

**Background K<sup>+</sup> channel currents in WEHI-231 cells, immature B lymphocytes**

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In our previous study, WEHI-231, an immature B cell line, showed intractable increase in  $[Ca^{2+}]_c$  after the B-cell receptor (BCR) ligation and treatment with 2-aminoethoxydiphenylborate (2-APB), which was never observed in Bal-17, a mature B cell line (Nam et al., 2003, FEBS Lett). In this study, a whole cell voltage clamp study revealed a specific expression of a novel type of K<sup>+</sup> current, namely voltage-independent background-type K<sup>+</sup> channels (IK-bg), in WEHI-231 cells. IK-bg was dramatically increase by the application of 2-APB (50  $\mu$ M), which induced severe hyperpolarization of WEHI-231 from -45 mV to -90 mV. When dialyzed with Mg<sup>2+</sup> and ATP-free pipette solution, a spontaneous development of IK-bg and membrane hyperpolarization were observed. IK-bg was insensitive to classical K<sup>+</sup> channel blockers (TEA, glibenclamide, Ba<sup>2+</sup> (1 mM)), whereas blocked by quinine and quinidine in a voltage-dependent manner (IC<sub>50</sub>=6~9  $\mu$ M at +60mV). Phorbol myrstate, a PKC activator, decreased the amplitude of IK-bg. Extracellular acidification (pH 6.5) slightly inhibited IK-bg. Arachidonic acid, riluzole, or hyposmotic stress could not affect the IK-bg after the full development by the intracellular dialysis with Mg-ATP-free solution. In a cell-attached mode of single channel recording from WEHI231, we found two types of voltage-independent K<sup>+</sup> channels with unitary conductance of 300 pS and 120 pS, respectively. Both channels showed very short mean open times and their open probabilities were increase by the application of 2-APB. In Bal-17 cells, no such K<sup>+</sup> current was observed in 50 cells tested. In summary, WEHI-231 immature B cells express background K<sup>+</sup> channels. The pharmacological properties and the large unitary conductance suggest that novel types of two-pore domain K<sup>+</sup> channels (2-P-K channels) might be expressed in WEHI-231, which may provide an intriguing targets of signal transduction in the immature B lymphocytes.