Phytoplankton biodiversity in the Atlangatepec Dam, a Ramsar Site of international importance

Biodiversidad de fitoplancton en la Presa de Atlangatepec, Sitio Ramsar de importancia internacional

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Abstract

The bodies of water provide ecosystem services such as: temperature regulation, hydric recharge, bird, terrestrial and aquatic species habitat, food, oxygen, landscape, tourism, among others. The objective of this work was to generate information on indicators of biological contamination in the Atlangatepec dam, which help to establish the Control and Management Plan (PCyM) for the Ramsar Site. The strategy consisted of determining physicochemical variables *in situ* and conducting a phytoplankton diversity study. The average temperature of the water body was 17.8 ° C, pH 8.23, dissolved oxygen 4.58 ppm and conductivity 215 µS.cm. These conditions favor the growth of cyanobacteria, which predominated in the body of water. The species *Pseudanabaena galeata* was the dominant species, followed by *Plankthorix agardhii*. 11 *Chlorophytas*, 15 *Cyanobacteria*, 1 *Charophyta* and 3 *Bacillariophytas* were identified. The cyanobacterial species identified have the potential to produce blooms and microtoxins, which negatively impact the food chains of the ecosystem. The impact of anthropogenic activities puts the dam's sustainable and sustained development at risk.

***Pseudanabaena galeata*, *Planktothrix agardhii*, cyanobacterial**

Resumen

Los cuerpos de agua aportan servicios ecosistemicos como son: regulación de temperatura, recarga hidríca, habitat de aves, especies terrestres y acuáticas, alimento, Oxígeno, paisaje, turismo entre otros. El objetivo del presente trabajo, fué generar información de indicadores de contaminación biologica en la presa de Atlangatepec, que ayuden a establecer del Plan de control y Manejo (PCyM) para el Sitio Ramsar. La estrategía consistió en determinar variables fisicoquímicas *in situ* y realizar un estudio de diversidad del fitoplancton. La temperatura promedio del cuerpo de agua fue de 17.8 °C, el pH 8.23, el Oxígeno disuelto 4.58 ppm y la conductividad de 215 µS.cm. Dichas condiciones favorecen el crecimiento de cianobacterias, las cuales predominaron en el cuerpo de agua. La especie *Pseudanabaena galeata* fue la especie dominante, seguida de *Plankthorix agardhii*. Se identificaron 11 *Chlorophytas*, 15 *Cyanobacterias*, 1 *Charophyta* y 3 *Bacillariophytas*. Las especies de cianobacterias identificadas, tienen el potencial de producir blooms y microtoxinas, las cuales impactan de forma negativa las cadenas tróficas del ecosistema. El impacto de las actividades antropogénicas pone en riesgo el desarrollo sustentable y sostenido de la Presa.

***Pseudanabaena galeata*, *Planktothrix agardhii*, cianobacteria**

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Introduction

The Atlangatepec dam is located 28 km from the city of Tlaxcala and 10 km from Taxco, in the State of Tlaxcala with coordinates of 19 ° 33'39 " N and 98 ° 10'49 " W, its area is of 1,200 hectares, it was declared a Ramsar site on February 2, 2009. The wetland receives tributaries from streams and the Zahuapan River, as well as discharge of sewage from the municipality of Tlaxco, the change of land use for agricultural and livestock activities, deforestation has caused high levels of siltation and etrophication in the body of water.

The location of the wetland gives it great importance, since it is a habitat for resident and migratory species, records of 125 species of birds associated with the wetland are kept and an average of 27,966 birds per season is estimated. The site is home to the Mexican Duck, Anas diazi, which is in category (A) threatened and endemic for Mexico, “protected” (Pr) the Gray Heron (*Ardea herodias*), Lesser Zambuillidor (*Tachybaptus dominicus*), (Pr), Northern Tular Heron (*Botaurus lentiginosus*) (A), Greyish Barred Ralo (*Rallus elegans*), (Pr) Reddish Barred Ralo (*Rallus limicola*) (Pr) and Axolotl (*Ambystoma tigrinum*) (A). The wetland is valuable in the production of fish, grains, hunting and tourism (I).

Based on the content of chlorophyll and dissolved solids, the water body was classified as eutrophic. Water bodies with eutrophic environments favor the proliferation of phytoplankton, with cyanobacteria being the dominant organisms. Cyanobacteria are unicellular, colonial prokaryotic organisms of great morphological diversity, growing in both terrestrial and freshwater or marine environments. (II)

Cyanobacteria predominate in temperature ranges close to 20 ° C, and have efficient mechanisms for capturing light. The presence of gas vacuoles allows cyanobacteria to move in the water column to access light and nutrients (III).

The warm temperatures and the availability of nutrients favor the establishment of blooms and the production of toxins. As part of phytoplankton, they are also the primary producers of aquatic systems.

