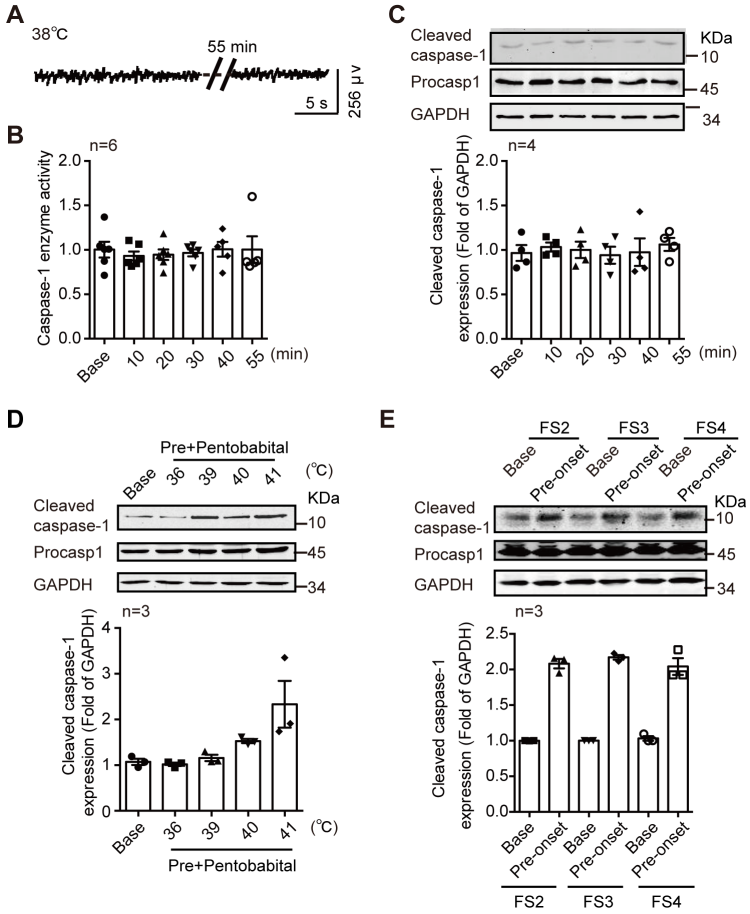
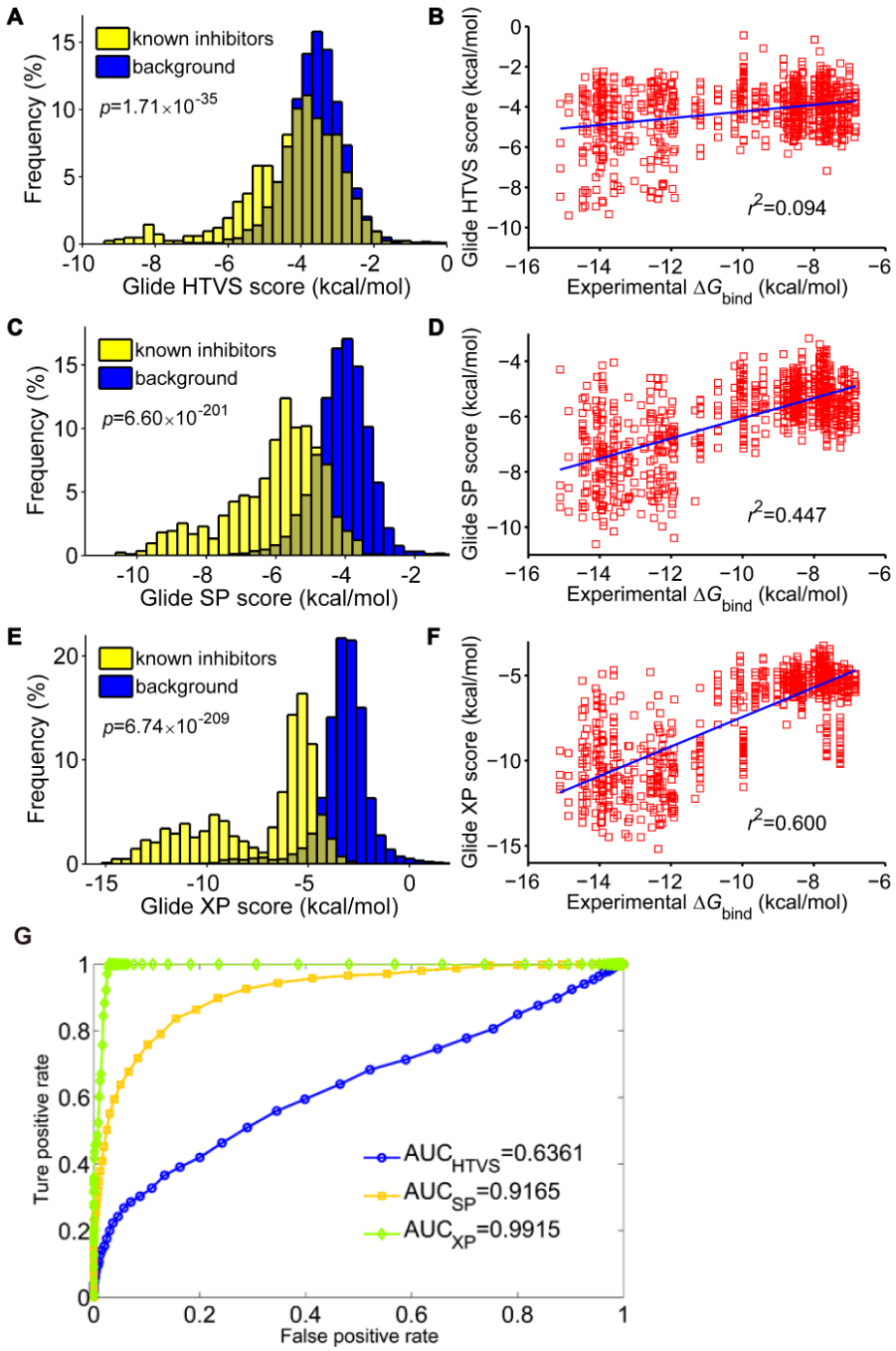
