**Protocol B9371039**

# A PHASE 3, SINGLE-ARM, OPEN-LABEL STUDY TO EVALUATE THE IMMUNOGENICITY, SAFETY, AND TOLERABILITY OF A TICK-BORNE ENCEPHALITIS VACCINE IN HEALTHY JAPANESE PARTICIPANTS 1 YEAR OF AGE AND OLDER

**Statistical Analysis Plan**
**(SAP)**

**Version:** 2

**Date:** 07-FEB-2022

[redacted]



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## APPENDICES

# 1. VERSION HISTORY

**Table 1. Summary of Changes**

| Version/ Date | Associated Protocol Amendment | Rationale | Specific Changes |
|---|---|---|---|
| 1/ 05-JAN-2021 | 1 10-Nov-2020 | N/A | N/A |
| 2/ 07-FEB-2022 | 1 10-Nov-2020 | Modification of description and update of analyses and summaries | Updated to ensure consistent description for CCI [redacted] in the document<br>Section 3.4.1<br>Minor addition<br>Section 3.5.2.1 (Table 3 and Table 4)<br>Revised the scale (cm) for the mild category<br>Section 5.3.1<br>Updated to match with the protocol<br>Section 6.1.2.1<br>Removed the number of occurrences of the event from the AE summary and updated the reporting summaries<br>Section 6.1.2.2.1 and Section 6.1.2.2.2<br>Removed moderate or severe reactions<br>Added the median in summary statistics for the duration of each local reaction or systemic event<br>Section 6.2.2.1<br>Minor modification<br>Section 6.3.1.2<br>Minor modification<br>Section 6.4<br>Minor modification<br>Added the definition of seropositivity for CCI [redacted]<br>Added the possibility of subset analyses for the evaluable immunogenicity population for the second dose<br>Section 6.5.1<br>Updated baseline summaries<br>Section 6.5.2<br>Removed screened participants and screen failure from the disposition summary<br>Section 6.5.3<br>Removed the summary of prior and concomitant nonstudy vaccinations by ATC 4<br>Section 6.5.4<br>Removed the summary of concomitant medications, as only the listing will be provided |

# 2. INTRODUCTION

This SAP provides the detailed methodology for summary and statistical analyses of the data collected in Study B9371039. This document may modify the plans outlined in the protocol; however, any major modifications of the primary endpoint definition or its analysis will also be reflected in a protocol amendment.

## 2.1. Study Objectives, Endpoints, and Estimands

The estimands corresponding to each primary, secondary, CCI [redacted] objective are described in Table 2 below.

The primary estimand of this study is the hypothetical estimand, which estimates the vaccine immune response in the hypothetical setting where participants follow the study schedule and protocol requirements as directed. It includes the following 4 attributes:

- Population: Healthy Japanese adult participants 16 years of age and older who received 3-dose TBE vaccine 0.5 mL in compliance with the key protocol criteria (evaluable adult participants [Section 4]) and healthy Japanese pediatric participants ≥1 to <16 years of age who received 3-dose TBE vaccine 0.25 mL in compliance with the key protocol criteria (evaluable pediatric participants [Section 4]);

- Variable: Seropositivity based on TBEV-neutralizing antibody levels (achieving NT titer ≥1:10) 4 weeks after the third dose;

- Intercurrent event: All data after an intercurrent event (protocol deviation, discontinuation of the study, etc.), if collected, will be excluded. Major protocol violations will be determined by clinical review (through the data handling memo);

- Population-level summary: The proportion of participants who are seropositive 4 weeks after the third dose in each age group.

**Table 2. Study Objectives, Endpoints, and Estimands**

| Primary Immunogenicity Objectives | Estimands | Primary Immunogenicity Endpoints |
|---|---|---|
| • To evaluate the immunogenicity of TBE vaccine 0.5 mL by NT.<br>• To evaluate the immunogenicity of TBE vaccine 0.25 mL by NT. | • In healthy Japanese adult participants 16 years of age and older who received 3-dose TBE vaccine 0.5 mL in compliance with the key protocol criteria (evaluable participants): The proportion who are seropositive (achieving NT titer ≥1:10) 4 weeks after the third dose.<br>• In healthy Japanese pediatric participants ≥1 to <16 years of age who received 3-dose TBE vaccine 0.25 mL in compliance with the key protocol criteria (evaluable participants): The proportion who are seropositive (achieving NT titer ≥1:10) 4 weeks after the third dose. | TBEV-neutralizing antibody titers. |

## Table 2. Study Objectives, Endpoints, and Estimands

| Primary Safety Objectives | Estimands | Primary Safety Endpoints |
|---|---|---|
| • To evaluate the safety profile of TBE vaccine 0.5 mL.<br>• To evaluate the safety profile of TBE vaccine 0.25 mL. | In healthy Japanese participants from each age group receiving at least 1 dose of investigational product:<br>• The percentage of participants reporting local reactions within 7 days after each vaccination.<br>• The percentage of participants reporting systemic events within 7 days after each vaccination.<br>• The percentage of participants reporting AEs within 1 month after each dose.<br>• The percentage of participants reporting SAEs during the study period. | • Prespecified local reactions (redness, swelling, and pain at the injection site).<br>• Prespecified systemic events (fever, decreased appetite, drowsiness, and irritability for participants 1 to ≤2 years of age; and fever, fatigue, headache, vomiting, diarrhea, muscle pain, and joint pain for participants >2 years of age).<br>• AEs within 1 month after vaccination.<br>• SAEs throughout the study. |
| **Secondary Immunogenicity Objectives** | **Estimands** | **Secondary Immunogenicity Endpoints** |
| • To describe the immunogenicity of TBE vaccine 0.5 mL by NT.<br>• To describe the immunogenicity of TBE vaccine 0.25 mL by NT. | In healthy Japanese adult participants 16 years of age and older who received 3-dose TBE vaccine 0.5 mL in compliance with the key protocol criteria (evaluable participants):<br>• The proportion who are seropositive (achieving NT titer ≥1:10) 4 weeks after the second dose.<br>• NT GMTs 4 weeks after the second and 4 weeks after the third dose.<br>• NT GMFRs 4 weeks after the second and 4 weeks after the third dose as compared to baseline.<br>• NT GMFR 4 weeks after the third dose as compared to 4 weeks after the second dose.<br><br>In healthy Japanese pediatric participants 1 through 15 years of age who received 3-dose TBE vaccine 0.25 mL in compliance with the key protocol criteria (evaluable participants):<br>• The proportion who are seropositive (achieving NT titer ≥1:10) 4 weeks after the second dose. | TBEV-neutralizing antibody titers. |

