Phytoplankton Community Dynamics in Tadlac Lake, Los Baños, Laguna, Philippines

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ABSTRACT

Tadlac Lake was heavily used for aquaculture until a massive fish kill occurred in the lake from December 1999 to February 2000 during its annual overturn. Subsequently, aquaculture activity was prohibited in the lake to improve its water quality. In this study, we assessed the trophic status of Tadlac Lake by characterizing the phytoplankton community and environmental variables collected from October 2017 to March 2018 to evaluate the effectiveness of the rehabilitation strategy. A total of 14 phytoplankton species were present in the seven stations sampled throughout the study period. Chlorophyta had the highest relative density (RD) (71.42 %), followed by Bacillariophyta (32.37 %) and Cyanophyta (8.57 %). Among phytoplankton taxa, *Aulacoseira granulata* had the highest density, followed by *Coelastrum microporum* and *Eudorina elegans*. Significant variation in phytoplankton density was documented among sampling months ($p<0.05$), but not among sampling sites ($p>0.05$). Differences in the phytoplankton density across sampling months were also supported by cluster analysis and analysis of similarities. Similarity percentage showed that the difference in plankton density could mainly be attributed to the density of the most abundant taxa. Canonical correspondence analysis revealed that phytoplankton communities in the lake were highly influenced by nitrate, phosphate, pH, temperature, and conductivity. Tadlac Lake is still in eutrophic condition based on the dominant phytoplankton species, high concentration of nitrate and phosphate, and calculated biotic indices. Therefore, further strategies are needed to more effectively control nutrient enrichment and restore the ecosystem of Tadlac Lake.

Keywords: Eutrophication, Limnology, Phytoplankton, Tadlac Lake, Trophic status, Water quality

INTRODUCTION

Eutrophication is one of the main issues in the lake ecosystem (Callisto *et al*., 2014). Eutrophication of lakes and rivers is a slow process in which there is an external supply of nitrogen and phosphorus minerals to these water systems that are derived from vast fluvial, atmospheric, and groundwater sources (Smith *et al*., 1999). This process has been dramatically accelerated by cultural (man-made) eutrophication (Mukherjee *et al*., 2010). Several activities such as agricultural fertilization, industrial and municipal waste release, irrigation, fishing, and recreation cause cultural eutrophication (Castro *et al*., 2005). These activities cause a disruption of balance among members of the aquatic community, such as plankton, benthos, and nekton (Jorgensen, 1976; Estlander *et al*., 2009). Therefore, assessing the trophic state of an aquatic body is essential for its effective management and conservation.

The phytoplankton community is widely used in assessing the trophic status and water quality of an aquatic system (Reynolds, 1999; Walsh *et al*., 2001). Phytoplankton species are considered suitable bioindicators as they can adapt to different

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environmental conditions, and their distributions are affected by several parameters such as temperature, light, and concentration of nutrients (Napiorkowska-Krzebietke *et al.*, 2013). The phytoplankton community is sensitive to pollutants, and its structure and metabolism change quickly in response to environmental changes. Moreover, phytoplankton has a short life cycle and fast reproduction rates, and collection is easy and inexpensive (Stevenson *et al.*, 1986).

Tadlac Lake is a closed lake in Barangay Tadlac Los Baños, Laguna, Philippines, and one of the eight crater lakes of Laguna de Bay (Laguna Lake), the Philippines' largest lake. Tadlac Lake has a surface area of 248,000 m^2, an average depth of 27 m, and an annual turnover from December to February (LLDA, 2007). Before the introduction of *Oreochromis* (tilapia) aquaculture in Tadlac Lake in 1986, the lake was considered an oligotrophic lake. Tilapia aquaculture quickly caused the lake to become eutrophic. At its peak, fish cages occupied almost 90 % of the surface area of the lake despite the Philippine law mandating that only 10 % of the lake's surface area could be utilized for aquaculture. A massive fish kill occurred in December 1999 during the lake's annual turnover, and all of the remaining fishes in the lake were wiped out in February 2000. This led the government agencies, namely Laguna Lake Development Authority (LLDA), Barangay Council, and the Barangay Fisheries and Aquatic Resource Management Council (BFARMC) to rehabilitate the lake via banning of aquaculture in the area (Santos-Borja, 2008). The ban on aquaculture in the lake persisted until this study was undertaken. Despite this intervention, there has been no post-rehabilitation assessment of the lake's water quality or trophic status. These data are empirical and are thus the primary tool that will gauge the recovery of the lake after rehabilitation. If shown to be effective, the rehabilitation efforts in the area must be continued. Otherwise, other forms of intervention and other restoration efforts must be employed. Thus, this study presents the phytoplankton community dynamics of Tadlac Lake, highlighting the lake's trophic status. The physico-chemical characteristics of the lake were also determined, and considered along with biotic indices to ascertain the lake's current trophic state.

MATERIALS AND METHODS

Study area

Samples were taken from Tadlac Lake at seven stations: five stations in the littoral zone to represent the surrounding land use, and two stations in the limnetic zone (Figure 1). Stations 1 and 2 were non-residential areas, and the shoreline was covered with vegetation only. Station 3 was in a commercial area, station 4 was in a residential area, while station 5 was near a poultry farm. Stations 6 and 7 represent the limnetic zone. Every station was divided into three sampling points with an interval of 100 m apart, and those were further divided into three sub-points separated by at least 10 m intervals. The lake is currently closed for aquaculture activity because of the rehabilitation done by the local government.

Sample collection

The study was conducted in Tadlac Lake, which is situated in Brgy. Tadlac Los Baños, Laguna, Philippines. Los Baños experiences Type I climate of the Philippines, with two pronounced seasons: dry from November to April and wet during the rest of the year (Lantican, 2001). Sampling was conducted on the 30th of each month from October 2017 to March 2018; wherein October and March represented the wet season and the remaining sampling months represented the dry season. Phytoplankton samples were collected using a 20-µm mesh-size plankton net. Horizontal towing was employed in the littoral zone, while vertical towing was used in the limnetic zone (Tingson and Tamayo-Zafaralla, 2018). Three replicates of water samples were collected for each sub-point for the littoral and limnetic zone. A total of nine samples were collected per sampling station. All water samples were placed in 1,000 mL polyethylene (PET) bottles and preserved with 1% buffered formalin.

