Faculty Resource Science and Management

PHYTOPLANKTON COMPOSITION OF UNIMAS LAKE IN EAST CAMPUS, SARAWAK

FIZAWATI BINTI KAMIS

Bachelor of Science with Honours
(Aquatic Resource Science and Management)
2012
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This project is submitted in partial fulfillment of the requirements for the degree of Bachelor of Science with Honours (Aquatic Resource Science and Management)

Department of Aquatic Science
Faculty of Resource Science and Technology
UNIVERSITI MALAYSIA SARAWAK
2012
I am very grateful to Allah for giving me the ability to complete my final year project.

I would like to extend my sincerest gratitude to my supervisor, Assoc. Prof. Dr. Norhadi Ismail, for his guidance and overall supervision throughout the course of my study. I also thank Assoc. Prof. Dr. Ling Teck Yee who assisted me on how to use the Statistical Package for the Social Sciences (SPSS) for data analysis.

I also would like to express my gratitude to my friend Diva Devirda Musa and the laboratory assistants, especially Mr. Zaidi Bin Haji Ibrahim, Mr. Mohd. Norazlan bin Bujang Belly, Mr. Zulkifli bin Ahmad, Mr. Nazri bin Latip, Mr. Richard Toh, Mr. Haris Norman @ Mustafa Kamal bin Mohd. Faizal and Mdm. Lucy Daru for their helps and support during my field work.

Finally, special thanks to my parents, Mr. Kamis bin Rosidi and Mrs. Doret binti Udai for supporting me throughout all my studies at University Malaysia Sarawak. Thank you to any individual that involved direct and indirectly in the completion of this final year project report.
DECLARATION

I hereby declare that the work in this project is my own except for quotations and summaries which have been duly acknowledged. No portion of the work referred to in this dissertation has been submitted in support of an application for another degree qualification of this or any other university or institution of higher learning.

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Fizawati Binti Kamis

Aquatic Resources Science and Management Programme

Faculty of Resource Science and Technology

University Malaysia Sarawak
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Phytoplankton Composition of UNIMAS Lake in East Campus, Sarawak

Fizawati Binti Kamis

Programme of Aquatic Resource Science and Management
Faculty of Resource Science and Technology
Universiti Malaysia Sarawak

ABSTRACT

A study on the phytoplankton composition in man-made lake was conducted at two lakes in East Campus of Universiti Malaysia Sarawak, Kota Samarahan. Sampling of phytoplankton from two of the lakes, were conducted from October 2011 until April 2012. Three stations were established at different parts of each lake, namely the inflow, middle and outflow of the lakes. Results showed that the phytoplankton biomass (chlorophyll a) in the lakes were negatively correlated with pH and dissolved oxygen (DO) but positively correlated with nitrate (NO₃⁻). A total of 5 algal classes were identified, which was dominated by members from the Chlorophyceae (13 genera), followed by Bacillariophyceae (10 genera), Cyanophyceae (6 genera), Euglenophyceae (3 genera) and Dinophyceae (1 genera). During the study period, water temperature recorded was in the range of 27.45°C – 30.87°C, with pH from 6.26 – 7.60. Dissolved oxygen varied from 1.07 mg/L – 6.14 mg/L with concentration of nitrate (NO₃⁻) and orthophosphate (PO₄³⁻) ranged from 0.01 mg/L – 0.09 mg/L and 0.05 mg/L – 0.23 mg/L respectively.

Key words: Phytoplankton, man-made lake, phytoplankton biomass, water parameters, phylum

