

Tick-borne *Borrelia* and *Rickettsia* in Sri Lankan wildlife: Molecular evidence and phylogenetic insights

Fazil KI¹, Amarakeerthi HKPU¹, Gamage SS², Thilakarathne DS¹

Introduction and Objectives: Ticks are vectors for numerous zoonotic pathogens, including *Borrelia* and *Rickettsia* species, posing significant risks to human health. Wildlife serves as natural reservoirs, contributing to the maintenance and transmission of these pathogens in the environment. This study aimed to molecularly investigate *Borrelia* and *Rickettsia* species in ticks collected from wildlife presented to the Veterinary Teaching Hospital (VTH), University of Peradeniya, Sri Lanka

Methods: Twenty ticks were collected from 20 different hosts out of the 50 wild animals examined at VTH (4 pythons, 4 mouse-deer, 4 barking-deer, 2 land monitors, 2 golden palm civet, 2 spotted deer and 2 wild boar) and preserved in 70% ethanol. Morphological identification was followed by aseptic dissection. DNA was extracted using the QIAGEN DNeasy Tissue Kit. Conventional PCR was conducted targeting the *gltA* gene (381 bp) for the spotted fever group of *Rickettsia* and 16S rRNA gene (674bp) for *Borrelia*. High-purity PCR amplicons, two from *Borrelia* samples and one from a *Rickettsia* isolate were submitted for Sanger sequencing, and the resulting sequences were subjected to nBLAST and phylogenetic analysis.

Results: Out of 50 wild animals examined, 70% were infested with ecto-parasites and 40% carried ticks. Among ticks, 30% were males while 70% were females. Tick genera identified included *Boophilus* (n=4), *Rhipicephalus* (n=2), *Aponomma* (n=7), *Ornithodoros* (n=1), *Haemaphysalis* (n=5), and *Amblyomma* (n=1). *Rickettsia* DNA was detected in 25% (5/20) of the samples, while *Borrelia* DNA was found in 45% (9/20). The local *Borrelia* sequence showed 88% nucleotide identity to *Borrelia turcica* and the *Rickettsia* sequence exhibited 100% homology to *R. montanensis*. In phylogenetic analysis, the local *Borrelia* sequence formed a sister clade with *B. turcica* reported from Germany and Turkey, and the local rickettsia sequence clustered with isolates recorded from Thailand and Japan.

Conclusions: This study provides evidence of the presence of *Borrelia* and *Rickettsia* species in ticks found in wildlife in Sri Lanka. Although the current study only detected species typically associated with wildlife, the presence of zoonotic species within these genera should not be overlooked. These findings highlight the need for comprehensive molecular surveillance that includes diverse wildlife species across various geographical locations to gain a better understanding of tick-borne zoonotic threats in Sri Lanka.

Keywords: *Borrelia*, *Rickettsia*, Ticks, Zoonotic, wildlife

Acknowledgement; URG/2022/67/V

¹ Department of Veterinary Pathobiology, Faculty of Veterinary Medicine and Animal Science, University of Peradeniya, Sri Lanka

² Department of Veterinary Clinical Science, Faculty of Veterinary Medicine and Animal Science, University of Peradeniya, Sri Lanka

Address for correspondence: Dr. Dulari S. Thilakarathne: Department of Veterinary Pathobiology, Faculty of Veterinary Medicine & Animal Science, University of Peradeniya, Sri Lanka ; Telephone:+94778881842

Email: dsamanthikat@gmail.com;  <https://orcid.org/0000-0002-8790-033X>