Eight physicochemical parameters were measured as possible factors that could influence plankton species composition and distribution in the sampled sites: (1) temperature (°C), (2) pH, (3) electrical conductivity ($\mu S \cdot cm^{-1}$), and (4) dissolved oxygen concentration ($mg \cdot L^{-1}$) were measured on-site with PASCO multi-parameter water quality meter; (5) transparency (meters) was measured using Secchi disk; (6) biochemical oxygen demand (BOD) ($mg \cdot L^{-1}$) was measured using BOD 5-day method; and (7) phosphate ($\mu M\ PO_4^{3-}$-P) and (8) nitrate ($\mu M\ NO_3^-$-N) concentrations were measured spectrophotometrically with Hach PhosVer® and NitraVer® reagent powder pillows, respectively.

Specimen processing and identification

Water samples were filtered with a 20-µm mesh net which separated the phytoplankton from the water. The residuum (remainder after the water has been removed) was then diluted with 10 mL of the water filtrate. The container was agitated thoroughly to homogenize the sample, and a 1 mL aliquot was drawn out using a pipette. The sample was dispensed on a haemocytometer and counted under an electric compound microscope (Mercurio *et al*., 2016). Phytoplankton samples were identified to the lowest possible taxon using the identification keys of Botes (2003), Vuuren *et al*. (2006), and Bellinger and Sigee (2010). The cell density ($cells \cdot L^{-1}$) was calculated using the formula of Pioli (2019):

$$N = \frac{\text{total number of cells} \times \text{dilution factor}}{\text{total volume scanned}}$$

Biotic indices

Algal Genus Pollution Index was employed to ascertain the trophic status of Tadlac Lake. A list of the most pollution-tolerant genera and species based on the Palmer index was determined for each sampling period. A pollution index factor was

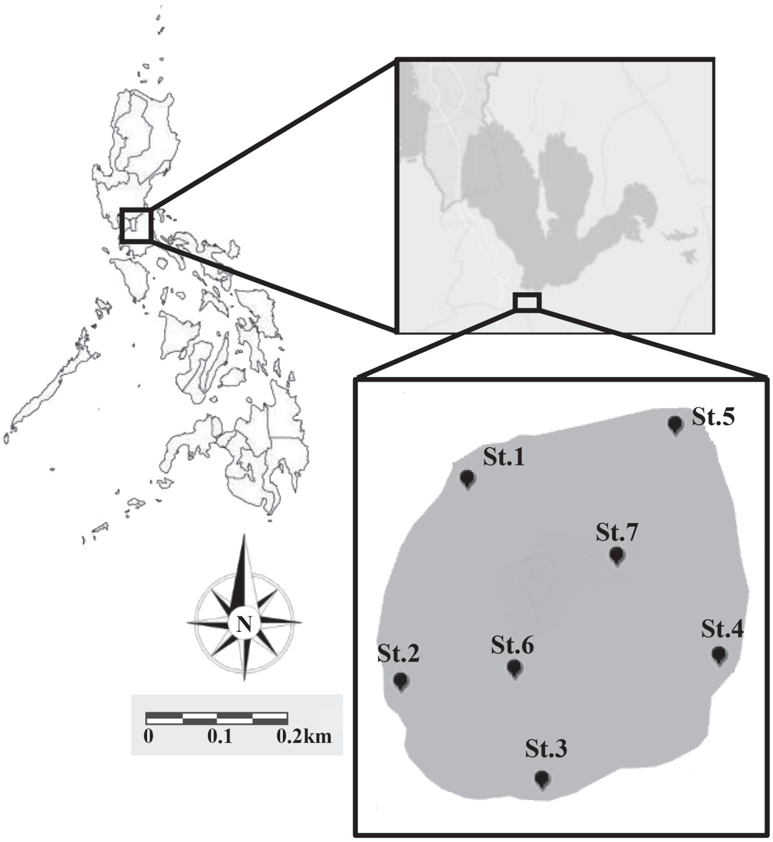

Figure 1. Map of Tadlac Lake showing the seven sampling stations. Littoral Zone: Stations 1 and 2, non-residential areas; Station 3, commercial area; Station 4, residential area; Station 5, poultry farm. Limnetic Zone: Stations 6 and Station 7.

assigned to each genus and species by determining each alga's relative number of total points (Palmer, 1969). A genus with a pollution index of 5 indicates high organic pollution, while a genus with a pollution index of 1 indicates low organic pollution. Any algae not included in the list have a pollution factor of zero. A total calculated score of ≤14 indicates low organic pollution, values ranging from 15-19 indicate moderate organic pollution, and values ≥20 indicate high organic pollution.

The trophic status of the lake was further assessed using the saprobity index. The sum of saprobic values for all the indicator species determined at the sampling point was divided by the sum of all frequency values for the indicator species, which can be calculated using the following formula:

$$SI = \frac{\sum s \times h}{\sum h}$$

where: SI is the Pantle and Buck (1955) saprobic index; s is the saprobic index of species "i" (Sladecek, 1983); and h is the relative frequency (1, uncommon; 3, common; 5, abundant).

The saprobic index (SI) is based on the following ranking scale: ≤1.5, oligosaprobic with low organic pollution; 1.6-2.6, mesosaprobic with moderate organic pollution; ≥2.7, eusosaprobic with high to very high organic pollution (Battes and Momeu, 2011).

Data analysis

The Shapiro-Wilk test was employed to assess the normality of the data. Since the data for phytoplankton density significantly departed from a normal distribution ($p<0.01$), plankton density data were log10 (x+1) transformed to improve the homoscedasticity and normality. One-way analysis of variance (ANOVA) with Tukey's honest significant difference (HSD) as the post-hoc analysis was used to check the differences between the physicochemical parameters measured between sampling periods and phytoplankton density between different sampling stations and sampling months. Cluster analysis with the Bray-Curtis similarity index was done to check the similarity of phytoplankton abundance between sampling months. Differences in phytoplankton assemblage between sampling stations were assessed via nonmetric dimensional scaling (NMDS) ordination technique, with Bray-Curtis dissimilarity index using the abundance data. Analysis of similarities (ANOSIM) was used to test for statistical differences in phytoplankton community structure among sampling sites and sampling months. Pairwise r values from ANOSIM were utilized to gauge absolute partition among assemblages. R values>0.25 indicate separated groups (Clarke and Green, 1988; Michelland *et al*., 2010). To determine the overall contribution of each species to the dissimilarity between sampling periods, similarity percentage (SIMPER) was applied (Clarke, 1993). Canonical correspondence analysis with type II scaling was used to determine the physicochemical parameters that contributed to the variation of phytoplankton density throughout the sampling period. Paleontological Statistics Software was used for all statistical analyses (Hammer *et al*., 2001) except for ANOVA and post hoc test, which were performed using Statistical Program for Social Science (SPSS) version 20 (trial version).

