Influences of a *Microcystis aeruginosa* Kützing bloom on zooplankton populations in Jacarepaguaí Lagoon (Rio de Janeiro, Brazil)

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Abstract

Jacarepaguaí Lagoon is a shallow, hypereutrophic, coastal lagoon located in Rio de Janeiro (RJ, Brazil), with recurrent blooms of cyanobacteria. This study was carried out with the aim to detect the effects of the cyanobacterium *Microcystis aeruginosa* on zooplankton populations (especially cladocerans) in the lagoon. At two sampling stations we measured temperature, pH, salinity, dissolved oxygen and transparency, and collected water samples for chemical analyses of particulate organic carbon (POC), chlorophyll-a and toxins (microcystins), and plankton samples for phytoplankton and zooplankton analyses, from August 1996 to September 1997. Laboratory experiments were also performed to test for toxicity of both natural assemblages and cultured *Microcystis*. The results showed that temperature, salinity and cyanobacteria biomass were the best descriptors of the population dynamics of Cladocera. In spite of high levels of microcystins in seston, toxins were not generally correlated with the density of Cladocera. Laboratory experiments, however, showed strong evidence of the toxicity of seston to Cladocera. Also, high levels of toxins in seston were associated with a collapse of cladoceran populations in the lagoon, suggesting toxic effects of mycrocystins on these organisms. Rotifers and copepods were less affected by cyanobacteria, maintaining high densities in the lagoon throughout the bloom. We conclude that blooms of *Microcystis aeruginosa* are potentially harmful for cladoceran populations in nature.

Key words: Cyanobacteria – tropical zooplankton – *Microcystis* – microcystins – toxic effects

Introduction

Most of the studies on cyanobacteria–zooplankton interactions have focused on the effects of toxic strains of cyanobacteria grown under controlled laboratory conditions, specially on temperate large zooplankton like Daphnia (Nizan et al. 1986; Lampert 1982; Fulton & Paerl 1987a; DeMott et al. 1991; Reinikainen et al. 1994; Hietala et al. 1995; Smith & Gilbert 1995; Rohrlack et al. 1999). However, there is a lack of data regarding the effects of cyanobacteria on tropical zooplankton communities, especially that ones composed of smaller species (Ferrão-Filho et al. 2000).

Studies on cyanobacteria–zooplankton interactions are relevant because of the harmful potential of cyanobacteria in aquatic communities, but up to date no study has clearly made the link between laboratory and field data (Burns 1987; Haney 1987; Lampert 1987; Benndorf...
& Henning 1989). Most of the work in this area has focused on descriptive approaches of phytoplankton and zooplankton communities and the possible effects of the appearance of cyanobacteria during the seasonal cycle. Besides this, quite a few studies have tested the effects of natural occurring populations of cyanobacteria on zooplankton and tried to correlate these data with the effects observed for laboratory cultures of cyanobacteria (Matveev & Balseiro 1990; Fulton & Jones 1991; Hanazato 1991; Nandini 2000; Ferrão-Filho & Azevedo, in press).

Here, we compare the data of a study on the phytoplankton and zooplankton communities of a tropical coastal lagoon with laboratory data regarding the effects of cultured and natural populations of the cyanobacterium *Microcystis aeruginosa* Kützing on cladocerans from the same lagoon. We consider several environmental factors such as food availability, colonial morphology and toxin concentration in the seston as responsible for the results observed in the field and relate them to laboratory experiments.

**Study site**

Jacarepaguá Lagoon is located in the South coast of Rio de Janeiro State, in the metropolitan zone of Rio de Janeiro City (Fig. 1), and has been investigated with respect to the consequences of cyanobacterial blooms on its aquatic community (Magalhães & Azevedo 1998; Ferrão-Filho et al. 2002). It is an oligohaline, shallow lagoon (*z*<sub>ave</sub> = 1 m), connected with other two lagoons and with the Atlantic Ocean by means of a straight channel. This system has received massive discharges from the surrounding urban areas, including domestic and industrial sewage and has become hypereutrophic. Recurrent blooms of *Microcystis aeruginosa* have been reported in this lagoon, with known toxicity to cladocerans (Ferrão-Filho et al. 2000). A year of intensive sampling was carried out in this lagoon from August 1996 through September 1997, at two sampling stations (Fig. 1). Sampling included phytoplankton, zooplankton and water for chlorophyll-α, particulate organic carbon and toxin (microcystin) analysis and measurements of temperature, pH, dissolved oxygen, transparency and salinity.

