

Non-volant tetrapods from Reserva Biológica de Duas Bocas, State of Espírito Santo, Southeastern Brazil

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Abstract: Reserva Biológica de Duas Bocas (2,910 ha) is one of the largest Atlantic forest remnants in the State of Espírito Santo, Southeastern Brazil. We recorded non-volant tetrapods in this area from May 2007 through April 2008, using pitfalls, live traps, camera traps, and diurnal and nocturnal opportunistic searches. In addition, we compiled available museum and literature records from this area. We documented 52 species of amphibians, 24 species of non-avian reptiles, and 39 species of non-volant mammals. Out of these 115 species, 47 are new records for this area and six other species had their geographic ranges expanded with the present study. Furthermore, we present the record of predation of the tree frog *Hypsiboas faber* by the snake *Chironius bicarinatus*. Out of the species listed, five species are listed as threatened with extinction in the State of Espírito Santo, and many others have uncertain conservation status. Reserva Biológica de Duas Bocas is an important wildlife refuge, especially considering the expansion of urban areas in its surroundings.

Keywords: amphibians, mammals, non-avian reptiles, Atlantic forest, inventory, biodiversity.

TONINI, J.F.R., CARÃO, L.M., PINTO, I.S., GASPARINI, J.L., LEITE, Y.L.R. & COSTA, L.P. **Tetrápodes não voadores da Reserva Biológica de Duas Bocas, Estado do Espírito Santo, Sudeste do Brasil.** *Biota Neotrop.* 10(3): <http://www.biotaneotropica.org.br/v10n3/pt/abstract?inventory+bn02710032010>.

Resumo: A Reserva Biológica de Duas Bocas (2.190 ha) é um dos maiores remanescentes de Mata Atlântica do Estado do Espírito Santo, Sudeste do Brasil. Nós amostramos tetrápodes não voadores nessa área entre maio de 2007 e abril de 2008, utilizando armadilhas de queda, armadilhas de isca, armadilhas fotográficas e buscas oportunísticas diurnas e noturnas. Além disso, nós compilamos registros de vertebrados não voadores ocorrentes nesta área disponíveis na literatura e através de espécimes em museus. Nós documentamos 52 espécies de anfíbios, 24 espécies de répteis não voadores e 39 espécies de mamíferos não voadores. Do total de 115 espécies, 47 configuram novos registros para a área e seis outras espécies tiveram sua distribuição geográfica ampliada com os resultados do presente estudo. Além disso, apresentamos o registro de predação da perereca *Hypsiboas faber* pela serpente *Chironius bicarinatus*. Cinco das espécies registradas são listadas como ameaçadas no Estado do Espírito Santo e muitas outras possuem estado de conservação incerto. A Reserva Biológica de Duas Bocas é um importante refúgio de vida selvagem, principalmente quando consideramos a expansão de áreas urbanas no seu entorno.

Palavras-chave: anfíbios, mamíferos, répteis não-avianos, Mata Atlântica, inventário, biodiversidade.

Introduction

Despite intense exploitation of its natural resources, the Brazilian Atlantic forest still maintains an extremely rich biodiversity, including a large number of endemic species of plants and animals (Morellato & Haddad 2000). It has unique floristic and climatic characteristics, and a marked altitudinal gradient, favoring geographic isolation among populations (Haddad et al. 1996).

Since European colonization, 500 years ago, the Atlantic forest cover in the State of Espírito Santo has been reduced to only 8.9% (Lederman & Padovan 2005), consisting typically of small, isolated fragments disturbed by human activities. One of the main Atlantic forest remnants is located in the metropolitan region of the state capital, Vitória (Grande Vitória). Reserva Biológica de Duas Bocas (RBDB) is an important buffer and refuge for the native fauna, and remained well preserved despite intense pressure from the expansion of urban areas and associated activities in its surroundings.

The destruction of the Atlantic forest in the State of Espírito Santo began in 1503, with the establishment of the first village, and later intensified with the indiscriminate extraction of hardwood (Secretaria... 1998). Coffee plantations expanded in the second half of the 19th century, leading to deforestation, soil erosion, and river aggradation and pollution (Schettino 2000). The state population was predominately rural until the 1960's, but industrialization led to an increase in urban population, especially around the state capital (Lederman & Padovan 2005). Nowadays, the main land use is for cattle ranches, which cover 40% of the state, especially in the northern region (Lederman & Padovan 2005).

According to Passamani & Mendes (2007), 197 animal species are threatened with extinction and 11 have been considered regionally extinct in the State of Espírito Santo. The number of threatened species in the state corresponds to 32.9% of the total number of species included on the Brazilian Endangered Species List (Machado et al. 2005). This is a worrisome problem, since the state occupies only 0.53% of the Brazilian territory. This situation if further aggravated by the lack of long-term inventories, leading to very few adequate species lists. The only two animal groups with reasonably well-documented faunas are birds (Simon 2009) and mammals (Moreira et al. 2008).

Here we provide a list of non-volant tetrapods from Reserva Biológica de Duas Bocas, based on a one-year survey using a combination of several sampling techniques. Our main goal is to provide a good preliminary inventory of the local biodiversity, and to increase scientific collections, which will subsidize further work on taxonomy, biogeography and conservation biology.

Materials and Methods

1. Study area

RBDB is located between latitudes 20° 14' 04" and 20° 18' 30" S, and longitudes 40° 28' 01" and 40° 32' 07" W (Figure 1). "Duas Bocas" means "two mouths" in Portuguese, and this name stems from the confluence of the rivers Panelas and Naiá-Assú, which flow into a dam built in 1951 on the eastern end of the reserve. RBDB has an area of 2,910 ha and the elevation ranges from 300 to 738 m. Most of the area shelters primary forest. The climate is tropical humid (average annual temperature between 19 and 22 °C, relative humidity > 70%), and rains are well distributed throughout the year, with an average annual rainfall of approximately 1,500 mm (Feitoza 1986).

2. Data sampling

We collected non-volant tetrapods at Alto Alegre, in the Southeastern area of RBDB (20° 16' 52" S and 40° 31' 19" W, elevation 550-738 m), during five days each month from May 2007 to April 2008, totaling 60 sampling days.

