

## Quantifying zooplankton species: use of richness estimators

Maria Carolina de A. Castilho<sup>1</sup>, Maria José dos S. Wisniewski<sup>2</sup>, Célio Wisniewski<sup>3</sup> & Érika dos S. Silva<sup>2</sup>

1. Departamento de Zoologia, Instituto de Biociências, Universidade Estadual Paulista, Caixa Postal 510, 18618-000, Botucatu, SP, Brazil. (mariacarolcastilho@gmail.com)
2. Instituto de Ciências da Natureza, Universidade Federal de Alfenas, 37130-000, Alfenas, MG, Brazil.
3. Instituto de Ciências Exatas, Universidade Federal de Alfenas, 37500-000, Alfenas, MG, Brazil.

Received 18 March 2015

Accepted 25 May 2016

DOI: 10.1590/1678-4766e2016011

**ABSTRACT.** Richness estimators (Jackknife 1, Bootstrap, Chao 1 and ACE) were used to relate zooplankton species richness with amount of water collected per sample and number of samples throughout the year for the limnetic region of Sapucaí River compartment of Furnas reservoir, state of Minas Gerais, Brazil. Seven 100 L samples were collected in sequence using a motor pump, and seven 70 L samples were collected in sequence using a plankton net (68  $\mu$ m mesh size) in vertical hauls, to totalize 450 L, in three stations of the reservoir. Twelve monthly samplings were carried out over a year. The assessment of richness was made by analyzing the asymptotic behavior of the estimator curves. The samplings reached the asymptote from 350 L of collection with trawls and 400 L using a suction motor pump and reached the plateau on the 8th collection, which included both dry and rainy seasons. Regardless of the type of sampling, the volume of 400 L and eight sessions throughout the year is enough to register 90% of the zooplankton richness in the environment.

**KEYWORDS.** Biodiversity, Cladocera, Rotifera, sampling effort, aquatic environments.

**RESUMO.** Quantificando espécies zooplânctônicas: uso de estimadores de riqueza. Buscando relacionar a riqueza de espécies zooplânctônicas com o volume de água coletado por amostra e com o número de amostragens ao longo do ano para a região limnética do compartimento do Rio Sapucaí no reservatório da UHE de Furnas, Estado de Minas Gerais, Brasil, foram utilizados estimadores de riqueza (Jackknife 1, Bootstrap, Chao 1 e ACE). Sete amostras de 100 litros foram coletadas com moto-bomba de sucção, e sete de 70 litros utilizando rede de plâncton de 68  $\mu$ m e arrastos verticais em um ponto do reservatório. Foram realizadas doze coletas mensais ao longo de um ano, nas quais foram coletados 450 litros de água por amostra, em três pontos do reservatório de Furnas. A avaliação da riqueza foi feita analisando o comportamento assintótico das curvas dos estimadores. Estes atingem a assíntota a partir de 350 litros para a coleta feita mediante arrastos e com 400 litros com utilização de moto-bomba de sucção, alcançando o platô na 8ª coleta, tendo sido abrangidas as estações seca e chuvosa. Independente do tipo de amostragem, o volume de 400 litros e oito coletas ao longo do ano mostra-se suficientes para registrar 90% da riqueza zooplânctônica do ambiente.

**PALAVRAS-CHAVE.** Biodiversidade, Cladocera, Rotifera, esforço amostral, ambientes aquáticos.

Biological diversity can be understood at three levels: species, habitat and genetic diversity. Diversity indices use the density and richness of species and infer on the distribution equitability of species in the environment (BEGON *et al.*, 2007; MELO, 2008).

Knowledge of aquatic diversity is fundamental to implement conservation programs and rational use of resources, to assess environmental impact for licensing of hydroelectric power plants and to regulate fish farm areas (KING & PORTER, 2005; ESKINAZI-SANT'ANNA *et al.*, 2005; SANTOS, 2006; PINTO-COELHO, 2004; ROCHA *et al.*, 2010; SANTOS-WISNIEWSKI *et al.*, 2011; BRITO *et al.*, 2011). These inventories become important because the degradation of natural ecosystems and biodiversity loss have increased in recent years.

To identify and count every species of a study area is very complex. Ecological studies search to estimate the species richness of these environments by sub-samples and the use of statistical techniques (MUIRHEAD *et al.*, 2006; MELO, 2008). Hence, the standardization of procedures and sampling effort is indispensable to reduce uncertainties and to make possible the comparison of species richness between different study environments (GOTELLI & COWELL, 2001). Among statistical techniques, non-parametric richness estimators, such as Chao, ACE and Jackknife estimators, have been used to improve sampling. These are based on the occurrence of rare species that appear in few samples or in low density (MAGURRAN, 2011). Other estimators, such as Bootstrap, give the same value for every species collected, whether they are rare or common, to estimate the

total richness (SANTOS, 2006).

The methodology and equipment used in samplings can interfere in species richness measurements, for example, the mesh size of the net used to collect aquatic organisms should consider organism size and escape capacity. In the same way, the collection procedure, such as the use of suction pumps or vertical hauls by net, integrating or not the water column, can interfere in results (PINTO-COELHO, 2004). Furthermore, the water volume for sampling should be enough to represent the environment diversity. For the aquatic environment, the standardization of sampling effort means that a minimal amount of collected water is established, per sample, for about 90% of the total environment richness to be registered (HECK *et al.*, 1975).

Species accumulation curves are good tools for assessing the effectiveness of a sampling method, since they represent the cumulative number of species observed in an area (or volume) as a function of sampling effort (KING & PORTER, 2005; MUIRHEAD *et al.*, 2006). On the other hand, rarefaction curves are used for direct comparisons between populations, or samples, to obtain the number of species expected in a random sample (MAGURRAN, 2011).

Due to seasonal variations in zooplankton composition and richness (DUMONT & SEGERS, 1996), a preliminary sampling is recommended to determine which sampling effort is required to access the highest possible richness in tropical aquatic environments, especially in temporary ponds. In the tropics, the high predation rates by planktivorous fish, cyanobacterial blooms and high pollution due to anthropogenic activities, are factors that influence the loss of diversity and have significant effects in water bodies (SARMA *et al.*, 2005).

This study aims to determine the minimum water volume needed per sample collected for the largest possible number of zooplankton species in the limnetic region of Sapucaí River compartment of Furnas reservoir, and the sufficient number of monthly samples over a year to reach maximum richness.

