UPREGULATIONS OF TRANSIENT RECEPTOR POTENTIAL (TRP) A1 AND V1 CHANNELS IN THE BLADDER AND THEIR FUNCTIONAL IMPLICATION IN MALE RATS WITH SUB-ACUTE PARTIAL BLADDER OUTLET OBSTRUCTION (BOO)

Hypothesis / aims of study
It has been reported that several TRP channels were expressed in the bladder urothelial cells and afferent nerves innervating the bladder, and that the expression of TRPA1 was up-regulated in the bladder mucosa taken from men with lower urinary tract symptoms/bladder outlet obstruction (male LUTS/BOO) (1). In addition, animal studies demonstrated that protein expressions of the TRPV1 and TRPV4 channels were increased in the bladder of the male rat with partial BOO (2, 3). However, possible expression changes of the other TRP channels in the bladder and its afferent pathways associated with BOO and their functional implication have not been fully investigated. In this study, we first evaluated the mRNA expressions of TRP channels systematically in the urinary bladder, L6 dorsal root ganglion (DRG), and spinal cord dorsal horn of rats with partial BOO at a sub-acute period. Second, we investigated the effects of antagonists for some TRP channels of which mRNA expressions were up-regulated on bladder function in this BOO rat model.

Study design, materials and methods
Forty-seven male Wistar rats were used and divided into Sham and BOO groups. To make partial BOO, the proximal urethra was ligated with a steel rod (1.2 mm in diameter) and then the steel rod was removed. Sham rats received similar surgery without urethral ligation. Ten days after the surgery, the urinary bladder (whole, mucosal, and detrusor layers), L6 DRG, and spinal cord dorsal horn were harvested. The mRNA expressions of TRP channels (TRP A1, C1, C2, C3, C4, C5, C6, C7, M2, M4, M8, V1, V2, and V4) were analyzed by RT-PCR. By focusing on the TRP channels of which mRNA was upregulated in the RT-PCR analysis, we further investigated the effect of antagonists for these up-regulated TRP channels on bladder function in the BOO rats in cystometry (CMG) measurements in case of such antagonists were commercially available. A catheter was placed into the bladder at 8 days following the surgery, and conscious and restraint CMG measurements were performed 2 days after the catheter-implantation. CMG was repeated 3 times and the third measurement served as the baseline, and then the measurements were further repeated after cumulative intravenous administrations of the antagonists or its vehicle.

Results
Compared with the Sham group, the BOO group showed significantly higher expressions of TRPA1 in the whole and mucosal layers of the bladder, TRPV1 in all the three types of layers, and TRPV2 in the whole and detrusor layers (Figure 1), whereas the expressions of the other TRP channels in any of the specimens investigated were not significantly different between the two groups (data not shown). Taking these results of RT-PCR, we further investigated the effects of HC-030031 (HC), a TRPA1 antagonist, and SB-366791 (SB), a TRPV1 antagonist, on bladder function in CMG measurements, whereas we could not perform similar CMG investigations with a TRPV2 antagonist as no TRPV2 antagonist was commercially available. As a result, HC increased bladder capacity in a dose-dependent manner in the BOO rats, and the effect was significant only at the highest dose when compared with vehicle- and before-administrations (Figure 2). On the other hand, SB did not change any CMG parameters compared with before-administration (Figure 3).

Interpretation of results
The present BOO rats showed the up-regulations of TRPA1, TRPV1 and TRPV2 channels in the bladder. The results of the up-regulated TRPA1 and TRPV1 channels were consistent with previous reports (1, 3), but the present result demonstrated for the first time that TRPV2 channel was also up-regulated following BOO. In this study, we further investigated the effect of TRPA1 and TRPV1 antagonists on CMG parameters in this BOO rats. The TRPA1 antagonist increased the bladder capacity, which may partly associate with the result of the up-regulated mRNA expression in the bladder mucosal layer, suggesting involvement of TRPA1 channel in modulation of the bladder mechano-sensory transduction in BOO rats. On the other hand, the role of the TRPV1 channel in the development of bladder dysfunction associated with BOO is doubtful as the TRPV1 antagonist did not affect any CMG parameters in the BOO rats.

Concluding message
The present study clearly demonstrated that mRNA expressions of TRPA1, TRPV1 and TRPV2 channels in the bladder were up-regulated, and that the inhibition of the TRPA1 channel increased bladder capacity in male rats with subacute BOO.

References

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Figure 1: Real-time PCR analyses of TRP channels mRNA expression in whole, mucosal, and detrusor layers of the bladder, and in DRG, and in spinal cord dorsal horn in Sham and BOO groups. *p<0.05, **p<0.01: significant differences from the Sham rats (unpaired t-test).

Figure 2: The effect of HC-030031 on cystometric parameters in the BOO rats. Values are expressed as mean ± SEM. *p<0.05: significant difference from before administration (one-way ANOVA Dunnett’s test), #p<0.05: significant difference from vehicle administration (unpaired t-test).

Figure 3: The effect of SB-366791 on cystometric parameters in the BOO rats. Values are expressed as mean ± SEM. No significant differences were found before and after SB administrations (one-way ANOVA Dunnett’s test).

Disclosures
Funding: None  Clinical Trial: No  Subjects: ANIMAL Species: Rat  Ethics Committee: Animal Ethics Committee, The University of Tokyo Graduate School of Medicine