# Phytoplankton Assemblages and Trophic Status of Wular Lake in Kashmir Himalaya

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## ABSTRACT

An ecological study was carried out to determine the trophic status of Wular Lake, in Kashmir Himalaya using physicochemical characteristics and phytoplankton assemblages from March 2002 to February 2004. Multivariate statistical techniques were employed to understand the anthropogenic and natural factors driving the spatio-temporal variation in the ecological and trophic status of Wular Lake. Based on the calculated trophic state index (TSI), Secchi disk transparency reflects the eutrophic nature while as per the concentration of total phosphorus, the lake falls under hypereutrophic category. Non-metric multidimensional scaling (NMDS) showed clear distinction in samples (abundance data) based on the seasons (color) and very small differentiation based on the sites (shape) with stress values 0.133. Analysis of similarity (ANOSIM) test revealed that there is a strong, statistically significant difference in phytoplankton communities based on seasons and no difference was noticed based on sites. Canonical corresponding analysis (CCA) was very successful in showing that the phytoplankton assemblages are positively and negatively correlated with physicochemical characteristics. The first two axes accounted for 87% of phytoplankton assemblage variance. This study provided special insight into the changes in physicochemical characteristics, phytoplankton, and trophic status of Wular Lake.

Keywords: TSI, Carlson, Trophic status, Wular, Wetland, Himalaya

# INTRODUCTION

Water is a critical resource and thus, it is necessary to find ecologically sustainable ways to protect and conserve valuable freshwater resources including lakes and wetlands (El-Serehy et al., 2018). Continuous monitoring and assessment of lake and wetland water quality is a key factor for proper conservation and management (Yu et al., 2011; Dar et al., 2020a). Wetlands are subjected to several environmental changes through natural factors and anthropogenic activities (water supply, agriculture, industries, tourism and recreation) (El-Serehy et al., 2018; Tibebe et al., 2019; Dar et *al.,* 2021a).

Wetlands are biologically diverse and productive ecosystems and cover only 6% of the earth's

surface (Mitsch and Gosselink, 1986; Maltby, 1988). Wetlands have often been regarded as "earth's kidneys" because of their inherent capacity to filter pollutants (Sharifi *et al.*, 2013). Wetlands are vital to human life and provide numerous socio-economic and ecosystem services including water purification, flood control, natural carbon stores, fisheries, water supply, and recreation (Junk *et al.*, 2013; Lamsal *et al.*, 2015; Kumar and Mahajan, 2020; Dar *et al.*, 2020b).

Wetlands are crucial for livelihoods, agriculture, and fisheries for rural populations among the developing nations of the world (Maltby and Acreman, 2011). Massive industrial and agricultural expansion, infrastructural development, and land system changes have drastically decreased their aerial extent and

declined socio-economic and ecosystem services (Bassi et al., 2014; Dar et al., 2021b). Globally, wetland loss and degradation are significantly more rapid than any other ecosystem (Davidson 2014; El-Serehy et al., 2018; Tibebe et al., 2019). Millennium Ecosystem Assessment (2005) report suggests that 50% of the world's wetland types have been lost during the 20<sup>th</sup> century. The deterioration of water quality and ecological health of wetland ecosystems is driven by increased water withdrawal, agricultural runoff, eutrophication, expansion of human settlements, and invasive species (MEA, 2005; McCartney et al., 2010). Monitoring and assessment of the wetland ecosystems for eutrophication are essential to mitigate or prevent adverse environmental and economic impacts (Devlin et al., 2011; Yu et al., 2011). To determine the trophic and nutrient status of lakes and wetlands, Secchi disk transparency, chlorophyll-a as a measure of algal activity and total phosphorus are required for the Carlson's Trophic Status Index (Carlson, 1977; Zhang et al., 2016). The trophic state index categorizes lakes as eutrophic, mesotrophic, or oligotrophic (Gholizadeh et al., 2016). The dynamic nature of the productivity and eutrophication due to natural and anthropogenic factors leaves no single assessment variable as a true measure of the trophic status of a given water body (Xu et al., 2001; Padisák et al., 2009) and a combination of physical and chemical parameters are widely used in determining the health of a lake ecosystem (Phillips et al., 2013). Phytoplankton, both in lotic and lentic environments are considered to be a reliable measure of environmental health reporting on different levels of eutrophication (Wetzel, 1983; Xu et al., 2001; Soylu and Gönülol, 2010; Demir et al., 2014), however with different

