Fig. S1

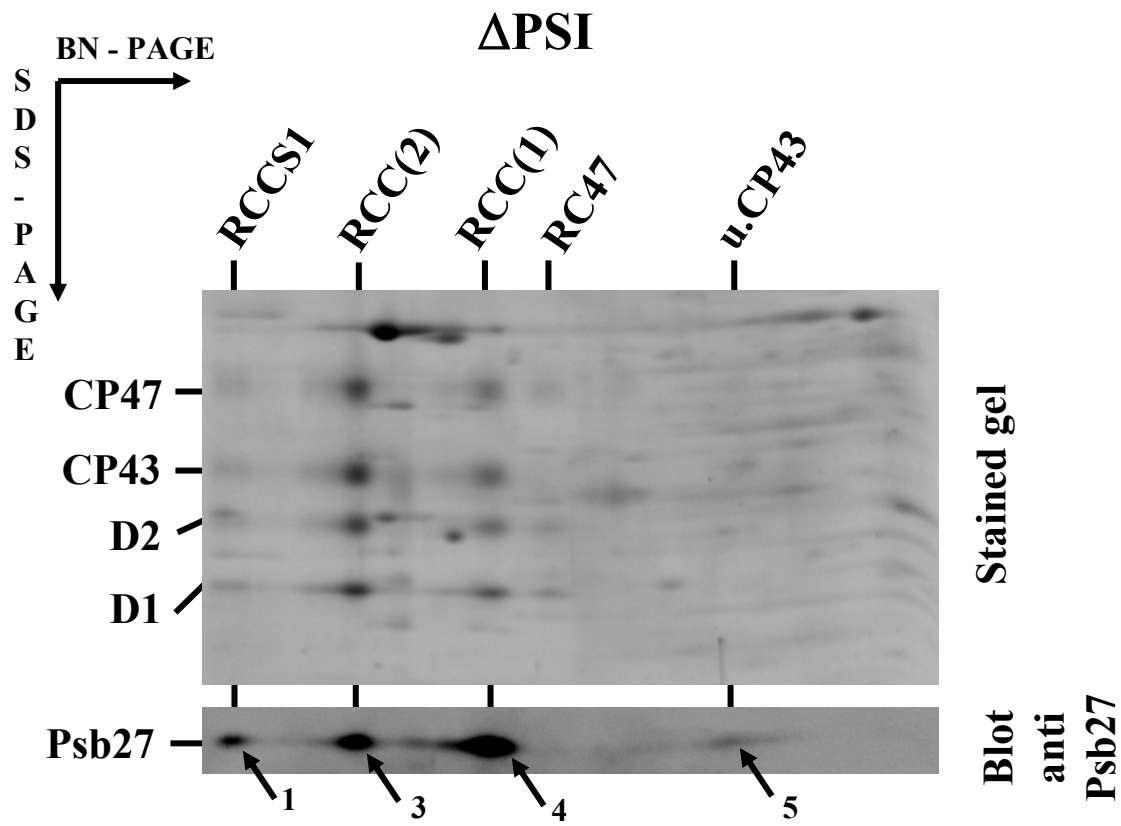

Fig. S1. Localization of the Psb27 protein by 2D-BN/SDS-PAGE in membranes of the *Synechocystis* PSI-less strain cultivated under 5 µmol photons m^{-2} s^{-1}. Membrane proteins were separated by 2D electrophoresis, the gel was either stained by Sypro Orange (Stained gel) or blotted to a PVDF membrane and Psb27 was detected by the specific antibody. Designation of complexes as in Fig. 1.

