Glycine-gated ion channels

Converging mechanism and therapeutic potentials

Li Zhang

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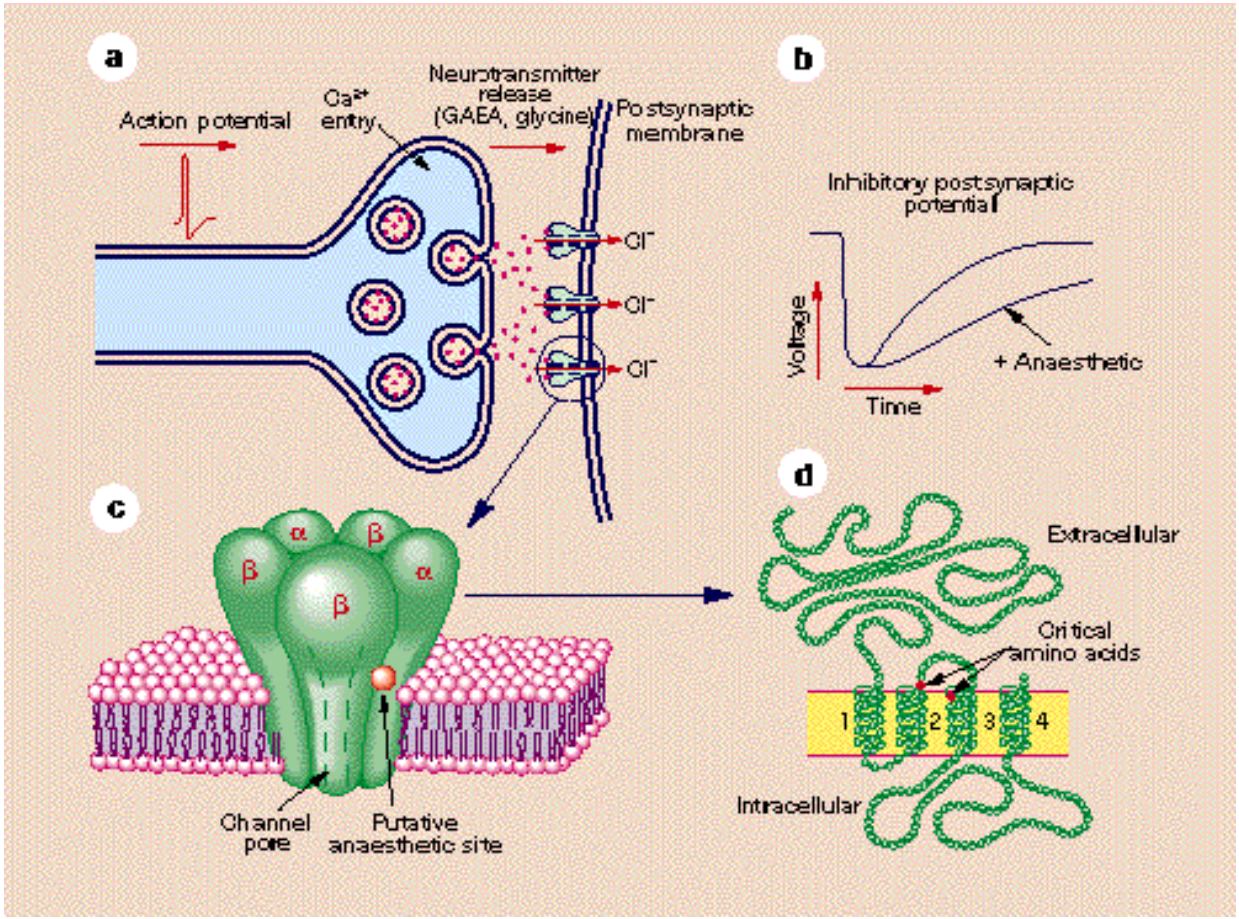
Glycine receptors are inhibitory neurotransmitter-gated ion channels

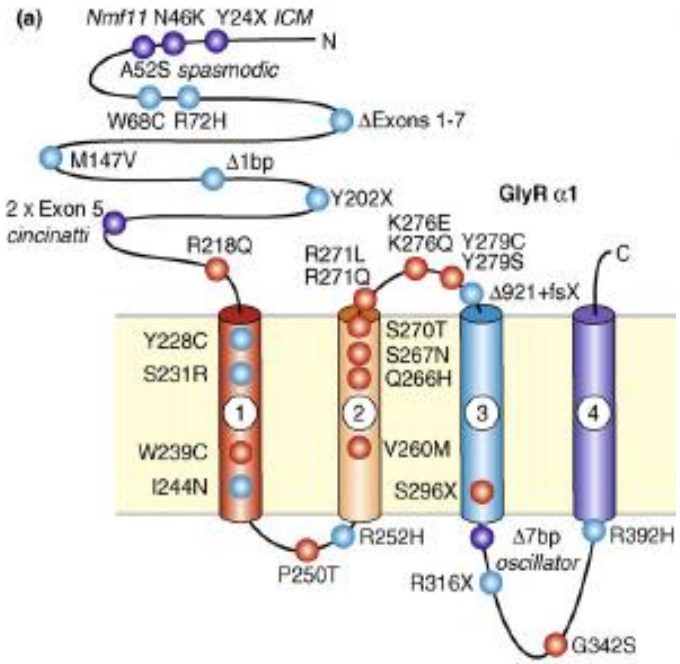


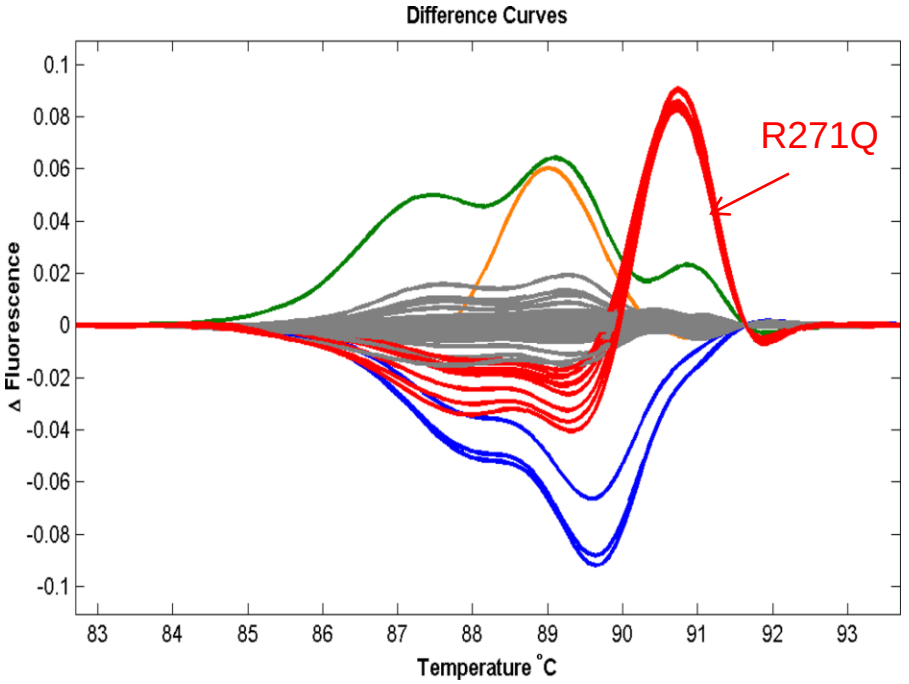
N. P. Franks and W. R. Lieb
Nature **389**, 334-335(25 September 1997)

GlyR subunits and neurological disorders

α1: abundantly expressed in sensory and motor neurons of brainstem and spinal cord, playing a critical role in neuromotor activity.

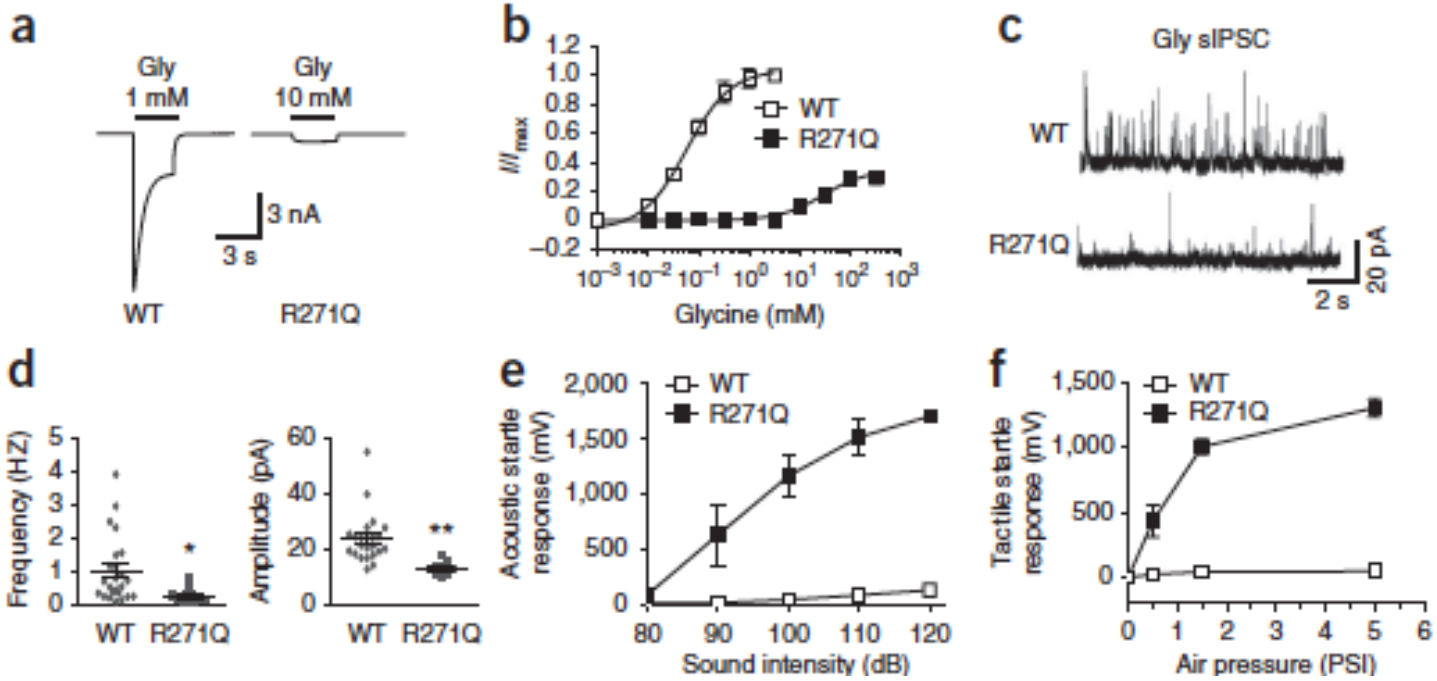
Multiple polymorphisms in the α1 GlyRs are associated with human and animal hyperekplexia (startle disease)





