Received: 25 April 2018 | Accepted: 4 September 2018
DOI: 10.1002/nau.23827

ORIGINAL BASIC SCIENCE ARTICLE

WILEY Neurourology Urodynamics ICS SUFU

Spinal cord stimulation ameliorates detrusor over-activity and visceromotor pain responses in rats with cystitis

Huiyi H. Chang[1,2] | **Jih-Chao Yeh**[3] | **Jackie Mao**[3] | **David A. Ginsberg**[3] | **Gamal Ghoniem**[1] | **Larissa V. Rodriguez**[3]

[1] Department of Urology, University of California Irvine, Irvine, California

[2] Reeve-Irvine Research Center, University of California Irvine, Irvine, California

[3] Department of Urology, University of Southern California, Los Angeles, California

Correspondence
Huiyi H. Chang, PhD, Urology and Reeve-Irvine Research Center, University of California Irvine, 837 Health Science Road, Gillespie 2111, Zot Code 4265, Irvine, CA 92697.
Email: harriet.chang@uci.edu

Funding information
National Institute of Diabetes and Digestive and Kidney Diseases, Grant number: R01 DK106181

Aim: Interstitial cystitis/painful bladder syndrome/(IC/PBS) results in recurring pain in the bladder and surrounding pelvic region caused by abnormal excitability of micturition reflexes. Spinal cord stimulation (SCS) is currently clinically used for the attenuation of neuropathic and visceral pain. The present study examined whether SCS at upper lumbar segments modulates detrusor overactivity and visceral hyperalgesia associated with cystitis in a rat model of cyclophosphamide (CYP)-induced cystitis.
Methods: Cystitis was induced by intraperitoneal injection of CYP (200 mg/kg) in six adult female Sprague Dawley rats 48 h prior to urodynamic recordings. Another six rats served as—controls with saline injection. Cystometry and the external urethral sphincter (EUS) electromyography during bladder infusion were evaluated under urethane anesthesia. The visceromotor reflexes (VMR) obtained from the external abdominal oblique muscle were quantified during bladder infusion and isotonic bladder distension (IBD), respectively. After baseline recordings were taken, SCS was applied on the dorsal surface of L3 for 25 min. Urodynamic recordings and VMR during bladder infusion and IBD were repeated 2 h after SCS.
Results: CYP resulted in detrusor overactivity, stronger EUS tonic contractions, and increased VMR. SCS significantly reduced non-voiding contractions, prolonged EUS relaxation, and delayed VMR appearance during bladder infusion as well as significantly decreased VMR during IBD in cystitis rats.
Conclusion: SCS improved bladder function and EUS relaxation during bladder infusion and significantly attenuated visceral nociceptive-related VMR during IBD in cystitis rats. SCS may have therapeutic potential for patients with hyperalgesia and IC/PBS.

KEYWORDS
electromyography, external urethral sphincter, neuromodulation, visceral pain

1 | INTRODUCTION

Neuromodulation of the sacral nerves and lumbosacral spinal roots has been established as an effective treatment for urge incontinence, urgency-frequency symptoms, non-obstructive urinary retention, and neurogenic bladder.[1] Spinal cord stimulation (SCS) of upper lumbar segments was previously reported to improve the urethral relaxation and promote efficient voiding.[2,3] SCS has also been employed for chronic neuropathic pain and visceral hyperalgesia. The mechanisms

Karl-Erik Andersson led the peer-review process as the Associate Editor responsible for the paper.

underlying SCS amelioration of visceral hypersensitivity are not well understood but may involve both spinal and supraspinal neural circuits.[4,5]

Bladder visceral hyperalgesia is often caused by damage to nociceptors, resulting from inflammation and cystitis. Cyclophosphamide (CYP)-induced cystitis in rodents has been used for studying interstitial cystitis/painful bladder syndrome (IC/PBS) characterized by pelvic pain, urinary urgency, and urinary frequency.[6,7] CYP-treated rodents also show bladder hyperalgesia and allodynia to noxious bladder distention.[6,7] In this study CYP-induced cystitis is used to evaluate the effect of SCS at the upper lumbar segment on detrusor overactivity (DO) and bladder hyperalgesia.

