Supplementary Information

Methods and materials

Bacterial strain, chemicals, and media

Synechococcus elongatus PCC7942 was obtained from ATCC (American Type Culture Collection, ATCC 33912). The BG-11 media contain the following composition per liter: 1.5 g $NaNO_3$, 75 mg $MgSO_4 \cdot 7H_2O$, 36 mg $CaCl_2 \cdot 2H_2O$, 6 mg citric acid, 2.86 mg H_3BO_3, 1.81 mg $MnCl_2 \cdot 4H_2O$, 222 μg $ZnSO_4 \cdot 7H_2O$, 390 μg $Na_2MoO_4 \cdot 2H_2O$, 79 μg $CuSO_4 \cdot 5H_2O$, 49.4 μg $Co(NO_3)_2 \cdot 6H_2O$, 6 mg ferric ammonium citrate, 20 mg Na_2CO_3, and 30.5 mg KH_2PO_4 [1]. Different concentrations of kanamycin (Aladdin Co., China) were added into the media when needed. All chemicals were purchased from Sinopharm Chemical Reagents Co. Ltd unless otherwise stated.

Cell growth and biofilm formation

S. elongatus was inoculated in BG-11 media at 30°C with constant light (2,000 - 3,000 Lux) till early exponential phase (6 d) and stationary phase (12 d). The growth profiles were monitered via recording the optical density (OD) at 730 nm with a UV-2000 spectrophotometer (UNICO, USA) at different time intervals. The cells were then harvested, normalized to OD_{730} 1.0, and inoculated to 12-well plates with BG-11 media and low-level concentrations of kanamycin (Aladdin Co., China) for biofilm formation experiments. The tested levels of kanamycin included 0, 0.05, 0.10 and 0.15 μg/mL,

which have neglectable effects on cell growth (Fig. S1). After 6 d, the whole culture and suspended cells in the 12-wells were quantified via measuring the amount of chlorophyll a by spectrophotometric method [2] and the biofilms were regarded as the differences of absorbance between whole culture and suspended cells.

Transcriptional analysis

The RNA was extracted from the *S. elongatus* cultured with kanamycin for 6 d using the RNAprep pure Cell/Bacteria Kit (Tiangen Biotech. Co. Ltd., Beijing, China), and the cDNA was synthesized using the Primescript™ RT reagent kit (Takara Biotech. Co. Ltd., Dalian, China). Quantitative real-time polymerase chain reaction was performed in 20 μL reaction mixture. The $2^{-\Delta\Delta CT}$ method was used to quantify the expression levels of *rbcL* (encoding the large subunit of RuBisCO) and *icfA* (encoding the β-form carbonic anhydrase) using the 16s rDNA sequence as the housekeeping gene [3]. All experiments were followed by the protocols from manufacturers and the primers used in this study were summarized in Table S1.

Reference

1. S. Diamond, D. Jun, B. E. Rubin, and S. S. Golden, *Proc. Natl. Acad. Sci. U.S.A.*, 2015, **112**, E1916–25.

2. D. Schatz, E. Nagar, E. Sendersky, R. Parnasa, S. Zilberman, S. Carmeli, Y. Mastai, E. Shimoni, E. Klein, O. Yeger, Z. Reich, and R. Schwarz, *Method Enzymol*, 2013, **15**, 1786–1794.

3. F. Pinto, C. C. Pacheco, D. Ferreira, P. Moradas-Ferreira, and P. Tamagnini, *PLoS ONE*, 2012, **7**, e34983.

Table S1 Primers used in this study

Primer Names	Sequences
rbcL forward	GGGTCTTCTTCACCCAAGATT
rbcL reverse	CGGAGTCATCACCGAAGATTT
icfA forward	TCAATCAGCTGCAAGAGGAC
icfA reverse	CCAAGTCGTCAGTCTCATAACC
16s rDNA forward	GTAGCGGTGAAATGCGTAGA
16s rDNA reverse	CGTCCATGAGCGTCAGTTAT

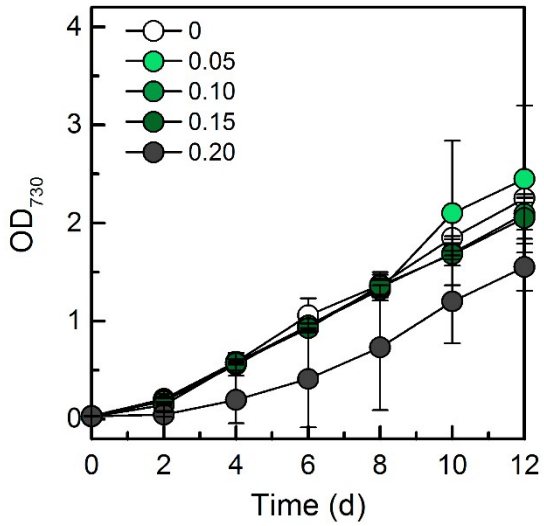

Fig. S1 Growth profiles of *S. elongatus* in the presence of different concentrations of kanamycin. All experiments were conducted at least in triplicate and the error bars denote ±1 SD from the means of independent experiments.