Additional Material

Occurrence of a single species cyanobacterial bloom in a lake in Cyprus: Monitoring and treatment with hydrogen peroxide releasing granules

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Introduction

The most applied stoichiometric reference is the Redfield ratio which describes the nutrient limitation of planktonic production in coastal waters based on the TN:TP (Total Nitrogen: Total Phosphorus) molar ratio [1,2]. Even though Redfield proposed this ratio for its use on oceanic studies, it was well adopted as a universal nutrient limitation threshold with multiple citations in different types of aquatic systems. Redfield referred to an average N:P molar ratio that when exceeds 16, phosphorus becomes the limiting element for phytoplankton growth while when the ratio is below 7, nitrogen is the limiting element. Over the years, different ratios and approaches have been developed for better understanding of the limiting element based on the specific conditions of each waterbody (e.g., nutrient sources, phytoplankton species present). Based on the cited literature, lakes with different characteristics showed different correlations of nutrients with their trophic status, and the efficiency of the Redfield ratio varied in each case [3]. Recent studies indicated that higher TN:TP molar ratios (>22, P-limitation) are more suitable for surface waters while DIN:TP (Dissolved Inorganic Nitrogen: Total Phosphorus) and NO_3^-:TP mass ratios have been used more often during the past years for determining the limiting elements in lakes [4]. Studies on dissolved nutrients mass ratios (DIN:TP, NO_3^-:TP, NH_4^+:TP suggested that in freshwater systems the DIN:TP mass ratio is a better indicator than the TN:TP molar ratio. Also, the NO_3^-:TP mass ratio performed even better than DIN:TP as DIN includes ammonium which in some studies it showed a weak correlation with N-limitation, resulting in a weaker model [5].

Table S1. Nutrient limitation approaches and thresholds based on TN:TP, DIN:TP and NO_3^-:TP ratios in different aquatic environments.

Ratio	Units	Threshold	Nutrient Limitation	Reference	Aquatic environment studied
TN:TP	Molar	<7	N – limitation	[1,2]	Ocean
		>22	P – limitation		
TN:TP	Molar	<20	N – limitation	[3]	Lakes; Ocean
		20-50	N or P limitation		
		>50	P – limitation		
DIN:TP	Mass	<9	N – limitation	[4]	Lakes; ponds
		9-22	N or P co-limitation		
		>22	P – limitation		
NO_3^-:TP	Mass	<9	N – limitation	[4,5]	Lakes
		9-22	N or P co-limitation		
		>22	P – limitation		

The main input of nutrients into the waterbodies comes from agriculture due to the misuse of fertilizers. The European Commission of Environment of the European Union has composed the Water Framework Directive (2000/60/EC) which aims to protect surface waters from chemical pollution [6]. Recently, it was proposed to include a group of cyanotoxins (microcystins) in a revised Drinking Water Directive on the Quality of Water Intended for Human Consumption [7]. If this comes through, it will enforce public authorities to include cyanobacteria and cyanotoxins into their monitoring and mitigation strategies in order to comply with the new directives and legislations of EU.

Additionally, the Organization for Economic Co-operation and Development (OECD) has actively participated in the development of frameworks and guidelines by reforming the surface water quality regulations in Eastern European and Central Asian countries [8-10]. Based on those, OECD proposed a surface water classification (I – V class; excellent – bad) system based on different water quality characteristics. Its use has been widely adopted for monitoring water quality and apply restorative measures when needed. In the present study, water class was evaluated based on the limits set by OECD for acidification status and total and dissolved nutrient concentration as shown in Table 2.

Table S2. Proposed surface water quality standards by OECD based on the EU Directive (2000/60/EC).

Parameter	Unit	Class I	Class II	Class III	Class IV	Class V
pH	[-]	6.5 – 9.0				
Total Nitrogen	[mg NL-1]	1.5	4	8	20	>20
Nitrate	[mg NL-1]	1	3	5.6	11.3	>11.3
Nitrite	[mg NL-1]	0.01	0.06	0.12	0.3	>0.3
Ammonium	[mg NL-1]	0.2	0.4	0.8	3.1	>3.1
Total Phosphorus	[mg NL-1]	0.1	0.2	0.4	1	>1
Orthophosphates	[mg NL-1]	0.05	0.1	0.2	0.5	>0.5

Sampling site and dates of sampling

Scheme S1. Lakes in the National Forest Park of Athalassa: (A) St. George Lake and (B) Athalassa Lake located at the Athalassa National Forest Park.

