Title: A Randomized, Observer-Blind, Placebo-Controlled, Phase 3 Trial to Investigate the Immunogenicity and Safety of a Tetravalent Dengue Vaccine Candidate and a Yellow Fever YF-17D Vaccine Administered Concomitantly and Sequentially in Healthy Subjects Aged 18 to 60 years in Non-Endemic Country(ies)

NCT Number: NCT03342898

Protocol Approve Date: 28 November 2018

Certain information within this protocol has been redacted (ie, specific content is masked irreversibly from view with a black/blue bar) to protect either personally identifiable information (PPD) or company confidential information (CCI).

This may include, but is not limited to, redaction of the following:

- Named persons or organizations associated with the study.
- Proprietary information, such as scales or coding systems, which are considered confidential information under prior agreements with license holder.
- Other information as needed to protect confidentiality of Takeda or partners, personal information, or to otherwise protect the integrity of the clinical study.

PROTOCOL

A Randomized, Observer-Blind, Placebo-Controlled, Phase 3 Trial to Investigate the Immunogenicity and Safety of a Tetravalent Dengue Vaccine Candidate and a Yellow Fever YF-17D Vaccine Administered Concomitantly and Sequentially in Healthy Subjects Aged 18 to 60 years in Non-Endemic Country(ies)

Immunogenicity and Safety of TDV Administered with a Yellow Fever Vaccine in Adults

Sponsor:	Takeda Vaccines, Inc. 40 Landsdowne Street, Cambridge, MA 02139, USA
Trial Identifier:	DEN-305
IND Number:	014292 **EudraCT Number:** Not Applicable
Vaccine Name:	• Investigational vaccine: Takeda's tetravalent dengue vaccine candidate (TDV) comprised of a molecularly characterized, attenuated dengue serotype 2 strain (TDV-2), a dengue serotypes 2/1 chimeric strain (TDV-1), a dengue serotypes 2/3 chimeric strain (TDV-3), and a dengue serotypes 2/4 chimeric strain (TDV-4) • Licensed live yellow fever (YF-17D) vaccine • Placebo: saline solution
Date:	28 November 2018
Version:	Version 4.0

1.0 ADMINISTRATIVE INFORMATION

1.1 Contacts

Table 1.a Contact Information

Issue	Contact
Serious adverse event and pregnancy reporting	PPD
Medical Monitor (medical advice on conduct of protocol or compound)	Emergency medical contact information will be provided to the site.
Responsible Medical Officer (carries overall responsibility for the conduct of the trial)	Emergency medical contact information will be provided to the site.

1.2 Approval

REPRESENTATIVES OF TAKEDA

This trial will be conducted with the highest respect for the individual participants in accordance with the requirements of this clinical trial protocol and also in accordance with the following:

- The ethical principles that have their origin in the Declaration of Helsinki.
- International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use E6 Good Clinical Practice: Consolidated Guideline.
- All applicable laws and regulations, including, but not limited to those related to data privacy and clinical trial disclosure.

SIGNATURES

PPD

INVESTIGATOR AGREEMENT

I confirm that I have read and that I understand this protocol, the Investigator's Brochure, and any other product information provided by the Sponsor. I agree to conduct this trial in accordance with the requirements of this protocol and also protect the rights, safety, privacy, and well-being of trial subjects in accordance with the following:

- The ethical principles that have their origin in the Declaration of Helsinki.
- International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use, E6 Good Clinical Practice: Consolidated Guideline.
- All applicable laws and regulations, including, but not limited to those related to data privacy and clinical trial disclosure.
- Regulatory requirements for reporting serious adverse events defined in Section 10.4 of this protocol.
- Terms outlined in the Clinical Trial Site Agreement.
- Appendix A – Responsibilities of the Investigator.

I further authorize that my personal information may be processed and transferred in accordance with the uses contemplated in Appendix B of this protocol.

Signature of Investigator Date

Investigator Name (print or type)

Investigator's Title

Location of Facility (City, State)

Location of Facility (Country)

1.3 Protocol Amendment Summary of Changes

This document describes the changes in reference to the Protocol incorporating Amendment No. 2.

The primary purpose of this amendment is to remove the planned Day 120 (Month 4 [M4]) interim analysis. Minor grammatical and editorial changes are included for clarification purposes only. Full details on changes of text are given in Section 1.3.2.

1.3.1 Amendment History

Date	Amendment Number	Amendment Type	Region
30 May 2017	Initial Protocol	Not applicable	Global
22 January 2018	1	Substantial	Global
28 November 2018	2	Non-substantial	Global

1.3.2 Summary of Changes

Amendment to Protocol Version 3.0, 22 January 2018

Rationale for the Amendment:

The rationale for removing the planned Day 120 (M4) interim analysis is that it is no longer required.

The removal of this interim analysis has no impact on the final statistical analyses planned for this study, the scientific value of the study, the study conduct, or the safety of subjects enrolled.

The administrative trial information has been updated and references list has been checked and updated as needed.

Details of the changes are outlined below. In this section only (ie, not in the protocol body) all new text is shown in bold italics and any redundant text is marked using strikethrough.

Section	Description of change
Title page	Date: ~~22 January~~***28 November*** 2018 Version ~~3.0~~***4.0***
1.1	Serious adverse event and pregnancy reporting PPD
1.1	Medical Monitor (medical advice on conduct of protocol or compound) PPD
1.1	Responsible Medical Officer (carries overall responsibility for the conduct of the trial) PPD
1.2	PPD
2.0; 6.1	***Replaced Figure 2.a and Figure 6.a in Sections 2 and 6.1, respectively, with a higher resolution image.***

Section	Description of change
2.0; 13.1.5	~~An interim analysis on safety and immunogenicity data is planned when all subjects have completed Visit 4 (Day 120 [M4]). This analysis will be performed by a separate group of unblinded statisticians and programmers at a selected independent contract research organization (CRO) who will have access to individual vaccine assignment but will not be involved in subsequent trial conduct. The rest of the personnel involved in the conduct of the trial, including those at Takeda, the CRO, and the trial sites, will remain blinded to individual vaccine assignment until unblinding after trial completion (database lock for data through the Day 360 [Month 12] follow-up visit). No modification to the trial will be made based on the results of this interim analysis. More details regarding the interim analysis will be provided in the SAP.~~ ***No interim analyses are planned.***
6.1	Data collection will be by electronic Case Report Form (eCRF). ~~Refer to Section 13.1.5 for the planned interim analysis.~~

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2.0 TRIAL SUMMARY

Name of Sponsor: Takeda Vaccines, Inc. 40 Landsdowne Street, Cambridge, MA 02139, USA		**Product Name:** Tetravalent Dengue Vaccine Candidate (TDV)
Trial Title: A Randomized, Observer-Blind, Placebo-Controlled Phase 3 Trial to Investigate the Immunogenicity and Safety of a Tetravalent Dengue Vaccine Candidate and a Yellow Fever YF-17D Vaccine Administered Concomitantly and Sequentially in Healthy Subjects Aged 18 to 60 years in Non-Endemic Country(ies)		
IND No.: 014292		**EudraCT No.:** Not Applicable
Trial Identifier: DEN-305	**Phase:** 3	**Trial Blinding Schema:** Observer-Blind

Background and Rationale:

Dengue fever is caused by infection with the wild type dengue virus (DENV), a ribonucleic acid virus that occurs as 4 recognized serotypes, DENV-1, DENV-2, DENV-3, or DENV-4. These dengue viruses are transmitted from human to human by mosquitoes (primarily *Aedes aegypti*). The 4 dengue viruses are endemic in Asia, Central and South America, the Caribbean, the Pacific Islands, and parts of Africa. There are an estimated 390 million dengue infections per year worldwide, which is more than 3 times the previous World Health Organization (WHO) estimate of 50 to 100 million cases. Every year, around 500,000 cases of dengue hemorrhagic fever (DHF) require hospitalization with an estimated death rate of 2.5%, primarily in children. It is estimated that 3.9 billion people are at risk of dengue infection.

Dengue fever is clinically defined as an acute febrile illness with 2 or more of the following manifestations: headache, retro-orbital pain, myalgia, arthralgia, rash, hemorrhagic manifestations, or leucopenia, and occurrence at the same location and time as other confirmed cases of dengue fever. The most severe forms of dengue infection – DHF and dengue shock syndrome (DSS) – are life threatening. Primary infection with any one of the 4 dengue serotypes is thought to result in life-long protection from re-infection by the same serotype, but does not protect against a secondary infection by one of the other 3 dengue serotypes and may lead to an increased risk of severe disease (DHF/DSS).

Treatment of dengue fever is based solely on signs and symptoms, with fluid replacement required for hemorrhagic or shock cases. An antiviral therapy for dengue virus infection is not available. Preventive measures that rely on mosquito control and individual protection are of limited efficacy, complex to implement and questionable in terms of cost-effectiveness. There is a great unmet global public health need for a safe and effective vaccine to reduce the morbidity and mortality associated with dengue disease. Vaccine development has focused on tetravalent vaccines that provide protection against all 4 dengue serotypes simultaneously since all 4 dengue serotypes commonly co-circulate in endemic areas. A first tetravalent dengue vaccine (chimeric yellow fever virus-dengue virus tetravalent dengue vaccine [CYD-TDV]) has been recently approved in some countries in Asia and Latin America. Initial findings showed that vaccine efficacy was different between serotypes and depended on dengue pre-exposure status. Additionally, recent analyses found that people who had not been infected by dengue virus before vaccination had a higher risk of getting severe disease when they were infected with dengue virus after vaccination with CYD-TDV. Hence, there is a continued unmet public health need for safer and more efficacious dengue vaccines.

Takeda's Tetravalent Dengue Vaccine Candidate (TDV) - Background:

Takeda's TDV consists of 1 molecularly characterized, attenuated dengue serotype 2 virus strain and 3 chimeric recombinant dengue virus strains expressing surface antigens corresponding to dengue serotypes 1–4. The dengue serotype 2 strain (TDV-2) is based upon the attenuated laboratory-derived virus, DENV-2 Primary Dog Kidney (PDK)-53, originally isolated at Mahidol University, Bangkok, Thailand. The chimeric, attenuated vaccine strains for dengue serotypes 1, 3 and 4 were engineered by substituting the structural genes, pre-membrane (prM) and envelope (E), of TDV-2 with the prM and E genes of the wild type (DENV) virus strains, DENV-1 16007, DENV-3 16562 or DENV-4 1036 virus, respectively. Thus, TDV is comprised of 4 dengue virus strains: a molecularly characterized, attenuated dengue serotype 2 strain (TDV-2), a dengue serotypes 2/1 chimeric strain (TDV-1), a dengue serotypes 2/3

chimeric strain (TDV-3), and a dengue serotypes 2/4 chimeric strain (TDV-4).

Non-clinical studies carried out in mice and non-human primates have demonstrated an acceptable safety, immunogenicity, and efficacy profile of TDV. Additionally, data from completed phase 1 and phase 2 trials have shown satisfactory reactogenicity, safety and immunogenicity profiles of TDV in adults in non-endemic areas as well as in adults and children in endemic areas in Asia and Latin America. Ongoing and completed phase 2 trials have enabled the selection of a final TDV dose (lyophilized formulation) and a 2-dose vaccination regimen 3 months apart by subcutaneous (SC) injection for use in the pivotal program.

The current Investigator's Brochure contains additional product information and a more detailed review of pre-clinical and clinical trials.

Rationale for the Proposed Trial:

Dengue and yellow fever (YF) viruses belong to the same family of *Flaviviridae* and share antigenic determinants, which may result in cross-reacting antibodies. They are both transmitted between humans by mosquitoes (primarily *Aedes aegypti*), and are endemic in tropical areas of Africa and Latin America with a high public health impact. The YF-17D vaccine which is based on a live, attenuated viral strain, is the only commercially available YF vaccine administered as a single SC injection. The YF-17D vaccine is highly effective (approaching 100%) and generally safe with the exception of very rare cases of vaccine-associated neurotropic and viscerotropic disease. Vaccination against YF is required for travelers to certain countries in accordance with the International Health Regulations, and is also recommended by the WHO for all subjects travelling to areas where there is evidence of persistent or periodic YF virus transmission.

The main purpose of this trial is to assess the immunogenicity and safety of the concomitant administration of YF-17D vaccine and TDV in healthy subjects aged 18 to 60 years living in country(ies) non-endemic for both dengue and YF. Because broader and enhanced immune responses following vaccination with a dengue vaccine have been reported in YF-immune subjects compared to YF non-immune subjects, a secondary purpose of this trial is to assess the immunogenicity and safety of the sequential administration of YF-17D vaccine and the 2-dose regimen of TDV. To maintain the trial blind, a placebo (0.9% sodium chloride [NaCl] solution) will be used. Approximately 900 subjects are planned to be enrolled in this trial.

This trial will be conducted in accordance with the protocol, the International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use and Good Clinical Practice (ICH-GCP) Guidelines and applicable regulatory requirements.

Trial Design:

This is a phase 3, observer-blind, randomized, multi-center trial in 900 healthy adults aged 18 to 60 years in non-endemic areas for dengue and YF to investigate the immunogenicity and safety of the concomitant and sequential administration of TDV and YF-17D vaccine. Subjects will be randomized equally (1:1:1 ratio) to one of the following 3 trial groups (300 subjects per trial group):

- Group 1: YF-17D vaccine+placebo concomitantly administered on Day 1 (Month 0 [M0]), first dose of TDV administered on Day 90 (Month 3 [M3]), and second dose of TDV administered on Day 180 (Month 6 [M6]).
- Group 2: first dose of TDV+placebo concomitantly administered on Day 1 (M0), second dose of TDV administered on Day 90 (M3), and YF-17D vaccine administered on Day 180 (M6).
- Group 3: first dose of TDV+YF-17D vaccine concomitantly administered on Day 1 (M0), second dose of TDV administered on Day 90 (M3), and placebo administered on Day 180 (M6).

Concomitantly administered vaccines will be injected into opposite arms. All subjects will be followed-up for 6 months after the third vaccination (administered approximately 6 months after the first vaccination), so the trial duration will be approximately 360 days (12 months) for each subject.

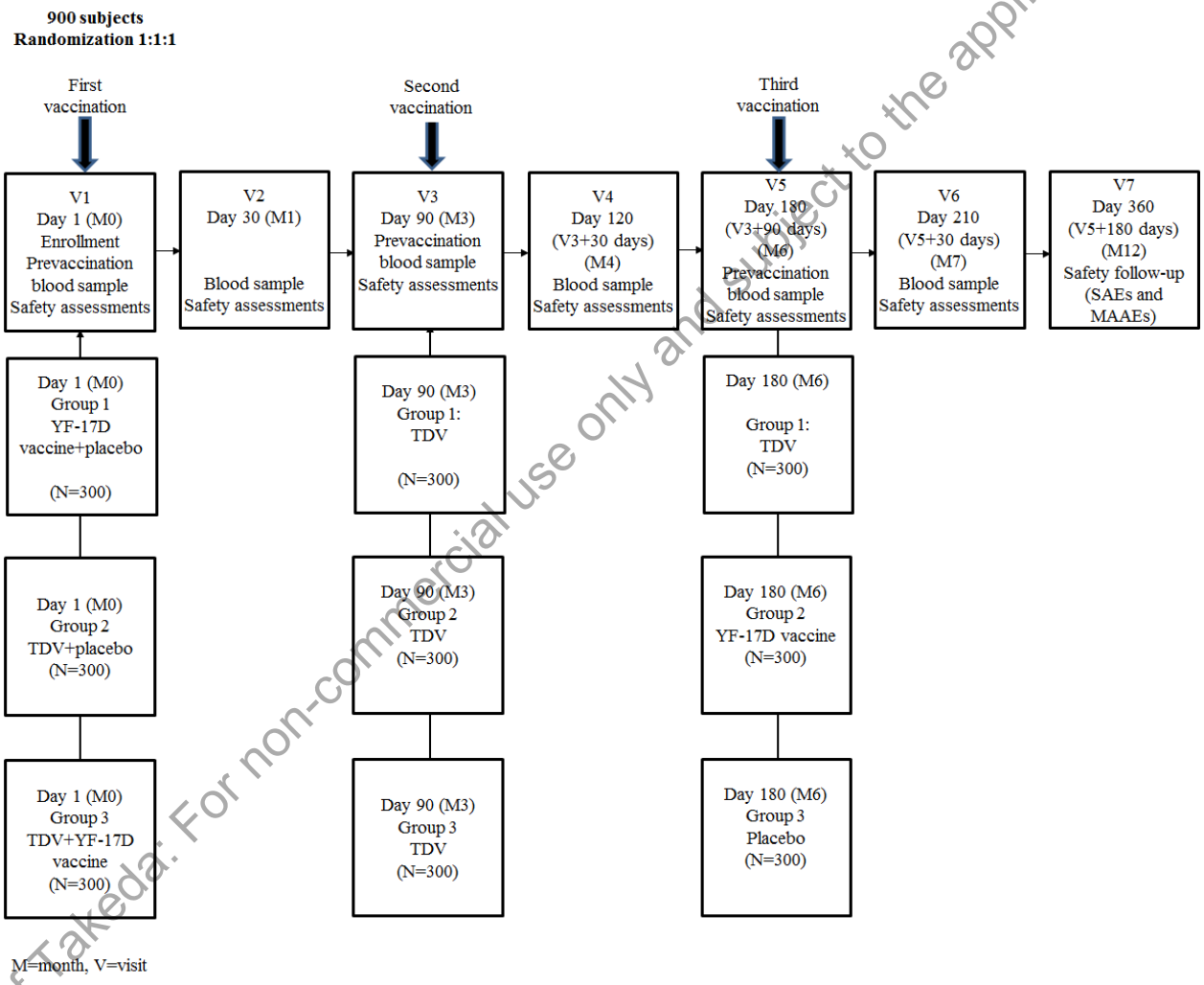
A trial population of 900 subjects (300 subjects per trial group) is considered sufficient for the non-inferiority (NI) assessment of the immune response to the YF-17D vaccine when concomitantly administered with TDV compared to placebo, and for the NI assessment of the immune response to TDV when TDV is concomitantly administered with

YF-17D vaccine compared to placebo.

Safety parameters include solicited local (injection site) and solicited systemic adverse events (AE) for 7 days (day of vaccination+6 days) and 14 days (day of vaccination+13 days) after each vaccination, respectively, unsolicited AEs for 28 days (day of vaccination+27 days) after each vaccination, medically attended AEs (MAAE) and serious adverse events (SAE) throughout the trial.

The schematic of the trial design is included as Figure 2.a.

