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ORIGINAL ARTICLE

Electrocutions in free-living black-tufted marmosets (*Callithrix penicillata*) in anthropogenic environments in the Federal District and surrounding areas, Brazil

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Abstract
Shrinking natural habitats exposes some non-human primates to the risk of accidents associated with electrical transmission lines. We examined dead marmosets (*Callithrix penicillata*) collected in the region from January 2015 to April 2018 to determine the animals' cause of death and for electrocuted animals we examined the locations the animals had died as well as the configuration of the power lines at these sites. We also recorded the sex of the animal, the body region affected, and characteristics of the injuries. We diagnosed electrocutions in 11% ($n=34$) of the marmosets studied. Most of the affected animals were male ($n=22$) with single or double sites of injury on the limbs. Animals were injured in urban ($n=26$) and peri-urban ($n=8$) areas on lower-voltage alternate current lines, and we detected no seasonality or hotspots of electrocution. Our findings suggest that movement along transmission lines composed of bundled conductors is a major factor in electrocutions of marmosets in the Federal District and surrounding areas. The planning of electrical power grid infrastructure should consider arboreal primates to prevent electrocutions.

Keywords Non-human primates · Electrical injuries · Conservation · Lower voltage

Introduction

Extensive loss of habitats due to urbanization, agriculture, and animal husbandry forces some primates to survive in rural, semi-urban, and urban areas (Corrêa et al. 2018; Estrada et al. 2012; Kumar and Kumar 2015). Such anthropogenic environments pose risks to non-human primates, including accidental and non-accidental trauma, predation by domestic dogs and cats, and electrocution (Corrêa et al. 2018; Kumar and Kumar 2015; Sinha and Vijayakrishnan 2017). Electrocutions and non-fatal electric shock injuries occur when wild animals encounter exposed electric wires and are usually caused by the alternating low-voltage currents of less than 1000 V commonly used in homes and small industries (Arnoldo and Purdue 2009; Sheikhazadi et al. 2010; Viner 2018). Electric shock injuries present highly variable morphological characteristics due to variation in tissue resistance to electrical energy and the amount of current. Electrocution injuries may vary considerably: some animals may have no visible marks, some may have first- to fourth-degree burns, while some may have completely carbonized tissues (Schulze et al. 2016).

Electric shock injuries occur in domestic animals (Boeve et al. 2004; Knox et al. 2014; Novales et al. 1998; Ros et al. 2015), and wild bird populations, and for some species electrocutions may represent a considerable threat to its survival (Dwyer et al. 2014; Lehman et al. 2007). In primates, such injuries generally occur when primates use the electrical power grid to travel across anthropogenic matrices

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(Chapman et al. 2016; Kumar and Kumar 2015; Lindshield 2016; Rodrigues and Martinez 2014).

Marmosets (genus *Callithrix*) are a diverse group of platyrrhine primates with 13-15 distinct taxa, many of which are considered endangered (Tagliaro et al. 1997). The black-tufted marmoset, *Callithrix penicillata,* is endemic to the savanna biome of Brazil (Miranda and Faria 2001; Vilela and Faria 2004). Black-tufted marmosets are one of the more abundant *Callithrix* species and adapt well to urbanized environments (Duarte et al. 2011; Sinha and Vijayakrishnan 2017). As a result, they have a high risk of contact with electric transmission lines and associated injuries.

The aim of this study is to determine the influence of seasonality, geographical distribution, and local standards of electrical transmission lines on electrocutions in black-tufted marmosets in the Federal District and surrounding areas in Brazil. We also describe morphological characteristics of electric shock injuries in these animals.

Methods

Study site and sample collection

The Federal District is the smallest unit of the Brazilian Federation, with an area of 5799.99 km^2, located in Central Brazil and surrounded by the states of Goiás and Minas Gerais. The Federal District and its surrounding comprise a large metropolitan area with close to 3 million human inhabitants and have one of the highest rates of urbanization in Brazil (Brandão 2015; IBGE 2010). Although extensively urbanized, the metropolitan area has the highest rate of reforestation of public roads (36.9%) in Brazil and is interspersed with fragments of native vegetation and artificially forested patches (Brandão 2015). Natural areas suffer great anthropic pressure due to rapid and unplanned population growth, real estate speculation, and occupation of natural areas by agriculture and livestock.

