A Survey of Wild and Introduced Marmosets (*Callithrix*: Callitrichidae) in the Southern and Eastern Portions of the State of Minas Gerais, Brazil

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Abstract: An understanding of current *Callithrix* distributions is important for species management and conservation in the wild because of widespread deforestation and the introduction, and spread, of marmosets outside their natural ranges. Here we report on a survey of the natural and introduced *Callithrix* distributions in five mesoregions of the southeastern Brazilian state of Minas Gerais: Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes and Vale do Rio Doce. Our study was based on on- and off-line literature searches, e-mail questionnaires, photographic records, and field observations. The collected data were superimposed on geopolitical and vegetation maps and compared to the historical records available in mammal collections from national museums and the currently recognized natural ranges of *Callithrix* species. *Callithrix geoffroyi* and *C. penicillata* were widely distributed in our study area, particularly in regions and biomes where they do not occur naturally. *Callithrix penicillata* has apparently expanded its range into the Atlantic Forest from its native Cerrado in Central Brazil. Although not native to Minas Gerais, *C. jacchus* was reported at several locations in our study area. These three species were reported even in the areas of natural occurrence of *C. aurita* and *C. flaviceps*. We detected the presence of five species of *Callithrix* (*C. aurita*, *C. flaviceps*, *C. geoffroyi*, *C. penicillata*, and *C. jacchus*) in some locations of the Zona da Mata. Marmoset groups are present in some urban areas, in close contact with human populations. Marmosets are found in artificial sympatry due to the introduction of species outside their native areas of distribution. Such introductions, together with the destruction of native marmoset habitats, are a threat due to ongoing hybridization.

Key words: Marmoset, threatened species, deforestation, introductions, Atlantic Forest, Cerrado

Resumo: A compreensão a respeito da distribuição atual das espécies de *Callithrix* é importante não apenas para seu manejo, mas também para sua conservação na natureza devido ao desmatamento e à introdução e disseminação de espécies fora das suas áreas naturais. Neste trabalho nós realizamos um levantamento sobre a distribuição natural e introduzida do gênero *Callithrix* em cinco mesorregiões do estado de Minas Gerais no sudeste do Brasil: Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes e Vale do Rio Doce. Nosso estudo baseou-se em pesquisas de literatura on-line e off-line, questionários enviados por e-mail, registros fotográficos e observações de campo. Os dados foram sobrepostos aos mapas geopolíticos e de vegetação, e comparados aos registros históricos disponíveis em coleções mastozoologicas de museus nacionais e as areas de distribuição natural atualmente reconhecidas para as espécies de *Callithrix*. O *Callithrix geoffroyi* e o *C. penicillata* foram amplamente distribuídos em nossa área de estudo, particularmente em regiões e biomas onde eles não ocorrem naturalmente. O *Callithrix penicillata*, aparentemente, expandiu sua área em direção à Mata Atlântica a partir de sua área nativa no Cerrado do

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Brasil Central. Embora não seja nativo de Minas Gerais, o *C. jacchus* foi relatado em vários locais da nossa área de estudo. Estas três espécies foram relatadas até mesmo nas áreas de ocorrência natural de *C. aurita* e *C. flaviceps*. Detectamos a presença de cinco espécies de *Callithrix* (*C. aurita*, *C. flaviceps*, *C. geoffroyi*, *C. penicillata* e *C. jacchus*) em alguns locais da Zona da Mata. Grupos de saguis estão presentes em algumas áreas urbanas, com contato muito próximo com populações humanas. As espécies de sagui são encontradas em simpatria artificial, devido à introdução de saguis fora de suas áreas nativas de distribuição. Essas introduções, juntamente com a destruição de seus habitats, são uma ameaça devido à hibridação.

Palavras-chave: Sagui, espécies ameaçadas, desmatamento, Mata Atlântica, Cerrado

Introduction

Of the non-Amazonian marmosets, *Callithrix*, *C. aurita* is classified as Vulnerable, *C. flaviceps* as Endangered, and *C. kuhlii* as Near Threatened; the remaining three species, *C. geoffroyi*, *C. penicillata*, and *C. jacchus* are ranked as Least Concern. *Callithrix aurita* and *C. flaviceps* are ranked as Endangered on Brazil's official list of threatened species (Brazil, ICMBio 2016). Both, along with *C. kuhlii*, are classified as Endangered on the threatened species list of the state of Minas Gerais (MG) (Fundação Biodiversitas 2007; Brazil, Minas Gerais, COPAM 2010)—the only Brazilian state that has wild populations of five of the six *Callithrix* species.

While *Callithrix* species are broadly allopatric (Coimbra-Filho *et al.* 1993), the illegal pet trade and deforestation have begun to break down the geographic separation of the different taxa (Rylands *et al.* 2009). Several accounts have reported the co-occurrence of native and allochthonous *Callithrix* across Brazil's Atlantic forest, mainly due to the introduction of *C. jacchus* (from the Northeast) and *C. penicillata* (from Central Brazil) (Ruiz-Miranda *et al.* 2000, 2006; Pereira *et al.* 2008; Rylands *et al.* 2009; Carvalho *et al.* 2013; Fuzessy *et al.* 2014; Rodrigues and Martinez 2014; Malukiewicz *et al.* 2014, 2015; Carvalho 2015). A consequence of these introductions is widespread hybridization among the different species in areas of artificial sympatry (Malukiewicz, in press).

In this study, we used on- and offline bibliographical searches, museum specimen localities, e-mail questionnaires, and photographic evidence to survey for the presence/absence of Callithrix species in key parts of the Brazilian state of Minas Gerais. Minas Gerais is the fourth largest Brazilian state. It has a population of 20 million inhabitants in 853 municipalities that are organized into geopolitical microregions and mesoregions (Brazil, IBGE 2010). The majority of the state (57%) is Cerrado (tropical bush savanna) and Atlantic Forest (41%) (Brazil, Minas Gerais, IEF-MG 2014). Minas Gerais has native populations of all *Callithrix* species except for the common marmoset, C. jacchus, the natural range of which is in Northeast Brazil, to the north (left bank) of the Rio São Francisco (Rylands et al. 1993, 2009; Malukiewicz et al. 2015). Callithrix jacchus was introduced to Minas Gerais (Mendes and Melo 2007; Rylands et al. 2009; J. Malukiewicz pers. obs.).

