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FUNCTIONAL MECHANISMS INDUCING HYPEREXCITABILITY OF BLADDER AFFERENT NEURONS THROUGH PROSTATE-TO-BLADDER AFFERENT CROSS-SENSITIZATION IN RATS WITH NON-BACTERIAL PROSTATITIS

Hypothesis / aims of study
Patients with chronic prostatitis/chronic pelvic pain syndrome (CP/CPPS) often exhibit irritative bladder symptoms despite no evidence of bladder inflammation. We previously reported that prostatic inflammation induces bladder overactivity through pelvic organ cross-sensitization in rats (2012 AUA). However, the underlying functional mechanisms are not fully elucidated. We therefore examined changes in electrophysiological properties of dichotomized (bladder and prostate) afferent neurons (D-AN) and bladder afferent neurons (B-AN) obtained from prostatitis rats.

Study design, materials and methods
Prostatic inflammation was induced by 5% formalin injection into the prostate of male SD rats. After 1 week, L6-S1 dorsal root ganglia (DRG) were removed and whole cell patch-clamp recordings were performed on dissociated D-AN and B-AN, which were labelled by retrograde axonal transport of fluorescent dyes, Fast Blue (FB) or DiI, injected into the bladder wall or the prostate, respectively. Since the majority of bladder C-fibres are sensitive to capsaicin, FB and/or DiI-labelled cells that exhibited inward currents in response to capsaicin application were selected for evaluation. The mRNA level of an A-type K^+ channel (K_A) subunit, Kv1.4 in D-AN and B-AN was evaluated by real-time RT-PCR using laser-capture microdissection methods.

Results
In the current clamp condition, capsaicin-sensitive D-AN or B-AN from prostatitis rats exhibited lower thresholds for spike activation (-28.3±1.4mV or -28.7±1.4mV) compared to control rats (-19.5±1.1mV or -21.3±2.2mV, respectively) (Fig.1, Table1). The number of action potentials of D-AN or B-AN during a 800 msec depolarizing pulse was significantly increased after prostatitis (5.1±0.7 or 3.5±0.4 spikes) compared to control rats (1.2±0.1 or 1.4±0.3 spikes, respectively) (Fig.1, Table1). In the voltage clamp condition, the peak densities of K_A currents during membrane depolarization to 0 mV were lower in neurons from prostatitis rats than in those from control rats (Fig. 2). Significant differences in current density were detected at depolarizing pulses greater than -20 mV. However, the peak current density of sustained delayed rectifier-type K^+ (K_{DR}) currents in capsaicin-sensitive bladder afferent neurons was not different between control and prostatitis rats. In prostatitis rats, the mRNA level of Kv1.4 α-subunit, which can form K_A channels, was significantly decreased in D-AN and B-AN compared to non-labeled neurons (Fig.3).

Interpretation of results
These results indicate that: (1) capsaicin-sensitive D-AN and B-AN from prostatitis rats exhibited increased cell excitability evidenced by lower thresholds for spike activation and tonic firing pattern, (2) the peak density of K_A currents in capsaicin-sensitive B-AN of prostatitis rats was significantly smaller than that from control rats and (3) prostatic inflammation induced downregulation of Kv1.4 α-subunit expression in prostate afferent neurons and D-AN innervating both organs as well as B-AN that do not innervate the prostate. Thus, it is assumed that bladder overactivity and electrophysiological changes in D-AN and B-AN after prostatic inflammation is induced at least in part by afferent cross-sensitization from the prostate to the bladder through activation of dichotomized afferent pathways that innervate both prostate and bladder.

Concluding message
The present study indicates that excitability of capsaicin-sensitive C-fiber D-AN and B-AN is increased due to reduced K_A channel activity associated with decreased expression of Kv1.4 α–subunit following prostatic inflammation. Thus, prostate-to-bladder afferent cross-sensitization through dichotomized afferents following prostatic inflammation is a potential mechanism inducing irritative bladder symptoms in CP/CPPS

