valneva

Clinical Trial Protocol

DRUG SUBSTANCE(S)	VLA1553
VERSION NO.	final 4.0
TRIAL CODE	VLA1553-221
DATE	01-February-2024

A RANDOMIZED, OBSERVER-BLINDED, DOSE RESPONSE PHASE 2 TRIAL TO ASSESS THE SAFETY AND IMMUNOGENICITY OF TWO DIFFERENT DOSE LEVELS OF A LIVE ATTENUATED CHIKUNGUNYA VIRUS VACCINE (VLA1553) IN HEALTHY CHILDREN AGED 1 TO 11 YEARS

PROTOCOL NUMBER:	**VLA1553-221**
IND NUMBER:	**17854**
BLA NUMBER:	**BL 125777**

Sponsor: **Valneva Austria GmbH**

[redacted]
Campus Vienna Biocenter 3
A-1030 Vienna, Austria

1. PROTOCOL SIGNATURE PAGE

Title of Clinical Trial:	**A RANDOMIZED, OBSERVER-BLINDED, DOSE RESPONSE PHASE 2 TRIAL TO ASSESS THE SAFETY AND IMMUNOGENICITY OF TWO DIFFERENT DOSE LEVELS OF A LIVE ATTENUATED CHIKUNGUNYA VIRUS VACCINE (VLA1553) IN HEALTHY CHILDREN AGED 1 TO 11 YEARS**
Protocol Number:	**VLA1553-221**
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With their signature, Investigators and Sponsor agree to conduct this trial in accordance with the Protocol, International Council on Harmonisation (ICH) and Good Clinical Practice (GCP) guidelines and with the applicable local regulatory requirements. Moreover, the site will keep all information obtained from the participation in this trial confidential unless otherwise agreed in writing.

Principal Investigator

Print Name	Signature	Date

[redacted]

Signature Date

[redacted]

Signature Date

[redacted]

Signature Date

2. TRIAL PERSONNEL

Head Clinical Operations Chikungunya	Sponsor's Chief Medical Officer
[redacted]	[redacted]

Director Clinical Strategy and Project Lead Chikungunya Vaccine
[redacted]

Trial Organization

The contact details of the organization/individuals involved in the trial (e.g., investigator(s), Sponsor's representative(s), laboratories, oversight committees [including institutional review boards (IRBs), as applicable] will be maintained by the Sponsor and provided to the Investigator.

3. TRIAL CHANGES IN RESPONSE TO COVID-19 PANDEMIC

In May 2023, the end to the global Public Health Emergency of International Concern (PHEIC) for COVID-19 was declared (WHO/IHR, 2023). The trial Sponsor will continuously monitor and evaluate the development of the COVID-19 progression in the area of trial sites to determine if any measures need to be implemented to mitigate undue risks to the participants or in response to local governmental recommendations. Any measure would be communicated to the relevant CA and IRB. For information on COVID-19 management refer to **Section 10.6.**

4. SERIOUS ADVERSE EVENT REPORTING

The Investigator will comply with applicable laws/requirements for reporting serious adverse events (SAEs) to the IRB. For information on the definition and assessment of adverse events (AEs), refer to **Section 17.1**.

All SAEs should be reported on the SAE Report Form to PrimeVigilance by eFax or email <u>within 24 hours</u> after the Investigator has become aware of the event.

eFax:	[redacted]
Email:	

5. ADVERSE EVENTS OF SPECIAL INTEREST REPORTING

The Investigator will comply with requirements set forth in this protocol for reporting adverse events of special interest (AESI). For information on the definition and assessment of adverse events of special interest (AESI), refer to **Section 17.1**.

All AESI should be reported on the AESI Report Form to PrimeVigilance by eFax or email within 24 hours after the Investigator has become aware of the cluster of events.

eFax:	[redacted]
Email:	

6. PREGNANCY REPORTING

The Investigator will comply with applicable laws/requirements for reporting pregnancies to the IRB. For information on the definition and assessment of pregnancies, refer to **Section 17.12**.

All Pregnancies* should be reported on the Pregnancy Report Form to PrimeVigilance by eFax or email within 24 hours after the Investigator has become aware of the event.

eFax:	[redacted]
Email:	

*A pregnancy is not considered an SAE. If a seriousness criterion applies in addition to the pregnancy (e.g. hospitalization, congenital anomaly/birth defect) the pregnancy qualifies as an SAE. In such case a Pregnancy Report Form **and** a SAE Report Form have to be filled out.

7. CLINICAL TRIAL SYNOPSIS

TRIAL TITLE	
A randomized, observer-blinded, dose response phase 2 trial to assess the safety and immunogenicity of two different dose levels of a live-attenuated chikungunya virus vaccine (VLA1553) in healthy children aged 1 to 11 years	
Short Title	A dose response phase 2 clinical trial of VLA1553 in healthy children aged 1 to 11 years
Trial No.	VLA1553-221
Trial Phase	Phase 2
Blinding Scheme	Observer-blinded
Trial Sites/ Countries	This multicenter trial will be conducted in chikungunya endemic countries in Latin America. Trial sites with and without recent local transmission of chikungunya virus (CHIKV) may be included.
CLINICAL CONDITION(S)/ INDICATION(S)	
Active immunization for the prevention of disease caused by CHIKV	
SCIENTIFIC BACKGROUND	

CHIKV is a small spherical RNA virus and a member of the Alphavirus genus in the family *Togaviridae*. The arthropod-borne virus is closely related to other viruses in Africa, South America and Australia that cause similar symptoms such as O'nyong-nyong-virus, Mayaro-virus, or Ross River Virus, respectively. The virus is vectored by the daytime-biting *Aedes aegypti* mosquito, which is also transmitting yellow fever, Zika and dengue viruses. CHIKV can also be transmitted by *Aedes albopictus* mosquitoes, a more cold-tolerant mosquito, which consequently could result in the spread of chikungunya to more temperate areas of the world.

CHIKV has been reported in over 100 countries with more than three million suspected cases alone in the Americas reported to the Pan American Health Organization (PAHO) from affected areas. CHIKV epidemics are explosive and rapidly moving, but not predictable.

An infection with CHIKV may result in chronic and incapacitating arthralgia affecting all gender and age groups accompanied by an acute febrile disease with headache, muscle pain, and skin rashes. Individuals who are at higher risk of more serious complications include infants, the elderly and individuals with underlying chronic medical conditions. Although children over two years of age have milder disease symptoms and higher rates of asymptomatic infection than adults, severe manifestations such as skin eruptions, meningoencephalitis and septic shock can occur.

VLA1553 is a live-attenuated chikungunya virus (CHIKV) vaccine designed for active immunization for the prevention of the disease caused by CHIKV. The vaccine is intended to prevent CHIKV infections in the general population living in endemic regions, as well as to serve as a prophylactic measure for travelers to epidemic areas or areas at risk for an upcoming outbreak. The VLA1553 vaccine comprises a large genetic deletion in the gene encoding the non-structural replicase protein nsP3. The deletion in nsP3 leads to attenuation of the virus *in vivo.*

Currently, neither specific antiviral treatment nor another vaccine than VLA1553 (INN: IXCHIQ ®) is available to prevent CHIKV infection. Prevention against CHIKV infection is otherwise limited to non-treatment interventions (e.g., restrict exposure to vector mosquitos/mosquito repellents).

RATIONALE FOR PERFORMING THE TRIAL	
Valneva is developing VLA1553 as a single dose vaccine for prophylaxis of the infection caused by CHIKV. Two Phase 3 trials with VLA1553 in healthy adults have been successfully completed. The first Phase 3 clinical trial investigating VLA1553 in an adolescent population was initiated in January 2022 and recruitment of 754 participants (randomized 2:1 to receive VLA1553 or placebo) was completed in February 2023. Initial safety data generated in trial VLA1553-321, Valneva's first clinical trial in an endemic area and with individuals previously infected with CHIKV, show that VLA1553 was generally safe and well tolerated in adolescents aged 12 to <18 years, regardless of the CHIKV antibody baseline serostatus. Data from ongoing DSMB reviews indicate that the vaccine is generally safe and well-tolerated in the adolescent population. The rationale for performing the present trial (VLA1553-221) is to evaluate the vaccine VLA1553 in the generally healthy pediatric population and to assess the safety and immunogenicity of a single dose of VLA1553 in line with an EMA and US FDA agreed pediatric development plan. In this Phase 2 trial (VLA1553-221), two different VLA1553 dose levels will be evaluated for tolerability, safety and immunogenicity in at least 300 healthy children (up to 10% baseline seropositive and at least 90% baseline seronegative for CHIKV antibodies) aged 1 to 11 years. This safety and immunogenicity trial will contribute to the safety database and will allow identification of the appropriate dose level in this pediatric population for testing in Phase 3. The trial will be carried out in CHIKV endemic countries in Latin America.	
PLANNED TRIAL PERIOD	
Initiation	Q4 2023
Duration	The overall trial duration (First Participant In – Last Participant Out) is estimated to be approximately 17 months. Individual participation is approximately 13 months from enrollment to trial completion, unless prematurely discontinued.
Completion	Part A (Visit 5, Day 29): Q3 2024 Part B (Visit 7, Month 6): Q1 2025 Part C (Visit 8, Month 12): Q3 2025 Part A (Day 29) analysis will be performed once the last participant has completed Day 29 (Visit 5). Part B (Month 6) analysis will be performed once the last participant has completed Month 6 (Visit 7). Part C (Month 12) analysis will be performed once the last participant has completed Month 12 (Visit 8).
TRIAL OBJECTIVES & PURPOSE	
Trial Purpose	To obtain tolerability, safety and immunogenicity data of two different dose levels of a live-attenuated CHIKV vaccine VLA1553 in healthy children aged 1 to 11 years. The outcome shall provide the basis for dose selection for further development of the vaccine in this age group.
Primary Objective	➢ To evaluate the tolerability of the full dose and half dose formulation of the live-attenuated CHIKV vaccine (VLA1553) following vaccination in healthy children aged 1 to 11 years after a single immunization.

Secondary Objective	➢ To assess the immunogenicity of the full dose and half dose formulation of VLA1553 in healthy children aged 1 to 11 years after a single immunization ➢ To assess the safety of the full dose and half dose formulation of VLA1553 in healthy children aged 1 to 11 years after a single immunization ➢ To identify the optimal dose level(s) of VLA1553 in healthy children aged 1 to 11 years
Exploratory Objective(s)	➢ To evaluate the efficacy of VLA1553 in pediatric participants in CHIKV endemic areas after a single immunization.

TRIAL DESIGN

This is a multicenter, prospective, randomized, observer-blinded, three arm, phase 2 clinical trial evaluating the full dose formulation (target 4.0 log 10 $TCID_{50}$ per 0.5mL) of VLA1553, half dose formulation (3.7 log 10 $TCID_{50}$ per 0.5mL) of VLA1553 and control (Nimenrix, a tetravalent meningococcal vaccine - Men ACWY).

1. At least 300 male and female healthy children aged 1 to 11 years will be enrolled and the overall distribution of participants will be 2:2:1 to the two VLA1553 dose groups (n=120 each) or control (Nimenrix) (n=60).
2. These volunteers will be screened for evidence of previous CHIKV exposure. Enrolment will then be stratified by CHIKV serostatus at baseline, targeting:
   - up to 10 % CHIKV seropositive participants (i.e., IgM+/IgG+ or IgM-/IgG+)
   - at least 90% CHIKV seronegative participants (i.e., IgM-/IgG-)

Participants who are IgM+/IgG- do not qualify for participation in this trial. Note: CHIKV IgM+/IgG- is suggestive for an early phase acute CHIKV infection. Therefore, as safety precaution, no vaccination with the attenuated CHIKV vaccine VLA1553 should be applied.

The clinical trial design is displayed in the

Figure 1 below.

Figure 1: Phase 2 Clinical Trial Design

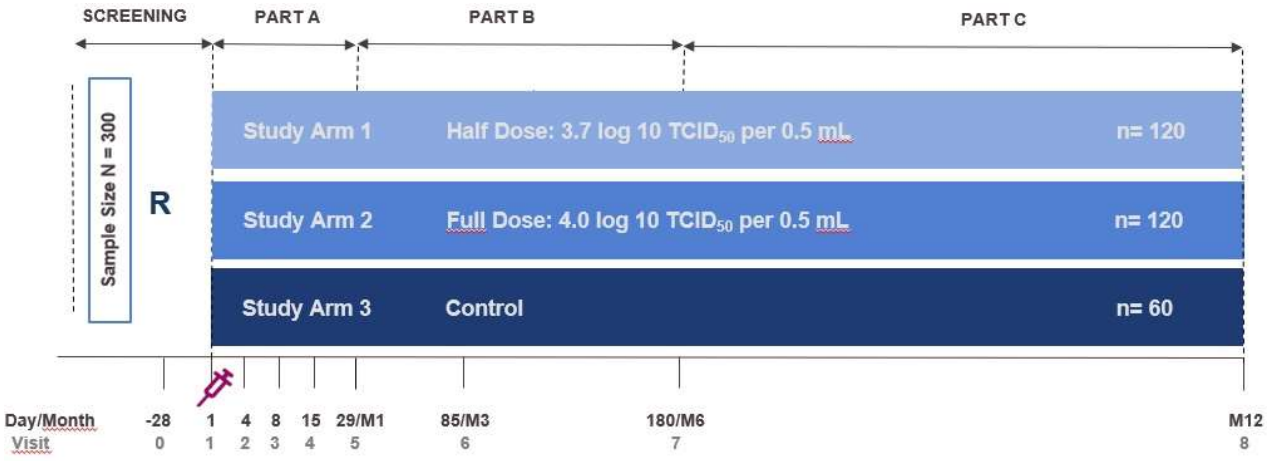
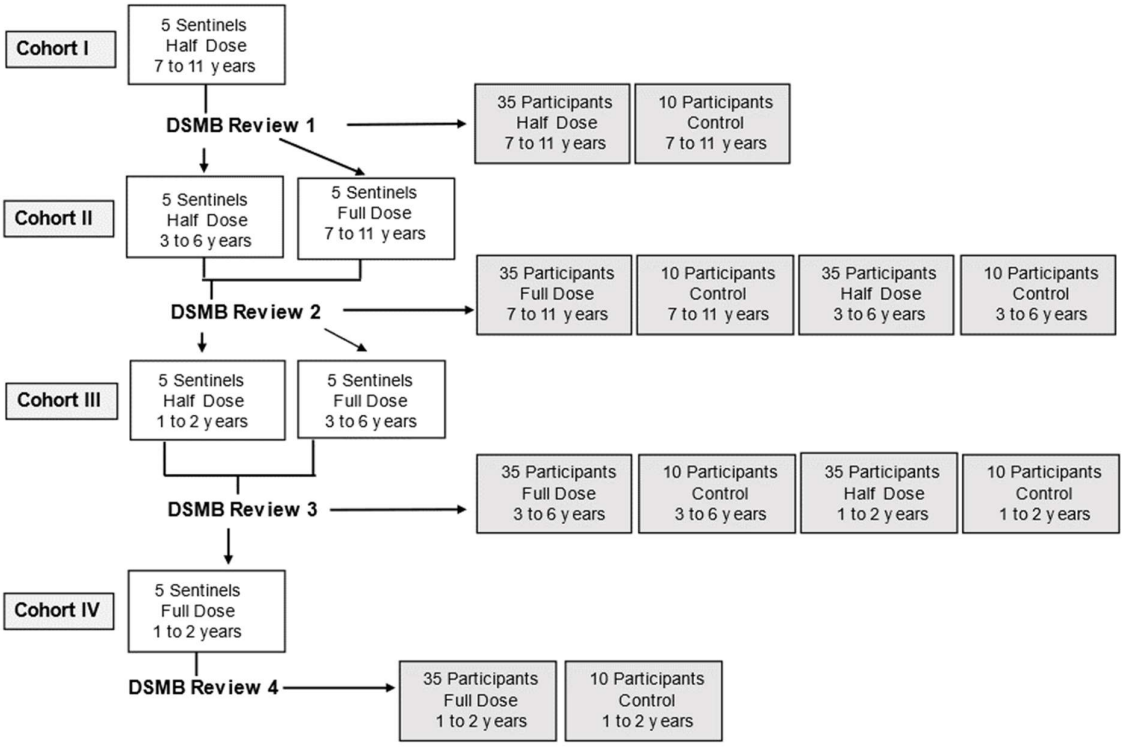


Figure 2 below describes the staggered enrolment approach and **Table 1** below illustrates the participant distribution.

Figure 2: Staggered enrolment



White box: open-label, Grey box: blinded, randomized

Table 1: Participant Distribution

Trial Arm	Dosage ($TCID_{50}$/dose)	Stratum	Cohort I	Cohort II	Cohort III	Cohort IV	Total N
1	VLA1553 half dose 3.7 log 10	A (7 to 11 yrs.)	5*+35	-		-	40
		B (3 to 6 yrs.)	-	5*+35	-		40
		C (1 to 2 yrs.)	-	-	5*+35	-	40
2	VLA1553 full dose 4.0 log 10	A (7 to 11 yrs.)	-	5*+35		-	40
		B (3 to 6 yrs.)	-	-	5*+35		40
		C (1 to 2 yrs.)	-	-	-	5*+35	40
3	Control						

A (7 to 11 yrs.)	10	10		-	20
B (3 to 6 yrs.)	-	10	10		20
C (1 to 2 yrs.)	-	-	10	10	20
Total	**50**	**100**	**100**	**50**	**300**

*Sentinel enrolment will be conducted in open-label fashion and sentinels recruited preferably at a single trial site. Sentinel vaccinations in the following cohort can be started based on the previous cohort sentinels' review by the Data Safety Monitoring Board (DSMB), DSMB's recommendation to the Sponsor and final Sponsor confirmation to proceed with the trial.

Staggered enrolment approach

As a safety precaution measure, 30 sentinel participants will be enrolled into the trial in an open-label fashion according to an age step down scheme outlined in **Figure 2**.

After sentinel analysis, participants will be enrolled in a blinded, randomized manner into three Trial Arms. Within each treatment arm participants will be stratified into three age strata:

Stratum A: 7 to 11 years -children from their 7th birthday until the day before their 12th birthday.

Stratum B: 3 to 6 years - children from their 3rd birthday until the day before their 7th birthday.

Stratum C: 1 to 2 years - children from their 1st birthday until the day before their 3rd birthday.

Age strata for the VLA1553 treatment arms are targeted to be equal in size, i.e., approximately 40 per age stratum.

The age step down phase of the trial will be preferably initiated at a single site to allow permanent oversight on safety data by one principal Investigator. Participants will be recruited in four cohorts as follows:

Cohort I:

Cohort I will be comprised of 5 sentinels of Stratum A (7 to 11 years) receiving the half dose formulation of VLA1553 in an open-label fashion, in a blinded fashion 35 participants of Stratum A (7 to 11 years) receiving the half dose formulation, 10 participants of Stratum A (7 to 11 years) receiving control.

A maximum of three sentinels of Cohort I are to be vaccinated on the first day with vaccinations evenly distributed during the day (e.g., at least 1 hour between participants). Specifically, one sentinel will be vaccinated, observed at the trial site for 1 hour after vaccination for safety and reactogenicity. If no immediate safety concerns arise as determined by the Investigator, then the next sentinel of this Cohort is to be vaccinated and again observed for one hour etc.

These three sentinels of Cohort I are to return to the site 3 days later (Visit 2) for safety follow-up. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of Cohort I are to be vaccinated and are to return to the site 3 days later (Visit 2) for safety follow-up. All sentinels of Cohort I are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

The available Day 8 safety data of these five sentinels of Cohort I will be reviewed by an independent Data Safety Monitoring Board (DSMB) during **DSMB Review 1.** Based on no reservations, a recommendation will be given to the Sponsor and final Sponsor confirmation will be provided on whether to proceed with the randomization of the remaining 45 participants of Stratum A in Cohort I and to start vaccination in Cohort II.

Cohort II:

Cohort II will consist of 5 sentinels of Stratum B (3 to 6 years) receiving the VLA1553 half dose and 5 sentinels of Stratum A (7 to 11 years) receiving the full dose in an open-label fashion, and in a blinded fashion 35 participants of Stratum B (3 to 6 years) receiving the half dose, 35 participants of Stratum A (7

to 11 years) receiving the full dose, 10 participants of Stratum A (7 to 11 years) and 10 participants of Stratum B (3 to 6 years) receiving control.

Similarly, a maximum of three sentinels per stratum are to be vaccinated open-label evenly distributed during the day. One hour observation time after vaccination for immediate safety and reactogenicity evaluation must be adhered. These three sentinels per stratum of Cohort II are to return to the site 3 days later (Visit 2) for safety follow-up. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of Cohort II are to be vaccinated and are to return to the site 3 days later (Visit 2) for safety follow-up. All sentinels of Cohort II are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

Available Day 8 safety data of Cohort II, as well as cumulative safety data, will be reviewed by the DSMB (as described earlier) during the **DSMB Review 2** and based on no reservations, a recommendation will be given to the Sponsor and final Sponsor confirmation will be provided on whether to proceed with the randomization of the remaining 90 participants in Cohort II and to start vaccination in Cohort III.

Cohort III:

Cohort III will consist of five sentinels of Stratum C (1 to 2 years) receiving the half dose formulation in an open-label fashion, five sentinels of Stratum B (3 to 6 years) receiving the full dose in an open-label fashion and in a blinded fashion 35 participants of Stratum C (1 to 2 years) receiving the half dose, 35 participants of Stratum B (3 to 6 years) receiving the full dose, 10 participants of Stratum B (3 to 6 years) and 10 participants of Stratum C (1 to 2 years) receiving control.

A maximum of three sentinels per stratum are to be vaccinated open-label evenly distributed during the day. One hour observation time after vaccination for immediate safety and reactogenicity evaluation must be adhered. These three sentinels per stratum of Cohort III (five sentinels of Stratum C receiving half dose formulation, and five sentinels of Stratum B receiving full dose) are to return to the site 3 days later (Visit 2) for safety follow-up. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of each age strata of Cohort III are to be vaccinated and are to return to the site 3 days later (Visit 2) for safety follow-up. All sentinels of Cohort III are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

Available Day 8 safety data of Cohort III, as well as cumulative safety data, will be reviewed by the DSMB (as described earlier) during the **DSMB Review 3** and based on no reservations, a recommendation will be given on whether to proceed with the randomization of the remaining 90 participants in Cohort III and to start vaccination in Cohort IV.

Cohort IV:

Cohort IV will consist of five sentinels of Stratum C (1 to 2 years) receiving the full dose in an open-label fashion, and in a blinded fashion 35 participants of Stratum C (1 to 2 years) receiving the full dose and 10 participants of Stratum C (1 to 2 years) receiving control.

A maximum of three sentinels of Stratum C receiving the full dose are to be vaccinated open-label evenly distributed during the day. One hour observation time after vaccination for immediate safety and reactogenicity evaluation must be adhered. These three sentinels of Cohort IV will be followed for safety (Visit 2) three days later. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of Cohort IV are to be vaccinated and are to return to the site 3 days later (Visit 2) for safety follow-up. All sentinels of Cohort IV are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

Available Day 8 safety data of Cohort IV, as well as cumulative safety data, will be reviewed by the DSMB (as described earlier) during the **DSMB Review 4** and based on no reservations, a recommendation will be given to the Sponsor and final Sponsor confirmation will be provided on whether to proceed with the randomization of the remaining 45 participants of Stratum C (1 to 2 years) in Cohort IV.

Randomization	Overall distribution of participants will be 2:2:1 to the VLA1553 half dose formulation, VLA1553 full dose formulation or control group.

TRIAL ENDPOINTS	
Primary Endpoint	Tolerability: ➢ Frequency and severity of solicited injection site and systemic reactions within 14 days post-vaccination.
Secondary Endpoint	Safety: ➢ Frequency and severity of any adverse event (AE) within 28 days post-vaccination; ➢ Frequency and severity of unsolicited AE until Month 6 (Day 180) and Month 12 (Day 365) post-vaccination; ➢ Frequency and severity of any serious adverse event (SAE) until Month 6 (Day 180) and Month 12 (Day 365) post-vaccination; ➢ Frequency and severity of any early onset adverse event of special interest (AESI) starting within 2 to 21 days post-vaccination (i.e. Day 3 – Day 22); ➢ Frequency and severity of any late onset adverse event of special interest (AESI) during the entire trial starting 22 days post-vaccination (i.e. Day 23 – trial end); ➢ Assessment of viremia on Days 1, 4, 8, and 15 and beyond as applicable; Immunogenicity: ➢ Immune response in baseline seronegative participants as measured by CHIKV-specific neutralizing antibody titers on Day 1, Day 15, Day 29, Day 85, Day 180 and Month 12 (Day 365) post-vaccination as determined by µPRNT assay; ➢ Proportion of participants with seroconversion [1] as compared to baseline at Day 15, Day 29, Day 85, Day 180 and Month 12 (Day 365) as determined by µPRNT assay; ➢ Proportion of participants with a seroresponse (defined as $\mu PRNT_{50} \geq 150$ for baseline negative participants) [2] on Day 15, Day 29, Day 85, Day 180 and Month 12 (Day 365) post-vaccination as determined by µPRNT assay; ➢ Fold increase of CHIKV-specific neutralizing antibody titers determined by µPRNT assay at Days 15, 29, 85, 180 and at Month 12 (Day 365) post-vaccination as compared to baseline; ➢ Proportion of participants reaching an at least 4-fold, 8-fold, 16-fold or 64-fold increase in CHIKV-specific neutralizing antibody titers compared to baseline at Day

[1] Seroconversion defined as >4-fold increase of $\mu PRNT_{50}$ compared to baseline $\mu PRNT_{50}$ titer at Day 1
[2] Seroresponse threshold derived from animal passive transfer experiments.

	15, Day 29, Day 85, Day 180 and Month 12 (Day 365) as measured by µPRNT assay; ➢ Antibody titers, seroresponse, seroconversion and fold increases for CHIKV-specific neutralizing antibodies, determined by µPRNT assay at Day 15, Day 29, Day 85, Day 180 and Month 12 (Day 365) post-vaccination stratified by baseline serostatus (based on µPRNT), age stratum and dose.
Exploratory Endpoint(s)	➢ Incidence of CHIKV infections with onset 15 days post-vaccination (i.e. Day 16) as evidenced by viremia by virus specific RT-qPCR, clinical diagnosis and seroconversion by µPRNT for the entire trial period* *measured in Stratum A and in sentinel participants of Stratum B and Stratum C

TRIAL POPULATION

At least 300 healthy children aged 1 to 11 years of either gender who satisfy the inclusion and exclusion criteria listed below will be invited to participate in the trial.

Inclusion Criteria

Participants who meet **ALL** of the following criteria are eligible for this trial:

1) Male or female healthy children aged 7 to 11 years[3] for Stratum A, 3 to 6 years[4] for Stratum B and 1 to 2 years[5] for Stratum C at the time of vaccination;
2) Written informed consent by the participant's parent(s)/Legally Acceptable Representative(s) (LAR(s)), according to local requirements, and written informed assent of the participant, if applicable;
3) Participant is seropositive for previous CHIKV exposure (i.e. IgM+/IgG+ or IgM-/IgG+) or seronegative (i.e. IgM-/IgG-) (see the protocol for details on CHIKV antibody screening and assessment of results);
4) Participant is generally healthy [6] as determined by the Investigator's clinical judgement based on medical history, physical examination and screening laboratory tests;
5) For participants in Stratum C 1 to 2 years:
   Participant was born at full term of pregnancy (≥37 weeks) with a birth weight ≥2.5 kg;
6) If participant is of childbearing potential (after onset of menarche) and if involved in activities that could result in pregnancy:
   a) Participant has a negative pregnancy test (in accordance with local regulations) at screening (Visit 0) and Day 1 (Visit 1), respectively
   b) Participant has practiced an adequate method of contraception during at least the 30 days before screening (Visit 0) and during the screening period
   c) Participant agrees to employ adequate birth control measures for the first three months post-vaccination (i.e. until Day 85, Visit 6). This includes one of the following measures:

[3] From the 7th birthday to the last day before the 12th birthday.
[4] From the 3rd birthday to the last day before the 7th birthday.
[5] From the 1st birthday (12 months) to the last day before the 3rd birthday (< 36 months).
[6] Participants are considered ***generally healthy*** if they have no (1) active disease (acute illness, exacerbation of a chronic condition or a chronic – progressive illness) which requires treatment and/or follow-up, or (2) disease that is identified as an exclusion criterion.

