BRAIN UPDATE

Dengue virus infection and neurological manifestations: an update

Si-Lei Fong,[1,†] Kum-Thong Wong[2,3,†] and Chong-Tin Tan[1,†]

[†]These authors contributed equally to this work.

Dengue virus is a flavivirus transmitted by the mosquitoes, *Aedes aegypti* and *Aedes albopictus*. Dengue infection by all four serotypes (DEN 1 to 4) is endemic globally in regions with tropical and subtropical climates, with an estimated 100–400 million infections annually. Among those hospitalized, the mortality is about 1%. Neurological involvement has been reported to be about 5%. The spectrum of neurological manifestations spans both the peripheral and central nervous systems. These manifestations could possibly be categorized into those directly related to dengue infection, i.e. acute and chronic encephalitis, indirect complications leading to dengue encephalopathy, and post-infectious syndrome due to immune-mediated reactions, and manifestations with uncertain mechanisms, such as acute transverse myelitis, acute cerebellitis and myositis.

The rising trend in global dengue incidence calls for attention to a more explicit definition of each neurological manifestation for more accurate epidemiological data. The actual global burden of dengue infection with neurological manifestation is essential for future planning and execution of strategies, especially in the development of effective antivirals and vaccines against the dengue virus.

In this article, we discuss the recent findings of different spectrums of neurological manifestations in dengue infection and provide an update on antiviral and vaccine development and their challenges.

1 Division of Neurology, Department of Medicine, Faculty of Medicine, University of Malaya, 50603 Federal Territory of Kuala Lumpur, Malaysia
2 Department of Pathology, Faculty of Medicine, University of Malaya, 50603 Federal Territory of Kuala Lumpur, Malaysia
3 Jeffrey Cheah School of Medicine and Health Sciences, Monash University Malaysia, 47500 Subang Jaya, Selangor, Malaysia

Correspondence to: Si-Lei Fong
Neurology Laboratory, Level 6 Menara Selatan
University Malaya Medical Centre, 50603 Kuala Lumpur
E-mail: fongsilei@gmail.com

Keywords: dengue; encephalitis; neurological manifestations

Introduction

Dengue virus (DENV), a single-stranded RNA virus (genus *Flavivirus*, family *Flaviviridae*) is transmitted by the mosquitoes, *Aedes aegypti* and *Aedes albopictus*. The four DENV virus serotypes (DEN 1 to 4), are all capable of causing human infection. Endemic globally, occurring in regions with tropical and subtropical climates, the annual incidence has increased tremendously over the years to 100 million–400 million infections.[1] The population at risk are estimated to increase by up to 4.7 additional billion people by 2070, particularly in urban areas and lowlands in the Western Pacific and the Eastern Mediterranean regions. Climate change with the rise of global mean temperature could increase the transmission susceptibility of DENV by increasing the DENV spatial range and length of transmission seasons, resulting in the expansion of the DENV epidemic belt to temperate areas, including a northward shift to central northern Europe and Northern United States.[2] While about 80% of infected patients were generally asymptomatic

Received June 12, 2023. Revised October 03, 2023. Accepted November 27, 2023. Advance access publication December 11, 2023

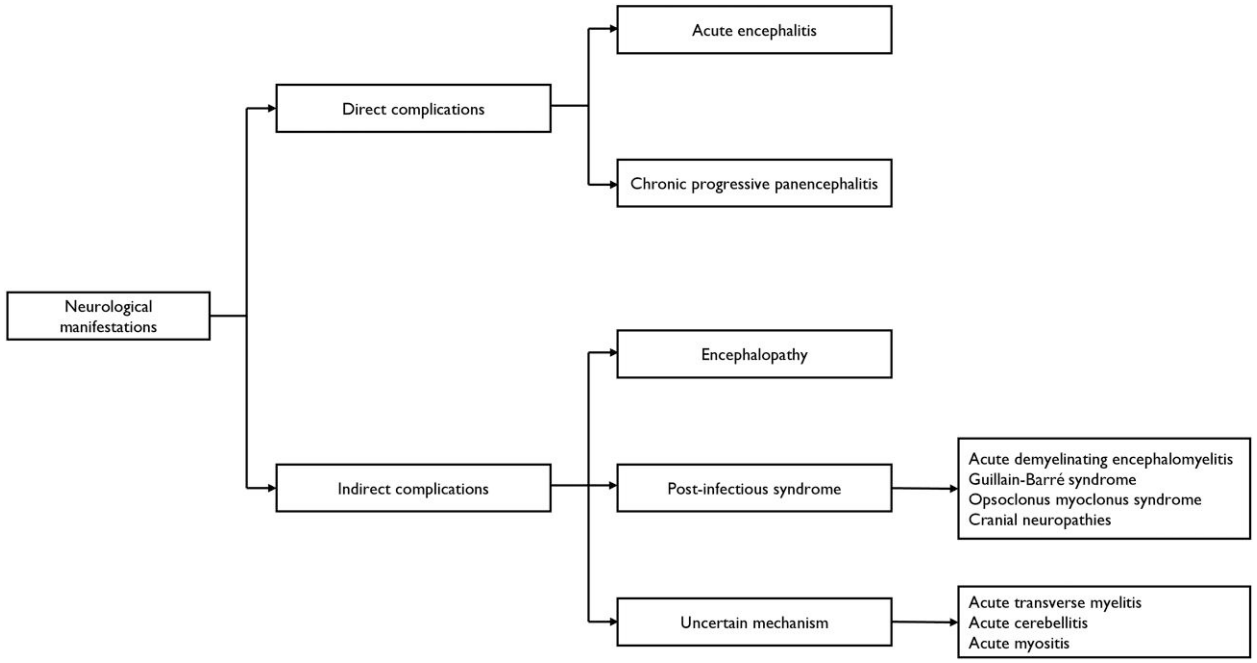

Figure 1 Categorization of neurological manifestations associated with DENV infection.

or only experienced mild symptoms, some may progress into more severe stages. World Health Organization (WHO) categorized DENV infection into two groups: (i) those with milder manifestations either with or without warning signs; (ii) and severe DENV. The first group of symptomatic DENV infection usually manifests with high-grade fever, headache, myalgia, arthralgia, nausea or non-persistent vomiting. Warning signs include persistent vomiting, abdominal pain, mucosal bleeding, lethargy, fluid accumulation, enlarged liver, and increasing haematocrit trend. The second group is the severe, life-threatening DENV infection, characterized by plasma leakage, bleeding and organ impairment (liver, renal or CNS). Impaired consciousness is one of the neurological symptoms categorized under severe DENV infection. However, this symptom is non-specific and could be due to encephalitis or encephalopathy. Primary DENV infection usually results in milder manifestations, while secondary infection with another DENV serotype may increase the risk of severe dengue due to antibody-dependent enhancement.[3,4]

The reported incidence of neurological involvement associated with DENV infection is about 5%,[5,6] and spans both the peripheral nervous system and the CNS. The neurological manifestations can be categorized into those presumed or proven to be directly related to DENV infection, indirect complications leading to DENV encephalopathy, post-infectious syndromes and manifestations with uncertain mechanisms. (Fig. 1) Nonetheless, the neuropathogenesis involved in many cases is presumptive and has yet to be proven. These neurological manifestations and complications associated with DENV infection are expected to increase in tandem with the increasing incidence of DENV worldwide. This worrisome trend calls for attention to a more explicit understanding and definition of each neurological manifestation in dengue infection for more accurate epidemiological data. Knowing the actual global burden of DENV infection with neurological manifestation is essential for future planning and execution of strategies especially in the development of effective antivirals and vaccines against dengue virus. In this update, we discuss the recent findings of different spectrum of neurological manifestations in dengue infection and update the development of antivirals and vaccines.

