

LETTER TO THE EDITOR

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Surveillance of arthropod-borne viruses in Benin, West Africa 2020–2021: detection of dengue virus 3 in *Aedes aegypti* (Diptera: Culicidae)

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Dear Editor,

Dengue virus (DENV, family *Flaviviridae*, genus *Flavivirus*) serotypes 1 to 4 (DENV-1, -2, -3, and -4) are responsible for more than 100 million infections per year worldwide. Symptoms of DENV infection can be diverse, reaching from an acute febrile illness to the more severe, sometimes fatal dengue haemorrhagic fever/dengue shock syndrome. After the primary infection, lifelong immunity against a specific serotype is built. However, secondary infections by heterologous serotypes increase the risk of severe disease. First epidemics in Africa date back to the 19th century, and since the 1960s laboratory-confirmed cases and outbreaks have been reported in many countries in sub-Saharan Africa [1]. All four serotypes circulate in Africa with DENV-1 and DENV-2 being described most frequently. Due to the lack of specific diagnostics, DENV infections are often misdiagnosed with diseases that share a similar clinical presentation, e.g., malaria, chikungunya, Zika, yellow fever, or typhoid fever. A valuable, non-invasive tool to detect the circulation of arthropod-borne viruses (arboviruses) is entomological surveillance. Monitoring arboviruses in field-collected mosquitoes can serve as a tool to guide the appropriate control measures prior to arbovirus epidemics. In addition, it allows the investigation of arbovirus evolution or adaptation, as well as the identification of new lineages, and provides data about vectors that are involved in the local arbovirus transmission.

In Africa, data about arbovirus distribution and prevalence mostly come from seroprevalence surveys or single case studies, i.e., travellers returning to countries outside of Africa.

Reported cases are suggested to significantly underestimate the underlying arboviral disease burden. In Benin, a country with a tropical wet-dry climate, the serological evidence of DENV infections in travellers returning from Benin have been described [2]. To gain data about the occurrence of arboviruses, a longitudinal arbovirus surveillance study was conducted.

Mosquitoes were collected from June 2020 until October 2021 at five sites in two bioclimatic regions in Benin (Additional file 1: Fig. S1). Specimens were morphologically identified on chilled tables and samples were tested with pan-*Flavivirus*, pan-*Alphavirus* and pan-*Orthobunyavirus* reverse transcription-polymerase chain reaction (RT-PCR) with primers shown in the Additional file 1: Table S1. The amplicons were sent for sequencing (LGC Genomics, Berlin, Germany) and sequences were analysed with Geneious v9.1.7 (Biomatters, Auckland, New Zealand).

A total of 3749 female mosquitoes, including two genera and four species, were captured during the sampling period (Additional file 1: Table S2). The dominant species was *Aedes aegypti* with 2970 specimens, followed by *Culex quinquefasciatus* with 753 specimens. DENV-3 specific RNA was detected in one sample (pool of 25 *Aedes aegypti*) collected with human landing catch (HLC) on 7th July 2021 in Porto Novo. Phylogenetic analysis revealed that the DENV-3 strain from Benin is grouped into the genotype III clade. The sequence clusters in a distinct clade within a monophyletic Africa clade (Fig. 1). Sequence analysis showed a close relationship to DENV-3 strains from humans in Burkina Faso and Ethiopia with a paired identity at the nucleotide level of 98.8% and 98.5%, respectively.

We report the first detection of DENV in mosquitoes in Benin, giving another indication that this virus is endemically