- Hormonal contraceptives (e.g. implants, birth control pills, patches);
- Intrauterine hormone-release systems;
- Barrier type of birth control measure (e.g. condoms, diaphragms, cervical caps);
- Vasectomy in the male sex partner ≥3 months prior to first vaccination;
- Same-sex relationships.

Exclusion Criteria

Participants who meet **ANY** of the following criteria are **NOT** eligible for this trial:

1. Participant who is anti-CHIKV IgM+/IgG- does not qualify for participation in this trial.
2. Participant is taking medication or other treatment for unresolved symptoms attributed to a previous CHIKV infection; or has participated in a clinical trial involving an investigational CHIKV vaccine;
3. Participant has an acute or recent infection (and who is not symptom-free in the week prior to the Screening Visit (Visit 0));
4. Parent(s)/LAR(s) or siblings have a known HIV infection;
5. Participant has received another live virus vaccine within 28 days or inactivated vaccine[7] within 14 days prior to vaccination in this trial or plans to receive a live virus vaccine within 28 days or inactivated vaccine within 14 days after vaccination;
6. Participant has abnormal findings in any required trial investigations (including medical history, physical examination, and clinical laboratory) considered clinically relevant by the Investigator which pose a risk for participation in the trial based on his/her judgement;
7. Participant has an ongoing medical history of or currently has acute or progressive, unstable or uncontrolled clinical conditions (e.g., cardiovascular, respiratory, neurologic, psychiatric, or rheumatologic conditions) that poses a risk for participation in the trial, based on Investigators clinical judgement. Examples include individuals with poorly controlled or unstable disease, ongoing suspected or active inflammation, or poor compliance with pharmacologic treatment, or presence of high-risk comorbidities (e.g., significant cardiopulmonary disease);
8. Participant has a history of immune-mediated or clinically relevant arthritis/arthralgia;
9. Participant has a history of malignancy;
10. Participant has a known or suspected defect of the immune system that can be expected to influence the immune response to the vaccine, such as Participants with congenital or acquired immunodeficiency, including infection with HIV, status post organ transplantation or immuno-suppressive therapy within 4 weeks prior to Visit 1. Immuno-suppressive therapy is defined as administration of chronic (longer than 14 days) prednisone or equivalent ≥0.05 mg/kg/day within 4 weeks prior to trial entry, radiation therapy or immunosuppressive cytotoxic drugs/ monoclonal antibodies in the previous 3 years; topical and inhaled steroids are allowed.
11. Participant has a history of any vaccine related contraindicating event (e.g., anaphylaxis, allergy to components of VLA1553 or the control vaccine, other known contraindications including febrile convulsions);
12. Participant presents with clinical conditions representing severe bleeding disorders and medications interfering with blood clotting;
13. Participant is pregnant (positive pregnancy test (in accordance with local regulations) at screening or Visit 1, respectively), lactating at the time of enrollment or unreliable contraception in female participants after onset of menarche and if involved in activities potentially leading to pregnancy;

[7] Includes mRNA vaccines.

14. Participant received blood-derived products (e.g. plasma) within 180 days prior to vaccination in this trial;
15. Participant has a rash, dermatological condition or tattoos that would, in the opinion of the Investigator, interfere with injection site reaction rating;
16. Participant has participated in another clinical trial involving an investigational medicinal product (IMP) or device within 30 days prior to vaccination or is scheduled to participate in another clinical trial involving an IMP, or device during the course of this trial;
17. Participant has any condition that, in the opinion of the Investigator, may compromise the participants well-being, might interfere with evaluation of trial endpoints, or would limit the participant's ability to complete the trial;
18. Participant/ Participant's parent(s)/LAR(s) is/are a member of the team conducting the trial or in a dependent relationship with one of the trial team members. Dependent relationships include close relatives (i.e., children, partner/spouse, siblings, parent(s)/LAR(s)s) as well as employees of the Investigator or site personnel conducting the trial.
19. Participant has received the control vaccine prior to the trial.

INVESTIGATIONAL PRODUCT

Investigational Product	Live-attenuated chikungunya vaccine (VLA1553) in a lyophilized formulation
Active Ingredient(s)	Live-attenuated CHIKV vaccine strain based on the La Reunion strain (LR-CHIKV clone LR2006-OPY1) of East/ Central/ South African (ECSA) genotype (del5nsP3)
Comparative Drug / Control Condition	Nimenrix (Men ACWY vaccine)
Dosage	VLA1553 Half dose formulation (target 3.7 log 10 $TCID_{50}$ per 0.5 mL) VLA1553 Full dose formulation (target 4.0 log 10 $TCID_{50}$ per 0.5 mL)
Route of Administration	Intramuscular (i.m.) in deltoid muscle for Stratum A and B; anterolateral thigh muscle for Stratum C

TRIAL ASSESSMENTS & PROCEDURES

For a tabular outline of trial procedures and assessments required at each visit, please see the Schedule of Trial Procedures and Assessments (**Table 5**).

Before enrollment and any trial-related procedures are performed, written informed consent of the participant's parent(s)/LAR(s) and participant's informed assent, as applicable, will be obtained.

At Day 1 (Visit 1) VLA1553 or control will be administered intramuscularly (in deltoid muscle for Stratum A &B; anterolateral thigh muscle for Stratum C) as a single dose immunization. Participants will return to trial site at Day 4 (i.e., Visit 2, three days after the first vaccination) and at Day 8 (Visit 3) for post-vaccination adverse event (AE) /adverse event of special interest (AESI) /serious adverse event (SAE) assessment, urinalysis and physical examination. Sampling for viremia assessment will be obtained from all participants of age Stratum A (7 to 11 yrs.) and all sentinel participants from Stratum B (3 to 6 yrs.) and Stratum C (1 to 2 yrs.) at Visit 1, Visit 2 and Visit 3. Furthermore, participants will return at Day 15 (Visit 4), Day 29 (Visit 5), Day 85 (Visit 6), Month 6 (Day 180, Visit 7) and Month 12 (Day 365, Visit 8) for AE /AESI /SAE assessment, physical examination and for collection of samples for immunogenicity assessments. Safety laboratory evaluations will be done at Day 29 (Visit 5) from all participants of age Stratum A (7 to 11 yrs.) and all sentinel participants from Stratum B (3 to 6 yrs.) and Stratum C (1 to 2

yrs.). In addition, urinalysis will be done at every trial visit from Day 1 (Visit 1) until Month 12 (Visit 8) from all participants, if possible. Viremia samples will be collected from all participants of Stratum A and sentinel participants from Stratum B and C only at Visit 1 to 8. Viremia samples from Day 1 - Day 15 (Visit 1- Visit 4) will be analysed. Should the sample on Day 15 (Visit 4) after vaccination test positive for viremia, the collected Day 29 (Visit 5) will be analysed. Other viremia samples will be tested for clinically indicated retrospective analysis.	
Trial Periods	Screening: Visit 0 (up to 28 days) Part A: Visit 1, 2, 3, 4, and 5 (Day 1 to Day 29) Part B: Visit 6 and 7 (Day 85 to Month 6) Part C: Visit 8 (until Month 12)
Delay Criteria for Vaccination	1. Participant has an acute febrile infection within 72 hours prior to vaccination or temperature ≥38.0°C(100.4°F) on the day of vaccination (participant may be rescheduled within the screening visit window until participant has completed 72 hours without fever); 2. Participant has received antipyretics or analgesics within 24 hours prior to the scheduled time of vaccination (participant may be rescheduled within the screening visit window); 3. Participant has received any live or inactivated vaccine [8] within 28 days or 14 days prior to vaccination, respectively (participant may be rescheduled within the screening visit window). 4. IMP is unavailable at the site (e.g., logistics problems) or under quarantine. 5. In case of no blood sample result available (e.g., out of stability samples) due to logistical problems at Screening (V0), and the permission of at least one parent/LAR to recollect the missing sample* In addition, for a rescheduled vaccination all inclusion and none of the exclusion criteria must be met; otherwise, the participant will be excluded from the trial. The rescheduled visit should be within the specified time window for the vaccination visit. In case the time window for the rescheduled visit cannot be met, the participant might be invited for a re-screening. *For recollection of a blood sample the permission of at least one parent/LAR has to be documented in the source documents and is only allowed once (if applicable in selected country).
Holding Rules	Trial specific and individual holding rules are specified in the protocol
Immunogenicity Assessments	Immunogenicity assessment measuring neutralizing antibodies will be performed on samples collected at Day 1, 15, 29, 85 and at Month 6 and 12 after a single dose immunization. Immunogenicity samples will be collected from participants at the indicated time points and may be used for additional testing procedures.

[8] Includes mRNA vaccines.

SAFETY ASSESSMENTS

Data Safety Monitoring Board (DSMB)

An independent Data Safety Monitoring Board (DSMB) will be installed for this trial. The DSMB will meet to review safety data during the staggered enrolment (four meetings) and accumulating safety data on a regular basis throughout the trial, as applicable and a recommendation will be given, whether to proceed with the next Cohort, which will be confirmed by the Sponsor in writing before proceeding.

The DSMB will thoroughly evaluate the following safety parameters:

1. AEs (including solicited and unsolicited), SAEs and AESI;
2. Results of physical examinations;

In addition, the DSMB will periodically review accruing safety information throughout the trial, as applicable. A meeting of the DSMB may be called at the discretion of the Sponsor, e.g., to address any safety concerns arising during the conduct of the trial.

The DSMB will ad-hoc review cases of SAEs and AESI. The DSMB will periodically review listings and summary tabulations of SAEs, deaths, solicited AEs and unsolicited AEs.

A DSMB charter including a detailed description will be prepared.

Safety Questionnaires

Parent(s)/LAR(s) will be provided with the following types of safety questionnaires covering the entire course of this trial:

- Participant Diary will be distributed to all parent(s)/LAR(s) for the collection of safety information from the day of vaccination until the first 14 post-vaccination days (i.e., including the day of vaccination (Day 1 until Day 15);
- Memory Aid will be distributed to all parent(s)/LAR(s) for the collection of safety information outside the first 14 post-vaccination (i.e., Day 16 – Month 12) days until the next Visit.

The following information will be collected in the **Participant Diary** (14 days post-vaccination (starting on day of vaccination), Day 1- Day 15):

- Body temperature (infrared measurements);
- Solicited injection site reactions[9], including severity (pain, tenderness, injection site erythema or redness, and injection site induration or swelling);
- Solicited systemic reactions9 for Stratum A and B (nausea or vomiting, headache, fatigue, myalgia, arthralgia, rash and fever) and Stratum C (nausea or vomiting, inconsolable or excessive crying, disturbed sleep, loss of appetite, drowsiness, rash and fever), including severity;
- Other AEs including severity;
- Any new concomitant medication or changes in medication taken after vaccination.

The following information will be collected in the **Memory Aid** (Day 16 until Month 12):

- AEs including severity;
- Solicited reactions ongoing beyond the participant diary period;
- Any new concomitant medication or changes in medication taken after vaccination.

Reporting of assessments for the **Participant Diary** should start on the day of vaccination, accomplished once daily at approximately the same time each day, starting approximately 8 hours after vaccination for a total of 14 consecutive days (until Day 15). Parent(s)/LAR(s) will be properly instructed on the reporting

[9] Solicited injection site and systemic reactions in the Participant Diary will be assessed by the parent/LAR for absence, presence, severity and duration.

requirements and how to complete and use the safety questionnaires, thermometer and measuring device for assessment of measurable injection site reactions.

Safety questionnaires are always to be returned at the next visit and will be reviewed by the Investigator together with the parent(s)/LAR(s), prior to these data being entered into the electronic case report form (eCRF). The Investigator will review and discuss the safety questionnaires with the parent(s)/LAR(s), ask about AEs occurring since the last visit and both the parent(s)/LAR(s) and Investigator have to sign the questionnaire to ensure completeness and reliability of self-reporting. The safety questionnaires are to be collected by the Investigator at indicated visits and new ones will be distributed.

The entries in the safety questionnaires shall be evaluated by the Investigator; solicited/unsolicited AEs are to be graded for severity (for grading refer to the section below) and relatedness to the vaccination (categories: probably, possibly, unlikely related or unrelated).

Note: Solicited AE should only be documented in the participant diary section of the eCRF. In case of solicited SAEs the data should additionally be documented at the AE eCRF page. Unsolicited AEs and SAEs are always documented at the AE eCRF page.

Solicited Adverse Events (Injection Site Reactions, Systemic Reactions)

Parent(s)/LAR(s) will be provided with an infrared thermometer and measuring device to individually assess solicited injection site and systemic reactions over a period of 14 consecutive days after vaccination (i.e., including the day of vaccination until Day 15).

1. Injection Site Reactions

Parent(s)/LAR(s) will be provided with a measuring device to measure the size of any measurable injection site reaction over a period of 14 consecutive days after vaccination.

Injection site reactions include injection site pain, tenderness, injection site erythema or redness, and injection site induration or swelling (**Table 2**). Any injection site reaction meeting the grade 4 criteria will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

Table 2: Injections Site Reactions 1 – 11 years of age

Vaccine specific Criteria	Grade 1 (Mild)	Grade 2 (Moderate)	Grade 3 (Severe)	Grade 4 [4] (Potentially Life-Threatening)
Injection Site Pain [A]	Does not interfere with activity	Repeated use of non-narcotic pain reliever >24 hours or interferes with activity	Any use of narcotic pain reliever or prevents daily activity	Emergency room (ER) visit or hospitalization
Tenderness [A]	Mild discomfort to touch	Discomfort with movement	Significant discomfort at rest	ER visit or hospitalization
Injection Site Erythema or Redness [B,1]	1.0 [2] - ≤2.5 cm in diameter	>2.5 - 5.0 cm [3] in diameter with <50% surface area of the extremity segment involved (e.g., upper arm or thigh)	>5 cm [3] OR ≥50% surface area of the extremity segment involved (e.g., upper arm or thigh) OR Ulceration OR Secondary infection OR Phlebitis OR Sterile abscess OR Drainage	Potentially life-threatening consequences (e.g. Skin necrosis)
Injection Site Induration/ Swelling [B]	1.0 [2] - ≤ 2.5 cm in diameter	>2.5 - 5.0 cm [3] in diameter with <50% surface area of the extremity segment involved (e.g., upper arm or thigh)	>5 cm [3] OR ≥ 50% surface area of the extremity segment involved (e.g., upper arm or thigh) OR Ulceration OR Secondary infection OR Phlebitis	Potentially life-threatening consequences (e.g. Skin necrosis)

A) Grading based on FDA Guidance on Toxicity Grading Scales Adults/ Adolescents.
B) Grading based on Division of AIDS, Table for Grading the Severity of Adult and Pediatric Adverse Events, corrected version 2.1, July 2017.
1) Injection Site Erythema or Redness should be evaluated and graded using the greatest single diameter or measured surface area.
2) Reactions less than 1cm are not considered AEs.
3) Modified grading based on Division of AIDS; 5.0 cm cut-off will be included as moderate instead of severe.4) Any injection site reaction meeting the grade 4 criterion will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

2. Systemic Reactions

Systemic reactions for **Stratum A and Stratum B** including nausea or vomiting, headache, fatigue, myalgia, arthralgia, rash and fever will be graded and reported in a standardized manner **(Table 3)**. Any systemic reaction meeting the grade 4 criteria according to **Table 3**, will need to be assessed by the investigator if a seriousness criterion is met and may require expedited reporting.

Table 3: Grading of Systemic Reactions Stratum A and Stratum B

Vaccine specific Criteria	Grade 1 mild	Grade 2 moderate	Grade 3 severe	Grade 4 [1] potentially life-threatening
Nausea/ Vomiting [A]	No interference with activity or 1 – 2 episodes/24 hours	Some interference with activity or >2 episodes/24 hours	Prevents daily activity, requires outpatient IV hydration	ER visit or hospitalization for hypotensive shock
Headache [A]	No interference with activity	Repeated use of non-narcotic pain reliever >24 hours or some interference with activity	Significant; any use of narcotic pain reliever or prevents daily activity	ER visit or hospitalization
Fatigue [A]	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalization
Myalgia [A]	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalization
Arthralgia [A]	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalization
Rash [B]	Macules/papules covering <10% body surface area (BSA) with or without symptoms (e.g., pruritus, burning, tightness)	Macules/papules covering 10- 30% BSA with or without symptom (e.g., pruritus, burning, tightness); limiting instrumental activity of daily living	Macules/papules covering >30% BSA with or without associated symptoms; limiting self-care activity of daily living	NA
Fever [C]	38.0 to < 38.6°C or 100.4 to < 101.5°F	≥ 38.6 to < 39.3°C or ≥ 101.5 to < 102.7°F	≥ 39.3 to < 40.0°C or ≥ 102.7 to < 104.0°F	≥ 40.0°C or ≥ 104.0°F

A) Grading based on FDA Guidance on Toxicity Grading Scales Adults/ Adolescents.
B) Grading based on Common Terminology Criteria for Adverse Events (CTCAE), NIH, v4.03, 2010.
C) Grading based on Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events Corrected Version 2.1 July 2017
1) Any grade 4 systemic reaction will need to be assessed by the investigator if a seriousness criterion is met and may require expedited reporting.

Systemic reactions for **Stratum C** including nausea/vomiting, inconsolable or excessive crying, disturbed sleep, loss of appetite, drowsiness and rash will be graded and reported in a standardized manner **(Table 4**). Any systemic reaction meeting the Grade 4 criterion will need to be assessed by the investigator if a seriousness criterion is met and may require expedited reporting.

Table 4: Grading of Systemic Reactions Stratum C

Vaccine specific Criteria	Grade 1 mild	Grade 2 moderate	Grade 3 severe	Grade 4 [3] potentially life-threatening
Nausea/Vomiting [A]	No interference with activity or 1 – 2 episodes/24 hours	Some interference with activity or >2 episodes/24 hours	Prevents daily activity, requires outpatient IV hydration	ER visit or hospitalization for hypotensive shock
Inconsolable or excessive crying [B]	presence of crying [1] which is continuous [2] AND unaltered for ≥3h.	presence of crying [1] which is continuous [2] AND likely to be unaltered for >3h OR unaltered for >3h AND likely to be continuous.	NA	NA
Disturbed sleep [C]	Mild difficulty falling asleep, staying asleep, or waking up early causing no or minimal interference with usual social & functional activities	Moderate difficulty falling asleep, staying asleep, or waking up early causing more than minimal interference with usual social & functional activities	Severe difficulty falling asleep, staying asleep, or waking up early causing inability to perform usual social & functional activities requiring intervention or hospitalization	NA
Loss of appetite [D]	Loss of appetite without alteration in eating habits	Oral intake altered without significant weight loss or malnutrition; oral nutritional supplements indicated	Associated with significant weight loss or malnutrition (e.g., inadequate oral caloric and/or fluid intake); tube feeding or TPN indicated	Life-threatening consequences; urgent intervention indicated
Drowsiness [D]	Mild but more than usual drowsiness or sleepiness	Moderate sedation; limiting instrumental activity of daily living	Obtundation or stupor	Life-threatening consequences; urgent intervention indicated
Rash [E]	Macules/papules covering <10% body surface area (BSA) with or without symptoms (e.g., pruritus, burning, tightness)	Macules/papules covering 10- 30% BSA with or without symptom (e.g., pruritus, burning, tightness); limiting instrumental activity of daily living	Macules/papules covering >30% BSA with or without associated symptoms; limiting self-care activity of daily living	NA
Fever [F]	38.0 to < 38.6°C or 100.4 to < 101.5°F	≥ 38.6 to < 39.3°C or ≥ 101.5 to < 102.7°F	≥ 39.3 to < 40.0°C or ≥ 102.7 to < 104.0°F	40.0°C or ≥ 104.0°F

[A)] Grading based on FDA Guidance on Toxicity Grading Scales Adults/ Adolescents.
[B)] Grading based on Persistent crying in infants and children as an adverse event following immunization: case definition and guidelines for data collection, analysis, and presentation; The Brighton Collaboration Persistent Crying Working Group; Vaccine 2004.

C) Grading based on Division of AIDS, Table for Grading the Severity of Adult and Pediatric Adverse Events, corrected version 2.1, July 2017.
D) Grading according to Common Toxicity Criteria (CTC) for cancer trials, Version 5.0, 2017.
E) Grading based on Common Terminology Criteria for Adverse Events (CTCAE), NIH, v4.03, 2010.
F) Grading based on Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events Corrected Version 2.1 July 2017

1) crying which parent(s)/LAR(s) may describe as "fearful", "angry", "sorrowful", "in pain", "pitiful", "plaintiff", "never heard before in this child".
2) not episodic, not interrupted within the time period of 3h (e.g. by naps).
3) Any Grade 4 systemic reaction will need to be assessed by the investigator if a seriousness criterion is met and may require expedited reporting.

2.1 Body Temperature Measurement

Parent(s)/LAR(s) will be provided with an infrared thermometer to individually measure the body temperature targeting the forehead once every evening from the day of vaccination until Day 14 after vaccination. In case fever (body temperature ≥38.0°C/100.4°F) occurs, the parent/LAR should measure the participant's body temperature every 4 to 8 hours until fever resolves (body temperature <38.0°C/100.4°F). The time at which the first body temperature of <38.0°C/100.4°F is recorded is considered to be the end of the fever episode. All fever measurements should be recorded in the Participant Diary including the first value that shows a return to normal body temperature. In case of more than one recorded body temperature value in the Participant Diary on a given day, the highest daily temperature reading will be recorded in the eCRF. In case of fever ≥39°C/102.1°F the site should be contacted immediately.

Body temperature measurements will be analyzed according to the "Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events Corrected Version 2.1 July 2017" for data analysis as described in **Table 4** above.

Note, in case a fever is not supported by a temperature reading it will be documented as "feverishness" in the eCRF.

Adverse Events of Special Interest (AESI)

In addition to nonspecific transient myalgia and arthralgia that may occur after vaccination with any vaccine, participants will be carefully monitored for symptoms suggesting an acute stage CHIKV-associated event.

If a participant develops symptoms suggestive of an AESI, parent(s)/LAR(s) will be asked to contact the site immediately for clinical evaluation at an unscheduled visit.

Therefore, the following cluster of symptoms with or without remissions or exacerbations will receive particular consideration and are defined as early onset AESI:

1. Fever (≥ 38.0 °c / 100.4 °f, infrared measurement) **AND**
2. symptoms suggesting acute (poly)arthralgia/arthritis, myalgia, neurological symptoms (e.g., for meningoencephalitis, acute encephalitis, headache, seizures), back pain, lymphadenopathy, cardiac or ocular symptoms (e.g., for uveitis and retinitis);

OR

one or more of the following signs and symptoms: macular to maculopapular rash (sometimes with cutaneous pruritus [foot plant]), pigmentary changes, bullous rash/ skin blistering, purpura, ecchymosis and

3. Onset of symptoms 2 to 21 days after vaccination (i.e., day 3 – day 22) **AND**
4. Duration of event ≥ 3 days.

The cluster of symptoms defined above but starting 22 or more days after vaccination (i.e. Day 23 or later) until trial end is defined as late onset AESI and will be documented and followed-up in the same manner as early onset AESI. Notably, the collection of late onset AESI is to be performed only in the absence of another diagnosis.

Events, AEs or (SAEs), which have an underlying (serious) suspected or confirmed diagnosis, stating a disease or a syndrome, where all or a part of the AESI qualifying symptoms (e.g., fever, headache, myalgia, arthralgia) can be, together with additional symptoms, attributed to, do not meet the AESI criteria.

Clinical Symptoms that may be a sign for a clinically significant condition need a medical work up (e.g. AESI laboratory parameter assessment) and a referral to a specialist as medically indicated. Treatment should be performed according to current medical standard of care. Additionally, participants presenting with acute arthralgia post-vaccination will be followed-up until resolution and monitored for recurrences until the end of the trial.

A blood sample for laboratory investigation will be taken upon presentation of the participant. Testing may include but is not limited to rheumatoid factor (RF), anticitrullinated protein antibody (ACPA), Ferritin and CRP. Retrospective investigation of a pre-vaccination sample may be considered for a thorough causality assessment.

Suspected AESI do not constitute a reason for withdrawal from the trial.

Unsolicited Adverse Events (post vaccination)

Parent(s)/LAR(s) will be provided with a Participant Diary up to Visit 4 and a Memory Aid from onward visits to be able to collect unsolicited AEs. Safety questionnaires are always to be returned at the next visit and will be jointly reviewed by the investigator and parent(s)/LAR(s), before these data will be entered into the electronic case report form (eCRF). The Investigator will review and discuss the safety questionnaires with the parent(s)/LAR(s), ask about AE occurring since the last visit and both parent(s)/LAR(s) and investigator have to sign the questionnaire to document completeness and reliability of self-reporting. The safety questionnaires are to be collected by the investigator at indicated visits and new ones will be distributed. Clinically relevant laboratory parameter changes constitute an unsolicited AE. In addition, any symptom noted during the symptom-driven physical examinations constitute an AE.

STATISTICAL ANALYSIS

Sample Size Justification	The total number of 240 children aged 1 to 11 years exposed to VLA1553 in this trial has been selected to provide a sufficient number of participants for proper safety evaluation. With 240 participants exposed, the trial will provide 95% confidence that an AE does not occur at a frequency of 1.24% or higher, if not observed in the trial. In terms of immunogenicity, up to 240 participants exposed to VLA1553 and assuming 10% seropositive participants and among seronegatives an expected rate of 10% of participants to be excluded due to drop-outs/major protocol deviations would leave 194 participants in the analysis and would allow determination of the GMT with meaningful accuracy. Ie., with an assumed Log_{10}-SD of 0.45 and an expected GMT of 2954 (based on pooled data from Phase 3 results in adults), the 95%CI for the GMT would be [2551, 3421] for combined VLA1553 groups and [2397, 3640] for one dose group. Furthermore, these 97 participants per dose group would allow for the detection of a difference in GMTs of 2954 vs. 1943 between the groups with a two-sided significance level of 5% and a statistical power of 80%.

Statistical Methods	All participants entered into the trial, who receive the vaccination, will be included in the safety analysis. The number and percentage of participants with solicited injection site and systemic reactions up to 14 days after vaccination, and with unsolicited AEs (including clinically relevant abnormal lab values), AESI and SAEs will be presented for treatment group overall and by body system / preferred term and will be compared using Fisher's exact test (Fisher-Freeman-Halton test). Changes in laboratory values and the frequency of clinically relevant abnormal values will be analyzed. The immunogenicity analysis will be a comparison of the GMTs in the per-protocol analysis set between the VLA1553 Trial Arms and control at Day 15, Day 29, Day 85, Day 180 and Month 12 post-vaccination by ANOVA (factor Trial Arm, age stratum and baseline serostatus (determined by µPRNT), including pair-wise comparisons (Tukey's HSD test) between VLA1553 Trial Arms. The models will be applied on logarithmic values, and estimates and two-sided 95% confidence intervals for GMTs and GMT ratios will be obtained by back-transformation. GMFI's (Geometric Mean Fold Increase) will be analyzed analogously at various time points. SRRs (Seroresponse Rate) and SCRs (Seroconversion Rate) will be compared using Fisher-Freeman-Halton test, amended by Fisher's exact test for pairwise comparisons. Two-sided 95% confidence intervals will be calculated for SRRs and SCRs 95% according to Altman (Wilson score interval). Immunogenicity analysis will also be stratified by age stratum and by baseline serostatus (determined by µPRNT). A statistical analysis plan will be prepared before database closure/snapshot.
Data Analysis	The following data analyses will be performed: ➢ Part A includes safety and immunogenicity data after all participants have completed Visit 5 (Day 29/ Month 1). ➢ Part B includes safety and immunogenicity data after all participants have completed Visit 7 (Month 6). ➢ Part C includes safety and immunogenicity data after all participants have completed Visit 8 (Month 12). Individual trial parts will be analyzed sequentially. Part A analysis will be performed unblinded (blind will be maintained for trial sites and participants) and a clinical trial report will be compiled. Following the Part B analysis another clinical trial report will be compiled (integrated Clinical Trial Report of Part A+B). For the Part C analysis, a final clinical trial report will be compiled (integrated Clinical Trial Report of Part A+B+C).

