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THE ROLE OF VOLTAGE-GATED K+ CHANNEL 2 IN RAT BLADDERS WITH DETRUSOR INSTABILITY(DI)

Hypothesis / aims of study

To investigate the role of voltage-gated K^+ channel 2 in rat bladders with detrusor instability(DI)

Study design, materials and methods

Healthy female Wistar rats of clean grade,2-3months of age, body mass 180-210g, were prepared. 40 rats were used to form bladder instability, the other 10 served as controls.The rat bladders with DI were identified by the urodynamics. The bladder cells with DI were cultured primarily, divided into three groups: instability detrusor group; pcDNA (1ug) transfection group; pcDNA (1ug) + Kv2.1 (1ug) transfection group.10 normal rats were used as control group. Detrusor cells in rats in vitro were originally cultured. Phase-contrast microscopy and staining of bladder smooth muscle cells were determined to conformed the success of primary culture. A stable transfected pcDNA3-Kv2.1 cell line was established.The whole-cell patch clamp method was applicated to measure the voltage-gated potassium current, and the methods of PCR and protein determination methods were evaluated to observe the effect of cell transfection.

Results

Five rats encountered accidental death in the obstruction group,and four rats appeared partial bladder leak. The successful rate of detrusor instability was 74.19% (23/31). The bladder capacity and bladder compliance were significantly defferent between the DI group and the control group. Bladder instability in the control group:7 rats; pcDNA (1ug) transfection group:8 rats; pcDNA (1ug) + Kv2.1 (1ug) transfection group:8 rats.10 rats was used as normal control group. Four groups of rats grew well, weight gain was no significant difference (P> 0.05).The detrusor cells of all groups were cultured primarily.The primary cells were round, smaller, uniform size. 3 hours after inoculation, the smooth muscle cells could be affixed to the wall.After 12-hour observation,we found that some cells began to stretch, and the cells became larger, arranged spindle-shaped. 6 days -7 days after inoculation,the conspicuous "peak" and "trough"were shaped when cell fusion,with positive staining by α-smooth muscle actin. ß-actin as a control, the expression levels of Kv2.1 mRNA in the control group was(0.86±0.12), while the DI group was (0.36±0.08). Compared with the DI group,the expression levels of Kv2.1 mRNA and protein in pcDNA (1ug) + Kv2.1 group were significantly different((P <0.05). Formed the whole-cell configuration, maintained at the voltage-70mV, imposed 10mV step voltage stimulation from-40mV to +50 mV,Kv2.1 current was recorded in 300ms. Compared with the DI group, the voltage-gated potassium currents and cell current density were statistically significant (P <0. 05) in the normal control group,pcDNA (1ug) transfection group and pcDNA (1ug) + Kv2.1 group.

Interpretation of results

Lower urinary obstruction is a dependable method for the generate of DI.Kv2.1 is important for the emerging of bladder detrusor excitability,which show Kv 2.1 might be one of the therapeutic targets for DI

Concluding message

Kv2.1 is important for the emerging of bladder detrusor excitability,which show Kv 2.1 might be one of the therapeutic targets for DI

Disclosures

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