# Mitigating harmful cyanobacterial blooms in drinking water reservoirs through *in-situ* sediment resuspension

Jiao Fang<sup>a,b</sup>, Yande Li<sup>d</sup>, Ming Su<sup>a,f,\*</sup>, Tengxin Cao<sup>a,f</sup>, Xufeng Sun<sup>c</sup>, Yufan Ai<sup>a,f</sup>, Jinyi Qin<sup>e</sup>, Jianwei Yu<sup>a,f</sup>, Min Yang<sup>a,f,\*</sup>

<sup>a</sup> State Key Laboratory of Environmental Aquatic Chemistry, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, P.O. Box 2871, Beijing, 100085,

<sup>b</sup>School of Environment and Spatial Informatics, China University of Mining and Technology, Xuzhou, 221116,
<sup>c</sup>Zhejiang Weicheng Huanbao Co. Ltd., Yunxiu North Road 1200, Huzhou, 313200,

<sup>d</sup>Management Station of Shuangxikou Reservoir, Reservoir Management Service Center of Yuyao, Ningbo, 315423,

<sup>e</sup>School of Civil Engineering, Chang'an University, Xi'an, 710054,

<sup>f</sup>University of Chinese Academy of Sciences, Beijing, 100049,

<sup>1</sup> State Key Laboratory of Environmental Aquatic Chemistry, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences

#### **Supplementary Material**

Figures and/or tables are provided below as the supplementary evidences to the main text.

<sup>&</sup>lt;sup>2</sup> School of Environment and Spatial Informatics, China University of Mining and Technology

<sup>&</sup>lt;sup>3</sup> Zhejiang Weicheng Huanbao Co. Ltd.

<sup>&</sup>lt;sup>4</sup> Management Station of Shuangxikou Reservoir, Reservoir Management Service Center of Yuyao

<sup>&</sup>lt;sup>5</sup> School of Civil Engineering, Chang'an University

<sup>&</sup>lt;sup>6</sup> University of Chinese Academy of Sciences

<sup>\*</sup> Correspondence: Ming Su <mingsu@rcees.ac.cn>, Min Yang <yangmin@rcees.ac.cn>

<sup>\*</sup>Corresponding author

Email addresses: mingsu@rcees.ac.cn (Ming Su), yangmin@rcees.ac.cn (Min Yang)

#### 1. Test in laboratory simulators

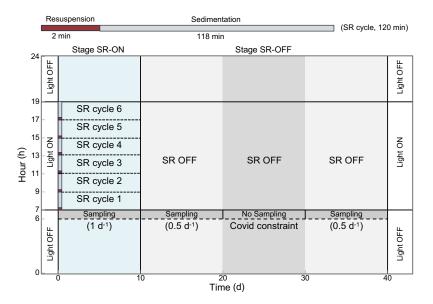


Fig. S1 The figure illustrated the protocol of Sediment Resuspension in laboratory simulators. As shown above, SR operation was periodically conducted in LEs for two minutes (then settled for the following 118 mins) every 2 hours from 7:00 to 19:00 during the first day (Stage SR-ON). No SR operation was performed for all three simulators from day 11 to day 40 (Stage SR-OFF). Besides, the unique light source was switched on from 7:00 to 19:00 during six SR cycles to provide light for the growth of algae. Water and sediments samples were collected before the first SR cycle began. The sampling frequency was every day during Stage SR-ON and one every 2 days during Stage SR-OFF (no samples were collected during day 21–30 due to COVID 19 constraints).

