Algological Studies 117 (Cyanobacterial Research 6)	393–406	Stuttgart, October 2005

Cyanobacteria blooms in Sete-Cidades lake (S. Miguel Island – Azores)

By MARIA DA CONCEIÇÃO RAIMUNDO SANTOS[1], DINA MARIA MEDEIROS PACHECO[2], FERNANDO SANTANA[1] and HELENA MUELLE[1]

[1] Department of Environmental Sciences and Engineering – Faculty of Sciences and Technology – New University of Lisbon, Caparica, Portugal
[2] Azores Government Environmental Secretary, Ponta Delgada, Portugal

With 8 figures and 2 tables in the text

Abstract: Sete-Cidades is a beautiful softwater lake located in a volcanic crater at S. Miguel island (Azores's archipelago). The lake has an area of 445.5 ha and in it's narrowest zone is crossed by a hand made bridge dividing it in two interconnected sub-units, Lagoa Azul (the Blue Lake – 357.3 ha, depth 24 m) and Lagoa Verde (the Green Lake – 88.2 ha, depth 21 m).

In the mid 20th century, some forested areas of their steep watersheds were transformed in grasslands and pastures. This human environmental disturbance and a rainy climate increased soil erosion and led to a progressive enrichment in nutrients of the water bodies. Sete-Cidades lake can still be classified as mesotrophic, having presented an average chlorophyll a concentration of 3–8 mg . m^{-3} and a total phosphorus average concentration of 15–23 mg. m^{-3} for the last three years.

As a result of eutrophication processes phytoplankton population increased along the years and cyanobacteria blooms began to develop, especially after the year 2000. Since the beginning of monitonng programs, in 1988, until 1998, phytoplankton population varied from 1.0 106 cells . l^{-1} to 8.0 106 cells . l^{-1}. In the autumn and winter seasons of 1997198, severe raining events gave rise to intense runoff from watersheds and edges leading to an increase of phytoplankton population during the next few years, reaching the maximum cell density of 4842.7 106 cells . l^{-1} inside Lagoa Azul, in July 2003. Cyanobacteria began to dominate and they represent now the major phytoplankton group all over the year.

Cyanobacteria blooms in this lake normally begin in April-May and continue through October. Bloom conditions were also detected in winter samples (446.7 10^6 cells . l^{-1} in a water sample collected from Lagoa Azul in February 2003). The dominant cyanobacteria species blooming in the lake was *Microcystis* sp., mainly *Microcystis aeruginosa*, which formed yellowish-green dense "scums" floating on the surface, when calm weather conditions prevailed.

Water and biomass samples analysis enabled to detect the presence of soluble and intracellular microcystins. Maximum levels were detected in Lagoa Azul samples collected in July 2002. The concentration of soluble toxins was only 0.03–0.05 mg . m^{-3} MC-LR eq., but in biomass samples from surface to 5 m deep, toxin concentrations reached 3.78–7.83 mg . m^{-3} MC-LR eq., corresponding to 154–3780 mg . Kg^{-1} dry weight.

0342-1120/05/0159-393 $ 3.50
© 2005 E. Schweizerbart'sche Verlagsbuchhandlung, D-70176 Stuttgart
Algological Studies 117 = Arch. Hydrobiol. Suppl. 159

This paper will present some of the results obtained in Sete-Cidades lake eutrophication monitoring program, being it's main objective the report and description of cyanobacteria blooms occurrence.

Key words: **Water quality, nutrients, eutrophication, cyanobacteria blooms.**

Introduction

S. Miguel island, also called the Green Island, is the largest island of the Archipelago of the Azores (37°50'N; 25°30'W). It has an area of 759.41 km^2, a length of 65 km and a width of 14 km. It was discovered by Portuguese ships in the XIVth century and became populated in 1439. Throughout time, agriculture always had a central role in the regional economy, however in the middle of the 20th century, the strategies of regional development have changed, giving primacy to cattle breeding and dairy farming.

The Sete Cidades lake is located in a volcanic crater of the island northwest region. The lake surface is 251 m above sea level, covers 445.5 ha and in it's narrowest zone it is crossed by a hand made bridge dividing it in two interconnected sub-units: Lagoa Azul (the Blue Lake – 357.3 ha, depth 24 m) and Lagoa Verde (the Green Lake – 88.2 ha, depth 21 m). The lake's area is about 23 % of its total drainage basin area. Average air temperatures in this region range from 11 ''C in winter to 23 ''C in summer, with a very short daily thermal amplitude. The Sete-Cidades climate is strongly influenced by site orography. Monthly average precipitation normally ranges from about 60 mm in August to 250 mm in January. However, rainy events greater than 100 mm are quite frequent. Total runoff from the drainage basin to the lake is about 14.4 10^6 m3. $year^{-1}$, flowing mainly from two small torrential streams.

In the middle of the last century, destruction of trees and natural vegetation in the catchment area around the lake, to enlarge pasture area, increased the runoff of nutrients, particularly phosphorus and nitrogen, from the steep land. The existence of a small village in one of the lake's edges, lacking a domestic sewage sewer system until five years ago, also contributed to a gradual and increasing pressure on the ecosystem, resulting in an overabundant development of rooted aquatic macrophytes and algae in the water bodies.

The eutrophication of Sete Cidades lakes, due to nutrient enrichment enhanced by human agricultural and domestic activities, began to be noticed in 1987 and led to the implementation of a monitoring programme, in order to evaluate the situation, in 1988189 (SANTOS et al. 1991, 1992). Carlson's trophic state index (CARLSON 1977) resulting from this earlier survey, based on six bimonthly sampling campaigns, ranged from 36 to 56, indicated that there was an eutrophication process in progress. At that time, the situation was worst in Lagoa Verde due to wind direction dominance. Chlorophyll water column average results ranged from 1.82 $mg.m^{-3}$ to 9.88 $mg.m^{-3}$ in Lagoa Azul samples and 3.43 $mg.m^{-3}$ to 13.81 $mg.m^{-3}$ in Lagoa Verde samples. The highest results were ob-

tained in winter **samples.** Inorganic **nitrogen** varied from 100 to 550 mg.m^{-3}, the **maximum** value **being characteristic** of a **meso-eutrophic situation** (**VOLLENWEIDER 1968** in HARPER 1992). **Phosphorus results** were **inconclusive due** to a **lack** of adequate **analytic** methodology.