Fig. S2

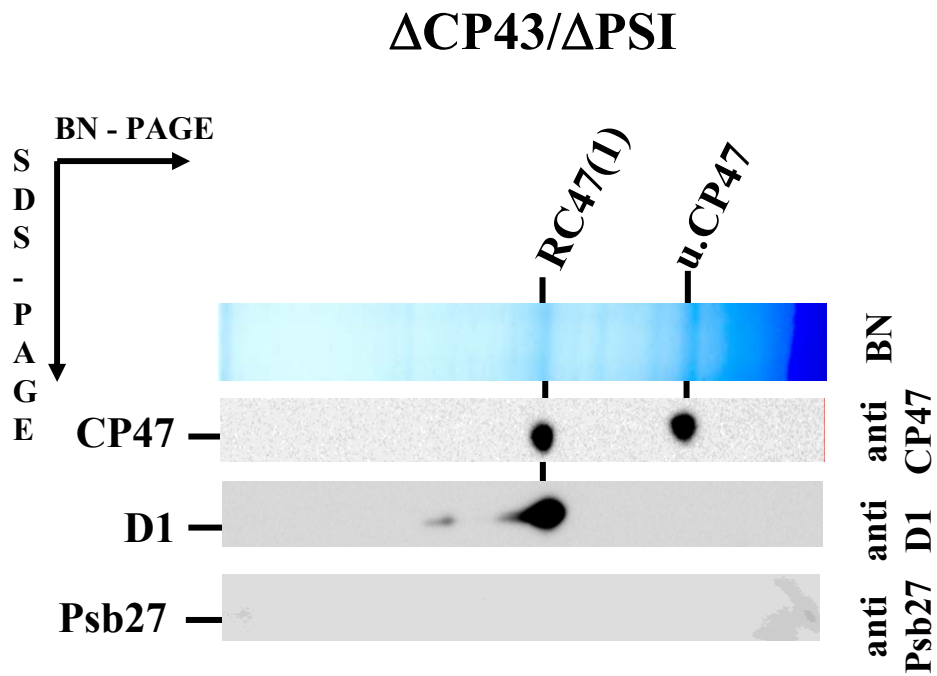

Fig. S2. Localization of the Psb27 protein by 2D-BN/SDS-PAGE in membranes of the *Synechocystis* strain lacking CP43 and PSI. Membrane proteins were separated by 2D electrophoresis, the gel was blotted to a PVDF membrane and CP47, D1 and Psb27 were detected by specific antibodies. Designation of complexes as in Fig.1

