

Supplementary Table 1. In vitro side effect profiling study for LDN/OSU-0212320.

Receptor	Percent Inhibition 10 μ M
Neurotransmitter Related	
Adenosine, Non-selective	7.29%
Adrenergic, Alpha 1, Non-selective	24.98%
Adrenergic, Alpha 2, Non-selective	27.18%
Adrenergic, Beta, Non-selective	-20.94%
Dopamine Transporter	8.69%
Dopamine, D1 (h)	8.48%
Dopamine, D2s (h)	4.06%
GABA A, Agonist Site	-16.15%
GABA A, BDZ, alpha 1 site	12.73%
GABA-B	13.60%
Glutamate, AMPA Site (Ionotropic)	12.06%
Glutamate, Kainate Site (Ionotropic)	-1.03%
Glutamate, NMDA Agonist Site (Ionotropic)	0.12%
Glutamate, NMDA, Glycine (Stry-insens Site) (Ionotropic)	9.84%
Glycine, Strychnine-sensitive	0.99%
Histamine, H1	-5.54%
Histamine, H2	16.54%
Histamine, H3	4.80%
Melatonin, Non-selective	-5.54%
Muscarinic, M1 (hr)	-1.88%
Muscarinic, M2 (h)	0.82%
Muscarinic, Non-selective, Central	29.04%
Muscarinic, Non-selective, Peripheral	0.29%
Nicotinic, Neuronal (α -BnTx insensitive)	7.85%
Norepinephrine Transporter	2.87%
Opioid, Non-selective	-0.09%
Opioid, Orphanin, ORL1 (h)	11.55%
Serotonin Transporter	-3.02%
Serotonin, Non-selective	26.33%
Sigma, Non-Selective	10.19%
Steroids	
Estrogen	11.16%

Receptor	Percent Inhibition
	10 μ M
Testosterone (cytosolic) (h)	12.50%
Ion Channels	
Calcium Channel, Type L (Dihydropyridine Site)	43.18%
Calcium Channel, Type N	4.15%
Potassium Channel, ATP-Sensitive	-4.05%
Potassium Channel, Ca ²⁺ Act., VI	17.80%
Potassium Channel, I(Kr) (hERG) (h)	-6.44%
Sodium, Site 2	-0.39%
Second Messengers	
Nitric Oxide, NOS (Neuronal-Binding)	-17.09%
Prostaglandins	
Leukotriene, LTB ₄ (BLT)	7.56%
Leukotriene, LTD ₄ (CysLT ₁)	-19.12%
Thromboxane A ₂ (h)	11.30%
Growth Factors/Hormones	
Corticotropin Releasing Factor, Non-selective	10.67%
Oxytocin	5.41%
Platelet Activating Factor, PAF	13.75%
Thyrotropin Releasing Hormone, TRH	14.11%
Brain/Gut Peptides	
Angiotensin II, AT ₁ (h)	22.25%
Angiotensin II, AT ₂	15.03%
Bradykinin, BK ₂	24.02%
Cholecystokinin, CCK ₁ (CCKA)	-10.75%
Cholecystokinin, CCK ₂ (CCKB)	4.17%
Endothelin, ET-A (h)	-2.53%
Endothelin, ET-B (h)	21.03%
Galanin, Non-Selective	-2.38%
Neurokinin, NK ₁	8.40%
Neurokinin, NK ₂ (NKA) (h)	23.17%
Neurokinin, NK ₃ (NKB)	-6.89%
Vasoactive Intestinal Peptide, Non-selective	18.80%
Vasopressin 1	-8.42%
Enzymes	
Decarboxylase, Glutamic Acid	-12.62%

Receptor	Percent Inhibition 10 μ M
Esterase, Acetylcholine	9.27%
Oxidase, MAO-A, Peripheral	-10.15%
Oxidase, MAO-B, Peripheral	-3.95%
Transferase, Choline Acetyl	7.43%
ADME-Tox	
Metbolism, Cytochrome P450, CYP1A2 (h)	68.51%
Metabolism, Cytochrom P450, CYP2A6 (h)	-0.76%
Metabolism, Cytochrome P450, CYP2C19 (h)	21.49%
Metabolism, Cytochrome P450, CYP2C9-1 (h)	28.48%
Metabolism, Cytochrome P450, CYP2D6 (h)	1.72%
Metabolism, Cytochrome P450, CYP2E1 (h)	2.11%
Metabolism, Cytochrome P450, CYP3A4 (h)	9.16%

Supplementary Table 2. Kinome wide profiling results for LDN/OSU-0212320.

