Supporting Information

Discovery of novel STAT3 inhibitors with anti-breast cancer activity: structure-based virtual screening, molecular dynamics and biological evaluation

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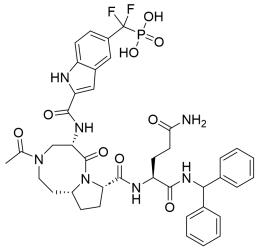
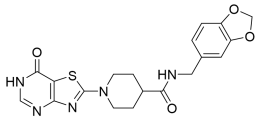
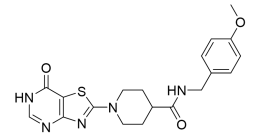
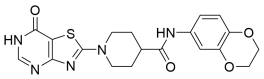
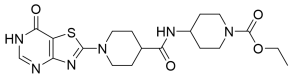
Table S1 Structures in G1 with their virtual screening results and BPMD values.

No.	Structures	Smiles	Docking score	Glide gscore	MM/GBSA ΔG_{bind} (kcal/mol)	PersScore	PoseScore	CompScore
SI-109		FC(F)(P(O)(O)=O)C1=CC=C(NC(C(N[C@H]2CN(C(C)=O)CC[C@@H](CC[C@H]3C(N[C@H](C(NC(C4=CC=CC=C4)C5=CC=CC=C5)=O)CCC(N)=O)=O)N3C2=O)=O)=C6)C6=C1	-9.764	-9.776	-99.65	0.827	1.896	-2.237
a1		O=C1C2=C(N=C(N3CCC(C(NCC4=CC(OCO5)=C5C=C4)=O)CC3)S2)N=CN1	-8.231	-8.941	-68.42	0.356	3.889	2.107
a2		O=C1C2=C(N=C(N3CCC(C(NCC4=CC=C(OC)C=C4)=O)CC3)S2)N=CN1	-8.999	-9.710	-65.13	0.452	3.926	1.665
a3		O=C1C2=C(N=C(N3CCC(C(NC4=CC(OCCO5)=C5C=C4)=O)CC3)S2)N=CN1	-8.834	-9.544	-63.09	0.255	3.028	1.755
a4		O=C1C2=C(N=C(N3CCC(C(NC4CCN(C(OCC)=O)CC4)=O)CC3)S2)N=CN1	-7.732	-8.443	-61.57	0.383	3.892	1.976

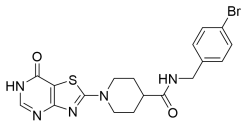
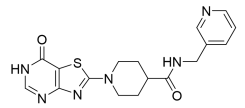
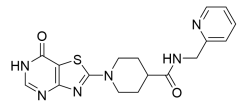
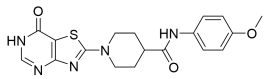
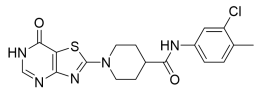
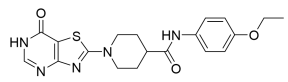
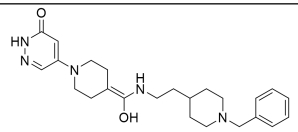
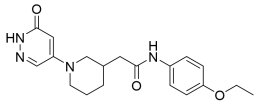
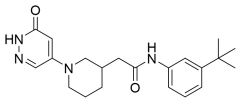
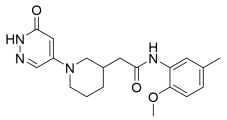
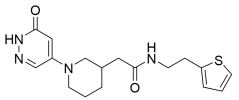
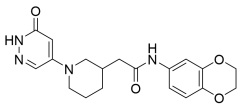
a5		BrC1=CC=C(CNC(C2CCN(C(S3)=NC4=C3C(NC=N4)=O)CC2)=O)C=C1	-8.131	-8.840	-61.37	0.545	3.348	0.621
a6		O=C1C2=C(N=C(N3CCC(C(NCC4=CC=CN=C4)=O)CC3)S2)N=CN1	-8.856	-9.575	-61.22	0.465	2.950	0.624
a7		O=C1C2=C(N=C(N3CCC(C(NCC4=CC=CC=N4)=O)CC3)S2)N=CN1	-8.445	-9.168	-61.12	0.320	2.475	0.876
a8		O=C1C2=C(N=C(N3CCC(C(NC4=CC=C(OC)C=C4)=O)CC3)S2)N=CN1	-8.620	-9.330	-60.85	0.361	2.864	1.058
a9		CC1=CC=C(NC(C2CCN(C(S3)=NC4=C3C(NC=N4)=O)CC2)=O)C=C1Cl	-7.934	-8.644	-60.08	0.400	2.955	0.955
a10		O=C1C2=C(N=C(N3CCC(C(NC4=CC=C(OCC)C=C4)=O)CC3)S2)N=CN1	-8.132	-8.842	-59.94	0.371	2.623	0.766

Table S2 Structures in G2 with their virtual screening results and BPMD values.

No.	Structures	Smiles	Docking score	Glide gscore	MM/GBSA ΔG_{bind} (kcal/mol)	PersScore	PoseScore	CompScore
b1		O=C1NN=CC(N2CC/C(CC2)=C(NCCC3CCN(CC4=CC=CC=C4)CC3)\O)=C1	-7.340	-8.396	-65.57	0.436	4.263	2.081
b2		O=C1NN=CC(N2CCCC(CC(NC3=CC=C(OCC)C=C3)=O)C2)=C1	-9.007	-9.075	-63.00	0.229	5.965	4.822
b3		O=C1NN=CC(N2CCCC(CC(NC3=CC=CC(C(C)(C)C)=C3)=O)C2)=C1	-8.160	-8.230	-61.47	0.315	4.035	2.459
b4		O=C1NN=CC(N2CCCC(CC(NC3=CC(C)=CC=C3OC)=O)C2)=C1	-8.570	-8.640	-59.52	0.358	2.962	1.171
b5		O=C1NN=CC(N2CCCC(CC(NCCC3=CC=CS3)=O)C2)=C1	-8.181	-8.247	-59.09	0.323	4.099	2.485
b6		O=C1NN=CC(N2CCCC(CC(NC3=CC4=C(OCCO4)C=C3)=O)C2)=C1	-9.447	-9.517	-59.01	0.144	6.692	5.972

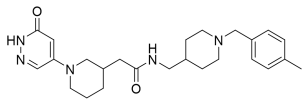
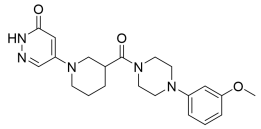
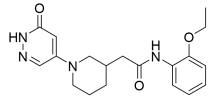
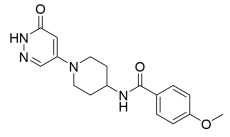
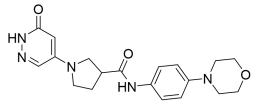
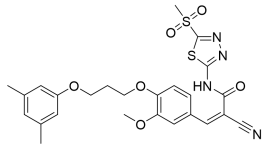
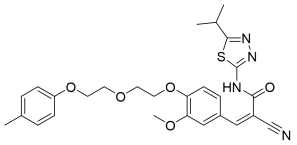
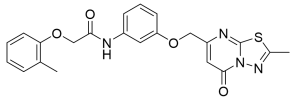
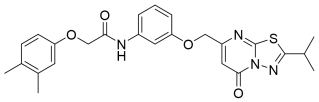
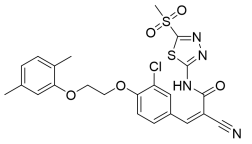
b7		O=C1NN=CC(N2CCCC(CC(NCC3CCN(CC4=CC=C(C)C=C4)CC3)=O)C2)=C1	-7.289	-7.355	-58.95	0.239	6.179	4.982
b8		O=C1NN=CC(N2CCCC(C(N3CCN(C4=CC=CC(OC)=C4)CC3)=O)C2)=C1	-9.436	-9.503	-58.77	0.226	4.962	3.834
b9		O=C1NN=CC(N2CCCC(CC(NC3=CC=CC=C3OCC)=O)C2)=C1	-7.793	-7.863	-58.61	0.236	2.761	1.580
b10		O=C1NN=CC(N2CCC(NC(C3=CC=C(OC)C=C3)=O)CC2)=C1	-7.596	-7.663	-58.23	0.171	3.855	2.998
b11		O=C1NN=CC(N2CCC(C(NC3=CC=C(N4CCOCC4)C=C3)=O)C2)=C1	-8.515	-8.581	-58.16	0.379	3.430	1.536

Table S3 Structures in G3 with their virtual screening results and BPMD values.

