INSTITUTIONEN FÖR KEMI OCH MOLEKYLÄRBIOLOGI

Ultrafast Structural Changes in a Bacterial Photosynthetic Reaction Center probed with XFEL Radiation

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Abstract

Photosynthesis is the process by which plants and many species of bacteria convert energy from sunlight into chemical energy used to power their metabolism. As these plants and bacteria are eaten, the chemical energy moves up the food chain and thus photosynthesis provides fuel for almost all life on Earth. Photosynthetic reaction centers are the workhorses of photosynthesis. Upon photo-excitation, these multi-domain integral membrane proteins drive an electron transport chain that results in a proton gradient across the cell membrane. The primary electron transport events are of great interest to the scientific community due to their near perfect efficiency and functional role in powering the biosphere. The articles that comprise this thesis deal with one such photosynthetic reaction center, that from the purple non-sulfur bacterium *Blastochloris viridis* (RC_{vir}). Spectroscopic studies of RC_{vir} have revealed that the initial charge-separation reactions occur on a time scale of picoseconds and raise interesting questions about the role of ultrafast structural changes in optimizing the efficiency of the overall process.

As X-ray free-electron lasers (XFELs) have been commissioned, the possibility of studying the initial light-driven reactions of the electron transport process through time-resolved crystallography has been realized. XFELs are powerful new X-ray sources that have a high peak brilliance and a pulse length three orders of magnitude shorter than the most advanced synchrotron source. Through the development of time-resolved crystallographic and solution scattering methods at XFELs, this thesis aims to deliver new information about the role structural changes play in guiding the charge separation reactions of photosynthesis.

A solution scattering experiment was performed to give physiological relevance to previous observations that multi-photon excitation led to quake like movements within RC_{vir} on the order of picoseconds. Oscillatory features were revealed following a single-photon absorption event, but these proved difficult to interpret structurally. This highlighted the need for time-resolved crystallography experiments that could directly visualize these structural changes. After optimizing crystallization methods to produce samples suitable for XFEL sources, a time-resolved crystallography experiment was conducted that captured the protein at two picosecond time-points following photo-excitation. These experiments allowed visualization of conformational changes that evolved over time and it is hypothesized these structural dynamics may play a role in altering the activation energies of the electron transport process.