In addition, urethral relaxation is associated with bladder contractions which is known as detrusor-sphincter coordination. Efficient voiding critically depends on input from the urethra. The urethral afferent response to stimulation activates spinal cord neurons and triggers CNS-mediated urethral-to-bladder reflexes that contribute to efficient voiding.[8,9] Besides investigating bladder contractions, the external urethral sphincter (EUS) activity during filling and voiding is examined before and after SCS.

The urotoxic metabolites of CYP (ie, acrolein) accumulate in the bladder and urethra, and contribute to the activation of C-fibers by direct contact with urothelium, leading to detrusor over-activity, and visceral hyperalgesia. We hypothesize that SCS over the upper lumbar level could inhibit the activated afferents conveying noxious stimuli from the bladder and urethra. Therefore, the present study was aimed to investigate the therapeutic effect of SCS on voiding function, EUS activation, and visceral nociception in CYP-induced cystitis rats.

2 | METHODS

All experiments were carried out in accordance with the National Institutes of Health guidelines and approved by the Institutional Animal Care and Use Committees at University of Southern California and University of California Irvine, CA, USA. Adult female Sprague-Dawley rats (237 ± 6 g, $n = 18$, Charles River Laboratories, CA) were housed and maintained in the animal facility on a 12 h light-dark cycle, and fed with standard food.

The rats received intraperitoneally (ip) administrated saline (controls, $n = 6$) or CYP (cystitis, 200 mg/kg, $n = 12$) 48 h prior to the urodynamic recordings and SCS. The cystitis rats were divided into two groups: 1) urodynamic recordings and visceromotor reflexes (VMR) during bladder infusion ($n = 6$) and 2) VMR during isotonic bladder distension (IBD, $n = 6$).

2.1 | Surgical preparations for urodynamic recordings and VMR

Rats received urethane (1.2 g/kg, ip) 1 h prior to surgical preparations and were then placed on a water-circulating heating pad to keep the body temperature between 36 and 38°C. A lower abdominal midline incision was made to expose the bladder and mid-urethra. A PE-50 catheter (Instech Laboratories, Plymouth Meeting, PA) with the flared tip was inserted into the top of the bladder dome and secured using a suture.[2,10] The other end of the catheter was connected to an infusion pump (KD Scientific Inc., Holliston, MA) and a pressure sensor (Biopac Systems Inc., Goleta, CA) via a 3-way connector. Two 50 μm insulated silver wire electrodes with exposed tips were inserted into the mid-urethra bilaterally, approximately 2 mm from the bladder neck, to obtain the external urethral sphincter (EUS) electromyography (EMG). Two additional wire electrodes with exposed tips were inserted into the external oblique muscle to record muscle activity associated with visceral pain-related VMR during isotonic bladder distension. The pressure sensor and two sets of EMG wires were all connected to the data acquisition system (MP150, Biopac Systems Inc.) and data were saved to the computer for data analysis.

2.2 | Surgical preparations for SCS

Laminectomy was performed at the thoracolumbar levels of T12-L1. The dura over the third lumbar spinal cord segment (L3) was exposed to place wire electrodes for SCS. After a small part of the Teflon coating (about 1 mm notch) was removed from the wires (AS632, Cooner Wire, Chatsworth, CA), the exposed part of the wire was positioned facing the dura of L3, and then secured as the anode and cathode.[2,10] Then, the muscle and skin layers were sutured. The wires were connected to a stimulus isolation unit and an electrical stimulator (SIU5 and S88, GRASS Technologies, West Warwick, RI). The threshold intensity was set as the appearance of EUS bursting responses, which represents voiding in rodents.[2] The stimulating intensity of SCS was defined at 1.5-2 fold of the threshold intensity (2.5-4 volts, 40 Hz, and 0.2 msec pulse width) without hind limb muscle twitching.

2.3 | Recording procedures

Baseline urodynamic and VMR parameters during a 30 min bladder infusion were obtained in the control ($n = 6$) and cystitis groups ($n = 6$) 1 hr after administration of urethane. The infusion rate was set at 0.1 mL per minute. The urethral outlet was opened to allow for urine expulsion. Then, the bladder infusion was stopped and SCS was applied for 25 min. Urodynamic recordings during bladder infusion were then obtained immediately and 2 h after SCS.

