

**THE PSBH PROTEIN IS BOUND TO CP47 PRIOR TO ITS  
ATTACHMENT TO THE D1-D2 HETERODIMER DURING ASSEMBLY  
OF PHOTOSYSTEM II COMPLEX IN THE CYANOBACTERIUM  
*Synechocystis* PCC 6803**

**JOSEF KOMENDA<sup>1,2</sup>, JULIE BENEŠOVÁ<sup>1,2</sup> and MARTIN TICHÝ<sup>1,2</sup>**

<sup>1</sup>Laboratory of Photosynthesis, Institute of Microbiology, Opatovický Mlýn, Třeboň, Czech Republic, <sup>2</sup>Institute of Physical Biology, University of South Bohemia, Zámek 136, Nové Hrady, Czech Republic

The PsbH protein, a small Photosystem II (PSII) subunit, has been previously identified in the cyanobacterium *Synechocystis* PCC 6803 as a component of the PSII core complex (RCC), the core complex lacking CP43 (RC47) but not the RC complex consisting of D1, D2 and cytochrome b-559 [1]. To follow the insertion of the protein into PSII during the assembly process, we constructed a mutant in which the original psbH gene was deleted and a modified psbH gene with the 6 x His tag at the N-terminus was inserted under the psbA2 promoter. Phenotype of this mutant was indistinguishable from the wild type indicating presence of fully active PSII complexes. Analysis of the PSII assembly in the mutant using blue native electrophoresis in combination with radioactive labeling and Western blot confirmed that the protein is incorporated into PSII during the step when CP47 is attached to the RC complex forming the RC47 complex. However, the labeled PsbH protein was found not only RCC and RC47 complex but also in the smaller complex corresponding to the unassembled CP47. In the double mutant unable to assemble RC complex due to deletion of the psbEFLJ operon, the PsbH protein has been found nearly exclusively in the form bound to unassembled CP47. Close structural relationship between PsbH and CP47 was confirmed by parallel variability of level of both proteins in various CP47 mutants. In the mutant completely lacking CP47 only negligible amount of the PsbH protein in a free form was found. The results showed that during the PSII assembly, the PsbH protein is associated with CP47 prior to its attachment to the D1-D2 heterodimer.

#### REFERENCE

1. Komenda, J., Lupinkova, L. and Kopecky, J. Absence of the *psbH* gene product destabilizes the Photosystem II complex and bicarbonate binding on its acceptor side in *Synechocystis* PCC 6803 **Eur. J. Biochem.** 269 (2002) 610-619.