Supplementary data for the article:

Vitorović-Todorović, M. D.; Koukoulitsa, C.; Juranić, I. O.; Mandić, L. M.; Drakulić, B. J. Structural Modifications of 4-Aryl-4-Oxo-2-Aminylbutanamides and Their Acetyl- and Butyrylcholinesterase Inhibitory Activity. Investigation of AChE-Ligand Interactions by Docking Calculations and Molecular Dynamics Simulations. *European Journal of Medicinal Chemistry* **2014**, *81*, 158–175. https://doi.org/10.1016/j.ejmech.2014.05.008

Structural modifications of 4-aryl-4-oxo-2-aminylbutyramides and their acetyl- and butyrylcholinesterase inhibitory activity. Investigation of AChE-ligand interactions by docking calculations and molecular dynamics simulations.

Maja D. Vitorović-Todorović[a*], Catherine Koukoulitsa[b], Ivan O. Juranić[c], Ljuba M. Mandić[d], Branko J. Drakulić[c]

[a] *Military Technical Institute, Ratka Resanovića 1, Belgrade, Serbia*
[b] *Department of Chemistry, University of Athens, Panepistimiopolis-Zografou, 15771, Greece*
[c] *Department of Chemistry-IChTM, University of Belgrade, Njegoševa 12, Belgrade, Serbia*
[d] *Faculty of Chemistry, University of Belgrade, Studentski trg 12-16, Belgrade, Serbia*

Table S1. Data of enzymatic analysis (K_i) for the compound **17**.

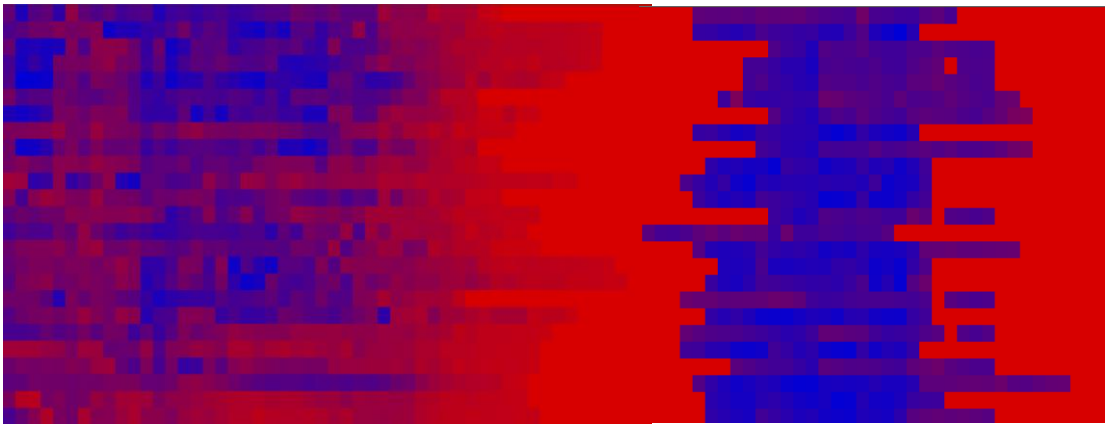
1/[S] x 10^3 (mol^{-1})	**V^{-1} (min/ΔA) for [I] (μM)**			
	0,00	**2,00**	**4,00**	**5,00**
5.05	3.571	5.102	8.064	13.888
5.618	3.676	5.555	8.333	15.625
6.329	3.789	5.814	8.621	16.666
7.246	4.111	6.25	9.615	17.857
8.928	4.386	6.579	10.417	19.23

Table S2. Predicted potency of the compounds based on model described in reference 22, given in the main text.

Compound N^{o}	**Exp. p(IC_{50})***	**Calc. p(IC_{50})**
2	5.348	5.478
6	5.633	5.634
14	5.481	5.433
17	5.708	5.555
18	5.688	5.503
21	5.264	5.088
22	5.226	5.329
25	5.196	5.197
26	5.491	5.544
30	5.627	5.759
34	4.699	5.224

* Recalculated from M concentrations.

* Corresponding author: mvitod@chem.bg.ac.rs



a) **b)**

Figure S3. Heatmaps of: a) DRY-DRY block of variables; b) N1-N1 block of variables. Compounds were arranged from the most to the least active one, from the top to the bottom of the Figure.

