Dissertation abstract

Photosynthesis is the process by which light is converted into chemical energy. Not only green plants but also some bacteria are capable of this task. One such bacterium is *Rhodobacter sphaeroides* in which the light-driven reaction takes place in a membrane-bound protein complex called the reaction center (RC). When a photon is absorbed by the RC an electron transfer reaction is initiated that ultimately leads to a translocation of protons across the membrane. As a consequence a proton gradient is maintained across the membrane and the energy stored in this gradient is then utilized to drive reactions neccesary to sustain life in the organism.

The RC has been subjected to intense research over the last three decades and evidence is emerging suggesting that it undergoes a change in structure as a response to illumination. The main focus of the work in this thesis has been to investigate this hypothesis.

In absence of exogenous electron donors isolated RCs form a charge-separated state when subjected to illumination. If the duration of the illumination is short (<1 s) the RC rapidly relaxes back to the ground state. However, when the time of illumination is extended to several minutes a slow accumulation of a semi-stable, charge-separated state is observed. This is followed by a slow relaxation when the light is turned off. The results from kinetic analysis of the formation and decay of the semi-stable state is consistent with it being in a different conformation than the initially formed charge-separated state. Further support for this interpretation comes from optical and EPR spectroscopy on both native and copper-substituted RCs that had been subjected to different illumination histories.

The most direct way to study conformational changes is to compare the 3D structures from different states. A first step in achieving this goal has been taken with the successful crystallization of the RC in the lipidic cubic phase system. The crystals diffract down to 2.35 Å and the previously reported binding of a cardiolipin molecule on the surface of the RC could be verified. In addition, a novel chloride-binding site was identified.

Key words: *Rhodobacter sphaeroides*, reaction center, conformational changes, copper substitution, EPR, crystallography

ISBN 91-628-5529-8