Fig. S3

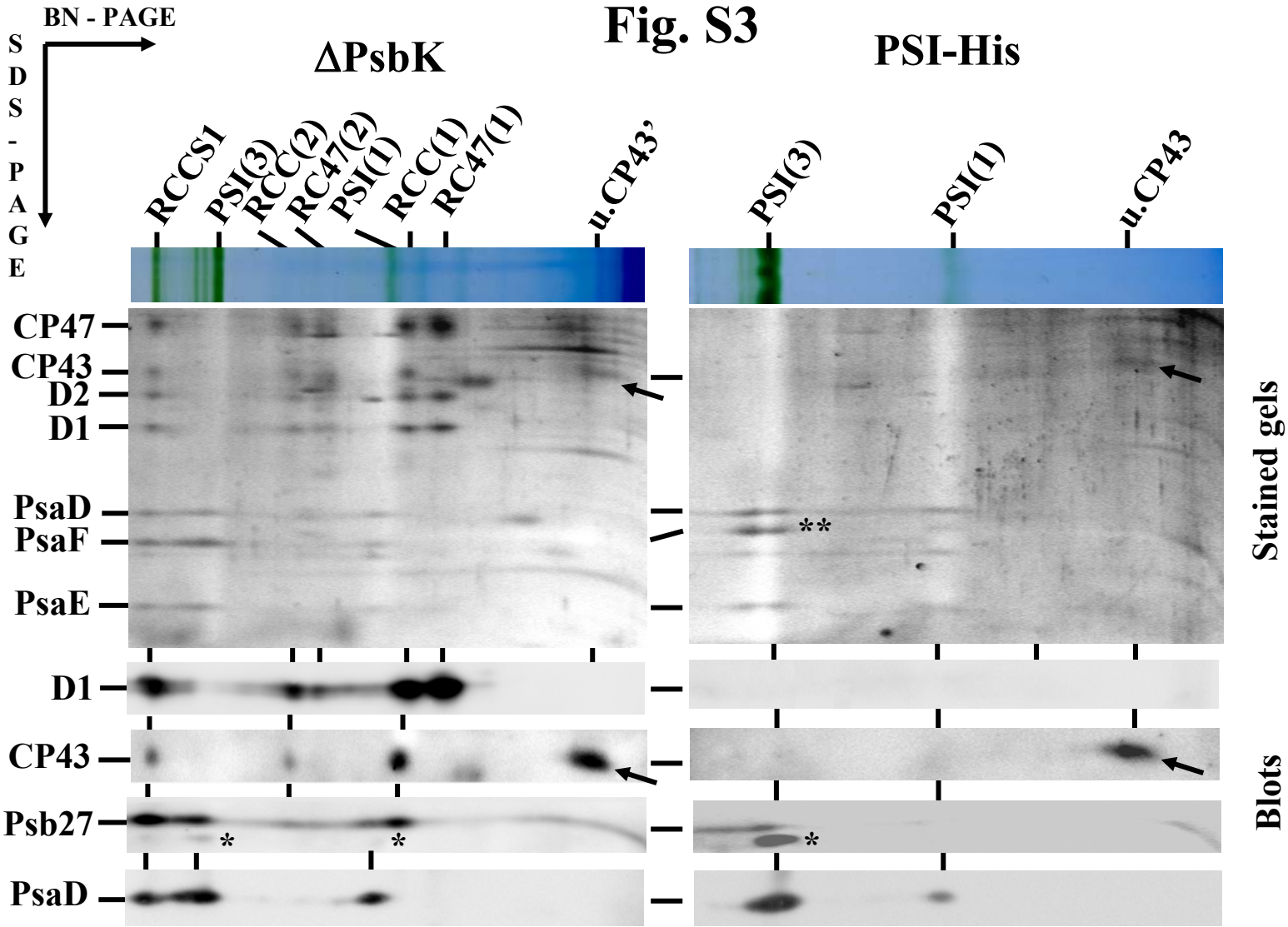

Fig. S3. Localization of the Psb27 protein by 2D-BN/SDS-PAGE in membranes of the *Synechocystis* psbK deletion mutant ΔPsbK (left panels) and PSI-His complex isolated by a single step Ni affinity chromatography (PSI-His, right panel). Membrane proteins were separated by 2D electrophoresis, the gel was either stained by Sypro Orange (Stained gel) or blotted to a PVDF membrane (Blot) and D1, CP43, Psb27 and PsaD were detected by specific antibodies. The identity of other designated proteins on the stained gel was verified by MS. Designation of complexes as in Figs.1. and 2.; oblique arrows, unassembled CP43; asterisk, cross-reactions of the anti Psb27 antibody with PsaE; double asterisk, His-tagged PsaF in PSI-His (note lower mobility in comparison with PsaF in ΔPsbK).

Fig. S4

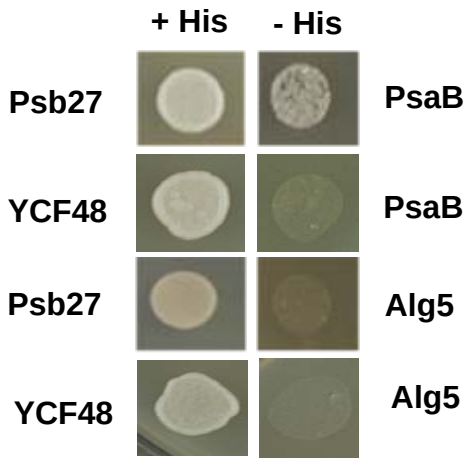

Fig. S4. Yeast Two-Hybdrid Analyses. The interaction between Psb27 and PsaB in the yeast strain DSY-1 was monitored by using the split ubiquitin system precisely as described (Dualsystems Biotech AG; Pasch et al. 2005). The psb27 coding region was PCR amplified from Synechocystis DNA, subcloned in the pDrive vector (Qiagen, Hilden) and control-sequenced. This construct lacks the N-terminal transit sequence of Psb27 comprising 24 amino acids to avoid localization problems of the NubG-Psb27 fusion protein in yeast. The protein in yeast is very likely not lipidated as the prokaryotic lipobox does not function in this organism. Subsequently, the construct was inserted into the pADSL vector via its oligonucleotide-derived BamHI and EcoRI restriction sites. The specificity of the construct was tested by co-transformation with the control-plasmid pMBV-Alg5 which encodes the Alg5 protein fused to the Cub fragment. Alg5 is an unrelated endoplasmic reticulum protein from yeast representing a dolichyl-phosphate glucosyltransferase (Pasch et al., 2005). The pTFB-1-psaB construct contains the entire psaB coding region inserted into the StuI restriction site of the vector pTFB-1 and was kindly provided by F. Ossenbühl. Interaction of the previously characterized lumenal PSII assembly factor YCF48 (Komenda et al., 2008) is also shown as negative control which exhibits no interaction with either PsaB or with Alg5 control protein. Yeast transformants expressing on one side either Psb27 or YCF48, and on the other side either PsaB or Alg5 fusion proteins were grown on non-selective medium (+His) and medium selecting for protein-protein interaction (-His).