VMR during IBD ($n = 6$) was measured by established methods.[11,12] The urethra was occluded to maintain the desired intravesical pressure (IVP). The bladder was distended using saline, and the IVP was maintained at 10, 20, 30, and 40 cm H_2O for 120 s. Following each IVP level, the bladder was emptied and allowed to rest for 120 s. After the VMR baseline was obtained, SCS was applied for 25 min with an empty bladder. VMR during IBD was then repeated 2 h after SCS.

2.4 | Outcome measurements

The urodynamic and VMR parameters were analyzed during bladder infusion for three consecutive voiding contractions. Cystometry measures included: non-voiding contractions that were larger than 15 cmH2O and did not result in voided volume, voiding efficiency, inter-contraction intervals, and residual volume. Voiding efficiency was calculated as the percentage of voided volume/total infused volume. The IVP- evoked VMR was referred to as VMR threshold (cmH2O). The normalized ratio of VMR threshold/maximum IVP was calculated. EUS-EMG measurements included the bursting duration during voiding and the active periods and silent periods during bursting duration.[13] The area under the curve (AUC) of during IBD VMR was measured at each IVP level.[11,12]

2.5 | Statistical analysis

All parameters obtained from urodynamic recordings and VMR are presented as mean values ± standard error. Comparisons of urodynamic measurements and VMR AUC between the control and cystitis groups were performed using the non-parametric Mann-Whitney test. Comparisons of urodynamic measurements and VMR AUC before and after SCS were performed using the non-parametric Wilcoxon matched-paired test. P value < 0.05 indicated statistically significant difference. GraphPad Prism 6 (GraphPad Software, La Jolla, CA) was used to conduct the statistical analysis and figures.

3 | RESULTS

3.1 | Urodynamic recordings during bladder infusion

Forty-eight hours after CYP administration, the cystitis rats ($n = 6$) showed increased non-voiding contractions ($P = 0.03$) compared to controls ($n = 6$) (Figure 1). These non-voiding contractions were not affected by acute SCS ($P = 0.38$; compared to before SCS). However, the non-voiding contractions were significantly decreased 2 h after SCS ($P = 0.04$; Figure 1). Therefore, the following data analysis was processed in the controls and cystitis rats before and 2 h after SCS.

Two hours after SCS, voiding efficiency was decreased in the cystitis rats ($61 \pm 5\%$, $n = 6$) compared to controls ($91 \pm 1\%$, $n = 6$) ($P = 0.002$) (Figure 2). Inter-contraction intervals in cystitis rats were significantly shorter (97 ± 17 s) compared to controls (162 ± 21 s, $P = 0.048$). The normalized ratio of VMR threshold/maximum IVP in cystitis rats ($74 \pm 4\%$) was significantly lower compared to controls ($87 \pm 2\%$, $P = 0.008$; Figure 2). The cystitis rats had significantly higher residual volume (71 ± 15 μL, $P = 0.011$) compared to controls (19 ± 5 μL).

The inter-contraction intervals analyzed at 2 h after SCS were prolonged to 175 ± 29 s (cystitis + SCS, $P = 0.046$) in the cystitis rats compared to before SCS (Figure 2). SCS in the cystitis rats significantly improved voiding efficiency compared to before SCS ($P = 0.03$). SCS significantly reduced the residual volume (35 ± 13 μL) in the cystitis rats (cystitis + SCS, $P = 0.03$) compared to the condition without SCS. SCS significantly delayed the VMR appearance, which is the normalized ratio of VMR threshold/maximum IVP, in cystitis rats during bladder infusion (cystitis + SCS, $P = 0.03$) compared to the condition without SCS. SCS had no significant effect on urodynamic recordings and VMR in the control rats.

During bladder filling, tonic EUS-EMG activity gradually increased and markedly changed to EUS bursting when

