



**Department of Health and Human Services
Food and Drug Administration (FDA)
Center for Biologics Evaluation and Research (CBER)
Office of Biostatistics and Pharmacovigilance (OBPV)**

FINAL RWE EPIDEMIOLOGY REVIEW MEMORANDUM

Date: November 1, 2023

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Subject: Final RWE review memo of chikungunya virus vaccine effectiveness study protocols; postmarket observational confirmatory studies for IXCHIQ (b) (4)

Sponsor: Valneva Austria GmbH

Product: IXCHIQ (VLA1553, a live attenuated, single-dose Chikungunya Virus vaccine)

Application Type/Number: BLA 125777/0

Submission Dates: August 17, 2022 - December 21, 2022

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1 REVIEW OBJECTIVE

The purpose of this review is to assess the observational study design of the proposed RWE study protocol (b) (4) -402), designed to confirm the vaccine effectiveness (VE) of the (b) (4) chikungunya virus (CHIKV) vaccine (IXCHIQ) in the adolescent and adult population in endemic areas of Brazil. This study is proposed to satisfy the postmarket confirmatory study necessary to support the licensure of IXCHIQ in the US. This memo also documents the RWE consult review of the VE elements of the second confirmatory study protocol, (b) (4) -404, a pragmatic Randomized Controlled Trial (RCT) submitted September 7, 2023.

2 BACKGROUND INFORMATION AND REGULATORY HISTORY

Currently there is no vaccine or specific drug against CHIKV infection. Therefore, Valneva applied for a Tropical Disease priority review voucher based on the Section 524 to the FD&C Act on December 21, 2022 with their last submission that completed the rolling BLA and started the review timeline. The target indication for (b) (4) is: "IXCHIQ is a vaccine indicated for active immunization for the prevention of disease caused by Chikungunya virus in individuals 18 years or older".

The Sponsor completed a pivotal Phase 3 clinical trial in March 2022 in the US in support of this BLA application under IND 17854. This clinical trial showed that VLA1553 induces antibody levels considered seroprotective against CHIKV based on surrogate protection ($\mu\text{PRNT}_{50} \geq 150$). Also, this vaccine was confirmed to be highly immunogenic in adults ≥ 65 years old. The immunogenicity profile from study VLA1553-301 was confirmed with a 96.0% sero-response rate at day 180.

The Sponsor informed CBER in the response to IR 38 (5/5/2023), that an RCT is infeasible given the explosive nature of the CHIKV epidemics. The challenges of an RCT were also discussed previously at a Vaccine and Related Biological Products Advisory Committee meeting in November 2019.

Valneva submitted a proposed RWE observational postmarket confirmatory study protocol for a Test-Negative Design (TND) study to be conducted in Brazil (b) (4) -402). They informed CBER the proposed observational study will initiate as soon as (b) (4) is licensed in Brazil and a pilot vaccine program is accepted by the (b) (4) to demonstrate the feasibility of (b) (4) incorporation into the (b) (4) and its benefits on the CHIKV burden of disease in endemic areas in Brazil; they expect to initiate the study in 2025.

The Sponsor provided the following background information about the incidence of Chikungunya cases in Brazil as rationale for choosing to conduct an observational study to confirm VE in Brazilian municipalities:

Chikungunya virus is transmitted by mosquitoes (Aedes aegypti and Aedes albopictus) which also transmit dengue and Zika (WHO, 2022). The most invasive mosquito species in the world is Ae. Albopictus, commonly known as the Asian tiger mosquito (Benedict et al, 2007). This species was first detected in Brazil in 1986 (Rocha et al, 2023), and since

¹ Valneva refers to the previous vaccine version tested and manufactured in the US under IND17854 as VLA1553. (b) (4)

then has rapidly expanded throughout Brazil on 26 of the 27 federative units of Brazil (Ferreira-de-Lima et al, 2020).

The circulation of the virus was first identified in Brazil in 2014 with an outbreak caused by the East/Central/South African (ECSA) genotype.

In 2021, a total of 131,630 suspected chikungunya cases were reported, including 11 deaths (all of them in Brazil), in 14 of the countries/territories in the region of the Americas. Among them, 99% of the cases were reported in three countries: Brazil with 127,487 (97%) cases, Guatemala with 1,951 (1.5%) cases, and Belize with 737 (0.6%) cases. The cumulative incidence rate in the region was 13 cases per 100,000 population. The countries with the highest incidence rates were Belize with 182 cases per 100,000 population, Brazil with 60 cases per 100,000 population, and Guatemala with 11 cases per 100,000 population. There were 21 imported cases of CHIKV in the United States of America.

Given the numerous RWE review concerns with the proposed study (b) (4) -402, Valneva submitted a proposal for sero-surveys to determine sero-prevalence in the targeted Brazilian municipalities and a second confirmatory study protocol (b) (4) -404, a pragmatic RCT, to address both safety and effectiveness in an endemic area.

Refer to the OVR Clinical and OBPV/Division of Pharmacovigilance reviewers' memos for additional information regarding clinical and safety concerns with the proposed studies.

2.1 Review timeline:

- August 17, 2022 – First submission of 125777/0
- December 21, 2022 – Submission of study protocol (b) (4) -402; initiates clock
- February 3, 2023 – Study protocol assigned for RWE review
- April 4, 2023 – Mid-Cycle Internal Meeting
- June 8, 2023 – Information Request (IR) letter #50 was sent based on review of the initial submission of (b) (4) -402
- June 19, 2023 – Response to IR #50 on Postmarketing confirmatory study (b) (4) -402 received
- June 6, 2023 – Late-Cycle Internal Meeting
- July 7, 2023 – IR #61 CBER comments on Valneva's responses to IR #50
- July 17, 2023 – Response to IR #61 on CBER's comments on Valneva's responses to IR #50
- July 25, 2023 – Post-marketing effectiveness study designs were submitted by the Sponsor
- August 2, 2023 – A revised study protocol for (b) (4) -402 related to IR# 60, 61 and 64 was submitted by the Sponsor
- August 11, 2023 – CBER acknowledged major amendment due to Valneva's submission of 7 possible second confirmatory study designs submitted July 25, 2023 – review clock extended to November 22, 2023

- August 16, 2023 – IR #77 Additional CBER comments on post marketing confirmatory study (b) (4) -402
- September 7, 2023 – Valneva submitted an updated version (4.0) of (b) (4) -402 observational effectiveness study protocol based on IR #77, and the (b) (4) -404 protocol concept for a pragmatic randomized controlled effectiveness trial in an endemic country.
- September 20, 2023 – Valneva submitted an updated version of (b) (4) -404 that incorporates a safety evaluation to investigate the safety of IXCHIQ with regard to chikungunya-like illness.
- September 22, 2023 – IR #85 Additional comments regarding protocol (b) (4) -402 (V4.0)
- September 26, 2023 – Telcon with Sponsor to discuss IR #85
- September 28, 2023 – Valneva submitted an updated version 4.1 of BLA (b) (4) -402 based on IR #85 along with written responses to IR #85.
- October 13, 2023 – IR #89 requesting milestone dates for data access and other pertinent activities for (b) (4) -402.
- October 17, 2023 – IR #91 requesting milestone dates for (b) (4) -404 and 402.
- October 20, 2023 – IR #93 requesting the DMP, DHT guidance, and clinical safety concerns
- October 26, 2023 – IR# 95, concern for data evaluation timeframe

3 MATERIALS REVIEWED

This RWE epidemiology review primarily addresses review of protocol (b) (4) -402, **Effectiveness of (b) (4) vaccine against chikungunya virus disease in adolescent and adult population in endemic areas of Brazil. Observation Study Protocol (b) (4) -402** – November 24, 2022, Version draft 0.4; September 7, 2023, Version 4.0; September 28, 2023, Version 4.1, the sponsor’s responses to our original review comments in IR #50, and the subsequent IRs 61, 77, 85 and 89.

The second confirmatory study, (b) (4) -404, **Trial of the Effectiveness and Safety of (b) (4) Vaccine Against Chikungunya Virus Disease in an Endemic Country: A Pragmatic Randomized Controlled Trial Concept Document (b) (4) -404**, September 2023, V3.0 was reviewed primarily by consult RWE review managers Dr. Richard Forshee (OBPV/CBER) and Dr. Hector Izurieta (OVR/RCBER) and by OVR/RCBER clinical reviewers (safety evaluation). The RWE review comments regarding the VE elements of the study design are included in this memo.

Table 1. Submission/Amendments and Documents Reviewed

Submission/Amendment	Date IR Sent	Date Received	Documents Reviewed
125777/0	NA	12/21/2022	Postmarket Confirmatory study (b) (4) -402 Version 0.4

125777/0.53	6/8/2023	6/19/2023	Response to IR #50 on Post-marketing confirmatory study (b) (4) -402
125777/0.64	7/7/2023	7/17/2023	Response to IR #61 on CBER comments on Valneva's responses to IR #50
125777/0.71	NA	7/25/2023	Second Post-marketing confirmatory effectiveness study designs
125777/0.75	NA	8/2/2023	A revised study protocol for (b) (4) -402 Version 3.0 related to IR# 60, 61 and 64
125777/0.84	8/16/2023	9/7/2023	(b) (4) -402 Version 4.0 observational effectiveness study protocol based on IR #77 and clarification telephone conference
125777/0.84	NA	9/7/2023	(b) (4) -404 protocol concept for a pragmatic randomized controlled effectiveness trial in an endemic county (V2.0)
125777/0.87	9/7/2023	9/20/2023	Updated (b) (4) -404 protocol for a pragmatic randomized controlled safety and effectiveness trial in an endemic county (V3.0)
125777/0.88	9/22/2023	9/28/2023	Updated (b) (4) -402 (V4.1) observational effectiveness study protocol based on IR #85 and clarification telephone conference. Written response to IR #85 on (b) (4) -402 (V4.0)
125777/0.93	10/13/2023	10/19/2023	Key Activities towards (b) (4) -402: Pilot Vaccination Program and complementary studies milestone dates; response to IR #89
125777/0.94	10/17/2023	10/20/2023	Milestone dates for both 402 and 404 studies; response to IR #91
125777/0.98	10/25/2023	10/31/2023	Data evaluation timeline, (b) (4) -402; response to IR #95

4 SUMMARY OF STUDY DOCUMENTS

4.1 Effectiveness of (b) (4) vaccine against chikungunya virus disease in adolescent and adult population in endemic areas of Brazil. Observation Study Protocol (b) (4) -402 – November 24, 2022, Version draft 0.4; September 7, 2023, Version 4.0; September 28, 2023, Version 4.1

4.1.1 RWE Summary/Rationale

A Phase 4 observational study has been proposed by the Sponsor to confirm the vaccine effectiveness (VE) of (b) (4) in populations aged 12 years and older in disease-endemic areas of Brazil. Rationale for conducting this study in Brazil is described in the Background section of this memo.

4.1.2 RWD Evaluation and Management

The Sponsor states they will submit a data management plan (DMP) prior to initiating the study, and it will describe all functions, processes, responsibilities, and specifications for data collection, data storage, quality checking, transfer, cleaning, and validations.

A workflow diagram to manage the data for this study provided by the Sponsor:



Figure 1. Study data management flowchart

This flow chart does not illustrate all data collection processes. Data will be collected from various Brazilian health database and questionnaires given to the study participants. All data will be manually entered into an electronic Clinical Record File (eCRF) as part of the study database.

- See Reviewers' Comments #8 and 16 (IR50), pages 29 and 35, respectively, of this memo regarding data collection and management concerns. Also refer to Comment #1 in IR85, page 36 regarding the use of the participant questionnaire as the primary source of demographic and clinical data.
- A Data Management Plan was not submitted despite several requests. See Reviewers' Comment #1a, IR 89, on page 35 of this memo regarding the timeline for submission of the DMP.

