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Urothelial Mucosal Signaling and the Overactive Bladder-ICI-RS 2013

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Abstract

There is abundant evidence that the lower urinary tract (LUT) mucosal layer is involved both in mechanosensory functions that regulate bladder contractile activity and in urethral sensation. Changes to the mucosa can be associated with a number of bladder pathologies. For example, alterations of the urothelium and underlying lamina propria at both the molecular and structural levels have been reported in both patients and animals associated with disorders such as bladder pain syndrome and diabetic cystopathy. In contrast to the urinary bladder, much less is known about the urothelium/lamina propria of the bladder neck/proximal urethra. There are important gender differences in the outflow region both anatomically and with respect to innervation, hormonal sensitivity, and location of the external urethral sphincter. There is reasonable evidence to support the view that the mucosal signaling pathway in the proximal urethra is important for normal voiding, but it has also been speculated that the proximal urethra can initiate bladder overactivity. When dysfunctional, the proximal urethra may be an interesting target, for example, botulinum toxin injections aiming at eliminating both urgency and incontinence due to detrusor overactivity.

Keywords

bladder nerves; epithelium; lamina propria; neuroendocrine; overactive bladder; sensation; urethra

INTRODUCTION

The urothelium as a part of a mucosal signaling pathway¹ has attracted increasing attention for its potential role in the pathophysiology of the overactive bladder syndrome/detrusor overactivity (=OAB). Most of the lower urinary tract (LUT) afferents are concentrated to the

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outflow region² (Fig. 1), which implies that the urothelium in this part may be of special importance for the generation of both normal and abnormal bladder activity. Since the outflow region in men and women are distinctly different, both anatomically and with respect to innervation and hormonal sensitivity, gender-dependent factors have to be considered when urothelial afferent signaling mechanisms from the urethra are discussed. For example, the female bladder neck is lacking the smooth muscle sphincter structure that serves as a protection against retrograde ejaculation in males (Fig. 2), and also the dense adrenergic innervation seen in the male bladder neck^{3,4} (Fig. 3). In addition, the location of the external striated sphincter is different (Fig. 2). In the human female LUT, estrogen receptors are found in squamous epithelia of the trigone and urethra, but not in bladder urothelial tissues⁵ (Fig. 4). There is no variation with estrogen status. In contrast, estrogen receptors (both alpha and beta) have been found in the rodent urothelium.^{6,7} In the human male LUT, estrogen receptors are found in the urethral epithelium, lamina propria, and periurethral glands.⁸

BLADDER AND URETHRAL EPITHELIUM

A prerequisite for conscious bladder control is adequate sensory input to the central nervous system (CNS), and it is well established that changes in afferent mechanisms may give rise to disturbances of bladder function. Morrison⁹ (referring to Nathan)¹⁰ stated that “the sense of filling and the desire to micturate probably depend on afferent endings in the bladder itself, whereas the sensation that micturition is about to happen probably originates from the urethra, as does the sensation that urine is being passed.” In addition, a variety of structures play an important role in terms of urethral closure (such as the urethral epithelium, vasculature, and smooth muscle) that are necessary to maintain continence.¹¹ The urothelium, forms the interface between the urinary space and the underlying lamina propria that contains a dense vasculature as well as connective, nervous, and muscle cells (muscularis mucosae).^{1,12,13}

In contrast to the urinary bladder, where the urothelium is composed of at least three layers of epithelial cells, very little has been described regarding the urethral epithelium. There seems to be no apparent difference between the urothelium of the trigone compared to the detrusor.¹⁴ This is in contrast to the proximal urethra where the urothelium transitions to a stratified or columnar epithelium accompanied by a lack of urothelial-specific differentiation markers.^{15,16} In terms of the underlying structures that are likely to contribute to function, there are similarities to that of the bladder body. For example, the urethral epithelium is likely to be part of a signaling system involving projections of neuroendocrine cells, interstitial cells and sensory nerve endings. There is speculation that these urethral-neuroendocrine cells (sometimes termed paraneurons)¹⁷ could release mediators such as serotonin, which via activation of adjacent sensory nerves can stimulate urethral reflexes. Such types of cells are not unlike that in other types of epithelia, such as the trachea, where a cell type termed brush cells have been described which are likely chemo-receptive and make contact with nearby nerve fibers.¹⁸ There is also a rich vascular network that lies beneath and a dense distribution of nerves throughout the bladder neck and initial part of the urethra^{2,19} where the nerves form a plexus adjacent to the urothelial lining.

