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Protocol MV-ZIKA-RSP-101

EudraCT 2019-000840-93

Observer blinded, randomized trial to evaluate safety and immunogenicity of a novel vaccine formulation MV-ZIKA-RSP

Sponsor	Themis Bioscience GmbH Muthgasse 11 1190 Vienna, Austria Tel: PPD
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Protocol Approval

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Investigator's Approval

I confirm that I have read the protocol and that I agree to conduct this study in accordance with the protocol, International Conference on Harmonisation and GCP guidelines and with the applicable local regulatory requirements. Moreover, the site will keep all information obtained from the participation in this study confidential unless otherwise agreed in writing.

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1. STUDY SYNOPSIS

Protocol Number	MV-ZIKA-RSP-101
Title of the Study	Observer blinded, randomized trial to evaluate safety and immunogenicity of a novel vaccine formulation MV-ZIKA-RSP
Study Centres	One site in Vienna, Austria
Study Period	Study start (FSI) August 2019
Study Phase	Clinical Phase I
Objectives	**Primary Objective** – To investigate safety and tolerability of MV-ZIKA-RSP, a novel vaccine formulation, up to day 56 after first immunization consisting in one or two vaccinations **Secondary Objective** – To investigate long-term safety, up to study day 182, after first immunization – To investigate immunogenicity up to day 56 after first immunization consisting in one or two vaccinations with different doses – To get information about the optimal dose of MV-ZIKA-RSP vaccine regarding safety, tolerability, and immunogenicity – To investigate specific cell mediated immunity induced by all different treatment groups
Study Design	Observer blinded, block-randomized, dose finding, single centre, phase I trial in 48 participants. After completion of screening procedures, participants are randomized to one of four treatment groups (A, B, C or D): – **A**: 14 participants will receive **two** high dose treatments with MV-ZIKA-RSP- 1 x10^5 (±0.5 log) $TCID_{50}$ /dose on day 0 and 28 – **B**: 14 participants will receive **two** low dose treatment with MV-ZIKA-RSP- 2,5 x10^4 (±0.5 log) $TCID_{50}$ on day 0 and 28

	– **C**: 12 participants will receive **<u>one</u>** high dose treatment with MV-ZIKA-RSP- 1 x10^5 (±0.5 log) $TCID_{50}$ /dose on day 0 and placebo on day 28 – **D**: 8 participants will receive placebo on day 0 and day 28 After the screening visit (V0), participants will perform three additional visits to the site. On study days 0 and 28 participants will receive the vaccination, as well as, safety assessments together with immunogenicity sample collection. On study day 56 no vaccination will be done, but safety assessments and sample collection are scheduled. Safety follow-up assessments will be done by a phone call (Visit 4) approximately 6 months (day 182) after the first vaccination. A preliminary analysis will be performed after all study subjects completed Visit 3 (day 56).
Number of Participants	48 healthy participants
Main Inclusion Criteria	Healthy female or male volunteers, 18 - 55 years old at screening.
Investigational Medicinal Product	MV-ZIKA RSP, presented as a liquid frozen (lf) formulation, is a live recombinant measles virus vaccine expressing ZIKA virus antigens; suspension for injection; administered by i.m. injection, used in two different dose levels: – High dose: 1 x10^5 (+/- 0.5 log) $TCID_{50}$ – Low dose: 2,5 x10^4 (+/- 0.5 log) $TCID_{50}$
Placebo	Physiological saline solution (0.9% NaCl), administered by i.m. injection
Duration of the Study	Study duration per subject will be 182 days (6 months), consisting of 4 weeks treatment period (study day 0 till study day 28) and 22 weeks follow up period (study day 28 till study day 182). The overall study duration is estimated to be approximately 18 months, from study approval until reporting.
Statistical Methods	The primary analysis will compare adverse events rates up to day 56 between treatment groups using Fisher exact tests. Solicited AEs, unsolicited AEs and SAEs will be summarized by severity and term up to day 56 in the preliminary analysis and AE up to 182 are summarized in the final analysis.

	The immunogenicity analysis will compare the anti-zika-rsp VNT antibody geometric mean titre (GMT) between the treatment groups. GMTs and GMT ratios will be estimated by applying an analysis of variance including the factor treatment group. This will be done using log10 transformed data and taking the anti-log of the resulting point estimates for the least squares means, least squares means differences and the corresponding 2-sided 95% CIs. P-values will also be provided to compare GMTs between the treatment groups adjusted for multiple comparisons according to Tukey-Kramer. Seroconversion rates will be compared between groups using Fisher's exact test.
Criteria for Evaluation	Primary endpoint – Rate of adverse events (AEs) up to study day 56 Secondary endpoints: – Rate of solicited and unsolicited AEs, as well as, reported serious adverse events (SAEs) up to study day 182 (long-term safety) compared between the 4 different treatment groups. – Immunogenicity on study days 0, 28 and 56 as confirmed by the presence of functional anti-zika antibodies as determined by VNT and by ELISA – Cell-mediated immunity specific for ZIKA-RSP, up to day 56, as confirmed by the presence of specific functional $CD4^+$ and $CD8^+$ T-cells – Safety laboratory parameters (haematology, serum chemistry, urinalysis)
Benefit/Risk Assessment	The measles virus vector is well known. Possible risks that are frequently associated with vaccinations are the occurrence of local reactions e.g. oedema, induration and erythema, transient local pain or tenderness at the injection site as well as mild to moderate headache, myalgia, flu-like symptoms or fatigue.

	Like any other vaccine, MV-ZIKA-RSP might induce allergic and anaphylactic reactions, apart from the described local reactions at the vaccination site and systemic flu-like reactions. In rare cases the injection can lead to a vasovagal reaction immediately after injection of the vaccine. The needle pricks for blood sampling may also cause local reactions such as oedema. The participants cannot expect direct benefit from study participation, except for potential boosting of measles immunity and increasing immunity to zika.

2. LIST OF ABBREVIATIONS

Ab	Antibody
AE	Adverse Event
AESI	Adverse Event of special interest
AGES	Österreichische Agentur für Gesundheit und Ernährungssicherheit GmbH
AMG	"Arzneimittelgesetz" [Austrian Law on Regulation of Therapeutic Goods]
ALT	Alanine Aminotransferase (SGPT)
aPTT	Abbreviated Partial Thrombin Time
AST	Aspartate Aminotransferase (SGOT)
CA	Competent Authority
CCID50	Cell Culture Infectious Dose 50%
CHIKV	Chikungunya Virus
CRO	Clinical Research Organisation
CV	Curriculum Vitae
DSMB	Data Safety Monitoring Board
(e)CRF	(electronic) Case Report Form
EC	Ethics Committee
ELISA	Enzyme Linked Immunosorbent Assay
ER	Emergency Room
FDA	(United States) Food and Drug Administration
FSI	First Subject In
GCP	Good Clinical Practice
GMO	Genetically Modified Organism
GMT	Geometric Mean Titre
HSA	Human Serum Albumin
HBV	Hepatitis B Virus
HCV	Hepatitis C Virus
HIV	Human Immunodeficiency Virus
HSD	Honest Significant Difference
IEC	Independent Ethics Committee
i.m.	Intra Muscular
IB	Investigator's Brochure
ICH	International Conference on Harmonisation
IMP	Investigational Medicinal Product
IRB	Institutional Review Board
ITT	Intent-To-Treat
MAE	Medically-attended Adverse Event
MedDRA	Medical Dictionary for Regulatory Activities
mITT	Modified Intent-To-Treat
Mo	Month
MV	Measles Virus
NHP	Non-Human Primate
PBMC	Peripheral Blood Mononuclear Cell
PP	Per-Protocol
$PRNT_{50}$	Plaque Reduction Neutralisation Test 50%
PT	Prothrombin Time
SAE	Serious Adverse Event
SAR	Serious Adverse Reaction
SPS®	Stabilising and Protecting Solutions
SUSAR	Suspected Unexpected Serious Adverse Reaction
$TCID_{50}$	Tissue Culture Infective Dose 50%
VNT	Virus Neutralization Test
WHO	World Health Organisation

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4. BACKGROUND INFORMATION AND RATIONAL

4.1 Investigational Drug

The MV-ZIKA-RSP vaccine candidate is a live attenuated recombinant viral vectored vaccine for prophylaxis of Zika virus infection. [redacted], chemically synthesized and inserted into the Schwarz vaccine strain of measles virus (MV) to produce the candidate vaccine MV-ZIKA-RSP.

[redacted] and colleagues at the Pasteur Institute in Paris recently introduced the live attenuated MV vaccine (Schwarz strain – MV/Schw) as a vector to express heterologous viral antigens [described in detail in Combredet, 2003]. This technology can be used as a platform for the generation of prophylactic vaccines against different indications (i.e. emerging infectious diseases).

The MV vector has been shown to stably express large, heterologous antigens up to 5kb long. To date, the vector has been used to generate several recombinant MV clones expressing heterologous viral antigens including Chikungunya Virus (CHIKV) (Brandler 2013), West Nile Virus (WNV) [Desprès, 2005; Brandler, 2012], Dengue virus (DENV) [Brandler, 2007; Brandler, 2010], and human immunodeficiency virus (HIV) [Lorin, 2004].

The measles vector-based Chikungunya vaccine is advanced to late stage clinical development. The Chikungunya vaccine was the first measles vectored infectious disease candidate tested in healthy adult subjects (Ramsauer et al. 2015). A Phase 2 clinical trial was recently completed showing the safety and immunogenicity of the vaccine candidate in 263 healthy adult subjects (Reisinger et al 2018).

A Zika vaccine candidate was previously developed by the study sponsor (Themis). A Phase 1 clinical trial was conducted in Austria in 2016/2017 (2016-004212-34/ NCT02996890), details are described below.

For a detailed description of the vaccine construct and mechanism of action please refer to the current version of the MV-ZIKA-RSP investigators' brochure.

4.2 Disease Background

Zika virus is an emerging mosquito-borne flavivirus initially isolated from a rhesus monkey in the Zika forest in Uganda in 1947 (Gubler, 2007; Dick, 1952). The first human infection was reported in Nigeria in 1954 (Macnamara, 1954). Like dengue and chikungunya viruses, Zika virus adapted from an ancestral transmission cycle involving non-human primates and a broad

spectrum of forest mosquito species as vectors to an urban cycle involving humans as reservoirs and the widely distributed Aedes mosquitoes as vectors. Since the 1950s, Zika virus had only been reported as circulating sporadically in Africa and Southeast Asia. In 2007, Zika virus was isolated for the first time in the Pacific, on the Micronesian island of Yap (Duffy, 2009). Between October 2013 and April 2014, French Polynesia experienced the largest Zika outbreak ever reported at that time (Cao-Lormeau, 2013). More than 32000 patients were suspected for Zika virus infection. Between 2014 and 2015, Zika virus had spread to other Pacific islands, notably the Cook Islands and Easter Island (Chile). In March 2015, Brazil reported the autochthonous transmission of Zika virus (Zanluca, 2015) and declared an unprecedented outbreak 6 months later with preliminary estimates of 440,000 to 1.3 million cases of infection through December 2015 (European Centre for Disease Prevention and Control, 2015). Since the outbreak in Brazil the virus has been found in over 60 countries and territories worldwide (Center for Disease Control and Prevention, Nov 2016). Although WHO has declared an end to its global health emergency in Nov 2016, the Zika crisis is not considered over. The virus is still spreading, and it remains a global health threat.

The current Zika epidemic is the largest epidemic ever recorded for this virus. Usually, a Zika infection is mild with symptoms such as fever, rash, conjunctivitis and joint pain lasting for several days to a week. People often do not realize they have been infected. Symptoms of Zika are often like other viruses spread through mosquito bites, like dengue and chikungunya. Although infection with Zika virus was usually associated to mild disease, its emergence in the Americas has coincided with a steep increase in patients developing Guillain-Barré syndrome. Moreover, infection with Zika virus has been linked to the birth of babies with neurological complications, in particular congenital microcephaly (WHO, 2016; ECDC 2015; Soares de Araújo, 2016), and it was shown that when pregnant women are exposed to Zika virus during the first trimester of pregnancy, the risk of microcephaly for the newborn is increased 50 times from 2/10 000 to 1/100 (Cauchemez, 2016). In Feb 2016, the WHO declared the suspected link between Zika virus and neurological disorders and neonatal malformations a Public Health Emergency of International Concern.

In this context, in March 2016, experts gathered at WHO agreed that the development of a preventive vaccine is a major priority to respond to Zika epidemics in the future (WHO, Media centre, 2016). A clear target product profile (TPP) for this vaccine is still under work, but pragmatic strategies were asked to fast track the development of a safe an effective product. Due to the established link between Zika virus infection and the appearance of congenital microcephaly in babies born to infected mothers, one could argue that a Zika vaccine has to be suitable for use in pregnant women. However, no licensed vaccine is currently recommended for the use in pregnant women. Moreover, with the demonstrated association of Zika infection with Guillain-Barré syndrome, the observation of possible sexual transmission,

and the appearance of developmental defects probably appearing very early in pregnancy, it is very likely that the TPP will be addressed to the general population. In any case a Zika vaccine will have to demonstrate an excellent safety profile, particularly concerning the risk of neurotropism.

4.3 Preclinical Studies

Brief Summaries of preclinical studies are given below. These studies are described in more detail in the current version of the MV-ZIKA-RSP investigators' brochure.

4.3.1 Non-Clinical Pharmacology Studies Performed

The immunogenicity and efficacy of MV-ZIKA-RSP vaccine candidates were examined in non-human primates and in transgenic CD46/IFNAR mice (CD46tg; IFNAR -/-), an animal model for the MV vector system. The vaccine immunogenicity was determined in mice by Enzyme-linked immuno-sorbent assay (ELISA) and plaque reduction neutralization assays (PRNT) from the sera of immunized mice. In addition, we showed the vaccine efficacy in two mouse challenge models, one lethal model and one disease model. In both models, all mice were protected against ZIKAV challenge.

Furthermore, the vaccine immunogenicity and efficacy were confirmed in non-human primates, which is a more relevant, immunocompetent animal model.

4.3.2 Primary Pharmacodynamics

<u>Analysis of the immunogenicity and protective efficacy of MV-ZIKA_RSP vaccine candidate in CD46-IFNAR mice after prime/boost immunization and subsequent Zika virus challenge model</u>

CD46-IFNAR mice were immunized twice intraperitoneally (ip) with MV-ZIKA RSP (10^6 $TCID_{50}$) one month apart. Control mice were immunized with the same dose of empty Schwarz vaccine strain of MV vector. Subsequently, one month after final immunization the mice were challenged by ip injection of 10^5 $TCID_{50}$ of an African Zika virus strain (Zika AFR). Blood samples were collected 1 month after each immunization. The sera were analyzed for the presence of functional, neutralizing antibodies by plaque reduction neutralization test (PRNT). In addition, enzyme-linked immunosorbent assays (ELISAs) were performed to detect anti-ZIKV binding antibodies. All animals that received two doses of the MV-ZIKA_RSP vaccine were protected against lethal challenge and did not show any signs of disease.

An additional animal model was used to show Zika virus viremia in the measles mouse model. Animals were immunized with two doses of MV-ZIKA RSP (10^6 $TCID_{50}$) at day 0 and day 28,

followed by challenge with Zika virus (South African Strain). This strain causes viremia but no signs of disease in the mouse model. The Zika virus viremia after challenge was analyzed at different time points (day 1, 2, 3, 5, 7, 10, and 21) by determination of viral ZIKV RNA in blood samples by quantitative PCR. Again, all animals that received the MV-ZIKA_RSP vaccine showed very high titers of neutralizing antibodies. Accordingly, were protected against Zika virus viremia.

Protective efficacy of the MV-ZIKA RSP vaccine in non-human primates.

The vaccine efficacy and safety were demonstrated in a mouse model that allows measles virus replication due to a deficiency in the Interferon system. Thus, the vaccine immunogenicity and efficacy were tested in non-human primates (Cynologous Macaques). This animal model is frequently used for vaccine studies with measles vectored vaccines as well as for Zika disease models.

Eight animals per groups were immunized with MV-ZIKA_RSP or with the measles vector backbone MV-Schwarz. The animals received two MV-ZIKA RSP immunizations at 10^5 $TCID_{50}$, 4 weeks apart. The immunogenicity was demonstrated at 4 weeks after the first and the second dose. All animals that received MV-ZIKA_RSP showed seroconversion to anti-ZIKA antibodies by ELISA and all animals showed high levels of neutralizing antibodies.

Seventeen weeks after the first dose all animals were challenged with $1x10^4$ $TCID_{50}$ Zika virus. All animals in the MV-Schwarz groups showed a viremia between days 1 -9 after challenge. The viremia in the serum was as high as $1x10^7$ RNA copies per mL serum. In contrast, animals that received the MV-ZIKA_RSP vaccine were fully protected against challenge. Three out of eight animals showed a small increase in virus copy numbers at peak viremia on day 4, but the levels were at or below the limit of quantification.

In summary, the MV-ZIKA vaccine induced a strong and protective immune response in a relevant animal's model.

Repeated-dose toxicity study by intramuscular route in cynomolgus monkeys followed by a 3-week treatment free period

The objective of this study was to evaluate the potential toxicity of the test item, MV-ZIKA RSP, following an immunization schedule corresponding to three intramuscular administrations of the Intended Human Dose (IHD), administered every 2 weeks, to cynomolgus monkeys. On completion of the treatment period, designated animals were held for a 3-week treatment-free period in order to evaluate the reversibility of any findings or potential delayed effects.

One group of six male and six female cynomolgus monkeys received the test item MV-ZIKA RSP, by intramuscular administration on Days 1, 15 and 29 at the maximal intended human dose of 2 x 106 TCID50/injection. A control group of three animals per sex received the control

item (sterile saline), under the same experimental conditions. Two days after the last injection (i.e. Day 31), the animals were euthanized (early sacrifice), except for three animals per sex in the test item-treated group which were kept for a 3-week treatment-free period and euthanized on Day 50 (late sacrifice).

Measles and Zika serological status of each animal were checked before allocation to the study and only seronegative animals were included. The animals were checked daily for mortality and clinical signs. Local tolerance at the injection sites was assessed on each day of administration, before, 4 and 24 hours after administration. Rectal temperature was recorded on Days 1 and 29, before dosing, 4 and 24 hours after administration. Body weight was recorded pre-test, on the first day of administration and then daily during the week after each administration, and at least twice weekly until the end of the study. Group food consumption was checked daily. Ophthalmological examinations were performed before the beginning of the treatment period and once in the week before early and late sacrifice. Hematology, coagulation, blood biochemistry and urinary investigations were performed once before the beginning of the treatment period, on Day 4 and at scheduled euthanasia. Blood samples for the determination of C-Reactive Protein (CRP) levels were taken 24 hours and 4 days after administration on Days 1 and 29. Blood samples for the determination of serum levels of antibodies against Measles antigen and Zika antigen were taken once pre-test, on Days 15 (before administration), 31 and then 50. Blood samples for the determination of serum levels of antibodies against Zika antigen were also taken on Day 29 (before administration). On completion of the treatment (early sacrifice) or treatment-free period (late sacrifice), the animals were sacrificed, and a full macroscopic post-mortem examination was performed. Designated organs were weighed, and selected tissue specimens were preserved. A microscopic examination was performed on all animals.

Results

No unscheduled deaths occurred during the study and there were no clinical signs or local reactions at the injection sites. Rectal temperature and body weight were unaffected. No ophthalmological changes were observed at early and late sacrifice. No effects were observed among hematology, coagulation, blood biochemistry and urinary parameters during the study. CRP levels were increased 24 hours after administration on Days 1 and 29 in all test item-treated animals and had returned to control values 4 days after the injections.

All monkeys seroconverted to measles after the first immunization with the test item. In addition, it resulted in the generation of functional neutralizing antibodies against Zika virus in all test item treated animals and therefore confirmed the immunogenicity of the MV-ZIKA RSP vaccine candidate.

At pathology, there were no systemic findings, but the test item administration induced mild, non-adverse, partially reversible inflammatory changes at the injection sites. At early sacrifice (Day 31), in test item-treated animals, an increased and more acute inflammation compared to controls was present at the injection site that was injected two days before sacrifice. It was characterized by infiltrates of granulocytes mainly, with increased incidence/severity of lymphoid infiltrates. At the injection site that was injected on Day 1, a mild inflammatory (granulomatous) reaction composed of macrophages and multinucleated giant cells was present with similar incidence and severity in both controls and test item-treated animals and was suggestive of an adjuvant-related reaction. None of the changes were considered to be adverse. At late sacrifice (Day 50), there was complete recovery of the granulocytic infiltrates at the injection site injected on Day 29. Minimal lymphoid infiltrates and minimal to slight inflammatory granulomatous reaction were still observed at the injection sites.

Conclusion

The potential toxicity of the test item, MV-ZIKA RSP, was evaluated in cynomolgus monkeys following an immunization schedule corresponding to 3 intramuscular administrations of the Intended Human Dose (IHD), administered every 2 weeks. Administration of the MV-ZIKA RSP vaccine candidate resulted in the generation of functional neutralizing antibodies against Zika virus in all immunized cynomolgus macaques and therefore confirmed the immunogenicity of the MV-ZIKA RSP vaccine candidate. The test item was well tolerated, inducing only a transient increase in CRP levels after administration and mild, non-adverse, partially reversible inflammatory changes at the injection sites.

4.4 Clinical Studies

As this is the first clinical study with MV-ZIKA-RSP vaccine, no clinical data are available for this IMP. However, a different vaccine (MV-ZIKA) against zika have been evaluated by our group in one completed phase I study (MV-ZIKA-101, EudraCT 2016-004212-34). In this case, the MV-ZIKA vaccine tested is a recombinant live attenuated measles vaccine, based on the backbone of the measles Schwarz virus strain for prophylaxis of Zika infection. The heterologous antigens are derived from the Zika virus.

Safety data from this study was shared in real time with the sponsor and communicated to all clinical trial sites in order to ensure that any safety signals that emerge are promptly addressed. Based on this previous clinical study, acceptable safety and tolerability as well as immunogenicity are expected.

4.4.1 Phase I Study

MV-ZIKA-RSP:

No clinical study with MV-ZIKA-RSP have been performed until now. One phase 1 study with a similar Zika-vaccine candidate has been performed and is described below.

MV-ZIKA-101 (EudraCT 2016-004212-34):

A phase I study was conducted in two sites in Vienna, Austria. Site # 01 at the Department of Clinical Pharmacology and Site # 02 the Institute of Specific Prophylaxis and Tropical Medicine both from the Medical University Vienna.

