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New isolates of *Trichoderma* antagonistic to *Sclerotinia sclerotiorum*

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Abstract: Forty-nine isolates of *Trichoderma* from the Brazilian Midwest were evaluated for their antagonistic activity *in vitro* against *Sclerotinia sclerotiorum* (causal agent of white mold), which were then identified based on their nuclear ribosomal ITS sequences. Paired culture tests showed that all isolates exhibited some antagonism, with a maximum of 77% mycelial inhibition and complete inhibition of sclerotia production. Two isolates were found to be the most promising biocontrol agents, considering both antagonistic parameters (CEN1253 - *T. koningiopsis* and CEN1265 - *T. brevicompactum*). Five different species were identified: *T. harzianum* (23), *T. spirale* (9), *T. koningiopsis* (8), *T. brevicompactum* (7) and *T. asperellum* (2). These isolates are stored in the Embrapa Fungi Collection for Biological Control and the information obtained in the experiments will be incorporated into the database of biological assets within the genetic resources information system (Allele) and be made available for further studies.

Keywords: *Microbial genetic resources, molecular identification, biocontrol, white mold.*

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Resumo: Quarenta e nove isolados de *Trichoderma* obtidos no centro-oeste do Brasil foram avaliados quanto a sua atividade antagônica *in vitro* contra *Sclerotinia sclerotiorum* (agente causal do mofo branco) e identificados com base nas sequências ITS do DNA ribossômico nuclear. Os testes de cultivo pareado mostram que todos os isolados exibiram algum antagonismo, com um máximo de 77% de inibiação micelial e inibição total da produção de escleródios. Dois isolados se destacaram como os mais promissores, considerando ambos os parâmetros avaliados (CEN1253 - *T. koningiopsis* e CEN1265 - *T. brevicompactum*). Cinco espécies diferentes foram identificadas: *T. harzianum* (23), *T. spirale* (9), *T. koningiopsis* (8), *T. brevicompactum* (7) and *T. asperellum* (2). Estes isolados estão armazenados na Coleção de Fungos para Controle Biológico da Embrapa e as informações obtidas nos experimentos serão incorporadas na base de dados de ativos biológicos, no sistema de informações de recursos genéticos, e disponibilizados para estudos futuros.

Palavras-chave: *Recursos genéticos microbianos, identificação molecular, biocontrole, mofo branco.*

Introduction

Sclerotinia sclerotiorum Lib. de Bary (*Ss*) is the causal agent of white mold, a disease that affects many hosts such as legumes, sunflowers, canola, most vegetables, and others. This pathogen is widespread in Brazil and the fungus can cause serious losses, mainly due to its contamination or infestation of seeds (Heffer Link & Johnson 2007). Furthermore, the pathogen survives for long periods using specialized resistant structures, called sclerotia (Bianchini et al. 2005). For soil pathogens, chemical control rarely leads to satisfactory results. Furthermore, chemical pesticides pose health risks, lead to the reduction of beneficial soil microflora and increase production costs (Fahmi et al. 2012). In this context, biological control of plant diseases using antagonists has already shown good results in Brazil, as in other countries with more tradition in this practice, such as USA, Canada, Australia and France (Campanhola & Bettiol 2003). Particularly in the areas of organic production, there is high demand for biological control agents, because they have a low impact on the environment, leave no toxic residues in food and are fully compatible with other alternative control measures. However, one of the keys to success in the use of biofungicides is that they must be developed from strains with high activity against the pathogens in question and be adapted to the environmental conditions under which they will operate.

It is necessary to maintain a continuous flow of collection, isolation, evaluation and characterization of biocontrol agents to improve and maintain their efficiency (Lopes et al. 2013). Species of *Trichoderma* Pers. are widely used in the biological control of plant pathogens, but commercial formulations have only become available recently. The lack of properly registered formulations has been a serious obstacle in the use of bioproducts in Brazil (Machado et al. 2012). According to the Brazilian Ministry of Agriculture Livestock and Food Supply (MAPA) database (Agrofit 2016), there are five commercial products recommended for *Ss* control, formulated with either *T. asperellum* and *T. harzianum*. In order to select isolates for new formulations, some studies have reported the effectiveness of different species of *Trichoderma* in control of *Ss*, showing both inhibition of mycelial growth (Amin et al. 2010, Samuel et al. 2010, Matroudi et al. 2009) and the parasitism of sclerotia (Abdullah et al. 2008).

The current taxonomy of *Trichoderma* is based mainly on molecular analysis (Druzhinina et al. 2010, Kullning et al. 2000), since morphological identification of the anamorph forms are unreliable (Druzhinina et al. 2006). With the development of molecular phylogeny, the genus *Trichoderma* has been grouped into taxonomic sections (Bissett 1984); based on phylogenetic clades (Druzhinina et al. 2010). As an extension of this work, Druzhinina and colleagues (2005) developed a DNA barcode for molecular identification of *Trichoderma* species, called *Trich*OKEY. Given the importance of biological control within the sustainable management of agriculture and environmental protection, the current investigation was undertaken to prospect for *Trichoderma* isolates with potential for inhibition of *Ss* in soil of the Federal District, Brazil and to identify them using nrRNA gene ITS sequences.

Material and Methods

The samples were obtained from soils from organic farming systems and native vegetation, from Rural Center Rajadinha (Planaltina, Federal District). The region is part of the Brazilian central highlands, with an altitude of around 954 m. The main soil type that predominates in the region is red latosol. The collected soils were extracted from fractions of rhizoplane, rhizosphere and roots, up to 5 cm from the following plants: maize (*Zea mays* L.); eggplant (*Solanum melongena* L.); okra (*Abelmoschus esculentus* L. Moench); tomato (*Solanum lycopersicum* L.); squash (*Cucurbita* sp.); cassava (*Manihot esculenta* Crantz); cabbage (*Brassica oleracea* var. *capitata* L.); ornamental fern (*Pleopeltis* sp.), kale (*Brassica oleracea* L. var. *acephala* DC.); *Cestrum* sp. and *Miconia* sp. (Table 2). Pure cultures of the antagonist, originating from monosporic cultures, were tested against *Ss* strain CEN1157 in paired culture experiments, according to Dennis & Webster (1971). The plates were incubated aerobically for seven days at 25 °C, in a photoperiod of 12 hours.

The confrontation tests were performed in triplicate and the control was a disc of culture medium with *Ss* in the center of plate. Radial mycelial growth of the pathogen (cm) was measured with the aid of a millimeter ruler and used for calculating inhibition of mycelial growth rate (Menten et al. 1976) using the equation: $MGI = [(D_{test}-D_{trat}/D_{trat})*100]$; where D_{test} is the diameter of the radial mycelial growth of *Ss* in the control treatment without *Trichoderma*), D_{trat}, is the diameter of the radial mycelial growth of *Ss* in treatment with *Trichoderma*. The number of sclerotia produced by *Ss* was also counted in the confrontation cultures zones.

Comparison of means was performed using the Scott-Knott clustering test. The model residues were subjected to Box-Cox transformation when they had normal variance. Box-Cox consisted of transforming the data according to expression: $Y' = (Y\lambda\text{-}1)/\lambda$, where Y is the response variable under investigation and λ is a parameter to be estimated. Analyses were performed in the statistical program R (http://www.r-project.org/). The significance level for all analyses was 5%.

Genomic DNA was extracted using a phenol-chloroform method (Raeder & Broda 1985) from *Trichoderma* isolates grown in potato dextrose broth in a shaking incubator at 150 rpm for five days. The gDNA was quantified by comparison to a high molecular weight DNA Mass Ladder (Invitrogen). The nuclear ribossomal ITS locus (rDNA; ITS1-5.8S-ITS2) was amplified by PCR using the primers ITS1 5'-TTCCGTAGGTGAACCTGCGG-3' and ITS4 5'-TCCTCCGCTTATTGATATGC-3' (White et al. 1990). The PCR reaction volume was 25 µL and contained: 5 ng template DNA; 1 X PCR buffer; 0.2 mM dNTPs; 1.5 mM $MgCl_2$; 0.4 µM each primer; 1 U Taq DNA polymerase. PCR cycling consisted of an initial 96 °C for 5 min, then 30 cycles 94 °C for 45 s, 60 °C for 45 s and 72 °C for 1 min with a final extension at 72 °C for 10 min. PCR products were visualized using 1% agarose gels and the products subjected to Sanger chain termination sequencing (both strands) using the amplification primers (Macrogen Inc.; Seoul, South Korea).

Contigs were assembled using DNA BASER (http://www.dnabaser.com/index.html). BLAST (Basic Local Alignment Search Tool) searches of the NCBI (National Center for Biotechnology Information) Genbank nucleotide database and *Trich*OKEY 2.0 (http://www.isth.info/) were used to verify the ITS sequences. Experimentally derived sequences and related reference ITS sequences obtained from Genbank were aligned using MAFFT with its G-INSI option (Katoh & Standley 2013). A phylogenetic hypothesis was calculated using MrBayes (v3.2.5; Ronquist & Huelsenbeck 2003). The Bayesian analysis was conducted using default program settings except that reversible-jump

MCMC (Huelsenbeck et al. 2004) was used to optimize base substitution model parameters within the general time reversible (GTR) framework. MCMC sampling was run for 2 million generations, which allowed the standard deviation of split frequencies to fall to 0.006853, strongly indicating stable convergence. The analysis was repeated to confirm the result obtained. The first 25% of trees were discarded as *burn-in* prior to calculation of the 50% majority rule consensus phylogram. The ITS sequence from *T. longibrachiatum* (Table 1) was used as outgroup for tree rooting.

Table 1. Reference ITS sequences used for identification.

GenBank accession number	Species	Reference
GU198318.1	*T. asperellum*	Samuels et al. (2010)
EU330949.1	*T. brevicompactum*	Degenkolb et al. (2008)
AY857253.1	*T. harzianum*	Druzhinina et al. (2005)
DQ313142.1	*T. koningiopsis*	Samuels et al. (2006)
EU401576.1	*T. longibrachiatum*	Druzhinina et al. (2008)
AY857246.1	*T. spirale*	Druzhinina et al. (2005)

Results

In the paired culture experiments, we observed inhibition of mycelial growth of *Ss* ranging between 28, and 77% (Table 2).

In the statistical analysis of MGI, we observed the formation of five distinct groups (horizontal bars; Figure 1) at the 5% significance level in the Scott-Knott test, as shown (Figure 1). The data were transformed (Box-Cox, λ = 0.42) and the model was able to explain 91.69% of the total variability in the data (SQResidues= 33.14, SQTreat= 109.98, R^2=69%, $F_{48,98}$= 6.775, p-value= 7.8e-16).

Production of *Ss* sclerotia was also reduced in the confrontation cultures by the action of *Trichoderma* isolates (Figure 2). At the 5% level of significance, the ScottKnott test showed that the isolates formed two separate groups. The data were transformed (Box-Cox, λ = 0.55) and the model was able to explain 66% of the total variability in the data (SQResidues= 222.6, SQTreat= 660.3, R^2=66%, $F_{37,76}$= 6.092, p-value < 1.61e-11). The isolates CEN1253, CEN1314, CEN1313, CEN1257 and CEN1304 all caused complete inhibition of sclerotia production (Table 2). The first group of 25 isolates had less inhibition of sclerotia production, ranging between 27 and 10 sclerotia, while in the second group, between eight and two sclerotia were produced.

In the phylogenetic analysis of ITS sequences, rooted with *T. longibrachiatum,* we observed five major clades (Figure 3). The first 23 isolates grouped with the *T. harzianum* reference with a posterior probability of one, although some variability within the clade was present. The second clade contained nine isolates clustering with *T. spirale*, again with a posterior probability of one. The third group comprised into two related clades, one containing eight isolates and *T. koningiopsis*, and the other two isolates clustering with *T. asperellum*. Finally, the fifth clade contained seven isolates and *T. brevicompactum*.

Table 2. List of the *Trichoderma* isolates, results of the *in vitro* mycelial growth inhibition test (MGI) percentage and number of sclerotia (NS) produced by *Sclerotinia sclerotiorum*.

Embrapa Culture collection number	Origin of isolates		Observations	
	Plant	Niche	MGI(%)	NS
CEN1242	Maize	Soil	48	8
CEN1243	Eggplant	Soil	57	23
CEN1244	Okra	Rhizoplane	54	22
CEN1245	Tomato	Rhizosphere	53	27
CEN1247	Squash	Rhizosphere	50	6
CEN1248	Squash	Rhizoplane	51	18
CEN1249	Tomato	Rhizoplane	62	2
CEN1250	Tomato	Soil	63	10
CEN1251	Maize	Soil	53	14
CEN1252	Maize	Soil	50	16
CEN1253	Cassava	Soil	77	0
CEN1254	Tomato	Rhizoplane	60	14
CEN1256	Tomato	Rhizoplane	60	19
CEN1257	Tomato	Soil	59	0
CEN1258	Cassava	Soil	58	14
CEN1259	Cassava	Soil	46	1
CEN1260	Cassava	Rhizosphere	54	17
CEN1261	Cabbage	Rhizosphere	53	10
CEN1262	Eggplant	Rhizosphere	55	15
CEN1263	Eggplant	Rhizosphere	53	13
CEN1264	Eggplant	Rhizosphere	71	4

Continued Table 2.

Embrapa Culture collection number	Origin of isolates		Observations	
	Plant	**Niche**	**MGI(%)**	**NS**
CEN1265	Tomato	Rhizoplane	70	1
CEN1266	Ornamental fern	Rhizosphere	48	10
CEN1267	Maize	Rhizosphere	44	12
CEN1268	Maize	Rhizoplane	51	6
CEN1269	*Miconia* sp.	Soil	28	18
CEN1270	*Miconia* sp.	Soil	53	26
CEN1271	Cassava	Rhizosphere	62	3
CEN1272	*Cestrum* sp.	Soil	36	19
CEN1274	Kale	Soil	48	11
CEN1298	Tomato	Rhizoplane	63	2
CEN1299	Kale	Solo	65	11
CEN1302	Tomato	Soil	58	6
CEN1303	Eggplant	Soil	61	18
CEN1304	Squash	Rhizoplane	58	0
CEN1306	Tomato	Rhizoplane	59	1
CEN1311	Cassava	Soil	54	2
CEN1313	Cabbage	Rhizosphere	62	0
CEN1314	Eggplant	Rhizosphere	66	0
CEN1319	Ornamental fern	Rhizosphere	69	7
CEN1326	Kale	Rhizoplane	59	3
CEN1333	Cassava	Rhizoplane	56	4
CEN1337	*Miconia* sp.	Rhizoplane	52	7
CEN1339	*Miconia* sp.	Soil	60	7
CEN1345	Squash	Soil	50	6
CEN1346	Cassava	Soil	51	17
CEN1347	Squash	Rhizoplane	65	9
CEN1348	Squash	Soil	63	10
Control treatment	-	-	-	30

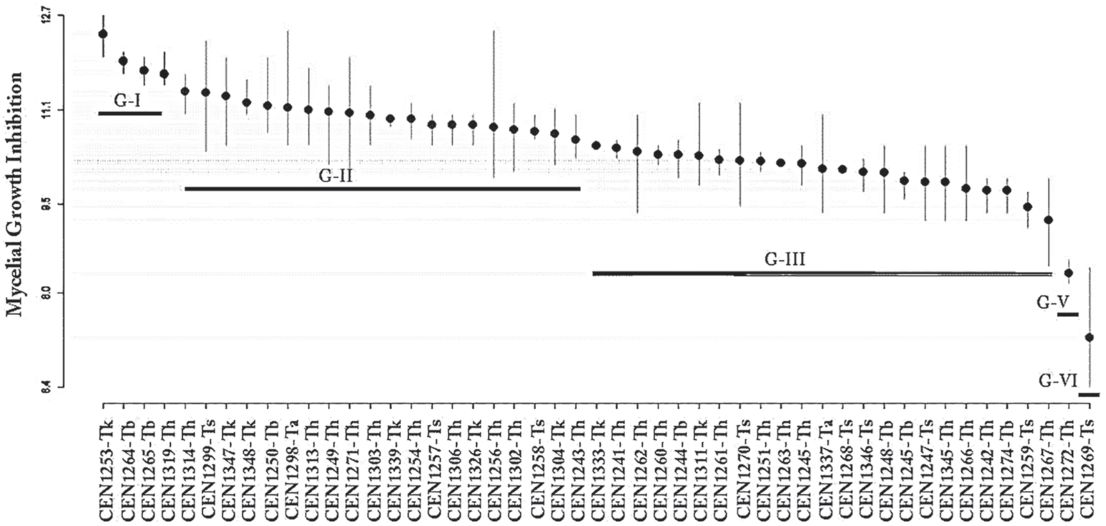

Figure 1. Average inhibition of mycelial growth of *Sclerotinia sclerotiorum* by *Trichoderma* spp., according to the Scott-Knott test.

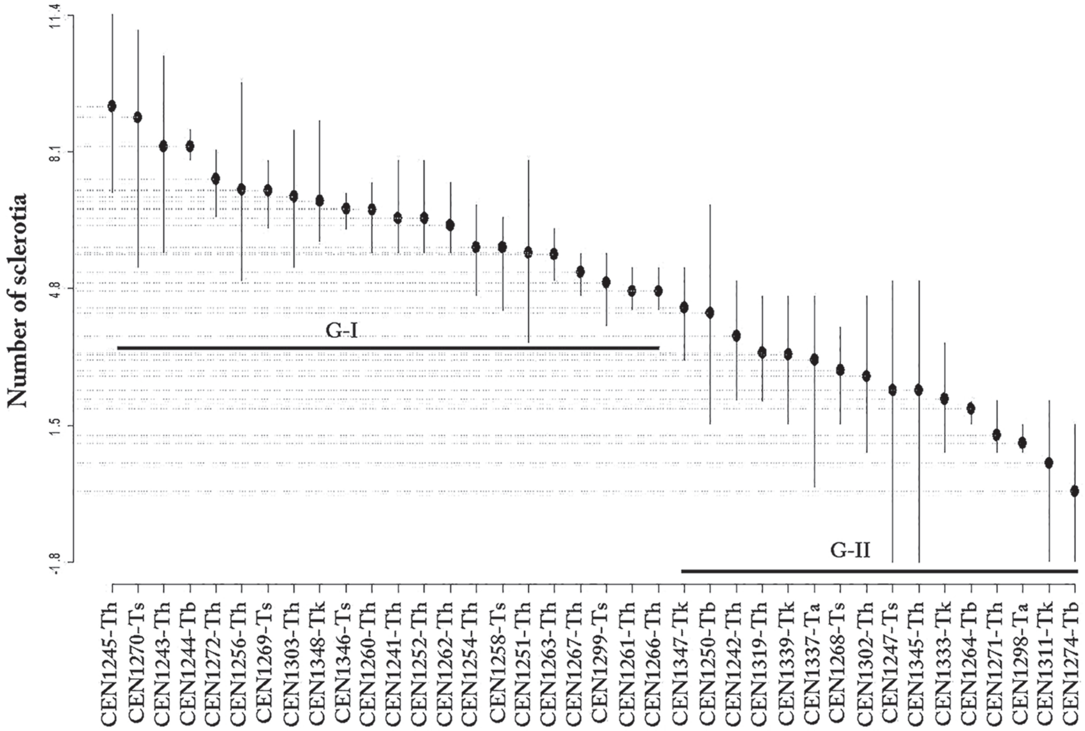

Figure 2. Mean production of *Ss* sclerotia in the presence of *Trichoderma*, according to the Scott-Knott test.

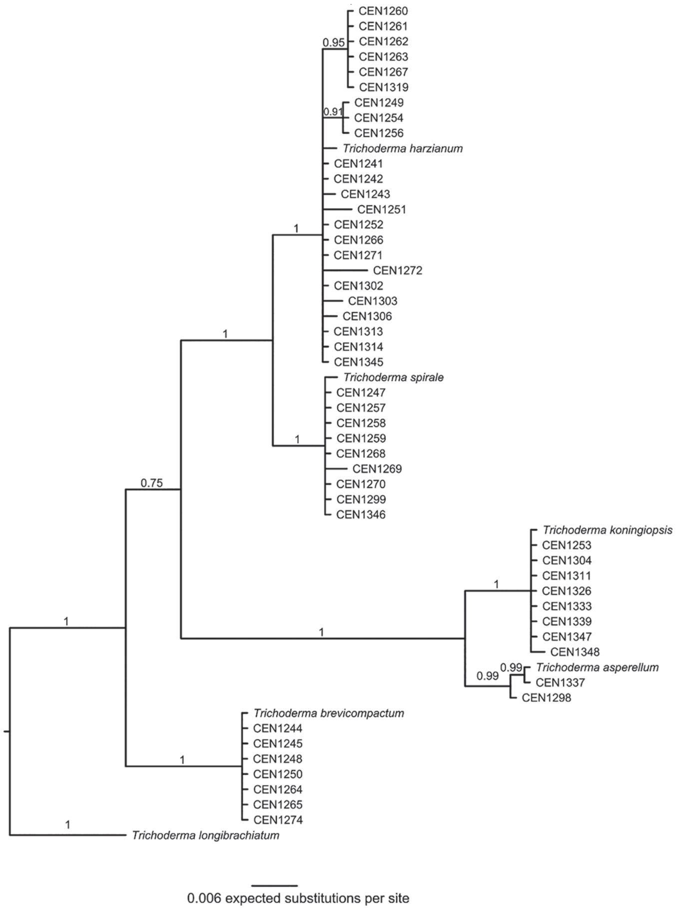

Figure 3. Bayesian phylogenetic tree based on ITS sequences from 49 *Trichoderma* isolates.

Discussion

In the present work, isolates with greater inhibitory potential were found to belong to the species *T. koningiopsis*, *T. brevicompactum* and *T. harzianum*. Experiments carried out by Amin et al. (2010) showed that *T. viride* was most inhibitory to *S. sclerotiorum* (66%), while two isolates of *T. harzinaum* exhibited MGI values of 56.57% and 38.12% respectively. Matroudi et al. (2009) used an *Ss* isolate from canola, to evaluate antagonistic *Trichoderma* spp., reporting variation between isolates. However, *T. atroviride* was the most efficient species and reduced growth between 85-93% compared to *T. harzianum* that presented an MGI of 70-80%. Likewise, there was no correlation between the source niche of the antagonist and its inhibitory potential *in vitro*. Here, the most efficient isolates were obtained from both the rhizosphere and rhizoplane as well from soil samples.

The ability to reduce the production of sclerotia is a valuable quality for the selection of *Trichoderma* isolates for biocontrol in view of the importance of the resistant structure on the survival of the pathogen in soil (Bianchini et al. 2005). Some isolates from this study (*T. koningiopsis*, *T. harzinaum* and *T. spirale*) completely inhibited the production of *Ss* sclerotia. In the same sense, Abdullah et al. (2008) observed inhibition of the formation of *Ss* sclerotia by *T. harzianum*, that decreased from 31.66 (treatment control) to 12.07 and 18.12 sclerotia. On the other hand, the most inhibitory isolate from the work reported by Amin et al. (2010) was *T. viride*.

Following the classification of Druzhinina et al. (2010), we observed some ITS sequence variation among the isolates clustering with *T. harzianum*. The formation of subgroups within *T. harzianum* is recognized, since it is regarded as a species complex (Kullnig et al. 2000). The second clade included *T. spirale* and these first two species have phylogenetic affinity (Druzhinina & Kubicek 2005), belonging to *Trichoderma* Section *Pachybasium* B. The third group (*T. koningiopsis* and *T. asperellum*) with related clades are classified in Section *Pachybasium A*. The fifth and final clade contained *T. brevicompactum* are placed in Section *Lutea* (http://www.isth.info/biodiversity/). Although the ITS sequences showed multiple hits in BLASTn for each query, these were clarified and confirmed by the *Trich*OKEY program which corroborates the phylogenetic analysis. The advantage of this online tool is that it was developed by taxonomists skilled in *Trichoderma*/*Hypocrea,* where the database is restricted to correctly identified sequences (Druzhinina et al. 2005).

A majority of the isolates were identified as *T. harzianum*, followed by *T. spirale*, *T. koningiopsis*, *T. brevicompactum* and *T. asperellum*, respectively. This distribution was expected, since it is known that *T. harzianum* is a global species, colonizing the most diverse substrates and ecological niches with a broad geographic distribution (Kubicek et al. 2008).

This study permitted the selection of isolates with good antagonistic potential for *S. sclerotiorum* and two of them: CEN1253 and CEN1265, identified as *T. koningiopsis* and *T. brevicompactum*, respectively, were considered to be promising for both biocontrol parameters (inhibition of mycelial growth and reduction of sclerotia). These fungi are stored in the Embrapa Fungi Collection for Biological Control and the information obtained in the experiments will be incorporated into the database of biological assets and genetic resources system (Allele, http://alelomicro.cenargen.embrapa.br/) for further studies required for the process of development of commercial products and procedures established by the Brazilian Ministry of Agriculture Livestock and Food Supply, until they are available for use by farmers.

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Ichthyofauna of streams from the lower Iguaçu River basin, Paraná State, Brazil

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Abstract: The drainages of the Iguaçu River basin, as well as the main channel of the river, have peculiar characteristics resulting from geomorphological processes in this area, including the formation of the Iguaçu River Falls. This slope in the lower portion of the basin is a biogeographical barrier to many organisms. In this study was inventoried the fish fauna of streams of the lower Iguaçu River basin, evaluating possible differences in the species composition upstream and downstream of this biogeographical barrier. Sampling were conducted between 2004 and 2013, in five streams, three upstream and two downstream of the Iguaçu River Falls, using electrofishing. The nMDS analysis was run to investigate possible groupings of similar fauna between the streams sampled. The scores of this test were tested as to the significance of groupings with the Hotelling T^2 test. The indicator value method (IndVal) was used to detect the distribution of species among the groups of the streams upstream and downstream of the Iguaçu River Falls. We collected 18,908 individuals of six orders, 11 families, and 40 species. Siluriformes and Characiformes had the highest species richness; Cyprinodontiformes presented the highest abundance. Considering the species recorded, 21 are considered natives to the Iguaçu River basin, including 15 endemic, wich were found only in streams upstream of the falls. Additional 18 species were verified only in the streams downstream of such barrier. Four species were common to both stretches. The axis 1 of the nMDS separated two groups: streams upstream (S1, S2 and S3) and streams downstream of the Iguaçu River Falls (S4 and S5). The indicator species analysis also indicated a distinction between the groups of streams, that were significantly different (Hotelling's $T^2 = 234.36$, $p < 0.0001$). The number of endemic species in the streams upstream of the Falls (15 spp.) evidences a significant effect of isolation promoted by the Iguaçu River Falls, and confirms the role of this barrier in the vicariant processes and endemism typical of this basin. These results emphasize the importance of conserving these ecosystems, once the extinction of species in this region means the irreversible loss of them.
Keywords: *Biogeographic barrier, endemics species, fish.*

LARENTIS, C., DELARIVA, R.L., GOMES, L.C., BAUMGARTNER, D., RAMOS, I.P., SEREIA, D.A.O. **Ictiofauna de riachos da bacia do baixo rio Iguaçu, estado do Paraná, Brasil**. Biota Neotropica. 16(3): e20150117. http://dx.doi.org/10.1590/1676-0611-BN-2015-0117

Resumo: As drenagens da bacia do rio Iguaçu, bem como o canal principal do rio, possuem características peculiares resultantes de processos geomorfológicos dessa área, incluindo a formação das Cataratas do rio Iguaçu. Esse desnível na porção inferior da bacia constitui uma barreira biogeográfica para muitos organismos. Neste trabalho inventariamos a fauna de peixes de riachos na porção do baixo rio Iguaçu, avaliando possíveis diferenças na composição de espécies acima e abaixo dessa barreira biogeográfica. Entre 2004 e 2013, foram amostrados cinco riachos, sendo três localizados acima das Cataratas do Iguaçu e dois abaixo, por meio de pesca elétrica. Para investigar possíveis padrões de agrupamentos de fauna similares entre os riachos amostrados foi realizada uma análise nMDS, cujos escores foram testados quanto à significância dos agrupamentos utilizando o teste de T^2 de Hotelling. O valor indicador (IndVal) foi usado para detectar a distribuição das espécies entre os grupos de riachos. Foram amostrados 18.908 indivíduos, pertencentes a seis ordens, 11 famílias e 40 espécies. As ordens Siluriformes e Characiformes destacaram-se em riqueza de espécies; Cyprinodontiformes apresentou maior abundância. Do total de espécies amostradas, 21 são consideradas nativas da bacia do rio Iguaçu, sendo 15 delas endêmicas e registradas somente nos riachos acima das Cataratas do rio Iguaçu. Outras 18 espécies foram registradas somente nos riachos que desembocam abaixo das cataratas. Quatro espécies foram comuns em ambos os ambientes. A dimensão do eixo 1 da análise nMDS resultou na separação de dois grupos: riachos acima (S1, S2 e S3) e riachos abaixo das cataratas (S4 e S5). A análise de espécies indicadoras também indicou a distinção entre os grupos de riachos, sendo que esses agrupamentos foram significativamente diferentes (Hotelling's $T^2 = 234.36$, $p < 0.0001$). O número de espécies endêmicas registrado nos riachos acima das cataratas (15 spp.), demonstra um significativo efeito de isolamento da fauna proporcionado pelas Cataratas do rio Iguaçu e confirmam o papel dessa barreira nos processos vicariantes e endemismos característicos dessa bacia. Esses resultados ressaltam a importância da preservação desses ecossistemas, pois a extinção de espécies nessa região significa a perda global dessas entidades biológicas.

Palavras-chave: *Barreira biogeográfica, espécies endêmicas, peixes.*

Introduction

The Paraná State, Southern Brazil, has 16 basins (Parolin et al. 2010), including the Iguaçu River basin, formed by the longest river in the Paraná State with approximately 1,080 km (Garavello et al. 2012). The formation of its basin dates back to the Mesozoic era and the beginning of the Paleozoic and was associated with staggered movements of uplift of the Serra do Mar (Parolin et al. 2010). Considering the confluence of the Atuba and Iraí rivers as its origin, the Iguaçu River runs in the east-west direction from its headwaters in the Serra do Mar to its mouth on the Paraná River (Baumgartner et al. 2012). Along its course, it crosses different geological and geomorphological units, with an altitude range of 830 m, considered as a typical plateau river, with numerous rapids and falls. Among these, stands out the Iguaçu River Falls with 82 meters, in addition to other hundreds of waterfalls in the basin, which result from the geological structure caused by basaltic spills. The Iguaçu River and its tributaries features a fauna of small-sized fish, with a high degree of endemism (Vicari et al. 2006), possibly due to the geographical isolation given by the Iguaçu River Falls that isolated this river from the lower Paraná River basin (Baumgartner et al. 2012).

Biogeographical barriers are a key factor in the composition of regional faunas and especially in promoting endemism (Rahel 2007). Besides, the presence of barriers, such as waterfalls, promotes geographical isolation of habitats that increase the speciation rate in these isolated environments (Cracraft 1982), mainly by limiting gene flow, and promoting the diversification of species (Meeuwig et al. 2010). These barriers determine the degree of connectivity of water bodies within the basin, directing the speciation process by influencing the availability of habitats for aquatic species (Dias et al. 2012).

The geological event that formed the Iguaçu River Falls back to the Cretaceous period 130 million years ago (Parolin et al. 2010), which has possibly generated a cladogenic event for most fish species of the Iguaçu River. According to Rahel (2007), the Iguaçu River Falls are classified as a barrier level three for the movement of fish, that is, they promote the isolation of the fish fauna within a basin, ensuring that these species do not colonize other regions unless by human intervention.

In the Iguaçu River basin, 75% of fish species are endemic (Zawadzki et al. 1999), but this fauna has undergone changes mainly caused by human activities (Baumgartner et al. 2006). Rapid population growth leads to urban sprawl and expansion of agriculture increasingly closer to freshwater environments, affecting negatively the physical and biological structure of these environments (Cunico et al. 2006, Pereira et al. 2014). In this way, besides the threat that human activities cause, there are few studies on streams in the Paraná State (Pavanelli & Caramaschi 2003, Cunico et al. 2006, 2009, Galves et al. 2007, Araujo et al. 2011, Daga et al. 2012, Delariva & Silva 2013). In the Iguaçu River basin, stand out the studies conducted in the upper reach of the basin (Suzuki et al. 1997, Vitule & Abilhoa 2003) and surveys of species in streams of the lower Iguaçu River (Suzuki et al. 1997, Bifi et al. 2006, Baumgartner et al. 2012).

Most rivers and streams forming the Iguaçu River basin are located upstream of the Iguaçu River Falls, but some streams have connection downstream of this barrier, which is located about 18 km upstream of the mouth of the Iguaçu River into the lower Paraná River basin. Although belonging to the Iguaçu River basin, these streams may have been affected by the same cladogenic event and hold a fish fauna similar to that observed for the Paraná River basin. In this sense, aiming to investigate the fish fauna composition of streams upstream and downstream of the Iguaçu River Falls, this study surveyed the fish fauna in first- and second-order streams of the lower Iguaçu River basin. Thus, this study provides a list of species, showing the occurrence and expansion of distribution and identified possible dissimilarities between the fish faunas located in these sections (up- and downstream of the falls).

Material and Methods

1. *Study Area*

The Iguaçu River basin encompasses an area of 55,108.04 km² in the Paraná State, covering 116 municipalities (Parolin et al. 2010) and is subdivided into three hydrographical units: upper, middle, and lower Iguaçu River. In the lower stretch is located the Iguaçu River Falls, in the municipality of Foz do Iguaçu, PR, considered the largest falls in volume of water on the planet, which fall through a deep crevice, with an gap of 72 meters and an average volume of 1.551 m³/second (SEMA 2013). On the Brazilian side of the Iguaçu River Falls and vicinity of the Iguaçu River in this region, is situated the Iguaçu National Park, a conservation unit essential for the country's biodiversity, because it protects the Iguaçu River Falls and constitutes the largest remnant area of preservation of semideciduous forest of the Paraná State. In the lower Iguaçu River region, the main economic activity is agriculture, especially the cultivation of soybeans and corn, and pastures, which was the prevailing scenario in the vicinity of the streams sampled.

The basins of the studied streams drain, directly or indirectly, into the Iguaçu River, however, flow into upstream (S1, S2, S3) and downstream (S4 and S5) of the Iguaçu River Falls (Figure 1). So, they are separated and susceptible to geographic isolation generated by this natural barrier.

2. *Fish sampling*

The sampling of the fish fauna was performed between 2004 and 2013 in five streams, totaling 11 sampling sites. Samplings were conducted in different periods: Padres River (S1) - quarterly at two sites, between July 2004 and December 2005, with two samplings in 2006 (March and May); Cascavel River (S2) - quarterly samples at two sites between August 2011 and February 2012; Jumelo Stream (S3) - an unique site on the headwaters in September and November 2011, March and July 2012 and July 2013; Poço Preto Stream (S4) and Arroio Dourado Stream (S5) - sampled at three sites every four months between May 2010 and April 2011. The main physical characteristics and geographical coordinates are shown in Table 1. Fish were collected by electrofishing technique with three paces of 40 minute in 50 meters length at each reach. The electrofishing equipment was powered by a portable generator (Honda, 2.5 kW, 220 V, 3 - 4 A) connected to a DC transformer, with two electrified net rings (anode and cathode). Output voltage varied from 400 to 600 V, and we installed blocking nets (mesh size 5 mm) at the ends of each reach to prevent the escape of fish. After sampling, specimens were sacrificed in a solution of clove oil (Eugenol, 2 drops per liter; *cf.* American Veterinary Medical Association 2001), and fixed in 10% formalin and then preserved in 70% alcohol.

Specimens were identified according to Baumgartner et al. (2012), Garavello et al. (2012), Garavello & Sampaio (2010), and Graça & Pavanelli (2007). Voucher specimens were deposited at the fish collection of Nupélia (Núcleo de Pesquisas em Limnologia, Ictiologia

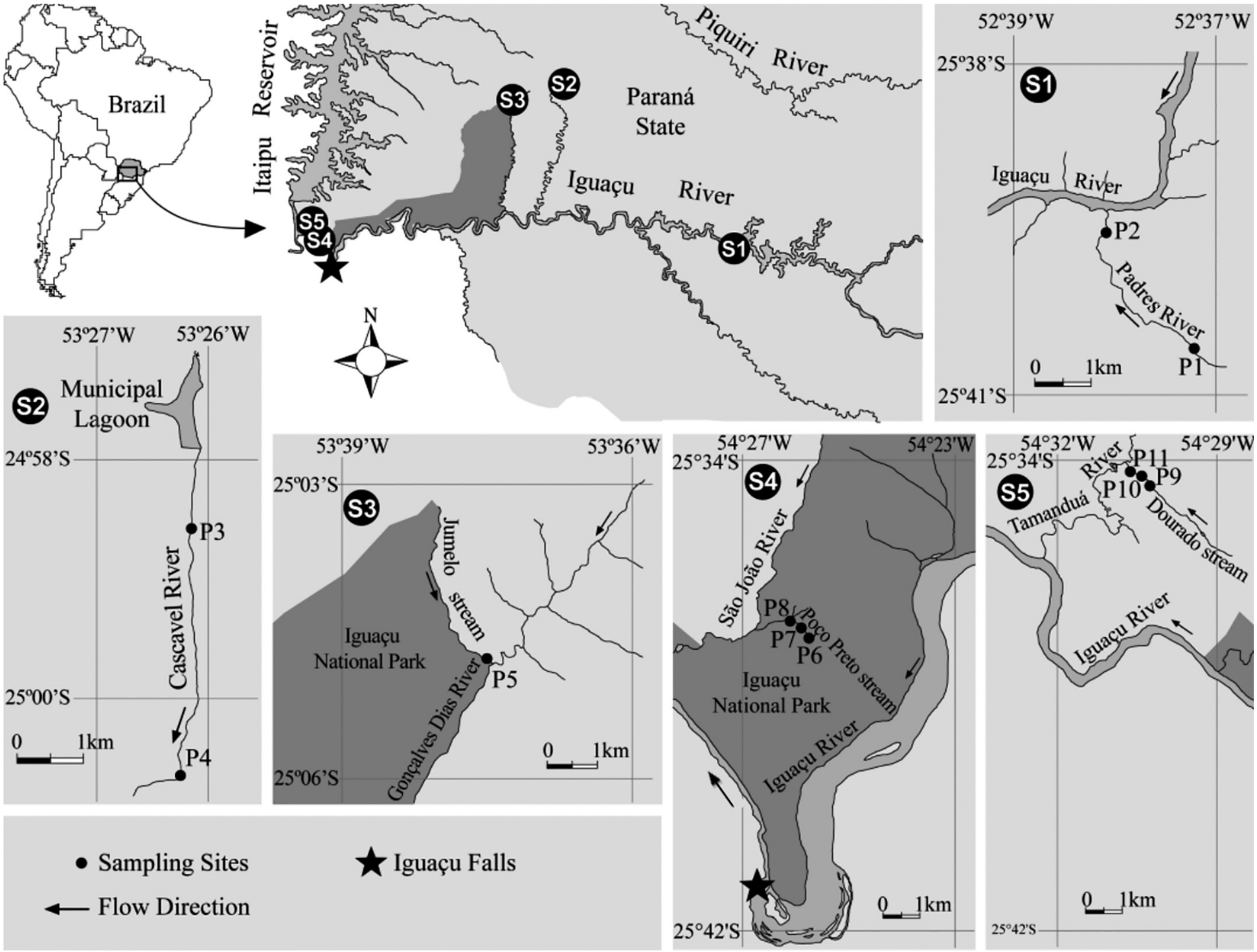

Figure 1. Streams studied in the lower Iguaçu River basin, Paraná State, Brazil.

Table 1. Physical characteristics of the sampled sites in Iguaçu River basin, Paraná State. Brazil. S1, S2 until S5 = Stream's number. P1, P2, P3 until P11 = sampled portions.

SITES	COORDINATES	WIDTH (m)	DEPTH (m)	PREDOMINANT SUBSTRATE	RIPARIAN VEGETATION	PHYSIOGRAPHY CHANNEL	OBSERVATIONS
Padres River (S1)	P1-25°40'34.81"S 52°37'6.05"W P2-25°39'34.777"S 52°38'2.514"W	P1-2.00-4.10 P2-2.00- 4.10	P1-0.10-0.28 P2-0.10-0.55	Muddy alternated with argil in the lower stretch, rubble in the upper stretch, and small amount of shelters.	Variables ranges of trees and shrubs	Low-speed stream in both of the points.	Wide range of soil devoid of vegetation, because is within of rural areas.
Cascavel River (S2)	P3-24°58'33.57"S 53°26'6.41"W P4-25°0'32.10"S 53°26'11.10"W	P3-1.00-2.50 P4-1.00-3.00	P3-0.30-0.60 P4-0.40-0.70	Rubble predominant in both of the points.	P3- present in only one of the edges; P4- present in both of the banks	Low-speed stream in all of the sites sampled.	The sites sampled are within of the urban areas.
Jumelo (S3) (headwater stretch)	P5-25°04'46.62"S 53°37'26.42" W	P5-2.00-4.00	P5-0.20-1.00	Rocky, rubble, pebble and silt.	Right bank with Riparian vegetation preserved. Left bank with range of trees and shrubs between 5 and 10 m.	Rapids with mean depth of 0.30 m, and pools of up to 1.00 m in depth.	The right bank belongs to Iguaçu National Park, a area of preservation. The left bank is within of rural areas.
Poço Preto (S4)	P6-25°36'48.04"S 54°25'54.09"W P7-25°37'19.3"S 54°26'52.6"W P8-25°37'40.5"S 54°26'53"W	P6-1.00 -1.89 P7-1.60-2.88 P8-2.70-3.41	P6-0.20-0.49 P7-0.55-0.90 P8-2.30-2.95	Partially rocky substrate with different grain textures.	Riparian vegetation on both banks composed of pionners formations of rainforest river influence.	Predominance of rapids.	Located inside of a area of preservation (Iguaçu National Park).
Arroio Dourado (S5)	P9-25°34'19.3"S 54°30'16.4"W P10- 25°34'16"S 54°30'19.2"W P11- 25°34'14.8"S 54°30'24.1"W	P9-1.80-2.24 P10- 2.00- 2.96 P11- 2.50- 3.10	P9-0.10-0.29 P10- 0.22- 0.39 P11- 0.10- 0.15	Partially rocky substrate with different grain textures.	Riparian vegetation on both banks composed of pionners formations of rainforest river influence.	Predominance of rapids.	Soil around mainly occupied by pastures.

e Aquicultura), Universidade Estadual de Maringá- UEM, and at the fish collection of Gerpel (Grupo de Pesquisas em Recursos Pesqueiros e Limnologia) - Universidade Estadual do Oeste do Paraná - UNIOESTE.

3. *Data Analysis*

To identify possible groupings of similar fauna between streams, we applied a non-metric multidimensional scaling analysis (nMDS) to summarize the composition of fish assemblages using the Jaccard distance and following the general nMDS procedure proposed by McCune & Grace (2002). We obtained a confidence index (Stress), which determines the degree of proximity of the graphical representation to the real data. Stress values below 0.20 allow potential two-dimensional (2D) use of the nMDS. Data were randomized 100 times, and the stability criterion was 0.005 standard deviations in the stress after 200 iterations. This analysis was carried out using the numerical abundance matrix of each sampling site and month.

To test the null hypothesis of no difference in composition of the fish assemblage between sites upstream and downstream of the falls summarized by the nMDS, we used a multivariate pairwise comparisons between two groups (upstream and downstream of the falls), using the Hotelling T^2 test. This analysis indicates whether a multidimensional sample belongs to a particular multidimensional group. As closer sites have a higher chance to be similar, we applied a Euclidean distance on UTM (site coordinates), obtaining proportional geographic distances between them. We used a Mantel's test on the distances between fish assemblage abundances matrix (nMDS) and geographic distances matrix with the objective of controlling possible geographical effects. Both results were reported based on 10.000 permutations.

The indicator value method (IndVal; Dufrêne & Legendre 1997) was used to detect how differently each species is distributed among the groups of the streams upstream and downstream of the Iguaçu River Falls. The indicator value of a species varies from 0 to 100, and it reaches its maximum value when all individuals of a species occur in all sites within a single nMDS group.

All analyses were run in the software Past (Hammer et al. 2001), except the indicator value method, performed in the PCOrd® 6.0 (McCune & Mefford 2011).

Results

In total, we collected 18,908 individuals belonging to six orders, 11 families, and 40 species (Table 2). Siluriformes was the most representative with 50% of the species richness, followed by Characiformes (31%). As to numerical abundance, the most representative was Cyprinodontiformes (64%), followed by Characiformes (23%) and Siluriformes (11%). Characidae and Loricariidae were the highest in number of species (22% each). The largest-sized species was *Synbranchus* cf. *marmoratus* Bloch, 1795 (53.7 cm total length), and the smallest specimen was *Phalloceros harpagos* Lucinda, 2008, with standard length (SL) of 0.6 cm.

Among the species sampled, 12 are possibly not described, already that they presented morphological characters different from their congeners currently described for the region. In such cases, *Characidium* sp. 2, *Trichomycterus* sp. 1, *Trichomycterus* sp. 2, and *Ancistrus* sp. were *sensu* Baumgartner et al. (2012), and *Characidium* sp. 1 was *sensu* Graça & Pavanelli (2007). Regarding the origin of the species, 21 were native

Table 2. List of fish species and their respective abundances at sampling stations in Iguaçu River basin. Systematic positions were based on Nelson (2006) to all orders, and Siluriformes families; Reis et al. (2003) to others families, except Characidae that follow Mirande (2009). The column "Origin" refers to species classified in native (N) and non-native (NN) to the of lower Paraná River and Iguaçu River basins. S = sites of sampling. NUP - vouchers specimens deposited in Fish Collection of Núcleo de Pesquisas em Limnologia, Ictiologia e Aquicultura - Nupélia; and CIG - vouchers specimens deposited in Fish Collection of Grupo de Pesquisas em Recursos Pesqueiros e Limnologia - Gerpel.

Taxon	**Common name**	**Origin**								
		Lower Paraná River	**Iguaçu River**	**S1**	**S2**	**S3**	**S4**	**S5**	**Total**	**Voucher specimens**
CHARACIFORMES										
Crenuchidae										
Characidium sp. 1		N	NN				x	x	42	CIG 1410, CIG 1422
Characidium sp. 2		NN	N	x					27	NUP 2110
Characidae										
Astyanax sp. 1		N	NN				x	x	621	CIG 2277
Astyanax sp. 2		N	NN					x	77	CIG 2259, CIG 2283
Astyanax sp. 3		N	NN					x	154	CIG 2284
Astyanax lacustris (Lütken, 1875) ex *Astyanax altiparanae* Garutti & Britski, 2000 (Lucena & Soares, 2016)	"lambari-do-rabo-amarelo"	N	N				x	x	30	CIG 1480
Astyanax bifasciatus Garavello & Sampaio, 2010	"lambari-do-rabo-vermelho"	NN	N	x	x	x			3764	CIG 2239, NUP 16945
Astyanax dissimilis Garavello & Sampaio, 2010	"lambari"	NN	N			x			7	NUP 16940
Astyanax serratus Garavello & Sampaio, 2010	"lambari"	NN	N			x			11	CIG 2381
Bryconamericus iheringii (Boulenger, 1887)	"lambarizinho"	N	NN					x	13	CIG 1445
Oligosarcus longirostris Menezes & Géry, 1983	"saicanga"	NN	N	x					1	NUP 11856
Bryconamericus pyahu Azpelicueta, Casciotta & Almirón, 2003	"lambarizinho"	NN	N			x			2	NUP 12089
Erythrinidae										
Hoplias aff. *malabaricus* (Bloch, 1794)	"traíra"	N	N	x	x				15	CIG 2242
Erythrinus erythrinus (Block & Schneider, 1801)	"jejú"	N	NN					x	2	CIG 1446
SILURIFORMES										
Trichomycteridae										
Trichomycterus sp. 1	"candiru"	NN	N			x			317	NUP 12660
Trichomycterus sp. 2	"candiru"	NN	N			x			83	NUP 12661
Trichomycterus sp. 3	"candiru"	N	NN				x	x	126	CIG 1416, CIG 1432
Trichomycterus davisi (Haseman, 1911)	"candiru"	NN	N	x					186	CIG 2241, CIG 2245
Trichomycterus taroba Wosiacki & Garavello, 2004	"candiru"	N	N	x					169	NUP 3125
Callichthyidae										
Corydoras sp.	"limpa-vidro"	N	NN				x		151	NUP 14261
Loricariidae										
Ancistrus sp.	"cascudo-roseta"	NN	N	x					30	NUP 4140, NUP 4489

Continued Table 2.

Taxon	Common name	Origin							Total	Voucher specimens
		Lower Paraná River	**Iguaçu River**	**S1**	**S2**	**S3**	**S4**	**S5**		
Ancistrus mullerae Bifi, Pavanelli & Zawadzki, 2009	"cascudo-roseta"	NN	N			x			489	NUP 16942
Hisonotus sp.	"cascudinho limpa-vidro"	NN	N				x	x	43	CIG 2261
Hypostomus ancistroides (Ihering, 1911)	"cascudo"	N	NN					x	2	CIG 1448
Hypostomus albopunctatus (Regan, 1908)	"cascudo"	N	N				x	x	15	CIG 1444
Hypostomus derbyi (Haseman, 1911)	"cascudo"	NN	N	x	x				129	NUP 4134
Hypostomus myersi (Gosline, 1947)	"cascudo"	NN	N	x					4	NUP 6239
Hypostomus sp.	"cascudo"	N	NN					x	4	CIG 1420, CIG 1485
Heptapteridae										
Heptapterus mustelinus (Valenciennes, 1835)	"bagrinho, bagre-pedra"	N	NN					x	46	CIG 1439
Rhamdia branneri Haseman, 1911	"bagre", "jundiá"	NN	N	x		x			23	CIG 2258, NUP 16941
Rhamdia voulezi Haseman, 1911	"bagre", "jundiá"	NN	N	x		x			54	NUP 11185, NUP 5678
Rhamdia aff. *quelen* (Quoy & Gaimard, 1824)	"bagre", "jundiá"	N	NN				x	x	119	CIG 1475, CIG 1418
GYMNOTIFORMES										
Gymnotidae										
Gymnotus sylvius Albert & Fernandes-Matioli, 1999	"morenita"	NN	NN		x			x	123	CIG 1442
Gymnotus inaequilabiatus (Valenciennes, 1839)	"morenita"	N	NN					x	10	CIG 1441
Gymnotus pantanal Fernandes, Albert, Daniel-Silva, Lopes, Crampton & Almeida-Toledo, 2005	"morenita"	N	NN					x	9	CIG 1451
CYPRINODONTIFORMES										
Poeciliidae										
Phalloceros harpagos Lucinda, 2008	"barrigudinho", "guaru"	N	N	x	x	x	x		11940	CIG 1427, NUP16943
Poecilia reticulata Peters, 1859	"barrigudinho", "guaru"	NN	NN		x				10	CIG 2272
SYNBRANCHIFORMES										
Synbranchidae										
Synbranchus cf. *marmoratus* Bloch, 1795	"muçum"	N	NN		x		x	x	16	CIG 1449, CIG 1434
PERCIFORMES										
Cichlidae										
Crenicichla britskii Kullander, 1982	"joaninha"	N	NN				x	x	21	CIG 1483
Oreochromis niloticus (Linnaeus, 1758)	"tilápia"	NN	NN		x			x	23	CIG 2273
Total number				15826	262	1056	1270	494	18908	
Richness									**40**	

to the Iguaçu River basin (Table 2). In terms of occurrence, 27 species have already been recorded in previous studies and other 13 are new records for the Iguaçu River basin (*Astyanax* sp. 3, *Bryconamericus iheringii* (Boulenger, 1887), *Hoplias* aff. *Malabaricus* (Bloch, 1794), *Erythrinus erythrinus* (Block & Schneider, 1801), *Trichomycterus* sp. 3, *Corydoras* sp., *Hypostomus ancistroides* (Ihering, 1911), *Hypostomus* sp., *Heptapterus mustelinus* (Valenciennes, 1835), *Rhamdia* aff. *quelen* (Quoy & Gaimard, 1824), *Gymnotus pantanal* Fernandes, Albert, Daniel-Silva, Lopes, Crampton & Almeida-Toledo, 2005, *Poecilia reticulata* Peters, 1859, and *Crenicichla britskii* Kullander, 1982). Of the total species sampled, 15 were endemic to the Iguaçu River basin, and observed only in streams which discharge into upstream of the Iguaçu River Falls. Still, 18 species were sampled only in streams which flow into downstream of this barrier. Only four species were common to both environments (*Gymnotus sylvius* Albert & Fernandes-Matioli, 1999, *Oreochromis niloticus* (Linnaeus, 1758), *Phalloceros harpagos*, and *Synbranchus* cf. *marmoratus*).

In the stream S1, we collected 12 species, where the most abundant in number was *Phalloceros harpagos* and in biomass was *Astyanax bifasciatus* Garavello & Sampaio, 2010. The high numerical abundance of *Phalloceros harpagos* in this stream gave greater prominence to Cyprinodontiformes. Of the 12 species recorded in this stream, nine are endemic and the other three (*Hoplias* aff. *malabaricus*, *Trichomycterus davisi* (Haseman, 1911), and *Phalloceros harpagos*) are considered autochthones species (with distribution in other basins). Among the endemic species, two are considered not described (*Ancistrus* sp. e *Characidium* sp. 2) and are in the process of description (Baumgartner, D., pers. comm.).

In the stream S2, we sampled eight species, wherein *Astyanax bifasciatus* was the most abundant in number (45%), and *Gymnotus sylvius* showed the highest biomass. This environment contained only one species endemic to the Iguaçu basin (*Astyanax bifasciatus*). Among the others, two are native with distribution in other basins (*Hoplias* aff. *malabaricus* and *Phalloceros harpagos*) and four were considered introduced (*Gymnotus sylvius*, *Poecilia reticulata*, *Oreochromis niloticus*, and *Synbranchus* cf. *marmoratus*).

For the stream S3, we identified 10 species, including *Ancistrus mullerae* Bifi, Pavanelli & Zawadzki, 2009, a species endemic to the lower Iguaçu River, with the highest abundance, both in number and biomass (46% and 37%, respectively). In this study, it was only registered in this stream. All species caught in this stream were considered endemic, except for *Phalloceros harpagos*, with wide distribution in other basins. Two species (*Trichomycterus* sp. 1 e *Trichomycterus* sp. 2) are still possibly not described (*sensu* Baumgartner et al. 2012).

In the stream S4, we captured 11 species: *Astyanax* sp. 1 presented higher abundances both number and biomass (48% and 41%, respectively). Of the species caught, five were identified only to the genus level and probably they are still not described, because they do not present the same morphological characters of other already found in the literature (*Astyanax*, *Characidium*, *Corydoras*, *Hisonotus*, and *Trichomycterus*). It should be noted that *Corydoras* sp. has been recorded exclusively in this stream (Tencatt, L. pers. comm.).

In the stream S5, we sampled 21 species, and observed high numerical abundance of the *Astyanax* sp. 3 and higher biomass of *Gymnotus sylvius* (28% and 24%, respectively). In this stream, as in the stream S4, seven species have possibly not been described. Six species were considered as non-native, one exotic (*Oreochromis niloticus*) and five allochthonous (*Bryconamericus iheringii*, *Erythrinus erythrinus*, *Gymnotus inaequilabiaus* (Valenciennes, 1839), *Gymnotus sylvius* and *Gymnotus pantanal*) (Graça & Pavanelli 2007, Langeani et al. 2007, Baumgartner et al. 2012). Six species (*Astyanax altiparanae* Garutti & Britski, 2000, *Hypostomus albopunctatus* (Regan, 1908), *Gymnotus sylvius*, *Gymnotus inaequilabiatus*, *Synbranchus* cf. *marmoratus* and *Oreochromis niloticus*) were previously recorded in the lower Iguaçu River (*sensu* Baumgartner et al. 2012). Nevertheless, none of them is considered endemic to this basin. Only two species are native to the Iguaçu River, but widely found in the Paraná River.

The NMDS summarized the composition of the fish assemblages and separated the studied streams (Figure 2). After 200 iterations, the stability criterion was met with a final stress of 0.12 (Monte Carlo test, $p < 0.004$) for the two-dimensional solution. The proportion of variance represented by each axis, based on the R^2 between distance in the ordination space and distance in the original space, was 0.50 for the axis 1 and 0.16 for the axis 2. The distribution of points along the axis 1 segregated the streams upstream and downstream of the Iguaçu River Falls at the multidimensional space, suggesting that the composition of fish assemblages were highly affected by the geographical barrier (Iguaçu River Falls). We found significant differences in the composition of fish assemblages between the group of streams upstream and downstream of the falls (Hotelling's T^2 = 234.36, $p < 0.0001$). These results are corroborated by the Mantel test that evidenced no correlation between the similarity matrix of fish fauna composition and geographical distances among the sampled streams (Mantel r = 0.024, p = 0.5), demonstrating that the clusters formed in the nMDS axis 1 were not influenced by spatial autocorrelation arising from the proximity of streams.

In general, the indicator species analysis also indicated a distinction between the groups of streams. The streams upstream and downstream were characterized by different sets of indicator species (IndVal; Table 3). Six species characterized the streams upstream of the falls, except for *Phalloceros harpagos*, all these species were endemic to the Iguaçu basin. On the other hand, the streams flow into downstream of such biogeographical barrier were characterized by 15 indicator species, and with exception of *Synbranchus* cf. *marmoratus*, all were exclusive to these streams.

Discussion

The number of species observed in this study (40 spp.) can be considered high, especially if evaluated some issues: (i) the number of species recorded for other water bodies with similar size in the Neotropics (Bertaco 2009, Couto & Aquino 2011, Marceniuk et al. 2011, Oyakawa & Menezes 2011, Cetra et al. 2012, Daga et al. 2012), as well in other streams in the Paraná State (Araújo et al. 2011, Delariva & Silva 2013, Gubiani et al. 2010); (ii) the proportion of species registered herein represents 37.7% of the total number of species recorded for the Iguaçu River basin (106 spp.), including those observed in the main channel of the river (Baumgartner et al. 2012). This high number of species is because two streams (S4 and S5) belong to the Iguaçu River basin but flow into it downstream of the Iguaçu River Falls, thus have greater connection and exchange with the fish fauna of the Paraná River basin; (iii) besides the considerable number of species that have not yet been described. Moreover, most of the records held in the Iguaçu River basin compiled in Baumgartner et al. (2012) refers to surveys in the main channel of the Iguaçu River, with the use of other fishing gears, *i.e.*, the majority of streams in this basin have never been sampled. This condition combined with the geomorphological characteristics of the basin provides the record of species of which there is no confirmation if they are only new records or are still unknown to science.

The highest richness of the Siluriformes and Characiformes, as well as the families Characidae and Loricariidae, is a common observation in streams in the Neotropics (Lowe-McConnell 1999, Shibatta et al. 2002, Buckup et al. 2007; Langeani et al. 2007), and corroborates studies in small streams in other basins of the Paraná State (Galves et al. 2007, Cunico et al. 2009; Gubiani et al. 2010, Araújo et al. 2011, Pagotto et al. 2012, Delariva & Silva 2013). However, for the lower Iguaçu River, were registered 11 *Astyanax* spp., popularly known as "lambari", of which eight species are endemic, and two possibly endemic to this basin

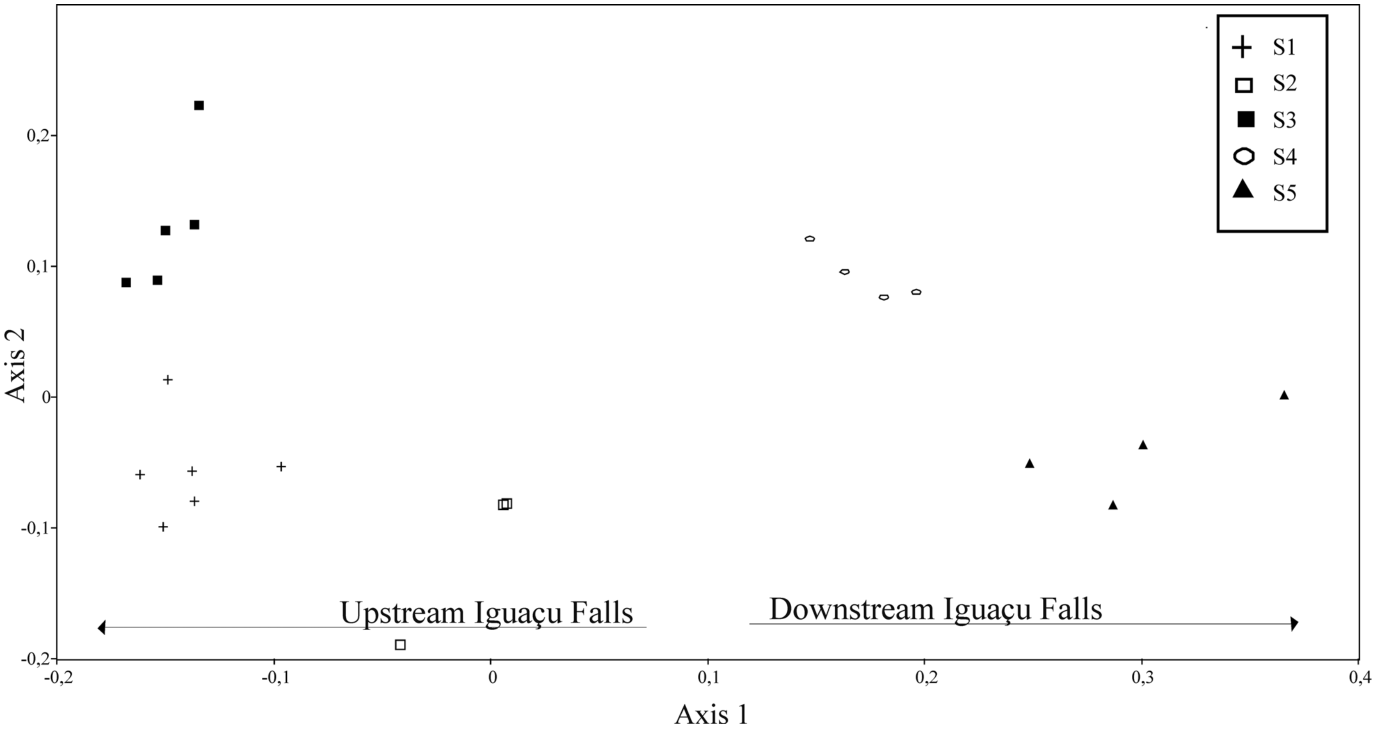

Figure 2. Non-metric multidimensional scaling (nMDS) ordination of the fish assemblages in the streams sampled in lower Iguaçu River basin, Paraná State, Brazil.

Table 3. Summary of the indicator species analysis: relative abundance, relative frequency, and indicator value for the groups of streams (up and downstream of Iguaçu River Falls) studied in lower Iguaçu River basin, Paraná State, Brazil. Only species with significant values are shown. Values in boldface indicate significant indicator values ($p < 0.05$, Monte Carlo test).

Species	Groups	Relative abundance		Relative frequency		Indicators values	
Astyanax bifasciatus	1	100	0	94	0	**94**	0
Trichomycterus davisi	1	100	0	50	0	**50**	0
Trichomycterus taroba	1	100	0	50	0	**50**	0
Hypostomus derbyi	1	100	0	56	0	**56**	0
Rhamdia voulezi	1	100	0	50	0	**50**	0
Phalloceros harpagos	1	97	3	94	50	**91**	2
Characidium sp. 1	2	0	100	0	63	0	**63**
Astyanax sp. 1	2	0	100	0	75	0	**75**
Astyanax sp. 2	2	0	100	0	50	0	**50**
Astyanax sp. 3	2	0	100	0	50	0	**50**
Astyanax lacustris	2	0	100	0	50	0	**50**
Briconamericus iheringii	2	0	100	0	50	0	**50**
Trichomycterus sp. 3	2	0	100	0	63	0	**63**
Corydoras sp.	2	0	100	0	50	0	**50**
Hisonotus sp.	2	0	100	0	63	0	**63**
Hypostomus albopunctatus	2	0	100	0	63	0	**63**
Heptapterus mustelinus	2	0	100	0	50	0	**50**
Rhamdia aff. *quelen*	2	0	100	0	100	0	**100**
Gymnotus pantanal	2	0	100	0	38	0	**38**
Synbranchus cf. *marmoratus*	2	7	93	13	75	1	**70**
Crenicichla britskii	2	0	100	0	50	0	**50**

(Baumgartner et al. 2012). In our study, we recorded four *Astyanax* spp., three possibly endemic. Other three species are possibly new, totaling seven *Astyanax* spp., which explains the high contribution of Characidae in our study.

Most of the species verified in this study is considered small size, which is expected to small water bodies (Bifi et al. 2006, Araújo et al. 2011, Cionek et al. 2012). According to Castro (1999), this pattern is shared by the fauna of South American streams and is mainly related to historical and evolutionary factors. It is noteworthy that a high number of small-sized species is even more evident in the fish fauna of the Iguaçu basin, even considering the species occurring in the main channel of the river. According to Garavello et al. (1997), the scarcity of rheophilic or large migratory species in this basin can be attributed to the formation of the Iguaçu River Falls, with an abrupt elevation, which interrupted the faunal exchange with the Paraná River basin.

In general, a high degree of endemism is reported to streams (Castro 1999, Casatti 2010). However, it was more evident in the fauna of the Iguaçu River, with about 70% of endemic fish (Baumgartner et al. 2012). The formation of the falls isolated the upper from the lower reach of the Iguaçu River basin (Parolin et al. 2010), providing different scenarios for the evolution of species. This resulted in species with unique characteristics in the upper section. The indicator species for the two groups of streams corroborate this hypothesis, especially, because all indicator species of the group of streams upstream of the falls are endemic to the Iguaçu River. On the other hand, downstream of the falls was observed a higher number of indicator species, wherein, except *Synbranchus* cf. *marmoratus*, all were common to the fauna of the Paraná River. The occurrence of the distinct species in downstream indicates that the streams S4 and S5 remained connected below of the falls. In this way, although these streams belong to the Iguaçu River basin, its fish fauna evolved following similar patterns those observed for the Paraná River basin. This result strongly supports the hypothesis of distinction between the stream fish fauna upstream and downstream of the Iguaçu River Falls. Accordingly, corroborates with the already postulated effects of biogeographical barriers in the differentiation of the fish fauna and especially in endemism (Olden et al. 2010, Torrente-Vilara et al. 2011, Dias et al. 2012, Vitule et al. 2012).

Despite these general patterns, the fish fauna of each stream can be considered exclusive to their microbasins, as demonstrated by the high number of species not yet described. The presence of species common to both environments, such as *Gymnotus sylvius*, *Oreochromis niloticus*, *Phalloceros harpagos*, and *Synbranchus* cf. *marmoratus*, should be considered with reservation, because except for *Phalloceros harpagos*, for the other species there is the possibility of escapes or introductions via their use as bait (Graça & Pavanelli 2007, Baumgartner et al. 2012).

Regarding the differences in fish composition between streams upstream of the barrier (Iguaçu River Falls), it may be associated with conditions of preservation of streams sampled. For example, we have a high numerical abundance of *Phalloceros harpagos* in one of the streams (S1), where, besides the activity of livestock, human activities, such as the construction of a bridge, altered the normal course of the river, favoring the proliferation of less demanding species (Bifi et al. 2006, Lange et al. 2014). On the other hand, the presence of *Poecilia reticulata* in the stream S2, is possibly because to its location, near the urban center of municipality of Cascavel, since this species could be released or escape from aquarium. Tolerant non-native species in altered streams were recorded in several studies in streams of other basins in the Paraná State (Cunico et al. 2009, Araújo et al. 2011, Pagotto et al. 2012). The presence of non-native species is considered a major factor in the reduction of the native community (Jackson 2002, Di Prinzio et al. 2009, Eros et al. 2014, Zeni & Casatti 2014). In this study, the occurrences of *Gymnotus* and *Synbranchus* species can be attributed to the use of species of such genera as live bait (Graça & Pavanelli 2007, Baumgartner et al. 2012). These results confirm that huge changes in the use and occupation of land can cause changes in native fish assemblages in inland freshwater ecosystems.

Furthermore, despite having completely different fish fauna composition, the streams S3 and S4 showed a high number of native species (to the Iguaçu River and Paraná River, respectively). In addition, these streams are located near the ‘Parque Nacional do Iguaçu’, which suggests a higher degree of preservation. This fact shows that preserved environments offers better conditions for the permanence of native species (Lyons et al. 1995, Saunders et al. 2002, Casatti et al. 2012), which are more sensitive to changes, ensuring the preservation of endemism of this fish fauna.

The results summarized in this study show strongly the significant effect of the Iguaçu River Falls as a natural biogeographic barrier for speciation of fish species and endemism in streams of the Iguaçu River basin, leading to the development of distinct faunas in water bodies located upstream and downstream of such waterfalls. Although others studies had shown that geologic processes provide the capture of headwaters by adjacent basins (Ribeiro 2006; Dagosta et al. 2014), this seems not to be the case of this study. So, the main insight of our study is related to fact that differentiation has occurred in species composition between streams up and downstream and that these follow a similar pattern for the registered to the fish fauna of the principal channel. Thereby, it is stressed the importance of further surveys in headwaters streams, especially in the Iguaçu River to confirm and consolidate this findings. Peculiarities of this basin as the presence of geographical barriers (falls) with different levels of isolation, should provide different scenarios for speciation of the fish fauna.

The high number of endemic species not yet described deserves the attention of researchers and experts, so that they can be recognized by science and thus conserved, in order to maintain the biodiversity of freshwater ecosystems. In this context, we confirm the claim of Nogueira et al. (2010) who reported that a comprehensive strategy for the conservation of freshwater fish species in Brazil needs to address studies for restricted-range endemic species. Also, the presence of non-native species was more closely associated with streams under the influence of adjacent urban areas. These findings, along with urban sprawl in the lower Iguaçu River region, reinforce the imminent need for knowledge of this fauna, considering that the extinction of species in this region means the irreparable loss of them. In addition, considering that the basic unit of management is the basin, we suggested that it should be considered geographical spaces delimited by natural or artificial barriers as units separated in the basin when developing and implementing monitoring programs, impact studies and conservation plans.

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Inventory of the fish fauna from Ivaí River basin, Paraná State, Brazil

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Abstract: We compiled data on fish fauna of the Ivaí River basin from recent specialised literature, standardised sampling and records of species deposited in fish collections. There were 118 fish species of eight orders and 29 families. Of these, 100 species are autochthonous (84.8%), 13, allochthonous (11.0%) and five, exotic (4.2%). The main causes for the occurrence of non-native species are escapes from aquaculture, introduction for fishing purposes and the construction of the Itaipu hydroelectric plant. The predominance of small and medium-sized Characiformes and Siluriformes, including 13 species new to science, accounts for approximately 11.0% of all species and 13.0% of all native species. About 10.2% of all species and 12.0% of all native species are endemic to the upper stretch of the Ivaí River, isolated by numerous waterfalls in tributary rivers and streams. The Ivaí River basin is subjected to various anthropogenic interferences such as pollution, eutrophication, siltation, construction of dams, flood control, fisheries, species introduction and release of fingerlings. These activities raise concerns about biodiversity of Brazilian inland waters especially regarding the fish fauna; the basin of the Ivaí River already has species classified in categories of extinction risk: *Brycon nattereri* and *Apareiodon vladii* (Vulnerable) and *Characidium heirmostigmata* and *Steindachneridion scriptum* (Endangered). The high species richness of native fish, endemism of some, high environmental heterogeneity, high risk of extinction and lack of knowledge of several other species along with the eminent human activities raise the need to enrich the scientific knowledge for future conservation efforts for the studied basin.

Keywords: *ichthyofauna, upper Paraná River, checklist, biogeographic barriers, conservation.*

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Resumo: Nós compilamos dados sobre a diversidade da ictiofauna da bacia do rio Ivaí proveniente de recentes informações contidas em estudos divulgados na literatura especializada, coletas padronizadas e registros das espécies depositadas em coleções ictiológicas. Foram registradas 118 espécies de peixes pertencentes a oito ordens e 29 famílias. Dessas, 100 são autóctones, (84.8%), 13 são alóctones (11.0%) e cinco são exóticas (4.2%). As principais causas da ocorrência de espécies não nativas são escapes da piscicultura, introduções para pesca e a construção da usina hidrelétrica de Itaipu. Ocorre o predomínio de espécies de Characiformes e Siluriformes com porte pequeno e médio, sendo que 13 espécies são novas, o que representa aproximadamente 11.0% do total de espécies e 13.0% do total de espécies nativas. Ainda, aproximadamente 10.2% do total de espécies e 12.0% do total de espécies nativas correspondem a espécies endêmicas, isoladas pela presença de inúmeras cachoeiras em rios e riachos afluentes no trecho superior do rio Ivaí. A bacia do rio Ivaí está sujeita a uma variedade de interferências antrópicas como poluição, eutrofização, assoreamento, construção de represas, controle do regime de cheias, pesca, introduções de espécies e soltura de alevinos. Tais atividades apontam alarmantes preocupações com a biodiversidade das águas continentais brasileiras sobretudo para a ictiofauna, sendo que a bacia do rio Ivaí já apresenta espécies listadas em categorias de ameaças de extinção como *Brycon nattereri* e *Apareiodon vladii* (Vulneráveis) e *Characidium heirmostigmata* e *Steindachneridion scriptum* (Em Perigo). Devido à alta riqueza de espécies de peixes nativos, endemismo de algumas, alta heterogeneidade ambiental, sérios riscos de extinções e desconhecimento de várias outras espécies somados às eminentes ações antrópicas deve-se enriquecer o aporte científico de futuros apelos conservacionistas para a bacia aqui inventariada.

