Desalination and Water Treatment
www.deswater.com

doi: 10.1080/19443994.2014.930933

55 (2015) 2237–2252
August

Taylor & Francis
Taylor & Francis Group

Spatio-temporal variations of phytoplankton structure and water quality in the eutrophic freshwater reservoir of Macau

Weiying Zhang[a], Inchio Lou[a,*], Wai Kin Ung[b], Yijun Kong[b], Kai Meng Mok[a]

[a]*Faculty of Science and Technology, Department of Civil and Environmental Engineering, University of Macau, Av. Padre Tomás Pereira Taipa, Macau SAR, China, Tel. +853 8397 8469; email: jane1874@hotmail.com (W. Zhang), Tel. +853 8397 8469; Fax: +853 2883 8314; email: iclou@umac.mo (I. Lou), Tel. +853 8397 4971; email: kmmok@umac.mo (K.M. Mok)*
[b]*Laboratory and Research Center, Macao Water Co. Ltd., 718, Avenida do Conselheiro Borja, Macau SAR, China, Tel. +853 8299 0603; email: kin.ung@macaowater.com (W.K. Ung), Tel. +853 8299 0652; email: edwards.kong@macaowater.com (Y. Kong)*

Received 13 October 2013; Accepted 29 May 2014

ABSTRACT

In small-scale pumped-storage reservoirs, physical disturbances have been suggested to be one of the main factors influencing phytoplankton structure and water quality. This study presented data on dynamic changes of the phytoplankton structure and the quality of raw water sampled monthly from January 2011 to June 2012, in three locations (with two different water levels each) of a small pumped-storage reservoir of Macau main storage reservoir. The trophic state index, phytoplankton structure indices, and multivariate statistical techniques were applied for assessing trophic state, phytoplankton community, and spatio-temporal variations of the reservoir, respectively. The results showed that the reservoir was categorized as a eutrophic–hypereutrophic reservoir, with the dominance of *Cyanophyta* in 2011, and of *Chlorophyta* and *Bacillariophyta* in 2012. Lowest diversity/evenness and highest dominance happened in June 2011, while highest diversity/evenness and lowest dominance occurred in May 2012. Principle component analysis identified four factors that can explain 80.8% of the total variance of the water quality data, and cluster analysis generated two clusters of spatial similarity among the six sampling points and two clusters of temporal similarity among the 18 months. Discriminant analysis results revealed only three parameters (TP, NO_3-N, and Chl-a) that could afford 100% correct assignation in temporal analysis, while no spatial variation was found in spatial analysis. This study highlighted the usefulness of combination of these methods for the evaluation and interpretation of complex water quality data-sets and assessment of pollution level of small-scale eutrophic reservoirs. The results from the study can be used in developing monitoring program of freshwater bodies.

Keywords: Spatio-temporal variations; Phytoplankton structure; Water quality; Multivariate statistical techniques; Freshwater reservoirs

---

*Corresponding author.

1. Introduction

Phytoplankton structure and water quality in freshwater eutrophic reservoirs are becoming a serious concern, which may lead to proliferation of harmful phytoplankton (called algal blooms) under favorable conditions. Algal blooms disturb the ecosystem, deteriorate the water quality, and most importantly produce cyanotoxins that pose a serious health hazard for humans [1,2]. To better understand the problem of algal blooms and prevent their occurrences, it is imperative to analyze the phytoplankton structure and the corresponding water quality. Understanding the spatial and temporal variations of phytoplankton community and water quality could help in developing regular monitoring program that is a helpful tool not only to evaluate the impacts of pollution sources but also to ensure an efficient management of water resources and the protection of aquatic ecosystems.

Human activities such as periodic pulses of mixing have influenced reservoirs features including retention times and water level fluctuations [3]. Due to physical disturbances in small-scale pumped-storage eutrophic reservoirs, changes of phytoplankton community structure in response to water quality tend to be more pronounced [4,5]. These changes can reveal important aspects of susceptibilities and tolerances of the species present that form the community, which is fundamental to biological monitoring of the reservoirs. Previous studies on reservoirs have generally dealt with the structural compositions of the phytoplankton community, focusing on the taxonomic surveys and ecological studies [6–8]. Diversity and community comparison indices of Shannon–Wiener diversity, Simpson diversity, Pielou's evenness, Stander's similarity index (SIMI), and Margalef's richness have been recognized as useful ways for analyzing phytoplankton structure in different water bodies all over the world [9–12].

For analyzing the complex data-set obtained from large numbers of samples and water quality parameters at different times and places, univariate and bivariate statistical techniques were traditionally used, which could be far from adequate. Recently, multivariate statistical techniques including principle component analysis (PCA), cluster analysis (CA), and discriminant analysis (DA) have been proved to be more helpful in the interpretation of complex data matrices to better understand the water quality. These methods have been employed to evaluate and examine the spatial and temporal variations and trends in water bodies [13–15]. PCA is a very powerful technique applied to reduce the dimensionality of a data-set consisting of a large number of inter-related variables, while retaining as much as possible the variability present in data-set. It uses an orthogonal transformation to convert a set of observations of possibly correlated variables into a set of values of uncorrelated variables called PCs, thus reducing the complexity of multidimensional system by maximization of component loadings variance and elimination of invalid components. PCA technique was previously used alone or in combination with other methods, to simplify the interpretation of the relationship within complex data-set and model aquatic, environmental, and ecological processes [16,17]. In analyzing the temporal and spatial difference of the samples, CA and DA analyses were applied. CA can group objects (cases) into classes (clusters) on the basis of similarities within a class and dissimilarities between different classes, so that the objects in the same cluster are more similar to each other than to those in other clusters, helping in mining the data and indicate patterns [18]. DA provides statistical classification of samples and it is performed with prior knowledge of membership of objects to particular group or cluster (such as temporal or spatial grouping of a sample is known from its sampling time or site). The DA results are able to group the samples sharing common properties and help in prediction [19].

Main storage reservoir (MSR), a small pumped MSR of Macau, was reported to have experienced increasing frequency of algal blooms, with high concentrations of *Cylindrospermopsis* spp. and *Microcystis* spp. which were producing cyanotoxins [20]. However, detailed information about the phytoplankton composition and the spatio-temporal variations in water quality of MSR are still lacking. There is also only sparse literature integrating phytoplankton structure indices and multiple statistical analysis for studying the spatio-temporal variation of phytoplankton community and water quality in small-size freshwater reservoirs. We hypothesized that phytoplankton composition and water quality would be affected greatly in such small-scale ecosystem. The objectives of this study were to analyze the spatio-temporal variations of phytoplankton structure including dominant species, diversity, evenness, and similarity, and to perform multivariate statistical techniques (PCA, CA, and DA) for dynamic changes of water quality in MSR. These results will be used in evaluating the pollution level and developing a water monitoring program in the reservoir.

2. Materials and methods

2.1. Site description and sampling

MSR (22º12´12´´N, 113º33´´12´´E), located in the east part of Macau peninsula, is the biggest reservoir in

Macau, with the capacity of about 1.9 million m^3 and the water surface area of 0.35 km^2. It is a small pumped-storage reservoir that receives raw water from the West River of the Pearl River network and can provide water supply to the whole area of Macau for about one week. MSR is particularly important as the temporary water source during the salty tide period, when high salinity concentration is caused by intrusion of sea water to the water intake location. However, algal bloom problems occur from time to time in the summers, and the situation appeared to be more worsening in recent years, with high phytoplankton abundance in which *Microcystis* spp. and *Cylindrospermopsis* spp. were detected as the dominant species.

During the study period, water samples were collected monthly in MSR from January 2011 to June 2012. Six sampling points were selected at three stations S1–S3 (Fig. 1) with two different water depths (0.5 and 3.5 m below the water surface) each. Stations S1 and S3 are located in the inlet and outlet, respectively, while station S2 is at the center of MSR. P1 (P2), P3 (P4), and P5 (P6) are defined as the sampling points at 0.5 m (3.5 m) below the water surface at station S1, S2, and S3, respectively.

