

# Dynamics of Cladocera Community in a Tropical Hypereutrophic Environment (Garças Reservoir, São Paulo, Brazil)

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## Abstract

This study was performed at Garças Reservoir, a hypereutrophic urban lake within São Paulo metropolitan area, Southeast Brazil. This reservoir underwent to a marked limnological change after the harvest of free floating macrophytes, shifting to a hypereutrophic state. Present purposes were to characterize the Cladocera community and verify its association with the water deterioration after the macrophytes removal period as well as to compare it to previous studies, before macrophytes harvesting. Samplings were collected from October 2007 until September 2008 and the results were compared to data obtained during pre-removal period (1997). Principal Component Analysis (PCA) showed that the highest values for water transparency were associated with the macrophytes pre-removal period while the highest values of electrical conductivity, chlorophyll *a* and total phosphorus were associated with the macrophytes post-removal period, indicating the degradation of the water quality. During this period, large cladocerans disappeared and the small sized species of *Bosmina* predominated. Male organisms of this genus were found as well as elevated densities of *Bosmina huaruensis*, suggesting overcrowding. Furthermore, it was detected a reduction of 82% in the number of species compared to data from 1997. The shifts to the hypereutrophic state led to aggressive alterations in Cladocera community, suggesting a strong stress on populations.

## Keywords

Eutrophication, Zooplankton, Cladocerans, *Bosmina*, Tropical Reservoir

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## 1. Introduction

The aquatic macrophytes play an important role in the aquatic ecosystems. They participate in a variety of mechanisms including nutrient competition [1], allelopathic substances [2] and refugia for herbivorous zooplankton [3] [4].

The disappearance of submerged aquatic vegetation in shallow lakes due to eutrophication leads to a turbid state and provokes a dramatic change in the structure of lake biotic community [5]. Dominance of free floating macrophytes is another example of a stable state in shallow ecosystems [6], and a well-known consequence of eutrophication in tropical, subtropical and temperate systems is the outbreak of *Eichhornia crassipes* [7]-[9].

Long-term studies have been carried out at Garças Reservoir, which is the object of this study. A comprehensive limnological data set collected monthly during 8 years (1997-2004), including periods before and after the harvest of the water hyacinth, demonstrated this ecosystem shifted to a stable degraded state after the macrophytes removal, from eutrophic to hypereutrophic state with permanent cyanobacteria blooms [9] [10].

Despite the increasing knowledge about Garças Reservoir on phytoplankton ecology [11]-[13], algal richness loss in long time series [14], cyanobacteria toxicity [15], trophic state changes [16] [17], and more recently on paleolimnology [18].

The information on zooplankton community is very scarce. The only contribution available in [19] characterized this community in 1997, the macrophytes pre-removal period, and demonstrated the occurrence of trophic conditions indicator species such as rotifers *Brachionus angularis*, *B. calyciflorus*, *Keratella tecta*, the co-occurrence of cyclopoids copepods *Thermocyclops decipiens* and *Metacyclops mendocinus*, the calanoid copepods *Scolodiaptomus cordeiroi* and the cladocerans *Daphnia gessneri*, *Moina micrura* and *Moina minuta*. The role of zooplankton community as indicator of ecological state in biomonitoring programs is well recognized by considering these organisms respond quickly to changes in the water column.

This is the second study about zooplankton community at Garças Reservoir. In this context, the purposes of this study were to characterize the Cladocera community in a hypereutrophic state with permanent cyanobacteria blooms in a shallow tropical reservoir (Garças Reservoir), and compare it with previous study performed during the eutrophic phase (before water hyacinth removal). The results showed how zooplankton behaved in a permanent bloom stage environment and would contribute to the better management of shallow tropical reservoirs.

## 2. Material and Methods

### 2.1. Study Area

Garças Reservoir (23°38'S and 46°37'W) is located in the reserve of the Parque Estadual das Fontes do Ipiranga (São Paulo, Southeastern Brazil). It is a shallow reservoir, with maximum depth of 4.7 m and mean depth of 2.1 m, surface area of 88,156 m<sup>2</sup>, volume of 188,785 m<sup>3</sup> and mean theoretical residence time of 71 days [9].

