SUPPLEMENTARY DATA

Anti-Alzheimer Potential of a New (+)-Pinitol Glycoside Isolated from *Tamarindus indica* Pulp: *In Vivo* and *In Silico* Evaluations

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Table S1. The information of 82 targets related to Alzheimer's disease.

No.	Target	Full name
1	APP	Beta amyloid A4 (resveratrol and ferulic acid and Chrysin)
2	MAOA	Monoamine oxidase A (Resveratrol and chrysin)
3	MAOB	Monoamine oxidase B (Ferulic acid and chrysin)
4	ACHE	Acetylcholinesterase
5	BCHE	Butyrylcholinesterase
6	PIK3CB	PI3-kinase p110-beta subunit
7	PIK3CA	PI3-kinase p110-alpha subunit
8	BACE1	Beta-Secretase 1
9	BACE2	Beta secretase 2
10	HTR7	Serotonin 7 (5-HT7) receptor
11	HTR6	Serotonin 6 (5-HT6) receptor
12	MAPT	Microtubule-associated protein tau
13	CDK5	Cyclin-dependent kinase 5/CDK5 activator 1
14	PTGS2	Cyclooxygenase-2 (and resveratrol)
15	PSEN1	Presenilin 1
16	PSEN2	Presenilin 2
17	APOE	Apolipoprotein E
18	ABCA7	ATP Binding Cassette Subfamily A Member 7

19	SNCA	Synuclein Alpha
20	SORL1	Sortilin Related Receptor 1
21	ADAM10	ADAM Metallopeptidase Domain 10
22	A2M	Alpha-2-Macroglobulin
23	NOS3	Nitric Oxide Synthase 3
24	PRNP	Prion Protein
25	TREM2	Triggering Receptor Expressed On Myeloid Cells 2
26	MT-ND1	Mitochondrially Encoded NADH:Ubiquinone Oxidoreductase Core Subunit 1
27	HFE	Homeostatic Iron Regulator
28	PLAU	Plasminogen Activator, Urokinase
29	SNCB	Synuclein Beta
30	GRN	Granulin Precursor
31	MT-ND2	Mitochondrially Encoded NADH:Ubiquinone Oxidoreductase Core Subunit 2
32	UNC5C	Unc-5 Netrin Receptor C
33	TOMM40	Translocase Of Outer Mitochondrial Membrane 40
34	MPO	Myeloperoxidase
35	GSK3B	Glycogen Synthase Kinase 3 Beta
36	COMT	Catechol-O-Methyltransferase
37	PRKN	Parkin RBR E3 Ubiquitin Protein Ligase
38	CLU	Clusterin
39	HTR2A	5-Hydroxytryptamine Receptor 2A
40	LRP1	LDL Receptor Related Protein 1
41	Serpin Family A Member 3 MIR29A	Serpin Family A Member 3 MicroRNA 29a
42	DRD3	Dopamine Receptor D3
43	MIR29B1	MicroRNA 29b-1
44	IDE	Insulin Degrading Enzyme
45	MIR107	MicroRNA 107
46	MIR146A	MicroRNA 146a
47	CHAT	Choline O-Acetyltransferase
48	CTSD	Cathepsin D

49	TNF	Tumor Necrosis Factor
50	APBB1	Amyloid Beta Precursor Protein Binding Family B Member 1
51	BDNF	Brain Derived Neurotrophic Factor
52	NCSTN	Nicastrin
53	PICALM	Phosphatidylinositol Binding Clathrin Assembly Protein
54	HSD17B10	Hydroxysteroid 17-Beta Dehydrogenase 10
55	CDK5R1	Cyclin Dependent Kinase 5 Regulatory Subunit 1
56	COL25A1	Collagen Type XXV Alpha 1 Chain
57	ACE	Angiotensin I Converting Enzyme
58	PLD3	Phospholipase D Family Member 3
59	PSENEN	Presenilin Enhancer, Gamma-Secretase Subunit
60	CASP3	Caspase 3
61	CR1	Complement C3b/C4b Receptor 1
62	IL1B	Interleukin 1 Beta
63	MME	Membrane Metalloendopeptidase
64	CALHM1	Calcium Homeostasis Modulator 1
65	IL1A	Interleukin 1 Alpha
66	TF	Transferrin
67	GAPDH	Glyceraldehyde-3-Phosphate Dehydrogenase
68	SOD1	Superoxide Dismutase 1
69	IL6	Interleukin 6
70	PTGS2	Prostaglandin-Endoperoxide Synthase 2
71	VLDLR	Very Low Density Lipoprotein Receptor
72	MAPK1	Mitogen-Activated Protein Kinase 1
73	DHCR24	24-Dehydrocholesterol Reductase
74	CAPN1	Calpain 1
75	VCP	Valosin Containing Protein