2.2. Water quality parameters

2.2.1. Abiotic parameters

Sampling, preservation, and transportation of the water samples to the laboratory were performed according to standard methods [21]. The samples were analyzed for 15 abiotic parameters including water temperature (WT), secchi depth (SD), electrical conductivity (EC), pH, dissolved oxygen (DO), total nitrogen (TN), nitrate nitrogen (NO_3-N), nitrite nitrogen (NO_2-N), ammonia nitrogen (NH_3-N), total phosphorus (TP), orthophosphate phosphorus, chlorophyll-a (Chl-a), microcystin (MC) and cylindrospermopsin (CYN) concentrations, and precipitation. Precipitation was obtained from Macau Meteorological Center (http://www.smg.gov.mo/www/te_smgmail.php).
WT and SD were measured *in situ* with a mercury thermometer and a Secchi disk. pH was determined in the laboratory with a pH meter (DKKTOA, HM-30R). Conductivity was measured with an EC meter (DKK-TOA, CM-30R). DO, NH_3-N, NO_3-N, NO_2-N, TN, TP, and PO_4^{3-} were measured according to the standard methods [21]. Chl-a was determined by UV–vis recording Spectrophotometer (SHIMADZU, UV-2401PC). Identification of the planktons was conducted following Smith [22]. MC and CYN concentrations were measured using HPLC technique.

2.2.2. Biotic parameters

The phytoplankton samples were immediately fixed using Lugol's iodine solution for phytoplankton counting with an inverted microscope following the method of McAlice [23]. After 72 h sedimentation, algal species were identified based on morphological

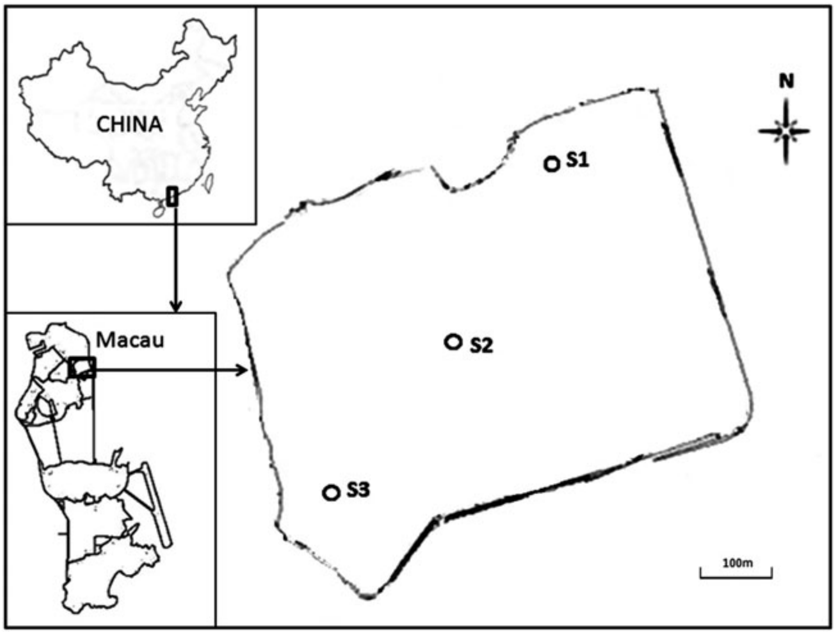

Fig. 1. Layout of MSR and location of water sampling.

criteria and quantified at ×100 magnification along a Sedgewick-Rafter chamber, according to the method described by Utermöhl [24].

2.3. Data analysis

2.3.1. Trophic state index

Trophic status was assessed using trophic state index (TSI), the most commonly used index, to describe trophic levels of lakes and reservoirs. The overall TSI was calculated based on TP concentration, Chl-a concentration, and SD, according to the following equations [25].

$$\mathrm{TSI(SD)} = 10 \times \left(6 - \frac{\ln \mathrm{SD}}{\ln 2}\right) \tag{1}$$

$$\mathrm{TSI(TP)} = 10 \times \left(6 - \frac{\ln\left(\frac{48}{\mathrm{TP}}\right)}{\ln 2}\right) \tag{2}$$

$$\mathrm{TSI(Chl)} = 10 \times \left(6 - \frac{2.04 - 0.68 \ln \mathrm{Chl}}{\ln 2}\right) \tag{3}$$

$$\mathrm{TSI(overall)} = \frac{\mathrm{TSI(SD)} + \mathrm{TSI(TP)} + \mathrm{TSI(Chl)}}{3} \tag{4}$$

Four classes, oligotrophic, mesotrophic, eutrophic, and hypereutrophic states with the corresponding TSI of < 30–40, 40–50, 50–70, 70–100+ are defined, from low to high primary productivity.

2.3.2. Community comparison indices

(1) The diversity was estimated using the Shannon and Wiener index [26]:

$$H' = -\sum_{i=1}^{s} p_i \log_2 p_i \tag{5}$$

where p_i is the proportion of individuals in species i and s is the number of species encountered.

(2) The Simpson index (D) of diversity was first introduced by Simpson [27], and it is used to measure the degree of dominance:

$$D = \sum_{i=1}^{s} p_i^2 \tag{6}$$

(3) The evenness was assessed using H' [28] as follows:

$$J' = \frac{H'}{\log_2 s} \tag{7}$$

where H' is the Shannon's index in a sample and s is the number of species.

(4) Margalef's richness [29] index:

$$\frac{s-1}{\ln N_i} \tag{8}$$

where s is the number of species and N_i is the number of individuals. The richness referred to the number of algal taxa registered in each sample. Generally, in a healthy environment, Margalef's richness index is higher in the range of 2.5–3.5 [30].

(5) The Stander's [31] SIMI is calculated to compare two successive phytoplankton communities:

$$\mathrm{SIMI} = \frac{\sum a_i b_i}{\sqrt{\sum a_i^2 \sum b_i^2}} \tag{9}$$

where a_i is the ratio of the number of individuals of species i to the total number of individuals N in sample A; b_i is the ratio of the number of individuals of species i to the total number of individuals N in sample B; and s is the total number of species in both samples. The range of SIMI value is from 0 (no similarity) to 1 (identical). The criteria [32] can be divided into five categories to evaluate the meaning of the SIMI index: 0.00–0.199 represents dissimilarity, 0.20–0.499 is low similarity, 0.50–0.699 is medium similarity, 0.70–0.899 is similarity, and 0.90–0.999 is high similarity. Our study period was divided into six groups (January–Mar 2011, April–June 2011, July–September 2011, October–December 2011, January–March 2012, and April–June 2012) to compare the similarity and quantify the differences in the kinds of species present and their abundance data.

2.3.3. Statistical analysis

Statistical analyses (PCA, CA and DA) were carried out using PASW 19 software package (SPSS Inc.). DA was applied to the raw data, whereas PCA and CA were performed on standardized data through normalized transformation due to the wide ranges of data dimensionality and different units of measurements [15,33,34].

2.4. Principal component analysis

Kaiser–Meyer–Olkin (KMO) and Bartlett's sphericity tests were performed to examine the suitability of the data for PCA [15,19]. KMO is a measure of sampling adequacy which indicates the proportion of variance that is common, while Bartlett's test of sphericity indicates whether a correlation matrix is an identity matrix, which would indicate that variables are unrelated. The significance level indicated that whether there were significant relationships among the variables. In our case, the selected 14 variables with complete data-set were accessed with KMO and Bartlett's test of sphericity to verify the applicability of PCA. Only parameters with communalities great than 0.5 were used for analysis.

2.5. Cluster analysis

The purpose of CA is to group a set of objects into different clusters based on their similarity to each other. It is the most common approach to decide which clusters should be combined or formed. The clusters are formed sequentially by starting with the most similar pair of objects and grouping higher clusters in a step-by-step method. The Ward's method with Euclidean distance [35] is usually applied to show similarities between two samples, and a "distance" can be represented by the "difference" between analytical values from both of the samples. CA determines the variability of the data-set using the linkage distance, which is expressed as Dlink/Dmax. It is traditional to use the quotient multiplied by 100 as a way to standardize the linkage distance [36]. In our case, CA was applied to the water quality data-set to group the similar spatial (6 points) and temporal (18 months) among all the water samples, resulting in the spatial and temporal dendrograms.

2.6. Discriminant analysis

DA is the analysis used to determine which continuous variables discriminate between two or more naturally occurring groups. The details have been described in previous studies [34,37]. It performs analysis on raw data and its technique set up a discriminant function (DF) for each group, as written by the following equation:

$$f(G_i) = k_i + \sum_{j=1}^{n} W_{ij} P_{ij} \tag{10}$$

where i is the number of groups (G), k_i is the constant inherent to each group, n is the number of parameters used to classify a set of data into a given group, W_j is the weight coefficient, assigned by DA to a given selected parameters (P_j). However, there is only one DF for a 2-group DA. Wilk's lambda is used to test if the discriminant model is significant, where "Sig." p value <0.05 is required. If DA is effective for a set of data, the classification table of correct and incorrect estimates will yield a high correct percentage. In this study, DA was performed on grouped periods and sections based on the CA results.