**Methods**

**Sampling and analysis**

Phytoplankton was collected at biweekly intervals at the subsurface with an amber glass flask and preserved with acid Lugol’s solution. The fixed and live samples were examined with a Light Microscope. The densities were estimated as proposed by Utzschneider (1958) and the biomass was calculated following Edler (1979), assuming a specific density of phytoplankton cells of 1 g · cm<sup>−3</sup>. Zooplankton was collected monthly, with a Van Dorn horizontal bottle (3 x 3 liters) at 0.5 m depth, concentrated in a 68 μm mesh size plankton net and preserved with a 4% formalin + borax (10%) solution. Five liters of surface water from the lagoon were collected and stored in ice and taken to the laboratory between 2 and 4 hours later for processing. For chlorophyll-α analysis, 100–200 ml subsamples were filtered onto glass fiber filters (Sartorius AG 37070, Goettingen, Germany), immediately extracted with methanol for 3 hours, and extract absorbance measured at 665 and 750 nm after centrifugation (Lorenzen 1967). For particulate organic carbon (POC) analysis, 100–250 ml of water were passed through a 200 μm net to remove large zooplankton, filtered onto glass-fiber filters, and dried overnight at 60 °C. POC was determined with a dichromate-sulfuric acid oxidative method following Strickland & Parsons (1972). Microcystin content was determined in seston samples obtained from 2 liters of water filtered onto glass-fiber filters, which freeze-dried prior to the analysis. Toxin (microcystins) extraction was performed with a butanol:methanol:H<sub>2</sub>O solution (5:20:75 v/v) following a method modified from Krishna-Murthy et al. (1986). The analysis and quantification of microcystins was carried out by HPLC technique in a Shimadzu chromatographic apparatus with a diode array UV/Vis. SPDA-M10A [see Ferrão-Filho et al. (2000) for details].

Temperature, pH, dissolved oxygen and salinity were measured at the surface using field instruments. Transparency was measured using a Secchi disc. Significant relationships among biotic and abiotic variables were analyzed by Pearson correlations pooling the field data from both stations. In order to increase the power to detect significant effects, the correlation tables were corrected for multiple testing using the sequential Bonferroni technique (Rice 1989).

**Laboratory experiments**

Acute toxicity, life-table and growth experiments were conducted at the laboratory with cladocerans isolated from the lagoon and using both cultured and natural occurring populations of *Microcystis aeruginosa*. The two cladocerans occurring in the lagoon, Ceriodaphnia cornuta Sars and Moina micrura KURS, were cultured in the lab using filtered-autoclaved lagoon water. Animals were fed with the green algae Ankistrodesmus falcatus (Braun) or Chlamydomonas reinhardtii Dangeard at a total concentration of 1.0 mg C · l<sup>−1</sup>. Green algae used as food were cultured in MBL medium (Stemberger 1981) at 40 μE · m<sup>−2</sup> · s<sup>−1</sup>, 23 °C and 12/12 hours (light/dark) photoperiod.
Four acute toxicity experiments were performed using the two cladocerans and both cultured and naturally occurring populations of *Microcystis*. Cultures of the *Microcystis* strain NPLJ-2, isolated from the lagoon, were maintained in the laboratory with ASM-1 medium (GORHAM et al. 1964), at the same light intensity, temperature and photoperiod as green algae. For these experiments, exponential growing cells of laboratory cultured *Microcystis* were concentrated in a centrifuge (4000 g x 10 min.) and resuspended in the ASM-1 medium. Seston samples containing natural populations of *Microcystis* were obtained by centrifuging lagoon water and resuspending the seston in filtered-autoclaved lagoon water. These samples were kept at 4 °C during the experiments. Concentrations of both laboratory cultures and seston used in the acute experiments ranged from 0.25 to 1.0 mg C·l⁻¹, with the concentrations lower than 1.0 mg C·l⁻¹ ("100%") having an addition of the green algae used as food, so that the final concentration of all food suspensions totalled 1.0 mg C·l⁻¹. Controls consisted of animals both starved and fed only with green algae ("Food"). Ten newborns less than 24 h old of each...

Fig. 1. Localization of Jacarepaguá Lagoon in Brazil and in the State of Rio de Janeiro (small rectangle). The sampling stations are indicated by numbers (1 and 2).
cladoceran species were placed in glass flat-bottom tubes with 30 ml of each food suspension with four replicate tubes per treatment. Every day, during the five days of the experiment, animals were transferred to new algal and seston suspensions and the survivors were counted. For each treatment, the median lethal time (LT₅₀), in hours, was estimated by PROBIT regression analysis (SPSS Statistical Package, SPSS Inc., Chicago, IL, USA) and these values were statistically compared by ANOVA.

One life-table experiment was performed in order to verify the effects of both cultured and naturally occurring Microcystis on the reproduction of cladocerans. In this experiment, newborns less than 24 h old of each cladoceran species were placed individually in glass flat-bottom tubes with 30 ml of each food suspension with 16 replicate tubes per treatment. Every day, survivors were transferred to new food suspensions and checked for the appearance of eggs and newborns. Treatments included cultured Microcystis and seston samples collected on 20 March 1997 from station 2 and processed in the same way as in the acute toxicity experiments. There was only one concentration of seston (100% = 1.0 mg C • 1⁻¹) and only one concentration of cultured Microcystis (10% = 0.10 mg C • 1⁻¹). In this treatment Microcystis was mixed with the green algae Ankistrodesmus (90% = 0.90 mg C • 1⁻¹). Ankistrodesmus was also used as control. Population parameters calculated were the age at first reproduction, mean clutch size, and total offspring. The intrinsic rate of natural increase (r) was estimated by bootstrap technique (Taberner et al. 1993), with 500 replicates per run and bias adjusted correction for small cohorts (Meyer et al. 1986). Population parameters were compared among treatments by ANOVA and Tukey multiple comparison test. Statistical differences between r values in each treatment were tested by t-tests.

