

Supplementary Materials:

Indolizine-based scaffolds as efficient and versatile tools – Application to the synthesis of biotin-tagged anti-angiogenic drugs

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Figure S1: Stability of the COB223/236 modified agarose S2

Table S1. Affinity chromatography-proteomics data. S3

Figures S2-S18: NMR-HPLC data S5

Stability of the COB223/236 modified agarose.

In 2 ml Eppendorf vials, 0.5 mL of the modified agaroses were diluted with 0.5 mL of the buffer used for the affinity chromatography (Tris 20 mM-HCl buffer pH 7.5 supplemented with 0.075M NaCl and 0.05% Triton-X100) and the resulting suspensions were shaken in a thermal shaker at 37°C for 72h or at 70°C for 72h. The suspensions were then centrifuged and the supernatants were separated. The different fractions were irradiated at 365 nm to reveal the liberation of the fluorescent molecules (COB223- or COB236-indolizines). The results of the heating at 70°C are shown in Fig. 1 by comparison with solutions of COB223/236-biotins.

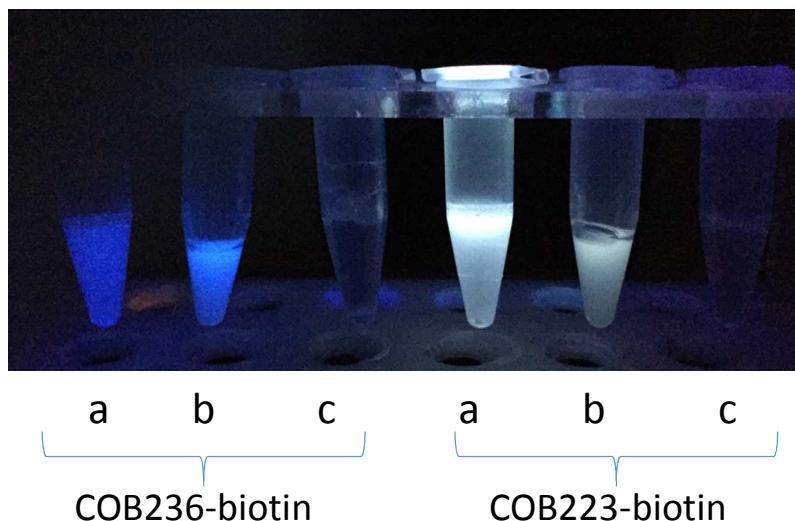


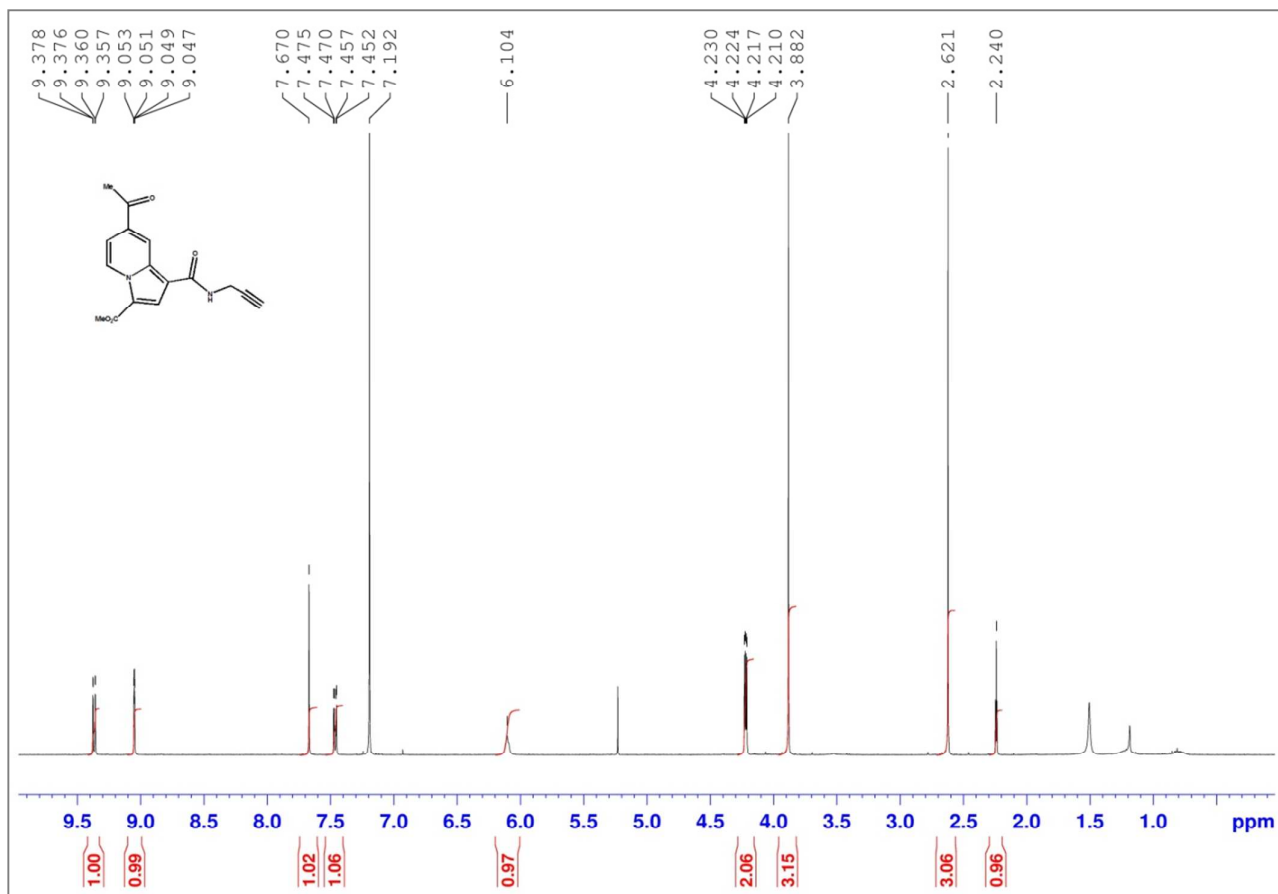
Figure S1. The vials were irradiated at 365 nm using a portable UV/vis lamp. a: solutions of COB223- or CO236-biotin in Tris 20 mM-HCl buffer pH 7.5; b: COB223- or COB236-modified agaroses after 3 days at 70°C in Tris 20 mM-HCl buffer pH 7.5; c: supernatants separated by centrifugation of the suspensions of COB223- or COB236-modified agarose stirred 3 days at 70°C in Tris 20 mM-HCl buffer pH 7.5

COB223/COB236 enrichment fold	Protein name	Gene Symbol
17	Proliferation-associated protein 2G4	PA2G4
15	Cysteine and glycine-rich protein 2	CSRP2
7	Polypyrimidine tract-binding protein 1	PTBP1
6	Heterogeneous nuclear ribonucleoprotein M	HNRNPM
6	Phosphatidylinositol 5-phosphate 4-kinase type-2 alpha	PIP4K2A
6	Serine/threonine-protein kinase 10	STK10
6	Protein lin-28 homolog B	LIN28B
5.5	Ribonuclease 4	RNASE4
5	cDNA FLJ77858, highly similar to Homo sapiens N-acylsphingosine amidohydrolase (acid ceramidase) 1 (ASAH1)	
5	LanC-like protein 1	LANCL1
5	Hypoxia up-regulated protein 1	HYOU1
5	Glutamine--fructose-6-phosphate aminotransferase [isomerizing] 1	GFPT1
5	Ubiquitin-conjugating enzyme E2 N	UBE2N
5	Complement component C6	C6
5	Poly(rC)-binding protein 1	PCBP1
COB223 only	Growth/differentiation factor 2	GDF2
COB223 only	Vigilin	HDLBP
COB223 only	40S ribosomal protein S20	RPS20
COB223 only	Calponin-1	CNN1
COB223 only	Pleiotropic regulator 1	PLRG1
COB223 only	DnaJ homolog subfamily B member 1	DNAJB1
COB223 only	Malignant T-cell-amplified sequence 1	MCTS1
COB223 only	Putative uncharacterized protein DKFZp686I04196 (Fragment)	DKFZp686I04196
COB223 only	Myosin-2	MYH2
COB223 only	Myosin-10	MYH10
COB223 only	Glutathione S-transferase kappa 1	GSTK1
COB223 only	Nuclear transport factor 2	NUTF2
COB223 only	RNA-binding protein Raly	RALY
COB223 only	EF-hand domain-containing protein D1	EFHD1
COB223 only	Extended synaptotagmin-2	ESYT2
COB223 only	Inorganic pyrophosphatase	PPA1
COB223 only	Glutathione S-transferase omega-1	GSTO1
COB223 only	26S protease regulatory subunit 7	PSMC2
COB223 only	IBM-A3 heavy chain variable region (Fragment)	
COB223 only	Aspartate aminotransferase, cytoplasmic	GOT1
COB223 only	Cysteine-rich secretory protein LCCL domain-containing 2	CRISPLD2
COB223 only	Syntaxin-5	STX5
COB223 only	Polyadenylate-binding protein 1	PABPC1
COB223 only	40S ribosomal protein S14	RPS14
COB223 only	Single-stranded DNA-binding protein, mitochondrial	SSBP1

Table S1: Proteins differentially bound to COB223-biotin-avidin-agarose versus COB236-biotin-avidin-agarose. The ratio of intensities (when detectable under both conditions) was determined from MS/MS analyses

NMR and HPLC spectra:

Figure. S2. 7-Acetyl-3-methyl-1-[(prop-2-yn-1-yl)carbamoyl] indolizine-3-carboxylate **3**



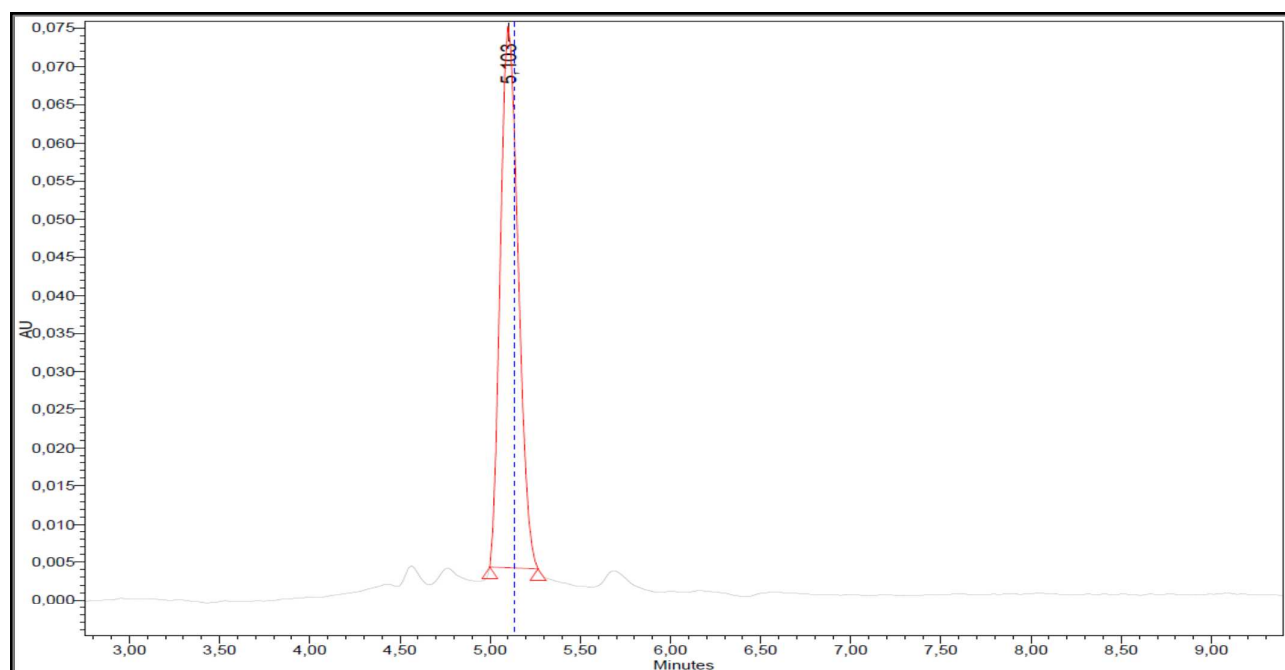
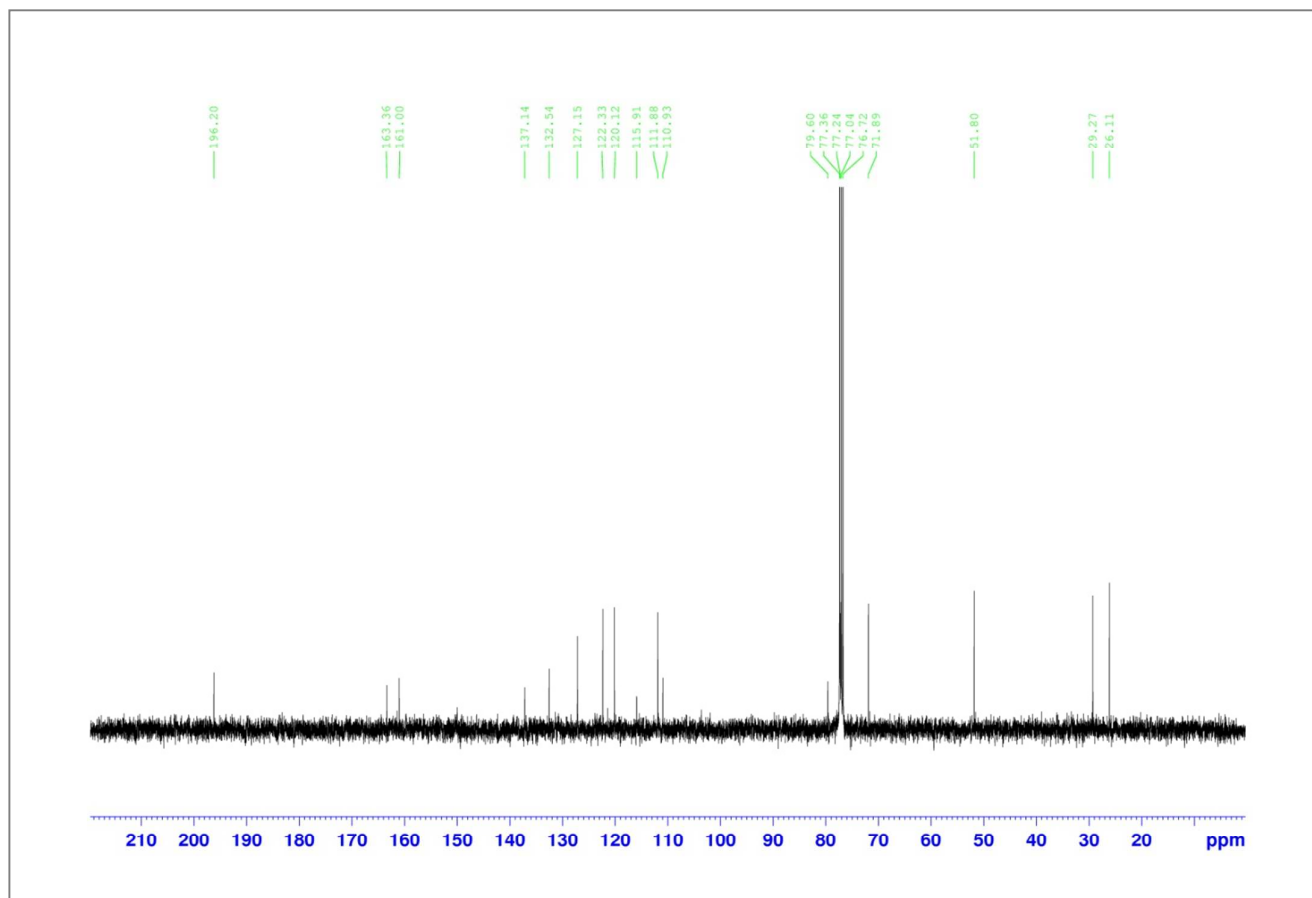
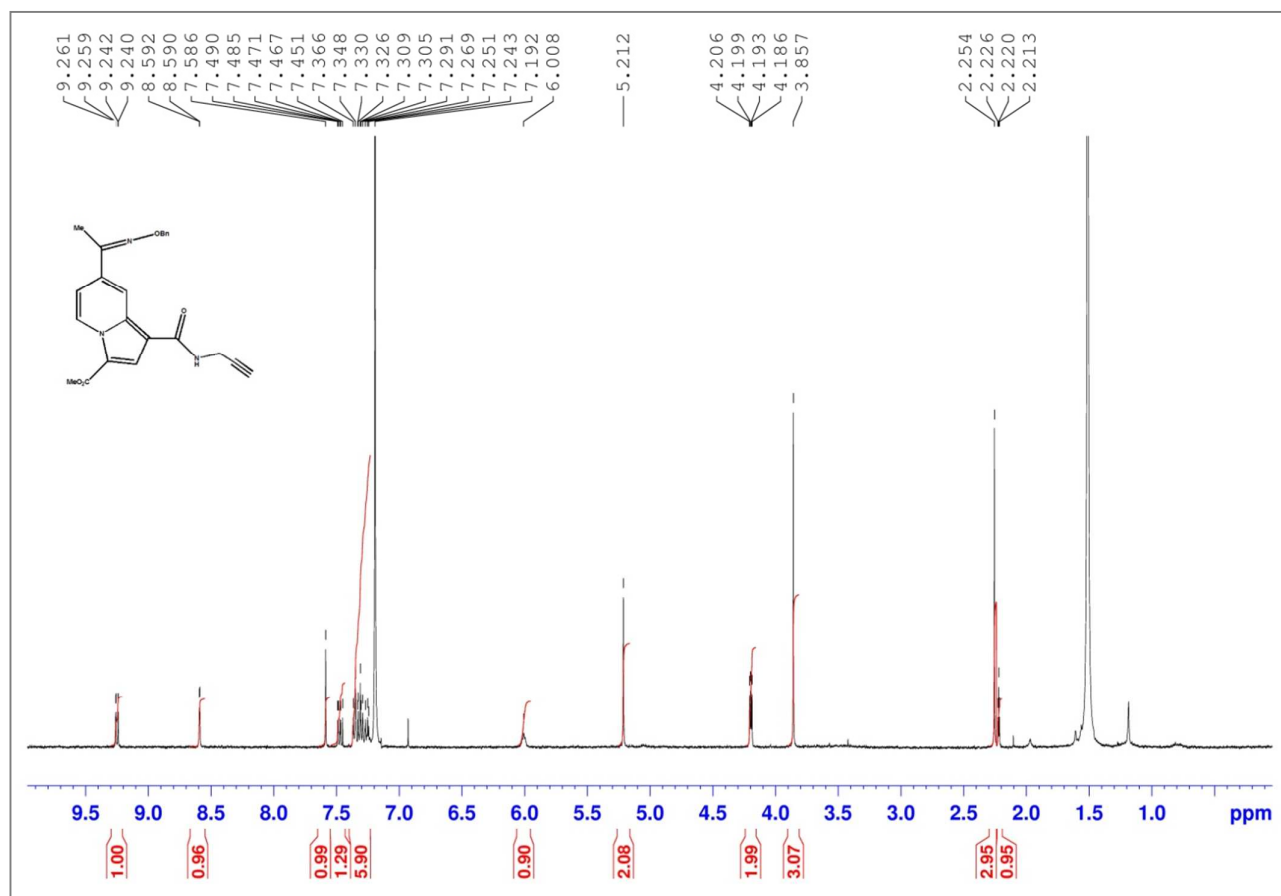


Figure. S3 7-[(1-(benzyloxy)imino)ethyl]-3-methyl-1-[(prop-2-yn-1-yl)carbamoyl] indolizine-3-carboxylate 4



