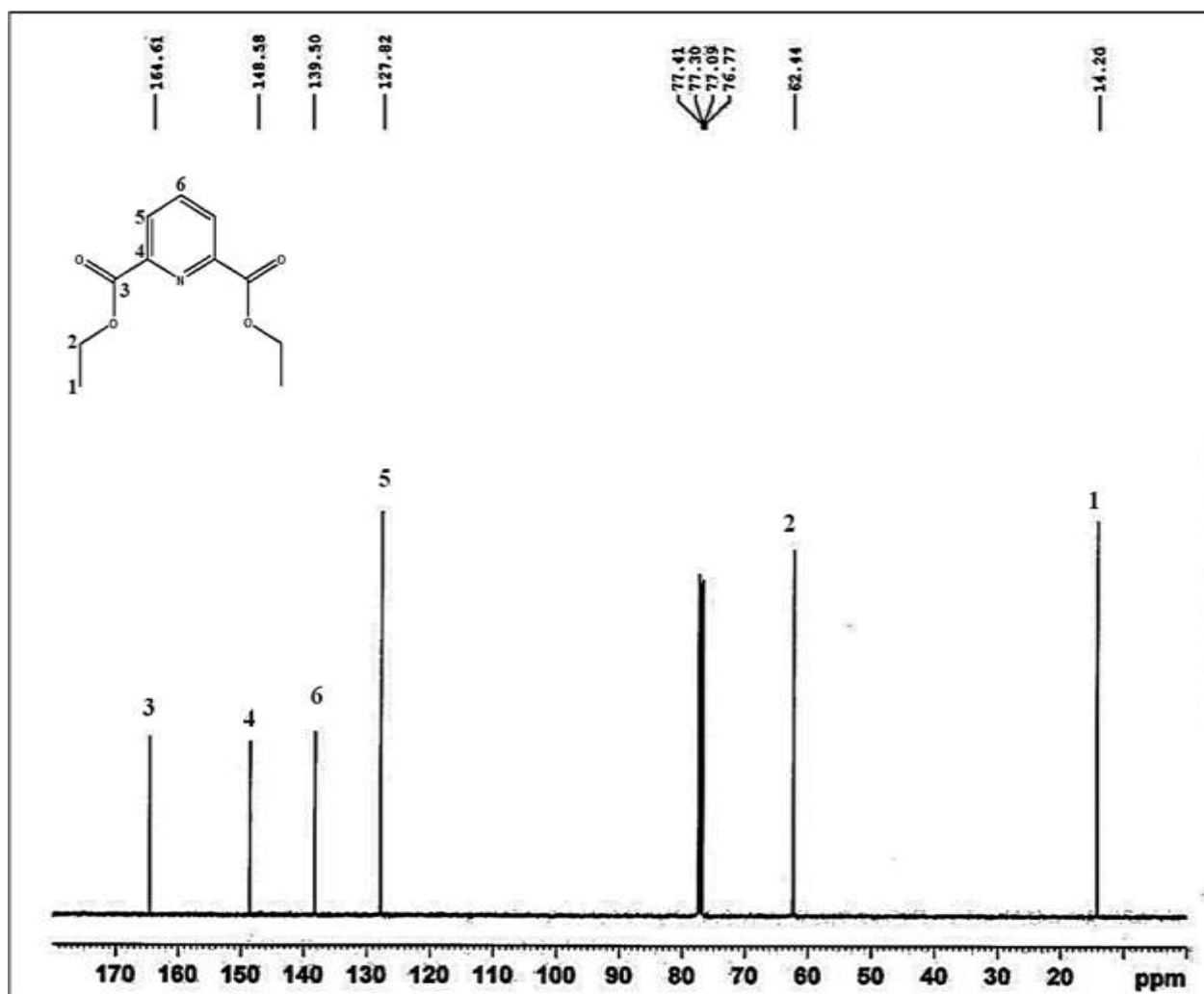


Figure S2. ^{13}C NMR spectrum of diethyl pyridine-2,6-di carboxylate (2)

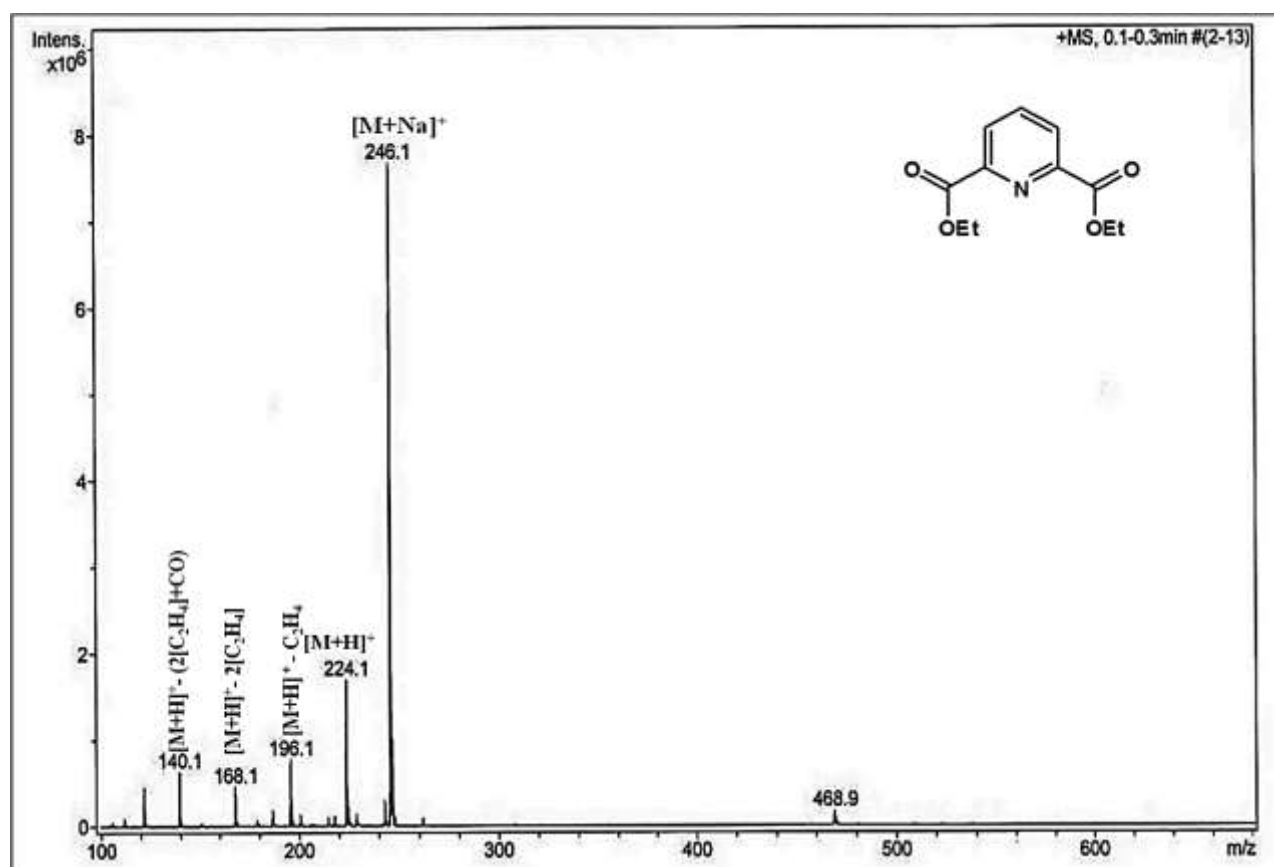


Figure S3. Mass spectrum of diethyl pyridine-2,6-di carboxylate (2)

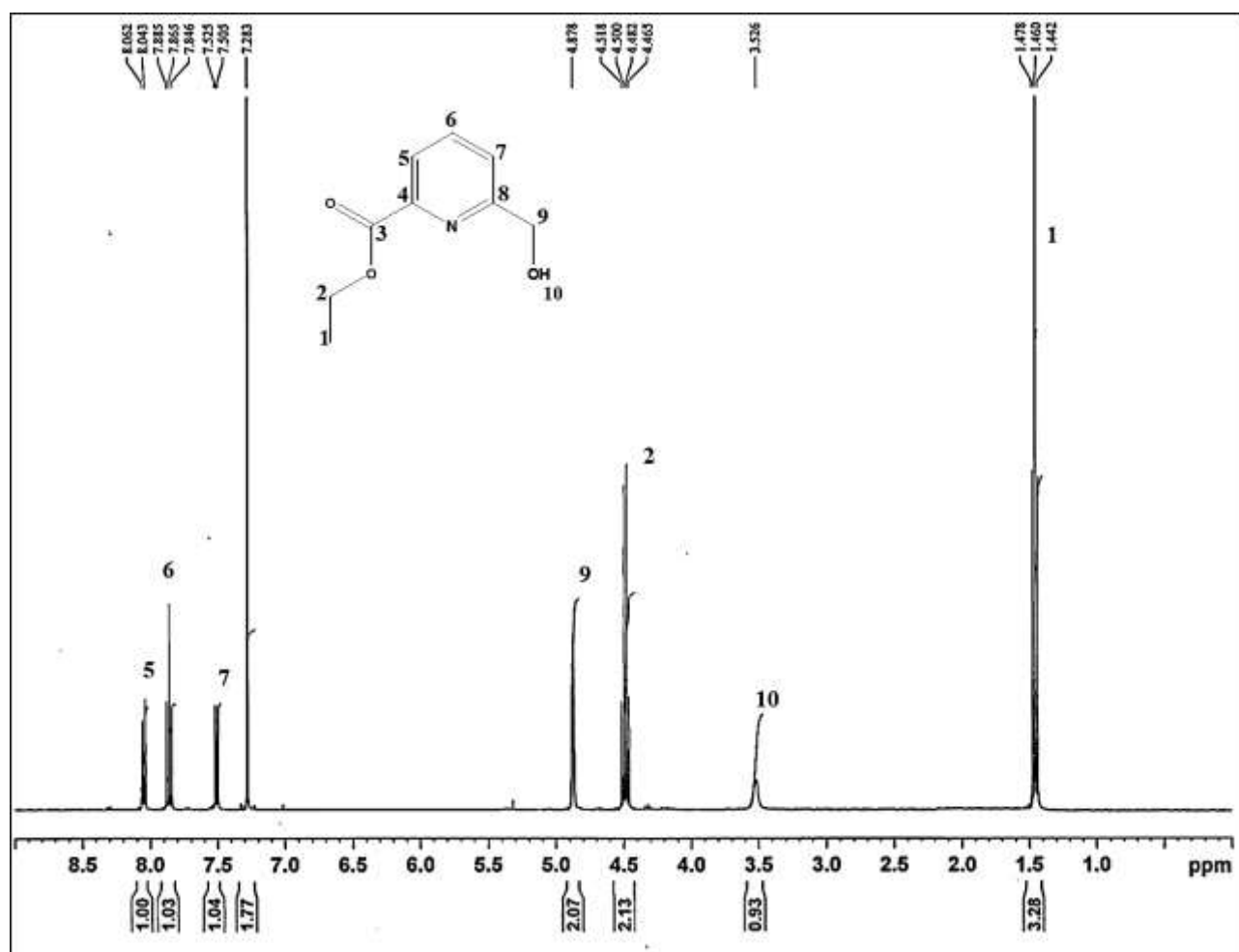


Figure S4. ^1H NMR spectrum of ethyl 6-(hydroxymethyl)picolinate (3)

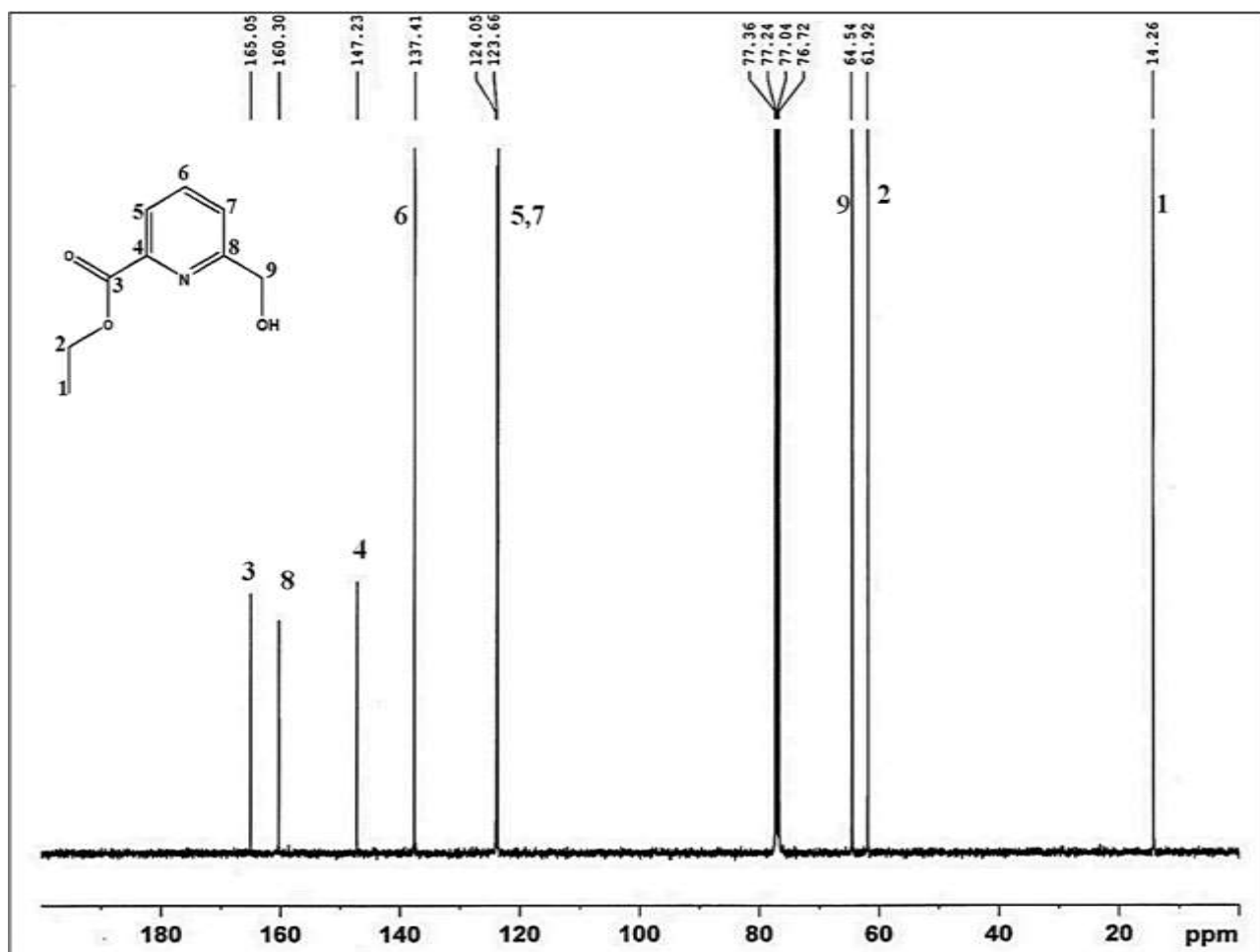


Figure S5. ^{13}C NMR spectrum of ethyl 6-(hydroxymethyl)picolinate (3)

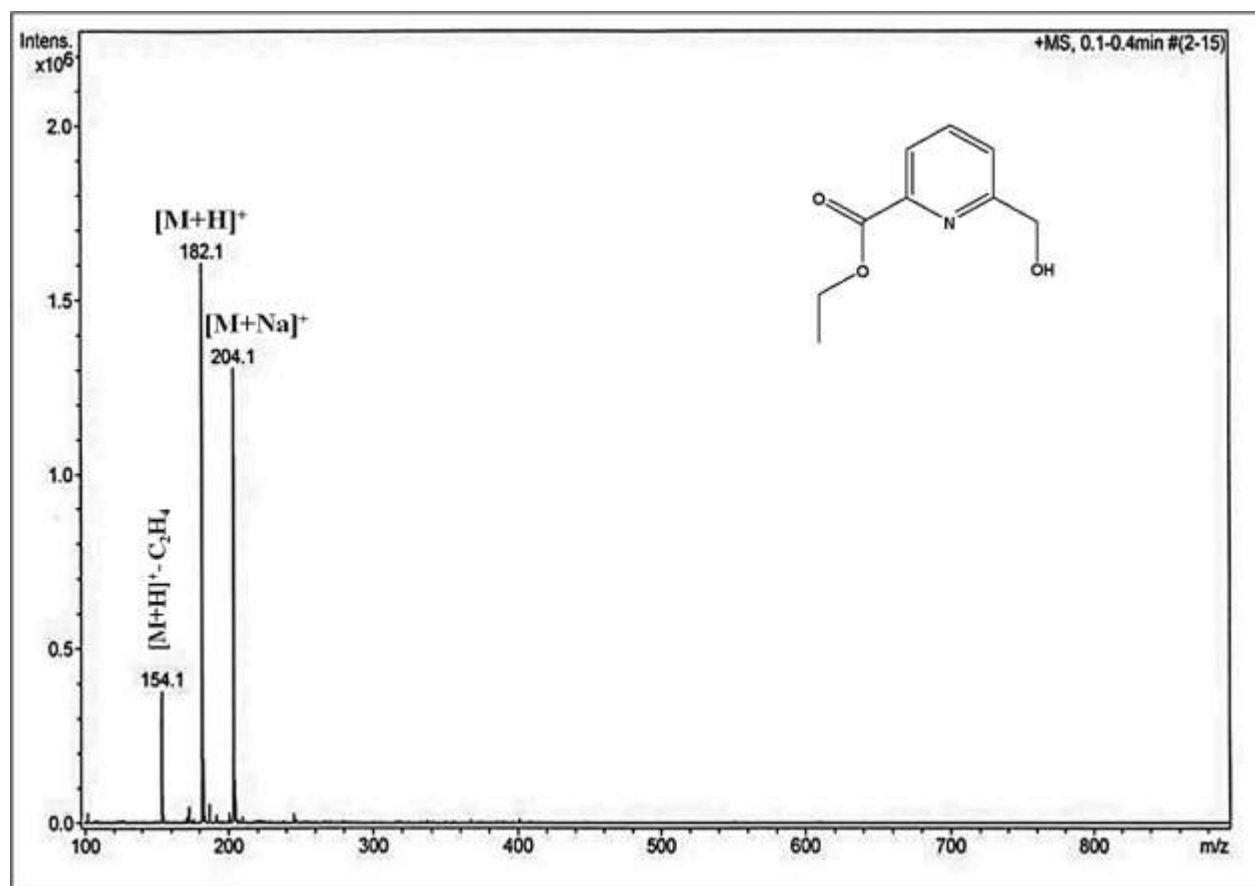


Figure S6. Mass spectrum ethyl 6-(hydroxymethyl)picolinate (**3**)