Phytoplankton **community** studies, performed by OLIVEIRA in 1987188 (OLIVEIRA 1989) **concluded** that the **dominant** species in these lakes were:

Lagoa Azul	winter	***Aulacoseira ambigua*** (Melosira)
		Gloeocystis sp.
		Anabaena ***cylindrica***
	spring	***Melosira ambigua***
	summer	***Botryococcus braunii***
		Anabaena cylindrica
		Aphanizomenon ***flos-aquae***
Lagoa Verde	winter	***Melosira ambigua***
	spring	Synedra ***acus***
		Pseudanabaena planktonica (Oscillatoria)
	summer	***Aphanizomenon flos-aquae***

The **same study** noticed the **disappearance** of Dinobryon sp. that **had been detected** in the end of the **19th century** (**BARROIS** 1896).

According to SANTOS et al. **(1991,1992),** in 1988189, phytoplankton **populations** varied from 0.57 10^6 **cells**.l^{-1} to **1,1** 10^6 celis.l^{-1} **in Lagoa** Azul and from 1.5 10^6 celis.l^{-1} to 4.8 10^6 cells.l^{-1} **in Lagoa** Verde. **Phytoplankton populations** in **Lagoa** Azul were **dominated** by **Bacillariophyceae** in **autumn** (***Aulacoseira ambigua (Melosira)***), **Cyanobacteria in** winter (***Pseudanabaena planktonica (Oscil-***latoria)) and **spring** (*Aphanizomenon flos-aquae*) and **Dinophyceae** (***Peridinium*** spp.) in **summer.** In **Lagoa** Verde, **phytoplankton populations** were **dominated** all over the year by Cyanobacteria (Pseudanabaena planktonica (Oscillatoria), Aphunizomenon ***flos-aquae*** and ***Anabaena*** cylindrica).

The macrophyte population was, and **still** is, **composed** of exotic submerged species, **mainly** ***Elodea canadensis,*** Egeria densa, ***Potamogeton polygonofolius*** and ***Myriophillum alterniflorum*** (**PACHECO** et al. 1998). They were **first discovered** in the early 1970s and **since** then they have **spread** very **rapidly** in the **shallow** zones of the lake. It is believed that these **macrophytes** were **introduced accidentally** by **individuals** who emptied aquariums into the **water,** or **deliberately** to **provide habitat** for some fish species. **Despite most** experts **scepticism regarding** aquatic weed **harvesting** as a lake **restoration** and management **tool** (**MADSEN 1993),** this method has **been used** in Sete-Cidades lake, since 1995, especially to **improve its aesthetic** value and **facilitate recreational activities.**

Studies performed by PORTEIRO et al. (1998) showed that **this** lake **nutrient** loading, **arriving through** watercourses, was **about** 4.59 g N.m^{-2} per year and 0.72 g P. m^{-2} **year**$^{-1}$. According to **VOLLENWEIDER** (in HARPER **1992),** permissible loadiig in **lakes** as deep as Sete-Cidades lake **is** up to 2.40 g.m^{-2} per year of N and 0.16 g.m^{-2} **per** year of P and **dangerous loading is** up to 4.88 g.m^{-2} per year of N and 0.32 $g.m^{-2}$ per year of P. So, the estimated nitrogen loading flowing from the rivers was 1.9 times higher than the permissible value but still bellow a dangerous level. On what concerns phosphonis, the nutrient more frequently responsible for eutrophication, the loading estimated was 4.5 times higher than the permissible and 2.3 times above the dangerous level.

Sete-Cidades steep watersheds can cause important diffuse surface runoff, mainly when strong rainy events occur. As nutrient losses from permanent pastures can attain 10–40 % of added nitrogen (ANONYMOUS 1983 in HARPER 1992) and 6–12 % of added phosphorus (COOKE & WILLIAMS 197011973 in HARPER 1992), nitrogen and phosphonis loading from Sete-Cidades pastures area can be as high as 9.63 $g\ N.m^{-2}$ per year and 0.35 $g\ P.m^{-2}$ per year (SANTOS et al. 2002). Nitrogen losses from undisturbed forested catchments was estimated by BORMANN et al. (1968) (in HARPER 1992) to be 2–5 kg $N.ha^{-1}\ year^{-1}$. Phosphorus average losses from igneous forested catchments was estimated to be 48 g $P.ha^{-1}\ year^{-1}$, but they can be as high as 720 g $P.ha^{-1}$ per year in volcanic catchments (DILLON & KIRCHNER 1974 in HARPER 1992). Based on these figures and on Sete-Cidades basin forested area, SANTOS et al. (2002) concluded that it could contribute with a nutrient loading of 0.37 g $N.m^{-2}\ year^{-1}$ and 0.13 g $P.m^{-2}\ year^{-1}$. These approaches led to a diffuse loading estimation of 9.97 g $N.m^{-2}\ year^{-1}$ and 0.48 g $P.m^{-2}\ year^{-1}$, corresponding to 2.0 and 1.5 times the dangerous loading of N and P to this lake.

The nutrient loading that Sete-Cidades lake has been receiving during the last years might be even higher. In fact, some preliminary analysis of runoff water samples (PORTEIRO et al. 1998) indicated that the loading could attain 24.6 g $N.m^{-2}$ per year and 2.% g $P.m^{-2}\ year^{-1}$. Nevertheless, these results must be confirmed.

