Supporting Information

Syntheses of Morpholine based Nucleotide Analogs for Hepatic siRNA-Targeting and Stabilization

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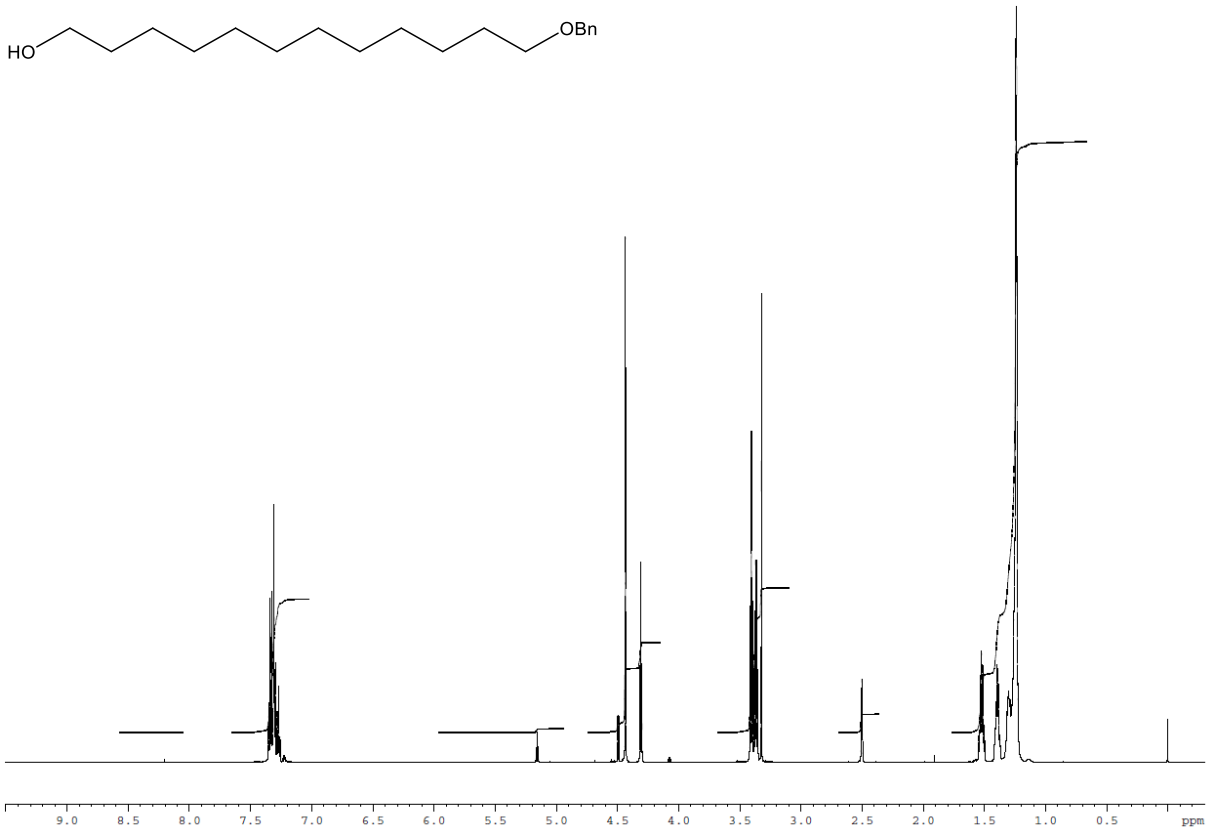
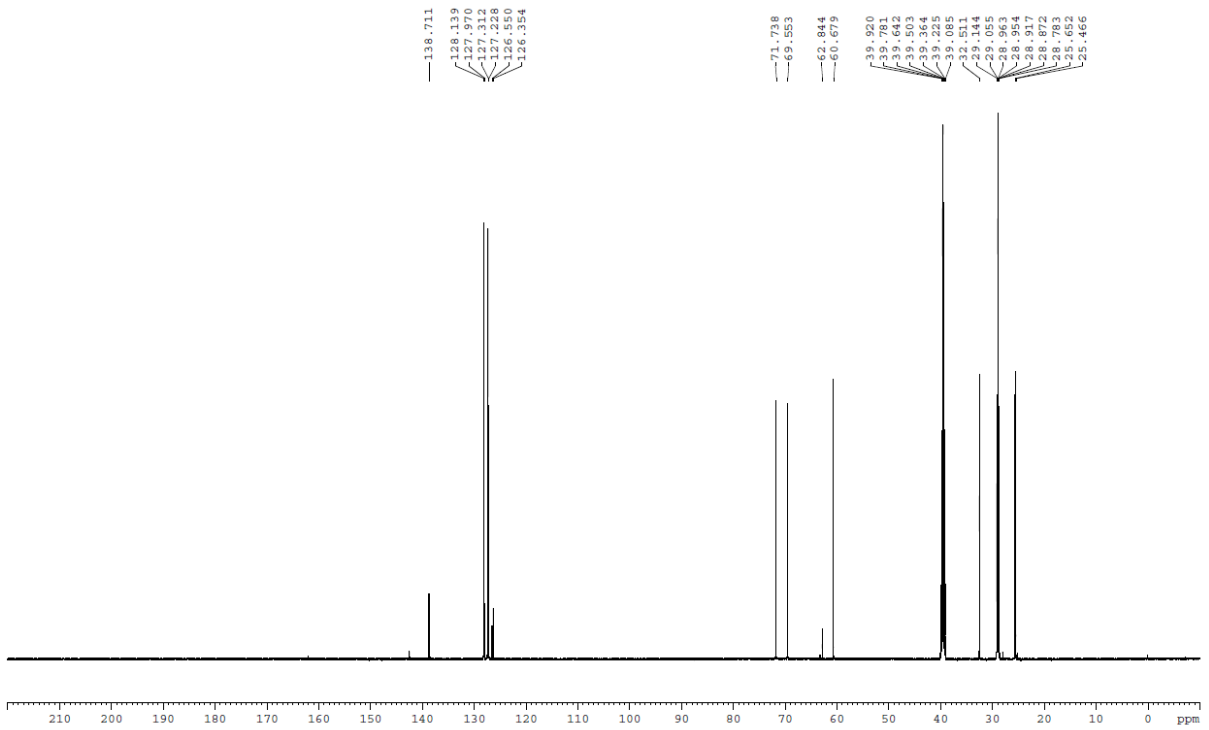
NMR-spectra **11b**





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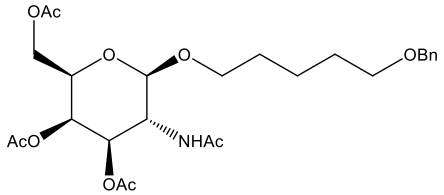
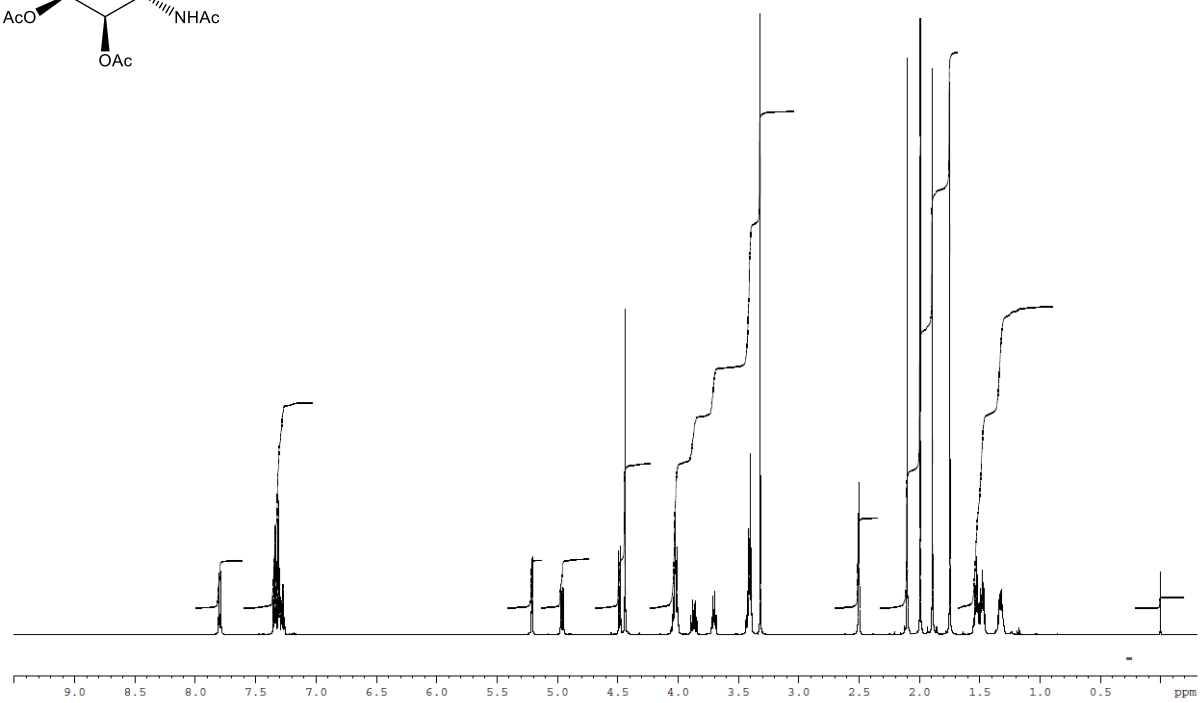
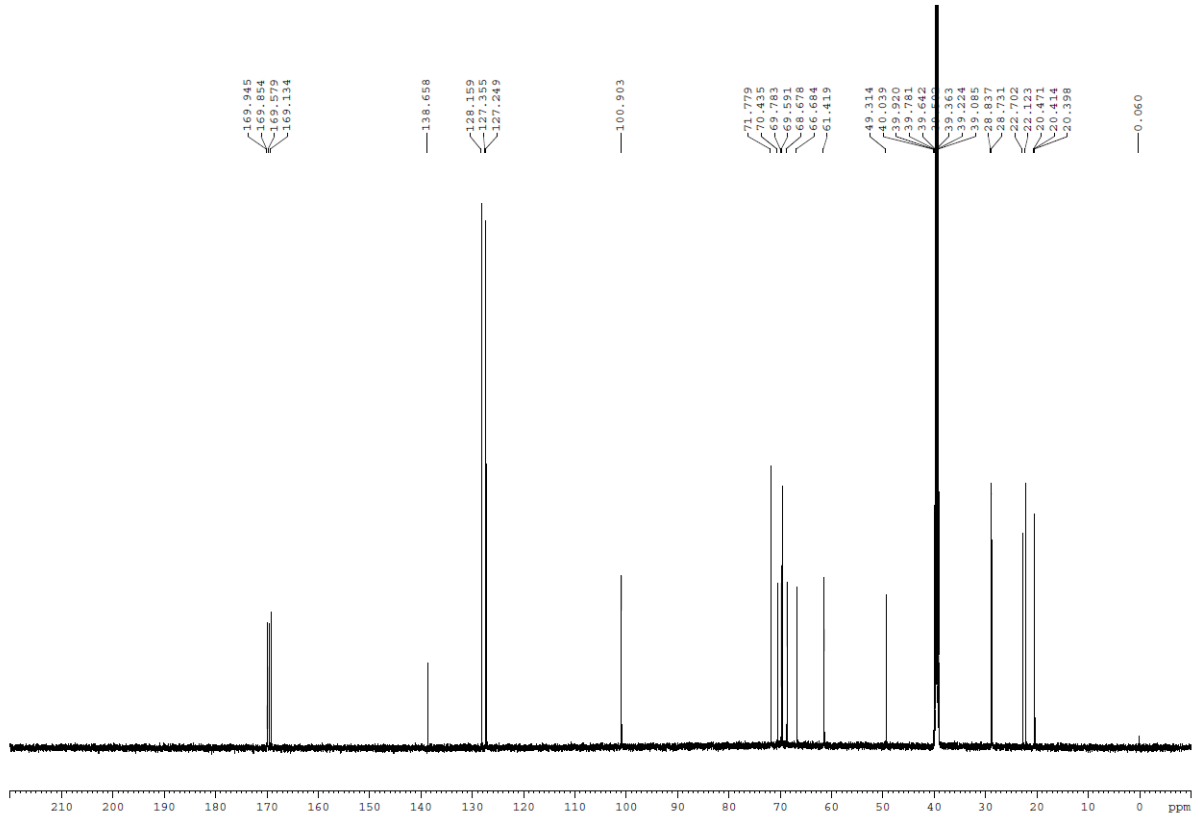
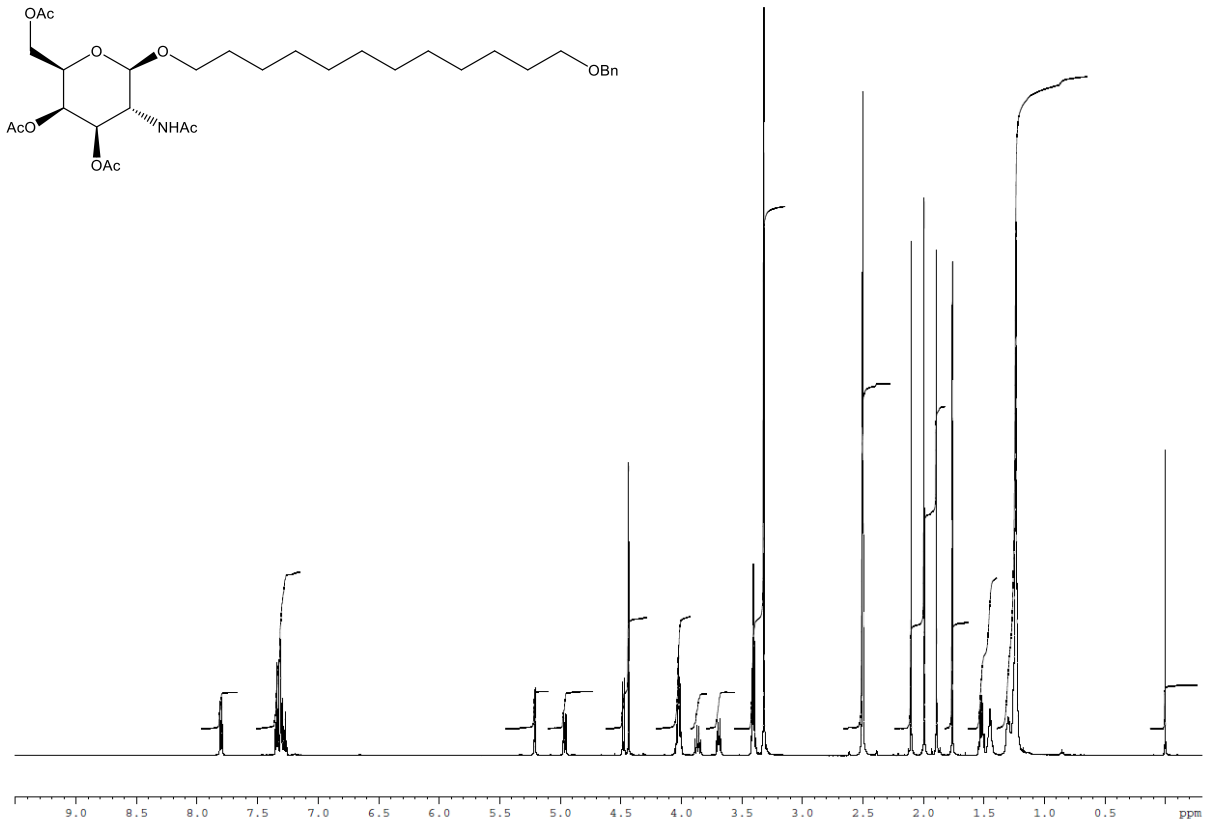
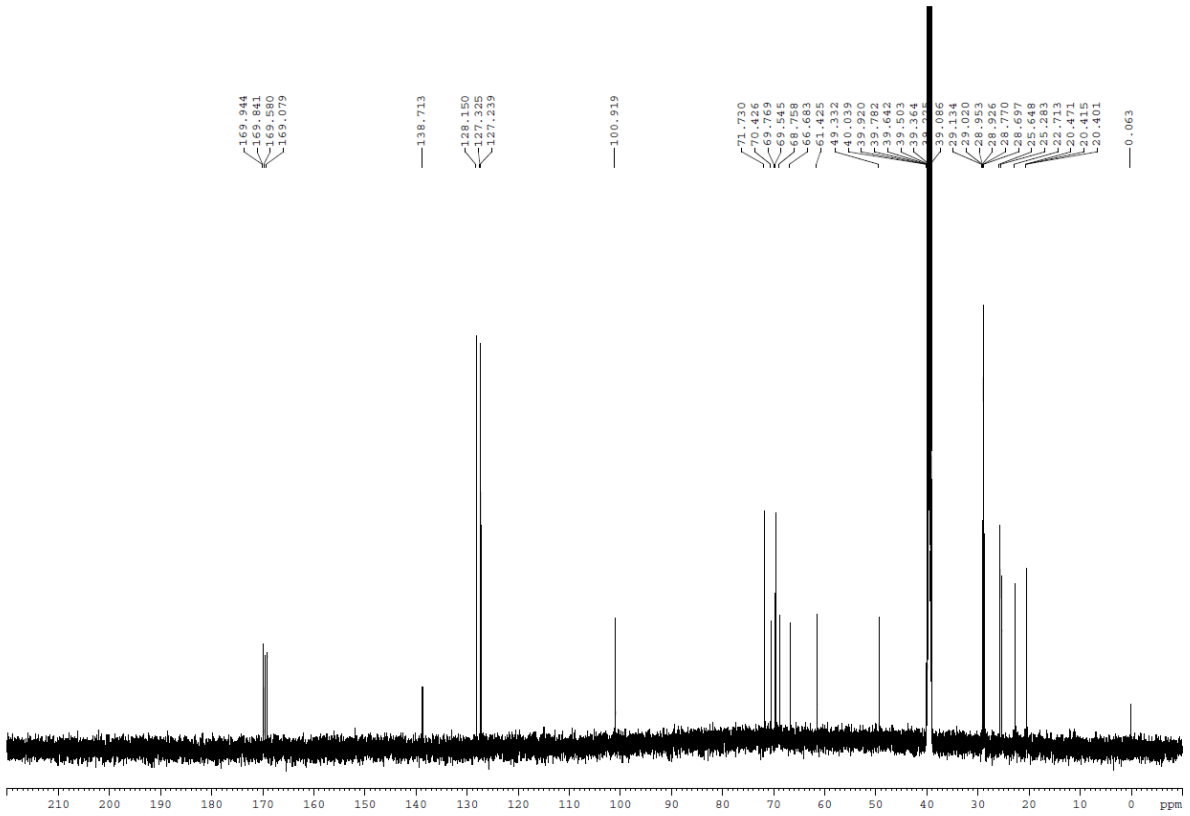




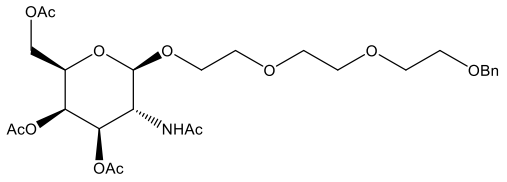


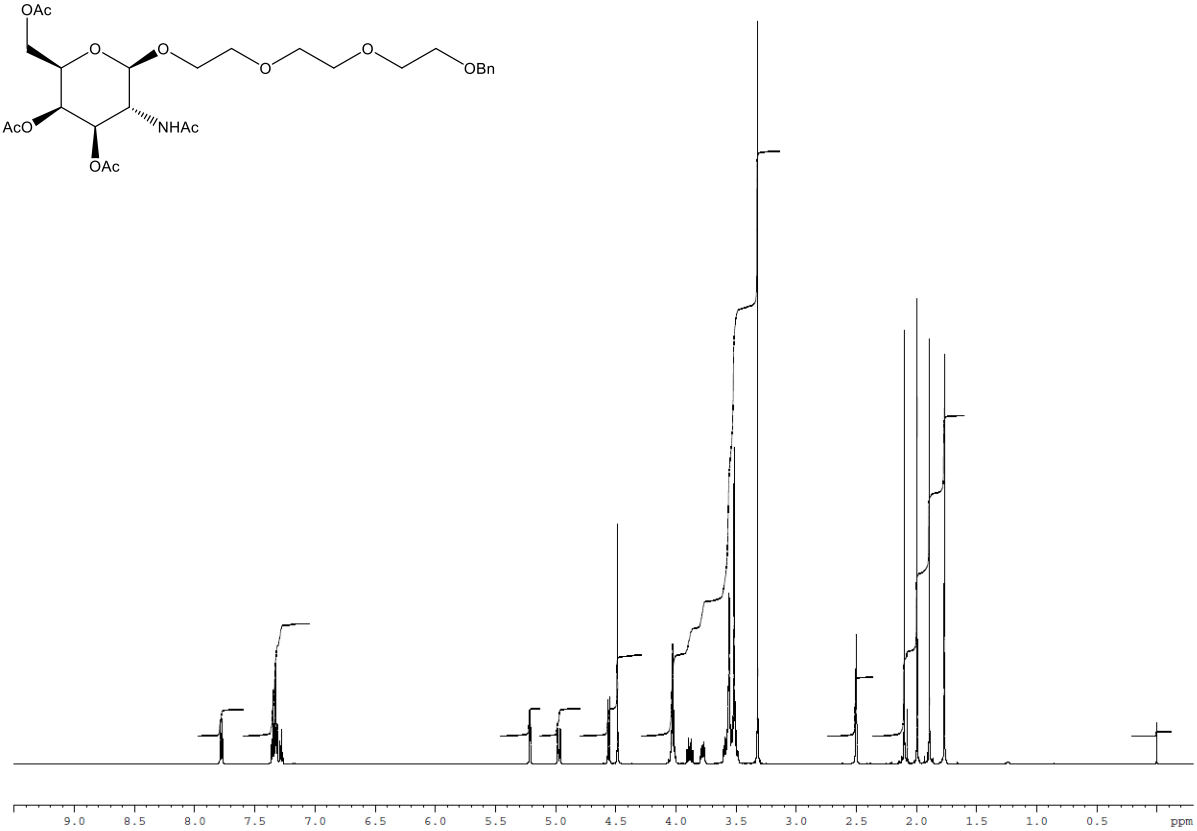
NMR-spectra **12b**

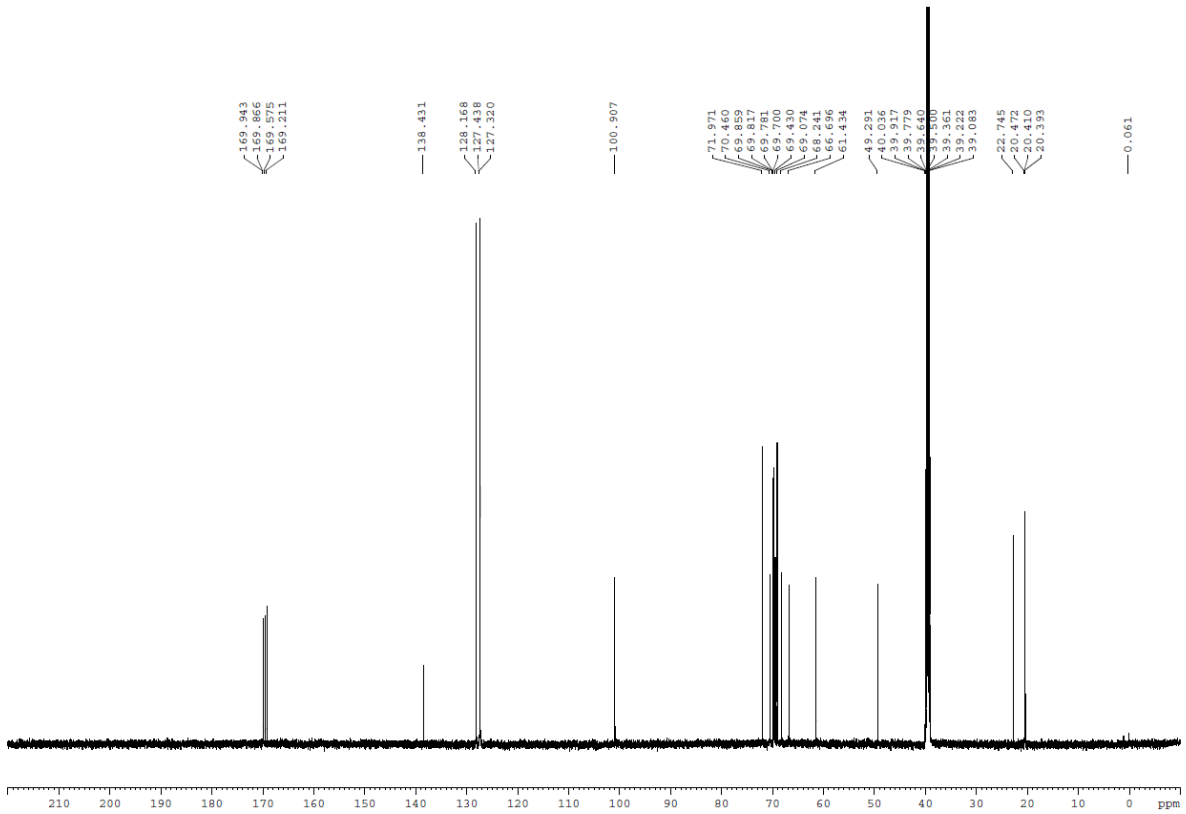




NMR-spectra **12c**







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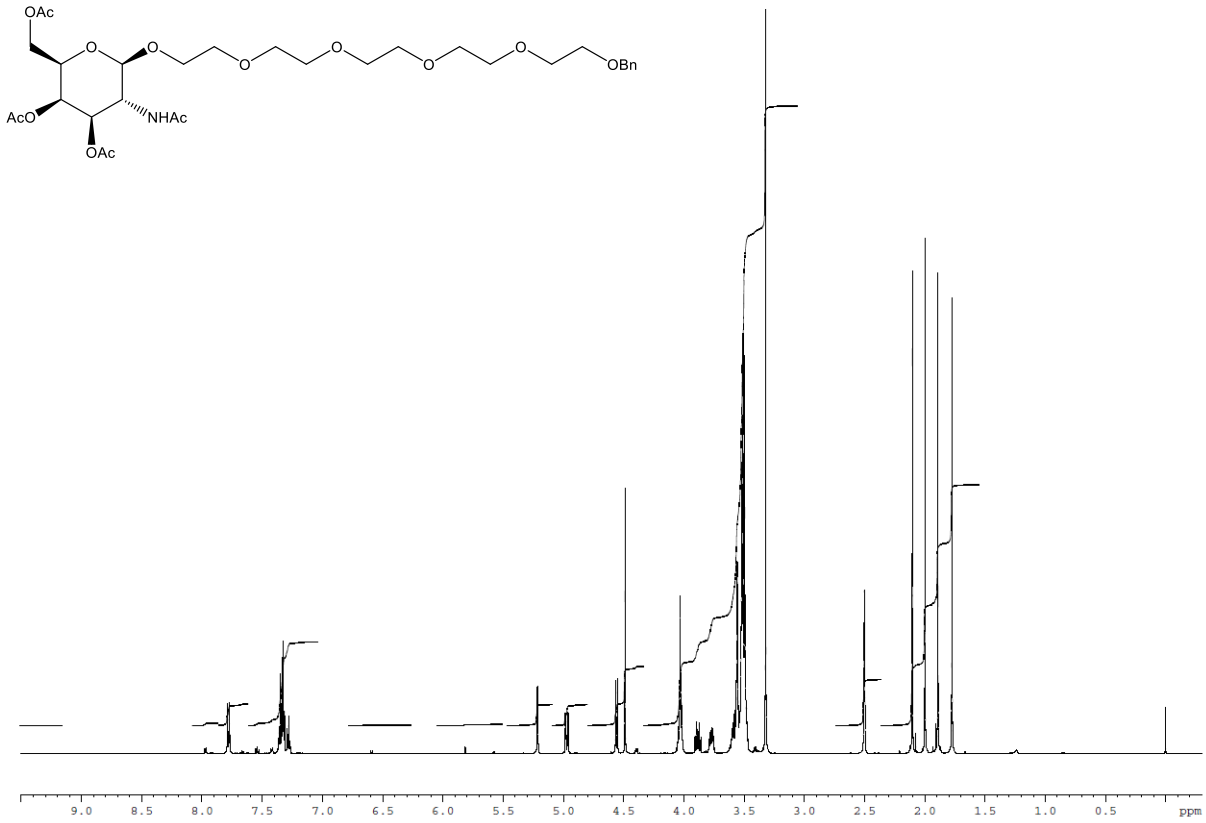
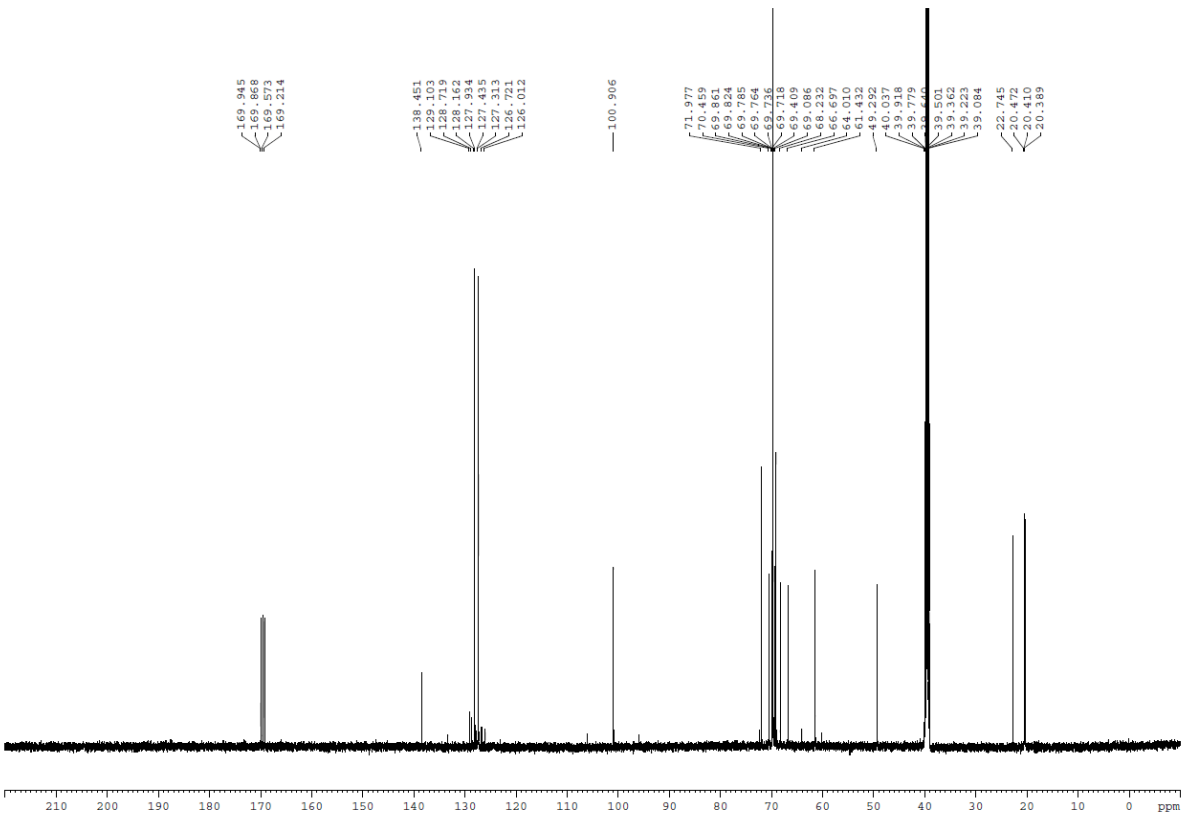
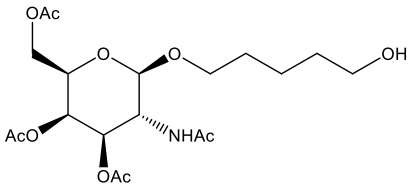
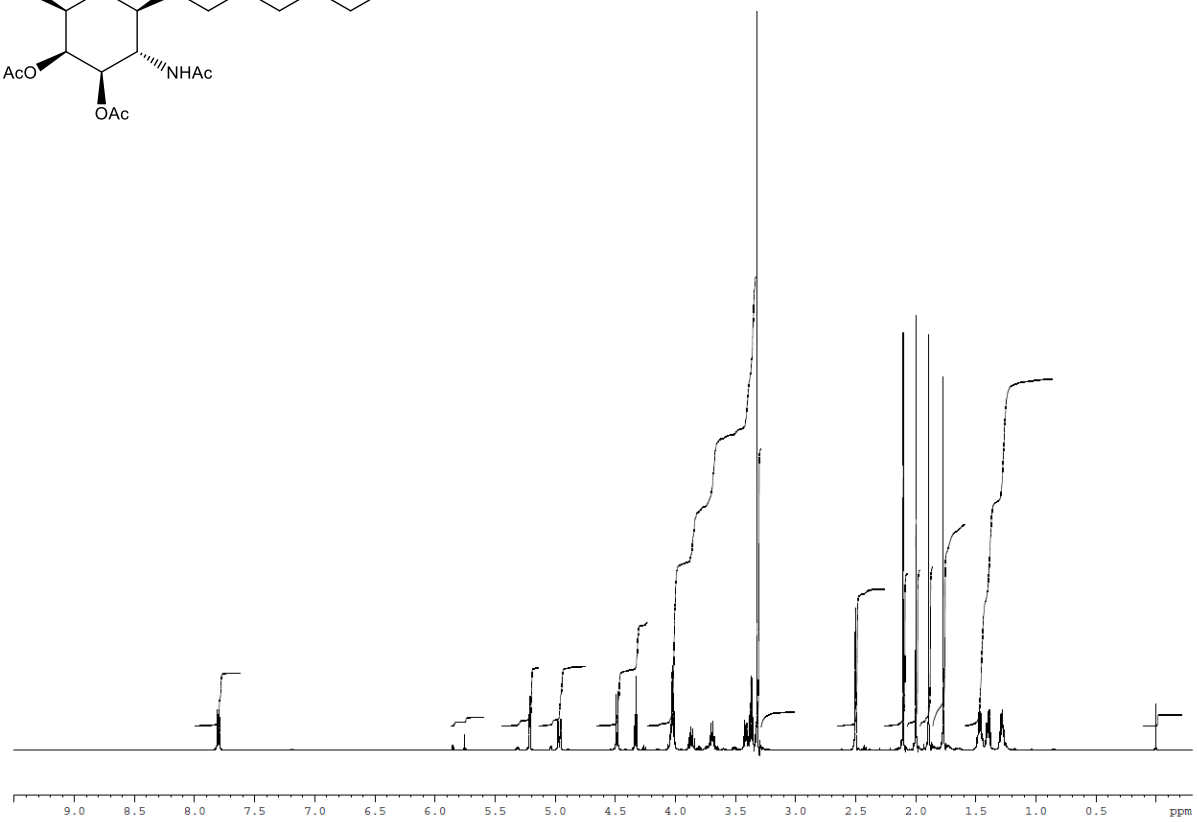
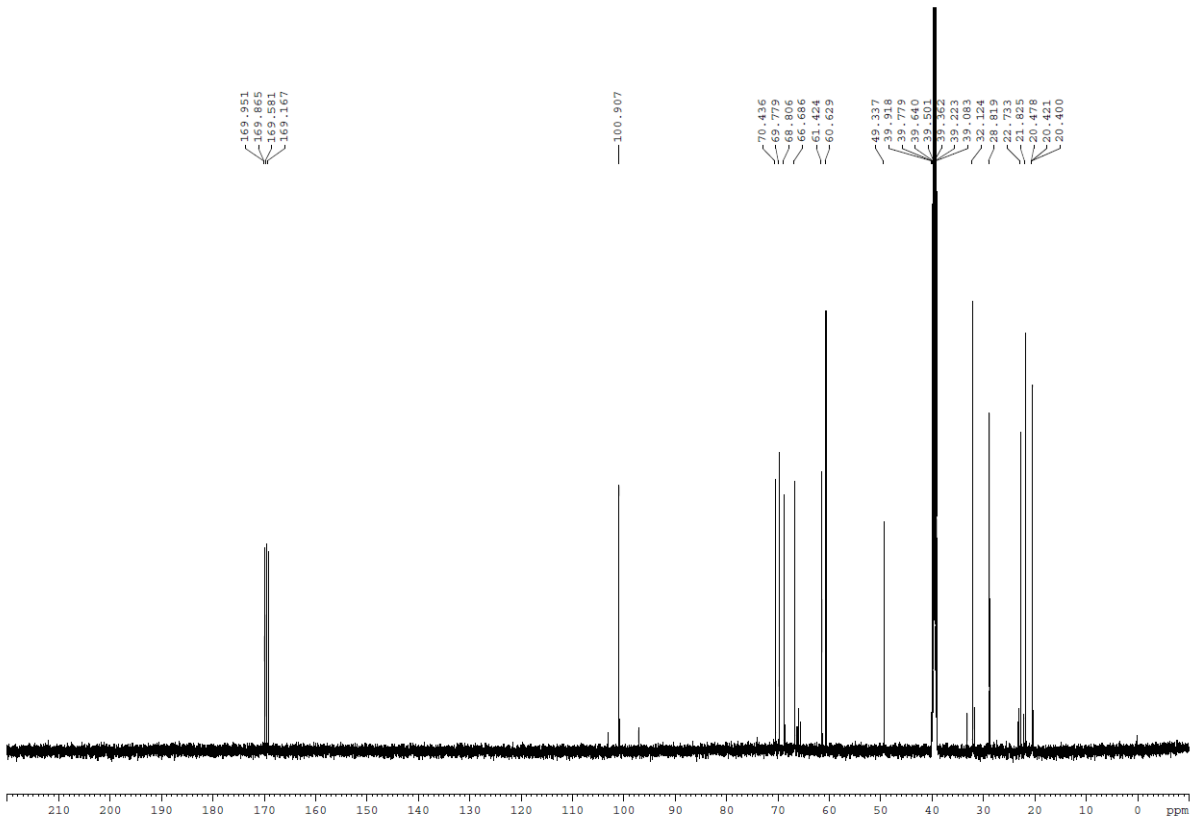