The name of this Phase I clinical study was “Observer blinded, randomized, placebo controlled, dose finding trial to evaluate the optimal dose of MV-ZIKA, a new vaccine against Zika virus, regarding immunogenicity, safety, and tolerability in healthy volunteers”.

The purpose of this first-in-man study was to evaluate the optimal dose considering immunogenicity, safety and tolerability of the MV-ZIKA vaccine. We further investigate the immunogenicity, safety, and tolerability of MV-ZIKA during the treatment period up to 84 days after first investigational medicinal product (IMP) administration.

All participants received study treatment on Days 0, 28, and 56. Participants randomized to Group A received a dose of $6x10^5$ TCID50 MV-ZIKA on Day 0 and received placebo on Days 28 and 56; participants in Group B received a dose of 1 $x10^5$ TCID50 MV-ZIKA on Days 0, 28, and 56; participants in Group C received a dose of $6x10^5$ TCID50 MV-ZIKA on Days 0, 28, and 56; participants in Group D (placebo) received 0.6 mL of physiological saline on Days 0, 28, and 56.

Doses (volumes administered):

Group	Treatment
A	MV-ZIKA 0.6 mL ($6x10^5$ $TCID_{50}$/dose) Placebo 0.6 mL
B	MV-ZIKA 0.1 mL ($1x10^5$ $TCID_{50}$/dose)
C	MV-ZIKA 0.6 mL ($6x10^5$ $TCID_{50}$/dose)
D	Placebo 0.6 mL

The phase I clinical batch of this vaccine was present at a ready to use liquid formulation at a single concentration.

The mean (SD) age of participants in the safety/mITT population was 36.8 (10.7) years and 47 (97.9%) participants were Caucasian/White. The proportion of males (20 [41.7%]) was smaller than that of females (28 [58.3%]). Of the 28 females, 24 (85.7%) participants were of

childbearing potential. Demographic data for the PP population were similar to the safety/mITT population.

Participants were enrolled into the study from May to October 2017.

Immunogenicity Analysis:

The primary endpoint was immunogenicity on Study Day 56 confirmed by the presence of functional anti-zika antibodies as determined by a neutralizing antibody assay. An increase in Zika neutralizing antibody GMT was observed only in Group C (three high doses of MV-ZIKA [$6x10^5$ TCID50/dose]). The largest increase in neutralizing antibody GMT occurred after the second vaccination, increasing from 6.6 to 7.9; no increase occurred after the first vaccination and a reduction of 0.7 was observed after the third vaccination. No statistically significant difference was detected between Group C and placebo. The Group A regimen of one high dose of MV-ZIKA ($6x10^5$ TCID50/dose) and two placebos showed no increase in GMT. The Group B regimen of three low doses of MV-ZIKA showed no notable differences when compared with placebo (Group D). Assessment of the effect of baseline measles titer on GMT of neutralizing antibodies at Day 56 showed no effect.

Seroconversions were primarily observed in participants who received high dose MV-ZIKA ($6x10^5$ TCID50/dose). Overall, seven participants (one in Group A, one in Group B, and five in Group C) showed seroconversion after MV-ZIKA vaccination and one after placebo. Variations in seroconversion across all treatment groups during the study did not achieve statistical significance.

Participants who received the triple high-dose regimen in Group C experienced the greatest increase in Zika antibody GMT. The increases in GMT for Zika antibodies were notable and were sustained throughout the study. An increase of approximately 30% was observed after three high doses of MV-ZIKA, although the increase compared with placebo did not achieve statistical significance. Group B showed an increase in GMT for Zika antibodies after two vaccinations with low dose MV-ZIKA, which was of the same magnitude as the change seen in participants who received placebo.

Notable increases in GMT for measles antibodies were observed in all three MV-ZIKA treatment groups compared with placebo but the pattern of change differed. Geometric mean titer increased intensely after the first vaccination with high dose MV-ZIKA, in both Group A and Group C; the increase in Group C was sustained and finally achieved a 1.3-fold increase compared with baseline after the third vaccination. In Group B, GMT for measles antibodies did not started to increase until after the first vaccination; the increase was sustained after the second vaccination and stayed at the same level after the third vaccination.

Due to the low number of participants showing seroconversion in the ELISA tests, it was decided that evaluating the cell mediated immunity would not yield additional insights and hence no T-cell assay was performed.

Safety Analysis:

MV-ZIKA was well tolerated in this population of healthy men and women. Adverse events were predominantly mild or moderate. Only one subject experienced a severe solicited AE (fever) and five participants experienced severe unsolicited TEAEs (dysmenorrhea, arthralgia, hyperbilirubinemia, hypercholesterolemia, hyperlipidaemia and hypertriglyceridemia); all were judged unlikely or not related to IMP. No AE was judged serious.

Overall, 36 (75.0%) participants reported 200 solicited AEs and 39 (81.3%) participants reported 126 unsolicited AEs (TEAEs). The proportion of participants that experienced solicited and unsolicited AEs were higher in Group D (placebo) (100% and 87.5%, respectively) than Total ZIKA (70.0% and 80.0%, respectively). Only the proportion of participants that experienced unsolicited AEs in Group C (three high doses of MV-ZIKA [$6x10^5$ TCID50/dose]; 100%) were higher than Group D (87.5%).

Solicited local AEs occurred more frequently in participants who received MV-ZIKA than those who received placebo; more participants in Group C experienced solicited local AEs than Group A and Group B in all categories. The most frequently reported solicited local AE was injection site tenderness, followed by injection site pain. Injection site edema occurred only in Group C.

Solicited systemic AEs occurred across all treatment groups. The most frequently reported AE was headache; although the proportion of participants who experienced headache in Group D (placebo) was notably higher than any MV-ZIKA treatment group (62.5% versus 12.5–43.8%). Adverse events of myalgia, limb discomfort, rash and fever were only reported by participants who received MV-ZIKA.

The most frequently reported unsolicited AEs were nasopharyngitis and headache; the frequencies of these in participants who received MV-ZIKA were similar or less than those observed in participants who received placebo. A number of AEs were reported by participants who received MV-ZIKA but were not reported by those who received placebo. Most notably, these included general disorders and administration site conditions, and musculoskeletal and connective tissue disorders. Diarrhea was reported only by participants who received three high doses of MV-ZIKA (Group C).

Drug-related TEAEs were reported by 18 (37.5%) participants. The proportion of participants who experienced drug-related TEAEs was the highest in Group C (10 [62.5%]), although Total ZIKA showed the same percentage (37.5%) as placebo (37.5%) and no statistically significant

differences in AE rate were observed between any MV-ZIKA treatment group and placebo. The most frequently reported drug-related AE was diarrhea, which occurred only in Group C; five (10.4%) participants experienced six episodes. No subject who received single dose of MV-ZIKA or placebo experienced diarrhea. The second most frequently reported drug-related AE was vertigo, which was distributed evenly across groups (one each in Groups A, B and D).

The proportion of participants who experienced medically attended TEAEs was higher in Group D (50.0%) than any MV-ZIKA treatment group (25.0–31.3%) but no statistically significant differences in AE rate were observed between any MV-ZIKA treatment group and Group D. Overall, 15 (31.3%) participants experienced medically attended TEAEs. The most frequently reported were arthralgia, dizziness, abdominal pain upper, hepatic steaosis and oropharyngeal pain, each reported by two participants.

The proportion of participants who experienced TEAEs for which action was taken was notably higher in those who received placebo (87.5%) than MV-ZIKA (31.3–37.5%). There were statistically significant differences in AE rate in Group B versus Group D and Group C versus Group D. The most frequently reported AEs with action taken were nasopharyngitis (five [10.4%] participants) followed by headache (four [8.3%] participants). All participants who reported unsolicited AEs for which action was taken took medicines except one subject who had a broken tooth and received an amalgam filling, one subject where action taken was unknown and one subject who had increased liver values and was supervised by the general practitioner.

No trends or changes were observed in hematology, coagulation or urinalysis. A small increase in mean ALT was observed in Group C. Four participants had abnormal clinical laboratory evaluations that were considered to be of clinical relevance but unlikely related to the study drug.

No changes in vital sign parameters were considered clinically significant. No abnormalities in vital sign parameters were reported directly as AEs, although one subject experienced an AE of fever and one an AE of tachycardia. Nine participants reported 13 events of presented symptoms. The most frequently reported symptom was flu-like symptoms (one subject at Visit 3 and four participants at Visit 5). One subject reported excessive fatigue. None of the participants reported any symptom of fever, nausea vomiting.

More participants who received MV-ZIKA experienced local tolerability symptoms than placebo. The main local symptoms experienced were tenderness and injection site pain. No pruritus or edema was presented by any subject after injection.

4.5 Study Rationale

This study is designed to investigate, at first, safety and tolerability of a novel liquid vaccine formulation named MV-ZIKA-RSP, in healthy adults aged 18 to 55 years.

For this purpose, two different doses of MV-ZIKA-RSP ($1x10^5$ $TCID_{50}$ and $2,5\ x10^4$ $TCID_{50}$ per dose), and one group receiving only placebo, will be assessed in healthy adults for one or two immunizations and at different time points.

The induction of functional anti-zika-rsp antibodies by MV-ZIKA-RSP will be investigated, by means of immunoassays like VNT and supported with assays like ELISA and T-cell analysis. In our previous clinical studies, it is demonstrated that the timepoints day 28 and day 56, after immunisation, are suitable for elicitation of immunogenicity.
The different vaccination schedules:

- two MV-ZIKA-RSP vaccinations (high dose) on days 0 and 28
- two MV-ZIKA-RSP vaccination (low dose) on day 0 and 28
- one MV-ZIKA-RSP vaccination (high dose) on day 0 and placebo on day 28
- two placebo injections on day 0 and 28

will allow us a comparison between immunogenicity of these dosing regimens. A single shot or two vaccinations within a short period of time are desirable to allow for a quick immunisation before travelling to areas with active transmission of the zika virus and at the same time will reveal valuable information on future treatment recommendations (single shot treatment vs. double immunisation).

With a design of randomised, observer-blinded study we will be able to record and assess all adverse events.

Since the induction of virus specific T cells is a critical step in the generation of a functional immune response, the rate of zika virus specific T cells will be analysed *in vitro.*

This study was designed according to the Note for Guidance on Clinical Evaluation of New Vaccines (CHMP/VWP/164653/2005), and will be conducted in compliance with the protocol, Good Clinical Practice (GCP) as set forth in the International Council on Harmonisation (ICH) guidelines on GCP (ICH E6), and applicable local regulatory requirements.

4.6 Benefit/Risk Assessment

General

The measles virus vector is well known. Possible risks that are frequently associated with vaccination are the occurrence of local reactions e.g. edema, induration and erythema, transient local pain or tenderness at the injection site as well as mild to moderate headache, myalgia, flu-like symptoms or fatigue.

Currently no vaccines against Zika virus are commercially available and few vaccines are in pre-clinical or clinical testing.

The MV-ZIKA-RSP vaccine is based on the measles virus technology. [redacted], chemically synthesized and inserted into the Schwarz vaccine strain of measles virus (MV) to produce the candidate vaccine MV-ZIKA-RSP. Importantly, no live attenuated Zika virus is used during generation or manufacture of MV-ZIKA-RSP vaccine. Thus, induction of Zika like symptoms are not expected. However, the participants will be monitored for occurrence of adverse events and tolerability findings.

As any other vaccine, MV-ZIKA-RSP might induce allergic and anaphylactic reactions, apart from the described local reactions at the vaccination site and systemic flu-like reactions. In rare cases, the injection can lead to a vasovagal reaction immediately after injection of the vaccine. The needle pricks for blood sampling may also cause local reactions such as edema. The participants cannot expect direct benefit from study participation, except for potential boosting of measles immunity and mounting immunity to Zika virus.

Potential Benefit

If the clinical development of MV-ZIKA-RSP is successful, this vaccine might help people, who are at risk of acquiring an infection with Zika virus and might help to prevent congenital effects occurring in infants of infected mothers.

A boost in measles immunity may be an additional benefit. The impact of pre-existing immunity is a major point of interest for Measles-vectored vaccines, as large parts of the population have been vaccinated at least once, while some have undergone natural infections. In previous phase I (Ramsauer et al. 2015) and phase II (Reisinger et al. 2018) clinical trials we have shown that the immunogenicity of another vaccine using the same vector (MV-CHIK) is not influenced by the levels of Measles-specific serum IgG before vaccination. In addition, we found that the frequency of Adverse Events (AEs) in participants with low (<200 iU/l) and participants with high (≥200 U/l) frequency of anti-Measles IgG is similar (p=0.0658 when comparing unsolicited AEs, p=1.0000 when comparing solicited AEs).

Conclusion

Considering the small risk of adverse events and the potential benefits if immunogenicity can be shown, the benefit risk ratio is considered positive and the clinical study is justified.

5. TRIAL OBJECTIVES AND MEASUREMENTS

5.1 Primary Objective

- To investigate safety and tolerability of MV-ZIKA-RSP, a novel liquid vaccine formulation, up to day 56 after first immunization consisting in one or two vaccinations

Safety and tolerability will be assessed by the rate adverse events (AEs) after the first vaccination up to day 56.

5.2 Secondary Objectives

- To investigate long-term safety, up to study day 182, after first immunization

Long term safety and tolerability will be assessed by means of a phone call where participants will be asked about any adverse event (solicited, unsolicited or serious adverse event) experimented after the Visit 3 (day 56)

- To investigate immunogenicity up to day 56 after first immunization consisting in one or two vaccinations with different doses

To cover this objective the presence of functional anti-zika antibodies will be determined by means VNT and ELISA at the following time points: Visit 1 (day 0), Visit 2 (day 28) and Visit 3 (day 56) after first vaccination

- To get information about the optimal dose of MV-ZIKA-RSP vaccine regarding safety, tolerability, and immunogenicity

An evaluation of the optimal dose will be determined after analysis of the safety, tolerability, and immunogenicity (VNT)

- To investigate specific cell mediated immunity induced by all different treatment groups

Cell mediated immunity up to day 56 will be assessed by T cell analysis of PBMC isolated on study days 0, 28 and 56

6. TRIAL DESIGN

6.1 Overall Study Design

This is an observer-blinded, block-randomised, dose finding, phase I trial, comparing different dose levels of MV-ZIKA-RSP to evaluate safety, tolerability and immunogenicity, of this novel ZIKA-RSP vaccine. Placebo (physiological saline solution) will be applied to blind the different treatment schedules.

The study will be conducted at a single study site, Medical University Vienna and is registered online at http://www.ClinicalTrials.gov under EudraCT number 2019-000840-93.

After the screening procedures, 48 healthy male and female volunteers aged 18-55 years will be randomly assigned to one of four treatment groups (A, B, C or D). Participants will be assessed for immunogenicity on days 0, 28 and 56 (treatment period), as confirmed by the presence of functional anti-zika-rsp antibodies determined by (VNT) and by ELISA, at the same time safety will be also assessed. After the treatment period, participants will be call by phone (day 182) for evaluation of safety follow-up.

The investigator and site personnel assessing AEs, all participants, as well as one of the sponsor's representatives involved in the monitoring and conduct of the study will be blinded to which vaccine was administered. Only the unblinded monitor, site personnel performing randomisation and preparation of IMP will be unblinded.

Study duration per subject will be 182 days (6 months), consisting of 4 weeks treatment period (study day 0 till study day 28) and 22 weeks follow up period (study day 2 till study day 182)

6.2 Study Endpoints

6.2.1 Primary Endpoint

- Rate of adverse events (AEs) up to study day 56

6.2.2 Secondary Endpoints

- Rate of solicited and unsolicited AEs, as well as, reported serious adverse events (SAEs) up to study day 182 (long-term safety) compared between the 4 different treatment groups.

- Immunogenicity on study days up to study day 56 as confirmed by the presence of functional anti-zika antibodies as determined by VNT and by ELISA

- Cell-mediated immunity specific for ZIKA-RSP, up to study day 56, confirmed by the presence of specific functional $CD4^+$ and $CD8^+$ T-cells
- Safety laboratory parameters (haematology, serum chemistry, urinalysis)

6.3 Treatment Groups

After the screening procedure, 48 participants eligible for the study will be randomised to one of the following 4 treatment groups:

Group A:

two vaccinations with MV-ZIKA-RSP, high dose on day 0 and day 28

Group B:

two vaccinations with MV-ZIKA-RSP, low dose on day 0 and day 28

Group C:

one vaccination with MV-ZIKA-RSP, high dose, on day 0 and placebo on day 28

Group D:

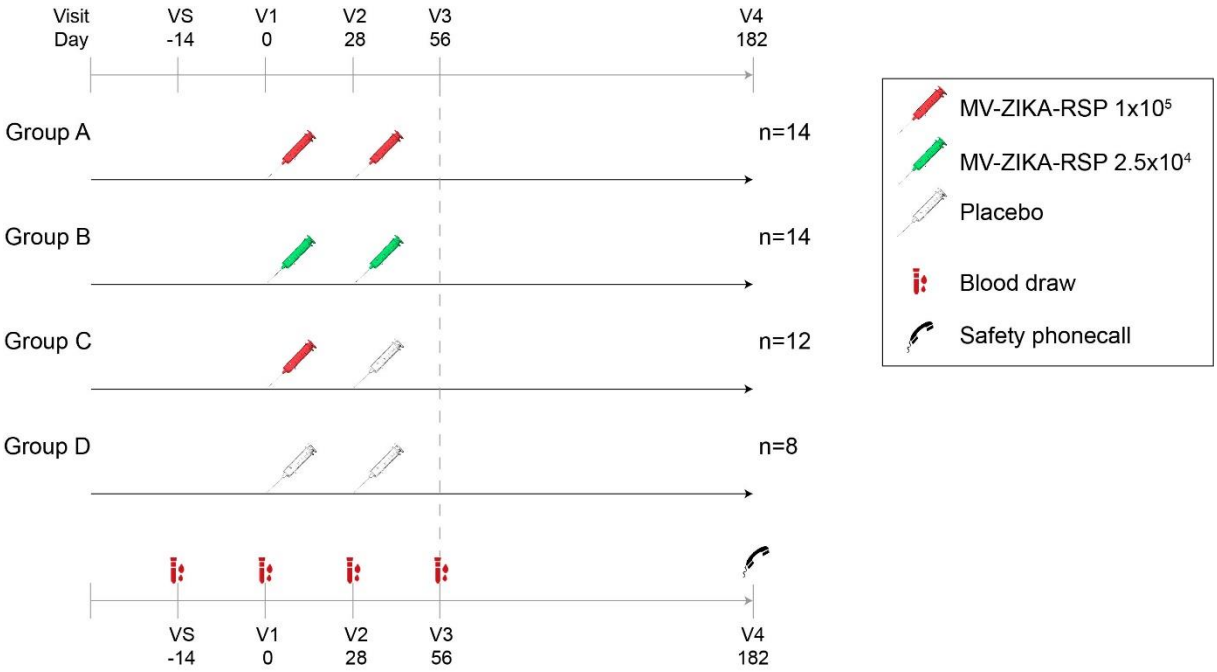
placebo on day 0 and day 28

Table 1. Treatment groups and vaccination schedule

Group	Treatment	Dose	Schedule	Participants
A	MV-ZIKA-RSP high dose	1 x10^5 $TCID_{50}$	days 0 & 28	14
B	MV-ZIKA-RSP low dose	2,5 x10^4 $TCID_{50}$	days 0 & 28	14
C	MV-ZIKA-RSP high dose + Placebo	1 x10^5 $TCID_{50}$ + Placebo	MV-ZIKA-RSP high dose day 0 + Placebo day 28	12
D	Placebo	Placebo	Placebo day 0 + Placebo day 28	8

The actual vaccine dosages are dependent on the manufacturing process, specifying and allowing a window of ±0.5 log.

6.4 Study Flowchart



6.5 Study Procedures

Table 2. Study procedures

	Screening	Treatment				Post Treatment Safety Follow Up	ET
Visit	**VS**	**V1**	Post dose safety FU	**V2**	**V3** Primary Endpoint	**V4**	(1)
Study Day	-14 to -1	0		28± 3	56 ±3	182 ±14	
Informed Consent	x						
Inclusion/Exclusion Criteria	x	x					
Medical History	x	x					
Vaccination History	x	x					
Vital Signs (2)	x	x		x	x		x

Symptom-directed Physical Examination		x		x	x		x
System-based Physical Examination (6)	x	x		x	x		x
Adverse Events (3) (7)		x	x	x	x	x	x
Prior/Concomitant Medication (4)	x	x	x	x	x	x	x
Clinical Laboratory (5)	x			x	x		x
Randomization		x					
Urine Pregnancy Test (12)	x	x		x	x		x
Study Treatment		**x**		**x**			
Immunogenicity (VNT) (12)		x		x	x		x
T-cell immunity (PBMCs) (12)		x		x	x		x (11)
Zika antibody (ELISA) (12)		x		x	x		x
Measles antibody (ELISA) (12)		x		x	x		x
Local Tolerability (7)		x		x	x		x
Dispense subject diary		x		x			
Collect and review subject diary				x	x		x
Safety phone call (9)			x (9)			x (10)	

(1) Early termination visit should – if possible - be conducted 28±3 days after last IMP administration.
(2) Systolic/diastolic blood pressure, pulse rate, body temperature
(3) Conditions/symptoms noted prior to randomization should not be reported as AE, but should be reported as medical history
(4) Medications administered up to 30 days prior to screening until end of study participation should be documented
(5) The clinical laboratory comprises: Hematology (hemoglobin, hematocrit, erythrocyte count, differential white blood count, platelets), Chemistry (creatinine, potassium, sodium, calcium, AST, ALT, alkaline phosphatase, bilirubin), Coagulation (prothrombin time (PT), activated partial thromboplastin time (aPTT), fibrinogen), Urinalysis (standard urine test stick for determining pH-value, glucose, protein, bilirubin, urobilinogen, red blood cells, white blood cells, nitrite, ketone and specific gravity), Virology (only at screening; HBs-Ag, anti-HBc-Ab, anti-HCV-Ab, HIV 1/2 Ab/HIV-1 Antigens if not done within 30 days before screening)
(6) To be performed if necessary, according to findings in symptom-directed physical examination
(7) To be performed 1 hour after each IMP administration and additionally at the subsequent visit (prior to IMP administration, if applicable)
(8) Participants should be interviewed regarding potential AEs since the last study visit. If indicated, participants should be called in for an unscheduled visit.
(9) The post dose safety call follows up should be conducted 7±1 days (one week) after the first vaccination.
(10) The post treatment safety call follows up should be conducted 182 days (6 months) after the first study treatment also in case of an early termination.
(11) The amount of blood sample taken during the early termination (ET) visit depends on the time point of the visit. If the ET-visit replaces the primary endpoint visit on day 56±3, 48 to 60 mL blood should be taken; otherwise 24 to 48 mL are enough.
(12) Urine for pregnancy test should be taken before vaccination

7. PARTICIPANT IDENTIFICATION

7.1 Trial Participants

Healthy male and female volunteers will be screened for inclusion and exclusion criteria and randomised to one of 4 treatment groups in case of eligibility.