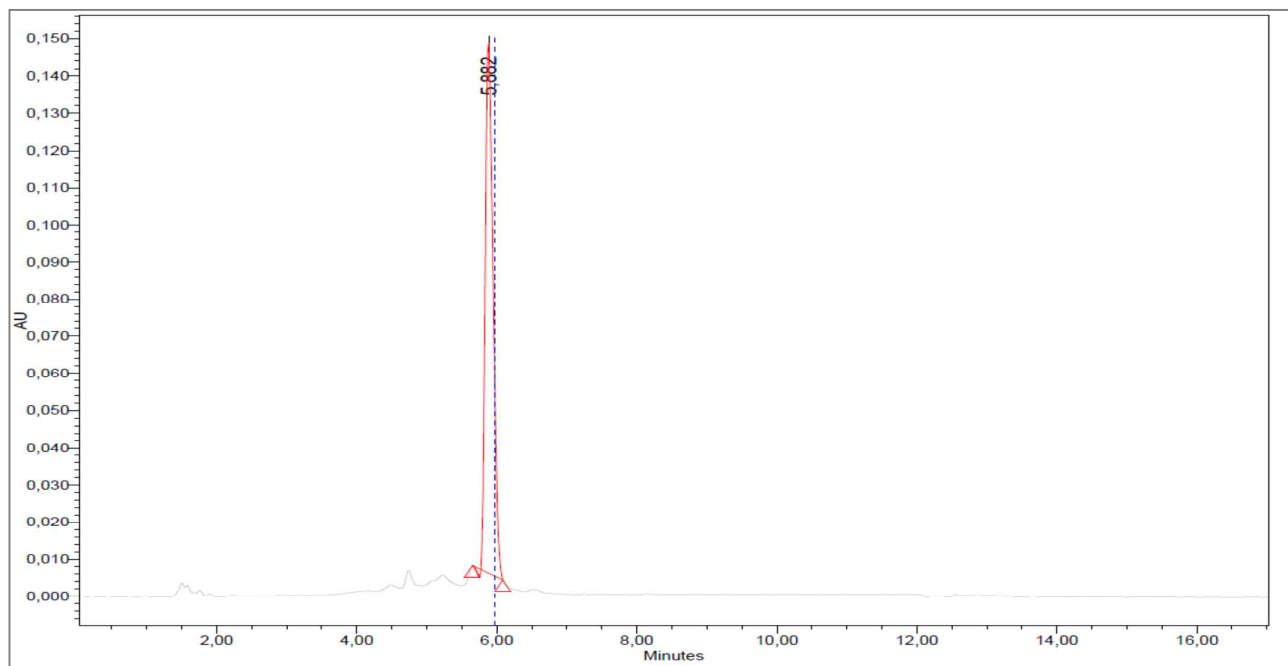
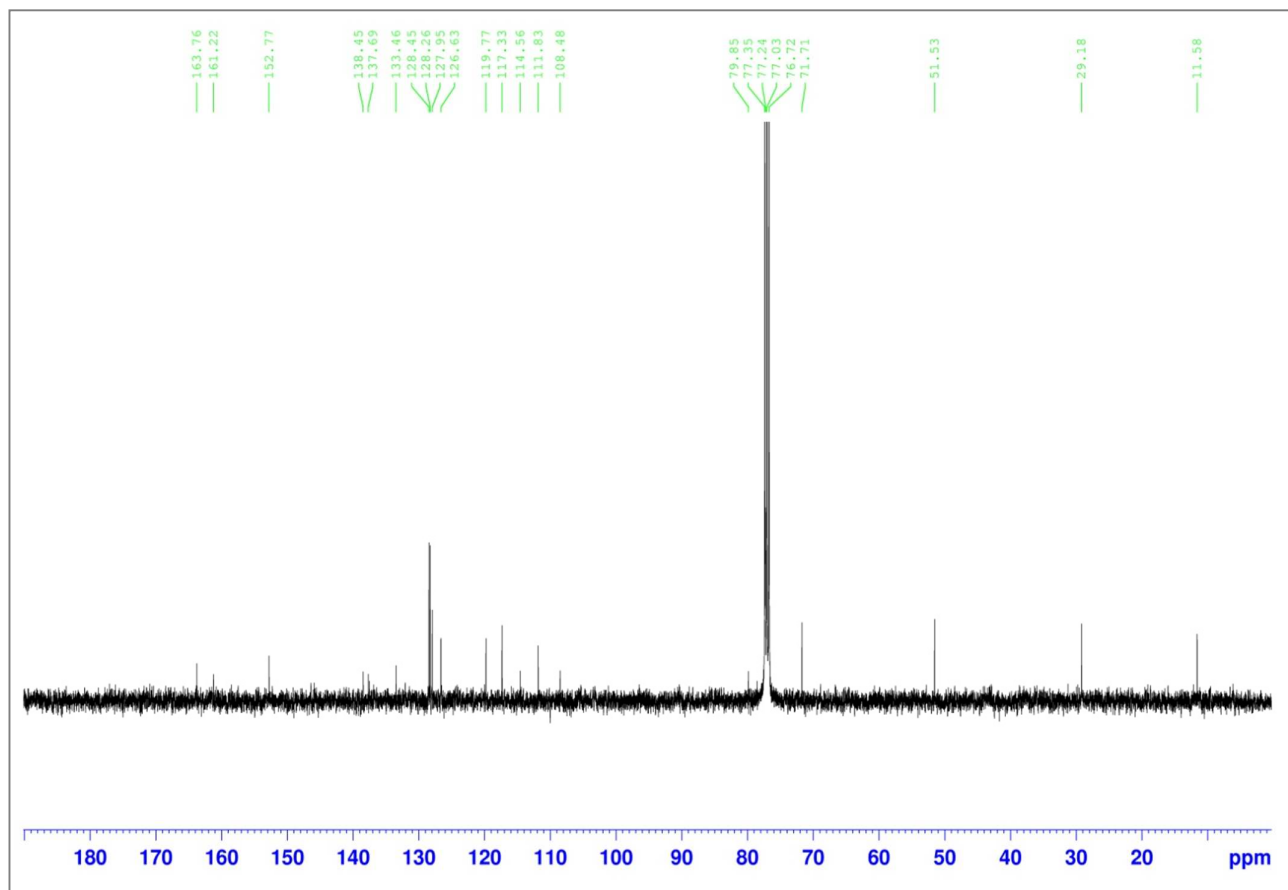
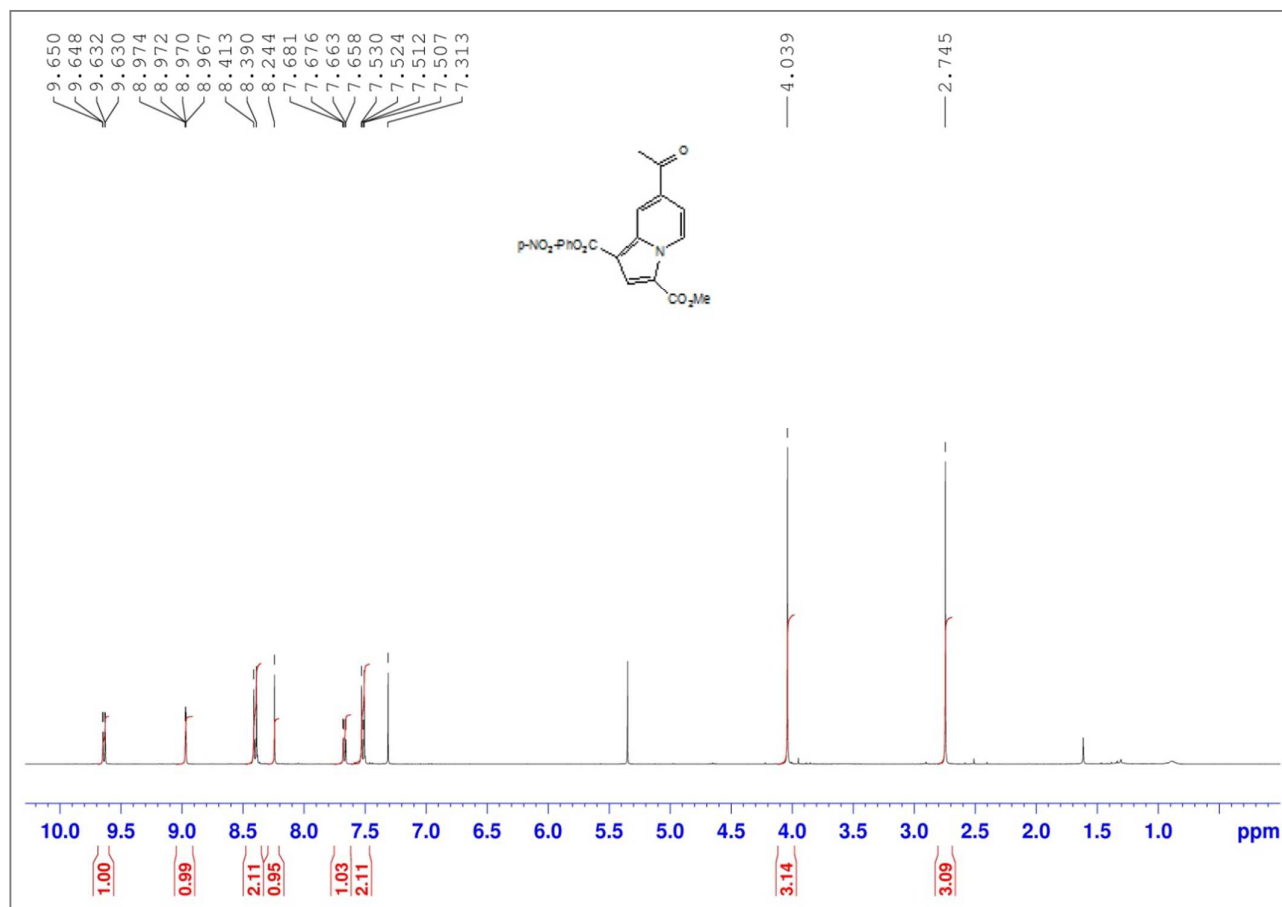


Figure. S4 7-Acetyl-1-(4-nitrophenyl)-3-methyl-indolizine-1,3-dicarboxylate 6



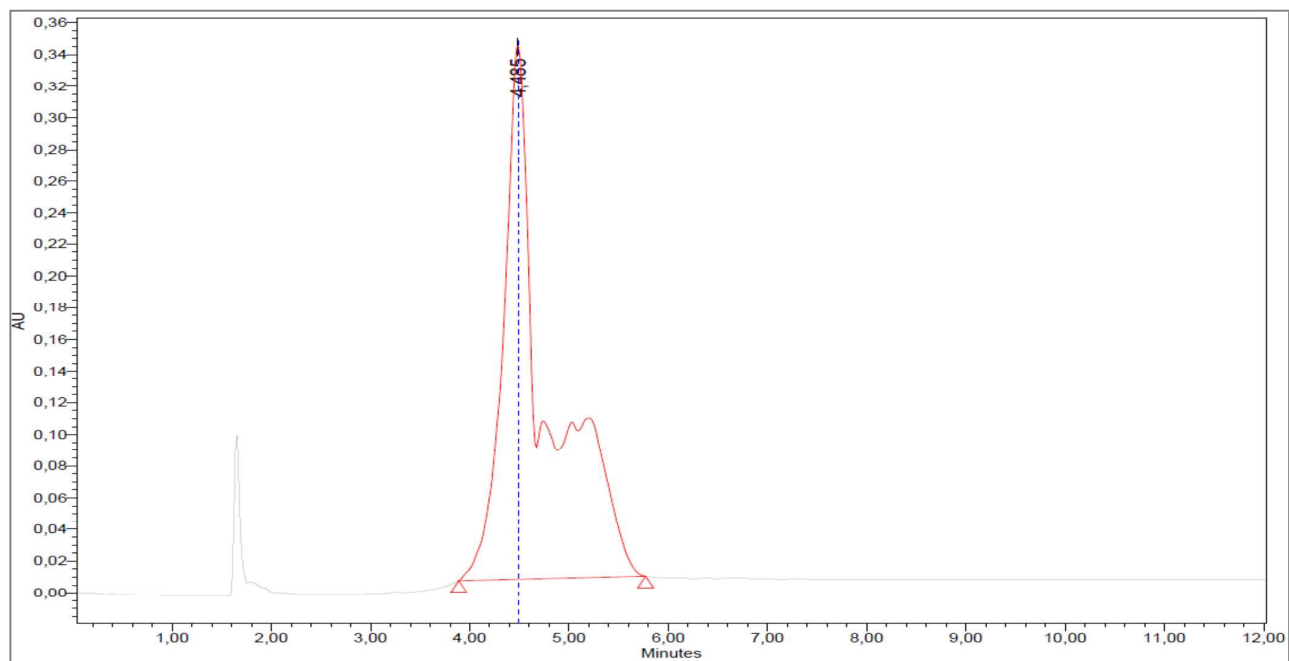
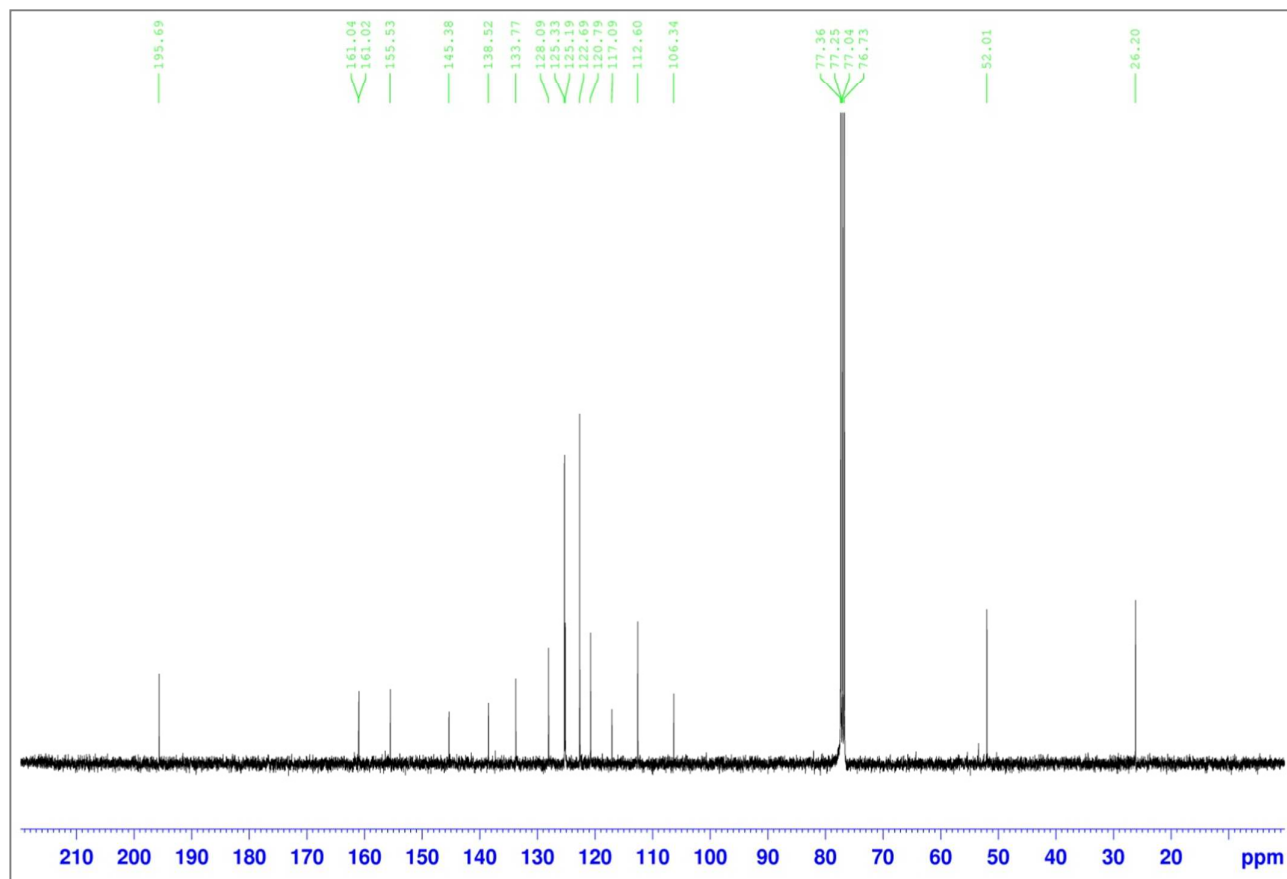
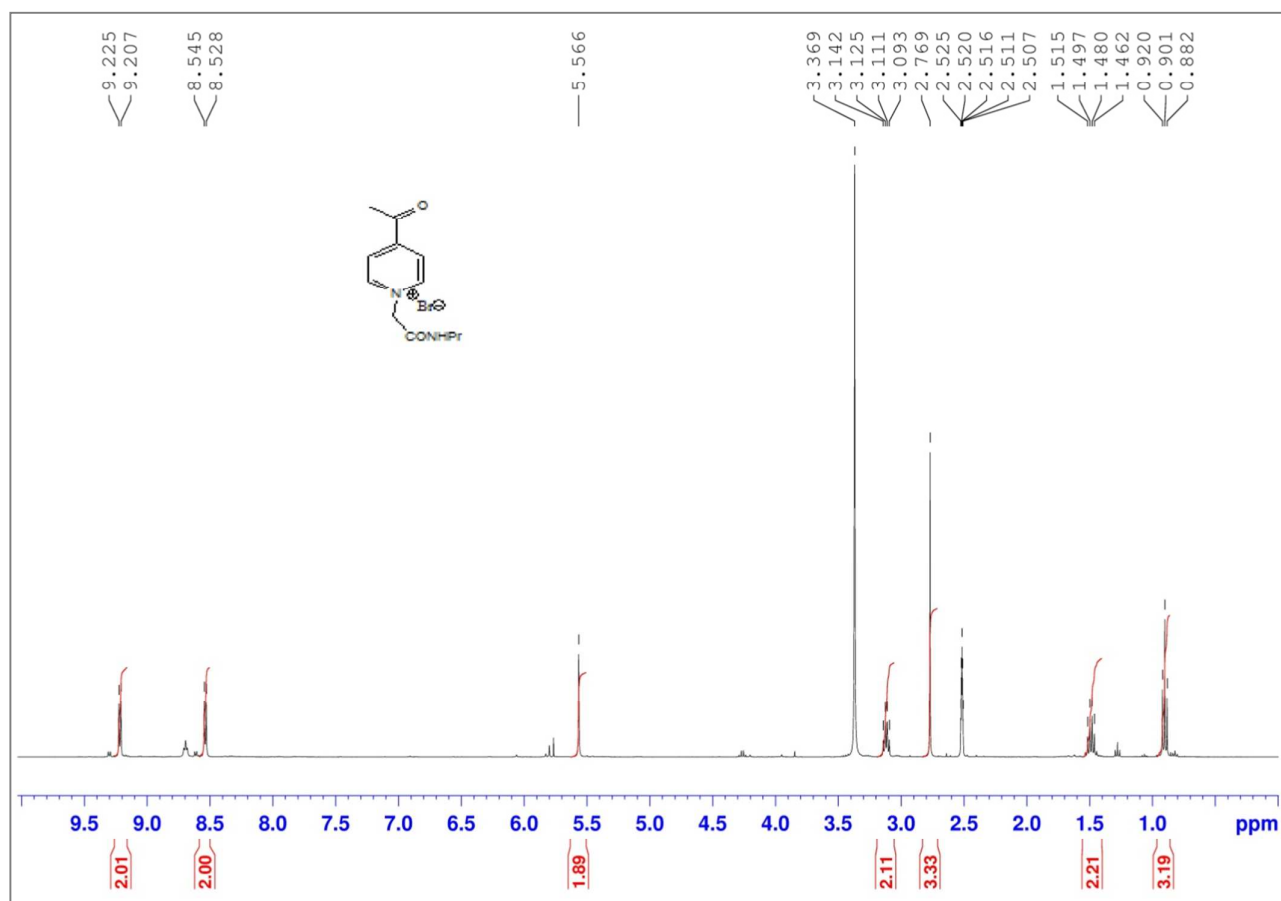


