DOI: 10.7589/JWD-D-20-00044

Journal of Wildlife Diseases, 57(3), 2021, pp. 561–568

EXPOSURE OF WILD BOAR (*SUS SCROFA*) TO THE COMMON VAMPIRE BAT AND LACK OF IMMUNE PROTECTION TO RABIES VIRUS IN BRAZILIAN HUNTERS

Pedro Irineu Teider-Junior,[1] Laís Giuliani Felipetto,[1] Louise Bach Kmetiuk,[2] Fernanda Pistori Machado,[1] Luciana Botelho Chaves,[3] Rene dos Santos Cunha Neto,[3] Thaila Francini Corona,[4] Camila Marinelli Martins,[5,6] Renato van Wilpe Bach,[7] Ivan Roque de Barros-Filho,[1] Andrea Pires dos Santos,[8] and Alexander Welker Biondo[1,8,9]

[1] Graduate College of Veterinary Science, Federal University of Paraná (UFPR), Funcionários Street, 1540, Curitiba, Paraná, PR 80035-050, Brazil
[2] Graduate College of Cellular and Molecular Biology, Federal University of Paraná (UFPR), Coronel Francisco Heráclito dos Santos Avenue, 100, Curitiba, PR 81531-970, Brazil
[3] Pasteur Institute, Paulista Avenue, 393, São Paulo, SP 01311-000, Brazil
[4] Paraná State Reference Laboratory, Sebastiana Santana Fraga Street, 403, São José dos Pinhais, PR 83060-500, Brazil
[5] Department of Nursing and Public Health, State University of Ponta Grossa, General Carlos Cavalcanti, 4748, Ponta Grossa, PR 84030-900, Brazil
[6] AAC&T Research Consulting LTDA, Curitiba, PR 82540-096, Brazil
[7] Department of Medicine, State University of Ponta Grossa, General Carlos Cavalcanti, 4748, Ponta Grossa, PR 84030-900, Brazil
[8] Department of Comparative Pathobiology, Purdue University, 725 Harrison Street, West Lafayette, Indiana 47907-2027, USA
[9] Corresponding author (email: abiondo@ufpr.br).

ABSTRACT: Rabies virus is recognized as one of the most fatal zoonotic agents affecting all mammals. Wild boars (*Sus scrofa*), classified as a large-size exotic invasive species in Brazil with nationwide hunting permitted, may serve as an extra blood source for the common vampire bat (*Desmodus rotundus*). Our aim was to document wild boar exposure to vampire bats to determine the seroprevalence of rabies virus antibodies in wild boars and to determine the immune status of hunters in southern and central-western Brazilian regions. Serum samples were collected from 80 wild boars and 49 hunters from natural and degraded areas of the Atlantic Forest biome of southern Brazil and in degraded areas of the Cerrado biome of central-western Brazil. The rabies-modified rapid fluorescent focus inhibition test was performed to detect the presence of rabies virus neutralizing antibodies in wild boars and considered seropositive when ≥0.10 IU/mL. The simplified fluorescence inhibition microtest was used for samples from hunters with a titer of ≥0.50 IU/mL and considered indicative of seroconversion. While 11% (9/80) of wild boars had serum titers for rabies exposure (≥0.10 IU/mL), 88% (43/49) of corresponding hunters lacked immune protective titers (<0.50 IU/mL). Wild boars showed serum titers for rabies likely due to contact with contaminated saliva of vampire bats or from infected carcass consumption. Additionally, Brazilian wild boars can be exposed to rabies and may play an important role in the sylvatic rabies cycle by providing a blood supply for vampire bats, highlighting the possibility of direct transmission of rabies virus to hunting dogs and hunters. These findings suggested hunters are a potential risk group for contracting rabies, and the World Health Organization may consider adding this occupation to their recommendations of who should receive the pre-exposure rabies vaccination.

Key words: Hunters, neutralizing antibodies, rabies, wild boars.

INTRODUCTION

The wild boar (*Sus scrofa*) is an exotic invasive species in Brazil that is under official hunting control and has been found in all six Brazilian biomes (IBAMA 2017). Roaming, rooting, and predatory habits of wild boars have impacts on biodiversity, agriculture crops, livestock pasture, and spreading of pathogens (IBAMA 2017; Kmetiuk et al. 2019). Two isolated rabies cases were detected in feral pigs in India (Daly et al. 2014; Nair and Jayson 2020).