When temperature, light and nutrient availability are adequate to phytoplankton growth, surface waters may host algae or cyanobacteria blooms. The term bloom used here means a celi concentration greater then 20 10^6 celis. l^{-1}, the definition normally adopted in potable and recreational waters (OLIVER & GANF in WHITTON & POTTS 2000). In eutrophic waters, cyanobacteria often dominate the summer and early autumn phytoplankton, while during winter and spring seasons they are replaced by diatoms. However, cyanobacteria can be present and even dominate all over the year (CHORUS & BARTRAM 1999). Cyanobacteria dominance in temperate areas warmer months was explained by a better adaptation of these organisms to higher temperatures, their ability to capture reduced photosynthetic photon flux densities and to utilise low N:P ratios (SCHREURS 1992 in CHORUS & BARTRAM 1999) and low carbon dioxide concentrations. According to ROBARTS & ZOHARY 1987 (in CHORUS & BARTRAM 1999), maximum growth rates are attained by most bloom-forming cyanobacteria at temperatures above 25 °C. SMITH 1983 concluded that a low ratio between nitrogen and phosphonis concentration may favour cyanobacterial blooms development. Others have found little evidence that TN : TP rabos could be important to determine cyanobacterial dominance (HARRIS 1986, PICK & LEAN 1987, ELSER et al. 1990, JENSEN et al. 1994, SCHEFFER et al. 1997, OLIVER & GANF in WHITTON & POTTS 2000). Cyanobacteria that form surface scums seem to have higher tolerance to high light intensities while the ability of other cyanobacteria genera to grow at low light intensities and to harvest certain specific light qualities enable them to grow in turbid waters (CHORUS & BARTRAM 1999).

Other characteristics, exclusive to only some of cyanobacteria genera, like buoyancy regulation, phosphorus storage capacity and reduced zooplankton grazing may enhance bloom development (STEINBURG & HARTMANN 1988, SHAPIRO 1990, BLOMQVIST 1994, OLIVER & GANF in WHITTON & POTTS 2000). Buoyancy and its regulation provide gas-vacuolate cyanobacteria with significant advantage over micro-algae, especially when water turbulence is low and waters are thermally stratified, because it enables them to maintain their cells at maximum growth conditions and prevent sedimentation. Inorganic nitrogen sources available seem to influence cyanobacteria success. Ammonium nitrogen seem to favour non-nitrogen fixing cyanobacteria, while nitrate enables micro-algae development and nitrogen-fixing cyanobacteria are favoured by nitrogen deficiency (OLIVER & GANF in WHITTON & POTTS 2000).

According to CHORUS & BARTRAM (1999), although eutrophication has been recognised as a water quality problem of growing concern since 1950s, only recently cyanobacterial toxins were recognised as a human health problem deriving from eutrophication. A significant proportion of cyanobacteria can produce one or more types of toxins which, when present in drinking or recreational waters, might lead to human health problems. In aquatic environments, these toxins usually remain within cyanobacterial cells and are only released when cell lysis occur. The most frequently cyanobacterial toxins found in freshwaters blooms are microcystins and nodularin, which are cyclic peptides hepatotoxins.

As the existence of a eutrophication process in Sete-Cidades lake was already known since 1980's, the arise of cyanobacteria blooms in 2000 wasn't a surprise. These blooms became more serious after severe raining events that occured in 1997's autumn and 1998's winter which lead to an increase of nutrient transportation to the water bodies. This paper presents some of the information collected during sampling field campaigns performed, from 2000 until 2003, for water quality monitoring, mainly in some aspects related to cyanobacteria blooms formation.

Materiais and methods

The monitonng program was initiated in February 2000 and lasted until October 2003. Fourteen sampling expeditions were conducted, four in 2000, 2002 and 2003 and two in 2001.

Water samples were collected, from a boat, at surface, 2.5 m, 5 m, 10 m and 15 m deep and 0.5 m above sediments, in one sampling point located in the middle of each lagoon, Azul and Verde, at the deepest zone. Samples were collected with a Van Dom type device, transported in refrigerated polyethylene bottles (glass for samples addressed to phosphorus analysis) to a laboratory and analysed (Standard Methods 1998) for physical and chemical parameters (acidity; alkalinity; total suspended solids; DOC; nitrogen and phosphorus compounds; chlorophyll a and phaeophytin a). The variables temperature, pH, dissolved oxygen, turbidity and conductivity were determined *in situ*, with a multiparameters meter. Transparency was determined with a Secchi disc at sampling points.

Samples for phytoplankton analysis were collected at the same levels of deepness, equal volumes mixed and one aliquot placed in a polyethylene flask and preserved with a 1% Lugol solution. Phytoplankton identifications were performed with an optical microscope Leica DML and for cell (colony) counting the method used was the UTERMÖHL's (LUND et al. 1958).

Microcystins determinations were performed in water and biomass samples collected at the same deep levels previously described. Water samples were filtered in a glass fibre membrane (GF/F, pore size ≈ 0.7 µm) and the filters with the biomass were kept frozen at −18 °C until toxin extraction with methanol. Soluble toxins were extracted at once with methanol using a SPE method. The filtered samples were passed through a C18 cartridge (Sep-Pak Waters Corporation) preconditioned with methanol and washed with water, at a flow rate not exceeding 10 ml/min. The cartridge was then eluted with 3 ml methanol acidified with 0,1% TFA. The extracts were evaporated until dryness with a nitrogen stream, the residue resolved in 250 µl methanol and centrifuged for 10 min at 10000 rpm. Filter discs were placed in glass beakers containing 20 ml of methanol and allowed to extract for 24 hours at −4 °C. The samples were evaporated under pressure at 40 °C until dry, the residue resolved in 500 µl methanol and centrifuged for 10 min at 10000 rpm. Centrifuged extracts were analysed in a HPLC system with photodiode array detection, with a Symmetry C18 column (4.6 x 150 mm; 5 µm). Results were expressed in equivalent units of microcystin LR, the standard used for calibration (LAWTON et al. 1994, CHORUS & BARTRAM 1999).