Current therapeutic agents in the treatment of startle disease do not target GlyRs

The R271Q point-mutation reduces glycinergic synaptic transmission in dorsal horn neurons in mice

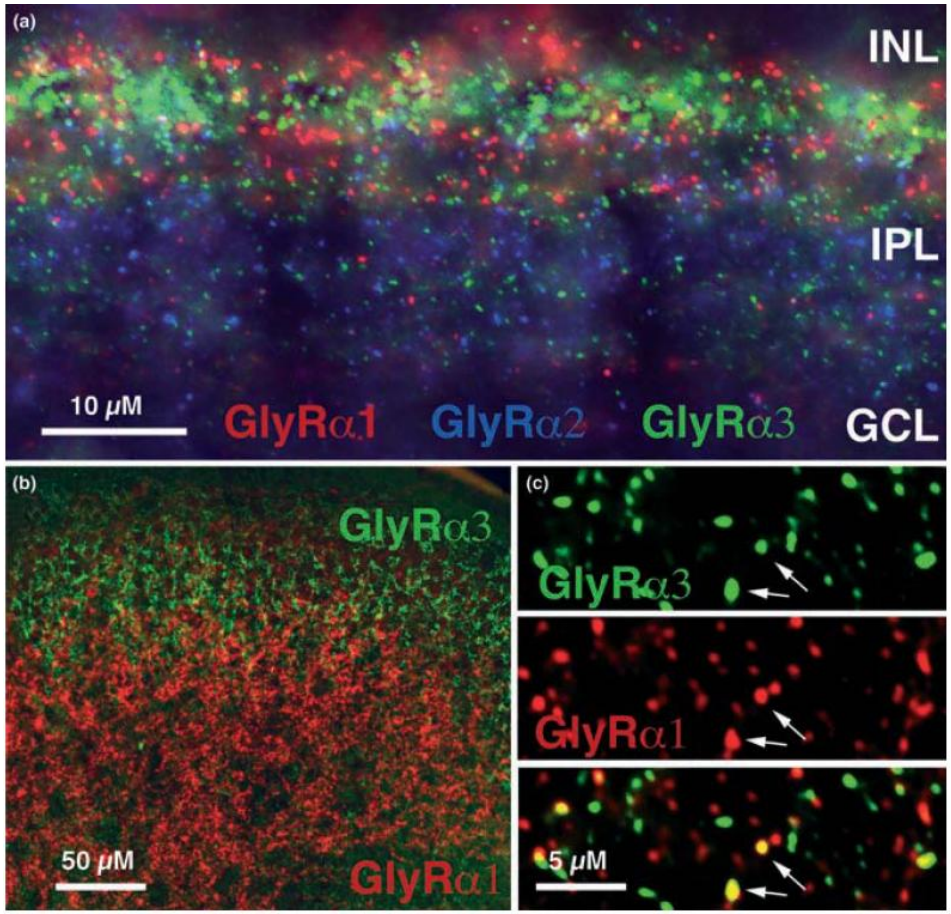


Xiong et al., Nat. Neurosci., 2014

GlyR subunits and neurological disorders

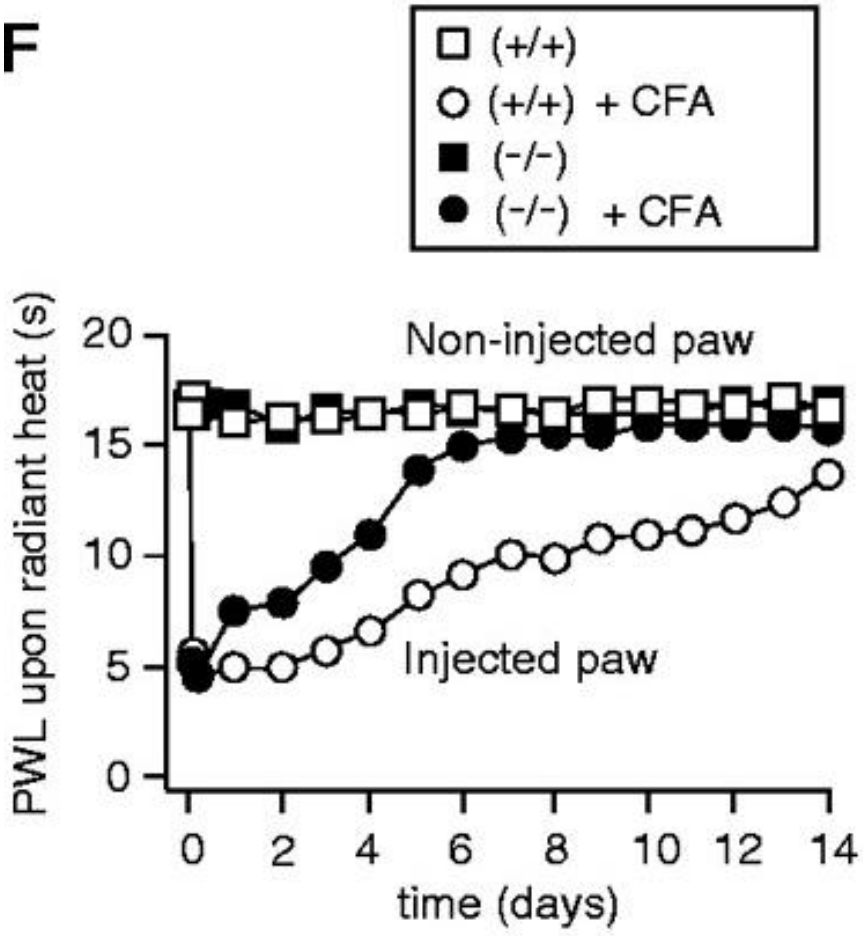
α3: highly expressed in superficial layer of the spinal dorsal horn. These receptors are involved in antinociceptive process.

Distribution of GlyR α subunits in spinal cord



Betz and Laube, 2008

α3GlyRs: an essential target for PGE2-induced chronic inflammatory pain



Harvey et al, 2005, Science

GlyRs: therapeutic orphan receptors

- No therapeutic agents are developed from GlyRs
- No subunit selective antagonist or agonist for α and β subunits

Questions to address:

- Can we find therapeutic agents in the treatment of rare and common diseases by targeting at GlyRs?

- How can these agents interact with GlyRs?
- What subtypes of GlyRs are therapeutic sensitive?
- Synaptic localization?

Marijuana was used as a anti-pain medicine in Han Dynasty (206 BCE – 220 AD)

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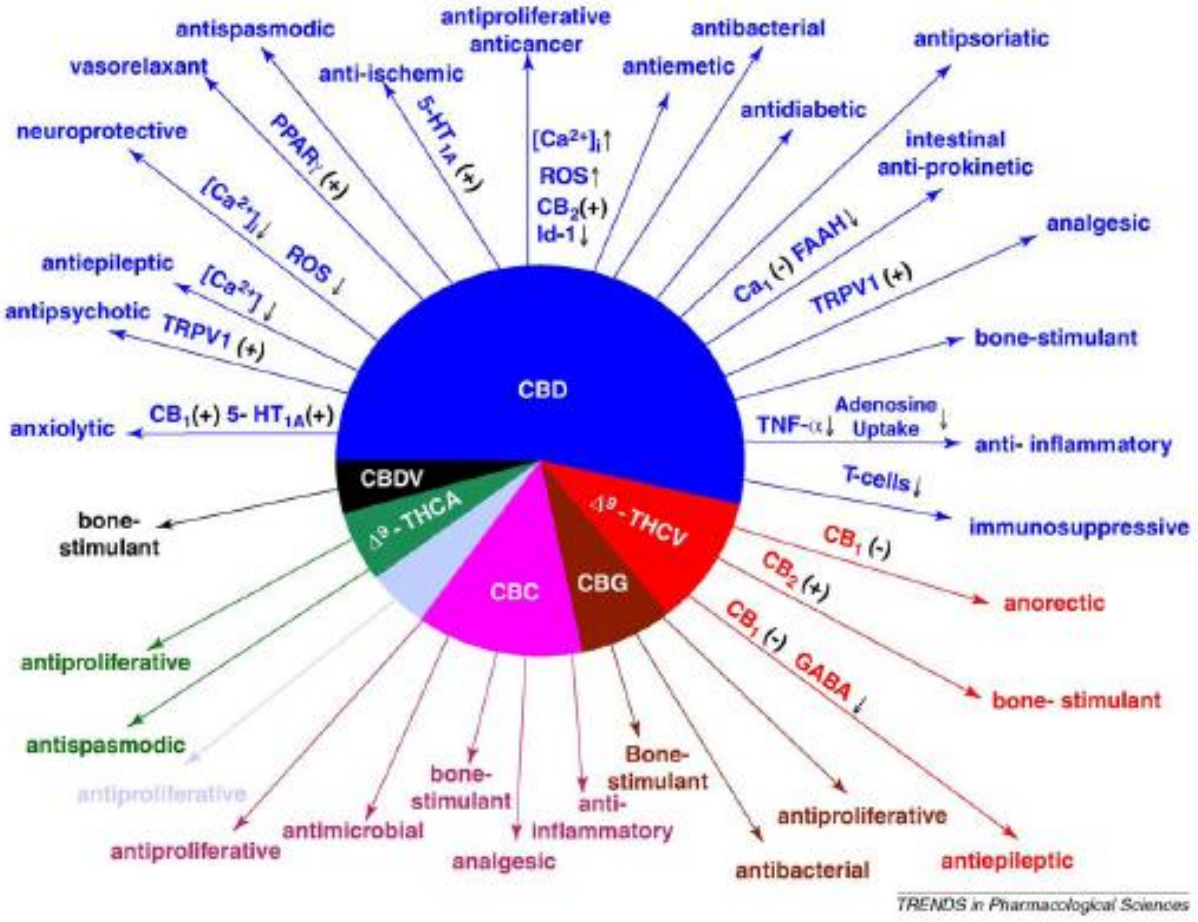
Review

Non-psychotropic plant cannabinoids: new therapeutic opportunities from an ancient herb

Angelo A. Izzo[1,4], Francesca Borrelli[1,4], Raffaele Capasso[1,4], Vincenzo Di Marzo[2,4] and Raphael Mechoulam[3]