Rabies is recognized as a lethal zoonotic disease affecting all mammals and one that is maintained in sylvatic areas by native terres-

trial and aerial animals that serve as reservoirs, particularly *Desmodus rotundus*, the common vampire bat, in Neotropical countries (Daly et al. 2014; WHO 2018a). Rabies primarily causes death in susceptive mammals by acute encephalomyelitis (Machado et al. 2012; Nair and Jayson 2020). Although oral vaccination against rabies by baiting, designed for foxes, has been used and monitored in wild boars of Europe, differentiation between natural infection and immunization may be challenging (Vengušt et al. 2011). Regardless, vaccination of native or exotic wildlife fauna, as observed in other parts of the world, has never been employed or recommended by the Brazilian Ministry of Health, likely due to a historical understanding that massive attenuated virus vaccination could lead to a return to virulence of the vaccine strain in nonvalidated native wildlife species, despite the fact that current vaccines contain only inactivated rabies virus.

From 2000 to 2017, a total of 188 cases of human rabies were registered in Brazil, of which 44% (82/188) were transmitted by bats (Vargas et al. 2019), with case growth associated with areas that overlap with the range of vampire bats (Johnson et al. 2014). Although exotic, free-range wild boars have reportedly been shown to provide an extra blood source for vampire bats in Brazil (Galetti et al. 2016; Pereira et al. 2016), no epidemiologic serosurvey of rabies virus exposure has been conducted in wild boars to date. Accordingly, our goals were to assess the exposure of wild boars to vampire bats by determining the seroprevalence of rabies virus antibodies and to evaluate the immune protection of hunters in southern and central-western Brazilian regions.

MATERIALS AND METHODS

Blood samples from wild boars and hunters were collected between October 2016 and May 2018 (Fig. 1). Free-ranging wild boars from degraded areas of the Atlantic Forest biome in southern Brazil and the Cerrado biome in central-western Brazil, defined as having environmental deterioration due to depletion of natural resources, were obtained from legally registered hunters. Blood samples were collected and the carcasses were buried following Brazilian hunting laws (IBAMA 2013). Both biome regions were selected based on distance apart, located in two different Brazilian biomes, and via previous agreement of local hunters to participate in concurrent hunter and wild boar samplings. In addition, free-range wild boars from a natural area in the Vila Velha State Park of southern Brazil were sampled. The presence of wild boars in the park has been evidenced by signs of rooting. Five areas in-park have been photo monitored, and wild boars have been attracted with corn and livestock salt. In these areas, box traps were installed and wild boars were baited, killed by firearm, and buried following Brazilian hunting laws. Finally, previously captured free-range piglets, kept and raised at two local farms of anthropized areas in southern Brazil, considered as captured wild boars, were also sampled after physical restraint and sedation with tiletamine-zolazepam (3 mg/kg) and xylazine (2.5 mg/kg) given intramuscularly (Allan et al. 1997). Blood was collected by jugular puncture.

Sample sizes were determined by using the maximum wild boar and hunter sampling within the available timeframe. Blood collection was performed by intracardiac puncture immediately after death in slaughtered wild boars and by cephalic venipuncture in hunters who agreed to participate in the study after signing a standard informed consent form and answering a questionnaire that contained several questions; among them, whether they had received rabies pre-exposure vaccination. Samples were placed in tubes without anticoagulant and kept at 25 C until visible clot retraction. Serum was then separated by centrifugation at $25 \times G$ for 5 min and stored at −20 C until processing. An epidemiologic questionnaire was used to collect data about sample location, sex, age, free-range or captured, capture area, biome, presence of vampire bats blood feeding, and body size of wild boars. Based on previous reports of vampire bat blood feeding (Galetti et al. 2016; Pereira et al. 2016), five trap cameras were placed in five locations within Vila Velha State Park from November 2017 to January 2018. Images obtained were individually analyzed for the presence of vampire bat blood-feeding on wild boars. Although all the wild boars were killed, only blood samples were taken for testing, as all animals were apparently healthy at the time of hunting. As some wild boars have shown presence of titers, all hunter samples were immediately tested as a public health emergency due to potential rabies infection during hunting, slaughter, and consumption.

