Seasonal and Spatial Variation of Ciliate Abundance in Lake Lanao along Wato-Balindong, Lanao Del Sur, Philippines

F Abamo\textsuperscript{1,2*}, H Cosain\textsuperscript{2}, J Abato\textsuperscript{1}, C Disoma\textsuperscript{2}

\textsuperscript{1} Mamitua Saber Research & Technology Center, Mindanao State University (MSU)-Main Campus, Marawi City 9700 Philippines
\textsuperscript{2} Biology Department, MSU-Main Campus, Marawi City 9700 Philippines

*Corresponding author: yadfem@yahoo.com

Abstract. Ciliated protozoans are one-celled eukaryotes characterized by hair-like structures called cilia. Ciliate abundance has been used as bioindicators of organic pollution in a freshwater system. This study was conducted to determine the spatio-temporal variation of ciliate abundance in Lake Lanao, Philippines, along Wato-Balindong’s littoral and pelagic zones during dry (May 2018) and wet (August 2018) seasons. It intended to assess the trophic state of the lake based on planktonic ciliate abundance. In each zone, three sites were sampled for ciliates using a conical plankton net. The ciliates were fixed with formalin-acidified logul’s iodine for microscopic observation and counting. Spatially, the data on ciliate abundance did not vary between littoral and pelagic zones. However, it did vary significantly between seasons; ciliates had higher abundance in the wet season than in the dry season. This variation could be attributed to a possible increase of nitrogen and phosphorus washed into the lake by rain. Bioindication using ciliate abundance revealed that Lake Lanao in Wato-Balindong is ultraoligotrophic, i.e., the lake is pristine, well-oxygenated and low in nutrients during sampling.

1. Introduction
Lake Lanao in the province of Lanao del Sur (LDS) is located in the southern part of the Philippines. It is considered as the largest freshwater lake in the Philippines, one of the world’s ancient lakes [1] and the major source of hydroelectric energy in Mindanao. Countless floral and faunal organism are thriving in the lake including the endangered endemic cyprinid fish [2]. It is the home of the Maranao, “people of the lake” whose culture is so intertwined with the lake such as food source, the water source for domestic and agricultural uses, livelihood, sports, transport as well as religious ablution activity. The lake is reportedly deteriorating due to the increased human population and activities not to mention the damage it sustained by the recent Marawi siege.

Protozoan ciliates are unicellular eukaryotes characterized by hairy organelles called cilia [3]. They comprise a significant component of the abundance of freshwater plankton communities such as lakes. Ciliate abundance may vary from 1-24 cells/ml in oligotrophic and mesotrophic lakes to over 1000 cells/ml in eutrophic lakes. Their abundance increases as the trophic state of freshwater changes from oligotrophic to mesotrophic or to the eutrophic conditions [4]. Moreover, in the freshwater community they serve as prey for zooplankton and fish larvae but as predators for bacteria and nanoplankton. They are abundant in organically polluted rivers and streams thus their presence and abundance are good indicators of organic pollution; ciliate communities were studied as an early warning indicator of changes in the environment [5,6]. This study, therefore, aimed to study the ciliate abundance in the Lake Lanao particularly focusing only in the lake waters bordered by the municipality of Wato-Balindong,
Lanao, del Sur (which is only part and partial of a big project). Specifically, it intended to compare the ciliate abundance between the littoral and pelagic zones and between the dry and wet seasons. It also intended to utilize the ciliate abundance as bioindicators of the trophic status of the lake and organic pollution. Data generated by this study could serve as a basis for the local government to formulate policies in conserving the lake’s biological resources.

2. Methods

2.1. Sampling location and schedule

The study was conducted in the western part of the Lake bordered by Wato-Balindong, LDS during dry (May 2018) and wet or rainy (August 2018) seasons (Figure 1). Most of the houses in this area are built along the shoreline of the lake where people have easy access for fishing, transportation and other domestic uses of the lake. The nearby land has a mixture of trees and agricultural farms planted with corn.

![Map of the Philippines and Lake Lanao](source: http://www.gpsvisualizer.com/map?output_google)

**Figure 1.** A. Map of the Philippines showing the Lake Lanao in the inset. B. Lake Lanao showing the sampling location (circle) in Wato-Balindong, Lanao del Sur.