## MATERIALS AND METHODS

The reservoir of the Hydroelectric Power Plant (HPP) of Furnas is located in the south of the state of Minas Gerais, Brazil. It has a flooded area of 1,450 km<sup>2</sup> and 250 km of length, in both of their two main sub-axes or compartments, the Sapucaí and Grande rivers, respectively (CORGOSINHO & PINTO-COELHO, 2006). The average depth is 13 m and the maximum reaches 90 m, near the dam.

Samplings were carried out in HPP Furnas reservoir at the junction of rivers Verde and Sapucaí (VSJ) (21°27'03"S and 45°40'24"W) in December 2009, with a plankton net of 68 µm mesh size. First, seven samples of 100 L were collected sequentially integrating one-meter of water column, using a suction motor pump. After this, seven other samples of 70 L were collected with vertical hauls, using constant speed. Both methods were limited to one-meter depth from the water surface. The organisms were

subjected to narcotization process with CO<sub>2</sub> saturation by the addition of carbonated water to avoid body contraction of the zooplankton individuals. The samples were maintained in polyethylene bottles and fixed with formalin 4% (v/v). Additional single samples of 400 L were collected monthly, in Barranco Alto region in this same reservoir, from March 2011 to February 2012. They were collected at stations BA1 (21°10'33"S; 46°00'51"W), BA2 (21°10'17"S; 46°00'38"W) and BA3 (21°10'04"S; 46°00'26"W). The suction motor pump was used to integrate the water column, from the station depth to the water surface, and the sample was prepared and maintained as above.

The quantitative analysis, restricted to cladocerans and rotifers, was carried out on a gridded acrylic slide, under a stereoscope microscope (50x magnification), in sub-samples or the whole sample, depending on the sample characteristics. An optical microscope (up to 2000x magnification) and specialized bibliography were used for species identification (KOSTE, 1978; JERSABEK *et al.*, 2003; DE MELO & HEBERT, 1994; ELMOOR-LOUREIRO, 1997; ORLOVA-BIENKOWSKAJA, 1998; SMIRNOV, 1998; SEGERS, 2007; KOTOV, 2009; SINEV & ELMOOR-LOUREIRO, 2010). The frequency of species was determined considering the number of samples in which they occurred in relation to the total number of samples. The species were classified as constant (present in more than 80% of samples), frequent (between 50 to 80%), common (from 20 to 50%) and rare (< 20%) (DAJOZ, 1983).

From the sample analysis the non-parametric estimators (ACE, Chao1, Jackknife1 and Bootstrap) of species richness were calculated based on abundance using the EstimateS 8.2 software (COLWELL, 2009). Generally, these estimators make comparisons between rare species (*uniques* or *singletons*), present in a unique sample, and species present in, at least, two samples (*doubletons* or *duplicates*). These estimates were chosen according to KING & PORTER (2005) and SOUSA *et al.* (2014).

The richness evaluation was done by asymptotic behavior analysis of curves obtained from richness estimators.

## RESULTS

Firstly, the analysis of samples from the VSJ station (unique sampling data) was carried out. The curves of ACE, Chao 1, Jackknife 1 and Bootstrap estimators, as well as the observed richness curve (Sobs), for the two types of samplings (vertical hauls with mesh and suction motor pump) both adjusted for increasing volume using the seven collected samples, in sequence, showed that 90% of the maximum richness was reached, on average, with up to 400 L of filtered water (see Figs 2, 3).

For the suction pump sampling, the asymptote was obtained by filtration of 600 L. Only the Jackknife 1 estimator curve showed the organism richness increasing up to 700 L of sample, given that the curve of *uniques* began to decrease again (Fig. 2). For vertical haul sampling, about 80% of the maximum richness was reached using up to 140 L and above 95% using up to 350 L, for every richness curve. The