The testing for neutralizing antibodies to rabies virus was performed at the Pasteur Institute (São Paulo, Brazil). Wild boar samples were tested by the modified rapid fluorescent focus inhibition test (RFFIT; Smith et al. 1996), which detects

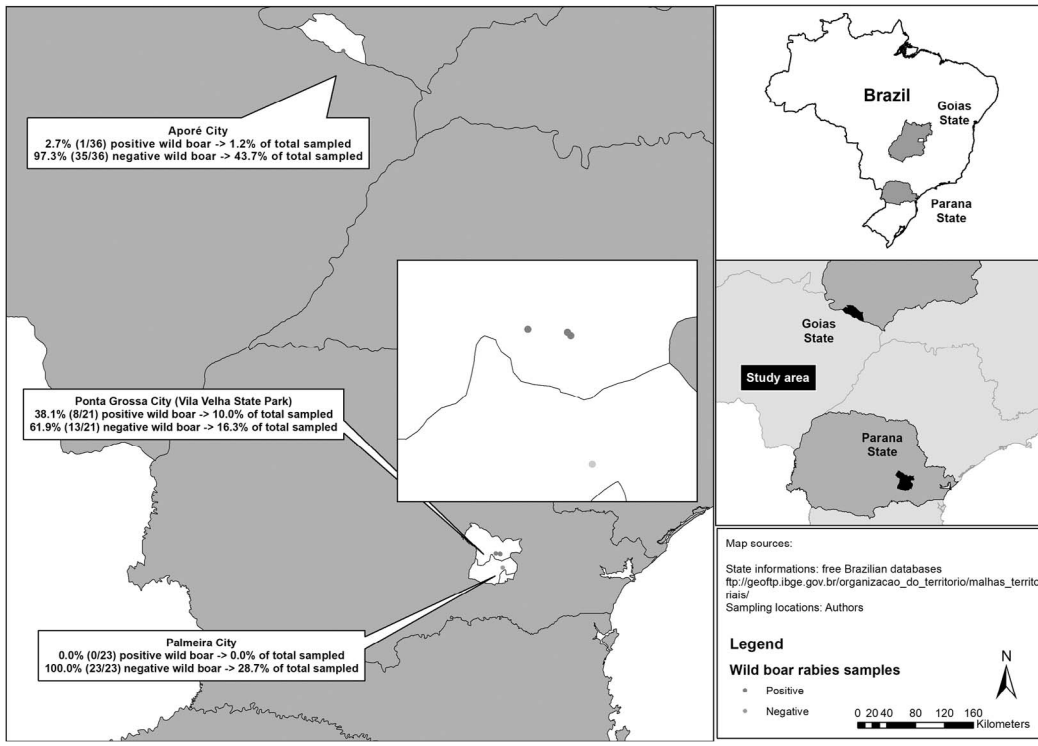

FIGURE 1. Locations in southern and central-western Brazil where wild boars (*Sus scrofa*) and hunters were sampled for rabies exposure.

neutralizing antibodies to rabies virus (Favoretto et al. 1993) The presence of neutralizing antibodies in unvaccinated animals indicates natural virus exposure (WHO 2018a). The 50% tissue culture infective dose ($TCID_{50}$) is the measure of infectious virus titer, and the test sera and were calculated according to the Spearman-Kärber method (Aubert 1996). The log 50% focus forming dose (FFD_{50}) titers were converted to an IU/mL value by comparing sample titer with the reference serum titer at 0.50 IU/mL (WHO 2018b) and considered seropositive when $\geq$0.10 IU/mL (WHO 2018a,b). Hunter samples were evaluated for rabies virus antibodies and tested by the simplified fluorescence inhibition microtest (SFIMT), a test with the same reproducibility as the RFFIT (Favoretto et al. 1993) in which the titer of $\geq$0.50 IU/mL is considered as adequate and indicative of seroconversion (WHO 2018b). The cutoff levels for rabies virus neutralizing antibody tests followed the international guidelines, where the value of 0.50 IU/mL is indicative of seroconversion after vaccination and 0.10 IU/mL is indicative of the presence of rabies virus neutralizing antibodies in wild animals or unvaccinated animals (WHO 2018b). The absolute and relative frequency of infection was calculated by stratifying the observations according to hunters and wild boars as well as the biome and capture area in which samples were collected. Epidemiologic analyses were performed based on a questionnaire associated with wild boar exposure to rabies antibodies; this included age, sex, sample location, and free-range or captured wild boar. These variables were compared by proportions using the chi-square test and significant differences were declared when $P<0.05$. This study has been approved by the Ethics Committees of Animal Use (no. 059/2017) and Brazilian National Human Use (no. 97639017.7.0000.0102). Wild boar trapping, slaughter, and burial were additionally approved by the Environmental Institute of Paraná (no. 30/17).