76	APLP2	Amyloid Beta Precursor Like Protein 2
77	CHRNA7	Cholinergic Receptor Nicotinic Alpha 7 Subunit
78	NGF	Nerve Growth Factor
79	CLSTN1	Calsyntenin 1
80	OLR1	Oxidized Low Density Lipoprotein Receptor 1
81	INS	Insulin
82	NOS1	Nitric Oxide Synthase 1

Table S2. Proteins predicted to be potential targets for compound1

No.	Target	Full name	Uniport ID	PharmMapper (Fit score score)	Vina score kcal/mol	$\Delta G_{binding}$ kcal/mol
1	APP	Beta amyloid A4	P05067	8.3	-4.9	-4.1
2	MAOA	Monoamine oxidase A	P21397	8.4	-5.5	-4.8
3	PIK3CB	PI3-kinase p110-beta subunit	P42338	8.5	-7.1	-5.4
4	PIK3CA	PI3-kinase p110-alpha subunit	P42336	8.3	-4.2	-4.5
5	MAOB	Monoamine oxidase B	P27338	8.2	-7.6	-5.1
6	**BACE1***	**Beta secretase 1**	**P56817**	**8.7**	**-8.1**	**-7.8**
7	HTR7	Serotonin 7 (5-HT7) receptor	P34969	8.1	-5.3	-4.2
8	HTR6	Serotonin 6 (5-HT6) receptor	P50406	7.6	-5.2	-5.8
9	**ACHE***	**Acetylcholinesterase**	**P22303**	**7.4**	**-7.7**	**-7.1**
10	BCHE	Butyrylcholinesterase	P06276	8.5	-6.3	-5.6
11	MAPT	Microtubule-associated protein tau	P10636	7.2	-6.7	-5.1

*The best scoring protein targets

Molecular Docking

Docking experiments were performed using Auto Dock Vina software according the previously reported protocol[1]. The binding site in each docked protein was determined according to the co-crystalized ligand. To account for these binding sites' flexibility, we used their MDS-derived conformers sampled every 10 ns for docking experiments (i.e., ensemble docking). Subsequently, the retrieved top hits were ranked

according to their binding energies. The generated docking poses were visualized and analyzed using Pymol softwar [2].

Molecular Dynamic Simulation

MD simulations were performed by Desmond v. 2.2 [3] the MDS machine of Maestro software [4] using the OPLS3 forcefield. The protein-ligand systems were built via System Builder option, where it was embedded in an orthorhombic box of TIP3P waters together with 0.15 M Na^+ and Cl^- ions with 20 A° solvent buffer from the molecular surface of the centrally placedreceptor. Afterwards, the prepared system was energy minimized and equilibrated for 10 ns. Desmond software automatically parameterizes inputted ligands during the system building step according OPLS force field. For simulations performed by NAMD, the parameters and topologies of the compounds were calculated either using Charmm27 force field by the online software Ligand Reader & Modeler (http://www.charmm-gui.org/?doc=input/ligandrm) [5] or by the VMD plugin Force Field Toolkit (ffTK) [6]. Afterward, the generated parameters and topology files were loaded to VMD so that it can readily read the protein-ligand complexes without errors and then conduct the simulation step. Simulations were run for 50 ns at 310 K in the NPT ensemble with the Nose-Hoover thermostat and Martyna-Tobias-Klein barostat using an anisotropic coupling. We used the best binding poses for each compound-protein complex as starting systems to investigate their binding stability and mode of interactions.

Binding free energy calculations (ΔG) were performed using the free energy perturbation (FEP) method. We first prepared the input files and script NAMD by the online-based software CHARMM-GUI Free Energy Calculator (http://www.charmm-gui.org/input/fec). Afterwards, these inputs were loaded to NAMD for simulations, where the equilibration was performed in the NPT ensemble at 300 K and 1 atm (1.01325 bar) with Langevin piston pressure (for "Complex" and "Ligand") in the presence of TIP3P water model. 10 ns FEP simulations were performed fro each compound, and the last 5 ns of the free enegry values was measured for the final free energy values [7].

