Status of vaccine research and development of vaccines for dengue

Kirsten S. Vannice a, Anna Durbin b, Joachim Hombach a,∗

a Department of Immunizations, Vaccines and Biologicals, World Health Organization, Avenue Appia 20, Geneva 1211, Switzerland
b Department of International Health, Johns Hopkins Bloomberg School of Public Health, 615N. Wolfe St, Baltimore, MD, 21205, USA

A R T I C L E   I N F O

Article history:
Available online 11 March 2016

Keywords:
Dengue vaccine
Vaccine development
Clinical pipeline

A B S T R A C T

This review on the dengue vaccine pipeline was a solicited article and drafted based on the pre-defined template for PD-VAC.
© 2016 World Health Organization; licensee Elsevier Ltd. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/3.0/).

1. About the disease and pathogen

Dengue virus is a single-stranded RNA virus in the genus Flavivirus, family Flaviridae. There are four distinct serotypes (DENV1–DENV4). They are antigenically diverse and only share about 60–75% identity at the amino acid level [1]. Due to genetic variations leading to changes in viral fitness, virulence, and transmission, serotypes and lineages may manifest different patterns of clinical disease and severity. The mature spherical dengue viral particle contains multiple copies of the three structural proteins (capsid, C, prM, the precursor of membrane, M, protein and envelope, E), as well as a host-derived membrane bilayer and a single copy of a positive-sense, single-stranded RNA genome. Human antibodies raised against the DENV virion are mostly targeted at the E and prM proteins.

The virus is transmitted to humans by infectious bites of Aedes mosquitoes, in particular Aedes aegypti but also Aedes albopictus. These vectors are urban day-biting mosquitoes, such that insecticide treated bednets, which have been very important for malaria control, are ineffective [2]. Infected humans are the main carriers and multipliers of the virus, which then transmit DENV to uninfected mosquitoes for subsequent transmission. The geographic distribution of dengue is determined in large part by the vector [3]. During the past five decades, the incidence of dengue worldwide has increased 30-fold [4]. In 2013 the WHO ranked dengue as the fastest spreading vector-borne viral disease, with an epidemic potential. This expansion is believed to be due to global trade (increased transportation and expansion of the vectors), increased global travel (importations of dengue virus to new areas), and urbanization (multiple transmission opportunities from an infected mosquito), possibly enhanced by global warming [5]. Today, all five WHO regions are affected by dengue, with nearly 4 billion people believed to be at risk of dengue infection. The numbers of dengue cases submitted to WHO are underreported and many cases are misclassified because illness is mild or cannot be differentiated from other viral diseases that manifest high fever [6]. One recent modelling estimate suggests 390 million dengue infections occur globally each year, of which 96 million are clinical, and up to one million considered severe [7]. Dengue control is a major public health priority in disease endemic countries. However, the burden of disease in many regions, particularly Africa, is poorly understood.

In endemic areas, dengue has been traditionally a pediatric disease of children less than 15 years of age. However, in some settings there has been a shift toward older age groups; it has been suggested this is related to changing demographics, including smaller susceptible birth cohorts and a larger immune aging population [8].

Dengue can be diagnosed either by virus isolation, serology (MAC-ELISA, IgG ELISA, NS1 ELISA, and PRNT), or molecular methods (RT-PCR). PCR is considered the gold-standard for dengue diagnosis (80–90% sensitivity and 95% specificity if applied in the adequate time window), as serological tests suffer from cross-reactivity, variable sensitivity by timing of specimen collection, and the need for multiple samples (IgG acute and convalescent samples) [9,10]. Due to limited capacity for PCR around the world, definitive dengue diagnosis is difficult in many settings.
Clinical dengue, in particular during epidemics, puts a significant strain on health care facilities. WHO classifies dengue into two categories, dengue (with or without warning signs) and severe dengue [9]. Dengue without warning signs can still lead to significant patient discomfort and debilitation from high fever, vomiting, myalgia, and joint pain lasting 3–7 days, leading to school absenteeism and loss of work. Because it is difficult to know which dengue cases will become severe, non-severe patients are often admitted to the hospital for monitoring. Severe dengue can be life-threatening due to plasma leakage, fluid accumulation, respiratory distress, severe bleeding, and/or organ impairment. Through improved supportive clinical case management, case fatality rates from severe dengue have decreased from more than 20% to less than 1% [11,12]. Proper maintenance of the patient's body fluid volume is critical to patient success.