Figure. S5 4-Acetyl-1-[(N-propylcarbamoyl)methyl]pyridinium bromide 7



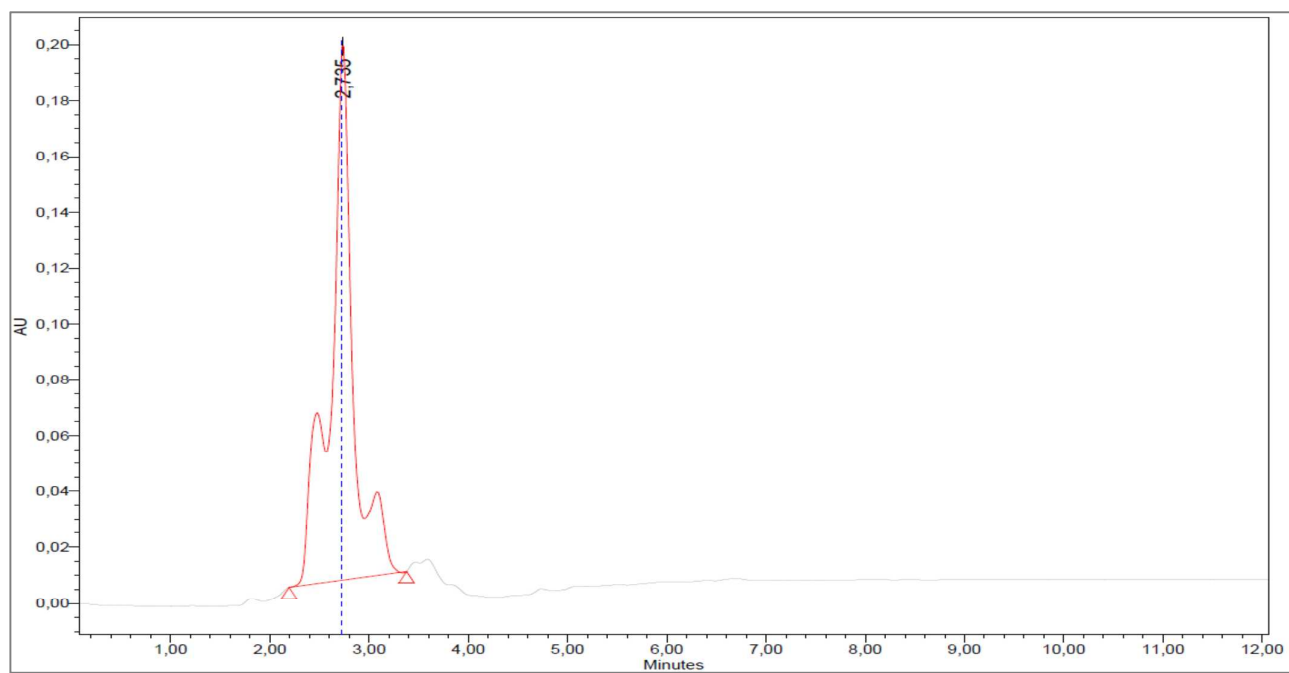
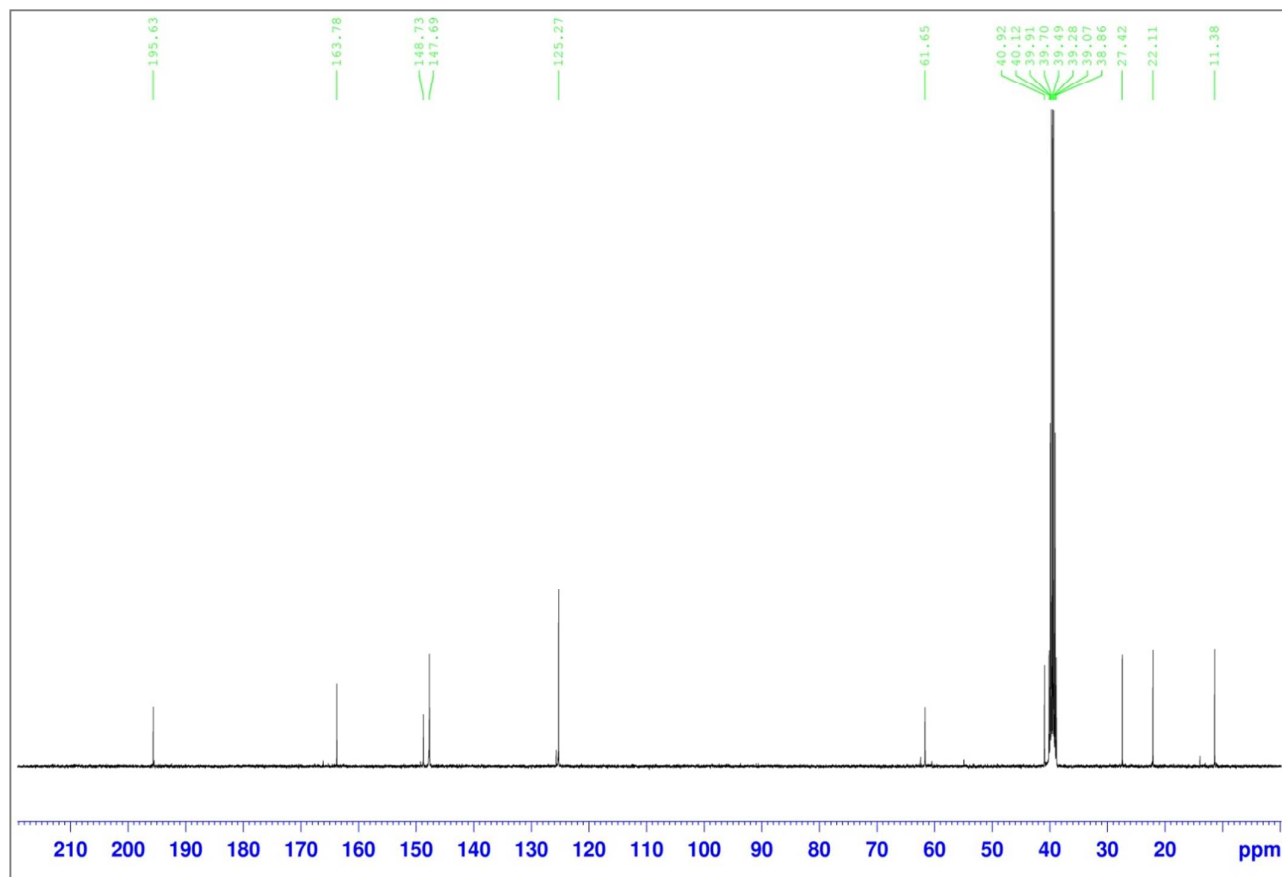
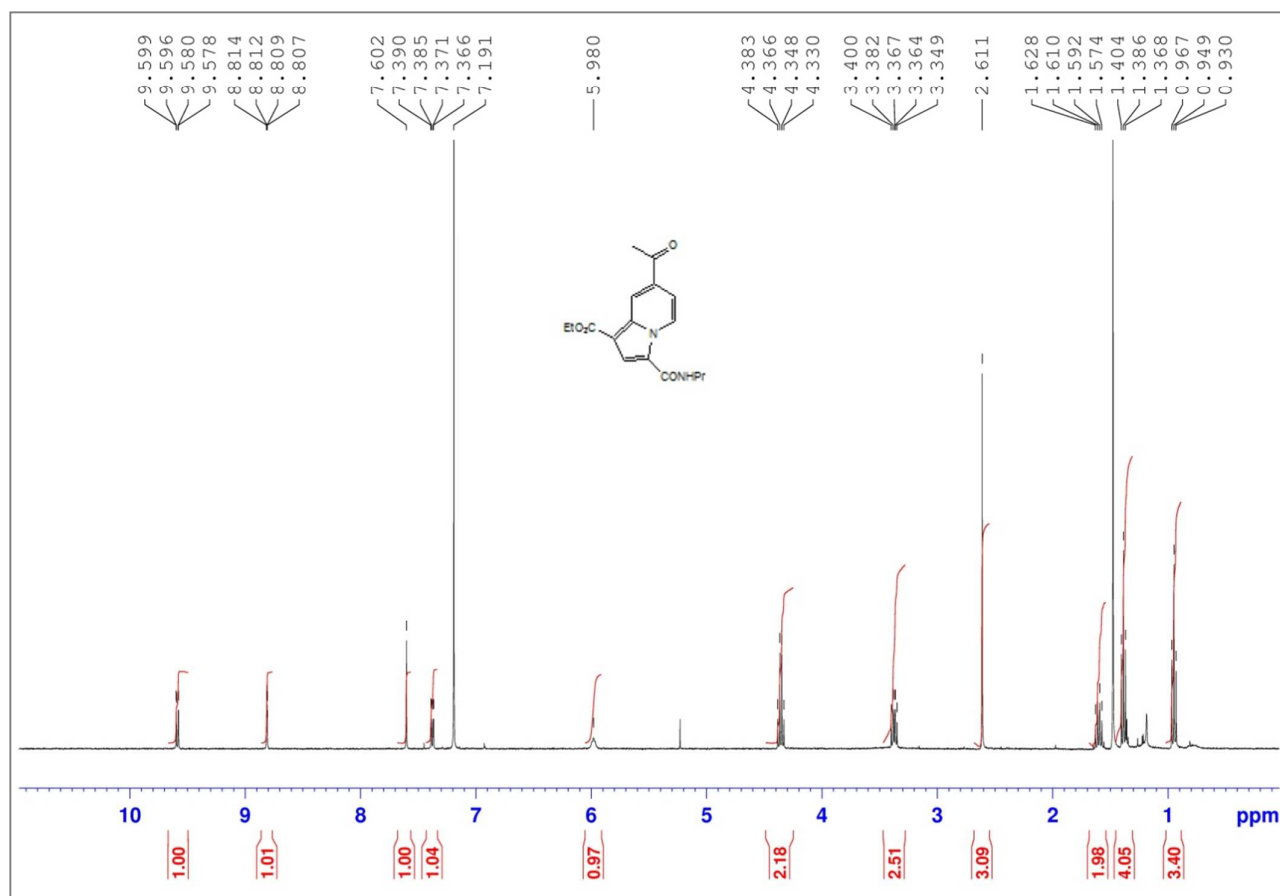


Figure. S6 7-Acetyl-1-ethyl-3-(N-propylcarbamoyl)indolizine-1-carboxylate 8



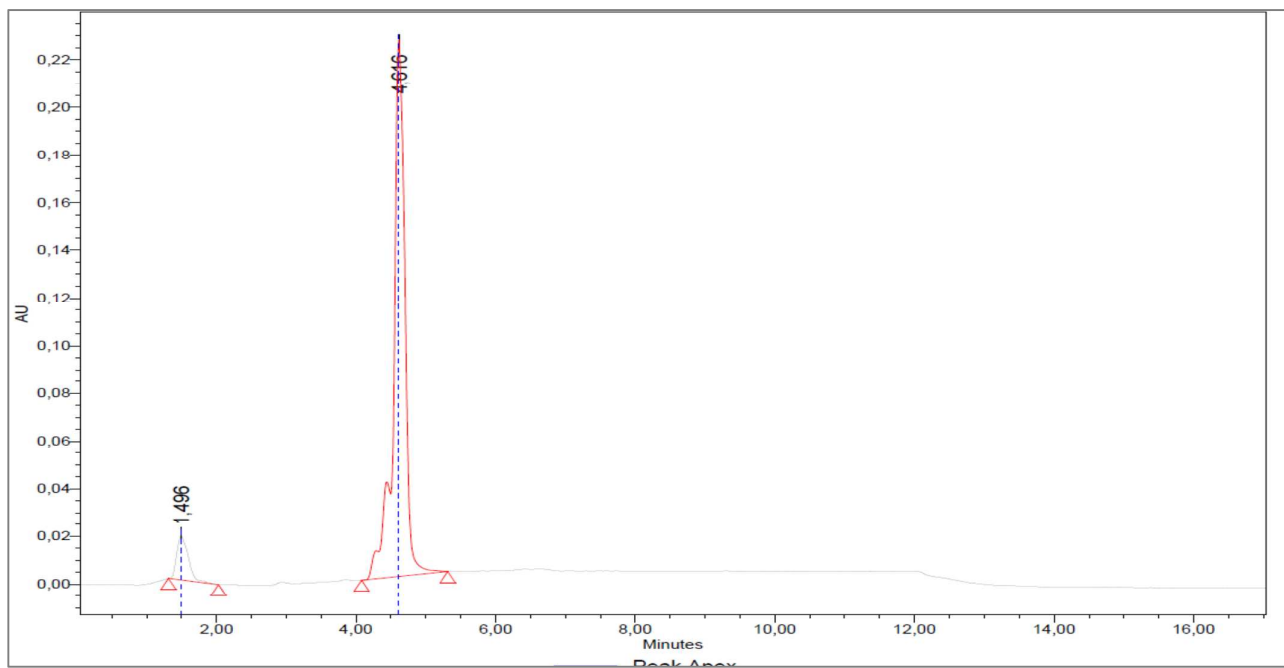
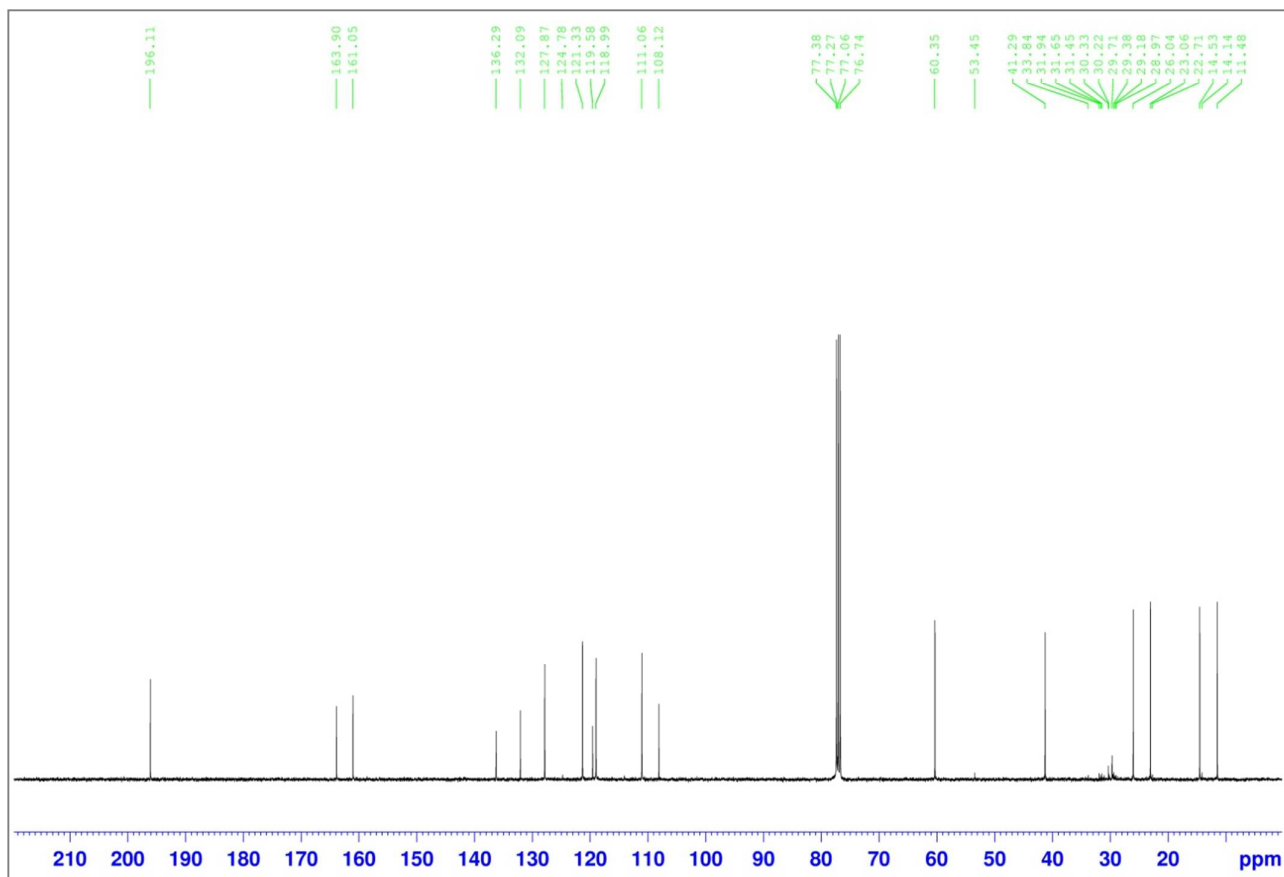
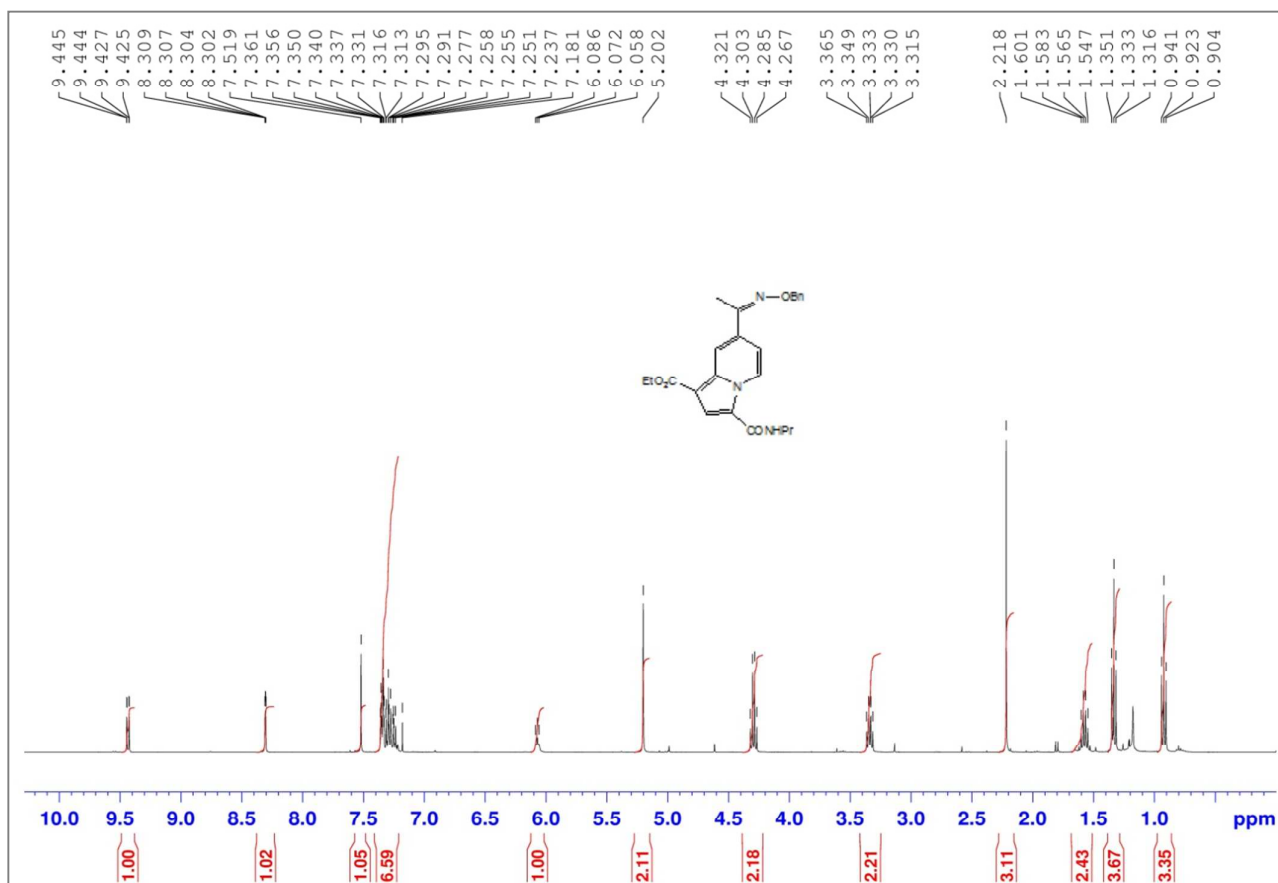


Figure. S7 7-[(1-(benzyloxy)imino)ethyl]-1-ethyl-3-(N-propylcarbamoyl) indolizine-1-carboxylate 9