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levels of accuracy. Eutrophication is characterized by the excessive increase in algal and plant growth, due to the availability of one or more growth limiting factors (key plant nutrients) (Schindler, 2006). In the recent past, humans have accelerated the rate of eutrophication through point and non-point sources of nutrient loading (cultural eutrophication) (Carpenter, 2005; Dar et al., 2021c, d). Eutrophication has serious consequences for domestic drinking water sources, aquaculture and recreation, and the estimated cost of damage caused by the eutrophication is around \$ 2.2 billion annually (Dodds et al., 2009). Water guality deterioration is widespread in Wular Lake and has increased significantly owing to the increasing anthropogenic activities (agricultural activities, sewage discharge, and urban expansion) (Rumysa et al., 2012; Shah and Pandit, 2012; Hassan et al., 2015; Bano et al., 2018). Turbidity and transparency (Secchi disk) are measures of water clarity and vary spatiotemporally (Davies-Colley and Smith, 2001). High turbidity and low transparency of water bodies are associated with increased water temperature, low dissolved oxygen and excessive algal growth (Kirk, 1994).

Wular Lake, one of the largest freshwater bodies in India was designated as a Wetland of International importance under the Ramsar convention in 1990 (Matthews, 1993). It is located in the North-West of Kashmir, about 64 km from Srinagar city and is a rural wetland. The lake is the main flood drainage basin of the Kashmir valley mainly fed by River Jhelum. In addition to this nallah Erin, Madhumati and a large number of small tributaries add a huge volume of water to it. Wular Lake plays a significant role in fishery, water supply, flood

control, agriculture, hydrological regime and (Wetlands International, recreation 2007). Enhanced anthropogenic activities and anthropogenically driven changes in natural processes have degraded this ecosystem at an alarming rate, affecting its ecological health and water quality (Bhat and Pandit, 2014). The sustainable conservation and management of Wular Lake are vital from its socio-economic and ecological importance. Wular Lake is valued as a habitat for migratory birds that visit Kashmir from different parts of the world. The present study provides important insights into the spatiotemporal changes in phytoplankton assemblages and the water quality of Wular Lake. Furthermore, the trophic state was evaluated using transparency and total phosphorus concentrations which can provide baseline information for future research and conservation efforts for the Wular Lake.

## **MATERIAL AND METHODS**

## Study area and study sites

Geographically the Wular Lake, one of the largest wetlands of India, is situated at an altitude of

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1,580 m (a.m.s.l), between 34°16'-34°20'N latitudes and 74°33'-74°44'E longitudes (Fig. 1). Wular Lake, an ox-bow type lake, is of fluviatile origin located in the northwest of Kashmir about 35 km from Srinagar city. Wular Lake is formed by the meandering of River Jhelum, which is the main feeding channel besides other tributaries. It plays a significant role in the hydrography of the Kashmir valley by not only acting as a huge absorption basin for floodwaters but also for maintaining flows to support agriculture and hydropower generation, as well as sports activities. The lake along with the extensive marshes surrounding is an important habitat for fish, accounting for 60% of the fish production within the state of Jammu and Kashmir (Farooq et al., 2017). The lake is largely shallow, with a maximum depth of 5.8 m, the deeper part being on the Western side opposite the hills of Baba Shakur Din. The lake is drained in the northeast by the only single outlet in the form of River Jhelum. For the present study, seven sites were selected from the Wular Lake shown in Fig. 1.



Fig. 1. Map of Wular Lake showing various study sites selected for the present investigation.

The catchment of the lake is comprised of sloping hills of the Zanskar ranges of the western Himalaya on the Northeastern and Northwestern sides which drain their runoff through various nallahs, where Erin and Madhumati are prominent. On the Eastern and Southern sides are the low-lying areas of Sonawari which used to get inundated almost every year until numerous crisscrossing embankments were constructed along River Jhelum. The wetland area thus reclaimed has in the recent past been brought under cultivation of paddy and plantations of willow, poplar and fruit trees. On the western side in the Sopore-Watlab section, low-lying areas have also been brought under paddy cultivation. In 1986, the lake was designated as a wetland of importance under national the Indian Governments Wetlands Programme and in 1990 it was enlisted as a wetland of international importance under the Ramsar Convention of 1971.

