Table S1. Strains and plasmids used in this study

Strains	Relevant Characteristic(s)	source
Nostoc punctiforme strains		
ATCC 29133	wild type	ATCC
UCD153	Laboratory derivative of *N. punctiforme* ATCC 29133 with reduced motility	(1)
TNM620	UCD153 with a Tn5-1063 insertion after nucleotide 796 of *hrmK* (Npun_R3825)*	This study
TNM710	UCD153 with a Tn5-1063 insertion after nucleotide 757 of *hrmK*	This study
TNM14194	UCD153 with a Tn5-1063 insertion after nucleotide 1713 of *hrmK*	This study
UOP131	Δ*sigC* (Npun_F0996)	This study
UOP132	Δ*sigJ* (Npun_R1337)	This study
UOP139	Δ*hrmK*	This study
Plasmids		
pAM504	Mobilizable shuttle vector	(2)
pRL278	Mobilizable suicide vector	(3)
pRL1063a	Suicide vector carrying Tn5-1063, a Tn5 derivative transposon	(4)
pDDR420	Suicide vector for in-frame deletion of *hrmK* [1-4]†	This study
pDDR456	Shuttle vector containing *hrmK* and 5' intergenic region [5-6]	This study

* locus tag denoted in parentheses

† numbers in brackets correspond to primers used to construct plasmid. Detailed information on primers can be found in Table S2

Table S2. Oligonucleotides used in this study

Oligonucleotide	Sequence	Number	qPCR primer set target gene
NpR3825-5'-F	ATATAGGATCCGAATCACTGAGGCAAATAACG	1	
NpR3825-5'-R	CTATATTGGCGCTGCTGCATGGTCAACTAG	2	
NpR3825-3'-F	CATGCAGCAGCGCCAATATAGAGAGCAGG	3	
NpR3825-3'-R	ATATAGAGCTCATCTGTCGTTAACACAAACTC	4	
PNpR3825-BamHI-F	ATATAGGATCCCATAAGTCTTAAGTTGCTTTCAC	5	
NpR3825-SacI-R	ATATAGAGCTCCTATATTGGCGGTTTGCTG	6	
qNpR3825-F2	TGAAAATCAACAGCCACCAA	7	*hrmK*
qNpR3825-R2	AGTTGGCAGACCTGGACATC	8	
qNpun_F0996-F2	GGCGATCGCAACTTCTAGTC	9	*sigC*
qNpun_F0996-R2	ACTTGGGTCGGTGTCATCTC	10	
qNpun_R1337-F1	TGAGATGCTGCACTTTTTGC	11	*sigJ*
qNpun_R1337-R2	TTTTGAGCGGCTAACTTGGT	12	
qNpun_F4811-F2	TGTTTGGGAGAATTGGTTCC	13	*sigF*
qNpun_F4811-R2	ATCCCGAGATGTTCTGCAAC	14	
qhfq-F2	GCTTACCGAATTTGACACCAC	15	*hfq*
qhfq-R2	TGTATGCGATCGCTTGTTTC	16	
qNpun_R0118-F2	AATGGTGTCGGCTACAAAGG	17	*pilB*
qNpun_R0118-R2	TCGGCTTCCAAACCAGTATC	18	
qpilA-F2	TCTGGTTGCCAACAATGGTA	19	*pilA*
qpilA-R2	ACTTCAGCACTCCGATCACC	20	
qNpun_F0070-F2	GGTAGCCAAATTCACCCTGA	21	*hpsE*
qNpun_F0070-R2	TTGCCTTGAACTCTCCCAGT	22	
qNpun_r018_F1	TAAGAGCGCACCAGCAGTAT	23	*rnpB*
qNpun_r018_R1	CATTGAGCGGAACTGGTAAA	24	
Tn5-seq-F	CGATGAAGAGCAGAAGTTATC	25	
Tn5-seq-R	GGCTCTATTCAGGATAAATC	26	
Tn5-seq-F-nest	CGTTACCATGTTAGGAGGTC	27	