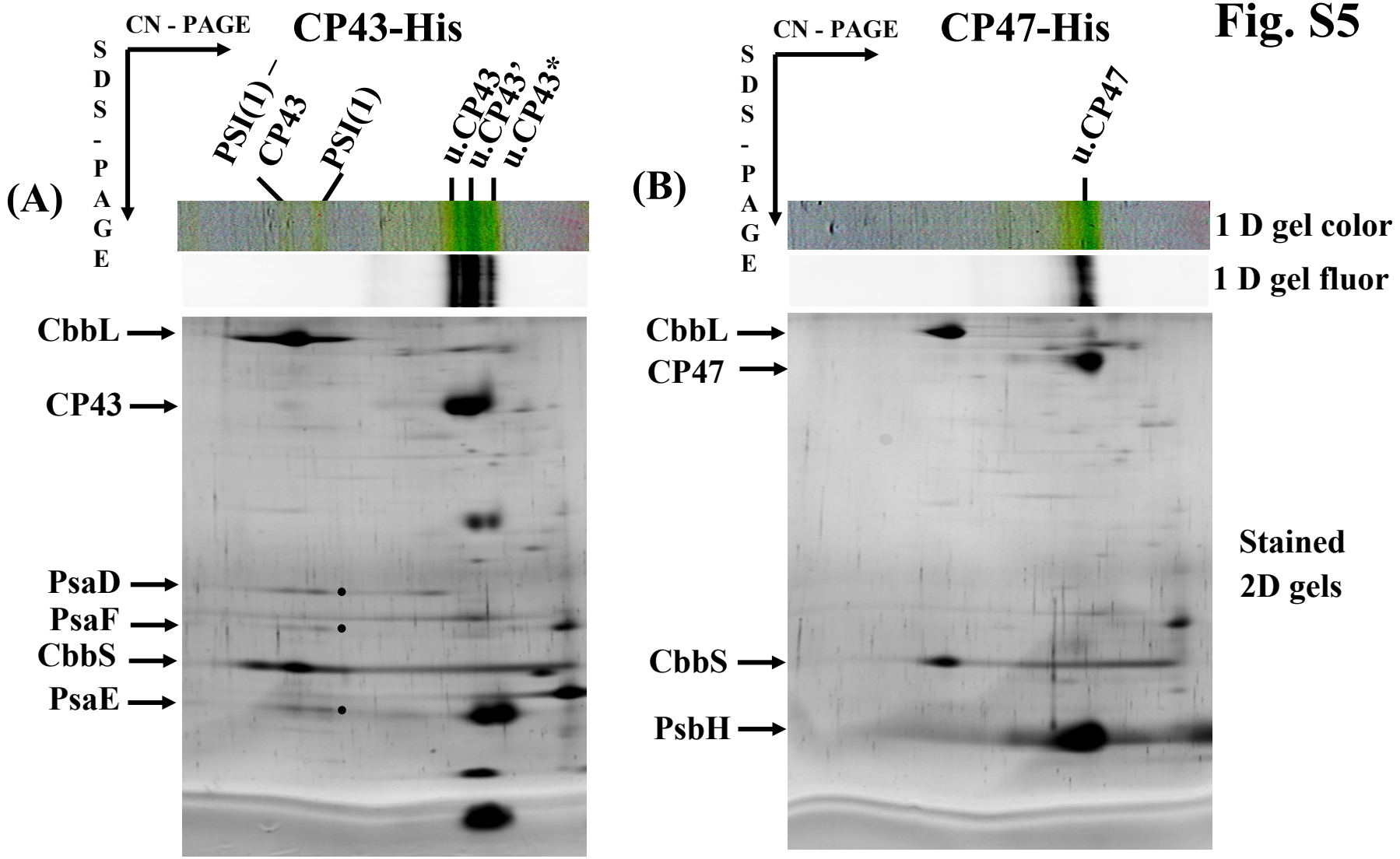

Figure S5. Separation of pigment-protein complexes from isolated preparations of His-tagged CP43 (A) and His-tagged CP47 (B) by 2D-CN/SDS-PAGE. The CP43-His and CP47His isolated by Ni-metal affinity chromatography were analyzed by CN PAGE, the native gel was photographed (1D gel color), scanned for fluorescence (1D fluor) by LAS 4000 and reelectrophoresed (stained 2D gel). 2D gels were stained by silver and identity of proteins was verified by sequencing. The identity of PSII-related spots as in Figs. 5 and 6, the small PSI subunits PsaD, PsaF and PsaE are designated by dots directly in the gel.