Forty-eight participants will be enrolled if all the following inclusion criteria and none of the following exclusion criteria apply:

7.2 Inclusion Criteria

1. Signed informed consent obtained before any trial-related activities
2. Healthy men or women aged 18 to 55 years on the day of consenting
3. Ability to comprehend the full nature and purpose of the study, including possible risks and side effects; ability to cooperate with the investigator and to comply with the requirements of the entire study
4. All female participants must have a negative urine pregnancy-test at screening
5. Willingness not to become pregnant or to father a child during the entire study period by practicing reliable methods of contraception as specified in protocol section 8.11.4
6. Availability during the duration of the trial
7. Normal findings in medical history and physical examination or the investigator considers all abnormalities to be clinically irrelevant
8. Normal laboratory values or the investigator considers all abnormalities to be clinically irrelevant (unless otherwise specified in exclusion criteria)

7.3 Exclusion Criteria

The subject may not be enrolled in the study if any of the following applies:

1. Participation in another clinical study (including exposure to an investigational medicinal product ore device) within one month before the screening visit or planned concurrent participation in another clinical study before completion of the treatment period (day 56)
2. History of immunodeficiency, known human immunodeficiency virus (HIV) infection or current hepatitis B/C infection
3. Strong anamnestic evidence for or confirmed history of or current infection with Zika- or Dengue-virus
4. History of drug addiction including alcohol dependence within the last 2 years

5. Inability or unwillingness to avoid intake of more than around 20g alcohol per day during 48 hours after each vaccination (equals roughly 0.5 L beer or 0.25 L of wine)
6. Vaccination within 4 weeks prior to first vaccination or planning to receive any non-study vaccine until end of treatment period (day 56)
7. Prior receipt of any Zika or Chikungunya vaccine
8. History of moderate or severe arthritis or arthralgia within the past 3 months prior to Screening Visit
9. Recent infection within 1 week prior to Screening Visit (possibility of deferral)
10. Blood donations including plasma donations, 90 days prior to Screening Visit and anticipated blood, plasma, tissue, sperm, or organ donation, throughout the study until end of treatment period (day 56)
11. Clinically relevant history of renal, hepatic, gastrointestinal, cardiovascular, respiratory, skin, haematological, endocrine, inflammatory, autoimmune, or neurological diseases or clinically relevant abnormal laboratory values, that in the opinion of the investigator may interfere with the aim of the study
12. History of neoplastic disease (excluding non-melanoma skin cancer that was successfully treated) within the past 5 years or a history of any haematological malignancy
13. Behavioural, cognitive, or psychiatric condition that in the opinion of the investigator affects the ability of the participant to understand and cooperate with the study protocol
14. History of severe adverse reactions to vaccine administration, including anaphylaxis and related symptoms, such as urticaria, respiratory difficulty, angioedema and abdominal pain to vaccines, or history of allergic reaction likely to be exacerbated by any component of the vaccine
15. History of anaphylaxis to drugs or other allergic reactions, which the investigator considers compromising the safety of the volunteer
16. Abnormal laboratory values which, at the discretion of the investigator should lead to the exclusion of the subject
17. Use of medication during 2 weeks before the first vaccination and throughout the study, which the investigator considers affecting the validity of the study, except hormonal contraception or hormonal replacement therapy in female participants. (Prior to taking any medication within 72 h before study vaccination, the subject should consult the investigator)
18. Use of immunosuppressive drugs like corticosteroids (excluding topical preparations) within 30 days prior to first IMP administration, or anticipated use until completion of the end of treatment visit Receipt of blood products or immunoglobulins within 120 days prior to the Screening Visit or anticipated receipt of any blood product or immunoglobulin before completion of the treatment period (day 56)

19. Pregnancy (positive pregnancy test at screening or during the treatment period) or lactation at screening, or planning to become pregnant during the treatment period
20. Unreliable contraception methods (for details please refer to protocol section 8.11.4)
21. Persons in a direct relationship with the sponsor, an investigator or other study team members. Direct dependent relationships include close relatives (i.e. children, parents, partner/spouse, siblings) as well as employees of the study site or the sponsor

8. TRIAL PROCEDURES

8.1 Informed Consent (AMG § 38)

In obtaining and documenting informed consent, the investigator must comply with the applicable regulatory requirement(s) and adhere to ICH guideline for GCP, the requirements in the Declaration of Helsinki and the EU directive 2005/28/EC.

Before any trial specific procedures are performed participants will be informed about the exact nature of the trial, what it will involve for the participant, all procedures as determined by the clinical protocol, the known side effects and any risks involved in taking part. It will be clearly stated that the subject is free to withdraw from the trial at any time for any reason without prejudice to future care, without affecting their legal rights and with no obligation to give the reason for withdrawal.

The participant will be allowed as much time as needed to consider the information, and the opportunity to question the investigator or other independent parties to decide whether they will participate in the trial.

Written informed consent will then be obtained by means of participant's dated signature and dated signature of the qualified and experienced person who presented and obtained the informed consent. A copy of the signed informed consent will be provided to the subject and the original signed form will be retained at the site.

If information becomes available that may be relevant to the subject's willingness to continue participating in the trial, the investigator must inform the subject in a timely manner, and a revised written informed consent must be obtained.

8.2 Randomisation

At Visit 1 (day 0) 48 eligible participants will be randomly assigned to one of four treatment groups (A, B, C or D) by means of randomisation envelopes provided by data management.

Each subject will be assigned a unique three-digit randomisation number in ascending order, by opening the randomisation envelope with the lowest free (unassigned) randomisation number available. Randomisation should be performed as late as possible, i.e. when knowledge of the assigned treatment group becomes necessary. Only trained members of the unblinded study team will be authorised to open one envelope per subject, containing the information about the allocated treatment group.

Upon opening, the randomisation envelope shall be signed with date and time of randomisation and the corresponding subject number.

Please note:

As this is a first in humans' study, the site should initially enroll a group of six participants and then pause with enrollment until these participants completed their Post Dose Safety Follow Up one week after the first vaccination. If no events as defined in section 8.9.1 "Safety Stopping Rule" are identified, the site should resume enrolling participants. If such events were identified, the investigators and the sponsor need to assess the risk of continuing the study and based on this assessment to decide if or if not, the study should be continued.

The 48 participants will be distributed between the 4 groups using a randomisation ratio of 14 : 14 : 12 : 8 = A : B : C : D, with an adequate representation of high dose, low dose and placebo subjects (2 : 2 : 0 : 2 = A : B : C : D) within the first 6 subjects to allow for the safety evaluation after the first vaccination.

A subject will be considered enrolled once the first IMP administration has been performed according to randomisation.

8.3 Blinding and Code-breaking

As this study will be conducted in an observer-blinded manner, unblinded study team members responsible for randomisation, monitoring (unblinded monitor) and preparation of IMP, will otherwise not be involved in the conduct of the study.

All participants, the investigator and site personnel performing study related assessments, as well as the sponsor's representatives involved in the monitoring (blinded monitor) and conduct of the study, will be blinded to treatment assignment.

Besides one set of randomisation envelopes, the site will receive an additional set of emergency envelopes stored in the ISF for unblinding purposes. As the knowledge of the subject's treatment might become mandatory (e.g. in case of emergency) the investigator has the possibility to open the emergency envelope identified with the subject's randomisation number and unblind the subject.

Please note that this code-breaking should only be done by the investigator if the knowledge of the subject's treatment influences the decision on further procedures. If unblinding would not make any difference for further treatment, the study team should remain blinded.

8.4 Screening Procedures

8.4.1 Screening Visit (VS, 1-14 days prior to Visit 1)

After participants have signed the informed consent form, the screening visit will be performed within two weeks prior to Visit 1.

During this visit, all participants will be checked for inclusion and exclusion criteria, undergo a full physical examination and evaluation of vital signs (systolic and diastolic blood pressure, pulse rate and body temperature). Medical history, vaccination history (covering the last three years prior to screening), prior/concomitant medication within 30 days prior to screening and demographic data will be recorded.

Blood and urine samples will be collected for clinical laboratory assays (HIV, hepatitis B and C, haematology, coagulation parameters, clinical chemistry and urinalysis). HIV tests with available results performed up to 30 days before screening are acceptable. A urine pregnancy test will be performed in women of childbearing potential. For details on laboratory sampling and testing, see section 8.11.1.

All participants eligible for the clinical study will be asked to return to the study site within 14 days after the screening visit for Visit 1.

8.5 Treatment Period

8.5.1 Visit 1 (day 0)

Participants found to be eligible will return to site for Visit 1 within 14 days after the screening visit. Inclusion and exclusion criteria will be checked again and (in case of changes since the screening visit) the medical history will be updated. Symptom-directed physical examination, system-based assessment (only if necessary, according to findings in symptom-directed physical exam) and vital signs (systolic and diastolic blood pressure, pulse and body temperature) will be recorded. In addition, the participants will be asked regarding changes in concomitant medications since the last visit.

Blood samples for baseline immunogenicity assessments (Zika antibody ELISA, VNT) will be drawn from an appropriate forearm or cubital vein. For T-cell assays blood samples will be collected and peripheral blood mononuclear cells (PBMCs) will be prepared. A urine pregnancy test will be performed in women of childbearing potential. For details on laboratory sampling and testing, see section 8.11.

Participants will be randomised to treatment group A, B, C or D and receive either the first MV-ZIKA-RSP vaccination or placebo, according the assigned randomized treatment group.

The participants' diaries will be explained and handed over. Participants will be asked to report their body temperature and any adverse event in these diaries for 7 days, starting 6 hours after the injection.

After observation of the participants for 1-hour, local reactions and adverse events (if any occurred) will be recorded.

Blood pressure and pulse rate will be checked, and participants will be discharged only if the investigating physician considers it to be safe for them to leave the department.

The participants are invited to return to study site for Visit 2 on study day 28 (± 3 days).

8.5.2 Post Dose Safety Follow-up (day 7 ±1 days)

The investigator will ensure phone contact with the participants one week after first vaccination was performed and ask about any new concomitant medications or AEs occurred since the last visit. If deemed necessary, the investigator may ask participants to come to study site for an unscheduled visit to assess severity of the AEs.

8.5.3 Visit 2 (day 28 ±3 days)

The investigator or designee will examine local tolerability (of previous vaccination site) and ask about any new concomitant medications or AEs occurred since the last visit.

In addition, symptom-directed physical examination, system-based assessment (only if necessary, according to findings in symptom-directed physical exam) and vital signs (systolic and diastolic blood pressure, pulse rate and body temperature) will be recorded.

A urine pregnancy test will be performed in women of childbearing potential.

Blood samples will be taken for clinical chemistry, haematology, and coagulation parameters.

Additional blood samples for immunogenicity assessments (Zika and Chikungunya ELISA, VNT) will be drawn from an appropriate forearm or cubital vein. For T-cell assays blood samples will be collected and PBMCs will be prepared. For details on laboratory sampling and testing, see section 8.11.

Participants will receive their second vaccination either with MV-ZIKA-RSP, or placebo, according to the assigned randomised treatment group.

The participants' diaries will be explained and handed over. Participants will be asked to report their body temperature and any adverse event in these diaries for 7 days, starting 6 hours after the injection.

After observation of the participants for 1-hour, local reactions, and adverse events (if any occurred) will be recorded.

Blood pressure and pulse rate will be measured, and participants will be discharged only if the investigating physician considers it to be safe for them to leave the department.

The participants are invited to return to study site for Visit 3 on study day 56 (± 3 days).

8.5.4 Visit 3 (day 56 ±3 days)

The investigator or designee will examine and document local tolerability (of the previous vaccination site) and ask about any new concomitant medications or AEs occurred since the last visit.

Symptom-directed physical examination, system-based assessment (only if necessary, according to findings in symptom-directed physical exam) and vital signs (systolic and diastolic blood pressure, pulse rate and body temperature) will be recorded.

A urine pregnancy test will be performed in women of childbearing potential.

Blood samples will be taken for clinical chemistry, haematology, coagulation parameters and urine samples will be collected for urinalysis.

Additionally, blood samples for immunogenicity assessments (Zika and Chikungunya ELISA, VNT). For T-cell assays blood samples will be collected and PBMCs will be prepared. For details on laboratory sampling and testing, see section 8.11.

Upon Visit 3, the treatment period of this study will be completed.

Participants are informed about the approximated date for a safety follow up call for follow up Visit 4 on study day 182 (± 14 days).

8.6 Post Treatment Safety Follow up Period

8.6.1 Visit 4 (day 182 ±14 days)

Participants will be call and will be ask if they experienced any new or changes in adverse events (AE) or concomitant medications since the last visit.

8.7 Early Termination Visit (ET)

Participants who are withdrawn or wish to discontinue the study prematurely for any reason shall be asked to attend an early termination visit.

If the early termination occurs within the treatment period (at or before Visit 4), participants shall perform all procedures as defined for Visit 3. If the early termination occurs within the follow up period (after Visit 3), participants shall perform all procedures defined for Visit 4.

8.8 Unscheduled Visits

Unscheduled visits can be conducted whenever deemed necessary. The extent of the examination performed at an unscheduled visit is at the discretion of the investigator. All procedures completed during an unscheduled visit should be documented in the source data and should also be entered to the electronic case report form (eCRF).

8.9 Discontinuation/Withdrawal of Participants

Participants may withdraw from participation in the study at any time, without the need to justify. If a subject withdraws or is withdrawn prior to completion of the study, the reason for this decision should be recorded in the electronic case report forms (eCRFs) although it is recognised that the subject may refuse to provide a reason.

Generally, any enrolled/vaccinated subject may withdraw or be withdrawn from the study for the following reasons:

- The subject withdraws consent
- The subject fails to comply with the requirements of the protocol
- At the discretion of the investigator
- For an adverse event, which at the discretion of the investigator requires discontinuation of the subject or results in inability to continue to comply with trial procedures
- if the administration of a drug, which is not permitted by the study protocol, becomes necessary (this should be discussed and agreed between the investigator and the sponsor)
- violation of exclusion criteria 1, 4, 5, 7, 12, 15, 16, 17, 18, 19 or 20 during the study (this should be discussed and agreed between the investigator and the sponsor)

All participants who withdraw or are withdrawn prematurely from the study will undergo an early termination visit (see section 8.7).

8.9.1 Safety Stopping Rules

Stopping rules are established at subject level and entire study level.

Individual Participants Stopping Rules

The following are specific criteria for discontinuing individual participants from further vaccination, but not from completing scheduled follow-up assessments, unless the participant is explicitly withdrawn from the study:

- Participants who develop serious adverse reaction (SAR) at the discretion of the investigator
- Participants who develop a potentially life-threatening (grade 4) solicited systemic adverse reaction that occurs without alternative etiology within 7 days after the study vaccination
- Participants who develop a medical condition for which continued participation, in the opinion of the investigator, would pose a risk to the subject or would be likely to confound interpretation of the results
- Participants who become pregnant during the treatment phase of the study
- Participants who experience anaphylaxis within 24 hours or generalised urticaria within 72 hours after administration of the study vaccine

If any of these occur, appropriate measures to treat the subject will be taken and the sponsor or the sponsor designee will be notified immediately.

Entire Study Stopping Rules

The study will be stopped (no new enrolments and no additional vaccine administered) pending a safety review by the investigator and the sponsor, if any of the following occurs:

- One or more participants experience a serious adverse reaction (SAR), based on the current reference safety information (Investigator's Brochure)
- One or more participants' deaths assessed by the investigator, safety manager or sponsor as related to investigational product
- One or more participants develop laryngospasm, bronchospasm or anaphylaxis within 24 hours of vaccine administration
- Two or more participants develop injection-site ulceration, abscess or necrosis
- Two or more participants experience generalised urticaria within 72 hours after administration of study product

Upon completion of this review, the sponsor will determine if study enrolment or treatment should be discontinued, or if study enrolment and treatment may continue according to the protocol.

Irrespective of the stopping rules the study may be temporarily suspended or prematurely terminated at any time if there is sufficient reasonable cause, and if agreed to by both the investigator and the sponsor as being in the best interests of participants and justified on either medical or ethical grounds. In terminating the study, the sponsor, the CRO, and the investigator will ensure that adequate consideration is given to the protection of the participants' interests.

Whenever the study is stopped the competent authorities and ethical committees will be informed.

In case the entire study has to be discontinued prematurely, all concerned participants should be asked to perform an early termination visit.

8.9.2 Replacement of Participants

Participants who are discontinued before randomisation will be regarded as screening failures and screening will be continued until 48 participants will be randomised.

Participants who withdraw or are withdrawn after randomisation will not be replaced. This may result in a decreased number of participants in the predefined sample size.

8.10 End of Study

The end of study will be defined as the date of the last visit of the last subject. The study will then be stopped. For premature termination or suspension of the study, refer to section 8.9.1

8.11 Handling of Samples

The procedures of sample collection, preparation and storage will be described in detail in a study specific Lab-Manual. Blood sampling kits and labels will be supplied by the supply vendor. Timepoints and blood volumes are summarised in Table 3.

8.11.1 Samples for Safety Laboratory Parameters

Safety laboratory parameters will be analysed by local laboratories according to applicable laboratory SOP.

The following parameters will be measured to evaluate inclusion and exclusion criteria only once at screening visit (SV).

- Virology: Hepatitis B surface-Ag, anti-Hepatitis B core-Ab, anti-Hepatitis C-Ab, HIV Ab. HIV tests that were performed up to 30 days before screening and where results are available are acceptable and do not require a new test.

The following laboratory parameters will be assessed for subject's safety at screening (VS), Visit 2, Visit 3 and additionally at Early Termination (if applicable) as depicted in Table 2.

- Haematology: haemoglobin, haematocrit, erythrocyte count, differential and total white blood count, platelets
- Chemistry: creatinine, potassium, sodium, calcium, AST, ALT, alkaline phosphatase, bilirubin
- Coagulation parameter: prothrombin time (PT), activated partial thromboplastin time (aPTT), fibrinogen
- Urinalysis: a standard urine test dipstick for determining pH-value, glucose, protein, bilirubin, urobilinogen, blood, leucocytes esterase, nitrite, ketone and specific gravity will be used.
- Laboratory values deviating from the normal ranges defined by the local laboratory will be evaluated by the investigator. Clinically relevant deviations have to be reported as adverse events and graded according to the FDA Guidance for Industry (Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials, Sept. 2007)

8.11.2 Immunogenicity Samples

Virus Neutralization Test (VNT)

Humoral systemic immune response will be determined by anti-zika-rsp virus neutralisation test. The virus neutralisation titre (VNT) will be measured to identify immunogenicity in sera of all participants.

At Visits 1, 2, 3 and ET (if applicable) a volume of 8ml blood will be collected, to isolate serum and freeze a minimum of three aliquots (0.5 ml /vial) at -80°C.

Enzyme linked immunosorbent assay (ELISA)

In addition, humoral systemic immune response mediated by serum IgG antibodies against zika-rsp will be determined by IgG ELISA.

At Visits 1, 2, 3 and ET (if applicable) a volume of 8 ml blood will be collected, to isolate serum and freeze a minimum of three aliquots (1 ml /round bottom cryovial) at -80°C for determination of anti-zika antibodies by ELISA.

Anti-measles antibodies will be determined by ELISA out of the same samples collected at Visits 1, 2, 3 and ET (if applicable). Therefor no additional blood sapling will be required for this assay.

T cell Analysis

Cellular immunogenicity will be determined by evaluation of T cell immune response. Therefore, PBMCs will be isolated from 24-48 ml of fresh collected blood within 8 hours after blood draw, aliquoted in approximately 3-5 cryovials (~1 ml /vial), to an approximated concentration of 10x 10^6 cells per vial, frozen at -80°C, and storage in N_2, for further analysis. The T cells will be re-stimulated in vitro with zika virus structural protein peptides to determine the number of zika-virus specific T cells after one or two immunisations.

At Visits 1, 2, 3 and ET (if applicable) a volume of 24-48 ml blood will be collected for isolation of peripheral blood mononuclear cells (PBMCs).

Table 3. Blood sample collection

Test	Type of Vials	Volume ml	Visits	Total Volume ml
Virology (HIV, HBV, HCV)	serum	* 0	VS, V2, V3	* 0
Haematology	EDTA	4 - 5	VS, V2, V3	12-15
Coagulation	citrate tubes	4 - 5	VS, V2, V3	12-15
Clinical Chemistry	serum	8	VS, V2, V3	24
VNT	serum	8	V1, V2, V3	24
ELISA ZIKA	serum	8	V1, V2, V3	24
ELISA measles	serum	** 0	V1, V2, V3	** 0
PBMCs for T-cell Analysis	citrate tubes	24-48	V1, V2, V3	96
				192-198

* 0 ml: Virology testing at screening, V2 and V3 will be performed from the same sample as Clinical Chemistry, no additional blood draw will be necessary

** 0 ml: ELISA for determination of measles antibodies will be performed from the same sample as the ELISA for ZIKA-Ab, no additional blood draw will be necessary

Total blood volume collected during the study duration per subject (182 days/6 months) will be approximately –192-198 ml.

8.11.3 Pregnancy Test – Contraception

For females of childbearing potential, urine pregnancy tests will be performed at all scheduled visits and during the V4-phone call, participants will be ask whether they are pregnant. Only females with a negative urine test will be allowed to receive study vaccinations.

At visits where IMP is to be administered, a negative urine test outcome must be available prior to administration of study treatment.

Participants with a positive urine pregnancy test at Screening Visit cannot be enrolled into the study. In case of pregnancy during the study the investigator will inform the sponsor immediately by completing and sending the applicable form (Section 10.6). The participant should attend planned visits without further vaccinations for follow-up. If at least one dose was received, participants will be monitored until pregnancy outcome.