RESULTS

Blood samples were collected from 80 clinically healthy wild boars, including 56% (45/80) free-range following shooting, 17% (14/80) captured with chemical sedation and under physical restraint, and 26% (21/80) trapped and killed in natural areas of the state

TABLE 1. Results of associated risk factors for seropositivity of rabies virus antibodies in blood samples from 80 wild boars (*Sus scrofa*) in Brazil tested by the rapid fluorescent focus inhibition test.

Independent variables	Total		Seropositive		Seronegative		Odds ratio (95% confidence interval)	*P* value
	%	*n*	%	*n*	%	*n*		
Sample location								
Central-western Brazil	45	36/80	3	1/36	97	35/36	Reference	
Vila Velha State Park	26	21/80	38	8/21	62	13/21	21.54 (2.45–189.42)	0.001
Southern Brazil	29	23/80	0	0	100	23/23	Not calculated	0.610
Sex								
Female	63	51/80	10	5/51	90	46/51	Reference	
Male	36	29/80	14	4/29	86	25/29	0.68 (0.17–2.76)	0.716
Age								
<6 mo	22	18/80	17	3/18	83	15/18	Reference	
>6 mo and <1 yr	17	14/80	7	1/14	93	13/14	0.38 (0.04–4.16)	0.613
>1 yr	60	48/80	10	5/48	90	43/48	0.58 (0.12–2.73)	0.673
Captured[a] or free-range wild boars								
Captured (from January 2018 to May 2018)	17	14/80	0	0	100	14/14	Reference	
Free-range (from October 2016 to May 2018)	82	66/80	14	9/66	86	57/66	Not calculated	0.160
Capture areas								
Agricultural	56	45/80	2	1/45	98	44/45	Reference	
Natural	26	21/80	38	8/21	62	13/21	27.08 (3.09–236.90)	0.0002
Anthropized	17	14/80	0	0	100	14/14	Not calculated	0.012
Biome								
Cerrado	45	36/80	3	1/36	97	35/36	Reference	
Atlantic Forest	26	21/80	38	8/21	62	13/21	21.54 (2.45–189.42)	0.001
Degraded Atlantic Forest	29	23/80	0	0	100	23/23	Not calculated	0.001
Presence of bat blood feeding								
Yes	64	7/11	29	2/7	71	5/7	7.50 (0.46–122.70)	0.242
No	36	4/11	75	3/4	25	1/4	Reference	
Body size								
Medium	21	17/80	35	6/17	65	11/17	10.91 (2.37–50.27)	0.002
Large	79	63/80	5	3/63	95	60/63	Reference	

[a] Free-range piglets, kept and raised at two local farms of anthropized areas in southern Brazil.

park. A total of 49 hunters were sampled during visits to both regions. Overall, 11% (9/80) of hunted wild boar samples tested by RFFIT were considered seropositive, including 38% (8/21) from within the park and 3% (1/36) from central-western Brazil, with titers of 0.12, 0.13, 0.13, 0.14, 0.16, 0.20, 0.22, and 0.35 IU/mL in southern and 0.10 IU/mL in central-western Brazil (Table 1).

A total of 88% (43/49) of hunter blood samples were tested by the SFIMT, and results were below 0.5 IU/mL in this study, indicating a lack of effective immune protection against rabies. Only 18% (9/49) of hunters reported rabies vaccination and 67% (6/9) had their titers previously tested. A total of 11,112 images were registered by trap cameras installed at the Vila Velha State Park that showed 94 different wild boars, 14% (13/94) with vampire bats blood feeding.