We used pitfall traps for capturing small mammals, amphibians and non-avian reptiles. We established six 100 m transects where eleven 60-liter buckets (40 cm in diameter by 54 cm in depth) were installed, 10 m apart. The buckets were connected by 50 cm-high drift fences secured by wooden stakes. In addition, we used conventional live traps for small mammals: one Sherman (23 × 9 × 9 cm) and one wire cage (32 × 15 × 15 cm) were arranged within a radius of about five meters of each pitfall. These traps were baited with pineapple and peanut butter.

We also placed three camera traps (Tigrinus® 4.0C) for detecting medium to large mammals. They were installed 50 cm above the ground, attached to tree trunks near creeks or possible animal routes. Cameras were set to operate both night and day with a minimum interval of 90 seconds between pictures. They were checked monthly, when film and batteries were changed and the total sampling effort was 1,095 camera trap-nights. Additionally, we conducted opportunistic diurnal and nocturnal searches for animals, nests, shelter, tracks, or other signs of non-volant tetrapods in the study area. These searches were conducted throughout the year, 5 hours a day on average.

Series of specimens were collected to serve as museum vouchers (listed in Appendix 1), and were prepared according to standard techniques (Auricchio & Salomão 2002). Vouchers are deposited in the mammal collection at Universidade Federal do Espírito Santo, Vitória (UFES); Museu de Biologia Professor Mello Leitão, Santa Teresa (MBML); and Célio Fernando Baptista Haddad collection (CFBH) at Universidade Estadual Paulista Júlio de Mesquita Filho, Rio Claro, but some specimens still will be deposited in MBML. The remaining animals were identified and released (in the case of amphibians and non-avian reptiles), or marked with numbered ear tags (National Band and Tag Co.) and released (in the case of mammals). We also used published records (Paresque et al. 2004, Prado & Pombal 2005) and museum data available at the Reference Center on Environmental Information – CRIA (<http://splink.cria.org.br>) for MBML, CFBH, and "Alphonse Richard Hoge" herpetological collection (IBSP-Herpeto).

The taxonomic arrangement used for mammals follows Wilson & Reeder (2005), but includes recent updates (Bonvicino et al. 2008). For amphibians, we followed Frost (2010) and for non-avian reptiles, taxonomic arrangement follows Frost et al. (2001), Castoe et al. (2004), Gamble et al. (2008), Vidal & Hedges (2009) and Zaher et al. (2009), with some modifications, since the phylogeny of higher reptile taxa is still uncertain.

Results

Our trapping effort was 3,960 pitfall trap-nights, 7,920 live trap-nights resulting in 427 captures of mammals (3.59% success), 433 of amphibians (10.9% success), and 18 of non-avian reptiles (0.45% success). The sampling effort in the opportunistic searches was 300 hours and we captured 25 non-avian reptiles and 151 amphibians.

We registered 114 species of non-volant tetrapods at RBDB: 39 mammals (Table 1; Figure 4i-p), 52 amphibians (Table 2; Figures 2 and 3a-n) and 24 non-avian reptiles (Table 3; Figures 3o and 4a-h).

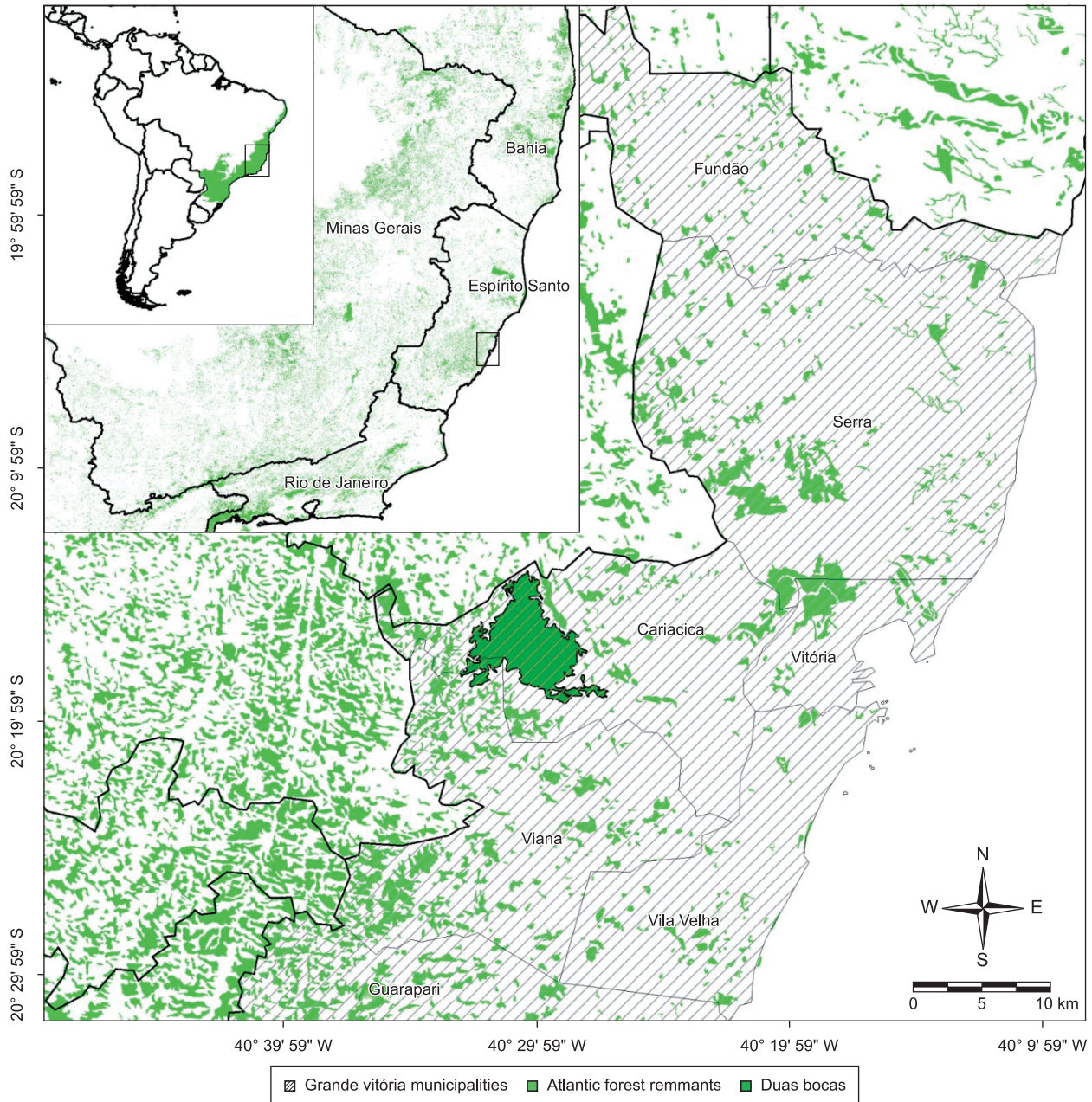


Figure 1. Location of Reserva Biológica de Duas Bocas, State of Espírito Santo, Southeastern Brazil.