## **Physicochemical characteristics**

Fifteen physicochemical variables were measured at seven sites from March 2002 to February 2004 on monthly basis, making a total of 8 seasonal samples. Water samples from each site were collected in a one-litre pre-rinsed polyethylene bottle. Transparency of Wular waters at each site was determined using Secchi disk and depth was measured using the graduated rod. Water temperature (WT) was determined in situ by using a mercury-in-glass thermometer while a pre-calibrated digital pH and conductivity meter were used to record the pH (pH) and conductivity (Cond, µS/cm) (APHA, 1998). Dissolved oxygen (DO, mg/L), total alkalinity (TA, mg/L), free carbon dioxide (FC, mg/L), chloride (Cl, mg/L), calcium (Ca, mg/L) and magnesium (Mg, mg/L) were

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determined by titrimetric methods (APHA, 1998). Sodium (Na, mg/L) and Potassium (K, mg/L) using flame photometry (APHA, 1998). Ammoniacalnitrogen (NH<sub>3</sub>N), nitrate-nitrogen (NO<sub>3</sub>N,  $\mu$ g/L), total phosphorus (TP,  $\mu$ g/L), and orthophosphorus (Ortho-P,  $\mu$ g/L) were determined by colorimetric assays using spectrophotometer following standard methods (APHA, 1998).

# Phytoplankton Collection

For microscopic identification, enumeration and determination, phytoplankton samples were collected every month from March 2002 to February 2004. Phytoplankton was collected by horizontal phytoplankton hauls using plankton net with a mesh size of 64  $\mu$ m connected with a sample bucket at the end. The obtained samples were fixed with acetic Lugol's solution with a final of (Throndsen, concentration 1% 1978). Phytoplankton taxa were identified using an inverted microscope (magnification 200X, 400X, and 600X). The most commonly used literature was consulted to ascertain the taxonomy of the phytoplankton's (Krammer and Lange-Bertalot, 1986; Sournia, 1986; Popovski and Pfiester, 1990; Cox, 1996; Komarek and Anagnostidis, 1999; Hickman, 2000; John et al., 2002; Sheath and Wehr, 2015).

# **Plankton Biomass**

Lohman's (1908) formula was applied for the estimation of phytoplankton biomass. Since the volume and fresh weight of phytoplankton very much correspond to each other, the average density being 1.01–1.03 (Hutchinson, 1967), the biomass is expressed as micrograms per liter (=  $10^{6} \mu^{3}L^{-1}$  or mg m<sup>-3</sup> assuming  $1\mu^{3} = 10^{-9}$  mg (Jumppanen, 1976; Pandit, 1980).

## Data analysis

We performed non-metric multidimensional scaling (NMDS) using the vegan package (Oksanen et al., 2016), a robust ordination technique that produces an ordination based on similarity or dissimilarity among sites. The NMDS attempts to represent the pairwise dissimilarity between the sites in a reduced number of dimensions, as closely as possible, using Bray-Curtis dissimilarity (McCune and Grace, 2002; Clarke and Gorley, 2006). The sites that are more similar to one another in terms of biological assemblages are ordinated together. As a rule of thumb, an NMDS ordination plot with a stress value below or close to 0.05 is considered a good fit (Shepard 1962; Mahecha et al., 2007). To establish the results obtained from NMDS, analysis of similarity (ANOSIM, P=0.05) using dissimilarity index package "vegan" (Clarke, 1993) was employed. Canonical corresponding analysis (CCA) was used to evaluate the impact of environmental variables on the distribution patterns of the epilithic algal population at each location, using a robust statistical method. CCA is used to describe composite environmental factors that represent the patterns of species occurrence as closely as possible (ter Braak, 1987; Stevenson et al., 1989). Axes are derived, which are combinations of environmental variables. linear Individual organisms are directly connected to these axes assuming the environmental variables are affected by unimodal species. CCA is a means of investigating not only the direct relationship between modern diatom assemblages and associated environmental variables but also the indirectly implementation of weighted average calibration (Palmer, 1993). The CCA was carried using Package 'labdsv' (Roberts, 2019). The

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environmental and biological datasets were subjected to analysis of variance (ANOVA) to check difference among means within and between groups using package "ggpubr". The statistical analysis of data was carried out in R Software Version 3.4.0 (R Core Team, 2017). Both physicochemical and biological variables were transformed and normalized to allow comparison at the same scale.

