Diversity of *Anopheles* species and trophic behavior of putative malaria vectors in two malaria endemic areas of northwestern Thailand

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ABSTRACT: We determined the species diversity, blood-feeding behavior, and host preference of *Anopheles* mosquitoes in two malaria endemic areas of Tak (Mae Sot District) and Mae Hong Son (Sop Moei District) Provinces, located along the Thai border with Myanmar, during a consecutive two-year period. Anopheline mosquitoes were collected using indoor and outdoor human-landing captures and outdoor cow-baited collections. Mosquitoes were initially identified using morphological characters, followed by the appropriate multiplex AS-PCR assay for the identification of sibling species within *Anopheles* (*Cellia*) complexes and groups present. Real-time PCR was performed for parasite-specific detection in mosquitoes (*Plasmodium* spp. and *Wuchereria bancrofti*). A total of 7,129 *Anopheles* females were captured, 3,939 from Mae Sot and 3,190 from Sop Moei, with 58.6% and 37% of all anophelines identified as *An. minimus*, respectively. All three malaria vector complexes were detected in both areas. One species within the Minimus Complex (*An. minimus*) was present along with two related species in the Funestus Group, (*An. aconitus*, *An. varuna*), two species within the Dirus Complex (*An. dirus*, *An. baimaii*), and four species within the Maculatus Group (*An. maculatus*, *An. sawadwongporni*, *An. pseudowillmori*, and *An. dravidicus*). The trophic behavior of *An. minimus*, *An. dirus*, *An. baimaii*, *An. maculatus*, and *An. sawadwongporni* are described herein. The highest *An. minimus* densities were detected from February through April of both years. One specimen of *An. minimus* from Mae Sot was found positive for *Plasmodium vivax*. *Journal of Vector Ecology* 39 (2): 424-436. 2014.

Keyword Index: Anopheles dirus, Anopheles minimus, Anopheles maculatus, species complexes, malaria, Thailand.

INTRODUCTION

After years of intensive, well-organized malaria control activities in Thailand, malaria still remains prevalent in vulnerable areas, especially along the less developed international borders with Myanmar. Approximately 85% of all reported malaria cases in the country occur in this poorly monitored border region, primarily related to transient employment opportunities or occupational activities including agriculture, hunting, and gem mining. Several primary malaria vector species in Thailand are abundant in forested areas along the international border (Chareonviriyaphap et al. 2003, Corbel et al. 2013). Bancroftian filariasis is also endemic along the Thai-Myanmar border but of low prevalence. However, the detection of this parasite is not uncommon among Burmese working in Thailand (Khamboonruang et al. 1987, Jitpakdi et al. 1998, Bhumiratana et al. 2002). Nocturnally subperiodic (NSP) Wuchereria bancrofti is transmitted by a wide variety of mosquito species including anophelines also capable of malaria transmission (Pothikasikorn et al. 2008).

Of the 73 known *Anopheles* species in Thailand, various sibling species in the Minimus and Dirus Complexes and the Maculatus Group have been recognized as the main malaria vectors in the country (Rattanarithikul et al. 2006, Sinka et al. 2011). Molecular techniques based on polymerase chain reaction (PCR) have allowed for precise and reliable

differentiation and identification of the sibling species of medical importance (Manguin et al. 2008, Sinka et al. 2011). Among these 73 species, five to six species, depending on the literature, are incriminated as primary malaria vectors in Thailand (Rattanarithikul et al. 2006), including Anopheles baimaii (previously An. dirus D) (Green et al. 1991) and Anopheles dirus (Rosenberg et al. 1990, Green et al. 1991) of the Dirus Complex, Anopheles minimus (previously An. minimus A) (Rattanarithikul et al. 1996a) of the Minimus Complex, and Anopheles pseudowillmori (Green et al. 1991), An. maculatus and An. sawadwongporni of the Maculatus Group (Saeung 2012). Additionally, An. campestris and An. epiroticus (Sundaicus Complex) have also been incriminated as potential malaria vectors in Thailand (Apiwathnasor et al. 2002). Wuchereria bancrofti develops experimentally in An. minimus and An. maculatus (Pothikasikorn et al. 2008). Natural infections of this parasite have also been found in An. minimus, An. maculatus and An. vagus in Thailand (Pothikasikorn et al. 2008).

While the number of cryptic species has increased, Thailand has been identifying sibling species within the complexes for several decades, yet information on the distribution, ecology and behavior of many of these species remains poor or lacking. This information is quite critical in defining the vector capacity of each species (Takken and Verhulst 2013). Knowledge on mosquito behavior is crucial