Micturition Reflex Elicited by Stimulation of the Pudendal Nerve

**Boggs JW** 1,2, **Wenzel BJ** 1,2, **Gustafson KJ** 1, **Grill WM** 2

1 Department of Biomedical Engineering, Case Western Reserve University, Cleveland, OH
2 Department of Biomedical Engineering, Duke University, Durham, NC

warren.grill@duke.edu

Abstract

Persons with spinal cord injury cannot voluntarily contract their bladder to produce micturition (bladder emptying). A micturition reflex can be initiated and sustained in cats after spinal cord transection (SCT) through activation of afferent fibers in the deep perineal nerve (DP), but placing an electrode near the DP is surgically cumbersome. This study examined the ability to activate selectively the DP afferent pathway contained in the surgically accessible pudendal nerve trunk (PN) to generate a micturition-like response.

A micturition-like bladder contraction without urethral sphincter contraction was obtained in response to PN stimulation with the same parameters capable of producing a micturition-like response to DP stimulation. Similar to DP stimulation, PN stimulation only evoked bladder contractions when the bladder contained sufficient volume both pre- and post-SCT. PN stimulation evoked micturition comparable to that of distention-evoked reflex contractions.

The ability to activate a micturition reflex from the compound pudendal nerve trunk holds promise for a neural prosthesis capable of restoring bladder emptying to persons with SCT.

1. INTRODUCTION

There are two complementary phases in the micturition cycle: continence, when the bladder stores urine, and micturition, when the bladder contracts and the external urethral sphincter relaxes to void urine. Micturition can be inhibited and continence augmented by stimulation of the compound pudendal nerve (PN) [13] via activation of genital [12] and/or anal [10] sensory pathways in the pudendal nerve. The pudendal nerve also contains urethral sensory pathways that initiate a micturition-like response when they are activated independently of the genital and anal branches [3,4,9]. The objective of this study was to determine if electrical stimulation of the compound pudendal nerve trunk could activate a micturition-like response.

2. METHODS

Acute experiments were conducted in 6 sexually intact, adult male cats (3.3–5.8 kg) initially anesthetized with ketamine HCl (35 mg/kg, i.m. and maintained with α-chloralose (65 mg/kg, i.v., supplemented at 15 mg/kg). Blood pressure was measured through a catheter placed in the carotid artery, body temperature was maintained between 38°C and 39°C with a heating blanket, 0.9% saline with 8.4 mg/ml sodium bicarbonate and 5% dextrose added was administered i.v. (10-15 ml/kg/h), and artificial respiration was used to maintain end tidal CO₂ between 3-4%. The bladder was exposed via a ventral midline incision. The ureters were ligated, transected proximal to the ligation, and drained externally. The bladder was cannulated with a catheter to measure bladder pressure, and the urethra was occluded with a catheter. After collection of data with the spinal cord intact, the spinal cord was transacted (T12) and the response to pudendal nerve stimulation was measured again in 3 animals.

A solid-state pressure transducer connected to the suprapubic catheter (Deltran DPT-100, Utah Medical Products, Midvale, UT) was used to measure bladder pressure. The pressure signal was amplified, low pass filtered (cutoff frequency = 300 Hz), recorded continuously on a strip chart recorder, sampled at 24 kHz, and stored digitally on tape. The pudendal nerve was isolated and stimulated through platinum bipolar hook electrodes with monophasic cathodic pulses with duration of 100 μs, amplitude of 100 μA – 900 μA, and a pulse frequency between 2 Hz and 40 Hz. The minimum current amplitude required to elicit bladder contractions was called threshold (PNthr) and ranged from 100 μA to 450 μA (n = 4 cats). The data presented in this study were obtained at 2*PNthr to 4*PNthr.

The electroneurogram (ENG) activity of fibers innervating the external urethral sphincter
(EUS) was recorded through a tripolar cuff electrode placed on the contralateral deep perineal branch of the pudendal nerve. The ENG was preamplified (100x) and filtered (300 Hz – 10 kHz), further amplified (1,000x) and filtered (300 Hz – 3 kHz), sampled at 24 kHz, and stored digitally on tape. The deep perineal branch ipsilateral to stimulation was transected distal to the trunk of the pudendal nerve prior to ENG recording to prevent direct activation of the EUS and the resulting afferent response to the EUS contraction that would be recorded by the contralateral cuff electrode. An overdose of sodium pentobarbital was used to euthanize the animal at the conclusion of the experiment.