### 2. Field applications

## 2.1. Field applications in five drinking water reservoirs



Fig. S2 The figure displayed the field applications of SR operations in five drinking water reservoirs: SMH Reservoir (A), SXK Reservoir (B), NJ Reservoir (C), CX Reservoir (D) and HM Reservoir (E)

### 2.2. Equipment operational parameters, basic feature and sampling of five reservoirs

Reservoir	SMH	SXK	NJ	CX	НМ
Depth (m)	10.5	28.7	7.8	4	6.4
Work area (ha)	25	18	26.5	3.8	12
Risk area (ha)	12	6.4	14.3	1.3	4.12
Numbers of boats	1	1	2	3	3
Work time (d)	180~210	180~210	14	11	10
Work hour (h/d)	10	10	10	10	10
Frequency (1/d)	0.625	1.172	1.049	17.31	5.461
Investigation time	Apr.~Oct., 2023	Apr.~Oct., 2023	Sep., 2021	Jul., 2022	Oct., 2023
Dominant genus	Achnanthes;	Pseudanabaena;	Microcystis;	Microcystis	Raphidiopsis
	Microcystis	Raphidiopsis	Synedra		
Sampling frequency	biweekly	biweekly	daily	daily	daily
Sampling depth (m)	0.5;3.0;6.0	0.5;3.0;6.0	0.5;3.0;6.0	0.5;2.0;4.0	0.5;3.0;6.0
Collect sediment?	yes	yes	no	no	no

 $Table \, S1 \, Equipment \, operational \, parameters, \, basic \, characteristics \, of \, algal \, blooms, \, and \, sampling \, protocols \, in \, five \, drinking \, water \, reservoirs.$ 

### 3. The changes in turbidity and extinction coefficient during a single SR operation

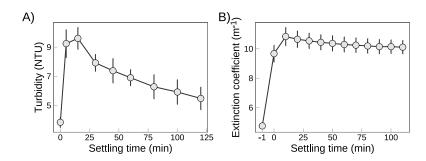


Fig. S3 The changes in turbidity over time during a single SR operation (repeated three times, n=3) in the field test of SXK Reservoir (A); The changes in extinction coefficient over time in SR cycles (n=60) during LEs tests (B). Each data point represents the mean value of the respective measurements.

### 4. Evaluation of nitrogen changes for SR operation in laboratory simulators

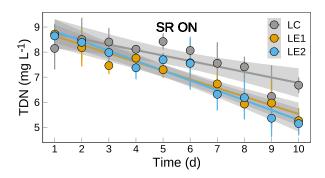


Fig. S4 The dynamics of nitrogen concentration (B) in laboratory simulators, comprising two simulators with Sediment Resuspension (SR) operations (designated as LE1 and LE2), and one simulator without SR serving as an untreated control (LC).

#### 5. The difference in dissolved nitrogen and dissolved organic carbon during SR operation

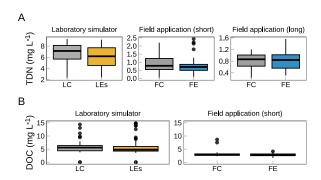


Fig. S5 Comparison of total dissolved nitrogen (TDN) and dissolved organic carbon (DOC) between SR-operated samples (LC, FC) and untreated samples (LEs, FE) in laboratory simulators and field applications. Panel A illustrates the difference in TDN between SR-operated samples and untreated samples within laboratory simulators (p-value = 0.0116), three short-term SR-operated reservoirs (CX, NJ and HM Reservoirs, p-value = 0.4445), and two long-term SR-operated reservoirs (SMH and SXK Reservoirs, p-value = 0.8324). Panel B illustrates the difference in DOC between SR-operated samples and untreated samples within laboratory simulators (p-value = 0.6713) and three short-term SR-operated reservoirs (p-value = 0.1104).

### 6. Improvement of sediment quality in laboratory simulators

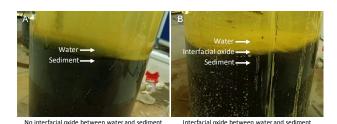


Fig. S6 Interfacial oxide layer was formed between the sediment and the water phase in laboratory simulators with 10 days' SR operation (LE, B) in comparison with untreated control set (LC, A).

# 7. Evaluation of algae control effect and phosphorus changes during SR OFF in laboratory simulators

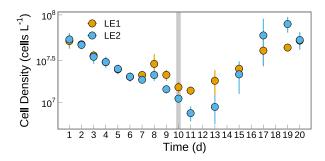


Fig. S7 The dynamics of Microcystis cell density in LEs (LE1 & LE2) during Stage SR-ON (days 1–10) and Stage SR-OFF (days 11–20) are depicted. *Microcystis* cell density decreased during Stage SR-ON but subsequently increased during Stage SR-OFF, providing additional evidence of the impact of SR on algae control.