Table 5: Schedule of Trial Procedures and Assessments

		Part A					Part B		Part C	
	Visit 0 Screening (Day -28 to Day 0)	**Visit 1** [j] Day 1	**Visit 2** Day 4 (+/-1d)	**Visit 3** Day 8 (+/- 1d)	**Visit 4** Day 15 (+/-1d)	**Visit 5** Day 29 M1 (+/- 4d)	**Visit 6** Day 85 M3 (+/- 7d)	**Visit 7** Day 180 M6 (+/- 14d)	**Visit 8** Day 365 M12 (+/- 14d)	**Visit ET**
Informed consent/ assent [a]	X									
Inclusion/ Exclusion criteria	X	X (Review)								
Demographics [b]	X									
Medical history [c] (including vaccination history)	X	X (Update)								
Randomization		X								
Prior / Concomitant medications	X	X	X	X	X	X	X	X	X	X
Physical examination [d]	X	X	X	X	X	X	X	X	X	X
Vital signs [e]	X	X								
CHIKV Screening sample [1.2 mL]	X									
Pregnancy test (if applicable) [f]	X	X								
Immunogenicity sample [g] [2 mL]		X			X	X	X	X	X	X
Safety sample [h] [4 mL]	X					X				X [n]
Urinalysis [l]		X	X	X	X	X	X	X	X	X
Viremia sample [i] [2 mL]		X	X	X	X	X [k]	X [m]	X [m]	X [m]	X [m]
Maximum total approximate blood volume [mL] Stratum A sentinels/non-sentinels Stratum B sentinels/non-sentinels Stratum C sentinels/non-sentinels	5.2/5.2 5.2/5.2 5.2/5.2	4.0/4.0 4.0/2.0 4.0/2.0	2.0/2.0 2.0/none 2.0/none	2.0/2.0 2.0/none 2.0/none	4.0/4.0 4.0/2.0 4.0/2.0	8.0/4.0 8.0/2.0 8.0/2.0	4.0/4.0 4.0/2.0 4.0/2.0	4.0/4.0 4.0/2.0 4.0/2.0	4.0/4.0 4.0/2.0 4.0/2.0	8.0/6.0 8.0/6.0 8.0/6.0
VACCINATION		X								
Participant Diary		D	R/C/D	R/C/D	R/C					R/C
Memory Aid					D	R/C/D	R/C/D	R/C/D	R/C	R/C
AE/ AESI/ SAE Assessment	X [o]	X	X	X	X	X	X	X	X	X

ET … early termination; R … review; C … collect; D … distribute

[a] Occurs at enrollment before Screening.
[b] Demographics include date of birth, age, height, weight, BMI, gender, race and ethnicity.
[c] Symptoms noted (except those qualifying as a SAE) at Visit 1 (prior to first vaccination) are not considered AEs but will be recorded as medical history. Any prior vaccination should be documented in the Medical History.
[d] A physical examination will be performed on the following body systems being described as normal or abnormal: general appearance, head and neck, eyes and ears, nose and throat, chest, lungs, heart, abdomen, extremities and joints, lymph nodes, skin, and neurological.
[e] Vital signs will include systolic and diastolic blood pressure and pulse rate while seated and at rest, and body temperature measured with infrared thermometer targeting the forehead before vaccination. In addition, after an observation period of 60 minutes (open label phase) or 30 minutes (blinded phase) following vaccination, pulse rate and blood pressure while seated and at rest will again be assessed.
[f] At the screening visit and at indicated visits (i.e., V1) a pregnancy test (in accordance with local regulations) will be performed for all female Participants after onset of menarche and if involved in activities that could result in pregnancy.
[g] Immunogenicity sample [blood: 2 mL] for CHIKV-specific neutralizing antibody titer evaluation.
[h] Safety laboratory sample for hematology and coagulation panel [blood: 4 mL]. Safety sample will be taken from all participants at the screening visit; at subsequent visits (i.e., V5 or ET if earlier than V5) the safety sample will be only taken from the sentinel participants. If deemed necessary by the investigator a safety sample can be taken any time if safety concerns arise.
[i] Viremia sample for investigation of viremia by RT-qPCR [plasma: 2 mL]. Viremia sample will be taken from all participants of Stratum A and only taken from sentinel participants from Stratum B and C.
[j] All procedures/assessments (apart from Participant Diary distribution and AE assessment) occur prior to vaccination (unless stated otherwise).
[k] Visit 5 (Day 29) will be only tested if Visit 4 (Day 15) tests positive.
[l] Urinalysis (i.e., pH, specific gravity, leucocytes, nitrite, protein, glucose, ketones, urobilinogen, bilirubin, erythrocytes) - if possible.
[m] Samples should be obtained for clinically indicated retrospective investigation of viremia by RT-qPCR.
[n] Safety sample should be collected if ET is before Day 29 (Visit 5)-sentinels only.
[o] SAE(s) should be recorded from signed ICF onwards

8. TABLE OF CONTENTS

9. LIST OF ABBREVIATIONS

Abbreviation	Definitions
ACPA	Anticitrullinated Protein Antibody
AE	Adverse Event
AESI	Adverse Event of Special Interest
ALT	Alanine aminotransferase
aPTT	Activated Partial Thromboplastin Time
ART	Anti-retroviral therapy
AST	Aspartate aminotransferase
BMI	Body Mass Index
BSA	Body Surface Area
BLA	Biologics License Application
CDC	Centers of Disease Control and Prevention
CHIKV	Chikungunya virus
CI	Confidence interval
CRA	Clinical Research Associate
CRO	Clinical Research Organization
CRP	C-reactive protein
CSR	Clinical Trial Report
DSMB	Data Safety Monitoring Board
eCRF	Electronic Case Report Form
e.g.	for example
ESR	Erythrocyte sedimentation rate
ER	Emergency Room
ET	Early Termination
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GMT	Geometric mean titer
GMFI	Geometric mean fold increase
Hb	Hemoglobin
HbsAg	Hepatitis B surface antigen
HCV	Hepatitis C virus
Hct	Hematocrit
IB	Investigator’s Brochure
ICF	Informed Consent Form

ICH	International Council on Harmonisation
i.e.	That is
IMM	Immunogenicity Analysis Population
IgG	Immunoglobulin G
IgM	Immunoglobulin M
IMP	Investigational Medicinal Product
kDa	Kilo Dallton
MedDRA	Medical Dictionary for Regulatory Activities
LAR	Legally Acceptable Representative
µL	Microliter
mL	Mililiter(s)
n.a.	Not applicable
NT	Neutralization Test
NT_{50}	Antibody titer at 50% virus neutralization
PCR	Polymerase chain reaction
PIC	Participant Identification Code
pH	Potential of hydrogen
µPRNT	Micro Plaque reduction neutralization test
RF	Rheumatoid Factor
RBC	Red blood cells
RT qPCR	Quantitative reverse transcription polymerase chain reaction
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SRR	Seroresponse Rate
SCR	Seroconversion Rate
TCID	Tissue Culture Infectious Dose
WBC	White blood cell
wt	wild-type

10. INTRODUCTION

10.1 VLA1553

Valneva Austria GmbH has successfully completed phase 1 and two pivotal phase 3 clinical trials of a novel live-attenuated vaccine for prophylaxis of the disease caused by Chikungunya virus (CHIKV) in the adult population. Currently a long-term follow-up trial in adults and pediatric development in adolescents in a CHIKV endemic country, i.e. Brazil, are ongoing. The vaccine (development code VLA1553) is designed to target all globally circulating CHIKV strains. VLA1553 is based on the chikungunya La Reunion strain (LR-CHIKV clone LR2006-OPY1) of the East/Central/South African (ECSA) genotype and is characterized by a genetically engineered large deletion in its nsP3 viral replicase complex which leads to an attenuation of the virus in vivo. VLA1553 is genetically stable and passaged three times on African green monkey kidney (VERO) cells and contains no adjuvant. VLA1553 is intended to facilitate a rapid development of a protective antibody titer.

For further details please refer to the Investigator's Brochure.

10.2 Clinical Condition/ Indication

10.2.1 Transmission, Disease and Diagnosis

CHIKV is a mosquito-borne virus which was first isolated in 1953, during an epidemic of polyarthralgia in Tanzania (Robinson, 1955). The word 'chikungunya' means 'that which bends up' in Swahili, in reference to the stooped posture of participants afflicted with the severe joint pain associated with this disease. Since its first isolation CHIKV was found to occasionally cause smaller outbreaks during the following decades in Africa, India, the Indian Ocean Islands, and Southeast Asia (Suhrbier, Jaffar-Bandjee et al. 2012; Simon, Javelle et al. 2015).

Within the human population, CHIKV is maintained by a human-mosquito-human transmission probably following a dengue-like model characterized by the absence of an animal reservoir and the ability to spread rapidly among human beings via domestic and peri-domestic mosquitoes (Her et al., 2009; Pialoux et al., 2007). CHIKV was typically transmitted only by *Aedes aegypti* mosquitos, however coinciding with an adaptation enabling unusually efficient transmission by *Aedes albopictus* mosquitos, the virus re-merged in 2004 and rapidly spread over Africa, Asia and locally also in Europe (Delisle, Rousseau et al. 2015). More recently CHIKV also has spread across the Americas with millions of people becoming infected (Rezza, Weaver et al. 2019).

CHIKV is an enveloped alphavirus of the family *Togaviridae*. It carries a single-stranded positive sense genomic RNA of around 12 kb containing two open reading frames that encode four non-structural replicase proteins (nsP1-4) and a structural polyprotein consisting of the capsid protein I and the envelope proteins (E3-E2-6K/TF-E1), respectively. The replicase complex serves two functions: it replicates the genomic RNA for incorporation into new virus particles and it also has transcriptase activity to produce a mRNA from a subgenomic promoter that encodes the structural proteins (Simizu, Yamamoto et al. 1984). Mature virions that bud from the plasma membrane of infected cells carry 240 copies of E2-E1 glycoproteins arranged into 80 heterotrimeric spike complexes. The E2 protein is the receptor-binding moiety, whereas the E1 protein is involved in fusion of the virion envelope with the target cell endosomal membrane (Voss, Vaney et al. 2010). Accordingly, it is the spike complex and in particular the E2 envelope protein that is the target for neutralizing antibodies.

An infection with CHIKV may result in chronic and incapacitating arthralgia in some participants affecting all gender and age groups (Couderc, Khandoudi et al. 2009; Suhrbier, Jaffar-Bandjee et al. 2012; Simon, Javelle et al. 2015). CHIKV clinical symptoms typically are presented in three stages that differ in clinical features and treatment. During the acute stage, clinical symptoms manifest with common features such as fever, transient rash and multiple arthralgia/arthritis episodes. This is followed by a multi-morbid post-acute and chronic stage characterized by persisting rheumatic symptoms up to months and years after infection and impaired quality-of-life (Marimoutou, Ferraro et al. 2015; Simon, Javelle et al. 2015). Symptoms of CHIKV disease usually appear 4–7 days post-infection and include rapid onset of fever,

viremia, severe joint pain, recurring mild joint pain, maculopapular rash, and may lead to death (Simon, Javelle et al. 2015).

Individuals who are at higher risk of more serious complications include infants, elderly and individuals with underlying chronic medical conditions. Although children over two years of age have milder disease symptoms and higher rates of asymptomatic infection than adults, severe manifestations such as skin eruptions, meningoencephalitis and septic shock can occur.

Currently, neither specific antiviral treatment nor a vaccine is available to prevent CHIKV infection. Prevention against CHIKV infection is therefore limited to non-treatment interventions such as the employment of insecticides, wearing long sleeves and pants and use of repellents, and other means to restrict exposure to vector mosquitos. The treatment of CHIKV disease is mainly supportive such as bed rest, adequate fluids, and symptomatic relief by using analgesics, antipyretics like paracetamol, or anti-inflammatory drugs like ibuprofen and naproxen for the control of fever and joint pain. Persons who have persistent joint pain may require analgesic or long-term anti-inflammatory therapy (Simon, Javelle et al. 2015).

For detailed information on the disease and epidemiology please refer to the Investigator's Brochure.

10.2.2 Prospects for Vaccine Development

Two Phase 3 trials in adults were successfully completed for this vaccine. In November 2023, the FDA granted Valneva marketing authorization for IXCHIQ®, a vaccine indicated for the prevention of disease caused by chikungunya virus in individuals 18 years of age and older who are at increased risk of exposure to CHIKV (FDA 2023). Currently, Valneva is in preparation for this vaccine becoming available to travelers and persons residing in areas endemic for chikungunya virus.

The first clinical trial investigating VLA1553 in an adolescent population was initiated in January 2022 and recruitment of 754 participants (randomized 2:1 to receive VLA1553 or placebo) was completed in February 2023. Data from ongoing DSMB reviews indicate that the vaccine is generally safe and well-tolerated in the adolescent population. In line with an agreed pediatric development plan with EMA and FDA, we aim to progress age step down studies to younger healthy children.

10.3 Findings From Clinical Studies

10.3.1 Clinical Summary

10.3.1.1 VLA1553-101

In a Phase 1 clinical trial (trial code: VLA1553-101), three dose levels of VLA1553 were administered intramuscularly (i.m.) into the deltoid muscle as a single dose immunization. As primary outcome safety and as secondary outcomes immunogenicity and antibody persistence were investigated. 120 healthy volunteers aged 18 to 45 years were randomly assigned 1:1:2 to one of three escalating dose groups (Group L, low dose of $3.2x10^3$ $TCID_{50}$/0.1 mL; Group M, medium dose of $3.2x10^4$ $TCID_{50}$/1 mL or Group H, high dose of $3.2x10^5$ $TCID_{50}$/1 mL) and received a single-shot immunization on Day 1. Individuals were re-vaccinated with the highest dose on either Month 6 (half of Group H) or 12 (Group L, Group M, and half of Group H) and followed up for 28 days post re-vaccination.

The vaccine was generally safe in all and well tolerated in the low and medium dose group. VLA1553 showed an excellent immunogenicity profile in all dose groups after a single vaccination. 100% seroconversion (defined as the proportion of participants achieving a CHIKV-specific neutralizing antibody titer of NT_{50} ≥20) was achieved 14 days after a single vaccination in all dose groups and sustained at 100% until Month 12. The absence of an anamnestic response following re-vaccination demonstrates that a single vaccination of VLA1553 is sufficient to induce sustaining high titer neutralizing antibodies at all dose levels one year after priming. Even at the Month 12 re-vaccination („intrinsic human viral challenge") vaccinees were protected from vaccine induced viremia and associated clinical symptoms as early indication of VLA1553's efficacy at all dose levels. Based on a superior safety profile along with an improved immunogenicity profile in terms of antibody kinetics the medium dose

($3.2x10^4$ $TCID_{50}$/1 mL) was selected for further development (Wressnigg et al., 2020). For further safety findings please see Section 10.3.1.4.

10.3.1.2 VLA1553-301

A pivotal Phase 3 trial (trial code: VLA1553-301) was performed across 43 U.S. sites (Schneider et al, 2023). The final dose of VLA1553 (target 1x 10^4 $TCID_{50}$/ 0.5 mL) or placebo as control was administered i.m. into the deltoid muscle as a single-shot immunization on Day 1 to 4,115 adults, aged 18 years and above. The trial met its primary endpoint by inducing protective CHIKV neutralizing antibody titers in 98.9% of participants 28 days after receiving a single dose (263 of 266 participants from the per-protocol subgroup tested for immunogenicity), the seroresponse rate [10] (SRR) of 98.9% (95% confidence interval [CI]: 96.7%-99.8%) exceeded the 70% threshold for the lower bound of the 95% CI (for non-acceptance) agreed with the US FDA. The SRR remained high, with 241/246 (98.0%) and 233/242 (96.3%) participants in the VLA1553 group on Day 85 and Day 180, respectively. In this context, seroresponse was defined as an immune response with CHIKV specific neutralizing antibody titers of $\mu PRNT_{50}$ of 150 or higher, which was considered as a threshold reasonably likely to predict clinical benefit against CHIKV infection and was used as a surrogate endpoint to evaluate the efficacy of the vaccine. The titer was accepted by regulatory agencies, including the US FDA and the European Medicines Agency.

Safety data

VLA1553 was generally well tolerated among the 3,082 participants evaluated for safety in trial VLA1553-301. An independent Data Safety Monitoring Board (DSMB) continuously monitored the trial and identified no safety concerns. The safety profile was consistent with clinical results across all age groups (Lot to lot consistency trial, VLA1553-302, described below and preceding Phase 1 clinical trial) and comparable with other vaccines. For further safety findings please see Section 10.3.1.4.

10.3.1.3 VLA1553-302

The above described immunogenicity and safety profile was mirrored in the pivotal lot-to-lot consistency trial (trial code: VLA1553-302), where 408 healthy adults were block randomized 1:1:1 to receive one of three manufacturing lots of VLA1553. The primary endpoint was met as the Confidence Intervals (CIs) of the Geometric Mean Titer (GMT) ratios of all lots were within the defined acceptance margins of 0.67 and 1.5. The antibody titer, determined by µPRNT, peaked at Day 29 with a GMT of 2,643.2; titers subsequently decreased to 846.1 at Day 85 and 708.8 at Day 180. The seroresponse rate was 97.8% (348/356) on Day 29 and remained high, with 97.3% (321/330) and 96.0% (316/329) on Day 85 and Day 180, respectively. Moreover, seroconversion rate, defined as a $\mu PRNT_{50}$ titer of ≥20 in participants seronegative for CHIKV at baseline (Day 1), was achieved in 97.5% (353/362) on Day 29 and remained stable throughout the trial, with 97.6% (327/335) participants maintaining seroconversion on Day 180. For safety findings please see Section 10.3.1.4.

10.3.1.4 Pooled safety analysis

Pooled safety data reflect the findings from the above mentioned trials (VLA1553-301, VLA1553-302, and VLA1553-101) see also Sections 10.3.1.1, 10.3.1.2 and 10.3.1.3. The pooled safety population consists of 4,643 participants: 3,610 randomized and vaccinated with VLA1553 (3,082, 408, and 120 participants, respectively) and 1,033 participants randomized and vaccinated with placebo.

Most solicited systemic AEs described in this population were of mild or moderate severity with a notable difference in the frequency of solicited systemic AEs between the pooled VLA1553 group and the placebo group:

- Mild: 37.5% [95%CI: 35.9%-39.1%] versus 23.4% [95% CI: 20.9%-26.1%];

[10] Also labeled seroprotection rate in the individual clinical trial reports.

- Moderate: 11.1% [95%CI: 10.1%-12.2%] versus 3.1% [95%CI: 2.2%-4.3%];
- Severe: (2.3% [95%CI: 1.8%-2.8%] versus 0.1% [95%CI: 0.0%-0.5%].

A total of 82 (2.3%) participants reported severe solicited systemic AEs: mostly fever (60 participants), arthralgia (10 participants), myalgia (9 participants), fatigue (7 participants), and headache (5 participants).

Similar to the pattern observed for solicited systemic AEs, solicited injection site AEs showed a slightly higher overall frequency between the pooled VLA1553 group than the placebo group (15.2%, 95% CI: 14.1%-16.4% versus 11.1%, 95% CI: 9.4%-13.2%) driven by tenderness and injection site pain.

All solicited injection site AEs were graded as mild or moderate in severity, except for a single solicited injection site AE of pain graded as severe in the pooled VLA1553 group classified as related adverse event.

Tenderness at the injection site was the most common related solicited injection site AE in the pooled VLA1553 group and in the placebo group (10.5% versus 8.0%).

There was a notable difference in overall frequency of unsolicited AEs occurring within 6 months post-vaccination between treatment groups. The overall frequency of unsolicited AEs in the pooled VLA1553 group was 31.6% (95% CI: 30.1%-33.1%) versus 23.9% (95% CI: 21.4%-26.6%) in the placebo group. In decreasing order of frequency, this difference included chills, arthralgia, COVID-19, neutropenia, headache, diarrhea, back pain, leukopenia, and lymphadenopathy.

Up to six months post-vaccination, the frequency of unsolicited reports of musculoskeletal stiffness, joint stiffness, joint swelling, and arthritis was similar between the pooled VLA1553 group and placebo (range: 0.1% to 0.5%).

Another phase 3 clinical trial (VLA1553-304) is currently at initiation status. The trial's objective is to assess additional safety and immunogenicity data in moderately immunocompromised adult participants (aged 18 years and above) infected with HIV living in CHIKV endemic areas. The trial is planned to recruit approx. 75 participants. The first participant is planned to be enrolled in Q1 2024.

More information on the clinical profile of VLA1553 can be found in the Investigator's Brochure.

10.4 Trial Rationale and Justification for Dosage and Trial Design

Two Phase 3 trials with VLA1553 in healthy adults have been successfully completed. The first clinical trial investigating VLA1553 in an adolescent population was initiated in January 2022 and recruitment of 754 participants is completed. Initial safety data generated in trial VLA1553-321, Valneva's first clinical trial in an endemic area and with individuals previously infected with CHIKV, show that VLA1553 was generally safe and well tolerated in adolescents aged 12 to <18 years, regardless of the CHIKV antibody baseline serostatus. Data from trial interim analysis at Day 29 and also from ongoing DSMB reviews confirmed that the vaccine is generally safe and well-tolerated in the adolescent population (please also refer to the Investigator's Brochure). The rationale for performing the present trial (VLA1553-221) is to evaluate the vaccine VLA1553 in the generally healthy pediatric population and to assess the safety and immunogenicity of a single dose of VLA1553 in line with an EMA and US FDA agreed pediatric development plan. In this Phase 2 trial (VLA1553-221), two different VLA1553 dose levels will be evaluated for tolerability, safety and immunogenicity in at least 300 healthy children (up to 10% baseline seropositive and at least 90% baseline seronegative for CHIKV antibodies) aged 1 to 11 years. This safety and immunogenicity trial will contribute to the safety database and will allow identification of the appropriate dose level in this pediatric population for testing in Phase 3. The trial will be carried out in CHIKV endemic countries in Latin America.

10.4.1 Selection of Trial Population

At least 300 healthy children aged 1 to 11 years of either gender who meet all inclusion criteria and none of the exclusion criteria and for whom their parent(s)/LAR(s) provide a written informed consent and participants provide a written informed assent if applicable will be invited to participate in the trial.

Participants will be screened for evidence of previous CHIKV exposure and up to 10% baseline seropositive and at least 90% baseline seronegative healthy participants will be enrolled.

10.4.2 Selection of Seroresponse Threshold

The seroresponse threshold titer, i.e., a $\mu PRNT_{50}$ titer ≥150 (Roques et al., 2022) was accepted by regulators such as US FDA and EMA as a threshold reasonably likely to predict clinical benefit against CHIKV infection, as induced by the VLA1553 vaccine.

10.5 Evaluation of Anticipated Risks and Benefits of the Investigational Product(s) to Human Participants

10.5.1 Possible Benefits for Participants

The benefit of participation for the participants in this trial is the possible formation of antibodies against CHIKV after vaccination with the VLA1553 vaccine or Neisseria meningitides in the control (Nimenrix) arm. During their participation in this trial, the participants' CHIKV antibody development will be monitored. Following vaccination with Valneva's VLA1553 vaccine, participants may acquire immunity to CHIKV as indicated by antibody levels above the pre-defined threshold. Following vaccination with the Nimenrix vaccine in the control arm, participants may acquire immunity to meningococcal disease.

Another benefit for participants involved in this trial is derived from safety monitoring and follow-up as prophylactic measure for an early diagnosis of a new illness.

10.5.2 Possible Benefits for Society

CHIKV is currently regarded as one of the most-likely re-emerging viruses to spread globally (Simon, Savini et al. 2008), an issue which raises an urgent demand for efficient prophylaxis. At present IXCHIQ® is the first approved vaccine available against this CHIKV-induced debilitating disease (FDA 2023) and its various symptoms (Ahola et al. 2015; Weaver et al. 2012; Weaver and Lecuit 2015). Thus, morbidity due to this virus is a serious threat to global health and CHIKV has now been listed as a priority pathogen by the National Institutes of Allergy and Infectious Disease (NIAID) in the United States (Ahola, Couderc et al. 2015; Weaver and Lecuit 2015; NIAID October 26, 2016).

Chikungunya virus epidemics are explosive, rapidly-spreading and unpredictable. Originating in tropical and sub-tropical countries, outbreaks have spread in recent years to more temperate regions of the world, including Europe, due to the increase in global travel and trade. Infection is not usually life-threatening for healthy individuals including pediatrics, but the disease burden associated with the illness is a serious threat to global public health. Studies indicate that children over 2 years of age typically have milder disease symptoms and higher rates of asymptomatic infection than adults, however there can be a significant loss in the quality of life for children and adolescents who are affected by inflammatory relapses, rheumatic arthritis and joint deformation resulting in pain and debilitation, which is long-lasting in a substantial number of cases and even irreversible in some. Severe outcomes have been reported in infants under 2 years of age, which have been associated with neurological conditions and fever due to CHIKV with associated secondary bacterial infections. As currently no specific treatment is available to prevent or cure CHIKV disease in either the adult or pediatric populations, there is therefore an urgent medical need for a prophylactic approach against CHIKV.

The development of life-long immunity to natural CHIKV infection suggests that a vaccine containing live-attenuated virus which mimics natural infection but is unable to cause CHIKV disease, has the potential to generate life-long protection against it. The availability of a vaccine which could be used in the pediatric population, to provide active immunization for the prevention of disease caused by CHIKV infections of globally circulating chikungunya genotypes, and protect infants, children, and adolescents who live in or travel to endemic or high-risk areas, is anticipated to fulfil a therapeutic need in this population.

Through the participation in this Phase 2 trial, the participants will contribute to further development of a novel vaccine against chikungunya.

10.5.3 Possible Risks / Inconveniences for the Participant

In trial VLA1553-301, solicited AEs were reported by 52.8% of participants, and 49.0% of participants experienced any related solicited AE. The most frequently reported related solicited systemic AEs following vaccination with VLA1553 included headache (31.4%; 969/3082), fatigue (28.5%; 879/3082) and myalgia (23.8%; 735/3082). A total of 58 (1.9%) participants reported related severe solicited AEs, predominantly fever (39 participants), arthralgia (9 participants), myalgia (8 participants), fatigue (4 participants), and headache (3 participants).

With regard to local AEs, 15.0% (463/3082) of participants reported any local AE following vaccine administration. The most common solicited local AEs after vaccination with VLA1553 were tenderness (10.6%; 328/3082) and pain (6.2%; 191/3082) with the majority of local AEs being either mild or moderate in severity. In total 30.3% (933/3092) of participants who received VLA1553 reported any unsolicited AE. Related unsolicited AEs were reported by 9.8% (303/3082) of participants.

Eleven participants, of which 10 had received VLA1553, met the criteria of an AESI. Five cases of fever in combination with arthralgia (4 participants in the VLA1553 arm and 1 in the placebo arm), three cases of fever in combination with arthralgia and back pain, including one participant who reported in addition lymphadenopathy, two cases of fever in combination with back pain, including one participant who reported in addition rash, and one case of fever in combination with oedema and arthralgia/arthritis (in the VLA1553 arm). However, most of the symptoms contributing to an AESI were mild or moderate and were self-limited after 2 to 4 days. Furthermore, each AESI case was assessed by the DSMB. The DSMB performed a thorough review of each case and advised whether additional clinical work-up was required.

During the licensure procedure of VLA1553, the FDA requested a post-hoc analysis with a revised AESI definition i.e., fever (≥38 °C / 100.4 °F) and one or more of any of the following: arthralgia or arthritis, myalgia, headache, back pain, rash, lymphadenopathy, or certain neurological, cardiac or ocular symptoms occurring within 30 days after the vaccination, regardless of the order of their onset and their duration.

Using this broader AESI definition, 361 (11.7%) in the VLA1553 group (n=3,082) reported AESI (synonym: chikungunya-like adverse reaction), including 48 participants (1.6%) who reported severe[11] AESI (synonym: severe chikungunya-like adverse reaction). Six (0.6%) participants in the placebo group (n=1,033) reported AESI, none of which were severe.

Using the AESI definition according to FDA criteria, prolonged (duration at least 30 days) AESI were identified in fourteen VLA1553 recipients (median duration 94 days, range 30 days to at least 6 months) (IXCHIQ Prescribing Information 2023).

In the VLA1553-302 trial solicited Injection site AEs were reported by 19.4% (79/408) of participants vaccinated with VLA1553. All of the solicited local AEs were mild or moderate. The most common (≥ 10%) related solicited local AE was tenderness (14.2%). Solicited systemic AEs were reported by 57.1% of participants. Most solicited systemic AEs were mild or moderate; 11 were severe and all were related. Of the 11 participants who reported related severe solicited AEs, they reported: fever (9 participants), fatigue (2 participants), arthralgia (1 participant), myalgia (1 participant), and headache (1 participant). The most common (≥ 20%) related solicited systemic AEs were fatigue (36.3 %), headache (33.6 %) and myalgia (22.3 %). 1 participant met the criteria of AESI, the events experienced were mild arthralgia (6 days) and severe fever (3 days) but the events were self-limited. Unsolicited AEs were reported by 34.1% of participants; 1.5% (6 participants/8 events) were severe.