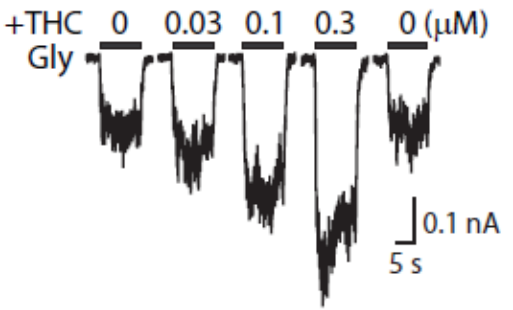
TRENDS in Pharmacological Sciences

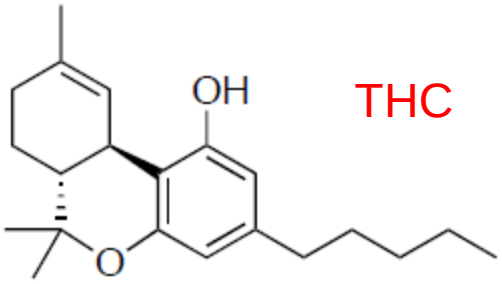
Ligand gated-ion channels are cannabinoid targets

- CB_1 receptors
- CB_2 receptors

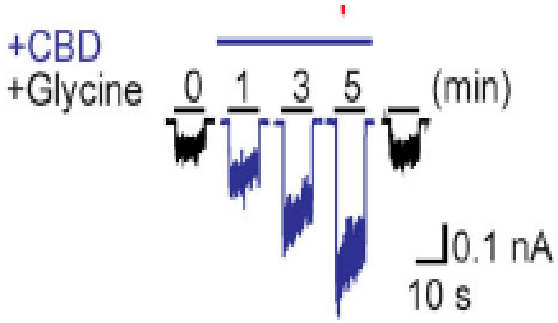
- Glycine receptors
- TRPV channels
- $GABA_A$ receptors
- $5\text{-}HT_{3A}$ receptors
- NMDA receptors

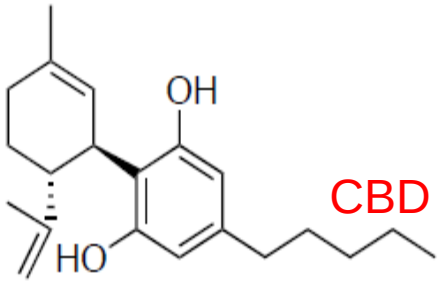
Cannabinoids potentiate GlyRs in a non CB1 and CB2 dependent mechanism





Psychoactive





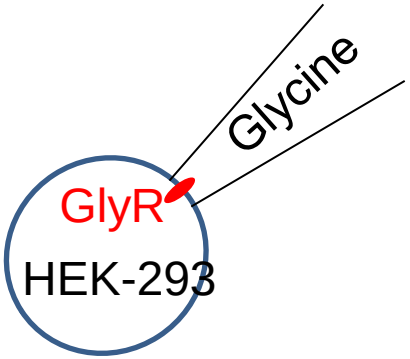
Nonpsychoactive

Xiong et al, J. Exp. Med. 2012

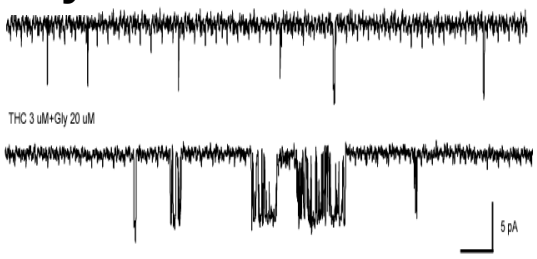
THC and CBD potentiation of GlyRs is mediated by a non CB1-dependent mechanism

Hejazi et al, 2006
Yang et al, 2010
Xiong et al, 2011, 2012
Yevenev & Zeilhofer, 2012

Single Channel Recording analysis: cannabinoid increases GlyR open probability



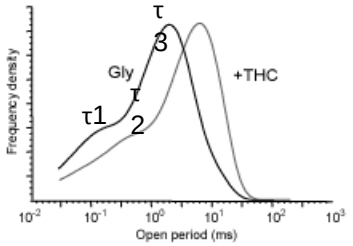
Gly

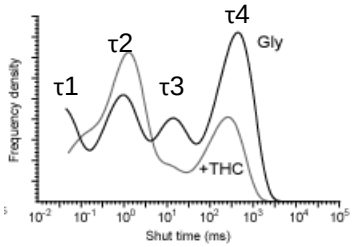


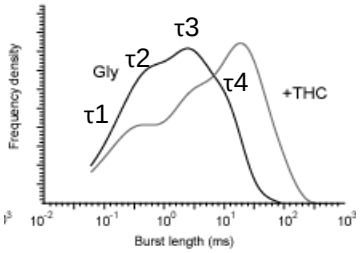
Gly+THC

Table 1. Mean fitted exponential densities

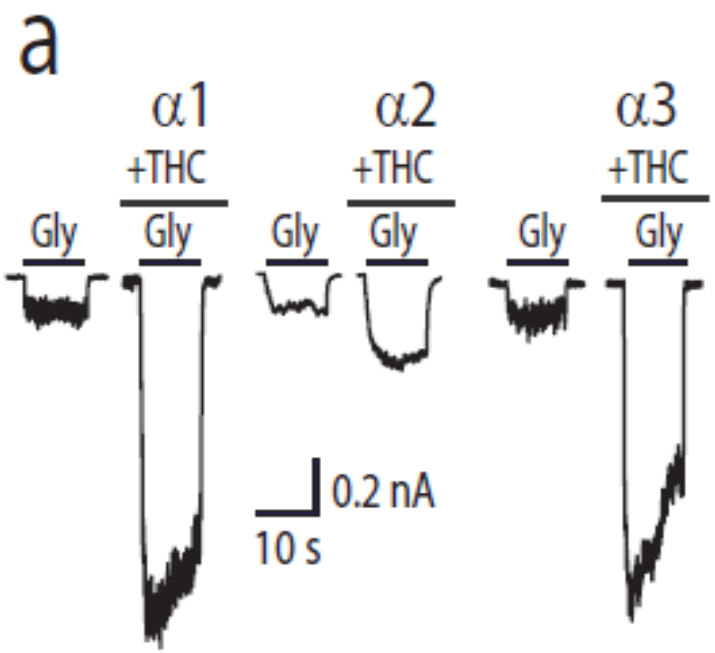
	Gly (n=7)	THC+Gly (n=12)
Open periods		
τ1 (ms)	0.099±0.075	0.063±0.033
(%)	17.4±0.8	8.2±2.0
τ2 (ms)	1.78±0.047	0.79±0.036
(%)	62.8±3.4	**18.7±1.8
τ3 (ms)	6.13±0.14	7.57±0.27
(%)	19.7±3.5	**73.1±17.9
Shut times		
τ1 (ms)	0.034±0.005	0.072±0.004
(%)	20.1±1.9	12.5±1.1
τ2 (ms)	0.85±0.043	1.1±0.038
(%)	22.2±1.0	47.7±6.1
τ3 (ms)	12.02±4.89	7.0±0.17
(%)	17.0±2.0	10.4±1.35
τ4 (ms)	457.1±43.1	*246.5±95.9
(%)	40.7±14.1	29.4±2.5
Burst length		
τ1 (ms)	0.24±0.14	0.42±0.11
(%)	23.7±3.2	16.7±1.3
τ2 (ms)	1.22±0.19	3.2±1.2
(%)	33.0±4.0	20.5±1.6
τ3 (ms)	5.70±0.22	**25.13±3.5
(%)	39.5±5.1	38.2±5.7
τ4 (ms)	21.96±1.59	**76.5±12.5
(%)	37.3±6.7	24.6±6.3