No.	Structures	Smiles	Docking score	Glide gscore	MM/GBSA ΔG_{bind} (kcal/mol)	PersScore	PoseScore	CompScore
c1		CC1=CC(C)=CC(OCCCOC2=C(OC)C=C(/C=C(C#N)\C(NC3=NN=C(S(=O)(C)=O)S3)=O)C=C2)=C1	-7.355	-7.355	-61.90	0.128	4.214	3.572
c2		COC(C=C(/C=C(C#N)\C(NC1=NN=C(C(C)C)S1)=O)C=C2)=C2OCCOCCOC3=CC=C(C)C=C3	-7.069	-7.072	-58.97	0.029	4.780	4.637
c3		CC(S1)=NN2C1=NC(COC3=CC(NC(COC4=C(C)C=CC=C4)=O)=CC=C3)=CC2=O	-6.986	-6.986	-58.84	0.090	3.574	3.126
c4		O=C(NC1=CC=CC(OCC(N=C2N3N=C(C(C)C)S2)=CC3=O)=C1)COC4=CC(C)=C(C)C=C4	-6.674	-6.674	-58.32	0.099	4.923	4.429
c5		CC1=CC=C(C)C(OCCOC2=CC=C(/C=C(C#N)\C(NC3=NN=C(S(=O)(C)=O)S3)=O)C=C2Cl)=C1	-6.383	-6.383	-58.05	0.082	4.911	4.502

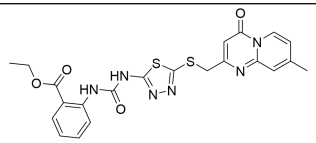
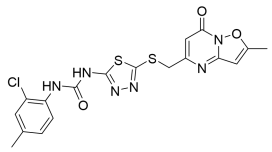
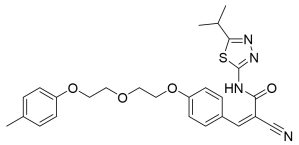
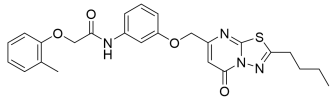
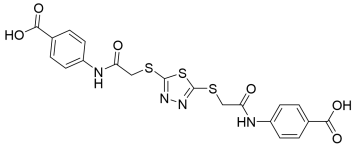
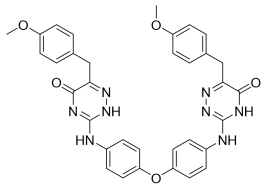
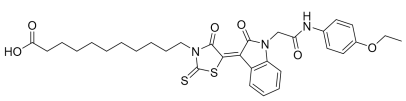
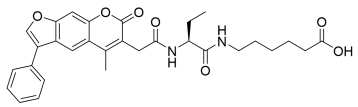
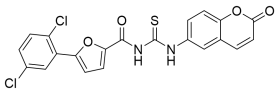
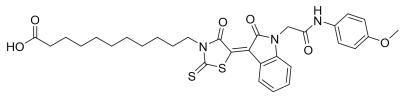
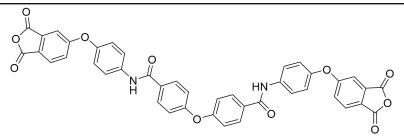
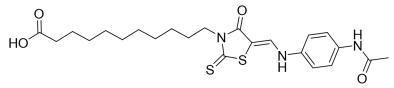
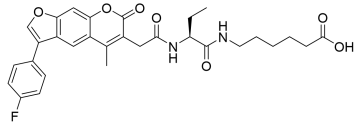
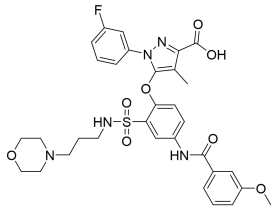
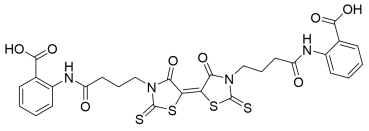
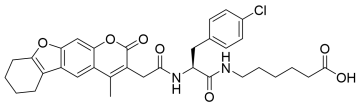
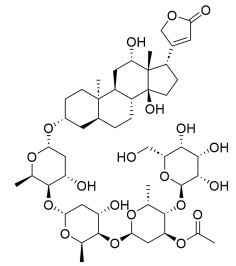
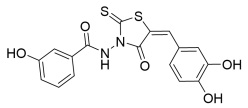
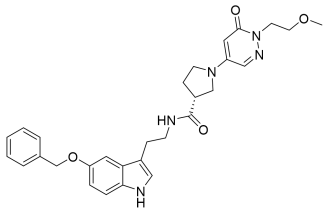
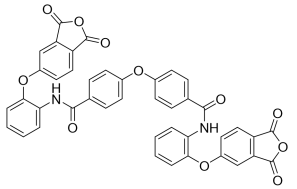
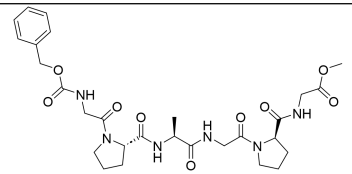
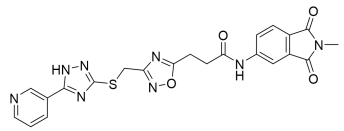
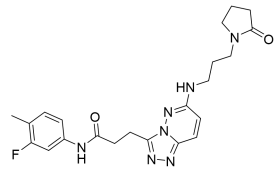
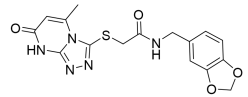
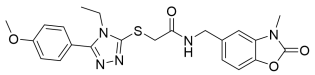
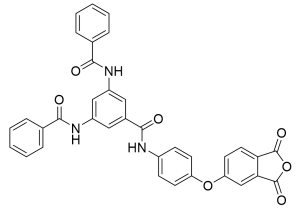
c6		O=C(OCC)C1=C(NC(NC2=NN=C(SCC(N=C3N4C=CC(C)=C3)=CC4=O)S2)=O)C=CC=C1	-6.528	-6.528	-57.99	0.115	4.367	3.791
c7		ClC1=C(NC(NC2=NN=C(SCC(N=C3N4OC(C)=C3)=CC4=O)S2)=O)C=CC(C)=C1	-6.408	-6.408	-57.96	0.109	4.688	4.143
c8		CC(C)C(S1)=NN=C1NC(/C(C#N)=C\C(C=C2)=CC=C2OCCOCCOC3=CC=C(C)C=C3)=O	-6.330	-6.333	-56.99	0.057	4.301	4.015
c9		CC1=C(OCC(NC2=CC=CC(OCC(N=C3N4N=C(CCCC)S3)=CC4=O)=C2)=O)C=CC=C1	-6.279	-6.279	-56.88	0.220	4.016	2.916
c10		O=C(CSC1=NN=C(SCC(NC2=CC=C(C(O)=O)C=C2)=O)S1)NC3=CC=C(C(O)=O)C=C3	-6.684	-6.684	-56.41	0.343	4.195	2.480

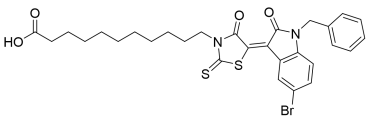
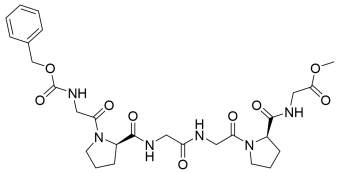
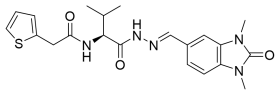
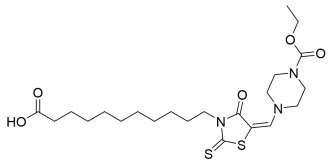
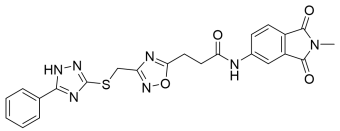
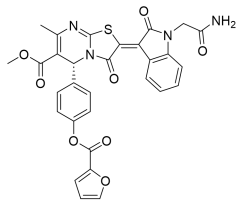
Table S4 Structures in G4 with their virtual screening results and BPMD values.

