

SUPPRESSION OF NEUROGENIC DETRUSOR OVERACTIVITY BY GLYCINE TRANSPORTER TYPE 2 (GLYT2) INHIBITOR IN RATS WITH SPINAL CORD INJURY

Hypothesis / aims of study

Glycine is a major inhibitory neurotransmitter in the central nervous system including the spinal cord. The concentration of glycine at synaptic nerve terminals is regulated by two types of glycine transporters (GlyTs): GlyT1 and GlyT2. GlyT1 is mainly located at glial cells and/or glutamatergic nerve terminals to modulate glycine concentration at NMDA receptor expressing synapses. On the other hand, GlyT2 is located on inhibitory glycinergic nerve terminals with high degree overlap with the expression pattern with strychnine sensitive glycine receptor (GlyR). A recent study demonstrated that a GlyT2 inhibitor suppresses bladder overactivity induced by chemical bladder irritation in rats [1] although it is not known whether GlyT inhibitors have therapeutic effects on bladder overactivity in chronic disease conditions. Therefore, this study utilized rats with chronic spinal cord injury (SCI) to investigate the SCI-induced changes in spinal GlyT and GlyR expression and the effects of GlyT inhibitors, which can increase the glycine concentration at nerve terminals, on neurogenic detrusor overactivity as shown by non-voiding contractions (NVCs) during the storage phase.

Study design, materials and methods

Adult female Sprague-Dawley rats were used. SCI was produced by complete transection of the Th8-9 spinal cord. After 4 weeks, cystometry under an awake condition was performed with continuous infusion of saline (0.08 ml/min) into the bladder. Selective GlyT1 or GlyT2 inhibitors (sarcosine or ALX-1393, respectively) were administered intrathecally via a PE-10 catheter connected with a 30G needle and placed at the L6 spinal cord level to examine the effect of GlyT inhibition on SCI-induced detrusor overactivity. Cystometric parameters evaluated included the amplitude (cmH₂O) and frequency (number/min) of NVCs. GlyT1, GlyT2 and glycine receptor (GlyR) levels such as GlyR α 1, GlyR α 2, GlyR α 3 and GlyR β in the L6-S1 spinal cord were measured by RT-PCR in spinal intact and SCI rats (n=5 each).

Results

Intrathecal application of sarcosine, a selective GlyT1 inhibitor, did not elicit significant changes in any cystometric parameters (n=5) even at a high dose (250 μ g). In contrast, intrathecal application of ALX-1393 (3, 10 and 30 μ g, n=5 each), a selective GlyT2 inhibitor, dose-dependently reduced the amplitude and frequency of NVCs with significant changes at 10 μ g (26 and 65% reductions, respectively) and 30 μ g (37 and 76 % reductions, respectively) (Fig. 1.). SCI rats had a significantly higher GlyT2 mRNA level, but not a GlyT1 level, without changes of GlyR levels in the L6-S1 spinal cord compared to spinal intact rats (Fig. 2.).