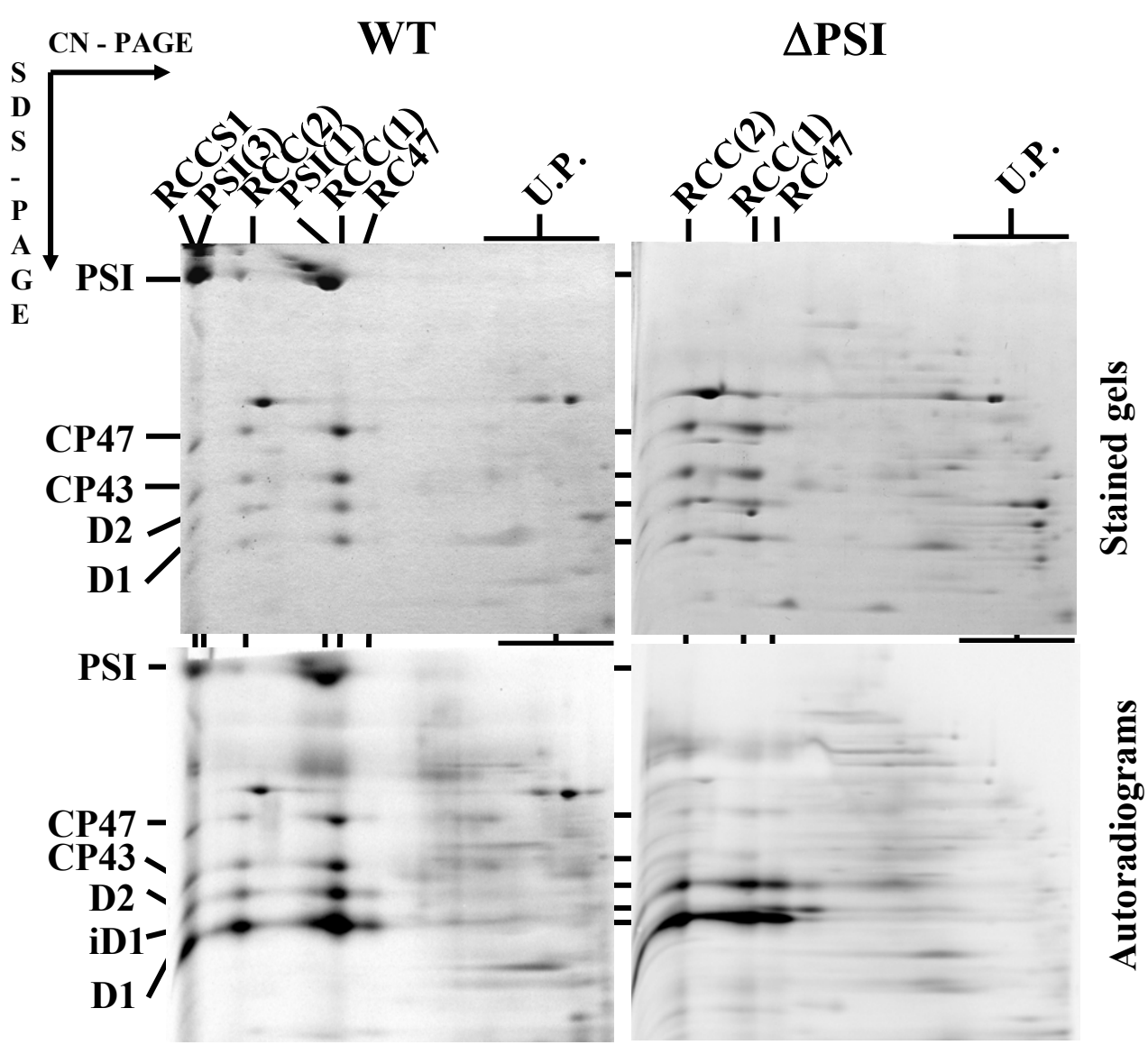

Fig. S6

Figure S6. 2D analysis of radioactively labeled membrane protein complexes of WT and the PSI-less strain ΔPSI. Membranes isolated from radioactively labeled cells of WT and ΔPSI cultivated under 5 µmol photons m^{-2} s^{-1} were analyzed by 2D CN/SDS-PAGE. The gels were stained (Stained gel) and exposed to Phosphorimager plate (Autoradiograms). Complexes are designated as in Fig. 1. 5 µg of Chl were loaded for membranes of WT and 1 µg of Chl for membranes of ΔPSI.