Contraception

Birth control methods which may be considered as highly effective are those that can achieve a failure rate of less than 1% per year when used consistently and correctly are considered as highly effective birth control methods. Such methods include:

_ combined (estrogen and progestogen containing) hormonal contraception associated with inhibition of ovulation:

- Oral
- Intravaginal
- transdermal

_ progestogen-only hormonal contraception associated with inhibition of ovulation:

- oral
- injectable
- implantable

_ intrauterine device (IUD)

_ intrauterine hormone-releasing system (IUS)

_ bilateral tubal occlusion

_ vasectomised partner

_ sexual abstinence

Acceptable birth control methods which may not be considered as highly effective are those that result in a failure rate of more than 1% per year include:

_ progestogen-only oral hormonal contraception, where inhibition of ovulation is not the primary mode of action

_ male or female condom with or without spermicide

_ cap, diaphragm, or sponge with spermicide

Birth control methods which are considered unacceptable include:

Periodic abstinence (calendar, symptothermal, post-ovulation methods), withdrawal (coitus interruptus), spermicides only, and lactational amenorrhoea method (LAM) are not acceptable methods of contraception. Female condom and male condom should not be used together

8.11.4 Labelling, Storage and Transport of Samples

Please refer to the study specific Lab-Manual for detailed information about labelling, storage and transport of study serum samples vials and blood for PBMC isolation.

Labelling of Samples

Each sample tube will be clearly and unequivocally identified by a label resistant to the storage temperature which contains at least the following information:

study code, subject number, visit number, test allocation (e.g. ELISA-ZIKA, ELISA-measles, VNT)

Samples Storage

Aliquots of samples will be stored at -80°C in a temperature monitored freezer. The temperature will be controlled either by a connected central monitoring system or a min/max thermometer, which will be controlled and documented each working day.

Handling and storage of blood for PMBC preparation will be explained in a corresponding section in the above-mentioned Lab-Manual.

Transport of Samples

Study samples packed in sufficient dry ice will be shipped by an authorised courier from the study site either to the sponsor, a dedicated analytical laboratory or a sample storage facility. Pick up of serum samples will be organised together with the site personnel at the end of the study or during the study as necessary.

9. INVESTIGATIONAL MEDICINAL PRODUCT (IMP)

9.1 Investigational Medicinal Product Description

9.1.1 MV-ZIKA-RSP vaccine-test product

The MV-ZIKA-RSP vaccine candidate is a live attenuated recombinant viral vectored vaccine for prophylaxis of Zika virus infection. [redacted] chemically synthesized and inserted into the Schwarz vaccine strain of measles virus (MV) to produce the candidate vaccine MV-ZIKA-RSP (further details under section 4.1; also refer to IB and IMPD).

The MV-ZIKA-RSP vaccine is provided by the sponsor in glass vials.

The study drug used in the study is a liquid frozen suspension for injection with an extractable dose of:

High dose: Concentration: 3.0 x 10^5 $TCID_{50}$ per vial

Low dose: Concentration: 6.0 x 10^4 $TCID_{50}$ per vial

Two dosages will be administered to the participants:

High dose: Concentration: 0.5 mL 1 x10^5 $TCID_{50}$/dose

Low dose: Concentration: 0.5 mL 2,5 x10^4 $TCID_{50}$/dose

Dosage strengths have been selected based on experience with this type of vaccine (Schwarz strain measles virus vectored vaccine).

Liquid frozen MV-ZIKA-RSP vaccine will be thawed at room temperature (for about 30 min), 0.5 mL will be drawn up into a 1-mL syringe. The preparer may inject the subject or can hand over the syringe to an injector. The injecting preparer must not participate in the study as observer; the injector, if participating in later study conduct must be blinded (e.g. by blinding the content of the syringe with the provided syringe label).

9.1.2 Placebo

A sterile physiological saline solution will be used as placebo to ensure blinding of the treatment schedule in case of single vaccination regimen (treatment group C and D). The physiological saline solution (0.9% NaCl) is a clear colourless solution ready to use, stored at room temperature and provided by the study site.

9.2 Packaging and Labelling

Individual cardboard boxes will contain single 2 ml transparent glass vials, with rubber stopper and aluminium seal, containing frozen liquid. The labels on vials and boxes will be in English language.

For further details of IMP labelling, please refer to the study specific IMP-Manual.

9.3 Dose Regimen

All 48 eligible participants will receive at their first treatment MV-ZIKA-RSP, at Visit 1 (day 0), and 4 weeks later the second treatment either with MV-ZIKA-RSP or placebo, at Visit 2 (day 28 ±3)

Visit	VS	Visit 1	Visit 2	Visit 3	Visit 4
Study Day		0	28 ±3	56 ±3	182 ±14
Study Treatment		❶	❷		

The 4 treatments groups (A, B, C and D) differ in the concentration of MV-ZIKA-RSP, as well as, in single or double shot treatment regimen.

Group A: MV-ZIKA-RSP 1 $x10^5$ (±0.5 log) $TCID_{50}$ /dose day 0 and day 28

Group B: MV-ZIKA-RSP 2,5 $x10^4$ (±0.5 log) $TCID_{50}$ /dose day 0 and day 28

Group C: MV-ZIKA-RSP 1 $x10^5$ (±0.5 log) $TCID_{50}$ /dose day 0 and placebo day 28

Group D: Placebo day 0 and day 28

9.4 Justification of Clinical Dose Regime

Two different doses of MV-ZIKA: $2,5x10^4$ (± 0.5 log) - low dose- and $1x10^5$ (± 0.5 log) -high dose- 50% tissue culture infective dose ($TCID_{50}$) will be assessed in healthy adults for safety and immunogenicity as determined by the induction of functional, neutralizing antibodies (Table 4). A dose of $2x10^6$ $TCID_{50}$ was assessed for toxicity in a GLP compliant repeated dose toxicity study. The 20-fold higher dose compared to the clinical dose showed no adverse local or systemic effects.

The clinical dose was selected based on findings in non-clinical pharmacology studies, clinical dose finding studies of comparable vaccine candidates and on assessment for cost-effective manufacturing of a vaccine for use in low-and middle-income countries. A dose of $1x10^5$ $TCID_{50}$

in NHP conferred 100% protection against Zika virus challenge. In addition, clinical trial on a comparable, measles vector-based vaccine candidate against Chikungunya virus (CHIKV) was effective and safe in the here proposed dose range. A dose of $5x10^5$ $TCID_{50}$ induced up to 100% seroconversion in healthy adult subjects after two immunizations.

[redacted]. However, a reduction to a potentially highly effective lower dose will substantially affect manufacturing costs. Zika virus is predominantly endemic in low- and middle-income countries. Thus, a vaccine will need to meet the qualitative and economical requirements.

Table 4. Dose regime

Treatment Group	N	Vaccine	Formulation	Vaccine on day	Placebo on day
A	14	MV-ZIKA-RSP 1 $x10^5$ /0.5 ml	liquid frozen	0 + 28	
B	14	MV-ZIKA-RSP 2,5 $x10^4$/0.5 ml	liquid frozen	0 + 28	
C	12	MV-ZIKA-RSP 1 $x10^5$ /0.5 ml	liquid frozen/placebo liquid	0	Placebo on day 28
D	8	Placebo/0.5 ml	liquid	Placebo on day 0	Placebo on day 28

9.5 Storage and Transport Conditions

The IMP must be stored under continuously temperature-controlled condition in a lockable room, or lockable freezer with limited access.

The minimum and maximum temperature will be recorded daily in the temperature logs throughout the study, except on weekends and holidays if min/max thermometer will be used.

In case the freezer/fridge is connected to an alarm system and corresponding copies of the readout can be filed, a min/max temperature does not need to be recorded daily. It is acceptable to use a site-specific temperature log, which covers all the required information.

MV-ZIKA-RSP have to be kept in the outer package to be protected from light.

The liquid frozen formulation of MV-ZIKA-RSP will be stored and transported at -80°C and shall be administered within 30 min after preparation, the ready to use formulation can be stored at 2-8°C for up to 1 hour.

The 0.9% saline solution and sterile water diluent will be stored according to instruction leaflet. It is under the site's responsibility to ensure the storage conditions according to the summary product characteristics.

After administration, all used containers properly labelled with subject ID and appropriate visit identifier (e.g. visit number, date of administration) will be stored at site until checked for accountability by the unblinded monitor.

9.6 IMP Preparation and Distribution

MV-ZIKA-RSP and placebo will be prepared and administered by unblinded, authorised and qualified staff members (investigator, pharmacist, or nurse) otherwise not involved in the conduct of the trial after randomisation. Preparation of MV-ZIKA-RSP for injection will be performed in a dedicated lab appropriate for handling of GMOs applying biosafety level 2 (BSL2) standards although MV-ZIKA-RSP are assessed as a biosafety level 1 product. BSL2 is applied because there is a negative system biological safety level 2 cabinet available within a containment room, which usually is used for preparing IMPs.

MV-ZIKA-RSP and placebo will be exclusively used for the present clinical trial and will only be administered to participants enrolled in the study.

For detailed description of receipt, storage, preparation, accountability, and destruction of IMP, please refer to the study specific IMP-Manual.

9.7 GMO Handling

MV-ZIKA-RSP vaccine are a genetically modified organism requiring biosafety level 1 standards. Nevertheless, preparation and disposal of IMP will be performed in a dedicated lab according to applicable hygiene standards of biosafety level 2. Needles and syringes that have been in contact with MV-ZIKA-RSP, as well as all other potentially contaminated materials, will be collected in dedicated containers and will be destroyed in a safe manner.

This study will be conducted under regulations for contained use of a GMO.

9.8 Route and Method of Administration of IMP

All participants will receive intramuscular (i.m.) injections of MV-ZIKA-RSP vaccine or placebo in the deltoid region of the non-dominant arm (preferred) or the dominant arm. In case of ongoing local AEs from previous vaccinations at the respective injection site, vaccination in the contra-lateral arm should be performed.

9.9 Treatment Compliance

The study medication will be prepared at site and administered during the study Visit 1 and 2. Each treatment will be documented in the subject's charts, the eCRF, an IMP Preparation Log and an IMP Inventory Log. Participants must receive all assigned vaccinations to be considered compliant.

9.10 Study Drug Accountability

IMP accountability will be performed throughout the entire study, starting with the initial receipt of medication.

The authorised, unblinded study staff members will confirm the number and condition of received vials, by signing and dating an IMP receipt form.

Upon receipt an IMP inventory log will be kept current by the site, detailing the batch numbers, dates and quantity of IMP obtained, used for administration on a per subject basis and destroyed or returned to the sponsor. This documentation will only be available to an unblinded monitor verifying drug accountability during the study.

At the end of the study, all unused and used MV-ZIKA-RSP vials (marked with subject ID and visit number) will be returned to the sponsor or destroyed by the investigator in a confidential manner, after final reconciliation and confirmation of correctness of drug accountability.

9.11 Prior and Concomitant Therapy

9.11.1 Permitted Prior and Concomitant Therapy

Any prior vaccination within the last three years prior screening and any medication within 30 days prior screening has to be documented.

Any medication taken during the study up to Visit 4 (study day 182) has to be reported to the investigator and will be documented.

Additionally, any treatment that will be considered necessary for the subject's welfare may be given at the discretion of the Investigator. All concomitant medications must be reported in the appropriate section of the eCRF along with dosage information, dates of administration, and reasons for use. Generic names for concomitant medication should be preferred, if possible.

All concomitant medications will be coded using the WHO Drug Dictionary.

9.11.2 Forbidden Prior and Concomitant Therapy

Treatments with non-study licensed vaccines within 4 weeks prior to first study vaccination until the end of the study (day 182) are not allowed.

Use of immunosuppressive drugs like corticosteroids (excluding topical preparations) within 30 days prior to the first vaccination or anticipated use before completion of the study (day 182) is not allowed.

Receipt of blood products or immunoglobulins within 120 days prior to the Screening Visit or anticipated receipt of any blood product or immunoglobulin before completion of the study (day 182) is not allowed.

Participants must be asked about concomitant medication and vaccinations at each visit. Any concomitant medication or vaccination must be documented.

10. SAFETY REPORTING

10.1 Definitions

10.1.1 Adverse Event (AE)

Any untoward medical occurrence in a participant to whom an investigational medicinal product (IMP) has been administered, not necessarily caused by or related to that product.

An AE can therefore be any unfavourable or unintended sign, abnormal laboratory finding, symptom or disease temporally associated with the use of an IMP whether or not considered related to the IMP.

10.1.2 Clinical Laboratory Adverse Event

A clinical laboratory AE is any clinical laboratory abnormality that suggests a disease and/or organ toxicity and is of a severity, which requires active management (i.e., changes of dose, discontinuation of drug, more frequent follow-up or diagnostic investigation).

10.1.3 Adverse Reaction (AR)

An untoward and unintended response in a participant to an investigational medicinal product which is at least possibly related to any dose administered to that participant.

All cases judged by either the reporting medically qualified professional or the sponsor as having a reasonable suspected causal relationship to the trial medication qualify as adverse reactions.

10.1.4 Serious Adverse Event (SAE)

A serious adverse event is any untoward medical occurrence that:

- results in death
- is life-threatening
- requires inpatient hospitalisation or prolongation of existing hospitalisation
- results in persistent or significant disability/incapacity
- consists of a congenital anomaly or birth defect

Other ‘important medical events’ may also be considered serious if they jeopardise the participant or require an intervention to prevent one of the above consequences.

The term "life-threatening" in the definition of "serious" refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event which hypothetically might have caused death if it were more severe.

Any pregnancy occurring during the clinical trial and the outcome of the pregnancy will be recorded and followed up for congenital abnormality or birth defect, at which point it would fall within the definition of “serious”.

NOTE: to avoid confusion or misunderstanding of the difference between the terms “serious” and “severe”, the following note of clarification is provided: “Severe” is used to describe the intensity of a specific adverse event, while “seriousness” is used for defining regulatory reporting obligations as supplied above.

10.1.5 Suspected Unexpected Serious Adverse Reaction (SUSAR)

A serious adverse reaction, the nature, severity, or outcome of which is not consistent with the applicable product information.

10.1.6 Causality

The relationship of each AE to the IMP must be determined by a medically qualified person according to the following definitions:

Definitely: Temporal relationship to the administration of the study drug and course following a known reaction pattern

Probably: Good reasons and sufficient documentation to assume a causal relationship

Possibly: A causal relationship is conceivable and cannot be dismissed

Unlikely: The event is most likely related to an etiology other than the trial treatment

Not Related: No temporal relationship to the administration of the drug or other factors have caused the event

10.1.7 Severity

AEs must be graded by a medically qualified person as being mild, moderate, severe, or life-threatening and their approximate duration given. Definitions of severity are as follows:

Mild: an AE that requires minimal or no treatment and does not interfere with daily activities.

Moderate: an AE that is sufficiently discomforting to interfere with normal activities.

Severe: an AE that is incapacitating or prevents normal activities and may require systemic drug therapy or other treatment.

Potentially life threatening: an AE that requires immediate intervention to prevent death.

"life-threatening" refers to an AE in which the subject was at risk of death at the time of the event, it does not refer to an AE which hypothetically might have caused death if it were more severe.

Please see Appendix 1 for stencil to be used for grading local injection site reactions and Table 5 and 6 for grading of systemic and local reactions.

10.1.8 Expectedness

Expectedness will be determined considering the current Investigators' Brochure.

Expected: an AE that is listed in the current Investigator's Brochure of MV-CHIK.

Unexpected: an AE that is not listed in the current Investigator's Brochure or differs due to greater severity or greater specificity.

All SAEs assessed as unexpected and suspected to be related to the IMP qualify for a SUSAR (suspected unexpected serious adverse reaction) and require expedited reporting.

10.1.9 Outcome

Recovered: A subject has recovered from an AE,when all signs or symptoms returned to normal.

Stabilised: An AE is stabilised when, according to the investigator, the subject is in a clinically stable condition. This term should only be used for chronic conditions and for a given subject only when he/she has completed the study.

Recovered with sequelae: As a result of the SAE, the subject is suffering from persistent or significant disability/incapacity (e.g. became blind, deaf, paralysed). Any AE recovered with sequelae should be rated as an SAE since an SAE criterion is fulfilled.

Not Recovered: An AE currently ongoing.

Ongoing at final examination: An AE ongoing at the subject's last visit.

Died: An AE that caused death.

10.2 Recording of Adverse Events

All AEs occurring during the treatment period starting after the first vaccination at Visit 1 until termination of Visit 4 (day 182) observed by the investigator or reported by the subject (verbally or in the subject diary), will be recorded, whether or not related to the study treatment.

AEs will be requested at each visit during the treatment period up to Visit 3 (day 56) and additionally solicited by a subject's diary for 7 days after each vaccination. (see section Subject Diary)

The participants will be asked generally (e.g. "How are you?") as well as specifically (e.g. "Have you experienced any problems since the last visit?").

The investigator must record all adverse events in the eCRF. One single adverse event page must be used per adverse event, from start to resolution, stating the following information:
AE description, date of onset and end date, severity, relatedness, seriousness, and action taken. Please consider definitions of section 10.1. Follow-up information should be provided, as necessary.

If possible, the investigator should record the diagnosis. If a diagnosis is not available, the investigator should record each sign and symptom as individual adverse events. However, as soon as a diagnosis becomes available, all signs and symptoms should be summarised and replaced by the underlying adverse event term (e.g. common cold instead of separate AEs for headache, rhinorrhoea, arthralgia, fever etc.)

The AE should be documented stating the highest severity (i.e. if an AE goes from mild severity to moderate severity or vice versa, the whole episode of the AE should be reported with the moderate severity). Please consider Table 5 and 6 for grading local injection site reactions and systemic reactions.

For causality reporting purposes, the categories "definitely", "probably" and "possibly" qualify an event as "suspected" adverse reaction. AEs with missing causality assessment will be regarded as possibly related unless further specified.

Any medical condition that is present at screening will be considered as medical history and not reported as an AE. However, if the subject's condition deteriorates at any time during the study, it will be recorded as an AE.

It will be left to the investigator's clinical judgment to decide whether an AE requires the subject's removal from the study. In case of discontinuation, the subject will undergo an early termination visit.

10.3 Reporting of Serious Adverse Events

The investigator should report all SAEs occurring during the treatment period (whether or not related to the IMP) within 24 hours of the site study team becoming aware of the event, to the Assign Safety Desk by completing an SAE Report Form.

The completed SAE Report Form should be submitted to the Assign Safety Desk either by fax or as pdf per e-mail:

Fax: PPD

Email

For urgent questions, please call the 24-hours Safety Hotline: PPD

Under certain circumstances, the first notification can be done by phone, nevertheless, a written Serious Adverse Event Report Form should be submitted for confirmation to Assign Safety Desk.

After the initial SAE report the investigator will follow up proactively each subject and provide further information like copies of hospital case reports, laboratory reports or autopsy reports, to the Assign Safety Desk within 24 h after receipt.

Assign Safety Desk will assess initial and follow-up SAE reports from the site for expectedness, which will be determined based on the most recent edition of the Investigator's Brochure.

Reports of suspected unexpected serious adverse reactions (SUSARs) will be reported in an expedited manner to the CA as required per local regulations within the required time frame.

The original copy of the SAE Report Form and the fax confirmation sheet must be kept with the documentation at the study site. Each SAE has also to be documented in the eCRF as a serious adverse event in the corresponding AE section.

10.4 Follow-up of Adverse Events

During and following a subject's participation in a clinical trial, the investigator will ensure that adequate medical care is provided to the subject for any adverse events, including clinically significant laboratory values related to the trial.

All non-serious and serious adverse events will be followed until the subject has recovered, stabilised, recovered with sequelae or died until end of the study.

Follow-up information about a previously reported SAE must also be reported within 24 hours of the investigator receiving it. The initial SAE report form should be used, stating that this is a

follow-up report to the previously reported SAE and giving the number and date of the follow-up report. The information provided should describe whether the event has resolved or continues, if and how it was treated, and whether the subject continued or discontinued study participation, if not already stated in the initial report.

10.5 Reporting of SUSARs

A serious adverse reaction that is not expected according to the current IB and were the causality is assessed as "definitely", "probably" or "possibly", qualifies as "suspected" unexpected serious adverse reaction.

On behalf of the sponsor, Assign Safety Desk will be responsible to fulfil the requirements of expedited reporting of all SUSARs (suspected unexpected serious adverse reactions) to the Competent Authorities, Ethics Committees and principal investigators concerned according to national and European law and regulations.

For fatal and life-threatening SUSARs, this will be done no later than 7 calendar days after the sponsor or delegate is first aware of the reaction. Any additional relevant information to the initial report that was not available within the 7 calendar days will be reported within 8 additional calendar days. All other SUSARs will be reported within 15 calendar days.

Treatment codes will be unblinded for specific participants.

Principal investigators will be informed of all SUSARs related to the relevant IMP for all studies conducted with the same IMP, whether or not the event occurred in the current trial.

10.6 Reporting of Pregnancies

Women should not become pregnant during the treatment period of the study. If a subject becomes pregnant within the treatment period up to day 56, she should immediately inform the investigator about the pregnancy.

The investigator will complete a corresponding Pregnancy Report Form and send the form within 24 hours after becoming aware to Assign Safety Desk via fax or as pdf per e-mail:

Fax: PPD

Email

The first notification can be done via phone 24-hours Safety Hotline: PPD

Nevertheless, a written Pregnancy Report Form must be submitted for confirmation. The subject should attend follow-up visits as planned. In addition, participants who become pregnant within the treatment period up to day 56 (and who received at least one dose) will be followed until the pregnancy outcome, even if this occurs after the study is completed.

Pregnancy itself is not regarded as an AE unless there is a suspicion that the IMP may have interfered with the effectiveness of a contraceptive medication. However, the outcome of all pregnancies that begin before day 56 (spontaneous miscarriage, elective termination, normal birth or congenital abnormality) will be followed up and documented even if the subject was discontinued from the study.

A pregnancy outcome of a congenital abnormality/birth defect would qualify as an SAE and will be reported to Assign Safety Desk for processing to the regulatory authority. A pregnancy outcome of an elective abortion without evidence of complications would not be processed as an AE.

10.7 Development Safety Update Reports

Assign Data Management and Biostatistics GmbH will submit a DSUR once a year throughout the clinical trial to the competent authority (AGES), ethics committee, according to applicable regulations and requirements.

11. ASSESSMENT OF SAFETY

Adverse event rates compared between 4 different treatment groups will be used to evaluate the secondary study objective. Please also refer to Table 2, summarising study procedures including safety assessments.

11.1 Safety Laboratory Parameter Assessments

Haematology, blood chemistry, coagulation parameters and urinalysis will be assessed for subject's safety at screening, Visit 2, Visit 4 and additionally at Early Termination (if applicable). Please refer to section 10.