Figure 2.a Schematic of Trial Design



Immunogenicity evaluation:

Blood samples for the measurement of dengue neutralizing antibodies (microneutralization test 50% [MNT_{50}]) will be collected at pre-first vaccination (Day 1 [M0]), 1 month post first vaccination (Day 30 [Month 1; M1]), pre-second vaccination (Day 90 [M3]), 1 month post second vaccination (Day 120 [Month 4; M4]), pre-third vaccination (Day 180 [M6]), and 1 month post third vaccination (Day 210 [Month 7; M7]).

Blood samples for the measurement of YF neutralizing antibodies (plaque reduction neutralization test [PRNT]) will be collected at pre-first vaccination (Day 1 [M0]), 1 month post first vaccination (Day 30 [M1]), pre-third vaccination

(Day 180 [M6]), and 1 month post third vaccination (Day 210 [M7]).

Safety evaluation:

- Diary cards (paper or electronic) will be distributed to all subjects for the recording of:
  - Solicited local AEs for 7 days following each vaccination (day of vaccination+6 days). These will include: injection site pain, injection site erythema, and injection site swelling, and will be collected at each injection site.
  - Solicited systemic AEs for 14 days following each vaccination (day of vaccination+13 days). These will include: fever, headache, asthenia, malaise, and myalgia.
- Unsolicited AEs for 28 days following each vaccination (day of vaccination+27 days).
- SAEs and MAAEs will be recorded for the trial duration. MAAEs are defined as AEs leading to an unscheduled visit to or by a healthcare professional including visits to an emergency department, but not fulfilling seriousness criteria.

Data collection will be by electronic Case Report Form.

Primary Objective:

- To demonstrate NI of the YF seroprotection rate response to 1 dose of YF-17D vaccine, 1 month following concomitant administration with 1 dose of TDV compared to placebo.

Secondary Objectives:

Immunogenicity

- To demonstrate NI of the geometric mean titer (GMT) response to TDV for all 4 dengue serotypes, 1 month post second dose of TDV following concomitant administration of the first dose of TDV with YF-17D vaccine or placebo.
- To demonstrate NI of the GMT response to 1 dose of YF-17D vaccine, 1 month following concomitant administration with 1 dose of TDV compared to placebo.
- To describe the seropositivity rates for all 4 dengue serotypes, 1 month post second dose of TDV following concomitant administration of the first dose of TDV with YF-17D vaccine or placebo.
- To describe the GMTs of neutralizing antibodies and the seropositivity rates for all 4 dengue serotypes, 1 month post first dose of TDV administered concomitantly with YF-17D vaccine or placebo.
- To describe the GMT and seroprotection rate response to YF-17D vaccine, 1 month after sequential administration of a 2-dose regimen of TDV at 0 and 3 months, followed by YF-17D vaccine 3 months later.
- To describe the GMTs of neutralizing antibodies and the seropositivity rates for all 4 dengue serotypes, 1 month after sequential administration of YF-17D vaccine at month 0, followed by a 2-dose regimen of TDV 3 months later at 3 and 6 months.

Safety

- To assess the safety profile after each vaccine injection in Groups 1, 2 and 3.

Subject Population:

Healthy Subjects: Yes

Planned Minimum Age: 18 years

Planned Maximum Age: 60 years

Planned Number of Subjects: 900

Planned Number of Groups: Three groups in a 1:1:1 ratio (300 subjects in each of Groups 1, 2, and 3)

Criteria for Inclusion:

1. The subject is aged 18 to 60 years, inclusive.
2. Individuals who are in good health at the time of entry into the trial as determined by medical history, physical

examination (including vital signs), and the clinical judgment of the Investigator.

3. The subject signs and dates a written informed consent form and any required privacy authorization prior to the initiation of any trial procedures, and after the nature of the trial has been explained according to local regulatory requirements.
4. Individuals who can comply with trial procedures and are available for the duration of follow-up.

Criteria for Exclusion:

1. Individuals with an elevated oral temperature ≥ 38°C (100.4°F) within 3 days of the intended date of vaccination (consider whether applicable as an exclusion criterion or criterion for delay, see below).
2. Known hypersensitivity or allergy to any of the trial vaccine components (including excipients of the investigational vaccines and placebo).
3. Individuals with contraindications, warnings and/or precautions to vaccination with the YF-17D vaccine as specified within the product information (especially history of thymus dysfunction).
4. Individuals with behavioral or cognitive impairment or psychiatric disease that, in the opinion of the Investigator, may interfere with the subject's ability to participate in the trial.
5. Individuals with any history of progressive or severe neurologic disorder, seizure disorder or neuro-inflammatory disease (e.g., Guillain-Barré syndrome).
6. Individuals with a history of or any current illness that, in the opinion of the Investigator, might interfere with the results of the trial or pose additional risk to the subjects due to participation in the trial.
7. Known or suspected impairment/alteration of immune function, including:
   a) Chronic use of oral steroids (equivalent to 20 mg/day prednisone ≥12 weeks/≥2 mg/kg body weight/day prednisone ≥2 weeks) within 60 days prior to Day 1 (M0) (use of inhaled, intranasal, or topical corticosteroids is allowed).
   b) Receipt of parenteral steroids (equivalent to 20 mg/day prednisone ≥12 weeks/≥ 2 mg/kg body weight/day prednisone ≥2 weeks) within 60 days prior to Day 1 (M0).
   c) Administration of immunoglobulins and/or any blood products within the 3 months prior to Day 1 (M0) or planned administration during the trial.
   d) Receipt of immunostimulants within 60 days prior to Day 1 (M0).
   e) Immunosuppressive therapy such as anti-cancer chemotherapy or radiation therapy within 6 months prior to Day 1 (M0).
   f) Human immunodeficiency virus (HIV) infection or HIV-related disease.
   g) Hepatitis C virus infection.
   h) Genetic immunodeficiency.
8. Abnormalities of splenic or thymic function.
9. Individuals with a known bleeding diathesis, or any condition that may be associated with a prolonged bleeding time.
10. Individuals with any serious chronic or progressive disease according to the judgment of the Investigator (e.g., neoplasm, insulin dependent diabetes, cardiac, renal or hepatic disease).
11. Individuals with body mass index (BMI) greater than or equal to 35 kg/m^2 (=weight in kg/[height in meters^2]).
12. Individuals or their first degree relatives involved in the trial conduct.
13. Intent to travel to dengue or YF endemic countries during the trial period.
14. Individuals with history of substance or alcohol abuse within the past 2 years.

15. Female subjects who are pregnant or breastfeeding.
16. Any positive or indeterminate pregnancy test.
17. Females of childbearing potential[1] who are sexually active and who have not used any of the acceptable contraceptive methods[2] for at least 2 months prior to Day 1 (M0).
18. Females of childbearing potential who are sexually active and who refuse to use an acceptable contraceptive method up to 6 weeks post third vaccination on Day 180 (M6). In addition, they must be advised not to donate ova during this period.
19. Individuals participating in any clinical trial with another investigational product 30 days prior to Day 1 (M0) or intent to participate in another clinical trial at any time during the conduct of this trial.
20. Individuals who received any other vaccines within 14 days (for inactivated vaccines) or 28 days (for live vaccines) prior to enrollment in this trial or who are planning to receive any non-trial vaccine within 28 days of trial vaccine administration.
21. Previous and planned vaccination (during the trial conduct), against any flavivirus including dengue, YF, Japanese encephalitis (JE) or tick-borne encephalitis viruses.
22. Previous participation in any clinical trial of a dengue or other flavivirus (e.g., West Nile [WN] virus) candidate vaccine, except for subjects who received placebo in those trials.
23. Subjects with a current or previous infection with a flavivirus such as dengue, Zika, YF, JE, WN fever, or Saint Louis encephalitis viruses and subjects with a history of prolonged (≥1 year) habitation in a dengue endemic area.

There may be instances when individuals meet all entry criteria except one that relates to transient clinical circumstances (e.g., body temperature elevation or recent use of excluded medication or vaccine). Under these circumstances, eligibility for trial enrollment may be considered if the appropriate window for delay has passed, inclusion/exclusion criteria have been rechecked, and if the subject is confirmed to be eligible.

Criteria for delay of second or third vaccination at Day 90 (M3) and Day 180 (M6):

After enrollment, subjects may encounter clinical circumstances that warrant a delay in the administration of the trial vaccination. These situations are listed below. In the event that a subject meets a criterion for delay of vaccination, the subject may receive trial vaccination once the window for delay has passed as long as the subject is otherwise eligible for trial participation. The decision to vaccinate in those situations will be made by the Investigator.

1. Body temperature ≥38.0°C (≥100.4°F) within 3 days of intended trial vaccination, and/or use of antipyretics and/or analgesic medication for either reason within 24 hours prior to vaccine administration.
2. Receipt of any vaccine other than the trial vaccines within 14 days (for inactivated vaccines) or 28 days (for live vaccines) of intended trial vaccination.
3. Known or suspected altered or impaired immune function as specified under the exclusion criteria.

Contraindications to vaccination at Day 90 (M3) and Day 180 (M6):

[1] Defined as status post onset of menarche and not meeting any of the following conditions: bilateral tubal ligation (at least 1 year previously), bilateral oophorectomy (at least 1 year previously) or hysterectomy.
[2] One or more of the following: hormonal contraceptive (such as oral, injection, transdermal patch, implant, cervical ring), barrier method (condom with spermicide or diaphragm with spermicide) each and every time during intercourse; intrauterine device, monogamous relationship with vasectomized partner (partner must have been vasectomized for at least 6 months prior to Day 1 [M0]). Other contraceptive methods may be considered in agreement with the Sponsor and will be approved by the appropriate ethics committee.

There are also circumstances under which receipt of the vaccination at Day 90 (M3) and Day 180 (M6) is contraindicated in this trial. These circumstances include but are not limited to anaphylaxis or severe hypersensitivity reactions following the administration of first or second trial vaccination at Day 1 (M0) or Day 90 (M3), respectively. If these reactions occur, the subject must not receive the trial vaccination at Day 90 (M3) or Day 180 (M6), respectively, but is encouraged to continue trial participation for safety follow up.

Trial Vaccines

TDV

The investigational vaccine, TDV, is a tetravalent dengue vaccine candidate comprised of 1 molecularly characterized, attenuated dengue virus strain, and 3 chimeric dengue virus strains with CCI [redacted] TDV-1, TDV-2, TDV-3, and TDV-4, respectively. TDV is a lyophilized vaccine that will be reconstituted in diluent (37 mM NaCl solution) prior to administration.

Licensed YF-17D vaccine

The licensed YF-17D vaccine (Sanofi Pasteur) is a live attenuated YF vaccine containing not less than 4.74 $\log_{10}$ plaque forming units or 1000 IU per dose of the 17D-204 strain of the YF virus. YF-17D vaccine is lyophilized and will be reconstituted in diluent (NaCl for injection) prior to administration.

Placebo

Placebo is a normal saline solution (0.9% NaCl) for injection.

Route of administration: TDV, YF-17D vaccine and placebo will be administered by the SC route.

Duration of the Trial:	**Period of Evaluation:**
The trial duration for each subject will be approximately 360 days (12 months).	For the duration of a subject's participation.

Main Criteria for Evaluation and Analyses:

Primary Endpoint:

- Proportion of subjects YF and DENV-naive at Baseline who are seroprotected against YF on Day 30 (M1) as measured by PRNT (YF seroprotection rate). YF seroprotection is defined as reciprocal anti-YF neutralizing antibody titer ≥10. Immunological naivety to YF and DENV is defined as Baseline reciprocal neutralizing antibody titers <10 for YF and for the 4 dengue serotypes.

Secondary Endpoints:

Immunogenicity

- GMT of neutralizing antibodies (MNT_{50}) for each of the 4 dengue serotypes at pre-second and -third vaccinations (Day 90 [Month 3; M3] and Day 180 [Month 6; M6], respectively); and 1 month post first, second, and third vaccinations (Day 30 [M1], Day 120 [Month 4; M4], and Day 210 [Month 7; M7], respectively) in subjects YF and DENV-naive at Baseline.
- GMTs of anti-YF neutralizing antibodies at 1 month post first and third vaccinations (Day 30 [M1] and Day 210 [M7], respectively) in subjects YF and DENV-naive at Baseline.
- Seropositivity rates (% of subjects seropositive) for each of the 4 dengue serotypes at pre-second and -third vaccinations (Day 90 [M3] and Day 180 [M6], respectively); and 1 month post first, second, and third vaccinations (Day 30 [M1], Day 120 [M4], and Day 210 [M7], respectively) in subjects YF and DENV-naive at Baseline, where seropositivity is defined as a reciprocal neutralizing antibody titer ≥10.
- Seropositivity rates (% of subjects seropositive) for multiple (2, 3 or 4) dengue serotypes at pre-second and -third vaccinations (Day 90 [M3] and Day 180 [M6], respectively); and 1 month post first, second, and third vaccinations (Day 30 [M1], Day 120 [M4], and Day 210 [M7], respectively) in subjects YF and DENV-naive at Baseline, where seropositivity is defined as a reciprocal neutralizing antibody titer ≥10.
- Proportion of subjects YF and DENV-naive at Baseline who are seroprotected against YF at 1 month post third

vaccination (Day 210 [M7]) as measured by PRNT.

Safety

- Frequency and severity of solicited local (injection site[s]) AEs for 7 days (day of vaccination+6 days) and solicited systemic AEs for 14 days (day of vaccination+13 days) after each vaccination.
- Percentage of subjects with any unsolicited AEs within 28 days (day of vaccination+27 days) after each vaccination.
- Percentage of subjects with MAAEs and SAEs throughout the trial.

Statistical Considerations:

Analysis sets

Safety set: The safety set will consist of all subjects who received at least 1 dose of trial vaccine.

Full analysis set (FAS): The FAS will include all randomized subjects who received at least 1 dose of trial vaccine and for whom a valid pre-dosing and at least 1 post-dosing blood sample have been received for immunogenicity assessments.

Per–protocol set (PPS): The PPS will exclude all subjects seropositive for dengue virus and seroprotected for YF virus at Baseline (seropositivity is defined as a reciprocal neutralizing antibody titer $\geq$10 for the 4 dengue serotypes; YF seroprotection is defined as a reciprocal anti-YF neutralizing antibody titer $\geq$10) and will include all subjects in the FAS who have no major protocol violations. The major protocol violation criteria will be defined as part of the blinded data review prior to the unblinding of subject's trial vaccine assignment. The categories of major protocol violations include: (1) not meeting selected entry criteria, (2) not receiving the planned trial vaccinations or receiving a wrong trial vaccine, (3) receiving prohibited therapies, and (4) other major protocol violations that may be identified during blinded data reviews.

All summaries and analyses of safety data will be based on the safety set. The primary immunogenicity analyses will be based on the PPS; additional immunogenicity analyses may be provided based on the FAS.

Analysis of demographics and other Baseline characteristics

Age, gender, race, and other Baseline characteristics will be summarized descriptively by trial group for all randomized subjects. Unless specified otherwise, number of subjects with non-missing observations, mean, SD, median, minimum and maximum will be presented for continuous data; and frequency and percent will be presented for categorical data.

Immunogenicity analysis

Descriptive statistics including 95% CI for the primary and secondary endpoints, including seroprotection rates, seropositivity rates and GMTs, will be computed by trial group for all available assays at all relevant time points.

As a primary analysis, the NI of the immune response to the YF-17D vaccine when concomitantly administered with TDV (Group 3) compared to concomitant administration of the YF-17D vaccine with placebo (Group 1) will be assessed in terms of YF seroprotection rates on Day 30 (M1). NI will be concluded if the upper bound of the 95% CI for the seroprotection rate difference (Group 1 minus Group 3) is less than the NI margin of 5%. The Newcombe score method will be used to compute the 95% CI of the rate difference.

Secondary analyses will be performed as follows:

The NI of the immune response to TDV when concomitantly administered with YF-17D vaccine (Group 3) compared to concomitant administration of TDV with placebo (Group 2) will be assessed in terms of the GMTs of neutralizing antibodies for all 4 dengue serotypes on Day 120 (M4). NI will be concluded if the upper bound of the 95% CI for the GMT ratio (Group 2/Group 3) is less than the NI margin of 2.0. An analysis of variance (ANOVA) model will be used for this assessment.

The NI of the immune response to YF-17D vaccine when concomitantly administered with TDV (Group 3) compared to concomitant administration of YF-17D vaccine with placebo (Group 1) will be assessed in terms of the GMTs of

anti-YF neutralizing antibodies on Day 30 (M1). NI will be concluded if the upper bound of the 95% CI for the GMT ratio (Group 1/Group 3) is less than the NI margin of 2.0. An ANOVA model will be used for this assessment.

Handling of missing data and of values below the lower limit of quantification will be described in the statistical analysis plan (SAP).

Safety analysis

Solicited AEs

In all subjects the presence and severity (Grade) of solicited local (injection site[s]) AEs (pain, erythema and swelling) and solicited systemic AEs (fever, asthenia, malaise, headache and myalgia) will be collected for 7 days and 14 days respectively, following each vaccination (including the day of vaccination) via collection of diary cards.

For each solicited AE, the number and percentage of subjects with local (injection site[s]) and systemic AEs will be summarized by trial group and event severity for each day after each vaccination (i.e., Day 1 through Day 7 for local [injection site(s)] AEs and Day 1 through Day 14 for systemic AEs), and overall. Summaries of first onset of each event and the number of days subjects reported experiencing each event will also be provided. For subjects with more than 1 episode of the same event, the maximum severity will be used for tabulations.

Persistent/prolonged solicited local or systemic AEs continuing on Day 8 and Day 15, respectively, following each trial vaccination will be assessed separately. Unless otherwise specified these AEs will not be included in the analyses/tabulations of unsolicited AEs and will have separate listings.

Unsolicited AEs

In all subjects, unsolicited AEs will be assessed for 28 days following each vaccination (day of vaccination+27 days).

Unsolicited AEs will be coded using the Medical Dictionary for Regulatory Activities (MedDRA), and summarized by Preferred Term (PT) and System Organ Class (SOC) for each trial group.

Unsolicited AEs will be summarized as follows: by PT including events with frequency greater than a pre-defined frequency (the percentage will be specified in the SAP); by SOC and PT; by SOC, PT, and severity; and by SOC, PT, and relationship to the trial vaccine(s). Subjects reporting more than 1 occurrence for the term (level) being summarized will be counted only once.

AEs leading to trial or trial vaccine withdrawal will be collected and summarized for the entire study period.

MAAEs

In all subjects, MAAEs will be collected throughout the trial. MAAEs will be coded using MedDRA, and summarized by PT and SOC for each trial group.

SAEs

In all subjects, SAEs will be collected throughout the trial. SAEs will be coded using MedDRA, and summarized by PT and SOC for each trial group.

Sample Size Justification:

The immunogenicity set sample size calculation assumes a significance level of 0.025 (one-sided).

For the primary objective of showing NI in YF seroprotection rates, the calculation assumes a NI margin of 5%, and a YF seroprotection rate of 98% at 1 month after YF-17D vaccination in the two trial groups (Group 1 and Group 3).