RESULTS AND DISCUSSION

Environmental parameters

Statistical variation was observed in all measured physico-chemical parameters (Table 1). Higher mean surface water temperatures were recorded during October (31.00 °C), November (32.00 °C), February (30.78 °C), and March (31.74 °C). Lower temperatures were noted in December (26.30 °C) and January (29.00 °C). Likewise, a drastic drop in DO was observed during December (2.48 $mg\cdot L^{-1}$) and January (0.51 $mg\cdot L^{-1}$). The mean DO ranged from 0.51 $mg\cdot L^{-1}$ to 10.43 $mg\cdot L^{-1}$. As with BOD, the lowest value was recorded during December and January, whereas the highest BOD was noted in November at 3.20 $mg\cdot L^{-1}$. The highest pH values were recorded in October (8.95) and November (10.90), while the lowest pH (7.70) was documented in December. Mean Secchi disk transparency (SDT) readings ranged from 1.17 m to 2.58 m. The highest recorded SDT was 2.58 meters and 1.92 meters for October and March respectively, while November (1.17 m) and December (1.29 m) had the lowest SDT values. Electrical conductivity (EC) ranged from 847.00 µS·cm^{-1} (October) to 1,048 µS·cm^{-1} (January). The highest nitrate concentration was observed in December (25.00 µM), and the lowest was found in January (7.42 µM). Phosphate concentration was also highest in December (7.16 µM) and lowest in November (2.42 µM).

The Philippines Department of Environment and Natural Resources water quality classification ranges from public water supply class 1 (Class AA) to industrial water supply class II (Class D) (DENR, 2019). The mean values of the measured environmental parameters are within the normal ranges described for lake ecosystems. The water quality of Tadlac Lake falls under the "Class B Water" designation, intended for primary contact recreation, and propagation and growth of fish and other aquatic resources.

Gonzales and Flavier (1996) assessed the physicochemical parameters of Tadlac Lake when the lake was heavily used for aquaculture (Table 2). Compared to their findings, it can be concluded that there has been an improvement in the water quality of the lake, particularly in terms of dissolved oxygen, transparency, biological oxygen demand, and nitrate. Although the parameters are within the normal range for a lake system, the nutrient levels recorded indicate eutrophic conditions.

Lakes with nitrate-nitrogen levels between 0.5 mg·L^{-1} (8.06 µM) and 1.5 mg·L^{-1} (24.19 µM) are considered eutrophic (Gibson *et al*., 2020). Lakes with a phosphate concentration of less than 0.010 mg·L^{-1} (0.11 µM) are classified as oligotrophic, while those greater than 0.020 mg·L^{-1} (0.21 µM) are eutrophic (Muller and Helsel, 1996). The mean nitrate concentration in the lake was 17.15±0.21 µM, whereas the mean phosphate concentration was 3.86±0.01 µM. Both of these values suggest that Tadlac Lake is eutrophic. Effluent discharge from domestic sewage, urban run-off, and farm waste are the primary sources of organic pollution (Meijide *et al*., 2008, Zhu *et al*., 2018). Despite the current absence of aquaculture, which had been the major contributor of nutrient and organic matter loading in the lake, Tadlac Lake is still in a eutrophic state. It was observed that there are some residential areas, an active poultry farm, and resorts present along the shoreline of Tadlac Lake. These establishments may currently be significant contributors of organic matter loading in the lake, and since Tadlac Lake is a closed lake, discharge from these sources tends to stay and accumulate in the lake throughout time.

Table 1. Summary of the measured physico-chemical parameters from six monthly samples (October 2017 to March 2018) of Tadlac Lake, Los Baños, Laguna, Philippines.

Environmental Parameter	**Sampling Period**						
	October	**November**	**December**	**January**	**February**	**March**	**Mean**
Temperature (°C)	31.00±0.27^{c}	32.00±0.10^{b}	26.30±0.05^{a}	29.00±0.11^{c}	30.78±0.01^{c}	31.74±0.17^{c}	29.97
pH	8.95±0.04^{b}	10.00±0.01^{c}	7.70±0.02^{a}	8.73±0.02^{b}	8.36±0.04^{b}	8.55±0.01^{b}	8.87
Transparency (m)	2.58±0.05^{c}	1.17±0.01^{a}	1.29±0.04^{a}	1.62±0.02^{b}	1.60±0.01^{b}	1.92±0.01^{c}	1.70
Conductivity (µS·cm^{-1})	847.00±4.9^{a}	848.00±6.20^{a}	939.00±12.60^{b}	1048±8.21^{c}	967.57±10.07^{b}	865.48±3.97^{a}	919.18
DO (mg·L^{-1})	7.58±0.07^{c}	7.44±0.05^{c}	2.48±0.12^{b}	0.51±0.02^{a}	8.51±0.01^{d}	10.43±0.18^{e}	6.17
BOD (mg·L^{-1})	2.74±0.15^{c}	3.20±0.10^{c}	1.09±0.07^{b}	0.15±0.01^{a}	2.23±0.18^{c}	2.97±0.14^{c}	2.06
Nitrate (µM)	16.13±0.05^{b}	17.58±0.04^{b}	25.00±0.06^{d}	7.42±0.02^{a}	15.97±0.18^{c}	20.81±0.34^{b}	17.15
Phosphate (µM)	2.74±0.02^{b}	2.42±0.02^{b}	7.16±0.02^{a}	3.68±0.02^{b}	2.53±0.01^{b}	4.63±0.01^{b}	3.86

Table 2. Comparison of physico-chemical parameters of Tadlac Lake before and after the aquaculture ban.

