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Lee K[1], Isogai A[2], Mitsui R[2], Kajioka S[3], Eto M[4], Hashitani H[2]

1. *Department of Cell Physiology, Nagoya City University, Department of Applied Urology and Molecular Medicine, Kyushu University, Department of Urology, Kyushu University,* ***2.*** *Department of Cell Physiology, Graduate School of Medical Sciences, Nagoya City University,* ***3.*** *Department of Applied Urology and Molecular Medicine, Kyushu University,* ***4.*** *Department of Urology, Graduate school of Medical Sciences, Kyushu university*

FUNCTIONAL COUPLING OF TRPV4 CHANNELS AND BK CHANNELS IN REGULATING DETRUSOR SPONTANEOUS CONTRACTIONS IN THE GUINEA-PIG BLADDER

Hypothesis / aims of study

Transient receptor potential vanilloid 4 (TRPV4) channels function as a stretch-induced Ca^{2+} influx pathway triggering urothelial ATP release to initiate a micturition reflex in the bladder. TRPV4 are also expressed in detrusor smooth muscle (DSM) and may play a role in regulating DSM contractility upon bladder filling. GSK 10167090A (GSK), a TRPV4 channel agonist, potentiated the spontaneous contractions of DSM presumably by causing membrane depolarization and subsequent opening of L-type voltage-dependent Ca^{2+} channels (LVDCCs)[1]. Although these observed responses upon GSK administration are consistent with the opening of TRPV4 channels, the increased contractility of DSM would be rather undesirable during bladder storage.

A recent study demonstrated that TRPV4 channel knock out mice showed markedly higher voiding frequency and increased non-voiding contractions, which are believed to be myogenic origin [2]. Since the activation of TRPV4 channels in the urothelium would trigger a micturition reflex, the higher voiding frequency of TRPV4 knock out mice is rather unexpected. Thus TRPV4 channels may have an inhibitory role in DSM in addition to having an excitatory function in the urothelium-sensory nerve signal transmission. In cerebral arteriolar smooth muscle, stimulation of TRPV4 channels evokes the opening of large-conductance Ca^{2+}-activated K^+ (BK) channels to cause membrane hyperpolarization and vasodilatation [3]. Similar functional coupling of TRPV4 and BK channels may be operating in DSM.

Study design, materials and methods

Changes in the contractility of DSM bundles taken from guinea-pig bladders were measured using isometric tension recording. Membrane potential changes were recorded using intracellular microelectrode technique, while intracellular Ca^{2+} dynamics were visualized by Cal-520 fluorescent Ca^{2+} imaging. Distribution of TRPV4 in DSM was also examined by fluorescence immunohistochemistry.

Results

GSK10167090A (GSK, 1 nM), a TRPV4 channel agonist, caused a sustained contraction in DSM associated with a cessation of spontaneous phasic contractions in a manner sensitive to *HC*-067047 (10 μM), a TRPV4 channel antagonist (Figure 1A). DSM expressed TRPV4 channel immunoreactivity. GSK-induced cessation of spontaneous contractions were reversed by Iberiotoxin (100 nM) or paxilline (1 μM), BK channel blockers (Figure 1B), but not by apamin, a blocker for small-conductance Ca^{2+}-activated K^+ channels. The sustained contractions in GSK were reduced by nifedipine (10 μM), a blocker of LVDCCs by about 40%. GSK (1 nM) prevented spontaneous action potentials and hyperpolarized the membrane in DSM in a manner sensitive to iberiotoxin (100μM, Figure 1C). GSK (1 nM) also caused a cessation of spontaneous Ca^{2+} transients and an increase in the basal Ca^{2+} levels that were reversed by Iberiotoxin (100μM, Figure 1D).

Interpretation of results

The activation of TRPV4 channels with GSK exerted two opposing effects on the contractility of DSM in the guinea-pig bladder, i.e., GSK blocked spontaneous phasic contractions and evoked a sustained contraction, in a manner sensitive to a TRPV4 channel antagonist.

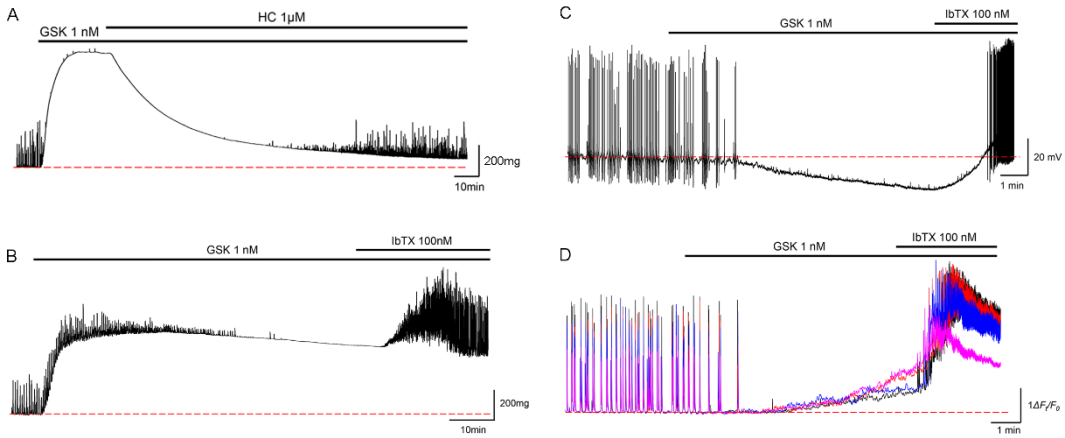
GSK-induced cessation of spontaneous contractions, action potentials and Ca^{2+} transients were reversed by BK channels blockers, indicating that TRPV4 channels in the guinea-pig DSM are functionally coupled with BK channels.

GSK evoked sustained contractions that were reduced by nifedipine even in the presence of a BK channel-mediated membrane hyperpolarization. Since LVDCCs are known to undergo Ca^{2+}-sensitive inactivation and facilitation, the massive increase in intracellular Ca^{2+} may tonically facilitate LVDCCs presumably by Ca^{2+}/calmodulin-dependent phosphorylation.

Concluding message

This study, for the first time, demonstrated that stretch-activated, Ca^{2+}-permeable TRPV4 channels appear to be functionally coupled with BK channels to stabilize DSM excitability. Thus, DSM TRPV4 channels may paradoxically act as self-limiting mechanism upon bladder filling, while urothelial TRPV4 channels may play a critical role in the initiation of the micturition reflex. Pharmacological manipulation of urothelial TRPV4 channels could have potential for the treatment of overactive bladder, but particular caution should be paid to deleterious modulations of DSM contractility to avoid unexpected outcomes.

Figure 1. Effects of GSK on spontaneous contractile, electrical and intracellular calcium activity in DSM



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Disclosures

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