3. Results

3.1. Water quality in MSR

3.1.1. Abiotic parameters

The rainfall in the study period (data not shown here) was 2,131.8 mm yearly in average, with two peaks in the summers (June 2011 and April 2012) and nearly no precipitation in the dry and cold seasons. Other water quality parameters were measured and summarized in Fig. 2. It was showed that the physical parameters, temperature, SD, conductivity, and Chl-a fluctuated from time to time, while DO and pH maintained relatively stable (Fig. 2(a)). Variations in nitrogen and phosphorus concentrations (Fig. 2(b) and (c)) were observed during the study. High concentrations of TN and TP were observed from July to January, resulting in high phytoplankton density in summer (Fig. 3). Low phytoplankton abundance from November to January was because of the low temperature in winter which is another more important factor affecting the micro-organisms' growth. Furthermore, low TN/TP ratio (<10) for most time was found in MSR, thus favored the blue algae (Fig. 3). The concentrations of CYN and MC (Fig. 2(d)) showed one peak in February 2011, after which both concentrations decreased to nearly zero in March and April of 2011. Then both cyanotoxins showed different behaviors: the CYN dramatically increased and kept at a high level until the end of 2011, while MC maintained at a low level until the April of 2012 and started to increase in May and June. These results were consistent with the dynamic changes of the corresponding species, *Cylindrospermopsis* spp. and *Microcystis* spp. (Fig. 5), particularly *Microcystis* spp. maintained an extremely low level of cell number during the whole year of 2011 and gradually increased in 2012, while *Cylindrospermopsis* spp. had high concentrations in 2011 and dramatically decreased in 2012.

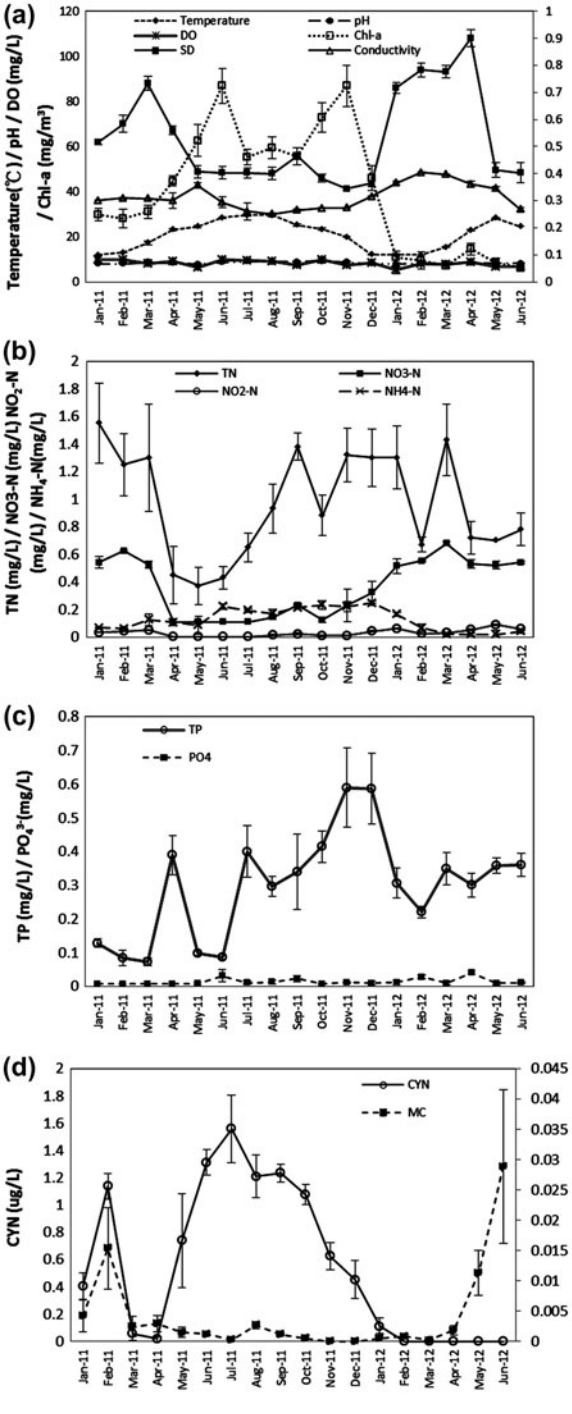

Fig. 2. Variations of 14 abiotic water parameters, with (a) temperature, pH, DO, Chl-a, SD, and conductivity; (b) TN, NO_3^--N, NO_2^--N, and NH_4^+-N; (c) TP and PO_4^{3-}; and (d) CYN and MC. The error bars represented the standard deviations of the six samples.

The TSI (SD), TSI (TP), and TSI (Chl) of the reservoir were calculated as 62–75, 66–96, and 49–74, respectively, from which the overall TSI was estimated as 65–82 by taking the average of the three values. The results indicated that MSR was categorized as a reservoir between eutrophic and hypereutrophic status.

3.2. MSR phytoplankton community

3.2.1. Phytoplankton species compositions and densities

Thirty-five taxa from the six divisions including *Cyanophyta, Chlorophyta, Bacillariophyta, Cryptophyta, Pyrrophyta*, and *Euglenophyta* were identified and shown in Table 1.

Cyanophyta, Chlorophyta, and *Bacillariophyta* were the most important phytoplankton constituents, occupying more than 95% of the total phytoplankton, with small variations of the monthly counting data among different sampling points observed (Fig. 3). It should be noted that *Cyanophyta* was dominant in 2011, especially during the summer and autumn, while *Chlorophyta* and *Bacillariophyta* were the dominant constitutes in 2012 (Fig. 4). Further species-level microscopic counting results indicated that *Pseudanabaena* spp., *Cylindrospermopsis* spp., *Dactylococcopsis* spp., *Merismopedia* spp., *Scenedesmus* spp., and *Chlorella* spp. were the dominant species in the study period (Fig. 5). Starting from March 2011, cyanobacteria dominated with relative abundances more than 95%. It became dominant very rapidly and remained at high level until the end of the year. Though *Pseudanabaena* spp. was the dominant species of cyanobacteria for most of the time, it was not considered to be the notorious species, as it releases no cyanotoxins, reported from literature, but may only clog the filters during the treatment process. Different from *Microcystis* spp. that was the dominant species causing algal blooms in previous years, *Cylindrospermopsis raciborskii* was the dominant toxic species in the study period. It was also found that, compared to that of 2011, phytoplankton abundance in the first half of the year 2012 was dramatically decreased, which was due to the partial change of source water from the mainland China. This change would definitely affect the water quality, thus resulting in the change of phytoplankton structure, in the small-scale pumped-storage reservoir.

The total phytoplankton maintained at a high level during April–November 2011, which was positively correlated with temperature ($r_s = 0.58$, $p < 0.01$), pH ($r_s = 0.73$, $p < 0.01$), NH_4-N ($r_s = 0.74$, $p < 0.01$), and Chl-a ($r_s = 0.90$, $p < 0.01$), and anti-correlated with SD ($r_s = -0.56$, $p < 0.01$), conductivity ($r_s = -0.62$, $p < 0.01$), NO_3-N ($r_s = -0.88$, $p < 0.01$), and NO_2-N ($r_s = -0.76$, $p < 0.01$). These results suggested that the most important water parameters associated with the

Table 1
Phytoplankton species composition in MSR

Phyla	Species composition
Cyanophyta	*Pseudanabaena galeata*, *Cylindrospermopsis* spp., *Planktothrix* spp., *Dactylococcopsis* spp., *Merismopedia* spp., *Chroococcus* spp., *Microcystis* spp., *Oscillatoria* spp., *Aphanocapsa* spp., *Anabaena* spp.
Chlorophyta	*Scenedesmus* spp., *Chlorella* spp., *Tetraedron minimum*, *Ankistrodesmus falcatus*, *Chlamydomonas* spp., *Schroederia* spp., *Cosmarium* spp., *Selenastrum* spp., *Oocystia* spp., *Pediastrum* spp., *Coelastrum* spp., *Staurastrum* spp., *Tetraedron caudatum*, *Micractinium* spp., *Westella* spp.
Bacillariophyta	*Cyclotella* spp., *Achnanthes* spp., *Navicula* spp., *Fragilaria* spp., *Aulacoseira granulata*
Cryptophyta	*Cryptomonads* spp.
Pyrrophyta	*Peridinium* spp., *Ceratium hirundinella*
Euglenophyta	*Phacus* spp., *Trachelomonas* spp.

development of the algal blooms were temperature, pH, nitrogen source, and conductivity. Besides, the high correlation between phytoplankton abundances and Chl-a concentrations confirmed that the Chl-a is a good indicator of the phytoplankton measurement, reflecting the abundance of algae in the reservoirs.