For this study, we focused our survey on five mesoregions (and their corresponding microregions) in Minas Gerais: 1) Campo das Vertentes, 2) Metropolitana de Belo

Horizonte (region of the state's capital), 3) Sul/Sudoeste de Minas, 4) Vale do Rio Doce, 5) Zona da Mata (Fig. 1). These mesoregions include the Cerrado and the Atlantic Forest and their transitional areas, besides heavily urbanized areas (Brazil, IBGE 2010). The Metropolitana de Belo Horizonte mesoregion is of particular importance as the economic, political, and administrative center of Minas Gerais (http://www.mg.gov.br). The mesoregions of Zona da Mata and Vale do Rio Doce are within the natural ranges of the endangered *C. aurita* and *C. flaviceps* (Hirsch 2003; Mendes and Melo 2007). Many major animal-trafficking routes pass through these mesoregions (RENCTAS 2001), resulting in the introduction of exotic species, particularly in fringe urban areas (Paula *et al.* 2005; Begotti and Landesmann 2008; Traad *et al.* 2012; Fuzessy *et al.* 2014).

In this study, we update information on the occurrence of introduced and native *Callithrix* and, comparing them with previously published reports, provide updated distribution maps of native and introduced marmosets for the mesoregions of the Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes and Vale do Rio Doce.

Methods

Data Collection

The protocol for this survey was approved by the Ethics Committee on Human Research of the Universidade Federal de Viçosa, number 1.064.333 (CAAE: 42727015.7.0000.5153). The data sources for the distribution of native and exotic marmosets in the municipalities included: museum specimen localities, on- and offline literature searches, e-mail questionnaires, literature reviews, photographic records, and sightings in the field. We carried out the e-mail-based survey from September 2012 to February 2013. It consisted of a questionnaire of eight questions, along with illustrations of all six Callithrix species (see Supplementary Information 1). We asked respondents to confirm the presence of marmosets within a given municipality, to identify the species, and to identify whether the observed setting was rural, urban, or captive. Questionnaire illustrations were those of Stephen D. Nash, featured in Ford et al. (2009).

We preferentially, but not exclusively, opted to send the questionnaire out to people and institutions that were knowledgeable about local faunas and rural areas outside of cities.

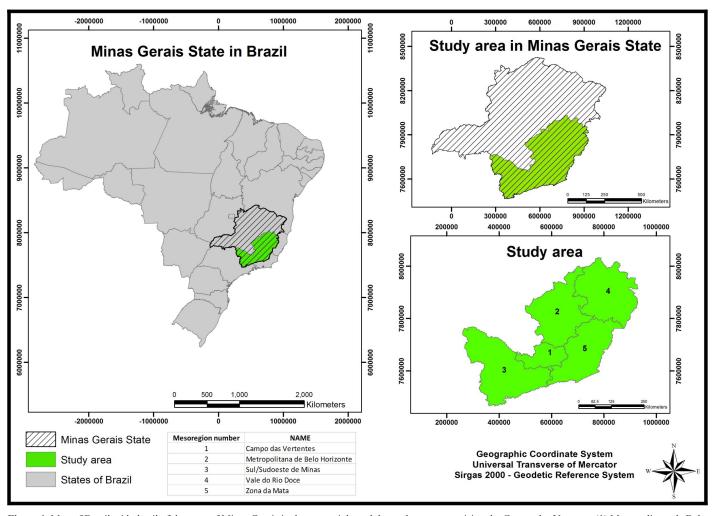


Figure 1. Map of Brazil with detail of the state of Minas Gerais in the upper right and the study area comprising the Campo das Vertentes (1) Metropolitana de Belo Horizonte (2), Sul/Sudoeste de Minas (3), Vale do Rio Doce (4), and Zona da Mata (5) in the lower right.

The institutions and people that we contacted included the Instituto Mineiro de Agropecuária (IMA), the Empresa de Assistência Técnica e Extensão Rural do Estado de Minas Gerais (EMATER), and biology teachers, and colleges of environmental sciences, biology, and agriculture. Potential contact information was found through search engines (such as Google) and social networking sites (such as Facebook). The e-mail message included an explanation about our research objectives and the questionnaire (see Supplementary Information 1). Besides our standardized survey questions, e-mail respondents were encouraged to add personal observations of physical and behavioral marmoset descriptions at the end of the questionnaire.

We scaled the number of contacts we made in each municipality for the mesoregions of Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes, and Vale do Rio Doce, in accordance with the number of inhabitants in each. For example, (1) for municipalities with up to 20,000 inhabitants at least three potential respondents were contacted, (2) for municipalities with 20,000 to 50,000 inhabitants, a minimum of six potential respondents were contacted, and (3) for municipalities having between 50,000 and 100,000 inhabitants, a minimum of nine potential respondents were contacted. E-mail questionnaire replies

were compiled into a spreadsheet and analyzed qualitatively as frequency data in SPSS 17.0 (see data analysis below). Marmoset photographs received from respondents helped us confirm the presence of marmosets in the municipalities of our mesoregions of interest. We acquired many photographs through colleagues who observed and photographed marmosets while in the field.

Our literature search included the use of online resources such as Google search, Google Scholar search, and the Web of Science bibliographic data. Keywords used included the following in both Portuguese and English: "Callithrix," "sagui," "Minas Gerais," "Mata Atlântica," "Cerrado," "Metropolitana de Belo Horizonte, " "Vale do Rio Doce," "Sul/Sudoeste de Minas," "Campo das Vertentes," "Zona da Mata," as well as the names of the microregions and municipalities located in the mesoregions. We also used articles, books and faunal inventories (reports made available online by environmental consulting agencies of faunal assemblages within a specific geographic area) collected opportunistically. Databases of biological conference proceedings were also searched using the same keywords as above. The proceedings searched covered all volumes from 2002 to 2013 for the following conferences: Brazilian Ecology Congress (Congresso de Ecologia do Brasil), Annual Ethology Meeting (Encontro Anual

de Etologia), Brazilian Mastozoology Congress (Congresso Brasileiro de Mastozoologia), Brazilian Primatology Congress (Congresso Brasileiro de Primatologia) and Brazilian Zoology Congress (Congresso Brasileiro de Zoologia). We also included in our final dataset the marmoset sightings made by the authors.

Finally, we visited several Brazilian mammal collections to verify historical marmoset distributions in Minas Gerais. The Museu de Zoologia João Moojen UFV (MZUFV) was visited by F. Silva and F. Coelho. Records from the Museu de Zoologia da USP (MZUSP), Museu Nacional (MNRJ), Museu de Ciências Naturais PUC Minas (MCN-M) and Coleção Mastozoologica da UFMG (UFMG) were provided by V. Bardi. Curators of these collections authorized our use of the information for the purposes of our survey.