Eutrophication was triggered by the construction of the city zoo (1958) and installation of the São Paulo State Department of Agriculture (1975) within the Garças watershed, increasing loads of untreated sewage from these institutions [18]. These authors inferred that deterioration in water quality began after ~1975 and markedly accelerated after ~1990. Based on a comprehensive data set of eight-year study, reference [9] identified three limnological phases in this environment according to physical, chemical and biological characteristics and considering the water hyacinth cover (*Eichhornia crassipes*):

Phase I (January 1997-March 1998): water surface coverage with 10% - 20% of water hyacinth, presence of cyanobacterial blooms during the spring, high concentrations of chlorophyll *a* and high values of pH on water surface.

Phase II (April 1998-August 1999): macrophytes covered 40% - 70% of the reservoir water surface. The large macrophyte patches triggered a serious problem with mosquitoes (*Mansonia* sp.), thus demanding its mechanical removal. This phase was characterized by the harvest of water hyacinth, low algal biomass, and the increase of water transparency and decrease of dissolved oxygen (DO).

Phase III (September 1999-December 2004) was characterized by the drastic reduction of the macrophyte cover on the water surface, along with an abrupt limnological changes such as a significant increase in chlorophyll *a*, cyanobacterial biovolume, total phosphorus (TP) concentrations and pH values and a drastic decrease in water transparency and DO at the deepest layers of the reservoir. During this phase, the spring cyanobacteria bloom was replaced by permanent multi-specific cyanobacteria blooms, and the reservoir shifted to a hyper-

trophic state [9] [10].

## 2.2. Abiotic and Biotic Variables

Sampling was carried out on a monthly basis from October 2007 to September 2008. A multiprobe HORIBA U-22 was used for measuring temperature, electrical conductivity (EC) and pH of the water column vertical profile at five different depths (subsurface, 1, 2, 3 m and bottom). Water transparency was measured with a Secchi disk. Water samples for determining dissolved oxygen (DO), chlorophyll *a*, total nitrogen (TN), and total phosphorus (TP) were collected at the same five different depths with a *van Dorn* sampler. The analyses were carried out at the Aquatic Ecology Laboratory of the Instituto de Botânica.

For determining the concentrations of DO, *Winkler* method was applied as described by [20], TN and TP ( $\mu\text{g/L}$ ) concentrations were measured according to reference [21], and chlorophyll *a* ( $\mu\text{g/L}$ ) concentrations corrected for phaeophytin were determined using 90% ethanol extraction [22]. Further details are available in reference [9].

Zooplankton samples were taken in vertical hauls of the entire water column using a plankton net of 68  $\mu\text{m}$  mesh and preserved in 4% formalin. Qualitative analysis was performed under stereoscopic microscope and the identification of the organisms according to specialized literature [23] [24]. The microcrustaceans were counted in subsamples in an acrylic chamber.

Numerical density of organisms was expressed in numbers of individuals per cubic meter ( $\text{ind/m}^3$ ) and the abundance in percentage (number of organisms of a taxon present in a sample relative to the total number of organisms).

Ecological indices such as richness (total number of taxa in a sample), diversity (bits/ind) [25] and evenness [26] were calculated in order to verify changes in biodiversity comparing 1997 with 2007 data.

## 2.3. Data Analysis

The water column mean was calculated per month considering the physical, chemical, and biological variables (water temperature, DO, EC, pH, TN, TP and chlorophyll *a*).

Principal Component Analysis (PCA) was performed based on data transformed by ranging and a covariance matrix of the following variables: water transparency, and monthly mean of water temperature, DO, EC, chlorophyll *a*, pH, TN and TP obtained in 1997 and in 2007/2008 (present study). PC-ORD version 4.0 for Windows [27] was used for the analysis.

## 3. Results

During the study period, water transparency remained low, pH ranged from slightly acid to neutral, electrical conductivity and nutrients concentrations, especially nitrogen and phosphorus were high (Table 1). Thermal stratification occurred in the rainy season (October/2007-March/2008) and more pronounced from December/2007 to February/2008, and the mixing of the water column occurred in April and May/2008, during the dry period (April-September/2008) (Figure 1(a)). Pronounced dissolved oxygen stratification occurred almost over the entire study period, leading to extended periods of oxygen depletion at the bottom layers (Figure 1(b)). Algal biomass (chlorophyll *a*) remained high, with annual mean of 119.4  $\mu\text{g/L}$  (Table 1).