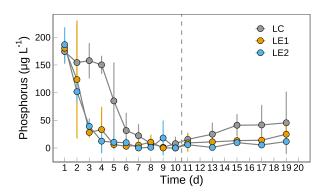
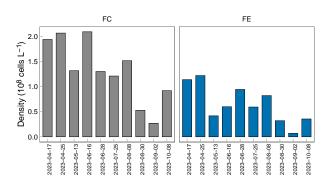


Fig. S8 The dynamics of dissolved phosphorus concentration in LC and LEs over the first 20 days are illustrated. Dissolved phosphorus decreased to extremely low levels during SR in both LC and LEs. However, in LC, dissolved phosphorus began to rise significantly between days 11 and 20, with an increase rate of 4.29  $\mu$ g L<sup>-1</sup> d<sup>-1</sup> (R<sup>2</sup> = 0.92, p-value = 0.0027). In contrast, increasing rate of LEs was much lower (1.55  $\mu$ g L<sup>-1</sup> d<sup>-1</sup>, R<sup>2</sup> = 0.53, p-value = 0.007).

### 8. Algal control effect of SR in drinking water reservoirs



 $Fig. \ S9\ Changes\ in\ total\ algal\ density\ over\ time\ in\ FC\ (without\ SR\ operation)\ and\ FE\ (with\ SR\ operation)\ of\ SMH\ Reservoir.$ 

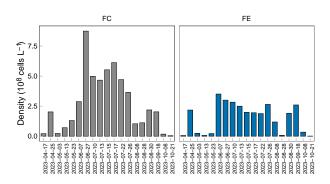


Fig. S10 Changes in total algal density over time in FC (without SR operation) and FE (with SR operation) of SXK Reservoir.

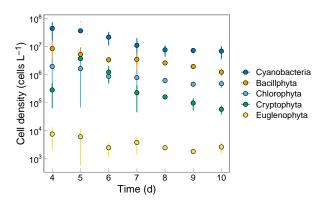


Fig. S11 Changes in different phylum density over time in CX Reservoir. Specifically, severe *Microcystis* bloom occurred in CX reservoir in October of 2021 and one boat was used to resuspend sediment for emergency algae control. Dynamics of cell density for different phylum in the Reservoir during 10 days SR operation were shown here. Significant decline in cell density especially density of Cyanobacteria occurred after the fourth day of SR operation.

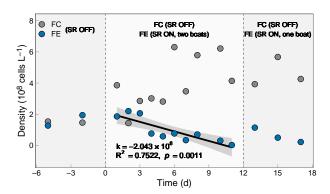


Fig. S12 The changes in total algal density over time in FC (without SR operation) and FE (with SR operation) of CX Reservoir. During day -6 to day 0, severe cyanobacteria bloom was found in the whole reservoir including FE and FC. During day 1–11, two boats were employed for CX reservoir and conducted SR operation in FE. Decrease rate of  $2.043 \times 10^8$  cells L<sup>-1</sup> d<sup>-1</sup> in cell density was found in FE, while density remained rising in FC. After day 11, the algae control effect had been confirmed, and the number of boat decreased to one to maintain effectiveness. The following are additional explanations: (1) Comparison of cell densities between SR-operated site (FE) and untreated site (FC) during day 1–11 is shown in Fig. 4B; (2) several images illustrating the changes in water surface from day 0 to day 10 is shown in Fig. 4D.

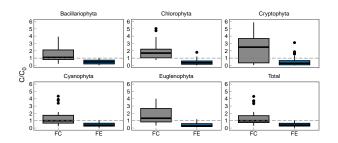


Fig. S13 Comparison of cell density between FC and FE in NJ Reservoir. Here,  $C_0$  represented original cell density of FC and FE, and C represented cell density during SR operation in FE and FC. The dotted line in this figure represented  $C/C_0=1$ . In more detail,  $C_0$  represented original cell density of FC and FE, and C represented cell density during SR operation in FE and FC. Therefore,  $C/C_0>1$  denoted increase in cell density during our investigation, while  $C/C_0<1$  denoted decrease in cell density. Data points of 42 were used for FC and FE in each figure.

