Parasites & Vectors

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New vectors in northern Sarawak, Malaysian Borneo, for the zoonotic malaria parasite, *Plasmodium knowlesi*

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Abstract

Background: *Plasmodium knowlesi* is a significant cause of human malaria in Sarawak, Malaysian Borneo. Only one study has been previously undertaken in Sarawak to identify vectors of *P. knowlesi*, where *Anopheles latens* was incriminated as the vector in Kapit, central Sarawak. A study was therefore undertaken to identify malaria vectors in a different location in Sarawak.

Methods: Mosquitoes found landing on humans and resting on leaves over a 5-day period at two sites in the Lawas District of northern Sarawak were collected and identified. DNA samples extracted from salivary glands of *Anopheles* mosquitoes were subjected to nested PCR malaria-detection assays. The small subunit ribosomal RNA (*SSU* rRNA) gene of *Plasmodium* was sequenced, and the internal transcribed spacer 2 (ITS2) and mitochondrial cytochrome *c* oxidase subunit 1 (*cox*1) gene of the mosquitoes were sequenced from the *Plasmodium*-positive samples for phylogenetic analysis.

Results: Totals of 65 anophelines and 127 culicines were collected. By PCR, 6 *An. balabacensis* and 5 *An. donaldi* were found to have single *P. knowlesi* infections while 3 other *An. balabacensis* had either single, double or triple infections with *P. inui*, *P. fieldi*, *P. cynomolgi* and *P. knowlesi*. Phylogenetic analysis of the *Plasmodium SSU* rRNA gene confirmed 3 *An. donaldi* and 3 *An. balabacensis* with single *P. knowlesi* infections, while 3 other *An. balabacensis* had two or more *Plasmodium* species of *P. inui*, *P. knowlesi*, *P. cynomolgi* and some species of *Plasmodium* that could not be conclusively identified. Phylogenies inferred from the ITS2 and/or *cox*1 sequences of *An. balabacensis* and *An. donaldi* indicate that they are genetically indistinguishable from *An. balabacensis* and *An. donaldi*, respectively, found in Sabah, Malaysian Borneo.

Conclusions: Previously *An. latens* was identified as the vector for *P. knowlesi* in Kapit, central Sarawak, Malaysian Borneo, and now *An. balabacensis* and *An. donaldi* have been incriminated as vectors for zoonotic malaria in Lawas, northern Sarawak.

Keywords: Zoonosis, Malaria, *Plasmodium knowlesi*, Vector, *Anopheles balabacensis*, *Anopheles donaldi*

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