The first two axes of principal component analysis (Figure 2) accounted for 70% of the limnological data variation. The first axis was positively related with water transparency, and negatively related with conductivity, total phosphorus, total nitrogen and chlorophyll (Table 2). In general, it separated the samples of the macrophyte pre-removal period (1997) from the post-removal one (2007/2008), characterized by lower transparency, high nutrients and chlorophyll *a* concentrations. The second axis separated the sampling units according to the seasonal variation, distinguishing the rainy period from the dry one in both studied periods (1997 and 2007/2008). Chlorophyll *a* and DO were positively related while water temperature was negatively related with this axis (Table 2 and Figure 2).

Cladocera was represented by five species: *Bosmina freyi*, *Bosmina huaruensis*, *Daphnia ambigua*, *Diaphanosoma birgei* and *Moina minuta*. *Bosmina* spp. represented 95% of total Cladocera, with dominance of *Bosmina huaruensis* (52%) followed by juveniles from *Bosmina* (36%), *Bosmina freyi* (5%) and males (2%). *Diaphanosoma birgei* represented 5% of total Cladocera and *Daphnia ambigua* and *Moina minuta* registered the lowest

**Table 1.** Mean values of water column and annual minimum, maximum, mean and standard deviation of limnological variables at Garças Reservoir from October 2007 to September 2008.

Months	Variables							
	SD (m)	T (°C)	DO (mg/L)	pH	EC (µS/cm)	TN (µg/L)	TP (µg/L)	Chlor <i>a</i> (µg/L)
Oct/07	0.19	19.6	3.4	7.2	351.6	4310.3	278.2	154.5
Nov/07	0.18	20.6	6.2	7.9	301.6	3612.6	264.2	198.7
Dec/07	0.20	22.4	0.4	6.1	457.2	6194.0	282.3	52.7
Jan/08	0.30	21.7	1.7	6.8	346.2	5073.3	290.0	76.8
Feb/08	0.26	23.2	3.4	7.2	340.2	1951.8	292.6	72.0
Mar/08	0.30	21.9	3.0	6.5	258.8	2226.9	225.9	133.7
Apr/08	0.30	21.4	3.1	6.9	294.8	6772.0	110.8	132.4
May/08	0.30	17.9	5.7	5.1	273.4	4847.0	143.5	149.4
Jun/08	0.23	18.1	4.0	6.4	164.8	5078.4	146.2	99.8
Jul/08	0.25	17.5	4.5	7.2	194.0	3805.8	130.4	69.8
Aug/08	0.22	18.6	4.3	7.5	190.0	3011.6	109.8	101.1
Sep/08	0.30	18.3	7.4	7.4	212.0	195.2	285.7	191.8
Annual mean	0.25	20.1	3.9	6.9	282.1	3923.2	213.3	119.4
Standard deviation	0.05	2.0	1.9	0.8	84.8	1871.7	77.8	48.4
Minimum	0.18	17.5	0.4	5.1	164.8	195.2	109.8	52.7
Maximum	0.30	23.2	7.4	7.9	457.2	6772.0	292.6	198.7

SD = Secchi depth; T = Water temperature; DO = Dissolved oxygen; EC = Electrical conductivity; TN = Total nitrogen; TP = Total phosphorus; Chlor *a* = Chlorophyll *a*.