ABSTRAK

Satu kajian mengenai komposisi fitoplankton dalam tasik buatan manusia telah dijalankan di dua tasik kampus timur Universiti Malaysia Sarawak, Kota Samarahan. Pensampelan fitoplankton dari dua tasik, telah dijalankan dari Oktober 2011 hingga April 2012. Tiga stesen telah ditetapkan di bahagian-bahagian yang berlainan di setiap tasik, iaitu saluran masuk, bahagian tengah dan saluran keluar tasik. Hasil kajian menunjukkan bahawa biojisim fitoplankton (klorofil a) dalam tasik ini mempunyai hubung kait yang negatif dengan pH dan oksigen terlarut (DO) tetapi mempunyai hubung kait positif dengan nitrat (NO₃⁻). Sebanyak 5 kelas alga yang telah dikenalpasti, iaitu didominasi oleh Chlorophyceae (13 genera), diikuti oleh Bacillariophyceae (10 genera), Cyanophyceae (6 genera), Euglenophyceae (3 genera) dan Dinophyceae (1 genera). Semasa tempoh kajian dijalankan, suhu air yang dicatatkan adalah dalam linkungan 27.45°C – 30.87°C, bacaan pH dari 6.26 – 7.60. Bacaan untuk oksigen terlarut berubah dari 1.07 mg/L – 6.14 mg/L dengan bacaan nitrat (NO₃⁻) dan orthophosphat (PO₄³⁻) adalah dari, 0.01 mg/L – 0.09 mg/L dan 0.05 mg/L – 0.23 mg/L untuk setiap bacaan.

Kata kunci: Fitoplankton, tasik buatan, biojisim fitoplankton, parameter air, phylum
1.0 Introduction

Lakes in Malaysia are the example of the tropical lake with difference of temperature between the surface and bottom is only 4.5°C (Szyper & Lin, 1990). According to Szyper and Lin (1990), the tropical lakes usually drop 0.3°C during the stratification. Stratification prevents dissolved oxygen and nutrient exchange within the upper and lower layers in the water column which restrict photosynthesis and production (Szyper & Lin, 1990).

Lakes zonation consist of three major zones which are euphotic zone, dysphotic zone and aphotic or profundal zone. The euphotic zone refers to the total illuminated stratum of the water and has 1% of light intensity of the surface which enough light for photosynthesis. The euphotic zone then subdivides horizontally into two subzones; littoral subzones and limnetic subzone. The littoral subzone is at the lake margin and support greater biodiversity of organisms. While, the limnetic subzone is the open water that away from the shore which only contain phytoplankton (Agrawal, 1999). The second zone is dysphotic zone where the small amount of light can reach this zone and has lower light intensity, below 1% of the surface. At this zone, the rate of respiration is higher than the rate of photosynthesis. Aphotic or profundal zone is the zone where the light penetration is zero due to it depth. This zone also has low amount of dissolved oxygen because active of decomposition by the organic detritus which release inorganic nutrients for the uptake by primary producers (Agrawal, 1999).

The most abundant phytoplankton in freshwater lake are the member of Chlorophyceae and Cyanophyceae. While the members of Bacillariophyceae, Dinophyceae, Cryptophyceae, Chrysophyceae and Euglenophyceae are more abundant in small lakes or ponds (Agrawal, 1999). Chlorophyceae are the green algae that mostly
found in all freshwater lakes which can be categorized as unicellular, colonial and filamentous. They also exist as motile and nonmotile organisms. The filamentous green alga is not considered as true planktonic algae but these filamentous algae resistant to grazing due to it larger size (Agrawal, 1999).

A few study of phytoplankton in lakes were conducted especially in Sarawak. The information available is limited especially on phytoplankton assemblages and their correlation with environment parameters (e.g., temperature, pH, DO, conductivity nitrate, phosphate and chlorophyll \(a\)) in tropical countries. In Malaysia, the studies are mostly on phytoplankton composition and productivity, such as Paya Bungor, Pahang (Fatimah et al., 1984), Tasik Aman, Tasik Kundang and Tasik Rawang, Kuala Lumpur (Sulaiman et al., 1991). According to the study done by Elora (2005), Unimas lakes are the example of the lake that undergoes eutrophication due to the input from the surrounding areas.

The main purpose of this study is to determine the composition and monthly variations of phytoplankton of UNIMAS lakes in east campus. The study is also intended to update and compare the information of phytoplankton composition from the previous study and also to relate the phytoplankton biomass (chlorophyll \(a\)) with the water parameters, namely temperature, pH, DO, nitrate and phosphate.
2.0 LITERATURE REVIEW

2.1 Morphology of Phytoplankton

According to Sieburth et al. (1978), phytoplankton are identified as planktonic photoautotrophs and major producer of the pelagic zone and there are three types of phytoplankton (based on their location); limnoplankton (plankton of lakes), helioplankton (plankton of ponds) and potamoplankton (plankton of the rivers).