## Trophic state determination

The trophic state was determined based on the Trophic State Index (TSI) using a logarithmic transformation (Ln) of the Secchi disk transparency (SDT) in meters and the total phosphorus (TP) in microgram per liter according to the following equations (Carlson, 1977):

$$TSI(SD) = 60 - \frac{14}{41} \ln(SD) (m) \quad (1)$$
$$TSI(TP) = \frac{14}{41} \ln(TP) + \frac{4}{15} (\mu g/L) \quad (2)$$

Vollenweider's criteria for assessing the trophic state of a water body, accepted protocol by the Organization for Economic Co-Operation and Development (OECD) (Ryding and Rast, 1994), based on the average values of selected parameters were used in the present study (Vollenweider, 1989).

# **RESULTS AND DISCUSSION**

# Physico-chemical properties of the Wular Lake

The minimum and maximum values of different physicochemical parameters measured at the seven sampling sites selected from the Wular Lake are listed in Fig. 2a, b. The surface water temperature varied between 4.0 °C in winter at site VII and 28 °C in summer at site VI. Depth levels ranged between 4.8 m (spring, at site V)

and 0.5 m (winter at Site IV), SDT (Secchi disk transparency) varied from 0.18m (site IV, winter) and 2.23m (winter Site V). pH ranged between 7.11 at site VII (summer) and 7.72 at site V (autumn), conductivity varied between 230 µS/cm (autumn, site IV) and 295.66 µS/cm (summer, site VI), FC varied from 12.5 mg/L at site VII (autumn) to 20.16 mg/L at site VI (summer), DO levels ranged between 10.5 mg/L (spring, at site VII) and 6.11 mg/L (summer at Site V), Cl ranged between 28.5 mg/L at site VII (summer) and 9.66 mg/L at the site I (winter), TA varied between 201 mg/L (winter, site VI) and 85.66 mg/L (spring, site IV) (Fig. 2a). Ca varied from 55 mg/L at site V (winter) to 21.33 mg/L at site IV (summer), Mg ranged between 7.33 mg/L at site VII (summer) and 22.13 mg/L at site VI (winter), Na varied between 4.23 mg/L (autumn, site I) and 7.98 mg/L (summer, site I), K varied from 2.66 mg/L at site IV (winter) to 4.8 mg/L at the site I (summer). NO<sub>3</sub>-N concentration ranged from 214.66 µg/L at site VII (winter) to 457.22  $\mu$ g/L at site V (summer) while NH<sub>3</sub>-N varied from 42.5 µg/L (site II, summer) to 162.165 µg/L (site VI, winter). The concentration of TP and Ortho-P

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remained moderate, fluctuating between 101  $\mu$ g/L (site VII, winter) - 215  $\mu$ g/L (site IV, summer) and 15.33 µg/L (site VII, winter) -32 µg/L (site IV, spring), respectively (Fig. 2b). Transparency of wetlands is affected by several factors, including suspended and dissolved solids. Low transparency reflects a large number of suspended sediments and planktonic material (Wetzel 2001; Shah et al., 2019). Low transparency in some of the Kashmir Himalayan lakes is due to increased sediment load and suspended planktonic material (Sinha et al., 2002). The increased concentration of phosphorus recorded in the Wular Lake is driven by agricultural activities, sewage discharge, and litter decomposition (Del Rosario et al., 2002; Pandit and Yousuf, 2002: Bhat et al., 2014; Bhat et al., 2015; Kumar and Mahajan, 2020). The concentration of phosphorus and transparency values in Wular Lake were found to fall in hypereutrophic and eutrophic category respectively as per the standards set by OECD for trophic status classification of wetlands (Ryding and Rast, 1994). Analysis of variance (ANOVA, p<0.05) showed that physico-chemical characteristics didn't vary significantly across sites except for depth, transparency, and ortho-phosphorus (p<0.05) while significant variations were observed across seasons (p<0.05) (Fig. 2a, 2b).