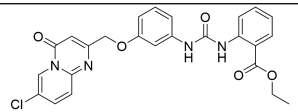
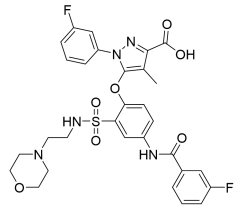
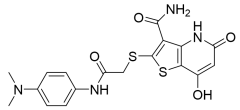
No.	Structures	Smiles	Docking score	Glide gscore	MM/GBSA ΔG_{bind} (kcal/mol)	PersScore	PoseScore	CompScore
d1		COC1=CC=C(CC2=NNC(NC3=CC=C(OC4=CC=C(NC(N5)=NN=C(CC6=CC=C(OC)C=C6)C5=O)C=C4)C=C3)=NC2=O)C=C1	-6.662	-7.259	-75.71	0.277	3.283	1.900
d2		O=C(N(CCCCCCCCCCC(O)=O)C(S/1)=S)C1=C(C(C=CC=C2)=C2N3CC(NC4=CC=C(OCC)C=C4)=O)/C3=O	-6.862	-6.867	-71.79	0.627	2.691	-0.446
d3		CC(C1=CC2=C(OC=C2C3=CC=CC=C3)C=C1O4)=C(CC(N[C@H](C(NCCCCCC(O)=O)=O)CC)=O)C4=O	-6.703	-6.707	-70.03	0.251	1.794	0.539
d4		ClC1=C(C2=CC=C(C(NC(NC3=CC=C4C(C=CC(O4)=O)=C3)=S)=O)O2)C=C(Cl)C=C1	-6.234	-6.441	-68.17	0.068	3.691	3.350
d5		O=C(N(CCCCCCCCCCC(O)=O)C(S/1)=S)C1=C(C(C=CC=C2)=C2N3CC(NC4=CC=C(OC)C=C4)=O)/C3=O	-6.849	-6.854	-68.08	0.698	2.426	-1.067

d6		O=C1C2=C(C=CC(OC3=CC=C(NC(C4=CC=C(OC5=CC=C(C(NC6=CC=C(OC7=CC=C(C(OC8=O)=O)C8=C7)C=C6)=O)C=C5)C=C4)=O)C=C3)=C2)C(O1)=O	-7.350	-7.350	-67.21	0.048	4.215	3.975
d7		O=C(N(CCCCCCCCCCC(O)=O)C(S/1)=S)C1=C\NC2=CC=C(NC(C)=O)C=C2	-6.494	-6.909	-65.25	0.448	2.636	0.395
d8		CC(C1=CC2=C(OC=C2C3=CC=C(F)C=C3)C=C1O4)=C(CC(N[C@H](C(NCCCCCC(O)=O)=O)CC)=O)C4=O	-6.451	-6.456	-64.96	0.364	2.105	0.287
d9		O=S(NCCCN1CCOCC1)(C2=CC(NC(C3=CC=CC(OC)=C3)=O)=CC=C2OC4=C(C)C(C(O)=O)=NN4C5=CC(F)=CC=C5)=O	-7.876	-8.255	-62.84	0.502	4.440	1.928
d10		OC(C1=C(NC(CCCN2C(S/C(C2=O)=C3SC(N(CCCC(NC4=CC=CC=C4C(O)=O)=O)C\3=O)=S)=S)=O)C=CC=C1)=O	-7.009	-7.010	-62.51	0.562	3.016	0.203
d11		CC(C1=CC2=C(OC3=C2CCCC3)C=C1O4)=C(CC(N[C@H](C(NCCCCCC(O)=O)=O)CC5=CC=C(Cl)C=C5)=O)C4=O	-6.544	-6.549	-62.40	0.235	3.029	1.853

d12		C[C@]12[C@@H](CC[C@@H]3[C@@H]2C[C@H](O)[C@@]4(C)[C@]3(O)CC[C@H]4C5=CC(OC5)=O)C[C@H](O[C@@H]6C[C@H](O)[C@@H](O[C@H]7O[C@H](C)[C@H](O[C@H]8O[C@H](C)[C@H](O[C@H]9O[C@H](CO)[C@H](O)[C@H](O)[C@@H]9O)[C@@H](OC(C)=O)C8)[C@@H](O)C7)[C@@H](C)O6)CC1	-8.095	-8.095	-62.12	0.031	6.349	6.192
d13		O=C(NN(C(/C(S1)=C\C2=CC(O)=C(O)C=C2)=O)C1=S)C3=CC(O)=CC=C3	-6.571	-6.985	-61.69	0.222	2.054	0.945
d14		O=C([C@@H]1CCN(C(C=NN2CCOC)=CC2=O)C1)NCCC3=CNC4=CC=C(OCC5=CC=CC=C5)C=C43	-6.601	-6.601	-61.67	0.057	5.907	5.622
d15		O=C1C2=C(C=CC(OC3=C(NC(C4=CC=C(OC5=CC=C(C(NC6=CC=CC=C6OC7=CC=C(C(OC8=O)=O)C8=C7)=O)C=C5)C=C4)=O)C=CC=C3)=C2)C(O1)=O	-7.904	-7.904	-61.06	0.069	5.265	4.919

d16		O=C(NCC(N1CCC[C@H]1C(N[C@H](C(NCC(N2CCC[C@@H]2C(NCC(OC)=O)=O)=O)=O)C)=O)=O)OCC3=CC=CC=C3	-6.497	-6.497	-60.67	0.306	4.296	2.764
d17		O=C(NC1=CC(C(N2C)=O)=C(C=C1)C2=O)CCC3=NC(CSC4=NNC(C5=CN=CC=C5)=N4)=NO3	-7.604	-7.674	-60.21	0.195	4.385	3.411
d18		CC1=C(F)C=C(NC(CCC2=NN=C3N2N=C(NCCCN4C(CCC4)=O)C=C3)=O)C=C1	-6.652	-6.652	-60.17	0.111	4.313	3.760
d19		O=C1NC2=NN=C(SCC(NCC3=CC4=C(C=C3)OCO4)=O)N2C(C)=C1	-8.554	-8.554	-60.12	0.260	3.858	2.559
d20		COC1=CC=C(C2=NN=C(SCC(NCC3=CC=C(OC(N4C)=O)C4=C3)=O)N2CC)C=C1	-7.288	-7.288	-59.23	0.115	5.416	4.840
d21		O=C(NC1=CC(NC(C2=CC=CC=C2)=O)=CC(C(NC3=CC=C(OC4=CC=C5C(C(OC5=O)=O)=C4)C=C3)=O)=C1)C6=CC=CC=C6	-6.790	-6.790	-58.93	0.066	4.071	3.741

d22		O=C(N(CCCCCCCCCCC(O)=O)C(S/1)=S)C1=C(C(C=C(Br)C=C2)=C2N3CC4=CC=CC=C4)/C3=O	-6.756	-6.761	-58.83	0.447	4.335	2.102
d23		O=C(NCC(N1CCC[C@@H]1C(NCC(NCC(N2CCC[C@@H]2C(NCC(OC)=O)=O)=O)=O)=O)=O)OCC3=CC=CC=C3	-6.733	-6.733	-58.65	0.189	4.037	3.092
d24		O=C(N[C@H](C(N/N=C/C(C=C1)=CC2=C1N(C)C(N2C)=O)=O)C(C)C)CC3=CC=CS3	-6.813	-6.813	-58.52	0.215	2.726	1.650
d25		O=C(N(CCCCCCCCCCC(O)=O)C(S/1)=S)C1=C/N2CCN(C(OCC)=O)CC2	-6.337	-6.342	-58.35	0.369	2.588	0.744
d26		O=C(NC1=CC(C(N2C)=O)=C(C=C1)C2=O)CCC3=NC(CSC4=NNC(C5=CC=CC=C5)=N4)=NO3	-6.866	-6.939	-57.58	0.183	5.829	4.915
d27		O=C(/C(S1)=C(C(C=CC=C2)=C2N3CC(N)=O)/C3=O)N(C1=NC(C)=C4C(OC)=O)[C@@H]4C5=CC=C(OC(C6=CC=CO6)=O)C=C5	-6.356	-6.356	-57.39	0.000	7.303	7.303

d28		ClC(C=C1)=CN2C1=NC(COC3=CC=CC(NC(NC4=CC=CC=C4C(OCC)=O)=O)=C3)=CC2=O	-6.386	-6.386	-57.22	0.193	2.804	1.838
d29		O=S(NCCN1CCOCC1)(C2=CC(NC(C3=CC=CC(F)=C3)=O)=CC=C2OC4=C(C)C(C(O)=O)=NN4C5=CC(F)=CC=C5)=O	-6.781	-8.012	-56.35	0.534	3.533	0.862
d30		CN(C)C1=CC=C(NC(CSC2=C(C(N)=O)C3=C(C(O)=CC(N3)=O)S2)=O)C=C1	-9.478	-9.488	-52.93	0.130	3.681	3.029

