

# Unique surface density layers promote formation of harmful algal blooms in the Pengxi River, Three Gorges Reservoir

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**Abstract:** The Three Gorges Reservoir (TGR), China, is the largest man-made reservoir in the world. Harmful algal blooms (HABs) have become common since the reservoir's impoundment in 2003. To investigate the mechanisms of HAB formation in the reservoir and to determine possible mitigation measures, we conducted surveys over a range of spatial scales and temporal resolutions over a 2-y period (March 2013–December 2014). The large-scale survey (the portion of the reservoir on the main stem of the Yangtze River and 22 tributaries) revealed that cyanobacteria blooms were restricted to the upper reaches of the tributaries. The medium-scale survey (1 tributary: Pengxi River) showed that cyanobacteria blooms were confined to the early-spring period with the initiation of thermal stratification in the deep-water column. The small-scale survey (a local, backwater lake in the Pengxi River), which was of higher-temporal resolution than the other 2 surveys, demonstrated that the bloom occurred at the same time as the formation of a surface-density layer unique to the geomorphology and water-control management of the reservoir. The vertical distributions of the bloom and surface-density layer appeared to be related, although the density layer persisted beyond the duration of the HABs. We hypothesized that limited nutrient diffusion into these density layers could result in nutrient limitation despite the hyper-eutrophic conditions that generally characterize the TGR basin. In the main stem of the Yangtze River and lower reaches of the tributaries in the TGR, algal blooms were not observed because of continuous, deep mixing throughout the year. We conclude that the hydrological stability and geomorphological characteristics of the TGR play critical roles in regulating the temporal and spatial patterns of algal blooms and that artificial mixing of the water column is currently the best option to limit HAB formation, especially in upper tributaries.

**Key words:** Three Gorges Reservoir, geomorphology, harmful algal blooms, surface-density layers, nutrients

There are 16.7 million reservoirs >0.01 ha (surface area) on Earth (Lehner et al. 2011), and another 3700 major dams are either planned or under construction (Zarfl et al. 2015). Water-flow-control programs have fragmented over ½ of

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the large rivers on Earth (Wu et al. 2019). Reduced connectivity of rivers resulting from dam construction changes both physicochemical conditions and habitat utilization. For example, changed hydrological conditions, sediment transport, and nutrient stoichiometry have been suggested as critical triggers of water-quality deterioration and the development of algal blooms in the impounded areas. The occurrence of harmful algal blooms (HABs) has raised critical concerns among the public, scientists, and policymakers about both human and environmental health (Jeong et al. 2014). Understanding the causes of such threats is needed so that sustainable management strategies can be implemented to protect the surface waters of the planet.

The Three Gorges Reservoir (TGR) in China is the largest hydroelectric and flood control reservoir in the world, producing an annual electrical output of 85 billion kWh. This energy source allows for a reduction of 50 million Mg of coal consumption, preventing 120 million Mg of CO<sub>2</sub> and 10,000 Mg of CO emissions/y (Zhang 2014). Completed in 2009, the impoundment of the reservoir resulted in a 100-m water-level rise, reaching 175 m above sea level (ASL). This project transformed a 660-km reach of the Yangtze River into a highly-regulated reservoir. At high water levels, the serpentine reservoir contains 40 billion m<sup>3</sup> of water with an increased water-surface area of 645 to 1200 km<sup>2</sup> (Fu et al. 2010). There are >40 major tributaries (watershed areas >100 km<sup>2</sup>) flowing directly into the TGR (Ji et al. 2017). These characteristics provide a unique opportunity to study how dam construction and water-flow management can result in novel threats to human and environmental health.

Before the construction of the dam, algal blooms were not common in the TGR basin (Zeng et al. 2007), but following construction, the frequency and magnitude of HABs have intensified and become a serious concern (Cai and Hu 2006, Zeng et al. 2007, Li et al. 2011, Fu et al. 2015, Zhang et al. 2015). Many studies have attributed these blooms to excessive nutrient inputs to the TGR (Cai and Hu 2006, Zeng et al. 2006, 2007, Ye et al. 2007, Zhang et al. 2010, Gao et al. 2016). The elevated concentrations of nutrients such as total nitrogen (TN), total phosphorus (TP), and silica (Si), are 2 to 3× higher than the reported threshold of eutrophication (Thomann and Mueller 1987) as a result of high population densities and intensive agriculture in the TGR basin.

Decreased water velocities in tributaries have also been linked to the formation of HABs (Wang 2012) as has the development of stable water columns (Davis and Koop 2006, Yang et al. 2010, Liu et al. 2012, Conroy et al. 2014, McKay et al. 2018). As a result of flow control, water velocities in the tributaries of the TGR have declined from 0.5–1 to 0.05 m/s. The main channel, however, has maintained a constant flow of 1 m/s (Wang 2012). A study in the Daning River demonstrated that phytoplankton biomass was positively related to relative water-column stability (Zhu et al. 2013). Regulation of daily water-level fluctuations has been

used by the reservoir managers to reduce the relative water-column stability to mitigate HABs in some tributaries (e.g., Xiangxi River) but with limited success (Zheng et al. 2011, Yang et al. 2013, Sha et al. 2015, Ji et al. 2017). Similar approaches modifying water-column stability have proven effective in controlling HABs in a relatively-small reservoir in Spain (Rigosi and Rueda 2012). However, the size of the TGR (660 km long and an average of 1 km wide) limits the use of water flow to manage water-column stability as an approach for HAB control. Water-level fluctuations in tributaries far from the dam are relatively minor.

Only a few studies have integrated nutrient dynamics with spatial and temporal variability of hydrological conditions to address the problem of HABs in the TGR. These past studies have been focused on limited spatial scales, e.g., single tributary (Zheng et al. 2011, Yang et al. 2013, Sha et al. 2015, Ji et al. 2017), and to date, no full-reservoir studies have been reported. This study aims to provide a spatially-integrated investigation of HABs in the TGR at the basin, tributary, and upper-reach scales. Specifically, we address the relative importance of nutrient concentrations and hydrological variability as regulators of where and when HABs develop in the reservoir. We hypothesize that differing hydrological conditions and water quality in the main channel, tributaries, and upper reaches have the potential to regulate the development of HABs. We predict that algal blooms reflect unique combinations of nutrients and hydrological conditions within the morphologically-complex reservoir. To achieve these goals, we conducted several surveys on different spatial and temporal scales: 1) a large-scale investigation of the entire TGR basin across both low and high water-level seasons to determine the overall spatial distribution of HABs, 2) a higher spatial-resolution surveys in a tributary of the TGR to investigate factors (nutrient conditions and water-column stratification) that regulate the timing and spatial distribution of HABs, and 3) a temporally-intensive study in the upper reach of the same tributary to characterize its chlorophyll *a* (Chl *a*) concentration and hydrology at the time of algal blooms.

## METHODS

To address our research objectives, we conducted field surveys at 3 different spatial scales (basin, tributary, and upper-reach) and 2 different temporal resolutions (2 sampling periods over 1 y and biweekly sampling periods over 13 mo). We characterized physical and chemical components including hydrological data, total and bioavailable nitrogen and phosphorus concentrations, Chl *a* concentrations, and the phytoplankton community. The 3 studies overlapped temporally from March 2013 to June 2014. Distribution of algal blooms and eutrophication among all 77 monitoring sites of the Ministry of Environmental Protection in the TGR remained similar between 2013 and 2014 (Ministry of Environmental Protection, China 2013, 2014). In

addition, the climate was relatively stable during the study period, according to the climate report of Yunyang County (Table S1; CMDC 2015), the location of the Pengxi River, which makes the data in these 2 sampling years comparable.