Palavras-chave: *peixes, alto rio Paraná, lista de espécies, barreiras biogeográficas, conservação.*

Introduction

Of the Brazilian basins, the Paraná River basin is the second largest drainage area after the Amazon basin (Stevaux et al. 1997, Galves et al. 2009). According to Agostinho et al. (2007), the upper section of the Paraná River is the most investigated with regards to Brazilian freshwater fish. This stretch covers water systems that cross the states of Goiás, Minas Gerais, São Paulo, Mato Grosso do Sul and Paraná. In the latter, it extends to the upstream region of the city of Guaíra, formerly Sete Quedas, now submerged by the Itaipu lake.

Studies on fish of the upper Paraná River basin have increased in recent years but are still mainly concentrated in basins of the São Paulo State (Langeani et al. 2007). Although these authors have surveyed the number of species with records in the upper Paraná River basin and totaled 310 valid species and 50 likely new species, a study performed by Galves et al. (2009) on the fish fauna surveys of the main tributaries of the upper Paraná River basin indicated a gap of these studies in relation to the Ivaí River basin, which is an important left bank tributary of the Paraná River, in the Paraná State.

There are only few surveys on fish fauna in the Ivaí River basin, especially for the tributaries, Barra Bonita River (Maier et al. 2008), Bonito River (Viana et al. 2013), some streams located in the Perobas Biological Reserve (Delariva & Silva 2013) and a first-order stream in the municipality of Marialva (Araújo et al. 2011); and there are few genetic studies with some species of Loricariidae (Zawadzki et al. 2004, Portela-Castro et al. 2007, Paiva et al. 2013) or ecological studies (Luiz et al. 2003, Luiz et al. 2005) and description of new species (Graça & Pavanelli 2008, Roxo et al. 2014, Tencatt et al. 2014, Zawadzki et al. 2016) with fish of this basin.

This study aims to provide a compilation of data on the diversity of the fish fauna of the Ivaí River basin from recent specialised literature, standardised sampling and records of species deposited in fish collections. In addition, endemism and threats to the species are discussed.

Material and Methods

1. *Study area*

A dense drainage network with many tributaries composes the watershed of the Ivaí River, the second largest in the Paraná State, located at the geographical coordinates 22º56’17” – 25º35’27” S and 50º44’17” – 53º41’43” W (Destefani 2005). The Ivaí River is a left bank tributary of the Paraná River in the Paraná State and has 35,845 km^2 drainage area, which is approximately 685 km in length (Maack 1981). This river is formed in the municipality of Ivaí by the confluence of the rivers Patos and São João (Santos et al. 2008), both in the State Park of Serra da Esperança, on the border between the second and third plateau of the Paraná State (Maack 1981).

The Ivaí River basin has different geological and geomorphological characteristics, since it runs through different lithologies and drains distinct morphological and topographical environments (Destefani 2005). In this way, this author has defined three sections for the basin considering the geology, geomorphology, topography and slope: upper, middle and lower stretches (Figure 1).

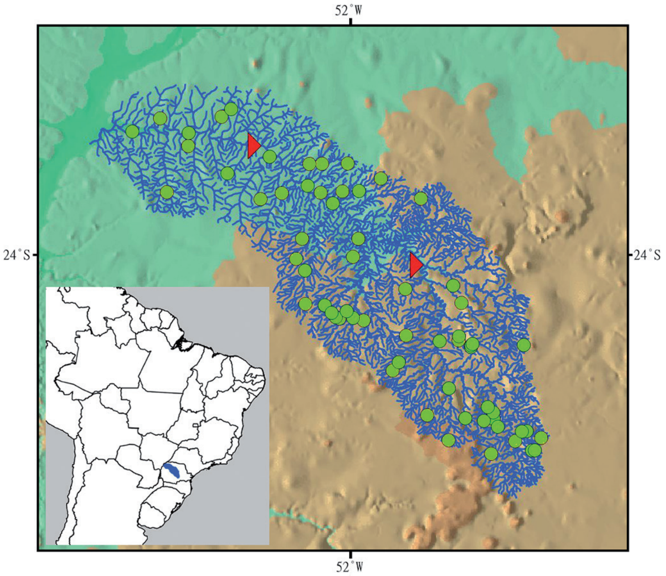

Figure 1. Map of the Ivaí River basin, showing the most thoroughly sampled sites (green dots). Each dot may correspond to more than one collection site. Limits between upper, medium and lower sections are represented by red triangles.

The upper section is the longest with about 440 km and has the highest slopes of the whole basin, especially from the source of the Patos River until its confluence with the São João River; also there are rapids and waterfalls due to the type of geological substrate formed by sedimentary rocks, which, according to Maack (1981), promote a stepped relief formed by ridges. By crossing the slope of the Serra da Esperança, the Ivaí River enters the third plateau and flows upon basaltic rocks, giving rise to the middle section, which is approximately 170 km. In this section, the slope is much smaller, with a less energetic relief; there are small and shallow waterfalls as well as important rapids as the Ferro and Índio rapids, intercalated by backwaters of gentle slope (Destefani 2005). The lower section is the floodplain and is approximately 164 km in length. Therein, the river flows over sandstones of the Caiuá formation and of alluvial sediments, with a very small slope of 20 meters until flowing into the Paraná River (Destefani 2005).

Other studies show some physical characteristics of each part of the Ivaí River basin, such as altitude, temperature and rainfall. Paiva (2008) stated that the altitude in the upper section of this basin has, on average, 800 m, but it can reach up to 1,250 m, in the middle stretch, the average altitude is 500 meters, and in the lower reaches, 250 m, on average. Ichiba (2006) reports the average annual temperatures for each section of the basin: 18ºC for the upper reaches, 20ºC in the middle and 22ºC in the lower reaches. The research of Sousa (2006) determined the average annual rainfall for the same sections: 1,800 mm for the upper reaches, 1,600 mm in the middle and 1,400 mm for the lower reaches.

2. *Database*

The list of species for the Ivaí River basin was developed by consulting the material deposited in the fish collections of the Universidade Estadual de Londrina, Londrina (MZUEL); Museu de Zoologia da Universidade de São Paulo, São Paulo (MZUSP); Museu de História Natural Capão da Imbuia, Curitiba (MHNCI), Museu de Ciências e Tecnologia da PUCRS, Porto Alegre (MCP) and the Coleção Ictiológica do Nupélia, Maringá (NUP), which are available at http://splink.cria.org.br. This list was compared with the species listed for the upper Paraná River basin in Langeani et al. (2007). Species with doubtful occurrence or identification were reviewed by experts.

To complement the information, recent collections were conducted under the permission #14028-1 (Sistema de Informação e Autorização em Biodiversidade - Sisbio, Instituto Chico Mendes de Conservação da Biodiversidade) in 21 streams of the upper reaches of the basin, the region with the lowest number of samples. The origin of each species was determined according to by Langeani et al. (2007), in which autochthonous species are native to the upper Paraná River basin, the allochthonous species were introduced from other basins belonging to the Neotropical region and the exotic were introduced from other continents. The origin of each species were obtained from Reis et al. (2003), Langeani et al. (2007), Graça & Pavanelli (2007) and Julio Júnior et al. (2009), which compile data of several long-term studies carried out by the Nupélia – Universidade Estadual de Maringá.

Threat level was determined in accordance with the Portaria do Ministério do Meio Ambiente Nº 445, of December 17th, 2014 and changed by Nº 98, of April 28th, 2015, which recognize species of fish and aquatic invertebrates of Brazilian fauna threatened with extinction classified in the following categories: Extinct in the Wild (EW), Critically Endangered (CR), Endangered (EN) and Vulnerable (VU). For endemism determinations, some species were considered as restricted to the basin by Graça & Pavanelli (2008), Roxo et al. (2014), Tencatt et al. (2014), Zawadzki et al. (2016) and personal communications from Cláudio H. Zawadzki (UEM); other species were considered endemic by means of analysis and comparison of data obtained with comparative material from adjacent basins.

3. *Fish identification*

Identification follows Graça & Pavanelli (2007), Graça & Pavanelli (2008), Tencatt et al. (2014) and Roxo et al. (2014). Some individuals of *Trichomycterus* identified as *Trichomycterus* sp., *Trichomycterus* sp. 1 *Trichomycterus* sp. 2 and *Trichomycterus* sp. 3 in the studies of Maier et al. (2008), Araújo et al. (2011), Delariva & Silva (2013) and Viana et al. (2013), which are deposited in the fish collection of Nupélia, were re-examined and identified as *Trichomycterus davisi* (Haseman, 1911). In addition, some species recorded in the collections analysed were re-examined and identifications were rectified, namely: *Hypostomus aspilogaster* (Cope, 1894) is *Hypostomus ancistroides* (Ihering, 1911); *Hypostomus* sp. is *Hypostomus* sp. 1; *Hypostomus* sp. 1 is *Hypostomus* sp. 2; *Hypostomus* sp. 3 is *Hypostomus hermanni* (Ihering, 1905); *Corumbataia* sp. is *Hisonotus pachysarkos* Zawadzki, Roxo & Graça, 2016; *Bryconamericus exodon* (Eigenmann, 1907) is *Piabarchus* aff. *stramineus* (Eigenmann, 1908); *Creagrutus* sp. is *Bryconamericus* sp.; *Hyphessobrycon* sp. is *Diapoma guarani* (Mahnert & Géry, 1987); *Hyphessobrycon* aff. *guarani* Mahnert & Géry, 1987 is *Planaltina* sp.; *Mimagoniates microlepis* (Steindachner, 1877) is *Piabarchus* aff. *stramineus; Oligosarcus* sp. is *Oligosarcus paranensis* Menezes & Géry, 1983; *Piabina* sp. is *Piabina argentea* Reinhardt, 1867; *Serrasalmus spilopleura* Kner, 1858 is *Serrasalmus maculatus* Kner, 1858; *Phalloceros* aff. *caudimaculatus* (Hensel, 1868) is *Phalloceros harpagos* Lucinda, 2008; *Gymnotus* aff. *carapo* (Linnaeus, 1758) is *Gymnotus inaequilabiatus* (Valenciennes, 1839), *Crenicichla niederleinii* (Holmberg, 1891) and *Crenicichla haroldoi* Luengo & Britski, 1974 are *Crenicichla jaguarensis* Haseman, 1911. Five species have had taxonomic changes recently: *Astyanax altiparanae* Garutti & Britski, 2000 considered a junior synonym of *A. lacustris* (Lütken, 1875) by Lucena and Soares (2016); three new genus changes of *Bryconamericus stramineus* to *Piabarchus*, *Hyphessobrycon guarani* to *Diapoma* by Thomaz et al. (2015) and *Hisonotus insperatus* Britski & Garavello, 2003 and *H. oliveirai* to *Curculionichthys* by Roxo et al. (2015).

Results

There were 118 fish species registered in the Ivaí River basin (Table 1) belonging to eight orders and 29 families (Table 2). The most species-rich orders are Siluriformes and Characiformes (Table 1), with 51 and 48 species, respectively (Table 2). Together, they represent 83.9% of all species registered. The richest families are Loricariidae and Characidae with, respectively, 24 and 20 species (Table 2), representing approximately 37.2% of the species.

The Ivaí River basin has 13 new species to science (species marked with an asterisk are endemic to the Ivaí River basin; Figure 2, Table 1): *Ancistrus* sp., *Apareiodon* sp.*, *Aphyocharax* sp., *Bryconamericus* sp.*, *Cnesterodon* sp.*, *Hoplias* sp. 2, *Hoplias* sp. 3, *Hypostomus* sp. 1*, *Hypostomus* sp. 2*, *Hypostomus* sp. 3*, *Neoplecostomus* sp.*, *Odontostilbe* sp. and *Planaltina* sp.*, representing 11.0% of all species and 13.0% of native species. Eight of the new species plus *Characidium heirmostigmata* Graça & Pavanelli, 2008, *Corydoras lacrimostigmata* Tencatt, Britto & Pavanelli, 2014, *Curculionichthys oliveirai* (Roxo, Zawadzki & Troy, 2014) and *Hisonotus pachysarkos* Zawadzki, Roxo & Graça, 2016 make a total of twelve endemic species (Figure 2, Table 1), corresponding to 10.2% of all species and 12.0% of all native species. Fish species of the Ivaí River basin listed in the Portaria do Ministério do Meio Ambiente Nº 445 and changed by Nº 98 fall into the conservation status categories Endangered (EN) and Vulnerable (VU) (Table 1). The following species are considered to be the most threatened: *Apareiodon vladii* Pavanelli, 2006; *Brycon nattereri* Günther, 1864; *Characidium heirmostigmata* Graça & Pavanelli, 2008; and *Steindachneridion scriptum* (Miranda-Ribeiro, 1918).

Five exotic species were captured (Figure 3). Of these, the carp, *Cyprinus carpio* (Linnaeus, 1758), and the tilapias, *Oreochromis niloticus* (Linnaeus, 1758) and *Tilapia rendalli* (Boulenger, 1897), probably colonised the basin by escaping from fish farms. This also explains the presence of the other two exotic species, the channel catfish, *Ictalurus punctatus* (Rafinesque, 1818), and the black bass, *Micropterus salmoides* (La Cépède, 1802), along with introduction for sport-fishing purposes. From the 13 allochthonous species (Figure 2, Table 1), nine invaded the upper Paraná River and the Ivaí River from the lower Paraná basin through the spawning channel of the Itaipu Reservoir (Canal da Piracema), opened December 2002. Those species are: *Apteronotus* aff. *albifrons* (Linnaeus, 1766), *Hemiodus orthonops* Eigenmann & Kennedy, 1903, *Hypostomus commersoni* Valenciennes, 1836, *Pimelodus ornatus* Kner, 1858, *Porotergus ellisi* Arámburu, 1957, *Potamotrygon falkneri* Castex & Maciel, 1963, *Roeboides descalvadensis* Fowler, 1932, *Steindachnerina brevipinna* (Eigenmann & Eigenmann, 1889) and *Trachydoras paraguayensis* (Eigenmann & Ward, 1907). Two other allochthonous species, *Erythrinus erythrinus* (Bloch & Schneider, 1801) and *Hoplerythrinus unitaeniatus* (Agassiz, 1829), were introduced by sport fishers; one species, *Plagioscion squamosissimus* (Heckel, 1840), was introduced by escape from fish farming; and another species, *Poecilia reticulata* Peters, 1859, was introduced for mosquito control.

Table 1. Ichthyofauna from the Ivaí River basin: species, voucher specimens, origin of each species, threat level and endemism. The symbol # refers to species added to the list due to personal observation. The asterisk (*) represents endemic species.

Species	Voucher	Origin/Threat level
ELASMOBRANCHII		
Myliobatiformes		
Potamotrygonidae		
1 *Potamotrygon falkneri* Castex & Maciel, 1963	NUP10918	Allochthonous
ACTINOPTERYGII		
Characiformes		
Acestrorhynchidae		
2 *Acestrorhynchus lacustris* (Lütken, 1875)	NUP5541	Autochthonous
Anostomidae		
3 *Leporinus amblyrhynchus* Garavello & Britski, 1987	NUP5554	Autochthonous
4 *Leporinus friderici* (Bloch, 1794)	#	Autochthonous
5 *Leporinus octofasciatus* Steindachner, 1915	NUP10636	Autochthonous
6 *Schizodon nasutus* Kner, 1858	NUP1440	Autochthonous
Bryconidae		
Bryconinae		
7 *Brycon nattereri* Günther, 1864	NUP8534	Autochthonous/(VU)
Salmininae		
8 *Salminus brasiliensis* (Cuvier, 1816)	#	Autochthonous
Characidae		
9 *Astyanax bockmanni* Vari & Castro, 2007	NUP5487	Autochthonous
10 *Astyanax lacustris* (Lütken, 1875)	NUP3941	Autochthonous
11 *Astyanax* aff. *fasciatus* (Cuvier, 1819)	NUP11538	Autochthonous
12 *Astyanax* aff. *paranae* Eigenmann, 1914	NUP11794	Autochthonous
13 *Hemigrammus* cf. *marginatus* Ellis, 1911	NUP1498	Autochthonous
14 *Moenkhausia* aff. *gracilima* Eigenmann, 1908	NUP9746	Autochthonous
15 *Oligosarcus pintoi* Campos, 1945	NUP16388	Autochthonous
16 *Oligosarcus paranensis* Menezes & Géry, 1983	NUP16381	Autochthonous
Aphyocharacinae		
17 *Aphyocharax* sp.	NUP5550	Autochthonous
Characinae		
18 *Galeocharax knerii* (Steindachner, 1879)	NUP10737	Autochthonous
19 *Roeboides descalvadensis* Fowler, 1932	NUP5531	Allochthonous
Cheirodontinae		
20 *Odontostilbe* sp.	NUP5533	Autochthonous
21 *Serrapinnus notomelas* (Eigenmann, 1915)	NUP17827	Autochthonous
Stevardiinae		
22 *Bryconamerius* aff. *iheringii* (Boulenger, 1887)	NUP16083	Autochthonous
23 *Bryconamericus turiuba* Langeani, Lucena, Pedrini & Tarelho-Pereira, 2005	NUP16369	Autochthonous
24 *Bryconamericus* sp.	NUP17150	Autochthonous*
25 *Diapoma guarani* (Mahnert & Géry, 1987)	NUP5066	Autochthonous
26 *Piabarchus* aff. *stramineus* (Eigenmann, 1908)	NUP16385	Autochthonous
27 *Piabina argentea* Reinhardt, 1867	NUP5010	Autochthonous
28 *Planaltina* sp.	NUP17152	Autochthonous*
Curimatidae		
29 *Cyphocharax modestus* (Fernández-Yépez, 1948)	NUP11730	Autochthonous

Continued Table 1.

30 *Cyphocharax nagelii* (Steindachner, 1881)	NUP5530	Autochthonous
31 *Steindachnerina brevipinna* (Eigenmann & Eigenmann, 1889)	NUP5538	Allochthonous
32 *Steindachnerina* cf. *corumbae* Pavanelli & Britski, 1999	NUP16396	Autochthonous
33 *Steindachnerina insculpta* (Fernández-Yépez, 1948)	NUP5529	Autochthonous
Crenuchidae		
34 *Characidium* aff. *zebra* Eigenmann, 1909	NUP1383	Autochthonous
35 *Characidium gomesi* Travassos, 1956	NUP11718	Autochthonous
36 *Characidium heirmostigmata* Graça & Pavanelli, 2008	NUP17136	Autochthonous*/(EN)
Erythrinidae		
37 *Erythrinus erythrinus* (Bloch & Schneider, 1801)	NUP11729	Allochthonous
38 *Hoplerythrinus unitaeniatus* (Spix & Agassiz, 1829)	NUP1499	Allochthonous
39 *Hoplias intermedius* (Günther, 1864)	NUP10020	Autochthonous
40 *Hoplias* sp. 2	NUP14323	Autochthonous
41 *Hoplias* sp. 3	NUP15907	Autochthonous
Hemiodontidae		
42 *Hemiodus orthonops* Eigenmann & Kennedy, 1903	#	Allochthonous
Parodontidae		
43 *Apareiodon affinis* (Steindachner, 1879)	NUP5539	Autochthonous
44 *Apareiodon piracicabae* (Eigenmann, 1907)	NUP4548	Autochthonous
45 *Apareiodon vladii* Pavanelli, 2006	NUP16081	Autochthonous/(VU)
46 *Apareiodon* sp.	NUP1501	Autochthonous*
47 *Parodon nasus* Kner, 1859	NUP9857	Autochthonous
Prochilodontidae		
48 *Prochilodus lineatus* (Valenciennes, 1850)	NUP6064	Autochthonous
Serrasalmidae		
49 *Serrasalmus maculatus* Kner, 1858	#	Autochthonous
Cypriniformes		
Cyprinidae		
50 *Cyprinus carpio* Linnaeus, 1758	NUP860	Exotic
Cyprinodontiformes		
Poeciliidae		
51 *Cnesterodon* sp.	NUP5475	Autochthonous*
52 *Phalloceros harpagos* Lucinda, 2008	NUP5551	Autochthonous
53 *Poecilia reticulata* Peters, 1859	NUP11792	Allochthonous
Gymnotiformes		
Apteronotidae		
54 *Apteronotus* aff. *albifrons* (Linnaeus, 1766)	NUP3058	Allochthonous
55 *Apteronotus caudimaculosus* Santana, 2003	NUP10613	Autochthonous
56 *Porotergus ellisi* Arámburu, 1957	NUP3057	Allochthonous
Gymnotidae		
57 *Gymnotus inaequilabiatus* (Valenciennes, 1839)	NUP15364	Autochthonous
58 *Gymnotus sylvius* Albert & Fernandes-Matioli, 1999	NUP11281	Autochthonous
Perciformes		
Centrarchidae		
59 *Micropterus salmoides* (La Cepède, 1802)	NUP2672	Exotic

Continued Table 1.

Cichlidae		
60 *Cichlasoma paranaense* Kullander, 1983	NUP11728	Autochthonous
61 *Crenicichla britskii* Kullander, 1982	NUP3575	Autochthonous
62 *Crenicichla jaguarensis* Haseman, 1911	NUP10797	Autochthonous
63 *Geophagus brasiliensis* (Quoy & Gaimard, 1824)	NUP5524	Autochthonous
64 *Oreochromis niloticus* (Linnaeus, 1758)	NUP15900	Exotic
65 *Tilapia rendalli* (Boulenger, 1897)	NUP858	Exotic
Sciaenidae		
66 *Plagioscion squamosissimus* (Heckel, 1840)	#	Allochthonous
Siluriformes		
Auchenipteridae		
67 *Glanidium* cf. *cesarpintoi* Ihering, 1928	NUP5543	Autochthonous
68 *Tatia neivai* (Ihering, 1930)	NUP11045	Autochthonous
69 *Parauchenipterus galeatus* (Linnaeus, 1766)	#	Autochthonous
Callichthyidae		
70 *Callichthys callichthys* (Linnaeus, 1758)	NUP6122	Autochthonous
71 *Corydoras* aff. *aeneus* (Gill, 1858)	NUP11736	Autochthonous
72 *Corydoras ehrhardti* Steindachner, 1910	NUP15899	Autochthonous
73 *Corydoras lacrimostigmata* Tencatt, Britto & Pavanelli, 2014	NUP1446	Autochthonous*
Cetopsidae		
74 *Cetopsis gobioides* Kner, 1858	NUP11673	Autochthonous
Doradidae		
75 *Trachydoras paraguayensis* (Eigenmann & Ward, 1907)	NUP2084	Allochthonous
Loricariidae		
Hypoptopomatinae		
76 *Curculionichthys insperatus* (Britski & Garavello, 2003)	NUP3578	Autochthonous
77 Curculionichthys oliveirai (Roxo, Zawadzki & Troy, 2014)	NUP16070	Autochthonous*
78 *Hisonotus francirochai* (Ihering, 1928)	NUP16379	Autochthonous
79 *Hisonotus pachysarkos* Zawadzki, Roxo & Graça, 2016	NUP16258	Autochthonous*
Hypostominae		
80 *Ancistrus* sp.	NUP15978	Autochthonous
81 *Hypostomus ancistroides* (Ihering, 1911)	NUP16080	Autochthonous
82 *Hypostomus albopunctatus* (Regan, 1908)	NUP10109	Autochthonous
83 *Hypostomus commersoni* Valenciennes, 1836	NUP856	Allochthonous
84 *Hypostomus hermanni* (Ihering, 1905)	NUP9806	Autochthonous
85 *Hypostomus iheringii* (Regan, 1908)	NUP 4837	Autochthonous
86 *Hypostomus margaritifer* (Regan, 1908)	NUP4921	Autochthonous
87 *Hypostomus* aff. *paulinus* (Ihering, 1905)	NUP15329	Autochthonous
88 *Hypostomus strigaticeps* (Regan, 1908)	NUP4530	Autochthonous
89 *Hypostomus regani* (Ihering, 1905)	NUP4979	Autochthonous
90 *Hypostomus* cf. *topavae* (Godoy, 1969)	NUP4529	Autochthonous
91 *Hypostomus* sp. 1	NUP 2597	Autochthonous*
92 *Hypostomus* sp. 2	NUP10917	Autochthonous*
93 *Hypostomus* sp. 3	NUP 4745	Autochthonous*
94 *Megalancistrus parananus* (Peters, 1881)	NUP4466	Autochthonous

Continued Table 1.

Loricariinae		
95 *Farlowella amazonum* (Günther, 1864)	NUP1450	Autochthonous
96 *Loricaria prolixa* Isbrücker & Nijssen, 1978	NUP3359	Autochthonous
97 *Rineloricaria pentamaculata* Langeani & Araújo, 1994	NUP16079	Autochthonous
98 *Rineloricaria* cf. *latirostris* (Boulenger, 1900)	NUP5527	Autochthonous
Neoplecostominae		
99 *Neoplecostomus* sp.	NUP10113	Autochthonous*
Heptapteridae		
100 *Cetopsorhamdia iheringi* Schubart & Gomes, 1959	NUP5483	Autochthonous
101 *Heptapterus mustelinus* (Vallenciennes, 1835)	NUP11667	Autochthonous
102 *Imparfinis borodini* Mees & Cala, 1989	NUP4549	Autochthonous
103 *Imparfinis mirini* Haseman, 1911	NUP3582	Autochthonous
104 *Imparfinis schubarti* (Gomes, 1956)	NUP11809	Autochthonous
105 *Pimelodella avanhandavae* Eigenmann, 1917	NUP5553	Autochthonous
106 *Pimelodella gracilis* (Valenciennes, 1835)	NUP1783	Autochthonous
107 *Rhamdia quelen* (Quoy & Gaimard, 1824)	NUP16090	Autochthonous
108 *Phenacorhamdia tenebrosa* (Schubart, 1964)	NUP16072	Autochthonous
Ictaluridae		
109 *Ictalurus punctatus* (Rafinesque, 1818)	NUP757	Exotic
Pimelodidae		
110 *Iheringichthys labrosus* (Lütken, 1874)	NUP5544	Autochthonous
111 *Pimelodus microstoma* Steindachner, 1877	NUP5534	Autochthonous
112 *Pimelodus ornatus* Kner, 1858	NUP10747	Allochthonous
113 *Pimelodus paranaensis* Britski & Langeani, 1988	NUP9143	Autochthonous
114 *Pseudoplatystoma corruscans* (Spix & Agassiz, 1829)	#	Autochthonous
115 *Steindachneridion scriptum* (Miranda-Ribeiro, 1918)	NUP2511	Autochthonous/(EN)
Trichomycteridae		
116 *Trichomycterus davisi* (Haseman, 1911)	NUP16071	Autochthonous
117 *Trichomycterus diabolus* Bockmann, Casatti & de Pinna, 2004	NUP5482	Autochthonous
Synbranchiformes		
Synbranchidae		
118 *Synbranchus marmoratus* Bloch, 1795	NUP5552	Autochthonous

Discussion

The ichthyofauna of the Ivaí River basin, comprised of 118 species, has an alpha diversity superior to that of all other river systems confined to the Paraná State. There are 110 species in the Tibagi River basin (Shibatta et al. 2002), 76 in the Pirapó River basin (Pagotto et al. 2012), 62 in the Piquiri River basin (Gubiani et al. 2006), 54 in the Jordão River basin (Frota et al. in prep.) and 48 in the Areia river basin (Frota et al. in prep.). The Iguaçu River basin, another important system in the Paraná State, although not completely comprised in it, lacks a complete inventory, but Baumgartner et al. (2012) found in its lower stretch 106 species, an amount similar to that of the Ivaí River as a whole. Other comparable systems within the upper Paraná River basin for which inventories are available show similar species richness. The Mogi Guaçu River has about 135 species (Meschiatti & Arcifa 2009, Oliveira et al. 2009), and the Corumbá Reservoir and its influence area, 119 (Pavanelli et al. 2007). The upper Paraná floodplain harbors 182 fish species, a number considerably higher than that observed in the Ivaí River basin, but that region also presents a higher habitat diversity (Graça & Pavanelli 2007).

In the headwaters of the Ivaí River, there is a predominance of small and medium-sized (15 cm or less in length) Characiformes and Siluriformes. As already highlighted by Viana et al. (2013) in their study of the Bonito River, backwaters separated by rapids typically bear a wide variety of microhabitats. Those environments favour species with greater ability to stay in the water column (*e.g.*, *Astyanax* and *Bryconamericus*), as well as benthic species equipped with spines that

Table 2. Number of species in the Ivaí River basin, sorted by Order and Family.

Order	Family	Number of species	%
1. Characiformes		48	40.7
	1. Acestrorhynchidae	1	0.8
	2. Anostomidae	4	3.4
	3. Bryconidae	2	1.7
	4. Characidae	20	16.9
	5. Curimatidae	5	4.2
	6. Crenuchidae	3	2.5
	7. Erythrinidae	5	4.2
	8. Hemiodontidae	1	0.8
	9. Parodontidae	5	4.2
	10. Prochilodontidae	1	0.8
	11. Serrasalmidae	1	0.8
2. Cypriniformes		1	0.8
	12. Cyprinidae	1	0.8
3. Cyprinodontiformes		3	2.5
	13. Poeciliidae	3	2.5
4. Gymnotiformes		5	4.2
	14. Apteronotidae	3	2.5
	15. Gymnotidae	2	1.7
5. Perciformes		8	6.8
	16. Centrarchidae	1	0.8
	17. Cichlidae	6	5.1
	18. Sciaenidae	1	0.8
6. Siluriformes		51	43.2
	19. Auchenipteridae	3	2.5
	20. Callichthyidae	4	3.4
	21. Cetopsidae	1	0.8
	22. Doradidae	1	0.8
	23. Loricariidae	24	20.3
	24. Heptapteridae	9	7.6
	25. Ictaluridae	1	0.8
	26. Pimelodidae	6	5.1
	27. Trichomycteridae	2	1.7
7. Synbranchiformes		1	0.8
	28. Synbranchidae	1	0.8
8. Myliobatiformes		1	0.8
	29. Potamotrygonidae	1	0.8

allow them to attach to rocks and resist the water flow (*e.g.*, loricariids and *Trichomycterus*). On the other hand, higher-order waterbodies have greater water volume and light incidence, which result in increased primary production and greater availability of resources (Ferreira et al. 2010). Thus, the lower reaches of the Ivaí River basin bear large-sized pelagic (*e.g.*, the characiforms *Salminus* and *Leporinus*) and benthic (*e.g.*, the siluriforms *Pseudoplatystoma* and *Steindachneridion*) species. In addition, higher-order streams offer a wider array of food items, which allows the coexistence of several trophic guilds ranging from detritivores and planktivores to piscivores, also contributing to their species richness (Ferreira et al. 2010).

The proportion of new species in relation to the total observed in the Ivaí River basin (11.0%) is close to the 14.4% reported by Langeani et al. (2007) for the entire upper Paraná River. Of the new species from the Ivaí River, only five are not endemic, and have already been recognized as so in previous studies performed in other river systems (*e.g.*, Graça & Pavanelli 2007). That shows how poorly known the Ivaí River basin is and that the sampling of low-order streams can reveal unknown species even in a relatively well-sampled river basin such as that of the upper Paraná River. The identification of areas of endemism is extremely important considering that they compose basic geographic units that allow understanding of the evolution of complex regional biota (Morrone 1994) and that should be prioritised for biodiversity conservation (Löwenberg-Neto & Carvalho 2004). Historical biogeography may explain this endemism as a result of speciation processes caused by the emergence of geographic barriers mainly in the upper reaches of the basin, located on the Serra da Esperança. Maack (1981) stated that this mountain range is part of the Triassic-Jurassic relief and corresponds, among other basins, to areas draining the headwaters of the Ivaí River. The sandy and clayey rocks of the geological formations that underlie the Serra da Esperança, combined with the dense drainage network, allow the development of a very uneven relief, with altitudes up to 1,200 m (Maack 1981). Thus, tectonism imposed deformations to the basis, forming some local inflections on the ground, as the Ponta Grossa Arch, and had close relationship with the basaltic effusion in the Mesozoic. Because of these orogenetic features, in that region there are numerous waterfalls, some over 50 meters high, such as Saltos São João and São Francisco (Maack 1981). According to Ribeiro (2006), the formation of some of the Paraná State river basins was strongly affected by the origin of the Ponta Grossa Arch, which may have had an influence on speciation processes.

According to IUCN (2012), the categories Vulnerable and Endangered contain species to which the best available evidence indicates they are at, respectively, high and very high risk of extinction in the wild. Environments with species listed in these categories should be prioritized in terms of conservation. It is therefore important to create more Conservation Units and National Plans that focus the full protection of these sites, such as stretches of the upper Ivaí River basin where most records of *Characidium heirmostigmata* (Endangered), *Apareiodon vladii* and *Brycon nattereri* (vulnerable) were made. Additionally, for many species there is no adequate information to assess, directly or indirectly, the risk of extinction, therefore more studies are necessary to determine a more appropriate threat classification.

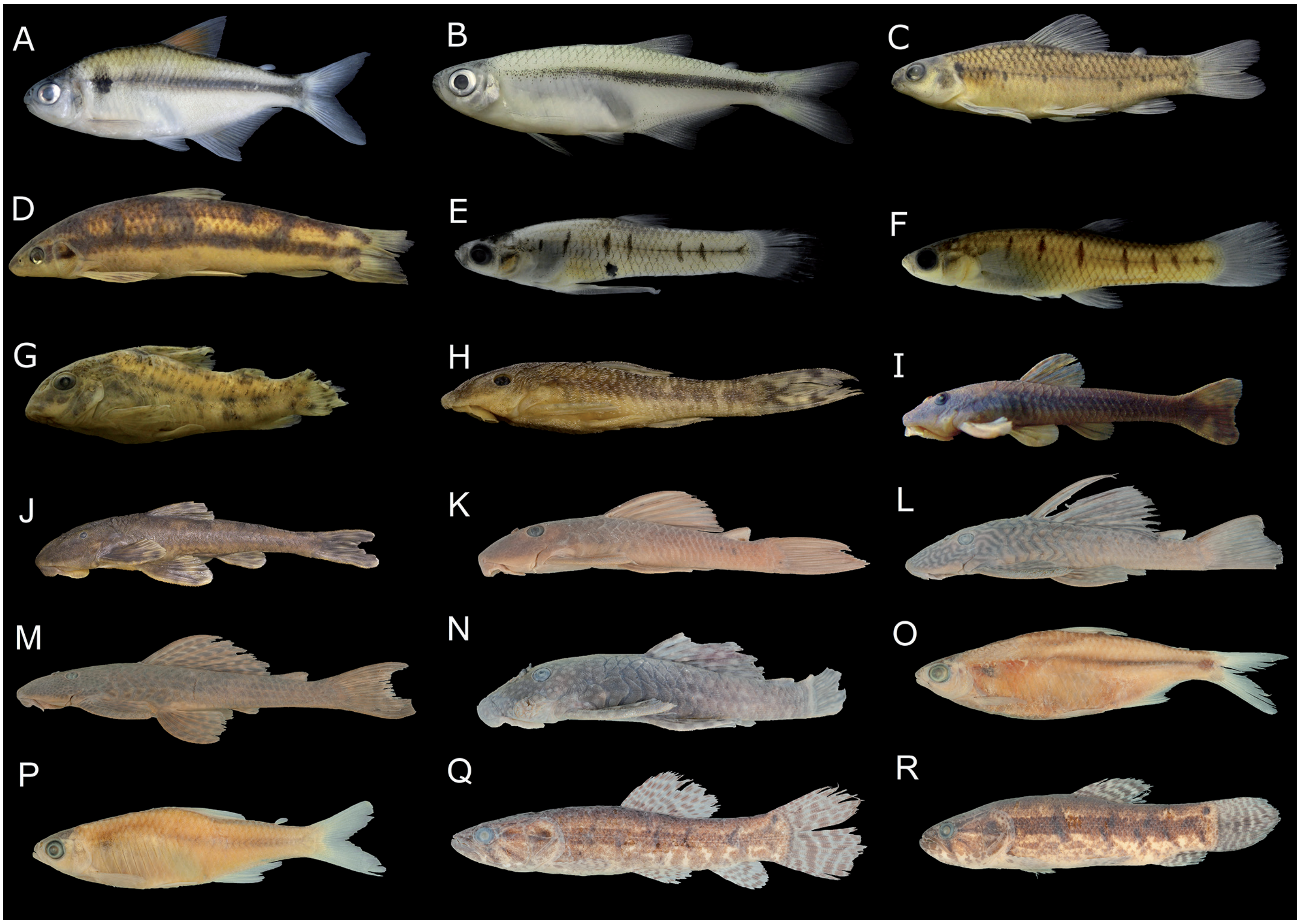

Figure 2. Representative specimens of endemic and new fish species to the Ivaí River basin. Their catalogue numbers in the Coleção Ictiológica do Nupélia (NUP) and standard lengths are presented after the names of species. A) *Bryconamericus* sp., NUP 17150, 66.7 mm; B) *Planaltina* sp., NUP 17152, 39.6 mm; C) *Characidium heirmostigmata*, NUP 17136, 46.6 mm; D) *Apareiodon* sp., NUP 1501, 89.7 mm; E) *Cnesterodon* sp. (male), NUP 4167, 19.2 mm; F) *Cnesterodon* sp. (female), NUP 5475, 30.3 mm; G) *Corydoras lacrimostigmata*, NUP 1446, 33.6 mm; H) *Curculionichthys oliveirai* - NUP 16070, 28.7 mm; I) *Hisonotus pachysarkos*, NUP 16258, 35.8 mm; J) *Neoplecostomus* sp., NUP 10113, 81.1 mm; K) *Hypostomus* sp. 1, NUP 2597, 86.8 mm; L) *Hypostomus* sp. 2; NUP 10917, 141.1 mm; M) *Hypostomus* sp. 3; NUP 4745, 139.97 mm; N) *Ancistrus* sp., NUP 15978, 101.7 mm; O) *Odontostilbe* sp., NUP 5553, 70.3 mm; P) *Aphyocharax* sp., NUP 5550, 39.1 mm; Q) *Hoplias* sp. 2, NUP 14323, 94.2 mm; R) *Hoplias* sp. 3, NUP 15907, 101.7 mm.

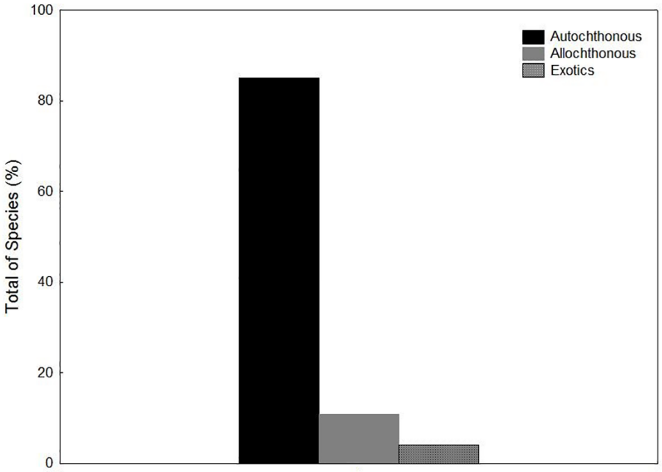

Figure 3. Graphic showing the percentages of the origins of species in the Ivaí River basin.

Agostinho et al. (2005) provided alarming data on the habitat conditions of Brazilian freshwater species and listed the main causes of biodiversity loss: pollution, eutrophication, siltation, construction of dams, flood control, fisheries and species introduction. This is also true for the Ivaí River basin (Affonso et al. 2015). In its drainage area, there are nearly one million inhabitants, of which only 36% have domestic sewage treatment (Paraná 2010). Also contributing to the pollution of its waters, the area comprises virtually no conservation units and little riparian vegetation due to the intensive use of the soil for agriculture and cattle raising (Paraná 2010). These activities greatly contribute to the siltation of the waterbodies, which was observed in the sites sampled herein and by Viana et al. (2013), and lead to destruction of native vegetation that provide important food items in the diet of some species.

Hydropower enterprises threaten fish biodiversity by controlling the hydrological regime and changing the longitudinal distribution of the species (Petry et al. 2011). This affects the reproductive success of the migratory species by influencing the timing, duration and intensity of floods and droughts (Pelicice et al. 2015). In the last few years, several projects for building hydroelectric plants of different proportions in the Ivaí River basin have been put forward. Those projects were received with apprehension not only by ecologists (Affonso et al. 2015), but also by communities that would be directly affected by their implantation. By demonstrating that the number of new, endemic and threatened species in the Ivaí River is greater than previously imagined, the present paper reinforces the need to promote popular manifestations against those dams.

Although overfishing is not a major problem in the Ivaí River basin, sport fishing contributes to the accidental or intentional introduction of species. In that basin, escapes from fish farms and invasion from the lower Paraná River basin also contribute to the presence of non-native species. Among the ones observed in the Ivaí River basin, few have been studied for their potential of invasion and possible antagonistic interactions with native species. That is the case of *Plagioscion squamosissimus*, which has been demonstrated to consume the same food items as other piscivores in the upper Paraná flodplain and thus, to be a probable competitor (Pereira et al. 2015). Similar studies are not available for *Micropterus salmoides*, but the fact that this species is also a piscivore indicate that it also has a potential to affect negatively native species from the Ivaí River. This calls for a close observation upon invasive species, as in the study of Pelicice & Agostinho (2009), who reported on the relationship between the expansion of *Cichla kelberi* Kullander & Ferreira in the Rosana Reservoir and the decline of fish communities associated with macrophyte stands. Although this species is absent from the Ivaí River Basin, a similar threat may be posed by *M. salmoides* and *P. squamosissimus*.

An activity that has become quite common in the Ivaí River is the careless release of thousands of fingerlings of the species of sport fishes, *e.g. Leporinus* spp., *Piaractus mesopotamicus*, *Pseudoplatystoma* spp., *Prochilodus* spp. and *Salminus* spp. Although welcomed by the population, this practice is stated by Agostinho et al. (2007) as frequently inefficient (because the river was devoid of its ancestral capacity of supporting large fish populations) or even harmful to the natural populations. It potentially causes loss of genetic diversity and of important alleles selected along many years by their advantages to the survival of the species in that particular habitat, as well as introduction of new pathogens and parasites (Agostinho et al. 2010). Thus, until those actions are carried out based on a large body of knowledge on the life history and population genetics of the species that are being reintroduced, they will continue to be no more than a waste of public money and an additional threat to the native species, as well as a deceiving electoral strategy.

From the results obtained, it becomes evident the need for efforts to preserve the Ivaí River basin, given its high environmental heterogeneity, high species richness, endemism and high risk of extinction of some species. In addition, the lack of information on biology and ecology of various species and the eminent human activities that affect much of the waterbodies in the basin, raise the need for continuity of studies on fish fauna in this basin.

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Influence of abiotic variables on the bat fauna of a granitic cave and its surroundings in the state of São Paulo, Brazil

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Abstract: In the present study we analyzed the bat assemblage of the granitic cave Gruta do Riacho Subterrâneo and its surroundings (Itu, São Paulo state, Brazil) aiming to verify the influence of seasonality on its species composition and population abundances. Five samplings were carried out with three days of duration each, along the period from October 2013 to September 2014. Captures of bats were performed by setting mist nets in cave entrances, its interior and surroundings, making a total capture effort of 6,090 m^2.h. Our results indicate that this cave is shelter for a rich bat assemblage with fifteen species captured. *Carollia perspicillata*, *Desmodus rotundus* and *Myotis* sp. were the most abundant species. A comparison of the assemblage composition with that of other caves of São Paulo state revealed that its composition is very similar and typical of the Atlantic Forest Atlantic cave chiropterofauna independently of cave lithology. A multiple regression analysis performed to check for the existence of correlation between the seasonal fluctuation of the climatic variables temperature, pluviosity and air humidity did not reveal significant relationships among these and the changes in the abundance of bats. However, the analysis of canonical correspondence including these variables and also moonlight luminosity indicated a significant relationship of the changes in bat abundance with the air relative humidity. Changes in bat abundances are probably related to the seasonality in food availability. The accumulation curve obtained from the relationship between the accumulated richness of species and the number of samples showed that more samplings are required to reach the asymptote of species richness. Considering that Gruta do Riacho Subterrâneo is the largest granitic cave in Brazil and that it shelters a high number of bat species, including common and rare species, we suggests the preservation of this cave for maintenance of bat diversity in São Paulo state.
Keywords: *bats, granitic cave, seasonality, biodiversity.*

ROCHA, A.D., BICHUETTE, M.E. **Influência de variáveis abióticas sobre a fauna de morcegos de uma caverna granítica e seu entorno no estado de São Paulo, Brasil**. Biota Neotropica. 16(3): e20150032. http://dx.doi.org/10.1590/1676-0611-BN-2015-0032

Resumo: No presente estudo analisamos a assembleia de morcegos da caverna granítica Gruta do Riacho Subterrâneo e sua área de entorno (Itu, estado de São Paulo, Brasil) com o objetivo de verificar a influência da sazonalidade na composição de espécies e na abundância das populações. Foram realizadas cinco amostragens com duração de três dias cada, ao longo do período de outubro de 2013 a setembro de 2014, utilizando redes-de-neblina instaladas nas entradas, no interior da caverna e em seu entorno, totalizando um esforço de captura de 6.090 m^2.h. Nossos resultados indicam que esta caverna abriga uma assembleia rica, onde foram capturadas quinze espécies. *Carollia perspicillata*, *Desmodus rotundus* e *Myotis* sp. foram as espécies mais abundantes. A comparação da composição desta assembleia com a de outras cavernas do estado de São Paulo revelou que suas espécies são similares e típicas da quiropterofauna de cavernas da Mata Atlântica, independentemente da litologia das mesmas. A análise de regressão múltipla utilizada para a relação entre a variação sazonal das variáveis temperatura, pluviosidade e umidade relativa do ar não revelou correlação significativa entre estas e as variações na abundância das diferentes espécies de morcegos. Contudo a análise de correspondência canônica incluindo, além destas, a variável luminosidade da lua indicou uma correlação significativa com a umidade relativa do ar. As variações nas abundâncias dos morcegos provavelmente estão relacionadas à sazonalidade na disponibilidade de alimento. A curva de acumulação obtida pela relação entre a riqueza de espécies e o número de amostragens acumuladas mostrou que mais amostragens são necessárias para atingir a assíntota da riqueza de espécies. Considerando-se que a Gruta do Riacho Subterrâneo é a maior caverna granítica do Brasil e que abriga uma elevada riqueza de espécies de morcegos, incluido raras e comuns, nós sugerimos a preservação desta caverna para a manutenção da diversidade morcegos no estado de São Paulo.

Palavras-chave: *morcegos, caverna granítica, sazonalidade, biodiversidade.*

Introduction

Bats are one of the most successful groups of mammals on Earth. This, in part, is due to several evolutionary traits acquired, such as flight capacity, echolocation ability and nocturnal habits. In Brazil, bats represent 25% of the mammals described, totaling 179 species distributed in 68 genera and nine families, a significatively high diversity that represents 16% of the species described in the world and still increasing with the new findings in this decade (Gardner 2007, Paglia et al. 2012, Nogueira et al. 2014).

Bats are the only mammals that successfully explore the caves (Kunz 1982), permanently establishing trogloxenes populations (organisms with epigean source populations) that use resources from subterranean habitats, as shelter, for instance, but also bringing organic material from the epigean environment (Trajano 2012). Ecologically, bats have an important role on caves, because their faeces, known as guano, represent an energy source to the subterranean biota (Gnaspini & Trajano 2000).

Many species of bats use caves as shelters (Kunz & Lumsden 2003) seeking protection against climate variation, predation and potential competitors (Kunz 1982). Their preferences regarding shelters are also influenced by structure, distribution and diversity of caves and abundance of resources (Trajano 1985, Kunz & Lumsden 2003).

There are a number of studies in the Southeast Brazil regarding cave chiropteran assemblages. Among them are the studies on cave-dwelling bats at the Atlantic Forest in Vale do Ribeira, São Paulo carried out by Trajano (1985) and by Campanhã & Fowler (1993) also in remnant fragments of Atlantic Forest in Corumbataí municipality, São Paulo. For other regions of Brazil, in the North it can be cited the study of Cajaiba (2014) in sandstone caves in Uruará, Pará; in the west central region Bredt et al. (1999) studied the bat assemblages of 20 caves in Federal District, which have the Cerrado as the predominant vegetation and Esbérard et al. (2005), who studied the caves of an Environmental Protection Area caves also predominantly in the Cerrado, in the state of Goiás. In the South region Arnone & Passos (2007) studied the bat assemblages caves located in mixed Atlantic Rainforest in the state of Paraná. Bat assemblages of Northeast have were analized by Sbragia & Cardoso (2008) for caves in Cerrado Atlantic Forest and Caatinga in Bahia. These authors evidenced the relevance of studies on cave environments, due to their capacity to provide shelters for species that are relevant to the functioning of these habitats and associated communities. A review of data on bat assemblages of several Brazilian caves is provided by Guimarães & Ferreira (2014).

Bats occupy caves of different lithologies, as observed by Trajano (1985) and by Bredt et al. (1999), who concluded that the selection of cave shelters can be more influenced by the amount of other caves in the area than by the cave characteristics. Àvilla-Flores & Medellín (2001) found that for bat assemblages of 18 caves in Mexico, the lithology (limestone and silisiclastic caves) had no significant effect on bat selection of caves. Nevertheless, cave attributes as size and complexity can be important for bat assemblage species composition. For example, Brunet & Meddellin (2001) also for bat cave assemblages in Mexico found a positive relationship between species richness and cave surface area and complexity.

Granitic caves are relatively rare when compared to carbonatic ones (Juberthie 2000). Usually granitic caves are small, may vary in shape (Twidale & Bourne 2008) and their final structure is shaped mainly by the power of water (Romaní et al. 2010). They are often formed by large agglomerates of blocks with many openings, becoming natural entrances to the subterranean environment. The high number of openings observed in granitic caves leads to a direct influence of the surface environment on their communities (Romaní et al. 2010). Studies comparing granitic and other lithology caves regarding bats species composition have not yet been published in Brazil.

Several studies on bat populations from different regions analyze the influence of seasonality (dry and wet seasons) on the assemblage composition, diet and population densities showing that the main factor for changes is the food availability throughout the year, which is in turn influenced by seasonality (Stevens & Amarilla-Stevens 2012). This fact was evidenced for bat assemblages from the Amazonian forest (Bobrowiec et al. 2014) and from Atlantic Rainforest (Ribeiro-Mello 2009, Ortêncio-Filho et al. 2010, Stevens & Amarilla-Stevens 2012, Stevens 2013, Lourenço et al. 2014). For cave bats from Atlantic Rainforest in the Alto Ribeira region, São Paulo state, Trajano (1985) and Arnone (2008) suggested the existence of seasonality, at least for some species.

We analyzed herein the bat assemblage of the granitic cave Gruta do Riacho Subterrâneo and its surroundings. The analysis is based on the changes in the species composition and population relative abundances during twelve months, involving periods of contrasting pluviosity to test the hypothesis that seasonality affect bat assemblages.

Methodology

1. *Studied Area.*

The cave studied is the “Gruta do Riacho Subterrâneo” (23°16’10.40”S, 47°13’49.79”W, 726 m above sea level), located in the city of Itu - state of São Paulo, Brazil, and belongs to Itu’s post-orogenic granite suite (Grupo Pierre Martin de Espeleologia 2014) (Figure 1 A, B). This cave is amongst the six largest granite caves in the world, and it is so far the largest of the Southern Hemisphere, with approximately 1,415 meters of linear development, formed by the superposition of several blocks. The cave has many entrances offering various routes for bats.

The region where the cave is located is characterized by humid subtropical climate with dry winters and warm summers (Cfa) according to Köeppen (1948). The regional vegetation is a transition of Cerrado,

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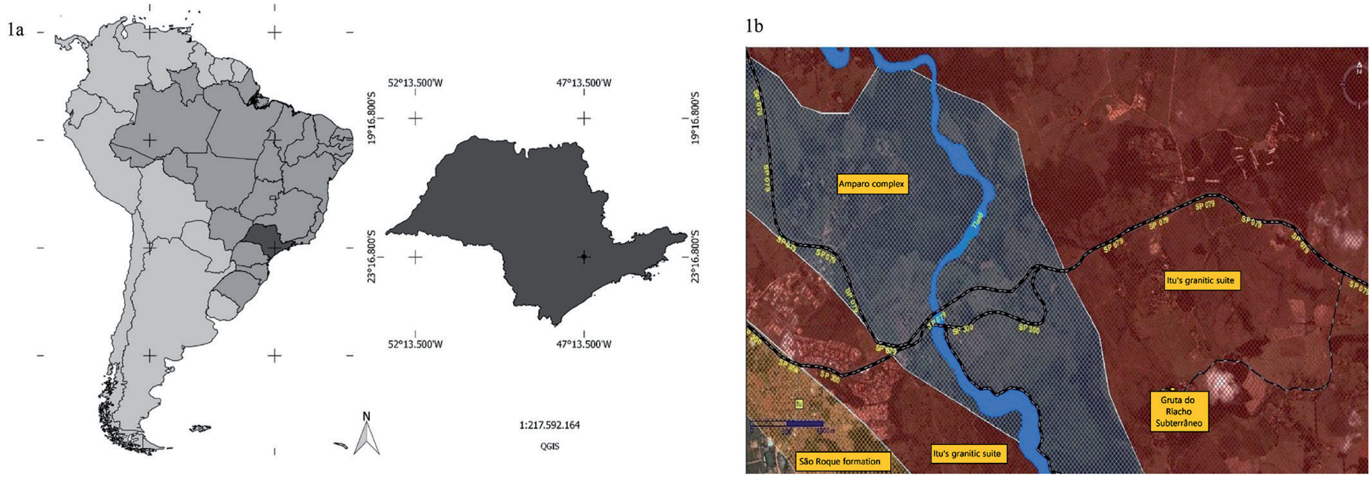


Figure 1. (A) Maps showing the location of Gruta do Riacho Subterrâneo in South America and in Itu municipality, SP. (B) Geological formation (Source: Grupo Pierre Martin de Espeleologia, 2014).

ombrophilous dense forest, stational semidecidual forest, and the ecotone zone among these vegetations (Rizzini 1997). Vegetation around the cave is nevertheless under strong anthropogenic pressure since the beginning of economic development in the São Paulo state highlands (Grupo Pierre Martin de Espeleologia 2014). In the surrounding area there is a small stream close to the cave and another in its interior, providing water resources to the bat assemblage and other communities present. Also, there is a small cave at a few meters distance and a large one at approximately 6 Km. It must be pointed out that several fires happened in 2010 in the surroundings of Gruta do Riacho Subterrâneo cave (Bichuette, M. E. personal observation) evidencing another type of disturbance the cave and its surrounding area undergo, making them even more altered.

Due to the granitic origin of the cave it is difficult to precisely establish the amount of existing saloons, since this is a non-regular cave along its whole extension being formed by superimposed granitic blocks, thus offering many entrances and possible routes to bats.

2. *Bat Samplings and Environmental Variables Measurements*

Samplings were carried out every two or three months in the period from October 2013 to September 2014. The first sampling (pilot) took place in August 2013, in order to define where the mist-nets could be placed. Table 1 shows the sampling dates, with October, December and March representing the rainy season and June and September representing the dry one (INMET 2014).

Table 1. Sampling days at Gruta do Riacho Subterrâneo and surroundings, Itu municipality, SP.

Year	Month and period	Days		
2013	October (rainy)	19	20	21
	December (rainy)	16	17	18
2014	March (rainy)	17	18	19
	June (dry)	28	29	30
	September (dry)	2	3	[a]

[a] No sampling due to rainfall.

Four or five mist-nets were set at the entrances of the caves in each of the three sampling days. Two other nets were set outside the cave (in a pasture and near a stream), in the first and in the third sampling days. These places were defined considering their dimensions and accessibility (Figure 2). It is important to emphasize that the cave has many entrances, so it is difficult to cover all possible routes for bats. The mist nets had three different sizes (4 x 3 m; 6 x 3 m and 7 x 3 m), installed in according to the entrance sizes. In the pasture we used a 7 x 3 m net and close the stream a 4 x 3 m net. The nets were set before sunset and were exposed for five hours, with regular checks every 20-30 minutes.

After captured, the specimens were identified on-site at species or genus level, and some were kept to allow a more accurate identification in the laboratory by utilizing specific identification keys (Vizzoto & Taddei 1973, Gardner 2007 and Reis et al. 2013). The specimens were deposited in the reference collection at Subterranean Studies Laboratory of the Federal University of São Carlos (LES - UFSCar).

Measurements of temperature (°C) and relative humidity (%) were obtained with a thermo-hygrometer (Instrutherm THAL-300; 0.1 resolution ± 5.0% accuracy) and monthly precipitation values were obtained from the National Institute of Meteorology (INMET, 2014), from the nearest station of Itu municipality. The moon phase was recorded in each collection day and posteriorly checked in a moon phase Table for each sampling day and month. To attribute a value for the moonlight luminosity of each lunar phase we used the following scale: new moon = one, horning = two, waning = tree, and full moon = four, as usually suggested for quantitative analysis based on qualitative evaluations.

3. *Data Analysis*

Capture effort was calculated according to Straube & Bianconi (2002). A sample-based species accumulation curve was plotted to analyze the Sufficiency Sampling Size, using "EstimateS" software (Colwell 2005). The relative abundance of each species was calculated and expressed as a percentage of the total assemblage abundance, in order to show their representativeness in the assemblage.

Figure 2. A view of sampling mist-nets at Gruta do Riacho Subterrâneo cave, Itu municipality, SP. (Author: Ives Arnone).

A multiple regression analysis was performed to verify the influence of the abiotic variables (temperature, relative humidity and pluviosity) over the abundance of the assemblage. Later on, the Akaike information criterion (AIC) was applied, in order to select the best representative model (Akaike 1970). A canonical correspondence analysis (CCA) was performed to verify the influence of the abiotic data matrix (temperature, relative humidity, moonligth and pluviosity) over the species abundance and assemblage richness. These analyses were carried out using the statistical software "R Development Core Team 2011".

Results

The capture effort was 6,090 m². h resulting on 113 captured individuals belonging to 15 species and two families. Eight species were recorded in the mist-nets settled in the cave (entrance and interior) and 11 were recorded in the cave surroundings, approximately 20 meters from the rocky outcrop that contains the cave (near the external stream and near the pasture) (Table 2). Amongst the captured bats, around 80% belonged to the Phyllostomidae family, while the others were represented by species belonging to the Vespertilionidae family. In Figure 3 the species accumulation curve is presented showing that the curve asymptote was not reached.

Figure 4 shows the relative abundances of the species, organized according to the environment in which they were found (outside or inside the cave). Some species were more abundant, such as *Carollia perspicillata* (30.8%), *Desmodus rotundus* (29.6%) and a species of genus *Myotis* sp. (23.4%), while others were much less abundant and even had single capture events, such as *Anoura geoffroyi*, *Micronycteris* sp., *Chiroderma doriae*, *Sturnira tildae* and *Eptesicus diminutus*.

Bat abundances were not related to environmental variables as indicated by the multiple regression analysis (F = 0.344, df = 3, p = 0.833; temperature, p = 0.495; relative humidity, p = 0.587 and pluviosity, p = 0.525). The application of the Akaike information criterion showed that using the three abiotic variables together would be the model resulting less information loss in data analysis. In the canonical correspondence analysis the relative humidity was the most important factor in the first axis, positively related to the bat assemblage richness and population abundances of the species *A. caudifer*, *P. lineatus, A. fimbriatus* and *S. lilium* in the months of March and September and negatively related with the abundances of *Micronycteris* sp. and *A. geoffroyi* in the months of December and October (Table 3). Pluviosity representing the main factor in axis two was not significantly related with bat abundances (Figure 5 A). The variables temperature and moonlight were not significantly related to the abundance of the species (Fig. 5 B).

Table 2. Species recorded and feeding habits for bat assemblage of Gruta do Riacho Subterrâneo and surroundings, Itu municipality, SP.

Family	Subfamily	Specie	Feeding habit	Cave	Surroundings	Captures
Phyllostomidae	Desmodontinae	*Desmodus rotundus* (E. Geoffroy, 1810)	Haematophagus	33	3	36
	Glossophaginae	*Anoura caudifer* (E. Geoffroy, 1818)	Nectarivores/Onivores	1	1	2
		Anoura geoffroyi Gray, 1838	Nectarivores/Onivores	1	0	1
		Glossophaga soricina (Pallas, 1766)	Nectarivores/Onivores	8	0	8
	Phyllostominae	*Chrotopterus auritus* (Peters, 1856)	Carnivores	2	1	3
		Micronycteris sp. Gray, 1866	Insectivores/Frugivores	1	0	1
	Carolliinae	*Carollia perspicillata* (Linnaeus, 1758)	Frugivores/Onivores	32	14	46
	Stenodermatinae	*Artibeus fimbriatus* Gray, 1838	Frugivores/Onivores	0	2	2
		Artibeus lituratus (Olfers, 1818)	Frugivores/Onivores	0	2	2
		Chiroderma doriae Thomas, 1891	Frugivores	0	1	1
		Platyrrhinus lineatus (E. Geoffroy, 1810)	Frugivores	0	2	2
		Sturnira lilium (E. Geoffroy, 1810)	Frugivores/Onivores	0	10	10
		Sturnira tildae de la Torre, 1959	Frugivores/Nectarivores	0	1	1
Vespertilionidae		*Eptesicus diminutus* Osgood, 1815	Insectivores	0	1	1
		Myotis sp. Kaup, 1829	Insectivores	26	0	26
Total: 2 families		15 species		104	38	142

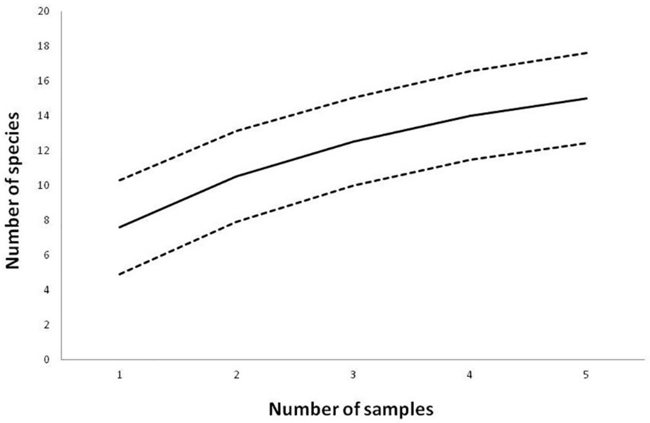


Figure 3. Accumulation curve for the species sampled at Gruta do Riacho Subterrâneo cave, Itu municipality, SP. Solid line represents the rarefaction curve and the dotted lines represent the 95% confidence limits.

Discussion

1. Bat Assemblage Composition

Among the 15 species recorded in this study only eight were found in the interior of Gruta do Riacho Subterrâneo, whereas the other seven were captured in the surroundings. From the eight species found in the interior of this granitic cave, six were already recorded by Trajano (1985) and by Arnone (2008) in Vale do Ribeira Atlantic Rainforest caves. Individuals from two genera, *Myotis* sp. and *Micronycteris* sp. could not be included in the comparison because they were not yet identified to the species level.

Among the seven species collected only in the surroundings of the Gruta do Riacho Subterrâneo (pasture or native vegetation) six have also been previously recorded by Trajano (1985) and by Arnone (2008) in Vale do Ribeira caves. Only one species, *Epitesicus diminutus,* captured in the pasture nearby the cave, was not previously recorded by these two

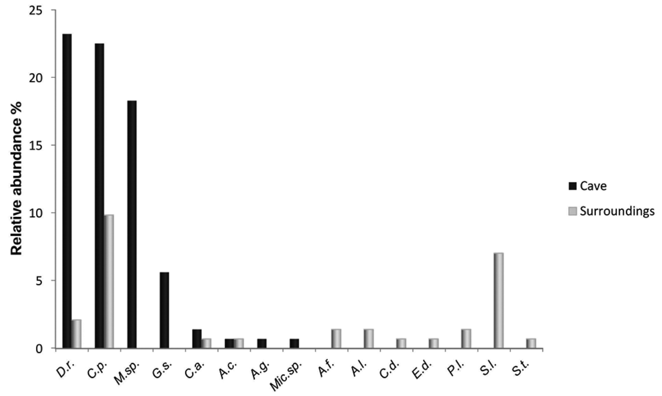
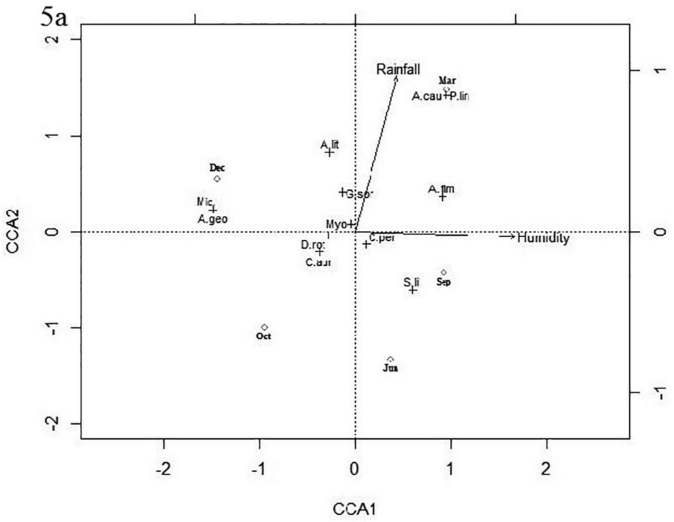


Figure 4. Bat relative abundances recorded for Gruta do Riacho Subterrâneo cave and surroundings, Itu municipality, SP. D. r. = *Desmodus rotundus*; C. p. = *Carollia perspicillata*; M. sp = *Myotis* sp.; G. s. = *Glossophaga soricina*; C. a. = *Chrotopterus auritus*; A. c. = *Anoura caudifer*; A. g. = *Anoura geoffroyi*; Mic.sp = *Micronycteris* sp.; A. f. = *Artibeus fimbriatus*; A.l. = *Artibeus lituratus*; C.d. = *Chiroderma doriae*; E.d.= *Eptesicus diminutus*; P.l. = *Platirhynus lineatus*; S. l. = *Sturnira lilium*; S.t. = *Sturnira tildae*.

Table 3. Canonical correspondence analysis valoes of the correlation between abiotic variables and bat species abundances.

	Axis 1	Axis 2	Pr (>r)
Temperature (°C)	-0.35900	0.93334	0.1417
Relative humidity (%)	0.99873	-0.05032	0.0166
Rainfall (mm)	0.2816	0.95952	0.2000
Moonlight	-0.58784	0.80898	0.2000
Cumulative proportion of explicability (humidity and Rainfall)	0.2717	0.4811	
Cumulative proportion of explicability (Temperature and moonlight)	0.2333	0.4414	



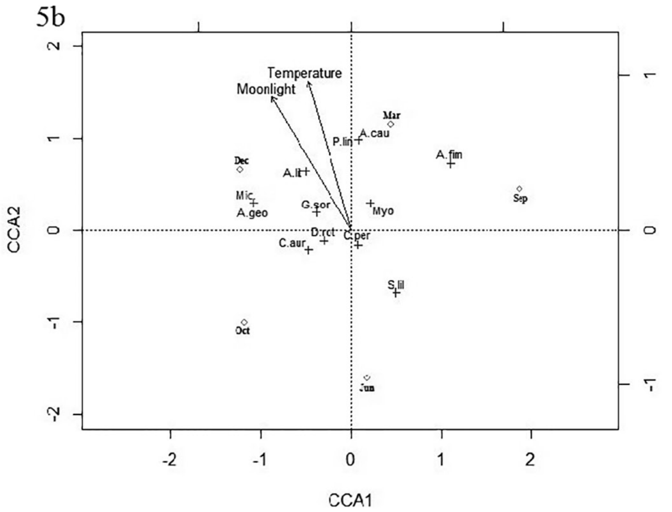


Figure 5A. Canonical correspondence analysis relating relative humidity and pluviosity and bat richness and abundances at Gruta do Riacho Subterrâneo and surroundings, Itu municipality, SP. A. cau = *Anoura caudifer*; P. lin. = *Platirhynus lineatus*; A. fim = *Artibeus fimbriatus*; A. lit = *Artibeus lituratus*; G. sor. = *Glossophaga soricina*; Mic = *Micronycteris* sp.; A. geo = *Anoura geoffroyi*; Myo = *Myotis* sp.; D. rot. = *Desmodus rotundus*; C. aur = *Chrotopterus auritus*; S. lil = *Sturnira lilium*; C. per = *Carollia perspicillata*.; Oct = October 2013; Dec. = December 2013; Mar = March, 2014; Jun = June 2014 and Sep = September, 2014.

Figure 5B. Canonical correspondence analysis relating temperature and moonlight on bat richness and abundances at Gruta do Riacho Subterrâneo and surroundings, Itu municipality, SP. A. cau = *Anoura caudifer*; P. lin. = *Platirhynus lineatus*; A. fim = *Artibeus fimbriatus*; A.lit = *Artibeus lituratus*; G.sor. = *Glossophaga soricina*; Mic = *Micronycteris* sp.; A. geo = *Anoura geoffroyi*; Myo = *Myotis* sp.; D. rot. = *Desmodus rotundus*; C. aur = *Chrotopterus auritus*; S. lil = *Sturnira lilium*; C. per = *Carollia perspicillata*.; Oct = October 2013; Dec. = December 2013; Mar = March, 2014; Jun = June 2014 and Sep = September, 2014.

authors in Vale do Ribeira caves. According to Reis et al. (2013) this species is found in primary and secondary forests and forest fragments, but also in open areas and urban environments. In São Paulo State it was previously recorded in the Atlantic Forest at Morro do Diabo State Park, where it is also a rare species (Reis et al. 1995).

Only three species abundant in the assemblage studied, the species *C. perspicillata* and *D. rotundus* and *Myotis* sp.. Two of them were also abundant in bat assemblages of other caves in São Paulo state (Trajano 1985, Campanhã & Fowler 1993, Arnone 2008).

Comparing the species composition of the bat assemblage in the granitic cave Gruta do Riacho Subterrâneo with those from caves with different lithologies in São Paulo State, as the calcareous caves of Alto do Ribeira (Trajano 1985, Arnone 2008) and the sandstone caves in the Preservation Area of Corumbataí (Campanhã & Fowler, 1999) it was found that they have very similar composition, although differing in the number of species. The assemblages of 36 caves so far inventoried probably belong to the same regional chiropterofauna, thus suggesting that cave lithology is not a determinant factor for bat assemblages taxonomic structure. One possible explanation for this similarity and also for the relatively high number of bat individuals captured in Gruta do Riacho Suberrâneo could be the fact that all these caves are located in remnants of the Atlantic Rainforest.

Bats from Gruta do Riacho Subterrâneo and its surroundings were classified into five trophic guilds with predominance of the frugivore guild, a fact that is corroborated in other studies that indicate that there is a predominance of this guild in the Neotropical Region (Sipinski & Reis 1995, Ribeiro-Mello 2009, Stevens & Amarilha-Stevens 2012, Bobrowiec et al. 2014, Lourenço et al. 2014). The predominance of frugivorous could, at least in part, be influenced by the sampling method used (mist nets) which is more effective for this guild, but less effective for insectivores due to their more refined echolocation system (Handley 1967, Trajano 1985, Arita 1993, Sipinski & Reis 1995).

Some species were rare being captured only once (*Anoura geoffroyi, Micronycteris* sp.*, Chiroderma doriae, Sturnira tildae* and *Eptesicus diminutus*), which could be due to the rarity of the species themselves, which usually tend to form small colonies and also to travel long distances to obtain food (Stevens & Amarilla-Stevens 2011). For instance, *Micronycteris* sp. live in small groups of six or less individuals (Emmons, 1990). *Chiroderma doriae* has not been found in the caves in the studies of Trajano (1985) and of Campanhã and Fowler (1999), but it was recorded by Arnone (2008) in Vale do Ribeira caves, who captured only one individual in this study site. It also occurred in small numbers in a semi-deciduous forest in the Ecological Station of Caetetus, São Paulo state (Pedro et al. 2001).

2. *Seasonality*

The results of multiple linear regression analysis did not show significance for the correlations among abiotic variables and the abundance of bats captured along the year. On contrary, the multivariate canonical analysis showed a significant positive relationship between the relative humidity and bat abundances. In the case of multiple regression, the tested variables are redundant (relative humidty is influenced by pluviosity and air temperature), which probably caused a noise in the analysis. Certainly the pluviosity and air temperature (affecting the relative humidity) influence the bat abundances and the canonical analysis supported this.