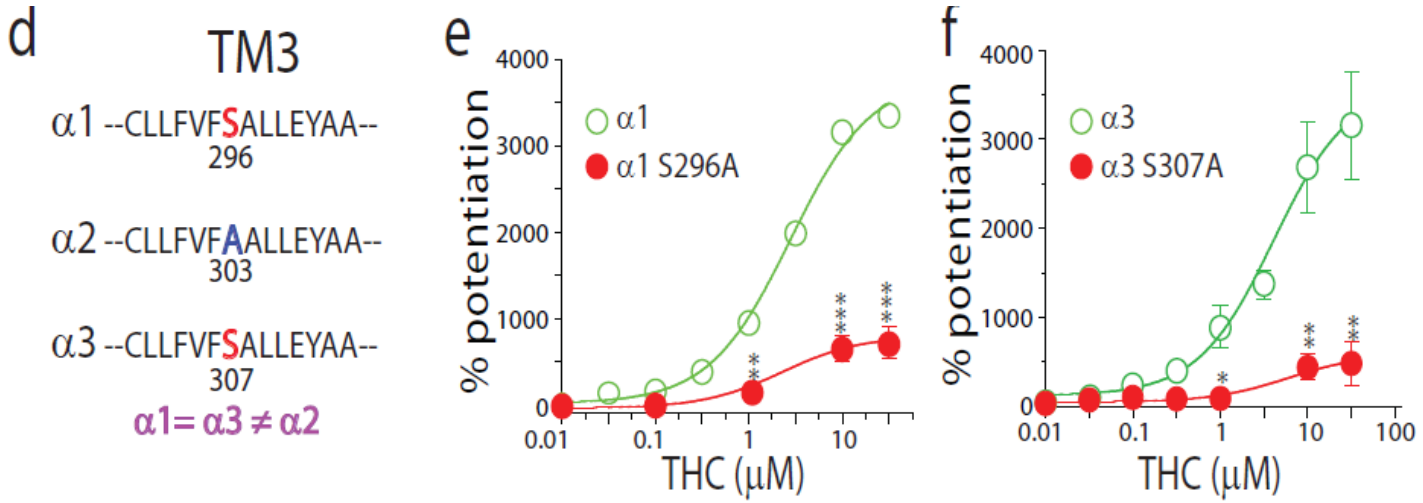


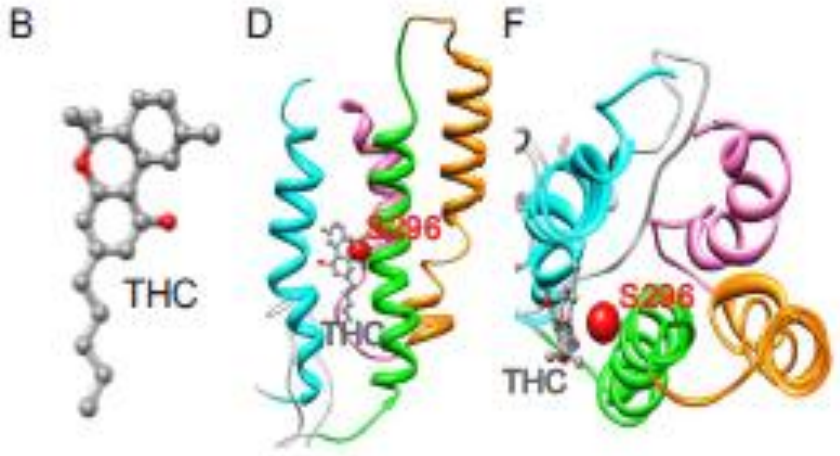


α1 and α3 GlyRs are cannabinoid high sensitive receptors



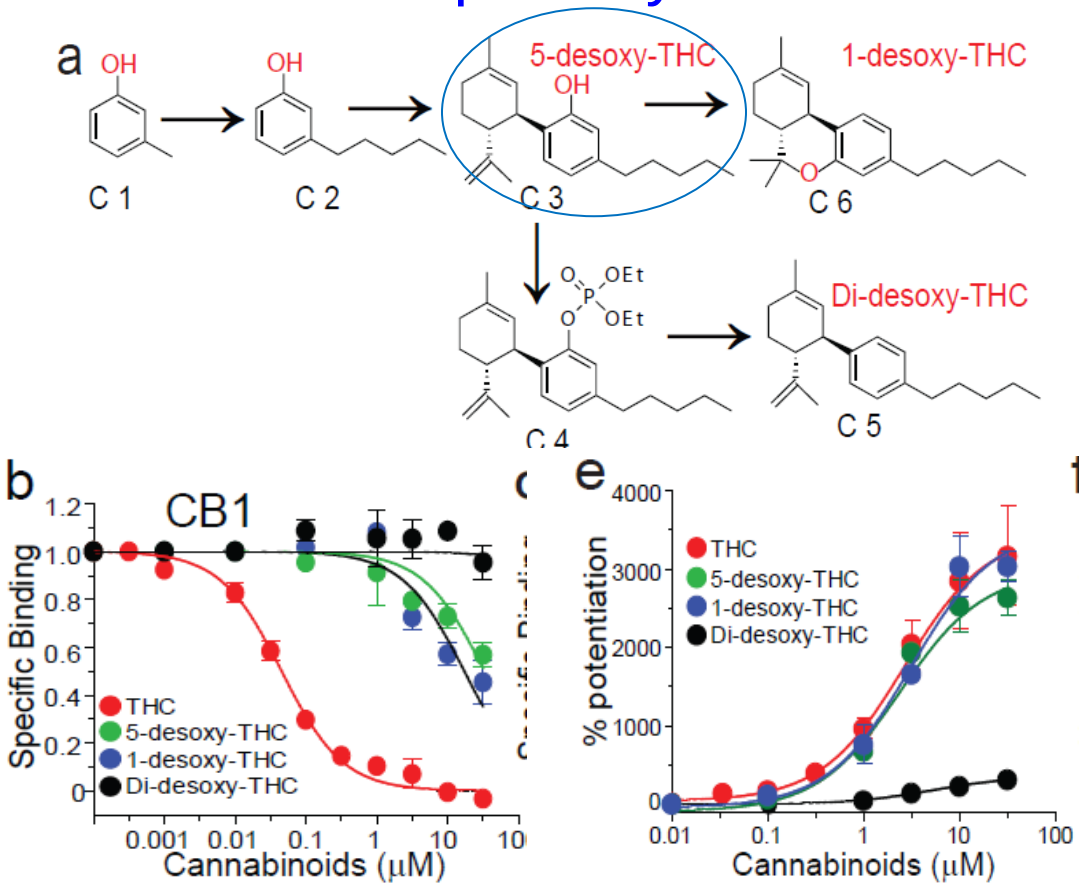
Identification a distinct site for cannabinoid potentiation of α1 and α3 GlyRs





Xiong et al, J. Neurosci. 2012

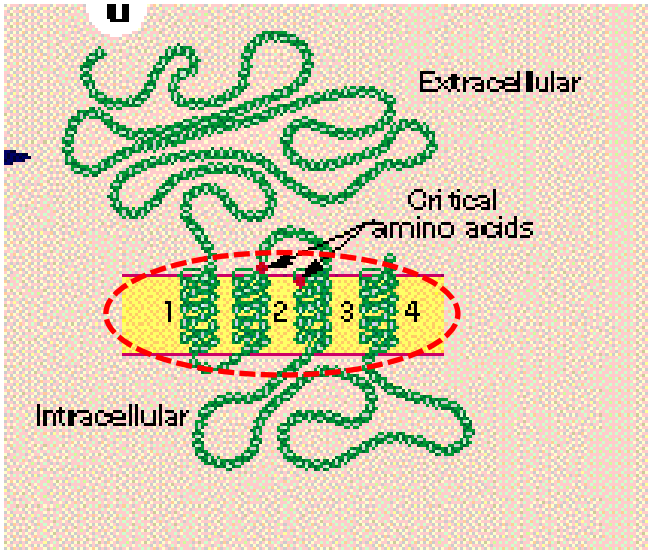
Chemical modification of THC: 5-desoxy-THC (DH-CBD) retains its potency in potentiating IGly but losses its binding potency to CB1R

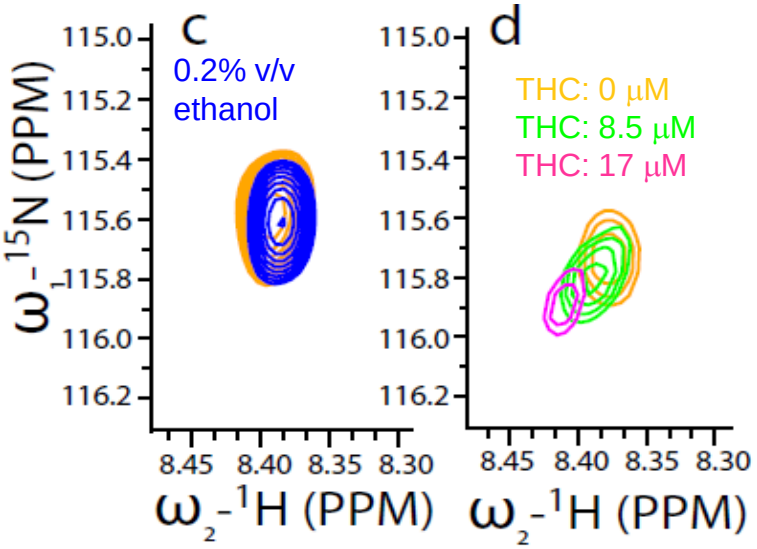


DH-CBD does not produce psychoactive effects associated with cannabis

Xiong et al, 2011, Nat. Chem. Biol.

NMR analysis: a direct interaction of cannabinoid with S296-containing domain in the α3-TM4 protein

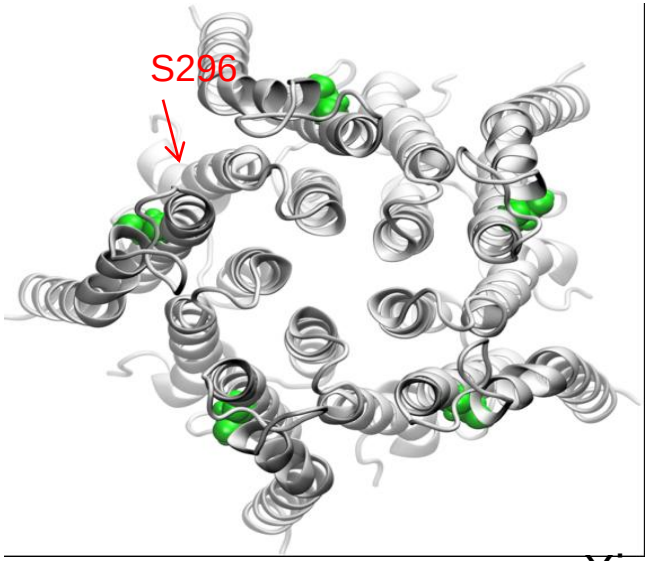


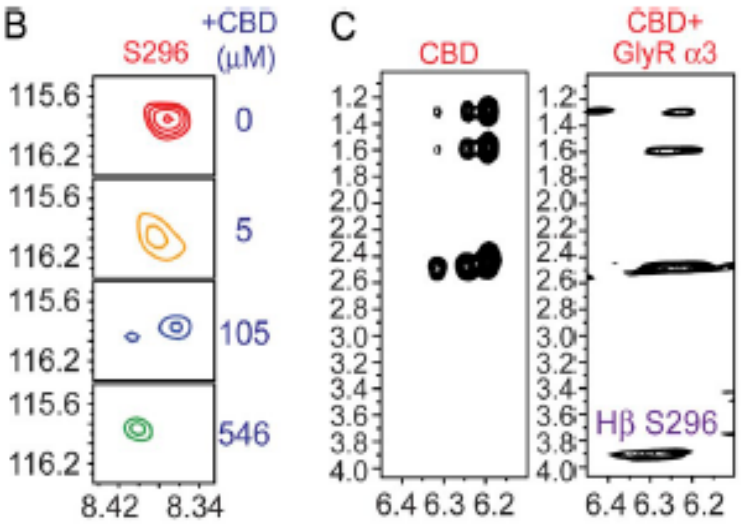


α1 GlyR

Xiong et al, 2011

Intermolecular cross peak





α3 GlyR

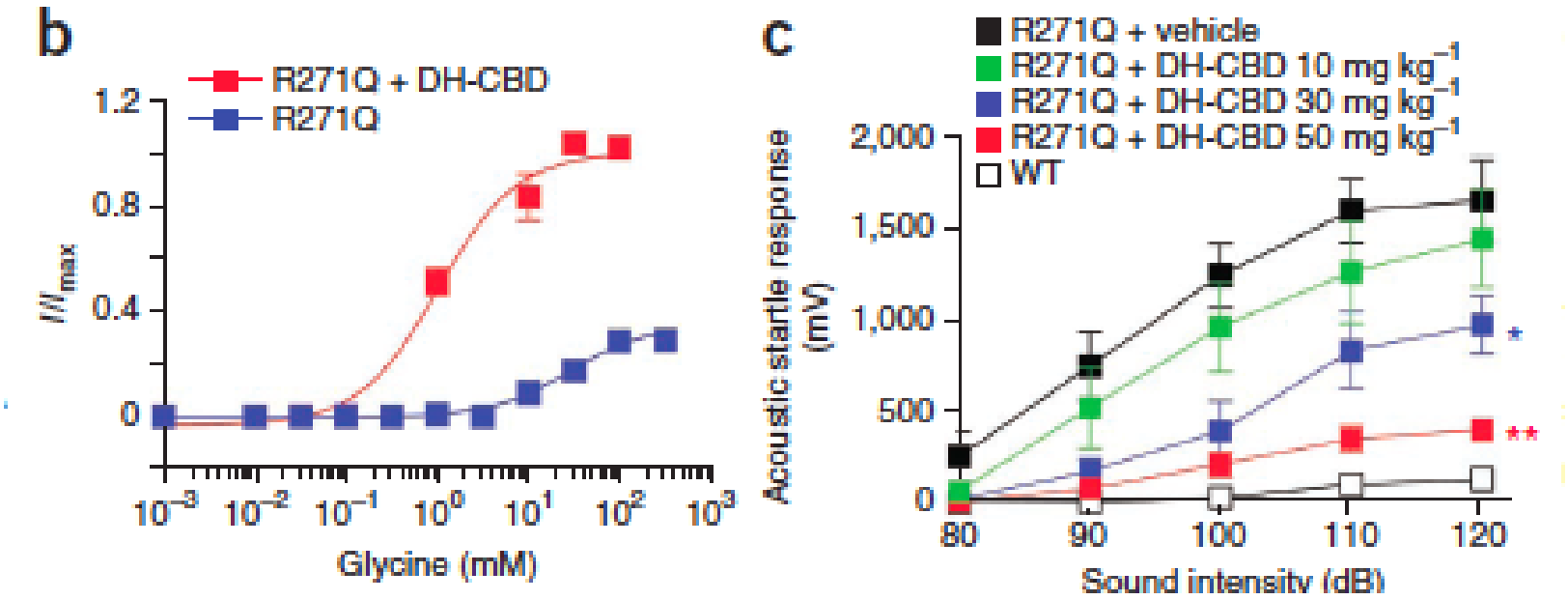
Xiong et al, 2012

Xiong et al., 2011, Nat. Chem. Biol., & 2012 J. Exp. Med.