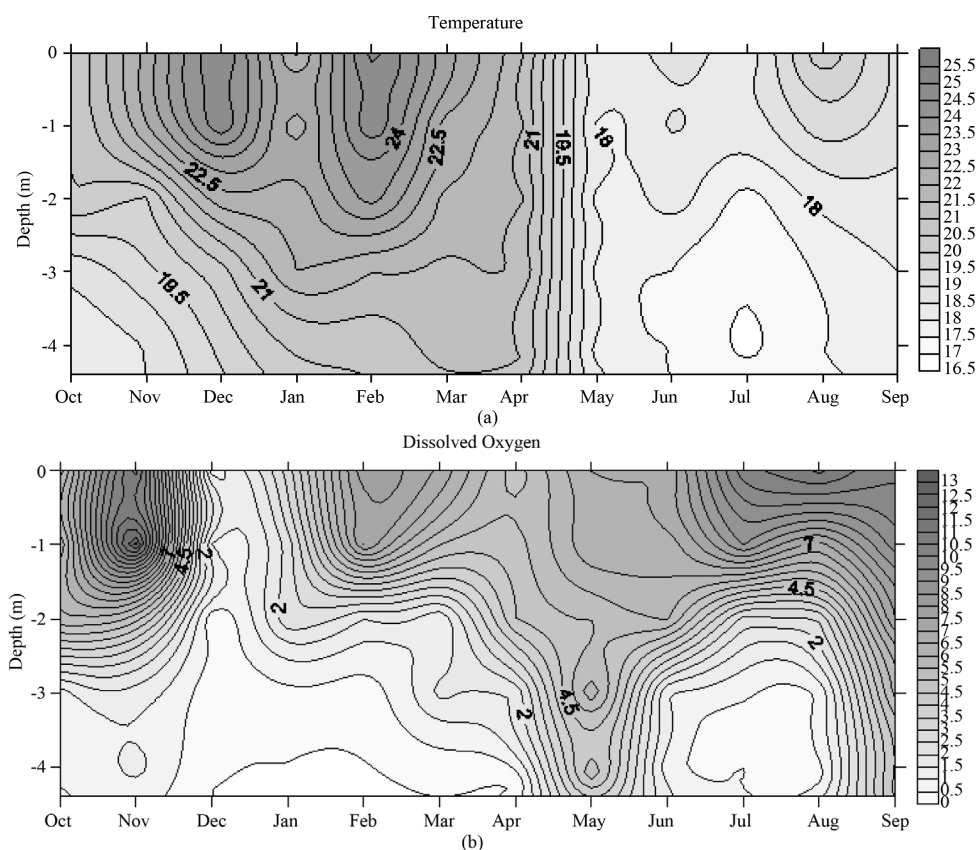
**Table 2.** Loadings of limnological variables on the first two principal components and the proportion of variance explained by each component of PCA in 1997 and 2007/2008.

Variables	Main Components	
	Axis 1	Axis 2
Secchi depth	0.759	-0.281
Water temperature	-0.293	-0.756
Dissolved oxygen	0.220	0.843
Electrical conductivity	-0.919	-0.144
Chlorophyll <i>a</i>	-0.590	0.665
Total nitrogen	-0.630	-0.392
Total phosphorus	-0.911	0.155
pH	0.023	0.023
Explained variation	45%	25%

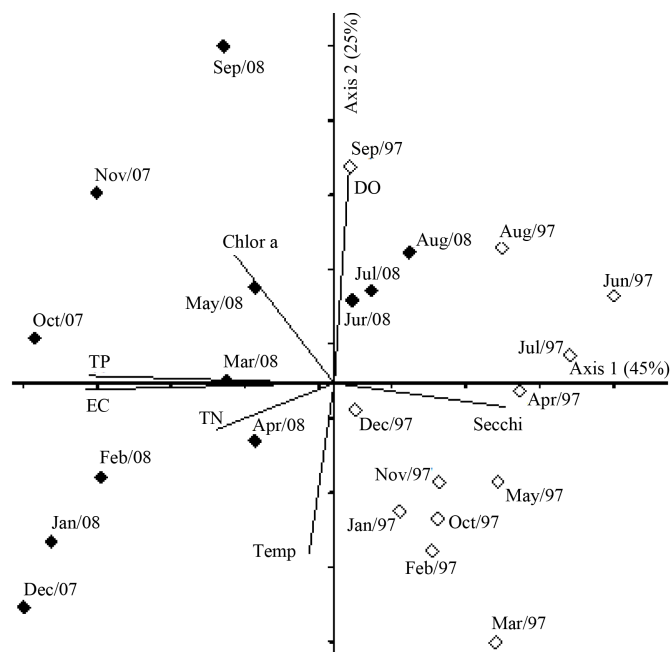
abundance (**Figure 3(b)**). In 1997, *Daphnia gessneri* dominated this group considering relative abundance (**Figure 3(a)**).

The highest numerical density of community occurred in October/2007 (1,226,800 ind/m<sup>3</sup>), and the lowest in December/2007 (159 ind/m<sup>3</sup>). Compared to 1997, the highest densities occurred during the macrophytes post-removal period (**Figure 4**).