**Timing and spatial distribution of HABs at the basin scale**

To characterize the timing and spatial distribution of HABs across the entire TGR (large spatial scale), we con-

ducted surveys across the TGR, which is located between lat 29°41'43"–31°03'57"N, long 106°58'38"–110°55'55"E in the middle reach of the Yangtze River (Fig. 1A). This area has a monsoon-influenced humid subtropical climate with 4 distinct seasons. Average annual precipitation is 1000 to 1350 mm. Monthly precipitation during March through August, when the reservoir stores water, is 200 to 300 mm and, during the rest of the year, 100 to 150 mm (Holbach et al. 2014). The wind speed on the surface of the reservoir

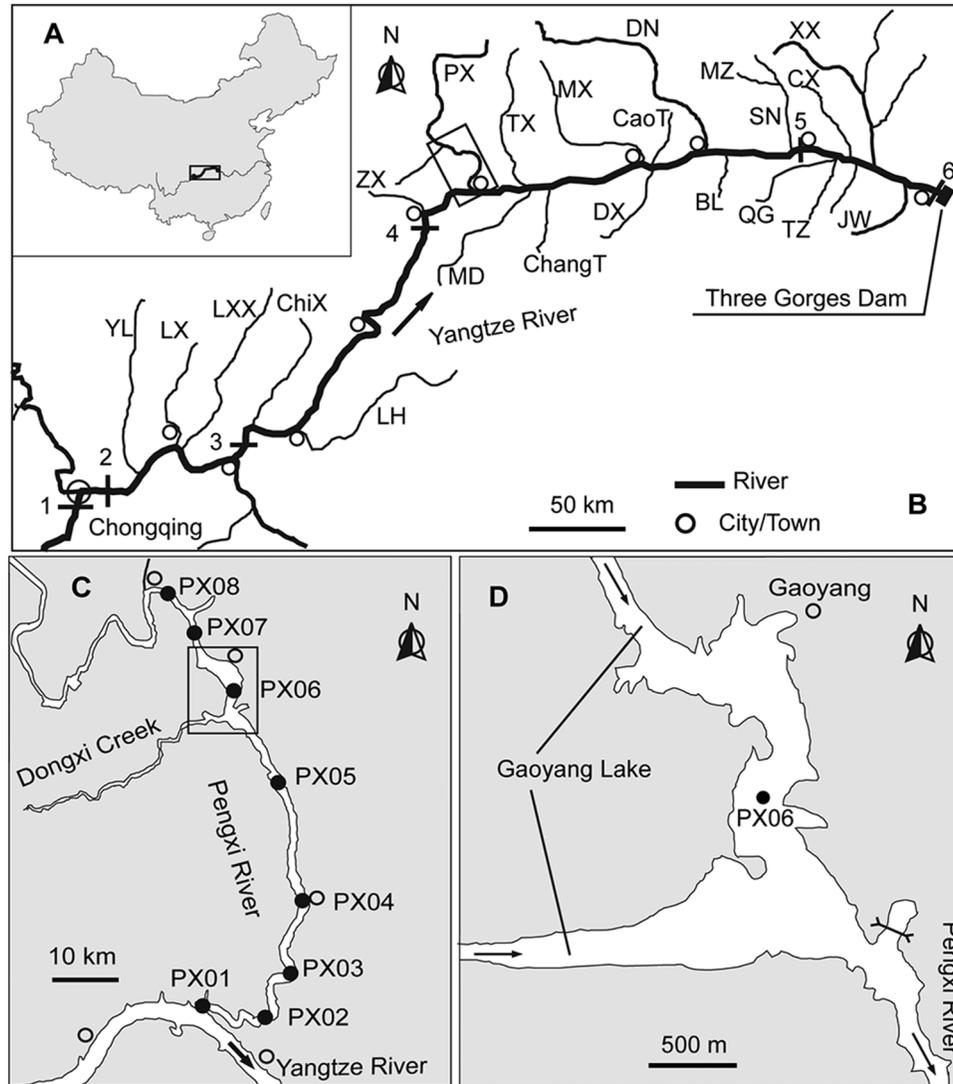


Figure 1. Map of sampling sites showing the location of the study area in China (A), sampling sections in the main stem (black bars) of the Three Gorges Reservoir and the 22 tributaries (see below for the full list of names) sampled in the basin-scale survey (B), sampling sites (black dots) in the Pengxi River (PX) in the tributary-scale survey (C), and sampling site (black dot) in the upper-reach-scale study with high temporal resolution in Gaoyang Lake (D). Arrows show direction of river flow. The full names of tributaries are (from left to right in panel B): YL, Yuling River; LX, Longxi River; LXX, Lixiangxi River; ChiX, Chixi River (池溪河)\*; LH, Longhe River; ZX, Zhuxi River; PX, Pengxi River; TX, Tangxi River; MD, Modao River; ChangT, Changtan River; MX, Meixi River; CaoT, Caotang River; DX, Daxi River; DN, Daning River; BL, Baolong River; SN, Shennong River; MZ, Meizhuxia River; QG, Qinggan River; TZ, Tongzhuang River; CX, Chixi River (叱溪河)\*; XX, Xiangxi River; JW, Jiuwanxi River.

\*The names of these 2 rivers are written and read differently in the local language, but they are translated as the same word in English. We added their Chinese local names so readers can tell the difference.

is very low because of the steep sides and ranges from 0.9 to 2.1 m/s (Wang 2012). The average annual water temperature of the reservoir is 16 to 18°C, but extreme surface water temperatures of >35°C have been reported during the summer (Yang et al. 2017) as a result of high humidity and decreased surface evaporation. Water levels of the reservoir are managed to maintain a minimum water level of 145 m ASL in May through August for transportation and flood-control purposes and a maximum of 175 m ASL in September through April for electricity generation (Holbach et al. 2014).

We conducted 2 reservoir-wide surveys in 2013: 1 during the high-water phase (8–25 March 2013) and 1 during the low-water phase (9–25 September 2013). In March, we sampled 6 transects along the main stem of the Yangtze River (hereafter, main stem) and 22 major tributaries (Fig. 1B). In September, we resampled the same main-stem transects and 17 of the 22 previously-sampled tributaries. The remaining 5 tributaries were inaccessible because of low water levels. We sampled each tributary at the inflow where flow was continuous throughout the year, in the mid-reach where backwater conditions (low-velocity flow caused by a backup effect from the main stem) occur during the filling of the reservoir, and at the confluence at 0.5 km upstream from the main stem. Specifically, we sampled 1 transect in the inflow, 1 transect at the confluence, and 1 to 3 equally-spaced transects in the backwater areas based on their length, yielding a total of 3 to 5 transects/tributary. Exceptions to this sampling scheme were the Modao River (MD), Changtan River (CT), Meixi River (MX), and Tongzhuang River (TZ) (Fig. 1B), in which we only sampled the backwater and confluence because inflow stations were not accessible.

To characterize HABs and chemical and physical characteristics of the waters in the TGR, we collected water samples for nutrient and phytoplankton analyses from 0.5 m under the surface. For tributaries with widths <50 m, 50–100 m, and >100 m, we collected 1, 2, and 3 water samples, respectively. We pooled the multiple samples taken from the wider individual tributaries. We used a data sonde (model 6600 V2; Yellow Springs Incorporated, Yellow Springs, Ohio) to record in-situ water temperature, dissolved oxygen, water depth, Chl *a*, conductivity, pH, and turbidity. We used a Secchi disk to measure water transparency (Tilzer 1988) and a hydrometric propeller (model LJ12-1A; Shengrong Facility Limited, Nanjing, China) to measure water velocity. We recorded the weather, wave height, and visible algal blooms at the time of sampling. We collected a 500-mL sample for phytoplankton identification and enumeration at each transect and fixed samples on site with Lugol's iodine. We enumerated phytoplankton (at 400× magnification) in a counting chamber (Hamilton et al. 2001), and we used the keys in Hu and Wei (2006) to identify phytoplankton to genus level. We used spectrophotometric measures to measure water chemistry including TN, TP, am-

moniacal nitrogen (NH<sub>3</sub>-N), orthophosphate (PO<sub>4</sub>-P), and permanganate index (COD) immediately on board the sampling vessel, so no special preservation was required. Water samples were digested for measuring TN and TP (Molen et al. 1994). Before analysis we used 0.45-μm-pore, glass-fiber membranes to filter water samples for NH<sub>3</sub>-N and PO<sub>4</sub>-P (Rice et al. 2012). We calculated retention time, which is the ratio of storage capacity to flow:  $RT = V_i/Q_i$ , where  $RT$  is the retention time,  $V_i$  is the storage capacity, and  $Q_i$  is the average monthly flow (Quinn 1992). We obtained the average monthly flow from Wang (2012), and we calculated the storage capacity as the product of the TGR area (Xu 2018) and average depth (Jiang 2017).

To identify environmental factors possibly associated with the algal blooms observed in the backwater areas in March, we used a generalized linear model in the MASS package (Venables and Ripley 2002) in R (version 3.6.2; R Project for Statistical Computing, Vienna, Austria) to relate Chl *a* concentration (response variable) to physicochemical parameters (explanatory variables; Tables 1, S4). We used bi-directional model selection based on the Akaike information criterion to determine the reduced model (Giri et al. 2018). In addition, we used SPSS<sup>®</sup> software (version 17.0; IBM Corporation, Armonk, New York) to run correlation analysis and identify the relationships between concentrations of dissolved and total nutrients (NH<sub>3</sub>-N ~ TN, PO<sub>4</sub>-P ~ TP) for March and September 2013.