Fig. S7

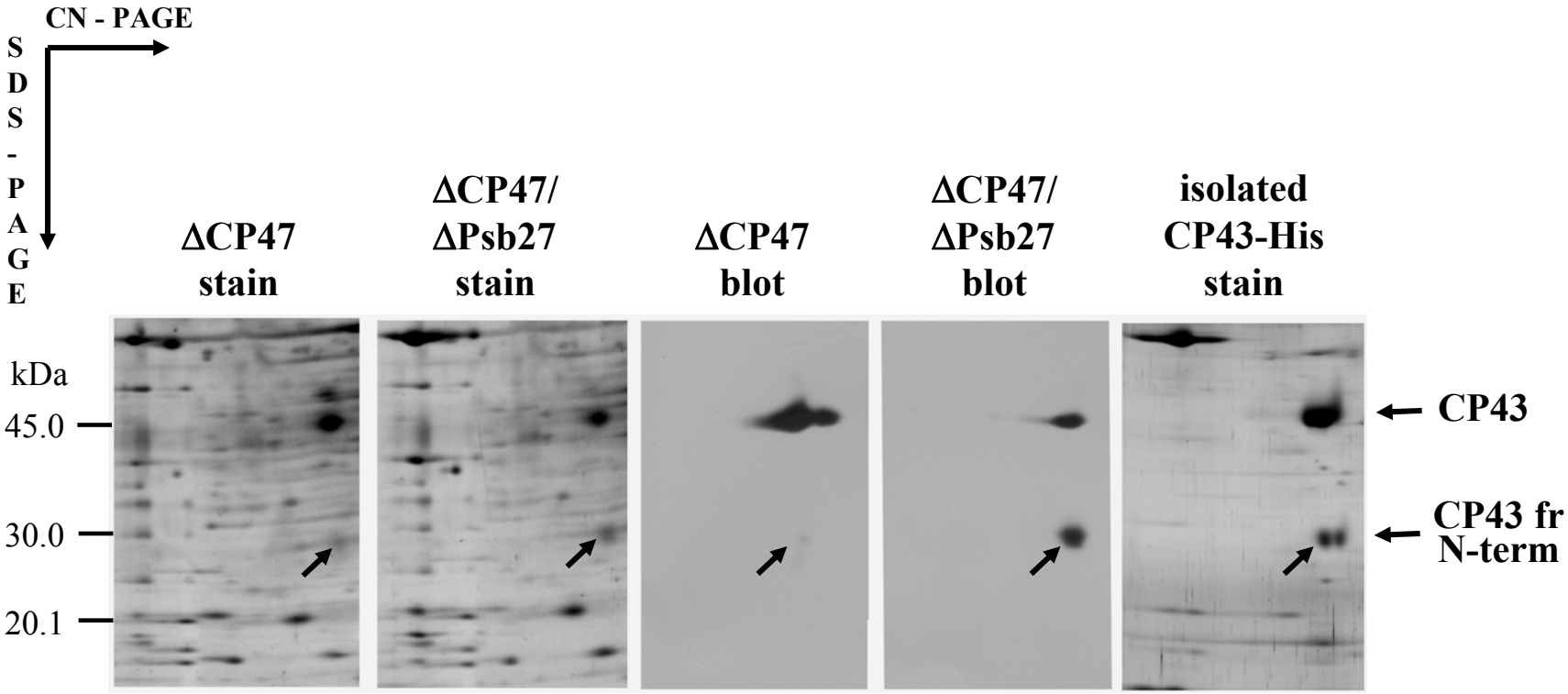

Fig. S7. Comparison of CP43 fragmentation in membranes of the ΔCP47 and ΔCP47/ΔPsb27 strains and in the preparation of isolated CP43-His. Membranes from both strains and CP43-His isolated from the ΔD1/CP43-His strain by Ni-metal affinity chromatography were analyzed by 2D-BN/SDS-PAGE and identity of the proteins was confirmed by CP43-specific antibodies. The N-terminal fragment is designated by an oblique arrow.