Acute encephalitis

The operational or clinical diagnosis of acute DENV encephalitis in literature was based on the presence of fever, altered sensorium with evidence of systemic DENV infection, with or without the demonstration of DENV virus (antigen, RNA or virus particle) in the CSF or brain tissue. These features served as the backbone for the definitions and criteria for DENV encephalitis by Soares *et al.*[7] and Carod-Artal *et al.*,[6] after the exclusion of other causes of encephalitis and encephalopathy. We applied more stringent criteria for the diagnosis of acute DENV encephalitis, by accounting for potential anti-Japanese encephalitis (JE) virus antibody cross-reactivity with DENV in endemic regions and strictly using CSF findings as core evidence of encephalitis. We defined definite and probable DENV encephalitis using criteria shown in Table 1.[8] Using these criteria, we found only 42 of 121 publications (34.7%) in the literature could be considered as either 'definite' (30/121; 24.8%) or 'probable' (12/121; 9.9%) DENV encephalitis.[8] Of the 181 cases in these publications, 125 cases (69.1%) were 'definite' and 56 (31.0%) were 'probable' DENV encephalitis based on our criteria. In this systemic review and critique, we found that the well-known serological cross-reactivity between anti-DENV and anti-JE antibodies in serum or CSF was often ignored or overlooked although these two flaviviruses are endemic in the same geographical distribution in the South East Asia and Western Pacific regions.[9] The co-detection rate for CSF anti-DENV and anti-JE IgM may range from 9.0% to 50.0% in patients with encephalitis.[10-13] Three possibilities could account for this finding: serological cross-reactivity, sequential or coinfection by DENV and JE viruses. This indicates that a clinical diagnosis of DENV encephalitis based on antibody detection alone is often fraught. Nonetheless, a higher CSF IgM titre for a particular virus should be considered as the current infection.[11,13,14]

Table 1 Inclusion criteria for definite and probable DENV encephalitis

Definite DENV encephalitis (all items to be satisfied)
(1) Encephalitic symptoms and signs[a]
(2) Detection of DENV in CSF and/or brain tissue by one or more of the following:
NS-1 antigen
RNA by PCR
Virus isolation by culture
Viral antigens by immunohistochemistry
Probable DENV encephalitis (all items to be satisfied)
(1) Encephalitic symptoms and signs
(2) Evidence of anti-DENV IgM[b] in the CSF AND presence of one or more of the following in the serum:
NS-1 antigen
RNA by PCR
Virus isolation by culture
Anti-DENV IgM

JEV = Japanese encephalitis virus.
[a]Altered sensorium or change in behaviour or mental status.
[b]In endemic areas for Japanese encephalitis, concurrent CSF/serum anti-JEV IgM should be tested and titres compared with CSF/serum anti-DENV IgM. The higher IgM titre obtained should be considered as the infecting virus.

While a strong emphasis was made on the CSF evidence of DENV and exclusion of concurrent JE, the diagnosis of DENV encephalitis is often hampered by concurrent thrombocytopenia that may preclude a lumbar puncture or result in blood contamination, leading to false positive results and inaccurate interpretation of positive viral NS1 antigen or viral RNA in the CSF. Since IgM does not cross the blood–brain barrier (BBB), DENV-specific IgM in the CSF could theoretically indicate CNS infection after JE is ruled out. Serum IgM in the acute phase and a 4-fold increase of serum IgG may serve as second-line evidence for recent infection. Clinicians should be cautious in the interpretation of standalone positive serum or CSF anti-DENV IgM or IgG.

The transition from the peak viraemic phase to the beginning of IgM seroconversion at about Day 5 of illness when the low level of viral particles and CSF IgM may escape detection, and result in false negative results. This was consistent with the reported cases of which most positive CSF DENV PCR were taken within 3 to 5 days from the onset of illness. The sensitivity of real-time PCR assays range from 71.7% to 95.7% among different commercial kits,[15] and virus isolate rates range from 71.5% to 84% depending on the DENV serotype.[16]

Brain MRI in both DENV and JE may show thalamic fluid attenuation inversion resolution (FLAIR) hyperintense signals with or without central blooming artefact.[8,17,18] Other radiological findings, such as reversible splenium lesions, cortical and brainstem FLAIR lesions, are non-specific for DENV encephalitis.[8,19]

From the pathological perspective, acute DENV encephalitis, as in other acute viral encephalitides, should be defined by evidence of typical viral encephalitic changes in the CNS, which includes perivascular cuffing and parenchymal infiltration by inflammatory cells, oedema and necrosis. Viral isolation and identification from the CSF or CNS tissues by cell culture and/or PCR, or by demonstration of virus particles, viral antigens and/or viral genome within neuroglial or vascular tissues are desirable to confirm viral aetiology.[20] Unlike the other flaviviral encephalitides such as JE, West Nile encephalitis,[21] tick-borne encephalitis,[22] St. Louis encephalitis,[23] and Murray Valley encephalitis,[24] in which autopsy studies have adequately proven encephalitis and demonstrated the presence of virus mainly in neurons, we think that this has not been adequately shown in DENV encephalitis. Bhoopat *et al.*[25] reported on three autopsies of DENV shock syndrome that apparently showed positive immunoperoxidase staining for DENV in the brain parenchyma and other tissues. Another series of five autopsies was reported sequentially by Chimelli *et al.*[26] and Miagostovich *et al.*[27] showed perivascular lymphocytes and demyelination, and oedema. There was no neuronal necrosis. In one patient, DENV virus antigen by immunohistochemistry was apparently demonstrated in brain tissues.[27] Another case reported by Ramos *et al.*[28] only found leptomeningeal venous inflammation but no neuronal death, neuronophagia or activated microglia. Immunostaining for viral antigens was apparently positive in neuroglial cells and capillary endothelium. In one of the three meningoencephalitis cases attributed to DENV by Jois *et al.*[29] post-mortem revealed focal cerebral oedema, inflammation, haemorrhage and microinfarcts, without any attempt to perform immunohistochemistry for viral antigen. Overall however, we believe that the published illustrations in these papers are of poor quality and not convincing to confirm viral localization in neuroglial tissues.[25-27,29] Other authors have similarly opined that current literature on viral localization in the brain to be conflicting and that the issue of DENV neuroinvasion awaits further resolution.[30] We support the call for enhanced global efforts towards more pathological studies to establish acute DENV encephalitis.