The values of Carlson's throphic state index (TSI), associated to parameters Secchi disc transparency (SD), chlorophyll a (chl a) and total phosphorus (TP) concentrations were calculated from the following equations (CARLSON 1977 in COOKE et al. 1993):

$$TSI = 10\,(6 - \log_2 SD)$$
$$10\,(6 - \log_2 7.7/\text{chl a}^{0.68})$$
$$10\,(6 - \log_2 48/TP)$$

Results

Surface water temperature of this lake, at eleven o'clock a. m., ranged from 12–13 °C in late autumn and winter samplings to 22–23 "C in summer. Thermal stratification lasted from May to September/October enabling us to classify Sete-Cidades as a warm monomictic lake. The transparency of Lagoa Azul varied from 4.8 m, in July 2000, to 0.2 m in July 2002. In Lagoa Verde, the maximum transparency recorded was 4.6 m, in July 2000 and minimum vaiue 1.3 m in May 2003. In 1992 (SANTOS et ai. 2002), transparency values of 7.5 m were recorded in Lagoa Azul, but Lagoa Verde, smailer than Lagoa Azul and more gloomy, never reached a transparency greater than 4.6 m (Fig 1). The smallest transparencies in both lakes were recorded during surface bloom situations.

Inorganic nitrogen water column average concentrations were quite variable (Fig. 2). In the 1992193 time (SANTOS et al. 2002). they fell to 90 mg N. m^{-3} (Lagoa Azul) and 125 mg N. m^{-3} (Lagoa Verde), values corresponding to ultra-oligotrophy (VOLLENWEIDER 1968 in HARPER 1992). In 2000/2001 they raised to 540 mg N. m^{-3} in Lagoa Azul and 620 mg N. m^{-3} in Lagoa Verde (meso-eutrophy) and in 200212003 they fell again, in the same water bodies, to 104 mg N. m^{-3} and 258 mg N. m^{-3}. Inorganic nitrogen in 200112003 was mainly in the ammonium form. Nitrate accounted for 68–73% of inorganic nitrogen in 2000,

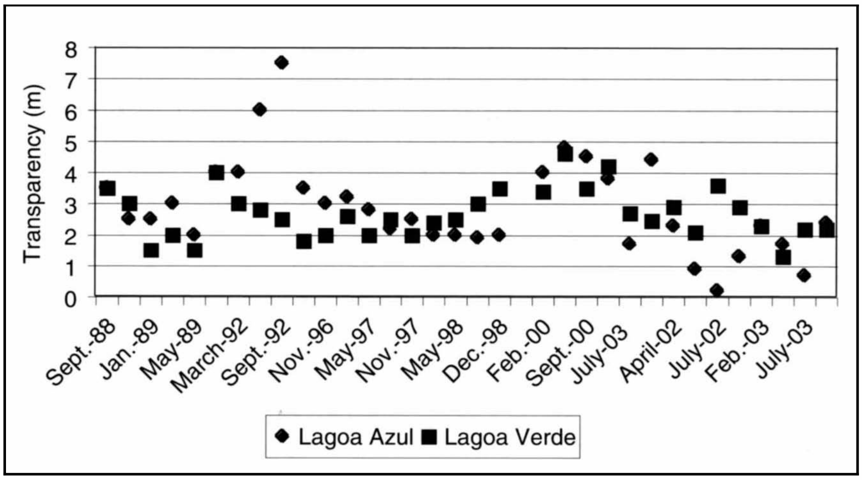

Fig. 1. Transparency variation in Sete-Cidades lake from 1988 until 2003 (SANTOS et al. 2002, INOVA, 1996, 1998).

but, after that, it became very scarce or absent in the samples collected inside the lagoons, perhaps because it had been readily assimilated as soon as it reached the water bodies.

Total phosphorus water column average concentrations were not as variable as inorganic nitrogen was (Fig.2). Until 1996 there were no reliable results of total phosphorus. Total phosphorus average concentrations of water column were 20 mg P.m^{-3} and 27 mg P.m^{-3}, in Lagoa Azul and Lagoa Verde, in 1996/98 (INOVA 1998) and 15 mg P.m^{-3} and 21 mg P.m^{-3} in 2000/2002. According to OECD (ANON. 1982 in HARPER 1992), these results correspond to a mesotrophic situation. Nevertheless, levels as high as 43 mg P.m^{-3} and 97 mg P.m^{-3} were measured in some winter samples from Sete-Cidades lagoons.

In 1988/89 (SANTOS et al. 1992), water column chlorophyll *a* average concentrations were 2.59 mg.m^{-3} in Lagoa Azul and 6.02 mg.m^{-3} in Lagoa Verde. In 2000/2003, chlorophyll average values, raised to 6.65 mg.m^{-3} and 9.27 mg.m^{-3}, respectively, which are characteristics of meso-eutrophic aquatic environments, according to OECD boundary values for trophic categories. Chlorophyll maximum values in this period were 19.08 mg.m^{-3} in Lagoa Azul (July 2002) and 20.59 mg.m^{-3} in Lagoa Verde (November 2001).