Juvenile growth rate for Moina micrura was estimated on the same cohort of animals used in the life-table experiment. Fifty < 24 h newborns were placed in 500 ml bottles filled with food suspensions and with three replicate bottles per treatment. Treatments were the same as in the life-table experiment. Initially and after 2, 4 and 6 days, a group of 5 to 10 animals was taken from each treatment and placed in pre-tared aluminum foil containers and dried overnight in 60 °C. These containers were then weighed in a microbalance (Mettler Toledo UMT-2; GmbH, Greifensee, Switzerland) with a precision of 0.1 µg. Growth rates were calculated using the equation:

$$ GR = \frac{\ln M_t - \ln M_i}{t}, $$

where M₀ and Mₜ are mean individual mass initially and after t days. Treatments were compared by ANOVA and Tukey multiple comparison test (P < 0.05).

**Results**

**Field data**

Temperature varied between 20 to 33.6 °C during the study, with minimum temperatures occurring in June and maximum temperatures in January (Fig. 2a). Salinity varied from 0.0 to 10.0%, with lower values occurring in the rainy season (December to March; Fig. 2b). The pH varied between 6.2 and 9.3, with no defined seasonal pattern (Fig. 2c), but lower values at station 1 were probably associated to the influence of Marinho River. Dissolved oxygen (D.O.) varied highly during the study, reaching values close to anoxia (0.5 mg • 1⁻¹) and as high as 18.5 mg 1⁻¹ (Fig. 2d). Transparency (Secchi depth) was low in the lagoon, varying from only 10 to 60 cm, with higher values occurring in the wet-warm season (January to April; Fig. 2e). Particulate organic carbon (POC) values were also very high, varying from 2.7 to 38.1 mg • 1⁻¹ (Fig. 2f). Chlorophyll-a concentrations were characteristic of hypereutrophic systems (Vollenweider 1982), ranging from 65.2 to 304.3 µg • 1⁻¹, with higher values occurring from the end of spring (November) through the rainy season and winter of 1997 (Fig. 2g). Microcystin concentrations in seston varied from undetectable values at the beginning of the study to more than 900 µg 1⁻¹ at station 2 at the end of the sampling period (Fig. 2h).

Seasonal changes in phytoplankton composition in the lagoon suggest three distinct periods (Fig. 3). From August to December 1996, the phytoplankton community was dominated by Chlorophyceae and Bacillariophyceae. Chlorophyceae were represented mostly by Chlorococcales, including Ankistrodesmus, Coelastrum, Crucigenia, Monoraphidium, Oocystis, Scenedesmus and Tetrastrum, which were in the edible size range for zooplankton. Bacillariophyceae were represented mostly by centric diatoms (Cyclotella sp.). From late December 1996 to early January 1997, phytoplankton composition changed dramatically to a community dominated by cyanobacteria (Cyanophyceae). This group was represented mostly by Microcystis aeruginosa, which reached very high biomass in December and comprised 88% and 95% of the phytoplankton at stations 1 and 2, respectively. January to April were marked by rapid changes in the phytoplankton community, shifting in dominance between cyanobacteria and the other groups. The final five months of the study (May to September) were marked by the complete dominance of cyanobacteria, especially M. aeruginosa (72% to 80%) and Aphanizomenon sp. (9% to 11%). During this bloom, Microcystis presented both unicellular and colonial forms, with an increase in colonial forms from the beginning to the end of the bloom. Most of these colonies were composed by hundreds or thousands of cells, reaching sizes above the edibility range for zooplankton (> 50 µm).
Zooplankton was represented by seven species of rotifers (Brachionus angularis GOSSE, B. calyciflorus PALLAS, B. plicatilis, Filinia sp., Hexarthra oxyuris SERNOV, Monomata sp. and Polyarthra sp.), two cladocerans (Ceriodaphnia cornuta SARS and Moina micrura KURS) and one copepod (Metacyclops mendocinus WIERZBICKI). Rotifers and copepods were quantitatively more abundant than cladocerans, dominating the zooplankton community throughout all the study (Fig. 4). The dominant rotifers B. angularis and B. plicatilis reached peak densities in January and February at station 1, respectively, and in February at station 2. From March to June, B. plicatilis was dominant over B. angularis at station 1, whereas both rotifers showed similar densities at station 2. In August, density of rotifers peaked again, with B. angularis as the dominant species and reaching 4282 ind. • 1 • 1 • 1 • 1 at station 1 and 10617 ind. • 1 • 1 • 1 • 1 at station 2. Cladocerans occurred only from August to May, declining gradually from March until undetectable densities in June. The dominant cladoceran, M. micrura, reached maximum densities in January, with more than 350 and 400 ind. • 1 • 1 • 1 • 1 at station 1 and 2, respectively, while C. cornuta reached maximum densities in April, with 100 and 70 ind. • 1 • 1 • 1 • 1 at station 1 and 2, respectively. M. micrura populations appeared to collapse twice during the study, once in November and again in February, followed by a rapid recovery of the initial densities only at station 1. At station 2, cladocerans did not recover the same densities after the February breakdown. Copepods had a more homogeneous distribution along the year, with peak densities in August.