Discussion

With the results presented above, we increase the species list of non-volant tetrapods from RBDB by 40%. These records correspond to 30% of the mammal species in the State of Espírito Santo (Moreira et al. 2008). The same fraction cannot be calculated for non-avian reptiles and amphibians because there is no state wide consistent species list of these animals.

Sixteen native mammal species had not been recorded in the reserve before: the marsupials *Monodelphis iheringi* (Figure 4i) and *Chironectes minimus*; the xenarthrans *Dasypus novemcinctus*, *Dasypus septemcinctus*, and *Bradypus torquatus*; the primates *Alouatta guariba*, *Callithrix geoffroyi*, and *Cebus nigrinus*; the carnivores *Eira barbara*, *Nasua nasua* (Figure 4p), *Procyon*

cancrivorus, and *Cerdocyon thous*, and the rodents *Blarinomys breviceps*, *Juliomys pictipes* (Figure 4m), *Thaptomys nigrita*, and *Cuniculus paca*.

In addition, we confirmed the presence of two exotic species: domestic dogs (*Canis familiaris*) were often seen and photographed by camera traps, and one specimen of black rat (*Rattus rattus*) was trapped in the middle of a very pristine forest, at least 3 km away from the forest edge. Domestic or free-roaming dogs can be frequent visitors to wet tropical forest preserves of the Neotropics, and are potential competitors with other large mammals, including some large cats and smaller carnivores, like *Cerdocyon thous* (Srbek-Araújo & Chiarello 2008). Several researchers have documented the presence of domestic dogs in Brazilian protected areas (e.g., Horowitz 1992,

Table 1. Non-volant mammal species from Reserva Biológica de Duas Bocas, State of Espírito Santo, southeastern Brazil. MBML = Museu de Biologia Prof. Mello Leitão.

Taxon	Type of record	References
Didelphimorphia		
Didelphidae		
<i>Chironectes minimus</i> (Zimmermann, 1780)	Visual	This study (J.L. Gasparini)
<i>Didelphis aurita</i> (Wied-Neuwied, 1826)	Voucher	This study/MBML
<i>Gracilinanus microtarsus</i> (Wagner, 1842)	Voucher	This study/MBML
<i>Marmosa murina</i> (Linnaeus, 1758)	Voucher	MBML/Paresque et al. (2004)
<i>Marmosops incanus</i> (Lund, 1840)	Voucher	This study/MBML
<i>Metachirus nudicaudatus</i> (É. Geoffroy, 1803)	Voucher	This study/MBML
<i>Micoureus paraguayanus</i> (Tate, 1931)	Voucher	This study/Paresque et al. (2004)
<i>Monodelphis americana</i> (Müller, 1776)	Voucher	This study/ Paresque et al. (2004)
<i>Monodelphis iheringi</i> (Thomas, 1888)	Voucher	This study
<i>Philander frenatus</i> (Olfers, 1818)	Voucher	This study/MBML
Cingulata		
Dasypodidae		
<i>Dasybus novemcinctus</i> Linnaeus, 1758	Voucher	This study
<i>Dasybus septemcinctus</i> Linnaeus, 1758	Voucher	This study
Pilosa		
Bradypodidae		
<i>Bradypus torquatus</i> Illiger, 1811	Visual	This study
Primates		
Cebidae		
<i>Callithrix geoffroyi</i> (É. Geoffroy in Humboldt, 1812)	Visual	This study
<i>Cebus nigratus</i> (Goldfuss, 1809)	Visual	This study
Atelidae		
<i>Alouatta guariba</i> (Humboldt, 1812)	Vocalization	This study
Carnivora		
Canidae		
<i>Canis familiaris</i> Linnaeus, 1758	Visual/Camera trap	This study
<i>Cerdocyon thous</i> (Linnaeus, 1766)	Visual	This study
Mustelidae		
<i>Eira barbara</i> (Linnaeus, 1758)	Visual/Camera trap	This study
<i>Lontra longicaudis</i> (Olfers, 1818)	-	Amado (2004)
Procyonidae		
<i>Nasua nasua</i> (Linnaeus, 1766)	Visual/Camera trap	This study
<i>Procyon cancrivorus</i> (G. Cuvier, 1798)	Camera trap	This study
Rodentia		
Sciuridae		
<i>Guerlinguetus ingrani</i> (Thomas, 1901)	Visual/Voucher	This study/MBML
Cricetidae		
<i>Akodon cursor</i> (Winge, 1887)	Voucher	This study/Paresque et al. (2004)
<i>Blarinomys breviceps</i> (Winge, 1887)	Voucher	This study
<i>Euryoryzomys russatus</i> (Wagner, 1848)	Voucher	MBML/ Paresque et al. (2004)
<i>Juliomys pictipes</i> (Osgood, 1933)	Voucher	This study
<i>Nectomys squamipes</i> (Brants, 1827)	Voucher	This study/ MBML
<i>Oecomys catherinae</i> Thomas, 1909	Voucher	This study/ Paresque et al. (2004)
<i>Oligoryzomys nigripes</i> (Olfers, 1818)	Voucher	This study/ Paresque et. al (2004)
<i>Oxymycterus</i> sp.	Voucher	MBML
<i>Rhipidomys mastacalis</i> (Lund,1840)	Voucher	This study/Paresque et. al (2004)
<i>Thaptomys nigrita</i> (Lichtenstein, 1829)	Voucher	This study
Muridae		
<i>Rattus rattus</i> (Linnaeus, 1758)	Voucher	This study
Erethizontidae		
<i>Sphiggurus villosus</i> (F. Cuvier, 1823)	-	Paresque et al. (2004)
Cuniculidae		
<i>Cuniculus paca</i> ,(Linnaeus, 1758)	Camera trap	This study
Dasyproctidae		
<i>Dasyprocta azarae</i> Lichtenstein, 1823	Camera trap	This study/MBML
Echimyidae		
<i>Phyllomys pattoni</i> Emmons et al., 2002	Voucher	This study/MBML
<i>Trinomys paratus</i> (Moojen, 1948)	Voucher	This study/MBML/Paresque et al. (2004)

Table 2. Amphibian species from Reserva Biológica de Duas Bocas, State of Espírito Santo, southeastern Brazil. MBML = Museu de Biologia Prof. Mello Leitão; CFBH = Célio Fernando Batista Haddad collection.