3.2.2. Diversity, dominance, evenness, richness, and similarity

The seasonal variations in phytoplankton species were estimated using five indices, Shannon and Wiener index, Simpson index, Evenness index, Margalef's richness, and Stander's SIMI, to reveal the diversity, dominance, evenness, richness, and similarity of MSR, respectively, and the results were summarized in Fig. 6. The indices calculation was based on the number of individuals for each species, thus showing more information and further relationship of all the identified species than those of the only three dominant phyla as shown in Figs. 3 and 4. Small spatial variations of the monthly calculated indices data were observed, indicating that there is no much difference of those indices among different sampling points.

It was showed that diversity and dominance varied irregularly throughout the study period. The highest diversity ($H' = 3.09$) and the lowest dominance ($D = 0.19$) occurred in May 2012, while the lowest diversity ($H' = 0.29$) and highest dominance ($D = 0.94$) happened in June 2011.

Pielou's index revealed the evenness of distribution of various species in the samples. Our results showed that the evenness indices had a similar pattern to the diversity indices, indicating that the phytoplankton community was satisfactorily even in May 2012 ($J' = 0.73$), while it was uneven in June 2011 ($J' = 0.07$). These results could be explained by the number of countable species and their percentage (Fig. 5). *Pseudanabaena* spp. had a peak in June 2011, occupying 96.89% of the total phytoplankton, i.e. other species only had a very small portion, resulting in low diversity and evenness. On the contrary, in May 2012

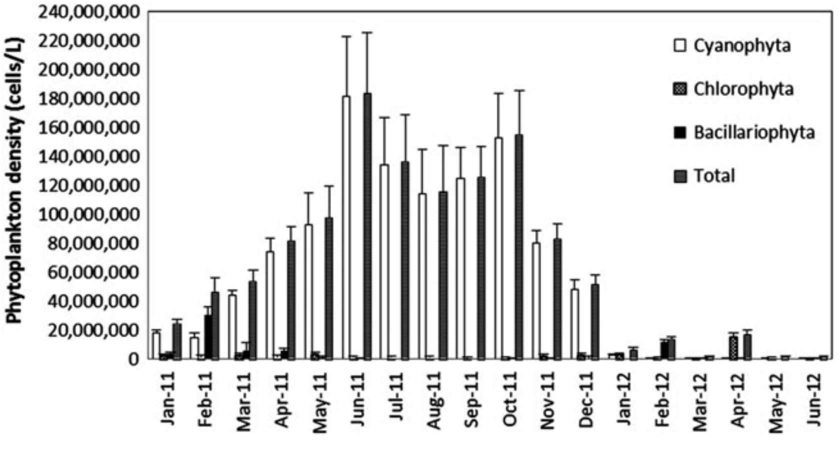

Fig. 3. Densities of phytoplankton and principal phyla during study period. The error bars represented the standard deviations of the six sampling points.

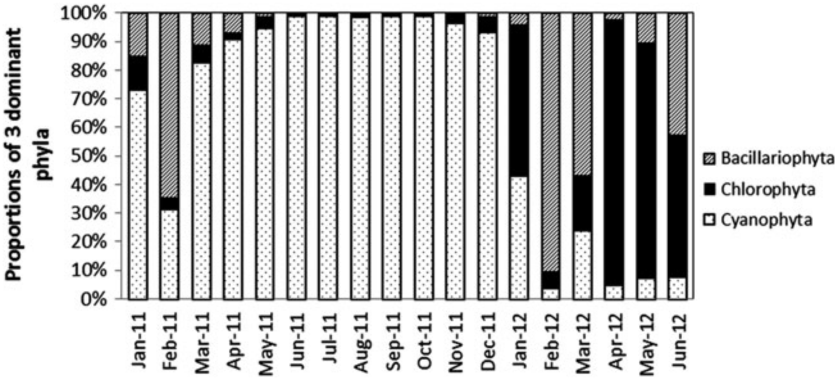

Fig. 4. Monthly proportions of *Cyanophyta*, *Chlorophyta*, and *Bacillariophyta* in total phytoplankton.

the total phytoplankton grew in low density (Fig. 3), with the species appeared at balanced percentages (33.78% *Scenedesmus* spp., 9.44% *Tetraedron minimum*, 6.95% *Cyclotella* spp., 4.97% *Chlorella* spp., and 4.47% *Pseudanabaena* spp. of the total population), leading to the highest levels of diversity and evenness.

The Margalef's richness index in MSR was estimated as 0.43–1.24 (Fig. 6), which was far below the range of 2.5–3.5 as reported by Khan et al. [30].

The SIMI results varied irregularly in our study. Comparison between two pairs of each 3-month period (Table 2) showed that MSR had similarity or high similarity (>0.7) in 2011. However, there was much dissimilarity or very low similarity (<0.2) between January–March 2012 and April–June 2012, indicating that the variations of phytoplankton species were high in 2012. This result was consistent with the variations of densities and percentages of the phytoplankton (Figs. 3 and 4), which was probably due to the partial change of the source water from the Mainland China in the January of 2012.

Further statistical analysis indicated that the diversity was positively correlated with evenness ($r_s = 0.98$, $p < 0.01$), while anti-correlated with both the dominance ($r_s = -0.88$, $p < 0.01$) and phytoplankton density ($r_s = -0.73$, $p < 0.01$). However, the correlation between diversity and Margalef's richness was not high ($r_s = 0.47$, $p < 0.01$).

3.3. Principal component analysis

The PCA was performed on 14 selected variables (13 abiotic parameters and 1 biotic parameter) and total phytoplankton to compare the compositional pattern between the water samples and determine the factors influencing each other in MSR. The value of KMO was 0.602, which is above the criteria value of 0.6. The value of χ^2, calculated as 210.06 with *p*-value

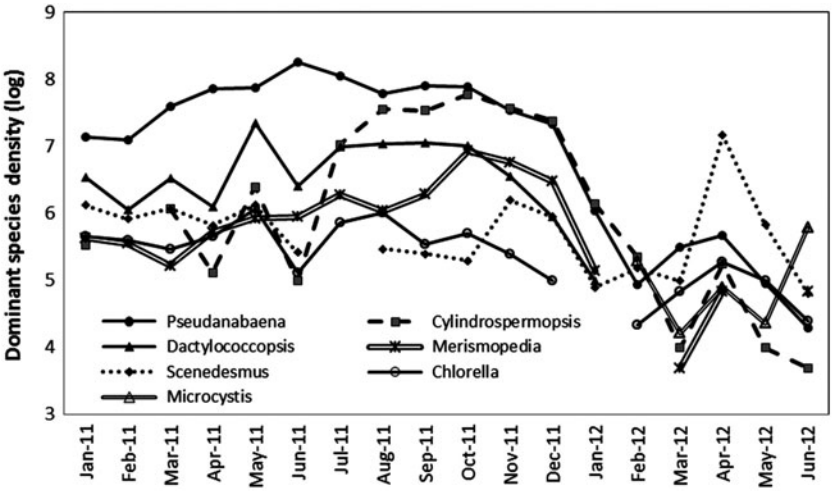

Fig. 5. Densities of dominant species (belonging to *Cyanophyta*, *Chlorophyta*, and *Bacillariophyta*) during the study period.

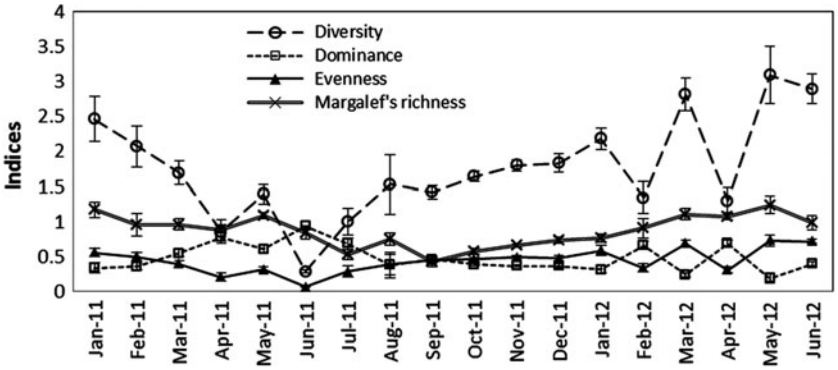

Fig. 6. Indices of diversity, dominance, evenness, and richness over time. The error bars represented the standard deviations of the six sampling points.