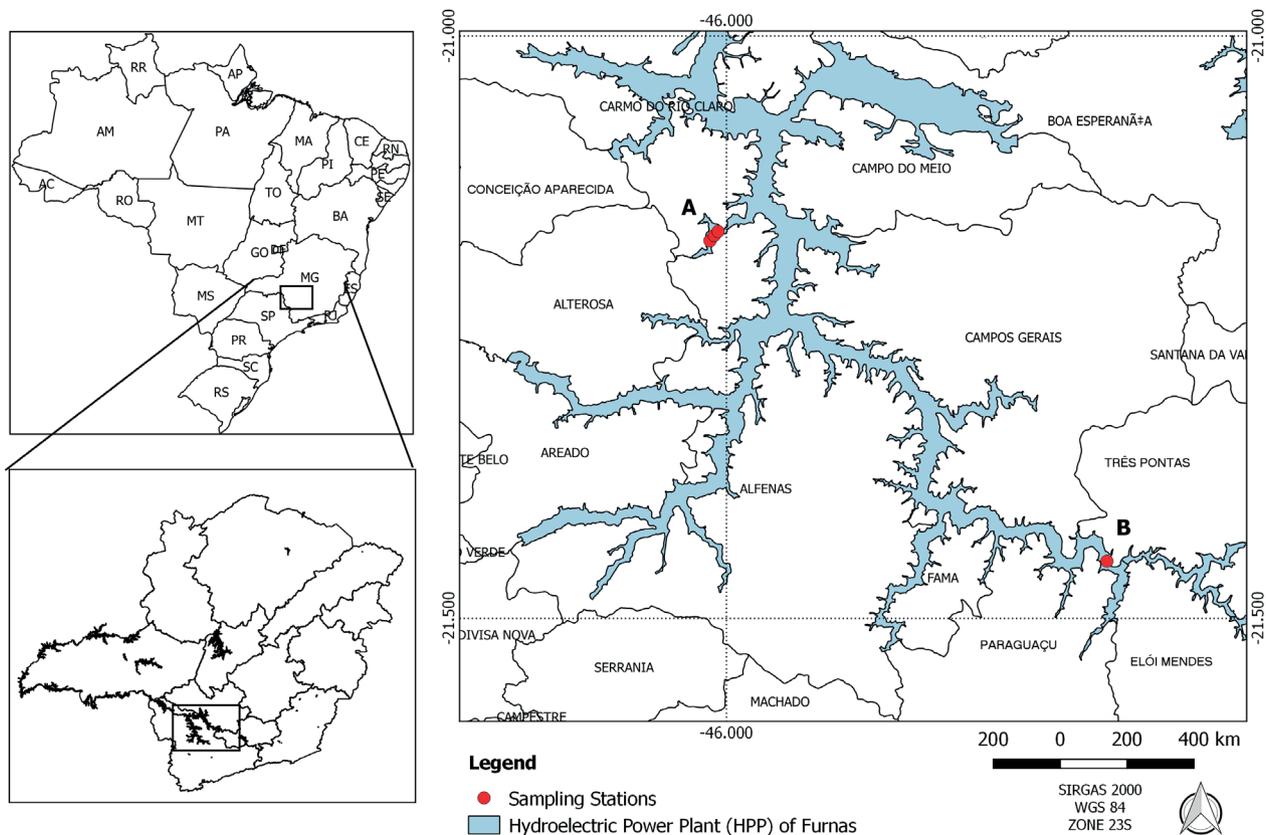


Fig 1. Sampling stations in the Hydroelectric Power Plant of Furnas reservoir, state of Minas Gerais, Brazil (A, Barranco Alto region; B, junction of rivers Verde and Sapucaí - VSJ).

observation of estimator curves showed that the asymptote was reached using up to 420 L (Fig. 3).

*Bosmina freyi*, *Bosminopsis deitersi*, *Moina minuta*, *Asplanchna sieboldi*, *Filinia longiseta*, *Hexarthra intermedia*, *Kellicotia bostoniensis* and *Synchaeta* sp. were found for every one of seven samples which were collected with suction motor pump and vertical haul samplings. However, *Ceriodaphnia cornuta rigaudi*, *Brachionus falcatus*, *Collotheca* sp., *Conochilus* sp., *Keratella americana*, *Lecane curvicornis*, *Platyas quadricornis*, *Plationus patulus*, *Polyarthra* sp. and *Testudinella patina* were constant only in samplings using vertical hauls (Tabs I, II).

The rare species *Tricocherca bicristata*, *Beucampiella* sp., *Chydorus eurynotus* and *Ascomorpha* sp., as well as *Lecane cornuta*, *Ptygura libera*, *Brachionus calyciflorus*, *Iliocryptus spinifer* and *Alonella lineolata* were recorded in only one of the seven samples, being the earliest obtained from vertical hauls and the latest from suction pump sampling.

For analysis in time scale, twelve samples were collected every month for one year. Standard species accumulation curves were similar for the three sampling stations BA1, BA2 and BA3, showed by Figures 3, 5 and 6. Chao 1 and ACE estimator curves approximately overlapped with the Sobs curve, because there were no singletons or doubletons in the samples and species occurred in more than 10 ind.L<sup>-1</sup>. Species with low frequency (*uniques* and *duplicates*) were recorded at high density, so Jackknife 1

and Bootstrap estimator curves showed species richness above those of the Sobs curve, in all samples. The minimal sampling effort was reached, for all curves, on the 8<sup>th</sup> sampling (asymptote), for both dry and wet seasons samplings.

*Bosmina hagemanni*, *Ceriodaphnia cornuta cornuta*, *C. cornuta rigaudi*, *C. silvestrii*, *Daphnia gessneri*, *Diaphanosoma birgei*, *D. spinulosum*, *Moina minuta*, *Conochilus unicornis* and *Keratella americana* were constant in all monthly samples (Tab. III).

## DISCUSSION

The two tested sampling methods, suction pump and vertical hauls, showed similar results and included in the inventory most of the species already registered for this reservoir, thus the two sampling methods are comparable. At 400 L water volume about 90% of the environmental species had been accessed for samplings using suction pump and a similar percentage was recorded with 350 L for samplings with vertical hauls using plankton net. It is considered a satisfactory inventory of species when between 50 and 75% of the species which could potentially occur in the environment are registered and the more frequently found species (or common) should be included in this percentage (HECK *et al.*, 1975).

The greater species richness was recorded for samplings using vertical hauls with a plankton net. Rotifers

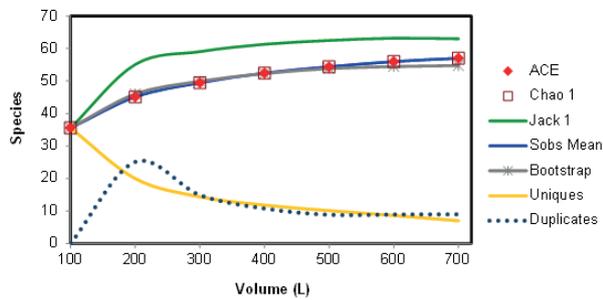


Fig. 2. Species accumulation curves, *uniques* and *duplicates* for the VSJ station in Furnas reservoir, state of Minas Gerais, Brazil, collected by suction pump.

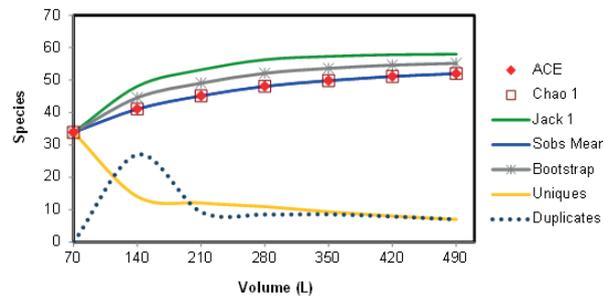


Fig. 3. Species accumulation curves, *uniques* and *duplicates* for the VSJ station in Furnas reservoir, state of Minas Gerais, Brazil, collected with vertical hauls.

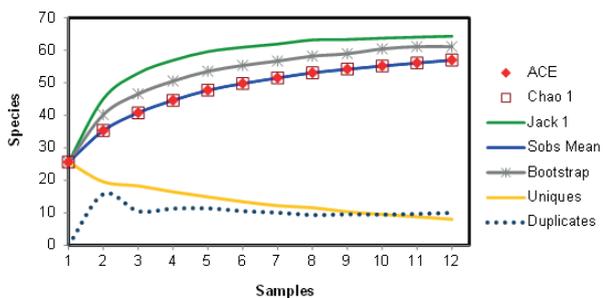


Fig. 4. Species accumulation curves, *uniques* and *duplicates* for the BA1 station of Furnas reservoir, state of Minas Gerais, Brazil, from March 2011 to February 2012.