The strain on the health system and wider economic consequences of non-severe and severe dengue are significant. The cost of illness includes lost wages and decreased productivity as well as care-seeking and direct medical expenses. Sixty percent of the economic strain is attributable to indirect costs [13]. The global economic burden is not well described, but in the Americas alone it is estimated at $2.1 billion USD each year.

Natural immunity to wild-type infection is not completely understood. Humans infected with one serotype of dengue appear to remain protected for the rest of their life to subsequent symptomatic infection with the infecting serotype (homotypic immunity) [14]. Following a first clinically manifested infection, there is a period of cross-protection (heterotypic immunity) against symptomatic infection with the other three serotypes for approximately two years [15]. As cross-protection wanes, individuals who have only had a primary infection are at an increased risk of severe dengue with a secondary infection of a heterologous serotype [16]. It is commonly believed that this increased risk is due to antibody-dependent enhancement of infection, but other mechanisms may contribute [1]. Following a secondary infection, symptomatic dengue due to a third or fourth infection is rare. Thus it is presumed that a secondary infection reinforces non-type specific immunity that provides additional protection against the remaining serotypes (multitypic immunity) [14]. This phenomenon with wild type infection has been an important consideration for the strategy to develop a vaccine and the necessary follow up in clinical trials [17].

2. Overview of current efforts

2.1. EITHER Vaccines currently available and their limitations OR Biological feasibility for vaccine development

In December, 2015, the first dengue vaccine, Dengvaxia® (CYD-TDV) developed by Sanofi Pasteur, was licensed in Mexico [18]. The vaccine was licensed in individuals 9–45 years living in endemic areas. CYD-TDV has since been licensed by several endemic countries National Regulatory Authorities (NRA).

CYD-TDV is a 3-dose live recombinant tetravalent dengue vaccine administered on a 0/6/12 month schedule. It is based on the YF17D backbone, which is also the basis for the licensed JE vaccine IMOJEV [19]. CYD-TDV includes all three structural proteins, but because of the YF backbone, there are no dengue non-structural proteins included. This vaccine has been evaluated in two large pivotal Phase 3 trials in 5 countries in Asia and 5 countries in Latin America, in participants aged 2–16 across the two trials [20,21]. Pooled vaccine efficacy against symptomatic virologically-confirmed dengue (VCD) of any serotype in the year starting 1 month after the third dose was 59.2% (95%CI 52.3, 65.0) [22]. Vaccine efficacy varied by participant age, serostatus at baseline, severity of dengue disease, and infecting serotype. Vaccine efficacy was higher against serotypes 3 and 4 (71.6% and 76.9%, respectively) than against serotypes 1 and 2 (54.7% and 43.0%, respectively), with the lower confidence bound above zero for all serotypes. Surprisingly, vaccine efficacy was substantially higher among participants who had already been exposed to dengue (pooled VE from immunological subset: 78.2%, 95% CI 65.4, 86.3) compared with participants who were naïve at baseline (pooled VE: 38.1%, 95% CI –3.4, 62.9). Interim results from long-term safety follow up demonstrated an elevated risk of hospitalization and severe dengue among 2–5 year old participants (at vaccination) in the third year after receipt of the first dose (RR = 7.45, 95% CI 1.15, 313.80). This younger age group was thus not included in the initial indication. No safety signals were identified in older age groups.

The mechanism behind the imbalance seen in the youngest age group is not currently understood, although there are a number of hypotheses, including age-specific susceptibility to severe disease, serostatus at baseline, waning immunity, and clustering of cases in the CYD group [23,24]. While differences in risk are associated with age, there may be factors in addition to or highly correlated with age that are important. There is a need to better characterize and assess the potential increased risk of dengue among some vaccinees looking at both characteristics of the vaccine and vaccinees, which will also inform any implications for other vaccine candidates [25]. An optimal pediatric vaccine would need to elicit long-term protection against dengue from all four serotypes in seronegative individuals, and hence should have strong immunological priming capacity against all four DENV serotypes.