For the secondary objective of showing NI in GMTs of neutralizing antibodies for all the 4 dengue serotypes, the calculation assumes a NI margin of 2.0, that the true GMT ratio for the two trial groups (Group 2 and Group 3) is 1, and that the natural logarithm of titers are distributed as normal distributions with SDs of 1.35, 0.86, 1.21, and 1.27.

For the secondary objective of showing NI in GMTs of anti-YF neutralizing antibodies, the calculation assumes a NI margin of 2.0, that the true GMT ratio for the two trial groups (Group 1 and Group 3) is 1, and that the natural logarithm of titers are distributed as normal distributions with a SD of 1.31.

A sample size of 300 subjects per trial group, with approximately 255 evaluable subjects per trial group (assuming approximately 15% dropouts and non-evaluable subjects), is sufficient to achieve approximately 90% overall power for showing NI for the above primary and secondary objectives. A total sample size of 900 subjects also ensures that a sufficient number of healthy YF/DENV-naive adults will be vaccinated to support the safe use of TDV in travelers.

The power calculations were based on nQuery Advisor® 6.01.

Interim Analysis:

No interim analyses are planned.

Data Monitoring Committee:

A data monitoring committee (DMC) will have oversight of this trial. The DMC functions at a program level and further information is available in the DMC Charter.

2.1 Schedule of Trial Procedures

Visit number		**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	**V7**
Day/Month		**Day 1** **M0**	**Day 30** **M1**	**Day 90** **M3**	**Day 120 (V3+30 days)** **M4**	**Day 180 (V3+90 days)** **M6**	**Day 210 (V5+30 days)** **M7**	**Day 360 (V5+180 days)** **M12 (ET) [a]**
Visit window (days)		**±0**	**-1/+7**	**-4/+7**	**-1/+7**	**-4/+7**	**-1/+7**	**-7/+14**
Signed informed consent [b]		X						
Assessment of eligibility criteria [c]		X						
Demographics [b]		X						
Medical history [b]		X						
Concomitant medications/ vaccinations [d]		X	X	X	X	X	X	X
Check contraindications to vaccination				X		X		
Check criteria for delay of vaccination				X		X		
Complete physical examination [e]		X		X		X		
Targeted physical examination [f]			X		X		X	X
Vital signs [g]		X	X	X	X	X	X	X
Pregnancy test [h]		X		X		X		
Pregnancy avoidance guidance [i]		X	X	X	X	X	X	
Randomization		X						
Blood Collection	YF neutralizing antibodies (10 mL) [j]	X	X			X	X	
	Dengue neutralizing antibodies (5 mL) [j]	X	X	X	X	X	X	
Vaccine administration		X		X		X		
30 min post-vaccination in-clinic observation including injection site evaluation and body temperature measurement [k]		X		X		X		

Visit number		**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	**V7**
Day/Month		**Day 1** **M0**	**Day 30** **M1**	**Day 90** **M3**	**Day 120 (V3+30 days)** **M4**	**Day 180 (V3+90 days)** **M6**	**Day 210 (V5+30 days)** **M7**	**Day 360 (V5+180 days)** **M12 (ET) [a]**
Visit window (days)		**±0**	**-1/+7**	**-4/+7**	**-1/+7**	**-4/+7**	**-1/+7**	**-7/+14**
Diary Card [l]	Distribution	X		X		X		
	Review/collection		X		X		X	
Unsolicited AEs [m]		X	X	X	X	X	X	
SAEs [n]		X	X	X	X	X	X	X
MAAEs [n]		X	X	X	X	X	X	X
AEs leading to discontinuation or withdrawal [n]		X	X	X	X	X	X	X

Note: AE=adverse event, ET=early termination, M=Month, MAAE=medically attended adverse event, SAE=serious adverse event, V=visit, YF=yellow fever

Footnotes:

(a) If the subject terminates early, Day 360 (M12) procedures should be performed.

(b) Prior to the subject entering into the trial, and before any protocol-directed procedures are performed.

(c) Review of inclusion/exclusion criteria will be performed prior to the first trial vaccination at Day 1 (M0). After written informed consent has been obtained and eligibility is assessed, subjects will be randomized to one of the 3 trial groups.

(d) Any other vaccination against any flavivirus (licensed or investigational, including any other dengue vaccine) during the entire trial period, all concomitant medications and vaccine history from 1 month (minimum 28 days) prior to administration of each dose of trial vaccine(s) up to 1 month (minimum 28 days) thereafter, steroids and immunostimulants within 60 days prior to Day 1 (M0), immunoglobulins and blood products within 3 months prior to Day 1 (M0), and immunosuppressive therapy within 6 months prior to Day 1 (M0).

(e) Physical examination at Day 1 (M0) including measurement of weight and height; BMI will be calculated automatically. Measurement of height is only required at Day 1 (M0).

(f) Subjects may undergo a brief symptom-directed physical examination. Clinically significant changes from the Baseline examination should be recorded in the subject's source documents and electronic Case Report Form (eCRF).

(g) Vital signs including (but not limited to) the measurement of systolic blood pressure/diastolic blood pressure, heart rate, and body temperature.

(h) Pregnancy testing (serum or urine) for females of childbearing potential. Results must be confirmed and documented as negative prior to each vaccination. Additional pregnancy tests may be performed during the trial if deemed necessary by the Investigator.

(i) Females of childbearing potential who are sexually active will be provided with information on acceptable methods of contraception and will be asked prior to vaccination on Day 1 (M0) to sign the informed consent form stating that they understand the requirements for avoidance of pregnancy and donation of ova. Subjects will be reminded during trial visits to adhere to acceptable contraceptive methods and not donate ova up to 6 weeks post last trial vaccination at Day 180 (M6).

(j) The blood sample on Day 1 (M0), Day 90 (M3) and Day 180 (M6) should be taken prior to trial vaccination. The blood sample on Day 120 (M4) and Day 210 (M7) should be taken at least 29 days after the vaccination on Day 90 (M3) and Day 180 (M6), respectively.

(k) After each trial vaccination, the subject will be observed for at least 30 minutes including observation for solicited local (injection site) and systemic AEs, unsolicited AEs (non-serious and serious), and body temperature measurement.

(l) Diary (paper or electronic) cards will be used for the collection of:
1) Solicited local (injection site) AEs for 7 days after each vaccination (including the day of vaccination). If solicited local AEs continue on Day 8 following each trial vaccination, record the extended information on the Adverse Event eCRF.
2) Solicited systemic AEs for 14 days after each vaccination (including the day of vaccination). If solicited systemic AEs continue on Day 15 following each trial vaccination, record the extended information on the Adverse Event eCRF.
The Investigator will categorize events by severity (mild, moderate or severe) and will assess causality to vaccine administration for solicited systemic events (related or not related).

(m) Unsolicited AEs for 28 days (including the day of vaccination) after each vaccination will be collected by interview and recorded for all subjects at Day 30 (M1), Day 120 (M4) and Day 210 (M7). The Investigator will categorize events by severity (mild, moderate or severe) and will assess causality to vaccine administration (related or not related).

(n) AEs leading to discontinuation or withdrawal, MAAEs and SAEs will be collected for the trial duration. The Investigator will categorize events by severity (mild, moderate or severe) and will assess causality to vaccine administration (related or not related).

3.0 TRIAL REFERENCE INFORMATION

3.1 Trial-Related Responsibilities

The Sponsor will perform all trial-related activities with the exception of those identified in the 'Trial-Related Responsibilities' template. The identified vendors in the template for specific trial-related activities will perform these activities in full or in partnership with the Sponsor.

3.2 Principal Investigator/Coordinating Investigator

The Sponsor will select a Signatory Principal Investigator/Coordinating Investigator from the Investigators who participate in the trial. Selection criteria for this Investigator will include significant knowledge of the trial protocol, the investigational vaccine, expertise in the therapeutic area and the conduct of clinical research as well as trial participation. The Signatory Principal Investigator/Coordinating Investigator will be required to review and sign the clinical study report and by doing so agrees that it accurately describes the results of the trial.

3.3 List of Abbreviations

AE	Adverse Event
ANOVA	Analysis Of Variance
BMI	Body Mass Index
CFR	Code of Federal Regulations
CYD-TDV	Chimeric Yellow fever virus-Dengue virus Tetravalent Dengue Vaccine
DENV	Wild Type Dengue Virus
DENV-1, -2, -3, -4	Wild Type Dengue Virus Serotypes 1, 2, 3, and 4
DHF	Dengue Hemorrhagic Fever
DMC	Data Monitoring Committee
DSS	Dengue Shock Syndrome
eCRF	electronic Case Report Form
EudraCT	European Clinical Trials Database
FAS	Full Analysis Set
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GMT	Geometric Mean Titer
HIV	Human Immunodeficiency Virus
ICH	International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use
IEC	Independent Ethics Committee
IND	Investigational New Drug
IRB	Institutional Review Board
IRT	Interactive Response Technology
JE	Japanese Encephalitis
M0, 1, 3, 4, 6, 7, 12	Month 0, 1, 3, 4, 6, 7, 12
MAAE	Medically Attended Adverse Event
MedDRA	Medical Dictionary for Regulatory Activities
MNT_{50}	Microneutralization Test 50%
NaCl	Sodium Chloride
NI	Non-inferiority
PPS	Per–Protocol Set
PRNT	Plaque Reduction Neutralization Test
PT	Preferred Term
SAE	Serious Adverse Event

SAP	Statistical Analysis Plan
SC	Subcutaneous
SOC	System Organ Class
SUSAR	Suspected Unexpected Serious Adverse Reactions
TDV	Tetravalent Dengue Vaccine Candidate
TDV-1	Dengue serotypes 2/1 chimeric strain
TDV-2	Molecularly characterized, attenuated dengue serotype 2 strain
TDV-3	Dengue serotypes 2/3 chimeric strain
TDV-4	Dengue serotypes 2/4 chimeric strain
US	United States
WHO	World Health Organization
WN	West Nile
YF	Yellow Fever

3.4 Corporate Identification

TV Takeda Vaccines, Inc.

4.0 INTRODUCTION

4.1 Background

Dengue fever is caused by infection with the wild type dengue virus (DENV), a ribonucleic acid virus that occurs as 4 recognized serotypes, DENV-1, DENV-2, DENV-3, or DENV-4. These dengue viruses are transmitted from human to human by mosquitoes (primarily *Aedes aegypti*). The 4 dengue viruses are endemic in Asia, Central and South America, the Caribbean, the Pacific Islands, and parts of Africa. There are an estimated 390 million dengue infections per year worldwide, which is more than 3 times the previous World Health Organization (WHO) estimate of 50 to 100 million cases. Every year, around 500,000 cases of dengue hemorrhagic fever (DHF) require hospitalization with an estimated death rate of 2.5%, primarily in children. It is estimated that 3.9 billion people are at risk of dengue infection [1-4].

Dengue fever is clinically defined as an acute febrile illness with 2 or more of the following manifestations: headache, retro-orbital pain, myalgia, arthralgia, rash, hemorrhagic manifestations, or leucopenia, and occurrence at the same location and time as other confirmed cases of dengue fever. The most severe forms of dengue infection – DHF and dengue shock syndrome (DSS) – are life threatening. Primary infection with any one of the 4 dengue serotypes is thought to result in life-long protection from re-infection by the same serotype, but does not protect against a secondary infection by one of the other 3 dengue serotypes and may lead to an increased risk of severe disease (DHF/DSS) [3-6].

Treatment of dengue fever is based solely on signs and symptoms, with fluid replacement required for hemorrhagic or shock cases. An antiviral therapy for dengue virus infection is not available. Preventive measures that rely on mosquito control and individual protection are of limited efficacy, complex to implement and questionable in terms of cost-effectiveness. There is a great unmet global public health need for a safe and effective vaccine to reduce the morbidity and mortality associated with dengue disease [1-7]. Vaccine development has focused on tetravalent vaccines that provide protection against all 4 dengue serotypes simultaneously since all 4 dengue serotypes commonly co-circulate in endemic areas. A first tetravalent dengue vaccine (chimeric yellow fever virus-dengue virus tetravalent dengue vaccine [CYD-TDV]) has been recently approved in some countries in Asia and Latin America. [8]. Initial findings showed that vaccine efficacy was different between serotypes and depended on dengue pre-exposure status [9]. Additionally, recent analyses found that people who had not been infected by dengue virus before vaccination had a higher risk of getting severe disease when they were infected with dengue virus after vaccination with CYD-TDV [10]. Hence, there is a continued unmet public health need for safer and more efficacious dengue vaccines.

Takeda's Tetravalent Dengue Vaccine Candidate (TDV) - Background:

Takeda's TDV consists of 1 molecularly characterized, attenuated dengue serotype 2 virus strain and 3 chimeric recombinant dengue virus strains expressing surface antigens corresponding to dengue serotypes 1-4. The dengue serotype 2 strain (TDV-2) is based upon the attenuated laboratory-derived virus, DENV-2 Primary Dog Kidney (PDK)-53, originally isolated at Mahidol University, Bangkok, Thailand [11]. The chimeric, attenuated vaccine strains for dengue serotypes

1, 3 and 4 were engineered by substituting the structural genes, pre-membrane (prM) and envelope (E), of TDV-2 with the prM and E genes of the wild type (DENV) virus strains, DENV-1 16007, DENV-3 16562 or DENV-4 1036 virus, respectively [12]. Thus TDV is comprised of 4 dengue virus strains: a molecularly characterized, attenuated dengue serotype 2 strain (TDV-2), a dengue serotypes 2/1 chimeric strain (TDV-1), a dengue serotypes 2/3 chimeric strain (TDV-3), and a dengue serotypes 2/4 chimeric strain (TDV- 4).

Non-clinical studies carried out in mice and non-human primates have demonstrated an acceptable safety, immunogenicity, and efficacy profile of TDV. Additionally, data from completed phase 1 and phase 2 trials have shown satisfactory reactogenicity, safety and immunogenicity profiles of TDV in adults in non-endemic areas as well as in adults and children in endemic areas in Asia and Latin America. Ongoing and completed phase 2 trials have enabled the selection of a final TDV dose (lyophilized formulation) and a 2-dose vaccination regimen 3 months apart by subcutaneous (SC) injection for use in the pivotal program.

The current Investigator's Brochure for TDV contains additional product information and a more detailed review of pre-clinical and clinical trials [13].

4.2 Rationale for the Proposed Trial

Dengue and yellow fever (YF) viruses belong to the same family of *Flaviviridae* and share antigenic determinants, which may result in cross-reacting antibodies [14]. They are both transmitted between humans by mosquitoes (primarily *Aedes aegypti*), and are endemic in tropical areas of Africa and Latin America with a high public health impact. The YF-17D vaccine which is based on a live, attenuated viral strain, is the only commercially available YF vaccine administered as a single SC injection. The YF-17D vaccine is highly effective (approaching 100%) and generally safe with the exception of very rare cases of vaccine-associated neurotropic and viscerotropic disease [15]. Vaccination against YF is required for travelers to certain countries in accordance with the International Health Regulations [16], and is also recommended by the WHO for all subjects travelling to areas where there is evidence of persistent or periodic YF virus transmission [15].

The main purpose of this trial is to assess the immunogenicity and safety of the concomitant administration of YF-17D vaccine and TDV in healthy subjects aged 18 to 60 years living in country(ies) non-endemic for both dengue and YF. Because broader and enhanced immune responses following vaccination with a dengue vaccine have been reported in YF-immune subjects compared to YF non-immune subjects, a secondary purpose of this trial is to assess the immunogenicity and safety of the sequential administration of YF-17D vaccine and the 2-dose regimen of TDV [17, 18]. To maintain the trial blind, a placebo (0.9% sodium chloride [NaCl] solution) will be used. Approximately 900 subjects are planned to be enrolled in the trial.

This trial will be conducted in accordance with the protocol, the International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use and Good Clinical Practice (ICH-GCP) Guidelines, and applicable regulatory requirements.

5.0 TRIAL OBJECTIVES AND ENDPOINTS

5.1 Objectives

5.1.1 Primary Objective

- To demonstrate non-inferiority (NI) of the YF seroprotection rate response to 1 dose of YF-17D vaccine, 1 month following concomitant administration with 1 dose of TDV compared to placebo.

5.1.2 Secondary Objectives

Immunogenicity:

- To demonstrate NI of the geometric mean titer (GMT) response to TDV for all 4 dengue serotypes, 1 month post second dose of TDV following concomitant administration of the first dose of TDV with YF-17D vaccine or placebo.
- To demonstrate NI of the GMT response to 1 dose of YF-17D vaccine, 1 month following concomitant administration with 1 dose of TDV compared to placebo.
- To describe the seropositivity rates for all 4 dengue serotypes, 1 month post second dose of TDV following concomitant administration of the first dose of TDV with YF-17D vaccine or placebo.
- To describe the GMTs of neutralizing antibodies and the seropositivity rates for all 4 dengue serotypes, 1 month post first dose of TDV administered concomitantly with YF-17D vaccine or placebo.
- To describe the GMT and seroprotection rate response to YF-17D vaccine, 1 month after sequential administration of a 2-dose regimen of TDV at 0 and 3 months, followed by YF-17D vaccine 3 months later.
- To describe the GMTs of neutralizing antibodies and the seropositivity rates for all 4 dengue serotypes, 1 month after sequential administration of YF-17D vaccine at month 0, followed by a 2-dose regimen of TDV 3 months later at 3 and 6 months.

Safety:

- To assess the safety profile after each vaccine injection in Groups 1, 2 and 3.

5.2 Endpoints

5.2.1 Primary Endpoint

- Proportion of subjects YF and DENV-naive at Baseline who are seroprotected against YF on Day 30 (Month 1 [M1]) as measured by plaque reduction neutralization test (PRNT) (YF seroprotection rate). YF seroprotection is defined as reciprocal anti-YF neutralizing antibody

titer ≥10. Immunological naivety to YF and DENV is defined as Baseline reciprocal neutralizing antibody titers <10 for YF and for the 4 dengue serotypes.