11.2 Symptom-directed Physical Examination

A full physical exam will be performed at screening and a system based physical examination will be done, if necessary, according to findings in a symptom-directed physical examination at Visit 1,2 and 3. This means if a symptom is reported by the subject, a system-based assessment will be performed, if needed, for a detailed check of the affected body system.

Any symptom reported after first vaccination, including worsening of pre-existing conditions (i.e. medical history/concomitant diseases), will be recorded as AE.

11.3 System-based Physical Examination

All participants will undergo a system-based physical examination, including but not limited to assessment of general appearance and skin, head/eyes/ears/nose/throat, respiratory system, cardiovascular system, abdominal and gastrointestinal system, musculoskeletal system, neurological system and lymph nodes.

This system-based physical examination will only be performed if necessary, according to findings of the symptom-directed physical exam at Visit screening as well as Visits 1,2 and 3.

11.4 Vital Signs and Body Temperature

Systolic and diastolic blood pressure and pulse rate as well as body temperature will be recorded at screening and Visits 1,2 and 3 with the subject at rest in a sitting position.

11.5 Post Vaccination Reactogenicity Assessments

All participants enrolled in the study and who receive at least one study treatment (MV-ZIKA-RSP or placebo) will be followed up 28 days after each study treatment. Solicited and unsolicited AEs will be recorded at each visit until the end of the study at Visit 4.

11.6 Local and Systemic Tolerability

The local tolerability (i.e. injection site reactions) will be inspected and evaluated by the investigator one hour after each IMP administration.

Additionally, local tolerability (i.e. inspection of the previous injection site) will be assessed at the following visits and prior to application of the subsequent vaccination at Visit 2 (day 28). Furthermore, local tolerability will be evaluated by the participants in the subject diaries for 7 days starting 6 hours after each treatment.

The grading of local reaction will be performed by means of a stencil (provided to the subject as shown in Appendix 1) and per "Guidance for Industry: Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials", modified, reflecting the guidance of the Brighton Collaboration (Table 5).

Findings in the local tolerability examination by the investigator as well as local tolerability findings recorded in the subject diary will be covered as adverse events in the eCRF.

11.7 Subject Diary

The participants will be instructed to complete a diary for 7 days after each vaccination to document local and systemic tolerability as well as body temperature.

Solicited local and solicited systemic AEs will be assessed by the participants themselves by checking for presence of the listed symptoms and measuring the size of the affected area where appropriate (a template will be provided for local reaction grading, as shown in Appendix 1). The diary will also provide space for recording unsolicited AEs and concomitant medication.

Recording should be done approximately at the same time each day, starting on the day of vaccination. The first entry should be made 6 hours ±1h after vaccination at Visit 1 and Visit 2.

The participants' diaries will be collected and verified for completeness by the investigator at Visit 2 and Visit 3. Any AE recorded in the subject diaries will be entered in the eCRF by the investigator or authorised delegates. The investigator will re-evaluate the severity of the reported local and systemic AEs according to the below Tables 5 and 6.

Diary solicited systemic AEs include nausea, vomiting, headache, and fatigue, also to be documented in the eCRF.

In addition, body temperature will be recorded daily for 7 days and fever will be graded by the investigator according to the FDA Guidance for Industry. For details see below Table 6.Table 5. Grading of local reactions

Local reaction to injectable product	Mild (grade 1)	Moderate (grade 2)	Severe (grade 3)	Potentially life threatening (grade 4)

Pain	Does not interfere with activity	Interfere with activity or repeated use of non-narcotic pain reliever	Prevents daily activity or repeated use of non-narcotic pain reliever	Emergency room (ER) visit or hospitalisation
Tenderness	Mild pain to touch	Pain with movement	Significant pain at rest	ER visit or hospitalisation
Erythema/ Redness[1]	≤ 5 cm	5.1–10 cm	> 10 cm	Necrosis or exfoliate dermatitis
Induration/ Swelling[2]	≤ 5 cm and does not interfere with activity	5.1–10 cm or interfere with activity	> 10 cm or prevents daily activity	Necrosis
Itching	Does not interfere with activity	Interferes with activity or repeated use of non-narcotic pain reliever	Prevents daily activity or repeated use of anti-inflammation and pain-relieving ointment	Emergency room (ER) visit or hospitalisation

[1] In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.
[2] Swelling should be evaluated and graded using the functional scale as well as the actual measurement.

Table 6. Grading of systemic reactions

Systemic Reaction (General)	**Mild (grade 1)**	**Moderate (grade 2)**	**Severe (grade 3)**	**Potentially life threatening (grade 4)**
Nausea/ vomiting	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	Emergency room (ER) visit or hospitalisation for hypotensive shock
Diarrhoea	2-3 loose stools or < 400 gms/ 24hours	4-5 stools or < 400-800 gms/ 24hours	6 or more watery stools or >800 gms/ 24 hours or requires outpatient IV hydration	ER visit or hospitalisation
Headache	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 hospitalisation
Fatigue	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalisation
Myalgia	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalisation
Illness or clinical AE (as defined according to applicable regulations)	No interference with activity	Some interference with activity not requiring medical intervention	Prevents daily activity and requires medical intervention	ER visit or hospitalisation
Fever (°C)	38.0 - 38.4	38.5 - 38.9	39.0 - 40.0	>40.0

12. STATISTICS

The data will be analysed by Assign Data Management and Biostatistics GmbH. A Statistical Analysis Plan (SAP) will be finalised before database snapshot for the preliminary analysis providing a detailed description on the statistical methods and evaluation of study results.

A preliminary analysis will be performed after all participants completed the treatment period and the final analysis will be conducted after the last subject completed the Post Treatment Safety Follow-Up.

12.1 Preliminary Analysis

A preliminary analysis including safety data will be performed after all participants completed Visit 3 (day 56) and therefore terminated the treatment period of the study. This analysis will cover the primary safety endpoint (adverse events up to day 56. The purpose of this analysis is to provide highly valuable data for further zika vaccine development.

Results from this preliminary analysis will not have any impact on the study design or the participants during the follow up period of the study.

12.2 Methods of Statistical Immunogenicity Analysis

The immunogenicity analysis will compare the anti-zika-rsp VNT antibody geometric mean titre (GMT) in the Per Protocol (PP) analysis population between the treatment groups. GMTs and GMT ratios will be estimated by applying an analysis of variance including the factor treatment group. This will be done using $\log_{10}$ transformed data and taking the anti-log of the resulting point estimates for the least squares means, least squares means differences and the corresponding 2-sided 95% CIs.

P-values will also be provided to compare GMTs between the treatment groups adjusted for multiple comparisons according to Tukey-Kramer. Seroconversion rates will be compared between groups using Fisher's exact test.

Seroconversion will be defined as anti-zika-rsp VNT titres ≥20.

ZIKA-RSP- and measles- ELISA titres will be analysed as described above.

12.3 Methods of Statistical Safety Analysis

The analysis of safety will be performed in the Safety Population.

AEs and concomitant diseases will be coded using the MedDRA coding dictionary.

<u>Solicited AEs (reactogenicity):</u>

Solicited local and systemic AEs gathered by the participants' diaries will be summarised for the 7-day period, by AE grading (mild, moderate, severe) and by treatment group. The number and percentage of participants with AEs will be presented. Fisher exact tests will be provided for a comparison of AE rates between treatment groups in summary tables.

Unsolicited AEs:

Separately, the number and percentage of participants with unsolicited/spontaneous adverse events (AEs)and serious adverse events (SAEs) will be presented for each treatment group overall, and by system organ class/preferred term, by AE grade, and relatedness. Fisher exact tests will be provided for a comparison of AE rates between treatment groups.

Laboratory values and vital signs as well as changes in laboratory values from baseline will be analysed descriptively by time point and treatment group.

12.4 Determination and Justification of Sample Size

A formal sample size calculation was not conducted. The sample size of 48 participants has been determined based on prior experience in evaluating the safety and immunogenicity of vaccines and is typical for early phase clinical studies. Sample size for this study was determined on grounds of feasibility and common practice in similar trials.

12.5 Analysis Populations

12.5.1 Safety Population

All safety analyses will be based on the safety population, defined as participants who entered into the study and received at least one IMP administration. All analysis based on the Safety Population will be carried out using the actual treatment received.

12.5.2 Modified Intent-to-Treat (mITT) Population

The secondary immunogenicity analyses will be based on the modified ITT population. The modified intent-to-treat (mITT) analysis population is defined to include all participants randomised who receive at least one IMP administration. Participants will be analysed according to the treatment group they had been allocated to, rather than by the actual treatment they received.

12.5.3 Per-Protocol (PP) Population

The immunogenicity analysis will be assessed primarily on the per-protocol (PP) population. The PP population includes participants without protocol deviations that could impact immune response. Examples that may lead to exclusion from the PP population are provided here (further criteria may be defined in the SAP):

- Immunosuppressive drugs: Use of corticosteroids (excluding topical preparations) or immunosuppressive drugs within 30 days prior to first IMP administration, or anticipated use during the trial.
- Participants with any confirmed immunosuppressive or immunodeficient condition, including human immunodeficiency virus (HIV), hepatitis A, B or C infection or a family history of congenital or hereditary immunodeficiency
- Participants who received the wrong or no IMP

These criteria for potential protocol violation are identified at the time of planning the study. 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 population will be made by the sponsor on a case by case basis in a blinded manner (and prior to study unblinding at the preliminary analysis). Sample testing issues may also lead to exclusion from the PP population for particular time points.

12.6 Statistical Analysis Plan

A Statistical Analysis Plan (SAP) will be written and finalised prior to the preliminary analysis. The SAP will provide a detailed description of the statistical methods and expand on the details provided in the protocol. Additional analyses may be added.

12.7 Missing Data

All attempts will be made to prevent missing data and generally, missing data will not be imputed, and analysis will be limited to observed values. However, for missing data in AE evaluation (e.g. missing information about severity or causality) a worst-case approach will be applied.

13. QUALITY CONTROL AND QUALITY ASSURANCE

The trial will be conducted in accordance with the current approved protocol, ICH GCP guidelines, relevant regulations and standard operating procedures.

13.1 Periodic Monitoring

A designated Clinical Research Associate (CRA) will monitor study progress by scheduling and performing on-site study visits throughout the study including site initiation visit, several site monitoring visits and site close out visit. At regular intervals throughout the study monitoring visits will be performed to verify completeness, accuracy and consistency of data in the eCRFs, protocol adherence, adherence to GCP and the applicable regulatory requirements. Frequency and scope of source data verification will be determined before study start and detailed in the Monitoring Plan.
Therefore, the monitor must have access to all source records needed to verify the entries on the eCRFs. The investigator will cooperate with the monitor to ensure that any discrepancies identified are resolved.
The periodic monitoring will be performed by Neox Clinical Research.

13.2 Audit and Inspection

Upon request, the investigator will make all study-related source data and records available to a qualified quality assurance auditor mandated by the sponsor or to regulatory inspectors.

The investigator or designee should contact the sponsor/CRO immediately upon announcement of an audit by the regulatory authority. He/she further agrees to fully cooperate with competent authorities and participate with audits conducted at a convenient time in a reasonable manner.

The main purposes of an audit or inspection are to confirm that the rights and welfare of the participants have been adequately protected, to assess whether ethics, regulatory and quality requirements are met and to verify that all data relevant for the assessment of safety and immunogenicity of the investigational product have appropriately been reported to the sponsor.

14. ETHICS

14.1 Ethical Conduct of the Study

This study will be conducted in compliance with relevant regulatory requirements, ICH GCP guidelines of the Declaration of Helsinki for biomedical research involving human participants and the EU directive 2005/28/EC.

14.2 Ethics Committees (EC) (AMG §§ 40,41)

Prior to initiation of the study, the protocol, subject informed consent form, any other written information to be provided to the subject, any proposed advertising material, investigator's brochure (IB), information about payments and compensation available to participants if not mentioned in the subject information, investigator's current CV and other documentation evidencing qualifications, and other documents as required by the ethics committee should be submitted.

Written approval/favourable opinion must be obtained from the IEC prior to commencement of the study. This approval should include a statement that these documents comply with GCP requirements and must identify the documents and versions reviewed.

During the trial, the investigator must promptly report the following to the IEC: amendments to the protocol, updates to IB, unexpected SAEs where a causal relationship cannot be ruled out, notes of administrative changes, deviations to the protocol implemented to eliminate immediate hazards to the trial participants, new information that may affect adversely the safety of the participants or the conduct of the trial, annually written summaries of the trial status, and other documents as required by the local IEC.

Amendments must not be implemented before approval/favourable opinion, unless necessary to eliminate immediate hazards to the participants.

14.3 Competent Authority (CA)

The CA will receive the protocol, amendments, reports on SAEs, and related relevant safety information, including the final report according to EU Directive and national regulations.

Written approval/favourable opinion must be obtained from the CA prior to commencement of the study and for all substantial amendments to the original documents before implementation.

15. DATA HANDLING AND RECORD KEEPING

15.1 Source Data and Records

Source documents are where data are first recorded and from which participants' eCRF data are obtained. These include, but are not limited to, original charts (participants' records), laboratory and pharmacy printouts, diaries, and data from automated instruments. On all trial-specific documents, other than the signed consent, the participant will be referred to by subject number, not by name.

At least the following data will be documented in the source records:

- Study identification, date of subject's study entry and termination
- Subject number
- Documentation of informed consent procedure
- Date of each study visit
- Medical history, demographic data
- Any examination findings, including local injection site reactions
- Adverse events
- Concomitant medication intake
- Early withdrawal date and withdrawal reason, if applicable
- Treatment dates
- Completed subject diaries

Source data entries must be made in accordance with GCP and local requirements.

The investigator will permit study-related monitoring, audits, IRB/IEC review and regulatory inspections, by providing direct access to source data. Source records should be preserved for the maximum period of time required by local regulations.

15.2 Periodic Monitoring

A designated monitor will inspect the eCRFs at regular intervals throughout the study to verify completeness, accuracy and consistency of the data, protocol adherence, and adherence to Good Clinical Practice guidelines. The monitor should have access to all source records needed to verify the entries on the eCRFs. The investigator will cooperate with the monitor to ensure that any discrepancies identified are resolved

15.3 Access to Data

Direct access to source data and documents will be granted to authorised representatives from the sponsor, host institution and the competent authorities to permit trial-related monitoring, IRB/IEC review, audits and inspections.

15.4 Data Collection

During each study visit, the investigator will collect and maintain notes in the subject's study records to document all procedures, significant observations and assessments, which are regarded as source data. Additionally, diaries and laboratory result reports signed and dated by the investigator, have to be kept within the subject's records. Changes to information in the study record and other source documents will be initiated and dated on the day the change is made. All documents will be stored safely under confidential conditions.

15.5 Electronic Case Report Forms (eCRFs)

15.4.1 eCRF Entries

Information from the participants' study records and other source documents will be entered into a 21 CFR Part 11-compliant validated electronic Case Report Form (eCRF) as soon as possible after each visit. The participants will be identified by a unique trial specific subject number. The name or any other identifying detail will NOT be included in the eCRF.

Entries and corrections in the eCRF may only be performed by the investigator or authorised study site staff via personal user account and password for access. This password must be kept confidential and must only be used by the person to whom it was assigned to ensure that each entry/change can be allocated to the person who performed the entry/change.

An automatic audit trail will log each data entry/change performed in the eCRF.

The eCRF system includes internal quality checks, to identify data that appear inconsistent, incomplete or inaccurate. Maintenance of the eCRF and the study database will be performed by the data management centre at Assign Data Management and Biostatistics GmbH.

15.4.2 Changes to eCRF Data

Necessary data changes of the eCRF data may be identified as follows:

- Entries are checked by the eCRF during data entry or when the eCRF page has been submitted/saved. If the data does not fulfil particular quality criteria, a message will specify the type of problem and assist in its correction.

- Monitors may ask for correction of data during monitoring (e.g., if the eCRF entry does not match the source data).
- Computerised data-check programs and/or manual checks will identify clinical data discrepancies. Corresponding queries aiming at the resolution of these discrepancies will be created within the eCRF system and the study site will be informed about new issues to be resolved on-line.

All discrepancies will be resolved on-line directly by the investigator or by authorised staff.

As long as an eCRF page is not locked, required data changes can be conducted by the investigator or authorised site staff at any time.

15.4.3 eCRF Entry Validation

The principal investigator or the authorised delegate will thoroughly review the eCRF data and will finally certify the contents of the eCRF by electronically signing the eCRFs within the data capturing system directly. If a correction was made to the eCRF data after the investigator's approval, the certification must be repeated after the changes were performed.

15.5 Confidentiality of Subject's Data

The investigator will ensure that all subject's information obtained during the conduct of the study will be kept confidential maintaining subject's anonymity.

On eCRFs or any other documents submitted to the sponsor, participants will only be identified by subject number and therefore remain confidential in all reports or publications related to the study. Documents revealing subject's identification, e.g. subject identification log and original informed consent forms, will be maintained by the investigator in strict confidence, except to the extent necessary to allow checking by CRAs, the Sponsor or competent authorities.

15.6 Record Maintenance, Archiving and Destruction

The investigator is responsible for compiling and keeping all essential documents in the Investigator Site File (ISF) during the study and for the maximum period of time in accordance with ICH GCP guidelines and local regulatory requirements.

The ISF has to be stored in a safe and secure location with restricted access to staff personnel only and will include all essential documents like the clinical protocol, competent authorities' approvals, original informed consent forms, source data, IMP dispensing and accountability logs, participants' logs and all correspondence pertaining to the study.

The investigator will agree to archive the study documentation and will not dispose of any records relevant to this study without prior written permission from the sponsor. If an investigator moves, withdraws, or retires, the responsibility for maintaining the records may be transferred to another person upon sponsor's agreement.

The sponsor or their representative will notify the investigator when study documents need no longer be retained.

16. CHANGES IN THE CONDUCT OF THE STUDY

16.1 Protocol Amendments

Any proposed changes to the protocol must be covered in a written amendment to the protocol and submitted to the appropriate ethics committees or competent authorities. Amendments may only be implemented after approval of the appropriate ethics committees or competent authorities have been obtained except where necessary to eliminate apparent immediate hazard to participants.

16.2 Study Termination

If the sponsor or the investigator decides to terminate the study before it is completed, they will notify each other in writing stating the reasons for early termination. In terminating the study, the sponsor and the investigator will ensure that adequate consideration is given to the protection of the participants' interest. The investigator, sponsor or CRO will notify the relevant ethics committee and regulatory authority in writing in accordance with local requirements. Documentation will be submitted for filing in Central File and Investigator Site File.

17. DEVIATIONS FROM THE PROTOCOL

17.1 Relevant Protocol Deviations

A protocol deviation is any noncompliance with the clinical trial protocol, ICH GCP, manuals or local requirements. The noncompliance may be either on the part of the subject, the Investigator, or the study site staff. Consequently, corrective actions are to be developed and implemented promptly.

Important protocol deviations are deviations that might significantly affect the completeness, accuracy and/or reliability of the study data or that might significantly affect a subject's rights, safety, or well-being.

All protocol deviations will be listed in the study report and assessed as to their influence on the quality of the study analysis. No deviations from the protocol of any type will be made without complying with all the ethics committee's or regulatory authority's established procedures in accordance with applicable regulations.

17.2 Serious Breaches

The Medicines for Human Use (Clinical Trials) Regulations contain a requirement for the notification of "serious breaches" to the AGES within 7 days of the sponsor becoming aware of the breach.

A serious breach is defined as "A breach of GCP or the trial protocol which is likely to affect to a significant degree:

- the safety or physical or mental integrity of the participants of the trial, or

- the scientific value of the trial".

If a serious breach is suspected the sponsor must be contacted within 1 working day. In collaboration with the principal investigator, the serious breach will be reviewed by the sponsor and, if appropriate, the sponsor will report it to the Regulatory Authority within 7 calendar days.

17.3 Premature Subject Withdrawal

Participants have the right to withdraw from the study at any time for any reason, without the need to justify. The investigator also has the right to withdraw participants in case of AEs, protocol violations or administrative reasons. Please refer to section 8.9 for details.

Since an excessive rate of withdrawal can render the study inconclusively, the unnecessary withdrawal of participants must be avoided.

18. FINANCE AND INSURANCE

18.1 Contractual and Financial Details

The investigator, the sponsor and CRO will sign a clinical study agreement prior to the start of the study outlining overall sponsor and investigator responsibilities in relation to the study. The contract will describe costs for pharmacy, laboratory and other protocol-required services.

Financial Disclosure Statements will be completed, as required by 21 CFR part 54.

18.2 Insurance (AMG §32(2))

All participants participating in this clinical study will be insured through Themis Biosciences GmbH.

19. REPORTING AND PUBLICATION

19.1 Clinical Study Report

After all participants terminated their study participation (either by completion of Visit 4 (day 182) or prematurely by early termination or are confirmed as lost to follow up), a clinical study report will be prepared by the sponsor or delegate in accordance with relevant guidelines.

19.1 Publication Policy

All results generated in this study will be considered to be strictly confidential. The investigators 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.

20. REVISION INDEX

Section	Modification	Reason
Version 1.7 (22 Apr 2019)		
N/A	First version	New Document
Version 1.8 (13 Jun 2019)		
	The Clinical Study Protocol was amended to comply with the requirements and feedback received from the Ethics Committee. Following changes have been performed:	
8.2	Statistics: Request from the Ethic Committee: For safety reasons, only 6 patients should be included for the time being, and if no serious AE occur, the study will be continued. However, due to the randomization, it cannot be ruled out that all or most of these 6 patients will be assigned to the placebo arm. This appears problematic, so make sure that enough patients from the highest dose group are included in these 6 patients.	Request implemented in the Protocol Section 8.2. Randomisation
8.11.2	Blood volumes drawn for the Immunogenicity testing updated.	To specify more preciously the volumes.
8.11.3	Pregnancy test specified.	To clarify the sample for pregnancy test.
9.4	Request of an explicit justification for selecting for the clinical trial a lower dose than used for animal experiments.	Justification was included in the protocol (Section 9.4)
Version 1.9 (26 Jun 2019)		
8.2	Specification of the adequate representation of the randomization was added.	Request from Ethic Committee
Version 2.0 (25 Mar 2020)		
11.7	Grading of local reactions was updated.	The update was done to be in accordance with definition in the referenced FDA Guidance for Industry “Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive

		Vaccine Clinical Trials", September 2007.
12.1	Preliminary Analysis of Immunogenicity data will not be performed after all participants completed Visit 3 (day 56).	The reason is that the labs don't have capacity for those tests due to the current Covid-19 pandemic situation.
17.1	Paragraph removed from this Section since do not proceed here.	Paragraph removed.
Version 2.1 (28 Sep 2020)		
1.	$PRNT_{50}$ was replaced with VNT.	A Plaque Reduction Neutralization Test method was replaced with another, highly similar serum sample test termed Virus Neutralization Test.
4.5		
5.2		
6.1		
6.2.2		
6.5		
8.5.1		
8.5.3		
8.5.4		
8.11.2	Description of usage of a Plaque reduction neutralization assay was replaced with description of usage of a Virus Neutralization Test.	
8.11.4	$PRNT_{50}$ was replaced with VNT.	
12.2		
12.2	Seroconversion will be defined as anti-zika-rsp VNT titres ≥20 ~~10~~.	Neutralizing antibodies were quantified with a slightly different assay than originally planned. This VNT has a Lower Limit of Quantification (LLOQ) of 20, requiring us to adapt the protocol to accommodate the more stringent cut-off.

