

INHIBITION OF P75 NEUROTROPHIN RECEPTORS REDUCES NEURAL DEGENERATION AND LOWER URINARY TRACT DYSFUNCTION FOLLOWING SPINAL CORD INJURY

Hypothesis / aims of study

Spinal cord injury (SCI) causes detrimental effects to lower urinary tract (LUT) function. Primarily, SCI is associated with the development of detrusor-sphincter dyssynergia (DSD) which can lead to obstructive uropathy and further damage to the LUT. Neurotrophic factors, including nerve growth factor (NGF) and brain-derived neurotrophic factor (BDNF), are believed to contribute to the development of DSD [1]. NGF/BDNF are released as pro-forms that are later converted to mature proteins. The pro and mature proteins preferentially signal through p75 neurotrophin (p75^{NTR}) or TrkA/B receptors, respectively, and elicit differential effects on cell survival. The p75^{NTR} has been implicated in neural degeneration following SCI and selective inhibition by a centrally acting small molecule drug, LM11A-31, has been shown to be protective [2]. The role of proneurotrophins in the CNS and bladder wall following SCI has not been fully characterized. Therefore, our aims were to investigate the sites of action and therapeutic benefits of p75^{NTR} inhibitors on LUT dysfunction following thoracic SCI.

Study design, materials and methods

Adult (6-8 weeks) female C57Bl/6 mice were used for this study. SCI was performed by anesthetizing mice with isoflurane (5% induction, 2% maintenance in 100% oxygen), a laminectomy performed at T8-T9 vertebrae and the spinal cord completely transected. The transection site was packed with haemostatic sponge and the wound sutured. Animals were allowed to recover and given prophylactic antibiotics and analgesics for 7 days. Daily LM11A-31 or LM11A-24 (peripherally acting p75^{NTR} ligand; not commercially available and synthesized in-house) treatment (100 mg/kg/day in water) was given by oral gavage starting one day prior to SCI. LUT function was assessed by weekly 2-hour urine spot tests. At experimental end points (1 day to 8 weeks post-SCI), mice were used for decerebrate cystometry or bladder transepithelial resistance (TER) measurements and bladders and spinal cords collected for histological analysis.

Results

Between 2 to 8 weeks post-SCI, cystometries demonstrated the development of large amplitude non-voiding contractions and significant decrease in bladder compliance (Fig 1A). Daily LM11A-31 treatment improved voiding function and bladder compliance compared to untreated SCI mice. Urine spot tests showed similar improvements where LM11A-31, but not LM11A-24, treatment resulted in increased voided volumes in SCI mice (not shown). Bladder histology revealed LM11A-31 and -24 pre-treatment prevented the loss of the urothelium at 1 day post-SCI (Fig 1B) and this was reflected by maintenance of a high TER comparable to uninjured control mouse bladders (Fig 1C). Lastly, chronic SCI (> 8 weeks post-SCI) resulted in significant atrophy of the spinal cord. LM11A-31 and not the peripherally acting LM11A-24 reduced spinal cord atrophy (Fig 1D).