4.1.3 Key Study/Research Question(s)

The main objective of the study is to assess the effectiveness of (b) (4) vaccination in Brazil in the prevention of symptomatic laboratory-confirmed CHIKV cases after a single dose of (b) (4). A secondary objective is to estimate VE in the prevention of symptomatic laboratory-confirmed CHIKV cases after a single dose of (b) (4) in selected endemic areas of Brazil, stratified by age groups (12-17, 18-64, and ≥ 65 years). As an exploratory objective, the Sponsor will evaluate the VE by health status (with and without comorbidities).

4.1.4 Study Design & Methods

This is a test-negative case-control study which will be initiated after the implementation of the pilot vaccination program in selected municipalities, when vaccination coverage has reached 15-20% of the eligible population and an increase in CHIKV transmission has been detected through CHIKV routine epidemiological surveillance in the same areas. (See Reviewers' Comment #2, page 19 of this memo)

The Sponsor states that the study will comply with the basic principles of the classic case-control studies, namely, both cases and controls are subjects who visit the same institution for CHIKV-like symptoms; both cases and controls should have the same risk of exposure to CHIKV; and both cases and controls should be selected independently of the vaccination status.

Updated Version 4.0: The study will be initiated when vaccination coverage has reached 20% of the eligible population. If CHIKV case numbers start to rise, the sponsor may decide to start the study earlier as long as vaccination coverage has reached at least 15%.

- a. **Study Period/Setting:** The total study period is estimated to be approximately 18 months or until the sample size is reached. The study period can be modified depending on the epidemiological situation of CHIKV in the study areas. They expect the pilot vaccination program to begin in Brazil in 2025, after licensure.

Valneva states the study will be carried out in approximately five to six medium-sized municipalities in different regions of Brazil where the pilot vaccination program will be implemented. The selection of municipalities is based on the following criteria:

- Medium-sized municipalities (no more than 500,000 residents)
- Municipalities with a good relationship with local research institutions
- Municipalities with a good laboratory infrastructure for arboviruses diagnosis (i.e., possibility to perform CHIKV RT-PCR)
- Municipalities with a historical good performance of arboviruses surveillance (database quality, timelines of case notification, etc.)
- Historically proven successful vaccination campaigns (e.g., use COVID-19 vaccination campaigns as indicator)
- Previous successful experience with vaccination campaigns and successful interactions between the health and education sectors (mainly for children and adolescents' vaccination strategy)
- Potential for a chikungunya outbreak

The vaccine will be offered free of charge by the public health system to the residing population during the pilot vaccine roll-out of (b) (4). The expected indication is active immunization for prevention of disease caused by chikungunya virus in persons 12 years and older; contraindications are immunocompromised individuals.

- See Reviewers' Comment #7, page 27 of this memo regarding accessibility to the official databases and Comments #1, 2 and 3 (pg. 19 and 22) regarding concerns of the study setting and vaccine coverage.

The sponsor addressed these and other comments in Version 4.0 of the study protocol:

Updated Version 4.0: A feasibility assessment will be conducted for the identification of the study municipalities to determine the data representativeness, the data relevance, the data quality, and the accessibility to official databases such as GAL, NIP, and SINAN to demonstrate that each data source contains the detail and completeness needed to capture study population, vaccine exposure, key covariates, outcomes of interest, and other important parameters that are relevant to the study question and the TN design.

The municipalities will be selected according to a prediction model based on mosquito incidence rates (dengue incidence used as proxy) and historical pluviometry patterns. In addition to the criteria listed in Version 0.4, the following are included:

- Previous chikungunya outbreak(s) with the goal of selecting municipalities with limited seroprevalence of chikungunya.
- The CHIKV health seeking behavior during the previous years will be considered as an additional selection criterion.

- See Reviewers' Comment #1a and 1e of IR 89 (10/13/2023) on page 28 for request for timelines for an agreement with Brazil, access to data, and the feasibility assessment and the sponsor's responses.

b. Participants/Eligibility Criteria:

Inclusion criteria:

Participants must satisfy the following criteria for eligibility:

1. Male or female aged 12 years of age or above
2. Provisions of informed consent/assent
3. Reside in the selected municipalities of Brazil where the pilot vaccination program was implemented
4. Suspected CHIKV cases seeking medical care (outpatients or admitted or hospitalized with CHIKV-like symptoms or confirmed CHIKV infection including previously suspected, e.g., dengue or Zika cases seeking medical attention, admitted, or hospitalized)
5. Have performed RT-PCR testing to investigate CHIKV infection

The test-positive cases are study participants (vaccinated and unvaccinated) that meet the inclusion criteria (above), the case definition of CHIKV², and the test positive for reverse transcription polymerase chain reaction (RT-PCR) for CHIKV.

The test-negative controls are study participants (vaccinated and unvaccinated) that meet the inclusion criteria (above), the CHIKV-like case definition, and test negative for CHIKV RT-PCR when the sample was collected up to 5 days after symptom onset.

Exclusion criteria:

Participants that fulfill one or more of the following criteria will not be eligible for the study:

1. Individuals with a history of CHIKV infection before vaccination with (b) (4) that is registered in the medical records, in the SINAN database, and/or self-reported.
2. Individuals for whom it was not possible to perform a CHIKV RT-PCR.
3. Individuals with undetermined CHIKV results for RT-PCR or negative results for samples collected after six days of symptoms onset.
4. Individuals for whom data is incomplete on age, sex, location of residence, vaccination status, testing status or dates.
5. Individuals who were not eligible for vaccination with (b) (4) will also be ineligible to participate in the study, such as individuals with immunodeficiency or immunosuppression due to disease or therapy.
6. Individuals who have been vaccinated with an investigational CHIKV vaccine or with a licensed CHIKV vaccine other than (b) (4) .

Updated Version 4.0/4.1: The sponsor revised the following inclusion and exclusion criteria according to our comments:

Inclusion criteria:

4. Suspected CHIKV or other arboviruses (e.g., Dengue or Zika) cases have performed RT-PCR testing to investigate CHIKV infection.

² Cunha RVD, Trinta KS. Chikungunya virus: clinical aspects and treatment - A Review. *Mem Inst Oswaldo Cruz* 2017; **112**(8): 523-31.

Exclusion criteria:

1. Individuals with a history of CHIKV infection before vaccination with (b) (4) self-reported or registered in the GAL or SINAN databases or in the sero-survey database. Note: Valneva will use the sero-survey results to perform additional analyses but will not exclude participants based on their sero-status from that survey (per telcon on 9/26/2023).
2. Individuals without a CHIKV RT-PVR test registered in the GAL database.
3. Individuals with undetermined CHIKV results for RT-PCR.
4. Individuals for whom data is incomplete on age, sex, municipality of residence, vaccination status, testing status date and date of symptoms onset.
5. Individuals who were not eligible for vaccination with (b) (4) according to contraindications of the vaccine per the local marketing authorization will also be ineligible to participate in the study, such as e.g., individuals with immunodeficiency or immunosuppression due to disease or therapy.
7. Self-reported receipt of donor blood within 90 days prior to vaccination.
8. Individuals who have been away from endemic area for 8 consecutive days during the 12 days prior to symptom onset.

As per the sponsor: *Eligibility criteria are intended to be aligned with the contraindications for the vaccine once approved in Brazil; thus, the criteria above may be adapted in the final protocol.* (Telcon 9/22/2023)

- See Reviewers' Comments #9b, 9c, and 12, page 30 and 33 of this memo regarding inclusion and exclusion criteria.

c. Exposure of interest/ascertainment: Vaccination status is considered exposure and it is determined by vaccinated (had received a single dose of the (b) (4) vaccine 14 or more days before onset of symptoms), partially vaccinated (had received a single dose of the vaccine up to 13 days before onset of symptoms), and unvaccinated (did not receive a single dose). An official vaccination card serves as the primary source of information on vaccination status. Participants shall be informed in the informed consent/assent form that, where appropriate, these sources will be accessed to confirm their vaccine status. Documentation on vaccination should include the date of vaccination, the brand name of the vaccine, and the batch number of the vaccine.

Updated Version 4.0: The primary source of vaccination status will be the SI-PNI at municipal level. Vaccination status will also be verified during the interview of participants. A secondary source is the individual immunization card and vaccination self-report (b) (4) requested during the participant interview.

d. Variables:

- Outcomes: CHIKV infection (lab-confirmed), CHIKV symptoms
- Exposure: Vaccination status (vaccination against CHIKV with (b) (4) and time of vaccination): vaccinated, partially vaccinated, or unvaccinated
- Predictors: Vaccine effectiveness
- Potential biases/confounders: Age, sex, calendar time, presence of comorbidities, health status, and geographical area.
- Diagnostic criteria (if applicable): Dates of RT-PCR tests and results, date of symptoms onset, severity of symptoms, and treatment site.

- See Reviewers' Comment #12, page 33 of this memo regarding previous CHIKV exposure and Comments #9a, page 29 regarding variables and effect modifiers.

Some variables were revised in Version 4.0:

Updated Version 4.0: Potential confounders: Age, sex, presence of comorbidities.

Effect modifiers: To be provided in the SAP.

- e. **Outcomes/ascertainment:** The outcome of interest is the detection of acute CHIKV in (b) (4) -vaccinated and unvaccinated study participants with CHIKV-like symptoms confirmed by RT-PCR. Ascertainment of CHIKV RNA positive samples via the Brazilian laboratory database.
 - See Reviewers' Comment #6, page 26 of this memo regarding lab validation.
- f. **Data Sources, Settings & Collection:** Official Brazil CHIKV surveillance data sources will be used to ensure that controls are residents of the same area as the cases and to identify the health facility where both cases and controls seek care for symptoms. To identify potential participants, data from Brazil's databases, the Notifiable Diseases Information System (SINAN database) and Laboratory Information System (GAL database), will be used. Questionnaires will be given to participants to collect demographic, medical history, CHIKV signs and symptoms, and knowledge of CHIKV infection.
 - See Reviewers' Comments #7 and 8, page 27 and 29 of this memo regarding access to these databases; Comment #1 in IR85, page 36, and Comment #1 (IR89) on page 35 of this memo.

The sponsor has reported that data collection (enrollment of subjects) will start when the pilot vaccination program with (b) (4) has reached 15 to 20% of vaccine uptake in the selected areas of Brazil. However, a lower vaccination coverage might be considered depending on the epidemiological status (sporadic epidemic occurrence and uncertainty about the duration of the outbreak) of CHIKV in the selected areas.

- See Reviewers' Comment #2b, page 19 of this memo regarding vaccine coverage.

Designated study team members at the municipal reference laboratory will identify potentially eligible individuals who were tested by CHIKV RT-PCR registered in the GAL database. A member of the study team will assess the inclusion and exclusion criteria of potential participants to classify them into cases (with positive CHIKV RT-PCR) and controls (with negative CHIKV RT-PCR). Once the inclusion and exclusion criteria for eligible participants have been verified, a member of the study team will enter their data into the study database.

The individual vaccination card will be used as the primary source to assess vaccination status. The immunization database will be used to check the vaccination status of all vaccinated individuals. To obtain access to the immunization database the study team will coordinate with the local health authorities.

- See Reviewers' Comment #7 (IR50), page 27 of this memo regarding accessibility to the databases and Valneva's response to IR 95 on page 28 regarding the sponsor's timelines.

The Sponsor plans to collect participant's demographic characteristics, CHIKV vaccination status, history of comorbidities, and medical treatments, as well as individual preventative measures such as personal use of repellents and active monitoring of mosquitoes.