Therapeutic application of modified cannabinoid (glycinergic cannabinoid):

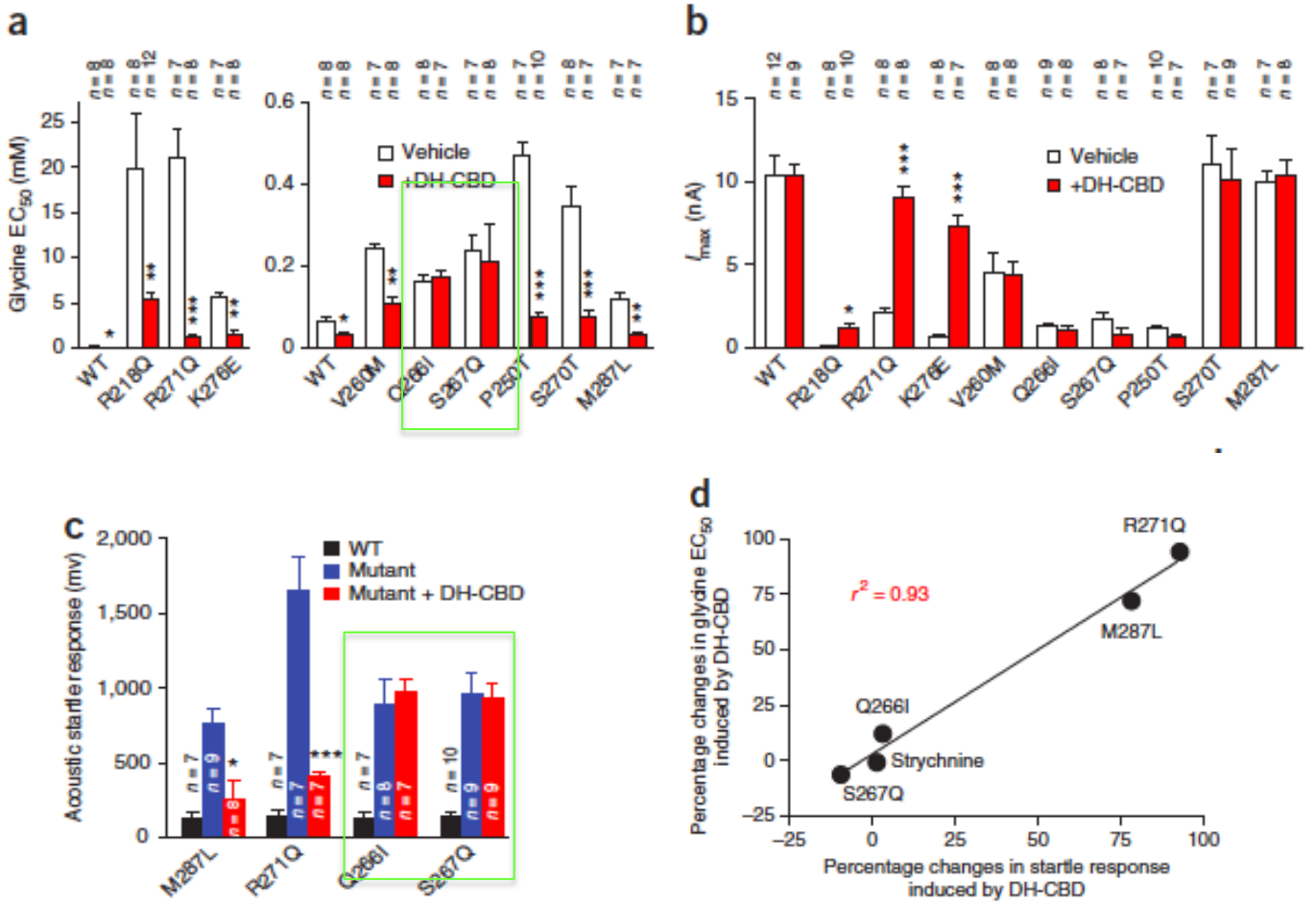
restoration of the functional deficiency of α1GlyRs
and exaggerated behavior

Glycinergic cannabinoid rescues exaggerated startle response in R271Q mutant mice



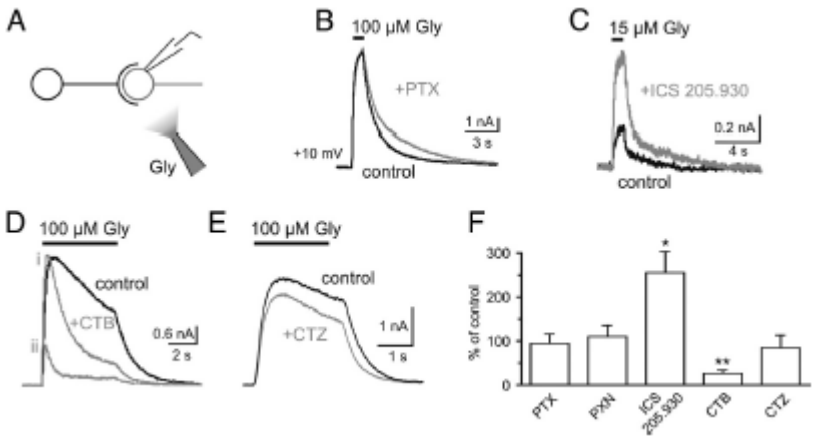
Xiong et al, 2014

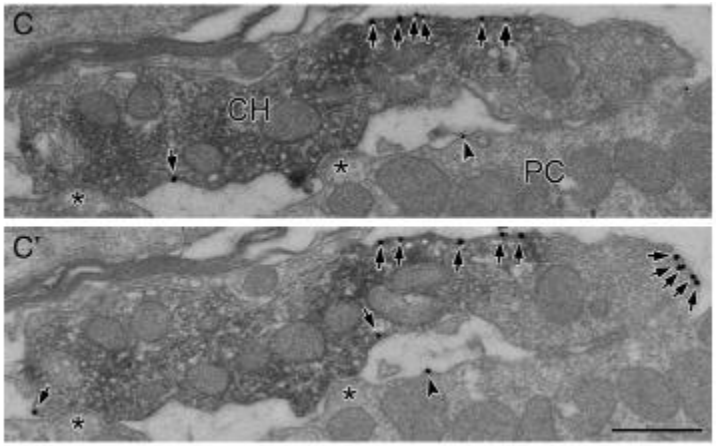
Glycinergic cannabinoid restoration of GlyR function and exaggerated startle response is genotype-specific

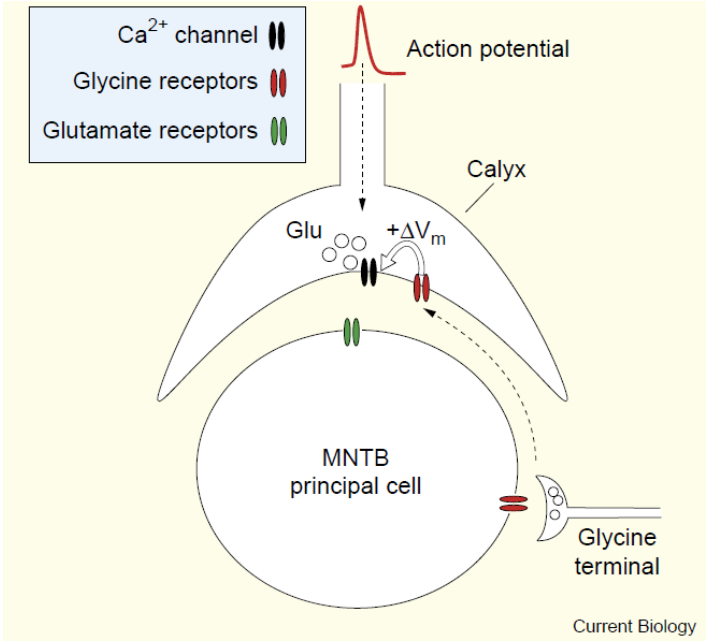


Xiong et al., 2014 Nat. Neurosci.

