Evidence of leptospiral exposure in neotropical primates rescued from illegal trade and a Zoo in Bahia, Brazil

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ABSTRACT. - Almeida D.S, Santos A.C., Silva C.L.R., Oriá A.P., Oliveira A.V.D., Libório F.A., Athanazio D.A. & Pinna M.H. 2016. Evidence of leptospiral exposure in neotropical primates rescued from illegal trade and a Zoo in Bahia, Brazil. Pesquisa Veterinária Brasileira 36(9):864-868. Departamento de Patologia e Medicina Legal, Praça XV de Novembro s/n, Largo do Terreiro de Jesus, Salvador, BA 40025-010, Brazil. E-mail: daa@ufba.br

Few studies have compared the seroprevalence of antileptospiral agglutinins with the demonstration of urinary shedding of leptospira or evidence of active infection in the bloodstreams of non-human primates. The study population consists of 58 animals, including 42 monkeys from the Zoological Park of Salvador (Parque Zoológico Getúlio Vargas), Bahia, Brazil. The study also evaluated 16 primates (Cebus sp.) rescued from illegal trade that were housed in the Wildlife Rehabilitation Center of Salvador (CETAS), Bahia, Brazil. The seroprevalence of antileptospiral antibodies was low (2%) in the animals from the Zoo. A higher rate (31%) was observed among the animals that were rescued from illegal trade in the state of Bahia. Even if all the blood and urine samples were negative for leptospiral DNA fragments, the high frequency of serological evidence of exposure suggests a potential risk of leptospirosis transmission when keeping these animals as pets.

INDEX TERMS: Leptospirosis, Leptospira, Primates.

INTRODUCTION

Leptospirosis is a widespread zoonosis caused by pathogenic leptospira. Humans acquire infection by direct exposure to contaminated urine from mammalian reservoirs or, more commonly, by exposure to contaminated soil or water. The disease occurs in different settings associated with a wide range of reservoirs, such as field mice in paddy fields, farm animals in occupational exposures, urban rodents...
in large urban areas with poor sanitation, and wild animals in water sports and ecotourism related exposures (Bharti et al. 2003). Most human infections are asymptomatic or cause mild febrile illness that is indistinguishable from diseases caused by other infectious agents. However, 5-10% of human infections will become severe, resulting in the Weil's triad of acute renal failure, hemorrhages, and jaundice (with 5-30% fatality) or severe pulmonary hemorrhagic syndrome (SPHS, with ≥ 50% fatality) (WHO 2003a, McBride et al. 2005, Medeiros et al. 2010).

Non-human primates usually develop a self resolving, mild illness in naturally acquired or experimental infections (Minette 1966, Minette & Shaffer 1968, Hambleton et al. 2005, Medeiros et al. 2010). Most human infections are asymptomatic or produce mild febrile illness that is indistinguishable from diarrheal syndromes caused by other infectious agents. However, 5-10% of human infections cause mild febrile illness that is indistinguishable from diarrheal syndromes caused by other infectious agents. However, 5-10% of human infections will become severe, resulting in the Weil's triad of acute renal failure, hemorrhages, and jaundice (with 5-30% fatality) or severe pulmonary hemorrhagic syndrome (SPHS, with ≥ 50% fatality) (WHO 2003a, McBride et al. 2005, Medeiros et al. 2010). There is a significant public health concern associated with illegal wildlife ownership, as wild animals are estimated to be the source of over 70% of all emerging infections (Chomel et al. 2007). Seroprevalence of antileptospiral antibodies in Neotropical primates from zoos in Latin America or from wildlife is variable, partly due to the diverse species studied and the different cutoffs employed (Table 1 and 2) (Baulu et al. 1987, Kessler & Everard 1988, Perolat et al. 1992, Luna-Alvarez et al. 1996, Ibanez-Contreras et al. 2010*).

Table 1. Studies in Latin America on the serologic evidence of exposure to leptospires in non-human primates in captivity