Environmental Parameter	Present Study	Gonzales and Flavier (1996)	Optimum Value based on DENR Water Quality Standard 2019
Temperature (°C)	29.97	26.60	25-31
pH	8.87	7.97	6.5-9.0
Transparency (m)	1.70	1.14	*
Conductivity ($\mu S \cdot cm^{-1}$)	919.18	421.16	*
DO ($mg \cdot L^{-1}$)	6.17	2.64	5 $mg \cdot L^{-1}$ minimum
BOD ($mg \cdot L^{-1}$)	2.06	2.63	7 $mg \cdot L^{-1}$ maximum allowable limit
Nitrate (µM)	1.06	2.29	112.90 µM (7 $mg \cdot L^{-1}$) maximum allowable limit
Phosphate (µM)	0.37	0.32	5.26 µM (0.50 $mg \cdot L^{-1}$) maximum allowable limit

Note: *no optimum values were set

Phytoplankton species composition and distribution

A total of 14 phytoplankton species were collected and identified from the samples collected from Tadlac Lake. Of these, ten were members of Chlorophyta, two were Bacillariophyta, and two were Cyanophyta. Chlorophyta had the highest overall relative density (56.16 %), the highest abundance in the littoral zone, and the second highest abundance in the limnetic zone. This was followed by Bacillariophyta species (35.27 %), the most abundant group in the limnetic zone and the second most abundant group in the littoral zone. Cyanophyta was the least abundant group in both zones (Figure 2)

Green algae (Chlorophyta) dominate Tadlac Lake. The abundance of Chlorophyta may be attributed to their competitive abilities, such as colonizing, growing, and tolerating multiple stresses (Cox, 1990). Moreover, in most tropical lakes, phytoplankton taxa are usually dominated by the Chlorophyceae (Sterner, 1989; Paerl, 1996). Lakes in the tropics have relatively less fluctuation in terms of water temperature and total radiation than temperate lakes, as there is almost constant solar

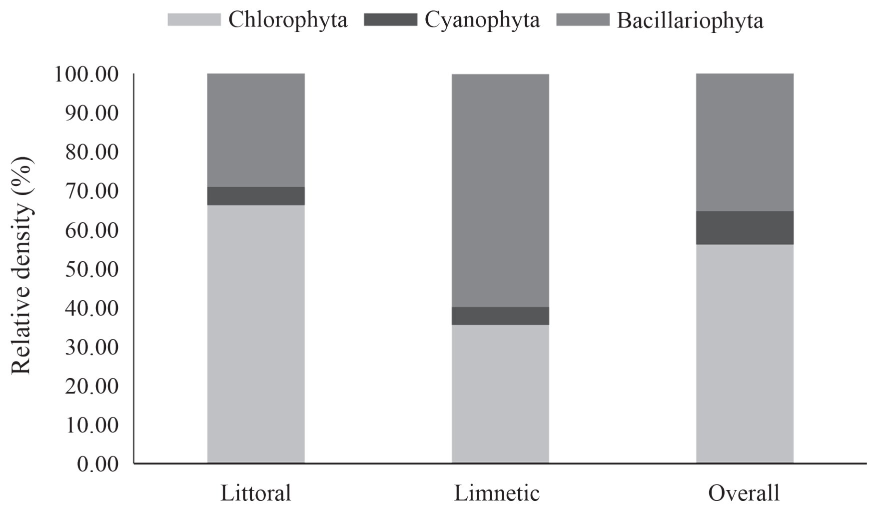

Figure 2. Relative density of different phytoplankton groups in Tadlac Lake.

energy input in the tropical region; this favors the abundance of phytoplankton species, especially the green algae (Talling, 1987). Furthermore, sampling showed that the relative density of phytoplankton in the littoral zone was higher than in the limnetic zone, except for Bacillariophyta, which had higher density in the limnetic zone. Results indicated that the relative density of taxa in the phytoplankton community of the lake was influenced by the differences in the conditions and activities of the two zones. Although the littoral and limnetic zones are connected, there are differences in environmental parameters between the two areas producing a variation in phytoplankton abundance (Jamal *et al*., 2014). Moreover, microhabitats are produced by the two zones that also influence the abundance of species, as well as their life strategies, growth, and development (Twardochleb and Olden, 2016). The abundance of Cyanophyta and Chlorophyta in the littoral zone might be attributed to their ability to live in a more disturbed zone where the presence of nutrient and sediment-laden run-off from fertilizer use and manure favors their growth (Ward and Waniek, 2007).

One-way ANOVA revealed significant variation in overall phytoplankton density and density of individual species among the six sampling months ($p<0.05$). The highest densities were recorded in October, November, and December when all of the 14 phytoplankton species were recorded. A sudden drop in the overall density and the number of species (n = 10) was recorded in January, then density increased in February and March. However, the number of species declined to eight in February and increased to nine in March (Figure 4). *Aulacoseira granulata*, *Planktothrix agardhi*, *Staurastrum anatinum*, *Eudorina elegans*, and *Coelastrum microporum* were consistently abundant throughout the six sampling periods. *Aulacoseira granulata* had the highest mean density and relative density (39,240 cells·L^{-1}; 35.07 %) followed by *Eudorina elegans*, (22,150 cells·L^{-1}; 19.80 %) and *Coelastrum microporum* (21,019 cells·L^{-1}; 18.79 %). On the other hand, no significant variation ($p>0.05$) in phytoplankton density was detected among sampling stations (Figure 5). This would signify that despite the different human activities and environmental conditions in the limnetic and littoral areas of Tadlac Lake, it did not induce a significant change in the phytoplankton community.

The three dominant phytoplankton taxa distributed in Tadlac Lake are known as indicators of eutrophic conditions (Figure 3). Several studies demonstrated that *A. granulata* is commonly found in fresh to brackish and eutrophic water (Hutchinson, 1967; Abubacker *et al*., 1996; Vuuren *et al*., 2006; Shruthi *et al*., 2011). *Aulacoseira granulata* has been linked with trophic conditions where there is a high nutrient supply (Bicudo *et al*., 2016). Eutrophication commonly stimulates the development of algal cells and colonies (Khan and Ansari, 2005). The abundance of these species could indicate that the lake is eutrophic. A similar study was conducted by Hanpongkittikul and Wongrat (2005), wherein