Table S3. Sample number and date of sampling event in St. George Lake.

Sample nº	Date
1	25/02/2019
2	04/03/2019
3	18/04/2019
4	12/07/2019
5	06/08/2019
6	22/08/2019
7	09/09/2019
8	15/10/2019
9	12/11/2019
10	09/12/2019

Results

Monitoring

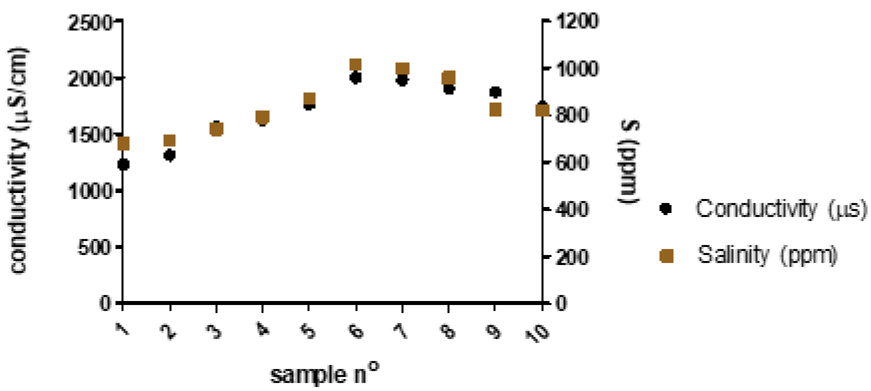

Figure S1. Conductivity (left axis) and salinity (right axis) measured with EXTECH portable probe in St. George Lake surface samples.

Cyanotoxins analysis

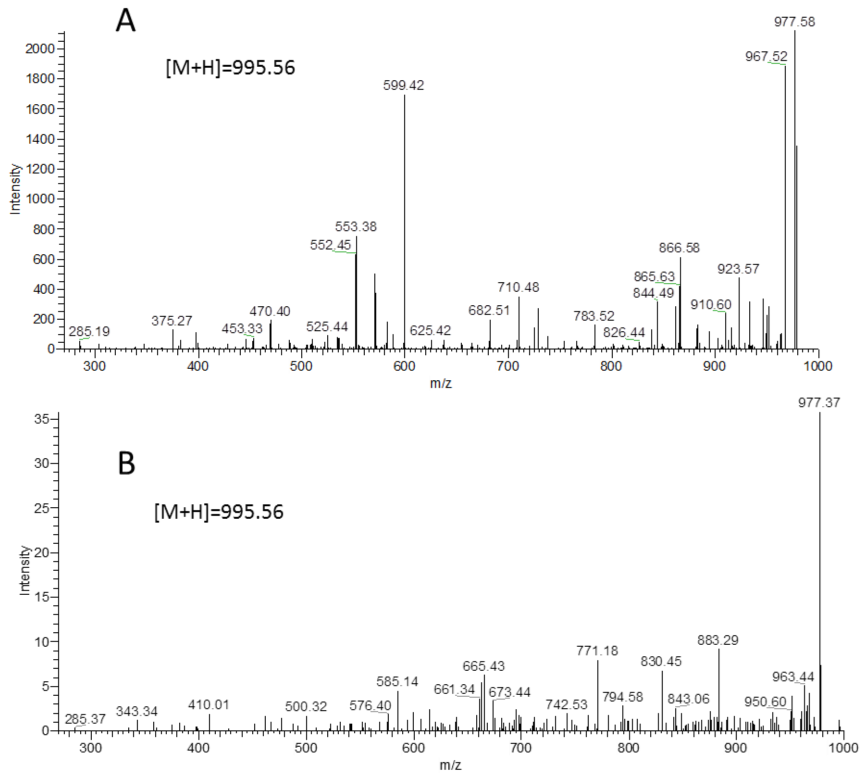

Figure S2. MS/MS spectra of the (A) MC-LR standard compound and (B) compound detected in samples with [M+H] =995.56 but different fragmentation pattern.

References

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