Phytoplankton can be classified into two major groups which are nonmotile and motile plankton. The nonmotile plankton commonly consists of fast-growing diatoms, which also can be found into two groups; pennate diatoms, where their structure can be found in long and flat shape and centric diatoms, where their shaped can be found in pillboxes and may have spines through from their cell walls. The motile plankton such as flagellated and dinoflagellated can migrate vertically in the water body for the response to light (Langlois and Smith, 2001).

Phytoplankton can be found in different sizes and shapes. Phytoplankton commonly can be classified into four sizes which are macroplankton (more than 200 μm in diameter), microplankton (20-200 μm in diameter), nanoplankton (2-20 μm in diameter) and picoplankton (0.2-2 μm in diameter) (Sze, 1993).

Algae were coloured differently from green to golden brown algae which contain chlorophyll a and c through the chromatography. Each group of phytoplankton exhibits characteristic colours, depending on its relative abundance of the major groups of photosynthetic pigments: green (chlorophylls), yellow (carotenes), or pink or blue (phycobilins). The relative abundance of phytoplankton groups varies seasonally and geographically. Therefore, small amount of the phytoplankton groups are seldom found in the same plankton sample.
Cyanobacteria can be found in various shape and size with few cellular features visible with light microscopes due to its individual cells which are small (Fogg et al., 1973). They may occur singly or form simple linear chains of cells. They contain chlorophyll $a$, carotenoids, and phycobilin pigments. Cynobacteria consist of varying type of colours from olive-green, grey-green, yellow-brown, purplish and red (Prescott, 1968). Despite the protection provided by thick cell envelopes and polar plugs, these morphological adaptations are not absolutely effective in shielding oxygen. Several members have capability to store the phosphorus as polymeric polyphosphate bodies to maintain and survive during external phosphorus deficiency. Some of them exhibit of nitrogen-deficient water because of their ability to fix the atmospheric nitrogen into ammonium (Wyatt & Silvey, 1969).

Diatoms exhibit fine lines, or *striae*, on the frustules surface. These striae are actually rows of very small pores. Exchange across the cell wall occurs through these pores. Most of diatoms contain chlorophyll pigments a and c and a wide variety of carotenoids (Robert, 1989). As a consequence, diatoms often appear brown. Both types of diatoms, pennates and centric have an external cell wall, and frustules, composed of silicon dioxide (SiO2).

According to Armstrong and Brasier (2005), dinoflagellates distinguished from their special form of eukaryote nucleus or known as dinokaryon. It has a cellulose cell wall, integrated by many pores. This groove separates the dinophyte's cellulose cell wall into two portions, the *epicone* and *hypocone*. Spines, wings and horns may decorate the cell wall. Most forms have an equatorial groove that contains a ribbon flagellum. Another groove perpendicular to the equatorial groove contains a longitudinal flagellum. The long longitudinal flagellum gives the cells mobility (Armstrong & Brasier, 2005).
Euglenophyta is a free-living microalga contains chlorophyll $a$ and $b$ which is a grass-green motile unicells (monads) for undergoes photosynthesis (Sze, 1993). It commonly found at freshwater environment which exhibit heterotrophy and absorb extracellular organic material (Sze, 1993). Euglenophyta does not require starch to reserve but it need paramylon to reserve food. For morphological of euglenophyta is not derived from endoplasmic reticulum which then represent the plasmalemman for original green symbiont (Sze, 1993).