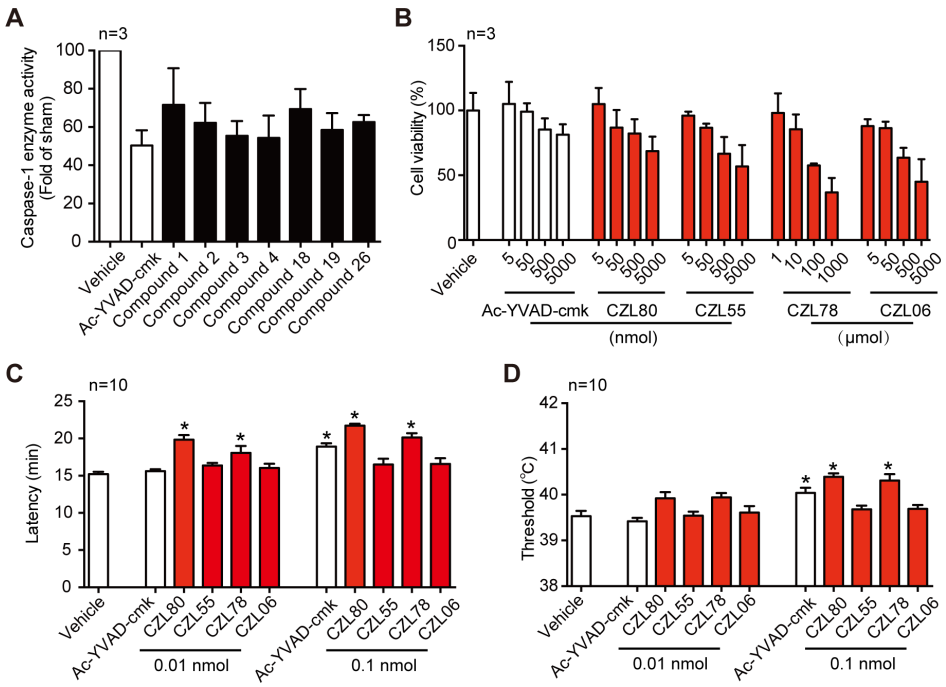


