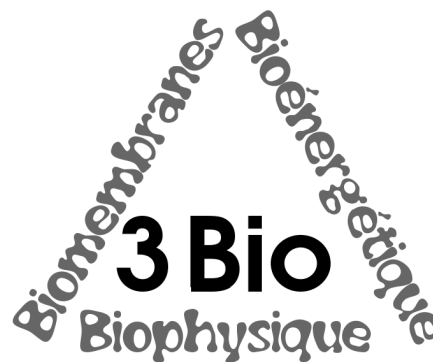


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New insights into the photosystem II mechanism from the 1.9 Å resolution structure

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Photosystem II (PSII) performs light-induced electron transfer and water-splitting reactions, which lead to the formation of molecular oxygen. PSII from thermophilic cyanobacteria consists of seventeen membrane-spanning subunits, three hydrophilic, peripheral subunits, and many cofactors with a total molecular weight of 350 kDa for a monomer. X-ray crystal structures of PSII have been reported at 3.8–2.9 Å resolutions for PSII isolated from *Thermosynechococcus elongatus* or *T. vulcanus*, which provide arrangement of protein subunits and most of the cofactors involved in the electron transfer reactions. However, the detailed structure of Mn₄Ca-cluster, the catalytic center of light-induced oxygen evolution, has not been resolved. In order to elucidate the detailed structure of PSII, especially the structure of the Mn₄Ca cluster, PSII crystals with a higher resolution are required. In order to obtain higher resolution PSII crystals, we screened the crystallization conditions for PSII dimers purified from *T. vulcanus* extensively, and optimized the conditions for crystallization, post-crystallization treatment and crystal harvesting. As a result, we succeeded in improving the crystal resolution significantly. Diffraction experiments with the new-type of PSII crystals using X-rays from a Synchrotron radiation source gave rise to diffraction spots visible beyond 1.8 Å resolution and its diffraction data was processed to 1.9 Å resolution. Electron density distributions for each of the 5 metal ions in the Mn₄Ca-cluster are clearly separated, allowing us to locate the individual metal ions and all of the ligands to the metal cluster unambiguously. 5 oxygen atoms forming oxo-bridges between the metal ions are identified, and 4 water molecules are found to be associated with the metal cluster, some of which may serve as substrates in the oxygen-evolving reaction. The most significant structural feature of the Mn₄CaO₅ cluster is that the distances between O5 and Mn1, Mn4, and Ca are remarkably longer. This implies that the bonds between O5 and each of the metal atoms are very weak and that O5 is different from the other 4 oxygen atoms and may have a higher reactivity. In addition, W2 and W3 are located most closely to O5. These distances are 3.1 Å and 3.0 Å, respectively. Moreover, the distance between W2 and W3 is 3.3 Å. Thus, it is most probable that O–O bond formation is taken place within the 3 species of W2, W3 and O5.

Invitation: Alain Boussac

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