THE ROLE OF PURINERGIC SIGNALING AND DEITERS CELLS IN THE INNER EAR

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Besides conventional neurotransmitters purinergic signal transduction may play a significant role in the Ca^{2+}-dependent cochlear adaptation processes. The first part of the dissertation examines the expression of purinergic receptors in outer hair cells (OHCs) of guinea pig and the relationship between receptor expression and the characteristics of the Ca^{2+} signal along with the characterization of changes in the above mentioned parameters upon noise exposure. We found that extracellular ATP triggers a fast and non-desensitizing rise in the cytosolic free Ca^{2+} concentration ($[Ca^{2+}]_i$) in some cells whereas other cells display slow and desensitizing $[Ca^{2+}]_i$ response to ATP exposure. Some cells responding to ATP did not show a characteristic rise in $[Ca^{2+}]_i$ upon exposure to UTP. Based on these data we hypothesized that the rise in the $[Ca^{2+}]_i$ is mediated by the activation of multiple P_2 receptor types. Immunocytochemical staining of OHCs showed marked signal upon labeling with fluorescence-conjugated antibodies against P_{2X1}, P_{2X2}, P_{2X4}, P_{2Y1}, $P_{2Y2,}$ P_{2Y4} receptors whereas only a weak signal was detected with the antibody directed to the P_{2X7} receptor. The distribution of the receptor subtypes along the outer hair cell was also different. Whereas the distribution of P_{2X1} P_{2X4} and P_{2Y1} receptors was homogenous, the P_{2X7} and P_{2Y4} receptors could only be observed weakly at the basal pole whereas the P_{2X2} and P_{2Y2} localized mostly at the apical part of the OHCs, close to the cuticular plate. Moderate chronic noise exposure of the animals (80 dB, 14 days) resulted in elevated resting $[Ca^{2+}]_i$ in OHCs isolated from both the basal and the apical turns of the cochlea. The rise in the resting $[Ca^{2+}]_i$ was more prominent in OHCs isolated from the basal turns where OHCs are more susceptible to noise-induced damage. The ATP-induced $[Ca^{2+}]_i$ transients had smaller amplitude and longer duration in OHCs isolated form noise-exposed animals with a more prominent decrease in the amplitude of the $[Ca^{2+}]_i$ transients in OHCs isolated from the basal turns. Noise exposure also decreased the expression of P_{2X1}, P_{2X2}, P_{2X4}, P_{2X7} és P_{2Y1}, P_{2Y4} purinoceptor subtypes whereas that of the P_{2Y2} subtype did not change as compared to the control. The alterations in the purinergic signal transduction pathways described in this study may contribute to the noise exposure-induced changes in the function of outer hair cells which leads to sensoryneural hearing loss.

Due to their anatomical location Deiters cells and OHCs constitute a micromechanical unit of the inner ear. Membrane-potential driven processes of Deiters are determined by the activity of K^+ channels located in the cytoplasm membrane. The second part of the dissertation addresses the biophysical and pharmacological characterization of the voltage-gated K^+ channels of Deiters cells along with the cell-shape dependent expression of the K^+ channels. Based on the biphasic inactivation kinetics and the analysis of the voltage-dependence of steady-state activation and inactivation of the whole cell currents we concluded that two different K^+ channels types are expressed in Deiters cells. This conclusion was justified by pharmacological separation of the currents using TEA and charybdotoxin. Based on the pharmacological and biophysical properties, Kv1.3 channels may be responsible for the more rapidly inactivating current component. The kinetic analysis of whole-cell currents showed that morphologically different Deiters cells (corpulent and lanky cells) express the same K^+ channel types although cells having lanky cell body express more channels with faster inactivation kinetics, which also leads to larger overall peak currents in these cells. The difference in the K^+ channel expression of Deiters cells of different cochlear locations and morphology might allow differential regulation of the micromechanical properties of the inner ear as well as the contribution of these cells to K^+ recirculation.