5.2.2 Secondary Endpoints

Immunogenicity:

- GMT of neutralizing antibodies (microneutralization test 50% [MNT_{50}]) for each of the 4 dengue serotypes at pre-second and -third vaccinations (Day 90 [Month 3; M3] and Day 180 [Month 6; M6], respectively); and 1 month post first, second, and third vaccinations (Day 30 [M1], Day 120 [Month 4; M4], and Day 210 [Month 7; M7], respectively) in subjects YF and DENV-naive at Baseline.
- GMTs of anti-YF neutralizing antibodies at 1 month post first and third vaccinations (Day 30 [M1] and Day 210 [M7], respectively) in subjects YF and DENV-naive at Baseline.
- Seropositivity rates (% of subjects seropositive) for each of the 4 dengue serotypes at pre-second and -third vaccinations (Day 90 [M3] and Day 180 [M6], respectively); and 1 month post first, second, and third vaccinations (Day 30 [M1], Day 120 [M4], and Day 210 [M7], respectively) in subjects YF and DENV-naive at Baseline, where seropositivity is defined as a reciprocal neutralizing antibody titer ≥10.
- Seropositivity rates (% of subjects seropositive) for multiple (2, 3 or 4) dengue serotypes at pre-second and -third vaccinations (Day 90 [M3] and Day 180 [M6], respectively); and 1 month post first, second, and third vaccinations (Day 30 [M1], Day 120 [M4], and Day 210 [M7], respectively) in subjects YF and DENV-naive at Baseline, where seropositivity is defined as a reciprocal neutralizing antibody titer ≥10.
- Proportion of subjects YF and DENV-naive at Baseline who are seroprotected against YF at 1 month post third vaccination (Day 210 [M7]) as measured by PRNT.

Safety:

- Frequency and severity of solicited local (injection site[s]) Adverse Events (AE) for 7 days (day of vaccination+6 days) and solicited systemic AEs for 14 days (day of vaccination+13 days) after each vaccination.
- Percentage of subjects with any unsolicited AEs within 28 days (day of vaccination+27 days) after each vaccination.
- Percentage of subjects with Medically Attended AEs (MAAE) and Serious AEs (SAE) throughout the trial.

6.0 TRIAL DESIGN AND DESCRIPTION

6.1 Trial Design

This is a phase 3, observer-blind, randomized, multi-center trial in 900 healthy adults aged 18 to 60 years in non-endemic areas for dengue and YF to investigate the immunogenicity and safety of the concomitant and sequential administration of TDV and YF-17D vaccine. Subjects will be randomized equally (1:1:1 ratio) to one of the following 3 trial groups (300 subjects per trial group):

- Group 1: YF-17D vaccine+placebo concomitantly administered on Day 1 (Month [M0]), first dose of TDV administered on Day 90 (M3) and second dose of TDV administered on Day 180 (M6).
- Group 2: first dose of TDV+placebo concomitantly administered on Day 1 (M0), second dose of TDV administered on Day 90 (M3) and YF-17D vaccine administered on Day 180 (M6).
- Group 3: first dose of TDV+YF-17D vaccine concomitantly administered on Day 1 (M0), second dose of TDV administered on Day 90 (M3) and placebo administered on Day 180 (M6).

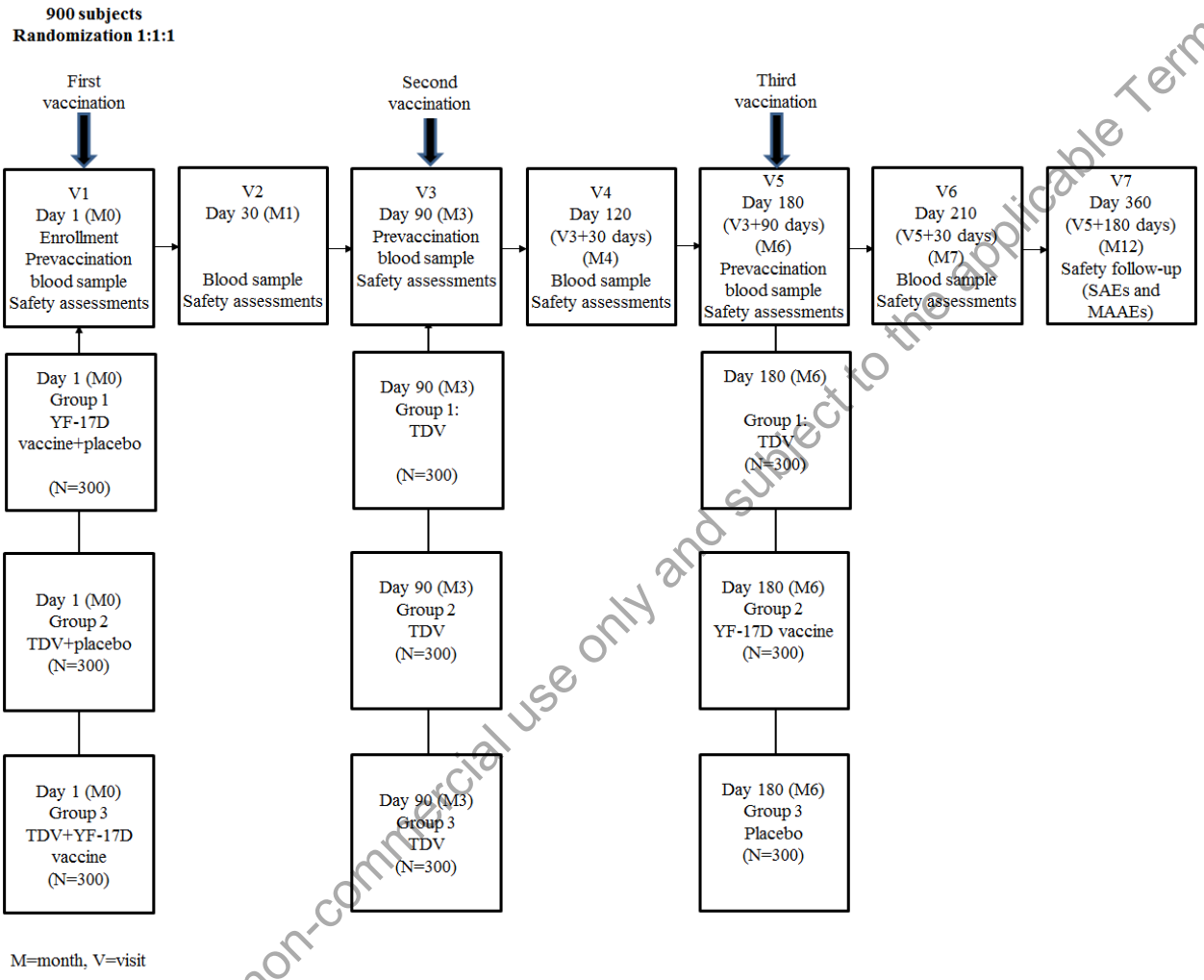
Concomitantly administered vaccines will be injected into opposite arms. All subjects will be followed-up for 6 months after the third vaccination (administered approximately 6 months after the first vaccination), so the trial duration will be approximately 360 days (12 months) for each subject.

A trial population of 900 subjects (300 subjects per trial group) is considered sufficient for the NI assessment of the immune response to the YF-17D vaccine when concomitantly administered with TDV compared to placebo, and for the NI assessment of the immune response to TDV when TDV is concomitantly administered with YF-17D vaccine compared to placebo.

Safety parameters include solicited local (injection site) and solicited systemic AEs for 7 days (day of vaccination+6 days) and 14 days (day of vaccination+13 days) after each vaccination, respectively, unsolicited AEs for 28 days (day of vaccination+27 days) after each vaccination, MAAEs and SAEs throughout the trial.

A schematic of the trial design is included as Figure 6.a. A schedule of trial procedures is provided in Section 2.1.

Figure 6.a Schematic of Trial Design



Immunogenicity evaluation:

Blood samples for the measurement of dengue neutralizing antibodies (MNT_{50}) will be collected at pre-first vaccination (Day 1 [M0]), 1 month post first vaccination (Day 30 [M1]), pre-second vaccination (Day 90 [M3]), 1 month post second vaccination (Day 120 [M4]), pre-third vaccination (Day 180 [M6]), and 1 month post third vaccination (Day 210 [M7]).

Blood samples for the measurement of YF neutralizing antibodies (PRNT) will be collected at pre-first vaccination (Day 1 [M0]), 1 month post first vaccination (Day 30 [M1]), pre-third vaccination (Day 180 [M6]), and 1 month post third vaccination (Day 210 [M7]).

Safety evaluation:

- Diary cards (paper or electronic) will be distributed to all subjects for the recording of:

  - Solicited local AEs for 7 days following each vaccination (day of vaccination+6 days). These will include: injection site pain, injection site erythema, and injection site swelling, and will be collected at each injection site.
  - Solicited systemic AEs for 14 days following each vaccination (day of vaccination+13 days). These will include: fever, headache, asthenia, malaise, and myalgia.
- Unsolicited AEs for 28 days following each vaccination (day of vaccination+27 days).
- SAEs and MAAEs will be recorded for the trial duration. MAAEs are defined as AEs leading to an unscheduled visit to or by a healthcare professional including visits to an emergency department, but not fulfilling seriousness criteria.

Data collection will be by electronic Case Report Form (eCRF).

6.2 Justification for Trial Design, Dose, and Endpoints

The trial design and the collection of solicited AEs, unsolicited AEs (non-serious AEs and SAEs), and MAAEs following vaccination are consistent with vaccine evaluation trials.

Ongoing and completed phase 2 trials have enabled the selection of a final TDV dose (lyophilized formulation) and a 2-dose vaccination regimen 3 months apart by SC injection for use in the pivotal program.

The timing of the primary and secondary endpoints after vaccination is consistent with previous trials with TDV. Dengue neutralizing antibodies have been generally accepted as the immune response endpoint for dengue vaccine trials.

The trial is observer-blind. A placebo (0.9% NaCl solution) will be used at the appropriate time point to minimize bias.

As the trial will be conducted in non-endemic areas for dengue, a 6-month follow-up period after the third trial vaccination is considered adequate.

A sample size of 300 subjects per trial group (i.e., 900 subjects in total) is considered sufficient for the immunogenicity assessments. The total sample size of 900 subjects is also to ensure that a sufficient number of healthy YF/DENV-naive adults will be vaccinated to support the safe use of TDV in travelers. Refer to Section 13.2.

The rationale for the proposed trial is given in Section 4.2.

6.3 Duration of Subject's Expected Participation in the Entire Trial

The trial duration for each subject will be approximately 360 days (12 months) including vaccination (Day 1 [M0], Day 90 [M3] and Day 180 [M6]) and follow-up through Day 360 (Month 12 [M12]).

6.4 Premature Termination or Suspension of Trial or Investigational Site

6.4.1 Criteria for Premature Termination or Suspension of the Trial

The trial will be completed as planned unless one or more of the following criteria are satisfied that require temporary suspension or early termination of the trial.

- New information or other evaluation regarding the safety or efficacy of the investigational vaccine that indicates a change in the known risk/benefit profile, such that the risk/benefit is no longer acceptable for subjects participating in the trial.
- The data monitoring committee (DMC) recommends that the trial should be suspended or terminated.
- Significant deviation from GCP that compromises the ability to achieve the primary trial objectives or compromises subject safety.

6.4.2 Criteria for Premature Termination or Suspension of Investigational Sites

A trial site may be terminated prematurely or suspended if the site (including the Investigator) is found in significant deviation from GCP, protocol, or contractual agreement, is unable to ensure adequate performance of the trial, or as otherwise permitted by the contractual agreement.

6.4.3 Procedures for Premature Termination or Suspension of the Trial or the Participation of Investigational Site(s)

In the event that the Sponsor, an institutional review board (IRB)/independent ethics committee (IEC) or regulatory authority elects to terminate or suspend the trial or the participation of an investigational site, a trial-specific procedure for early termination or suspension will be provided by the Sponsor; the procedure will be followed by applicable investigational sites during the course of termination or trial suspension.

7.0 SELECTION AND DISCONTINUATION/WITHDRAWAL OF SUBJECTS

All entry criteria, including test results, need to be confirmed prior to randomization.

7.1 Inclusion Criteria

Subject eligibility is determined according to the following criteria:

1. The subject is aged 18 to 60 years, inclusive.
2. Individuals who are in good health at the time of entry into the trial as determined by medical history, physical examination (including vital signs), and the clinical judgment of the Investigator.
3. The subject signs and dates a written informed consent form and any required privacy authorization prior to the initiation of any trial procedures, and after the nature of the trial has been explained according to local regulatory requirements.
4. Individuals who can comply with trial procedures and are available for the duration of follow-up.

7.2 Exclusion Criteria

Any subject who meets any of the following criteria will not qualify for entry into the trial:

1. Individuals with an elevated oral temperature ≥ 38°C (100.4°F) within 3 days of the intended date of vaccination (consider whether applicable as an exclusion criterion or criterion for delay, see below).
2. Known hypersensitivity or allergy to any of the trial vaccine components (including excipients of the investigational vaccines and placebo).
3. Individuals with contraindications, warnings and/or precautions to vaccination with the YF-17D vaccine as specified within the product information (especially history of thymus dysfunction).
4. Individuals with behavioral or cognitive impairment or psychiatric disease that, in the opinion of the Investigator, may interfere with the subject's ability to participate in the trial.
5. Individuals with any history of progressive or severe neurologic disorder, seizure disorder or neuro-inflammatory disease (e.g., Guillain-Barré syndrome).
6. Individuals with a history of or any current illness that, in the opinion of the Investigator, might interfere with the results of the trial or pose additional risk to the subjects due to participation in the trial.
7. Known or suspected impairment/alteration of immune function, including:
    a. Chronic use of oral steroids (equivalent to 20 mg/day prednisone ≥12 weeks/≥2 mg/kg body weight/day prednisone ≥2 weeks) within 60 days prior to Day 1 (M0) (use of inhaled, intranasal, or topical corticosteroids is allowed).

b. Receipt of parenteral steroids (equivalent to 20 mg/day prednisone ≥12 weeks/≥2 mg/kg body weight/day prednisone ≥2 weeks) within 60 days prior to Day 1 (M0).

c. Administration of immunoglobulins and/or any blood products within the 3 months prior to Day 1 (M0) or planned administration during the trial.

d. Receipt of immunostimulants within 60 days prior to Day 1 (M0).

e. Immunosuppressive therapy such as anti-cancer chemotherapy or radiation therapy within 6 months prior to Day 1 (M0).

f. Human immunodeficiency virus (HIV) infection or HIV-related disease.

g. Hepatitis C virus infection.

h. Genetic immunodeficiency.

8. Abnormalities of splenic or thymic function.
9. Individuals with a known bleeding diathesis, or any condition that may be associated with a prolonged bleeding time.
10. Individuals with any serious chronic or progressive disease according to the judgment of the Investigator (e.g., neoplasm, insulin dependent diabetes, cardiac, renal or hepatic disease).
11. Individuals with body mass index (BMI) greater than or equal to 35 kg/m^2 (=weight in kg/[height in meters^2]).
12. Individuals or their first degree relatives involved in the trial conduct.
13. Intent to travel to dengue or YF endemic countries during the trial period.
14. Individuals with history of substance or alcohol abuse within the past 2 years.
15. Female subjects who are pregnant or breastfeeding.
16. Any positive or indeterminate pregnancy test.
17. Females of childbearing potential[1] who are sexually active and who have not used any of the acceptable contraceptive methods[2] for at least 2 months prior to Day 1 (M0).
18. Females of childbearing potential who are sexually active and who refuse to use an acceptable contraceptive method up to 6 weeks post third vaccination on Day 180 (M6). In addition, they must be advised not to donate ova during this period.

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[1] Defined as status post onset of menarche and not meeting any of the following conditions: bilateral tubal ligation (at least 1 year previously), bilateral oophorectomy (at least 1 year previously) or hysterectomy.

[2] One or more of the following: hormonal contraceptive (such as oral, injection, transdermal patch, implant, cervical ring), barrier method (condom with spermicide or diaphragm with spermicide) each and every time during intercourse; intrauterine device, monogamous relationship with vasectomized partner (partner must have been vasectomized for at least 6 months prior to Day 1 [M0]). Other contraceptive methods may be considered in agreement with the Sponsor and will be approved by the appropriate ethics committee.

19. Individuals participating in any clinical trial with another investigational product 30 days prior to Day 1 (M0) or intent to participate in another clinical trial at any time during the conduct of this trial.
20. Individuals who received any other vaccines within 14 days (for inactivated vaccines) or 28 days (for live vaccines) prior to enrollment in this trial or who are planning to receive any non-trial vaccine within 28 days of trial vaccine administration.
21. Previous and planned vaccination (during the trial conduct), against any flavivirus including dengue, YF, Japanese encephalitis (JE) or tick-borne encephalitis viruses.
22. Previous participation in any clinical trial of a dengue or other flavivirus (e.g., West Nile [WN] virus) candidate vaccine, except for subjects who received placebo in those trials.
23. Subjects with a current or previous infection with a flavivirus such as dengue, Zika, YF, JE, WN fever, or Saint Louis encephalitis viruses and subjects with a history of prolonged (≥1 year) habitation in a dengue endemic area.

There may be instances when individuals meet all entry criteria except one that relates to transient clinical circumstances (e.g., body temperature elevation or recent use of excluded medication or vaccine). Under these circumstances, eligibility for trial enrollment may be considered if the appropriate window for delay has passed, inclusion/exclusion criteria have been rechecked, and if the subject is confirmed to be eligible.

7.3 Criteria for Delay of Vaccination and/or Blood Sampling

After enrollment, subjects may encounter clinical circumstances that warrant a delay in the administration of trial vaccination. These situations are listed below. In the event that a subject meets a criterion for delay of vaccination, the subject may receive trial vaccination once the window for delay has passed as long as the subject is otherwise eligible for trial participation. The decision to vaccinate in those situations will be made by the Investigator.

1. Body temperature ≥38.0°C (≥100.4°F) within 3 days of intended trial vaccination, and/or use of antipyretics and/or analgesic medication for either reason within 24 hours prior to vaccine administration.
2. Receipt of any vaccine other than the trial vaccines within 14 days (for inactivated vaccines) or 28 days (for live vaccines) of intended trial vaccination.
3. Known or suspected altered or impairment of immune function as specified under the exclusion criteria.

<u>Contraindications to vaccination at Day 90 (M3) and Day 180 (M6):</u>

There are also circumstances under which receipt of the vaccination at Day 90 (M3) and Day 180 (M6) is contraindicated in this trial. These circumstances include but are not limited to anaphylaxis or severe hypersensitivity reactions following the administration of first or second trial vaccination at Day 1 (M0) or Day 90 (M3), respectively. If these reactions occur, the subject

must not receive the trial vaccination at Day 90 (M3) or Day 180 (M6), respectively, but is encouraged to continue trial participation for safety follow up.

7.4 Criteria for Early Trial Termination of a Subject

Under some circumstances, a subject's trial participation may be terminated early. This means that no further trial procedures (including data collection) will be performed on that subject beyond the specific date of early termination. The primary reason for early termination of the subject from the trial should be recorded in the eCRF "end of trial visit" page using the following categories. For screen failure subjects, refer to Section 9.1.10.

