

*Supplementary Material*

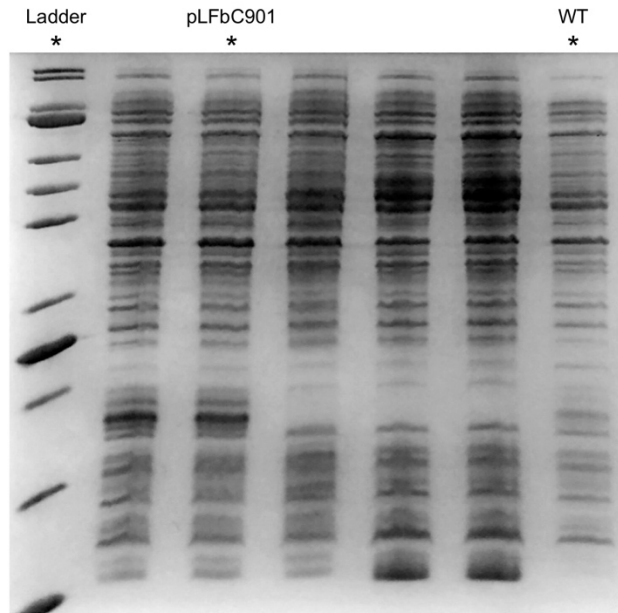
# Engineering and modulating functional cyanobacterial CO<sub>2</sub>-fixing organelles

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**Supplementary Figures and Tables**



**Supplementary Figure 1. Raw SDS-PAGE gel image of the total cell extracts of the recombinant *E. coli* pLFbC901 and the *E. coli* WT cells.** The sample lanes of protein ladder, WT and pLFbC901 were selected to be shown in Figure 1C.

**A**

**CbbL** ----MAVKKYSAGVKEYRQTYWMPEYTPLDSDILACFKITPQPGVDREAAAAVAESST  
**RbcL** MPKTQSAAGYKAGVKDYKLTYYTPDYTPKDTDLLAAFRFSPQPGVPADEAGAAIAESST  
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**CbbL** GTWTTVWTDLLTDMDYKGRAYRIEDVPGDDAAFYAFIAYPIDLFEEGSVVNVFTSLVGN  
**RbcL** GTWTTVWTDLLTDMDRYKGYKCYHIEPVQGEENSYFAFIAYPLDLFEEGSVTNILTSIVGN  
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**CbbL** VFGFKAVRGLRLEDVRFPLAYVKTCGGPPHGIQVERDKMNKYGRPLLGCITKPKLGLSAK  
**RbcL** VFGFKAIRSLRLEDIRFPVALVKTFQGGPPHGIQVERDLLNKYGRPMLGCITKPKLGLSAK  
 \*\*\*\*\*:\*.\*\*\*\*\*:\*\*\*:\* \*\*\* \*\*\*\*\* :\*\*\*\*\*:\*\*\*\*\*\*\*\*\*\*

**CbbL** NYGRAVYECLRGGLDFTKDDENINSQPFMRWRDRFLFVQDATETAEAOQTGERKGHYLNVT  
**RbcL** NYGRAVYECLRGGLDFTKDDENINSQPFQWRDRFLFVADAIHKSQAETGEIKGHYLNVT  
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**CbbL** APTPEEMYKRAEFAKEIGAPIIMHDYITGGFTANTGLAKWCQDNGVLLHIHRAMHAVIDR  
**RbcL** APTCEEMKRAEFAKELGMPIIMHDFTAGFTANTTLAKWCRDNGVLLHIHRAMHAVIDR  
 \*\*\* \*\* \*\*\*\*\*:\* \*\*\*\*\*:\*.\*\*\*\*\* \*\*\*\*\*:\*\*\*\*\*\*\*\*\*\*

**CbbL** NPNHGIHFRVLTKILRLSGGDHLHTGTVVGKLEGDRASTLGWIDLLRESFIPEDRSRGIF  
**RbcL** QRNHGIHFRVLAKCLRLSGGDHLHSGTVVGKLEGDKASTLGFVDLMREDHIEADRSRGVF  
 : \*\*\*\*\*:\* \*\*\*\*\*:\*\*\*\*\*:\*\*\*\*\*:\*\*\*:\*. \* \*\*\*\*\*:\*

