

## ROLE OF $\beta_3$ -ADRENOCEPTORS IN THE CONTROL OF THE URETHRAL CONTINENCE FUNCTION IN FEMALE RATS

### Hypothesis / aims of study

Mirabegron, a  $\beta_3$ -adrenoceptor agonist, has been used for the treatment of overactive bladder (OAB) because of its major action inducing bladder smooth muscle relaxation. However, the role of  $\beta_3$ -adrenoceptors or action of mirabegron in the control of urethral continence function is not well elucidated although the existence of  $\beta_3$ -adrenoceptors in the human external urethral sphincter has been shown previously [1]. Also, a recent study demonstrated that mirabegron induce smooth muscle relaxation of rat urethral strips by blocking  $\alpha_1$ -adrenoceptors, rather than an agonistic action on  $\beta_3$ -adrenoceptors [2]. Therefore, this study tried to investigate the effect of mirabegron and selective  $\beta$ -adrenoceptor antagonists on the urethral contractile function using female rats.

### Study design, materials and methods

Twenty female Sprague-Dawley rats were used and divided into 3 groups. In group A, 2 mg/kg mirabegron was administered intravenously (IV) in 5 rats and 10 mg/kg mirabegron in other 5 rats. In group B, 50  $\mu$ g/kg L-748,337, a selective  $\beta_3$ -adrenoceptor antagonist, was injected IV prior to 10mg/kg mirabegron in 5 rats. In group C, 3 mg/kg propranolol, a non-selective  $\beta$ -adrenoceptor antagonist, and 50  $\mu$ g/kg L-748,337 was injected IV to inhibit all 3  $\beta$ -adrenoceptor subtypes; and then 10 mg/kg mirabegron was injected in 5 rats.

Before drug administration, the T8-9 spinal cord was transected to block the spino-bulbo-spinal voiding reflex under isoflurane anesthesia. Thereafter, a PE60 catheter and a microtransducer-tipped catheter were inserted into the bladder through the dome and into the mid urethra from the external urethral orifice, respectively. Then, LPP (leak point pressure), UBP (urethral baseline pressure), and dUP (differential values of urethral pressure during intravesical pressure elevation) were measured before and after drug administration under urethane anesthesia. LPP was defined as the intravesical pressure at which the fluid leakage occurred by increasing the intravesical pressure gradually using a water reservoir connected to the bladder. UBP was measured as the urethral pressure maintained stable at 0 cmH<sub>2</sub>O intravesical pressure for more than 2 minutes. dUP was measured in the mid urethra at 10-14 mm from the urethral meatus and defined as the greatest urethral pressure differential value just prior to the fluid leakage when the intravesical pressure was gradually elevated (Fig. 1). For statistical analysis, Wilcoxon's signed rank test was used to compare the changes in parameters within the same group, and Mann-Whitney U test among the different groups.

### Results

In group A, LPP, UBP, and dUP were not changed after 2 mg/kg mirabegron, but showed significant decreases in these 3 parameters from  $45.0 \pm 2.1$  to  $29.5 \pm 2.3$  cmH<sub>2</sub>O, from  $23.9 \pm 1.1$  to  $14.0 \pm 1.8$  cmH<sub>2</sub>O and from  $21.2 \pm 0.9$  to  $10.3 \pm 0.7$  cmH<sub>2</sub>O, respectively, after 10 mg/kg mirabegron (Fig. 2). In group B, after administration of L-748,337 ( $\beta_3$  antagonist), there were no changes in LPP or dUP whereas UBP was slightly, but significantly, increased from  $20.3 \pm 2.1$  to  $23.0 \pm 1.7$  cmH<sub>2</sub>O. 10 mg mirabegron following L-748,337 still significantly reduced LPP from  $41.5 \pm 2.9$  to  $26.5 \pm 1.3$  cmH<sub>2</sub>O, UBP from  $23.0 \pm 1.7$  to  $12.9 \pm 1.6$  cmH<sub>2</sub>O, and dUP from  $19.9 \pm 0.8$  to  $10.9 \pm 0.7$  cmH<sub>2</sub>O compared to L-748,337 alone (Fig. 3). In group C, any of 3 parameters were not changed by the combined administration of propranolol and L-748,337. However 10 mg/kg mirabegron following propranolol and L-748,337 still reduced LPP from  $43.5 \pm 1.3$  to  $29.0 \pm 1.3$  cmH<sub>2</sub>O, UBP from  $24.8 \pm 1.6$  to  $16.3 \pm 1.2$  cmH<sub>2</sub>O, and dUP from  $21.2 \pm 1.4$  to  $11.0 \pm 1.2$  cmH<sub>2</sub>O (Fig. 4).

### Interpretation of results

Mirabegron did not affect urethral pressure within the clinically approved dose (2mg/kg) in rats, but, at a higher dose, it reduced the baseline urethral activity and reflex urethral contractions during passive bladder pressure elevation. Because this action of high-dose mirabegron was not interrupted by selective  $\beta_3$ -adrenoceptor antagonist or non-selective  $\beta$ -receptor antagonist, suggesting that mirabegron can exert its action via other receptors than  $\beta$ -adrenoceptors, possibly including blockade of  $\alpha_1$ -adrenoceptors [2]. In addition, the selective  $\beta_3$  adrenoceptor antagonist showed a slight, but significant increase in urethral baseline tone, suggesting that  $\beta_3$ -receptor activation is tonically active to reduce baseline urethral activity although this mechanism does not seem to be involved in mirabegron-induced effects on urethral function.