### Timing and spatial distribution of HABs at the tributary scale

To investigate associations between nutrient conditions and water-column stratification with the timing and spatial distribution of HABs, we conducted a study at a medium spatial scale in the Pengxi River. We selected this river (lat 30°57'03.8"–31°11'61.5"N, long 108°24'00"–108°42'39.5"E) because it is the largest tributary on the northern bank of the TGR (5173-km<sup>2</sup> watershed, 182 km long; Fig. 1C). The Pengxi River's confluence is 247 km from the Three Gorges Dam and has an average slope of 1.25%. Annual precipitation in this watershed is 1100 to 1500 mm, occurring primarily from May to September (Jiang 2017).

Algal blooms have frequently been reported in the Pengxi River following the completion of the dam (Wang 2012, Jiang 2017, Jiang et al. 2017). To determine the timing and spatial distribution of the algal blooms in this tributary, we sampled 8 transects from the confluence to the upper reach in the mornings of 17 April 2013 during the high-water phase and 27 July 2013 when the water level dropped to 145 m ASL. We used an RBRmaestro<sup>3</sup> data logger (RBR Limited, Ottawa, Ontario, Canada) to measure in-situ water temperature, water depth, conductivity, and Chl *a* throughout the water column. We used a Secchi disk to measure water transparency (Tilzer 1988) at each transect and calculated euphotic depth ( $Z_{eu}$ : a measurement of the

Table 1. Hydrological and water-quality parameters of the backwater sections in tributaries from upstream to downstream of the Three Gorges Reservoir, China, during the basin-scale survey in March 2013. Chl *a* = chlorophyll *a*, Temp = temperature, TP = total phosphorus, TN = total nitrogen, PO<sub>4</sub>-P = orthophosphate as phosphorus, NH<sub>3</sub>-N = ammoniacal nitrogen, LogA = log<sub>10</sub> (algal density), Cond = conductivity, Turb = turbidity, NTU = Nephelometric Turbidity Units, DO = dissolved oxygen, COD = permanganate index, NA = not available.

Tributary	Chl <i>a</i> (µg/L)	Temp (°C)	pH	TP (mg/L)	TN (mg/L)	PO <sub>4</sub> -P (mg/L)	NH <sub>3</sub> -N (mg/L)	LogA	Cond (µS/cm)	Turb (NTU)	DO (mg/L)	COD (mg/L)
Yuling River	24	18.1	8.5	0.073	1.58	0.005	0.035	6.70	440	2.3	10.92	4.16
Longxi River	7.2	17.8	8.0	0.084	1.75	NA	0.108	7.74	451	1.9	9.76	4.42
Lixiangxi River	30.8	17.2	8.8	0.116	1.39	NA	0.034	7.04	274	4.2	12.15	4.26
Chixi River	2.7	17.0	7.8	0.184	1.85	NA	0.164	5.44	573	0.7	8.19	2.26
Longhe River	1.7	16.0	7.9	0.059	1.73	0.005	0.187	5.40	358	2.3	9.81	1.46
Zhuxi River	22.5	19.8	8.1	0.691	6.21	0.554	3.395	6.72	374	4.2	7.97	6.50
Pengxi River	21.9	17.2	8.7	0.133	2.06	0.046	0.038	6.55	444	4.2	13.04	4.34
Tangxi River	8.2	17.4	8.2	0.039	1.09	NA	0.056	5.83	350	3.3	11.18	1.69
Modao River	1.4	15.8	8.3	0.061	1.54	0.052	0.145	5.92	402	1.6	10.81	1.90
Changtan River	2.0	15.4	8.4	0.041	1.07	0.005	0.022	5.84	418	6.6	12.63	2.12
Meixi River	38.8	15.8	8.9	0.273	2.07	0.027	0.082	5.90	344	1.6	12.80	5.56
Caotang River	82.2	15.7	9.2	0.306	2.76	NA	0.015	7.49	250	4.2	18.54	4.46
Daxi River	0.3	14.3	8.2	0.018	1.81	NA	0.015	5.21	315	1.2	17.85	1.36
Daning River	3.9	14.1	8.3	0.019	1.29	0.005	0.058	5.95	401	0.9	10.98	1.77
Baolong River	70.9	14.5	8.4	0.103	1.31	NA	0.015	6.85	368	11	12.60	4.68
Mianzhuxia River	18.3	15.1	7.7	0.089	0.30	0.020	0.032	6.79	1045	4.3	8.84	2.17
Shennong River	27.4	13.1	8.3	0.041	0.46	0.011	0.016	6.75	373	1.4	13.92	2.85
Qinggan River	35.6	13.4	8.5	0.046	1.12	0.032	0.021	6.55	284	5.7	14.63	3.50
Chixi River	52.9	11.6	8.5	0.532	3.62	0.071	0.024	7.34	312	12.3	11.74	11.05
Tongzhuang River	5.9	12.9	8.2	0.094	1.89	0.054	0.026	7.36	392	4.1	12.12	1.75
Xiangxi River	55.6	13.7	8.7	1.140	1.67	0.753	0.093	6.81	337	4.2	13.99	6.51
Jiuwanxi River	3.0	14.0	8.1	0.061	1.93	0.036	0.058	5.08	375	3.0	10.38	1.29

depth at which 1% of the photosynthetically-active radiation at the surface remains) as  $2.7 \times$  the Secchi depth, in accordance with Tilzer (1988) and Zhang et al. (2012). We calculated retention time as described for the basin-scale study. At each transect we collected three 1-L water samples across the river at a depth of 0.5 m. We transported water samples on ice to the laboratory within 3 h of collection and stored them at 4°C before chemical analysis for TP, TN, NH<sub>3</sub>-N, and nitrate nitrogen (NO<sub>3</sub>-N). We performed chemical analysis within 48 h of collection (as described in Rice et al. 2012). As in the large-scale study, water samples were digested for measuring TP and TN (Molen et al. 1994), and we used 0.45-µm-pore, glass-fiber membranes to filter water samples for NH<sub>3</sub>-N and NO<sub>3</sub>-N before digestion (Rice et al. 2012). We identified phytoplankton to genus level as described for the basin-scale study.

We used Surfer (version 15; Golden Software, Golden, Colorado) to visualize the spatial distribution of Chl *a* in the Pengxi River based on data collected by the RBR data logger readings. We used paired-sample *t*-tests in SPSS

to compare concentrations of TN and TP between April and July 2013. An algal bloom occurred in April 2013 (see the results of the upper-reach-scale study for details), and we used simple linear regressions in SPSS to relate the concentration of Chl *a* (i.e., biomass) to nutrient concentrations (TP, TN, NH<sub>3</sub>-N, and NO<sub>3</sub>-N).

#### Timing and distribution of HABs at the tributary upper-reach scale

To characterize HABs and hydrology at a small spatial scale, we conducted a study with high temporal resolution on Gaoyang Lake (lat 31°05'27.4"N, long 108°40'41.6"E; Fig. 1D). The lake was formed in 2003 as a result of increased water level associated with the Three Gorges Dam. Water velocity in the lake was up to 0.32 m/s from May to August but decreased to 0.02 to 0.05 m/s from September to April 2011, changing this reach of the Pengxi River into a riverine lake (Wang 2012).

We performed biweekly water-column sampling from 21 May 2013 to 30 June 2014. We used the RBRmaestro<sup>3</sup>

Table 2. Water velocity and retention time in the main stem of the Yangtze River and the Pengxi River in the Three Gorges Reservoir, China, in 2013.

Variable	Locations	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Velocity (m/s)	Main stem	0.18	0.2	0.24	0.31	0.36	0.71	0.81	0.8	0.79	0.4	0.24	0.2
	Pengxi River	0.015	0.02	0.02	0.02	0.21	0.21	0.21	0.21	0.02	0.015	0.015	0.015
Retention time (d)	Main stem	122.2	120.2	108.2	70.6	49.8	30.5	13.4	12.9	16.1	41	57.9	114.4
	Pengxi River	356.9	459.7	195.2	48.0	15.4	12.9	20.9	33.8	28.7	55.5	123.3	246.5

data logger to measure in-situ water temperature, water depth, conductivity, and Chl *a* throughout the water column. We collected water samples from the surface, mid-depth, and bottom according to the thermal profile of the water column. Specifically, we collected a 1-L water sample at 0.5 m below the surface and another sample at 0.5 m above the lake bottom to represent the surface and hypolimnion, respectively. We collected a mid-water-column sample from either the depth of maximum temperature change during stratified periods or from the mid-depth of the water column during isothermal periods. Water transparency and euphotic depth ( $Z_{eu}$ ) were determined as described previously. We conducted preservation and transportation as in the tributary-scale study and analyzed Chl *a* in the lab (Rice et al. 2012).