3. RESULTS

Stimulation of the compound pudendal nerve trunk (PN) evoked bladder contractions (n = 6 of 6 cats) without an increase in urethral sphincter ENG activity (n = 3 of 3 cats) both before and after (n = 3 of 3 cats) acute spinal transection (SCT). The response was dependent on stimulus frequency and bladder volume before and after SCT. Increasing stimulation amplitudes beyond twice the threshold to evoke a bladder contraction did not increase the magnitude of the bladder contractions nor did it change the ability of the stimulation to sustain a bladder contraction. The duration of the response was linked to stimulation because when stimulation ended, bladder pressure and sphincter ENG activity typically returned to baseline. Immediately following SCT, PN stimulation did not elicit bladder contractions without increases in EUS ENG activity, but the synergic responses returned as time progressed past acute spinalization.

PN stimulation evoked a micturition-like response analogous to the micturition-like response evoked by DP stimulation (Fig 1). Stimulation evoked an increase in bladder pressure that was sustained for the duration of stimulation. Stimulation elicited a transient burst in EUS ENG, but the ENG activity returned to the pre-stimulus level as bladder pressure increased.

Stimulation frequency determined the character of the bladder pressure responses to PN stimulation similar to that of DP stimulation (p < 0.01 for responses to both PN and DP stimulation). Higher frequencies (20 – 40 Hz) were more successful in generating sustained bladder contractions than lower frequencies (< 20 Hz) before and after SCT (p < 0.05 for pre and post SCT) (Fig. 2).

The bladder response to PN stimulation and DP stimulation depended on bladder volume in all cats. The bladder was filled in 1 ml increments and stimulation was applied to the PN or DP between each bolus of saline. PN and DP stimulation could evoke bladder contractions only when the bladder contained more than a minimum threshold volume. PN stimulation elicited contractions at 66 ± 17% of the volume required for the saline bolus to evoke a contraction (V_{thr}) (n = 12 cats), and DP stimulation evoked contractions at 78 ± 17% of V_{thr} (n = 4 cats). The minimum bladder volume necessary to evoke contractions via stimulation decreased slightly but not significantly after SCT (p > 0.10).

The ability to evoke micturition through PN stimulation was validated in 2 cats by stimulating the pudendal nerve after ipsilateral transection of the PN branch innervating the

![Fig. 1 Responses evoked by selective and non-selective stimulation of pudendal nerve afferents. (A) The deep perineal nerve (DP) is a branch of the pudendal nerve (PN) (adapted from [6]). (B) PN stimulation evoked a sustained increase in bladder pressure (top black trace) without a sustained increase in EUS ENG activity (bottom gray trace). (C) A similar micturition-like response was evoked by DP stimulation.](image)
Fig. 2 PN stimulation elicits bladder contractions in response to the higher stimulus frequencies similar to DP stimulation. Stimulation of the PN evokes a greater percentage of sustained contractions with high frequency stimulation before (A) and after (B) acute spinal cord transection (SCT). Stimulation of the DP also evokes a greater percentage of sustained contractions with high frequency stimulation before (C) and after (D) SCT. Stimulation amplitude was 500 - 900 µA. Bladder volumes were above the minimum volume required to evoked bladder contractions.

urethral sphincter. In both cats, PN stimulation generated voiding (56 ± 10%, n = 4 trials across 2 cats) comparable to voiding produced by DP stimulation (63 ± 20% n = 3 trials across 2 cats) and bladder emptying produced by distention-evoked bladder contractions (42 ± 8%, n = 4 trials across 2 cats).

4. DISCUSSION AND CONCLUSIONS

Stimulation of the compound pudendal nerve trunk (PN) evoked a coordinated micturition-like response before and after spinal cord transection (SCT), consistent with the micturition-like response elicited by deep perineal nerve (DP) stimulation. Similar to DP stimulation, the micturition-like response could be produced only if the bladder contained a sufficient volume and stimulation was applied at a frequency greater than 20 Hz. The coordinated increase in bladder pressure and reduction in external urethral sphincter (EUS) ENG activity evoked by PN stimulation were confirmed to be a micturition reflex because PN stimulation produced voiding comparable to that produced by distention-evoked bladder contractions.

The present results support future investigation to determine if this micturition reflex can be activated in persons with chronic spinal cord injury. The ability to access a spinal micturition reflex at the level of the compound pudendal nerve trunk suggests the potential to develop a neural prosthesis to restore bladder emptying to persons with spinal cord injury.

References
[2] Barrington FJF. The component reflexes of micturition in the cat, Parts I and II. Brain 54: 177-188, 1931

Acknowledgements
This work was supported by NIH grants R21 NS-43450 (WMG), R01 NS-50514 (WMG), K25 HD-40298 (KJG), and a Whitaker Graduate Student Fellowship (BJW).