2.2. Sampling of ciliates

Ciliates from the lake water samples were collected using a conical plankton net with a mesh size of 53 µm and a mouth diameter of 0.48895 m. Three sampling sites at 100-m intervals were established in each zone serving as replicates. For the littoral zone, approximately up to 100 m perpendicular from the lakeshore, the plankton net was towed obliquely for 10 m to get an integrated water sample representing the surface and middle parts of the water column. For the pelagic zones with a minimum depth of about 20 m, the plankton net was towed vertically at a distance of 10 m in each specific sampling site.

Only the planktonic ciliates were considered in this study. Water samples were concentrated to 50-100 mL by a modified sieve with a mesh size smaller than the plankton net and the ciliates were fixed by adding a formalin-acidified Lugol’s solution. For every 10 mL of concentrated sample, 1 mL of 5% formalin and a drop of acidified Lugol’s solution were added after transferring the water samples into the plastic sampling bottles. Ciliates were counted manually using the right-hand rule method for
microscopy. During microscopic counting, only 30-50 mL of water sample in each replicate was scored for ciliate abundance (cells/m³). Abundance was calculated according to Harris et al. [7]:

\[
\text{Abundance} \left(A = \frac{\text{indiv}}{m^2}\right) = \frac{n+k}{V} \quad \text{eq. 1}
\]

where:
- \( n \) = number of count for a particular ciliate species
- \( k \) = the part of sample counted (i.e. the proportion of the total volume to subsample volume)
- \( V \) = the volume of water filtered by the plankton net
- \( m \) = the mouth area of the plankton net x distance towed

2.3. Physical and chemical parameters
Physico-chemical parameters namely temperature, pH and Secchi depth were also measured during the sampling. The temperature was measured at the surface level (within a few centimeters from the surface) by a hand-held thermometer. A portable pH meter (Hanna Instrument, USA) was used for pH. The Secchi depth was measured using a Secchi disc. Trophic state index (TSI) based on Secchi disc transparency was also calculated following Carlson [8]:

\[
\text{TSI} (SD) = 10 \left(6 - \frac{\ln SD}{\ln 2}\right) \quad \text{eq. 2}
\]

where:
- SD=Secchi depth

2.4. Statistical Analysis
Statistical Package for Social Sciences (SPSS), Version 22 was used to analyze the data. All data in abundance, temperature, and pH were subjected to the Shapiro-Wilk test to determine if means were normally distributed. The Sample T-test for Equality of Means was then used should the data satisfy the assumption of a normal distribution, otherwise, Mann-Whitney U would be used.

3. Results and discussion
A total of 27 morphologically distinct ciliates were observed in all sampling sites. Figure 2 shows the total abundance of ciliates between littoral and pelagic zones and between dry and wet seasons. The spatial abundance of ciliates between littoral and pelagic zones in each season did not very much. Results of the t-test comparing the mean abundance of ciliates between zones confirmed that the variation did not significantly differ. However, comparing the abundance of ciliates between seasons (Figure 3) revealed a significant variation. The abundance was significantly higher in the wet season than in the dry season. This result actually conformed to the expectation since there could be an increase of nitrogen and phosphorus washed into the lake by rain during wet season [9].

Bioindication involving ciliate abundance to determine the trophic state of the lake is made possible by comparing the data of the present study with the work of Beaver and Crisman [4] (Table 1). As pointed out in the table, the spatio-seasonal abundance of ciliate in Lake Lanao along Wato Balingdong is ultraoligothphic, i.e, the lake is pristine, well-oxygenated and low in nutrients during the sampling seasons.
**Figure 2.** Ciliates abundance (cells/m³) between littoral and pelagic zones and between dry and wet seasons. Results of the t-test comparing the spatial variation of mean abundance are also shown.

**Table 1.** Summary of data on annual abundances of planktonic ciliates in lakes [4].

| Trophic state      | Range of observed abundances (cell/ml) |
|--------------------|----------------------------------------|
| Ultraoligotrophic  | 2.4                                    |
| Oligotrophic       | 2.3-10.8                               |
| Mesotrophic        | 18.0-70.9                              |
| Eutrophic          | 55.5-145.1                             |
| Hypereutrophic     | 90.0-215.0                             |

**Figure 3.** The total ciliate abundance (cells/m³) between dry and rainy seasons. Results of the t-test comparing the seasonal variation of mean abundance is also shown.