Supplementary Figure 1. Fever, but not seizure, increases the expression of cleaved caspase-1. (A) Typical EEG of FS onset under 38°C condition. **(B)** The enzyme activity of caspase-1 at different time points under 38°C condition. **(C)** The amount of caspase-1 at different time points under 38°C condition was analyzed by western blotting with indicated antibodies. Data were quantified and normalized against the base. **(D)** Hippocampus samples of indicated body temperature were analyzed by western blotting with indicated antibodies in mice with pentobarbital pretreatment, and data were quantified and normalized with the base. **(E)** The level of cleaved caspase-1 increased prior to recurrence FS. Hippocampus samples were analyzed by western blotting, and data were quantified and normalized with the base. Data are presented as means ± s.e.m..

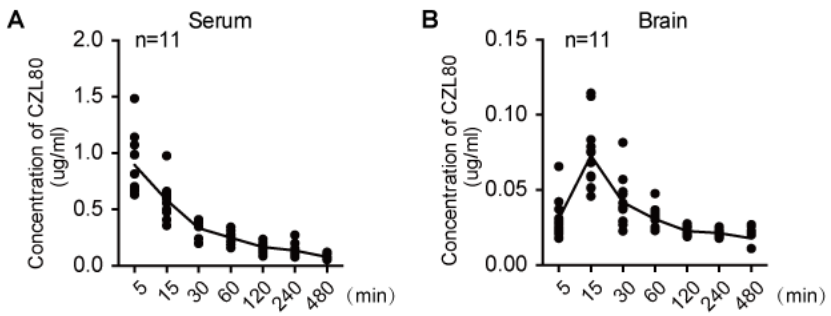


Supplementary Figure 2. Validation of different docking approaches on the test dataset. (A-F) The distribution of the known inhibitors and chemical background (non-inhibitors) are illustrated in panels **A**, **C**, and **E** for glide HTVS docking (High Throughput Virtual Screening), glide SP docking (Standard Precision), and glide XP docking (Extra Precision), respectively. The corresponding Pearson correlation coefficients are shown in panels **B**, **D**, and **F**, respectively. **Actives set:** A total of 824

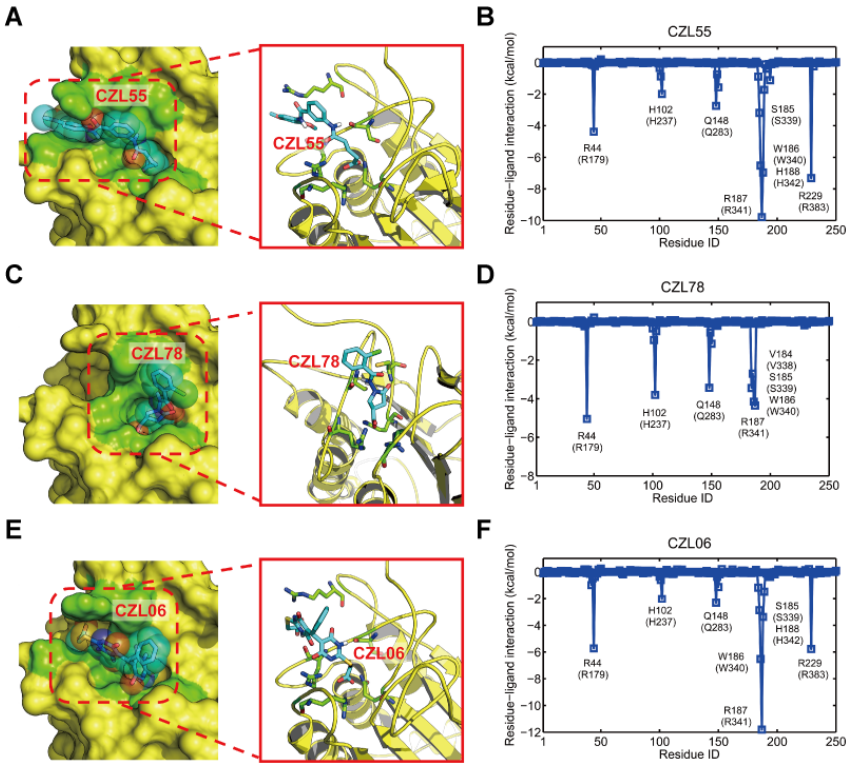
known inhibitors (with IC50 < 10 μM) were collected from the BindingDB database. **Inactives set:** To mimic the fact that few compounds can exhibit activity to a certain target, 17,000 compounds randomly chosen from the ChemBridge database were used as the non-inhibitors, making the ratio of inhibitors and non-inhibitors 1:20. These molecules have the maximum diversity evaluated by the 2-D similarities (Tanimoto coefficient) based on the FCFP_4 fingerprints in the Find Diverse Molecules module in Discovery Studio 2.5, and thereafter can represent a real chemical background. **(G)** ROC curves of different docking methods. The ROC curves of HTVS, SP, and XP docking methods on the test dataset are shown in blue cycle line, orange square line, and green diamond line, respectively,