- See Reviewers' Comment #1 (IR85), page 36 regarding the collection of participant demographic and medical data via a questionnaire.

A study team will collect information on demographic data, vaccination status, presence of comorbidities, assessment of inclusion and exclusion criteria for both cases and controls, and reasons for non-participation and manually enter into an eCRF for the study database.

- See Reviewers' Comments #7 and 8 (IR50), page 27 of this memo regarding data collection concerns and the blinding of certain study team members.

Updated Version 4.0/4.1: GAL database will be used as the primary source to identify potential participants undergoing testing for CHIKV, with participants being considered cases or controls based on the results of the RT-PCR test for CHIKV infection. A study member will have remote access to GAL's database using a password, who will gather the potential cases and controls from the laboratory data (GAL database) into the study database. The SINAN database will be used as a back-up source to verify demographic data, medical history, and contact information of participants. The National Immunization Program Information System (SI-PNI) will be the primary source to determine vaccination status at the municipal level. The study team will coordinate with local health authorities prior to the start of data collection to obtain access to the SI-PNI database.

A structured questionnaire (personal interview) will be the primary data source for demographic data, presenting signs and symptoms, comorbidities, whether they have received the (b) (4) vaccine and whether they have any immunosuppressive diseases. According to the Sponsor (telcon 9/23/2023), a medically trained study member will administer this interview questions to the participant.

All data required for the study will be collected using an eCRF and stored in a study-specific database. These data include information on demographics, vaccination status, presence of comorbidities, assessment of inclusion and exclusion criteria for both cases and controls, and reasons for non-participation. Information on the use of protective measures to avoid mosquito bites, and the participant's knowledge of CHIKV will be collected for an exploratory analysis.

A unique code (national identification number) will be used to identify the participant in all study data sources. The team member who will classify the cases and controls will be blinded to the vaccination status of the eligible participants, to ensure that the controls are selected independently of the vaccination status and prevent introducing bias to the estimation of VE.

- g. Potential Bias/Control for Confounding:** Variables that could be associated with both exposure (CHIKV vaccination) and outcome (CHIKV infection) were identified a priori such as age, sex, health status (presence of comorbidities), geographical area, and calendar time.

The Sponsor proposes the following to control for confounding:

- Age: categorization of age as a continuous variable should also be considered in the analysis, as it could lead to residual confounding, the importance of which is greatest in VE studies when small changes in age correspond to large differences in immunologic competence. *Version 4.0:* Age will be also analyzed as a categorical data.
- Health care-seeking behavior in terms of vaccination and care-seeking for symptoms
- Calendar time: since vaccination uptake and risk of disease may vary over time, in this study the controls will be selected in the same epidemiological week (EW) in which the case was detected and failing that, in one EW before or one EW after the identification of the case.
- Patient's history of previous exposure to CHIKV: given that it is believed that exposure to CHIKV induces lifelong cellular and humoral immunity, the disease exposure (CHIKV confirmed by RT-PCR or suspected) will be assessed.

Additionally, to minimize biases in this study for lack of data on comorbidities and misclassification related to the status of previous CHIKV infection, the study team will cross-check the participants' statement with SINAN database and/or medical records.

Potential bias issues for this study design include the lack of randomization of persons to vaccination, underreporting of cases in real-world settings, differential previous exposure to CHIKV, which CBER noted as a major potential source of bias.

Quantitative Bias Analysis (internal): The RWE review team (primarily Dr. Rodriguez Messan) performed a QBA to assess the potential biases from the TND study on the VE study results that may be caused by differences in seroprevalence rates (i.e., prior CHIKV infections) among vaccinated and non-vaccinated. We assumed the same sample size as proposed by the sponsor (401 cases and 802 controls) for easy comparison. The vaccine coverage was assumed to reach 15%. Four scenarios were considered for assumed VE (50%, 60%, 75%, 90%). We tested a range of prior infection rates (10-30%) with different distributions among vaccinated and un-vaccinated (e.g., vaccinated with prior infection of 10%, while non-vaccinated at 20%). Our results indicated that 1) bias may be large if prior infection rate among the un-vaccinated is larger than for vaccinated; 2) bias may be minimal for any prior infection rate when assumed VE is high; 3) bias may be minimal if both the vaccinated and un-vaccinated have the same prior infection rate regardless of true VE. Overall, bias on VE study results may be minimized if both vaccinated and unvaccinated individuals have similar prior infection rate.

Sero-survey Proposal: In response to our concerns about this potential bias, Valneva submitted a brief sero-survey outline in an annex to (b) (4) -402. They proposed to collect dried blood spots from 2000 individuals in a targeted municipality; 1000 who are receiving vaccinations and 1000 in shopping centers for the

unvaccinated. According to the Sponsor, a concept document for this sero-survey study will be submitted by December 30, 2023 and the draft protocol will be submitted by June 30, 2024. The purpose of the sero-survey is to ensure that both vaccinated and unvaccinated people have similar serostatus and the survey will also be used as part of the feasibility study for each municipality.

- See Reviewers' Comment #2, 4 and 12 (IR50) on page 19, 23 and 33 of this memo regarding previous exposure to CHIKV and the proposed sero-survey; Comment #2 in IR85 on page 37 and Comment 1d in IR 89 (10/13/2023) on page 34.

Updated Version 4.0/4.1: Exposure to disease will be also considered as a confounding variable. Exposure to the disease will be assessed by interviewing the participant, verifying the information in the laboratory database (GAL) and SINAN database and the results of a serological test (b) (4) performed during the serological survey during the implementation of the pilot vaccination program. Prior CHIKV infection of individuals who were not documented as seropositive in the sero-survey may be an unmeasured confounding. The sponsor plans to perform sensitivity analysis to assess the impact on the VE from these individuals.

- h. **Study Size:** Considering an estimation of 75% VE, a two-sided hypothesis test (H_0 : $OR = 1$; H_A : $OR = 0.25$); a type I error $\alpha = 0.05$; 80% for the power test; a ratio of 1 case to 2 controls; a proportion of 20% of cases in vaccine group under H_0 , and $R^2=0.81$, it was obtained a sample size of 1203 participants, using the software GPower. Applying a rate of loss of 10%, the overall sample size is 1338.

- Refer to Statistician's review memo for issues related to study size and power.

Updated Version 4.0/4.1: Null hypothesis (H_0): The lower limit of the 95% CI of the $VE \leq 35\%$; Alternative hypothesis (H_a): The lower limit of the 95% CI of the $VE > 35\%$. This results in a number of 401 cases and 802 controls needed to reject the null hypothesis with 98% and 99% power, for vaccination coverages of 15% and 20%, respectively. To compensate for a potential sample loss of up to 10% of the subjects, the target sample size was increased to 446 cases and 892 controls (1,338 participants).

- i. **Data Analyses:** The Sponsor will develop a full statistical analysis plan (SAP) prior to the conduct of the analysis and both the SAP and the currently reviewed protocol will be amended as needed according to the epidemiological situation of CHIKV in the selected study regions of Brazil.

Demographic and baseline characteristics, including distribution of subjects enrolled by study center will be tabulated by group and overall. Continuous variables will be summarized with descriptive statistics and categorical variables with frequency counts and percentages.

Crude and adjusted VE estimates (compared to fully vaccinated) will be estimated to address primary and secondary objectives. The VE will be estimated with the formula: $VE = (1-OR) \times 100$, where OR denotes the odds ratio for CHIKV vaccination amongst CHIKV-positive cases against CHIKV-negative controls. The OR will be unadjusted for crude VE estimates. For adjusted VE estimates, multivariate logistic

regression models will be used to obtain confounder-adjusted OR which will then be used in the VE calculation with a 95% CI.

To monitor for a potential waning of the vaccine effect, the distribution of breakthrough cases will be tabulated by number of months since vaccination. A seasonal VE estimate will be produced for each of the CHIKV seasons covered by the study.

Detailed statistical methodology which includes the full multivariable model specification, sensitivity and subgroup analyses will be pre-specified in the SAP.

- Refer to the Statistician's review memo for issues regarding the analyses and the SAP. Also refer to Comments #1 in IR 89 (10/13/2023) for submission of the SAP.

4.1.5 Results

a. Key Results:

The Sponsor has not initiated the study and therefore no results are available.

Limitations: The Sponsor has identified some limitations of the research methods. The Sponsor acknowledges that a TND case-control study is susceptible to bias due to the lack of randomization of persons to vaccination, underreporting of cases in real-world settings, and other issues. However, the Sponsor chose to use a TND because it is less susceptible to bias due to misclassification of infection and to confounding by health care-seeking behavior, relative to traditional case-control or cohort studies. In this study, age is identified as a confounder since it is likely associated with the odds of being vaccinated (exposure) and infected (outcome). Another potential confounder recognized by the Sponsor is the differential health care-seeking behavior in terms of vaccination and care-seeking for symptoms that can occur due to the wide variation in CHIKV symptomatology, which would affect the probability of being tested.

Our review also identified the potential bias of differential serostatus between the cases and controls. This issue was resolved with the Sponsor's proposal for a sero-survey in the intended municipalities of the study. Refer to page 13 of this memo regarding the sero-survey.

Due to these limitations and other concerns in the IRs, the sponsor proposed a sero-survey and a second study, nested within the study (b) (4) -402. The second confirmatory study was later submitted as a pragmatic RCT design in (b) (4) -404 to evaluate both safety and effectiveness postmarket.

4.1.6 Overall Study Assessment for (b) (4) -402

The study protocol is well-written and contains some detailed information about the rationale, study design, limitations of research methods, and ethical considerations, however, both the RWE and clinical reviewers had numerous concerns about this TND study serving as the only VE evidence to confirm clinical benefit. The RWE reviewers also had concerns about seroprevalence in the vaccinated and non-vaccinated populations introducing a bias via differentials in serostatus. Throughout the review cycle, the sponsor updated the study protocol to incorporate CBER's concerns and as

mentioned above, a sero-survey (described in an Annex to 402), feasibility studies, and a second study were proposed to address many of the concerns, providing more robust evidence of clinical benefit.

- See Reviewers' Comment #4 (IR77), page 23 of this memo regarding additional proposed studies.

4.2 Trial of the Effectiveness and Safety of (b) (4) Vaccine Against Chikungunya Virus Disease in an Endemic Country: A Pragmatic Randomized Controlled Trial Concept Document (b) (4) -404, September 2023, V3.0

4.2.1 RWE Study Protocol Summary

This section of the memo provides only a summary of the proposed study, as it was primarily reviewed by Drs. Forshee and Izurieta for effectiveness, DABRA DHT for the DHT elements, and OVR Clinical reviewers for the safety elements of the study. RWE comments are captured in the Reviewers' Comments section of this memo (page 18).

4.2.2 RWE Study Rationale

This Phase 4 observational study is a supplementary study being proposed by the Sponsor to provide additional confirmatory evidence for the safety and effectiveness of (b) (4) in an endemic population (TBD). After numerous IRs to address issues with the (b) (4) -402 TND VE study in Brazil, FDA requested a second study, agreeing to a pragmatic RCT design, to provide more robust data to confirm the VE of the Chikungunya virus vaccine; this study, 404, was later resubmitted to include a safety element.

4.2.3 Research Question and Objectives

To evaluate the effectiveness and safety of a single dose of (b) (4) vaccine in preventing symptomatic virologically-confirmed Chikungunya virus (CHIKV) disease among adults in an endemic country.

Primary objective:

1. To assess vaccine effectiveness (VE) in preventing symptomatic virologically-confirmed Chikungunya virus (CHIKV) disease among adults, in (b) (4) vaccinees compared to unvaccinated control participants during the same trial period, both overall and by age groups.

