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ROLE OF P38 MAP KINASE SIGNALLING PATHWAYS IN STORAGE AND VOIDING DYSFUNCTION IN MICE WITH SPINAL CORD INJURY

Hypothesis / aims of study

Chronic spinal cord injury (SCI) rostral to the lumbosacral level induces detrusor overactivity (DO) during the storage phase, which is mediated by spinal reflexes triggered by hyperexcitable C-fiber afferent pathways. During the voiding phase, inefficient voiding is commonly observed due to detrusor-sphincter dyssynergia or DSD after SCI. It has also been shown that neurotrophic factors such as nerve growth factor (NGF) released within the urinary bladder or the spinal cord are an important mediator inducing DO after SCI [1]. It is also known that the second messenger signalling pathways activated by NGF involve activation of p38 MAP kinase (MAPK) [2], which is a serine-threonine kinase that is activated by phosphorylation and mediates cellular responses to a variety of chemical and physical insults [3]. However, it remains to be elucidated whether the p38 MAPK pathway is also involved in lower urinary tract dysfunction induced by SCI. Therefore, we investigated the effects of a p38 MAPK inhibitor treatment using SCI mice to clarify the role of the p38 MAPK pathway in SCI-induced storage and voiding dysfunction.

Figure 1; Single CMG recordings in SCI mice

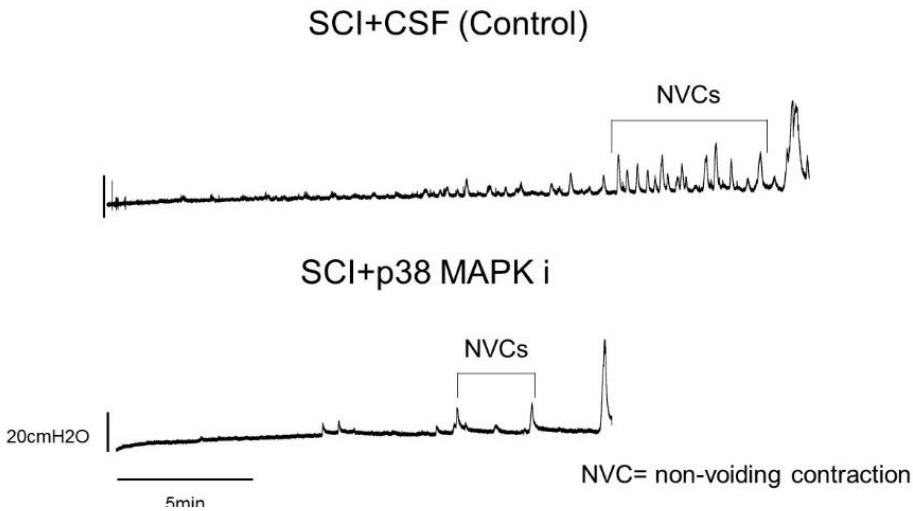


Figure 2; Urodynamic parameters in p38 MAPK i -treated and CSF treated SCI mice

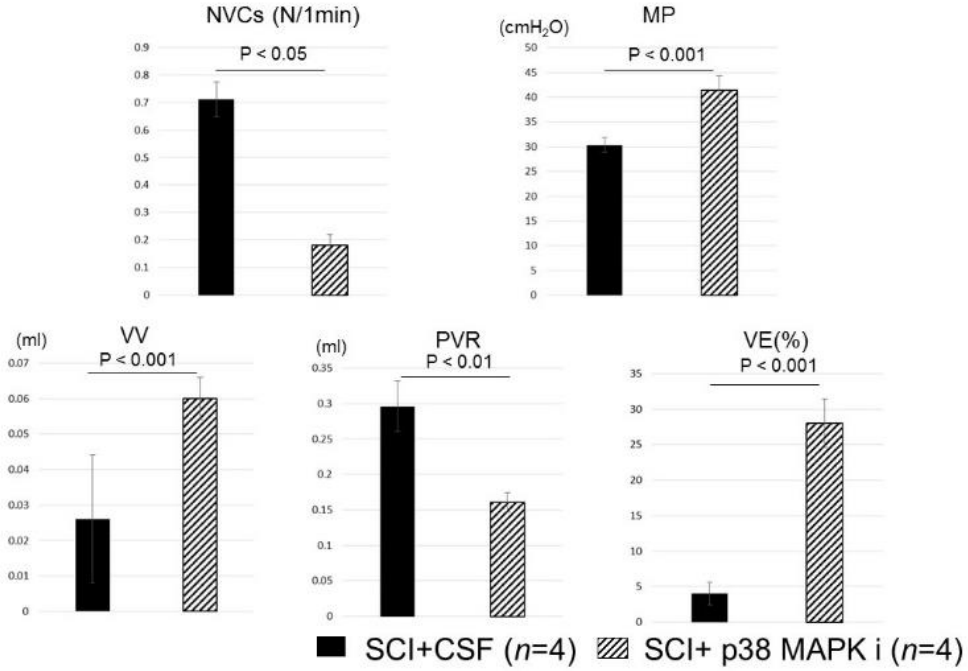
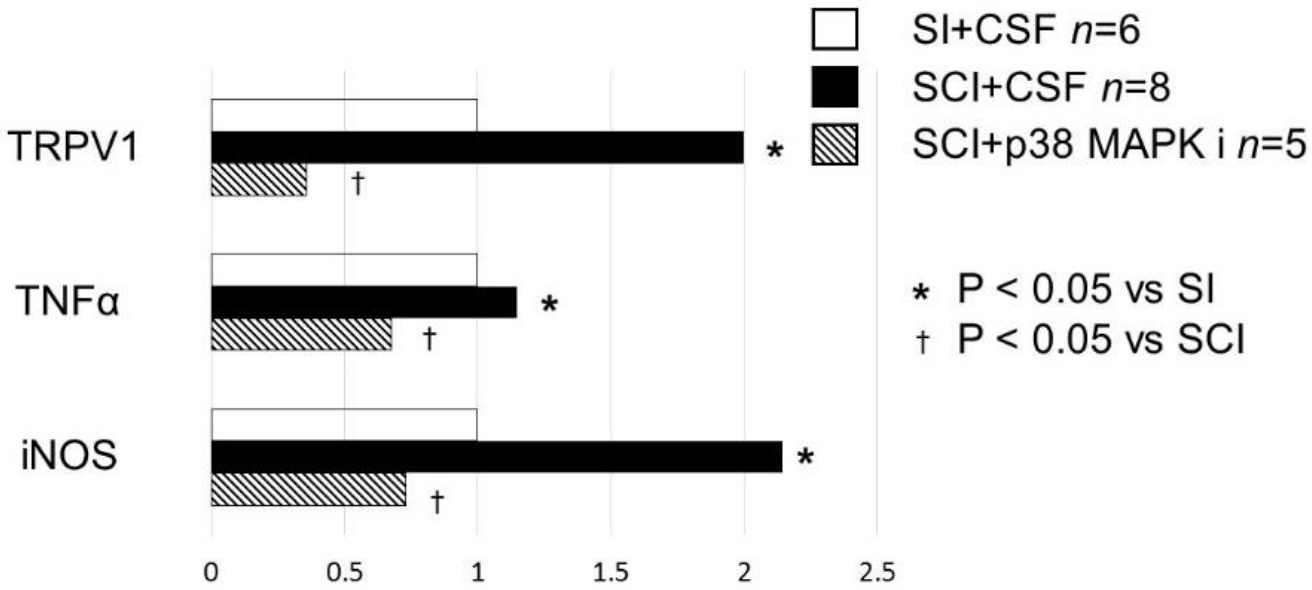