Taxon	Type of record	References
Anura		
Craugastoridae		
<i>Haddadus binotatus</i> (Spix, 1824)	Voucher	This study/CFBH/MBML/Prado & Pombal (2005)
Brachycephalidae		
<i>Ischnocnema guentheri</i> (Steindachner, 1864)	Voucher	This study
<i>Ischnocnema oea</i> (Heyer, 1984)	Voucher	This study
<i>Ischnocnema</i> sp.	Voucher	This study
<i>Ischnocnema verrucosa</i> Reinhardt and Lütken, 1862	Voucher	This study/CFBH/Prado & Pombal (2005)
Strabomantidae		
<i>Euparkerella tridactyla</i> Izecksohn, 1988	Voucher	This study/CFBH/MBML
Hylidae		
<i>Aplastodiscus cavicola</i> (Cruz and Peixoto, 1985)	Voucher	This study/Prado & Pombal (2005)
<i>Aplastodiscus weygoldti</i> (Cruz and Peixoto, 1987)	Voucher	Prado & Pombal (2005)
<i>Bokermannohyla caramaschii</i> (Napoli, 2005)	Voucher	This study/Prado & Pombal (2005)
<i>Dendropsophus bipunctatus</i> (Spix, 1824)	Voucher	This study/CFBH/MBML/Prado & Pombal (2005)
<i>Dendropsophus branneri</i> (Cochran, 1948)	Voucher	CFBH/MBML/Prado & Pombal (2005)
<i>Dendropsophus decipiens</i> (A. Lutz, 1925)	-	Prado & Pombal (2005)
<i>Dendropsophus elegans</i> (Wied-Neuwied, 1824)	Voucher	This study/MBML/Prado & Pombal (2005)
<i>Dendropsophus microps</i> (Peters, 1872)	-	Prado & Pombal (2005)
<i>Dendropsophus minutus</i> (Peters, 1872)	Voucher	MBML
<i>Dendropsophus seniculus</i> (Cope, 1868)	Voucher	This study/MBML/Prado & Pombal (2005)
<i>Hypsiboas albomarginatus</i> (Spix, 1824)	Voucher	This study/CFBH/MBML/Prado & Pombal (2005)
<i>Hypsiboas albopunctatus</i> (Spix, 1824)	Voucher	This study/MBML/Prado & Pombal (2005)
<i>Hypsiboas faber</i> (Wied-Neuwied, 1821)	Voucher	This study/ MBML/Prado & Pombal (2005)
<i>Hypsiboas pardalis</i> (Spix, 1824)	Voucher	This study/MBML/Prado & Pombal (2005)
<i>Hypsiboas semilineatus</i> (Spix, 1824)	Voucher	CFBH/MBML/Prado & Pombal, (2005)
<i>Phasmahyla exilis</i> (Cruz, 1980)	Voucher	This study/CFBH/MBML/Prado & Pombal (2005)
<i>Phyllomedusa burmeisteri</i> Boulenger, 1882	-	Prado & Pombal (2005)
<i>Scinax alter</i> (B. Lutz, 1973)	Voucher	This study/ MBML/Prado & Pombal (2005)
<i>Scinax argyreornatus</i> (Miranda-Ribeiro, 1926)	Voucher	Thus study/MBML/Prado & Pombal (2005)
<i>Scinax cuspidatus</i> (A. Lutz, 1925)	-	Prado & Pombal (2005)
<i>Scinax fuscovarius</i> (A. Lutz, 1925)	Voucher	This study/MBML/Prado & Pombal (2005)
<i>Scinax</i> gr. <i>catharinae</i>	-	Prado & Pombal (2005)
<i>Scinax hayii</i> (Barbour, 1909)	Voucher	This study
<i>Scinax heyeri</i> (Peixoto and Weygoldt in Weygoldt, 1986)	Voucher	MBML
<i>Scinax humilis</i> (Lutz, 1954)	Voucher	MBML
<i>Scinax kautskyi</i> (Carvalho e Silva and Peixoto, 1991)	Voucher	This study/MBML
<i>Trachycephalus mesophaeus</i> (Hensel, 1867)	Voucher	Prado & Pombal (2005)
Centrolenidae		
<i>Vitreorana uranoscopa</i> (Müller, 1924)	Voucher	This study
Leptodactylidae		
<i>Leptodactylus</i> aff. <i>cupreus</i>	Voucher	This study
<i>Leptodactylus natalensis</i> A. Lutz, 1930	-	Prado & Pombal (2005)
<i>Leptodactylus ocellatus</i> (Linnaeus, 1758)	Voucher	This study/MBML/Prado & Pombal (2005)
Cycloramphidae		
<i>Proceratophrys laticeps</i> Izecksohn and Peixoto, 1981	Voucher	This study/CFBH/MBML/Prado & Pombal (2005)
<i>Proceratophrys schirchi</i> (Miranda-Ribeiro, 1937)	Voucher	This study/CFBH/MBML/Prado & Pombal (2005)
<i>Thoropa miliaris</i> (Spix, 1824)	Voucher	This study/MBML/Prado & Pombal (2005)
<i>Zachaeus carvalhoi</i> Izecksohn, 1983	Voucher	This study
Leiuperidae		
<i>Physalaemus crombiei</i> Heyer and Wolf, 1989	Voucher	This study/Prado & Pombal (2005)
<i>Physalaemus cuvieri</i> Fitzinger, 1826	Voucher	This study
Bufonidae		
<i>Dendrophryniscus</i> sp.	Voucher	This study
<i>Rhinella crucifer</i> (Wied-Neuwied, 1821)	Voucher	This study/MBML/Prado & Pombal (2005)
<i>Rhinella granulosa</i> (Spix, 1824)	Voucher	This study/MBML/Prado & Pombal (2005)

Table 2. Continued...