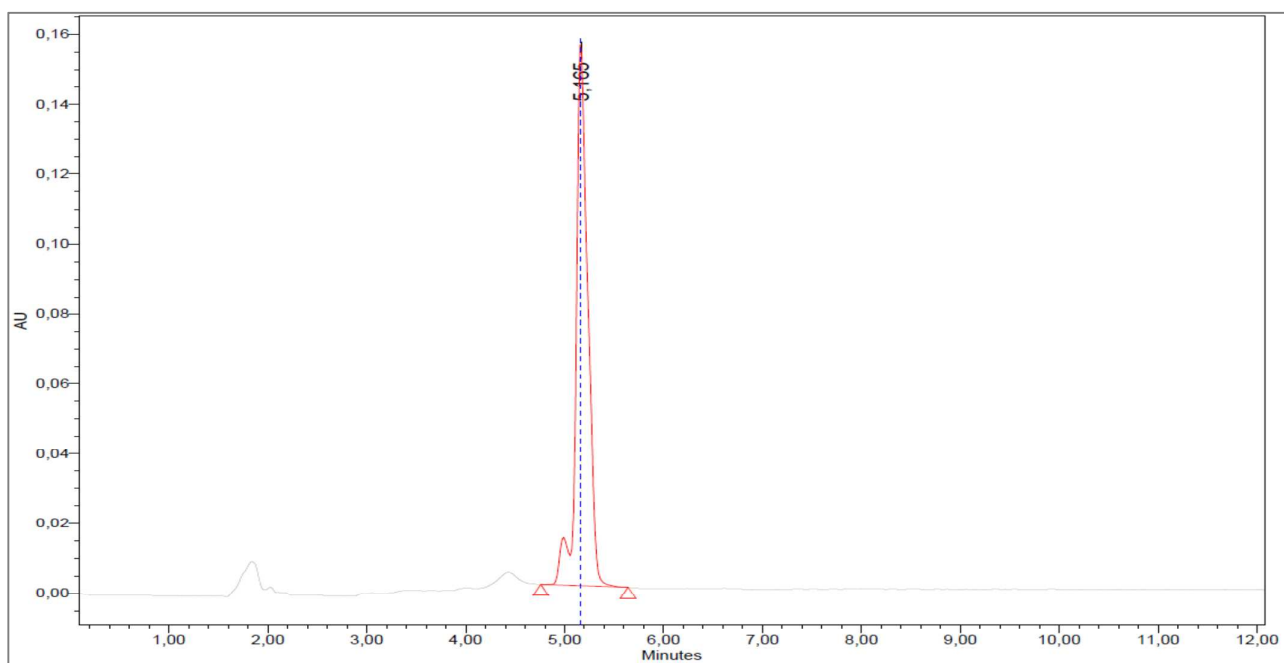
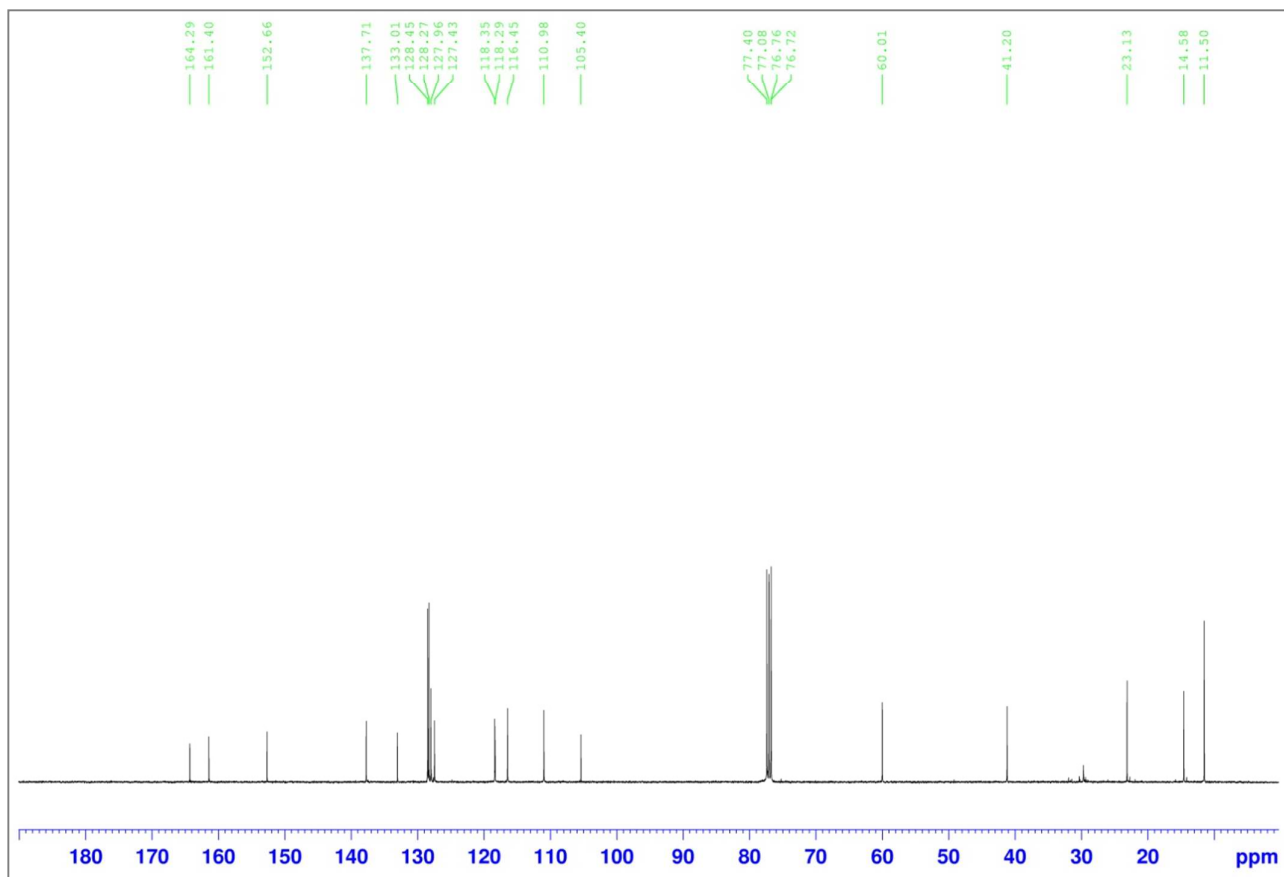
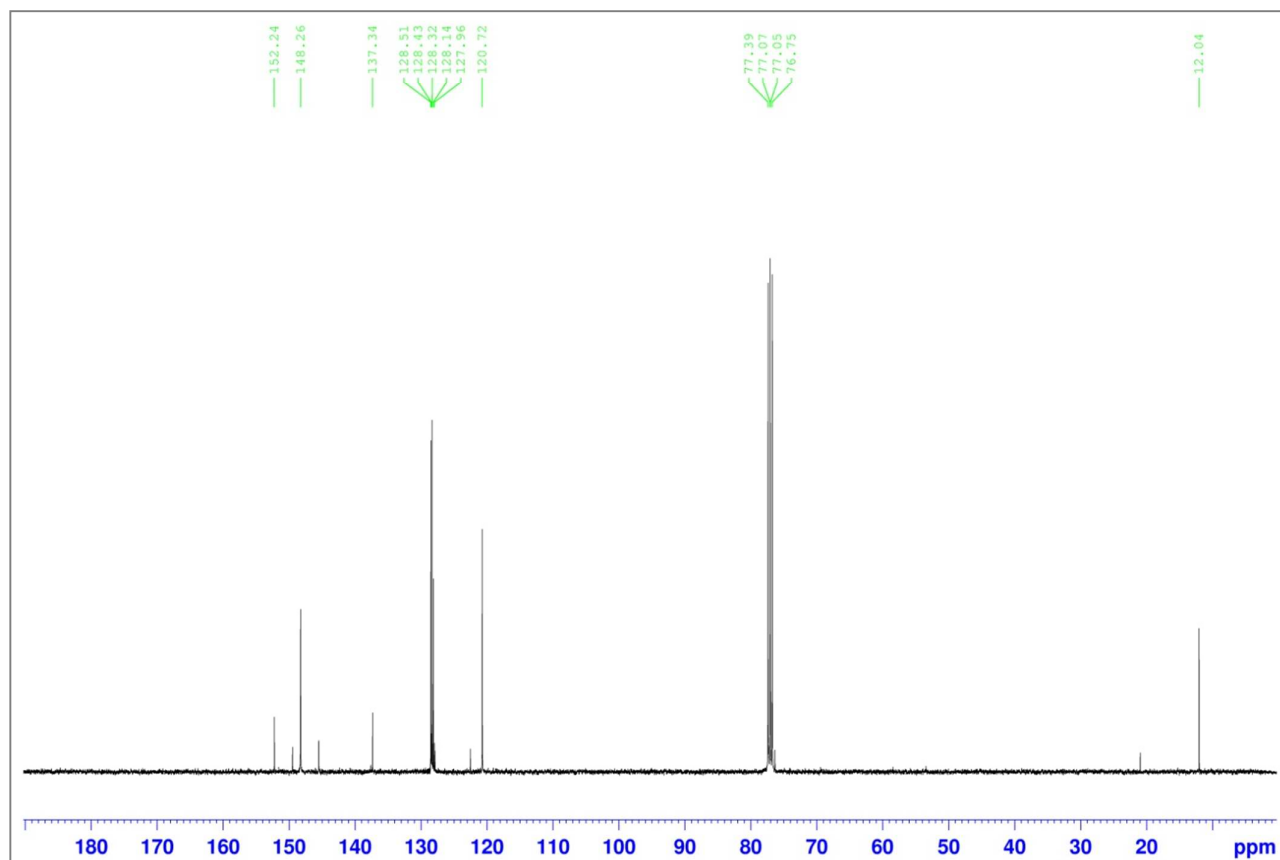
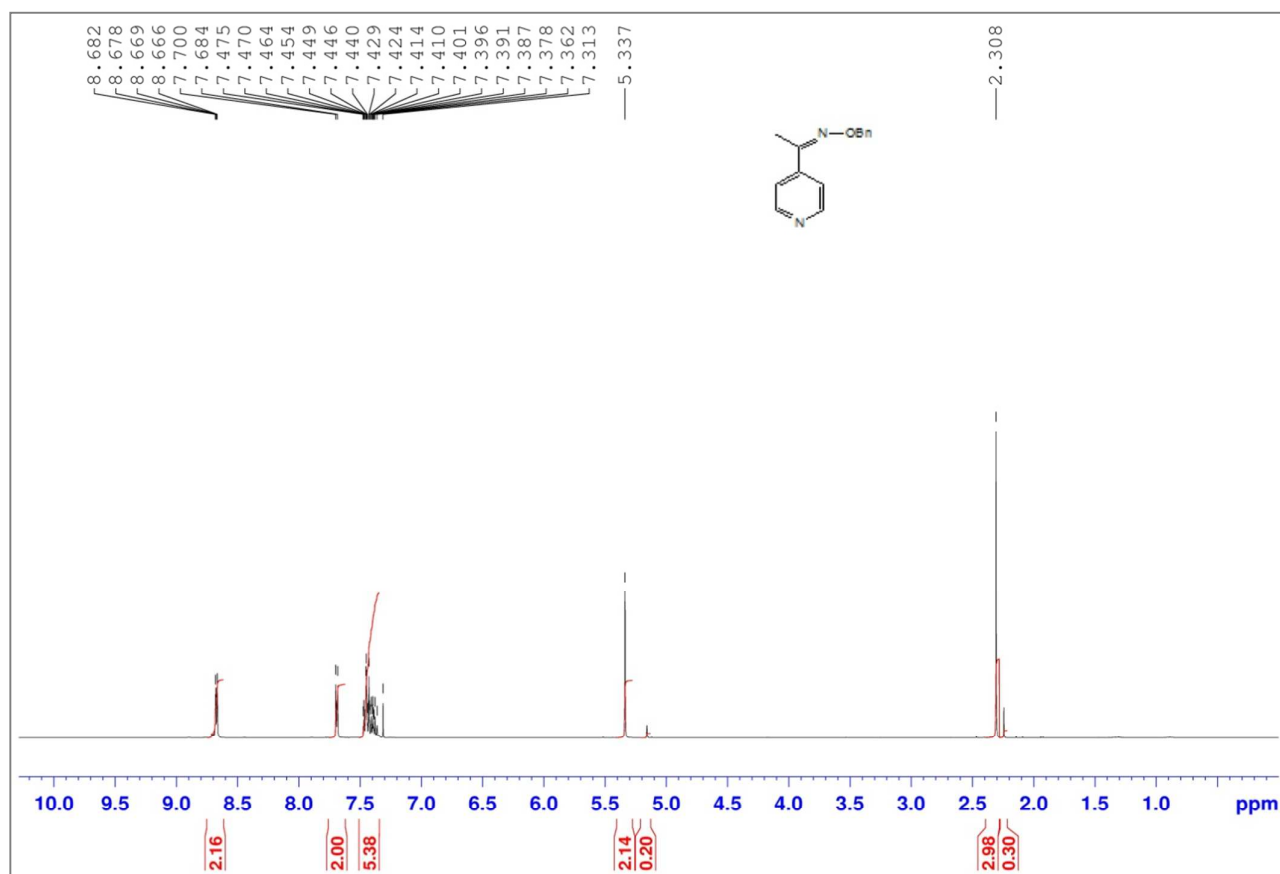


Figure. S8 4-[(1-(benzyloxy)imino)ethyl]pyridine 10



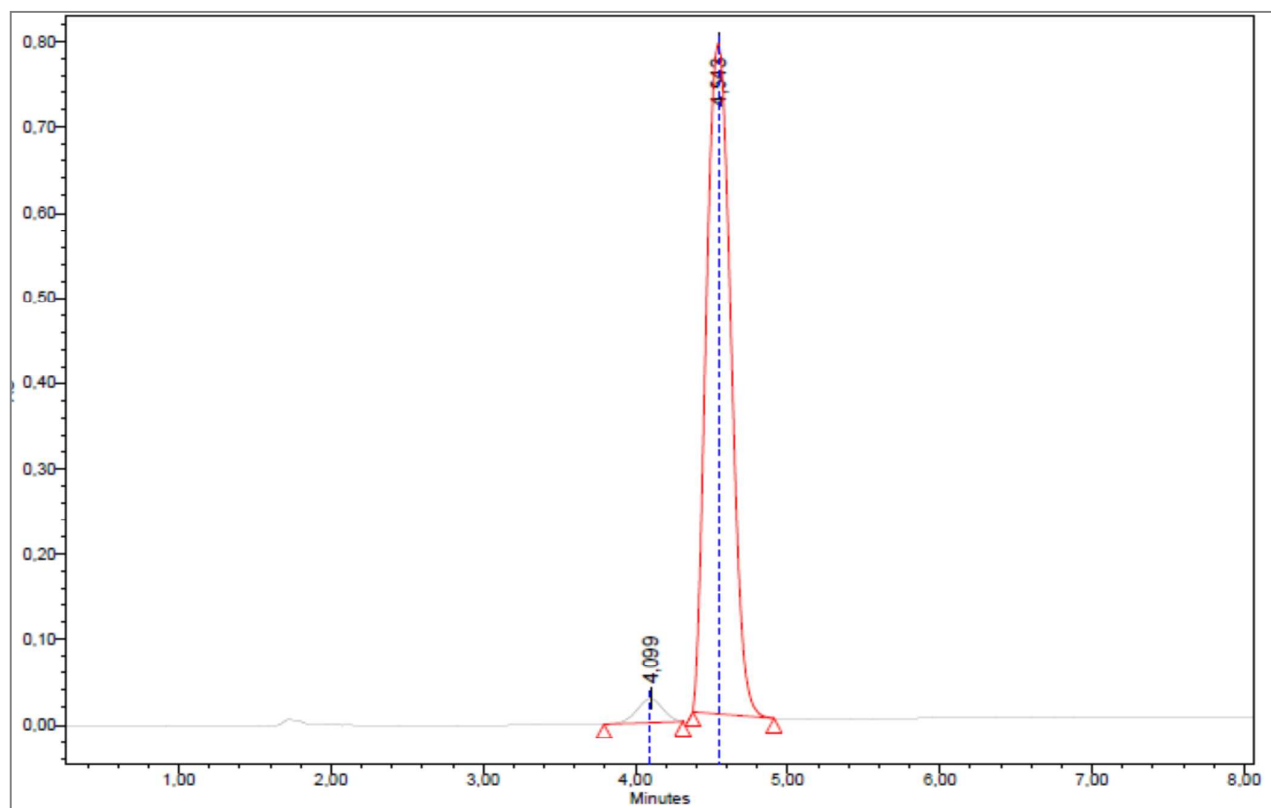
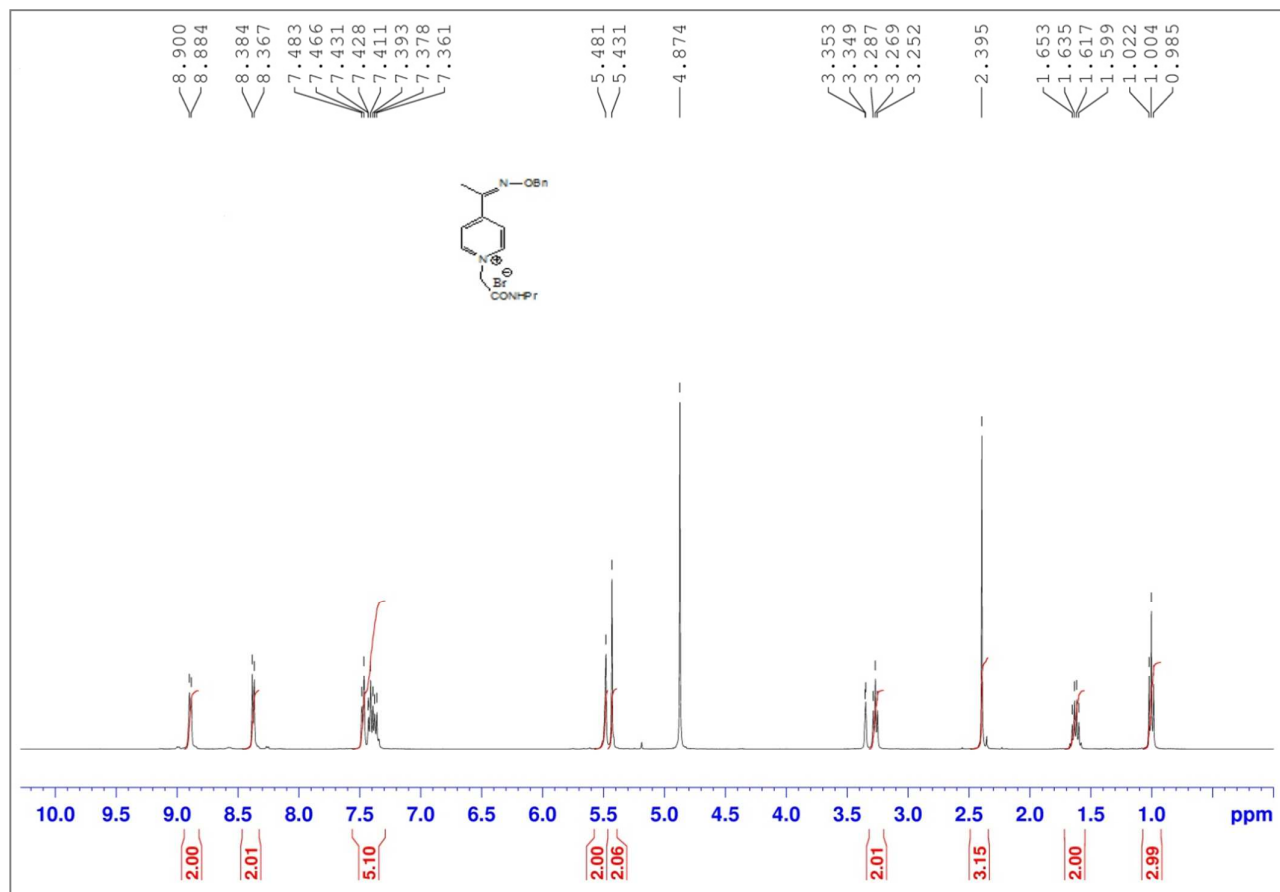


Figure. S9 4-[(1-(benzyloxy)imino)ethyl]-1-[(N-propylcarbamoyl)methyl]pyridinium bromide 11