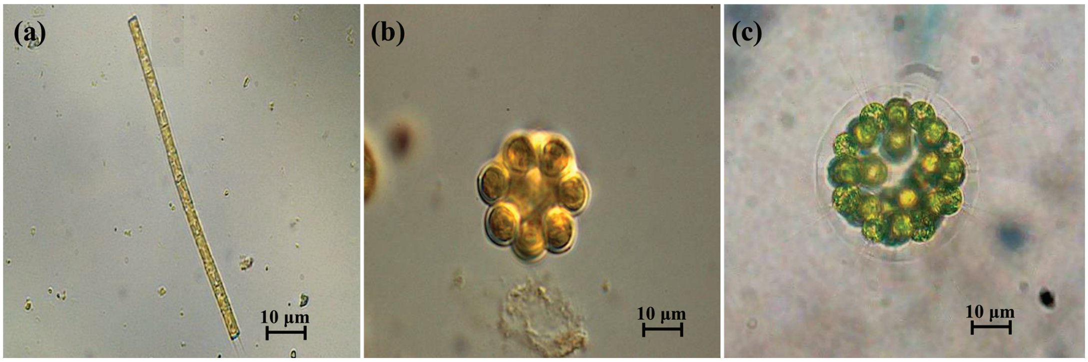

Figure 3. Three phytoplankton taxa with the highest densities in Tadlac Lake during six sampling periods (October 2017 to March 2018): (a) *Aulacoseira granulata*, (b) *Coelastrum microporum* and (c) *Eudorina elegans*

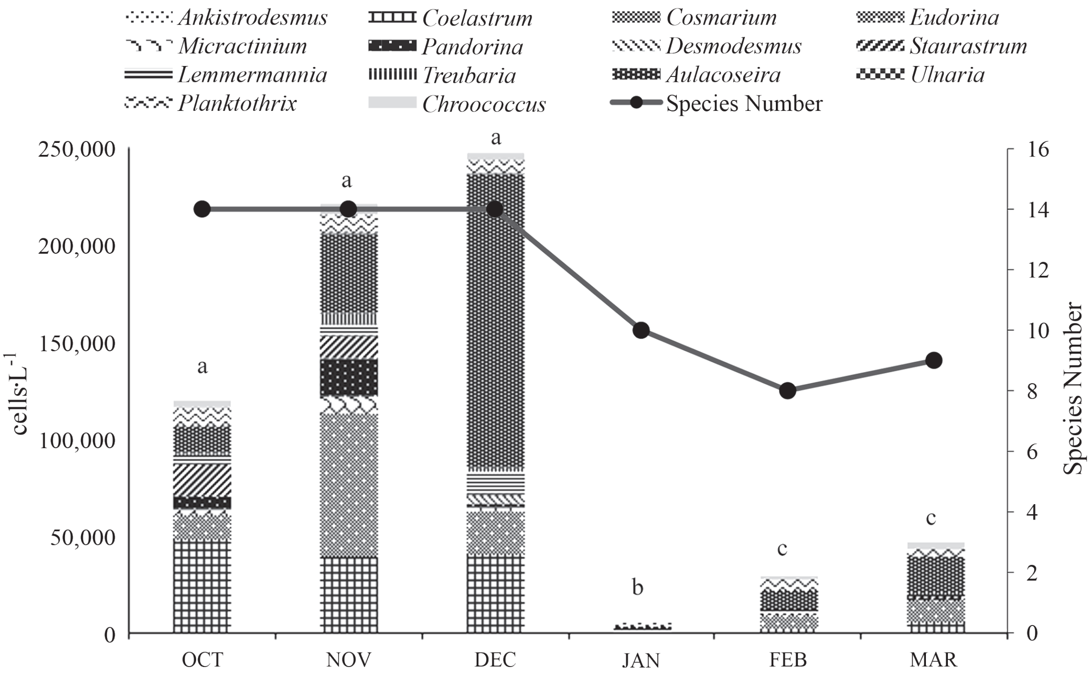

Figure 4. Mean density (cells·L^{-1}) and number of phytoplankton species collected in Tadlac Lake in each sampling period (October 2017 to March 2018). Different letters above bars denote significant ($p<0.05$) difference between mean overall phytoplankton densities.

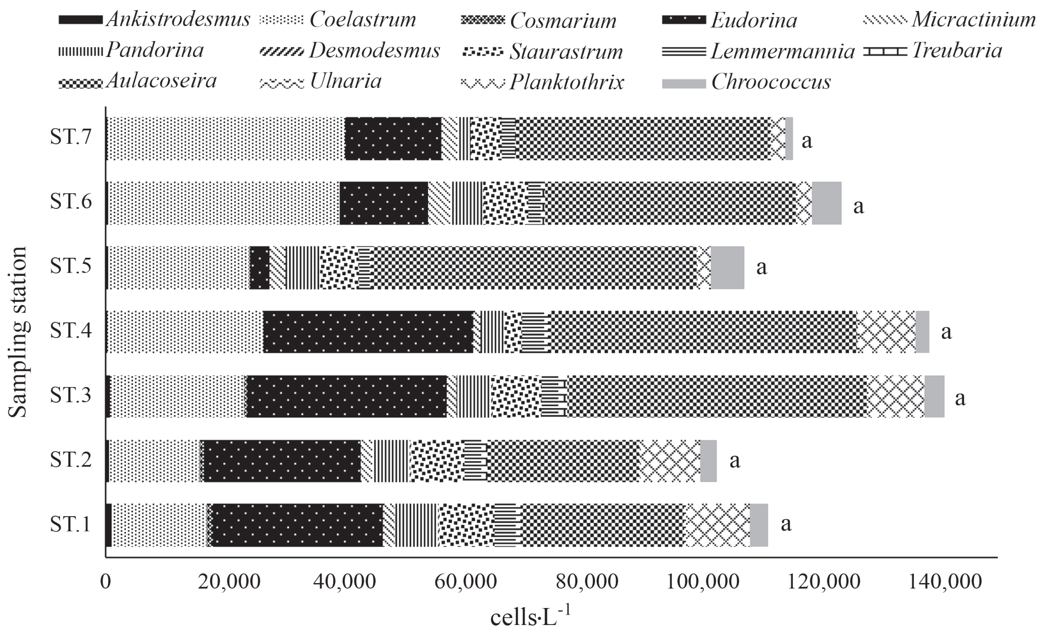

Figure 5. Mean density (no. of cells·L^{-1}) of phytoplankton in Tadlac Lake by sampling station (October 2017 to March 2018). Bars with the same letters are not significantly different ($p>0.05$).

they assessed the phytoplankton community in the Pasak Jolasid Reservoir in Lop Buri Province, Thailand. The study revealed that the reservoir is experiencing eutrophic conditions based on the abundance of eutrophic phytoplankton species such as *Aulacoseira granulata*, *Desmodesmus*, *Pediastrum*, *Closterium*, and *Coelastrum*.