They have the ability to fix nitrogen and store phosphorus, they can grow with low light intensity and produce substances that inhibit the growth of other organisms (VII).

Toxicological studies carried out with strains of *Pseudanabaena galeata* show that it has the ability to produce toxins in natural environments. Identified as: microtoxins, anatoxin-a and saxitoxin. Extracts prepared from said cyanobacteria with a single oral dose of 1,000 mg.kg-1 of body weight, caused the death of mice in 12 days.

Extracts prepared from laboratory-grown P galeata CCIBt3082 strains are classified in the category of compounds with relatively low toxicity, but with effects on the health of humans exposed to them for long periods of time. The effects it produces are microscopic lesions in the intestine, dilation of lymphatic and intestinal vessels and liver congestion. (IV)

The oscillatorial order is made up of filamentous bacteria, within them the Planktothrix genus is grouped, with the capacity to produce: anatoxins, microcystins and saxitoxins. Its production has been confirmed worldwide (VI).

It is worth mentioning that according to Pineda 2011, P. agardhii does not have the ability to produce cream in bodies of water since its growth is dispersed (II). However, it has been reported as a dominant species in European lakes with a higher toxic capacity compared to other cyanobacteria (VII).

The present work aims to study the temporal diversity of phytopankton in the Atlangatepec Dam and its trophic status, in order to generate scientific knowledge that helps define the complementary measures for the Development of the Conservation and Management Plan of the dam of Atlangatepec.

Hypothesis the eutrophication conditions reported for the Atlangatepec Dam favors the establishment of cyanobacteria and negatively affects the diversity of phytoplankton and the structure of trophic networks.

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Methodology

The present study was carried out in the Atlangatepec Dam, located in Tlaxcala, Mexico on November 3 and 4, 2021.

The environmental monitoring consisted of the monitoring of physicochemical parameters: pH, temperature, conductivity, dissolved oxygen, with the use of a HANNA Instruments brand multiparametric probe, model HI9829.

In each monitoring station (See Table 1) samples were collected for biodiversity analysis by dragging with a 20 µm mesh (1 minute), the samples were kept in a sterile flask and formaldehyde was used to preserve them.

The classification was carried out based on the traditional morphological characters with the use of a compound microscope brand Leica and lugol. The study was carried out at the Microbiology Laboratory of the Polytechnic University of the Guadalajara Metropolitan Area (UPZMG). Located in Tlajomulco de Zuñiga Jalisco. In collaboration with the students of the UPZMG Biotechnology career.

Station	Location
1.- Zacacuexco	19°33´01" N 98°10´30.9" W
2.- The Shearing	19°33´00.09" N 98°10´30.9" W
3.- Airport	19°32´58.7" N 98°10´31.6" W
4.- San José Atlangatepec	19° 33´34.7" N 98° 11´56.8" W
5.- Santa Clara	19° 34´37.3" N 98° 11´04.6" W
6.- Embarcadero	19° 33´42.4" N 98° 12´04.2" W

Table 1 Coordinates of the monitoring stations at the Atlangatecpec dam, phytoplankton biodiversity analysis
Source: Own elaboration

Phylogenetic analysis. The search for gene and protein sequences was carried out based on NCBI data. BLAST (Basic Local Alignment Search Tool) was used using a cyanobacterial protein of 275 aa Oxidoreductase as zone. Sequence cleansing was performed visually, removing the sequences that did not contain conserved sites or the repeated ones. NCBI diagrams were used.

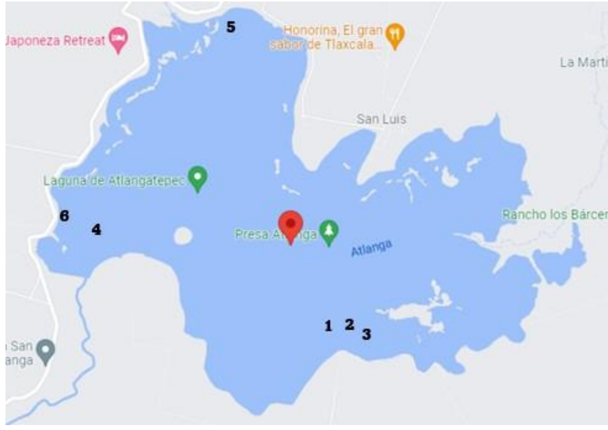

Figure 1 Monitoring points in the Atlangatepec Dam Tlaxcala Mexico, phytoplankton Biodiversity analysis
Source: Own elaboration

Results

Figure 2 shows the physical-chemical data collection and samples at the Santa Clara monitoring station.

Figure 2 Record of physicochemical variables and phytoplankton sampling at the Santa Clara station
Source: Own elaboration

Table 2 contains the results of the monitored physicochemical variables. The pH presented an average value of 8.23 with a range of 7.98 to 8.45. The average dissolved oxygen was 4.58 ppm. The conductivity range was from 215 to 245 µS.cm. The average temperature was 17.87 ° C.

