

Supporting Information

Discovery of Potent Apoptosis Signal-Regulating Kinase 1 Inhibitors via Integrated Computational Strategy and Biological Evaluation

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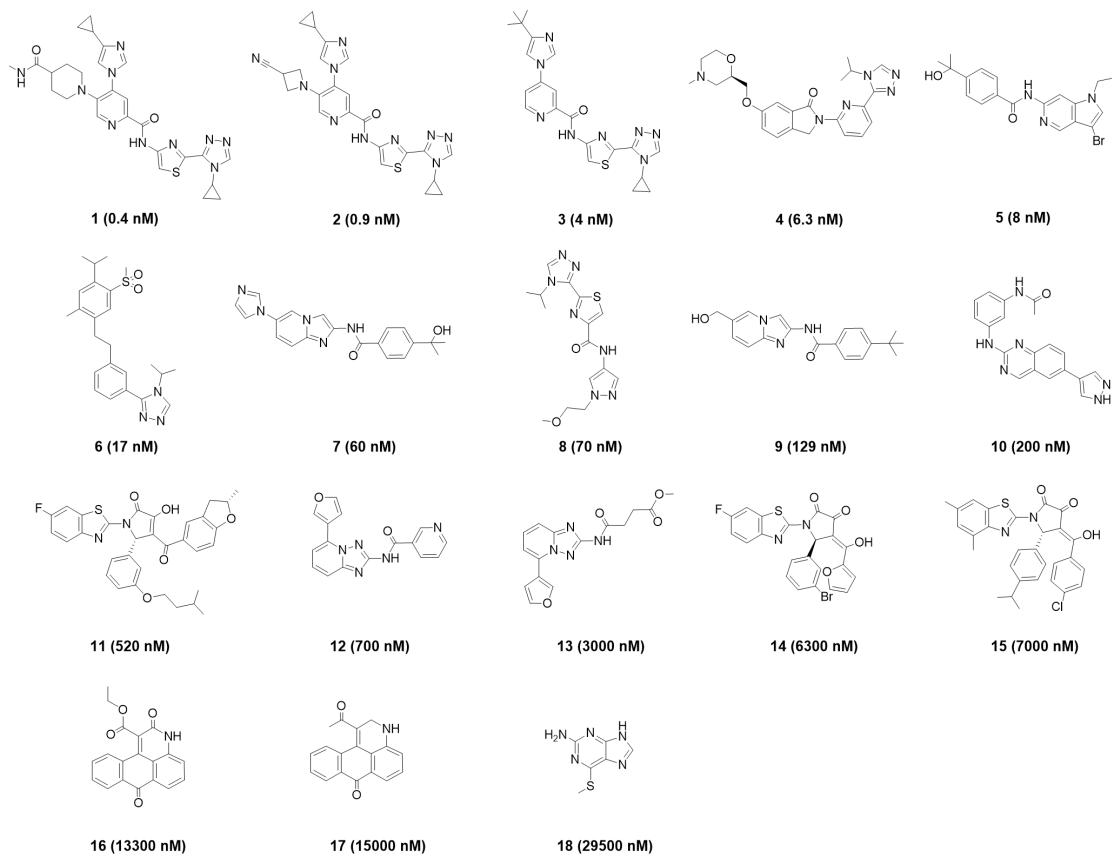


Figure S1. Chemical structures of 18 ASK1 inhibitors in the training set together with their biological activity values (IC_{50} , nM).