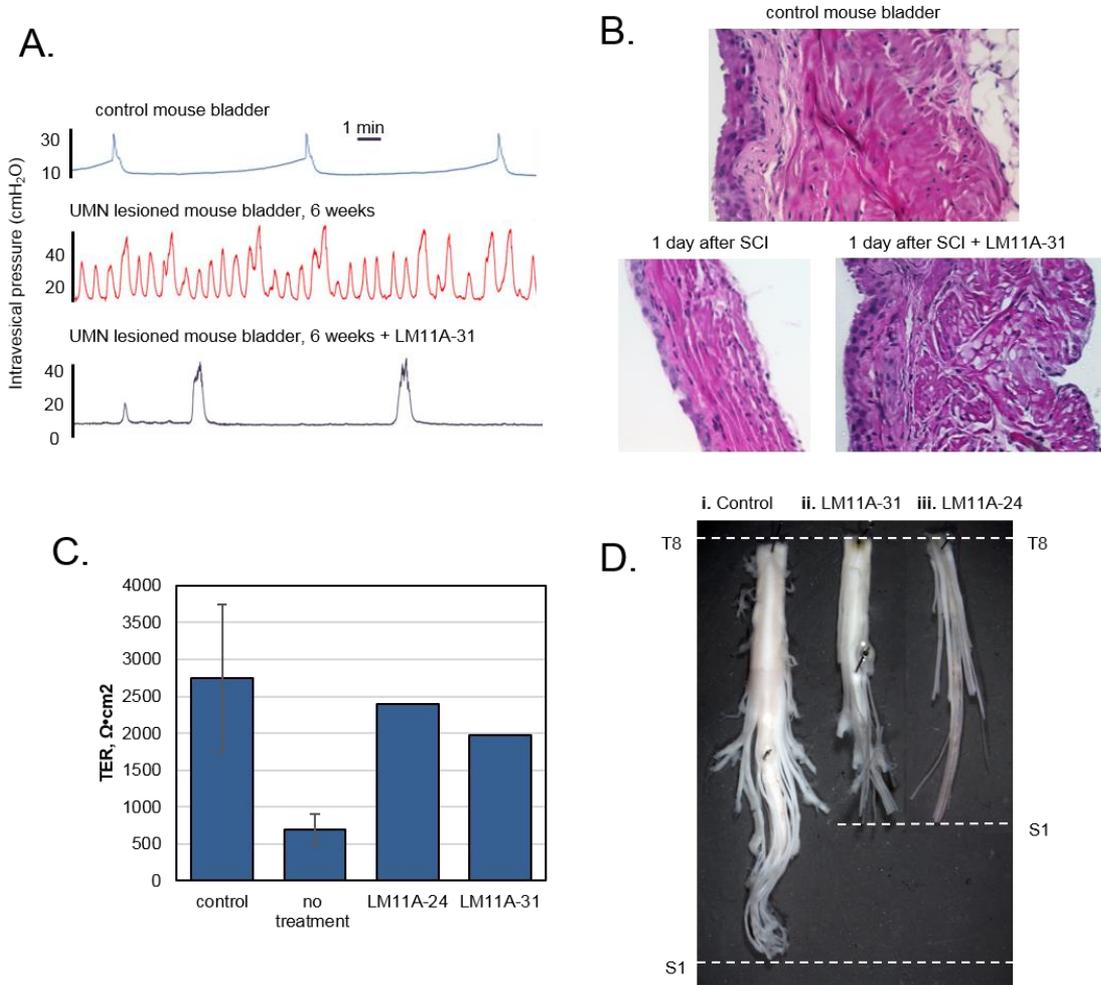
Interpretation of results

This study has demonstrated p75^{NTR} inhibition prevents loss of the urothelial layer and neural degeneration following SCI. Proneurotrophins are released immediately following SCI and could be responsible for apoptosis of the urothelium in acute stages and degeneration of spinal cord neurons chronically. Inhibition of p75^{NTR} by LM11A-31 protected the integrity of the urothelium and reduced the degree of spinal cord atrophy following thoracic SCI. The peripherally acting LM11A-24 better protected against urothelial damage which was reflected in higher TER, but did not prevent neural degeneration. These results suggest that p75^{NTR} activation in both peripheral and central sites are responsible for SCI-induced LUT pathology. Accordingly, combination therapy using LM11A-31 and -24 may be beneficial.

Concluding message

Proneurotrophins contribute to SCI-induced urothelial and neural dysfunction, which can be prevented by administration of small molecule p75^{NTR} inhibitors. Centrally acting LM11A-31, currently in clinical trials for treatment of neural degenerative diseases, and peripherally acting LM11A-24 may have utility for treating LUT dysfunction following SCI.

Figure 1



References

1. Ochojnicky P et al., Neurotrophins as regulators of urinary bladder function, *Nat Rev Urol*;9(11):628-37, 2012.
2. Tep C et al., Oral administration of a small molecule targeted to block proNGF binding to p75 promotes myelin sparing and functional recovery after spinal cord injury, *J Neurosci*, 9;33(2):397-410, 2013.

Disclosures

Funding: NIH/NIDDK grants (DK093424, DK071085 and DK098361) **Clinical Trial:** No **Subjects:** ANIMAL **Species:** Mouse, C57Bl/6 strain **Ethics Committee:** University of Pittsburgh Institutional Animal Care and Use Committee (IACUC)