The temperature varies according to the geographic location and climatic conditions, its impact determines the availability of dissolved oxygen in the body of water, influencing the diversity of microorganisms in the water column and seasonal succession of aquatic communities (VIII).

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The oxygen concentration is determined by the temperature, the atmospheric pressure, the aquatic organisms that, due to the photosynthesis process, release it in the euphotic zone and the supersaturation in the surfaces is associated with high concentrations of phytoplankton. However, in the aphotic zone, the decomposition of organic matter by microorganisms consumes oxygen and releases $C0_2$.

When the pH is neutral or slightly alkaline, in the range of 7.5 and 10 and the temperatures are warm, they favor the growth of cyanobacteria and the development of blooms since it allows them to assimilate the inorganic carbon in the form of bicarbonate (II).

Electrical conductivity is a numerical expression of the ability of water to conduct electric current and is related to the presence of ions in the water, in the study carried out the range was found from 215 to 245 µS.cm (VIII).

Variable	1	2	3	4	5	6
pH	8.1	8.2	8.1	8.4	7.9	8.0
OD ppm	4.6	4.9	5.3	4.6	3.3	4.6
Conductivity (µS.cm)	244	215	222	224	245	238
Temperature (°C)	17.5	17.8	17.5	19	16.4	18.9
Atmospheric pressure (psi)	11.02	11.07	11.09	11.04	11.037	11.09

Table 2 Physico-chemical variables determined in situ at the Atlangatepec Dam, Tlaxcala, Mexico, November 2021
Source: Own elaboration

Regarding the biodiversity of phytoplankton, filamentous cyanobacteria predominated (see Figure 3). These results agree with those reported by Romo et al., 2008, in the Albufera Lake of Valencia during the period 1980-2006; filamentous cyanobacteria predominated, representing 76% abundance; It is worth mentioning that between 1985 and 1988 P. agardhii represented 67% of the average annual biovolume in the lake and was not found when the nutrient supply was diverted to the water body. During 1998-2006 *P. gaelata* predominated with 34% of total mean abundance (V).

Similar results were found in three lakes in Mexico in which cyanobacteria predominated: *Microcystis* sp., *Planktothrix*, *Anabaenopsis*, *Pseudanabaena* and *Phormidium*, all of them with the capacity to produce microcystins (I).

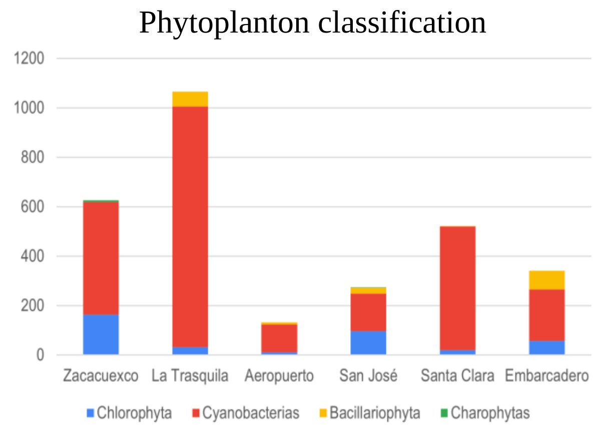

Figure 3 Classification of phytoplankton present in the Atlangatepec dam, Tlaxcala, Mexico, November 2021
Source: Own elaboration

Morphologically, 10 chlorophytes were identified: *Pediastrum simple var. duodenarium, Pediastrum simplex Meyen, Pediastrum duplex Meyen, Volvox sp, Chlorella saccharophila, Scenedesmus opoliensis, Sphaerocystis agardhiii, Tetraëdron, Ankistrodesmus falcatus, Ulotrhrix tenuissima.* Fifteen cyanophytas were classified: *Dolichospermun planctonicum, Planktothrix agardhii, Pseudanabaena moliniformis. Pseudanabaena galeata, Pseudanabaena linmetica, Anabaena, Coelosphaerium, Aphanizomenom gracile, Aphanizomenom issatschenkoi, Chroococcus limneticus Lemmermann, Lyngbya, Microcystis pulverea f. incerta, Aphanocapsa incerta, Anabaena Spiroides, Raphidiopsis mediterranea.*

A species of Charophyta: *Zyg. and four Bacillariophyta species: Coscinodiscus Ehrenberg, Aulacoseira granulata var. angustissima, Guinardia flaccida, Asterionellopsis glacialis* (see Figure 4).