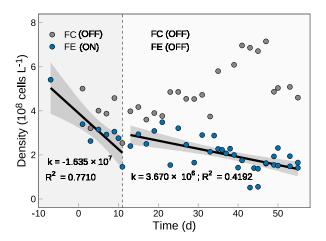


Fig. S14 The changes in total algal density over time in FC (without SR operation) and FE (with SR operation) of HM Reservoir. As shown in the figure, algal density decreased during Stage SR-ON (day 1–11) and stayed in a relatively low level during Stage SR-OFF (day 12–55) in FE. While in FC, algal density increased and stabled at a higher level during the whole investigation in FC. Additionally, comparison of cell densities between FE and FC during day 12–55 is shown in Fig.

#### 9. Summary of algal growth rates under various light conditions

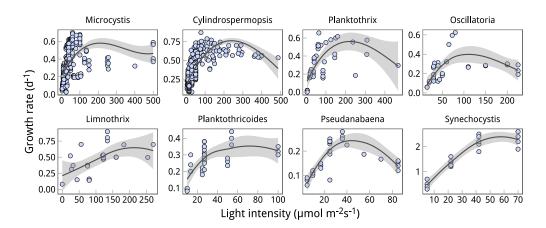


Fig. S15 The effect of light intensity on the growth of cyanobacteria. Data are collected from references: *Cylindrospermopsis* (Briand et al., 2004; Xiao et al., 2017; Bonilla et al., 2012; Xiao et al., 2020; Willis et al., 2016), *Microcystis* (Marinho et al., 2013; Li et al., 2014; Salvador et al., 2016; Xiao et al., 2017; Torres et al., 2016; Hesse et al., 2001; Xiao et al., 2020; Thomas and Litchman, 2015; Bañares-España et al., 2012; e. Mello et al., 2012; Briand et al., 2012; Wiedner et al., 2003; Sugimoto et al., 2015; Lürling et al., 2013; Wu et al., 2011), *Pseudanabaena* (Zhang et al., 2016; Cao et al., 2023), *Limnothrix* (Nicklisch, 1998; Tiwari et al., 2001; Nicklisch, 1999; Shatwell et al., 2012; Daniels, 2016), *Planktothrix* (Jia et al., 2019; Torres et al., 2016; Nicklisch, 1998), *Planktothricoides* (Lu et al., 2022; Mohanty et al., 2022), *Oscillatoria* (Foy, 1983; Van Der Ploeg et al., 1995; Tiwari et al., 2001; Cai et al., 2017; Tang et al., 1997) and *Synechocystis* (Luimstra et al., 2018).

## 10. The changes in Chlorophyll a during a single operation in field tests

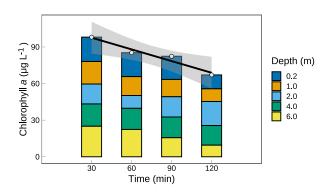


Fig. S16 The changes in Chlorophyll a over time during a single SR operation (repeated three times, n = 3) in the field test of SXK Reservoir (A)

### 11. Investigation of dissolved iron and manganese during SR operations in five Reservoirs

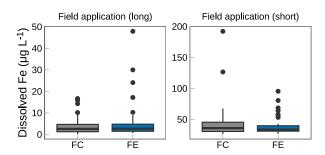


Fig. S17 Comparison of dissolved iron between FC and FE in two reservoirs with long-term SR operation (A) and NJ reservoirs with short-term SR operation (B).

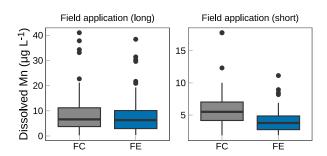


Fig. S18 Comparison of dissolved manganese between FC and FE in two reservoirs with long-term SR operation (A) and NJ reservoirs with short-term SR operation (B).