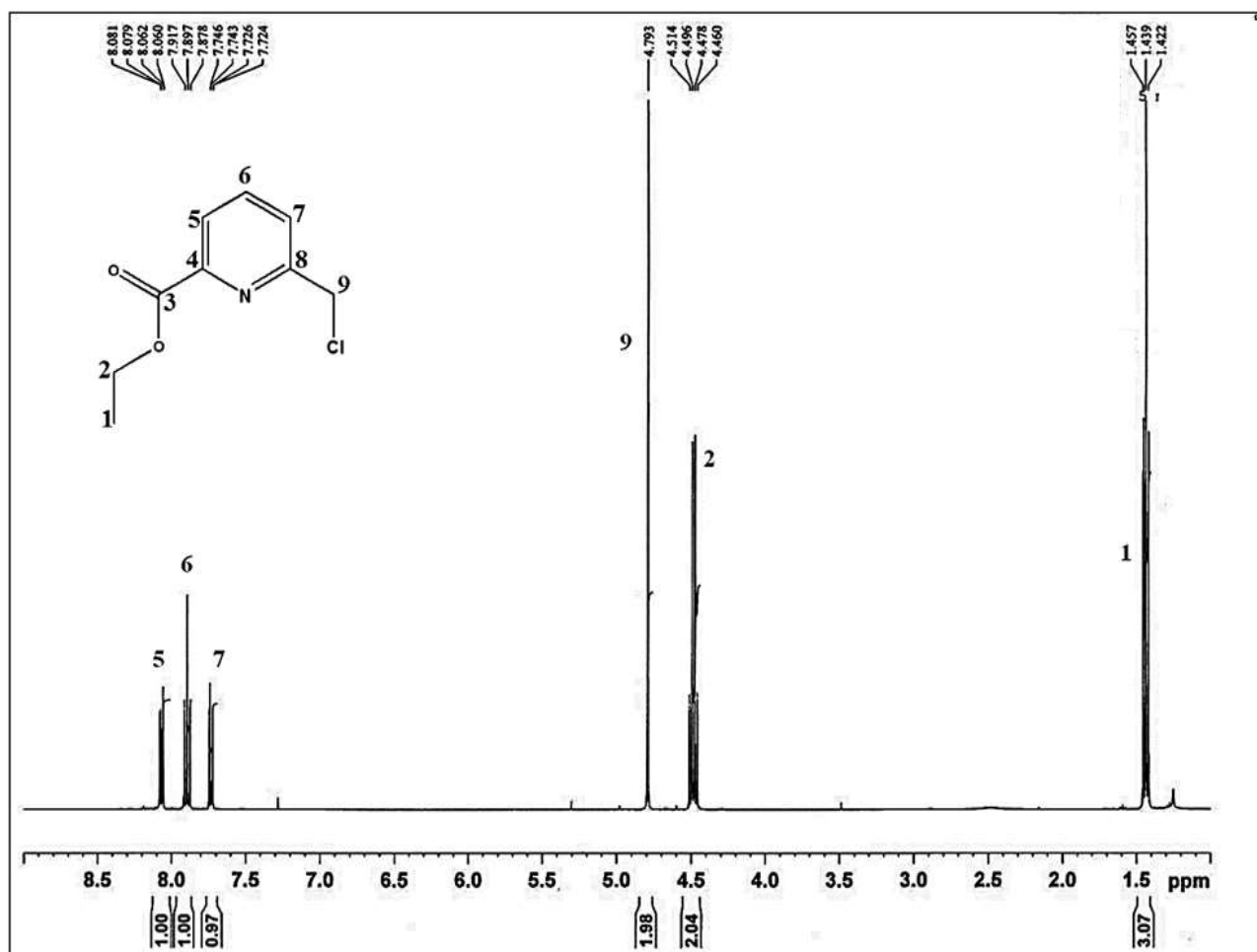


Figure S7. ^1H NMR spectrum of ethyl 6-(chloromethyl)picolinate (**4**)

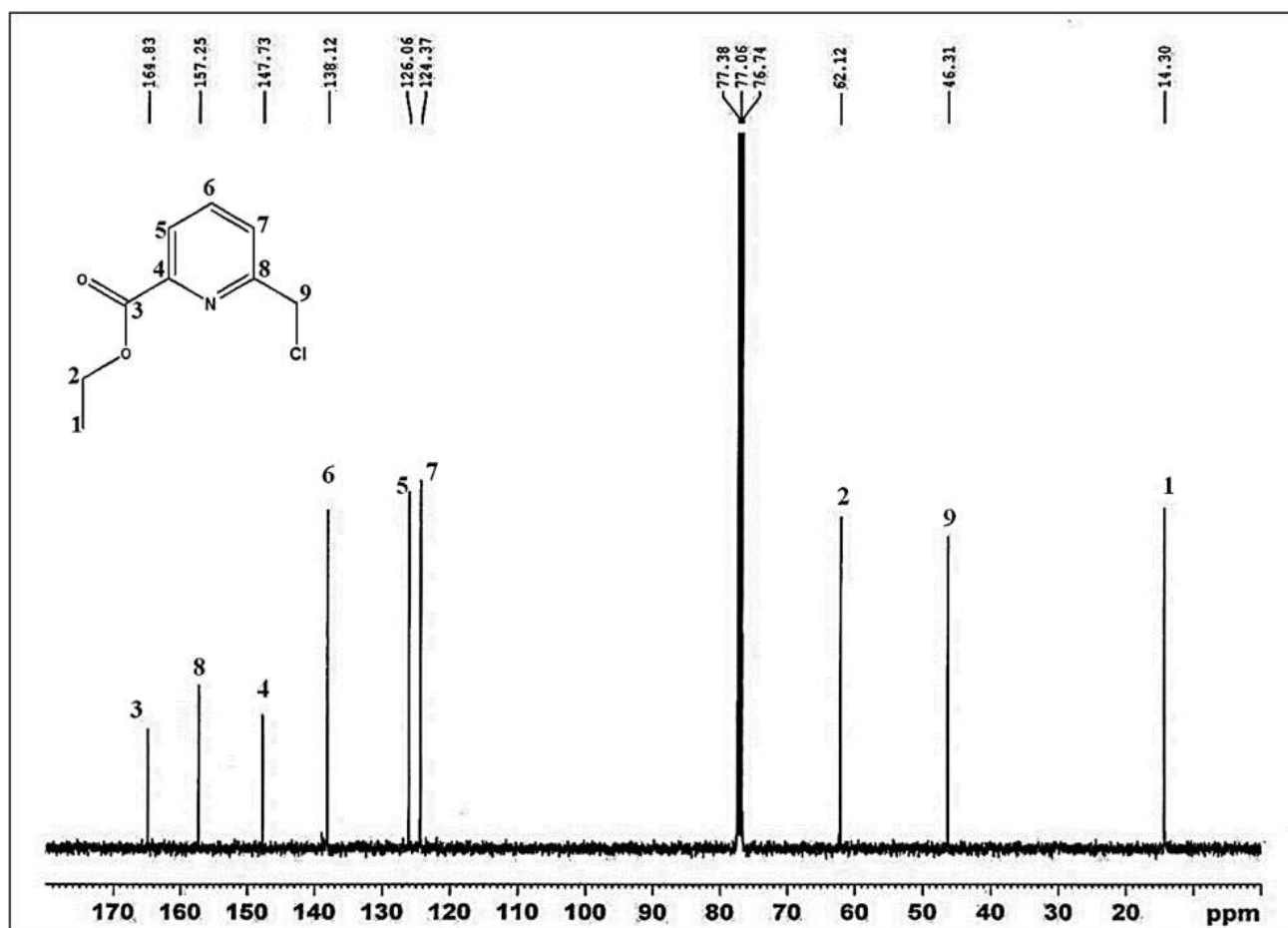


Figure S8. ^{13}C NMR spectrum of ethyl 6-(chloromethyl)picolinate (**4**)

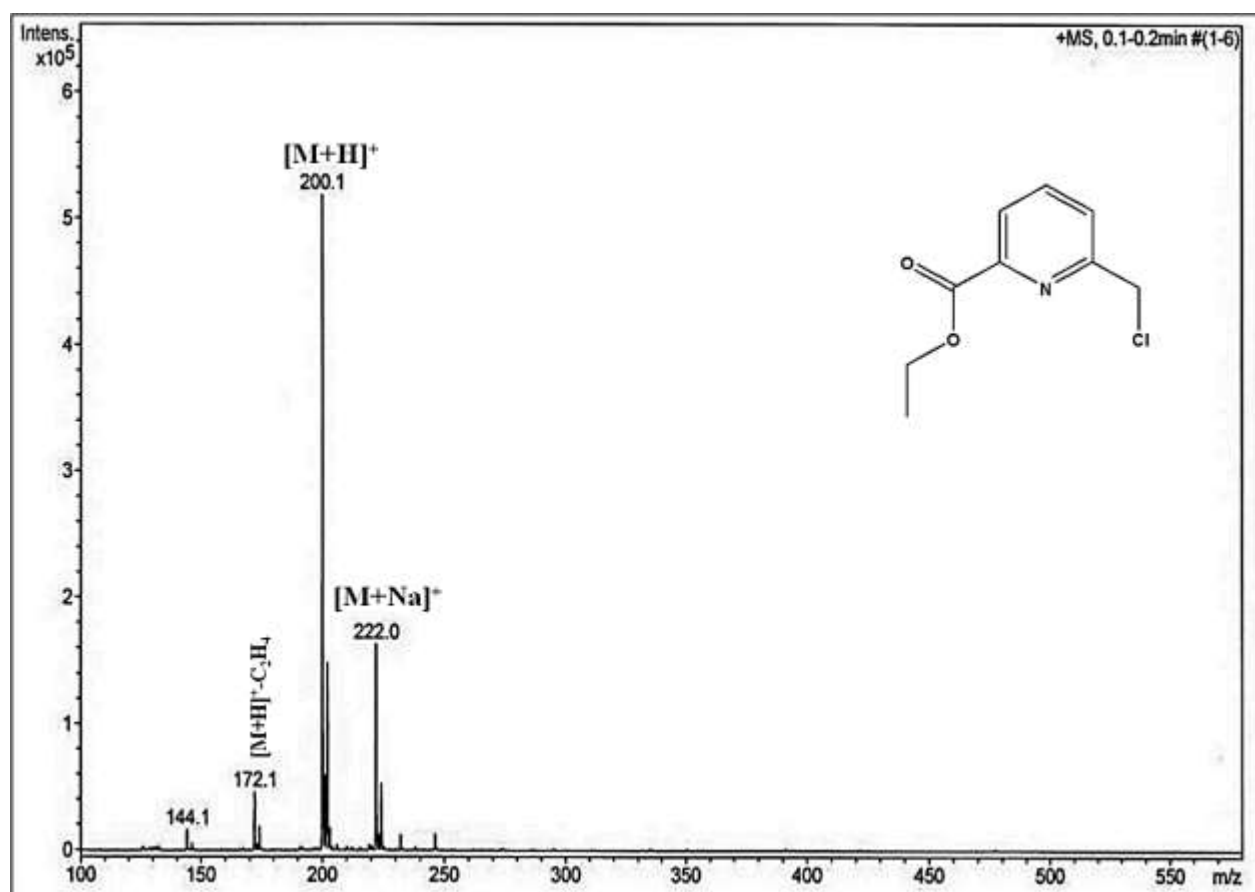


Figure S9. Mass spectrum of ethyl 6-(chloromethyl)picolinate (**4**)

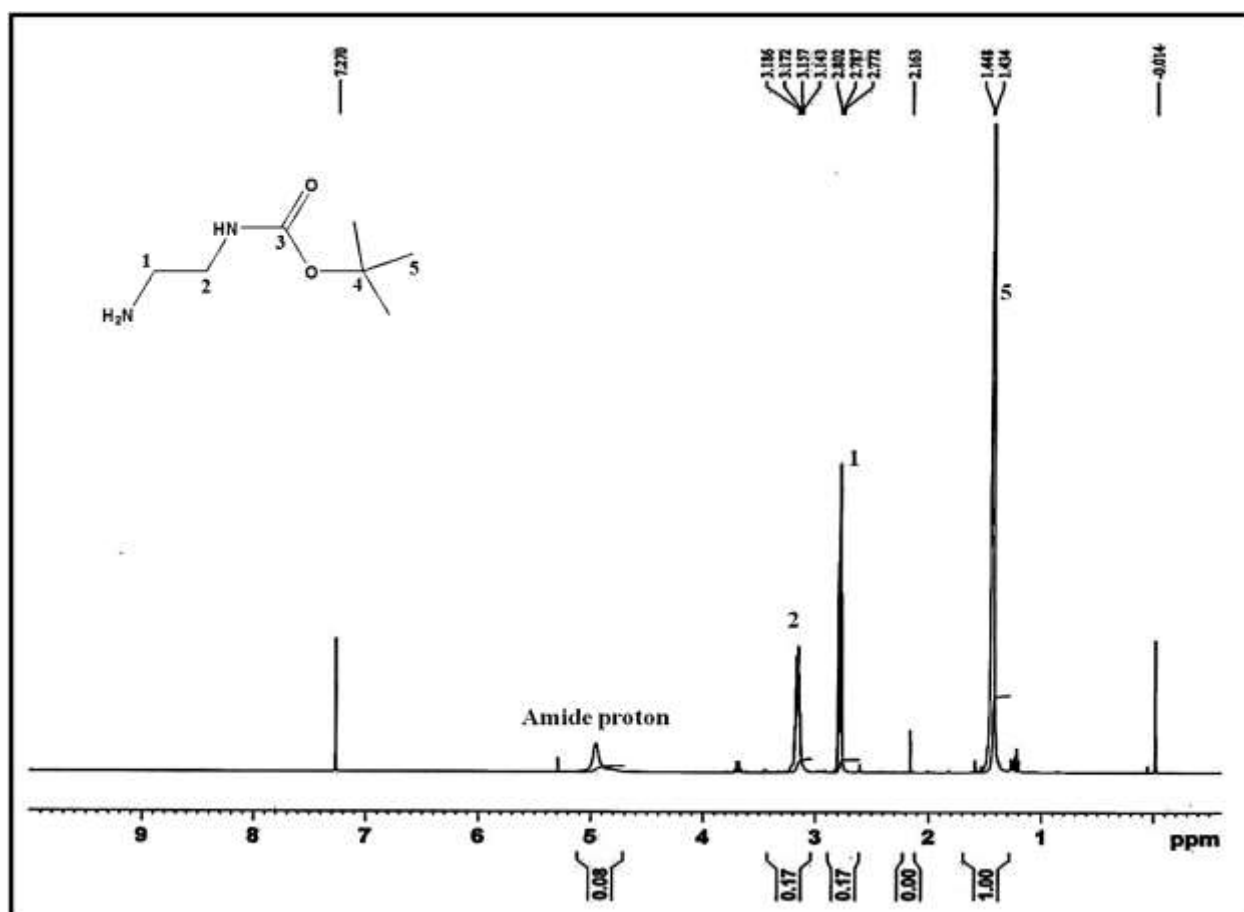


Figure S10. ¹H NMR spectrum of *tert*-butyl 2-aminoethylcarbamate (6)