Corroborating this idea, we observed that the lowest captures of bats occurred in October 2013 and September 2014, which correspond to the transition period between dry and rainy seasons at the studied site. Bat populations were affected by seasonality, since the greatest bat abundances occurred in the warm and rainy summer months: December 2013 and March 2014, although a high number of bats was also captured in June 2014. The population of *Carollia perspicillata*, the most abundant species in Gruta do Riacho Subterrâneo cave followed this pattern, also displayed by less abundant species, such as *Sturnira lilium* and *Glossophaga soricina.* These species belongs to the frugivorous and nectarivorous trophic guilds, that are directly dependent on flowers and fruit production, which in Atlantic Rainforest and Cerrado have pronounced seasonality (Morellato et al. 2000; Batalha et al. 2004; Marques et al. 2007).

The different pattern observed for *Desmodus rotundus*, which showed high abundance along all the year (except for the last capture event in September 2014) is probably related to its hematophagous trophic guild, dependent only on the availability of mammals blood (as cattle and horses), its main food source, which is constant throughout the year in the region.

Stevens & Amarilla-Stevens (2011) in a study on bats in the Atlantic Forest in Paraguay found that some species displayed greater population changes in abundance, which were more related with seasonality than others. For example, the species *Artibeus lituratus* had a decline of more than 30% of its population during the colder season, but other species such as *Carollia perspicillata* and *Artibeus fimbriatus* did not show significant population density changes. The pattern observed for the latter species is similar to that of *Carollia perspicilata,* in which there was no direct relationship between this species abundance and temperature changes. The species *A. lituratus* was also captured, but its extremely low abundance in Gruta do Riacho Subterrâneo, with only two captures during all study, does not allow inferences regarding seasonality. Also, Ribeiro-Mello (2009) and Ortencio-Filho et al. (2010) showed that changes in temperature throughout the year influences the structure of bat community, and that this can be associated with a change in the feeding sites due to the decrease in food availability in the local area. Similarly, in Gruta do Riacho Subterrâneo cave, the lowest abundances of bats in September 2014 were probably due to the lower food availability, since this was the driest period in this one year sampling.

Another environmental factor that can influence cave bat capture is the moonlight intensity. It was shown by Crespo et al. (1972) in Mexico that *Desmodus rotundus,* a haematophagous species, and by Morrison (1978, 1980) that some frugivorous species in Panamá, Canal Zone, presented lunar phobia. In the present study there was no relationship between the number of bat captures and the moon light phases.

Conclusion

This study showed that Gruta do Riacho Subterrâneo cave is an important shelter for a rich assemblage of bats, including common and rare species. Althought the climatic variables tested had not shown a significance according to the multiple regression analysis, the changes in population abundances could be due the seasonality in food availability (corroborated by canonical analysis, considering the air relative humidity). The preservation of this cave is important and urgent to the maintenance of bat diversity in general, also contributing to the conservation of the chiropterofauna of São Paulo state.

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Addisonia enodis (Vetigastropoda: Lepetelloidea) associated with an elasmobranch egg capsule from the South Atlantic Ocean and the discovery of the species from deep waters off northeastern Brazil

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LIMA, S.F.B., SIMONE, L.R.L., GUIMARÃES, C.R.P. ***Addisonia enodis* (Vetigastropoda: Lepetelloidea) associated with an elasmobranch egg capsule from the South Atlantic Ocean and the discovery of the species from deep waters off northeastern Brazil**. Biota Neotropica. 16(3): e20160202. http://dx.doi.org/10.1590/1676-0611-BN-2016-0202

Abstract: A gastropod specimen of the subfamily Addisoniinae Dall, 1882 is reported here for the first time associated with an elasmobranch egg capsule from the South Atlantic Ocean. A specimen of *Addisonia enodis* Simone, 1996 was found living inside an egg capsule of *Atlantoraja castelnaui* (Miranda Ribeiro, 1907) (Arhynchobatidae Fowler, 1934) from shallow waters off southeastern Brazil. Previous studies have reported the association of members of the genus *Addisonia* Dall, 1882 only with the egg capsules of sharks from the family Scyliorhinidae Gill, 1862 and skates from the family Rajidae de Blainville, 1816. Other specimens of *A. enodis* are also here reported to occur off northeastern Brazil based on shells found in deep waters off the state of Sergipe, which fills a gap in its distribution in the Southwestern Atlantic to the north of this region. *Addisonia enodis* was recognized as a synonym of *A. excentrica* (Tiberi, 1855). However, we consider *A. enodis* as a valid species until further data clarify this issue based on a large sample of *Addisonia* from Brazilian waters.
Keywords: *Gastropoda, Addisoniidae,* Addisonia excentrica*, South America, deep sea.*

LIMA, S.F.B., SIMONE, L.R.L., GUIMARÃES, C.R.P. ***Addisonia enodis* (Vetigastropoda: Lepetelloidea) associada a uma cápsula ovígera de elasmobrânquio no Oceano Atlântico sul e a descoberta da espécie em águas profundas no nordeste do Brasil**. Biota Neotropica. 16(3): e20160202. http://dx.doi.org/10.1590/1676-0611-BN-2016-0202

Resumo: Um espécime de gastrópode da subfamília Addisoniinae Dall, 1882 é reportado aqui pela primeira vez associado a uma cápsula ovígera de elasmobrânquio encontrada no Oceano Atlântico sul. Um espécime de *Addisonia enodis* Simone, 1996 foi achado vivendo na parte interna da cápsula ovígera de *Atlantoraja castelnaui* (Miranda Ribeiro, 1907) (Arhynchobatidae Fowler, 1934) em águas rasas do sudeste do Brasil. Estudos prévios mencionaram a associação de membros do gênero *Addisonia* Dall, 1882 somente com desovas de tubarões da família Scyliorhinidae Gill, 1862 e raias da família Rajidae de Blainville, 1816. Outros espécimes de *A. enodis* também são aqui reportados para o nordeste do Brasil com base em conchas coletadas em águas profundas ao largo de Sergipe, as quais preenchem uma lacuna em sua distribuição no sudoeste do Atlântico. *Addisonia enodis* foi reconhecida como um sinônimo de *A. excentrica* (Tiberi, 1855). No entanto, essa espécie é considerada válida, neste estudo, até que mais dados demonstrem com clareza que tratam-se da mesma espécie.
Palavras-chave: *Gastropoda, Addisoniidae,* Addisonia excentrica*, América do Sul, mar profundo.*

Introduction

Lepetelloidea is one of the least investigated groups of marine gastropods, whose species richness is sporadically approached by alpha-taxonomic studies, especially from the South Atlantic Ocean (Simone 1996, Leal & Simone 2000, Simone & Cunha 2003, Lima 2014, Lima et al. 2016).

Lepetelloidean gastropods are far from being well known from shallow and deep waters off Brazil, where so far a total of three families and only five species were reported (Simone 1996, Leal & Simone 2000, Simone & Cunha 2003, Lima 2014, Lima et al. 2016): *Addisonia enodis* Simone, 1996 (Addisoniidae), *Copulabyssia riosi* Leal & Simone, 2000, *Lepetella furuncula* Lima, Guimarães & Simone, 2016 (Lepetellidae), *Notocrater christofferseni* Lima, 2014 and *Pseudococculina rimula* Simone & Cunha, 2003 (Pseudococculinidae).

Members of the subfamily Addisoniinae are classified into the single genus *Addisonia* Dall, 1882 (Roldán & Luque 2010), which was once represented by six Recent species (Dantart & Luque 1994, Bouchet & Gofas 2014), but currently only two congeners are considered valid (Roldán & Luque 2010, Bouchet & Gofas 2014): the amphi-Atlantic *A. excentrica* (Tiberi, 1855) (Roldán & Luque 1999, 2010) and the eastern Pacific (California) *A. brophyi* McLean, 1985 (McLean 1985, Haszprunar 1987, Dantart & Luque 1994, Simone 1996, Roldán & Luque 2010). *Addisonia brophyi* and *A. excentrica* are biologically associated with the elasmobranch egg capsules (McLean 1985, 1992, Dantart & Luque 1994, Roldán & Luque 1999, 2010) from shallow waters to bathyal depths (McLean 1985, Dantart & Luque 1994, Roldán & Luque 2010).

Addisonia enodis Simone, 1996 was described based on three specimens, one of which had soft parts that were dissected and supported the anatomical definition of the species. The main characters that differentiate *A. enodis* from congener species were the lack of radial sculpture, the position of the gill and shell muscle, as well as the shape of the rachidian teeth (Simone 1996: 784). Roldán & Luque (2010: 209) did not recognize conchological, radular and geographical differences in *A. enodis* Simone, 1996 for its separation as a different species, considering it a synonym of *A. excentrica*.

The present paper recognizes *Addisonia enodis* as a valid species and extends its distribution to northeastern Brazil based on specimens found in deep waters off the state of Sergipe. Furthermore, this genus is reported here for the first time associated with an elasmobranch egg capsule from the South Atlantic Ocean.

Material and methods

The present paper is based on the study of specimens collected by the R/V 'Natureza' using a trawling dredge between 365 to 500 meters depth on the continental slope of the states off Sergipe (northeastern Brazil) as part of benthic studies of the "Programa de Avaliação do Potencial Sustentável de Recursos Vivos na Zona Econômica Exclusiva (REVIZEE/2000–2001)", and specimens collected using a trawling dredge and trawl between 50 to 60 meters depth on the continental shelf of the states off Rio de Janeiro and São Paulo (southeastern Brazil) in 2001 to 2002. Other specimens collected in 1987 by the R/V 'Prof. W. Besnard' from the continental shelf off southeastern Brazil were reexamined (Simone 1996).

Elasmobranch egg capsules were obtained using a trawl net during the pink shrimp fishery on the continental shelf of the state off São Paulo. Capsules were placed in a container with ice and then taken to the Laboratory of Malacology (MZSP - Museu de Zoologia, Universidade de São Paulo, São Paulo, Brazil) for opening and removal of embryos. A living specimen of *Addisonia enodis* was found inside a capsule and then fixed in 70% ethanol.

Results

Addisoniidae Dall, 1882

***Addisonia enodis* Simone, 1996** (Figure 1)

1. *Material examined*

Types; BRAZIL, Sergipe: off São Francisco River mount (10º41.4'S, 36º18.7'W, dredged, 365 m), 28.X.2000, R/V 'Natureza' col. (MZSP 121236, 2 shells (Figure 1A-B): length 1.62 to 1.84 mm x width 1.19 to 1.57 mm x height 0.96 to 0.99 mm). Rio de Janeiro: off Arraial do Cabo (dredged, 50 m), 13.IX.2002 (MZSP 35845, 1 specimen: length 4.0 mm x width 3.04 mm x height 0.8 mm), (dredged, 107 m), 19.II.2002 (MZSP 35846, 1 shell: length 7.74 mm x width 6.7 mm x height 3.3 mm). São Paulo: off Santos (trawl, 50-60 m), IX.2001 (MZSP 54617, 1 specimen, inside capsule of *Atlantoraja castelnaui* (Ribeiro, 1907) (Figure 1C-D): length 10.5 mm x width 8.45 mm x height 4.3 mm).

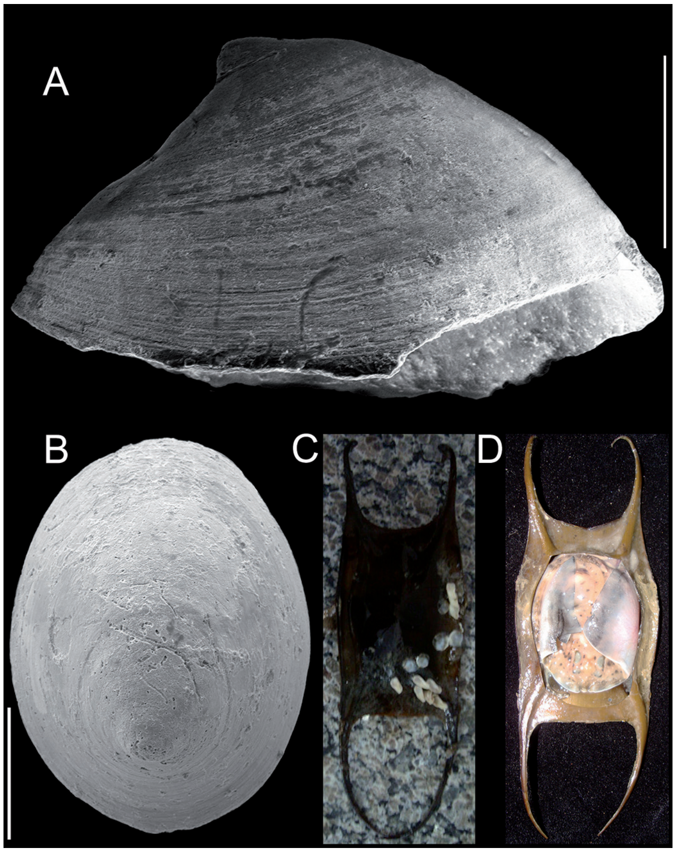

Figure 1. Specimens of *Addisonia enodis* and ovigerous capsule of *Atlantoraja castelnaui*. A, lateral view of *A. enodis* (scale = 500 µm); B, dorsal view of *A. enodis* (scale = 500 µm); C and D, ovigerous capsules of *Atlantoraja castelnaui* in which *A. enodis* were found.

2. *Ecology*

This species may be found on sand to mud bottoms between 50 to 365 m depth from Brazilian waters, associated with egg capsules of *Atlantoraja castelnaui*.

3. *Geographic distribution*

Northeastern (Sergipe state - present study) to Southeastern Brazil (Rio de Janeiro and São Paulo states - Simone 1996).

Discussion

We believe that conchological, anatomical and radular variations among specimens of *A. excentrica* and *A. enodis* need to be further investigated, especially based on a large sample of *Addisonia* from Brazilian waters, to ascertain if both species are actually conspecific. In this case, it is important to keep its status as a valid species rather than synonymize them, until further data, including molecular analyses, clarify this issue.

The present paper reports the association of *Addisonia* with an elasmobranch egg capsule of the family Arhynchobatidae Nelson, 2006 for the first time. *Addisonia enodis* was found living inside the egg capsule of *Atlantoraja castelnaui* from Brazilian shallow waters (between 50 to 60 m depth). This skate occurs from the Southeastern Brazil to Argentina waters, usually living between 10 to 100 m depth, and lay pairs of eggs on sandy or muddy bottoms (Bornatowski & Abilhoa 2012). Further information is not yet available for *A. enodis*, despite the fact that some of the abovementioned sharks and rays occur along the Brazilian coast.

The samples studied here from the continental slope off northeastern Brazil reinforce the possibility of live specimens of *Addisonia* being less abundant in deeper waters. It is likely that the specimens usually live in shallower waters (up to 100 m) due to greater abundance of elasmobranch egg capsules (especially of skates of the family Arhynchobatidae Nelson, 2006) which are biologically associated. Particularly, Scyliorhinidae, Rajidae, and now Arhynchobatidae, are potential families to find specimens of *Addisonia* and possibly other gastropods [e.g., *Choristella* Bush, 1897 (McLean 1992)] associated with their eggs in Brazilian waters based on common records in other eco-regions (McLean 1985, Ragozzi 1985, Villa 1985, Mclean 1992, Dantart & Luque 1994, Roldán & Luque 1999, 2010) and the greater richness of known elasmobranchs (especially Rajidae) on the Atlantic Coast of South America.

Most published studies on *Addisonia* have been based on specimens of *A. excentrica* collected from the north Atlantic and Mediterranean Sea (Dall 1882, 1889a, b, McLean 1985, Dantart & Luque 1994, Roldán & Luque 1999, 2010). These studies have shown considerable variability in the shell morphology (Dantart & Luque 1994), such as specimens with a lowly curved to moderately arched shell (Dall 1889a, b: pl. 25, fig. 1, pl. 44, fig. 11, pl. 63, fig. 100, Abbott 1974: fig. 206, McLean 1985: figs 1, 3, Dantart & Luque 1994: figs 68–71, Simone 1996: fig. 14, Roldán & Luque 2010: fig. 6A), while other shells are rather highly arched (McLean 1985: fig. 2); shells with a concentric sculpture poorly developed, bearing obsolete to fine lines (Dall 1889a,b: pl. 25, fig. 1, pl. 44, fig. 11, pl. 63, fig. 100, Abbott 1974: fig. 206, McLean 1985: figs 1–4, Dantart & Luque 1994: figs 68–69, Roldán & Luque 2010: fig. 6A) as well as a well-marked ornamentation represented by riblets (Dantart & Luque 1994: figs 70–72). These studies also show that the species has a variably deflected apex. Furthermore, teleoconch proportions vary considerably: 1.0 to 20.3 mm in length (McLean 1985, Roldán & Luque 1999). Specimens measuring 2.6 mm in length can already act as functional males (Roldán & Luque 1999) and represent adult shells. This considerable degree of variation appears more an assembly of different species rather than phenotypic plasticity of such a specific environmental strategist, as the samples of *A. enodis* has been more uniform in the shell shape.

Addisonia excentrica has been found living in association with egg capsules of sharks from the family Scyliorhinidae [e.g., *Scyliorhinus canicula* (Linnaeus, 1758)] and skates from the family Rajidae de Blainville, 1816 [e.g., *Raja clavata* Linnaeus, 1758 and *Raja* spp.] (Ragozzi 1985, Villa 1985, Dantart & Luque 1994, Roldán & Luque 1999, 2010). Arhynchobatidae, with three known species (Bornatowski & Abilhoa 2012), Chimaeridae, with three known species, Scyliorhinidae, with seven known species (*S. canicula* is not reported for Brazil), and Rajidae, with 28 known species (Rosa & Gadig 2014) are the families (with their respective numbers of species) of oviparous elasmobranchs recorded for the Brazilian coast (Bornatowski & Abilhoa 2012, Rosa & Gadig 2014). In general, members of these groups are demersal, living from the continental shelf down to the continental slope. Females deposit egg capsules on the sea floor and/or attached to structures on the bottom (Ebert & Stehmann 2013). *Addisonia excentrica* has been found in egg capsules of *S. canicula* and *Raja* Linnaeus, 1758 from the Mediterranean Sea between 47 to 426 m (Ragozzi 1985, Villa 1985, Roldán & Luque 2010). However, this species is collected more frequently between 50 to 200 m (Dantart & Luque 1994, Roldán & Luque 2010).

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The composition and new records of micro- and mesophytoplankton near the Vitória-Trindade Seamount Chain

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Abstract: In spite of the length of the Brazilian coastline, studies of marine phytoplankton species in offshore areas have been largely neglected. Regarding phytoplankton species composition, the aim was to assess biodiversity status, species frequency, and the similarities at different sites along the Vitoria-Trindade Seamount Chain. Phytoplankton net samples were analyzed. One hundred and seventy five infrageneric taxa were identified. They represented four phyla, viz., Cyanobacteria, Bacillariophyta, Dinophyta, and Ochrophyta. Dinophyta was the most diverse, and its genus *Tripos* the largest contributor. This genus also represented more than half of the most common taxa in the sampled area. Thirty-five new infrageneric taxa were recorded, 12 of which are new reports for Brazil. Neritic stations assemblages were markedly different from those of seamounts and deep offshore. The high species diversity, especially among dinoflagellates can be considered typical of tropical oligotrophic waters. The new records demonstrate the gap in knowledge of phytoplankton biodiversity in Brazil.
Keywords: *South Atlantic, Tripos, oceanic islands, phytoplankton, biodiversity.*

LUBIANA, K.M.F., DIAS JÚNIOR, C. **Composição e novos registros do micro e mesofitoplâncton nas proximidades da Cadeia Vitória-Trindade**. Biota Neotropica. 16(3): e20160164. http://dx.doi.org/10.1590/1676-0611-BN-2016-0164

Resumo: O fitoplâncton marinho é negligenciado nas áreas fora da plataforma continental brasileira. Este estudo objetiva avaliar o status da biodiversidade, a frequência de ocorrência das espécies e a relação de proximidade entre as estações amostradas ao longo da cadeia Vitória-Trindade de acordo com a composição de espécies do fitoplâncton. Analisamos amostras de rede e identificamos 175 táxons infra genéricos de quatro filos: Cyanobacteria, Bacillariophyta, Dinophyta e Ochrophyta. Dinophyta teve o maior número de táxons e seu gênero *Tripos* foi o mais diversificado. Esse também representou mais da metade dos táxons muito comuns. Foram identificados 35 novos registros de táxons infra genéricos na área estudada, dentre eles 12 são novos para o Brasil. A composição de espécies nas estações neríticas foi mais distinta das estações oceânicas e sobre os montes da cadeia submersa. A alta diversidade de espécies, especialmente dinoflagelados, é típica de ambiente de águas tropicais oligotróficas. Os novos registros demonstraram a lacuna de conhecimento a respeito da biodiversidade fitoplanctônica nas regiões oceânicas no Brasil.
Palavras-chave: *Atlântico Sul, Tripos, ilhas oceânicas, fitoplâncton, diversidade.*

Introduction

In spite of the long extent of the Brazilian coastline, the study of phytoplankton in offshore areas is incipient, notwithstanding the important role of this marine community as the source of more than 45% of primary production worldwide (Field 1998). Regarding group diversity, a high range in phyla is found, since many organisms are not descended from the same lineage. The greatest species-diversity is found among diatoms (13.776 species) and dinoflagellates (3.281) (Guiry & Guiry 2015), although other groups such as haptophytes, prasinophytes and cyanobacteria, are also significant. According to Barton et al. (2010), phytoplankton composition is driven by a latitudinal gradient of species richness, whereby tropical and subtropical zones prove to be the richest.

The Vitória-Trindade Seamount Chain is located off the central coast of Brazil. Starting 175km off the coast of Espírito Santo State and extending for 950km eastward, the seamounts are disposed almost linearly at 20° and 21°S (Almeida 2006), and situated between the parallels 28° and 38°W. The chain emerged on the seafloor during the Cenozoic, simultaneously with the westward movement of the South America Plate (Almeida 2006). Trindade Island and Martin Vaz Archipelago are located at the eastern end, the farthest location from the coast (Almeida 2006, Motoki et al. 2012). The last 17 submarine banks rise higher than 2.5km from the seafloor, with more than half reaching the euphotic zone (Motoki et al. 2012).

The Brazilian current, originating from the South Equatorial Current at 10°S (Silveira et al. 2000), passes through the region transporting an oligotrophic, warm and highly saline water mass (Brandini et al. 1997, Gaeta et al. 1999). Due to low nutrient availability, these environmental conditions favor phytoplankton species that are mixotrophic, heterotrophic or diazotrophic.

The only two studies available of phytoplankton close to the Vitória-Trindade Seamount Chain, both as a part of the REVIZEE program (Tenenbaum et al. 2006, 2007), were published in Portuguese, thereby precluding access by the international scientific community. Other organisms from this region, such as cetaceans (Wedekin et al. 2014), fishes (Pinheiro et al. 2009, 2015), barnacles (Young 1999), rhodolites (Pereira-Filho et al. 2012), etc, received more attention.

On considering the extreme importance of investigating the fundamental role of phytoplankton community in marine ecosystems, their high biodiversity in tropical waters, and the few studies currently available, the aims of this study were to assess: *(i)* biodiversity status, *(ii)* frequency of occurrence of each taxon, and *(iii)* station similarity, as regards taxonomic composition of micro- and mesophytoplankton communities, close to the Vitória-Trindade Seamount Chain.

Material and Methods

Samples were collected during November of the austral spring of 2003, aboard R.V. Antares of the Brazilian Navy. Collection was by vertical trawling up to 100m deep, using a specific net (20µm mesh size). After sample concentration, a formaldehyde solution was added (final concentration 2%). The samples were then incorporated into the Phytoplankton Collection of the Federal University of Espírito Santo. Vertical profiles of salinity, temperature, and dissolved oxygen were obtained with a CTD and oximeter coupled to a rosette.

Sixteen sampling stations were arranged in two transects (Figure 1), the first comprising nine stations located over the seamount chain (19°S), and the second, seven, most of which close to individual seamounts (20°S). Distances between stations varied from 150 to 200km. One station was located at the southern end of the Abrolhos Bank (E26), another two on the continental slope (E23 and E27), and two more over the Vitória and Montague seamounts (E30 and E32, respectively). A further three were relatively close to the Jasur, Dogaressa and Columbia seamounts (E34, E36, and E38, respectively), and just one station close to Trindade Island (E40). The last seven were situated in deep offshore areas (E14, E15, E16, E17, E18, E19 and E20). The Geographical Information System (GIS) approach with Diva-Gis program v7.5 (http://www.diva-gis.org/), as well as a shape file from the Brazilian Institute of Geography and Statistics (IBGE) (http://mapas.ibge.gov.br/en/interativos/arquivos/downloads), were applied to assessing submarine bathymetry at the sampled sites.

Phytoplankton species diversity was analysed with an Olympus CX41 light microscope. Detected organisms were drawn, measured and photographed under 200x and 400x magnification (USB camera Bel IS500 5.0mp and TSView 7 images acquisition program). Digital images and drawings are available upon request. Three slides were observed for each sample using transect methodology under 100x magnification. In sequence, these taxa were identified with the appropriate bibliography (Kofoid 1905, Cupp 1943, Wood 1954, Abé 1967a, b, Balech 1967, 1971, 1979, Hallegraeff 1987, Balech 1988, Hernández-Becerril 1991, Fernández & García 1998, Botes 2002, Polat & Koray 2002, Gómez 2005, Koening & Lira 2005, Tenenbaum et al. 2006, Haraguchi & Odebrecht 2007, Islabão & Odebrecht 2011, Garcia & Odebrecht 2012, Okolodkov 2014).

The International Code of Nomenclature for Algae, Fungi and Plants (IAPT) was applied to taxonomic classification, and AlgaeBase (Guiry & Guiry 2015) to checking nomenclature. In sequence, a table was set up for indicating taxa occurrence per station, whereby percentages were obtained. This facilitated classification into categories as, *(i)* very common (≥ 70% of stations); *(ii)* common (<70% and ≥ 30%); *(iii)* uncommon (<30% and ≥ 10%); and *(iv)* rare (<10%).

Subsequently, the search focused on phytoplankton species already reported in marine waters of Brazil, with the aim of finding new records (Koening & Lira 2005, Procopiak et al. 2006, Tenenbaum et al. 2006, 2007, Haraguchi & Odebrecht, 2007, Sousa et al. 2008, Villac et al. 2008, Villac & Tenenbaum 2010, Islabão & Odebrecht 2011, Proença et al. 2011, Tiburcio et al. 2011, Garcia & Odebrecht 2012, Jardim & Cardoso 2013, Menezes et al. 2015). All species synonyms were checked through AlgaeBase, in order to confirm the records in the publications consulted.

Hierarchical clustering methods are useful for evaluating species-composition correlations between stations, thereby revealing biogeographical connections with subjacent processes (Kreft & Jetz 2010). The pvclust package v1.32 (Suzuki & Shimodaira 2006), available in R program (http://www.r-project.org/), was applied in the present case. The Ward agglomerative method, based on the binary distance of taxonomic composition among stations, was employed for inferring hierarchical clustering. Statistical confidence of dendrogram nodes was defined by approximately unbiased (AU) support values that are less biased than the traditional bootstrap (Suzuki & Shimodaira 2006).

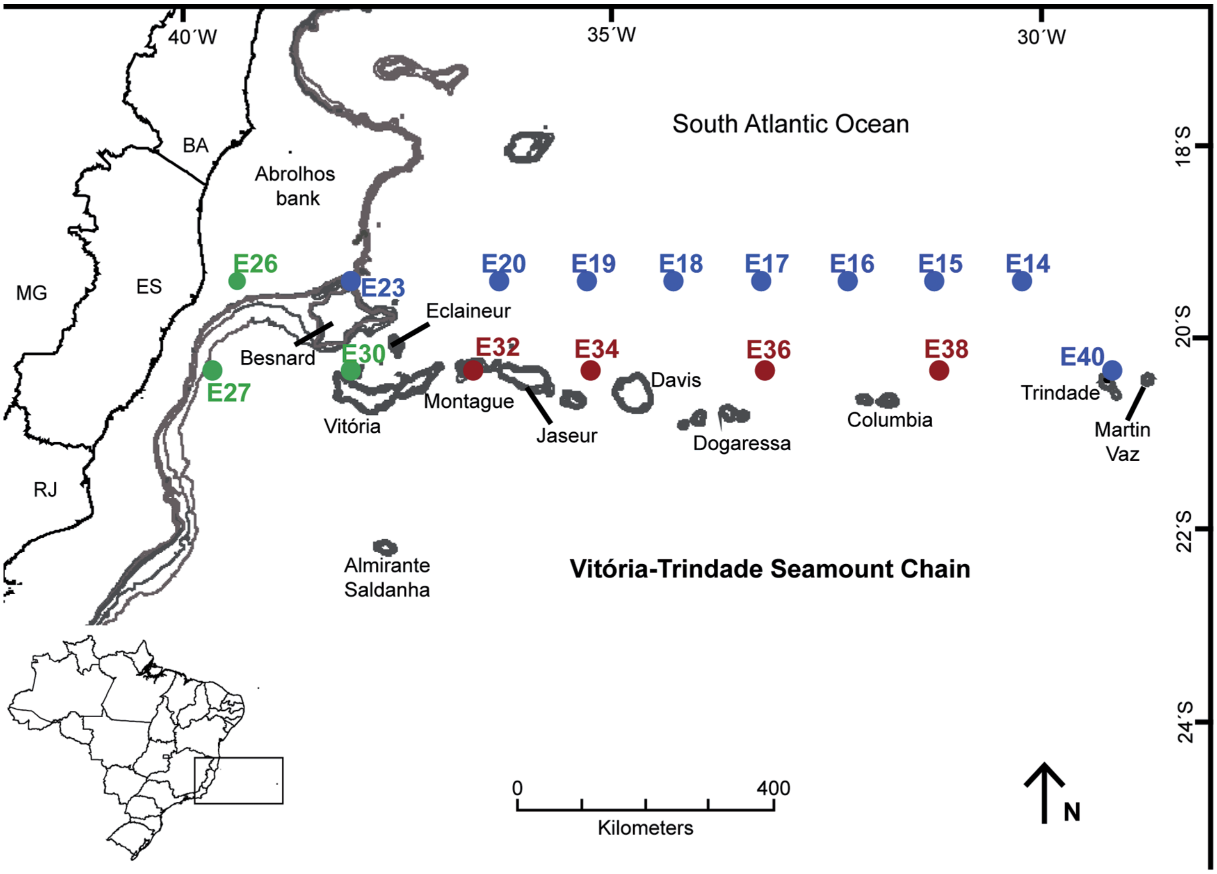

Figure 1. Map of the study area and sampling station sites. The colors of the stations are in accordance with Figure 5. Green stations are neritic, red, those closer to the Vitória-Trindade Seamount Chain, and bluethose located over deep offshore oceanic waters. The grey lines are isobaths (100- 2000 deep meters).

Results

The 175 identified infrageneric taxa of the micro and meso-phytoplankton identified were distributed among four phyla, viz., Bacillariophyta L.S.Dillon, Cyanobacteria Stanier ex Cavalier-Smith, Dinophyta F.E.Round and Ochrophyta Cavalier-Smith in Cavalier-Smiith & E.E.Chao (Table 1). The number of species registered per sample varied between 29 and 90 (average 54 ± 15.59).

The least represented phylum was Ochrophyta with only one species (0.57%) of Dictyochophyceae P.C.Silva, *Dictyocha fibula* Ehrenberg, followed by Cyanobacteria, with two species of *Trichodesmium* Ehrenberg ex Gomont (1.14%), Bacillariophyta, with 21 genera and 36 species (20.57%), and finally, Dinophyta, the most numerous, with 24 genera and 131 infrageneric taxa (74.85%) (Figure 2).

The family Rhizosoleniaceae De Toni contributed most to species diversity among the diatoms (9 species, 5.14%). Chaetocerotaceae Ralfs in Pritchard came next (5, 2.86%), followed by Coscinodiscaceae Kützing (4, 2.3%), and finally, Asterolampraceae H.L.Smith (4, 2.3%).

The order Gonyaulacales F.J.R.Taylor presented the highest number of identified taxa among all taxa (62, 35%), while its family Ceratiaceae Kofoid was the most diverse, due to the large number of *Tripos* Bory de St.-Vincent (45, 25.7%). The second, as regards orders and families, were Dinophysiales Kofoid (40 taxa, 22.52%) and Dinophysaceae Bütschli (30 taxa, 17.14%). In the Dinophysaceae, *Dinophysis* Ehrenberg and *Histioneis* Stein were the most representative, comprising 11 (6.3%) and 10 (5.7%) species, respectively.

According to frequency of occurrence, 17 taxa (9%) were very common, having been reported in more than 70% of the stations (Table 2 and Figure 3). All were dinoflagellates, ten of which *Tripos* (58.8%). *Ceratocorys horrida* Stein and *Tripos contrarius* (Gourret) F.Gómez were observed in all the stations. The common category consisted of 55 (31%) infrageneric taxa, two of which Cyanobacteria, five Bacillariophyta, and 48 Dinophyta, whereas the uncommon, 51 (29%), was comprised of one Ochrophyta, 17 Bacillariophyta, and 33 Dinophyta. Among rare taxa, 50 (28.5%) were recorded. Once again, dinoflagellates were the most diverse (31 species), followed by diatoms (19 species).

Thirty five new records of micro-and mesophytoplankton infrageneric taxa were found in the Vitória-Trindade Seamount Chain and continental shelf of Espírito Santo State, this including 12 new reports for Brazil (Table 3 and Figure 4). At a depth of 100 meters, and among stations, there was no significant change in environmental variables. Salinity varied between 37.39 and 37.055 (average 37.25) and temperatures 25.55 and 24.01°C (average 24.7ºC), thereby showing the predominance of tropical water in surface layers. Average dissolved oxygen was 4.64mg/l, this varing from 4.95 to 4.32mg/l.

Table 1. Taxa occurrence frequency throughout the sampled stations

Taxa Frequency Occurrence																		
Taxa	**Sampling stations**																	
	E14	**E15**	**E16**	**E17**	**E18**	**E19**	**E20**	**E23**	**E26**	**E27**	**E30**	**E32**	**E34**	**E36**	**E38**	**E40**	**Number of occurrences**	**% of occurrences**
Cyanobacteria																		
Cyanophyceae																		
Oscillatoriales																		
Microcoleaceae																		
Trichodesmium thiebautii Gomont ex Gomont 1890		X		X					X		X	X				X	6	37.5
Trichodesmium erythraeum Ehrenberg ex Gomont 1892		X						X	X	X	X	X	X			X	8	50
Ochrophyta																		
Dictyochophyceae																		
Dictyochales																		
Dictyochaceae																		
Dictyocha fibula Ehrenberg 1839				X							X	X				X	4	25
Bacillariophyta																		
Bacillariophyceae																		
Bacillariales																		
Bacillariaceae																		
										X	X						2	12.5
Bacillariales sp. 1											X			X	X		3	18.75
Bacillariales sp. 2									X								1	6.25
Bacillariales sp. 3													X				1	6.25
Bacillariales sp. 4		X															1	6.25
Fragilariales																		
Fragilariaceae																		
Ceratoneis closterium Ehrenberg 1839										X							1	6.25
Naviculales																		
Pleurosigmataceae																		
Pleurosigma sp.1											X						1	6.25

Continued Table 1.

Taxa Frequency Occurrence																		
	Sampling stations																	
Taxa	**E14**	**E15**	**E16**	**E17**	**E18**	**E19**	**E20**	**E23**	**E26**	**E27**	**E30**	**E32**	**E34**	**E36**	**E38**	**E40**	**Number of occurrences**	**% of occurrences**
Coscinodiscophyceae																		
Asterolamprales																		
Asterolampraceae																		
Asterolampra marylandica Ehrenberg 1844	X	X	X	X			X						X	X	X	X	9	56.25
**Asteromphalus flabellatus* (Brébisson) Greville 1859		X															1	6.25
Asteromphalus heptactis (Brébisson) Ralfs in Pritchard 1861	X	X	X	X		X	X							X	X	X	9	56.25
***Asteromphalus stellatus* (Greville) J.J.Ralfs	X	X											X	X			4	25
Biddulphiales																		
Biddulphiaceae																		
Isthmia enervis Ehrenberg 1838											X	X	X				3	18.75
**Trigonium formosum* (Brightwell) Cleve 1867																X	1	6.25
Triceratilaes																		
Triceratiaceae																		
**Lampriscus shadboltianum* (Greville) Peragallo & Peragallo 1902											X						1	6.25
Chaetocerotales																		
Chaetocerotaceae																		
Bacteriastrum cf. *hyalinum* Lauder 1864									X								1	6.25
Chaetoceros cf. *coarctatus* Lauder 1864					X		X					X	X		X		5	31.25
**Chaetoceros* cf. *concavicornis* Mangin 1917		X															1	6.25
**Chaetoceros mitra* (Bailey) Cleve 1896		X													X		2	12.5
Chaetoceros peruvianus Brightwell 1856		X											X		X		3	18.75
Coscinodiscales																		
Coscinodiscaceae																		
Coscinodiscus cf. *centralis* Ehrenberg 1844						X					X		X				3	18.75
Coscinodiscus granii Gough 1905														X		X	2	12.5
Coscinodiscus sp.1							X			X			X		X		4	25
Coscinodiscus sp.2									X								1	6.25

Continued Table 1.

Taxa Frequency Occurrence																		
	Sampling stations																	
Taxa	**E14**	**E15**	**E16**	**E17**	**E18**	**E19**	**E20**	**E23**	**E26**	**E27**	**E30**	**E32**	**E34**	**E36**	**E38**	**E40**	**Number of occurrences**	**% of occurrences**
Hemidiscaceae																		
Hemidiscus cuneiformis Wallich 1860															X		1	6.25
Hemiaulales																		
Hemiaulaceae																		
Cerataulina pelagica (Cleve) Hendey 1937									X								1	6.25
Hemiaulus hauckii Grunow ex Van Heurck 1882		X		X									X	X	X		5	31.25
**Hemiaulus membranaceus* Cleve									X								1	6.25
Leptocylindrales																		
Leptocylindraceae																		
Leptocylindrus danicus Cleve 1889									X								1	6.25
Paraliales																		
Paraliaceae																		
Paralia sulcata (Ehrenberg) Cleve 1873									X								1	6.25
Rhizosoleniales																		
Rhizosoleniaceae																		
Guinardia delicatula (Cleve) Hasle in Hasle & Syvertsen 1997											X						1	6.25
Guinardia flaccida (Castracane) H.Peragallo 1892										X	X						2	12.5
Neocalyptrella robusta (G.Norman ex Ralfs) Hernández-Becerril & Meave del Castillo 1997									X	X							2	12.5
Proboscia alata (Brightwell) Sundström 1986										X		X	X				3	18.75
**Rhizosolenia castracanei* H.Peragallo 1888		X								X				X	X		4	25
Rhizosolenia hebetata Bailey 1856				X			X			X		X	X	X	X		7	43.75
Rhizosolenia imbricata Brightwell 1858		X								X							2	12.5
Rhizosolenia setigera Brightwell 1858										X							1	6.25
Rhizosolenia styliformis T.Brightwell 1858				X						X					X		3	18.75
Thalassiosirales																		
Thalassiosiraceae																		
**Thalassiosira leptopus* (Grunow ex Van Heurck) Hasle & G.Fryxell 1977					X											X	2	12.5

Continued Table 1.

Taxa Frequency Occurrence																		
Taxa	**Sampling stations**																	
	E14	**E15**	**E16**	**E17**	**E18**	**E19**	**E20**	**E23**	**E26**	**E27**	**E30**	**E32**	**E34**	**E36**	**E38**	**E40**	**Number of occurrences**	**% of occurrences**
Thalassiosira sp.1												X	X	X	X		4	25
Thalassiosira sp.2							X										1	6.25
Dinophyta																		
Dinophyceae																		
Dinophysiales																		
Amphisoleniaceae																		
Amphisolenia bidentata Schröder 1900	X	X										X			X	X	5	31.25
**Amphisolenia bifurcata* Murray & Whitting 1899	X		X	X												X	4	25
Amphisolenia globifera Stein 1883		X				X									X	X	4	25
**Amphisolenia schauinslandii* Lemmermann 1899	X	X	X				X	X			X	X				X	8	50
***Amphisolenia schroederi* Kofoid 1907							X										1	6.25
Dinophysaceae																		
Citharistes apsteinii F.Schütt 1895	X	X				X										X	4	25
**Citharistes regius* Stein 1883				X		X											2	12.5
Dinophysis acuminata Claparède & Lachmann 1859											X						1	6.25
Dinophysis argus (Stein) Abé		X	X				X									X	4	25
**Dinophysis caudata* Saville-Kent 1881									X								1	6.25
**Dinophysis fortii* Pavillard 1923									X								1	6.25
Dinophysis hastata Stein 1883	X					X	X					X			X		5	31.25
Dinophysis cf. *operculoides* (Schütt) Balech 1967		X		X		X	X	X	X		X	X	X	X	X	X	12	75
**Dinophysis pusilla* Jørgensen 1923	X	X		X								X		X	X		6	37.5
Dinophysis schuettii Murray & Whitting 1899	X	X	X	X		X	X	X			X	X	X	X	X	X	13	81.25
Dinophysis sp. 1									X								1	6.25
Dinophysis sp. 2							X					X					2	12.5
Dinophysis sp. 3		X															1	6.25
Histioneis cymbalaria Stein 1883	X	X	X	X												X	5	31.25
Histioneis elongata Kofoid & Michener 1911						X											1	6.25
***Histioneis garrettii* Kofoid & Michener 1907		X				X											2	12.5
Histioneis highleyi Murray & Whitting 1899												X					1	6.25
Histioneis inclinata Kofoid & Michener 1911													X				1	6.25
***Histioneis joergensenii* Schiller 1928						X											1	6.25

Continued Table 1.

	Taxa Frequency Occurrence																	
	Sampling stations																	
Taxa	**E14**	**E15**	**E16**	**E17**	**E18**	**E19**	**E20**	**E23**	**E26**	**E27**	**E30**	**E32**	**E34**	**E36**	**E38**	**E40**	**Number of occurrences**	**% of occurrences**
Histioneis megalocopa Stein 1883	X	X	X				X	X				X	X	X	X	X	10	62.5
Histioneis milneri Murray & Whitting 1899														X	X		2	12.5
Histioneis panaria Kofoid & Skogsberg 1928	X		X														2	12.5
Histioneis para Murray & Whitting 1899	X	X	X	X										X		X	6	37.5
***Metaphalacroma skogsbergii* L.-S.Tai in L.-S.Tai & Skogsberg 1934		X															1	6.25
***Ornithocercus cristatus* Matzenauer 1933		X		X													2	12.5
Ornithocercus heteroporus Kofoid 1907	X	X		X		X					X	X		X	X	X	9	56.25
Ornithocercus magnificus Stein 1883	X	X	X	X		X	X	X		X	X	X	X		X	X	13	81.25
Ornithocercus quadratus Schütt 1900	X	X		X		X											4	25
Ornithocercus steinii Schütt 1900	X	X	X	X	X	X	X	X		X			X	X	X	X	13	81.25
Ornithocercus thumii (Schmidt) Kofoid & Skogsberg 1928				X			X						X	X		X	5	31.25
Oxyphysiaceae																		
Phalacroma cf. *doryphorum* Stein 1883	X					X										X	3	18.75
Phalacroma circumcinctum Kofoid & Michener 1911		X	X														2	12.5
Phalacroma cuneus F.Schütt 1895	X	X	X	X		X	X			X			X		X	X	10	62.5
Phalacroma hindmarchii Murray & Whitting 1899		X				X								X			3	18.75
Phalacroma rapa Jørgensen 1923		X							X			X		X			4	25
Gonyaulacales																		
Ceratiaceae																		
Tripos arietinus (Cleve) F.Gómez 2013											X	X				X	3	18.75
Tripos azoricus (Cleve) F.Gómez 2013	X	X		X	X	X	X	X	X				X	X	X	X	12	75
Tripos candelabrus (Ehrenberg) F.Gómez 2013		X			X	X	X	X		X	X	X	X		X	X	11	68.75
Tripos carriensis (Gourret) F.Gómez 2013	X			X		X	X					X	X	X	X	X	9	56.25
**Tripos cephalotus* (Lemmermann) F.Gómez 2013	X	X		X	X		X			X					X	X	8	50
Tripos concilians (Jørgenen) F.Gómez 2013	X									X		X	X		X		5	31.25
Tripos contortus (Gourret) F.Gómez 2013	X	X		X	X	X	X	X	X				X	X	X		11	68.75
Tripos contrarius (Gourret) F.Gómez 2013	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	16	100
Tripos declinatus (Karsten) F.Gómez 2013	X	X	X	X	X	X	X	X	X		X	X	X	X	X	X	15	93.75
Tripos digitatus (Schütt) F.Gómez 2013				X			X										2	12.5
Tripos euarcuatus (Jörgenen) F.Gómez 2013	X	X	X		X	X				X		X	X	X	X	X	11	68.75

Continued Table 1.

Taxa	Taxa Frequency Occurrence																	
	Sampling stations																	
	E14	E15	E16	E17	E18	E19	E20	E23	E26	E27	E30	E32	E34	E36	E38	E40	Number of occurrences	% of occurrences
Tripos extensus (Gourret) F.Gómez 2013	X	X	X		X	X	X	X			X	X	X	X		X	12	75
Tripos furca (Ehrenberg) F.Gómez 2013									X								1	6.25
Tripos fusus (Ehrenberg) F.Gómez 2013	X	X	X		X		X	X		X		X	X	X	X	X	12	75
Tripos geniculatus (Lemmermann) F.Gómez 2013							X					X					2	12.5
Tripos gibberus (Gourret) F.Gómez 1883	X	X	X		X	X				X			X	X	X	X	10	62.5
Tripos gravidus (Gourret) F.Gómez 2013	X	X		X		X							X		X		6	37.5
Tripos hexacanthus (Gourret) F.Gómez 2013	X			X		X	X	X		X	X		X	X	X	X	11	68.75
Tripos horridus (Cleve) F.Gómez 2013	X	X	X		X		X	X		X							7	43.75
**Tripos horridus molle* (Kofoid) F. Gómez 2013									X	X							2	12.5
Tripos inflatus (Karsten) F.Gómez 2013		X															1	6.25
Tripos karstenii (Pavillard) F.Gómez 2013	X	X	X	X		X	X	X					X			X	9	56.25
Tripos limulus (Pouchet) F.Gómez 2013	X	X														X	3	18.75
Tripos lineatus (Ehrenberg) F.Gómez 2013		X															1	6.25
***Tripos longinus* (Karsten) F.Gómez 2013	X	X					X	X		X	X	X		X	X	X	10	62.5
Tripos longirostrus (Gourret 1883) F. Gómez 2013	X		X			X			X		X	X	X		X		8	50
Tripos lunula (Schimper ex Karsten) F.Gómez 2013	X		X	X	X	X				X			X	X	X		9	56.25
**Tripos macroceros* var. *gallicum* (Kofoid) F.Gómez 2013										X			X	X			3	18.75
Tripos macroceros (Ehrenberg) F.Gómez 2013	X	X	X	X		X	X	X	X	X	X		X	X	X	X	14	87.5
**Tripos massiliensis* var. *armatum* (Karsten) F. Gómez									X					X			2	12.5
Tripos massiliensis (Gourret) F.Gómez 2013	X	X	X	X		X	X		X	X	X		X	X	X	X	13	81.25
Tripos muelleri Bory de Saint-Vincent in J.V.Lamouroux et al. 1824	X		X	X					X	X	X		X			X	8	50
Tripos pentagonus (Gourret) F.Gómez 2013	X	X	X	X		X		X	X	X	X	X		X	X	X	13	81.25
Tripos praelongus (Lemmermann) Gómez 2013				X			X									X	3	18.75
Tripos pulchellus (Schröder) F.Gómez 2013	X	X	X	X	X	X	X	X				X	X	X	X		12	75
***Tripos pulchellus* f. *tripodioides* (Jørgensen) F. Gómez 2013	X	X	X			X	X		X	X	X	X	X	X	X	X	13	81.25
**Tripos ranipes* (Cleve) F.Gómez 2013	X	X		X	X	X	X		X				X			X	9	56.25
Tripos reflexus (Cleve) F.Gómez 2013		X	X	X		X										X	5	31.25
Tripos setaceum (Jörgensen) F. Gómez 2013					X												1	6.25
Tripos sumatranus (Karsten) F.Gómez 2013	X	X				X	X		X	X			X	X		X	9	56.25
**Tripos symmetricus* (Pavillard) F.Gómez 2013	X		X		X		X	X								X	6	37.5
Tripos teres (Kofoid) F. Gómez 2013	X	X	X	X	X	X	X	X			X	X	X	X	X	X	14	87.5

Continued Table 1.

Taxa	Taxa Frequency Occurrence																	
	Sampling stations																	
	E14	E15	E16	E17	E18	E19	E20	E23	E26	E27	E30	E32	E34	E36	E38	E40	Number of occurrences	% of occurrences
Tripos trichoceros (Ehrenberg) Gómez 2013									X	X							2	12.5
Tripos vultur (Cleve) F.Gómez 2013																X	1	6.25
**Tripos vultur* var. *japonicum* (Schröder) F. Gómez 2013	X			X	X	X				X		X	X	X	X	X	10	62.5
Cladopyxidaceae																		
Cladopyxis brachiolata Stein 1883	X	X	X	X		X	X					X	X	X	X	X	11	68.75
***Cladopyxis hemibrachiata* Balech 1964	X	X	X	X	X		X							X	X	X	9	56.25
Goniodomataceae																		
Triadinium polyedricum (Pouchet) Dodge 1981		X	X	X	X	X	X	X		X	X	X	X	X	X	X	14	87.5
Goniodoma sp.		X	X					X					X	X		X	6	37.5
Gonyaulacaceae																		
Gonyaulax birostris Stein 1883	X	X					X					X	X	X	X	X	8	50
***Gonyaulax milneri* (Murray & Whitting) Kofoid 1911g)		X															1	6.25
Gonyaulax polygramma Stein 1883	X	X					X			X	X	X	X	X	X	X	10	62.5
Gonyaulax sp.													X				1	6.25
**Spiraulax kofoidii* H.W.Graham 1942	X		X													X	3	18.75
Heterodiniaceae																		
***Heterodinium dispar* Kofoid & Adamson 1933		X												X	X		3	18.75
Heterodinium sp.1	X	X					X					X				X	5	31.25
Heterodinium sp.2		X															1	6.25
Protoceratiaceae																		
Ceratocorys gourretii Paulsen 1931	X	X	X			X		X				X		X	X	X	9	56.25
Ceratocorys horrida Stein 1883	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	16	100
Ceratocorys armata (Schütt) Kofoid 1910														X			1	6.25
Protoceratium cf. *aculeatum* (Stein) Schiller 1937		X	X				X							X			4	25
Protoceratium spinulosum (Murray & Whitting) Schiller 1937		X															1	6.25

Continued Table 1.

Taxa	Taxa Frequency Occurrence																	
	Sampling stations																	
	E14	**E15**	**E16**	**E17**	**E18**	**E19**	**E20**	**E23**	**E26**	**E27**	**E30**	**E32**	**E34**	**E36**	**E38**	**E40**	**Number of occurrences**	**% of occurrences**
Gymnodiniales																		
Gymnodiniaceae																		
Gymnodinium sp.														X			1	6.25
Noctilucales																		
Kofoidiniaceae																		
***Kofoidinium velleloides* Pavillard 1929												X					1	6.25
Peridiniales																		
Oxytoxaceae																		
Oxytoxum milneri Murray & Whitting 1899	X	X	X	X	X	X	X				X	X	X	X	X	X	13	81.25
Oxytoxum scolopax Stein 1883		X	X		X	X	X					X	X		X	X	9	56.25
Peridiniales inc. sed.																		
Corythodinium tesselatum (Stein) Loeblich Jr. & Loeblich III 1966		X															1	6.25
Corythodinium constrictum (Stein) F.J.R.Taylor 1976				X									X				2	12.5
Corythodinium sp. 1				X													1	6.25
Podolampaceae																		
Podolampas bipes Stein 1883	X	X		X		X	X	X	X			X	X	X		X	11	68.75
Podolampas elegans Schütt 1895	X	X	X	X	X	X			X				X		X		9	56.25
Podolampas palmipes Stein 1883	X	X										X	X	X		X	6	37.5
Podolampas spinifera Okamura 1912		X			X				X			X	X	X	X	X	8	50
Protoperidiniaceae																		
**Protoperidinium oceanicum* (Vanhöffen) Balech 1974									X								1	6.25
**Protoperidinium elegans* (Cleve) Balech 1974	X	X	X	X	X	X							X		X	X	9	56.25
Protoperidinium ovatum Pouchet 1883													X	X			2	12.5
**Protoperidinium pentagonum* (Gran) Balech 1974													X				1	6.25
Protoperidinium sp.1		X															2	12.5
Protoperidinium sp.2	X					X						X	X			X	5	31.25
Protoperidinium sp.3		X								X	X	X			X		5	31.25
Protoperidinium sp.4			X														1	6.25

Continued Table 1.

Taxa Frequency Occurrence																		
	Sampling stations																	
Taxa	**E14**	**E15**	**E16**	**E17**	**E18**	**E19**	**E20**	**E23**	**E26**	**E27**	**E30**	**E32**	**E34**	**E36**	**E38**	**E40**	**Number of occurrences**	**% of occurrences**
Prorocentrales																		
Prorocentraceae																		
Prorocentrum compressum (Bailey) Abé ex J.D.Dodge 1975		X				X											2	12.5
Prorocentrum gracile Schütt 1895				X	X												2	12.5
Prorocentrum balticum (Lohmann) Loeblich 1970			X				X		X				X			X	5	31.25
Pyrocystales																		
Pyrocystaceae																		
Pyrocystis fusiformis C.W.Thomson in J.Murray 1876	X	X		X		X		X	X					X		X	8	50
***Pyrocystis hamulus* var. *semicircularis* Schröder 1900					X												1	6.25
Pyrocystis pseudonoctiluca Wyville-Thompson in Murray 1876		X								X		X	X	X	X		6	37.5
Pyrocystis robusta Kofoid 1907		X	X	X	X	X			X	X		X	X	X	X	X	12	75
Dinophyceae not identified																		
Dinophyceae sp.1											X						1	6.25
Dinophyceae sp.2					X						X						2	12.5
Dinophyceae sp.3			X														1	6.25

** New occurence for Brazil; * New occurence for the state of Espírito Santo.

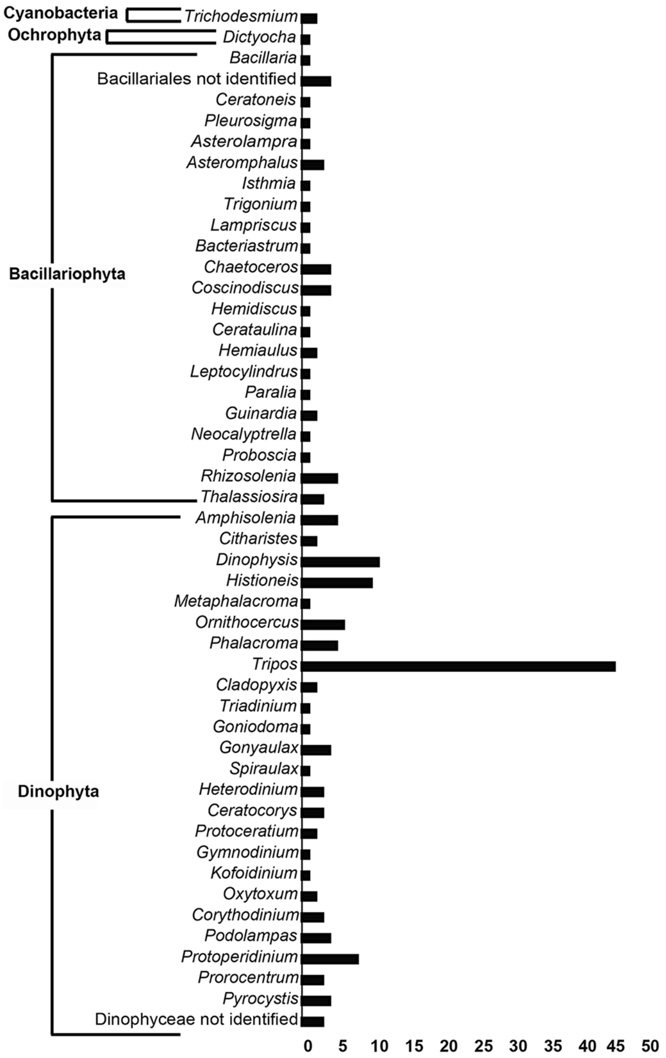

Figure 2. Genera biodiversity in the proximity of the Vitória-Trindade Seamount Chain. Four different phyla were identified. The number of species found for each genus is represented by bars.

Cluster analysis, by revealing differences in taxonomic composition among the various stations (Figure 5), facilitated their division into three main groups (Figure 1 and 5). The first, marked in green, and located closer to the continental shelf, were the most distant (71% AU value), the second, in red, were near the Vitória-Trindade Seamount Chain (81% AU), and the third, in blue, were in deep offshore areas (81% AU).

Discussion

The high infrageneric diversity among the micro and mesophytoplankton observed corroborates previous findings for tropical oligotrophic regions (Hallegraeff & Jeffrey 1984, Balech 1988). The high diversity of dinoflagellates collected in this area seems to be correlated with its preference for warm oligotrophic oceanic zones (Taylor et al 2007). Recently, the marine species of the genus *Ceratium* F.Schrank were renamed *Tripos* (Gómez et al. 2010, Gómez 2013) marine species of Ceratium (Dinophyceae, Alveolata. This presents, by far, the highest number of infrageneric taxa. Incidentally, it was the most common genus in the samples collected. Furthermore, besides being more specious in tropical regions (Dodge 1993, Dodge & Marshall 1994, Okolodkov & Dodge 1996, Tunin-Ley & Lemée 2013), it is usually the most diverse of the dinoflagellates (Taylor et al. 2007).

Table 2. Very common taxa in the stations (≥70% of occurrence).

Taxa	number occurrence	% occurrence
Ceratocorys horrida Stein 1883	16	100
Tripos contrarius (Gourret) F.Gómez 2013	16	100
Tripos declinatus (Karsten) F.Gómez 2013	15	93.75
Triadinium polyedricum (Pouchet) Dodge 1981	14	87.5
Tripos macroceros (Ehrenberg) F.Gómez 2013	14	87.5
Tripos teres (Kofoid) F. Gómez 2013	14	87.5
Dinophysis schuettii Murray & Whitting 1899	13	81.25
Ornithocercus magnificus Stein 1883	13	81.25
Ornithocercus steinii Schütt 1900	13	81.25
Oxytoxum milneri Murray & Whitting 1899	13	81.25
Tripos massiliensis (Gourret) F.Gómez 2013	13	81.25
Tripos pentagonus (Gourret) F.Gómez 2013	13	81.25
Tripos pulchellus f. *tripodioides* (Jørgensen) F. Gómez 2013	13	81.25
Dinophysis cf. *operculoides* (Schütt) Balech 1967	12	75
Tripos azoricus (Cleve) F.Gómez 2013	12	75
Tripos extensus (Gourret) F.Gómez 2013	12	75
Tripos fusus (Ehrenberg) F.Gómez 2013	12	75

In an environment with low nutrient availability, nitrogen fixation is an important feature for ensuring fitness. Hence, the prevalence of the Cyanobacteria *Trichodesmium* can be understood. It is a diazotrophic organism that has been well documented both in the marine waters of Brazil (Satô et al. 1963, Brandini et al. 1997, Carvalho et al. 2008, Proença et al. 2009, Monteiro et al. 2010) and worldwide (Sellner 1997, Janson et al. 1999).

Several micro- and mesophytoplankton organisms accompany cyanobacteria nitrogen fixers. *Richelia intracellularis* J.Schmidt in Ostenfeld & Schmidt is a diazotrophic cyanobacteria found in symbiotic relationship with certain diatoms, such as *Rhizosolenia* Brightwell (Padmakumaret al. 2010), *Hemiaulus* Heiberg (Kimor et al. 1978, Villareal 1994) and *Chaetoceros* Ehrenberg (Gómez et al. 2005). Furthermore, some oceanic dinoflagellates abide in symbiosis with unicellular diazotrophic bacteria, commonly found in certain genera of the non-photosynthetic Dinophysiales order, such as *Histioneis*, *Ornithocercus* Stein and *Amphisolenia* Stein (Foster et al. 2006, Farnelid et al. 2010). This corroborates our results on the high diversity of the above cited genera in the region of the Vitória-Trindade Seamount Chain.

When considering the lack of research on phytoplankton assemblages in Brazilian offshore waters, novel reports on infrageneric taxa can be expected. Other propitious factors could be (*i*) the fluctuation of phytoplankton composition due to environmental changes, such as

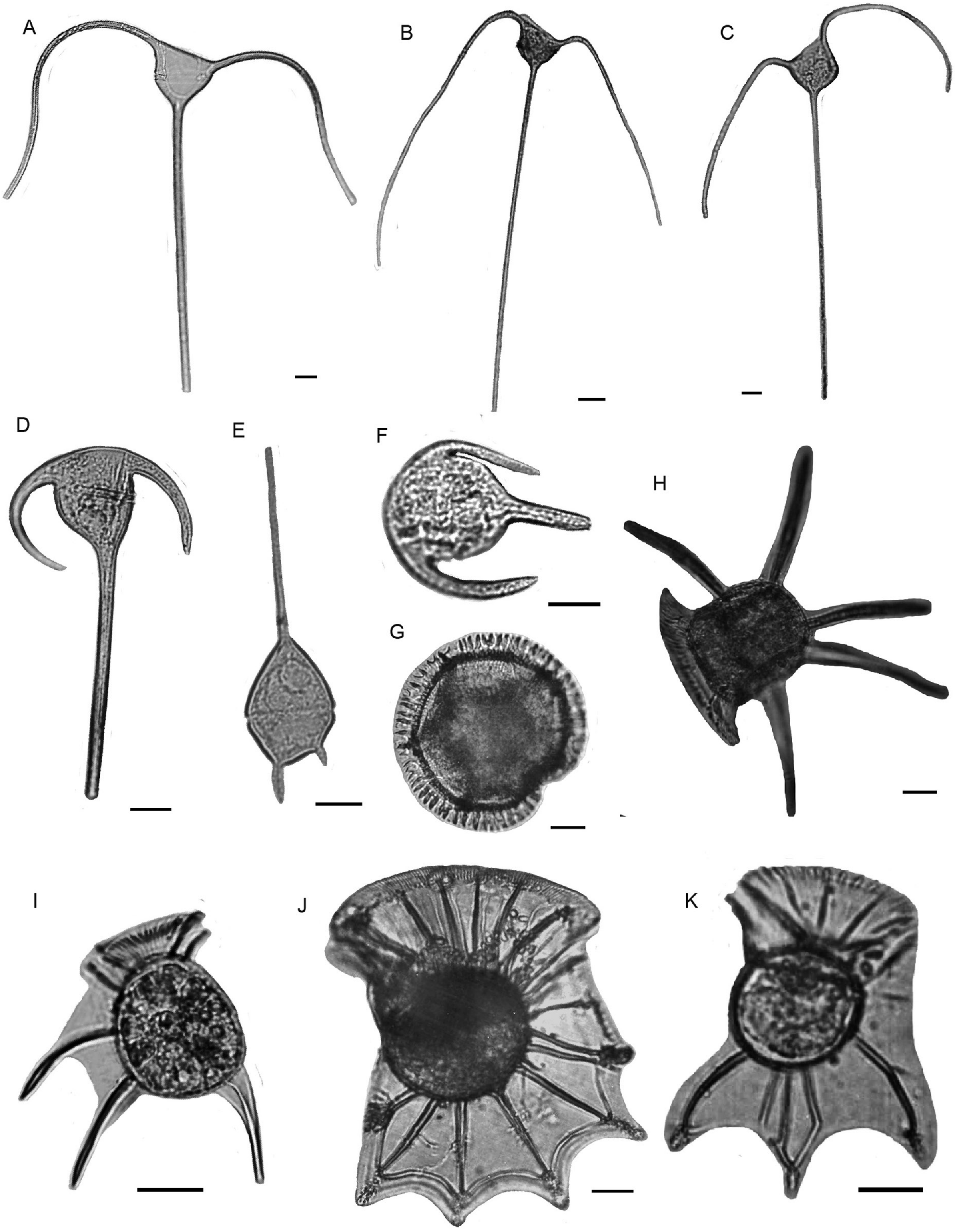

Figure 3. Very common taxa (≥70% of occurrence). Scale bar sizes are between brackets. (A) *Tripos contrarius* (20µm); (B) *Tripos massiliensis* (50 µm); (C) *Tripos macroceros* (20 µm); (D) *Tripos declinatus* (20µm); (E) *Tripos teres* (scale bar20µm); (F) *Tripos azoricus* (20µm); (G) *Triadinium polyedricum* (20µm); (H) *Ceratocorys horrida* (20 µm); (I) *Dinophysis schuettii* (20µm); (J) *Ornithocercus steinii* (20 µm); (K) *Ornithocercus magnificus* (20µm).

Table 3. New infrageneric taxa reports. New records for Brazil. (*) New records for waters of the Espírito Santo continental shelf and near the Vitória-Trindade seamount chain.

Phylum	Species
Bacillariophyta	*Asteromphalus stellatus* (Greville) J.J.Ralfs
Dinophyta	*Amphisolenia schroederi* Kofoid 1907
Dinophyta	*Cladopyxis hemibrachiata* Balech 1964
Dinophyta	*Heterodinium dispar* Kofoid & Adamson 1933
Dinophyta	*Histioneis garrettii* Kofoid & Michener 1907
Dinophyta	*Histioneis joergensenii* Schiller 1928
Dinophyta	*Kofoidinium velleloides* Pavillard 1929
Dinophyta	*Metaphalacroma skogsbergii* L.-S.Tai in L.-S.Tai & Skogsberg 1934
Dinophyta	*Ornithocercus cristatus* Matzenauer 1933
Dinophyta	*Pyrocystis hamulus* var. *semicircularis* Schröder 1900
Dinophyta	*Tripos longinus* (Karsten) F.Gómez 2013
Dinophyta	*Tripos pulchellus* f. *tripodioides* (Jørgensen) F. Gómez 2013
Bacillariophyta	**Asteromphalus flabellatus* (Brébisson) Greville 1859
Bacillariophyta	**Chaetoceros* cf. *concavicornis* Mangin 1917
Bacillariophyta	**Chaetoceros mitra* (Bailey) Cleve 1896
Bacillariophyta	**Rhizosolenia castracanei* H.Peragallo 1888
Bacillariophyta	**Hemiaulus membranaceus* Cleve
Bacillariophyta	**Lampriscus shadboltianum* (Greville) Peragallo & Peragallo 1902
Bacillariophyta	**Thalassiosira leptopus* (Grunow ex Van Heurck) Hasle & G.Fryxell 1977
Bacillariophyta	**Trigonium formosum* (Brightwell) Cleve 1867
Dinophyta	**Amphisolenia bifurcata* Murray & Whitting 1899
Dinophyta	**Amphisolenia schauinslandii* Lemmermann 1899
Dinophyta	**Citharistes regius* Stein 1883
Dinophyta	**Dinophysis caudata* Saville-Kent 1881
Dinophyta	**Dinophysis fortii* Pavillard 1923
Dinophyta	**Dinophysis pusilla* Jørgensen 1923
Dinophyta	**Protoperidinium elegans* (Cleve) Balech 1974
Dinophyta	**Protoperidinium oceanicum* (Vanhöffen) Balech 1974
Dinophyta	**Protoperidinium pentagonum* (Gran) Balech 1974
Dinophyta	**Spiraulax kofoidii* H.W.Graham 1942
Dinophyta	**Tripos cephalotus* (Lemmermann) F.Gómez 2013
Dinophyta	**Tripos macroceros* var. *gallicum* (Kofoid) F.Gómez 2013
Dinophyta	**Tripos massiliense* var. *armatum* (Karsten) F. Gómez
Dinophyta	**Tripos symmetricus* (Pavillard) F.Gómez 2013
Dinophyta	**Tripos vultur* var. *japonicum* (Schröder) F. Gómez 2013

seasonality, (*ii*) misidentification of species, and (*iii*) very low density of some species, thereby precluding their perception in previous studies.

There was little variation in environmental data. In temperature, although the highest range, the values varied between 1.5°C. Considering the variables obtained, the conditions in the study area could be considered stable. Therefore, it was impossible to associate the differences in phytoplankton composition with the data obtained. Maybe, quantifying and qualifying nutrients could give a better indication for the findings.

Cluster analysis indicated how close the stations were in terms of taxonomic composition, and also that geographic distances between the stations were correlated with species composition: the closer the stations to one another, the more species they shared in common. The same association was found in a biogeographic study of *Tripos* (Dodge 1993). Furthermore, the grouping of stations into three different clusters could be justified by the high number of uncommon and rare taxa (101 taxa, 57.5%), thereby indicating that many species are not extensively disposed. It was noteworthy that the three clusters seemed to be correlated to continental closeness and seafloor bathymetry. This is in accordance with previous findings, whereby neritic species assemblage differs from the oceanic (Dodge 1993, Raine et al. 2002, Taylor et al. 2007), and seamounts can influence species composition and density (Genin & Boehlert 1985, Pitcher et al. 2007).

The high micro- and mesophytoplankton species diversity is apparently related to tropical oligotrophic oceanic zones. Several of the organisms identified were either mixotrophic, or maintained a symbiotic association with diazotrophic bacteria. The results are important in defining micro-and mesophytoplankton diversity, especially among the dinoflagellates. The numerous new infrageneric taxa reported for the marine waters off Espírito Santo State, and Brazil as a whole, indicate the importance of inventory surveys, and the lack of studies of phytoplankton assemblages. Bathymetric dissemblance in the area studied could be associated with differences in species composition. Neritic stations formed a distinct cluster from the oceanic and seamount. Furthermore, geographic distances between stations possibly exert an influence on species distribution. Further studies of phytoplankton should be extended to the area, especially those of other phytoplankton size-fractions, such as pico- and nanoplankton. The application of other methodologies, such as cultures, metabarcoding and metagenomics, would be useful for enhance the knowledge about the community.

