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Palea S[1], Deba A[1], Lluel P[1]
***1.*** *UROsphere*

# ACTIVATION OF ß3-ADRENOCEPTORS INHIBITS NEUROGENIC CONTRACTION IN MOUSE ISOLATED URINARY BLADDER.

Hypothesis / aims of study

Since $\beta_3$-adrenoceptors ($\beta_3$-ARs) have been reported to predominantly mediate the relaxation of the human detrusor muscle (1), they represent an interesting target for the treatment of overactive bladder.

In previous studies we demonstrated that a selective $\beta_3$-AR agonist (CL316,243) increased bladder capacity in anesthetized mice and relaxed the basal tension and spontaneous motility of the mouse isolated detrusor muscle (2).

The aim of this study was to investigate the effects of CL316,243 and SR 59230A (a selective $\beta_3$-AR antagonist) on contractions induced by electrical field stimulation (EFS) in mouse isolated urinary bladder.

Study design, materials and methods

Two detrusor muscle strips were obtained from each animal and mounted in organ bath filled with a Krebs solution containing propranolol (1 µM) and prazosin (1 µM) in order to block $\beta_1/\beta_2$-ARs and $\alpha_1$-adrenoceptors, respectively. An initial tension of 0.5 g was applied and strips were equilibrated for a 60 min period. Then, detrusor muscle strips were exposed to 80 mM KCl and a further 45 min re-equilibration period was applied.

Concentration-response curves (CRC) to CL316,243 (0.01 to 100 µM) or its solvent (Krebs solution) were obtained in detrusor muscle strips submitted to EFS (800 mA, pulse duration 0.3 ms, train of pulses 2 s every 60 s). CRCs were constructed in the presence of SR 59230A (0.3, 1, 3 µM) or its solvent (distilled water) incubated for 45 min.

CL316,243 effects were expressed as % inhibition (mean ± s.e.m) of basal response to EFS taken 5 min before the start of the CRC.

Results

EFS-induced contractions were totally abolished by tetrodotoxin 1 µM, confirming their neurogenic origin.

In a preliminary study we found that frequency-dependent detrusor contractions (range 0.625-20 Hz) were inhibited by 10 µM CL316,243 and the maximal effect was observed at 2.5 Hz (n=6, data not shown). This frequency was therefore used for further experiments. The vehicle for CL316,243 (n=14) slightly decreased EFS-induced contractions with a maximal effect of 9 ± 1 % (data not shown) whereas CL316,243 (n=13), in the range 0.01 – 100 µM, significantly and concentration-dependently inhibited these contractions (Figure 1). The maximal effect of CL316,243 was observed at 10 µM (-30 ± 2 %). By using a non-linear fitting, a $pIC_{50}$ value of 6.9 ± 0.1 was calculated for CL316,243. SR 59230A, tested at 0.3, 1 and 3 µM shifted to the right the CRC to CL316,243 in a concentration dependent manner without modification of the maximal effect (Figure 1). The $pA_2$ value was equal to 6.8 (Schild plot slope = 1.1 ± 0.1; Figure 1).

Interpretation of results

These data demonstrated that CL316,243 inhibited mouse detrusor muscle neurogenic contractions with the same potency found in the relaxation of basal tone in the same preparation (2). The $\beta_3$-AR antagonist, SR 59230A, antagonized the effect of CL316,243 with a potency comparable to that ($pA_2$ = 6.71) previously reported by us on the relaxation of basal tone (2). Moreover SR 59230A potency reported in the mouse urinary bladder is very similar to the potency ($pA_2$ = 7.01) displayed versus another $\beta_3$-ARs agonist on the relaxation of the basal tone in human detrusor muscle (3).

These results confirm the presence of functional $\beta_3$-ARs in mouse urinary bladder. Whether these receptors are located not only on the detrusor muscle but also on parasympathetic nerve terminals deserves further investigation.

Concluding message

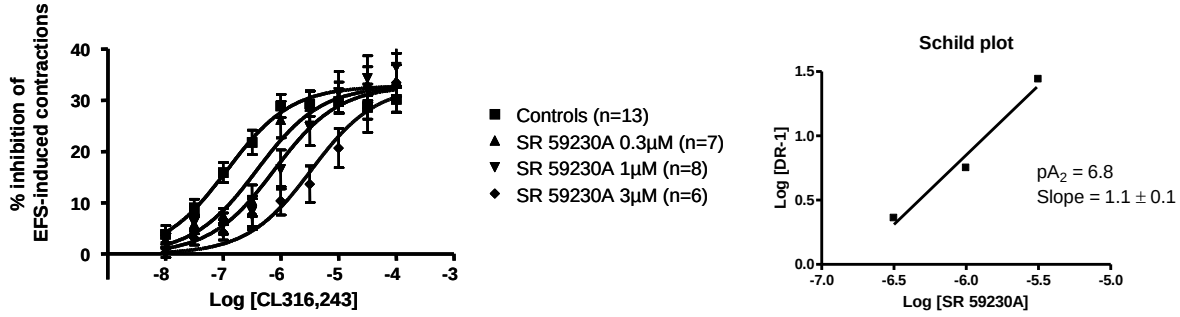
A selective $\beta_3$-AR agonist inhibits neurogenic contractions (present results) as well as basal tension and spontaneous motility in mouse isolated detrusor muscle (2), confirming that $\beta_3$-ARs agonist could be useful to treat overactive bladder in humans.

In addition, we noticed that SR 59230A displays similar antagonistic potencies on human and mouse urinary bladder, suggesting that the mouse can provide a predictive model for the selection of new $\beta_3$-AR agonists for human use.

References

1. J Pharmacol Exp Ther (1999) 288; 1367-1373.
2. Eur. Urol. (2008) 53 (Issue 3); Abstract n°448.
3. Neurourol Urodyn (2006) 25; 815-819.

Figure 1: Concentration-response curves for the effects of CL316,243 on neurogenic contractions in mouse isolated urinary bladder in the presence or absence of SR 59230A; Schild plot for SR 59230A.

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| ***Is this a clinical trial?*** | **No** |
| ***What were the subjects in the study?*** | **ANIMAL** |
| ***Were guidelines for care and use of laboratory animals followed or ethical committee approval obtained?*** | **Yes** |
| ***Name of ethics committee*** | **Comité Régional d'ethique Midi-Pyrénées (Toulouse, France)** |