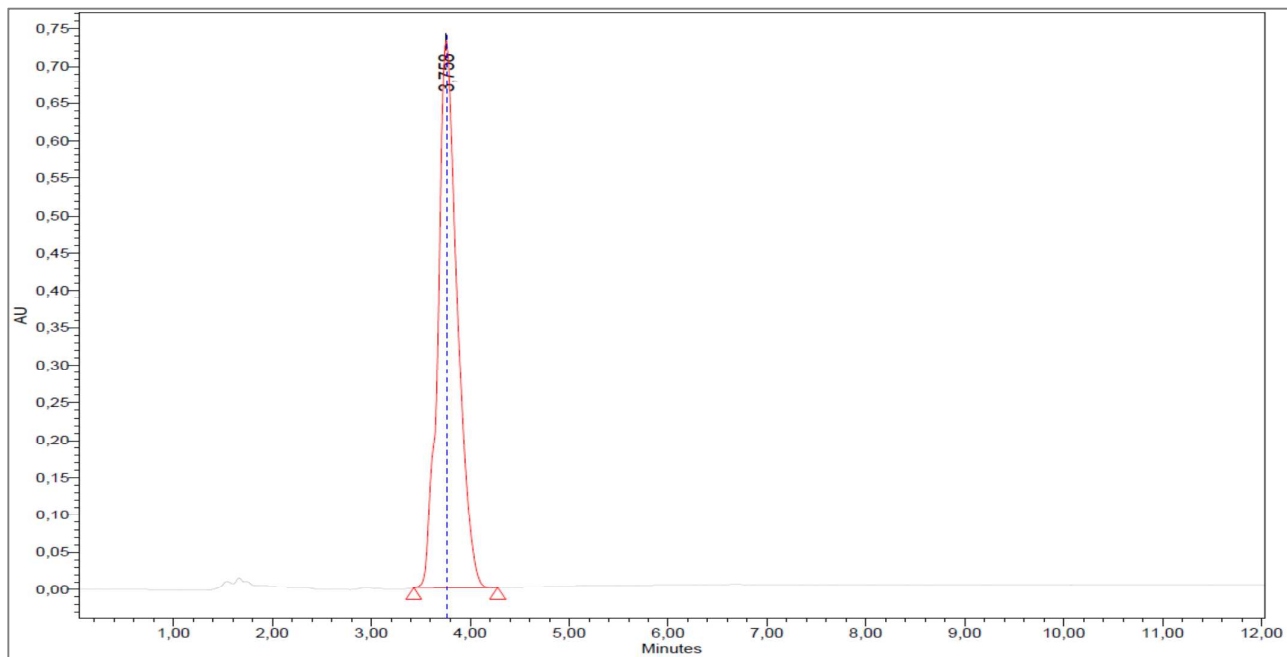
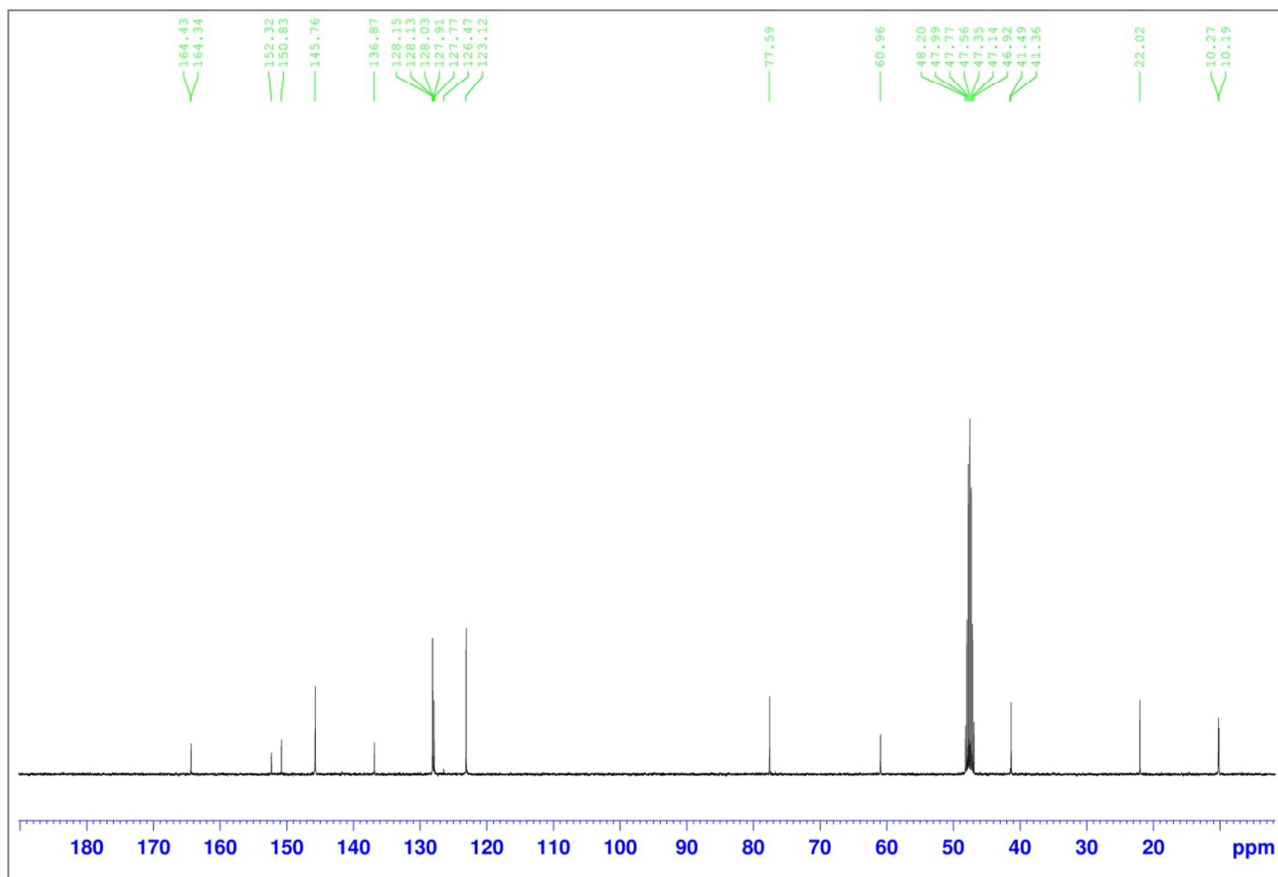
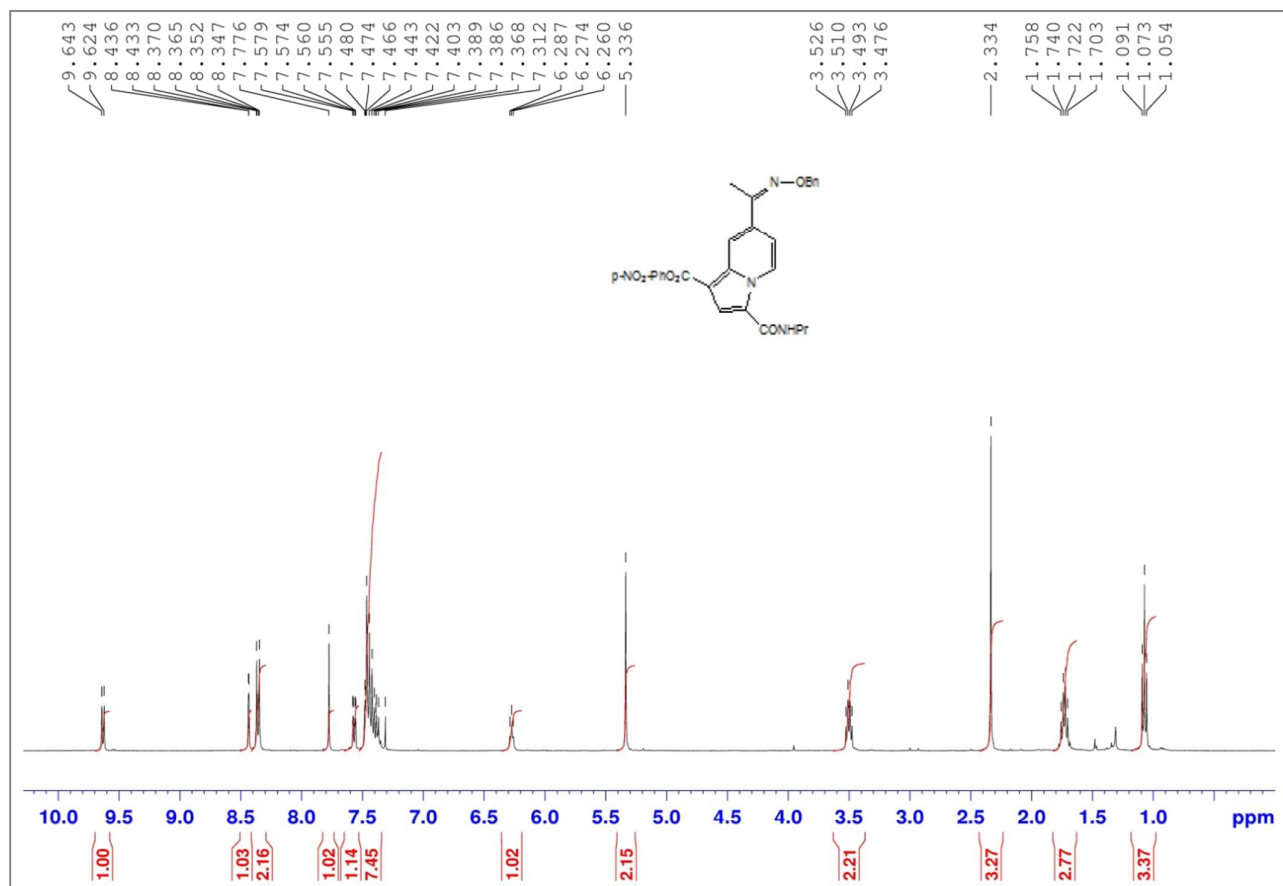


Figure. S10 7-[(1-(benzyloxy)imino)ethyl]-1-(4-nitrophenyl)-3-(N-propylcarbamoyl) indolizine-1-carboxylate 12



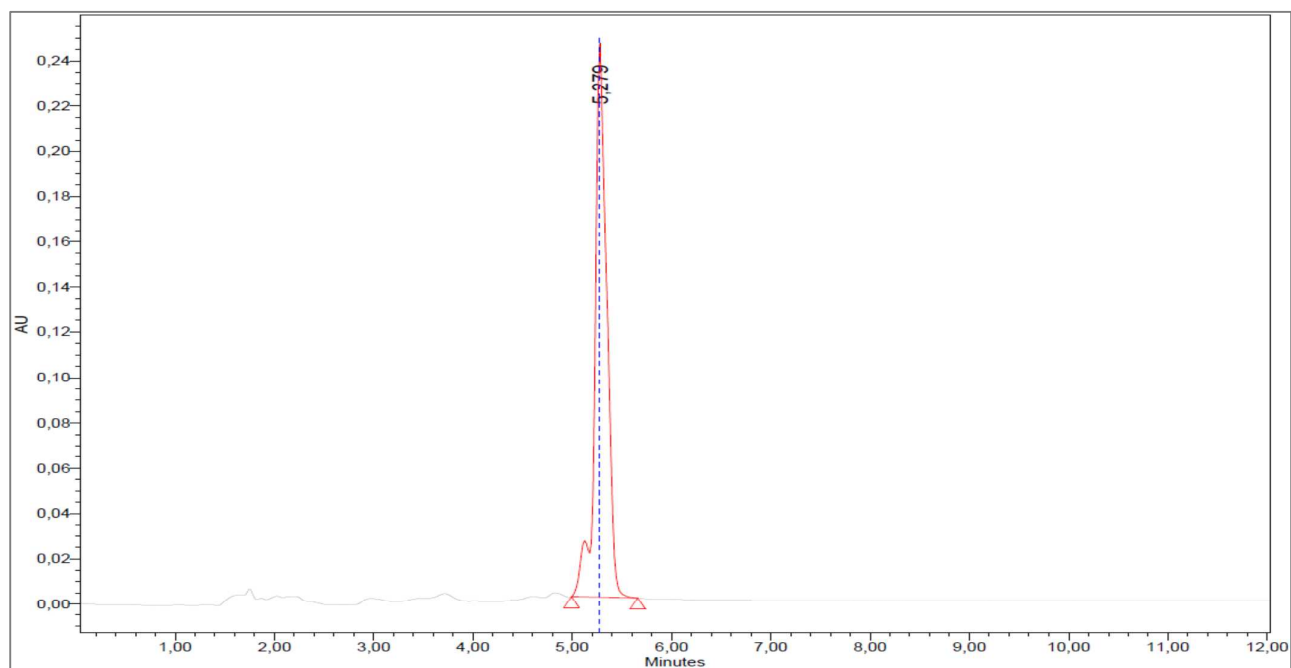
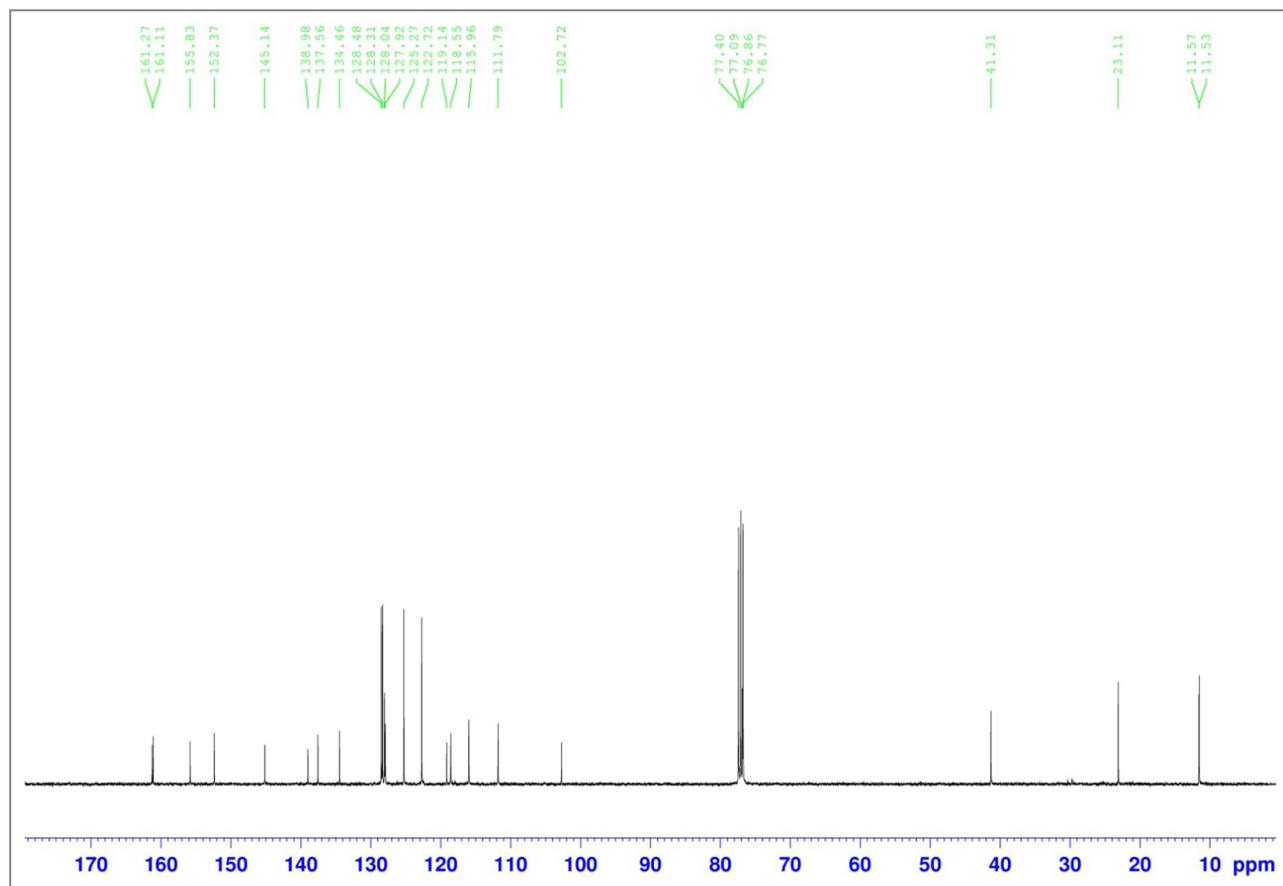
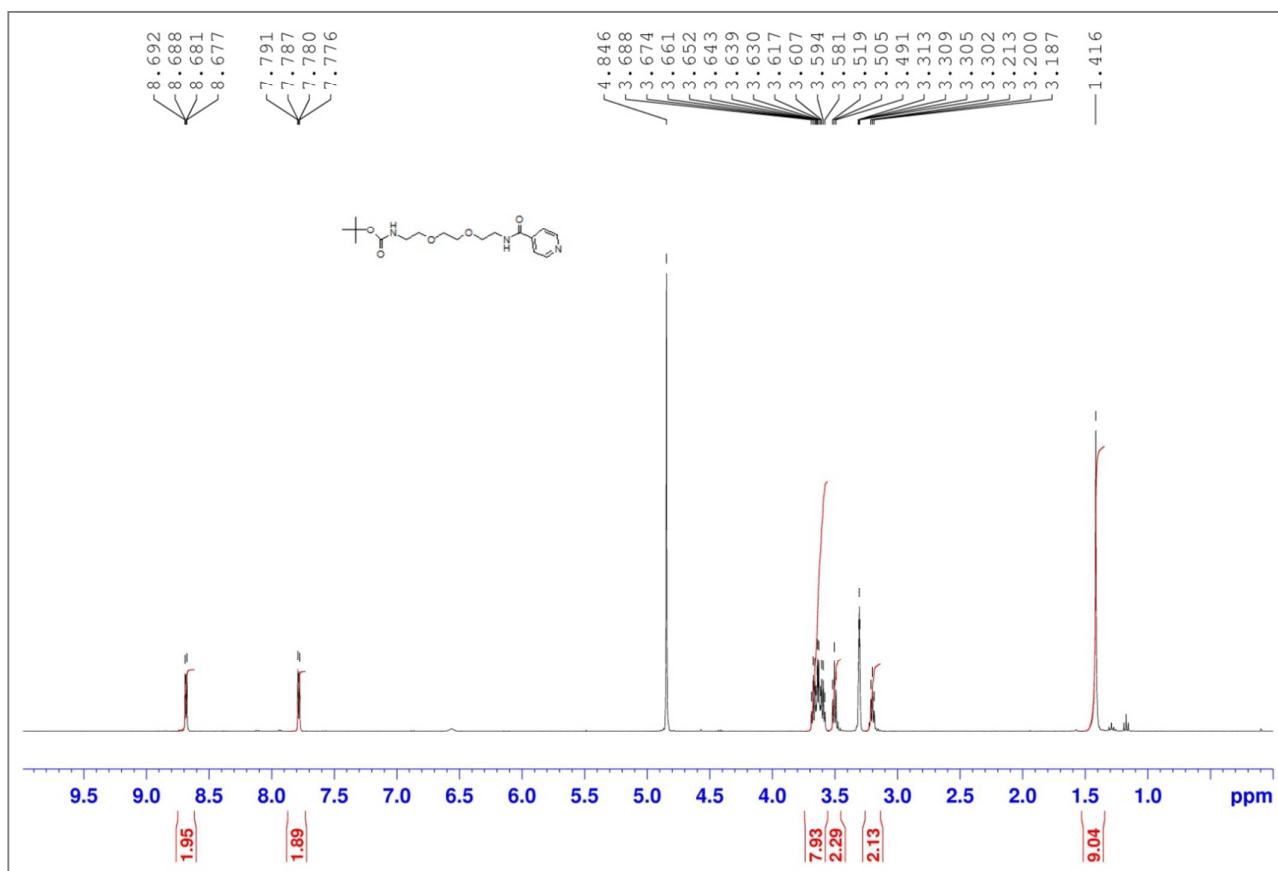


Figure. S11 N-[2-(2-(2-tert-butyloxycarbonylaminoethoxy)ethoxy) ethyl]pyridine-4-carboxamide 14