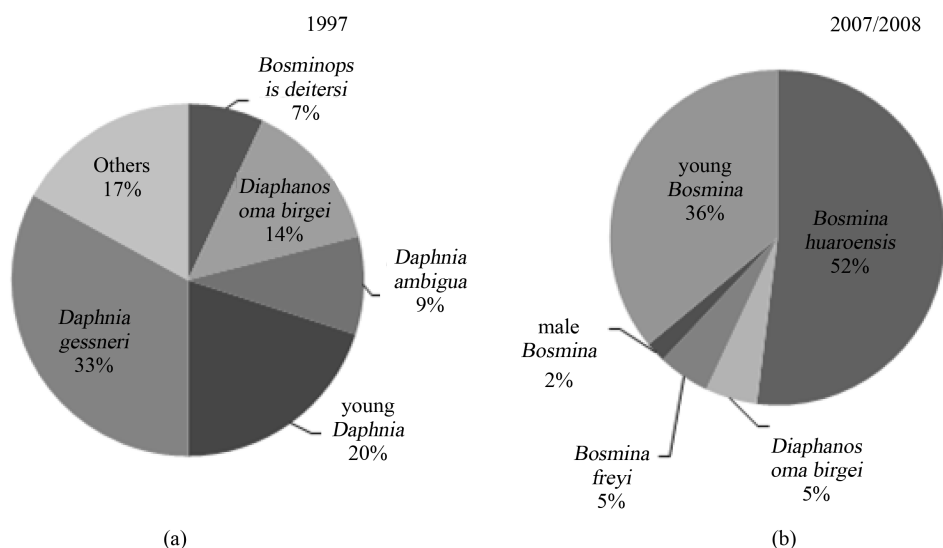
In 1997 28 taxa were registered while in 2007/2008 only five taxa were registered. Species richness and diversity were markedly higher in 1997, and evenness was slightly higher in 2007/2008 (**Figure 5**).



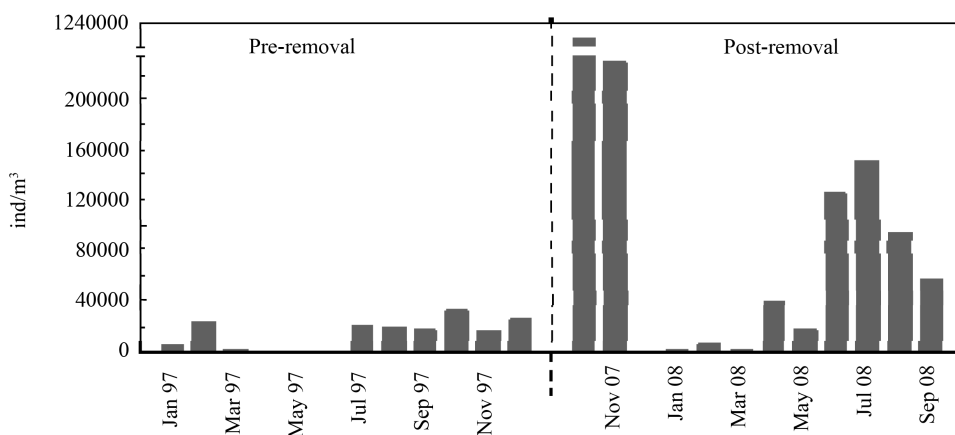
**Figure 1.** Depth-time diagram of (a) water temperature and (b) dissolved oxygen at Garças Reservoir from October 2007 to September 2008.



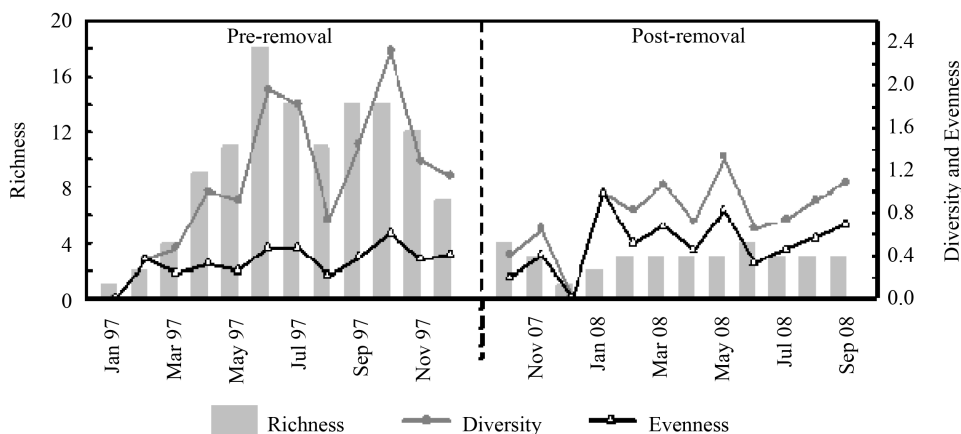
**Figure 2.** PCA ordination of limnological variables at Garças Reservoir in 1997 and 2007/2008. Abbreviations: Secchi: Secchi depth; Temp: Water Temperature; DO: Dissolved oxygen; EC: Electrical conductivity; TN: Total nitrogen; TP: Total phosphorus; Chlor *a*: Chlorophyll *a*.