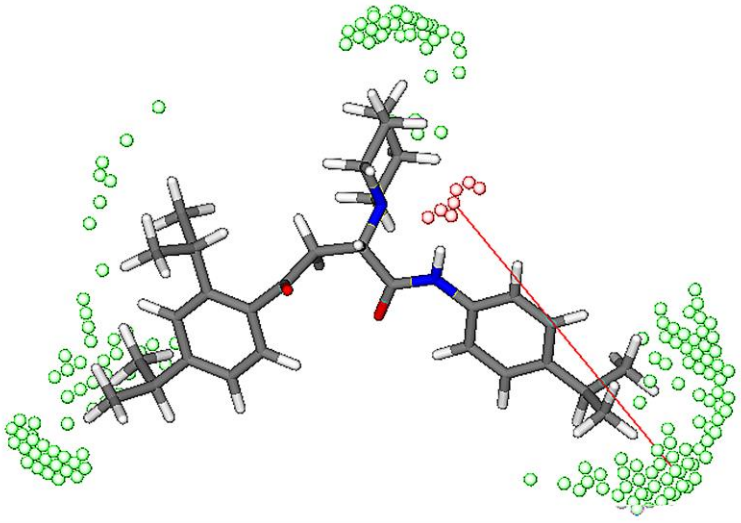


Figure S4. Variable O-TIP 473 (10.0-10.4 Å), on the example of the most active derivative **18**.

Table S5. Experimental *vs*. calculated p(IC_{50}) values obtained with 3LV from PLS model described in the main text. In the last column, association of variable O-TIP 473 with compounds is shown.

Compound Nº	**Exp. p(IC_{50})**	**Calc. p(IC_{50})**	O-TIP 473 Variable*
1a	5.460	5.203	–
2a	5.085	5.031	+
4a	5.313	5.372	+
5a	5.810	5.716	+
7a	5.249	5.365	+
8a	5.198	5.199	+
2	5.348	5.451	+
4	4.553	4.589	+
6	5.633	5.561	+
9	4.638	4.657	–
10	4.602	4.646	–

11	4.398	4.401	–
12	4.398	4.400	–
14	5.482	5.506	+
17	5.708	5.794	+
18	5.688	5.660	+
19	5.131	5.302	+
20	5.143	5.218	+
21	5.264	5.185	+
22	5.226	5.126	+
25	5.196	5.204	+
26	5.491	5.401	+
30	5.627	5.619	+
32	4.921	5.019	+
34	4.699	4.634	–

* (+) - Variable has a value different from 0. (–) - Variable has a value 0.

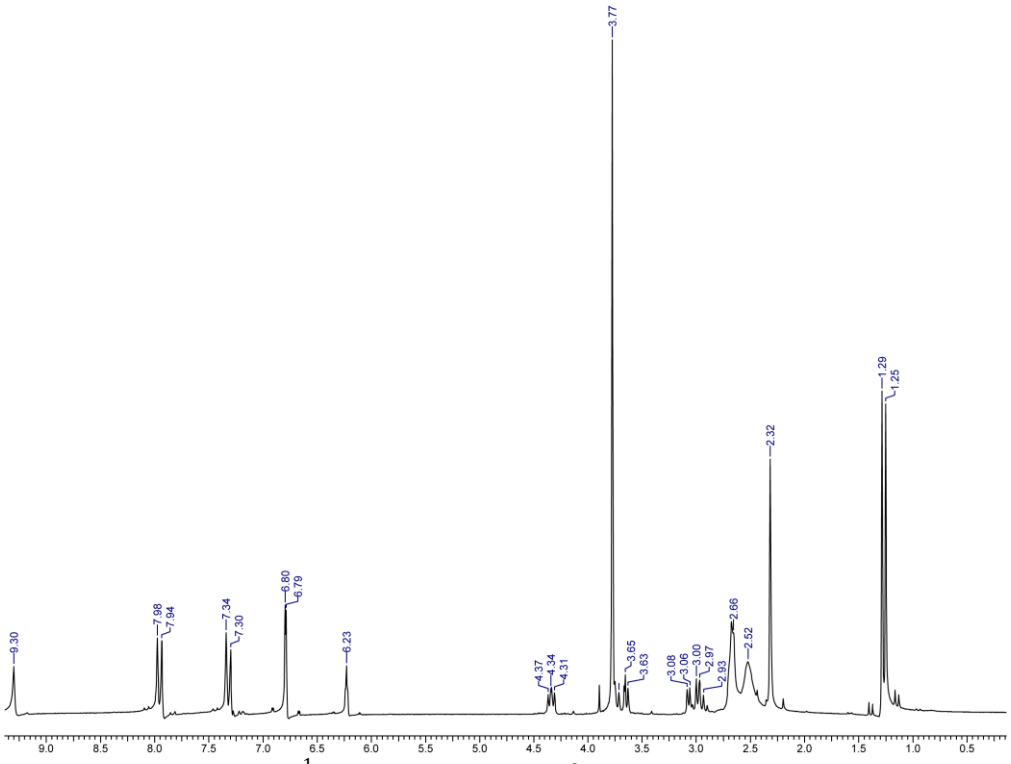
Table S6. Statistics of PCA model

Component	SSX	SSX_{acc}	VarX	$VarX_{acc}$
1	24.47	24.47	21.04	21.04
2	16.33	40.8	14.15	35.19
3	9.80	50.60	8.05	43.24
4	7.75	58.35	6.42	49.66
5	5.33	63.68	4.05	53.71

