16P.6 Large conductance potassium channel opener NS1619 regulates endothelial function

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Mitochondria play crucial role both in energetic and regulatory pathways within the cell. Inner mitochondrial membrane contains various ion channels, among which potassium channels are well described due to protective activities. Large conductance calcium activated potassium channel (BK_{Ca}) can be activated by channel openers such as NS1619 (1,3-dihydro-1-[2-hydroxy-5-(trifluoromethyl)phenyl]-5-(trifluoromethyl)-2*H*-benzimidazole-2-one). NS1619 can regulate functioning of endothelial cells EA.hy 926 in many aspects. In our study it was shown that NS1619 changes mitochondrial function both by decreasing mitochondrial potential and by increasing oxygen consumption probably due to activating BK_{Ca} channels present in the inner mitochondrial membrane and thus promoting K^+ flux. Additionally NS1619 caused increase in calcium concentration within the endothelial cells. Calcium is well known regulator of many signaling pathways within the cells. Ionophore A23187 (1 μM) causes increase in calcium concentration, which subsequently increased nitric oxide (NO) production in EA.hy 926 cells *via* activation of nitric oxide synthase. Similar activity is proposed for NS1619. Along with these results it was observed that NS1619 increased coronary flow in isolated guinea pig hearts in NO dependent manner (100 μM L-NAME, inhibitor of nitric oxide synthase, partially reversed the effect of NS1619). It seems that NS1619 can have beneficial effect on endothelium *via* vasodilating activity, however, the exact mechanism which seems to involve both BK_{Ca} channel activation and other places of action, needs further investigation.

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16P.7 Cytoprotective action of the potassium channel opener NS1619 under conditions of disrupted calcium homeostasis

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Cytoprotective properties of potassium channel openers (KCOs) have already been shown in several models of cell injury, mainly in ischemia–reperfusion-induced damage of cardiac muscle. The mechanism responsible for the observed cytoprotection as well as the relative contribution of potassium channels located in the plasma membrane and in the inner mitochondrial membrane to the beneficial effects exerted by KCOs remains unclear. This work demonstrates the cytoprotective properties of NS1619, an opener of large-conductance calcium-activated potassium channels (BK_{Ca} channels), in C2C12 myoblasts injured by calcium ionophore A23187 treatment. Application of two BK_{Ca} channel inhibitors, paxilline and iberiotoxin, abolished this cytoprotective effect. At the applied concentrations (10–100 μM), NS1619 increased the respiration rate of C2C12 cells in a dose-dependent manner. However 0.2 μM paxilline, which effectively abolished the protective effect of NS1619, failed to counteract the opener-induced increase in cellular respiration. This result indicates that the NS1619-mediated increase in the survival rate of A23187-treated C2C12 cells is distinct from its effect on mitochondrial functioning and suggests that activation of BK_{Ca} channels in the plasma membrane is responsible for cytoprotection by NS1619.

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16P.8 Influence of ATP-sensitive potassium channel activities on respiration and membrane potential in plant mitochondria

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We describe the existence of a potassium ion transport mechanism in the mitochondrial inner membrane of plants. We found that substances known to modulate ATP-sensitive potassium channel ($mitoK_{ATP}$) activity influenced the bioenergetics of potato (*Solanum tuberosum)* tuber mitochondria, i.e. the rate of resting respiration and membrane potential. In isolated mitochondria, diazoxide (a potassium channel opener) was found to depolarize the mitochondrial membrane potential (measured with a TPP^+-specific electrode) and to stimulate resting respiration. These effects were blocked by glibenclamide and ATP, potassium channel blockers, dependently on the presence of potassium ions in the incubation medium. We investigated monovalent cation (chloride salts) selectivity of the diazoxide-induced ATP-sensitive mitochondrial membrane depolarization. Pharmacological profile and immunoreactivity with specific antibodies indicate that the plant $mitoK_{ATP}$ channel belong to inward rectifier K^+ channel family — Kir.6.x. Our results suggest that an ATP-sensitive potassium channel similar to that of mammalian mitochondria is present in plant mitochondria.

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16P.9 Kidney cortex mitochondria are non-functional in a potassium-based media whereas heart mitochondria improve with increasing potassium concentration

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A medium of containing high levels of potassium chloride (KCl) is commonly used when assessing respiratory function of isolated mitochondria from various tissues. However, the measured intracellular [K^+] in kidney proximal tubular cells is about 60 mM and in cardiac myocytes approximately 130 mM. Therefore, the use of a similar media [K^+] for all tissues seems unsupported. Here we investigated the effect of different [K^+] on respiratory function in mitochondria isolated from kidney cortex and heart of healthy male Sprague–Dawley rats. Oxygen consumptions and the respiratory