From 2000 to 2003, Carlson's Trophic State Index (TSI) values, associated with parameters Secchi disc depth (transparency), total phosphorus and chlorophyll, varied from 37 to 60, but most of the results were above 45 (Fig. 4). During July 2002 bloom, Lagoa Azul TSI result associated to transparency reached 83, due to the accumulation of dense scums at lake surface. At the same period, the corresponding TSI value for chlorophyll was 60. Despite the TSI associated with total phosphorus was only 44 (oligo-mesotrophic conditions), these results and the observed shifts in taxonomic composition of phytoplankton presented further,

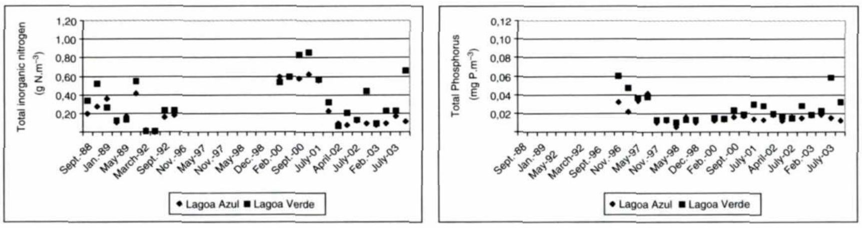

Fig. 2. Inorganic nitrogen and total phosphorus water column average concentration variation in Sete-Cidades lake from 1988 to 2003.

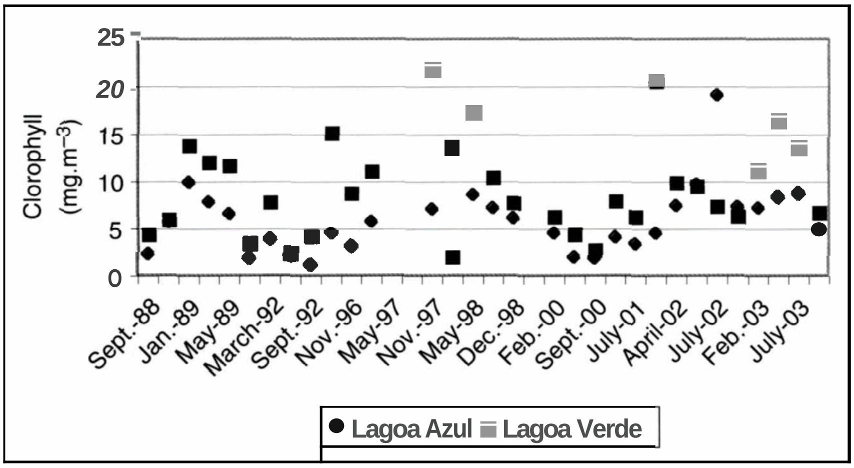

Fig. 3. Chlorophyll a concentration variation in Sete-Cidades lake from 1988 to 2003.

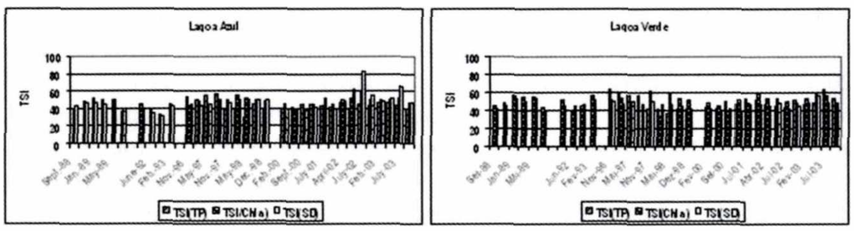

Fig. 4. Carlson's Trophic State Index, associated with Secchi disc depth (transparency), total phosphorus and chlorophyll in Sete-Cidades lake (1988/2003 periods).

show, with no doubts, that Sete-Cidades is now a meso-eutrophic lake and it's water quality is worse than ten years ago.

From 1988 to 1998 (SANTOS et al. 1992, INOVA 1996,1998), phytoplankton cell densities in these lagoons never exceeded 9 10^6 cells.l^{-1} (Fig. 5). In the

Table 1 – Phytoplankton density in Sete-Cidades lake (2000/2003) (10^6 cells . l^{-1})

	Feb. 00	**Set. 00**	**Dec. 00**	**July 01**	**Nov. 01**			
L. Azul	2.8	2.7	3.7	25.5	4.8			
L. Verde	15.7	12.5	11.5	11.5	45.8			
	Apr. 02	**May 02**	**July 02**	**Oct. 02**	**Feb. 03**	**May 03**	**July 03**	**Oct. 03**
L. Azul	45.2	93.9	312.8	2830.4	446.7	224.5	4842.7	1960.9
L. Verde	96.9	53.9	29.2	2828.3	17.0	190.8	370.9	19.8

1988/89 monitoring campaign, the maximum cell density (4.8 10^6 cells . l^{-1}) was achieved in a Lagoa Verde sample (May 1989), in which cyanobacteria represented only 59 % of total phytoplankton. Cell densities and cyanobacteria percentages at that period were normally greater in Lagoa Verde than in Lagoa Azul. The same happened in 1992/96 with smaller cell densities being observed in 1992 for both lagoons samples. In 1997/98, increasing cell densities began to be observed but cyanobacteria were dominant (67 % of total phytoplankton) only in one sample collected from Lagoa Verde in December 1998. There are no records of 1999 samplings.

Results from phytoplankton analysis in 2000/2003 period are presented in Table 1. The first bloom by scum forming cyanobacteria was noticed in Lagoa Verde. Very dense yellow scums formed, in July 2000, at the surface of this lagoon. In Lagoa Azul the bloom was not as perceptible as in Lagoa Verde. Phytoplankton analysis weren't performed during that event, but results from samples collected in February, September and December of 2000, in the same lagoon, showed that 86 % to 89 % of total phytoplankton was cyanobacteria. This means that this phytoplanktic group became dominant all year long. During the 2001 late spring and early summer, a similar bloom took place in Lagoa Azul, having been recorded 25.5 10^6 cells . l^{-1} (92 % of cyanobacteria) in a sample collected in July. Since that time until October 2003, the last sampling with results already available, the situation has been always worst in Lagoa Azul than in Lagoa Verde (Fig. 5). In fact, cyanobacteria began to account for Lagoa Azul phytoplankton population in percentages never less than 95 % (Fig. 6). The highest phytoplankton cell density in this lagoon (4842.7 10^6 cells . l^{-1}) was recorded in July 2003.