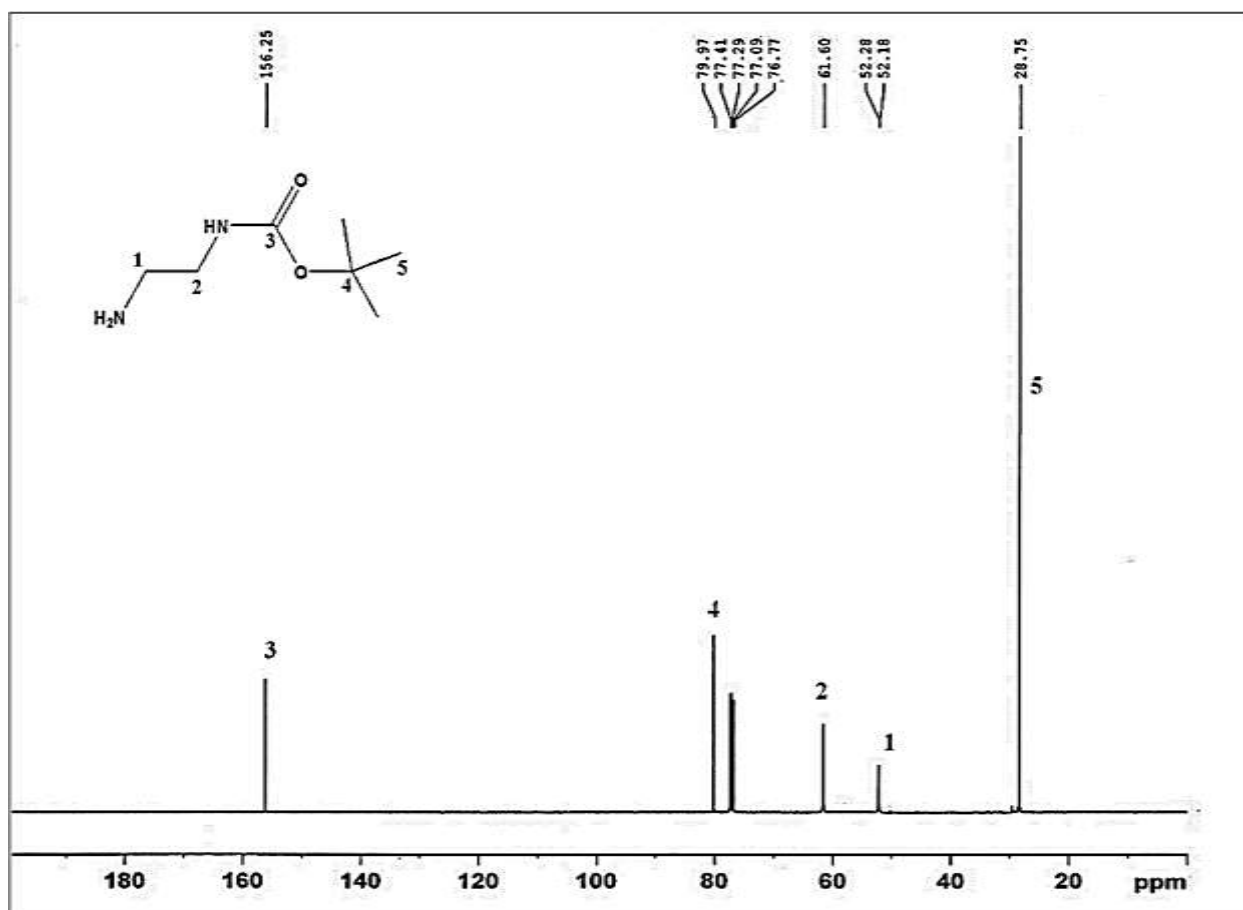


Figure S11. ^{13}C NMR spectrum of *tert*-butyl 2-aminoethylcarbamate (6)

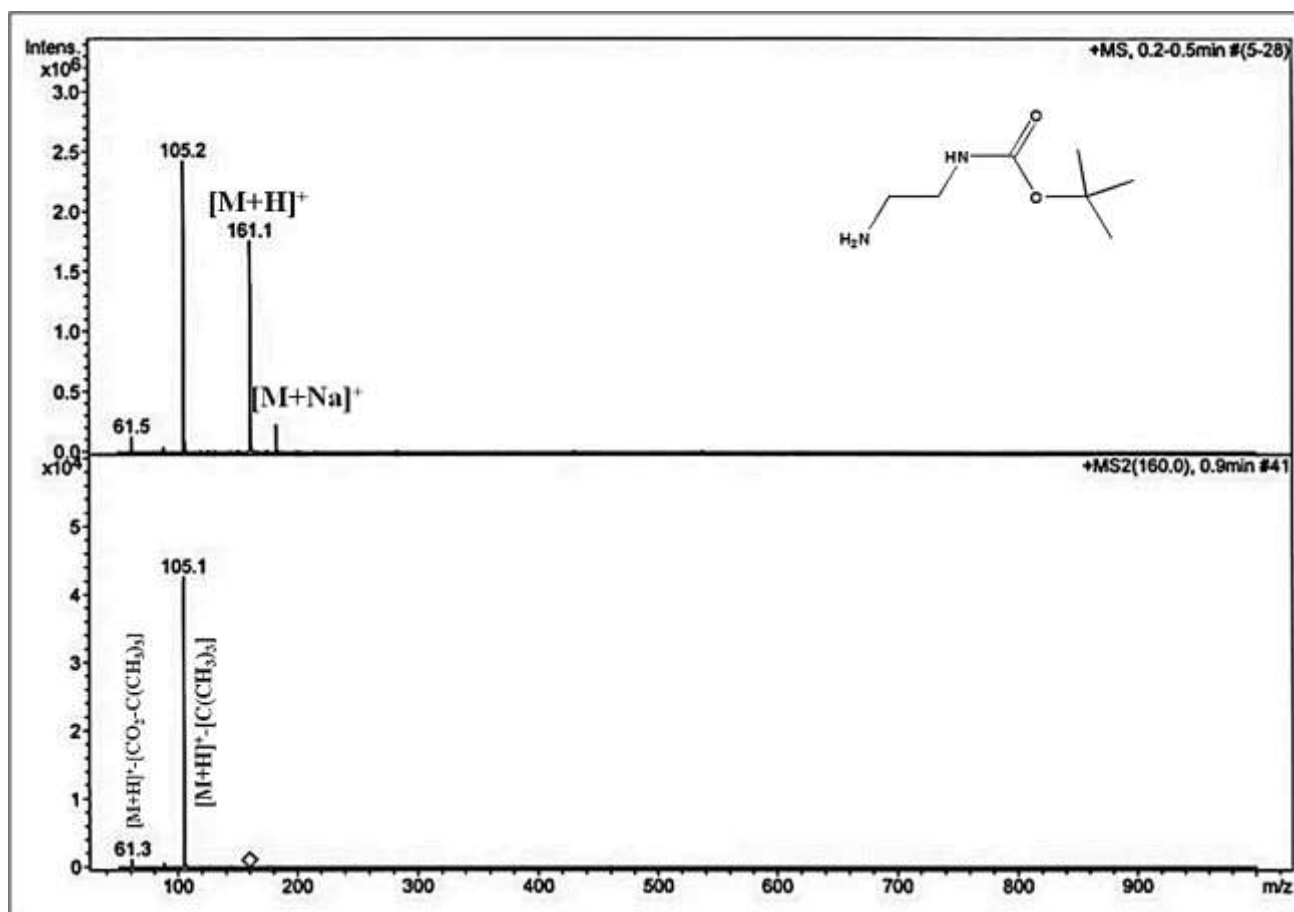


Figure S12. Mass spectrum *tert*-butyl 2-aminoethylcarbamate (**6**)

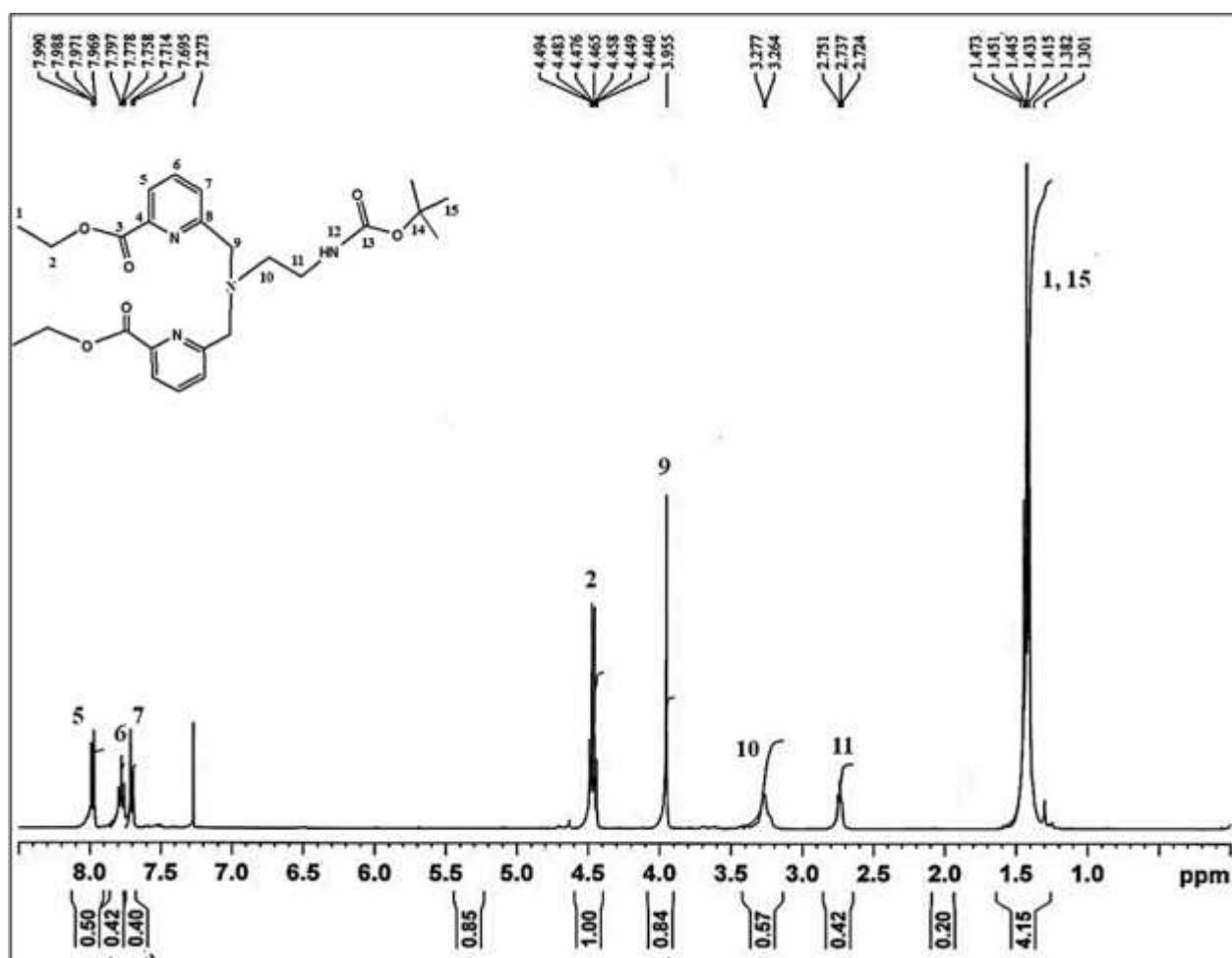


Figure S13. ^1H NMR spectrum of diethyl 6,6'-(2(*tert*-butoxycarbonylamino)ethylazanediyloxy)bis(methylene)dipicolinate (**7**)

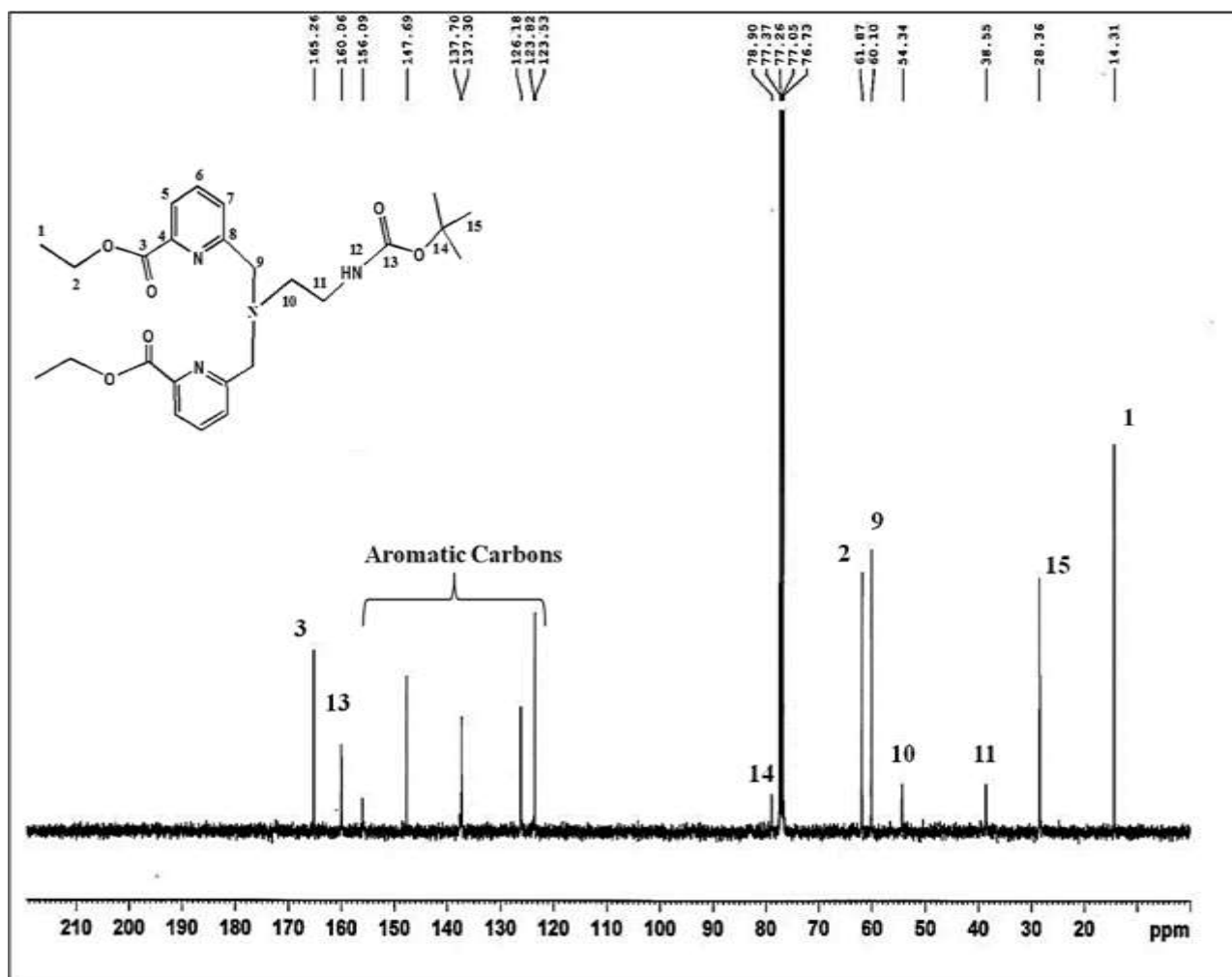


Figure S14. ^{13}C NMR spectrum of diethyl 6,6'-(2(*tert*-butoxycarbonylamino)ethylazanediy)bis(methylene)dipicolinate (**7**)

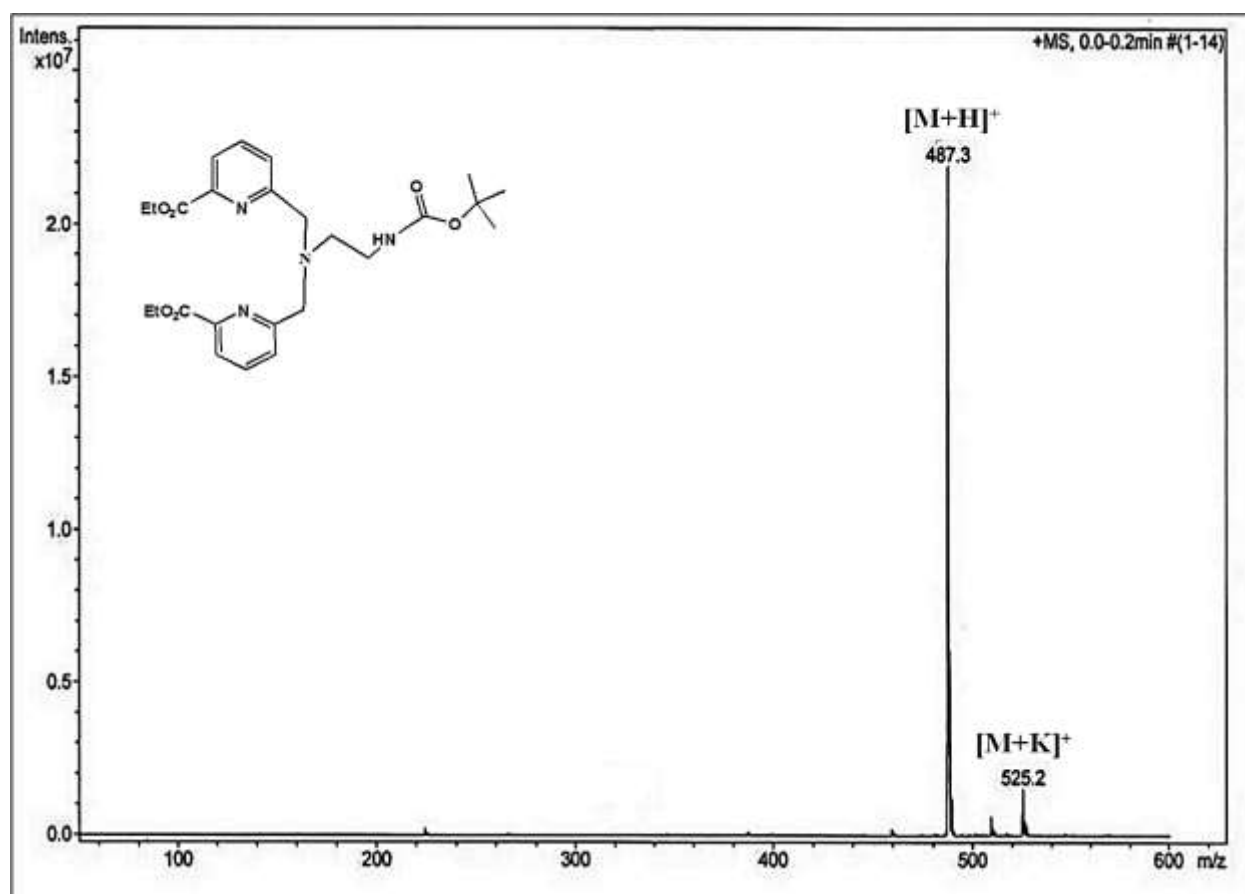


Figure S15. Mass spectrum of diethyl 6,6'-(2(*tert*-butoxycarbonylamino)ethylazanediy)bis(methylene)dipicolinate (**7**)