1. Adverse Event: The subject has experienced an AE (irrespective of being related/unrelated to the trial vaccine or trial-related procedures) that requires early termination because continued participation imposes an unacceptable risk to the subject's health and/or the subject is unwilling to continue participation because of the AE. If the subject is unwilling to continue because of the AE the primary reason for early termination in this case will be 'withdrawal due to AE' and not 'withdrawal of consent', see below. Any ongoing AEs leading to early termination should be followed by the Investigator until resolution or stabilization.
2. Lost to follow-up: The subject did not return to the clinic and at least 3 attempts to contact the subject were unsuccessful. Attempts to contact the subject must be documented.
3. Withdrawal of consent: The subject wishes to withdraw from the trial. The primary reason for early termination will be 'withdrawal of consent" if the subject withdraws from participation due to a non-medical reason (i.e., reason other than AE). The reason for withdrawal, if provided, should be recorded in the eCRF.

   Note: All attempts should be made to determine the underlying reason for the withdrawal and, where possible, the primary underlying reason should be recorded.
4. Premature trial termination by Sponsor, a regulatory agency, the IEC/IRB, or any other authority.

   If the clinical trial is prematurely terminated by the Sponsor, the Investigator is to promptly inform the trial subjects and local EC/IRB and should ensure appropriate follow up for the subjects. The primary reason for early termination in this case will be 'trial termination".
5. Subject's death during trial participation.
6. Other.

   Note: The specific reasons should be recorded in the "specify" field of the eCRF.

7.5 Criteria for Premature Discontinuation of Trial Vaccine Administration

Early trial termination of a subject will by default prevent the subject from continued trial vaccine administration, as the subject will no longer be participating in the trial. In addition to early termination criteria (see Section 7.4), other situations may apply in which subjects may continue participating in the trial (e.g., contributing safety data according to protocol) but trial vaccine administration is discontinued selectively. Regardless of the reasons for discontinuation of trial vaccine administration, this must be documented as protocol deviation. Even if the subject is deemed ineligible to receive trial vaccine, all efforts should be made to continue the collection of safety data according to protocol. In addition, the primary reason for premature discontinuation of trial vaccine administration should be recorded in the eCRF "end of trial vaccine administration" page using the following categories:

1. Adverse Event: The subject has experienced an AE (irrespective of being related/unrelated to the trial vaccine or trial-related procedures) for which subsequent trial vaccine administrations impose an unacceptable risk to the subject's health, but the subject may continue trial participation for safety, or a subset of other trial procedures.
2. Lost to follow-up: The subject did not return to the clinic and at least 3 attempts to contact the subject were unsuccessful. Attempts to contact the subject must be documented.
3. Withdrawal of consent: The subject wishes to withdraw from the trial. The primary reason for early termination will be "withdrawal of consent" if the subject withdraws from participation due to a non-medical reason (i.e., reason other than AE). The reason for withdrawal, if provided, should be recorded in the eCRF.
4. Premature trial termination by Sponsor, a regulatory agency, the IEC/IRB, or any other authority. If the clinical trial is prematurely terminated by the Sponsor, the Investigator is to promptly inform the trial subjects and local IEC/IRB and should ensure appropriate follow up for the subjects. The primary reason for early termination in this case will be "trial termination".
5. Subject´s death during trial participation.
6. Protocol deviation: A protocol deviation is any change, divergence, or departure from the trial design or procedures of a trial protocol. The subject may remain in the trial unless continuation in the trial jeopardizes the subject's health, safety or rights (see Section 7.4).
7. Pregnancy: Any subject who, despite the requirement for adequate contraception, becomes pregnant during the trial will not receive further trial vaccine administrations. Pregnant subjects should, however, be asked to continue participating in the trial contributing data to the safety follow-up according to protocol (i.e., the pregnant subject will not participate in any further trial interventions [e.g., blood collection], except for safety follow-up if the subject agrees). In addition, the site should maintain contact with the pregnant subject and complete a "Pregnancy Form" as soon as possible. The subject should be followed-up until the birth of the child, or spontaneous or voluntary termination; when pregnancy outcome information

becomes available, the information should be captured using the same form. Data obtained from the "Pregnancy Form" will be captured in the safety database.

8. Receipt of any other dengue vaccines (investigational or licensed) during the trial.
9. Other

   Note: The specific reasons should be recorded in the "specify" field of the eCRF.

8.0 CLINICAL TRIAL MATERIAL MANAGEMENT

This section contains information regarding all vaccines and materials provided directly by the Sponsor, and/or sourced by other means, that are required by the trial protocol, including important sections describing the management of clinical trial material.

8.1 Trial Vaccines and Materials

TDV

The investigational vaccine is TDV, a tetravalent dengue vaccine comprised of 1 molecularly characterized, attenuated dengue virus strain, and 3 chimeric dengue virus strains with CCI [redacted] TDV-1, TDV-2, TDV-3, and TDV-4, respectively. TDV is a lyophilized vaccine that will be reconstituted in diluent (37 mM NaCl solution) prior to administration.

Licensed YF-17D vaccine

The licensed YF-17D vaccine (Sanofi Pasteur) is a live attenuated YF vaccine containing CCI [redacted] of the YF virus. YF- 17D vaccine is lyophilized and will be reconstituted in diluent (NaCl for injection) prior to administration.

Placebo

Placebo is a normal saline solution (0.9% NaCl) for injection.

8.1.1 Dosage Form, Packaging, and Labeling

TDV kits (TDV and TDV diluent):

Manufacturing of monovalent bulk vaccine substances of TDV, mixing of the 4 TDV vaccine substances, filling into vials, and lyophilization of TDV is done at CCI [redacted]

Lyophilized TDV is presented in a single-dose 2CCI [redacted]

TDV diluent (37 mM NaCl solution) is manufactured by CCI [redacted]

TDV and TDV diluent vials are packaged together into single dose dispensing cartons. The vials will be labeled to contain pertinent trial information in local languages.

Licensed YF-17D vaccine and diluent:

Lyophilized YF-17D vaccine and YF-17D vaccine diluent (NaCl for injection) are manufactured by Sanofi Pasteur.

Lyophilized YF-17D vaccine is presented in a single-dose vial with a stopper and flip-off cap.

The diluent (0.5 mL) is provided in a single-use pre-filled syringe.

Placebo:

Commercially available normal saline solution for injection (0.9% NaCl) will be used as placebo. The placebo is presented in single dose units for 0.5 mL dosing.

The Sponsor will supply study sites with packaged and labeled TDV, TDV diluent, YF-17D vaccine, YF-17D vaccine diluent and placebo. The labels will contain pertinent trial information in local languages. Further details can be found in the Pharmacy Manual.

8.1.2 Storage

TDV, YF-17D vaccine, TDV diluent, and YF-17D vaccine diluent will be shipped in refrigerated containers at 2°C to 8°C. From receipt and prior to use, TDV, YF-17D vaccine,TDV diluent, and YF-17D vaccine diluent must be stored at 2°C to 8°C in a refrigerator, do not freeze. TDV and YF-17D vaccine must also be protected from light.

The placebo will be shipped in ambient conditions and should be stored per the manufacturer's label.

All clinical trial material must be kept in an appropriate, limited-access, secure place until it is used or returned to the Sponsor or designee for destruction. All Sponsor-supplied vaccines must be stored under the conditions specified on the label, and remain in the original container until dispensed. A daily temperature log of the vaccine storage area must be maintained every working day. Temperature excursions must be reported to the Sponsor as soon as possible and use of these trial vaccines and diluents requires Sponsor approval.

8.1.3 Dose and Regimen

Trial vaccine doses that will be provided to each trial group are presented in Table 8.a.

The 0.5 mL trial vaccine doses will be prepared and administered by an unblinded pharmacist or vaccine administrator according to the instructions in the Pharmacy Manual or per Sponsor instructions.

TDV, YF-17D vaccine, and placebo will be administered by the SC route.

Table 8.a Sponsor-Supplied Vaccines and Placebo

Trial Group	Number of subjects	Day 1 (M0)	Day 90 (M3)	Day 180 (M6)
Group 1	300	YF-17D vaccine+placebo	TDV	TDV
Group 2	300	TDV+placebo	TDV	YF-17D vaccine
Group 3	300	TDV+YF-17D vaccine	TDV	Placebo

Note: Concomitantly administered trial vaccines will be injected in opposite arms.

8.2 Trial Vaccine Assignment and Dispensing Procedures

The vaccine to be used will be identifiable by a unique identification number and managed by Interactive Response Technology (IRT). Refer to Section 8.6 for accountability of Sponsor-supplied vaccines.

The Investigator or designee will use IRT at subject enrollment to obtain the subject number. This number will be used throughout the trial.

The Investigator or designee will use IRT on the day of first dosing (Day 1 [M0]) to provide the necessary subject identifying information.

The Investigator or designee will use IRT at each dispensing visit to obtain the vaccination identification number for the vaccine dose.

The Investigator's designee(s) will be responsible for overseeing the administration of vaccine to subjects enrolled in the trial according to the procedures stipulated in this trial protocol. The vaccine will be administered only by an unblinded pharmacist or vaccine administrator who is qualified to perform that function under applicable laws and regulations for that specific trial.

If Sponsor-supplied vaccine is lost or damaged, the site can request a replacement. Expired vaccines must not be administered.

Prior to vaccination, a subject must be determined to be eligible for trial vaccination and it must be clinically appropriate in the judgment of the Investigator to vaccinate. Eligibility for vaccination prior to first trial vaccine administration is determined by evaluating the entry criteria outlined in this protocol (Section 7.1 and Section 7.2).

Eligibility for subsequent trial vaccination is determined by the criteria outlined in Section 7.3.

Trial vaccines should not be administered to individuals with known hypersensitivity to any component of the vaccines.

Standard immunization practices are to be observed and care should be taken to administer the injection subcutaneously. In addition, the WHO recommendations to reduce anxiety and pain at the time of vaccination should be followed [19]. Before administering the vaccine, the vaccination site is to be disinfected with a skin disinfectant (e.g., 70% alcohol). Allow the skin to dry. DO NOT inject intravascularly.

As with all injectable vaccines, trained medical personnel and appropriate medical treatment should be readily available in case of anaphylactic reactions following vaccination. For example, epinephrine 1:1000, diphenhydramine, and/or other medications for treating anaphylaxis should be available. These rescue medications will not be supplied by the Sponsor.

8.3 Randomization Code Creation and Storage

The Sponsor or designee's randomization personnel will generate the randomization schedule. Randomization information will be stored in a secured area, accessible only by authorized personnel.

8.4 Trial Vaccine Blind Maintenance

The trial will be conducted in an observer-blind manner; i.e., during the course of the trial, the subject and those responsible for the evaluation of any trial endpoint will all be unaware of which trial vaccine(s) was administered.

The Investigator is responsible for the conduct of the trial according to the protocol, however, they will be unaware of which trial vaccine(s) was administered. The unblinded Investigator designee(s) will be responsible for preparation, administration, and accountability of trial vaccines as specified within the Site Responsibility Delegation Log. The trial vaccines will be prepared and administered by an unblinded pharmacist or vaccine administrator according to the instructions in the Pharmacy Manual or per Sponsor instructions (see also Section 8.2). The pharmacist (or designated individual) at each site will be responsible for vaccine accountability (see also Section 8.6). These unblinded designees will maintain the trial vaccine blind and will have no role in the assessment of subject safety.

8.5 Unblinding Procedure

The trial vaccine blind shall not be broken by the Investigator unless information concerning the trial vaccine(s) is necessary for the medical treatment of the subject or in case of pregnancy if requested by the subject. In the event of a medical emergency or pregnancy, if possible, the medical monitor should be contacted before the trial vaccine blind is broken to discuss the need for unblinding.

For unblinding a subject, the trial vaccine blind can be obtained by the Investigator, by accessing IRT.

The Sponsor's Pharmacovigilance Department must be notified as soon as possible if the trial vaccine blind is broken by the Investigator and the completed SAE or pregnancy form must be sent within 24 hours (Section 10.4.4 and Section 9.1.9, respectively). The date, time, and reason the blind is broken must be recorded in the source document and the same information (except the time) must be recorded on the eCRF.

If any subject is unblinded, no further dose(s) of trial vaccine are to be administered and the subject must be withdrawn from the trial.

8.6 Accountability and Destruction of Sponsor-Supplied Vaccine(s)

Vaccine supplies will be counted and reconciled at the site before being locally destroyed or returned to the Sponsor or designee as noted below. Sites will maintain source documents in addition to entering data in IRT.

The Investigator or designee must ensure that the Sponsor-supplied vaccines (TDV, YF-17D vaccine, and placebo) are used in accordance with the approved protocol and are administered only to subjects enrolled in the trial. To document appropriate use of Sponsor-supplied vaccines (TDV, YF-17D vaccine, and placebo), the Investigator or designee must maintain records of all Sponsor-supplied vaccine delivery to the site, site inventory, administration and use by each subject, and return to the Sponsor or designee.

Upon receipt of Sponsor-supplied vaccines the Investigator or designee must verify the contents of the shipments against the packing list. The verifier should ensure that the quantity is correct, the vaccines are received within the labeled storage conditions (i.e., no cold chain break has occurred during transit), and are in good condition. If quantity and conditions are acceptable, the Investigator or designees will record receipt of the shipment in the IRT.

If there are any discrepancies between the packing list and the actual product received, the Sponsor or designee must be contacted to resolve the issue. The packing list should be filed in the Investigator's essential document file by a qualified Investigator designee.

The pharmacist (or designated individual) at each site must maintain 100% accountability for all Sponsor-supplied vaccines received and administered during their entire participation in the trial. Proper vaccine accountability includes, but is not limited to:

- Verifying that the actual inventory matches the documented inventory.
- Verifying that the log is completed for the vaccine lot number used to prepare each dose.
- Verifying that all containers used are documented accurately on the log.
- Verifying that required fields are completed accurately and legibly.

If any dispensing errors or discrepancies are discovered, the Sponsor must be notified immediately.

The pharmacist (or designated individual) at each site must record the current inventory of all Sponsor-supplied vaccines (TDV, YF-17D vaccine, and placebo) including TDV diluent and YF-17D vaccine diluent on a Sponsor-approved vaccine accountability log. The following information will be recorded at a minimum: protocol number and title, name of Investigator, site identifier and number, description of Sponsor-supplied vaccines, expiry date and/or retest date, and amount. The log (IRT) should include all required information as a separate entry for each subject to whom Sponsor-supplied vaccine is administered.

Prior to site closure or at appropriate intervals throughout the trial, before any clinical trial materials are destroyed locally or returned to the Sponsor or its designee for destruction, a representative from the Sponsor or its designee will perform clinical trial material accountability and reconciliation. The Investigator will retain a copy of the documentation regarding clinical trial material accountability, return, and/or destruction, and originals will be sent to the Sponsor or designee.

The pharmacist (or designated individual) at each site will be responsible for vaccine accountability and will document receipt, use, return, or destruction of trial vaccines (TDV, YF-17D vaccine, and placebo) including TDV diluent and YF-17D vaccine diluent.

Vaccine accountability documentation will be reviewed by the unblinded monitor during clinical monitoring visits.

9.0 TRIAL PLAN

9.1 Trial Procedures

The following sections describe the trial procedures and data to be collected. For each procedure, subjects are to be assessed by the same Investigator or site personnel whenever possible. The Schedule of Trial Procedures is presented in Section 2.1.

9.1.1 Informed Consent Form

The requirements of the informed consent form are described in Section 15.2.

Informed consent must be obtained prior to the subject entering into the trial, and before any protocol-directed procedures are performed.

A unique subject number from IRT will be assigned to each subject after informed consent is obtained. If all eligibility criteria are fulfilled, this subject number will be used throughout the trial. Subject numbers assigned to subjects who fail screening should not be reused (see Section 9.1.10).

9.1.2 Demographics, Medical History and Prior Medications

Demographic information to be obtained will include date of birth, sex, race and ethnicity as described by the subject.

Medical history will also be collected, including but not limited to any medical history that may be relevant to subject eligibility for trial participation, such as prior medications/vaccinations, concomitant medications/vaccinations, and previous and ongoing illnesses or injuries. Relevant medical history can also include any medical history that contributes to the understanding of an AE that occurs during trial participation, if it represents an exacerbation of an underlying disease/pre-existing problem.

Any other vaccination against any flavivirus (licensed or investigational, including any other dengue vaccine) during the entire trial period will be recorded in the eCRF irrespective of time of administration, and including the vaccine type.

All concomitant medications and vaccine history from 1 month (minimum 28 days) prior to administration of each dose of trial vaccine(s) up to 1 month (minimum 28 days) thereafter, steroids and immunostimulants within 60 days prior to Day 1 (M0), immunoglobulins and blood products within 3 months prior to Day 1 (M0), and immunosuppressive therapy within 6 months prior to Day 1 (M0) are to be recorded on the "Prior and Concomitant Medications" eCRF page and in the subject's source document. The use of antipyretics and/or analgesic medications within 24 hours prior to vaccination must be identified and the reason for their use (prophylaxis versus treatment) must be described in the source documents or the eCRF. Trial vaccination should be delayed if subjects have used antipyretics and/or analgesic medication for either reason within 24 hours prior to vaccine administration.

Medications taken for prophylaxis are those intended to prevent the onset of AEs following vaccination. Medications taken for treatment are intended to reduce or eliminate the presence of symptoms that are present.

Prohibited Therapies (see also Section 7.2):

- Parenteral immunoglobulin preparation, blood products, and/or blood-derived products within the 3 months prior to Day 1 (M0).
- Immunosuppressive therapy within 6 months or systemic (e.g., oral or parenteral) corticosteroid treatment within 60 days prior to Day 1 (M0) or immunostimulants within 60 days prior to Day 1 (M0).
- Any other vaccines within 14 days (for inactivated vaccines) or 28 days (for live vaccines) prior to Day 1 (M0), Day 90 (M3) and Day 180 (M6), and 28 days after each trial vaccination.
- Any other dengue vaccines (investigational or licensed) for the entire trial period.
- Receipt of any other clinical trial product for the entire trial period.

These data must be written in the source documents.

Medical history (including corresponding medication) to be obtained will include any significant conditions or diseases that have disappeared or resolved at or prior to signing of the informed consent form.

9.1.3 Documentation of Trial Entrance/Randomization

Only subjects who have a signed informed consent form, meet all of the inclusion criteria and none of the exclusion criteria are eligible for randomization into the vaccination phase. The randomization schedule will be created and controlled by the IRT provider. The randomization specification will be approved by the Sponsor's trial statistician or designee.

If the subject is found to be ineligible for the randomization/vaccination phase, the Investigator should record the primary reason for failure on the subject enrollment log.

9.1.4 Physical Examination

Physical examinations must be performed by a qualified health professional in accordance with local regulations and licensing requirements designated within the Site Responsibility Delegation Log. Complete physical examination will be performed prior to vaccination on Day 1 (M0), Day 90 (M3), and Day 180 (M6). A complete physical examination includes but is not limited to: auscultation of heart and lungs, palpation of the abdomen, inspection of extremities (including skin over intended vaccination site[s]), review of systems, a check of general appearance and the measurement of weight and height (measurement of height is only required at Day 1 [M0]); BMI will be calculated automatically. Additional physical examinations may be performed if indicated by review of the subject's medical history. The findings should be documented in the subject's source document.

