Subunit and chlorophyll organization of the plant photosystem II supercomplex

Laura S. van Bezouwen¹, Stefano Caffarri², Ravindra S. Kale³, Roman Kouřil³, Andy-Mark W. H. Thunnissen¹, Gert T. Oostergetel¹ and Egbert J. Boekema¹*

¹Electron microscopy and Protein crystallography group, Groningen Biomolecular Sciences and Biotechnology Institute, University of Groningen, 9747 AG Groningen, The Netherlands. ²Aix Marseille Université, CEA, CNRS, BIAM, Laboratoire de Génétique et Biophysique des Plantes, 13009 Marseille, France. ³Centre of the Region Haná for Biotechnological and Agricultural Research, Department of Biophysics, Faculty of Science, Palacký University, Šlechtitelů 27, 783 71 Olomouc, Czech Republic. ¹Present address: Cryo-Electron Microscopy, Bijvoet Centre for Biomolecular Research, Faculty of Science, Utrecht University, 3584 CH Utrecht, Netherlands (L.S.v.B.). *e-mail: e.j.boekema@rug.nl
**Supplementary Figure 1 | Original EM images.** Images are sums of frames corrected for beam induced motion and drift. (a) Image taken at a defocus of 1.2 μm. (b) Image taken at a defocus of 3.0 μm. Some side views (white arrowheads) and top views (black arrowheads) are marked.

**Supplementary Figure 2 | Angular distribution of angles of cryo-EM projections with respect to the 3D model.** The length and the colour of the bars represent the population of different projection directions. The 3D model is seen from aside.
Supplementary Figure 3 | A gallery of 2D averages that reconstitute the final cryo-TEM map. 16 2D maps show the PSII supercomplex in top-view, side-views and intermediate views.

Supplementary Figure 4 | Local resolution of the cryo-EM density map. (a) Slice through the PSII supercomplex in the centre of the membrane plane to show the differences in local resolution. At the core region the resolution is at its best. The S-LHCII trimer and CP26 have the highest resolution for the antenna proteins. The M-LHCII trimer and the CP24 have the lowest resolution of the supercomplex. Details in the detergent shell were not resolved. (b) Resolution inside the protein. The extrinsic part is well resolved. The helices inside the membrane of the core have the highest resolution.
Supplementary figure 5 | FSC curve of the map vs model. The resolution is 7.7 Å, based on the 0.5 criterion. This is lower than the final resolution 5.3 Å in figure 1c, because there are large resolution differences within the map. In addition, there are areas left empty, where most likely pigments and lipids are present, due to limited resolution.

Supplementary Figure 6 | The region near the expected location of the manganese cluster in plants. Subunits PsbA (purple), PsbB (cyan), PsbD (blue) and PsbO (yellow) are shown and their expected coordination of the manganese cluster (manganese yellow, calcium green and waters in red). There is no density present for the manganese cluster. The C-terminus of PsbA is highly disordered, a slight disorder at the C-terminus of PsbD, and there is a disorder at the loop of PsbO. The regions of PsbA and PsbD interact with manganese cluster and the loop of PsbO interact with the C-terminus of PsbA in cyanobacteria. The density for these regions is missing in the cryo-EM map.
Supplementary Figure 7 | Fit of the antenna subunits inside the cryo-EM structure. (a) Fit of a Lhcb1 subunit of the S-trimer, (b) Fit of a Lhcb1 subunit of the M-trimer, (c) Fit of CP26, (d) fit of CP29 and (e) fit of CP24.

Supplementary Figure 8 | Comparison of the 3D cryo structure with a negative stain 2D projection map. (a) The 3D cryo-EM density map, showing all the main regions. Color coding is the same as in Fig. 2. (b) 2D projection map of the C2S2M2 particle20. The core is from cyanobacteria64 and the Lhcb subunits are from spinach12 for both trimeric LHCII (blue and green) and monomeric Lhcb (red, yellow and pink). The LHCII-S and LHCII-M trimer are contoured in light green. Note that the exact orientation of CP26 and CP29 is slightly different in the two models, while for the orientation of CP24 is significantly different. The scale bar is 100 Å.