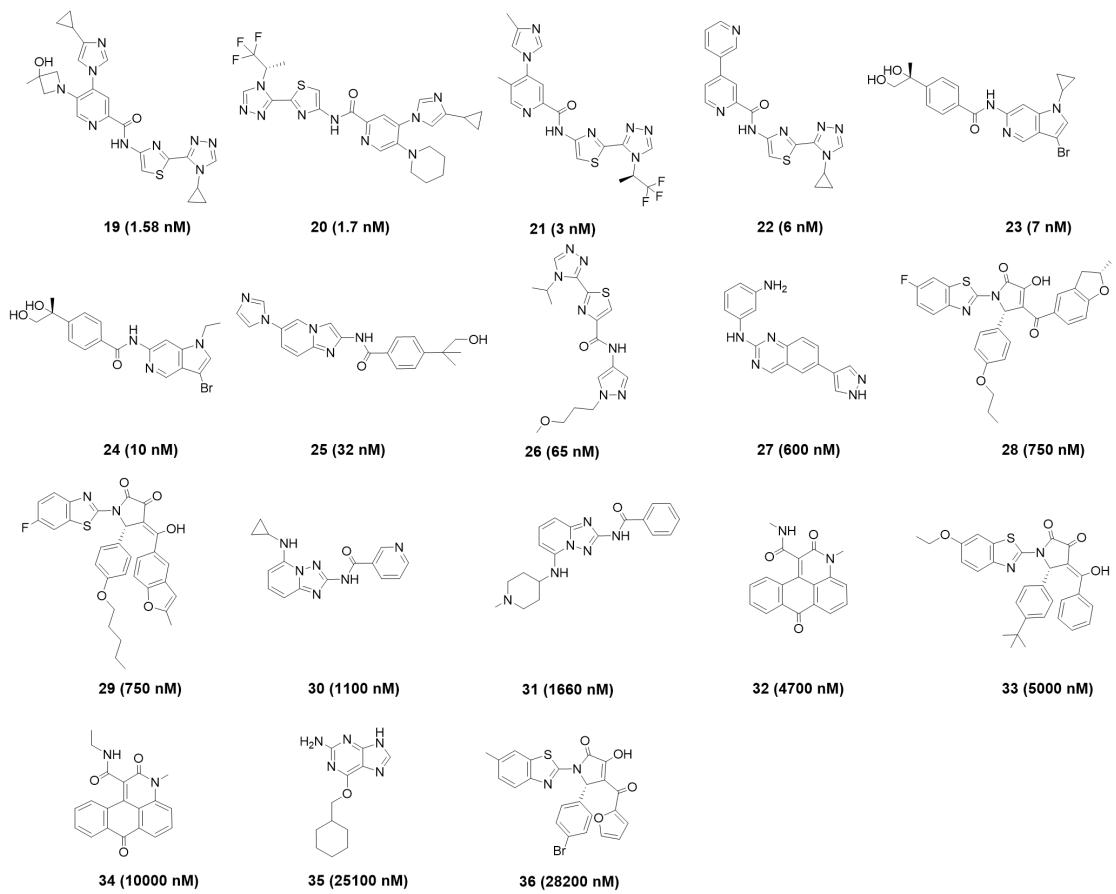


Figure S2. Chemical structures of 18 ASK1 inhibitors in the test set together with their biological activity values (IC_{50} , nM).