Data Analysis

Responses to our e-mail survey were collated and entered into a single EXCEL spreadsheet. Questionnaire data were classified as qualitative or nominal, and SPSS 17.0 was used for descriptive analyses to calculate the frequency of occurrence of each questionnaire response. Confidence indices were created to evaluate the reliability of the collected data according to the following criteria: 1) minimally reliable due to three or less questionnaire responses indicating the presence of a species in a region; 2) reliable with more than three questionnaire responses indicating the presence of a species in a region; and 3) highly reliable with more than three questionnaire responses and/or an additional data source (photograph, the literature, or field sighting) indicating species presence in an area.

To visualize the data, we superimposed the reported occurrences of Callithrix marmosets on maps based on biomes, microregions, and the currently recognized natural distributions of Callithrix as proposed by Rylands et al. (2009). Due to the similarity of *Callithrix* geographical distribution maps in Rylands et al. (2009) and those of the IUCN Red List of Threatened Species, shapefiles for the Callithrix distributions used in this work were downloaded directly from the IUCN site (http://www.iucnredlist.org/). Our maps were created with the ArcGIS10.4.1 geoprocessing software. Base data shapefiles were downloaded from files labeled as "Brasil e Estados Políticos", "Minas Gerais Político", "Mesorregiões", "Microrregiões", "Municípios de Minas Gerais" and "Biomas de Minas Gerais" from the website <ftp://geoftp.ibge.gov.br>. Data for presence/absence of Callithrix species for each city were inserted into these maps with the "Join" tool, where an Excel table was added to the table of attributes for cities in the study area.

Results

The totals of different records and resources obtained for each species are as follows: (1) *C. aurita* – 49 questionnaire responses, seven photographic records, 21 bibliographic references, and two field sightings; (2) *C. flaviceps* – 12

questionnaire responses, three photographic records, 23 bibliographic references and two field sightings; (3) C. geoffroyi - 63 questionnaire responses, seven photographic records, 12 bibliographic references and no field sightings; (4) C. jacchus - 59 questionnaire responses, five photographic records, three bibliographic references and one field observation; and (5) C. penicillata - 73 questionnaire responses, 48 photographic records, 44 bibliographic references and 15 field sightings. None of our information sources indicated the presence of Callithrix kuhlii in our geographic region of interest. Table 1 shows the confidence indices for the results compiled from these various sources. All reported results are organized in Table S1, Supplementary Information, by the city, microregion, and mesoregion from which these data were originally obtained. A selection of photos obtained during our survey is shown in an appendix.

Questionnaire responses

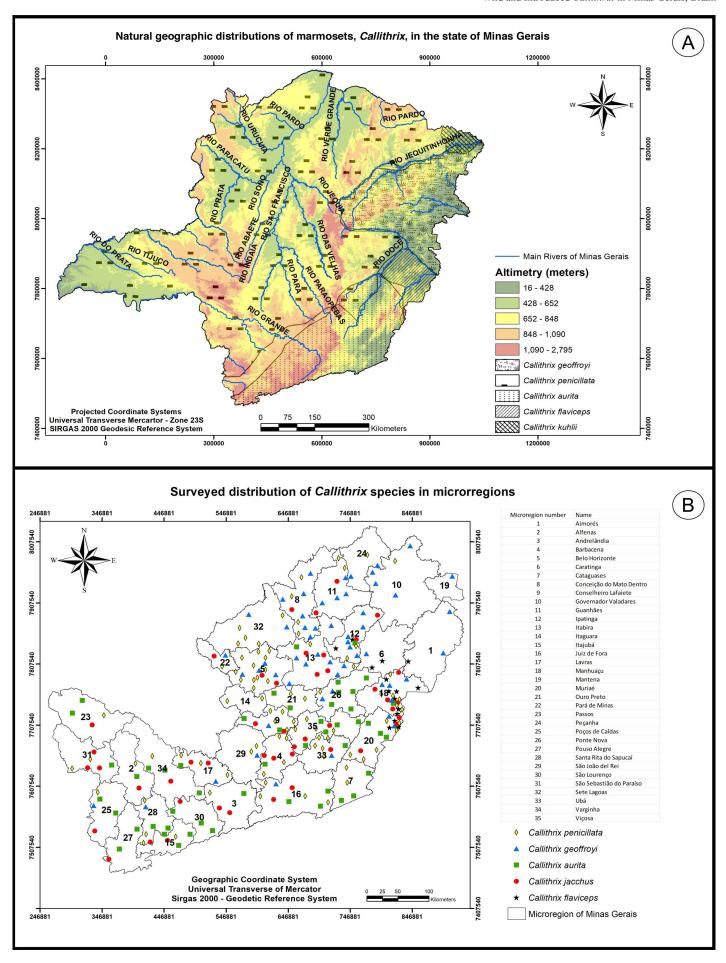
A total of 2,391 e-mail questionnaires were sent out, and we considered e-mail transmission to the intended recipient a success if the message did not bounce back in error. In all, 2291 e-mails were successfully transmitted to recipients, and we received 279 responses (12% of all successfully transmitted emails). Table 2 shows the response results of our questionnaire, broken down by five out of the eight questionnaire questions. The three omitted questions dealt with marmoset species identifications from supplied pictures, and whether respondents noted marmoset presence in any locations beyond their own area of residence.

The qualitative analysis indicated that most respondents (92%) had observed some marmosets within their respective municipalities. Respondents indicated that the animals were frequently present in urban areas (69.5%), areas of preserved forest (36%), and on rural properties (57%). Regarding group size, 45% of respondent answers indicated that social groups had five to ten animals, but 43% of the respondents did not know if the numbers of animals had recently changed or not in their respective regions. Fifty-seven percent of the respondents indicated that the marmosets they observed seemed accustomed to human presence.

Museum specimen records

Information on the *Callithrix* specimens present in different Brazilian mammal collections is shown in Table 3. These data indicated the presence of *C. aurita* in the Zona da Mata mesoregion, in the microregions of Cataguases (record from 1934 to 1943), Juiz de Fora (records from 1934 to 1953), and Viçosa (record in 1939), all in the Atlantic Forest. *Callithrix flaviceps* was indicated for the microregion of Caratinga, in the mesoregion of Vale do Rio Doce, Atlantic Forest (year 1995).