21. REFERENCES

CDC/PAHO. Preparedness and Response for Chikungunya virus introduction in the Americas. www.paho.org/hq/index.php?option=com_docman&task=doc_download&gid=16984&Itemid=&lang=en

Brandler S, Lucas-Hourani M, Moris A, Frenkiel MP, Combredet C, Février M, Bedouelle H, Schwartz O, Desprès P, Tangy F. Pediatric measles vaccine expressing a dengue antigen induces durable serotype-specific neutralizing antibodies to dengue virus. PLoS Negl Trop Dis. 2007;1(3):e96.

Brandler S, Marianneau P, Loth P, Lacôte S, Combredet C, Frenkiel MP, Desprès P, Contamin H, Tangy F. Measles vaccine expressing the secreted form of West Nile virus envelope glycoprotein induces protective immunity in squirrel monkeys, a new model of West Nile virus infection. J Infect Dis. 2012;206:212-9.

Brandler S, Ruffie C, Najburg V, Frenkiel MP, Bedouelle H, Desprès P, Tangy F. Pediatric measles vaccine expressing a dengue tetravalent antigen elicits neutralizing antibodies against all four dengue viruses. Vaccine. 2010;28:6730-9.

Brandler S, Ruffié C, Combredet C, Brault JB, Najburg V, Prevost MC, Habel A, Tauber E, Desprès P, Tangy F. A recombinant measles vaccine expressing chikungunya virus-like particles is strongly immunogenic and protects mice from lethal challenge with chikungunya virus. Vaccine. 2013;31(36):3718-25.

Cao-Lormeau VM et al, Emerg Infect Dis 2013; 20: 1085–86.

Cauchemez S et al, The Lancet 2016

Combredet C, Labrousse V, Mollet L, Lorin C, Delebecque F, Hurtrel B, McClure H, Feinberg MB, Brahic M, Tangy F. A molecularly cloned Schwarz strain of measles virus vaccine induces strong immune responses in macaques and transgenic mice. J Virol. 2003;77(21):11546-54.

Commission Directive 2005/28/EC laying down principles and detailed guidelines for good clinical practice as regards investigational medicinal products for human use, as well as the requirements for authorisation of the manufacturing or importation of such products; see

https://ec.europa.eu/health/files/eudralex/vol-1/dir_2005_28/dir_2005_28_en.pdf

Committee for Human Medicinal Products (CHMP): Note for Guidance on the Clinical Evaluation of Vaccines (CHMP/VWP/164653/2005); see: http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2009/09/WC500003875.pdf

Desprès P, Combredet C, Frenkiel MP, Lorin C, Brahic M, Tangy F. Live measles vaccine expressing the secreted form of the West Nile virus envelope glycoprotein protects against West Nile virus encephalitis. J Infect Dis. 2005;191(2):207-14.

Dick GWA et al, Trans R Soc Trop Med Hyg 1952; 46: 509–20

Duffy MR et al, N Engl J Med 2009; 360: 2536–43.

ECDC. Rapid risk assessment. Zika virus epidemic in the Americas: potential association with microcephaly and Guillain-Barré syndrome. Dec 10, 2015. http://ecdc.europa.eu/en/publications/ Publications/zika-virus-americas-association-with-microcephaly- rapid-risk-assessment.pdf (accessed Feb 5, 2016).

Enserik M (2008) Entomology. A mosquito goes global. Science 320: 864-866

European Centre for Disease Prevention and Control (ECDC), December 10, 2015 (http://ecdc.europa.eu/en/publications/Publications/zika-virus-americas-association-with-microcephaly-rapid-risk-assessment.pdf)

FDA Guidance for Industry: Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials (Sept. 2007)

http://www.fda.gov/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/Guidances/Vaccines/ucm074775.htm

Gubler D et al. Flaviviruses. In: Knipe DM, Howley PM, Griffin DE, Lamb RA, Martin MA, et al, eds. Fields Virology, 5th edn. Philadelphia, PA: Lippincott Williams & Wilkins Publishers, 2007: 1155–227.

Gerardin P, Barau G, Michault A, Bintner M, Randrianaivo H, et al. (2008) Multidisciplinary prospective study of mother-to-child chikungunya virus infections on the island of La Reunion. PLoS Med 5: e60. doi:10.1371/journal.pmed.0050060

International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use: ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6 (R1); https://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines /Efficacy/E6/E6_R1_Guideline.pdf

Jain M, Rai S, Chakravarti A (2008) Chikungunya: a review. Trop Doct 38: 70-72

Lorin C, Mollet L, Delebecque F, Combredet C, Hurtrel B, Charneau P, Brahic M, Tangy F. A single injection of recombinant measles virus vaccines expressing human immunodeficiency virus (HIV) type 1 clade B envelope glycoproteins induces neutralizing antibodies and cellular immune responses to HIV. J Virol. 2004;78(1):146-57.

Macnamara FN, Trans R Soc Trop Med Hyg 1954; 48: 139–45

Msaouel P, Opyrchal M, Domingo Musibay E, Galantis E. Expert Oncolytic measles virus strains as novel anticancer agents Opin Biol Ther. 2013 Apr;13(4):483-502.

Paul BJ, Pannarkady G, Moni SP, Thachil EJ. Clinical profile and long-term sequelae of Chikungunya fever. Indian Journal of Rheumatology. Volume 6, Issue 1, March 2011, 12–19.

Ramsauer K, Schwameis M, Firbas C, Müllner M, Putnak RJ, Thomas SJ, Desprès P, Tauber E, Jilma B, Tangy F. Immunogenicity, safety, and tolerability of a recombinant measles-virus-based chikungunya vaccine: a randomised, double-blind, placebo-controlled, active-comparator, first-in-man trial. Lancet Infect Dis. 2015 May;15(5):519-27.

Immunogenicity, safety, and tolerability of the measles-vectored chikungunya virus vaccine MV-CHIK: a double-blind, randomised, placebo-controlled and active-controlled phase 2 trial

EC Reisinger, R Tschismarov, E Beubler, U Wiedermann, C Firbas, M Löbermann, A Pfeiffer, M Muellner, E Tauber, K Ramsauer. Immunogenicity, safety, and tolerability of the measles-vectored chikungunya virus vaccine MV-CHIK: a double-blind, randomised, placebo-controlled and active-controlled phase 2 trial. Published online www.thelancet.com, November 5, 2018; http://dx.doi.org/10.1016/S0140-6736(18)32488-7

Rezza G, Nicoletti L, Angelini R, Romi R, Finarelli AC, et al. (2007) Infection with chikungunya virus in Italy: an outbreak in a temperate region. Lancet 370:1840-1846

Schilte C, Staikowsky F, Couderc T, Madec Y, Carpentier F, Kassab S, Albert ML, Lecuit M, Michault A. Chikungunya virus-associated long-term arthralgia: a 36-month prospective longitudinal study. PLoS Negl Trop Dis. 2013;7(3):e2137.

Simon F, Parola P, Grandadam M, Fourcade S, Oliver M, Brouqui P, Hance P, Kraemer P, Ali Mohamed A, de Lamballerie X, Charrel R, Tolou H. Chikungunya infection: an emerging rheumatism among travelers returned from Indian Ocean islands. Report of 47 cases. Medicine (Baltimore). 2007 May;86(3):123-37.

Soares de Araújo J, Regis CT, Silva Gomes RG, et al. Microcephaly in northeast Brazil: a review of 16 208 births between 2012 and 2015. http://who.int/bulletin/online_first/16-170639.pdf?ua=1 (accessed Feb 8, 2016).

Staples JE, Breiman RF, Powers AM. Chikungunya fever: an epidemiological review of a re-emerging infectious disease. Clin Infect Dis. 2009 Sep 15;49(6):942-8.

Stebbings R, Février M, Li B, Lorin C, Koutsoukos M, Mee E, et al. Immunogenicity of a recombinant measles-HIV-1 clade B candidate vaccine. PLoS One. 2012;7:e50397.

Tsetsarkin KA, Vanlandingham DL, McGee CE, Higgs S (2007) A single mutation in chikungunya virus affects vector specifity and epidemic potential. PLoS Pathog 3: e201. doi:10.1371/journal.ppat.0030201

U.S. Department of Health and Human Services, Food and Drug Administration, Center for Biologics Evaluation and Research, September 2007: Guidance for Industry: Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials; see http://www.fda.gov/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/Guidances/Vaccines/ucm074775.htm

Vazeille M, Moutailler S, Coudrier D, Rousseaux C, Khun H, et al. (2007) Two Chikungunya isolates from the outbreak of La Reunion (Indian Ocean) exhibit different patterns of infection in the mosquito, Aedes albopictus. PLoS ONE 2: e1168. doi:10.1371/journal.pone.0001168

Wang E, Volkova E, Adams AP, Forrester N, Xiao SY, et al. (2008) Chimeric alphavirus vaccine candidates for chikungunya. Vaccine 26: 5030-5039

WMA Declaration of Helsinki - Ethical Principles for Medical Research Involving Human Participants; JAMA. 2013;310(20):2191-2194.

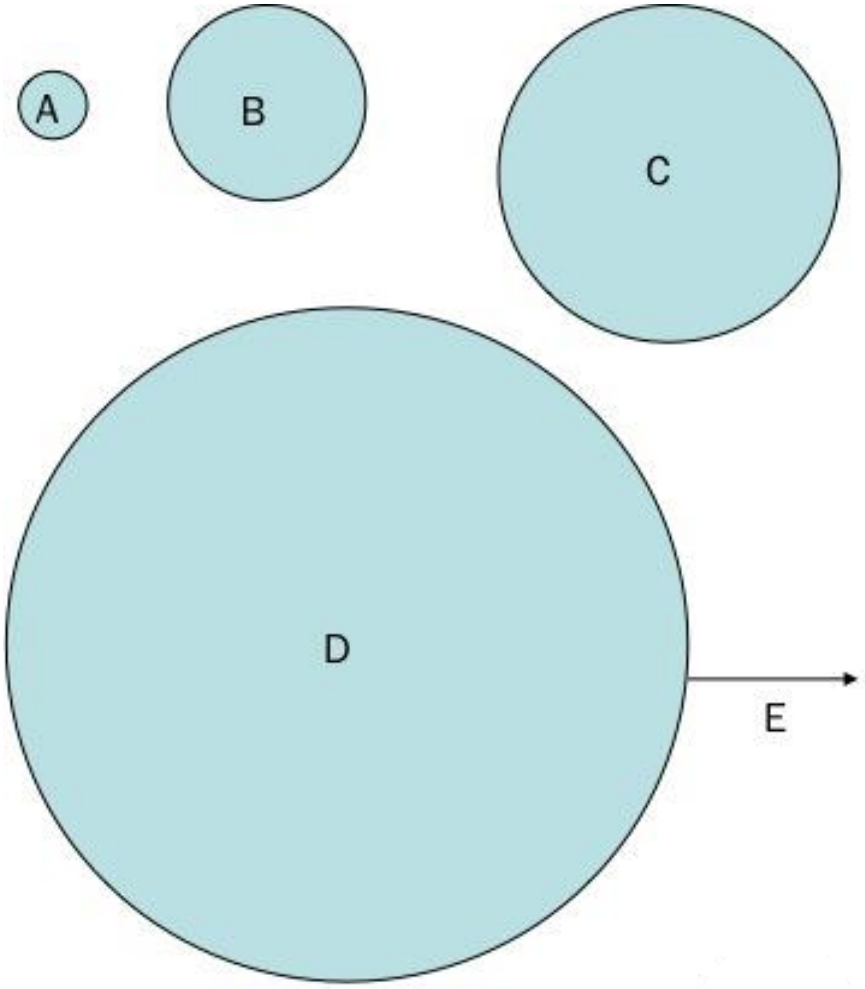
WHO. Guillain-Barré syndrome – El Salvador. Jan 21, 2016. http://www.who.int/csr/don/21-january-2016-gbs-el-salvador/en/ (accessed Feb 5, 2016).

WHO, Media centre, 2016: See: http://www.who.int/mediacentre/news/notes/2016/research-development-zika/en/

Zanluca C et al, . Mem Inst Oswaldo Cruz 2015; 110: 569–72.

22. APPENDICES

Appendix 1: Template distributed to participants to assess local reaction



MV-ZIKA-RSP-101_CSP_v2.1_28Sep2020_cl_SP1

Final Audit Report 2020-10-01

Created:	2020-10-01
By:	PPD
Status:	Signed
Transaction ID:	CBJCHBCAABAAbSvYbhJP0DvQxmmu3V_6sSiN468pg008

"MV-ZIKA-RSP-101_CSP_v2.1_28Sep2020_cl_SP1" History

PPD

Agreement completed.
2020-10-01 - 6:55:43 AM GMT

Statistical Analysis Plan

Preliminary and Final Analysis

Observer blinded, randomized trial to evaluate safety and immunogenicity of a novel vaccine formulation MV-ZIKA-RSP

Protocol: MV-ZIKA-RSP-101 / EudraCT 2019-000840-93

Confidential

Sponsor: Themis Bioscience GmbH

Table of Contents

Document	
Study	MV-ZIKA-RSP-101
Document	Statistical Analysis Plan (SAP)
Version	Final 3.0
Date	24-Sep-2020

Revision History

Version	Date	Reason for Revision
Final 1.0	24-Mar-2020	First version;
Final 2.0	10-Sep-2020	Immunogenicity analysis performed at final analysis instead of preliminary analysis since laboratory results are not available for preliminary analysis;
Final 3.0	24-Sep-2020	Replaced $PRNT_{50}$ with VNT to be consistent with updated CSP (Version 2.1);

Approval

Name	Role	Date	Signature
Author			
PPD	Biostatistician		
Assign DMB			
PPD	Senior Biostatistician		
Themis Bioscience GmbH			
PPD	Clinical Project Manager		

List of Abbreviations

AE	Adverse Event
ALT	Alanine Aminotransferase
ANOVA	Analysis of Variance
aPTT	Activated Partial Thromboplastin Time
AST	Aspartate Aminotransferase
ATC	Anatomical Therapeutic Chemical
BDRM	Blind Data Review Meeting
CI	Confidence Interval
CSP	Clinical Study Protocol
CSR	Clinical Study Report
DRM	Data Review Meeting
eCRF	Electronic Case Report Form
ELISA	Enzyme Linked Immunosorbent Assay
GMFI	Geometric Mean Fold Increase
GMT	Geometric Mean Titer
HIV	Human Immunodeficiency Virus
IMP	Investigational Medicinal Product
IMP	Investigational Medical Product
LLOQ	Lower Limit of Quantification
mITT	Modified Intent-to-Treat
PP	Per-Protocol
$PRNT_{50}$	Plaque Reduction Neutralization Test 50 %
PT	Prothrombin Time
SOP	Standard Operating Procedure
SPS®	Stabilizing and Protecting Solutions
TLF	Tables Listings and Figures
ULOQ	Upper Limit of Quantification
VNT	Virus Neutralization Test

1. OVERVIEW

1.1 Study Objectives

1.1.1 *Primary Objective*

- To investigate safety and tolerability of MV-ZIKA-RSP, a novel liquid vaccine formulation, up to day 56 after first immunization consisting in one or two vaccinations

Safety and tolerability will be assessed by the rate adverse events (AEs) after the first vaccination up to day 56.

1.1.2 *Secondary Objectives*

- To investigate long-term safety, up to study day 182, after first immunization

Long term safety and tolerability will be assessed by means of a phone call where participants will be asked about any adverse event (solicited, unsolicited or serious adverse event) experimented after the Visit 3 (day 56)

- To investigate immunogenicity up to day 56 after first immunization consisting in one or two vaccinations with different doses

To cover this objective the presence of functional anti-zika antibodies will be determined by means VNT and ELISA at the following time points: Visit 1 (day 0), Visit 2 (day 28) and Visit 3 (day 56) after first vaccination

- To get information about the optimal dose of MV-ZIKA-RSP vaccine regarding safety, tolerability and immunogenicity

An evaluation of the optimal dose will be determined after analysis of the safety, tolerability and immunogenicity (VNT)

- To investigate specific cell mediated immunity induced by all different treatment groups

Cell mediated immunity up to day 56 will be assessed by T cell analysis of PBMC isolated on study days 0, 28 and 56

1.2 Study Design

This is an observer-blinded, block-randomised, dose finding, phase I trial, comparing different dose levels of MV-ZIKA-RSP to evaluate safety, tolerability and immunogenicity, of this novel ZIKA-RSP vaccine. Placebo (physiological saline solution) will be applied to blind the different treatment schedules.

The study will be conducted at a single study site, Medical University Vienna.
After the screening procedures, 48 healthy male and female volunteers aged 18-55 years will be randomly assigned to one of four treatment groups (A, B, C or D). Participants will be assessed for immunogenicity on days 0, 28 and 56 (treatment period), as confirmed by the presence of functional anti-zika-rsp antibodies determined by (VNT) and

by ELISA, at the same time safety will be also assessed. After the treatment period, participants will be call by phone (day 182) for evaluation of safety follow-up.

Study duration per subject will be 182 days (6 months), consisting of 4 weeks treatment period (study day 0 till study day 28) and 22 weeks follow up period (study day 2 till study day 182)

1.3 Endpoints

1.3.1 *Primary Endpoint*

- Rate of adverse events (AEs) up to study day 56

1.3.2 *Secondary Endpoints*

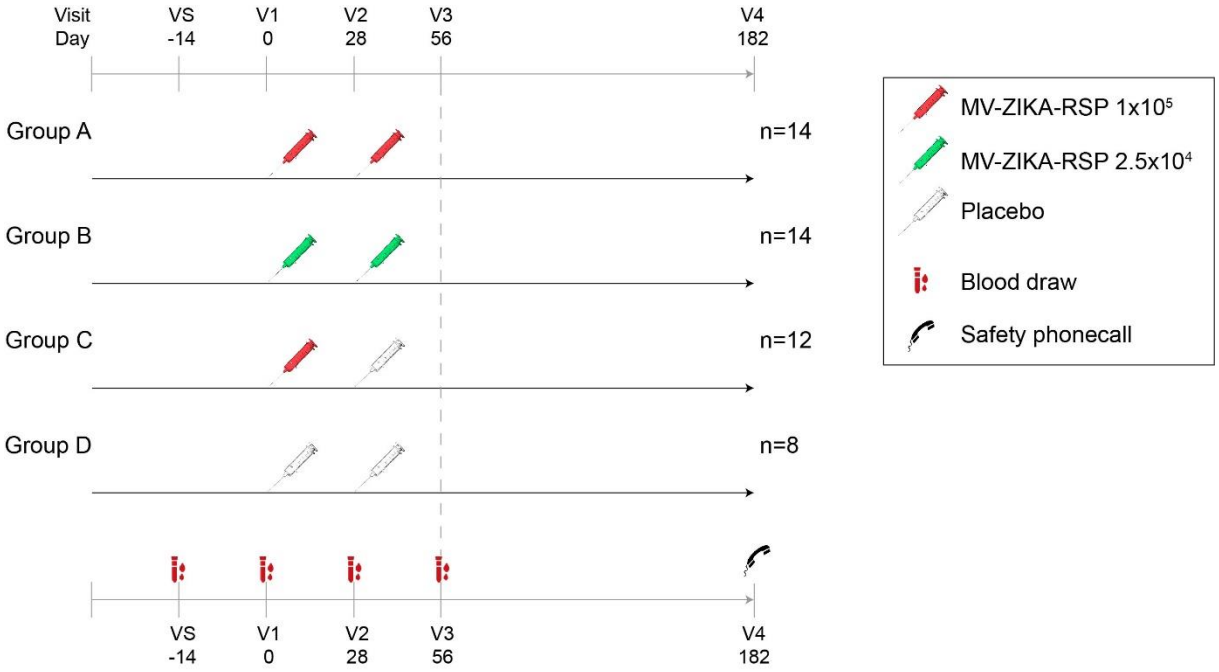
- Rate of solicited and unsolicited AEs, as well as, reported serious adverse events (SAEs) up to study day 182 (long-term safety) compared between the 4 different treatment groups.
- Immunogenicity on study days up to study day 56 as confirmed by the presence of functional anti-zika antibodies as determined by VNT and by ELISA
- Cell-mediated immunity specific for ZIKA-RSP, up to study day 56, confirmed by the presence of specific functional $CD4^+$ and $CD8^+$ T-cells
- Safety laboratory parameters (haematology, serum chemistry, urinalysis)

1.4 Sample Size Calculation

A formal sample size calculation was not conducted. The sample size of 48 participants has been determined based on prior experience in evaluating the safety and immunogenicity of vaccines and is typical for early phase clinical studies. Sample size for this study was determined on grounds of feasibility and common practice in similar trials.

1.5 Study Flowchart and Study Procedures

Study flowchart:



Study procedures:

	Screening	Treatment				Post Treatment Safety Follow Up	ET
Visit	**VS**	**V1**	Post dose safety FU	**V2**	**V3** Primary Endpoint	**V4**	(1)
Study Day	-14 to -1	0		28± 3	56 ±3	182 ±14	
Informed Consent	x						
Inclusion/Exclusion Criteria	x	x					
Medical History	x	x					
Vaccination History	x	x					
Vital Signs (2)	x	x		x	x		x
Symptom-directed Physical Examination		x		x	x		x
System-based Physical Examination (6)	x	x		x	x		x
Adverse Events (3) (7)		x	x	x	x	x	x
Prior/Concomitant Medication (4)	x	x	x	x	x	x	x
Clinical Laboratory (5)	x			x	x		x
Randomization		x					
Urine Pregnancy Test (12)	x	x		x	x		x
Study Treatment		**x**		**x**			
Immunogenicity (VNT) (12)		x		x	x		x
T-cell immunity (PBMCs) (12)		x		x	x		x (11)
Zika antibody (ELISA) (12)		x		x	x		x
Measles antibody (ELISA) (12)		x		x	x		x
Local Tolerability (7)		x		x	x		x
Dispense subject diary		x		x			

Collect and review subject diary				x	x		x
Safety phone call (9)			x (9)			x (10)	

(1) Early termination visit should – if possible - be conducted 28±3 days after last IMP administration.