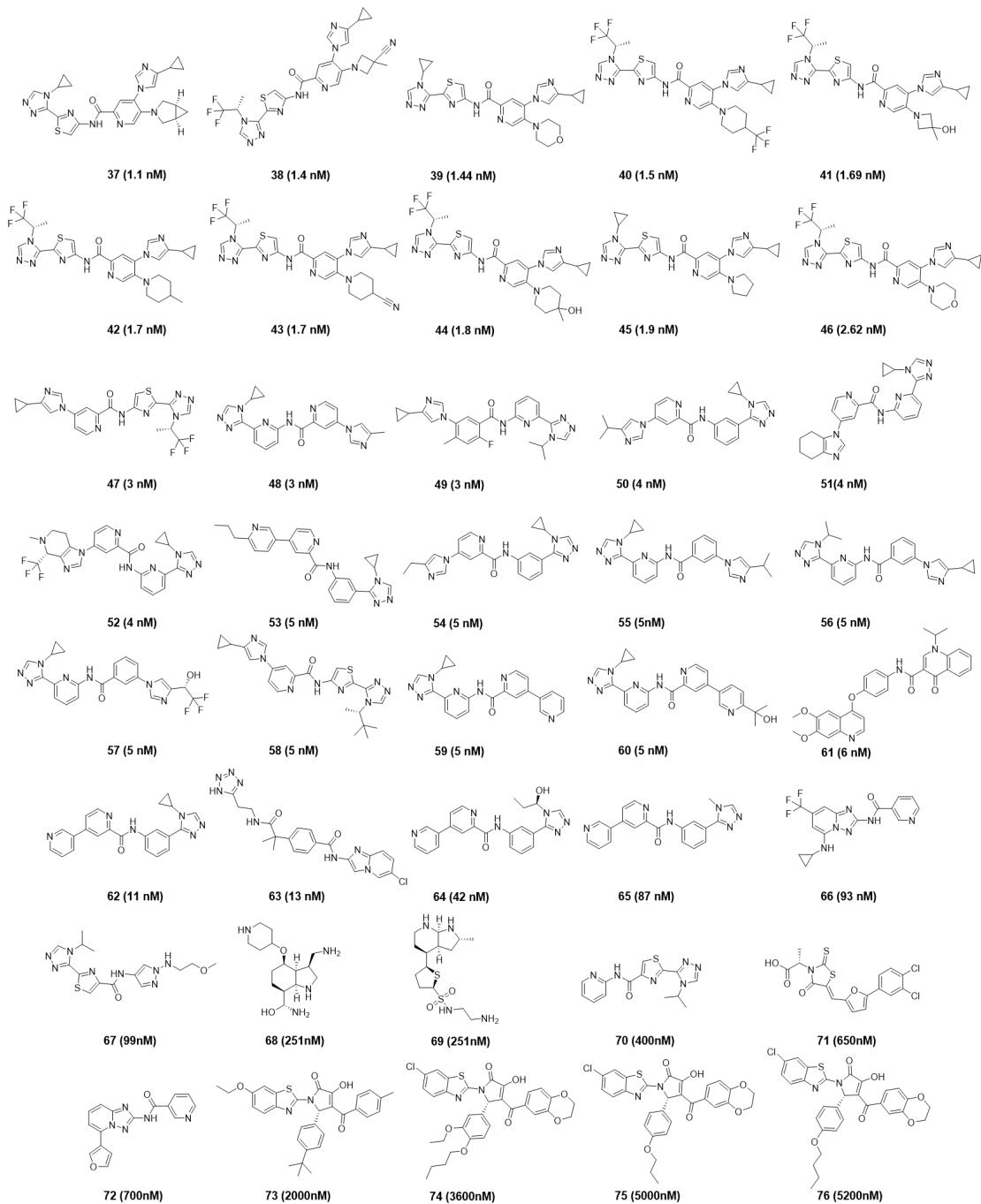


Figure S3. Chemical structures of 40 ASK1 inhibitors in the validation set together with their biological activity values (IC_{50} , nM).

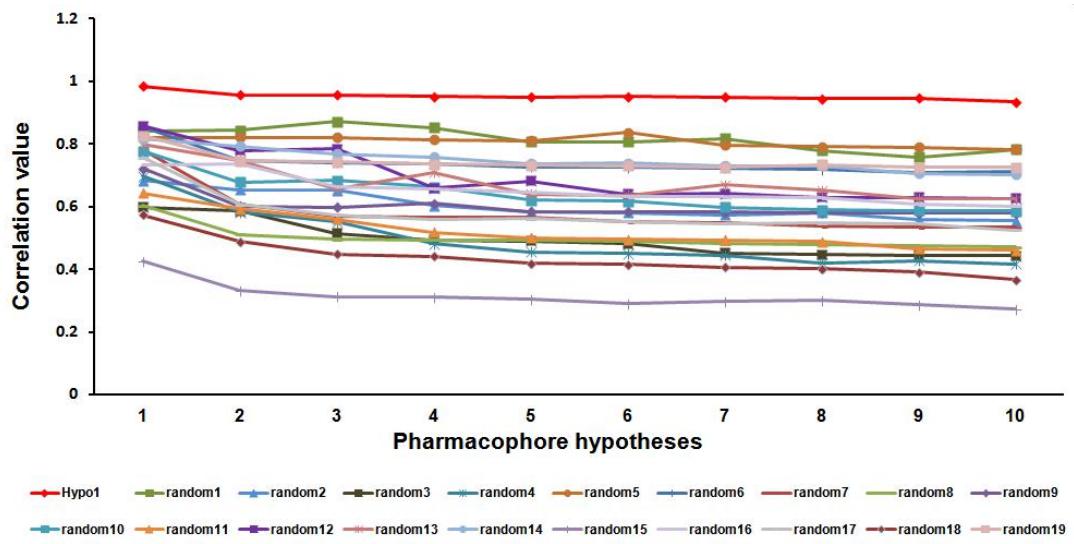


Figure S4. Comparison on correlation values of Hypo 1 and 19 randomly generated hypotheses at the 95% confidence level. Among the 20 broken lines, the red one located at the top, meaning that Hypo 1 had the highest correlation value.