Associated risk factors for wild boars were statistically significant for sample location and biome (P=0.001). Wild boars inside the parks had a 21.5-fold greater chance to be seropositive than did those in the central-western area. Wild boars in natural areas had a 27.1-fold greater chance to be seropositive than did agricultural areas. Although wild boars with

medium body scores were 10.9-fold more likely to be seropositive than were wild boars with high body scores, most of the wild boars inside the park were in the large body category and may have biased the results. No significant differences were observed for wild boar seropositivity and sex, age, captured or free-range wild boars, and presence of blood feeding (Table 1).

DISCUSSION

In our study, 14% (13/94) of wild boars with vampire bats blood feeding were detected by trap cameras. These results corroborated previous studies that identified wild boars as a blood source for vampire bats in the Pantanal and Atlantic Forest biomes (Galetti et al. 2016; Pereira et al. 2016). A previous comprehensive survey of bat rabies in Brazil from 1996 to 2009 found 41 nonhematophagous rabies virus-infected species belonging to 25 genera and three families, of which 15 genera were in the Phyllostomidae family, 29% (12/41) of species and six genera were in the Vespertilionidae family, and 27% (11/41) of species and four genera were in the Molossidae family (Sodré et al. 2010). In another study, a total of 5% (38/828) of bats was found in northeastern Brazil by fluorescent antibody test (FAT) and the mouse inoculation test (MIT; Cordeiro et al. 2016), while 17% (183/1,047) of bats were reactive in northern Brazil by SFIMT (Almeida et al. 2019). A previous study of two domestic pigs (*Sus scrofa*) from northeastern Brazil with suggestive neurologic signs confirmed rabies caused by a strain phylogenetically related to the rabies virus strain frequently found in vampire bats (Pessoa et al. 2011).

Routine, laboratory-based surveillance for rabies in wildlife living in study areas herein, and throughout Brazil, has been based on submission of the central nervous tissue of wild animals with suspected disease, usually based on aggressiveness and human contact, to the State or National Reference Laboratories. The labs used FAT for a rapid result, MIT for confirmation of the rabies diagnosis, and culture for viral characterization. Of all human rabies cases reported from 1992 to 2012, a total of 28% (121/428) were caused by wildlife encounters, with 86% (104/121) associated with bats, 11% (13/121) with primates, 2% (2/121) with foxes, and 2% (2/121) with raccoons. According to the Brazilian Rabies Annual Reports, no wild boars were tested in Brazil (Ministry of Health [Brazil] 2020).

In our study, 11% (9/80) of slaughtered wild boars were considered seropositive. Although the remaining wild boars lacked neutralizing antibodies against rabies virus, antibody presence may appear only in advanced clinical disease. Despite contaminated saliva as the main transmission route of rabies (Jorge et al. 2010), experimental ingestion of rabid mouse carcasses has infected red foxes (*Vulpes vulpes*) and striped skunks (*Mephitis mephitis*), with some adults surviving oral intake and having circulating neutralizing antibodies (Ramsden and Johnston 1975). Thus, the low titers of neutralizing antibodies found herein in wild boars may be the result of nonlethal contact, such as less virulent or more immunogenic rabies virus strains (Rosatte and Gunson 1984), through the consumption of infected carcasses of mammals that died of rabies (Mshelbwala et al. 2013) or rabies virus proteins presented by bat saliva during blood feeding (Johnson et al. 2014). In addition, a factor complicating the interpretation of rabies serology tests is the possibility of cross-reactivity with other lyssaviruses. There have been human clinical cases in which the indirect FAT gave false rabies seropositive results in patients with encephalitis caused by West Nile virus, but this is probably unlikely in Brazil.

Studies evaluating the presence of neutralizing antibodies to rabies in wild boars conducted in Europe examined whether the boars had ingested oral rabies vaccine baits that were targeting foxes (Vengušt et al. 2011; Dascalu et al. 2019). They found that 16% (122/746) of wild boar samples captured in Slovenia showed neutralizing antibody titers to rabies virus tested with the fluorescent antibody virus neutralization assay (Vengušt et

al. 2011), while in Romania, 42% (132/312) of wild boars had antibodies against rabies by an enzyme-linked immunosorbent assay (Dascalu et al. 2019).