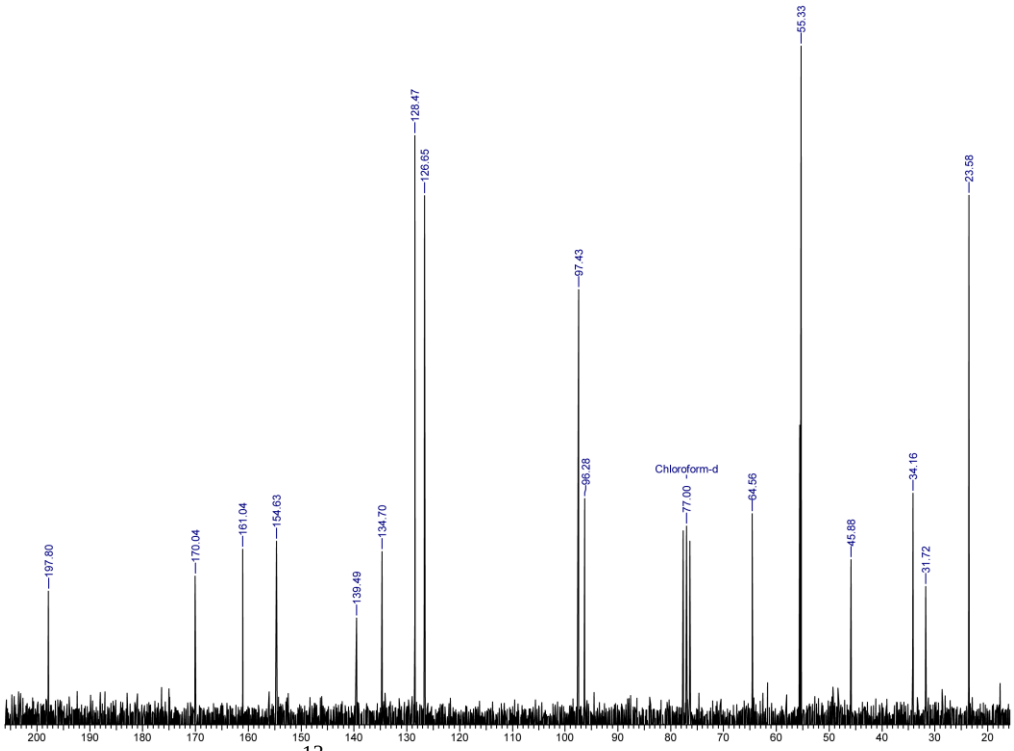
Abbreviations: SSX – percentage of the X sum of the squares; VarX – percentage of the X variance. The 'acc' states cumulative value.

Table S7. RMSD of the atomic positions between docked poses of *R* and *S* enantiomers, for the energetically best ranked (E), and the most populated (P) pose of compounds **6**, **17** and **18**.

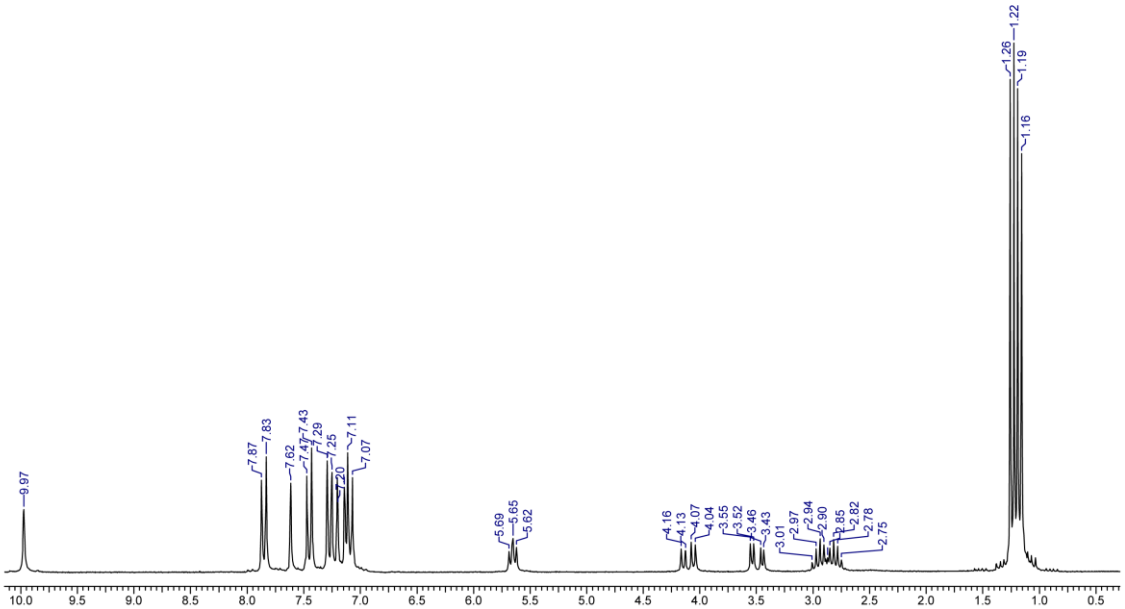
Compound Nº	RMSD (Å)	
	H atoms included	heavy atoms only
6 (E, P)	1.811	1.810
17 (E)	2.350	1.480
17 (P)	2.077	2.077
18 (E)	1.874	1.220
18 (P)	2.312	1.440