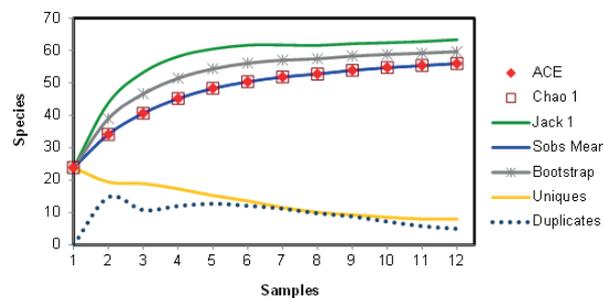


Fig. 5. Species accumulation curves, *uniques* and *duplicates* for the BA2 station of Furnas reservoir, state of Minas Gerais, Brazil from March 2011 to February 2012.

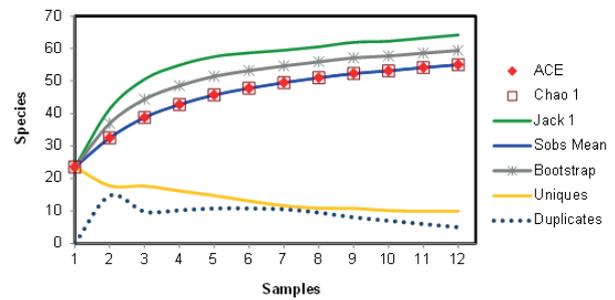


Fig. 6. Species accumulation curves, *uniques* and *duplicates* for the BA3 station of Furnas reservoir, state of Minas Gerais from March 2011 to February 2012.

were the most specious in this type of sampling while cladocerans were more representative with suction motor pump samplings. The most sensitive organisms, such as rotifers, may be damaged (including breakup and destruction of some individuals) during suction pump samplings, which makes their identification difficult (KOZŁOWSKY-SUZUKI & BOZELLI, 1998; PINTO-COELHO, 2004). On the other hand, the suction motor pump samplings select organisms with reduced escape capability and low (and slow) swimming movements, such as Cladocera (PINTO-COELHO, 2004).

The species richness for the three stations was very similar regarding the sampling throughout the year. With increasing sampling effort, the rarest species were identified. However, when the sampling effort is excessive, errant species are recorded, contributing to an increase in the *uniques* curve and even in this case the estimators extrapolate the total richness of the environment (MAGURRAN, 2011). Thus, it is believed that in 20 rare species sampled; only 8% are truly rare (KING & PORTER, 2005).

Many of the species identified in other studies in the Furnas reservoir were not recorded in this study, as they covered a larger area of this reservoir surrounding the limnetic and littoral regions and the two compartments (Rivers Grande and Sapucaí), resulting in greater richness. In the review by SANTOS-WISNIEWSKI *et al.* (2011) for Cladocera fauna of Minas Gerais, 94 species were recorded for the state distributed in 88 water bodies. Of these, 62 species were identified in the Furnas reservoir. Among the species not recorded in this study in the limnetic region were *Bosmina longirostris*, *Bosmina tubicen*, *Daphnia ambigua*, *Simocephalus latirostris*, *Simocephalus serrulatus* and *Moina micrura*.

Generally, the records of *Bosmina longirostris* for Brazil are dubious, because according to DE MELO & HEBERT (1994), the species occurs in North America and *B. freyi* in South America. In the present study and in earlier studies in the Furnas reservoir, only *B. freyi* was recorded.

Although the study was carried out in the limnetic region, typical species of the littoral region were recorded, such as *Alona intermedia*, *Alona yara*, *Alona guttata*, *Camptocercus australis*, *Chydorus pubescens*, *Chydorus eurynotus*, *Ephemeroporus* sp., *Ilyocryptus spinifer* and

Tab. I. List of occurrence of zooplankton species and Dajoz Constancy Index (DCI) for the seven samples (S) collected by suction pump at the VSJ station in Furnas reservoir, state of Minas Gerais, Brazil (Ct, constants; F, frequent; C, common; R, rare).

Taxa	S1	S2	S3	S4	S5	S6	S7	%	DCI
CLADOCERA									
<i>Alona</i> sp.	X	X					X	43	C
<i>Alonella lineolata</i> Sars, 1901			X					14	R
<i>Bosmina freyi</i> De Melo & Hebert, 1994	X	X	X	X	X	X	X	100	Ct
<i>Bosmina hagmanni</i> Stingelin, 1904				X	X	X		43	C
<i>Bosminopsis deitersi</i> Richard, 1895	X	X	X	X	X	X	X	100	Ct
<i>Ceriodaphnia cornuta</i> Sars, 1886		X						14	R
<i>Ceriodaphnia cornuta cornuta</i> Sars, 1886			X		X	X	X	57	F
<i>Ceriodaphnia cornuta intermedia</i> Sars, 1886	X						X	29	C
<i>Ceriodaphnia cornuta rigaudi</i> Sars, 1886	X		X	X	X	X		71	F
<i>Ceriodaphnia silvestrii</i> Daday, 1902	X		X	X	X	X		71	F
<i>Daphnia gessneri</i> Herbest, 1967		X		X	X	X		57	F
<i>Diaphanosoma birgei</i> Korineck, 1981	X	X		X	X	X	X	86	Ct
<i>Diaphanosoma spinulosum</i> Korineck, 1981	X		X	X	X	X	X	86	Ct
<i>Diaphanosoma</i> sp.	X		X			X		43	C
<i>Ilyocryptus spinifer</i> Korineck, 1981						X		14	R
<i>Leydigia striata</i> Berabén, 1939			X	X				29	C
<i>Macrothrix</i> sp.	X	X	X	X		X	X	86	Ct
<i>Moina minuta</i> Hansen, 1899	X	X	X	X	X	X	X	100	Ct
ROTIFERA									
<i>Asplanchna sieboldi</i> (Leydig, 1854)	X	X	X	X	X	X	X	100	Ct
<i>Beucampiella</i> sp.	X		X	X	X	X	X	86	Ct
<i>Brachionus calyciflorus</i> Pallas, 1766				X				14	R
<i>Brachionus dolabratus</i> Harring, 1915				X		X	X	43	C
<i>Brachionus falcatus</i> Zacharias, 1898		X	X	X	X		X	71	F
<i>Brachionus mirus</i> (Daday, 1905)	X	X					X	43	C
<i>Collotheca</i> sp.		X		X	X	X	X	71	F
<i>Conochilus</i> sp.				X	X	X	X	57	F
<i>Conochilus unicornis</i> (Rousselet, 1892)		X				X		29	C
<i>Euchlanis dilatata</i> Ehrenber, 1832		X	X					29	C
<i>Filinia longiseta</i> (Ehrenberg, 1834)	X	X	X	X	X	X	X	100	Ct
<i>Filinia opoliensis</i> (Zacharias, 1898)				X		X		29	C
<i>Hexartia intermedia</i> (Weiszniewski, 1929)	X	X	X	X	X	X	X	100	Ct
<i>Kellicottia bostoniensis</i> (Rousselete, 1908)	X	X	X	X	X	X	X	100	Ct
<i>Keratella americana</i> (Carlin, 1943)		X	X	X	X	X	X	86	Ct
<i>Keratella cochlearis</i> (Gosse, 1851)	X	X		X	X		X	71	F
<i>Keratella lenzi</i> (Hauer, 1953)		X	X					29	C
<i>Keratella tropica</i> (Apstein 1907)		X	X		X		X	57	F
<i>Lecane bulla</i> (Gosse, 1886)				X		X		29	C
<i>Lecane cornuta</i> (Müller, 1786)		X						14	R
<i>Lecane curvicornis</i> (Murray, 1913)	X	X		X	X	X	X	86	Ct
<i>Lecane prolecta</i> Hauer, 1956	X	X	X	X		X		71	F
<i>Lepadella</i> sp.			X	X		X		43	C
<i>Platyas quadricornis</i> (Ehrenberg, 1832)	X	X	X		X	X	X	86	Ct
<i>Platyonus patulus</i> (Müller, 1953)	X	X	X	X	X		X	86	Ct
<i>Polyarthra</i> sp.	X	X	X	X	X		X	71	F
<i>Proales</i> sp.	X	X	X	X	X	X		86	Ct
<i>Ptygura libera</i> Myers, 1934							X	14	R
<i>Sinchaeta</i> sp.	X	X	X	X	X	X	X	100	Ct
<i>Testudinella patina</i> (Hermann, 1783)	X	X	X		X		X	71	F
<i>Trichocerca cylindrica</i> (Sudzuki 1956)				X	X		X	43	C