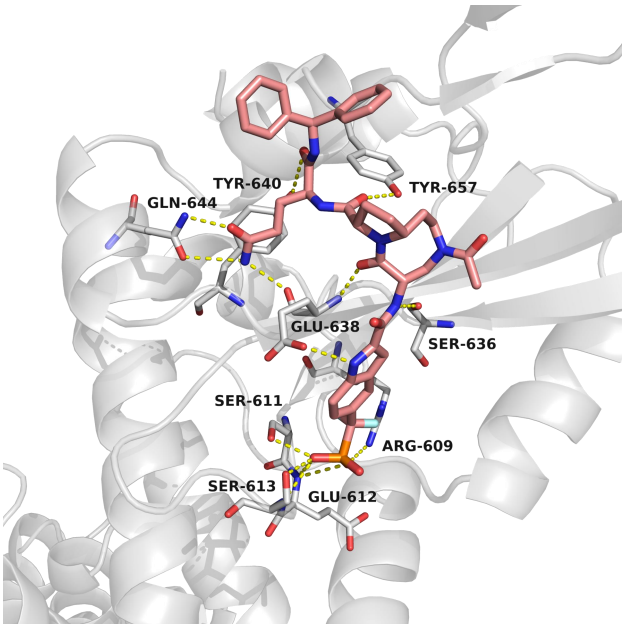


Fig. S1 Diagram of the interactions between SI-109 and the STAT3 protein (PDB: 6NJS). H-bonds is represented by yellow dashed lines.

