PRIZE AWARD: Best Basic Science Abstract

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Minagawa T[1], Wyndaele J J[1], Aizawa N[2], Igawa Y[2]

1. *Department of Urology, Antwerp University Hospital and University of Antwerp, Faculty of Medicine, Antwerp, Belgium,* ***2.*** *Department of Continence Medicine, The University of Tokyo Graduate School of Medicine, Tokyo, Japan*

THE ROLE OF TRPA1 CHANNELS IN ACTIVATION OF SINGLE UNIT MECHANOSENSITIVE BLADDER AFFERENT ACTIVITIES IN THE RAT

Hypothesis / aims of study

Transient receptor potential ankyrin 1 (TRPA1) is a member of the TRP channel superfamily, which has been shown to be involved in nociception and mechanosensory transduction in various organ systems. It has been reported TRPA1 ion channels are expressed in the urothelial cells and sensory nerve fibers in the urothelium and suburothelial space of the rat bladder (1), as well as in urothelial cells of the human bladder (2). Moreover, induction of detrusor overactivity by intravesical instillation of TRPA1 activators in the rat and overexpression of epithelial TRPA1 in the bladder and prostatic gland in patients with bladder outlet obstruction (BOO) have been demonstrated (2). These findings suggest that TRPA1 might be involved in the bladder sensory transduction and the induction process of overactive bladder (OAB) by BOO. However, no direct demonstration of such activation of the bladder afferent activities has been reported. Therefore, the aim of the present study is to determine the role of TRPA1 channel in activation of bladder mechanosensory transduction by using cystometry (CMG) and single unit mechanosensitive bladder afferent activities (SAAs) measurement in the rat.

Study design, materials and methods

Female Sprague-Dawley rats were used. CMG and intraarterial blood pressure (BP) measurements were performed with continuous saline instillation at a rate of 0.08 ml/min in conscious and free-moving condition before and after intravenous (i.v.), cumulative administration of HC-030031 (HC, a selective TRPA1 antagonist) at three doses, 0.1, 1.0, and 10.0 mg/kg. In separate rats SAAs measurements were performed under urethane (1.5 g/kg intraperitoneally) anesthesia. Both L6 dorsal roots were cut and fine filaments were dissected from the left L6 dorsal roots and placed across a bipolar electrode for monitoring SAAs. Nerve fibers primarily originating from the bladder were identified by electrical stimulation of the left pelvic nerve and by bladder distension. Nerves with conduction velocities (CV) more than 2.5 m/sec were designated as Aδ-fibers and those with CV less than 2.5 m/sec as C-fibers. The baselines of SAAs were recorded with saline instillation at a rate of 0.08 ml/min until the intravesical pressure reached 30cmH_2O. Then, the SAAs investigation was repeated three times with i.v. cumulative administration of HC at three doses, 0.1, 1.0, and 10.0 mg/kg or i.v. single administration of vehicle (8% dimethyl sulfoxide + 2% Tween 80). In separate experiments, after the baseline investigation with saline instillation, SAAs investigation was repeated three times with intravesical instillation of vehicle (0.01 % of ethanol) or ally isothiocyanate (AI, a TRPA1 agonist,10 μM) with or without pretreatment with HC (1.0 mg/kg i.v., 3 min before each intravesical instillation of AI).

Results

Four rats were used for CMG and BP measurements. There was no significant change in any cystometric parameters or BP after HC-administrations (Table1). A total of 54 SAAs (25 Aδ-fibers; CV: 8.60 ± 6.57 m/sec and 29 C-fibers; CV: 1.12 ± 0.58 m/sec) were isolated in 28 rats. SAAs of both fibers significantly decreased after HC-administrations in a dose-dependent manner (Figs. 1A and 2). And SAAs of both fibers significantly increased after AI-instillations (Figs. 1B and 3). Pretreatment with HC suppressed the excitatory effect of AI-instillation on SAAs (Figs. 1C and 3). Bladder compliance did not change with any of drug-administrations (data not shown).

Interpretation of results

Although no significant cystometric changes were observed with inhibition of TRPA1 channel by HC in conscious rats, mechanosensitive SAAs of both Aδ- and C-fibers were attenuated with HC-administrations in similar dose-ranges in urethane-anesthetized rats. The induction of both fiber-activation with AI, the TRPA1 channel agonist, was counteracted by HC. These findings indicate that there is no major physiological role of TRPA1 channel in the regulation of the micturiton reflex in normal rats, but also suggest that mechanosensitive bladder afferent activities of both Aδ- and C-fibers can be activated by stimulation of TRPA1 channel, and pathologically activated TRPA1 channel may enhance mechanosensation of the bladder, thereby trigger OAB.