Coelastrum microporum, the second most abundant phytoplankton species in Tadlac Lake, is an indicator of mesotrophic to eutrophic conditions (Vuuren *et al*., 2006; Hulyal and Kaliwal, 2009). Similarly, colonies of *Eudorina colonies*, the third most abundant species, are frequently found in waters associated with nutrient enrichment (Vuuren *et al*., 2006). Several studies also noted eutrophication of aquatic ecosystems based on the abundance of these species. Ochocka and Pasztaleniec (2016) investigated the water quality and trophic state of 10 lakes in northeastern Poland using phytoplankton and zooplankton communities. One of the abundant phytoplankton species documented was a member of the genus *Coelastrum*, which signifies eutrophication of the water bodies studied. Kemka *et al*. (2006) assessed the trophic status of Yaounde Municipal Lake in Cameroon, Central Africa. As in this study, their team also noted the abundance of *Eudorina* spp. in the lake and showed that it was in a eutrophic state.

Aside from these three species, 10 more species observed in the lake are also eutrophic indicator species according to Vuuren *et al*. (2006), namely *Cosmarium turpinii*, *Chroococcus dispersus*, *Micractinium*, *P. agardhi*, *Pandorina morum*, *Desmodesmus communis*, *S. anatinum*, *Ulnaria ulna*, *Lemmermannia triangularis*, and *Treubaria triappendicularia*.

Cluster analysis divulged a distinct difference in phytoplankton abundance among the sampling periods, thus showing a temporal variation in the phytoplankton community (Figure 6). Three major clusters were formed based on phytoplankton abundance per month. Phytoplankton abundance during February and March formed one cluster, while another tight group was observed for samples from October, November, and December, with November and December being highly similar. Meanwhile, phytoplankton abundance for January formed a separate group, showing disparity from the rest of the sampling months. Analysis of similarities (ANOSIM) revealed that sampling periods that were grouped in the dendrogram are significantly different from one another (r-value = 0.83, $p<0.05$). These results coincide with the previously described ANOVA findings (Figure 4) and indicate that there is temporal variation in the density of phytoplankton species.

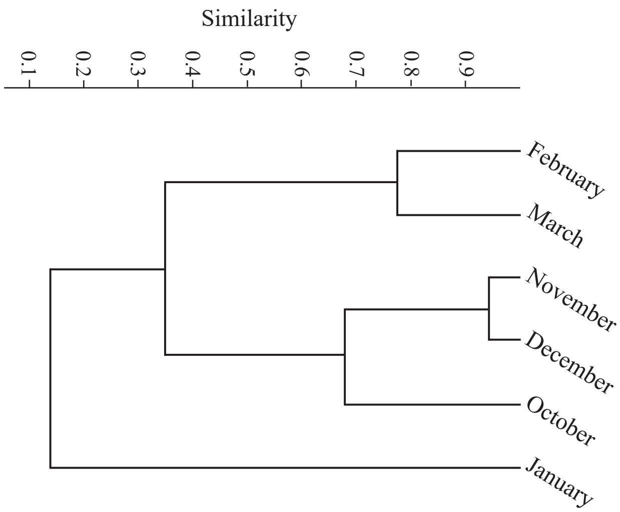

Figure 6. Cluster analysis based on the abundance of phytoplankton species in Tadlac Lake in six monthly samples.

Similarity percentage (SIMPER) was calculated to determine which taxa contributed to the observed variation in the phytoplankton density throughout the study sampling period. Findings of the SIMPER analysis further validate the ANOSIM results, where significant differences were observed by cluster analysis. High average dissimilarity percentages were calculated between months, ranging from 37.76 % to 96.16 %. The taxa that contributed most to the observed variation were the three most abundant taxa (and also eutrophic indicators), namely *C. microporum*, *E. elegans*, and *A. granulata* (Table 3).

Table 3. Mean percentage dissimilarity between monthly samples from Tadlac Lake, Philippines, based on phytoplankton and taxon most responsible for the dissimilarity.

Comparison of Sampling Months		**Average Dissimilarity (%)**	**Leading Contributing Taxon for Dissimilarity**	**Percentage Contribution per Taxon**
October	November	50.81	*Eudorina*	34.47
	December	57.04	*Melosira*	65.30
	January	91.74	*Coelastrum*	39.92
	February	64.59	*Coelastrum*	43.58
	March	58.63	*Coelastrum*	41.19
November	October	50.81	*Eudorina*	34.47
	December	53.06	*Melosira*	43.77
	January	95.47	*Eudorina*	33.46
	February	76.78	*Eudorina*	34.31
	March	68.92	*Eudorina*	33.23
December	October	57.04	*Melosira*	65.3
	November	53.06	*Melosira*	43.77
	January	96.16	*Melosira*	61.81
	February	79.29	*Melosira*	63.47
	March	70.77	*Melosira*	63.49
January	October	91.74	*Coelastrum*	39.92
	November	95.47	*Eudorina*	33.46
	December	96.16	*Melosira*	61.81
	February	75.63	*Melosira*	33.46
	March	88.32	*Melosira*	40.51
February	October	64.59	*Coelastrum*	43.58
	November	76.78	*Eudorina*	34.31
	December	79.29	*Melosira*	63.47
	January	75.63	*Melosira*	33.46
	March	37.76	*Melosira*	29.87
March	October	58.63	*Coelastrum*	41.19
	November	68.92	*Eudorina*	33.23
	December	70.77	*Melosira*	63.49
	January	88.32	*Melosira*	35.78
	February	37.76	*Melosira*	29.87

In contrast to the monthly comparisons, no statistical differences were observed in phytoplankton abundance across sampling stations, as previously indicated by ANOVA. This is further supported by NMDS, which showed that abundances of phytoplankton taxa were clustered into a single distinct group at the 95 % similarity level and at a stress value of 0.07 (Figure 7). This is evident in the overlapping pattern in the phytoplankton abundance with very few separations between samples. Furthermore, findings of the ANOSIM (p>1.00: r = -0.1388) validated the NMDS results. R values can range from -1 to +1, with a value close to zero indicating no separation between groups, and +1 indicating that all similarities within groups are less than any similarity between groups (Clarke and Gorley, 2001). The pairwise value from ANOSIM (r = -0.1388) would also indicate no separation based on phytoplankton abundance between sites. This finding would suggest that the various land use activities along the lake's shoreline do not induce significant variation in the density of phytoplankton species in the water nearby.

