Available online at www.sciencedirect.com
ScienceDirect
Plant Physiology and Biochemistry 46 (2008) 238–264
Plant Physiology and Biochemistry
www.elsevier.com/locate/plaphy

Review

Crystal structure of cyanobacterial photosystem II at 3.0 Å resolution: A closer look at the antenna system and the small membrane-intrinsic subunits

Frank Müh [a,b], Thomas Renger [a], Athina Zouni [b,*]

[a] Institut für Chemie und Biochemie/Kristallographie, Freie Universität Berlin, Takustrasse 6, D-14195 Berlin, Germany
[b] Institut für Chemie/Max-Volmer-Laboratorium für Biophysikalische Chemie, Technische Universität Berlin, Strasse des 17. Juni 135, D-10623 Berlin, Germany

Received 29 November 2007
Available online 12 January 2008

Abstract

Photosystem II (PSII) is a homodimeric protein-cofactor complex embedded in the thylakoid membrane that catalyses light-driven charge separation accompanied by the water splitting reaction during oxygenic photosynthesis. In the first part of this review, we describe the current state of the crystal structure at 3.0 Å resolution of cyanobacterial PSII from *Thermosynechococcus elongatus* [B. Loll et al., Towards complete cofactor arrangement in the 3.0 Å resolution structure of photosystem II, Nature 438 (2005) 1040–1044] with emphasis on the core antenna subunits CP43 and CP47 and the small membrane-intrinsic subunits. The second part describes first the general theory of optical spectra and excitation energy transfer and how the parameters of the theory can be obtained from the structural data. Next, structure–function relationships are discussed that were identified from stationary and time-resolved experiments and simulations of optical spectra and energy transfer processes.

Keywords: Antenna protein; Chlorophyll; Crystal structure; Excitation energy transfer; Optical spectra; Photosystem II; Small subunits

1. Introduction

Solar energy is harnessed by different types of organisms in a process called photosynthesis. The variant carried out by cyanobacteria, some algae (Protista) and higher plants (Plantae) is called oxygenic photosynthesis, since it uses water as the ultimate source of reducing equivalents, thereby producing molecular oxygen. The key reactions, in which light energy is transformed into chemical energy, take place in two multimeric pigment-protein complexes (PPCs) called photosystem I (PSI) and photosystem II (PSII). These photosystems are part of a complex machinery associated with the thylakoid membrane [1].

From the enzymatic point of view, PSII is a membrane-embedded water:plastoquinone oxidoreductase [2,3]. It contains the site of water-cleavage and utilizes the electrons extracted from water to reduce plastoquinone. The formed plastoquinole diffuses through the thylakoid membrane (quinone pool) until it finds its way to the cytochrome-b_6f-complex, another membrane protein [4]. Here, plastoquinole is re-oxidized and the electrons are transferred to a water-soluble electron carrier (plastocyanin or cytochrome c_6). This carrier in turn is oxidized by PSI, a membrane-standing plastocyanin:ferredoxin oxidoreductase [5], that delivers the electrons via ferredoxin

Abbreviations: AAR, amino acid residue; βCar, β-carotene; CD, circular dichroism; Chl*a*, chlorophyll *a*; cyt, cytochrome; DGDG, digalactosyldiacylglycerol; EDA, extended dipole approximation; EET, excitation energy transfer; ERPE, excited-state radical-pair equilibrium; ET, electron transfer; ETC, electron transport chain; FMO, Fenna–Matthews–Olson; FPU, fixed photosynthetic unit; LD, linear dichroism; LHC, light-harvesting complex; MGDG, monogalactosyldiacylglycerol; PG, phosphatidylglycerol; Pheo*a*, pheophytin *a*; PPC, pigment–protein complex; PSIcc, photosystem I core complex; PSIIcc, photosystem II core complex; RC, reaction centre; SQDG, sulfoquinovosyldiacylglycerol; TDC, transition density cube; TMA, transition monopole approximation; TMH, transmembrane α-helix; TrEsp, transition charges from electrostatic potentials; TTT, transfer to the trap; VPU, variable photosynthetic unit.

* Corresponding author. Tel.: +49 (0)30 3142 5650; fax: +49 (0)30 3142 1122.
E-mail address: athina.zouni@tu-berlin.de (A. Zouni).

0981-9428/$ - see front matter © 2008 Elsevier Masson SAS. All rights reserved.
doi:10.1016/j.plaphy.2008.01.003

to the enzymes producing NADPH. During the action of these enzymes, a proton gradient across the thylakoid membrane, termed proton motive force, is build up that is utilized in ATP synthesis [6].

During the last six years, significant progress has been made in elucidating the spatial structures of membrane proteins involved in oxygenic photosynthesis. Besides the structures of PSI of cyanobacteria [7] and higher plants [8,9], the cytochrome-b_6f-complex of cyanobacteria [10] and algae [11], and the major light-harvesting complex LHC-II of higher plants [12,13], a number of medium resolution crystallographic models of cyanobacterial PSII appeared, ranging from 3.8 to 3.2 Å [14–18]. Recently, the crystal structure of PSII from *Thermosynechococcus elongatus* could be improved to 3.0 Å resolution [19]. The latter structural model revealed many novelties concerning the cofactor inventory and uncovered misinterpretations of previous models at lower resolution. Therefore, the present review is based exclusively on this most recent crystal structure (pdb entry 2AXT).