DSMB data from the currently ongoing adolescent VLA1553-321 do not indicate an issue so far in a pediatric population. Initial safety data generated in trial VLA1553-321, Valneva's first clinical trial in an

[11] Prevented daily activity and/or required medical intervention.

endemic area and with individuals previously infected with CHIKV, show that VLA1553 was generally safe and well tolerated in adolescents aged 12 to <18 years, regardless of the CHIKV antibody baseline serostatus.

To date no clinical data with VLA1553 is available in children aged 1 to 11 years.

Allergic reactions to components of the vaccine - in the worst case – an anaphylactic shock cannot be excluded, although such reactions have been reported in very rare cases with other vaccinations.

The possibility that vaccination may activate an autoimmune disease (e.g., multiple sclerosis) in predisposed participants cannot be excluded.

In addition, as with any IMP, there may be unforeseeable risks associated with the use of the VLA1553 vaccine.

The blood draws performed during the trial carry the possible risks of pain, hematoma, and in very rare cases an infection at the venipuncture site. The process of vaccination may also trigger syncope.

For further details on known and potential risks of VLA1553, please refer to the Investigator's Brochure.

Nimenrix

Nimenrix is a vaccine used to protect adults, adolescents and children from the age of 6 weeks against invasive meningococcal disease caused by the bacterium Neisseria meningitidis (group A, C, W-135, and Y). **Table 6** shows the adverse reactions reported from the studies in participants aged from 6 weeks up to 55 years of age and post-marketing experience. Adverse reactions reported are listed according to the following frequency categories:

Very common: (≥1/10)

Common: (≥1/100 to <1/10)

Uncommon: (≥1/1,000 to <1/100)

Rare: (≥1/10,000 to <1/1,000)

Very rare: (<1/10,000)

Not known (cannot be estimated from available data)

Table 6: Tabulated summary of adverse reactions by system organ class – Nimenrix

System Organ Class	Frequency	Adverse reactions
Blood and lymphatic system disorders	Not known***	Lymphadenopathy
Metabolism and nutrition disorders	Very common	Appetite lost
Psychiatric disorders	Very common	Irritability
	Uncommon	Insomnia Crying
Nervous system disorders	Very common	Drowsiness Headache
	Uncommon	Hypoaesthesia Dizziness
	Rare	Febrile convulsion
Gastrointestinal disorders	Common	Diarrhoea Vomiting Nausea*

Skin and subcutaneous tissue disorders	Uncommon	Pruritus Urticaria Rash**
Musculoskeletal and connective tissue disorders	Uncommon	Myalgia Pain in extremity
General disorders and administration site conditions	Very common	Fever Swelling at injection site Pain at injection site Redness at injection site Fatigue
	Common	Injection site haematoma*
	Uncommon	Malaise Injection site induration Injection site pruritus Injection site warmth Injection site anaesthesia
	Not known***	Extensive limb swelling at the injection site, frequently associated with erythema, sometimes involving the adjacent joint or swelling of the entire injected limb

*Nausea and Injection site haematoma occurred at a frequency of Uncommon in infants
**Rash occurred at a frequency of Common in infants
***ADR identified post-marketing

10.6 COVID-19 Management

In May 2023, the end to the global Public Health Emergency of International Concern (PHEIC) for COVID-19 was declared (WHO/IHR 2023). The trial sponsor will continuously monitor and evaluate the development of the COVID-19 progression in the area of trial sites to determine if any measures need to be implemented to mitigate undue risks to the participants or in response to local governmental recommendations. Any measure would be communicated to the relevant CA and IRB.

11. TRIAL PURPOSE AND OBJECTIVES

11.1 Trial Purpose

To obtain tolerability, safety and immunogenicity data of two different dose levels of a live-attenuated CHIKV vaccine (VLA1553) in healthy children aged 1 to 11 years. The outcome shall provide the basis for dose selection for further development of the vaccine in this age group.

11.2 Primary Objective

To evaluate the tolerability of the full dose and half dose formulation of the live-attenuated CHIKV vaccine (VLA1553) following vaccination in healthy children aged 1 to 11 years after a single immunization.

11.3 Secondary Objectives

- To assess the immunogenicity of the full dose and half dose formulation of VLA1553 in healthy children aged 1 to 11 years after a single immunization
- To assess the safety of the full dose and half dose formulation of VLA1553 in healthy children aged 1 to 11 years after a single immunization
- To identify the optimal dose level(s) of VLA1553 in healthy children aged 1 to 11 years.

11.4 Exploratory Objective

- To evaluate the efficacy of VLA1553 in pediatric participants in CHIKV endemic areas after a single immunization.

12. TRIAL DESIGN

12.1 Overall Trial Design

This is a multicenter, prospective, randomized, observer-blinded, three arm, phase 2 clinical trial evaluating the full dose formulation (target 4.0 log 10 TCID50 per 0.5mL) of VLA1553, half dose formulation (target 3.7 log 10 TCID50 per 0.5mL) and control (Nimenrix).

At least 300 male and female healthy children aged 1 to 11 years will be enrolled and the overall distribution of participants will be 2:2:1 to the two VLA1553 dose groups (n=120 each) or control (Nimenrix) (n=60).

These volunteers will be screened for evidence of previous CHIKV exposure. Enrolment will then be stratified by CHIKV serostatus at baseline, targeting:

- up to 10 % CHIKV seropositive participants (i.e. IgM+/IgG+ or IgM-/IgG+)
- at least 90% CHIKV seronegative participants (i.e. IgM-/IgG-)

Participants who are IgM+/IgG- do not qualify for participation in this trial. Please see Section 17.8.4 for details on CHIKV antibody screening and assessment of results.

Note: CHIKV IgM+/IgG- is suggestive for an early phase acute CHIKV infection. Therefore, as safety precaution, no vaccination with the attenuated CHIKV vaccine VLA1553 should be applied.

The clinical trial design is displayed in the

Figure 1 below.

Figure 1: Phase 2 Clinical Trial Design

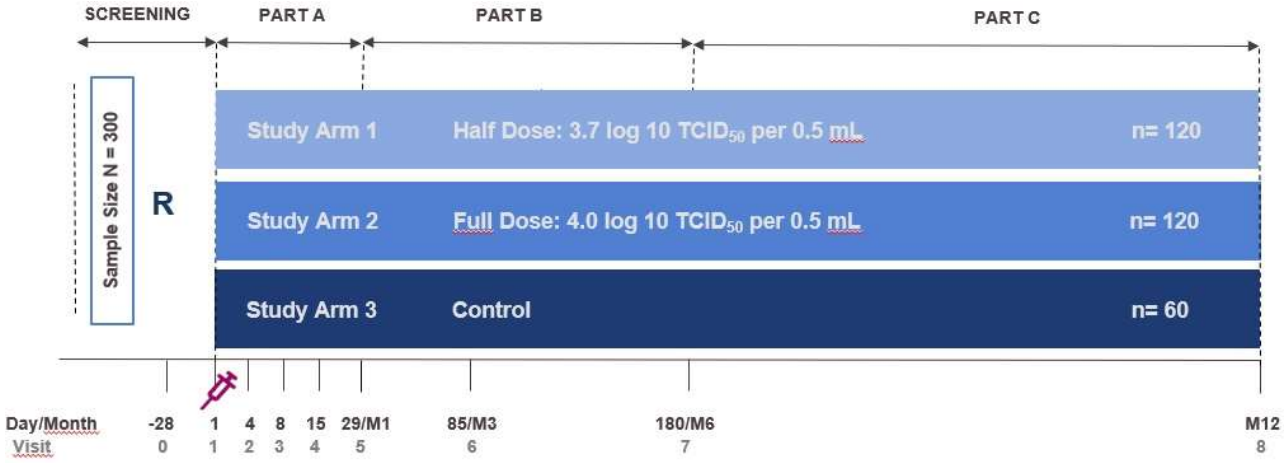


Table 7 below illustrates the participant distribution.

Table 7: Participant Distribution

Trial Arm	Dosage ($TCID_{50}$/dose)	Stratum	Cohort I	Cohort II	Cohort III	Cohort IV	Total N
1	VLA1553 half dose 3.7 log 10	A (7 to 11 yrs.)	5*+35	-		-	40
		B (3 to 6 yrs.)	-	5*+35	-		40
		C (1 to 2 yrs.)	-	-	5*+35	-	40
2	VLA1553 full dose 4.0 log 10	A (7 to 11 yrs.)	-	5*+35		-	40
		B (3 to 6 yrs.)	-	-	5*+35		40
		C (1 to 2 yrs.)	-	-	-	5*+35	40
3	Control	A (7 to 11 yrs.)	10	10		-	20
		B (3 to 6 yrs.)	-	10	10		20
		C (1 to 2 yrs.)	-	-	10	10	20
		C (1 to 2 yrs.)	-	-	10	10	20

*Sentinel enrolment will be conducted in open-label fashion and sentinels recruited at a preferably single trial site. Vaccinations in the following cohort can be started based on the previous cohort sentinels' review by the Data Safety Monitoring Board and recommendation to the Sponsor and final Sponsor confirmation will be provided to proceed with the trial.

At Day 1 (Visit 1) VLA1553 or Nimenrix (control) will be administered intramuscularly (in deltoid muscle for Stratum A and B; anterolateral thigh muscle for Stratum C) as a single dose immunization. Participants will return to trial site at Day 4 (i.e., Visit 2, three days after vaccination) and Day 8 (i.e. Visit 3, seven days after vaccination) for physical examination, safety evaluation and viremia assessments (as applicable). Furthermore, participants will visit trial site at Day 15 (Visit 4), Day 29 (Visit 5), Month 3 (Day 85, Visit 6), Month 6 (Day 180, Visit 7) and Month 12 (Day 365) for safety evaluations and collection of samples for immunogenicity assessments. Safety laboratory evaluations will be done at Visit 5 (Day 29) from all participants of age Stratum A (7 to 11 yrs.) and all sentinel participants from Stratum B (3 to 6 yrs.) and Stratum C (1 to 2 yrs.). In addition, urinalysis will be done at every trial visit from Day 1 (Visit 1) until Month 12 (Visit 8) from all participants, if possible. Viremia samples will be collected from all participants of Stratum A and sentinel participants from Stratum B and C only at Visit 1 to 8. Viremia samples from Day 1 - Day 15 (Visit 1- Visit 4) will be analyzed. Should the sample on Day 15 (Visit 4) after vaccination test positive for viremia, the collected Day 29 (Visit 5) sample will be analysed. Other viremia samples will be tested for clinically indicated retrospective analysis.

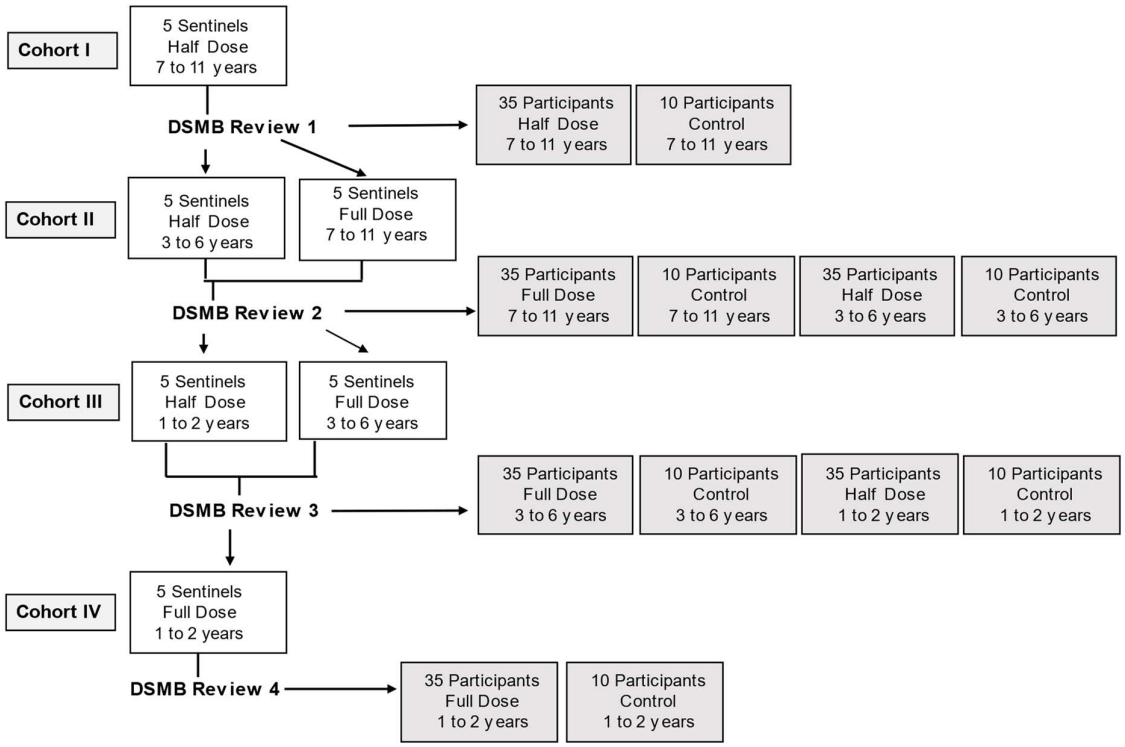
For a tabular outline of trial procedures and assessments required at each visit, please see the Schedule of Trial Procedures and Assessments (**Table 5**).

12.1.1 Participants Enrollment

Staggered enrolment approach

As a safety precaution measure, 30 sentinel participants will be enrolled into the trial in an open-label fashion according to an age step down scheme outlined in **Figure 2.**

Figure 2: Staggered enrolment



White box: open-label, Grey box: blinded, randomized

After sentinel analysis, participants will be enrolled in a blinded, randomized manner into three Trial Arms. Within each treatment arm participants will be stratified into three age strata:

Stratum A: 7 to 11 years - children from their 7th birthday until the day before their 12th birthday.

Stratum B: 3 to 6 years - children from their 3rd birthday until the day before their 7th birthday.

Stratum C: 1 to 2 years - children from their 1st birthday until the day before their 3rd birthday.

Age strata for the VLA1553 treatment arms are targeted to be equal in size, i.e., approximately 40 per age stratum.

The age step down phase of the trial will be preferably initiated at a single site to allow permanent oversight on safety data by one principal Investigator. Participants will be recruited in four cohorts as follows:

Cohort I:

Cohort I will be comprised of 5 sentinels of Stratum A (7 to 11 years) receiving the half dose formulation of VLA1553 in an open-label fashion, in a blinded fashion 35 participants of Stratum A (7 to 11 years) receiving the half dose formulation, 10 participants of Stratum A (7 to 11 years) receiving control.

A maximum of three sentinels of Cohort I are to be vaccinated on the first day with vaccinations evenly distributed during the day (e.g. at least 1 hour between participants). Specifically, one sentinel will be vaccinated, observed at the trial site for 1 hour after vaccination for safety and reactogenicity. If no immediate safety concerns arise as determined by the Investigator, then the next sentinel of this Cohort is to be vaccinated and again observed for one hour etc.

These three sentinels of Cohort I are to return to the site 3 days later (Visit 2) for safety follow-up. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of Cohort I are to be vaccinated and are to return to the site 3 days later (Visit 2) for safety

follow-up. All sentinels of Cohort I are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

The available Day 8 safety data of these five sentinels of Cohort I will be reviewed by an independent Data Safety Monitoring Board (DSMB) during **DSMB Review 1.** Based on no reservations, a recommendation will be given on whether to proceed with the randomization of the remaining 45 participants of Stratum A in Cohort I and to start vaccination in Cohort II.

Cohort II:

Cohort II will consist of 5 sentinels of Stratum B (3 to 6 years) receiving the VLA1553 half dose and 5 sentinels of Stratum A (7 to 11 years) receiving the full dose in an open-label fashion, and in a blinded fashion 35 participants of Stratum B (3 to 6 years) receiving the half dose, 35 participants of Stratum A (7 to 11 years) receiving the full dose, 10 participants of Stratum A (7 to 11 years) and 10 participants of Stratum B (3 to 6 years) receiving control.

Similarly, a maximum of three sentinels per stratum are to be vaccinated open-label evenly distributed during the day. One hour observation time after vaccination for immediate safety and reactogenicity evaluation must be adhered. These three sentinels per stratum of Cohort II are to return to the site 3 days later (Visit 2) for safety follow-up. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of Cohort II are to be vaccinated and are to return to the site 3 days later (Visit 2) for safety follow-up. All sentinels of Cohort II are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

Available Day 8 safety data of Cohort II, as well as cumulative safety data, will be reviewed by the DSMB (as described earlier) during the **DSMB Review 2** and based on no reservations, a recommendation will be given on whether to proceed with the randomization of the remaining 90 participants in Cohort II and to start vaccination in Cohort III.

Cohort III:

Cohort III will consist of five sentinels of Stratum C (1 to 2 years) receiving the half dose formulation in an open-label fashion, five sentinels of Stratum B (3 to 6 years) receiving the full dose in an open-label fashion and in a blinded fashion 35 participants of Stratum C (1 to 2 years) receiving the half dose, 35 participants of Stratum B (3 to 6 years) receiving the full dose, 10 participants of Stratum B (3 to 6 years) and 10 participants of Stratum C (1 to 2 years) receiving control.

A maximum of three sentinels per stratum are to be vaccinated open-label evenly distributed during the day. One hour observation time after vaccination for immediate safety and reactogenicity evaluation must be adhered. These three sentinels per stratum of Cohort III (five sentinels of Stratum C receiving half dose formulation, and five sentinels of Stratum B receiving full dose) are to return to the site 3 days later (Visit 2) for safety follow-up. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of each age strata of Cohort III are to be vaccinated and are to return to the site 3 days later (Visit 2) for safety follow-up. All sentinels of Cohort III are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

Available Day 8 safety data of Cohort III, as well as cumulative safety data, will be reviewed by the DSMB (as described earlier) during the **DSMB Review 3** and based on no reservations, a recommendation will be given on whether to proceed with the randomization of the remaining 90 participants in Cohort III and to start vaccination in Cohort IV.

Cohort IV:

Cohort IV will consist of five sentinels of Stratum C (1 to 2 years) receiving the full dose in an open-label fashion, and in a blinded fashion 35 participants of Stratum C (1 to 2 years) receiving the full dose, 10 participants of Stratum C (1 to 2 years) receiving control.

A maximum of three sentinels of Stratum C receiving the full dose are to be vaccinated open-label evenly distributed during the day. One hour observation time after vaccination for immediate safety and reactogenicity evaluation must be adhered. These three sentinels of Cohort IV will be followed for safety (Visit 2) three days later. If no safety concern arises during Visit 2 and none of the pre-defined holding rules apply, the remaining two sentinels of Cohort IV are to be vaccinated and are to return to the site 3

days later (Visit 2) for safety follow-up. All sentinels of Cohort IV are to return to the trial site 7 days post-vaccination for safety follow-up (Visit 3).

Available Day 8 safety data of Cohort IV, as well as cumulative safety data, will be reviewed by the DSMB (as described earlier) during the **DSMB Review 4** and based on no reservations, a recommendation will be given on whether to proceed with the randomization of the remaining 45 participants of Stratum C (1 to 2 years) in Cohort IV.

12.1.2 Holding rules

The safety holding rules defined below will be assessed by the CRO and the Sponsor on a continuous basis. The holding rules trigger recruitment, screening and vaccination stop until available safety data has been reviewed by the DSMB and their recommendation and Sponsor decision is available whether or not to proceed with enrollment and vaccinations:

- **One** or more participants experience an SAE with no likelier alternative cause than the trial vaccine (i.e. possibly or probably related);
- **Two** or more participants experience the same Grade 3 (severe) or higher solicited injection-site reaction that (1) occurs following vaccination and (2) lasts longer than 3 days and (3) has no likelier alternative cause than the trial vaccine;
- **Two** or more participants experience the same Grade 3 (severe) or higher solicited systemic reaction that (1) occurs following vaccination, (2) lasts longer than 3 days, and (3) has no likelier alternative cause than the trial vaccine;
- **Two** or more participants experience the same Grade 3 (severe) or higher unsolicited AE, including Grade 3 or higher abnormal laboratory values that are assessed to be clinically relevant by the Investigator, that (1) occurs after vaccination, and (2) has no likelier alternative cause than the trial vaccine.

The DSMB can issue a recommendation to stop the trial, amend the trial protocol or to discontinue a trial group during planned or ad-hoc DSMB meetings, e.g., in response to an excess rate of AEs with the same suspected underlying pathological mechanism.

12.2 Trial Termination – Stopping Rules

The trial may be put on hold or prematurely terminated, after consultation with the DSMB, if vaccine-related SAEs or other significant vaccine-related side effects occur and result in a safety concern.

Additionally, vaccination of further participants may be put on hold during the sentinel recruitment phase in case an ad-hoc DSMB meeting is triggered (for more information see Section 17.13.1).

If the Sponsor decides to terminate the trial before it is completed, the Investigator will be notified in writing, stating the reasons for early termination. In terminating the trial, the Sponsor and the Investigator will ensure that adequate consideration is given to the protection of the participants' interests. The Sponsor or Investigator will notify the relevant regulatory authorities or IRB in writing in accordance with local requirements. Documentation will be submitted for filing in the Investigator Site File and the Trial Master File.

12.3 Trial Duration

The overall trial duration (First Participant In – Last Participant Out) is estimated to be approximately 17 months.

Individual participation is approximately 13 months from enrollment to trial completion, unless prematurely discontinued.

12.4 Trial Endpoint

12.4.1 Primary Endpoint

The primary endpoint is to assess tolerability through the frequency and severity of solicited injection site and systemic reactions within 14 days post-vaccination.

12.4.2 Secondary Endpoints

Safety

- Frequency and severity of any adverse event (AE) within 28 days post-vaccination;
- Frequency and severity of unsolicited AE until Month 6 (Day 180) and Month 12 (Day 365) post-vaccination;
- Frequency and severity of any serious adverse event (SAE) until Month 6 (Day 180) and Month 12 (Day 365) post-vaccination;
- Frequency and severity of any early onset adverse event of special interest (AESI) starting within 2 to 21 days post-vaccination (i.e. Day 3 – Day 22);
- Frequency and severity of any late onset adverse event of special interest (AESI) during the entire trial starting 22 days post-vaccination (i.e. Day 23 – trial end);
- Assessment of viremia on Days 1, 4, 8, and 15 and beyond as applicable;

Immunogenicity

- Immune response in baseline seronegative participants as measured by CHIKV-specific neutralizing antibody titers on Day 1, Day 15, Day 29, Day 85, Day 180 and Month 12 post-vaccination as determined by µPRNT assay;
- Proportion of participants with seroconversion[12] as compared to baseline at Day 15, Day 29, Day 85, Day 180 and Month 12 as determined by µPRNT assay;
- Proportion of participants with a seroresponse (defined as $\mu PRNT_{50}$ ≥150 for baseline negative participants) [13] on Day 15, Day 29, Day 85, Day 180 and Month 12 post-vaccination as determined by µPRNT assay;
- Fold increase of CHIKV-specific neutralizing antibody titers determined by µPRNT assay at Days 15, 29, 85, 180 and at Month 12 post-vaccination as compared to baseline;
- Proportion of participants reaching an at least 4-fold, 8-fold, 16-fold or 64-fold increase in CHIKV-specific neutralizing antibody titers compared to baseline at Day 15, Day 29, Day 85, Day 180 and Month 12 as measured by µPRNT assay;
- Antibody titers, seroresponse, seroconversion and fold increases for CHIKV-specific neutralizing antibodies, determined by µPRNT assay at Day 15, Day 29, Day 85, Day 180 and Month 12 post-vaccination stratified by baseline serostatus (based on µPRNT), age stratum and dose.

12.4.3 Exploratory Endpoint

Incidence of CHIKV infections with onset after 15 days post-vaccination (i.e. Day 16) as evidenced by viremia by virus specific RT-qPCR, clinical diagnosis and seroconversion by µPRNT for the entire trial period - measured in Stratum A and in sentinel participants of Stratum B and Stratum C

[12] Seroconversion defined as >4-fold increase of $\mu PRNT_{50}$ compared to baseline $\mu PRNT_{50}$ titer at Day 1.
[13] Seroresponse threshold derived from animal passive transfer experiments (Roques et al., 2022) .

12.5 Randomization and Blinding

At least 300 male and female healthy children aged 1 to 11 years will be enrolled and will be randomized in a 2:2:1 ratio to VLA1553 half dose formulation, VLA1553 full dose or control group as described in Section 12.1.

In order to minimize/avoid bias, assignment into one of these trial arms will be blinded for the participants and the site staff performing the safety assessments as well as the biostatistician (i.e. double-blind).

Each participant will have a unique participant number obtained via the randomization and trial supply management system (RTSM) and assigned at the screening visit. The Investigator will keep a record (i.e. the participant screening log) of participants who entered screening.

Randomization will be performed via the RTSM. At Day 1 (Visit 1, Day of vaccination) eligible participants will be assigned to VLA1553 or control. Participants will be allocated to trial arms according to the randomization code.

The IMP will be prepared by unblinded trial staff in accordance with the information obtained from the RTSM.

An overview of persons who will be (un)blinded is provided below:

Unblinded:

- Designated trial staff who randomize participants to trial arms and are responsible for IMP handling (i.e. perform preparation and administration of the trial vaccine, maintain the drug dispensing log). These unblinded trial staff members will not be involved in any other trial procedures/assessments;
- CRA's responsible for monitoring of IMP handling and for verifying drug accountability during the trial and performing overall drug accountability;
- DSMB members;
- Dedicated statistical team at the CRO performing statistical analyses for generation of unblinded safety data tables for the DSMB;
- Designated trial staff at the Sponsor and CRO/CMO involved in IMP oversight, IMP supply and IMP temperature excursions;
- Designated trial staff at the Sponsor and at central laboratory for immunogenicity and viremia assessments;
- Clinical Safety Team: Clinical Safety Manager and Clinical Safety Physician (at Valneva), and Clinical Safety Team at Safety Service Provider.

Blinded:

- Investigators and other trial staff involved in general trial conduct and safety assessments;
- Trial participants and their parent(s)/LAR(s);
- CRA's responsible for monitoring trial data;
- All laboratory personnel at central and local laboratories for safety laboratory assessments;
- All other Sponsor and CRO staff including medical monitor.

For further details please refer to the trial-specific Blind Maintenance Plan.

12.5.1 Blinding Process

In order to ensure the blinding of trial participants and site staff performing the safety assessments with respect to the vaccine dose, preparation of IMP must be done by unblinded staff members in a separate room applying the 4-eyes principle, unobserved by blinded staff members and the participant/parent(s)/LAR(s). Administration of the vaccine must be performed by unblinded trial staff members. The unblinded trial staff must not discuss randomizations with blinded trial clinicians/trial staff members and will not be involved in any other trial related procedures/assessments.

For further details please refer to the IMP Manual and Blind Maintenance Plan.

12.5.2 Unblinding

The randomization assignment is not to be revealed except in emergency cases in which unblinding is necessary for the clinical management of an SAE. In such events, the Investigator may consult the Sponsor before breaking the blind. In any case the Investigator must inform the Sponsor immediately after unblinding has been performed.

In case of emergency, the vaccine administered to the trial participant can be revealed by the Investigator through the RTSM.

13. PARTICIPANT SELECTION, WITHDRAWAL AND DISCONTINUATION

At least 300 healthy children aged 1 to 11 years of either gender who satisfy the inclusion and exclusion criteria listed below will be invited to participate in the trial.

13.1 Inclusion Criteria

Participants who meet **ALL** of the following criteria are eligible for this trial:

1. Male or female healthy children aged 7 to 11 years[14] for Stratum A, 3 to 6 years[15] for Stratum B and 1 to 2 years[16] for Stratum C at the time of vaccination;
2. Written informed consent by the participant's parent(s)/Legally Acceptable Representative(s) ((LAR(s)), according to local requirements, and written informed assent of the participant, if applicable;
3. Participant is seropositive for previous CHIKV exposure (i.e. IgM+/IgG+ or IgM-/IgG+) or seronegative (i.e. IgM-/IgG-) (see Section 17.8.4 for details on CHIKV antibody screening and assessment of results);
4. Participant is generally healthy [17] as determined by the Investigator's clinical judgement based on medical history, physical examination and screening laboratory tests;
5. For participants in Stratum C 1 to 2 years:
   Participant was born at full term of pregnancy (≥37 weeks) with a birth weight ≥2.5 kg;
6. If participant is of childbearing potential (after onset of menarche) and if involved in activities that could result in pregnancy:
   d) Participant has a negative pregnancy test (in accordance with local regulations) at screening (Visit 0) and Day 1 (Visit 1), respectively
   e) Participant has practiced an adequate method of contraception during at least the 30 days before screening (Visit 0) and during the screening period
   f) Participant agrees to employ adequate birth control measures for the first three months post-vaccination (i.e. until Day 85, Visit 6). This includes one of the following measures:
      - Hormonal contraceptives (e.g. implants, birth control pills, patches);
      - Intrauterine hormone-release systems;
      - Barrier type of birth control measure (e.g. condoms, diaphragms, cervical caps);
      - Vasectomy in the male sex partner ≥3 months prior to first vaccination;
      - Same-sex relationships.