Museum specimen records indicated the occurrence of *C. geoffroyi* in various mesoregions. In the Zona da Mata, specimens were collected in Atlantic Forest of the microregion of Ponte Nova in 1940. *Callithrix geoffroy* specimens were also collected between 1954 and 1960 in the Metropolitana de



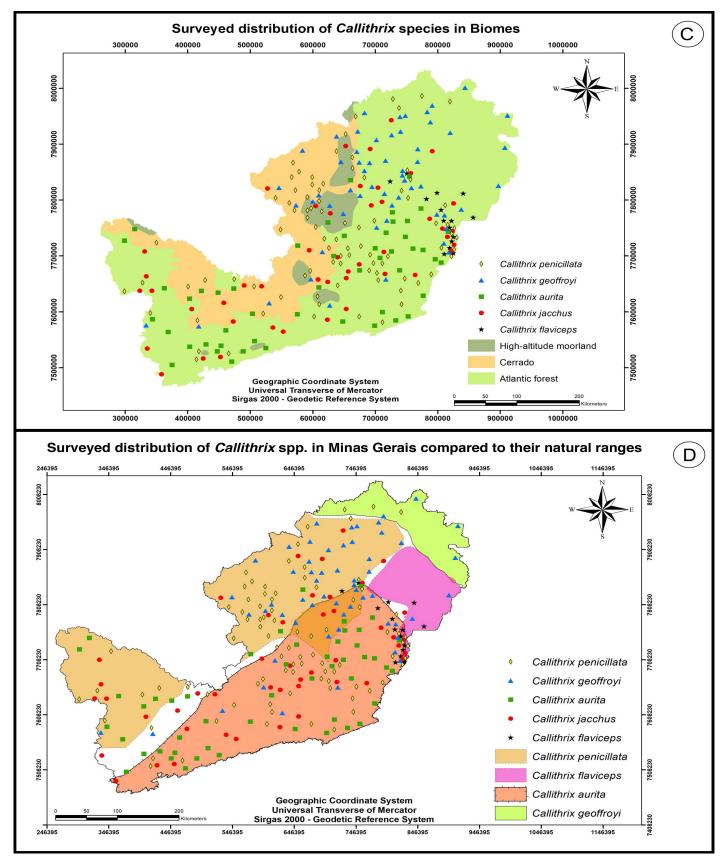


Figure 2 A-D. Survey records of *Callithrix* occurrences superimposed on the geographical species distributions based on IUCN shapefiles and Rylands *et al.* (2009). (2A) Map of altimetry and principle river drainages in Minas Gerais superimposed on the natural distribution of *Callithrix* following IUCN shapefiles and Rylands *et al.* (2009). In 2B-2D, geometric shapes for *Callithrix* species indicate surveyed species occurrences throughout Minas Gerais. (2B) Map is based on the 35 microregions that encompass the MG mesoregions of Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes and Vale do Rio Doce. (2C) Represents the biomes encompassing the MG mesoregions of Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes and Vale do Rio Doce. (2D) Shows natural *Callithrix* distributions within the five MG mesoregions of Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes and Vale do Rio Doce.

Belo Horizonte, in the transition between Atlantic Forest and Cerrado, microregion of Conceição do Mato Dentro.

The occurrence of *C. penicillata* was indicated for three mesoregions. Specimens were collected in 1946 in the Sul/Sudoeste de Minas, in a transition between Atlantic Forest and Cerrado in the microregion of Passos. The species was also collected in the Metropolitana de Belo Horizonte in the microregions of Ouro Preto, a transitional area between the Atlantic Forest, Cerrado and Campo Rupestre (high elevation moorland) in 1988; Belo Horizonte, in a transition of Cerrado and Campo Rupestre in 1942, 1999 and 2000; Sete Lagoas in Cerrado in 2006; and Itabira, in Atlantic Forest/Cerrado in 2008. It was also collected in the Zona da Mata, in the Atlantic Forest in the microregions of Viçosa in 1996, 1998, and 2002, and Juiz de Fora in 2002.

Survey results

Table 1 lists the reported occurrences of *Callithrix* in Minas Gerais in the mesoregions of Metropolitana de Belo Horizonte, Sul/Sudoeste de Minas, Zona da Mata, Campo das Vertentes, and Vale do Rio Doce. Figure 2A shows the natural distribution of *Callithrix* in Minas Gerais, following IUCN (2017) and Rylands *et al.* (2009), superimposed on a map of the state's principal rivers and mountain chains. In the rest of Figure 2, we show our records superimposed on three different maps: Fig. 2B shows the 35 microregions that encompass our five mesoregions; Fig. 2C, shows the records superimposed on the regions' biomes; and Fig. 2D shows the natural areas of occurrence (Rylands *et al.* 2009).

We received reports of C. aurita in 56 municipalities in the five mesoregions. The majority (40+) were within its natural range as proposed by Rylands et al. (2009). The most reliable reports (confidence score 3) of the occurrence of C. aurita in its natural range were in the Atlantic Forest in all microregions of the Zona da Mata mesoregion except for Ubá, Ponte Nova and Manhuaçu (confidence score 1); and in the Sul/Sudoeste de Minas in the microregions of São Sebastião do Paraíso (Atlantic Forest), Alfenas (Cerrado/Atlantic Forest transition), Poços de Caldas (Atlantic Forest), Pouso Alegre (Atlantic Forest), Santa Rita do Sapucaí (Cerrado/Atlantic Forest transition) and São Lourenço (Atlantic Forest). The most reliable reports (confidence score of 3) of C. aurita found outside of its supposed natural range (Rylands et al. 2009) were in the microregions of São Sebastião do Paraíso, Poços de Caldas and Alfenas. All were believed to form part of the natural range of *C. penicillata*.

Callithrix flaviceps was described in 15 municipalities. It was reported in the natural area of occurrence of *C. aurita* in the microregion of Muriaé, and close to the limits of the ranges of the two species in the microregion of Manhuaçu; both are in the Zona da Mata. Additionally, *C. flaviceps* was reported in two cities from the Ipatinga microregion (Vale do Rio Doce), described by Rylands *et al.* (2009), to be an area close to the natural range borders of *C. penicillata*, *C. flaviceps*, and *C. aurita*. All reported occurrences for *C. flaviceps*

had a high confidence score of 3, and all these microregions where this species was reported are within the Atlantic Forest.