Tab. II. List of occurrence of zooplankton species and Dajoz Constancy Index (DCI) for the seven samples (S) collected with vertical hauls in VSJ station in Furnas reservoir, state of Minas Gerais, Brazil. (Ct, constants; F, frequent; C, common; R, rare).

Taxa	S1	S2	S3	S4	S5	S6	S7	%	DCI
CLADOCERA									
<i>Alona</i> sp.		X						14	R
<i>Alonella dadayi</i> Birgei, 1910		X	X					29	C
<i>Bosmina freyi</i> De Melo & Hebert, 1994	X	X	X	X	X	X	X	100	Ct
<i>Bosmina hagmanni</i> Stingelin, 1904	X	X		X		X	X	71	F
<i>Bosminopsis deitersi</i> Richard, 1895	X	X	X	X	X	X	X	100	Ct
<i>Ceriodaphnia cornuta cornuta</i> Sars, 1886	X	X	X	X	X		X	86	Ct
<i>Ceriodaphnia cornuta rigaudi</i> Sars, 1886	X	X	X	X	X	X	X	100	Ct
<i>Ceriodaphnia silvestrii</i> Daday, 1902	X	X				X	X	57	F

Tab. II. Cont.

Taxa	S1	S2	S3	S4	S5	S6	S7		
<i>Chydorus eurynotus</i> Sars, 1901			X					14	R
<i>Daphnia gessneri</i> Herbest, 1967				X		X	X	43	C
<i>Diaphanosoma spinulosum</i> Korineck, 1981	X	X	X	X	X	X		86	Ct
<i>Diaphanosoma</i> sp.	X	X	X	X	X	X	X	100	Ct
<i>Macrothrix</i> sp.	X			X	X	X		57	F
<i>Moina minuta</i> Hansen, 1899	X	X	X	X	X	X	X	100	Ct
ROTIFERA		X						14	R
<i>Ascomorpha</i> sp.									
<i>Asplanchna sieboldi</i> (Leydig, 1854)	X	X	X	X	X	X	X	100	Ct
<i>Beucampiella</i> sp.	X							14	R
<i>Brachionus calyciflorus</i> Pallas, 1766			X				X	29	C
<i>Brachionus dolabratus</i> Harring, 1915		X	X	X				43	C
<i>Brachionus falcatus</i> Zacharias, 1898	X	X	X	X	X	X	X	100	Ct
<i>Brachionus mirus</i> (Daday, 1905)					X	X	X	43	C
<i>Cephalodella</i> sp.				X	X			29	C
<i>Collotheca</i> sp.	X	X	X	X	X	X	X	100	Ct
<i>Conochilus</i> sp.	X	X	X	X	X	X	X	100	Ct
<i>Conochilus coenobasis</i> (Skorikov, 1914)		X						14	R
<i>Conochilus unicornis</i> (Rousselet, 1892)	X	X						29	C
<i>Euchlanis dilatata</i> Ehrenber, 1832		X			X		X	43	C
<i>Filinia longiseta</i> (Ehrenberg, 1834)	X	X	X	X	X	X	X	100	Ct
<i>Filinia opoliensis</i> (Zacharias, 1898)					X	X	X	43	C
<i>Hexarthra intermedia</i> (Weiszniewski, 1929)	X	X	X	X	X	X	X	100	Ct
<i>Kellicottia bostoniensis</i> (Rousselete, 1908)	X	X	X	X	X	X	X	100	Ct
<i>Keratella americana</i> (Carlin, 1943)	X	X	X	X	X	X	X	100	Ct
<i>Keratella cochlearis</i> (Gosse, 1851)	X		X	X	X	X	X	86	Ct
<i>Keratella lenzi</i> (Hauer, 1953)	X	X	X		X	X	X	86	Ct
<i>Keratella tropica</i> (Apstein 1907)			X		X	X	X	57	F
<i>Lecane bulla</i> (Gosse, 1886)	X		X			X	X	57	F
<i>Lecane curvicornis</i> (Murray, 1913)	X	X	X	X	X	X	X	100	Ct
<i>Lecane leontina</i> (Turner, 1892)	X	X	X	X	X			71	F
<i>Lecane papuana</i> Murray 1913		X	X	X				43	C
<i>Lecane proiecta</i> Hauer, 1956			X			X		29	F
<i>Lecane quadridentata</i> (Ehrenberg, 1832)			X	X				29	F
<i>Lecane</i> sp.			X					14	R
<i>Platyas quadricornis</i> (Ehrenberg, 1832)	X	X	X	X	X	X	X	100	Ct
<i>Platyonus macracanthus</i> (Daday, 1905)	X	X	X	X	X	X		86	Ct
<i>Platyonus patulus</i> (Müller, 1953)	X	X	X	X	X	X	X	100	Ct
<i>Polyarthra</i> sp.	X	X	X	X	X	X	X	100	Ct
<i>Proales</i> sp.	X	X	X	X	X	X		86	Ct
<i>Synchaeta</i> sp.	X	X	X	X	X	X	X	100	Ct
<i>Testudinella patina</i> (Hermann, 1783)	X	X	X	X	X	X	X	100	Ct
<i>Trichocerca bicristata</i> (Gosse, 1887)				X				14	R
<i>Trichocerca cylindrica</i> (Suzuki 1956)					X		X	29	C
<i>Trichotria tetractis</i> (Ehrenberg, 1830)		X			X	X		43	F