Fig. S8

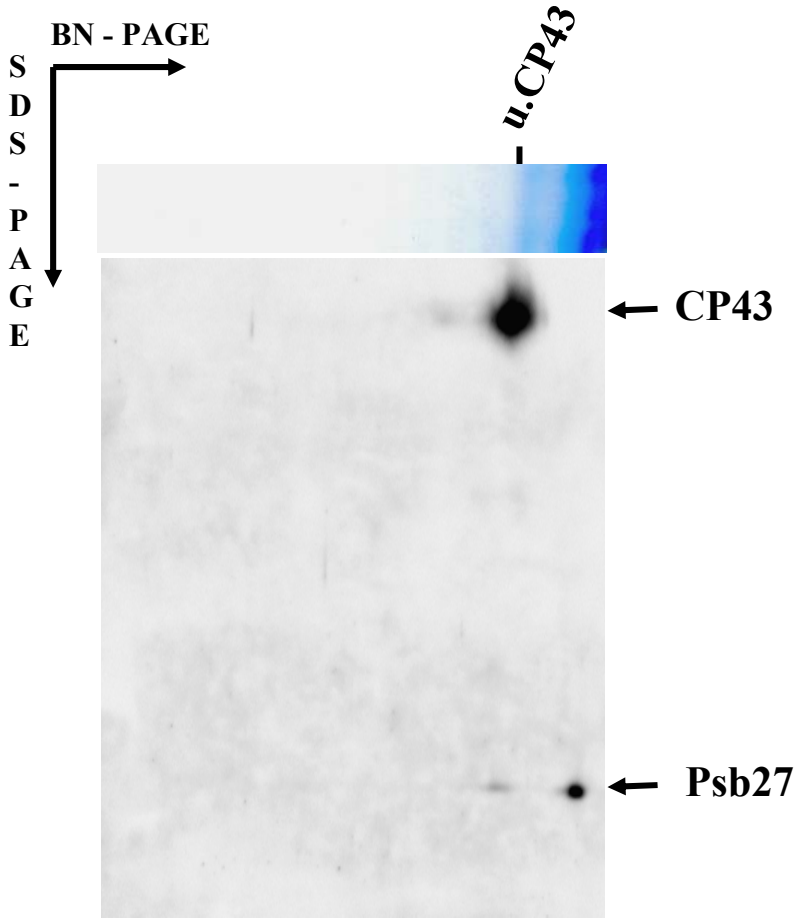

Fig. S8. Separation of His-tagged CP43 by 2D-BN/SDS-PAGE. The 6xHis-CP43 isolated from the 6xHis-CP43/ΔD1 strain by Ni-metal affinity chromatography was analyzed by 2D-BN/SDS-PAGE and separated proteins were identified by antibodies specific for CP43 and Psb27.

Fig. S9

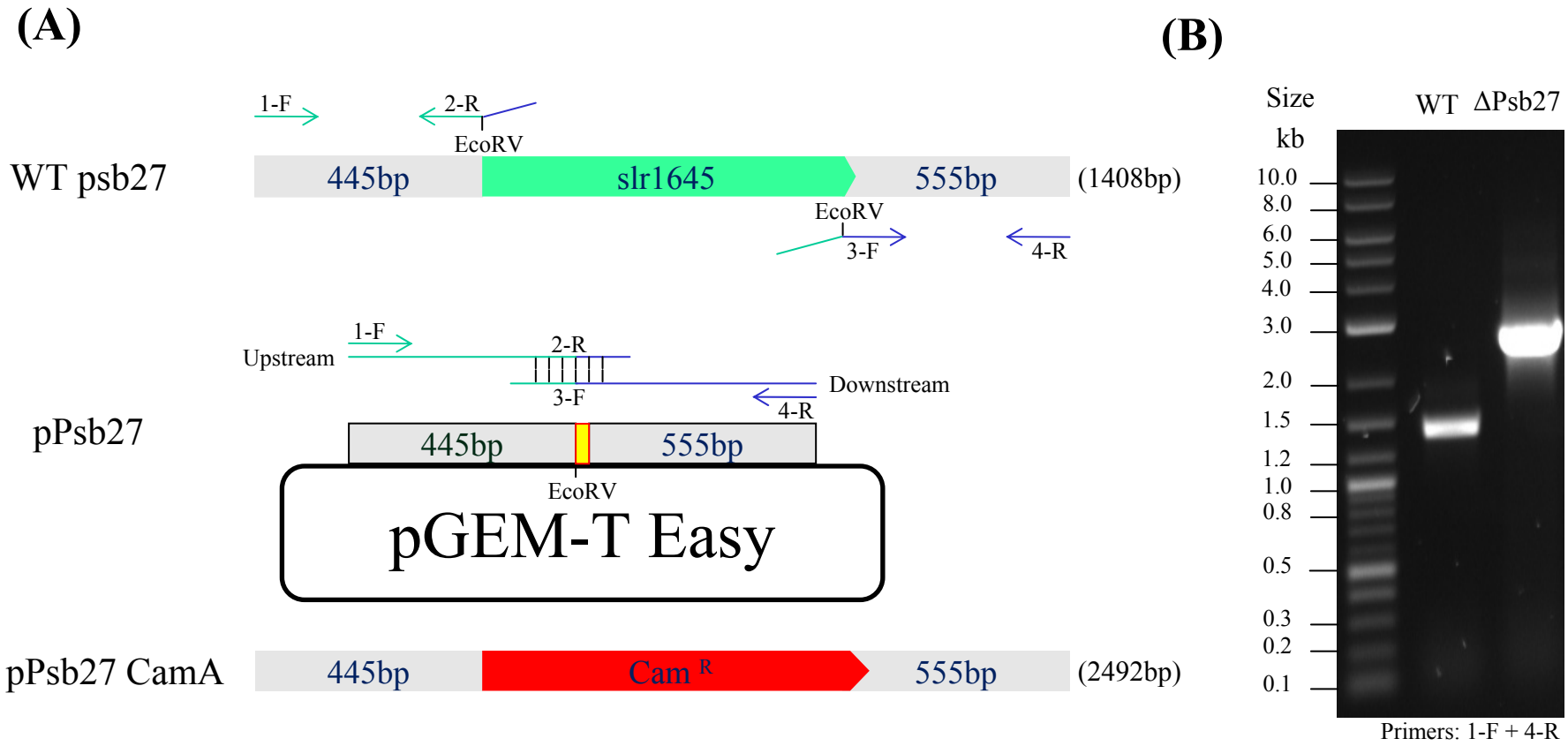

Fig. S9. Construction of the pPsb27 CamA vector for transformation of the *Synechocystis* 6803 WT-G strain (A) and confirmation of the complete segregation of the deletion mutant ΔPsb27 by PCR (B).