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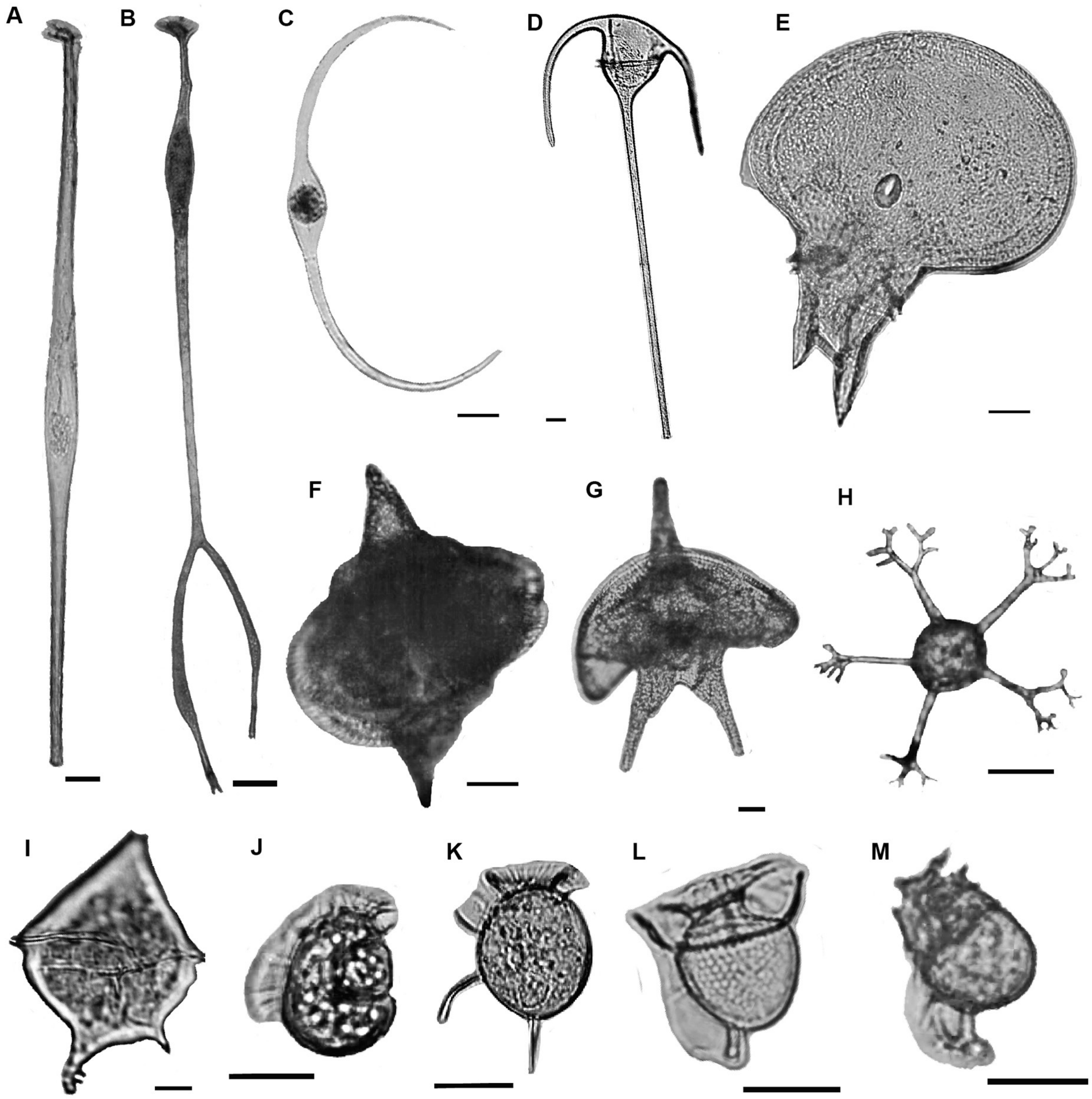

Figure 4. Some of the new records for marine waters of Brazil and Espírito Santo State (*). Scale bar sizes are between brackets. (A) **Amphisolenia schauinslandii* (20 µm); (B) **Amphisolenia bifurcata* (10µm); (C) *Pyrocystis hamulus* var. *semicircularis* (50 µm); (D) *Tripos pulchellus* f. *tripodioides* (20µm); (E) **Tripos cephalotus* (20µm); (F) **Spiraulax kofoidii* (20µm); (G) **Protoperidinium elegans* (20µm); (H) *Cladopyxis hemibrachiata* (20µm); (I) *Heterodinium dispar* (10µm); (J) **Citharistes regius* (20µm); (K) **Dinophysis pusilla* (20µm); (L) *Histioneis garrettii* (20µm); (M) *Histioneis joergensenii* (20µm)

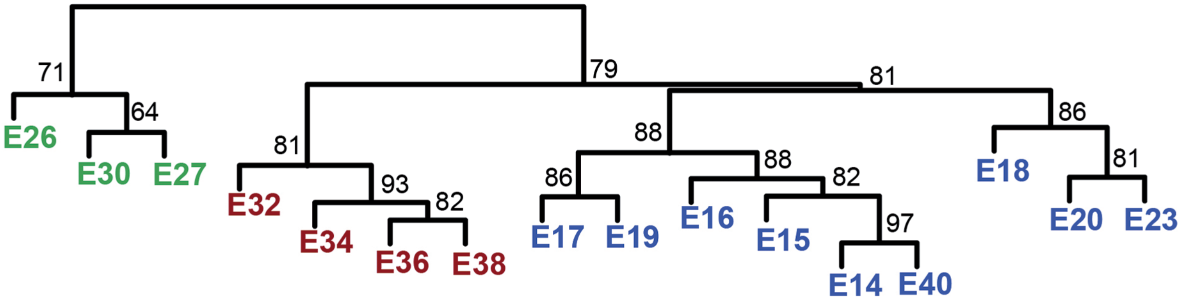

Figure 5. Hierarchical cluster dendrogram representing binary distances between stations according to phytoplankton species composition. Green represents the neritic group, red those closer to the seamounts, and blue those in deep offshore areas.

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article

Bat fauna of the Cerrado savanna of eastern Maranhão, Brazil, with new species occurrences

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Abstract: In Brazil, 179 bat species have been recorded to date, representing 68 genera and nine families. Few data are available on the bat fauna of the state of Maranhão, so the present study was based on a survey of the bat fauna of the Inhamum Municipal Environmental Protection Area (APA Inhamum) in the Cerrado savanna of the eastern extreme of the state. This inventory provides new records of bat species for the state of Maranhão and the Cerrado biome. A total of 31 species (four families) were collected, representing 39% of the bat species known to occur in Maranhão and 28.71% of the species described for the Cerrado. The family Phyllostomidae was the most diverse, with 23 species (74.20% of the total), and also the most abundant, with 86.09% of the specimens collected. The Molossidae (four species) was the second most diverse, followed by the Vespertilionidae (three species) and the Emballonuridae (one species). *Carollia perspicillata* was the most abundant species (28%).The records of *Artibeus fimbriatus* and *Platyrrhinus fusciventris* are the first for the Cerrado and were also recorded in Maranhão for the first time. The cumulative species curve did not reach an asymptote. The species *Dermanura gnoma*, *Lasiurus blossevillii*, *Lasiurus ega*, *Micronycteris schmidtorum*, *Molossops temminckii*, *Platyrrhinus cf. recifinus*, *Phylloderma stenops* and *Trachops cirrhosus*, were also recorded in Maranhão for the first time demonstrating the value of for mammal inventories and emphasizing the need for further surveys in this poorly-known region.
Keywords: *Bats, Faunal survey, APA Inhamum, Maranhão, New records, Chiroptera.*

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Resumo: Atualmente são registrados para o Brasil, nove famílias, 68 gêneros e 179 espécies de morcegos. No Maranhão, poucas são as informações sobre a fauna de morcegos, portanto objetivou-se realizar um levantamento faunístico e reportar os novos registros de morcegos para o Cerrado leste maranhense na Área de Proteção Ambiental Municipal do Inhamum. Foram coletadas 31 espécies, distribuídas em quatro famílias representando 39% das espécies de morcegos registradas no Maranhão e 28,71% das espécies descritas para o Cerrado. A família Phyllostomidae apresentou a maior riqueza (23 espécies - 74,20%) e abundância (86,09%). A família Molossidae (quatro espécies) foi a segunda mais representativa, seguida de Vespertilionidae (três espécies) e Emballonuridae (uma espécie). *Carollia perspicillata* foi a espécie mais abundante (28%). As espécies *Artibeus fimbriatus* e *Platyrrhinus fusciventris* constituem novos registros para o Cerrado e o primeiro registro para o Maranhão. A curva de acumulação de espécies mostrou-se não estabilizada. As espécies *Dermanura gnoma*, *Lasiurus blossevillii*, *Lasiurus ega*, *Micronycteris schmidtorum*, *Molossops temminckii*, *Platyrrhinus cf. recifinus*, *Phylloderma stenops* e *Trachops cirrhosus* constituem novos registros para o Maranhão. As espécies coletadas na APA do Inhamum correspondem a novos registros para o Cerrado do leste maranhense evidenciando-a como uma área de extrema importância para levantamentos mastozoológicos, mostrando que a região necessita de mais estudos.
Palavras-chave: *Morcegos, Levantamento faunístico, APA do Inhamum, Maranhão, Novos registros, Chiroptera.*

Introduction

Bats constitute an important component of the mammal communities of all Neotropical ecosystems in terms of species richness and ecological diversity. In some areas, bats contribute a significant proportion of total diversity, representing up to 40-50% of the total number of species in mammalian communities (Bianconi et al. 2004). In Brazil, the reviewed list includes nine families, 68 genera, and 179 species (Nogueira et al. 2014, Feijó et al. 2015).

The Brazilian state of Maranhão covers a total of 21,656,866 hectares (Neres & Conceição 2010) and is located centrally on the country's northern coast, in a strategic position at the convergence point of the Amazon, Cerrado and Caatinga biomes (Dias et al. 2009),which is reflected in the state's considerable diversity of animals and plants, and makes it potentially important for the inventory of mammals (Oliveira et al. 2008). Despite this richness of ecosystems and biological diversity, few studies are available on the bat fauna of the state, with the exception of the recent efforts of Cruz et al. (2007), Dias et al. (2009), Santos et al. (2009) and Bernard et al. (2011). Most of these studies have focused on sites in the north and west of the state, with few data available from its other regions. In this context, the present study is based on an inventory of the bat fauna of an area of Cerrado in eastern Maranhão, providing new records of the occurrence of species in this region, and other important data on the characteristics of this important group of mammals.

Material and methods

1. *Study area*

The bat specimens were collected during three field excursions to the Inhamum Municipal Environmental Protection Area (APA Inhamum) in April and September 2014, and January 2015. The APA Inhamum (04º53'30"S, 43º24'53"W) is located in the municipality of Caxias, in eastern Maranhão, Brazil (Figure 1), and has a total area of approximately 3,500 ha, which is covered primarily by Cerrado savanna, with two distinct types of vegetation (shrubby-arboreal and grassland) on the flatter terrain, with stands of buriti palms being associated with the watercourses, located in depressions (Neres & Conceição 2010).

2. *Specimen collection and data analysis*

The collection of specimens during this study was authorized by the federal Chico Mendes Institute for Conservation Biology (ICMBio) through special license IBAMA/SISBIO 42670-1. The bats were captured between 18:00 h and 23:00 h using mist-nets of different sizes (12 m x 3 m, 9 m x 3 m, and 6 m x 3 m), which were set in areas adjacent to food sources and probable flight paths, at different locations within the APA Inhamum. All specimens collected have been deposited provisionally in the Genetics and Molecular Biology Laboratory (GENBIMOL) at CESC/UEMA, where they were photographed, identified, sexed, weighed, and measured using a digital caliper (300mm - 12"). The specimens will be deposited in the UFMA vertebrate collection on the Chapadinha campus (Maranhão).

The life stage of each specimen was classified as juvenile or adult based on morphological traits. Adult females were examined through the palpation of the abdomen and teats to determine whether they were lactating or gestating, while the testicles of the males were recorded as abdominal or external. Some of the specimens were anesthetized and sacrificed to provide vouchers and were stored in containers with cotton wool soaked in ether. These specimens were fixed in a standard position with 10% formaldehyde and then conserved in 70% ethanol.

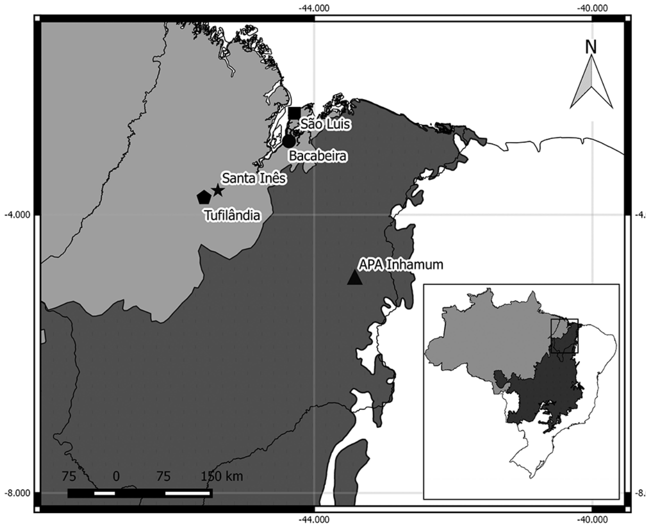

Figure 1. Map of Brazil and the state of Maranhão, showing the study area: the Inhamum Municipal Environmental Protection Area (APA Inhamum). Sites of previous surveys (Amazon biome - Bacabeiras, Santa Inês, Tufilândia and São Luís) and the present study (Cerrado biome - APA Inhamum) of state of Maranhão.

Morphometric data were also collected, including the right (RF) and left forearms (LF), ear (E), tragus (TG), foot (F), and tail (TL). The identification of the taxa was based on the classification keys of Gregorin &Taddei (2002) and Gardner (2008), and subsequently confirmed by specialists. Sampling effort was calculated using the formula proposed by Straube & Bianconi (2002). The cumulative species curve was determined using the Jackknife 1 estimator run in EstimateS, version 9.0.0 (Colwell 2012).

Results

1. *Species diversity*

With a total sampling effort of 24,030 m^2/h, 226 specimens were captured, of 31 species representing four families - Phyllostomidae, Molossidae, Vespertilionidae and Emballonuridae (Figure 2). The family Phyllostomidae was the taxonomically richest, with 23 species (74.20% of the total) distributed in 15 genera, as well as being the most abundant, with 86.09% of the total number of netted individuals (Table 1). The family Molossidae was the second most diverse, being represented by four species in four genera, followed by the Vespertilionidae with three species in two genera, and the Emballonuridae, with a single species.

Carollia perspicillata was the most abundant, representing 28% of the captures. Despite the fact that the species recorded in the APA Inhamum represent 39% of the total number known to occur in Maranhão and 28.71% of the species described for the Cerrado, *Artibeus fimbriatus* and *Platyrrhinus fusciventris* represent new records for this biome and together with *Dermanura gnoma*, *Lasiurus ega, Lasiurus blossevillii, Micronycteris schmidtorum, Molossops temminckii, Phylloderma stenops*, *Platyrrhinus cf. recifinus* and *Trachops cirrhosus* constitute new records for the state of Maranhão. The species curve

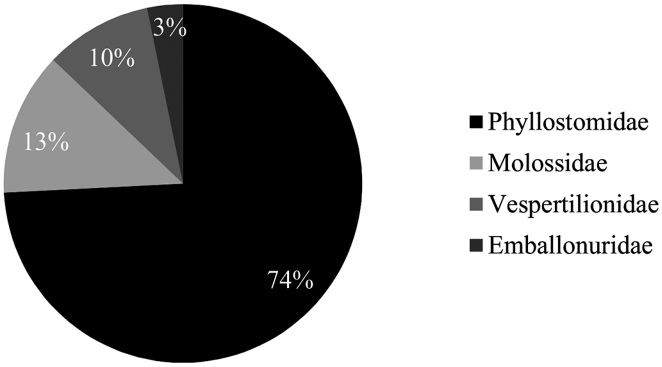

Figure 2. Bat species richness recorded in the APA Inhamum by chiropteran family. The numbers represent the species of families.

Table 1. Bat species recorded in the APA Inhamum, Caxias, Maranhão (Brazil). The species marked with an asterisk were recorded in Maranhão for the first time.

Taxon
Family Emballonuridae
Subfamily Emballonurinae
Rhynchonycteris naso (Wied-Neuwied, 1820)
Família Phyllostomidae
Subfamily Desmodontinae
Desmodus rotundus (E. Geoffroy, 1810)
Diaemus youngii (Jentink, 1893)
Subfamily Glossophaginae
Glossophaga soricina (Pallas, 1766)
Subfamily Phyllostominae
Lophostoma brasiliense Peters, 1866
Lophostoma silvicola d'Orbigny, 1836
Micronycteris minuta (Gervais, 1856)
Micronycteris schmidtorum Sanborn, 1935*
Gardnerycteris crenulatum (E. Geoffroy, 1803)
Phylloderma stenops Peters, 1865*
Phyllostomus discolor Wagner, 1843
Phyllostomus hastatus (Pallas, 1767)
Trachops cirrhosus (Spix, 1823)*
Subfamily Carollinae
Carollia perspicillata (Linnaeus, 1758)
Rhinophylla pumilio Peters, 1865
Subfamily Stenodermatinae
Artibeus fimbriatus Gray, 1838*
Artibeus lituratus (Olfers, 1818)
Artibeus obscurus (Schinz, 1821)
Artibeus planirostris (Spix, 1823)
Dermanura cinerea (Handley, 1987)
Dermanura gnoma Handley, 1987*
Platyrrhinus fusciventris Velazco, Gardner & Patterson 2010*
Platyrrhinus cf. recifinus (Thomas, 1901)*
Sturnira lilium (E. Geoffroy, 1810)
Family Molossidae
Cynomops abrasus (Temmincki, 1827)
Molossops temminckii (Burmeister, 1854)*
Molossus rufus E. Geoffroy, 1805
Nyctinomops laticaudatus (E. Geoffroy, 1805)
Family Vespertilionidae
Subfamily Vespertilioninae
Lasiurus blossevillii ([Lesson, 1826])*
Lasiurus ega (Gervais, 1856)*
Subfamily Myotinae
Myotis nigricans (Schinz, 1821)

had not reached the asymptote by the end of the study (Figure 3). This indicates that further sampling would be required to record the total number of bat species that occur in the study area.

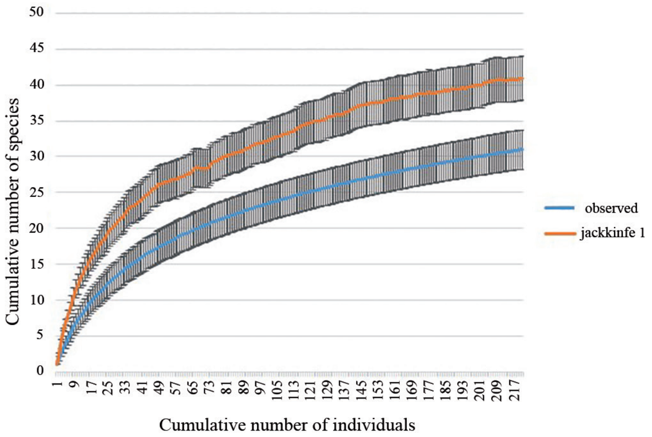

Figure 3. Cumulative species curve for the bats recorded in the APA Inhamum, Caxias, Maranhão, Brazil.

The survey of bats in Maranhão closest to the APA Inhamum was conducted by Dias et al. (2009) in the municipality of Bacabeira (Figure 1). In this case, the results of the present study extend by more than 241 km to the east, the geographic ranges of *Artibeus planirostris*, *A. lituratus*, *A. obscurus*, *C. perspicillata*, *Diaemus youngii*, *Desmodus rotundus*, *Glossophaga soricina*, *Lophostoma silvicola*, *Lophostoma brasiliense, Phyllostomus discolor*, *Sturnira lilium*, *Mimon crenulatum*, *Micronycteris minuta* and *Phyllostomus hastatus*. These same authors also conducted surveys in the municipalities of São Luís, Santa Inês and Tufilândia. The results of the present study also represent an extension of 274 km to eastern Maranhão, of the ranges of *Dermanura cinerea*, *Rhinophylla pumilio*, *Rhynchonycteris naso* and *Myotis nigricans*, previously known to occur only as far east as São Luís (Cruz et al. 2007). The species *Molossus rufus*, *Nyctinomops laticaudatus*, and *Cynomops abrasus* have been recorded previously in Maranhão although the localities have not been documented. Recent studies in the state have all focused on the Amazon biome, and the records from the APA Inhamum represent the first data for the Cerrado biome in Maranhão.

2. *Characterization of the new records of bat species for the Brazilian state of Maranhão*

Family Phyllostomidae

Artibeus fimbriatus

An adult male specimen (body weight 53 g) was captured at 20:50 h on 18 September 2014 and a non-gestating adult female (body weight 59 g) at 22:10 h on 22 January 2015. Forearm length in the male was 69.16-70.98 mm, and 69.07-69.56 mm in the female. The specimens present gray pelage with white-based hairs.

Dermanura gnoma

Two specimens were collected, a male weighing 14 g, at 21:33 h on 10 June 2015, and an adult female weighing 11.5g at 18:25 h on 8 June 2015. Forearm length was 40.19 - 41.16 mm in the male and 40.35-39.83 mm in the female. The dorsal pelage is light chestnut, with the ventral pelage slightly lighter in color, and face with incipient light striping.

Micronycteris schmidtorum

An adult female (non-gestating) weighing 6 g was collected at 20:30 h on 23 January 2015. Right forearm length 32.8 mm, left forearm 32.78 mm. Brown dorsal coloration with ventral coloring in tones of light yellow.

Platyrrhinus fusciventris

Juvenile female weighing 11 g was collected at 19:27 h on 24 January 2015. Right forearm length 37.36 mm, left forearm 39.28 mm. Brown coloration.

Platyrrhinus cf. recifinus

An adult male was captured at 21:24 h on 18 September 2014. Body weight 12.5 g, right forearm 37.32 mm and left forearm 38.22 mm. Coloration predominantly gray, with a continuous white stripe extending from the top of the head to near the uropatagium, as well as yellowish extremities to the ears and tragus.

Phylloderma stenops

An adult male weighing 40.4 g was collected at 18:25 h on 8 June 2015. Right forearm length 70.19 mm, left forearm 69.1 mm. Light brown coloration.

Trachops cirrhosus

An adult male was captured at 18:22 h on 2 May 2014. Body weight 29 g, forearms 62.42-63.22 mm, and tail of 19.52 m. Pelage coloration in varying shades of gray, lips with warts, and ears large.

Family Molossidae

Molossops temminckii

This species was rare, with a single non-gestating adult female being captured at 18:32 h on 1 May 2014. Body weight 4.5 g. Right forearm 30.05 mm, left forearm 29.98 mm. The dorsal pelage ranged from medium to dark chestnut, with the ventral fur light chestnut to cream, with white-based hairs.

Family Vespertilionidae

Lasiurus ega

Four adult males and a juvenile were captured after 19:15 h on 1 May 2014, all with their testicles in an abdominal position. Body weights varied from 9 g to 12 g, and forearm lengths from 43.92 mm to 48.44 mm, and tails of 41.02 mm to 57.19 mm. The specimens were buffy-yellowish in color, with a lighter-colored venter, and a striping pattern on the wings.

Lasiurus blossevillii

Two adult males with abdominal testicles were collected at 18:46 h on 19 September 2014 and a non-gestating female at 19:16 h on 22 January 2015. Body weights varied from 6 g to 7 g for males and 8 g for female. Forearm length varied from 36.64 mm to 40.41 mm, and tails from 30.00 mm to 45.55 mm. Coloration was reddish buffy, with white-tipped hairs, and a lighter-colored venter in yellowish tones.

Discussion

Only 10% of surveys have focused on sites in the northeastern region of the country, which includes Maranhão (Feijó et al. 2011). The results of the present study indicate that the APA Inhamum is characterized by a considerable diversity and abundance of bats, with ten new species recorded for the state and two for the Cerrado biome. In their survey of the bats of the Amazon biome of Maranhão, Bernard et al. (2011) recorded 21 species, of which seven were also found in the present study of the state's Cerrado. In the light of the results of the studies of Olímpio et al. (2015), Nascimento et al. (2015) and Costa et al. (2012) the present study presents evidence of the occurrence in the APA Inhamum of species typical of the state's different biomes, in addition to species endemic to the Cerrado, emphasizing the importance of this site for the understanding of the distribution patterns of the region's chiropterans.

In the opinion of Feijó et al. (2011), inventories such as that reported in the present study are important for the understanding of the diversity and distribution of the fauna of a given region, permitting the resolution of taxonomic questions and the identification of priorities for biogeographic and ecological studies. In addition, Bernard et al. (2011) concluded that this type of study can contribute to the evaluation of environmental impacts and provide important data for decision-making in the management of natural environments.

The inventory of the bat fauna of the APA Inhamum represents 39% of the species known to occur in Maranhão. The phyllostomids were the most diverse of these species, with 74.20% of species richness and 86.09% of total abundance. This predominance of phyllostomids is typical of the chiropteran fauna of Brazil (Bolzan 2011) and may also be accounted for by the collection of specimens using only mist-nets, a selective method (Muylaert et al. 2014).

The genus *Platyrrhinus* is diagnosed from other genera of the subfamily Stenodermatinae by a combination of three characters: two accessory cusps on the posterior face of P4, presence of three upper molars, and presence of a fringe of hair along the trailing margin of the uropatagium. Although other genera also have these characters, no other genera possess all three at the same time (Carvalho. & Fabián, 2011). According to Gardner (2007) little is known about *P. recifinus* and Jones and Carter (1976) suggested that *P. recifinus* may prove to be a junior synonym of *P. lineatus*. The *status* specific of the specimen remains "*Platyrrhinus cf. recifinus*".

The Cerrado biome covers almost a quarter (23%) of Brazil and is characterized by rich, but still poorly-understood biological diversity, which includes 101 bat species, corresponding to 60% of the total number known to occur in the country. These species belong to 42 genera, distributed in eight families (except Natalidae), of which the Phyllostomidae is the most diverse (55 species), followed by the Molossidae (20) and Vespertilionidae with 12 (Sousa et al. 2013). A similar pattern of diversity was recorded in the present study, given that the predominant family was the Phyllostomidae, with 23 species, followed by the Molossidae (four species) and Vespertilionidae, with three species. With the exception of the Emballonuridae (one species), none of the other families known to occur in the Cerrado were recorded in the APA Inhamum. *Artibeus fimbriatus* and *P. fusciventris* represent new records for the Cerrado.

Despite these new records, the number of species captured, and the overall sampling effort, the cumulative species curve did not stabilize, indicating the need for further sampling efforts based on additional, complementary techniques. The results of the present study were consistent with those of Cunha et al. (2011), Ávilla-Cabadilla et al. (2014), and Muylaert et al. (2014) in the Cerrado, in which the cumulative species curves also failed to reach an asymptote. This may have been related to the exclusive use of mist-nets for the collection of specimens, given that this technique favors the capture of phyllostomids, to the detriment of species of other families (Kunz & Parsons 2009), even though specimens of both vespertilionids and molossids were collected in the present study, despite the fact that they tend to fly at very high levels and are not usually collected in mist-nets (Muylaert et al. 2014).

The bat fauna of the Brazilian state of Maranhão is still poorly-known, and the species *D. gnoma*, *L. ega, L. blossevillii, M. schmidtorum, M. temminckii, P. stenops*, *Platyrrhinus cf. recifinus, P. fusciventris, T. cirrhosus, A. fimbriatus* and *P. fusciventris* were all recorded in the state for the first time. The records of *A. fimbriatus* and *P. fusciventris* also represent new occurrences for the Cerrado biome. Range extensions were also recorded for a number of species. In addition to the intrinsic value of these findings, the data represent a valuable resource for the planning of conservation measures, and emphasize the need for additional surveys in the Cerrado of Maranhão, given that the biological diversity of this system is still virtually unknown.

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Biology and description of immature stages of *Gymnetis rufilateris* (Illiger, 1800) (Coleoptera: Cetoniidae: Cetoniinae)

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Abstract: Larvae, pupae and adults of *Gymnetis rufilateris* (Illiger, 1800) (Coleoptera: Cetoniidae: Cetoniinae) were collected in the municipality of Rochedo, Mato Grosso do Sul State. Studies on biology and morphology were conducted at the Laboratory of Entomology of the Universidade Estadual de Mato Grosso do Sul in Aquidauana, Mato Grosso do Sul, Brazil. Females of couples formed in the laboratory oviposited and allowed the biology studies. The eggs lasted for 15.5 days. Larvae of the 1st, 2nd and 3rd instars lasted 13.5, 30.6 and 113.1 days, respectively. The pupal period lasted 12.8 days. The egg-to-adult period lasts 186.8 days. The larvae fed on decomposing feces of poultry. In the laboratory, adults fed on ripe banana (*Musa sp.*) (Musaceae). Descriptions of the 3rd larval instar and pupa of *G. rufilateris* are presented. A key to known larvae of *Gymnetis* is also included.

Keywords: *Biology, behavior, Gymnetini, immature stages, morphology.*

RODRIGUES, S.R., GARCIA, F.P., FALCO, J.S., MORÓN, M.A. **Biologia e descrição dos estágios imaturos de *Gymnetis rufilateris* (Illiger, 1800) (Coleoptera: Cetoniidae: Cetoniinae)**. Biota Neotropica. 16(3): e20140176. http://dx.doi.org/10.1590/1676-0611-BN-2014-0176

Resumo: Larvas, pupas e adultos de *Gymnetis rufilateris* (Illiger, 1800) (Coleoptera: Cetoniidae: Cetoniinae) foram coletados em Rochedo, MS. Estudos sobre a biologia e morfologia foram conduzidos no Laboratório de Entomologia da Universidade Estadual de Mato Grosso do Sul em Aquidauana, MS. Fêmeas de casais formados em laboratório ovipositaram e foram iniciados os estudos de biologia. O período embrionário dura 15,5 dias. As larvas de primeiro, segundo e terceiro instares duram 13,5, 30,6 e 113,1 dias, respectivamente. O período pupal dura 12,8 dias. O período de ovo a adulto completa-se em 186,8 dias. As larvas se alimentam de fezes de aves em decomposição. Em laboratório, os adultos foram alimentados com pedaços de banana (*Musa sp.*) (Musaceae) madura. As descrições da larva de terceiro instar e pupa de *G. rufilateris* são apresentadas. Uma chave para larvas de *Gymnetis* conhecidas também está incluída.

Palavras-chave: *Biologia, comportamento, Gymnetini, estágios imaturos, morfologia.*

Introduction

For *Gymnetis* MacLeay, 1819 (Coleoptera: Cetoniidae: Cetoniinae), 26 species are known, occurring from the United States to Argentina (Morón & Arce 2002). The coloration of adults is variable, including intraspecific variation, as observed by Morón (1995), Antonie (2001), Solís (2004), Orozco & Pardo-Locarno (2004) and Di Iorio (2013). According to Micó et al. (2008), Morón (2010) and Cherman & Morón (2014), Cetoniidae gather enough characteristics to be considered as a family different from Scarabaeidae and Melolonthidae. The *Gymnetis* is comprised by rougly 30 species (Krikken 1984). For the genera of Gymnetini, Antoine (2001) proposed accurately the use of the old nomenclature and considered *Paragymnetis* Schürhoff, 1937 and *Gymnetosoma* Martínez, 1949 as *Gymnetis* MacLeay, 1819.

In Colombia, Orozco & Parco-Locarno (2004) reared *Gymnetis holosericea* (Voet, 1779) in laboratory and observed that the egg-to-adult period is completed in seven months and adult insects showed mutilation behavior and for *G. pantherina* Blanchard, 1837, the larvae show cannibalism. In studies conducted in Colombia, Neita et al. (2006) collected adults of *G. coturnix* (Burmeister, 1842) with fruit traps or on flowers of *Annona muricata* L. (Annonaceae), and larvae are found in decaying logs of *Brosimum utile* (Kunth) Oken (Moraceae). Morón & Arce (2002) collected adults of *G. hebraica difficilis* Burmeister, 1842 and *G. flavomarginata sallei* Schaum, 1849 (= *G. sallei*) in rotten banana (*Musa* sp.) (Musaceae) in a forest in Mexico.

Regarding the morphology of *Gymnetis* immatures, there are descriptions of the 3rd larval instar and pupa of *G. flavomarginata sallei*

Schaum, 1849 (Richter 1966, Morón & Arce 2002), 3rd larval instar of *G. hebraica difficilis* Burmeister, 1842 (Morón & Arce 2002), 3rd larval instar and pupa of *G.* (*Paragymnetis*) *chalcipes* (Gory & Percheron, 1833) (Morelli 2000) and 3rd larval instar and pupa of *G. holosericea* (Voet, 1779) and *G. pantherina* Blanchard, 1837 (Orozco & Parco-Locarno 2004).

In Brazil, works carried out on *Gymnetis* are restricted to studies on the occurrence and diversity of the species (Morón 2004, Ratcliffe 2004, Gonçalves & Louzada 2005, Rodrigues et al. 2013, Puker et al. 2014). In this work, we present for the first time information on the biology and descriptions of 3rd instar larva and pupa of *G. rufilateris* (Illiger, 1800).

Material and Methods

We collected adults, pupae and larvae of *G. rufilateris* between 22-25 December 2012 and 15-20 March 2013 at Bela Vista farm, in the municipality of Rochedo, Mato Grosso do Sul, in a poultry shed (4 x 5 m) in decomposing feces. The samples were sent for identification at the Laboratory of Entomology at the Universidade Estadual de Mato Grosso do Sul (UEMS) in Aquidauana.

We collected 104 larvae of *G. rufilateris* (four of 1st instar, 24 of 2nd instar and 76 of 3rd instar). In the laboratory, the larvae were kept in plastic containers of 500 mL with 2/3 of their volume completed with organic material collected at the site where the larvae were found.

We collected 47 pupae and seven adults from the in plastic containers that was closed with voile fabric. The couples were kept in plastic containers of 4,000 mL, which was covered with voile fabric to prevent them from escaping. Inside the container, we added approximately 2,000 mL of organic material collected from the poultry shed, and ripe banana to feed the adults.

In the containers with adults, we carried out daily observations of mating behavior. Each two days, we inspected and screened material for egg extraction. We collected 200 eggs, which were put in Petri dishes containing a layer of soil, and subsequently placed in a temperature-controlled chamber, which remained with photophase of 12 hours and temperature of 26 ± 2° C. We measured egg width and length with a caliper and the weight on an analytical scale.

After hatching, the larvae were kept in 250 mL containers with organic material and feces of poultry. The larvae remained in the containers until emergence of adults. For larvae and pupae, we performed measurements with a caliper, obtaining information of body length, thorax width and cephalic capsule width. The weight was verified on a digital analytical scale. To monitor the development and duration of larval stage, 50 newly hatched larvae were kept in containers properly numbered 1-50 and we measured the width of the cephalic capsule every three days.

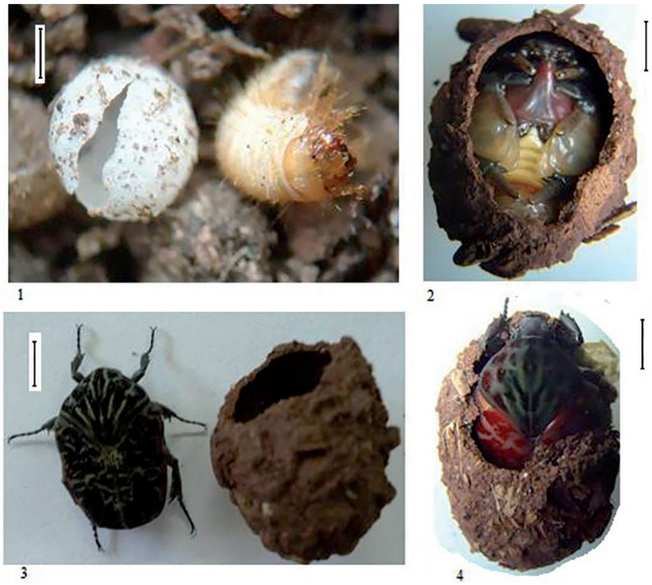
Some adults were mounted with an entomological pin and, through comparisons with insects preserved in the entomological pinned collection of the Universidade Estadual de Mato Grosso do Sul, we obtained their identification.

Ten laboratory-reared larvae were killed in boiling water and preserved in alcohol for description. Larvae and body parts were drawn on stereomicroscope coupled with a camera lucida. Mouthparts were mounted on slides in Hoyer liquid. Descriptions of the larvae were obtained from observations of characters, following the terminology used by Costa et al. (1988) and Morón & Arce (2002). We measured and preserved in alcohol 10 larvae and nine pupae laboratory-reared by Felix P. Garcia on April 24, 2013. The specimens are deposited at UEMS and Museu de Zoologia, Universidade de São Paulo, São Paulo, Brazil (MZSP).

Results

Biology. In the poultry shed, *G. rufilateris* was usually found up to 50 cm from the sides of the shed walls at 10-20 cm deep. The organic material where the insects developed was humid and easy to handle, unlike the organic material inside the shed, which was dry and compact. Several larvae, pupae and adults were found in this substrate, configuring as suitable food for larval development of *G. rufilateris*.

Newly-laid eggs had average sizes of 1.0 x 0.8 mm and near hatching time, average sizes reached 2.5 x 3.0 mm (n = 50). The embryonic period lasted on average 15.5 days and 1st instar larvae, remained in that stadium for 13.5 days, with cephalic capsule width of 1.35 mm (Table 1 and Figure 1). The larvae had 6.3 mm of body length, 1.94 mm thorax width and 13.34 mg weight (Table 2). Larvae in the 2nd instar remained in this stadium for 30.6 days (Table 1), with 2.06 mm cephalic capsule width. Body length was 16.6 mm, thorax width 4.48 mm and weight 257.05 mg (Table 2). Larvae in the 3rd instar remained in that stadium for 113.1 days (Table 1), with 3.6 mm of cephalic capsule width. They were 29.7 mm long, 6.7 mm wide and weighed 1,352.34 mg (Table 2).



Figures 1-4. *Gymnetis rufilateris* (Illiger, 1800). **1)** Chorion and first instar larva. **2)** Pupa inside pupal chamber, **3)** Adult and pupal chamber, **4)** Newly emerged adult inside pupal chamber. Scale bars: Fig. 1, 1 mm; 2-4, 5 mm.

At the end of the third instar, the larvae acquire a whitish color and construct a pupal chamber, where they remained. The pupal chamber was constructed with organic material, soil, own excrement and probably saliva. The chambers had an average size of 23.84 mm long and 17.48 mm wide (Figures 2-4).

Table 1. Duration (mean ± SE) of developmental stages of *Gymnetis rufilateris* (laboratory conditions, 26 ± 1°C, 12-h photoperiod).

Stage	Duration (days)	Interval of variation (days)	N	Viability (%)
Egg	15.5 ± 0.04	15 - 16	200	100
1st instar	13.5 ± 0.16	12 - 17	200	100
2nd instar	30.6 ± 0.20	30 - 35	180	90
3rd instar	113.1 ± 3.91	102 - 138	142	77.8
Pupae	12.8 ± 0.55	10 - 16	138	97.2
Adult	38.5 ± 0.25	32 - 45	138	100
Egg-to-adult	186.8 ± 1.25	102 - 214	138	69

Table 2. Mean value (± SE) for length, width, and weight of developmental stages of *Gymnetis rufilateris* (in laboratory, 26 ± 1°C, 12-h photoperiod).

Stage	Length (mm)		Width (mm)		Weight (mg)	
	Mean ± SE	Interval	Mean ± SE	Interval	Mean ± SE	Interval
Egg	1.0 ± 0.02	0.9 - 1.1	0.8 ± 0.02	0.7 - 1.0	-	-
1st instar	6.3 ± 0.24	4.5 - 12.0	1.9 ± 0.01	1.4 - 3.8	13.34 ± 0.01	6.1 - 80.0
2nd instar	16.6 ± 0.85	9.9 - 22.4	4.5 ± 0.21	2.6 - 6.0	257.06 ± 32.83	60 - 544
3rd instar	29.7 ± 0.58	19.3 - 34.3	6.7 ± 0.15	5.3 - 8.3	1,352.34 ± 71.11	380 - 2,153
Pupa	23.8 ± 0.26	18.5 - 27.7	17.5 ± 0.25	12.8 - 20.6	697.55 ± 77.99	436 - 1,211

The pupae remained in this stage for 12.8 days on average with 23.8 mm long and 17.5 mm wide (Table 1 and Figure 2). The egg-to-adult period of *G. rufilateris* was completed in 186.8 (102 - 214) days on average.

After emergence adults have predominantly red color (Figure 4). After sclerotized, adults head were predominantly slightly green with shades of black. On the pronotum and elytra, dark-green color predominated with black streaks and red color predominated in spots on the edges (Figure 3). Adults of *G. rufilateris* showed average longevity of 38.5 days (n = 50).

Copulation in the laboratory was observed with a duration 150 seconds (n = 1). Four other copulation attempts were observed in the laboratory. Copulation and copulation attempts occurred between 07:30 and 09:00 a.m. Copulation showed the following behavior: the male meets the female and touches this with antennae and tarsi of the first pair of legs. Then the male climbs onto the female, positions the final portion of the abdomen lined with female pygidium, exposes the aedeagus and inserts it into the female genital chamber. After copulation, the male climbs off the female and both separate.

In copulation attempts, the male climbs onto the female, exposes the aedeagus and initiates rapid movement with the abdomen, where the exposed aedeagus touches quickly on the female pygidium, in an attempt to initiate copulation. This behavior lasts 4 minutes on average and, immediately ceases. Next, the male climbs off the female, retracts the aedeagus, walks near the female, climbs onto the female again and repeats the behavior for 4 minute again (n = 4). If the female does not accept the male for copulation, the male climbs off the female, retracts the aedeagus and both separate. During the 4 minute attempts of copulation, the male show intense rhythmic activity back and forth with the aedeagus exposed.

Ripe bananas were offered to adults, which remained feeding for 2-3 hours. According to information from Mr. Nilson Falco, owner of Bela Vista Farm, the 2nd and 3rd instar larvae of *G. rufilateris* are used as excellent baits for deep-sea fishing of piraputanga (*Brycon microlepis* Perugia, 1897) (Characidae).

Gymnetis rufilateris (Illiger, 1800)
3rd instar larva

Description. Dorsal length 29.7 (20.3 - 35.4) mm on average, prothoracic width 6.7 mm, head capsule width 3.6 mm (Figure 5). **Head** (Figure 6): Hypognathous, strongly sclerotized, light brown. The epicranial arms is slightly sinuous in its initial third. Setae with the following distribution: five dorsoepicranial pairs adjacent to coronal suture, two pairs near the antennifer, one pair in the postero-frontal region, and one pair in the anterior frontal angle. Stemmata absent. Well-sclerotized antennifer about half of length of pedicel. Antennae (Figures 5, 6, 13) long, 4 articles, well-sclerotized, articles I-III enlarging to apex, short sensorial cone at apex on article III, article IV (Figure 13) with two dorsal and three ventral sensorial spots. Clypeus (Figure 6) trapezoidal, with one posterior clypeal setae and two exterior clypeal setae. Labrum (Figure 6) anterior border trilobate, 5-6 anterior marginal setae on each side, posterior-lateral margin with three setae per side, basal region with two groups of four short setae.

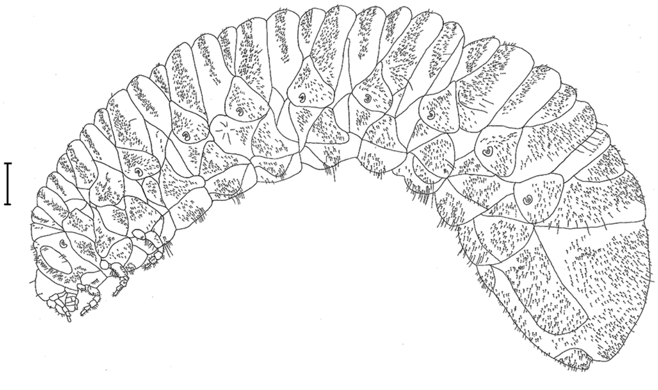
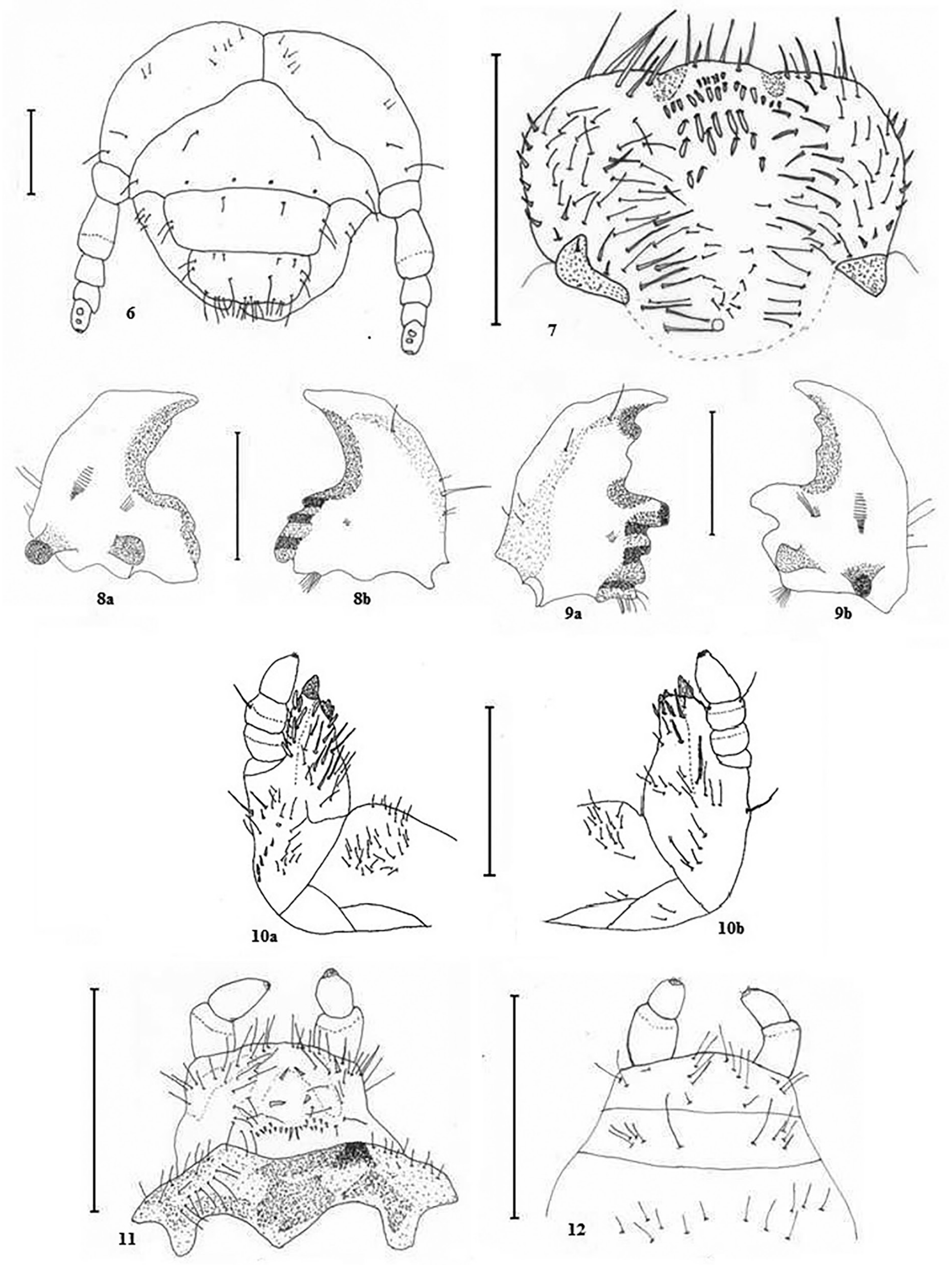
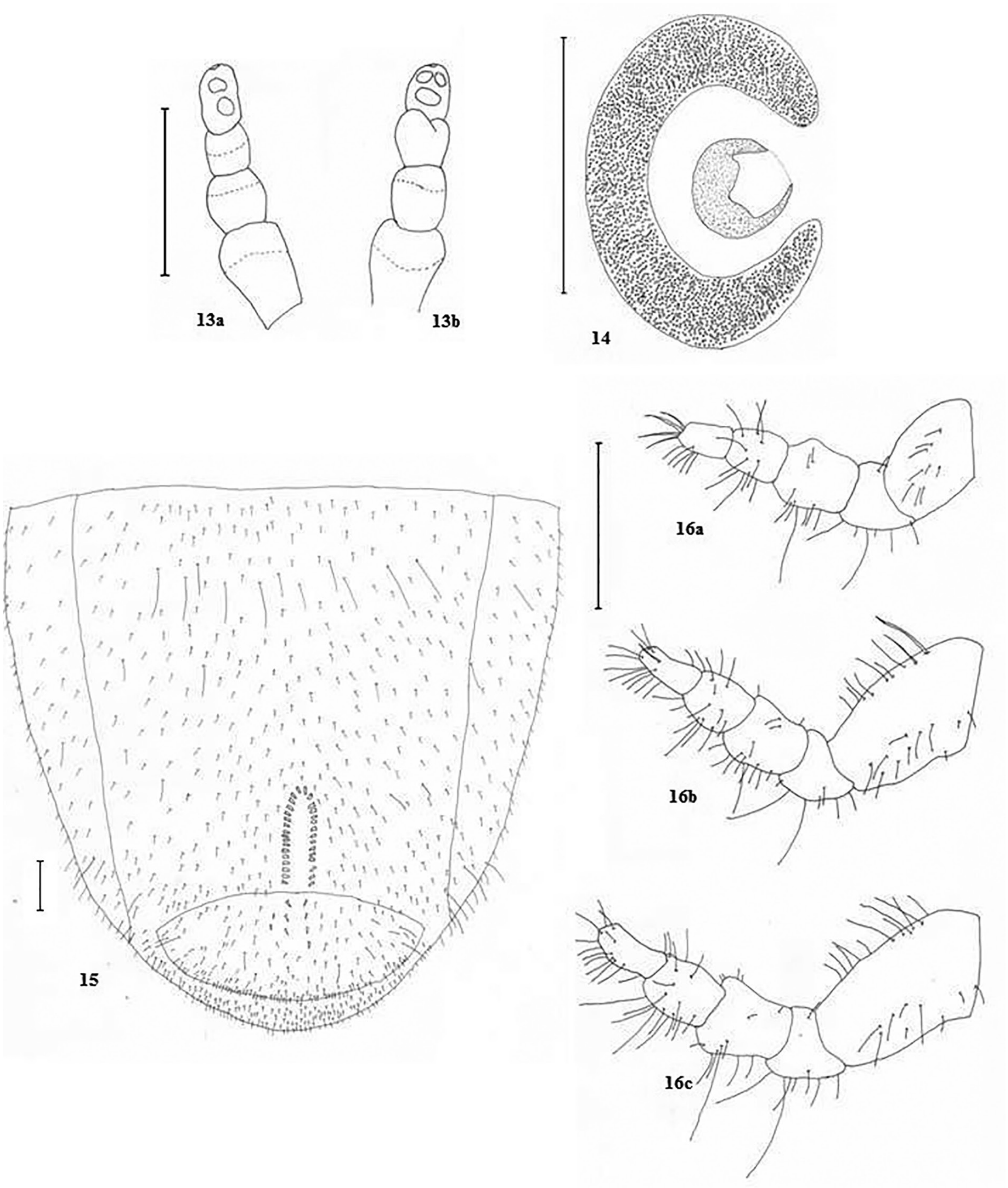


Figure 5. *Gymnetis rufilateris* (Illiger, 1800), third instar (lateral view). Scale bar: 2 mm.



Figures 6-12. *Gymnetis rufilateris* (Illiger, 1800), third instar larva. **6)** Head, frontal view; **7)** Epipharynx; **8)** Right mandible, a) ventral view, b) dorsal view; **9)** Left mandible, a) dorsal view, b) ventral view; **10)** Left maxilla, a) dorsal view, b) ventral view; **11)** Hypopharynx; **12)** Labium. Scale bars: 1 mm.



Figures 13-16. *Gymnetis rufilateris* (Illiger, 1800), third instar larva. **13)** Antennal apex, a) dorsal view, b) ventral view; **14)** Thoracic spiracle; **15)** Urosternite X; **16)** Left legs, a) prothoracic, b) mesothoracic, c) metathoracic. Scale bars: Fig. 14, 0.5 mm; Figs. 13, 15-16, 1 mm.

Epipharynx (Figure 7): Clithra present. Corypha with 6 setae. Haptomeral process present, with distal projection and 14 heli. Above heli 6 small setae. Behind the heli, 12 long setae irregularly distributed. Acroparia with 6 long setae on left side, 7 on right side. Acanthoparia with 6 setae on left and on right. Chaetoparia with short spiniform setae, 51 setae on left side, 43 on right side. Dexiotorma transverse, short sclerotized plate. Laeotorma present. Sensorial cone present.

Right mandible (Figure 8ab) with scissorial teeth S1 and S2 fused, separated from S3 by a furrow. Dorsal molar area with four setae. Dorsal surface with a large setae below base of S3 scissorial. Scrobe with four setae. Brustia with nine setae. Ventral region with well-marked stridulatory area consisting of 17 subparallel ridges. Well-developed ventral process. Four setae in molar region, with two basolateral setae.

Left mandible (Figure 9ab) with tooth S1 separated from S2 scissorial tooth by incised furrow. Base of scissorial teeth S2 with a long setae. Scrobe with three setae. Dorsomolar region with about five setae. Brustia with 10 setae. Ventral surface with stridulatory area with 17 well-defined ridges. Ventral process present. Well-developed molar lobe. Molar region with 6-7 setae.

Maxillae (Figure 10ab) symmetrical. Cardo long, with a few setae and stipes with various setae. Galea and lacinia fused, forming mala. Mala with three unci, one apical and two pre-apical. Palpifer slightly shorter than palpomere I. Palp 3-segmented, segment II with a setae. Maxilla with rows containing five acute denticles and an intricate distal process.

Hypopharynx (Figure 11). Sclerome strongly sclerotized, asymmetric, with right side more prominent. Left lobe with18 setae, right lobe with 10 short setae. Posterior left margin with 18 long, slender setae and posterior right margin with 17 long, slender setae. Glossa with a transverse row of small and short setae.

Labium (Fig. 12). Submentum with various setae. Mentum with 5 pairs of setae. Prementum with 5 pairs of setae on median region and 3 pairs of seta on lateral region. Labial palpi 2-segmented, small sensory hairs on apex.

Thorax: Thoracic segments with several dorsal and lateral setae disposed in rows. Pronotum with two lobes and a brown sclerotized plate on each side with three setae (Figure 5). Thoracic spiracle 0.54 mm long, 0.47 mm wide (Figure 14). Respiratory plate with about 19 holes across the middle section. Respiratory orifice with irregular shaped. Meso and metanotum each with three lobes, respectively. Legs. Coxa, trochanter, femur, tibia and tarsungulus of all legs with numerous, long, stout setae. Anterior and middle legs shorter than hind legs (Figure 16abc). Tarsungulus cylindrical rounded apically with 9-13 setae.

Abdomen: Ten-segmented. I-VII each with 3 dorsal lobes. Segments IX and X united. Spiracles I, II and IV 0.55 mm long, 0.41 mm wide; III and V 0.51 mm long, 0.42 mm wide; VI 0.60 mm long, 0.50 mm wide; VII 0.58 mm long, 0.43 mm wide; and VIII 0.57 long, 0.40 mm wide. Raster with one pair of palidia and with 15-17 setae in each pali. Palidia opened posteriorly and closed anteriorly. Lower anal lip with about 168 small, thick setae (Figure 15).

Pupa (Figure 17ab)

Length: 23.8 (18.5 - 27.7) mm, maximal width: 11 mm. Body elongated, exarate and light yellow. Completely glabrous. Antennae and mouthparts clearly separated. Ocular canthus, antennae, and compound well-differentiated eyes. Clypeus concavo. Pronotum trapezoidal. Meso- and metasternum with wide process with apex emerging between pro- and mesocoxa. Pterothecae free, compressed around the body and extended posteriorly to fifth abdominal segment. Pterothecae I superposed to pterothecae II. Prior margin of pterothecae I subposed by medial leg. Pterothecae II covering proximal half of posterior leg. Spurs of meso and metatibiae visible. All tarsomeres little defined on apex. Tergites convex. Seven spiracles visible dorsally. Elongated spiracle I, not prominent, partially protected. Spiracles II - IV tuberculiform, sclerotized peritreme. Spiracles V-VII closed, tuberculiform, prominent. Urogomphi absent. Female ventrite VIII with medial genital pore, male ventrite VIII with genital ampulla.

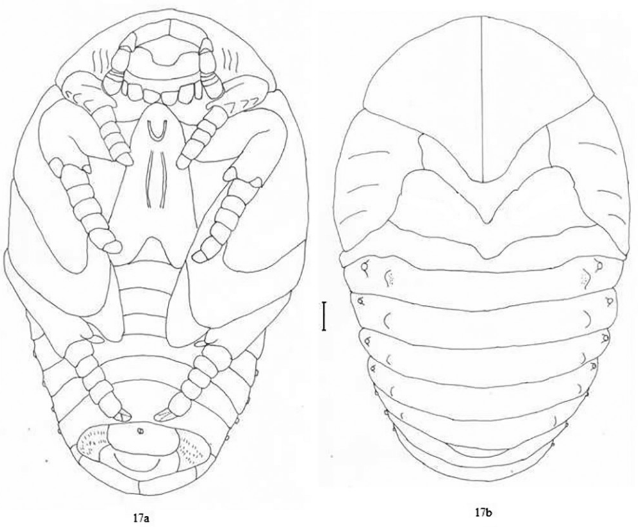


Figure 17. *Gymnetis rufilateris* (Illiger, 1800), pupa, a) ventral view, b) dorsal view. Scale bar: 1 mm.

Material Examined. BRAZIL. Mato Grosso do Sul: Rochedo (Bela Vista Farm), on poultry feces, 22.XII.2012 - 20.III.2013, F. P. Garcia, 10 larvae, 9 pupae (measured) (UEMS), 3 larvae (3rd instar), 1 pupa, 2 adults (MZSP).

Key to the Known Third Larval Instar of *Gymnetis* MacLeay, 1819

(Adapted from Orozco & Pardo-Locarno 2004)

1. Surface of last antennomere with four dorsal sensory spots. Each palidium consisting of a row of 13-14 pali. Tarsungulus bearing six setae..................***G.* (*Paragymnetis*) *chalcipes*** (Gory & Percheron)

1´. Surface of last antennomere with 2-3 dorsal sensory spots. Each palidium consisting of a row of 10-26 pali. Tarsungulus with 8-13 setae..2

2. Tentorial pits absent. Spiracles of abdominal segments slightly increasing in size toward posterior segments...***G. difficilis*** Burmeister (cited as ***G. hebraica difficilis*** Burmeister by Morón & Arce (2002)

2´. Tentorial pits evident. Spiracles of abdominal segments similar in size...3

3. Dorsoepicranium with central row of 9-13 setae. Surface of last antennomere with 3-5 ventral sensory spots. Each palidium with a row of 18-26 pali....................................***G. pantherina*** Blanchard

3´. Dorsoepicranium with central row of 5-7 setae. Surface of last antennomere with 2-3 ventral sensory spots. Each palidium with 10-17 pali..4

4. First antennomere slightly shorter than the two following segments together. Palidia joined anteriorly..................................
....***G. chevrolati sallei*** Schaum (cited as ***G. flavomarginata sallei*** Schaum by Morón & Arce 2002)
4´. First antennomere longer than the two following segments together. Palidia open or joined anteriorly.....................................5
5. Palidia open anteriorly.................................***G. holosericea*** Voet
5'. Palidia joined anteriorly...........................***G. rufilateris*** (Illiger)

Discussion

Gymnetis rufilateris completed the cycle of 186.8 days (about 6 months), and showed no cannibalism behavior in the laboratory. Its life cycle is seven months shorter than that of *G. holosericae*, which also presented cannibalism behavior according Orozco & Locarno Parco (2004). Immature of *G. rufilateris* develop in decaying poultry droppings. These observations agree with Arce & Morón (1999), who reported that *Gymnetis* larvae develop in organic material.

The third instar larvae of *G. rufilateris* differ from those of other species of *Gymnetis* by having five dorsoepicranial pairs setae adjacent to coronal suture, antennomere IV with 2 dorsal and three ventral sensorial spots, spiracles of abdominal segments similar in size and palidium with a row of 15-17 pali opened posteriorly.

Acknowledgments

To Mr. Nilson Falco, owner of Bela Vista Farm, for allowing collection of *G. rufilateris*.

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article

Structure and composition of fish assemblages from São Sebastião Channel tide pools, southwestern Atlantic

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PASTRO, G., DIAS, G.M., GIBRAN, F.Z. **Structure and composition of fish assemblages from São Sebastião Channel tide pools, southwestern Atlantic**. Biota Neotropica. 16(3): e20150128. http://dx.doi.org/10.1590/1676-0611-BN-2015-0128

Abstract: Organisms living in tide pools in the intertidal zone are exposed to daily and abrupt changes in temperature, dissolved oxygen, pH and salinity, during tidal cycles. Fish assemblages in tide pools are usually very different from those in surrounding areas; resident species exhibit morphological and ecological adaptations to deal with such a variable environment. In this study, we described the environmental conditions and the fish diversity and composition from four tide pools at the continental margin of the São Sebastião Channel, São Sebastião (23º41'-23º54'S; 45º19'-45º30'W), in southeastern Brazil. Monthly, from January to December 2011, we sampled four tide pools (12 samples per pool), applying a standard protocol to record environmental variables, and visual censuses to record abundance, richness and composition of fish assemblages. Environmental and fish data were compared among tide pools and periods throughout the year (warm and cold) using both univariate and multivariate analyses of variance. Tide pools showed significant environmental differences, mainly in area, volume and complexity of substrata. We observed 13 species of fishes (10 families), which belong to five trophic guilds (roving herbivores, territorial herbivores, mobile invertebrate feeders, omnivores and carnivores). Density and richness of fish were almost constant during the studied period, except for a reduction in fish density in São Francisco's Beach during a cold period. However, assemblages from the sampled tide pools differed from each other in composition and relative abundance of species, resulting in four distinct assemblage structures. *Bathygobius soporator* was the dominant species in most of the tide pools, corresponding to 66% of all recorded fish. The tide pool from Baleeiro's Point was intermediate in size, but was covered by a complex algae community and supported the most diverse fish assemblage, suggesting that habitat complexity may contribute to the diversity of fish.
Keywords: *Subtropical fish, intertidal zone, environmental factors, species richness, Southeastern Brazil.*

PASTRO, G., DIAS, G.M., GIBRAN, F.Z. **Estrutura e composição das assembleias de peixes de poças de marés do Canal de São Sebastião, Atlântico sudoeste**. Biota Neotropica. 16(3): e20150128. http://dx.doi.org/10.1590/1676-0611-BN-2015-0128

Resumo: Organismos que habitam poças na zona entre marés estão sujeitos a mudanças diárias e abruptas na temperatura, oxigênio dissolvido, pH e salinidade durante os ciclos das marés. Por isso, as assembleias de peixes dessas poças são geralmente bem distintas daquelas das áreas adjacentes, do infralitoral, apresentando espécies residentes com adaptações morfológicas e ecológicas para lidar com tal ambiente variável. Neste estudo, descrevemos e comparamos as condições ambientais e a diversidade e composição da ictiofauna de quatro poças de marés verdadeiras na margem continental do Canal de São Sebastião, São Sebastião (23º41'-23º54'S; 45º19'-45º30'W), sudeste do Brasil. Mensalmente, de janeiro a dezembro de 2011, amostramos quatro poças (12 amostras por poça), aplicando um protocolo padrão para registrar as variáveis ambientais, e censos visuais para registrar a abundância, riqueza e composição da assembleia de peixes. Dados abióticos e da ictiofauna foram comparados entre poças de maré e períodos do ano (quente e frio) usando análises de variância univariadas e multivariadas. As poças de marés diferiram uma das outras principalmente em área, volume e complexidade do substrato. Observamos 13 espécies de peixes (10 famílias) pertencentes a cinco guildas tróficas (herbívoros vágeis, herbívoros territoriais, invertívoros, onívoros e carnívoros). A densidade e riqueza de peixes foram praticamente constantes ao longo do período estudado, exceto pela redução na densidade de peixes observada na poça da Praia de São Francisco durante o período frio. Entretanto, as assembleias das quatro poças de maré diferiram uma das outras quanto à composição e abundância relativa das espécies, resultando em quatro poças com diferentes estruturas de assembleias. O amboré *Bathygobius soporator* foi a espécie dominante na maioria das localidades, correspondendo a 66% de todos os peixes registrados. A poça da Ponta do Baleeiro apresentou tamanho intermediário, mas esteve coberta por uma complexa comunidade de algas e abrigou a assembleia de peixes mais diversa nesse estudo, sugerindo que a complexidade de habitat pode contribuir para a diversidade de peixes.
Palavras-chave: *peixes subtropicas, entremarés, fatores ambientais, riqueza de espécies, sudeste do Brasil.*

Introduction

Tide pools are formed during low tides when water is retained in depressions in rocky or sandy shores, and serve as a refuge for living organisms during intertidal zone exposure (Griggs 2007). Because of their cyclic isolation during low tides, organisms living in tide pools are exposed to rapid changes in physical-chemical conditions (i.e. temperature, salinity and incident irradiance) that can be more or less marked depending on the time of year, making tide pools a diversified, but very selective habitats for organisms adapted to such oscillations (Fuji 1988, Horn et al. 1999, Griggs 2007). Even small differences in the size, depth and position of tide pools through the intertidal gradient can affect the diversity and structure of their associated communities. For instance, taxon richness can be affected by tide pool height and depth (Firth et al. 2014). Large and deep tide pools located close to the subtidal fringe are usually more stable and support more diverse benthic and nektonic assemblages (e.g. Godinho & Lotufo 2010). On the other hand, pools more distant from the subtidal fringe are exposed to extreme environmental conditions, such as lower oxygen concentration and, thus, tide pools spatially close may be more similar (e.g. Rosa et al.1997).

Substrate composition of tide pools is another factor that affects the species richness and abundance of fishes (e.g. Arakaki et al. 2014). For example, pools of non-limestone rock formations tend to support more abundant and diverse tide pool assemblages than those composed of limestone at the same latitude. As a global pattern, tropical areas tend to support a more diverse community than subtropical or temperate areas. Southwestern Atlantic subtropical intertidal zones are mainly composed of rocky shores formed by igneous and/or metamorphic rocks. At these sites, local assemblages of fishes are generally influenced by interactions among larval availability and effectiveness of recruitment, as well as many levels and frequencies of disturbances, predation pressures and resource competition, being strongly context-dependent (Sale 1991a, 1991b, Rocha et al. 2005, Robertson et al. 2006, Sale 2006, Gibran & Moura 2012).

Studies on organisms from tide pools have focused on settlement patterns (e.g. Beckley 1985), population dynamics (e.g. Sideman & Mathieson 1985, Emson & Whitfield, 1989), a single species or genera (e.g. Jonhsson 1994, Valentine et al. 2007), larval supply and recruitment (e.g. Pfister 1996), diversity (e.g. Leon et al. 1993, Barreiros et al. 2004), bottom-up and top-down effects on ecosystem functioning (e.g. Menge 2000, Methratta 2004, Masterson et al. 2008) and, more recently, on ecophysiology *vs*. environmental conditions concerning microbial communities or experimental ecology (e.g. Nguyen & Byrne 2014, Ntougias 2014). The diversity and composition of fish assemblages from tide pools have been studied in different regions, mainly at temperate latitudes (e.g. Arruda 1979, Mahon & Mahon 1994, Beja 1995) and for most of the studied regions there is a positive relationship between pool size and fish assemblage attributes such as species richness, abundance and biomass. At the southwestern Atlantic, studies on intertidal fishes are mostly faunal surveys (e.g. Almeida 1973, Rosa et al. 1997, Machado et al. 2015), but in the last decade this has changed, particularly in the Northeast (e.g. Cunha et al. 2007, Cunha et al. 2008, Godinho & Lotufo 2010) and Southeast Brazil (e.g. Barreiros et al. 2004, Macieira & Joyeux 2011), with many studies focusing on community ecology of tide pools (but see Soares et al. 2013 for an ecomorphological approach).

Tide pool fishes can be divided into: (1) true residents, which inhabit these sites permanently including Gobiidae and Blennidae families (Gibson 1982); (2) partial residents, generally juveniles of several families as Labridae (Scarinae), Pomacentridae and Chaetodontidae, in addition to some species of economic interest from families Lutjanidae, Epinephelidae and Haemulidae (Gibson 1982, Mahon & Mahon 1994, Cunha et al. 2007); and (3) occasional, those whose occurrence is only accidental or sporadic (Mahon & Mahon 1994, Cunha et al. 2007). True residents generally display morphological and/or physiological traits (such as tolerance to salinity changes, and pelvic fins modifications for better stability) as adaptations to permanent life in such peculiar habitats (see Zander et al. 1999). Generally, these individuals are rarely found at the infralittoral, because the tide pools are their principal habitat, even at high tide. In a different way, juveniles of partial residents use these habitats to obtain food and protection during critical development phases; so, tide pools also contribute to fish diversity outside the pools (Horn et al. 1999).

Marine construction and other human activities are responsible for the increasing loss of coastal habitats. By being located in an area of transition between the terrestrial and marine environment, tide pools are subject to more impacts and disturbances than more remote areas of the coast. The dynamics of their community structures can be easily tampered with, especially when changes occur to the local hydrodynamics, which leads to bottom-up effects that reach the fish assemblages (e.g. Freitas et al. 2009). Within this framework, this study aims to characterize and compare four subtropical tide pools based on their environmental variables and fish assemblage composition and abundances. The study was conducted in a poorly managed region that suffers severe impacts from an expanding seaport, oil terminals, sewage discharge, dredging and coastal development (Gibran & Moura 2012).

Material and Methods

1. *Study area*

The São Sebastião Channel (23º41'-23º54'S; 45º19'-45º30'W) is a 25 km strait in the northern coast of São Paulo State, southeastern Brazil, located between the São Sebastião Island and the mainland (Figure 1). The coastline of the São Sebastião Channel has several tide pools. However, most of those are very small, lacking macroorganisms, or are not completely isolated as "true pools" from the adjacent areas (personal observations during sampling). The only isolated tide pools, during low tide periods, in which we observed macroorganisms at the study area were the four pools studied here and hence restricting our sample size. The pools are located at different sites along the Channel: São Francisco's Beach (SF), Pitangueiras's Beach (PB), Baleeiro's Isthmus (BI) and Baleeiro's Point (BP) (Figure 1). Pitangueiras's Beach, Baleeiro's Isthmus and Baleeiro's Point are located inside no-take Marine Protected Areas (MPAs); the latter two are associated with the Center for Marine Biology, University of São Paulo (CEBIMar-USP) and, thus, have better access to control and implemented conservation policies. Baleeiro's Point and Baleeiro's Isthmus are located parallel to the coast and are 80 m apart. Baleeiro's Point and Baleeiro's Isthmus are 800 m away from Pitangueiras's Beach and 10 km from São Francisco's Beach (Figure 1).

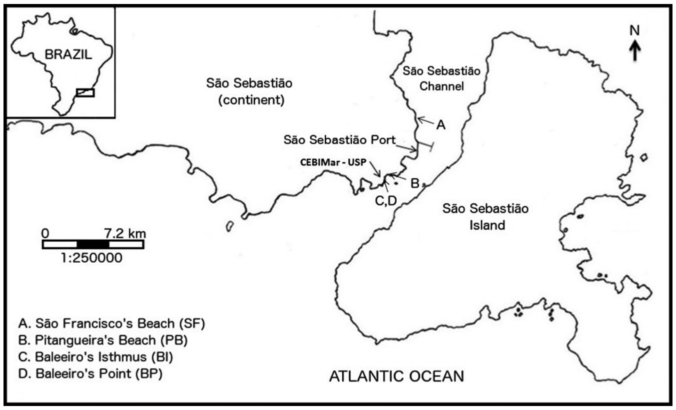

Figure 1. Sampled sites A, B, C, D at the continental margin of São Sebastião Channel, southeastern Brazil. Sites B, C and D are inside a protected area. Modified from Gibran (2010) and (Gibran & Moura (2012).

2. *Sampling design*

We sampled between January and December 2011 (92 hours of fieldwork). Samples were diurnal, monthly, replicated for each tide pool (12 samples per pool, totaling 48 samples in a year), and performed during 3-4 consecutive days always during low spring tides. The protocol included measures of the environmental variables and visual censuses of fish assemblages as described in the next sections.

3. *Environmental variables*

During each sampling interval we recorded biotic (percent of algal cover, benthic complexity, richness and abundance of fishes) and abiotic variables (perimeter, average depth and average rugosity of tide pools, number of crevices and type and proportion of mineral substrata). To estimate the average depth of tide pools, we measured depth in five haphazardly selected locations within each tide pool. We calculated rugosity using the method suggested by Macieira & Joyeux (2011), considering linear and real distances along one segment on the length of each tide pool. Surface areas of tide pools were estimated by adding up triangular areas of known sides, until the whole pool was measured. Tide pool volume was estimated by calculating the product of surface area and average depth.

We visually estimated the algal cover on a scale ranging between 0 and 100% with intervals of ten points (see Bennett & Griffiths 1984). Substrate types were categorized as sand, gravel or rock and were measured by the same method described above. To record the benthic complexity, the benthic cover was scored based on the percent cover of dominant organisms (cf. Gibran & Moura 2012): (0) bare rock, sediment or little cover of filamentous and encrusting calcareous algae; (1) uniform cover of up to two dominant species (e.g. barnacles, *Sargassum* spp., mussels); (2) at least three dominant benthic cover species; (3) diversified, colorful and heterogeneous benthic cover with no obviously dominant species.

4. *Visual censuses of fish assemblages*

We determined species richness, abundance and composition of fishes from visual censuses (12 samples per pool, totaling 48 samples in a year) with the observer outside the pools and always with good water visibility to see the bottom and the edge of each pool. To ensure that all fishes were recorded we also checked all crevices and hiding places manually or with a stick. Each census lasted between 10-15 minutes depending on the size and number of fishes of the sampled pool. When species identification was not possible during census, we temporarily collected and photographed the individuals for subsequent determination. Fish density was calculated by dividing the number of individuals of each species by tide pool volume. Fishes were then classified according to: (1) level of association with the pools (i.e. permanent, partial or occasional residents) based on Gibson (1982), Mahon & Mahon (1994) and Cunha et al. (2007); (2) vertical distribution in the water column (i.e. benthic, nektobenthic or nektonic) according to Lincoln et al. (2009); and (3) trophic guild according to Ferreira et al. (2004). For species whose juveniles and adults belong to different trophic guilds, only the guild of the observed life phase was considered.

5. *Statistical analysis*

To compare environmental conditions among tide pools we used normalized environmental variables to build a similarity matrix among samples using Euclidian distance to provide a general overview of similarities among pools. We tested multivariate differences among pools using analysis of similarity (ANOSIM) and the environmental variables that most contributed to the differences were identified using the SIMPER procedure. We also performed univariate ANOVA using each sampled variable, both biotic and abiotic, followed by post-hoc pair wise comparisons using a Tukey test.

Fish density and species richness data were log x+1 transformed to achieve parametric prerequisites of normality and homoscedasticity and then compared among tide pools and periods (warm/wet and cold/dry) using a two-way ANOVA on monthly replicates. Pairwise comparisons were performed using a Tukey post-hoc test.

