

# Evaluation of rabies virus in the *Cerdocyon thous* in the atlantic forest

## Abstract

The ability of the virus to invade and sustain in a variety of mammalian hosts is an issue of serious concern and wild rabies may emerge as a new public health problem, due to its global distribution. In Latin America, countries are working to decrease the incidence of the virus by controlling the disease in domestic animals. The present study evaluated dead specimens of *Cerdocyon thous* due motor vehicles accidents on state highway ES-060. The study involved 16 crab-eating foxes with their necropsy examinations and their brain tissues were collected. Biological samples were identified, frozen and sent to the Rabies Diagnosis Laboratory of the Agricultural and Forestry Defense Institute of the State of Espírito Santo for further tests using direct immunofluorescence technique and biological test by intracerebral inoculation in Swiss mice. This is the first investigation of rabies virus in *Cerdocyon thous* from a region comprising of two biological reserves in south eastern Brazil, with negative results for all samples. Despite the absence of cases in the analyzed samples, improvements in the surveillance of rabies virus in wild animals can be pointed out, as the fauna research is still very small.

**Keywords:** public health, zoonotic diseases, wild carnivores, conservation

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## Introduction

The occurrence of rabies virus, (a zoonosis of the genus Lyssavirus, family *Rhabdoviridae*) is considered as a major threat to the conservation of wild animals, public and animal health.<sup>1</sup> Transmitted by the saliva of terrestrial mammals and Chiroptera infected in episodes of biting, rabies causes acute viral encephalomyelitis framework of progressive character, affecting the central nervous system of all species of mammals.<sup>2-4</sup> Added to the urbanization, the change in environment and ecology of the host favors the pathogen, in transmitting the diseases.<sup>5,6</sup> Despite the fact, that rabies is a global issue of health concern and is spread all over the world, several, countries have attempted to reduce the incidence of rabies in humans through the control of the disease in dogs. But it is likely that the wild rabies can infect a range of mammalian hosts.<sup>7,8</sup> As reported in northeastern region of Brazil, wild canids as crab-eating fox are considered as the primary reservoir of rabies and can affect domestic animals and humans.<sup>8-11</sup> This in turn complicates the practice of control measures, which are reflected in the small number of vaccinated dogs.<sup>12</sup> This study evaluated specimens of dead *Cerdocyon thous* on State Highway ES-060, to disclose the occurrence of rabies.

## Materials and methods

The present study evaluated a total of 16 crab-eating foxes free-living collected dead, due to collision with vehicles. The study took place from 2012 to 2015 and the stretch of State Highway ES-060, was investigated to reveal the prevalence of the rabies virus. The State Highway ES-060 links the municipalities of Vitória to Guarapari, passes through two areas of environmental protection, the Parque Estadual Paulo César Vinha (1,500ha) and the Municipal Park of Jacarenema (307ha). Regularly, the company which administers the

highway 67 km of track monitors, each 1:30h, 24 hours a day. The animals found living and/or dead are promptly collected, and the place and time of the occurrence are recorded. Biodiversity Information and Authorization System with registration n° 49417-1 and the Ethics Committee of the University Vila Velha with registration n° 351-2015, performed necropsies exams and subsequently collected the injured material, after opening the braincase of the specimens, through dental use diamond blades, attached to a Dremel® micro-retífica. Authorized by the Biological samples were identified, frozen and were later forwarded to the laboratory for Diagnosis of rabies for tests carried out using the Immunofluorescence technique (IFT) and biological evidence through intracerebral inoculation in Swiss albino mice, as recommended by the World Organisation for Animal Health, through the technique described in the normative instruction n° 8 April 12, 2012, with some modifications.

## Results and discussion

All 16 samples obtained negative results for the rabies virus. This suggests that the cause of the accident was not due to neurological changes resulting from infection by rabies virus in species involved. Although the animals were killed by accidents, is necessary to monitor the highway for every 24 hours in a day, to reduce the decomposition rate. In addition, studies have shown that the variation in temperature interferes with test results to some extent which is negligible, showing positive tests to the rabies virus, in carcasses of animals subjected to high temperatures, as well as low temperatures and in stage of putrefaction.<sup>13,14</sup> Two techniques have been used to confirm the diagnosis, but IFT is not very effective when the viral protein is in smaller quantity.<sup>15</sup> Technique of intra cerebral inoculation in mice was used, but there may be loss of replication capacity in cases where there is a decomposed sample where the virus may no longer

replicate. Although there is a small probability of false-negative exams it is known that several surveys in the states and regions on Brasil.<sup>16</sup> as well as in other countries of Latin American there is a decrease in the urban cycle. Despite the success of controlling rabies in dogs and cats, the epidemiological characteristics of rabies in other domestic animals and bats are still very high, demonstrating the importance of wildlife as a potential source of human rabies.<sup>17-19</sup> The increase in aerial rabies cycles composed of bats hematophagous and non-hematophagous, from rural and wild cycles, consequently can promote the transmission of the virus among the same.<sup>12</sup> In this sense, some studies have shown evidence with antigenic variants related to the virus maintained and transmitted by domestic canids in countries like Brazil, Argentina, Bolivia and Paraguay.<sup>20-22</sup> Therefore, it is evident that the virus of rabies is circulating between states of the same country.<sup>23</sup> But due to the fact that the country has a very complex mammalian fauna, the main hosts of wild rabies cycle can vary according to specificity and regional character.<sup>24</sup> However, it is necessary to mention the importance of the collaboration between states and countries for the control of infectious diseases, as the number of samples analyzed from wild terrestrial animals is small compared to other species of domestic animals.

## Conclusion

Although this study presents negative results for the rabies virus in the *Cerdcoyon thous* species of the study region, a better profile of the rabies situation in wild animals is necessary for surveillance to be expanded to increase the number and constancy with which the samples are sent for laboratory diagnosis. Thus it is possible to trace rabies virus occurrence in these populations and disclose the role of wild population in spreading the infection or not. The dead animals along highways used in this study facilitate the access to research material which in turn provides the further insights of the research investigation and epidemiological surveillance of wild species.

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## Conflicts of interest

The authors declare no conflict of interest.

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