There is some evidence that the urothelium in the region of the urethra may play a role in continence and sensation. The mucosal pathway (often referred to as a sensory web)^{1,12,20} within the proximal urethra also involves a cascade of epithelial inhibitory and stimulatory transmitters/mediators. Release of these factors may be involved in a complex transduction scheme underlying the activation of bladder nerves to play a prominent role in sensation. In addition, it has been suggested that symptoms of pain that arise from the lower urinary tract may originate from the bladder neck and proximal urethra.²¹ The bladder neck and proximal urethral contain the largest density of nerves^{2,19} and the epithelial cells that line the surface exhibit “neuronal-like” properties including expression of proteins sensitive to chemical and physical stimuli.²⁰ The proximity of afferent nerves to the epithelium suggests that epithelial cells could be targets for transmitters released from nerves and/or that chemicals released by epithelial cells influence afferent nerve excitability. Thus, urethral epithelial-neural interactions (via release of mediators) may lead to a “urethral instability” that can influence storage and voiding reflexes and result in symptoms including urgency and pain.

URETHRAL SENSATION

Sensory signals from the urethra arise during urine flow and a fundamental question arises whether urethral stretch and/or shear stress caused by urine flow contribute to sensory signals. By analogy with the bladder, lateral stretch of the urothelium releases transmitters such as ATP²² as does equivalent relaxation.²³ This therefore suggests that there is a dynamic component to stretch in the urothelium, which in the urethra would derive from variations in urine flow.

Sensory signals from urethral afferents can initiate detrusor contractions and also maintain this contraction by positive feedback through continuous flow of urine through the urethra.^{24,25} If this urethro-vesical reflex is eliminated by urethral anesthesia, the detrusor contraction fails and excessive straining is needed to achieve bladder evacuation in multiple spurts.²⁵

There is evidence for slowly and rapidly adapting afferents to steady urethral flow,²⁶ indicating that both the rate and duration of urine flow may be sensed. However, the afferent nerve population is not homogenous, and more than one population of sacral interneurons seem to be involved.²⁷ This further suggests that discharge may depend on both the urethral flow itself and the rate of change of flow.

The mechanism of afferent activation remains unclear, but by analogy with the bladder itself one possibility is that shear stress on the urethral urothelium induces the release of transmitter molecules to mediate the effect. In other tubular structures, such as blood vessels shear-stress releases mediator molecules such as ATP from the endothelium²⁸ and a hydrodynamic analysis would indicate a similar phenomenon is feasible in the urethra. In the left-anterior descending coronary artery (diameter 3.5–4.0 mm) peak diastolic velocity is about 22 ml/sec.^{29,30} A similar maximum urine flow rate (25 ml/min) occurs in normal adult men and women through a urethra that varies in diameter from 1 to 12 mm.^{31,32} These two variables are key determinants of the shear stress on a vessel wall³³ and their similar values

in blood vessels and the urethral walls and will suggest that flow-mediated shear stress could indeed evoke mediator release from the urethral wall.

The evidence for release of mediators from urethral urothelium is weaker than in the bladder. Histological evidence shows that urothelial cells secrete mucus, similar to that of other cells with microvilli and small granules and which can be interpreted as evidence of secretion.³⁴ Endogenous nitric oxide (NO) synthase is present in urethral urothelium³⁵ and nitric oxide may be released during stress either from lamina propria nerves or from the urothelium itself.^{36,26} Local prostaglandin release may influence urethral function as exogenous PGE2 reduces urethral resistance.³⁷ Evidence for stress-activated release of ATP or acetylcholine from urethral urothelium, as occurs in bladder urothelium, is lacking.

THE PROXIMAL URETHRAL MUCOSA AS A POSSIBLE INITIATOR OF OVERACTIVE BLADDER

The pathophysiology of lower urinary tract (LUT) symptoms, including the overactive bladder syndrome, is multifactorial³⁸ and may involve various LUT structures. As mentioned previously, the distal urethral epithelium is composed of stratified squamous cells, arranged in longitudinal folds, which variably becomes transitional as the bladder neck is approached. The epithelium is supported by a loose lamina propria, consisting of collagen fibrils and elastic fibers, arranged both circularly and longitudinally, and a rich network of blood vessels and nerves.¹³

The epithelium together with the interstitial cells and the suburothelial afferent nerves have been suggested to form a functional unit (“the mucosal signaling pathway”) able to initiate a bladder contraction.³⁹ Gabella and Davis,² studying the distribution of bladder afferent nerves in the rat bladder, found afferent axons distributed over four distinct areas: at the base of the epithelium, inside the epithelium, on blood vessels (both arteries and veins) and along muscle bundles. In the lamina propria, all the afferent axons, except the perivascular ones, were found either inside the epithelium or in a subepithelial plexus very close to the basal surface of the epithelium. The plexus was thickest in the neck of the bladder and in the proximal portion of the urethra, and it became progressively less dense in the adjacent regions. The cranial region of the bladder had no afferent axons.