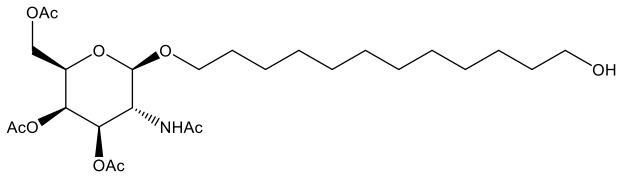
NMR-spectra **13a**

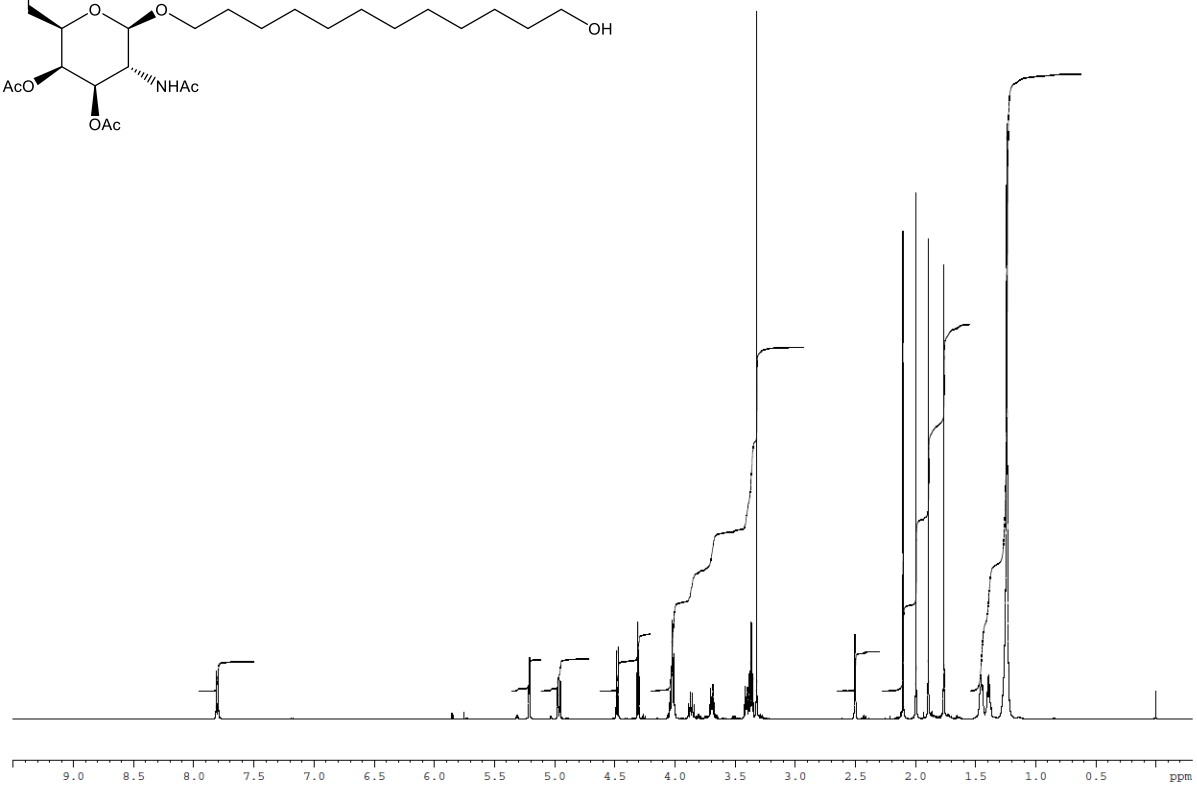


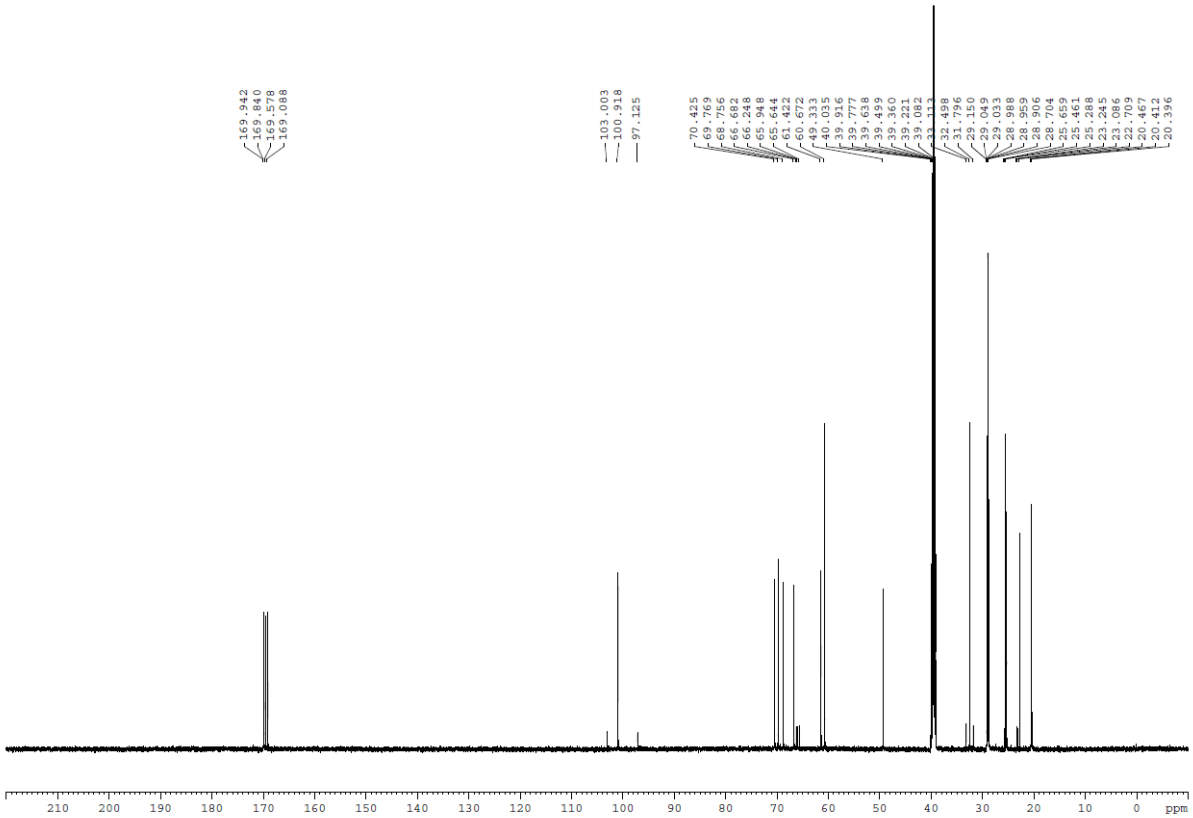




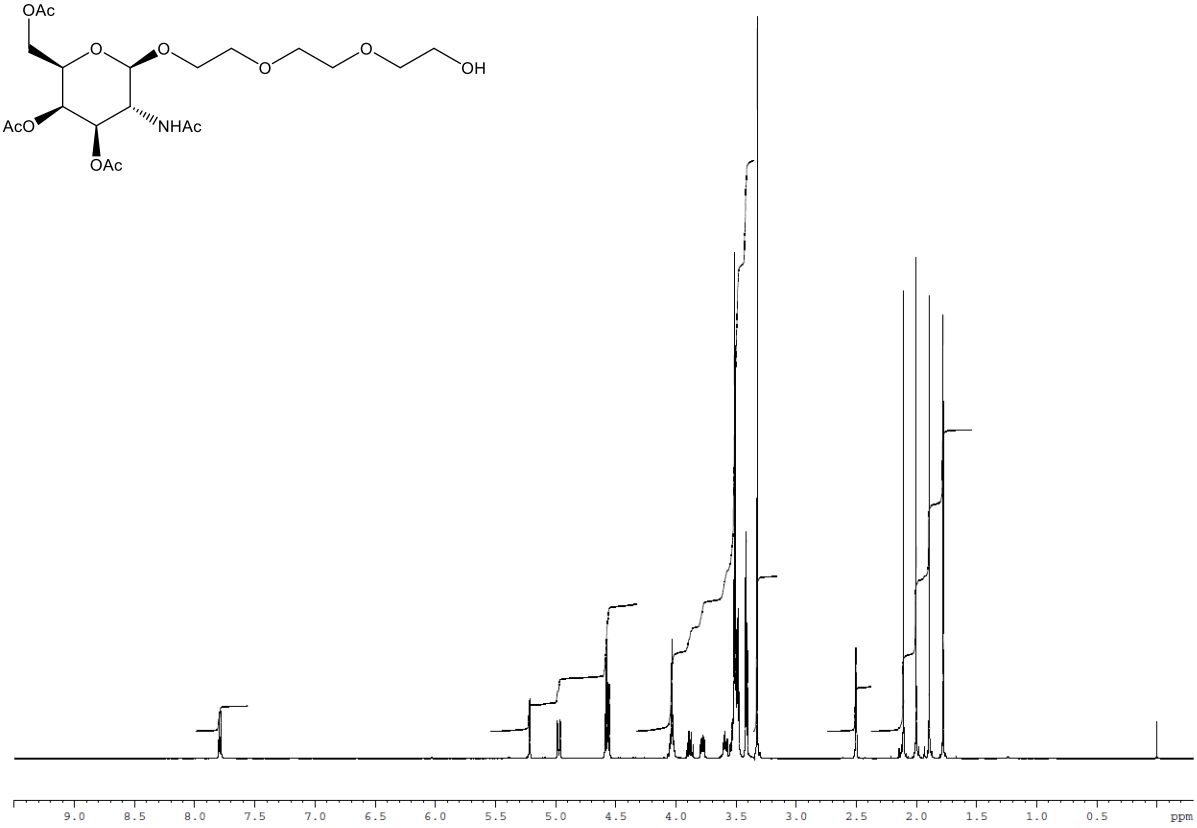
NMR-spectra **13b**

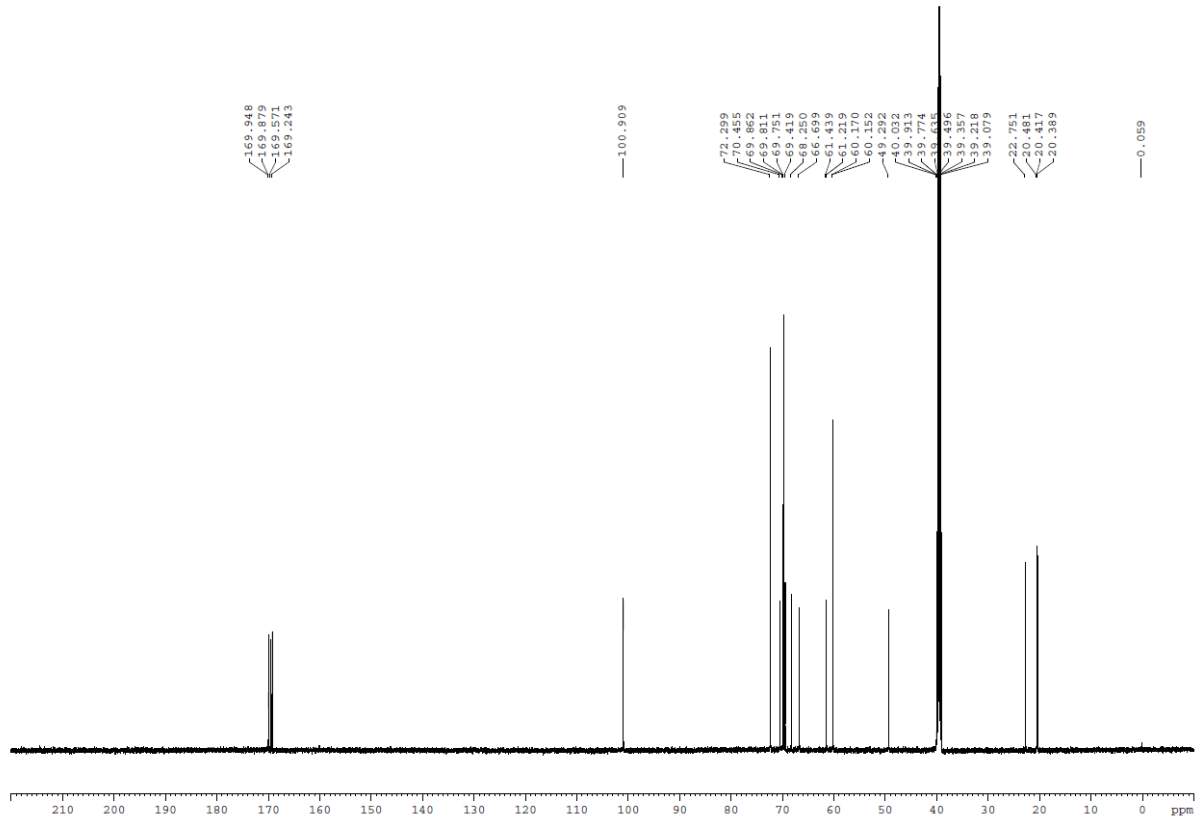




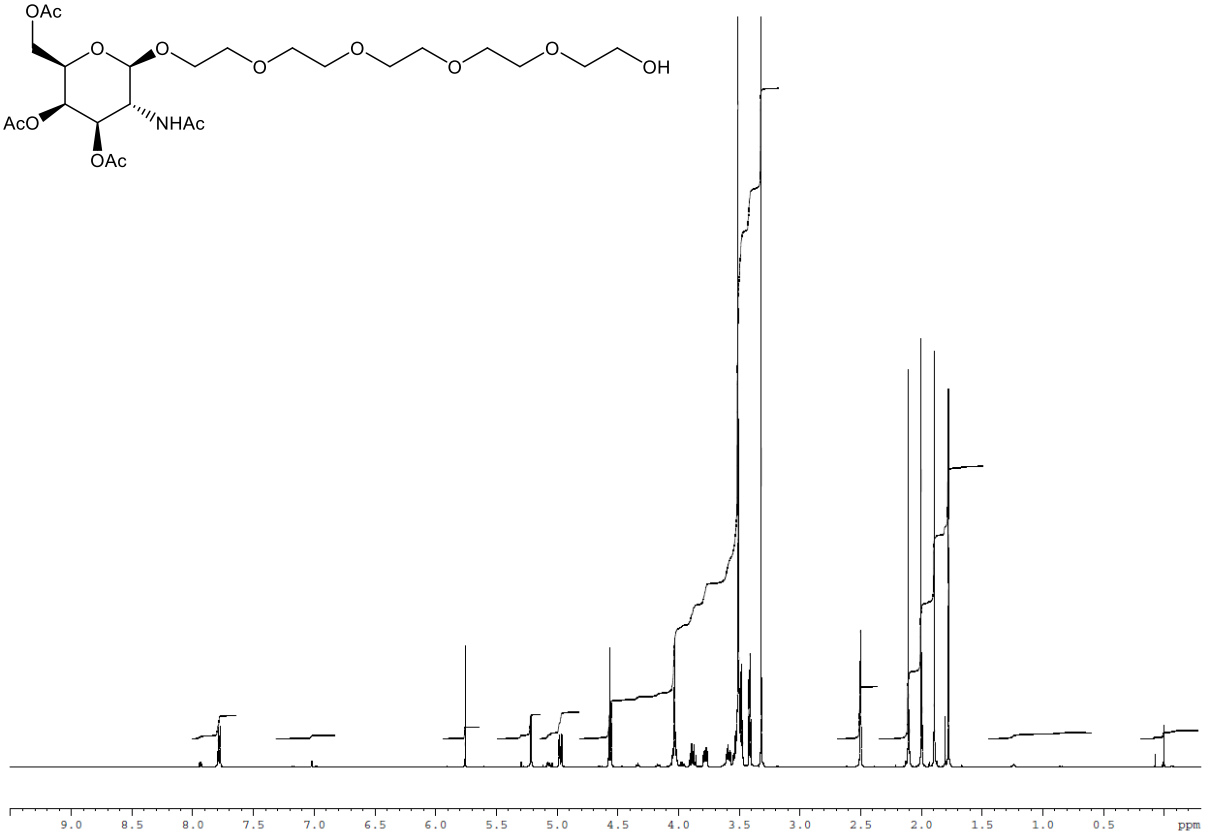


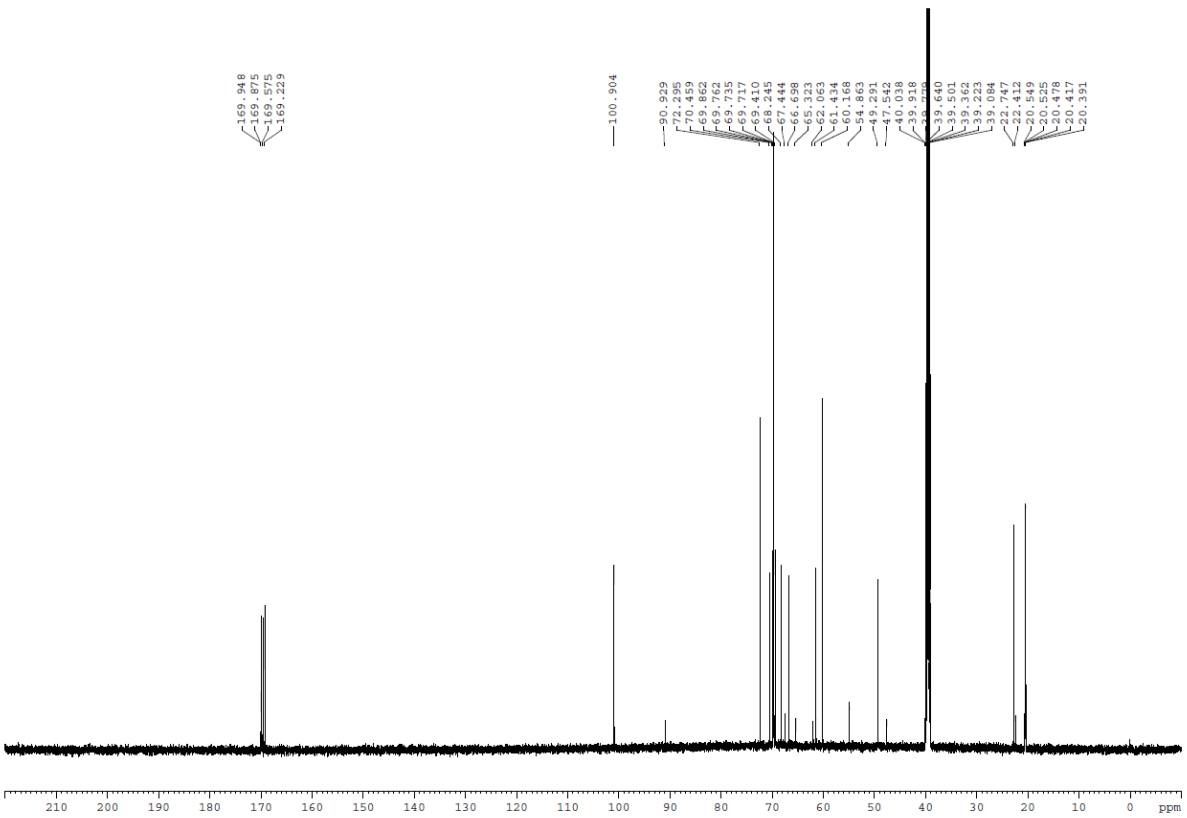
NMR-spectra **13c**



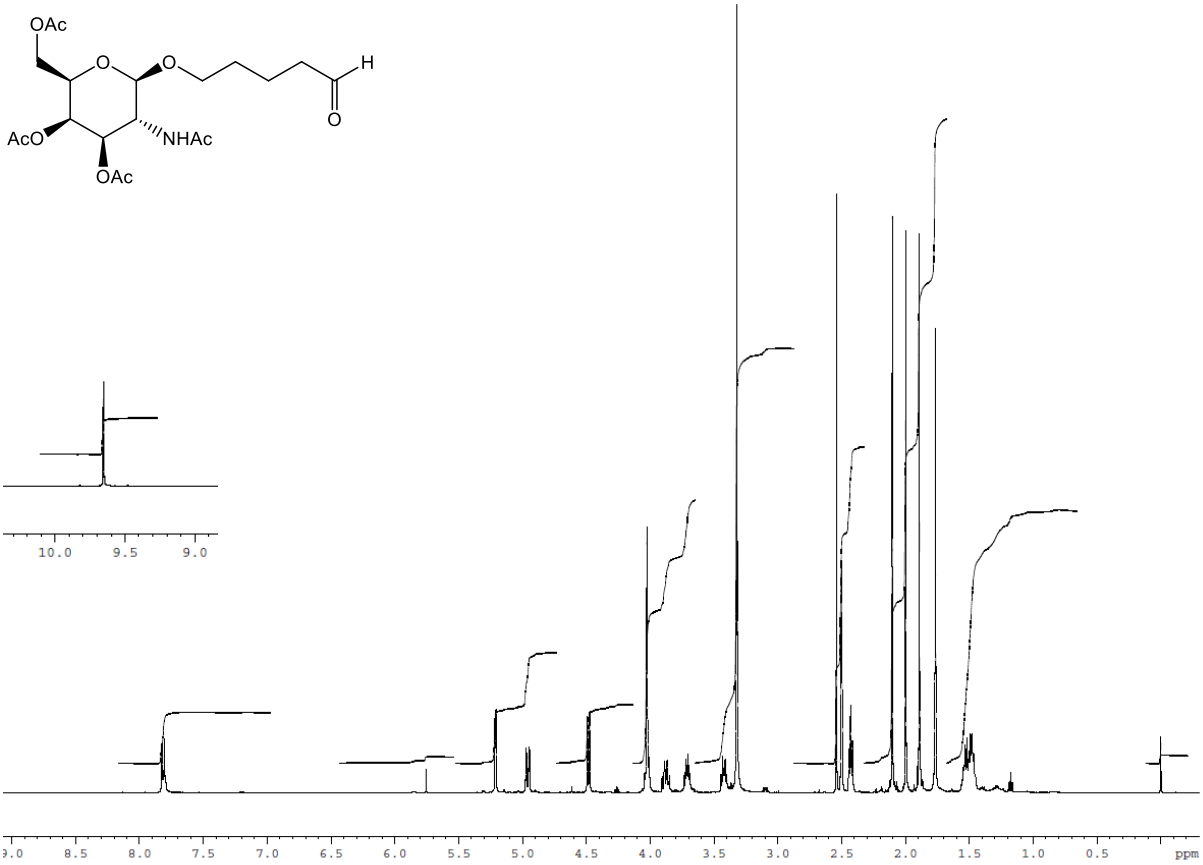


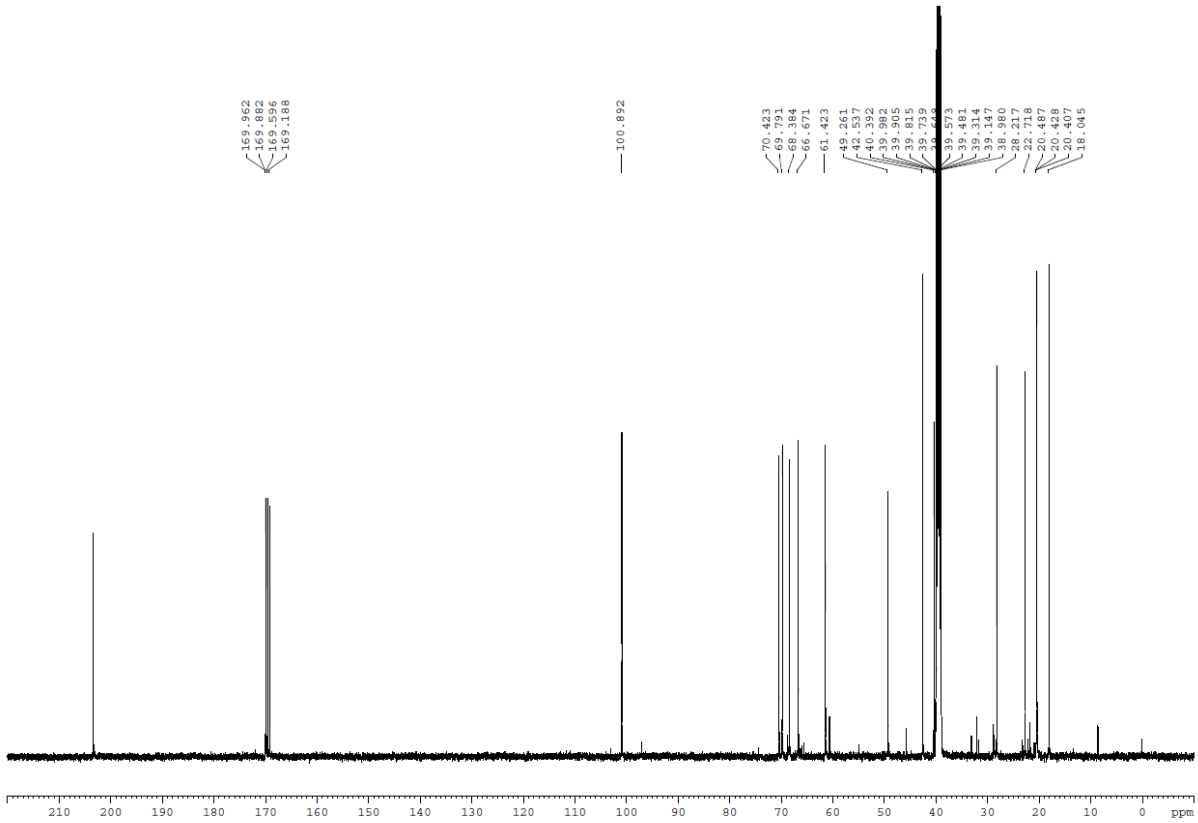
NMR-spectra **13d**



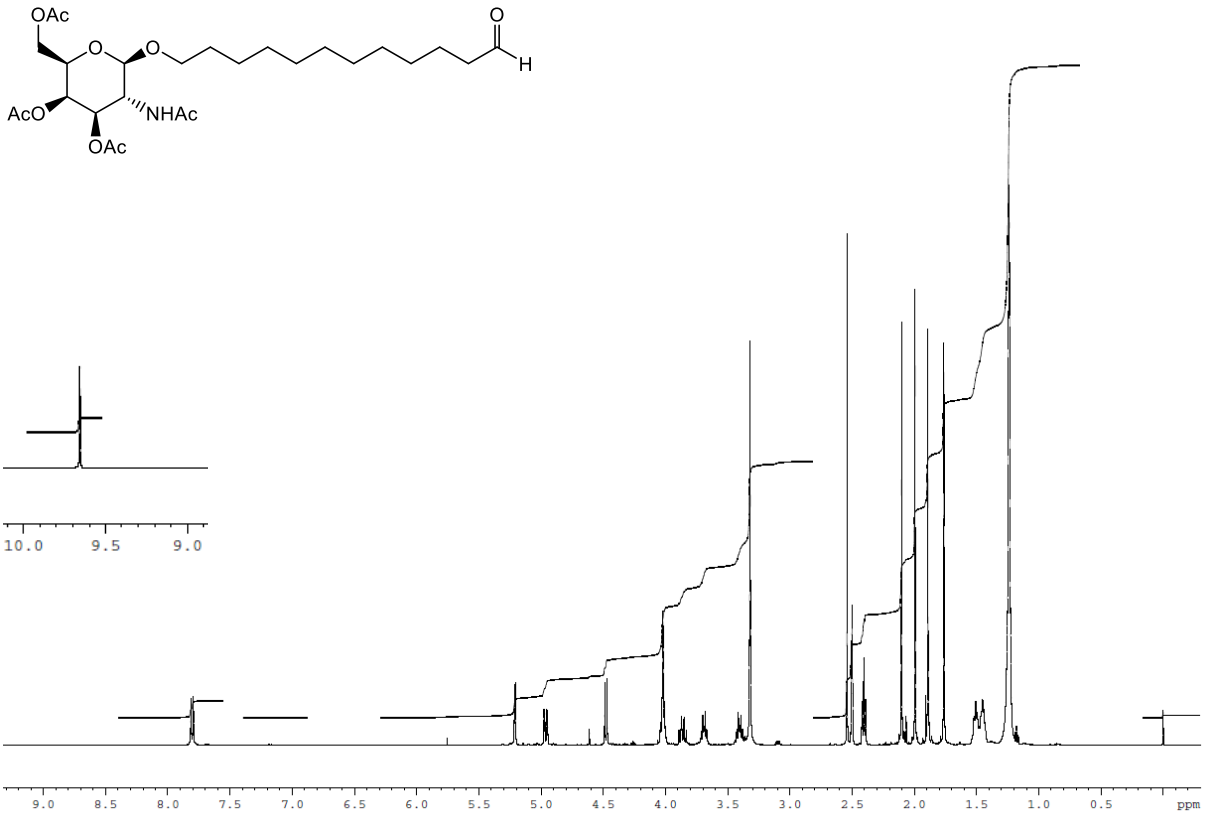


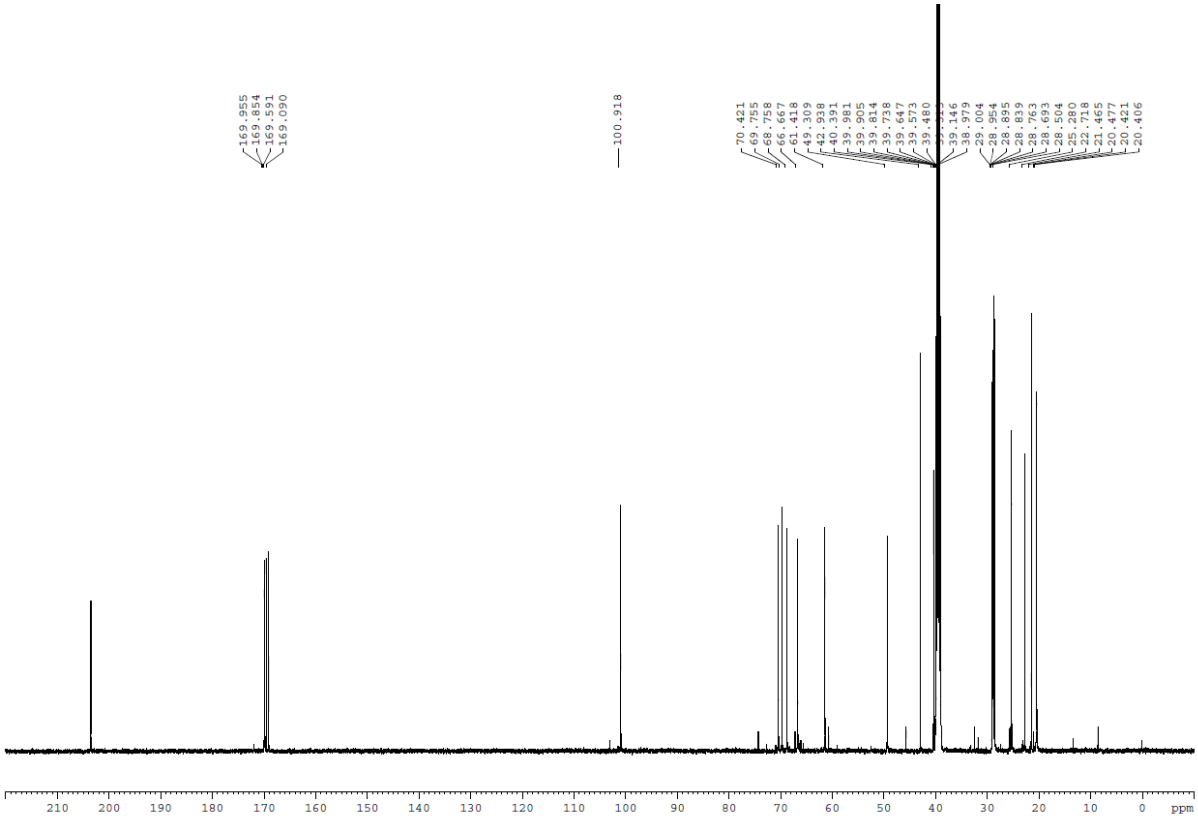
NMR-spectra **14a**



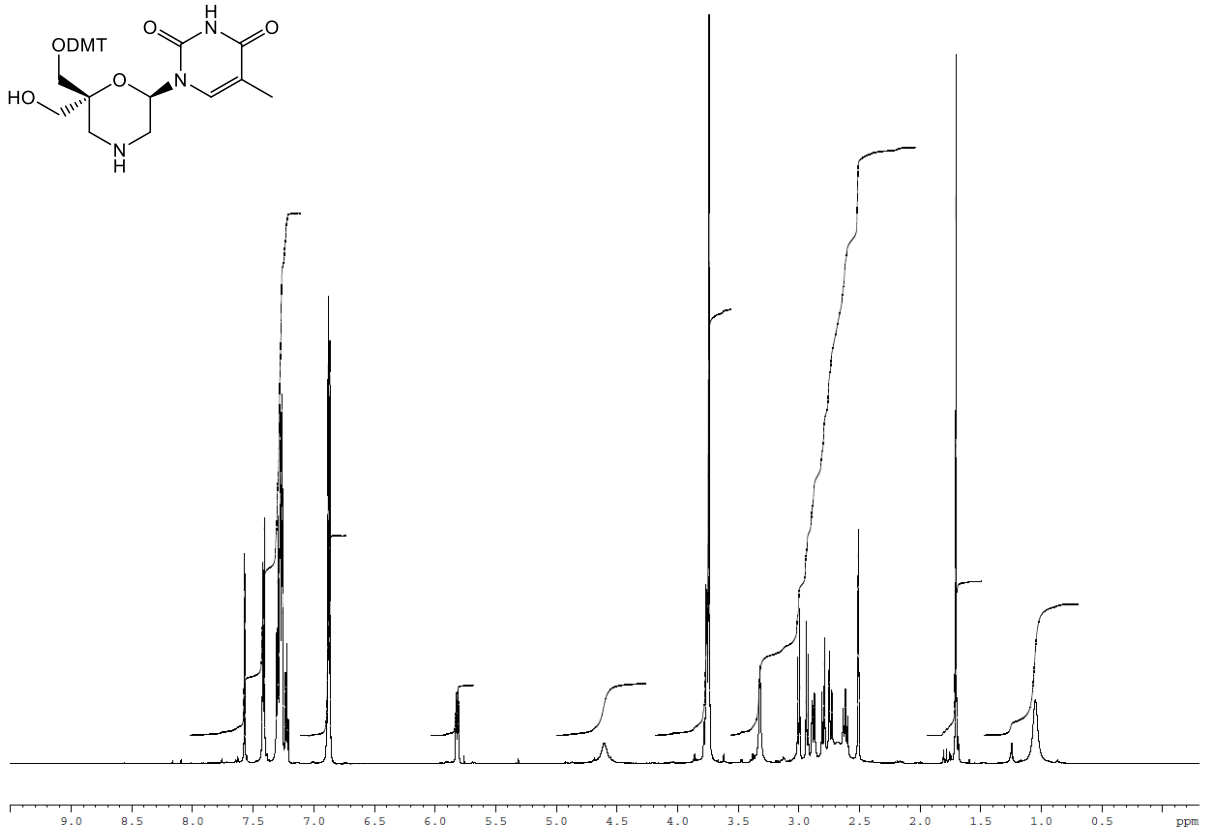


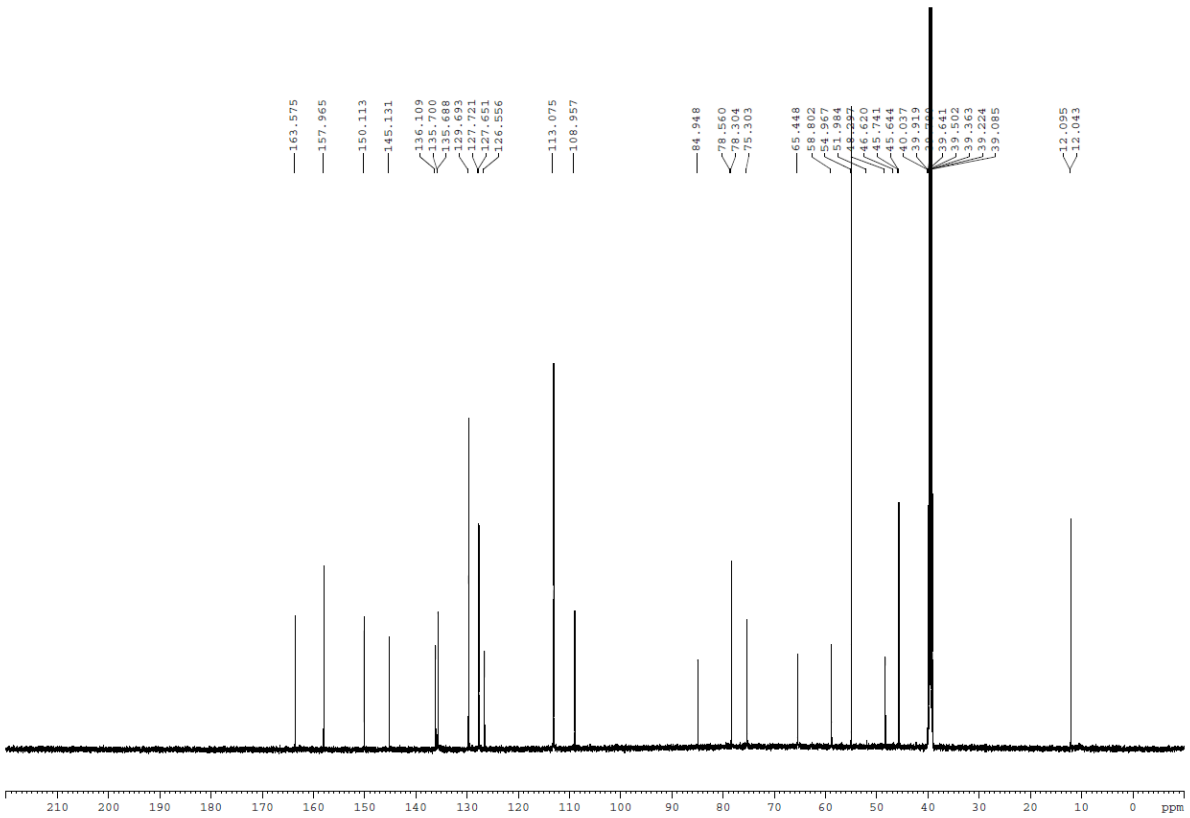
NMR-spectra **14b**