Targeted physical examination including but not limited to measurement of vital signs (see Section 9.1.5) will be performed at Day 30 (M1), Day 120 (M4), Day 210 (M7) and Day 360 (M12). Clinical significant changes from the Baseline assessment must be recorded in the subject's source documents and the eCRF.

9.1.5 Vital Signs

These will include (but are not limited to) the measurement of systolic blood pressure, diastolic blood pressure, heart rate, and body temperature at all scheduled visits (Day 1 [M0], Day 30[M1], Day 90 [M3], Day 120 [M4], Day180 [M6], Day 210 [M7] and Day 360 [M12]).

9.1.6 Immunogenicity Assessments

Blood samples for the measurement of dengue neutralizing antibodies (MNT_{50}) will be collected at pre-first vaccination (Day 1 [M0]), 1 month post first vaccination (Day 30 [M1]), pre-second vaccination (Day 90 [M3]), 1 month post second vaccination (Day 120 [M4]), pre-third vaccination (Day 180 [M6]), and 1 month post third vaccination (Day 210 [M7]). Blood samples for the measurement of YF neutralizing antibodies (PRNT) will be collected at pre-first vaccination (Day 1 [M0]), 1 month post first vaccination (Day 30 [M1]), pre-third vaccination (Day 180 [M6]), and 1 month post third vaccination (Day 210 [M7]). The maximum volume of blood taken at any single visit is approximately 15 mL, and the approximate total volume of blood for the trial is maximum 70 ml. Refer also to Appendix D.

All samples will be collected in accordance with acceptable laboratory procedures. Blood samples will be processed and stored at the trial site according to the Laboratory Guidelines as provided in the Laboratory Manual.

9.1.7 Safety Assessments

Safety assessments will include collection and recording of solicited local (injection site) and systemic AEs, unsolicited AEs (non-serious AEs and SAEs), and MAAEs. Refer to Section 10.1 for safety definitions. Details on collection and reporting of AEs are in Section 10.4.

9.1.8 Contraception and Pregnancy Avoidance Procedure

For female subjects of childbearing potential, serum or urine pregnancy testing will be performed prior to vaccination at Day 1 (M0), Day 90 (M3), and Day 180 (M6). Additional pregnancy tests may be performed during the trial if deemed necessary by the Investigator. Females of childbearing potential who are sexually active will be reminded during trial visits to adhere to acceptable contraceptive methods up to 6 weeks post third vaccination at Day 180 (M6). Subjects will be provided with information on acceptable methods of contraception and will be asked prior to vaccination on Day 1 (M0) to sign the informed consent form stating that they understand the requirements for avoidance of pregnancy and donation of ova. During the course of the trial, subjects of childbearing potential will receive continued guidance with respect to the avoidance of pregnancy and donation of ova as part of the trial procedures (Section 2.1); refer also to Section 7.2.

9.1.9 Pregnancy

To ensure subject safety and the safety of the unborn child, each pregnancy in a subject having received a trial vaccine must be reported to the Sponsor within 24 hours of the site learning of its occurrence. If the subject becomes pregnant during the trial, she will not receive any further doses of any Sponsor-supplied trial vaccine. The pregnancy must be followed to determine outcome, including spontaneous or voluntary termination, details of birth, and the presence or absence of any birth defects, congenital abnormalities, or maternal and/or newborn complications. This follow-up should occur even if the intended duration of safety follow-up for the trial has ended. If the trial is still ongoing at the time of birth or termination, the women should be further followed up for safety until the end of the trial.

Any pregnancy occurring following trial vaccine administration should be reported immediately, using the pregnancy form, to the contact listed in the Investigator Site File.

The Investigator must inform the subject of their right to receive trial vaccine information. If the subject chooses to receive unblinded trial vaccine information, the individual blind should be broken by the Investigator and procedures must be followed as described in Section 8.5.

9.1.10 Documentation of Subjects who are not Randomized

Investigators must account for all subjects who sign an informed consent form. If the subject is found to be not eligible at Day 1 (M0), the Investigator should complete the eCRF. The IRT supplier should be contacted as a notification of non-randomization.

The primary reason for non-randomization is to be recorded in the eCRF using the following categories:

- Screen failure (did not meet one or more inclusion criteria or met one or more exclusion criteria).
- Withdrawal by subject.
- Trial terminated by Sponsor.

Subject numbers assigned to subjects who fail screening should not be reused.

9.2 Monitoring Subject Trial Vaccine Compliance

The Investigator records all injections of trial vaccine given to the subject in the eCRF.

9.3 Schedule of Observations and Procedures

The schedule for all trial-related procedures for all evaluations is shown in Section 2.1. Assessments should be completed at the designated visit/time point(s).

9.3.1 Pre-Vaccination Procedures (Day 1 [M0], Day 90 [M3], and Day 180 [M6])

1. Before performing any other trial procedure, the signed informed consent needs to be obtained (Day 1 [M0]). Refer to Section 9.1.1.

2. Check inclusion and exclusion criteria (Day 1 [M0]). Refer to Section 7.1 and Section 7.2.
3. Collect demographic data (Day 1 [M0]). Refer to Section 9.1.2.
4. Collect medical history (Day 1 [M0]). Refer to Section 9.1.2.
5. Collect concomitant medications/vaccinations. Refer to Section 9.1.2.
6. Perform a complete physical examination. Refer to Section 9.1.4.
7. Check vital signs. Refer to Section 9.1.5.
8. Review of systems: Review of systems is a structured interview that queries the subject as to any complaints the subject has experienced across each organ system. Refer to Section 9.1.4.
9. Perform pregnancy testing (serum or urine) for female subjects of childbearing potential. Refer to Section 9.1.8 and Section 9.1.9.
10. Randomize subject (Day 1 [M0]). Refer to Section 9.1.3.
11. Collect pre-vaccination blood sample(s) from all subjects. Refer to Section 9.1.6.

    Blood should be taken from the subject using an aseptic venipuncture technique for serological immunogenicity testing. Refer to the detailed collection and handling procedures outlined in the Laboratory Manual.

9.3.2 Vaccination Procedures (Day 1 [M0], Day 90 [M3], and Day 180 [M6])

1. Check criteria for delay of trial vaccination (Day 90 [M3] and Day 180 [M6]). Refer to Section 7.3.
2. Check contraindications to trial vaccination (Day 90 [M3] and Day 180 [M6]). Refer to Section 7.3.
3. Vaccinate the subject. Refer to Section 8.1.3.

9.3.3 Post-Vaccination Procedures (Day 1 [M0], Day 90 [M3], and Day 180 [M6])

1. Careful training of the subject on how to measure solicited local (injection site) AEs and body temperature, how to complete the diary card and how often to complete the diary card. Training should be directed at the individual(s) who will perform the measurements of local AEs and those who will enter the information into the diary card. This individual may or may not be the subject, but if a person other than the subject enters information into the diary card, this person's identity must be documented in the trial file and this person must receive training on the diary card. Training of the subject on how to measure an injection site AE should be performed while the subject is under 30-minute observation after vaccination.

   Diary card instructions must include the following:

   - The subject must understand that timely completion of the diary card on a daily basis is a critical component of trial participation. In case a paper diary card is used, the subject should also be instructed to write clearly and to complete the diary card in pen. Any

corrections to the paper diary card that are performed by the person completing the diary card should include a single strikethrough line with a brief explanation for any change and be initialed and dated.

Please note:

Diary cards will be the only source document allowed for remote collection of solicited local (injection site) AEs and systemic AEs (including body temperature measurements). The following additional rules apply to the documentation of safety information collected by diary card:

- The diary card should be reviewed with the subject.
- No corrections or additions to the diary card will be allowed after it is reviewed with the Investigator/designee.
- Any data that are identified as implausible or incorrect, and confirmed by the subject to be a transcription error should be corrected by the subject on the diary card (for paper diary cards, the correction should include a single strikethrough line and should be initialed and dated by the subject).
- Any blank or illegible fields on the diary card not otherwise corrected as above will be missing in the eCRF.
- The site must enter all readable entries on the paper diary card into the eCRF.
- Any newly described solicited safety information should be added to the diary card by the subject (for paper diary, the new information should be initialed and dated). Any new unsolicited safety information would be recorded in the subject's source document as a verbally reported event and therefore captured as an AE and recorded in the Adverse Event eCRF.
- Starting on the day of vaccination, the subject will record specific types of events at the injection site (solicited local AEs), any specific generalized symptoms (solicited systemic AEs), daily body temperature (any method), any other symptoms or change in the subject's health status, and any medications taken (excluding vitamins and minerals). These solicited AEs and body temperature will be recorded on the diary card. Assessments should preferably take place in the evening at day's end.
- Body temperature measurement is to be performed using the thermometer provided by the site. If the subject feels unusually hot or cold during the day, the subject should check their temperature. The highest body temperature observed that day should be recorded on the diary card.
- The measurement of solicited local (injection site) AEs (pain, erythema, and swelling) is to be performed using the ruler provided by the site.

2. The collection on the diary card of solicited local (injection site) AEs, and solicited systemic AEs (including body temperature) will continue for a total of 7 days and 14 days, respectively,

following each trial vaccination. Persistent/prolonged solicited local or systemic AEs continuing on Day 8 and Day 15 respectively, following each trial vaccination will be recorded in the subject source document as a verbally reported event and therefore captured as an AE and recorded in the Adverse Event eCRF. Refer to Section 10.4.2.

3. Provide pregnancy avoidance counseling. Refer to Section 9.1.8.
4. Collect and record solicited AEs. Refer to Section 10.1.2.
5. Collect and record unsolicited AEs. Refer to Section 10.4.1.
6. Collect and record MAAEs. Refer to Section 10.4.3.
7. Collect and record SAEs. Refer to Section 10.4.4.
8. Collect and record AEs leading to subject discontinuation or withdrawal. Refer to Section 10.4.1.

After each trial vaccination, the subject will be observed for at least 30 minutes including observation for unsolicited AEs (non-serious and serious), solicited local (injection site) and systemic AEs, and body temperature measurement. Information should be recorded in the electronic data capture system. The Investigator or delegate will take the opportunity to remind the subject how to measure solicited AEs and body temperature as part of this observation period. All safety data will be collected in the subject's source documents.

The site should schedule the next trial activity with the subject.

The subject will receive a written reminder of the next planned trial activity. The subject will be reminded to complete the diary card daily and to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject becomes pregnant or has a medical condition that leads to a hospitalization or an emergency room visit or is otherwise perceived as serious. All contact details will be provided to the subject.

9.3.4 Clinic Visits after Vaccination (Day 30 [M1], Day 120 [M4], Day 210 [M7], and Day 360 [M12])

1. Collect and review the diary card (Day 30 [M1], Day 120 [M4], and Day 210 [M7]).
2. Collect concomitant medications/vaccinations. Refer to Section 9.1.2.
3. Perform a targeted physical examination. Refer to Section 9.1.4.
4. Check vital signs. Refer to Section 9.1.5.
5. Provide pregnancy avoidance counseling (Day 30 [M1], Day 120 [M4], and Day 210 [M7]). Refer to Section 9.1.8.
6. Record persistent/prolonged solicited AEs (Day 30 [M1], Day 120 [M4], and Day 210 [M7]). Refer to Sections 9.3.3 and 10.4.2.
7. Collect and record unsolicited AEs (Day 30 [M1], Day 120 [M4], and Day 210 [M7]). Refer to Section 10.4.1.

8. Collect and record MAAEs. Refer to Section 10.4.3.
9. Collect and record SAEs. Refer to Section 10.4.4.
10. Collect and record AEs leading to subject discontinuation or withdrawal. Refer to Section 10.4.1.
11. Collect blood sample(s) from all subjects (Day 30 [M1], Day 120 [M4], and Day 210 [M7]). Refer to Section 9.1.6.

Blood should be taken from the subject using an aseptic venipuncture technique for serological immunogenicity testing. Refer to the detailed collection and handling procedures outlined in the Laboratory Manual.

The site should schedule the next trial activity with the subject (Day 30 [M1], Day 120 [M4], and Day 210 [M7]).

The subject will receive a written reminder of the next planned trial activity, as applicable. The subject will be reminded to complete the diary card daily and to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject becomes pregnant or has a medical condition that leads to a hospitalization or an emergency room visit or is otherwise perceived as serious. All contact details will be provided to the subject.

9.3.5 Final (End of Trial) Visit

The Final Visit will be performed at Day 360 (M12). If a subject terminates earlier, end of trial visit procedures should be performed if possible. For all subjects receiving trial vaccine or placebo, the Investigator must complete the End of Trial eCRF page.

9.3.6 Post-Trial Care

No post-trial care will be provided.

9.4 Biological Sample Retention and Destruction

In this trial, specimens for immune response testing will be collected as described in Section 9.1.6. After blood draw and serum processing, the serum samples will be preserved and retained at a central laboratory contracted by the Sponsor for this purpose for up to but no longer than 20 years or as required by applicable law. The Sponsor has put into place a system to protect the subject's personal information to ensure optimal confidentiality and defined standard processes for sample and data collection, storage, analysis, and destruction.

Serum samples will be used for the analyses defined in this protocol, but could also, with permission from the subject, be used to assess, improve or develop tests related to dengue and other disease(s) or the vaccine(s) under trial that will allow more reliable measurement of the response to the vaccine(s).

10.0 ADVERSE EVENTS

10.1 Definitions

10.1.1 Adverse Events

An AE is defined as any untoward medical occurrence in a clinical investigation subject administered a trial vaccine; it does not necessarily have to have a causal relationship with trial vaccine administration.

An AE can therefore be any unfavorable and unintended sign (e.g., a clinically significant abnormal laboratory finding), symptom, or disease temporally associated with the administration of a trial vaccine whether or not it is considered related to the trial vaccine.

AEs will be graded by the Investigator in the following manner:

Mild	Grade 1	Awareness of symptoms that are easily tolerated, causing minimal discomfort and not interfering with everyday activities. Relieved with or without symptomatic treatment.
Moderate	Grade 2	Sufficient discomfort is present to cause interference with normal activity. Only partially relieved with symptomatic treatment.
Severe	Grade 3	Extreme distress, causing significant impairment of functioning or incapacitation. Prevents normal everyday activities. Not relieved with symptomatic treatment.

10.1.2 Solicited Adverse Events

The occurrence of selected indicators of safety as listed in Table 10.a will be measured/collected for 7 days (solicited local [injection site] AEs) and 14 days (solicited systemic AEs) following each trial vaccination (including the day of vaccination) and will be recorded on the eCRF, as applicable. These will be summarized in the final report under the category "solicited AEs" to differentiate them from other AEs which were not solicited. Any solicited local or systemic AE observed as continuing on Day 8 and Day 15, respectively, following each trial vaccination will be recorded as an AE on the Adverse Event eCRF for follow-up. For these persistent/prolonged solicited AEs the end date will be captured on the Adverse Event eCRF to permit a separate analysis from the unsolicited AEs (see Section 10.4.2).

Table 10.a Solicited Local and Systemic AEs

Solicited local (injection site) AEs:	Pain Erythema Swelling
Solicited systemic AEs:	Fever [(a)] Headache Asthenia Malaise Myalgia

(a) Fever is defined as greater than or equal to 38°C (100.4°F) regardless of method taken [20].

The severity of solicited safety parameters will be assessed as described in Table 10.b.

Table 10.b Severity Scales for Solicited Safety Parameters

Adverse Event	Severity grade	Severity
Pain at injection site	0	None
	1	Mild: No interference with daily activity
	2	Moderate: Interference with daily activity with or without treatment
	3	Severe: Prevents daily activity with or without treatment
Erythema at injection site [(a)]	0	<25 mm
	1	Mild: ≥25 – ≤50 mm
	2	Moderate: >50–≤100 mm
	3	Severe: >100 mm
Swelling at injection site [(a)]	0	<25 mm
	1	Mild: ≥25 – ≤50 mm
	2	Moderate: >50-≤100 mm
	3	Severe: >100 mm
Headache	0	None
	1	Mild: No interference with daily activity
	2	Moderate: Interference with daily activity with or without treatment
	3	Severe: Prevents normal activity with or without treatment
Asthenia	0	None
	1	Mild: No interference with daily activity
	2	Moderate: Interference with daily activity
	3	Severe: Prevents daily activity
Malaise	0	None
	1	Mild: No interference with daily activity
	2	Moderate: Interference with daily activity
	3	Severe: Prevents daily activity
Myalgia	0	None
	1	Mild: No interference with daily activity
	2	Moderate: Interference with daily activity
	3	Severe: Prevents daily activity
Fever [(b)]	Record body temperature in °C/°F	

(a) Subjects are to record greatest surface diameter in mm on the diary card.
(b) Fever is defined as greater than or equal to 38°C (100.4°F) regardless of method taken [20].

10.1.3 Medically Attended Adverse Events

MAAEs are defined as AEs leading to an unscheduled visit to or by a healthcare professional including visits to an emergency department, but not fulfilling seriousness criteria.

10.1.4 Serious Adverse Events

An SAE is defined as any untoward medical occurrence that:

1. Results in DEATH.
2. Is LIFE THREATENING.
    - The term "life threatening" refers to an event in which the subject was at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death if it were more severe.
3. Requires inpatient HOSPITALIZATION or prolongation of existing hospitalization.
4. Results in persistent or significant DISABILITY/INCAPACITY.
5. Leads to a CONGENITAL ANOMALY/BIRTH DEFECT in the offspring of a subject.
6. Is an IMPORTANT MEDICAL EVENT that satisfies any of the following:
    - May require intervention to prevent items 1 through 5 above.
    - May expose the subject to danger, even though the event is not immediately life threatening or fatal or does not result in hospitalization.

10.2 Causality of Adverse Events

Relatedness (causality) to vaccine will also be assessed by the Investigator. The relationship of each AE, including solicited systemic AEs (all solicited local AEs are considered as related) to trial vaccine(s) will be assessed using the following categories:

Related:	There is suspicion that there is a relationship between the trial vaccine(s) and the AE (without determining the extent of probability); there is a reasonable possibility that the trial vaccine(s) contributed to the AE.
Not Related:	There is no suspicion that there is a relationship between the trial vaccine(s) and the AE; there are other more likely causes and administration of the trial vaccine(s) is not suspected to have contributed to the AE.

10.2.1 Relationship to Trial Procedures

Relationship (causality) to trial procedures should be determined for all AEs.

The relationship should be assessed as "Yes" if the Investigator considers that there is reasonable possibility that an event is due to a trial procedure. Otherwise, the relationship should be assessed as "No".

