

PATIENT AND SAMPLE-RELATED FACTORS THAT EFFECT THE SUCCESS OF *IN VITRO* ISOLATION OF *ORIENTIA TSUTSUGAMUSHI*

Rungnapa Luksameetanasan¹, Stuart D Blacksell^{1,2}, Thareerat Kalambaheti¹, Vanaporn Wuthiekanun¹, Wirongrong Chierakul¹, Sunee Chueasuwanchai¹, Apichat Apiwattanaporn³, John Stenos⁴, Stephen Graves⁴, Sharon J Peacock^{1,2} and Nicholas PJ Day^{1,2}

¹Faculty of Tropical Medicine, Mahidol University, Bangkok, Thailand; ²Centre for Tropical Medicine, Nuffield Department of Clinical Medicine, John Radcliffe Hospital, Oxford, United Kingdom; ³Udon Thani Provincial Hospital, Udon Thani, Thailand; ⁴Australian Rickettsial Reference Laboratory, Geelong Hospital, Geelong, Australia

Abstract. *Orientia tsutsugamushi* is the causative agent of scrub typhus infection, a major cause of human disease in rural areas of Southeast Asia. Twenty-six blood samples collected from patients with serologically proven scrub typhus during a six month period were sent to Bangkok (535 km from the clinical site) by road at ambient temperature (average daily temperature range: 27.1-29.1°C) for attempted *in vitro* isolation in Vero cells. *O. tsutsugamushi* was isolated from 12 samples (sensitivity 46.7%) with the time to isolation ranging from 16 to 37 days [median 27 days, inter-quartile range (IQR) 22.5-33.5 days]. Patient factors such as days of fever and *O. tsutsugamushi* IgM antibody titer, transport factors such as transit time, and isolate genotype (Karp and Gilliam/Kawasaki) were assessed to determine their influence on the outcome of *in vitro* isolation. None of the factors significantly influenced the isolation outcome. This study demonstrates that *O. tsutsugamushi* can often be isolated *in vitro* from the blood of scrub typhus patients when transported at ambient tropical temperatures for many days.