Networks Construction

We constructed two networks (Figures2 and 3): (i) compound-protein interaction (CPI) network depending on the results of prediction. In this network we constructed connections between the metabolites that showed drug-like properties and predicted to pass the BBB, and target proteins relevant to AD; (ii) protein-protein interaction (PPI) network that showed the interactions between proteins relevant to AD. The proteins that were found to be potential targets for the select metabolites were submitted to the STRING database (https://string-db.org/) [8] for protein–protein interaction (PPI) analysis, and "Homosapiens" was selected as the search species., the lowest interaction score was set to 0.4, and the rest of the parameters were set to the default setting to obtain the PPI network .All of the above networks were constructed and summarized in two figures (Figure 2 and 3) using Cytoscape 3.8.2 (https://www.cytoscape.org/) [9], which is a software package for visualizing and analyzing networks.

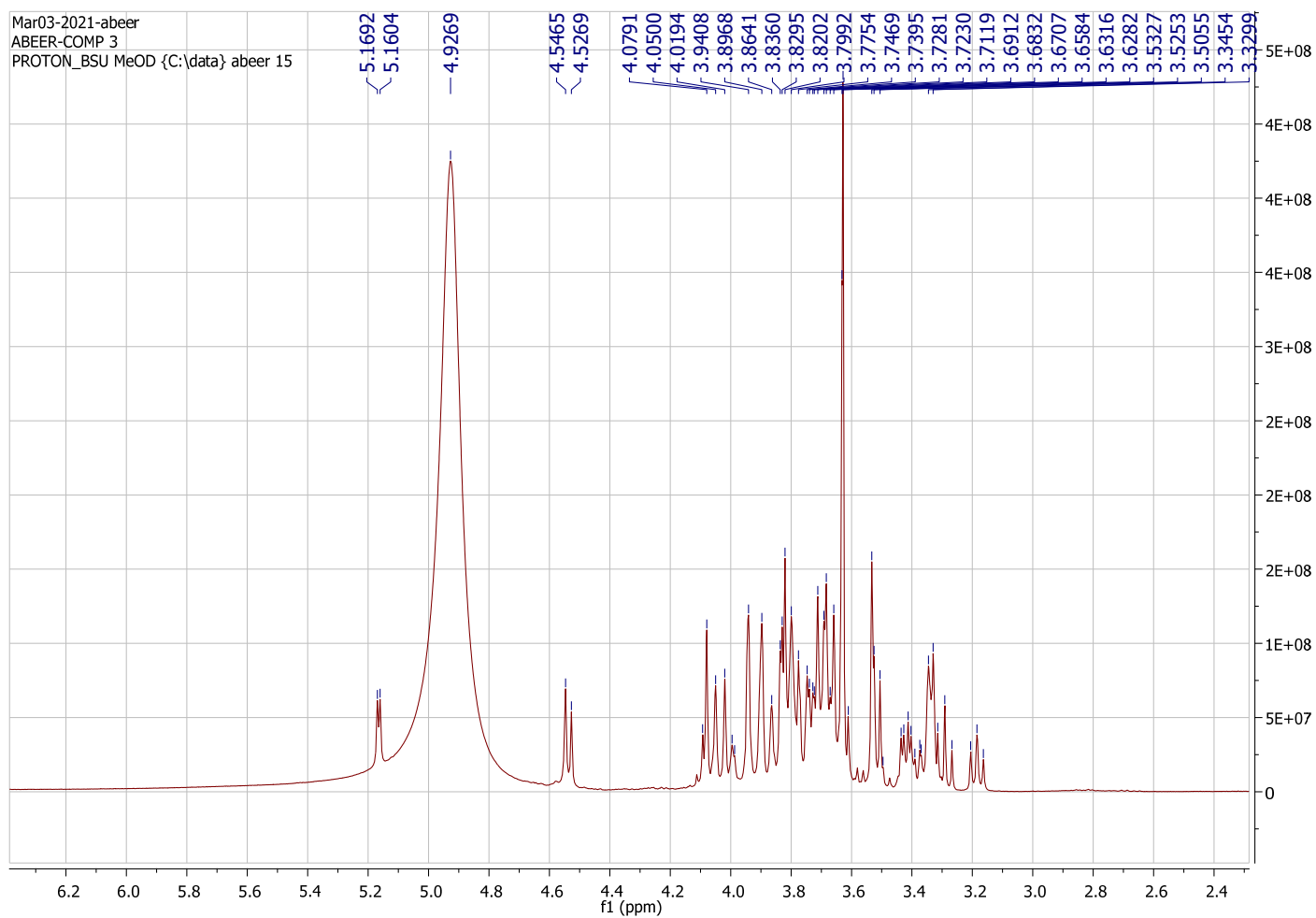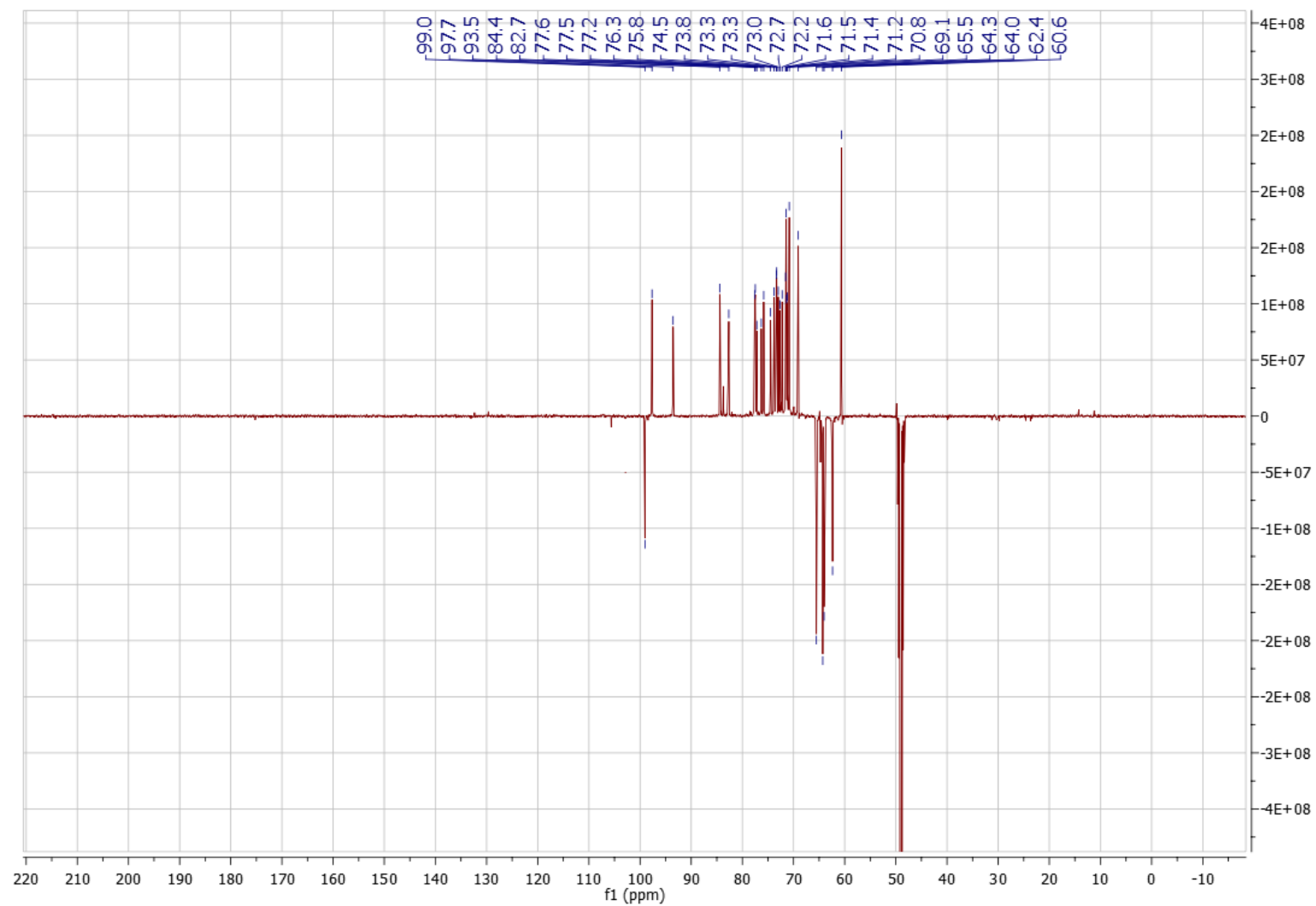**Figure S1.** 1H NMR spectrum of compound **1** measured in CD₃OD-d_4 at 400 MHz