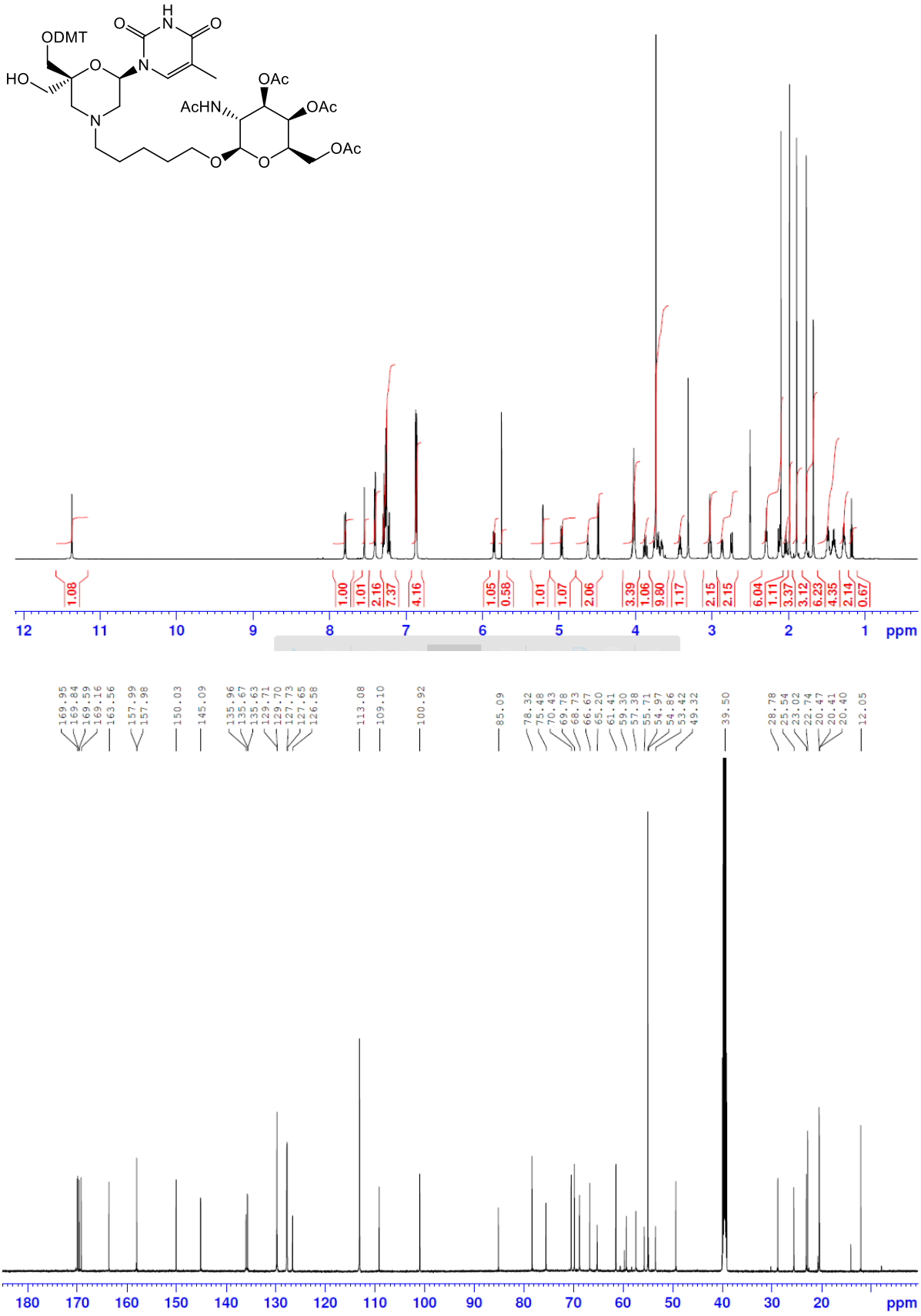


NMR-spectra **15**





NMR-spectra **16a**



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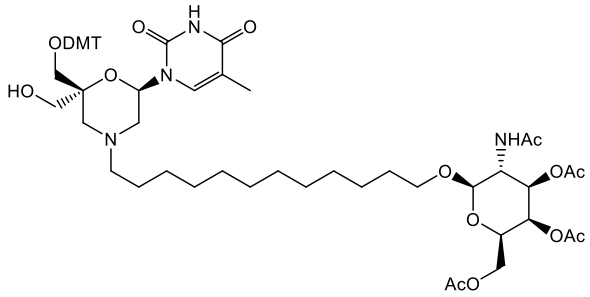
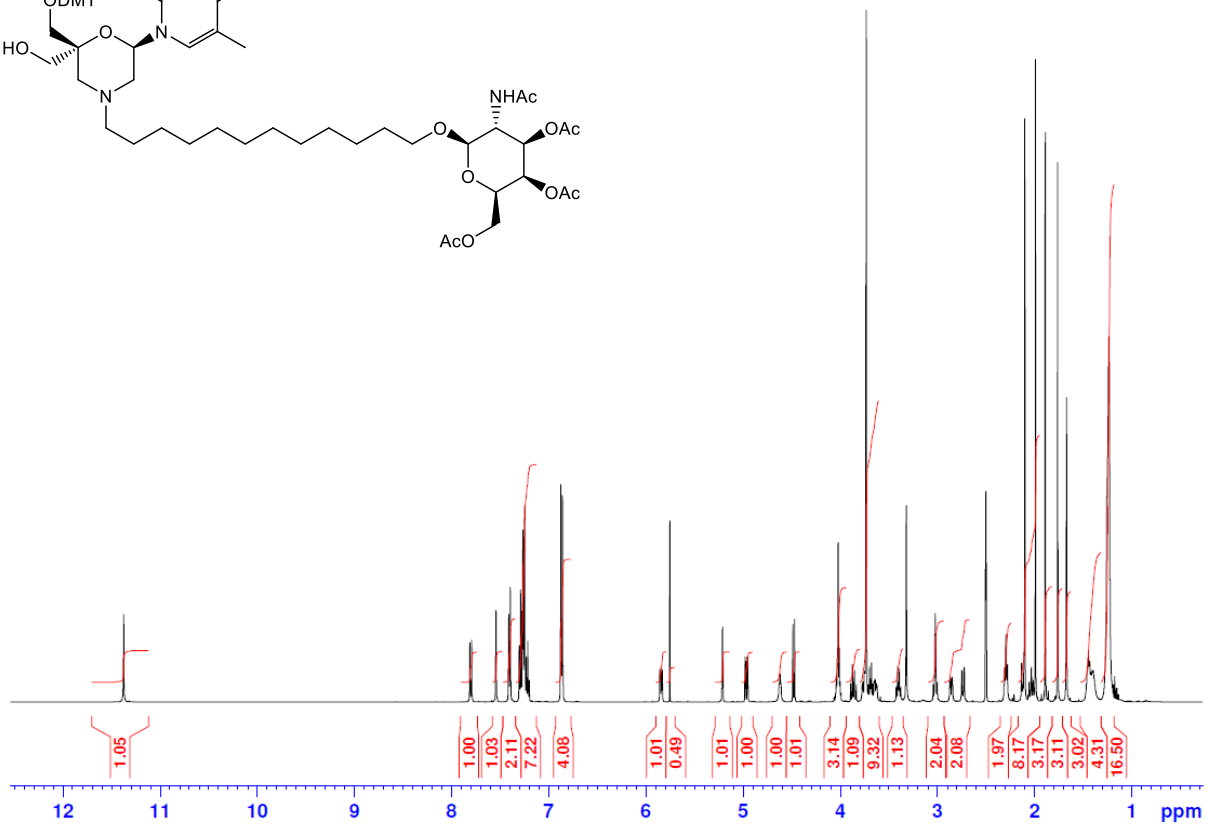
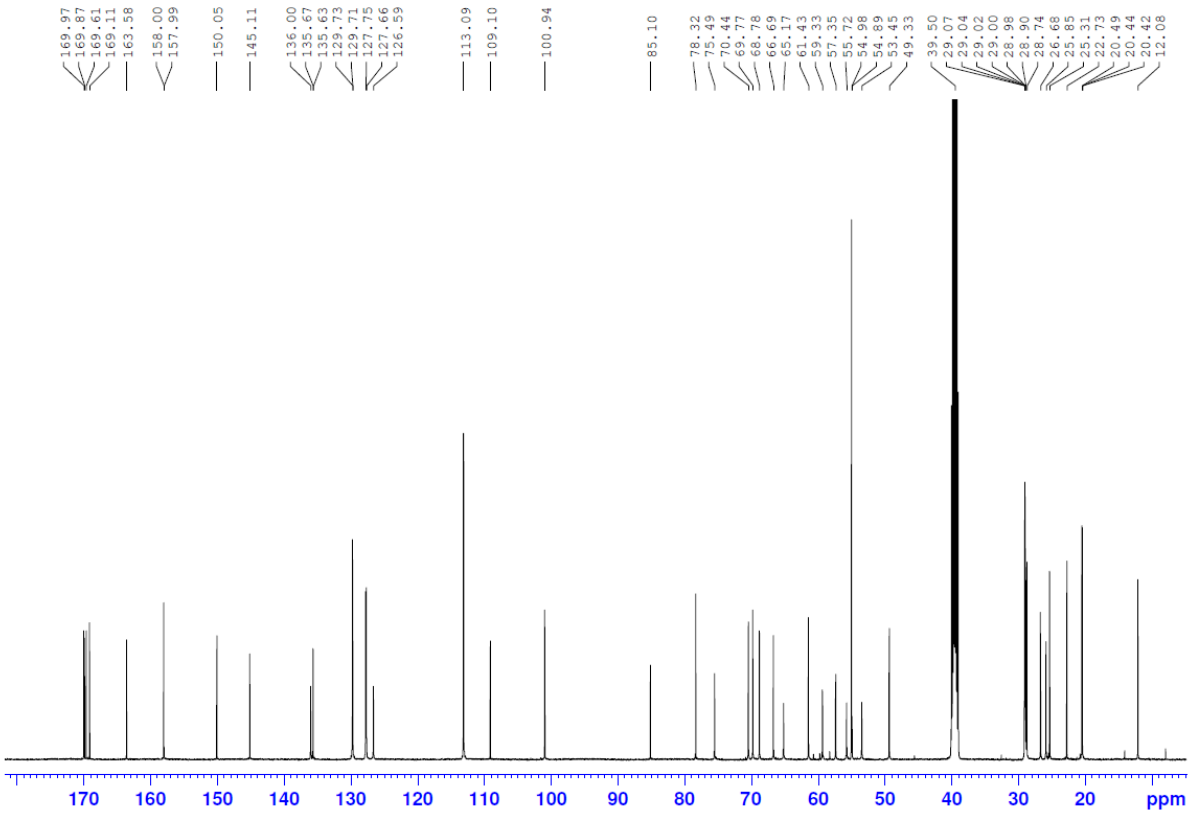
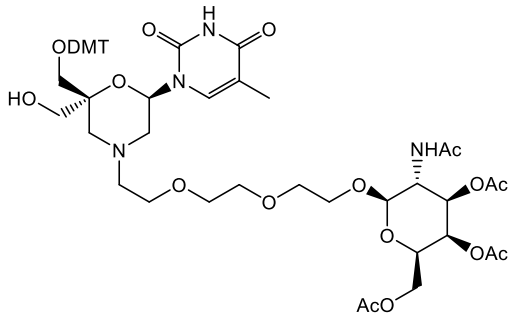
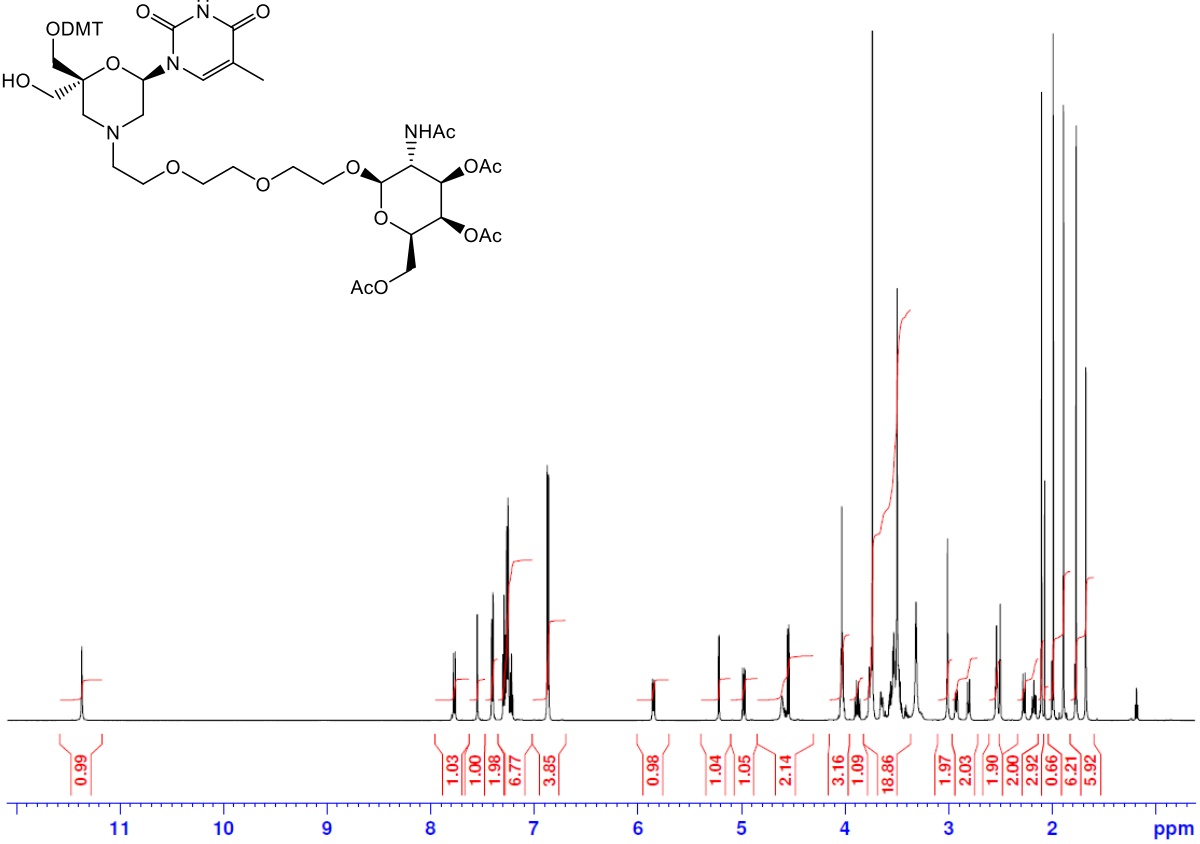


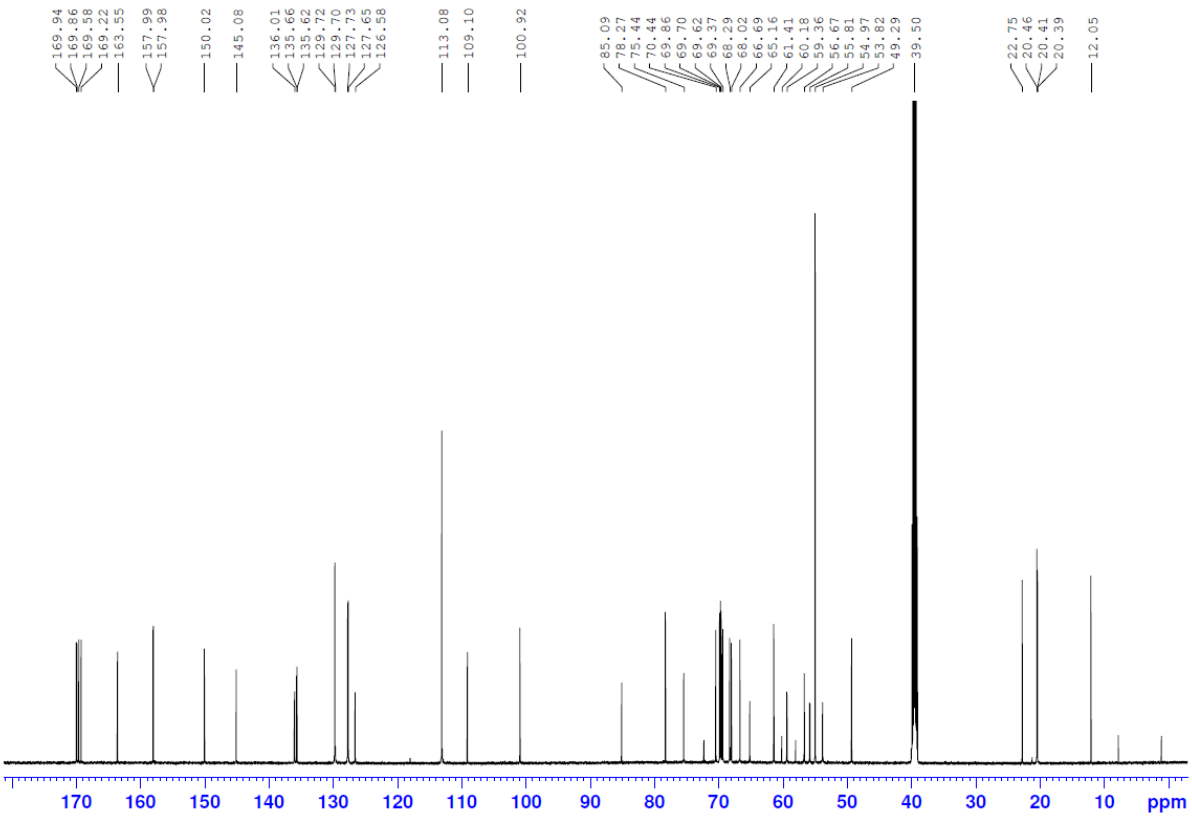




NMR-spectra **16c**







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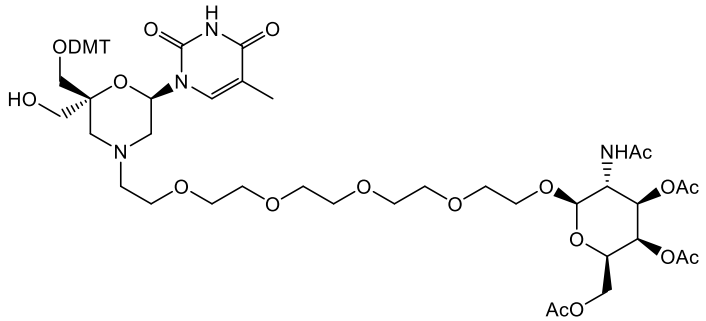
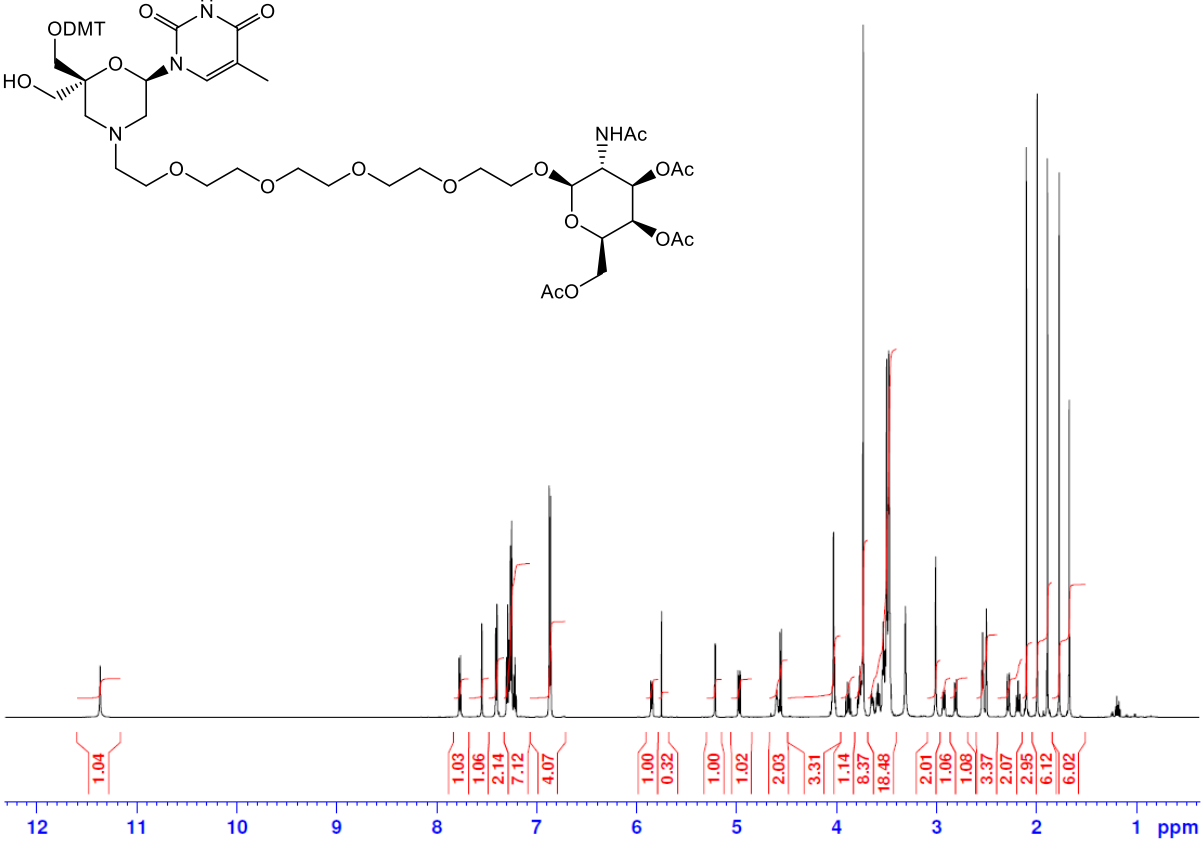
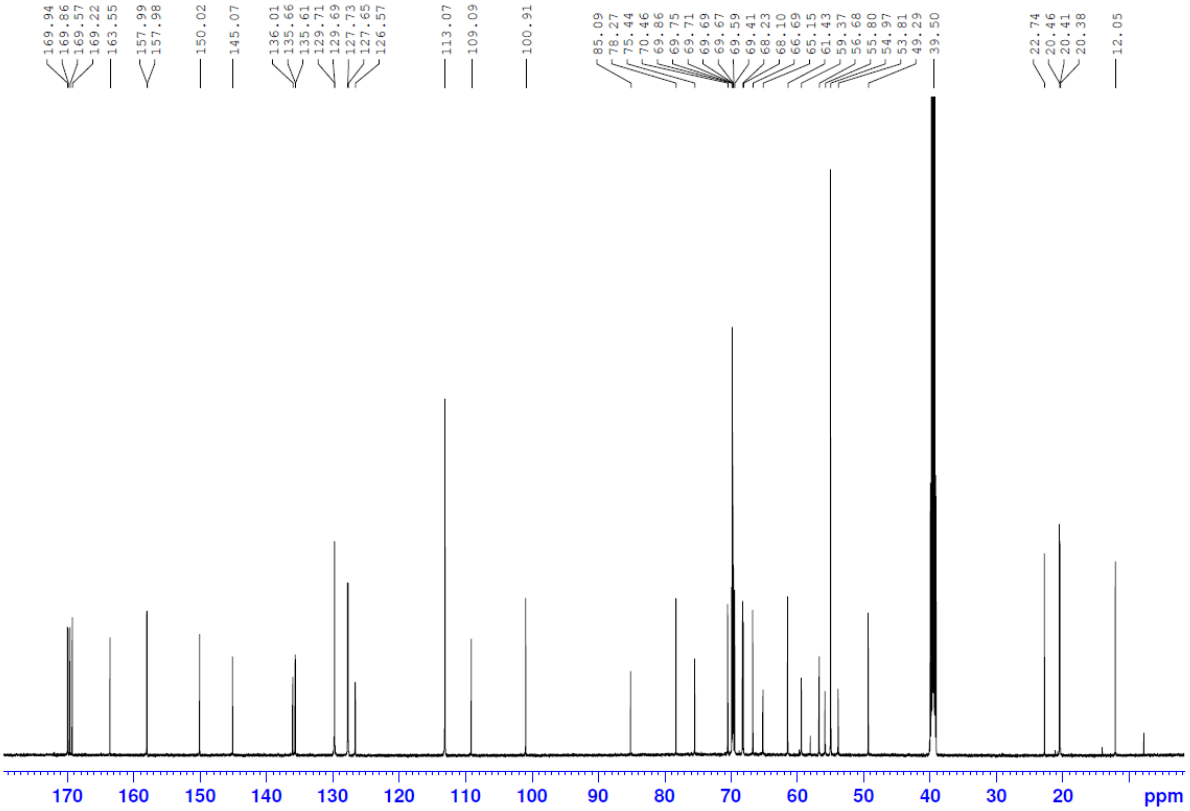
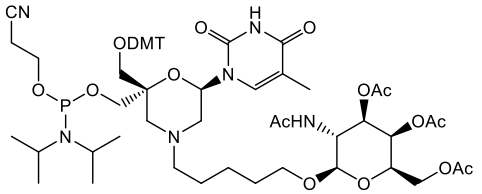
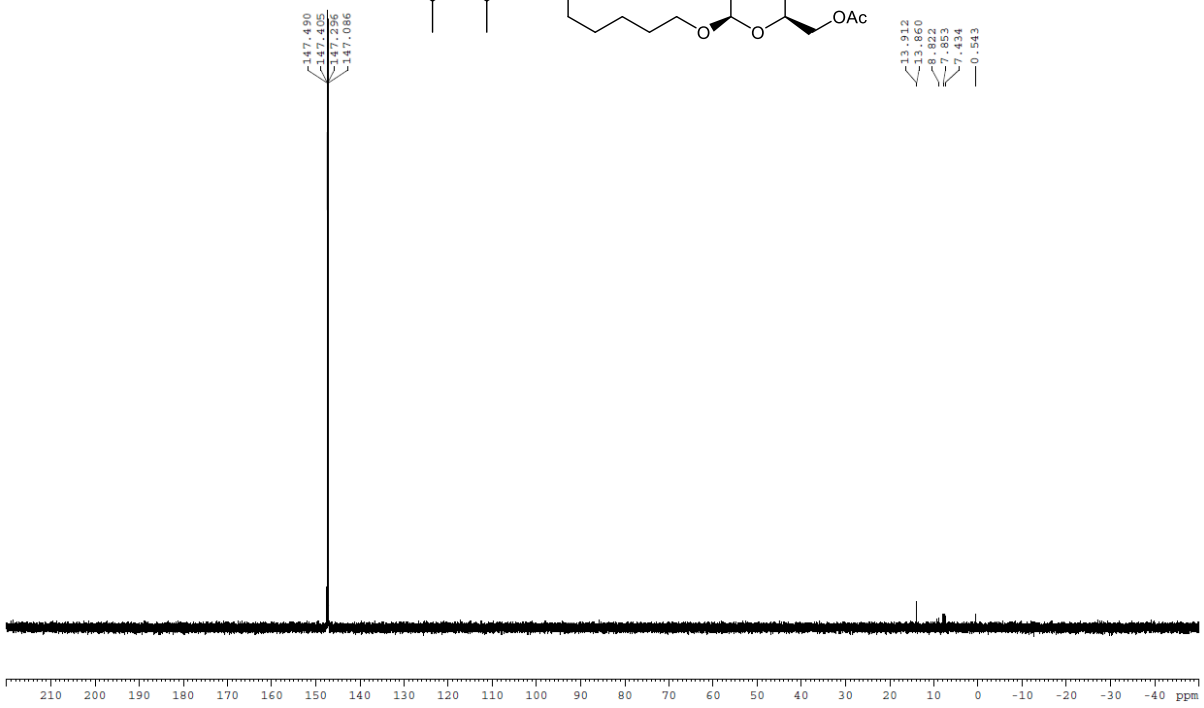