Table 2
SIMI comparisons in the study period

		2011				2012	
		January–March	April–June	July–September	October–December	January–March	April–June
2011	January–March	1					
	April–June	0.910	1				
	July–September	0.873	0.955	1			
	October–December	0.686	0.744	0.906	1		
2012	January–March	0.214	0.134	0.155	0.185	1	
	April–June	0.086	0.046	0.042	0.053	0.105	1

less than 0.0005 by Bartlett's test of sphericity test, indicated that the analysis was applicable [38]. The scree test suggested that there were four components (PC) with the eigen values greater than 1, in which all the 14 variables were included. PCA explained 80.84% of the variance from the total data in PC1–PC4. The biplot (Fig. 7) indicated that PC1 (44.92%) was mainly composed of nitrogen sources, physical, and biological parameters. PC2 (17.77%) was mostly influenced by climatic parameters and TN. PC3 (10.13%) and PC4 (8.03%) were defined as the phosphorus source and SD (not shown here), respectively. The results from PCA suggested that most of the variations can be explained by the nutrients, physical, and soluble salts. Though PCA did not reduce the number of data in this study (as all the variables were included), it served as a means to identify those parameters that had the greatest contribution to variation in the water quality of reservoirs and indicated possible sets of pollutant sources.

3.4. Temporal variations

3.4.1. Cluster analysis

The difference of all the sampling months was represented by the length of each two branch lines as shown in the dendrogram: the greater the length, the greater the difference. Temporal CA generated the dendrogram (Fig. 8) that grouped the 18 months into two clusters at (Dlink/Dmax) *100 = 20, with Cluster 1 in April–December 2011, and Cluster 2 in January–March 2011 and January–June 2012. The two clusters classification was in accordance with the cyanobacteria-blooming period and cyanobacteria-depression period (Figs. 3 and 4) and was also consistent with the corresponding SD, conductivity, NO_3-N, NH_4-N, and Chl-a (Fig. 2) in both the periods, implying that sampling only during these two periods in a year possibly sufficed for assessment of temporal variations in water quality of MSR, assuming that other factors were insignificant.

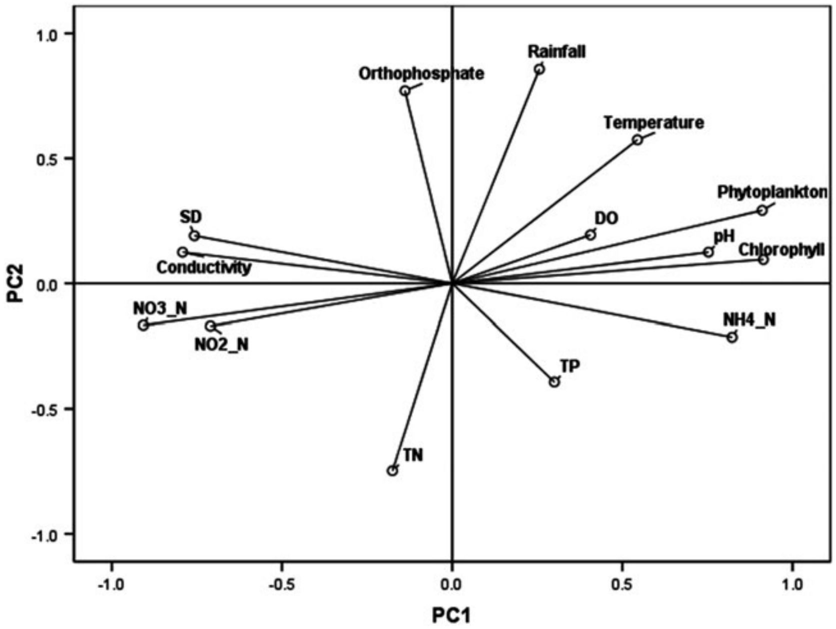

Fig. 7. PCA results in the ordination space of the first and second PCA axis (PC1 and PC2).

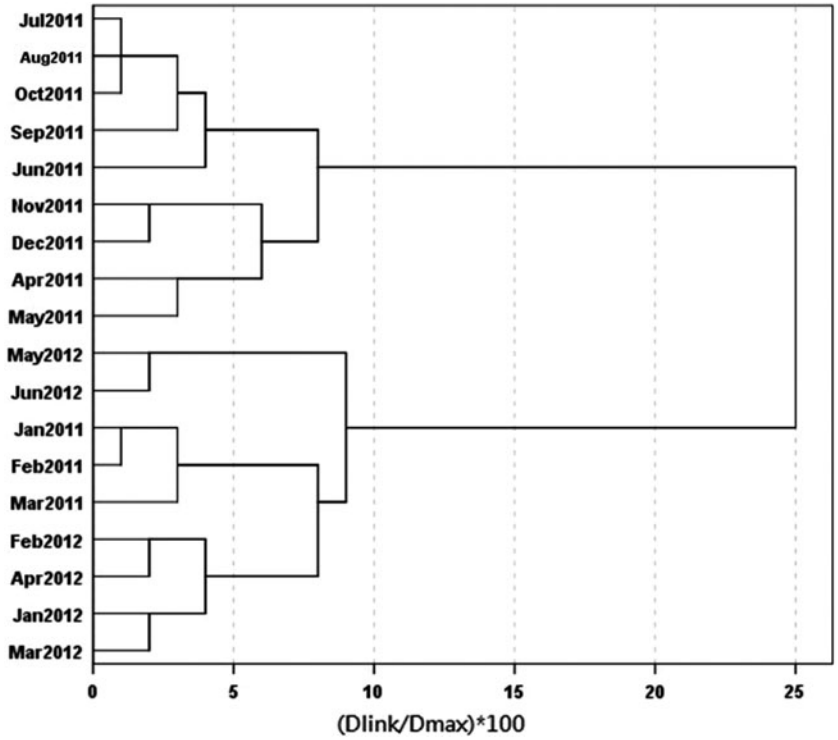

Fig. 8. Dendrogram showing hierarchical clustering of monitoring periods according to Ward’s method with Euclidean distance (CA).

3.4.2. Discriminant analysis

Temporal DA was performed on the two period clusters (April–December 2011/January–March 2011 and January–June 2012). DFs obtained from the standard and stepwise modes of DA were listed in Table 3, in which in the standard mode all water parameters were included, while in the stepwise mode, only three parameters were considered, thus reducing variables in the further analysis. However, whatever the standard mode or stepwise mode was used, it rendered the corresponding classification matrices (CMs) assigning 100% cases correctly. These results showed that TP, NO_3-N, and Chl-a were the most significant variables to discriminate between the two periods, i.e. accounting for most of the expected temporal variations in MSR. Therefore, nitrogen and phosphorus sources may contribute to the concentration of Chl-a, the indicator of phytoplankton abundance.

3.5. Spatial variations

3.5.1. Cluster analysis

Spatial CA results were shown in a dendrogram (Fig. 9) where all six sampling points on the reservoirs were grouped into two statistically significant clusters at (Dlink/Dmax) *100 = 20. Cluster 1 consisted of four sampling points (p1, p2, p3, and p4, i.e. the center and the inlet of MSR) and cluster two consisted of the other points (p5 and p6, i.e. the outlet of MSR). These results showed that there was a certain level of spatial fluctuation on the water quality between the inlet/center and the outlet, in spite of the small-scale reservoir of MSR.

Both Figs. 8 and 9 confirmed that MSR is a small pumped-storage reservoir with no significant spatial difference but with high temporal difference. Thus, this information could provide a guide for the Macao Water Utility to monitor the water quality only at different times without taking samples at different locations.