Fig.1
Dichotomized afferent Bladder Afferent

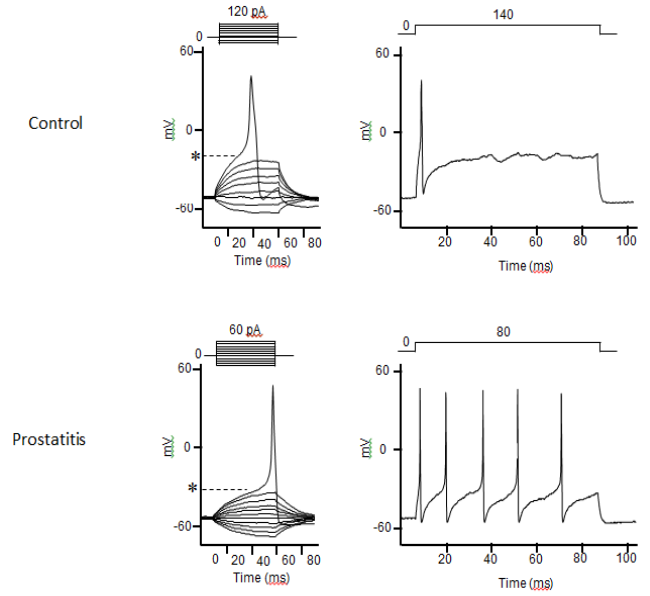


Fig.2

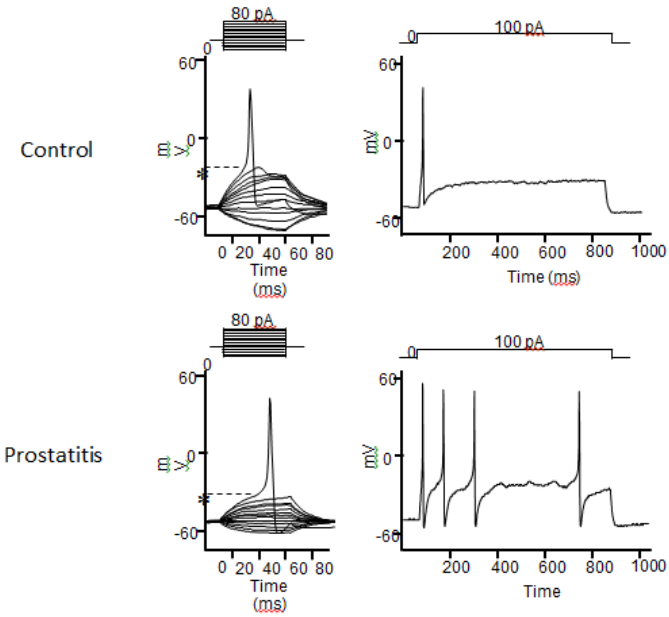
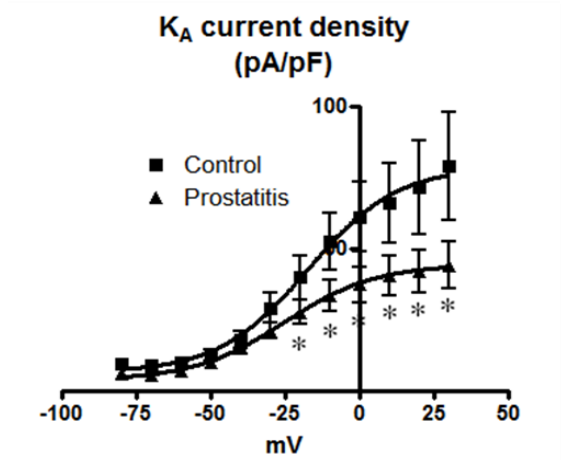


Fig.3



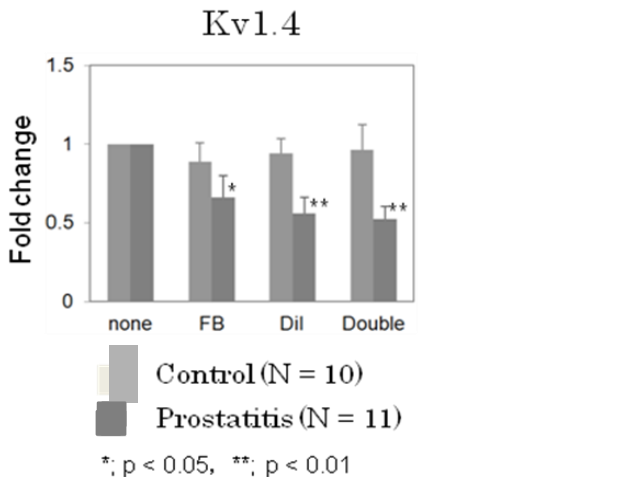


Table 1

	Dual afferent		Bladder afferent	
	Control	Prostatitis	Contro	Prostatiti
No. of cells	8	7	8	10
Diameter (μm)	28.4 ± 0.8	30.4 ± 1.0	27.7 ± 1.1	28.9 ± 0.6
Input capacitance (pF)	27.0 ± 1.5	37.5± 3.2 *	30.8 ± 2.6	33.3 ± 2.4
Membrane potentials (mV)				
Restin	-50.3 ± 1.2	-47.2 ± 1.4	-50.8 ± 0.9	-48.6 ± 0.8
Spike threshold	-19.5 ± 1.1	-28.3 ± 1.4 **	-21.6 ± 2.4	-28.7 ± 1.4 *
Pea	36.3 ± 2.2	46.3 ± 2.6 *	38.2 ± 3.7	39.4 ± 2.6
Spike duration (ms)	6.4 ± 0.9	3.6 ± 0.4 *	5.6 ± 0.7	4.4 ± 0.4
No. of action potentials, 800-ms depo	1.2 ± 0.1	5.1 ± 0.7 **	1.5 ± 0.3	3.5 ± 0.4 **

Data are means ± s.e.m ** P < 0.01 vs. control * P < 0.05 vs. control

Disclosures

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