**Table 2.** Mean data on the Secchi depths between sites and seasons and the corresponding calculated TSI of the Secchi depths.

| Seasons | Zones     | Secchi Depth (m) | TSI  |
|---------|-----------|------------------|------|
| Dry     | Littoral  | 4.25             | 39.12|
|         | Pelagic   | 5.125            | 36.42|
| Wet     | Littoral  | 2.25             | 48.30|
|         | Pelagic   | 4.5              | 38.30|

**Figure 4.** The mean temperature (left) and pH (right) data between seasons. Results of the t-tests comparing the mean variation between seasons are also shown.
Figure 4 illustrates the mean seasonal temperature and pH data of the sampling sites. Both factors were significantly higher in the wet season than in the dry season. A closer look at the temperature and pH data of the lake at Wato-Balindong supported the prevailing abundance of ciliates during seasonal sampling. The higher temperature of the lake during the wet season coincided with the higher abundance of ciliates since temperatures tend to increase as the metabolic reaction increases [4]. Moreover, the higher pH data also concurred with higher ciliate abundance during wet season. A similar result was obtained from a study on planktonic ciliates in subtropical lakes in which the ciliate abundance generally decreased with low pH. Kwiatkowski & Roof [10] also reported that primary productivity and abundance in lakes were reduced in low pH.

Table 2 shows the data of the Secchi depths in both zones and seasons. The values of the Secchi depths in this study were used to calculate the TSI of the lake following the work of Carlson and Simpson [11] which revolves around the idea that the water transparency decreases as the productivity increases thus the Secchi depth decreases. The calculated TSI ranged from 36.42 to 48.30 which is interpreted as an oligotrophic category. The calculated TSI corroborated the indication by ciliate abundance which positively demonstrated that Lake Lanao waters in the sampling sites of Wato-Balindong are clean.

4. Conclusion and Recommendation
A part of Lake Lanao bordered by the municipality of Wato-Balindong, LDS was studied for ciliate abundance between littoral and pelagic zones and between dry and wet seasons. Ciliate abundance did not vary spatially but varied significantly between seasons. It was higher in the wet than in the dry season. Based on ciliate abundance during sampling, Lake Lanao in Wato-Balindong is ultraoligotrophic, indicating that the water is clean, well-oxygenated and low in nutrients during the sampling. Local government must be informed of the lake’s condition from this study to formulate evidence-based plans and policies for Lake Lanao management and conservation.

Acknowledgement
This study is funded by the National Research Council of the Philippines (NRCP)-Grants-In-Aid.

References
[1] Duker L and Borre L 2001 Biodiversity conservation of the world's lakes: A preliminary framework for identifying priorities LakeNet Report Series Number 2 (Maryland (USA): LakeNet Secretariat Annapolis)
[2] Ismail G Sampson D and Noakes D 2014 Environ. Biol. Fishes 9 74
[3] Jahn T L, Bovee E C and Jahn F F 1979 How to know the protozoa 2nd ed (McGrawHill Science/Engineering/Math)
[4] Beaver J and Crisman T 1989 Microb. Ecol. 17 2
[5] Mohr J 1952 Sci Mon 74 1
[6] Mason C 1996 Biology of Freshwater Pollution 3rd ed (Singapore Publishers, Ltd.)
[7] Harris R et al. 2000 ICES zooplankton methodology manual (Acad Press)
[8] Carlson R 1977 Limnol. Oceanogr. 22 2
[9] Polameesanaporn Y 2008. Biodiversity of Protozoa in Freshwater Case Study in the Chao-Praya River, Thailand Proceedings of Taal 2007: The 12th World Lake Conference 484
[10] Kwiatkowski R and Roof J 2011 Can. J Bot 54 22
[11] Carlson R and Simpson J 1996 A Coordinator’s Guide to Volunteer Lake Monitoring Methods North American Lake Management Society pp 96