The warm/wet period corresponded to the Austral Spring and Summer months (January to March and October to December) while the cold/dry period corresponded to Austral Autumn and Winter months (April to September). Our aim in considering the period effect was not to discuss seasonality of fish assemblage or to attribute the differences observed in time to specific environmental conditions since we did not replicate the seasons or have a proper test of environmental conditions effects on fish assemblages. However, including periods (warm/wet and cold/dry) as a factor, allowed us to increase the amount of data variation explained by our test, and thus to better understand the variation of fish assemblages within the studied period and among tide pools.

We used Bray-Curtis distances to calculate the similarities among fish assemblages to perform an nMDS plot (Clarke 1993) and a PERMANOVA test with 999 permutations (Anderson 2001), considering tide pool and periods (warm/wet and cold/dry) as factors. Significant sources of variation were subjected to pairwise comparisons with 999 permutations. To compare the multivariate dispersions of samples among tide pools we used a PERMDISP analysis. Taxonomic groups responsible for differences among-tide pools and periods were identified using the SIMPER procedure. SIMPER was restricted to the first three ranked taxa holding a contribution for the total variance of 8% or higher.

Results

The four studied tide pools differed from each other regarding the environmental variables (ANOSIM, Global R = 0.61, $p = 0.001$, all pairwise $p < 0.05$) mainly in area, volume and composition of the substrata (Table 1). PB is a small and shallow tide pool composed of rocks and several crevices, most of it covered by turf algae (90%); BP is a tide pool with intermediate area and volume, and also has a high proportion of algae (90%), while SF and BI are large tide pools covered by sand with few algae (30-35% coverage). BP had the most diverse benthic cover, being dominated (75% coverage) by a diversified, colorful and heterogeneous benthic community with no obviously dominant species (level 3 of benthic complexity), while the remaining tide pools were covered (76-90%) by a uniform cover of up to two dominant species (level 1).

We recorded 2,557 fish belonging to 13 species (10 families) and to five of the eight trophic categories: roving herbivores, territorial herbivores, mobile invertebrate feeders, omnivores and carnivores. Most fishes (60 to 83% of total abundance) were permanent residents, except in BP where the abundance of permanent and partial residents was similar, respectively 37% and 46% of the total. Occasional species corresponded on average to 0-15% of the total abundance. At all sites juvenile fishes were abundant (SF = 42%; PB = 62%; BI = 42%; BP = 44% of total). In PB and BI, juveniles from permanent resident species were more abundant than non-residents, but in SF and BP we observed the opposite. According to vertical distribution in the water column we recorded six species of benthic, five nektobenthic and two nektonic fishes (Table 2).

The most abundant species was *Bathygobius soporator* (Valenciennes 1837), corresponding to 66% of fishes recorded, followed by *Diplodus argenteus* (Valenciennes 1830) (12%) and *Mugil curema* (Valenciennes 1836) (10%). *Ctenogobius boleosoma* (Jordan & Gilbert 1882), *Malacoctenus* spp., *Parablennius pilicornis* (Cuvier 1929), *Scartella cristata* (Linnaeus 1758), *Sphoeroides greeleyi* (Gilbert 1900), *Stegastes fuscus* (Cuvier 1830) and *Micrognathus crinitus* (Jenyns 1842) were only observed in BP. Altogether, tide pools were dominated almost always by a single species (*B. soporator* or *M. curema*), with the exception of BP, which was dominated by two species, *B. soporator* and *D. argenteus* (Figure 2).

During the warm period all of the studied tide pools held higher densities of fishes than the cold period. However, during the cold period the density of fishes was very low in SF (Tables 3 and 4). During the whole study BP held the richest assemblage with 4.9 ± 0.64 (mean ± SE) fish species per sample. The mean species richness from BI, PB and SF did not differ from each other, ranging from 1.08 to 1.9 species per sample (Tables 3 and 4). BP along with SF displayed more functional groups than PB and BI, which were dominated by carnivores (Figure 3). Carnivores and omnivores were present in all tide pools, and carnivores were the more abundant guild. Roving and territorial herbivores were recorded just in SF and BP, respectively (Figure 3).

The four tide pools displayed different fish assemblages (all pairwise tests $p < 0.05$) with dissimilarities ranging from 39% between PB and BI to 68% between SF and BP (Table 5; Figure 4). The assemblages from the tide pools SF and BP were more variable through the year period than assemblages from BI and PB (PERMDISP, $p<0.001$). The species that contributed the most to the differences in fish assemblages among tide pools were *B. soporator* and *A. saxatilis,* which were abundant in PB, BI and BP but scarce or absent in SF; *D. argenteus* that occurred in SF, BI and BP, but not in PB; and *M. curema,* which was only observed in SF (SIMPER analysis, Table 5). Temporal variation of the fish assemblages was caused mainly by larger abundance of *B. soporator* during the warm period and by the larger abundance of *D. argenteus* during the cold period (SIMPER analysis, Table 5).

Discussion

Each tide pool studied here had peculiar characteristics, and even the spatially closest pools showed significant environmental differences, mainly in area, volume and complexity of substrata. Tide pools were dominated mostly by juveniles of permanent residents and common species of the southeastern Brazil, such as *Bathygobius soporator* and *Abudefduf saxatilis.* Density and richness of fish showed a small amount of variation during the studied period, except for a reduction in fish density in SF during the cold period. However, the proportion of the most abundant fish within tide pools was extremely variable, resulting in four distinct assemblage structures. The tide pool with the least algal coverage (BI), even being among the largest tide pool in our study area, supported a less diverse fish assemblage than a tide pool of intermediate size covered by a more diversified benthic community (BP), suggesting that habitat complexity may contribute to the diversity of fish. The other large tide pool, SF, was also covered by algae, and had only four species of fish. However, each species belongs to a different functional group, resulting in a high functional diversity. On the other hand, the shallowest pool (PB), held only two fish species and was dominated by *Bathygobius soporator.*

The studied pools had smaller size and volume than those found in other regions of southeastern Brazil. Even the largest tide pools (SF and BI) had a smaller area and volume than those from tropical areas from the Southwestern Atlantic, such as those from the northeastern coast of Brazil (e.g. Rosa et al. 1997, Cunha et al. 2008, Godinho & Lotufo 2010). In our study area, rocky shores are formed by igneous

Table 1. Mean and standard error of the values obtained for tide pool characteristics and the contributions of different mineral substrates in the four studied tide pools, together with the p and F values obtained through statistical analysis, where: SF = São Francisco's Beach, PB = Pitangueiras's Beach, BI = Baleeiro's Isthmus, BP = Baleeiro's Point.

Characteristics	SF	PB	BI	BP	p	F
Depth (m)	0.12 ± 0.01	0.05 ± 0.00	0.12 ± 0.01	0.17 ± 0.01	< 0.001	39.3
Area (m^2)	7.54 ± 0.76	1.59 ± 0.13	7.46 ± 0.64	3.48 ± 0.33	< 0.001	32.9
Volume (m^3)	0.94 ± 0.13	0.08 ± 0.01	0.93 ± 0.10	0.59 ± 0.06	< 0.001	21.7
Rugosity	1.04 ± 0.00	1.14 ± 0.02	1.13 ± 0.01	1.10 ± 0.01	< 0.001	9.5
Sand (%)	66.67 ± 6.55	6.67 ± 4.97	58.33 ± 4.05	0.83 ± 0.83	< 0.001	55.0
Rock (%)	26.67 ± 7.91	75.00 ± 7.02	35.83 ± 4.52	75.83 ± 4.68	< 0.001	8.6
Gravel (%)	6.67 ± 1.88	16.67 ± 3.10	5.83 ± 1.93	23.33 ± 3.96	< 0.001	17.2

Table 2. Total fish abundance recorded per tide pool between January and December 2011 with classification of trophic guilds (see reference below), vertical distribution and level of association. OMN = omnivore, TERH = territorial herbivore, MIF = mobile invertebrate feeder, CAR = carnivore, ROVH = roving herbivore, N = nektonic, B = benthic, NC = nektobenthic, RPA = resident partial, RPE = resident permanent, O = occasional, SF = São Francisco's Beach, PB = Pitangueiras's Beach, BI = Baleeiro's Isthmus, BP = Baleeiro's Point.

Families/Species	Trophic guilds	Vertical distribution	Level of association	SF	PB	BI	BP
Atherinopsidae							
Atherinellla brasiliensis	OMN[1,2]	N	O	-	-	2	2
Blenniidae							
Parablennius pilicornis	OMN[3,4]	B	RPA	-	-	-	10
Scartella cristata	TERH[3,5]	B	RPA	-	-	-	14
Gerreidae							
Eucinostomus gula	MIF[2,1]	NB	RPE	28	-	3	80
Gobiidae							
Bathygobius soporator	CAR[6]	B	RPA	54	200	1176	260
Ctenogobius boleosoma	MIF[6,7]	B	RPA	-	-	-	6
Labrisomidae							
Malacoctenus spp.	MIF[4,8]	B	RPA	-	-	-	4
Mugilidae							
Mugil curema	ROVH[2]	N	O	275	-	-	-
Pomacentridae							
Abudefduf saxatilis	OMN[5,8]	NB	RPA	-	3	40	82
Stegastes fuscus	TERH[8,9]	NB	RPA	-	-	-	3
Sparidae							
Diplodus argenteus	OMN[8,9]	NB	RPA	44	-	102	162
Syngnathidae							
Micrognathus crinitus	MIF[4,7]	B	O	-	-	-	5
Tetraodontidae							
Sphoeroides greeleyi	MIF[5,6]	NB	RPA	-	-	-	2

[1]Figueiredo & Menezes (1978), [2]Paiva et al. (2008), [3]Dinslaken (2008), [4]Halpern & Floeter (2008), [5]Chaves & Monteiro-Neto (2009), [6]Corrêa & Uieda (2007), [7]Carvalho-Filho (1999), [8]Floeter et al. (2007), [9]Ferreira et al. (2004).

and/or metamorphic rocks with tide pools occurring inside natural depressions, while on the northeastern coast and other reef-forming areas, the tide pools are mainly formed by large crevices in sandstone reefs (i.e. higher structural complexity). Also, tide pools from tropical areas tend to support more diverse assemblages than tide pools from temperate areas, regardless the substrata. In our study, we observed 13 species of fishes, as already reported for other tide pools under similar latitude, while those in the tropics had at least 40 species (e.g. Rosa et al. 1997, Cunha et al. 2008). These differences from other studied areas could be a sampling effect, since we only studied four tide pools, which may restrict the generalization of our conclusions. On the other hand, as far as we know, the tide pools studied here are the only real pools from the continental portion of the São Sebastião Channel.

The small volume of the São Sebastião Channel tide pools may contribute to their low species richness, as expected by the species/area relationship. Larger pools, besides having a larger area, exhibit a more temporally constant habitat, allowing less tolerant species to occur, which increases diversity (Rosa et al. 1997). However, in our study, the largest tide pools (SF and BI) held a less specious assemblage than BP, which is a tide pool of intermediate size, differing from other studies, where abundance and richness are positively correlated to tide pool volume (Cunha et al. 2007). Other factors, such as tide pool isolation, hydrodynamics and algal cover, may interact with volume and area to determine the distribution and structure of fish assemblages inside tide pools (Godinho & Lotufo 2010, Macieira & Joyeux 2011). In the intertidal zone, intense water turbulence restricts biological activities such as feeding and reproduction of some benthic fishes, such as labrisomids and blenniid species like *Malacoctenus* spp. and *Parablennius pilicornis*. Besides being protected from wave and wind actions, BP is also mostly covered by a diverse benthic community, allowing the occurrence of fishes with specific diets, such as the territorial herbivorous damselfish *Stegastes fuscus*. On the other hand, the largest tide pools SF and BI are covered by sand and bare rock, that may restrict the occurrence of some species. Although PB is the smallest pool, it is also the most elevated, which isolates the fish assemblages and reduces water inflow and colonization during high tides, resulting in an assemblage with only two species. Considering that we only sampled four tide pools and that those differed from each other on several physical and biological attributes, a larger sample size is necessary to properly test how tide pool size and complexity interact to determine fish diversity.

While species composition might differ among tide pools around the world, Gobiidae and Bleniidae families are frequently reported as dominant in several localities, such as Japan (Arakaki et al. 2014),

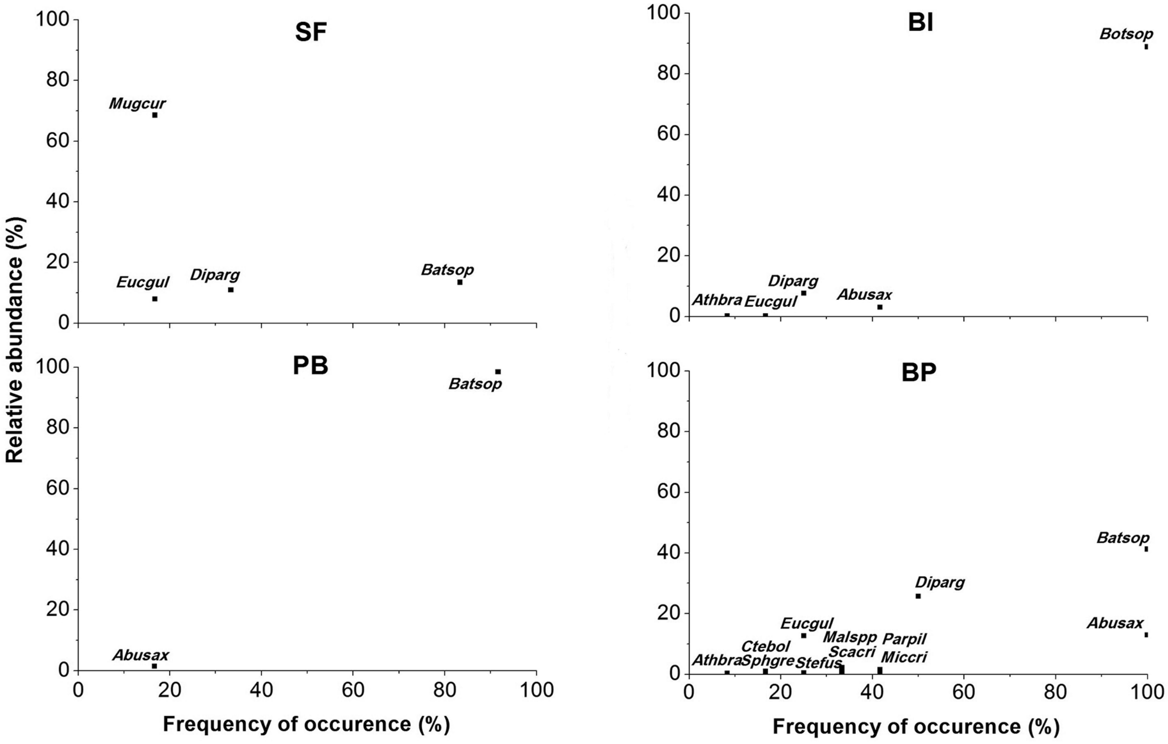

Figure 2. Relative abundance and frequency of occurrence of fish fauna in the tide pools of São Francisco's Beach (SF), Pitangueiras's Beach (PB), the Baleeiro's Isthmus (BI) and Baleeiro's Point (BP), between January and December 2011. Species codes: first three letters of the genus name followed by the first three letters of the specific epithet: *Abusax* = *Abudefduf saxatilis*, *Athbra* = *Atherinella brasiliensis*, *Batsop* = *Bathygobius soporator*, *Ctebol* = *Ctenogobius boleosoma*, *Diparg* = *Diplodus argenteus*, *Eucgul* = *Eucinostomus gula*, *Malspp* = *Malacoctenus spp.*, *Miccri* = *Micrognathus crinitus*, *Mugcur* = *Mugil curema*, *Parpil* = *Parablennius pilicornis*, *Scacri* = *Scartella cristata*, *Sphgre* = *Sphoeroides greeleyi*, *Stefus* = *Stegastes fuscus*. Tide pools codes: SF = São Francisco's Beach, PB = Pitangueiras's Beach, BI = Baleeiro's Isthmus, BP = Baleeiro's Point.

Table 3. Fish density and species richness in the four studied tide pools (average ± standard error)

Tide pool	Abundance (individuals/m^3)		Richness (number of species)
Period	Warm/wet	Cold/dry	
São Francisco's Beach (SF)	67.7 ± 30.3	9.81 ± 4.1	1.5 ± 0.2
Pitangueiras's Beach (PB)	290.0 ± 85.6	165.3 ± 63.5	1.1 ± 0.1
Baleeiro's Isthmus (BI)	136.6 ± 30.7	121.9 ± 10.3	1.9 ± 0.2
Baleeiro's Point (BP)	119.7 ± 13.0	77.2 ± 19.4	4.9 ± 0.6

Table 4. Analyse of variance considering the effects of tide pools and period (warm/wet or cold/dry) on fish density and species richness and PERMANOVA on the structure of fish assemblage. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$.

	Species richness			Fish density			Fish assemblage		
Source	Df	MS	F	Df	MS	F	Df	MS	Pseudo-F
Season (SE)	1	1.69	1.05	1	0.06	0.26	1	93172.9	3.52*
Tide pool (TP)	3	36.41	22.69***	3	2.41	9.70***	3	9600.9	10.66**
SE x TP	3	0.63	0.76	3	0.80	3.24*	3	1055.5	1.17
Error	40	1.60		40	0.25		39	900.18	

Mediterranean Sea (Macpherson 1994) and Hawaiian Island (Cox et al. 2011). The most abundant species in this study, *B. soporator*, is also abundant in other tide pools of the Brazilian coast (e.g. Rosa et al. 1997, Barreiros et al. 2004). In the studied area, it accounts for the preponderance of permanent residents, followed by *Scartella cristata*, *P. pilicornis* and *Ctenogobius boleosoma*. Permanent resident fishes have morphological, physiological and behavioral adaptations to live in these environments (Floeter et al. 2001, Barreiros et al. 2004, Macieira 2008), and *B. soporator* is rarely found outside tide pools (see Gibran & Moura 2012). On the other hand, occasional visitants are less frequent and abundant, because they enter the pools during high tides and are trapped when the tide recedes. They are more frequent in pools closer to the

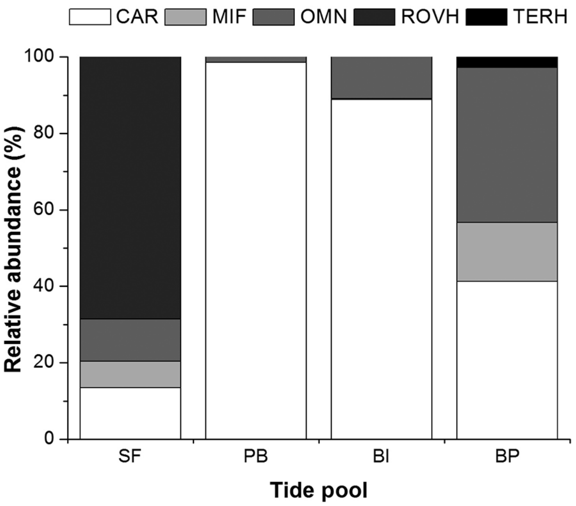

Figure 3. Relative abundance of each trophic guild present in the tide pools of São Francisco's Beach (SF), Pitangueiras's Beach (PB), the Baleeiro's Isthmus (BI) and Baleeiro's Point (BP), between January and December 2011. CAR = carnivore, MIF = mobile invertebrate feeder, OMN = omnivore, ROVH = roving herbivore, TERH = territorial herbivore.

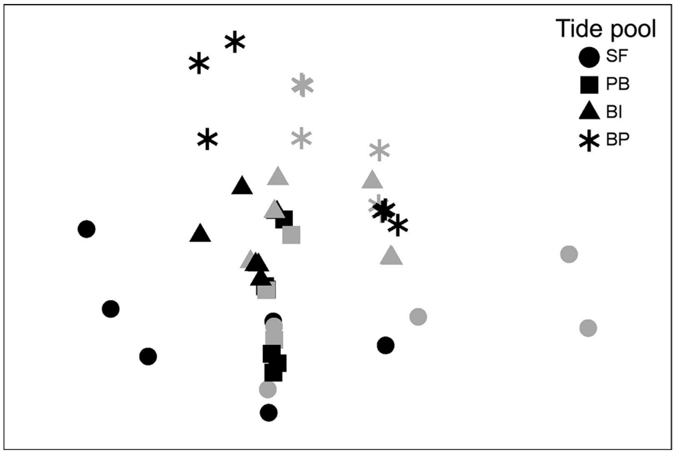

Figure 4. Non-metric multidimensional scalling plot of fish assemblages from tide pools of São Francisco's Beach (SF), Pitangueiras's Beach (PB), the Baleeiro's Isthmus (BI) and Baleeiro's Point (BP) in the studied tide pools. Black symbols represent the Warm/Wet season, while Grey symbols represent the Cold/Dry season. Stress = 0.12.

Table 5. Simper analyses showing the contribution of species for the dissimilarity among tide pools and periods of the year, where: SF = São Francisco's Beach, PB = Pitangueiras's Beach, BI = Baleeiro's Isthmus, BP = Baleeiro's Point.

SF x PB	**Dissimilarity = 49%**
Species	**Contribution (%)**
Bathygobius soporator	42.5
Diplodus argenteus	26.2
Mugil curema	14.3
SF x BI	**Dissimilarity = 62%**
Species	**Contribution (%)**
Bathygobius soporator	49.4
Diplodus argenteus	19.8
Abudefduf saxatilis	8.8
SF x BP	**Dissimilarity = 68%**
Species	**Contribution (%)**
Abudefduf saxatilis	23.2
Diplodus argenteus	19.0
Bathygobius soporator	15.1
PB x BI	**Dissimilarity = 39%**
Species	**Contribution (%)**
Bathygobius soporator	50.4
Abudefduf saxatilis	23.8
Diplodus argenteus	19.2
PB x BP	**Dissimilarity = 58%**
Species	**Contribution (%)**
Abudefduf saxatilis	25.2
Diplodus argenteus	24.6
Bathygobius soporator	8.7
BI x BP	**Dissimilarity = 50%**
Species	**Contribution (%)**
Diplodus argenteus	22.0
Abudefduf saxatilis	18.8
Bathygobius soporator	17.5
Warm/Wet x Cold/Dry	**Dissimilarity = 51%**
Species	**Contribution (%)**
Bathygobius soporator	27.7
Diplodus argenteus	23.0
Abudefduf saxatilis	17.3

infralittoral (Barreiros et al. 2004), as SF and BL. The high percentage of occasional individuals in SF result from numerous juveniles of *Mugil curema*, that can be attributed to the schooling behavior of this species, while in BI, *Sphoeroides greeleyi*, *Micrognathus crinitus* and *Atherinella brasiliensis* were the most abundant occasional species.

Considering the degree of conservation of each pool, we propose that despite three pools being in protected areas (BP, BI and PB) and two with controlled access (BP and BI), this factor was not strongly related to diversity and trophic complexity. In this study, intrinsic factors of each pool, such as algal coverage (primary productivity), may be the most responsible for the distribution of species found, but a proper test of this hypothesis is needed. The importance of the temporal variation of fish assemblages was also small when compared to the variation among tide pools. The richness of fish was constant during the whole study and we only observed differences in the abundance of fish for the tide pool in SF. The differences in the organization of the fish assemblages observed between the warm and cold period seems to be related to variation in the abundance of the most common species and not to a turnover of species during the year, and was more pronounced in SF and BP, which were the pools with the smallest abundance of fish during the study. This small variation of the fish assemblage during the year can be related to the small number of species in tide pools, to an intrinsic variation of conditions during the studied period, or to the study area location in a subtropical zone, in which temporal variation seems to play an underestimated small role in the diversity of fish when compared to tropical areas.

Because most of the partial resident individuals are recruits and juveniles, tide pools seem to work as nursery sites for nektobenthic species that use tide pools for reproduction and early development (Horn et al. 1999). Because of (1) their importance to fishes that do not occur at the infralittoral zone and for juvenile fishes from adjacent seawater during reproduction, and also (2) of their location in a transition zone between ocean and mainland, highly influenced by continental development and human activities, tide pools need to be considered as a peculiar marine habitat and also considered during Marine Protected Area (MPA) establishment (see Myres et al. 2011). Our study described the fish assemblages from the few real tide pools from the continental portion of the São Sebastião Channel and shows that while most of the species occurring in the four tide pools are very similar, there is a large differentiation between fish assemblages among tide pools caused mainly by differences in the abundance of *B. soporator, A. saxatilis* and *D. argenteus*.

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List of Odonates from the Floresta Nacional de São Francisco de Paula (FLONA - SFP), with two new distribution records for Rio Grande do Sul, Brazil

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RENNER S., PÉRICO, E., SAHLÉN, G. **List of Odonates from the Floresta Nacional de São Francisco de Paula (FLONA - SFP), with two new distribution records for Rio Grande do Sul, Brazil**. Biota Neotropica. 16(3): e20150132. http://dx.doi.org/10.1590/1676-0611-BN-2015-0132

Abstract: A survey of Odonata was carried out in the National Forest FLONA - SFP, Northeastern region of the Rio Grande do Sul state, Brazil. This conservation unit is mainly covered by Mixed Ombrophilous Forest (MOF), a subtype of Atlantic Forest biome, being also areas covered in planted *Pinus*, planted *Araucaria* and open fields. Our sampling efforts were conducted in thirty aquatic environments inside this reserve during the period between January 2014 and November 2014. The sampling sites were selected randomly, comprehending lakes, bogs, small streams and river sections, all inserted in the four vegetation types occurring in the reserve. Fortysix species of Odonata were collected and grouped into 23 genera and seven families. The dominant families were Coenagrionidae (32%), Libellulidae (32%), Aeshnidae (12%), and, Calopterygidae and Lestidae (9%). As expected, the findings revealed the presence of a highly diverse Odonate assemblage, mainly represented by generalist species in the most human disturbed sectors (*Pinus* and Open fields) and some specialist species in the pristine forest. Two species were registered for the first time in the state of Rio Grande do Sul, Brazil: *Libellula herculea* Karsch, 1889 (Libellulidae) and *Heteragrion luizfelipei* Machado, 2006 (Heteragrionidae).
Keywords: *Ecology, Odonata, inventory, Atlantic Forest, distribution.*

RENNER S., PÉRICO, E., SAHLÉN, G. **Lista de Odonata da Floresta Nacional de São Francisco de Paula (FLONA - SFP), com dois novos registros de distribuição para o Rio Grande do Sul, Brasil**. Biota Neotropica. 16(3): e20150132. http://dx.doi.org/10.1590/1676-0611-BN-2015-0132

Resumo: Uma pesquisa de Odonatas foi desenvolvida na Floresta Nacional de São Francisco de Paula (FLONA - SFP), Rio Grande do Sul, Brasil. Esta unidade de conservação é, em grande parte, coberta por Floresta Ombrófila Mista, um subtipo de floresta do Bioma Mata Atlântica, sendo também áreas cobertas com plantação de *Pinus*, *Araucaria*, além de áreas de campo aberto. Nossos esforços de coleta abrangeram 30 ambientes aquáticos dentro da reserva, no período de Janeiro de 2014 e Novembro de 2014. Os locais de coleta foram selecionados aleatoriamente, compreendendo lagos, banhados, pequenos arroios e seções de rio, todos inseridos nos quatro tipos de vegetação ocorrentes na reserva. Foram coletadas 46 espécies de Odonata, agrupadas em 23 gêneros e sete famílias. As famílias dominantes foram Coenagrionidae (32%), Libellulidae (32%), Aeshnidae (12%), e, Calopterygidae e Lestidae (9%). Como esperado, os resultados revelaram a presença de uma alta diversidade de Odonata, em sua maioria generalistas, encontrados nos setores mais impactados pelo homem (*Pinus* e Campos abertos) e algumas espécies especialistas na floresta primária. Duas espécies foram registradas pela primeira vez no estado do Rio Grande do Sul, Brasil: *Libellula herculea* Karsch, 1889 (Libellulidae) e *Heteragrion luizfelipei* Machado, 2006 (Heteragrionidae).
Palavras-chave: *Ecologia, Odonata, inventário, Floresta Atlântica, distribuição.*

Introduction

A big number of studies to describe the diverse fauna and flora in the Neotropics are already being published or under way. Even with so intense efforts, we are still far from the knowledge levels achieved in Europe and North America, or the northern hemisphere in general. In this part of the world, we even still lack reliable estimates of how many species occurs (May 1998, Scotland & Wortley 2003). Every day the information on the diversity of species becomes more relevant, as well as the knowledge on the species distribution and richness, which can provide several tools to put focus on human impact on these environments (Lewis 2006). To prioritize among areas under consideration for conservation, biologists and decision makers need concise information on species diversity, especially in threatened habitats like the Atlantic Forest (Kerr et al. 2000).

The Brazilian Odonata are still poorly described, despite a few publications with ecological approach, species lists are known only from São Paulo (Costa et al. 2000); Espírito Santo (Costa & Oldrini 2005); Minas Gerais (Souza et al. 2013) for Mata do Baú specifically; and Mato Grosso (Calvão et al. 2014) this last one being restricted only to a small region of the state. The most recent Odonata inventory for the state of Rio Grande do Sul is Renner et al. (2015), for the central region, and Kittel & Engels (2014), from a nature reserve (Pró-mata/PUCRS), with sampling performed only in open areas in forest surroundings and restricted to the Zygoptera suborder.

In this paper, we aim to improve the knowledge of the Odonata of the northeastern region of the Rio Grande do Sul state, acquiring a general overview of the species distribution over several types of aquatic environments distributed in Atlantic Forest at relatively high altitudes. As consequence we also expanded species distributions for this state.

Material and Methods

1. *Study area*

All the sampling areas are located within the National Forest FLONA - SFP, at the São Francisco de Paula municipality (Figure 1). This reserve is a National Conservation Unit, administered by de ICMBio (Instituto Chico Mendes de Conservação da Biodiversidade) together with the Brazilian Environment Ministry (Ministério do Meio Ambiente). These units are part of a sustainability plan, being so, part of the total areas are cultivated with planted forest: *Araucaria angustifolia* (Bertol.) Kuntze and *Pinus elliottii* Engelm., producing commercial wood for economical purposes.

The climate in the region is classified as Cfb in the Koppen system, Mesothermic Humid, Temperate Subtropical, with well defined seasons, mean temperatures varying in between 12°C and 16°C, altitudes reaching from 900 m to 1,100 m ASL, and precipitation means above 2,000 mm annually (INPE 2014). The reserve area (1,572 ha) is mainly (720 ha) covered in pristine Mixed Ombrophilous Forest (MOF), a subtype of Atlantic Forest, some areas are covered in planted *Araucaria* (AR), planted *Pinus* (PN) and Open fields (OP), these areas being altered since the 1950's decade. In each of these four vegetation types, three types of aquatic environments were selected: rivers (R), lakes (L) and swamps (S) to provide an overall view of the species occurring in the area.

2. *Data collection*

In total 30 aquatic environments (Figure 1) were sampled: MOF (n = 12), AR (n = 8), PN (n = 6) and OP (n = 4). The aquatic environments were divided in 3 types: rivers (R) n = 14, lakes (L) n = 7 and swamps (S) n = 9. We sampled for dragonflies during the year of 2014, all the sampling sites were visited three times per year, once per season: summer (2 - 11 January); autumn (10 - 18 May); spring (2 - 11 November); excluding the winter season due to the lack of activity of adult Odonata in the low temperatures prevailing. Our sampling efforts were concentrated on adults of Odonata, since the larvae can present great difficulties to the determination work, as only scarce information is available for this region (Garrison et al. 2006). The sampling method used handhold insect nets, performed by a team of two persons, in sunny days, during the peak time of Odonata activities (between 09:00 h to 16:00 h), which is an adaptation of Renner et al. (2015). The collection authorization process was issued by IBAMA, through SISBio system under the number 38928-1.

Species richness was determined by the absolute number of collected specimens. Collection efforts and the number of sampled species were based on the richness percentage estimated by the mean of non-parametric estimator building the collector's curve by Jack1 and Chao1 to estimate the actual number of species. The indexes Jackknife1 and Chao1 were calculated using the Software Estimates (Colwell 2009), with 1000 repetitions. Our sampling efforts were also validated using the estimation of total richness according to Smith & van Belle (1984).

All specimens collected were immediately preserved in 96% ethanol and deposited in the MCNU, afterwards determined in the Evolution and Ecology Laboratory according to Garrison et al. (2006, 2010), Heckman (2006, 2010), Lencioni (2006) and original species descriptions. For the systematics classification, we followed Dijkstra et al. (2014).

Results

1. *Species list*

We found 46 species, belonging to 23 genera in seven families, collected in the 30 aquatic environments inside the National Forest FLONA - SFP, except two, which are located in the immediate surroundings (main access road). This is a relatively large number of species, reflecting the diversity of sampling sites, which includes three types of aquatic systems, and four types of vegetation. In total 454 specimens were collected. The estimated number of species through non-parametric indexes (Jacknife1 and Chao1) are shown in Figure 2, the total estimated number of species through Smith & van Belle (1984) was 55,67, showing we reached 82% (n = 46) of the total estimated number of species on the three sampling occasions.

The Odonate list records from the National Forest FLONA - SFP is presented in Table 1, each species accompanied by the acronym of the vegetation type and aquatic environments with occurrence and collection/voucher ID number.

The families with the higher number of species were Coenagrionidae (32%), Libellulidae (32%), Aeshindae (12%), Calopterygidae and Lestidae (both 9%), as shown in the Table 2. The most common genera were *Erythrodiplax* and *Oxyagrion*, represented by six and four species, respectively. *Erythrodiplax* was the most abundant genus, occurring virtually in all sampling sites. The rarest species found with only one occurrence and in only one season were *Macrothemis marmorata*, *Dasythemys m. mincki* and *Peristicta gauchae*.

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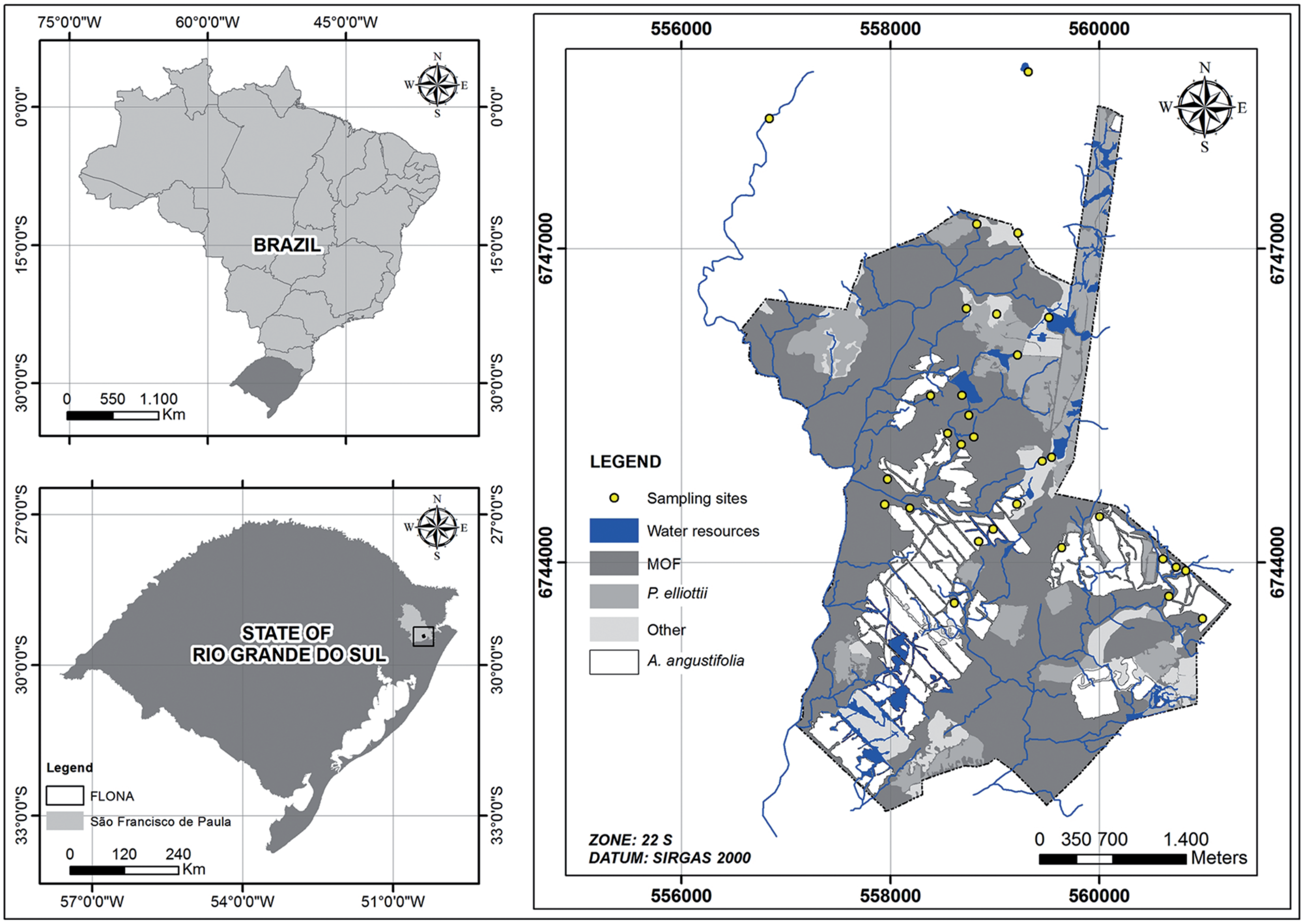

Figure 1. Brazilian map with the insets of the state of Rio Grande do Sul (RS); São Francisco de Paula municipality (light gray); and the complete map of National Forest FLONA - SFP, yellow dots representing the sampling sites in the reserve.

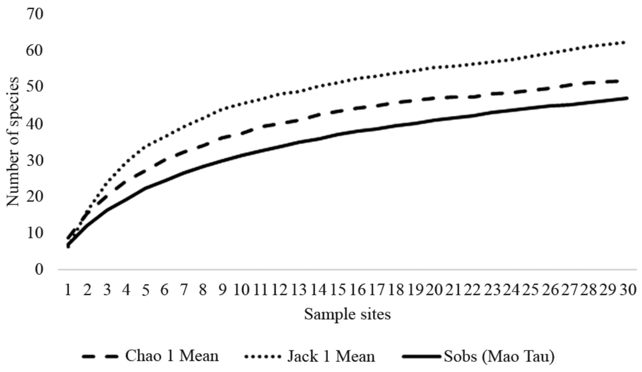

Figure 2. Number of species of Odonata collected (sobs) on the 30 sampling sites in relation to the number of species expected by Jacknife1 and Chao1 estimators in the National Forest FLONA - SFP, state of Rio Grande do Sul (RS), Brazil.

2. *New records*

We found two new records of odonate species for Rio Grande do Sul, one belonging to Libellulidae: *Libellula herculea* Karsch, 1889; and one belonging to Heteragrionidae: *Heteragrion luizfelipei* Machado, 2006.

2.1. *Libellula herculea* Karsch, 1889

Libellula is a Holarctic genus, which occurs also in Europe and Asia, in the New World its species are most diverse in North America: from Alaska and N Canada and to N Argentina in South America. The only species known to be widespread in South America is *Libellula herculea* (Garrison et al. 2006). According to Carle & Kjer (2002) this genus is well diagnosed and is considered a sister group of Orthemis, however an undescribed species is known from Brazil (Garrison et al. 2006). The two species belonging to the genus Libellula recorded here are: *Libellula herculea* and *Libellula* sp. both distributed widely in North, Central America and northern regions of South America, and with our records expanding their distribution southwards. The current distribution records were derived from De Marmels (1982) and Heckman (2006). According to Garrison (2006) the genus distribution ranges from North America to the Paraná state in Brazil, around 250 km northwards of our sampling site. Regarding *Libellula herculea*, the records from Brazil are from Marambaia, Ilha Grande and Itatiaia in the state of Rio de Janeiro (Santos 1970). Our specimens of *Libellula herculea* were found only in a permanent, highly vegetated swamp, inside the *Pinus* (PN) plantation (Figure 3a).

Table 1. Inventory list of Odonata species from the National Forest FLONA - SFP.

Suborder	Family	Species	Vegetation type	Env. type	Collection ID
Zygoptera	Calopterygidae	*Hetaerina longipes* Hagen *in* Selys, 1853	MOF, AR, OP	R, S	ZAUMCN928
		Hetaerina rosea Selys, 1853	MOF, PN	R, S	ZAUMCN929
		Mnesarete borchgravii (Selys, 1869)	MOF, AR	R	ZAUMCN930
		Mnesarete pruinosa (Hagen *in* Selys, 1853)	MOF, OP	R	ZAUMCN931
	Coenagrionidae	*Acanthagrion gracile* (Rambur, 1842)	OP	L	ZAUMCN932
		Acanthagrion lancea Selys, 1876	AR, PN, OP	L, S	ZAUMCN933
		Acanthagrion sp.	MOF, PN	R, L	ZAUMCN934
		Argentagrion ambiguum (Ris, 1904)	AR	S	ZAUMCN935
		Argia indocilis Navás, 1934 (j. syn. *A. croceipennis*)	MOF, AR, OP	R	ZAUMCN936
		Argia sp.	MOF	R	ZAUMCN937
		Homeoura chelifera (Selys, 1876)	AR, PN, OP	L	ZAUMCN938
		Ischnura capreolus (Hagen, 1861)	AR, PN, OP	L	ZAUMCN939
		Ischnura fluviatilis Selys, 1876	AR, PN, OP	L	ZAUMCN940
		Oxyagrion hempeli Calvert, 1909	OP	R	ZAUMCN942
		Oxyagrion microstigma Selys, 1876	PN, OP	R, L	ZAUMCN943
		Oxyagrion terminale Selys, 1876	MOF, AR, PN, OP	R, L, S	ZAUMCN944
		Oxyagrion sp.	MOF, AR, PN, OP	R, L, S	ZAUMCN945
		Peristicta gauchae Santos, 1968	OP	R	ZAUMCN946
		Telebasis theodori (Navás, 1934)	PN, OP	L	ZAUMCN947
	Heteragrionidae	*Heteragrion luizfelipei* Machado, 2006	MOF	R	ZAUMCN948
	Lestidae	*Lestes auritus* Hagen *in* Selys, 1862	PN, OP	L, S	ZAUMCN949
		Lestes bipupillatus Calvert, 1909	OP	S	ZAUMCN950
		Lestes pictus Hagen *in* Selys, 1862	PN	L	ZAUMCN952
		Lestes undulatus Say, 1840	PN, OP	L	ZAUMCN951
Anisoptera	Aeshnidae	*Castoraeschna* sp.	AR, PN	S	ZAUMCN953
		Limnetron sp.	MOF	R, S	ZAUMCN954
		Rhionaeschna bonariensis Rambur, 1842	MOF, AR, PN, OP	R, L, S	ZAUMCN956
		Rhionaeschna brasiliensis Ellenrieder and Costa, 2002	MOF, PN	R, L, S	ZAUMCN955
		Rhionaeschna planaltica (Calvert, 1952)	MOF, AR, PN, OP	R, L, S	ZAUMCN957
	Gomphidae	*Phyllogomphoides regularis* (Selys, 1873)	MOF	R	ZAUMCN958
		Progomphus gracilis Hagen *in* Selys, 1854	MOF, AR, OP	R	ZAUMCN959
	Libellulidae	*Dasythemis mincki mincki* (Karsh, 1890)	AR	S	ZAUMCN960
		Erythrodiplax atroterminata Ris, 1911	MOF	R	ZAUMCN961
		Erythrodiplax fusca (Rambur, 1842)	AR, PN, OP	L	ZAUMCN963
		Erythrodiplax hyalina Förster, 1907	MOF, AR, PN, OP	R, L, S	ZAUMCN964
		Erythrodiplax media Borror, 1942	MOF, AR, PN, OP	R, L, S	ZAUMCN965
		Erythrodiplax sp. (1)	MOF, AR, PN, OP	R, L, S	ZAUMCN962
		Erythrodiplax sp. (2)	OP	L	ZAUMCN966
		Libellula sp.	MOF, PN	S	ZAUMCN967
		Libellula herculea Karsch, 1889	PN	S	ZAUMCN968
		Macrothemis marmorata Hagen, 1868	AR	S	ZAUMCN969
		Micrathyria artemis Ris, 1911	MOF	R	ZAUMCN970
		Oligoclada laetitia Ris, 1911	AR, PN	L, S	ZAUMCN971
		Orthemis discolor (Burmeister, 1839)	PN	S	ZAUMCN972
		Perithemis icteroptera (Selys *in* Sagra, 1857)	PN	L	ZAUMCN973
		Perithemis mooma Kirby, 1889	PN	L	ZAUMCN974

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Table 2. Number of Odonata species collected per family in the National Forest (FLONA - SFP).

FAMILY	Nº	%
ZYGOPTERA		
Coenagrionidae	15	32
Lestidae	4	9
Calopterygidae	4	9
Heteragrionidae	1	2
ANISOPTERA		
Libellulidae	15	32
Aeshnidae	5	12
Gomphidae	2	4
TOTAL	46	100%

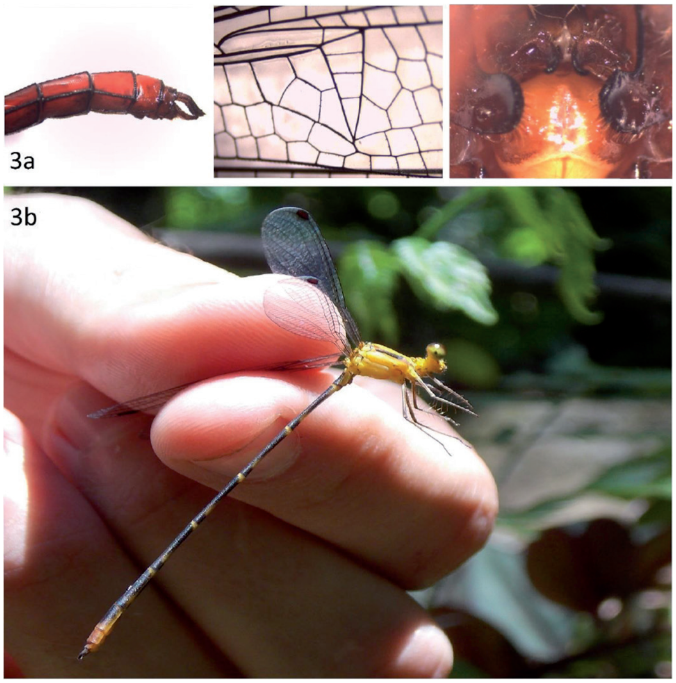

Figure 3. a) Main features of *Libellula herculea*: Male cercus (lateral view); FW base, arculus sectors separated; Genital fossa, ventral view. b) *Heteragrion luizfelipei*, adult male.

2.2. *Heteragrion luizfelipei* Machado, 2006

This species (Figure 3b) have its current distribution restricted to southern and southeastern Brazil, from São Paulo (Machado, 2006), and Paraná to its southern limit in the Santa Catarina state as reported by Lencioni (2013). Here we expand the distribution around 250 km southwards in the Rio Grande do Sul state. No further reported records could be found for this species. Possibly the lack of sampling efforts in the Atlantic coastal zones covered by the Atlantic Forest biome can explain why no sighting of this species have taken place in this region, as it is known as a typical forest genus (Loiola & De Marco 2011). The specimens were collected in the proximities of a slow flowing stream (ca. 50 - 100 cm wide), with boulder/gravel bottom, mostly shaded by the dense pristine Atlantic Forest (MOF).

Most of the biodiversity in the Atlantic Forest still unknown, especially so in the MOF subtype (Southern Brazil), and more studies are needed as shown by the discovery of these two new records. The order Odonata in the Neotropics in general is in need of many taxonomic revisions which will only be made possible through sampling and inventory studies as this one. We also strongly recommend the creation of more protected areas in the region, since it is considered a “hotspot” for biodiversity.

Discussion

The size of regional species pools for dragonflies has only been studied in certain regions of the world. In temperate regions of northern Europe, specifically southern and central Sweden, several studies have reported a regional species pool of between 24 and 30 (Wittwer et al. 2010; Flenner & Sahlén 2008). According to studies from Brazil, Pires et al. (2013) found 30 genera of dragonfly larvae in the Jacuí River Basin; De Marco et al. (2014), listed a species pool of 56, in central Brazi; Monteiro et al. (2013), noted 32 species in Manaus, Amazonas; Anjos-Santos & Costa (2006), registered 77 species, from Rio de Janeiro; and Souza et al. (2013) recorded 47 in Mata do Baú, Minas Gerais. Thus, 46 species fit well into the general pattern for a Subtropical zone, although there may be some additional rare species occurring in the region, the estimated indexes indicate that the size of the species pool is correctly estimated although up to ten more species might occur.

The big numbers of Libellulids occurring in the PN and OP (the most human impacted areas) consists to large part by widespread generalists, a fact that can be attributed to the mosaic of forest and open areas, which by its time favors the occupation by the fast and agile flying dragonflies corroborating the findings from Machado (2001).

There are clear relations between environmental factors (biotic and abiotic) and species composition, these factors acting as determinants of presence and absence of some species due to ecological and physical restrictions (Paulson 2006, Juen et al. 2007). In most preserved areas or bigger forested areas it is expected to find a bigger number of Calopterygids and Heteragrionids, which have more environmental restrictions and ecological needs to occur (Carvalho et al. 2013).

Despite that the occurrence of rare species was mostly restricted to the MOF and AR sectors, the finding of a good number of Coenagrionids in the other sectors (PN, OP) can also mirror good conditions, since some genera have also ecological restrictions related to aquatic vegetation and water quality (e.g. *Homeoura*, *Argia*, *Oxyagrion*) as stated by Garrison et al. (2010).

Regarding the occurrence indicator species, we found a selection which could fit into an indicator of general species richness for fragmented Atlantic forest biome, following Renner et al. (2015), as for example *Ischnura capreolus* and *Homeoura chelifera*, occurring only in the non-natural environments (AR, PN and OP). No endangered species were found, since most of the records are now attributed to “Least concern” or “Data deficient”, according to the IUCN.

The Atlantic forest is one of the most endangered ecosystems in South America, and in this case, more specifically, the Mixed Ombrophilous Forest, one of the less known forest subtypes was explored. For these environments, species inventories can produce a decisive tool for the management actions in order to keep and restore forests and natural resources. This survey made a deepened approach of the poorly known Odonata from Southern Brazil, showing that a great diversity is still occurring despite the human impacted habitats included in this study. This knowledge is useful in providing support data for future conservation measurements in the studied landscape.

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The herpetofauna of Parque Nacional da Serra das Confusões, state of Piauí, Brazil, with a regional species list from an ecotonal area of Cerrado and Caatinga

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Abstract: Different physiognomies at Parque Nacional da Serra das Confusões (PNSCo) were intensively sampled aiming to access the distribution pattern of its herpetofauna. Sixty six species were found in the park (47 reptiles and 19 amphibians); the rarefaction curve for lizards, although not fully stabilized in an asymptote, indicates that the sampling effort was enough to reveal most lizard species occurring in the area; and richness estimators recovered values close to observed. For amphibians, the curve shows a weak tendency to stabilization with richness estimators indicating that additional records could be done. Field work carried out at PNSCo has highlighted an unique herpetofauna: five new species were described and there are three candidates as new species. The regional list including Cerrados's units - Estação Ecológica Serra Geral do Tocantins (EESGT) and Estação Ecológica de Uruçuí-Una (EEUU) with Caatinga's ones - PNSCo and Parque Nacional da Serra da Capivara (PNSCa), shows a high herpetofaunal diversity (191 species) to the region. The cluster analysis recovered the Cerrados's units and Caatinga's ones, in separate clusters evidencing a species turnover between domains, despite its geographical proximity. Thus, although there is widespread fauna throughout region shared by the units, each reserve holds its own faunal identity, harboring a singular assemblage of species.
Keywords: *Reptiles, Amphibians, Herpetofaunal Inventory, Herpetofaunistic similarities.*

DAL VECHIO, F., TEIXEIRA JR., M., RECODER, R.S., RODRIGUES, M.T., ZAHER, H. **A herpetofauna do Parque Nacional da Serra das Confusões, Piauí, Brasil, com uma lista regional para uma área ecotonal entre o Cerrado e a Caatinga**. Biota Neotropica. 16(3): e20150105. http://dx.doi.org/10.1590/1676-0611-BN-2015-0105

Resumo: Diferentes fisionomias foram intensivamente amostradas no Parque Nacional da Serra das Confusões (PNSCo) com o objetivo de acessar o padrão de distribuição da sua herpetofauna. Sessenta e seis espécies foram encontradas no parque (47 répteis e 19 anfíbios); a curva de rarefação de lagartos, embora não totalmente estabilizada em uma assíntota, indica que o esforço amostral foi suficiente para revelar a maioria das espécies de lagartos que ocorrem na área; e estimadores de riqueza recuperaram valores perto dos observados. Para os anfíbios, a curva mostra uma fraca tendência para a estabilização com estimadores de riqueza indicando que registros adicionais podem ser feitos. O trabalho de campo realizado no PNSCo destacou uma herpetofauna única: cinco novas espécies foram descritas e há três candidatos como novas espécies. A lista regional, incluindo unidades de Cerrado - Estação Ecológica Serra Geral do Tocantins (EESGT) e Estação Ecológica Uruçuí-Una (EEUU) com aqueles da Caatinga - PNSCo e Parque Nacional da Serra da Capivara (PNSCa), mostra uma alta diversidade da herpetofauna (191 espécies) para a região. A análise de agrupamento recupera as unidades do Cerrado e as da Caatinga em grupos separados, evidenciando uma substituição de espécies entre os domínios, apesar da sua proximidade geográfica. Assim, embora haja fauna ocorrente em toda a região e compartilhada pelas unidades, cada uma das reservas mantém a sua própria identidade, abrigando um conjunto singular de espécies.
Palavras-chave: *Répteis, Anfíbios, Inventário da herpetofauna, Similaridade herpetofaunística.*

Introduction

For many years, our limited knowledge of the herpetofaunal diversity present in the Brazilian Cerrado and Caatinga biomes led us to the erroneous idea these open formation areas were characterized by low endemism rates (Sick 1965, Vanzolini 1963, 1976, 1988). However, extensive field work carried out in last two decades showed the opposite, with elevated local diversity recorded throughout their extension (**Cerrado:** Colli et al. 2002, Rodrigues 1987, Nogueira et al. 2009, Vitt et al. 2005, França & Araújo 2006, Silveira 2006, Recoder & Nogueira 2007, Recoder et al. 2011, Dal Vechio et al. 2013; **Caatinga:** Borges-Nojosa & Cascon 2005, Ribeiro et al. 2008, Loebmann & Haddad 2010, Moura et al. 2011a, Moura et al. 2011b) and exceptional regional diversity along the contact zone between the Cerrado and Caatinga biomes (Rodrigues 1984a,b, 1987, 1988, 1996; Nogueira et al. 2010, Nogueira et al. 2011).

Such efforts also resulted in the description of several new species for the region (**Cerrado:** Colli et al. 2003, Ferrarezzi et al. 2005, Nogueira & Rodrigues 2006, Rodrigues et al. 2007, 2008, Pinna et al. 2010, Teixeira Jr. et al. 2012; Arias et al. 2014; Recoder et al. 2014; **Caatinga:** Rodrigues et al. 2001, Rodrigues et al. 2003b, Bour and Zaher 2005, Nogueira & Rodrigues 2006, Arias et al. 2011a, Arias et al. 2011b, Passos et al. 2011) and the discovery of large populations of species that were considered rare until now (Rodrigues et al. 2013). High diversity in Cerrado faunal assemblages can be explained by local physiognomic heterogeneity, (Eiten 1972, Oliveira-Filho & Ratter 2002, Colli et al. 2002, Nogueira et al. 2009) and historical climatic stability (Werneck et al. 2012). The Caatinga domain, on the other hand, presents more homogeneous drier habitats, and a singular fauna that is associated with these phytoecological conditions (Rodrigues 2003a).

Both Cerrado and Caatinga biomes lost extensive areas of original vegetation, mostly due to fast expansion of agricultural enterprises (Myers et al. 2000, Cavalcanti & Joly 2002, Klink & Machado 2005, Silva et al. 2006). With this continuous process of habitat loss, it seems imperative to prioritize faunal inventories within both biomes in order to fill the geographical gaps in our knowledge, allowing more accurate decisions when defining areas for conservation (Brooks et al. 1992; Greene 1994; Silva & Bates 2002). According to Castro (2000), the state of Piauí harbors an important center of biodiversity, mainly for its highly diversified vegetation that accounts for a large ecotonal zone that includes Cerrado, Dry Forest, Caatinga and pre-Amazon elements.

Here, we present a taxonomic list of the local herpetofauna present at the Parque Nacional da Serra das Confusões (PNSCo), located in southwestern Piauí, one of the least studied areas of the Cerrado-Caatinga ecotone. We also provide a regional list of the herpetofauna that includes three intensively sampled conservation units that are geographically closer to PNSCo, as follows: Parque Nacional da Serra da Capivara (PNSCa), Estação Ecológica de Uruçuí-Una (EEUU) and Estação Ecológica Serra Geral do Tocantins (EESGT). Finally, we compare the herpetofaunal composition of PNSCo with other local lists from the Caatinga and neighboring biomes.

Materials and Methods

1. *Study area*

The Parque Nacional da Serra das Confusões (PNSCo) (approximate coordinates: 08°32' - 09°16'S, 43°15' - 43°51'W, Figures 1, 2) was created in October 1998, and preserves an area of approximately 500.000 ha in southwestern Piauí, close to the border of the state of Bahia. The park is placed in an ecotonal area of Tropical Dry Forest and Caatinga (Olson et al. 2001), with a predominance of deciduous forest and arboreal Caatinga. Climate is considered to be tropical semi-arid. The park is situated within the Parnaíba drainage, in a sandstone plateau with altitudes ranging from about 450 to 700 m above the sea level [a.s.l.]) (Rodrigues et al. 2001; Bour & Zaher 2005). The plateau is locally known as "Chapada da Serra Grande," contains most of the area belonging to the park, and is dissected on its southwestern portion by the intermittent Santana River drainage (Rodrigues et al., 2001; Bour and Zaher, 2005). The top of the plateau, averages 550 m a.s.l., being covered by a tall arboreal caatinga that grows on sandy soil and produces an abundant leaf litter. The plateau is dissected by the intermitent Itaueira River in its central portion and by the "Lagoa do Jacu" depression created by erosive processes in its northern portion. The southern and western borders of the park are deeply dissected, with scattered rocky outcrops over sandy soil originated from the erosion of the Chapada da Serra Grande plateau. Lowland areas are characterized by dense rocky outcrops with low arboreal Caatinga growing over a sandy soil. This vegetation is widespread on the open lowlands. Several canyons, dissected by intermittent rivers with an evergreen forest and a dense leaf litter, are also present, but account for only a minor portion of the area.

2. *Sampling method*

Sampling was carried out in two campaigns in the years 2000 and 2002, one at the end of the dry season (September 26th to October 10th 2000) and other during the rainy season (January 10th to 26th 2002). The main physiognomic subunits present in the region were sampled using pitfall traps with drift fences, complemented by active visual searches. Twelve lines of pitfall traps were installed, each composed of 10 sampling units that consisted of four 30 liters buckets arranged in Y-shape, with a central one connecting to three peripherals by 4 m long plastic fences. Habitat types, geographic coordinates, campaigns and the total sampling effort are summarized in Table 1. The following habitats were sampled:

1. Dry Forest (Lagoa do Jacu) - Line 1. Mesophitic forest, growing on sandy soil, embedded within the river valley. Dominated by arboreal elements, with canopy reaching about 20 m and understory with sparse shrubs. Forest with variable width, reaching 25 m wider sections.
2. High Arboreal Caatinga (Serra Grande Plateau) - Lines 2 to 5. Vegetation on top of the plateau, consisting of arboreal and shrubby elements, characterized by Euphorbiaceae and Leguminosae such as "mimosa" (*Anadenanthera macrocarpa*). Canopy at about 6 m, growing over sandy soil.
3. High Arboreal Caatinga (Serra das Confusões Plateau) - Lines 6 and 7. Predominantly arboreal vegetation growing over sandy/clay soil, covered with leaf litter. Canopy reaching 15 m in height with understory undeveloped.
4. Low Arboreal Caatinga (Lowland) - Lines 8 and 9. Sandstone outcrops, covered by xeromorphic vegetation, dominated by Cactaceae, Bromeliaceae and Velloziaceae.
5. Grooved Forest (Lowland) - Line 10. Evergreen forest along canyon bottoms, with great number of bromeliads, pteridophytes and tall trees, around 30-40 m in height, such as jatobá *(Hymenaea* spp.), forming continuous canopy.

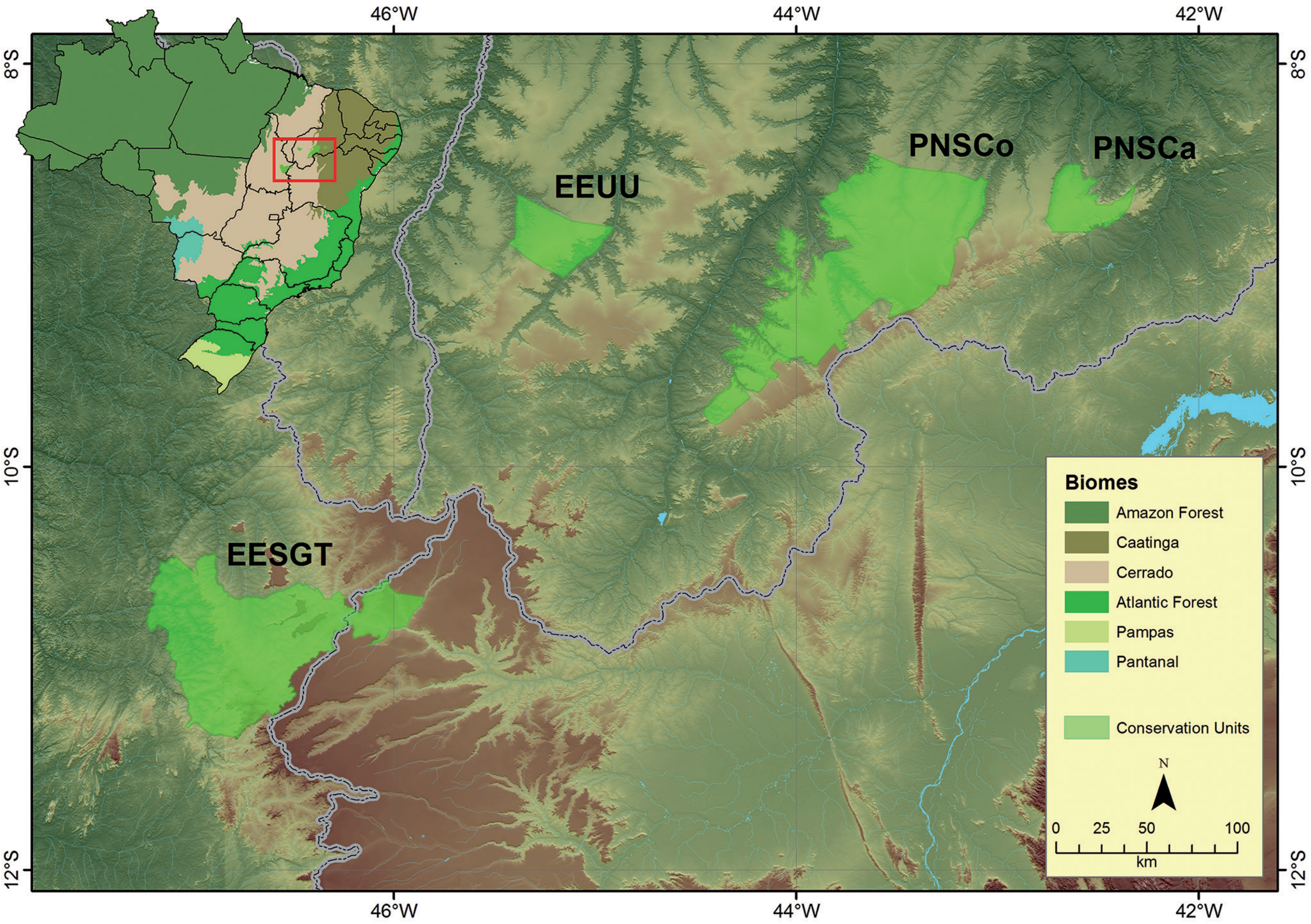

Figure 1. Map of northwestern Brazil showing the conservation units: Estação Ecológica Serra Geral do Tocantins (EESGT), Estação Ecológica de Uruçuí-Una (EEUU) Parque Nacional da Serra das Confusões (PNSCo) and Parque Nacional da Serra da Capivara (PNSCa).

Figure 2. View of plateau with rock outcrop and arboreal caatinga in its base.

It remains constantly in the shade, and accumulates large layers of leaf litter over sandy soil.

6. High arboreal Caatinga (Lowland) - Lines 11 and 12. High arboreal elements, with canopy at 10 m above sandy soil rich in organic matter and closed understory. This type of vegetation predominates in the valleys.

A representative sample of specimens was collected and housed at the herpetological collection of the Museu de Zoologia, Universidade de São Paulo, São Paulo, Brazil (MZUSP). Voucher specimens were fixed in 10% formalin and preserved in 70% alcohol. The remaining individuals were marked by toe clipping and released near the capture site.

1. *Data Analysis*

The effectiveness of the sampling effort was estimated based on rarefaction curves, through 10.000 randomizations without replacement, of a matrix containing the number of individuals per species at each sampling day, including data from both pitfall traps and active search, for lizards and anurans. This analysis was performed using EstimateS v.8.0.0 (Colwell 2006). We also estimated species richness with the Chao2 and Jacknife1 estimators. Snakes were excluded from this analysis because capture rates were low, and the resulting list for the group may be underestimated, which could compromise the behavior of the rarefaction curves.

Initially, we performed faunistic comparisons with herpetofaunal lists available for intensively sampled (long-term sampling) conservation

Table 1. All pitfall lines with its accurate coordinate and the effort spent in each campaign.

Pitfall Lines	Drier Season (1st campaign)	Wet season (2nd campaign)
Line 1 - Dry Forest (Lagoa do Jacu) 08°40'37" S, 43°29'09" W	320	
Line 2 - Toca da Cabocla High Arboreal Caatinga 08°55'28" S, 43°26'58" W	400	
Line 3 - Morrinhos High Arboreal Caatinga 08°57'38" S, 43°26'50" W	360	
Line 4 - Camaçari High Arboreal Caatinga 08°59'10" S, 43°26'45" W		560
Line 5 - Camaçari High Arboreal Caatinga 08°59'10" S, 43°26'45" W		560
Line 6 - Guarita IBAMA 1 High Arboreal Caatinga 09°13'38" S, 43°27'05" W	440	
Line 7 - Guarita IBAMA 2 High Arboreal Caatinga 08°59'10" S, 43°26'45" W		560
Line 8 - Olho d'água Santa Low Arboreal Caatinga (Low Land) 09°13'03" S, 43°29'26" W	520	600
Line 9 - Baixão do Fausto Low Arboreal Caatinga (Low Land) 09°09'53" S, 43°33'58" W		440
Line 10 - Grooved Forest (Low Land) 09°13'10" S, 43°29'27" W	400	
Line 11 - Pinga Velho 1 High arboreal Caatinga (Low Land) 09°03'46" S, 43°44'27" W		80
Line 12 - Pinga Velho 2 High arboreal Caatinga (Low Land) 09°03'42" S, 43°44'23" W		480
Sampling effort	2440 buckets. days	3280 buckets. days

units geographically closer to PNSCo: Estação Ecológica Serra Geral do Tocantins (EESGT) (Recoder et al. 2011, Valdujo et al. 2011), Estação Ecológica de Uruçuí-Una (EEUU) (Dal Vechio et al. 2013), and Parque Nacional da Serra da Capivara (PNSCa) (Cavalcanti et al. 2014). An additional motivation in using these listings is the similar latitude in which these areas are located, forming a Cerrado-Caatinga transect. EESGT and EEUU are mostly within the Cerrado (with EEUU having some Caatinga's elements) while PNSCo and PNSCa are mostly within a Caatinga physiognomy (with PNSCo including Cerrado elements).

A dendrogram was estimated using the UPGMA clustering algorithm and Jaccard's similarity index with the program MVSP v.3.1 (Kovach 2000). Subsequently, we performed a cluster analysis using the Jaccard's coefficient, considering only the presence and absence of species and using the following local lists from different Brazilian domains: **Caatinga**: Planalto da Ibiapaba, Ceará (Loebmann & Haddad 2010), Serra das Almas, Ceará (Borges-Nojosa & Cascon 2002), Parque Nacional do Catimbau, Pernambuco (Moura et al. 2011a, b; Pedrosa et al. 2014), Ouricuri, Pernambuco (Moura et al. 2011a, b), Fazenda Saco, Pernambuco (Moura et al. 2011a, b); **Cerrado**: UHE Espora, Goiás (Vaz-Silva 2007), Parque Nacional das Emas-PNE, Goiás (Nogueira et al. 2009; Valdujo et al. 2009; Kopp et al. 2010), Northern Tocantins River basin-BTN, Tocantins and Maranhão (*i.e.* municipalities of Estreito, Babaçulândia, Carolina and Palmeirante) (Pavan 2007), Southern Tocantins River basin-BTS, Tocantins and Goiás (*i.e.* municipalities of Peixe, São Salvador do Tocantins, Paranã and Minaçu) (Pavan 2007); **Atlantic Forest:** Mata do Buraquinho, Paraíba (Santana et al. 2008), Estação Ecológica do Tapacurá-EET, Pernambuco (Moura et al. 2011a, b); **Amazon:** Espigão do Oeste, Rondônia (Bernarde & Abe 2006; Bernarde 2007; Macedo et al. 2008), Carajás, Pará (Cunha et al. 1985; Pinheiro 2010), Cacoal, Rondônia (Turci & Bernarde 2008) Reserva Ducke, Amazonas (Lima et al. 2006; Vitt et al. 2008; Fraga et al. 2014).

Results

Sixty six species of reptiles and amphibians were found at PNSCo: 47 reptiles (21 lizards, 20 Snakes, 2 turtles and 4 amphisbaenids) and 19 amphibians (18 anurans and one caecilian) (Table 2, Figures 3, 4, 5).

Table 2. List of the herpetofauna recored at Parque Nacional Serra da Confusões (PNSCo). N: number the specimens registered; DF: Dry Forest; HAC-SGP: High Arboreal Caatinga-Serra Grande plateau; HAC-SCP: High Arboreal Caatinga-Serra das Confusões plateau; LAC: Low Arboreal Caatinga; GF: Groove Forest; HAC-LL: High Arboreal Caatinga-Lowland; "?" without information. * recorded in a recent field trip.

	N	Habitat of capture
REPTILIA		
SQUAMATA		
LIZARDS		
HOPLOCERCIDAE		
Hoplocercus spinosus Fitzinger, 1843	18	HAC-SCP, LAC, HAC-LL
IGUANIDAE		
Iguana iguana (Linnaeus, 1758)	2	?
TROPIDURIDAE		
Stenocercus squarrosus Nogueira & Rodrigues, 2006	13	HAC-SGP, HAC-LL
Tropidurus hispidus (Spix, 1825)	84	HAC-SGP, HAC-SCP, LAC, H AC-LL
Tropidurus semitaeniatus (Spix, 1825)	32	HAC-SGP
LEIOSAURIDAE		
Enyalius bibronii Boulenger, 1885	18	HAC-SGP, HAC-SCP, LAC, HAC-LL
PHYLLODACTYLIDAE		
Phyllopezus pollicaris (Spix, 1825)	12	HAC-SGP, LAC, GF
SPHAERODACTYLIDAE		
Coleodactylus brachystoma (Amaral, 1935)	1	LAC
GEKKONIDAE		
Hemidactylus brasilianus (Amaral, 1935)	16	DF, HAC-SGP, HAC-SCP, LAC, HAC-LL
SCINCIDAE		
Mabuya heathi (Schmidt & Inger, 1951)	11	HAC-SGP, HAC-SCP

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Continued Table 2.