Secondary objective:

1. To evaluate VE against symptomatic probable or suspected CHIKV disease among adults, in (b) (4) vaccinees compared to unvaccinated control participants during the same trial period, both overall and by age groups.

2. To assess (b) (4) safety, in a subset of (b) (4) vaccinees compared to control participants during the same trial period.

Exploratory objective:

1. To examine the vaccine's effectiveness across the initial and subsequent transmission seasons.

2. To evaluate VE against occurrence of chronic arthralgia in confirmed and suspected cases of CHIKV disease.

3. To assess the impact of CHIKV infection disease on Quality of Life (QoL).

4.2.4 Study Trial Design, Endpoints & Assessments

This is an individual-level randomized, observer-blind, controlled trial conducted across multiple centers in an endemic country to assess the CHIKV (b) (4) vaccine safety and effectiveness against symptomatic, confirmed, and suspected CHIKV disease. Participants will be assigned at random to either the (b) (4) vaccine or a placebo/active control in a 1:1 ratio. Trial size is calculated to be 21,828.

Baseline Assessment: A baseline assessment will be conducted to measure participant's health status at the start of the study.

Primary Effectiveness Endpoint and Assessment: The trial will evaluate effectiveness by passively monitoring for CHIKV cases among participants. For the primary objective, virologically-confirmed chikungunya disease cases by RT-PCR occurring ≥ 2 weeks after vaccination.

Secondary Effectiveness Endpoint and Assessment: For the secondary objective, suspected cases of CHIKV disease occurring ≥ 2 weeks after vaccination. Participants attending the trial health clinics with chikungunya-like symptoms and CHIKV disease is suspected, a dried blood spot will be collected for virological confirmation.

Secondary Safety Endpoint and Assessment: Relevant safety information i.e., Adverse Events of Special Interest (AESIs) and Serious Adverse Events (SAEs) will be collected for all participants. AESIs include severe typical and atypical chikungunya-like illness, related hospitalizations, and prolonged arthralgia, starting 2-21 days post-vaccination. Participants will be followed up periodically after vaccination at weeks 1, 2, 4, 8, 12 and 24 via mobile app or phone calls to collect information about relevant adverse events, i.e., adverse event of special interest and serious adverse events.

Exploratory Endpoints and Assessment Methods:

Transmission Seasons: The primary effectiveness endpoint of symptomatic confirmed CHIKV disease will be examined by calculating the VE in different transmission seasons. This analysis will be repeated for the secondary effectiveness endpoint of suspected CHIKV disease.

Chronic Chikungunya Disease: Participants with either confirmed or suspected CHIKV disease will be followed up via mobile app or phone call at week 12 post-disease identification to collect data and determine whether the condition has resolved.

Quality of Life: The EQ-5D-5L, a European tool that assesses health-related QoL (HRQoL) across five key dimensions, will be incorporated into the trial's data collection to standardize health state measurements and evaluate the impact of health interventions.

- Refer to the Reviewers' Comments on page 38 below for issues communicated to the sponsor regarding RWE in this second confirmatory study.
- We noted that an eDiary will be used to collect safety data from participants via a phone app, therefore a Digital Health Technology reviewer was added to the review team. The DHT comment is included in this memo under the comments to study 404, on page 38 below.

4.2.5 Overall Study Assessment for (b) (4) -404

This study is a RCT with pragmatic elements to be conducted in a CHIKV endemic country and is a stronger design than the TND study. The two studies proposed will provide more robust confirmation of clinical benefit for the CHIKV vaccine. However, clinical reviewers had concerns regarding the safety evaluation in the proposed study and there were RWE concerns with the

sero-survey and the lack of a data management plan. These issues were communicated to the sponsor in IR #93, see page 37 below for RWE issues. Note: Since this an RCT, OVRP was lead reviewer on this protocol and their comments were also captured in IR #93.

4.3 Studies Milestone Dates

As requested in our IR 89, Valneva provided the following dates as milestones moving forward with developing study 402:

- Draft SAP by November 30, 2023
- Sero-survey design concept – December 31, 2023
- Engage with Brazilian MoH – January 1, 2024
- Pre-selection of municipalities – March 31, 2024
- Sero-survey draft protocol – June 30, 2024
- Quality Review of state laboratories – September 30, 2024
- Agreement with Brazilian MoH – September 30, 2024
- Approval for access to the databases – September 30, 2024
- Draft Date Management Plan – June 30, 2024
- Assessment of the data in the databases for fit for use and quality – February 28, 2025
- (b) (4) -402 Final agreed upon protocol – May 31, 2025

As requested in our IR 91 (10/17/2023), Valneva provided the following dates as milestones moving forward with developing study (b) (4) -404:

- Study implementation readiness verification (includes agreements with partners, IRB and local regulatory approvals, serological assay validation documentation, final SAP, final DMP) – June 30, 2025
- Draft (b) (4) -404 protocol – February 28, 2024
- (b) (4) -404 Final agreed upon protocol – September 30, 2024
- Study period – October 1, 2025 – July 31, 2029
- Submission of final study report – December 31, 2029

See Reviewer's Comment #1 (IR89) below on page 28 regarding the dates for data access and RWD evaluation for (b) (4) -402 and Comment #1 for IR# 91 on page 36 below.

5 REVIEWER'S COMMENTS

The RWE summaries of the study designs are described above. Several IR's were sent to Valneva to address our outstanding concerns with study protocol (b) (4) -402 which was submitted in the original BLA application (V0.4).

Based on our RWE review, the below comments on this study protocol (b) (4) -402 (version: Draft 0.4, dated November 24, 2022) were first sent to the sponsor on June 8, 2023 in IR #50. Their June 19, 2023 response to each issue is below each comment, as are the subsequent IRs and responses, including our comments on the updated V. 4.0/4.1 (submitted in September 2023).

Review comments for study (b) (4) -404 are in section 5.2 below. **Note:** FDA comments are in bold; Valneva comments are in italics.

5.1 STUDY PROTOCOL (b) (4) -402 (V0.4)

5.1.1 **Comment # 1 (IR50 6/8/23):** Please provide more information about how the vaccine will be offered in each participating municipality and if certain sub-populations are more likely to be vaccinated.

Sponsor's Response (6/19/23): The vaccine will be made available free of charge to the population of the selected municipalities through the local pilot vaccination program, in accordance with the Brazilian public health system (SUS). In Brazil, vaccination is not mandatory, and there is a risk that certain groups may not decide to get the vaccination, as expected in a real world evidence study conducted with limited ability to influence the selection of vaccinees.

However, there will be efforts undertaken in alignment with local Health Authorities to increase acceptance by appropriate community-directed communication efforts. Demographic information on vaccinated cases and controls can be contrasted against general population census information to determine representativeness. In addition, we consider it possible to monitor the vaccine uptake in sub-groups during the pilot phase.

Comment (IR61 7/7/23): The potential differences described in your response highlight the need for additional efforts to ensure that vaccinated and unvaccinated participants are comparable (except of course for the protection afforded by vaccination).

Sponsor's Response (7/17/23): We acknowledge the agency's comments and address this in our responses to other questions below.

- Comment #1 was fully addressed and resolved. See also their proposed sero-survey on page 13-14 to address our concerns of differences in sero-status introducing bias.

5.1.2 **Comment # 2 (IR50 6/8/23):** Please address the following concerns: **2b. The adequacy of 15-20% community vaccine coverage to initiate the study.**

Sponsor's Response (6/19/23): We calculated the vaccination rate in the target population necessary for a case-control study to estimate a vaccine effectiveness of 75% with the width of the 95% confidence interval of approximately 22% (see (b) (4) -402 study protocol for more details, section 4.8).

Based on our Phase 3 trials, where we have seen high seroresponse rates exceeding 97% using a very conservative definition of seroresponse (titer that resulted in sterilizing immunity in the passive transfer model, and exceeding a titer that was seen as protective in a seroepidemiological trial), we consider that the 75% VE is a conservative estimate. In addition, the resulting CI width of 22%, ranging between 26% to 17.6% depending on the coverage rate observed in the controls at the time of starting the study, translates to a lower bound of the 95% CI of 62 in the least favourable case. We consider that demonstrating VE with such a high lower bound of the confidence interval will still provide very robust evidence, and hence consider the vaccine coverage as indicated sufficient to start the study. Due to explosiveness and sporadic occurrence of outbreaks, we do not want to miss the opportunity to start the study in case we see CHIKV cases rising and hence need to allow for a certain flexibility.

Comment (IR61 7/7/23): Given the explosiveness and sporadic occurrence of outbreaks, and potentially low vaccination coverage, please consider the possibility of extending the study duration for additional seasons, if needed.

Sponsor's Response (7/17/23): We would like to reiterate that the study (b) (4) -402 will not be started before an at least 15% vaccination coverage has been reached by the means of the pilot vaccination program. The possibility of extending the (b) (4) -402 study duration is planned. If the sample size, i.e., number of cases is not reached within the planned timelines, study recruitment will continue until the required number of cases has been reached, including, if needed, a subsequent transmission season.

Comment (IR77 8/16/2023): Although we basically agree, please refer also to RWE comment #1. Specifically, on comment 1b, we are asking for prospective study participants to be serologically tested to assess prior CHIKV infection before they have the opportunity to get vaccinated or become study cases. Because a vaccine-induced immune response is indistinguishable from the immune response to a prior natural infection, samples for serological testing must be collected before or immediately after vaccination (i.e., prior to mounting a vaccine-induced immune response). Regarding vaccinees, collection of a sample for serologic testing from prospective participants on the same day of vaccination is thus an acceptable alternative to collection pre-vaccination. Such data should improve our understanding of potential differences in the rate of prior CHIKV infection between vaccinated and unvaccinated study participants. We recognize the difficulties in serologically sampling all study participants. Thus, alternatively, we would be willing to consider a proposal for serological sampling of a representative sample. In addition, please submit laboratory and epidemiological data showing absence of major CHIKV outbreaks in the prior years in areas in which the study is being planned.

- Valneva's proposed a sero-survey study, see description on pages 13-14. This issue is resolved.

Comment #2c. (IR50 6/8/23): How time-varying vaccine coverage and the potential for waning immunity could impact the study results and how you plan to account for waning immunity in your study.

Sponsor's Response (6/19/23): On the waning immunity, we are currently performing a prospective long-term persistence study (VLA1553-303, clinicaltrials.gov identifier: NCT04838444). It is evaluating the persistence of antibodies and long-term safety (SAEs) in a subset of participants of the pivotal Phase 3 study VLA1553-301. The primary objective of the study is to evaluate persistence of antibodies annually from 1 to 5 years after the single immunization with VLA1553.

Currently data are available at 1 year after vaccination in all 363 study participants. GMTs for CHIKV-specific neutralizing antibody titer remained stable from 6 months post-vaccination for this cohort (1070 at 1 year versus 1281 at 6 months), underscoring continuing protective immunity, and the seroresponse rate remained very high (99.5%). At the time of initiating the observational study (b) (4) -402 additional antibody persistence data (at least up to 3 years post vaccination) will be available from the ongoing long-term persistence study. We acknowledge the Agency's concern on waning immunity. We will take further data coming from study (b) (4) -402 into account and can modify or include

respective stratified analysis as necessary, because vaccination date and disease date will be collected. We will also analyse the proportion of breakthrough cases among vaccinated, by time since vaccination.

On the time-varying vaccine coverage, in the test-negative case-control (b) (4) -402 study design, the time-varying vaccine coverage can be neglected.

Comment (IR61 7/7/23): We agree that stratification by time since vaccination will be useful for analyzing waning of immunity. Please consider the implications on the study power.