Raw data of Western Blots Images

Fig. 8D Effect of **d2** on the expression of STAT3 in TNBC cells

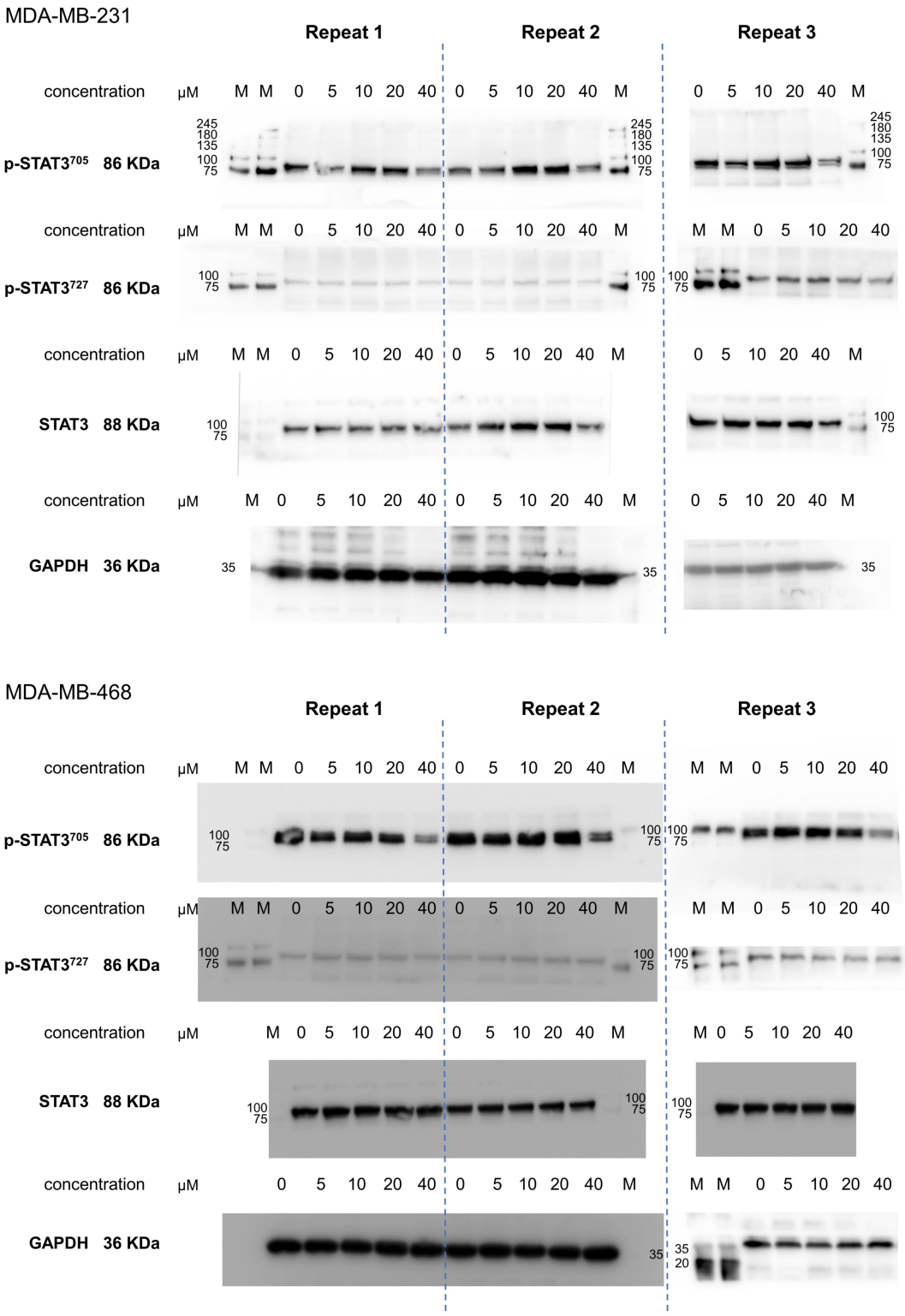


Fig. 8F Effect of **d10** on the expression of STAT3 in MDA-MB-468 cells

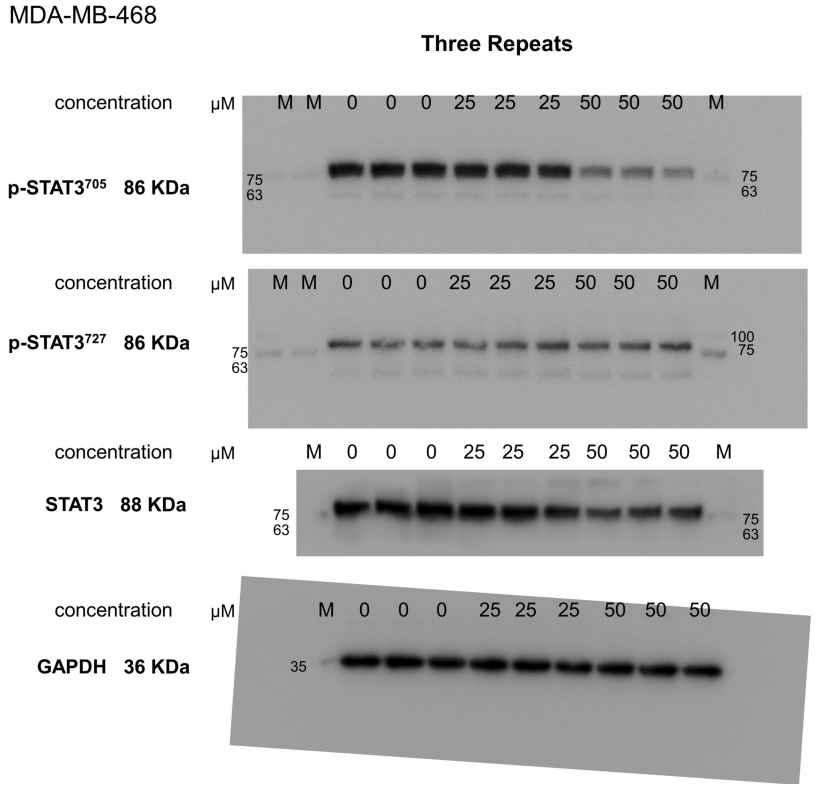
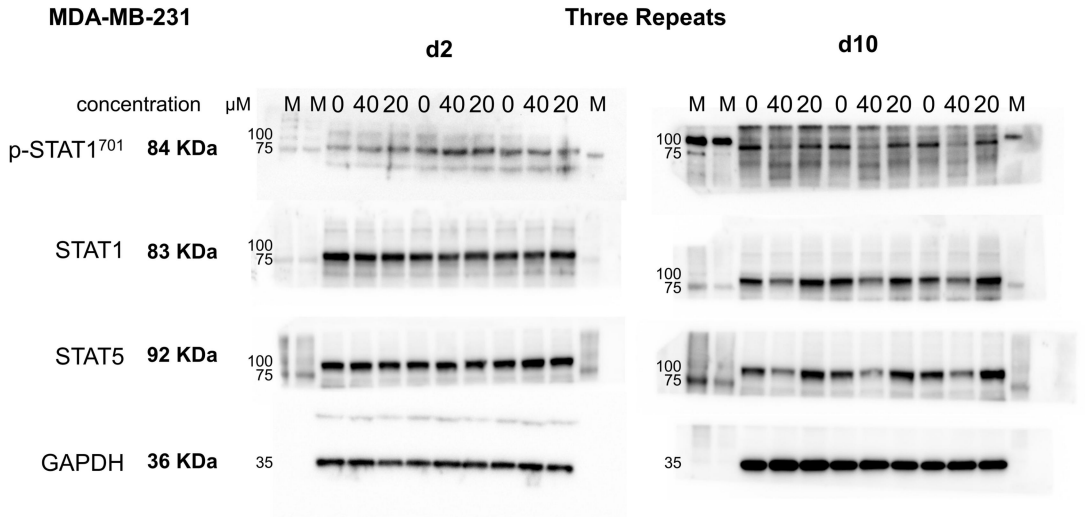
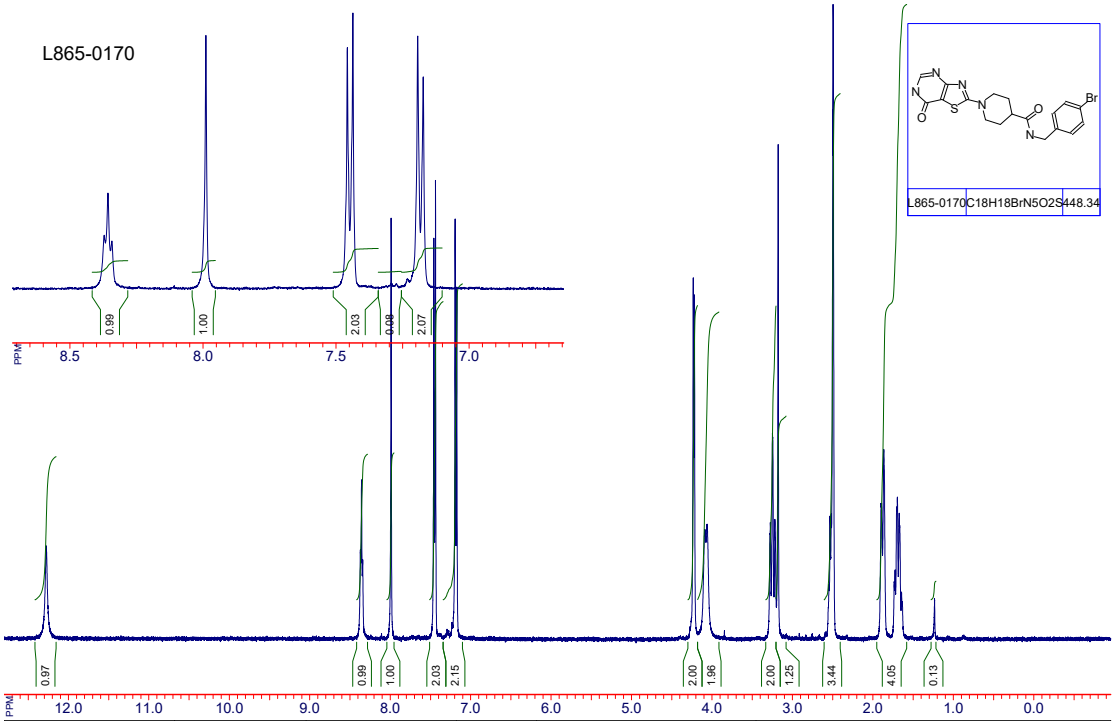


Fig. 8G The selectivity of **d2** and **d10** against STAT isoforms.