**Figure 3.** Relative abundance (%) of Cladocera community at Garças Reservoir in (a) 1997 [19] and (b) 2007/2008.



**Figure 4.** Numerical densities (ind/m<sup>3</sup>) of Cladocera community at Garças Reservoir during macrophytes pre- (1997 [19]) and post-removal (2007/2008) periods.



**Figure 5.** Richness, Shannon diversity (bit/ind) and evenness of Cladocera community at Garças Reservoir during macrophytes pre- (1997 [19]) and post-removal (2007/2008) periods.

During the latter period, three species were numerically relevant. *Bosmina huarioensis* was dominant in October and November 2007 and August 2008. After registering the highest densities in October and November (706,692 and 134,302 ind/m<sup>3</sup>, respectively), this species disappeared during the following two months (December 2007 and January 2008) and re-appeared in February 2008 in dramatically lower densities (108 ind/m<sup>3</sup>).

*Bosmina freyi* was present all along this study and dominated the community in February and April 2008. *Bosmina* young individuals were registered the whole studied period and were dominant in December 2007, June and July 2008. Male individuals were observed in October and November 2007 and in June, July, August and September 2008. High density of male organisms occurred in the first month of this study (32,664 ind/m<sup>3</sup>).

*Diaphanosoma birgei* was the third more relevant species of Cladocera and was absent in December 2007 and January 2008. Nevertheless, it was the dominant Cladocera species in May and September 2008 (dry season). *Moina minuta* was dominant over other Cladocera species in January 2008, reaching 631 ind/m<sup>3</sup>.

#### 4. Discussion

The anoxic conditions observed in the deepest layers were propitious to the release of phosphorus by sediment (via feedback mechanisms), thus accelerating the eutrophication process as discussed in previous studies by [9] [17]. According to these authors, anoxic conditions were persistent after the macrophyte removal, and phosphorus dynamics, initially driven by allochthonous loads, was replaced by internal (autochthonous) ecological processes.

In this period when the high values of TP were detected, it was also observed low richness of the Cladocera community at Garças reservoir. Reference [28] showed that zooplankton species richness declined considerably with increasing total phosphorus concentration on Danish lakes. The decline was particularly noticeable for the number of cladoceran species. Reference [29] also found Cladocera assemblage changes with trophic state mainly due to total phosphorus concentration.

In Garças reservoir, only three species were common to the pre-removal (1997 [19]) and post-removal periods (2007/2008): *Daphnia ambigua*, *Diaphanosoma birgei* and *Moina minuta*.

Comparing 1997 with 2007/2008, twenty-five cladoceran species disappeared (*Alona guttata*, *A. monocantha*, *Alona* sp., *Biapertura* spp., *Bosmina hagmanni*, *B. tubicen*, *Bosmina* sp., *Bosminopsis deitersi*, *Camptocercus daday*, *Ceriodaphnia cornuta cornuta*, *C. cornuta rigaudi*, *C. cf. reticulata*, *Chydorus pubecens*, *C. sphaericus*, *Chydorus* sp., *Daphnia gessneri*, *Daphnia* sp., *Diaphanosoma* sp., *Disparalona daday*, *Ilyocryptus spnifer*, *Macrothrix spinosa*, *Macrothrix* sp., *Moina micrura*, *Moina* sp. and *Simocephalus* sp.). The disappearance of non-planktonic species can be attributed to the macrophytes removal because these organisms use these plants as refuge and food source.

The dense cyanobacterial blooms recorded continuously at Garças Reservoir [9] [10] probably led to the replacement of large cladocerans, like *Daphnia gessneri*, predominant species in 1997 [19], for small cladocerans, like *Bosmina huarioensis*, predominant in this study.

Other studies also found decrease in contribution of *Daphnia* spp. to the total biomass of cladocerans with increasing total phosphorus concentration [28]. Replacement of *Daphnia* spp. for *Bosmina huarioensis* was also reported in a hypereutrophic lake in Argentina, Don Tomás Lake [30].