Fig. S10

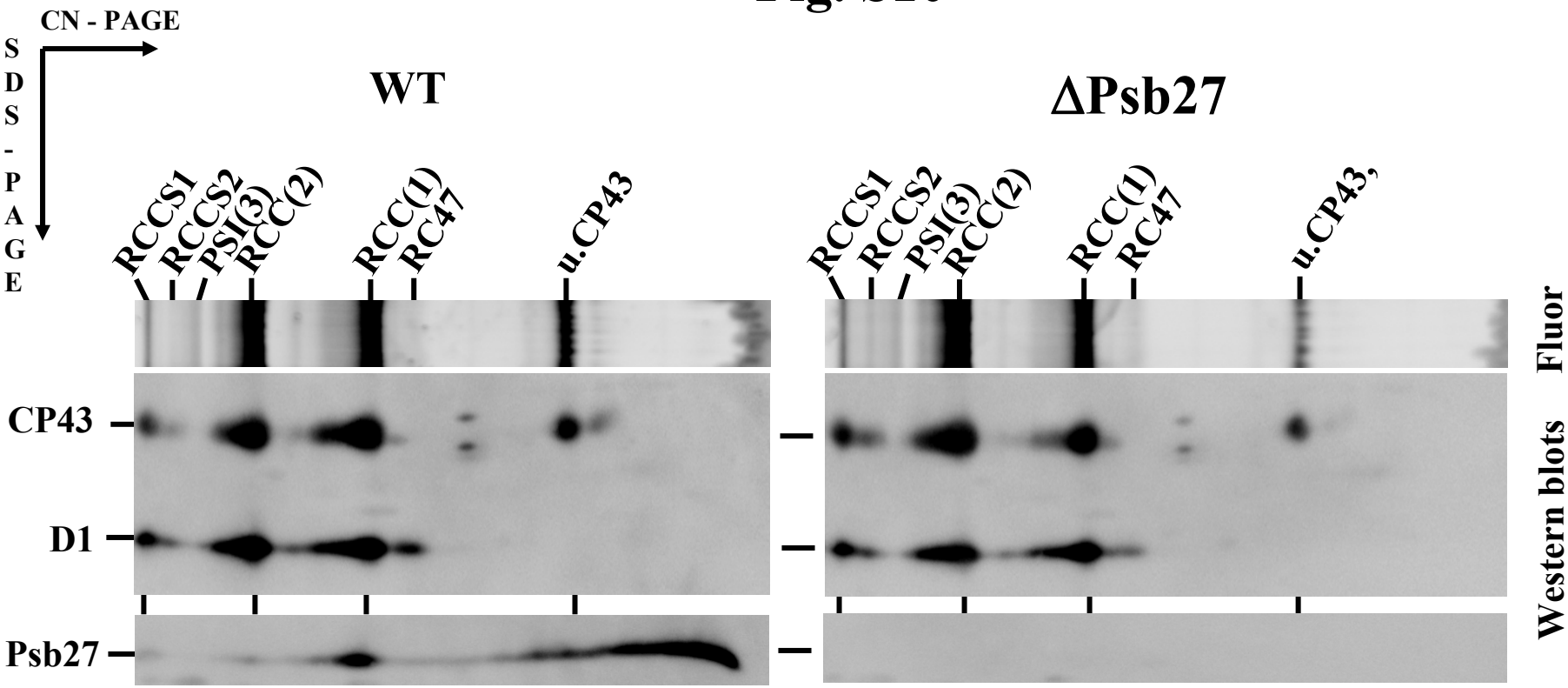

Fig. S10. Analysis of CP43, D1 and Psb27 distribution in complexes of WT and ΔPsb27 strains by 2D-CN/SDS-PAGE. Membrane complexes were separated by CN PAGE, the gel was scanned for Chl fluorescence by LAS4000 (Fluor) and subunits of complexes were separated by 2D electrophoresis, the gel was blotted to a PVDF membrane and CP43, D1 and Psb27 were detected by specific antibodies (Western blots). Designation of complexes as in Fig. 1.