Previous study on phytoplankton composition and productivity of a shallow tropical lake Paya Bungor, Pahang, revealed that most of the phytoplankton there were genera belong to *Merismopedia*, *Microcystis*, *Lyngbya*, *Oscillatoria*, *Spirulina*, *Anabaena* and *Aphanizomenon* (Fatimah et al., 1984). Sulaiman et al.(1991), reported 38 species of phytoplankton found in Tasik Aman, 36 species in Tasik Rawang and 10 species in Tasik Kundang. In Tasik Chini, there were 135 species from 81 genera of phytoplanklon which were dominated by Chlorophyta. The genera that have been recorded were *Staurastrum*, *Cosmarium* and *Ankistrodesmus* (Sulaiman et al.,1991).

### 2.2 Reproduction of Phytoplankton

Phytoplankton undergoes reproduction for the perpetuation of the species and to increase the number of individuals of a species. There are two types of reproduction found in phytoplankton, which are sexual and asexual reproduction (Sze, 1993).

Asexual reproduction is the separation from the parent specialized cell or group of cells by creating a genetic carbon copy of itself to form new individual with same characteristics with the parent cells. Asexual reproduction is uniparental and undergoes variety of formation for new offspring such as, spore formation, fission and fragmentation (Sze, 1993).
The spore formation consists of zoospores, aplanospores, hypnospores, akinetes and tetraspores. Spores commonly one-celled structures and formed mostly by the algae. They may produce sporangia where each sporangium produce large or small spore which are may be motile (zoospores) or non motile (aplanospores) (Sze, 1993).

Sexual reproduction involve of two reproductive cells, male and female gametes either from the same plant (monoecious) or from different plants (dioecious). The fertilization on two gametes will produce a zygote which is always haploid. There are two type of sexual reproduction, isogamy and heterogamy; consist of anisogamy and oogamy (Hickel, 1988).

Some species of diatoms and dinoflagellates produce a resting stage (Noraini, 2008). Resting spore in diatom and cysts in dinoflagellates are allows their species to survive in unfavorable conditions. Some of the dinoflagellates have motile phase either to become cysts or normal vegetative cell which depends on prevailing conditions (Langois and Smith, 2001).

2.3 Ecological Importance of Phytoplankton

Phytoplankton is the ‘plant’ in the water body due to it position in aquatic food web, where they are the primary producers, providing food energy for animal-like zooplankton to multi-ton whales (Kideys et al., 2005). Phytoplankton also acts as an index of the tropic status in the water body where the developed amounts of phytoplankton become indicators for aquatic enrichment (Wetzel, 1975).

Phytoplankton also be the harbinger of death or disease where they are the indicator of the area with have greater amount of nutrient input by causing the powerful biotoxins to produce ‘red tides’ or harmful algal blooms. These toxic blooms can kill marine life and people who eat the contaminated seafood (Pan et al., 1999).
Massive blooms of freshwater algae often cause die-off of fish and other organisms when the algal populations suddenly collapse. The death and rapid decomposition of the algae quickly lead to anoxia and asphyxiation of fish and other aquatic animals (CCME, 1992). Responsible

The cumulative irradiance experienced by phytoplankton during transit within the pool was found to be a good predictor of autotrophic potential and for interpreting complex interactions arising from seasonal hydrologic cycles and the influence of water regulation structures (Sellers & Bukaveckas, 2003).

Phytoplankton is responsible for gas exchange of carbon dioxide, between the lakes or any water body with the atmosphere (Paterson et al., 2008). Carbon dioxide is consumed by the phytoplankton during the photosynthesis process and exchange the oxygen for the organism uptake. The carbon dioxide that release during the phytoplankton decompose or respiration by the organisms will return to the near-surface water and some of carbon dioxide is carried to the deep area when the phytoplankton die (Paterson et al., 2008).
3.0 Materials and Methods

3.1 Study Area

The study was conducted at the man made lake of UNIMAS at east campus. These man-made lakes were shallow with depth around 0.6 to 0.8 meters. Two lakes were selected as study site and three stations were established in each lake (refer to the Figure 1 to Figure 3).

In Lake 1, station 1 was established at the inflow from the cafeteria, station 2 was established at the outflow and station 3 was established in the middle part of the same lake. While, in Lake 2, station 4 was established at the inflow from the surrounding area, station 5 was established at the outflow and station 6 was established in the middle part of the same lake. The coordinate for each station was determined by a Global Positioning System (GPS) as shown in Table 1.