## RESULTS

### Basin-scale study

As a result of water level regulation in the TGR, water velocity in the main stem during March 2013, when water levels were beginning to decline, was 0.24 m/s, which maintained isothermal conditions in the water column. In contrast, water flows in the upper reaches of the Pengxi River, as an example of the tributaries, during this time period were <0.02 m/s (Table 2). In September 2013, the end of the drawdown period, water velocity in the main stem had increased to 0.79 m/s. In the Pengxi River, water velocity increased to ~0.2 m/s in the backwater reaches in summer months but returned to ~0.02 m/s in September. Water-retention time in the main stem during the high-water-level months (i.e., January–March and November–December) ranged from 57.9 to 122.2 d. Retention time in the Pengxi River varied considerably because of changes in velocity and water level throughout the year and ranged from 123.3 to 459.7 d. The monthly retention time of the Pengxi River was 1.3 to 3.8× that in the main stem except during April, May, and June as a result of water discharge at the Three Gorges Dam (Table 2).

Surficial algal blooms were visible scums in 15 of 22 (68%) and 13 of 17 (76%) of the tributaries in March and September, respectively. Most of the March blooms (87%) and September blooms (93%) occurred in backwater areas of the tributaries (Tables S2, S3). Elevated Chl *a* (March: 20–25  $\mu\text{g/L}$ ; September: 9–15  $\mu\text{g/L}$ ) and algal den-

sities (> $10^6$  cells/L in both periods) were restricted to the backwater sections of the tributaries (Fig. 2). Furthermore, algal biomass, measured as Chl *a*, was up to 3× higher in March than in September (Fig. 2, Tables S2, S3). In contrast, transects on the main stem of the reservoir did not exhibit high algal biomass (Chl *a* < 1.5  $\mu\text{g/L}$ ) in either sampling season (Fig. 2, Tables S2, S3).

Concentrations of TN (>1.5 mg/L), TP (>0.1 mg/L), and the dissolved inorganic nutrients  $\text{NH}_3\text{-N}$  (0.01–2.718 mg/L), and  $\text{PO}_4\text{-P}$  (0.013–0.482 mg/L) were consistently high throughout the TGR, including the main stem and tributaries, during both the March and September surveys (Fig. 2, Tables 1,

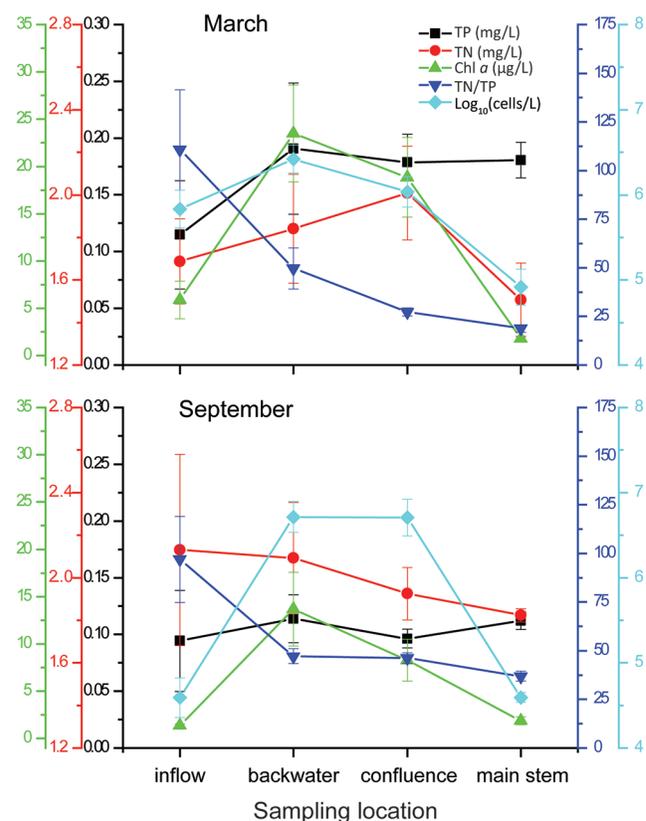
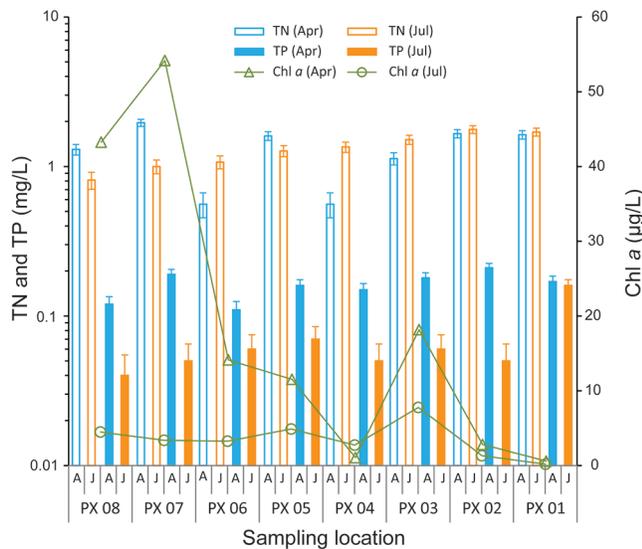


Figure 2. Concentration (mean  $\pm$  SD) of nutrients and Chl *a* and density (mean  $\pm$  SD) of algal cells in the inflow, backwater, and confluence of tributaries and the main stem of the Three Gorges Reservoir in March and September 2013. TN = total nitrogen, TP = total phosphorous. TN/TP is given as molar ratio.

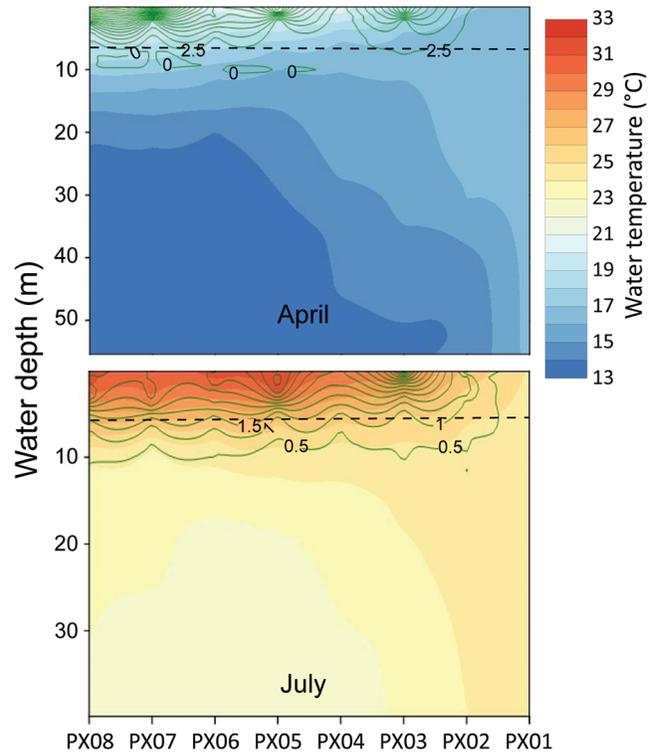
S2, S3). At all sampling sites, concentrations of  $\text{NH}_3\text{-N}$  and  $\text{PO}_4\text{-P}$  were strongly positively related to TN and TP, respectively, in both March (Pearson's  $r = 0.754\text{--}0.831$ ,  $p < 0.01$ ) and September (Pearson's  $r = 0.596\text{--}0.926$ ,  $p < 0.001$ ). TN/TP molar ratios were  $>16$  and decreased from the inflows to the main stem in all tributaries during both sampling periods (Fig. 2). Generalized linear modeling with 11 physicochemical parameters (Table 1) as explanatory variables demonstrated that TN,  $\text{NH}_3\text{-N}$ , TP, and  $\text{PO}_4\text{-P}$ , which were removed from the reduced model during model selection, were not closely related to Chl  $a$  (dependent variable;  $p = 0.883\text{--}0.947$  in the initial model; Table S4) in the backwaters in March when algal blooms were most pronounced.