<table>
<thead>
<tr>
<th>Author, year</th>
<th>Site, City, Country</th>
<th>Species</th>
<th>N / n (%)</th>
<th>Predominant serogroup</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baulu et al. 1987</td>
<td>Research Center, Saint Joseph, Barbados</td>
<td><em>Saimiri sciureus</em> (Common Squirrel Monkey)</td>
<td>24/93 (26)</td>
<td>Icterohaemorrhagiae</td>
</tr>
<tr>
<td>Perolat et al. 1992</td>
<td>Instituto Pasteur; Caiena, Guiana Francesa</td>
<td><em>Pongo pygmaeus</em> (Bornean Orangutan)</td>
<td>2/3 (66)</td>
<td>Canicola</td>
</tr>
<tr>
<td>Luna-Alvarez et al. 1996</td>
<td>Chapultepec Zoo, Cidade do México, México</td>
<td><em>Ateles belzebuth</em> (White-fronted Spider Monkey)</td>
<td>2/2 (100)</td>
<td></td>
</tr>
<tr>
<td>Ibanez-Contreras et al. 2010*</td>
<td>Research Center, Cidade do México, México</td>
<td><em>Macaca mulatta</em> (Rhesus Monkey)</td>
<td>38/59 (63)</td>
<td>Panama, Icterohaemorrhagiae, Australis, and Shermani</td>
</tr>
<tr>
<td>Lilenbaum et al. 2002**</td>
<td>Zoo, Rio de Janeiro, Brazil</td>
<td><em>Ateles belzebuth</em> (White-fronted Spider Monkey)</td>
<td>2/2 (100)</td>
<td>Icterohaemorrhagiae</td>
</tr>
<tr>
<td>Corrêa et al. 2004</td>
<td>Zoo, São Paulo, Brazil</td>
<td><em>Callithrix jacchus</em> (Common Marmoset)</td>
<td>1/1 (100)</td>
<td></td>
</tr>
<tr>
<td>Lilenbaum et al. 2005</td>
<td>Primatology center, Rio de Janeiro, Brazil</td>
<td><em>Cebus capucinus</em> (White Faced Capuchin)</td>
<td>7/11 (64)</td>
<td></td>
</tr>
<tr>
<td>Pinna et al. 2012</td>
<td>CETAS, Salvador, Brazil</td>
<td><em>Ateles fusciceps</em> (Black-headed Spider Monkey)</td>
<td>5/19 (33)</td>
<td>Icterohamorhagiae</td>
</tr>
<tr>
<td>Ullman et al. 2012</td>
<td>Zoo, Sorocab, Brazil</td>
<td><em>Callithrix jaccus</em> (Common Marmoset)</td>
<td>16/28 (57)</td>
<td>Icterohaemorrhagiae</td>
</tr>
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</table>

Note: All studies used the cutoff of 1:100 MAT titers except for * and **, which used 1:20 and 1:200 cutoffs, respectively.
The population included 42 monkeys from the Zoological Park of Salvador (Parque Zoobotânico Getúlio Vargas), Bahia, Brazil. These animals belonged to the following species: Cebus xanthosternos (n=23), Cebus flavus (n=11), Alouatta caraya (n=3), Aotus sp. (n=4), and Saimiri sciureus (n=1). The study also evaluated 16 primates rescued from illegal trade that were housed in the wildlife rehabilitation center of Salvador (Centro de Triagem de Animais Silvestres Chico Mendes, CETAS), Bahia, Brazil. All these animals were identified as Cebus sp., as many of them are hybrids.

Microscopic agglutination tests (MAT) were performed, according to recommended protocols (Faine et al. 1999, WHO 2003), and included twenty-three WHO reference strains and a local isolate (serovar Copenhageni strain L1130) (Ko et al. 1999, Nascimento et al. 2004). Titters ≥1:100 were considered positive. Polymerase chain reaction for the detection of the lipL32 gene was performed as previously described (Rojas et al. 2010, Chagas-Junior et al. 2012).

The research protocols were approved by the Research Ethics Committee of the Faculdade Franca - SP (025/2009-A). In addition, they were in accordance with guidelines System Authorization and Information on Biodiversity the Ministry of Environment of Brazil (number 20831-1).

RESULTS AND DISCUSSION

In the present study, only one of 42 primates (2%) from the Zoological Park of Salvador had positive serum samples, according to the MAT. This animal was an adult female Alouatta caraya (black howler) that had a positive serum sample of 1:100 with mixed reactions for the Bratislava and Icterohaemorrhagiae serogroups. This animal was housed in a cage with two other adults of the same species. The positive animal and one negative black howler were born in the Zoological Park, while the other negative adult came from illegal trade (CETAS). Urine and blood samples were negative for all 42 monkeys evaluated at the Zoological Park.

