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Molecular typing of Dengue virus circulating in Kuching district of Sarawak, Malaysian Borneo, from 2014 to 2016

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Background: Dengue fever is endemic to Malaysia and the past five years has seen a large increase in recorded dengue cases. All four dengue serotypes have been recorded in Malaysia and the state of Sarawak. Historically for Sarawak, DENV-1 and DENV-2 were first serologically detected in 1962, while DENV-3 and DENV-4 were picked up by PCR and Sanger sequencing in 1997–1999. However, no serotype sequence data for Sarawak has been published in recent years.

Methods and materials: A total of 790 clinically suspected dengue patient serum samples were collected from Borneo Medical Centre (BMC) and Sarawak General Hospital (SGH) from September 2014 to July 2016. Of these, 61 DENV isolates were retrieved from sera cultured in C6/36 cells and confirmed by a published pan-flavi PCR assay followed by sequencing. Complete envelope gene sequence using in-house designed primers followed by Sanger sequencing were obtained for 27 DENV isolates.

Results: BLAST analysis of the 27 complete E gene sequences revealed three DENV serotypes including 10 DENV-1, 10 DENV-2 and 7 DENV-3. All three serotypes were found in each year of the study period. Phylogenetic analyses showed that all Sarawak DENV-1 were grouped within Genotype 1. All Sarawak DENV-2 clustered within the Cosmopolitan genotype. Previously published Sarawak DENV-2 sequences (from 1997 to 2002) were from the same genotype. For DENV-3, circulating strains were predominantly from Genotype 3, with a single isolate detected from Genotype 1 that was similar to published strains observed in 1997. The isolates from all three serotypes were closely related with strains from Peninsular Malaysia, neighboring Southeast Asian countries and China. Clustering of a number of Sarawak isolates are seen in all three DENV serotype phylotrees.

Conclusion: Three serotypes DENV-1, DENV-2 and DENV-3 co-circulate in the Kuching district of Sarawak, with the clustering of some local strains demonstrating internal circulation. The close relationship of Sarawak strains with other Malaysian, Southeast Asian nations and China strains in the same time period, coupled with the emergence of DENV-3 Genotype 3 in Sarawak, also indicates the inter-state and international nature of DENV circulation in Sarawak.

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0900

Molecular epidemiology of Coxsackievirus A6 in Sarawak, Malaysian Borneo, from 2000 to 2015

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Background: Hand, foot and mouth disease (HFMD) affects mostly children with millions of infections notified every year particularly in Asia. In the last decade, Coxsackievirus A6 (CVA-6) has emerged as an important pathogen in HFMD epidemics replacing CVA-16 as a predominant serotype associated with uncomplicated HFMD. In Sarawak, CVA-6 has been detected since 2000. However a comprehensive study on the circulation of this virus has not been carried out to date. In this study, we investigated the molecular epidemiology of CVA-6 in Sarawak from 2000 to 2015 associated with HFMD.

Methods and materials: A total of 106 CVA-6 isolates collected in Sarawak from 2000 to 2015 were investigated in this study. A nested PCR approach was used to generate VP1 sequences from all these isolates. Phylogenetic analysis using a maximum likelihood approach was used to determine the diversity and relatedness of CVA-6 strains circulating in Sarawak.

Results: The phylogenetic analysis of the VP1 gene sequence, revealed that Sarawak CVA-6 viruses were grouped into genotype D following a previously published taxonomy naming convention. All 106 Sarawak isolates were further distributed into subgenotype D1 (isolated from years 2000–2001), D2 (2003,2005) and D3 (2006–2008, 2013, 2015). A Bayesian MCMC analysis estimated the rate of substitution to be 6.17×10^{-3} substitutions/site/year (95% HPD: $5.30–7.01 \times 10^{-3}$).

Conclusion: Data from this study shown that CVA-6 viruses collected in Sarawak from 2000 to 2015 belonged to genotype D which could be further subdivided into subgenotypes D1, D2 and D3. A gradual subgenotype replacement from D1 to D3 in subsequent years since 2000 was observed.

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0904

Plasmodium falciparum and *Plasmodium vivax* epidemiology in an era of malaria elimination

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Background: Dramatic reductions in malaria burden have been achieved in recent years, consequently the epidemiology of malaria in low burden settings is increasingly important. It is widely held dogma that in *Plasmodium vivax* will be harder to eliminate than *Plasmodium falciparum* due to its distinctive characteristics, such as the hypnozoite stage.

Methods and materials: The Malaria Atlas Project incidence and prevalence databases were refined to only include countries with both *P. falciparum* and *P. vivax* malaria, with at least 5 years of data from 2000–2017. Empirical and statistical tests were carried out to assess if *P. vivax* declines faster than *P. falciparum*, there