*A total of 230 kinases were tested. Only kinases with $\geq 10\%$ inhibition at 10 μM are shown.

$\geq 10\%*$	Mean Percent Inhibition		
	Kinase	Conc. Tested (μM)	LDN-212320
	ABL1	1	2
	ABL1	10	21
	CK1-GAMMA3	1	5
	CK1-GAMMA3	10	12
	EPH-A1	1	7
	EPH-A1	10	37
	EPHA2	1	2
	EPHA2	10	24
	EPH-A4	1	5
	EPH-A4	10	44
	EPH-A5	1	7
	EPH-A5	10	44
	EPH-A8	1	1
	EPH-A8	10	12
	EPH-B1	1	4
	EPH-B1	10	33
	EPHB2	1	8
	EPHB2	10	34
	EPHB4	1	1
	EPHB4	10	25
	FGR	1	8
	FGR	10	14
	FYN	1	14
	FYN	10	23
	HCK	1	1
	HCK	10	22
	KIT	1	14
	KIT	10	34
	KIT-D816V	1	0
	KIT-D816V	10	10
	LYNA	1	4
	LYNA	10	12
	LYNB	1	4

LYNB	10	16
MKNK1	1	3
MKNK1	10	10
MSK2	1	10
MSK2	10	8
PDGFR-ALPHA	1	5
PDGFR-ALPHA	10	23
RET	1	14
RET	10	4
SPHK1	1	-1
SPHK1	10	12
SPHK2	1	1
SPHK2	10	32
TEC	1	3
TEC	10	16
TXK	1	2
TXK	10	25
YES	1	3
YES	10	21

Supplementary Table 3. Kinome wide profiling list

No.	Kinase	Kinase Name
1	ABL1	c-abl oncogene 1, non-receptor tyrosine kinase
2	ABL-T315I	c-abl oncogene 1, non-receptor tyrosine kinase (mutant variant)
3	AKT1	v-akt murine thymoma viral oncogene homolog 1
4	AKT2	v-akt murine thymoma viral oncogene homolog 2
5	AKT3	v-akt murine thymoma viral oncogene homolog 3 (protein kinase B, gamma)
6	ALK	anaplastic lymphoma receptor tyrosine kinase
7	ALK4	activin A receptor, type IB
8	AMPKA1 (A1B1G1)	protein kinase, AMP-activated, alpha 1 catalytic subunit
9	AMPKA2 (A2B1G1)	protein kinase, AMP-activated, alpha 2 catalytic subunit
10	ARG	v-abl Abelson murine leukemia viral oncogene homolog 2
11	ARK5	NUAK family, SNF1-like kinase, 1
12	AuroraA	aurora kinase A
13	AuroraB	aurora kinase B
14	AuroraC	aurora kinase C
15	AXL	AXL receptor tyrosine kinase
16	BLK	B lymphoid tyrosine kinase
17	BMX	BMX non-receptor tyrosine kinase
18	BRAF	v-raf murine sarcoma viral oncogene homolog B1
19	BRK	PTK6 protein tyrosine kinase 6
20	BRSK1	BR serine/threonine kinase 1
21	BRSK2	BR serine/threonine kinase 2
22	BTK	Bruton agammaglobulinemia tyrosine kinase
23	CAMK1D	calcium/calmodulin-dependent protein kinase ID
24	CAMK2A	calcium/calmodulin-dependent protein kinase II alpha
25	CAMK2D	calcium/calmodulin-dependent protein kinase II delta
26	CAMK4	calcium/calmodulin-dependent protein kinase IV