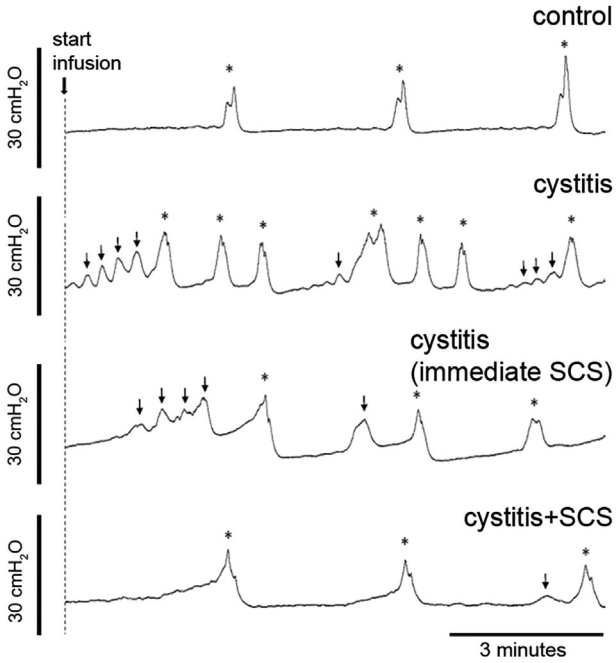

FIGURE 1 Representative examples of cystometry during bladder infusion. The voiding and non-voiding contractions were more frequent in the cystitis rat compared to the control. The numbers of non-voiding contractions were decreased immediately after SCS. Two hours after SCS, the voiding and non-voiding contractions in cystitis rats were significantly decreased compared to the cystometry without SCS. Asterisk and arrows indicated voiding and non-voiding contractions, representatively

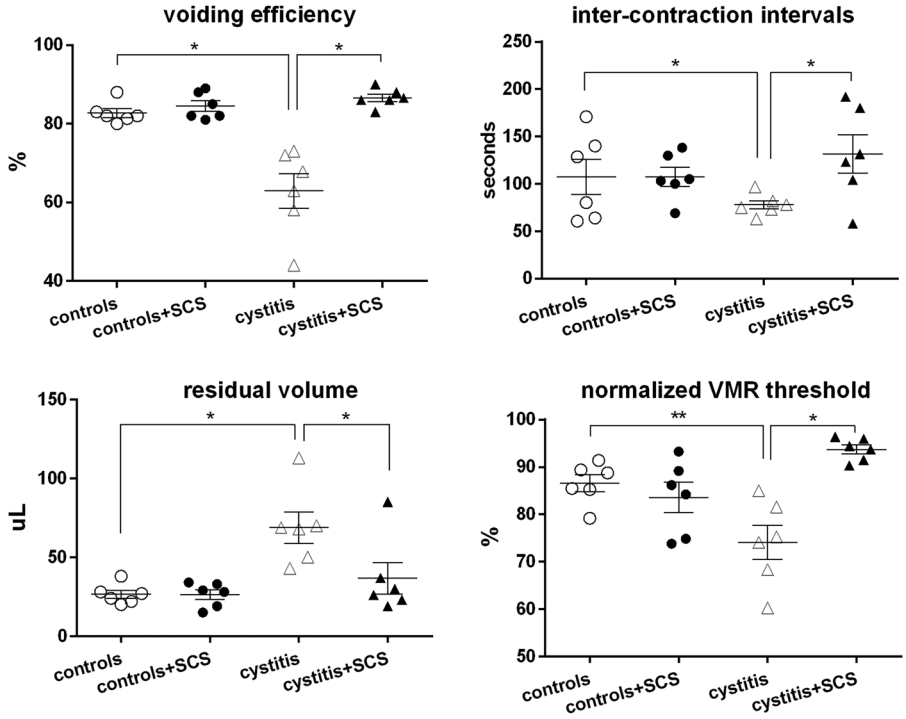

FIGURE 2 Statistical analysis of cystometry during bladder infusion in controls ($n = 6$) and cystitis rats ($n = 6$) before and 2 hr after SCS. * and ** indicated the $p < 0.05$ and $p < 0.01$, respectively

bladder voiding contraction started (Figure 3A). EUS bursting activity was comprised of active and silent periods (Figure 3B). The cystitis rats, compared to controls, showed significant changes including shorter bursting duration ($P = 0.025$), prolonged active periods ($P = 0.03$), and shorter silent periods ($P = 0.04$) (Figures 3B and 3C). Two hours after SCS in the cystitis rats, the bursting duration was significantly increased (cystitis + SCS, $P = 0.03$). SCS did not change the active periods in the cystitis rats; however, the silent periods were significantly prolonged by SCS (cystitis + SCS, $P = 0.03$) (Figures 3B and 3C).

