



# The threshold responses of phytoplankton community to nutrient gradient in a shallow eutrophic Chinese lake



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## ABSTRACT

Excessive nutrient loads resulted in cascading trophic effects and ecosystem responses. Aims of this study were to determine if the thresholds in nutrient gradient related to phytoplankton community composition could be identified in eutrophic lake, and further to analyze the change of phytoplankton assemblage along the nutrient concentration based on Threshold Indicator Taxa ANalysis (TITAN). The results presented the significant community thresholds estimate for negative taxa declining at 1.650 mg/L TN and 131.5  $\mu$ g/L TP, as well as simultaneously increasing for positive taxa at 1.665 mg/L TN and 151.5  $\mu$ g/L TP along nutrient enrichment gradient. However, there was unremarkable change point determined for TN:TP ratios in Lake Dianchi. Elevated TN and TP altered the phytoplankton assemblage, even may induce the fade of algal blooms across the threshold in the hypertrophic lake. The findings could provide implications for deeply deciphering abrupt transitions for phytoplankton assemblage and developing nutrient tactics to protect the lake ecosystems.

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## 1. Introduction

Anthropogenic activities have altered the input of nitrogen and phosphorus to freshwater ecosystems and resulted in severer eutrophication, which are expected to continue increasing in the coming decades due to growing populations, further development, and greater demand for food production and energy production (Carpenter et al., 1998; Smucker et al., 2013). Excessive loads of nutrients to lakes and streams negatively affect ecosystem structure and function by changing dissolved oxygen regimes, increasing algal biomass, altering biological communities, food webs, rates of nutrient and carbon cycling (Smucker et al., 2013). Especially, this emerging threat has significant impacts on algal biodiversity (Proulx et al., 1996; Palmer et al., 2009; Dudgeon, 2010; Taylor et al., 2014) and often leads to harmful algal blooms (Carpenter and Lathrop, 2008), which means there is considerable risk of crossing a threshold to sustained eutrophication with high concentrations of nitrogen and/or phosphorus (Civin-Aralar et al., 2004; Carpenter and Lathrop, 2008; Conley et al., 2009).

As early as in the 1970s, Schindler (1974) reported that phosphorus control alone was an effective means of overcoming eutrophication problem after a long-term full-lake experiment in Canada. However, nitrogen has clearly been established as the nutrient limiting phytoplankton production and leads to a “vicious circle” of eutrophication (Conley et al., 2009). Furthermore, subsequent studies in lakes established that nutrient ratios influence the growth, physiological state, and community structure of phytoplankton (Pick, 1989), while Civin-Aralar et al. (2004) considered that nutrient concentrations, at least for nitrogen and phosphorus, are more critical in determining algal population and community structure rather than the ratios of these two nutrients. These different opinions resulting in a debate are whether one or both nutrients or the ratios of these two nutrients (Conley et al., 2009) as the key factor should be controlled to reverse the detrimental effects of eutrophication of lakes (Schindler et al., 2008; Schindler and Hecky, 2009; Scott and McCarthy, 2011; Slomp, 2011).

No matter which opinion is supported, it is still necessary to identify which nutrient element is the key factor on eutrophic freshwater ecosystems and whether there is a nutrient threshold for phytoplankton community response as useful benchmarks to support management strategy for protecting and restoring freshwater ecosystem structure and functions (Dodds, 2007; Dodds et al., 2010; Taylor et al., 2014). Nutrient threshold is a

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well-known type of ecological thresholds, which usually is focused on a shift from the clear, macrophyte-dominated state to the turbid state dominated by phytoplankton in lakes related to an increase in nutrient loads (Scheffer, 2004; Loverde-Oliveira et al., 2009). Recent studies examining biotic community structure across nutrient gradients in freshwater ecosystems have reported threshold or wedge-shaped patterns in biotic diversity and index metrics (King and Richardson, 2003; Evans-White et al., 2009; Brenden et al., 2008; King and Baker, 2010; Smucker et al., 2013). These thresholds can be identified by characterizing nonlinear relationships between nutrients and the structure of community assembly among different taxonomic groups based on statistical methods (Toms and Lesperance, 2003; King and Richardson, 2003; Brenden et al., 2008; King and Baker, 2010; Maloney et al., 2012). However, to our knowledge, few studies have examined whether nutrient enrichment might partly drive patterns of phytoplankton community structure across an eutrophication gradient. Especially, Phytoplankton community consists of three functional group, i.e., diatoms, green algae, and cyanobacteria (Zhao et al., 2008), and plays a central role in trophic transfers through aquatic food webs in freshwater ecosystems (Winder and Schindler, 2004). Its composition and structure are assumed to reflect to the stress derived from nutrient enrichment (Pick, 1989; Zohary et al., 1996; Cuvin-Aralar et al., 2004; Cai and Kong, 2013). Meanwhile, the main taxa of algal blooms are supposed to be *Microcystis*, *Anabaena*, *Aphanizomenon*, *Cylindrospermopsis*, *Planktothrix*, etc., which are belonging to phytoplankton community (Graham et al., 2010; Huber et al., 2012; Cai and Kong, 2013). Thus, to identify the nutrient threshold relevant to phytoplankton community response in eutrophic lake will be particularly useful for early warning for lake ecosystems (Biggs, 2000; Dodds et al., 2002; Smucker et al., 2013).