It is worth mentioning that in a study of diatom biodiversity carried out in 2011, in the Atlangatepec Dam, 16 species were identified: *Achnanthidium exiguum, Amphora copulate, Aulacoseira granulate, Cocconeis placentula, Cymbella Mexicana, Encyonema silesiacum, Epithemia sorex Kützingzing, Gomphonema affine Kützing , Hantzschia amphioxys, Navicula gregaria Donkin, Nitzschia amphibia Grunow, Nitzschia filiformis, Rhoicosphenia abbreviate, Sellaphora pupula, Tryblionella apiculata W. Gregory, Ulnaria ulna (IX).*

Our results with less diversity of diatoms differ from those reported, this is because the monitoring of these species is generally carried out in the littoral zone and our study was carried out in the limnetic zone of the body of water.

Figure 4 Fitoplanton recovered from the Atlangatepec dam. From left to right *Pediastrum duplex*, *Raphidiopsis mediterranea*, *Anabaena*, *Scenedesmus opoliensis*, zig, *Aulacoseira granulata*, *Pediastrum simple*. 40X compound microscope, lugol was used as contrast.

In the study carried out, the dominant species was *Pseudanabaena galeata*, and it predominated at the 6 monitoring points, followed by *Planktothrix agardhii* (see Figure 5). These results differ from a study carried out in three urban lakes in Mexico in which *Microcystis* sp., As well as *Arthrospira* sp. and *Planktothrix agardhii*, were found in abundance (II).

According to the literature (II), the genus *Pseudanabaena* has unmistakable morphological characteristics, which clearly distinguish it from other filamentous cyanobacteria, such as its solitary and short growth as well as constrictions on its walls.

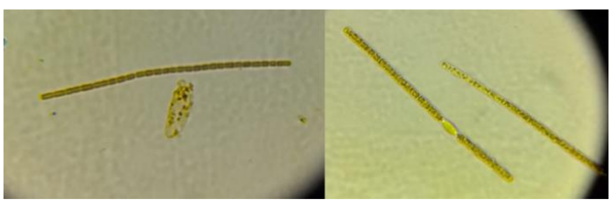

Figure 5 Photograph obtained with a Leica microscope, with a 40X objective, *Pseudanabaena galeata* and *Plankthotrix*, recovered from the Atlangatepec dam
Source: Own

Figure 6 shows the phylogenetic tree of the identified cyanobacterial species. In the topology of the tree, two main branches can be observed, which separates *Plankthotrix agardhii* from the rest of cyanobacteria. In the second bifurcation *Holothece* sp. At the next level of branching it is possible to distinguish *Pseudanabaena* from another group of cyanobacteria.

Abiotic (physical-chemical) and biotic (biological) criteria define water quality and its potential use. However, when the permitted limits of some parameters are exceeded, they infer the conservation of the aquatic ecosystem and the protection of human health.

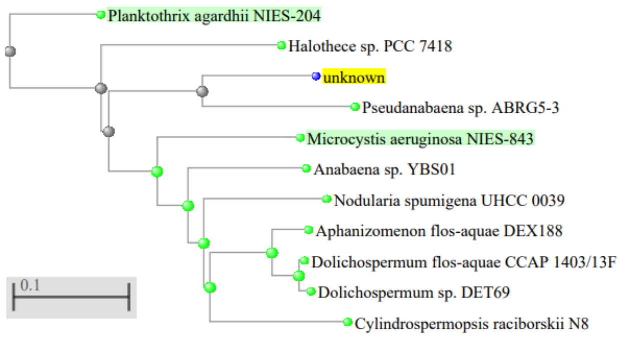

Figure 6 Phylogenetic tree obtained by Blastn, of cyanobacterial species and their accession numbers of the identified sequences, in the Atlangatepec dam, Tlaxcala, Mexico, November 2021

Cyanobacterial blooms have negative impacts on aquatic systems, they inhibit the growth of other microalgae and macrophytes due to the formation of secondary metabolites that are toxic to other species. The degradation of the algal flowering decreases the oxygen concentration in the water body promoting the death of fish, therefore the formation of blooms affects the aquatic diversity and the ecosystem (VII).

The monitoring activities carried out, on the one hand, allowed to know the impact of anthropogenic activities in the dam by obtaining data and using them, they will help in taking actions for the sanitation and sustainable use of the dam.

The dominance of cyanobacteria implies risks that must be known and managed, because they retain the ability to produce microtoxins, considered freshwater pollutants that outweigh heavy metals and pesticides (VI).

It is necessary to reduce the entry of phosphorus into the body of water since it promotes the massive growth of cyanobacteria. It is necessary to establish a conservation and sustainable management plan (PCyM), which promotes sanitation activities and compromises the participation of society for the establishment of good environmental practices and avoids the direct discharge of wastewater, which increases the content of organic matter and eutrophication processes.

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Conclusions

The predominance of *Pseudanabaena galeata* and *Plancthotrix agardhii* puts the diversity of phytoplankton and the balance of trophic chains in the water body at risk because these species are potentially cyanotoxin-producing.

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