3.2 | VMR during IBD

Two hours post SCS, the VMR AUC significantly increased during IBD at the IVP level of 40 cmH_2O in the cystitis rats compared to controls ($P = 0.03$) (Figure 4). SCS had no significant effect in the control rats. SCS significantly reduced the VMR AUC at the IVP levels of 20, 30, and 40 in the cystitis rats ($P = 0.04$, 0.03, and 0.03, respectively) (Figure 4).

4 | DISCUSSION

The present study investigated the effect of SCS at the dorsal surface of the L3 spinal segment in attempts to modulate DO and bladder hyperalgesia in rats with CYP-induced cystitis. CYP-induced cystitis elicited DO as evidenced by increased non-voiding contractions and shorter inter-contraction intervals. CYP-induced cystitis also induced visceral hyperalgesia as demonstrated by the increased VMR AUC. SCS significantly reduced DO, improved voiding function, and reduced signs of visceral pain in cystitis rats. Previously, SCS was applied over L3 to induce EUS bursting in the neurologically intact and spinal cord injured rats, but cystometry was not investigated.[2] Previous studies have shown that a variety of neuromodulation targets to the sacral and/or lumbosacral spinal nerves significantly reduce voiding frequency and/or non-voiding contractions.[1,7,14–16] The present study is the first to demonstrate that CYP-induced DO is reduced by L3 SCS.

4.1 | Poor voiding efficiency in the cystitis rats

It has been reported that accumulation of acrolein, a hydroxylated metabolite of CYP, induces irritation of the urothelium resulting in bladder inflammation.[17,18] During inflammatory reaction, the C-fiber afferents are activated and elicit detrusor over-activity, resulting in a reduction in bladder capacity and a consequent increase in contraction frequency.[19] CYP-induced cystitis, which is an established and clinically relevant model of the study of visceral pain.[6] The resulting DO causes decreased voiding efficiency and bladder capacity.[20] The present study demonstrated that CYP-induced cystitis elicited the significant increases of non-voiding contractions and residual volumes as well as a