Table 3
Classification functions for DA of temporal variations in MSR

	Coefficient	
Parameters	Standard mode Function	Stepwise mode Function
Temperature	0.709	
SD	14.159	
Conductivity	0.173	
pH	0.748	
DO	2.430	
TN	16.720	
TP	3.582	5.327
NO_3-N	−71.442	−14.276
NO_2-N	0.023	
NH_4-N	43.465	
Orthophosphate	148.374	
Chl-a	0.126	0.046
MC	817.505	
CYN	1.544	
Rainfall	−0.006	
Phytoplankton	0.000	
(Constant)	−103.023	1.801
Sig.	0.005	0.000

3.5.2. Discriminant analysis

Spatial DA was also performed on the two section clusters (p1-p2-p3-p4/p5-p6) using standard and stepwise modes. However, the results using both modes showed the significance level approaching to 1, i.e. failing to complete DA. The reason was mainly because in such a small pumped-reservoir of MSR, water quality characteristics at different points were similar. This result was also consistent with that performed by CA where the water parameters in MSR are relatively uniform, and nearly no spatial variations were found between the different sampling points.

Based on the results from CA and DA, we can conclude that there is no spatial difference for water quality variables, while much difference between different periods exists. It is recommended that, in the future water quality monitoring program and strategy, more efforts should be placed to increase the sampling frequency at different times, instead of increasing sampling points of MSR. Probably a couple of sampling points are enough to extract enough information for further analysis.

4. Discussion

MSR is a small pumped-storage reservoir with a short hydraulic retention time (HRT) (~90 d), thus its water qualities are greatly influenced by the external source water and the anthropogenic activities. Due to the change of source water from the Mainland China in the January of 2012, the SD and Chl-a concentration varied dramatically from 0.4 to 0.75 m and 45 to 10 mg/m^3, respectively. Our overall monthly TSI results indicated that MSR was classified as eutrophic reservoir in rainy and cold seasons (December–March) and as hypereutrophic reservoir in rainy and hot seasons (April–November). These findings were similar to our previous study [20], which can be explained by

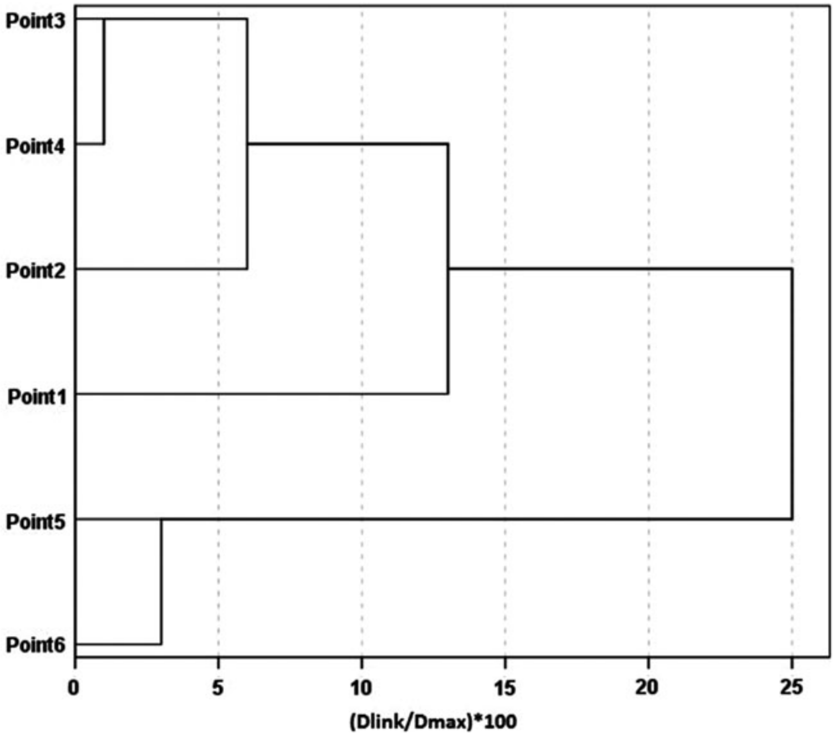

Fig. 9. Dendrogram showing hierarchical clustering of monitoring sites according to Ward's method with Euclidean distance (CA).

that during the rainy and hot period, it increases the risk of the pollutants from the land that are washed out and enter the reservoir, resulting in high level of eutrophic state in the small-scale ecosystem. In addition, the maximum density of *Cyanophyta* (April–November) can influence the TSI and water quality in reservoir. Katsiapi [39] showed that high percentage of cyanobacteria deteriorates the water quality from good to moderate, based on the Alert Levels Framework established by the World Health Organization.

Due to the small capacity of the reservoir, only 35 phytoplankton species are found, which showed fewer taxa than the large water bodies with the species more than 100. Besides, the Margalef's richness index in our study was far below the range of 2.5–3.5 stated by Khan et al. [30], indicating that the biodiversity level of MSR was not high enough. The report on low values for phytoplankton species and biodiversity indices in small eutrophic reservoir is common, as environmental conditions in a situation of trophy tend to favor a small number of species that have large densities and alternate in the dominance of the community [40]. Other researches [41,42] also found that compared with mesotrophic reservoirs in which coexistence of more species is possible, a trend to lower diversity indices in small-scale eutrophic reservoir further confirmed that diversity was more affected by the evenness than by phytoplankton density and richness [43].

Cyanophyta, *Chlorophyta*, and *Bacillariophyta* were the most important phytoplankton constituents in MSR, which was observed in other trophic reservoirs [7,12]. Both the total phytoplankton and *Cyanophyta* exhibited larger densities during hypereutrophic period of 2011, and the dominance of cyanobacteria in eutrophic reservoirs has been mentioned in the previous studies [12,44,45], involving the factors to favor cyanobacterial blooms: high nutrient status and high WT [45–47]. Interestingly, starting from the late summer to the end of 2011, *Cylindrospermopsis* spp. increased dramatically and became one of the dominant species, coexisting with *Pseudanabaena* spp. High temperature seems to be essential for *Cylindrospermopsis* spp. to develop [48], and perennial populations have been observed in tropical areas [49–51]. *Cylindrospermopsis* spp. allows it to grow in relatively low levels of phosphate [52,53], and their preferred nitrogen source is ammonium [48,52], which also happened in MSR. It was also noted that *Microcystis* spp. was not counted in most of the time during 2011, while it was the common species in 2012. This is probably because *Cylindrospermopsis raciborskii*, the dominant toxic species in 2011, release high concentration of CYN that reduced the growth of *Microcystis* spp., as CYN is able to inhibit the protein synthesis of phytoplankton [54]. Furthermore, the two important cyanotoxin-producing species, *Cylindrospermopsis raciborskii* and *Microcystis* spp., alternatively became the dominant toxic species in such a relatively simple ecosystem as MSR. Though the mechanisms of such changes are still unclear, it should be taken into consideration when developing the monitoring program of the raw water and treated water, particularly for *Cylindrospermopsis raciborskii* whose releasing toxin (CYN) is not a regulatory parameter.

Phytoplankton experienced low growth rates in January–March 2011 and January–June 2012, and the dominant organisms changed from *Cyanophyta* to *Chlorophyta* and *Bacillariophyta,* which was also observed in Fernández et al. [12]. However, the explanations for such a change were complicated, and the physical disturbances due to temperature and partial change of the raw water from the mainland China would probably be the major two reasons. Generally, *Chlorophyta* and *Bacillariophyta* favor in the shallow and enriched systems [55], and their dominant species, *Scenedesmus* spp., *Chlorella* spp., *Achnanthes* spp., and *Fragilaria* spp., varied greatly, which was consistent with the SIMI results indicating that the large variations in phytoplankton compositions happened in 2012.

Shannon index is not only the indicator of diversity, but also the indicator of pollution level of water bodies, with the H´ of 3.0–4.5 as slight, 2.0–3.0 as light, 1.0–2.0 as moderate, and 0.1–1 as heavy pollution [56]. Our results showed that 12 of 18 months in the study were classified as moderate pollution state, which was similar to that in Mumbai coast [56]. This can be explained by that Macau urban area is densely populated and susceptible to heavy anthropogenic stresses.

To further study on the spatial and temporal variations in water quality, PCA was served as a means to identify those parameters that had the greatest contribution to variation in the water quality of reservoirs and indicated possible sets of pollutant sources [34,57]. In our study, phytoplankton was found to be correlated with water temperature, pH, DO, NH_4-N, and Chl-a, and anti-correlated with conductivity, NO_3-N, NO_2-N, and SD, suggesting that high temperature, sufficient DO, appropriate alkalinity condition, and soluble nitrogen sources favor the phytoplankton abundance. High density of phytoplankton resulted in high concentrations of Chl-a, NH_4-N, and low SD.