Tab. III. List of occurrence of zooplankton species and Dajoz Constancy Index (DCI) in site BA1, BA2 and BA3 stations, from March 2011 to February 2012 (Ct, constants; F, frequent; C, common; R, rare; \*, occurrence in 1 station; \*\*, occurrence in 2 station; \*\*\*, occurrence in 3 stations).

Taxa	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	%	ICD
CLADOCERA														
<i>Alona guttata</i> Sars, 1862				*	*	***	*	*		*		*	25	c
<i>Alona yara</i> Sinev & Elmoor-Loureiro, 2010									**	*	**	*	17	R
<i>Alona intermedia</i> Sars, 1862			**										6	R
<i>Bosmina freyi</i> De Melo & Hebert, 1994	**		***	***	***	***	***	**	***	***	**	***	83	Ct
<i>Bosmina hagmanni</i> Stingelin, 1904	***	***	***	***	***	***	***	***	***	***	***	***	100	Ct
<i>Camptochercus australis</i> Sars, 1896											*		3	R
<i>Ceriodaphnia cornuta cornuta</i> Sars, 1886	***	***	***	***	***	***	***	***	***	***	**	***	97	Ct
<i>Ceriodaphnia cornuta intermedia</i> Sars, 1886		*	**	**	*	*			***	***	*	***	47	c
<i>Ceriodaphnia cornuta rigaudi</i> Sars, 1886	***	***	***	***	***	***	***	***	***	***	***	***	100	Ct
<i>Ceriodaphnia silvestrii</i> Daday, 1902	***	***	***	***	***	***	***	***	***	***	***	***	97	Ct
<i>Daphnia gessneri</i> Herbest, 1967	**	***	**	***	***	**	***	***	***	*	**	***	86	Ct
<i>Daphnia laevis</i> Birge, 1878		*					**	***					17	R
<i>Diaphanosoma birgei</i> Korineck, 1981	***	***	***	***	***	**	**	***	***	***	***	***	94	Ct
<i>Diaphanosoma brevireme</i> Sars, 1901	***	**	**			**	**	***	**	**	**	***	64	F

Tab. III. Cont.

Taxa	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Jan	Feb	%	ICD
<i>Diaphanosoma spinulosum</i> Herbst, 1967	***	***	***	**	***	**	**	**	***	***	***	***	89	Ct
<i>Diaphanosoma</i> sp.		*	*	*	*	*	*		*	*		*	25	c
<i>Diaphanosoma fluviatile</i> Hansen, 1899	*	**			**	**	**		***	***	***	***	58	F
<i>Ilyocryptus spinifer</i> Herrick, 1882		*		*									6	R
<i>Macrothrix</i> sp.							**	*		**			14	R
<i>Moina minuta</i> Hansen, 1899	***	***	***	***	***	***	***	***	***	***	***	***	100	Ct
<i>Simocephalus mixtus</i> Orlova-Bienkowskaja, 2001			*	**		**	***	*	*				28	c
<i>Chydorus pubescens</i> Sars, 1901			*	*		***	*	**			*		25	c
<i>Chydorus</i> sp.									*				3	R
ROTIFERA														
<i>Anuraeopsis</i> sp.		*							*			*	8	R
<i>Ascomorpha saltans</i> Bartsch, 1870			*		**		*		**	*	*		22	c
<i>Ascomorpha ovalis</i> (Bergendal, 1892)			**	*	*	***	***	***	**	*	*	**	53	F
<i>Asplanchna sieboldi</i> (Leydig, 1854)	***	*		**	*	***	***	*	***	***		*	58	F
<i>Brachionus calyciflorus</i> Pallas, 1766										*			3	R
<i>Brachionus dolabratus</i> Hanning, 1915	*	*								***	***	***	31	c
<i>Brachionus falcatus</i> Zacharias, 1898		***	*		**						***	**	31	c
<i>Brachionus mirus</i> (Daday, 1905)			*				*	*	**	**	***	**	33	c
<i>Brachionus</i> sp.				*									3	R
<i>Collotheca</i> sp.	*	***	**	*							***	*	31	c
<i>Conochilus natans</i> (Seligo, 1990)	***	**		*			*						19	R
<i>Conochilus</i> sp.				*	*	***		*					17	R
<i>Conochilus coenobasis</i> (Skorikov, 1914)		*	*								**	**	17	R
<i>Conochilus unicornis</i> (Rousselet, 1892)	***	***	***	***	***	***	***	***	***	***	**	***	97	Ct
<i>Euchlanis dilatata</i> Ehrenber, 1832		*				***	***	**	***	***	*		44	c
<i>Filinia longiseta</i> (Ehrenberg, 1834)	**	**						*	*	*	***	**	33	c
<i>Filinia opoliensis</i> (Zacharias, 1898)	*	***	**	**			*	**	*	***	**	***	56	F
<i>Filinia</i> sp.				*									3	R
<i>Gastropus</i> sp.	*	***		*	**	*	**	*					31	c
<i>Hexartra intermedia</i> (Weiszniewski, 1929)	***	**	**	**					*	***	***	***	56	F
<i>Kellicotia bostoniensis</i> (Rousselete, 1908)			*	***	***		**	**		*	**	*	42	c
<i>Keratella americana</i> (Carlin, 1943)	*	***	***	***	***	***	***	***	***	***	***	***	94	Ct
<i>Keratella cochlearis</i> (Gosse, 1851)	*	**		*	**		**	**	**	***	***	***	58	F
<i>Keratella lenzi</i> (Hauer, 1953)	**	***	**	***	***	***	***	**	*	*			64	F
<i>Keratella</i> sp.							*		*				6	R
<i>Keratella tecta</i> Turner, 1986									**	***	**	**	25	c
<i>Keratella quadrata</i> Plate, 1886					*					*			6	R
<i>Keratella tropica</i> (Apstein 1907)									*	***	*	*	17	R
<i>Lecane bulla</i> (Gosse, 1886)												*	44	c
<i>Lecane luna</i> (Müller, 1776)		**											6	R
<i>Lecane leontina</i> (Turner, 1892)			*										3	R
<i>Lecane lunaris</i> (Ehrenberg 1832)					*	*						*	8	R
<i>Lecane papuana</i> Murray 1913					*	*		*					6	R
<i>Lecane proietta</i> Hauer, 1956	***	***	***	***	**	*			*			*	47	c
<i>Lepadella</i> sp.					*	*	*						8	R
<i>Plationus macracanthus</i> (Daday, 1905)				*	*								6	R
<i>Polyarthra vulgaris</i> Carlin, 1943		*	**	***	**		**	**	**	*	**	**	53	F
<i>Ptygura libera</i> Myers, 1934		*										***	11	R
<i>Synchaeta jollyii</i> Shiel & Koste, 1993	***	**	***	**	***	***	**	**	***	**			69	F
<i>Testudinella patina</i> (Hermann, 1783)		*						*					6	R
<i>Trichocerca cylindrica</i> (Sudzuki, 1956)		***	*	*	***	**		*	***	***	***	**	61	F
<i>Trichocerca chatonni</i> (de Beauchamp, 1907)	*	**	**	**	*	**	**	*	***	**	***	*	61	F
<i>Trichocerca similis grandis</i> (Wierzejski, 1983)	*	**	*	***	*	***	***	***	**		***	*	64	F