Environmental parameters as factors determining phytoplankton assemblage

The ordination of the phytoplankton assemblages from the lake around two main CCA axes is presented in Figure 8. The parameters that influenced the lake's temporal variation of phytoplankton density were nitrates, phosphates, pH, temperature, and conductivity. The first canonical axis (55.36 % of variance, an Eigenvalue of 0.26) was positively correlated with nitrates and conductivity. Axis 2 (variance 25.2 % of variance, an Eigenvalue of 0.12) was positively correlated with temperature, pH, and phosphate. The ordination diagram shows the approximate community composition at different sampling periods. The phytoplankton community was positioned differently in the first and second planes of the canonical axes.

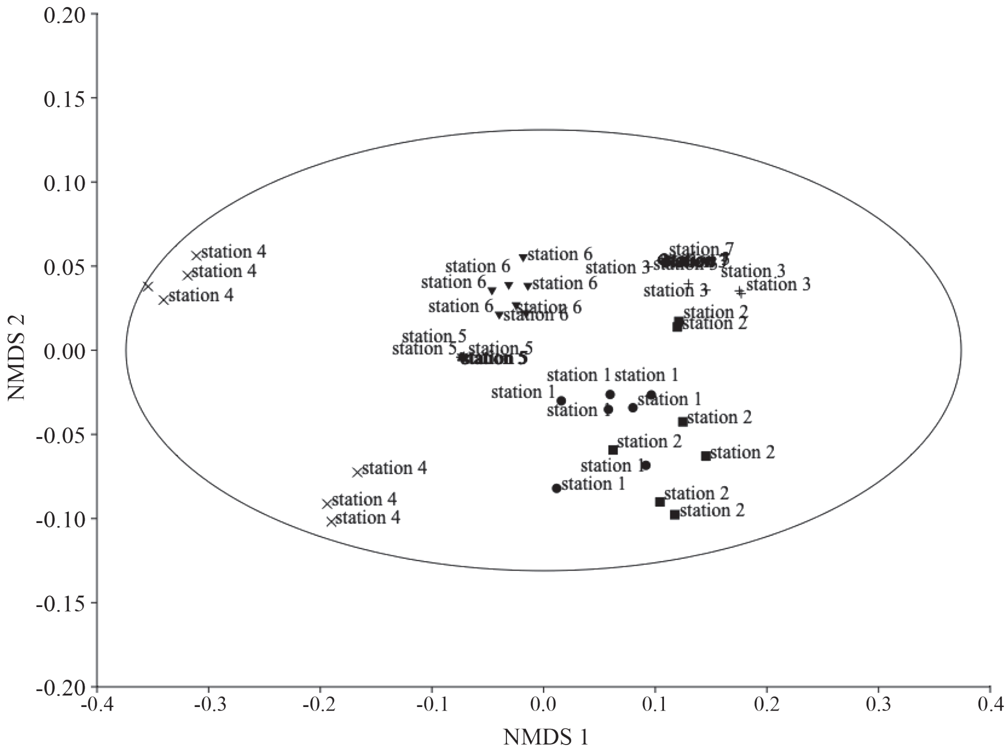

Figure 7. Multidimensional scaling ordination of sampling sites in Tadlac Lake, Philippines, based on Bray-Curtis similarities of phytoplankton abundance.

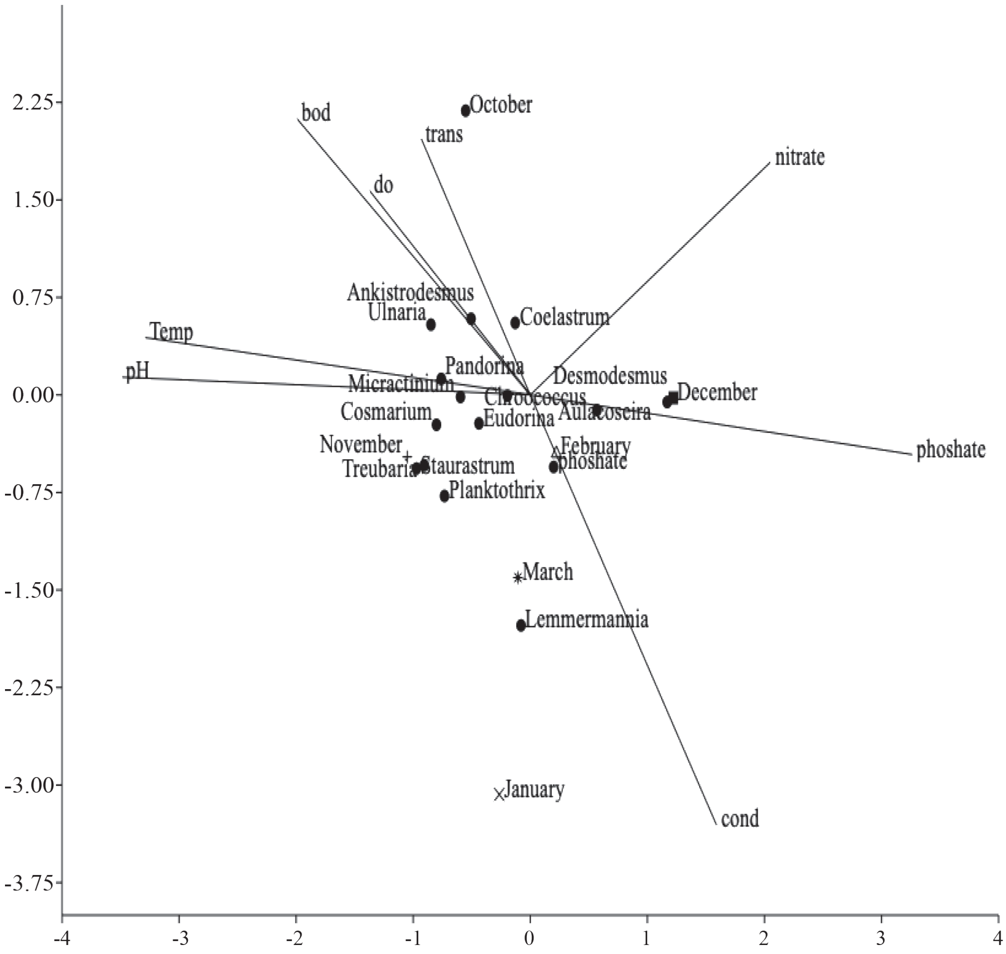

Figure 8. Canonical correspondence analysis based on the abundance of phytoplankton and environmental parameters of Tadlac Lake, Philippines, in six monthly samples.

