Photosystem II Reaction Centre Assembly Complex (RCII) - from Purification to 3D Structure

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Photosystem II (PSII) is a unique multi-component oxidoreductase embedded in the thylakoid membranes (TM) of cyanobacteria and chloroplasts. It catalyses the solar energy-powered water splitting which provides reducing equivalents to fix carbon dioxide and, as a by-product, it produces molecular oxygen. The biogenesis of PSII occurs in a step-wise manner, by the association of so called assembly modules, each formed by one of the large reaction centre core proteins (D1, D2, CP47, or CP43) together with low-molecular-mass subunits, pigments, and auxiliary proteins (Komenda et al. 2012). In the early stage of the assembly the D1 and D2 modules combine to form the first PSII intermediate, reaction center assembly complex, RCII (Komenda et al. 2008; Knoppová et al. 2014).

In the current study, the RCII has been isolated using a *Synechocystis* PCC 6803 mutant expressing the 6xHistidine-tagged D2 protein employed as an anchor for the complex purification, and unable to form the functional Photosystem II because of the lack of the CP47 antenna (6xHis-D2/ΔCP47). The obtained preparation contains two previously described variants of the reaction centre, RCIIa and RCII*, differing in the presence of RCII-specific auxiliary proteins (Komenda et al. 2008; Knoppová et al. 2014). Both RCIIs contain D1, D2, cytochrome b-559 and PsbI subunits, and the essential Ycf48 (Yu et al. 2018) and RubA (Kiss et al. 2019) factors. Additionally, the RCII* binds also the Ycf39/HliC/HliD auxiliary complex implicated in RCII photoprotection (Knoppová et al. 2014; Komenda and Sobotka 2016). Further, the complexes contain pigment and heme cofactors, i.e. chlorophyll, pheophytin, beta-carotene and cytochrome b-559, in ratios close to those expected from the PSII crystal structure, and the preparation is already capable of the primary photochemical reactions.

The RCII preparation contains also a high molecular weight complex of the RCII anchored to Photosystem I monomer [RCII-PSI(I)]. Although this association is highly likely an artifact, its size, abundance and stability gave us an opportunity to gain a high resolution cryo-EM 3D structure of the RCII assembly complex. The preliminary results provided by the team of Prof. Sazanov (IST Austria) will be also presented.

References:

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