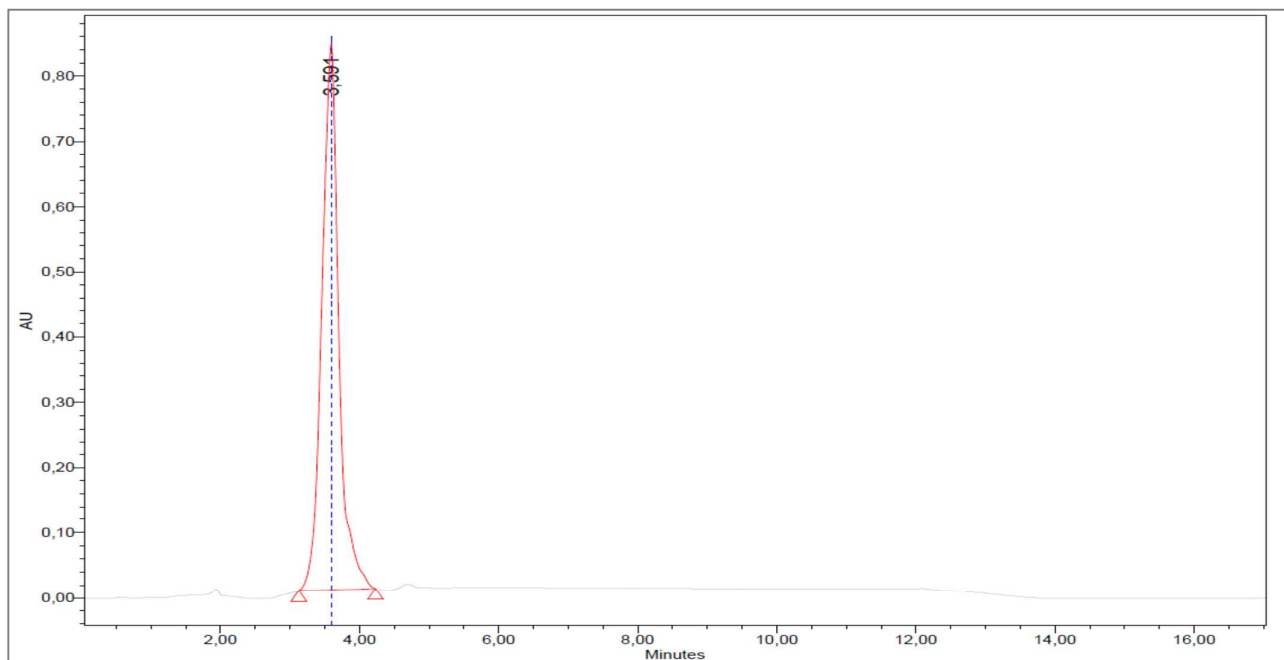
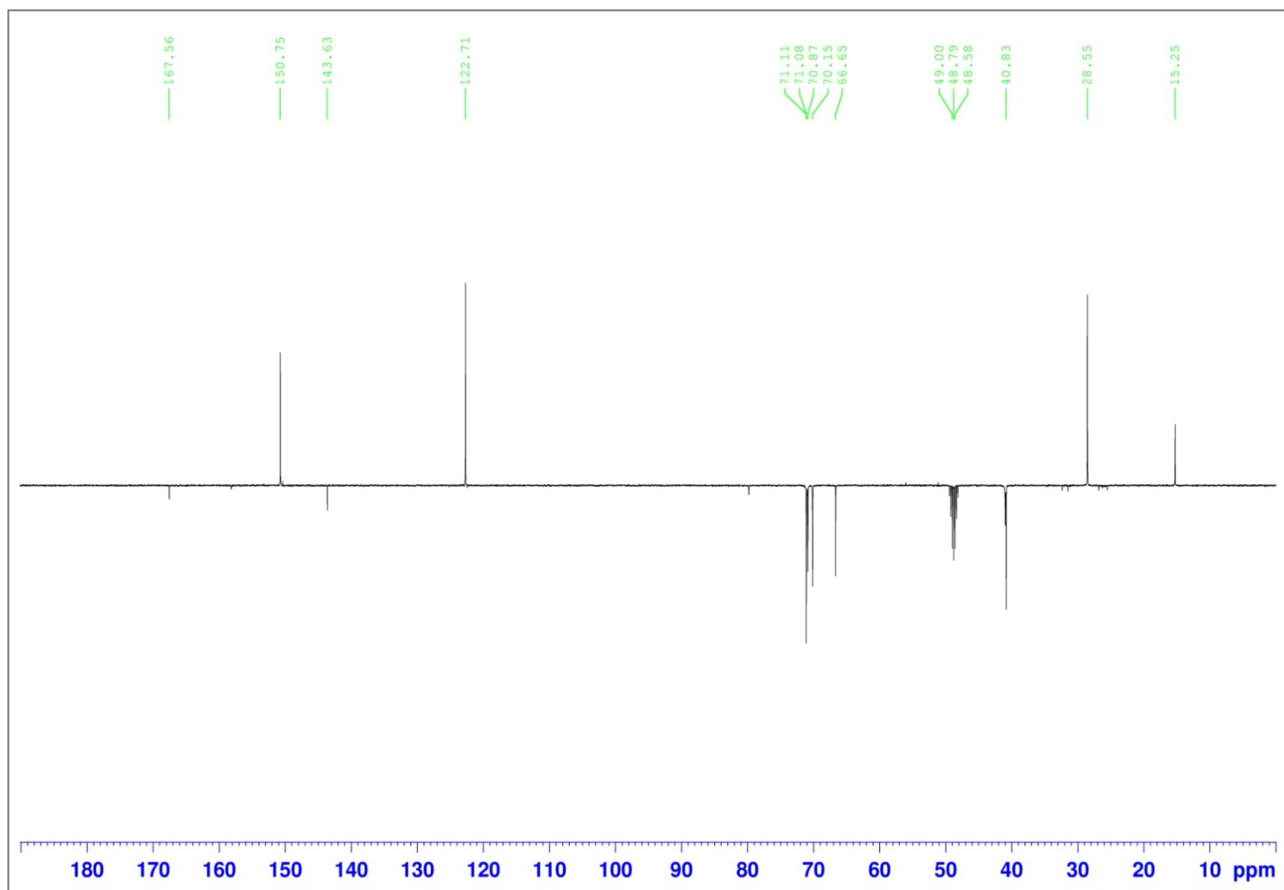
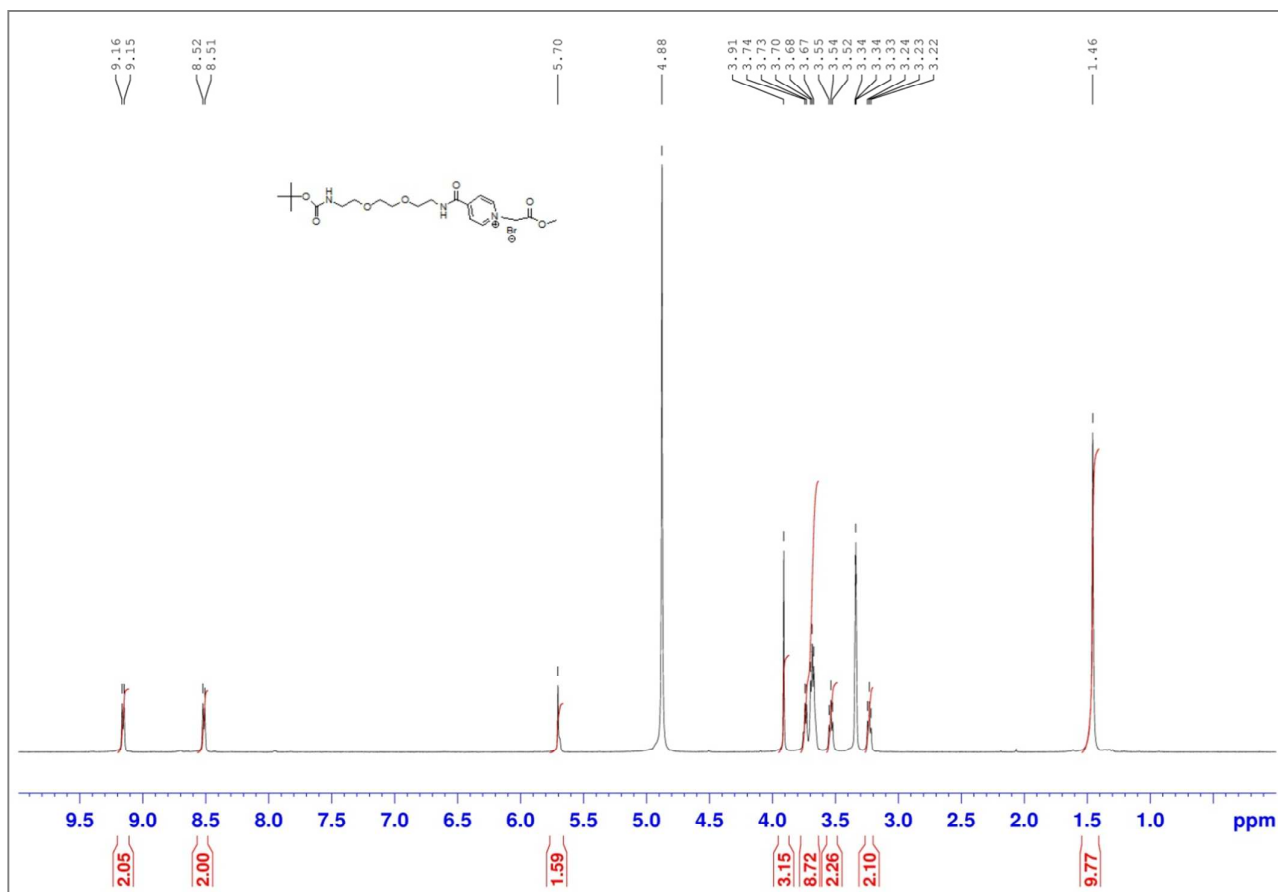


Figure. S12 4-[(2-(2-(2-tert-butyloxycarbonylaminoethoxy)ethoxy) ethyl)carbamoyl]-1-(2-methoxy-2-oxoethyl)pyridinium bromide 15a



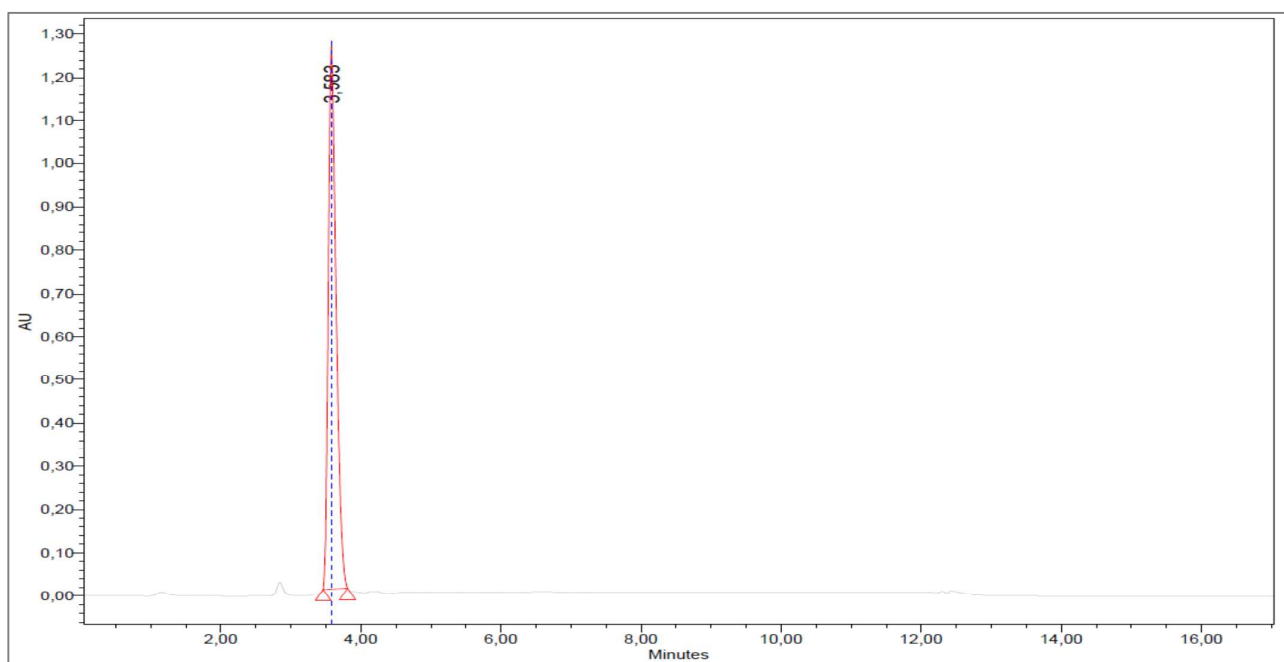
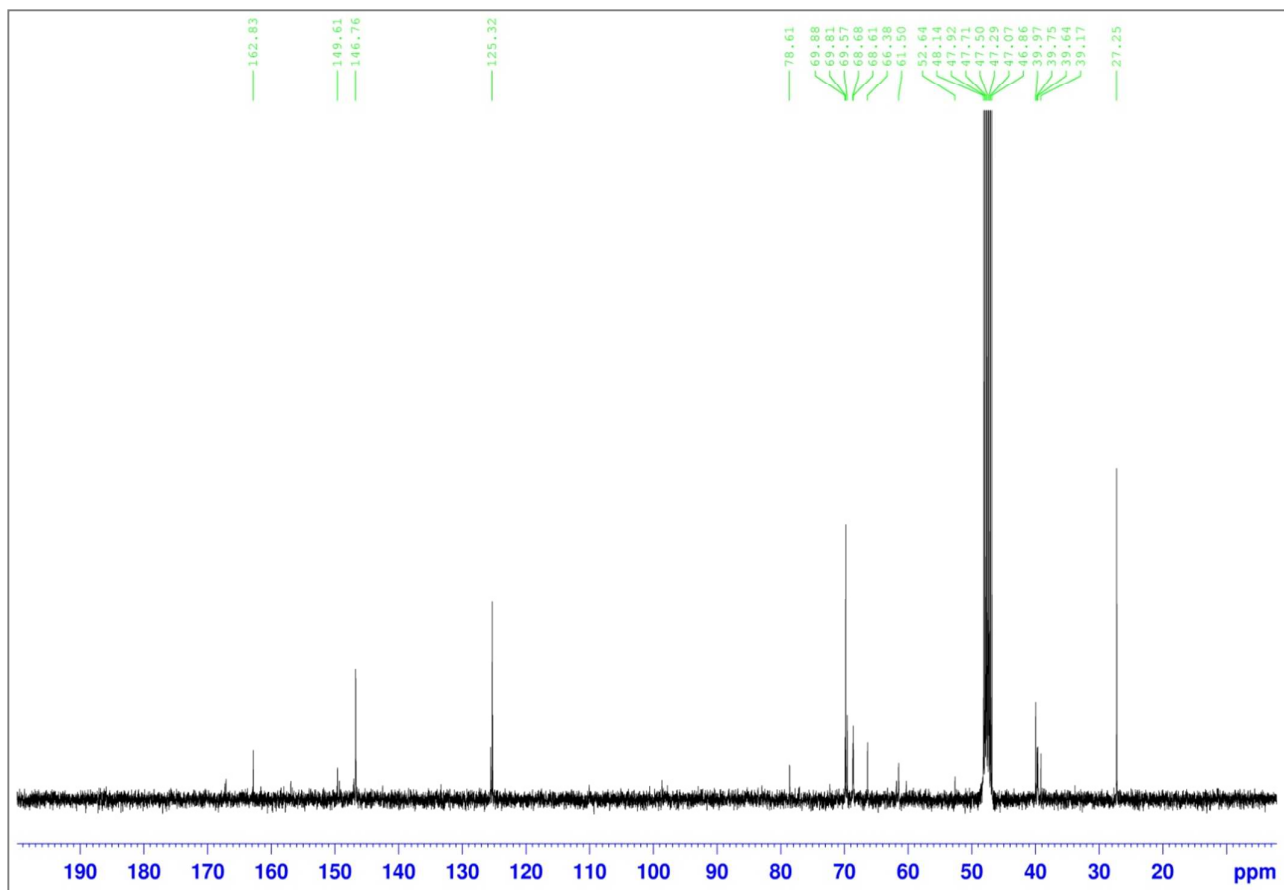
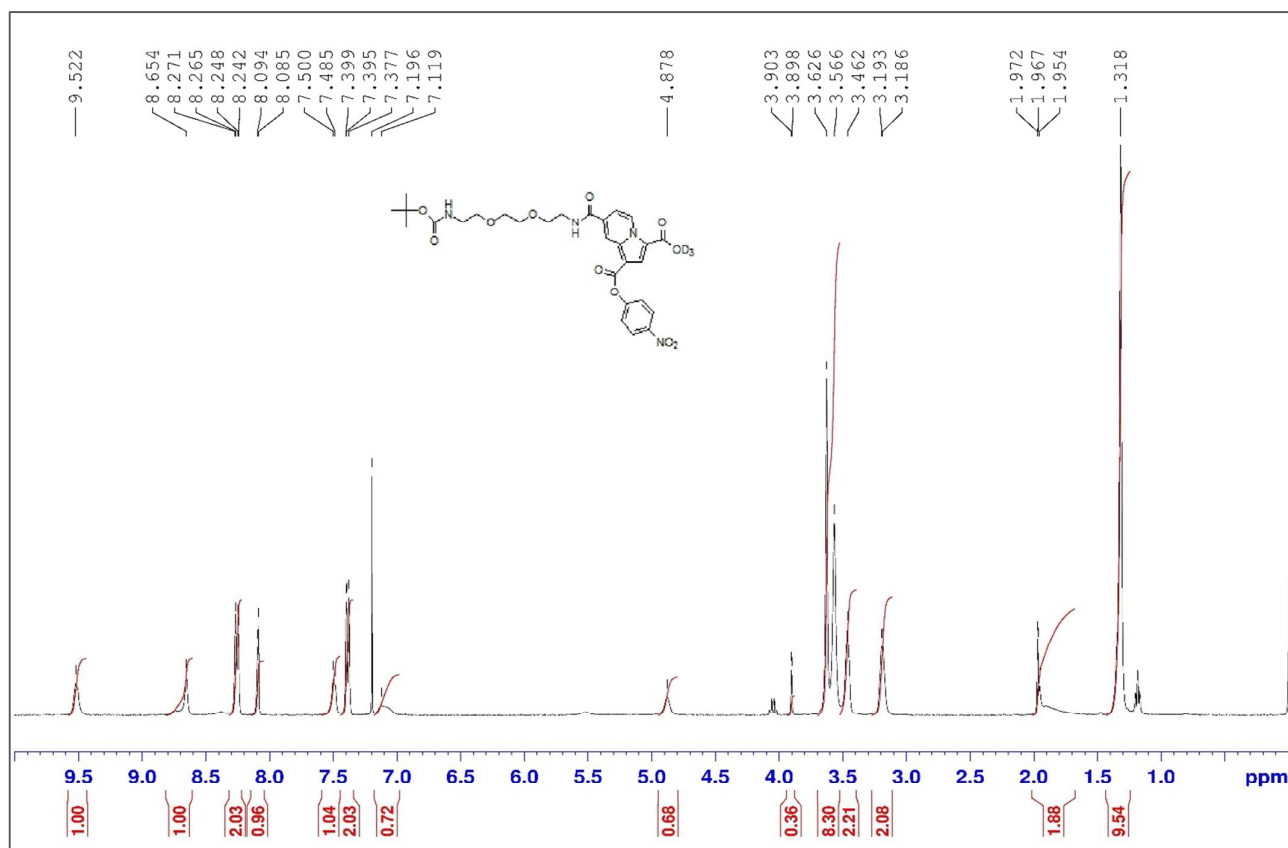


Figure. S13 7-[(2-(2-(2-tert-butyloxycarbonylaminoethoxy)ethoxy) ethyl)carbamoyl]-1-(4-nitrophenyl)-3-tri-deuteromethyl-indolizine-1,3-dicarboxylate 16.



