

Cryogenic X-ray Crystallography of Light-Harvesting

Complex of Photo System II (LHC-II)

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In photosynthesis of green plants, the solar energy captured by the light-harvesting chlorophyll a/b-protein complex (LHC-II) is transferred efficiently to the reaction centres of photosystems. LHC-II is the most abundant membrane protein in chloroplasts, accounting for half of the chlorophyll pigments involved in plant photosynthesis. Each polypeptide consists of 232 amino acids and binds >12 chlorophylls and 2 carotenoids (xanthophylls). The three-dimensional structure of LHC-II has recently been determined at 3.4 Å resolution by electron crystallography of two dimensional crystals.

In order to determine the protein structure at a higher resolution, we have developed a crystallization method by which an well-ordered three-dimensional crystal of LHC-II is produced. In this study, the chlorophyll-protein complex purified from pea chloroplast (Burke, J.J. 1978 Arch. Biochem. Biophys.) was crystallized according to the method

of Kühlbrandt with slight modification. When a mixture of LHC-II, nonylglucoside, KCl and sucrose was concentrated by vapor diffusion at room temperature, a hexagonal crystal (0.1 × 0.1 × 0.01) grew in several days. A single crystal was picked up with a nylon wire and rapidly cooled by liquid propane.

Diffraction data were collected with a weissenberg camera at BL41XU beamline. The wavelength was 0.708 Å. The crystal-to-camera distance was 500mm, and the coupling constant was 0.5mm/deg. The cryo-stream cooler was operated at 100K.

At 100 K, radiation damage was not significant, and diffraction spots up to 3 Å resolution were observed. The unit cell dimensions were determined as follows: $a = b = 128 \text{ \AA}$, $c = 146 \text{ \AA}$, $\alpha = \beta = 90^\circ$ and $\gamma = 120^\circ$. Unfortunately the crystal used exhibited a significant mosaicity in the direction parallel to the crystal c axis, making it difficult to determine the space group unambiguously.