#### References

- Bañares-España, E., Kromkamp, J.C., López-Rodas, V., Costas, E., Flores-Moya, A., 2012. Photoacclimation of cultured strains of the cyanobacteriumr *Microcystis aeruginosa* to high-light and low-light conditions. FEMS Microbiology Ecology 83, 700–710. URL: https://doi.org/10.1111/1574-6941.12025, doi:10.1111/1574-6941.12025.
- Bonilla, S., Aubriot, L., Soares, M.C.S., González-piana, M., Fabre, A., Huszar, V.L., Lürling, M., Antoniades, D., Padisák, J., Kruk, C., 2012. What drives the distribution of the bloom-forming cyanobacteria *Planktothrix agardhii* and *Cylindrospermopsis Raciborskii*? Fems Microbiology Ecology 79, 594–607. URL: http://dx.doi.org/10.1111/j.1574-6941.2011.01242.x.
- Briand, E., Bormans, M., Quiblier, C., Salençon, M.J., Humbert, J.F., 2012. Evidence of the cost of the production of microcystins by microcystis aeruginosa under differing light and nitrate environmental conditions. PLoS ONE 7, e29981. URL: https://doi.org/10.1371/journal.pone.0029981, doi:10.1371/journal.pone.0029981.
- Briand, J.F., Leboulanger, C., Humbert, J.F., Bernard, C., Dufour, P., 2004. CYLINDROSPERMOPSIS RACIBORSKII (CYANOBACTERIA) INVASION AT MID-LATITUDES: SELECTION, WIDE PHYSIOLOGICAL TOLERANCE, ORGLOBALWARM-ING?1. Journal of Phycology 40, 231–238. URL: https://doi.org/10.1111/j.1529-8817.2004.03118.x, doi:10.1111/j.1529-8817.2004.03118.x.
- Cai, F., Yu, G., Zhang, K., Chen, Y., Li, Q., Yang, Y., Xie, J., Wang, Y., Li, R., 2017. Geosmin production and polyphasic characterization of *Oscillatoria limosa* agardh ex gomont isolated from the open canal of a large drinking water system in tianjin city, china. Harmful Algae 69, 28–37. URL: https://doi.org/10.1016/j.hal.2017.09.006, doi:10.1016/j.hal.2017.09.006.
- Cao, T., Fang, J., Jia, Z., Zhu, Y., Su, M., Zhang, Q., Song, Y., Yu, J., Yang, M., 2023. Early warning of mib episode based on gene abundance and expression in drinking water reservoirs. Water Research 231, 119667. URL: https://www.sciencedirect.com/science/article/pii/S0043135423001021, doi:https://doi.org/10.1016/j.watres. 2023.119667.
- Daniels, O., 2016. Autecology, allelopathy and toxicity of *Limnothrix* (strain AC0243): Multiple-organism studies using laboratory cultures. Ph.D. thesis. University Queensland, Australia.
- Foy, R., 1983. Interaction of temperature and light on the growth rates of two planktonic *Oscillatoria* species under a short photoperiod regime. British Phycological Journal 18, 267–273. URL: https://doi.org/10.1080/00071618300650281, doi:10.1080/00071618300650281, arXiv:https://doi.org/10.1080/00071618300650281.
- Hesse, K., Dittmann, E., B¶rner, T., 2001. Consequences of impaired microcystin production for light-dependent growth and pigmentation of microcystis aeruginosa PCC 7806. FEMS Microbiology Ecology 37, 39–43. URL: https://doi.org/10.1111/j.1574-6941.2001.tb00851.x.
- Jia, Z., Su, M., Liu, T., Guo, Q., Wang, Q., Burch, M., Yu, J., Yang, M., 2019. Light as a possible regulator of MIB-producing *Planktothrix* in source water reservoir, mechanism and *in-situ* verification. Harmful Algae 88, 101658. URL: http://www.sciencedirect.com/science/article/pii/s1568988319301313, doi:10.1016/j.hal.2019.101658.