We examined records of electrocutions in marmosets at the Regional Reference Laboratory of the Ministry of Health for the Diagnosis of Yellow Fever in non-human primates—University of Brasilia, Brasília, Federal District, Brazil, from January 2015 to April 2018. The Directorate of Environmental Health Surveillance—Secretary of Health collected all dead non-human primates found by people in the Federal District and surrounding areas (Goiás State) in accordance with the National Epizootic Surveillance Program for the Control of Yellow Fever of the Brazilian Ministry of Health. Public campaigns encourage the population to collaborate with the Yellow Fever surveillance and control program and to report the location of dead animals to the Directorate of Environmental Health Surveillance. We recorded the location of each marmoset using GPS and georeferenced it using QGIS 2.18.0™ software.

Sample processing

We necropsied and collected samples of organs and tissues from all marmosets, and fixed them in 10% buffered formalin, processed and embedded them in paraffin, and stained histological sections with hematoxylin and eosin (H&E). Skin samples with current marks from the palm or plantar surfaces were stained with Pearls' Prussian blue to detect metallization lines formed by microscopic particles of metal from electric wiring transferred to the skin (Bellini et al. 2016; Schulze et al. 2016). We recorded the morphological characteristics, location, distribution, and frequency of external and internal injuries. The diagnosis of electrocution was based on the animal's location at the time of death (e.g., under electrical lines), witness reports, skin lesion characteristics, electrical burns, and histological changes (Merck and Miller 2013; Schulze et al. 2016; Viner 2018).

GPS coordinates were used to investigate the distribution of electrocutions across urban and peri-urban areas. Standards of the electrical transmission lines (voltage, disposition, type, and distance between wires) at locations of electrocutions were determined on the technical standards used by Energy Company of Brasília (Ferreira et al. 2011) and local inspection. We recorded the frequency of marmoset electrocutions by sex, age group (adult or young based on external morphological characteristics: Decanini and Macedo 2008), year, month, season (spring, summer, autumn, winter) and precipitation period (rainy season from October to April vs. dry season from May to September: INMET 2018).

Data analysis

We used Fisher's exact test to compare electrocutions with sex (male or female), age range (adult or young), geographical location of electrocutions (urban or peri-urban areas, and Federal District or surrounding areas) and precipitation period (moisture on electrical transmission lines could favor electrocutions) (Cooper 1995; Koumbourlis 2002). We used Chi-square tests to compare frequencies of external electrical lesions on body regions (upper limbs, lower limbs, torso, and tail), the number of electric shock injuries detected in each animal (single, double, triple, or more), and the geographical location at which the electrocution occurred (to detect hotspots) (Katsis et al. 2018), years, months, and seasons of the year (may interfere with behavioral and ecological aspects of the marmosets) (Vilela and Faria 2004). We calculated the incidence (%) of electrocutions in relation to other causes of death of marmosets in the study period. We categorized the geographical location of electrocutions (urban or peri-urban areas, and Federal District or surrounding areas) according to data from the Planning Company of the Federal District (Brandão 2015). All data and samples from this study are available in the archives of Regional Reference Laboratory of the Brazilian Ministry of Health for the Diagnosis of Yellow Fever in Non-Human Primates—University of Brasilia, Brasília, Federal District, Brazil. The monthly rainfall data are available from the National Institute of Meteorology (INMET 2018).

Results

We reviewed 355 necropsy records of non-human primates, 306 of which involved black-tufted marmosets. Electrocutions were diagnosed in 34 marmosets (incidence of 11%). Electrocutions were randomly distributed across the months of the year (January, $n=4$; February, $n=2$; March, $n=3$; April, $n=4$; May, $n=2$; June, $n=5$; July, $n=1$; August, $n=1$; September, $n=3$; October, $n=5$; November, $n=2$; December, $n=2$; $P=0.816$) without a seasonal pattern (dry, $n=20$, wet season, $n=14$, $P=0.225$; autumn, $n=9$; winter, $n=7$; spring, $n=10$; summer, $n=8$; $P=0.853$). Among the 4 years of surveillance, electric shock injuries in marmosets were more frequent in 2016 ($n=12/42$; $P<0.001$) (Fig. 1).

