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THE EFFECT OF B-ADRENOCEPTOR AGONISTS ON DETRUSOR CONTRACTION IN THE PRESENCE AND ABSENCE OF UROTHELIUM

Hypothesis / aims of study

It is known that stimulating muscarinic receptors in bladder urothelium causes the release of nitric oxide and an unidentified diffusible factor that inhibits contractions of the detrusor muscle. The relaxation of detrusor muscle via β-adrenoceptors is thought to contribute to urine storage during bladder filling. That is why β-adrenoceptor agonists are thought to be a potential treatment for detrusor overactivity. However, there is little information about the relationship between urothelium and β-adrenoceptors. Therefore we investigated whether the responses of pig bladder induced by β-adrenoceptor agonists are affected by the urothelium. Furthermore, we also studied whether adrenergic-evoked nitric oxide, which is reported to be released from urothelium, was affected by β-adrenoceptor agonists.

Study design, materials and methods

Fresh pig bladders were obtained from a local abattoir and immediately placed in (4 degrees Celsius) cold Krebs solution (composition in mM: NaCl 118.4, KCl 4.7, $CaCl_2$ 1.9, $NaHCO_3$ 24.9, $MgSO_4$ 1.15, KH_2PO_4 1.15, glucose 11.7). Paired longitudinal strips of pig bladder dome were isolated. The urothelium was carefully removed from 1 strip per pair before suspension in 5 ml organ bath. Tissues were bathed in Krebs solution, maintained at 37 degrees Celsius and gassed with 95% O_2-5% CO_2. Tissues were attached to force transducers to measure isometric tension and a tension of 1 g was applied. Tension developed by tissues was recorded to a Macintosh computer using “Chart” software. In the relaxation experiments, following the concentration-response curves to carbachol, the tissues were pre-contracted with carbachol. When the contraction had stabilized, increasing concentrations of β-adrenoceptor agonists were added cumulatively in 0.5 log unit increments and concentration-relaxation curves obtained to isoprenaline (non-selective β-adrenoceptor agonist) and BRL37344 (β3-adrenoceptor selective agonist). On the other hand, we also performed inhibition experiments. First, we constructed the carbachol-response curves and after washout then added isoprenaline before the second curves to carbachol were constructed. Furthermore, to study nitric oxide interactions, we added 100 μM L-NNA before obtaining responses to isoprenaline. Results are expressed as a mean ± SEM, with paired Student’s t-test used for statistical analysis.

Results

Contractile responses to carbachol were depressed by approximately 35% in the presence of the urothelium. In relaxation experiments, isoprenaline relaxed carbachol precontracted tissues by approximately 60%, the potency (pEC_{50}) and maximal relaxation were similar in the absence (pEC_{50}=7.38 ± 0.33, maximum=63.1 ± 5.0 %) and presence of the urothelium (pEC_{50}=7.19 ± 0.16, maximum=56.5 ± 6.0 %). BRL37344 also relaxed tissues equally in the absence and presence of urothelium. In inhibition experiments, pretreatment with isoprenaline (0.1 μM) caused parallel shifts to the right of the carbachol-induced response curves, but pretreatment with isoprenaline did not inhibit the maximal contractions. There was a significant difference in the pEC_{50} values for carbachol between the absence (5.46 ± 0.07) and presence (5.14 ± 0.09) of urothelium. There was no significant difference in the responses to isoprenaline in the presence of L-NNA compared to the control.

Interpretation of results

These data demonstrate that the urothelium does not influence detrusor relaxation to β-adrenoceptor agonists, but does have some influence on the inhibition of smooth muscle contraction by these agents. It is possible that β-adrenoceptor agonists cause the release of an unidentified diffusible inhibitory factor as described previously for carbachol. Isoprenaline has been reported to cause nitric oxide released from the urothelium, but this study suggests that nitric oxide does not have a direct effect on detrusor smooth muscle. Isoprenaline relaxed carbachol precontracted tissues well, but did not inhibit carbachol-induced maximal contraction. This can be interpreted as isoprenaline having an effect on the filling phase and less effect on the voiding phase. That is why isoprenaline has a good relaxant effect and a weak inhibitory effect in detrusor smooth muscle.

Concluding message

These data suggest β-adrenoceptor agonists stimulate the release of an unidentified diffusible factor from urothelium, which inhibits contractions of the detrusor muscle. Nitric Oxide also was not associated with the responses to β-adrenoceptor agonists in vitro. These findings suggest an interesting mechanism of action for this class of drugs potential a therapeutic role in the treatment of storage conditions such as overactive bladder. Further work in man is clearly essential before definitive conclusions can be drawn.

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