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UDP-SENSITIVE P2Y6 RECEPTORS PLAY A DUAL ROLE IN THE HUMAN URINARY BLADDER INDIRECTLY VIA THE RELEASE OF ATP FROM UROTHELIUM

Hypothesis / aims of study

Urothelium is the major source of purines and pyrimidines (e.g. ATP and UTP) in the urinary bladder. Bladder distension, inflammation and chemical irritation stimulate the release of nucleotides leading to bladder overactivity. ATP-sensitive P2X1 receptors prevail on the detrusor smooth muscle, whereas P2X3 receptors found on pelvic nerve afferent are implicated in micturition triggered by bladder filling. ATP stimulation of micturition may be partially counteracted by its catabolism into ADP by ecto-NTPDases at the urinary bladder; ADP acts through inhibitory P2Y1 receptors on cholinergic nerves. In contrast to the compelling evidence for the extracellular signalling role of ATP, the hypothesis that uridine nucleotides may also fulfil an autocrine/paracrine role has only recently gained experimental support. Metabotropic P2Y2 recognizes both ATP and UTP as the most potent agonists, while P2Y4 and P2Y6 receptors were recently identified and characterized as UTP- and UDP-selective receptors in humans, respectively. Preliminary data using the rat *in vivo* demonstrated that UDP (100 µM) and its analogue, PSB0474 (100 nM, a selective P2Y6 agonist), increase the voiding frequency through the release of ATP from the urothelium, without affecting the amplitude and the duration of bladder contractions (1).

Study design, materials and methods

Human bladder samples were collected from cadaveric organ donors. Urothelium and detrusor strips were mounted in organ baths and were superfused with oxygenated Tyrode's solution at 37°C. [³H]ACh outflow from strips loaded with [³H]choline was induced by electrical field stimulation (10 Hz, 200 pulses). ATP content of the samples used to measure the release of [³H]ACh was quantified using the luciferin-luciferase bioluminescence assay. Immunolocalization studies of P2 receptor and ecto-NTPDase subtypes were performed by immunofluorescence confocal microscopy (Olympus, FV1000).

Results

We showed that PSB0474 (100 nM) increased the release of ATP and [³H]-ACh induced by electrical stimulation of the human urothelium, an effect antagonized by the P2Y6 antagonist, MRS2578 (50 nM). The excitatory effect of UDP (100 µM) and PSB0474 (100 nM) in the bladder urothelium contrast with the inhibitory (~50%) responses observed on [³H]-ACh release from stimulated cholinergic nerves innervating the detrusor smooth muscle, both in the rat and in human samples; the P2Y1 antagonist, MRS2179 (0.3 µM), prevented the inhibitory effects of both UDP (100 µM) and PSB0474 (100 nM).

Interpretation of results

Our findings indicate that UDP, via P2Y6 receptors activation, operates a dual role in the human urinary bladder indirectly by releasing ATP from the urothelium. Data suggest that UDP-induced ATP release operate bladder excitability via P2X3 and P2X1 receptors located respectively on suburothelial sensory nerve fibres and smooth muscle fibres. However, ATP-mediated excitation may be partially counteracted by ADP formation through ecto-NTPDases 2 and 3 expressed in urothelial and suburothelial layers, as demonstrated by immunofluorescence confocal microscopy. The inhibitory effects of ADP are mediated by P2Y1 receptors, which activation restrains ACh release from stimulated cholinergic motoneurons thus contributing to indirectly reduce smooth muscle activity.

Concluding message

In this study, we gathered information regarding the role of uridine nucleotides, namely UDP via P2Y6 receptors, in the human urinary bladder. Our results confirmed that ATP released from the urothelium as a consequence of P2Y6 receptors activation plays a key role to control bladder activity. Nucleotide effects in the human bladder (as well as in many other tissues) occur through a series of complex events involving subtype specific P2 (and P1) purinoceptors, ecto-NTPDases and the local production of secondary metabolites (e.g. ADP/UDP). The (patho)physiological significance of these arrays, as well as their geographical distribution and kinetics are just beginning to be unravelled, which certainly will lead to novel therapeutic targets to control urinary bladder function and dysfunction.

References

1. (1) Timóteo M A, Carneiro I, Vieira C, Silva N, Marinhos J, Monteiro D, Ferreirinha F, Sévigny J, Silva-Ramos M, Correia-de-Sá P (2011) ATP release from the urothelium mediates bladder excitation triggered by udp-sensitive p2y6 receptors. Poster presented at ICS 2011, Glasgow, abstract n° 514 retrieved from <http://www.icsoffice.org/Abstracts/Publish/106/000514.pdf>.

Disclosures

Funding: This work was supported by FCT (FEDER funding, PTDC/SAU-OSM/104369/2008 and UMIB-215/94), Associação Portuguesa de Urologia and Univ. Porto / Caixa Geral de Depósitos. **Clinical Trial:** No **Subjects:** HUMAN **Ethics Committee:** Comissão de Ética do Centro Hospitalar do Porto and Comissão de Ética do Instituto de Ciências Biomédicas Abel Salazar **Helsinki:** Yes **Informed Consent:** Yes