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online data

SUPPLEMENTARY ONLINE DATA

Elucidation of the preferred routes of C8-vinyl reduction in chlorophyll and bacteriochlorophyll biosynthesis

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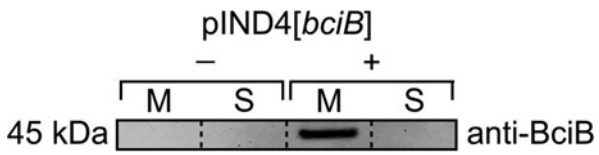

Figure S1 Expression of recombinant *bciB* in *R. sphaeroides* detected by Western blotting

Separated membrane (M) and soluble (S) fractions of Δ*bchCXF*/Δ*bciA* lacking (–) and containing (+) pIND4[*bciB*] grown under expression-inducing conditions were separated by SDS/PAGE and transferred to a membrane that was probed with an anti-BciB antibody.

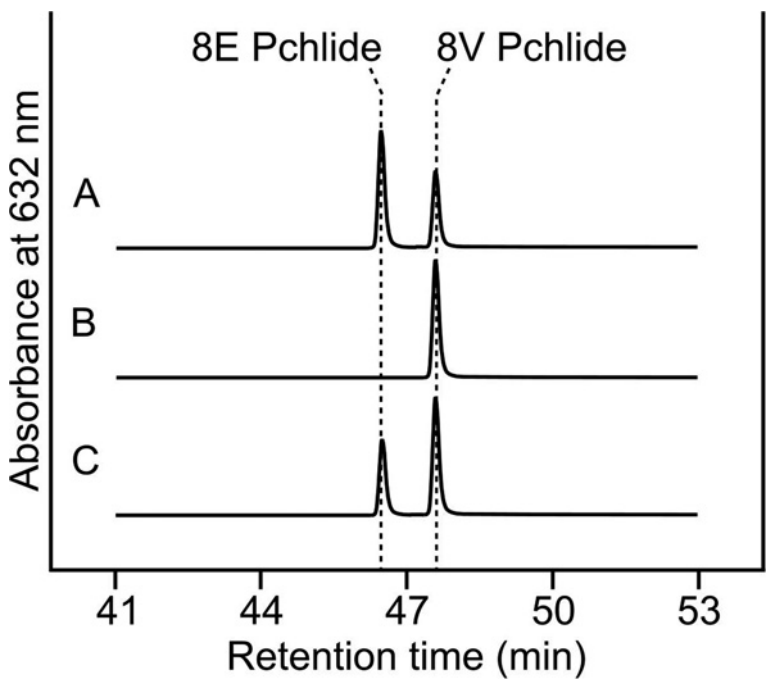

Figure S2 HPLC elution profiles of pigments extracted from *Synechocystis* strains lacking *chlB*

HPLC elution profiles of extracts from pellets of (A) Δ*chlB*, (B) Δ*bciB*/Δ*chlB* and (C) Δ*chlB*/Δ*bciB*::*bciA*^{Rs} strains after 16 h growth without illumination. Traces are normalized to major peak height for clarity.

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Table S1 List of strains and plasmids described in the present study