**Table 2. Study Objectives, Endpoints, and Estimands**

| | | |
|---|---|---|
| | • NT GMTs 4 weeks after the second and 4 weeks after the third dose.<br>• NT GMFRs 4 weeks after the second and 4 weeks after the third dose as compared to baseline.<br>• NT GMFR 4 weeks after the third dose as compared to 4 weeks after the second dose. | |
| CCI | | |
| | | . |

**Table 2. Study Objectives, Endpoints, and Estimands**

## 2.2. Study Design

This Phase 3, multicenter, single-arm, open-label study will be conducted at investigator sites in Japan. This study is part of the Phase 3 clinical development plan to support use of TBE vaccine 0.5 mL and 0.25 mL in healthy Japanese adults (≥16 years of age) and the pediatric population (≥1 to <16 years of age). The purpose of this study is to provide key safety and immunogenicity data in Japanese participants. Study duration is approximately 7 to 15 months for each participant. Approximately 100 adults (≥16 years of age at the time of consent) and 65 children (≥1 to <16 years of age at the time of consent) will be enrolled to receive investigational product such that approximately 150 evaluable participants complete the study.

Vaccinations will occur at Visits 1, 2, and 4 and blood draw will be taken prior to the first dose (Visit 1), 1 month after the second dose (Visit 3), prior to the third dose (Visit 4), and 1 month after the third dose (Visit 5) to assess immune responses (Section 1.3 of the protocol).

Local reactions (redness, swelling, and pain at the injection site), systemic events (fever, decreased appetite, drowsiness, and irritability for participants 1 to ≤2 years of age; and fever, fatigue, headache, vomiting, diarrhea, muscle pain, and joint pain for participants >2 years of age), and use of antipyretic/pain medication will be collected in the e-diary within 7 days after each vaccination.

AEs will be collected from the time the participant/parent(s)/legal guardian/legally authorized representative provides informed consent, through and including Visit 3, and from Visit 4 to Visit 5. SAEs will be collected throughout the study.

No data monitoring committee will be established for the study.

# 3. ENDPOINTS AND BASELINE VARIABLES: DEFINITIONS AND CONVENTIONS

## 3.1. Primary Endpoint

- TBEV-neutralizing antibody levels
    - These will be determined 4 weeks after the third dose using Adner NT assay for the analysis of the primary estimands.

- To support the primary estimands, an NT titer will be classified as seropositive if the titer is ≥1:10 at each time point.

### 3.2. Secondary Endpoints

- TBEV-neutralizing antibody levels
  - These will be determined at baseline, 4 weeks after the second dose, and before the third dose using Adner NT assay for the analysis of the secondary estimands.
  - To support the secondary estimands, an NT titer will be classified as seropositive if the titer is ≥1:10 at each time point.
  - To support the secondary estimands, the NT fold rise between the following time points will be calculated:
    - Baseline and 4 weeks after the second dose
    - Baseline and 4 weeks after the third dose
    - 4 Weeks after the second dose and 4 weeks after the third dose

CCI

[redacted]

### 3.4. Baseline and Other Variables

#### 3.4.1. Demographics, Medical History, and Physical Examination

The demographic variables are age (in years) at the time of informed consent (Day 1), sex, race, and ethnicity.

Age at Day 1 will be directly calculated based on the participant's date of birth recorded in the CRF. For example, if the vaccination day is 1 day before the participant's 21st birthday, the participant is considered to be 20 years of age.

Day 1 is also defined as the day of the first vaccination and the start of the subsequent reporting period for local reactions and systemic events in the e-diary.

Day 1 is considered the baseline visit for the immunogenicity assessments.

Medical history will be categorized according to MedDRA.

Physical examination (at Visit 1) will include, at a minimum, assessments of the cardiovascular, respiratory, gastrointestinal, and neurological systems. Height and weight will also be measured and recorded. Prior to subsequent vaccination (at Visits 2 and 4), physical examination should be performed if the clinical assessment indicates that it is necessary to comprehensively evaluate the participant.

[redacted]

### 3.4.2. E-Diary Transmission

An e-diary will be considered transmitted if any data for the local reactions, systemic events, or use of antipyretic/pain medication are present for any day. If all data are missing for all items on the e-diary for all 7 days after vaccination, then the e-diary will be considered not transmitted. An e-diary will be considered completed if all expected data for all 7 days are available (ie, not missing). Otherwise, the e-diary will be considered incomplete. For any given day, an e-diary will be considered complete if all expected data are available.

### 3.4.3. Prior/Concomitant Nonstudy Vaccines and Concomitant Treatments

The name and date of administration for any nonstudy vaccinations received from the time of signing of the ICD until the final visit will be collected and recorded in the CRF.

History of JEV vaccination of participants will be collected at Visit 1 as completely as possible.

Nonstudy vaccines and medications taken during the study will be categorized according to the WHO Drug Dictionary and summarized in accordance with the sponsor reporting standards.

## 3.5. Safety Endpoints

### 3.5.1. Adverse Events

AEs will be captured and reported in accordance with Pfizer reporting standards.

The time period for actively eliciting and collecting AEs and SAEs ("active collection period") for each participant begins from the time the participant/parent(s)/legal guardian/legally authorized representative provides informed consent, which is obtained before the participant's participation in the study (ie, before undergoing any study-related procedure and/or receiving investigational product), through and including Visit 3, and from Visit 4 to Visit 5. Between Visit 3 and Visit 4, only SAEs will be reported.