The combination of a urothelial mechanosensory function¹ and the distribution of afferent nerves have focused interest on the proximal urethra as an initiator of detrusor overactivity, and several previous studies support such a possibility. Tanagho and Miller⁴⁰ observed that urethral relaxation occurs a number of seconds before detrusor contraction under normal circumstances, and Low,⁴¹ demonstrated a similar sequence occurring in female patients with idiopathic detrusor overactivity. The presence of urethral pressure variations during filling cystometry (urethral instability) has been well documented by various investigators, and said to be responsible for the sensation of urgency and for incontinence.⁴² Mechanical stimulation of the urethra, especially the flow of urine, may facilitate the micturition reflex as mentioned above, and reflexes between the urethra and bladder play an integral role in the neural control of the lower urinary tract.²⁵ Studies in animals have demonstrated the basis for an excitatory urethra-to-bladder reflex, showing that sensory nerves in the wall of the

urethra fire in response to urethral urine flow and electrical stimulation, and that this activity initiates bladder contractions in the quiescent bladder and augments ongoing contractions in the active bladder. Mazieres et al.⁴³ showed in cats that electrical stimulation of pudendal urethral sensory nerves can activate bladder efferent neurons and initiate detrusor contractions. Urethra-to-bladder reflexes thus seem to be mediated by afferent inputs traveling through the pudendal nerve to the sacral spinal cord⁴⁴ and brain.^{45,46} Attempts to identify an excitatory urethra-to bladder reflex in humans using urethral urine flow have been difficult,^{25,47} and produced results either opposing or supporting the presence of such a reflex. Gustafson et al. stimulated the prostatic urethra electrically via a catheter-based electrode in five men with complete spinal cord injury and succeeded in initiating detrusor contractions in four of five individuals when the bladder volume was sufficiently large.⁴⁸ These results demonstrate in humans the presence of a volume-dependent excitatory bladder reflex mediated by urethral afferent nerve fibers. The neural circuitry was suggested to exist in the lumbosacral spinal cord not requiring a brainstem pathway.

There is thus reasonable evidence supporting the view that mucosal signaling pathways in the proximal urethra are important for normal voiding, and that when dysfunctional can be an interesting target for example, botulinum toxin.^{49,50} The ability of botulinum toxin injected into the urethral suburothelial space to block afferent nerves involved in urgency and DO remains to be established. The urethral mucosal signaling pathway is as yet not fully explored and promises to be an interesting and rewarding research field many research questions can be raised.

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RESEARCH QUESTIONS

1. Which are the mechanisms by which bladder and urethral epithelial cells, when exposed to different sensory “inputs” (mechanical, chemical), evokes different afferent discharges? In what ways are such mechanisms influenced by pathology?
2. Can urethral flow, or rate of change of flow, differentially effect discharge from urethral afferents?
3. Is urethral sensitivity to urine flow is modulated by exogenous agents (ATP, acetylcholine)?
4. If the proximal urethra is a generator of detrusor overactivity—then how does treatments (blocking nerves in the suburothelial space) affect such overactivity?

Fig. 1.
Whole mounts of rat bladder stained with calcitonin-gene related peptide (CGRP) antibodies from different regions.²

Fig. 2.

Anatomical gender difference. Green color: urethral smooth muscle; red color: striated urethral muscle. Note the presence of an internal smooth muscle sphincter in the male (arrow 1: “sexual” sphincter, preventing retrograde ejaculation), the location of the external urethral sphincter in the male (arrow 3), and the presence of the prostate (arrow 2).

Fig. 3.
Gender difference on the distribution of noradrenergic nerves in the human bladder neck.⁵¹

Fig. 4. Estrogen receptor (ER) distribution in the female LUT. The bladder urothelium (A) is ER-negative, whereas the urethra is ER-positive.⁵