(2) Systolic/diastolic blood pressure, pulse rate, body temperature

(3) Conditions/symptoms noted prior to randomization should not be reported as AE, but should be reported as medical history

(4) Medications administered up to 30 days prior to screening until end of study participation should be documented

(5) The clinical laboratory comprises: Hematology (hemoglobin, hematocrit, erythrocyte count, differential white blood count, platelets), Chemistry (creatinine, potassium, sodium, calcium, AST, ALT, alkaline phosphatase, bilirubin), Coagulation (prothrombin time (PT), activated partial thromboplastin time (aPTT), fibrinogen), Urinalysis (standard urine test stick for determining pH-value, glucose, protein, bilirubin, urobilinogen, red blood cells, white blood cells, nitrite, ketone and specific gravity), Virology (only at screening; HBs-Ag, anti-HBc-Ab, anti-HCV-Ab, HIV 1/2 Ab/HIV-1 Antigens if not done within 30 days before screening)

(6) To be performed if necessary, according to findings in symptom-directed physical examination

(7) To be performed 1 hour after each IMP administration and additionally at the subsequent visit (prior to IMP administration, if applicable)

(8) Participants should be interviewed regarding potential AEs since the last study visit. If indicated, participants should be called in for an unscheduled visit.

(9) The post dose safety call follows up should be conducted 7±1 days (one week) after the first vaccination.

(10) The post treatment safety call follows up should be conducted 182 days (6 months) after the first study treatment also in case of an early termination.

(11) The amount of blood sample taken during the early termination (ET) visit depends on the time point of the visit. If the ET-visit replaces the primary endpoint visit on day 56±3, 48 to 60 mL blood should be taken; otherwise 24 to 48 mL are enough.

(12) Urine for pregnancy test should be taken before vaccination

2. GENERAL CONSIDERATIONS

2.1 Conduct of Analysis

A preliminary analysis including safety and immunogenicity data will be performed after all participants completed Visit 3 (day 56) and therefore terminated the treatment period of the study. This analysis will cover the primary safety endpoint (adverse events up to day 56) and some immunogenicity endpoints. The purpose of this analysis is to provide highly valuable data for further zika vaccine development.
Results from this preliminary analysis will not have any impact on the study design or the participants during the follow up period of the study.

The final analysis will be conducted after the last subject completed the Post Treatment Safety Follow-Up and the database was closed. All data of the study will already be available at the preliminary analysis except the Adverse Events and Prior/Concomitant Medication data from Visit 4 (Day 182). Thus, only Adverse Events and Prior/Concomitant Medication data will be analyzed at the final analysis. If unexpected changes occur e.g. in the immunogenicity data, analyses may also be repeated at the final analysis.

SAP update (Final 2.0): Due to the covid pandemic, at the time of analysis of the samples, laboratory capacities were limited, and no results were available. For this reason, immunogenicity analysis was not conducted for the preliminary analysis. Instead, the immunogenicity analysis will be done as described in Section 5 for the final analysis only.

2.2 Statistical Software and Quality Control

All statistical analyses will be performed using SAS® version 9.3 or higher. Tables, figures and data listings will be generated in Microsoft® Word® as well as PDF® format.

Quality control of SAS® programs will include a review of the whole process of result generation:

- Review of all analysis SAS® programs
- Review of SAS® log for errors, warnings and other notes that could indicate mistakes in the programs
- Review of all tables, listings and figures for completeness and correctness

2.3 Applicable Standard Operation Procedures

The applicable Standard Operating Procedures (SOPs) of Assign DMB for this study are:

STAT01 Statistical Analysis File
STAT03 Statistical Analysis Plan
STAT04 Interim Analysis
STAT05 Randomization and Unblinding
STAT06 Data Review Meeting

STAT07	Report Writing
SAS01	SAS General Principles
SAS04	Handling of Statistical Analyses

2.4 Blinding and Randomization

Blinding:

As this study will be conducted in an observer-blinded manner, unblinded study team members responsible for randomisation, monitoring (unblinded monitor) and preparation of IMP, will otherwise not be involved in the conduct of the study.

All participants, the investigator and site personnel performing study related assessments, as well as the sponsor's representatives involved in the monitoring (blinded monitor) and conduct of the study, will be blinded to treatment assignment.

Besides one set of randomisation envelopes, the site will receive an additional set of emergency envelopes stored in the ISF for unblinding purposes. As the knowledge of the subject's treatment might become mandatory (e.g. in case of emergency) the investigator has the possibility to open the emergency envelope, identified with the subject's randomisation number, and unblind the subject.

The investigator and site personnel assessing AEs, all participants, as well as one of the sponsor's representatives involved in the monitoring and conduct of the study will be blinded to which vaccine was administered. Only the unblinded monitor, site personnel performing randomisation and preparation of IMP will be unblinded.

The primary endpoint as well as most of the secondary endpoints will be covered in the preliminary analysis after Day 56 and only a safety follow-up phone call will be performed after Day 56. The sponsor personnel and ADMB team, involved in generation and review of the preliminary analysis and underlying data management activities, will be unblinded after the Blind Data Review Meeting (Section 2.9) and database snapshot for preliminary analysis.

Randomization:

At Visit 1 (day 0) 48 eligible participants will be randomly assigned to one of four treatment groups (A, B, C or D) by means of randomisation envelopes provided by data management.

Each subject will be assigned a unique three-digit randomisation number in ascending order, by opening the randomisation envelope with the lowest free (unassigned) randomisation number available. Randomisation should be performed as late as possible, i.e. when knowledge of the assigned treatment group becomes necessary. Only trained members of the unblinded study team will be authorised to open one envelope per subject, containing the information about the allocated treatment group.

The 48 participants will be distributed between the 4 groups using a randomisation ratio of 14 : 14 : 12 : 8 = A : B : C : D, with an adequate representation of high dose, low dose and placebo subjects (2 : 2 : 0 : 2 = A : B : C : D) within the first 6 subjects to allow for the safety evaluation after the first vaccination.

A subject will be considered enrolled once the first IMP administration has been performed according to randomisation.

2.5 Descriptive Analyses

Descriptive analyses of continuous variables (summary statistics) will be described with the number of non-missing observations, arithmetic mean, standard deviation (±SD), median, quartiles (Q1 and Q3) and range (minimum and maximum).

Descriptive analysis of continuous immunogenicity variables (i.e. tables for the GMT and GMFI) will be described with the number of non-missing observations, geometric mean, confidence intervals for the geometric mean, standard deviation of logarithmic values, median, quartiles (Q1 and Q3) and range (minimum and maximum).

Categorical variables (frequency statistics) will be described with the number of non-missing observations and percentages (%). Percentages will be calculated within each stratum on the total number of non-missing observations, if not stated otherwise.

2.6 Inferential Analyses

The analysis for primary endpoint will compare adverse events rates up to day 56 between treatment groups using Fisher-Freeman-Halton test. The same overall test between treatment groups will be performed for all adverse event tables in the Day 56 analysis as well as the final analysis.

The immunogenicity analysis will compare the anti-zika-rsp VNT antibody geometric mean titer (GMT) in the Per Protocol (PP) analysis population between the treatment groups. GMTs and GMT ratios will be estimated by applying an analysis of variance including the factor treatment group. This will be done using log10 transformed data and taking the anti-log of the resulting point estimates for the least squares means, least squares means differences and the corresponding 2-sided 95% CIs.

P-values will also be provided to compare GMTs between the treatment groups adjusted for multiple comparisons according to Tukey-Kramer.

For the primary immunogenicity analysis, values below the lower limit of detection are replaced by half of the limit and values above the upper limit are replaced by the upper limit.
As sensitivity analysis, an analysis of immunogenicity will be applied, that does not use such imputation. Therefore, observations will be interval-censored and GMTs will be estimated by assuming normal distribution for the log-transformed values (intervals). Values below the lower limit of detection will be left-censored at the lower limit and values above the upper limit (if applicable) will be right-censored at the upper limit.

Seroconversion rates and positivity rates will be compared between groups using Fisher-Freeman-Halton test.

2.7 Center and Country Effect

Not applicable since this is a single-center study.

2.8 Handling Missing Data

Generally, missing values will not be imputed, and the analysis will be limited to observed values. But for missing data in the AE (e.g. missing information about seriousness, severity or causality) a worst case approach will be applied. For example, adverse events with missing severity assessments will be considered as "Potentially life threatening" and events with missing relationship to study treatment are counted as "Definitely" related to study treatment. In case of missing assignment to solicited or unsolicited, this AE will neither be counted in tables for solicited AEs nor in tables for unsolicited AEs but in tables for all AEs.

2.9 Protocol Deviations

Protocol deviations will be collected only for the preliminary analysis. A listing of the potential protocol deviations will be prepared by Assign Data Management and Biostatistics GmbH using information from the eCRF documentation and monitoring reports. These protocol deviations will be classified into major or minor protocol deviations based on their possible impact on the study results in a Blind Data Review Meeting (BDRM) prior to the preliminary analysis attended by Assign Data Management GmbH and Themis Bioscience GmbH (sponsor).

Major protocol deviations will include but are not limited to the following:

- Subjects with any confirmed immunosuppressive or immunodeficient condition, including human immunodeficiency virus (HIV), hepatitis A, B or C infection or a family history of congenital or hereditary immunodeficiency
- Subjects who received the wrong or no IMP
- Subjects who received less than the protocol-defined number of vaccinations
- Subjects who received a wrong dose other than they were randomized to
- Subjects who entered the study even though they did not satisfy the entry criteria
- Subjects with substantial time window violations on vaccination visits
- Subjects who received a forbidden prior or concomitant medication
    - Immunosuppressive drugs: Use of corticosteroids (excluding topical preparations) or immunosuppressive drugs within 30 days prior to first IMP administration, or anticipated use during the trial
    - Treatments with non-study licensed vaccines within 4 weeks prior to first study vaccination until the end of the treatment period (day 56)
    - Receipt of blood products or immunoglobulins within 120 days prior to the Screening Visit or anticipated receipt of any blood product or immunoglobulin before completion of the treatment period (day 56)
- Further protocol deviations that could impact immune response that have not been described above

These criteria for potential protocol violation are identified at the time of planning the study. 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 per-protocol (PP) population will be made by the sponsor on a case by case basis in a blinded manner (and prior to study unblinding).

All protocol deviations will be listed in the analysis and described in the Clinical Study Report (CSR).

In case of unexpected changes or further protocol deviations documented after the BDRM, it may be discussed in a second DRM prior to database closure which analyses may be repeated for the final analysis.

2.10 Medical Coding

Adverse events, medical history and concomitant procedures will be coded using MedDRA in the most current version. Concomitant medications and vaccination history will be coded using WHO Drug Reference List and Anatomical Therapeutic Chemical (ATC) Classification System in the most current version. The used dictionary versions will be indicated in the respective tables and listings.

2.11 Analysis Populations

2.11.1 *Safety Population*

All safety analyses will be based on the safety population, defined as participants who entered into the study and received at least one IMP administration. All analysis based on the Safety Population will be carried out using the actual treatment received.

Possible issues with wrong treatment and assignment of subjects to actual treatment group will be discussed in the DRM. In general, this assignment will be done in a conservative way to not bias the safety results towards MV-ZIKA-RSP. If a subject in the high dose group receives a low dose, the subject will be analysed in the low dose group, for example.

2.11.2 *Modified Intent-to-Treat (mITT) Population*

The secondary immunogenicity analyses will be based on the modified ITT population. The modified intent-to-treat (mITT) analysis population is defined to include all participants randomised who receive at least one IMP administration. Participants will be analysed according to the treatment group they had been allocated to, rather than by the actual treatment they received.

When no subjects received wrong treatment, mITT and Safety population do not differ and tabulations will only be done for safety population.

2.11.3 *Per-Protocol (PP) Population*

The immunogenicity analysis will be assessed primarily on the per-protocol (PP) population. The PP population includes participants without protocol deviations that could impact immune response. Examples that may lead to exclusion from the PP population are provided here:

- Immunosuppressive drugs: Use of corticosteroids (excluding topical preparations) or immunosuppressive drugs within 30 days prior to first IMP administration, or anticipated use during the trial.
- Participants with any confirmed immunosuppressive or immunodeficient condition, including human immunodeficiency virus (HIV), hepatitis A, B or C infection or a family history of congenital or hereditary immunodeficiency
- Participants who received the wrong or no IMP

These criteria for potential protocol violation are identified at the time of planning the study. 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 population will be made by the sponsor on a case by case basis in a blinded manner (and prior to study unblinding at the preliminary analysis). Sample testing issues may also lead to exclusion from the PP population for particular time points.

2.12 Subject Data Listings

All treated subjects will be included in listings if not stated otherwise (e.g. subject overview listings, screening failure listing). Data listings will include the subject number as identifier (and parameter and/or visit if available) and will be sorted by treatment group, subject number and parameter and/or visit if available (in this order). Columns that indicate the treatment group will be shown in all listings.

2.13 Columns in Tables

Every treatment group has its own column in the analysis and an additional total column will be added. For OV tables, where all subjects are shown, there is an additional column with subject not randomized.

2.14 Changes in the Conduct of the Study or Planned Analysis

No changes to the statistical analysis as compared to CSP are planned.

3. OVERALL STUDY INFORMATION

For visit attendance and study status the safety population is used. For the other OV tables all subjects are used.

3.1 Data points

The following information will be analyzed descriptively and corresponding details on the subject level will be provided in data listings:

- Subject overview (screened, randomized, 1st/2nd IMP administered, study populations…)
- Visit attendance
- Protocol deviations
- Study status (incl. drop-outs and reason)
- Screening failures

For details on table and listing content, see Section 7.

3.2 Derivations and Definitions

Details about study population can be found in Section 2.11.

4. BASELINE EVALUATION

Baseline analysis will be presented for the Safety, mITT population and for the PP population. In case populations do not differ, tabulations will not be repeated.

4.1 Data points

The following information will be analyzed descriptively and corresponding details on the subject level will be provided in data listings:

- Demographic information (age, sex, race)
- System-based Physical examination (will be listed only)
- Vaccination history (by ATC level 3)
- Virology (will be listed only)
- Medical history
- Prior and concomitant medications
- Pregnancy Test (will only be listed)
- Concomitant Procedure (will only be listed)
- Vital Signs at screening

For details on table and listing content, see Section 7.

4.2 Derivations and Definitions

Medications stopped clearly prior (<) to Day 0 (Visit 1) will be considered prior medications, all other medications are considered to be concomitant. Medications with a missing or incomplete stop date where it cannot clearly be decided if the stop date was before or after Day 0 will be considered concomitant.

5. IMMUNOGENICITY ANALYSIS

The immunogenicity analysis will be performed for the mITT and PP population. All tables will be presented by treatment group and overall. Data from the early termination visit or unscheduled visits will be listed only and will not be shown in summary tables by time point.

5.1 Data points

The following information will be analyzed by visit descriptively in tables and corresponding details on the subject level will be provided in data listings:

- GMT of Zika-RSP VNT antibody (incl. ANOVA with fixed treatment group as described in Section 2.6)
- Seroconversion rate for Zika-RSP VNT antibody (incl. test as described in Section 2.6)
- Positive/Negative results for NT
- GMT of Zika-RSP-ELISA antibodies (incl. ANOVA with fixed treatment group)
- Seroconversion rate for Zika-RSP-ELISA antibodies
- Positive/Negative results for ZIKA ELISA
- GMT of Measles-ELISA antibodies (incl. ANOVA with fixed treatment group)
- Seroconversion rate for Measles-ELISA antibodies
- Positive/Negative results for MV ELISA
- specific functional CD4+ T-cells by visit (without geometric mean):
    - E+prM positive/negative
    - E values
    - prM values

Sampling details for VNT, ELISA Zika-RSP, ELISA measles and T-cells immunity (will be listed only)

Specifications on TLFs are provided in Section 7.

5.2 Derivations and Definitions

- For final analysis all time points, including Visit 1 (Day 0), Visit 2 (Day 28) and Visit 3 (Day 56) will be analyzed.
- Seroconversion rate for anti-zika-rsp VNT titers is defined as proportion of subjects achieving anti-zika-rsp VNT titers ≥ 20 at a certain time point.

- Seroconversion rate for anti-zika-rsp ELISA titers is defined as 4-fold increase over baseline (Visit 1 –Day 0). If no Visit 1 (Day 0) sample is available, no seroconversion will be determined.
- For MV ELISA, Zika ELISA, NT and T-Cell an analysis of positive versus negative values is planned. Since such a definition could not be discussed and communicated with the central laboratory at the time of generation of this SAP, details about used limits will be shown as footnotes in the corresponding tables and explained in the Clinical Study Report.
- For the different immunogenicity parameter there are different lower limit of quantification (LLOQ). [redacted]
    - In data listings, the original reported value "<LLOQ" will be displayed.
    - For calculations (GMT, median…) the half of the true LLOQ value will be used for such values.
    - If another unexpected value is reported as LLOQ in the data than mentioned here, the half value of the reported LLOQ will be used for the calculations.
- For the different immunogenicity parameters there are different upper limits of quantification (ULOQ) [redacted]
    - In data listings, the original reported value ">ULOQ" will be displayed.
    - For calculations (GMT, median…) the ULOQ value will be used for such values.
    - If another unexpected value is reported as ULOQ in the data than mentioned here, this ULOQ value will be used for calculations.
- For analysis using interval-censored data an interval will be defined for every observation. Observations between LLOQ and ULOQ will be handled as interval with left limit and right limit being the observed value. For values <LLOQ the interval will be left-censored with the LLOQ as right limit. For values >ULOQ the interval will be right-censored with the ULOQ as left limit.

6. SAFETY ANALYSIS

All safety analyses will be performed for the safety population.

6.1 Extent of Exposure

6.1.1 *Data points*

The following information will be analyzed descriptively and corresponding details on the subject level will be provided in data listings:

- Treatment duration
- Study duration
- Study vaccine administration (will be listed only)

For details on table and listing content, see Section 7.

6.1.2 *Derivations and Definitions*

- Treatment duration [days] = date of last administered vaccine – date of first vaccination +1 (only if more than one vaccine administered)
- Study duration [days] = date of last attended visit (or date of study discontinuation, if applicable) – date of signed informed consent + 1

6.2 Adverse Events

6.2.1 *Data points*

The following analyses will be provided for all Adverse Events:

- Adverse event overview for solicited and unsolicited events (e.g. any AE, any SAE, any related AE).
- Solicited Adverse Events by symptom
- Solicited Adverse Events by symptom and severity
- Unsolicited Adverse Events by SOC and PT, overall and for specific types of Adverse Events (e.g. SAEs, related AEs)
- Unsolicited Adverse Events by severity and for specific types of Adverse Events
- Unsolicited Adverse Events by causality and for specific types of Adverse Events

For details on table and listing content, see Section 7.

6.2.2 *Derivations and Definitions*

- All solicited and unsolicited Adverse Events up to Day 56 (Visit 3) will be analyzed for the preliminary analysis and all AE in the database for the Final Analysis. The following rules will be applied to identify AEs up to Day 56 for preliminary analysis: In general, all AE with start date prior to or at Visit 3 are considered. If a subject has no Day 56 visit (Visit 3) then all AEs within 56 days from Visit 1 (Day 0) (Visit 1 + 56 days) are included. If start date of AE is incomplete (e.g. missing start day) and the AE could theoretically by within Visit 3, or if Visit 3 is not available, within 56 days from Visit 1 (Visit 1 + 56 days), it will be handled as AE up to Day 56 (Visit 3) and thus be included in the preliminary analysis. If start date of AE is unknown it will be handled as AE up to Day 56 visit (Visit 3) and thus be included in the preliminary analysis.
- Percentages for AE rates are based on N (treatment group totals).
- An Adverse Events is considered to be related to study treatment if “Relationship to Study Treatment” is answered with “definitely”, “probably” or “possibly” or the causality is missing.
- Tables showing “severe” events will include events with grade 3 or 4 or missing grade.
- In tables showing subjects with AEs by maximum severity, subjects will be counted only in the highest grading category (=”potentially life threatening”), but events will be counted in each reported grading category.

- In tables showing AEs by causality, subjects will be counted only in the strongest relationship category (="definitely"), but events will be counted in each category.

6.3 Laboratory Parameters

Data from the early termination visit or unscheduled visits will be listed only and will not be shown in summary tables by time point. Laboratory data from scheduled visits (VS, Visit 2 and Visit 3) will be tabulated by time point. Specifications on TLFs are provided in Section 7:

For details on table and listing content, see Section 7.

6.3.1 *Data points*

- Hematology
  - Hemoglobin
  - Hematocrit
  - Erythrocytes
  - Leukocytes
  - Neutrophils
  - Lymphocytes
  - Monocytes
  - Eosinophils
  - Basophils
  - Platelets
- Clinical Chemistry
  - Creatinine
  - Potassium
  - Sodium
  - Calcium
  - Aspartate Aminotransferase
  - Alanine Aminotransferase
  - Alkaline Phosphatase
  - Bilirubin
- Coagulation
  - Prothrombin Time
  - Activated Partial Thromboplastin Time
  - Fibrinogen
- Urinalysis
  - Specific gravity
  - pH

- Leukocytes
- Nitrite
- Protein
- Glucose
- Ketone
- Urobilinogen
- Bilirubin
- Erythrocytes

The following parameters will be analyzed descriptively by time point:

- Absolute values (summary statistics for hematology, clinical chemistry and coagulation)
- Absolute change from baseline (Screening) (summary statistics for hematology, clinical chemistry and coagulation)
- Number of subjects with values above/below normal range (for hematology, clinical chemistry and coagulation)
- Urinalysis results (summary statistics for quantitative parameters, frequency statistics for qualitative parameters)

6.3.2 *Derivations and Definitions*

- No conversion of laboratory parameters will be performed. The parameters listed above will be tabulated and listed in the units reported by the study site.
- Absolute change from baseline will be calculated as difference from value of the current visit and baseline value (screening).

6.4 Other Safety Parameters

6.4.1 *Data points*

Data from the early termination visit or unscheduled visits will be listed only and will not be shown in summary tables by time point. The following vital sign parameters will be summarized descriptively:

- Diastolic blood pressure
- Systolic blood pressure
- Pulse rate
- Temperature

The following data will be listed only:

- Pregnancy test
- Local tolerability
- Local tolerability – post vaccination

- Symptom-directed physical examination
- System-based physical examination

For details on table and listing content, see Section 7.

