Supplementary figures and tables



Figure S1: Level of the unassembled PsbI protein is controlled by the protease FtsH (slr0228). Thylakoid proteins from cells of wild-type (WT), mutant lacking FtsH (slr0228) protease (Δ FtsH), D1-less mutant (Δ D1) and the double mutant lacking both D1 and FtsH (Δ D1/ Δ FtsH) were separated by denaturing SDS-PAGE and electroblotted onto PVDF membrane. Proteins were detected using antibodies specific for D2, PsbI and CP43 proteins from *Synechocystis* sp. PCC 6803. Correct protein loading was proven by blotting using antibody against CP43. Level of CP43 is not affected by FtsH (slr0228) (Komenda et al. 2006) and can be used as an internal standard for protein loading. 4, 2, 1 and 0.5 µg of chlorophyll were loaded onto the gel.



Figure S2: PsbI does not affect assembly of the PSII RC47 complex in the CP43-less strain. Thylakoid

proteins from cells of *psbC* deletion mutant Δ CP43 and *psbC/psbI* double mutant Δ CP43/ Δ PsbI were separated by 2D-BN/SDS-PAGE, stained by Coomassie Blue (Gel stain) and radioactive proteins visualized by autoradiography (autoradiogram).



Figure S3: Effect of PsbI absence on levels of the D1 protein in the wild-type, *psbB* (Δ CP47) and *psbEFLJ* (Δ CYT) deletion mutants. Thylakoid proteins from designated strains were separated by denaturing SDS-PAGE, electroblotted onto PVDF membrane and immunodecorated using antibodies specific for D1 (anti-D1) and PsbI (anti-PsbI). Correct protein loading was checked by Poinceau staining and documented by the staining intensity of the α and β subunits of ATP synthase ($\alpha\beta$ ATPsynth stain). 2, 1 and 0.5 µg of chlorophyll was loaded onto the gel for each strain.

10010011	Table S1.	Characteristics	of the wild	d-type strain	WT and	the mutant	ΔPsbI
----------	-----------	-----------------	-------------	---------------	--------	------------	-------

	Doubling	O ₂ evolution ^b	O ₂ evolution ^c	Fv/Fm ^d	
	time ^a	$(\mu mol O_2 mg Chl^{-1} h^{-1})$	$(BQ + K_3Fe(CN)_6)$	(PAM)	
	(h)		$(\mu mol O_2 mg Chl^{-1} h^{-1})$		
WT	$15,2 \pm 0,1$	372 ± 27	562 ± 32	0,47±0.02	
		100%	100%	100%	
ΔPsbI	$16,4 \pm 0,1$	258 ± 13	343 ± 30	0,32±0.02	
		69%	61%	68%	

^ameasured in microtitration plates at an irradiance of 25 μ mol photons m⁻² s⁻¹; average of 11 measurements ± SD.

^blight-saturated rate of oxygen evolution measured in the absence of artificial electron acceptors; average of 6 measurements \pm SD.

^clight-saturated rate of oxygen evolution measured in the presence of 1 mM p–benzoquinone and 5 mM potassium ferricyanide; average of 10 measurements \pm SD.

^dvalues obtained by PAM modulated fluorimeter in the presence of 10^{-5} M DCMU; average of 3 measurements ± SD.

	RCC(2)	RCC-	RC47(2)	RCC(1)	RC47	RCa	U.P.
		RC47					
WT pulse	6250	n.d.	n.d.	8552	773	n.d.	n.d.
stain ^a	(41%)			(54%)	(5%)		
WT pulse	46197	n.d.	n.d.	294830	158952	410564	1154399
autorad ^b	(2%)			(15%)	(8%)	(20%)	(55%)
WT pulse-	322479	n.d.	n.d.	1033171	154853		82759
chase autorad ^b	(20%)			(65%)	(10%)	n.d.	(5%)
ΔPsbI	2355	2490	680	3591	3050	n.d.	n.d.
stain ^a	(20%)	(19%)	(6%)	(30%)	(25%)		
∆PsbI pulse	208971	214816	94332	508849	1637857	n.d.	n.d.
$autorad^{b}$	(8%)	(8%)	(4%)	(19%)	(61%)		
ΔPsbI pulse-	312289	318470	142945	452931	461994	n.d.	n.d.
chase autorad ^b	(19%)	(19%)	(8%)	(27%)	(27%)		

Table S2. Distribution of the stained and labeled D1 protein among PSII complexes after pulse and pulse-chase labeling of WT and the Δ PsbI mutant

^aquantification of stained D1 bands by ImageQuant software in 2D gels stained with Coomassie Blue, average of three measurements, SD did not exceed 6%. Identical amount of Chl (6 μ g) was loaded for each sample; samples of each strain after pulse and after pulsechase were run on a single 2D gel, staining pattern of both samples were identical and only quantification of the samples after pulse is shown. Values indicate arbitrary counts, values in parenthesis show percentage of the overall D1 level in the sample. Designation of complexes as in Fig. 2, n.d. means not detected.

^bquantification of labeled D1 bands by ImageQuant software from 2D autoradiogram obtained by exposure to Phosphoimager plate, average of three measurements, SD did not exceed 5%. Identical amount of Chl ($6 \mu g$) was loaded for each sample; samples from particular strain after pulse and after pulse-chase were run on a single 2D gel and were directly compared. Values indicate arbitrary counts, values in parenthesis show percentage of the overall level of labeled D1 in the sample. Designation of complexes as in Fig. 2, n.d. means not detected.