DENV neuroinvasion was investigated in mouse models. The overexpression of pro-inflammatory cytokines such as interleukin-6, interleukin-10, tumour necrosis factor-alpha, interferon-gamma, and MMP-9 *in vitro* infected microglial cells could lead to the disruption of the inter-endothelial tight junctions and to allow DENV entry into the CNS via the BBB.[31-33] Virus internalization via clathrin-mediated or non-classical clathrin-independent endocytosis may also be important.[34] *In vitro* models of the BBB also showed that the viruses which replicated in the brain microvascular endothelial cells, could also induce the downregulation of tight junction proteins and increase the barrier permeability.[35] Despite the growing evidence of DENV neuropathogenesis in animal models, the exact molecular event and receptors involved in the virus entry into CNS in humans are unclear.

Chronic progressive panencephalitis

In 2019, a unique case of a 45-year-old male who had a 5-year history of severe, progressive dementia with extrapyramidal features, was diagnosed with chronic progressive panencephalitis due to DENV infection.[36] The brain MRI showed marked cerebral atrophy.[36] The brain at autopsy in 2016 confirmed what was found earlier in a 2013 biopsy that showed microglial nodules and inflammatory cell infiltration composed of predominantly CD8 cells in the meninges and parenchyma. The viral envelope protein was also found in the cerebral vasculature, neurons and glial cells throughout the brain, including the hippocampus, basal ganglia and cerebellum, while viral RNA was detected in the hippocampus, basal ganglia, and cerebellum. It is of note that the demonstration of DENV in CNS was challenging as the RNA was only detected in the brain tissue but not in CSF. The virus identified strongly suggested DENV-1, genotype V. Interestingly, the patient had travelled repeatedly to regions endemic for DENV, but he had no previous history of acute encephalitis.[36] An outbreak of DENV-1 genotype V occurred in India in 2008, consistent with the onset of the patient's symptoms in 2009.[37,38] DENV persistence in this patient, was not due to immunodeficiency, since there was a robust

immune response with high-titre of neutralizing antibodies and T-cell responses in the tissues found to harbour the virus. Whole exome sequencing did not reveal any reported or potentially pathogenic mutations in genes known to be associated with primary immunodeficiency. West Nile virus[39,40] and Zika virus[41] are both flaviviruses known to persist in human tissues. Herpes simplex virus,[42] herpes zoster virus, cytomegalovirus[43] and henipavirus[44] can persist in the CNS to manifest as relapsing CNS diseases.

Encephalopathy

Encephalopathy refers to a clinical state of altered mental status presenting with either confusion, behavioural changes or other cognitive impairments, with or without brain tissue inflammation.[20] The terms 'encephalitis' and 'encephalopathy' are two clinically similar but pathologically distinct conditions. In the context of DENV infection, encephalopathy could be a result of metabolic derangements secondary to liver or renal failure, hyponatraemia or metabolic acidosis, cerebral hypoperfusion in hypovolaemic shock, intracranial haemorrhage or disseminated intravascular coagulopathy. Many of these complications have long been thought to be caused by antibody-dependent enhancement of DENV infection of macrophages leading to a cytokine storm and DENV vascular permeability syndrome.[3,4] This phenomenon is mainly caused by sub-neutralizing antibodies arising from a previous primary infection by one DENV serotype enhancing a subsequent infection by DENV of a different serotype.

Post-infection syndromes

Similar to other flaviviruses such as the JE virus, Zika virus, West Nile virus and St. Louis encephalitis virus, DENV is associated with post-infection Guillain-Barré syndrome (GBS).[45] The reported duration from onset of DENV infection to GBS ranges from 5 to 15 days.[46] In a large series of DENV-associated GBS, T'an *et al.*[47] found that 20% of the 97 patients with GBS had recent DENV infection as evidenced by the positive serum DENV IgM. These patients were more likely to have diarrhoea, facial palsies, and more severe disease with lower Medical Research Council (MRC) sum score, higher GBS disability scale at nadir and requirement for ventilation.[47] Acute inflammatory demyelinating polyneuropathies were the most common electrodiagnostic feature in these GBS patients,[47] although Fragoso *et al.*[46] found acute motor and sensory axonal neuropathy in all the 10 patients in their study. The mechanism of DENV-associated GBS is most likely to be similar to other GBS in which the molecular mimicry between the GBS-related human proteins and polyprotein of the virus.[48] The treatment was the same as other GBS with no difference in prognosis among patients with recent DENV infection. Opsoclonus myoclonus ataxia syndrome (OMAS), is a rare neurological disorder, characterized by irregular multidirectional chaotic eye movements (opsoclonus), myoclonus, cerebellar ataxia and cognitive impairment. DENV virus was one of the infectious agents reported to be associated with OMAS in adults.[49-51] This manifestation has a benign course and usually resolves spontaneously without immunotherapy.

Acute demyelinating encephalomyelitis (ADEM) is a rare neurological disorder which could occur in 0.4% of DENV-infected patients.[52] A meta-analysis by Kamel *et al.*[52] and recent case reports showed that the onset of neurological symptoms of DENV-associated ADEM ranges from 3 to 19 days (median of 7 days).[52-55] Most of them had fever on the ADEM onset and the majority of the cases had serologically-confirmed DENV status by serum IgM followed by IgG.[52] Some cases also had concurrent positive serum NS-1 antigen at the onset of neurological symptoms.[52,55] The diagnosis of ADEM was made from the brain MRI in these patients which often shows bilateral or diffuse demyelination in centrum semiovale, corona radiata, cerebellar peduncles, subcortical grey matter, cervico-medullary junction and thoracic spinal cord.[53-56]

The prediction models by Kamel *et al.*[52] also found that earlier onset days of neurological manifestations (<9.5 days) and higher fever when presenting ADEM (>38.4°C) were associated with worse outcomes and partial recovery.

However, given the short latency (i.e. median of 7 days) between DENV infection and ADEM-like symptoms, the differentiation between encephalopathy, acute DENV encephalitis and ADEM could be challenging. This is especially true for cases with concurrent positive serum NS-1 antigen which suggest an early viraemic phase. In addition, the MRI brain findings of ADEM maybe non-specific. Therefore, we speculate that the few cases of ADEM reported in the literature may in fact represent part of the clinical manifestation of acute encephalitis.