Cyanobacterial populations (Table 2) were almost always dominated by *Microcystis* sp., mainly *Microcystis aeruginosa* but, sometimes, also *Microcystis pulverea*. Other genera were less frequent but in some samples they reached densities higher than 10^6 cells . l^{-1}. That's the case of *Anabaena* sp. (L. Verde – Sept./Dec. 2000 and May 2003), *Aphanocapsa* sp. (L. Verde – Feb./May 2003), *Aphanotece* (L. Verde – May/July 2003), *Coelosphaerium* sp. (L. Azul – all 2002 samples; L. Verde – October 2002), *Pseudanabaena* sp. (L. Verde – Feb. 03), *Oscillatoria* sp. (L. Azul – November 2001) and *Woronichinia naegeliana* (L. Verde – Oct. 2003).

Bloom densities (> 20 10^6 cells . l^{-1}) were detected, in Lagoa Azul, after April 2002 and lasted for the rest of the sampling campaigns performed. In Lagoa Verde, blooms were first noticed in November 2001 (37.9 10^6 cells . l^{-1}), repeated

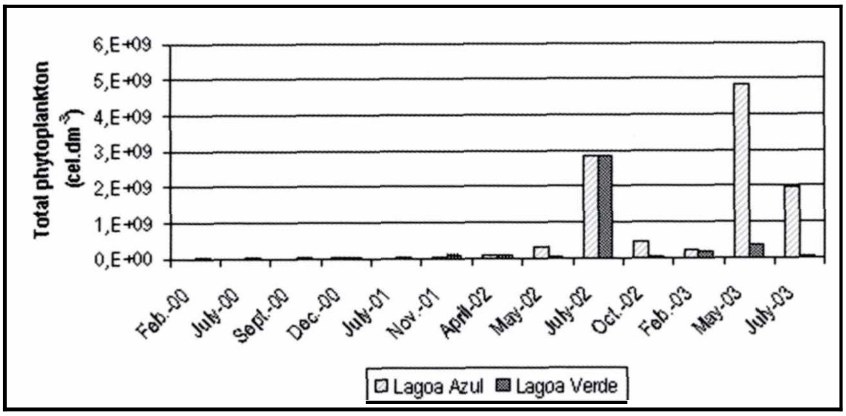

Fig. 5. Phytoplankton densities in Sete-Cidades lake (200012003)

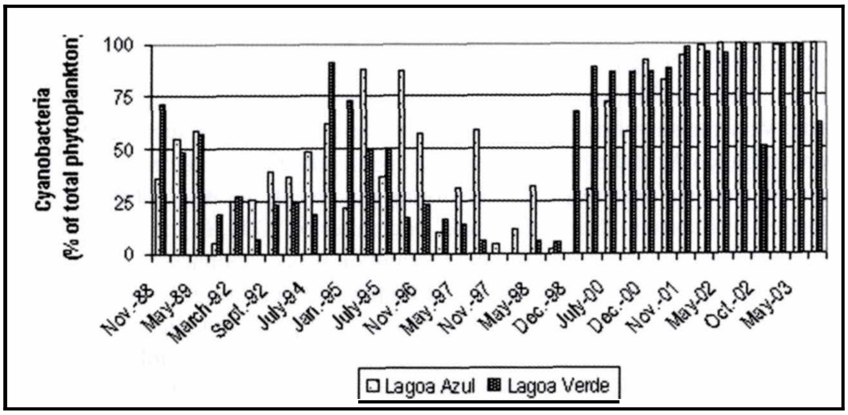

Fig. 6. Cyanobacteria percentages variation in Sete-Cidades lake (198812003)

themselves in ali 2002 campaigns and also during spring and summer 2003 samplings. Blooms were almost always dominated by *Microcystis* sp., except for July 2003, in Lagoa Azul. At that time *Aphanotece* sp. (3846.5 10^6 cells $\cdot l^{-1}$) was the most abundant, and *Aphanocapsa* sp. and *Coelosphaerium* sp. also attained bloom densities (880.3 10^6 cells $\cdot l^{-1}$ and 181.3 10^6 cells $\cdot l^{-1}$, respectively). Although blooms began to appear in Lagoa Verde, the highest cell counts were observed in Lagoa Azul.

Toxins in solution (filtered samples) were detected, in both lagoons samples collected in July and November 2001 (Lagoa Azul: 0.02–0.64 $mg \cdot m^{-3}$; Lagoa Verde: 0.05–0.45 $mg \cdot m^{-3}$) and in July and October 2002 (Lagoa Azul: 0.03–0.05 $mg \cdot m^{-3}$; Lagoa Verde: 0.01–0.24 $mg \cdot m^{-3}$). Results from 2003 samplings are not yet available. In Lagoa Azul soluble toxins were most frequently found at

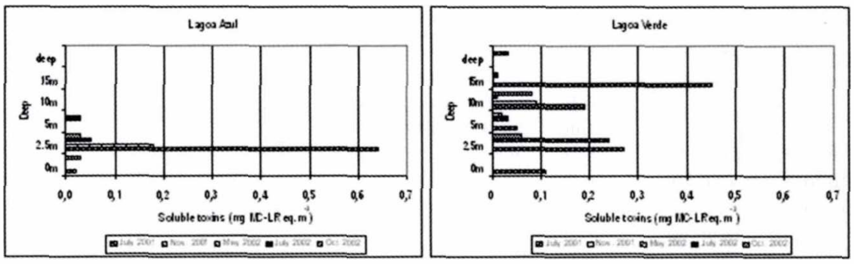

Fig. 7 Toxins in water column filtered samples of Lagoa Azul and Lagoa Verde (2001/2002).