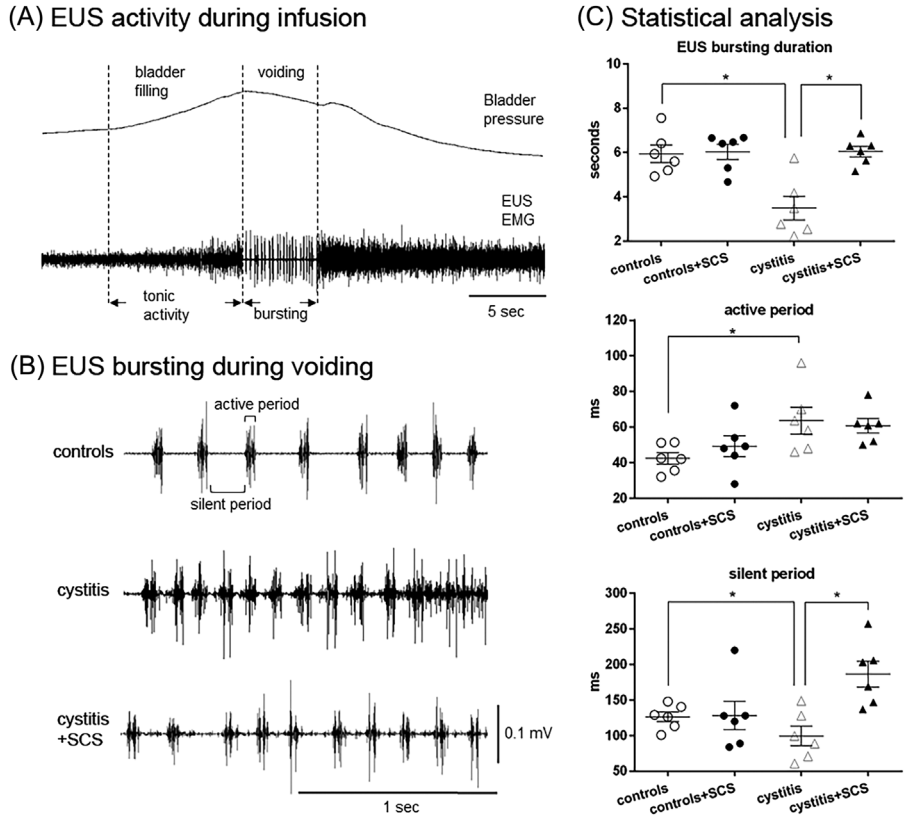

FIGURE 3 EUS EMG activity during bladder infusion in controls and cystitis rats before and 2 h after SCS. EUS EMG activity consisted of tonic activity during filling and EUS bursting during voiding (A). EUS bursting is composed of active periods and silent periods (B). Statistical analysis of EUS bursting duration including active and silent periods was shown here (C). The cystitis rats had significant shorter bursting duration composed of longer active periods and shorter silent periods. SCS had a significant increase in bursting period with longer silent periods in the cystitis rats (cystitis + SCS). * indicated the $P < 0.05$

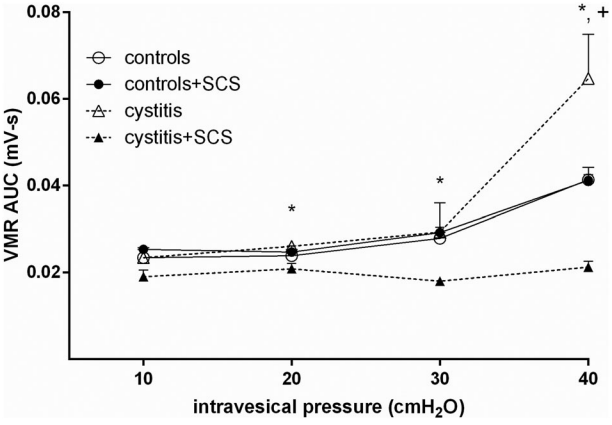

FIGURE 4 Effect of SCS on the visceromotor reflex (VMR) induced by isotonic bladder distension (IBD). The bladder was distended using saline from 10 cmH_2O to 40 cmH_2O. Prior to SCS, cystitis rats showed a significantly increase in the VMR AUC. Two hours after SCS, the inhibitory effect on the VMR AUC during IBD was significantly shown at 20, 30, and 40 cmH_2O in the cystitis rats (cystitis + SCS). + indicated $P < 0.05$ between the control and cystitis groups. * indicated the $P < 0.05$ between the cystitis and cystitis + SCS groups

significant decrease of inter-contraction intervals as previously reported.[18–20] These changes resulted in poor voiding efficiency in the cystitis rats compared to controls.

The present study further examined the EUS EMG activity during filling and voiding in both control and cystitis groups. EUS bursting in rodents demonstrates intermittent urethral relaxation and opening of the outlet, which is essential for achieving efficient voiding.[13] In cystitis rats, there were significant decreases of EUS bursting duration and silent periods, suggesting a decrease of urethral relaxation. It is likely this was playing a role in inefficient voiding observed in the present setting.

4.2 | SCS promoted voiding function

Neuromodulation with SCS does not seem to affect the normal physiological status in controls. SCS only modulated the irregular reflex pathways induced by cystitis. Assessment of DO and urodynamic parameters immediately (ie, 25 min; Figure 1) after SCS revealed modest changes. However, assessment of the same parameters 2 h after SCS demonstrated profound changes. This suggests that the mechanisms

underlying SCS modulation of bladder function are time-dependent. These could involve changes in synaptic circuits in the spinal cord involving short term plasticity and/or peripheral mechanisms. Whether the relief of DO lasts for longer time was not determined in this study, and future studies are necessary to test the same parameters at different times after SCS.

Our previous study indicated that L3 could be the EUS bursting controlling center in spinal cord injured rats as well as that SCS over L3 resulted in EUS bursting with intermittent urethral relaxation in control rats when the bladder was partially full.[2,10] Here, SCS induced prolonged silent periods during EUS bursting in the cystitis rats and showed the similar result of urethral relaxation during voiding.