	N	Habitat of capture
Mabuya nigropunctata (Spix, 1825)	17	HAC-SGP, HAC-SCP, LAC, HAC-LL
Mabuya frenata (Cope, 1862)	10	DF, HAC-SGP, HAC-SCP
GYMNOPHTHALMIDAE		
Calyptommatus confusionibus Rodrigues, Zaher & Curcio, 2001	14	HAC-SGP, HAC-LL
Colobosaura modesta (Reinhardt & Lütken, 1862)	45	HAC-SGP, HAC-SCP, LAC, HAC-LL
Micrablepharus maximiliani (Reinhardt & Lütken, 1862)	32	HAC-SGP, HAC-SCP, LAC, HAC-LL
Procellosaurinus erythrocercus Rodrigues, 1991	16	HAC-SGP, HAC-LL
TEIIDAE		
Ameiva ameiva (Linnaeus, 1758)	29	DF, HAC-SGP, HAC-SCP, LAC, HAC-LL
Ameivula confusioniba (Arias, De Carvalho, Rodrigues & Zaher, 2011)		
Ameivula sp.		HAC-SGP, HAC-SCP, LAC, HAC-LL
Glaucomastix venetacauda (Arias, De Carvalho, Rodrigues & Zaher, 2011)	23	HAC-SGP, LAC
Salvator merianae (Duméril & Bibron, 1839)	2	LAC
AMPHISBAENA		
AMPHISBAENIDAE		
Amphisbaena aff. *meringoera**	1	?
Amphisbaena frontalis Vanzolini, 1991	5	HAC-SGP, LAC
Amphisbaena polystega (Duméril, 1851)	1	?
Amphisbaena vermicularis Wagler, 1824	2	?
SNAKES		
LEPTOTYPHLOPIDAE		
Trilepida cf. *fuliginosa*	5	HAC-SGP, HAC-SCP, HAC-LL
BOIDAE		
Corallus hortulanus (Linnaeus, 1758)	3	DF, HAC-SCP
Epicrates assisi Machado, 1945	1	?
COLUBRIDAE		
Drymarchon corais (Boie, 1827)	3	LAC, GF
Drymoluber brazili (Gomes, 1918)	2	GF
Leptophis ahaetulla (Linnaeus, 1758)	1	?
Oxybelis aeneus (Wagler, 1824)	1	?
Spilotes pullatus (Linnaeus, 1758)	3	HAC-SGP, LAC, GF
Tantilla melanocephala (Linnaeus, 1758)	2	DF
DIPSADIDAE		
Apostolepis cearensis Gomes, 1915	10	HAC-SGP
Erythrolamprus miliaris (Linnaeus, 1758)	1	?
Erythrolamprus viridis Günther, 1862	1	?
Oxyrhopus trigeminus Duméril, Bibron & Duméril, 1854	3	?
Philodryas nattereri Steindachner, 1870	1	HAC-SCP

Continued Table 2.

	N	Habitat of capture
Philodryas olfersii (Lichtenstein, 1823)	1	DF
Pseudoboa nigra (Duméril, Bibron & Duméril, 1854)	5	HAC-SGP
Rodriguesophis iglesiasi (Gomes, 1915)	1	?
Xenodon merremii (Wagler, 1824)	1	?
Xenodon nattereri (Steindachner, 1867)	1	?
VIPERIDAE		
Crotalus durissus (Linnaeus, 1758)	1	?
ARCHOSAURIA		
ANAPSIDA		
TESTUDINES		
CHELIDAE		
Mesoclemmys perplexa Bour & Zaher, 2005	3	LAC, GF
Mesoclemmys tuberculata (Luederwaldt, 1926)	6	GF
AMPHIBIA		
ANURAN		
HYLIDAE		
Corythomantis greeningi Boulenger, 1896	11	LAC
Dendropsophus soaresi (Caramaschi and Jim, 1983)	1	?
Phyllomedusa nordestina Caramaschi, 2006	11	LAC
Scinax gr. *ruber*	7	LAC
Scinax sp.	1	?
LEPTODACTYLIDAE		
Adenomera sp. n.	25	LAC, GF
Leptodactylus aff. *syphax*	10	LAC
Leptodactylus fuscus (Schneider, 1799)	2	LAC
Leptodactylus macrosternum Miranda-Ribeiro, 1926	3	?
Leptodactylus mystaceus (Spix, 1824)	9	LAC
Leptodactylus troglodytes Lutz, 1926	32	HAC-SGP, LAC, HAC-LL
Leptodactylus vastus Lutz, 1930	5	LAC, GF
Physalaemus albifrons (Spix, 1824)	6	LAC
Physalaemus cuvieri Fitzinger, 1826	59	HAC-SGP, LAC, GF
BUFONIDAE		
Rhinella granulosa (Spix, 1824)	18	LAC, GF
Rhinella jimi (Stevaux, 2002)	4	GF
ODONTOPHRYNIDAE		
Proceratophrys cristiceps (Müller, 1883)	19	HAC-SGP, LAC, GF
MICROHYLIDAE		
Dermatonotus muelleri (Boettger, 1885)	20	HAC-SGP, LAC
GYMNOPHIONA		
CAECILIIDAE		
Siphonops paulensis Boettger, 1892	2	HAC-SGP, LAC

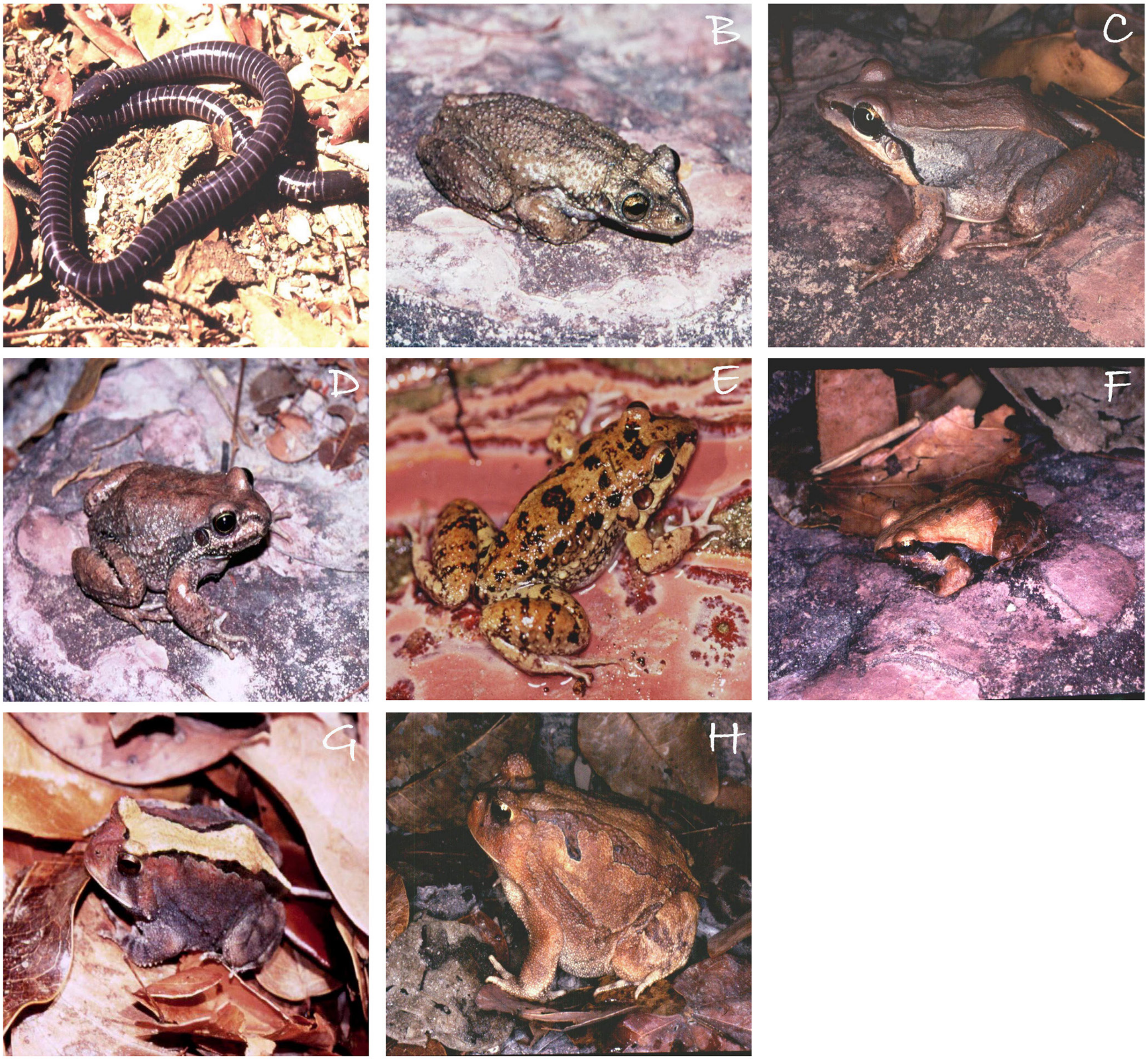

Figure 3. Some of the amphibians sampled at PNSCoe. A - *Siphonops paulensis*; B - *Corythomantis greeningi*; C - *Leptodactylus mystaceus*; D - *Leptodactylus* aff. *syphax*; E - *Leptodactylus troglodytes*; F - *Physalaemus albifrons*; G - *Proceratophrys cristiceps* (male) and H - *Proceratophrys cristiceps* (female).

For lizards, the rarefaction curve showed a tendency to asymptote, with decreasing standard deviation after 34 sampling days (two campaigns together) (Figure 6A), same pattern found in each campaign separately (Figure 6B). The richness estimators Chao2 and Jacknife1 recovered 19 and 20 species, respectively. For amphibians, the rarefaction curve for the two campaigns together showed only a weak tendency towards stabilization (Figure 7A), differently from the results attained for each campaign separately (Figure 7B). The richness estimators Chao2 and Jacknife1 recovered 17 and 18 species, respectively.

The complex PNSCo-PNSCa (Caatinga's units) harbors at least 74 species, and share 30 species, as follows: 52 reptiles (24 lizards, 4 amphisbaenas, 22 snakes and 2 turtles) and 22 amphibians (20 anurans and two caecilians). Adding the closer EEUU and EESGT Cerrado's units, the number of species increases to 191 (about 12.5% of Brazilian species): 130 reptiles (45 lizards, 10 amphisbaenas, 69 snakes, 4 turtles and 2 crocodilians) and 61 amphibians (59 anurans and two caecilians) (Table 3).

Sixteen species occured in all four units while 17 species were shared among three of them, 32 species were shared only between EEUU and EESGT, 8 species between PNSCo and PNSCa, 5 species between PNSCo and EEUU, and 4 between PNSCo and EESGT. Only one species was shared between PNSCa and EEUU and none were exclusive of PNSCa and EESGT together. Seven species occurred exclusively in PNSCa, 17 in PNSCo, 20 in EEUU, and 64 in EESGT (Table 3).

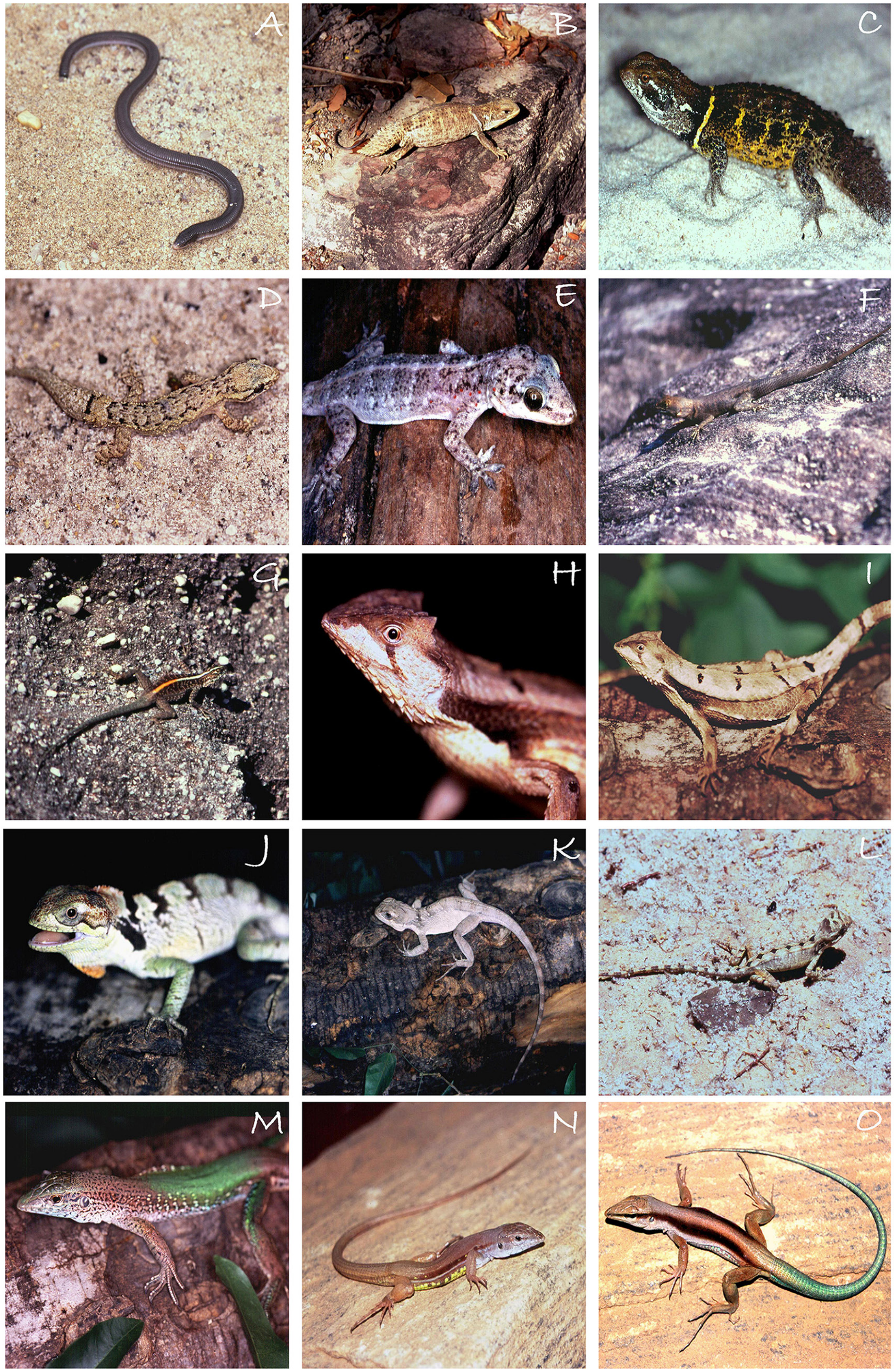

Figure 4. Some of the reptile sampled at PNSCo. A - *Amphisbaena vermicularis*; B - *Hoplocercus spinosus* (female); C - *Hoplocercus spinosus* (male); D - *Hemidactylus brasilianus*; E - *Phyllopezus pollicaris*; F - *Tropidurus hispidus*; G - *Tropidurus semitaeniatus*; H and I - *Stenocercus squarrosus*; J - *Enyalius bibronii* (male); K - *Enyalius bibronii* (female); L - *Enyalius bibronii* (juvenile); M - *Ameiva ameiva*; N - *Ameivula confusioniba*; O - *Glaucomastix venetacauda*.

Figure 5. Some of the reptile sampled at PNSCo. A - *Colobosaura modesta*; B - *Procellosaurinus erythrocercus*; C - *Boa constrictor*; D - *Corallus hortulanus*; E - *Epicrates assisi*; F - *Drymarchon corais*; G - *Drymoluber brazili*; H - *Apostolepis cearensis*; I - *Erythrolamprus miliaris*; J - *Oxyrhopus trigeminus*; K - *Philodryas nattereri*; L - *Pseudoboa nigra*.

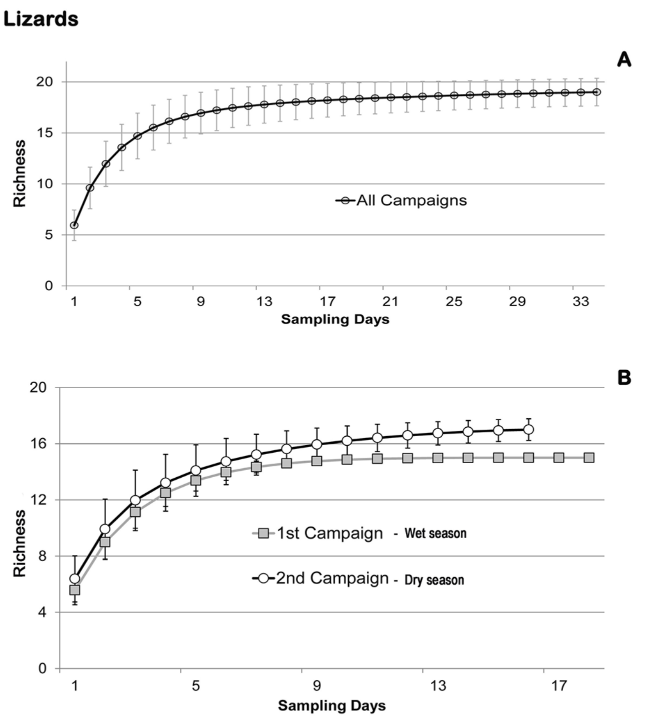

Figure 6. Rarefaction curve for lizards. A - after 34 sampling days (two campaign together); B - for each campaign separately.

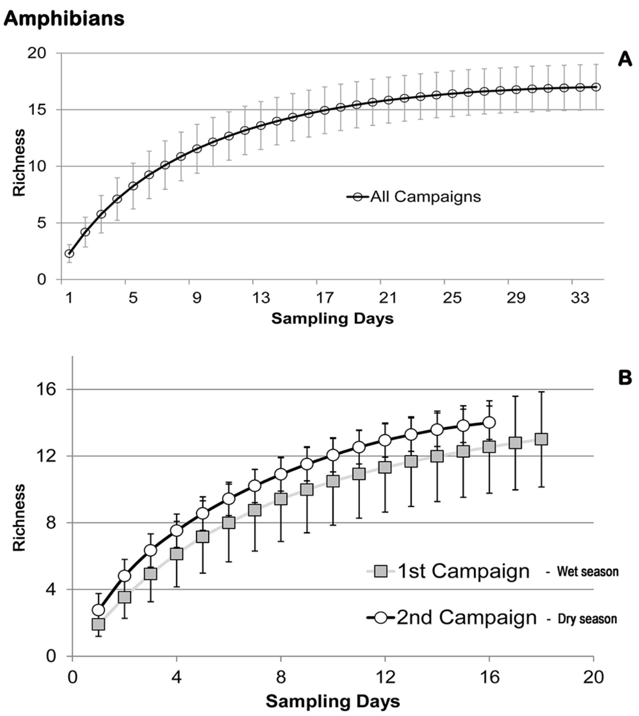

Figure 7. Rarefaction curve for amphibians. A - after 34 sampling days (two campaign together); B - for each campaign separately.

Table 3. Regional Herpetofaunal list, with presence "X" or "-" absence of species for each conservation unit. PNSCo: Parque Nacional da Serra das Confusões (present work); EEUU: Estação Ecológica de Uruçuí-Una (Dal Vechio et al. 2013); EESGT: Estação Ecológica Serra Geral do Tocantins (Recoder et al. 2011, Valdujo et al. 2011); PNSCa: Parque Nacional da Serra da Capivara (Cavalcanti et al. 2014).

	PNSCa (Caatinga)	PNSCo (Caatinga with Cerrado elements)	EEUU (Cerrado with Caatinga elements)	EESGT (Cerrado)
REPTILIA				
SQUAMATA				
LAGARTOS				
HOPLOCERCIDAE				
Hoplocercus spinosus Fitzinger, 1843	-	X	X	X
IGUANIDAE				
Iguana iguana (Linnaeus, 1758)	X	X	X	X
TROPIDURIDAE				
Stenocercus squarrosus Nogueira & Rodrigues, 2006	X	X	-	-
Stenocercus quinarius Nogueira & Rodrigues, 2006	-	-	-	X
Tropidurus cf. *oreadicus*	-	-	-	X
Tropidurus helenae (Manzani & Abe, 1990)	X	-	-	-
Tropidurus oreadicus Rodrigues, 1987	-	-	X	-
Tropidurus semitaeniatus (Spix, 1825)	X	X	X	-
Tropidurus hispidus (Spix, 1825)	X	X	-	-
LEIOSAURIDAE				
Enyalius bibronii Boulenger, 1885	X	X	-	-
POLYCHROTIDAE				
Norops chrysolepis Duméril & Bibron, 1837	-	-	-	X
Norops brasiliensis Vanzolini & Williams 1970	-	-	X	-
Norops meridionalis Boettger, 1885	-	-	X	-

Continued Table 3.

	PNSCa (Caatinga)	PNSCo (Caatinga with Cerrado elements)	EEUU (Cerrado with Caatinga elements)	EESGT (Cerrado)
Polychrus acutirostris Spix, 1825	-	-	X	X
PHYLLODACTYLIDAE				
Gymnodactylus amarali Barbour, 1925	-	-	-	X
Phyllopezus pollicaris (Spix, 1825)	X	X	X	X
SPHAERODACTYLIDAE				
Coleodactylus brachystoma (Amaral, 1935)	-	X	X	X
Coleodactylus cf. *meridionalis*	-	-	-	X
GEKKONIDAE				
Hemidactylus brasilianus (Amaral, 1935)	X	X	X	X
Hemidactylus mabouia (Moreau de Jonnès, 1818)	-	-	X	X
Lygodactylus klugei (Smith, Martin & Swain, 1977)	X	-	-	-
SCINCIDAE				
Mabuya cf. *heathi*	-	-	-	X
Mabuya heathi (Schmidt & Inger, 1951)	-	X	-	-
Mabuya cf. *nigropunctata*	-	-	-	X
Mabuya nigropunctata (Spix, 1825)	X	X	X	-
Mabuya frenata (Cope, 1862)	-	X	-	X
GYMNOPHTHALMIDAE				
Bachia oxyrhina Rodrigues et al., 2008	-	-	-	X
Calyptommatus confusionibus Rodrigues, Zaher & Curcio, 2001	-	X	-	-
Cercosaura ocellata Wagler, 1830	-	-	-	X
Colobosaura modesta (Reinhardt & Lütken, 1862)	X	X	X	X
Micrablepharus maximiliani (Reinhardt & Lütken, 1862)	X	X	X	X
Procellosaurinus erythrocercus Rodrigues, 1991	X	X	-	-
Vanzosaura savanicola Recoder, Werneck, Teixeira Jr., Colli, Sites & Rodrigues, 2014	-	-	-	X
TEIIDAE				
Ameiva ameiva (Linnaeus, 1758)	X	X	X	X
Ameivula confusioniba (Arias, De Carvalho, Rodrigues & Zaher, 2011)	-	X	-	-
Ameivula cf. *mumbuca*	-	-	X	-
Ameivula jalapensis (Colli et al., 2009)	-	-	-	X
Ameivula mumbuca (Colli et al., 2003)	-	-	-	X
Ameivula ocellifera (Spix, 1825)	X	-	-	-
Ameivula sp.	-	X	-	-
Glaucomastix venetacauda (Arias, De Carvalho, Rodrigues & Zaher, 2011)	X	X	-	-
Kentropyx aff. *paulensis* Boettger, 1893	-	-	-	X
Kentropyx calcarata Spix, 1825	-	-	X	-
Salvator duseni (Lönnberg, 1910)	-	-	-	X
Salvator merianae (Duméril & Bibron, 1839)	X	X	X	X
Tupinambis quadrilineatus Manzani & Abe, 1997	-	-	X	X
AMPHISBAENAS				
AMPHISBAENIDAE				
Amphisbaena acrobeles (Ribeiro, Castro-Mello & Nogueira, 2009)	-	-	-	X
Amphisbaena aff. *miringoera**	X	X	X	X

Continued Table 3.

	PNSCa (Caatinga)	**PNSCo (Caatinga with Cerrado elements)**	**EEUU (Cerrado with Caatinga elements)**	**EESGT (Cerrado)**
Amphisbaena alba Linnaeus, 1758	-	-	X	X
Amphisbaena cf. *ibijara**	X	-	-	-
Amphisbaena frontalis Vanzolini, 1991	-	X	-	-
Amphisbaena kraoh (Vanzolini, 1971)	-	-	-	X
Amphisbaena sp. nov.	-	-	-	X
Amphisbaena polystega (Duméril, 1851)	-	X	X	X
Amphisbaena sp.*	X	-	-	-
Amphisbaena vermicularis Wagler, 1824	-	X	X	X
SERPENTES				
LEPTOTYPHLOPIDAE				
Trilepida cf. *fuliginosa*	-	X	-	-
Tricheilostoma brasiliensis (Laurent, 1949)	-	-	X	-
Siagonodon acutirostris Pinto & Curcio, 2011	-	-	-	X
TYPHLOPIDAE	-			
Typhlops brongersmianus Vanzolini, 1976	-	-	X	X
BOIDAE				
Boa constrictor Linnaeus, 1758	-	-	X	X
Corallus hortulanus (Linnaeus, 1758)	X	X	X	X
Epicrates assisi Machado, 1945	-	X	X	-
Epicrates crassus Cope, 1862	-	-	-	X
Eunectes murinus (Linnaeus, 1758)	-	-	X	X
COLUBRIDAE	-			
Chironius exoletus (Linnaeus, 1758)	-	-	X	-
Chironius flavolineatus (Boettger, 1885)	-	-	X	X
Chironius quadricarinatus (Boie, 1827)	-	-	-	X
Drymarchon corais (Boie, 1827)	-	X	-	X
Drymoluber brazili (Gomes, 1918)	-	X	-	-
Leptophis ahaetulla (Linnaeus, 1758)	-	X	X	-
Mastigodryas bifossatus (Raddi, 1820)	-	-	X	X
Mastigodryas boddaerti (Sentzen, 1796)	-	-	X	X
Oxybelis aeneus (Wagler, 1824)	X	X	-	X
Spilotes pullatus (Linnaeus, 1758)	X	X	X	-
Tantilla melanocephala (Linnaeus, 1758)	-	X	X	X
DIPSADIDAE				
Apostolepis ammodites Ferrarezzi, Barbo & Albuquerque, 2005	-	-	-	X
Apostolepis cf. *longicaudata* Amaral, 1921	-	-	-	X
Apostolepis cearensis Gomes, 1915	-	X	X	-
Apostolepis nelsonjorgei Lema & Renner 2004	-	-	-	X
Apostolepis polylepis Amaral, 1921	-	-	X	X
Atractus pantostictus Fernandes & Puorto, 1993	-	-	-	X
Boiruna cf. *sertaneja*	-	-	-	X
Clelia plumbea (Wied, 1820)	-	-	X	X
Erythrolamprus almadensis (Wagler, 1824)	-	-	X	X
Erythrolamprus cf. *frenatus*	-	-	-	X
Erythrolamprus maryellenae (Dixon, 1985)	-	-	-	X

Continued Table 3.

	PNSCa (Caatinga)	PNSCo (Caatinga with Cerrado elements)	EEUU (Cerrado with Caatinga elements)	EESGT (Cerrado)
Erythrolamprus poecilogyrus (Wied, 1825)	-	-	X	-
Erythrolamprus miliaris (Linnaeus, 1758)	-	X	-	-
Erythrolamprus reginae (Linnaeus, 1758)	-	-	X	X
Erythrolamprus taeniogaster (Jan, 1863)	-	-	X	-
Erythrolamprus viridis (Günther, 1862)	-	X	-	-
Helicops angulatus (Linnaeus, 1758)	-	-	X	X
Helicops leopardinus (Schlegel, 1837)	-	-	-	X
Hydrodynastes gigas (Duméril, Bibron & Duméril, 1854)	-	-	-	X
Hydrops triangularis (Wagler, 1824)	-	-	X	-
Leptodeira annulata (Linnaeus, 1758)	-	-	-	X
Lygophis meridionalis (Schenkel, 1902)	-	-	-	X
Lygophis paucidens Hoge, 1953	-	-	X	X
Oxyrhopus guibei Hoge & Romano, 1977	-	-	-	X
Oxyrhopus rhombifer Duméril, Bibron & Duméril, 1854	-	-	X	X
Oxyrhopus trigeminus Duméril, Bibron & Duméril, 1854	X	X	X	X
Phalotris labiomaculatus Lema, 2002	-	-	-	X
Philodryas nattereri Steindachner, 1870	X	X	X	X
Philodryas olfersii (Lichtenstein, 1823)	X	X	X	-
Philodryas patagoniensis Girard, 1858	-	-	-	X
Phimophis guerini (Duméril, Bibron & Duméril, 1854)	-	-	-	X
Pseudoboa nigra (Duméril, Bibron & Duméril, 1854)	X	X	X	X
Psomophis joberti (Sauvage, 1884)	-	-	X	X
Rodriguesophis iglesiasi (Gomes, 1915)	X	X	X	X
Sibynomorphus mikanii (Schlegel, 1837)	-	-	X	X
Taeniophallus occipitalis (Jan, 1863)	-	-	X	X
Thamnodynastes sp.	X	-	X	-
Thamnodynastes hypoconia (Cope, 1860)	-	-	-	X
Thamnodynastes cf. *pallidus*	-	-	-	X
Xenodon merremii (Wagler, 1824)	X	X	X	X
Xenodon nattereri (Steindachner, 1867)	-	X	X	-
ELAPIDAE				
Micrurus brasiliensis Roze, 1967	-	-	-	X
Micrurus ibiboboca (Merrem, 1820)	-	-	X	-
VIPERIDAE	-			
Bothrops lutzi (Miranda-Ribeiro, 1915)	X	-	X	X
Bothrops mattogrossensis Amaral, 1925	-	-	-	X
Bothrops neuwiedi Wagler, 1824	-	-	-	X
Bothrops pauloensis Amaral, 1925	-	-	-	X
Bothrops moojeni Hoge, 1966	-	-	X	X
Crotalus durissus (Linnaeus, 1758)	-	X	X	X
ARCHOSAURIA				
ANAPSIDA				
TESTUDINES				
CHELIDAE				
Mesoclemmys perplexa Bour & Zaher, 2005	-	X	-	-

Continued Table 3.

	PNSCa (Caatinga)	PNSCo (Caatinga with Cerrado elements)	EEUU (Cerrado with Caatinga elements)	EESGT (Cerrado)
Mesoclemmys tuberculata (luederwaldt, 1926)	X	X	-	-
Phrynops geoffroanus (Schweigger, 1812)	-	-	-	X
Phrynops cf. *tuberosus*	-	-	X	-
CROCODYLIA				
ALLIGATORIDAE				
Caiman crocodilus (Linnaeus, 1758)	-	-	X	X
Paleosuchus palpebrosus (Cuvier, 1807) 124spp	-	-	-	X
AMPHIBIA				
ANURA				
CRAUGASTORIDAE				
Barycholos ternetzi Caramaschi & Pombal, 2001	-	-	-	X
HYLIDAE				
Corythomantis greeningi Boulenger, 1896	-	X	-	X
Dendropsophus cruzi (Pombal & Bastos, 1998)	-	-	-	X
Dendropsophus minutus (Peters, 1872)	-	-	X	X
Dendropsophus nanus (Boulenger, 1889)	-	-	X	-
Dendropsophus rubicundulus (Reinhardt and Lütken, 1862)	-	-	X	X
Dendropsophus soaresi (Caramaschi and Jim, 1983)	-	X	X	X
Hypsiboas albopunctatus (Spix, 1824)	-	-	-	X
Hypsiboas punctatus (Schneider, 1799)	-	-	-	X
Hypsiboas raniceps Cope, 1862	-	-	-	X
Hypsiboas multifasciatus (Günther, 1859)	-	-	X	-
Osteocephalus cf. *taurinus*	-	-	-	X
Osteocephalus taurinus Steindachner, 1862	-	-	X	-
Phyllomedusa azurea Cope, 1862	-	-	X	X
Phyllomedusa nordestina Caramaschi, 2006	-	X	-	-
Scinax constrictus (lima, Bastos & Giaretta, 2005)	-	-	-	X
Scinax fuscomarginatus (Lutz, 1925)	-	-	X	X
Scinax fuscovarius Lutz, 1925)	-	-	-	X
Scinax gr. *ruber* sp.1	-	-	-	X
Scinax gr. *ruber* sp.2	-	-	X	-
Scinax gr. *ruber* sp.3	-	-	X	-
Scinax gr. *ruber* sp.4	-	X	-	-
Scinax sp.	-	X	-	-
Scinax x-signatus (Spix, 1824)	X	-	-	-
Trachycephalus venulosus (Laurenti, 1768)	-	-	X	X
LEPTODACTYLIDAE				
Adenomera sp. n.	-	X	X	-
Leptodactylus fuscus (Schneider, 1799)	-	X	X	X
Leptodactylus aff. *syphax*	X	X	-	-
Leptodactylus hylaedactylus Müller, 1923	-	-	-	X
Leptodactylus labyrinthicus (Spix, 1824)	-	-	-	X
Leptodactylus latrans (Steffen, 1815)	-	-	-	X
Leptodactylus macrosternum Miranda-Ribeiro, 1926	-	X	-	-

Continued Table 3.

	PNSCa (Caatinga)	**PNSCo (Caatinga with Cerrado elements)**	**EEUU (Cerrado with Caatinga elements)**	**EESGT (Cerrado)**
Leptodactylus martinezi Bokermann, 1956	-	-	-	X
Leptodactylus mystaceus (Spix, 1824)	-	X	-	-
Leptodactylus podicipinus (Cope, 1862)	-	-	X	X
Leptodactylus sertanejo (Giaretta & Costa, 2007)	-	-	-	X
Leptodactylus syphax Bokermann, 1969	-	-	-	X
Leptodactylus troglodytes Lutz, 1926	X	X	X	X
Leptodactylus vastus Lutz, 1930	X	X	X	-
Eupemphix nattereri (Steindachner, 1863)	-	-	-	X
Physalaemus albifrons (Spix, 1824)	-	X	-	-
Physalaemus centralis Bokermann, 1962	-	-	X	X
Physalaemus cuvieri Fitzinger, 1826	X	X	X	X
Pleurodema diplolistris (Peters, 1870)	-	-	-	X
Pseudopaludicola cf. *mystacalis*	-	-	X	-
Pseudopaludicola mystacalis (Cope, 1887)	-	-	-	X
Pseudopaludicola saltica (Cope, 1887)	-	-	-	X
BUFONIDAE				
Rhaebo guttatus (Schneider, 1799)	-	-	X	X
Rhinella granulosa (Spix, 1824)	X	X	-	-
Rhinella jimi (Stevaux, 2002)	X	X	X	-
Rhinella mirandaribeiroi (Gallardo, 1965)	-	-	X	X
Rhinella ocellata (Günther, 1858)	-	-	X	X
Rhinella schneideri (Werner, 1894)	-	-	-	X
Rhinella veredas (Brandão, Maciel & Sebben, 2007)	-	-	X	-
ODONTOPHRYNIDAE				
Proceratophrys cristiceps (Müller, 1883)	-	X	-	-
Proceratophrys goyana (Miranda-Ribeiro, 1937)	-	-	-	X
MICROHYLIDAE				
Dermatonotus muelleri (Boettger, 1885)	-	X	X	X
Elachistocleis carvalhoi Caramaschi, 2010	-	-	X	-
Elachistocleis cesarii (Miranda-Ribeiro, 1920)	-	-	-	X
GYMNOPHIONA				
CAECILIIDAE				
Siphonops sp.	X	-	-	-
Siphonops paulensis Boettger, 1892	-	X	-	X

The cluster analysis recovered PNSCo and PNSCa within the Caatinga group, and EEUU and EESGT within the Cerrado group, indicating a strong species turnover in the contact region between Caatinga and Cerrado in southern Piauí (Figure 8). The same pattern was recovered for lizards and amphibians when analyzed separately (Data not shown).

Discussion

The herpetofauna of PNSCo is one of the most diverse of the Caatinga biome, except for the fauna from the isolated forest patches of northeastern Brazil, usually referred as "Brejos Nordestinos". A total of 66 species were recorded (74 species when taking together PNSCo and PNSCa), equating in diversity with previously well sampled areas like Exu in Pernambuco, Valença in Piauí, and Xingó in Alagoas (with 53, 42 and 41 species, respectively) (Rodrigues, 2003a).

Part of the high diversity found in the PNSCo and PNSCa taken together can be explained by landscape heterogeneity, which, in turn, also characterizes the Cerrado units and strongly differs from Caatinga (*sensu stricto*) areas where the landscape physiognomy is far more homogeneus. However, since the PNSCo and PNSCa are situated in an ecotonal area between these two biomes, they harbor a mix of

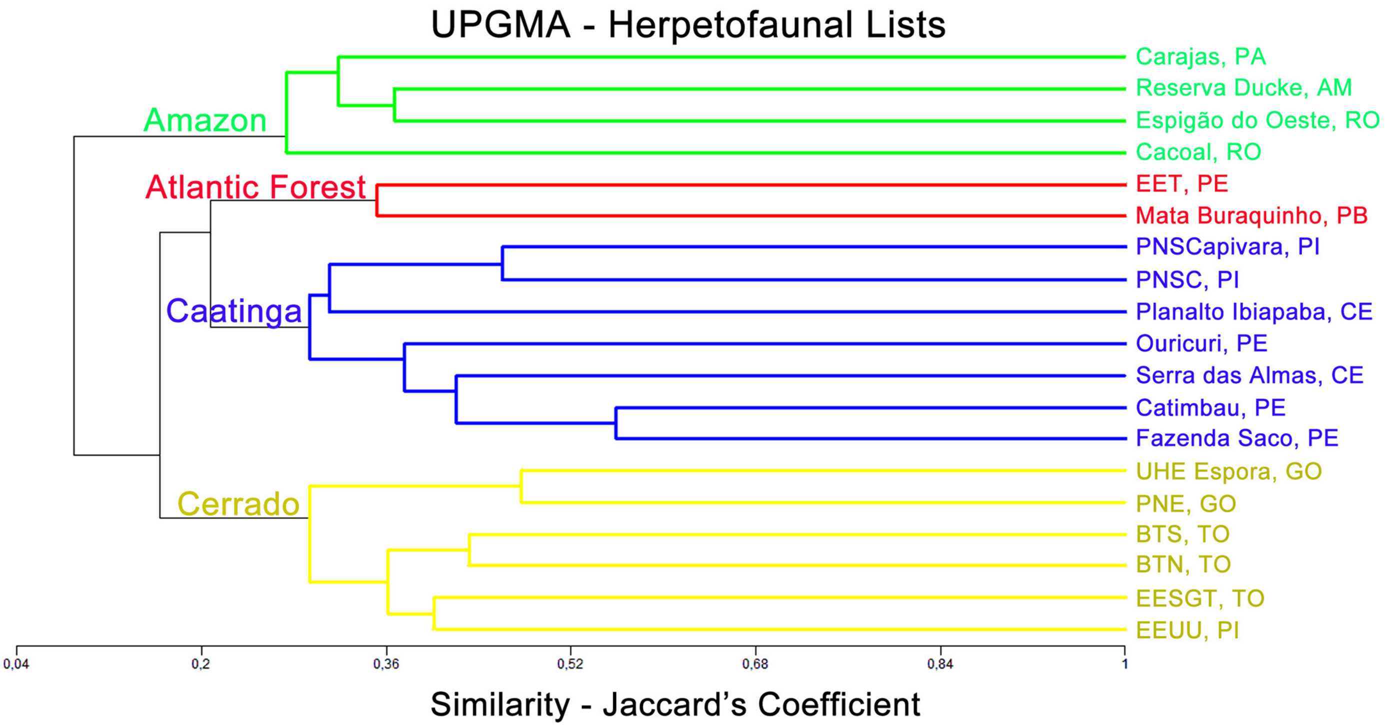

Figure 8. UPGMA of herpetofaunal lists from different Brazilian biomes. Green: Amazon; Red: northwest Atlantic Forest; Blue: Caatinga and Yellow: Cerrado.

Caatinga and Cerrado elements, forming an heterogeneous landscape. This results in high species diversity, with species typical from Cerrado (*e.g. Hoplocercus spinosus, Coleodactylus brachystoma, Colobosaura modesta*) and Caatinga *(e.g. Epicrates assisi, Dermatonotus mulleri, Physalaemus albifrons, Corythomantis greeningi, Rhinella jimi, Proceratophrys cristiceps*) coexisting in the area. Adding to phyto-geomorphological features, historical events also could be acting to increase the local diversity in the southwestern Piauí, as the historical contacts now lost between sandy soils from this region and those from the São Francisco dunes (Rodrigues et al. 2001).

The observed richness of lizards and amphibians might be close to the real diversity in PNSCo since species rarefaction curves for both groups tend toward an asymptote after 34 sampling days (stronger for lizards) and richness estimators recover close values to those obtained. However, some new species records are expected in the future since arboreal, aquatic, fossorial species and ones with explosive reproduction are always more difficult to sample and tend to be underestimated even in long-term and/or large-scale inventories.

Indeed, the most diverse lists of local herpetofauna are those based on intensive field work with long periods of sampling along different climatic seasons and/or mega infrastructure projects with extensive devastation of natural habitats, such as hydroelectric power plants (Pavan & Dixo 2004, Vaz-Silva et al. 2007, Silva Junior et al. 2005, Loebmann & Haddad 2010, Nogueira et al. 2010, Silveira et al. 2010, Recoder et al. 2011, Valdujo et al. 2011, Dal Vechio et al. 2013, present work). In addition, long-term surveys provide necessary material to understand the basic biology of the species (Vitt 1982; Cruz 1994; Mesquita & Colli 2003; Werneck et al. 2009; Dal Vechio et al. 2014, 2015).

Sampling efforts at PNSCo resulted in the discovery and description of five new species: *Calyptommatus confusionibus* (Rodrigues et al. 2001), *Mesoclemmys perplexa* (Bour & Zaher 2005), *Stenocercus squarrosus* (Nogueira & Rodrigues 2006), *Ameivula confusioniba* and *Glaucomastix venetacauda* (Arias et al. 2011a). Another three are candidates to new species. *Adenomera* sp., was detected as a possible new species in a recent molecular study (Fouquet et al. 2014). This species also occurs at EEUU, being probably distributed along the northern Cerrado and ecotonal areas between this domain and the neighbors. *Leptodactylus* aff. *syphax* has recently been recognized as an undescribed new species (Loebmann & Haddad 2010; Andrade et al. 2011; Ribeiro et al. 2012; Cavalcanti et al. 2014). *Ameivula* sp. seems to be a hybrid between *A. confusioniba* and *G. venetacauda* (Arias pers. comun).

Five additional species are worthy of comment. The five specimens of *Trilepida* sampled in the PNSCo have scale counts that are intermediate between *T. koppesi* and *T. fuliginosa*. Although quantitatively more similar to *T. koppesi,* they present qualitative characters that resemble *T. fuliginosa* (Passos et al. 2006). *Trilepida koppesi* is distributed throughout the southern Cerrado region (São Paulo, eastern Mato Grosso do Sul and southern Goiás states), while *T. fuliginosa* is distributed in the northern part of the Cerrado, with the nearest record in Palmas (Tocantins basin), about 520 km in straight line far from PNSCo (Passos et al. 2006). Sampled specimens in PNSCo could either be an undescribed species or represent a large distribution extension for *Trilepida fuliginosa* and the first record of the species for the Caatinga domain.

Other interesting species is *Siphonops paulensis*, which is widely distributed in Brazil, with only a few records for the Caatinga biome

(Taylor 1968, 1970; Santana et al. 2015). The specimen of *S. paulensis* represents the first record for the state of Piauí and fills an important distribution gap for the group in northwestern Brazil. However, it is likely that *S. paulensis* represents a complex of cryptic species (Wilkinson, pers. comm.). Indeed, Loebmann and Haddad (2010) already pointed out the complex taxonomic nature of this species when they registered *Siphonops* for the "Brejo Nordestino" Planalto de Ibiapaba and considered their record as possibly representing an undescribed species related to *S. paulensis*. Here we treat the population in PNSCo as *S. paulensis*, pending further studies that could clarify the taxonomic status of populations considered to belong to this widely distributed species.

A recent phylogenic work of *Tropidurus semitaeniatus* showed the complexity of the the species, with several lineages candidates to fully species, inclusive, there are two distinct of these lineages that occur in PNSCo, being one of them exclusive to the park; and there is another exclusive lineage to EEUU (Werneck et al. 2015). Here, we maintain *T. semitaeniatus* as just one species until further taxonomic approach.

Amphisbaena frontalis is until now known only from the type locality at the left bank of middle São Francisco River in Alagoado, Bahia state. The record in PNSCo expands its distribution on approximately 270 km northwest of the type locality (Vanzolini 1991); and highlights an historical event of sand contact with southwestern Piauí and São Francisco Dunes as mentioned above. Finally, *Hoplocercus spinosus* is a typical Cerrado species that also occurs in contact areas with forested biomes. In PNSCo, *H. spinosus* was found in a forested Caatinga, representing the easternmost record and first for the species within the Caatinga domain (Torres-Carvajal et al. 2011).

The PNSCo together with other regional conservation units of similar latitude (*i.e.* EESGT, EEUU and PNSCa) protect an area of ca. 200.000 Km^2 in northern Cerrado and western Caatinga. There is a widespread herpetofaunal species throughout this landscape, occuring in all these four units, however, the fauna is more similar between Cerrado's units (EESGT + EEUU) and Caatinga's ones (PNSCo and PNSCa) but not so much between them. Despite the geographical proximity between these Cerrado and Caatinga parks, there is a turnover of congeneric species that seems to be related to the shift of domains (*e.g. Tropirurus oreadicus* X *Tropirurus hispidus; Stenocercus quinarius* X *Stenocercus squarrosus*; *Ameivula mumbuca* X *Ameivula confusioniba; Procellosaurinus erythrocercus* X *Vanzosaura savanicola; Epicrates crassus* X *Epicrates assisi*; *Phyllomedusa azurea* X *Phyllomedusa nordestina*; *Leptodactylus syphax* X *Leptodactylus aff. syphax*; *Physalaemus centralis* X *Physalaemus albifrons*; *Rhinella mirandaribeiroi* X *Rhinella granulosa*; *Rhinella schneideri* X *Rhinella jimi*; *Proceratophrys goyana* X *Proceratophrys cristiceps*). This shift on is also recovered in the similarity analysis, with PNSCo and PNSCa included within the "Caatinga" group while EESGT and EEUU units cluster within the "Cerrado" group, indicating that the units harbor a typical faunal of each biome.

Our results show that the PNSCo harbors one of the most diverse herpetofauna among the inventoried localities within the Caatinga domain, conferring to the park a strategic role for the conservation of the remaining regions of this vanishing domain. Our results also indicate that, despite geographical proximity, the northeastern Cerrado and Caatinga units studied still retain high levels of diversity and uniqueness with low faunal similarities between domains, evidencing a high species turnover.

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Periphytic algae in a floodplain lake and river under low water conditions

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Abstract: Drought events will become more frequent due the climate change. In floodplains, periphytic algae are responsible for part of the primary production, are the principal source of organic carbon deposition, play an important role in mineralization and nutrient cycling, and are the base of the food web for many organisms. As algae distribution in aquatic environments is a strong indicator of physical and chemical conditions of the sites, we aimed to determine the structure of periphytic algae in lentic and lotic environments during drought conditions and to uncover the main local abiotic factors in community structuring. We hypothesized diatoms would be more frequent than green algae and desmids at both sites, due to their resistance characteristics, and that higher periphyton algal richness, density and diversity would occur in the lake due to the greater availability of nutrients and the absence of flow. The study was carried out in the Finado Raimundo lake and the Ivinhema river in the Upper Paraná river floodplain during the low water period of 2011. Petioles of the aquatic macrophyte *Eichhornia azurea* (Sw.) Kunth were used as a substrate for periphytic algae. We found a total of 171 species, 104 species in the lake and 80 in the river. Diatoms were predominant at both sites due to their strategic traits, and between sites, there were different patterns in the periphytic algal community structure, owing to the distinctive physical and chemical characteristics of the lake and the river. *Achnanthidium minutissimum* (Kützing) Czarnecki and *Nitzschia palea* (Kützing) W. Smith were the most abundant species in both environments. Our results showed patterns of periphytic algae in a floodplain during drought conditions, which will assist in understanding their structuring during future drought scenarios.
Keywords: *diatoms, flood pulse, freshwater, drought events.*

BICHOFF, A., OSÓRIO, N.C., DUNCK, B., RODRIGUES, L. **Algas perifíticas em lago e rio de planície de inundação sob condições de águas baixas**. Biota Neotropica. 16(3): e20160159. http://dx.doi.org/10.1590/1676-0611-BN-2016-0159

Resumo: Eventos de seca serão mais frequentes, devido as mudanças climáticas. Em planícies de inundação, as algas perifíticas são responsáveis por parte da produção primária, são a principal fonte de deposição de carbono orgânico, desempenham um papel importante na mineralização e ciclagem de nutrientes, e são a base da cadeia alimentar de muitos organismos. Considerando que a distribuição de algas nos ambientes aquáticos é uma forte indicadora das condições físicas e químicas dos locais, objetivamos determinar a estrutura de algas perifíticas em ambientes lênticos e lóticos em condições de seca e de descobrir os principais fatores abióticos locais na estruturação da comunidade. Nossa hipótese é que as diatomáceas seriam mais frequentes do que as algas verdes e desmídias em ambos os locais, devido as suas características de resistência, e que a maior riqueza, densidade e diversidade de algas perifíticas que ocorreria no lago devido à maior disponibilidade de nutrientes e a ausência de fluxo. O estudo foi realizado no lago Finado Raimundo e rio Ivinhema na planície de inundação do alto rio Paraná, durante o período de águas baixas de 2011. Pecíolos das macrófitas aquáticas *Eichhornia azurea* (Sw.) Kunth foram utilizados como substrato para algas perifíticas. As diatomáceas foram predominantes em ambos os locais, devido às suas características estratégicas, e entre os locais, houve padrões diferentes na estrutura da comunidade de algas perifíticas, devido as características físicas e químicas distintas do lago e do rio. *Achnanthidium minutissimum* (Kützing) Czarnecki e *Nitzschia palea* (Kützing) W. Smith foram as espécies mais abundantes em ambos os ambientes. Nossos resultados mostraram padrões de algas perifíticas em uma planície de inundação em condições de seca, o que vai ajudar na compreensão de sua estruturação em futuros cenários de secas.
Palavras-chave: *diatomáceas, pulso de inundação, água doce, eventos de seca.*

Introduction

Floodplains are among the most productive ecosystems on the planet and represent major centres of biological diversity (Junk 1996). The extreme diversity of habitats in such environments provide high environmental heterogeneity, due to the typical limnological characteristics found in each habitat, which may determine the species pattern distribution and elevated biodiversity. Among floodplain habitats, lakes can be considered more complex than rivers in physical structures and limnological factors (Margalef 1983).

The Upper Paraná river floodplain is the last remaining wetland area of the Paraná River that is not dammed in Brazil (Agostinho 1997, Gois et al. 2015). This area has high habitat diversity, which endows it with unique limnological characteristics that allows conservation of high species diversity (Thomaz et al. 1997, Thomaz et al. 2007). In this floodplain, great floods (a water level of over 4.5 m) promote greater connectivity and homogenization between habitats (Thomaz et al. 2007), affecting aquatic communities (Agostinho et al. 2001) and changing ecosystem properties (Neiff 1990). However, the presence of reservoirs upstream of the Upper Paraná river (approximately 35 dams) controls the hydrological regime, influencing nutrient cycling, and therefore, the structure and composition of communities within this system (Agostinho et al. 2000, 2008, 2009, Stevaux et al. 2009, Souza-Filho et al. 2010). Flood periods in these areas have been found to have reduced frequency, duration, and intensity, thus increasing the periods of low water and drought in these environments (Souza-Filho 2009). In this context, it is expected that global drought events would be more frequent in the near future due to climate change and decreases in precipitation (Barnett et al. 2005, Woodwand et al. 2010).

The presence and distribution of algae in aquatic environments is a strong indicator of the physical and chemical conditions of these sites (Biggs 1996, Stevenson 1997, Moschini-Carlos & Henry 1997, Domitrovic et al. 2013, Moresco & Rodrigues 2013, Rodrigues et al. 2013). In floodplains, periphytic algae is responsible for part of the primary production, is the principal source of organic carbon deposition, plays an important role in mineralization and nutrient cycling, and is the base of the food web for many organisms (Stevenson 1996, Felisberto & Murakami 2013). These organisms are influenced by several factors in the floodplain, such as the water level, current flow, macro and micro nutrients, light, temperature, predation, and the substrate type (Rodrigues et al. 2003, Algarte et al. 2014, Dunck et al. 2016). The alterations caused by drought events and changes in water level also affect these communities, particularly in the availability of substrates for adhesion, reproduction and development (Stevenson 1996). Furthermore, in periods of drought, algae species tend to be more persistent and endure the drought despite the disturbance, compared with how they react in periods of flood (Schneck 2011).

In this context, we aimed to evaluate the periphytic algae community in floodplain lakes and rivers in drought conditions, taking into account the importance of studying drought events in a quickly changing world. We aimed to answer the following questions surrounding the lake and river: i) What is the pattern of the periphytic algae attributes (composition, richness, density and diversity) in each type of environment (lentic or lotic) in drought conditions? ii) What are the main local abiotic factors in the community structuring in both environments in drought conditions? We hypothesize that diatoms are more frequent at both sites, over green algae and desmids, due to their resistance characteristics (Round et al. 1990, Passy 2007). Between sites, we hypothesize that higher periphyton algal richness, density and diversity occurs in the lake due to the greater availability of nutrients and the absence of flow (Rodrigues & Bicudo 2001, Stevenson 1996) and habitat complexity. Then a record of aquatic community structure during this event may help us understand how organisms may behave in the face of climate change and droughts. Furthermore, we believe our study may provide evidence that is potentially applicable to future drought scenarios in wetlands worldwide.

Materials and Methods

The study was carried out at two sites, the Finado Raimundo lake (22º47' 57.6"S and 53º32'29.16"W) and Ivinhema river (22º47'59.64"S e 53º32' 21.3"W) of the Upper Paraná river floodplain, which belong to the State Park of Ivinhema River, State of Mato Grosso do Sul, Brazil. The Ivinhema river runs parallel to the Paraná river in its lower reaches (sampled area), connecting to the Baia river by the Curutuba channel and to the Paraná river by the Ipoitã channel. The Finado Raimundo lake has an elongated shape and is approximately 2.9 km in length with an average width of 500 m, connecting to the Paraná river through a channel 50-m long and 20-m wide (Souza-Filho et al. 2000) (Figure 1).

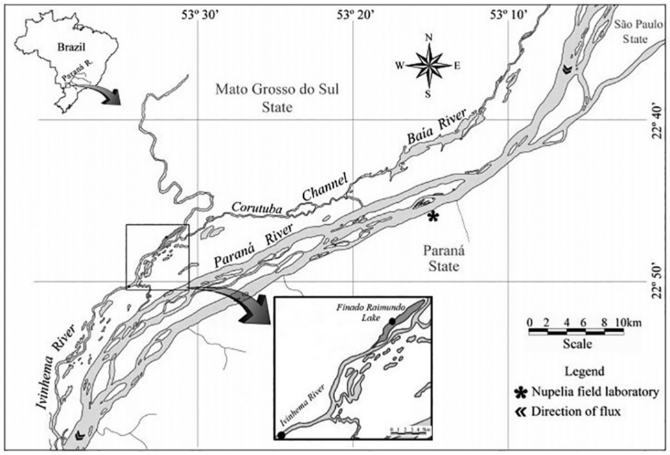

Figure 1. Upper Paraná river floodplain and the location of the Ivinhema river and Finado Raimundo lake.

The samplings were performed three times in 2011 during drought conditions (low water period, June, September and December) because there was no direct influence of the Paraná river on the study sites (Ivinhema River and Finado Raimundo Lake, Table 1).

In each sampling, limnological variables were sampled (simultaneously to biotic sample) in the subsurface of the limnetic zone in both environments. The following variables were measured: depth (m), water temperature (ºC) (oxímetro- YSI55A), dissolved oxygen (mg.L^{-1}) (oximeter- YSI55A), conductivity (µS.cm^{-1}) and pH (Digimed digital potentiometers), turbidity (NTU) (turbidimeter- LaMotte2020e), inorganic and organic dissolved solids (mg.L^{-1}), total alkalinity (µEq.L^{-1}, Carmouze 1994), ammonium (NH_4^+), orthophosphate (PO_4^{++}), total nitrogen and total phosphorus (µg.L^{-1}) (Table 1). Water samples were filtered through Whatman GF/F filters, under low pressure (< 0.5 atm) and stored at -20ºC for later determination of the dissolved nutrient

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Table 1. Morphometric and limnological parameters of Finado Raimundo lake and Ivinhema river in the Upper Paraná River floodplain during sampling in 2011. (NH_4^+): ammonium, (PO_4^{++}): orthophosphate.

Variables	Lake Finado Raimundo			River Ivinhema		
	June	**September**	**December**	**June**	**September**	**December**
Depth (m)	4	4.25	3.5	4.5	4.3	4
Water temperature (°C)	19.2	24.3	29.1	17.8	23.3	28.2
Dissolved O_2 (mg L^{-1})	7.72	10.02	6.99	7.92	7	6.31
pH	6.57	8.95	7.01	7.23	7.09	6.77
Conductivity (uS/cm)	35.5	38.9	39.7	34.5	39.7	42.7
Turbity (NUT)	10.13	15.95	11.09	18	23.5	24
Inorganic dissolved solids (mg L^{-1})	0.35	0.47	0.09	2.6	7	2.26
Dissolved organic solids (mg L^{-1})	0.3	1.19	0.54	0.55	1.64	0.64
Alkalinity (mEq L^{-1})	172	441.3	342.2	190.3	323.1	339.4
Total Nitrogen (ug L^{-1})	764	1226.4	1319.6	725.9	1101.1	790.5
NH_4^+ (ug L^{-1})	28	8.2	17.8	28.2	6.9	14.9
Total Phosphorus (ug L^{-1})	28.5	51.7	42.6	30.9	49.5	83.7
PO_4^{++} (ug L^{-1})	6.99	10.5	7.8	18.07	22.6	19.8

fractions and suspended material (Bergamin et al. 1978, Mackereth et al. 1978) (Table 1). The daily water level of the Paraná river is available at http://www.peld.uem.br/.

Two banks of the macrophyte *Eichhornia azurea* (Sw.) Kunth were sampled in each environment (lake and river) and these same banks were used in all samplings. These macrophyte banks were located in opposite margins. In each macrophyte bank it was selected a random branch and periphytic algae were collected from the sixth or seventh plant internode. In each environment we collected two petioles, totaling two sub-samples per site, and all together totaling twelve samples in the study. The general pattern of results are reported as the sample means for each environment per month. The periphytic material was taken by scraping part of the *E. azurea* petioles, using a stainless-steel blade wrapped in aluminium foil and jets of distilled water (Bicudo 1990, Schwarzbold 1990). The area scraped from the substrate (cm^2) was calculated from measurements of the length and width of each petiole. The material removed was preserved with acidified (0.5% acetic acid) Lugol's solution for later counting, and the material for the qualitative analysis was preserved in Transeau's solution at a ratio of 1:1 as recommended by Bicudo & Menezes (2006).

The periphytic algal samples were quantified using sedimentation chambers in an Olympus® M021 inverted microscope following the Utermöhl method (1958). The counts were carried out in random fields until reaching at least 100 individuals (cells, colonies, or filaments) from the most common species in each sample and according to the species accumulation curve following Ferragut & Bicudo (2012). Qualitative analyses were performed by temporary glass slides in a binocular optical microscope, with micrometre oculars of 400x and 1000x, for species identification alone. The species identification was performed using classical literature such as Prescott (1982), Round et al. (1990), Bourrelly & Couté (1991), Dillard (1991), John et al. (2002), Wehr & Sheath (2003) and Bicudo & Menezes (2006) and regional studies (Fonseca & Rodrigues 2005a, Moresco & Rodrigues 2006, Biolo & Rodrigues 2010, 2011, Bartozek et al. 2013) for reference. The classification system adopted was that proposed by Round (1965, 1971).

Species density was estimated according to Ros (1979), and the results were expressed as number of individuals per unit area (ind cm^{-2}). The algal richness was estimated from algal density, using the species accumulation curve. We determined the dominance and abundance of species as proposed by Lobo & Leighton (1986), where the dominant species are those with densities greater than 50% of the total density of the community, and the abundant species were those with densities exceeding the mean population density of each sample. The species diversity was estimated by the Shannon-Weaver index (1963) for the sites in all samplings.

Data analysis

A principal component analysis (PCA) was applied to analyse the spatial and temporal variation of limnological variables of the environments. We tested the correlation of limnological variables to avoid multicollinearity and used water temperature, dissolved oxygen, conductivity, pH, turbidity, inorganic and organic dissolved solids, ammonia and orthophosphate for this analysis. The data were previously standardized [(Xij-Xi)/Si] (Legendre & Legendre 1998). Axis retention was evaluated under the broken-stick criterion (Jackson 1993).

T-tests were used to test for differences in community attributes (richness, density and diversity) between the environments (the lake and the river). A nonmetric multidimensional scaling (NMDS) was applied to the dominant and abundant species matrix to investigate the species distribution in the sites for all samplings. The Bray-Curtis index was used to calculate the distances and check the stress level of adjustment of the newly generated axes and the ordering of the dissimilarity matrix (Legendre & Legendre 1998). The analyses were performed using R software version 3.1.2 (R Development Core Team 2014) and graphics in Statistica, version 7.1 (StatSoft 2005).

Results

The PCA results showed that 77.5% of the limnological data variability was summarized in the first two axes with eigenvalues 3.51 and 3.45, respectively. The first axis was most influenced by pH (loading r = 0.95) and turbidity (loading r = 0.21) (Figure 2). The second axis had a greater relationship with turbidity (loading r = 0.70) and inorganic dissolved solids (loading r = 0.57) (Figure 2). The PCA 1 axis showed a temporal difference between the samplings, with similarity between LS and RS; and in the other side RJ and LJ (Figure 2). The second axis showed differences between the river and the lake (Figure 2). In general, the lake was more influenced by dissolved oxygen (DO) and NH_4^+, whereas the river was most influenced by dissolved solids, conductivity and PO_4^{++} (Table 1).

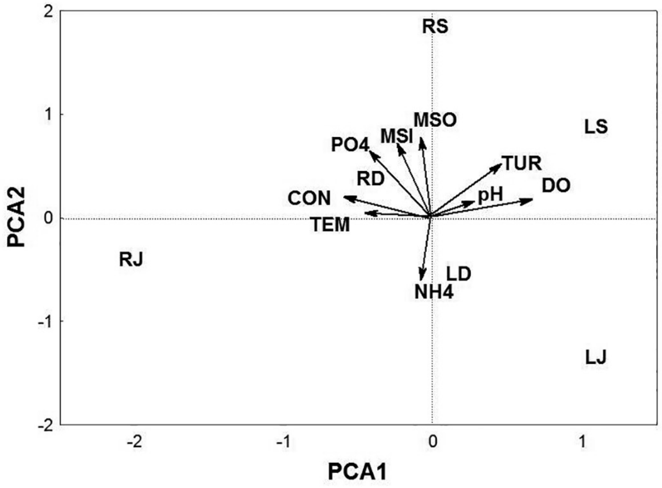

Figure 2. Site scores derived from a principal component analysis applied to the limnological dataset. (LJ): Lake sampling in June, (LS): Lake sampling in September, (LD): Lake sampling in December, (RJ): River sampling in June, (RS): River sampling in September, (RD): River sampling in December, (CON): conductivity, (NH4): ammonium, (TEM): temperature, (PO4): orthophosphate, (MSI): inorganic dissolved solids, (MSO): dissolved organic solids, (ALK): total alkalinity, (NT): total nitrogen, (DO): dissolved oxygen, (TUR): turbidity.

We found 171 species and the lake was represented by 104 species divided into eight classes presented in the following descending order: Bacillariophyceae, Zygnemaphyceae, Chlorophyceae, Cyanophyceae, Chrysophyceae, Euglenophyceae, Oedogoniophyceae and Xanthophyceae (Figure 3a). The river was characterized by 80 species distributed among six classes: Bacillariophyceae, Zygnemaphyceae, Chlorophyceae, Cyanophyceae, Oedogoniophyceae and Rhodophyceae (Figure 3b). Chrysophyceae, Euglenophyceae and Xanthophyceae belong exclusively to the lake, whereas species belonging to Rhodophyceae were found only in the river.

Among the species found in both sites (supplementary material), 13 species were common to both environments, with 10 species belonging to Bacillariophyceae (*Achnanthidium minutissimum* (Kützing) Czarnecki, *Fragilaria capucina* Desmazières*, Frustulia saxonica* Rabenhorst*, Gomphonema parvulum* (Kützing) Kützing*, Melosira varians* C. Agardh, *Navicula cryptotenella* Lange-Bertalot, *Nitzschia palea* (Kützing) W. Smith, *Pinnularia latarea* Krammer, *Synedra goulardii* Brébisson ex Cleve and Grunow and *Ulnaria ulna* (Nitzsch) P. Compère), two species belonging to Zygnemaphyceae (*Closterium leibleinii* Kützing ex Ralfs and *Staurastrum trifidum* Nordstedt) and one species belonging to Chlorophyceae (*Stigeoclonium* sp.).

The lake had a species density 2.45 times higher than the river (t = 2.45, p = 0.02, Fig. 4). Bacillariophyceae was the most abundant class in both environments (Figure 4). The most abundant species in the two environments were *A. minutissimum* and *N. palea*. In the river, *M. varians* was the most abundant species, and in the lake, *F. capucina* was most abundant species. September was the month that both sites showed the highest species richness and density (Figures 3 and 4).

There was no mean difference in species richness between the sites (t = 1.87, p = 0.13) and a similar pattern was found for species diversity (t = 1.79, p = 0.14). In contrast, the NMDS showed a clearly difference between the communities in both types of environments (stress = 0.065) (Fig. 5). Among the river species, the most abundant in June were *Frustulia pumilio* Lange-Bertalot and U. Rumrich (Frp), *Encyonema silesiacum* (Bleisch) D.G. Mann (Eco) and *Gomphonema gracile* Ehrenberg (Gog) (Figure 5). In September, the most abundant species were *Nitzschia clausii* Hantzsch (Nic) and *Surirella angusta* Kützing (Sui), and in December, the most abundant species were *Gomphonema pumilum* (Grunow) E. Reichard and Lange-Bertalot (Gol) and *Gomphonema mexicanum* Grunow (Gom) (Figure 5). The *Gomphonema* species had the highest representation in the river. However, in the lake communities, the most abundant species in June were *Oedogonium* sp. 2 (Oem) and *Oedogonium* sp. 3 (Oep) (Figure 5). The most abundant species in September was *Gomphonema brasiliense* Grunow (Goe), and the most abundant species in December were *F. capucina* (Frc) and *N. palea* (Nip) (Figure 5).

Discussion

Climate change represents a complex amalgam of stressors, which include the increased frequency and intensity of droughts and extreme flow events (Woodward et al. 2010). This study demonstrated that in low water levels, diatoms are predominant in both sites. Between the environments, there are different patterns in periphytic algal community structure, which agrees with our hypothesis. These differences were attributed mainly due to the physical and chemical characteristics of the lake and the river and their different hydrodynamics (Rodrigues et al. 2003).

There is an important spatial and temporal variability in the limnological characteristics of the Upper Paraná river floodplain habitats (Thomaz et al. 2004, Algarte et al. 2006). Studies in this floodplain that have evaluated periods of both high and low water levels have shown that diatoms dominate the periphyton under these two hydrological conditions (Fonseca & Rodrigues 2005b, Algarte et al. 2006, Biolo & Rodrigues 2013, Carapunarla et al. 2014). In these studies, at low water levels, the diatoms were favoured by environmental conditions such as low temperatures and concentrations of phosphorus forms, higher concentrations of dissolved oxygen and nitrogen for both environments. In the present study, both environments were related to the higher values of dissolved oxygen in June and September corroborating with these other studies. In general, the lake had higher values of nitrogen and dissolved oxygen, whereas the river had higher values of conductivity, suspended solids and phosphorus. The high values of dissolved oxygen in the lake and suspended solids in the river in June and September may explained the high density of diatoms, which, by having chlorophyll c, develop better in low-light conditions (Round et al. 1990). In the lake, the dominance of diatoms maybe by it be excellent competitors

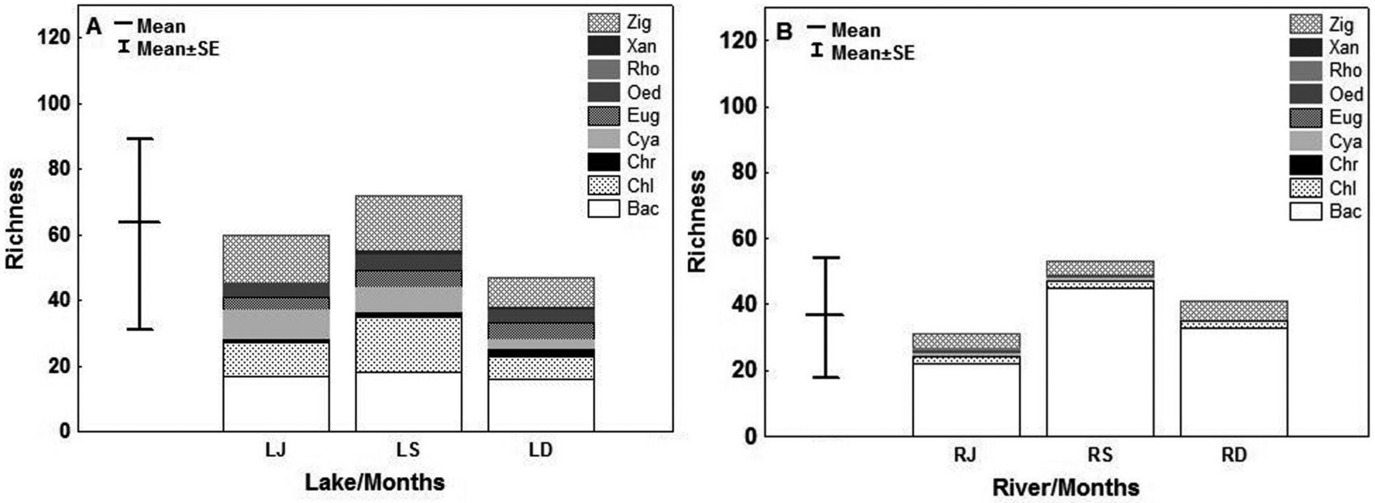

Figure 3. Algal richness (mean ± standard error) during samplings and algal groups richness between sites (a: Lake; b: River) and samplings, (LJ): Lake sampling in June, (LS): Lake sampling in September, (LD): Lake sampling in December, (RJ): River sampling in June, (RS): River sampling in September, (RD): River sampling in December, (Bac): Bacillariophyceae, (Chl): Chlorophyceae, (Chr): Chrysophyceae, (Cya): Cyanophyceae, (Eug): Euglenophyceae, (Oed): Oedogoniophyceae, (Rho): Rhodophyceae, (Xan): Xanthophyceae, (Zig): Zygnemaphyceae.

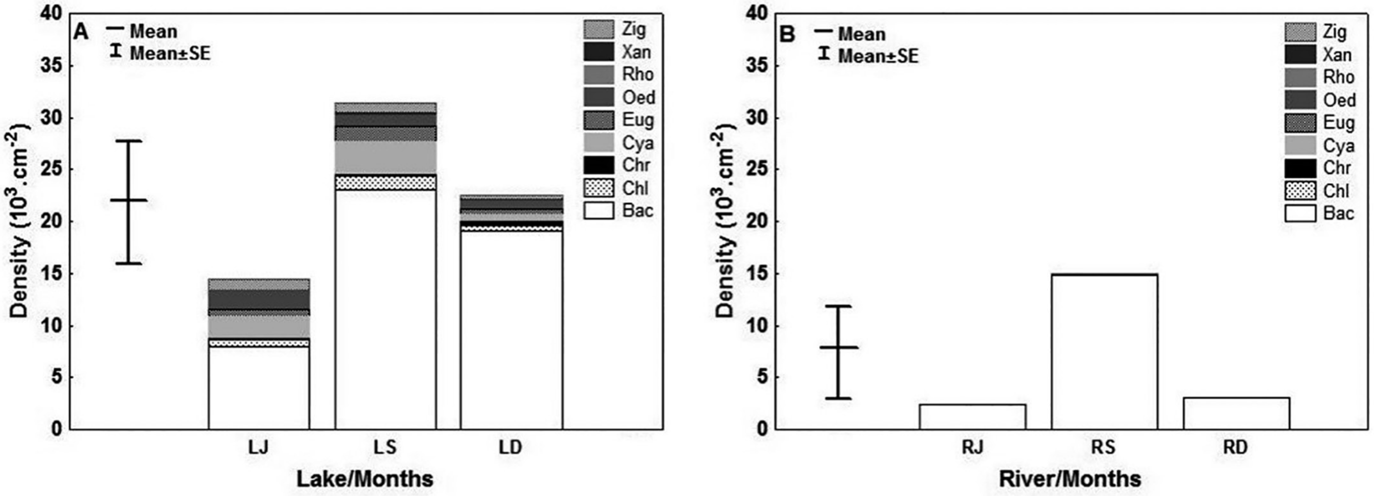

Figure 4. Algal density (mean ± standard error) during samplings and algal group density between sites (a: Lake; b: River) and samplings, (LJ): Lake sampling in June, (LS): Lake sampling in September, (LD): Lake sampling in December, (RJ): River sampling in June, (RS): River sampling in September, (RD): River sampling in December, (Bac): Bacillariophyceae, (Chl): Chlorophyceae, (Chr): Chrysophyceae, (Cya): Cyanophyceae, (Eug): Euglenophyceae, (Oed): Oedogoniophyceae, (Rho): Rhodophyceae, (Xan): Xanthophyceae, (Zig): Zygnemaphyceae.

in turbid environments and low phosphorus concentrations (Cetto et al. 2004). Our results showed that physical variables are important factors to distinguish the limnological characteristics between lentic and lotic environments, which agrees with data by Rodrigues & Bicudo (2001).