Sponsor's Response (7/17/23): In order to monitor for a potential waning of the vaccine effect, the distribution of breakthrough cases will be tabulated by number of months since vaccination. A seasonal VE estimate will be produced for each of the CHIKV seasons covered by the study (if multiple seasons are needed). In addition, as an exploratory analysis we will also estimate the VE by weeks or months since vaccination and produce a plot showing VE by week/month, with its 95% CI. However, at this stage we do not plan to test any hypothesis concerning waning.

Comment (IR77 8/16/2023): We agree.

- This issue is resolved.

Comment #2d. (IR50 6/8/23): How the difference in CHIKV attack rates and vaccine coverage in the various municipalities could impact the VE calculations.

Sponsor's Response (6/19/23): The attack rate of CHIKV and the vaccination coverage will not have any impact on the VE due to the study's design, which is a case-control study with a ratio of 1:2, as per protocol. However, in the event of a low attack rate, the trial period would need to be extended in order to identify a sufficient number of cases. The VE will be calculated using the odds ratio through logistic regression, adjusting for confounding variables such as age, sex, comorbidities status, geographical area, and calendar time. An additional sensitivity analysis will be performed using a propensity score matching in order to have matched analysis, considering the geographic area and epidemiological period (Austin 2011; Austin 2011a; Bergstra et al., 2019; Rosenbaum et al., 1983; Parsons, undated). A detailed explanation of such methodology will be stated in the Statistical Analysis Plan.

Comment (IR61 7/7/23): Low vaccination coverage and variable attack rates might make it necessary to extend the study period, please consider this possible scenario. Please provide criteria to specify conditions under which the study should be extended for additional seasons.

Sponsor's Response (7/17/23): Indeed, we consider this a possible scenario. The conditions for study extension may comprise low vaccination coverage (below 15%), or too few cases, i.e. recruitment of mainly controls and the lack of real cases, might make it necessary to extend the study period.

Comment (IR77 8/16/2023): We agree.

- Issue is resolved.

- 5.1.3 Comment # 3 (IR50 6/8/23):** Often, people who want to be vaccinated are more likely or willing to use nonvaccine approaches to protect themselves from infection compared to people who do not want to be vaccinated. Please explain how you will match these kinds of behavior between vaccinees and non-vaccinees in the proposed observational study. Consider performing an additional analysis in a subpopulation restricted to individuals who had received Dengue vaccination (or other non-mandatory vaccines) in the past, to help decrease the likelihood of health seeking behavior differences between the vaccinated and unvaccinated population.

Sponsor's Response (6/19/23): In terms of mitigating the potential bias arising from the disparate probabilities of achieving the outcome between vaccinated and unvaccinated individuals due to factors unrelated to the direct impact of the vaccine, in this study, we will assume that the probability of vaccination is conditioned by the level of health awareness. To measure this, we will apply a standardized questionnaire to measure the level of knowledge about chikungunya and the use of preventive measures. The responses to the questionnaire will be analyzed to create a range between high levels of health awareness and low levels of health awareness. By including this covariate in the analysis model, we will be able to estimate the VE controlled for this factor.

Attached is a structured questionnaire to assess the knowledge and prevention measures among vaccinated and non-vaccinated participants.

Comment (IR61 7/7/23): We appreciate your plan to use a survey to collect additional information to address potential major confounders. However, we would like additional details of how you will assess the validity of the survey instrument and how the data will be used in your analysis. Please consider local factors such as socioeconomic status and cultural differences when designing and pretesting the questionnaire. We suggest that this questionnaire once finalized and properly tested should be administered to all study participants. Similar awareness of CHIK does not necessarily mean similar willingness or likelihood to get the vaccine or use of non-vaccine preventive approaches. The questionnaire should also include questions to measure the likelihood of being vaccinated with the CHIK vaccine when it is available, and what non-vaccine measures they usually use to prevent themselves from CHIKV infection. Please provide specifics on any algorithms you are planning to use for interpretation of survey responses.

Furthermore, depending on the final survey instrument proposed and intent of the data collected, additional Agency review may be required. Please refer to the FDA Guidance for Industry Patient-Reported Outcome Measures: Use in Medical Product Development to Support Labeling Claims (<https://www.fda.gov/media/77832/download>) and Clinical Outcome Assessment (COA): Frequently Asked Questions | FDA.

Sponsor's Response (7/17/23): We acknowledge the agency's feedback. If we will not be able to identify an available, validated questionnaire to address the agency's request, we will ensure necessary testing of a Valneva developed questionnaire. However, please note that this questionnaire is intended to be used as part of study (b) (4) -402 (test negative case control study) when participants already have been vaccinated in the pilot vaccination program. Therefore, questions on likelihood of being vaccinated will not be applicable in this setting. Kindly refer also to Company Response to IR#61 Question 4

The survey data will be analyzed by descriptive statistics and statistical inferences from ANOVA. In addition, multivariate regression techniques will be used to classify the patient willingness to take the vaccine based on their responses, the level of knowledge about CHIKV, and use of protective measures to avoid mosquito bites.

Comment (IR77 8/16/2023): More specifically, regarding the survey instrument, please ensure:

- a. The questions will assess your specific construct of interest; provide documentation on how this process will be operationalized.
- b. There is a valid language translation from English to the language(s) of each municipality and perform preliminary pilot testing.
- c. The survey is pilot tested in a sufficient representative sample of respondents to ensure reliability and validity of the overall questionnaire, and its ability to measure the construct.
- d. The algorithm utilized to produce a covariate in the model, based on this construct, is also validated, and provide the documentation.

Sponsor's Response: After a telcon with Valneva to clarify comments in IR77, they decided to forgo the survey instrument to determine participant knowledge of Chikungunya and the potential "healthy vaccinee" effect and instead add exploratory knowledge questions to the participant questionnaire. To further address CBER concerns regarding potential bias, they agreed to conduct a sero-survey in the municipalities where they plan to enroll participants.

As described on page 14, the Sponsor submitted sero-survey proposal to collect dried blood spots from 2000 individuals in a targeted municipality; 1000 who are receiving vaccinations and 1000 in shopping centers for the unvaccinated. A brief outline of the survey was added as an annex to the 402 study protocol. A concept document for this sero-survey study will be submitted by December 30, 2023 and the draft protocol will be submitted by June 30, 2024. The purpose of the sero-survey is to ensure that both vaccinated and unvaccinated people have similar serostatus and the survey may provide information about previous epidemics in that municipality.

- This issue is resolved.

5.1.4 Comment # 4 (IR50 6/8/23): We recommend you perform a Quantitative Bias Analysis (QBA) to evaluate the likelihood that differences in the proportion of undetected prior CHIKV infection between the vaccinated and unvaccinated in the source population could bias study results. Include a range of assumptions about the amount of undetected prior CHIKV infection. Use results of the QBA to produce likely lower and upper bounds for the potential bias. Also, as possible, use such assumptions to make corrections to your study power calculations, to decrease the likelihood of a type 2 error in your study.

Sponsor's Response (6/19/23): We performed a simple bias analysis (similar to the ones described in Lash et al., 2009). For simplicity, a non-matched case-control study was considered, easy to summarize in a 2 x 2 table. The number of enrolled cases and controls (after the 10% drop-out loss) was assumed to be as in the sample size calculation, 401, respectively 802. The vaccine coverage was assumed to reach 20%. Three scenarios were considered for the assumed VE (60%, 75%, 90%). Using the assumed VE and the total

number of cases and controls, the number of vaccinated and non-vaccinated cases and controls were derived for each scenario.

Then, three scenarios were considered for the cumulative percent of persons with a previous CHIKV undetected infection (15%, 30%, 50%). Since a CHIKV infection can only be contracted once, and the infection ensures lifelong immunity against re-infection (e.g., de Souza et al., 2023), it can be inferred that cases arriving in healthcare facilities, confirmed as having a CHIKV infection right then, could not have had a previously undetected infection.

Therefore, only the controls could have had a previous CHIKV infection. By assuming there's no relationship between the unknown previous CHIKV infection and the vaccination status, the correct number of vaccinated and non-vaccinated controls (those without a previous CHIKV infection) was calculated. The corrected VE with its 95% CI was estimated, using the same methods. As presented in the Quantitative Bias Analysis (Appendix 2), in all these scenarios, the presence of previously undetected infections will not bias the point estimates. The impact on the 95% CI will be relatively small, even for an assumed proportion of 50% previously undetected infections, making a sample size correction unnecessary.

Comment (IR61 7/7/23): There could be an association between prior CHIKV infection and likelihood of vaccination. For instance, if previously infected people are convinced that they are protected without vaccination, that could discourage vaccination among them. The opposite could occur if they believe instead that prior infection is not protective or is insufficiently protective. Also, if previously infected people are less “health seekers” (less prone to seek medical care when sick or to get vaccinated), they may be unaware of having had an infection and could also be less likely to be vaccinated anyway. Because prior infection not only alters the risk of disease but also the patient’s perception of disease severity and need for prevention, this could bias study results, depending on the scenario, in either direction.

Thus, finding, prior to Test negative design implementation, whether there are significant differences in the rates of prior CHIKV infection between the vaccinated and the unvaccinated would be important in order to reassure FDA that such potential imbalance is not a significant problem. Confirming prior infection, however, would, require serological testing of prospective participants, as requested elsewhere. Please let us know of your plans to address this concern. Although very useful, quantitative bias analysis may be insufficient to resolve this important concern without additional information on the potential magnitude of these imbalances. Because of CBER’s concerns regarding potential confounding by unmeasured differences in rates of prior CHIKV infection between vaccinated and unvaccinated participants, please also consider the possibility of restricting the primary analysis to municipalities that had not suffered any significant CHIKV outbreaks in prior years.

Serosurveillance data for your proposed study sites will be critical for determining the acceptability of your protocol to satisfy the PMR. Please describe the status of your agreements with national and local health authorities in Brazil, as well as respective Institutional Review Boards, for implementation of this protocol for your proposed study sites. Furthermore, we suggest that you incorporate study sites in additional Chikungunya endemic countries, as consistent with your development program.

Sponsor's Response (7/17/23): We agree with the agency's view on health seeking behavior in the first paragraph. The municipalities will be selected according to a prediction model based on mosquito incidence rates (using dengue incidence as proxy) and historical pluviometry patterns. Then, the CHIKV behaviours during the previous years can be considered as an additional selection criterion.

Additionally, before the pilot vaccination program, in order to understand the seroprevalence of CHIKV in each municipality, we propose to implement a sero-survey to develop an understanding of the levels of previous exposure to CHIKV among the population in the respective municipality. At the same time, among the survey population, we would implement the questionnaire to measure likelihood of health seeking behaviour, likelihood of vaccination and use of other protective measures to avoid any vector-borne disease. These characteristics will be assessed by serostatus (positive or negative, and the potential different behavior in persons with known prior CHIKV infection) and will be used as a co-variate to be adjusted at the study (b) (4) -402 analysis.

In the absence of a final, agreed protocol, discussions with the national health authorities are taking place at high-level and further details will be provided as soon as the potential municipalities were selected. All required agreements (regulatory, ethics) including with the local health authorities will be obtained prior to the study onset.

Sero-survey data will be crucial and will be discussed once we propose the sero-survey design concept to the agency in Q4/2023.

We will consider involvement of other chikungunya endemic countries than Brazil for demonstration of effectiveness for the second effectiveness study currently under discussion. At the planned start of study (b) (4) -402 the vaccine will not yet have been licensed in other chikungunya endemic countries; in addition, the entire design and execution of study (b) (4) -402 has been developed to make best use of RWE data collection in Brazil (i.e. use of existing/ routine assays, established databases).