Cranial neuropathies are rare manifestations of acute DENV infection. There are only a few case reports of isolated oculomotor nerve palsy in the context of DENV encephalitis and rhombencephalitis.[57,58] Cranial neuropathies are more commonly reported as immune-mediated post-infection complications such as optic neuritis, oculomotor nerve palsy, abducens nerve palsy, and unilateral or bilateral lower motor neuron motor facial nerve palsy.[59-63] The evidence of nerve enhancement on brain MRI was only reported by Bhate *et al.*[61] in a case of isolated oculomotor nerve palsy after a DENV infection. A combination of cranial nerve palsies secondary to brainstem stroke (oculomotor and facial nerve palsies) or in the form of mononeuritis multiplex (abducens and common peroneal nerve palsies) have also been reported.[64,65]

Transverse myelitis, cerebellitis and myositis

Badat *et al.*[66] reviewed a total of 25 publications with DENV-associated transverse myelitis and found that this manifestation occurs at the mean age of 33 years, and at 11.7 days after the onset of acute symptoms.[66] The pathogenetic mechanisms for this uncommon neurological complication are uncertain but speculated to include direct viral invasion of the cord,[67] which would account for the cases that occur during the febrile phase with DENV IgM or antigen detected in the CSF. A post-infectious immune response may account for cases with later onset.[68] Steroids were the commonest treatment given to DENV-associated transverse myelitis; however, only 50.8% of patients achieved full recovery.[66]

DENV-associated cerebellitis has been rarely reported in the literature. In a systematic review in 2019, there were only eight case reports/series on DENV-associated cerebellar syndrome.[69] The majority of cases presented with cerebellar ataxia during the febrile phase of DENV infection,[70-73] some in the context of DENV encephalitis with detectable DENV virus via PCR.[72] However, only eight patients in the series by Hegde *et al.*[72] and one patient by Weeratunga *et al.*[73] reported brain MRI FLAIR and T2 hyperintense signals in the vermis, or bilateral cerebellar hemispheres, microhaemorrhages within the parenchymal lesions, with and without the involvement in the thalamus, basal ganglia, brainstem and spinal cord. The pathogenetic mechanisms for this DENV-associated cerebellar syndrome remain unclear. Post-infection immune-mediated response may be another possible mechanism.

Myositis associated with DENV infection could range from subclinical elevation of creatinine phosphokinase (CK), self-limiting myalgia and weakness, and rhabdomyolysis, to life-threatening severe fulminant myositis resulting in severe quadriparesis and respiratory failure.[74-76] DENV-associated myositis commonly occurs during the febrile phase in young male patients between 25 and 33 years of age with serum CK elevated to between 500 and 100 000 IU/l. EMG usually showed polyphasic and normal-to-short duration motor unit potentials without spontaneous activities.[76] Although Warke *et al.*[77] have shown that DENV is capable of infecting human muscle satellite cells *in vitro*, the study also found that infected cells are unable to upregulate MHC-1 expression, thus facilitating possible immune evasion by the virus. Muscle biopsies in the reported series to date only revealed interstitial haemorrhage, inflammatory infiltrates, occasional myonecrosis and myophagocytosis, without demonstration of viral particles in the muscles.[74-76] This suggests that myositis is likely to be related to cytokine-mediated inflammatory reactions. Fortunately, most cases were self-limiting and achieved complete recovery without corticosteroids.[76,78]

Neurological disease without dengue systemic symptoms

Solomon *et al.*[79] reported from an infectious disease referral hospital in Ho Chi Minh City, Vietnam, 378 patients with acute febrile CNS infection,[79] of which 16 (4.2%) were infected by DENV, compared with four (1.4%) of 286 hospital controls. The diagnosis was mainly based on serology, but 10 patients had DENV isolated or detected by PCR, and three had DENV antibodies in CSF. This accounted for 1% of the 1675 patients admitted with suspected DENV infection. There were more patients with neurological manifestations of dengue fever than dengue haemorrhagic fever grade IV. Of the 21 patients described in detail (16 patients from the 1-year study and an additional five patients in subsequent years), 12 patients did not experience characteristic features of DENV on admission. Of the presenting symptoms listed, only six (29%) had petechiae. Coma and convulsions were the most common manifestations.

Tan *et al.*[47] reported 95 patients with GBS presenting to the University Malaya Medical Centre, Kuala Lumpur, Malaysia between 2010 and 2018. The sera of these patients taken at a mean of 16.5 days after the onset of infective illness were examined for DENV IgM. This was compared to 68 controls. Of the 95 patients with GBS, 70 (73.7%) had antecedent infectious symptoms. DENV IgM antibodies were positive in 19 (20%) patients with GBS compared with five (7.4%) control ($P = 0.034$). Based on the clinical characteristics listed in the report, of the 19 DENV-associated GBS patients, 10 (53%) had fever, five (26%) had cough, four (21%) had diarrhoea (none with anti-*Campylobacter jejuni* antibodies); one (5.2%) had headache; and four (21%) did not have any infectious symptoms.

Taken together, neurological diseases may develop in association with DENV infection, even in the absence of systemic DENV symptoms. It has been estimated that three-quarters of the DENV virus infection is asymptomatic. It is thus possible that asymptomatic patients, or those with mild symptoms may also develop DENV-associated neurological diseases.

Antivirals and current status

There are no specific treatments for DENV infection or severe DENV to date. Judicious fluid management, blood product transfusion when needed and management of metabolic complications remained the mainstay of management.[1] The rising global disease burden warrants the development of an effective antiviral. The approaches used in DENV antiviral development were mainly (i) inhibition of the target host attachment factors or cell receptors (host-directed antivirals, HDA); and (ii) inhibition of specific viral components (direct-acting antivirals, DAA), targeting the structural and non-structural proteins.[80,81] To date, there have been 93 studies on HDA against DENV.[82,83] Among the few HDAs [chloroquine, lovastatin, prednisolone, celgosivir and imunosugar UV-4 hydrochloride (UV-4B)] that underwent clinical trials, celgosivir and UV-4B were shown to be safe and well tolerated in phase 1 studies.[84,85] However, the efficacy of celgosivir in viraemia reduction is insignificant,[84] and the follow up study on UV-4B in healthy subjects was terminated.[86] A total of 78 studies on DAA acting on DENV structural protein, E and C proteins; and non-structural (NS) protein NS4A, and NS4B; NS2B/NS3 protease, NS5 RNA-dependent RNA polymerase (RdRP), and NS5 methyltransferase (MTase) published to date.[82,87,88] NS3 and NS5 have been the main targets for the development of DENV antivirals due to their enzymatic and biological function in the viral replication process.[89] Until 2023, balapiravir (RdRP inhibitor) was the only DAA studied in clinical trials. Similar to the outcome in HDA studies, balapiravir did not show effective viraemia reduction, cytokine profile and fever duration improvement. Recently, a new pan-serotype NS3-NS4B inhibitor (JNJ-1802) was found to be safe and well tolerated in a phase 1 clinical trial[88,90] and is currently undergoing a phase 2 trial to determine its efficacy in humans.[91]

Various murine models (e.g. AG 129, C57BL/6, and BALB/c mice) were used to examine the antiviral efficacy *in vivo* as defined by the reduction of peak viraemia.[92,93] One of the challenges faced in the development of an antiviral was the difficulty in reproducing the clinical manifestation of severe DENV infection in murine models.[94] Nonetheless, using AG129 murine models, a few studies adopted changes to enhance the antiviral efficacy, e.g. celgosivir was found to show improved viraemia reduction effect after adjustment of the treatment regime from two to four times a day.[95] Cocktail regimens such as UV-4B and riboflavin,[96] and four types of bioflavonoids[97] have also been studied and showed positive results in their antiviral potential.