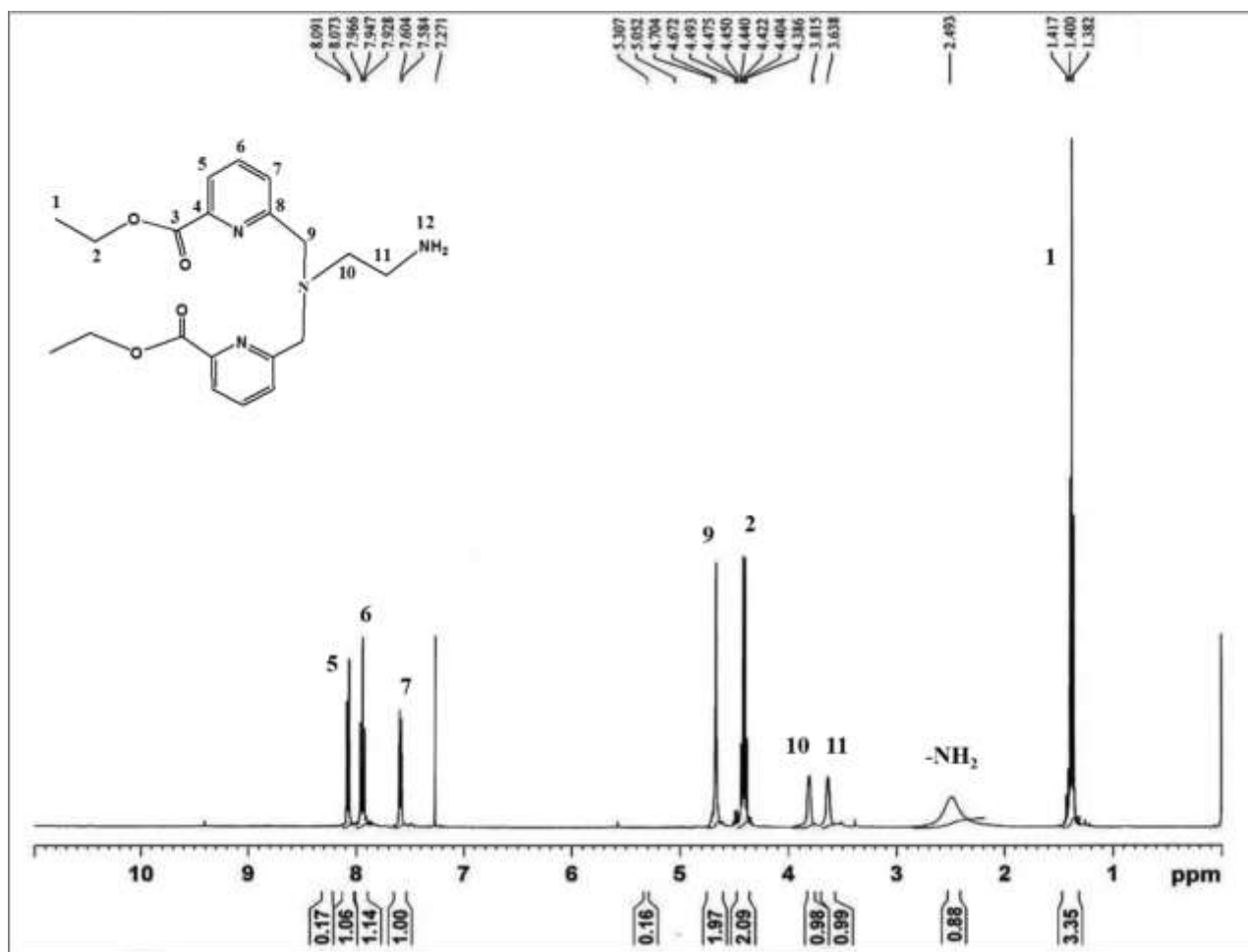


Figure S16. ¹H NMR spectrum of diethyl 6,6'-(2-aminoethylazanediyl)bis(methylene)dipicolinate (**8**)

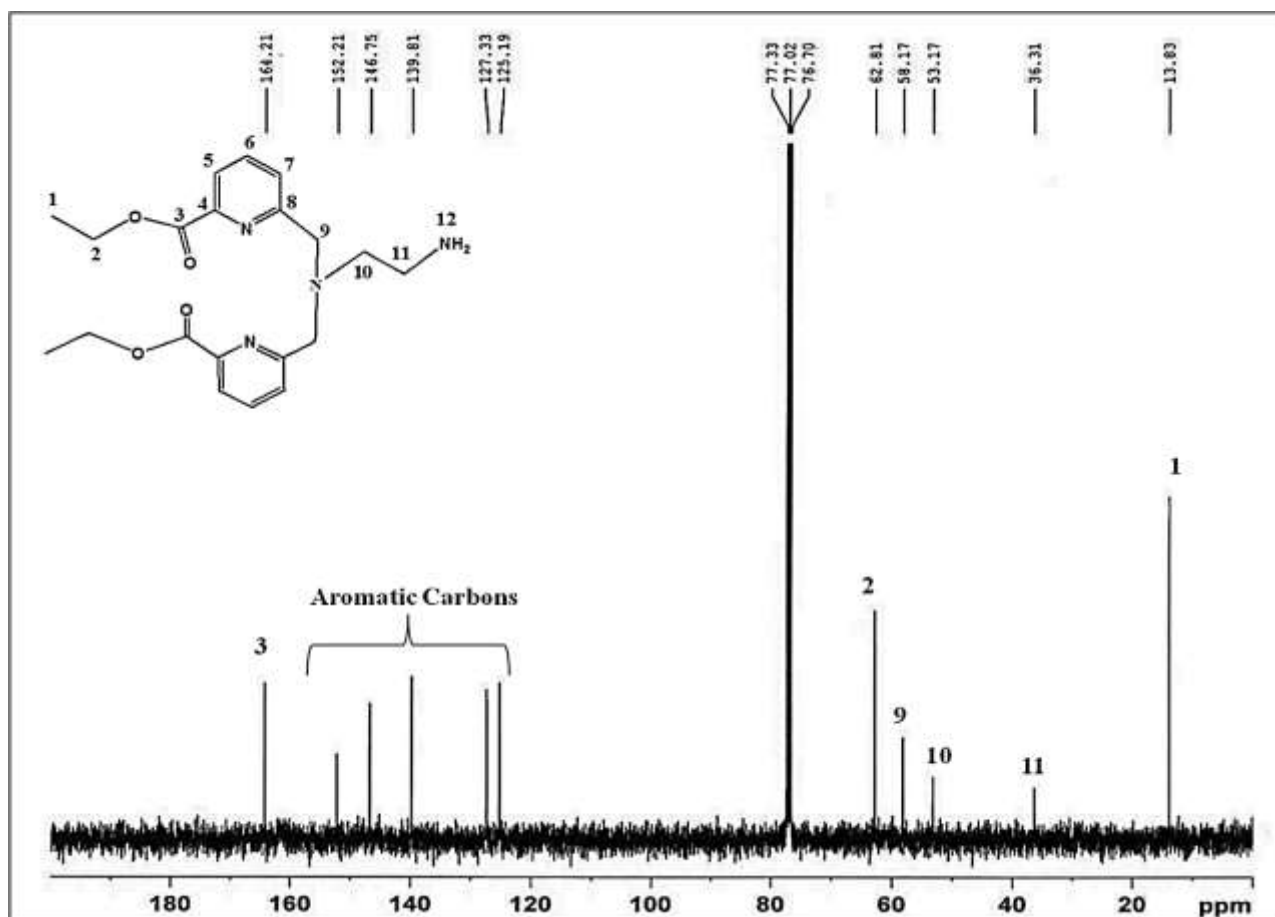


Figure S17. ^{13}C NMR spectrum of diethyl 6,6'-(2-aminoethylazanediyl)bis(methylene)dipicolinate (**8**)

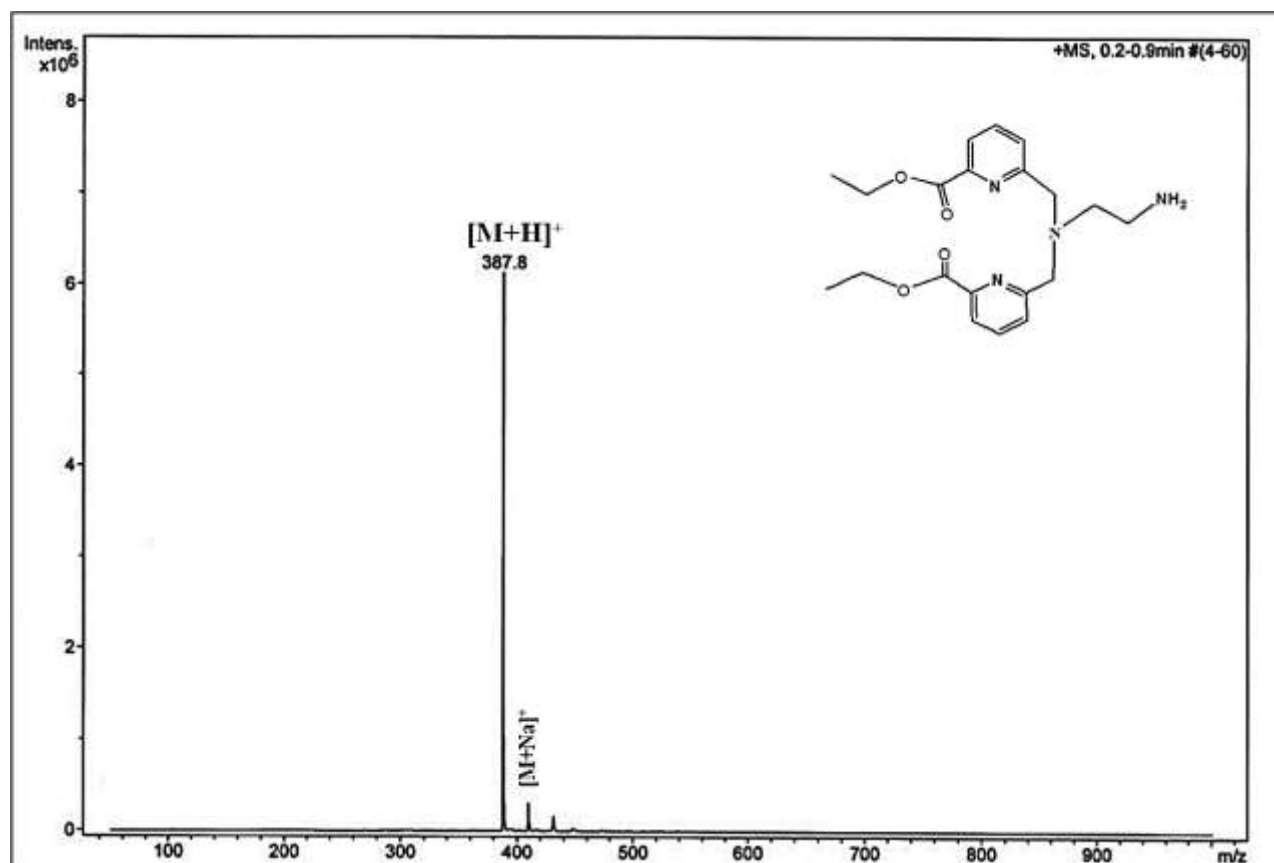


Figure S18. Mass spectrum of diethyl 6,6'-(2-aminoethylazanediy)bis(methylene)dipicolinate (**8**)

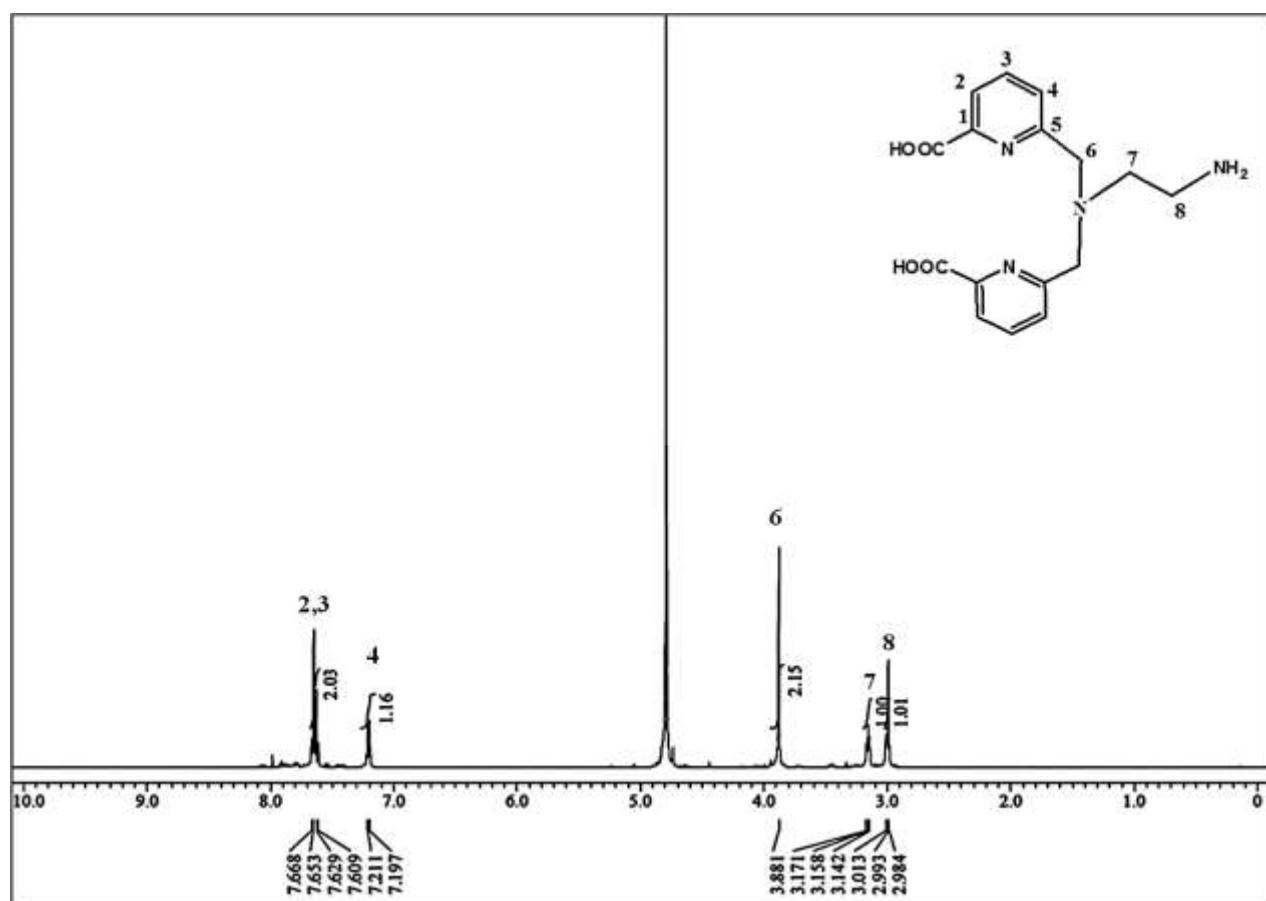


Figure S19. ^1H NMR spectrum of 6,6'-(2-aminoethylazanediyl)bis(methylene)dipicolinic acid ($\text{H}_2\text{pentapa-en-NH}_2$)

