Monitoring and treatment of cyanobacterial contaminated surface waters in France and Cyprus

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Abstract
Over the past few decades eutrophication of surface water has increased worldwide because of different anthropological activities including land fertilization and sewage run-offs; in combination with climate change. Excessive amount of nutrients (phosphorus and nitrogen), is now detected in freshwater lakes, artificially made reservoirs, and streams. Cyanobacterial nuisance in surface waters is among the most current environmental issues as it causes socio-economical problems. CYANOS is a two-year bilateral project funded from the Research Promotion Foundation of Cyprus and the Campus France of France with interdisciplinary activities that combines surface water monitoring and on-site water treatment for the restoration of eutrophic surface waters. Specifically, CYANOS aims to monitor the seasonal variation of cyanobacterial harmful algal blooms (cyano-HABs) in the surface waters of Cyprus and France and explore emerging on-site treatments to control their formation.
Keywords: eutrophication, cyanobacteria, cyanotoxins, in-lake treatment

1. Introduction

Cyanobacteria, also called "blue-green algae", are common, naturally-occurring micoorganisms of fresh, marine and brackish waters worldwide. They have key roles in natural environments, forming the basis of aquatic food chains and in the recycling of matter. However, when excess nutrient fluxes make it into a lake, cyanobacterial populations vastly increase (also known as "blooming") resulting in the formation of mats. These mats prevent oxygen and light penetration (Paerl & Otten, 2013). Cyanobacteria harmful algal blooms (cyano-HABs), besides greatly altering water's quality characteristics by adding undesirable color, odor and taste, they also have the ability to produce, release and excreted a wide range of secondary toxic metabolites (commonly known as cyanotoxins) for invertebrates and vertebrates including mammals. Cyanobacteria genera such as *Microcystis*, *Plankothrix*, and *Cylindrospermopsis* are known to produce a wide range of cyanotoxins. Cyanotoxins vary structurally (cyclic peptides, alkaloids, lipopolysaccharides) and can cause irritations of the skin and other organs (dermatotoxins and irritant toxins), cell damage (cytotoxins), and liver damage (hepatotoxins), or affect the nervous system (neurotoxins). Because of their high lethality (LD_{50} values are lower than the current priority substances), the World Health Organization (WHO) establish a guideline of 1.0 μg/L as a maximum concentration for the hepatotoxin microcystin-LR in drinking water supplies (WHO 2008). Economic impact of cyano-HABs is affecting different sectors including medical, tourism, fishing, and water management (Sanseverino, 2016).

Currently there are no data in the cited literature regarding the occurrence of cyano-HABs in Cyprus. Though the Water Development Department (WDD) of Cyprus takes sporadic samples to measure cyanobacterial biovolume, the agency does not check for the presence of cyanotoxins with advanced analytical techniques. In addition, these data are not readily available. Therefore, the main objectives of the CYANOS project are: 1) To identify the taxonomy of microalgae and cyanobacteria using microscopy and camera imaging tools, as well as molecular tools after sampling surface waterbodies of Cyprus. 2) To develop a monitoring strategy and an early warning system for diagnosing cyano-HABs on surface waters based on simple tools such as probes (i.e., for the detection of phycocyanin a pigment found only in cyanobacteria), followed by analysis of the physicochemical parameters of the water column. 3) To test the efficiency of a relative new on-site treatment (e.g. H_2O_2 treatment) for controlling the blooming and the appearance of cyanobacteria (Matthijs, 2012). Herein, data from the monitoring of the Polemidia Dam, located outside the city of Lemesos, during the summer of 2018 will be presented (CYANOS).

2. Methods

Seasonal sampling at cyanobacterial contaminated sites in Cyprus took place during 2017-2019. The most contaminated location in Cyprus is the Polemidia Dam located outside the city of Lemesos. The Water Development Department performs 6 sampling events annually by taking column samples from the euphotic zone (secchi disc depth multiplied by 2.5) of the dam.

After obtaining permission from the WDD, we have performed additional sampling but the water was collected from the surface. Since the dam is used for irrigation, the outlet of the dam was sampled as well. Fresh and preserved with lugol solution samples were taken to the lab and were observed under a trinocular microscope with a camera (Nikon), for species taxonomy (i.e., cyanobacteria, diatoms, dinophyta, chlorophyte). The water quality characteristics of water were recorded including nutrients, dissolved oxygen, temperature, total suspended solids, pH, and conductivity.

3. Results and Discussion

The collected water samples were processed based on the analysis that would have followed (nutrients, pigments, cyanotoxins, and cyanobacteria). Fresh samples were taken to the lab for observation under a trinocular microscope with a camera (Nikon). In addition, concentrated samples on a polycarbonate filter (porosity 5 μm) (volume of filtered water 100 mL) were observed as well. A chlorophyl-a and phycocyanin probe were used to monitor the concentration of the pigments in water from the Polemidia Dam and the outlet. Overall, it was found that phycocyanin's concentration was higher than chlorophyl's indicating the presence of higher concentration of cyanobacteria compared to green algae. Observation under the microscope revealed the taxonomy of the different species.

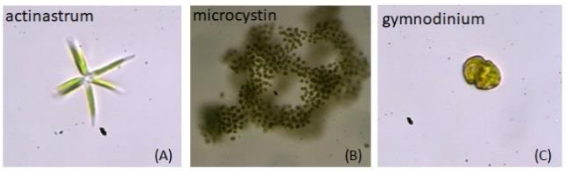

Figure 1. Species identified in the Polemidia Dam samples, observed under a trinocular microscope in year 2018. (A) Chlorophyta, (B) Cyanobacteria, (C) Dinophyta.

All the species were categorized into: cyanobacteria, diatoms, dinophyta, chlorophyta, and unidentified species. The outlet sample (~2m depth) had a significantly lower concentration of cyanobacteria and green algae than the dam sample (surface sample) and was mainly composed by green algae though a few colonies of *microcystis sp*. were observed. Cyanotoxins analysis with LC-MS/MS indicated that the intracellular cyanotoxin concentration was 3.4 ng MC-LF /L of filtered water. In addition, oligopeptides that belong to the categories of aeruginosins (AER) and anabaenopeptins (AP) were also detected.

Table 1. Chlorophyll-a and phycocyanin concentrations (μg/L) in Polemidia Dam (June 2018).

Polemidia

	Inlet Conc. (μg/L)	**Outlet Conc. (μg/L)**
Chlorophyll-a	4.47	0.07
Phycocyanin	67.00	1.12

4. Conclusions

To conclude, the CYANOS project will shed light on the seasonal variation of cyano-HABs under the climatic conditions of Cyprus (increasing sunlight and heat, variation in nutrient fluxes, and toxicity of bloom). The outlet of the dam is located so that it draws water below the euphotic zone to minimize the probability of cyanobacteria entering the water that goes for irrigation. Since our measurements still find low levels of phycocyanin, it is advised to make use of portable phycocyanin sensors to adjust the depth of the outlet to further insure that cyanobacteria are not found in the outlet water that is used for irrigation.

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