**Fig. 2a** Analysis of variance of environmental variables across seven sites (p<0.05). DEP: Depth (m), WT: Water Temperature (°C), TRANS: Transparency (m), pH, COND: Conductivity (μS/cm), TA: Total Alkalinity (mg/L), FC: Free Carbon dioxide (mg/L), DO: Dissolved oxygen (mg/L), Cl: Chloride (mg/L),



Fig. 2b. Analysis of variance of environmental variables across seven sites (p<0.05). Ca: Calcium (mg/L), Mg: Magnesium (mg/L), Na: Sodium (mg/L), K: Potassium (mg/L), TP: Total phosphorus (μg/L), Ortho-P: Ortho-phosphorus (μg/L), NH<sub>3</sub>N: Ammoniacal-nitrogen (μg/L), NO<sub>3</sub>N: Nitrate-nitrogen (μg/L).

# Planktonic algae of the Wular Lake

The phytoplankton community reported from the Wular Lake belongs to 4 classes: Bacillariophyceae, Chlorophyceae, Cyanophyceae, and Euglenophyceae. The percentage contribution of 49.6, 26, 13.8, and 9.1% to the phytoplankton community was recorded for Bacillariophyceae, Chlorophyceae, Cyanophyceae, and Euglenophyceae, respectively (Fig. 3).



**Fig. 3.** The percentage contribution, relative density, and biomass of Bacillariophyceae, Chlorophyceae, Cyanophyceae, and Euglenophyceae to the phytoplankton community at different sites of Wular Lake.

Overall, the Bacillariophyceae was the most abundant group of phytoplankton, followed by the Chlorophyceae rank and the Cyanophyceae. While the Bacillariophyceae formed the largest group almost every month, the Chlorophyceae group represented a large part of the phytoplankton community during the study period, although the population varied during the sampling seasons. In terms of relative density, Bacillariophyceae dominated the phytoplankton (Munawar, 1970, Munawar *et al.*, 1974; Ganai *et al.*, 2010) community followed by Chlorophyceae (Khan, 1978) and others while in terms of relative biomass, Chlorophyceae recorded the highest biomass at all sites, followed by Bacillariophyceae and Cyanophyceae (Fig. 3). The level and value of the biomass of phytoplankton classes were generally rich reaching the highest density (11466 Ind/liter) at Site IV in the Wular Lake, and a visible decline in the algal density reaching 4267 Ind/liter at site 1 (Fig. 4). Diatoms (Bacillariophyceae) formed the dominant component of the phytoplankton community in Wular Lake representing 49.6% of the total composition. Diatoms are very sensitive to changes in water quality and thus are used as biomonitoring tools

in aquatic ecosystems (Stevenson and Smol, 2003). Due to the short life-cycle diatoms respond quickly to the changes in physicochemical characteristics and nutrient enrichment from anthropogenic activities (Rahmati *et al.*, 2011; Darling, 2015; Shah *et al.*, 2017). Furthermore, diatoms show a strong correlation for total phosphorus concentrations (Wang *et al.*, 2014). Chlorophyceae were recorded throughout the year. On the other hand, members of Cyanophyceae were present in the samples received in some seasons and the numbers tend

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to remain low, a fact contrary to the other groups of phytoplankton. Phytoplankton assemblage patterns and structures are driven by spatiotemporal changes in physico-chemical and biological characteristics (Zhang *et al.,* 2011; Demir *et al.,* 2014). Analysis of variance (ANOVA, p<0.05) showed a significant difference in phytoplankton communities across seasons except for Euglenophyceae, while no significant difference was noticed across sites throughout the study period (Fig. 4).



**Fig. 4.** Analysis of variance (ANOVA) of phytoplankton (Ind./liter) dataset across sites and seasons (p<0.05). Bacill: Bacillariophyceae, Chloro: Chlorophyceae, Cyan: Cyanophyceae, Eugl: Euglenophyceae

# **Trophic status classification**

Data contained in Table 1 presents the complete spectrum of the trophic state index (TSI) and its associated parameters (Carlson, 1977). The values

calculated for TSI for the Wular Lake based on SDT and TP are shown in Fig. 5. The TSI value obtained for Wular Lake based on SDT and TP

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parameter were above 60. TSI values for Wular Lake results were compared with the internationally accepted criteria accepted by the OECD (1982) for the trophic status classification of the lake.