Taxon	Type of record	References
Hylodidae		
<i>Crossodactylus gaudichaudii</i> Duméril and Bibron, 1841	Voucher	This study/CFBH/MBML/Prado & Pombal (2005)
<i>Hylodes lateristrigatus</i> (Baumann, 1912)	Voucher	This study/Prado & Pombal (2005)
Mycrohylidae		
<i>Chiasmocleis capixaba</i> Cruz, Caramaschi and Izecksohn, 1997	Voucher	This study/Prado & Pombal (2005)
<i>Chiasmocleis carvalhoi</i> Cruz, Caramaschi, and Izecksohn, 1997	Voucher	This study
<i>Chiasmocleis schubarti</i> Bokermann, 1952	Voucher	This study/Prado & Pombal (2005)
Gymnophiona		
Caecilidae		
<i>Siphonops annulatus</i> (Mikan, 1820)	Voucher	This study

Table 3. Non-avian reptile species from Reserva Biológica de Duas Bocas, State of Espírito Santo, southeastern Brazil. MBML = Museu de Biologia Prof. Mello Leitão; IBSP-Herpeto = Alphonse Richard Hoge herpetological collection.

Taxon	Type of record	References
Testudines		
Chelidae		
<i>Hydromedusa maximiliani</i> (Mikan, 1820)	Voucher	This study/MBML
Squamata		
Polychrotidae		
<i>Anolis punctatus</i> Daudin, 1802	Voucher	This study
Leiosauridae		
<i>Enyalius brasiliensis</i> (Lesson, 1828)	Voucher	This study
Tropiduridae		
<i>Tropidurus torquatus</i> (Wied-Neuwied, 1820)	Voucher	This study
Phyllodactylidae		
<i>Gymnodactylus darwini</i> (Gray, 1845)	Voucher	This study
Gymnophthalmidae		
<i>Eupleopus gaudichaudii</i> Duméril & Bibron, 1839	Voucher	This study
<i>Heterodactylus imbricatus</i> Spix, 1825	Voucher	This study
<i>Leposoma scincoides</i> Spix, 1825	Voucher	This study
Teiidae		
<i>Tupinambis merianae</i> (Duméril and Bibron, 1839)	Voucher	This study
Scincidae		
<i>Mabuya macrorhyncha</i> Hoge, 1947	Voucher	This study
Anguidae		
<i>Diploglossus fasciatus</i> (Gray, 1831)	Voucher	This study/MBML
<i>Ophiodes</i> aff. <i>striatus</i>	Voucher	This study
Boidae		
<i>Boa constrictor</i> Linnaeus, 1758	Voucher	This study
Colubridae		
<i>Chironius bicarinatus</i> (Wied, 1820)	Voucher	This study
Dipsadidae		
<i>Dipsas catesbyi</i> (Sentzen, 1796)	Voucher	IBSP-Herpeto
<i>Dipsas incerta</i> Jan, 1863	Voucher	MBML
<i>Echianthera bilineata</i> (Fisher, 1885)	Voucher	This study
<i>Liophis reginae</i> (Linnaeus, 1758)	Voucher	This study
<i>Liophis poecilogyrus</i> (Wied, 1825)	Voucher	MBML
<i>Oxyrhopus formosus</i> (Wied-Neuwied, 1820)	Visual	This study
<i>Xenodon newwiedii</i> Günther, 1863	Voucher	This study
Elapidae		
<i>Micrurus corallinus</i> (Merrem, 1820)	Voucher	This study
Viperidae		
<i>Bothrops jararaca</i> (Wied-Neuwied, 1824)	Voucher	This study
<i>Bothrops jararacussu</i> Lacerda, 1884	Voucher	This study

Lacerda 2002, Srbek-Araújo & Chiarello 2008), but data showing the real impact of this species upon the native fauna are still scanty (Srbek-Araújo & Chiarello 2008). Although most dog attacks do not result in prey kill (Oliveira & Cavalcanti 2002), populations of *Canis familiaris* are potential disease and parasite reservoirs (Srbek-Araújo & Chiarello 2008).

Paresque et al. (2004) reported the occurrence of the short-tailed opossum *Monodelphis domestica* in this area, but we did not have access to their voucher specimen. We believe that its occurrence in the area, or even in the State of Espírito Santo, is unlikely because this species is found only in the drylands of central Brazil (Caatinga and Cerrado), eastern Bolivia, Northern Paraguay, and Northeastern Argentina (Chaco) (Gardner 2007). Considering the lack of a voucher and the geographic distribution of *M. domestica*, we did not include this species in our list of non-volant tetrapods from RBDB.

Among the amphibian species captured, approximately 80% are endemic to the Atlantic forest (Frost 2010). This result is expected, and is due to the fact that most of the evolutionary history of any frog community along the Brazilian coast is strongly connected to the Atlantic forest domain (Heyer et al. 1990). We obtained 12 new records of amphibians from RBDB: the anurans *Ischnocnema* sp. (Figure 2d), *Ischnocnema guentheri* (Figure 2b), *Ischnocnema oea* (Figure 2c), *Chiasmocleis carvalhoi*, *Dendrophryniscus* sp., *Zachaenus carvalhoi* (Figure 3f), *Bokermannohyla caramaschii* (Figure 2f), *Vitreorana uranoscopa* (Figure 3a), *Scinax hayii* (Figure 2o), *Physalaemus cuvieri* (Figure 3h), *Leptodactylus* aff. *cupreus* (Figure 3b), and the caecilian *Siphonops annulatus*. The species *Ischnocnema* sp. is a new species and is currently being described, and the specimen of *Dendrophryniscus* sp. was found dead and was in a bad state of conservation, making it difficult to identify the species level.

Nineteen new records of non-avian reptiles stem from the present paper: the lizards *Ophiodes* aff. *striatus*, *Ecpleopus gaudichaudii* (Figure 4b), *Heterodactylus imbricatus* (Figure 4c), *Leposoma scincoides* (Figure 4d), *Enyalius brasiliensis* (Figure 4a), *Anolis punctatus* (Figure 3p), *Gymnodactylus darwini*, *Mabuya macrorhyncha*, and the snakes *Boa constrictor*, *Chironius bicarinatus*, *Echinanthera bilineata*, *Liophis reginae* (Figure 4e), *Oxyrophus formosus* (Figure 4f), *Xenodon neuwiedii* (Figure 4g), and *Bothrops jararacussu*. These large numbers of new records of non-avian reptiles is due to the use of pitfalls (see below), and opportunistic searches.