10.2.2 Outcome of Adverse Events

Resolved:	The subject has fully recovered from the event or the condition has returned to the level observed at Baseline.
Resolving:	The event is improving but the subject is still not fully recovered.
Not resolved:	The event is ongoing at the time of reporting and the subject has still not recovered.
Resolved with sequelae:	As a result of the AE, the subject suffered persistent and significant disability/incapacity (e.g., became blind, deaf or paralysed).
Fatal:	The subject died due to the event. If the subject died due to other circumstances than the event, the outcome of the event per se should be stated otherwise (e.g., not resolved or resolving).
Unknown:	If outcome is not known or not reported.

10.3 Additional Points to Consider for Adverse Events

An untoward occurrence generally may:

- Indicate a new diagnosis or unexpected worsening of a pre-existing condition. Intermittent events for pre-existing conditions or underlying disease should not be considered as AEs.
- Necessitate therapeutic intervention.
- Require an invasive diagnostic procedure.
- Require vaccine discontinuation or a change in concomitant medication.
- Be considered unfavorable by the Investigator for any reason.

Diagnoses vs. signs and symptoms:

- Each event should be recorded to represent a single diagnosis. Accompanying signs (including abnormal laboratory values) or symptoms should NOT be recorded as additional AEs. If a diagnosis is unknown, signs or symptoms should be recorded appropriately as AEs.

Worsening of AEs:

- If the subject experiences a worsening or complication of an AE after starting administration of the trial vaccine(s), the worsening or complication should be recorded as a new AE. Investigators should ensure that the AE term recorded captures the change in the condition (e.g., "worsening of…").
- If the subject experiences a worsening or complication of an AE after any change in trial vaccine(s), the worsening or complication should be recorded as a new AE. Investigators should ensure that the AE term recorded captures the change in the condition (e.g., "worsening of…").

Changes in severity of AEs:

- If the subject experiences changes in severity of an AE, the event should be captured once with the maximum severity recorded.

Preplanned surgeries or procedures:

- Preplanned procedures (surgeries or therapies) that were scheduled prior to signing of the informed consent form are not considered AEs. Complications resulting from any planned surgery should be reported as AEs.

Elective surgeries or procedures:

- Elective procedures performed where there is no change in the subject's medical condition should not be recorded as AEs, but should be documented in the subject's source documents. Complications resulting from an elective surgery should be reported as AEs.

10.4 Procedures

10.4.1 Collection and Reporting of Adverse Events

All AEs, whether considered related with the use of the trial vaccine(s) or not, must be monitored until symptoms subside and any abnormal laboratory values have returned to Baseline, or until there is a satisfactory explanation for the changes observed, or until death, in which case a full pathologist's report should be supplied, if possible. All findings must be reported on an Adverse Event eCRF and on the SAE eCRF form, if necessary (see Section 10.4.2). All findings in subjects experiencing AEs must also be reported in the subject's source documents. Any unsolicited AEs for 28 days (day of vaccination+27 days) following each trial vaccination will be collected during site visits by interview. AEs leading to withdrawal from the study or discontinuation of trial vaccination have to be reported throughout the trial period.

The following information will be documented for each event:

- Reported term for the AE.
- Start and end date.
- Serious (Y/N).
- Severity.
- Investigator's opinion of the causality (relationship) between the event and administration of trial vaccine(s) ("related" or "not related").
- Investigator's opinion of the causality (relationship) to trial procedure(s), including the details of the suspected procedure.
- Action taken with the trial vaccine(s).
- Outcome of event.

10.4.2 Collection and Reporting of Solicited Adverse Events

The occurrence of selected indicators of safety will be collected using diary cards completed by the subjects for 7 days (solicited local [injection site] AEs) and 14 days (solicited systemic AEs) following each trial vaccination (including the day of vaccination) and will be recorded on the "Local and Systemic AE" eCRF, as applicable. These will be summarized in the final report under the category "solicited adverse events" to differentiate them from unsolicited AEs. Any solicited local (injection site) or systemic AE observed as continuing on Day 8 and Day 15, respectively, following each trial vaccination will be additionally recorded as an AE on the Adverse Event eCRF for follow-up. For these persistent/prolonged solicited AEs the end date will be captured on the Adverse Event eCRF to permit a separate analysis from the unsolicited AEs. Any solicited local (injection site) or systemic AE that resolved before 8 days and 15 days, respectively, following each trial vaccination, but which recurs at a later time (i.e., if discontinued), will be recorded as an unsolicited AE on the Adverse Event eCRF.

Any solicited AE that meets any of the following criteria must be entered as an AE on the Adverse Event eCRF:

- Solicited local (injection site) or solicited systemic AEs that lead the subject to withdraw from the trial.
- Solicited local (injection site) or solicited systemic AEs that lead to the subject being withdrawn from the trial by the Investigator.
- Solicited local (injection site) or solicited systemic AEs that otherwise meet the definition of an SAE (see Section 10.1.2).

10.4.3 Collection and Reporting of Medically Attended Adverse Events

MAAEs occurring from first trial vaccination (Day 1 [M0]) until the end of the trial (Day 360 [M12]) will be collected during site visits by interview and must be recorded as an AE on the Adverse Event eCRF.

10.4.4 Collection and Reporting of Serious Adverse Events

Collection of SAEs will commence from the time that the subject is first administered the trial vaccines (Day 1 [M0]). Routine collection of SAEs will continue until the end of the trial (Day 360 [M12]).

SAEs should be reported according to the following procedure:

A Sponsor SAE form must be completed, in English, and signed by the Investigator immediately or within 24 hours of first onset or notification of the event. The information should be completed as fully as possible but contain, at a minimum:

- A short description of the event and the reason why the event is categorized as serious.
- Subject identification number.
- Investigator's name.

- Name of the trial vaccine(s) – if no unblinding is necessary, in a blinded way.
- Causality assessment.

The SAE form should be transmitted within 24 hours to the attention of the contact(s) in the list provided to each site.

Note: For this study, SAE reporting will be performed via eCRF. Only if the electronic data capture system is unavailable, a paper Sponsor SAE form should be completed, signed by the investigator and transmitted within 24 hours. The SAE must be entered into the eCRF once access to the electronic data capture system is restored.

10.5 Follow-up Procedures

10.5.1 Adverse Events

All AEs will be monitored until resolution or a stable status is reached or until a formal diagnosis can be made or until the end of the trial, whichever occurs first. This could potentially be followed outside of this trial or in a planned extension trial.

10.5.2 Serious Adverse Events

If information not available at the time of the first report becomes available at a later date, the Investigator should complete a follow-up SAE form or provide other written documentation and fax it immediately within 24 hours of receipt. Copies of any relevant data from the hospital notes (e.g., laboratory tests, discharge summary, post-mortem results) should be sent to the Sponsor.

All SAEs should be followed up until resolution or permanent outcome of the event or is otherwise explained. The timelines and procedure for follow-up reports are the same as those for the initial report.

10.5.3 Safety Reporting to Investigators, Institutional Review Boards or Independent Ethics Committees, and Regulatory Authorities

The Sponsor or designee will be responsible for the reporting of all suspected unexpected serious adverse reactions (SUSAR) and any other applicable SAEs to regulatory authorities, investigators and IRBs or IECs, as applicable, in accordance with national regulations in the countries where the trial is conducted. Relative to the first awareness of the event by/or further provision to the Sponsor or Sponsor's designee, SUSARs will be submitted within 7 days for fatal and life-threatening events and 15 days for other SUSARs, unless otherwise required by national regulations. The Sponsor will also prepare an expedited report for other safety issues where these might materially alter the current benefit-risk assessment of an investigational medicinal product or that would be sufficient to consider changes in the trial vaccine administration or in the overall conduct of the trial. The investigational site also will forward a copy of all expedited reports to their IRB or IEC in accordance with national regulations.

10.5.4 Post-Trial Events

Any SAE that occurs outside of the protocol-specified observation period or after the end of the trial but considered to be caused by the trial vaccine(s) must be reported to the Sponsor. These SAEs will be processed by the Sponsor's Pharmacovigilance Department. Instructions for how to submit these SAEs will be provided in a handout in the Investigator Site File.

11.0 TRIAL-SPECIFIC REQUIREMENT(S)

11.1 Data Monitoring Committee

A DMC will have oversight of this trial. The DMC functions at a program level and further information is available in the DMC Charter.

12.0 DATA HANDLING AND RECORD KEEPING

The full details of procedures for data handling will be documented in the Data Management Plan. Adverse events, medical history, and concurrent conditions will be coded using the Medical Dictionary for Regulatory Activities (MedDRA, System Organ Class [SOC], High Level Group Term, High Level Term, Low Level Term, Preferred Term [PT], and their corresponding descriptive terms. Drugs will be coded using the WHO Drug Dictionary.

12.1 Electronic Case Report Forms

Completed eCRFs are required for each subject who provides a signed informed consent form.

The Sponsor or its designee will supply investigative sites with access to eCRFs. The Sponsor will make arrangements to train appropriate site staff in the use of the eCRF. Electronic CRFs must be completed in English.

After completion of the entry process, computer logic checks will be run to identify items, such as inconsistent dates, missing data, and questionable values. Queries may be issued by Sponsor personnel (or designees) and will be answered by the site.

Corrections to eCRFs are recorded in an audit trail that captures the old information, the new information, identification of the person making the correction, the date the correction was made, and the reason for change. Reasons for significant corrections should additionally be included.

The Principal Investigator or designee must review the eCRFs for completeness and accuracy and must sign and date the appropriate eCRFs as indicated. Furthermore, the Investigator must retain full responsibility for the accuracy and authenticity of all data entered on the eCRFs.

Electronic CRFs will be reviewed for completeness and acceptability at the trial site during periodic visits by trial monitors. The Sponsor or its designee will be permitted to review the subject's medical and hospital records pertinent to the trial to ensure accuracy of the eCRFs. The completed eCRFs are the sole property of the Sponsor and should not be made available in any form to third parties, except for authorized representatives of appropriate governmental health or regulatory authorities, without written permission of the Sponsor.

12.2 Record Retention

The Investigator agrees to keep the records stipulated in Section 12.0 and those documents that include (but are not limited to) the trial-specific documents, the identification log of all participating subjects, medical records. Temporary media such as thermal sensitive paper should be copied and certified, source worksheets, all original signed and dated informed consent forms, subject authorization forms regarding the use of personal health information (if separate from the informed consent form), electronic copy of eCRFs, including the audit trail, and detailed records of vaccine disposition to enable evaluations or audits from regulatory authorities, the Sponsor or its designees. Furthermore, ICH E6 Section 4.9.5 requires the Investigator to retain essential documents specified in ICH E6 (Section 8) until at least 2 years after the last approval of a marketing application for a specified vaccine indication being investigated or, if an application is not approved, until at least 2 years after the investigation is discontinued and regulatory authorities

are notified. In addition, ICH E6 Section 4.9.5 states that the trial records should be retained until an amount of time specified by applicable regulatory requirements or for a time specified in the Clinical Trial Site Agreement between the Investigator and Sponsor.

Refer to the Clinical Trial Site Agreement for the Sponsor's requirements on record retention. The Investigator should contact and receive written approval from the Sponsor before disposing of any such documents.

13.0 STATISTICAL METHODS

13.1 Statistical and Analytical Plans

A statistical analysis plan (SAP) will be prepared and finalized prior to unblinding of subject's trial vaccine assignment. This document will provide further details regarding the definition of analysis variables and analysis methodology to address all trial objectives.

A blinded data review will be conducted prior to unblinding of subject's trial vaccine assignment. This review will assess the accuracy and completeness of the trial database, subject evaluability, and appropriateness of the planned statistical methods.

13.1.1 Analysis Sets

Safety set: The safety set will consist of all subjects who received at least 1 dose of trial vaccine.

Full analysis set (FAS): The FAS will include all randomized subjects who received at least 1 dose of trial vaccine and for whom a valid pre-dosing and at least 1 post-dosing blood sample have been received for immunogenicity assessments.

Per–protocol set (PPS): The PPS will exclude all subjects seropositive for dengue virus and seroprotected for YF virus at Baseline (seropositivity is defined as a reciprocal neutralizing antibody titer ≥10 for the 4 dengue serotypes; seroprotection is defined as a reciprocal anti-YF neutralizing antibody titer ≥10) and will include all subjects in the FAS who have no major protocol violations. The major protocol violation criteria will be defined as part of the blinded data review prior to the unblinding of subject's trial vaccine assignment. The categories of major protocol violations include: (1) not meeting selected entry criteria, (2) not receiving the planned trial vaccinations or receiving a wrong trial vaccine, (3) receiving prohibited therapies, and (4) other major protocol violations that may be identified during blinded data reviews.

All summaries and analyses of safety data will be based on the safety set. The primary immunogenicity analyses will be based on the PPS; additional immunogenicity analyses may be provided based on the FAS.

13.1.2 Analysis of Demographics and Other Baseline Characteristics

Age, gender, race, and other Baseline characteristics will be summarized descriptively by trial group for all randomized subjects. Unless specified otherwise, number of subjects with non-missing observations, mean, SD, median, minimum and maximum will be presented for continuous data; and frequency and percent will be presented for categorical data.

13.1.3 Immunogenicity Analysis

Descriptive statistics including 95% CI for the primary and secondary endpoints, including seroprotection rates, seropositivity rates and GMTs, will be computed by trial group for all available assays at all relevant time points.

As a primary analysis, the NI of the immune response to the YF-17D vaccine when concomitantly administered with TDV (Group 3) compared to concomitant administration of the YF-17D

vaccine with placebo (Group 1) will be assessed in terms of YF seroprotection rates on Day 30 (M1). NI will be concluded if the upper bound of the 95% CI for the seroprotection rate difference (Group 1 minus Group 3) is less than the NI margin of 5%. The Newcombe score method [21] will be used to compute the 95% CI of the rate difference.

Secondary analyses will be performed as follows:

The NI of the immune response to TDV when concomitantly administered with YF-17D vaccine (Group 3) compared to concomitant administration of TDV with placebo (Group 2) will be assessed in terms of the GMTs of neutralizing antibodies for all 4 dengue serotypes on Day 120 (M4). NI will be concluded if the upper bound of the 95% CI for the GMT ratio (Group 2/Group 3) is less than the NI margin of 2.0. An analysis of variance (ANOVA) model will be used for this assessment.

The NI of the immune response to YF-17D vaccine when concomitantly administered with TDV (Group 3) compared to concomitant administration of YF-17D vaccine with placebo (Group 1) will be assessed in terms of the GMTs of anti-YF neutralizing antibodies on Day 30 (M1). NI will be concluded if the upper bound of the 95% CI for the GMT ratio (Group 1/Group 3) is less than the NI margin of 2.0. An ANOVA model will be used for this assessment.

Handling of missing data and of values below the lower limit of quantification will be described in the SAP.

13.1.4 Safety Analysis

Solicited AEs

In all subjects, the presence and severity (Grade) of solicited local (injection site[s]) AEs (pain, erythema and swelling) and solicited systemic AEs (fever, asthenia, malaise, headache and myalgia) will be collected for 7 days and 14 days respectively, following each vaccination (including the day of vaccination) via collection of diary cards.

For each solicited AE, the number and percentage of subjects with local (injection site[s]) and systemic AEs will be summarized by trial group and event severity for each day after each vaccination (i.e., Day 1 through Day 7 for local [injection site(s)] AEs and Day 1 through Day 14 for systemic AEs), and overall. Summaries of first onset of each event and the number of days subjects reported experiencing each event will also be provided. For subjects with more than 1 episode of the same event, the maximum severity will be used for tabulations.

Persistent/prolonged solicited local or systemic AEs continuing on Day 8 and Day 15, respectively, following each trial vaccination will be assessed separately. Unless otherwise specified these AEs will not be included in the analyses/tabulations of unsolicited AEs and will have separate listings.

Unsolicited AEs

In all subjects, unsolicited AEs will be assessed for 28 days following each vaccination (day of vaccination+27 days).

Unsolicited AEs will be coded using the MedDRA, and summarized by PT and SOC for each trial group.

Unsolicited AEs will be summarized as follows: by PT including events with frequency greater than a pre-defined frequency (the percentage will be specified in the SAP); by SOC and PT; by SOC, PT, and severity; and by SOC, PT, and relationship to the trial vaccine(s). Subjects reporting more than 1 occurrence for the term (level) being summarized will be counted only once.

AEs leading to trial or trial vaccine withdrawal will be collected and summarized for the entire study period.

MAAEs

In all subjects, MAAEs will be collected throughout the trial. MAAEs will be coded using MedDRA, and summarized by PT and SOC for each trial group.

SAEs

In all subjects, SAEs will be collected throughout the trial. SAEs will be coded using MedDRA, and summarized by PT and SOC for each trial group.

13.1.5 Interim Analysis and Criteria for Early Termination

No interim analyses are planned.

13.2 Determination of Sample Size

The immunogenicity set sample size calculation assumes a significance level of 0.025 (one-sided).

For the primary objective of showing NI in YF seroprotection rates, the calculation assumes a NI margin of 5%, and a YF seroprotection rate of 98% at 1 month after YF-17D vaccination in the two trial groups (Group 1 and Group 3).

For the secondary objective of showing NI in GMTs of neutralizing antibodies for all the 4 dengue serotypes, the calculation assumes a NI margin of 2.0, that the true GMT ratio for the two trial groups (Group 2 and Group 3) is 1, and that the natural logarithm of titers are distributed as normal distributions with SDs of 1.35, 0.86, 1.21, and 1.27.

For the secondary objective of showing NI in GMTs of anti-YF neutralizing antibodies, the calculation assumes a NI margin of 2.0, that the true GMT ratio for the two trial groups (Group 1 and Group 3) is 1, and that the natural logarithm of titers are distributed as normal distributions with a SD of 1.31 [22].

A sample size of 300 subjects per trial group, with approximately 255 evaluable subjects per trial group (assuming approximately 15% dropouts and non-evaluable subjects), is sufficient to achieve approximately 90% overall power for showing NI for the above primary and secondary objectives.

A total sample size of 900 subjects also ensures that a sufficient number of healthy YF/DENV-naive adults will be vaccinated to support the safe use of TDV in travelers.

The power calculations were based on nQuery Advisor® 6.01.

14.0 QUALITY CONTROL AND QUALITY ASSURANCE

14.1 Trial-Site Monitoring Visits

Monitoring visits to the trial site will be made periodically during the trial to ensure that all aspects of the protocol are followed. Source documents will be reviewed for verification of data recorded on the eCRFs. Source documents are defined as original documents, data, and records. The Investigator and institution guarantee access to source documents by the Sponsor or its designee (Contract Research Organization) and by the IRB or IEC.

All aspects of the trial and its documentation will be subject to review by the Sponsor or designee (as long as blinding is not jeopardized), including but not limited to the Investigator's Binder, trial vaccine, subject medical records, informed consent form documentation, documentation of subject authorization to use personal health information (if separate from the informed consent forms), and review of eCRFs and associated source documents. It is important that the Investigator and other trial personnel are available during the monitoring visits and that sufficient time is devoted to the process.