**CbbL** FDQDWGSM PGVFAVASGGIHVWHMPALVNI FGDDSVLQFGGGTLGHPWGNAAAGAAANRVA  
**RbcL** FTQDWASMPGVLPVASGGIHVWHMPALVEIFGDDSVLQFGGGTLGHPWGNAPGATANRVA  
 \* \*.\*\*.\*\*\*:\*.\*\*\*\*\*\*:\*\*\*\*\*:\*\*\*\*\*.\*:\*\*\*\*\*

**CbbL** LEACVEARNQGRDIEKEGKEILTAAAOHSPELKIAMETWKEIKFEFDTVDKLDTONR  
**RbcL** LEACVQARNEGRDLYREGGDILREAGKWSPELAAALDLWKEIKFEFETMDKL-----  
 \*\*\*\*\*:\*\*\*:\*\*\*: \*\*: \*\* \*.: \*\*\*\* \*: : \*\*\*\*\*:\*.\*\*\*

**B**

**CcmK2** ----MPIAVGMIETLGFPAVVEAADAMVKAARVTLVGYEKIGSGRVTVIVRGDVSEVQAS  
**CsoS1A** MADVTGIALGMIETRGLVPAIEAADAMTKAAEVRLVGRQFVGGGYVTVLVRGETGAVNAA  
**PduA** ---MQQEALGMVETKGLTAAIEAADAMVKSANVMLVGYEKIGSGLVTVIVRGDVGAVKAA  
 \*:\*\*\*:\*\* \*: .:\*\*\*\*\*.\*:\*. \* \*\* : :\*. \* \*\*\*:\*\*\*:.. \*:\*\*

**CcmK2** VSAGLDSAKRVAGGEVLSHHIIARPHENLEYVLPPIRYTEAVEQFRM  
**CsoS1A** VRAGADACERVGDG-LVAAHIIARVHSEVENILPKAPQA-----  
**PduA** TDAGAAAARN--VGEVKAVHVIPRPHTDVEKILPKGISQ-----  
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**Supplementary Figure 2. Sequence alignment of representative BMC proteins using Clustal Omega multiple sequence alignment program. (A) Sequence similarity (78%) between the Form 1B Rubisco large subunit CbbL from *H. neapolitanus* and the Form 1B Rubisco large subunit RbcL from Syn7942. (B) Sequence conservation of representative BMC shell hexamers, CcmK2 from Syn7942, CsoS1A from *H. neapolitanus* and PduA from *S. Typhimurium* (CsoS1A – CcmK2: 53%; PduA – CcmK2: 61%).**



CyanoBase	UniProt	Protein Description	Mascot score	Coverage (%)	Unique peptides
Synpcc7942_1426	Q31NB3	Rubisco large chain RbcL	206	14	KLTYYPDYTPKD KDTDLLAAFRF RAVYECLRG KASTLGFVDMRE REDHIEADRS KWSPELAAALDLWKE
Synpcc7942_1427	Q31NB2	Rubisco small chain RbcS	178	42	RFETFSYLPPLSDRQ KLPLFDCKS KSPQQVLDEVRE RSEYGDCYIRV RVAGFDNIQK
Synpcc7942_1421	Q03511	CcmK2	155	40	RVTLVGYEKI RVTVIVRG RGDVSEVQASVSAGLDSAKR RYTEAVEQFRM
Synpcc7942_1423	Q03513	CcmM	129	6	RLSSEVITQVRS RNLLAQGYRI RVFEALIQDPQGPVGSACA
Synpcc7942_0285	Q31RK2	CcmK4	112	24	RFAVNIRG KTAMDAGIEAAKN RLSAEGTGSGRR

**Supplementary Table 2. Proteomic analysis of identified  $\beta$ -carboxysome proteins expressed by pLFbC901 in the whole *E. coli* cell extract.**