## **ORIGINAL RESEARCH PAPER**



# A study on *Wolbachia*-dengue-carrying *Aedes* mosquitoes (diptera: culicidae) focuses on the sustainability and frequency of *Wolbachia* in high-rise buildings in Selangor, Malaysia

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# Abstract

The challenge of dengue control due to the unavailability of a specific medication stresses the importance of releasing *Wolbachia*-carrying mosquitoes through vector control programs. This study investigated the sustainability and frequency of *Wolbachia*-dengue-carrying mosquitoes in two dengue hotspot localities in Selangor. A modified sticky ovitrap was used to collect adult mosquitoes in two *Wolbachia*-releasing areas in Selangor, Kelana Puteri and Kelana D'Putera condominiums. All mosquito samples were subjected to PCR using wsp-specific primers for *Wolbachia* detection. Dengue virus was detected using RT-PCR, followed by multiplex-PCR. Out of the 80 *Aedes* spp. collected, *Ae. aegypti* was the most predominant species. More than one-third of *Ae. aegypti* were positive for *Wolbachia*, with 22.9% being superinfected with both *Wolbachia* A and B strains. About 61.4% of the species were uninfected with *Wolbachia*. *Ae. aegypti* carrying the *Wolbachia* A strain was also identified, which has previously never been reported. This strain was similar to the one found naturally in *Ae. albopictus*. None of the *Ae. aegypti* and *Ae. albopictus* were positive for dengue virus. This study could serve as a model for local researchers or health authorities to design and plan an effective field release and monitoring of *Wolbachia*-infected mosquitoes.

Keywords Aedes aegypti · Aedes albopictus · Dengue virus · Modified sticky ovitrap · Wolbachia

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# Introduction

Dengue is a serious global health issue caused by a virus transmitted through mosquitoes. Dengue cases have been increasing over the past few decades, with over 2.4 million cases reported worldwide. There has been a significant increase in dengue cases in Malaysia, with a record of 5.2 million cases in 2019. While the COVID-19 pandemic resulted in a reduction in the number of cases in 2020 and 2021, dengue remains a major health problem in Malaysia, with 43619 cases and 28 deaths reported. Selangor has been the most affected, with 22613 cases and 208 deaths reported between December 2019 and May 2023 (Ministry of Health 2023).

There are four types of dengue virus: DENV-I, DENV-II, DENV-III, and DENV-IV (Rodriguez-Roche and Gould 2013), with infected female *Aedes* mosquitoes being the primary cause of dengue virus transmission to humans. The primary vector responsible for transmitting the virus is *Aedes aegypti*, while *Aedes albopictus* is considered the

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Data availability All data are included in the manuscript.

## **Declarations**

**Conflict of interest** The authors declare that they have no competing interests.

**Consent of publication** All the authors read and approved the final version of the manuscript. All of the people involved in the study gave their consent for its publication.

# References

- Ab Hamid N, Mohd Noor SN, Isa NR, Md Rodzay R, Bachtiar Effendi AM, Hafisool AA, Azman FA, Abdullah SF, Kamarul Zaman MK, Mohd Norsham MI, Amanzuri NH, Abd Khalil N, Zambari IF, Mat Rani AN, Ariffin FD, Omar T, Wasi Ahmad N, Lee HL (2020) Vertical infestation profile of *Aedes* in selected urban highrise residences in Malaysia. Trop Med Infect Dis 5:114. https:// doi.org/10.3390/tropicalmed5030114
- Adams KL, Abernathy DG, Willett BC, Selland EK, Itoe MA, Catteruccia F (2021) *Wolbachia* cifB induces cytoplasmic incompatibility in the malaria mosquito vector. Nat Microbiol 6:1575–1582. https://doi.org/10.1038/s41564-021-00998-6
- Ahmad NA, Vythilingam I, Lim YAL, Zabari NZAM, Lee HL (2017) Detection of Wolbachia in Aedes albopictus and their effects on chikungunya virus. Am J Trop Med Hyg 96:148–156. https://doi. org/10.4269/ajtmh.16-0516
- Ant TH, Herd CS, Geoghegan V, Hoffmann AA, Sinkins P (2018) The Wolbachia strain wAu provides highly efficient virus transmission blocking in Aedes aegypti. PLoS Pathog 14:e1006815. https://doi. org/10.1371/journal.ppat.1006815
- Balaji S, Jayachandran S, Prabagaran SR (2019) Evidence for the natural occurrence of *Wolbachia* in *Aedes aegypti* mosquitoes. FEMS Microbiol Lett 366:fnz055. https://doi.org/10.1093/femsle/fnz055
- Bian G, Joshi D, Dong Y, Lu P, Zhou G, Pan X, Xu Y, Dimopoulos G, Xi Z (2013) Wolbachia invades Anopheles stephensi populations and induces refractoriness to Plasmodium infection. Science 340:748–751. https://doi.org/10.1126/science.1236192
- Brady OJ, Kharisma DD, Wilastonegoro NN, O'Reilly KM, Hendrickx E, Bastos LS, Yakob L, Shepard DS (2020) The cost-effectiveness of controlling dengue in Indonesia using wMel *Wolbachia* released at scale: a modelling study. BMC Med 18:186. https:// doi.org/10.1186/s12916-020-01638-2
- Buchori D, Mawan A, Nurhayati I, Aryati A, Kusnanto H, Hadi UK (2022) Risk assessment on the release of *Wolbachia*-infected *Aedes aegypti* in yogyakarta. Indones inSects 13:924. https://doi. org/10.3390/insects13100924
- Carvajal TM, Hashimoto K, Harnandika RK, Amalin DM, Watanabe K (2019) Detection of *Wolbachia* in field-collected *Aedes* aegypti mosquitoes in metropolitan manila. Philipp Parasit Vectors 12:361. https://doi.org/10.1186/s13071-019-3629-y
- Cheong YL, Nazni WA, Lee HL, NoorAfizah A, Mohd Khairuddin IC, Kamarul GMR, Nizam NMN, Arif MAK, NurZatilAqmar ZM, Irwan SM, Khadijah K, Paid YM, Topek O, Hasnor AH, AbuBakar R, Singh Gill B, Fadzilah K, Tahir A, Sinkins SP, Hoffmann