27	CDK1/cyclinB	cyclin-dependent kinase 1
28	CDK2/cyclinA	cyclin-dependent kinase 2
29	CDK2/cyclinE	cyclin-dependent kinase 2
30	CDK3/cyclinE	cyclin-dependent kinase 3
31	CDK4/cyclinD	cyclin-dependent kinase 4
32	CDK5/p35	cyclin-dependent kinase 5
33	CDK6/cyclinD3	cyclin-dependent kinase 6
34	CHEK1	CHK1 checkpoint human homolog (S. pombe)
35	CHEK2	CHK2 checkpoint human homolog (S. pombe)
36	CK1a	casein kinase 1, alpha 1
37	CK1g1	casein kinase 1, gamma 1
38	CK1g2	casein kinase 1, gamma 2
39	CK1g3	casein kinase 1, gamma 3
40	CK2	casein kinase 2, alpha 1 polypeptide
41	CLK2	CDC-like kinase 2
42	CLK3	CDC-like kinase 3
43	CRAF	v-raf-1 murine leukemia viral oncogene homolog 1
44	CSK	c-src tyrosine kinase
45	DAPK1	death-associated protein kinase 1
46	DCAMKL2	doublecortin-like kinase 2
47	DDR2	discoidin domain receptor tyrosine kinase 2
48	DYRK1A	dual-specificity tyrosine-(Y)-phosphorylation regulated kinase 1A
49	DYRK1B	dual-specificity tyrosine-(Y)-phosphorylation regulated kinase 1B
50	DYRK2	dual-specificity tyrosine-(Y)-phosphorylation regulated kinase 2
51	DYRK3	dual-specificity tyrosine-(Y)-phosphorylation regulated kinase 3
52	DYRK4	dual-specificity tyrosine-(Y)-phosphorylation regulated kinase 4
53	EGFR	epidermal growth factor receptor
54	EGFR-T790M	epidermal growth factor receptor (mutant variant)

55	EPHA1	EPH receptor A1
56	EPHA2	EPH receptor A2
57	EPHA3	EPH receptor A3
58	EPHA4	EPH receptor A4
59	EPHA5	EPH receptor A5
60	EPHA8	EPH receptor A8
61	EPHB1	EPH receptor B1
62	EPHB2	EPH receptor B2
63	EPHB3	EPH receptor B3
64	EPHB4	EPH receptor B4
65	ERBB2	v-erb-b2 erythroblastic leukemia viral oncogene homolog 2, neuro/glioblastoma derived oncogene homolog (avian)
66	ERBB4	v-erb-a erythroblastic leukemia viral oncogene homolog 4 (avian)
67	FER	fer (fps/fes related) tyrosine kinase
68	FES	feline sarcoma oncogene
69	FGFR1	fibroblast growth factor receptor 1
70	FGFR2	fibroblast growth factor receptor 2
71	FGFR3	fibroblast growth factor receptor 3
72	FGFR4	fibroblast growth factor receptor 4
73	FGR	Gardner-Rasheed feline sarcoma viral (v-fgr) oncogene homolog
74	FLT1	fms-related tyrosine kinase 1 (vascular endothelial growth factor/vascular permeability factor receptor)
75	FLT3	fms-related tyrosine kinase 3
76	FLT4	fms-related tyrosine kinase 4
77	FMS	colony stimulating factor 1 receptor
78	FYN	FYN oncogene related to SRC, FGR, YES
79	GRK6	G protein-coupled receptor kinase 6
80	GRK7	G protein-coupled receptor kinase 7
81	GSK3A	glycogen synthase kinase 3 alpha
82	GSK3B	glycogen synthase kinase 3 beta

83	Haspin	germ cell associated 2 (haspin)
84	HCK	hemopoietic cell kinase
85	HIPK1	homeodomain interacting protein kinase 1
86	HIPK2	homeodomain interacting protein kinase 2
87	HIPK3	homeodomain interacting protein kinase 3
88	HIPK4	homeodomain interacting protein kinase 4
89	IGF1R	insulin-like growth factor 1 receptor
90	IKKA	conserved helix-loop-helix ubiquitous kinase
91	IKKB	inhibitor of kappa light polypeptide gene enhancer in B-cells, kinase beta
92	IKKE	inhibitor of kappa light polypeptide gene enhancer in B-cells, kinase epsilon
93	INSR	insulin receptor
94	IRAK4	interleukin-1 receptor-associated kinase 4
95	IRR	insulin receptor-related receptor
96	ITK	IL2-inducible T-cell kinase
97	JAK1	Janus kinase 1
98	JAK2	Janus kinase 2
99	JAK3	Janus kinase 3
100	JNK1	mitogen-activated protein kinase 8
101	JNK2	mitogen-activated protein kinase 9
102	JNK3	mitogen-activated protein kinase 10
103	KDR	kinase insert domain receptor (a type III receptor tyrosine kinase)
104	KIT	v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog
105	KIT-D816V	v-kit Hardy-Zuckerman 4 feline sarcoma viral oncogene homolog (mutant variant)
106	LATS2	LATS, large tumor suppressor, homolog 2 (Drosophila)
107	LCK	lymphocyte-specific protein tyrosine kinase
108	LOK	serine/threonine kinase 10
109	LRRK2	leucine-rich repeat kinase 2
110	LTK	leukocyte receptor tyrosine kinase