**Tributary-scale study**

Water velocity in the Pengxi River was relatively low in April 2013, ranging from 0.01 to 0.04 m/s, but increased to 0.16 to 0.25 m/s in July as the reservoir reached minimum water levels. Water temperatures along the tributary revealed the development of a warm, upper-surface layer in April that became much more intense by July (Fig. 3). Interestingly, nowhere along the reach of the tributary was there a surface mixed layer (epilimnion) even though a strong thermal structure was evident. The maximum surface-water temperature in July ( $32^\circ\text{C}$ ) was  $\sim 10^\circ\text{C}$  higher than in April (Fig. 3), and it is possible these warm temperatures limited algal growth. The  $Z_{\text{eu}}$  declined from 5 m in April to 2.5 m in July. Concentrations of TN (0.56–1.96 mg/L) and dissolved inorganic nitrogen ( $\text{NH}_3\text{-N}$  and  $\text{NO}_3\text{-N}$ ;



**Figure 3.** Chl  $a$  ( $\mu\text{g/L}$ ) distribution contours (green lines) and temperature contours in the water column of the Pengxi River (PX), Three Gorges Reservoir, China, in April and July of 2013. Dashed line in each panel indicates approximate bottom of surface density layer.



**Figure 4.** Spatial distribution of TN, TP, and Chl  $a$  in the Pengxi River, Three Gorges Reservoir in April (A) and July (J) of 2013, showing results across sampling sites (PX01–PX08). Error bars indicate 1 standard deviation of the mean. TN = total nitrogen, TP = total phosphorous. The y-axis for TN and TP is scaled by  $\log_{10}$ .

0.07–0.53 mg/L) were high and varied along the tributary reach (Fig. 4, Table S5). Concentrations of TP were  $>0.1$  mg/L and were relatively constant along the length of the tributary in April (Fig. 4). TP concentrations were higher in April than in July ( $t_7 = 5.75$ ,  $p < 0.001$ ), but there was no clear difference in TN ( $t_7 = -0.04$ ,  $p = 0.969$ ) between the 2 mo. There were no strong relationships between Chl  $a$  and nutrients (TP, TN,  $\text{NH}_3\text{-N}$ , or  $\text{NO}_3\text{-N}$ ; linear regressions:  $r^2 = -0.03\text{--}0.27$ ; analysis of variance to test fit of linear regressions to the data:  $F_{1,6} = 0.06\text{--}2.24$ ,  $p = 0.185\text{--}0.807$ ) in either April or July.

An algal bloom in the upper reaches of the Pengxi River was initiated in April 2013 and extended into May 2013, with peak Chl  $a$  concentrations of  $50 \mu\text{g/L}$ . The sampling sites near the confluence with the main stem had much lower Chl  $a$  concentrations (Fig. 4, Table S5). The concentration of Chl  $a$  declined to  $\sim 5 \mu\text{g/L}$  throughout the tributary in July (Figs 3, 4). The phytoplankton assemblage was dominated by cyanobacteria *Microcystis* spp., *Anabaena* spp., *Dolichospermum* spp., and the dinoflagellate *Ceratium hirundinella* (Table S6). This algal biomass was mainly confined to the upper-surface layer extending down to a depth of 7 m (dashed line, Fig. 3 April). Similar to the pattern observed in April, the algal biomass in July was mainly confined

to the upper-surface layer (dashed line at 5.5-m depth, Fig. 3 July) but with much lower concentrations ( $\text{Chl } a \leq 7.8 \mu\text{g/L}$ ).

Conductivity–depth profiles in April revealed 2 distinct water masses in the Pengxi River, with the upper waters having higher conductivity ( $>550 \mu\text{S/cm}$ ) than the lower ( $<450 \mu\text{S/cm}$ ) in whole water columns (Fig. 5). Interestingly, conductivity in upper-reach sites (PX06–08) were greatest at  $\sim 7\text{-m}$  depth (dashed line), defining strong vertical structuring in the upper layers of the water column. The lower-reach conductivity profiles, however, revealed well-mixed conditions. Conductivity in July was generally lower than in April, ranging from 245 to  $430 \mu\text{S/cm}$  but with strong vertical structuring, i.e., peaking at  $\sim 5\text{ m}$  deep (dashed line) at all sites except the lower 2 stations (PX01, PX02), which were mostly regulated by inflows from the main stem.

### Upper-reach-scale study

The annual thermal structure of the water column in Gaoyang Lake (Fig. 6A–F) showed that the lake was thermally stratified from March to September (Fig. 6A–C) despite increased water flow with declining water levels. The dam began to store water in late September, which resulted in a well-mixed system in the lake for the rest of the year (Fig. 6B–C). Thermal stratification increased from its initiation in April until the middle of July when the surficial water temperature was at its highest ( $\sim 35^\circ\text{C}$ ) of the year. Most interesting was the lack of a surface mixed layer (epilimnion) during most of the year except September, supporting the observations of no epilimnion in the medium-scale study. These surface-temperature patterns resulted in the formation of strong **SDLs** when water was stratified. For ex-

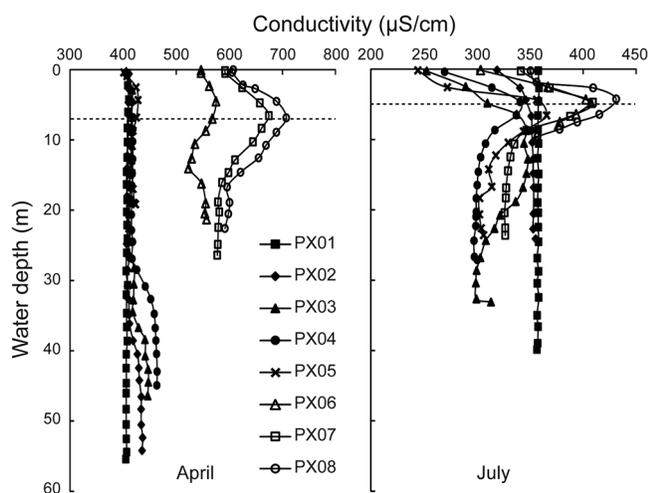


Figure 5. Water conductivity at different depths throughout the water column in the Pengxi River (PX), Three Gorges Reservoir, China, in April and July of 2013. Dashed lines show the approximate bottom of the surface density layer. Note: the x-axis scales differ between panels.

ample, the change in water density from  $30$  to  $33^\circ\text{C}$  is  $\sim 0.95 \text{ kg/m}^3$ , whereas water-density change from  $4$  to  $8^\circ\text{C}$ , commonly observed in lakes, is  $0.12 \text{ kg/m}^3$  (Tanaka et al. 2001). As shown by the conductivity profiles, the intensive changes happened in shallower water ( $<7\text{ m}$  deep) when the water column was stratified (Fig. 6D–F). Given that conductivity is an indicator of electrolyte (ions and soluble minerals) concentrations, we infer there was limited exchange of dissolved salts or nutrients between the SDL and the underlying water column.

The SDL that formed in Gaoyang Lake contained the highest algal biomass (Fig. 7) out of all layers and time periods. During the 13 mo of observation, the highest  $\text{Chl } a$  concentration in the SDL was observed in May of both years ( $>180 \mu\text{g/L}$  in May 2014) and then declined in summer. By late autumn, with the development of isothermal conditions,  $\text{Chl } a$  declined to  $<5 \mu\text{g/L}$ .

### DISCUSSION

The rapid development of dams has reduced the number of our planet's remaining free-flowing, large rivers by 21% (Zarfl et al. 2015). The ecological impacts of dams on water quality and the development of HABs is a topic of concern. Through a multi-scale study in the TGR, we aimed to identify the associations between excessive nutrients and decreased water velocity in the reservoir basin with the formation of HABs. By demonstrating HAB distributions, water quality, and hydrodynamic conditions throughout the TGR, this study reveals that the development of HABs does not always correlate with nutrient distributions, nor are HABs confined to periods of maximum temperature and water stratification. On the other hand, our discovery of the seasonal SDL in a tributary of the TGR offers implications for management of HABs in reservoirs.