**Correlation between the field data**

Table 1 shows the correlation coefficients between POC, chlorophyll-α and toxin and the biomass of phytoplank-
ton during the study. There were no significant correlations between POC or chlorophyll-a with the main phytoplankton groups in the lagoon. There was, however, a strong significant correlation between the toxin concentrations in seston and the biomass of Cyanophyceae. There was also a strong correlation between the total phytoplankton biomass and toxins, which was probably driven by the high biomass contribution of cyanobacteria.

Table 2 shows the correlation coefficients between zooplankton densities and the physical and chemical variables. There was a strong positive correlation of *M. micrura* density with temperature. There were also positive correlations of *C. cornuta* and *Polyarthra* sp. densities with transparency (Secchi depth), which is probably a consequence of the increase in the clearance rates of cladocerans and rotifers in summer. There were, however, no significant correlations between zooplankton and chlorophyll-a or POC in the lagoon. A strong negative correlation was observed between *M. micrura* density and salinity, while a positive correlation was observed between the density of copepods and salinity. There were no significant correlations between the density of cladocerans and toxin concentrations in the seston of the lagoon. Surprisingly, there was a strong and positive correlation between copepods and toxins as well as between *H. oxyuris* and toxins. These positive correlations were probably driven by the positive correlations between these zooplankton groups and the biomass of cyanobacteria (Table 3).

Table 3 shows the correlation coefficients between zooplankton densities and the biomass of the main phytoplankton groups during the study. The density of *M. micrura* was negatively correlated with cyanobacteriological biomass, whereas the density of copepods and the rotifer *H. oxyuris* were positively correlated with the biomass of cyanobacteria.

**Laboratory experiments**

Acute toxicity by seston was evident in December at station 2, and in February at station 1 and 2 (data for station 2 not shown; Fig. 5 and 6). In both periods, however, only *M. micrura* was affected. In December, microcystin concentration in seston varied from 2.6 µg · L⁻¹ (on 11th) to 2.0 µg · L⁻¹ (on 23rd), and animals fed seston showed a

![Station 1](image1)

![Station 2](image2)

*Fig. 3. Phytoplankton composition (as % biovolume) in both stations of Jacarepaguá Lagoon from August 1996 to September 1997.*
progressive decline in survivorship from the third day of the experiment (Fig. 5). The ANOVA revealed significant differences in the LT50 between the seston and controls with food (F3,48 = 32.1, P = 0.001) and without food (F3,48 = 32.1, P = 0.032). Animals died 1.4 to 1.7 times faster in seston treatments than in controls with food at the concentrations of 0.5 and 1.0 mg C \cdot l^{-1}, respectively. In late January, no toxin was detected in seston at station 2, and no cladoceran was affected by seston. In February, toxin concentration in seston was 14.0 \mu g \cdot l^{-1} and \textit{M. micrura} showed increased mortality from the second day of the experiment. There was a significant difference in LT50 between seston and the controls with food (F3,58 = 84.1, P < 0.001), and animals died 2.5 to 3.6 times faster in the seston treatments than in the control with food. Survivorship in the controls without food in this

Table 1. Pearson correlation coefficients between POC, chlorophyll-\textit{a} and toxin contents and the biomass of the main phytoplankton groups (*P < 0.05). n = sample size.

<table>
<thead>
<tr>
<th>Factors</th>
<th>Phytoplankton groups</th>
</tr>
</thead>
<tbody>
<tr>
<td>POC (n = 50)</td>
<td>0.228</td>
</tr>
<tr>
<td>Chl-\textit{a} (n = 42)</td>
<td>0.005</td>
</tr>
<tr>
<td>Toxin (n = 50)</td>
<td>0.890*</td>
</tr>
</tbody>
</table>

Fig. 4. Zooplankton densities in both sampling stations of Jacarepaguá Lagoon from August 1996 to September 1997.
Table 2. Pearson correlation coefficients between zooplankton density and physical and chemical variables (*P < 0.05).