Dengue vaccines

The various platforms and approaches for vaccine development and production include using live-attenuated, inactivated adjuvanted DNA and recombinant subunit vaccines. The primary aim is to elicit neutralizing antibodies against all four DENV serotypes to avoid the possibility of producing non-protective, cross-reactive, sub-neutralizing antibodies to any serotype or serotypes that might lead to antibody-dependent enhancement by a subsequent DENV infection.[80,98,99]

There are several DENV vaccines in various stages of development and clinical trials.[100] The earliest vaccine, Dengvaxia® by Sanofi Pasteur, and Qdenga (TAK-003)® by Takeda Pharmaceuticals are the two currently licenced DENV vaccines.[101-103] A comparison of the two vaccines is shown in Table 2. One of the main disadvantages of Dengvaxia® is that it is only available for previously infected individuals as it poses a significant risk of severe infection in seronegative individuals.[102] On the other hand, although Qdenga® is available for both seropositive and negative individuals, the overall efficacy decreased significantly to 44.7% in the third year, suggesting a need for booster doses.[104] Following these two vaccines, there is another vaccine, Butantan-DV, an analogue to the United States National Institute of Health (NIH) TV-003, which is currently in phase III trial by the Butantan Institution in Brazil, NIH, and Merck

Table 2 Currently licenced dengue vaccines (up to August 2023)

	DENGVAXIA®	QDENGA®
Compound	Tetravalent live-attenuated vaccine CYD-TDV (chimeric yellow fever virus–DENV–tetravalent dengue vaccine) Uses the yellow fever YF17D vaccine strain as the backbone	Tetravalent live-attenuated vaccine Recombinant chimeric attenuated vaccine with DEN-1, DEN-3, and DEN-4 components on DEN-2 non-structural protein as the backbone
Pharmaceutical companies	Sanofi Pasteur, France	Takeda Pharmaceuticals, Japan
Approval status	Mexico, Philippines, and Brazil in 2015 El Salvador, Costa Rica, Paraguay, Guatemala, Peru, Indonesia, Thailand and Singapore in 2016 European Commission in 2018 US Food and Drug Administration in 2019	European Commission in 2022 Indonesia regulatory board in 2022
Countries	More than 20 countries in Asia, USA and Latin America	European Union Indonesia
Eligible individuals	Children and adolescents 9–16 years of age with laboratory-confirmed previous DENV infection[a]	European Union: More than 4 years old Indonesia: 6 to 45 years of age
Regime	Two doses (6-monthly)	Three doses (3-monthly)
Serotype protection	Tetravalent (DEN 1–4)	Tetravalent (DEN 1–4)
Efficacy data (general)	Overall efficacy against VCD[b]: 82% Against hospitalization: 79% Against severe dengue: 84%	Overall efficacy against VCD:80.2% Against hospitalization: 90.4% Seronegative individuals at baseline: 74.9% Seropositive individuals at baseline: 82.2% Efficacy against VCD during the third year declined to 44.7% Efficacy against hospitalized VCD was sustained at 70.8%
Efficacy data (serotype-based)	DEN1–2:67% DEN-3: 80% DEN-4 89%	DEN-1: 73.7% DEN-2: 97.7% DEN-3: 62.6% DEN-4: Inconclusive

[a]Risk of severe infection in seronegative individuals.
[b]Virologically-confirmed dengue.

Pharmaceuticals (MSD).[105] The available efficacy data at 2 years showed that the overall efficacy against DEN-1 was 89.5%, and DEN-2 was 69.6%, with higher efficacy among the seropositive compared to seronegative individuals.[106]

The primary aim in DENV vaccine development is to elicit neutralizing antibodies against all 4 DENV serotypes to avoid the possibility of producing non-protective, cross-reactive, sub-neutralising antibodies to any serotype or serotypes that might lead to antibody-dependent enhancement by a subsequent DENV infection.[80,98,99] In addition, the ideal vaccine should offer protection against all serotypes and phenotypes ranging from mild to severe, for all age groups, regardless of their DENV immunity status. However, despite high neutralizing antibody titre to all serotypes in phase III trials, it seems that Dengvaxia® has greater protection against DEN-4 and Qdenga® against DEN-2.[101,107]

Future research directions

This current update indicates a great need to scrutinize the current definition of DENV encephalitis and other neurological manifestations. The current evidence and understanding of encephalitis rely mainly on the presence of viral particles, RNA or envelope proteins in the CSF or brain tissues in biopsy samples. However, more solid evidence is needed to prove true DENV acute encephalitis. Further studies are also needed to allow a greater understanding of the underlying mechanism of transverse myelitis, OMAS, myositis and cranial neuropathies. There is a need to be open to the possibility of dengue aetiology in patients with neurological disorders, even when there is no typical symptoms of DENV infection.

At the same time, while the development of DENV antiviral and vaccines is challenging and existing vaccines may not provide the ideal equal protection of all serotypes in all individuals. There are still unanswered questions on the degree of protection the current existing vaccines and antiviral could offer. Nevertheless, the current licensed vaccines should contribute significantly to reducing the global burden of DENV infection and its complications.

Funding

No funding was received towards this work.

Competing interests

The authors report no competing interests.

References

1. Dengue and severe dengue. World Health Organization. Accessed 30 January, 2023. https://www.who.int/news-room/fact-sheets/detail/dengue-and-severe-dengue
2. Colón-González FJ, Sewe MO, Tompkins AM, et al. Projecting the risk of mosquito-borne diseases in a warmer and more populated world: A multi-model, multi-scenario intercomparison modelling study. *Lancet Planet Health*. 2021;5:e404-e414.
3. Halstead SB. Dengue antibody-dependent enhancement: Knowns and unknowns. *Microbiol Spectr*. 2014;2:1-18.