### Algal blooms and hydrodynamic conditions in the TGR

We observed that algal blooms in the TGR were mainly confined to the mid and upper reaches of the tributaries, and they were not found in the continuously-flowing upper inflows or the confluence with the main stem. There is little evidence that this spatial pattern of bloom development is related to the levels of total and bioavailable nutrients found throughout the reservoir. Instead, the spatial distribution and timing of algal blooms appears to be a function of water-column stability regulated by both the dam and by the unique morphology of the reservoir, which restricts wind mixing. The operation of the dam leads to varying water velocities and retention times at different geographic locations, especially in the upper reaches of the tributaries. The initiation and distribution of algal blooms were consistently observed in the 3 different spatial-scale studies in the upper reaches of the tributaries during the spring period (Tables S2, S3, S4, Figs 3, 4, 7). These algal blooms

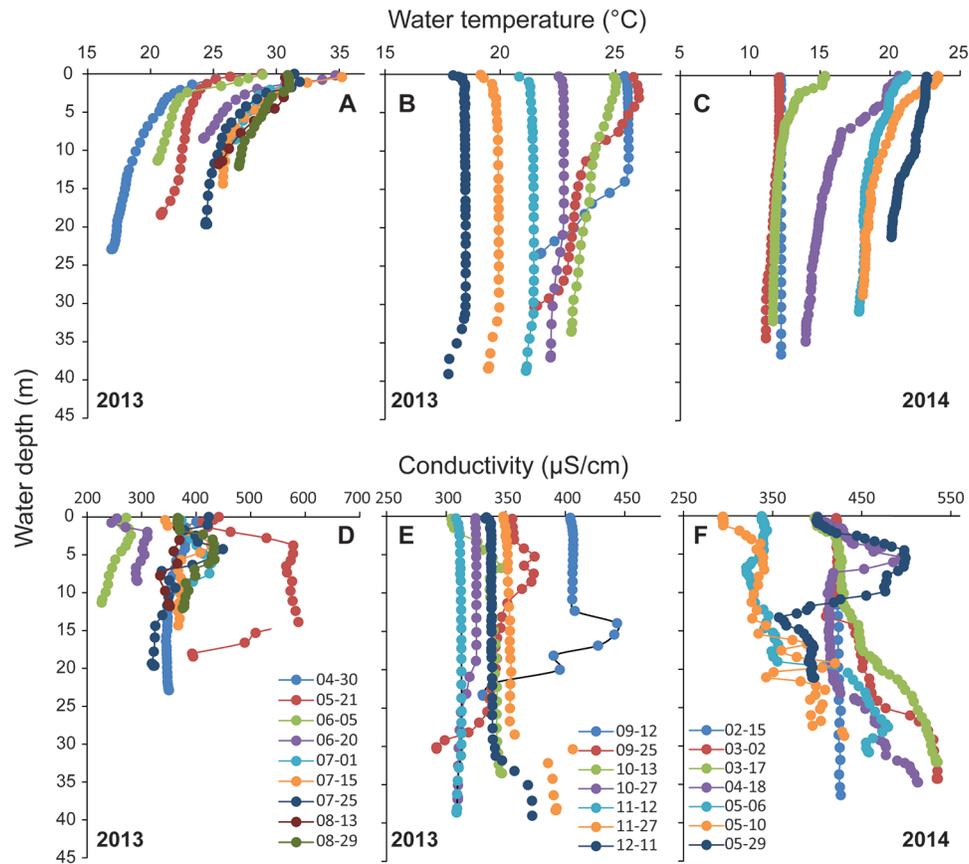


Figure 6. Water temperature (A–C) and conductivity (D–F) at different depths throughout the water column in Gaoyang Lake, Pengxi River, Three Gorges Reservoir, China, from 30 April 2013 to 29 May 2014. Legends give the sampling dates as mm–dd (e.g., 04–30 for 30 April). Each panel pair (i.e., A and D; B and E; C and F) denotes the same sampling period.

corresponded to times with low flow velocities and longer retention times (Table 2) because of water-level controls in TGR.

The size, vertical distribution, and duration of the blooms, however, were more influenced by the formation of the SDL, as observed in the upper reaches of the Pengxi River. The formation of the SDL, although dependent on water-flow control, was also a result of the unique morphology of the TGR. Over 95% of the TGR region is mountainous (Peng et al. 2006) with large elevation differences (~2000 m) between the water surface and mountain peaks (Li et al. 2001), resulting in very low wind speeds over the surface of the reservoir (e.g., 0.9–2.1 m/s; Yang et al. 2017). These characteristics, coupled with limited fetch at the water surface (<1 km), can prevent the development of breaking waves and the formation of an epilimnion. Thermal stratification patterns observed in tributaries, such as the Gaoyang Lake, do not conform to those typically observed in temperate lakes, where there is often a mixed epilimnion. In temperate-lake systems, algal blooms are frequently associated with periods of maximum heat content in late summer (Paerl and Huisman 2008). In the TGR, however, the formation of algal blooms relies on the initial development of an SDL.

The importance of stratification for the development of algal blooms has been reported previously. Davis and Koop (2006) concluded that stratification and light penetration were the triggers for blooms in impounded rivers in southeastern Australia. Similarly, Chl *a* concentration and algal density in this study were enhanced only in areas with strong thermal stratification. We also found that light penetration, as measured by the euphotic depth, was mostly confined within this SDL layer. Biomass of phytoplankton remained at a very low level in the main stem and the confluence of tributaries where limited light penetration and deep vertical mixing were likely responsible for maintaining the low phytoplankton biomass (e.g., Holbach et al. 2014, Jiang et al. 2018). High flow rates and vertical mixing might also apply to the inflows of tributaries, where strong mixing of the water mass and short retention time prevent the development of an SDL (Yang et al. 2013, Fu et al. 2015, Zhang et al. 2015).

Our study confirms the important role of physical processes, rather than nutrients, in determining the wax and wane of algal blooms in the tributaries of the TGR. Although nutrient enrichment has been shown to trigger HABs in many lakes (e.g., Lake Taihu, Lake Dianchi, and Lake Chaohu in China and Lake Erie in North America; Table S7), we did

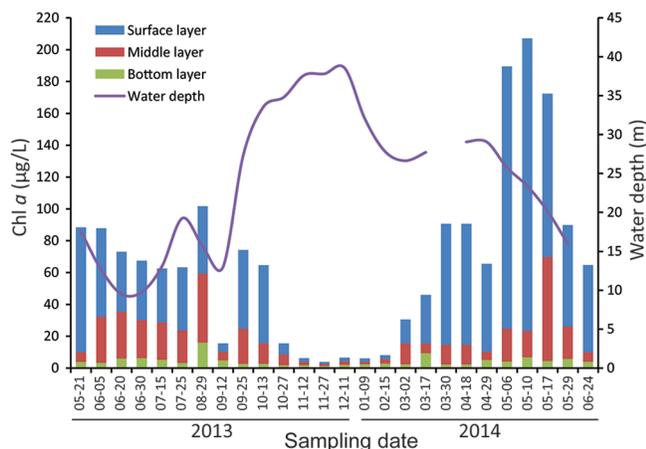


Figure 7. Changes of Chl *a* with water depth and time in Gaoyang Lake, Pengxi River, Three Gorges Reservoir, China. Surface (blue bars) refers to the uppermost layer in the water column (0–0.5 m below the water's surface), Middle (red bars) refers to the middle layer at the thermal line during stratification or middle depth without stratification, Bottom (green bars) refers to the bottom layer at 0.5 m above the riverbed. The water depth profile (purple line) is broken for 30 March and 18 April 2014 samples because of sensor malfunction.

not find a strong relationship between nutrient concentrations and algal blooms within the TGR. The discrepancy between occurrence of algal blooms and high levels of total and bioavailable nitrogen and phosphorous indicates that nutrients might not have been a driving factor of the timing and spatial distribution of the blooms (Fig. 2, Tables 1, S2, S3, S5; Dolman and Wiedner 2015). However, it does not rule out the potential role of nutrient limitation within the SDL at a finer spatial scale. Although the density layer continued to strengthen over the spring and into the early summer, algal blooms did not persist beyond May. These blooms might have become nutrient limited because there would be very little diffusion of bioavailable nutrients into the SDL from the nutrient-rich lower water column. Limited nutrient diffusion into the SDL in April, and even less in July, could have resulted in the depletion of bioavailable nutrients (e.g., Søndergaard et al. 1990) and restricted the growth of algae. Therefore, although the overall concentrations of total and bioavailable nitrogen and phosphorous suggest a nutrient-rich environment throughout the reservoir (Tables S2, S3), it is possible that nutrients could become limited within the SDL. Future research should study the potential for SDL nutrient limitations.