Comment (IR77 8/16/2023): You have not provided convincing evidence that this TND study, as proposed, can appropriately determine potential imbalances in prior Chikungunya virus (CHIKV) infection between vaccinated and unvaccinated study subjects, which could be a major source of bias.

In amendment 71 concerning the second confirmatory study, one of your proposals was a nested test negative study design in which participants are selected prospectively (i.e., prior to CHIKV vaccination or CHIKV disease onset) among individuals attending health centers which are chosen based on their potential for encountering Chikungunya cases. These participants would: (a) have blood spots taken at recruitment to determine serologically whether they may have had a prior CHIKV infection, and (b) as feasible, test-negative controls will be matched to test-positive cases based on age, group, sex, season, and location.

Further, you have indicated that you may also proceed to integrate a retrospective evaluation by healthcare providers of potential safety indicators into the evaluations of both cases and test-negative controls and subsequently associate this information with vaccination status. Please incorporate all of these elements from your proposed second confirmatory "nested test-negative study" into the (b) (4) -402 study protocol.

Sponsor's Response: After some discussion, the sponsor agreed to perform a sero-survey study described above and a second confirmatory study proposal submitted September 7, 2023 as a pragmatic RCT design in an endemic country, (b) (4) -404. This study is summarized on page 16 of this memo.

- This issue is resolved.

5.1.6 Comment #6 (IR50 6/8/23): Please note that the RT-PCR used to assess CHIKV infection status should be validated at all sites performing this test. We recommend the assay validation protocol be submitted to CBER for review before commencing the validation studies and that the validation report is approved by CBER before clinical sample testing begins.

Sponsor's Response (6/19/23): We appreciate the Agency's suggestion and understand the criticality of the issue. Our study will be conducted in a real-world setting and will leverage the data collected through the national surveillance laboratory system. Routine plasma and serum collection takes place in subjects with acute symptoms suggestive of arbovirus-like illness. The Brazilian lab network is well-structured and widely distributed across the country. It has established protocols in place for testing arboviruses using RT-PCR as the standard method. The sponsor asks the Agency to kindly acknowledge not submitting the RT-PCR protocol validation of each individual laboratory that is part of the lab network system for CBER's appreciation. In the Supplementary Material (Appendix 3), we provided a document that details the Brazilian laboratory network, the flow of samples from the health units, and the sample process flow and testing. At the time of the definition of the municipalities, we can perform a feasibility assessment to be sure that all labs have a minimal quality level required (considering that all public health labs should be certificated by ANVISA, International Organization for Standardization (IOS), and other regulators depending on the test that will be performed).

Comment (IR61 7/7/23): Although we acknowledge that the Brazilian laboratories have established protocols for testing CHIKV, the facilities that will be used as part of your study must demonstrate that the sample handling, testing, and reporting processes are validated. It is unclear from your response how many labs you expect to be utilized for the study, however, documentation indicating that the selected labs are proficient at CHIKV testing should be provided and made available for auditing purposes.

Sponsor's Response (7/17/23): The Brazilian public laboratories network has one central lab in each state and one reference lab for arboviruses investigation at the national level. For the pilot vaccination program (and consequently participation in study (b) (4) -402), we are aiming for the selection of municipalities from one or two states, thus, we expect to involve up to 2 public laboratories.

As part of the municipality's selection, we intend to include the verification of the availability of documentation demonstrating the lab process validation, as well as accessibility of such documentation for auditing purposes. If it is not already performed by the central lab, an agreement with the reference lab will be done to set-up for the validation before study (b) (4) -402 starts.

Comment (IR77 8/16/2023): We acknowledge receipt of the information provided on the Brazilian public laboratories network and your intention to include verification of

documentation demonstrating the lab process validation. As previously communicated in IR#50, the RT-PCR assay that will be used to assess CHIKV infection status should be validated at all of the laboratories prior to the start of sample testing. Review and approval of the assay validation will be required in support of regulatory decisions involving the use of RT-PCR results. We recommend that testing labs be identified prior to study start so that the assay validation protocols can be submitted to CBER for review. It will be necessary to include information on the handling and storage of samples at the collection sites, transport of samples to the testing labs, and study plans that ensure comparable performance of the assay at different sites.

Sponsor's Response: The Sponsor stated that samples will be handle appropriately and according to laboratory requirements. This question was posed by the CMC reviewers; refer to the CMC review memo for additional laboratory validation information.

- This issue is resolved.

5.1.7 Comment #7 (IR50 6/8/23): Please confirm that the study team has gained the necessary approvals for access to the official Brazilian databases, including the clinics and patient data, approvals from local and institutional review boards (IRB approvals), and provide the expected timeframe for commencement of the study.

Sponsor's Response (6/19/23): The municipality health service has already access to all databases needed, which means SINAN (surveillance system), GAL (laboratory system), and SI-PNI (vaccination system).

However, a more detailed assessment regarding the level of access will be performed as part of the feasibility evaluation during the selection process of municipalities. If necessary, a broader access to the local services will be requested from the respective authorities. All data required for the study will be collected using an eCRF (manual transcription) and will be stored at a specific study database. Other approvals (IRB) will be requested as soon as the final protocol is ready. However, a very similar program had been conducted for Dengvaxia in 2016-2019, using the same data sources; there is therefore precedent for conducting this type of studies and data access (de Moraes et al., 2022). It is expected that the pilot vaccination program may start in 2025 depending on the vaccine registration by the Regulatory Agency in Brazil.

Comment (IR61 7/7/23): Please ask for necessary access to the local services database so all covariates needed for the study can be analyzed.

Sponsor's Response (7/17/23): We acknowledge the comment and will ask for necessary access to local services database in due course.

Comment (IR77 8/16/2023): Please ensure the specific variables necessary for your analyses, required by your DMP, are available and comparable in each municipality.

Comment #1a (IR89, 10/13/2023): In this IR, we requested the milestone dates for submission of their DMP, SAP, data access, agreement with Brazil MoH, and other important aspects of their study development plan.

Sponsor's Response (10/19/2023): Valneva provided a timeline for milestones. These are reported in Section 4.3, page 18.

Comment #1 (IR 95 10/25/23): You provided timelines for your agreement with Brazil MoH (September 30, 2024), approval for access to the government databases (December 31, 2024), submission of a draft Data Management Plan (DMP) (December 31, 2024), the final agreed upon protocol (May 31, 2025), and the assessment of the data in the databases for fitness of use and quality (June 30, 2025). We understand that each subsequent step is contingent on the agreement with the MoH and data access, however we are concerned about the timeliness of your database evaluation to ensure the data are relevant and reliable for your study. FDA recognizes that evaluation of relevant data sources or databases is an important step in the design of a study and in evaluating a study's feasibility and should be performed earlier during your study development activities. Additionally, your DMP can be an evolving document and updated as needed as you prepare the study protocol. Please explain why you are anticipating the evaluation of data sources will be completed after the study protocol is finalized. You may refer to the FDA Guidance, Considerations for the Use of Real-World Data and Real-World Evidence to Support Regulatory Decision-Making for Drug and Biological Products, August 2023 at <https://www.fda.gov/media/171667/download> for more information regarding RWD evaluation.

Sponsor's Response (10/31/23): Valneva responded by providing new timelines for data access and evaluation:

Key activity	Tentative timeline for completion
Complete municipality selection & feasibility	30-Sep-2024
Engage with Brazilian MoH based on pre-final protocol (agreed with FDA; translated to Portuguese)	1-Jan-2024
Pre-selection of municipalities based on modelling to predict areas of high ChikV transmission activities	31-Mar-2024
Agreement reached with Brazilian MoH on supporting the pilot vaccination program (independent of municipality selection)	30-Jun-2024
Approval to access the databases (after agreement with MoH and after municipality selection)	30-Sep-2024
Initial engagement with relevant IRB(s) re (b) (4) -402 and complementary studies	30-Sep-2024
Quality review/ evaluation of state laboratories (for confirmation ChikV infection, PCR), incl. verification of availability of documentation demonstrating the lab process validation, accessibility of such documentation for auditing purposes	30-Sep-2024
Agreement reached with Brazilian MoH (based on near-final FDA agreed protocol) and public health systems of each municipality (following pre-selection of municipalities) on participation in pilot vaccination program and (b) (4) -402	30-Sep-2024
Assessment to determine the data representativeness, the data relevance, the data quality and the accessibility of GAL and SINAN (following approval for access)	28-Feb-2025

Reviewer's Comments: We note that the Sponsor adjusted the dates for data access and evaluation, as well sending an earlier draft DMP to CBER.

- This issue is resolved.

Comment #1e (IR89, 10/13/2023): Feasibility Assessment: You state on page 44, “For the identification of the study municipalities, we will conduct a feasibility assessment to determine the data representativeness, the data relevance, the data quality and the accessibility to official databases such as GAL (Laboratory Environment Management System), National Immunization Program, and SINAN (Notifiable Diseases Information System) in order to demonstrate that each data source contains the detail and completeness needed to capture the study population, exposure to (b) (4) vaccine, key covariates, outcomes of interest (RT-PCR for CHIKV results), and other important parameters (e.g., timing of exposure, timing of outcome) that are relevant to the study question and the test negative design.” You also include evaluation of laboratory processes and IRB considerations for municipality assessments. Please describe all activities necessary prior to trial implementation and include your expected timeline for completion of these activities, including initiation of your feasibility assessment.

- The Sponsor provided an overview (table) of key activities that need to be assessed and complete as they move forward implementing the study. The timeline of these activities is listed above. This issue is resolved.

5.1.8 Comment #8 (IR50 6/8/23): In Section 4.10.2 Data Collection of your study protocol, you mention that the designated study team will assess the inclusion and exclusion criteria of potential participants to classify them into cases and controls. In addition, the same study team will ascertain vaccination status by checking the vaccination card. Please be advised that the study team members who perform these two tasks should be blinded to each other, to ensure that the controls are selected independently of the vaccination status and therefore prevent introducing bias to the estimation of VE.

Sponsor's Response (6/19/23): We acknowledge the comment and will set it up accordingly in an updated (b) (4) -402 study protocol version.

Comment (IR61 7/7/23): We agree.

- This issue is resolved. The updated 402 protocol V4.0, Sept 2023, included questionnaires to collect participant demographic and pertinent health data which clarified that these data are not being collected from electronic health records (see Comment #1 IR85, page 36).

5.1.9 Comment #9 (IR50 6/8/23): Please explain the following from your study protocol:
9a. How you determined sex, age, and geography are potential confounders and your method for controlling these variables in your analyses (page 22).

Sponsor's Response (6/19/23): According to the references provided below these potential confounders may be considered as highly correlated with cases of the disease and vaccine responses. Age (Sissoko et al., 2010; Correia et al., 2021; Oh et al., 2019) and

sex (Sissoko et al., 2010; Correira et al., 2021; Delgado-Enciso et al., 2018) might be suggested as confounders since medical literature describes age and sex as common confounders to be discriminated in observational vaccine studies (Remschmidt et al., 2015; Souza et al., 2023). Geographic location with different socioeconomic status also presents variances in controlling comorbidity rates, which affects vaccine responses and clinical outcomes (Remschmidt et al., 2015; Souza et al., 2023). Therefore, the statistical adjustment aims to manage the data variability. The potential confounding variables will be included as independent covariates in the logistic regression model to adjust the VE estimation.

Comment (IR61 7/7/23): In addition, because geographic region could affect the likelihood of prior infection and other important variables, it could act as an effect modifier, please consider matching or another similar approach to address issues related to geographic region.

Sponsor's Response (7/17/23): During the analysis, cases and controls will be strata matched by municipality. A conditional logistic regression model will be used to account for the matching and will be included in the SAP

Comment (IR77 8/16/2023): We agree.

- Issue is resolved.