Figure S2. DEPT-Q NMR spectrum of compound **1** measured in CD₃OD-*d₄* at 100 MHz

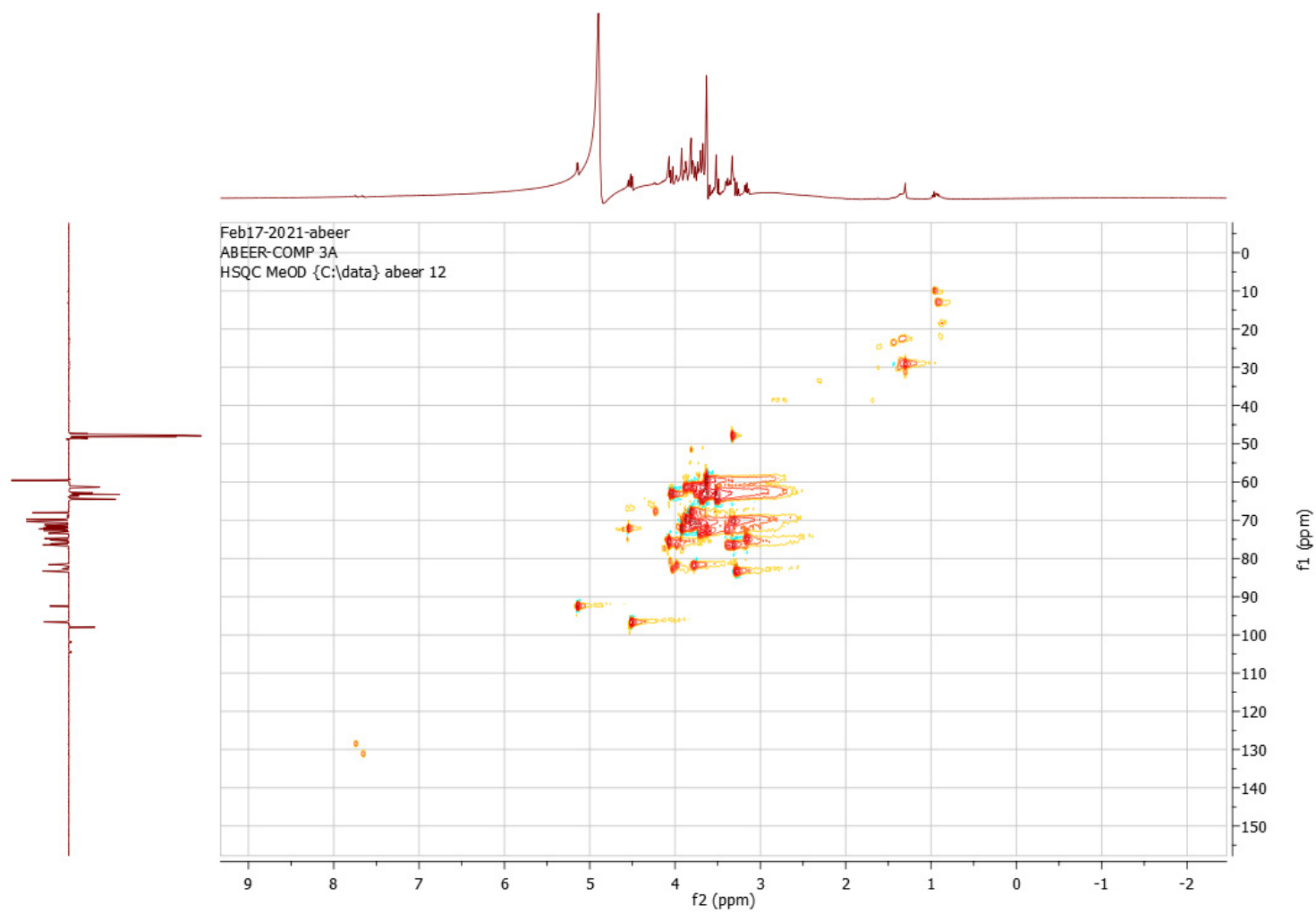**Figure S3.** HSQC spectrum of compound **1** measured in CD₃OD-d_4