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AA (2023) Spatial distribution and long-term persistence of *Wolbachia*-infected *Aedes aegypti* in the Mentari Court. Insects, Malaysia. https://doi.org/10.3390/insects14040373

- Chrostek E, Marialva MS, Esteves SS, Weinert LA, Martinez J, Jiggins FM, Teixeira L (2013) *Wolbachia* variants induce differential protection to viruses in *Drosophila melanogaster*: a phenotypic and phylogenomic analysis. PLoS Genet 9:e1003896. https://doi. org/10.1371/journal.pgen.1003896
- Coon KL, Brown MR, Strand MR (2016) Mosquitoes host communities of bacteria that are essential for development but vary greatly between local habitats. Mol Ecol 25:5806–5826. https://doi.org/ 10.1111/mec.13877
- Delatte H, Desvars A, Bouétard A, Bord S, Gimonneau G, Vourc'h G, Fontenille D (2010) Blood-feeding behavior of *Aedes albopictus*, a vector of Chikungunya on La Réunion. Vector Borne Zoonotic Dis 10:249–258. https://doi.org/10.1089/vbz.2009.0026
- Dobson SL, Rattanadechakul W, Marsland EJ (2004) Fitness advantage and cytoplasmic incompatibility in *Wolbachia* single- and superinfected *Aedes albopictus*. Heredity 93:135–142. https://doi.org/ 10.1038/sj.hdy.6800458
- Dorigatti I, McCormack C, Nedjati-Gilani G, Ferguson NM (2018) Using *Wolbachia* for dengue control: insights from modelling. Trends Parasitol 34:102–113. https://doi.org/10.1016/j.pt.2017. 11.002
- Ferguson NM, Kien DT, Clapham H, Aguas R, Trung VT, Chau TN, Popovici J, Ryan PA, O'Neill SL, McGraw EA, Long VT, Dui LT, Nguyen HL, Chau NV, Wills B, Simmons CP (2015) Modeling the impact on virus transmission of *Wolbachia*-mediated blocking of dengue virus infection of *Aedes aegypti*. Sci Transl Med. https://doi.org/10.1126/scitranslmed.3010370
- Harwood RF, James MT (1979) Entomology in human and animal health. MacMillan Publishing, New York
- Huber K, Ba Y, Dia I, Mathiot C, Sall AA, Diallo M (2008) *Aedes aegypti* in senegal: genetic diversity and genetic structure of domestic and sylvatic populations. Am J Trop Med Hyg 79:218–229
- Jayathilake TA, Wickramasinghe MB, de Silva BG (2015) Oviposition and vertical dispersal of *Aedes* mosquitoes in multiple storey buildings in Colombo district, Sri Lanka. J Vector Borne Dis 52:245–251
- Jin S, Fan J, Cao H, Zhang Z, Leng P, Gao Q (2022) Vertical dispersal of *Aedes albopictus* within multi-storey buildings in downtown shanghai. China Parasit Vectors 16:176. https://doi.org/10.1186/ s13071-023-05732-1
- Joanne S, Vythilingam I, Yugavahy N, Leong CS, Wong ML, AbuBakar S (2015) Distribution and dynamics of *Wolbachia* infection in Malaysian *Aedes albopictus*. Acta Trop 148:38–45. https://doi. org/10.1016/j.actatropica.2015.04.003
- Kittayapong P, Baisley KJ, Baimai V, O'Neill SL (2000) Distribution and diversity of *Wolbachia* infections in southeast asian mosquitoes (diptera: culicidae). J Med Entomol 37:340–345. https://doi. org/10.1093/jmedent/37.3.340
- Kittayapong P, Baimai V, O'Neill SL (2002) Field prevalence of Wolbachia in the mosquito vector Aedes albopictus. Am J Trop Med Hyg 66:108–111. https://doi.org/10.4269/ajtmh.2002.66.108
- Lau SM, Vythilingam I, Doss JI, Sekaran SD, Chua TH, Wan Sulaiman WY, Chinna K, Lim YA, Venugopalan B (2015) Surveillance of adult *Aedes* mosquitoes in Selangor, Malaysia. Trop Med Int Health 20:1271–1280. https://doi.org/10.1111/tmi.12555
- Laven H (1951) Crossing experiments with *culex* strains. Evolution 5:370–375
- Lee SF, White VL, Weeks AR, Hoffmann AA, Endersby NM (2012) High-throughput PCR assays to monitor *Wolbachia* infection in the dengue mosquito (*Aedes aegypti*) and *Drosophila simulans*. Appl Environ Microbiol 78:4740–4743. https://doi.org/10.1128/ AEM.00069-12