Therefore, the specific aims of this study are (i) to identify key nutrient factors influencing phytoplankton community assemblage and further detect whether there exist nutrient thresholds explaining the nonlinear response of community assemblage; (ii) to analyze the change of community structure along the nutrient gradient. Our results can provide initial support for deeply deciphering algal blooms mechanism and mitigating eutrophication in lake ecosystems.

## 2. Methods

### 2.1. Study sites and field sampling

The Lake Dianchi (24°40'–25°03' N, 102°03'–102°48' E) is the largest shallow lake in Yunnan-Guizhou plateau and the sixth largest one in China with approximately 300 km<sup>2</sup> total lake area, which has a distinctive monsoon climate with an annual mean temperature of 14.7 °C, an average precipitation of 797–1007 mm, and 227 frost-free days per year (Yang et al., 2010). No thermocline exists in Lake Dianchi. Strong wind-generated waves and currents enable complete mixing in this shallow lake (Zhang et al., 1996; Bai et al., 2012). A dike divided the lake into two parts, Waihai and Caohai. Caohai sits in the north of Lake Dianchi and Waihai with 97.2% of the whole area is the main water body of Lake Dianchi (Fig. 1). In the last two decades, runoff drains a significant amount of nitrogen and phosphorus nutrients into Lake Dianchi. It has been suffering algal blooms because of the drastic anthropogenic activities in this basin (Liu et al., 2013).

During August of 2011 and 2012, 57 sites distributed throughout Lake Dianchi were sampled for planktonic algae and water chemistry (Fig. 1). Underlying the spatially different nutrient concentration, 30 sites in 2011, two surface water samples of each site were collected following a uniform grid method. Another 27 sites with two surface water samples were uniformly collected in

2012. This sampling as the supplementary data to ensure the sampled phytoplankton was not re-sampled at the same locations of sampling sites in 2011. Water samples preserved in 1000 mL acid-washed plastic bottles. Samples were kept in 4 °C prior to chemical analysis.

### 2.2. Water quality analysis

Total nitrogen (TN), total phosphorus (TP), ammonia nitrogen (NH<sub>3</sub>N) and permanganate index (COD<sub>Mn</sub>, major contaminant in last few years) were analyzed in Kunming Environmental Monitoring Center following the standard methods described by the American Public Health Association (APHA, 1998). Another one was fixed with Lugol's iodine solution and settled for at least 24 h before determining species abundance microscopically.

### 2.3. Phytoplankton taxon analysis

Identification and enumeration of phytoplankton taxa were carried out according to Hu and Wei (2006). The biovolume of each species was calculated from cell numbers and size measurements. These biovolumes were converted into biomass (mg/L) on the basis of a phytoplankton cell density of 1 mg/mm<sup>3</sup> (Chen et al., 2003; Cai and Kong, 2013). Species were identified into genera, and average density was calculated for individual genera at each sites. Total algal density and the distribution of genera belonging to the major taxonomic groups were determined. Species richness, Shannon–Wiener index, Simpson index, and Evenness index were calculated according to Pielou (1974, 1975) for detecting the random distribution of individuals. Berger–Parker index was computed for expressing the proportional importance of the most abundant species (Magurran, 1988; Latini and Petrere, 2004). Succession of dominant genera in the different nutrient gradient was assessed based on each genera's percentage contribution to the overall density. The equations of three diversity indexes are defined as follows:

$$\text{Shannon–Wiener index} = -\sum_{i=1}^s p_i \ln p_i \quad (1)$$

$$\text{Simpson index} = \frac{1}{\sum_{i=1}^s p_i^2} \quad (2)$$

$$\text{Berger–Parker index} = \frac{N_{\max}}{N} \quad (3)$$

where  $p$  is the proportion ( $n/N$ ) of individuals of one particular species found ( $n$ ) divided by the total number of individuals found ( $N$ ),  $s$  is the number of species and  $N_{\max}$  is the number of individuals in the most abundant species.

### 2.4. Data analysis

Redundancy analysis (RDA) is a method to extract and summarize the variation in a set of variables that can be explained by a set of explanatory variables combining regression and principal component analysis (Legendre and Legendre, 1998), which was used to test the statistical significance of the main environmental variables that impact phytoplankton community structure. The Monte Carlo permutation test (permutations = 999,  $p < 0.05$ ) was applied to reveal the effect of the obtained explanatory variables on the community structure. Changes in phytoplankton diversity indices were then related to environmental variables using Pearson's correlation analysis. Both analyses were conducted using Vegan package (version 1.17-0; Oksanen et al., 2010) in R.2.9.2 (R Development Core Team, 2009).

Threshold Indicator Taxa ANalysis (TITAN) was also performed in R.2.9.2 combining indicator species analysis (Dufrene and

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