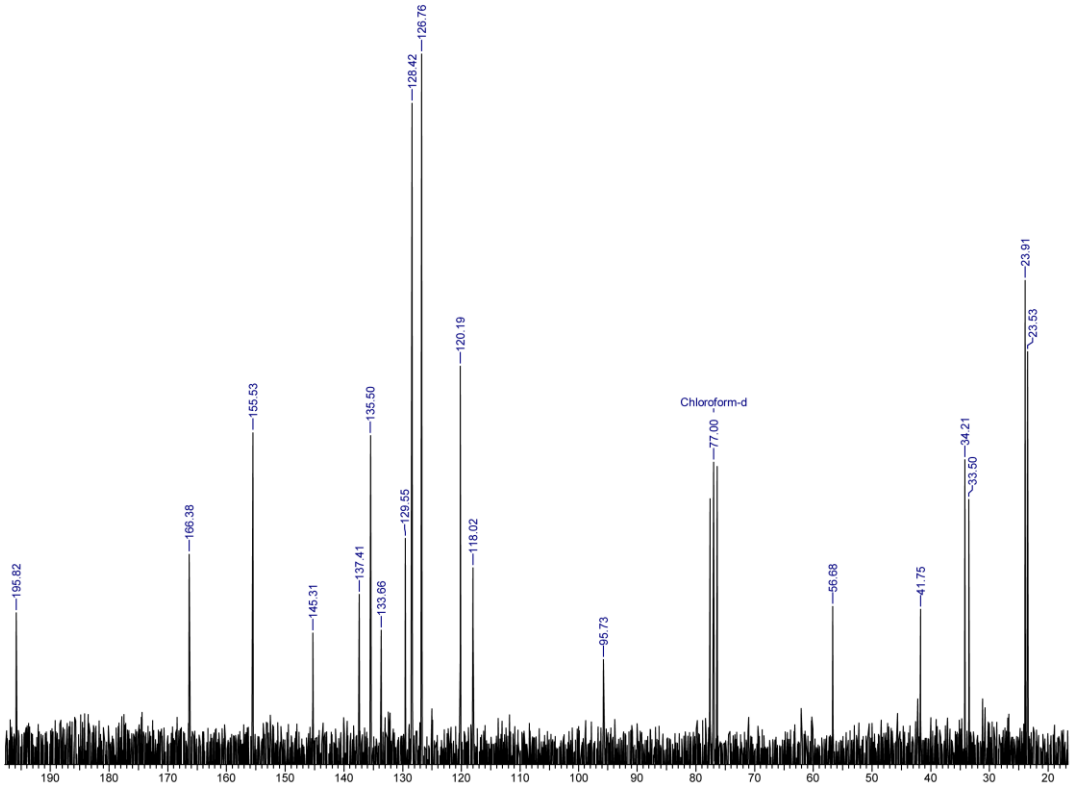
a) ^{1}H NMR spectrum of compound **4**



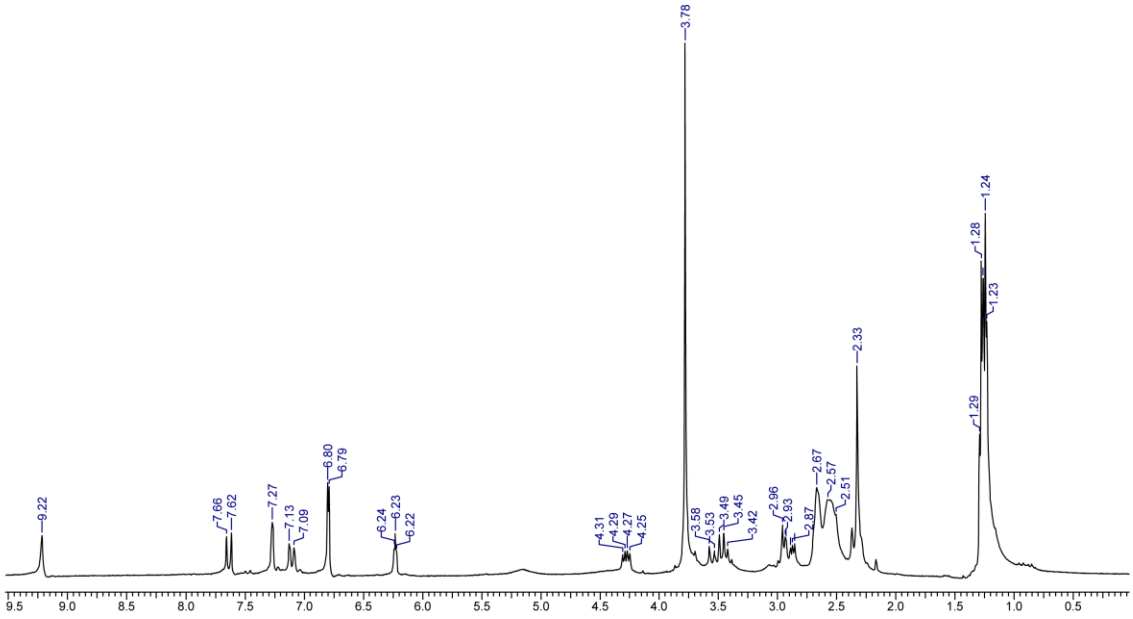
b) ^{13}C NMR spectrum of compound **4**



