Photochemistry and light-dark adaptation of monomeric and aggregated bacteriorhodopsin in various lipid environments

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The organisation of bacteriorhodopsin (BR) into a two-dimensional hexagonal lattice of protein trimers is a special feature of the purple membrane. In order to study the effect of this unusual state of aggregation on the functional properties of this light-driven proton pump, the photocycle and the light-dark adaptation reaction of monomeric and aggregated BR were compared. In BR-dimyristoylphosphatidylcholine vesicles reversible protein aggregation/disaggregation can be induced by temperature; above 23°C BR is monomeric whereas below 11°C it is aggregated in the same hexagonal lattice as in the purple membrane. The experiments indicate that the photochemical cycle of trans BR is qualitatively the same in the aggregated and monomeric state. The temperature dependence of the rate constant for the decay of the intermediate 411-T (M) was carefully measured between 1 and 45°C. No break in the Arrhenius plot of this reaction occurs at the temperature of the lipid phase transition (23°C). The plot exhibits, however, a small discontinuity around 16°C, i.e. in the temperature region of the BR aggregation/disaggregation transition. Activation energies of 14.4 and 14.1 kcal/mol for BR in the aggregated and monomeric state, respectively, have been calculated. Furthermore, the activation energies for dark adaptation of monomeric and aggregated BR are very similar (21.9 and 21.6 kcal/mol, respectively) and in close agreement with the value for intact purple membrane. Again, the Arrhenius plot shows a discontinuity around 16°C, which is even more pronounced for this reaction. Although the kinetics of dark adaptation are independent of BR's state of aggregation, its extent was found to be smaller for BR monomers. Whereas the aggregation state does not influence the kinetics of the photocycle and of the light-dark adaptation, these reactions are strongly affected by BR's microenvironment. The kinetic data and spectral features of BR in different solvents (e.g. detergent solutions) and lipids (DMPC, DPPC, asolectin, diphytanoylphosphatidylcholine) shall be discussed.