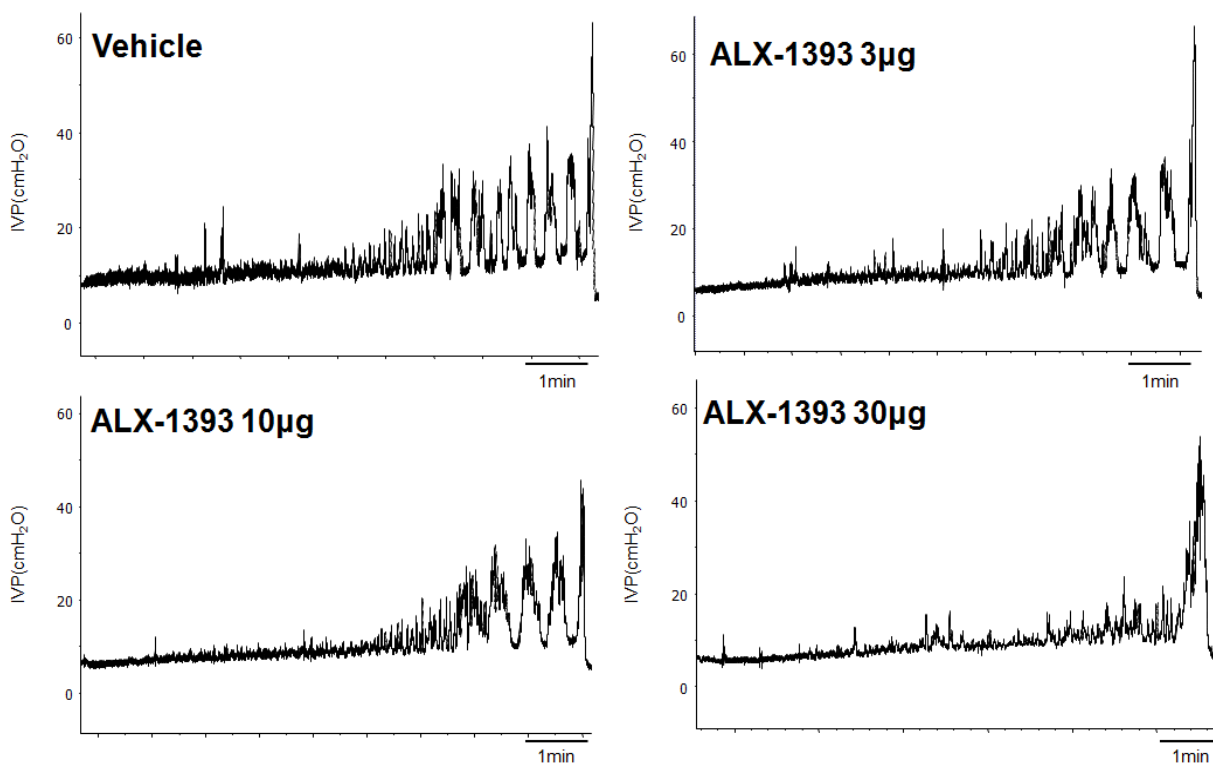
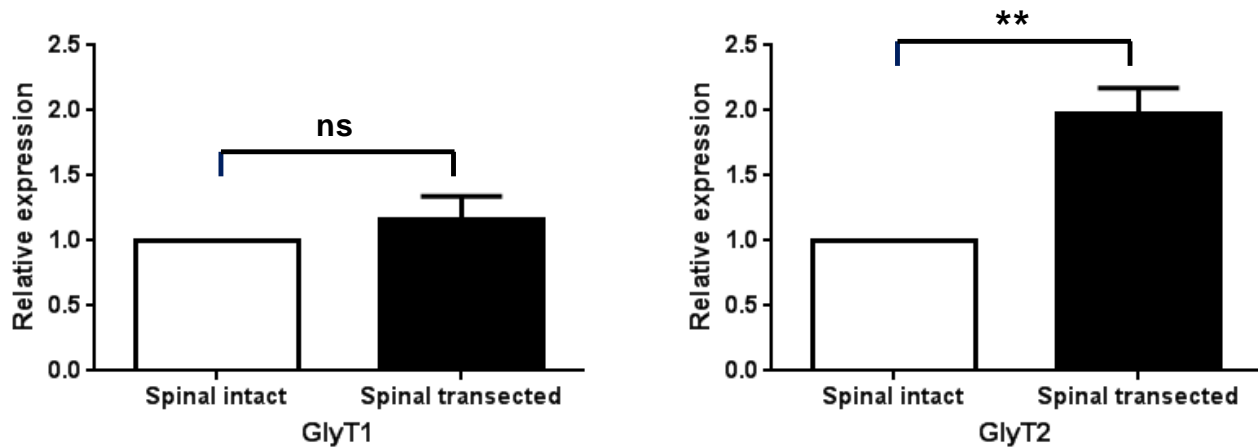


Fig. 1. Representative trace of cystometrograms after intrathecal (i.t.) application of Vehicle or ALX-1393, a GlyT2 inhibitor, (3, 10 and 30 μ g) in SCI rat.



n=5 rats each

Data are expressed as mean \pm s.e.m

** ; p<0.01, compared with the spinal intact group
ns; not significant

Fig. 2. GlyT1 and GlyT2 mRNA levels in the L6-S1 spinal cord.

Interpretation of results

These results indicate that: (1) the spinal inhibitory glycinergic mechanism is compromised in SCI due to increased expression of GlyT2, which enhances glycine uptake to reduce the concentration of glycine at synaptic nerve terminals, and (2) GlyT2 inhibition that enhances the spinal glycinergic system by inhibiting the glycine uptake at nerve terminals can suppress neurogenic detrusor overactivity as evidenced by the reduction of NVCs in chronic SCI rats. Because sensitization of bladder afferent pathways and increased signal transduction in the spinal cord have been proposed as important mechanisms inducing bladder overactivity [2], GlyT2 inhibitors might be effective for reducing neurogenic detrusor overactivity by suppressing sensory inputs from the bladder to the spinal cord.

Concluding message

Increased expression of glycine transporter type 2 (GlyT2) in the spinal cord, which reduces the glycine-mediated spinal inhibitory function, may contribute to neurogenic detrusor overactivity in SCI. Also, GlyT2 inhibitors could be a novel option for the treatment of overactive bladder with neurogenic detrusor overactivity by suppressing sensory inputs from the bladder to the spinal cord.

References

1. Eur Urology 2012; 62: 704-712.
2. Experimental Neurology 2012; 235: 123-132.

Disclosures

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References

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Disclosures

Funding: Funding: PVA 2793, DOD W81XWH-11-1-0763 and NIH DK088836, P01 DK093424 **Clinical Trial:** No **Subjects:** ANIMAL **Species:** Rat **Ethics Committee:** University of Pittsburgh Institutional Animal Care and Use Committee