The richness and diversity of periphytic algae were not significantly different between environments, and thus, they were not the attributes that best explained the spatial difference between the lotic and lentic sites. However, the species composition and density values between the environments were clearly distinct. The highest values for algal densities were recorded in September for both sites, in agreement with Algarte et al. (2006, 2009) for environments of the same floodplain. September presented the lowest hydrometric water level, allowing the development of loosely attached species due to the higher hydrometric water level and resultant physical disturbance, which made permanence difficult for species attached to the substrate (Algarte et al. 2006). The community structuring demonstrated that Bacillariophyceae was predominant in the river as well as in the lake. Its highest density occurred in the lake, and species richness and diversity were similar between sites, whereas September showed higher values for species richness and density. The diatoms were predominant in both environments, possibly due to their fast and efficient colonization, occupying the substrates within one to several weeks (Stevenson 1996, Hoagland et al. 1986). Furthermore, many diatoms have specialized structures for attachment to the substrate, such as mucilaginous peduncles in the *Gomphonema* species and the production of mucilaginous matrices as in *Frustulia* and *Navicula* (Round 1991). The highest values for density and richness of diatoms in the lentic system is partly related to their specialized structures that

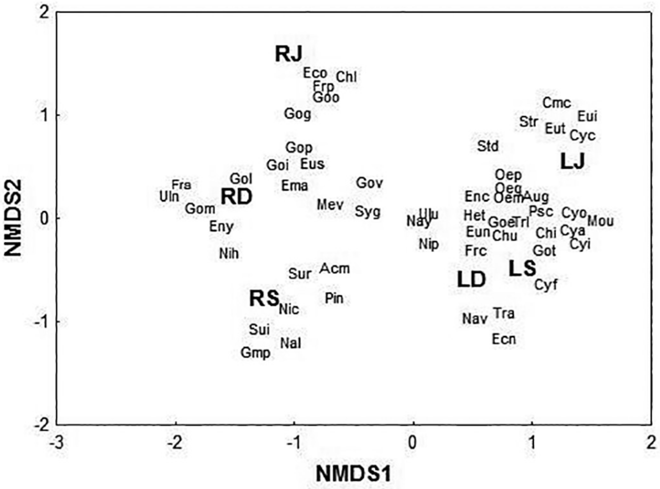

Figure 5. Nonmetric multidimensional scaling (NMDS) of periphytic algal communities between sites and samplings, (LJ): Lake sampling in June, (LS): Lake sampling in September, (LD): Lake sampling in December, (RJ): River sampling in June, (RS): River sampling in September, (RD): River sampling in December.

confer a competitive advantage over other species of different classes in stressful environmental conditions (Biggs 1996).

In term of species richness, the classes Zygnemaphyceae and Chlorophyceae were well represented in the lake. The presence of macrophyte stands in these sites favours the development of these groups because these plants release dissolved nutrients and reduce the turbulence of the water column (Rodrigues & Bicudo 2001, Algarte et al. 2006). The desmids have a selective advantage in more protected environments, are less disturbed (low current flow), and have a relatively large size, which makes them difficult to be removed by herbivory. According to Coesel (1996), truly planktonic desmids are rare and most of them use at least one substrate to reproduce.

Chrysophyceae, Euglenophyceae and Xanthophyceae were exclusive of the lentic environment. The chrysophytes are opportunists, present rapid growth, are adapted to growing in vegetated areas, have flagella and adaptive strategies for using different resource sources as well as are more related to dry periods and environments with higher dissolved oxygen concentrations (Almeida & Melo 2011, Golebski & Ferragut 2012). The euglenoids are optional heterotrophs and are commonly found in sediment (Wetzel 1983). The presence of these organisms in lentic environments may be related to a possible interaction with epipelic communities. The xanthophytes can benefit in environments with reduced depth, low transparency and medium levels of phosphorus, nitrate and ammonium (Bovo-Scomparin et al. 2005). Rhodophyceae occur only in lotic sites because they are typically found in environments with moderate current velocity, high transparency, and no organic pollution (Esteves 1998).

A. minutissimum and *N. palea* were the most abundant species in both sites. *A. minutissimum* is among the diatoms more often recorded in periphytic samples worldwide (Krammer & Lange-Bertalot 1991, Round 2004). This species can occur in acidic to alkaline environments (Round 2004) and is oligotrophic to eutrophic (Leandrini et al. 2013, Dunck et al. 2015). *N. palea* is generally considered tolerant to organic pollution (Bes & Torgan 2010) and may also have wide distribution, including lakes (Bes & Torgan 2010), rivers (Rusanov et al. 2009), streams (Dunck et al. 2015), and floodplains (Weilhoefer & Pan 2006).

In lotic environments, *M. varians* was the most abundant species. In contrast, McIntire (1968) showed these algae were more abundant in lentic environments. However, Fogg & Reimer (1995) reported that *M. varians* is cosmopolitan and can be found in a large range of ecological conditions, and its density is not determined by physical factors such as current velocity flow. *F. capucina* was important for both environments; however, this species was more abundant in the lake. Lobo et al. (2004) reported this species as highly tolerant to eutrophication, although there are studies that consider this species related to less polluted waters (Ten Cate et al. 1993) or very tolerant to low nutrient supplies (Hinojosa-Garro et al. 2010). Thus, these conflicting results concerning the response of *F. capucina* to environments suggest that it is a cosmopolitan species.

The NMDS results showed that more species of genus *Gomphonema* discriminated the lotic site (*G. pumilum*, *G. mexicanum* and *G. gracile)*. The *Gomphonema* genus has several cosmopolitan species and is well represented in richness and density in aquatic environments, usually in lotic environments (Tremarin et al. 2009). This species has cells that are typically attached to solid substrates by pads or mucilaginous peduncles (Hoagland et al. 1982, Burliga & Schwarzbold 2013), making this species more resistant to disturbances.

Other species that were more abundant in the river compared with the lake were *E. silesiacum*, *N. clausii* and *S. angusta*. Salomani & Torgan (2008) observed the presence of *E. silesiacum* in sites with low concentration of organic matter, in contrast with our results, as the river contained more suspended organic material. September was a period in which inorganic dissolved solids and total nitrogen were high, which may have contributed to the high density of *N. clausii* and *S. angusta* in the river.

Oedogoniophyceae were abundant in the lake, especially those of the *Oedogonium* genus. These algae increase the colonization surface, serving as a substrate for other algal classes, such as Bacillariophyceae (Carapunarla et al. 2014). These species are typically periphytic and can be found attached to many types of substrates (Lee 2008). They are efficient competitors for resources such as light and space and are associated with high concentrations of nutrients, electrical conductivity (Cavati & Fernandes 2008) and lower current velocity (Biggs 1996, Simons 1994), as in the present lentic environment.

In summary, we conclude that in floodplain drought conditions, periphytic algal species composition and density responded better to the effects of site hydrodynamics than algal richness and diversity. Diatoms were predominant in both sites due to their strategic traits, and between sites, there were different patterns in periphytic algal community structure, owing to the physical and chemical characteristics of the lake and the river. Our study revealed the primary importance of conductivity, suspended solids and phosphorus for the river and turbidity and nitrogen for the lake as abiotic local factors in periphytic algae structuring in drought conditions in the Upper Paraná river floodplain. Therefore, we have improved our understanding of the processes that govern the periphyton algal community in floodplain drought conditions.

Supplementary material

Supplement 1. Species occurrence (and its respective abbreviations) at the lentic and lotic environments in the sampling months. (LJ): Lake sampling in June, (LS): Lake sampling in September, (LD): Lake sampling in December, (RJ): River sampling in June, (RS): River sampling in September, (RD): River sampling in December.

Táxons	Abbreviation	LJ	LS	LD	RJ	RS	RD
Bacillariophyceae							
Achnathes sp.	Ach					X	X
Achnathes sp.1	Acn					X	
Achnanthidium minutissimum	Acm			X	X	X	X
Amphipleura lindheimeri	Amp				X	X	
Aulacoseira granulata	Aug	X	X	X			
Aulacoseira sp.	Aul		X	X			
Diploneis sp.	Dip					X	
Encyonema neomesianum	Ecn		X	X			
Encyonema silesiacum	Eco				X		
Encyonema sp.	Eny					X	X
Encyonema sp.1	Ema				X	X	X
Encyonema sp.2	Enc	X	X	X			
Eunotia sudetica	Eus				X	X	X
Eunotia sp.	Eun	X	X	X			
Eunotia sp.1	Eni						X
Eunotia sp.2	Euo	X					
Eunotia sp.3	Eut	X					
Eunotia sp.4	Eui	X					
Eunotia sp.5	Eua	X	X				
Eunotia sp.6	Eno						X
Eunotia sp.7	Ena						X
Eunotia sp.8	Ent					X	X
Eunotia sp.9	Eto						X
Eunotia sp.10	Etu					X	
Eunotia sp.11	Eti					X	
Eunotia sp.12	Eta				X		
Fragilaria capucina	Frc	X	X	X		X	
Fragilaria sp.	Fra					X	X
Frustulia pumilio	Frp				X		X
Frustulia saxonica	Fru	X	X	X	X	X	X
Gomphonema augur	Goa				X	X	X
Gomphonema brasiliense	Goe	X	X	X			
Gomphonema gracile	Gog				X		X
Gomphonema insigne	Goi				X	X	X
Gomphonema mexicanum	Gom					X	X
Gomphonema parvulum	Gov	X	X	X	X	X	X
Gomphonema pumilum	Gol				X	X	X
Gomphonema turris	Got	X	X				
Gomphonema sp.	Gop				X	X	X
Gomphonema sp.1	Goh					X	X
Gomphonema sp.2	Goo				X	X	
Gomphonema sp.3	Gos					X	
Gomphonema sp.4	Gon					X	

Continued Supplement 1.

Táxons	Abbreviation	LJ	LS	LD	RJ	RS	RD
Gomphonema sp.5	Gma					X	
Gomphonema sp.6	Gmp					X	
Gyrosigma sp.	Gyr					X	
Gyrosigma sp.1	Gyn						X
Melosira varians	Mev	X	X		X	X	X
Navicula cryptotenella	Nay	X	X	X	X	X	X
Navicula sp.	Nav		X	X			
Navicula sp.1	Nau				X		X
Navicula sp.2	Nal					X	
Navicula sp.3	Naa					X	
Navicula sp.4	Nac					X	
Navicula sp.5	Nvi					X	
Navicula sp.6	Nva					X	
Nitzschia clausii	Nic					X	X
Nitzschia palea	Nip	X	X	X		X	X
Nitzschia sp.	Nit		X				
Nitzschia sp.1	Niz						X
Nitzschia sp.2	Nih					X	X
Nitzschia sp.3	Nii					X	
Nitzschia sp.4	Nia					X	
Pinnularia latarea	Pin			X		X	X
Surirella angusta	Sui					X	
Surirella sp.	Sur				X	X	
Surirella sp.1	Sue					X	
Synedra goulardii	Syg	X	X	X	X	X	X
Synedra sp.	Syn				X		
Ulnaria ulna	Ulu	X	X	X	X	X	X
Ulnaria sp.	Uln						X
Chlorophyceae							
Ankistrodesmus sp.	Ank		X				
Chaetophora elegans	Cha	X	X				
Characium sp.	Chi	X	X				
Characium sp.1	Chu	X	X	X			
Characium sp.2	Chc			X			
Schroederia sp.	Sch		X				
Chlorophyceae 1	Chr						X
Chlorophyceae 2	Chl				X		
Chlorophyceae 3	Chb					X	
Chlorophyceae 4	Cor	X					
Chlorophyceae 5	Cho				X		
Chlorophyceae 6	Cbc		X				
Chlorophyceae 7	Clr	X	X	X			
Coelastrum cambricus	Coc			X			
Coelastrum sp.	Coe	X					
Coelastrum sp.	Cte	X	X				

Continued Supplement 1.

Táxons	**Abbreviation**	**LJ**	**LS**	**LD**	**RJ**	**RS**	**RD**
Chlorococcales sp.	Cna	X					
Glaucocystis sp.	Gla	X	X				
Pediastrum tetras	Ped		X	X			
Scenedesmus acutus	Sca	X	X	X			
Scenedesmus bijugus	Scb		X				
Scenedesmus rarceboski	Scr						X
Scenedesmus sp.	Scn		X	X			
Scenedesmus sp.1	Scd		X				
Selenastrum sp.	Sel		X				
Stigeoclonium sp.	Sti		X			X	
Treubaria sp.	Ter		X				
Chrysophyceae							
Chrysosphaera sp.	Cry			X			
Mallomonas sp.	Mll	X	X				
Mallomonas sp.1	Mal			X			
Cyanophyceae							
Anabaena sp.	Ana		X	X			
Calotrix sp.	Cal	X					
Chroococcus sp.	Cus			X			
Cyanophyceae 1	Cya	X	X				
Cyanophyceae 2	Cyn				X		
Cyanophyceae 3	Cyc	X					
Cyanophyceae 4	Cyo	X	X				
Cyanophyceae 5	Cyf	X	X				
Cyanophyceae 6	Cyi	X	X				
Heteroleibleinia sp.	Het	X	X	X			
Microcystis sp.	Mic		X				
Nostoc sp.	Nos	X					
Pseudanabaena catenata	Psc	X	X				
Pseudanabaena sp.	Pse					X	
Euglenophyceae							
Euglena sp.	Eug	X					
Phacus sp.	Pha			X			
Strombomonas fluviatilis	Stf		X				
Strombomonas sp.	Str	X					
Trachelomonas sp.	Trs	X		X			
Trachelomonas sp.1	Tra		X	X			
Trachelomonas sp.2	Trm		X	X			
Trachelomonas sp.3	Trn		X				
Trachelomonas sp.4	Trl	X	X	X			
Oedogoniophyceae							
Bulbochaete sp.	Bul	X	X	X			
Oedogonium sp.	Oed				X	X	
Oedogonium sp.1	Oeg	X	X	X			
Oedogonium sp.2	Oem	X	X	X			

Continued Supplement 1.

Táxons	Abbreviation	LJ	LS	LD	RJ	RS	RD
Oedogonium sp.3	Oep	X	X	X			
Oedogonium sp.4	Oez		X				
Rhodophyceae							
Audouinella pygmaea	Aud				X		
Xanthophyceae							
Characiopsis sp.	Cps		X	X			
Zygnemaphyceae							
Actinotaenium sp.	Act		X				
Closterium leibleinii	Cll	X	X			X	
Closterium moniliferum	Clm				X		
Closterium sp.	Clo		X				
Closterium sp.1	Clt				X		
Closterium sp.2	Cls		X				
Cosmarium cf. *vexatum*	Cmv					X	X
Cosmarium comissurales	Cms	X					
Cosmarium granatum	Cmg		X				
Cosmarium psedoconnatum	Cmp			X	X		
Cosmarium sp.	Com	X	X				
Cosmarium sp.1	Coi		X		X		
Cosmarium sp.2	Cou	X	X	X			
Cosmarium sp.3	Cma	X					
Cosmarium sp.4	Cmu		X				
Cosmarium sp.5	Cmb	X	X	X			
Cosmarium sp.6	Cmc	X					
Cosmarium sp.7	Cme	X	X	X			
Cosmarium sp.8	Cmm				X		X
Cosmarium sp.9	Csm					X	
Euastrum rectangulare	Eur		X				
Euastrum sp.	Eum	X					
Euastrum sp.1	Euf			X			
Euastrum sp.2	Eup					X	
Mougeotia sp.	Mog					X	
Mougeotia sp.1	Mot						X
Mougeotia sp.2	Mou	X	X				
Staurastrum cf. *teliferum*	Stt			X			
Staurastrum leptocanthum var. *borgei*	Stl			X			
Staurastrum margaritacium	Stm	X	X				
Staurastrum quadrangulare	Stq	X					
Staurastrum setierum	Ste						X
Staurastrum trifidum	Std	X	X				X
Staurastrum volans	Stv		X				
Staurodesmus cuspidatus	Stc		X	X			
Staurodesmus dickiei	Stk			X			
Staurastrum sp.	Sts						X
Staurastrum sp.1	Stu	X					
Staurastrum sp.2	Sta	X					

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short communication

Length-weight relationships of the ichthyofauna from a coastal subtropical system: a tool for biomass estimates and ecosystem modelling

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VAZ-DOS-SANTOS, A.M, GRIS, B. **Length-weight relationships of the ichthyofauna from a coastal subtropical system: a tool for biomass estimates and ecosystem modelling**. Biota Neotropica. 16(3): e20160192. http://dx.doi.org/10.1590/1676-0611-BN-2016-0192

Abstract: Aiming to analyse the growth pattern, to allow biomass estimates and consequently to subsidize the ecosystem modelling, the length-weight relationships (LWR) of 39 fish species from the Araçá Bay, a subtropical coastal area chosen as model for a holistic study comprising environmental, social and economic aspects have been estimated. The objective of this study was to provide LWR for the fishes from the area itself, accurately based on the life stages of fish populations present there. Particularly for *Albula vulpes*, *Trachinotus carolinus*, *T. falcatus*, *Archosargus rhomboidalis* and *Kyphosus sectatrix* these are the first records of LWR in Brazil.
Keywords: *Araçá Bay, relative growth, Huxley model, Brazil.*

VAZ-DOS-SANTOS, A.M, GRIS, B. **Relações comprimento-peso da ictiofauna de um sistema costeiro subtropical: uma ferramenta para estimativas de biomassa e modelagem de ecossistemas**. Biota Neotropica. 16(3): e20160192. http://dx.doi.org/10.1590/1676-0611-BN-2016-0192

Resumo: Com o objetivo de analisar o padrão de crescimento e viabilizar estimativas de biomassas e, consequentemente, subsidiar a modelagem ecossistêmica, foram estimadas as relações comprimento-peso (RCP) de 39 espécies de peixes da baía do Araçá, uma área costeira subtropical escolhida como modelo para um estudo holístico compreendendo aspectos ambientais, sociais e econômicos. O objetivo deste estudo foi fornecer RCP para os peixes da própria área, baseadas nas estágios de vida das populações ictiícas ali presentes. Especialmente para *Albula vulpes*, *Trachinotus carolinus*, *T. falcatus*, *Archosargus rhomboidalis* e *Kyphosus sectatrix* estas são as primeiras estimativas de RCP no Brasil.
Palavras-chave: *baía do Araçá, crescimento relativo, modelo de Huxley, Brasil.*

Introduction

In the current ichthyology, it is noteworthy that biodiversity conservation and sustainability are not dissociable. Socio-ecosystem approach for fishery management is a fact (Baigun et al. 2012). In the Southwestern Atlantic Ocean comprising the Brazilian coast, there are many marine environments with different levels of impact (Lana et al. 2001, Meniconi et al. 2012), consequence of global changes in local, medium and high scales (Ray & McCormick-Ray 2014). The Araçá Bay (23°48'47,3"S 45°24'22,1"W) (Figure 1) is one of them, and it was chosen as a model for a holistic and integrated study comprising biology, ecology, oceanography, economy, sociology and policy (BIOTA FAPESP Araçá 2015).

Araçá Bay is an area of approximately 1 Km^2 subjected to daily tides, showing tidal pools, mangroves, rocky substrates, sandy beaches and the typical pelagic and benthic habitats. It shelters three beaches and two small islands. The Mãe Isabel stream flows in its north portion. In addition, it is directly affected by pollution and by the São Sebastião Port, which is close to the area. Fishermen live surrounding the bay using it and its adjacent areas for survival. Biodiversity and species richness in Araçá Bay are surprisingly high, playing an important role in the productivity of the adjacent areas (Amaral et al. 2010, 2015).

Concerning the fish population dynamics and the community structure, length-weight relationships (LWR) are one of the most useful tools in applied ichthyology and fishery management (Pauly 1984, Froese 2006). Among their several applications (Vianna et al. 2004, Macieira & Joeux 2009, Joeux et al. 2009, Silveira & Vaz-dos-Santos 2015), LWR are used to estimate fish biomass, the basis for the ecosystem modelling (Pope et al. 2006, Gasalla et al. 2007, Maury et al. 2007, Froese et al. 2008), one of the main purposes of the Araçá Bay study (BIOTA FAPESP Araçá 2015). In Brazil, these contributions have been used to estimate the biomass and to subsidize the modelling, as it is the case of demersal (Haimovici & Velasco 2000, Nascimento et al. 2012) and small pelagic ichthyofauna (Cergole & Dias Neto 2011, Vaz-dos-Santos & Rossi-Wongtschowski 2013). In order to assess the growth pattern of the fish species and to provide an essential tool

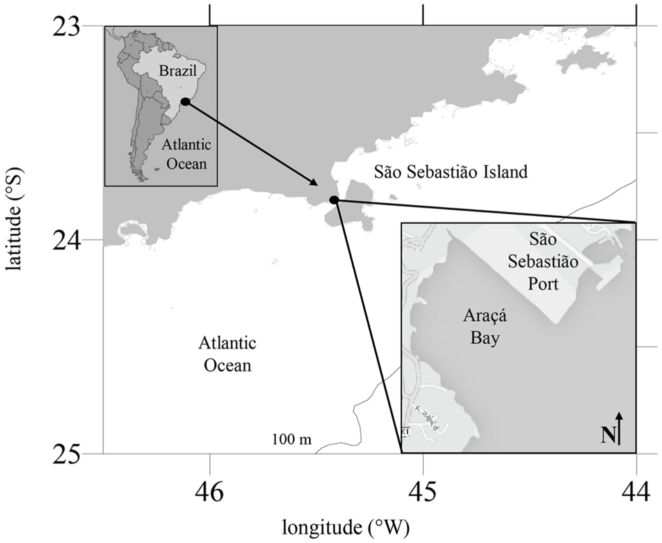

Figure 1. Map locating the Araçá Bay in the Brazilian coast, a coastal subtropical ecosystem.

to estimate the biomass with data from the area itself, the present study aimed to estimate the length-weight relationships (LWR) of the ichthyofauna in the Araçá Bay.

Material and Methods

Five samples (October 2012, March 2013, July 2013, October 2013 and January 2014) were attained by using nine different fishing gears, ensuring ontogenetic representativeness of the ichthyofauna. Fish species were identified, measured (total length, L_T, 0.1 mm) and weighed (total weight, W_T, 0.001 g). Nomenclature followed Eschmeyer (2015). Data from samples were pooled and the potential model $W_T = aL_T^{\,b}$ (Huxley 1993) was fitted through the non-linear iterative least squares method (Zar 2010). Fits were assessed through residual analysis and the coefficient of determination values, calculated as $r^2 = [\Sigma(y_p - y_a)^2 / \Sigma(y_o - y_a)^2]$, where y_p is the predicting weight for the individual *i*, y_a is the average weight and y_o is the observed weight for the individual *i* (Vieira 2006). Growth pattern (whether isometric or allometric in relation to the referential value 3) was verified through *t* confidence intervals (CI) of *b* estimates: every value inside the CI is statistically similar to the estimate. The proportion of young and adult fishes in the sampling was informed, thus allowing checking the life stage represented by the regressions. The young fish were those assigned as immature (never spawned) and those ones in the other phases as adults, in accordance to Brown-Peterson et al. (2011).

Results and Discussion

A total of 12,362 specimens belonging to 39 species, 21 families and 11 orders were analysed (Table 1). The allometric coefficient (*b*) varied between 2.55 and 3.97 (mean = 3.086, median = 3.096). The variation of the coefficient of determination (between 0.759 and 0.999, mean = 0.956, median = 0.978) and residual analysis ensured the acuity of regressions even in the cases in which r^2 values were reduced by the biological variability. Especially in these cases, it is important to highlight that these models represent the portion of the population and their condition in the Araçá Bay and they should be used for biomass estimates in this particular situation. Although almost all length ranges were represented in the sampling, the proportional contribution of small and young fishes (59% of species) was higher than that in adults and longer fishes (31% of species). The *Sardinella brasiliensis*, *Trachinotus* spp., *Caranx latus* and *Umbrina coroides* sampling were constituted only by young fishes, while for *Archosargus rhomboidalis*, *Menticirrhus americanus*, *Eugerres brasilianus*, *Cynoscion jamaicensis* and *Gymnothorax ocellatus* adults dominated the sampling. The remaining 10% corresponded to 3 species in which it was not possible to evaluate the life stage and one in which the proportion was exactly 1:1 (*Diapterus rhombeus*).

Isometric growth pattern was detected in 17 species, positive allometry in 16 and negative allometry in 6 species (Figure 2). While the coefficient *a* is the condition factor varying due to many factors related to the fish biology, physiology and body shape (Braga 1986, Froese 2006), the coefficient *b* represents mainly the growth pattern usually varying between 2.5 and 3.5 (Froese 2006). In *G. ocellatus* such upper limit was exceeded, and this was due both to the anguilliform body (Moyle & Cech 2004) and the narrow amplitude of lengths (Froese 2006).

Consulting the FishBase (Froese & Pauly 2015), for *Albula vulpes*, *Trachinotus carolinus*, *T. falcatus*, *Archosargus rhomboidalis* and *Kyphosus sectatrix*, these are the first records of LWR in Brazil. In spite of the availability of LWR parameters for the other species (Froese & Pauly 2015), only Muto *et al.* (2000) studied the ichthyofauna in the same area, but outside the coastal environments. These authors used mm-g and comparisons with other studies in cm-g can be done after the conversion using the equation of Froese (2006). Muto et al. (2000) provided LWR for 57 species based on samples attained from the continental shelf adjacent to Araçá Bay between 1993 and 1997, but only 18 species (Gerreidae, Haemulidae, Sciaenidae, flounders and some others) were the same ones. In comparison to this study, differences in the LWR were found mainly for the Gerreidae family with lower *b* amounts. Such heterogeneity in the species composition and growth pattern (mainly isometry and positive allometry) ensure that Araçá Bay is a growth ground for the ichthyofauna of the area revealed by the high amounts of *b* coefficients. Only *Ctenogobius boleosoma* and *Etropus crossotus* are resident species; the others go to the Araçá Bay to feed and grow (Vaz-dos-Santos et al. 2015).

In comparison with other LWR studies of the Southwestern Atlantic, it is possible to verify that Araçá Bay is shared by the ichthyofauna from different habitats. The continental shelf is dominated by the demersal sciaenids (Vianna et al. 2004), pelagic clupeiforms and carangids (Vaz-dos-Santos & Rossi-Wongtschowski 2013). In coastal environments (mangroves, rockpools), gerreids, gobiids and *Atherinella brasiliensis* usually predominate (Macieira & Joyeux 2009, Costa et al. 2014). Differences among LWR of these studies were expected due to different fishing gears, areas and periods, especially when it is considered the space (and time) scale of the present study. In such context, aiming the biomass estimates and ecosystem modelling, the present results are the most suitable: they are from the area itself and the use of nine fishing gears reduces (almost cancelled) selectivity. Data and results are representing properly and along a cycle (year) the different development phases of fishes using the Araçá Bay.

The presence of the young-of-the-year of the Brazilian sardine, *Sardinella brasiliensis*, in the Araçá Bay is noteworthy. This is the

Table 1. Number of fishes (n), total length range, parameters and *t* confidence intervals (CI) of the potential model ($W_T = aL_T^{\,b}$), coefficient of determination, relative growth pattern (i = isometric; a = allometric; + = positive; - = negative) and percentage of individuals by life stage.

Order	Family	Species		n	Total lenght (mm)			*a*	±	CI	*b*	±	CI	r²	growth pattern	% of life stage	
					minimun	average	maximum									youngs	adults
Albuliformes	Albulidae	*Albula vulpes*	(Linnaeus, 1758)	60	32	78	210	$6.571.10^{-6}$	±	$2,201.10^{-6}$	3,05	±	0,063	0,984	i	78	22
Anguiliformes	Muraenidae	*Gymnothorax ocellatus*	Agassiz, 1831	38	332	402	522	$4.169.10^{-9}$	±	$0,100.10^{-10}$	3,97	±	0,419	0,933	a+	16	84
Clupeiformes	Engraulidae	*Anchoa tricolor*	(Spix & Agassiz, 1829)	111	35	73	103	$7.670.10^{-6}$	±	$4,756.10^{-6}$	2,99	±	0,142	0,931	i	71	29
	Clupeidae	*Harengula clupeola*	(Cuvier, 1829)	654	39	74	212	$6.331.10^{-6}$	±	$7,854.10^{-7}$	3,12	±	0,025	0,983	a+	97	3
		Sardinella brasiliensis	(Steindachner, 1879)	538	43	78	116	$1.306.10^{-6}$	±	$3,929.10^{-7}$	3,41	±	0,057	0,990	a+	100	---
Aulopiformes	Synodontidae	*Synodus foetens*	(Linnaeus, 1766)	77	56	169	238	$2.990.10^{-6}$	±	$2,191.10^{-6}$	3,13	±	0,137	0,990	i	66	34
Mugiliformes	Mugilidae	*Mugil curema*	Valenciennes, 1836	1272	25	129	452	$1.672.10^{-5}$	±	$3,335.10^{-6}$	2,90	±	0,033	0,986	a-	73	27
Atheriniformes	Atherinopsidae	*Atherinella brasiliensis*	(Quoy & Gaimard, 1825)	1760	22	89	151	$4.542.10^{-6}$	±	$3,923.10^{-7}$	3,10	±	0,021	0,986	a+	42	58
Beloniformes	Hemiramphidae	*Hyporhamphus unifasciatus*	(Ranzani, 1841)	63	116	199	322	$2.562.10^{-7}$	±	$1,999.10^{-7}$	3,45	±	0,099	0,959	a+	34	66
Scorpaeniformes	Triglidae	*Prionotus punctatus*	(Bloch, 1793)	155	22	104	220	$1.394.10^{-5}$	±	$2,568.10^{-6}$	2,98	±	0,034	0,986	i	64	36
Perciformes	Centropomidae	*Centropomus parallelus*	Poey, 1860	24	200	318	403	$1.307.10^{-4}$	±	$2,610.10^{-4}$	2,55	±	0,342	0,896	a-	---	---
	Serranidae	*Diplectrum radiale*	(Quoy & Gaimard, 1824)	110	55	148	246	$2.645.10^{-6}$	±	$1,189.10^{-6}$	3,32	±	0,089	0,978	a+	49	51
	Carangidae	*Caranx latus*	Agassiz, 1831	37	59	171	225	$3.765.10^{-5}$	±	$4,056.10^{-5}$	2,82	±	0,208	0,956	i	100	---
		Chloroscombrus chrysurus	(Linnaeus, 1766)	50	38	118	210	$2.070.10^{-5}$	±	$1,166.10^{-5}$	2,85	±	0,108	0,999	a-	57	43
		Oligoplites saurus	(Bloch & Schneider, 1801)	211	20	54	254	$3.194.10^{-6}$	±	$3,943.10^{-7}$	3,17	±	0,022	0,999	a+	94	6
		Trachinotus carolinus	(Linnaeus, 1766)	91	18	34	62	$1.488.10^{-5}$	±	$6,755.10^{-6}$	2,99	±	0,112	0,999	i	94	6
		Trachinotus falcatus	(Linnaeus, 1758)	115	18	37	65	$3.511.10^{-5}$	±	$1,347.10^{-5}$	2,87	±	0,100	0,865	a-	100	---
		Trachinotus goodei	Jordan & Evermann, 1896	31	62	97	125	$1.139.10^{-5}$	±	$1,634.10^{-5}$	3,01	±	0,294	0,968	i	100	---
	Lutjanidae	*Lutjanus analis*	(Cuvier, 1828)	31	39	203	410	$1.296.10^{-5}$	±	$7,750.10^{-6}$	3,01	±	0,101	0,999	i	64	36
		Lutjanus synagris	(Linnaeus, 1758)	38	54	120	316	$3.779.10^{-5}$	±	$1,054.10^{-5}$	2,81	±	0,049	0,999	a-	77	23
	Gerreidae	*Diapterus rhombeus*	(Cuvier, 1829)	1038	14	124	258	$3.999.10^{-6}$	±	$5,887.10^{-7}$	3,27	±	0,033	0,971	a+	50	50
		Eucinostomus argenteus	Baird & Girard, 1855	2552	17	76	189	$1.049.10^{-5}$	±	$2,157.10^{-6}$	3,03	±	0,045	0,982	i	60	40
		Eucinostomus gula	(Quoy & Gaimard, 1824)	163	73	137	205	$6.515.10^{-6}$	±	$3,159.10^{-6}$	3,16	±	0,093	0,967	a+	51	49
		Eucinostomus melanopterus	(Bleeker, 1863)	87	28	174	230	$1.177.10^{-5}$	±	$1,392.10^{-5}$	3,02	±	0,231	0,951	i	36	64
		Eugerres brasilianus	(Cuvier, 1830)	35	152	194	269	$8.614.10^{-6}$	±	$1,016.10^{-5}$	3,10	±	0,226	0,991	i	11	89
	Haemulidae	*Haemulon steindachneri*	(Jordan & Gilbert, 1882)	56	99	154	258	$1.573.10^{-5}$	±	$8,818.10^{-6}$	2,99	±	0,105	0,968	i	71	29
		Orthopristis ruber	(Cuvier, 1830)	256	54	84	148	$1.953.10^{-5}$	±	$5,317.10^{-6}$	2,94	±	0,058	0,978	i	70	30
		Haemulopsis corvinaeformis	(Steindachner, 1868)	1477	46	120	178	$8.432.10^{-6}$	±	$1,177.10^{-6}$	3,11	±	0,028	0,973	a+	34	66
	Sparidae	*Archosargus rhomboidalis*	(Linnaeus, 1758)	46	16	135	191	$9.168.10^{-6}$	±	$5,438.10^{-6}$	3,14	±	0,120	0,999	a+	---	100
	Sciaenidae	*Ctenosciaena gracilicirrhus*	(Metzelaar, 1919)	187	42	107	145	$3.765.10^{-6}$	±	$1,973.10^{-6}$	3,26	±	0,104	0,892	a+	59	41
		Cynoscion jamaicensis	(Vaillant & Bocourt, 1883)	109	124	192	269	$6.382.10^{-6}$	±	$6,343.10^{-6}$	3,13	±	0,188	0,829	i	14	86
		Menticirrhus americanus	(Linnaeus, 1758)	61	28	234	425	$5.671.10^{-6}$	±	$5,201.10^{-6}$	3,13	±	0,156	0,984	i	5	95
		Umbrina coroides	Cuvier, 1830	38	70	111	185	$1.871.10^{-6}$	±	$1,418.10^{-6}$	3,38	±	0,145	0,999	a+	100	---
	Gobiidae	*Ctenogobius boleosoma*	(Jordan & Gilbert, 1882)	145	20	39	55	$1.706.10^{-5}$	±	$1,384.10^{-5}$	2,81	±	0,213	0,860	i	---	---
	Kyphosidae	*Kyphosus sectatrix*	(Linnaeus, 1758)	30	23	237	462	$5.829.10^{-6}$	±	$5,522.10^{-6}$	3,20	±	0,156	0,993	a+	---	---
Pleuronectiformes	Paralichthyidae	*Citharichthys spilopterus*	Günther, 1862	101	40	112	199	$2.704.10^{-6}$	±	$7,936.10^{-7}$	3,26	±	0,061	0,969	a+	63	37
		Etropus crossotus	Jordan & Gilbert, 1882	387	30	89	143	$2.907.10^{-6}$	±	$7,864.10^{-7}$	3,28	±	0,054	0,759	a+	28	72
		Syacium papillosum	(Linnaeus, 1758)	38	52	112	155	$1.104.10^{-5}$	±	$1,216.10^{-6}$	3,01	±	0,216	0,972	i	79	21
Tetraodontiformes	Tetraodontidae	*Sphoeroides greeleyi*	Gilbert, 1900	90	43	87	174	$1.126.10^{-4}$	±	$5,921.10^{-5}$	2,64	±	0,109	0,869	a-	46	54

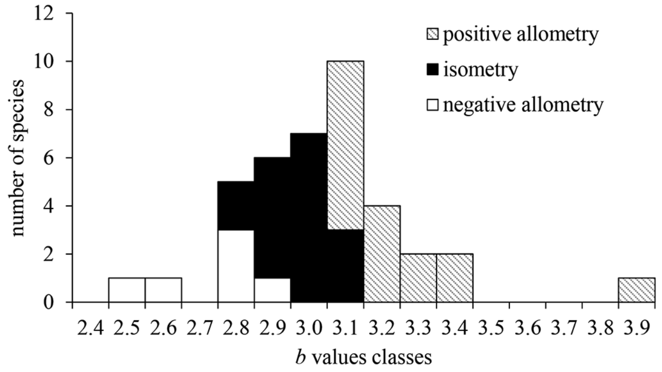

Figure 2. Frequency distribution of *b* values for 39 species caught in the Araçá Bay, a coastal subtropical ecosystem.

most important fishery resource in Brazil (MPA 2011), spawning along the continental shelf (Matsuura 1998). The displacement to a coastal ecosystem indicates the importance of Araçá Bay to the recruitment of the species.

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The South-American rattlesnake *Crotalus durissus*: feeding ecology in the central region of Brazil

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Abstract: We investigated food patterns in the diet composition of 452 specimens of *Crotalus durissus* from Central Brazil. Thirty-three items were recorded corresponding to four categories: rodents (75.76%), marsupials (6.06%), unidentified mammals (9.09%) and reptiles (9.09%). Adults of both sexes and some juveniles feed mostly on mammals, specifically rodents, it is the most active and abundant prey throughout the year mainly in areas from Cerrado. In addition, we observed that in *C. durissus* there is a trend in the diet of females could be more diverse, maybe this can be associated to sexual differences involving different strategies of the feeding biology of this rattlesnake.
Keywords: *diet composition, mammal specialization, lizards, Cerrado.*

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Resumen: Patrones de alimentación fueron investigados en la composición de la dieta de 452 ejemplares de *Crotalus durissus* de Brasil central. Treinta y tres ítems fueron registrados, correspondientes a cuatro categorías: roedores (75.76%), marsupiales (6.6%), mamíferos no identificados (9.09%) y reptiles (9.09%). Los adultos de ambos sexos e individuos jóvenes se alimentaron básicamente de mamíferos, en específico de roedores, esta es la presa más activa y abundante durante todo el año, principalmente en las áreas de Cerrado. Además fue observado que en *C. durissus* existe una tendencia a que la dieta de las hembras sea más diversa, factor que podría estar relacionado a las diferencias sexuales que implican diferentes estrategias de alimentación en la biología de esta serpiente cascabel.
Palabras clave: *composición de la dieta, especialista en mamíferos, lagartos, Cerrado.*

Introduction

The rattlesnakes of the genus *Crotalus* evolved in North America and subsequently spread across Central and South America (Echevarrigaray et al. 2000; Quijada-Mascareñas et al. 2007). Currently 41 species of *Crotalus* are recognized, with greater a diversity in Mexico and the United States (Uetz at al. 2016). This genus is frequently considered as a model for studies related to ecology, due to its widespread distribution covering several habitats, including deserts, flooded areas, forest environments and open habitats (Norman, 1994; Beaupre et al.1998; Place & Abramson, 2004).

In many species of *Crotalus* there is ontogenetic variation in diet, with juveniles feeding primarily on ectothermic prey whereas adults prefer endothermic animals as food. Moreover, there is extensive inter- and intraspecific variation in the diet of rattlesnakes. The smaller species, feed primarily on arthropods, amphibians and lizards whereas larger species feed on lizards and small mammals (Klauber 1956, 1972). For instance, adult *Crotalus horridus* from several regions of the United States (Clark 2002) as well as *C. viridis* in southeast Idaho (Diller & Wallace 1996) feed mainly on birds and small mammals. *Crotalus vegandris* from Venezuela (Pífano & Rodriguez-Acosta 1996), *C. pricei*, and *C. lepidus* from some areas in Mexico and the United States (Holycross et al. 2002; Prival et al. 2002), feed on centipedes, lizards, and snakes. However, a divergence in the diet composition was detected in two populations of *C. lepidus* at Big Bend National Park, Texas (Beaupre 1995). In a similar way, the diet of *C. viridis oreganus* from California is more diverse when compared to *C. viridis* from British Columbia, mainly because of the greater importance of ectothermic prey (Macartney 1989).

The South-American Rattlesnake *Crotalus durissus*, is restricted to South America (Campbell & Lamar 2004) and has a discontinuous distribution (Wüster et al. 2005) from Colombia to Argentina (Vanzolini et al. 1980). Some populations exhibit considerable ecological variation, with closeby populations differing greatly from each other (Campbell & Lamar, 2004).

In Brazil, *Crotalus durissus* is widely distributed, except for the states of Acre and Espírito Santo. In addition, there are isolated populations in open areas within savannas of the Amazonian Forest (Campbell & Lamar 1989). The diet of the species is assumed to be characterized by an extreme specialization in endothermic prey; a feature that exists for only a few species of rattlesnake and which is thought to represent an ancestral trait (Clark 2002). Studies of food composition in populations from southeastern Brazil indicated that rodents and small marsupials were the prey more widely eaten by rattlesnakes (Sant'Anna & Abe 2007)**;** however, birds (Vanzolini et al. 1980) and lizards (Santos & Germano 1996), can be considered as occasional food items in the *C. durissus* diet. In this work, we describe the feeding ecology of *Crotalus durissus* in the **c**entral **r**egion of Brazil and discus some strategies involved in the feeding of this rattlesnake.

Material and Methods

The snakes examined represent a total of 452 individuals of *Crotalus durissus* from the **c**entral **r**egion of Brazil covering forest formations of the Amazon and Atlantic forests, Caatinga and Cerrado; identified as 213 males, 167 females and 72 neonates and juveniles. We considered specimens to be neonates when their snout-vent length was smaller than 400mm (Hoyos, 2012). The specimens are housed in the scientific collections of Instituto Butantan (IBSP), Museu de Zoologia da Universidade de São Paulo (MZUSP)**,** and Universidade de Brasília (CHUNB). Animals kept in captivity prior to being deposited in a scientific collection were excluded from this study.

All food items were removed from the stomach and identified to the lowest possible taxonomic level. Each of the contents was deposited in a glass container with the same identification number as the specimen (voucher number). The intestinal content was determined while taking into account the type of food residue. In addition, available literature records of prey were included in this study, which analyzed the food composition for *Crotalus durissus* from southeastern Brazil (Santos & Germano 1996; Sant'Anna & Abe 2007).

Results

Thirty-three items were recorded in 30 (7%) stomachs. Approximately 52 (12%) of the intestines contained hair. Four prey categories were identified in the stomachs, the main being rodents 75.76% (Figure 2), marsupials 6.06%, followed by unidentified mammals 9.09% and reptiles 9.09% (Figure 3). The rodents were registered in almost all forest formations studied; marsupials were registered only in Cerrado sites, and lizards in areas of Caatinga and Cerrado (Figure 1) (Appendix 1). In addition, these results show a slight trend towards increased consumption of lizards in females. Evidence for ontogenetic or sexual variation was not detected (Table 1).

Overall, the diet composition of the South-American Rattlesnake *Crotalus durissus*, considering information of stomach contents identified from specimens in this study in combination with literature records (Santos & Germano 1996; Sant'Anna & Abe 2007), show a similar feeding patterns: rodents 66.05%, marsupials 3.98%, unidentified mammals 28.38% and reptiles 1.59%; in reference to the reptiles, only six specimens - four of which identified in females had reptiles in their stomach.

Discussion

Considering data to other viperids (Sawaya et al. 2008; Marques et al. 2009; Barbo et al. 2011) the frequency of individuals of *Crotalus durissus* with prey in the stomach was low.

The elapsed time between capture of the snake and its preservation probably is a reason for the low number of prey in the stomach. This is also supported by Sant'Anna & Abe 2007, where the stomach content of *C. durissus* from southeaster Brazil showed a much lower proportion than the gut content.

Approximately 90 percent of rattlesnakes of the genus *Crotalus*, exhibit ontogenetic shifts in food composition, changing from ectothermic to endothermic preys. These variations could be attributed to changes in morphological, behavioral and physiological characteristics during ontogeny (Mushinsky 1987). Alternatively a small percentage of rattlesnakes feed almost exclusively on endothermic preys, like *C. durissus* (Salomão et al. 1995; Duarte 2003), *C. horridus* and *C. molossus* (Clark 2002). Klauber (1956) gathered these three species and another, *C. basiliscus*, in the "*durissus* group", to demonstrate this specialization.

Mammal specialization may be related to several life history strategies (Martins et al. 2002). In some species of pitvipers, it could be associated with the increased venom toxicity in juveniles; this may be a consequence of the need to immobilize larger prey such as a mammal (Andrade & Abe 1999). Furthermore, the adoption of a juvenile diet based on mammals is more profitable energetically than an ecthothermic diet (Martins et al. 2002). In *Crotalus durissus* populations, the low frequency of ectothermic prey could explain such eventual facts.

Given this, behavioral and physiological changes necessary to switch from finding and consuming relatively sedentary ectotherms to finding and consuming fast, active endotherms might be costly (Clark 2002). Moreover, the microhabitat to which similar species are adapted to may not be used by local populations of reptiles that could serve as appropriate prey (Reinert 1984).

Several authors, point out the endothermic preys as a determinant element in food composition of *C. durissus*. For example, Beebe (1946) indicated that a specimen of this rattlesnake from Guyana contained a spiny rat. In populations from Uruguay, Achavál et al. (1978) reported that this rattlesnake commonly feeds on *Cavia pamparum*. Sant'Anna & Abe (2007), studying populations of southeastern Brazil, we observed that some species of rodents of the Caviidae and Muridae families followed by marsupials of the group Didelphidae, were the items most frequently consumed in this region.

In the current study, the diet composition of *C. durissus* was similar, characterized by a greater proportion of rodents. This mammals' group is terrestrial, nocturnal and dwell in open areas but they can also be found in forests (Emmons & Feer 1990; Nowak 1991). Similarly, in Cerrado endothermic prey types are rich and abundant, about 194 species of mammals are recognized, in which 51 are rodents and 17 marsupials (Marinho-Filho et al. 2002). This number has increased with the description of new species and the expansion of occurrence area of others (Bonvicino et al. 2003; Bonvicino et al. 2010).

In *C. durissus*, juveniles and adults feed predominantly on rodents (Sant'Anna & Abe 2007), a pattern attributed to the overall availability of this type of prey (Salomão et al. 1995). Field work confirmed an absence of ontogenetic variation in the diet which indicated that the juveniles of this rattlesnake could survive in dry locations with a supply

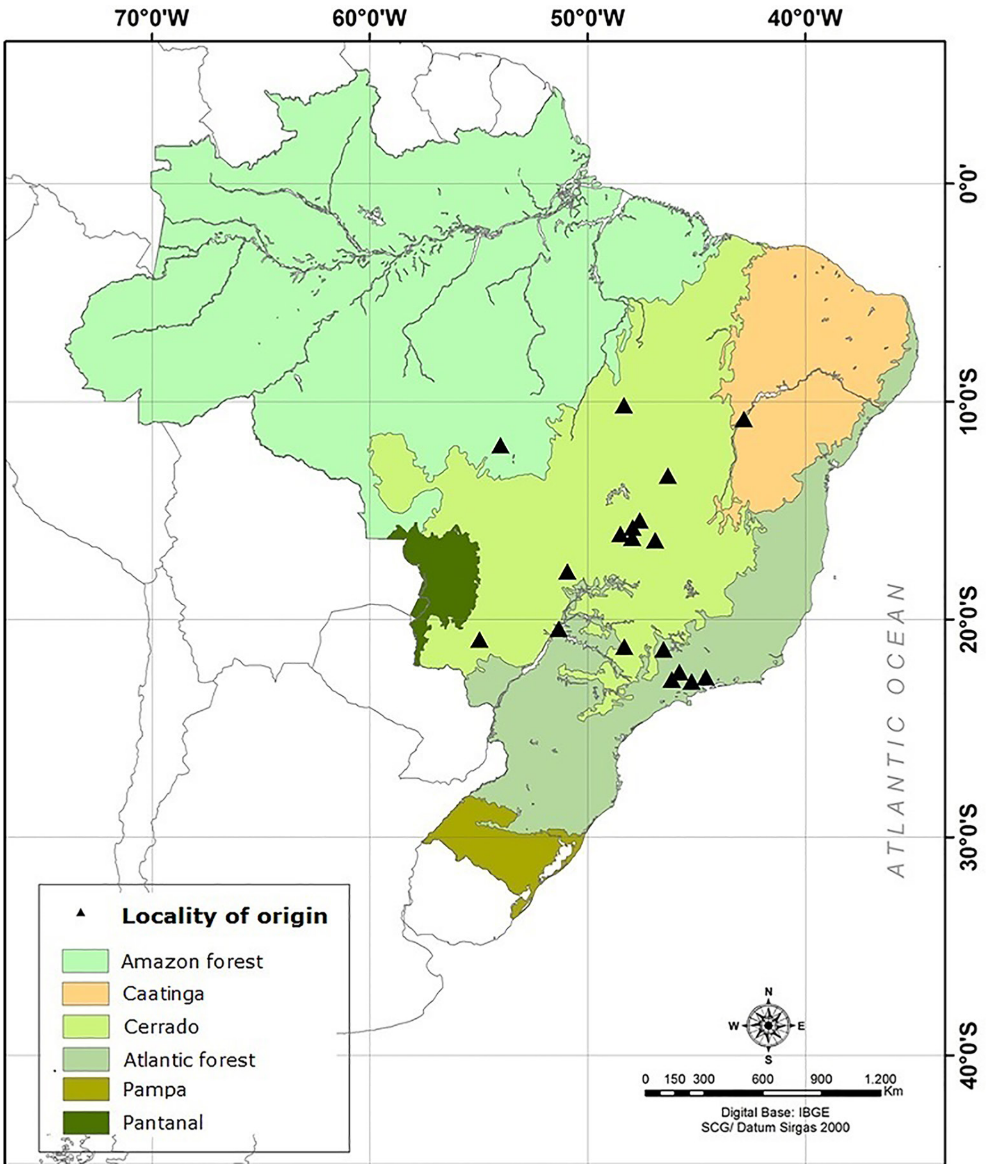

Figure 1. Geographical distribution of *Crotalus durissus* with stomach contents from Central Region of Brazil covering forest formations of Amazon and Atlantic forest, Caatinga and Cerrado.

Figure 2. Food category (rodent) of *Crotalus durissus* from Distrito Federal.

Figure 3. Food item, Ameiva ameiva, recorded in the stomach of a female *Crotalus durissus*. Specimen deposited in the Coleção Herpetológica da Universidade de Brasília (CHUNB 49673).

Table 1. Food composition in individual males, females, newborns and juveniles of *Crotalus durissus* from Central Region of Brazil (*n* = 30 snakes).

Categories of preys	Males	Females	Newborns/Juveniles	F	F%
Mammalia					
Not identified		3		3	9.09
Rodentia	9	10	6	25	75.76
Marsupialia					
Didelphidae	1		1	2	6.06
Reptilia					
Teiidae					
Ameiva ameiva		2	1	3	9.09
Total	10	15	8	33	100

of rodents (Tozetti & Martins 2008). The consumption of rodents in all age classes and both sexes is favored because this prey is the most abundant throughout the year. Yet teid lizards (*Ameiva*) (Santos & Germano 1996) can be a sporadic food item in rattlesnakes. The teid *Ameiva ameiva*, is relatively abundant in Cerrado sites (Araujo & Almeida-Santos 2012) and even near human settlements (Colli et al. 2002), usually occupying similar habitats to *Crotalus durissus*. This could be one of the reasons that this species is more frequently consumed rather than other ectothermic prey.

Additionally, some factors such as adaptations related to competition, niche utilization, foraging strategies, costs of time and energy and prey availability (Schoener 1969; Shine 1986; Shine et al. 2002), could reflect important characteristics in the understanding of feeding ecology of *C. durissus* from the central region of Brazil.

Appendix 1

Collection sites with voucher numbers of studied specimens of *Crotalus durissus* and stomach contents from Central Region of Brazil. CHUNB, Coleção Herpetológica da Universidade de Brasília; IBSP, Instituto Butantan; MZUSP, Museu de Zoologia da Universidade de São Paulo.

BAHIA: Ibiraba (MZUSP 10076) - one lizard. **DISTRITO FEDERAL**: Brasília (CHUNB 5439, 22100) - three rodents. **GOIÁS**: Alexânia (CHUNB 20432) - one unidentified mammal; Luziânia (CHUNB 20468) - one rodent; Planaltina (CHUNB 20444, 20446, 20467) - three rodents; Rio verde (CHUNB 49673, IBSP 11997) - one lizard and one marsupial; São Domingos (CHUNB 15499) - one unidentified mammal. **MINAS GERAIS**: Camanducaia (CHUNB 67995) - one rodent; Conceição dos Ouros (IBSP 68493) - one rodent; Muzambinho (CHUNB 24288) - one rodent; Unaí (CHUNB 20460, 20475-76, 24379) - one lizard, one unidentified mammal and two rodents. **MATO GROSSO**: Parque Xingú (CHUNB 28739) - one rodent. **MATO GROSSO DO SUL**: Ilha Solteira (IBSP 38087) - one rodent; Sidrolândia (IBSP 23922) - one rodent. **SÃO PAULO**: Aparecida do Norte (IBSP 73254) - one rodent; Jaboticabal (IBSP 23682) - one rodent; São José do Barreiro (IBSP 71480 - 71482) - three rodents. **TOCANTINS**: Palmas (CHUNB 14690) - one rodent. In relation to three specimens from Cerrado, two of them contained two rodents respectively and the other one marsupial; for these snakes the collection sites were undetermined.

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Headwater stream fish fauna from the Upper Paranapanema River basin

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CETRA M., MATTOX, G.M.T., FERREIRA, F.C., GUINATO, R.B., SILVA, F.V., PEDROSA, M. **Headwater stream fish fauna from the Upper Paranapanema River basin**. Biota Neotropica. 16(3): e20150145. http://dx.doi.org/10.1590/1676-0611-BN-2015-0145

Abstract: The Paraná River basin has about 600 fish species. In the Upper Paraná, 310 fish species were recorded, with 52 species were registered in the streams and headwaters of the Paranapanema River. The aim of this study was to characterize the stream fish communities in the Upper Paranapanema River basin. Samplings were conducted with electrofishing during the dry season in the year of 2014. The collection effort consisted of 30 streams stretches. As a result, 41 species of stream fish were recorded in the Upper Paranapanema River basin, distributed in 26 genera, 11 families and 7 orders. Thirty-nine percent of the species can be considered rare, 41% intermediate and 20% common. We captured approximately eight species by stream stretch and the estimated richness (Schao2) was 40 ± 6 species. Around 40% of the individuals had less than 50 mm in length.
Keywords: *fish communities, estimated richness, rare species.*

CETRA M., MATTOX, G.M.T., FERREIRA, F.C., GUINATO, R.B., SILVA, F.V., PEDROSA, M. **Ictiofauna de riachos das cabeceiras da bacia hidrográfica do Alto Paranapanema**. Biota Neotropica. 16(3): e20150145. http://dx.doi.org/10.1590/1676-0611-BN-2015-0145

Resumo: A bacia hidrográfica do Rio Paraná possui cerca de 600 espécies de peixes. No Alto Rio Paraná, 310 espécies de peixes foram registradas com 52 espécies registradas em riachos do Rio Paranapanema. O objetivo deste estudo foi caracterizar a comunidade de peixes de riachos da bacia do Alto Rio Paranapanema. O levantamento das espécies foi realizado com uso de pesca elétrica durante a estação seca de 2014. As coletas ocorreram em 30 trechos de riachos. Foram capturas 41 espécies distribuídas em 26 gêneros, 11 famílias e 7 ordens. Trinta e nove por cento das espécies podem ser consideradas raras, 41% intermediárias e 20% comuns. Foram capturadas cerca de 8 espécies por trecho de riacho e a riqueza estimada (Schao2) foi 40 ± 6 espécies. Cerca de 40% dos indivíduos possuem comprimento menor que 50 mm.
Palavras-chave: *comunidades de peixes, riqueza estimada, espécies raras.*

Introduction

The Paraná River basin is the second largest in area in the Neotropical region (Agostinho & Júlio Jr. 1999), with about 2.6 million km^2. Older estimates suggest that this basin has nearly 600 fish species (Bonetto 1986). Recently, 310 fish species were recorded in the Upper Paraná (Langeani et al. 2007), which comprises approximately 900,000 km^2 and the portion of this basin in São Paulo State includes 260 species along the Paraná River and its main tributaries, Paranapanema, Tietê and Grande (Oyakawa & Menezes 2011). About 65% of these species were small sized (less than 21 cm), and occurred in streams and headwaters (Langeani et al. 2007). Recent inventories documented a high diversity fish fauna for the Upper Paraná (Casatti et al. 2001, Castro & Casatti 1997, Castro et al. 2003, 2004, 2005, Langeani et al. 2005a b, Fagundes et al. 2015).

As recommended by Casatti et al. (2008), this study intended to perform the following actions:

(i) survey the fish populations in the Upper Paranapanema River basin, which currently presents a knowledge gap;
(ii) study the fish community from streams and headwaters that are especially interesting for sharing species with neighboring basins;
(iii) provide information on the fish populations that can be used in the monitoring of riparian forests restoration projects.

This study aimed to characterize the stream fish communities in the Upper Paranapanema River and supplement the species lists of Castro et al. (2003).

Material and Methods

1. *Study area*

The Upper Paranapanema River basin is located in the Unidade de Gerenciamento dos Recursos Hídricos [Water Resources Management Unit] (UGRHI 14). This unit presents agriculture activity and population density around 30 inhabitants/km^2. About 15% of the area is covered by native vegetation, and the headwaters are covered by reforested area (*Eucaliptus* spp.) and native forests (CETEC 1999). The headwater streams are in the Serra de Paranapiacaba, at about 1100 m altitude, and are tributaries of the Paranapanema River, which, after running along 500 km, will flow into the Paraná River.

2. *Fish Sampling*

Fish sampling was conducted from June to November 2014 as the associations between fish assemblage and environmental structure are better described in the dry season (Pinto et al. 2006). Furthermore, it is important to control the effect of sources of temporal variation.

The ichthyofauna sampling was performed with electrofishing between 10h and 16h (License SISBIO 13352-1/IBAMA/MMA). We collected the fish fauna in 30 streams stretches with 70 m. These streams are located in 13 watersheds in the municipalities of Piedade, Pilar do Sul, São Miguel Arcanjo, Capão Bonito, Ribeirão Grande, Guapiara, Apiaí, Ribeirão Branco, Itapeva, Itaberá, Itararé and Sengés (Figure 1, Table 1), covering stretches of 1st to 6th orders (Hydrology ArcGIS software tool).

The wadeable stream stretches have at least one sequence of riffle-run-pool meso-habitats. The organisms collected were fixed for 48h in 4% formalin and then transferred to 70% alcohol. Vouchers of the species collected were deposited in the collection of Laboratório de Ictiologia of the Departamento de Zoologia e Botânica da UNESP - São José do Rio Preto (DZSJRP 20.268 - 20.663) and of Laboratório de Ictiologia de Ribeirão Preto - FFCLRP/USP (LIRP 11826). The specimens were identified according to Castro et al. (2003), Castro & Vari (2004), Vari & Castro (2007), Lucinda (2008), Zawadzki et al. (2008), Lippert et al. (2014) and with the assistance of specialists in some groups.

Results and Discussion

We captured about 7 kg of fish: 1878 individuals, 41 species, 26 genera, 11 families and 7 orders (Table 2). Following the criteria of Teshima et al. (2015), 16 species (39%) can be considered rare, 17 species (41%) are intermediate and 7 species (20%) are common. On average, we captured approximately 8 species by stream stretch and the estimated richness (Schao2) was 40 ± 6 species. Around 40% of individuals had less than 50 mm in length.

The orders Siluriformes and Characiformes represented the majority of the species richness, 44% and 37% respectively, reflecting a well-known pattern recognized for South American rivers (Lowe-McConnell 1999). Twelve species are not on the list of species that occur in the Upper Paraná (Langeani et al. 2007), and 18 are not in the list by Castro et al. (2003) that collected fish in streams of the Paranapanema basin.

Some species have been listed with different names as presented in Castro et al. (2003) and Langeani et al. (2007) (Table 2, with asterisk). This happened because some species that had been recognized as distinct morphotypes were later formally described. This is the case of *Astyanax* sp1 and *Astyanax* sp2 (Castro et al. 2003), later described as *A. bockmanni* and *A. biotae* (Castro & Vari 2004, Vari & Castro 2007). A *Bryconamericus* morphotype with humeral spot was identified only at the genus level (Castro et al. 2003). Langeani et al. (2007) identified a morphotype of *Bryconamericus* with humeral spot as *B. iheringii*, highlightening that this could probably represent a new species. Also, these previous lists included *Phalloceros caudimaculatus* as a single species, but we found herein both species currently recognized in the genus for the Paranapanema basin (see below).

The São Paulo State fish fauna is relatively well known (Langeani et al. 2007, Oyakawa & Menezes 2011). However, there are still many taxa to be better studied and that probably represent new species. It is known, for instance, that *Astyanax scabripinnis* represents a group of species, some of which still undescribed, and further detailed analysis based on larger and more complete sample series may result in the description new species (e.g., Moreira-Filho & Bertollo 1991, Bertaco & Malabarba 2001, Bertaco & Lucena 2006, Fagundes et al. 2015).

Neoplecostomus selenae, *Phalloceros reisi* and *P. harpagos* were described in 2008 but were already cited as new species under description by Langeani et al. (2007). *Otothyropsis biamnicus* was recently described (Calegari et al. 2013) and included in a recent species list of the Upper Paraná (Fagundes et al. 2015).

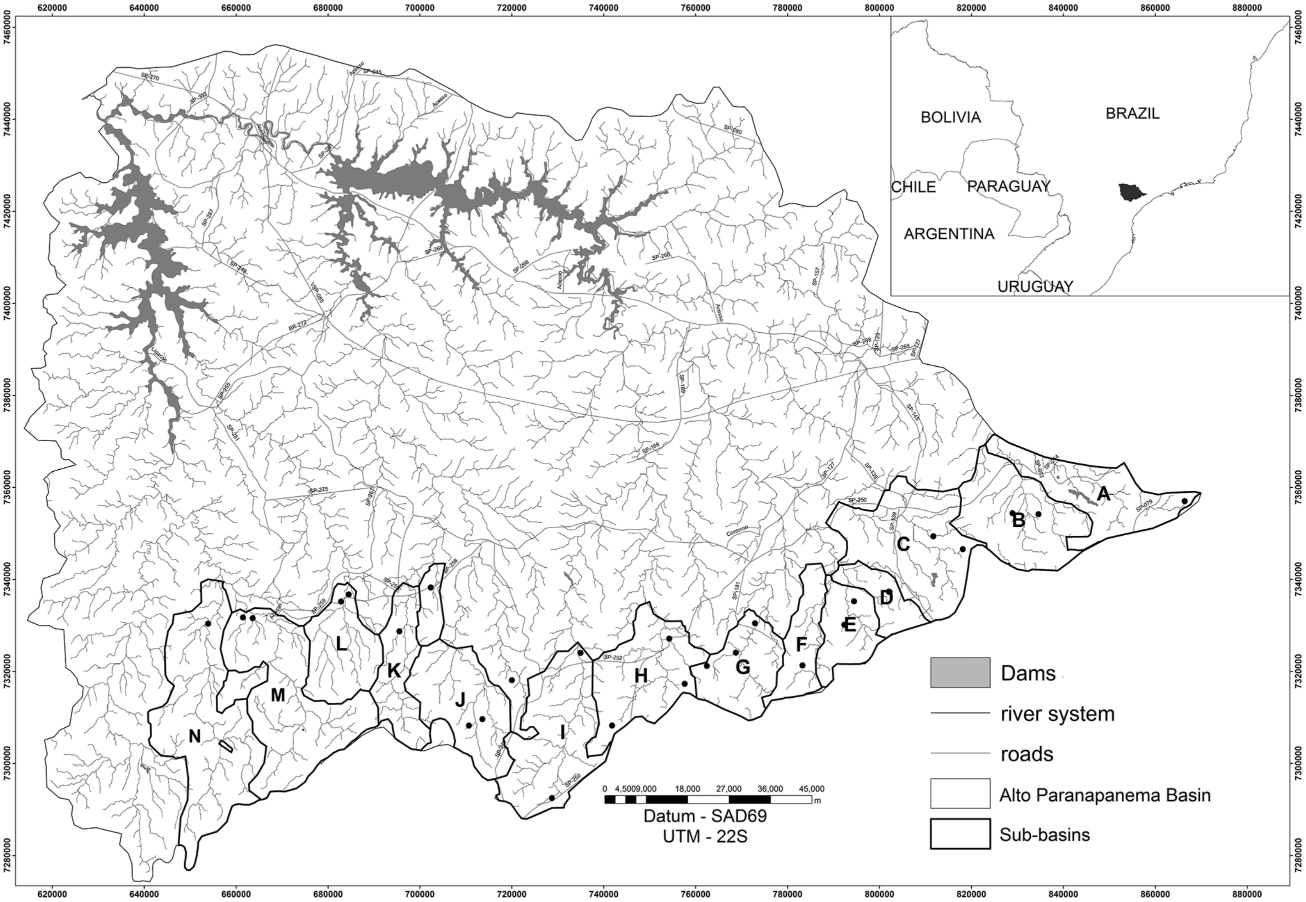

Figure 1. Sub-basins and stretch streams sampled: A (Turvo), B (Pinhal), C (Guarapú), D (Taquaral), E (Guapiara), F (Paranapanema), G (Almas), H (Apiaí-Mirim), I (Apiaí-Guaçu), J (Taquari-Guaçu), K (Pirituba), L (Verde), M (Itararé), N (Jaguaricatu).

Table 1. Geographical information of the stretch streams sampled.

Stretch	Stream	Sub-basin	Zone	X	Y	Municipality
P03	Turvo	Turvo	23	253797	7358842	Piedade
P17	Clarinho	Pinhal	23	223828	7355389	Pilar do Sul
P19	Pinhal	Pinhal	23	217944	7355372	Pilar do Sul
P23	Guarapu	Guarapu	23	207407	7347068	São Miguel Arcanjo
P24	Lageado	Guarapu	23	203595	7342984	São Miguel Arcanjo
APP01	Taquaral	Taquaral	22	802182	7337329	Capão Bonito
APP05	Grande	Taquaral	22	802002	7337026	Capão Bonito
APP04	Cristal	Guapiara	22	795363	7335461	Capão Bonito
APP25	Guapiara	Guapiara	22	790520	7330282	Capão Bonito
APP24	Guapiara tributary	Guapiara	22	790430	7330199	Capão Bonito
APP02	Panema	Paranapanema	22	783224	7321220	Capão Bonito
APP23	Conchas	Paranapanema	22	775540	7323894	Capão Bonito
APP03	Almas	Almas	22	768502	7324070	Ribeirão Grande
APP12	Bateia	Almas	22	762266	7321196	Ribeirão Grande
APP22	Pinheiros	Apiaí-Mirim	22	757668	7317396	Guapiara
APP13	Alegre	Apiaí-Mirim	22	754209	7327285	Guapiara
APP11	São José do Guapiara	Apiaí-Mirim	22	741811	7308275	Guapiara

Continued Table 1.

APP21	Afluente Apiaí Guaçu	Apiaí-Guaçu	22	734904	7324251	Ribeirão Branco
APP10	Apiai-Guaçu	Apiaí-Guaçu	22	728684	7292451	Apiaí
APP14	Taquari-Mirim	Taquari-Guaçu	22	720031	7318116	Ribeirão Branco
APP19	Afluente Taquari1	Taquari-Guaçu	22	713707	7309647	Ribeirão Branco
APP20	Afluente Taquari2	Taquari-Guaçu	22	710646	7308221	Ribeirão Branco
APP06	Papagaio	Pirituba	22	695536	7328740	Itapeva
APP18	Afluente Pirituba	Pirituba	22	702365	7338237	Itapeva
APP15	Da Gurita	Verde	22	684567	7336707	Itaberá
APP08	Verde	Verde	22	682849	7335189	Itararé
APP07	Afluente Verde	Verde	22	682786	7335250	Itararé
APP09	Funil	Itararé	22	663580	7331572	Sengés
APP16	Pelame	Itararé	22	661498	7331784	Sengés
APP17	Afluente Jaguaricatu	Jaguaricatu	22	653926	7330379	Sengés

Table 2. Species sampled in the headwater streams from Upper Paranapanema River basin with respective status. X: species listed in Castro et al. (2003) and/or Langeani et al. (2007). Asterisk indicates species listed with a different name in previous papers.

	Status	**Castro et al. (2003)**	**Langeani et al. (2007)**
Cypriniformes			
Cobitidae			
Misgurnus anguillicaudatus (Cantor, 1842)	rare	-	-
Characiformes			
Characidae			
Astyanax bockmanni Vari & Castro, 2007	common	X*	X*
Astyanax paranae Eigenmann, 1914	intermediate	-	X
Astyanax biotae Castro & Vari, 2004	rare	X*	X
Astyanax scabripinnis (Jenyns, 1842)	common	X	-
Bryconamericus stramineus Eigenmann, 1908	rare	X	X
Bryconamericus aff. *iheringii* (Boulenger, 1887)	rare	-	X
Hyphessobrycon anisitsi (Eigenmann, 1907)	rare	-	X
Oligosarcus paranensis Menezes & Géry, 1983	rare	X	X
Piabina argentea Reinhardt, 1867	intermediate	X	X
Crenuchidae			
Characidium gomesi Travassos, 1956	intermediate	X	X
Characidium schubarti Travassos, 1955	intermediate	-	X
Characidium aff. *zebra* Eigenmann, 1909	intermediate	X	X
Erythrinidae			
Hoplias malabaricus (Bloch, 1794)	intermediate	X	X
Parodontidae			
Apareiodon ibitiensis Campos, 1944	Rare	-	X
Parodon nasus Kner, 1859	Rare	-	X
Gymnotiformes			
Gymnotidae			
Gymnotus sylvius Albert & Fernandes-Matioli, 1999	Rare	X	X
Sternopygidae			
Eigenmannia virescens (Valenciennes, 1836)	Rare	X	X

Continued Table 2.

Siluriformes			
Heptapteridae			
Cetopsorhamdia iheringi Schubart & Gomes, 1959	Intermediate	X	X
Imparfinis borodini Mees & Cala, 1989	intermediate	-	X
Imparfinis mirini Haseman, 1911	Common	X	X
Phenacorhamdia tenebrosa (Schubart, 1964)	Rare	X	X
Pimelodella avanhandavae Eigenmann, 1917	Common	-	X
Rhamdia quelen (Quoy & Gaimard, 1824)	intermediate	X	X
Loricariidae			
Hypostomus ancistroides (Ihering, 1911)	Common	X	X
Hypostomus nigromaculatus (Schubart, 1964)	intermediate	X	X
Hypostomus sp1	Rare	-	-
Hypostomus sp2	Intermediate	-	-
Neoplecostomus selenae Zawadzki, Pavanelli & Langeani, 2008	Common	-	-
Otothyropsis biamnicus Calegari, Lehmann & Reis, 2013	Rare	-	-
Otothyropsis sp.	Rare	-	-
Rineloricaria pentamaculata Langeani & Araújo, 1994	intermediate	X	X
Trichomycteridae			
Trichomycterus davisi (Haseman, 1911)	intermediate	-	-
Trichomycterus diabolus Bockmann, Casatti & de Pinna, 2004	Rare	-	X
Trichomycterus sp1	intermediate	-	-
Trichomycterus sp2	intermediate	-	-
Cyprinodontiformes			
Poecilidae			
Poecilia vivipara Bloch & Schneider, 1801	Rare	-	X
Phalloceros reisi Lucinda, 2008	Common	*	*
Phalloceros harpagos Lucinda, 2008	intermediate	*	*
Synbranchiformes			
Synbranchidae			
Synbranchus marmoratus Bloch, 1795	intermediate	X	X
Labriformes			
Cichlidae			
Geophagus brasiliensis (Quoy & Gaimard, 1824)	Common	X	X

The species mentioned above were described in the last 10 years, hence discovered in relative recent times. In addition, some taxa could not be identified to species level, probably because they represent new taxa awaiting description. As noted by Langeani et al. (2007), this may be the case of *Hypostomus* sp1, *Hypostomus* sp2, *Trichomycterus* sp1, *Trichomycterus* sp2 and *Othothyropsis* sp, which should contribute to the increase in the species richness from the Upper Paraná.

The exotic species *Misgurnus anguillicaudatus* had been registered in the Iguaçu River basin (Ingenito et al. 2004, Vitule 2009) and was recently captured in preserved streams in the Ribeira de Iguape River basin (Gomes et al. 2011, Cetra et al. 2012). We herein present the first record of this species in the Paranapanema River basin in the São Paulo State.

Among the 41 fish species sampled in the present study, we highlight *Neoplecostomus selenae*, a species listed as vulnerable on the list of endangered species in São Paulo State (Oyakawa et al. 2009). This species was included in that list because it was known solely from the type locality in Ribeirão das Bateias basin, Ribeirão Grande municipality (Zawadzki et al 2008). For the conservation of this species, Oyakawa et al. (2009) suggested habitat protection and scientific research to obtain data on its biology, and inventories to better access the distribution of this species. The results in the present study expanded the area of occurrence of *N. selenae* (Apiaí-Guaçu, Apiaí-Mirim, Guapiara, Itararé, Panema, Taquaral, Taquari-Guaçu and Verde sub-basins) yielding new specimens for further studies.

