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EFFECTS OF TRPA1 RECEPTOR-AGONISTS ON THE ISOLATED DETRUSOR AND ON MICTURITION IN RATS

Hypothesis / aims of study

Transient receptor potential ion channels (TRPs) respond to e.g. various chemical or thermal stimuli and are likely involved in sensation of pain, heat, cold and taste. Activation of TRPV1 receptors in the urothelial / suburothelial region can produce detrusor overactivity. Similar to the TRPV1 receptor, the TRPA1 receptor has been located to primary sensory neurons which upon activation elicit pain, local vasodilation, inflammation, and hypersensitivity (Bandell et al, Neuron 41, 849, 2004, Bautista et al, PNAS, 102, 12248, 2005). It is not established if the TRPA1-receptor is located to the bladder or if activation of this receptor has any effects on bladder functions in vivo. H_2S has been shown to induce contraction in rat bladder strips and is proposed to activate the TRPA1 receptor (Patacchini et al, Eur J Pharmacol 21(509), 171, 2005). Allyl isothiocyanate (AI) and cinnamon aldehyde (CA) are two other agents that have been shown to act on the TRPA1 receptor in other tissues (Bandell et al, Neuron 41, 849, 2004). In the present study we aimed to study the effects of these TRPA1-agonists on the isolated detrusor and on micturition in awake rats.

Study design, materials and methods

Female Sprague-Dawley rats (200-250 g) were used in this study. For functional in vitro experiments, whole wall detrusor strips were prepared and mounted in tissue baths (37°C) containing aerated Krebs solution (pH 7.4). Isometric tension was recorded during cumulative addition of AI, NaHS, CA, and capsaicin, and effects, before and after pre-treatment with capcaisin (10μM), are expressed as percent of 60 mM K^+-induced contractions.
For the cystometry investigations, the bladder was exposed via a midline incision of the lower abdomen, and a catheter (PE-50) was introduced through the bladder apex into the lumen, and held in place with a purse-string suture. The sealed catheter was tunneled s.c. and anchored in the neck. Three days after bladder catheterization, cystometry was performed by infusing saline (0.9 % NaCl) (10 ml/h) into the bladder to obtain baseline values. After this, rats were given AI (10 and 100 μM), CA (100, and 500 μM), or NaHS (1 and 3 mM). Protamine sulfate (PS; 10 mg/ml for 30 min) was used as a pretreatment to facilitate penetration of NaHS and CA. All substances were dissolved in saline. After each substance, bladder pressure and voided volumes were recorded for 30 min. Different animals (n=4-11/group) were used for each substance.

Results

AI, CA, or capsaicin produced concentration-dependent contractions of detrusor preparations. At the highest investigated concentration the contractions amounted to 22 ± 2 %, 17 ± 11 %, and 18 ± 2 % of K^+_{60}-induced contractions for AI (n=6), CA (n=6), or capsaicin (n=6) respectively. After pre-treatment with 10 μM of capsaicin, none of the compounds affected tonus of the detrusor preparations. NaHS caused a 4 ± 1 % (n=6) reduction in tension that was unaffected by pre-treatment with capsaicin.
Neither NaHS nor CA *per se* produced any significant changes in urodynamic parameters. After pre-treatment with PS, NaHS (1 mM) increased maximal bladder pressure (110 cmH_2O ± SD 49 vs. control: 78 ± 33, p=0.01), reduced voided volume (0.7 ml ± 0.2 vs. control: 1.1 ± 0.5 (0.03), and reduced voiding interval (4.0 sec ± 1.3 vs. control: 5.6 ± 2.1, p=0.049). Similarily, after pretreatment with PS, CA increased maximal bladder pressure (100μM: 97 cmH_2O ± SD 35, p=0.006; 500μM: 111 ± SD 42 vs. control: 82 ± 33, p=0.001), reduced voided volume (500μM: 0.7 ml ± 0.2 vs. control: 1.1 ± 0.2, p=0.003), and reduced voiding interval (500μM: 4 sec ± 0.8 vs. control: 5 ± 1.5, p=0.02). PS treatment by itself produced no urodynamic changes.
AI induced an increase in maximal bladder pressure (10 μM: 188 cmH_2O ± SD 67, p=0.03; 100 μM: 200 ± 31, p=0.007 vs. control: 122 ± 38), reduced voided volume (0.4 ml ± 0.1, p=0.03; 0.1 ± 0.04, p=0.003, respectively vs. control: 0.8 ± 0.2), and reduced voiding interval (2 sec ± 0.7, p=0.03; 0.8 ± 0.2, p=0.007 vs. control: 4 ± 1.2). In addition, the basal bladder pressure was increased after 100 μM. See figure for details.

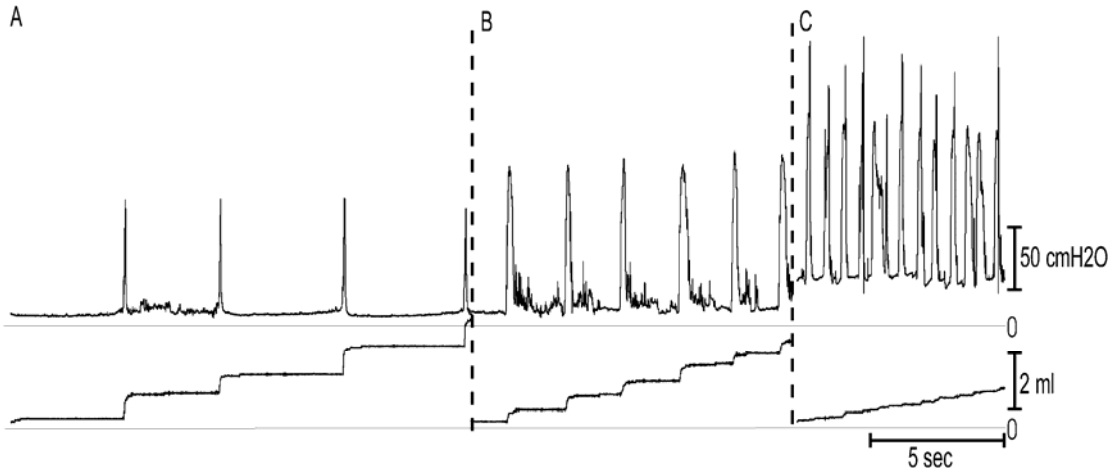

Figure. Cystometrogram from one rat showing bladder pressure (upper line) and voided volume (lower line) before (A) and after intravesical infusion of allyl isothiocyanate at 10 μM (B), and 100 μM (C).

Interpretation of results

The capsaicin-sensitive contractile effects by TRPA1-receptor agonists on isolated detrusor may reflect the ability of these agents to excite sensory neurons to release smooth muscle active peptides. In awake rats, intravesical administration of TRPA1 receptor agonists significantly increased micturition frequency at a constant rate of filling which is consistent with effects on bladder afferents. Increased micturition pressures may reflect postjunctional effects by release of peptides from sensory nerves secondarily to activation by TRPA1-receptor agonists, direct effects by the agents on the detrusor or urothelial region, or by effects of these agents on regulatory functions of the bladder outlet.

Concluding message

The current results suggest that functional TRPA1 receptors are present in the bladder and that activation of TRPA1-receptors can induce detrusor overactivity, probably by activation of capsaicin-sensitive afferent nerves.

FUNDING: NONE
DISCLOSURES: NONE
ANIMAL SUBJECTS: This study followed the guidelines for care and use of laboratory animals and was approved by Animal Ethics Committee, Court of the City of Lund, Sweden