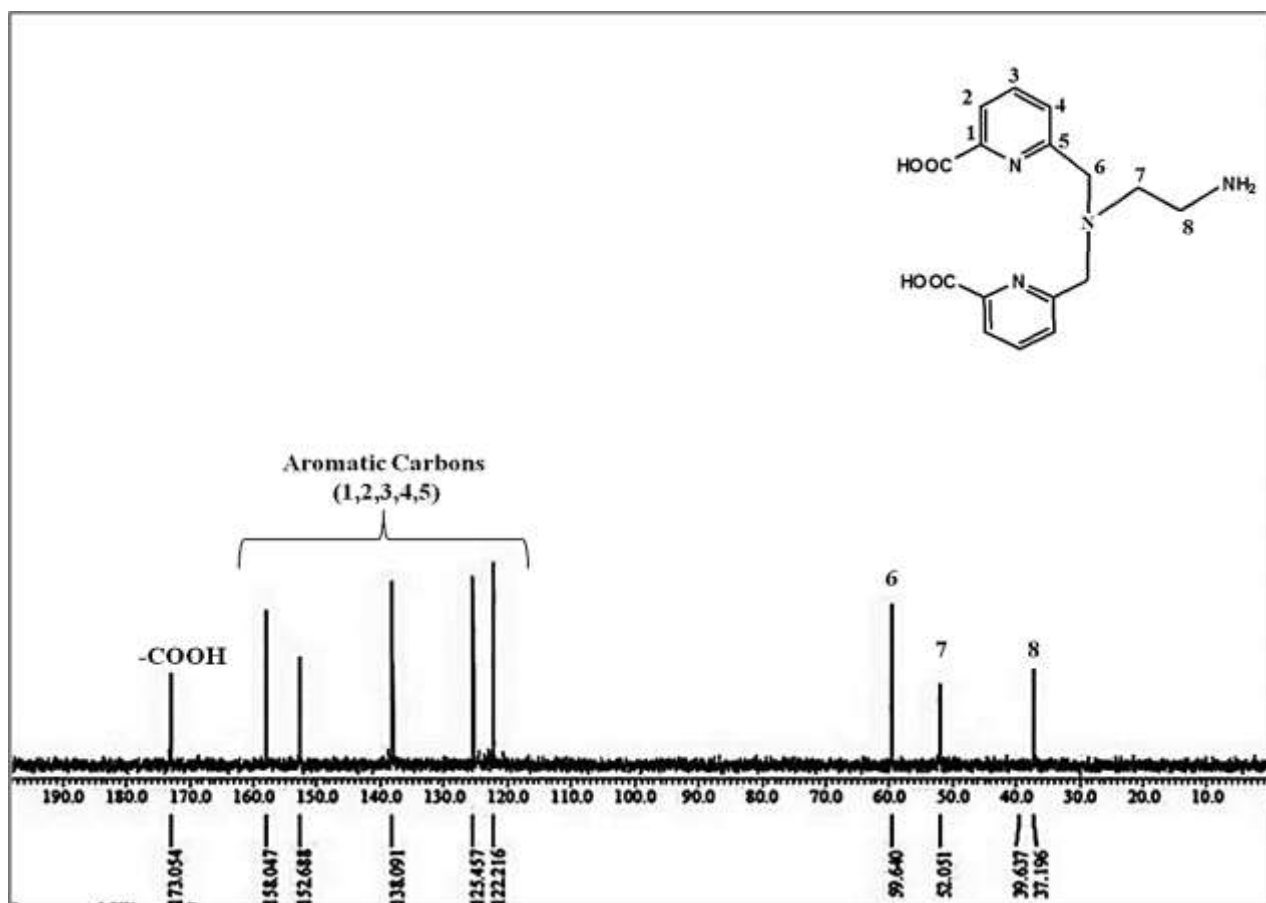


Figure S20. ^{13}C NMR spectrum of 6,6'-(2-aminoethylazanediyl)bis(methylene)dipicolinic acid ($\text{H}_2\text{pentapa-en-NH}_2$)

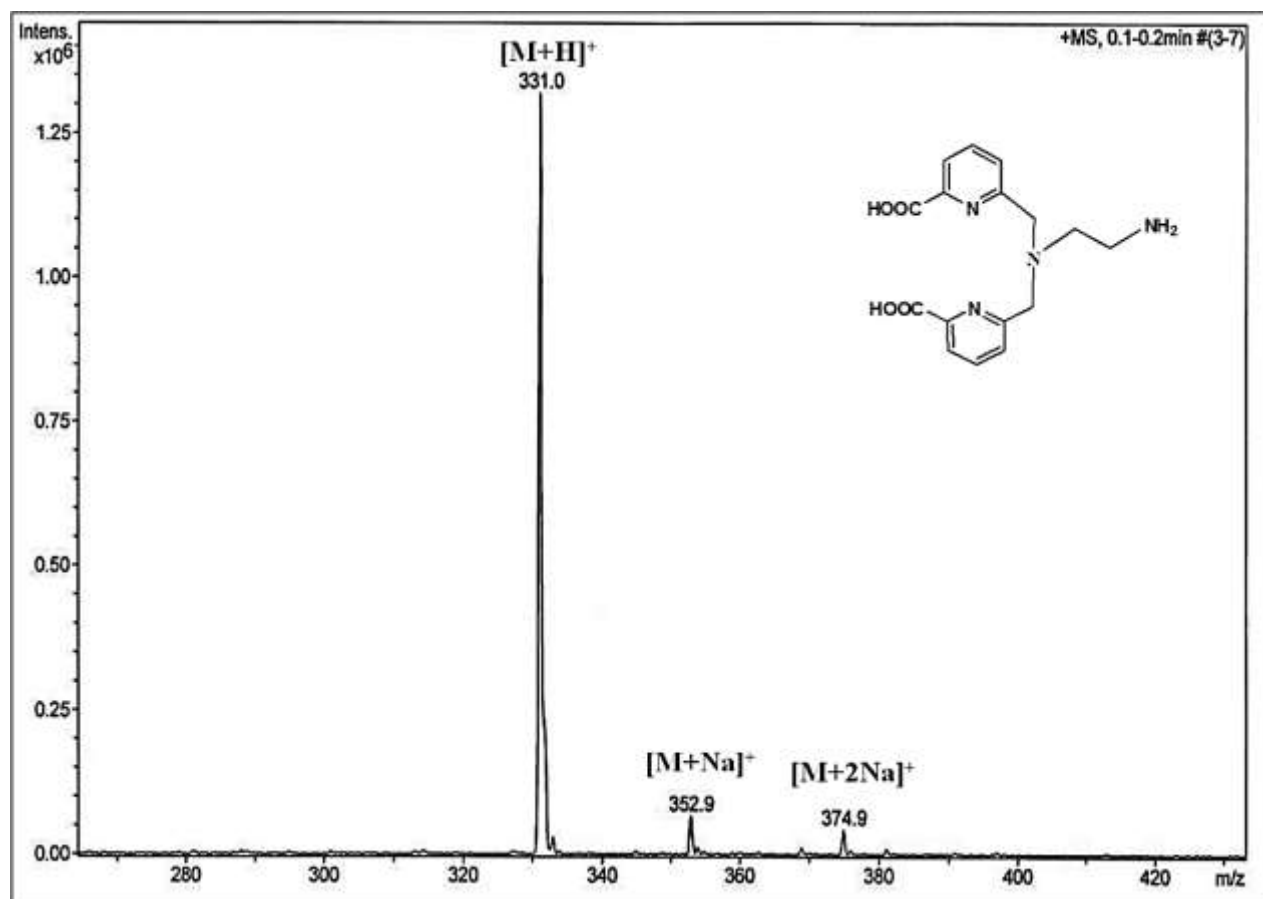


Figure S21. Mass spectrum of 6,6'-(2-aminoethylazanediyl)bis(methylene)dipicolinic acid ($H_2pentapa-en-NH_2$)

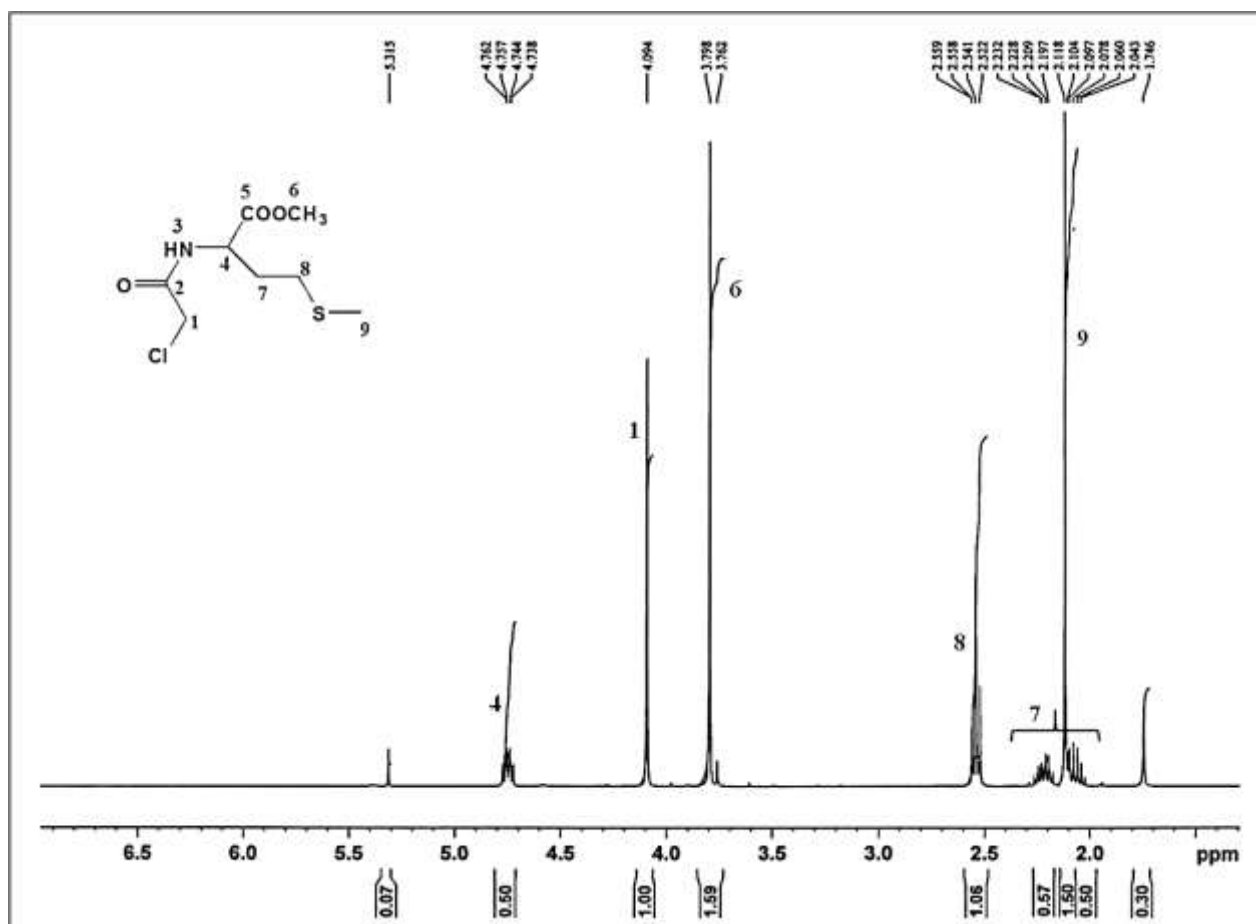


Figure S22. ¹H NMR spectrum of methyl 2-(2-chloroacetamido-4-(methylthio)butanoate (10)

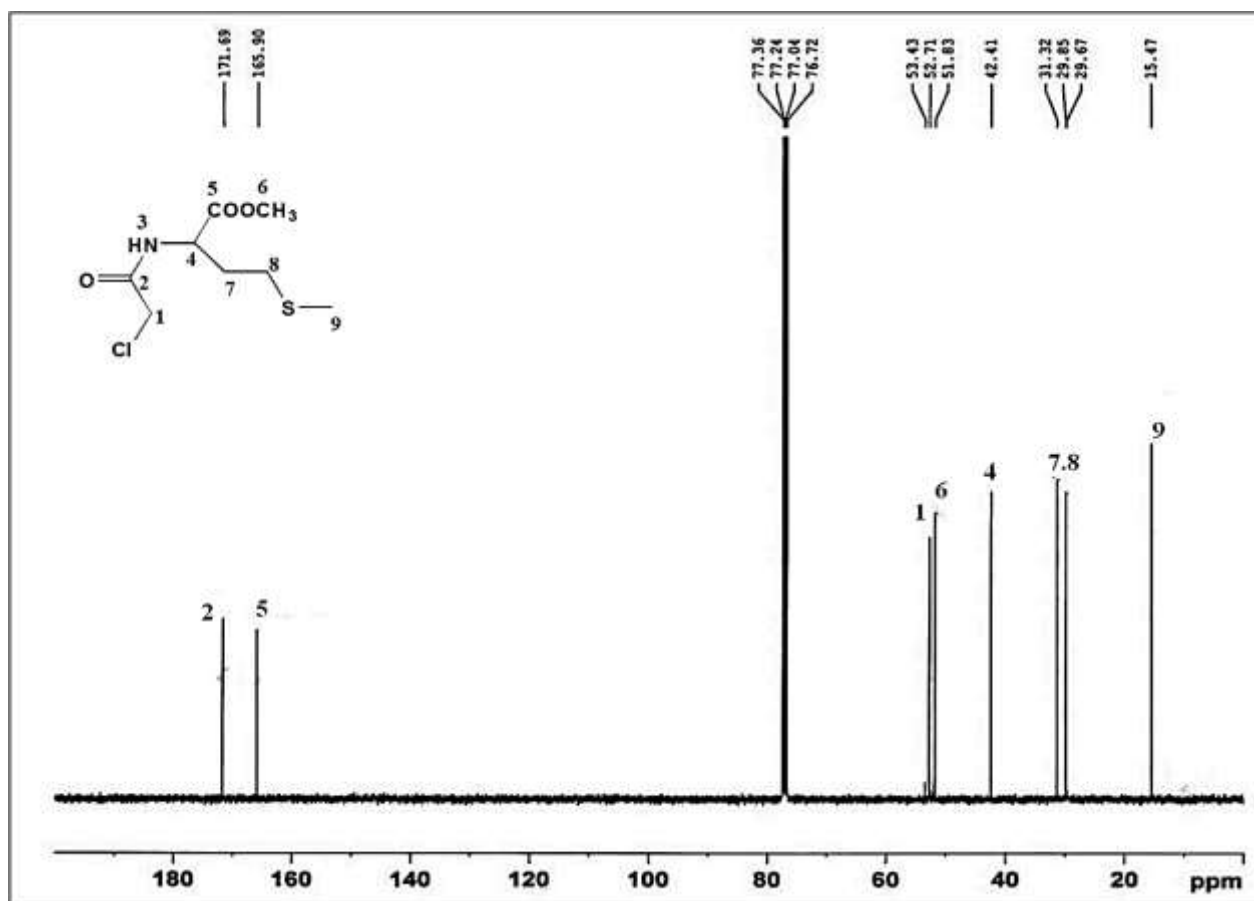


Figure S23. ¹³C NMR spectrum of methyl 2-(2-chloroacetamido-4-(methylthio)butanoate (10)

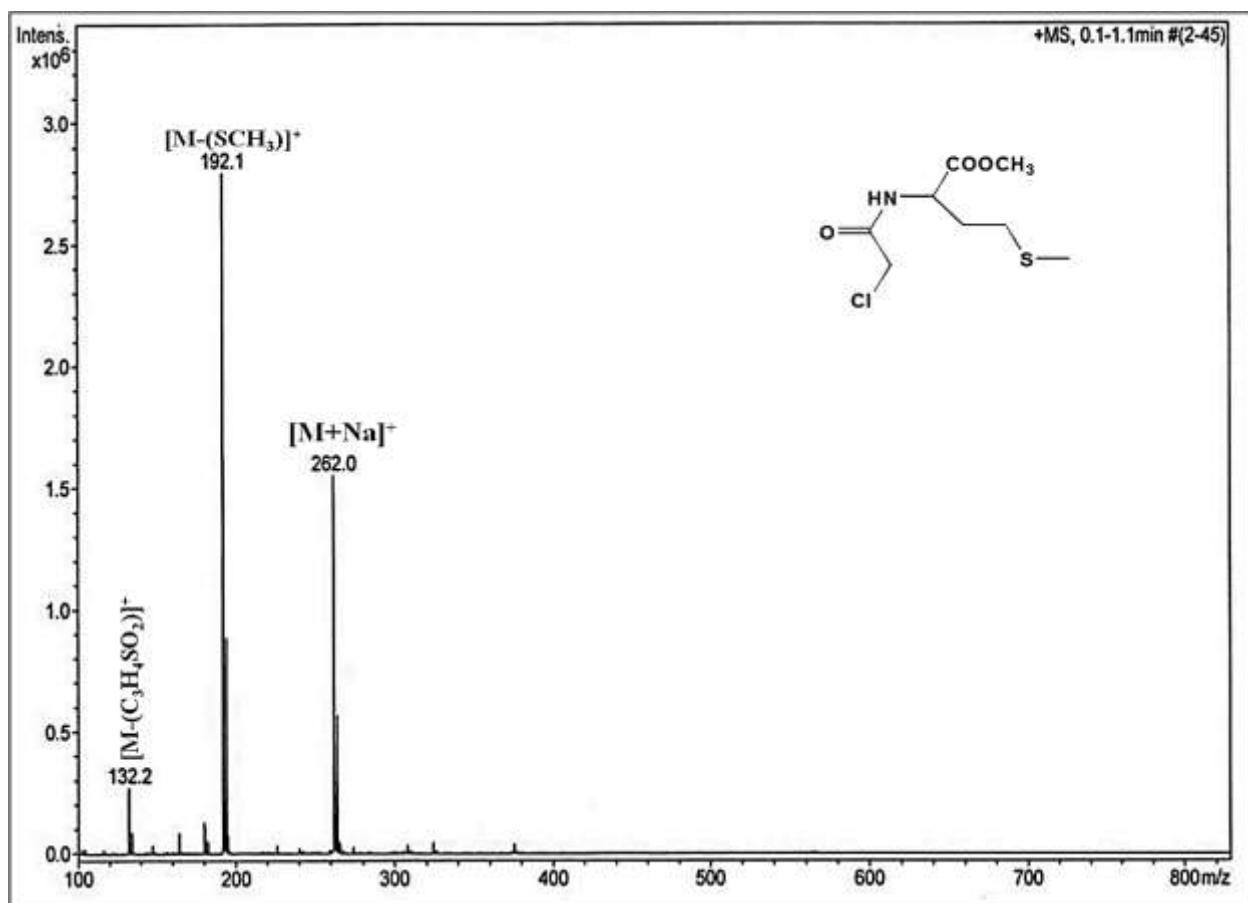


Figure S24. Mass spectrum of methyl 2-(2-chloroacetamido-4-(methylthio)butanoate (**10**)