- Li, M., Nkrumah, P.N., Xiao, M., 2014. Biochemical composition of microcystis aeruginosa related to specific growth rate: insight into the effects of abiotic factors. Inland Waters 4, 357–362. URL: https://www.tandfonline.com/doi/abs/10.5268/IW-4.4.710, doi:10.5268/IW-4.4.710, arXiv:https://www.tandfonline.com/doi/pdf/10.5268/IW-4.4.710.
- Lu, J., Su, M., Su, Y., Wu, B., Cao, T., Fang, J., Yu, J., Zhang, H., Yang, M., 2022. Driving forces for the growth of mib-producing *Planktothricoides raciborskii* in a low-latitude reservoir. Water Research, 118670URL: https://www.sciencedirect.com/science/article/pii/S0043135422006236, doi:10.1016/j.watres.2022.118670.
- Luimstra, V.M., Schuurmans, J.M., Verschoor, A.M., Hellingwerf, K.J., Huisman, J., Matthijs, H.C.P., 2018. Blue light reduces photosynthetic efficiency of cyanobacteria through an imbalance between photosystems I and II. Photosynthesis Research 138, 177–189. URL: https://doi.org/10.1007/s11120-018-0561-5, doi:10.1007/s11120-018-0561-5
- Lürling, M., Eshetu, F., Faassen, E.J., Kosten, S., Huszar, V.L.M., 2013. Comparison of cyanobacterial and green algal growth rates at different temperatures. Freshwater Biology 58, 552–559. doi:10.1111/j.1365-2427.2012. 02866.x.
- Marinho, M.M., Souza, M.B.G., Lürling, M., 2013. Light and phosphate competition between cylindrospermopsis raciborskii and microcystis aeruginosa is strain dependent. Microbial Ecology 66, 479–488. URL: https://doi.org/10.1007/s00248-013-0232-1.
- e. Mello, M.M., Soares, M.C.S., Roland, F., Lurling, M., 2012. Growth inhibition and colony formation in the cyanobacterium microcystis aeruginosa induced by the cyanobacterium cylindrospermopsis raciborskii. Journal of Plankton Research 34, 987–994. URL: https://doi.org/10.1093/plankt/fbs056, doi:10.1093/plankt/fbs056.
- Mohanty, B., Majedi, S.M., Pavagadhi, S., Te, S.H., Boo, C.Y., Gin, K.Y.H., Swarup, S., 2022. Effects of light and temperature on the metabolic profiling of two habitat-dependent bloom-forming cyanobacteria. Metabolites 12, 406. URL: http://dx.doi.org/10.3390/metabo12050406, doi:10.3390/metabo12050406.
- Nicklisch, A., 1998. Growth and light absorption of some planktonic cyanobacteria, diatoms and chlorophyceae under simulated natural light fluctuations. Journal of Plankton Research 20, 105–119. URL: https://doi.org/10.1093/plankt/20.1.105, doi:10.1093/plankt/20.1.105.
- Nicklisch, A., 1999. Competition between the cyanobacterium limnothrix redekei and some spring species of diatoms under p-limitation. International Review of Hydrobiology 84, 233–241. URL: https://onlinelibrary.wiley.com/doi/abs/10.1002/iroh.199900024, doi:https://doi.org/10.1002/iroh.199900024, arXiv:https://onlinelibrary.wiley.com/doi/pdf/10.1002/iroh.199900024.
- Salvador, D., Churro, C., Valério, E., 2016. Evaluating the influence of light intensity in mcyA gene expression and microcystin production in toxic strains of *Planktothrix agardhii* and *Microcystis aeruginosa*. Journal of Microbiological Methods 123, 4–12. URL: https://doi.org/10.1016/j.mimet.2016.02.002, doi:10.1016/j.mimet.2016.02.002.
- Shatwell, T., Nicklisch, A., Köhler, J., 2012. Temperature and photoperiod effects on phytoplankton growing under simulated mixed layer light fluctuations. Limnology and Oceanography 57, 541–553. URL: https://doi.org/10.4319/lo.