In addition, six other candidates are in clinical development using a variety of technological approaches (Table 1). A strong case for the feasibility of developing a dengue vaccine can be made based on the assumed life-long homotypic immunity conferred by natural infection [14]. Due to the theoretical risk of immune enhancement, the dogma has been that a tetravalent vaccine inducing a balanced immune response was needed [26]. The interim results of long-term follow up of CYD-TDV show these concerns to be relevant (though not confirmed), and ongoing/future development efforts will need to have practices in place to closely monitor for changes in risk, including in subgroups, and make all efforts to ensure the safety of trial participants [27].

2.2. General approaches to vaccine development for this disease for low and middle income country markets

Many dengue-endemic countries are middle-high income economies and provide a large market to drive development. Candidates under development are being designed primarily for use in endemic settings, which are predominantly low and middle income countries. For this reason and for easier implementation into immunization programs, there are efforts to minimize the number of doses needed, ideally for single-dose vaccines. One candidate vaccines have also been studied for having a low cost of goods [28]. Given the age distribution of symptomatic dengue, which is quite broad and dependent upon the transmission setting, there is attention both to vaccine use in young children as well as in adults. There is consensus that a vaccine that can be provided in early childhood is needed for those countries in which substantial disease in childhood would require early vaccination. Live attenuated candidates under development have ongoing age de-escalation studies with a target lower bound of 1 or 2 years due to interference with maternally derived antibodies and ADE and are both currently being evaluated as single dose vaccines [29–32].

3. Technical and regulatory assessment

While many dengue vaccine trials are conducted under US Investigational New Drug (IND) supervision, dengue vaccines have
been considered independently by NRAs in endemic countries for clinical trials and registration without reliance on WHO prequalification. WHO has provided support to NRAs that first received the CYD-TDV dengue vaccine dossier to assist informed decision-making for vaccine registration.

In 2007, WHO developed guidelines for the clinical evaluation of dengue vaccine in endemic countries and recommended symptomatic virologically-confirmed dengue as the primary endpoint [17]. Such an end-point requires an active surveillance system that captures all febrile illness to avoid missing detection of mild dengue, which may never present to a hospital. Other secondary endpoints were suggested, such as efficacy against each of the four distinct virus types; efficacy after the first of two or more doses of vaccine; effect on duration of hospitalization for dengue; severity of laboratory-confirmed dengue cases; vaccine efficacy against “possible” or “probable” dengue infection. The 2007 guidelines were later on integrated into WHO’s regulatory guidance [33].

No correlate of protection for dengue has been identified as yet, so large efficacy trials are required at this time [17,34], although some correlation has been described between vaccine-induced neutralizing antibody titers (as measured by PRNT50) and protection from disease for a given serotype [35]. However, data from both clinical trials as well as observational studies suggest that thresholds may be different across vaccines as well as serotypes, and there has not been a clear match between seroconversion as measured by PRNT50 and protection from disease [36,37]. Cellular immunity may also contribute to protection. It has been demonstrated that the majority of CD8+ T cell epitopes are located in the NS proteins and that CD8+ T cells have a role in the protective immune response against dengue [38]. New neutralization and CMI assays are currently being developed and validated.

There are only 3 natural hosts for dengue; humans, non-human primates, and mosquitoes. Nonhuman primates may be infected with dengue but do not manifest clinical symptoms; nonhuman
primate studies have been used to demonstrate protection against viremia [39]. Other animal models are of limited value. Human challenge models, both of infection and disease, are at the early stages and should provide useful data for candidate selection in the future, as well as possibly identifying correlates of protection [40–42]. Recent data from one candidate (TV003, described below) showed 100% protection against viremia, rash, and neutropenia following challenge with an attenuated DENV2 virus 6 months post vaccination [43]. Many candidates are in the process of being evaluated or have plans for evaluation using a human challenge model.

4. Status of vaccine R&D activities

4.1. Clinical pipeline

In addition to CYD-TDV, two other tetravalent live recombinant vaccines, TV003/TV005 and DENVax, have just begun or are close to beginning Phase 3 trials and do contain dengue virus non-structural proteins for at least one serotype [44].

TV003 and TV005 (which are identical except for the dosing level of the dengue 2 component) were developed by the US National Institutes of Health and are based on wild-type strains with genetic mutations to attenuate the virus [44]. Several monovalent candidates were first tested in Phase 1 trials to optimize each of the four vaccine virus strains [45]. Vaccine virus serotypes 1, 3, and 4 are based on complete viruses, while serotype 2 is a recombinant virus based on the serotype 4 vaccine strain with the structural proteins replaced by those of serotype 2. One dose of TV005 elicits serumconversion rates over 90% against each serotype, and 90% of flavivirus-naive recipients mounted a tetravalent response [46]. TV003 or TV005 has been licensed to several manufacturers, including Butantan, VaBiotech, and Merck [47]. Phase 2 studies are underway in Brazil and Thailand, and a Phase 3 trial led by Butantan began in February, 2016, in Brazil [32,47].