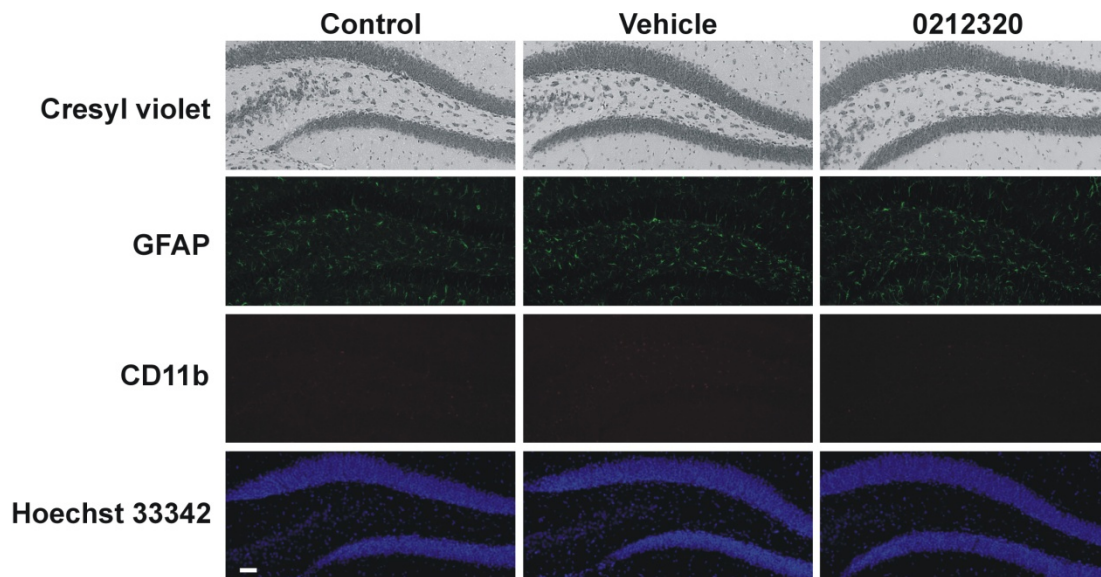
111	LYNA	v-yes-1 Yamaguchi sarcoma viral related oncogene homolog, isoform A
112	LYNB	v-yes-1 Yamaguchi sarcoma viral related oncogene homolog, isoform B
113	MAP4K2	mitogen-activated protein kinase kinase kinase kinase 2
114	MAP4K4	mitogen-activated protein kinase kinase kinase kinase 4
115	MAP4K5	mitogen-activated protein kinase kinase kinase kinase 5
116	MAPK1	mitogen-activated protein kinase 1
117	MAPK3	mitogen-activated protein kinase 3
118	MAPKAPK2	mitogen-activated protein kinase-activated protein kinase 2
119	MAPKAPK3	mitogen-activated protein kinase-activated protein kinase 3
120	MARK1	MAP/microtubule affinity-regulating kinase 1
121	MARK3	MAP/microtubule affinity-regulating kinase 3
122	MARK4	MAP/microtubule affinity-regulating kinase 4
123	MEK1	mitogen-activated protein kinase kinase 1
124	MELK	maternal embryonic leucine zipper kinase
125	MER	c-mer proto-oncogene tyrosine kinase
126	MET	met proto-oncogene (hepatocyte growth factor receptor)
127	MINK	misshapen-like kinase 1
128	MKNK1	MAP kinase interacting serine/threonine kinase 1
129	MNK2	MAP kinase interacting serine/threonine kinase 2
130	MRCKA	CDC42 binding protein kinase alpha (DMPK-like)
131	MRCKB	CDC42 binding protein kinase beta (DMPK-like)
132	MSK1	ribosomal protein S6 kinase, 90kDa, polypeptide 5
133	MSK2	ribosomal protein S6 kinase, 90kDa, polypeptide 4
134	MSSK1	SRSF protein kinase 3
135	MST1	serine/threonine kinase 4
136	MST2	serine/threonine kinase 3
137	MST3	serine/threonine kinase 24
138	MST4	MST3 and SOK1-Related Kinase