For participants who are screen failures, the active collection period ends when screen failure status is determined.

If the participant withdraws from the study and also withdraws consent for the collection of future information, the active collection period ends when consent is withdrawn.

If a participant definitively discontinues or temporarily discontinues investigational product because of an AE or SAE, the AE or SAE must be recorded on the CRF and the SAE reported using the Vaccine SAE Reporting Form.

Both nonserious AEs and SAEs are recorded on the CRF and will be categorized according to the current version (at the time of reporting) of MedDRA.

An immediate AE is defined as any AE that occurred within the first 30 minutes after administration of the investigational product.

The following AEs will be summarized at the participant level:

- AEs reported within 1 month after the first dose (from Visit 1 to Visit 2).
- AEs reported within 1 month after the second dose (from Visit 2 to Visit 3).
- AEs reported within 1 month after the third dose (from Visit 4 to Visit 5).
- AEs reported during the vaccination phase.
- Related AEs, severe AEs, related severe AEs, life-threatening AEs, related life-threatening AEs, AEs leading to discontinuation, related AEs leading to discontinuation, and immediate AEs will be summarized in the same way.
- SAEs reported from the first dose until 1 month after the third dose.
- Related SAEs will be summarized in the same way.

### 3.5.2. Reactogenicity Data

Reactogenicity data are prompted AEs collected using an e-diary, during Days 1 through 7, starting on the day of each vaccination. The reactogenicity data will include local reactions (pain, redness, and swelling at the injection site) and systemic events (decreased appetite, drowsiness, and irritability for participants 1 to ≤2 years of age; and fever, fatigue, headache, vomiting, diarrhea, muscle pain, and joint pain for participants >2 years of age).

#### 3.5.2.1. Local Reactions

Local reactions reported in the e-diary are redness, swelling, and pain at the injection site.

**Presence of Local Reactions (Proportion of Participants Reporting)**

The participant or participant's legally acceptable representative/parent/legal guardian will record the presence or absence of pain at the injection site in the e-diary as "mild," "moderate," "severe," or "none". Redness and swelling will be measured and recorded in measuring device units and then categorized during analysis as mild, moderate, or severe based on the grading scale in Table 3, Table 4, and Table 5. Measuring device units can be converted to centimeters according to the following scale: 1 measuring device unit = 0.5 cm. The participant or participant's parent(s)/legal guardian(s) will be prompted to contact the investigator if the participant experiences a severe (Grade 3 or above) local reaction to assess the reaction and to perform an unscheduled assessment or visit as appropriate.

Only an investigator is able to classify a participant's local reaction as Grade 4, after physical examination of the participant or documentation from another medically qualified source (eg, emergency room or hospital record) or, in the case of pain at the injection site only, contact with the participant. If a participant experiences a Grade 4 local reaction, the investigator must immediately notify the sponsor. Grade 4 reactions will be collected on the unscheduled reactogenicity page and as an AE on the CRF.

The AE event will be graded using the AE intensity grading scale as indicated in the table "Assessment of Intensity" in Section 10.3 of the protocol.

The presence or absence of each local reaction on a given day is defined as follows:

= "Missing," if the value is missing on a given day;

= "Yes," if the participant reports the reaction as "yes" for redness or swelling (with a specific diameter or more as defined in Table 3, Table 4, and Table 5) **or** "mild," "moderate," "severe," or "Grade 4" for pain at the injection site on a given day;

= "No," if the participant reports the reaction as "no" for redness or swelling **or** "none" for pain at the injection site on a given day.

**Grading Scale for Local Reactions**

The grading of local reactions is listed below in Table 3, Table 4, and Table 5.

**Participants 1 to ≤2 Years of Age**

**Table 3. Grading Scale for Local Reactions for Participants 1 to ≤2 Years of Age**

| | Mild Grade 1 | Moderate Grade 2 | Severe Grade 3[a] | Grade 4[b] |
|---|---|---|---|---|
| Redness | >0 cm to 2.0 cm (1 to 4 measuring device units) | >2.0 cm to 7.0 cm (5 to 14 measuring device units) | >7.0 cm (>14 measuring device units) | Necrosis or exfoliative dermatitis[b] |
| Swelling | >0 cm to 2.0 cm (1 to 4 measuring device units) | >2.0 cm to 7.0 cm (5 to 14 measuring device units) | >7.0 cm (>14 measuring device units) | Necrosis[b] |
| Pain at the injection site (tenderness) | Hurts if gently touched (eg, whimpers, winces, protests, or withdraws) | Hurts if gently touched with crying | Causes limitation of limb movement | Emergency room visit or hospitalization for severe pain at the injection site |

a. Participants experiencing local reactions >14 caliper units (>7.0 cm) are to be seen by or contact the study site; an unscheduled visit may be required.
b. Grade 4 assessment must be made by an investigator; Grade 4 will not be collected in the e-diary but will be collected in the case report form. The severity of the local reaction should be graded using the AE severity grading scale provided in the protocol.

**Participants >2 to <12 Years of Age**

**Table 4. Grading Scale for Local Reactions for Participants >2 to <12 Years of Age**

| | Mild Grade 1 | Moderate Grade 2 | Severe Grade 3[a] | Grade 4[b] |
|---|---|---|---|---|
| Redness | >0 cm to 2.0 cm (1 to 4 measuring device units) | >2.0 cm to 7.0 cm (5 to 14 measuring device units) | >7.0 cm (>14 measuring device units) | Necrosis or exfoliative dermatitis[b] |

**Table 4. Grading Scale for Local Reactions for Participants >2 to <12 Years of Age**

| | Mild Grade 1 | Moderate Grade 2 | Severe Grade 3[a] | Grade 4[b] |
|---|---|---|---|---|
| Swelling | >0 cm to 2.0 cm (1 to 4 measuring device units) | >2.0 cm to 7.0 cm (5 to 14 measuring device units) | >7.0 cm (>14 measuring device units) | Necrosis[b] |
| Pain at the injection site | Does not interfere with activity | Interferes with activity | Prevents daily activity | Emergency room visit or hospitalization for severe pain at the injection site |

a. Participants experiencing local reactions >14 caliper units (>7.0 cm) are to be seen by or contact the study site; an unscheduled visit may be required.
b. Grade 4 assessment must be made by an investigator; Grade 4 will not be collected in the e-diary but will be collected in the case report form. The severity of the local reaction should be graded using the AE severity grading scale provided in the protocol.