Moreover, the success of sampling can be attributed to the fact that opportunistic searches were conducted at different times of day over a year, allowing the detection of species with seasonal occurrence and with different ecological habits (Auricchio & Salomão 2002).

The increase in the number of non-volant tetrapods species from RBDB is probably due to the use of multiple methods of capture. Other researchers carried out field expeditions in the area, but using mostly visual methods for detecting animals (Prado & Pombal 2005). The use of pitfall traps in this study greatly increased trapping success, probably being responsible for most new records. Many species recorded in pitfalls usually inhabit the leaf-litter, have fossorial habits, and are rarely observed during opportunistic searches.

The tree frog *Phasmahyla exilis*, the glass frog *Vitreorana uranoscopa*, the snake-necked turtle *Hydromedusa maximiliani* (Figure 3o) are on the list of threatened species in the State of Espírito Santo (Gasparini et al. 2007, Almeida et al. 2007) (Table 4). We found these three species in the Pau Oco Creek. We observed more than one individual of *Phasmahyla exilis* and usually heard its vocalization when we walked through the creek at night. *Vitreorana uranoscopa*, on the other hand, was detected only once in December 2007, when we found a male vocalizing. We found a male, a female and an egg of the turtle *Hydromedusa maximiliani* during the study. The male was recorded during an active search at night along the Pau Oco Creek, the female was accidentally captured in a Tomahawk trap set for small mammals, and the egg was collected during the opportunistic searches and later proved to belong to this species.

The study area houses the maned sloth *Bradypus torquatus* and the water opossum *Chironectes minimus*, which are both on the state list of threatened species (Chiarello et al. 2007). Mother and baby maned sloths were observed in the forest canopy (about 20 m above the ground) near the main trail for 15 minutes. Water opossums are seldom captured by traditional methods used for other small mammal species because of their semiaquatic and nocturnal habits (Galliez et al. 2009). One of us (J.L. Gasparini) saw one water opossum in the study area a few years before the beginning of the present fieldwork, but despite some directed effort to capture this species in the area by setting tomahawk traps along a creek and waterfall during the present study, we were unable to trap any individual. It is included on the checklist because *C. minimus* can be easily recognized in the field since it is the only semi-aquatic marsupial and shows a very characteristic black and white pelage.

Table 4. Conservation status of threatened non-volant tetrapods species recorded at Reserva Biológica de Duas Bocas according to regional, national, and international lists.

Taxon	Regional	National	International
Mammalia			
<i>Bradypus torquatus</i> Illiger, 1811	EN	VU	EN
<i>Chironectes minimus</i> (Zimmermann, 1780)	CR	-	LC
Amphibia			
<i>Euparkerella tridactyla</i> Izecksohn, 1988	DD	-	VU
<i>Ischnocnema oea</i> (Heyer, 1984)	DD	-	NT
<i>Phasmahyla exilis</i> (Cruz, 1980)	EN	-	LC
<i>Scinax kautskyi</i> (Carvalho e Silva and Peixoto, 1991)	DD	-	DD
<i>Vitreorana uranoscopa</i> (Müller, 1924)	VU	-	
<i>Zachaenus carvalhoi</i> Izecksohn, 1983	DD	-	DD
Testudines			
<i>Hydromedusa maximiliani</i> (Mikan, 1820)	VU	-	VU

DD: Data Deficient; LC: Least Concern; NT: Near Threatened; VU: Vulnerable; EN: Endangered; CR: Critically Endangered.



Figure 2. a) *Haddadus binotatus*; b) *Ischnocnema guentheri*; c) *Ischnocnema oea*; d) *Ischnocnema* sp.; e) *Euparkerella tridactyla*; f) *Bokermannohyla caramaschii*; g) *Dendropsophus branneri*; h) *Dendropsophus elegans*; i) *Dendropsophus seniculus*; j) *Hypsiboas albomarginatus*; l) *Hypsiboas albopunctatus*; m) *Hypsiboas faber*; n) *Phasmahyla exilis*; o) *Scinax hayii*; p) *Scinax kautskyi*. Photos by J.F.R. Tonini.

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Figure 3. a) *Vitreorana uranoscopa*; b) *Leptodactylus* aff. *cupreus*; c) *Leptodactylus ocellatus*; d) *Proceratophrys laticeps*; e) *Proceratophrys schirchi*; f) *Zachaenus carvalhoi*; g) *Physalaemus crombiei*; h) *Physalaemus cuvieri*; i) *Rhinella crucifer*; j) *Crossodactylus gaudichaudii*; l) *Hylodes lateristrigatus*; m) *Chiasmocleis capixaba*; n) *Chiasmocleis schubarti*; o) *Hydromedusa maximiliani*; p) *Anolis punctatus*. Photos by J.F.R. Tonini.