13.2 Exclusion Criteria

Participants who meet **ANY** of the following criteria are **NOT** eligible for this trial:

1. Participant who is anti-CHIKV IgM+/IgG- does not qualify for participation in this trial.
2. Participant is taking medication or other treatment for unresolved symptoms attributed to a previous CHIKV infection; or has participated in a clinical trial involving an investigational CHIKV vaccine;

[14] From the 7th birthday to the last day before the 12th birthday.
[15] From the 3rd birthday to the last day before the 7th birthday.
[16] From the 1st birthday (12 months) to the last day before the 3rd birthday (< 36 months).
[17] Participants are considered ***generally healthy*** if they have no (1) active disease (acute illness, exacerbation of a chronic condition or a chronic – progressive illness) which requires treatment and/or follow-up, or (2) disease that is identified as an exclusion criterion.

3. Participant has an acute or recent infection (and who is not symptom-free in the week prior to the Screening Visit (Visit 0);
4. Parent(s)/LAR(s) or siblings have a known HIV infection;
5. Participant has received another live virus vaccine within 28 days or inactivated vaccine [18] within 14 days prior to vaccination in this trial or plans to receive a live virus vaccine within 28 days or inactivated vaccine within 14 days after vaccination;
6. Participant has abnormal findings in any required trial investigations (including medical history, physical examination, and clinical laboratory) considered clinically relevant by the Investigator which pose a risk for participation in the trial based on his/her judgement;
7. Participant has an ongoing medical history of or currently has acute or progressive, unstable or uncontrolled clinical conditions (e.g. cardiovascular, respiratory, neurologic, psychiatric, or rheumatologic conditions) that poses a risk for participation in the trial, based on Investigators clinical judgement. Examples include individuals with poorly controlled or unstable disease, ongoing suspected or active inflammation, or poor compliance with pharmacologic treatment, or presence of high-risk comorbidities (e.g. significant cardiopulmonary disease);
8. Participant has a history of immune-mediated or clinically relevant arthritis/arthralgia;
9. Participant has a history of malignancy;
10. Participant has a known or suspected defect of the immune system that can be expected to influence the immune response to the vaccine, such as Participants with congenital or acquired immunodeficiency, including infection with HIV, status post organ transplantation or immuno-suppressive therapy within 4 weeks prior to Visit 1. Immuno-suppressive therapy is defined as administration of chronic (longer than 14 days) prednisone or equivalent ≥0.05 mg/kg/day within 4 weeks prior to trial entry, radiation therapy or immunosuppressive cytotoxic drugs/ monoclonal antibodies in the previous 3 years; topical and inhaled steroids are allowed.
11. Participant has a history of any vaccine related contraindicating event (e.g., anaphylaxis, allergy to components of VLA1553 or the control vaccine, other known contraindications including febrile convulsions);
12. Participant presents with clinical conditions representing severe bleeding disorders and medications interfering with blood clotting;
13. Participant is pregnant (positive pregnancy test (in accordance with local regulations) at screening or Visit 1, respectively), lactating at the time of enrollment or unreliable contraception in female participants after onset of menarche and if involved in activities potentially leading to pregnancy;
14. Participant received blood-derived products (e.g. plasma) within 180 days prior to vaccination in this trial;
15. Participant has a rash, dermatological condition or tattoos that would, in the opinion of the Investigator, interfere with injection site reaction rating;
16. Participant has participated in another clinical trial involving an investigational medicinal product (IMP) or device within 30 days prior to vaccination or is scheduled to participate in another clinical trial involving an IMP, or device during the course of this trial;
17. Participant has any condition that, in the opinion of the Investigator, may compromise the participants well-being, might interfere with evaluation of trial endpoints, or would limit the participant's ability to complete the trial;

[18] Includes mRNA vaccines

18. Participant/partipant's parent(s)/LAR(s) is/are a member of the team conducting the trial or in a dependent relationship with one of the trial team members. Dependent relationships include close relatives (i.e., children, partner/spouse, siblings, parent(s)/LAR(s)s) as well as employees of the Investigator or site personnel conducting the trial.
19. Participant has received the control vaccine prior to the trial.

13.3 Delay Criteria

Vaccination will be delayed if:

1. Participant has an acute febrile infection within 72 hours prior to vaccination or temperature ≥38.0°C (100.4°F) on the day of vaccination (participant may be rescheduled within the screening visit window until participant has completed 72 hours without fever);
2. Participant has received antipyretics or analgesics within 24 hours prior to the scheduled time of vaccination (participant may be rescheduled within the screening visit window);
3. Participant has received any live or inactivated vaccine[19] within 28 days or 14 days prior to vaccination, respectively (participant may be rescheduled within the screening visit window).
4. IMP is unavailable at the site (e.g. logistics problems) or under quarantine.
   In addition, for a rescheduled vaccination all inclusion and none of the exclusion criteria must be met; in case not all of these criteria are met, the participant will be excluded from the trial.
   The rescheduled visit should be within the specified time window for the vaccination visit. In case the time window for the rescheduled visit cannot be met, the participant might be invited for a re-screening.
5. In case of no blood sample result available (e.g., out of stability samples) due to logistical problems at Screening (V0), and the permission of at least one parent/LAR to recollect the missing sample*
   In addition, for a rescheduled vaccination all inclusion and none of the exclusion criteria must be met; otherwise the participant will be excluded from the trial. The rescheduled visit should be within the specified time window for the vaccination visit. In case the time window for the rescheduled visit cannot be met, the participant might be invited for a re-screening.

* For recollection of a blood sample the permission of at least one parent/LAR has to be documented in the source documents and is only allowed once (if applicable in selected country)

13.4 Re-screening of Participants

A participant is eligible for re-screening if she or he completed the screening visit and met eligibility criteria but cannot be randomized or vaccinated for administrative or logistical reasons within the given time window.

13.5 Pregnancy Testing and Birth Control

The risk of maternal-to-fetal transmission of chikungunya vaccine virus during pregnancy cannot be excluded. Therefore, female participants of childbearing potential and if involved in activities that could result in pregnancy, presenting with a negative pregnancy test and practicing the use of adequate birth control before conduct and during the first three months of the trial are eligible for inclusion into the trial.

[19] Includes mRNA vaccines

A female participant is considered of childbearing potential after onset of menarche. Female participants of childbearing potential will be asked during the screening visit (Visit 0) if they are involved in activities that could lead to pregnancy. Birth control measures only have to be performed, if involved in activities that could result in pregnancy.

Participant of childbearing potential and if involved in activities that could result in pregnancy must have practiced an adequate contraceptive method during the 30 days before Visit 0 (Screening Visit) and the first three months until Visit 6 (Day 85/ Month 3) and present with a negative pregnancy test at Visit 0 (Screening Visit), a negative pregnancy test prior to vaccination.

Participants of childbearing potential and if involved in activities that could result in pregnancy are required to practice an acceptable method of birth control for the first three months post-vaccination. An acceptable method of birth control is defined as those, which result in a low failure rate (i.e., less than 1% per year) when used consistently and correctly.

This includes one of the following measures:

- Hormonal contraceptives (e.g. implants, birth control pills, patches);
- Intrauterine hormone-releasing system;
- Barrier type of birth control measure (e.g., condoms, diaphragms, cervical caps);
- Vasectomy in the male sex partner ≥ 3 months prior to first vaccination;
- Sexual abstinence;
- Same-sex relationships;

If a participant becomes pregnant during the trial, the parent(s)/Legally Authorized Representative(s) (LARs) must immediately inform the Investigator and the participant is asked to attend all remaining visits according to schedule.

13.6 Participant Withdrawal or Discontinuation

Any parent/LAR has the right to withdraw their child from the trial at any time for any reason, without the need to justify. The Investigator and Sponsor also have the right to prematurely terminate a participant's further participation in the trial, e. g. in the case of non-compliance or if – in the judgment of the Investigator and/or Sponsor – continued participation would pose an unacceptable risk for the participant.

The primary reason for withdrawal / discontinuation of a participant from the trial should be documented in the electronic Case Report Form (eCRF) (e.g. withdrawal of consent, Investigator/Sponsor recommended withdrawal, lost to follow-up, death).

The primary reasons for discontinuation will be reported on the Discontinuation eCRF, including:

- Withdrawal of consent (not due to AE);
- Withdrawal due to AE;
- Lost to follow-up (defined as 3 documented unsuccessful attempts to contact the participant´s parent(s)/LAR(s)[20]);
- Investigator decision (e.g. non-compliance with protocol);
- Trial terminated by Sponsor;
- Death;
- Or other (reason to be specified by the Investigator, e.g. technical problems).

Regardless of the reason, all data available for the participant up to the time of completion/discontinuation should be recorded on the appropriate CRF.

[20] Note that a participant who is lost to follow-up but later returns to the trial site for a follow-up can still complete the trial provided he/she has received the vaccination according to the protocol.

Data collected on withdrawn participants will be used in the analysis and included in the clinical trial report.

Participants who do not complete the entire trial due to withdrawal or discontinuation for any reason will not be replaced.

14. INVESTIGATIONAL MEDICINAL PRODUCT

In this trial the Sponsor's VLA1553 vaccine in two different doses or Nimenrix in the control group will be administered to trial participants.

14.1 Description of VLA1553

The CHIKV vaccine (VLA1553) is a live-attenuated vaccine comprising a large deletion in the nsP3 viral replicase complex, which leads to attenuation of the virus *in vivo*. VLA1553 is present in a freeze-dried presentation and must be reconstituted with a solvent consisting of sterile water for injection in a prefilled syringe before use.

The appearance of the reconstituted solution for administration is a clear pale to slightly yellow liquid solution.

The VLA1553 vaccine will be tested both as full dose formulation (4.0 log 10 $TCID_{50}$ per 0.5mL) and as a half dose formulation (3.7 log 10 $TCID_{50}$ per 0.5mL)

One full dose (0.5 mL) of the VLA1553 vaccine contains between 3.2 log 10 and 4.4 log 10 $TCID_{50}$ per dose (target dose 4.0 log 10 $TCID_{50}$ per 0.5 mL).

One half dose (0.5 mL) of the VLA1553 vaccine contains between 2.9 log 10 and 4.1 log 10 TCID50 per dose (target dose 3.7 log 10 TCID50 per 0.5 mL).

The active ingredient is suspended in a formulation buffer of pH 7.3, before freeze drying.

Table 8 shows details of the full dose Drug Product.

Table 8: Composition of the VLA1553 full dose lyophilized Drug Product

Active substance	
Live-attenuated CHIKV	**Target $TCID_{50}$/dose**
	4.0 log 10 $TCID_{50}$
Excipients and buffer components	**Freeze Dried Formulation/ dose**
di-Potassium Hydrogen Phosphate	0.313 mg
Potassium di-Hydrogen Phosphate	0.098 mg
Trisodium citrate dihydrate	3.68 mg
Sucrose	25 mg
Magnesium Chloride hexahydrate	0.51 mg
D-Sorbitol	2.5 mg
L-Methionine	0.75 mg
Recombinant Human Albumin	0.01% (equals 0.05mg)
pH	7.3

Table 9 shows details of the half dose Drug Product.

Table 9: Composition of the VLA1553 half dose lyophilized Drug Product

Active substance	
Live-attenuated CHIKV	**Target $TCID_{50}$/dose**
	3.7 log 10 TCID50
Excipients and buffer components	**Freeze Dried Formulation/ dose**
di-Potassium Hydrogen Phosphate	0.157 mg
Potassium di-Hydrogen Phosphate	0.048 mg
Trisodium citrate dihydrate	1.84 mg
Sucrose	12.5 mg
Magnesium Chloride hexahydrate	0.255 mg
D-Sorbitol	1.25 mg
L-Methionine	0.375 mg
Recombinant Human Albumin	0.005% (equals 0.025mg)
pH	7.3

14.2 Description of Nimenrix

Nimenrix is a conjugate vaccine indicated for the active immunization of individuals from the age of 6 weeks against invasive meningococcal disease caused by Neisseria meningitidis groups A, C, W-135, and Y.

Nimenrix is present as powder and solvent for solution for injection in pre-filled syringe.

The reconstituted vaccine is a clear colourless solution.

Nimenrix should be used in accordance with available official recommendations. Primary immunization in infants from 6 months of age, children, adolescents and adults: a single 0.5 mL dose should be administered.

Table 10: Composition of Nimenrix

	Nimenrix	
Formulation:	MenA (5 µg)-TT MenC(5 µg)-TT MenW135(5 µg)-TT MenY(5 µg)-TT	Sodium chloride; Water for injections q.s. 0.5 mL
Presentation:	Powder for solution for injection (vial)	Solution for solution for injection
Type:	Biologic/Combination Product	
Route of administration:	Intramuscular injection (i.m.)	
Location	Upper arm for Stratum A and B; thigh for Stratum C	
Directionality	Anterolateral	
Number of doses to be administered:	1	
Volume to be administered:	0.5 mL	
Packaging and labeling	Refer to IMP manual for more details	

14.3 IMP Packaging

14.3.1 VLA1553 Packaging

The full dose VLA1553 will be provided in kit containing one single-use 2R vial with the lyophilized powder of VLA1553 vaccine, one solvent consisting of 0.5 mL sterile water for injection in a prefilled syringe and a sterile needle set. One (1x 25G x 1,5” 0.50x40 mm) for reconstitution and one (1x 25G x 1”, 0.50x25 mm) for administration.

The half dose VLA1553 will be provided in kit containing one single-use 2R vial with the lyophilized powder of VLA1553 vaccine, one solvent consisting of 0.5 mL sterile water for injection in a prefilled syringe and a sterile needle set. One (1x 25G x 1,5” 0.50x40 mm) for reconstitution and one (1x 25G x 1”, 0.50x25 mm) for administration.

14.3.2 Nimenrix Packaging

Powder and solvent for solution for injection in pre-filled syringe.

14.4 IMP Labelling

Labeling of VLA1553 and Nimenrix are performed by a qualified CMO. The IMP will be labeled according to the valid regulatory requirements for clinical trials.

14.5 IMP Storage

14.5.1 VLA1553 Storage

VLA1553 has an expected retest date of 24 months from the date of manufacture, considering the specified storage conditions of +2-8°C (35.6 °F to +46.4 °F). After reconstitution, the vaccine should be used promptly. If not used within 2 hours, do not administer the vaccine. A stability program is currently ongoing and if any impact on the stability profile is observed, the trial sites will be informed accordingly. The vaccine must not be used after the retest or expiry date indicated on the package. VLA1553 must be stored in a refrigerator in a room not accessible to unauthorized persons. Storage at room temperature or higher should be avoided because of potential impairment to immunogenicity and tolerability. Temperature monitoring systems will be used.

14.5.2 Nimenrix Storage

Nimenrix must be stored in a refrigerator at +2-8°C (35.6 °F to +46.4 °F) in a room not accessible to unauthorized persons. After reconstitution, the vaccine should be used promptly. If not used within 2 hours, do not administer the vaccine. The vaccine must not be used after the end of shelf-life retest date indicated on the package. Storage at room temperature or higher should be avoided because of potential impairment to immunogenicity and tolerability. Temperature monitoring systems will be used.

14.6 Dispensing and Accountability of IMP

Drug accountability logs have to be maintained current, detailing the dates and quantities of IMP administered to each participant. Records will be maintained that include participant identification code (PIC), administration date, and amount administered. This documentation will be available to the designated unblinded CRA to verify drug accountability during the trial and to perform overall drug accountability.

Used and unused IMP will be accounted for and returned to the CMO/ Sponsor. In addition, used IMP can be destroyed on-site upon Sponsor approval.

A trial specific IMP manual with further details on IMP handling, administration, return/destruction and documentation will be provided.

15. TRIAL PROCEDURES

15.1 Informed Consent/ Assent and Enrollment

Any volunteer child for who their parent(s)/LAR(s) provides informed consent (i.e., signs and dates the informed consent form) and assent from the participant if applicable is considered enrolled in the trial.

For further details refer to **Section 19.3**.

15.2 Participant Identification Code

The following format of the Participant Identification Code is agreed within the VLA1553 trial program by Valneva: At Visit 0, a 13-character participant identification code shall be assigned to each participant. The first four digits are the product identifier (e.g., 1553 for this product) provided by Valneva. The fifth digit is the trial identifier (i.e., 1553-2 for this Phase 2 trial), the sixth and seventh digits are site identification number (i.e. 1553-2-01). The last three digits are assigned in ascending order as the participants are enrolled (i.e., signing the informed consent form, e.g. 1553-2-01-001).

15.3 Investigational Treatment

15.3.1 Description of Treatment

All of the participants will receive a single intramuscular vaccination - in the deltoid region of the arm for Stratum A and B or in the anterolateral thigh for Stratum C - of VLA1553 or Nimenrix according to the vaccination schedule as described in **Section 24** of Trial Procedures. Participants will be followed up for approximately 12 months following the vaccination (see also **Section 12.1**).

15.3.2 Vaccine Preparation and Administration

VLA1553 Full dose

VLA1553 is available as a suspension after reconstitution of targeted 4.0 log 10 $TCID_{50}$ per 0.5 mL dose.

IMP preparation and administration will be done according to the procedure outlined in the trial specific IMP manual.

VLA1553 Half dose

Half dose of VLA1553 is available as a suspension after reconstitution of targeted 3.7 log 10 $TCID_{50}$ per 0.5 mL dose.

IMP preparation and administration will be done according to the procedure outlined in the trial specific IMP manual.

Under no circumstances should the VLA1553 vaccine be administered intravascularly, as this could lead to hypersensitivity reactions such as shock.

Nimenrix

Instructions for reconstitution and administration will be done according to the procedure outlined in the trial specific IMP manual.

Anaphylaxis or other possible severe acute, post-vaccination adverse reactions to vaccines, including VLA1553 and Nimenrix vaccines, are very rare, but can occur. Therefore, appropriate emergency equipment and medication as well as adequately trained personnel must be on site whenever a vaccination is performed and during the observation period.

15.3.3 Post-Vaccination Observation

Following vaccination, the participant will be observed for at least 30 minutes (at least 1 hour for sentinel participants of Cohort I, II, III and IV) at the trial site in order to provide appropriate emergency treatment should this be necessary. In addition, vital signs including pulse rate and blood pressure while seated and at rest will be measured prior to discharge (for further information see **Section 17.6**). Any injection site and systemic reactions will be recorded.

Prior to leaving the trial site, the parent(s)/LAR(s) will be given the respective Participant Diary for documentation of AEs (for further information see **Section 17.3**), an infrared thermometer for measuring body temperature and a measuring device for assessing injection site reactions (at the vaccination visit only).

15.4 Screening and Trial Visits

The overall trial design is illustrated in

Figure 1 (in **Section 12.1**). For a tabular outline of trial procedures and assessments required at each visit, please see **Table 11**, **Table 12** and **Table 13** as well as the Schedule of Trial Procedures and Assessments in **Table 5**.

15.4.1 Part A

Table 11: Trial VLA1553-221 Trial Visit Schedule – Part A

VISIT	TIME	ACTION
Visit 0 Screening	**Day -28 to 0 (prior to Visit 1)**	1. Informed consent [a] 2. Inclusion and exclusion criteria (**Section 13.1 and 13.2**) 3. Demographics [b] 4. Medical history incl. vaccination history [c] (**Section 17.5.1**) 5. Prior and concomitant Medications (**Section 17.5.2**) 6. Physical examination [d] (**Section 17.7**) 7. Vital Signs [e] (**Section 17.6**) 8. Blood draw and urine sample for: a) CHIKV Screening b) Pregnancy test (if applicable)[f] c) Safety [h]
Visit 1[j]	**Day 1**	1. Inclusion and exclusion criteria (review) (**Section 13.1 and 13.2**) 2. Medical History incl. vaccination history (update) [c] (**Section 17.5.1**) 3. Concomitant Medication (**Section 17.5.2**) 4. Physical examination [d] (**Section 17.7**) 5. Vital Signs [e] (**Section 17.6**) 6. Blood draw and urine sample for: a) Pregnancy test (if applicable)[f] b) Immunogenicity [g] c) Viremia [i] d) Urinalysis [l] 7. Randomization 8. **VACCINATION** (**Section 15.3.2**) 9. Post-vaccination observation (**Section 15.3.3**) 10. AE documentation (**Section 17.2**) 11. Distribute Participant Diary (**Section 17.3**) 12. Distribute Safety Card (**Section 17.4**)
Visit 2	**Day 4 (+/- 1d)**	1. Review/Collect/Distribute Participant Diary (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**) 4. Physical examination [d] (**Section 17.7**) 5. Blood draw and urine sample for: a) Urinalysis [l] b) Viremia [i]

VISIT	TIME	ACTION
Visit 3	**Day 8 (+/- 1d)**	1. Review/Collect/Distribute Participant Diary (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**) 4. Physical examination [d] (**Section 17.7**) 5. Blood draw and urine sample for: a) Urinalysis [l] b) Viremia [i]
Visit 4	**Day 15 (+/- 1d)**	1. Review/Collect Participant Diary (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**) 4. Physical examination [d] (**Section 17.7**) 5. Blood draw and urine sample for: a) Immunogenicity [g] b) Urinalysis [l] c) Viremia[i] 6. Distribute Memory Aid (**Section 17.3**)
Visit 5	**Day 29 Month 1 (+/- 4d)**	1. Review/Collect Participant Memory Aid (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**) 4. Physical examination [d] (**Section 17.7**) 5. Blood draw and urine sample for: a) Immunogenicity [g] b) Safety [h] c) Urinalysis [l] d) Viremia [k] 6. Distribute Memory Aid (**Section 17.3**)

[a] Occurs at enrollment before Screening. Note: SAEs are recorded from ICF signature onwards.
[b] Demographics include date of birth, age, height, weight, BMI, gender, race and ethnicity.
[c] Symptoms noted (except those qualifying as a SAE) at Visit 1 (prior to first vaccination) are not considered AEs, but will be recorded as medical history. Any prior vaccination should be documented in the Medical History.
[d] A physical examination will be performed on the following body systems being described as normal or abnormal: general appearance, head and neck, eyes and ears, nose and throat, chest, lungs, heart, abdomen, extremities and joints, lymph nodes, skin, and neurological.
[e] Vital signs will include systolic and diastolic blood pressure and pulse rate while seated and at rest, and body temperature measured with infrared thermometer targeting the forehead before vaccination. In addition, after an observation period of 60 minutes (open label phase) or 30 minutes (blinded phase) following vaccination, pulse rate and blood pressure while seated and at rest will again be assessed.
[f] A pregnancy test (in accordance with local regulations) will be performed for all female Participants after onset of menarche and if involved in activities that could result in pregnancy at the screening visit and at indicated visits (i.e., V1).
[g] Immunogenicity sample [blood: 2 mL] for CHIKV-specific neutralizing antibody titer evaluation.
[h] Safety laboratory sample for hematology and coagulation panel [blood: 4 mL]. Safety sample will be taken from all participants at the screening visit; at subsequent visits (i.e. V5 or ET if earlier than V5) the safety sample will be only taken from the sentinel participants. If deemed necessary by the investigator a safety sample can be taken any time if safety concerns arise.
[l] Viremia sample for investigation of viremia by RT-qPCR [plasma: 2 mL]. Viremia sample will be taken from all participants of Stratum A and only taken from sentinel participants from Stratum B and C.
[j] All procedures/assessments (apart from Participant Diary distribution and AE assessment) occur prior to vaccination (unless stated otherwise).
[k] Visit 5 (Day 29) will be only tested if Visit 4 (Day 15) tests positive.
[l] Urinalysis (i.e. pH, specific gravity, leucocytes, nitrite, protein, glucose, ketones, urobilinogen, bilirubin, erythrocytes)- if possible.
[m] Samples should be obtained for clinically indicated retrospective investigation of viremia by RT-qPCR.
[n] Safety sample should be collected if ET is before Day 29 (Visit 5)- sentinels only.

15.4.2 Part B

Table 12: Trial VLA1553-221 Trial Visit Schedule – Part B

VISIT	TIME	ACTION
Visit 6	**Day 85 Month 3 (+/- 7d)**	1. Review/Collect/Distribute Memory Aid (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**)

		4. Physical examination [a] (**Section 17.7**) 5. Blood draw and urine sample for: a) Immunogenicity sample [b] b) Urinalysis [c] c) Viremia sample [d/e]
Visit 7	**Day 180 Month 6 (+/- 14d)**	1. Review/Collect/Distribute Memory Aid (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**) 4. Physical examination [a] (**Section 17.7**) 5. Blood draw and urine sample for: a) Immunogenicity sample [b] b) Urinalysis [c] c) Viremia sample [d/e]

[a] A physical examination will be performed on the following body systems being described as normal or abnormal: general appearance, head and neck, eyes and ears, nose and throat, chest, lungs, heart, abdomen, extremities and joints, lymph nodes, skin, and neurological
[b] Immunogenicity sample [blood: 2 mL] for CHIKV-specific neutralizing antibody titer evaluation.
[c] Urinalysis (i.e., pH, specific gravity, leucocytes, nitrite, protein, glucose, ketones, urobilinogen, bilirubin, erythrocytes)- if possible.
[d] Viremia sample for investigation of viremia by RT-qPCR [plasma: 2 mL]. Viremia sample will be taken from all participants of Stratum A and only taken from sentinel participants from Stratum B and C.
[e] Samples should be obtained for clinically indicated retrospective investigation of viremia by RT-qPCR.

15.4.3 Part C

Table 13: Trial VLA1553-221 Trial Visit Schedule – Part C

VISIT	TIME	ACTION
Visit 8	**Day 365 Month 12 (+/- 14d)**	1. Review/Collect Memory Aid (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**) 4. Physical examination [a] (**Section 17.7**) 5. Blood draw and urine sample for: a) Immunogenicity sample [b] b) Urinalysis [c] c) Viremia sample [d/e/] [f]

[a] A physical examination will be performed on the following body systems being described as normal or abnormal: general appearance, head and neck, eyes and ears, nose and throat, chest, lungs, heart, abdomen, extremities and joints, lymph nodes, skin, and neurological
[b] Immunogenicity sample [blood: 2 mL] for CHIKV-specific neutralizing antibody titer evaluation.
[c] Urinalysis (i.e. pH, specific gravity, leucocytes, nitrite, protein, glucose, ketones, urobilinogen, bilirubin, erythrocytes)- if possible.
[d] Viremia sample for investigation of viremia by RT-qPCR [plasma: 2 mL]. Viremia sample will be taken from all participants of Stratum A and only taken from sentinel participants from Stratum B and C.
[e] Samples should be obtained for clinically indicated retrospective investigation of viremia by RT-qPCR.

15.4.4 Unscheduled Visit

An unscheduled visit can be held at any time during the trial if deemed necessary by the Investigator (e.g., follow-up on unexpected AEs or SAEs) or the DSMB. Assessments performed at an unscheduled visit will be at the Investigator's or DSMB's discretion. Unscheduled visits and any procedures/assessments performed during such a visit (e.g., physical examination, laboratory test) should be documented in the source data and the eCRF.

15.4.5 Early Termination Visit

Participants for who their parent(s)/LAR(s) terminate participation or who are withdrawn from the trial prematurely will undergo investigations as outlined below during an Early Termination Visit, if possible. Every effort should be made to have discontinued participants complete the trial Early Termination (ET) Visit (see **Table 14**).