**Participants ≥12 Years of Age**

**Table 5. Grading Scale for Local Reactions for Participants ≥12 Years of Age**

| | Mild Grade 1 | Moderate Grade 2 | Severe Grade 3[a] | Grade 4[b] |
|---|---|---|---|---|
| Redness | >2.0 cm to 5.0 cm (5 to 10 measuring device units) | >5.0 cm to 10.0 cm (11 to 20 measuring device units) | >10 cm (>20 measuring device units) | Necrosis or exfoliative dermatitis[b] |
| Swelling | >2.0 cm to 5.0 cm (5 to 10 measuring device units) | >5.0 cm to 10.0 cm (11 to 20 measuring device units) | >10 cm (>20 measuring device units) | Necrosis[b] |
| Pain at the injection site | Does not interfere with activity | Interferes with activity | Prevents daily activity | Emergency room visit or hospitalization for severe pain at the injection site |

a. Participants experiencing local reactions >20 caliper units (>10 cm) are to be seen by or contact the study site; an unscheduled visit may be required.
b. Grade 4 assessment must be made by an investigator; Grade 4 will not be collected in the e-diary but will be collected in the case report form. The severity of the local reaction should be graded using the AE severity grading scale provided in the protocol.

For each local reaction, the derivation of whether or not the specific reaction occurred on "any day (Day 1-7)" will be made. The derivation of this variable is given in Table 6 below.

**Table 6. Derived Variables for Each Local Reaction**

| Variable[a] | Yes (1) | No (0) | Missing (.) |
|---|---|---|---|
| Any day (Day 1-7) | Participant reports the reaction as "yes" on any day from Day 1 through Day 7. | Participant reports the reaction as "no" on all 7 days or as a combination of "no" and missing on all 7 days. | Participant's response to the reaction is missing on all 7 days. |

a. The variable will be defined for each of the 3 local reactions.

For "any local reaction" on any day, a similar definition can be applied as given in Table 7 below.

**Table 7. Derived Variables for Any Local Reaction**

| Variable | Yes (1) | No (0) | Missing (.) |
|---|---|---|---|
| Any day (Day 1-7) | Participant reports any reaction as "yes" on any day during Days 1 through 7. | Participant reports all reactions as "no" on all 7 days or as a combination of "no" and missing on all 7 days for all 3 local reactions. | Participant's response to all of the local reactions is missing on all 7 days. |

**Maximum Severity for Local Reactions**

The maximum severity (highest grading) of each local reaction within 7 days after each vaccination will be derived. The maximum severity will be derived as follows:

= "Missing," if values are missing for all days from Day 1 through Day 7 after each vaccination;

= 0, if the participant reports all reactions as "no" or a combination of missing and "no" for all days from Day 1 through Day 7 after each vaccination;

= *highest grade* (maximum severity) within 7 days after each vaccination if the answer is not "no" for at least 1 day.

**Duration of Each Local Reaction**

The duration of each local reaction will be calculated in days as (resolution date of reaction - start date of reaction + 1). Resolution of the reaction is the last day on which the reaction is recorded in the e-diary or the date the reaction ends if it is unresolved during the participant diary-recording period (end date collected on the CRF), unless chronicity is established. If there is no known end date, the duration will be considered unknown and set to missing. However, if a reaction is ongoing at the time of a subsequent vaccination, the end date/day for the ongoing event would be the date/day that the next vaccine is administered, which will be used for the duration computation.

**Onset of Local Reaction**

The onset day of each local reaction and any local reaction will be derived.

For the onset day of each local reaction, if participants report severity change of the local reaction, the first day of initial reporting of that specific local reaction will be counted.

For the onset day of any local reaction, the first day of reporting any severity of any local reaction will be counted.

In summary, the following variables will be derived for local reactions:

1. Presence or absence of each local reaction on each day (Days 1-7) after each vaccination.
2. Presence or absence of each local reaction on "any day (Day 1-7)" after each vaccination.
3. Maximum severity of each local reaction on "any day (Day 1-7)" after each vaccination.
4. Maximum severity of each local reaction on "any day (Day 1-7)" after any vaccination.
5. Presence or absence of any local reaction on each day (Days 1-7) after each vaccination.
6. Presence or absence of any local reaction on "any day (Day 1-7)" after each vaccination.
7. Duration of each local reaction after each vaccination.
8. Onset day of each local reaction after each vaccination.
9. Onset day of any local reaction after each vaccination.

### 3.5.2.2. Systemic Events

Systemic events reported via the e-diary are fever, decreased appetite, drowsiness, and irritability for participants 1 to ≤2 years of age; and fever, fatigue, headache, vomiting, diarrhea, muscle pain, and joint pain for participants >2 years of age. The participant or participant's legally acceptable representative/parent/legal guardian is to document the presence or absence of systemic events in the e-diary as "mild," "moderate," "severe," or "none". The participant or participant's legally acceptable representative/parent/legal guardian is to be asked to assess the severity of each event according to Table 8 and Table 9 below.

Only an investigator is able to classify a participant's systemic event as Grade 4, after physical examination of the participant or documentation from another medically qualified source (eg, emergency room or hospital record) or contact with the participant. If a participant experiences a Grade 4 systemic event, the investigator must immediately notify the sponsor. A Grade 4 event will be collected on the unscheduled reactogenicity page and as an AE on the CRF. The event will be graded using the AE severity grading scale (see Section 10.3 of the protocol). Further, for all ongoing systemic events on Day 7, the stop date will be recorded in the CRF.