Supplementary Figure 3. Screening the top-ranked compounds *in vitro* and *in vivo*. **(A)** The enzyme activity of caspase-1 of the selected 7 compounds and Ac-YVAD-cmk on LPS-pretreated PBMCs compared with the sham group. **(B)** Cell viability following administration of four indicated compounds and Ac-YVAD-cmk tested by MTT. **(C-D)** The latency **(C)** and threshold **(D)** of FS generation in WT mice treated with vehicle, Ac-YVAD-cmk, CZL80, CZL55, CZL78, and CZL06 (0.01 nmol and 0.1 nmol, *i.c.v*). Data are presented as means ± s.e.m.. **P* <0.05, One-way ANOVA followed by Dunnett *post hoc* test was used for **C, D**.



Supplementary Figure 4. CZL80 penetrates the BBB. (A and B) Concentration of CZL80 in serum **(A)** and brain **(B)** were measured at 5, 15, 30, 60, 120, 240, 480 min after CZL80 (0.75 mg/kg) was administered intravenously.



Supplementary Figure 5. Structural and energetic representations of the 3 compounds (CZL55, CZL78, and CZL06) interacting with caspase-1. (A, C, E) The solvent-accessible surfaces of the 3 compounds to the binding pocket of caspase-1. **(B, D, F)** The corresponding hydrogen bond networks of the 3 compounds to the binding pocket of caspase-1. The compounds, the key residues, and the protein backbones are shown in cyan stick model, green stick model, and yellow cartoon model, respectively.

Supplementary Tables

Supplementary Table 1. Detailed information of the selected 50 compounds obtained and tested in bioassays.

ID	IC_{50} (μM)	Score[a]	MW[b]	ClogP[c]	LogS[d]	Caspase-1 enzyme activity (Fold of Sham)	Rank[e]
CZL80	0.010	-11.5398	365.39	3.22	-4.621	38.14±9.21***	7
CZL55	0.024	-10.5803	385.4	1.63	-3.409	53.81±3.02**	48
CZL78	3.742	-10.7835	281.678	0.55	-1.732	58.88±1.92*	33
Compound 1	46.03	-12.0615	429.862	3.33	-5.185	56.51±1.59*	2
Compound 2	46.28	-10.8508	339.374	2.534	-3.846	57.38±4.39*	30
Compound 3	100.6	-10.6441	375.792	0.41	-2.297	58.59±3.92*	40
Compound 4	153.8	-11.0186	438.397	1.5	-3.691	59.74±4.73*	21
CZL06	11.31	-11.3766	489.465	3.19	-5.119	54.48±2.90**	8
Compound 5	-	-10.6586	189.982	0.839	-1.311	91.89±3.56	38
Compound 6	-	-11.9513	446.468	1.26	-3.538	96.06±2.72	3
Compound 7	-	-10.5369	330.343	2.53	-3.77	94.15±1.17	52
Compound 8	-	-10.6414	341.347	2.39	-3.737	92.83±1.23	41
Compound 9	-	-10.9581	255.315	1.97	-2.752	97.10±2.00	24
Compound 10	-	-10.5766	352.433	4.32	-5.466	94.78±2.45	49
Compound 11	-	-11.2351	401.402	4.6	-6.327	93.50±1.90	12
Compound 12	-	-11.185	299.239	-3.05	-0.862	92.9±2.53	15
Compound 13	-	-11.6575	420.338	4.78	-6.751	93.74±1.69	6
Compound 14	-	-10.5924	283.67	0.98	-2.121	95.15±2.23	46
Compound 15	-	-10.6024	317.281	0.74	-2.153	92.26±1.16	45
Compound 16	-	-10.9598	327.296	2.55	-3.779	94.94±2.33	23
Compound 17	-	-10.7602	311.122	-2.14	0.506	77.08±19.55	35
Compound 18	-	-10.757	359.792	1.69	-3.274	48.98±4.10***	36
Compound 19	-	-10.5105	349.775	1.43	-2.979	48.61±2.78***	58
Compound 20	-	-10.7535	357.776	1.75	-3.31	90.86±2.17	37
Compound 21	-	-10.4968	455.495	5.1	-7.148	91.93±1.05	59
Compound 22	-	-10.5298	459.954	4.23	-6.172	97.54±1.65	53
Compound 23	-	-10.9201	479.467	4.41	-6.468	94.94±1.06	26
Compound 24	-	-11.2813	469.519	3.08	-5.26	95.94±1.43	10
Compound 25	-	-10.9369	438.427	4.13	-5.93	84.89±2.63	25
Compound 26	-	-10.8375	387.438	3.7	-5.192	57.30±4.52*	31
Compound 27	-	-10.8673	491.346	5.33	-7.34	86.73±13.27	28
Compound 28	-	-11.2215	351.362	1.068	-2.482	74.03±16.98	13
Compound 29	-	-10.6029	447.394	3.622	-6.7	91.86±4.23	44
Compound 30	-	-10.5834	363.353	2.515	-3.901	82.11±8.41	47
Compound 31	-	-10.8958	277.236	1.358	-2.416	92.53±2.78	27
Compound 32	-	-10.4713	422.424	5.509	-6.318	93.23±1.77	61
Compound 33	-	-10.862	493.352	2.439	-4.886	92.74±2.78	29
Compound 34	-	-12.3631	503.452	3.881	-6.191	95.28±4.65	1
Compound 35	-	-10.5465	262.244	0.957	-1.938	95.20±2.41	50
Compound 36	-	-11.1631	328.348	2.62	-3.839	74.57±5.36	16
Compound 37	-	-11.2671	371.395	2.82	-4.323	85.09±9.05	11
Compound 38	-	-10.8281	341.368	2.271	-3.636	90.84±0.99	32
Compound 39	-	-10.5192	326.356	1.885	-2.222	91.50±5.77	57
Compound 40	-	-11.2069	353.401	3.897	-5.112	90.87±7.17	14
Compound 41	-	-10.6249	456.953	4.357	-6.259	86.51±10.67	43
Compound 42	-	-10.4667	507.302	3.19	-5.629	60.33±25.64	64
Compound 43	-	-11.776	370.41	3.324	-4.747	62.97±21.21	5
Compound 44	-	-11.1201	445.886	4.857	-6.87	89.04±10.40	18
Compound 45	-	-11.3567	277.299	-0.381	-2.215	83.19±10.38	9
Compound 46	-	-10.5244	314.324	0.503	-1.929	76.77±11.18	54