We received reports of C. geoffroyi in 57 municipalities, but only four were from the species' natural range indicated by Rylands et al. (2009). The species was found in more than 30 municipalities in the range of C. penicillata as portrayed by Rylands et al. (2009). Although the species is considered be native to the Atlantic Forest (Rylands et al. 1996), high confidence reports (confidence score 3) were obtained for the presence of the species in Cerrado (microregions of Sete Lagoas and Conceição do Mato Dentro, Metropolitana de Belo Horizonte). Reports with a confidence score of 2 indicated its occurrence in Campo Rupestre (microregion of Belo Horizonte). This species was also reported in the ranges of C. aurita and C. flaviceps and in the limits to the ranges C. flaviceps and C. aurita, as well as C. penicillata and C. aurita. The IUCN Callithrix distribution shapefiles (http://www.aperiles.com/ iucnredlist.org/>) show overlap in the ranges of *C. penicillata* and C. aurita in the microregion of Itabira, though Rylands et al. (2009) limit these species' distribution in this area with no overlap. Nonetheless, we also received reliable reports (confidence score 3) for the presence there of C. geoffroyi. In the mesoregions of Campo das Vertentes (microregions Lavras and São João del Rei) and Sul/Sudoeste de Minas (microregions Poços de Caldas and Santa Rita do Sapucaí) the occurrence of C. geoffroyi was supported by data with confidence scores of just 1. Confidence index values for the remaining mesoregions varied between the respective microregions, as can be seen in Table 1.

Callithrix penicillata was reported in 88 municipalities in all five mesoregions, and more than 30 are located in the natural range *C. aurita*, as indicated by Rylands *et al.* (2009). Although *C. penicillata* is broadly endemic to the Cerrado (Rylands *et al.* 1996, 2009), we obtained reports (confidence score 3) of its occurrence in the Atlantic Forest in the microregions of Santa Rita do Sapucaí, Juiz de Fora, Cataguases, Barbacena and São João del Rei. Of the 35 microregions of the study, the presence of *C. penicillata* was reported with a confidence score of 3 for 23 microregions, with a confidence score of 1 for seven microregions, and not reported for just five microregions (Table 1).

We have records of the presence of *C. jacchus* in 46 municipalities in Minas Gerais. Those with a confidence score of 3 were in the Zona da Mata (microregions of Manhuaçu, Viçosa, Muriaé and Juiz de Fora in Atlantic Forest); the Sul/Sudoeste de Minas (microregions of São Sebastião do Paraíso and Itajubá in the Atlantic Forest), and Campo das Vertentes (microregions of Lavras and Barbacena in a Cerrado and Atlantic Forest transition). Records with a confidence score of 2 came from Campo Rupestre in the microregion of Belo Horizonte, and those with a confidence score of 1 came from areas in the microregions of Pará de Minas and Conselheiro Lafaiete. We also received photos of mixed groups of *C. jacchus* and *C. penicillata* in Cerrado in the Lavras microregion. In the microregion of Muriaé (Atlantic Forest in the

Table 1. Callithrix species found in each microregion of Metropolitana de Belo Horizonte, Vale do Rio Doce, Sul/Sudoeste de Minas, Campo das Vertentes and Zona da Mata. The numbers indicate data confidence rates as follows: "1" minimally reliable; "2" reliable; "3" highly reliable. Empty cells indicate absence of information for a region

Mesoregion	Microregion	Microregion coordinates	C. aurita	C. flaviceps	C. geoffroyi	C. jacchus	C. penicillata
Campo das Vertentes	Lavras	44°53'22"W, 21°21'15"S	1	-	1	3	3
	São João del-Rei	44°17'08"W, 21°08'35"S	-	-	1	ı	3
	Barbacena	43°44'40"W, 21°12'31"S	1	-	-	2	3
Metropolitana de Belo Horizonte	Sete Lagoas	44°04'16"W, 19°16'51"S	-	-	3	-	3
	Conceição do. Mato Dentro	43°26'52"W, 18°48'52"S	-	-	3	1	3
	Pará de Minas	44°39'06"W, 19°47'07"S	-	-	1	1	3
	Belo Horizonte	44°02'00"W, 19°54'33"S	-	-	2	2	3
	Itabira	43°10'53"W, 19°50'50"S	1	-	3	2	3
	Itaguara	44°13'42"W, 20°24'49"S	-	-	-	-	3
	Ouro Preto	43°28'18"W, 20°23'23"S	1	-	-	-	3
	Conselheiro Lafaiete	43°52'17"W, 20°42'14"S	1	-	1	1	3
Vale do Rio Doce	Guanhães	42°52'36"W, 18°46'52"S	-	-	2	1	1
	Peçanha	42°26'54"W, 18°13'28"S	-	-	3	-	1
	Governador Valadares	41°51'30"W, 18°40'53"S	-	-	2	-	1
	Mantena	41°11'31"W, 18°39'10"S	-	-	1	-	-
	Ipatinga	42°36'20"W, 19°22'17"S	1	3	3	2	3
	Caratinga	42°08'04"W, 19°37'42"S	-	3	3	-	-
	Aimorés	41°24'40"W, 19°37'08"S	-	3	3	-	-
Sul/Sudoeste de Minas	Passos	46°40'45"W, 20°37'05"S	1	-	-	1	3
	São Sebastião do Paraíso	46°45'53"W, 21°10'13"S	3	-	-	3	3
	Alfenas	46°00'12"W, 21°24'29"S	3	-	-	1	1
	Varginha	45°31'22"W, 21°20'47"S	1	-	-	1	3
	Poços de Caldas	46°22'28"W, 21°58'11"S	3	-	1	1	1
	Pouso Alegre	46°05'55"W, 22°25'53"S	3	-	-	1	-
	Sanat Rita Sapucaí	45°38'13"W, 22°05'19"S	3	-	1	1	3
	São Lourenço	44°57'06"W, 22°05'02"S	3	-	-	1	-
	Andrelândia	44°25'07"W, 21°55'20"S	1	-	-	1	1
	Itajubá	45°21'39"W, 22°26'44"S	1	-	-	3	3
Zona da Mata	Ponte Nova	42°46'15"W, 20°1445"S	1	-	3	-	1
	Manhuaçu	42°03'47"W, 20°16'53"S	1	3	3	3	3
	Viçosa	43°00"53"W, 20°44'44"S	3	-	-	3	3
	Muriaé	42°17'20"W, 20°55'48"S	3	3	3	3	3
	Ubá	43°0114"W, 21°09'43"S	1	-	1	1	3
	Cataguases	42°35'54"W, 21°33'59"S	3	-	-	-	3
	Juiz de Fora	43°28'00"W, 21°45'46"S	3	-	1	3	3

