PERSISTENCE AND DISSEMINATION OF VACCINIA VIRUS ZOONOTIC INFECTION IN CENTRAL WEST REGION OF SÃO PAULO STATE- BRAZIL

PERSISTÊNCIA E DISSEMINAÇÃO DE INFECÇÃO ZOONÓTICA DO VÍRUS VACCINIA NA REGIÃO CENTRO OESTE DO ESTADO DE SÃO PAULO-BRASIL

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ABSTRACT
The aim of this study was to describe the persistence and dissemination of Vaccinia Virus (VACV) in Central West region of São Paulo State-Brazil and the zoonotic character of the disease. The work suggests viral circulation and maintenance of VACV by possible wildlife reservoir.

KEY WORDS: Vaccinia virus; São Paulo state; persistence; dissemination, wildlife.

INTRODUCTION
In São Paulo State, Brazil, the Vaccinia Virus (VACV) strain was obtained in 1966 from United States to produce vaccines Trindade et al (2007). Orthopoxviruses have been repeatedly isolated in Brazil and identified as VACV by classical immunologic, virologic, and molecular methods Trindade et al (2007). According to Chomel et al (2007) most emerging infectious diseases are zoonotic, and wildlife constitutes a large and often unknown reservoir. The reservoirs for these viruses in the wild are not well understood, and in Brazilian wilderness the isolates from monkeys revealed a VACV strain commonly associated with the outbreaks among cattle and humans (3). Since 1999, a number of outbreaks affecting cattle and humans have been reported in Brazil, being most of the outbreaks occurred in southeastern Brazil; some outbreaks, however, are 2500 km away from the main affected region indicating circulation of viral strains Trindade et al (2007). The aim of this study was to describe circulation and persistence of VACV in Central West region of São Paulo State-Brazil.
In São Paulo State, VACV was reported in 2003 Trindade et al, 2004 Nagasse-Sugahara et al, 2008 Megid et al and 2012 Megid et al. In Torre de Pedra city (23° 14′ 38″ S, 48° 11′ 42″ W) an outbreak of VACV with lesions in cattle (Figure 1B) and milkers (Figure 1C; 1D) was described Megid et al (2008), however, after one year of this first outbreak (outbreak 1), another outbreak was detected in Itatinga city (23° 6′ 7″ S, 48° 36′ 57″ W) (outbreak 2) and again in Torre de Pedra city (23° 14′ 38″ S, 48° 11′ 42″ W) (outbreak 3) resulting positive for VACV Megid et al (2012) (Figure 1A). These outbreaks (outbreak 1 and 2) have 100 km of distance from each other and natural geographic barriers exist between the farms. The clinical signs described in the milkers during the outbreaks Megid et al (2008), Megid et al (2012) were mainly headache, limphadenopathy, fever and ulcerated lesions on fingers and fists; reinforcing the zoonotic character of the infection. The strain responsible for the second and third VACV outbreaks grouped with the others Brazilian VACV, including ARAV Trindade et al (2003), Megid et al (2012) indicating that the same virus is responsible for these outbreaks independently from the distance between them, and suggesting a possible common source of infection or virus reservoirs. The animal movement is the cause of establishment of VACV in Amazon biome, Brazil Quixabeira-Santos et al (2011). However, in our report the owners denied animal movement between the farms where the outbreaks occurred. According to Medaglia et al (2009), the virus can spread in different regions; Cantagalo virus (CTGV) that was isolate in Rio de Janeiro State in 1999 was re-isolated in northern Brazil in two cities in the State of Tocantins, which are 12 km distant from each other. The authors considered the infection in Tocantins State a spread of CTGV to northern Brazil, reflecting propagation of the virus due to movement of animals or people Quixabeira-Santos et al (2009). Transmission among animals occurs mainly through the milker’s hands or mechanical milking equipment, while the disease is transmitted from animals to humans thorough contact with the lesions in cow’s teats Medeiros-Silva et al (2010). In Brazil, since 1960, innumerous outbreaks related to VACV have been documented by several research groups in different regions, and usually occurs in small rural properties with dissemination from cattle to milkers and vice-versa Medeiros-Silva et al (2010). Emerging infectious diseases have a major effect on human health and animals, particularly wild animals, are the source of >70% of all emerging infections Kuiken et al (2003). Leading factors for emergence of zoonoses are forest exploitation and aggressive agricultural development Chomel et al
The affected farms have in common the presence of rodents, forest and eventually others wild animals. The properties had deforestation for agricultural activities purposes and due human population expansion. In Namibia and Malaysia reported outbreaks of zoonoses after ecologic and environmental changes associated with deforestation and increase of human population Daszak et al (2001), Moussatche and Damaso et al 2008. The occurrence of outbreaks in new areas, where the disease was never diagnosed, as well as in the region where the disease was previously observed indicates viral spread and persistence (Table 1; Figure 1A).

Table 1- Dissemination of VACV in animals and human beings.

<table>
<thead>
<tr>
<th>Animals</th>
<th>Milkers/relatives</th>
</tr>
</thead>
<tbody>
<tr>
<td>Outbreak 1</td>
<td>PCR, RFLP, virus isolation in cell culture, and electron microscopy and serology positive</td>
</tr>
<tr>
<td>PCR, RFLP, virus isolation in cell culture, and electron microscopy positive</td>
<td></td>
</tr>
<tr>
<td>Outbreak 2</td>
<td>PCR, RFLP, virus isolation in cell culture, and electron microscopy and serology positive</td>
</tr>
<tr>
<td>PCR, RFLP, virus isolation in cell culture, and electron microscopy positive</td>
<td></td>
</tr>
<tr>
<td>Outbreak 3</td>
<td>PCR positive</td>
</tr>
<tr>
<td>Serology positive</td>
<td></td>
</tr>
</tbody>
</table>

PCR: Polymerase Chain Reaction; RFLP: Restriction Fragment Length Polymorphism

The results suggest that the VACV is able to persist in a reservoir not yet defined in nature. According to Abrahão et al (2009), this reservoir may be a rodent responsible for viral persistence as well as viral spread, transmission, circulation and environmental maintenance. The three reported episodes of VACV in central west of São Paulo state, and at the same farms two years after the first outbreak, including the distance between the farms, suggest viral circulation, dissemination and persistence of the virus. The study raises the possibility that the VACV have been established in wildlife host or peridomestic animals and the transmission occurred due to accidental contact with cattle. Most animal diseases for which surveillance programs exist are related to farm animals, therefore, development of programs for surveillance and for monitoring diseases in wildlife reservoirs are essential. The Brazilian
public health authorities and research groups should pay particular attention to this potential dissemination, persistence and zoonotic potential of VACV.

**Figure 1.** Persistence of Vaccinia virus in Central West region of São Paulo State- Brazil. A- Map from São Paulo State showing the outbreaks. The first outbreak reported in 2008 in Torre de Pedra city 1 (outbreak 1) following to the Itatinga city (outbreak 2), and finally returned to the third episode in Torre de Pedra city again (outbreak 3); B- Crusted lesions of VACV on udders; C-D crusted lesions of VACV in the hands (C) and finger (D) of milkers.

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