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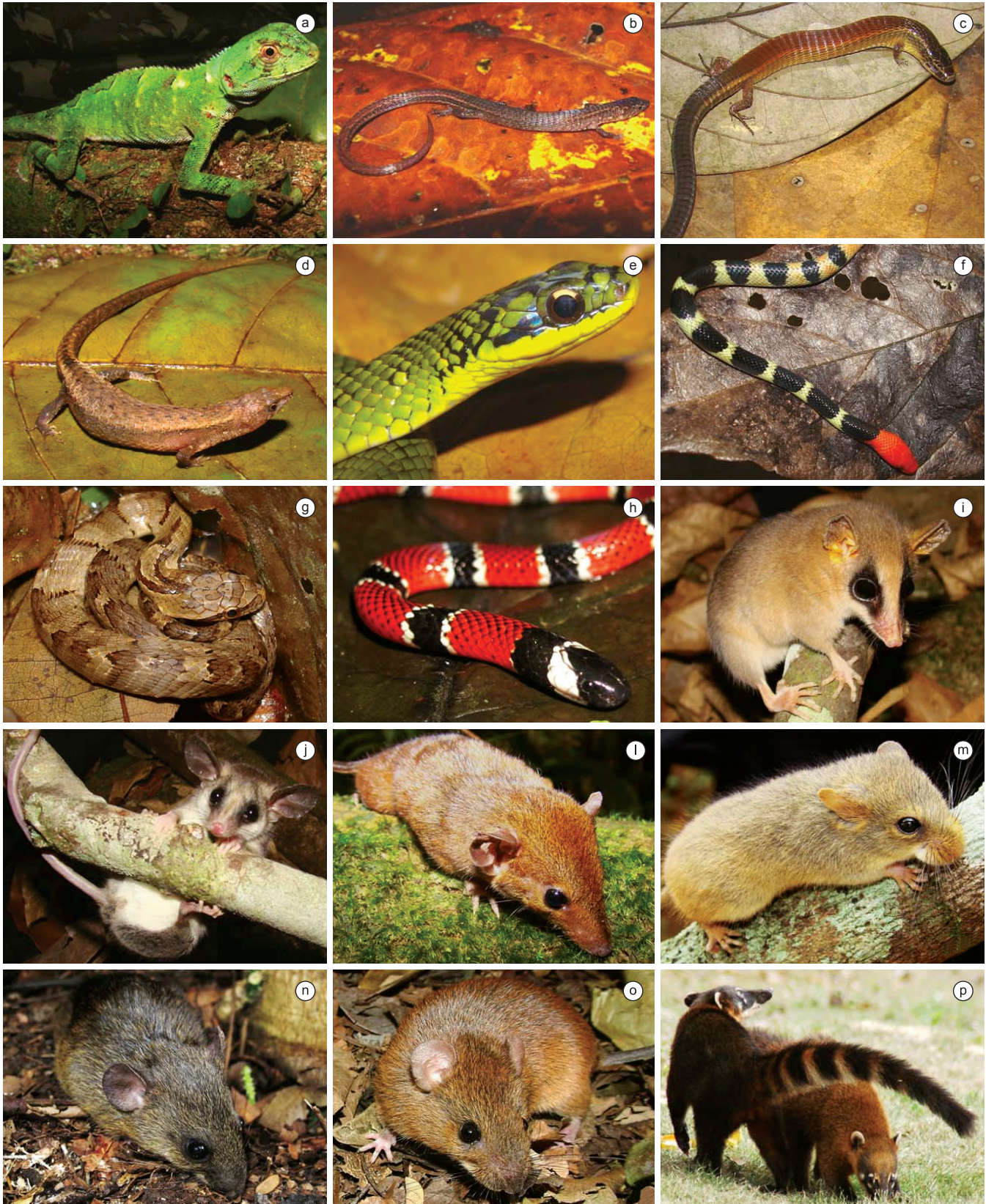


Figure 4. a) *Enyalius brasiliensis*; b) *Ecleopus gaudichaudii*; c) *Heterodactylus imbricatus*; d) *Leposoma scincoides*; e) *Liophis reginae*; f) *Oxyrophus formosus*; g) *Xenodon neuwiedii*; h) *Micrurus coralinus*; i) *Gracilinanus microtarsus*; j) *Marmosops incanus*; l) *Monodelphis iheringi*; m) *Juliomyia pictipes*; n) *Nectomys squamipes*; o) *Oecomys catherinae*; p) *Nasua nasua*. Photos by E. Ferreira, J.F.R. Tonini, L.P. Costa, P.L.V. Peloso and Y.L.R. Leite.

Our record of the snake *Oxyrophus formosus* is the southernmost, and highest in terms of elevation. Its previous southern limit was Mucuri, in the Southern part of the State of Bahia (Argôlo 2004). A.P. Almeida, J.L. Gasparini & A. Argôlo (unpublished data) have also recorded one specimen near Reserva Biológica de Sooretama, in the Northern part of Espírito Santo. The only individual of *O. formosus* captured during the present study escaped. Despite the lack of a voucher, this conspicuous species was included on the list, since our field identification was confirmed by specialists who examined its picture (A. Argôlo and A.P. Almeida, pers. commun.). *O. formosus* has a disjunct distribution, occurring in the Amazon and the Atlantic forest, and Argôlo (2004) discussed the possibility that this taxon in fact represents a composite species. We hope that the record of this species at Duas Bocas will stimulate and promote further field research in the area and its surroundings.

The reserve still houses little known anurans of uncertain conservation status, such as *Scinax kautskyi* (Figure 2p), *Ischnocnema oea*, *Euparkerella tridactyla*, and *Zachaeus carvalhoi* (Gasparini et al. 2007) (Table 4). Before this study, published records for *S. kautskyi*, *I. oea* and *Z. carvalhoi* were known only from their type locality, at Santa Teresa, State of the Espírito Santo, which is 40 km from RBDB. These four species had already been collected at RBDB by J.L. Gasparini, in 2003, and by C. Zamprogno, R.P. Bastos and A.P. Almeida, in 1989 (museum data; Appendix 1), but these records have not been published yet. Ferreira et al. (2010) recently expanded the geographic distribution of *Euparkerella tridactyla* to other localities in the State of Espírito Santo.

Chiasmocleis carvalhoi occurs in the States of São Paulo, Rio de Janeiro, and Bahia (Ilhéus), and it was recently recorded from Espírito Santo (Silva-Soares et al. 2010). In the present paper, we expand the occurrence of this species to RBDB, about 40 km NW of the previous record (Setiba, Espírito Santo) and to an elevation above 550 m, given that previous records are all from coastal lowlands.

We found the snake *Chironius bicarinatus* preying on an adult tree frog *Hypsiboas faber* near the Alto Alegre field station, where they both fell on the ground from a tree branch in December 2007. Pombal Jr. (2007) considered *Chironius bicarinatus* as a potential predator of anurans and Oliveira (2008) reported this snake preying on the tree frog *Hypsiboas pulchellus*.

All anurans and most mammals recorded at RBDB also occur at Santa Teresa, State of Espírito Santo, which is a hotspot of frog and mammal diversity (Rodder et al. 2007, Passamani et al. 2000). This status has been achieved as a direct result of constant collecting by well-trained herpetologists and mammalogists and a research tradition associated to a 50-year old local natural history museum (Museu de Biologia Professor Mello Leitão). Other important Atlantic forest fragments such as RBDB have been historically neglected in terms of faunal surveys, and we are only now beginning to evaluate their biological diversity. Reserva Biológica de Duas Bocas undoubtedly houses additional unrecorded species and long-term fieldwork is needed to complete its inventory. For example, park rangers mentioned the occurrence of the snake *Bothrops bilineatus*, but we did not find it. RBDB is located very close to the state capital, which is witnessing a wave of economical growth in recent years. Therefore human impact is rapidly growing in the surroundings, especially eucalyptus plantations, hunting and agriculture. This growth increases the fragility of the ecosystem in this area, which is becoming an island of forest within a matrix of urban and agricultural land uses. All efforts should be devoted to survey and protect the biodiversity of this important Atlantic forest remnant. More information about our project, including pictures of most species, can be found in the "Virtual Guide to the Fauna from Reserva Biológica de Duas Bocas" available at <http://www.chn.ufes.br/dbio/labs/lamab/duasbocas> (in Portuguese).