<table>
<thead>
<tr>
<th>Species</th>
<th>Variables</th>
<th>Temperat. (n = 30)</th>
<th>pH (n = 30)</th>
<th>D.O. (n = 30)</th>
<th>Secchi depth (n = 30)</th>
<th>Chl-a (n = 30)</th>
<th>POC (n = 28)</th>
<th>Salinity (n = 30)</th>
<th>Toxin (n = 25)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Cladocera</strong></td>
<td>M. micrura</td>
<td>0.796*</td>
<td>-0.451</td>
<td>-0.211</td>
<td>0.521</td>
<td>-0.070</td>
<td>0.323</td>
<td>-0.707*</td>
<td>-0.262</td>
</tr>
<tr>
<td></td>
<td>C. cornuta</td>
<td>0.205</td>
<td>-0.301</td>
<td>-0.138</td>
<td>0.691*</td>
<td>-0.003</td>
<td>-0.155</td>
<td>-0.117</td>
<td>-0.122</td>
</tr>
<tr>
<td><strong>Copepoda</strong></td>
<td>Nauplii</td>
<td>-0.003</td>
<td>0.174</td>
<td>0.317</td>
<td>-0.243</td>
<td>-0.230</td>
<td>-0.270</td>
<td>0.554</td>
<td>0.454</td>
</tr>
<tr>
<td></td>
<td>Copepodites</td>
<td>-0.043</td>
<td>0.222</td>
<td>0.212</td>
<td>-0.459</td>
<td>-0.218</td>
<td>-0.250</td>
<td>0.610*</td>
<td>0.709*</td>
</tr>
<tr>
<td><strong>Rotifera</strong></td>
<td>B. plicatilis</td>
<td>0.375</td>
<td>-0.013</td>
<td>-0.184</td>
<td>0.203</td>
<td>0.251</td>
<td>0.175</td>
<td>-0.102</td>
<td>0.552</td>
</tr>
<tr>
<td></td>
<td>B. angularis</td>
<td>0.324</td>
<td>-0.123</td>
<td>0.218</td>
<td>0.200</td>
<td>-0.101</td>
<td>-0.244</td>
<td>0.255</td>
<td>0.528</td>
</tr>
<tr>
<td></td>
<td>B. calyciflorus</td>
<td>0.547</td>
<td>-0.215</td>
<td>-0.134</td>
<td>0.218</td>
<td>0.215</td>
<td>-0.154</td>
<td>-0.365</td>
<td>-0.072</td>
</tr>
<tr>
<td></td>
<td>Polyarthra sp.</td>
<td>0.209</td>
<td>-0.144</td>
<td>-0.174</td>
<td>0.611*</td>
<td>0.020</td>
<td>-0.161</td>
<td>-0.199</td>
<td>-0.150</td>
</tr>
<tr>
<td></td>
<td>H. oxyuris</td>
<td>0.001</td>
<td>0.216</td>
<td>0.465</td>
<td>-0.355</td>
<td>-0.227</td>
<td>-0.195</td>
<td>0.396</td>
<td>0.763*</td>
</tr>
</tbody>
</table>

Table 3. Pearson correlation coefficients between zooplankton density and the biomass of the main phytoplankton groups (*P < 0.05).

<table>
<thead>
<tr>
<th>Species</th>
<th>Phytoplankton groups</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cyanoph. (n = 30)</td>
</tr>
<tr>
<td><strong>Cladocera</strong></td>
<td>M. micrura</td>
</tr>
<tr>
<td></td>
<td>C. cornuta</td>
</tr>
<tr>
<td><strong>Copepoda</strong></td>
<td>Nauplii</td>
</tr>
<tr>
<td></td>
<td>Copepodites</td>
</tr>
<tr>
<td><strong>Rotifera</strong></td>
<td>B. plicatilis</td>
</tr>
<tr>
<td></td>
<td>B. angularis</td>
</tr>
<tr>
<td></td>
<td>B. calyciflorus</td>
</tr>
<tr>
<td></td>
<td>Polyarthra sp.</td>
</tr>
<tr>
<td></td>
<td>H. oxyuris</td>
</tr>
</tbody>
</table>

period was about the same as in seston treatments (LT50 = 63 h for starved and 73 h for the higher concentration of seston). The treatments with mixtures of laboratory cultured *Microcystis* and green algae caused similar effects to *M. micruca*, which died 1.7 to 2.1 times faster in the mixtures than in controls with only good food in December (F3,48 = 32.1, P < 0.001), and 3.0 to 6.5 times faster in the mixtures in February (F4,70 = 84.1, P < 0.001). Both treatments with seston and cultured *Microcystis* showed a concentration-dependent effect in *M. micrurca*, which is typical of an acute intoxication. *C. cornuta*, however, was not significantly affected by strain NPLJ-2, showing good survivorship in the *Microcystis* treatments (Fig. 6).