Such low seroprevalence was not expected. Salvador is the third most populated city in Brazil, with an estimated 2.6 million inhabitants, and 60% of those inhabitants live in slum communities (Riley et al. 2007). It is a large urban center with poor sanitation, and incidence of severe leptospirosis cases peak during rainy seasons and in association with floods (Ko et al. 1999, Costa et al. 2001, Riley et al. 2007). Reports from the Zoological personnel indicated that there is a high population of rodents in the park, and rodents are frequently observed to be in contact with the animals in their cages. In some surveys from Latin American zoos, the seroprevalence of antileptospiral antigens is highly variable among different species (Table 1), and higher rates were attributed to the contact of primates with urban rodents. Additionally, there is a high prevalence of Icterohaemorrhagiae as the predicted infecting serogroup in some of these studies (as implied by MAT highest titers), and this suggests that rodents are the source of non-primate infection because these serovars are known to be selectively carried by urban rodents such as the Brown (Rattus norvegicus) and Black rats (R. rattus) (Bharti et al. 2003). In a survey of captured rats in Salvador, 80% of a total 142 animals had positive cultures for leptospires from kidney or urine samples, and all 59 serotypes isolated by monoclonal antibodies were characterized as serovar Copenhageni (serogroup Icterohaemorrhagiae) (de Faria et al. 2008), which is the main cause of severe leptospirosis in Salvador and in other large urban Brazilian centers (Ko et al. 1999, Pereira et al. 2000). It is not possible to infer that the single Black Howler with a positive MAT acquired the infection from exposure to rats because it had a mixed reaction with the Bratislava serogroup, which has no known selectivity for rodents.

In summary, the results from the Zoological Park suggest that the Neotropical primates’ contact with rodents was not associated with an increased seroprevalence of antileptospiral antibodies. These species may be intrinsically more resistant than others in terms of acquiring leptospiral infection, or conversely, these animals may have had previous exposure in the wild that is associated with a progressive decrease in antileptospiral antibodies during the period of captivity.

In 2009, a previous survey performed in the wildlife rehabilitation center of Salvador (CETAS) found 57% (25/44) seropositivity by MAT. In that study, marmosets were mainly affected, and positive samples were distributed as follows: Callithrix jacchus (common marmoset, 16/28: 57%), Callithrix penicillata (Black-tufted marmoset, 4/8: 50%), and Cebus sp. (capuchin monkeys, 5/8: 63%) (Pinna et al. 2012). In this study we found 5/16 (31%) positive serum samples by MAT from Cebus sp.

Table 2. Studies in Latin America on the serologic evidence of exposure to leptospires in non-human primates captured from the wild

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<tr>
<td>Souza Júnior et al. 2002</td>
<td>Lajeado/ Ipueras, Brazil</td>
<td>Alouatta caraya (Black howler)</td>
<td>46/82 (16)</td>
<td>Pomona</td>
</tr>
<tr>
<td>Cebus sp. (Capuchin)</td>
<td>2/286 (2)</td>
<td>Panama</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baulu et al. 1987</td>
<td>Research Center, Barbados, Caribe</td>
<td>Cercocephalus aethiops sabeaus (Vervet Monkey)</td>
<td>150/501 (30)</td>
<td>Ballum</td>
</tr>
<tr>
<td>Kessler &amp; Everard 1988</td>
<td>Caio Santiago, Porto Rico</td>
<td>Macaca mulatta (Rhesus Macaque)</td>
<td>5/169 (3)</td>
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Thus, the high rate of serological evidence of Leptospira exposure seems to be a consistent finding among animals from the wild that are rescued from illegal trade in the state of Bahia. It is important to note, however, that the predicted infecting serogroup inferred by MAT highest titers was Icterohaemorrhagiae in 84% of the cases in a former study. In this study, the five positive samples were distributed in the following predicted serogroups: Ballum (1:100), Semaranga (1:200), Grippotyphosa (1:100), Cy

The study also evaluated the possible association of serological evidence of exposure with active infection and renal carriage using PCR detection of leptospiral DNA in blood and urine samples, respectively. None of the evaluated samples yielded positive results. Further studies with larger sample groups of animals rescued from illegal trade are warranted to estimate the risks of infection from intimate contact with exotic animals, including Neotropical primates, when kept as pets.

CONCLUSIONS

The seroprevalence of antileptospiral antibodies was low (2%) in the Zoological Park of Salvador, Brazil, despite the high frequency of rodents in the area and the endemi

d of human leptospirosis in Salvador.

A higher rate (31%) was observed among the animals rescued from illegal trade in the state of Bahia.

Serological evidence of exposure does not predict an active infection or the renal carrier state in non-human pri

mates. Even if all the blood and urine samples were negative for leptospiral DNA fragments, the high frequency of serological evidence of exposure suggests a potential risk of leptospirosis transmission when keeping these animals as pets.

Acknowledgements- The authors are grateful to Victor Pereira Curvelo for technical assistance on animal capture and confinement during the survey performed in the Zoological Park of Salvador, Brazil.

REFERENCES


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