543

Imamura T[1], Ishizuka O[2], Sudha G[1], Lei Z[2], Noguchi W[2], Yamagishi T[2], Yokoyama H[2], Kurizaki Y[2], Nishizawa O[2]
1. *Department of Lower Urinary Tract Medicine, Shinshu University School of Medicine,* ***2.*** *Department of Urology, Shinshu University School of Medicine*

AN EXTRACT OF BA-WEI-DIE-HUANG-WAN (THC-002) INCREASES SKIN TEMPERATURE AND DECREASES TRPM8 EXPRESSION LEVEL, AND INHIBITS COLD-STRESS INDUCED DETRUSOR OVERACTIVITY IN CONSCIOUS RATS

Hypothesis / aims of study
We showed that sudden whole body cooling with exposure of low temperature, cold-stress induced detrusor overactivity of conscious health rats, and a portion of the response was mediated by C-fiber sensitive neurological pathway [1], and transient receptor potential channel M8 (TRPM8) expressing on the skin [2]. In addition, we showed that Ba-Wei-Die-Huang-Wan Extract (THC-002) could partially inhibit detrusor overactivity induced with C-fiber sensitive neurological pathway [3]. In this study, we determined if THC-002 could increase skin temperature, and decrease expression level of TRPM8 mRNA, and then inhibit detrusor overactivity induced by cold-stress in rats.

Study design, materials and methods
Twenty-four female 10-weeks Sprague-Dawley (SD) rats were randomly separated into two groups (n=12 in each). Legs of the rats were shaved, and then the skin temperature was measured with a thermal imaging camera. One group was orally given 100 mg / 5 ml per kg-body weight THC-002 daily for one week (THC-002-treatment group). Another was similarly given 5 ml per kg-body weight THC-002-free saline (control group). At 7 days after, cystometric investigations of the conscious and free-moving rats were performed at room temperature (RT, 27±2°C) for 20 min, and at low temperature (LT, 4±2°C) for 40 min. The LT exposure was equally divided into two phases (Phase I and II). During the cystometric investigations, the micturition parameters were recorded, and the skin temperature was simultaneously measured at RT and LT 5, 10, 20, 30, and 40 min. After the cystometric investigations, TRPM8 mRNA expression level on the leg skin was estimated.

Results
At 7 days after administration, the skin temperature of THC-002-treated rats significantly increased (from 35.56±0.21℃ to 37.95±0.75℃) compared to the control rats (from 35.74±0.18℃ to 35.70±0.36℃, $P<0.05$). Exposure of LT affected skin temperature and micturition patterns of the rats (Figure 1). During the first 20 min after transferring to LT (phase I), the skin temperature of the both groups significantly decreased (Figure 2A). Voiding interval (from 4.85±0.24 min to 3.16±0.27 min) and bladder capacity (from 0.84±0.04 ml to 0.55±0.04 ml) of the THC-002-treated rats did not decrease compared to the control rats (from 4.46±0.35 min to 2.36±0.28 min, and from 0.93±0.08 ml to 0.45±0.05 ml, respectively, Figure 2B and C). During the second 20 min of LT (phase II), the skin temperature of the both groups did not alter (Figure 2A), and the voiding interval and bladder capacity of the both group increased (Figure 2B and C). TRPM8 mRNA expression level of the THC-002-treated rats (0.89±0.15) was lower than that of the control rats (1.37±0.21, $P<0.01$).

Interpretation of results
During the first 20 min after transferring to LT, the skin temperature of control rats was significantly decreased, and the rats exhibited decreases of voiding interval and bladder capacity. During the second 20 min, the skin temperature of the control rats did not decrease, and their voiding interval and bladder capacity were longer and larger. These results suggested that the decrease of skin temperature might be related with cold-stress induced detrusor overactivity. In contrast, while the skin temperature of THC-002-treated rats was also decreased by exposure of LT, the voiding interval and bladder capacity of THC-002-treated rats did not decrease compared to the control rats. Also, we showed that THC-002 increased skin temperature, and decreased TRPM8 mRNA expression level. Therefore, THC-002 can improve physical condition for the cold-stress tolerance through not only inactivity of C-fiber sensitive neurological pathway, but also increase of the skin temperature, and decrease of the expression level of TRPM8.

Concluding message
The skin temperature of THC-002-treated rats significantly increased. The expression level of TRPM8 mRNA on the THC-002-treated rat skin significantly decreased. The cold-stress induced detrusor overactivity in the THC-002-treated rats was partially inhibited.