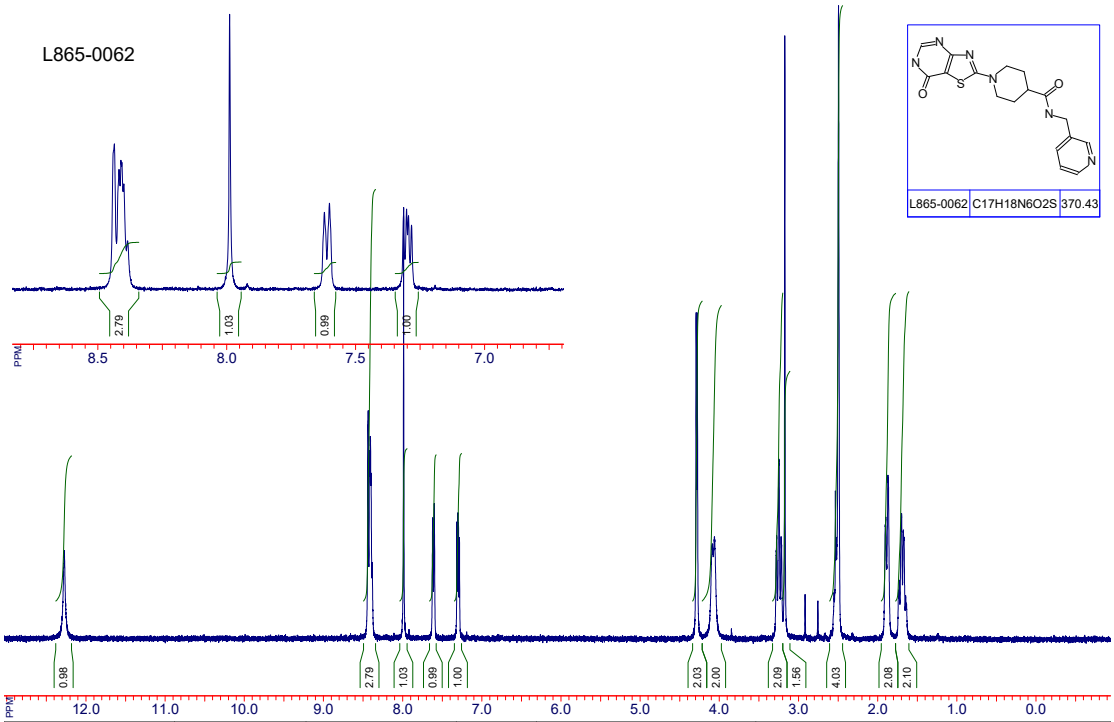


Characterization information of the 11 test compounds provided by the supplier



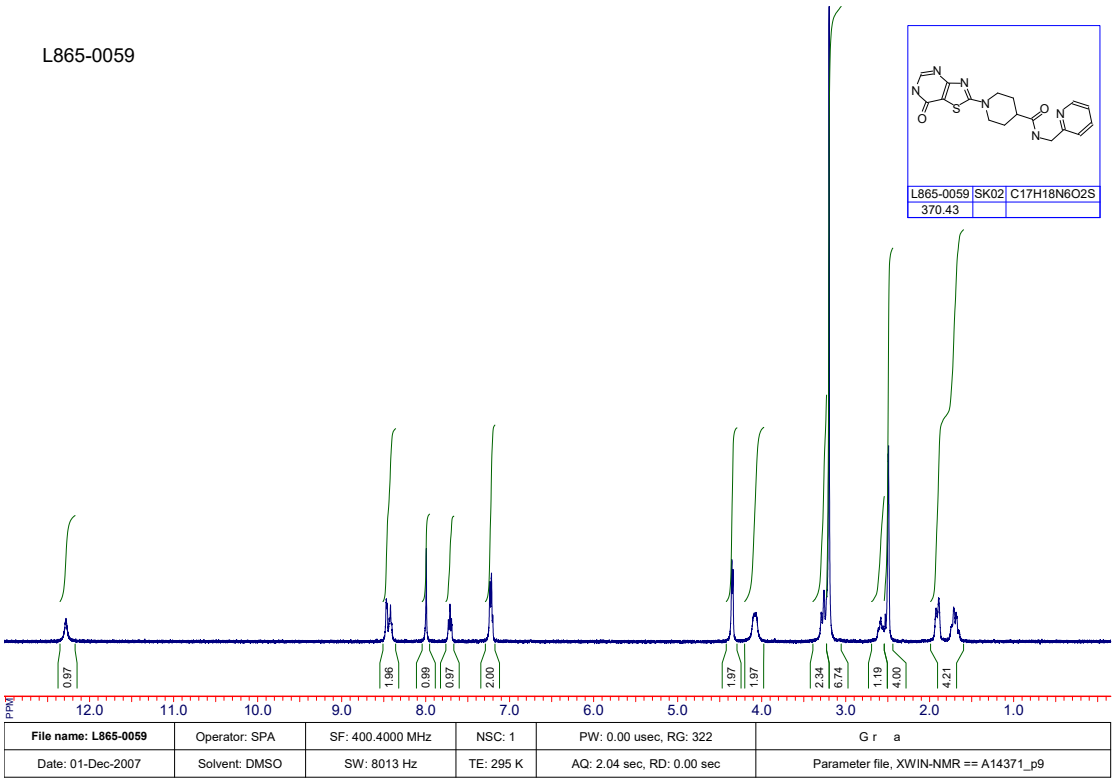
File name: L865-0170	Operator: MEL	SF: 400.4000 MHz	NSC: 1	PW: 0.00 usec, RG: 181	G r a
Date: 22-Nov-2007	Solvent: DMSO	SW: 8013 Hz	TE: 296 K	AQ: 2.04 sec, RD: 0.00 sec	Parameter file, XWIN-NMR == A14365_p5

^{1}H NMR spectrum of compound **a5**

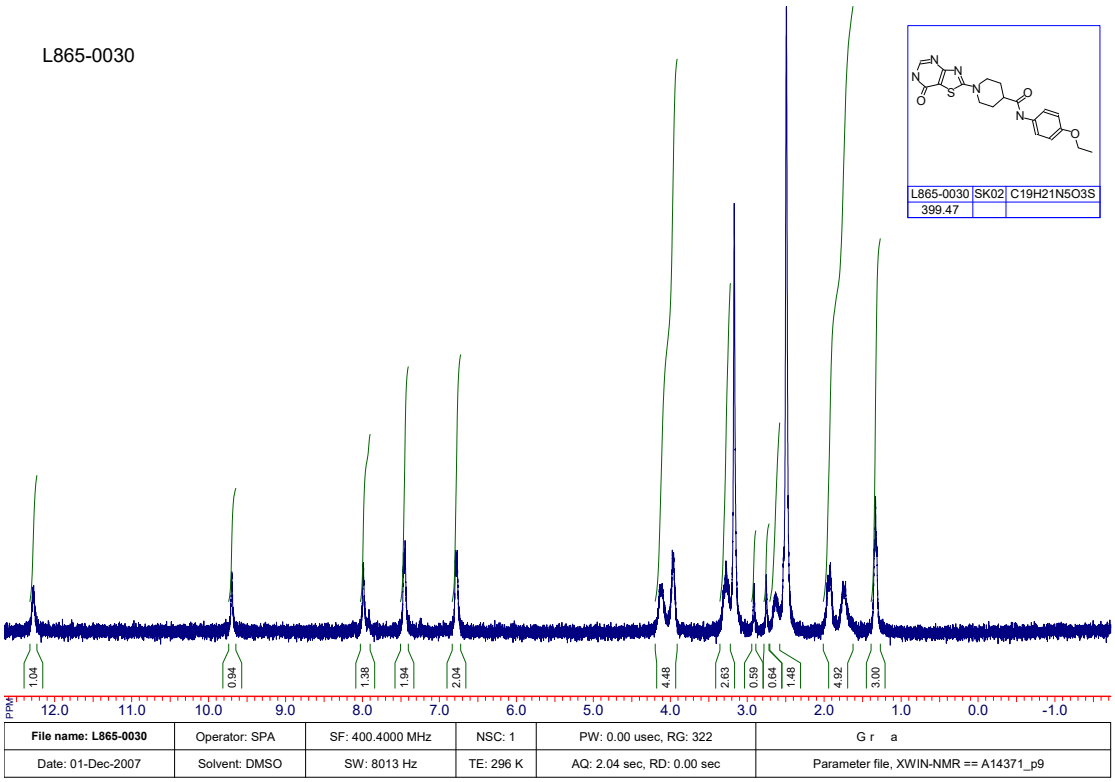


File name: L865-0062	Operator: MEL	SF: 400.4000 MHz	NSC: 1	PW: 0.00 usec, RG: 181	G r a
Date: 22-Nov-2007	Solvent: DMSO	SW: 8013 Hz	TE: 296 K	AQ: 2.04 sec, RD: 0.00 sec	Parameter file, XWIN-NMR == A14365_p5