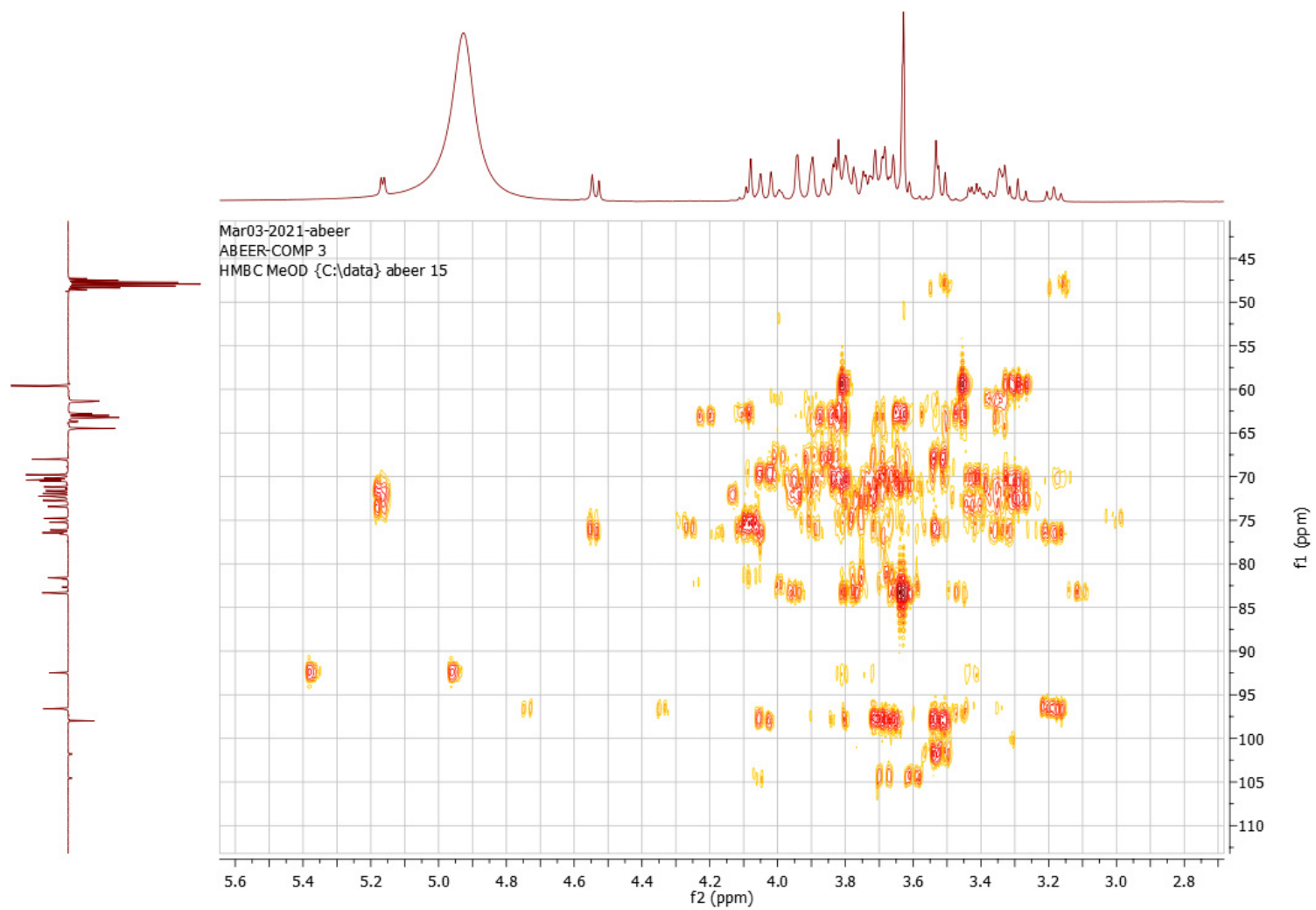**Figure S4.** HMBC spectrum of compound **1** measured in CD₃OD-*d₄*

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