Muscarinic Receptors in the Urinary Bladder

The role of the urothelium regarding cholinergic and nitrergic effects in inflammation

Akademisk avhandling

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av

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I. **Andersson MC.**, G. Tobin & D. Giglio, 2007
Cholinergic nitric oxide release from the urinary bladder mucosa in cyclophosphamide-induced cystitis of the anaesthetized rat
Br J Pharm 2008 Apr;153(7):1438-44. Epub 2008 Feb 4

II. **Andersson M.**, P. Aronsson, D. Doufish, A. Lampert & G. Tobin
The muscarinic M5 receptor is the primary mediator of urothelium-derived nitric oxide effects in the rat urinary bladder
Manuscript

III. **Andersson M.**, P. Aronsson, D. Giglio, A. Wilhelmson, P. Jeřábek & G. Tobin, 2010
Pharmacological modulation of the micturition pattern in normal and cyclophosphamide-pretreated conscious rats
Auton Neurosci 2010. Epub 2010 Sep 17

IV. **Andersson M**., P. Aronsson, D. Eskandari, D. Giglio & G. Tobin
Characterization of receptor-mediated proliferation in the human bladder urothelial UROtsa and T24 cell line
Manuscript

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The role of the urothelium regarding cholinergic and nitrergic effects in inflammation

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Abstract

Inflammation alters the functional properties of the urinary bladder. Interstitial cystitis (IC) is a non-infectious chronic syndrome that is characterized by painful urination, urgency, frequency, inflammation, stiffening of the bladder wall and visceral pain. The aim of this study was to investigate how the physiological properties of the urinary bladder are affected by inflammation, and what specific part the urothelium plays in this.

Methods: Cystitis was induced in rats by a single injection of cyclophosphamide (CYP; 100 mg/kg). This treatment causes a disease state which is highly comparable to IC. Data comparing the properties of the healthy and inflamed bladder were gathered from (1) contraction experiments *in vitro* in an organ bath set-up, (2) cystometrical studies *in vivo* in anaesthetized rats and (3) wake, freely moving rats in a metabolism cage. Cell cultures were also cultivated in order to investigate if proliferation of urothelial cells is influenced by receptor activation.

Key findings: Induction of cystitis by CYP altered the cholinergic response of the urinary bladder. *In vitro* studies showed a significantly lower response to carbachol in the inflamed bladder. We could see that the altered cholinergic response could be normalized by either removal of the urothelium, blockade of nitric oxide synthase or blockade of muscarinic M1/M3/M5 receptors, both *in vitro* and *in vivo*. Further characterization *in vitro* revealed the M5 receptor to be the most likely candidate for mediating these effects. *In vivo* experiments carried out in the metabolism cage showed that micturition parameters are affected by CYP-induced cystitis. Increasing doses of a muscarinic antagonist eliminated these differences, and a connection between the effects of antimuscarinic and antinitrergic drugs was observed. These findings underline the importance of muscarinic receptors and NO in the alterations seen during cystitis. The proliferation experiments indicated that adrenergic, but not muscarinic, nicotinic or EGF receptors, are involved in the regulation of urothelial cell proliferation.

Conclusions: In CYP-induced cystitis in the rat, the urothelium exerts an inhibitory influence on the cholinergic response of the urinary bladder. We conclude that this is caused by the release of NO upon activation of urothelial muscarinic M5 receptors.

Keywords: urinary bladder, muscarinic receptor, urothelium, cyclophosphamide-induced cystitis, inflammation, nitric oxide, M5 receptor, proliferation, micturition, rat

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