**Table 1.** OECD criteria are used for trophic state classification of the water bodies.

Trophic Status	TP (µg/L)	Transparency (m)		
OECD criteria		Mean	Maximum	
Ultra-oligotrophic	<4	>6	>12	
Oligotrophic	<10	>3	>6	
Mesotrophic	10-35	1.5-3	3-6	
Eutrophic	35-100	0.7-1.5	1.5-3	
Hypereutrophic	>100	<0.7	<1.5	
Wular Lake results	175	1.08	1.94	

The present study employed Carlson's Trophic State Index (TSI) with the understanding that it is a well-tested, robust quantitative method and replicable method considering biological and physical parameters, and the findings are presented in Table 1. A TSI value between 40 and 50 is usually associated with mesotrophic (moderate productivity), values greater than 50 are associated with eutrophic (high productivity), and values less than 40 are associated with oligotrophic nature (low productivity) of the water body (Murthy *et al.*, 2008; Kumar and Mahajan, 2020). TSI results for seven sites revealed that the Wular Lake has TSI Index above 60 (Fig. 5)



**Fig. 5.** The calculated trophic state index (TSI), using Carlson's method (CTSI) with the measured Secchi disk transparency (SDT), and total phosphorous (TP) values for the Wular Lake during the present study.

## **Statistical Analysis**

Non-metric multidimensional scaling (NMDS) showed quite a distinction in samples (abundance data) based on the seasons (color) and very small differentiation based on the sites (shape) with stress values 0.133 (Fig. 6). Analysis of similarity

(ANOSIM) test revealed that there is a strong, statistically significant difference in phytoplankton communities based on seasons (R = 0.42, p = 0.0001) and no difference was noticed based on sites (R = 0.01349, p = 0.404).



**Fig. 6.** NMDS based on phytoplankton and zooplankton abundance data across seven sites and four seasons in Wular Lake.

In the present study, the CCA was very successful. About 0.14886/0.33374 or 0.8051 of the total variability was captured in the CCA (Table 2). The first two eigenvalues value sum to 0.1318. The first axis accounts for approximately 75% of the constrained variability, with the second at 12% (Table 2). The phytoplankton groups and biomass are shown as red, sites as black and environmental variables as blue arrows. In the present analysis, the first axis is associated with increasing DEP, WT, TRANS, Cl, TP (negative values) and DO, TA, Ca, Mg (positive values). The second axis is associated with Na, K (positive values) and pH, Ortho-P (negative values). In axis 1, Bacillariophyceae (positive values) are opposed to Chlorophyceae, Cyanophyceae, Xanthophyceae, and their biomass (RBCyan, RBChl, RBEugl, RBBac) (negative values). In axis 2, R and GPP (positive values) are opposed to RBCyan, RBChl, RBEugl, RBBac (negative values) (Fig. 7). Furthermore, the Chi-square test tells us that the phytoplankton community is closely linked with the environmental variables (p<0.001).



**Fig. 7.** CCA ordination plot showing the distribution of phytoplankton abundance and biomass communities along an environmental gradient.

# CONCLUSIONS

Wular Lake can be categorized as a lake falling between eutrophic to hypereutrophic category concerning calculated TSI values and greater diversity and density of phytoplankton community. The trophic status of the Wular Lake has been confirmed by Carlson's numerical index. Wular Lake is facing serious water quality problems due to anthropogenic activities (domestic sewage and agriculture runoff). Statistical techniques revealed that seasonal factors played a strong and dominant role in changing the pattern of physico-chemical characteristics, which in turn controlled the abundance of the phytoplankton community throughout the study period. CCA showed that the gradient created by the physico-chemical characteristics has a strong influence on the phytoplankton community. The conservation and management of Wular Lake (Wetland of international importance) have become а pressing environmental issue in the wake of population increase, climate change, and pollution. Management and conservation of Wular Lake would require a collective effort from the scientists, policymakers, and citizens to economically viable, develop an socially acceptable, and scientifically reliable long-term bio-manipulative method to restore this important ecosystem.

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# **Conflict of interest**

The authors declare that they have no conflict of interest

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