Question	Response Percentages						
Have you observed marmosets in your municipality?	Yes (92%)		No (8%)				
Where have you observed marmosets in your municipality?	Preserved forest area (36%)	Rural area (57%)	Urban (69,5%)	Captivity (4%)	Other location (15%)		
How many animals do you observe regularly?	Up to 4 animals (36%)	Between 5 and 10 animals (45%)	More than 10 animals (15%)	Respondents indicated more than one response (4%)			
Has the number of marmosets within your municipality changed in the last few years?	Yes, the quantity of animals has decreased (20%)	Yes, the quantity of animals has increased (28%)	No, the quantity of animals has not changed (9%)	I don't know (43%)			
Do marmosets seem habituated to people?	Yes (57%)	No (37%)	I don't know (6%)				

Table 2. Results presented as percentages for each set of possible responses to select questions from the email-questionnaire

Zona da Mata), there are reliable data (confidence score 3) for the simultaneous presence of *C. aurita*, *C. flaviceps*, *C. geoffroyi*, *C. penicillata*, and *C. jacchus*.

Discussion

The questionnaire response rate was considerable (12%), given that online surveys usually produce a low response rate (Gonçalves 2008) of around 7% (Kuter and Yilmaz 2001). Only in rare cases, do online survey response rates reach more than 20% (Masser and Schonrok 2006). Caveats regarding our data collection approach include the difficulty of fully gauging the capability of e-mail survey respondents to identify marmoset species using the illustrations we provided. In some instances, we were unable to determine whether a species was native or introduced, and whether some species expansions were natural or human-induced. We tried to overcome some of this possible error by combining information from collective evidence gathered from our searches and surveys to establish a credibility rating for our data.

Historical occurrence of Callithrix in the five mesoregions

Specimens of *Callithrix* collected during the early to late 20th century corroborated accounts of their currently accepted distributions as described in the literature, as well as our data, for four of the six of species. Specimen records for *C. aurita* show the presence of this species in the Atlantic Forest in the Zona da Mata mesoregion of Minas Gerais, as indicated by Rylands *et al.* (2009). These observations were corroborated by the findings from our study. The single *C. flaviceps* specimen we had access to was collected in the area of Caratinga in the Vale do Rio Doce mesoregion, which fits the species' distribution indicated by our data and Rylands *et al.* (2009). Specimen records for *C. aurita* are biased towards

the early to mid-20th century, and trail off toward the latter part of the century. One of the last published accounts of *C. aurita* in the Zona da Mata mesoregion was from the town of Viçosa in 1995 (Pereira *et al.* 1995) that today is occupied by invasive marmosets (Fuzessy *et al.* 2014). Although a group of *C. aurita* was recently encountered in Viçosa for the first time in decades (Vital 2017), the species' conservation status still remains alarming and worrisome.

One surprising result indicated by museum specimen localities was the presence of C. geoffroyi in Rio Doce (microregion of Ponte Nova / mesoregion of Zona da Mata) and Conceição do Mato Dentro (microregion of Conceição do Mato Dentro / mesoregion of Metropolitana de Belo Horizonte). While this pattern was further corroborated by our data, these areas were not included in the range of C. geoffroyi as indicated by Rylands et al. (2009). Ávila-Pires (1969) and Hershkovitz (1977) do, however, mention the occurrence of C. geoffroyi in these two areas. Ávila-Pires (1969) discussed the reports of C. geoffroyi in the Zona da Mata mesoregion during the 1930s and 1940s. However, it was not clear the whether the name of "Rio Doce" where museum specimens were sampled and the place referred to by Ávila-Pires (1969) and Hershkovitz (1977) meant the municipality or river of the same name. Hershkovitz (1977) also cited the occurrence of this species in the locality of Visconde do Rio Branco, Zona da Mata mesoregion. More recently, Carvalho (2015) found possible mitochondrial DNA evidence of C. geoffroyi in an individual from a C. penicillata group in Juiz de Fora (Zona da Mata). The isolated instances of C. geoffroyi in the state of Minas Gerais could be the result of deforestation of the Atlantic Forest along with introductions (Ávila-Pires 1969; Coimbra-Filho 1971; Coimbra-Filho et al. 1993).

Museum specimen localities showing the occurrence of *C. penicillata* in the mesoregions of Sul/Sudoeste de Minas

Table 3. *Callithrix* specimens present in the collections of the Museu de Zoologia da Universidade de São Paulo (MZUSP), Museu Nacional do Rio de Janeiro (MNRJ), Museu de Ciências Naturais da Pontificia Universidade Católica de Minas Gerais (MCN-M), Museu da Universidade Federal de Minas Gerais (UFMG), and Museu de Zoologia João Moojen da Universidade Federal de Viçosa (MZUFV).

Species	Museum collection	Specimen Number	Year	City	Microregion	Mesoregion
C. aurita	MNRJ	1354 1355	1934 1934	Além Paraíba	Cataguases	Zona da Mata
C. aurita	MNRJ	2603	1943	Volta Grande	Cataguases	Zona da Mata
C. aurita	MNRJ	23737 23739 23740	1953 1953 1953	Rio Novo	Juiz de Fora	Zona da Mata
C. aurita	MNRJ	2817	1934	Belmiro Braga	Juiz de Fora	Zona da Mata
C. aurita	MZUFV	0004	1939	Viçosa	Viçosa	Zona da Mata
C. flaviceps	MZUFV	0489	1995	Caratinga	Caratinga	Vale do Rio Doce
C. geoffroyi*	MZUSP	5923 5924 5926 5927	1940 1940 1940 1940	Rio Doce*	Ponte Nova	Zona da Mata
C. geoffroyi	MNRJ	13481 13482 13483 23756	1954 1954 1954 1969	Conceição do Mato Dentro	Conceição do Mato Dentro	Metropolitana de Belo Horizonte
C. penicillata	MNRJ	8810 9091 9848 10378 10657 10949 28857 28858 28859	1946 1946 1946 1946 1946 1946 1946 1946	São João Batista do Glória	Passos	Sul/Sudoeste de Minas
C. penicillata	MZUSP	28534	1988	Itabirito	Ouro Preto	Metropolitana de Belo Horizonte
C. penicillata	MNRJ	4230 4231 4232 4233 4234	1942 1942 1942 1942 1942	Lagoa Santa	Belo Horizonte	Metropolitana de Belo Horizonte
C. penicillata	MCN-M	667	1999	Lagoa Santa	Belo Horizonte	Metropolitana de Belo Horizonte
C. penicillata	MCN-M	1952	2006	Santana do Riacho	Sete Lagoas	Metropolitana de Belo Horizonte
C. penicillata	MCN-M	2470	2008	Rio Piracicaba	Itabira	Metropolitana de Belo Horizonte
C. penicillata	UFMG	958	-	Baldim	Sete Lagoas	Metropolitana de Belo Horizonte
C. penicillata	UFMG	3823 4093	2000	Belo Horizonte	Belo Horizonte	Metropolitana de Belo Horizonte
C. penicillata	MZUFV	0684 0685 1096	1998 1996 2002	Viçosa	Viçosa Zona da Mata	
C. penicillata	MZUFV	0792	2002	Rio Novo	Juiz de Fora	Zona da Mata