Presynaptic glycine receptors at brainstem calyx of held



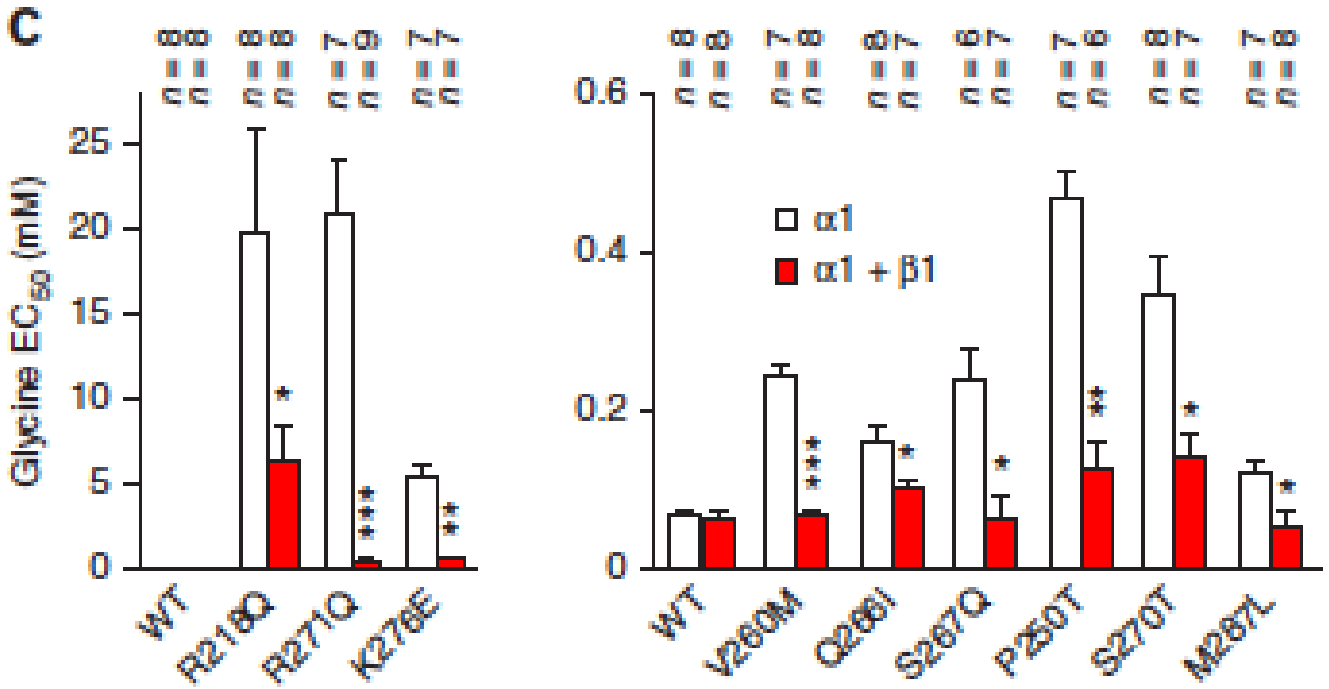




- α1GlyRs
- homomeric subunits

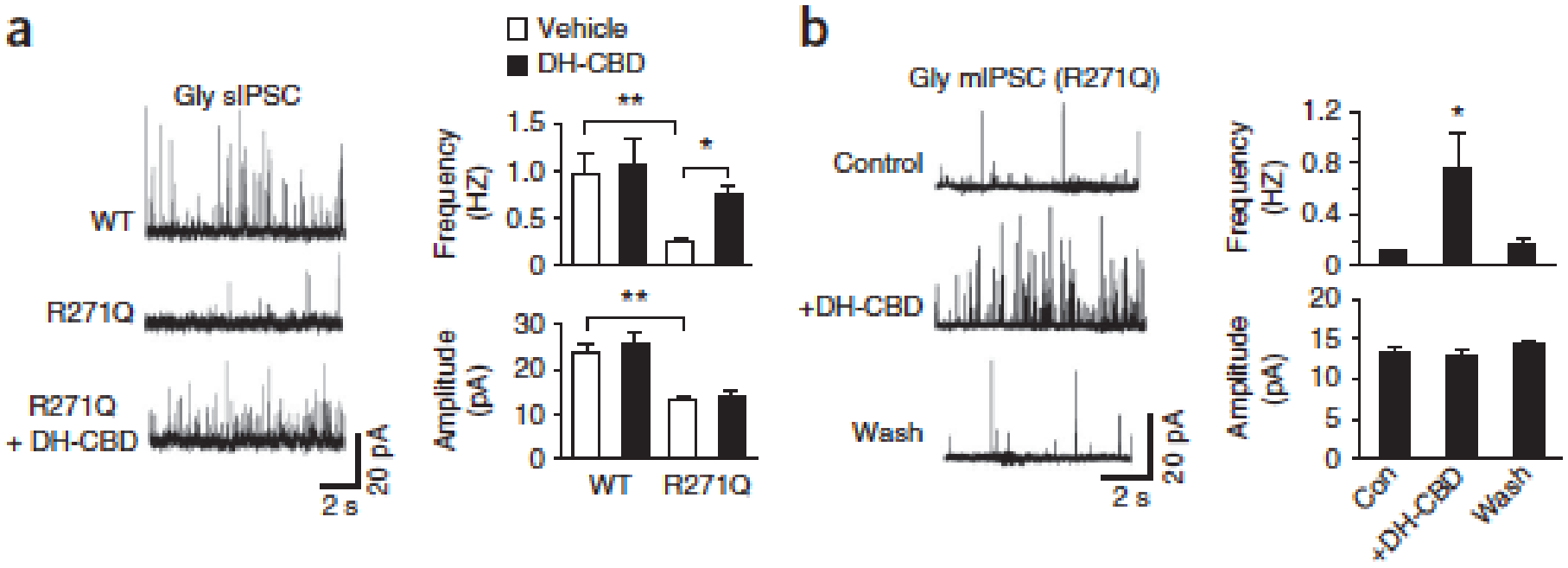
Huesic et al., 2012 J. Neurosci.

Homomeric GlyRs are more sensitive than heteromers to glycinergic cannabinoid



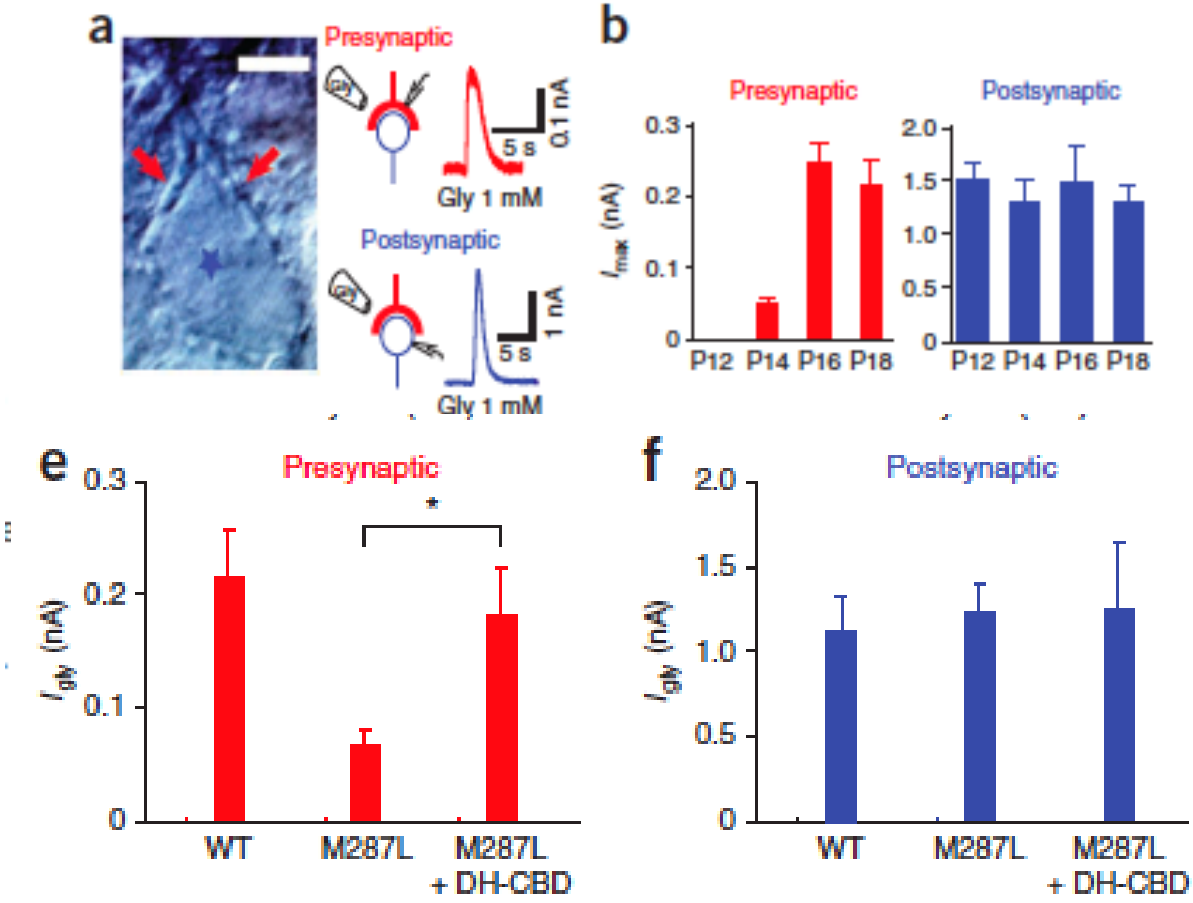
Xiong et al., 2014 Nat. Neurosci.

DH-CBD enhances diminished frequency of Gly sIPSC and mIPSCs in spinal slices of R271Q mutant mice



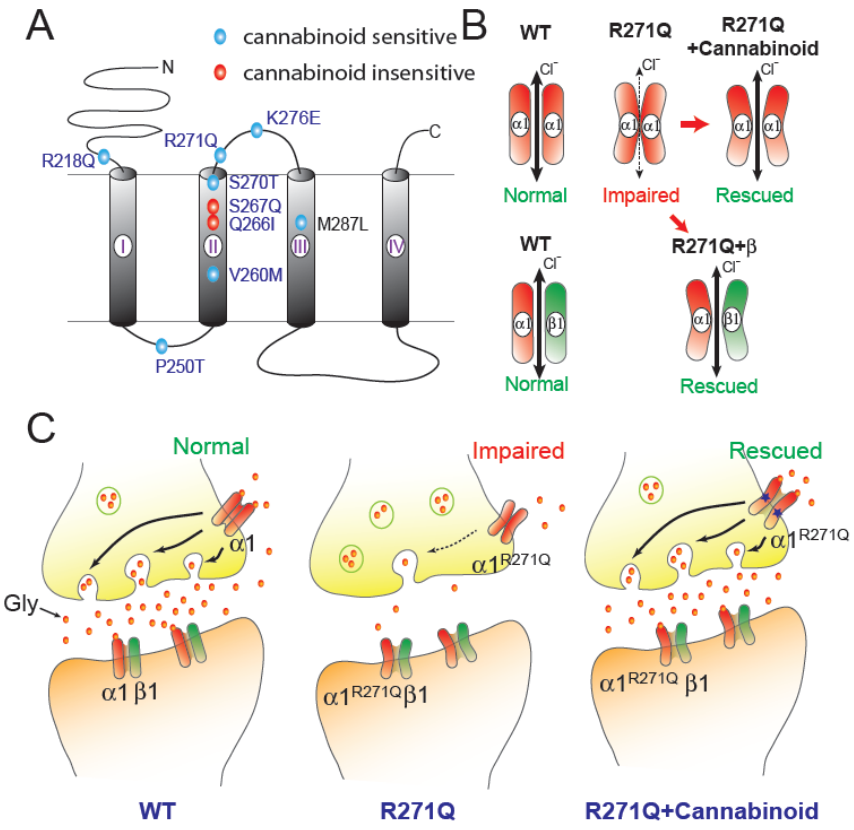
Xiong et al, 2014 Nat. Neurosci.

DH-CBD selectively rescues presynaptic GlyR deficiency in hyperekplexic mutant mice



Xiong et al, 2014, Nat. Neuroci.