7. LIST OF TABLES, DATA LISTINGS AND FIGURES

7.1 List of Tables

No. for prel. Analysis	No. for FA	Legend	Content
Overall Subject Information			
1.1.1.1	2.1.1.1	Subject Overview (All subjects)	• Randomized • Safety Population • mITT Population • PP Population • Number of subjects that received first vaccination • Number of subjects that received second vaccination • Number of subjects that missed any vaccination
1.1.1.2	2.1.1.2	Protocol Deviations by Deviation Type (All subjects)	• Any major PD • Any major PD by category • Any minor PD Any minor PD by category
1.1.1.3	2.1.1.3	Number of Screening Failures and Reasons	• Number of screening failures • Reason for screening failure
1.1.1.4	2.1.1.4	Subjects by Visit (Safety Population)	
1.1.1.5	2.1.1.5	Study Status (Safety Population)	• Visit attendance status • Primary reason for early termination
1.1.2.4-1.1.2.5	2.1.2.4-2.1.2.5	Repeat tables x.1.1.4-x.1.1.5 for mITT population (if not the same as Safety Population)	
1.1.3.4-1.1.3.5	2.1.3.4-2.1.3.5	Repeat tables x.1.1.4-x.1.1.5 for PP population	
NOTE: Overall Subject Information tables will also be provided for final analysis including all information reported during the study			
Baseline Evaluation			
1.2.1.1	n/a	Summary Table of Demographic Information (Safety Population)	• Age • Gender • Race • Height [cm] • Weight [kg]
1.2.1.2	n/a	Vaccination History by and ATC Level 3 (Safety Population)	
1.2.1.3	n/a	Medical History Entry by SOC and PT (Safety Population)	
1.2.1.4	2.2.1.4	Prior Medications by ATC Level 2 and 3 (Safety Population)	
1.2.1.5	2.2.1.5	Concomitant Medications by ATC Level 2 and 3 (Safety Population)	
1.2.1.6	n/a	Vital Signs at Baseline (Safety Population)	• Systolic blood pressure [mmHg] • Diastolic blood pressure [mmHg] • Pulse rate [beats/min]

			• Temperature [C]
1.2.2.1-1.2.2.6	2.2.2.4-2.2.2.5	For preliminary analysis repeat tables 1.2.1.1-1.2.1.6 and for final analysis tables 2.2.1.4-2.2.1.5 for mITT population (if not the same as Safety Population).	
NOTE: Prior/Concomitant Medication tables will also be provided for final analysis but including all Prior/Concomitant Medications reported during the study			
Immunogenicity Analysis			
Zika-RSP VNT Antibody			
n/a	2.3.1.1	GMT of Zika-RSP VNT Antibody by Visit (mITT Population)	
n/a	2.3.1.2	GMT of Zika-RSP VNT Antibody based on interval-censored data by Visit (mITT Population)	
n/a	2.3.1.3	ANOVA for GMT of Zika-RSP VNT Antibody between treatment groups by Visit (mITT Population)	
n/a	2.3.1.4	Seroconversion rate for of Zika-RSP VNT Antibody by Visit (mITT Population)	
n/a	2.3.1.5	Positivity of Zika-RSP VNT Antibody by time point	
Zika-RSP ELISA Antibody			
n/a	2.3.1.6	GMT of Zika-RSP-ELISA Antibodies by Visit (mITT Population)	
n/a	2.3.1.7	GMT of Zika-RSP-ELISA Antibodies based on interval-censored data by Visit (mITT Population)	
n/a	2.3.1.8	ANOVA for GMT of Zika-RSP-ELISA Antibodies between treatment groups by Visit (mITT Population)	
n/a	2.3.1.9	Seroconversion rate of Zika-RSP-ELISA Antibodies by Visit (mITT Population)	
n/a	2.3.1.10	Positivity of Zika-RSP-ELISA Antibodies by time point	
Measles-ELISA antibodies			
n/a	2.3.1.11	GMT of Measles-ELISA Antibodies by Visit (mITT Population)	
n/a	2.3.1.12	GMT of Measles-ELISA Antibodies based on interval-censored data by Visit (mITT Population)	
n/a	2.3.1.13	ANOVA for GMT Measles-ELISA Antibodies between treatment groups by Visit (mITT Population)	
n/a	2.3.1.14	Seroconversion rate of Measles-ELISA Antibodies by Visit (mITT Population)	
n/a	2.3.1.15	Positivity of Measles-ELISA Antibodies by time point	

T cell immunity			
n/a	2.3.1.16	Median of SFC of T cell immunity-PBMCs by Visit (mITT Population)	
n/a	2.3.1.17	Positivity of SFC of T cell immunity-PBMCs by time point	
n/a	2.3.2.1-2.3.2.17	Repeat Immunogenicity tables 2.3.1.1-2.3.1.17 for PP Population.	
Safety Results			
Extent of Exposure			
1.4.1.1	2.4.1.1	Extent of Exposure (Safety Population)	• Study duration • Treatment duration
Adverse Events			
1.4.1.2	2.4.1.2	Summary Table of solicited and unsolicited Adverse Events (Safety Population)	Number of subjects with: • Any AEs • Any related AEs • Any severe AEs • Any related severe AEs • Any SAEs • Any related SAEs • Any medically attended AEs • Any related medically attended AEs • Any AEs leading to second vaccination not administered
1.4.1.3	2.4.1.3	Summary Table of solicited Adverse Events (Safety Population)	• Any solicited AEs • Any related solicited AEs • Any severe solicited AEs • Any related severe solicited AEs • Any solicited SAEs • Any related solicited SAEs • Any medically attended solicited AEs • Any related medically attended solicited AEs • Any solicited AEs leading to second vaccination not administered
1.4.1.4	2.4.1.4	Summary Table of solicited local Adverse Events (Safety Population)	• Any solicited local AEs • Any related solicited local AEs • Any severe solicited local AEs • Any related severe solicited local AEs • Any solicited local SAEs • Any related solicited local SAEs • Any medically attended solicited local AEs • Any related medically attended solicited local AEs • Any solicited local AEs leading to second vaccination not administered
1.4.1.5	2.4.1.5	Summary Table of solicited systemic Adverse Events (Safety Population)	• Any solicited systemic AEs • Any related solicited AEs • Any severe solicited AEs • Any related severe solicited systemic AEs

			• Any solicited systemic SAEs • Any related solicited systemic SAEs • Any medically attended solicited systemic AEs • Any related medically attended solicited systemic AEs • Any solicited systemic AEs leading to second vaccination not administered
1.4.1.6	2.4.1.6	Summary Table of unsolicited Adverse Events (Safety Population)	• Any unsolicited AEs • Any related unsolicited AEs • Any severe unsolicited AEs • Any serious unsolicited AEs • Any medically attended unsolicited AEs • Any related severe unsolicited AEs • Any related serious unsolicited AEs • Any related medically attended unsolicited AEs • Any unsolicited AEs leading to second vaccination not administered
1.4.1.7	2.4.1.7	Subjects with at least one Adverse Event classified by Maximum Severity (Safety Population)	
Solicited Adverse Events			
1.4.1.8	2.4.1.8	Subjects with solicited local Adverse Events by Symptom (Safety Population)	
1.4.1.9	2.4.1.9	Subjects with solicited systemic Adverse Events by Symptom (Safety Population)	
1.4.1.10	2.4.1.10	Subjects with solicited Adverse Events classified by Maximum Severity (Safety Population)	
1.4.1.11	2.4.1.11	Subjects with solicited local Adverse Events by Symptom classified by Maximum Severity (Safety Population)	
1.4.1.12	2.4.1.12	Subjects with solicited systemic Adverse Events by Symptom classified by Maximum Severity (Safety Population)	
Unsolicited Adverse Events			
1.4.1.13	2.4.1.13	Subjects with unsolicited Adverse Events by PT (Safety Population)	
1.4.1.14	2.4.1.14	Subjects with unsolicited Adverse Events by SOC and PT (Safety Population)	
1.4.1.15	2.4.1.15	Subjects with related unsolicited Adverse Events by SOC and PT (Safety Population)	
1.4.1.16	2.4.1.16	Subjects with severe unsolicited Adverse Events by SOC and PT (Safety Population)	

1.4.1.17	2.4.1.17	Subjects with serious unsolicited Adverse Events by SOC and PT (Safety Population)	
1.4.1.18	2.4.1.18	Subjects with medically attended unsolicited Adverse Events by SOC and PT (Safety Population)	
1.4.1.19	2.4.1.19	Subjects with related severe unsolicited Adverse Events by SOC and PT (Safety Population)	
1.4.1.20	2.4.1.20	Subjects with related serious unsolicited Adverse Events by SOC and PT (Safety Population)	
1.4.1.21	2.4.1.21	Subjects with related medically attended unsolicited Adverse Events by SOC and PT (Safety Population)	
1.4.1.22	2.4.1.22	Subjects with unsolicited Adverse Events leading to second Vaccination not administered (Safety Population)	
1.4.1.23	2.4.1.23	Subjects with unsolicited Adverse Events caused study discontinuation (Safety Population)	
1.4.1.24	2.4.1.24	Subjects with at least one unsolicited Adverse Event by Maximum Severity (Safety Population)	
1.4.1.25	2.4.1.25	Subjects with at least one medically attended unsolicited Adverse Event by Maximum Severity (Safety Population)	
1.4.1.26	2.4.1.26	Subjects with at least one unsolicited Adverse Event by Causality (Safety Population)	
1.4.1.27	2.4.1.27	Subjects with at least one medically attended unsolicited Adverse Event by Causality (Safety Population)	
1.4.1.28	2.4.1.28	Subjects with at least one serious unsolicited Adverse Event by Causality (Safety Population)	
NOTE: Adverse Event tables will also be provided for final analysis but including all AEs reported during the study. For the preliminary analysis the wording “up to Day 56” and for the final analysis “up to study end” will be added in the title of AE Table x.4.1.2-x.4.1.28.			
Laboratory Parameters			
1.4.1.29	n/a	Absolute Values for Hematology Parameters by Parameter and Visit (Safety Population)	
1.4.1.30	n/a	Absolute Values for Clinical Chemistry Parameters by Parameter and Visit (Safety Population)	

1.4.1.31	n/a	Absolute Values for Coagulation Parameters by Parameter and Visit (Safety Population)	
1.4.1.32	n/a	Absolute Changes from Baseline for Hematology Parameters by Parameter and Visit (Safety Population)	
1.4.1.33	n/a	Absolute Changes from Baseline for Clinical Chemistry Parameters by Parameter and Visit (Safety Population)	
1.4.1.34	n/a	Absolute Changes from Baseline for Coagulation Parameters by Parameter and Visit (Safety Population)	
1.4.1.35	n/a	Subjects with Hematology Parameters Outside Normal Range by Parameter and Visit (Safety Population)	
1.4.1.36	n/a	Subjects with Clinical Chemistry Parameters Outside Normal Range by Parameter and Visit (Safety Population)	
1.4.1.37	n/a	Subjects with Coagulation Parameters Outside Normal Range by Parameter and Visit (Safety Population)	
1.4.1.38	n/a	Urinalysis Results by Parameter and Visit (Safety Population)	
1.4.1.39	n/a	Absolute Changes from Baseline for pH and Specific Gravity by Visit (Safety Population)	
Other Safety Parameters			
1.4.1.40	n/a	Systolic Blood Pressure [mmHg] by Visit (Safety Population)	
1.4.1.41	n/a	Diastolic Blood Pressure [mmHg] by Visit (Safety Population)	
1.4.1.42	n/a	Pulse Rate [beats/min] by Visit (Safety Population)	
1.4.1.43	n/a	Body Temperature [°C] by Visit (Safety Population)	

7.2 List of Data Listings

No. for prel. Analysis	No. for FA	Legend	Content
Overall Subject Information			
1.1.1	2.1.1	Analysis Population Details	Subject number, Treatment, received IMP, Safety Population, mITT Population, PP Population, Reason not in PP Population, Randomization performed, Reason randomization not performed, randomization date/time, randomization number
1.1.2	2.1.2	Screening Failures with Reason	Subject number, Date of informed consent, Reason for screening failure (Subject's withdrawal of consent, In/Ex criterion not fulfilled / exclusion criterion fulfilled, Other), Specification other reason
1.1.3	2.1.3	Violated Entry Criteria	Subject number, Visit, Criterion ID not met, Criterion description
1.1.4	2.1.4	Attended Visits with Dates (Safety Population)	Subject number, Treatment, Visit, date, age, Reason for visit not performed, reason for outside time window, reason for unscheduled visit
1.1.5	2.1.5	Protocol Deviations (Safety Population)	Subject number, Treatment, PD Category, Classification, Reason for classification, PD Description
1.1.6	2.1.6	Study Status (Safety Population)	Subject number, Treatment, Visit attendance status, Primary reason early termination/treatment discontinuation, Other reason specification, AE Term, Specify individual stopping rules, AE term (Stopping rules), Primary cause of death, Death date, Date of study discontinuation
Baseline Evaluation			
1.2.1	n/a	Demographic Information (Safety Population)	Subject number, Treatment, Date of birth, Age [years], Gender, Childbearing potential, reason, specification other, Race, Specification other race
1.2.2	n/a	Full Physical Examination at Visit 0 (Safety Population)	Subject number, Treatment, Examination performed, Reason not performed, Exam Date
1.2.3	n/a	Medical History (Safety Population)	Subject number, Treatment, Condition, Preferred Term (MedDRA [Version]), SOC (MedDRA [Version]), Start date, End date, Ongoing at study end
1.2.4	2.2.4	Prior Medication (Safety Population)	Subject number, Treatment, Medication or therapy, ATC Level 2 (WHODD [Version]), ATC Level 3 (WHODD [Version]), Indication category, Indication, Dose, Dose Unit, Dose Unit other, Frequency, Frequency other, Route, Route other, Start Date, Ongoing at study end, End Date
1.2.5	2.2.5	Concomitant Medication (Safety Population)	Subject number, Treatment, Medication or therapy, ATC Level 2 (WHODD [Version]), ATC Level 3 (WHODD [Version]), Indication category, Indication, Dose, Dose Unit, Dose Unit other, Frequency, Frequency other, Route,

			Route other, Start Date, Ongoing at study end, End Date
1.2.6	n/a	Vaccination History (Safety Population)	Subject number, Treatment, Visit, Any vaccination history within the last three years prior to screening, Any new vaccination history since screening, Date, Vaccination/Indication, ATC Level 2 (WHODD [Version]), ATC Level 3 (WHODD [Version]),
1.2.7	n/a	Concomitant Procedure (Safety Population)	Subject number, Treatment, Procedure, Start/End date, Ongoing at study end, Indication category, Indication
1.2.8	n/a	Virology	Subject number, Treatment, Virology test, Test performed, Reason, Result, Date
Immunogenicity Analysis			
n/a	2.3.1	Functional Antibodies Assessment (VNT) (mITT Population)	Subject number, Treatment, Visit, Sample performed, Reason not performed, Sample ID, Date/Time of sample, Titer [IU/ml], Serconversion, positive/negative value
n/a	2.3.2	Functional Antibodies Assessment (ELISA) m(ITT Population)	Subject number, Treatment, Visit, Sample performed, Reason not performed, Sample ID, Date/Time of sample, Titer [IU/ml] , Serconversion, positive/negative value
n/a	2.3.3	Measles Antibodies Assessment (mITT Population)	Subject number, Treatment, Visit, Sample performed, Reason not performed, Sample ID, Date/Time of sample, Titer [IU/ml] , positive/negative value
n/a	2.3.4	T-cell details (mITT Population)	Subject number, Treatment, Visit, Sample performed, Reason not performed, Sample ID, Date/Time of sample, Titer [IU/ml] , positive/negative value
Safety Analysis			
Extent of Exposure			
1.4.1	2.4.1	Extent of Exposure (Safety Population)	Subject number, Treatment, Date first vaccination, Date second vaccination, Treatment duration [days], Date informed consent, Date of last attended visit, Study duration [days]
1.4.2	2.4.2	Study Medication Details (Safety Population)	Subject number, Treatment, Visit, Vaccination performed, Reason, Date/time
Adverse Events			
1.4.3	2.4.3	Adverse Events Part 1 (Safety Population)	Subject number, Treatment, AE Term, MedDRA PT, MedDRA SOC, AE type, Start date/time, End date/time, Medically attended, Severity, Serious AE, SAE Criteria, Relationship to Study Treatment
1.4.4	2.4.4	Adverse Events Part 2 (Safety Population)	Subject number, Treatment, AE Term, Action taken with Study Treatment, Other Action taken, specification other action taken, outcome, Ongoing at final examination
1.4.5	2.4.5	Unsolicited Adverse Events Part 1 (Safety Population)	Subject number, Treatment, AE Term, MedDRA PT, MedDRA SOC, AE type, Start date/time, End date/time, Medically attended, Severity,

			Serious AE, SAE Criteria, Relationship to Study Treatment
1.4.6	2.4.6	Unsolicited Adverse Events Part 2 (Safety Population)	Subject number, Treatment, AE Term, Action taken with Study Treatment, Other Action taken, specification other action taken, outcome, Ongoing at final examination
1.4.7	2.4.7	Related Unsolicited Adverse Events Part 1 (Safety Population)	See Listing x.4.5
1.4.8	2.4.8	Related Unsolicited Adverse Events Part 2 (Safety Population)	See Listing x.4.6
1.4.9	2.4.9	Severe Unsolicited Adverse Events Part 1 (Safety Population)	See Listing x.4.5
1.4.10	2.4.10	Severe Unsolicited Adverse Events Part 2 (Safety Population)	See Listing x.4.6
1.4.11	2.4.11	Serious Unsolicited Adverse Events Part 1 (Safety Population)	See Listing x.4.5
1.4.12	2.4.12	Serious Unsolicited Adverse Events Part 2 (Safety Population)	See Listing x.4.6
1.4.13	2.4.13	Medically attended unsolicited Adverse Events Part 1 (Safety Population)	See Listing x.4.5
1.4.14	2.4.14	Medically attended unsolicited Adverse Events Part 2 (Safety Population)	See Listing x.4.6
1.4.15	2.4.15	Unsolicited Adverse Events caused Study Discontinuation Part 1 (Safety Population)	See Listing x.4.5
1.4.16	2.4.16	Unsolicited Adverse Events caused Study Discontinuation Part 2 (Safety Population)	See Listing x.4.6
1.4.17	2.4.17	Unsolicited Adverse Events leading to second Vaccination not administered Part 1 (Safety Population)	See Listing x.4.5
1.4.18	2.4.18	Unsolicited Adverse Events leading to second Vaccination not administered Part 2 (Safety Population)	See Listing x.4.6
1.4.19	2.4.19	Solicited Adverse Events Part 1 (Safety Population)	Subject number, Treatment (Safety), Record ID, AE Term, Start date/time, End date/time, Medically attended, Severity, Serious AE, SAE Criteria, Causality
1.4.20	2.4.20	Solicited Adverse Events Part 2 (Safety Population)	Subject number, Treatment (Safety), Record ID, AE Term, Action taken with Study Treatment, Other Action taken, Outcome, Ongoing at final examination, Caused study discontinuation
1.4.21	2.4.21	Related Solicited Adverse Events Part 1 (Safety Population)	See Listing x.4.19
1.4.22	2.4.22	Related Solicited Adverse Events Part 2 (Safety Population)	See Listing x.4.20
1.4.23	2.4.23	Severe Solicited Adverse Events Part 1 (Safety Population)	See Listing x.4.19
1.4.24	2.4.24	Severe Solicited Adverse Events Part 2 (Safety Population)	See Listing x.4.20

1.4.25	2.4.25	Serious Solicited Adverse Events Part 1 (Safety Population)	See Listing x.4.19
1.4.26	2.4.26	Serious Solicited Adverse Events Part 2 (Safety Population)	See Listing x.4.20
1.4.27	2.4.27	Medically attended Solicited Adverse Events Part 1 (Safety Population)	See Listing x.4.19
1.4.28	2.4.28	Medically attended Solicited Adverse Events Part 2 (Safety Population)	See Listing x.4.20
1.4.29	2.4.29	Solicited Adverse Events caused Study Discontinuation Part 1 (Safety Population)	See Listing x.4.19
1.4.30	2.4.30	Solicited Adverse Events caused Study Discontinuation Part 2 (Safety Population)	See Listing x.4.20
1.4.31	2.4.31	Solicited Adverse Events leading to second Vaccination not administered Part 1 (Safety Population)	See Listing x.4.19
1.4.32	2.4.32	Solicited Adverse Events leading to second Vaccination not administered Part 2 (Safety Population)	See Listing x.4.20
Laboratory Parameters			
1.4.33	n/a	Hematology Values outside Normal Range (Safety Population)	Subject number, Treatment, Visit, Date/Time, Parameter, Value, Unit, Not done, Reason not done, Clinically significant, Likely cause, Change from baseline
1.4.34	n/a	Clinical Chemistry Values outside Normal Range (Safety Population)	Subject number, Treatment, Visit, Date/Time, Parameter, Value, Unit, Not done, Reason not done, Clinically significant, Likely cause, Change from baseline
1.4.35	n/a	Coagulation Values outside Normal Range (Safety Population)	Subject number, Treatment, Visit, Date/Time, Parameter, Value, Unit, Not done, Reason not done, Clinically significant, Likely cause, Change from baseline
1.4.36	n/a	Abnormal and Positive Urinalysis Values (Safety Population)	Subject number, Treatment, Visit, Date/Time, Parameter, Value, Unit, Not done, Reason not done, Clinically significant, Abnormal finding, Likely cause
Other Safety Parameters			
1.4.37	n/a	Vital Signs (Safety Population)	Subject number, Treatment, Visit, visit specifier, visit specifier, Vital signs collected, Reason not collected, Date/Time, Test, Result, Unit, Clinically significant, not done
1.4.38	n/a	Symptom-directed Physical Exam (Safety Population)	Subject number, Treatment, Exam performed, Reason not performed, Exam date/Time
1.4.39	n/a	System Based Physical Exam (Safety Population)	Subject number, Treatment, Exam performed, Reason not performed, other reason, Exam date/time
1.4.40	n/a	Local Tolerability (Safety Population)	Subject number, Treatment, visit, visit specifier, Assessment performed, Reason not performed, Exam Date/Time

1.4.41	n/a	Pregnancy test (Safety Population)	Subject number, Treatment, visit, Pregnancy test performed, Reason not performed, Date/time, Result

8. REFERENCES

Not applicable.