ELECTROPHYSIOLOGICAL STUDY OF VOLTAGE-GATED ION CHANNELS

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Characteristics of voltage-gated cation channels in the cell membrane were studied in this thesis.

1. We examined the effect of certain fatty acids (FAs) on the kinetics of $K_V1.3$ channel gating of PHA- and IL-2-stimulated human peripheral lymphocytes (HPLs). Our study indicated that the effect carried out by FAs depended on the degree of unsaturation and/or the chain length of the FAs. All of the studied polyunsaturated FAs (PUFAs) [linoleic acid (LA), arachidonic acid (AA) and docosahexaenoic acid (DHA)] influenced the activation and inactivation kinetics of the $K_V1.3$ channels of HPLs (decreased the time constants mentioned above), whereas the monounsaturated FA (MUFA) oleic acid (OA) and the saturated FAs (SFAs) [palmitic acid (PA) and stearic acid (SA)] were ineffective. This pattern parallels the efficacy of FAs to interfere with lymphocyte activation processes. However, these FAs did not affect the voltage-dependence of steady-state activation and steady-state inactivation of the channels. The efficacy of the incorporation of FAs into the HPL membrane was confirmed by gas chromatographic measurements, each treatment specifically increased the relative contribution of the corresponding FA to the total amount of FAs in plasma membrane of HPLs.

   We have demonstrated here that enrichment of the membrane with various PUFAs can modulate the kinetics of $K_V1.3$ channels in HPLs probably via alteration of microenvironment in the lipid bilayer. In this way, a diet rich in PUFAs can be important in maintaining the suitable lipid composition of the membrane for proper operation of these channels or induction of lipid correction. T-cell activation is based on the operation of the voltage-gated K^+ channels, the Ca^{2+} activated K^+ channels and the Ca^{2+} release activated Ca^{2+} channels, and $K_V1.3$ channels expressed in T-lymphocytes control the membrane potential and signal transduction. Therefore the PUFA-induced modification of $K_V1.3$ kinetics might have important consequences regarding the inhibition of the activation of T-cells under pathophysiological conditions. This might contribute to the beneficial effects of dietary PUFA supplement in autoimmune reactions and/or during chronic inflammation.
2. We have revealed a relationship between the order of K^+ currents and adenosine receptor stimulation or inhibition in DDT_1 MF-2 smooth muscle cells. On the basis of both this present study and our earlier measurements presented by Bálint Rubovszky in his Ph.D. thesis, we can conclude that applied adenosine agonists (NECA, F-NECA, CPA, CGS 21680) and antagonist (ZM 241385) take part in the adenosine mediated process and verify the presence of A_1 and A_2 adenosine receptors in the studied cell line. During the changes in the properties of K^+ current, receptors act on the channels in a G protein independent way. On the other hand, the examined P_1 agonists and antagonists did not influence the conductance of Na^+ channels in this smooth muscle cells.

   DDT_1 MF-2 cells have proved to be unique to response to both the A_1 receptor activation with increase in K^+ conductance and hyperpolarization, and the A_2 receptor activation with decreased K^+ current and depolarization. In this way, these cells can be useful targets of pharmacological research, because they provide the possibility of tracking A_1 and A_2 receptor mediated mechanisms on the same cell line.

Keywords: fatty acids, T-lymphocyte, adenosine agonist, adenosine antagonist, smooth muscle cell