Compared to PCA which is used to identify the principle parameters for explaining the variations of water qualities, CA is a method to classify the similar and dissimilar groups. It has been successfully applied in water quality assessment programs and designed the sampling strategy [19,34,36]. Different from SIMI index which is calculated based on the phytoplankton structure, CA was based on water quality including abiotic and biotic parameters. In the present temporal analysis of CA, two periods identified was consistent with the classification of cyanobacteria-blooming period and cyanobacteria-depression period. In the spatial analysis of CA, if the criterion of (Dlink /Dmax)*100 < 60 in previous studies [34,58] was applied, our sampling points did not show any difference, implying that only one sampling point may be enough for the rapid assessment of water quality in MSR. This is because in such a small reservoir with a short HRT, the hydrography is homogenous and the hydraulic exchange is high [59]. The stratification in MSR is thus absent or very weak, which is likely to have an impact on phytoplankton compositions and water quality from different depths that are sharing the same characteristics.

Besides, our DA results suggested that TP, NO_3-N, and Chl-a were the most significant variables among all the water parameters in temporal analysis, which was consistent with the PCA results that NO_3-N and Chl-a were the principle components for explaining the variations, and the temporal CA study showed that the two periods, algal blooms and non-algal blooms periods, can be divided. Actually, the TP and Chl-a were parameters used in calculating the TSI of the reservoir, while NO_3-N is the most important nutrient factor for phytoplankton growth. In spite of TP being a relatively independent factor in MSR, it was often selected as one of the most important parameters to monitor and reported as the environmental factor affecting the blue-green algae biomass [12,60].

5. Conclusions

In small-scale eutrophic lakes or reservoirs, physical disturbances caused by natural and anthropogenic activities have great impacts on the variations of phytoplankton structure and water quality. This study presented data on dynamic changes of the phytoplankton structure and water quality of a short HRT reservoir that is experiencing algal blooms in recent years. The reservoir showed a high TSI level of 65–82 and was determined to be in eutrophic–hypereutrophic status. Lowest diversity/evenness and highest dominance happened in June 2011, while highest diversity/evenness and lowest dominance occurred in May 2012, which was consistent with the number of countable species and their relative proportions. The SIMI index revealed that the variations of phytoplankton species were significant in 2012, while maintained relatively stable in 2011. Margalef's richness index revealed that the overall phytoplankton species richness level of MSR was not too high and within the range of 0.43–1.24.

PCA identified four factors, which were responsible for the data structure explaining 80.84% of the total variance of the complete data-set. CA generated two groups of spatial similarity from six sampling points and two groups of temporal similarity among 18 months, and DA provided an important data reduction, with only three parameters (TP, NO_3-N, and Chl-a) that could afford 100% right assignations in temporal analysis. However, there is no spatial variation found, mainly due to the small capacity of the reservoir. This study highlighted the usefulness of combination of various methods for the evaluation and interpretation of complex water quality data-sets and assessment of pollution level of small-scale eutrophic reservoirs. The results from this study confirmed the variations of phytoplankton structures and water quality in temporal distribution, as well as relative homogeneity in spatial distribution, which can help in developing a future plan to determine the optimal sampling locations and sampling frequency in the monitoring program of MSR.

Acknowledgments

We thank the technical staffs at Macao Water Supply Co. Ltd. for water sampling and measurement. The research project was supported by the Fundo para o Desenvolvimento das Ciências e da Tecnologia (FDCT) and the Research Committee of University of Macau, under grant number MYRG106(Y1-L3)-FST12-LIC.

References

[1] K. Sivonen, G. Jones, Cyanobacterial toxins, in: I. Chorus, J. Bartram (Eds.), Toxic Cyanobacteria in Water: A Guide to their Public Health Consequences, Monitoring, Management, E & FN Spoon, London, 1999, pp. 55–71.

[2] H.W. Paerl, J. Huisman, Climate change: A catalyst for global expansion of harmful cyanobacterial blooms, Environ. Microbiol. Rep. 1 (2009) 27–37.

[3] J.G. Tundisi, T. Matsumura-Tundisi, O. Rocha, Theoretical basis for reservoir management, in: J.G. Tundisi, M. Straškraba (Eds.), Theoretical Reservoir Ecology and Its Applications, Backhuys, Leiden, 1999, pp. 505–528.

[4] L. Naselli-Flores, Phytoplankton assemblages in twenty-one Sicilian reservoirs: Relationships between species composition and environmental factors, Hydrobiologia 424 (2000) 1–11.

[5] L. Naselli-Flores, R. Barone, Water-level fluctuations in Mediterranean reservoirs: Setting a dewatering threshold as a management tool to improve water quality, Hydrobiologia 548 (2005) 85–99.

[6] M.G. Nogueira, M. Ferrareze, M.L. Moreira, R.M. Gouvêa, Phytoplankton assemblages in a reservoir cascade of a large tropical–subtropical river (SE, Brazil), Braz. J. Biol. 70 (2010) 781–793.

[7] G.A.S.T. Lira, E.L. Araújo, M.C. Bittencourt-Oliveira, A.N. Moura, Phytoplankton abundance, dominance and coexistence in an eutrophic reservoir in the state of Pernambuco, Northeast Brazil, Ann. Acad. Bras. Ciênc. 83 (2011) 1313–1326.

[8] T. Baykal, I. Açikgöz, A.U. Udoh, K. Yildiz, Seasonal variations in phytoplankton composition and biomass in a small lowland river-lake system (Melen River, Turkey), Turk. J. Biol. 35 (2011) 485–501.

[9] K.W. Pontasch, E.P. Smith, J. Cairns, Diversity indices, community comparison indices and canonical discriminant analysis: Interpreting the results of multispecies toxicity tests, Water Res. 23 (1989) 1229–1238.

[10] G.A.S.T. Lira, M.C. Bittencourt-Oliveira, A.N. Moura, Structure and dynamics of phytoplankton community in the Botafogo reservoir-Pernambuco-Brazil, Braz. Arch. Biol. Technol. 52 (2009) 493–501.

[11] D.S. Lymperopoulou, K.A. Kormas, M. Moustaka-Gouni, A.D. Karagouni, Diversity of cyanobacterial phylotypes in a Mediterranean drinking water reservoir (Marathonas, Greece), Environ. Monit. Assess. 173 (2011) 155–165.

[12] C. Fernández, E.R. Parodi, E.J. Cáceres, Phytoplankton structure and diversity in the eutrophic-hypereutrophic reservoir Paso de las Piedras, Argentina, Limnology 13 (2012) 13–25.

[13] B. Helena, R. Pardo, M. Vega, E. Barrado, J.M. Fernández, L. Fernández, Temporal evolution of groundwater composition in an alluvial aquifer (Pisuerga river, Spain) by principal component analysis, Water Res. 34 (2000) 807–816.

[14] J. Lee, J. Cheon, K. Lee, S. Lee, M. Lee, Statistical evaluation of geochemical parameter distribution in a ground water system contaminated with petroleum hydrocarbons, J. Environ. Qual. 30 (2001) 1548–1563.

[15] S. Shrestha, F. Kazama, Assessment of surface water quality using multivariate statistical techniques: A case study of the Fuji river basin, Japan, Environ. Model. Softw. 22 (2007) 464–475.

[16] H. Çamdevýren, N. Demýr, A. Kanik, S. Keskýn, Use of principal component scores in multiple linear regression models for prediction of Chlorophyll-a in reservoirs, Ecol. Model. 181 (2005) 581–589.

[17] S.H. Te, K.Y. Gin, The dynamics of cyanobacteria and microcystin production in a tropical reservoir of Singapore, Harmful Algae 10 (2011) 319–329.

[18] M. Vega, R. Pardo, E. Barrado, L. Debán, Assessment of seasonal and polluting effects on the quality of river water by exploratory data analysis, Water Res. 32 (1998) 3581–3592.

[19] M. Varol, B. Gökot, A. Bekleyen, B. Şen, Spatial and temporal variations in surface water quality of the dam reservoirs in the Tigris River basin, Turkey, Catena 92 (2012) 11–21.

[20] W. Zhang, I. Lou, Y. Kong, W.K. Ung, K.M. Mok, Eutrophication analyses and principle component regression for two subtropical storage reservoirs in Macau, Desalin. Water Treat. 51 (2013) 7331–7340.

[21] APHA, Standard Methods for the Examination of Water and Wastewater, 21st ed., AWWA and WEF, American Public Health Association, Washington, DC, 1999.