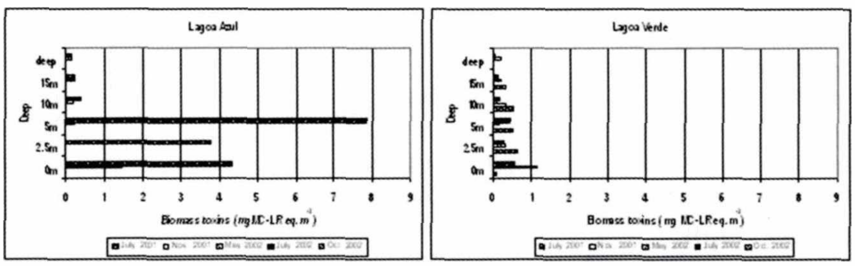

Fig. 8. Toxins in phytoplankton biomass samples of Lagoa Azul and Lagoa Verde (2001/2002).

surface and 2.5m deep. In Lagoa Verde, toxins were found mostly between 2.5 m and 10 m dephts, but twice also in surface and deep samples (Fig. 7).

Toxins were found in Lagoa Azul phytoplankton biomass samples collected in May and July 2002. Levels recorded ranged from 0.01 mg.m^{-3} to 7.83 mg.m^{-3}. In Lagoa Verde biomass, toxins were found in July and November 2001 and May and July 2002 samples. Maximum concentration recorded was 1.17 mg.m^{-3} (Fig. 8).

The highest levels were recorded in Lagoa Azul July 2002 samples. Toxins concentration of 2.5 m deep sample (3.78 mg.m^{-3}) corresponded to 3780 mg.kg^{-1} of dry biomass weight and 0.47 mg.mg^{-1} of chlorophyll a. The maximum level of 7.83 mg.m^{-3}, found in the 5m deep sample, corresponded to 1566 mg.kg^{-1} of dry biomass weight and 0.873 mg.mg^{-1} of chlorophyll *a*. These results are similar to those mentioned by CHORUS et al. (in CHORUS 2001) for bloom conditions (1000 to 6000 mg.kg^{-1} dry weight).

Discussion

The results obtained in the 2000/2003 period confirmed the Sete-Cidades lake tendency for eutrophication detected in previous monitoring programs. Average Trophic State Index (TSI) values for Lagoa Azul and Lagoa Verde were higher than 45 and near 50, which enable to classify this water body as meso-eutrophic.

Table 2. Cyanobacteria identification in Sete-Cidades lake (10^6 cells $\cdot l^{-1}$)

LAGOA AZUL

	Feb. 2000	Sept. 2000	Dec. 2000	July 2001	Nov. 2001
Anabaena spp.	0.51	0.08	0.21	0.71	0.71
Aphanizomenon sp.	0.26	0.07	0.20		
Aphanotece sp.			0.03	0.26	0.26
Coelosphaerium sp.		0.10	0.17		
Merismopedia sp.		0.22	0.01		
Microcystis spp.		1.26	0.98	14.4	14.4
Pseudanabaena spp.	0.10	0.10	0.52	8.18	3.34

	Apr. 2002	May 2002	July 2002	Oct. 2002	Feb. 2003	May 2003	July 2003	Oct. 2003
Anabaena spp.	0.21	0.62			0.45		0.02	0.01
Aphanizomenon sp.				0.01			0.31	0.52
Aphanocapsa sp.				0.79	8.20	37.6	880.3	0.24
Aphanotece sp.						0.01	3846.5	11.7
Coelosphaerium sp.				3.89	39.7		181.3	183.4
Lyngbya sp.				0.02	0.01	0.01	0.20	1.04
Micmcystis spp.	42.1	92.7	311.9	2825.1	395.1	184.9	811.7	1762.2
Pseudanabaena sp.	0.31		0.23	0.02	0.04		0.02	0.012

LAGOA VERDE

	Feb. 2000	Sept. 2000	Dec. 2000	Juiy 2001	Nov. 2001
Anabaena spp.	0.03	7.74	5.12	0.09	1.90
Aphanizomenon sp.			0.14		
Aphanotece sp.			0.20		
Coelosphaerium sp.		0.16			
Snowella sp.	0.03	0.42	0.26	0.38	0.38
Merismopedia sp.			0.03	0.04	
Microcystis spp.	5.84	5.66	4.99	9.39	37.9
Pseudanabaena spp.	0.26			0.01	0.18

	Apr. 2002	May 2002	Juiy 2002	Oct. 2002	Feb. 2003	May 2003	July 2003	Oct. 2003
Anabaena spp.		0.24		0.01	0.13	2.56	0.05	0.06
Aphanizomenon sp.							0.05	0.28
Aphanocapsa sp.				0.04	5.39	8.24	0.38	0.60
Aphanotece sp.						15.6	5.51	
Coelosphaerium sp.				2.28	0.62	2.37	4.62	
Lyngbya sp.				0.01				
Microcystis spp.	95.2	51.5	27.6	2818.5	2.42	159.4	357.3	6.62
Pseudanabaena sp.					8.74			
Oscillatoria spp.		0.09	0.05	0.01				0.02
Womnichinia naegeliana								4.63

Although average total phosphorus concentration inside the lake was not very high, concentrations found in winter samples proved that there is an important runoff of fertilised water from the watershed. Internal recycling, resulting from thermal stratification and sediment anoxia, also contributed with phosphorus to the water column. Most of the time nitrogen concentrations were also non-limiting for phytoplankton growth, being the ammonium form the most abundant.