Deaths of marmosets by electrocution occurred predominantly in urban areas ($n=26$, $P<0.001$) and in Federal District ($n=31$, $P<0.001$) with fewer cases ($n=4$) in surrounding areas in Goiás State (Fig. 2). Of the 34 cases of electrocution, four were witnessed. On one occasion, four marmosets (one adult male, one adult female, two juveniles) were electrocuted simultaneously. Cases were located throughout the region, with no hotspots of electrocution ($P=1.0$) (Fig. 2).

According to the technical standards for power transmission lines used by the Energy Company of Brasília, and our observations at electrocution locations, all electric shock injuries occurred on lower-voltage (< 1000 V) alternate current transmission lines. Transmission lines in the Federal District and surrounding areas mainly supply households (86.6%) and are composed of bundled conductors made up of four sub-conductors or vertical parallel non-coated conductors spaced with 20 cm between the wires.

Adults were electrocuted more than juveniles ($n=31$, $P<0.001$) and males were electrocuted more than females ($n=22$, $P=0.028$). Electrocuted animals exhibited high frequencies of both single ($n=16$) and double site injuries ($n=13$) more than triple injuries ($n=6$, $P=0.032$). External electric shock injuries were more frequent on the upper ($n=14$) and lower limbs ($n=10$) and torso ($n=10$) than on the tail ($n=2$, $P<0.001$) (Fig. 3). Most electrical lesions were on the extremities of the limbs, especially on the palm and plantar surfaces (Fig. 4a). We observed fractures in the spinal column close to electrocution marks in one marmoset, and on the limbs in four marmosets, affecting the radius, ulna, tibia, fibula, or phalanges (Fig. 4b). Amputation of the upper right limb with complete carbonization of the surrounding tissues was also observed in one marmoset.

Direct electrothermal injuries on the skin (Fig. 5a) and adjacent tissues, characterized by mild to severe wiring burns (Fig. 5b), were the hallmarks of electrocution in the marmosets. External current marks ranged from slight superficial ashy discoloration to deep ulcers, with carbonized borders and exposure of the underlying tissues including subcutaneous, muscles and bones (first- to fourth-degree

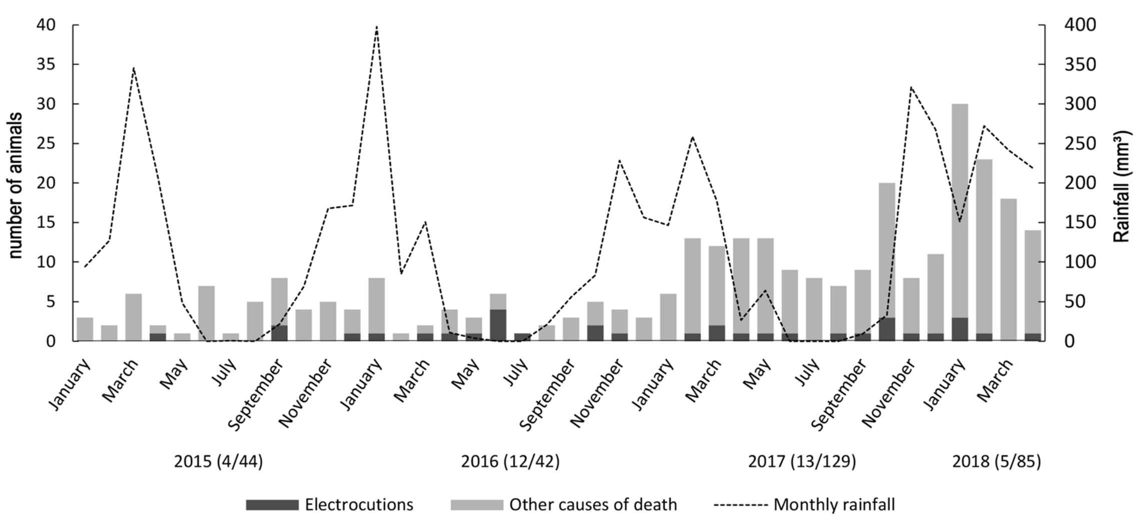

Fig. 1 Occurrence of electrocutions in black-tufted marmosets and monthly rainfall from January 2015 to April 2018 in Federal District and surroundings, Brazil