Cladocerans showed also different response patterns in the life-table experiment (Fig. 7). While *C. cornuta* was little or positively affected by seston and *Microcystis* treatments, *M. micrurca* had a negative reproductive response in seston and in the cultured *Microcystis*. In the seston treatment, *C. cornuta* reproduced significantly earlier than in the control with green algae (F3,48 = 10.4, P < 0.001). In both seston and *Microcystis* treatments, *C. cornuta* had a significant increase in fecundity (F2,76 = 36.4, P < 0.001) and in total offspring number (F2,76 = 9.8, P < 0.001) relative to controls, and a significant increase in the intrinsic rate of natural increase (r) only in seston (t-test; P < 0.001). On the other hand, *M. micrura* had a significant reduction in fecundity in the seston treatment and in the treatment with the cultured *Microcystis* (F2,29 = 14.4, P < 0.001). However, age at first reproduction (F2,29 = 2.6, P = 0.095), total offspring number (F2,29 = 2.7, P = 0.084) and r-values (t-test; P = 0.591) of this species fed with seston were not significantly different from the control with green algae. The cultured *Microcystis*, however, decreased significantly the r-value of *M. micrura* compared to control (t-test; P = 0.028). The microcystin concentration in the seston treatment in this experiment was 2.4 µg · l⁻¹, a value close to the nominal concentration of this toxin in the cultured *Microcystis* treatment (2.3 µg · l⁻¹).

In the growth experiment, animals showed exponential growth in all treatments until the fourth day and a decrease in growth rates after the fourth day, associated with allocation of energy to reproduction (Fig. 8). Therefore, we compared statistically the treatments only for four days of growth. Animals in the control group
showed a higher growth rate than the other treatments, whereas there were any significant differences between the seston and the Microcystis treatments (Table 4).

Discussion

As shown in the correlation analyses, temperature, salinity and the bloom of cyanobacteria seem to be the main driving factors of zooplankton populations in the lagoon. While temperature had a positive effect on zooplankton (especially cladocerans), salinity and cyanobacteria had a negative impact on the cladoceran populations.

As temperature increased from spring to summer (August–December), there was an increase in cladoceran populations, probably caused by the increase in food availability. The effects of temperature on primary metabolism and reproduction rates of zooplankton are well known (Marsh 1973; Gophen 1976; Lampert 1977; MacArthur & Baille 1979). Also, temperature can affect the response of zooplankton to toxic cyanobacteria (Threlkeld 1985; Gilbert 1996a; Clark & Gilbert 1998). However, minimum temperatures

Table 4. Results of one-way ANOVA for the growth experiment with Moina micrura. Data are mean growth rates (d⁻¹) ± SE for the first 2 and 4 days of the experiment. There were three replicates per treatment, which included (1) Ankistrodesmus (control), (2) Microcystis mixed with Ankistrodesmus in a proportion of 10% of the total food concentration of 1.0 mg C ⋅ l⁻¹ and (3) lake seston. df = degrees of freedom.

<table>
<thead>
<tr>
<th>Days</th>
<th>Treatments</th>
<th>F-ratio</th>
<th>df</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ankistrodesmus</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0–2nd day</td>
<td>0.383 ± 0.036</td>
<td>0.213 ± 0.017</td>
<td>0.138 ± 0.043</td>
<td>14.0</td>
</tr>
<tr>
<td>0–4th day</td>
<td>0.381 ± 0.016</td>
<td>0.239 ± 0.008</td>
<td>0.233 ± 0.028</td>
<td>17.3</td>
</tr>
</tbody>
</table>

Fig. 5. Acute toxicity experiments with Moina micrura. The three treatments are indicated above each column. (1) Controls consisted of animals fed with green algae (A. falcatus) and starved animals. (2) Mixtures of seston and green algae (25% and 50%) and seston alone (100%) in a total concentration of 1.0 mg C ⋅ l⁻¹. (3) Microcystis strain NPLJ-2 was mixed with green algae in two proportions (25% and 50%) at a total concentration of 1.0 mg C ⋅ l⁻¹.
Fig. 6. Acute toxicity experiments with Ceriodaphnia cornuta. Experimental conditions were the same as for M. micrura in Fig. 5, except that December experiments were done on another date.

Fig. 7. Life-table experiment with Ceriodaphnia cornuta and Moina micrura. Treatments included (1) a control with the green alga Ankistrodesmus (Ank), (2) a mixture of 10% toxic Microcystis (Micro) and 90% Ankistrodesmus and (3) seston (100%) at a total concentration of 1.0 mg C l⁻¹.
in Jacarepaguá Lagoon were around 20–21 °C and maximum temperatures never exceeded 35 °C, which is considered a threshold for some tropical zooplankton species (HARDY & DUNCAN 1994). Temperature range in the tropics is, however, narrower than in the temperate zone, and can hardly be considered as a limiting factor for zooplankton.

Salinity seems to be another strong driving force for zooplankton populations in the lagoon. There were strong negative correlations between the density of cladocerans and salinity, suggesting that the increase in salinity was inhibitory for cladocerans. Actually, cladoceran populations increased mostly during the oligohaline and freshwater phase of the annual cycle and disappeared after salinity elevated up to 8‰ (mesohalinity). Other studies in coastal lagoons in the State of Rio de Janeiro have shown the effects of salinity in reducing cladoceran populations (BRANCO 1998). For copepods, salinity seems to have exerted a positive effect. Although Metacyclops mendocinus is a species frequently found in freshwater eutrophic conditions, the increase in its density at the end of the study, when salinity was high, shows that this copepod is highly tolerant to mesohaline conditions.