14.2 Protocol Deviations

The Investigator should not deviate from the protocol, except where necessary to eliminate an immediate hazard to trial subjects. Should other unexpected circumstances arise that will require deviation from protocol-specified procedures, the Investigator should consult with the medical monitor (and IRB or IEC, as required) to determine the appropriate course of action. There will be no exemptions (a prospective approved deviation) from the inclusion or exclusion criteria.

14.3 Quality Assurance Audits and Regulatory Agency Inspections

The trial site also may be subject to quality assurance audits by the Sponsor or designees. In this circumstance, the Sponsor-designated auditor will contact the site in advance to arrange an auditing visit. The auditor may ask to visit the facilities where laboratory samples are collected, where the vaccine is stored and prepared, and any other facility used during the trial. In addition, there is the possibility that this trial may be inspected by regulatory agencies, including those of foreign governments (e.g., the Food and Drug Administration [FDA], the United Kingdom Medicines and Healthcare Products Regulatory Agency, the Pharmaceuticals and Medical Devices Agency of Japan). If the trial site is contacted for an inspection by a regulatory body, the Sponsor should be notified immediately. The Investigator and institution guarantee access for quality assurance auditors to all trial documents as described in Section 14.1.

15.0 ETHICAL ASPECTS OF THE TRIAL

This trial will be conducted with the highest respect for the individual participants (i.e., subjects) according to the protocol, the ethical principles that have their origin in the Declaration of Helsinki, and the ICH Harmonized Tripartite Guideline for GCP. Each Investigator will conduct the trial according to applicable local or regional regulatory requirements and align his or her conduct in accordance with the "Responsibilities of the Investigator" that are listed in Appendix A. The principles of Helsinki are addressed through the protocol and through appendices containing requirements for informed consent and Investigator responsibilities.

15.1 Institutional Review Board and/or Independent Ethics Committee Approval

IRBs and IECs must be constituted according to the applicable federal/local requirements of each participating region. The Sponsor or designee will require documentation noting all names and titles of members who make up the respective IRB or IEC. If any member of the IRB or IEC has direct participation in this trial, written notification regarding his or her abstinence from voting must also be obtained. Those United States (US) sites unwilling to provide names and titles of all members due to privacy and conflict of interest concerns should instead provide a Federal Wide Assurance Number or comparable number assigned by the Department of Health and Human Services.

The Sponsor or designee will supply relevant documents for submission to the respective IRB or IEC for the protocol's review and approval. This protocol, the Investigator's Brochure, a copy of the informed consent form, and, if applicable, subject recruitment materials and/or advertisements and other documents required by all applicable laws and regulations, must be submitted to a central or local IRB or IEC for approval. The IRB's or IEC's written approval of the protocol and subject informed consent form must be obtained and submitted to the Sponsor or designee before commencement of the trial (i.e., before shipment of the Sponsor-supplied vaccine or trial-specific screening activity). The IRB or IEC approval must refer to the trial by exact protocol title, number, and version date; identify versions of other documents (e.g., informed consent form) reviewed; and state the approval date. The Sponsor will notify the site once the Sponsor has confirmed the adequacy of site regulatory documentation and, when applicable, the Sponsor has received permission from the competent authority to begin the trial. Until the site receives notification no protocol activities, including screening may occur.

Sites must adhere to all requirements stipulated by their respective IRB or IEC. This may include notification to the IRB or IEC regarding protocol amendments, updates to the informed consent form, recruitment materials intended for viewing by subjects, local safety reporting requirements, reports and updates regarding the ongoing review of the trial at intervals specified by the respective IRB or IEC, and submission of the Investigator's final status report to IRB or IEC. All IRB and IEC approvals and relevant documentation for these items must be provided to the Sponsor or its designee.

Subject incentives should not exert undue influence for participation. Payments to subjects must be approved by the IRB or IEC and Sponsor.

15.2 Subject Information, Informed Consent, and Subject Authorization

Written consent documents will embody the elements of informed consent as described in the Declaration of Helsinki and the ICH Guidelines for GCP and will be in accordance with all applicable laws and regulations. The informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) describe the planned and permitted uses, transfers, and disclosures of the subject's personal and personal health information for purposes of conducting the trial. The informed consent form and the subject information sheet (if applicable) further explain the nature of the trial, its objectives, and potential risks and benefits, as well as the date informed consent is given. The informed consent form will detail the requirements of the participant and the fact that he or she is free to withdraw at any time without giving a reason and without prejudice to his or her further medical care.

The Investigator is responsible for the preparation, content, and IRB or IEC approval of the informed consent form and if applicable, the subject authorization form. The informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) must be approved by both the IRB or IEC and the Sponsor prior to use.

The informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) must be written in a language fully comprehensible to the prospective subject. It is the responsibility of the Investigator to explain the detailed elements of the informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) to the subject. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB or IEC.

The subject must be given ample opportunity to: (1) inquire about details of the trial and (2) decide whether or not to participate in the trial. If the subject, determines he or she will participate in the trial, then the informed consent form and subject authorization form (if applicable) must be signed and dated by the subject at the time of consent and prior to the subject entering into the trial. The subject should be instructed to sign using their legal names, not nicknames, using blue or black ballpoint ink. The Investigator must also sign and date the informed consent form and subject authorization (if applicable) at the time of consent and prior to subject entering into the trial; however, the Sponsor may allow a designee of the Investigator to sign to the extent permitted by applicable law.

Once signed, the original informed consent form, subject authorization form (if applicable), and subject information sheet (if applicable) will be stored in the Investigator's site file. The Investigator must document the date the subject signs the informed consent form in the subject's medical record and eCRF. Copies of the signed informed consent form, the signed subject authorization form (if applicable), and subject information sheet (if applicable) shall be given to the subject.

All revised informed consent forms must be reviewed and signed by the relevant subject in the same manner as the original informed consent form. The date the revised consent was obtained should be recorded in the subject's medical record and eCRF, and the subject should receive a copy of the revised informed consent form.

15.3 Subject Confidentiality

The Sponsor and designees affirm and uphold the principle of the subject's right to protection against invasion of privacy. Throughout this trial, a subject's source data will only be linked to the Sponsor's clinical trial database or documentation via a unique identification number. As permitted by all applicable laws and regulations, limited subject attributes, such as sex, age, or date of birth, and subject initials may be used to verify the subject and accuracy of the subject's unique identification number.

To comply with ICH Guidelines for GCP and to verify compliance with this protocol, the Sponsor requires the Investigator to permit its monitor or designee's monitor, representatives from any regulatory authority (e.g., FDA, Medicines and Healthcare products Regulatory Agency, Pharmaceuticals and Medical Devices Agency), the Sponsor's designated auditors, and the appropriate IRBs and IECs to review the subject's original medical records (source data or documents), including, but not limited to, laboratory test result reports, electrocardiogram (ECG) reports, admission and discharge summaries for hospital admissions occurring during a subject's trial participation, and autopsy reports. Access to a subject's original medical records requires the specific authorization of the subject as part of the informed consent form process (see Section 15.2).

Copies of any subject source documents that are provided to the Sponsor must have certain personally identifiable information removed (i.e., subject name, address, and other identifier fields not collected on the subject's eCRF).

15.4 Publication, Disclosure, and Clinical Trial Registration Policy

15.4.1 Publication and Disclosure

The results of this trial are expected to be published in a scientific journal. It is anticipated that clinical and laboratory co-investigators will participate in authorship. The order of authorship and choice of journal will be proposed by the Sponsor to the Principal Investigator(s), to be eventually agreed upon by all authors. The data analysis center for this trial will provide the analyses needed for publication. Information regarding this trial will be posted on ClinicalTrials.gov.

15.4.2 Clinical Trial Registration

In order to ensure that information on clinical trials reaches the public in a timely manner and to comply with applicable law, regulation and guidance, the Sponsor will, at a minimum register all clinical trials conducted in subjects that it sponsors anywhere in the world on ClinicalTrials.gov or other publicly accessible websites before trial initiation. The Sponsor contact information, along with Investigator's city, country, and recruiting status will be registered and available for public viewing.

15.4.3 Clinical Trial Results Disclosure

The Sponsor will post the results of this clinical trial, regardless of outcome, on ClinicalTrials.gov or other publicly accessible websites, as required by applicable laws and/or regulations.

Trial completion corresponds to the date on which the final subject was examined or received an intervention for the purposes of final collection of data for the primary outcome, whether the clinical trial concluded according to the pre-specified protocol or was terminated.

15.5 Insurance and Compensation for Injury

Each subject in the trial must be insured in accordance with the regulations applicable to the site where the subject is participating. If a local underwriter is required, then the Sponsor or Sponsor's designee will obtain clinical trial insurance against the risk of injury to clinical trial subjects. Refer to the Clinical Trial Site Agreement regarding the Sponsor's policy on subject compensation and treatment for injury. If the Investigator has questions regarding this policy, he or she should contact the Sponsor or Sponsor's designee.

16.0 REFERENCES

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APPENDIX A RESPONSIBILITIES OF THE INVESTIGATOR

Clinical research trials sponsored by the Sponsor are subject to ICH GCP and all the applicable local laws and regulations.

The Investigator agrees to assume the following responsibilities:

1. Conduct the trial in accordance with the protocol.
2. Personally conduct or supervise the staff who will assist in the protocol.
3. Ensure that trial-related procedures, including trial-specific (non-routine/non-standard panel) screening assessments are NOT performed on potential subjects, prior to the receipt of written approval from relevant governing bodies/authorities.
4. Ensure that all colleagues and employees assisting in the conduct of the trial are informed of these obligations.
5. Secure prior approval of the trial and any changes by an appropriate IRB/IEC that conform to 21 Code of Federal Regulations (CFR) Part 56, ICH, and local regulatory requirements.
6. Ensure that the IRB/IEC will be responsible for initial review, continuing review, and approval of the protocol. Promptly report to the IRB/IEC all changes in research activity and all anticipated risks to subjects. Make at least yearly reports on the progress of the trial to the IRB/IEC, and issue a final report within 3 months of trial completion.
7. Ensure that requirements for informed consent, as outlined in 21 CFR Part 56, ICH and local regulations, are met.
8. Obtain valid informed consent from each subject who participates in the trial, and document the date of consent in the subject's medical chart. Valid informed consent form is the most current version approved by the IRB/IEC. Each informed consent form should contain a subject authorization Section that describes the uses and disclosures of a subject's personal information (including personal health information) that will take place in connection with the trial. If an informed consent form does not include such a subject authorization, then the Investigator must obtain a separate subject authorization form from each subject.
9. Prepare and maintain adequate case histories of all persons entered into the trial, including eCRFs, hospital records, laboratory results, etc, and maintain these data for a minimum of 2 years following notification by the Sponsor that all investigations have been discontinued or that the regulatory authority has approved the marketing application. The Investigator should contact and receive written approval from the Sponsor before disposing of any such documents.
10. Allow possible inspection and copying by the regulatory authority of GCP-specified essential documents.
11. Maintain current records of the receipt, administration, and disposition of Sponsor-supplied vaccines, and return all unused Sponsor-supplied vaccines to the Sponsor.

12. Report AEs to the Sponsor promptly. In the event of an SAE, notify the Sponsor within 24 hours.
13. Review and provide a signature as approval of the content of the clinical study report.

APPENDIX B INVESTIGATOR CONSENT TO USE OF PERSONAL INFORMATION

Takeda will collect and retain personal information of Investigator, including his or her name, address, and other personally identifiable information. In addition, Investigator's personal information may be transferred to other parties located in countries throughout the world (e.g., the United Kingdom, US, and Japan), including the following:

- Takeda, its affiliates, and licensing partners.
- Business partners assisting Takeda, its affiliates, and licensing partners.
- Regulatory agencies and other health authorities.
- IRBs and IECs.

Investigator's personal information may be retained, processed, and transferred by Takeda and these other parties for research purposes including the following:

- Assessment of the suitability of Investigator for the trial and/or other clinical trials.
- Management, monitoring, inspection, and audit of the trial.
- Analysis, review, and verification of the trial results.
- Safety reporting and pharmacovigilance relating to the trial.
- Preparation and submission of regulatory filings, correspondence, and communications to regulatory agencies relating to the trial.
- Preparation and submission of regulatory filings, correspondence, and communications to regulatory agencies relating to other vaccines used in other clinical trials that may contain the same chemical compound present in the investigational vaccine.
- Inspections and investigations by regulatory authorities relating to the trial.
- Self-inspection and internal audit within Takeda, its affiliates, and licensing partners.
- Archiving and audit of trial records.
- Posting Investigator site contact information, trial details and results on publicly accessible clinical trial registries, databases, and websites.

Investigator's personal information may be transferred to other countries that do not have data protection laws that offer the same level of protection as data protection laws in Investigator's own country. Investigator acknowledges and consents to the use of his or her personal information by Takeda and other parties for the purposes described above.

Please note that this consent will not cover Investigator personnel.

APPENDIX C ELEMENTS OF THE SUBJECT INFORMED CONSENT FORM

In seeking informed consent, the following information shall be provided to each subject:

1. A statement that the trial involves research.
2. An explanation of the purposes of the research.
3. The expected duration of the subject's participation.
4. A description of the procedures to be followed, including invasive procedures.
5. The identification of any procedures that are experimental.
6. The estimated number of subjects involved in the trial.
7. A description of the subject's responsibilities.
8. A description of the conduct of the trial.
9. A statement describing the vaccination(s) and the probability for random assignment to each trial group.
10. A description of the possible side effects following vaccine administration that the subject may receive.
11. A description of any reasonably foreseeable risks or discomforts to the subject and, when applicable, to an embryo, fetus, or nursing infant.
12. A statement that the vaccinations may involve risks to the subject (or to the embryo or fetus, if the subject is or may become pregnant) which are currently unforeseeable.
13. A description of any benefits to the subject or to others that reasonably may be expected from the research. When there is no intended clinical benefit to the subject, the subject should be made aware of this.
14. Disclosures of appropriate alternative procedures or courses of treatment, if any, that might be advantageous to the subject and their important potential risks and benefits.
15. A statement describing the extent to which confidentiality of records identifying the subject will be maintained, and a note of the possibility that regulatory agencies, auditor(s), IRB/IEC, and the monitor may inspect the records. By signing a written informed consent form, the subject is authorizing such access.
16. For research involving more than minimal risk, an explanation as to whether any compensation and an explanation as to whether any medical treatments are available if injury occurs and, if so, what they consist of or where further information may be obtained.
17. The anticipated prorated payment(s), if any, to the subject for participating in the trial.
18. The anticipated expenses, if any, to the subject for participating in the trial.

19. An explanation of whom to contact for answers to pertinent questions about the research (Investigator), subject's rights, and IRB/IEC and whom to contact in the event of a research-related injury to the subject.
20. A statement that participation is voluntary, that refusal to participate will involve no penalty or loss of benefits to which the subject otherwise is entitled, and that the subject may discontinue participation at any time without penalty or loss of benefits to which the subject is otherwise entitled.
21. The consequences of a subject's decision to withdraw from the research and procedures for orderly termination of participation by the subject.
22. A statement that the subject will be informed in a timely manner if information becomes available that may be relevant to the subject's willingness to continue participation in the trial.
23. A statement that results of pharmacogenomic analysis will not be disclosed to an individual, unless prevailing laws require the Sponsor to do so.
24. The foreseeable circumstances or reasons under which the subject's participation in the trial may be terminated.
25. A written subject authorization (either contained within the informed consent form or provided as a separate document) describing to the subject the contemplated and permissible uses and disclosures of the subject's personal information (including personal health information) for purposes of conducting the trial. The subject authorization must contain the following statements regarding the uses and disclosures of the subject's personal information:
    a) that personal information (including personal health information) may be processed by or transferred to other parties in other countries for clinical research and safety reporting purposes, including, without limitation, to the following: (1) Takeda, its affiliates, and licensing partners; (2) business partners assisting Takeda, its affiliates, and licensing partners; (3) regulatory agencies and other health authorities; and (4) IRBs/IECs;
    b) it is possible that personal information (including personal health information) may be processed and transferred to countries that do not have data protection laws that offer subjects the same level of protection as the data protection laws within this country; however, Takeda will make every effort to keep your personal information confidential, and your name will not be disclosed outside the clinic unless required by law;
    c) that personal information (including personal health information) may be added to Takeda's research databases for purposes of developing a better understanding of the safety and effectiveness of the investigational vaccine(s), studying other therapies for patients, developing a better understanding of disease, and improving the efficiency of future clinical trials;
    d) that subjects agree not to restrict the use and disclosure of their personal information (including personal health information) upon withdrawal from the trial to the extent that the restricted use or disclosure of such information may impact the scientific integrity of the research; and

e) that the subject's identity will remain confidential in the event that trial results are published.

26. Female subjects of childbearing potential (e.g., non-sterilized, premenopausal female subjects) who are sexually active must use adequate contraception (as defined in the informed consent form) from at least 2 months prior to Day 1 (M0) up to 6 weeks post third vaccination on Day 180 (M6). Pregnancy tests will be performed prior to vaccination at Day 1 (M0), Day 90 (M3), and Day 180 (M6). Additional pregnancy tests may be performed during the trial if deemed necessary by the Investigator. Females of childbearing potential who are sexually active will be reminded during trial visits to adhere to acceptable contraceptive methods up to 6 weeks post third vaccination on Day 180 (M6). In addition, they must be advised not to donate ova during this period. If a subject is found to be pregnant during the trial, no further vaccine doses will be administered.
27. A statement that clinical trial information from this trial will be publicly disclosed in a publicly accessible website, such as ClinicalTrials.gov.

APPENDIX D SEROLOGY PLAN

Table 16.a Serology Plan

Timing	Blood volume	Assessments
Day 1 (M0), Day 30 (M1), Day180 (M6), Day 210 days (M7)	10 mL	YF neutralizing antibodies (PRNT)
Day 1 (M0), Day 30 (M1), Day 90 (M3), Day 120 (M4), Day180 (M6) and Day 210 (M7)	5 mL	Dengue neutralizing antibodies (MNT_{50})

M=month, MNT_{50}=microneutralization test 50%, PRNT=plaque reduction neutralization test.
Note: Blood sampling at Day 1 (M0), Day 90 (M3) and Day 180 (M6) should be performed prior to vaccination.
Note: The blood sample on Day 120 (M4) and Day 210 (M7) should be taken at least 29 days after the vaccination on Day 90 (M3) and Day 180 (M6), respectively.

Signature Page for DEN-305 Protocol Amendment 2, Version 4.0, 28 November 2018
Title: A Randomized, Observer-Blind, Placebo-Controlled, Phase 3 Trial to Invest

Approval	PPD
Approval	PPD
Approval	PPD
Approval	PPD

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