Objectives

While it is well established that stimulation of β-adrenoceptors (β-ARs) produces urinary bladder smooth muscle relaxation, very little is known about how β-AR stimulation affects neurally mediated bladder contractions. Relaxation of basal tension and pre-contracted urinary bladder strips has been shown to be mediated by both β₂- and β₃-ARs in rat (1) and pig (2) and by β₂-AR in dog (3), primates (4, 5) and human (6, 7). Recently, the inhibition of EFS-induced contractions of human urinary bladder by stimulation of β₂-ARs has been demonstrated (8).

The purpose of the present study was to characterize the β-AR subtypes involved in the inhibition of neurogenic contractions of mouse urinary bladder.

Methods

Urinary bladders were obtained from adult female C57Bl/6 mice (aged 11-13 weeks) sacrificed by cervical dislocation.
Bladders were bisected and bladder halves mounted into 5 mL organ baths under 0.5 g of initial tension in the presence of prazosin (1 μM) in order to block α₁-ARs.
ICI118,551 (β₂-AR antagonist at 30 nM), L748,337 (β₃-AR antagonist at 3 or 10 μM) or vehicle (0.001% DMSO in distilled water) were added to the organ bath.
15 min later, tissues were subjected to EFS using the following parameters: maximal current, frequency of 2.5 Hz, pulse duration 0.3 ms, trains of pulses 2 s every minute.

Once responses to EFS stabilized, cumulative concentration-response curves to β-AR agonists (fenoterol, a β₂-AR selective agonist; CL316,243, a β₃-AR selective agonist; isoproterenol, a non-selective β-AR agonist) were constructed.

Results

Involvement of both β₂- and β₃-adrenoceptors in the inhibition of neurogenic contractions of mouse isolated urinary bladder

Effects of the selective β₂-AR agonist CL316,243 on EFS-induced contractions of mouse urinary bladder strips

- CL316,243 concentration-dependently inhibited EFS-induced contractions of mouse urinary bladder strips with a pEC₅₀ value of 7.48 and an E₅₀ value of 36 ± 5%.
- L748,337 (but not ICI118,551) significantly inhibited the effects of CL316,243 without affecting the maximal response with a pA₂ value of 7.00, that is similar to the affinity value (pKᵢ = 6.5) reported at rat recombinant β₂-ARs (7).

Effects of β-AR antagonists on fenoterol and isoproterenol-mediated inhibition of EFS-induced mouse urinary bladder strip contractions

- Fenoterol and isoproterenol inhibited EFS-induced contractions of mouse urinary bladder strips in a concentration-dependent manner with a pEC₅₀ and E₅₀ values of 6.85 and 68 ± 2% and 6.96 and 65 ± 4%, respectively.
- ICI118,551 potently blocked the relaxing effects of fenoterol (pA₂ = 8.80) and isoproterenol (pA₂ = 8.53), values that are very similar to the pA₂ value (8.97) reported in rat isolated myometrium, a tissue known to express β₂-ARs (8).
- In contrast, L748,337 inhibited the fenoterol and isoproterenol effects only at relatively high concentrations (pA₂ = 5.79 and 5.49, respectively).

Comparison of inhibitory effects of β-AR agonists on EFS-induced mouse urinary bladder strip contractions

Both fenoterol and isoproterenol were able to inhibit EFS-induced contractions of mouse urinary bladder strips with similar potency and maximal inhibition of approximately 65%.
CL316,243 had similar potency, but only produced a maximal inhibition of 36%.

Conclusions

The current results demonstrate that stimulation of both β₂- and β₃-ARs produces inhibition of neurally-mediated contractions of mouse urinary bladder. CL316,243 acts exclusively through stimulation of β₂-ARs, whereas fenoterol and isoproterenol appear to activate mainly β₂-ARs. In addition, the stimulation of β₂-ARs produces a greater degree of inhibition than stimulation of β₃-ARs, suggesting a predominant role of β₂-ARs. Further studies are required to define whether the effects of β-AR stimulation on EFS-induced contractions are solely due to a post-junctional action or whether pre-junctional receptors are also involved.

References

(6) Rekik et al, Poster n° Tue 204, poster session FC08 of this meeting.