In spite of the relatively high POC concentrations in the lagoon, food availability was most conditioned by the phytoplankton composition. From August to December, POC concentrations averaged 10.4 to 15.1 mg C · l⁻¹ in station 1 and 2, respectively, with phytoplankton represented mostly by edible green algae such as Ankistrodesmus, Scenedesmus and Monoraphidium, diatoms such as Cyclotella, and nanoflagellates such as Pyramimonas. This high availability of good food likely fueled the increase in zooplankton densities, especially cladocerans, in this period. From January to April, POC concentrations remained high, averaging 10.2 to 15.1 mg C · l⁻¹ in station 1 and 2, respectively. However, there was an increase in the biomass and relative abundance of cyanobacteria, which was comprised almost by Microcystis aeruginosa. This period coincided with depressions in M. micrura populations (Fig. 4). From May to September, when cyanobacteria dominated over other phytoplankton taxa, POC averaged 9.2 and 9.8 mg C · l⁻¹ in station 1 and 2, respectively, with edible algae comprising only a small percentage of algal carbon. This period was marked by the complete disappearance of cladocerans in the lagoon.

As GULATI & DEMOTT (1997) pointed out, among the factors that determine food quality for zooplankton are size and shape of food particles, morphological defenses against digestion (digestion resistance), nutritional deficiency (P, N and fatty acids) and the presence of toxins. Although there was no significant correlation between cladocerans and toxins of cyanobacteria, there was a negative correlation between the density of cladocerans and the biomass of cyanobacteria, suggesting a possible detrimental effect of the Microcystis bloom on these organisms. These effects can be categorized as nutritional, since many cyanobacteria are considered a poor food for cladocerans (PORTER & ORCUTT 1980; DEBERNARDI & GIUSSANI 1990), and toxic effects, which are caused by the presence of toxins, such as microcystins (DEMOTT et al. 1991; RÖHRLÄK et al. 1999). However, as the acute experiments showed (Fig. 5), there was evidence of acute toxicity in December, immediately after the appearance of the Microcystis bloom in November 1996, and in February, coinciding with a decrease in M. micrura densities in the same period in the lagoon. Since we used both starved and fed animals in the controls during the acute experiments, nutritional deficiency seems to be a less plausible explanation for the high mortality of M. micrura in the Microcystis treatments, since animals in the food mixtures died faster than starved ones.

Therefore, it is likely that cyanobacterial toxins may have been the main factor responsible for the negative effect on cladocerans populations. The lack of significant correlations between cladocerans and toxins may have been caused by interfering factors such as colony size, which causes a lack of linearity between toxin concentrations in seston and ingestion rates of zooplankton. As shown by FERRÃO-FILHO & AZEVEDO (in press), the presence of large colonies in the seston of Jacarepaguá Lagoon may lead to low ingestion of Microcystis by cladocerans, mitigating toxic effects.

The reproduction of cladocerans was also affected by Microcystis. The reduction observed in mean fecundity and in intrinsic rate of natural increase (r) of M. micrura in the life-table experiment, both with seston and cultured Microcystis, suggests that Microcystis was the main cause of cladoceran declines in the lagoon. However, the higher fecundity and r-value for C. cornuta in the seston diet was probably sustained by the contribution of high quality algae and the low toxin content in the seston in this period. The growth experiment carried out with the same seston sample showed similar results.

![Fig. 8](image-url) Growth experiment with Moina micrura. Treatments were the same as in the life-table experiment (Fig. 7).
to the life-table experiment, corroborating the hypothesis of toxic effects of *Microcystis* on *M. micrura*.

The higher survivorship and fecundity of *Ceriodaphnia cornuta* in both seston and *Microcystis* treatments suggest that this cladoceran is less affected by the toxins of *Microcystis*. Also, its late appearance and increase during the *Microcystis* bloom and *Moina micrura* declining phase suggests that it is a more resistant species and probably has a superior competitive ability during blooms of cyanobacteria. The same conclusion was taken in a previous study by Ferrão-Filho et al. (2000), in which laboratory strains of *Microcystis* were used. As a small, slow-growing species, *Ceriodaphnia* seems to be more resistant to starvation and to toxins of *Microcystis* than the fast-growing *Moina*, which invests much of its energy in reproduction. Also, as Ferrão-Filho & Azevedo (in press) showed, the feeding rate of *Moina* is much more inhibited than that of *Ceriodaphnia* in the presence of toxic cells of *Microcystis* from natural assemblages. Thus, these results strongly support the hypothesis that *Ceriodaphnia* is more resistant to *Microcystis* toxins than *Moina*.

