

PHYLOGENETIC ANALYSIS OF *AEDES ALBOPICTUS* ISOLATED FROM TAMAN PUNCAK KINRARA, SELANGOR BASED ON THE NADH DEHYDROGENASE 5 SUBUNIT (ND5) GENE

By

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ABSTRACT

PHYLOGENETIC ANALYSIS OF *AEDES ALBOPICTUS* ISOLATED FROM TAMAN PUNCAK KINRARA, SELANGOR BASED ON THE NADH DEHYDROGENASE 5 SUBUNIT (ND5) GENE

Ae. albopictus is a vector that can transmit a variety of viruses such as dengue (DENV), chikungunya (CHIKV) and zika virus (ZIKV). In Malaysia, number of dengue cases reported has increased on a yearly basis. Vector control methods can be enhanced through the understanding of the genetic structure and diversity of the Ae. albopictus population. In particular, the ND5 gene is a protein-coding gene that has been widely used in intraspecific evolutionary studies of anopheline mosquitoes and Ae. albopictus, but no study has reported its use in the local settings, especially in Selangor, Malaysia. Therefore, the aim of this study is to identify the genetic variation and unravel the phylogenetic distribution of Ae. albopictus isolated at Taman Puncak Kinrara (TPK), Selangor. The eggs of Aedes mosquitoes were collected using standardized ovitraps and reared until adulthood in the insectarium. Ten individual adult females of Ae. albopictus were morphologically identified use pictorial keys. DNA was extracted and quantified. The amplified ND5 regions of extracted DNA (450bp) were sequenced and subjected to BLAST revealing 99% of similarities compared to the references ND5 regions. The polymorphic sites and phylogenetic distribution of the local Ae. albopictus were determined using MEGA6 and MrBayes v3.2.4 based on the aligned sequences derived from ClustalX v2.1. Our findings revealed that Ae. albopictus isolated from TPK have low genetic variation and are closely related to each other. Even though TPK 3 and 4 have polymorphic sites and longer branch, they still have close relationship with other samples and countries such as USA, Vietnam, Cambodia, Madagascar, France, Reunion, Lebanon and Thailand as they are grouped together in group 2. This indicates that Ae. albopictus in TPK area has low variation which could be due to several factors such as repetitive use of insecticides, choice of marker, effects of Wolbachia infestations and the environmental adaptability trait of Ae. albopictus. The use of other markers and studies on Wolbachia are warranted to substantiate the findings.

Keywords: *Aedes albopictus,* ND5 gene; sequence alignment; polymorphism; phylogenetic analysis

CHAPTER 1

INTRODUCTION

1.1 Background of study

Aedes albopictus otherwise known as the "Asian Tiger Mosquito" is able to transmit a number of human viral diseases such as dengue fever, chikungunya fever, yellow fever and others (Chen *et al.*, 2015). *Ae. albopictus* is able to affect the human health because of its rapid and aggressive spread, ecological adaptability in different traits, feeding behavior, diapause and vector competence (Paupy, Delatte, Bagny, Corbel, & Fontenille, 2009). The rapid expansion of *Ae. albopictus* can be mainly attributed to human-aided dispersal (Tatem, Hay, & Rogers, 2006) such as international shipping trade of used car tires infested with *Ae. albopictus* eggs and larvae that undergo diapause (Lourenço de Oliveira, Vazeille, de Filippis, & Failloux, 2003) while adults *Ae. albopictus* are passively transported inside the cars and truck cabins (Miller & Loaiza, 2015).

In recent tries, the spread of dengue is a cause of worldwide great concern especially in Asia. The Star2.com online newspaper documented that dengue cases in Asia have increased with an estimated 1.8 billion of people at risk corresponding to 75% of the world's population (Milton, L., 2015, May 24). In Malaysia, up to 19th October 2015, there are 96,222 cases of dengue reported, which increased by 19.4% compared to 2014 (WHO, 2015). The number of dengue cases in 2016 has risen dramatically within only 5 months for 3rd January 2016 to 17th May 2016. A total of 47,438 cases reported with 109 mortality (ARSM & MOSTI, 2016). According to WHO (2016), the number of dengue cases reported for year 2016 was higher than the cases in 2015 for the same period. But the number of death reported for 2016 decreased as compared to the same period of 2015.

Ae. albopictus is difficult to eliminate and eradicate due to its numerous features, necessitating effective control strategies (Bonizzoni, Gasperi, Chen, & James, 2013).