Comment # 9b (IR 50 6/8/2023): If travel to areas outside of the endemic study areas and receipt of donor blood products will be added to the exclusionary criteria, as these subjects may have different risks of exposure and outcome.

Sponsor's Response (6/19/23): We have considered introducing an exclusion criterion related to traveling to areas outside of the epidemic study area. Our concern is the impact of this exclusion criterion on the potential cases since we cannot ensure that those travelers will not be exposed to local risk – some examples: 1. subject who lives in the study municipality but works in another city and performs daily travel; 2. subject who stays in the study municipality during the week and travels at the weekends; 3. how much time should we consider for the traveler to stay outside of the municipality to be sure that he/she is at a lower risk? We are reasoning that there are many variables to consider to be sure that the subject is exposed to a lower risk. So, this exclusion criterion in general could limit our sample size and reduce the inclusion of potential cases in the study – therefore, we propose to not implement this exclusion criterion in the study protocol. As we are performing a study in a real world setting we need to be careful in choosing which variables we should control.

With respect to the use of prior blood products, we will establish an exclusion criterion based on self-reported receipt of donor blood within 90 days prior to vaccination. The main aim of the study is to understand the vaccine's effectiveness against vector-borne chikungunya, as opposed to non-vectorial transmission routes.

Comment (IR61 7/7/23): Please provide rules regarding exclusionary criteria for individuals who have been away from endemic areas for extended periods of time.

Sponsor's Response (7/17/23): In our considerations for definition of an exclusion criterion for individuals "who have been away from endemic areas for extended periods of time" we

suggest linking to the CHIK incubation period. CDC states the incubation period to be typically 3-7 days, with a range of 1-12 days (Source: Centers for Disease Control and Prevention, National Center for Emerging and Zoonotic Infectious Diseases (NCEZID), Division of Vector-Borne Diseases (DVBD), last accessed 12-Jul-2023). Therefore, we suggest $\frac{3}{4}$ of the incubation period range as definition for extended periods of time being away from endemic areas, i.e., 8 consecutive days within the 12 days prior symptom onset would be an exclusion criterion.

Comment (IR77 8/16/2023): We agree.

- This issue is resolved.

Comment #9c (IR50 6/8/2023): Why exclusionary criteria on page 10 includes negative results for samples collected after 6 days of symptom onset, and page 19 defines inclusionary test negative control as negative results for samples collected up to 5 days after symptom onset. We recommend that all RT-PCR samples be collected within 5 days of symptom onset for both cases and controls.

Sponsor's Response (6/19/23): We acknowledge the agency's recommendation but would like to suggest a different approach. We suggest staying with an exclusion criterion which will exclude individuals with negative results for CHIKV for samples collected outside 5 days of symptom onset from the controls. This will ensure that only individuals will serve as controls who indeed have not presented with CHIKV.

On the other hand, we suggest extending the time window for acceptance of individuals testing positive for CHIKV (i.e., cases) with samples collected within 8 days of symptom onset. This approach is supported by the observation that during the first 8 days of symptom onset, chikungunya viral RNA often can be identified in serum (CDC Yellow Book 2024, <https://www.cdc.gov/chikungunya/hc/diagnostic.html>).

This is also in line with Brazilian surveillance guidelines for samples collection and the expectation for a positive result from different tests that states that the RT-PCR is still detected until the 8th day of symptoms onset (see the Supplementary Material Appendix 3, Figure 2).

- Validation of the laboratory procedures will determine the timeframe for sample collection and symptom onset. Refer to CMC review memo for lab-related issues.

Comment (IR61 7/7/23): To avoid bias caused by differences in timeliness to seek health care when sick between cases (those who have the disease of interest) and controls (who have another disease), it is better to allow the same time intervals between symptoms onset and medical visit (testing date), for both cases and controls. Differences in timeliness to seek help when sick could, for instance, be associated with differences in likelihood to seek vaccination, which might bias study results.

Sponsor's Response (7/17/23): We suggest an exclusion criterion for controls which will exclude individuals for whom only negative CHIKV results for samples collected outside 8 days intervals between symptom onset and medical visit (date sample obtained, not testing date at the lab) are available.

For cases, we suggest acceptance of positive CHIKV results collected within 8 days of symptom onset and medical visit (date sample obtained, not testing date at the lab). This approach is supported by the observation that during the first 8 days of symptom onset, chikungunya viral RNA often can be identified in serum (CDC Yellow Book 2024, <https://www.cdc.gov/chikungunya/hc/diagnostic.html>).

Comment (IR77 8/16/2023): We agree.

- The updated 402 protocol V4.0, Sept 2023, included an updated exclusion criteria list for which Comment #2 was sent out in IR85 (page 37). This issue is resolved.

Comment #9d (IR50 6/8/2023): The following statement on page 23 of the protocol, “The categorization of age as a continuous variable should also be considered in the analysis, as it could lead to residual confounding.”

Sponsor’s Response (6/19/23): Age will be considered as continuous variable for overall VE calculations. However, the categorization of age will be evaluated for stratified analysis as described in secondary outcomes of the protocol.

Comment (IR61 7/7/23): We agree with your response.

- Issue is resolved.

5.1.10 Comment #10 (IR50 6/8/23): Please perform a matched analysis or another weighting method to improve the comparability between cases and controls and explain the methodology that you will use.

Sponsor’s Response (6/19/23): A propensity score matching will be applied in order to perform additional matched analysis, and detailed explanation of such methodology will be stated in the Statistical Analysis Plan (Austin 2011; Austin 2011a; Bergstra et al., 2019; Rosenbaum et al., 1983; Parsons, undated).

Comment (IR61 7/7/23): Despite power concerns, we suggest that the matched analysis (or similar design), including by onset time and geographic region, become the primary analysis to improve comparability between cases and controls. Please provide additional details in the SAP on how the propensity score matching will be implemented.

Sponsor’s Response (7/17/23): Please consider our answer to question 9a. We plan to deliver the SAP by 30-Sep-2023. The SAP will include details on these topics. Note: the SAP submission date was later changed to November 30, 2023.

- This issue is resolved.

5.1.11 Comment #11 (IR50 6/8/23): You define partially vaccinated individuals as those who received the vaccine up to 13 days before symptom onset (page 19). Please explain how you plan to analyze these subjects.

Sponsor’s Response (6/19/23): A sensitivity analysis will be performed to estimate the vaccine effectiveness of “any vaccination”, for which people will be considered vaccinated if they received the vaccine any time before symptom onset. Thus, partially vaccinated will

be pooled together with fully vaccinated and compared with the non-vaccinated persons. We would expect this analysis to yield a slightly lower VE estimate than the primary analysis.

Comment (IR61 7/7/23): We agree.

- Issue is resolved.

5.1.12 Comment #12 (IR50 6/8/23): We acknowledge that you plan to exclude subjects with previous CHIKV infection. However, it has been reported that seroprevalence rates were 20% and 51% in two studies in Brazil with the corresponding asymptomatic (or inapparent) CHIKV infection rates of 46% and 63% among seropositive subjects, respectively (PLoS Negl Trop Dis 16(1): e0010069). Since the proposed observational study is not randomized, and there is not a way to ensure participants with pre-existing immunity to CHIKV are equally distributed between vaccine and control groups leading to a potential for biased study results. Please perform serological testing of participants to assess whether they had prior CHIKV infection or similar cross-reactive alphaviruses. Please exclude individuals with prior CHIKV infection from the main analysis. If you believe it is infeasible to exclude participants with prior CHIKV infection (including asymptomatic infection), please explain how you will address the potential biases caused by pre-existing immunity to CHIKV. [Comment written by Dr. Izurieta]

Sponsor's Response (6/19/23): It will be infeasible to test all these subjects. Asymptomatic (or inapparent) CHIKV infection rates were estimated as ranging from 3.8% to 27.7% of cases (Thiberville et al., 2013), about 15% (Burt et al., 2017), 23.9% of cases in the municipality Quixadá Ceará (Braga et al., 2021), 58.8% to 67.3% of cases in Bahia state (Dias et al., 2018; note that while the asymptomatic rate in this study is very high in comparison to other sources, the chronic manifestation rate is also higher in comparison to other studies, suggesting a recall bias among those who made a full recovery, since the sero-survey was done a year after disease activity) of cases, and for participants who did not report to have symptoms, 43 (14.3%) had reactive antibodies against the virus, and had been classified as asymptomatic or oligosymptomatic cases (Barreto et al., 2020). Symptomatic prior infection will be excluded by self-report. Asymptomatic prior infection will be not identified, however, considering that CHIKV infection can be contracted only once, and the infection ensures lifelong immunity against re-infection (e.g., de Souza et al 2023), we expect that it is more likely to find these participants in the control group, then the VE could be underestimated.

If feasible, sero-surveys (adopting CHIKV RT-PCR assay) will be considered in 1-2 selected study municipalities prior to vaccination roll-out to better understand previous CHIKV exposure in those sites, and with that we could potentially infer the proportion of CHIKV experienced individuals that would take part in the study.

Comment (IR61 7/7/23): For the groups to be comparable, the likelihood of prior infection has to be similar between the vaccinated and unvaccinated populations. Because participants with pre-existing immunity to CHIKV may be unequally distributed between vaccinated and unvaccinated groups, this could lead to potentially biased study results, as stated above. Therefore, to help determine whether vaccinees are as likely to have prior CHIKV infection as the population to which they belong, we strongly advise that you consider performing sero-surveys in the study municipalities/administrative regions in

conjunction with the vaccination roll-out, including among vaccinees and those who do not get vaccinated, to determine rates of prior CHIKV exposure in both groups by administrative region.

Sponsor's Response (7/17/23): The Sponsor added a sero-survey to the (b) (4) -402 protocol, as described on page 13.

Comment #1d (IR89, 10/13/2023): Sero-survey Study: On page 74 of the protocol, you provide a sero-survey outline that states: "A separate study protocol will be developed to investigate CHIKV pre-exposure at the time of the pilot vaccination program roll-out." Please provide a timeline for submission of this sero-survey study protocol.

Sponsor's Response (10/18/2023): *A sero-survey design concept will be submitted to the agency by December 31, 2023; a draft protocol is planned to be available for the agency's review by 30-Jun-2024. Please also refer to the timetable (section Sero-survey for identification ChikV prevalence) provided in Company Response to IR#89 CBER Comment 1e.*

- This issue is resolved.

5.1.13 Comment #13 (IR50 6/8/23): Also, because of the possibility of still including some individuals who had prior CHIKV infection despite serological testing, please consider performing a sensitivity analysis stratifying by CHIKV seroprevalence in the population or including an appropriate interaction term to address this concern.

Sponsor's Response (6/19/23): *We acknowledge the comment and will set it up accordingly in an updated (b) (4) -402 study protocol version and SAP.*

Comment (IR61 7/7/23): Please provide the timeline for submission of the SAP. Please provide details regarding proposed sensitivity analysis, including specifics of how you plan to account for differences in baseline serostatus for vaccinees and the unvaccinated in each administrative region included in the study.

Sponsor's Response (7/17/23): *We plan to deliver the SAP by the 30th of September 2023.*

In order to quantify the potential bias resulted from unmeasured confounding due to prior CHIKV infection, a probabilistic bias analysis will be used. For this analysis, the VE estimates will be first transformed to OR ($VE = 1 - OR$) and then transformed again to VE estimates after applying the bias correction. We will quantify the order and the direction of the presumed level of bias due to unmeasured prior CHIKV infection, by applying a bias factor to the unadjusted estimate of association.

More details about the probability distributions and the other assumptions used to generate the MC simulations will be provided in the SAP.

Note: the sponsor changed the submission of the SAP to November 30, 2023.