**Table 8. Grading Scale for Systemic Events for Participants 1 to ≤2 Years of Age**

| | Mild<br>Grade 1 | Moderate<br>Grade 2 | Severe<br>Grade 3 | Grade 4[a] |
|---|---|---|---|---|
| Decreased appetite (loss of appetite) | Decreased interest in eating | Decreased oral intake | Refusal to feed | Emergency room visit or hospitalization for severe decreased appetite (loss of appetite) |

**Table 8. Grading Scale for Systemic Events for Participants 1 to ≤2 Years of Age**

| | Mild Grade 1 | Moderate Grade 2 | Severe Grade 3 | Grade 4[a] |
|---|---|---|---|---|
| Drowsiness (increased sleep) | Increased or prolonged sleeping bouts | Slightly subdued interfering with daily activity | Disabling not interested in usual daily activity | Emergency room visit or hospitalization for severe drowsiness (increased sleep) |
| Irritability (fussiness)<br>(synonymous with restless sleep; decreased sleep) | Easily consolable | Requiring increased attention | Inconsolable; crying cannot be comforted | Emergency room visit or hospitalization for severe irritability (fussiness) |

a. Grade 4 assessment should be made by an investigator; Grade 4 will not be collected in the e-diary but will be collected in the case report form.

**Table 9. Grading Scale for Systemic Events for Participants >2 Years of Age**

| | Mild Grade 1 | Moderate Grade 2 | Severe Grade 3 | Grade 4[a] |
|---|---|---|---|---|
| Fatigue (= tiredness in diaries) | Does not interfere with activity | Some interference with activity | Prevents daily routine activity | Emergency room visit or hospitalization for severe fatigue |
| Headache | Does not interfere with activity | Some interference with activity | Prevents daily routine activity | Emergency room visit or hospitalization for severe headache |
| Vomiting | 1 to 2 times in 24 hours | >2 times in 24 hours | Requires intravenous hydration | Emergency room visit or hospitalization for severe vomiting |
| Diarrhea | 2 to 3 loose stools in 24 hours | 4 to 5 loose stools in 24 hours | 6 or more loose stools in 24 hours | Emergency room visit or hospitalization for severe diarrhea |
| Muscle pain | Does not interfere with activity | Some interference with activity | Prevents daily routine activity | Emergency room visit or hospitalization for severe muscle pain |
| Joint pain | Does not interfere with activity | Some interference with activity | Prevents daily routine activity | Emergency room visit or hospitalization for severe joint pain |

a. Grade 4 assessment should be made by an investigator; Grade 4 will not be collected in the e-diary but will be collected in the case report form.

The highest temperature for each day for 7 days after vaccination is to be recorded in the e-diary. The protocol defines fever as an axillary temperature ≥37.5°C. For ongoing fever on Day 7, the stop date will be recorded in the CRF. For reporting purposes, fever will be analyzed using the following temperature grading scale as displayed in Table 10 for all participants.

**Table 10. Ranges for Fever**

| | Mild Grade 1 | Moderate Grade 2 | Severe Grade 3 | Grade 4 |
|---|---|---|---|---|
| Fever | ≥37.5°C; ≤38.4°C | ≥38.5°C; ≤38.9°C | ≥39.0°C; ≤40.0°C | >40.0°C |

The presence or absence of each systemic event on a given day is defined as follows:

= "Missing," if the value is missing on a given day;

= "Yes," if the participant reports a temperature ≥37.5°C for fever **or** "mild," "moderate," "severe," or "Grade 4" for the remaining events on a given day;

= "No," if the participant reports a temperature <37.5°C for fever **or** "none" for the remaining events on a given day.

For each systemic event, the following variables will be derived:

1. Presence or absence of each systemic event on each day (Days 1-7) after each vaccination.
2. Presence or absence of each systemic event on "any day (Day 1-7)" after each vaccination.
3. Maximum severity of each systemic event on "any day (Day 1-7)" after each vaccination.
4. Maximum severity of each systemic event on "any day (Day 1-7)" after any vaccination.
5. Presence or absence of any systemic event on each day (Days 1-7) after each vaccination.
6. Presence or absence of any systemic event on "any day (Day 1-7)" after each vaccination.
7. Duration of each systemic event after each vaccination.
8. Onset day of each systemic event after each vaccination.
9. Onset day of any systemic event after each vaccination.

The derivation of these variables is similar to the derivation of the variables for local reactions (Section 3.5.2.1). "Any systemic event" includes any fever, decreased appetite, drowsiness, or irritability for participants 1 to ≤2 years of age; and any fever, fatigue, headache, vomiting, diarrhea, muscle pain, or joint pain for participants >2 years of age.

### 3.5.3. Use of Antipyretic/Pain Medication

Use of antipyretic and/or pain medication will be reported via the e-diary. The participant or participant's parent(s)/legal guardian(s) is to document the presence or absence of use of antipyretic and/or pain medication in the e-diary as "yes" or "no". Variables other than those related to severity are derived following the same rule as for local reactions (Section 3.5.2.1).

### 3.5.4. Physical Examination

Physical examination will be performed prior to vaccination at the first vaccination visit and will include any clinically significant abnormalities within the following body systems: general appearance; lungs; heart; abdomen; musculoskeletal; neurological; skin; and lymph nodes. Prior to subsequent vaccination (at Visits 2 and 4), physical examination will be performed if the clinical assessment indicates that it is necessary to comprehensively evaluate the participant.

