374

Suzuki T[1], Shimizu T[2], Kwon J[2], Wada N[2], Takai S[2], Shimizu N[2], Takaoka E[2], Miyake H[3], Ozono S[3], Yoshimura N[2]
1. *University of Pittsburgh, Hamamatsu University School of Medicine,* ***2.*** *University of Pittsburgh,* ***3.*** *Hamamatsu University School of Medicine*

ROLE OF THE SEROTONERGIC SYSTEM IN URETHRAL CONTINENCE REFLEXES DURING SNEEZING IN RATS

Hypothesis / aims of study

The spinal serotonergic pathways are reportedly involved in the control of urethral continence reflexes that prevent stress urinary incontinence (SUI). Previous studies described that serotonin (5-HT) receptor subtypes, 5-HT_{1A} and 5-HT_{2C}, respectively reduce and enhance the urethral continence reflex during sneezing in rats [1]. However, because there are other multiple excitatory and inhibitory 5-HT receptors, the overall effects of the 5-HT system on the urethral function remain to be elucidated. Therefore, in this study, we examined the effects of 5-HT depletion induced by p-chlorophenylalanine (PCPA) that inhibits 5-HT synthesis and 5-HT_{2C} or 5-HT_7 subtype agonists on urethral baseline activity and reflex contractions during sneezing in rats.

Study design, materials and methods

We investigated the effects of intraperitoneal application of PCPA (200 mg/kg/day), and intravenous application of a 5-HT_{2C} agonist (CP-809101) or a 5-HT_7 agonist (LP44) on neurally evoked urethral continence reflexes during sneezing using female rats. Female Sprague-Dawley rats (12 weeks old) were divided into two groups; either Normal group (n = 6) or PCPA-treatment group without (n = 5) or with 5-HT drug administration (n=21). The PCPA-treatment + drug administration group received intravenous injection of; (1) a 5-HT_{2C} agonist without (n = 4) or with a 5-HT_{2C} antagonist (n = 4) or (2) a 5-HT_7 agonist without (n = 7) or with a 5-HT_7 antagonist (n = 6). In the PCPA-treatment + 5-HT_7 agonist group, a 5-HT_{1A} antagonist (WAY-100635) was also administered before LP44 administration to suppress the partial 5-HT_{1A} agonistic effect of LP44. Amplitudes of urethral pressure responses during sneezing (A-URS) and urethral baseline pressure (UBP) were measured before and after drug administration under urethane anesthesia by using a microtransducer-tipped catheter inserted to at the middle urethra. UBP was determined from a plateau section of pressure recordings just before inducing sneezing, which was induced by intranasal stimulation with a whisker. A-URS values were measured as the maximal urethral pressure change from baseline during sneezing. To evaluate induced sneeze intensity, abdominal pressure during sneezing (Pabd) was also measured via an intraabdominal balloon catheter. All data are shown in cmH_2O. Student's t-test or one way analysis of variance followed by Bonferroni's multiple comparison tests were used to compare before and after two or three kinds of drugs administrations. P values 0.05 were considered to be significant.

Results

5-HT depletion by PCPA treatment significantly decreased A-URS from 71.8 ± 7.1 to 36.7 ± 4.3 cmH_2O ($p < 0.01$), and also UBP from 31.1 ± 3.0 to 17.8 ± 2.2 cmH_2O ($p < 0.01$) compared to normal rats (Fig. 1). On the other hand, in PCPA-treated rats, CP-809101 alone (Fig. 2) or LP44 with WAY-100635 (Fig. 3) significantly increased A-URS from 42.1 ± 5.7 to 66.2 ± 6.5 cmH_2O ($p < 0.01$) or from 30.0 ± 2.7 to 50.5 ± 5.3 cmH_2O ($p < 0.01$) as well as UBP from 17.3 ± 1.2 to 32.4 ± 2.7 cmH_2O ($p < 0.05$) or from 15.2 ± 1.6 to 26.6 ± 2.1 cmH_2O ($p < 0.01$), respectively. The enhancing effects of 5-HT_{2C} or 5-HT_7 agonist on A-URS and UBP were antagonized by respective 5-HT receptor antagonist.

