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Report:

The bacterial reaction center from *Rhodobacter sphaeroides* belong to type II reaction centers where the final electron acceptor is a ubiquinon molecule. Photosynthetic reaction centres are the basic functional protein complexes that can perform the primary photochemical event in photosynthesis: light driven charge separation. Following photoexcitation an electron flows from the special pair of chlorophylls to the dissociable ubiquinon (Q_B), which is consequently reduced to semiquinol. The semiquinol is further reduced to ubiquinol in a second light driven electron transfer step.

At Chalmers we have grown crystals of bacterial reaction centre from *Rhodobacter sphaeroides* using lipidic phase crystallisation[1]. This crystal form, which forms stacked two dimensional crystals, enables us to study the structure of an integral membrane protein in a “native like” environment. Crystals of photosynthetic reaction center from were grown in the presence of 60 % monoolein and 40 % protein solution. Plate like crystals of about 0.05 mm x 0.05 mm x 0.02 mm were grown in three days at room temperature. These crystals were screened for diffraction at MAX-LAB beamline I711, and the best crystals diffracted to 3.3 Å resolution in Lund. The well diffracting crystals were saved at liquid nitrogen temperatures, and data were

collected from the same crystals at the ESRF. Due to the higher brilliance at the ESRF the structure could be refined to 2.35 Å resolution.

A paper describing this new crystal form was published in Journal of Molecular Biology.[2] We believe that this work is of interest since this is the first lipidic cubic phase structure to be reported from a membrane protein not belonging to the archeal rhodopsin family. There are also some interesting new insights regarding the plane of the membrane, crystal contacts mediated via lipid molecules, and a minor conformational change relative to the highest resolution detergent structures[3]. Furthermore the crystals screened in Lund were used for intermediate trapping experiment at the ESRF. There, the crystals were illuminated with an IR laser to generate the $P^{+}Q_{A}^{-}$ charge separated state. Statistical analysis of the data showed that a stable conformational intermediate builds up at 100 K. Conformational changes were previously reported in this state by using limited proteolysis experiments and kinetic analysis [4; 5]. The observed changes from the limited proteolysis experiment agrees remarkably well with our results. A manuscript describing these conformational changes is under preparation.

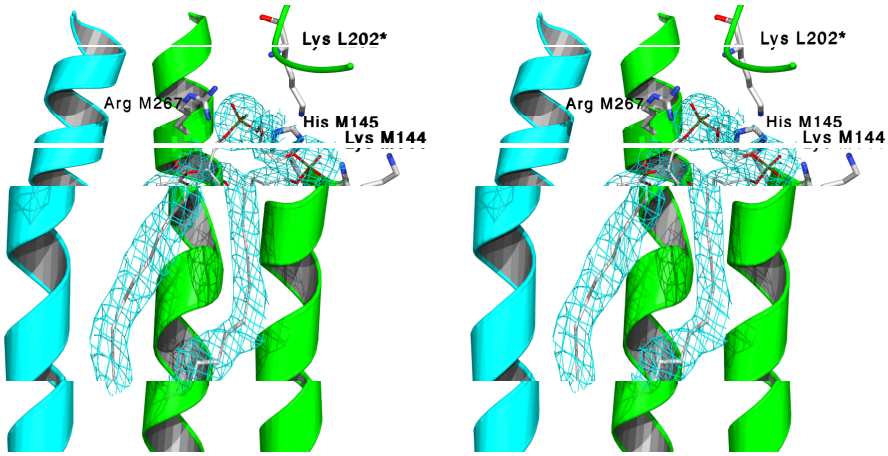

Figure 1: Electron density for the cardiolipin molecule, which is involved in an important crystal contact

References:

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