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Structure of Protochlorophyllide Reductase: a Greening Mechanism for Plants in the Dark.

<u>Norifumi Muraki</u>^a, Jiro Nomata^b, Yuichi Fujita^b, Genji Kurisu^a. ^aInstitute for Protein Research, Osaka University, 3-2 Yamadaoka, Suita, Osaka 565-0871, Japan. ^bGraduate School of Bioagricultural Sciences, Nagoya University, Furo-cho, Nagoya, Aichi 464-8601, Japan.

E-mail: nmuraki@protein.osaka-u.ac.jp

Chlorophyll (Chl) is a tetrapyrrole macrocycle containing Mg and phytol chain. The Chl biosynthetic pathway consists of the multi-enzymatic reactions. An asymmetric conjugated double bond system of Chl a, which is crucial for efficient light absorption, is formed in the penultimate step of biosynthesis, reducing protochlorophyllide (Pchlide) to form chlorophyllide a. Photosynthetic organisms adopt two different strategies for the reduction of Pchlide; one is the dark-operative (lightindependent) Pchlide oxidoreductase (DPOR) that operates even in the dark. The greening ability of plant in the dark is attributed to the activity of DPOR. DPOR is characterized to be a nitrogenase-like enzyme requiring ATP hydrolysis and electron supply from Ferredoxin. As same as nitrogenase which catalyzes reduction of nitrogen, DPOR consists of the electron transfer component, L-protein and the catalytic component, NB-protein.

We show a crystal structure of the catalytic component, NBprotein, of the DPOR from Rhodobacter capsulatus at 2.3angstrom resolution. Overall structure with two copies of homologous BchN and BchB subunits is similar to that of nitrogenase MoFe-protein. Each catalytic BchN-BchB unit contains one Pchlide held without any axial ligations from amino acid residues and one [4Fe-4S] cluster (NB-cluster) at the subunit interface. A surprise of the structure is direct coordination of BchB-Asp36 to the cluster, instead of BchB-Cys95 anticipated to coordinate the cluster based on the sequence similarity. The orientation of bound Pchlide is mainly provided by hydrophobic interaction, keeping the reduction site of Pchlide away from the NB-cluster. The structure in the presence or absence of Pchlide has revealed the displacement of C-terminal helix of BchB when accommodating Pchlide. Intriguingly, NB-cluster and Pchlide are arranged spatially as almost identical to P-cluster and FeMo-cofactor in MoFe-protein of nitrogenase, illustrating a common architectire to reduce chemically stable multi-bonds such as porphyrin and dinitrogen[1].

[1] Muraki, N., Nomata, J., Ebata, K., Mizoguchi, T., Shiba, T., Tamiaki, H., Kurisu, G. and Fujita, Y., *Nature*, 2010, *in press*

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