We focused on extracellular flagellar filament assembly and complementation of motility in *B. subtilis* flagellin mutant. First, we constructed the flagellin defect strain of *B. subtilis* using the chromosomal homologous recombination. Complementation test was carried out with *B. subtilis* flagellin mutant strain using *E. coli–B. subtilis* shuttle vector, harboring *B.* sp. PS3 flagellin gene. The results from swarming assay with soft agar plate indicated that the R91H, G185D and G202D mutation of *B.* sp. PS3 flagellin was found to complement the *B. subtilis* flagellin mutant and the wildtype flagellin gene failed to complement. However, it was about 30% of wildtype *B. subtilis* even if most restored G185D variant. We assumed that the slight motility complementation was probably caused by short flagellar filament. In fact, short filament was confirmed by dark-field microscopic observation of vortex mixing detached flagellar structure from motility restored cell. Additionally, to ascertain functional significance of the terminally conserved hydrophobic residues, we constructed substitutions of glutamate for these hydrophobic residues.

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S3.28 Potassium and chloride channel activities from potato *Solanum tuberosum* tuber mitochondria

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Mitochondrial potassium channels, such as ATP-regulated or large conductance Ca^{2+}-activated exist both in mammals and plants. Basic effects of these channel activity include changes in mitochondrial matrix volume, mitochondrial respiration and membrane potential, and generation of reactive oxygen species. The aim of this study was to describe chloride and potassium channels from potato tuber mitochondria. Single channel activities were measured after reconstitution of the inner mitochondrial membranes into planar lipid bilayers. Three potassium channels and two chloride channels were observed. After incorporation, in a gradient of 50/450 mM KCl (*cis*/*trans*), we found that 1 mM Mg/ATP and 200–800 nM iberiotoxin (IbTx) inhibited two potassium channel activities, the ATP-regulated and the Ca^{2+}-activated with large conductance, respectively. Furthermore, it was shown that the chloride channels are inhibited by 4,4′-diisothiocyanostilbene-2,2′-disulfonic acid (DIDS). We observed also that 1 mM Mg/ATP and 400 nM 5,6-dichloro-1-ethyl-1,3dihydro-2H-benzimidazol-2-one (DCEBIO) activated one of these channels. Moreover, we found that the substances known to modulate potassium channel activities (the ATP-regulated and the Ca^{2+}-activated) influenced the bioenergetics of isolated potato tuber mitochondria, i.e., the rate of resting respiration and membrane potential.

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S3.29 Ion channels from the inner mitochondrial membrane from rat heart — single channel properties

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Mitochondrial ion channels are objects of electrophysiological and pharmacological studies for over 10 years. It is known that they are involved in cytoprotection and apoptosis. In our study we investigated ion channels from the inner mitochondrial membrane of heart mitochondria. We recorded single channel activity using patch-clamp technique. An anion channel in the inner mitochondrial membrane from rat heart was observed. In symmetrical 150/150 mM KCl solution we recorded a chloride channel with conductance 120 pS. The effect of different channel inhibitors and activators (DIDS, SITS, DCEBIO) on the anion channel activity was studied. We plan to characterize its electrophysiological and pharmacological properties.

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S3.30 Cytoprotective effects of mitochondrial potassium channel opener BMS-191095

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Mitochondrial potassium channel openers (KCO's) were shown to be cytoprotective in models of ischemia–reperfusion induced injury in brain, heart and skeletal muscle tissue. The aim of this study was to identify the cellular events responsible for observed protection. We have investigated the cytoprotective potential of BMS-191095, an opener of the mitochondrial ATP-regulated potassium channel (mitoK_{ATP}), in C2C12 myoblasts. BMS-191095 did not protect the cells against *tert*-butyl hydroperoxide or H_2O_2-induced injury, but prevented calcium ionophore A23187-induced cell death. A23187 caused a transient increase in cytosolic calcium levels, which was not affected by the presence BMS-191095. On the contrary, the opener increased the cell survival and prevented the loss of cell membrane integrity and the appearance of sub-G1 fraction observed after A23187-treatment. At comparable concentrations the opener increased respiration rate and decreased mitochondrial membrane potential of C2C12 myoblasts, which confirms that mitochondria are the site of action of this drug. Since 'mild uncoupling' of mitochondrial oxidative phosphorylation is considered as a potential mechanism of cytoprotection, these results may at least partially explain the beneficial effects of KCO's on cell survival in the conditions of disrupted calcium homeostasis.

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S3.31 A *Bacillus* flagellar motor switches from proton to sodium gradients for powering motility at alkaline pH

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