- As described earlier, the RWE team performed a QBA to determine bias produced by prior infection rates that can impact the VE estimates. This issue is resolved.

5.1.14 Comment #16 (IR50 6/8/23): Please submit your Data Management Plan, including information on:

- a. Definitions and validation of study variables, including those captured in the case/control questionnaire and integration of data from the various sources (e.g., various clinics, laboratories) into the study database.
- b. Maintaining the provenance (intactness) and quality (reliability) of the data from collection to input into your study database and data analyses.
- c. How data are accurately matched to each study subject.
- d. How the data will be audited to ensure sufficient accuracy, consistency, and completeness and the mitigation strategies used to reduce error.

Sponsor's Response (6/19/23): We acknowledge the comments for the Data Management Plan (DMP) which will be set up accordingly in due course. The DMP will be submitted to allow incorporation of the Agency's comments prior to start of study (b) (4) -402.

Comment (IR61 7/7/23): The data and how they are managed are critical for establishing RWE for regulatory decision making. Without the DMP and the SAP, there are many open questions about data handling, variables, and analyses. Please provide an expected timeframe for submission of these key documents and note that transparency of your research processes is required for FDA to evaluate the quality of your methods and the applicability of the RWE generated. We strongly suggest you provide a flowchart of the study process including study participant inclusion and exclusion criteria, data sources and methods, testing processes, etc. Since the study will be conducted in a foreign country, please refer to the Agency guidance titled, "FDA Acceptance of Foreign Clinical Studies Not Conducted Under an IND" (<https://www.fda.gov/media/83209/download>), which states that "FDA may need to review source documents such as hospital records to verify data, whether during an on-site inspection or upon request. ... A review division within FDA may request submission of investigator, hospital, or institutional records outside of an inspectional context. If so, these records must be made available to the Agency for FDA to rely on the data."

Additionally, concerning the use of analyses and data collected from non-interventional studies, please refer to the Agency's Real-world evidence guidance documents (Real-World Evidence | FDA <https://www.fda.gov/science-research/science-and-research-special-topics/real-world-evidence>). In order to consider RWD as supportive evidence for regulatory decision making, "...sponsors must ensure that they are able to submit patient-level data for any RWD that have been analyzed as part of the clinical study included in a marketing application..." (FDA Draft Guidance: Considerations for the use of real-world data and real-world evidence to support regulatory decision-making for Drug and Biological products <https://www.fda.gov/media/154714/download>). If data are owned by third parties, sponsors should have agreements in place to ensure that all data may be provided to FDA for inspection and analysis.

Sponsor's Response (7/17/23): We agree.

Reviewer Comment #1 (IR89 10/13/2023): We again asked for the dates of the DMP and SAP submissions.

Sponsor's' Response: Valneva provided the following dates: November 30, 2023 for the SAP and December 30, 2024 for the DMP and other important milestones (see section 4.3).

Refer to the Review Comment IR #95 (10/26/23), on page 28 of this memo, regarding database and data element evaluation was communicated to the Sponsor.

Reviewer's Comments: Valneva provided an updated timeline for data access and evaluation for the 402 study.

- This issue has been addressed; refer to the Sponsor's Response (10/31/23) on page 28 of this memo for the updated timelines for data access and evaluation.

5.2 STUDY PROTOCOL (b) (4) -402 (V4.0/4.1)

Below are additional RWE comments for the updated versions to the original protocol:

5.2.1 Comment # 1 (IR85 9/22/23): We reviewed your updated participant interview/structured questionnaire which now includes the participants' medical history as well as specifics about their CHIK-like symptoms and knowledge of the illness. We have the following questions:

- You state that this questionnaire is the primary source of your demographic and medical data for the participants in this study, yet on page 45 you state "All questions are on a voluntary basis, if you do not wish to answer a question you have the right not to do so." Please explain how you will manage incomplete demographic and medical data that is required for this study.
- As none of the medical terms are defined, including neurological and other complications, please explain who will be completing these questionnaires and how responses will be standardized.
- We note in the sections for Medical History, Co-morbidities/Risk Factors and Complications, there are > 40 questions that include "do not know" as an answer. Please explain how you will address "do not know" answers and how they might impact your analyses, especially since many are pertinent clinical questions.

Sponsor's Response (9/28/23): 1a. In the updated (b) (4) -402 study protocol version 4.1, pages 10 and 20 (protocol section 4.2) participants are excluded if one or more of the following data items are incomplete: sex, age, municipality of residence, vaccination status, testing status date, and date of symptom onset. Thus, missing critical demographic data must be available for a participant to be included in the analysis.

As shared over our tc on 09/26, comorbidity variables (in Medical History (section C) of participant interview questionnaire) will be used in the multivariable regression analyses for estimating vaccine effectiveness, by first coding the categorical variable 'Number of comorbidities' (values: 0, 1, 2, ≥3), and including this variable as a confounder. This is interpreted as 'Number of [known] comorbidities', because only 'Yes' responses for each comorbidity are considered when constructing this categorical variable. To assess the impact of 'Don't know' or missing responses to questions in section C, vaccine effectiveness estimation will be repeated using missing data imputation methods, namely multiple imputation using chained equations (MICE) to impute 'Don't know' or missing responses. This approach has been introduced to the protocol (page 24, at the end of section 4.9.1) and will be described in more detail in the Statistical Analysis Plan (SAP). -Please note that for clarity the term "comorbidities" was replaced by "chronic conditions" throughout the protocol.

1b. As shared over our tc on 09/26, medically educated study team members will complete the

questionnaire. Training material which will provide detailed guidance on completion of the questionnaire and standardization of medical terms, e.g. list of events qualifying for neurological complications, will be developed and training sessions will be performed before start of study (b) (4) -402.

1c. Please see response to 1a above

- The RWE review team agreed to the response; issue resolved.

5.2.2 Comment # 2 (IR85 9/22/23): On page 11 of the (b) (4) -402 protocol, you indicate an exclusionary criterion is inclusion in the sero-survey database, yet on page 73, you state that participation in the sero-survey has no impact on the eligibility to participate in the 402 study. We agree that participation in the sero-survey is independent from enrollment in the 402 study if they are performed in the same geographic areas. Please update your protocol accordingly.

Sponsor's Response (9/28/23):

Thank you for pointing out this inconsistency. The respective exclusion criterion in the protocol has been updated to read as follows: Individuals with a history of CHIKV infection before vaccination with (b) (4) self-reported or registered in the GAL or SINAN databases.

As discussed over our t/c on 09/26, this approach means that only individuals known to be seropositive through routine medical practice will be excluded, more closely reflecting the real-world situation. In addition, we followed your recommendation by including a respective sensitivity analysis excluding also those individuals whose previous CHIKV illness was identified only during the sero-survey, addressed in the protocol on page 25 (protocol sections 4.9.2.3 and 4.9.2.4).

- The RWE review team agreed to the response; issue is resolved.

5.2.3 Comment # 3 (IR85 9/22/23): Please confirm that Informed Consent will be obtained from both vaccinees and non-vaccinees in your studies.

Sponsor's Response (9/28/23):

This is to confirm that an Informed Consent will be obtained from vaccinees as well as non-vaccinees. This has been made clearer in the latest revision of the protocol (see page 31, section 4.14., subsection).

- The RWE review team agreed to the response; issue is resolved.

5.2.4 Comment #4 (IR85 9/22/23): Please update your case definition for CHIK-like illness on page 44 by removing the following subjective descriptors which can potentially introduce bias by excluding possible cases:

A CHIKV-like case is defined as an individual with at least one of the following symptoms ~~not explained by other conditions~~:

- Sudden-onset of fever (>38.5°C)
- Intense (poly)arthralgia/arthritis of acute onset

Sponsor's Response (9/28/23):

As discussed over our t/c on 09/26, the definition for CHIKV-like illness in the (b) (4) -402 protocol has been based on the definition referred to by the Brazilian Ministry of Health

(MoH) and guides physicians'/ health care centers' decision making on individuals qualifying for chikungunya testing as part of Brazil's routine testing strategy. As such, we are not able to modify this definition as in this real-world evidence study, we will not be able to influence the practice on who will be sent for testing for chikungunya. As suggested during the call, wording has been added to the protocol (Annex 8.3) to clarify that the definition is based on the definition applied by the Brazilian MoH.

- This issue is deferred to the clinical team.

5.3 STUDY PROTOCOL (b) (4) -404

The RWE review team evaluated this protocol for the effectiveness and digital health technology (DHT) elements of the study. Refer to the clinical review team regarding the safety aspects and RCT components of the study design. The RWE team had the following comments:

5.3.1 Reviewer Comment #2f (IR93 10/20/23): As you will be collecting Real World Data from health facilities, laboratories, and the participants, please submit a Data Management Plan that includes all of the data elements, including definition and validation of study variables, maintaining provenance and quality of the study data, any data collection tools you will utilize, and your evaluation that the data are fit for your purpose. Please refer to the FDA Guidance for Industry Considerations for the Use of Real-World Data and Real World Evidence To Support Regulatory Decision-Making for Drug and Biological Products and Submitting Documents Using Real-World Data and Real-World Evidence to FDA for Drug and Biological Products.

Sponsor's Response 10/25/23: As indicated as part of Valneva's response to IR#91, the final DMP will be submitted as part of the study implementation readiness verification submission: Study implementation readiness verification submission (including agreements with required partners, IRB & local regulatory approvals as necessary, serological assay validation documentation, enrolment targets, final SAP, final DMP): 30-Jun-2025. Additional timelines: Final Protocol Submission (agreed between FDA and Valneva): 30-Sep-2024; Study initiation (defined as first subject enrolled): 01-Oct-2025; Study completion (last case collected + 12 weeks follow-up): 31-Jul-2029; and Final study report submission: 31-Dec-2029.

- These timelines were requested as part of the Sponsor's postmarket requirements (PMR). This issue has been addressed.

The RWE review team noted that an eDiary will be used for participants to track any health events on a mobile app. A DHT reviewer [Dr. Hussein Ezzeldin, OBPV/DABRA] evaluated the study proposal for data collection and Valneva was informed to consult the FDA guidance, [Digital Health Technologies for Remote Data Acquisition in Clinical Investigations](#) regarding the eDiary design and data collection methodology. More detailed information regarding DHT was provided to the Sponsor during the IND phase.

5.3.2 Reviewer Comment #3e (IR93 10/20/23): In Section 3.7, you state that participants will use a mobile app on their personal devices or will receive phone calls to record AESIs and SAEs. As recording safety data through the mobile app is considered an eDiary, please refer to FDA guidance, Digital Health Technologies for Remote Data Acquisition in Clinical Investigations, regarding your eDiary design and data collection methodology.

Sponsors Response (10/25/23): Acknowledged.

- The eDiary will be fully evaluated in the forthcoming study protocol.

6 REVIEWERS' RECOMMENDATIONS

This product was accepted under FDA's Accelerated Approval Program and therefore confirmatory trials for the IXCHIQ vaccine will be performed according to protocols reviewed and concurred with by CBER. It is expected that Valneva will work with due diligence to ensure the confirmatory trials meet FDA requirements and are performed according to the agreed upon protocols and timelines provided. We therefore recommend accelerated approval of the IXCHIQ Chikungunya Virus vaccine as the results of the confirmatory trials will be reviewed by FDA for traditional BLA approval in the future if it is shown to provide the anticipated clinical benefit.

7 REFERENCES

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3. Rocha RDC, Cardoso ADS, Souza JL, Pereira EDS, Amorim MF, Souza MSM, et al. First official record of *Aedes (Stegomyia) albopictus* (Diptera: Culicidae) in the Acre State, Northern Brazil. *Rev Inst Med Trop Sao Paulo.* 2023;65:e20.
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