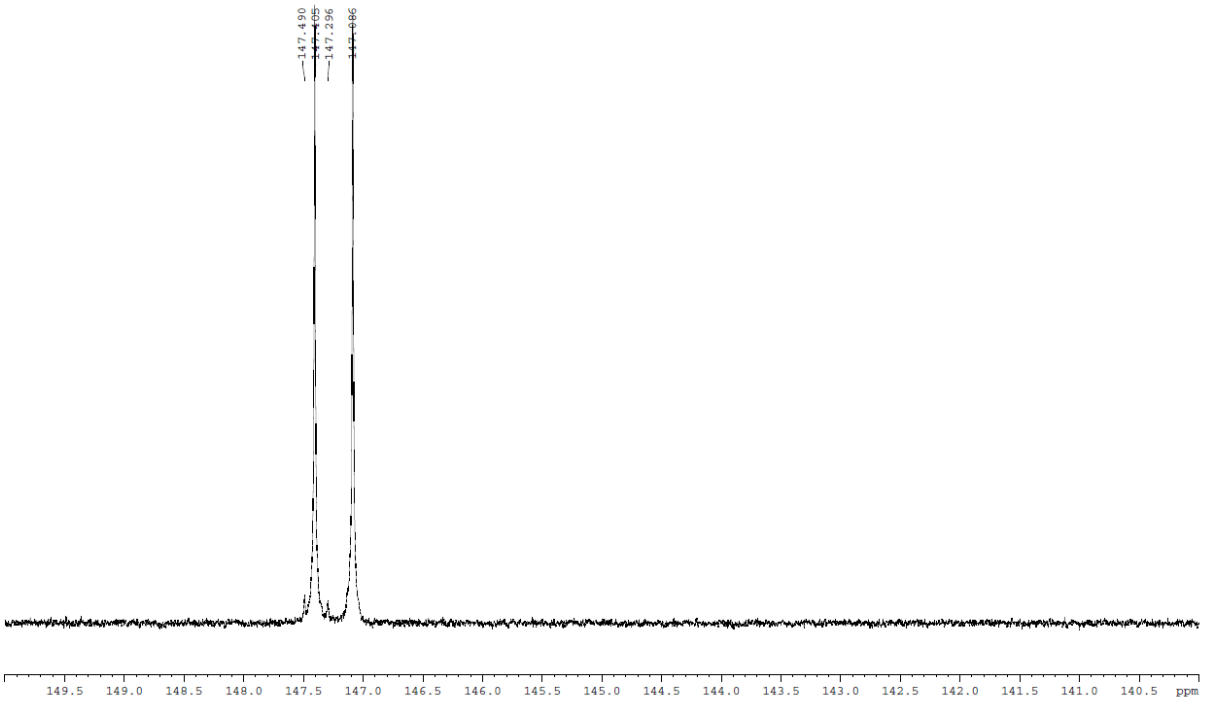




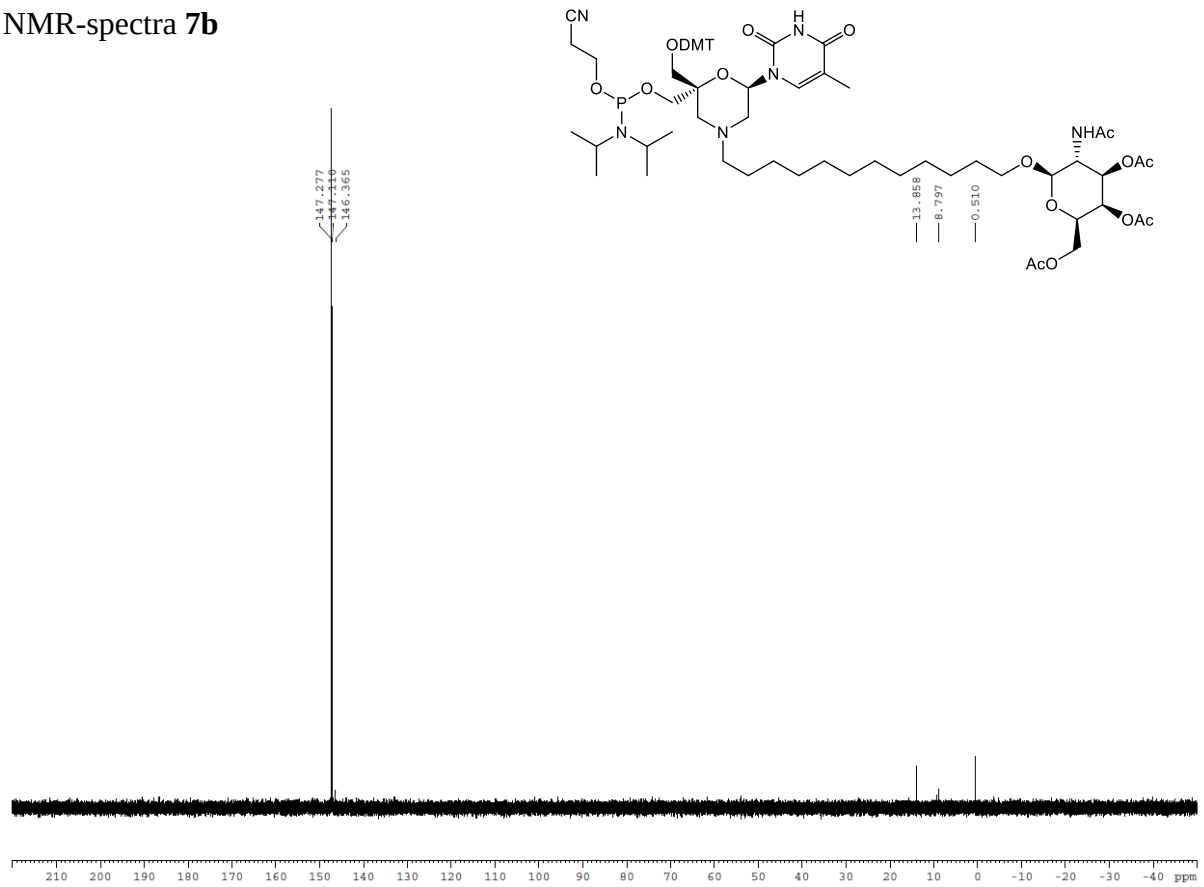
NMR-spectra **7a**

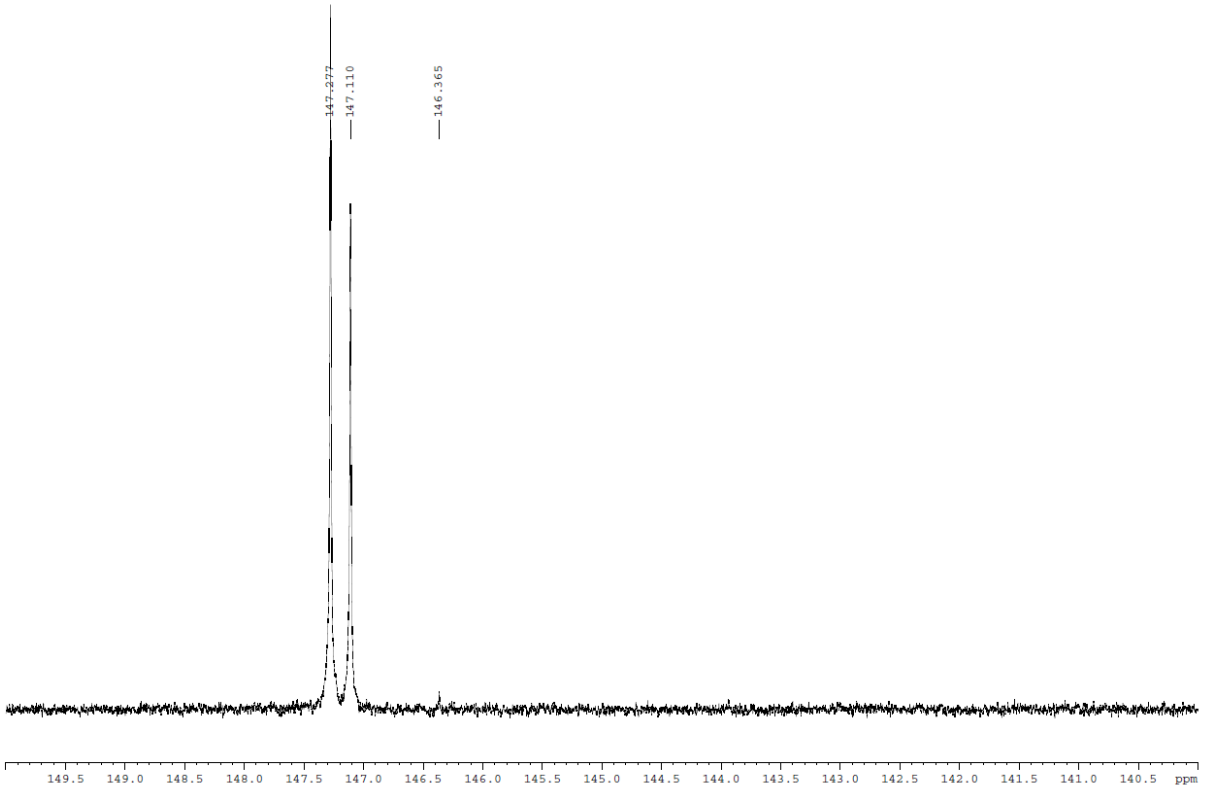




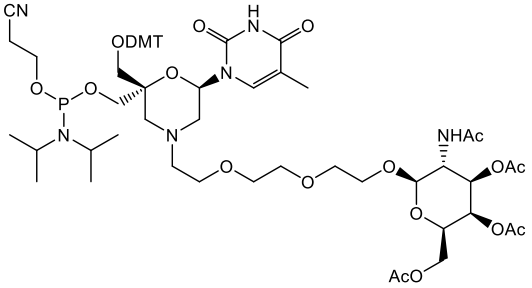


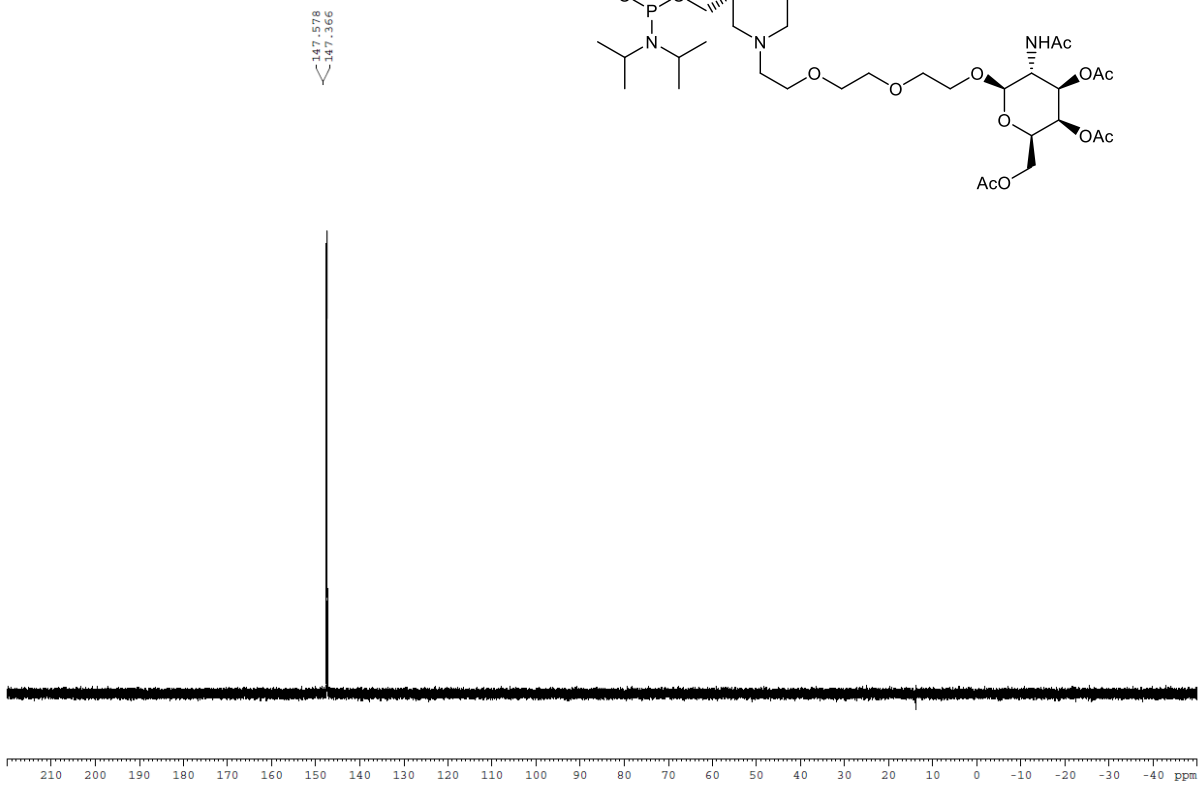
NMR-spectra **7b**

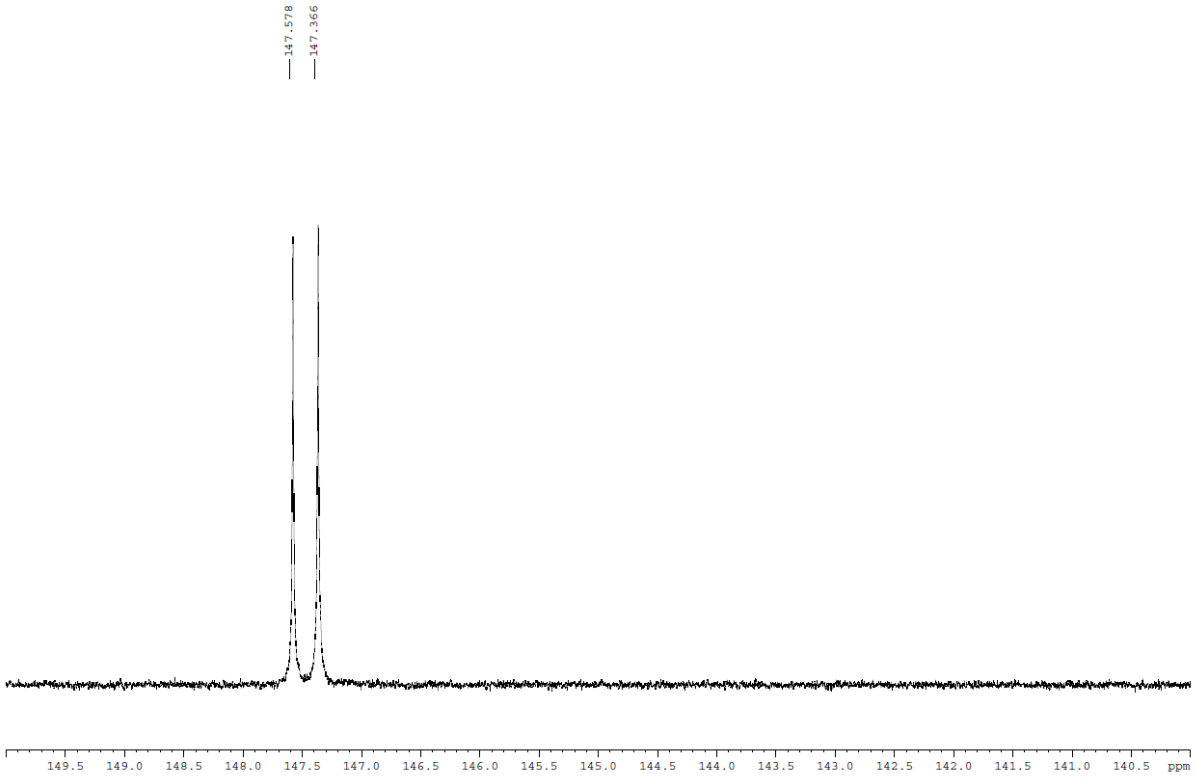




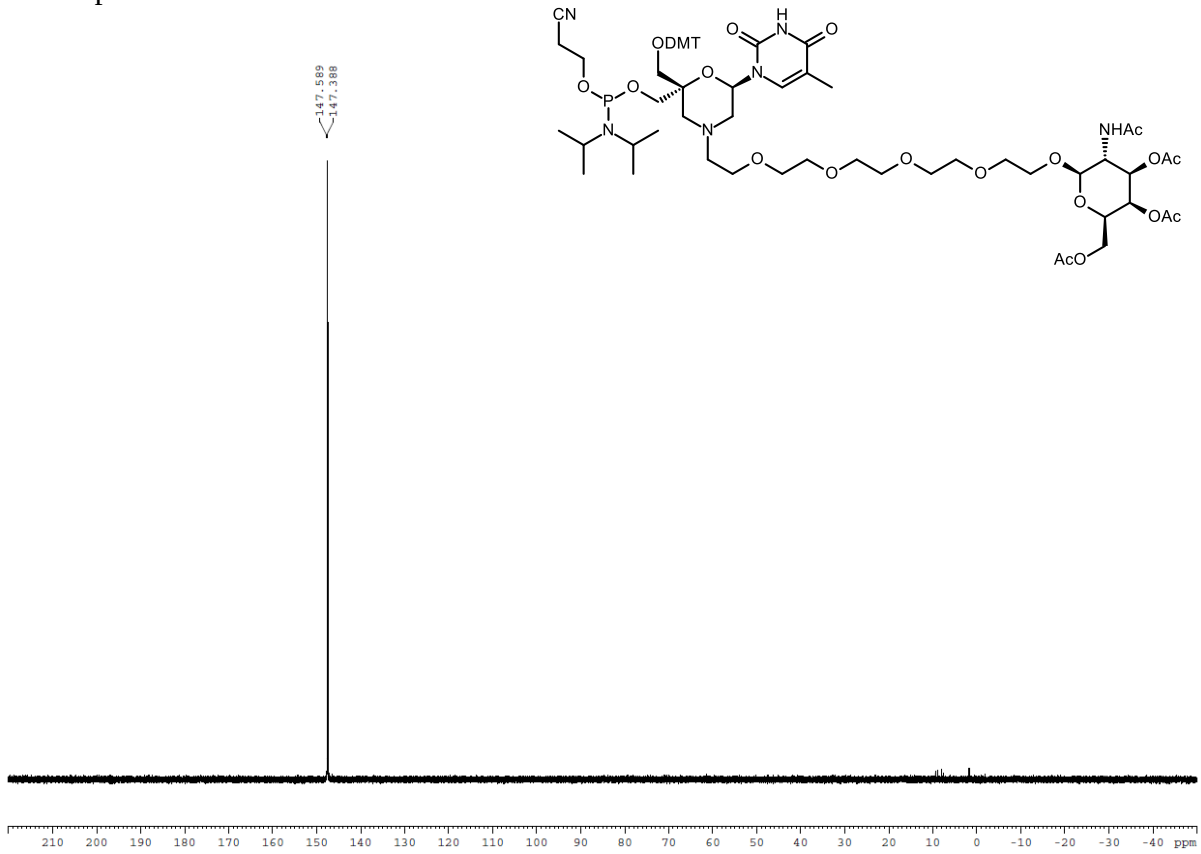
NMR-spectra **7c**

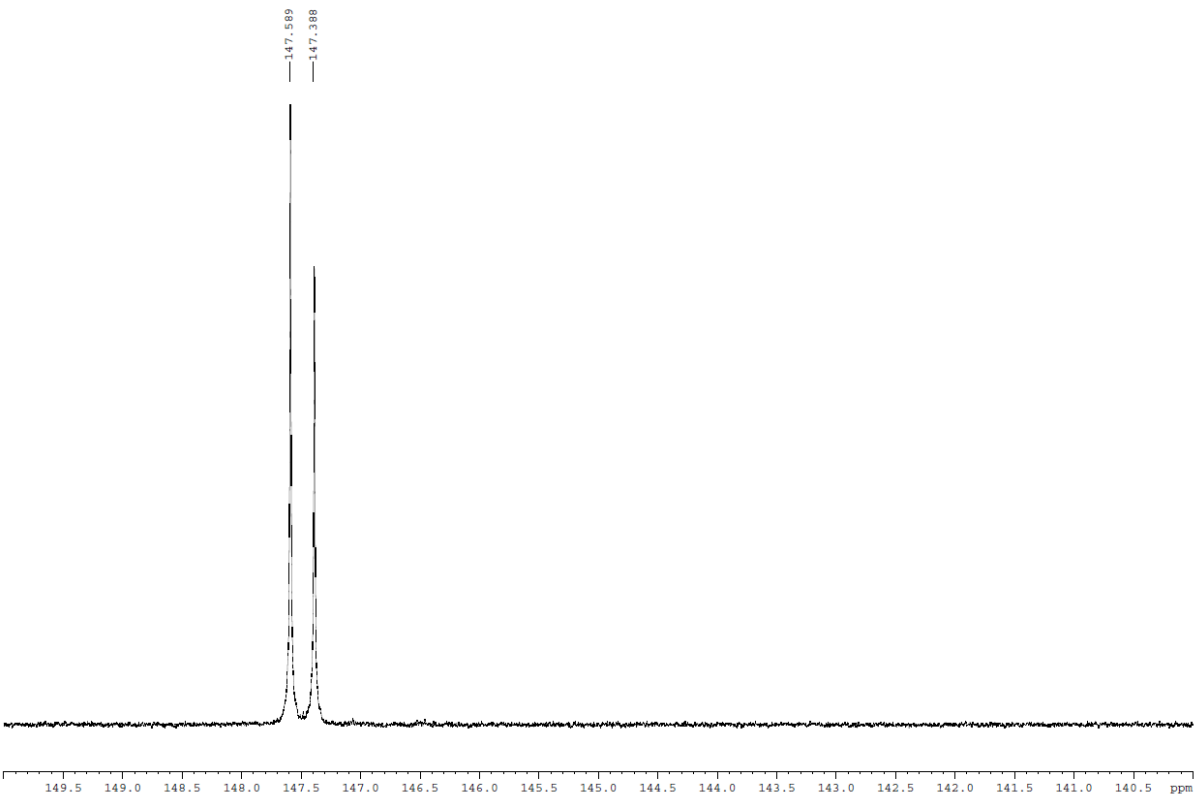




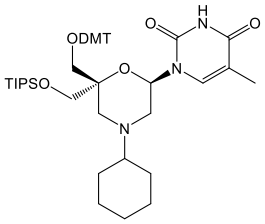


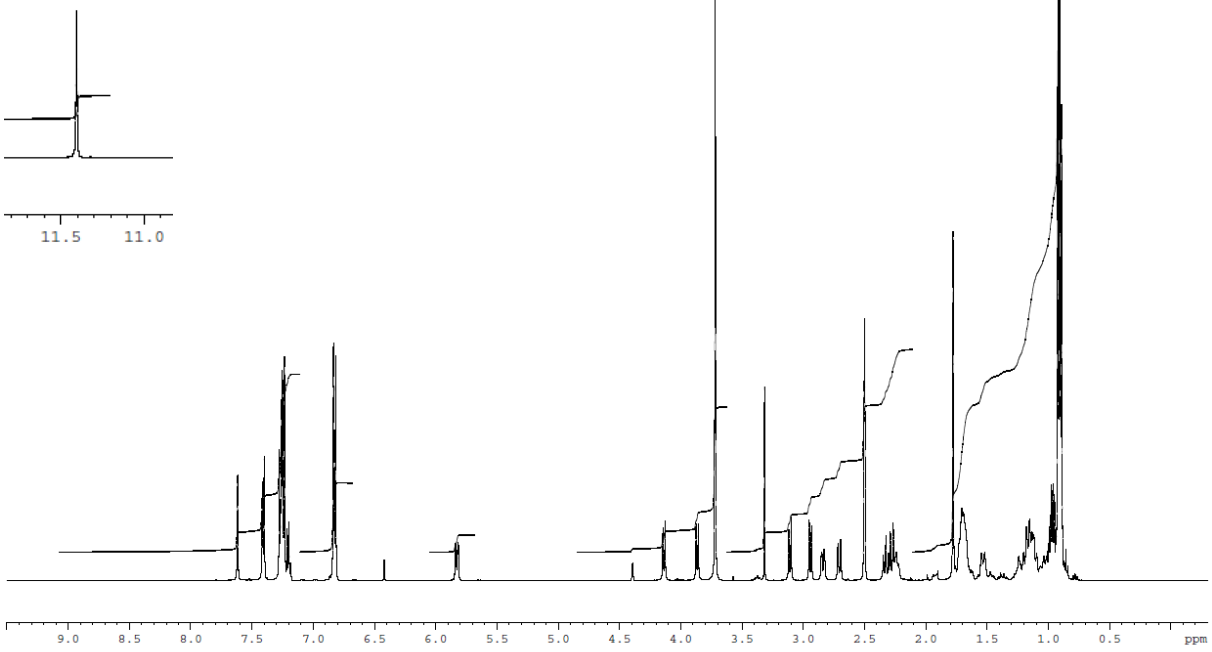
NMR-spectra **7d**

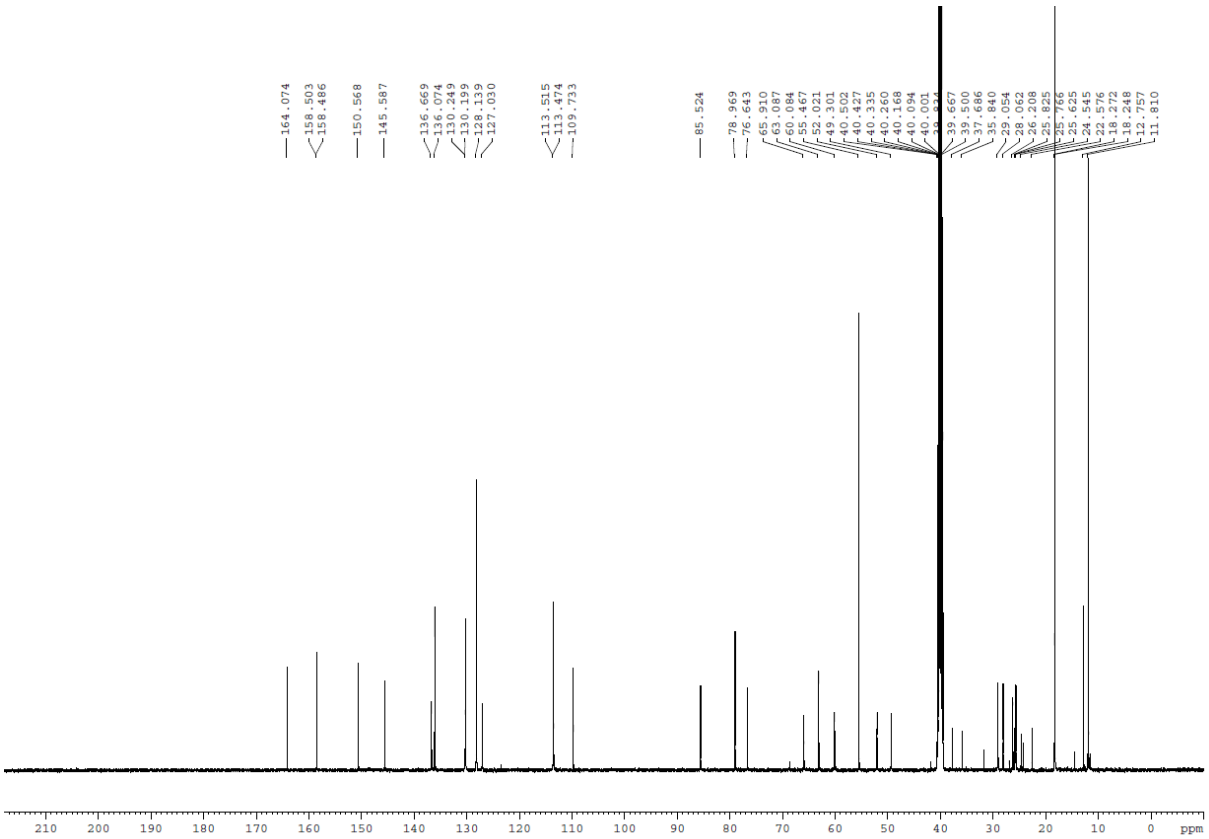




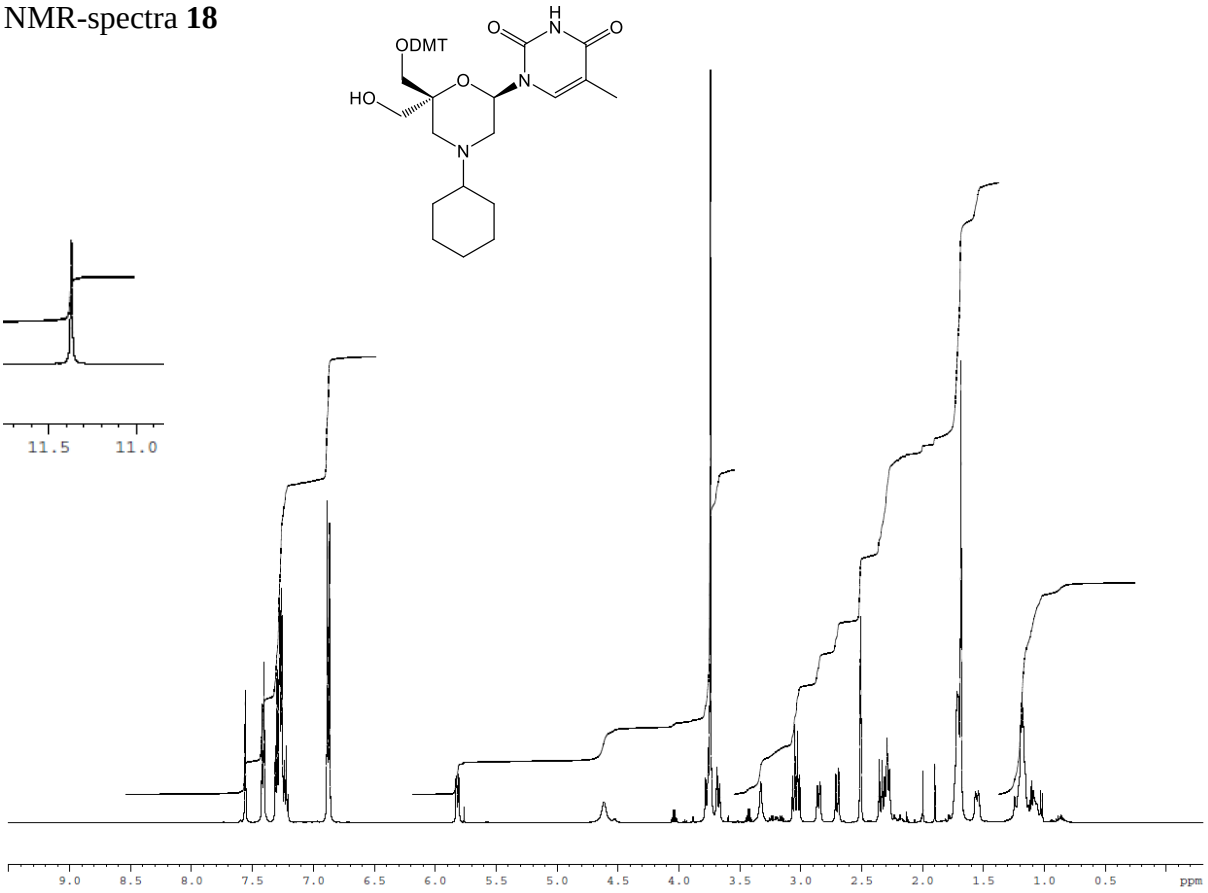
NMR-spectra **17**

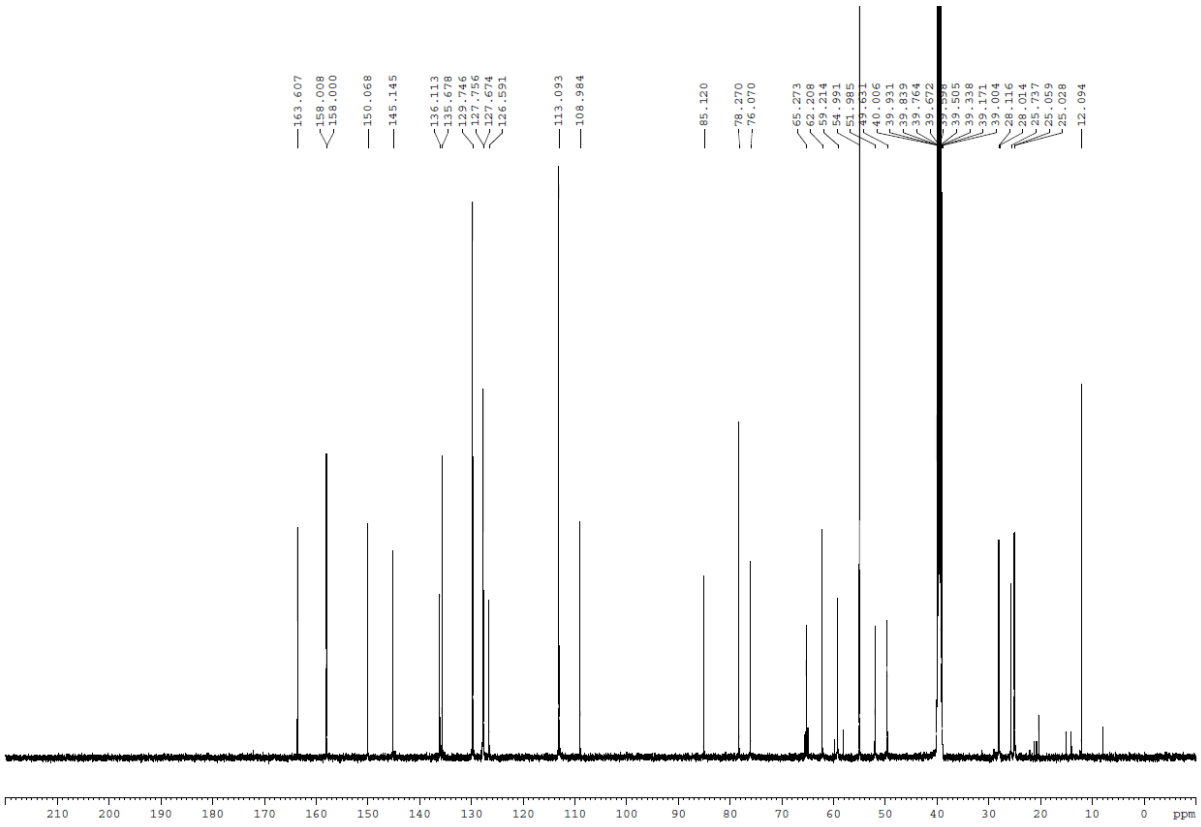




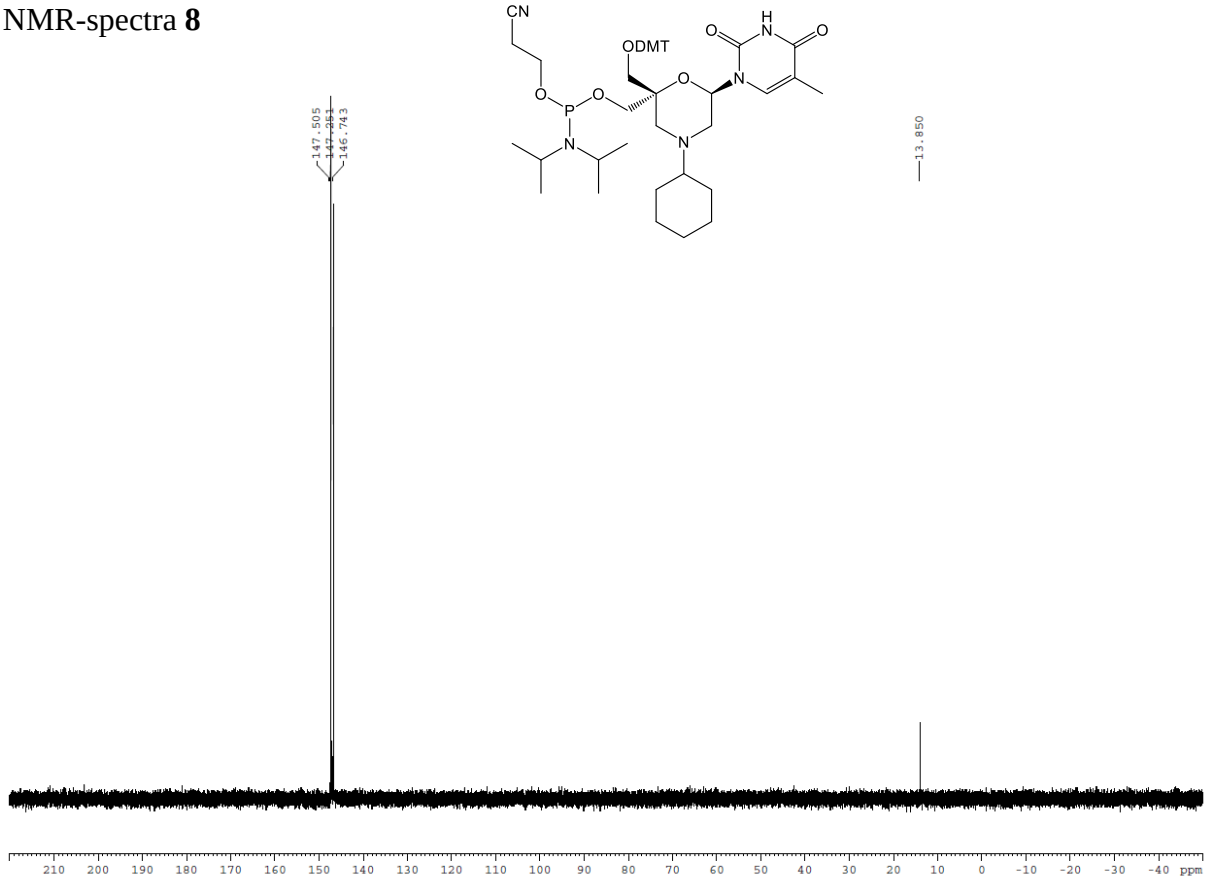


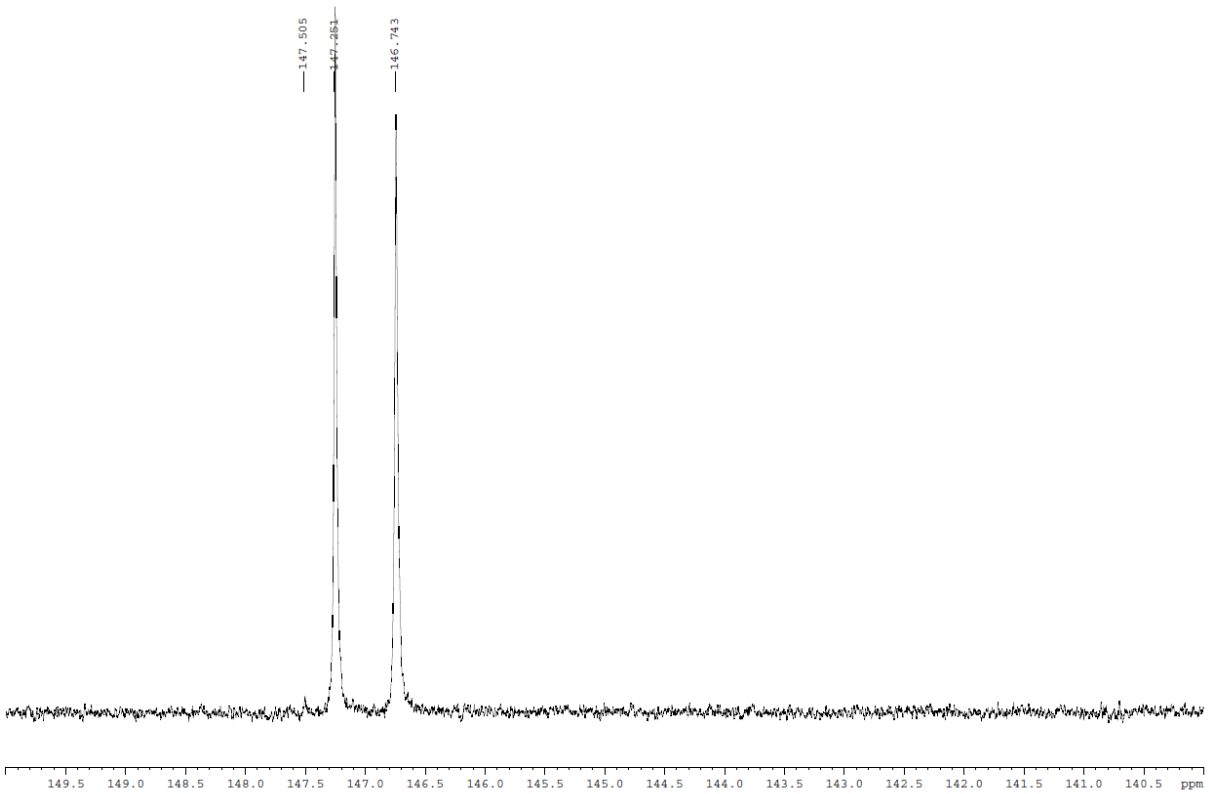