Water temperatures, ranging from 12–13 °C in winter and 22–23 °C in summer, low dissolved inorganic carbon (Sete-Cidades is a softwater lake), available phosphorus and ammonium nitrogen were optimal conditions for cyanobacteria development. Blooms dominated by colonial cyanobacteria (*Microcystis* spp.) began in 2000 and the events repeated in 2001 and 2002, with higher populations in spring and summer but with a tendency to last all year long. Species of *Microcystis* genera (*M. aeruginosa* and *M. pulverea*), non-nitrogen fixers and gas-vacuolate cyanobacteria, formed dense yellow scums at lake surface when calm weather conditions prevailed. From 2002 to 2003 Lagoa Azul cyanobacteria population shift, picocyanobacteria non-bloom forming (*Aphanotece* sp., *Aphanocapsa* sp. and *Coelosphaerium* sp.) became dominant and yellow scums were less intense.

High levels of cyanotoxins were found in 2002 samples, reaching 7.83 $mg \cdot m^{-3}$ in a Lagoa Azul sample. Sete-Cidades lake water is not used for human consumption but recreational use, like swimming and wind-surfing are common practices. Advisory signs were posted at Sete Cidades lake edges to warn people of the blooms toxicity and their associated health dangers.

Actions are being taken to revert eutrophication, namely the conversion of agricultural watersheds activities to others less pollutants. Limiting nutrient actions determined in the Sete Cidades Watershed Land Management Plan are expected to reduce the external loadings of nutrients benefiting water quality in general, as well as control phytoplankton productivity and ultimately prevent the occurrence of cyanobacteria blooms.

Acknowledgments

This research was supported by the Environment Secretary of Azores Regional Government and was performed with the contribution of the Environmental Science Department of the New University of Lisbon.

The authors thank Prof. António Rodrigues, algae specialist from the New University of Lisbon, and Dr. Maria Isabel Andrade, expert from the Portuguese Environment Institute, for phytoplankton analysis.

References

Barrois T. H. (1896): Faune des eaux douces des Açores. – 172 pp., Lille.

Carlson R. E. (1977) A trophic state index for lakes. – Limnol. Oceanogr. **22**: 361–9

Cooke G. D., Welch E. B., Peterson S. A. & Newroth P. R. (1993) Restoration and management of lakes and reservoirs, 2nd Ed. – 548 pp., Lewis Publishers, CRC Press, Boca Raton, Florida, USA.

INOVA (1996): Análise das águas das lagoas da Região autónoma dos Açores. – 547 pp., Technical Report, S. Miguel – Açores.

INOVA (1998): Análise das águas das lagoas da região autónoma dos Açores. – 580 pp., Technical Report, S. Miguel – Açores.

LAWTON L. A., EDWARDS C. & CODD G. A. (1994): Extraction and high-performance liquid chromatographic method for the determination of microcystins in raw and treated waters. – Analyst 119: 1525–1530.

LUND J. W. G., KIPLING C. & LE CREN E. D. (1958): The inverted microscope method of estimating algal numbers and the statistical basis of estimation by counting. – Hydrobiologia 11: 143–170.

MADSEN J. D. (1993): Biomass techniques for monitoring and assessing control of aquatic vegetation. – Lake and Reservoir Management 7 (2): 141–154.

OLIVEIRA M. R. L. (1989): Estrutura das comunidades de fitoplâncton nas lagoas das Sete-Cidades. Technical and Scientific Report. – 27p. INIP, Lisboa

PACHECO D.M., MACEDO M., GOULART M. E, SANTOS M. C. R., RODRIGUES A. M. F. & SANTANA F. (1998): Aquatic plants in Sete Cidades Lake (S. Miguel Island – Azores): An overview and attemptive management Proceedings of the 10th. EWRS International Symposium on Aquatic Weeds, Lisbon, 21–25 September.

PORTEIRO J. & CALADO H. (1998): Plano de ordenamento da bacia hidrográfica da lagoa das Sete-Cidades. 1ª Fase, Estudos de caracterização – Vol II, 174 pp.. Universidade dos Açores, Departamento de Biologia, Secção de Geografia, Ponta Delgada, Açores

SANTOS M. C. R., RODRIGUES A. M. F.. SOBRAL M. P. & SANTANA F. J. P (1992): A eutrofização de meios lacustres. Lagoas das Sete – Cidades e Lagoa das Fumas. – In: PIRES A. C., PIO C., BÓIA C. & NOGUEIRA T. (eds), Procc. 3ª Conferência nacional sobre a qualidade do ambiente, Comissão de Coordenação da Região Centro, Coimbra, p. 217–228.

SANTOS M. C. R., SANTANA F. J. P., RODRIGUES A. M. F. & SOBRAL, M. P.(1991): Controlo da eutrofização das lagoas de S. Miguel (Açores). Parte I – As Lagoas das Sete – Cidades – 136 pp., Technical Report, D. C. E. A.. Universidade Nova de Lisboa.

SMITH, V. H. (1983): Low nitrogen to phosphorus ratio favour dominance by bluegreen algae in lake phytoplankton. – Science 221: 669–671.

Standard Methods for the Examination of Water and Wastewater (1998) 20th Ed., APHA – AWWA – WEF, USA

WHITTON, B. A. & POTTS M. (eds)(2000): The Ecology of Cyanobacteria. Their diversity in time and space. – 669 pp., Kluwer Academic Publishers, Netherlands.

The authors' addresses:

Assist. Prof. MARIA DA CONCEIÇÃO RAIMUNDO SANTOS
Assoc. Prof. FERNANDO SANTANA
Dr. HELENA MUELLE
New University of Lisbon
Faculty of Sciences and Technology
Department of Environmental Sciences and Engineering
Quinta da Torre
P-2829-516 Caparica, Portugal
mcrs@fct.unl.pt

Dr. DINA MARIA MEDEIROS PACHECO
Azores Government Environmental Secretary
Avenida Antero de Quental
Edifício dos CTT 2° A
P-9 500-160 Ponta Delgada, Portugal
Dina.MD.Pacheco@azores.gov.pt