Aulacoseira granulata, the most abundant eutrophic indicator taxon, was placed close to the vector of phosphates and nitrates and was most abundant during December. *Desmodesmus communis* was also situated near the same vector. Most of the phytoplankton taxa were distributed along the vector of temperature and pH, and include *Pandorina morum*, *Chroococcus dispersus*, *Eudorina elegans*, *Cosmarium turpinii*, *Staurastrum anatinum*, *Treubaria triappendicularia*, *Planktothrix agardhi*, and *Micractinium*; these were found to be most abundant during November when high temperature and pH were noted. *Coelastrum microporum*, another eutrophic indicator taxon, was correlated with transparency and was most abundant during October.

Canonical correspondence analysis revealed the variable response of the phytoplankton community to water quality parameters, significantly affecting its temporal distribution. This was also evident in the results of the cluster analysis and ANOSIM. The sudden drop in the phytoplankton density and species number in January and February could be primarily due to changes in nutrient availability (nitrates and phosphates), temperature, pH, and conductivity. Furthermore, the observed clustering of phytoplankton abundance (Figure 7) could be best explained by these parameters. Several studies cited that nitrogen and phosphorus nutrients (in the form of nitrates and phosphates) are the key controlling factors for algal growth (Agius and Jaccarini, 1982; Smith, 2003; Jennifer *et al*., 2008). Nitrogen contributes to the structural components, genetic material, and metabolic compounds in plant and algal cells. Nitrogen is an essential part of chlorophyll, which is responsible for photosynthesis. Meanwhile, phosphorus is a vital macronutrient for algal growth, which is the primary constituent of the membrane of algae cells, is vital for nucleic acids, and forms the backbone of adenosine triphosphate (Berg *et al*., 2002). Temperature is an essential factor in phytoplankton growth since it significantly influences algal activity by controlling the enzymatic reaction of photosynthesis (Liu, 2000). The photosynthetic rate of phytoplankton is initiated and sped up by heat (Barber, 2014), and phytoplankton metabolism is also higher at high temperatures (Toseland *et al*., 2013). Phytoplankton organisms also increase their growth rate with increasing temperature (Dawes, 1998). Changes in pH can also influence algal cell growth by altering the distribution of carbon and the availability of carbon dioxide. It can also change the availability of trace metals and essential nutrients, and at extreme pH levels, potentially cause direct physiological effects (Chen and Durbin, 1994). The alkaline pH in an aquatic system is believed to provide the optimal conditions for the favorable growth of phytoplankton since alkaline pH can affect the supply of inorganic nutrients such as nitrate and phosphate (Moyle, 1946). These factors may explain why most of the phytoplankton species were highly associated with the alkaline pH value of Tadlac Lake as illustrated by the CCA triplot.

Lastly, conductivity measures the ability of a solution like water to pass an electric current. This indicates the concentration of dissolved electrolyte particles in the water, such as nitrate, phosphate, and sodium (Behar *et al*., 1996). Li *et al*. (2005) also stated that changes in nitrogen and phosphorus concentrations lead to immediate changes in conductivity. The more eutrophication there is, the more nitrogen and phosphorus contribute to conductivity. Several studies noted that conductivity values above 100 $\mu S \cdot cm^{-1}$ indicate eutrophication because of the high degree of decomposition of organic matter, which releases a significant amount of ions into the water column (Margalef, 1984; Tundisi *et al*., 2008; Gemelgo *et al*., 2009; Silva *et al*., 2011). The measured conductivity in Tadlac Lake ranged from 847 $\mu S \cdot cm^{-1}$ to 1,048 $\mu S \cdot cm^{-1}$; this could denote high nutrient availability in the water, which could stimulate algal growth.

Biotic indices

The algal pollution index computed for October, November, and December was 16, which indicates moderate organic pollution condition (Table 4). In December, the calculated value was 13, which signifies low organic pollution; the difference was probably due to the absence of *Ankistrodesmus falcatus* and *Pandorina morum*. For February and March, the calculated index was 14. Overall, the mean algal pollution index computed for the six-month study period was 15. It signifies that Tadlac Lake is experiencing moderate organic pollution.

Table 4. Summary of calculated values for algal genus pollution index and saprobic index for Lake Tadlc, Philippines.

Month	Algal Genus Pollution Index	Saprobic Index
October	16.00	2.20
November	16.00	2.13
December	16.00	2.21
January	13.00	2.35
February	14.00	2.37
March	14.00	2.35
Mean	15.00	2.27

The calculated saprobity index values ranged from 2.13 to 2.37, having a mean of 2.27. During the six months, the calculated values fell within the β-mesosaprobic zone, representing moderate organic contamination.

As stated by Edokpayi *et al*. (2017), effluent discharge from domestic sewage, urban run-off, and farm waste are the major sources of organic pollution in aquatic ecosystems. During the sampling, it was observed that were some residential areas, an active poultry farm, and a resort (Laresio Resort) present along the banks of the lake. These establishments are possibly major contributors of organic matter loading in the lake, as they were observed disposing their waste into the lake. This could explain why the lake is still in eutrophic condition despite the removal of aquaculture activity.

CONCLUSION

A phytoplankton community assessment was carried out in Tadlac Lake. A total of 14 phytoplankton taxa were detected, spanning three major groups, namely Chlorophyta, Cyanophyta, and Bacilliariophyta. Green algae (Chlorophyta) was the most dominant among all algal groups in Tadlac Lake in terms of density and number of species. Eutrophic indicator species *Aulacoseira granulata*, *Eudorina elegans*, and *Coelastrum microporum* were recorded with the highest density among the phytoplankton species. Statistical variation in the phytoplankton density was only noted among sampling months, and was generally influenced by nutrient availability, temperature, pH, and conductivity. Water quality parameters (particularly nutrient concentrations), the abundance of eutrophic indicator species, and the calculated values of the Algal Genus Pollution Index and Saprobity Index revealed that the lake is still experiencing eutrophication despite the rehabilitation. The local government must consider the mitigation of organic matter discharge into the lake. This could help improve the trophic status and water quality of the lake.

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