NMR-spectra **18**



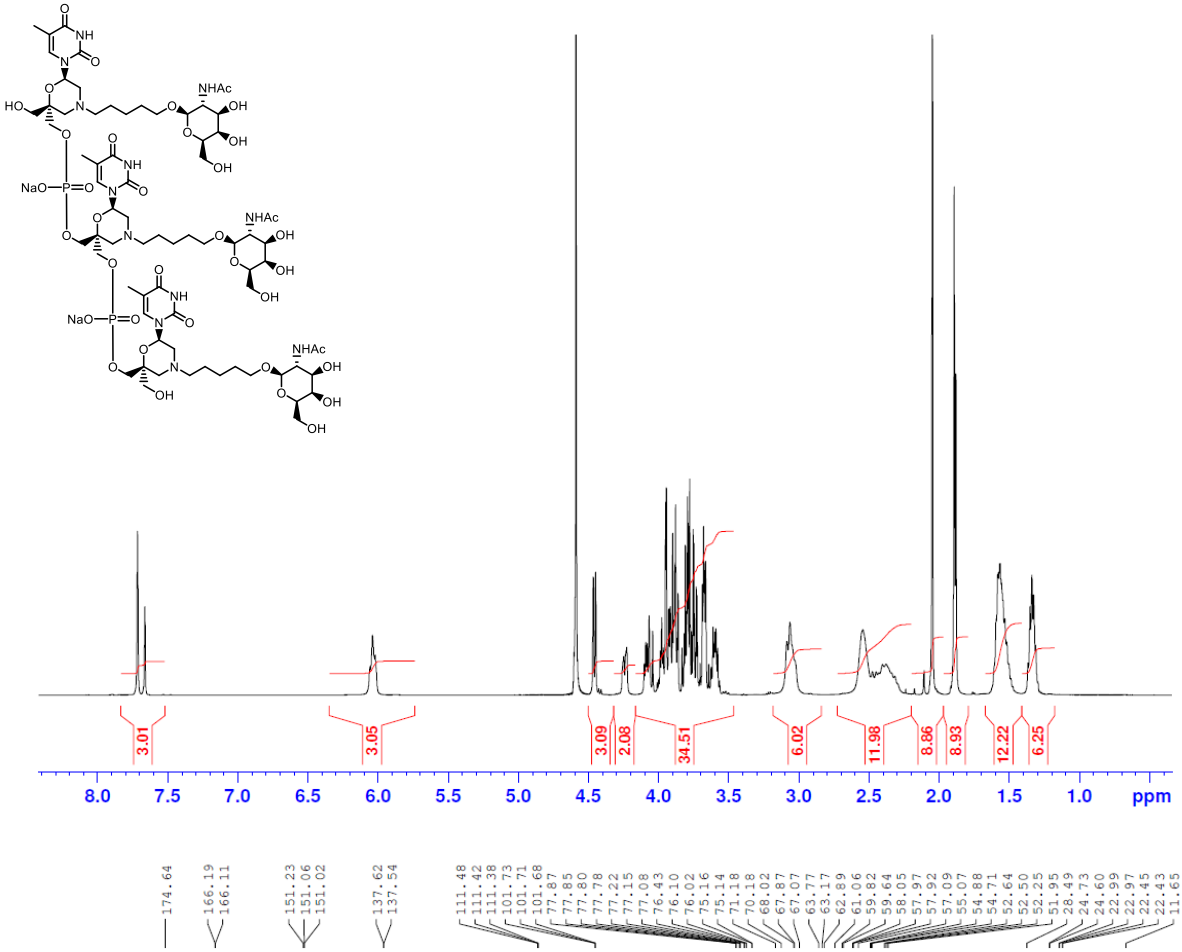


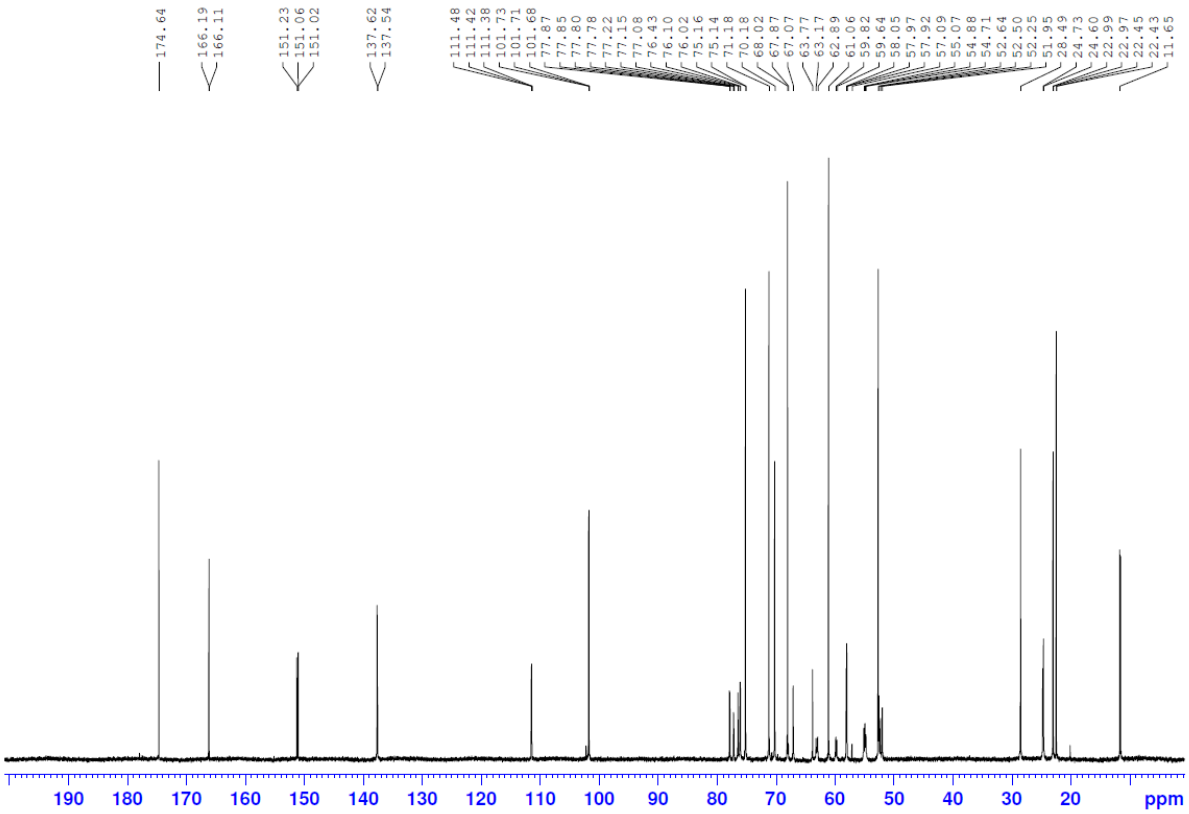
NMR-spectra **8**



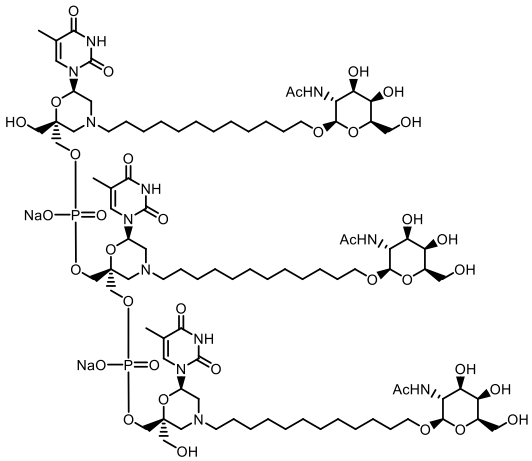


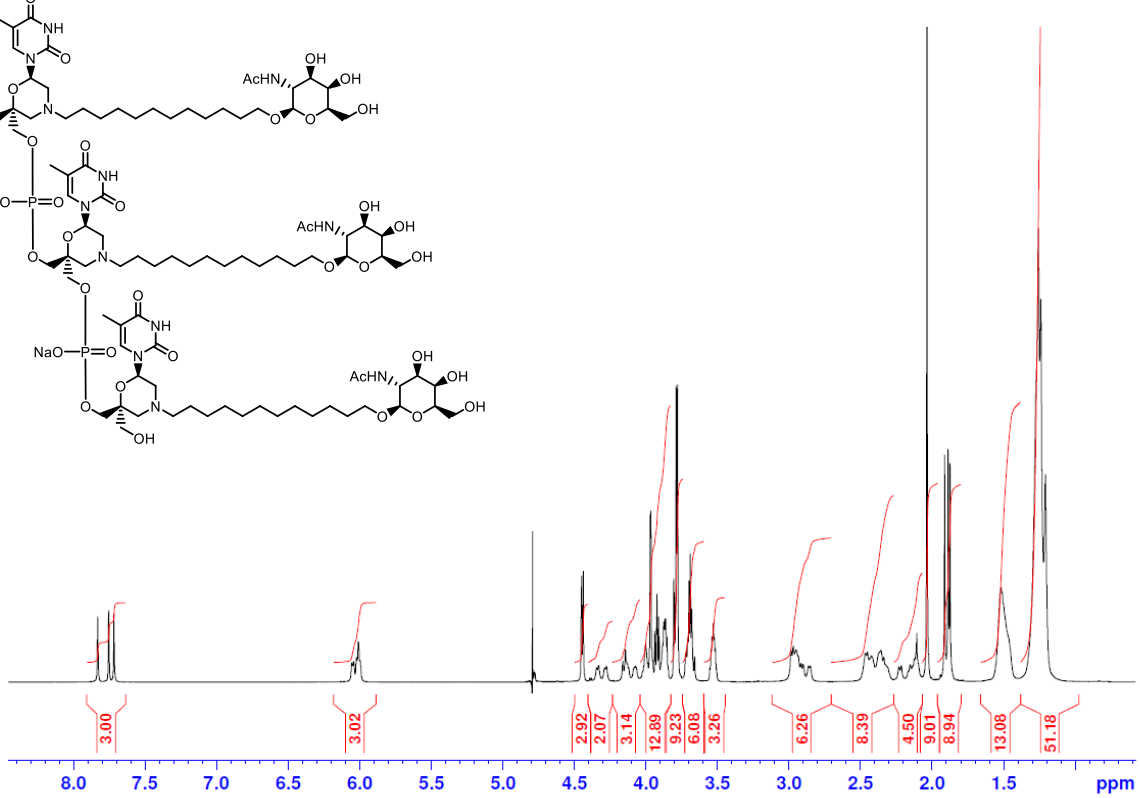
NMR-spectra **19a**

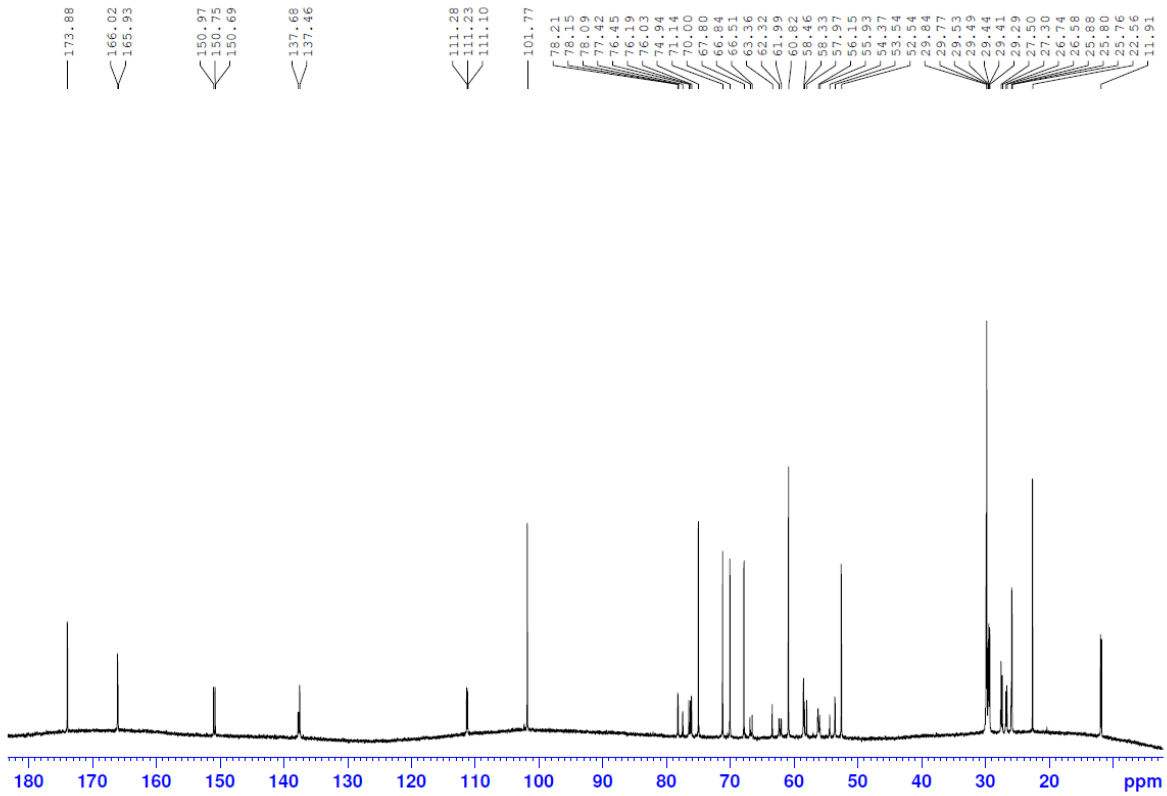




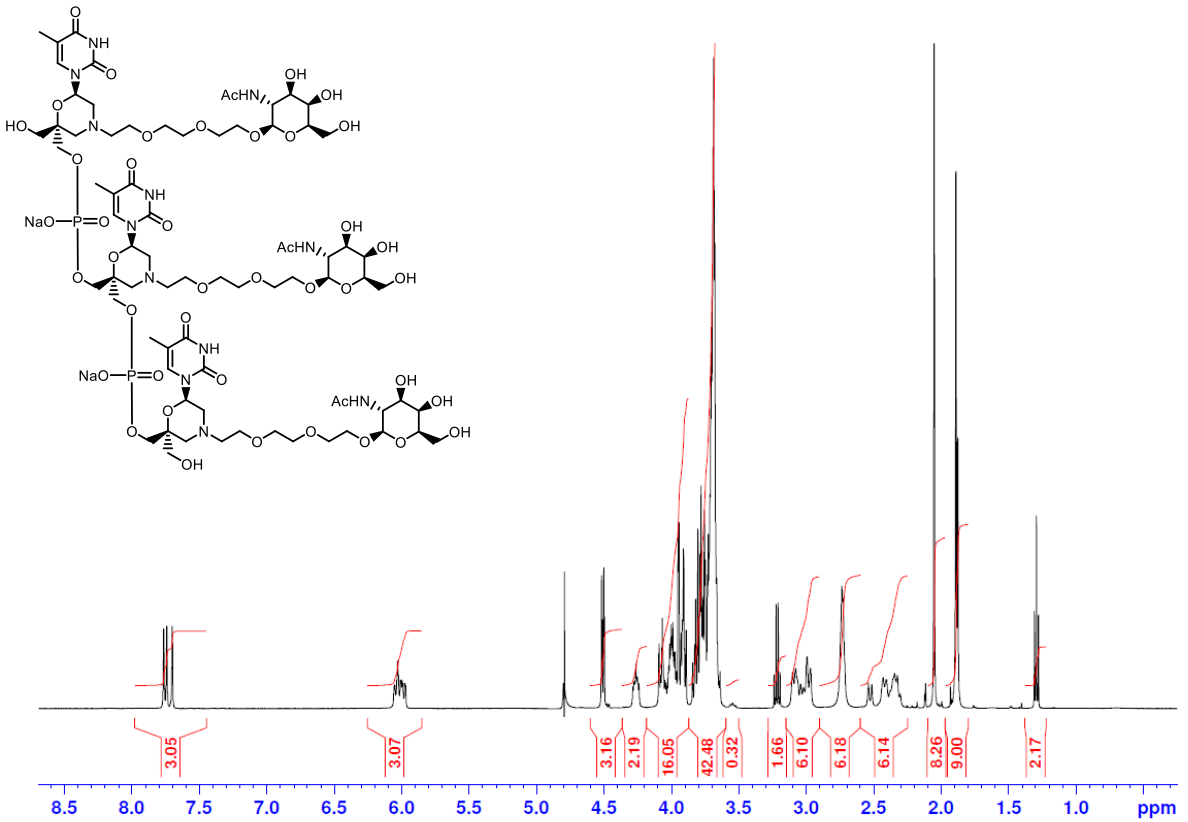
NMR-spectra **19b**

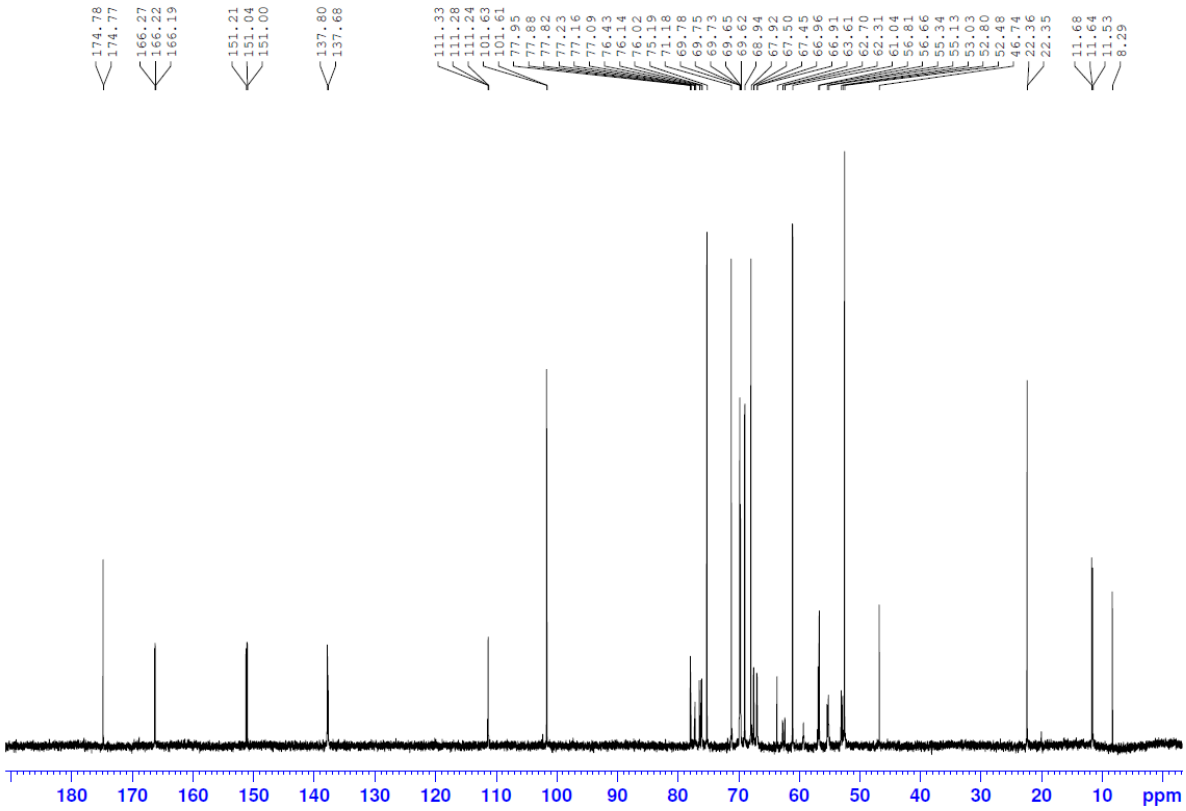




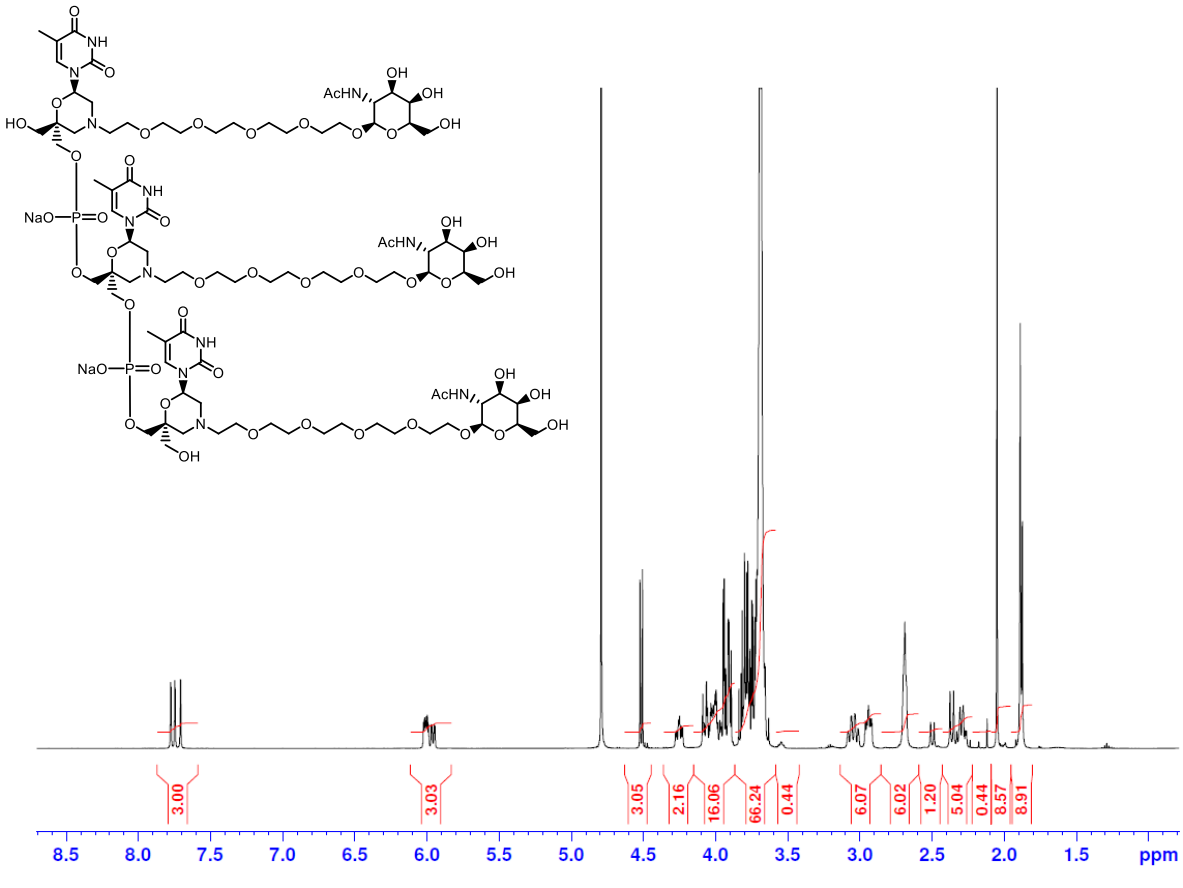


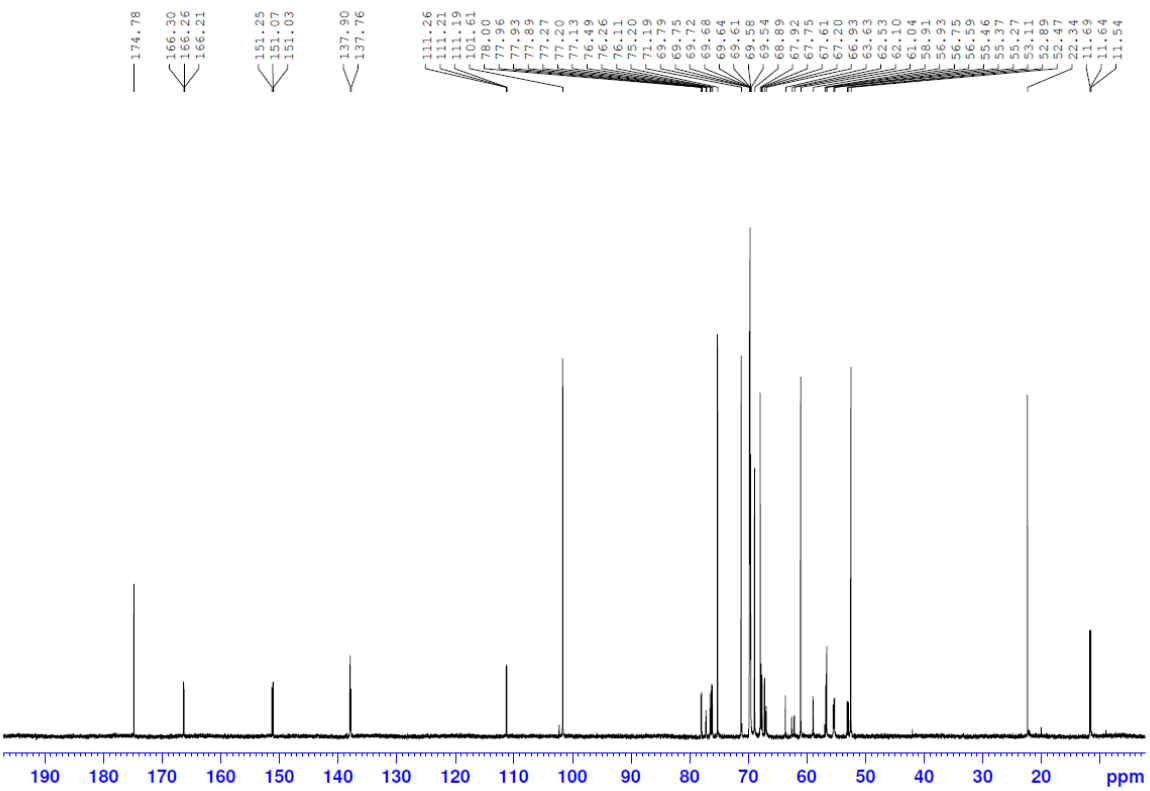
NMR-spectra **19c**





NMR-spectra **19d**

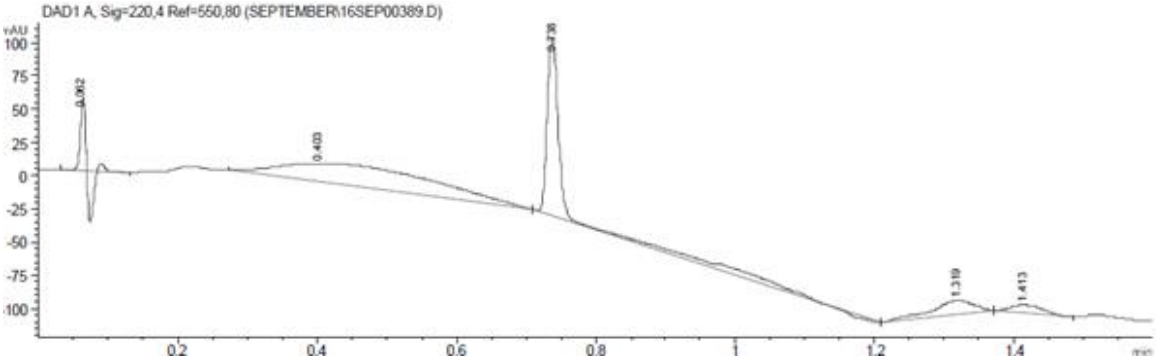




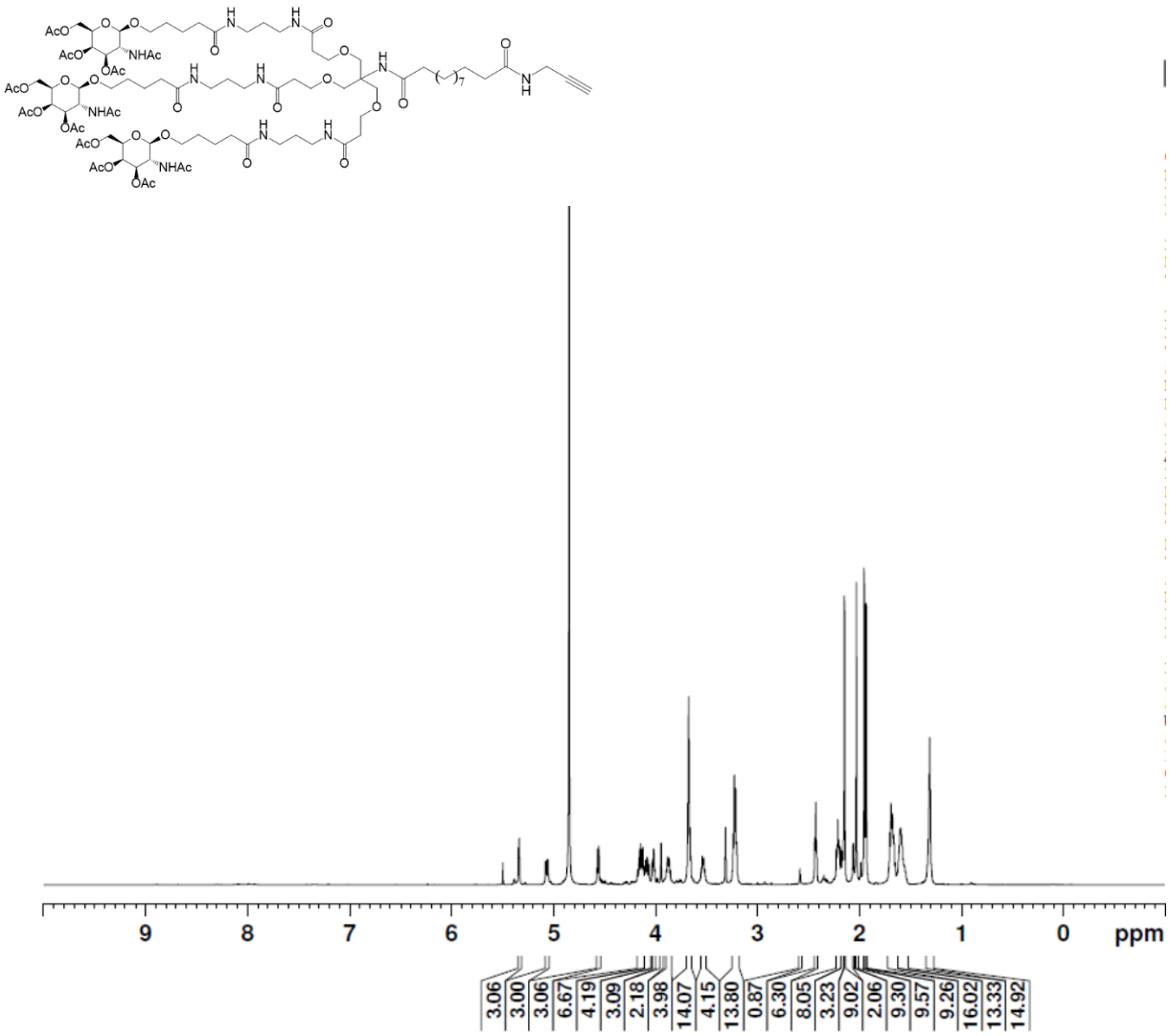
LCMS-UV (220 nm) spectrum of compound **21**

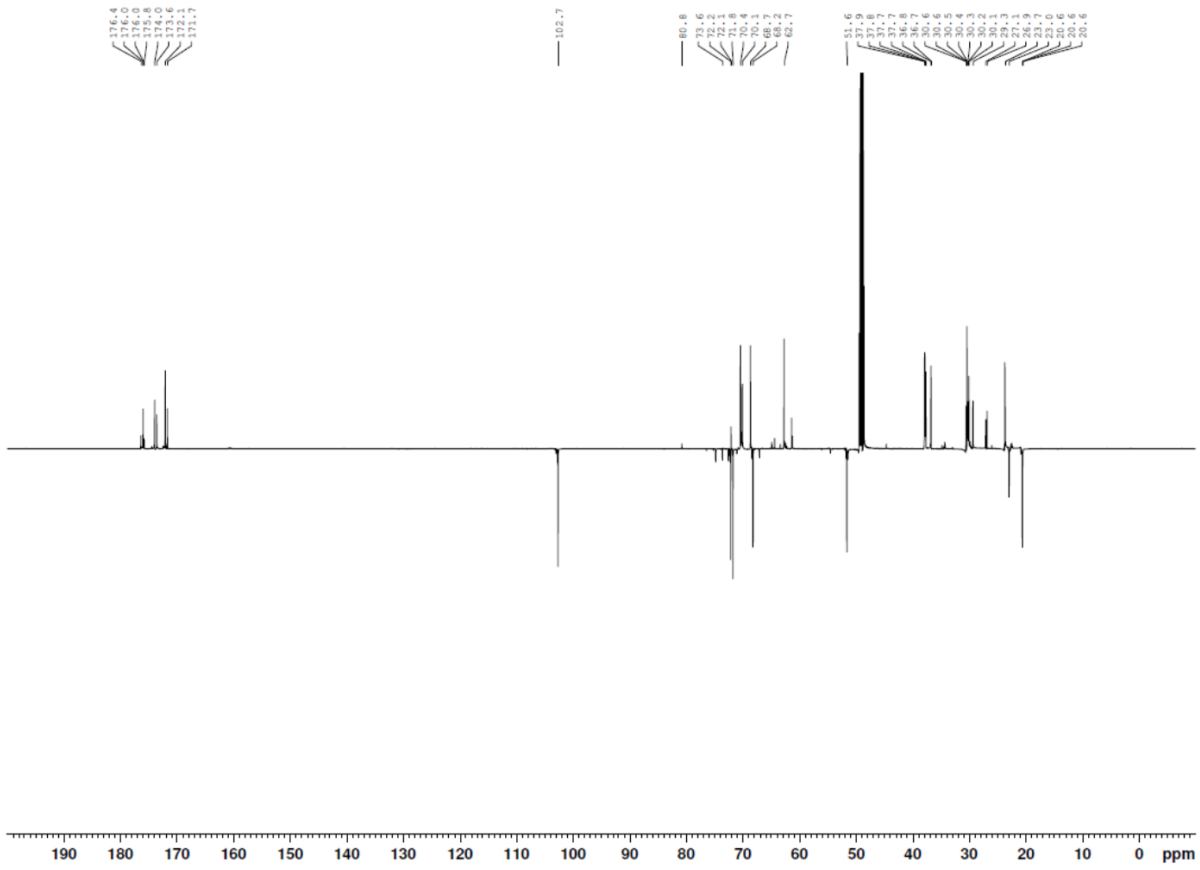
For compounds **21** to **25**, the following LCMS method was used:

Column: LUNA C18, 3µ, flow: 1.0 ml/min; gradient: 0 min 93% H_2O(0.05%TFA) 1.0 min 95% ACN; 95% ACN to 1.45 min; 7% ACN 1.50 min.

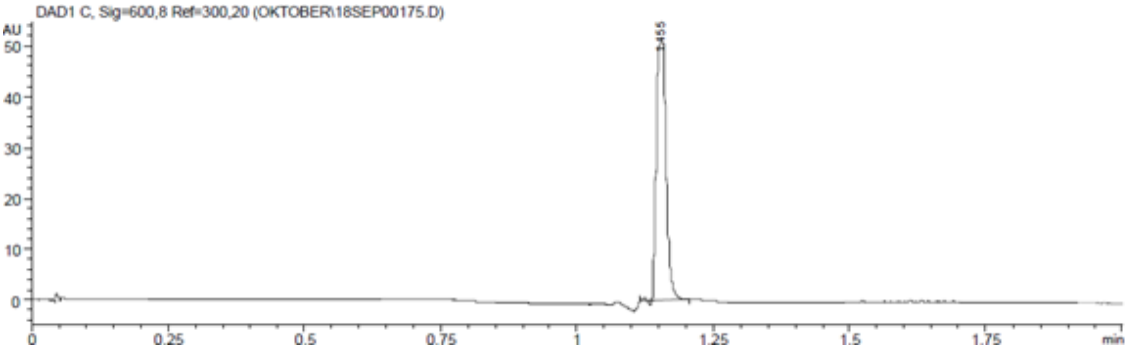


NMR spectra of compound **21**

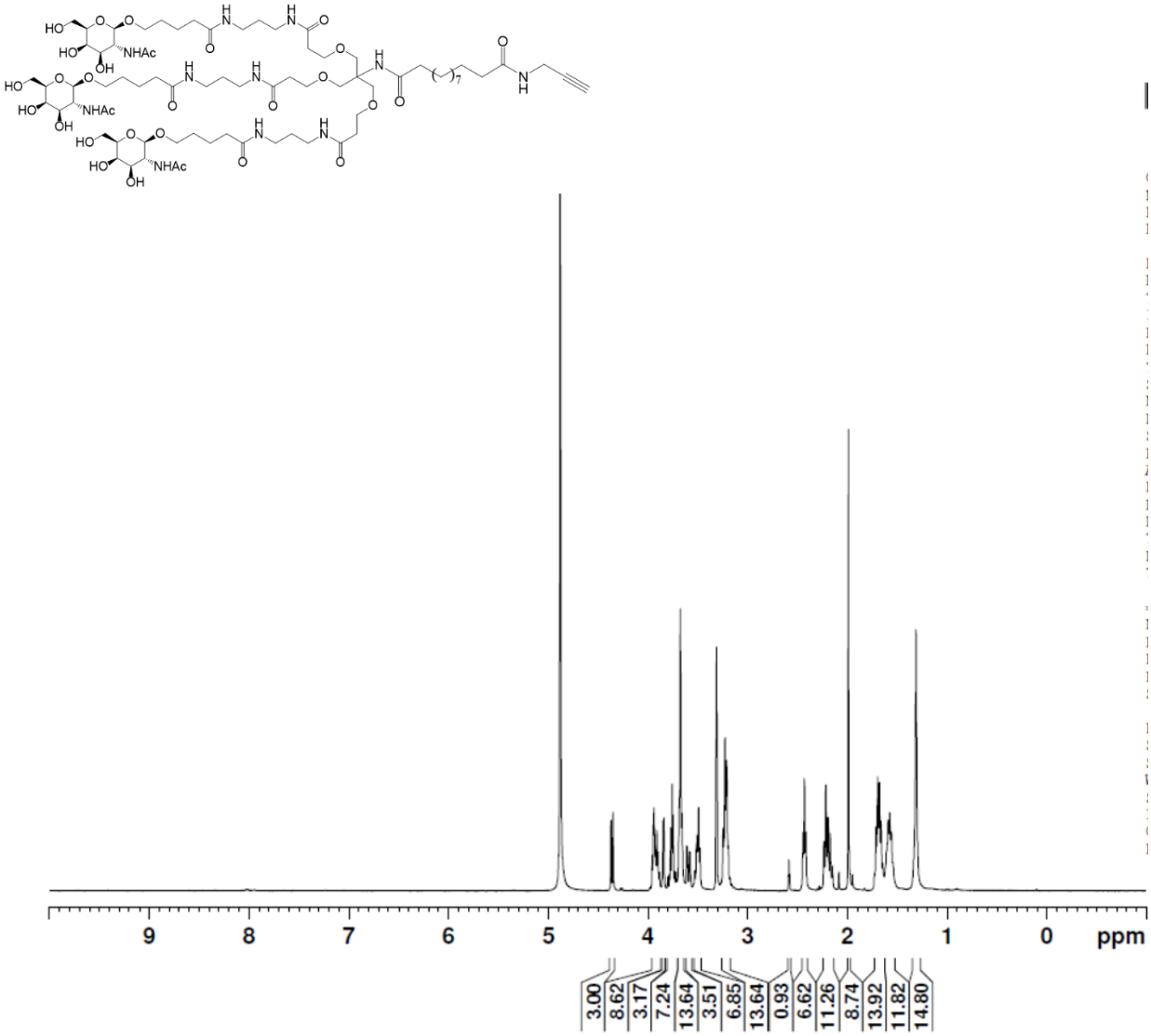


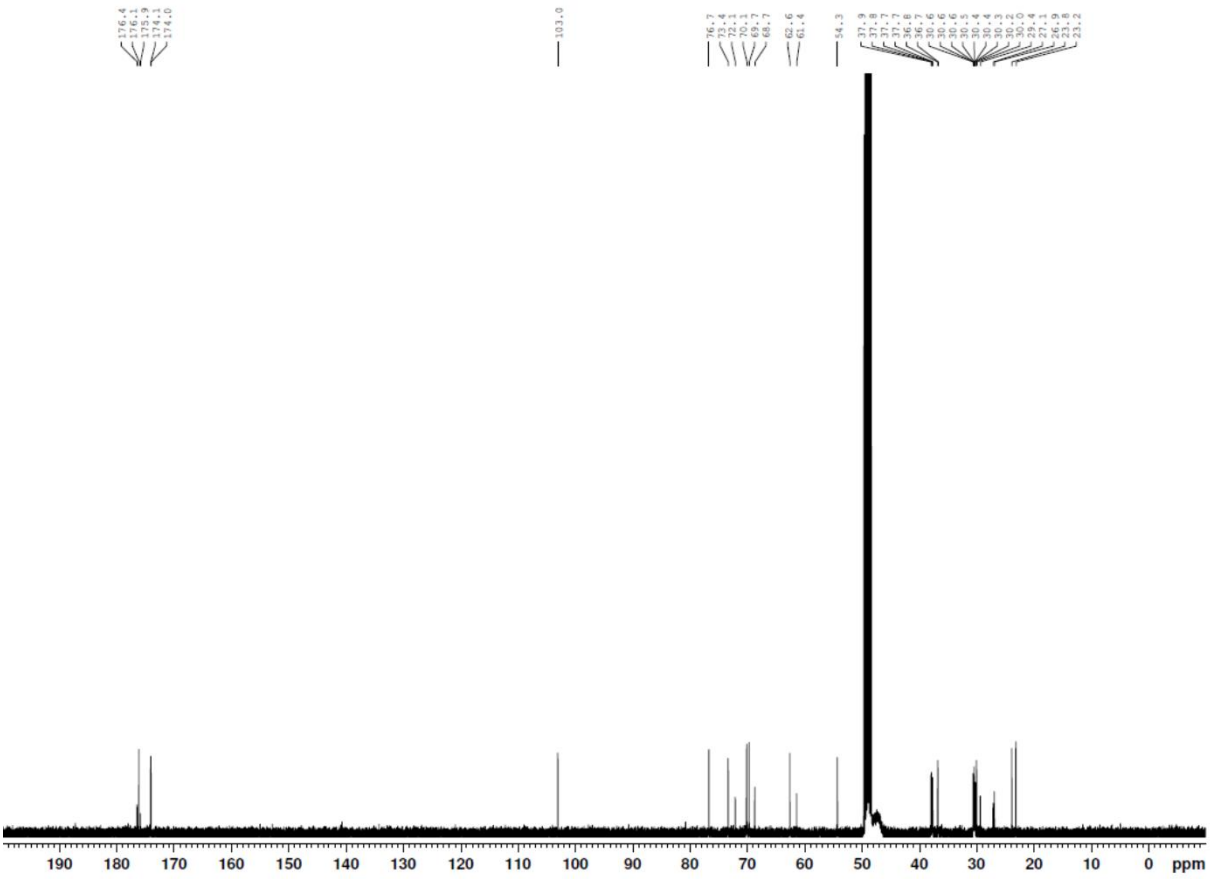


LCMS-UV (600 nm) spectrum of compound **22**

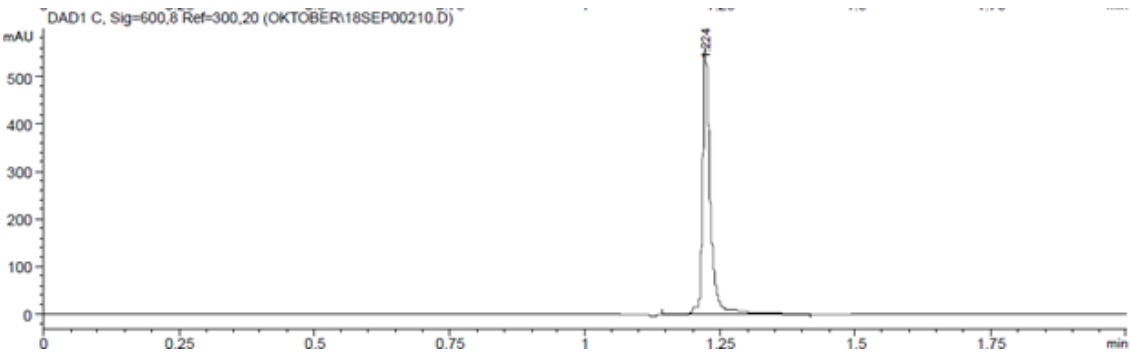


NMR spectra of compound **22**

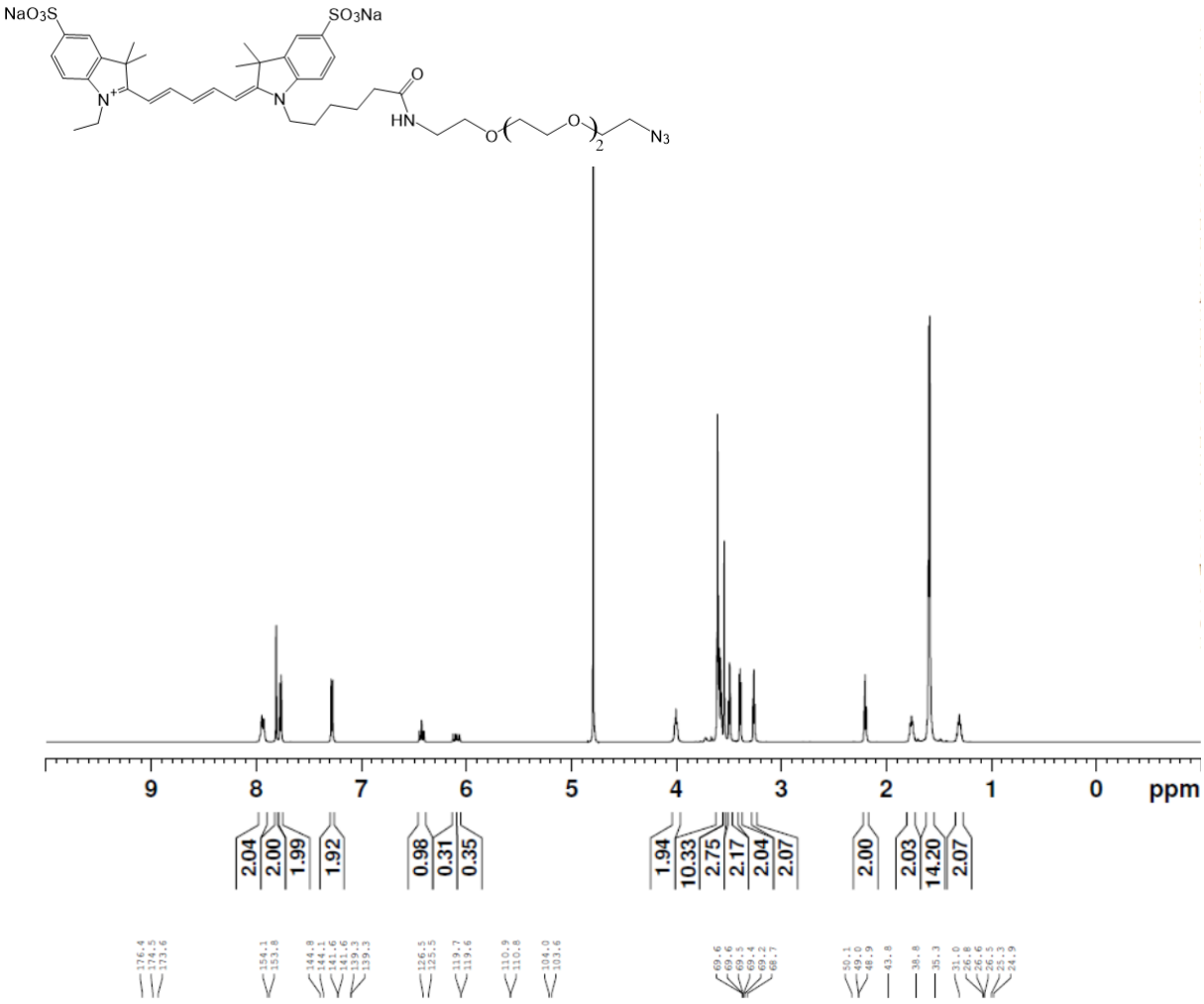


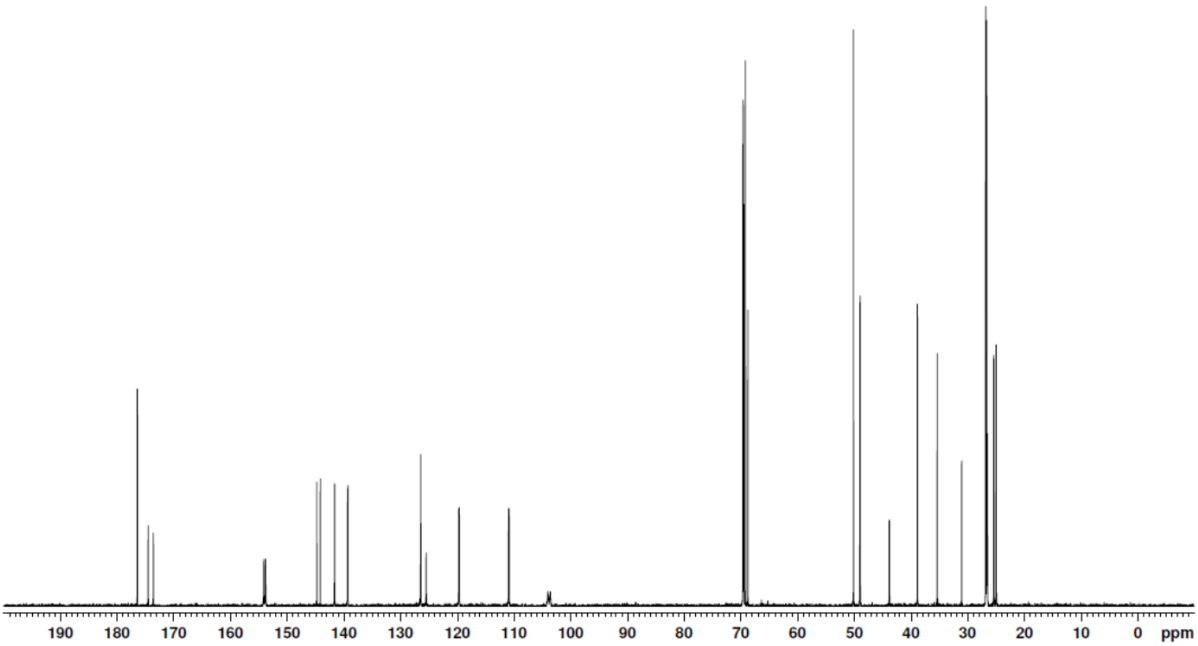


LCMS-UV (600 nm) spectrum of compound **24**

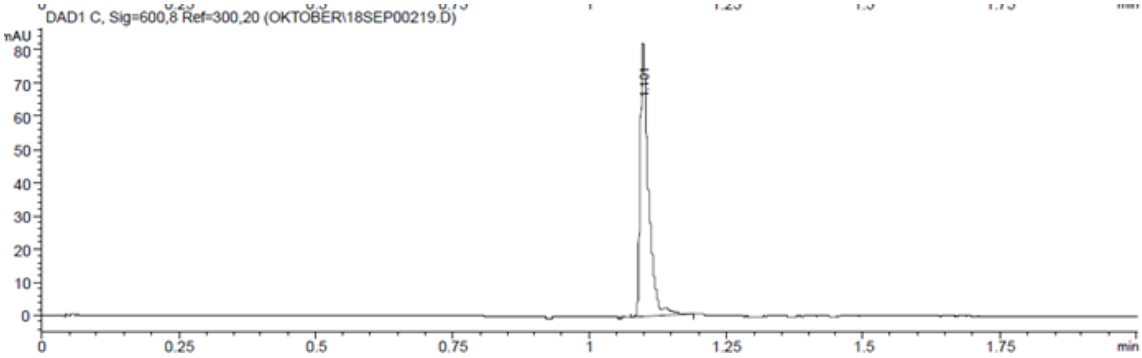


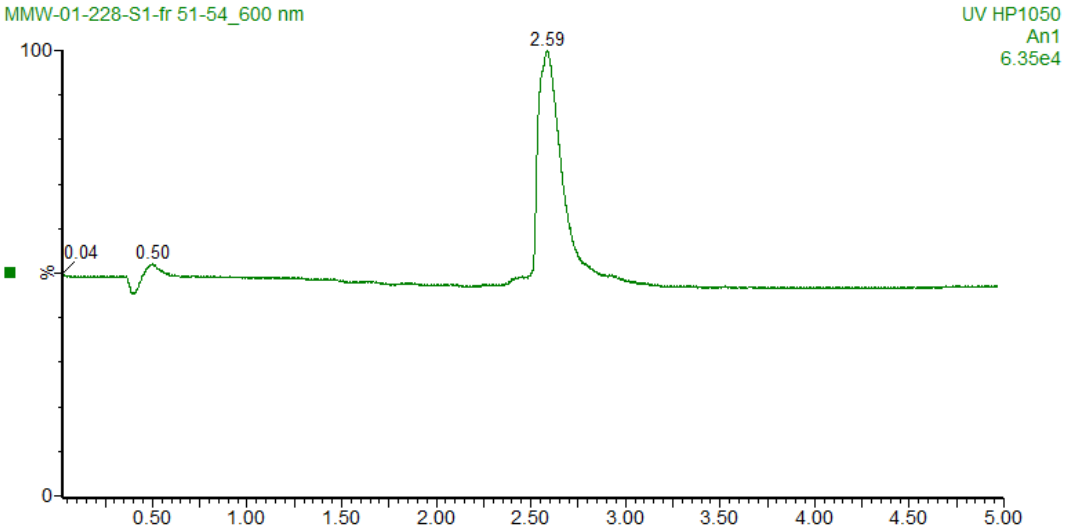
NMR spectra of compound **24**





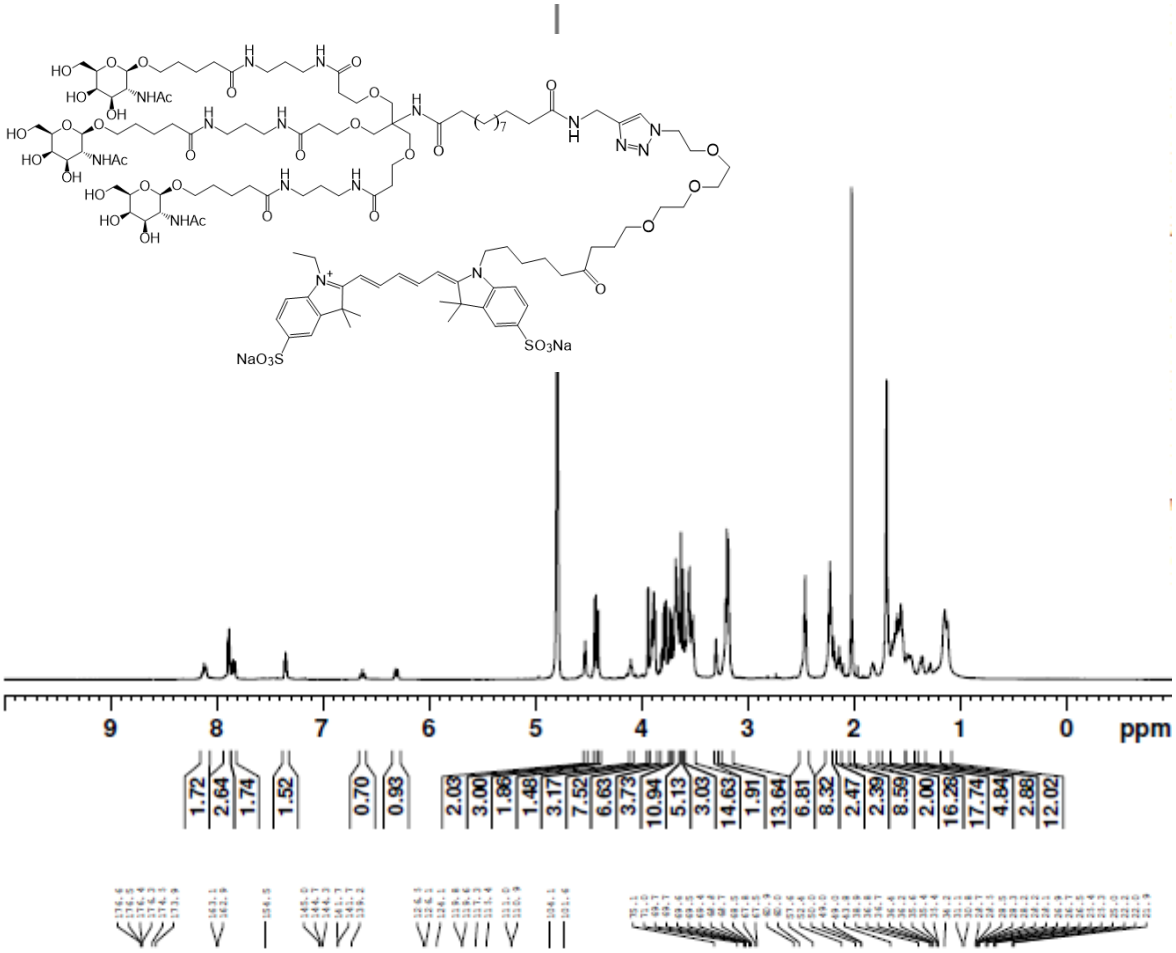
LCMS-UV (600 nm) spectrum of compound **25**