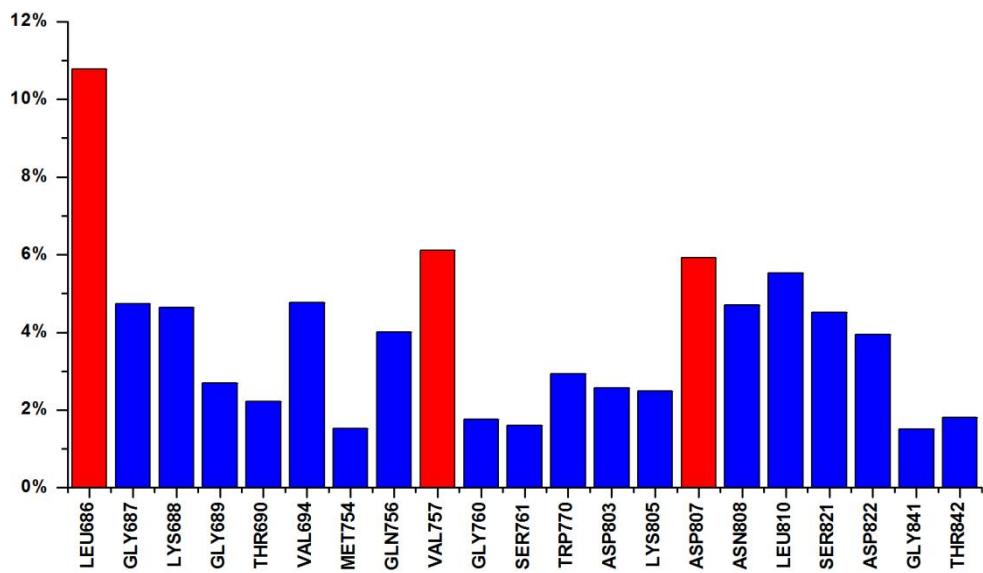
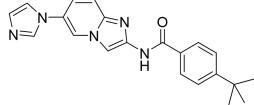
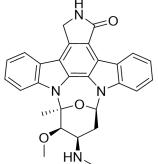
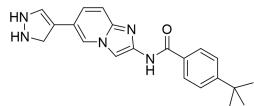
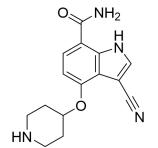
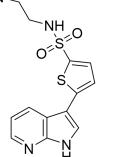
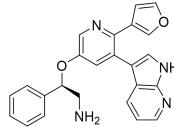
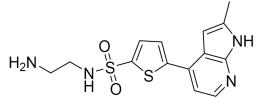
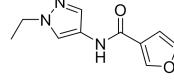
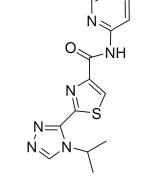
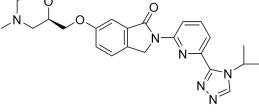
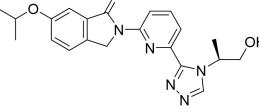
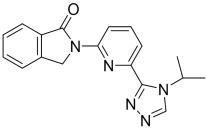


Figure S5. Proportion distribution of non-bonded interactions among the ASK1 residues. LEU686 had the highest proportion distribution, proving its importance for ligand binding of ASK1 protein.

Table S1. Detailed information of 12 ASK1 PDB Cognate Ligands.

No.	PDB_ID	Resolution (Å)	R _{free} Value	Ligand_ID	Ligand_Structure	Ligand IC ₅₀ (nM)	Release Date
1	3VW6	2.4	0.252	IM6		14	2012-10-31
2	4BF2	2.11	0.247	STU		NT ^a	2013-7-3
3	4BHN	2.3	0.235	BH9		6.3	2013-7-3
4	4BIB	2.43	0.239	IEO		251	2013-7-3
5	4BIC	2.62	0.265	IE4		50.1	2013-7-3
6	4BID	2.8	0.259	IE8		7.9	2013-7-3
7	4BIE	2.36	0.253	IE6		251	2013-7-3
8	5V19	3.1	0.285	8V4		500,000	2017-3-29
9	5V24	2.5	0.266	8V7		400	2017-3-29
10	5UOR	2.75	0.267	8GV		6.3	2017-6-7
11	5UOX	2.5	0.253	8GY		5.0	2017-6-7

12	5UP3	2.95	0.289	8GS		12.6	2017-6-7
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NT^a: No accurate IC₅₀ value is given.