The present study complemented data on fish populations of the Upper Paraná River basin, specifically of the tributary streams of the Paranapanema River (e.g., Castro et al. 2003, Langeani et al. 2007), representing a considerable increase in the richness in the latter drainage, in addition to a few putative new species. Although the fish fauna of the Paraná River basin is relativelky well known (eg, Langeani et al. 2007, Oyakawa

& Menezes 2011), several areas of this drainage system still remain unexplored and represent gaps in the knowledge of this ichthyofauna.

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Composition and diversity of anurans in the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil

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Abstract: The Conde municipality is located in the northern coast of the state of Bahia (NC), northeastern Brazil, and is part of the Atlantic Tropical domain. The anuran fauna of the northern portion of the NC is still poorly known if compared to the southern portion. The Restinga is one of the predominant environments of the coastal plains of the NC and it is characterized essentially by presenting sandy soil covered by herbaceous and shrubby vegetation. The objective of this study was to determine the anuran species composition and diversity for the Restinga of the Conde municipality. Sampling was carried out at night by active search over four periods of five consecutive days each, two over the 'main rainy season' and two in a 'lesser rainy season', using 14 sample units (SUs) and five extra sample plots (EPs). We calculated dominance and species diversity using the Berger-Parker and Shannon-Wiener H' indices, respectively. We used accumulation curves and the Jackknife 1 estimator to estimate anuran species richness, considering only the data obtained from the SUs. We recorded 713 anuran specimens distributed within 33 species, 13 genera and five families (Bufonidae, Craugastoridae, Hylidae, Leptodactylidae and Microhylidae). The Hylidae and Leptodactylidae families had the highest species richness. Considering only the SUs (Jackknife 1 estimator in brackets), we recorded 28 species in the study area (33.9 ± 2.3), 13 in Shrubby Vegetation Zones - SVZ (20.8 ± 2.9) and 25 in Freshwater Wetland Zones - FWZ (28.9 ± 1.9). The abundance and species diversity of the FWZ (n = 638 specimens; H'= 2.4) were higher than those recorded for the SVZ (n = 52 specimens; H' = 1.9). The SVZ and FWZ showed distinct dominant species, wherein *Pristimantis paulodutrai* was the dominant species in SVZ and *Scinax fuscomarginatus* in FWZ. The Restinga of the Conde municipality stands out as the one with the highest anuran species richness already recorded considering only SVZ and FWZ. Moreover, its anuran species composition represented 55% of the anuran species known for the NC and included taxa common to three different morphoclimatic domains (Tropical Atlantic, Cerrado and Caatinga).
Keywords: *inventory, species richness, wetland zones, shrubby vegetation zones, coastal plains.*

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Resumo: O município de Conde situa-se no litoral norte do estado da Bahia (LN), nordeste do Brasil, e está inserido no domínio Tropical Atlântico. A fauna de anuros da porção setentrional do LN ainda é pouco conhecida se comparada à porção sul. A Restinga é um dos ambientes predominantes das planícies costeiras do LN, sendo caracterizada essencialmente por apresentar solo arenoso e vegetação herbácea e arbustiva. O objetivo deste estudo foi determinar a composição e a diversidade de espécies de anuros da Restinga do município de Conde. A amostragem foi realizada por procura ativa noturna, durante quatro períodos de cinco dias consecutivos cada, dois na estação ‘mais chuvosa’ e dois em estação ‘menos chuvosa’, utilizando-se 14 unidades amostrais (UAs) e cinco pontos amostrais extras (PEs). Calculamos a dominância e a diversidade de espécies utilizando os índices de Berger-Parker e de Shannon-Wiener-H’. Utilizamos curvas de acumulação de espécies e o estimador Jackknife 1 para estimar a riqueza de espécies de anuros, considerando-se somente os dados obtidos nas UAs. Registramos 713 espécimes de anuros distribuídos em 33 espécies, 13 gêneros e cinco famílias (Bufonidae, Craugastoridae, Hylidae, Leptodactylidae e Microhylidae). As famílias Hylidae e Leptodactylidae apresentaram as maiores riquezas de espécies. Considerando-se somente UAs (estimador Jackknife 1 entre parêntesis) foram registradas 28 espécies na área de estudo (33,9 ± 2,3), 13 nas zonas de moitas (20,8 ± 2,9) e 25 nas zonas úmidas (28,9 ± 1,9). A abundância e a diversidade de espécies para as zonas úmidas (n = 638 espécimes; H’ = 2,4) foram superiores às registradas para as zonas de moitas (n = 52 espécimes; H’ = 1,9). As zonas de moitas e úmidas exibiram espécies dominantes distintas, sendo *Pristimantis paulodutrai* a espécie dominante para as zonas de moitas e *Scinax fuscomarginatus* para as zonas úmidas. A Restinga do município de Conde desponta como aquela de maior riqueza de espécies de anuros já registrada considerando-se somente zonas de moitas e zonas úmidas. Além disso, sua composição de espécies de anuros representou 55% do total de espécies de anuros conhecidas para o LN e incluiu táxons comuns a três diferentes domínios morfoclimáticos (Tropical Atlântico, Cerrado e Caatinga).

Palavras-chave: *inventário, riqueza de espécies, zonas úmidas, zonas de moitas, planícies costeiras.*

Introduction

The northern coast of the state of Bahia (NC) is part of the Tropical Atlantic Domain (Ab’Sáber 1977) and is essentially covered by the Atlantic Forest, a global biodiversity hotspot (Myers et al. 2000). The Restinga is one of the environments that shape this domain and is continuously distributed along the 200 km of coastal plains of the NC. This is a Quaternary habitat, characterized by sandy soils with high salt concentration, and covered predominantly by herbaceous and shrubby xerophytic vegetation under direct sunlight, the ‘open Restinga’ of Xavier et al. (2015).

The fauna and flora of the Restingas may represent a mosaic of species coming from adjacent and contiguous ecosystems, such as the Atlantic Forest (e.g., Freire 1990; Araújo 1992; Carvalho-e-Silva et al. 2000). Concerning the Restingas of northeastern Brazil, Xavier et al. (2015) identified them as a biotic element of distinct nature regarding ecological and biogeographical distribution of the anuran fauna, including species from three distinct morphoclimatic domains (Cerrado, Caatinga and Tropical Atlantic domains), which is of great importance because almost all previously known natural biogeographical units in northeastern Brazil were related to forested environments. Although presenting singular biogeographical importance, the NC is under pronounced degradation of its natural areas due to human activities such as agriculture, forestry and tourism (INEMA 2015), mainly after the construction of the highway BA-099 that has led to the progressive conversion of Restingas into resort areas (Bastazini et al. 2007). Furthermore, the faunistic information available in the literature for the NC is concentrated in its southern portion and is derived from occasional and specific studies on certain themes of interest to researchers, many of them not formally published (M.F. Napoli, unpublished data). This is also the case for the amphibian fauna, for which studies are concentrated in its southern portion and emphasizing forest patches in the Mata de São João municipality (e.g., Juncá 2006, Bastazini et al. 2007, Dabés et al. 2012). Indeed, the amphibians of the northern limit of the NC are represented in herpetological collections mainly by specimens obtained from sporadic and occasional inventories, which points to the urgency in the development of faunistic and ecologic studies for the region. The lack of information about amphibians in their natural environments hinders conservation actions (Rocha et al. 2008), as is the case for the Restingas of the NC.

The aim of this study is to determine the composition and diversity (richness, dominance and relative abundance of species) of the anuran species in the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil, considering both shrubby vegetation and freshwater wetland zones.

Material and Methods

1. *Study Area*

The study was carried out in the Conde municipality, northern coast of the state of Bahia (NC), northeastern Brazil (Figure 1). The NC is bordered by the municipalities of Salvador and Lauro de Freitas to the south and by the municipality of Rio Real to the north. Along its length there are state environmental protection areas (APAs), but no full conservation units of integral protection. The APA of the North Coast of the state of Bahia - APA NC (“APA do Litoral Norte do Estado da Bahia”; State Decree number 1,046 of 17 March 1992) has the greatest extent (ca. 10 km wide and 200 km long) and covers, from south to north, the municipalities of Mata de São João, Entre Rios, Esplanada, Conde (the study area) and Jandaíra. The main environmental conflicts in the APA of NC are, currently, the disorderly land use, lack of basic sanitation, substitution of natural areas by *Pinus* and *Eucalyptus* plantations, indiscriminate intensification of cattle raising, overfishing, degradation of mangroves and predatory tourism in coastal districts (INEMA 2015). The municipalities of Esplanada, Conde and Jandaíra were considered by Silva et al. (2008) as those with the most conserved areas in the NC due to their smaller degrees of urbanization.

The Conde municipality covers a total area of approximately 965 km² (SEI 2015) and presents the widest coastal plain (314 km^2) of the northern coast of the state of Bahia (Esquivel 2006). This municipality is bordered to the north by the Jandaíra municipality, to the west by the Rio Real municipality, to the south by the Esplanada municipality and to the east by the Atlantic Ocean. The climate is

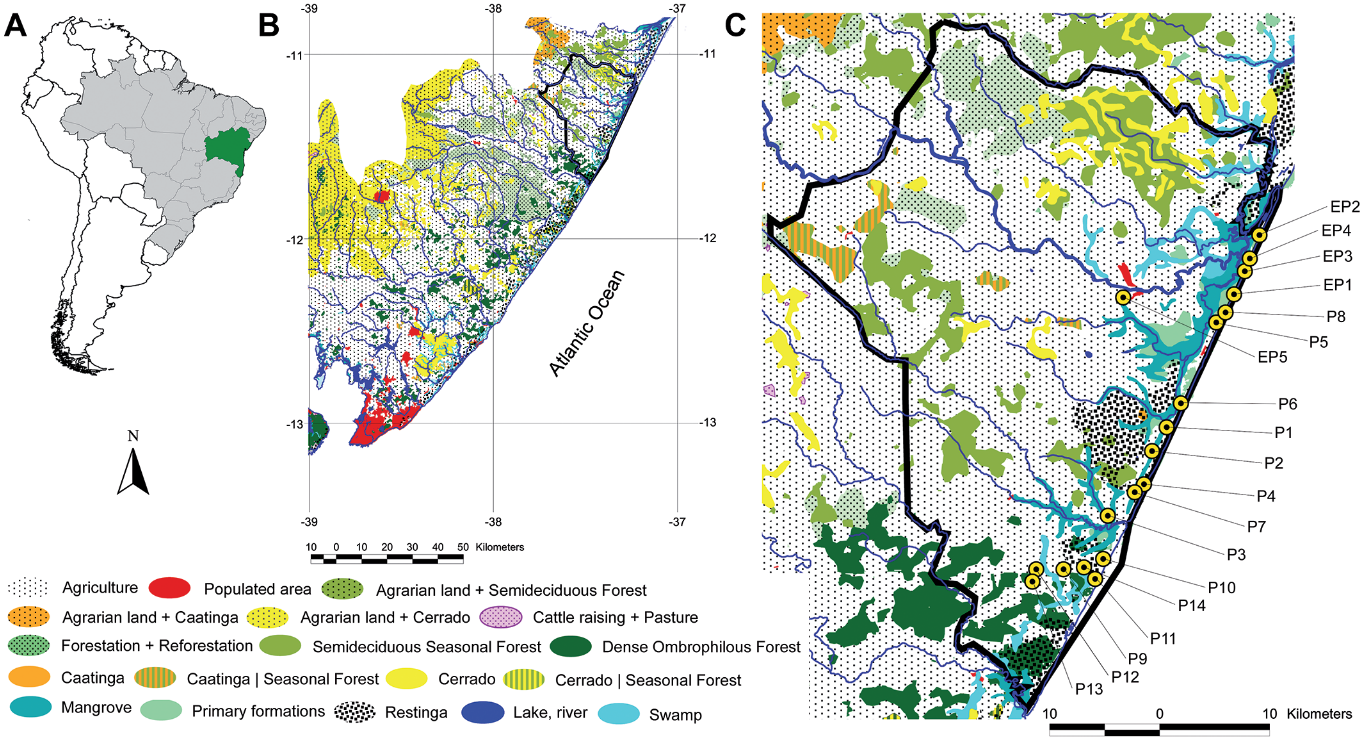

Figure 1. Distribution map of 14 sample units (SUs) and five extra sample plots (EPs) within the limits of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. (A) South America, Brazil in grey; (B) vegetation and hydrographic features of the northeastern state of Bahia, featuring the Conde municipality; (C) polygon of the Conde municipality, highlighting its vegetation, wetlands, Restingas, main types of anthropization, samples units (P1-P14) and extra sample plots (EP1-EP5). SUs/EPs from freshwater wetland zones, P4-P7, P12-P14, EP5. SUs/EPs from shrubby vegetation zones, P1-P3, P8-11, EP1-EP4.

classified by the Köppen climatic classification system as Tropical without dry season (Af). The average monthly temperature varies from 23°C to 27°C and the monthly rainfall from 61 to 264 mm, with more intense rainfall from April to July (Alvares et al. 2013).

The NC coastal plains are characterized as valleys partially filled with sandy deposits of marine and continental origins, in association with climate changes and marine transgressions and regressions during the Quaternary (Soares & Dominguez 2012). The sampled area studied herein was positioned alongside the coastline on areas of Holocene and Pleistocene deposits known as marine terraces (see Dominguez et al. 1999, Esquivel 2006, Nogueira 2006 and Bittencourt 2010 for geological-geomorphological descriptions and maps). Wetlands of a mosaic of shapes and sizes occur in the lowlands between these deposits, also present in the inland valleys associated with the Barreiras Formation of Tertiary age (Coastal Tablelands). This landscape holds a mosaic of different types of ecosystems, such as Restingas, wetlands (marshes and swamps), mangroves, dunes and riparian and ombrophilous forests (Esquivel 2006). Among them, we identified within the Restinga environment two main categories of physiognomies that could potentially be considered anuran habitats: shrubby vegetation zones and freshwater wetland zones (Figure 2). The shrubby vegetation zones (SVZ) were characterized by the presence of sandy soil covered by shrub and herbaceous vegetation, absence of water bodies, with or without terrestrial tank bromeliads. This physiognomy is equivalent to the 'shrub vegetation at the beach zone' of Bastazini et al. (2007) and to the 'Restinga em Moitas' of Menezes et al. (2009). The freshwater wetland zones (FWZ) are characterized by the presence of permanent or temporary freshwater bodies, with or without herbaceous and shrub vegetation on its margins. The wetlands sampled herein correspond to the freshwater marshes of Esquivel (2006) and Nogueira (2006), habitats dominated by herbaceous rather than woody plant species.

2. *Research Design and Field Methods*

For each physiognomy (SVZ and FWZ) we selected seven areas separated from one another by at least one kilometer to minimize biases due to pseudoreplication (Hurlbert 1984). In each sampling area a sample unit was established (SUs or Plots: SVZ P1-P3, P8-P11; FWZ P4-P7, P12-P14), totaling 14 SUs measuring 60 x 25 m each. In addition to these SUs, other five extra sample plots (EPs) were surveyed for anurans to complement the species inventory, four of them (EP1-EP4) located on the northernmost frontal dune near the Itapicuru river mouth and one located near the city of Conde (EP5). These extra sampling areas were not considered in the statistical analyses because they did not receive the same sampling effort designed for the SUs. Geographical coordinates of the SUs and EPs are in Table 1.

We sampled SUs in four periods of five consecutive days each: two periods over the main rainy season (15 to 21 May and 17 to 23 July 2015) and two in a lesser rainy period (10 to 16 September and 12 to 17 November 2015). To control for temporal biases in the characterization of the SUs, during each field campaign we distributed 50% of the SUs into shrubby vegetation zones and the other 50% into freshwater wetland zones. In the first and third sampling periods we sampled four SUs per physiognomy (P1-P8) and on the second and fourth sampling periods three SUs per physiognomy (P9-P14).

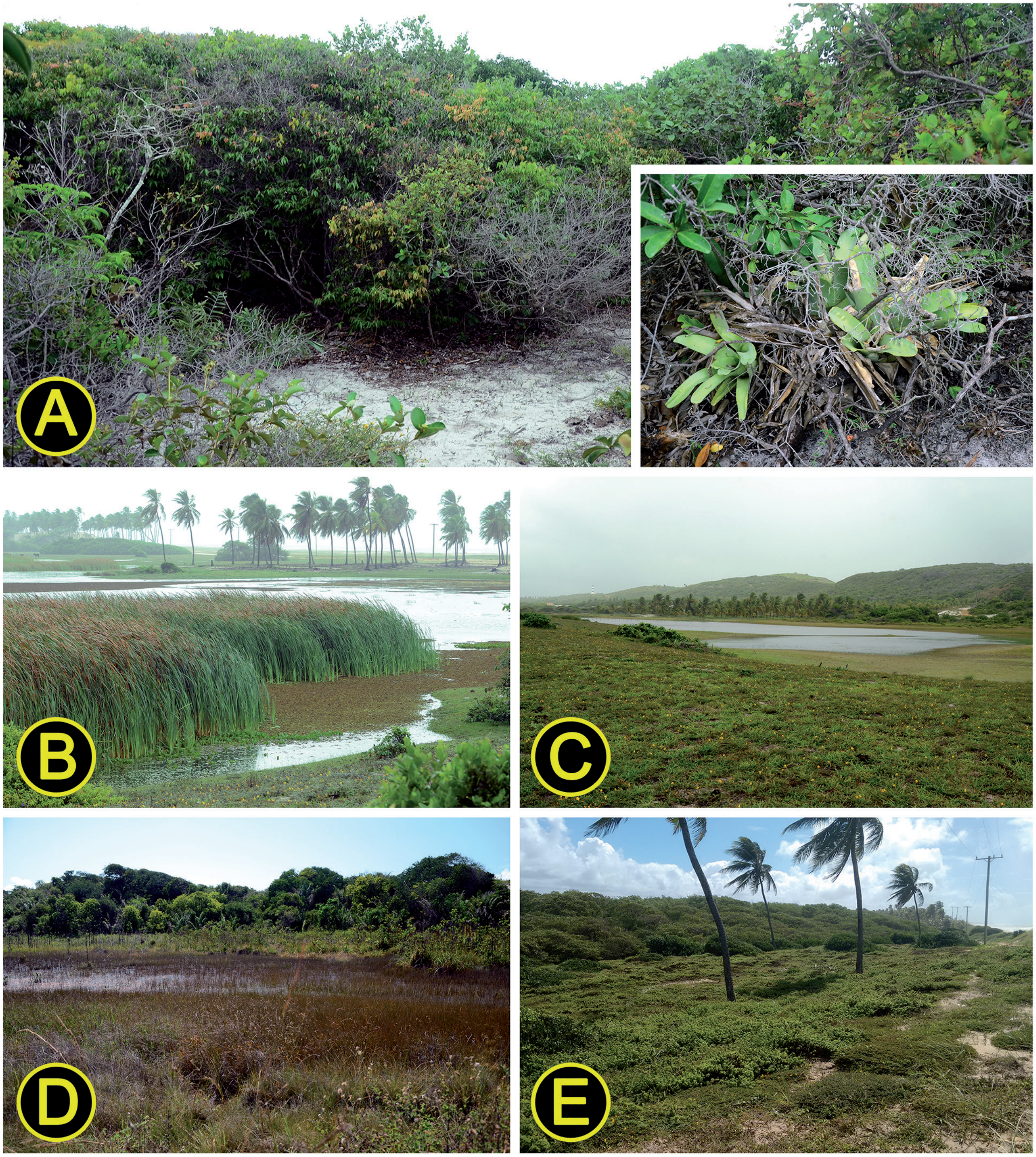

Figure 2. Landscapes in the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil, in which the sample units (SUs) and extra sample plots (EPs) were placed. (A) P1, a typical shrub zone, characterized by sandy soil covered by many shrubs and herbaceous vegetation; in detail at the bottom right corner terrestrial bromeliads, the main source of water and shelter for anuran species in shrubby vegetation zones; (B) P5, a marsh with reeds at its margins providing vegetal stratification; (C) P4, a freshwater wetland without vegetal stratification at its borders, characterized as a temporary pond on sandy soil the surroundings of which were covered by grasses; (D) P13, southernmost SU of the study area, placed at freshwater wetlands near forest remnants; (E) EP3, frontal dune near the northern coastal limits of the Conde municipality, featuring the shrub vegetation at the top of the image. Photographs: R.O. Abreu, 16 May 2015.

To obtain species composition, richness and abundance we sampled anuran species using only active search. We looked for anurans in each SU for 20 min a day, from 18:00 to 23:00 h, over five consecutive nights per campaign, totaling 500 min of active search per SU per field campaign. We covered each SU with homogeneous effort (i.e., without directing effort to a certain species or only vocalizing species) with a team of five researchers. All observers surveyed simultaneously each SU and the sequences of visitation to SUs were changed randomly during the sampling period. This procedure was performed in all campaigns for the 14 SUs, totaling 233 hours of sampling effort considering the four campaigns.

Table 1. Sample units (SUs: P1-P14) and extra sample plots (EP1-EP5) used in the survey of anurans at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil (Figure 1). Geographical coordinates in degrees, minutes and seconds, based on the WGS84 datum. Restinga physiognomies: FWZ, freshwater wetland zones; SVZ, shrubby vegetation zones.

SU/EP	Geographical coordinates	Physiognomy
P1	11°54’11.2”S; 37º35’30.3”W	SVZ
P2	11°55’8.8”S; 37°36’6.2”W	SVZ
P3	11º57’45.6”S; 37º37’52.8”W	SVZ
P4	11º56’29.2”S; 37º36’25.3”W	FWZ
P5	11º49’56.6”S; 37º33’29.6”W	FWZ
P6	11º53’13.1”S; 37º34’56.2”W	FWZ
P7	11º56’48.9”S; 37º36’47.7”W	FWZ
P8	11º49’33.2”S; 37º33’7.4”W	SVZ
P9	11º59’55.9”S; 37º39’41.3”W	SVZ
P10	11º59’30.1”S; 37º38’4.4”W	SVZ
P11	12º00’18.6”S; 37º38’23.9”W	SVZ
P12	11º59’56.4”S; 37º40’47.1”W	FWZ
P13	12º00’25.5”S; 37º40’55.8”W	FWZ
P14	11º59’51.2”S; 37º38’51”W	FWZ
EP1	11º48’48.9”S; 37º32’47” W	SVZ
EP2	11º46’25.3”S; 37º31’45.7”W	SVZ
EP3	11º47’53.3”S; 37º32’20.6”W	SVZ
EP4	11º47’22.1”S; 37º32’9.1”W	SVZ
EP5	11º48’55.9”S; 37º37’16.4”W	FWZ

Voucher specimens (n = 187) were collected and deposited in the Amphibian Collection of the Museu de Zoologia da Universidade Federal da Bahia (UFBA; Appendix 1), respecting the maximum number of individuals per species allowed by the collection license issued by the Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis (IBAMA). Specimens exceeding this maximum number were returned to nature after the sampling period was over in the same SUs where they were collected. Identification of the taxa was performed by analyzing the vocalizations and/or comparison with samples from the Museu de Zoologia da Universidade Federal da Bahia. We followed Frost (2015) for taxa nomenclature.

3. *Data Analysis*

For all statistical analyses we only considered anuran specimens captured in SUs. To estimate anuran species richness, we produced sample-based accumulation curves (*sensu* Gotelli & Colwell 2001) using 1000 randomizations on the order of samplings. Each night of observation corresponded to a sample, resulting in 70 samples per physiognomy and 140 samples for the overall study area. We used the species richness estimator Jackknife 1 (first order) to determine the expected richness of anurans (Magurran 2004). Both analyses were performed with EstimateS software, v. 9.1.0 (Colwell 2013). Species abundance distributions were analyzed visually through column charts. We used the Berger-Parker index as a measure of dominance (percentage of the most abundant species); the Gaston’s quartile criterion to define commonness and rarity (rare species are those that fall in the lower quartile of the species abundance distribution); the Shannon-Wiener index H’ to calculate species diversity; and the Pielou’s index J’ as an evenness measure (Magurran 2004). The latter analyses were performed with Past software, v. 2.15 (Hammer et al. 2001).

Results

We sampled a total of 713 individuals, distributed in five families (Bufonidae, Craugastoridae, Hylidae, Leptodactylidae and Microhylidae), 13 genera and 33 species (Table 2; Figures 3-6). Almost all species are categorized as Least Concern by the IUCN (2015), except for *Scinax cretatus* and *S. melanodactylus*, for which the conservation status has not yet been evaluated. The family Hylidae showed the highest species richness (20 species, 60.6%), followed by Leptodactylidae (9 species, 27.3%). The remaining families combined accounted for 12.1% of the species richness (4 species). Considering only the SUs, we recorded 690 individuals distributed in five families, 13 genera and 28 species, in which the most abundant species were *Scinax fuscomarginatus* (n = 145 individuals, 21%), *Pseudopaludicola mystacalis* (n = 121, 17.5%), *Leptodactylus macrosternum* (n = 66, 9.5%) and *Pleurodema diplolister* (n = 64, 9.2%). These four species accounted for 57.2% of the total number of sampled individuals (Figure 7). Three species were represented by one single individual (singletons): *Phyllodytes melanomystax*, *Phyllomedusa bahiana* and *Trachycephalus atlas*. Five species were represented by only two individuals (doubletons): *Dermatonotus muelleri*, *Hypsiboas faber*, *H. raniceps*, *Rhinella granulosa* and *Scinax eurydice*. In extra sample plots we recorded five additional hylid species (number of individuals in brackets): *Dendropsophus* cf. *oliveirai* (1), *D. elegans* (1), *Phyllomedusa nordestina* (18), *Physalaemus kroyeri* (2) and *Trachycephalus mesophaeus* (1). Only *P. bahiana* and *T. mesophaeus*

Table 2. Anuran species recorded for the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. Results obtained over four sampling periods in 2015: main rainy season (15 to 21 May and 17 to 23 July 2015); lesser rainy period (10 to 16 September and 12 to 17 November 2015). Categories of endemism in the Atlantic Forest biome (EN): 1, endemic, wide-range species; 2, endemic to northeastern Brazil; 3, endemic to the state of Bahia; W, wide distribution range in South America. Physiognomies (PY): FWZ, freshwater wetland zones; SVZ, shrubby vegetation zones. Sample units (SUs) and extra sample plots (EPs): shrubby vegetation zones, P1-P3, P8-P11, EP1-EP4; freshwater wetland zones, P4-P7, P12-P14, EP5. Breeding period (BP): MRS, main rainy season; LRS, lesser rainy period. Type of record (TR): Si, sighting; Ca, capture; Vo, vocalization. Number of individuals recorded (N).

Taxa	Popular Name	EN	PY	SU/EP	BP	TR	N
BUFONIDAE Gray, 1825							
1. *Rhinella granulosa* (Spix, 1824)	Sapo-granuloso	W	FWZ	P4	MRS	Ca/Vo	2
2. *Rhinela jimi* (Stevaux, 2002)	Sapo-cururu	EN 2	FWZ/SVZ	P2, P5, P12	LRS/MRS	Ca/Vo	10
CRAUGASTORIDAE Hedges, Duellman & Heinicke, 2008							
3. *Pristimantis paulodutrai* (Bokermann, 1975)	Rãzinha-do-folhiço	EN 2	FWZ/SVZ	P1-2, P7, P9-11, P14	LRS/MRS	Ca/Vo	26
HYLIDAE Rafinesque, 1815							
4. *Dendropsophus branneri* (Cochran, 1948)	Pererequinha	W	FWZ	P5, P7, P12	LRS/MRS	Ca/Vo	15
5. *Dendropsophus* cf. *oliveirai* (Bokermann, 1963)	Pererequinha	EN 3	FWZ	EP5	MRS	Ca	1
6. *Dendropsophus elegans* (Wied-Neuwied, 1824)	Perereca-de-moldura	EN 1	FWZ	EP5	MRS	Ca	1
7. *Dendropsophus minutus* (Peters, 1872)	Pererequinha-do-brejo	W	FWZ	P4, P12	MRS	Ca/Vo	3
8. *Dendropsophus nanus* (Boulenger, 1889)	Pererequinha-do-brejo	W	FWZ	P5-P7	LRS/MRS	Ca/Vo	15
9. *Dendropsophus rubicundulus* (Reinhardt & Lütken, 1862)	Pererequinha-verde	W	FWZ	P4, P6, P12	LRS/MRS	Ca/Vo	34
10. *Hypsiboas albomarginatus* (Spix, 1824)	Perereca-verde	EN 1	FWZ	P4, P12	LRS/MRS	Ca/Vo	3
11. *Hypsiboas faber* (Wied-Neuwied, 1821)	Sapo-martelo	W	FWZ	P4	MRS	Ca/Vo	2
12. *Hypsiboas raniceps* Cope, 1862	Perereca-zebrada	W	FWZ	P4	MRS	Ca/Vo	2
13. *Phyllodytes melanomystax* Caramaschi, Silva & Britto-Pereira, 1992	Pererequinha-de-bigode-de-bromélia	EN 2	SVZ	P2	MRS	Ca	1
14. *Phyllomedusa bahiana* Lutz, 1925	Perereca-folha	W	SVZ	P1	MRS	Vo	1
15. *Phyllomedusa nordestina* Caramaschi, 2006	Perereca-das-folhagens	W	FWZ	EP5	LRS/MRS	Ca/Vo	18
16. *Scinax auratus* (Wied-Neuwied, 1821)	Perereca-dourada	EN 2	FWZ	P4-6	LRS/MRS	Ca/Vo	8
17. *Scinax cretatus* Nunes & Pombal, 2011	Perereca-riscada-de-giz	EN 2	FWZ	P4, P6, P12, P14	LRS/MRS	Ca/Vo	37
18. *Scinax eurydice* (Bokermann, 1968)	Perereca-de-banheiro	W	FWZ	P4, P12	MRS	Ca/Vo	2
19. *Scinax fuscomarginatus* (Lutz, 1925)	Pererequinha-do-brejo	W	FWZ/SVZ	P1, P5-7, P12-14	LRS/MRS	Ca/Vo	145
20. *Scinax melanodactylus* (Lourenço, Luna & Pombal, 2014)	Perereca-de-bromélia	EN 2	FWZ/SVZ	P1, P3, P7, P12, P14	LRS/MRS	Ca/Vo	21
21. *Scinax x-signatus* (Spix, 1824)	Perereca-de-banheiro	W	FWZ/SVZ	P3, P4, P8	LRS/MRS	Ca/Vo	43
22. *Trachycephalus atlas* Bokermann, 1966	Perereca-de-capacete	EN 2	SVZ	P2	LRS	Ca	1
23. *Trachycephalus mesophaeus* (Hensel, 1867)	Perereca-grudenta	W	FWZ	EP5	LRS	Si	1
LEPTODACTYLIDAE Werner, 1896 (1838)							
24. *Leptodactylus fuscus* (Schneider, 1799)	Rã-assobiadora	W	FWZ/SVZ	P3-7, P12	LRS/MRS	Ca/Vo	33
25. *Leptodactylus macrosternum* Miranda-Ribeiro, 1926	Rã-manteiga	W	FWZ	P4-7, P12-14	LRS/MRS	Ca/Vo	66
26. *Leptodactylus natalensis* Lutz, 1930	Caçote-borbulhente	W	FWZ/SVZ	P5, P8	LRS/MRS	Ca/Vo	4
27. *Leptodactylus troglodytes* Lutz, 1926	Caçote/Rã-piadora	W	FWZ/SVZ	P2-4, P9	MRS	Ca/Vo	7
28. *Leptodactylus vastus* Lutz, 1930	Rã-pimenta	W	FWZ	P4, P12	MRS	Ca/Vo	7
29. *Physalaemus albifrons* (Spix, 1824)	Rã-cachorro	W	FWZ/SVZ	P3-5	LRS/MRS	Ca/Vo	15
30. *Physalaemus kroyeri* (Reinhardt & Lütken, 1862)	Rã-chorona	W	FWZ	EP5	LRS	Ca	2
31. *Pleurodema diplolister* (Peters, 1870)	Rãzinha-da-areia	W	FWZ/SVZ	P1, P4-5	LRS/MRS	Ca/Vo	64
32. *Pseudopaludicola mystacalis* (Cope, 1887)	Rãzinha-grilo	W	FWZ	P4, P6-7, P12, P14	LRS/MRS	Ca/Vo	121
MICROHYLIDAE Günther, 1858 (1843)							
33. *Dermatonotus muelleri* (Boettger, 1885)	Rã-manteiga	W	FWZ	P4	MRS	Ca/Vo	2

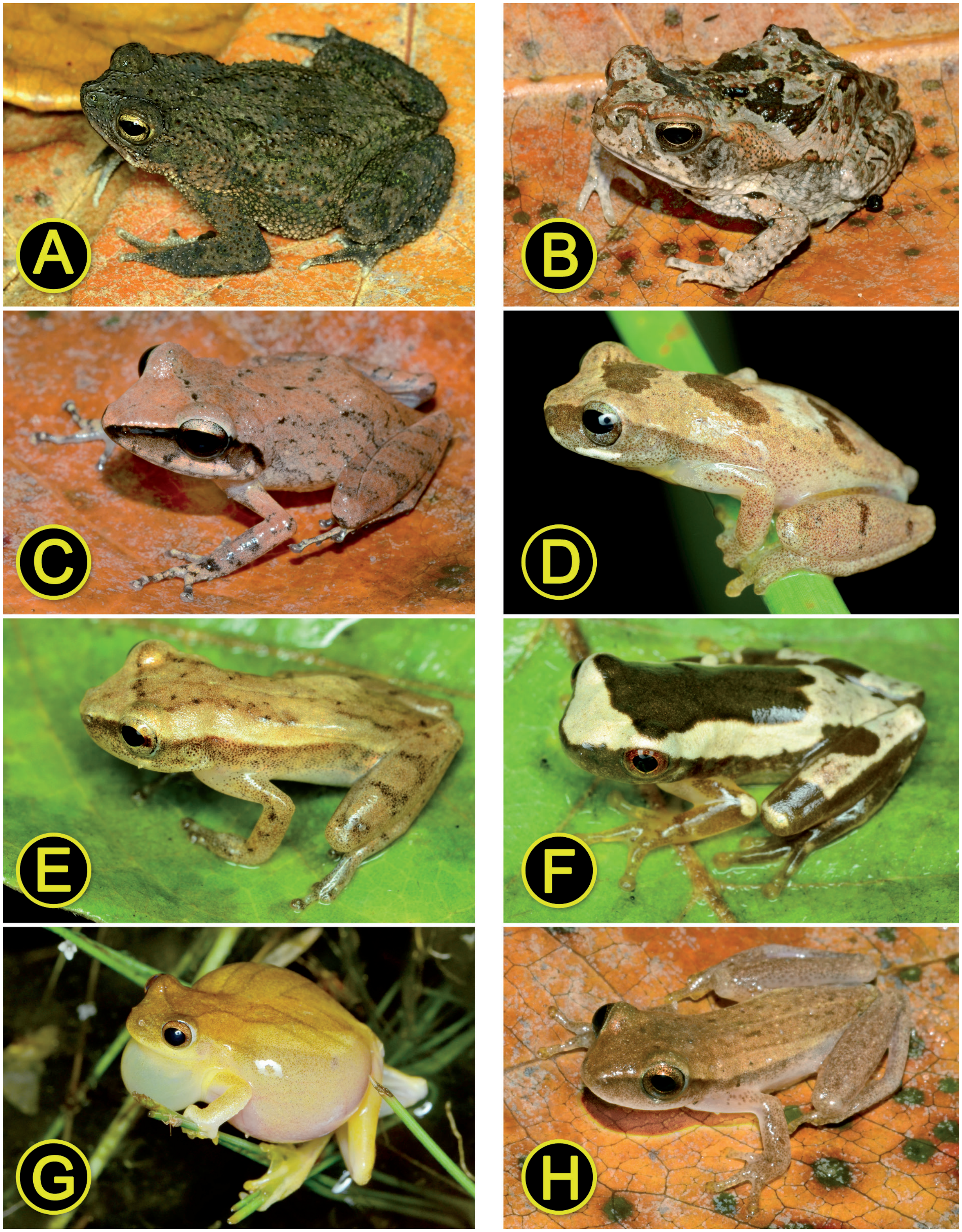

Figure 3. Anurans recorded at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. In brackets, for each vouchered specimen, is the acronym of the scientific collection followed by the respective institutional registration number and the specimen snout-vent length (SVL) in millimeters. (A) *Rhinella granulosa* (UFBA 13907; SVL 50.2); (B) *Rhinella jimi* (UFBA 13961; SVL 33.8); (C) *Pristimantis paulodutrai* (UFBA 13882; SVL 20.1); (D) *Dendropsophus branneri* (UFBA 13970; SVL 17.3); (E) *D.* cf. *oliveirai* (UFBA 14048; SVL 17.6); (F) *D. elegans* (UFBA 14044; SVL 12.0); (G) *D. minutus* (UFBA 13964; SVL 22.3); (H) *D. nanus* (UFBA 13973; SVL 21.7). Photographs: L. Menezes and R.O. Abreu.

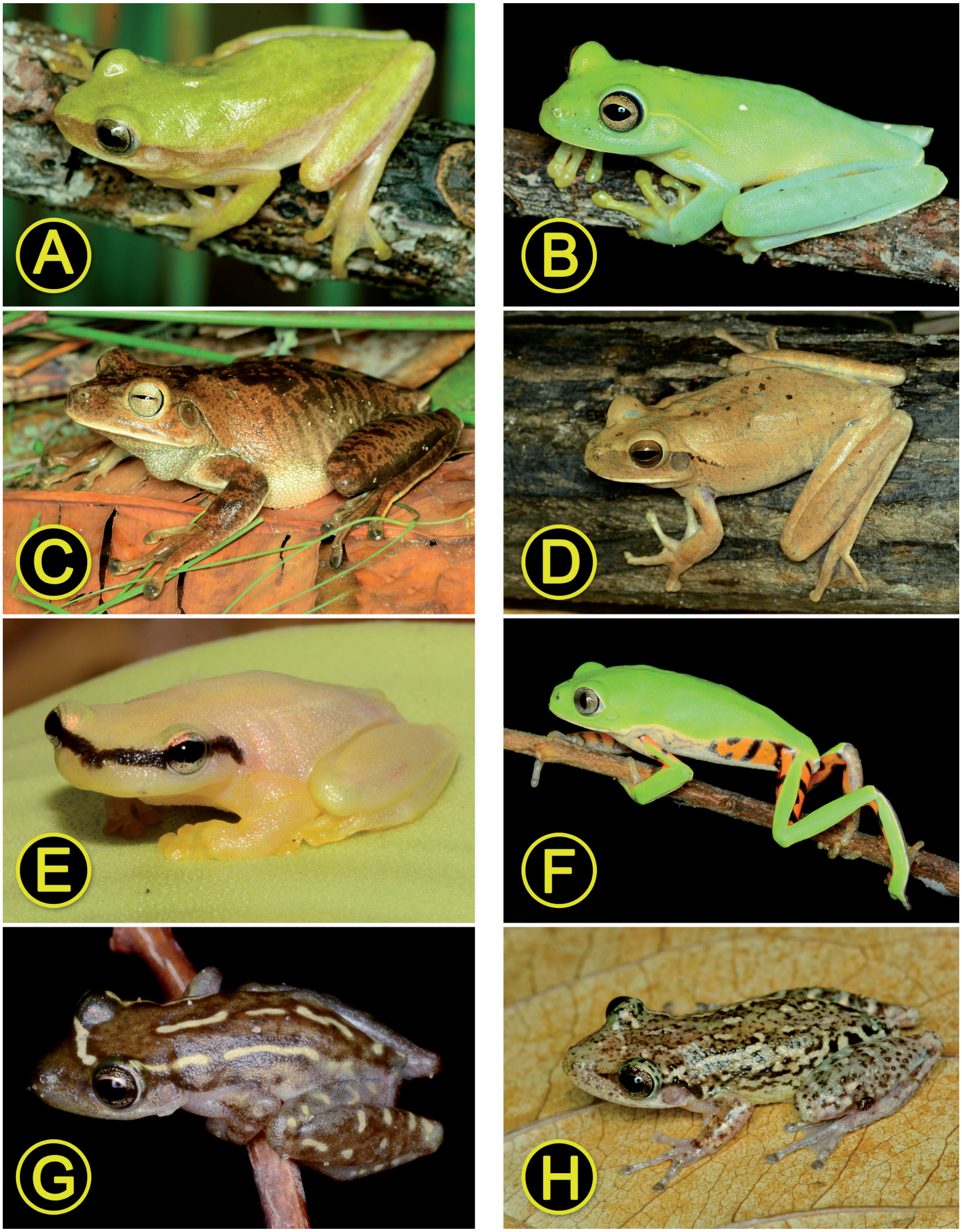

Figure 4. Anurans recorded at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. See caption of Figure 3 for additional information. (A) *Dendropsophus rubicundulus* (UFBA 13953; SVL 20.6); (B) *Hypsiboas albomarginatus* (UFBA 13971; SVL 43.5); (C) *H. faber* (UFBA 13909; SVL 86.7); (D) *H. raniceps* (UFBA 13898; SVL 68.6); (E) *Phyllodytes melanomystax* (UFBA 13960; SVL 23.4); (F) *Phyllomedusa nordestina* (UFBA 14070; SVL 35.2); (G) *Scinax auratus* (UFBA 13962; SVL 23.0); (H) *Scinax cretatus* (UFBA 14036; SVL 30.4).

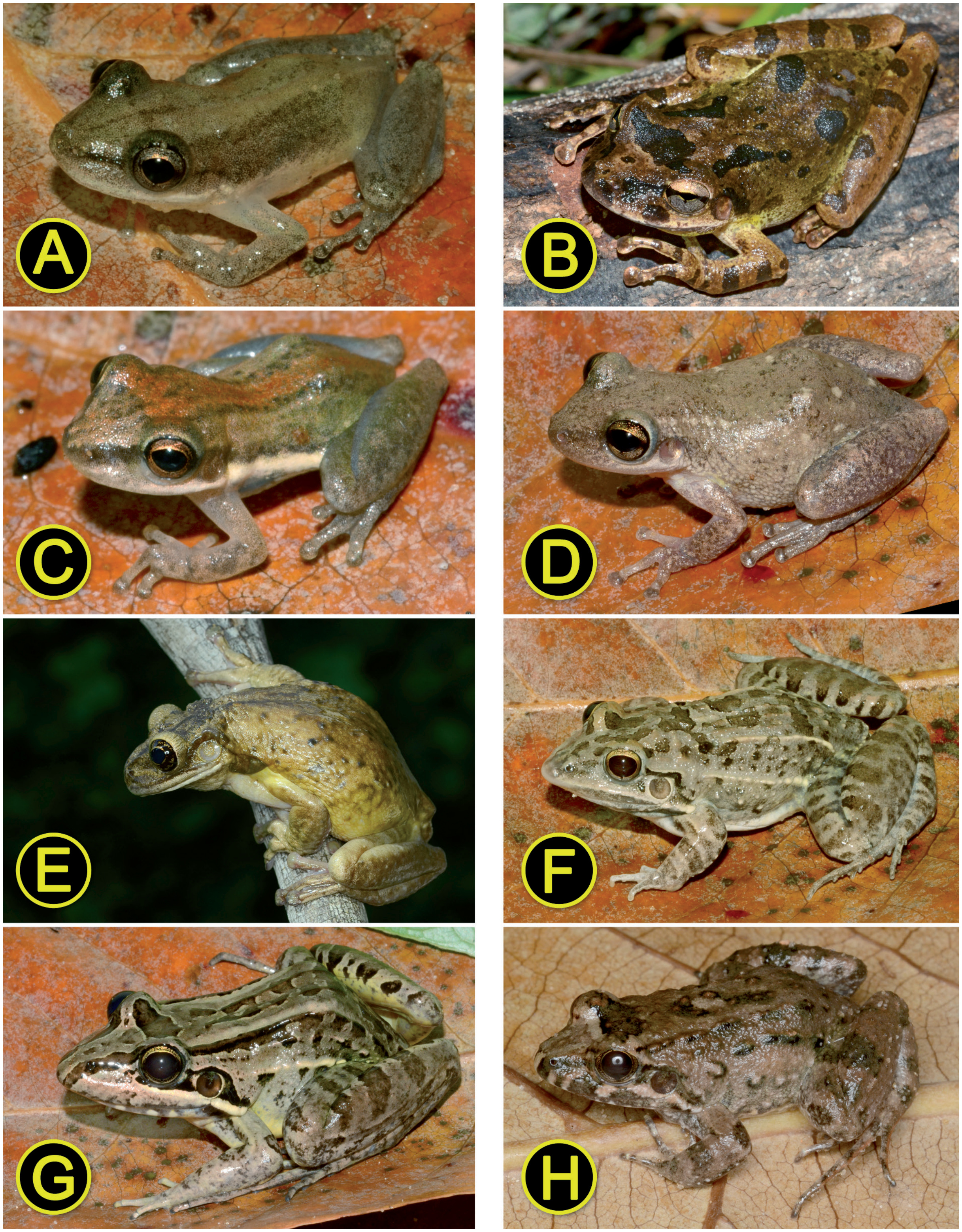

Figure 5. Anurans recorded at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. See caption of Figure 3 for additional information. (A) *Scinax fuscomarginatus* (UFBA 14010; SVL 18.6); (B) *S. eurydice* (UFBA 13893; SVL 42.3); (C) *S. melanodactylus* (UFBA 13939; SVL 12.4); (D) *S. x-signatus* (UFBA 13949; SVL 34.1); (E) *Trachycephalus atlas* (UFBA 14087; SVL 74.4); (F) *Leptodactylus fuscus* (UFBA 13936; SVL 41.8); (G) *L. macrosternum* (UFBA 13934; SVL 64.3); (H) *L. natalensis* (UFBA 13963; SVL 27.3).

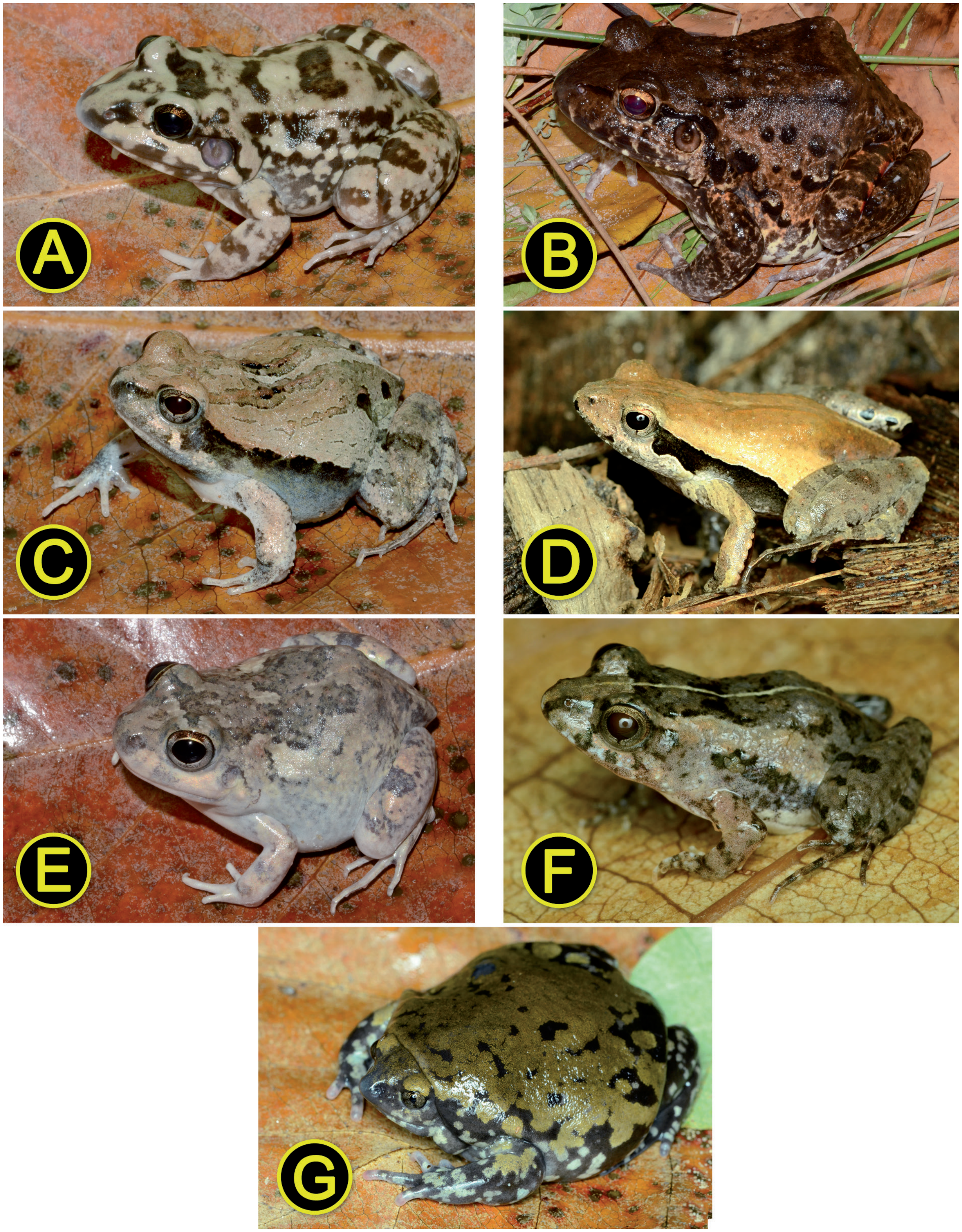

Figure 6. Anurans recorded at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. See caption of Figure 3 for additional information. (A) *Leptodactylus troglodytes* (UFBA 13902; 46.6); (B) *L. vastus* (UFBA 13941; SVL 147.8); (C) *Physalaemus albifrons* (UFBA 13926; SVL 32.0); (D) *P. kroyeri* (UFBA 14008; SVL 19.4); (E) *Pleurodema diplolister* (UFBA 13911; SVL 33.3); (F) *Pseudopaludicola mystacalis* (UFBA 13955; SVL 11.5); (G) *Dermatonotus muelleri* (UFBA 13900; SVL 55.2).

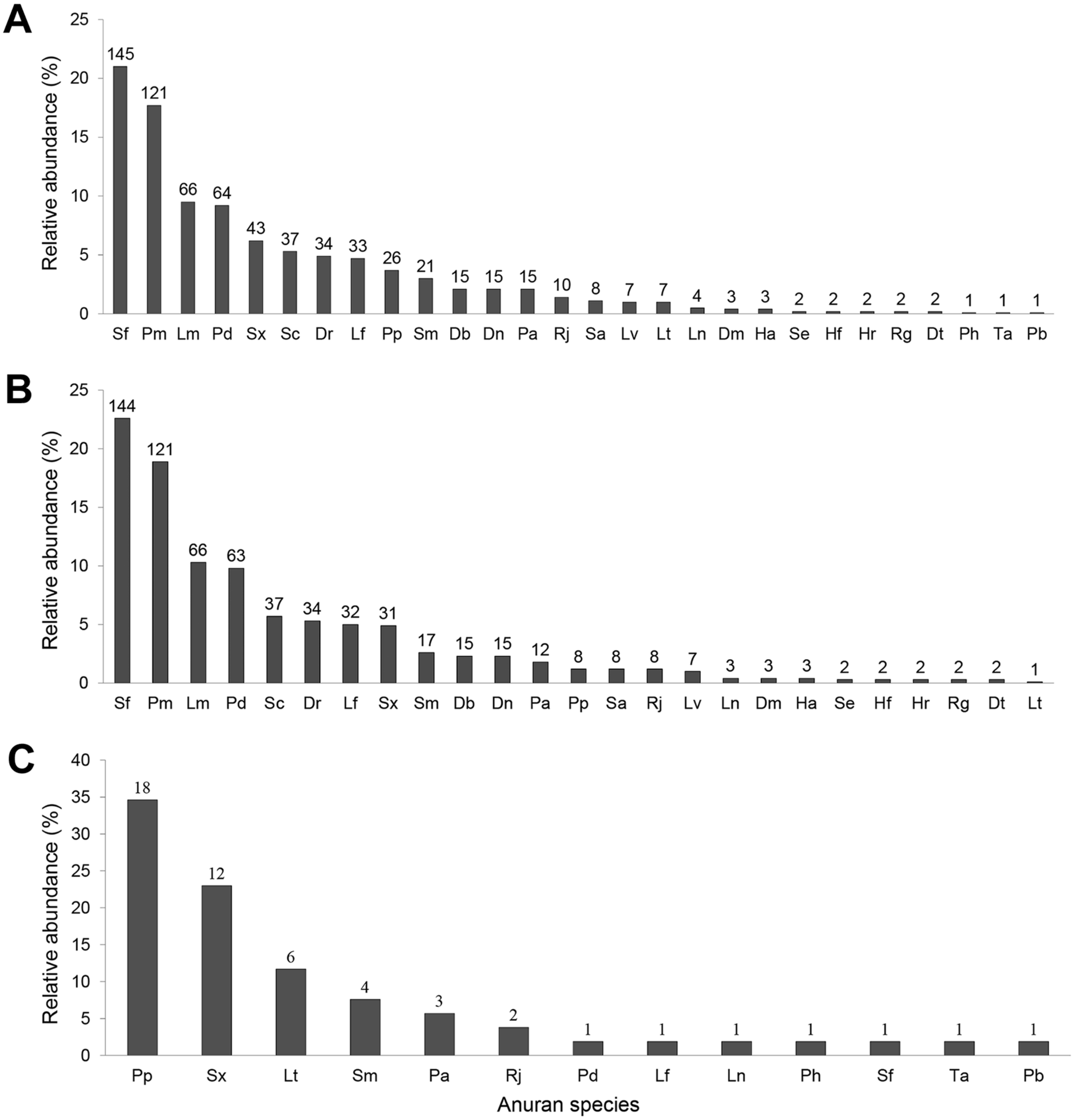

Figure 7. Relative abundance of anuran species (%) observed at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. (A) Overall Restinga; (B) wetland zones; (C) shrubby vegetation zones. The total number of recorded individuals per species is indicated above each column. Anuran species: Db, *Dendropsophus branneri*; Dn, *D. nanus*; Dm, *D. minutus*; Dr, *D. rubicundulus*; Dt, *Dermatonotus muelleri*; Ha, *Hypsiboas albomarginatus*; Hf, *H. faber*; Hr, *H. raniceps*; Lf, *Leptodactylus fuscus*; Lm, *L. macrosternum*; Ln, *L. natalensis*; Lt, *L. troglodytes*; Lv, *L. vastus*; Ph, *Phyllodytes melanomystax*; Pb, *Phyllomedusa bahiana*; Pa, *Physalaemus albifrons*; Pd, *Pleurodema diplolister*; Pp, *Pristimantis paulodutrai*; Pm, *Pseudopaludicola mystacalis*; Rg, *Rhinella granulosa*; Rj, *R. jimi*; Sa, *Scinax auratus*; Sc, *S. cretatus*; Se, *S. eurydice*; Sf, *S. fuscomarginatus*; Sm, *S. melanodactylus*; Sx, *S. x-signatus*; Ta, *Trachycephalus atlas*.

were not vouchered. The specimen identified here as *D. oliveirai* was putatively attributed to this species because it also resembles *D. decipiens* and *D. minusculus*. These species are highly polymorphic to external morphology and color patterns (Zina et al. 2014) and their specific limits remain unclear.

The species accumulation curves tended to asymptotes for FWZ and the overall study area, but not for SVZ (Figure 8), which suggests that not all anuran species that potentially inhabit the latter physiognomy were recorded. The species richness estimator Jackknife 1 estimated 33.9 ± 2.3 anuran species for the overall study area [28 species observed in SUs; 33 species combining SUs and EPs], 28.9 ± 1.9 species to FWZ (25 species observed in SUs; 30 species combining SUs and EPs) and 20.8 ± 2.9 species for SVZ (13 species observed both in SUs and SUs plus EPs). See Table 3 for additional diversity data.

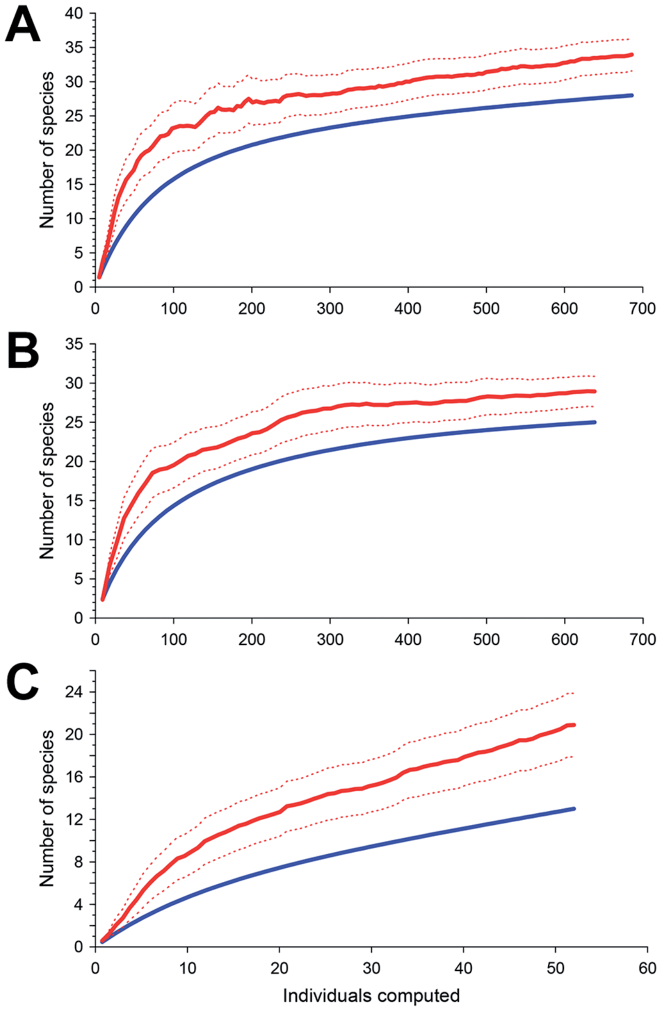

Figure 8. Accumulation curves for anurans sampled at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil, constructed from 1000 randomizations on the order of samplings. (A) Overall Restinga; (B) freshwater wetland zones only; and (C) shrubby vegetation zones only. Blue line represents accumulation curves, red line represents species estimates based on Jackknife 1 estimator and red dashed lines represents its interval of confidence.

The FWZ showed the highest species richness among the sampled environments (25 species, 89.3%) and SVZ the lowest (13 species; 46.4%) (Table 3). The family Hylidae had the highest species richness in both physiognomies, followed by Leptodactylidae: FWZ (14 species and eight species, respectively); SVZ (six species and five species, respectively). Regarding species abundances, the highest value was observed for the FWZ (638 individuals, 92.4%) and the lowest for SVZ (52 individuals, 7.5%). Ten species were common to both physiognomies: *Leptodactylus fuscus*, *L. natalensis*, *L. troglodytes*, *Physalaemus albifrons*, *Pleurodema diplolister*, *Pristimantis paulodutrai*, *Rhinella jimi*, *Scinax fuscomarginatus*, *S. melanodactylus* and *S. x-signatus*. The highest species diversity was obtained for FWZ (H' = 2.4) and the lowest for SVZ (H' = 1.9), although the evenness was equal for both physiognomies (J' = 0.76).

The species dominance pattern (evenness) was similar for both physiognomies and for the overall study area (Table 3; Figure 7). The same dominant species were observed for the FWZ and overall study area, but not for the SVZ that had a distinct dominant species, indicating that the former contributed more toward the dominance of the overall study area than the latter. In the FWZ, the dominant species was *Scinax fuscomarginatus* (144 individuals, 22.6%) and in the SVZ, *Pristimantis paulodutrai* (18 individuals, 34.6%). Using the Gaston's criterion for species commonness and rarity, in the overall study area eight species were considered common, 12 species intermediate and eight species rare; in the freshwater wetland zones, eight species were common, eight species intermediate and nine species rare; and in the shrubby vegetation zones, four were common, two were intermediate and seven were rare. There were exclusive species to each physiognomy: FWZ - *Dendropsophus branneri*, *D. minutus*, *D. nanus*, *D. rubicundulus*, *Dermatonotus muelleri*, *Hypsiboas albomarginatus*, *H. faber*, *H. raniceps*, *Leptodactylus macrosternum*, *L. vastus*, *Pseudopaludicola mystacalis*, *Rhinella granulosa*, *Scinax auratus*, *S. cretatus* and *S. eurydice*; SVZ - *Phyllodytes melanomystax*, *Phyllomedusa bahiana* and *Trachycephalus atlas*.

Discussion

The Restinga of the Conde municipality in the state of Bahia, Brazil, stands out as the second locality with the highest anuran species richness recorded for a Restinga environment (33 species), whereas the highest (34 species) was obtained for the Restinga of the Mata de São João municipality (Bastazini et al. 2007; Telles et al. 2012, Oliveira & Rocha 2015, Xavier et al. 2015), both of them located in the northern coast (NC) of the state of Bahia. Currently, 62 nominal species of amphibians are known for the NC (M.F. Napoli, L.M. Silva and R.O. Abreu, unpublished data), 60 species of Anura [including an exotic species, *Lithobates catesbeianus* (Shaw, 1802)] and two species of Gymnophiona [*Chthonerpeton noctinectes* da Silva, Britto-Pereira & Caramaschi, 2003 and *Siphonops annulatus* (Mikan, 1820)]. Of the 60 anuran species listed by M.F. Napoli, L.M. Silva and R.O. Abreu

Table 3. Anuran diversity of the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. Physiognomies: FWZ, freshwater wetland zones; SVZ, shrubby vegetation zones.

	Overall Restinga	**SVZ**	**FWZ**
Number of individuals	690	52	638
Species richness (observed)	28	13	25
Species richness (Jackknife 1)	33.9 ± 2.3	20.8 ± 2.9	28.9 ±1.9
Dominant species	*Scinax fuscomarginatus*	*Pristimantis paulodutrai*	*Scinax fuscomarginatus*
Dominance observed	21%	34.6%	22.6%
Shannon-Wiener index (H')	2.57	1.9	2.4
Pielou's index J' (evenness)	0.77	0.76	0.76

(*op. cit.*), 33 species (not exotic) were recorded for the 'open Restinga' of the Conde municipality, totaling 55% of the nominal species recorded for the NC. Previous studies conducted in the NC at the Mata de São João municipality (Juncá 2006, Bastazini et al. 2007) obtained similar anuran species richness (25 and 34 species, respectively). Nonetheless, Juncá (2006) sampled only forested environments at the Reserva Sapiranga and Bastazini et al. (2007) sampled the same habitats used by Juncá (2006) plus the riparian forest of the Timeantube River and shrub vegetation zones at Praia do Forte. As our study considered only shrubby vegetation zones and freshwater wetland zones of 'open' areas, the previous results do not permit direct comparisons with ours, with the exception of the study by Bastazini et al. (2007) for Praia do Forte.

In the Restinga of the Conde municipality we recorded 20 anuran species not registered for the Restinga of Praia do Forte (but six of them were registered at the Reserva Sapiranga), 12 species were common to both Restingas and two species were unique to Praia do Forte [but restricted to the riparian forest, *Chiasmocleis* sp. and *Itapotihyla langsdorffii* (Duméril & Bibron, 1841)]. Although this result represents 57.6% more anuran species for the Restinga of the Conde municipality, it should be analyzed with caution, because (i) the total extension area analyzed by Bastazini et al. (2007) at Praia do Forte was smaller than that considered in the present study, (ii) these authors did not emphasize the water bodies of the region and (iii) the exclusive species recorded at the Restinga of the Conde municipality are common species in the wetlands of the NC (M.F. Napoli, L.M. Silva and R.O. Abreu, unpublished data), which are likely to disperse throughout the contiguous coastal plains separating these two areas of study.

At the study area, the highest anuran species richness and diversity were observed in wetlands. The presence of water bodies is preponderant in determining species richness (Babbitt & Tanner 2000) and marshes with reeds also provide vertical substrates that increase the amount of microenvironments available to anurans (Ehrlich 1993, Stiling 1996). The lowest species richness was observed in the shrubby vegetation zones, which is an understandable consequence, considering that these environments have low water availability, a limiting factor for the occurrence of amphibians that depend on it for their osmotic balance and/or as breeding sites (Duellman & Trueb 1994). On the other hand, the accumulation curve obtained for the shrubby vegetation zones with the Jackknife 1 estimator did not reach an asymptote and estimated a higher species richness than that actually observed (estimated 20.8 ± 2.9 species, observed 13 species). This result is possibly due to the uniques and singletons found in the shrubby vegetation zones that did not decrease with increasing sampling effort, as discussed by Toti et al. (2000). Furthermore, it is noteworthy that three of these singletons were common species in wetlands (*S. fuscomarginatus*, *Pleurodema diplolister* and *Leptodactylus fuscus*) and therefore may be eventually using the shrubby vegetation zones.

Although the sampled shrubby vegetation zones lacked water bodies, more complex plant stratification environments, such as bushes, create different microenvironments, decrease the incidence of direct sunlight and favor the formation of wetter soils by organic matter accumulation, providing humidity and temperatures appropriate to maintain greater diversity of anurans than those areas devoid of vegetation stratification (Xavier & Napoli 2011). Furthermore, some SUs in shrubby vegetation zones had terrestrial tank bromeliads (Figure 2A), plants that retain moisture between their leaves and accumulate water inside the leaf-tank (Freitas et al. 2000, Cogliatti-Carvalho 2001). In shrubby vegetation zones, terrestrial tank bromeliads are the only source of water supply to maintain the minimum physiological needs of anurans (Dabés et al. 2012) and are considered fundamental ecological units in the Restinga ecosystem since they are used by many wildlife groups for shelter and/or as breeding sites (Peixoto 1995, Richardson 1999, Carvalho-e-Silva et al. 2000, Schneider & Teixeira 2001, Teixeira et al. 2002). The phytotelm-dweller hylids *Phyllodytes melanomystax* and *Trachycephalus atlas* were exclusively observed in these bromeliads, although as singletons. *Trachycephalus atlas* uses temporary puddles to oviposition and for the development of its tadpoles (Barreto et al. 2015), and therefore the presence of an adult specimen in a bromeliad of the shrubby vegetation zone is possibly associated with its use as shelter. *Phyllodytes melanomystax*, on the other hand, is a bromeligenous treefrog (*sensu* Peixoto 1995), i.e. it uses bromeliads throughout its life cycle for breeding and feeding, which explains its restriction to tank bromeliads of this physiognomy. Bastazini et al. (2007) indicated that terrestrial tank bromeliads were among the environmental variables that explained most of the changes in the anuran composition from the Restinga of the Mata de São João municipality. Moreover, *P. melanomystax* was the most frequent species in terrestrial tank bromeliads at Praia do Forte.

The family Hylidae showed the highest species richness in the Restinga of the Conde municipality, which coincides with the pattern already reported for the Restinga of the Mata de São João municipality (Juncá 2006, Bastazini et al. 2007) and for other Restingas in Brazil (e.g., Rocha et al. 2008, Narvaes et al. 2009, Zina et al. 2012, Oliveira & Rocha 2015). In the wetland zones, the dominant species was *Scinax fuscomarginatus* (Table 3). This small-sized hylid is commonly associated with reeds and other emergent plants in temporary ponds and lagoons (Brasileiro et al. 2005, Van-Silva et al. 2007, Serafim et al., 2008), similar to those observed in the wetlands of the study area (Figure 2B, D), except for the SU P4. This SU differed from the others by not showing plant stratification, being essentially characterized as a temporary pond in sandy soil the surroundings of which were covered by grasses (Figure 2C). The dominant anuran species at P4 was the leptodactylid *Pseudopaludicola mystacalis*, a small-sized terrestrial and semiaquatic species that is abundant in wetlands of opens formations (Silva et al. 2008, Ribeiro-Junior & Bertoluci 2009, Valdujo et al. 2011). In shrubby vegetation zones the dominant species was a Terrarana, the craugastorid *Pristimantis paulodutrai*, the most abundant species in forested areas of the NC (Bastazini et al. 2007). *Pristimantis paulodutrai* is a small-sized direct-developing frog with terrestrial breeding (Napoli et al. 2009), like other Terrarana (Hedges et al. 2008). This mode of reproduction allows this species to breed in environments deprived of water bodies (Xavier & Napoli 2011). Furthermore, *P. paulodutrai* has a low dehydration rate and high rate of rehydration from a neutral substrate (Dabés et al. 2012), which helps to explain its ability to occupy different types of environments, including those under high osmotic stress, as is the case of shrubby vegetation zones.

The species composition observed for the Restinga of the Conde municipality deserves additional comments. The Restingas of northeastern Brazil were singled out by Xavier et al. (2015) as a unique biotic element with respect to ecological and biogeographic distribution of anuran species, including taxa from three distinct morphoclimatic domains (Tropical Atlantic, Cerrado and Caatinga).

These authors considered eight anuran species from that biotic element as exclusive to the Atlantic Tropical domain, six (75%) of which were recorded in this study: *Leptodactylus vastus*, *Phyllodytes melanomystax*, *Pristimantis paulodutrai*, *Scinax auratus*, *S. cretatus* and *S. eurydice*. Seven species were common to the Cerrado and Caatinga domains, all recorded in the Restinga of the Conde municipality: *Dendropsophus nanus*, *D. rubicundulus*, *Dermatonotus muelleri*, *Hypsiboas raniceps*, *Leptodactylus troglodytes*, *Pleurodema diplolister* and *Rhinella jimi*. These authors also highlighted the existence of small and sparse relictual patches of Cerrado and Caatinga in the NC, some of them contiguous with Restinga environments, which could explain the presence of anuran species from three different morphoclimatic domains (see Xavier et al. 2015 for the discussion on the possible relationship between these relictual patches of Cerrado and Caatinga habitats and climatic fluctuations of the last period of the Quaternary).

The Restinga of the Conde municipality contains 87% of the anuran species found in the northeastern biotic element studied by Xavier et al. (2015), a natural biogeographical unit. Natural biogeographical units are basic units of comparison in many ecological and evolutionary large-scale studies and are areas that should be evaluated when planning the conservation of biota, once they allow the understanding of both the evolutionary history of taxa and areas (Lomolino et al. 2006, Xavier et al. 2015). Moreover, the Restinga of the Conde municipality stands out as the one with the highest anuran species richness already recorded considering only shrubby vegetation zones and freshwater wetland zones. In this context, the Restinga of the Conde municipality seems to be an important region for the conservation of the biota of the northern coast of the state of Bahia.

Appendix 1

Vouchers of anurans collected at the Restinga of the Conde municipality, northern coast of the state of Bahia, northeastern Brazil. The data were entered in the following order: family, species, acronym of the scientific collection (UFBA, Museu de Zoologia da Universidade Federal da Bahia) followed by their institutional record number (Arabic numerals).

BUFONIDAE. *Rhinella granulosa*, UFBA 13907-13908. *Rhinella jimi*, UFBA 13961.

CRAUGASTORIDAE. *Pristimantis paulodutrai*, UFBA 13882-13891, 14067-14069.

HYLIDAE. *Dendropsophus branneri*, UFBA 13970, 14056-14058. *Dendropsophus oliveirai*, UFBA 14048. *Dendropsophus elegans*, UFBA 14044. *Dendropsophus minutus*, UFBA 13964-13965, 14049. *Dendropsophus nanus*, UFBA 13972-13984, 14064. *Dendropsophus rubicundulus*, UFBA 13953-13954, 14051-14055, 14065-14066. *Hypsiboas albomarginatus*, UFBA 13971, 14047. *Hypsiboas faber*, UFBA 13909-13910. *Hypsiboas raniceps*, UFBA 13898-13899. *Phyllodytes melanomystax*, UFBA 13960. *Phyllomedusa nordestina*, UFBA 13985-13999, 14001-14003, 14070. *Scinax auratus*, UFBA 13962, 13966-13969. *Scinax cretatus*, UFBA 14037-14043. *Scinax eurydice*, UFBA 13893, 14050. *Scinax fuscomarginatus*, UFBA 14010-14025. *Scinax melanodactylus*, UFBA 13939-13940, 14034. *Scinax x-signatus*, UFBA 13892, 13942-13952. *Trachycephalus atlas*, UFBA 14087.

LEPTODACTYLIDAE. *Leptodactylus fuscus*, UFBA 13936-13938, 13897, 14061-14063. *Leptodactylus macrosternum*, UFBA 13934-13935, 13894-13896, 14060. *Leptodactylus natalensis*, UFBA 13963. *Leptodactylus troglodytes*, UFBA 13902-13906, 14035. *Leptodactylus vastus*, UFBA 13941, 14045-14046. *Physalaemus albifrons*, UFBA 13926-13933. *Physalaemus kroyeri*, UFBA 14008-14009. *Pleurodema diplolister*, UFBA 13911-13925. *Pseudopaludicola mystacalis*, UFBA 13955-13958, 14059.

MICROHYLIDAE. *Dermatonotus muelleri*, UFBA 13900-13901.

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