[22] G.M. Smith, The Freshwater Algae of the United States, second ed., McGraw-Hill, New York, NY, 1950.

[23] B.J. McAlice, Phytoplankton sampling with the Sedgwick-Rafter cell, Limnol. Oceanogr. 16 (1971) 19–28.

[24] H. Utermöhl, Zur vervollkommung der quantitativen phytolankton-methodik (Perfectible quantitative phytoplankton methodology), Mitteilungen Internationale Vereinigung Theorie Angewandte Limnologie 9 (1958) 1–39.

[25] R.E. Carlson, A trophic state index for lakes, Limnol. Oceanogr. 22 (1977) 361–369.

[26] C.E. Shannon, A mathematical theory of communication, Bell Syst. Tech. J. 27(4) (1948) 623–656.

[27] E.H. Simpson, Measurement of diversity, Nature 163 (1949) 688.

[28] E.C. Pielou, Species-diversity and pattern-diversity in the study of ecological succession, J. Theor. Biol. 10 (1966) 370–383.

[29] R. Margalef, Information theory in ecology, Int. J. Gen. Syst. 3 (1958) 36–71.

[30] S.A. Khan, P. Murugesan, P.S. Lyla, S. Jaganathan, A new indicator macroinvertebrate of pollution and utility of graphical tools and diversity indices in pollution monitoring studies, Curr. Sci. 87 (2004) 1508–1510.

[31] J.M. Stander, Diversity and Similarity of Benthic Fauna off the Coast of Oregon, MS thesis, Oregon State University, Corvallis, 1970.

[32] J.L. Rohr, Changes in Diatom Community Structure Due to Environmental Stress, Dissertation, Bowling Green State University, Bowling Green, OH, 1977.

[33] C. Liu, K. Lin, Y. Kuo, Application of factor analysis in the assessment of groundwater quality in a blackfoot disease area in Taiwan, Sci. Total Environ. 313 (2003) 77–89.

[34] K.P. Singh, A. Malik, D. Mohan, S. Sinha, Multivariate statistical techniques for the evaluation of spatial and temporal variations in water quality of Gomti River (India)—A case study, Water Res. 38 (2004) 3980–3992.

[35] M. Otto, Multivariate methods, in: R. Kellner, J.M. Mermet, M. Otto, H.M. Widmer (Eds.), Analytical Chemistry, Wiley-VCH, Weinheim, 1998.

[36] D.A. Wunderlin, M.P. Diaz, M.V. Ame, S.F. Pesce, A.C. Hued, M.A. Bistoni, Pattern recognition techniques for the evaluation of spatial and temporal variations in water quality. A case study: Suquia River basin (Cordoba-Argentina), Water Res. 35 (2001) 2881–2894.

[37] R.A. Johnson, D.W. Wichern, Applied Multivariate Statistical Analysis, seventh ed., Prentice-Hall International, Englewood Cliffs, NJ, 1992, p. 642.

[38] J. Pallant, I. Chorus, J. Bartram, Toxic Cyanobacteria in Water, SPSS Survival Manual, third ed., Open University Press, Berkshire, 2007.

[39] M. Katsiapi, M. Moustaka-Gouni, E. Michaloudi, K.A. Kormas, Phytoplankton and water quality in a Mediterranean drinking-water reservoir (Marathonas Reservoir, Greece), Environ. Monit. Assess. 181 (2011) 563–575.

[40] C.C. Figueredo, A. Giani, Seasonal variation in the diversity and species richness of phytoplankton in a tropical eutrophic reservoir, Hydrobiologia 445 (2001) 165–174.

[41] R. Holzmann, Seasonal fluctuations in the diversity and compositional stability of phytoplankton communities in small lakes in upper Bavaria, Hydrobiologia 249 (1993) 101–109.

[42] C.S. Reynolds, Scales of disturbance and their role in plankton ecology, Hydrobiologia 249 (1993) 157–171.

[43] M. Moustaka-Gouni, Phytoplankton succession and diversity in a warm monomictic, relatively shallow lake: Lake Volvi, Macedonia, Greece, Hydrobiologia 249 (1993) 33–42.

[44] A.K.M. Rahman, D. Bakri, P. Ford, T. Church, Limnological characteristics, eutrophication and cyanobacterial blooms in an inland reservoir, Australia, Lakes Reserv. Res. Manage. 10 (2005) 211–220.

[45] K.E. Havens, Cyanobacteria blooms: Effects on aquatic ecosystems, in: H.K. Hudnell (Ed.), Cyanobacterial Harmful Algal Blooms: State of the Science and Research Needs, Springer Science, New York, NY, 2008, pp. 733–747.

[46] H.W. Paerl, Growth and reproductive strategies of freshwater blue-green algae (Cyanobacteria), in: C.D. Sandgren (Ed.), Growth and Reproductive Strategies of Freshwater Phytoplankton, Cambridge University Press, New York, NY, 1988, pp. 261–315.

[47] K.E. Havens, E.J. Phlips, M.F. Cichra, B. Li, Light availability as a possible regulator of cyanobacteria species composition in a shallow subtropical lake, Freshwater Biol. 39 (1998) 547–556.

[48] M.L. Saker, A.D. Thomas, J.H. Norton, Cattle mortality attributed to the toxic cyanobacterium *Cylindrospermopsis raciborskii* in an outback region of North Queensland, Environ. Toxicol. 14 (1999) 179–182.

[49] L.D. Fabbro, L.J. Duivenvoorden, Profile of a bloom of the cyanobacterium *Cylindrospermopsis raciborskii* (Woloszynska) Seenaya and Subba Raju in the Fitzroy River in tropical Central Queensland, Mar. Freshwater Res. 47 (1996) 685–694.

[50] M. Bouvy, R. Molica, S. De Oliveira, M. Marinho, B. Beker, Dynamics of a toxic cyanobacterial bloom (*Cylindrospermopsis raciborskii*) in a shallow reservoir in the semi-arid region of northeast Brazil, Aquat. Microb. Ecol. 20 (1999) 285–297.

[51] J. Komarkova, R. Laudares-Silva, P.A.C. Senna, Extreme morphology of *Cylindrospermopsis raciborskii* (Nostocales, Cyanobacteria) in the Lagoa do Peri, a freshwater coastal lagoon, Santa Catarina, Brazil, Algol. Stud. 94 (1999) 207–222.

[52] C.W.C. Branco, P.A.C. Senna, Factors influencing the development of *Cylindrospermopsis raciborskii* and Microcystis aeruginosa in the Paranoa Reservoir, Brasilia, Brazil, Algol. Stud. 75 (1994) 85–96.

[53] M. Presing, S. Herodek, L. Vörös, I. Kobor, Nitrogen fixation, ammonium and nitrate uptake during a bloom of *Cylindrospermopsis raciborskii* in Lake Balaton, Archiv Für Hydrobiologie 136 (1996) 553–562.

[54] J.S. Metcalf, A. Barakate, G.A. Codd, Inhibition of plant protein synthesis by the cyanobacterial hepatotoxin, cylindrospermopsin, FEMS Microbiol. Lett. 235 (2004) 125–129.

[55] C.S. Reynolds, V. Huszar, C. Kruk, L. Naselli-flores, S. Melo, Towards a functional classification of the freshwater phytoplankton, J. Plankton Res. 24 (2002) 417–428.

[56] S.N. Datta, S.K. Chakraborty, A.K. Jaiswar, G. Ziauddin, A comparative study on intertidal faunal biodiversity of selected beaches of Mumbai coast, J. Environ. Biol. 31 (2010) 981–986.

[57] K.P. Singh, A. Malik, S. Sinha, Water quality assessment and apportionment of pollution sources of Gomti river (India) using multivariate statistical techniques—A case study, Anal. Chim. Acta 538 (2005) 355–374.

[58] X. Wang, Q. Cai, L. Ye, X. Qu, Evaluation of spatial and temporal variation in stream water quality by multivariate statistical techniques: A case study of the Xiangxi River basin, China, Quat. Int. 22 (2012) 137–144.

[59] C.S. Reynolds, Phytoplankton assemblages in reservoirs, in: J.G. Tundisi, M. Straskraba (Eds.), Theoretical Reservoir and its Application, Backhuys, Leiden, 1999, pp. 439–456.

[60] M.C. Calijuri, A.C.A. Santos, S. Jati, Temporal changes in the phytoplankton community structure in a tropical and eutrophic reservoir (Barra Bonita, S.P.-Brazil), J. Plankton Res. 24 (2002) 617–634.