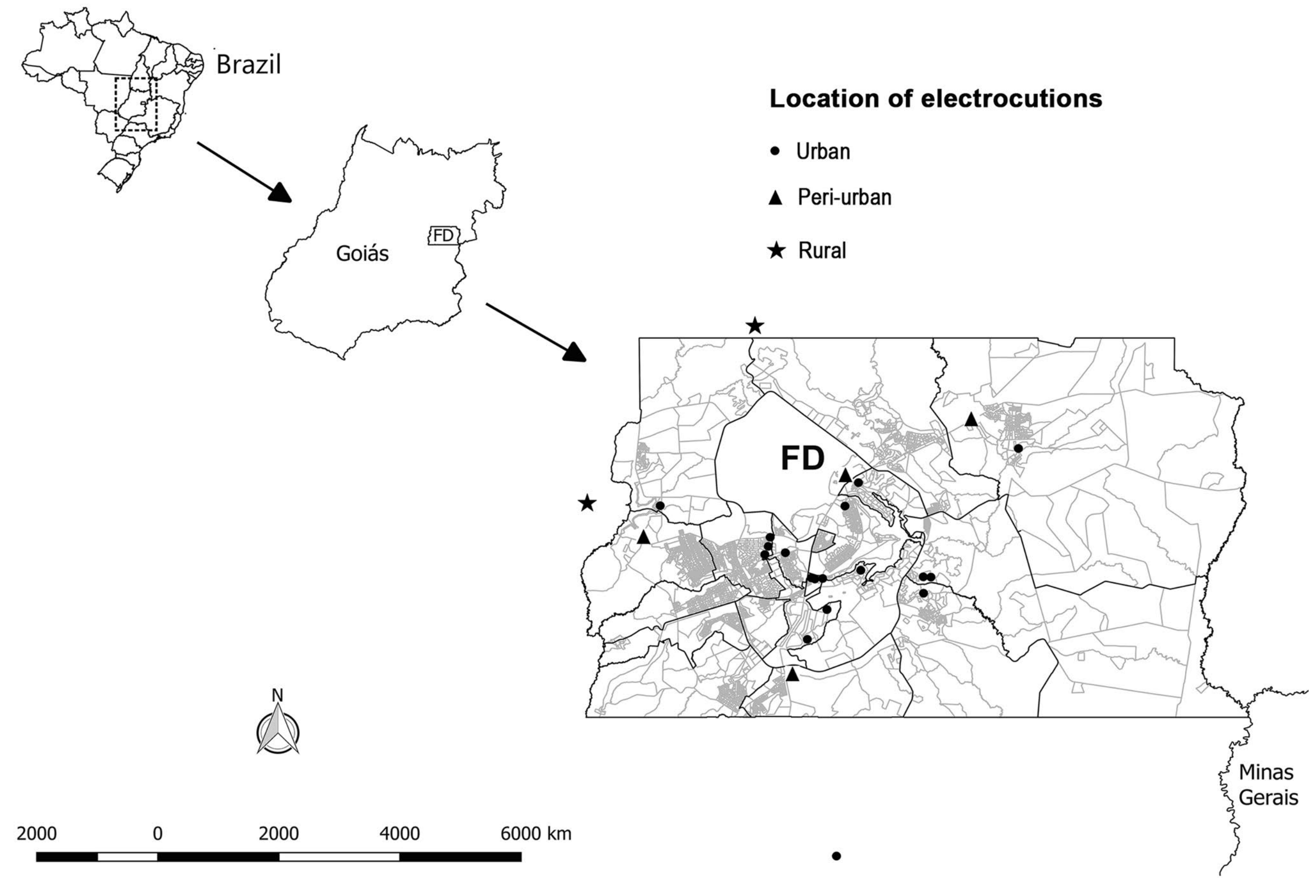

Fig. 2 Location of electrocutions of black-tufted marmosets from January 2015 to April 2018 in Federal District (FD) and surrounding areas (Goiás State), Brazil

Fig. 3 Number of electrocutions in black-tufted marmosets by sex, age, number of electric shock injury sites, and location of electric shock injuries from January 2015 to April 2018 in Federal District and surrounding areas (Goiás State), Brazil