Although there was a surprisingly strong positive correlation between copepods and toxins in both stations, there is no evidence that *Microcystis* toxins may be beneficial for any zooplankton species. This was probably an indirect effect of the positive correlations between the density of copepods and the biomass of cyanobacteria. In fact, copepods are selective against toxic cyanobacteria (DeMott & Moxter 1991), and this is probably the reason why copepods coexisted with *Microcystis* at high densities in the lagoon. However, this cyclopoid copepod species has omnivory habits, and it is likely that it may have benefited from the detritus of the decaying *Microcystis* bloom, especially at the end of the study period. In lake Kasumigaura (Japan), it was shown that cladocerans were able to utilize decomposing *Microcystis* and associated bacteria (Hanazato 1991). In addition, the lack of correlation between POC and chlorophyll-α with phytoplankton (Table 1) shows that detritus may be an important component in POC dynamics in the lagoon, as in Amazonian floodplain lakes (Ferrão-Filho 2000).

Some rotifer species such as *B. angularis* and *H. oxyuris* had also a strong correlation with *Microcystis* toxins at station 1 (*r* = 0.814 and 0.910, respectively) and *B. plicatilis* at station 2 (*r* = 0.611). However, except for *B. angularis* and *H. oxyuris*, these correlations cannot be explained by an indirect correlation with cyanobacteria. Also, as *Microcystis* was mostly in the form of large colonies, it is unlikely that it could represent a food source for rotifers, unless they were broken into an edible size or decomposed. In fact, rotifer species have been reported to have different degree of sensitivity to toxic cyanobacteria (Fulton & Paerl 1987a; Gilbert 1994; Smith & Gilbert 1995; Gilbert 1996b; Nandini 2000). Fulton & Paerl (1987b) showed that *B. calyciflorus* was unaffected by both single cells or colonial *Microcystis* having high feeding rates, survivorship and growth rates in the presence of high densities of this toxic cyanobacterium. In another study, however, Nandini (2000) found a higher sensitivity of this rotifer species to single cells of *Microcystis* than to colonial forms, although both forms decreased survivorship and reproduction. Since some rotifers have been reported to be sensitive to neurotoxins of *Anabaena* spp. (Kirk & Gilbert 1992; Gilbert 1996a and b), we should expect a decrease in the density of rotifers when *Aphanizomenon* appeared in the lagoon. This cyanobacterium, like *Anabaena*, has been reported to produce neurotoxins (Carmichael 1992). However, density of rotifers actually increased at the end of the study period, especially *B. angularis*, suggesting that *Aphanizomenon* was not producing toxins or that rotifers in Jacarepaguá Lagoon are resistant to neurotoxins.

Differences in the sensibility of zooplankton species to toxic cyanobacteria have been regarded as an important factor structuring zooplankton communities (Kirk & Gilbert 1992; Fulton & Paerl 1988; Gilbert 1994; Nandini 2000). Effects of cyanobacteria on zooplankton, however, will depend on the size and morphology of colonies and filaments (Kirk & Gilbert 1992; Fulton & Paerl 1987b; Gilbert 1994), on the competitive ability of zooplankton species to exploit resources in the presence of cyanobacteria (Fulton & Paerl 1988) and on the sensitivity of the zooplankton species to toxins (DeMott et al. 1991; Ferrão-Filho et al. 2000). Smaller species of herbivorous zooplankton such as rotifers and small cladocerans are less susceptible to toxic cyanobacteria when they are present as large colonies and filaments and may outcompete larger species in environments dominated by cyanobacteria (DeMott 1989). In Jacarepaguá Lagoon, the zooplankton community is composed by rotifers and small crustaceans, which explains the coexistence with toxic blooms of cyanobacteria.

It has been proposed that the absence of large cladocerans in the tropics is an impediment to biomanipulation and clearance of cyanobacterial blooms (Archibald et al. 1995; DeClerk et al. 1997). Also, it has been thought that the persistence of cyanobacterial blooms during the seasonal cycle in the tropics should lead to the appearance of resistant species of zooplankton (Nandini & Rao 1998; Ferrão-Filho et al. 2000). This study and the study of Ferrão-Filho et al. (2000), however, suggest that resistance to toxic cyanobacteria is more related to life history than to zooplankton size or geographic origin. That is, small tropical cladoceran species can be as sensitive to cyanobacteria as temperate large cladocerans (Ferrão-Filho et al. 2000). Therefore, our results
and other studies do not support the idea of controlling cyanobacteria by top-down approach in the tropics.

Evidence of direct poisoning by cyanobacteria in field situations is unknown (HANEY 1987), and a few studies have dealt with field-laboratory approach to detect toxic effects of cyanobacteria on zooplankton (FERRAO-FILHO & AZEVEDO, in press). Most correlational field studies may have failed because the expression of toxic effects on the population level may be delayed by one or two generations. Although our results failed to show a direct correlation between toxins of Microcystis and the density of cladocerans in the lagoon, there was a good agreement between the field data and laboratory tests, which showed evidences of direct toxicity of Microcystis on one cladoceran population. Thus, we conclude that cyanobacterial blooms may be potentially harmful to zooplankton in nature.

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