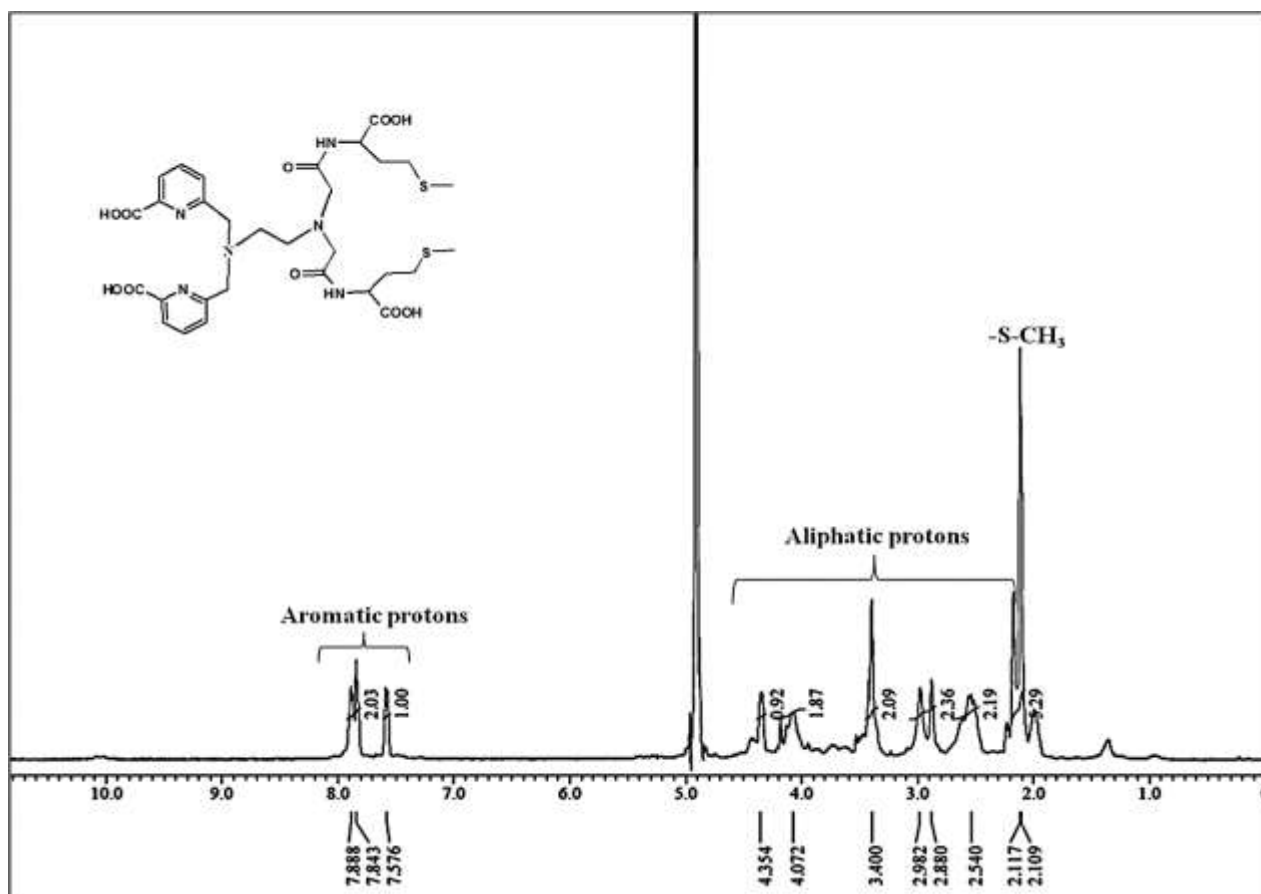


Figure S25. ¹H NMR spectrum of 6-(9-carboxy-5-(1-carboxy-3-(methylthio) propylamino)-2-oxoethyl)-2((6-carboxypyridin-2-yl)methyl)-7-oxo-12thia-2,5,8-triazatridecyl)picolonic acid (H_2 pentapa-en-met₂)

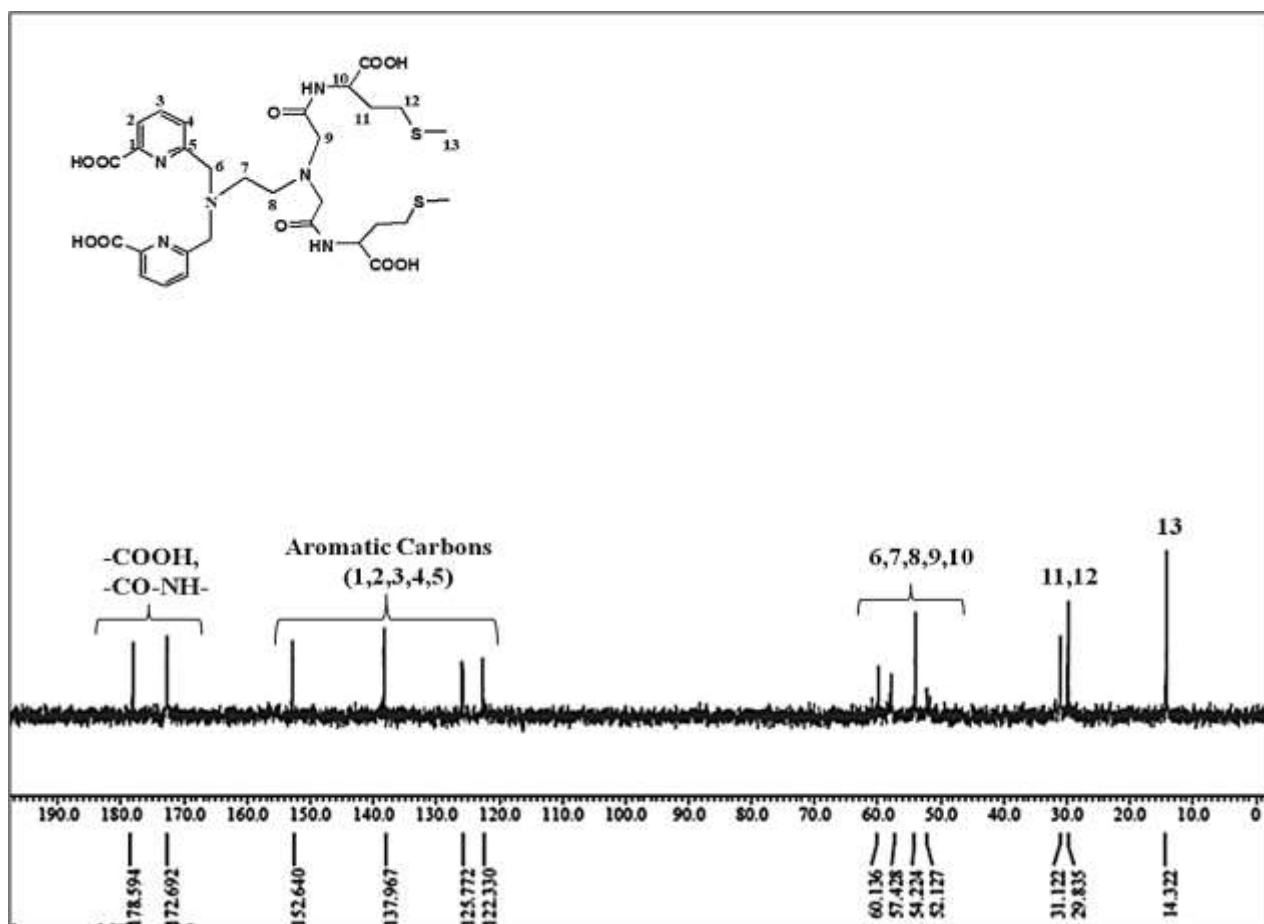


Figure S26. ^{13}C NMR spectrum of 6-(9-carboxy-5-(1-carboxy-3-(methylthio) propylamino)-2-oxoethyl)-2-((6-carboxypyridin-2-yl)methyl)-7-oxo-12thia-2,5,8-triazatridecyl)picolonic acid (**H₂pentapa-en-met₂**)

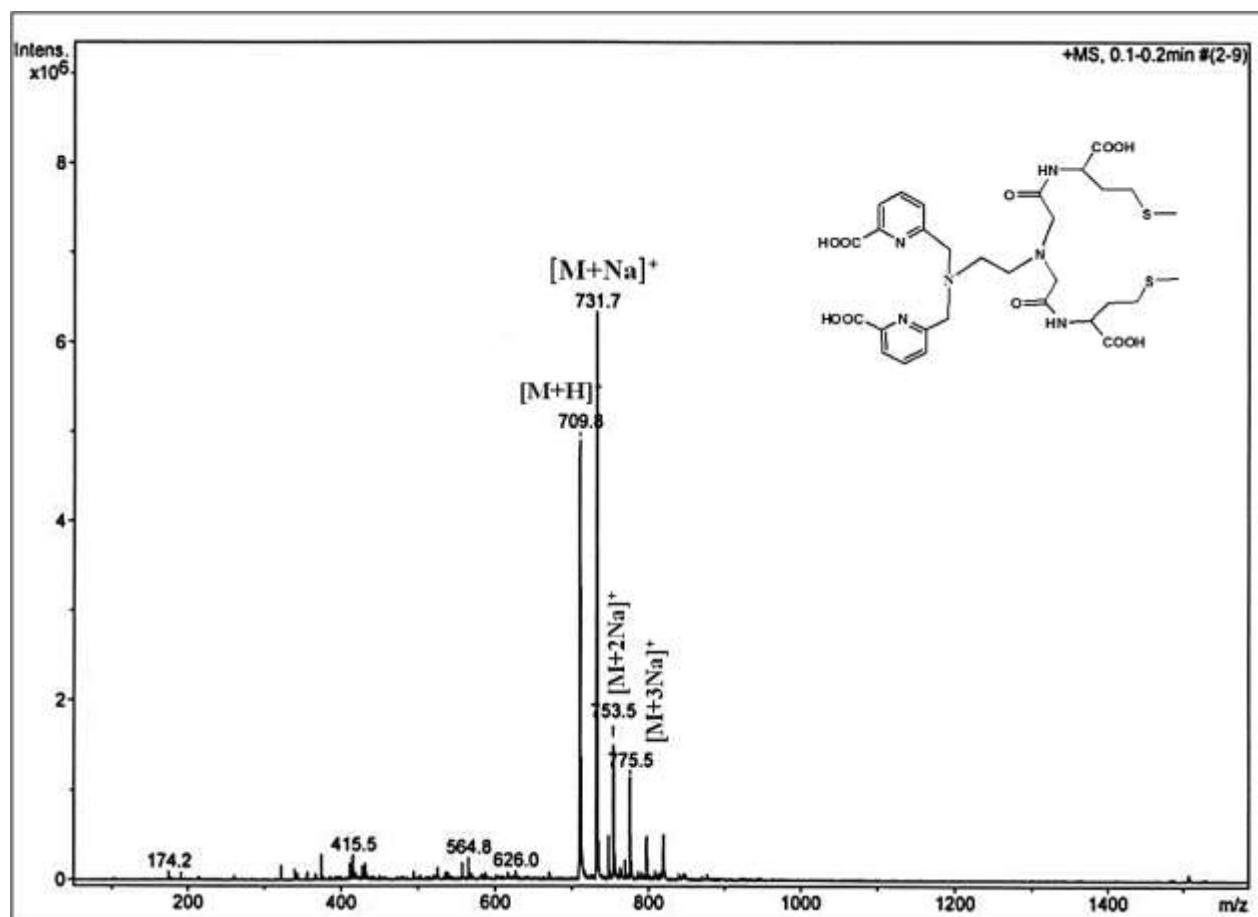


Figure S27. Mass spectrum of 6-(9-carboxy-5-(1-carboxy-3-(methylthio) propylamino)-2-oxoethyl)-2-((6-carboxypyridin-2-yl)methyl)-7-oxo-12thia-2,5,8-triazatridecyl)picolonic acid (**H₂pentapa-en-met₂**)