## 4. ANALYSIS SETS (POPULATIONS FOR ANALYSIS)

Data for all participants will be assessed to determine if participants meet the criteria for inclusion in each analysis population prior to releasing the database and classifications will be documented per standard operating procedures. For purpose of analysis, the following populations are defined for each age group.

| Population | Description |
|---|---|
| Enrolled | "Enrolled" means that a participant, or his or her legally authorized representative, has agreed to participate in a clinical study following completion of the informed consent process and is deemed eligible to participate. Potential participants who are screened for the purpose of determining eligibility for the study, but do not participate in the study, are not considered enrolled, unless otherwise specified by the protocol. |
| Evaluable immunogenicity | All participants who are eligible, receive all 3 doses of the investigational product, have blood drawn for assay testing within the specified time frame for baseline and 4 weeks (21-35 days) after the third vaccination, have valid and determinate assay result (NT titer) at baseline and 4 weeks after the third vaccination visit, are NT seronegative (NT titer <1:10) at baseline, and have no major protocol violations. |
| Evaluable immunogenicity for the second dose | All participants who are eligible, receive the first 2 doses of the investigational product, have blood drawn for assay testing within the specified time frame for baseline and 4 weeks (21-35 days) after the second vaccination, have valid and determinate assay result (NT titer) at baseline and 4 weeks after the second vaccination visit, are NT seronegative (NT titer <1:10) at baseline, and have no major protocol violations. |

| Population | Description |
|---|---|
| All-available immunogenicity | All enrolled participants who receive at least the first 2 doses of the investigational product and have valid and determinate assay results (NT titer CCI [redacted]) 4 weeks after the second and/or the third vaccination. |
| Safety | All enrolled participants who receive at least 1 dose of the investigational product. |

Major protocol violations will be determined by clinical review (through the data handling memo). A major protocol violation is a protocol violation that, in the opinion of the sponsor's study medical monitor, would materially affect assessment of safety and/or immunogenicity, eg, participant receipt of a prohibited vaccine or medication that might affect immune response or a medication error with suspected decrease in potency of the vaccine. The sponsor's medical monitor will identify those participants with protocol violations before any immunogenicity analysis is carried out.

For local reactions, systemic events, fever, and use of antipyretic and/or pain medication, the definition of the denominators for the percentage calculations are described in Section 3.5.2 based on e-diary record. In addition, these tables will be generated without e-diary data that have been confirmed by the participant to have been entered in error. Data-entry errors will be discussed in the clinical study report text and documented in a separate listing.

## 5. GENERAL METHODOLOGY AND CONVENTIONS

All analyses will be performed after completion of the study.

### 5.1. Hypotheses and Decision Rules

The primary immunogenicity objective of the study is to assess the immunogenicity of the investigational product by NT in Japanese healthy adults (16 years of age and older) and children (≥1 to <16 years of age) when administered on a 3-dose schedule. The null hypothesis to be tested concerns the seropositivity rate for the primary endpoint in each age group. The immune response induced by TBE vaccine will be evaluated in each age group, testing the hypotheses $H_0$: seropositivity rate ≤90% vs $H_1$: seropositivity rate >90%. These hypotheses will be tested by age group separately and no adjustment of multiplicity will be considered. For each age group, if the lower bound of the 95% CI on the NT seropositivity rate among the evaluable population is >90%, the objective is achieved.

### 5.2. General Methods

Descriptive summary statistics will be provided for all endpoints. Unless otherwise explicitly stated, descriptive statistics for continuous variables are n, mean, median, standard deviation, minimum, and maximum. Descriptive statistics for categorical variables are the proportion (%) and the n (the numerator) and N (the denominator) used in the calculation of the proportion.

All of the safety, tolerability, and immunogenicity data will be summarized by age group.

### 5.2.1. Analyses for Binary Endpoints

The number and percentage of participants in each category will be summarized. The 95% CI for percentage will also be presented, where appropriate.

The 95% CI for the proportion will be constructed by the Clopper-Pearson method described by Newcombe.[1] The 95% CI will be presented in terms of percentage. For the endpoint to be tested, evaluating 95% CI based on the Clopper-Pearson method comparing its lower limit to 90% corresponds to the statistical testing with the exact method.

### 5.2.2. Analyses for Continuous Endpoints

Unless otherwise specified, the CI for the mean of the continuous variables will be constructed by the standard method based on Student's t distribution.

#### 5.2.2.1. Geometric Mean (GM)

Continuous immunogenicity endpoints will be logarithmically transformed for analysis. GMT[redacted] and associated 2-sided 95% CI will be calculated at each available time point. The GM and associated 2-sided 95% CI will be calculated as the mean of the assay results on the natural logarithmic scale based on Student's t distribution and then exponentiating the results.

#### 5.2.2.2. Geometric Mean Fold Rise (GMFR)

GMFR will be calculated as the mean difference of an individual participant's logarithmically transformed antibody levels (postvaccination minus prevaccination) and back transformed to the original scale. Two-sided 95% CIs will be obtained by constructing CIs using 1-sample Student's t distribution for the mean difference of measures on the logarithmically transformed assay results and transforming the limits back to the original scale.

#### 5.2.2.3. Reverse Cumulative Distribution Curves (RCDCs)

Empirical RCDCs for TBEV-neutralizing antibody titers will be generated by each combination of available time points and age groups.

## 5.3. Methods to Manage Missing Data

### 5.3.1. Immunogenicity Data

Titers below the LLOQ or denoted as BLQ will be set to 0.5 × LLOQ for analysis.

Values that are designated as serum quantity not sufficient (QNS), designated as indeterminate results (IND), or recorded as "not done" will be set to missing. No imputation will be done for these missing values.

### 5.3.2. Safety Data

Standard algorithms on handling missing AE dates and missing AE severity will be applied as described in Pfizer's Vaccine Statistics Rulebook.

### 5.3.3. Reactogenicity Data

For derived variables based on reactogenicity data, if any day of the 7-day e-diary is available, the "any day (Day 1-7)" data will be considered nonmissing.

The reactogenicity data are collected through the e-diary, which does not allow participants to skip the question. Therefore, for a specific day, as long as the e-diary data are transmitted for that day, all of the reactogenicity data for the participant on that day are nonmissing. No missing reactogenicity data will be imputed other than what is described in Section 3.5.2.

## 6. ANALYSES AND SUMMARIES

### 6.1. Primary Endpoints

### 6.1.1. Seropositivity Based on TBE-Neutralizing Antibody Levels (Achieving NT Titer ≥1:10) 4 Weeks After the Third Dose

#### 6.1.1.1. Main Analysis

- Estimand strategy: Hypothetical (Section 2.1).
- Analysis set: Evaluable immunogenicity population (Section 4).
- Analysis methodology: Exact testing of the binomial proportion and descriptive summary statistics.
- Intercurrent events and missing data: For participants who discontinue, all collected data will be included; intermediate missing values will not be imputed.
- Reporting summaries: n (the number of seropositive participants), N (the size of the analysis population), point estimate (seropositivity rate), and the exact 2-sided 95% CI will be calculated using the Clopper-Pearson method for the seropositivity rate based on the immune response determined by NT 4 weeks after the third vaccination. NT titers ≥1:10 are considered seropositive.