Table 14: Trial VLA1553-221 Trial Visit Schedule – Early Termination

VISIT	TIME	ACTION
ET	**unscheduled**	1. Review/Collect Participant Diary/Memory Aid (**Section 17.3**) 2. AE documentation (**Section 17.2**) 3. Concomitant medication (**Section 17.5.2**) 4. Physical examination [a] (**Section 17.7**) 5. Blood draw and urine sample for: a) Immunogenicity sample [b] b) Safety sample [c/d] c) Viremia sample [e/f] d) Urinalysis [g] 6. Documentation of reason(s) for early termination

[a] A physical examination will be performed on the following body systems being described as normal or abnormal: general appearance, head and neck, eyes and ears, nose and throat, chest, lungs, heart, abdomen, extremities and joints, lymph nodes, skin, and neurological
[b] Immunogenicity sample [blood: 2 mL] for CHIKV-specific neutralizing antibody titer evaluation.
[c] Safety laboratory sample for hematology and coagulation panel [blood: 4 mL]. Safety sample will be taken from all participants at the screening visit; at subsequent visits the safety sample will be only taken from the sentinel participants. If deemed necessary by the investigator a safety sample can be taken any time if safety concerns arise.
[d] Safety sample should be collected if ET is before Day 29 (Visit 5).
[e] Viremia sample for investigation of viremia by RT-qPCR [plasma: 2 mL]. Viremia sample will be taken from all participants of Stratum A and only taken from sentinel participants from Stratum B and C.
[f] Samples should be obtained for clinically indicated retrospective investigation of viremia by RT-qPCR
[g] Urinalysis (i.e. pH, specific gravity, leucocytes, nitrite, protein, glucose, ketones, urobilinogen, bilirubin, erythrocytes)- if possible.

In case an ET Visit is not possible, a follow-up safety phone call should be made as soon as possible after termination to capture at least concomitant medications and solicited and unsolicited AEs since the last trial visit.

The reason for early termination should be clarified in as much detail as possible. If an AE was the reason for early trial termination details on that specific AE(s) should be captured (see **Section 13.6**).The reason for discontinuation will be recorded on the eCRF, and data collected up to the time of discontinuation will be used in the analysis and included in the clinical trial report.

In the event of participant discontinuation due to an AE, clinical and/or laboratory investigations that are beyond the scope of the required trial observations/assessments may be performed as part of the evaluation of the event. These investigations will take place under the direction of the Investigator in consultation with the Sponsor, and the details of the outcome may be reported to the appropriate regulatory authorities by the Sponsor.

16. ASSESSMENT OF IMMUNOGENICITY

All serum samples for neutralization titer determination will be handled according to the procedures supplied to each investigative site for the preparation, storage and shipment of samples (refer to Laboratory Manual). Each clinical trial site will be responsible for the separation of serum from whole blood samples and the safe and controlled storage of serum samples prior to shipment to the laboratory.

Immunogenicity samples (using CHIKV µPRNT) will be collected from all participants at the indicated time points and may be used for additional testing procedures (see **Section 16.2**). However, immunogenicity assessment measuring neutralizing antibodies will be performed on samples collected at Day 1, 15, 29, 85, 180 and 365 after a single immunization.

16.1 Determination of vaccine-induced neutralizing antibody response

Immunogenicity of VLA1553 will be evaluated using a micro–Plaque Reduction Neutralization Test (µPRNT), which is based on the same principle as a plaque reduction neutralization assay (PRNT), but allows testing with higher throughput. This assay differs from the µNT assay used for the assessment of neutralizing antibodies during Phase 1 clinical testing. Chikungunya virus neutralizing antibodies will be measured in the µPRNT using a serially passaged, live-attenuated CHIK vaccine virus strain (CHIKV 181/25, TSI-GSD-218 or 181/clone 25) developed by the Walter Reed Army Institute of Research (WRAIR, US).

The CHIKV µPRNT evaluates the levels of antibodies that neutralize CHIKV infection of Vero cells in human serum samples.

[redacted]

The cut-off of CHIKV-specific neutralization antibody titer to be used in the analysis of seroconversion has been defined during µPRNT assay validation as a >4-fold increase of $\mu PRNT_{50}$ compared to baseline (at Day 1). CHIKV-specific neutralizing antibody titer of $\mu PRNT_{50} \leq 40$ is regarded as seronegative.

16.2 Additional Testing Procedures

Samples obtained in this trial may, in addition to its use for assessment of CHIKV-specific neutralizing antibody titers, also be used for further development of the vaccine, including but not limited to the following assays:

- Passive immunization assays to evaluate the potency of immune sera to protect animals from infection after wild type CHIKV challenge;
- Development of additional neutralization assays (e.g,. µNT, PRNT, virus replicon particle neutralization assays) for the assessment of cross-neutralization of heterologous CHIKV strains or other related (alpha) viruses;

- Detection of anti-CHIKV antibodies
- Detection of viremia by viral culture;
- CHIKV sequencing;
- Clinical diagnostic work-up.

Such development may occur at laboratories other than the analytical laboratories used for this trial.

17. ASSESSMENT OF SAFETY

17.1 Definitions

17.1.1 Adverse Events

An adverse event (AE) is defined as any untoward medical occurrence in a participant administered an investigational product that does not necessarily have a causal relationship with the treatment. All new abnormalities or any exacerbation in intensity or frequency (worsening) of a pre-existing condition during or after vaccination have to be documented as AEs. If a Serious Adverse Event occurs from the time of ICF signature to vaccination, it will be documented as such.

Clinical Symptoms that may be indicative for a clinically significant condition may be subject to a medical work up at the discretion of the investigator which may include a referral to a specialist. The concerned treatment should be performed according to current medical standard of care.

17.1.2 Serious Adverse Event

A **serious** adverse event (SAE) is defined as any untoward medical occurrence that at any dose meets one or more of the following criteria:

- Outcome is fatal/results in death (including fetal death);
- Is life-threatening – defined as an event in which the participant was, in the judgment of the Investigator, at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death had it been more severe;
- Requires inpatient hospitalization or results in prolongation of an existing hospitalization;
- Results in persistent or significant disability/incapacity (i.e., a substantial disruption of a person's ability to conduct normal life functions);
- Results in congenital anomaly/birth defect;
- Is a medically important condition – a medical event that may not be immediately life-threatening or result in death or require hospitalization but may jeopardize the participant or may require medical or surgical intervention to prevent one of the other outcomes listed in the definitions above. This definition also applies to progression of disease leading to a serious outcome.

SAEs will be recorded from signed ICF onwards.

In case of hospitalization or prolonged hospitalization for diagnostic or elective medical procedures that were planned prior to vaccination to treat a pre-existing condition that did not change in severity, neither the condition leading to the hospitalization or prolonged hospitalization, nor the medical procedure itself need to be reported as an SAE. In this case, the underlying diagnosis or condition should be reported in the medical history section of the eCRF and the corresponding medical procedure should be documented as a comment to the underlying diagnosis or condition in the eCRF's medical history section.

The Sponsor's Clinical Safety Physician will classify the SAEs as either expected or unexpected:

- **Expected:** An AE that is listed in the current Investigator's Brochure (IB);
- **Unexpected:** An AE that is not listed in the current IB, or it differs because of greater severity or greater specificity.

Note: AEs graded as potentially life-threatening (Grade 4) (see Section 17.2.6 for AEs) need to be assessed by the investigator if a seriousness criterion is met and may require expedited reporting.

17.1.3 Adverse Events of Special Interest

An AESI (serious or non-serious) is an event of scientific and medical concern specific to the Sponsor's product, please refer to **Section 17.10** for further details.

17.1.4 Medically-Attended Adverse Event

All AEs where parent(s)/LAR(s) is/are seeking medical care (i.e. doctor's office, emergency service, hospital, but not including use of self-medication) for the participants.

17.1.5 Preexisting Diseases

Preexisting diseases that are present before entry into the trial, that are described in the medical history, and that manifest with the same severity, frequency, or duration after vaccine exposure, will not be recorded as AEs. Furthermore, routine health checks required due to pre-existing diseases will not be recorded. However, when there is an increase in the severity of a preexisting disease, the event must be reported on the AE CRF page.

17.2 Collection, Documentation and Assessment of Adverse Events

17.2.1 Collection Period for Adverse Events

Untoward medical occurrences (Adverse Events) experienced before vaccine exposure (i.e., from the time of signed informed consent up to but not including vaccine exposure) will be described in the medical history.

However, SAEs will be recorded from signed ICF onwards.

The following information will be documented for each AE: severity, causality, outcome, seriousness, medically-attended, action taken to treat AE, start and stop dates.

17.2.2 Severity

The investigator will assess the severity of AEs using his/her clinical expertise and judgment based on the most appropriate description below:

Mild (Grade 1):	Awareness of signs or symptoms, but easily tolerated, does not interfere with daily activities.
Moderate (Grade 2):	Discomfort enough to interfere with usual activity.
Severe (Grade 3):	Incapable of usual activity and requiring medical intervention. Such an AE would, for example, prevent attendance at a childcare facility and would cause the parent(s)/LAR(s) to seek medical advice.

If applicable (also refer to section 17.2.6 and section 21.2.2):

Potentially Life-Threatening (Grade 4):	Potentially life threatening or disabling (e.g., complicated by acute, life- threatening metabolic or cardiovascular complications such as circulatory failure, hemorrhage, sepsis; life–threatening physiologic consequences; need for intensive care or emergent invasive procedure; emergent interventional radiological procedure, therapeutic endoscopy or operation).

17.2.3 Causality

Causality is a determination of whether there is a reasonable possibility that the vaccine administration is etiologically related to/associated with the AE.

For AEs, the Investigator will assess the causal relationship between the IMP and the AE using his/her clinical expertise and judgement according to the following most appropriate algorithm for the circumstances of the AE:

Probable: Reaction that follows a reasonable temporal sequence from administration of the IMP, or that follows a known or expected response pattern to the suspected treatment; and that could not reasonably be explained by known characteristics of the participant's clinical state.

Possible: Reaction that follows a reasonable temporal sequence from administration of the IMP, or that follows a known or expected response pattern to the suspected treatment; but that could readily have been produced by a number of other factors.

Unlikely: Reports not following a reasonable temporal sequence from administration of the IMP; an event, which may have been produced by the participant's clinical state or by other environmental factors. A more likely alternative etiology exists.

Not related (unrelated): Events for which sufficient information exists to conclude that the etiology is unrelated to the IMP.

AEs with a causality reported as probable or possible will be considered related to the IMP. AEs with missing causality assessment will be regarded as related unless further specified. All other AEs will be considered as not related to IMP.

17.2.4 Assessment and Outcome of Adverse Events

AEs will be recorded in the eCRF (i.e., 1 AE per form) using the medical diagnosis (preferred), symptom, or sign, in standard medical terminology in order to avoid the use of vague, ambiguous, or colloquial expressions (see definition in **Section 17.1.1**). AEs will be evaluated by the Investigator for:

- Seriousness as defined in **Section 17.1.2**
- Severity as defined in **Section 17.2.2**
- Causal relationship to vaccine exposure as defined in **Section 17.2.3**

For each AE, the outcome will also be documented as either:

- recovering/resolving
- recovered/resolved with sequelae
- fatal
- recovered/resolved
- not recovered/not resolved
- unknown

If the severity rating for an ongoing AE changes before the event resolves, the AE will not be reported a second time. Instead, the original AE report will be revised. For purposes of data capture the highest severity rating during the course of a single AE will be the severity rating entered on the AE CRF. Any symptom is regarded as a separate AE. Please note that (possibly or probably) related solicited AEs should be documented as individual events even though they have the same origin, namely the trial vaccine. However, in the case of an underlying disease such as Influenza: involving a cluster of different events of recurrent nature and which are clearly linked to the same condition, (based on the PIs judgement) may be documented as one event with a long(er) duration and highest severity. The Investigator will follow-up on each AE until it is resolved or until the medical condition of the participant is stable. All relevant follow-up information will be reported to the Sponsor until the end of the trial for each

participant. SAEs ongoing at the time of Visit 8 will be followed until resolution or achievement of stable clinical conditions, latest until the overall end of the trial.

NOTE: *A participant's death per se is not an event, but an outcome. The event which resulted in the participant's death must be fully documented and reported, regardless of being considered related to treatment or not.*

17.2.5 Unsolicited Adverse Events (post-vaccination)

Parents/LARs will be provided with a Participant Diary up to Visit 4 and a Memory Aid from onward visits to be able to collect unsolicited AEs occurring until Day 365 (Visit 8, see **Section 17.3**). Additionally, the Investigator will enquire about AEs during trial visits. Clinically relevant laboratory parameter changes constitute unsolicited AEs, too, unless they are considered a symptom of an underlying AE or part of a syndrome that is reported as AE (e.g. presence of blood cells in urine in a person diagnosed with urinary tract infection). In addition, symptoms noted during physical examinations (unless already covered by an AE) constitute AEs.

All unsolicited AEs need to be documented in the respective AE section of the eCRF during applicable trial visit (Visits 1 to 8, or unscheduled visit(s), if applicable), regardless of their source (AEs noted in the Memory Aid (see **Section 17.3**), open question to participant, laboratory parameters, physical examination). SAEs will continue to be documented until the end of the trial (Visit 8, Day 365).

Any symptom is regarded as a separate AE. However, if the Investigator considers several symptoms to be in the context of one underlying diagnosis, the Investigator may merge these symptoms into one single appropriate AE. The AE term entered into the eCRF should contain all symptoms summarized to one event (e.g. “Influenza with flu-like-symptoms, fever and headache”).

The Investigator will follow-up on each AE until it is resolved or until the medical condition of the participant is stable. All relevant follow-up information will be reported to the Sponsor until the end of the trial for each participant. SAEs ongoing at the time of Visit 8 will be followed until resolution or achievement of stable clinical conditions, latest until the overall end of the trial.

The following information will be documented for each AE: severity, causality, outcome, seriousness, medically-attended, action taken to treat AE, start and stop dates.

17.2.6 Solicited Adverse Events

The list of Solicited AEs collected in this trial is inclusive for both the treatment arm and control arm (Nimenrix) and is based upon the available clinical data for VLA1553, the prescribing information for Nimenrix and general age considerations taking into account the Strata (Stratum A – 7 to 11 years, Stratum B – 3 to 6 years, Stratum C – 12 to 35 months)

17.2.6.1 Injection Site Reaction – Measurement and Evaluation

Parents/LARs will be provided with a measuring device to measure the size of any measurable injection site reaction that may develop after vaccination. The parent(s)/LAR(s) will be instructed on how to measure any such reactions over a period of 14 consecutive days (until Day 15) after vaccination along the longest diameter of the reaction area and record this measurement in the participant diary (see **Section 17.3**). Injection site reactions include injection site pain, tenderness, erythema/redness, induration/swelling (see **Table 2**). Severity of injection site reactions will be rated as described in **Table 2** by the investigator.
Any injection site reaction meeting the grade 4 criterion will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

Table 2: Injections Site Reactions 1 – 11 years of age

Vaccine specific Criteria	Grade 1 (Mild)	Grade 2 (Moderate)	Grade 3 (Severe)	Grade 4 [4] (Potentially Life-Threatening)
Injection Site Pain [A]	Does not interfere with activity	Repeated use of non-narcotic pain reliever >24 hours or interferes with activity	Any use of narcotic pain reliever or prevents daily activity	Emergency room (ER) visit or hospitalization
Tenderness [A]	Mild discomfort to touch	Discomfort with movement	Significant discomfort at rest	ER visit or hospitalization
Injection Site Erythema or Redness [B,1]	1.0 [2] - ≤2.5 cm in diameter	>2.5 - 5.0 cm [3] in diameter with <50% surface area of the extremity segment involved (e.g., upper arm or thigh)	>5 cm [3] OR ≥50% surface area of the extremity segment involved (e.g., upper arm or thigh) OR Ulceration OR Secondary infection OR Phlebitis OR Sterile abscess OR Drainage	Potentially life-threatening consequences (e.g. Skin necrosis)
Injection Site Induration/ Swelling [B]	1.0 [2] - ≤ 2.5 cm in diameter	>2.5 - 5.0 cm [3] in diameter with <50% surface area of the extremity segment involved (e.g., upper arm or thigh)	>5 cm [3] OR ≥ 50% surface area of the extremity segment involved (e.g., upper arm or thigh) OR Ulceration OR Secondary infection OR Phlebitis	Potentially life-threatening consequences (e.g. Skin necrosis)

[A)] Grading based on FDA Guidance on Toxicity Grading Scales Adults/ Adolescents.
[B)] Grading based on Division of AIDS, Table for Grading the Severity of Adult and Pediatric Adverse Events, corrected version 2.1, July 2017.
[1)] Injection Site Erythema or Redness should be evaluated and graded using the greatest single diameter or measured surface area.
[2)] Reactions less than 1cm are not considered AEs.
[3)] Modified grading based on Division of AIDS; 5.0 cm cut-off will be included as moderate instead of severe.
[4)] Any injection site reaction meeting the grade 4 criteria will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting

17.2.6.2 Systemic Reactions – Measurement and Evaluation

The list of collected systemic reactions for the individual participant will depend on which Stratum he/she belongs to.

Stratum A (7 to 11 years) and B (3 to 6 years)

Systemic reactions for Stratum A and B including nausea/vomiting, headache, fatigue, myalgia (muscle pain), arthralgia (joint pain) and rash will be reported in a standardized manner over a period of 14 consecutive days (until Day 15) after vaccination.
Systemic reactions will be rated as described in **Table 3** by the investigator.
Any systemic site reaction meeting the grade 4 criterion will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

Table 3: Grading of Systemic Reactions Stratum A and Stratum B

Vaccine specific Criteria	Grade 1 mild	Grade 2 moderate	Grade 3 severe	Grade 4 [1] potentially life-threatening
Nausea/ Vomiting [A]	No interference with activity or 1 – 2 episodes/24 hours	Some interference with activity or >2 episodes/24 hours	Prevents daily activity, requires outpatient IV hydration	ER visit or hospitalization for hypotensive shock
Headache [A]	No interference with activity	Repeated use of non-narcotic pain reliever >24 hours or some interference with activity	Significant; any use of narcotic pain reliever or prevents daily activity	ER visit or hospitalization
Fatigue [A]	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit orhospitalization
Myalgia [A]	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalization
Arthralgia [A]	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalization
Rash [B]	Macules/papules covering <10% body surface area (BSA) with or without symptoms (e.g., pruritus, burning, tightness)	Macules/papules covering 10- 30% BSA with or withoutsymptom (e.g., pruritus, burning, tightness); limiting instrumental activity of daily living	Macules/papules covering >30% BSA with or without associated symptoms; limitingself-care activity of daily living	NA
Fever [C]	38.0 to < 38.6°C or 100.4 to < 101.5°F	≥ 38.6 to < 39.3°C or ≥ 101.5 to < 102.7°F	≥ 39.3 to < 40.0°C or ≥ 102.7 to < 104.0°F	40.0°C or ≥ 104.0°F

[A)] Grading based on FDA Guidance on Toxicity Grading Scales Adults/ Adolescents.
[B)] Grading based on Common Terminology Criteria for Adverse Events (CTCAE), NIH, v4.03, 2010.
[C)] Grading based on Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events Corrected Version 2.1 July 2017
[1)] Any systemic reaction meeting the grade 4 criterion will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

Stratum C (1 to 2 years)

Systemic reactions for Stratum C including nausea/vomiting, inconsolable or excessive crying, disturbed sleep, loss of appetite, drowsiness and rash will be reported in a standardized manner over a period of 14 consecutive days (until Day 15) after vaccination.
Any systemic site reaction meeting the grade 4 criterion will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

Table 4: Grading of Systemic Reactions Stratum C

Vaccine specific Criteria	Grade 1 mild	Grade 2 moderate	Grade 3 severe	Grade 4 [3] potentially life-threatening
Nausea/Vomiting [A]	No interference with activity or 1 – 2 episodes/24 hours	Some interference with activity or >2 episodes/24 hours	Prevents daily activity, requires outpatient IV hydration	ER visit or hospitalization for hypotensive shock
Inconsolable or excessive crying [B]	presence of crying [1] which is continuous [2] AND unaltered for ≥3h.	presence of crying [1] which is continuous [2] AND likely to be unaltered for >3h OR unaltered for >3h AND likely to be continuous.	NA	NA
Disturbed sleep [C]	Mild difficulty falling asleep, staying asleep, or waking up early causing no or minimal interference with usual social & functional activities	Moderate difficulty falling asleep, staying asleep, or waking up early causing more than minimal interference with usual social & functional activities	Severe difficulty falling asleep, staying asleep, or waking up early causing inability to perform usual social & functional activities requiring intervention or hospitalization	NA
Loss of appetite [D]	Loss of appetite without alteration in eating habits	Oral intake altered without significant weight loss or malnutrition; oral nutritional supplements indicated	Associated with significant weight loss or malnutrition (e.g., inadequate oral caloric and/or fluid intake); tube feeding or TPN indicated	Life-threatening consequences; urgent intervention indicated
Drowsiness [D]	Mild but more than usual drowsiness or sleepiness	Moderate sedation; limiting instrumental activity of daily living	Obtundation or stupor	Life-threatening consequences; urgent intervention indicated
Rash [E]	Macules/papules covering <10% body surface area (BSA) with or without symptoms (e.g., pruritus, burning, tightness)	Macules/papules covering 10- 30% BSA with or without symptom (e.g., pruritus, burning, tightness); limiting instrumental activity of daily living	Macules/papules covering >30% BSA with or without associated symptoms; limiting self-care activity of daily living	NA
Fever [F]	38.0 to < 38.6°C or 100.4 to < 101.5°F	≥ 38.6 to < 39.3°C or ≥ 101.5 to < 102.7°F	≥ 39.3 to < 40.0°C or ≥ 102.7 to < 104.0°F	40.0°C or ≥ 104.0°F

[A)] Grading based on FDA Guidance on Toxicity Grading Scales Adults/ Adolescents.
[B)] Grading based on Persistent crying in infants and children as an adverse event following immunization: case definition and guidelines for data collection, analysis, and presentation; The Brighton Collaboration Persistent Crying Working Group; Vaccine 2004.
[C)] Grading based on Division of AIDS, Table for Grading the Severity of Adult and Pediatric Adverse Events, corrected version 2.1, July 2017.
[D)] Grading according to Common Toxicity Criteria (CTC) for cancer trials, Version 5.0, 2017.
[E)] Grading based on Common Terminology Criteria for Adverse Events (CTCAE), NIH, v4.03, 2010.
[F)] Grading based on Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events Corrected Version 2.1 July 2017
[1)] crying which parent(s)/LAR(s) may describe as "fearful", "angry", "sorrowful", "in pain", "pitiful", "plaintiff", "never heard before in this child".
[2)] not episodic, not interrupted within the time period of 3h (e.g. by naps).
[3)] Any systemic reaction meeting the grade 4 criteria will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

17.2.6.3 Body Temperature Measurement

From vaccination (Day 1) until Day 15 after vaccination (i.e., the first 14 post-vaccination days including the day of vaccination), the parent/LAR should measure the participant's body temperature with an infrared thermometer targeting the forehead every evening, assessments by the parent/LAR should occur at the same time each day, starting approximately 8 hours after vaccination. To optimize the comparability of the documented body temperatures, all parents/LARs will be provided with an infrared thermometer and be instructed in its use. The parent/LAR may keep the thermometer after trial termination.

If fever (body temperature ≥38.0°C or 100.4°F) occurs, body temperature should be measured at least every 4 to 8 hours until it returns to normal (<38.0°C or 100.4°F). All body temperature measurements including the date and time should be recorded in the participant diary (see **Section 17.3**). The time at which the last body temperature of <38.0°C/100.4°F is recorded is considered to be the end of the fever episode. In case of fever ≥39°C/102.1°F the site should be contacted immediately. Note, in case a fever is not supported by a temperature reading it will be termed "feverishness", otherwise it is considered subjective.

Body temperature measurements from vaccination (Day 1) until Day 15 after vaccination (i.e., the first 14 post-vaccination days including the day of vaccination) will be recorded by the Investigator in the eCRF. If more than one body temperature value is recorded in the participant diary for a given day, the highest daily temperature reading will be recorded in the eCRF.

Body temperature measurements will be analyzed according to the "Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events Corrected Version 2.1 July 2017" for data analysis as described in **Table 4** above.

Outside of the first 14 post-vaccination days, daily body temperature measurement is not required. However, for participants experiencing an AE with symptoms of fever it is recommended that the parent/LAR measures the participant's body temperature in order to document fever as an AE in the Memory Aid. If fever (body temperature ≥38.0°C or 100.4°F) occurs, it is recommended that the parent/LAR measures the participant's body temperature every 4 to 8 hours until fever resolves (<38.0°C or 100.4°F) in order to document onset and resolution, as defined by the date of first body temperature measurement of ≥38.0°C or 100.4°F (date of onset) and the date of last body temperature measurement of <38.0°C or 100.4°F (date of resolution).

Note: in case a fever is not supported by a temperature reading it should be documented as "feverishness" in the eCRF.

17.3 Safety Questionnaires

Parents/LARs will be provided with the following types of safety questionnaires throughout the course of this trial:

- **Participant Diary** will be distributed to all parents/LARs at Visit 1 for the collection of safety information from the day of vaccination until the first 14 post-vaccination days (i.e. including the day of vaccination until Day 15)
- **Memory Aid** will be distributed to all parents/LARs for the collection of safety information outside the first 14 post-vaccination days (i.e. Day 16-Day 365) until the next visit.

The information collected in Participant Diary card is dependent on the Strata.

Stratum A and B

The following information will be collected in the Participant Diary (14 days post-vaccination, Day 1- Day 15):

- Measurement of body temperature (For further information see **Section 17.2.6.3**);
- Solicited injection site reactions[21], including severity (injection site pain, tenderness, erythema, redness, induration, swelling), for further information see **Section 17.2.6.1**;
- Solicited systemic reactions[xx], including severity (nausea/vomiting, headache, fatigue, myalgia, arthralgia, rash, fever), for further information see **Section 17.2.6.2**;
- Other AEs including severity;
- Any new concomitant medication or changes in medication taken after vaccination;

Stratum C

The following information will be collected in the Participant Diary (14 days post-vaccination, Day 1- Day 15):

Measurement of body temperature (For further information see **Section 17.2.6.3**);

- Solicited injection site reactions[22], including severity (injection site pain, tenderness, erythema, redness, induration, swelling), for further information see **Section 17.2.6.1**;
- Solicited systemic reactions[xxi], including severity (nausea/vomiting, inconsolable or excessive crying, disturbed sleep, loss of appetite, drowsiness, rash and fever), for further information see **Section 17.2.6.2**;
- Other AEs including severity;
- Any new concomitant medication or changes in medication taken after vaccination;

The information collected in the Memory Aid is the same for all Strata.

The following information will be collected in the Memory Aid (outside 14 day post-vaccination period, Day 16- Day 180):

- Unsolicited AEs including severity;
- Solicited reactions ongoing beyond the participant diary period;
- Any new concomitant medication or changes in medication taken after vaccination;

Assessments for the Participant Diary should start on the day of vaccination and occur at the same time each day, starting approximately 8 hours after vaccination for a total of 14 consecutive days (Day 1 to Day 15). The parent(s)/LAR(s) will be properly instructed on the reporting requirements and how to complete and use the diary, thermometer and measuring device (for assessment of measurable injection site reactions).

Safety questionnaires are always to be returned at the next visit and will be assessed by the Investigator together with the parent(s)/LAR(s), prior to these data being entered into the electronic case report form (eCRF). The Investigator will review and discuss the safety questionnaires with the parent(s)/LAR(s), ask about AEs occurring since the last visit. The Investigator has to sign the safety questionnaires to ensure completeness and reliability of (self-)reporting. The safety questionnaires are to be collected by the Investigator at indicated visits and new ones will be distributed. The entries in the respective safety questionnaires shall be evaluated and solicited/ unsolicited AEs graded for severity and relatedness to the vaccination by the Investigator.

[21] Solicited injection site and systemic reactions in Participant Diary will be assessed by the parent/LAR for absence, presence, severity and duration
[22] Solicited injection site and systemic reactions in Participant Diary will be assessed by the parent/LAR for absence, presence, severity and duration

The safety questionnaires will serve as source documentation. Entries in the safety questionnaires will be transcribed onto the appropriate eCRFs. Solicited AEs should only be documented in the participant diary section of the eCRF. In case of solicited SAEs the data should additionally be documented at the AE eCRF page. Unsolicited AEs and SAEs are always documented at the AE eCRF page. Any entry on the eCRF that does not correspond with an entry in the safety questionnaires will be explained by the Investigator on the relevant safety questionnaire page.

17.4 Safety Card

Parent(s)/LAR(s) will be provided with a Safety Card at Visit 1 (Day 1), informing and instructing the parent(s)/LAR(s) in lay language to contact the trial site in case of SAEs or symptoms suggestive of CHIKV infection.