- 2012.57.2.0541, doi:10.4319/lo.2012.57.2.0541.
- Sugimoto, K., Negishi, Y., Amano, Y., Machida, M., Imazeki, F., 2015. Roles of dilution rate and nitrogen concentration in competition between the cyanobacterium microcystis aeruginosa and the diatom cyclotella sp. in eutrophic lakes. Journal of Applied Phycology 28, 2255–2263. URL: https://doi.org/10.1007/s10811-015-0754-7, doi:10.1007/s10811-015-0754-7.
- Tang, E.P.Y., Tremblay, R., Vincent, W.F., 1997. CYANOBACTERIAL DOMINANCE OF POLAR FRESHWATER ECOSYSTEMS: ARE HIGH-LATITUDE MAT-FORMERS ADAPTED TO LOW TEMPERATURE?1. Journal of Phycology 33, 171–181. URL: https://doi.org/10.1111/j.0022-3646.1997.00171.x.
- Thomas, M.K., Litchman, E., 2015. Effects of temperature and nitrogen availability on the growth of invasive and native cyanobacteria. Hydrobiologia 763, 357–369. URL: https://doi.org/10.1007/s10750-015-2390-2, doi:10.1007/s10750-015-2390-2.
- Tiwari, O., Prasanna, R., Yadav, A., Wattal, D., Singh, P., 2001. Growth potential and biocide tolerance of non-heterocystous filamentous cyanobacterial isolates from rice fields of uttar pradesh, india. Biology and Fertility of Soils 34, 291–295. URL: https://doi.org/10.1007/s003740100402, doi:10.1007/s003740100402.
- Torres, C.D.A., Lürling, M., Marinho, M.M., 2016. Assessment of the effects of light availability on growth and competition between strains of *Planktothrix agardhii* and *Microcystis aeruginosa*. Microbial Ecology 71, 802–813. doi:10.1007/s00248-015-0719-z.
- Van Der Ploeg, M., Dennis, M., De Regt, M., 1995. Biology of *Oscillatoria chalybea*, a 2-methylisoborneol producing bluegreen alga of Mississippi Catfish Ponds. Water Science and Technology 31, 173–180. URL: https://www.sciencedirect.com/science/article/pii/027312239500473Z, doi:10.1016/0273–1223(95)00473–Z.
- Wiedner, C., Visser, P.M., Fastner, J., Metcalf, J.S., Codd, G.A., Mur, L.R., 2003. Effects of light on the microcystin content of *Microcystis* strain PCC 7806. Applied and Environmental Microbiology 69, 1475–1481. URL: https://doi.org/10. 1128/aem.69.3.1475-1481.2003.
- Willis, A., Chuang, A.W., Woodhouse, J.N., Neilan, B.A., Burford, M.A., 2016. Intraspecific variation in growth, morphology and toxin quotas for the cyanobacterium, cylindrospermopsis raciborskii. Toxicon 119, 307–310. URL: https://doi.org/10.1016/j.toxicon.2016.07.005, doi:10.1016/j.toxicon.2016.07.005.
- Wu, X., Wu, Z., Song, L., 2011. Phenotype and temperature affect the affinity for dissolved inorganic carbon in a cyanobacterium microcystis. Hydrobiologia 675, 175–186. URL: https://doi.org/10.1007/s10750-011-0815-0, doi:10.1007/s10750-011-0815-0.
- Xiao, M., Hamilton, D.P., O'Brien, K.R., Adams, M.P., Willis, A., Burford, M.A., 2020. Are laboratory growth rate experiments relevant to explaining bloom-forming cyanobacteria distributions at global scale? Harmful Algae 92, 101732. URL: https://doi.org/10.1016/j.hal.2019.101732, doi:10.1016/j.hal.2019.101732.
- Xiao, M., Willis, A., Burford, M.A., 2017. Differences in cyanobacterial strain responses to light and temperature reflect species plasticity. Harmful Algae 62, 84–93. URL: https://doi.org/10.1016/j.hal.2016.12.008, doi:10.1016/j.hal.2016.12.008.

Zhang, T., Zheng, L., Li, L., Song, L., 2016. 2-methylisoborneol production characteristics of *Pseudanabaena* sp. FACHB 1277 isolated from Xionghe Reservoir, China. Journal of Applied Phycology , 1–10URL: http://dx.doi.org/10.1007/s10811-016-0864-x. doi:10.1007/s10811-016-0864-x.