It is also possible that within the SDL temperature might also have the potential to limit the duration of algal blooms. Many studies have demonstrated that high water temperature is essential for the formation of blooms (e.g., Kruger and Eloff 1978, Robarts and Zohary 1987, Nalewajko and Murphy 2001), but this necessity for high temperature

was not observed in the TGR. As a result of high humidity within the canyons of the TGR, surficial-water temperatures exceeding 30°C were maintained for ~2 mo (July–August 2013, Fig. 6A; Yang et al. 2017). Such high temperatures can harm algal growth because the optimum growth temperature for many problematic algal species, including cyanobacteria, is 20–30°C (Singh and Singh 2015). Future studies should examine the relative importance of nutrient diffusion into the SDL and high temperatures in limiting algal blooms.

### Management implications

Tributaries with different morphologies and land-use activities can experience different intensities of algal blooms. For example, the Modao River is near the Pengxi River (50 km away at the confluence; Fig. 1B) and shares noticeable similarities in climate and flow regulation. However, the Modao River has seldom been reported to have algal blooms, possibly because of its lower conductivity (i.e., fewer ions or soluble minerals) in its SDL compared with the Pengxi River (Jiang et al. 2017). Management of algal blooms in the TGR will need to account for differences among the tributaries with respect to how sensitive they are to dam operation and their varying morphologies and land-use patterns. For example, in order to interfere with the formation of SDLs, regulation of daily water-level fluctuations is effective in the tributaries close to the dam (Zheng et al. 2011, Yang et al. 2013, Sha et al. 2015, Ji et al. 2017), whereas local artificial mixing of the water column would be more effective for the tributaries in the middle and upper reach of the TGR. We suggest that tributary-specific management plans will be more effective than an overall reservoir management strategy in the TGR.

The construction of dams results in reservoirs that vary in their susceptibility to HABs (Rørslett and Johansen 1996, Holbach et al. 2014, Mao et al. 2015, Ji et al. 2017, Nogueira and Pomari 2018). Our study demonstrated that the upper reaches of tributaries in the TGR are the reservoir's locations that are most vulnerable to HABs because of both dam operations and the formation of an SDL found in the upper reaches. The formation of SDLs is a phenomenon that might be unique to reservoirs confined within deep canyons that prevent surface wind mixing. It is unlikely that nutrient management within the TGR will immediately address the control of HABs, as shown by the lack of association between nutrients and HABs in our study. Therefore, future research should address the interaction of hydrological phenomena such as the formation of SDLs and nutrient bioavailability. We could not determine if the spring algal growth observed in this study was a function of nutrient dynamics within the SDL or of the higher water temperatures observed in the SDL. The use of artificial mixing techniques for local control of the formation of SDLs, however, might be a promising interim management strategy to regulate HABs in critical areas such as the backwater reaches of tributaries.

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Author contributions: LZ, GDH, and DX conceived the study. ZX, CZ, LF, and JY collected the data. DJ, DL, and BZ implemented samplings. WDT, PBH, PVC, and MA analyzed the data. LZ drafted the manuscript, and ZX and GDH revised the manuscript. All authors reviewed and approved the final version of the manuscript.

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## LITERATURE CITED

- Cai, Q., and Z. Hu. 2006. Studies on eutrophication problem and control strategy in the Three Gorges Reservoir. *Acta Hydrobiologica Sinica* 30:7–11.
- CMDC (China Meteorological Data Service Center). 2015. Dataset of daily climate data from Chinese surface stations for global exchange 2013–2015. National Meteorological Information Center, Beijing, China. (Available from: [http://data.cma.cn/en/?r=dataService/index&datacode=SURF\\_CLI\\_CHN\\_MUL\\_DAY\\_CES](http://data.cma.cn/en/?r=dataService/index&datacode=SURF_CLI_CHN_MUL_DAY_CES))
- Conroy, J. D., D. D. Kane, R. D. Briland, and D. A. Culver. 2014. Systemic, early-season *Microcystis* blooms in western Lake Erie and two of its major agricultural tributaries (Maumee and Sandusky rivers). *Journal of Great Lakes Research* 40:518–523.
- Davis, J. R., and K. Koop. 2006. Eutrophication in Australian rivers, reservoirs and confluences—A Southern Hemisphere perspective on the science and its implications. *Hydrobiologia* 559:23–76.
- Dolman, A. M., and C. Wiedner. 2015. Predicting phytoplankton biomass and estimating critical N:P ratios with piecewise models that conform to Liebig's law of the minimum. *Freshwater Biology* 60:686–697.
- Fu, B., B. Wu, Y. Lü, Z. Xu, J. Cao, D. Niu, G. Yang, and Y. Zhou. 2010. Three Gorges Project: Efforts and challenges for the environment. *Progress in Physical Geography* 34:741–754.
- Fu, L., L. Zhang, J. Yu, C. Zhou, and G. D. Haffner. 2015. Water stratification and its relevance to growth of algal community on backwater area (Gaoyang Lake) in Three Gorges Reservoir. *Chinese Journal of Environmental Engineering* 9:38–44.
- Gao, Q., Y. Li, Q. Cheng, M. Yu, B. Hu, Z. Wang, and Z. Yu. 2016. Analysis and assessment of the nutrients, biochemical indexes and heavy metals in the Three Gorges Reservoir, China, from 2008 to 2013. *Water Research* 92:262–274.
- Giri, S., Z. Qiu, and Z. Zhang. 2018. Assessing the impacts of land use on downstream water quality using a hydrologically sensitive area concept. *Journal of Environmental Management* 213:309–319.
- Hamilton, P. B., M. Proulx, and C. Earle. 2001. Enumerating phytoplankton with an upright compound microscope using a modified settling chamber. *Hydrobiologia* 444:171–175.
- Holbach, A., S. Norra, L. Wang, Y. Yuan, W. Hu, B. Zheng, and Y. Bi. 2014. Three Gorges Reservoir: Density pump amplification of pollutant transport into tributaries. *Environmental Science & Technology* 48:7798–7806.
- Hu, H., and Y. Wei (editors). 2006. The freshwater algae of China: Systematics, taxonomy and ecology. Science Press, Beijing, China.
- Jeong, Y. H., J. S. Yang, and K. Park. 2014. Changes in water quality after the construction of a confluence dam in the Geum River confluence dam system, Korea. *Journal of Coastal Research* 6:1278–1286.
- Ji, D., S. A. Wells, Z. Yang, D. Liu, Y. Huang, J. Ma, and C. J. Berger. 2017. Impacts of water level rise on algal bloom prevention in the tributary of Three Gorges Reservoir, China. *Ecological Engineering* 98:70–81.
- Jiang, R., Y. Bao, Y. Shui, Y. Wang, M. Hu, Y. Cheng, A. Cai, P. Du, and Z. Ye. 2018. Spatio-temporal variations of the stable HO isotopes and characterization of mixing between the mainstream and tributary of the Three Gorges Reservoir. *Water* 10:563–575.
- Jiang, W. 2017. Relationship of water environment and internal sediment nutrient release in Gaoyang Lake of Pengxi River, Three Gorges Reservoir. PhD Thesis. Southwest University, Chongqing, China.
- Jiang, W., C. Zhou, D. Ji, D. Liu, Y. Ren, G. D. Haffner, D. Xie, and L. Zhang. 2017. Comparison of relationship between conduction and algal bloom in Pengxi River and Modao River in Three Gorges Reservoir. *Environmental Science* 38:2326–2335.
- Kruger, G., and J. N. Eloff. 1978. The effect of temperature on specific growth rate and activation energy of *Microcystis* and *Synechococcus* isolates relevant to the onset of natural blooms. *Journal of the Limnological Society of Southern Africa* 4:9–20.
- Lehner, B., C. R. Liermann, C. Revenga, C. Vorosmarty, B. Fekete, P. Crouzet, P. Doll, M. Endejan, K. Frenken, J. Magome, C. Nilsson, J. C. Robertson, R. Rodel, N. Sindorf, and D. Wisser. 2011. High-resolution mapping of the world's reservoirs and dams for sustainable river-flow management. *Frontiers in Ecology and the Environment* 9:494–502.
- Li, J., S. Xie, and M. Kuan. 2001. Geomorphic evolution of the Yangtze Gorges and the time of their formation. *Geomorphology* 41:125–135.
- Li, Y., J. Chen, Q. Zhao, C. Pu, Z. Qiu, R. Zhang, and W. Shu. 2011. A cross-sectional investigation of chronic exposure to microcystin in relationship to childhood liver damage in the Three Gorges Reservoir Region, China. *Environmental Health Perspectives* 119:1483–1491.
- Liu, L., D. Liu, D. M. Johnson, Z. Yi, and Y. Huang. 2012. Effects of vertical mixing on phytoplankton blooms in Xiangxi Bay of Three Gorges Reservoir: Implications for management. *Water Research* 46:2121–2130.
- Mao, J., D. Jiang, and H. Dai. 2015. Spatial–temporal hydrodynamic and algal bloom modelling analysis of a reservoir tributary embayment. *Journal of Hydro-Environment Research* 9:200–215.
- McKay, R. M. L., T. Tuttle, L. A. Reitz, G. S. Bullerjahn, W. R. Cody, A. J. McDowell, and T. W. Davis. 2018. Early onset of a microcystin-producing cyanobacterial bloom in an agriculturally-influenced Great Lakes tributary. *Journal of Oceanology and Limnology* 36:1112–1125.
- Ministry of Environmental Protection, China. 2013. Annual Reports of Ecology and Environment of Three Gorges Reservoir Engineering, 2013. (Available from: <http://www.cnemc.cn/jcbg/zjsxgcsyjhjcbg/>)