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Appendix 1: Voucher list

Abbreviations: CFBH = Célio Fernando Batista Haddad collection; MBML = Museu de Biologia Professor Mello Leitão; UFES = Universidade Federal do Espírito Santo; JFT = João Filipe Tonini's field numbers (these specimens will be catalogued at MBML). Specimens collected during the present study are underlined.

Amphibia

Anura: *Aplastodiscus cavicola* (CFBH 22496); *Aplastodiscus weygoldti* (MBML 6345); *Bokermannohyla caramaschii* (CFBH 22497); *Chiasmocleis capixaba* (CFBH 22498-99, 22500); *Chiasmocleis carvalhoi* (JFT311, 313, 323); *Chiasmocleis shubarti* (CFBH 22501); *Crossodactylus gaudichaudii* (JFT 127-29; CFBH 4023; MBML 5480); *Dendrophryniscus* sp. (CFBH 22502); *Dendropsophus bipunctatus* (CFBH 22503, 1361; MBML 5013); *Dendropsophus elegans* (CFBH 22504-05; MBML 5124); *Dendropsophus minutus* (MBML 5243); *Dendropsophus seniculus* (CFBH 22506; MBML 5488); *Euparkerella tridactyla* (CFBH 22526, 1360; MBML 5286-88); *Haddadus binotatus* (CFBH 22507-10, 940; MBML 4805); *Hylodes lateristrigatus* (CFBH 23606-08); *Hypsiboas albomarginatus* (CFBH 23617, 887; MBML 4824); *Hypsiboas albopunctatus* (CFBH 23618; MBML 5149); *Hypsiboas faber* (MBML 5216, 5368); *Hypsiboas pardalis* (CFBH 23631; MBML 5513); *Hypsiboas semilineatus* (CFBH 884; MBML 1344); *Ischnocnema guentheri* (JFT 468); *Ischnocnema oea* (CFBH 22517-20); *Ischnocnema* sp. (CFBH 22521-22); *Ischnocnema verrucosa* (CFBH 22523-25, 2272); *Leptodactylus* aff. *cupreus* (CFBH 23632, 23650); *Leptodactylus ocellatus* (CFBH 23633; MBML 4936); *Phasmahyla exilis* (CFBH 4022; MBML 1267); *Physalaemus crombiei* (CFBH 23636-39); *Physalaemus cuvieri* (CFBH 23640); *Proceratophrys laticeps* (CFBH 23609-12, 2271; MBML 4931); *Proceratophrys schirchi* (CFBH 2719-22, 4021; MBML 4940); *Rhinella crucifer* (CFBH 22492-94; MBML 1350); *Rhinella granulosa* (CFBH 22495; MBML 4953); *Scinax alter* (CFBH 23623; MBML 4772); *Scinax argyreornatus* (CFBH 23624; MBML 4904); *Scinax fuscovarius* (CFBH 23616; MBML 193); *Scinax hayii* (CFBH 23619-21); *Scinax heyeri* (MBML 5496); *Scinax humilis* (MBML 5410-12); *Scinax kautskyi* (CFBH 23625; MBML 6437); *Thoropa miliaris* (CFBH 23627-29; MBML 187); *Vitreorana uranoscopa* (CFBH 22527); *Zachaeus carvalhoi* (CFBH 23630).

Gymnophiona: *Siphonops annulatus* (CFBH 23626).

Sauropsida

Squamata: *Anolis punctatus* (JFT 459, 467); *Boa constrictor* (JFT 241); *Bothrops jararaca* (JFT 166, 261, 367, 422); *Bothrops jararacussu* (JFT 443); *Chironius bicarinatus* (JFT 369, 424, 439, 442); *Diploglossus fasciatus* (JFT 428; MBML 1844); *Dipsas catesbyi* (IBSP-Herpeto 25393); *Dipsas incerta* (MBML 1897); *Echivanthera bilineata* (JFT 458); *Ecpleopus gaudichaudii* (JFT 104, 282); *Enyalius brasiliensis* (JFT 20, 393); *Gymnodactylus darwini* (JFT 245-47); *Heterodactylus imbricatus* (JFT 36, 105-07, 133); *Hydromedusa maximiliani* (JFT 462; MBML 665); *Leposoma scincoides* (JFT 211-13); *Liophis reginae* (JFT 39, 423, 435, 460); *Mabuya macrorhyncha* (JFT 425); *Micrurus coralinus* (JFT 431); *Ophiodes striatus* (JFT 252); *Tropidurus torquatus* (JFT 121, 440); *Tupinambis merianae* (JFT 417). *Xenodon neuwiedii* (JFT 103, 441).

Mammalia

Didelphimorphia: *Didelphis aurita* (UFES 441-45; MBML 132); *Gracilinanus microtarsus* (UFES 446-58; MBML 82); *Marmosa murina* (MBML 99, 119, 97, 163); *Marmosops incanus* (UFES 461-74; MBML 96); *Metachirus nudicaudatus* (UFES 421-25, 475-86; MBML 103); *Micoureus paraguayanus* (UFES 496-501); *Monodelphis americana* (UFES 522-26); *Monodelphis iheringi* (UFES 509, 512-13, 516); *Philander frenatus* (UFES 546-47; MBML 122).

Cingulata: *Dasybus novemcinctus* (UFES 866); *Dasybus septemcinctus* (UFES 867-68);

Rodentia: *Akodon cursor* (UFES 548-52); *Blarinomys breviceps* (UFES 554-55); *Euryoryzomys russatus* (MBML 51, 138, 91); *Guerlinguetus ingrani* (MBML 78, 104); *Juliomys pictipes* (UFES 556-57); *Nectomys squamipes* (UFES 437, 558-568; MBML 89); *Oecomys catherinae* (UFES 569-70); *Oligoryzomys nigripes* (UFES 571-90); *Oxymycteris* sp. (MBML 117); *Phyllomys pattoni* (UFES 439-40, 620-24; MBML 226); *Rattus rattus* (UFES 618-19); *Rhipidomys mastacalis* (UFES 438, 598-615); *Thaptomys nigrita* (UFES 553, 616-17); *Trinomys paratus* (UFES 629-48; MBML 114).