A previous rabies study in Brazil from 2002 to 2012 using the standard 0.10 IU/mL cutoff identified 460 positive terrestrial wild mammals and 1,703 positive bats, with infected hematophagous bats found mostly in central-western Brazil (Rocha et al. 2017). Another study in central-western and southeastern Brazil, using the same cutoff, showed a 12% (26/211) seropositive rate in wild carnivore samples including maned wolf (*Chrysocyon brachyurus*), crab-eating fox (*Cerdocyon thous*), jaguar (*Panthera onca*), ocelot (*Leopardus pardalis*), bush dog (*Speothos venaticus*), puma (*Puma concolor*), pampas cat (*Leopardus pajeros*), and crab-eating raccoon (*Procyon cancrivorus*), thus identifying native carnivores as rabies reservoirs potentially infected by bat blood feeding or by ingesting bats (Jorge et al. 2010). In another study, 33% (105/317) of wild canids from northeastern Brazil tested seropositive by the FAT and the MIT (Cordeiro et al. 2016).

Native or exotic wildlife fauna may also be exposed to rabies virus in fragmented biomes or more-anthropized areas of Brazil, as rabies was documented in 11% (4/36) of capuchin monkeys (*Cebus capucinus*; Machado et al. 2012). Because wild boars are found in natural, agricultural, and anthropized areas worldwide, they may serve as rabies sentinels for public health and epidemiologic purposes.

Rabies infections in wild boars have been reported after attacks on three people, resulting in recommendations for mass rabies vaccination and titer monitoring in areas that have experienced an increase in wild boar aggression (Nair and Jayson 2020), based on WHO guidelines and recommendations (WHO 2018a). In Brazil, a total of 82 human rabies cases between 2002 and 2012 were due to transmission from wild animals, with 90% (74/82) originating from vampire bats, 5% (4/82) from nonhuman primates, and 5% (4/82) from wild or domestic herbivores (Rosa et al. 2018).

Hunter proximity to wildlife, particularly to serologically reactive animals, may create a major public health problem (Antunes et al. 2017; Rocha et al. 2017). Wild boar meat is frequently used as a protein source by Brazilian hunters, who can be exposed to saliva and brain or nervous system tissue while slaughtering the animals (Rosa et al. 2018). Furthermore, hunting dogs are the most popular method for wild boar tracking in Brazil, and dogs may be wounded by wild boar bites (Rosa et al. 2018). In addition, of the 25 human rabies cases reported in Brazil from 2010 to 2017, 32% (8/25) were caused by bats, 32% (8/25) by dogs, 16% (4/25) by cats, 16% (4/25) by nonhuman primates, and 4% (1/25) by unknown causes (Vargas et al. 2019). In the present study, all hunting dogs had been vaccinated for rabies, as reported by the owners. However, the frequency of vaccination and the presence of antibody titers of hunting dogs were not declared. Given that the hunters are at risk of contracting rabies, WHO may consider adding this occupation to their recommendations about who should receive the pre-exposure rabies vaccination, along with those in an already-identified job such as laboratory workers handling live rabies viruses, people (veterinarians, animal disease control staff, and wildlife rangers) whose activities may expose them to direct contact with bats or other potentially infected mammals, travelers visiting rabies-affected areas, and children living in or visiting high risk areas (WHO 2018a).

A major limitation of this study is the low number of samples for wild boars and hunters. Although the study was conducted between October 2016 and May 2018, camera observations occurred only from November 2017 through January 2018 due to wild boar capture authorization inside the park. Difficulties in obtaining simultaneous access to human and wild boar populations, or refusal to participate in a study, may explain the lack of previous studies. In addition, there is a chance of collecting serum samples of poor quality when samples are collected from hunted animals. Postmortem samples can have increased cytotoxicity, which can affect

neutralization tests, resulting in a higher false-positive rate. This needs to be taken into consideration when interpreting and reporting results.