4. Katzelnick LC, Gresh L, Halloran ME, et al. Antibody-dependent enhancement of severe dengue disease in humans. *Science*. 2017;358:929-932.
5. Cam BV, Fonsmark L, Hue NB, Phuong NT, Poulsen A, Heegaard ED. Prospective case-control study of encephalopathy in children with dengue hemorrhagic fever. *Am J Trop Med Hyg*. 2001;65:848-851.
6. Carod-Artal FJ, Wichmann O, Farrar J, Gascón J. Neurological complications of dengue virus infection. *Lancet Neurol*. 2013; 12:906-919.
7. Soares C, Puccioni-Sohler M. Dengue encephalitis: Suggestion for case definition response. *J Neurol Sci*. 2011;306:165.
8. Fong SL, Tan CT, Wong KT. Dengue virus encephalitis: A systematic review and critique. *Neurol Asia*. 2022;27:909-929.
9. Japanese Encephalitis. World Heath Organization. Accessed 19 February 2023. https://www.who.int/news-room/fact-sheets/detail/japanese-encephalitis
10. Atchareeya A, Panthuyosri N, Anantapreecha S, et al. Cross-reactive IgM responses in patients with dengue or Japanese encephalitis. *J Clin Virol*. 2008;42:75-77.
11. Garg RK, Malhotra HS, Gupta A, Kumar N, Jain A. Concurrent dengue virus and Japanese encephalitis virus infection of the brain: Is it co-infection or co-detection? *Infection*. 2012;40:589-593.
12. Singh KP, Mishra G, Jain P, et al. Co-positivity of anti-dengue virus and anti-Japanese encephalitis virus IgM in endemic area: Co-infection or cross reactivity? *Asian Pac J Trop Med*. 2014;7:124-129.
13. Sivamani K, Dhir V, Singh S, Sharma A. Diagnostic dilemma-dengue or Japanese encephalitis? *Neurol India*. 2017;65:105-107.
14. Horwood PF, Duong V, Laurent D, et al. Aetiology of acute meningoencephalitis in Cambodian children, 2010–2013. *Emerg Microbes Infect*. 2017;6:e35.
15. Najioullah F, Viron F, Césaire R. Evaluation of four commercial real-time RT-PCR kits for the detection of dengue viruses in clinical samples. *Virol J*. 2014;11:164.
16. Jarman RG, Nisalak A, Anderson KB, et al. Factors influencing dengue virus isolation by C6/36 cell culture and mosquito inoculation of nested PCR-positive clinical samples. *Am J Trop Med Hyg*. 2011;84:218-223.
17. Pichl T, Wedderburn CJ, Hoskote C, Turtle L, Bharucha T. A systematic review of brain imaging findings in neurological infection with Japanese encephalitis virus compared with dengue virus. *Int J Infect Dis*. 2022;119:102-110.
18. Lnu P, Sehgal V, Bhalla Sehgal L, Gulati N, Kapila S. The Spectrum of MRI findings in dengue encephalitis. *Cureus*. 2022;14:e29048.
19. Tetsuka S. Reversible lesion in the splenium of the corpus callosum. *Brain Behav*. 2019;9:e01440.
20. Venkatesan A, Tunkel AR, Bloch KC, et al. Case definitions, diagnostic algorithms, and priorities in encephalitis: Consensus statement of the international encephalitis consortium. *Clin Infect Dis*. 2013;57:1114-1128.
21. Hayes EB, Sejvar JJ, Zaki SR, Lanciotti RS, Bode AV, Campbell GL. Virology, pathology, and clinical manifestations of west Nile virus disease. *Emerg Infect Dis*. 2005;11:1174-1179.
22. Gelpi E, Preusser M, Garzuly F, Holzmann H, Heinz FX, Budka H. Visualization of central European tick-borne encephalitis infection in fatal human cases. *J Neuropathol Exp Neurol*. 2005;64: 506-512.
23. Reyes MG, Gardner JJ, Poland JD, Monath TP. St louis encephalitis. Quantitative histologic and immunofluorescent studies. *Arch Neurol*. 1981;38:329-334.
24. Fu TL, Ong KC, Tran YD, McLean CA, Wong KT. Viral neuronotropism is important in the pathogenesis of Murray valley encephalitis. *Neuropathol Appl Neurobiol*. 2016;42:307-310.
25. Bhoopat L, Bhamarapravati N, Attasiri C, et al. Immunohistochemical characterization of a new monoclonal antibody reactive with dengue virus-infected cells in frozen tissue using immunoperoxidase technique. *Asian Pac J Allergy Immunol*. 1996;14:107-113.
26. Chimelli L, Hahn MD, Netto MB, Ramos RG, Dias M, Gray F. Dengue: Neuropathological findings in 5 fatal cases from Brazil. *Clin Neuropathol*. 1990;9:157-162.
27. Miagostovich MP, Ramos RG, Nicol AF, et al. Retrospective study on dengue fatal cases. *Clin Neuropathol*. 1997;16:204-208.
28. Ramos C, Sánchez G, Pando RH, et al. Dengue virus in the brain of a fatal case of hemorrhagic dengue fever. *J Neurovirol*. 1998;4: 465-468.
29. Jois D, Moorchung N, Gupta S, Mutreja D, Patil S. Autopsy in dengue encephalitis: An analysis of three cases. *Neurol India*. 2018;66:1721-1725.
30. Johnson ES, Bilbao JM. *Flaviviruses 2: West Nile, St. Louis encephalitis, Murray valley encephalitis, yellow fever, and dengue*. In: Chretien F, Wong KT, Sharer LR, Keohane K, Gray F, eds. *Infections of the central nervous system*. Wiley; 2020:147-162.
31. Chaturvedi UC, Dhawan R, Khanna M, Mathur A. Breakdown of the blood-brain barrier during dengue virus infection of mice. *J Gen Virol*. 1991;72(Pt 4):859-866.
32. Salomão NG, Rabelo K, Póvoa TF, et al. BALB/c mice infected with DENV-2 strain 66985 by the intravenous route display injury in the central nervous system. *Sci Rep*. 2018;8:9754.
33. Bhatt RS, Kothari ST, Gohil DJ, D'Souza M, Chowdhary AS. Novel evidence of microglial immune response in impairment of dengue infection of CNS. *Immunobiology*. 2015;220:1170-1176.
34. Cruz-Oliveira C, Freire JM, Conceição TM, Higa LM, Castanho MA, Da Poian AT. Receptors and routes of dengue virus entry into the host cells. *FEMS Microbiol Rev*. 2015;39:155-170.
35. Mustafá YM, Meuren LM, Coelho SVA, de Arruda LB. Pathways exploited by flaviviruses to counteract the blood-brain barrier and invade the central nervous system. *Front Microbiol*. 2019;10: 525.
36. Johnson TP, Larman HB, Lee MH, et al. Chronic dengue virus panencephalitis in a patient with progressive dementia with extrapyramidal features. *Ann Neurol*. 2019;86:695-703.
37. Chakravarti A, Kumar A, Matlani M. Displacement of dengue virus type 3 and type 2 by dengue virus type 1 in Delhi during 2008. *Indian J Med Microbiol*. 2010;28:412.
38. Chakravarti A, Chauhan MS, Kumar S, Ashraf A. Genotypic characterization of dengue virus strains circulating during 2007–2009 in New Delhi. *Arch Virol*. 2013;158:571-581.
39. Murray K, Walker C, Herrington E, et al. Persistent infection with west Nile virus years after initial infection. *J Infect Dis*. 2010;201:2-4.
40. Murray KO, Kolodziej S, Ronca SE, et al. Visualization of west Nile virus in urine sediment using electron microscopy and immunogold up to nine years postinfection. *Am J Trop Med Hyg*. 2017;97:1913-1919.
41. Aid M, Abbink P, Larocca RA, et al. Zika virus persistence in the central nervous system and lymph nodes of rhesus monkeys. *Cell*. 2017;169:610-620.e14.
42. Nicoll JA, Maitland NJ, Love S. Autopsy neuropathological findings in 'burnt out' herpes simplex encephalitis and use of the polymerase chain reaction to detect viral DNA. *Neuropathol Appl Neurobiol*. 1991;17:375-382.
43. Adle-Biassette H, Teissier N. *Cytomegalovirus infections of the CNS*. In: Chretien F, Wong KT, Sharer LR, Keohane K, Gray F, eds. *Infections of the central nervous system*. Wiley; 2020:65-76.
44. Tan CT, Goh KJ, Wong KT, et al. Relapsed and late-onset nipah encephalitis. *Ann Neurol*. 2002;51:703-708.