Strain/plasmid	Properties	Source
E. coli		
JM109	Cloning strain for pK18*mobsacB* and pIND4 constructs	Promega
S17-1	Conjugative strain for pK18*mobsacB* and pIND4 constructs	[1]
R. sphaeroides		
WT	2.4.1	S. Kaplan*
V3	Unmapped mutant in a DPOR subunit-encoding gene	[2]
V3::*bciB*^{Syn}	V3 harbouring pIND4[*bciB*]	The present study
V3/Δ*bciA* V3/Δ*bciA*::*bciB*^{Syn}	Unmarked deletion mutant of rsp_3070 in V3 V3/Δ*bciA* harbouring pIND4[*bciB*]	The present study
Δ*bchCXF*	Unmarked deletion mutant of *bchC, bchX* and *bchF* in WT	The present study
Δ*bchCXF*/Δ*bciA*	Unmarked deletion mutant of rsp_3070 in Δ*bchCXF*	The present study
Δ*bchCXF*::*bciB*^{Syn}	Δ*bchCXF* harbouring pIND4[*bciB*]	The present study
Δ*bchCXF*/Δ*bciA*::*bciB*^{Syn}	Δ*bchCXF*/Δ*bciA* harbouring pIND4[*bciB*]	The present study
Synechocystis		
WT	sp. PCC6803	R. Sobotka†
Δ*bciB*	*Em*^{R} replacement of central portion of *slr1923* in WT	[3]
Δ*bciB*::*bciA*^{Rs}	rsp_3070 and *Kan*^{R} replacement of *psbAII* in Δ*bciB*	[3]
Δ*chlB*	*Zeo*^{R} replacement of central portion of slr0772 in WT	The present study
Δ*bciB*/Δ*chlB*	*Zeo*^{R} replacement of central portion of slr0772 in Δ*bciB*	The present study
Δ*bciB*/Δ*chlB*::*bciA*^{Rs}	*Zeo*^{R} replacement of central portion of slr0772 in Δ*bciB*::*bciA*	The present study
Plasmid		
pK18*mobsacB*	Allelic exchange vector, *Km*^{R}	J. Armitage‡
pIND4	IPTG-inducible expression vector for *R. sphaeroides*, *Km*^{R}	[4]
pIND4[*bciB*]	slr1923 cloned into the BamHI/HindIII sites of pIND4	The present study

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Table S2 List of primers used in the present study

Restriction enzyme cleavage sites used for cloning are underlined in the primer sequence.

Primer	Sequence (5′→3′)	Cleavage site
rsp_3070UpF	CCGGAATTCGGACATCCTGACCGGTTTCCTGTCC	EcoRI
rsp_3070UpR	GCTCTAGAGGACATGGCGGAACTCCTCGGG	XbaI
rsp_3070DownF	GCTCTAGACGTTGACATTGGTGCCGGTCGG	XbaI
rsp_3070DownR	CCCAAGCTTCGAAGGCGATGCGCGCGAGGC	HindIII
rsp_3070CheckF	GACGACGAGAAGCTGGCCTACGG	
rsp_3070CheckR	GGCAGGTACCGGAGAGCGGTTAGG	
bchCXUpF	CCGGAATTCGCTCCTGCACCGGGTGCGCG	EcoRI
bchCXUpR	GCGCTCTAGAAGCGTTTTCCCCCGCGCTCTTC	XbaI
bchCXDownF	GCGCTCTAGATCTCGATACCCTCGCGCGGC	XbaI
bchCXDownR	CCCCAAGCTTGTCCTCGAACAGCTTGCCCGTG	HindIII
bchCXCheckF	GACGATCCACTGCCGCTCGG	
bchCXCheckR	CAGCGGCACGCCCGAGGCG	
bchFUpF	CCGGAATTCCCCCGCCCTGTCTCTCTGCAAGCC	EcoRI
bchFUpR	CGGGGGCGGAAGGTCAAGGCTCATCTTGAGGTTCGCCTTCCGAGGAGGGCCC	
bchFDownF	CCTACGTCATCAACGCCGGGCAGTTCC	
bchFDownR	CCCAAGCTTGCGCCGAGAGCCGCTCGGCCGCCCGG	HindIII
bchFCheckF	GACGGCCACCGGGCCCTC	
bchFCheckR	GAGGCGAGGCAGGCATCCTC	
1923INDF	CGCGGATCCACCGTTCCTGCCCCCCACC	BamHI
1923INDR	CCCAAGCTTTTATTGCTGGGGAAGTTTATACTGC	HindIII
chlBUpF	GCATCGCTTATTGTTCTCAACG	
chlBUpR	ACATTAATTGCGTTGCGCTCACTGCCGTGCATAATGGCGTGGACG	
chlBDownF	CAACTTAATCGCCTTGCAGCACATGCAGAATTGAATAAAGTGCCAGGG	
chlBDownR	CCTTCAAAGGCCATCACCC	
zeo^RF	TGACCAGCGCCGTTCCGGTG	
zeo^RR	CGGGTCGCGCAGGGCGAAC	

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