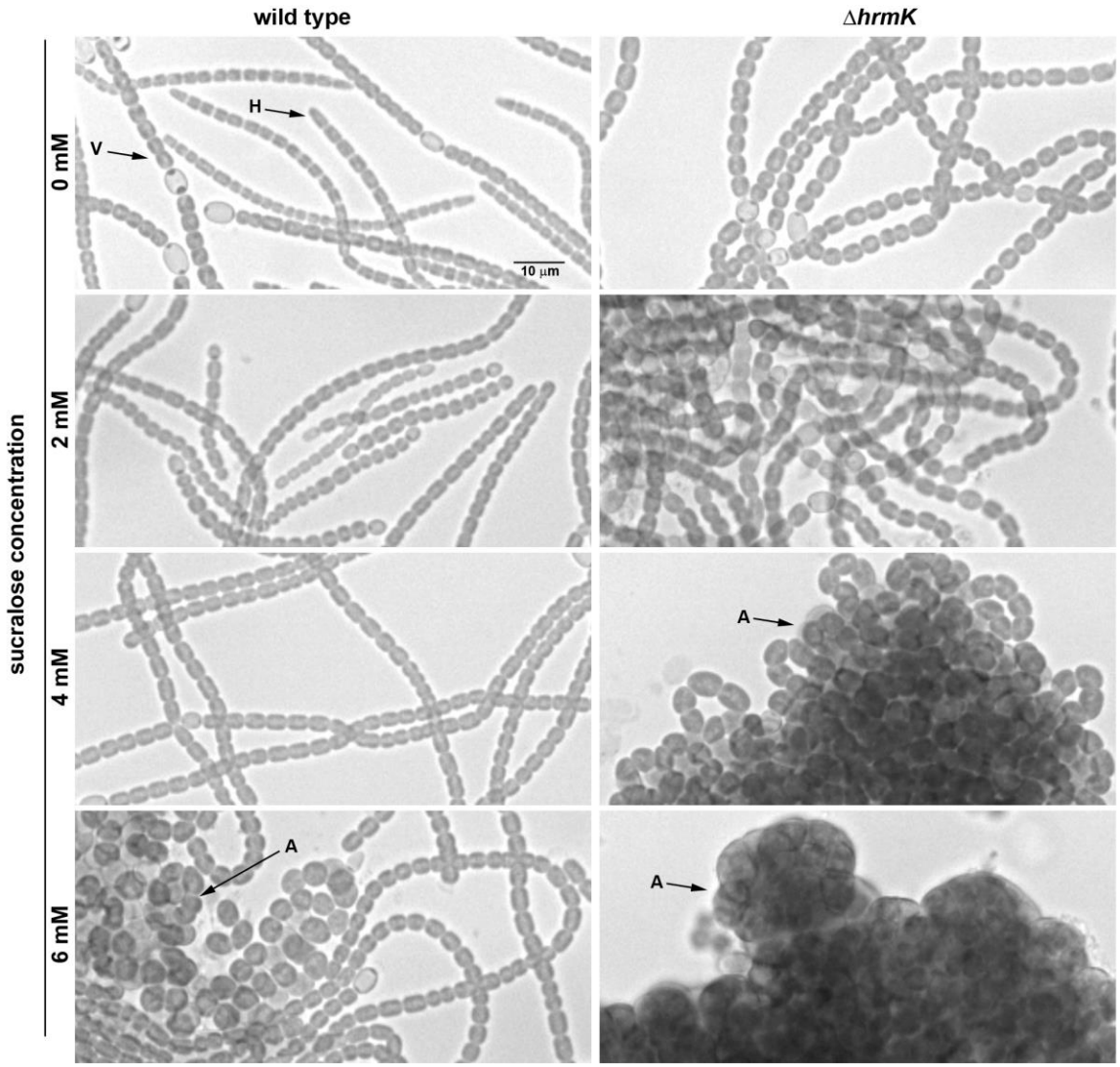

Figure S1. The effect of sucralose supplementation on the wild-type and Δ*hrmK* strain. Light micrographs of the wild-type and Δ*hrmK* strains from cultures supplemented with various concentrations of sucralose (as indicated). Examples of a vegetative (V) and hormogonium filaments (H), as well as aseriate colonies (A) are indicated.

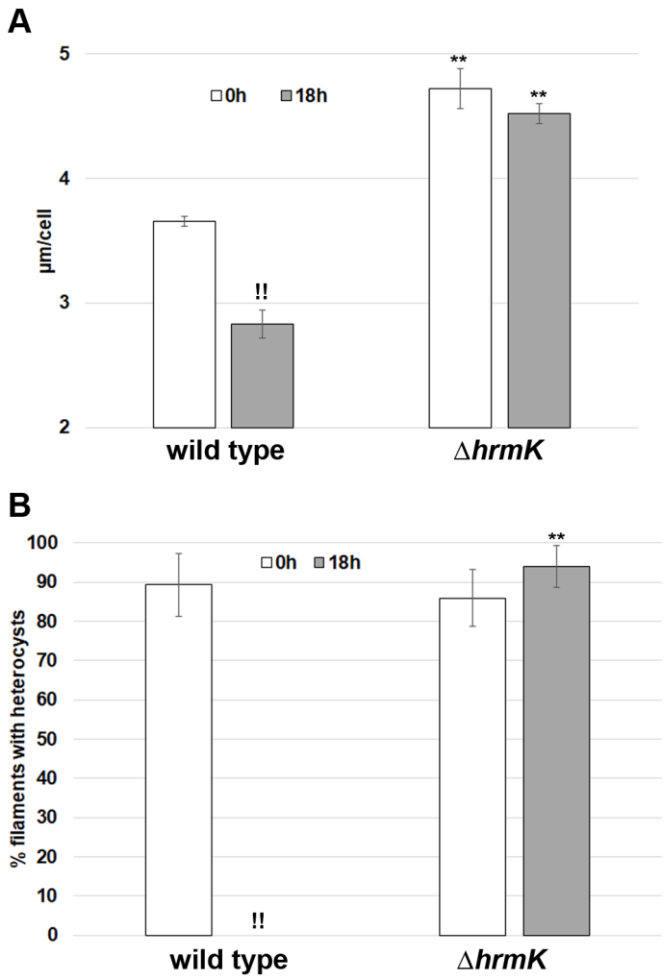

Figure S2. Quantitative analysis of hormogonium morphology (cell length and % filaments with heterocysts) in the wild type and Δ*hrmK* strain (n=3, error bars = +/-1 SD) 0 and 18 h post hormogonium induction. **(A)** Average cell length. **(B)** Percentage of filaments containing heterocysts. * = p-value<0.05, ** = p-value<0.01 as determined by two-tailed Student's t-Test between the wild type and Δ*hrmK* strain at the corresponding time point. ! = p-value<0.05, !! = p-value<0.01 as determined by two-tailed Student's t-Test between 0 and 18 h for the same strain.

SMOV1. Time lapse microscopy of the wild-type and Δ*hrmK* strains (as indicated).

Supplemental References Cited

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3. Cai YP, Wolk CP. 1990. Use of a conditionally lethal gene in *Anabaena* sp. strain PCC 7120 to select for double recombinants and to entrap insertion sequences. J Bacteriol 172:3138-3145.

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