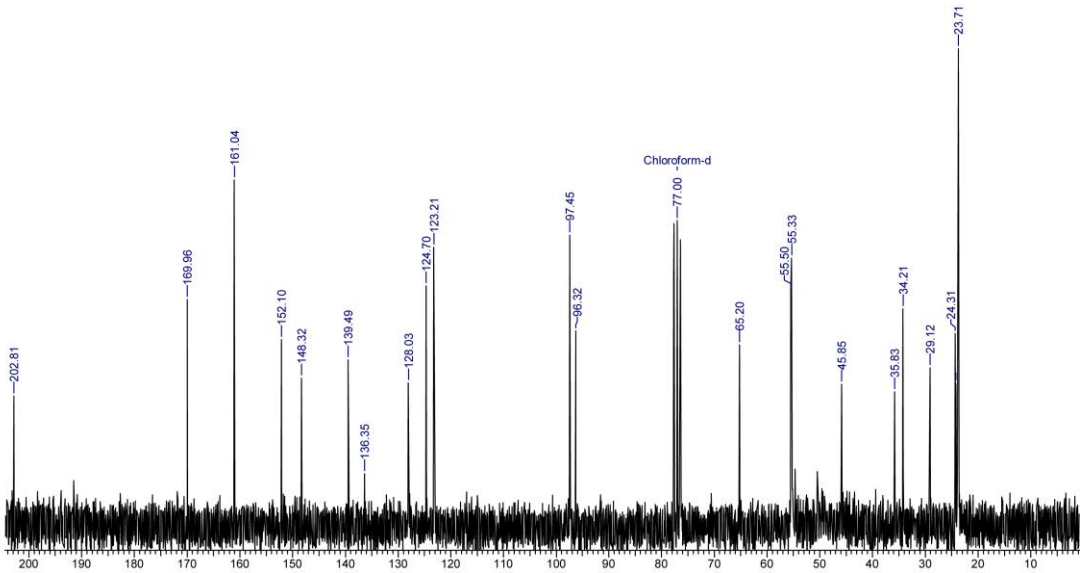
c) ^{1}H NMR spectrum of compound **6**



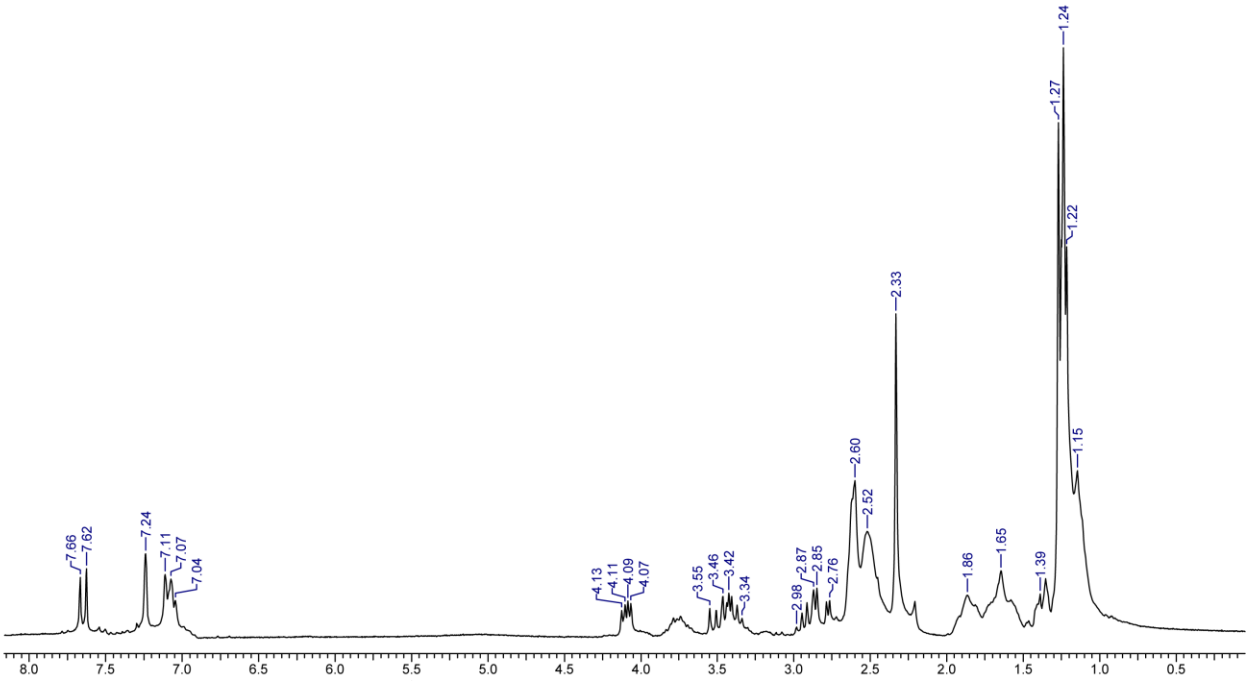
d) ^{13}C NMR spectrum of compound **6**