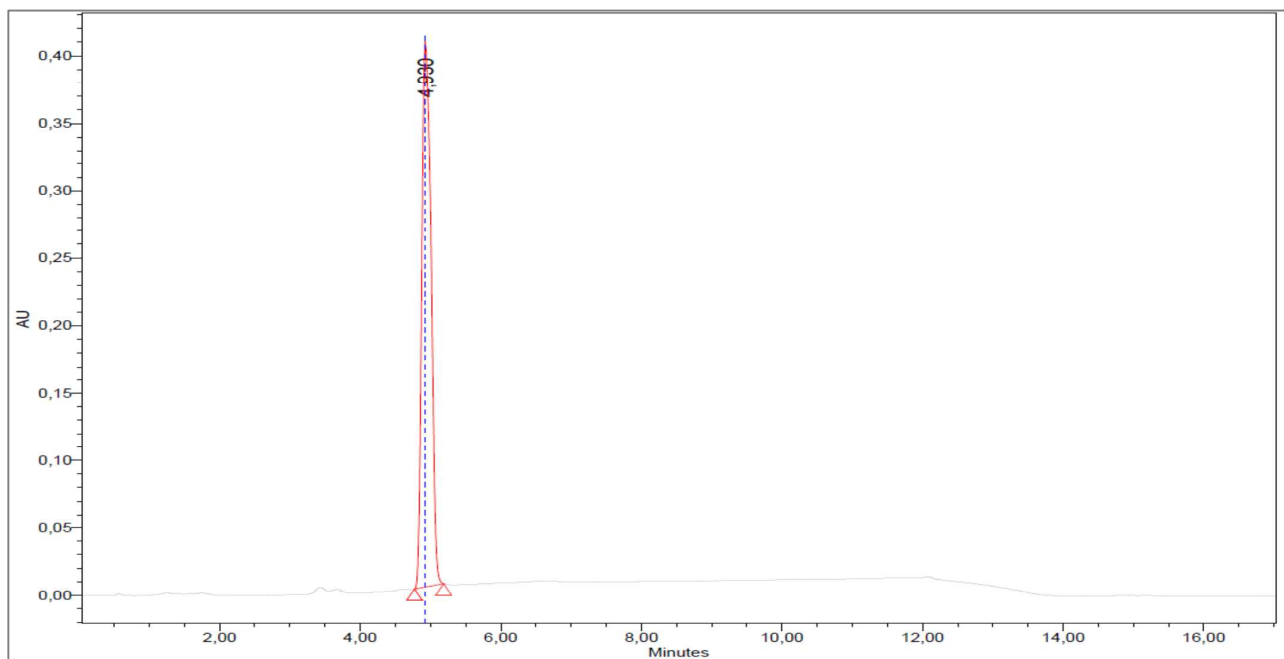
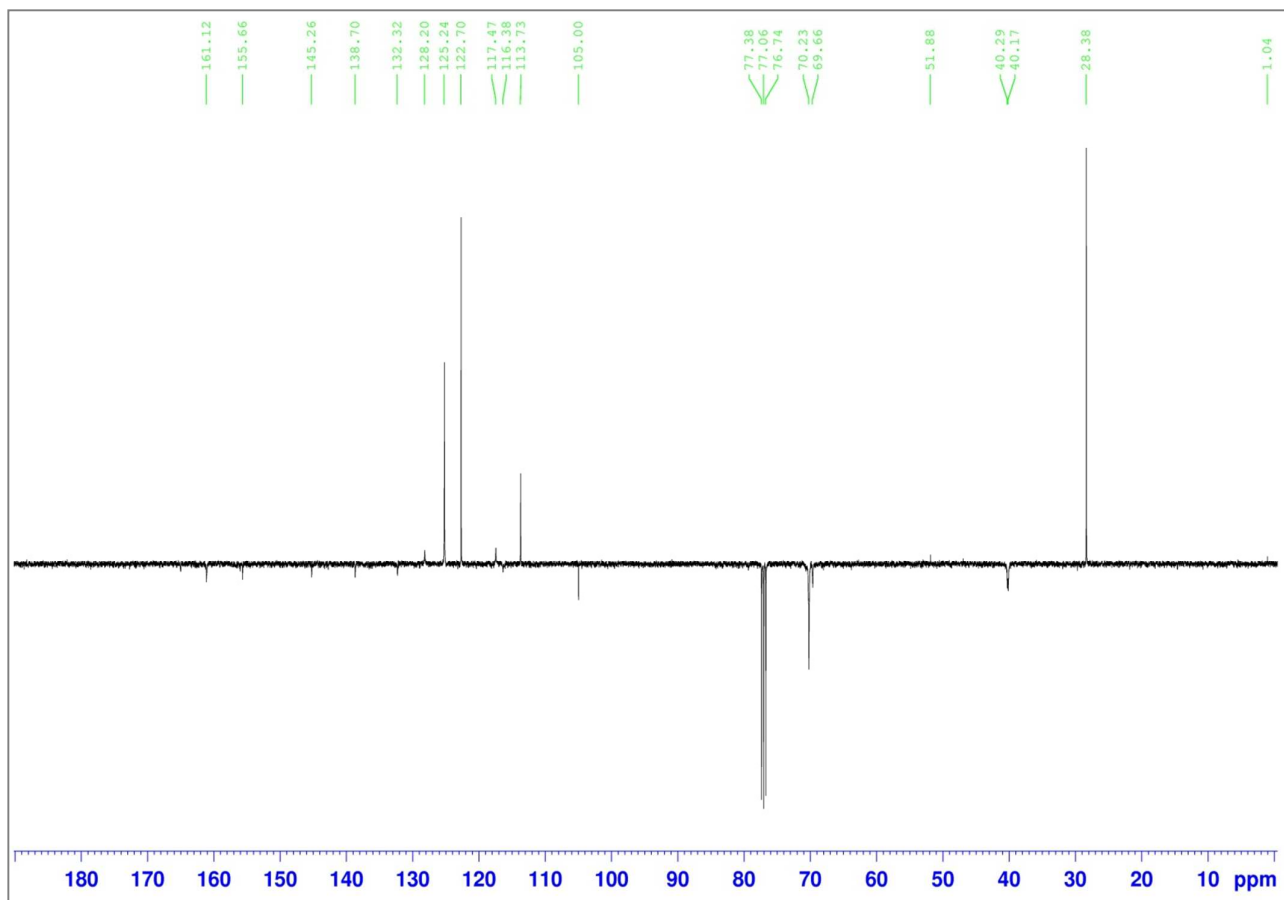
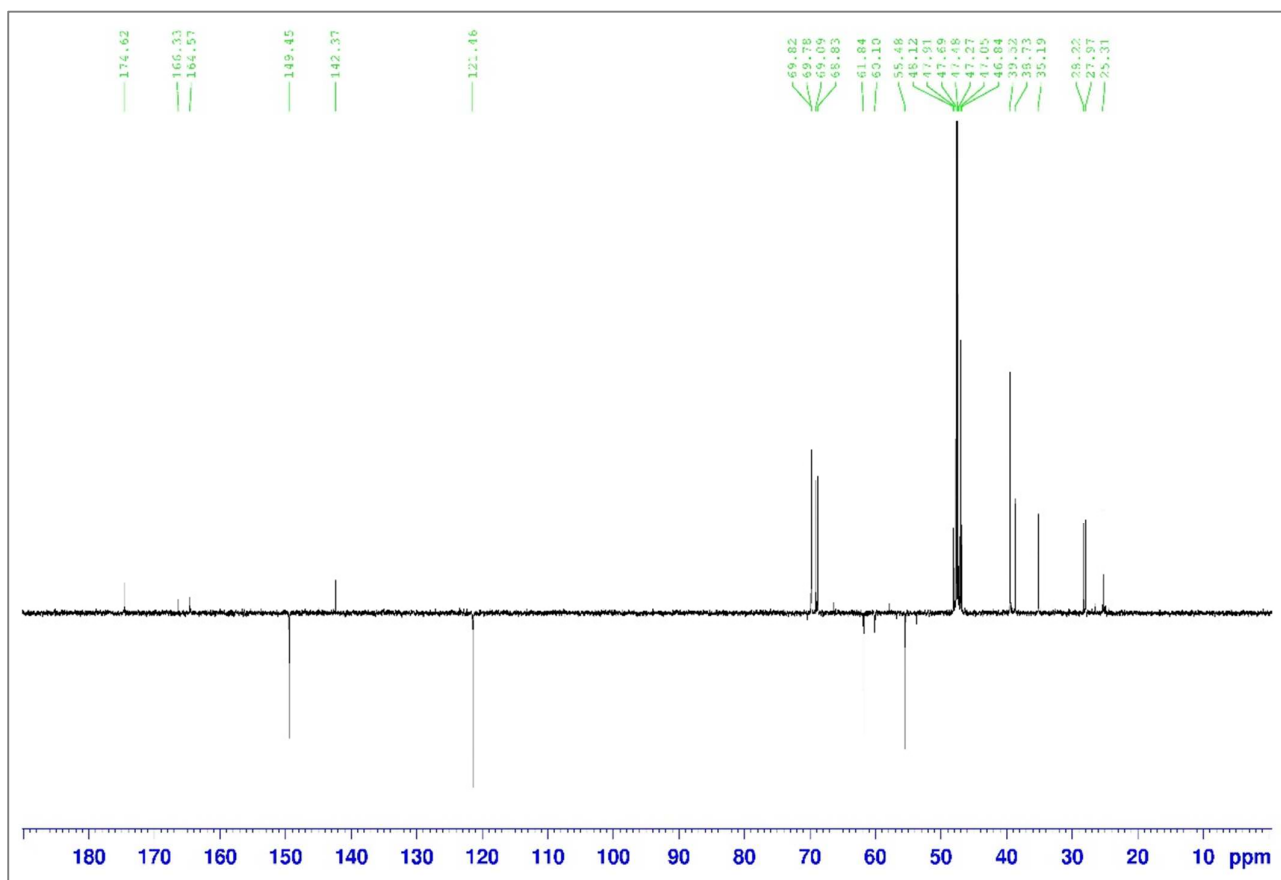
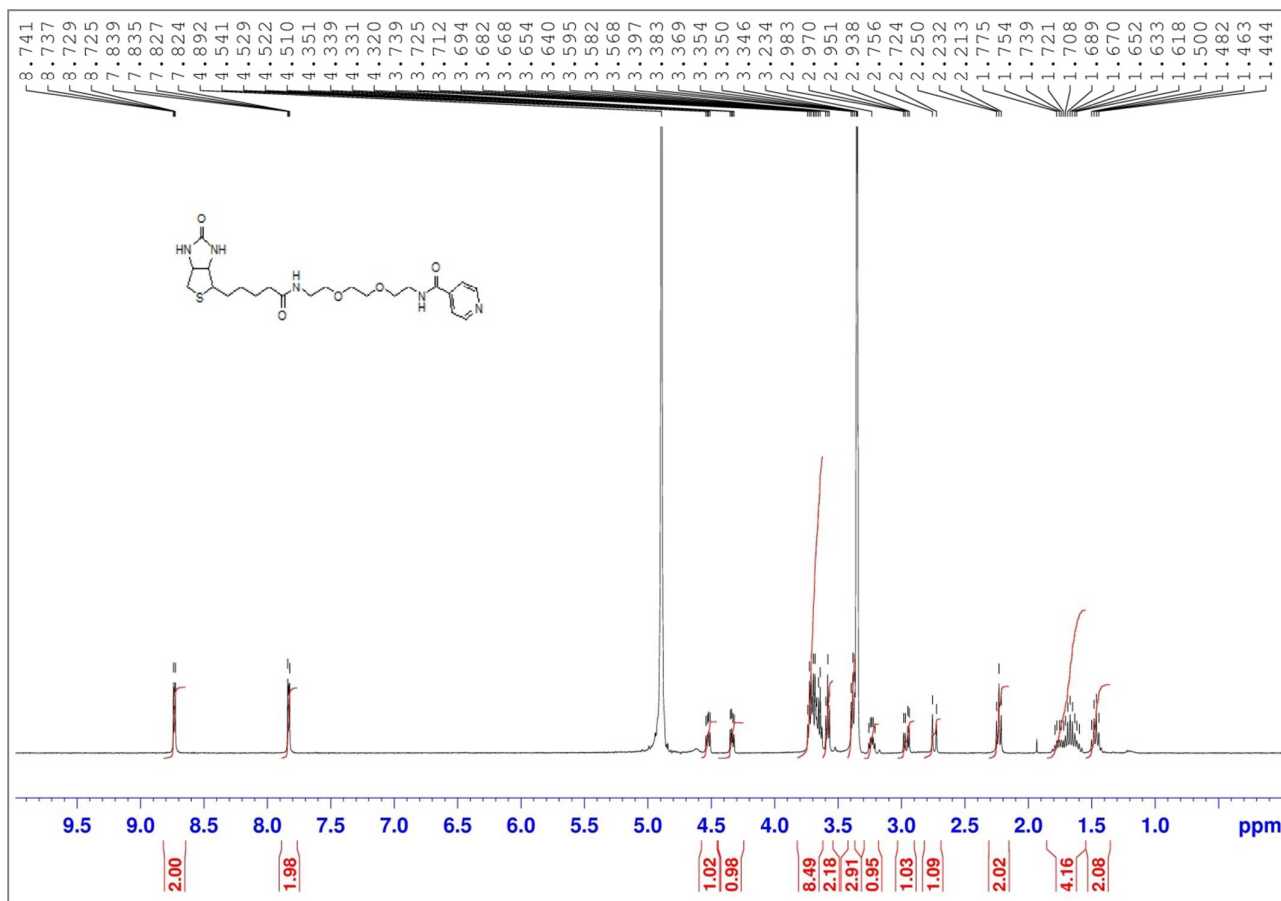


Figure. S14 Biotinylated-isoniconitamide 18



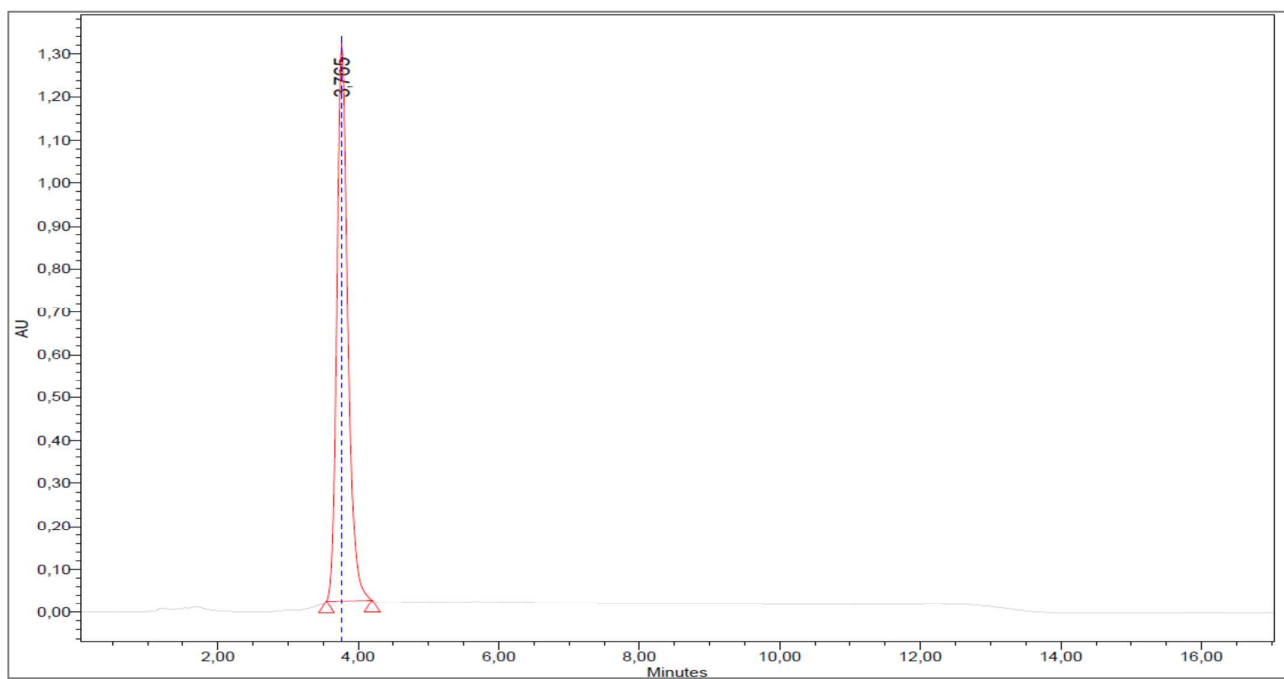
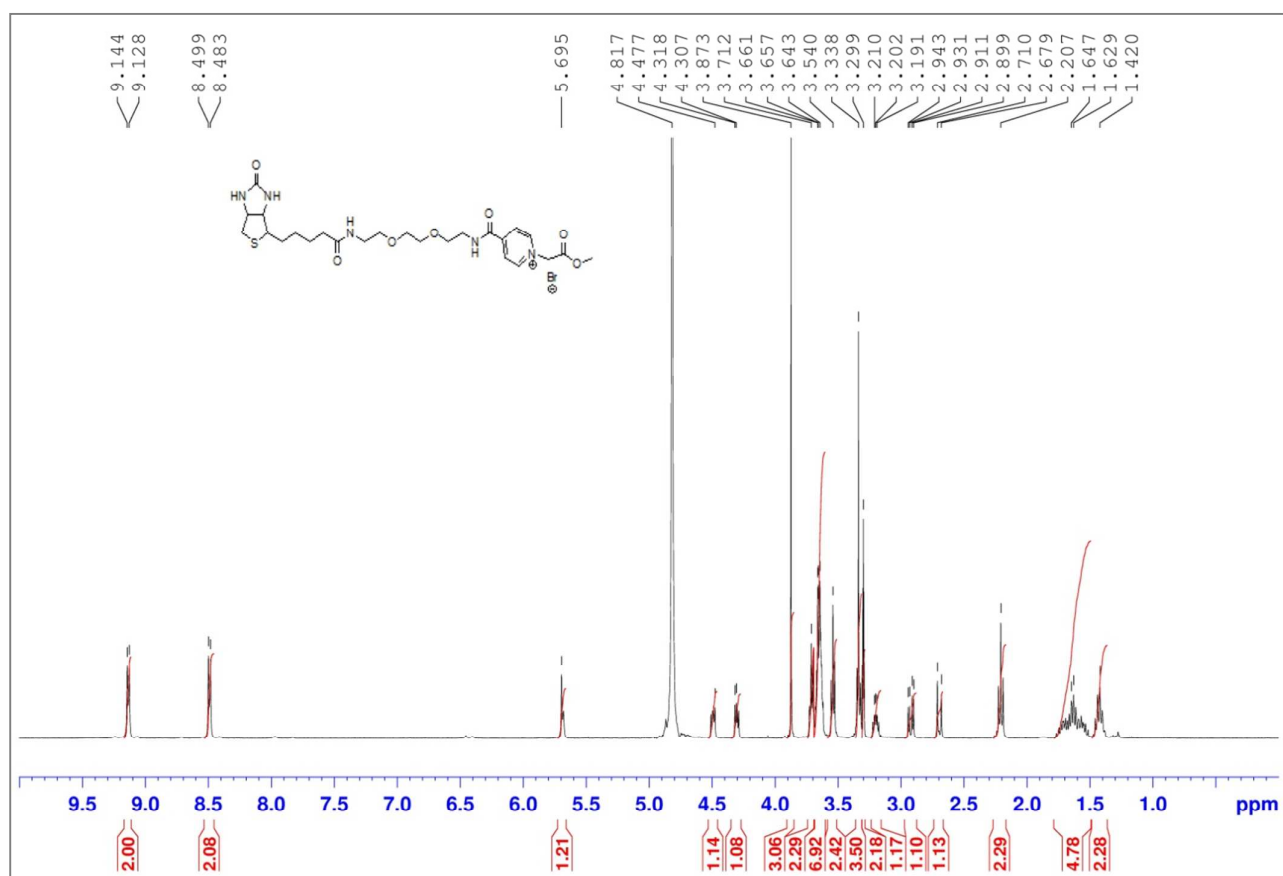