Our results show that invasive wild boars can be exposed to rabies and may play an important role in the sylvatic rabies cycle of Brazil, either indirectly by providing blood supplies for vampire bats, particularly in naturally preserved areas, or directly through transmission of rabies virus to hunting dogs and hunters.

ACKNOWLEDGMENTS

This research was supported by Veterinary Science Graduate Program at Federal University of Paraná, Pasteur Institute, and Paraná State Reference Laboratory. We thank Suzanne Pratt for editing and improving the article. We thank Ismail da Rocha Neto and Osvaldir Hartmann for helping with wild boar captures and samplings, biologist Mauro de Moura-Britto and the personnel of the Environmental Institute of Paraná for technical support, Vila Vela State Park for authorization and internal access, and Gerson Luiz Lopes Goularte of Paraná Agribusiness Defense Agency for his research collaboration. P.I.T.Jr. was supported by a graduate fellowship from the Coordination for the Improvement of Higher Education Personnel-Brasília, Distrito Federal 70.040-020 Brazil.

LITERATURE CITED

Allan FJ, Thompson KG, Frey B. 1997. The use of an anaesthetic combination in pigs. *N Z Vet J* 45:125.

Almeida MF, Rosa AR, Martorelli LFA, Kataoka APAG, Aires CC. 2019. Rabies virus monitoring in bat populations in Rondônia state, Brazil. *Rev Soc Bras Med Trop* 52:e20180199.

Antunes JMAP, Demoner LC, Cruvinel TMA, Kataoka AP, Martorelli LFA, Machado GP, Megid J. 2017. Rabies virus exposure of Brazilian free-ranging wildlife from municipalities without clinical cases in humans or in terrestrial wildlife. *J Wildl Dis* 53:662–666.

Aubert MFA. 1996. Methods for the calculation of titers. In: *Laboratory techniques in rabies*, 4th Ed., Meslin F-X, Kaplan MM, Koprowski H, editors. World Health Organization, Geneva, Switzerland, pp. 445–459.

Cordeiro RA, Duarte NFH, Rolim BN, Soares Júnior FA, Franco ICF, Ferrer LL, Almeida CP, Duarte BH, de Araújo DB, Rocha MF, et al. 2016. The importance of wild canids in the epidemiology of rabies in northeast Brazil: A retrospective study. *Zoonoses Public Health* 63:486–493.

Daly CD, Indu K, Vijayan. 2014. A case of rabies in a wild pig. *Indian J Sci Res Tech* 5:23–24.

Dascalu MA, Wasniewski M, Picard-Meyer E, Servat A, Bocaneti FD, Tanase OI. 2019. Detection of rabies antibodies in wild boars in north-east Romania by a rabies ELISA test. *BMC Vet Res* 15:466.

Favoretto SR, Carrieri ML, Tino MS, Zanetti CR, Pereira OAC. 1993. Simplified fluorescent inhibition microtest for the titration of rabies neutralizing antibodies. *Rev Inst Med Trop São Paulo* 35:171–175.

Galetti M, Pedrosa F, Keuroghlian A, Sazima I. 2016. Liquid lunch—Vampire bats feed on invasive feral pigs and other ungulates. *Front Ecol Environ* 14:505–506.

IBAMA (Brazilian Institute of Environment and Natural Renewable Resources). 2017. *National plan for prevention, control and monitoring of wild boars (*Sus scrofa*) in Brazil*. Brazilian Ministry of Environment and Ministry of Agriculture and Livestock, Brasilia, Brazil. https://www.mma.gov.br/images/arquivo/80046/Especies/2017-PlanoJavali-2017.2022.pdf. Accessed September 2020. Portuguese.

IBAMA. 2013. *Normative instruction, no. 3: January 31, 2013*. http://www.ibama.gov.br/legislacao/javali. Accessed April 2021.

Johnson N, Aréchiga-Ceballos N, Aguilar-Setien A. 2014. Vampire bat rabies: Ecology, epidemiology and control. *Viruses* 6:1911–1928.

Jorge RS, Pereira MS, Morato RG, Scheffer KC, Carnieli P Jr, Ferreira F, Furtado MM, Kashivakura CK, Silveira L, Jacomo AT, et al. 2010. Detection of rabies virus antibodies in Brazilian free-ranging wild carnivores. *J Wildl Dis* 46:1310–1315.