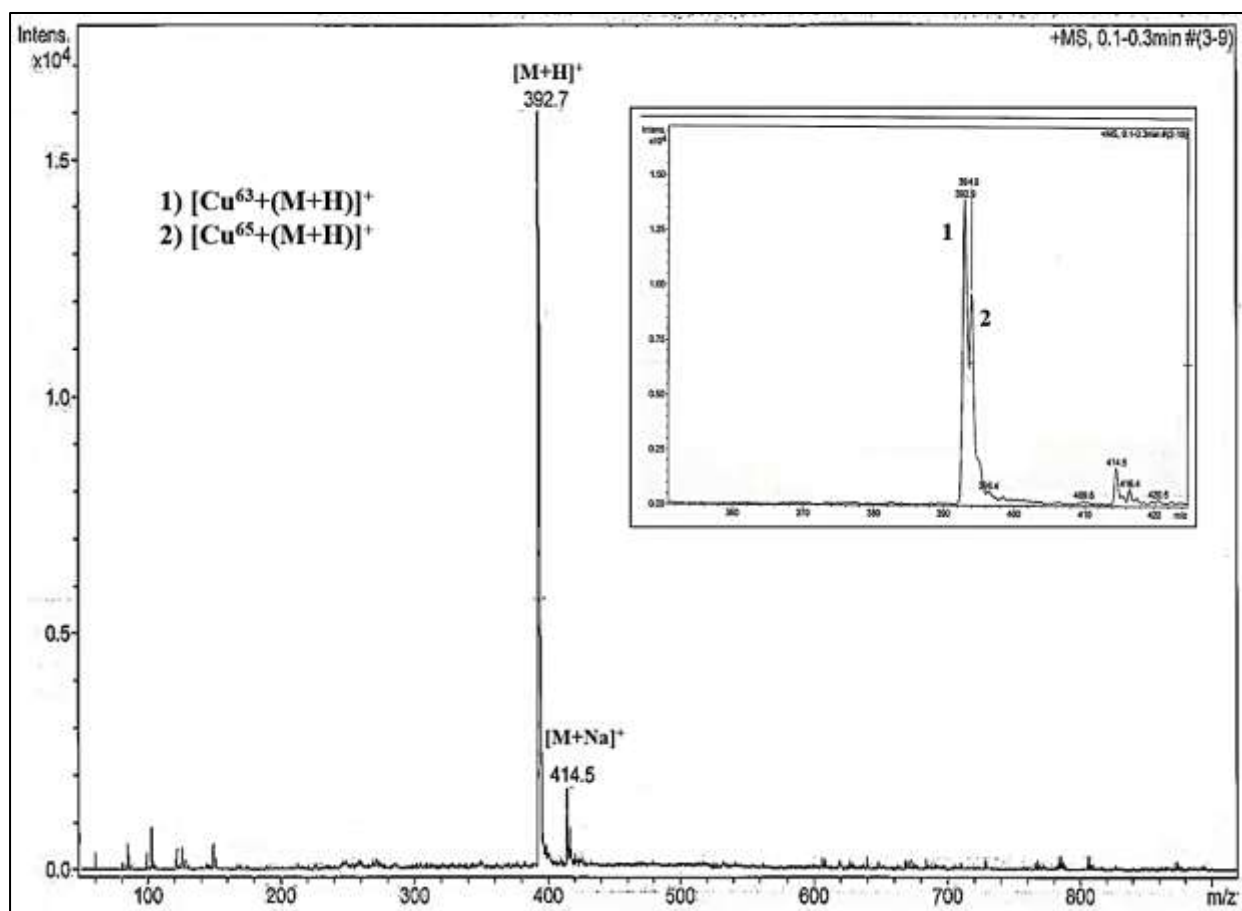


Figure S28. Mass spectrum of $\text{Cu-H}_2\text{pentapa-en-NH}_2$

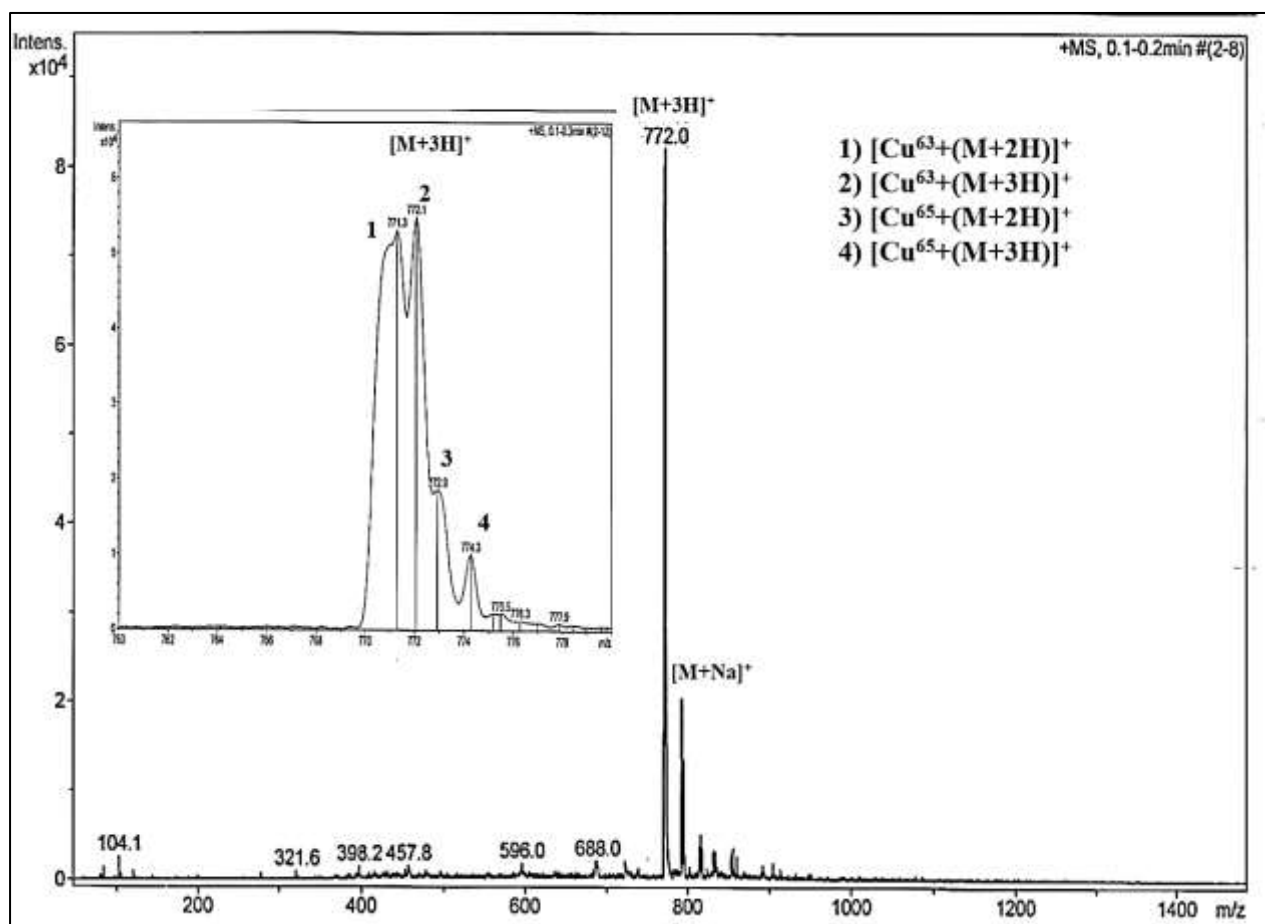


Figure S29. Mass spectrum of $\text{Cu-H}_2\text{pentapa-en-met}_2$

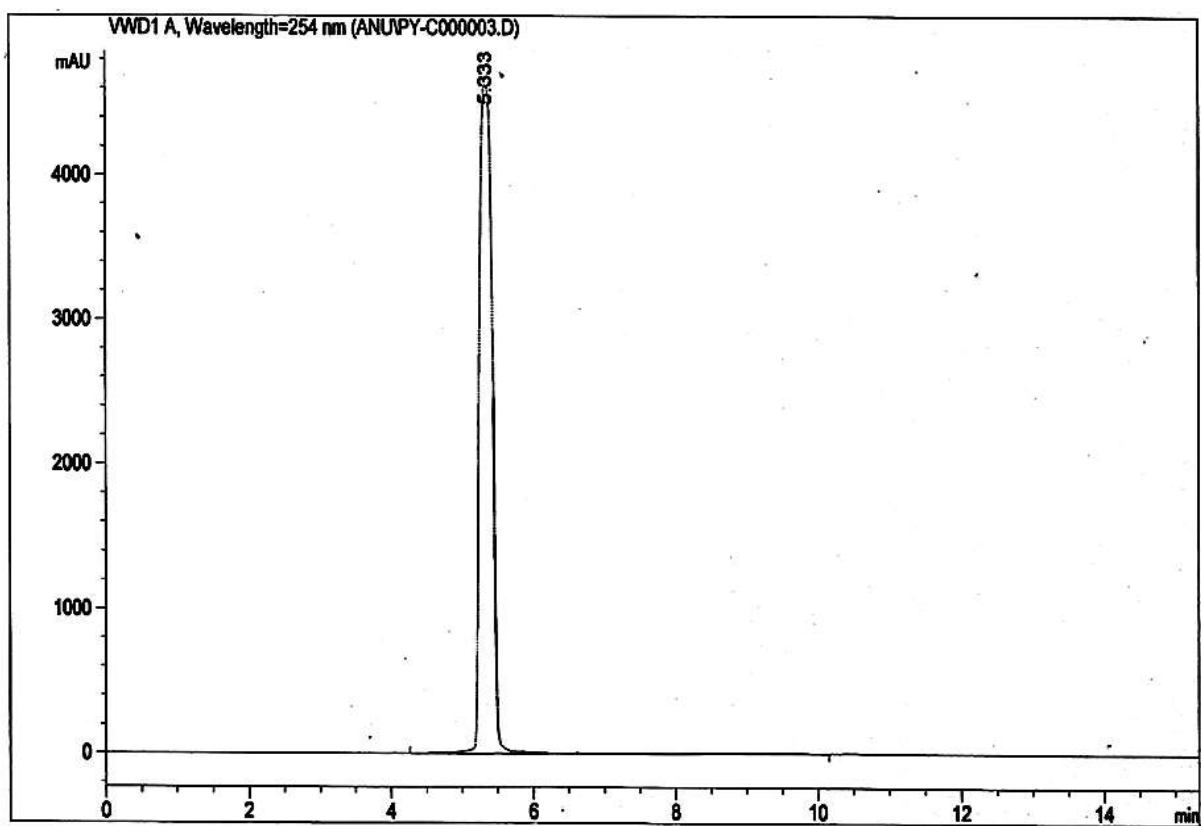


Figure S30. HPLC Profile of $\text{H}_2\text{pentapa-en-NH}_2$

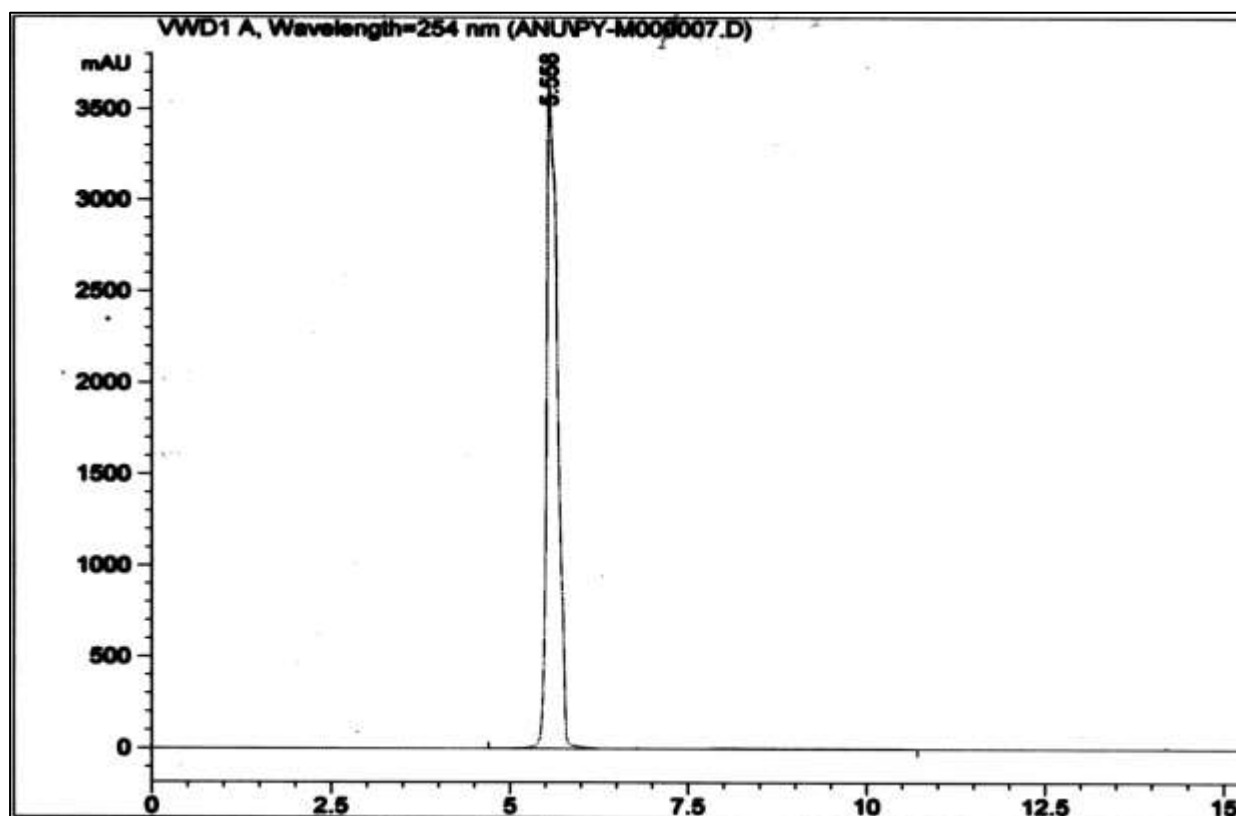


Figure S31. HPLC Profile of $\text{H}_2\text{pentapa-en-met}_2$

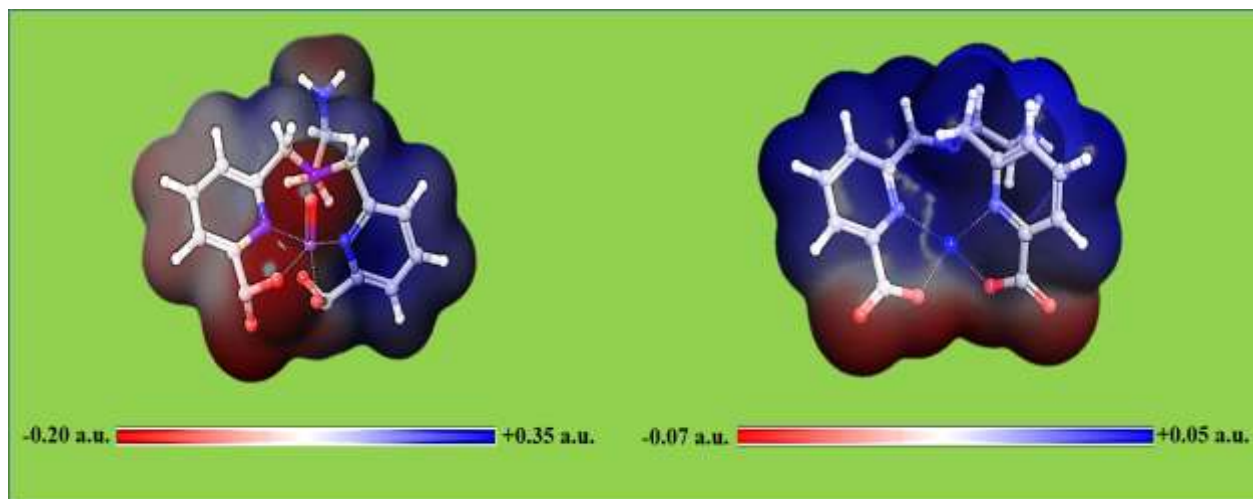


Figure S32. *In silico* DFT predicted electrostatic potential map of $[\text{ReO}(\text{pentapa-en-NH}_2)]^+$ and $\text{Cu-pentapa-en-NH}_2$ varying from -0.20 a.u. to +0.35 a.u. and -0.07 a.u. to +0.05 a.u. respectively

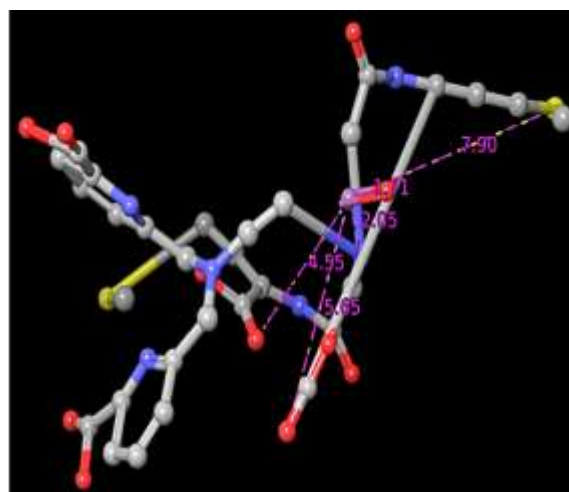
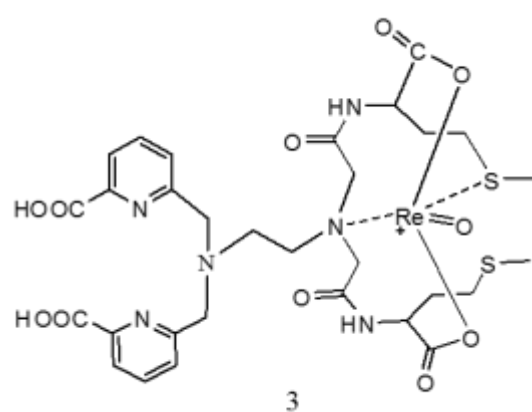
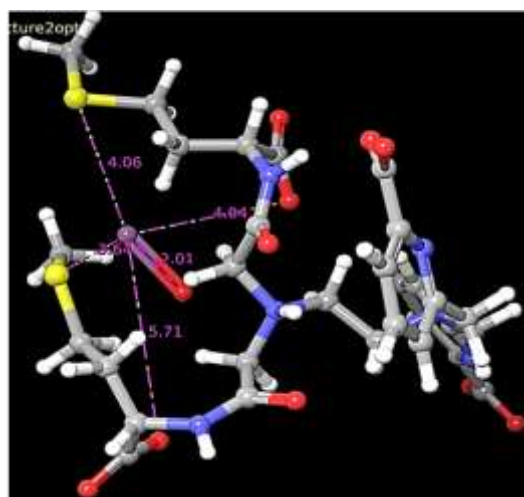
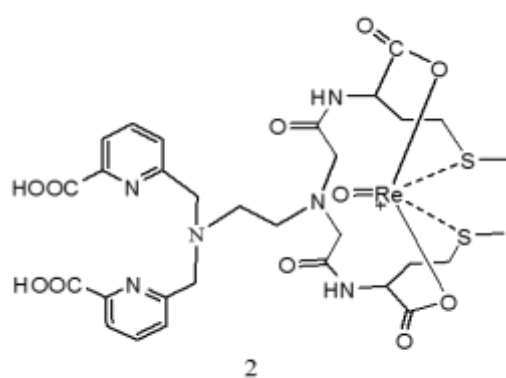
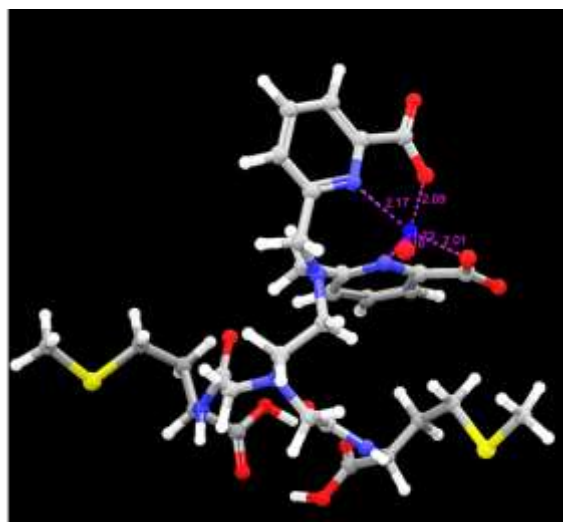
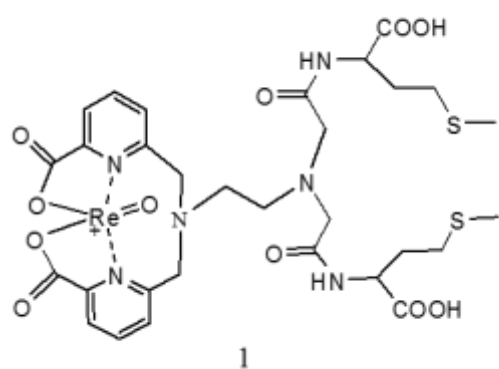


Figure S33. DFT studies showing the bond lengths after coordination of Rhenium oxocore to $\text{H}_2\text{pentapa-en-met}_2$

