223

Ost D[1], Blyweert W[1], Gevaert T[1], Roskams T[2], De Ridder D[1]
1. University Hospital Gasthuisberg, Dept of Urology, 2. University Hospital Gasthuisberg, Dept of Pathology

IMMUNOHISTOCHEMICAL EVIDENCE FOR A ROLE OF INTERSTITIAL CELLS IN THE ENDOCANNABINOID SYSTEM OF THE HUMAN URINARY BLADDER

Hypothesis / aims of study
Cannabinoids are known as mediators in peripheral and sensory neurotransmission (for review, see [2]). The main target for cannabinoids is the CB1 receptor. It has become clear that cannabinoids can act at prejunctional sites to modulate peripheral autonomic and sensory neurotransmission through the so-called endocannabinoid system. These endocannabinoids apparently exert also an activation of the TRPV-1 receptor, often co-expressed with CB-1. TRPV-1 is present on ICC.
The aim of this study was to study immunohistochemically the presence and relation between the endocannabinoid system (using antibodies for CB-1 (the target receptor for (endo)cannabinoids) and FAAH (the enzyme degrading endocannabinoids)) and interstitial cells (ICC) (using TRPV-1 and C-kit) in the human detrusor. PGP9.5 was used as pan-neuronal marker.

Study design, materials and methods
Appropriate ethical approval was obtained. 30 full thickness bladder specimens were obtained from 6 cystectomy specimens. Immunohistochemistry was performed using primary antibodies against CB-1, TRPV-1, C-kit, FAAH and PGP 9.5. For classic Immunohistochemical analysis 4µm slices were stained using a three step unlabeled peroxidase-anti-peroxidase method. Colocalisation studies were performed using confocal laserscanning microscopy in 20 µm slides. Controls consisted of omission of primary antibodies in subsequent slides.

Results
CB1 immunoreactivity was found on ICC in the suburothelium, lamina propria and some ICC in fibrovascular axes in the detrusor. There was a striking colocalisation with C-kit in all CB-1 immunoreactive cells, but not all C-kit immunoreactive cells were immunoreactive to CB-1. FAAH immunoreactivity was found on all CB1 immunoreactive ICC. CB-1 & FAAH immunoreactivity was also visible on the endothelium of small vascular structures. C-kit immunoreactivity appeared to be positive on ICC both in the suburothelium and the muscularis. Immunoreactivity to TRPV-1 and CB-1 was present in the suburothelium and in some (but clearly not all) ICC in the detrusor. TRPV-1 and C-kit positive ICC colocalised perfectly. No colocalisation was found between PGP9.5 and CB1 or FAAH, but nerve fibers were often in close contact with CB1 immunonegative ICC.

Interpretation of results
CB-1 and FAAH are localized in the human urinary bladder on ICC both in the suburothelium and the detrusor. The presence of CB1, TRPV-1 and FAAH provides evidence for the presence of an endocannabinoid system in the human urinary bladder involved in modulation of neurotransmission, as described in other organ systems[2]. The distribution of CB-1 immunoreactive ICC in the detrusor in close contact with CB-1 immunonegative ICC and neurons (PGP9.5 immunoreactive) supports the concept of cannabinoid signaling modulating neurotransmission[2].The perfect colocalisation of C-kit and CB-1 on ICC like cells implies that ICC are involved in this endocannabinoid system. The finding that not all ICC immunoreactive to C-kit and TRPV-1 are immunoreactive to CB-1 supports the newer concepts of several populations of ICC in the human detrusor[1].

Concluding message

The presence and distribution of CB-1 and FAAH immunoreactivity in the human bladder provides evidence for the presence of an endocannabinoid system as described in other organ systems. CB-1 and FAAH are apparently expressed by a different subpopulation of ICC, surrounding ICC in close contact to nerve endings. The detection of a subpopulation of ICC is in accordance with recent findings[1]. The endocannabinoid system in the bladder might become a new therapeutical target in modulating sensory and motoric neurotransmission of the human bladder.

Reference List

1. Davidson RA and McCloskey KD: Morphology and localization of interstitial cells in the guinea pig bladder: structural relationships with smooth muscle and neurons. J Urol **173**(4): 1385-1390, 2005.

2. Ralevic V: Cannabinoid modulation of peripheral autonomic and sensory neurotransmission. Eur J Pharmacol **472**(1-2): 1-21, 2003.