C4

Actin Filaments Regulate the Stretch Sensitivity of Large Conductance Ca^{2+}-Activated K^+ Channel in Rabbit Coronary Arterial Smooth Muscle Cells

Lin Piao*, Yung E Earm, Wonkyung Ho.

Department of Physiology, College of Medicine, Seoul National University

The large conductance Ca^{2+}-activated K^+ channels (BK_{Ca}) in vascular smooth muscle have been considered to function as a negative feedback in pressure-induced vasoconstriction. In the present study, the function of cytoskeletons in the regulation of BK_{Ca} and its stretch sensitivity was investigated. Using the inside-out patch clamp technique, we recorded single channel activities of BK_{Ca} with 150 mM KCl in the bath solution (pCa = 6.5). BK_{Ca} was identified by the large unitary conductance (about 300 pS), the voltage- and calcium-dependence, and by 100 nM iberiotoxin, a specific blocker of BK_{Ca}. Applying cytochalasin D (1 μM), an actin filaments disrupter, increased the open probability (NP_o) from 0.15 ± 0.03 to 0.26 ± 0.06 (mean ± SE, n = 4, $p < 0.01$), and this increase was largely reversed by phalloidin (1 μM), an actin filaments stabilizer, to 0.03 ± 0.01 (n = 4, $p < 0.01$). NP_o was also increased by chochicine (10 μM), a microtubules disrupter (from 0.14 ± 0.03 to 0.31 ± 0.07, n = 4, $p < 0.01$), and decreased by taxol (1 μM), a microtubules stabilizer to 0.04 ± 0.05 (n = 4, $p < 0.01$). NP_o with no pressure was 0.11 ± 0.03 and was increased to 0.54 ± 0.06 (n = 4, $p < 0.01$) at -30 cm H_2O. In the presence of phalloidin, applying negative pressures failed to induce a significant increase in NP_o. And NP_o at -30 cm H_2O and that at -40 cm H_2O were 0.07 ± 0.01 and 0.10 ± 0.01 (n = 4). On the contrary, in the presence of taxol, the negative pressure of -30 cm H_2O caused an increase in NP_o to the similar extent compared to the control (5.72 ± 1.03 folds increase in control; 6.50 ± 1.26 folds increase in the presence of taxol. n = 4, $p > 0.05$). So we concluded that membrane stretch activated BK_{Ca} in coronary arterial smooth muscle cells. Both actin filaments and micortubules modulate the activity of BK_{Ca}, while only actin filaments regulated stretch-sensitivity of BK_{Ca}.