In eutrophic lakes, large cladocerans (e.g. *Daphnia*) may disappear from the environment or be replaced by smaller organisms such as small cladocerans, rotifers and copepods due to the deterioration of water quality and cyanobacterial blooms [30]-[34]. Large cladocerans are impaired in water bodies dominated by cyanobacteria, among other factors for having a large carapace opening which allows the entry of cyanobacterial filaments and colonies in the filtering chamber, thus favoring the mechanical or chemical inhibition of the thoracic appendages movements by toxins. The reduction of the filtration rate mitigates the energy for growth and reproduction as a consequence. In smaller species of cladocerans, the carapace opening is too small for filaments and colonies to enter [35] what makes them less susceptible to the harmful effects of cyanobacteria.

Another reason for the successful presence of *Bosmina* in this environment is its dual-mode feeding which combines raptorial and filter-feeding that allows them to distinguish cyanobacteria from other particles. Besides the resistance to the harmful effects of cyanobacterial blooms, *Bosmina* can feed from bacteria and detritus [36]. This fact can explain their occurrence and predominance in eutrophic reservoirs.

By comparing the mean density of Cladocera at the macrophytes pre-removal period (13,395 ind/m<sup>3</sup> [19]) and post-removal period, an increase of approximately 1000% occurred in 2007/2008 (162,357 ind/m<sup>3</sup>). This oc-

curred due to the *Bosmina huaruensis* overcrowding in October and November 2007, when high densities of males of this genus were registered.

The reproductive strategies of cladocerans facilitate the development of large transitory population. These organisms are typically reproduced by asexual reproduction (parthenogenesis); however, environmental factors may induce sexual reproduction and production of resting eggs which allow the survival of the species during periods of harsh environmental conditions [37].

The resting eggs and male production are uncommon or unknown in some Cladocera species. It is generally related to adverse conditions such as changes in water temperature, overcrowding, that results in reduction of food supply, excretion products accumulation [38], decreasing of ingestion rate [39] and other factors probably still unknown.

*Bosmina huaruensis* disappeared for two subsequent months right after the period of overcrowding and appeared again in February 2008. The absence of male organisms was observed for six months until June 2008 when they were noticed again. It was detected harsh environmental conditions during the two-month period of *B. huaruensis* absence (December and January).

The disappearance of *Bosmina huaruensis* and of the male population, in addition to the low densities of cladocerans recorded in December may be associated with the phytoplankton collapse, followed by toxins release and the consequent high mortality rates of these organisms. Another factor resulting from the collapse, which might have harmed the zooplankton, is the low dissolved oxygen concentration throughout the highly stratified water column in December.

Changes in diversity and species richness were also detected. In 1997, the low diversity and evenness had already evidenced the predominance of few species, a typical condition of impacted environments. However, the reduction of 82% in richness value and the decrease of diversity values are indicative that changes have further restricted the environment to most adaptable species.

Although the evenness values did not present major changes, it should be considered that the number of species decreased significantly. Therefore the distribution of the relative abundances of species was more uniform when compared to the pre-removal period, which presented many species but few dominant and many rare ones.

Reference [40] investigated the relation between primary productivity and richness of species in lake communities (phytoplankton, rotifers, cladocerans, copepods, macrophytes and fish) and verified that the strongest and most consistent relation was the decline in richness of the crustacean zooplankton species with increased productivity. The variety of mechanisms that interfere in the relation richness-productivity includes the transient dynamics and the shift to a new system state as noticed at Garças Reservoir.

The free floating macrophytes removal in shallow systems may cause disastrous effects to the ecosystem as well as to biotic communities, as observed in this study for Cladocera. The deterioration of water quality after the hyper eutrophication triggered by the harvest of macrophytes promoted the reduction of species richness and diversity, changes in composition and structure of community with replacement of *Daphnia* by smaller cladocerans, signs of population stress with occurrence of male organisms and overcrowding.

## 5. Conclusion

We believe this study provides relevant information for the study of Cladocera as bioindicators of water quality and on the dynamics of these organisms in hypereutrophic tropical reservoirs, since it presents and discusses the data scarcely found in literature, like the occurrence of male organisms and overcrowding. Present results may contribute to further monitoring of zooplankton community as a tool to assess aquatic ecosystem with anthropogenic influences in tropical/subtropical areas.

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