Combined together, SCS over L3 modulated urethral relaxation during voiding. SCS seemed to modulate the lumbosacral spinal neurons receiving inputs from urinary bladder.[2,5,10] Although the urinary bladder and urethra are mainly innervated via the lumbosacral levels, SCS over L3 may still modulate micturition reflexes by interfering with signals among the bladder, the lumbosacral spinal cord, and the pontine micturition center.[2,10]

4.3 | SCS ameliorates bladder hyperalgesia

Cystitis changes the chemical communication between the urothelium and C-fiber afferent nerves, resulting in afferent sensitization. Released neurotransmitters from the irritated urothelium enhance signaling between the urothelium and afferent nerves and alter the second-messenger pathways, triggering painful bladder sensation.[21] During saline infusion to the bladder, the appearance of VMR associated with voiding contractions was induced earlier in cystitis rats compared to controls. Furthermore, the bladder was distended by saline to a constant IVP level between 10 to 40 cmH_2O. This range was used to grade mechanical stimuli of the bladder from innocuous to noxious.[12,22] This hypersensitivity in the cystitis rats was evident by significantly increased VMR AUC when the IVP reached the voiding threshold of 40 cmH_2O compared to controls. SCS seems suppressing the C-fiber activity in the CYP-induced cystitis, but more future investigations are needed to identify the inhibitory effects.

Our results also showed that the similar effect of SCS over L3 on the VMR in response to bladder distension in cystitis rats lasted 2 h. In addition, the effect of SCS over the upper lumbar cords on the reduced VMR in response to noxious colonic distention lasted 90 min.[4] Recent studies revealed that SCS increases blood flow via antidromic activation of sensory afferents to release neuromodulatory substances at the target organ.[23,24] This mechanism may play a role in the inhibitory effect of SCS on the VMR induced by bladder distention.

5 | CONCLUSION

In summary, the present study provided a novel method of SCS over the upper lumbar levels, specifically L3, to ameliorate DO, modulate urethral intermittent relaxation, and suppressed cystitis-induced bladder hyperalgesia. The present rodent model demonstrated the severity of the cystitis induced by the hazardous chemical irritation. Although it is not ideal to represent IC/PBS, SCS markedly improved the voiding function and ameliorated bladder hyperalgesia. This is a pre-clinical study with limited extrapolation to patients. Current technologies of SCS are well developed on the managements of spasticity and neuropathic pain, but are limited on the visceral pain.[25] The mechanisms involved in such modulation of visceral pain by SCS are currently unclear in both animal and human studies. The present results may support SCS as a potential therapeutic method for patients with hyperalgesia and IC/PBS.

ACKNOWLEDGMENTS

The authors thank Dr. F. Aura Kullmann at University of Pittsburgh and Dr. Oswald Steward at University of California Irvine for critical and helpful comments on this manuscript.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest to disclose.

ORCID

Huiyi H. Chang http://orcid.org/0000-0001-6439-2320
David A. Ginsberg http://orcid.org/0000-0002-0402-8970

REFERENCES

1. Hussain Z, Harrison SC. Neuromodulation for lower urinary tract dysfunction-an update. *ScientificWorldJournal*. 2007;7:1036–1045.
2. Abud EM, Ichiyama RM, Havton LA, Chang HH. Spinal stimulation of the upper lumbar spinal cord modulates urethral sphincter activity in rats after spinal cord injury. *Am J Physiol Renal Physiol*. 2015;308:F1032–F1040.
3. Chang HH, Yeh JC, Ichiyama RM, Rodriguez LV, Havton LA. Mapping and neuromodulation of lower urinary tract function using spinal cord stimulation in female rats. *Exp Neurol*. 2018;305:26–32.
4. Greenwood-Van Meerveld B, Johnson AC, Foreman RD, Linderoth B. Attenuation by spinal cord stimulation of a nociceptive reflex generated by colorectal distention in a rat model. *Auton Neurosci* 2003;104:17–24.
5. Qin C, Farber JP, Foreman RD. Spinal cord stimulation modulates activity of lumbosacral spinal neurons receiving input from urinary bladder in rats. *Neurosci Lett*. 2007;428:38–42.

