Monitoring the infection of a new strain of simian betaretrovirus in Southern Muriquis from Brazil

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BACKGROUND

Neotropical primates are natural hosts of several viruses, but many are poorly studied. One example was the identification through massive sequencing of a new strain of simian betaretrovirus (SRVbar) in an individual (2506) of Southern Muriqui (*Brachyteles arachnoides*), a critically endangered species, who died of immunodeficiency at the Primatology Center of Rio de Janeiro (CPRJ). The phylogenetic analysis showed that the SRVbar grouped close to the SRV of Asian primates, which also causes clinical signs similar to those presented by muriqui. Five muriquis from the same enclosure as 2506 were also positive for SRVbar, but were asymptomatic. Therefore, the objective of the study was to monitor the infection of SRVbar in specimen 2506 and in its asymptomatic contacts.

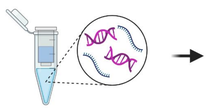
METHODS

1 Sample colection



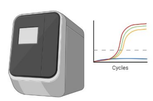
Oral swab and blood samples were collected from six adult muriquis living in the same CPRJ enclosure between 2016 and 2020.

2 Nucleic acids extraction



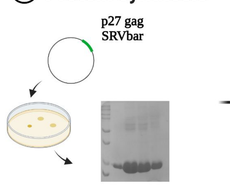
DNA was extracted from blood and oral swab samples and RNA was extracted from plasma samples.

3 Quantitative PCR



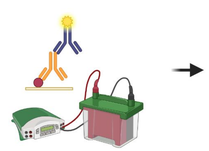
For analysis of proviral and viral load, a quantitative PCR was performed targeting a 310 bp region of the *env* gene of SRVbar

4 Protein synthesis



To analyze the presence of antibodies against SRVbar, a p27Gag recombinant protein was synthesized on the *Escherichia coli* cell and purified by affinity chromatography

5 Western Blot



The p27Gag protein was used as viral antigen in the Western Blot with plasma samples from muriquis.

6 Statistical analysis



The graphs and statistical analyzes were carried out in GraphPad prism 10.

For the first time, a study on the infection of a **simian betaretrovirus** with replicative potential and causing **immunodeficiency** in **Neotropical primates.**

Viral and proviral load of SRVbar were detected in asymptomatic and symptomatic animals, being 10x higher in the symptomatic animal.

It was detected antibodies against the SRVbar p27Gag protein, but with variation in detection over time and a possible correlation with viral load, as observed in Asian monkeys.

RESULTS

In two asymptomatic animals (CPRJ2049 and CPRJ3078) with more points collected, the proviral load in the blood was 10x higher than in the oral swab (p=0.01) **(Figure 1a**). Specimen CPRJ2506 showed similar proviral DNA loads in blood and oral swab (log 8.12 copies/10^6 cells and 7.66 copies/10^6 cells, respectively), 10x higher than asymptomatic animals (log 7.66 copies /10^6 cells and 6.57 copies/10^6 cells, respectively) (p=0.038). All asymptomatic animals tested had detectable viral loads and the average was 2.34 log10 copies/mL of plasma (1.4 - 3.18, min-max) while CPRJ2506 had a viral load greater than 3.96 log10 copies/mL of plasma (p=0.044) (**Figure 1b**). The seropositivity for p27Gag antigen varied between animals over time (**Figure 2a**) with an inverse correlation with viral load, being statistically significant (p=0.006) (**Figure 2b e 2c**).

Figure 1. Analysis of SRVbar proviral and viral load in samples from captive muriquis. a) Comparison of SRVbar proviral loads in blood and oral swab of each specimen and b) Analysis of RNA viral load in the plasma of symptomatic CPRJ2506 and asymptomatic animals (CPRJ3078 and CPRJ2049).

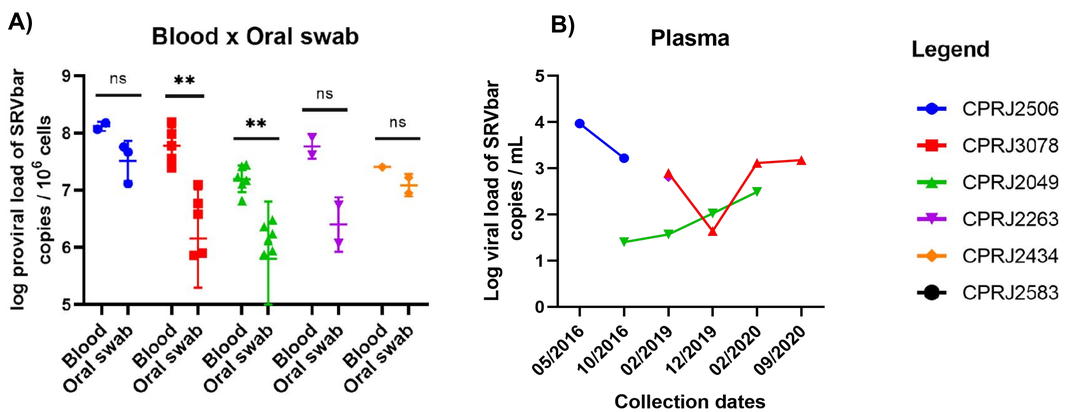
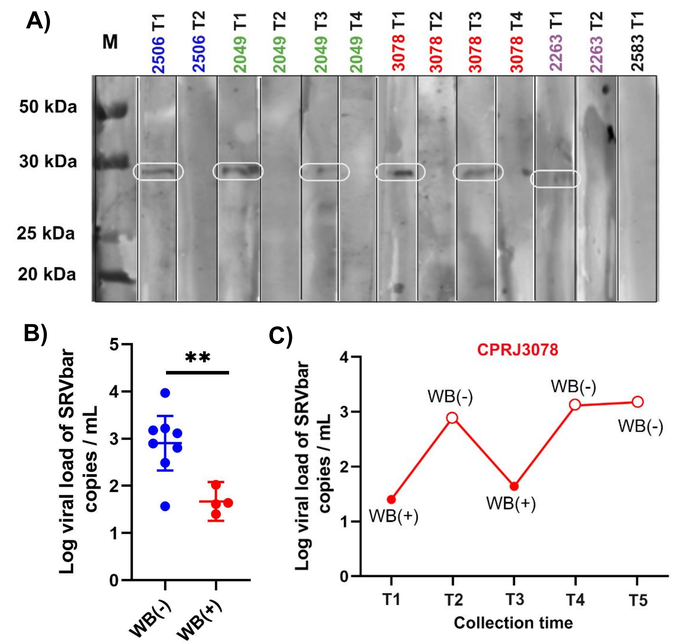


Figure 2. Analysis of the detection of antibodies against p27Gag of SRVbar and correlation with viral load. a) Western Blot for p27Gag of plasma samples collected at different times from muriquis. b) Analysis of the correlation between seropositivity and SRVbar viral load in plasma samples. c) Correlation of the variation in viral load and seropositivity in the CPRJ3078 specimen over time.



CONCLUSIONS

Although the sample size is low, it is possible to conclude that SRV infection in Neotropical primates has similarities to that observed in Asian monkeys, such as the presence of viral load in asymptomatic animals. Blood as main site of replication and consequently source of transmission and detectable antibodies appear to be inversely related to viral load. Except for CPRJ2506, none of the asymptomatic animals progressed to immunodeficiency, even with variation over time in the detection of antibodies and viral loads.

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