^{*}Please refer again to "Historical occurrence of Callithrix in the five mesoregions" in the text.

and Metropolitana de Belo Horizonte are in accordance with our data as well as the information given in Rylands *et al.* (2009). Specimens of *C. penicillata* from the Zona da Mata are not, however, part of the species' distribution as recognized by Rylands *et al.* (2009). Our survey data also indicated the species' occurrence there. While *C. penicillata* is considered broadly endemic to the Cerrado (Carmignotto *et al.* 2012), it is commonly introduced throughout the Brazilian Atlantic Forest (Coimbra-Filho 1971; Rylands *et al.* 1993, 2009; Malukiewicz *et al.* 2014, 2015). That specimen records for *C. penicillata* are much more recent than the records we obtained for any other *Callithrix* species, also suggests an expansion of the range of *C. penicillata* in our study area.

We found no museum specimen records for *C. jacchus* and *C. kuhlii*. Given that *C. jacchus* is a native of Northeast Brazil (Rylands *et al.* 2009), it was not surprising to find historical evidence lacking for the presence of *C. jacchus* in our Minas Gerais mesoregions. *Callithrix kuhlii* is the rarest native *Callithrix* species in Minas Gerais, given that it has a very small range, restricted to the extreme northeast of the state, and as such way outside our area of study (Rylands *et al.* 2009).

Distribution of native and introduced marmosets in the five mesoregions

Historically, large rivers and mountain chains in Brazil have acted as physical barriers that impeded contact between different populations and facilitated allopatric speciation (Rylands *et al.* 1996). Although natural secondary contact occurs between some *Callithrix* species, rivers and mountains in the past have helped isolate species from one another (Rylands *et al.* 1996). This scenario certainly applies to Minas Gerais and *Callithrix* distributions as can be seen in the altimetric map of Minas Gerais in Figure 2A.

Our survey results indicate the presence of numerous introduced *C. penicillata* populations in the southern and eastern parts of Minas Gerais and the Zona da Mata, where they are in close proximity to other introduced marmoset species, particularly *C. geoffroyi* and *C. jacchus*. Introduced marmosets are also present in the Metropolitana de Belo Horizonte, the natural home of *C. penicillata* (Rylands *et al.* 2009).

Callithrix penicillata is native to the Cerrado (Rylands et al. 1993), having adaptations that allow it to colonize open and more seasonal environments (Rylands and Faria 1993), and historically its range in Minas Gerais may have been entirely allopatric to other marmoset species (Rylands et al. 2009). Our survey results and the museum specimen records indicate clearly that the range of C. penicillata is expanding into areas originally covered by the Atlantic Forest in eastern and southern Minas Gerais. This expansion is related to deforestation (Rylands et al. 1993, 1996, 2009). Moreira et al. (2008) also argued that the expansion of the geographical distribution of mammals adapted to open environments, such as the Cerrado, can be attributed, at least in part to deforestation in the Atlantic Forest.

Another factor favoring the expansion of the range of C. penicillata, as well as C. jacchus, is illegal trafficking (RENCTAS 2001; Levacov et al. 2011; Freitas et al. 2015). Marmosets are commonly kept in captivity as pets and often abandoned in areas outside their range (Mendes 1997; Morais Júnior 2010). Just like introduced C. penicillata, introduced C. jacchus are found throughout the mesoregions, and associated microregions of Zona da Mata and Sul/Sudoeste de Minas, as well as the Campo das Vertentes. The southern and central portions of Minas Gerais may be particularly vulnerable to this problem due to the high human population densities there and the presence of regional highways used for the illegal transport of wild animals (RENCTAS 2001). Hybridization occurs wherever C. jacchus and C. penicillata meet (Alonso et al. 1987; Coimbra-Filho et al. 1993; Pereira et al. 1995; Pereira 2012; Ruiz-Miranda et al. 2006; Malukiewicz et al. 2014, 2015).

Callithrix flaviceps and C. aurita are more cryptic than the other species of the genus (Muskin 1983; Ferrari 1988; Rylands et al. 1997; J. Malukiewicz pers. obs.). The former is listed as Endangered, and the latter as Vulnerable on the IUCN Red List of Threatened Species (IUCN 2017). Fragmentation and habitat destruction have severely reduced their numbers (Rylands et al. 1997; IUCN 2017). The little data we were able to obtain indicated that C. aurita occurs in the mesoregion of Sul/Sudoeste de Minas, that both species are found in the Zona da Mata, and that both are in close proximity to introduced marmosets. They were not reported for localities any considerable distance outside of their supposed natural ranges. Callithrix aurita is naturally scarce and is not commonly kept as a pet (Brandão and Develey 1998), and the same is probably true for C. flaviceps. Trafficking in these two species is minimal. Their rarity means that the geographic distributions of C. aurita and C. flaviceps may be underestimated. Thus, future research on geographical distributions of these two species should focus on their known historical distributions and surrounding regions to determine if they can be found around those regions, but also to determine areas of recent declines.