<table>
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<tr>
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<th>Global Positioning System (GPS) reading</th>
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</thead>
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<tr>
<td>Station 1</td>
<td>N 01°27.571’, E 110°27.148’</td>
</tr>
<tr>
<td>Station 2</td>
<td>N 01°27.560’, E 110°27.176’</td>
</tr>
<tr>
<td>Station 3</td>
<td>N 01°27.548’, E 110°27.207’</td>
</tr>
<tr>
<td>Station 4</td>
<td>N 01°27.657’, E 110°27.185’</td>
</tr>
<tr>
<td>Station 5</td>
<td>N 01°27.640’, E 110°27.162’</td>
</tr>
<tr>
<td>Station 6</td>
<td>N 01°27.663’, E 110°27.211’</td>
</tr>
</tbody>
</table>
Figure 1: Map of sampling site in UNIMAS lakes at east campus
3.2 Sample Collections

3.2.1 Water Collections

The sampling was done twice per month in Lake 1 and 2, starting from 12 October 2011 and finished on 25 April 2012. Each sampling was done in the morning, around 9.00 am to 1.00 pm using a boat without engine.

Phytoplankton samples were collected using a plankton net (20 µm mesh size) at the sub surface of the lakes. The plankton net was towed horizontally. Samples were poured into Whirl-Pak, preserved with Lugol’s solution and labeled. The quantity of Lugol’s solution used was 3mL for 1 L of samples as suggested by Elora (2005). All samples were brought back to the Aquatic Botany laboratory and placed into a 1 L beaker for settlement of the phytoplankton.

Triplicate water samples were taken using a Van dorn water sampler and were kept in 1 L of acid washed bottle samples. The water samples were placed into cooler box and brought back to the laboratory for chlorophyll a and nutrients analysis. They were kept in refrigerator with the temperature (4°C) if the samples were to be analyzed within 24 to 48 hour later or placed the in the freezer (-20°C) for later analysis.

3.2.2 Physico-chemical Parameters

The physico-chemical parameters (DO, turbidity, depth, pH and temperature) reading were measured in-situ by using Eutech Multiparameter Series 600, DO meter, Secchi disk, depth finder, pH and temperature meter. Triplicate readings of each parameter at each sampling sites were collected.
3.3 Laboratory Studies

3.3.1 Phytoplankton Identification

The samples of each station were viewed under Nikon Eclipse E100, compound microscope (10 x 100 magnifications). Cell identification was done to the lowest possible taxon based on available reference sources such as Botes (2001), Mizuno (1978), Stafford (1999) and other relevant resources.

3.3.2 Permanent slides preparation for diatom microscopy

According to the method that suggested by Battarbee (1986), about 1 ml of the sample was placed into the centrifuge tube and double amount of the hydrogen peroxide was mixed into the same tube for cleaning the diatom. After 2 minutes reaction, distilled water was added into the tube and the diatom sample was centrifuged. After the first centrifugation, the supernatant was discharged and new distilled water was added into the tube. This step was repeated until the pH is neutral.

Clean sample was drop with the bulb onto the cover slip and placed onto the hotplate for drying process. The mounting media, Naphrax® solution was dropped on the dried sample and heated on the hotplate. After the Naphrax® toluence solutions was placed onto the cover slip, a piece of glass slide was put onto the slip, press gently to avoid bubble trap in the slides samples and than observed under Nikon Eclipse E100 (10 x 100 magnifications).
3.3.3 Chlorophyll a Analysis

Based on the method that suggested by APHA (1998), one liter of water samples were filtered using a fibre-glass filter paper Chlorophyll was extracted by adding 5 to 6 ml of 90% aqueous acetone and ground with mortar and grinder. The sample was transferred into test tube which was wrapped with aluminium foil and stored in 4°C in a refrigerator for 4 to 18 hours before centrifugation. The absorbance was measured using Hach DR 2800 and the chlorophyll concentrations calculated according to the equations of Jeffrey and Humphreys (1975):

\[ E_{664}\_C = E_{664} - E_{750} \]
\[ E_{647}\_C = E_{647} - E_{750} \]
\[ E_{630}\_C = E_{630} - E_{750} \]