Table S2. Actual and Predicted Activities of Test Set Molecules Calculated on the Basis of Hypothesis 1 of ASK1.

Cpd	FitValue	IC ₅₀ (nM) ^a		Error ^b	Activity scale ^c	
		Experimental	Estimated		Experimental	Estimated
19	7.96	1.58	7.47	4.73	+++	+++
20	8.05	1.7	6.02	3.54	+++	+++
21	8.40	3	2.68	-1.12	+++	+++
22	8.12	6	5.13	-1.17	+++	+++
23	7.57	7	18.25	2.61	+++	+++
24	7.47	10	23.13	2.31	+++	+++
25	7.18	32	44.87	1.40	+++	+++
26	7.25	65	37.63	-1.73	+++	+++
27	6.36	600	293.66	-2.04	++	++
28	7.14	750	49.17	-15.25	++	+++
29	6.70	750	135.41	-5.54	++	++
30	6.27	1,100	360.17	-3.05	+	++
31	6.68	1,660	140.86	-11.79	+	++
32	5.06	4,700	5,954.81	1.27	+	+
33	5.99	5,000	694.79	-7.20	+	++
34	5.10	10,000	5,406.07	-1.845	-	+
35	4.66	25,100	14,64	-1.71	-	+
36	6.14	28,200	490.88	-57.45	-	++

^a Data for activities of ASK1 inhibitors are from references listed in Materials and methods section.

^b Difference between the predicted and experimental values. ‘+’ indicates that the estimated activity is higher than experimental activity; ‘-’ indicates that the estimated activity is lower than experimental activity.

^c Activity scale: + + + (< 100 nM, highly active); + + (100 nM – 1 μM, moderately active); + (1 μM – 10 μM, less active); – (> 10 μM, inactive).

Table S3. Cross-Docking RMSD Values of 12 ASK1 Crystal Structures.

PDB ID	3VW6	4BF2	4BHN	4BIB	4BIC	4BID	4BIE	5UOR	5UOX	5UP3	5V19	5V24
3VW6	4.541	12.539	6.069	7.521	8.988	10.219	10.449	10.515	7.808	12.453	0.528	8.771
4BF2	11.665	4.588	7.041	5.505	5.689	—	4.162	8.716	7.032	10.246	6.972	7.639
4BHN	4.582	12.602	9.14	9.343	9.968	6.282	10.322	1.072	7.202	12.152	1.191	10.741
4BIB	11.592	3.997	4.754	4.703	3.864	7.46	5.458	5.766	5.846	8.13	1.296	1.364
4BIC	7.887	4.247	5.46	4.818	3.848	4.765	5.166	1.499	5.315	8.584	2.995	4.543
4BID	11.64	5.952	7.964	8.131	7.753	6.056	7.696	6.592	6.365	8.345	6.6	4.176
4BIE	8.668	4.861	5.89	5.631	4.988	6.472	6.085	3.67	5.981	7.359	3.802	3.801
5UOR	4.922	7.884	5.713	9.349	8.431	9.311	10.679	1.651	6.522	5.435	1.686	1.546
5UOX	4.994	5.922	4.298	7.224	6.287	6.283	8.007	0.662	4.683	5.41	0.566	0.66
5UP3	5.184	4.515	3.871	5.58	4.736	7.945	6.322	0.324	3.581	5.435	0.686	0.483
5V19	4.947	7.879	7.314	8.095	7.207	9.781	9.355	4.229	6.89	6.119	3.985	4.221
5V24	5.212	4.922	4.445	5.756	4.96	10.219	6.656	1.075	4.136	5.503	1.119	1.043
Mean (Å)	7.153	6.659	5.997	6.805	6.393	7.708	7.530	3.814	5.947	7.931	2.619	4.082
StdDev (Å)	2.867	2.915	1.536	1.609	1.965	1.824	2.148	3.264	1.242	2.459	2.188	3.253