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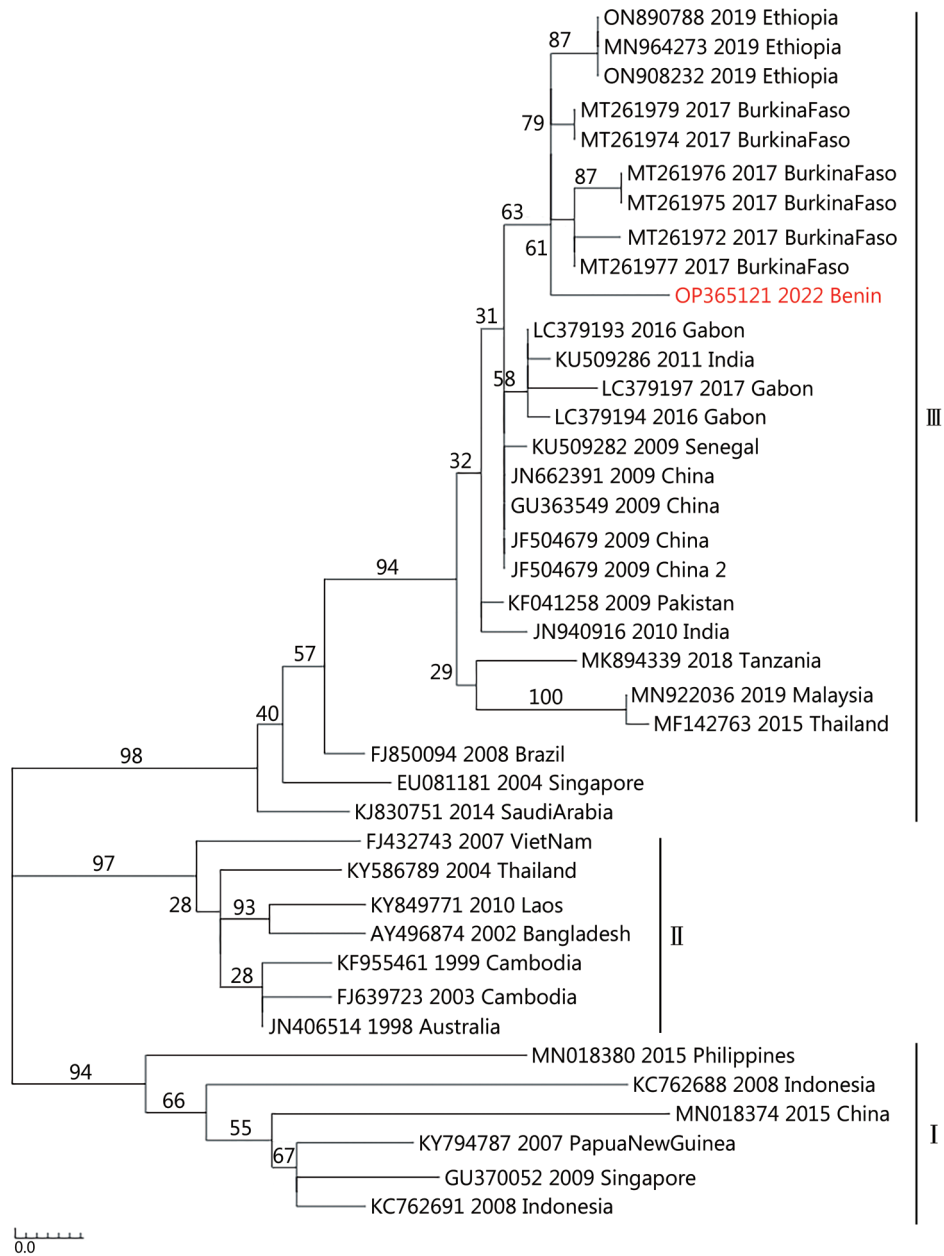


Fig. 1 Phylogenetic tree of detected dengue virus serotype 3.

The genetic analysis is based on a 483-nt sequence alignment comprising the C-prM gene sequences of DENV serotypes and reconstructed using Maximum Likelihood method and a General Time Reversible model implemented by using MEGA3 software (www.megasoftware.net). Values at nodes indicate bootstrap support (1000 replicates). Roman numerals denote the different genotypes of DENV-3. The sequence from Benin was gained using the primers D1 and D2 (Additional file 1: Table S1). The analysis involved 40 nucleotide sequences and GenBank accession numbers are provided for reference genomes. DENV. Dengue virus

occurring in this region. Phylogenetic analysis showed a close relationship to DENV strains from Burkina Faso and Ethiopia. Burkina Faso experienced a greater DENV outbreak in 2016 and 2017. The detection of DENV-3 genotype III strain in Benin and surrounding countries supports the suggestion that there is an ongoing circulation of different DENV strains in the West African countries. Specifically, DENV-3, has become

increasingly common in West and Central Africa in the last decade and has been responsible for several outbreaks in Senegal (DENV-3 accounting for 65% of all gained sequences)[3], Burkina Faso[4] and Gabon[5]. Until now, Benin did not report any outbreak of DENV, but it must be assumed that human infections occur regularly. Due to the restricted availability of diagnostic capacity, most investigations are

performed only during clearly noticeable outbreak periods. Our study highlights that there is a need to implement further investigations and surveillance strategies to prevent and control future outbreaks of mosquito-borne viruses in Western Africa.

Abbreviations

DENV: Dengue virus; HLC: Human landing catches; RNA: Ribonucleic acid; RT-PCR: Reverse transcription polymerase chain reaction.

Supplementary information

The online version contains supplementary material available at <https://doi.org/10.1186/s40779-022-00425-9>.

Additional file 1: Fig. S1 Map of Benin with trapping sites and number of collected mosquitoes. **Table S1** List of the primer sequences used for reverse transcription polymerase chain reaction. **Table S2** Total number of collected female mosquitoes.

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Authors' contributions

The study was conceived and designed by AY, HJ, JSC and RL. Data were collected by CT and GH. The data were analysed by CT, GH, AB and EA. The manuscript was drafted by AY, HJ and EA. The manuscript was critically revised by JSC and RL. All authors approved the final version of the manuscript.

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Availability of data and materials

Not applicable.

Declarations

Ethics approval and consent to participate

Ethical approval for this study was granted by the Ethical Committee of the Ministry of Health in Benin (045/MS/DC/SGM/DNSP/CJ/SA/027SGG2020). Verbal consent was asked to the head of each household for mosquito collection and consent of collectors was obtained prior to HLC. They received anti-malaria prophylaxis and yellow fever immunization. They were organized in teams of two

for each collection point and they rotated between locations within houses every two hours. In case of refusal, permission was sought from the next household.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

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