Where \( E_{750} \) = absorption at 750 nm wavelength
\( E_{664} \) = absorption at 664 nm wavelength
\( E_{647} \) = absorption at 647 nm wavelength
\( E_{630} \) = absorption at 630 nm wavelength
\( E_{x}\_C \) = corrected values of absorption for turbidity blanks

Chlorophyll concentration (µg / mL):

\[ \text{Chlorophyll } a = 11.85 \times E_{664}\_C - 1.54 \times E_{647}\_C - 0.08 \times E_{630}\_C \]

Conversion of Chlorophyll a concentration to mg/L in water samples

\[ \text{Chl } a (\mu g/L) = C_x \times v / VL \]

Where \( C_x = Ca \) is Chlorophyll concentration in (µg / mL)
\( v \) = volume of acetone in mL
\( V \) = volume of water samples in L

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3.3.4 Nitrate-Nitrogen Analysis

Nitrate was determined using a standard method 8192, *Cadmium Reduction Method* (Hach, 2000). A 15 ml of filtered water sample was added with Nitra ver 6 and continuously shaken for 3 minutes. The sample left for two minutes to allow cadmium reaction. After that, 10 ml of water sample was placed into the sample cell. The Nitra ver 3 was added to the water sample and inverted until dissolved before the substance was left for 15 minutes reaction. After the reaction completed, the nitrate concentration was measured using Hach DR 2800.

3.3.5 Orthophosphate Analysis

Orthophosphate was determined using a standard method 8048, *Powder Pillow Method* (Hach, 2000). A 10 ml of filtered water sample was added with Phos ver 3 of Phosphate Powder Pillow and then shaken to mix the substance for 30 second. After the mixing was complete, the orthophosphate concentration was measured using Hach DR 2800.
3.4 Statistical Analysis

The weekly mean of each physico-chemical parameters and nutrients from each station were calculated. One-way ANOVA followed by Tukey’s test were use to compare these means for its significant differences. Pearson’s Correlation was used to evaluate the relationships between phytoplankton biomass (Chl a) and the environmental parameters (α=0.05). The analysis was carried out using Statistical Package for the Social Sciences (SPSS) version 20.
4.0 Results

4.1 Phytoplankton composition

A total of 33 genera of phytoplankton were identified during this study (Table 2 until Table 8). Both of the lakes were dominated by Chlorophyceae with 13 genera. Other classes of phytoplankton were Bacillariophyceae with 10 genera found, 6 genera from Cyanophyceae, 3 genera from Euglenophyceae and 1 genus from Dinophyceae. The most common genera found at every station were *Scenedesmus, Navicula* and *Euglena* (Figure 2 and 3). Table 2 shows that the comparison between the previous study done by Elora (2005) with the current study (2012). The number of genera for each classes increase except for the member from Dinophyceae where only one genus was found.

<table>
<thead>
<tr>
<th>Class</th>
<th>Previous Study (Elora, 2005)</th>
<th>Current study (2012)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorophyceae</td>
<td>6 genera</td>
<td>13 genera</td>
</tr>
<tr>
<td>Bacillariophyceae</td>
<td>2 genera</td>
<td>10 genera</td>
</tr>
<tr>
<td>Cyanophyceae</td>
<td>3 genera</td>
<td>6 genera</td>
</tr>
<tr>
<td>Euglenophyceae</td>
<td>3 genera</td>
<td>3 genera</td>
</tr>
<tr>
<td>Dinophyceae</td>
<td>2 genera</td>
<td>1 genus</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>16 genera</strong></td>
<td><strong>33 genera</strong></td>
</tr>
</tbody>
</table>

Most of microalgae found in the UNIMAS lakes were from Class Chlorophyceae. More genera from this class were found in Lake 1 throughout the studied months. *Scenedesmus* was very common in both lakes and occurred at all stations. Other genera such as *Chodatella* and *Ankistrodesmus* have less common in both lakes.