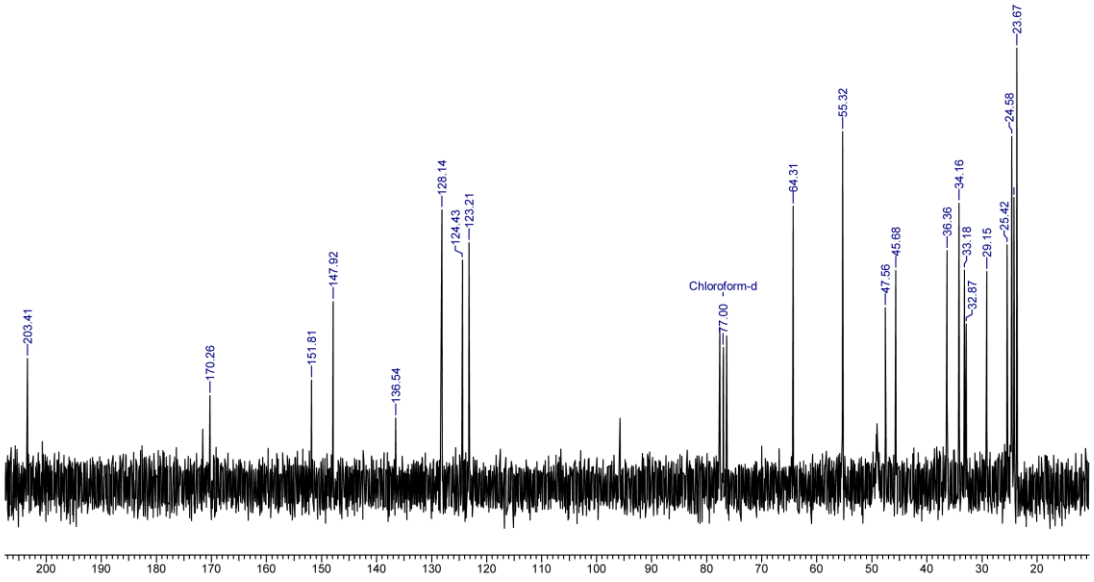
e) ^{1}H NMR spectrum of compound **16**



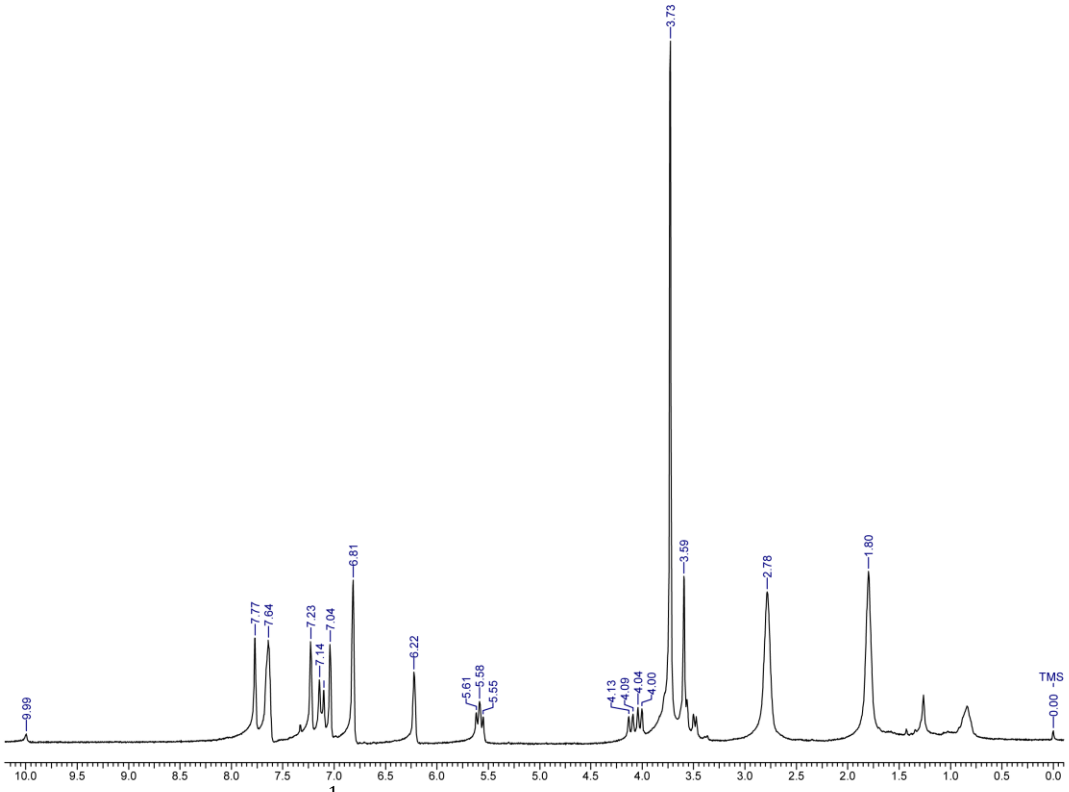
f) ^{13}C NMR spectrum of compound **16**