Callithrix geoffroyi is endemic to the Atlantic Forest in southeastern Brazil in the lower-montane and coastal lowland forest (Rylands et al. 1993, 1996, 2009). We obtained confident reports of the presence of this species in the Cerrado in areas that are naturally inhabited by C. penicillata. They also occur in forest patches in an area of Caatinga in the central valley of the Rio Jequitinhonha, south of the river, in northeastern Minas Gerais (Rylands et al. 1988). Hybrids of these two species have been noted in the municipalities of Antonio Dias (Vale do Rio Doce) (Passamani et al. 1997) and Viçosa (Zona da Mata) (Fuzessy et al. 2014). Reports exist of the illegal capture of C. geoffroyi in different regions of Minas Gerais as part of the illegal pet trade (Melo 2004), which favors the introduction of this species outside of its natural area of occurrence. Ávila-Pires (1969) and Coimbra-Filho (1971) also mentioned the possible introduction of C.

geoffroyi in the region of Lagoa Santa, Metropolitana de Belo Horizonte. A genetic survey comparing *C. geoffroyi* populations endemic to the Atlantic Forest and the Cerrado populations using a marker such as mitochondrial D-loop or whole mitochondrial genome (e.g., Malukiewicz *et al.* 2014, 2017) may shed light as to the origin of the latter populations.

Interactions with humans and urban settings

Introduced and native marmoset populations are frequently found in urban areas, in parks, public squares, and forest fragments (Goulart *et al.* 2010; Leite *et al.* 2011; Rodrigues and Martinez 2014). This pattern is confirmed in our data. Our questionnaire indicated that marmosets easily become habituated to human presence. Many survey respondents noted that people feed wild marmosets, and that marmosets develop a certain dependence on humans for food (Paula *et al.* 2005; Rodrigues and Martinez 2014; I. Silva and F. Silva pers. obs.). This provisioning (fruit, bread, sweets, and leftovers) disrupts their natural foraging patterns (Paula *et al.* 2005; Rodrigues and Martinez 2014) but means that they can thrive in often severely degraded forest patches and scrub (Pontes *et al.* 2007; Rodrigues and Martinez 2014; I. Silva and F. Silva pers. obs.).

The negative aspects of this close proximity to humans, include their exposure to parasites carried by humans, domestic animals or urban pests (Tavela *et al.* 2013; Mafra *et al.* 2015), and hybrid marmosets may be more susceptible to parasite infestations (Santos Sales *et al.* 2010). They are also hunted for pets, attacked by domesticated animals, and suffer from road kill and, when using power lines, electrocution (Paula *et al.* 2005; Rodrigues and Martinez 2014; I. Silva and F. Silva pers. obs.).

Conclusion

Overall, our results show that the geographical distributions of *Callithrix* in the area we covered are a hodgepodge of natural and allochthonous populations; a pattern suggesting that there are strong, imminent and future concerns regarding the conservation of these marmosets due to biological invasions and hybridization.

Our data importantly show to what extent the invasion of exotic *Callithrix* has proceeded in five mesoregions of Minas Gerais. This process is especially alarming for cases where invasive marmosets are found in contact with the endangered species, *C. aurita* and *C. flaviceps* occurring there naturally. The presence of invasive species creates the need for immediate studies regarding ecological dynamics between native and introduced species, such as competition for territory and food, sexual competition and hybridization, as well as adaptive species differences within the current geographic scenario. These invasive populations need to be managed, not only in our study area in Minas Gerais, but also in other regions of Brazil.

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Supplementary Information 1.

The questionnaire of the e-mail-based survey from September 2012 to February 2013.

Supplementary Information. Table SI 1.

Species found in MG municipalities by each method of data collection.

http://www.primate-sg.org/storage/pdf/PC32_SI_Silva_et_al_Primate_Conserv_32.pdf

Appendix

Photos of Callithrix specimens from different regions from Minas Gerais state that were received from questionnaire respondents and from online research. (A) Callithrix aurita found in Tombos, microregion of Muriaé, mesoregion of Zona da Mata (Photo by Daniel Ferraz); (B) Callithrix flaviceps found in Manhuaçu, microregion of Manhuaçu, mesoregion of Zona da Mata (Photo by Ana Paula Pereira); (C) Callithrix jacchus found in Viçosa, microregion of Viçosa, mesoregion of Zona da Mata (Photo by Orlando Vital); (D) Callithrix aurita found in Viçosa, microregion of Viçosa, mesoregion of Zona da Mata (Photo by Orlando Vital); (E) Callithrix penicillata found in Lamim, microregion of Viçosa, mesorregion of Zona da Mata (Photo by Vanner Boere); (F) Callithrix penicillata found in Leopoldina, microregion of Cataguases, mesoregion of Zona da Mata (Photo by Vanner Boere); (G) Callithrix jacchus and Callithrix penicillata found in Lavras, microregion of Lavras, mesoregion of Campos das Vertentes (Photo by Matusalém Miguel); (H) Callithrix penicillata found in Barbacena, microregion of Barbacena, mesoregion of Campo das Vertentes (Photo by João Marcos Junqueira Ribeiro); (I) Callithrix penicillata found in Ibertioga, microregion of Barbacena, mesoregion of Campo das Vertentes (Photo by Helberth Peixoto); (J) Callithrix geoffrovi found in Acucena, microregion of Ipatinga, mesoregion of Vale do Rio Doce (Photo by Helberth Peixoto); (K) Callithrix flaviceps found in Caratinga, microregion of Caratinga, mesoregion of Vale do Rio Doce (Photo by Douglas Eduardo Rocha); (L) Callithrix geoffroyi found in Cantagalo, microregion of Peçanha, mesoregion of Vale do Rio Doce (Photo by Ralph de Pinho Carvalho); (M) Callithrix aurita found in Santa Rita de Caldas, microregion Poços de Caldas, mesoregion of Sul/Sudoeste de Minas (Photo by Helberth Peixoto); (N) Callithrix penicillata found in Arceburgo, microregion of São Sebastião do Paraíso, mesoregion of Sul/Sudoeste de Minas (Photo by Ademir Carosia); (O) Callithrix aurita found in Pedralva, microregion of Santa Rita do Sapucaí, mesoregion of Sul/Sudoeste de Minas (Photo by João Paulo Braga); (P) Callithrix penicillata found in Ibirité, microregion of Belo Horizonte, mesoregion of Metropolitana de Belo Horizonte (Photo by Adriana Milagres); (Q) Callithrix penicillata found in Ouro Branco, microregion of Conselheiro Lafaiete, mesoregion of Metropolitana de Belo Horizonte (Photo by Flávia Queiroz Weysfield); (R) Callithrix penicillata found in Belo Horizonte, microregion of Belo Horizonte, mesoregion of Metropolitana de Belo Horizonte (Photo by Vinicius Herold Dornelas e Silva).



