

Supplementary Table 1 | Cryo-EM data collection and refinement.

	#1 Apo Rubisco (EMDB-22401) (PDB 7JN4) (EMPIAR-10503)	#2 EPYC1 ₄₉₋₇₂ peptide- bound Rubisco (EMDB-22308) (PDB 7JFO) (EMPIAR-10502)	#3 EPYC1 ₁₀₆₋₁₃₅ peptide- bound Rubisco (EMDB-22462) (PDB 7JSX) (EMPIAR-10501)
Voltage (kV)	300	300	300
Camera/detector	K2	K2	K3
Magnification	22,500	22,500	81,000
Pixel size (Å)	1.31	0.655	0.844
Defocus range (μm)	-1.5 to -3.0	-1.5 to -3.0	-1 to -1.6
Exposure time (s)	10	10	3.56
No. movie frames	50	50	60
Electron dose (e ⁻ /Å ²)	58	58	60
No. micrographs	1,834	2,500	13,727
No. initial particles	677,071	1,809,869	2,257,131
No. final particle	491,395	945,755	152,839
Symmetry	D4	D4	D4
Resolution (Å)	2.68	2.13	2.06
Map sharpening <i>B</i> factor (Å ²)	-100.04	-77.55	-47.75

Supplementary Table 2 | The amino acid residues that form the Rubisco-binding regions on EPYC1 homologs and the residues that form the EPYC1 binding site on the surface of Rubisco, appear to be conserved across the order Volvocales. Residues with roles in the binding interface are bolded. Residues that are different from the *Chlamydomonas reinhardtii* sequence are highlighted in grey.

Species	First Rubisco-binding region on EPYC1 homolog	Rubisco SSU helix A	Rubisco SSU helix B
<i>Chlamydomonas reinhardtii</i>	TRSVLPAN WRQELESLRN	DE QIAAQVDYIVA	PMQV LR EIVACTKA
<i>Tetrabaena socialis</i>	TRSVLPAN WRQELESLRG	DE QIAAQVDYIVA	PMQV LR EIV SCT RA
<i>Gonium pectorale</i>	TRSVLPAN WRQELESLRN	DE QIAAQVDYIVA	PMQV LR EIVACTKA
<i>Volvox carteri</i>	TRSVLPAN WRQELESLRN	DE QIAAQVDYIVA	PMQV LR EIVACTKA