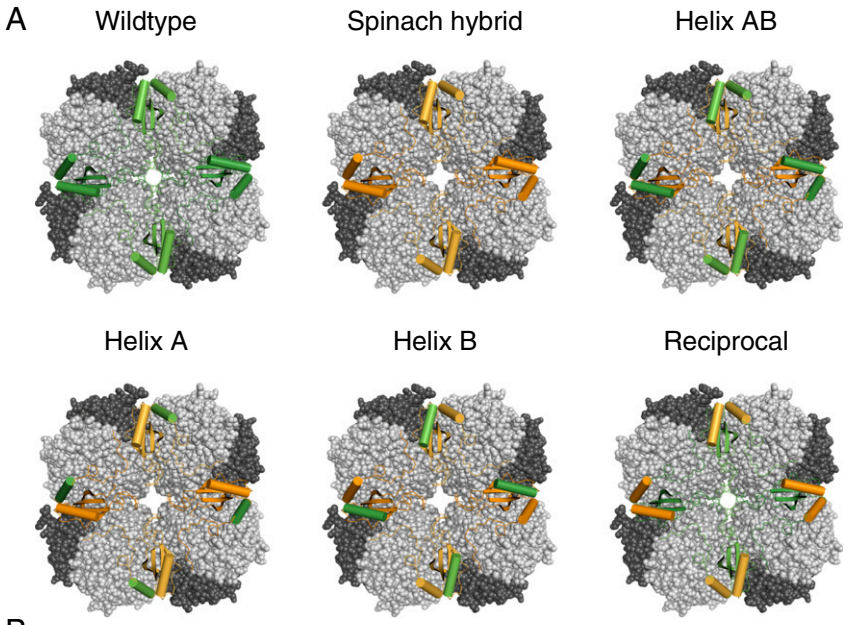
Supporting Information

Meyer et al. 10.1073/pnas.1210993109



B

Helix A *RbcS* sequence

ATGATGGTGTGGCCCACCCAGAACATGAAGCGCTACGAGACCCTGTCCTACCTGCCCCCCCTGACCGACGAGCAGATCGCCGC
CCAGGTCGACTACATCGTCGCCAACAAGTGGGTGCCCTGCCTGGAGTTCGAGACCGACCACGGCTTCGTGTACCGCGAGCACC
ACAACTCCCCCGGCTACTACGACGGCCGCTACTGGACCATGTGGAAGCTGCCCATGTTCGGCTGCACCGACCCCGCCCAGGTG
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Helix B *RbcS* sequence

ATGATGGTGTGGCCCACCCAGAACATGAAGCGCTACGAGACCCTGTCCTACCTGCCCCCCCTGACCACCGACCAGCTGGCCCG
CCAGGTGGACTACCTGCTGAACAACAAGTGGGTGCCCTGCCTGGAGTTCGAGACCGACCACGGCTTCGTGTACCGCGAGCACC
ACAACTCCCCCGGCTACTACGACGGCCGCTACTGGACCATGTGGAAGCTGCCCATGTTCGGCTGCACCGACCCCATGCAGGTG
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Helix AB *RbcS* sequence

ATGATGGTGTGGCCCACCCAGAACATGAAGCGCTACGAGACCCTGTCCTACCTGCCCCCCCTGACCGACGAGCAGATCGCCGC
CCAGGTCGACTACATCGTCGCCAACAAGTGGGTGCCCTGCCTGGAGTTCGAGACCGACCACGGCTTCGTGTACCGCGAGCACC
ACAACTCCCCCGGCTACTACGACGGCCGCTACTGGACCATGTGGAAGCTGCCCATGTTCGGCTGCACCGACCCCATGCAGGTG
CTGCGCGAGATCGTCGCCTGCACCAAGGCCTACCCCAACGCCTTCATCCGCATCATCGGCTTCGACTCCAACCGCCAGGTGCA
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Reciprocal *RbcS* sequence