17.5 Medical, Medication and Non-Drug Therapy History

17.5.1 Medical History

At screening, the participants' medical history will be described for the following body systems or surgery and start and end dates, if known: eyes, ears, nose, and throat; respiratory; cardiovascular; gastrointestinal; musculoskeletal; neurological; endocrine; metabolic; hematopoietic/lymphatic; dermatological; and genitourinary.

In addition, the medical history covering the last 3 years (if applicable) prior to screening will include but not limited to the following:

- Information on any prior vaccination
- Other vector-borne diseases in the past;
- Previous trauma to joints;
- Information on planned hospitalizations (including elective surgery) during the trial for medical conditions existing prior to or at trial entry. Such planned hospitalizations will not need to be reported as SAEs.

17.5.2 Concomitant Medications and Non-Drug Therapies

All medications received from 2 weeks prior to trial enrollment until completion/termination should be collected from all participants and recorded on the appropriate eCRF. In addition to product name (generic name), the dose, indication, route of administration and frequency as well as the start and end date of treatment will be documented.

In addition, medications to treat SAEs will be reported to the Sponsor on SAE Report Forms as described in **Section 4**. In context of this trial, information on non-drug therapies will only be collected in relation to SAEs.

The following medications are **not permitted** if administered within the specified trial periods (unless such treatment has to be administered in an emergency situation):

- Any blood products or immunoglobulins during the course of the trial;
- Prophylactic administration of antipyretics or analgesics within 24 hours prior to and during the first 72 hours after vaccination;

Note: Considering that low grade fever is not an emergency situation (if a low-grade fever gets treated with antipyretics: it is NOT prophylactic administration, but a therapeutic treatment for an elevated temperature).

For documentary purposes, any of the treatments listed above (including emergency treatment) given within these time periods requires special documentation and is to be documented as a protocol deviation.

The following medications and procedures will **delay** vaccination:

- Antipyretics or analgesics received within 24 hours prior to vaccination,
- any inactivated vaccines[23] within 14 days prior to vaccination,
- any live virus vaccine received within 28 days prior to vaccination,
- chronic steroid use[24] within 28 days prior to trial entry

Usage of any other medications or non-drug therapies is not restricted.

Additionally, medications that are not permitted prior to trial enrollment, resulting in exclusion from the trial, are reflected in the exclusion criteria in **Section 13.2**.

Note: from the ICF signature onwards events qualifying as a SAE will be documented and reported according to SAE requirements.

17.6 Vital Signs

Vital signs will include systolic and diastolic blood pressure (mmHg) and pulse rate (beats/min) while seated and at rest, and body temperature (°C).

Vital signs will be measured at screening (Visit 0) and at the vaccination visit (Visit 1) and are to be recorded before the vaccination is given. Vital sign values should be within normal values (upon investigator's discretions) before vaccination. In addition, after an observation period of 30 minutes (for sentinel participants 60 min) at the trial site following vaccination pulse rate as well as blood pressure while seated and at rest will again be assessed.

Vital sign values are to be recorded on the appropriate eCRF. For each vital sign value, the Investigator will determine whether the value is considered an AE (see definition in **Section 17.1.1**). If assessed as an AE, the medical diagnosis (preferably), symptom, or sign, will be recorded on the AE CRF. Additional tests and other evaluations required to establish the significance or etiology of an abnormal result, or to monitor the course of an AE should be obtained when clinically indicated. Any abnormal value that persists should be followed at the discretion of the Investigator.

17.7 Physical Examinations

Physical examination will be performed at all visits (Visit 0-8, ET) on the following body systems being described as **normal** or **abnormal**: general appearance, head and neck, eyes and ears, nose and throat, chest, lungs, heart, abdomen, extremities and joints, lymph nodes, skin, and neurological.

Abnormal conditions (except those qualifying as a SAE) detected at screening or prior to vaccination at Visit 1 will be recorded as medical history. At all other trial Visits, if a new abnormal or worsened abnormal pre-existing condition is detected, the condition will be recorded as an AE.

17.8 Clinical Laboratory Parameters

Blood and urine samples will be obtained for assessment of clinical laboratory parameters as outlined in the Schedule of Trial Procedures (see **Section 24**) or in the Screening and Trial Visits Section (see **Section 15.4**).

17.8.1 Urinalysis

A urine sample should be obtained from the participant and analyzed. Optional, only in the case the child is able to give urine during the trial visit. Sample collection should be performed in accordance with age considerations.

[23] Includes mRNA vaccines
[24] chronic steroid use defined as use of prednisone or equivalent ≥0.05 mg/kg/day longer than 14 days

Urinalysis — Standard urine dipstick for determining pH-value, specific gravity, leucocytes, nitrite, protein, glucose, ketones, urobilinogen, bilirubin, erythrocytes.

17.8.2 Safety Sample

A baseline safety laboratory blood [approx. 4mL] sample will be obtained from all participants at the screening visit; at Visit 5 (or at Early Termination Visit if it happens before Visit 5) the safety sample will be only taken from the sentinel participants in order to lessen the burden of blood draws for the majority of participants. If deemed necessary by the investigator a safety sample can be taken any time if safety concerns arise.

Hematology panel (approx.2 mL) — Hemoglobin, hematocrit, erythrocyte count, WBC count, differential WBC count (basophils, eosinophils, lymphocytes, monocytes, neutrophils), platelets, ESR.

Coagulation panel (approx. 2 mL) — Small blood coagulation (prothrombin time, aPTT and fibrinogen).

17.8.3 Assessment of Laboratory Values

Laboratory values will be evaluated according to the Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials (Food and Drug Administration). For the individual toxicity criteria refer to

Table 15.

Table 15: Toxicity Grading Scale for Abnormal Laboratory Assessments

	Mild (Grade 1)[1]	Moderate (Grade 2)	Severe (Grade 3)	Potentially life threatening (Grade 4)[2,6]
Hematology Parameters				
Hemoglobin (57 days of age to < 13 years of age, Female) - gm/dL	5.88 to 6.48	5.25 to < 5.88	4.03 to < 5.25	< 4.03
Hemoglobin (57 days of age to < 13 years of age, Male) – gm/dL	9.5 to 10.4	8.5 to < 9.5	6.5 to < 8.5	< 6.5
Hematocrit	Outside normal range[3]			
Erythrocyte count	Outside normal range[3]			
WBC Increase - cell/mm³	Outside normal range[3]			
WBC Decrease - cell/mm³	2,000 - 2,499[4]	1,500 - 1,999	1,000 - 1,499	< 1,000
Neutrophils Decrease - cell/mm³ (> 7 days of age)	800 - 1,000[4]	600 - 799	400 - 599	< 400
Lymphocytes Decrease - cell/mm³ (> 5 years of age, not HIV infected)	600 to < 650[4]	500 to < 600	350 to < 500	< 350
Monocytes	Outside normal range[3]			
Eosinophils - cell/mm³	Outside normal range[3]			
Basophils	Outside normal range[3]			

	Mild (Grade 1)[1]	Moderate (Grade 2)	Severe (Grade 3)	Potentially life threatening (Grade 4)[2,6]
Platelets Decreased - cell/mm³	125,000 – 140,000[4]	100,000 – 124,000	25,000 – 99,000	<25,000
ESR	Outside normal range[3]			
Coagulation Factors				
PT – increase by factor (prothrombin time)	1.1 to < 1.25 x ULN	1.25 to < 1.50 x ULN	1.50 to < 3.00 x ULN	≥ 3.00 x ULN
PTT (aPTT) – increase by factor (activated partial thromboplastin time)	1.1 to < 1.66 x ULN	1.66 to < 2.33 x ULN	2.33 to < 3.00 x ULN	≥ 3.00 x ULN
Fibrinogen increase - mg/dL	Outside normal range[3]			
Fibrinogen decrease - mg/dL	100 to < 200[4]	75 to < 100	50 to < 75	< 50

The table is made upon The Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Corrected Version 2.1 (July 2017). When a single parameter is not appropriate for grading an AE in both adult and pediatric populations, separate parameters with specified age ranges are provided. If no distinction between adult and pediatric populations has been made, the listed parameter should be used for grading an AE in both populations.
[1] In case the laboratory's normal ranges and absolute Grade 1 limits overlap, Grade 1 limits will prevail, i.e. the value will be classified as Grade 1 abnormality even if it is within laboratory normal ranges. Values between the laboratory normal ranges and absolute Grade 1 limits will be reported as no abnormality (Grade 0).
[2] The clinical signs or symptoms associated with laboratory abnormalities might result in characterization of the laboratory abnormalities as Potentially Life Threatening (Grade 4). For example, a low sodium value that falls within a grade 3 parameter (125-129 mE/L) should be recorded as a grade 4 hyponatremia unsolicited AE if the participant had a new seizure associated with the low sodium value.
[3] As the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events (Version 2.1 July 2017) does not provide any grading for Hematocrit, Erythrocyte count, Monocytes, Basophils, ESR, WBC increase, CRP and fibrinogen increase, these will only be analyzed as "outside normal range", as determined by laboratory standards and graded as described in **Section 17.2.2** upon investigator's judgement.
[4] Laboratory values should be adjusted to the DAIDS Table for Grading the Severity of Adult and Pediatric Adverse Events scale. Specifically, if laboratory reference range is more stringent than DAIDS grading scale the laboratory values should be reported as no abnormality (Grade 0). Similarly, if laboratory values are within the laboratory normal reference range, but fall into DAIDS grading scale, the values should be reported as indicated by the DAIDS grading scale.
[5] "ULN" is the upper limit of the normal range
[6] Any laboratory values that meet the Grade 4 criterion will need to be assessed by the Investigator if a seriousness criterion is met and may require expedited reporting.

Laboratory assessments for which no severity grading is described in

Table 15 are graded as described in **Section 17.2.2** upon Investigator's judgment.

The Investigator's assessment of each abnormal laboratory value, including its clinical significance, is to be recorded in the eCRF:

- Abnormal laboratory assessments that are considered clinically relevant, in the opinion of the Investigator, need to be documented as unsolicited AEs and assessed further for severity according to the toxicity grading scale provided in
- **Table** 15, causality and other assessments done for unsolicited AE (see **Section 17.2.5**).
- Abnormal laboratory assessments that are considered a symptom of an underlying AE or part of a syndrome that is reported as AE (e.g. presence of blood cells in urine in a person diagnosed with urinary tract infection) do NOT additionally need to be documented as unsolicited AE, but a respective comment should be added to the underlying AE.

Additional tests and other evaluations required to establish the significance or etiology of an abnormal laboratory result, or to monitor the course of an AE should be obtained when clinically indicated. Any abnormal value that persists should be followed at the discretion of the Investigator.

17.8.4 CHIKV Screening

A CHIKV Baseline Screening serum sample (approx. 1.2 mL blood) will be obtained from all participants at Visit 0 (Screening Visit) for CHIKV-specific IgG and IgM antibody testing using a rapid test. Only in case the rapid test is positive, the blood sample from the applicable participant will be subjected to a confirmatory anti-Chikungunya virus IgG and IgM ELISA testing.

Enrolment will then be stratified by CHIKV serostatus at baseline:

- up to 10 % CHIKV seropositive participants (i.e., IgM+/IgG+ or IgM-/IgG+)
- at least 90% CHIKV seronegative participants (i.e., IgM-/IgG-)

Participants who are IgM+/IgG- do not qualify for participation in this trial.

Note: A borderline ELISA result is considered negative. Hence, participants with ELISA results IgM borderline/IgG-, IgM-/IgG borderline, or IgM borderline/IgG borderline are mapped to seronegative group; participants with ELISA result IgM borderline/IgG+ are mapped to seropositive group; participants with ELISA result IgM+/IgG borderline are not eligible for the trial.

Note: Immunogenicity analysis will be stratified by baseline serostatus determined by µPRNT.

17.9 CHIKV Viremia

All participants will return to the trial site at Day 1 (Visit 1), Day 4 (Visit 2), Day 8 (Visit 3), Day 15 (Visit 4), Day 29 (Visit 5), Month 3 (Day 85, Visit 6), Month 6 (Day 180, Visit 7) and Month 12 (Day 365, Visit 8). Viremia sample [approx. 2 mL] will be obtained from all participants of Stratum A and only taken from sentinel participants from Stratum B and C in order to lessen the burden of blood draws for the majority of participants. Visit 5 (Day 29) samples will be only tested if Visit 4 (Day 15) tests were positive. For Visit 6, 7, 8 and ET (if applicable) viremia samples will be collected for clinically indicated retrospective analysis.

Viremia will be analyzed in plasma samples. Blood will be collected by venous puncture. Plasma samples will be shipped on dry ice, viral RNA will be isolated and viral CHIKV RNA will be specifically amplified by RT-qPCR. For discrimination between wild type and vaccine virus, samples tested positive in the nsp1 specific RT-qPCR will be analysed for the presence of the deletion in nsp3 of VLA1553, if required.

17.10 Adverse Events of Special Interest (AESI)

In addition to nonspecific transient myalgia and arthralgia that may occur after vaccination with any vaccine, participants will be carefully monitored for signs and symptoms similar to an acute stage CHIKV-associated event.

If a participant develops symptoms suggestive of an AESI, parent(s)/LAR(s)will be asked to contact the site immediately for clinical evaluation at an unscheduled visit.

Therefore, the following cluster of symptoms with or without remissions or exacerbations will receive particular consideration and are defined as early onset AESI:

1. Fever (≥ 38.0 °c / 100.4 °f, infrared measurement) **AND**

2. symptoms suggesting acute (poly)arthralgia/arthritis, myalgia, neurological symptoms (e.g., for meningoencephalitis, acute encephalitis, headache, seizures), back pain, lymphadenopathy, cardiac or ocular symptoms (e.g.,for uveitis and retinitis);

OR

one or more of the following signs and symptoms: macular to maculopapular rash (sometimes with cutaneous pruritus [foot plant]), pigmentary changes, bullous rash/ skin blistering, purpura, ecchymosis and

3. Onset of symptoms 2 to 21 days after vaccination (i.e., day 3 – day 22) **AND**
4. Duration of event ≥ 3 days.

The cluster of symptoms defined above but starting 22 or more days after vaccination (i.e., Day 23 or later) until trial end is defined as late onset AESI and will be documented and followed-up in the same manner as early onset AESI.

Note: AESIs with first symptom(s) occurring until 21 days after vaccination (Day 22) and additional symptom(s) occurring afterwards are still classified as early onset AESIs.

Events, AEs or (SAEs) which are part of the AESI qualifying symptoms (e.g. fever, headache, myalgia, arthralgia) but can be attributed (together with additional symptoms) to an underlying (serious) suspected or confirmed **diagnosis**, stating a disease or a syndrome, do not meet the AESI criteria.

The following interpretation guidance for handling of AESIs is to be observed:

- **Cluster of symptoms:** Fever plus at minimum one additional symptom need to overlap in their occurrence to make the cluster an AESI
- **On-set / end date of AESI:** The onset-date of the first symptom determines the overall onset-date of the AESI and the end date is the cessation of the last symptom.
- **Duration of the AESI of ≥ 3 days:** the overall AESI needs to last 3 days or more. It does not mean that each of the symptoms has to last 3 days.
- **AESI severity:** the most severe symptom determines the overall severity of the AESI
- **Seriousness of the AESI:** as soon as one of the AESI symptoms qualifies as SAE the overall classification of the AESI is a SAE.

Clinical Symptoms that may be a sign for a clinically significant condition need a medical work up (e.g. AESI laboratory parameter assessment) at the discretion of the investigator and a referral to a specialist as medically indicated. The concerned treatment should be performed according to current medical standard of care.

Additionally, participants presenting with acute arthralgia post-vaccination will be followed-up until resolution and monitored for recurrences until the end of the trial.

A blood sample for laboratory investigation will be taken upon presentation of the participant. Testing may include but is not limited to rheumatoid factor (RF), anticitrullinated protein antibody (ACPA), Ferritin and CRP. Retrospective investigation of a pre-vaccination sample may be considered for a thorough causality assessment.

Suspected AESI do not constitute a reason for withdrawal from the trial.

17.11 Adverse Event Reporting Procedures

17.11.1 Serious Adverse Event

Any SAE should be reported to the Safety Service Provider (PrimeVigilance) by fax or email within 24 hours after the Investigator has become aware of the event. Under certain circumstances the initial notification could be done by phone, but nevertheless a written SAE Report Form has to be submitted to the PrimeVigilance within 24 hours (see **Section 4**).

Correct SAE reporting will have to cite a diagnosis or a symptom. Any diagnosis and any symptom is regarded as separate SAE. However, if the Investigator considers several symptoms to be in the context of one underlying diagnosis, he/she may specify the diagnosis as the reportable SAE and describe the attendant symptoms in one single appropriate SAE report.

Medical or diagnostic procedures due to an underlying disease or symptom are not considered an AE but a consequent measure following an AE. A correct SAE report will therefore have to specify the disease or symptom as the reportable AE and the medical or diagnostic procedure as action taken.

In addition, expedited and periodic reporting to Competent Authorities and IRBs will be performed in accordance with local requirements. Further reporting details can be found in the trial-specific Safety Management Plan which is in accordance with respective US/EU requirements, International Council on Harmonisation (ICH) GCP, national laws and site-specific requirements. SAEs that are considered as probably or possibly related and additionally are unexpected need to be reported according to the requirements for suspected unexpected serious adverse reactions (SUSARs).

SAE reports will be reviewed by a trial site´s physician, PrimeVigilance, the Trial Medical Monitor, the Sponsor's Clinical Safety Physician and the DSMB.

17.11.2 Adverse Events of Special Interest (AESI)

Any AESI should be reported to PrimeVigilance by fax or email within 24 hours after the Investigator has become aware of the event. Under certain circumstances the initial notification could be done by phone, but nevertheless a written AESI Report Form has to be submitted to PrimeVigilance within 24 hours (see **Section 5**).

Participants will be carefully monitored for development of AESIs. A cluster of symptoms (see **Section 17.10)** associated with CHIKV infection, are defined as AESI. In case an AESI is identified, the Investigator will complete the AESI Report Form with all available information including medical records. This information will be provided to the Adjudication Committee which will perform a thorough review of each case and advise whether additional clinical work-up is required.

The DSMB will be presented all AESIs and will assess whether cases were new in onset and whether there is any relationship to administration of the trial vaccine. Narratives with detailed case descriptions will be provided for all AESIs.

17.12 Pregnancy

The risk of maternal to fetal transmission of Chikungunya virus during pregnancy cannot be excluded. Thus, female participants of childbearing potential and if involved in activities that could result in pregnancy must not become pregnant during the first three months post-vaccination. Contraceptive methods for all participants of reproductive potential will be provided by the sponsor.

However, if a female trial participant becomes pregnant, a respective Informed Consent/Assent needs to be obtained to enable data capture on the health condition during pregnancy/birth and up to three months after delivery or early termination of the pregnancy. The reporting of pregnancies starts with administration of the vaccination and last until trial completion (or ET Visit). All pregnancies that occur in this period, will be followed-up for three months after delivery or termination of the pregnancy. Any effect on either mother or fetus should be determined. A pregnancy which led to a congenital anomaly/birth defect must be followed-up by the Investigator longer or until resolution or stabilization. Duration of prolonged follow-up will be decided on an individual basis and in accordance with the Sponsor. The Investigator will prepare a narrative on the course of the pregnancy and the outcome.

The Investigator should report pregnancies within 24 hours of being notified using the Pregnancy Report Form. Reporting procedures are similar to SAE reporting procedures (contacts and processing), although a pregnancy is not considered an SAE.

If a seriousness criterion applies in addition to the pregnancy (e.g. hospitalization, congenital anomaly/birth defect) the pregnancy qualifies as an SAE. In such case a Pregnancy Report Form and an SAE Report Form have to be filled out (see **Section 4** and **Section 6**).

17.13 Safety Monitoring

17.13.1 Data Safety Monitoring Board

An independent Data Safety Monitoring Board (DSMB) will be installed for this trial. The DSMB will consist of four voting members who will be selected based upon their expertise with and understanding of infectious diseases, vaccines, clinical research and/or skills and knowledge in clinical medicine.

There are four DSMB reviews expected during the course of the trial to give recommendation on the trial continuation and the age step down upon review the safety data of the Cohorts in the following manner:

DSMB Review 1

Cohort I will be comprised of 5 sentinels of Stratum A (7 to 11 years) receiving the half dose formulation of VLA1553 in an open-label fashion, in a blinded fashion 35 participants of Stratum A (7 to 11 years) receiving the half dose formulation, 10 participants of Stratum A (7 to 11 years) receiving control.

The DSMB will meet to review available Day 8 safety data of sentinels in Cohort I. Based on no reservations, a recommendation will be given to the Sponsor and final Sponsor confirmation will be provided on whether to proceed with the randomization of the remaining 45 participants of Stratum A in Cohort I and to start vaccination in Cohort II.

DSMB Review 2

Cohort II will consist of 5 sentinels of Stratum B (3 to 6 years) receiving the VLA1553 half dose and 5 sentinels of Stratum A (7 to 11 years) receiving the full dose in an open-label fashion, and in a blinded fashion 35 participants of Stratum B (3 to 6 years) receiving the half dose, 35 participants of Stratum A (7 to 11 years) receiving the full dose, 10 participants of Stratum A (7 to 11 years) and 10 participants of Stratum B (3 to 6 years) receiving control.

The DSMB will meet to review available Day 8 safety data of sentinels in Cohort II as well as cumulative safety data. Based on no reservations, a recommendation will be given to the Sponsor and final Sponsor confirmation will be provided on whether to proceed with the randomization of the remaining 90 participants in Cohort II and to start vaccination in Cohort III.

DSMB Review 3

Cohort III will consist of five sentinels of Stratum C (1 to 2 years) receiving the half dose formulation in an open-label fashion, five sentinels of Stratum B (3 to 6 years) receiving the full dose in an open-label fashion and in a blinded fashion 35 participants of Stratum C (1 to 2 years) receiving the half dose, 35 participants of Stratum B (3 to 6 years) receiving the full dose, 10 participants of Stratum B (3 to 6 years) and 10 participants of Stratum C (1 to 2 years) receiving control.

The DSMB will meet to review available Day 8 safety data of sentinels in Cohort III as well as cumulative safety data. Based on no reservations, a recommendation will be given to the Sponsor and final Sponsor confirmation will be provided on whether to proceed with the randomization of the remaining 90 participants in Cohort III and to start vaccination in Cohort IV.

DSMB Review 4

Cohort IV will consist of five sentinels of Stratum C (1 to 2 years) receiving the full dose in an open-label fashion, and in a blinded fashion 35 participants of Stratum C (1 to 2 years) receiving the full dose, 10 participants of Stratum C (1 to 2 years) receiving control.

The DSMB will meet to review available Day 8 safety data of Cohort IV, as well as cumulative safety data. Based on no reservations, a recommendation will be given to the Sponsor and final Sponsor confirmation will be provided on whether to proceed with the randomization of the remaining 45 participants of Stratum C (1 to 2 years) in Cohort IV.

The DSMB will evaluate the following safety parameters in detail for all participants:

1. AEs (including solicited and unsolicited), SAE and AESI

2. Results of physical examinations

In addition, the DSMB will periodically review accruing safety information throughout the trial, as applicable. A meeting of the DSMB may be called at the discretion of the Sponsor, e.g. to address any safety concerns arising during the conduct of the trial.

The DSMB will ad-hoc review cases of SAEs and AESI. The DSMB will periodically review listings and summary tabulations of SAEs, Deaths, Solicited AEs and Unsolicited AEs.

A DSMB meeting is triggered if a holding rule is met (as described in **Section 12.1**) during recruitment of sentinel participants. The DSMB will review accruing safety data and decide whether to proceed with the vaccinations of the same Cohort and if recruitment for the next Cohort can be started without limitation.

A quorum of three DSMB voting members is required for a valid vote. If opposing votes are even, the vote of the DSMB chair will be decisive. A DSMB charter including a detailed description will be prepared.

A DSMB charter including a detailed description of the set-up, purpose, composition, responsibilities, types and schedule of DSMB meetings will be developed and effective prior to FPFV.

Responsibilities of the DSMB

- Provides independent monitoring of safety issues, which means it reviews and evaluates all SAE reports and trial discontinuations for: (i) increases in frequency of SAEs and trial discontinuations within the trial; and (ii) SAEs in the trial sample compared with the population based on what is known from the literature;
- Reviews data produced from the trial upon the Sponsor's request to determine whether the conditions on which trial design is based have remained the same or have changed. If changed, the DSMB will suggest whether or not the changes mandate changes to the protocol and suggest a protocol amendment;
- Upon the Sponsor's request, meets for discussion and gives recommendations to the Sponsor as to whether the trial should progress unchanged, or the trial requires changes, or the trial should be terminated prematurely;
- Can propose an unplanned safety analysis of clinical trial data;
- Can suggest unscheduled visits for specific evaluations for individual cases reviewed.

18. STATISTICS

18.1 Sample Size and Power Calculations

The total number of 240 children aged 1 to 11 years exposed to VLA1553 in this trial has been selected to provide a sufficient number of participants for proper safety evaluation. With 240 participants exposed, the trial will provide 95% confidence that an AE does not occur at a frequency of 1.24% or higher, if not observed in the trial.

18.2 Datasets and Analysis Cohorts

In terms of immunogenicity, up to 240 participants exposed to VLA1553, assuming 10% seropositive participants and among seronegatives an expected rate of 10% of drop-outs/major protocol deviations would leave 194 participants in the analysis and would allow determination of the GMT with meaningful accuracy. I.e., with an assumed Log10-SD of 0.45 and an expected GMT of 2954 (based on pooled data from Phase 3 results in adults), the 95%CI for the GMT would be [2551, 3421] for combined VLA1553 groups and [2397, 3640] for one dose group. Furthermore, these 97 participants per dose group would allow for the detection of a difference in GMTs of 2954 vs. 1943 between the groups with a two-sided significance level of 5% and a statistical power of 80%.

The safety analysis population contains all participants who entered into the trial and received one vaccination.

Statistical analyses for immunogenicity will be based on µPRNT serostatus. Detection of anti-CHIKV antibodies at screening will be used for enrollment at the sites only. The immunogenicity analysis population (IMM) is defined to include all vaccinated participants who have evaluable µPRNT antibody titer results at baseline and at least one post-baseline titer measurement after vaccination. The baseline seronegative immunogenicity analysis set (seronegative IMM) includes all IMM participants, who are seronegative ($\mu PRNT_{50} \leq 40$) at baseline (Day 1). The baseline seropositive immunogenicity analysis set (seropositive IMM) includes all IMM participants, who are seropositive ($\mu PRNT_{50} > 40$) at baseline (Day 1).

The per protocol analysis population (PP) contains all seronegative IMM participants who have no major protocol violations that could impact immune response. Examples that may lead to exclusion from the PP are provided here, further criteria may be defined in the SAP:

- Participant has a known or suspected defect of the immune system that can be expected to influence the immune response to the vaccine, such as participants with congenital or acquired immunodeficiency, status post organ transplantation or immuno-suppressive therapy within 4 weeks prior to Visit 1. Immuno-suppressive therapy is defined as administration of chronic (longer than 14 days) prednisone or equivalent ≥0.05 mg/kg/day within 4 weeks prior to trial entry, radiation therapy or immunosuppressive cytotoxic drugs/ monoclonal antibodies in the previous 3 years; topical and inhaled steroids are allowed;
- Participant has a history of immune-mediated or clinically relevant arthritis/arthralgia;
- Participant has received the wrong (not according to randomization) or no IMP.

These criteria for protocol violations are identified at the time of planning the trial. However, during the course of the trial unforeseen events may occur or new scientific knowledge may become available, therefore final decisions on whether any protocol violation could impact immune response and thus lead to exclusion from the PP will be made by the sponsor on a case by case basis. Sample testing issues may also lead to exclusion from the PP for particular time points.

18.3 Handling of Missing, Unused and Spurious Data

All statistical analysis will generally be based on observed values, missing values will not be imputed.

18.4 Methods of Analysis

A statistical analysis plan will be prepared before database closure/snapshot.

18.4.1 Safety Analysis

All participants entered into the trial, who receive the vaccination, will be included in the safety analysis.

The primary safety analysis will be the number and percentage of participants with solicited injection site reactions and solicited systemic reactions, by term and overall, up to 14 days after vaccination and will be presented by treatment group and overall. Groups will be compared using Fisher's exact test (Fisher-Freeman-Halton test).

For the secondary safety analyses tabulations will generally be provided separately for solicited AEs and unsolicited AEs, and for both types of AEs combined. 95% confidence intervals according to Altman (Wilson score interval) will generally be provided for all AE rates. Further details in addition to the outline below will be provided in the SAP.