*Macrothrix* cf. *elegans*. According to FERNANDO (2002), the distinction between limnetic and littoral zooplankton is often not observed in the tropics, which explains the occurrence of these phytoplous organisms in the samples.

In the review by ESKINAZI-SANT'ANNA *et al.* (2005), 300 species of Rotifera were recorded in Minas Gerais, which only six of these species are registered for the Furnas reservoir and recorded in this study, what indicates that an update for rotifers in Minas Gerais is necessary. SEGERS & DUMONT (1995) identified 102 species of rotifers in 20

points distributed in the Broa reservoir, including sampling in the limnetic and littoral regions of the reservoir, while in this study 42 species were recorded, which only covered the limnetic region.

Jackknife 1 and Bootstrap estimators curves showed richness above that verified by the observed richness curve (Sobs) throughout the study. This result is common when non-parametric estimators are used to estimate the richness of zooplankton species (DUMONT & SEGERS, 1996; MUIRHEAD *et al.*, 2006; SOUSA, 2014). The estimator richness obtained

by Jackknife 1 curve in BA1 station evidenced an asymptote, while for Bootstrap estimator curve, the asymptote was observed for BA1 and BA2 stations. The adjustments to other estimators curves showed a tendency to the formation of a asymptote, while for the *uniques* curves the asymptote was observed at all stations since 9<sup>th</sup> sampling.

The estimator curves showed that only between 3% and 16% of environmental potential richness was not recorded and the sampling may be considered satisfactory. The richness species determined by the *unique* curves showed the asymptote beginning at 9<sup>th</sup> successive monthly sampling. It indicates that the monthly sampling can be reduced to eight so that most of environment richness will be achieved in, in time scale. In the tropics, there is less variation in the seasonal succession of zooplankton species and species co-occur in both the summer and winter seasons (DUMONT, 1994). Thus, a smaller amount of samples is necessary and the time and sampling costs could be reduced.

The results obtained with the volume collected showed that 400 L are sufficient to record 90% of the environment richness, regardless of the sampling type (vertical hauls with a plankton net or suction motor pump). Also, another recommendation is to decrease from twelve to eight the monthly collections throughout the year, to goal a zooplankton limnetic species inventory, as the maximum number of species of Cladocera and Rotifera will be obtained in the waters of the Furnas reservoir in eight months.

**Acknowledgements.** We thank Professor Raoul Henry for his valuable comments which helped to improve the manuscript. This study was supported by “Eletrabras Furnas – Programa de P&D Aneel”.