ATGATGGTCTGGACCCCGGTCAACAACAAGATGTTCGAGACCTTCTCCTACCTGCCTCCTCTGACCACCGACCAGCTGGCCCG
CCAGGTGGACTACCTGCTGAACAACGGCTGGATCCCCTGCCTGGAGTTCGCTGAGGCCGACAAGGCCTACGTGTCCAACGAGT
CGGCCATCCGCTTCGGCAGCGTGTCTTGCCTGTACTACGACAACCGCTACTGGACCATGTGGAAGCTGCCCATGTTCGGCTGC
CGCGACCCCGCCCAGGTGCTGAACGAGCTGGAGGAGTGCAAGAAGGAGTTCCCCGATGCCTACGTGCGCCTGGTGGCCTTCGA
CAACCAGAAGCAGGTGCAGATCATGGGCTTCCTGGTCCAGCGCCCCAAGACTGCCCGCGACTTCCAGCCCGCCAACAAGCGCT
CCGTGTAA

Fig. S1. (*A*) Cartoon representation of all of the Rubisco variants used in this work. The hexadecameric holoenzyme is viewed from the "top," through the axis of the solvent channel. Rubisco large subunit dimers are in gray. Small subunits encoded by algal (*Chlamydomonas*) sequence are in green. Small subunits encoded by higher plant (spinach) sequence are in orange. The four mutants generated in this work (helix A, helix B, helix AB, and reciprocal) have a chimeric small subunit combining algal (green) and higher plant (orange) elements. The two small subunit α-helices are represented by barrels. (*B*) Codon-optimized coding sequences for the mature small subunits of chimeric variants of the spinach and *Chlamydomonas* Rubisco. These sequences were used to exactly replace the *Chlamydomonas* RbcS1 mature-protein coding sequence of pSS1-ITP (1) to generate plasmids used to transform a *Chlamydomonas* RbcS null mutant (2). α-Helix sequences are underlined.

1. Genkov T, Meyer M, Griffiths H, Spreitzer RJ (2010) Functional hybrid rubisco enzymes with plant small subunits and algal large subunits: Engineered rbcS cDNA for expression in *Chlamydomonas*. *J Biol Chem* 285(26):19833–19841.
2. Khrebtukova I, Spreitzer RJ (1996) Elimination of the *Chlamydomonas* gene family that encodes the small subunit of ribulose-1,5-bisphosphate carboxylase/oxygenase. *Proc Natl Acad Sci USA* 93(24):13689–13693.

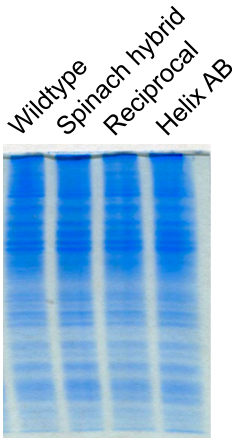

Fig. S2. Coomassie blue-stained gel corresponding to Western blot in Fig. 2*B*.

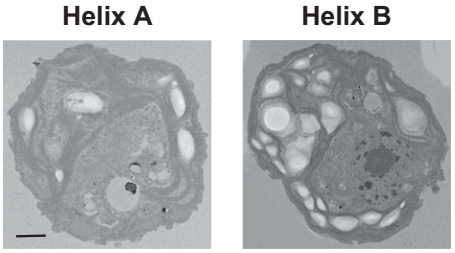

Fig. S3. Representative electron micrographs of the Rubisco SSU single α-helix mutants helix A and helix B. Both mutants systematically lacked a pyrenoid. (Scale bar, 0.5 μm.)

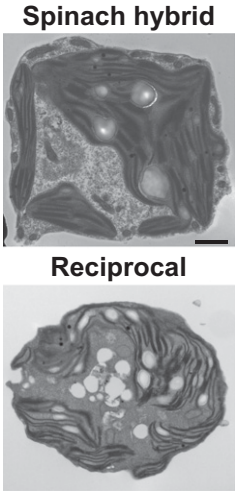

Fig. S4. Representative electron micrographs of pyrenoidless spinach hybrid (*Upper*) and reciprocal (*Lower*), displaying a marked phenotype of thylakoid membrane hyperstacking. (Scale bar, 1 μm.)