Kmetiuk LB, Krawczak FS, Machado FP, Paploski IAD, Martins TF, Teider-Junior PI, Serpa MCA, Barbieri ARM, Bach RVW, Barros-Filho IR, et al. 2019. Ticks and serosurvey of anti-*Rickettsia* spp. antibodies in wild boars (*Sus scrofa*), hunting dogs and hunters of Brazil. *PLoS Negl Trop Dis* 13:e0007405.

Machado GP, Antunes JMAP, Uieda W, Biondo AW, Cruvinel TMA, Kataoka AP, Martorelli LF, de Jong D, Amaral JM, Hoppe EG, et al. 2012. Exposure to rabies virus in a population of free-ranging capuchin monkeys (*Cebus apella nigritus*) in a fragmented, environmentally protected area in southeastern Brazil. *Primates* 53:227–231.

Ministry of Health (Brazil). 2020. *Epidemiological situation*. http://portalms.saude.gov.br/saude-de-a-z/raiva/situacao-epidemiologica. Accessed September 2020. Portuguese.

Mshelbwala PP, Ogunkoya AB, Maikai BV. 2013. Detection of rabies antigen in the saliva and brains of apparently healthy dogs slaughtered for human consumption and its public health implications in Abia State, Nigeria. *ISRN Vet Sci* 2013:468043.

Nair RP, Jayson EA. 2020. Wild pig rabies—A case study from Pathippara, Malappuram, Kerala. *Int J Res Med Basic Sci* 6:1–5.

Pereira AD, Batista CB, Bender D, dos Reis NR, Bazilio S. 2016. Report on *Desmodus rotundus* (Chiroptera, Phyllostomidae) feeding on *Sus scrofa* (Artiodactyla, Suidae) blood. *Bol Soc Bras Mastozool* 77:151–153.

Pessoa CRM, Silva MLCR, Gomes AB, Garcia AIE, Honmalto F, Franklin RC. 2011. Paralytic form of rabies in pigs. *Braz J Microbiol* 42:298–302.

Ramsden RO, Johnston DH. 1975. Studies on the oral infectivity of rabies virus in Carnivora. *J Wildl Dis* 11:318–324.

Rocha SM, de Oliveira SV, Heinemann MB, Gonçalves VSP. 2017. Epidemiological profile of wild rabies in Brazil (2002–2012). *Transbound Emerg Dis* 64:624–633.

Rosa CA, Wallau MO, Pedrosa F. 2018. Hunting as the main technique used to control wild pigs in Brazil. *Wildl Soc Bull* 42:111–118.

Rosatte RC, Gunson JR. 1984. Presence of neutralizing antibodies to rabies virus in striped skunks from areas free of skunk rabies in Alberta. *J Wildl Dis* 20:171–176.

Smith JS, Yager PA, Baer GM. 1996. A rapid fluorescent focus inhibition test (RFFIT) for determining rabies neutralising antibody. In: *Laboratory techniques in rabies*, 4th Ed., Meslin XF, Kaplan MM, Koprowski H, editors. World Health Organization, Geneva, Switzerland, pp. 200–208.

Sodré MM, Gama AR, Almeida MF. 2010. Updated list of bat species positive for rabies in Brazil. *Rev Inst Med Trop São Paulo* 52:75–81.

Vargas A, Romano APM, Merchán-Hamann E. 2019. Human rabies in Brazil: A descriptive study, 2000–2017. *Epidemiol Serv Saude* 28:e2018275.

Vengušt G, Hostnik P, Cerovšek M, Cilenšek P, Malovrh T. 2011. Presence of antibodies against rabies in wild boars. *Acta Vet Hung* 59:149–154.

WHO (World Health Organization). 2018a. *Laboratory techniques in rabies*, 5th Ed., Rupprecht CE, Fooks AR, Abela-Ridder B, editors. WHO, Geneva, Switzerland, pp. 1–289.

WHO. 2018b. Expert consultation on rabies. 3rd Report. *Technical Report Series 1012*. WHO, Geneva, Switzerland, pp. 1–184.

Submitted for publication 22 March 2020.
Accepted 29 August 2020.