The number and percentage of participants with any AE, any related solicited AE, any unsolicited AE, any related unsolicited AE, any related severe AE, any SAE, any related SAE, any medically attended AE, any AE leading to withdrawal from trial, and any AE occurring at a frequency of at least 10% up to Day 29, and up to Day 180, and any AESI (i.e. early and late onset AESI) and any related AESI up to Day 22, will be presented for treatment group overall and by system organ class (SOC) and preferred term (PT). Selected frequencies will be compared using Fisher's exact test (Fisher-Freeman-Halton test) as will be pre-defined in the analysis plan.

In addition, any AESI per FDA definition (synonym: chikungunya-like adverse reaction) will be provided as statistical output with details defined in the Statistical Analysis Plan. The FDA's definition of AESI is as follows: 'Fever (≥38 °C / 100.4 °F) and one or more of any of the following: arthralgia or arthritis, myalgia, headache, back pain, rash, lymphadenopathy, or certain neurological, cardiac or ocular symptoms that occurred with an onset within 30 days after vaccination', regardless of the order of their onset and their duration.

The occurrence of solicited local and systemic AEs will also be tabulated by participant diary day and mean duration of symptoms will be presented (for participants with symptoms).

Changes in any laboratory values from trial entry will be analyzed descriptively for sentinel participants and will be part of the unsolicited AE evaluation only in case of clinically relevant deviations. The rates of participants with laboratory assessments outside the normal range, and with abnormal laboratory parameters falling into the grade 0 vs. 1 through 3 will be calculated for treatment group overall, by visit and overall. The rate of participants with urinalysis results according to the test manufacturer's results categories will be calculated.

Selected safety analysis will be repeated stratified by baseline serostatus, these will be defined in the SAP.

18.4.2 Immunogenicity Analysis

All analyses for Part A, Part B and Part C of immunogenicity data will be performed primarily on the PP and secondarily on the IMM.

The secondary immunogenicity analysis will be a comparison of the GMTs in the per-protocol analysis set between the VLA1553 Trial Arms and control at Day 15, Day 29, Day 85, Day 180 and Month 12 post-vaccination by ANOVA (factor Trial Arm, age stratum and baseline serostatus), including pair-wise comparisons (Tukey's HSD test) between VLA1553 Trial Arms. The models will be applied on logarithmic

values and estimates and two-sided 95% confidence intervals for GMTs and GMT ratios will be obtained by back-transformation.

GMFI's will be analyzed at various time points analogously. The seroresponse rates and seroconversion for various trial days will be compared between the trial arms by Fisher-Freeman-Halton test, amended by Fisher's exact test for pairwise comparisons, and two-sided 95% confidence intervals will be calculated according to Altman (Wilson score interval). Select immunogenicity analyses will also be stratified by age stratum and by baseline serostatus (based on µPRNT), these will be defined in the SAP.

18.5 Planned Data Analysis of the Trial

The following data analyses will be performed:

- Part A includes safety and immunogenicity data after all participants have completed Visit 5 (Day 29).
- Part B includes safety and immunogenicity data after all participants have completed Visit 7 (Day 180).
- Part C includes safety and immunogenicity data after all participants have completed Visit 8 (Month 12).

Individual trial parts will be analyzed sequentially.

Part A analysis will be performed unblinded (blind will be maintained for trial sites) and a clinical trial report will be compiled. For the Part B analysis another clinical trial report will be compiled (integrated Clinical Trial Report of Part A+B). For the Part C analysis a final clinical trial report will be compiled (integrated Clinical Trial Report of Part A+B+C).

19. ETHICS AND REGULATORY ASPECTS

19.1 Compliance Statement

This trial will be conducted in accordance with this protocol, current ICH/GCP guidelines, Declaration of Helsinki, and with the applicable national and local regulatory requirements.

19.2 Institutional Review Board (IRB) and Regulatory Authorities

Before enrollment of participants into this trial, the protocol, informed consent/assent form, any promotional material/advertisements, and any other requested information will be reviewed and approved/given favorable opinion by the IRB and applicable regulatory authorities in accordance with local requirements. The trial will commence only upon the Sponsor's receipt of approval/favorable opinion from the IRB.

If the protocol and/or any other information given to the participant/parent(s)/LAR(s) is/are amended, the revised document(s) will be reviewed and approved/given favorable opinion by the IRB and applicable regulatory authorities in accordance with local requirements, where applicable. The protocol amendment will only be implemented upon the Sponsor's receipt of approval. Amendments that are intended to eliminate an apparent immediate hazard to participants may be implemented prior to receiving IRB and authority approval. However, in this case, approval must be obtained as soon as possible after implementation.

19.3 Participants Information and Informed Consent/ Assent

It is the Investigator's responsibility to obtain freely given written informed consent from parent(s)/LAR(s) and assent from the participant if applicable, as required by local regulations, depending on the participant's/parent's/LAR's/parents'/LARs' capability to understand the aims, methods, anticipated benefits and potential hazards of the trial. Written informed consent and assent if applicable has to be obtained before the participant is exposed to any trial-related procedures, including screening tests for eligibility.

The Investigator will explain that the parent(s)/LAR(s)/participants are completely free to refuse to enter the trial or to withdraw from it at any time, without any prejudice and need for justification. The parent(s)/LAR(s) will be informed that representatives of the Sponsor and health authority inspector may review their child's source records, and that these persons are bound by confidentiality obligations.

The informed consent form will include a comprehensive explanation of the proposed treatment without any exculpatory statements, in accordance with the elements required by ICH GCP and applicable regulatory requirements. Parent(s)/LAR(s) will be allowed sufficient time to consider their child's participation in the trial after having the nature and risks of the trial explained to them. By signing the informed consent form, parent(s)/LAR(s) agree(s) that all evaluations required by the trial will be completed, unless they withdraw voluntarily or are terminated from the trial for any reason.

The parent(s)/LAR(s), as applicable, will be given a copy of the signed informed consent documentation including the assent signed by the participant if applicable. The original of the signed and dated informed consent and assent if applicable must be retained in the site's records and is subject to inspection by representatives of the Sponsor, or representatives from regulatory agencies.

The Sponsor will provide to the Investigator in written form any new information that significantly bears on the participants' risks associated with trial vaccine exposure. The informed consent and assent form will be updated, if necessary. This new information and/or revised informed consent and assent form, that has been approved by the applicable IRB and regulatory authorities, where applicable, will be provided by the Investigator to the parent(s)/LAR(s) who consented for their child to participate in the trial.

20. QUALITY CONTROL AND QUALITY ASSURANCE

Quality tolerance limits will be pre-defined to identify systematic issues that can impact participant safety and/or the reliability of trial results. Important deviations from these limits and any remedial actions taken will be summarized in the clinical trial report.

20.1 Source Data and Records

Source data are defined as all information related to clinical findings, observations or other activities in the trial, captured in original records or certified copies of original records. The Investigator will permit trial-related monitoring, audits, IRB review and regulatory inspections, by providing direct access to source data/records. Source records should be preserved for the maximum period of time required by local regulations.

Source data entries must be made in accordance with local requirements. Signed and dated copies of the laboratory result reports have to be kept within the participant's source data file.

20.2 Investigator's Responsibility

The Investigator will comply with the protocol (which has been approved/given favorable opinion by the IRB), ICH GCP, and applicable regulatory requirements. The Investigator is ultimately responsible for the conduct of all aspects of the trial at the trial site and verifies by signature the integrity of all data transmitted to the Sponsor. The term "Investigator" as used in this protocol, and in trial documents refers to the Investigator or authorized trial personnel whom the Investigator has designated to perform certain duties. Sub-investigators or other authorized trial personnel are eligible to sign for the Investigator, except where the Investigator's signature is specifically required.

20.3 Training

The trial monitor will ensure that the Investigator and trial site personnel understand all requirements of the protocol, the investigational status of the vaccine, and his/her regulatory responsibilities as an Investigator. Training may be provided at an Investigator's meeting, at the trial site, and/or by instruction manuals. In addition, the trial monitor will be available for consultation with the Investigator and will serve as the liaison between the trial site and the Sponsor.

20.4 Monitoring

A designated monitor will check electronic system data and source data at regular intervals throughout the trial to verify completeness, accuracy and consistency of the data, protocol adherence, and adherence to GCP guidelines. The monitor will work and perform Source Data Verification according to the Clinical Management Plan. The Investigator will cooperate with the monitor to ensure that any discrepancies identified are resolved.

20.5 Audit and Inspection

Upon request, the Investigator will make all trial-related source data and records available to a qualified quality assurance auditor mandated by the Sponsor or to regulatory inspectors. The main purposes of an audit or inspection are to confirm that the rights and welfare of the participants have been adequately protected, and that all data relevant for the assessment of safety and efficiency of the investigational product have appropriately been reported to the Sponsor.

20.6 Non-compliance with the Protocol/ Deviations from the Protocol

Any deviations from the protocol will be tracked, actions defined, as feasible, and reviewed in Data Review Meetings for the trial part analysis and the final analysis for assessment of their influence on the quality of the trial analysis.

20.7 Confidentiality of Participant's Data

The Investigator will exercise all reasonable precautions within the constraints of the applicable regulatory requirements to maintain the confidentiality of participants' identities. On exported electronic source data or any other documents submitted to the Sponsor, participants will only be identified by participant number. Documents not for submission to the Sponsor, e.g. participant identification log and original ICF form, will be maintained by the Investigator in strict confidence.

21. DATA HANDLING AND RECORD KEEPING

21.1 Information of Investigators

An Investigator's Brochure containing all important data relating to the safe use of the IMP will be supplied to the Investigator prior to trial start.

The Investigator will be kept informed on new relevant safety data as the trial proceeds.

21.2 Electronic Case Report Forms (eCRFs)

21.2.1 Data Recorded Directly on Case Report Forms

An electronic Case Report Form (eCRF) will be used for this trial. Data will be recorded directly onto source documents before documentation in the eCRF.

21.2.2 eCRF entries

eCRF entries and corrections will only be performed by trial site staff authorized by the Investigator. Each user is informed of the clinical trial's web-site internet address and is allocated to a user account with personal password to access the confidential website. The personal password must be kept confidential and must only be used by the person to whom it was assigned. For additional authorized users at the site, a new user account has to be requested to ensure that each entry/change can be allocated to the person who performed the entry/change.

All visit data need to be recorded in the eCRF database as soon as possible after each trial visit.

21.2.3 Changes to eCRF data

Corrections may be requested as follows:

- Investigators' responses are checked as they are entered and are rejected if they do not fulfill quality criteria. A message will specify the type of error or syntax error and assist in its correction.
- If required, the CRA can ask for information to be corrected during monitoring.
- Computerized data-check programs and manual checks will identify clinical data discrepancies for resolution. Corresponding queries will be created within the data capturing system and the site will be informed about new issues to be resolved on-line.

All discrepancies will be solved on-line directly by the Investigator or by authorized staff.

Corrections of eCRF data may be performed by authorized staff only. The person performing the changes in the eCRF is required to electronically confirm the changes made.

21.2.4 eCRF Entry Validation

The Investigator will thoroughly review the data on the eCRF and will finally certify the contents of the eCRF by electronic signature after completion of each participant. If a correction is made to the eCRF data after the Investigator's final approval, the certification must be repeated after the changes have been performed.

21.2.5 Data Collection

All visits and assessments are entered into an interactive form. eCRFs will be source document verified following guidelines established before trial onset and detailed in the Monitoring Plan. Maintenance of

the trial database will be performed. Details to eCRF handling are provided in a trial specific eCRF manual.

21.2.6 Coding of Adverse Events, Drugs and Diseases

After data entry, AEs and medical history will be coded according to the latest MedDRA version. The same MedDRA version will be applied to all trial parts. Previous and concomitant medication and vaccines will be coded according to the latest version of the WHO Drug Reference List and Anatomical Therapeutic Chemical (ATC) Classification System.

21.3 Investigator File

21.3.1 Maintenance

The Investigator will maintain complete and accurate trial documentation in a separate file (i.e. Investigator File) provided during the initiation visit. The Investigator is responsible for maintaining complete, up to date and accurate trial records to enable the conduct of the trial to be fully documented. The records should include the clinical protocol as well as any amendments, trial approval letters, all original ICFs, drug dispensing and accountability logs and all relevant correspondence pertaining to the trial.

21.3.2 Archiving and Destruction

All trial related documents should be kept by the Investigator for the maximum period of time required by local regulations. No trial document should be destroyed without prior written agreement between the Investigator and the Sponsor. Should the Investigator elect to assign the trial documents to another party, or move them to another location, the Sponsor must be notified.

21.3.3 Provision of Additional Information

On request, the Investigator will supply the Sponsor with additional data relating to the trial or copies of relevant source records, duly anonymized. In case of particular issues or governmental queries, it is also necessary to have access to the complete trial records, provided that the participant's confidentiality is protected in accordance with applicable regulations.

22. PUBLICATION POLICY

All results generated in this trial will be considered to be strictly confidential. The Investigator may not submit the results for publication or presentation without prior written permission of the Sponsor. Authorship for any publication will be determined in mutual agreement. Within the scope of publication, co-authorship may be offered, at the sole discretion of the Sponsor, on a case-by-case basis taking scientific contribution into consideration. This is according to uniform requirements for manuscripts submitted to biomedical journals proposed by the International Committee of Medical Journal Editors.

All results of this trial will be made publicly available and published in accordance with globally accepted standards, in particular CEPI's publication policy

23. LIABILITIES AND INSURANCE

In case of any damage or injury occurring to a participant in association with the participation in the trial, insurance has been contracted.

The name, address and the insurance policy number will be given to both the Investigator prior to enrollment. Moreover, a copy of the insurance conditions will be filed on site.

The Investigator is responsible for dispensing the investigational product according to this protocol, and for its secure storage and safe handling throughout the trial.

24. SUPPLEMENTS

Figure 1: Phase 2 Clinical Trial Design

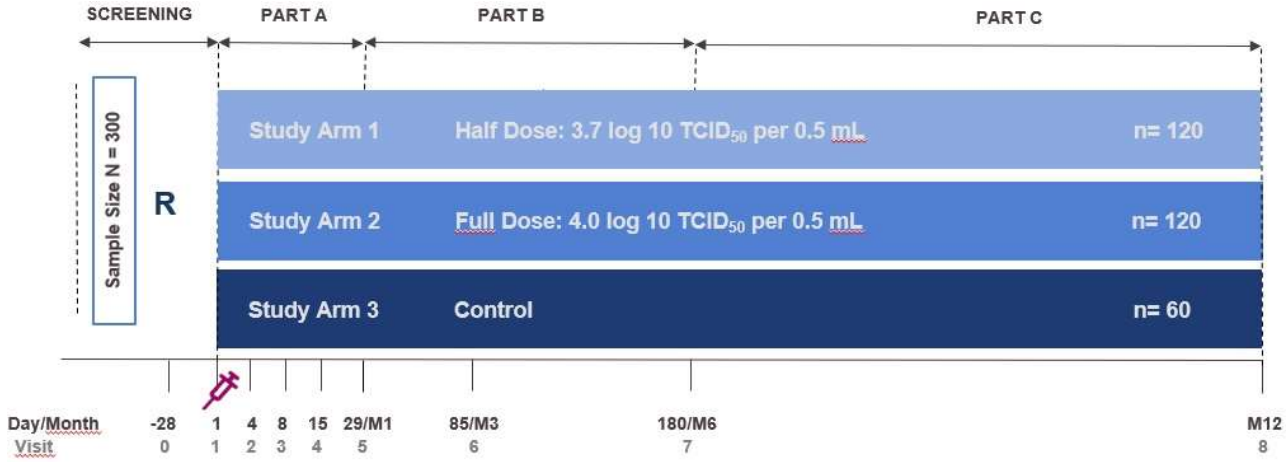


Figure 2: Staggered enrolment

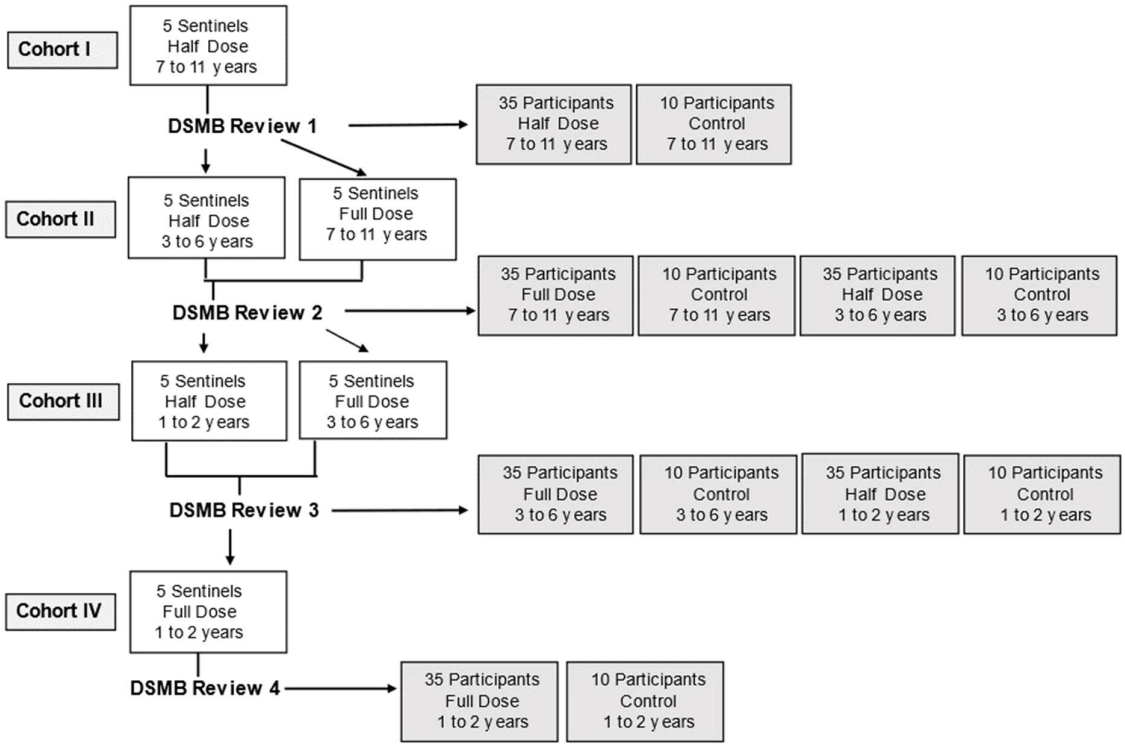


Table 1: Participant Distribution

Trial Arm	Dosage ($TCID_{50}$/dose)	Stratum	Cohort I	Cohort II	Cohort III	Cohort IV	Total N
1	VLA1553 half dose 3.7 log 10	A (7 to 11 yrs.)	5*+35	-		-	40
		B (3 to 6 yrs.)	-	5*+35	-		40
		C (1 to 2 yrs.)	-	-	5*+35	-	40
2	VLA1553 full dose 4.0 log 10	A (7 to 11 yrs.)	-	5*+35		-	40
		B (3 to 6 yrs.)	-	-	5*+35		40
		C (1 to 2 yrs.)	-	-	-	5*+35	40
3	Control	A (7 to 11 yrs.)	10	10		-	20
		B (3 to 6 yrs.)	-	10	10		20
		C (1 to 2 yrs.)	-	-	10	10	20
		Total	**50**	**100**	**100**	**50**	**300**

*Sentinel enrolment will be conducted in open-label fashion and sentinels recruited at a preferably single trial site. Sentinel vaccinations in the following cohort can be started based on the previous cohort sentinels' review by the Data Safety Monitoring Board and recommendation to the Sponsor and final Sponsor confirmation will be provided to proceed with the trial.

Table 5: Schedule of Trial Procedures and Assessments

		Part A					Part B		Part C	
	Visit 0 Screening (Day -28 to Day 0)	**Visit 1** [j] Day 1	**Visit 2** Day 4 (+/-1d)	**Visit 3** Day 8 (+/- 1d)	**Visit 4** Day 15 (+/-1d)	**Visit 5** Day 29 M1 (+/- 4d)	**Visit 6** Day 85 M3 (+/- 7d)	**Visit 7** Day 180 M6 (+/- 14d)	**Visit 8** Day 365 M12 (+/- 14d)	**Visit ET**
Informed consent/ assent [a]	X									
Inclusion/ Exclusion criteria	X	X (Review)								
Demographics [b]	X									
Medical history [c] (including vaccination history)	X	X (Update)								
Randomization		X								
Prior / Concomitant medications	X	X	X	X	X	X	X	X	X	X
Physical examination [d]	X	X	X	X	X	X	X	X	X	X
Vital signs [e]	X	X								
CHIKV Screening sample [1.2 mL]	X									
Pregnancy test (if applicable) [f]	X	X								
Immunogenicity sample [g] [2 mL]		X			X	X	X	X	X	X
Safety sample [h] [4 mL]	X					X				X [n]
Urinalysis [l]		X	X	X	X	X	X	X	X	X
Viremia sample [i] [2 mL]		X	X	X	X	X [k]	X [m]	X [m]	X [m]	X [m]
Maximum total approximate blood volume [mL] Stratum A sentinels/non-sentinels Stratum B sentinels/non-sentinels Stratum C sentinels/non-sentinels	5.2/5.2 5.2/5.2 5.2/5.2	4.0/4.0 4.0/2.0 4.0/2.0	2.0/2.0 2.0/none 2.0/none	2.0/2.0 2.0/none 2.0/none	4.0/4.0 4.0/2.0 4.0/2.0	8.0/4.0 8.0/2.0 8.0/2.0	4.0/4.0 4.0/2.0 4.0/2.0	4.0/4.0 4.0/2.0 4.0/2.0	4.0/4.0 4.0/2.0 4.0/2.0	8.0/6.0 8.0/6.0 8.0/6.0
VACCINATION		X								
Participant Diary		D	R/C/D	R/C/D	R/C					R/C
Memory Aid					D	R/C/D	R/C/D	R/C/D	R/C	R/C
AE/ AESI/ SAE Assessment	X [o]	X	X	X	X	X	X	X	X	X

ET … early termination; R … review; C … collect; D … distribute

[a] Occurs at enrollment before Screening.
[b] Demographics include date of birth, age, height, weight, BMI, gender, race and ethnicity.
[c] Symptoms noted (except those qualifying as a SAE) at Visit 1 (prior to first vaccination) are not considered AEs but will be recorded as medical history. Any prior vaccination should be documented in the Medical History.
[d] A physical examination will be performed on the following body systems being described as normal or abnormal: general appearance, head and neck, eyes and ears, nose and throat, chest, lungs, heart, abdomen, extremities and joints, lymph nodes, skin, and neurological.
[e] Vital signs will include systolic and diastolic blood pressure and pulse rate while seated and at rest, and body temperature measured with infrared thermometer targeting the forehead before vaccination. In addition, after an observation period of 60 minutes (open label phase) or 30 minutes (blinded phase) following vaccination, pulse rate and blood pressure while seated and at rest will again be assessed.
[f] At the screening visit and at indicated visits (i.e., V1) a pregnancy test (in accordance with local regulations) will be performed for all female Participants after onset of menarche and if involved in activities that could result in pregnancy.
[g] Immunogenicity sample [blood: 2 mL] for CHIKV-specific neutralizing antibody titer evaluation.
[h] Safety laboratory sample for hematology and coagulation panel [blood: 4 mL]. Safety sample will be taken from all participants at the screening visit; at subsequent visits (i.e., V5 or ET if earlier than V5) the safety sample will be only taken from the sentinel participants. If deemed necessary by the investigator a safety sample can be taken any time if safety concerns arise.
[i] Viremia sample for investigation of viremia by RT-qPCR [plasma: 2 mL]. Viremia sample will be taken from all participants of Stratum A and only taken from sentinel participants from Stratum B and C.
[j] All procedures/assessments (apart from Participant Diary distribution and AE assessment) occur prior to vaccination (unless stated otherwise).
[k] Visit 5 (Day 29) will be only tested if Visit 4 (Day 15) tests positive.
[l] Urinalysis (i.e. pH, specific gravity, leucocytes, nitrite, protein, glucose, ketones, urobilinogen, bilirubin, erythrocytes) - if possible.
[m] Samples should be obtained for clinically indicated retrospective investigation of viremia by RT-qPCR.
[n] Safety sample should be collected if ET is before Day 29 (Visit 5)-sentinels only.
[o] SAE(s) should be recorded from signed ICF onwards

Table 15: Toxicity Grading Scale for Abnormal Laboratory Assessments

	Mild (Grade 1)[1]	Moderate (Grade 2)	Severe (Grade 3)	Potentially life threatening (Grade 4)[2,6]
Hematology Parameters				
Hemoglobin (57 days of age to < 13 years of age, Female) - gm/dL	5.88 to 6.48	5.25 to < 5.88	4.03 to < 5.25	< 4.03
Hemoglobin (57 days of age to < 13 years of age, Male) – gm/dL	9.5 to 10.4	8.5 to < 9.5	6.5 to < 8.5	< 6.5
Hematocrit	Outside normal range[3]			
Erythrocyte count	Outside normal range[3]			
WBC Increase - cell/mm^3	Outside normal range[3]			
WBC Decrease - cell/mm^3	2,000 - 2,499[4]	1,500 - 1,999	1,000 - 1,499	< 1,000
Neutrophils Decrease - cell/mm^3 (> 7 days of age)	800 - 1,000[4]	600 - 799	400 - 599	< 400
Lymphocyte hours Decrease - cell/mm^3 (> 5 years of age, not HIV infected)	600 to < 650[4]	500 to < 600	350 to < 500	< 350
Monocytes	Outside normal range[3]			
Eosinophils - cell/mm^3	Outside normal range[3]			
Basophils	Outside normal range[3]			
Platelets Decreased - cell/mm^3	125,000 – 140,000[4]	100,000 – 124,000	25,000 – 99,000	<25,000
ESR	Outside normal range[3]			
Coagulation Factors				
PT – increase by factor (prothrombin time)	1.1 to < 1.25 x ULN	1.25 to < 1.50 x ULN	1.50 to < 3.00 x ULN	≥ 3.00 x ULN
PTT (aPTT) – increase by factor (activated partial thromboplastin time)	1.1 to < 1.66 x ULN	1.66 to < 2.33 x ULN	2.33 to < 3.00 x ULN	≥ 3.00 x ULN
Fibrinogen increase - mg/dL	Outside normal range[3]			
Fibrinogen decrease - mg/dL	100 to < 200[4]	75 to < 100	50 to < 75	< 50

The table is made upon The Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Corrected Version 2.1 (July 2017). When a single parameter is not appropriate for grading an AE in both adult and pediatric populations, separate parameters with specified age ranges are provided. If no distinction between adult and pediatric populations has been made, the listed parameter should be used for grading an AE in both populations.

[1] In case the laboratory's normal ranges and absolute Grade 1 limits overlap, Grade 1 limits will prevail, i.e. the value will be classified as Grade 1 abnormality even if it is within laboratory normal ranges. Values between the laboratory normal ranges and absolute Grade 1 limits will be reported as no abnormality (Grade 0).

[2] The clinical signs or symptoms associated with laboratory abnormalities might result in characterization of the laboratory abnormalities as Potentially Life Threatening (Grade 4). For example, a low sodium value that falls within a grade 3 parameter (125-129 mE/L) should be recorded as a grade 4 hyponatremia unsolicited AE if the participant had a new seizure associated with the low sodium value.

[3] As the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events

	Mild (Grade 1)[1]	Moderate (Grade 2)	Severe (Grade 3)	Potentially life threatening (Grade 4)[2,6]
(Version 2.1 July 2017) does not provide any grading for Hematocrit, Erythrocyte count, Monocytes, Basophils, ESR, WBC increase, CRP and fibrinogen increase, these will only be analyzed as "outside normal range", as determined by laboratory standards and graded as described in **Section 17.2.2** upon investigator's judgement. [4] Laboratory values should be adjusted to the DAIDS Table for Grading the Severity of Adult and Pediatric Adverse Events scale. Specifically, if laboratory reference range is more stringent than DAIDS grading scale the laboratory values should be reported as no abnormality (Grade 0). Similarly, if laboratory values are within the laboratory normal reference range, but fall into DAIDS grading scale, the values should be reported as indicated by the DAIDS grading scale. [5] "ULN" is the upper limit of the normal range [6] Any laboratory result meeting the Grade 4 criteria will need to be assessed by the investigator if a seriousness criterion is met and may require expedited reporting.				

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