

# Real-time RT-PCR for the detection and quantitation of Oropouche virus

Alejandra Rojas, Victoria Stittleburg, Fátima Cardozo, Nathen Bopp, César Cantero, Sanny López, Cynthia Bernal, Laura Mendoza, Patricia Aguilar, Benjamin A. Pinsky, Yvalena Guillén, Malvina Páez, Jesse J. Waggoner

## Abstract

Oropouche virus (OROV) causes an acute, systemic febrile illness, and in certain regions of South America, this represents the second most common human arboviral infection after dengue virus. A new real-time RT-PCR was developed for OROV and reassortant species. The new OROV rRT-PCR proved linear across 6–7 orders of magnitude with a lower limit of 95% detection of 5.6–10.8 copies/QL. Upon testing dilutions of OROV and Iquitos virus reference genomic RNA, all dilutions with >10 copies/QL were detected in both the OROV rRT-PCR and a comparator molecular assay, but the OROV rRT-PCR detected more samples with  $\leq 10$  copies/QL (8/14 vs 0/13, respectively,  $P = 0.002$ ). In a set of 100 acute-phase clinical samples from Paraguay patients with a suspected arboviral illness, no patients tested positive for OROV RNA using either assay. The OROV rRT-PCR provides a sensitive molecular assay for the study of this important yet neglected tropical arboviral infection.