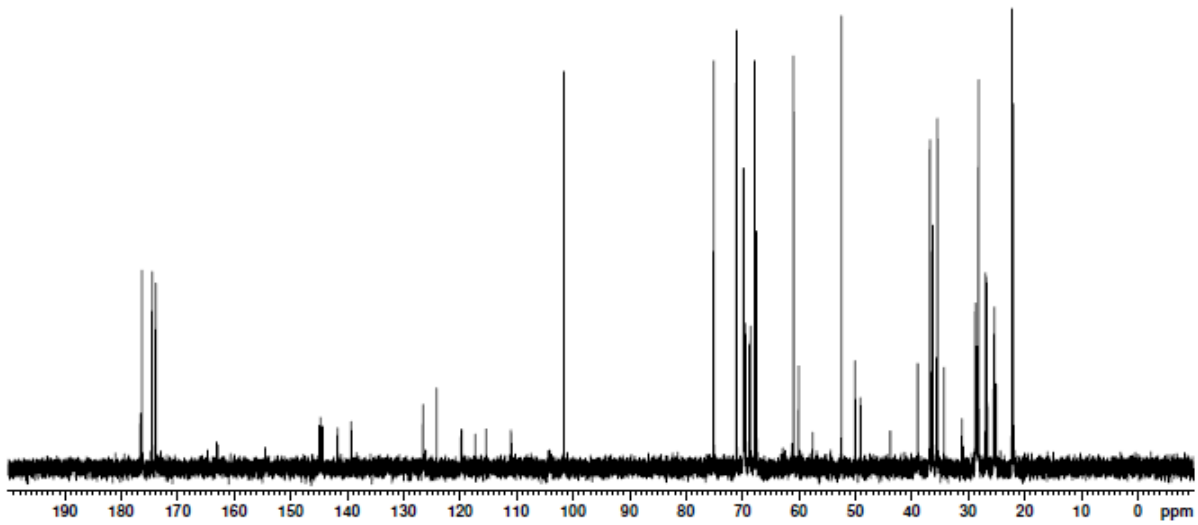




(Method II)

NMR spectra of compound **25**





Oligonucleotide analytics: Single strands

MS-results: siRNA **sense strands**

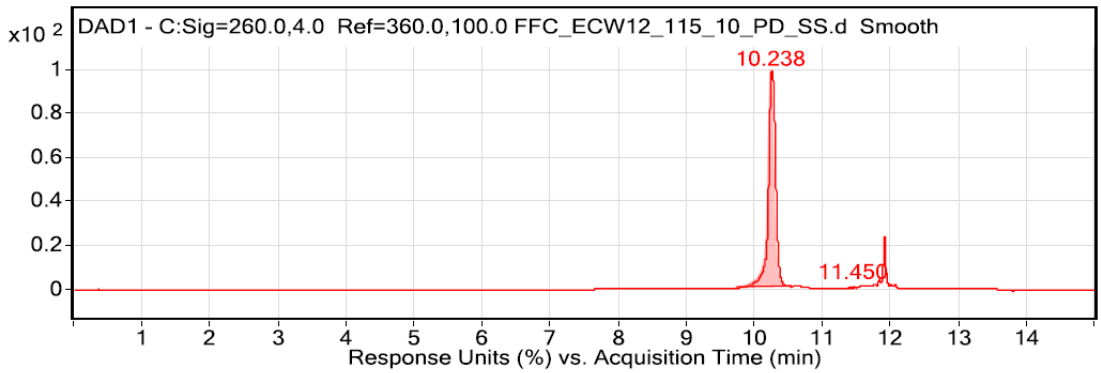
siRNA#	M(calc.) [m/z]	M(found) [m/z]	z
1-0	8965.45	2240.22	4
1-1	8668.93	8668.33	1
1-2	8963.50	8962.86	1
1-3	9071.33	9070.45	1
1-4	8668.93	8668.32	1
1-5	8963.50	8962.86	1
2-0	8044.59	2010.16	4
2-1	9071.33	9070.89	1
2-2	8807.01	8806.68	1
2-3	9071.33	9070.85	1
2-4	8807.01	8806.70	1
2-5	9467.58	9467.00	1
3-1	9605.65	9605.02	1

MS-results: siRNA **antisense strands**

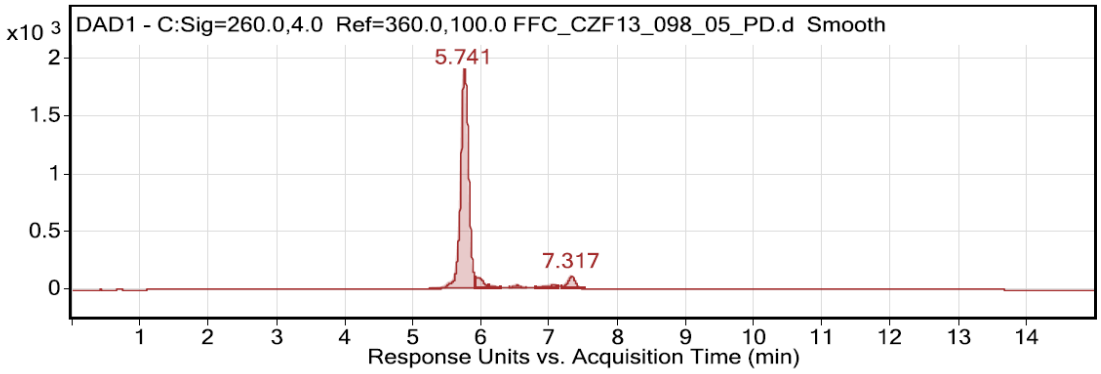
Antisense strand	M(calc.) [m/z]	M (found) [m/z]	z
neg. ctrl.	6903.58	1724.73	4
TTR	7595.96	7595.04	1

HPLC/UV-spectra (260 nm) of siRNA **sense strands**:

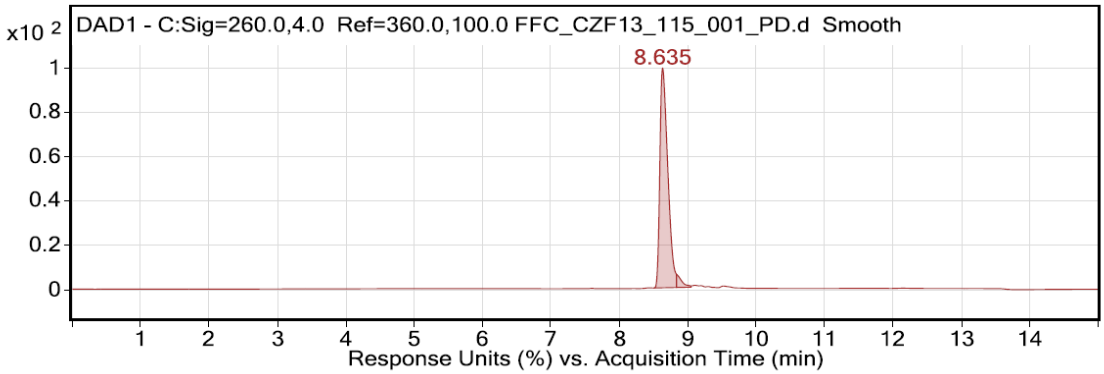
siRNA1-0 sense strand



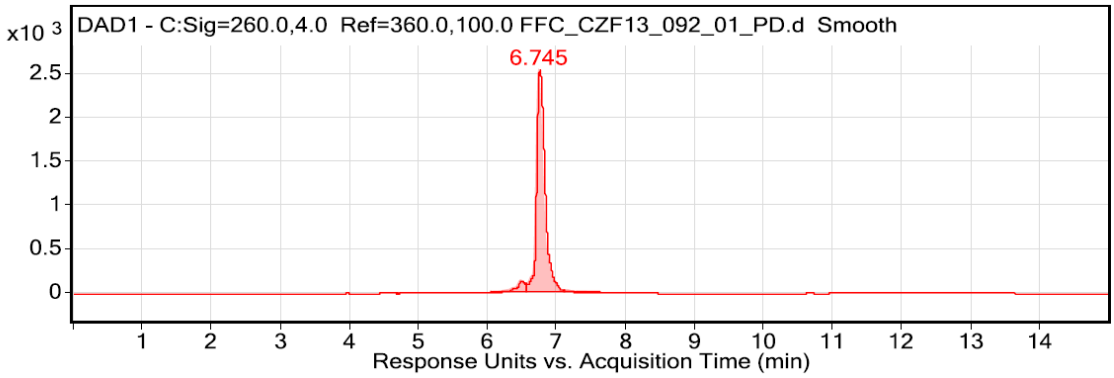
siRNA1-1 sense strand



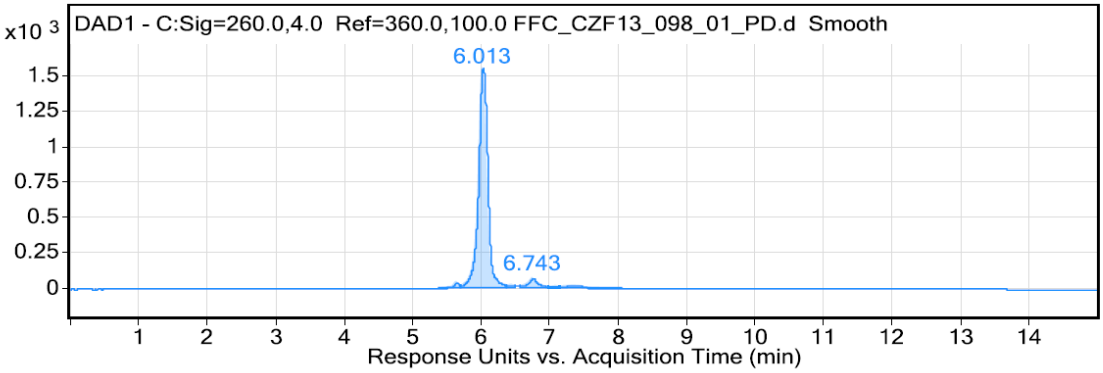
siRNA1-2 sense strand



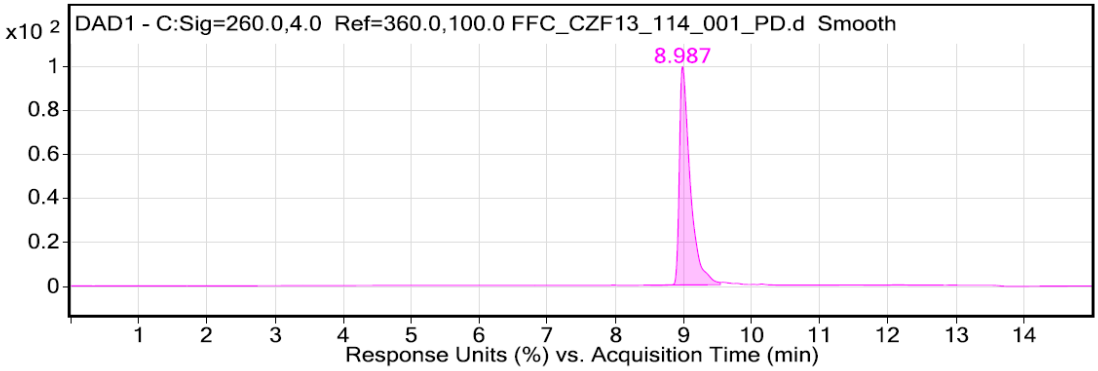
siRNA1-3 sense strand



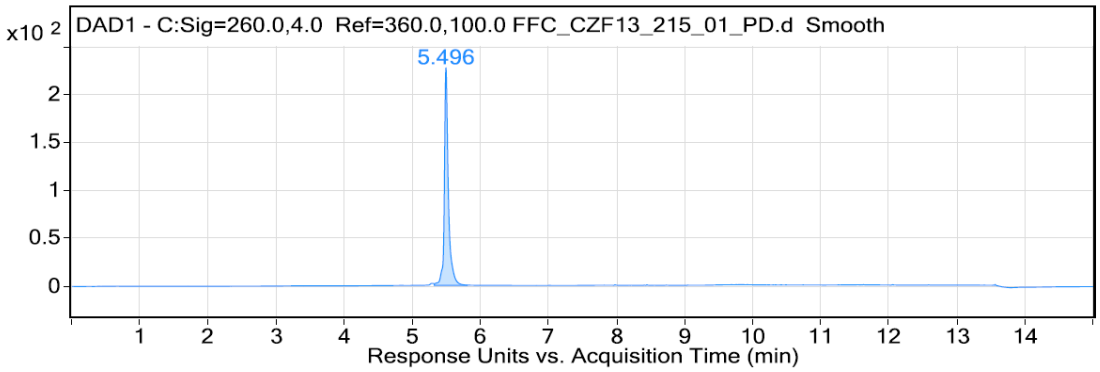
siRNA1-4 sense strand



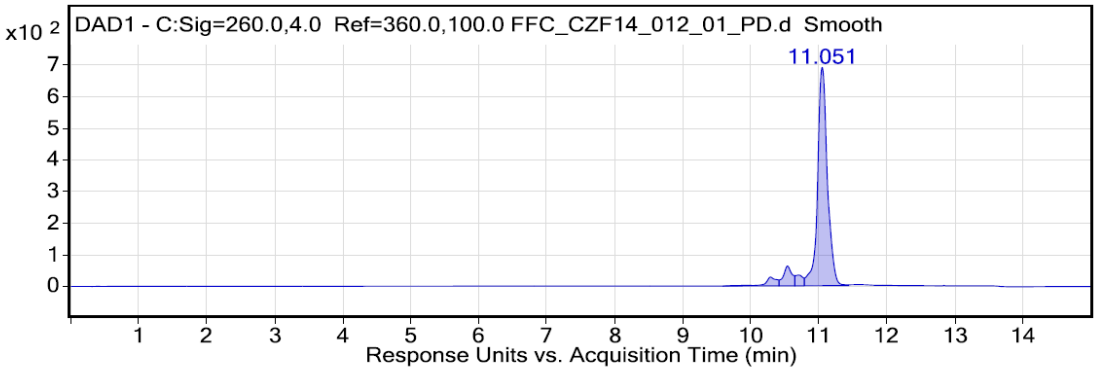
siRNA1-5 sense strand



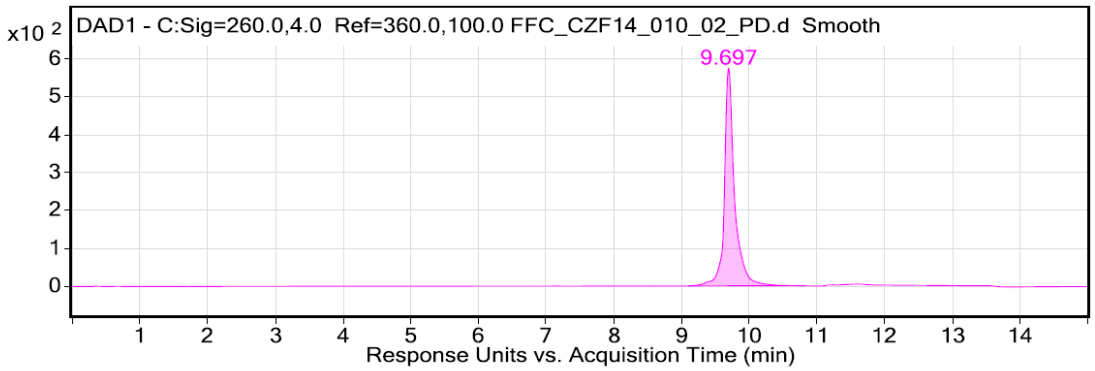
siRNA2-0 sense strand



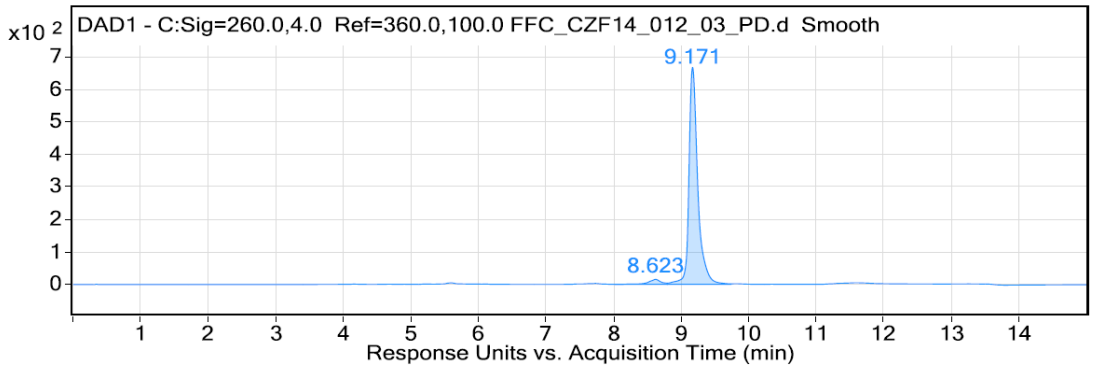
siRNA2-1 sense strand



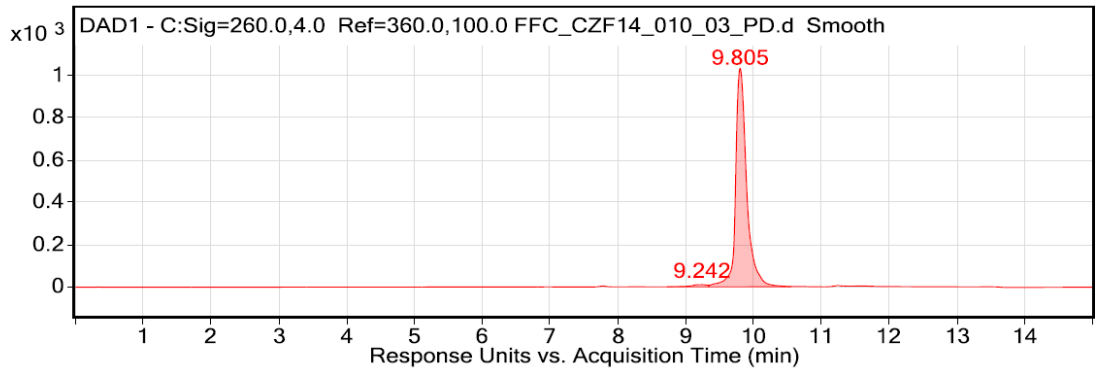
siRNA2-2 sense strand



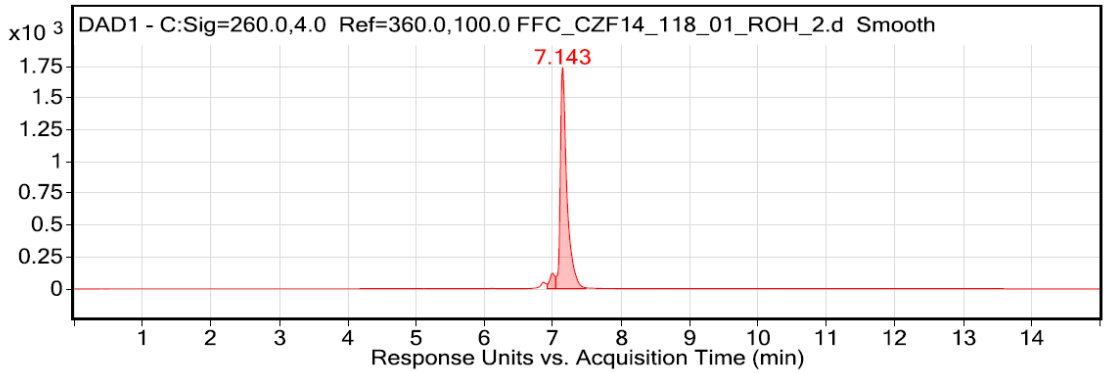
siRNA2-3 sense strand



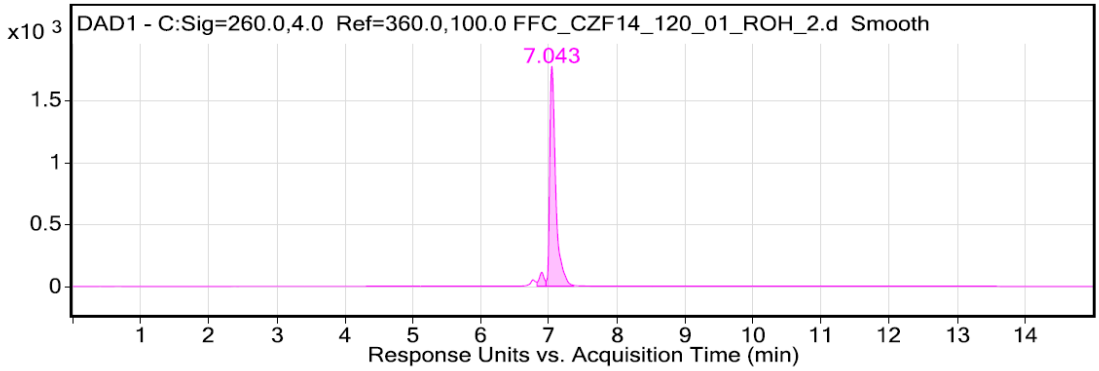
siRNA2-4 sense strand



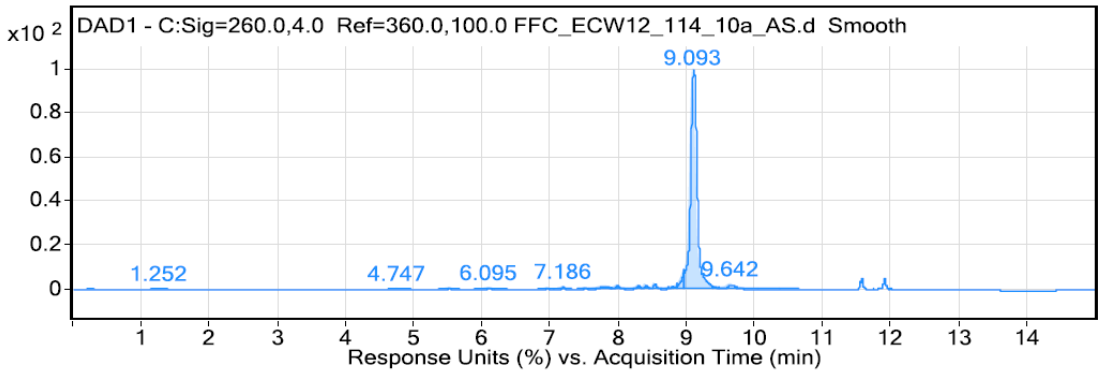
siRNA2-5 sense strand



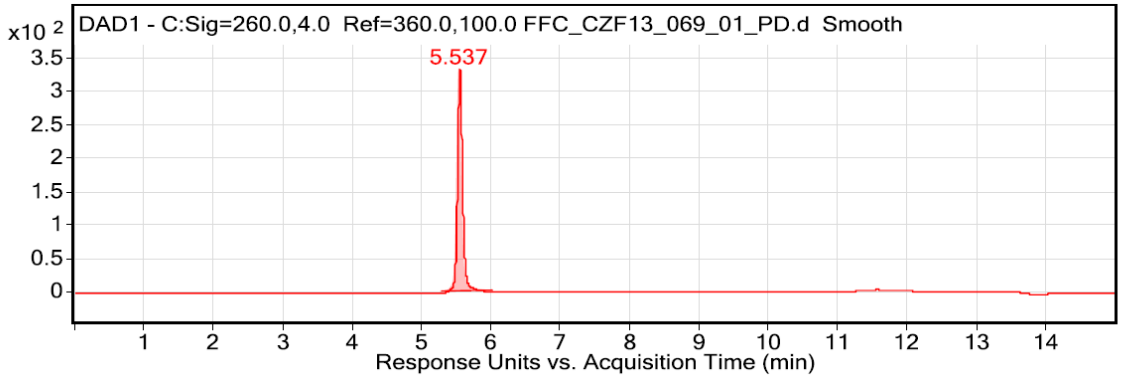
siRNA3-1 sense strand



antisense **neg. ctrl.**



antisense **TTR**



MS-results: siRNA **double strands**

MW of sense and antisense strand found

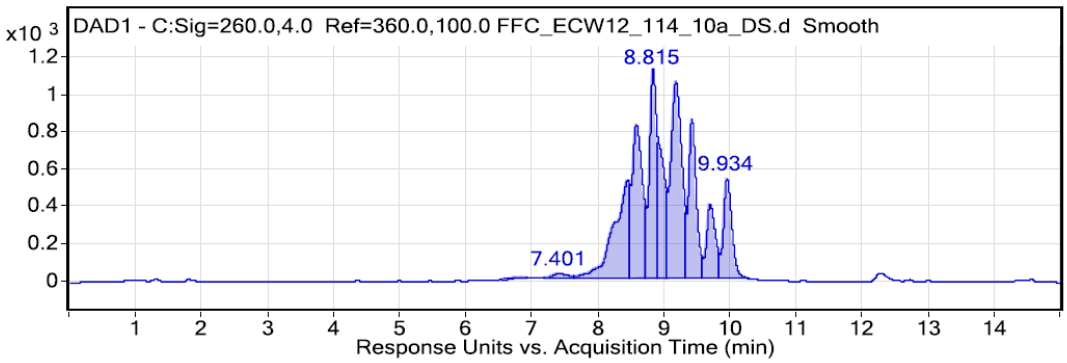
siRNA	**M(calc.) sense [m/z]**	**M (found) sense [m/z]**	**M (calc.) antisense [m/z]**	**M (found) antisense [m/z]**
1-1	8668.93	8668.62	7595.96	7595.10
1-2	8963.50	8962.82	7595.96	7594.99
1-3	9071.33	9070.79	7595.96	7595.10
1-4	8668.93	8668.59	7595.96	7595.07
1-5	8963.50	8962.84	7595.96	7596.00

MW of siRNA double strand found

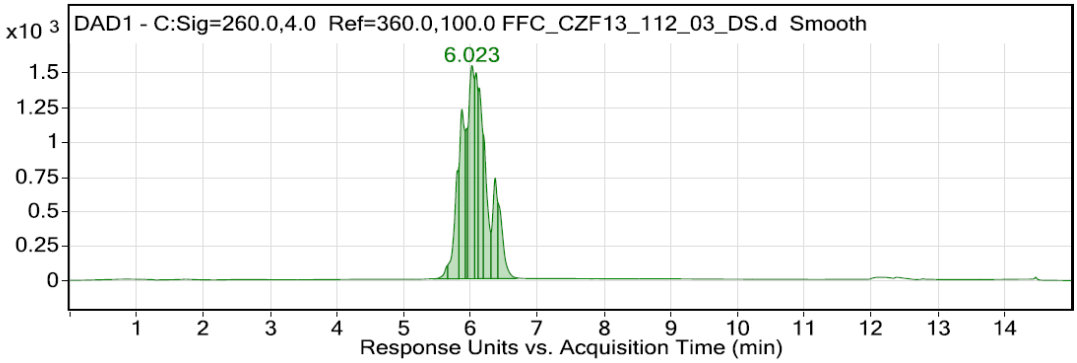
siRNA	**M(calc.) siRNA [m/z]**	**M(found) siRNA [m/z]**	**z**
1-0	15869.03	2643.77	6
2-0	14948.12	2490.19	6
2-1	16667.29	2776.78	6
2-2	16402.97	2732.76	6
2-3	16667.29	2776.78	6
2-4	16402.97	2732.76	6
2-5	17063.54	2842.83	6
3-1	17201.61	2865.84	6
3-2	17233.73	2871.16	6

HPLC/UV-spectra (260 nm) of siRNA **double strands**:

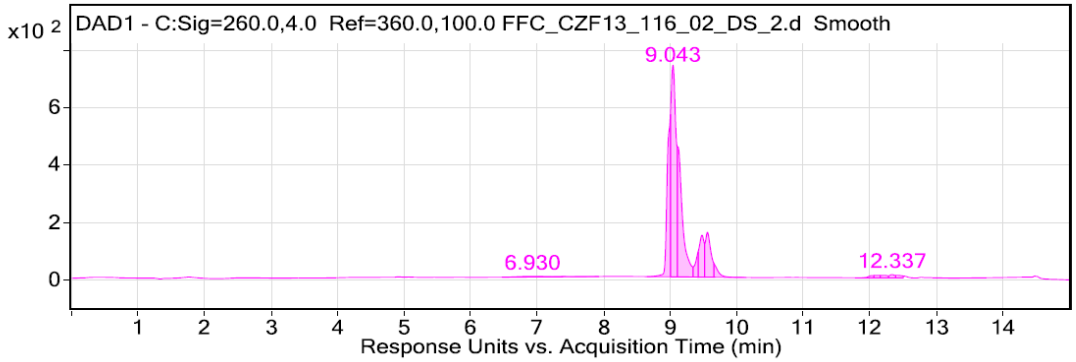
siRNA1-0



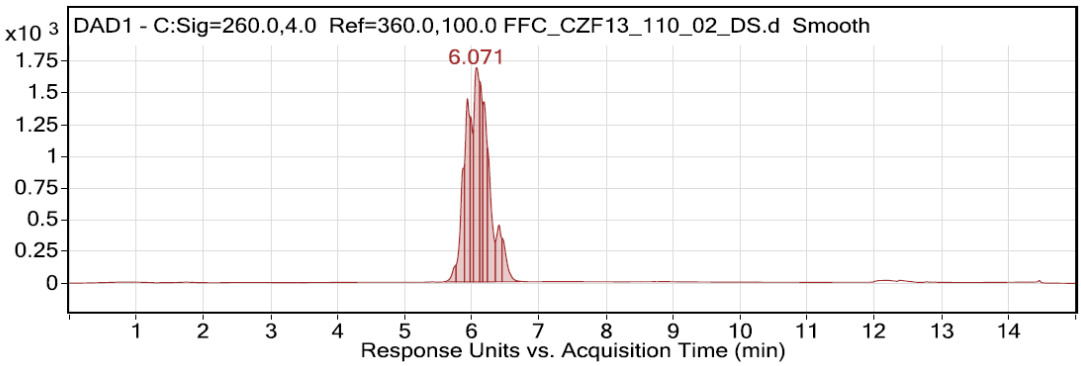
siRNA1-1



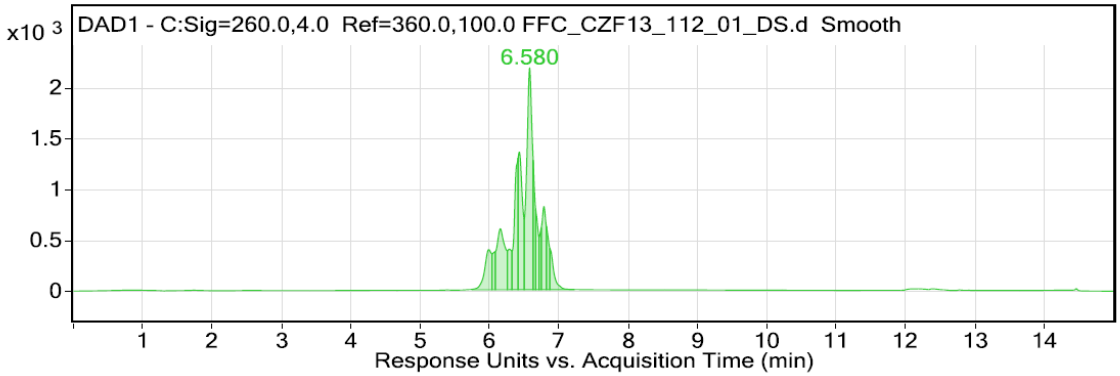
siRNA1-2



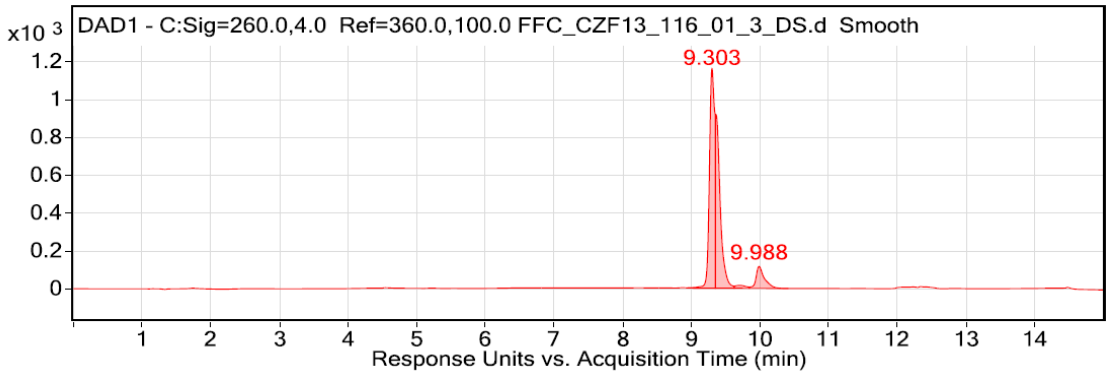
siRNA1-3



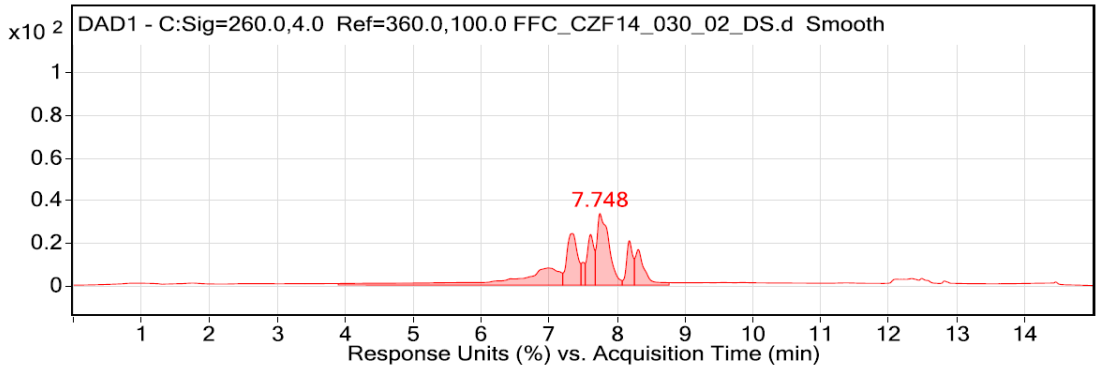
siRNA1-4



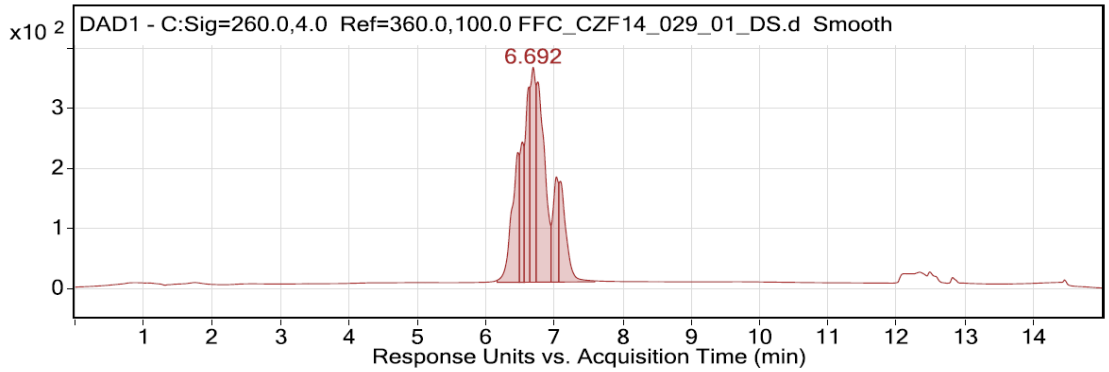
siRNA1-5



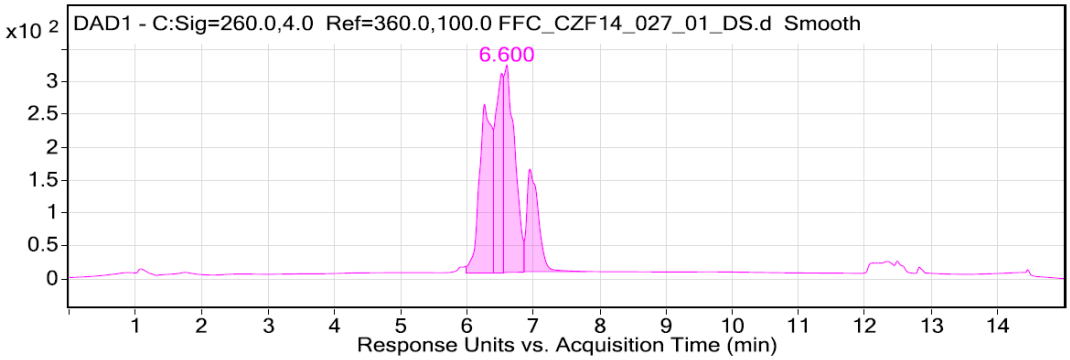
siRNA2-0



siRNA2-1



siRNA2-2



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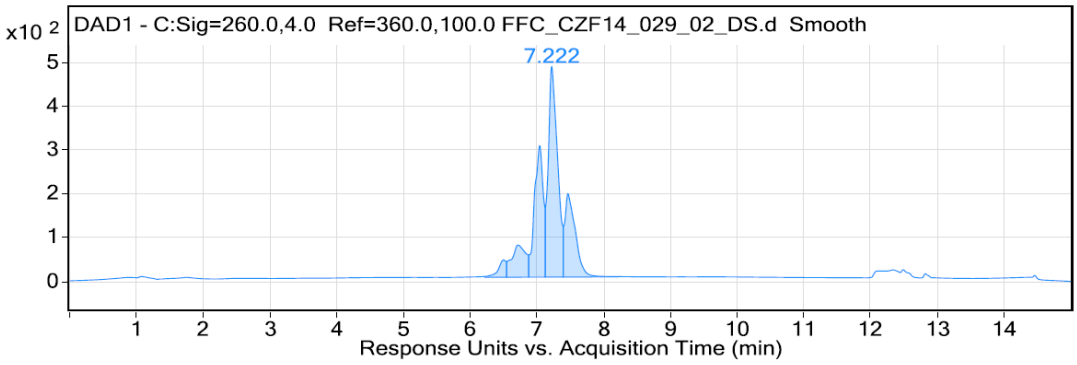