Figure 3; Real-time PCR results



Study design, materials and methods

Female C57BL/6N (8-9 week-old) mice weighing 18-20 g were used, and SCI was induced by complete transection of the Th8/9 spinal cord under isoflurane anesthesia. SCI mice were divided into 2 groups; (1) SCI mice treated with SB203580 (1 mg/ml), a p38 MAP kinase inhibitor (p38 MAPK i) (*n*=4), (2) SCI mice with artificial cerebrospinal fluid (CSF) (*n*=4). Two weeks after SCI, an intrathecal catheter connected to an osmotic pump was implanted into the intrathecal space of L6-S1 spinal cord for continuous intrathecal instillation at infusion rate of 0.51 µl/hr for 2 weeks. Osmotic pumps filled with p38 MAPK i or CSF were placed in the subcutaneous space between shoulder blades. After spinal cord transection, their bladders were manually squeezed to eliminate the urine once daily for 4 weeks until cystometric evaluation. SCI mice were evaluated using single-filling cystometry (CMG) and external urethral sphincter (EUS)-electromyogram (EMG) under an awake condition. In single CMG recordings, the number of non-voiding contractions (NVCs), micturition pressure (MP), post-void residual volume (PVR) and voiding efficiency (VE) were evaluated in each SCI mouse (Figure 1). In simultaneous CMG and EUS-EMG recordings, voiding contraction time, reduced EMG activity duration and the ratio of reduced EMG activity time to voiding contraction time were measured during the voiding phase to evaluate DSD in each SCI mouse. In real-time PCR analyses, L6 dorsal root ganglia (DRG) were removed from CSF-treated SCI mice (*n*=8) and p38 MAPK i-treated SCI mice (*n*=5) as well as CSF-treated normal (spinal intact) mice (*n*=6) and the levels of TRPV1, TNFα and iNOS transcripts were evaluated.

Results

Compared to CSF-treated SCI mice, NVCs during bladder filling were significantly reduced (Figure 1), and voiding efficiency was significantly improved with increased voided volume and micturition pressure in p38 MAPK i-treated SCI mice (Figure 2). In CMG and EUS-EMG recordings, the duration of reduced EMG activity or the ratio of reduced EMG activity time to voiding contraction time during the voiding phase was not significantly different between p38 MAPK i -treated and CSF-treated SCI mice.
The expression of TRPV1, TNFα and iNOS mRNA was increased in SCI mice compared to spinal intact mice, and significantly decreased after p38 MAPK i treatment in SCI mice(Figure 3).

Interpretation of results

The treatment with a p38 MAPK inhibitor improved DO evident as a decrease in NVCs in association with the reduction of the expression of TRPV1 in L6 DRG, which is predominantly expressed in C-fiber afferent pathways, and also voiding dysfunction as shown by increased voiding efficiency due to increased voided volume and micturition pressure without affecting DSD in SCI mice. Also, the p38 MAPK inhibitor treatment reduced the iNOS and TNFα expression in L6 DRG, suggesting that the p38 MAPK signaling pathway is involved in the inflammatory changes in bladder afferent pathways. Overall, the results of this study demonstrated that activation of the p38 MAPK signaling pathway significantly contributes to lower urinary tract dysfunction as well as inflammatory changes in bladder afferent pathways after SCI and that inhibition of the p38 MAPK pathway can improve SCI-induced voiding and storage dysfunction.

Concluding message

The p38 MAPK signalling pathway could be an effective target for the treatment of storage and voiding problems such as DO and inefficient voiding, respectively, after SCI.

References

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Disclosures

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