Tadalafil attenuates hypotonicity-induced Ca2+ influx via TRPV2 and TRPV4 channels as well as ATP release in primary bladder urothelial cell cultures.

Introduction & Objectives

Phosphodiesterase 5 (PDE5) inhibitor, tadalafil, improves lower urinary tract symptoms suggestive of benign prostatic hyperplasia. The mechanism is believed to smooth muscle relaxation, increased blood perfusion and modulation of sensory stimuli via increased activity of the NO/cGMP/protein kinase G pathway (1). The bladder urothelium is presumed to be involved in sensory mechanisms activated in response to physical and chemical stimuli (2). Urothelial cells express several receptors/ion channels including transient receptor potential (TRP) channels and secrete ATP capable of inhibiting sensory neurons (2). However study for tadalafil in urothelium is few, here we investigated the distribution of PDE5, as well as the molecular mechanism for tadalafil in signal transduction focusing on TRP channels and ATP release of bladder urothelium.

Methods

Primary urothelial cell cultures: Whole bladders were harvested from anesthetized rats, and urothelial cells were prepared as previously described (3).

Immunohistochemistry: Rat tissue was fixed with 4% PFA, cryosectioned (3 µM) and mounted on glass slides. Sections were incubated with primary antibody (rabbit anti-PDE5A [1:50] [Santa Cruz Biotechnology]) and subsequently, incubated with goat anti-rabbit HRP (1:1,000) (Santa Cruz Biotechnology). Samples were examined using an Olympus BX53 microscope.

Western blot analysis: Tissue specimens and primary rat urothelial cell cultures were homogenized using an M PER mammalian protein extraction reagent. Images were captured using an Image Quant LAS4000 imager. Anti-PDE5A (1:500) (Santa Cruz Biotechnology) and anti-β actin (1:20,000) (Santa Cruz Biotechnology)

Primary urothelial cell cultures: Whole bladders were harvested from anesthetized rats, and urothelial cells were prepared as previously described (3).

Results

Fig.1 PDE5 was expressed in the rat bladder urothelium

Fig.2 PDE5 was expressed in primary rat bladder urothelial cell cultures

Fig.3 Tadalafil-mediated inhibition of hypotonicity-induced Ca2+ influx

Fig.4 Tadalafil-mediated inhibition of Ca2+ influx induced by TRPV2 or TRPV4 agonism

Fig.5 Tadalafil-mediated inhibition of stretch-induced ATP release in mouse bladder urothelium

Conclusions

Tadalafil attenuated Ca2+ influx via TRPV4 and TRPV2 channels and inhibited ATP release in bladder urothelium. Tadalafil could act an inhibitory role in urothelial signal transduction. These novel insights into the mechanism underlying the effect of tadalafil on bladder sensation might inform the development of additional treatments for bladder disorders such as overactive bladder.

References


Conflict of Interest Disclosure

I have no potential conflict of interest to report