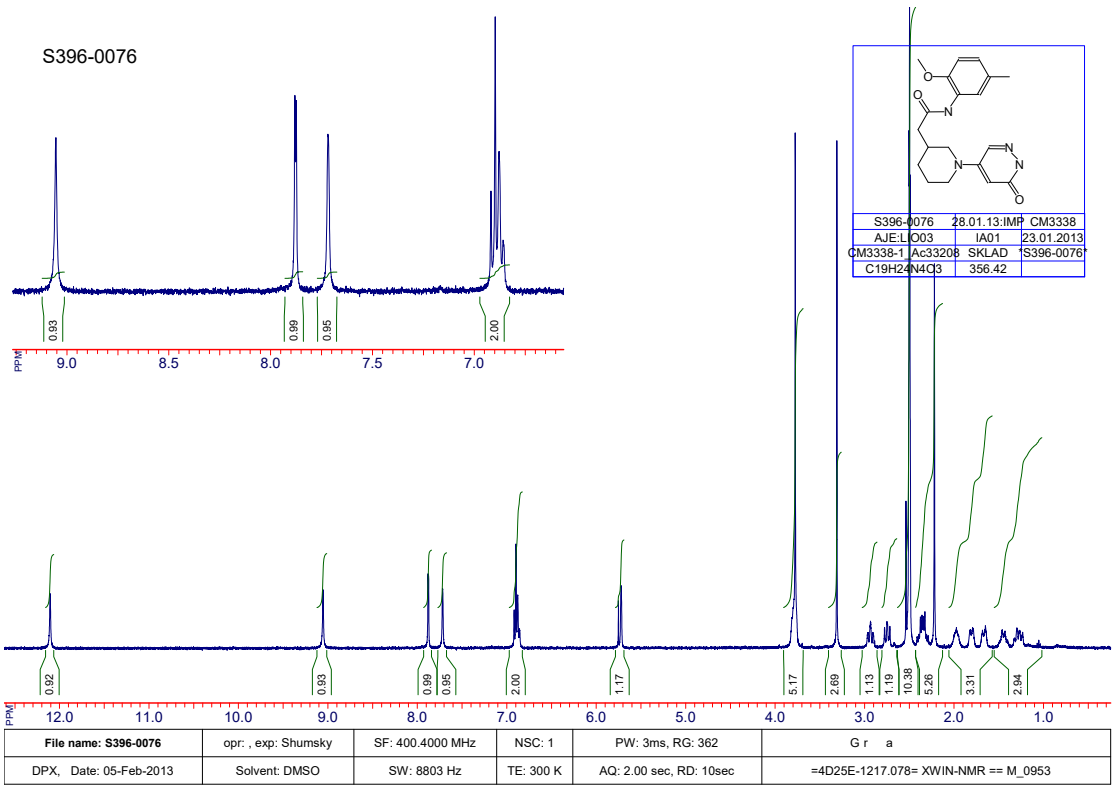
^{1}H NMR spectrum of compound **a6**



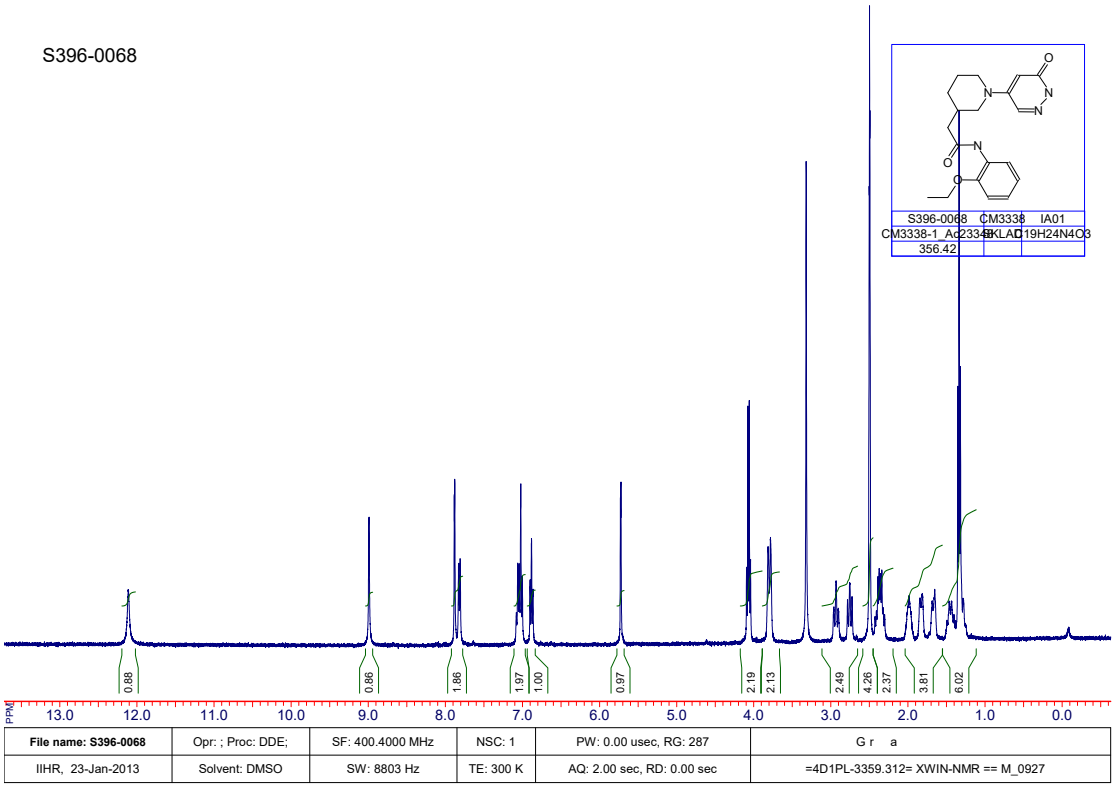
^{1}H NMR spectrum of compound **a7**



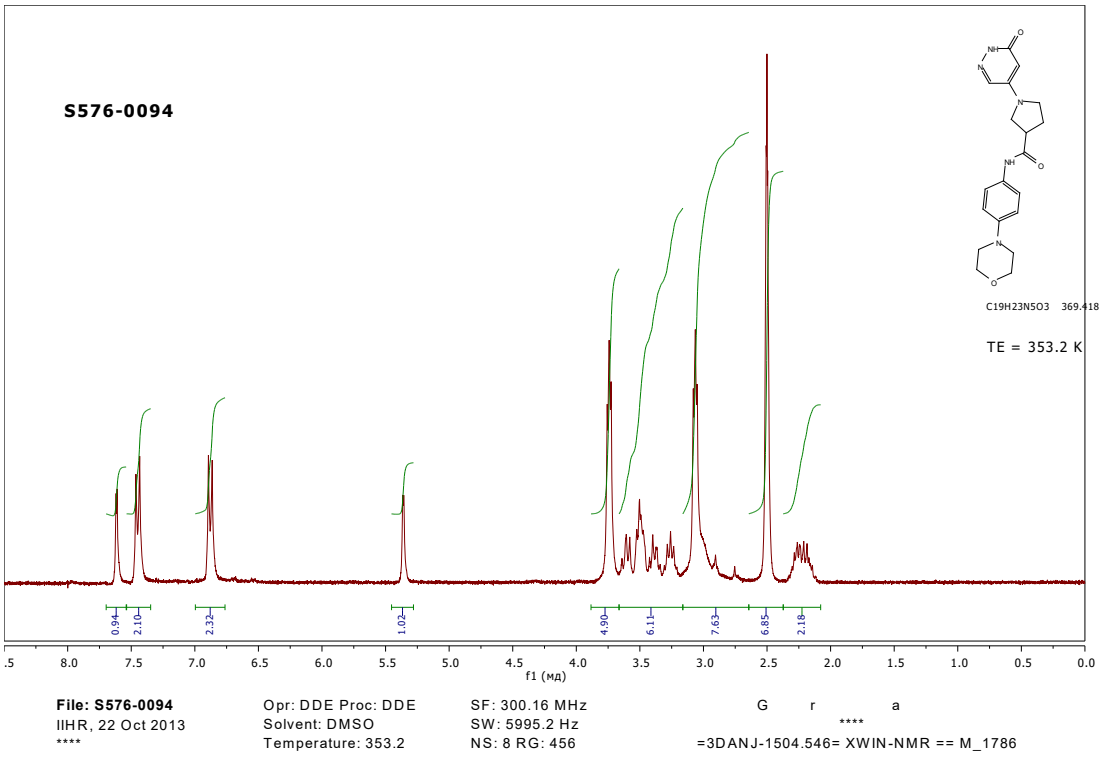
^{1}H NMR spectrum of compound **a10**



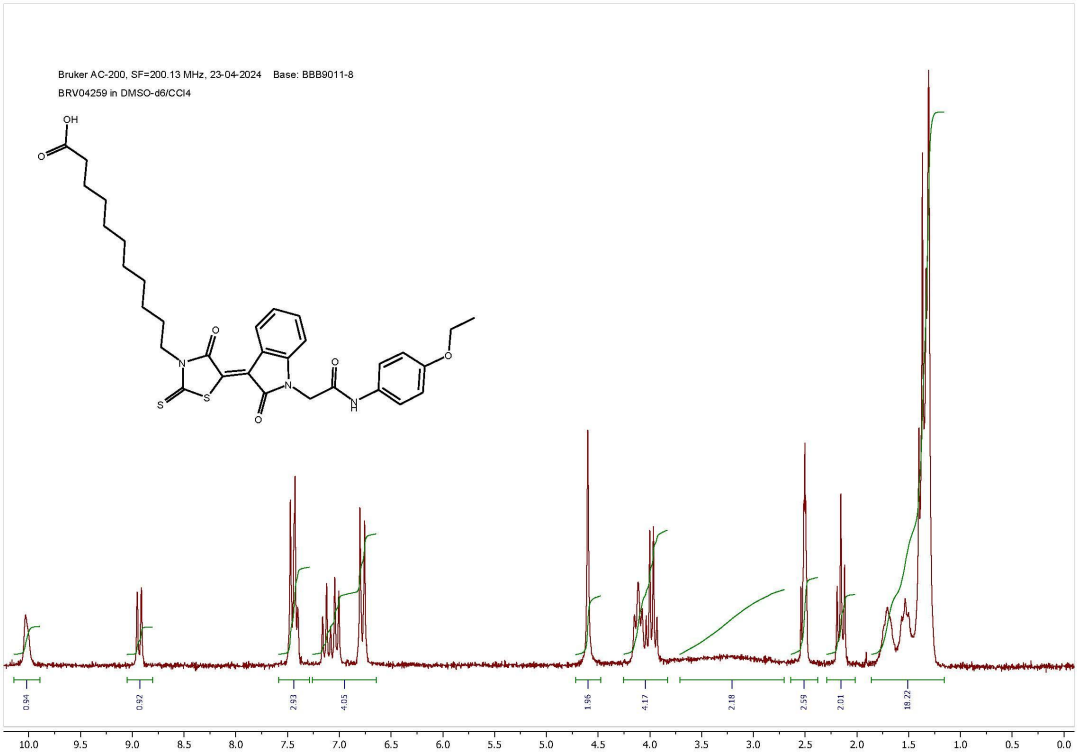
^{1}H NMR spectrum of compound **b4**



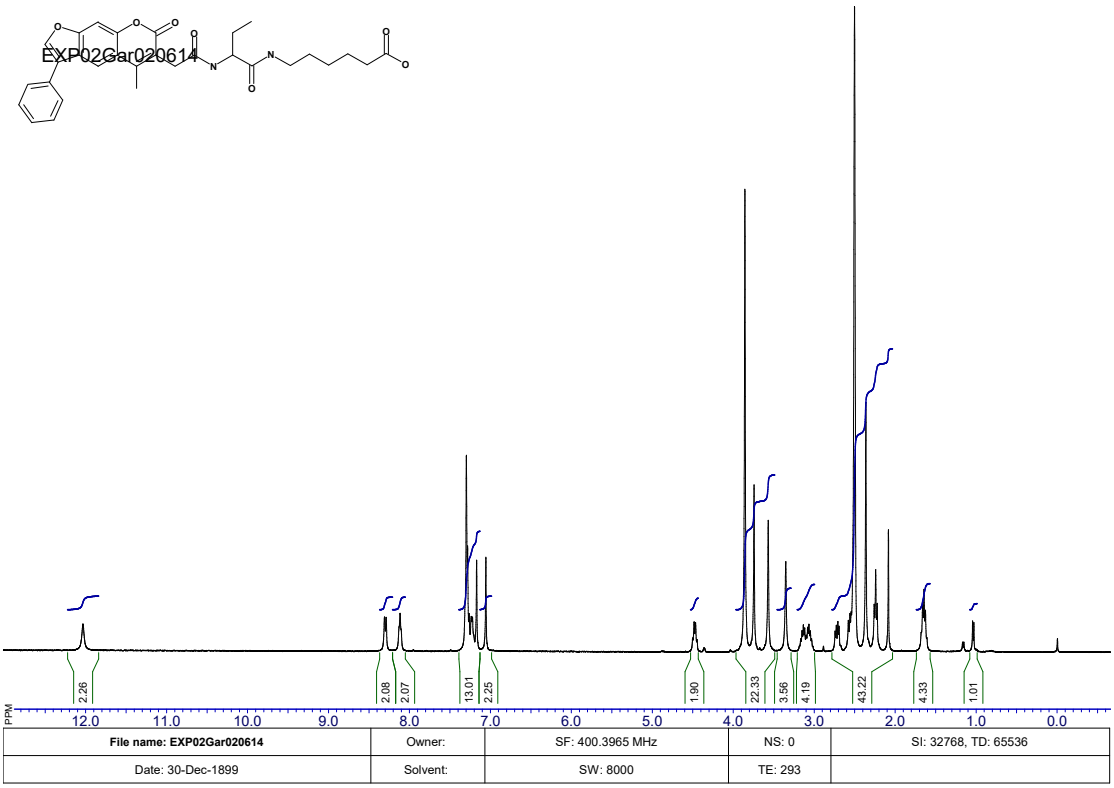
^{1}H NMR spectrum of compound **b9**



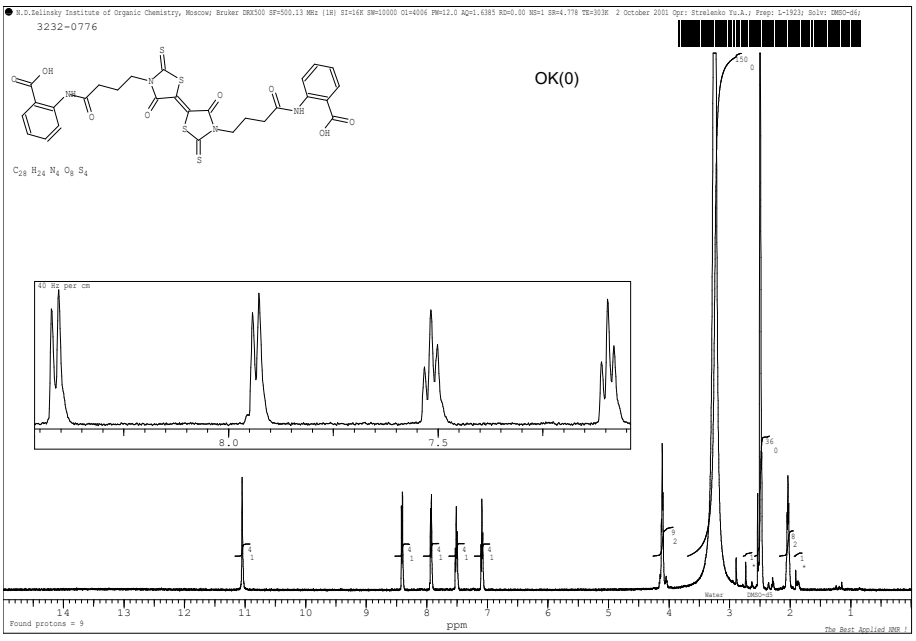