139	MUSK	muscle, skeletal, receptor tyrosine kinase
140	NEK1	NIMA (never in mitosis gene a)-related kinase 1
141	NEK2	NIMA (never in mitosis gene a)-related kinase 2
142	NEK6	NIMA (never in mitosis gene a)-related kinase 6
143	NEK7	NIMA (never in mitosis gene a)-related kinase 7
144	NEK9	NIMA (never in mitosis gene a)- related kinase 9
145	P38a	mitogen-activated protein kinase 14
146	P38b	mitogen-activated protein kinase 11
147	P38g	mitogen-activated protein kinase 12
148	P38d	mitogen-activated protein kinase 13
149	P70S6K1	ribosomal protein S6 kinase, 70kDa, polypeptide 1
150	PAK1	p21 protein (Cdc42/Rac)-activated kinase 1
151	PAK2	p21 protein (Cdc42/Rac)-activated kinase 2
152	PAK3	p21 protein (Cdc42/Rac)-activated kinase 3
153	PAK4	p21 protein (Cdc42/Rac)-activated kinase 4
154	PAK5	p21 protein (Cdc42/Rac)-activated kinase 7
155	PAK6	p21 protein (Cdc42/Rac)-activated kinase 6
156	PAR1Ba	MAP/microtubule affinity-regulating kinase 2
157	PASK	PAS domain containing serine/threonine kinase
158	PDGFRa	platelet-derived growth factor receptor, alpha polypeptide
159	PDGFRa-D842V	platelet-derived growth factor receptor, alpha polypeptide (mutant variant)
160	PDGFRb	platelet-derived growth factor receptor, beta polypeptide
161	PDK1	pyruvate dehydrogenase kinase, isozyme 1
162	PHKg1	phosphorylase kinase, gamma 1 (muscle)
163	PHKg2	phosphorylase kinase, gamma 2 (testis)
164	PI3Ka	phosphoinositide-3-kinase, catalytic, alpha polypeptide
165	PI3Kd	phosphoinositide-3-kinase, catalytic, delta polypeptide
166	PI4Kb	phosphatidylinositol 4-kinase, catalytic, beta

167	PIM1	pim-1 oncogene
168	PIM2	pim-2 oncogene
169	PIM3	pim-3 oncogene
170	PKA	protein kinase, cAMP-dependent, catalytic, alpha
171	PKCa	protein kinase C, alpha
172	PKCb1	protein kinase C, beta
173	PKCb2	protein kinase C, beta
174	PKCg	protein kinase C, gamma
175	PKCh	protein kinase C, eta
176	PKCq	protein kinase C, theta
177	PKCi	protein kinase C, iota
178	PKN1	protein kinase N1
179	PKN2	protein kinase N2
180	PLK1	polo-like kinase 1
181	PRAK	mitogen-activated protein kinase-activated protein kinase 5
182	PRKD1	protein kinase D1
183	PRKD2	protein kinase D2
184	PRKD3	protein kinase D3
185	PRKG1	protein kinase, cGMP-dependent, type I
186	PRKG2	protein kinase, cGMP-dependent, type II
187	PRKX	protein kinase, X-linked
188	PTK5	fyn-related kinase
189	PYK2	PTK2B protein tyrosine kinase 2 beta
190	RET	ret proto-oncogene
191	ROCK1	Rho-associated, coiled-coil containing protein kinase 1
192	ROCK2	Rho-associated, coiled-coil containing protein kinase 2
193	RON	macrophage stimulating 1 receptor (c-met-related tyrosine kinase)
194	ROS	c-ros oncogene 1 , receptor tyrosine kinase