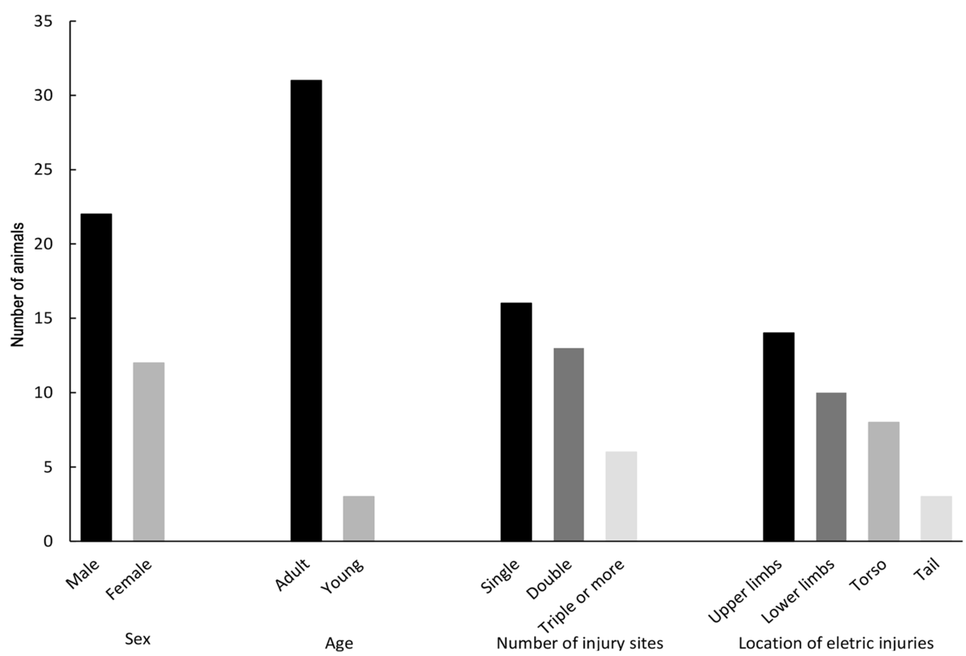

Fig. 4 Electrocuted black-tufted marmosets. **a** Lower limb with electrothermal injury on the plantar surface and ulcer formation on the skin. **b** Upper limb with severe electric shock injury with exposure of the underlying tissues and fracture of phalanges

Fig. 5 Electrocuted black-tufted marmosets. **a** Lower limb with carbonization of tissues and deep ulcer on the skin. **b** Torso with dark reddish parallel electrical wiring marks in the subcutaneous and muscle tissues

burns). Singed hairs at electric shock injury sites with haired skin were observed in 14 marmosets. In some cases, the exact distance between the points of contact with the electrical wiring based on external current marks were detected (Fig. 6). Congestion and hemorrhage of the lungs were the main lesions in internal organs, detected in 50% of the electrocuted animals.

Histological analysis of current marks on the skin in the palm or plantar surfaces of seven animals revealed intraepidermal separation (blister formation), discontinuity and coagulation necrosis of the epidermis and dermis, and hyalinization of collagen fibers (Fig. 7). Metallization lines on the epidermal surface were observed in three of the skin samples ($n = 7$) stained with Pearls' Prussian blue (Fig. 7).

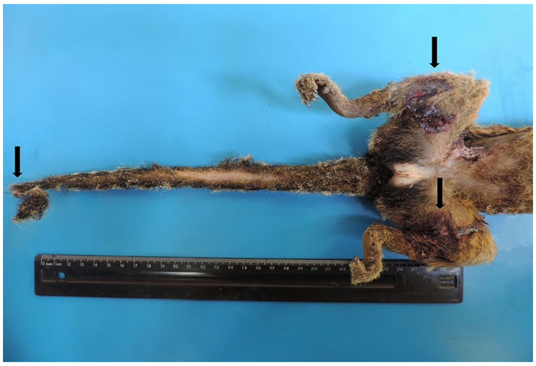

Fig. 6 Tail, lower limbs, and inguinal region of a black-tufted marmoset showing external contact points with wiring burns (*arrows*) and singed hairs