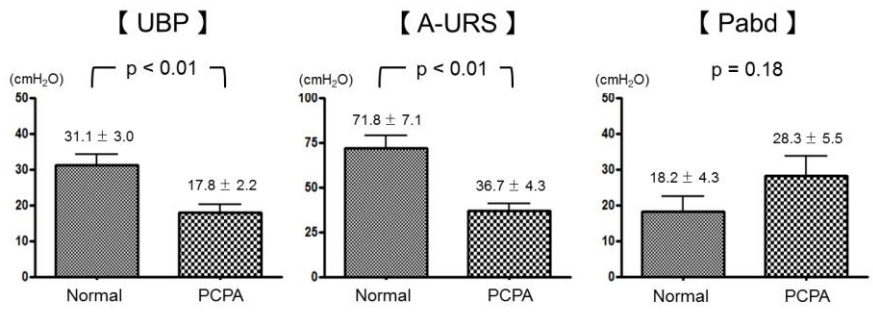
Interpretation of results

The site of the action of 5-HT in the lumbosacral spinal cord is found in the Onuf's nucleus, where dense 5-HT-containing nerve terminals onto urethral rhabdosphincter motoneurons are identified [2]. In the present study, 5-HT depletion by PCPA decreased A-URS and UBP, indicating that the overall 5-HT system plays a facilitatory role in the urethral continence function. Furthermore, both 5-HT_{2C} and 5-HT_7 agonists increased A-URS and UBP, indicating that not only 5-HT_{2C}, but also 5-HT_7 receptor subtypes can enhance the urethral continence reflex. In addition, previous studies reported that intrathecal application of a 5-HT_{2C} agonist, mCPP, increased A-URS without affecting UBP in rats without 5-HT depletion [1], suggesting that endogenous 5-HT may influence the 5-HT_{2C} receptor-mediate effect on UBP.

Concluding message

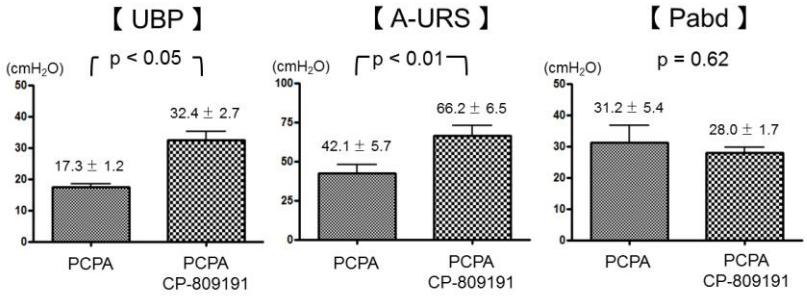
These results indicate that activation of 5-HT receptors such as 5-HT_{2C} and 5-HT_7 enhances the active urethral closure reflex during sneezing. Therefore, activation of these excitatory 5-HT receptor subtypes could be effective for the treatment of SUI.

Fig. 1



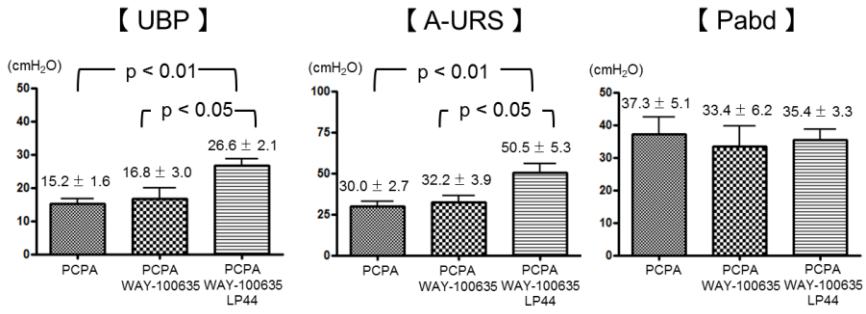
Mean ± SEM (cmH_2O); Unpaired t-test

Fig. 2



Mean ± SEM (cmH_2O); Paired t-test

Fig. 3



Mean ± SEM (cmH_2O); One-way analysis of variance followed by Bonferroni's multiple comparison tests

References
1. Am J Physiol Renal Physiol 295: F1024–F1031, 2009
2. J Comp Neurol 318: 1–17, 1992

Disclosures
Funding: NIH R01DK107450 **Clinical Trial:** No **Subjects:** ANIMAL **Species:** Rat **Ethics Committee:** University of Pittsburgh Institutional Animal Care and Use Committee