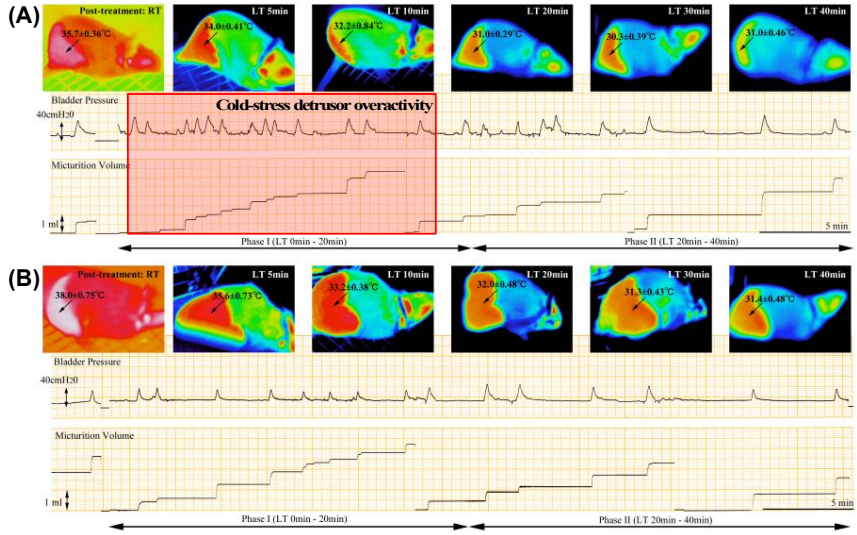


Figure 1. Skin temperature and micturition patterns under LT condition.
(A) THC-002-free control rats, (B) THC-002-treated rats

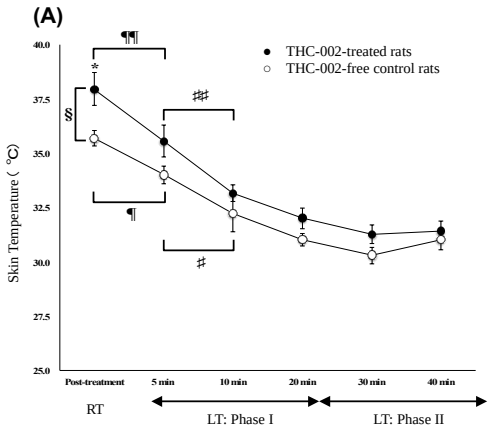
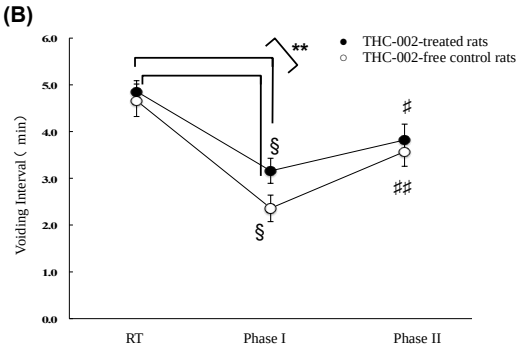
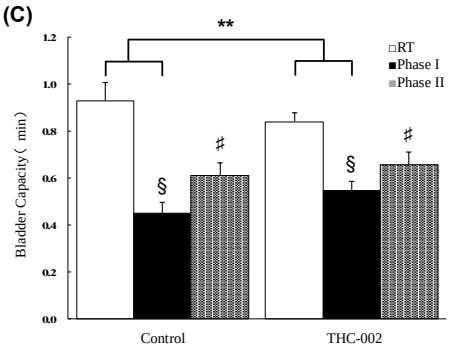


Figure 2. (A) Measurement of leg skin temperature in each condition. *P<0.05, compared to pre-treatment RT in each group, §P<0.05, compared to control at post-treatment RT, ¶P<0.05, ¶¶P<0.01, compared to RT in each group, and ♯P<0.05, ♯♯P<0.01, compared to LT 5min in each group. (B) voiding interval and (C) bladder capacity in each condition. **P<0.01, §P<0.01, and ♯P<0.05, ♯♯P<0.01.





References

1. Imamura, et al. Neurourol Urodyn 2008;27:348-52
2. Chen, et al. Neurourol Urodyn 2010;29:506-11
3. Imamura, et al. Neurourol Urodyn 2009;28:529-34

Disclosures

Funding: none **Clinical Trial:** No **Subjects:** ANIMAL **Species:** Rat **Ethics Committee:** The Animal Ethics Committee of Shinshu University School of Medicine (No. 190059).