Glycinergic cannabinoid in the treatment of hyperekplexia disease



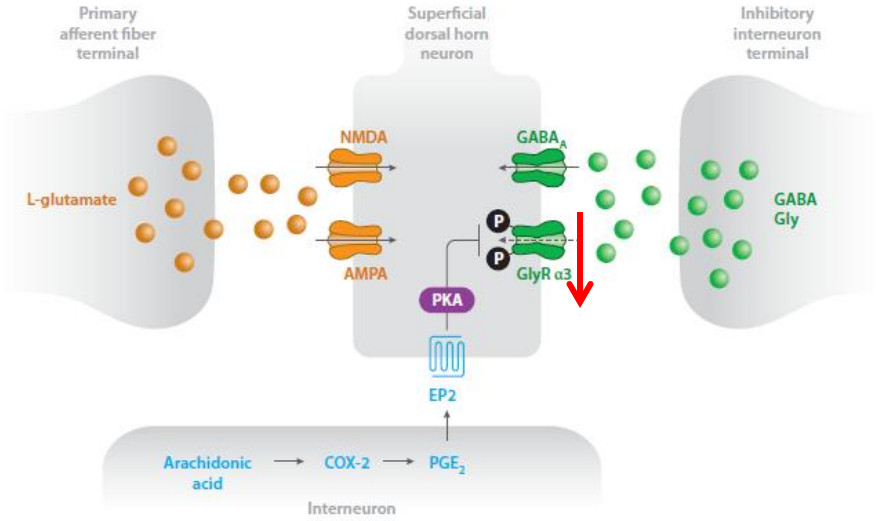
Cannabinoid sensitive α1 GlyRs

- α1 subunits with functional deficiency
- likely homomers
- mutation-site specific
- Targeting primarily presynaptic GlyRs

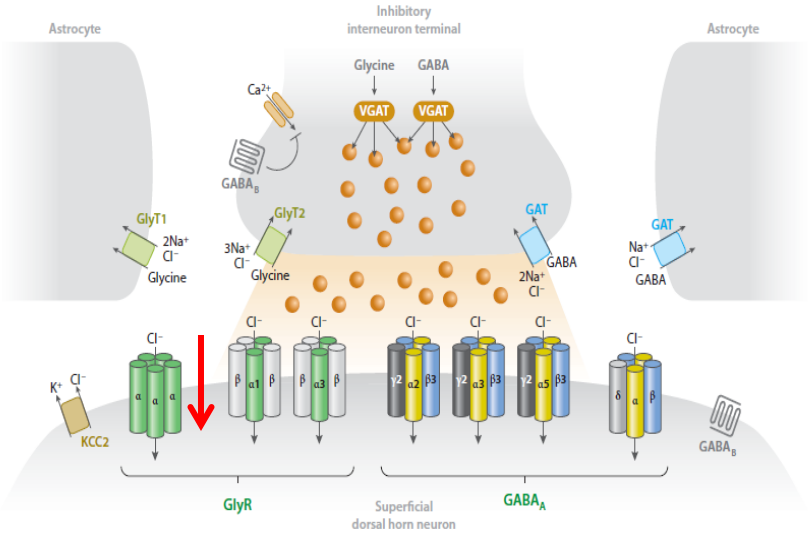
Cannabinoid sensitive GlyRs are Important therapeutic targets in sensory/motor dysfunction.

Role of spinal GlyRs in chronic pain: diminished synaptic inhibition in the dorsal horn

Inflammatory



Neuropathic



Zeilhofer, 2012

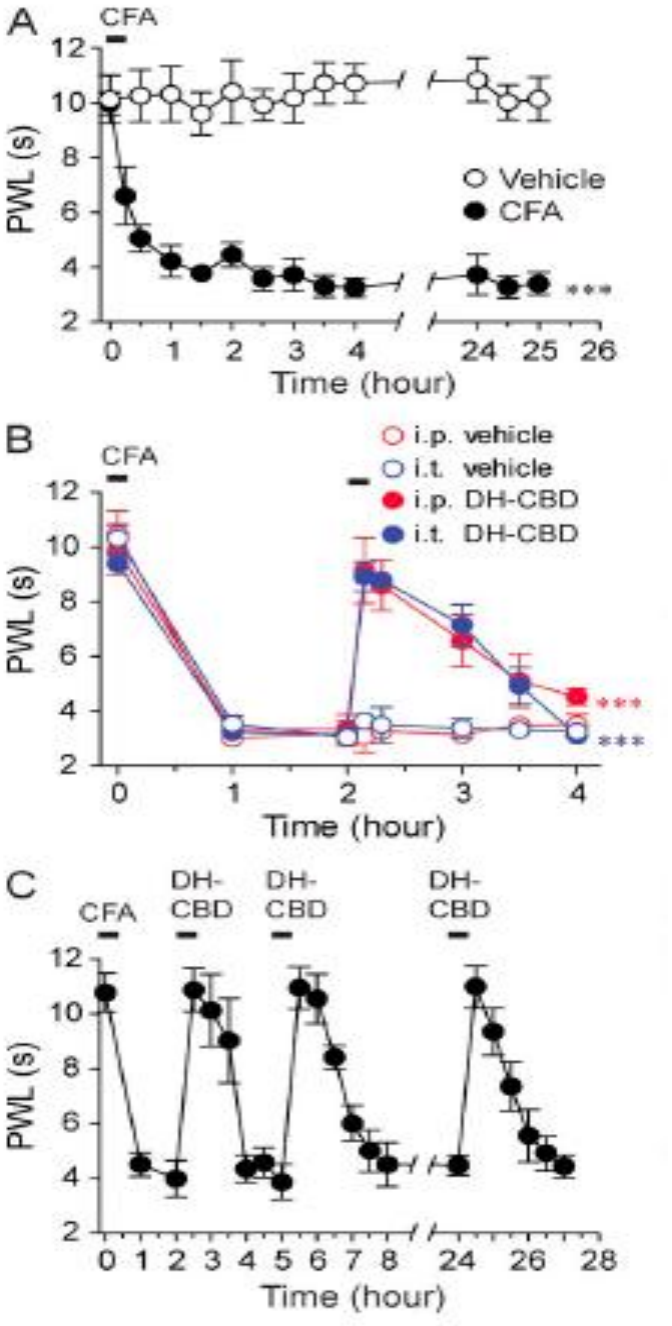
Ideas: Facilitation of glycinergic inhibition might be a particularly attractive approach through development of positive allosteric modulators.

DH-CBD can suppress acute and chronic inflammatory and neuropathic pain in rodents

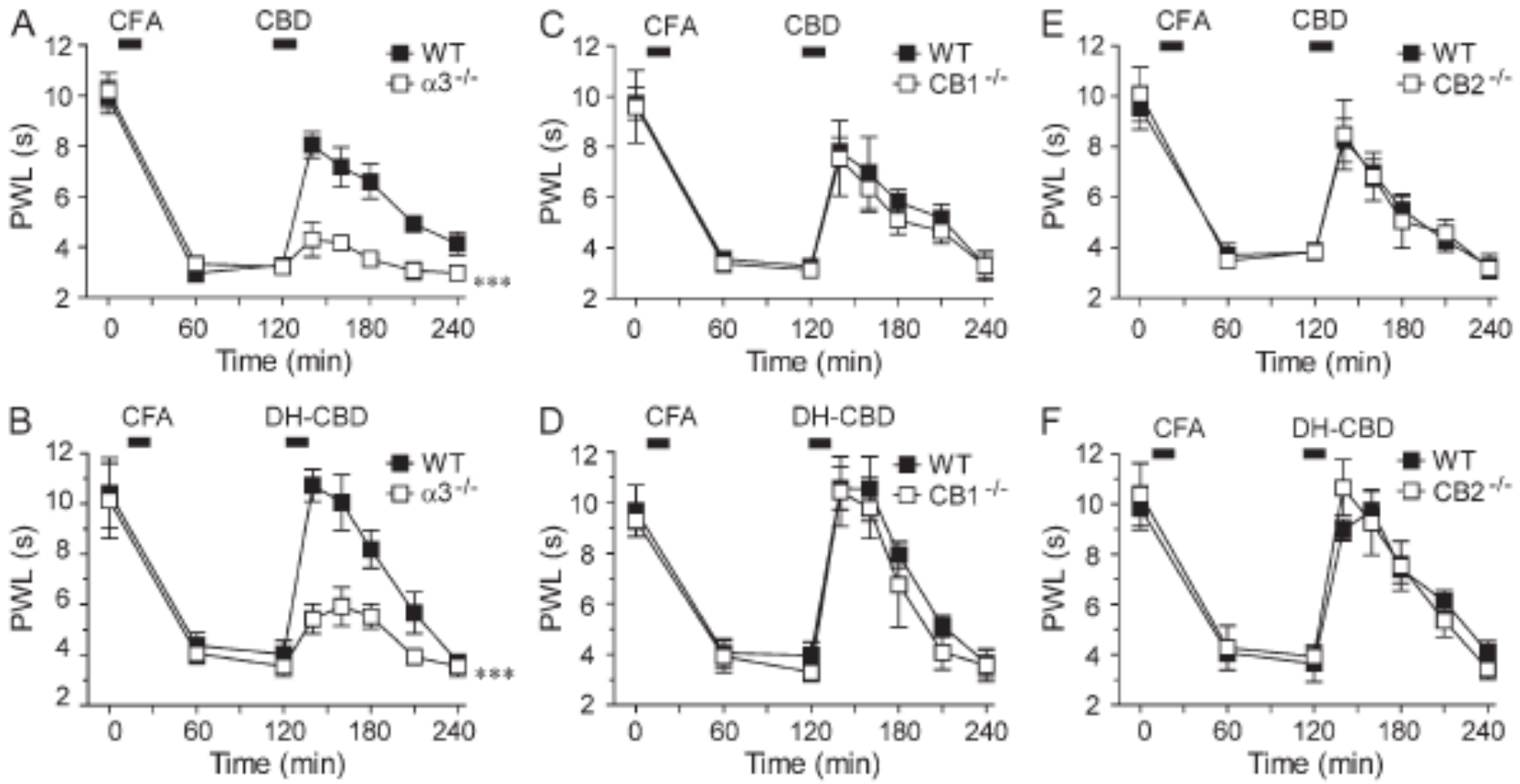
DH-CBD can suppress

- acute pain (Tail flick reflex)
- Chronic inflammatory pain (CFA paw injection)
- Neuropathic pain (spinal nerve ligation)

Xiong et al., 2012, J. Exp. Med.