45. Leung C. A lesson learnt from the emergence of Zika virus: What flaviviruses can trigger guillain-barré syndrome? *J Med Virol*. 2020;92:2938-2945.
46. Fragoso YD, Gomes S, Brooks JB, et al. Guillain-Barré syndrome and dengue fever: Report on ten new cases in Brazil. *Arq Neuropsiquiatr*. 2016;74:1039-1040.
47. Tan CY, Razali SNO, Goh KJ, Sam IC, Shahrizaila N. Association of dengue infection and guillain-barré syndrome in Malaysia. *J Neurol Neurosurg Psychiatry*. 2019;90:1298-1300.
48. Yuki N. Infectious origins of, and molecular mimicry in, guillain-barré and fisher syndromes. *Lancet Infect Dis*. 2001;1:29-37.
49. Aniket Patil SA, Shukla S. Opsoclonus-Myoclonus syndrome (OMS) due to dengue meningoencephalitis. *J Clin Diagn Res*. 2019;13:OL01.
50. Tan AH, Linn K, Ramli NM, et al. Opsoclonus-myoclonus-ataxia syndrome associated with dengue virus infection. *Parkinsonism Relat Disord*. 2014;20:1309-1310.
51. Desai SD, Gandhi FR, Vaishnav A. Opsoclonus myoclonus syndrome: A rare manifestation of dengue infection in a child. *J Pediatr Neurosci*. 2018;13:455-458.
52. Kamel MG, Nam NT, Han NHB, et al. Post-dengue acute disseminated encephalomyelitis: A case report and meta-analysis. *PLoS Negl Trop Dis*. 2017;11:e0005715.
53. Farooque U, Pillai B, Karimi S, Cheema AY, Saleem N. A rare case of dengue fever presenting with acute disseminated encephalomyelitis. *Cureus*. 2020;12:e10042.
54. Diallo A, Dembele Y, Michaud C, et al. Acute disseminated encephalomyelitis after dengue. *IDCases*. 2020;21:e00862.
55. Viswanathan S, Botross N, Rusli BN, Riad A. Acute disseminated encephalomyelitis complicating dengue infection with neuroimaging mimicking multiple sclerosis: A report of two cases. *Mult Scler Relat Disord*. 2016;10:112-115.
56. Ladak AA, Javed M, Kamal A. Acute disseminated encephalomyelitis as manifestation of dengue infection in an adult female nonresponsive to high dose steroids (1574). *Neurology*. 2021;96(15 Suppl):1574.
57. Verma R, Bharti K, Mehta M, Bansod A. Rhombencephalitis associated with dengue fever. *J Clin Virol*. 2016;78:99-101.
58. Chakrabarti U, Chaitanya V. Dengue encephalitis with unilateral 3rd nerve palsy. *Int J Sci Res Publ (IJSRP)*. 2019;9:9008.
59. Sardana V, Bhattiprolu RK. Dengue fever with facial palsy: A rare neurological manifestation. *Ann Indian Acad Neurol*. 2019; 22:517-519.
60. Peter S, Malhotra N, Peter P, Sood R. Isolated bell's palsy—An unusual presentation of dengue infection. *Asian Pac J Trop Med*. 2013;6:82-84.
61. Bhate M, Ponnaganti S. Isolated oculomotor neuritis after dengue fever. *J Neuroophthalmol*. 2021;41:e369-e371.
62. Boo YL, Lim SY, Chin PW, Hoo FK. Bilateral optic neuritis with maculopathy: A rare manifestation of dengue fever. *Malays Fam Physician*. 2017;12:32-34.
63. Mazliha M, Boo YL, Chin PW. Isolated unilateral sixth cranial nerve palsy: A rare presentation of dengue fever. *Malays Fam Physician*. 2016;11:25-26.
64. Herath HMM, Hewavithana JS, De Silva CM, Kularathna OAR, Weerasinghe NP. Cerebral vasculitis and lateral rectus palsy – two rare central nervous system complications of dengue fever: Two case reports and review of the literature. *J Med Case Rep*. 2018;12:100.
65. Ho JY, Liew YK, Loh J, Sohil P. Case report: Mononeuritis multiplex in the course of dengue fever. *BMC Infect Dis*. 2020;20:696.
66. Badat N, Abdulhussein D, Oligbu P, Ojubolamo O, Oligbu G. Risk of transverse myelitis following dengue infection: A systematic review of the literature. *Pharmacy*. 2019;7:3.
67. Kunishige M, Mitsui T, Tan BH, et al. Preferential gray matter involvement in dengue myelitis. *Neurology*. 2004;63:1980-1981.
68. An J, Zhou D-S, Kawasaki K, Yasui K. The pathogenesis of spinal cord involvement in dengue virus infection. *Virchows Arch*. 2003;442:472-481.
69. de Holanda AC, Maranhão E, Van Der Linden Ferreira Silva L, Bezerra MER, de Melo ES. Dengue fever presenting as acute cerebellar ataxia: Case report and literature review. *J Neurovirol*. 2022;28:460-463.
70. Khoo CS. Dengue cerebellitis: A case report and literature review. *Am J Case Rep*. 2018;19:864-867.
71. Withana M, Rodrigo C, Chang T, Karunanayake P, Rajapakse S. Dengue fever presenting with acute cerebellitis: A case report. *BMC Res Notes*. 2014;7:125.
72. Hegde V, Aziz Z, Kumar S, et al. Dengue encephalitis with predominant cerebellar involvement: Report of eight cases with MR and CT imaging features. *Eur Radiol*. 2015;25:719-725.
73. Weeratunga PN, Caldera HP, Gooneratne IK, et al. Spontaneously resolving cerebellar syndrome as a sequelae of dengue viral infection: A case series from Sri Lanka. *Pract Neurol*. 2014;14:176-178.
74. Misra UK, Kalita J, Maurya PK, Kumar P, Shankar SK, Mahadevan A. Dengue-associated transient muscle dysfunction: Clinical, electromyography and histopathological changes. *Infection*. 2012;40:125-130.
75. Paliwal VK, Garg RK, Juyal R, et al. Acute dengue virus myositis: A report of seven patients of varying clinical severity including two cases with severe fulminant myositis. *J Neurol Sci*. 2011;300:14-18.
76. Garg R, Malhotra H, Jain A, Malhotra K. Dengue-associated neuromuscular complications. Review article. *Neurol India*. 2015;63:497-516.
77. Warke RV, Becerra A, Zawadzka A, et al. Efficient dengue virus (DENV) infection of human muscle satellite cells upregulates type I interferon response genes and differentially modulates MHC I expression on bystander and DENV-infected cells. *J Gen Virol*. 2008;89(Pt 7):1605-1615.
78. Misra UK, Kalita J, Mani VE, Chauhan PS, Kumar P. Central nervous system and muscle involvement in dengue patients: A study from a tertiary care center. *J Clin Virol*. 2015;72:146-151.
79. Solomon T, Dung NM, Vaughn DW, et al. Neurological manifestations of dengue infection. *Lancet*. 2000;355:1053-1059.
80. Norshidah H, Vignesh R, Lai NS. Updates on dengue vaccine and antiviral: Where are we heading? *Molecules*. 2021;26:6768.
81. Lee MF, Wu YS, Poh CL. Molecular mechanisms of antiviral agents against dengue virus. *Viruses*. 2023;15:705.
82. Troost B, Smit JM. Recent advances in antiviral drug development towards dengue virus. *Curr Opin Virol*. 2020;43:9-21.
83. Piccini LE, Castilla V, Damonte EB. Inhibition of dengue virus infection by trifluoperazine. *Arch Virol*. 2022;167:2203-2212.
84. Low JG, Sung C, Wijaya L, et al. Efficacy and safety of celgosivir in patients with dengue fever (CELADEN): A phase 1b, randomised, double-blind, placebo-controlled, proof-of-concept trial. *Lancet Infect Dis*. 2014;14:706-715.
85. Callahan M, Treston AM, Lin G, et al. Randomized single oral dose phase 1 study of safety, tolerability, and pharmacokinetics of iminosugar UV-4 hydrochloride (UV-4B) in healthy subjects. *PLoS Negl Trop Dis*. 2022;16:e0010636.
86. Emergent BioSolutions. Safety and Pharmacokinetics of UV-4B Solution Administered Orally as Multiple Ascending Doses to Healthy Subjects. ClinicalTrials.gov. identifier: NCT02696291. Updated 11 March 2020. Accessed 28 August 2023. https://clinicaltrials.gov/study/NCT02696291
87. Kaptein SJF, Goethals O, Kiemel D, et al. A pan-serotype dengue virus inhibitor targeting the NS3–NS4B interaction. *Nature*. 2021;598:504-509.