[a]Glide-XP docking score; [b]Molecular weight; [c]Predicted octanol/water partition coefficient; [d]Predicted aqueous solubility, *S* in mol/L; [e]Ranks of the molecule based on glide XP docking score.

Supplementary Table 2. H-bond occupancy details in each active compound.

Donor	Acceptor	Occupancy[a]	Donor	Acceptor	Occupancy
CZL80			**CZL55**		
R179-Sidechain	CZL80	224.00%[b]	R179-Sidechain	CZL55	271.00%
R341-Sidechain	CZL80	108.00%	R341-Sidechain	CZL55	132.00%
Q283-Sidechain	CZL80	83.00%	Q283-Sidechain	CZL55	84.00%
R383-Sidechain	CZL80	69.00%	R383-Sidechain	CZL55	52.00%
CZL80	R341-Backbone	5.00%	CZL55	R341-Backbone	58.00%
			CZL55	S339-Backbone	67.00%
			S339-Sidechain	CZL55	6.00%
			R341-Backbone	CZL55	17.00%
CZL78			**CZL06**		
R179-Sidechain	CZL78	256.00%	R179-Sidechain	CZL06	280.00%
R341-Sidechain	CZL78	167.00%	R341-Sidechain	CZL06	197.00%
Q283-Sidechain	CZL78	83.00%	Q283-Sidechain	CZL06	92.00%
CZL78	S339-Backbone	10.00%	R383-Sidechain	CZL06	69.00%
C285-Backbone	CZL78	5.00%	CZL06	S339-Backbone	46.00%
C285-Sidechain	CZL78	15.00%	CZL06	R341-Backbone	12.00%
			R341-Backbone	CZL06	90.00%
			W340-Sidechain	CZL06	23.00%

[a]H-bond occupancy was calculated based on 30 ns MD trajectory; [b]H-bond occupancy >100% means that more than one hydrogen bond were formed between the donor and the acceptor.

Supplementary Table 3. Binding free energy of the four tight-binding compounds.

	ΔH	$-T\Delta S$	ΔG_{bind}	IC_{50} (μM)
CZL80	-34.90±0.39[a]	13.54±3.20	-21.36±0.39	0.0072
CZL55	-48.70±0.46	30.31±1.49	-18.39±0.46	0.024
CZL78	-32.17±0.46	16.32±1.85	-15.85±0.46	3.742
CZL06	-40.37±0.58	26.07±1.93	-14.30±0.58	11.31

[a]Deviations were estimated by two blocks (block1 1~15 ns; block2 15~30 ns).