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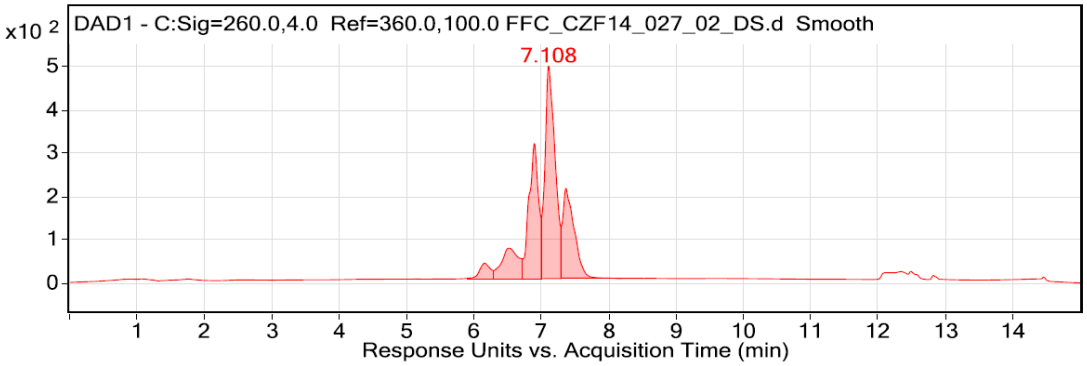

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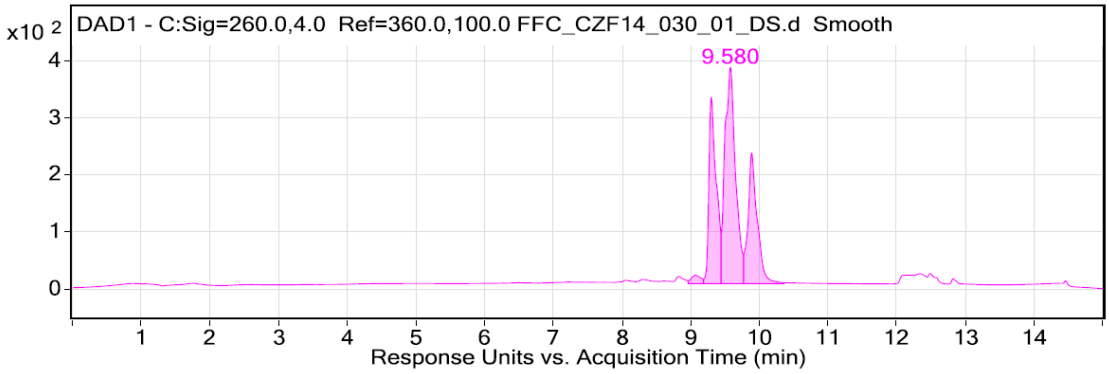
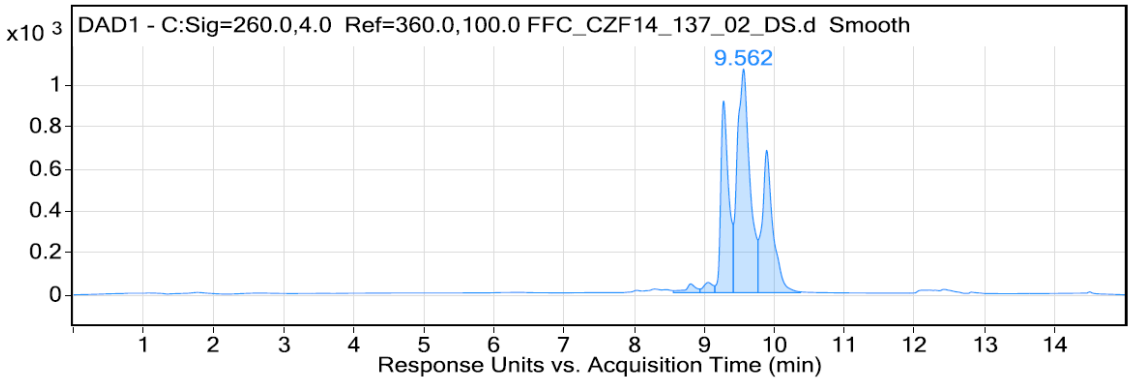


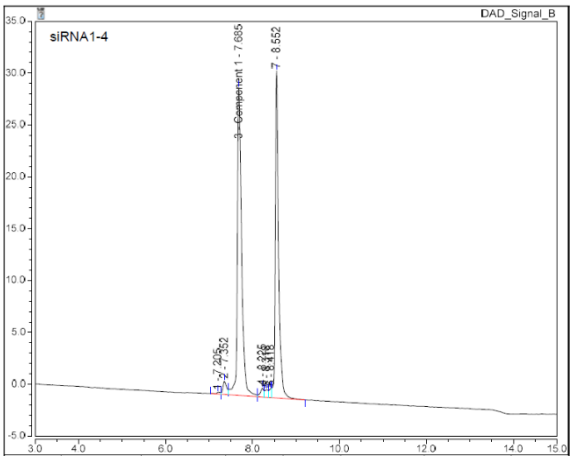
siRNA3-1



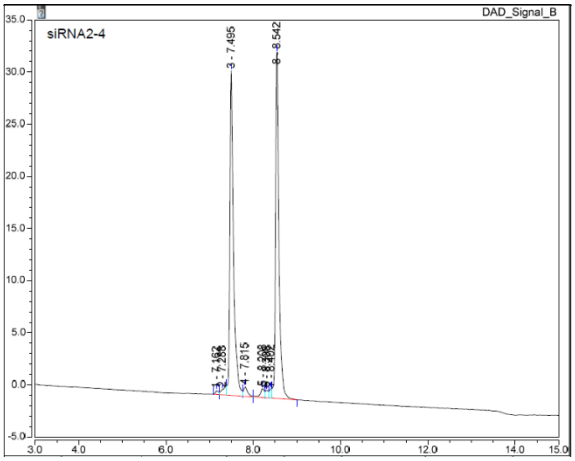
Ion exchange chromatograms from stability assay of siRNA1-4, siRNA2-4, siRNA2-5 and siRNA3-1 in mouse serum after 0, 24, 48, 72, 96 and 168 h.

<u>Starting materials siRNA1-4, siRNA2-4, siRNA2-5 and siRNA3-1</u>

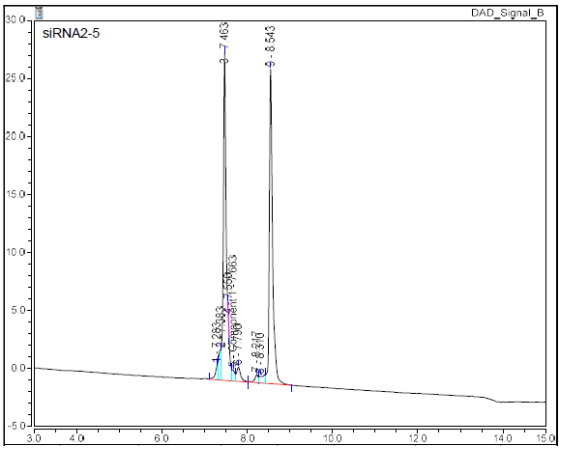
siRNA1-4



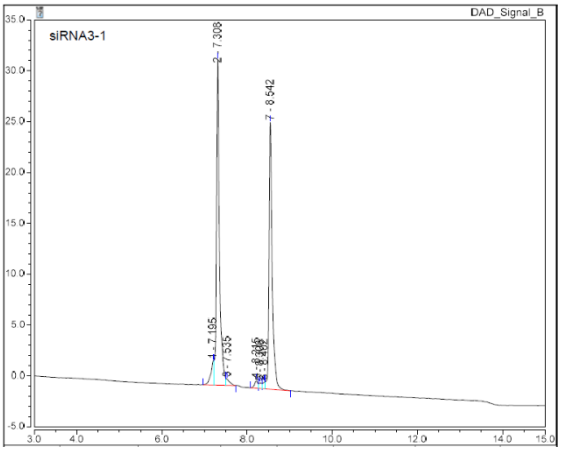
siRNA2-4



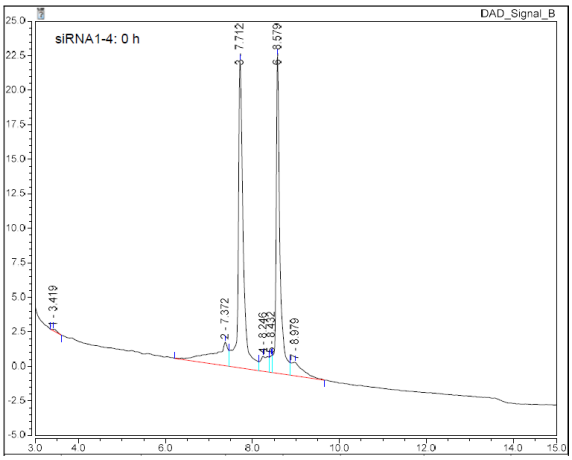
siRNA2-5

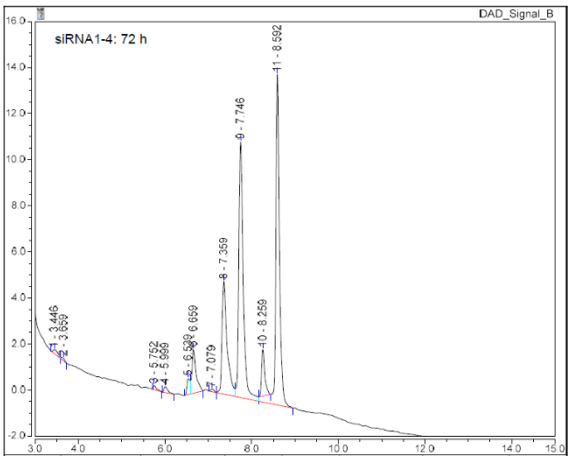


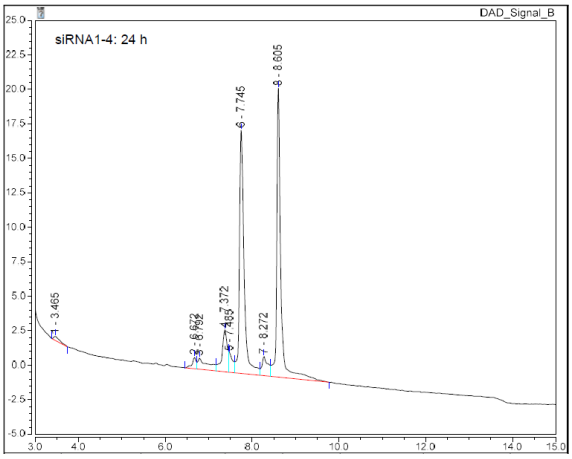
siRNA3-1

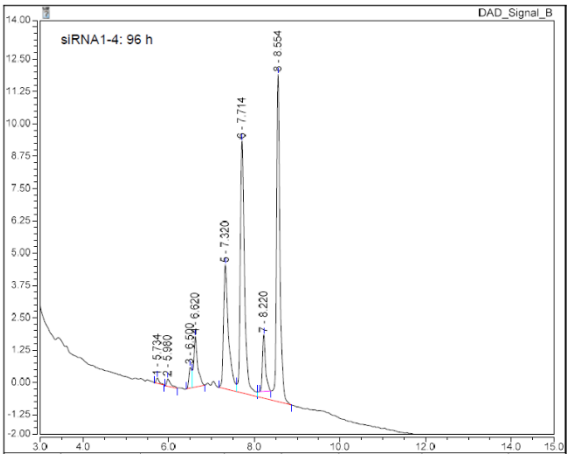


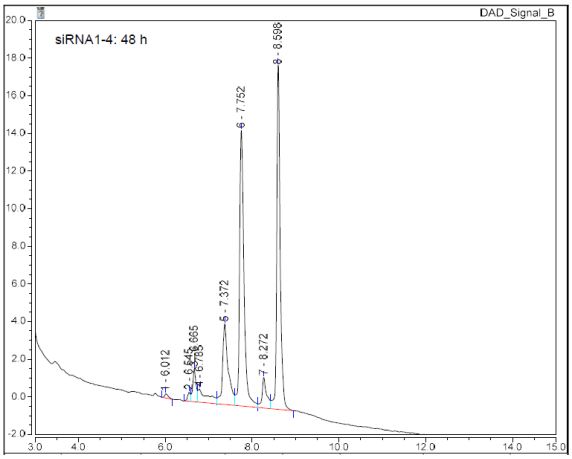
siRNA1-4: ion exchange chromatograms of test samples at 0, 24, 48, 72, 96 and 168 h

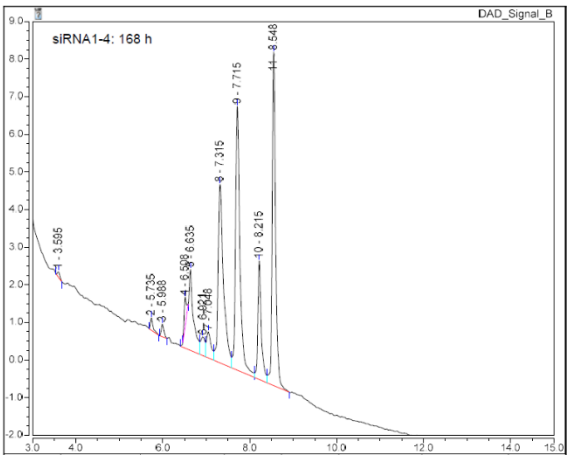




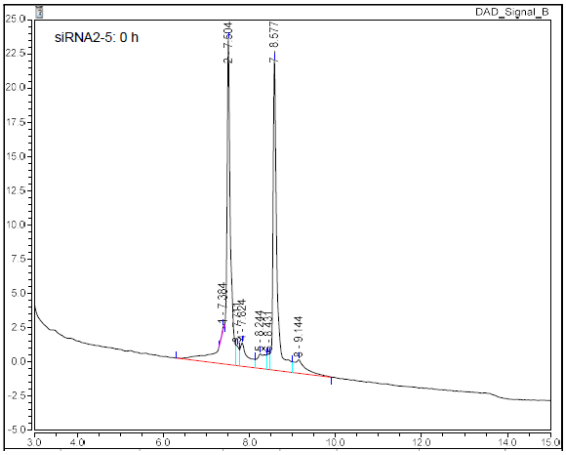


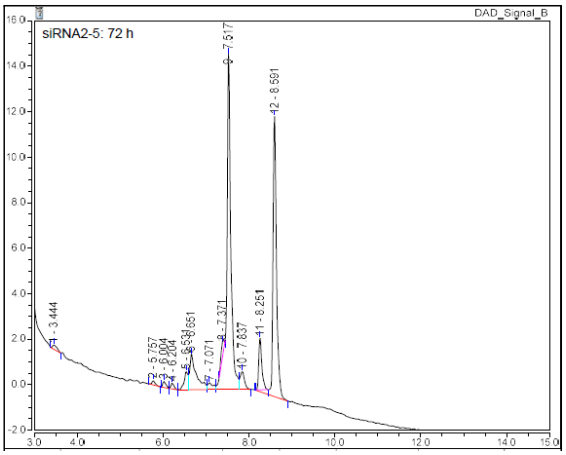


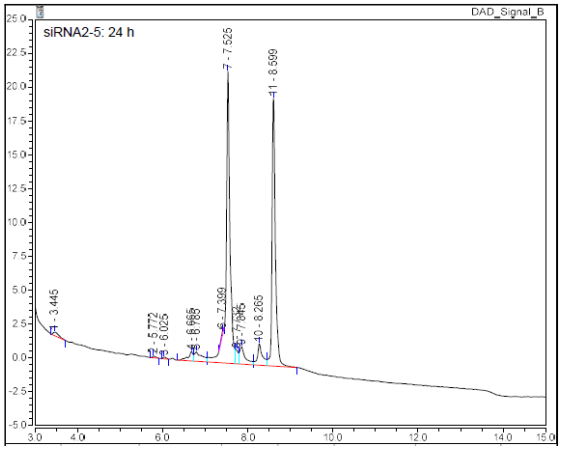


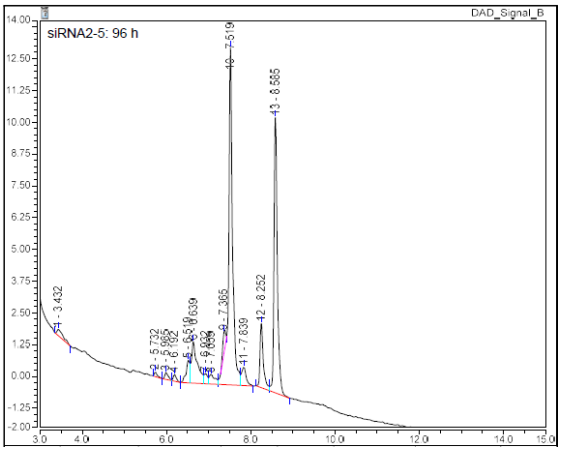


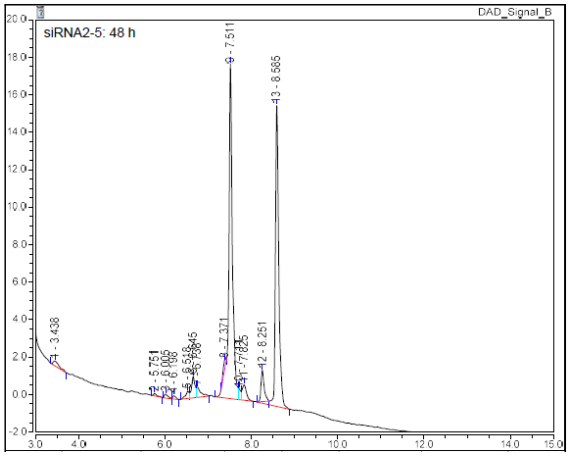
siRNA2-5: ion exchange chromatograms of test samples at 0, 24, 48, 72, 96 and 168 h

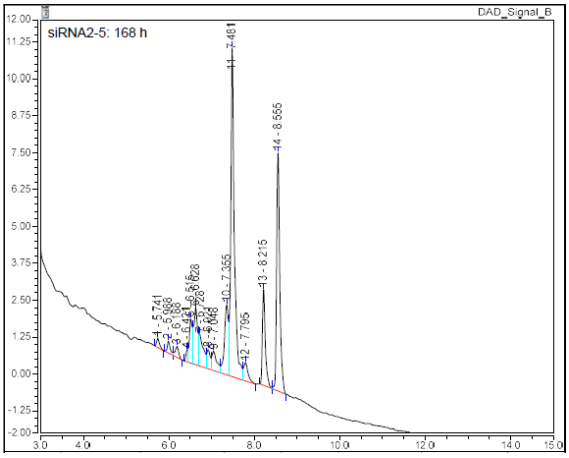




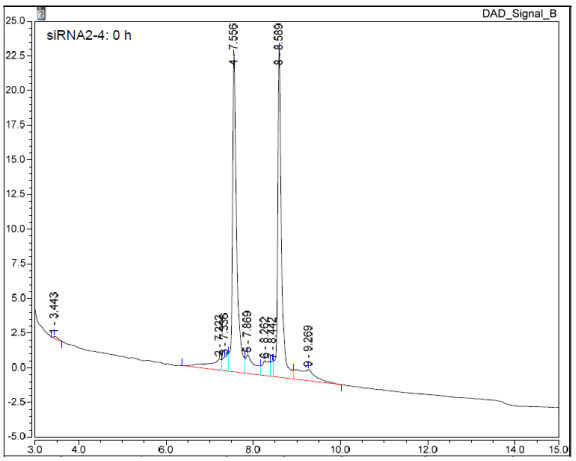


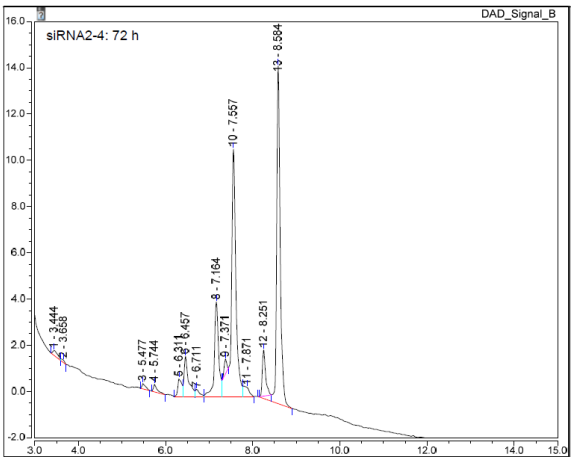


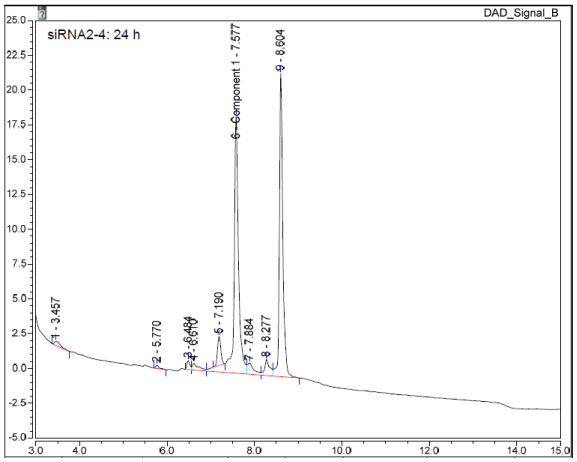


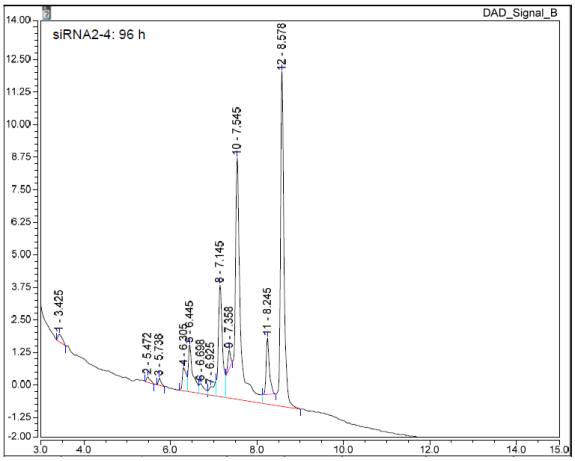


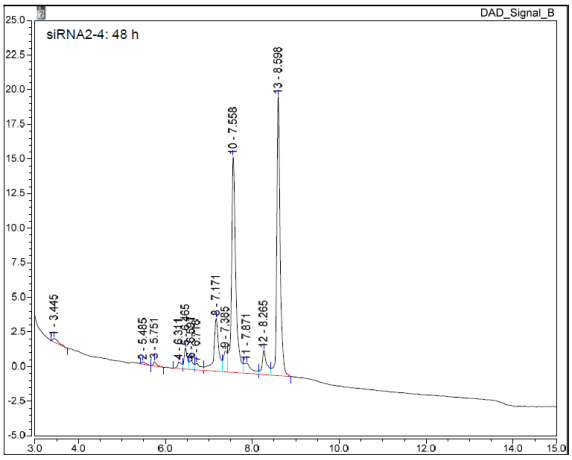
siRNA2-4: ion exchange chromatograms of test samples at 0, 24, 48, 72, 96 and 168 h

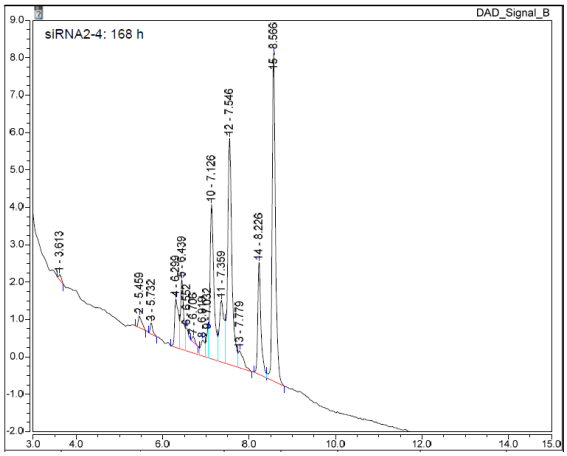
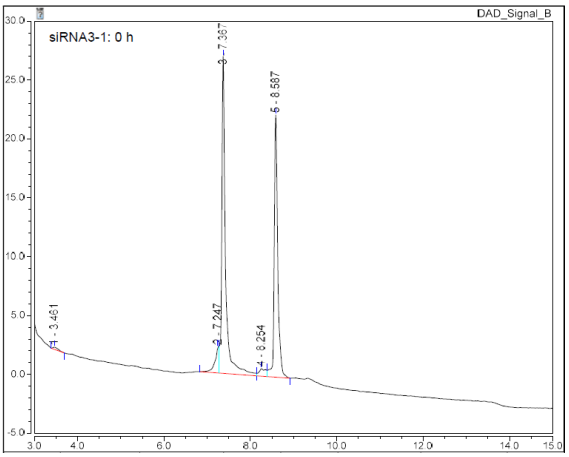
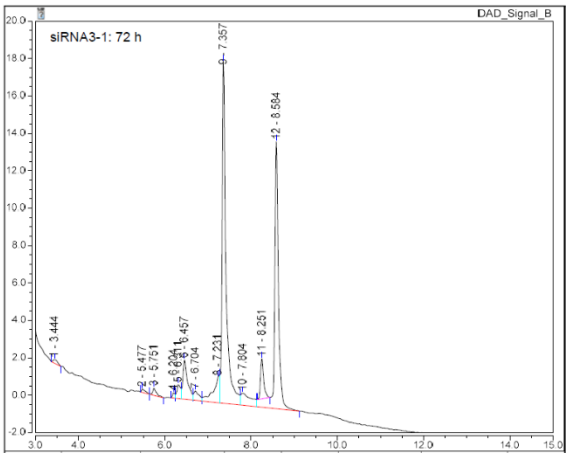
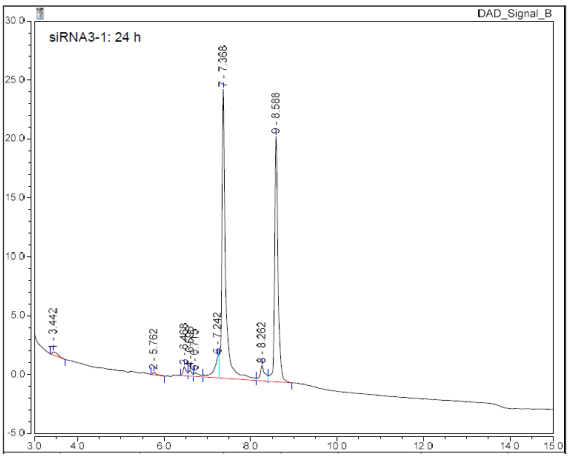
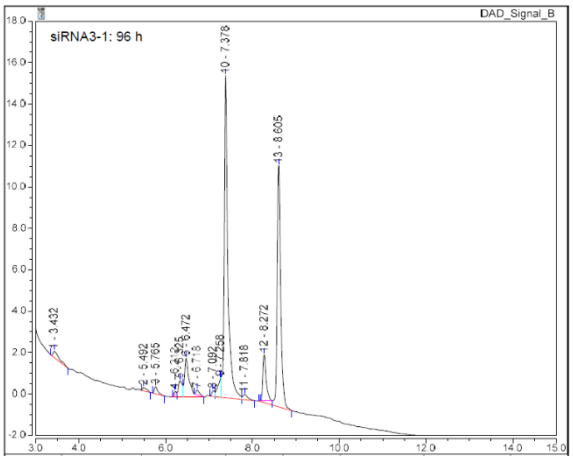
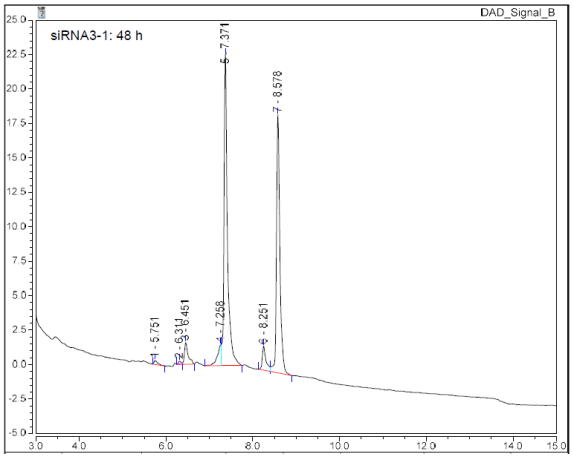
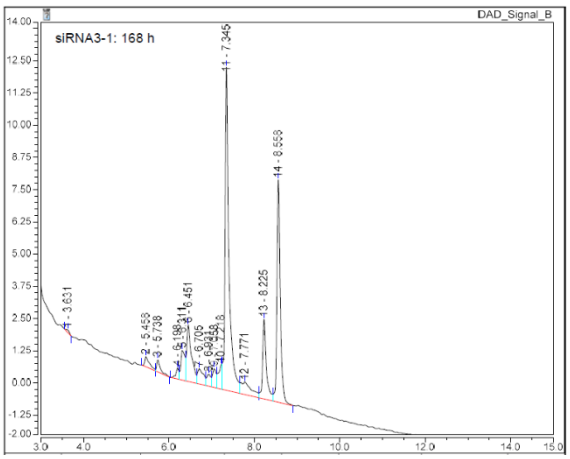












siRNA3-1: ion exchange chromatograms of test samples at 0, 24, 48, 72, 96 and 168 h