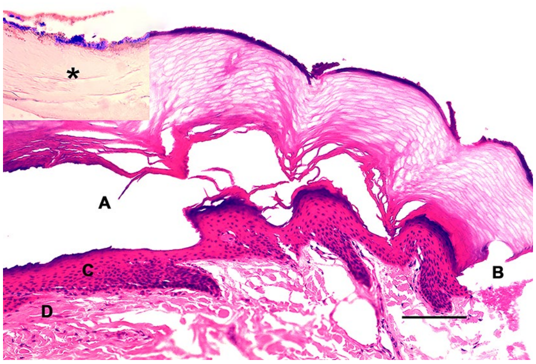

Fig. 7 Lower limb, plantar surface of a black-tufted marmoset with intraepidermal blister formation (**a**, intraepidermal separation), discontinuity (**b**) and coagulation necrosis of the epidermis (**c**), and hyalinization of collagen fibers (**d**) (H&E, bar = 100 µm). *Close view: lower limb, plantar surface of a black-tufted marmoset with metallization line on the epidermal surface stained with Pearls' Prussian blue

Discussion

We found that electrocutions are an important cause of death in free-living marmosets in the Federal District and surrounding areas, suggesting that the anthropogenic matrices in these areas are dangerous for marmosets. Other threats of the urbanized environment for marmosets have also to be considered as variable traumatic injuries such as car accidents, stoning, falling during wire crossing, predation by domestic animals and others (Rodrigues and Martinez 2014). In our study, the high number of animals necropsied with other causes of death than electrocution strengthens these observations. The dispersed occurrence of electrocuted animals shows that the region supports marmoset populations over a large urban area. A higher density of electrified cables in areas with higher rates of urbanization in the Federal District and surrounding areas possibly justify the electrocution of more marmosets in these locations. The abundant vegetation and food supply in urban and peri-urban areas may attract marmosets and facilitate their movement in highly urbanized areas. It is also important to consider differences between the scavenging activity from domestic and wildlife animals in urban and peri-urban areas (Antworth et al. 2005) that could cause variations in the number of dead marmosets collected.

Other arboreal primates such as brown howler monkeys (*Alouatta* sp.) also move between food patches using matrix elements such as trees, roofs and power lines, increasing the risk of electrocutions (Corrêa et al. 2018). Electrical power grid and infrastructure also pose a serious electrocution danger for rhesus macaques in India (Kumar and Kumar 2015).

The extensive local lower-voltage electrical power grid composed by non-coated conductors spaced 20 cm apart expose small primates such as marmosets to the risk of electrocution during the movement between areas in the Federal District. The standards for electrical power grid infrastructure used by the Energy Company of Brasília are regulated by the Brazilian's National Electric Energy Agency, and was not planned taking into account the movement of non-human primates across anthropogenic matrices and coexistence with wildlife.

We could not determine why marmosets move along electric transmission lines, as only four cases were witnessed, and the behavior of marmosets in the anthropogenic matrix of Federal District has not been studied. However, foraging for fruits, insects, and exudates (Vilela and Faria 2004), and the selection of home ranges to avoid noisy areas (Duarte et al. 2011), are likely to influence their movement and the risk of electrocutions.

Most electrocuted animals were adult males, which may be at greater risk of electrocution due to age- and sex-related behavioral differences. Groups of marmosets are composed of 3–15 individuals with one breeding dominant male and female (Schiel and Souto 2017). The male bias in electrocution risk of marmosets could be attributed to factors such as differences between sexes in their reach for foraging (Michels 1998; Miranda and Faria 2001) and dispersal (Ferrari 2009). Juvenile callitrichids have propensities to be overtly responsive to the ambient and objects (Box and Smith 1995; Kendal et al. 2005), which may expose them to greater risk than adults. However, cooperative breeding may protect infants from accidental injuries (Schiel and Souto 2017). The smaller size of young animals could minimize the incidence of electric shock injuries by preventing simultaneous contact with two or more wires.

Wet skin and moisture on electrical wiring during rainy seasons reduce electrical resistance, favoring electrocutions (Cooper 1995; Koumbourlis 2002). However, despite the marked dry and rainy periods in the Federal District and the surrounding areas, we found no seasonal pattern of electrocutions of marmosets. These findings contrast with electric shock injuries in free-range young male rhesus macaques, which occur predominantly during the rainy period (Kumar and Kumar 2015).