^{1}H NMR spectrum of compound **b11**



^{1}H NMR spectrum of compound **d2**

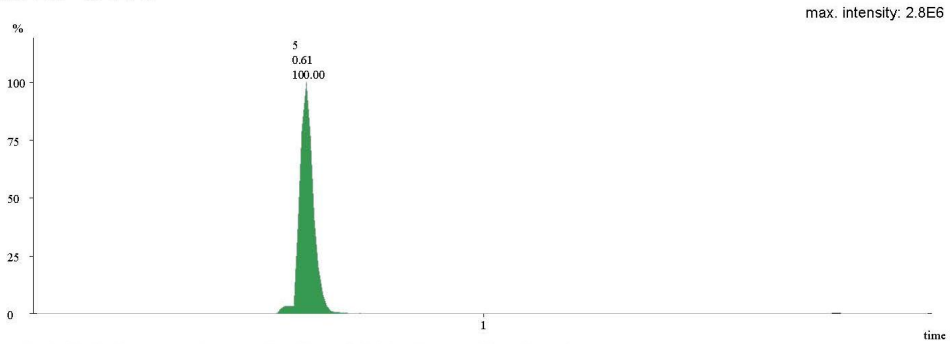


^{1}H NMR spectrum of compound **d3**



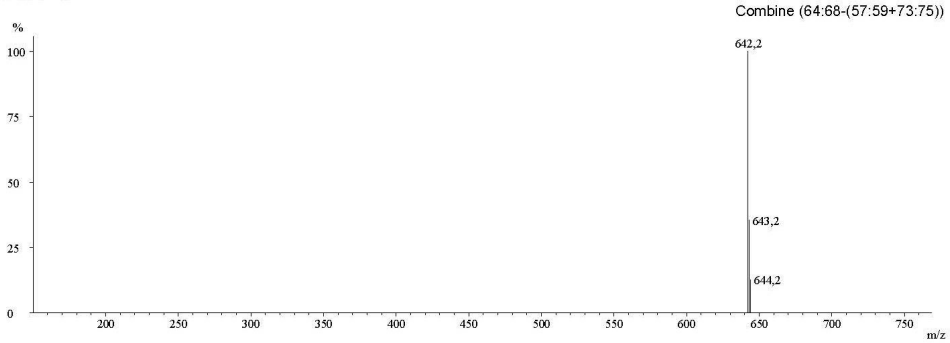
^{1}H NMR spectrum of compound **d10**

MS ES+ :642.18



Peak_ID	Peak	Area	Area%	Height	Time	Mass Found
5	0.52 0.71	1.E5	100	3.E6	0.61	641.18

MS: ES+



Peak_ID	Compound	Time	Mass found
5	Found	0.61	641.1800

MS spectrum of compound **d29**