CCI [redacted]

[redacted]



[redacted]

### 6.1.2. Safety Endpoints

#### 6.1.2.1. Adverse Events

It should be recognized that most studies are not designed to reliably demonstrate a causal relationship between the use of a pharmaceutical product and an AE or a group of AEs. Except for select events in unique situations, studies do not employ formal adjudication procedures for the purpose of event classification. CCI

- Endpoints: Participants reporting AEs and SAEs categorized according to MedDRA (Section 3.5.1).
- Analysis time points: From the time of informed consent through and including Visit 5 (1 month after the third vaccination):
  - After the first vaccination (Visit 1) and before the second vaccination (Visit 2)
  - After the second vaccination (Visit 2) and before the 1-month blood draw after the second vaccination (Visit 3)
  - After the third vaccination (Visit 4) and before the 1-month blood draw after the third vaccination (Visit 5)
  - During the vaccination phase
- Analysis population: Safety population (Section 4). Participants who lack any safety data will be excluded from that analysis.
- Reporting summaries:
  For each of the following AE categories, the number and percentage of participants reporting at least 1 event of each preferred term, arranged by system organ class with the associated Clopper-Pearson 95% CI. For deaths, only a listing will be provided.
  - AEs reported within 1 month after each vaccination
  - Related AEs reported within 1 month after each vaccination
  - SAEs reported throughout the study
  - Related SAEs reported throughout the study
  - Severe or life-threatening AEs reported within 1 month after each vaccination
  - Related severe or life-threatening AEs reported within 1 month after each vaccination
  - AEs leading to discontinuation reported within 1 month after each vaccination

- Related AEs leading to discontinuation reported within 1 month after each vaccination

- Immediate AEs reported within 1 month after each vaccination

Listings of participants reporting any AE and immediate AEs and listings of all reported AEs will be generated. In addition, AEs occurring after participants signed the informed consent and prior to vaccination will also be listed.

#### 6.1.2.2. Reactogenicity Data

##### 6.1.2.2.1. Local Reactions

- Endpoints: Participants reporting local reactions (redness, swelling, and pain at the injection site) and severity of the reactions (Section 3.5.2.1).

- Analysis time points: 7-Day period following each vaccination.

- Analysis population: Safety population (Section 4).

- Reporting summaries:

  - Maximum severity for local reactions
    The number of participants reporting e-diary data transmitted for at least 1 day (these values are used as the denominators for the percentage calculations), the number of participants reporting at least 1 occurrence of the specified reaction or any reaction by the maximum severity grade, the observed proportion of participants, and their exact 2-sided CIs (Clopper and Pearson).

  - Duration of each local reaction
    Number of participants reporting the specified reaction, mean, median, range (minimum, maximum), standard deviation, and number of unknown durations.

  - Onset of local reactions
    The number of participants reporting the specified reaction or any reaction, mean, range (minimum, maximum), and standard deviation.

  - Figures
    A bar graph showing the percent incidence for the specified reaction and for any reaction, by day.

##### 6.1.2.2.2. Systemic Events

- Endpoints: Participants reporting systemic events (fever, decreased appetite, drowsiness, and irritability for participants 1 to ≤2 years of age; and fever, fatigue, headache, vomiting, diarrhea, muscle pain, and joint pain for participants >2 years of age) and severity of the events (Section 3.5.2.2).

- Analysis time points: 7-Day period following each vaccination.
- Analysis population: Safety population (Section 4).
- Reporting summaries:
  - Maximum severity for systemic events
    The number of participants reporting e-diary data transmitted for at least 1 day (these values are used as the denominators for the percentage calculations), the number of participants reporting at least 1 occurrence of the specified event or any event by the maximum severity grade, the observed proportion of participants, and their exact 2-sided CIs (Clopper and Pearson).
  - Duration of each systemic event
    Number of participants reporting the specified event, mean, median, range (minimum, maximum), standard deviation, and number of unknown durations.
  - Onset of systemic events
    The number of participants reporting the specified event or any event, mean, range (minimum, maximum), and standard deviation.
  - Figures
    A bar graph showing the percent incidence for the specified event and for any event, by day.

## 6.2. Secondary Endpoints

### 6.2.1. Seropositivity Based on TBEV-Neutralizing Antibody Levels (Achieving NT Titer ≥1:10) 4 Weeks After the Second Dose

#### 6.2.1.1. Main Analysis

- Estimand strategy: Hypothetical.
- Analysis set: Evaluable immunogenicity population for the second dose (Section 4).
- Analysis methodology: Descriptive summary statistics.
- Intercurrent events and missing data: For participants who discontinue, all collected data will be included; intermediate missing values will not be imputed.
- Reporting results: n (the number of seropositive participants), N (the size of the analysis population), point estimate (seropositivity rate), and the exact 2-sided 95% CI will be calculated using the Clopper-Pearson method for the seropositivity rate based on the immune response determined by NT 4 weeks after the second vaccination. NT titers ≥1:10 are considered seropositive.