Electrocution frequency varied across the 4 years of the study. The increase in epidemiological surveillance and the collection of dead non-human primates associated with the severe outbreak of yellow fever in Brazil in 2017 and 2018 explain the increase in necropsies recorded and electrocutions detected in these years. However, the limited epidemiological data do not allow us to explain the highest frequency of electrocuted animals in 2016.

The morphological aspects of external current marks observed in electrocuted marmosets are similar to those reported in domestic and wild animals (Merck and Miller 2013; Schulze et al. 2016; Viner 2018), and humans (Mukherjee et al. 2015). In low-voltage electrocutions, electrical marks may be found around the contact point with power sources (Merck and Miller 2013). However, electrical injuries do not always produce external lesions that allow accurate diagnosis (Arnoldo and Purdue 2009; Schulze et al. 2016). It is possible that we underestimated the frequency of death by electrocution as electrical injuries do not always produce morphological changes.

In addition to the electrocutions we report, it is possible to suppose that a considerable number of marmosets may have suffered non-fatal electrical injuries. For example, electric shock injuries in rhesus monkeys result in 31.5% of mortality (Kumar and Kumar 2015). The small body size of marmosets implies reduced contact areas with electrical wiring, and may allow more energy to act per area of tissue (Fish and Geddes 2009; Hunt et al. 1976; Schulze et al. 2016). The concentrated powerful action of electric energy in a small region of the body may explain the severity of electric shock injuries on marmosets, especially on the limbs.

The few witnessed electrocutions involved contact between the limbs and wiring when the animals traveled on power transmission lines. This suggests that electric shock injuries occurred mainly on the extremity of limbs of marmosets due to contact between the hands and feet and the wiring, as in electrocuted rhesus macaques (Kumar and Kumar 2015). In some cases, marmosets also presented bone fractures and extensive tissue carbonization. This may be because exposure to alternate current (AC) causes strong muscle contraction, which can fracture the long bones and spine (Arnoldo and Purdue 2009; Merck and Miller 2013; Viner 2018). Muscular tetany in forearm flexors on contact with AC wiring increases the duration of the current flow and tissue damage by electrothermal heating (Arnoldo and Purdue 2009; Cooper 1995).

A considerable number of cases presented simultaneous electrical injury in more than one body site, mainly on the limbs. Electrical currents passing through the body can cause death and electrothermal injuries at the contact points between electrical source and skin, and in the inner organs (Fish and Geddes 2009; Schulze et al. 2016). The distances between skin contact points with the electrical wiring in some electrocuted marmosets were similar to the spacing standard between the wires used in the lower-voltage electrical transmission lines in the region. These findings reinforce the danger that electrical distribution lines composed of non-coated conductors spaced 20 cm apart present to small primates such as marmosets because they allow contact with two or more conductors at the same time, triggering electrocution.

Half of the marmosets analyzed showed pulmonary hemorrhage and congestion, similar to effects reported in electrocuted humans (Karger et al. 2002) and pigs (Giles and Simmons 1975). Exposure time, voltage, amperage, resistance of the tissues, size of the contact area, and the path of the current through the body are the main factors involved in the extent and severity of tissue damage (Arnoldo and Purdue 2009; Bier et al. 2005; Koumbourlis 2002; Schulze et al. 2016).

Histological changes in the current marks in the skin samples of marmosets were characterized by separation of the skin layers and coagulation necrosis of the dermal collagen, which are commonly reported in electrocuted animals (Merck and Miller 2013; Schulze et al. 2016; Viner 2018). The transfer of metal particles from conductors to the skin (metallization line) is an important cutaneous change in the diagnosis electrical burns, but may not be observed in all injured animals or humans (Merck and Miller 2013; Bellini et al. 2016; Schulze et al. 2016; Viner 2018).

The diagnosis of electrocution in free-living primates is challenging due to the frequently single and subtle lesions, nonspecific morphological characteristics, and the absence of clinical and epidemiological information. Mapping areas of risk and displacement, and the study of marmoset behavior are essential to understand the impact that electrical low-voltage grids have on marmoset populations. This knowledge can lead to actions to reduce or prevent mortality in free-range marmosets living in anthropogenic environments. In addition, public actions and policies for the standards of the electrical transmission lines should minimize accidents and the death of animals. For example, an increase in the distance between the conductors or the use of safe coated conductors should prevent simultaneous contact with wiring, reducing the risk of electrocutions of marmosets and other primates.

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