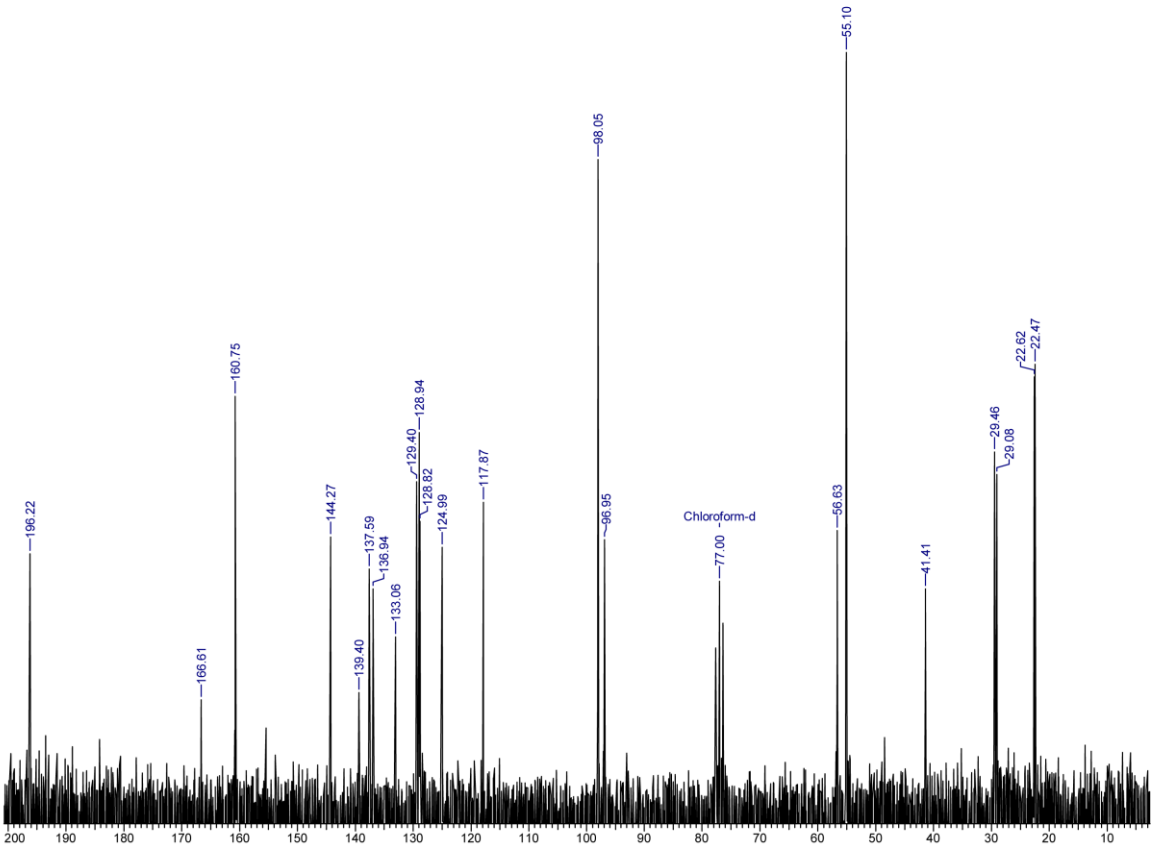
g) ^{1}H NMR spectrum of compound **24**