6. Boucher M, Meen M, Codron JP, Coudore F, Kemeny JL, Eschalier A. Cyclophosphamide-induced cystitis in freely-moving conscious rats: behavioral approach to a new model of visceral pain. *J Urol*. 2000;164:203–208.
7. Giuliano FA, Denys P, Chartier-Kastler E, Alexandre L, Bernabe J. L6-S1 spinal nerve stimulation reduces micturition frequency in anaesthetized rats with cyclophosphamide-induced cystitis. *BJU Int*. 2006;97:386–392.
8. Shafik A, el-Sibai O, Ahmed I. Effect of urethral dilation on vesical motor activity: identification of the urethrovesical reflex and its role in voiding. *J Urol*. 2003;169:1017–1019.
9. Danziger ZC, Grill WM. Sensory and circuit mechanisms mediating lower urinary tract reflexes. *Auton Neurosci*. 2016; 200:21–28.
10. Chang HH, Yeh JC, Ichiyama RM, Rodriguez LV, Havton LA. Mapping and neuromodulation of lower urinary tract function using spinal cord stimulation in female rats. *Exp Neurol*. 2018; 305:26–32.
11. Gao Y, Zhang R, Chang HH, Rodriguez LV. The role of C-fibers in the development of chronic psychological stress induced enhanced bladder sensations and nociceptive responses: a multidisciplinary approach to the study of urologic chronic pelvic pain syndrome (MAPP) research network study. *Neurourol Urodyn*. 2018;37:673–680.
12. Kullmann FA, Chang HH, Gauthier C, et al. Serotonergic paraneurones in the female mouse urethral epithelium and their potential role in peripheral sensory information processing. *Acta Physiol (Oxf)*. 2018;222. https://doi.org/10.1111/apha.12919
13. Cheng CL, de Groat WC. The role of capsaicin-sensitive afferent fibers in the lower urinary tract dysfunction induced by chronic spinal cord injury in rats. *Exp Neurol*. 2004;187:445–454.
14. Wang Y, Zhou Y, Mourad MS, Hassouna MM. Neuromodulation reduces urinary frequency in rats with hydrochloric acid-induced cystitis. *BJU Int*. 2000;86:726–730.
15. Riazimand SH, Mense S. Interaction between neurotransmitter antagonists and effects of sacral neuromodulation in rats with chronically hyperactive bladder. *BJU Int*. 2005;96:900–908.
16. Su X, Nickles A, Nelson DE. Neuromodulation in a rat model of the bladder micturition reflex. *Am J Physiol Renal Physiol*. 2012;302: F477–F486.
17. de Groat WC. The urothelium in overactive bladder: passive bystander or active participant? *Urology*. 2004;64:7–11.
18. Meotti FC, Forner S, Lima-Garcia JF, Viana AF, Calixto JB. Antagonism of the transient receptor potential ankyrin 1 (TRPA1) attenuates hyperalgesia and urinary bladder overactivity in cyclophosphamide-induced haemorrhagic cystitis. *Chem Biol Interact*. 2013;203:440–447.
19. Andersson KE, Soler R, Fullhase C. Rodent models for urodynamic investigation. *Neurourol Urodyn*. 2011;30:636–646.
20. Juszczak K, Ziomber A, Wyczolkowski M, Thor PJ. Urodynamic effects of the bladder C-fiber afferent activity modulation in chronic model of overactive bladder in rats. *J Physiol Pharmacol*. 2009;60:85–91.
21. Malley SE, Vizzard MA. Changes in urinary bladder cytokine mRNA and protein after cyclophosphamide-induced cystitis. *Physiol Genomics*. 2002;9:5–13.
22. Gao Y, Zhang R, Chang HH, Rodriguez LV. The role of C-fibers in the development of chronic psychological stress induced enhanced bladder sensations and nociceptive responses: a multidisciplinary approach to the study of urologic chronic pelvic pain syndrome (MAPP) research network study. *Neurourol Urodyn*. 2018;37: 673–680.
23. Croom JE, Foreman RD, Chandler MJ, Koss MC, Barron KW. Role of nitric oxide in cutaneous blood flow increases in the rat hindpaw during dorsal column stimulation. *Neurosurgery*. 1997;40:565–570. discussion 571.
24. Tanaka S, Barron KW, Chandler MJ, Linderoth B, Foreman RD. Low intensity spinal cord stimulation may induce cutaneous vasodilation via CGRP release. *Brain Res*. 2001;896:183–187.
25. Kapural L, Nagem H, Tlucek H, Sessler DI. Spinal cord stimulation for chronic visceral abdominal pain. *Pain Med*. 2010;11:347–355.

How to cite this article: Chang HH, Yeh J-C, Mao J, Ginsberg DA, Ghoniem G, Rodriguez LV. Spinal cord stimulation ameliorates detrusor over-activity and visceromotor pain responses in rats with cystitis. *Neurourology and Urodynamics*. 2019;38:116–122. https://doi.org/10.1002/nau.23827