Flash Induced Charge Displacement Signals in an Artificial Layer of Bovine Photoreceptor Membrane Fragments

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A new method has been developed for studying fast photovoltages (FPV) produced by bovine visual pigment rhodopsin in a layer of rod outer segment membrane fragments that is attached to cellulose acetate or nitrate filters. The filter is fixed into an electrolyte chamber with two low impedance electrodes. In this cell arrangement the aqueous medium has access to both sides of the layer and can easily be changed to any condition. In normal Ringer solution both the R_1 and R_2 component are present. The signal shows a close similarity to that obtained from rhodopsin in the intact receptor structure.The signal waveform is given by a superposition of an "active" membrane charge displacement and a "passive", simultaneous discharging process. In the low temperature range the passive time constant is much smaller than the active one. In this case the R_2 amplitude is markedly reduced and the active process determines the signal decay (s. Poster Hochstrate et al.). The temperature dependence of the R_2-generating process could be measured in the range between 10 and 60 °C. In this region the Arrhenius plot of the active time constant is nonlinear and shows a close correlation with that of the reaction time for the slow phase of the meta-transition in the rhodopsin photolysis. The active time constant decreases if the pH in the medium is lowered or if alcohol is added. These results can be explained by a corresponding change of the activation energy for the charge displacement process. Alcohol addition also affects the early part of the FPV. If 10 % ethanol is added at 0 °C the R_1-component is completely diminished. It is known from spectroscopic studies of the rhodopsin photolysis that the alcohol decreases the surface pressure of rhodopsin containing membranes. If the membrane pressure is markedly reduced by alcohol addition the FPV is largely comparable to those signals which are obtained from rhodopsin containing monolayers spread out at a hexane-water interface.

According to the free medium access to both sides of the rhodopsin layer the artificial system opens a new possibility to study in more detail the molecular events which generate both components of the fast electric signals of rhodopsin.