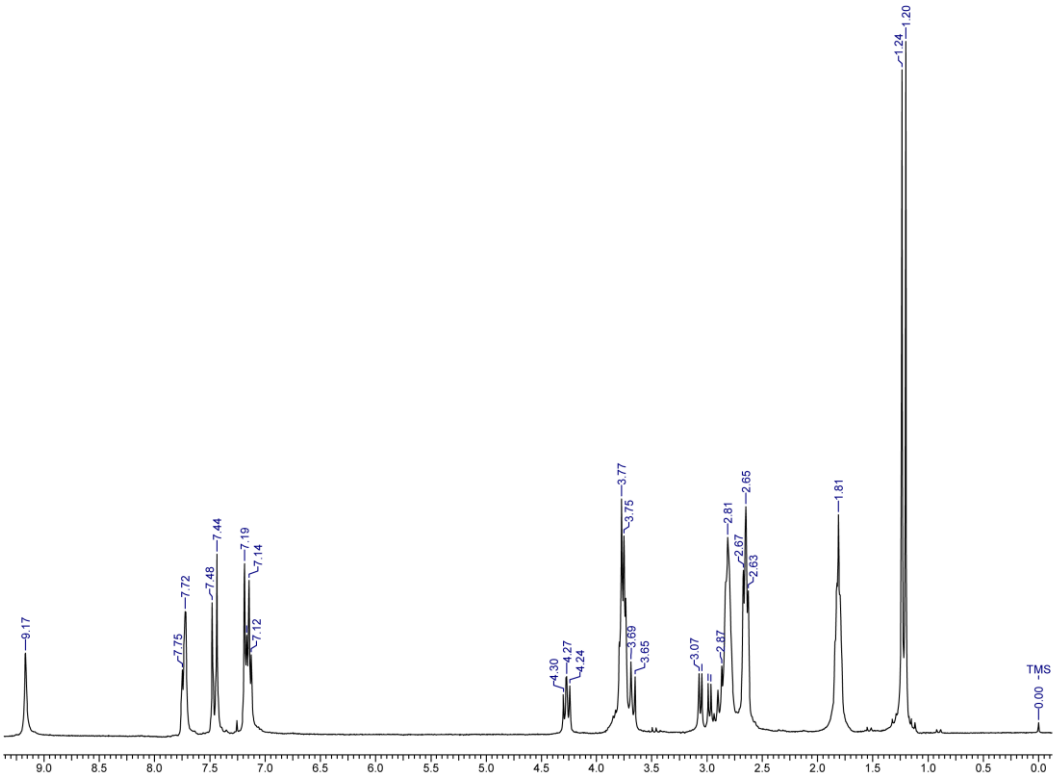
h) ^{13}C NMR spectrum of compound **24**



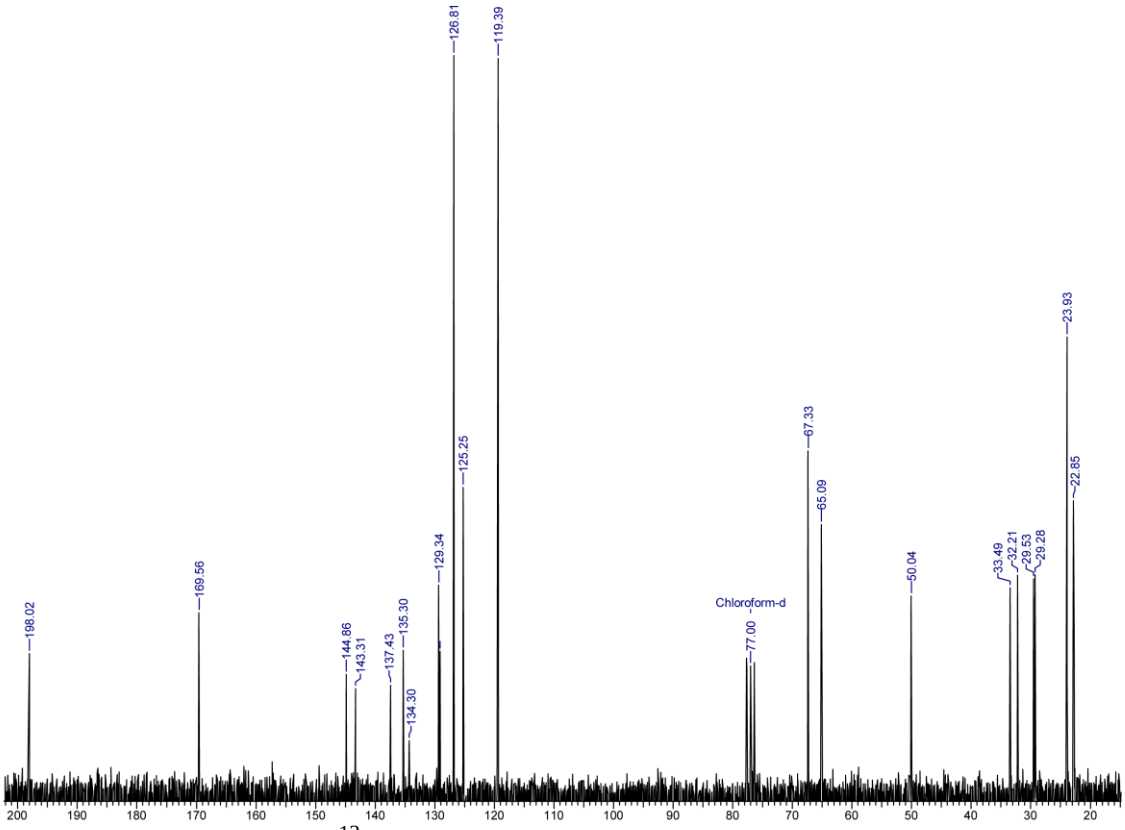
i) ^{1}H NMR spectrum of compound **26**



j) ^{13}C NMR spectrum of compound **26**



k) ^{1}H NMR spectrum of compound **31**



l) ^{13}C NMR spectrum of compound **31**

Figure S7. NMR spectra of representative compounds.