### Concluding message

$\beta_3$ -adrenoceptors play a minor role in the control of urethral baseline tone and reflex contractions of the urethral sphincter muscles. Mirabegron can induce urethral sphincter relaxation through  $\beta_3$ -receptor-independent mechanisms at a high dose, possibly through interactions with other receptor types such as  $\alpha_1$ -adrenoceptors.

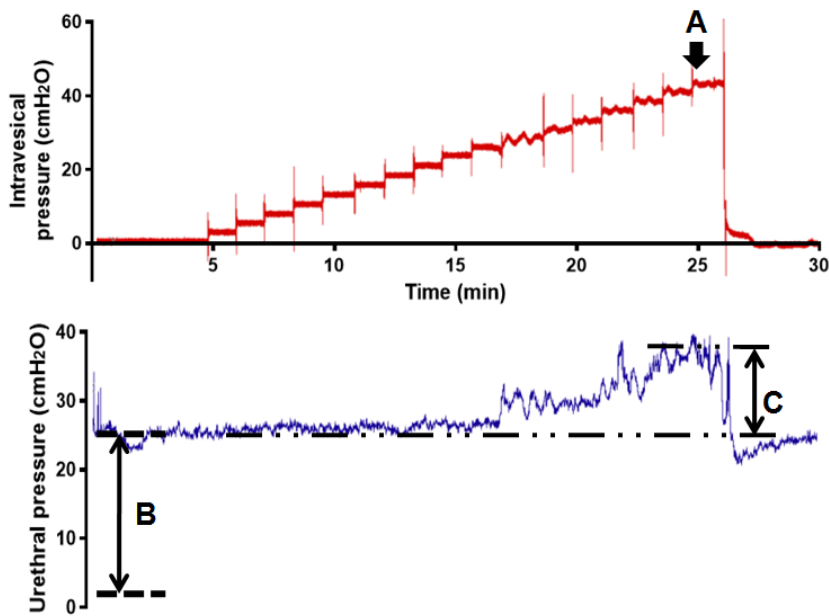


Fig. 1. Representative recordings of LPP (A; leak point pressure), UBP (B; urethral baseline pressure), dUP (C; differential values of urethral pressure during intravesical pressure elevation).

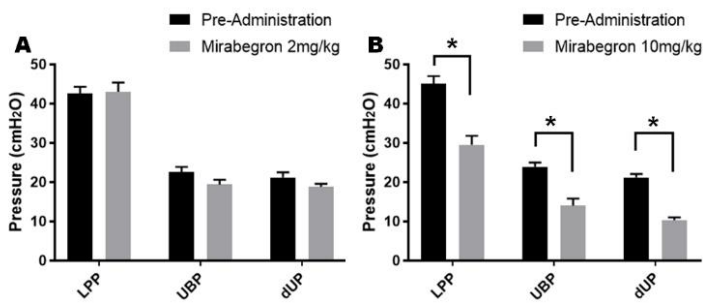


Fig. 2. Changes in LPP, UBP, and dUP responses to 2 mg/kg mirabegron (A) and 10 mg/kg mirabegron (B). \* p < 0.05 by Wilcoxon's signed rank test.

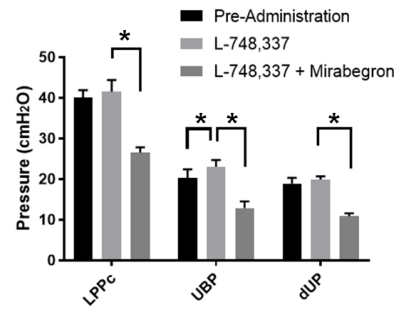


Fig. 3. Changes in LPP, UBP, and dUP responses to 50 µg/kg L-748,337 and following administration of 10 mg/kg mirabegron. \* p < 0.05 by Wilcoxon's signed rank test.

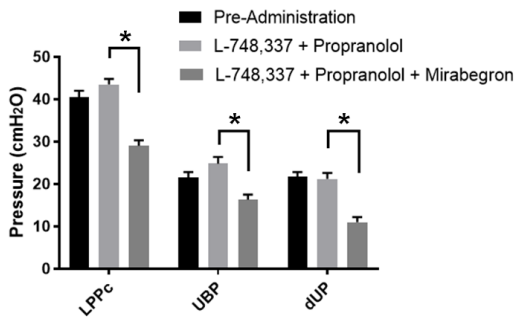


Fig. 4. Changes in LPP, UBP, and dUP responses to 50 µg/kg L-748,337 combined with 3 mg/kg propranolol and following administration of 10 mg/kg mirabegron. \* p < 0.05 by Wilcoxon's signed rank test.

## References

1. J Smooth Muscle Res. 2000;36:21-32.
2. Br J Pharmacol. 2016;173(3):415-28.

## Disclosures

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