## REFERENCES

- BEGON, M.; TOWNSEND, C. R. & HARPER, J. L. 2007. **Ecologia: de Indivíduos a Ecossistemas**. 4ed. Porto Alegre, Artmed. 752p.
- BRITO, S. L.; MAIA-BARBOSA, P. M. & PINTO-COELHO, R. M. 2011. Zooplankton as an indicators of trophic conditions in two large reservoirs in Brazil. **Lakes and Reservoirs: Reserch & Management** 16(4):253-264.
- COLWELL, R. K. 2009. **EstimateS: statistical estimation of species richness and shared species from samples**. Version 8.2 User's Guide and application. Available at <<http://viceroy.ebb.uconn.edu/EstimateS>>. Accessed on 6 July 2012.
- COLWELL, R. K.; MAO, C. X. & CHANG, J. 2004. Interpolating, extrapolating, and comparing incidence-based species accumulation curves. **Ecology** 85:2717-2727.
- CORGOSINHO, P. H. & PINTO-COELHO, R. M. 2006. Zooplankton biomass, abundance and allometric patterns along a eutrophic gradient at Furnas Reservoir (Minas Gerais, Brazil). **Acta Limnologia Brasiliensia** 18(2):213-224.
- DAJOZ, R. 1983. **Ecologia geral**. Vozes, Petrópolis. 472p.
- DE MELO, R. & HEBERT, P. D. N. 1994. A taxonomic reevaluation of North American Bosminidae. **Canadian Journal of Zoology** 72:1808-1825.
- DUMONT, H. J. 1994. The distribution and ecology of the fresh- and brackish-water medusae of the world. **Hydrobiologia** 272:1-12.
- DUMONT, H. J. & SEGERS, H. 1996. Estimating lacustrine zooplankton species richness and complementarity. **Hydrobiologia** 341:125-132.
- ELMOOR-LOUREIRO, L. M. A. 1997. **Manual de identificação de Cladóceros límnicos do Brasil**. Brasília, Universa. 156p.
- ESKINAZI-SANT'ANNA, E. M.; MAIA-BARBOSA, P. M.; BRITO, S. & RIETZLER, A. C. 2005. Zooplankton Biodiversity of Minas Gerais State: a Preliminary Synthesis of Present Knowledge. **Acta Limnologia Brasiliensia** 17(2):199-218.
- FERNANDO, C. H. 2002. Zooplankton and tropical freshwater fisheries. In: FERNANDO, C. H. ed. **Guide to Tropical Freshwater Zooplankton. Identification, Ecology and Impacts on Fisheries**. Leiden, Backhuys Publishers, p. 255-280.
- GOTELLI, N. & COLWELL, R. K. 2001. Quantifying biodiversity: procedures and pitfalls in the measurement and comparison of species richness. **Ecology Letters** 4:379-391.
- HECK, K. L.; VAN-BELLE, G. & SIMBERLOFF, D. 1975. Explicit Calculation of the Rarefaction Diversity Measurement and the Determination of Sufficient Sample Size. **Ecology** 56(6):1459-1461.
- JERSABEK, C. D.; SEGERS, H. & MORRIS, P. J. 2003. **An illustrated online catalog of the Rotifera in the Academy of Natural Sciences of Philadelphia (version 1.0: 2003-April-8)**. Available at <<http://rotifer.ansp.org/rotifer.php>>. Accessed on 14 December 2012.
- KING, J. R. & PORTER, S. D. 2005. Evaluation of Sampling Methods and Species Richness Estimators for Ants in Upland Ecosystems in Florida. **Environmental Entomology** 34(6):1566-1578.
- KOSTE, W. 1978. **Rotatoria**. Die RaÉdertiere Mitteleuropas, 2 vols. Berlim, Gebrüder Bornträger. 673p.
- KOTOV, A. A. 2009. A revision of *Leydigia* Kurz, 1875 (Anomopoda, Cladocera, Branchiopoda), and subgeneric differentiation within the genus. **Zootaxa** 2082:1-84.
- KOZŁOWSKY-SUZUKI, B. & BOZELLI, R. L. 1998. Avaliação da eficiência de três amostradores na estimativa de abundância de organismos zooplancônicos na lagoa de Cabiúnas. In: ESTEVES, F. A. ed. **Ecologia de lagoas costeiras do parque nacional da Restinga de Jurubatiba e do município de Macaé (RJ)**. Rio de Janeiro, Núcleo de Pesquisas Ecológicas de Macaé, UFRJ, p. 273-282.
- MAGURRAN, A. E. 2011. **Medindo a diversidade biológica**. Curitiba, Editora da UFPR. 261p.
- MELO, A. S. 2008. O que ganhamos ‘confundindo’ riqueza de espécies e equabilidade em um índice de diversidade? **Biota Neotropica** 8(3):21-27.
- MUIRHEAD, J. R.; EJSMONT-KARABIN, J. & MACISAAC, H. J. 2006. Quantifying rotifer species richness in temperate lakes. **Freshwater Biology** 51:1696-1709.
- ORLOVA-BIENKOWSKAJA, M. J. 1998. A revision of the cladoceran genus *Simocephalus* (Crustacea, Daphniidae). **Bulletin of the Natural History Museum** (Zoology series) 64(1):1-62.
- PINTO-COELHO, R. M. 2004. Métodos de Coleta, Preservação, Contagem e Determinação de Biomassa em Zooplâncton de Águas Epicontinentais. In: BICUDO, C. E. M. & BICUDO, D. orgs. **Amostragem em Limnologia**. São Carlos, RiMa, p.149-166.
- ROCHA, O.; SANTOS-WISNIEWSKI, M. J. & MATSUMURA-TUNDISI, T. 2010. Check List de Cladocera de Água Doce do Estado de São Paulo. **Biota Neotropica** 11(1):571-591.
- SANTOS, A. J. 2006. Estimativas de riqueza de espécies. In: CULLEN JR., L.; VALLADARES-PADUA, C. & RUDRAN, R. orgs. **Métodos de Estudos em Biologia da Conservação e Manejo da Vida Silvestre**. 2ed. Curitiba, Ed. Universidade Federal do Paraná. 652p.
- SANTOS-WISNIEWSKI, M. J.; MATSUMURA-TUNDISI, T.; NEGREIROS, N. F.; SILVA, L. C.; SANTOS, R. M. & ROCHA, O. 2011. O estado atual do conhecimento da diversidade dos Cladocera (Crustacea, Branchiopoda) nas águas doces do estado de Minas Gerais. **Biota Neotropica** 11(3):287-301.
- SARMA, S. S. S.; NANDINI, S. & GULATI, R. D. 2005. Life history strategies of cladocerans: comparisons of tropical and temperate taxa. **Hydrobiologia** 542:315-333.
- SEGERS, H. 2007. Annotated checklist of the rotifers (Phylum Rotifera), with notes on nomenclature, taxonomy and distribution. **Zootaxa** 1564:1-104.
- SEGERS, H. & DUMONT, H. J. 1995. 102+ rotifer species (Rotifera: Monogononta) in Broa reservoir (SP, Brazil) on 26 August 1994, with the description of three new species. **Hydrobiologia** 316:183-197.
- SINEV, A. Y. & ELMOOR-LOUREIRO, L. M. A. 2010. Three new species of chydorid cladocerans of subfamily Aloninae (Branchiopoda: Anomopoda: Chydoridae) from Brazil. **Zootaxa** 2390:1-25.
- SMIRNOV, N. N. 1998. A revision of the genus *Camptocercus* (Anomopoda, Chydoridae, Aloninae). **Hydrobiologia** 386:63-83.
- SOUSA, F. D. R.; ELMOOR-LOUREIRO, L. M. A.; MENDONÇA-GALVÃO, L. & PUJOL-LUZ, J. R. 2014. Evaluation of a new sampling method for assessing Cladocera richness (Crustacea, Branchiopoda) in macrophyte-rich wetlands. **Annales de Limnologie** 50:143-153.