88. Goethals O, Kaptein SJF, Kesteleyn B, et al. Blocking NS3–NS4B interaction inhibits dengue virus in non-human primates. *Nature*. 2023;615:678-686.
89. Obi JO, Gutiérrez-Barbosa H, Chua JV, Deredge DJ. Current trends and limitations in dengue antiviral research. *Trop Med Infect Dis*. 2021;6:180.
90. Ackaert O, Vanhoutte F, Verpoorten N, et al. Safety, tolerability, and pharmacokinetics of JNJ-1802, a pan-serotype dengue direct antiviral small molecule, in a phase 1, double-blind, randomized, dose-escalation study in healthy volunteers. *Clin Infect Dis*. 2023;77:857-865.
91. Johnson&Johnson. Janssen announces novel dengue antiviral demonstrates efficacy in pre-clinical data published in Nature. Published 15 March 2023. Accessed 28 August 2023. https://www.jnj.com/janssen-announces-novel-dengue-antiviral-demonstrates-efficacy-in-pre-clinical-data-published-in-nature
92. Watanabe S, Low JG, Vasudevan SG. Preclinical antiviral testing for dengue virus infection in mouse models and its association with clinical studies. *ACS Infect Dis*. 2018;4:1048-1057.
93. Kayesh MEH, Tsukiyama-Kohara K. Mammalian animal models for dengue virus infection: A recent overview. *Arch Virol*. 2022;167:31-44.
94. Zheng Z, Li M, Liu Z, Jin X, Sun J. Establishment of murine infection models with biological clones of dengue viruses derived from a single clinical viral isolate. *Virol Sin*. 2020;35:626-636.
95. Watanabe S, Chan KW-K, Dow G, Ooi EE, Low JG, Vasudevan SG. Optimizing celgosivir therapy in mouse models of dengue virus infection of serotypes 1 and 2: The search for a window for potential therapeutic efficacy. *Antiviral Res*. 2016;127:10-19.
96. Franco EJ, Pires de Mello CP, Brown AN. Antiviral evaluation of UV-4B and interferon-alpha combination regimens against dengue virus. *Viruses*. 2021;13:771.
97. Zandi K, Teoh B-T, Sam S-S, Wong P-F, Mustafa MR, AbuBakar S. Antiviral activity of four types of bioflavonoid against dengue virus type-2. *Virol J*. 2011;8:560.
98. Torres-Flores JM, Reyes-Sandoval A, Salazar MI. Dengue vaccines: An update. *BioDrugs*. 2022;36:325-336.
99. Prompetchara E, Ketloy C, Thomas SJ, Ruxrungtham K. Dengue vaccine: Global development update. *Asian Pac J Allergy Immunol*. 2020;38:178-185.
100. Palanichamy Kala M, St John AL, Rathore APS. Dengue: Update on clinically relevant therapeutic strategies and vaccines. *Curr Treat Options Infect Dis*. 2023;15:27-52.
101. Capeding MR, Tran NH, Hadinegoro SR, et al. Clinical efficacy and safety of a novel tetravalent dengue vaccine in healthy children in Asia: A phase 3, randomised, observer-masked, placebo-controlled trial. *Lancet*. 2014;384:1358-1365.
102. Sridhar S, Luedtke A, Langevin E, et al. Effect of dengue serostatus on dengue vaccine safety and efficacy. *N Engl J Med*. 2018;379:327-340.
103. Biswal S, Reynales H, Saez-Llorens X, et al. Efficacy of a tetravalent dengue vaccine in healthy children and adolescents. *N Engl J Med*. 2019;381:2009-2019.
104. Rivera L, Biswal S, Sáez-Llorens X, et al. Three-year efficacy and safety of takeda's dengue vaccine candidate (TAK-003). *Clin Infect Dis*. 2021;75:107-117.
105. Kallas EG, Precioso AR, Palacios R, et al. Safety and immunogenicity of the tetravalent, live-attenuated dengue vaccine butantan-DV in adults in Brazil: A two-step, double-blind, randomised placebo-controlled phase 2 trial. *Lancet Infect Dis*. 2020;20:839-850.
106. The Butantan Portal. Butantan's dengue vaccine has 79.6% efficacy, partial results from 2-year follow-up show. Published 16 December 2022. Accessed 28 August, 2023. https://butantan.gov.br/noticias/butantan%27s-dengue-vaccine-has-79.6-efficacy-partial-results-from-2-year-follow-up-show
107. Biswal S, Borja-Tabora C, Martinez Vargas L, et al. Efficacy of a tetravalent dengue vaccine in healthy children aged 4–16 years: A randomised, placebo-controlled, phase 3 trial. *Lancet*. 2020;395:1423-1433.