TDV (formerly DENVax) is also a live recombinant vaccine, developed by Takeda (originally Inviragen). This tetravalent formulation includes a whole attenuated DEN2 virus and recombinant DEN1, DEN3, and DEN4 using the DEN2 background [48]. There have been a number of ongoing and completed Phase 1 and Phase 2 trials in both endemic and non-endemic settings, evaluating 2 doses of a variety of formulations and routes of administration (including traditional needle-syringe mechanism, a needle-less injector, and a needle-free Pharmajet Injector) [44,49–51]. Two doses administered 90 days apart induced tetravalent seroconversion rates of 60% in dengue-naive subjects [51]. An ongoing study 1800 children in Asia and Latin America evaluating three different dosing schedules (day 0 only, 0 and 3 months, day 0 and 1 year) will help determine the optimal dosing schedule for TDV [29]. A Phase 3 trial is expected to begin soon.

A number of other candidates and approaches have been or are currently under evaluation in Phase 1 trials (Table 1) [44]. These include a tetravalent purified inactivated vaccine (GSK) [52], a tetravalent recombinant subunit vaccine based on the dengue wild-type premembrane and truncated envelope protein (Merck) [53,54], a monovalent plasmid DNA vaccine (US Navy Medical Research Center) [55], and an inactivated vaccine/live attenuated vaccine heterologous prime boost (Walter Reed Army Institute of Research) [56].

4.2. Preclinical pipeline

The preclinical pipeline for dengue vaccines includes both conventional as well as novel technological approaches, including recombinant subunit vaccines, DNA vaccines, VLP vaccines, virus-vectored vaccines, purified inactivated virus vaccines, live attenuated virus vaccines, heterologous prime-boost approaches, and simultaneous administration with two technologically different vaccine candidates (Table 1) [57]. Approximately 20 candidates have been or are in the process of being evaluated in NHP models, with some expected to move soon into the clinic. Some novel approaches include a measles vaccine viral vector (Themis Bioscience) [58] and the Japanese encephalitis SA14-14-2 vaccine viral vector (Beijing Institute) [59]. Fiocruz is exploring simultaneous vaccination with a recombinant YF17D vaccine viral vector together with a DNA vaccine [60].

5. Likelihood for financing

Dengue was not prioritized in the 2013 Gavi Vaccine Investment Strategy (VIS) due to reasons including the limited mortality and DALYs attributable to dengue as compared with other vaccines in the portfolio, in addition to uncertainty about the dengue vaccine pipeline at the time of the review [61]. It is expected to be reviewed again as part of the 2018 VIS. A better understanding of disease burden in Africa as well as better quantification of the economic burden of dengue may elevate dengue as a priority for financing by Gavi and others. It is expected that most dengue-endemic countries would need to fund the vaccine procurement and implementation independently.

6. Conclusions

The dengue vaccine clinical landscape is very dynamic. Despite multiple hurdles to vaccine development, tremendous progress has been made, and there is now a licensed dengue vaccine. There is still a significant research agenda to understand the mode of action of licensed and candidate dengue vaccines, and to ensure that vaccinees are not put at increased risk of dengue at some time period following vaccination. Efforts are also needed to be able to protect younger children through vaccination. Continued efforts in dengue vaccine development are critical to address the growing burden of dengue worldwide. A safe, effective and affordable dengue vaccine would represent a major advance for the control of the disease and could be an important tool for reaching the WHO goal of reducing dengue morbidity by at least 25% and mortality by at least 50% by 2020.

Disclaimer

Kirsten Vannice and Joachim Hombach are staff members of the World Health Organization. The authors alone are responsible for the views expressed in this publication and they do not necessarily represent the decisions or policies of the World Health Organization.

Conflict of interest

Joachim Hombach and Kirsten Vannice: No conflicts of interest.

Anna Durbin: Johns Hopkins University, is funded by the National Institutes of Health to conduct clinical trials of the NIH dengue vaccine under contract HHSN272200900010C.

References