CCI

[redacted]

### 6.2.2. TBEV-Specific Neutralizing Antibody Titer 4 Weeks After the Second and 4 Weeks After the Third Dose

#### 6.2.2.1. Main Analysis

- Estimand strategy: Hypothetical.
- Analysis set: Evaluable immunogenicity population (Section 4). GMT, LLOQ, and RCDC analysis after the second vaccination and GMFR from baseline to 4 weeks after the second vaccination will be conducted based on the evaluable immunogenicity population for the second dose as well.
- Analysis methodology: Descriptive summary statistics.
- Intercurrent events and missing data: For participants who discontinue, all collected data will be included; intermediate missing values will not be imputed.
- Reporting summaries:
  - GMTs of neutralizing antibody at each available time point will be descriptively summarized for each age group, along with associated 2-sided 95% CIs.
  - GMFRs and associated 2-sided 95% CIs will be provided for neutralizing antibody titer from baseline to 4 weeks after the second vaccination and 4 weeks after the third vaccination, and from 4 weeks after the second vaccination to 4 weeks after the third vaccination.
  - Numbers and proportions of participants with neutralizing antibody titer ≥LLOQ at each available time point and associated exact 2-sided CIs based upon the observed proportion of participants will be summarized.
  - Empirical RCDCs for TBEV-neutralizing antibody titers will be generated by each combination of available time points and age groups.

CCI

[redacted]



[redacted]

CCI

### 6.4. Subset Analyses

Subset analyses for all of the safety endpoints will be conducted based on the subset defined by age in the pediatric group. Three categories of age will be considered: 1 to ≤2 years of age, 3 to ≤6 years of age, and 7 to ≤15 years of age.

Subset analyses for the following endpoints will be conducted based on the subgroup by baseline CCI [redacted] All subset analyses will be performed by age group, and the analysis population will be the evaluable immunogenicity population and/or the evaluable immunogenicity population for the second dose as well, if appropriate. If there is no CCI [redacted] case at baseline, these subset analyses will not be conducted.

- Primary endpoint
  - Seropositivity based on TBEV-neutralizing antibody levels (achieving NT titer ≥1:10) 4 weeks after the third dose.
- Secondary endpoints
  - Seropositivity based on TBEV-neutralizing antibody levels (achieving NT titer ≥1:10) 4 weeks after the second dose.
  - TBEV-neutralizing antibody titer 4 weeks after the second and 4 weeks after the third dose.

CCI [redacted]

[redacted]

Additional subset analysis (not defined here) may also be performed if deemed necessary.

### 6.5. Baseline and Other Summaries and Analyses

#### 6.5.1. Baseline Summaries

For each age group, descriptive summary statistics for demographic characteristics (age at enrollment, sex, race, and ethnicity) will be generated based on the analysis populations (Section 4).

The number and proportion of participants with at least 1 medical history preferred term, arranged by system organ class, will be tabulated for each age group. The medical history summary is based on the safety population.

CCI [redacted]

[redacted]



[redacted]

Participant data listings for demography and baseline characteristics data will also be generated.

### 6.5.2. Study Conduct and Participant Disposition

The numbers and proportions of enrolled participants will be included in the participant disposition summary. In addition, participants who completed all study procedures, and those who withdrew in the middle of the study, along with the reasons for withdrawal, will be tabulated by age group separately. The reasons for withdrawal will be those as specified in the database.

Participants excluded from the evaluable immunogenicity and all-available immunogenicity populations will also be summarized with reasons for exclusion. These summaries will be generated by age group.

For each vaccination, the numbers and proportions of participants who were vaccinated, and had blood drawn within the protocol-specified time frame, and outside the specified window, will be tabulated by age group.

The numbers and proportions of participants with e-diary data not transmitted, transmitted by day (Days 1-7), and transmitted on "all days" will be summarized by age group.

Data listings of participants who withdrew during the study will be generated. Also, data listings of participants excluded from the evaluable and all-available immunogenicity populations will be generated separately.

The protocol deviations listings will be generated.

### 6.5.3. Prior and Concomitant Nonstudy Vaccinations

Any JEV vaccines received prior to signing of the ICD and any nonstudy vaccines received from signing of the ICD until completion of study participation will be categorized according to the WHO Drug Dictionary. The number and proportion of participants receiving each vaccine will be tabulated by age group. This tabulation will be performed for the safety population.

### 6.5.4. Concomitant Medications

Any medications used to treat SAEs taken after signing the informed consent will be categorized according to the WHO Drug Dictionary and be listed.

## 6.6. Safety Summaries and Analyses

### 6.6.1. Adverse Events

Refer to Section 6.1.2.1.

### 6.6.2. Reactogenicity Data

Refer to Section 6.1.2.2.

### 6.6.3. Use of Antipyretic/Pain Medication

Analysis time points and the analysis population will be the same as for systemic events described in Section 6.1.2.2.2. A summary will be included in the table for systemic events, where applicable.

### 6.6.4. Physical Examination

Physical examination results prior to each vaccination will be summarized. The number and percentage of participants with each type of finding (subcategories: normal, abnormal, or not performed) for the physical examination will be tabulated by age group. This tabulation will be performed for the safety population.

## 7. INTERIM ANALYSES

No formal interim analysis is planned for this study. However, as this is an open-label study, the sponsor may conduct unblinded reviews of the data during the course of the study for the purpose of safety assessment and/or to support clinical development.

## 8. REFERENCES

1. Newcombe RG. Two-sided confidence intervals for the single proportion: comparison of seven methods. Stat Med. 1998;17(8):857-72.

# 9. APPENDICES

## Appendix 1. List of Abbreviations

| Abbreviation | Term |
|---|---|
| AE | adverse event |
| ATC | Anatomic Therapeutic Chemical |
| BLQ | below the level of quantitation |
| CI | confidence interval |
| CRF | case report form |
| e-diary | electronic diary |
| CCI | [redacted] |
| GM | geometric mean |
| CCI | [redacted] |
| GMFR | geometric mean fold rise |
| GMT | geometric mean titer |
| HI | hemagglutination inhibition |
| ICD | informed consent document |
| CCI | [redacted] |
| IND | indeterminate |
| JEV | Japanese encephalitis virus |
| LLOQ | lower limit of quantitation |
| MedDRA | Medical Dictionary for Regulatory Activities |
| N/A | not applicable |
| NT | neutralization test |
| QNS | quantity not sufficient |
| RCDC | reverse cumulative distribution curve |
| SAE | serious adverse event |
| SAP | statistical analysis plan |
| TBE | tick-borne encephalitis |
| TBEV | tick-borne encephalitis virus |
| VIE U | Vienna units |
| WHO | World Health Organization |