Extensive biochemical characterization of the crystallizing material [20–22] indicated that the isolated, active PSII occurs as homodimers of core complexes with each monomeric part being composed of about 20 different protein subunits. On the basis of the 3.0 Å resolution structural model, 77 cofactors per monomer could be identified, including 35 chlorophyll *a* (Chl*a*), two pheophytin *a* (Pheo*a*), two plastoquinone, 11 β-carotene, 14 lipids and three detergent molecules [23], two haems, a non-haem iron, a calcium ion, one bicarbonate anion as well as the metal ions of the Mn_4Ca-cluster [24,25]. In addition, the total number of detergent molecules forming a belt around the PSII dimer in aqueous solution was estimated to ~350 [26].

A common feature of oxygenic and anoxygenic photosynthesis (where the latter is carried out by certain bacteria [27]) is that the chlorophyll (or bacteriochlorophyll) pigments are involved in two distinct types of processes: Excitation energy transfer (EET) and electron transfer (ET). Accordingly, the PPCs can be grouped into two classes: The reaction centres (RCs), which harbour only a small number of specialized pigments performing ET, and the light-harvesting complexes (LHCs) or antenna proteins [28,29], which contain the vast majority of photosynthetic pigments and deliver the energy of absorbed photons to the RC by performing EET. In this way, the LHCs effectively increase the absorption cross section of the RCs. The activity of the LHCs has to be adapted to supply and demand of photons. In this context, supply means the actual light conditions experienced by the organism, while demand refers to the need for NADPH and ATP reflected by, e.g., the redox state of the quinone pool and the proton motive force [30,31].

There are several ways of regulating light-harvesting activity. One possibility is non-photochemical quenching. In this process, the efficiency of EET from the antenna to the RCs is influenced by changing the rate of competing non-radiative transitions that lead to a depopulation of excited states in the antenna [32,33]. The detailed mechanism of non-photochemical quenching is not yet understood and an active field of research. Another possibility is to vary the amount of LHCs relative to RCs by means of changes in gene expression [34–36] and/or proteolysis [37]. In addition, LHCs may be shifted from one photosystem to the other in a process termed state transition [38,39]. Finally, one also has to consider the influence of photosystem stoichiometry, i.e. the relative amount of PSI and PSII, on the quantum efficiency of light-energy conversion [39,40]. It appears to be a common motif of photosynthetic organisms that there is always a minimal amount of antenna pigments associated with the RC in a fixed stoichiometry, forming what is called a fixed photosynthetic unit (FPU). The FPU-pigments are supplemented by those of separate antenna complexes, the amount of which is subject to regulation as described above. Altogether, therefore, the pigments form a variable photosynthetic unit (VPU) [41,42].

PSI and PSII could be considered as the FPUs of oxygenic photosynthesis, but this terminus is problematic in view of the variable PSI/PSII ratio [39,40]. Therefore, the photosystems PSI and PSII are referred to as *core complexes* (PSIcc and PSIIcc). Besides the RC architecture (type I or iron–sulphur type in PSI and type II or pheophytin-quinone type in PSII; for definition, see [43,44]), the two photosystems differ considerably in the size of the core antenna. Whereas there are about 100 Chl*a* pigments per RC in PSI (96 in the case of *T. elongatus* [7]), only about one third of this number is bound to PSII (35 in the case of *T. elongatus* [19]). Another important difference is the separability of the RC and the antenna part. The RC and antenna pigments are bound to different protein subunits in PSII (see below), so that they can (at least in principle) be separated from each other without denaturation or cutting of polypeptide chains [45,46]. This is not the case for PSI, where the two large subunits PsaA and PsaB bind both the RC and most of the antenna pigments [7].

In the first part of this review, we describe the structure of PSIIcc as it appears to us from X-ray crystallography at 3.0 Å resolution [19]. The focus will be on the two subunits PsbB (CP47) and PsbC (CP43) constituting the core antenna and the small intrinsic subunits connected to it. We believe that due to the high homology of cyanobacterial and plant PSIIcc [47,48], the data concerning *T. elongatus* to be presented here are of relevance to PSII in general.

Knowing the spatial arrangement of its components is essential for an understanding of a PPC, but the relationship between structure and spectroscopy cannot be established without structure-based calculations [29,49–55]. In the second part of the review, we shall, therefore, first explain basic principles of the theory in a simple way and then describe how the optical absorption spectra and EET processes of PSIIcc can be understood on the basis of the structural data.

2. Description of the structure

2.1. Overview

The crystals underlying the 3.0 Å resolution structural model are obtained from a fraction of the PSII preparation from *T. elongatus* containing dimeric PSIIcc [20–22]. Fig. 1 shows