Concluding message

TRPA1 channel has a role in activation of mechanosensitive afferent nerve activities of both Aδ- and C-fibers of the rat bladder and might be a possible therapeutic target of OAB

Table 1. The effects of HC-030031 (HC) on cystometric parameters

	Base (before)	After HC 0.1mg/kg	After HC 1.0mg/kg	After HC 10mg/kg
BaP (cmH_2O)	3.57 ± 0.4	3.89 ± 0.22	3.46 ± 0.31	3.38 ± 0.39
MT (cmH_2O)	10.80 ± 0.95	11.77 ± 0.93	9.63 ± 0.16	9.81 ± 1.44
MP (cmH_2O)	35.79 ± 6.07	34.75 ± 3.98	34.74 ± 5.47	37.22 ± 5.82
BC (ml)	1.58 ± 0.30	1.40 ± 0.16	1.48 ± 0.22	1.56 ± 0.26
VV (ml)	1.87 ± 0.30	1.86 ± 0.20	1.96 ± 0.22	1.83 ± 0.32
Mean BP (mmHg)	145.54 ± 7.84	148.92 ± 6.90	144.89 ± 4.8	141.68 ± 3.51

BaP: basal pressure, MT: micturition threshold, MP: micturition pressure, BC: bladder capacity, VV: voided volume, and BP: blood pressure

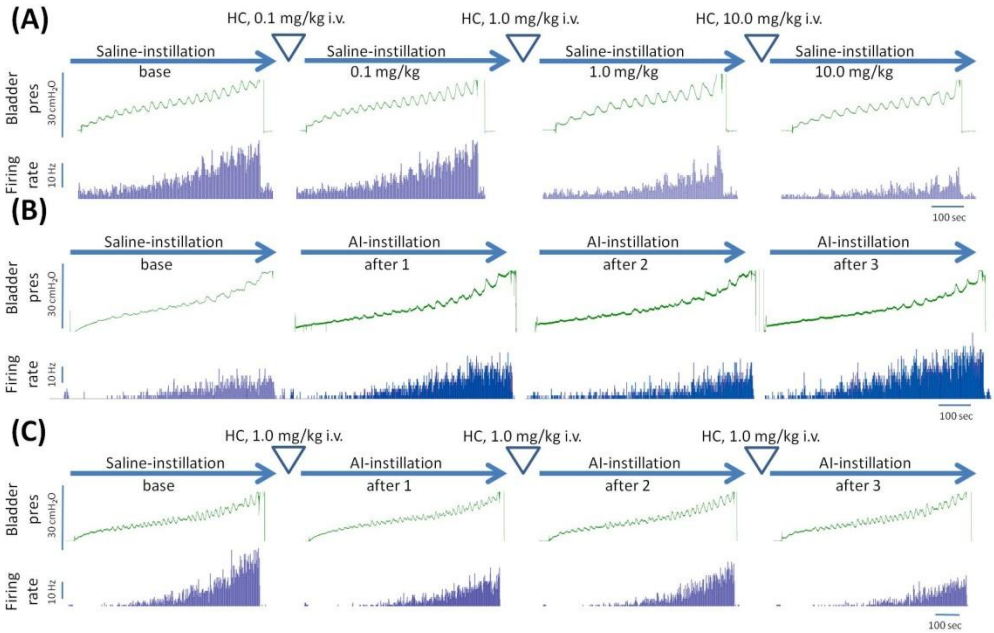

Fig. 1 Representative recordings indicating the effects of TRPA1 antagonist, HC (A), and of intravesial instillation of the TRPA1 agonist, AI, without (B) and with (C) pretreatment with HC on mechanosensitive SAAs of C-fibers during bladder filling .

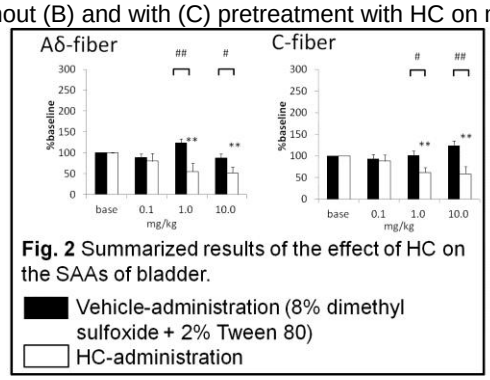

Fig. 2 Summarized results of the effect of HC on the SAAs of bladder.

Vehicle-administration (8% dimethyl sulfoxide + 2% Tween 80)

HC-administration

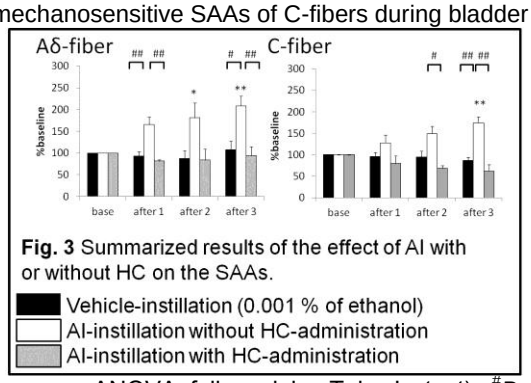

Fig. 3 Summarized results of the effect of AI with or without HC on the SAAs.

Vehicle-instillation (0.001 % of ethanol)

AI-instillation without HC-administration

AI-instillation with HC-administration

*P<0.05, **P<0.01: significant differences from the base (two-way ANOVA followed by Tukey's test). $^{\#}P$<0.05, $^{\#\#}P$<0.01: significant differences between the groups (unpaired Student's t-test).

References

1. Streng T, et al. Distribution and function of the hydrogen sulfide-sensitive TRPA1 ion channel in rat urinary bladder. Eur Urol. 2008; 53:391-9.
2. Du S, et al. Differential expression profile of cold (TRPA1) and cool (TRPM8) receptors in human urogenital organs. Urology 2008; 72: 450–5.

Disclosures

Funding: none **Clinical Trial:** No **Subjects:** ANIMAL **Species:** Rat **Ethics Committee:** The Animal Ethics Committee of the University of Antwerp, Faculty of Medicine