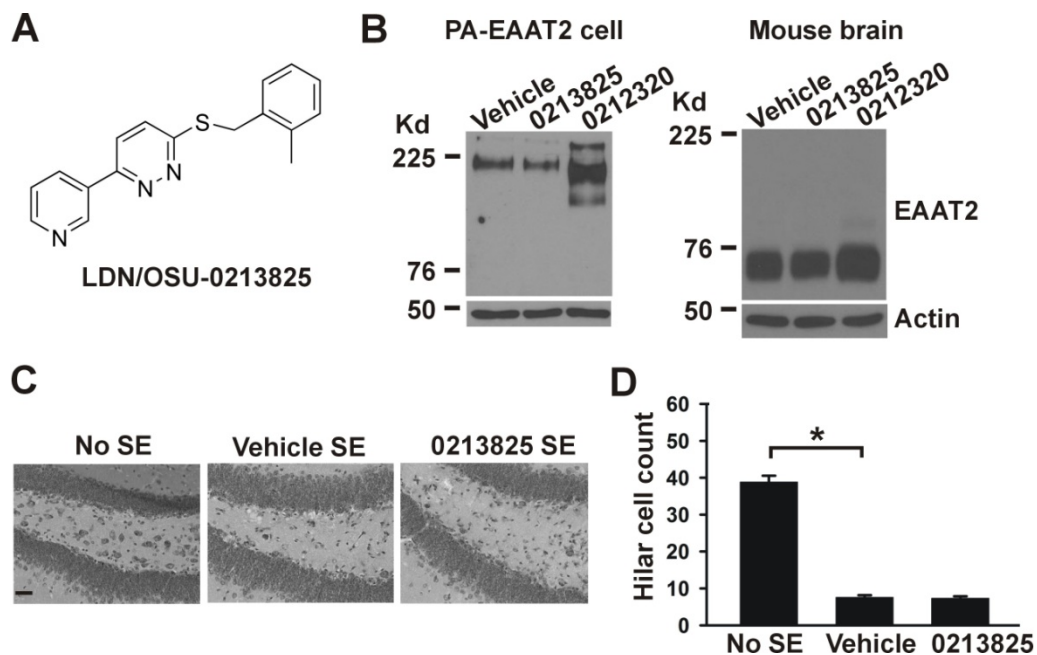
195	RSK1	ribosomal protein S6 kinase, 90kDa, polypeptide 1
196	RSK2	ribosomal protein S6 kinase, 90kDa, polypeptide 3
197	RSK3	ribosomal protein S6 kinase, 90kDa, polypeptide 2
198	RSK4	ribosomal protein S6 kinase, 90kDa, polypeptide 6
199	SGK1	serum/glucocorticoid regulated kinase 1
200	SGK2	serum/glucocorticoid regulated kinase 2
201	SGK3	serum/glucocorticoid regulated kinase family, member 3
202	SNF1LK (SIK)	salt-inducible kinase 1
203	SNF1LK2 (QIK)	salt-inducible kinase 2
204	SPHK1	sphingosine kinase 1
205	SPHK2	sphingosine kinase 2
206	SRC	v-src sarcoma (Schmidt-Ruppin A-2) viral oncogene homolog (avian)
207	SRMS	src-related kinase lacking C-terminal regulatory tyrosine and N-terminal myristylation sites
208	SRPK1	SRSF protein kinase 1
209	SRPK2	SRSF protein kinase 2
210	SYK	spleen tyrosine kinase
211	TAOK2	TAO kinase 2
212	TAOK3	TAO kinase 3
213	TBK1	TANK-binding kinase 1
214	TEC	tec protein tyrosine kinase
215	TIE2	TEK tyrosine kinase, endothelial
216	TNIK	TRAF2 and NCK interacting kinase
217	TNK1	tyrosine kinase, non-receptor, 1
218	TNK2	tyrosine kinase, non-receptor, 2
219	TRKA	neurotrophic tyrosine kinase, receptor, type 1
220	TRKB	neurotrophic tyrosine kinase, receptor, type 2
221	TRKC	neurotrophic tyrosine kinase, receptor, type 3
222	TSSK1	testis-specific serine kinase 1B

223	TSSK2	testis-specific serine kinase 2
224	TTK	TTK protein kinase
225	TXK	TXK tyrosine kinase
226	TYK2	tyrosine kinase 2
227	TYRO3	TYRO3 protein tyrosine kinase
228	YES	v-yes-1 Yamaguchi sarcoma viral oncogene homolog 1
229	ZAP70	zeta-chain (TCR) associated protein kinase 70kDa
230	ZIPK	death-associated protein kinase 3



Supplementary Figure 1

No toxicity was observed in the brain of mice after long-term treatment with LDN/OSU-0212320. Wild-type mice received 40 mg/kg of compound daily for two months. Brains were harvested for cresyl violet staining, nuclear staining with Hoechst 33342 and immunostaining with GFAP and CD11b. No neuronal loss or glial activation was found. Six animals for each group were analyzed. Scale bar: 50 μ m



Supplementary Figure 2

LDN/OSU-0213825, an inactive compound, does not provide protective effects in pilocarpine-induced status epilepticus mouse model. **(A)** Structure of LDN/OSU-0213825. **(B)** LDN/OSU-0213825 could not increase EAAT2 protein expression in PA-EAAT2 cells and mouse brains. **(C-D)** Hippocampal damage. Three sets of brains at 2 weeks post-SE were analyzed with cresyl violet staining (15-20 sections/mouse). **(C)** Representative images of dentate hilus. Scale bar: 20 μ m **(D)** Quantitative analysis of hilar cell quantity. LDN/OSU-0213825 could not protect pilocarpine-induced neuronal death. * $p < 0.05$