Figure. S15 Biotinylated-pyridinium bromide 19



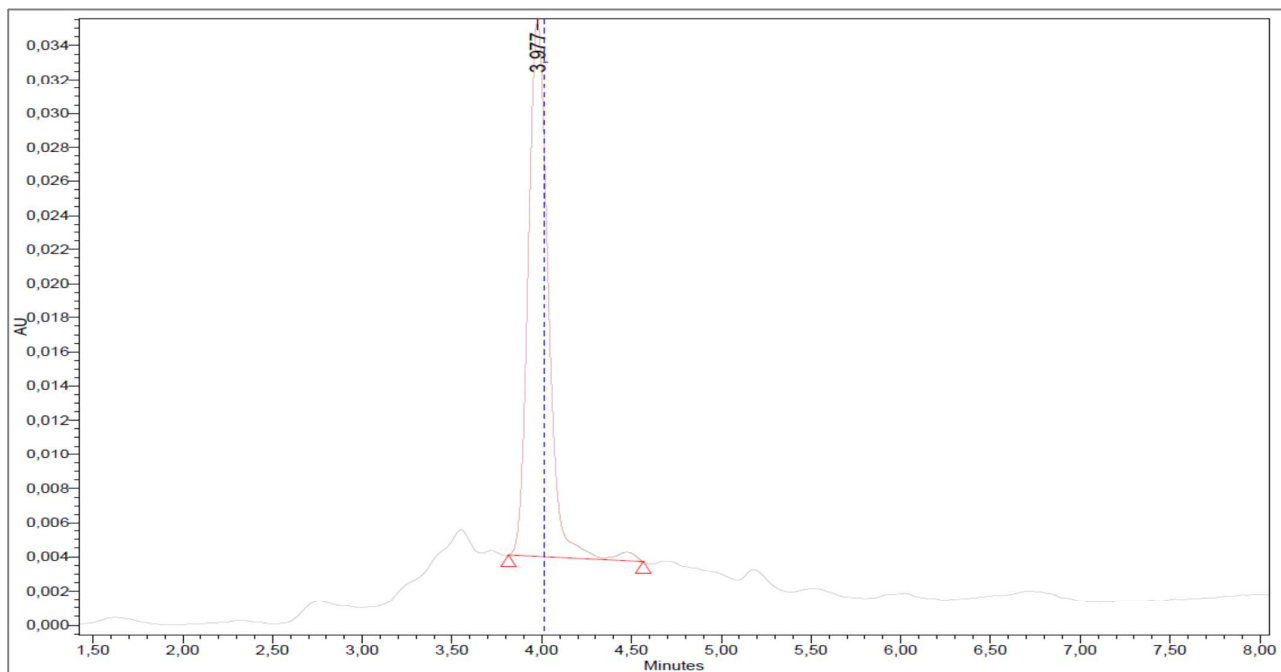
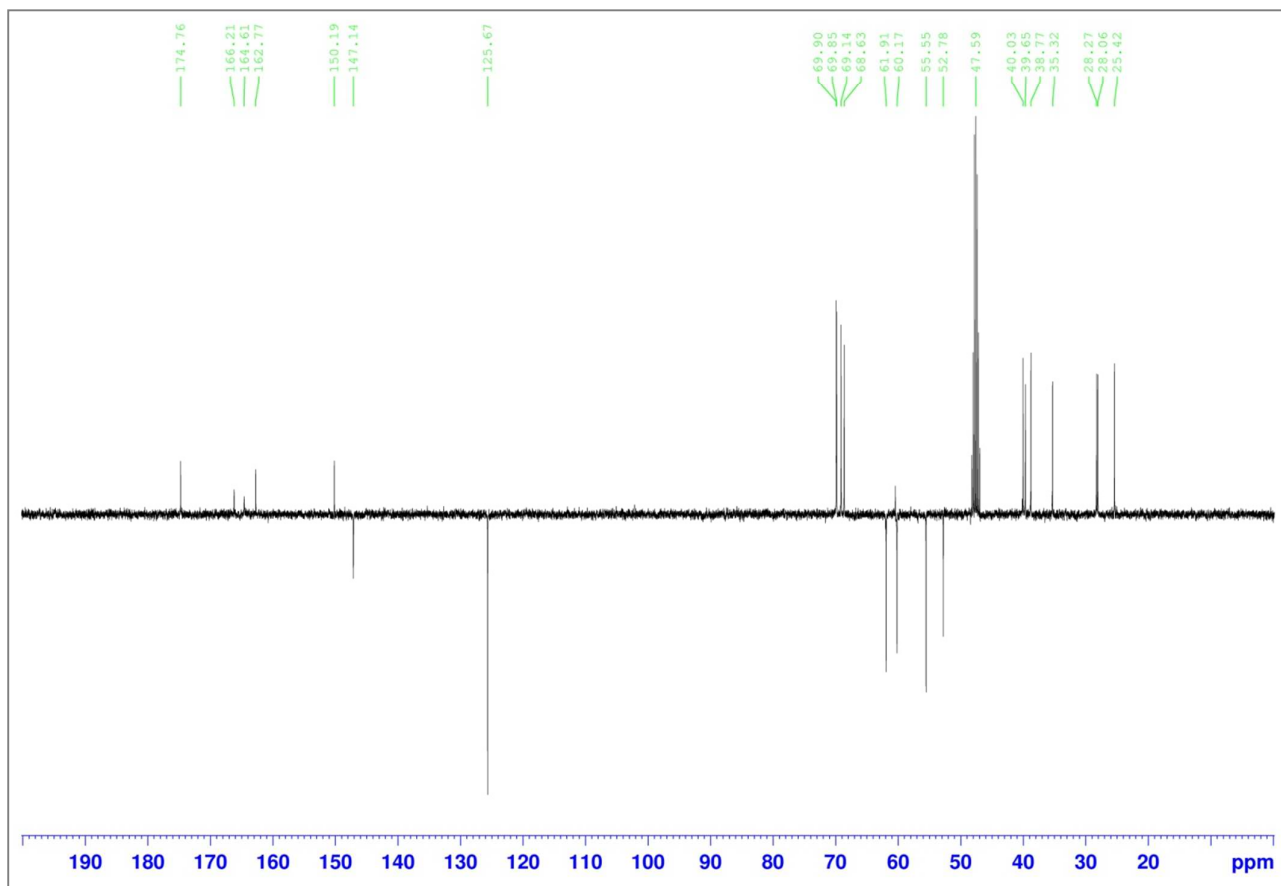
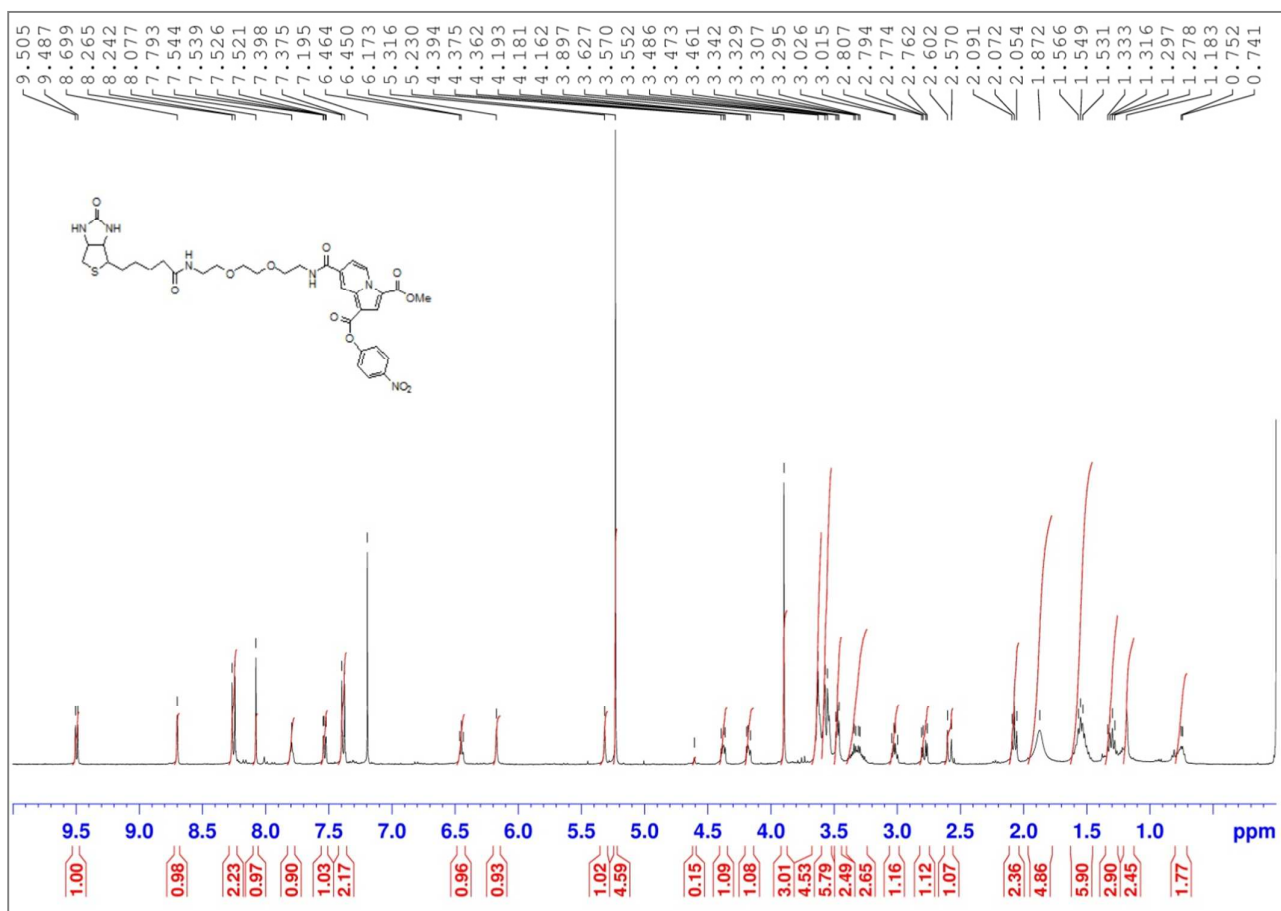


Figure. S16 Biotinylated-indolizine 20



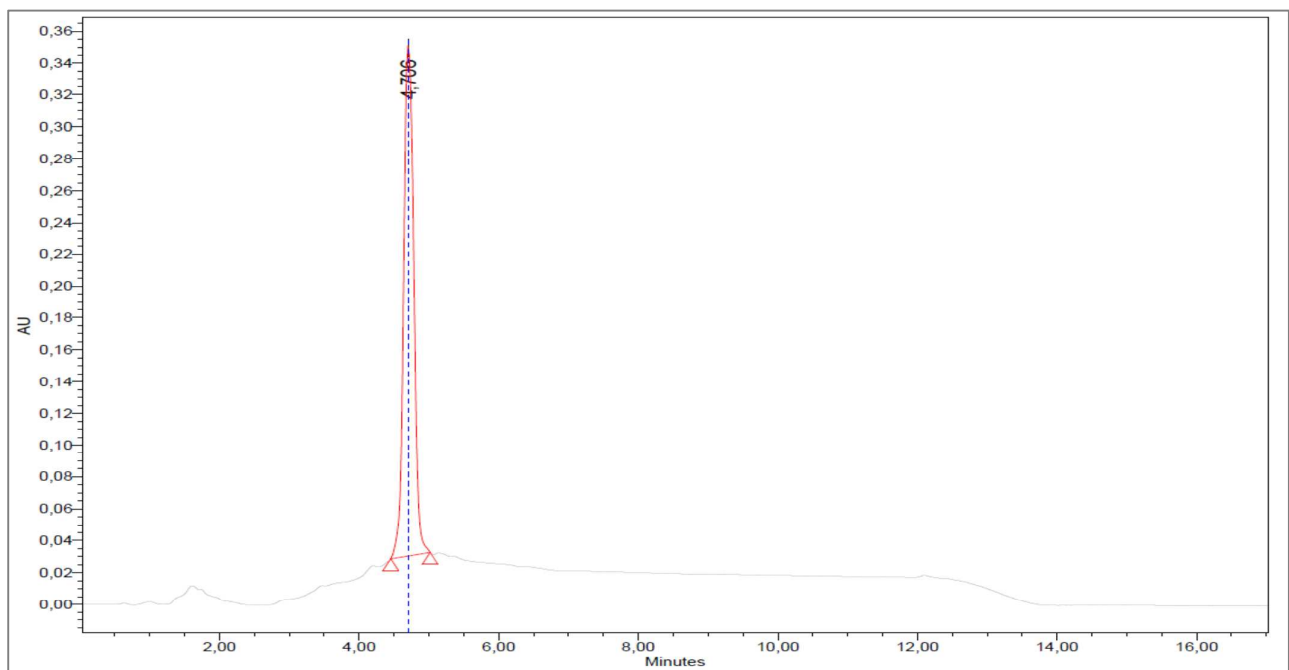
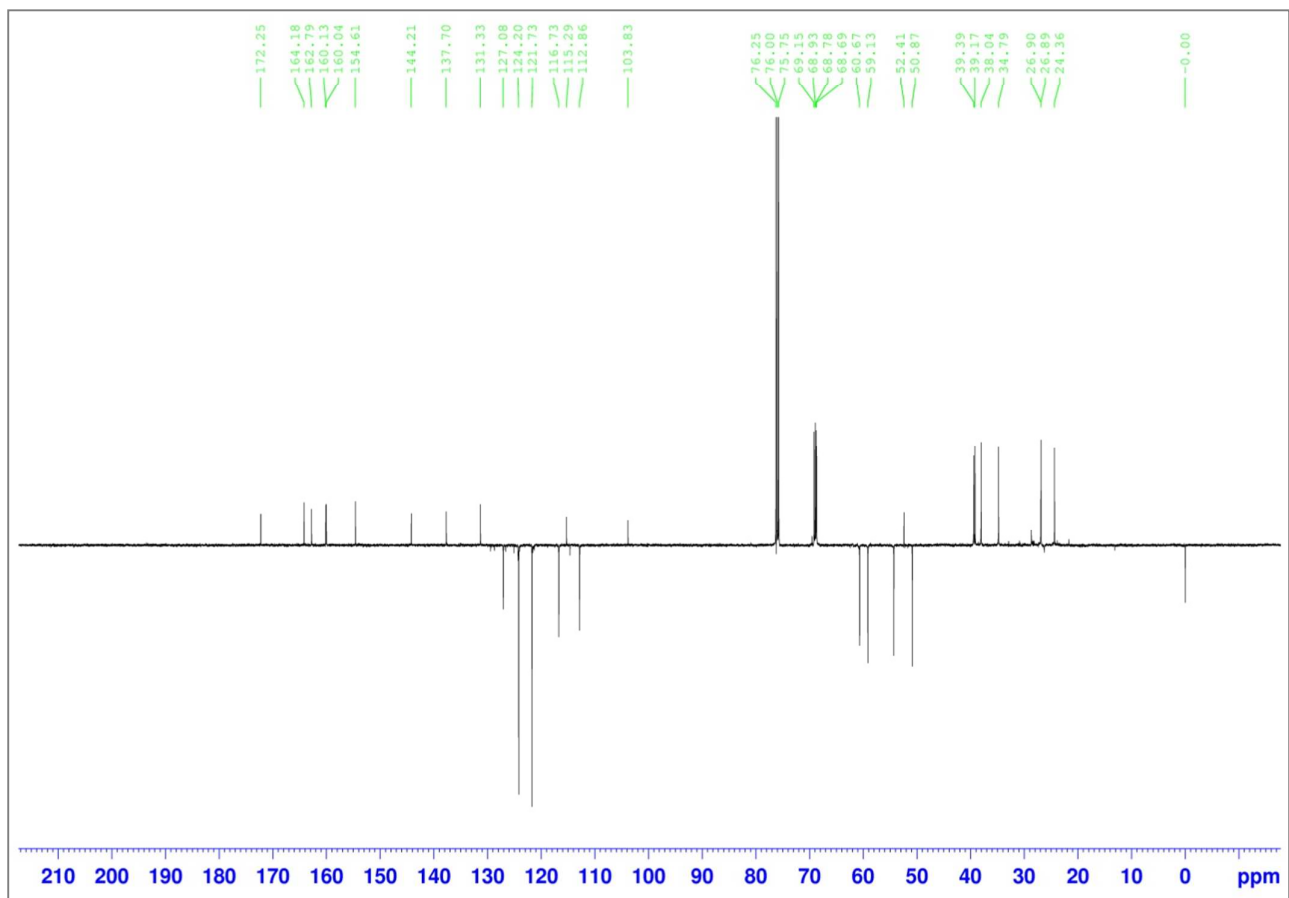


Figure. S17 Biotinylated-COB223

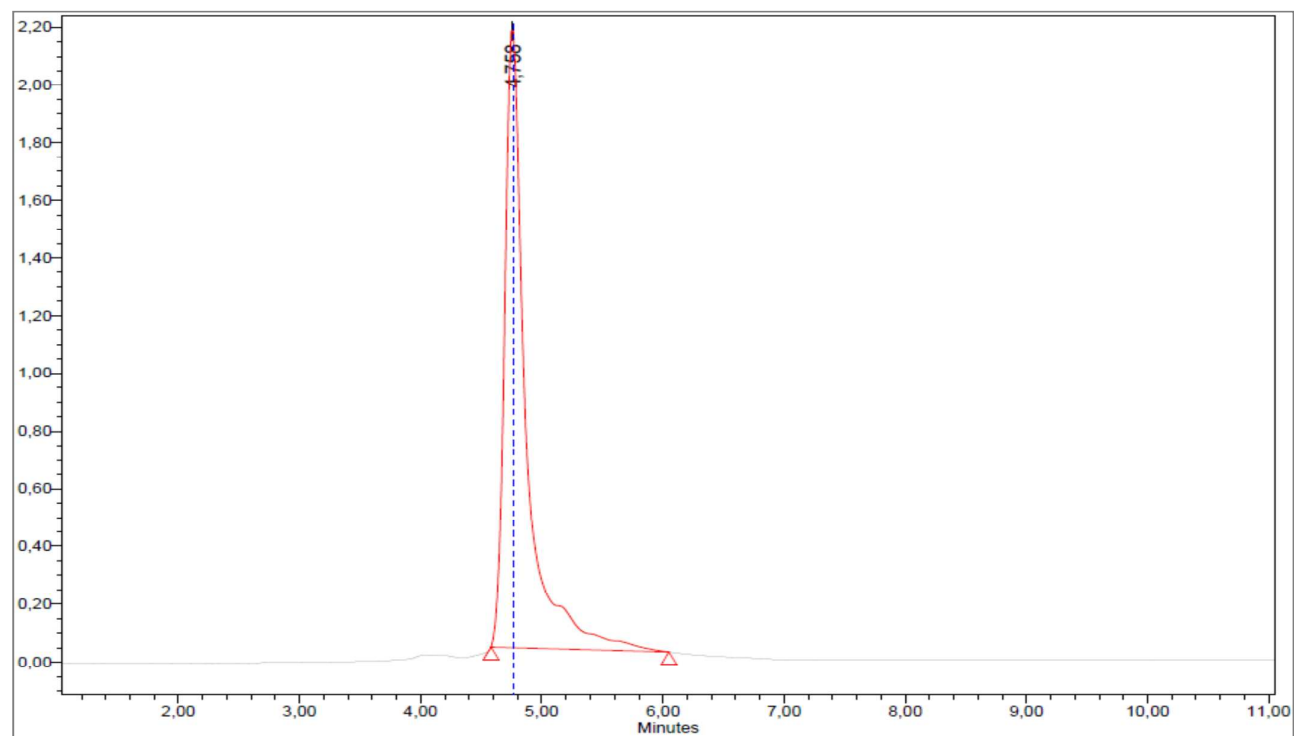
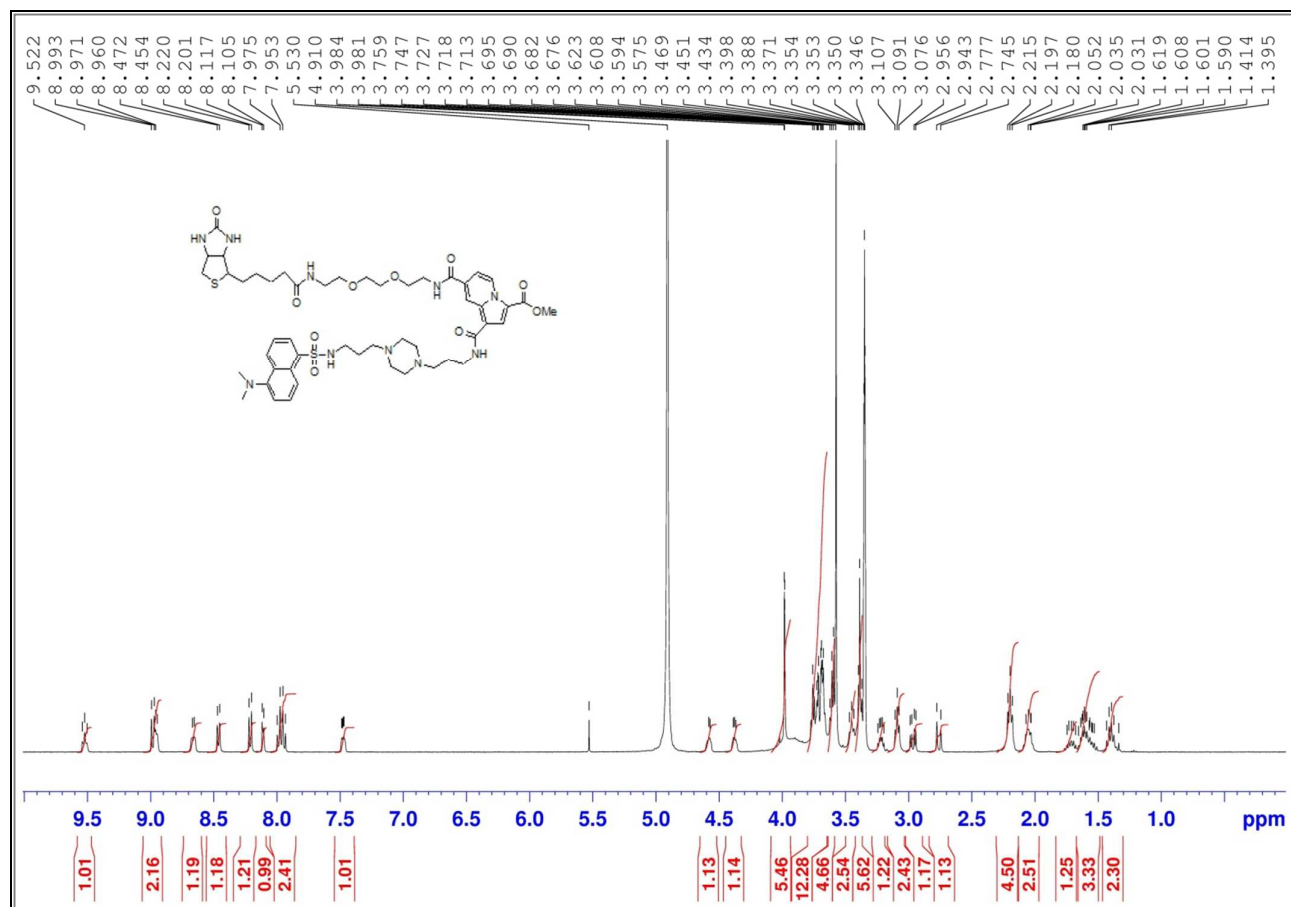


Figure. S18 Biotinylated-COB236