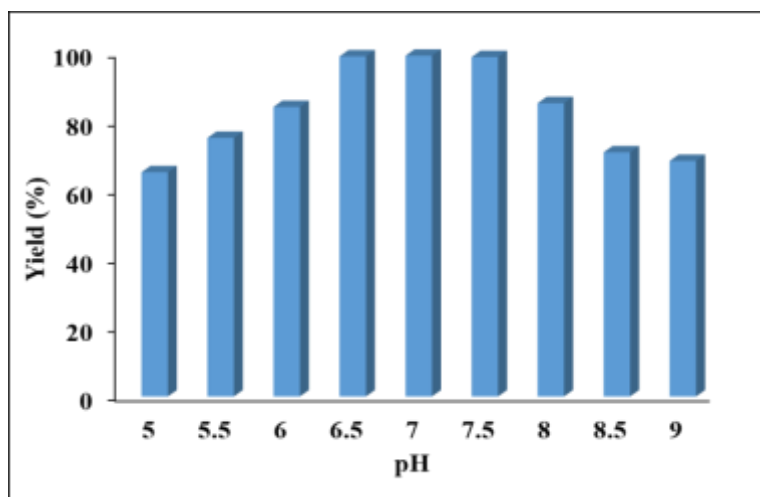


Figure S34. Effect of pH on radio chemical yield of H₂pentapa-en-NH₂ and H₂pentapa-en-met₂ with ^{99m}Tc

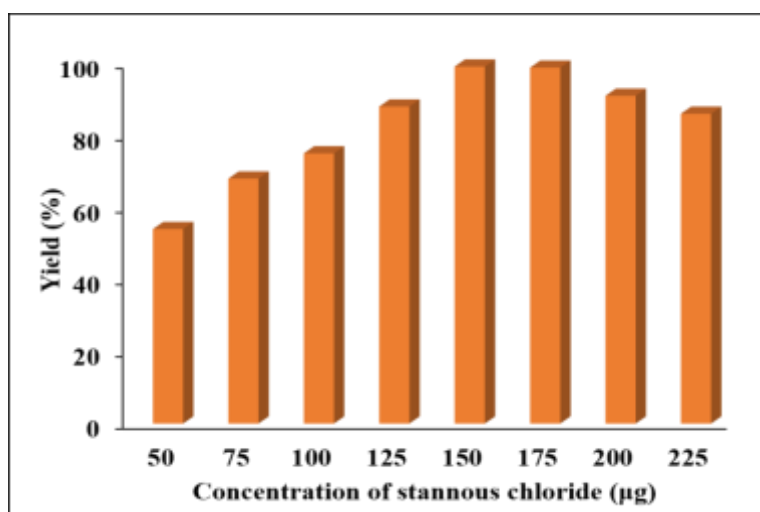


Figure S35. Effect of stannous chloride concentration on radio chemical yield of H₂pentapa-en-NH₂ and H₂pentapa-en-met₂ with ^{99m}Tc

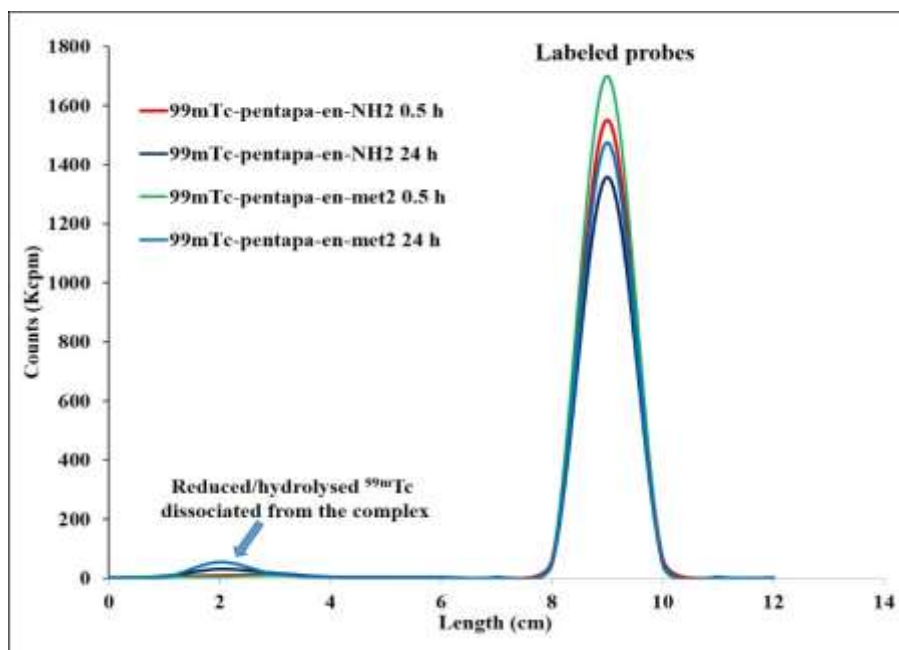


Figure S36. ITLC profile of ^{99m}Tc complexes of H₂pentapa-en-NH₂ and H₂pentapa-en-met₂ at various time intervals in PBS buffer using pyridine/acetic acid/water (3:5:1.5) as mobile phase

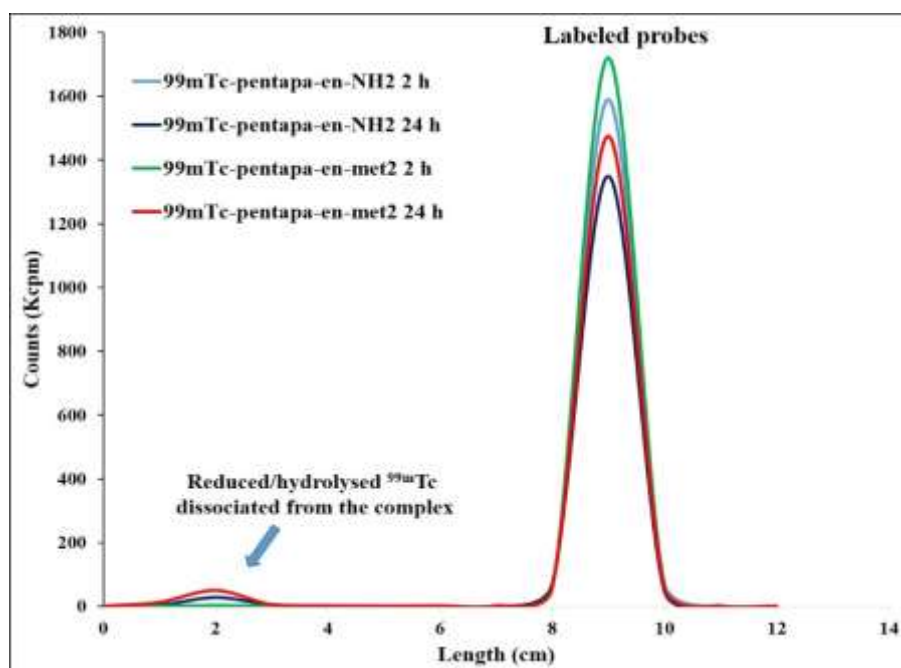


Figure S37. ITLC profile of ^{99m}Tc complexes of H₂pentapa-en-NH₂ and H₂pentapa-en-met₂ at various time intervals in human serum using pyridine/acetic acid/water (3:5:1.5) as mobile phase

Table S1. Comparison of bond lengths obtained through DFT studies after coordination of rhenium oxocore to penatapa-en-met₂ with bond lengths of reported XRD crystal structure of methionine conjugated rhenium oxocore.

	Re–O (Re=O)	Re–O (Re–O–C=O)	Re–N (Re–NH ₂)	Re–S
Met-Re [ReOX ₂ (Met)]	1.66 Å	2.05 Å	2.14 Å (Re–NH ₂)	2.42 Å
ReO[penatapa-en-met ₂] ⁺ (1)	1.72 Å	2.01 Å, 2.09 Å	2.16 Å, 2.17 Å	-
ReO[penatapa-en-met ₂] ⁺ (2)	2.01 Å	4.04 Å, 5.71 Å	-	4.06 Å, 3.64 Å
ReO[penatapa-en-met ₂] ⁺ (3)	1.71 Å	4.55 Å, 5.65 Å	2.65 Å	7.90 Å

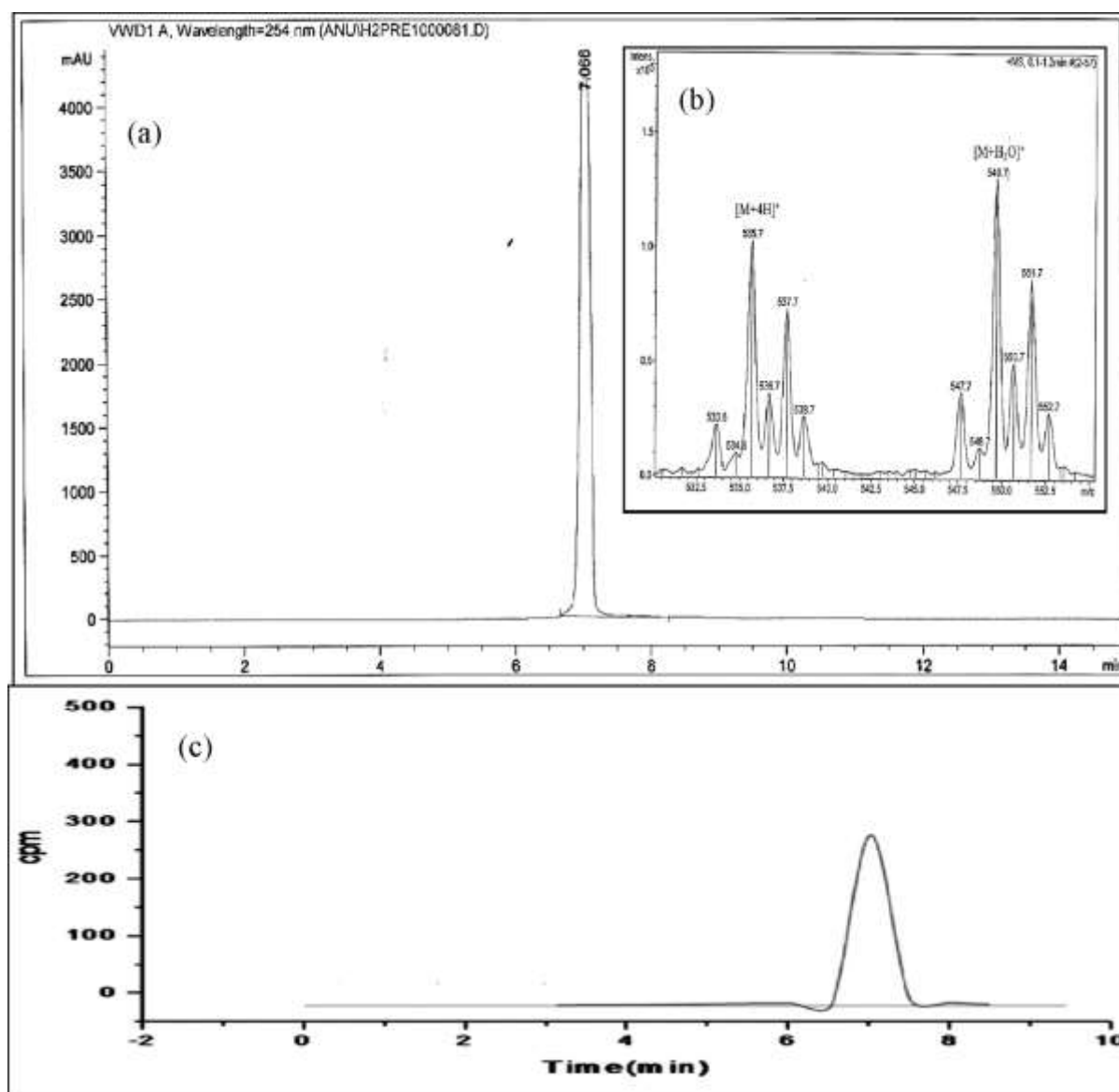


Figure S38. (a) HPLC profile and (b) mass spectrum of Re-pentapa-en-NH₂; (c) HPLC profile (γ - detector) of ^{99m}Tc-pentapa-en-NH₂

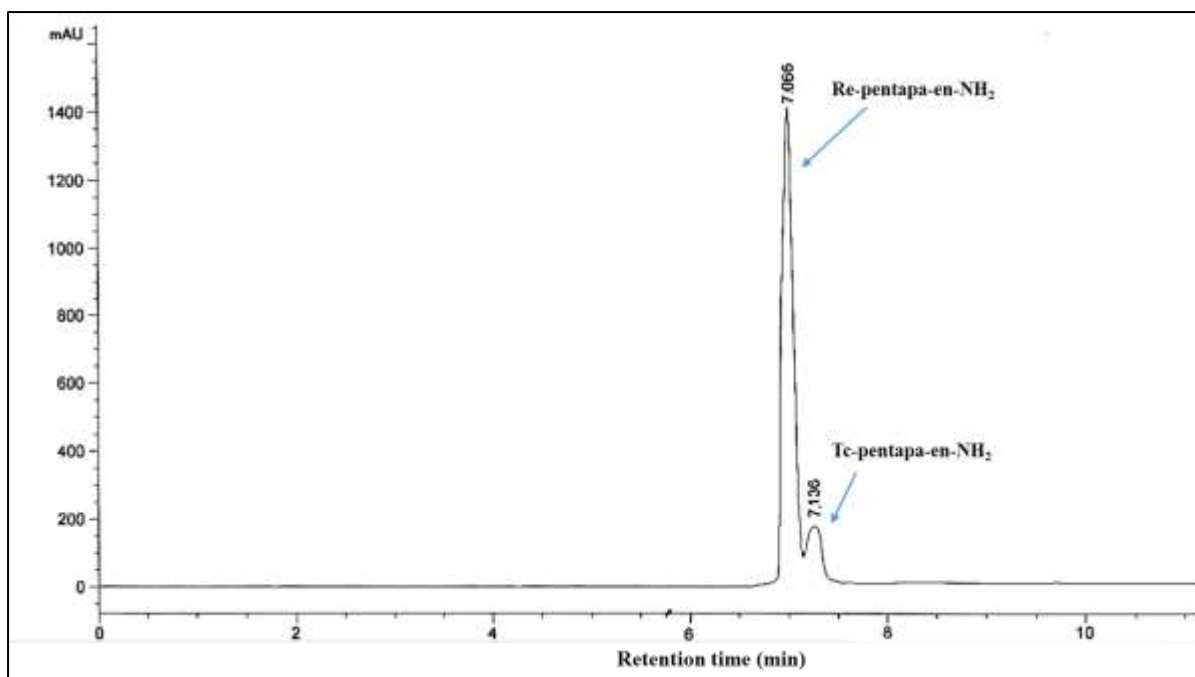


Figure S39. HPLC profile of co-injected metal complexes of Re-pentapa-en-NH₂ and Tc-pentapa-en-NH₂

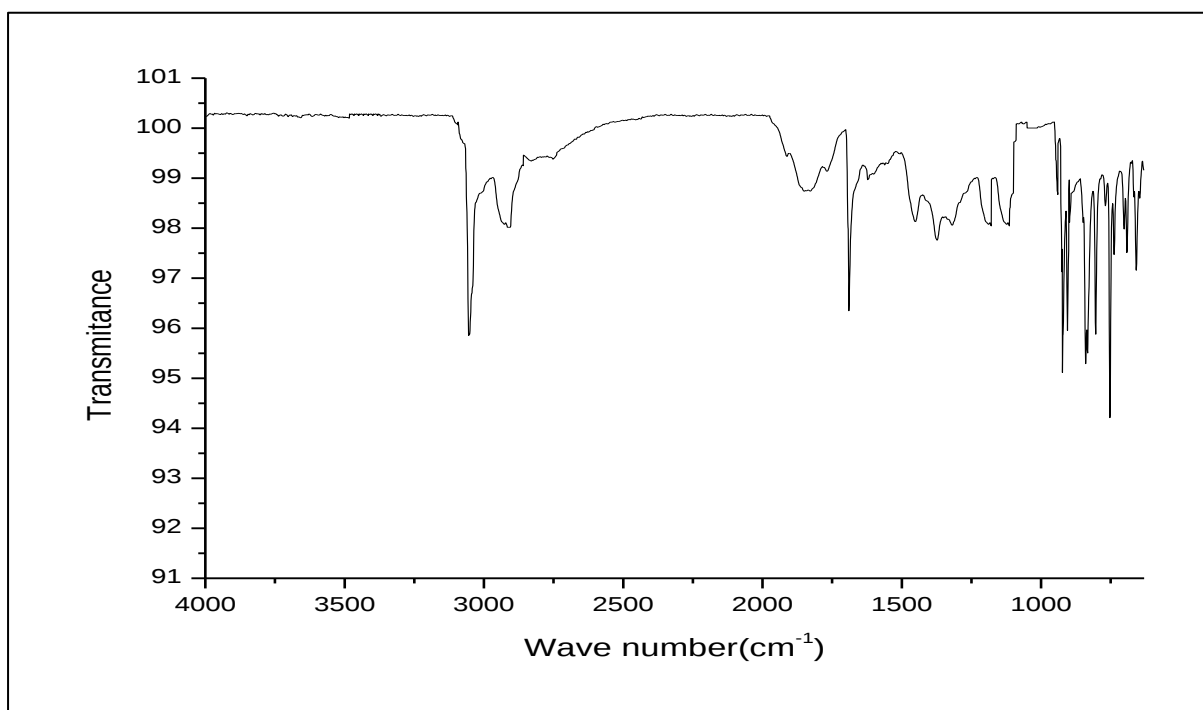


Figure S40. IR spectrum of Re-pentapa-en-NH₂