Cannabinoid-induced analgesia is significantly reduced In α3 GlyR KO mice but not in CB1 or CB2 KO mice

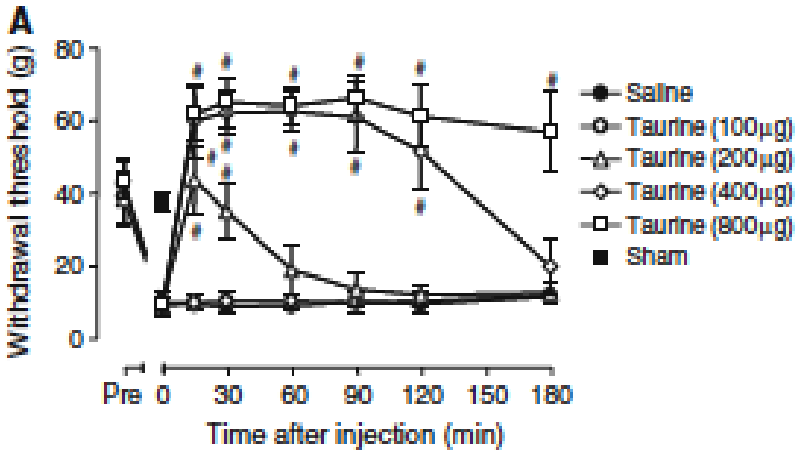


Xiong, et al.2012, J. Exp. Med.

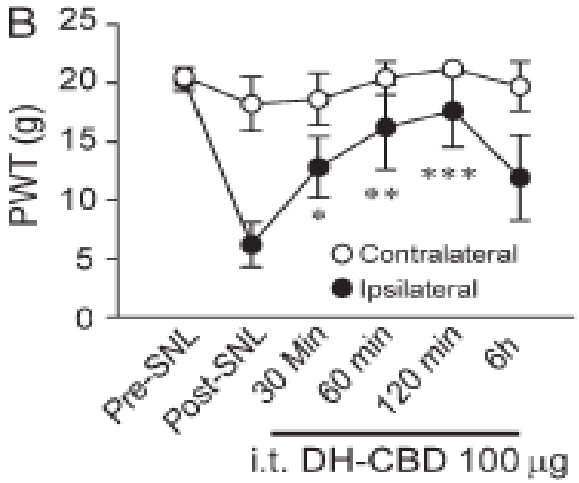
Therapeutic advantage of glycinergic cannabinoid vs traditional GlyR agonist in the treatment of pain

more efficacious and safer

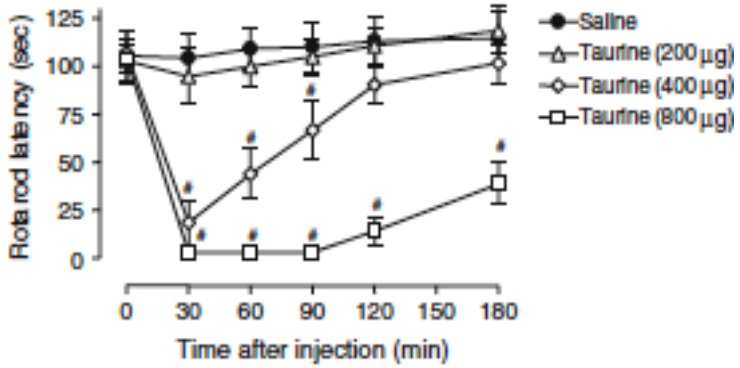
Endogenous GlyR agonist, taurine, suppresses neuropathic pain **but also impairs neuromotor activity**



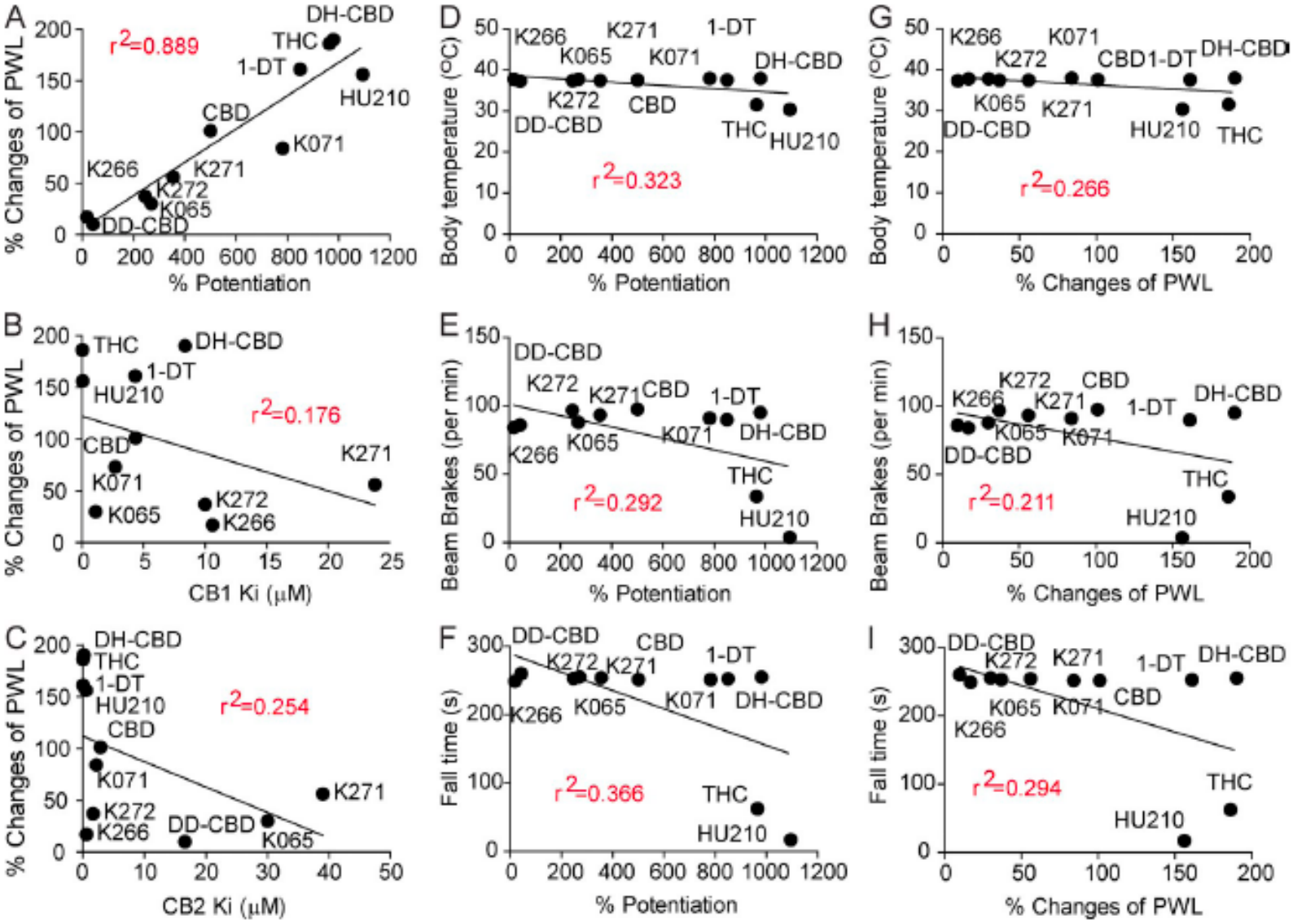
Terada, et al, 2011, Can. J. Anal.



Xiong, et al, 2012, JEM.

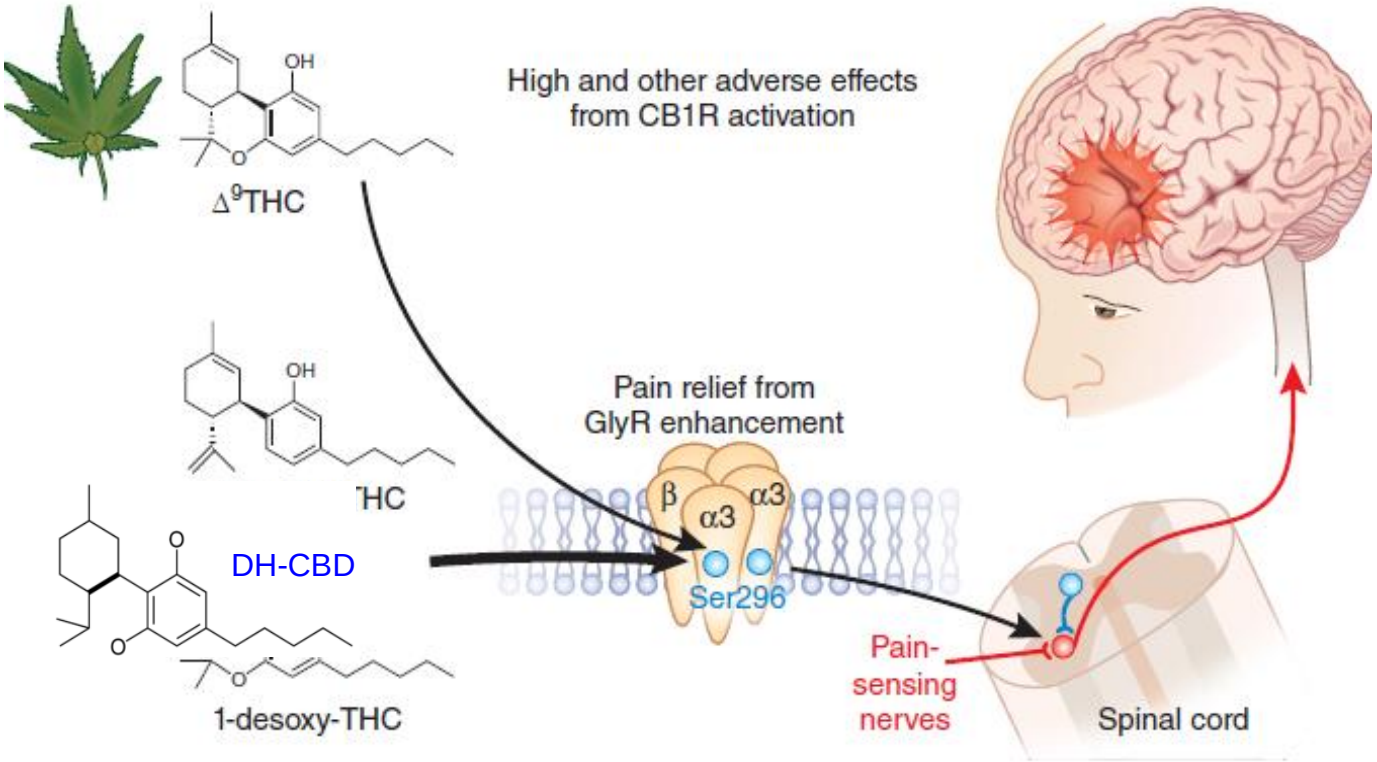


Gly α3 receptors is an ideal target for the treatment of chronic pain



Xiong et al, 2012 JEM

α3 GlyRs contribute to cannabis-induced pain relief



NATURE CHEMICAL BIOLOGY | VOL 7 | MAY 2011

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