- Ministry of Environmental Protection, China. 2014. Annual Reports of Ecology and Environment of Three Gorges Reservoir Engineering, 2014. (Available from: <http://www.cnemc.cn/jcbg/zjsxgcstyhjcbg/>)
- Molen, V. D., F. J. Los, V. L. Ballegooijen, and M. P. van Vat. 1994. Mathematical modelling as a tool for management in eutrophication control of shallow lakes. *Hydrobiologia* 275:479–492.
- Nalewajko, C., and T. P. Murphy. 2001. Effects of temperature, and availability of nitrogen and phosphorus on the abundance of *Anabaena* and *Microcystis* in Lake Biwa, Japan: An experimental approach. *Limnology* 2:45–48.
- Nogueira, M. G., and J. Pomari. 2018. Limnological patterns in a large subtropical reservoir cascade. Chapter 6 in D. Gökçe (editor). *Limnology—Some new aspects of inland water ecology*. IntechOpen Limited, London, United Kingdom. (Available from: <https://www.intechopen.com/books/limnology-some-new-aspects-of-inland-water-ecology/limnological-patterns-in-a-large-subtropical-reservoir-cascade>)
- Paerl, H. W., and J. Huisman. 2008. Blooms like it hot. *Science* 320:57–58.
- Peng, J., Y. L. Wang, J. S. Wu, J. Yue, Y. Zhang, and W. F. Li. 2006. Ecological effects associated with land-use change in China's southwest agricultural landscape. *International Journal of Sustainable Development and World Ecology* 13:315–325.
- Quinn, F. H. 1992. Hydraulic residence times for the Laurentian Great Lakes. *Journal of Great Lakes Research* 18:22–28.
- Rice, E. W., R. B. Baird, A. D. Eaton, and L. S. Clesceri (editors). 2012. *Standard methods for the examination of water and wastewater*. 22<sup>nd</sup> edition. American Public Health Association, American Water Works, Water Environment Federation, Washington, DC.
- Rigosi, A., and F. J. Rueda. 2012. Hydraulic control of short-term successional changes in the phytoplankton assemblage in stratified reservoirs. *Ecological Engineering* 44:216–226.
- Robarts, R. D., and T. Zohary. 1987. Temperature effects on photosynthetic capacity, respiration and growth rates of bloom-forming cyanobacteria. *New Zealand Journal of Marine and Freshwater Research* 21:391–399.
- Rørslett, B., and S. W. Johansen. 1996. Remedial measures connected with aquatic macrophytes in Norwegian regulated rivers and reservoirs. *Regulated Rivers: Research & Management* 12:509–522.
- Sha, Y., Y. Wei, W. Li, J. Fan, and G. Cheng. 2015. Artificial tide generation and its effects on the water environment in the backwater of the Three Gorges Reservoir. *Journal of Hydrology* 528:230–237.
- Singh, S. P., and P. Singh. 2015. Effect of temperature and light on the growth of algae species: A review. *Renewable and Sustainable Energy Reviews* 50:431–444.
- Søndergaard, M., E. Jeppesen, P. Kristensen, and O. Sortkjaer. 1990. Interaction between sediment and water in a shallow and hypertrophic lake: A study on phytoplankton collapses in Lake Søbygård, Denmark. *Hydrobiologia* 191:139–148.
- Tanaka, M., G. Girard, R. Davis, A. Peuto, and N. Bignell. 2001. Recommended table for the density of water between 0°C and 40°C based on recent experimental reports. *Metrologia* 38:301–311.
- Thomann, R. V., and J. A. Mueller (editors). 1987. *Principle of surface water quality modeling and control*. Harper & Row Publishers, New York, New York.
- Tilzer, M. M. 1988. Secchi disk–chlorophyll relationships in a lake with highly variable phytoplankton biomass. *Hydrobiologia* 162:163–171.
- Venables, W. N., and B. D. Ripley. 2002. *Modern applied statistics with S*. 4<sup>th</sup> edition. Springer, New York, New York.
- Wang, X. 2012. Study on the eutrophication and models of hydrodynamic and water quality in the Pengxi (Xiaojiang) River of Three Gorges Reservoir. PhD Thesis. Chongqing University, Chongqing, China.
- Wu, H., J. Chen, J. Xu, G. Zeng, L. Sang, Q. Liu, Z. Yin, J. Dai, D. Yin, J. Liang, and S. Ye. 2019. Effects of dam construction on biodiversity: A review. *Journal of Cleaner Production* 221:481–487.
- Xu, Q. 2018. The analysis of the change of water area in reservoir area based on the remote sensing image of the Three Gorges Dam—A case study of pre-dam to Jiangjin section. PhD Thesis. Sichuan Normal University, Sichuan, China.
- Yang, B., B. He, and D. Wang. 2017. Hanfeng pre-reservoir commissioning time variation feature and water quality in Three Gorges Reservoir. *Environmental Science* 38:1366–1375.
- Yang, Z., D. Liu, D. Ji, and S. Xiao. 2010. Influence of the impounding process of the Three Gorges Reservoir up to water level 172.5 m on water eutrophication in the Xiangxi Bay. *Science China Technological Sciences* 53:1114–1125.
- Yang, Z., D. Liu, D. Ji, S. Xiao, Y. Huang, and J. Ma. 2013. An eco-environmental friendly operation: An effective method to mitigate the harmful blooms in the tributary bays of Three Gorges Reservoir. *Science China Technological Sciences* 56:1458–1470.
- Ye, L., X. Q. Han, Y. Xu, and Q. Cai. 2007. Spatial analysis for spring bloom and nutrient limitation in Xiangxi Bay of Three Gorges Reservoir. *Environmental Monitoring and Assessment* 127:135–145.
- Zarfl, C., A. E. Lumsdon, J. Berlekamp, L. Tydecks, and K. Tockner. 2015. A global boom in hydropower dam construction. *Aquatic Science* 77:161–170.
- Zeng, H., L. Song, Z. Yu, and H. Chen. 2006. Distribution of phytoplankton in the Three Gorge Reservoir during rainy and dry seasons. *Science of the Total Environment* 367:999–1009.
- Zeng, H., L. Song, Z. Yu, and H. Chen. 2007. Preliminary study on algal blooms within the Three Gorges Reservoir. *Resources and Environment in the Yangtze Basin* 16:336–339.
- Zhang, J., B. H. Zheng, L. Liu, L. Wang, M. Huang, and G. Wu. 2010. Seasonal variation of phytoplankton in the Daning River and its relationships with environmental factors after impounding of the Three Gorges Reservoir: A four-year study. *Procedia Environmental Sciences* 2:1479–1490.
- Zhang, L., Z. Xia, W. Zhou, P. B. Hamilton, and G. D. Haffner. 2015. Spatial distribution of nutrients and phytoplankton and causes for their differences in Three Gorges Reservoir in spring. *Research of Environmental Sciences* 28:1069–1077.
- Zhang, W. 2014. Weighing the pros and cons: Transformation of angle of view for Three Gorges Dam. *Natural Resources* 5:1048–1056.
- Zhang, Y., X. Liu, Y. Yin, M. Wang, and B. Qin. 2012. Predicting the light attenuation coefficient through Secchi disk depth and beam attenuation coefficient in a large, shallow, freshwater lake. *Hydrobiologia* 693:29–37.

Zheng, T., J. Mao, H. Dai, and D. Liu. 2011. Impacts of water release operations on algal blooms in a tributary bay of Three Gorges Reservoir. *Science China Technological Sciences* 54:1588–1598.

Zhu, K., Y. Bi, and Z. Hu. 2013. Responses of phytoplankton functional groups to the hydrologic regime in the Daning River, a tributary of Three Gorges Reservoir, China. *Science of the Total Environment* 450:169–177.

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