Immunogenicity and Safety of a Purified Vero Rabies Vaccine – Serum Free in Comparison with Verorab® and Imovax® Rabies, in a Pre-exposure Regimen in Both Pediatric and Adult Populations and a Single Booster Dose of Purified Vero Rabies Vaccine – Serum Free Administered at 1 Year Post-3-Dose Primary Series, and between 2 up to 3 Years Post-One Week 2-Dose Primary Series in a Subset of Adults in Thailand

Multi-center, observer-blind, controlled, randomized, Phase III study in 1700 subjects; ie, 505 subjects aged 1 year to <18 years and 1195 adult subjects aged ≥ 18 years. Subjects will receive 2 or 3 vaccine injections in a primary series.

Booster phase will be conducted in a blinded manner which include a subset of 170 adult subjects who will receive a single booster dose of Purified Vero Rabies Vaccine – Serum Free 1 year post-3-dose primary series, in addition to be conducted in an open-label manner which include a subset of 230 adult subjects who will receive a single booster dose of Purified Vero Rabies Vaccine – Serum Free between 2 up to 3 years post-one week 2-dose primary series, regardless of the vaccine used in the primary series.

Statistical Analysis Plan (SAP) - Core Body Part

Trial Code:	VRV12
Development Phase:	Phase III
Sponsor:	Sanofi Pasteur 14 Espace Henry Vallée, 69007 Lyon, France
Investigational Product(s):	Purified Vero Rabies Vaccine - Serum Free (VRVg): Purified inactivated rabies vaccine prepared on Vero cell line
Form / Route:	Freeze-dried + solvent/Intramuscular
Indication For This Study:	Pre-exposure regimen in pediatric and adult populations
Version and date of the protocol:	Version 8.0 dated 12JUN2023
Version and Date of the SAP core body part:	Version 1.0 dated 14AUG2023

Table of Contents

List of Tables

List of Abbreviations

ACIP	Advisory committee on immunization practices
AE	adverse event
AESI	adverse event of special interest
AR	adverse reaction
BL	Blood sampling
CI	confidence interval
COVID-19	Coronavirus Disease 2019
CRB	(electronic) case report book [all the case report forms for a subject]
CSR	Clinical Study Report
D	Day
DC	diary card
DNA	Deoxyribonucleic acid
eCRF	electronic case report form
FAS	Full Analysis Set
FASB1	Full Analysis Set for Booster at M12
FASB2	The Full Analysis Set for Booster at M24 up to M36
FASI	Full Analysis Set for Immunogenicity
FASIB1	Full Analysis Set for Immunogenicity for Booster at M12
FASIB2	Full Analysis Set for Immunogenicity for Booster at M24 up to M36
FASIP	Full Analysis Set for Immunogenicity for Immunogenicity Persistence
FASP	Full Analysis Set for Immunogenicity Persistence
FDA	Food and Drug Administration
GM	geometric mean
GMT	geometric mean of titers
GMTR	geometric mean of titer ratio
HDCV	human diploid cell vaccine
ID	intradermal
IM	intramuscular
IRT	Interactive response technology
IU	international unit
LLOQ	lower limit of quantitation
LLT	lowest level term
MA	Memory aid
MD	missing data
MedDRA	Medical Dictionary for Regulatory Activities
NA	not applicable

NI	non-inferiority
NM	non-measurable
PEP	post-exposure prophylaxis
PrEP	pre-exposure prophylaxis
PPAS	Per-Protocol Analysis Set
PT	preferred term
PVRV	Purified Vero rabies vaccine
Q1; Q2; Q3	first quartile; second quartile (median); third quartile
RCDC	Reverse Cumulative Distribution Curve
RFFIT	Rapid Fluorescent Focus Inhibition test
RVNA	rabies virus neutralizing antibody
SAE	serious adverse event
SafAS	safety analysis set
SafASB	safety analysis set for booster
SAP	Statistical Analysis Plan
SD	standard deviation
SF	serum free
SOC	system organ class (primary)
TLF	table(s), listing(s), and figure(s)
ULOQ	upper limit of quantitation
US	United States
V	visit
VAC	vaccination
WHO	World Health Organization

1 Introduction

Currently, Sanofi Pasteur has 2 rabies vaccines registered worldwide and available on the market: Imovax® Rabies and Verorab®. Imovax Rabies, a human diploid cell vaccine (HDCV), was first licensed in 1975. It has been registered and marketed in 15 countries, including the United States (US), Canada, Australia and 9 European countries. On the other hand, a purified Vero rabies vaccine (PVRV), was first licensed in France in 1985 under the commercial name of Verorab and is extensively registered worldwide in 80 countries including 10 European countries but not in the US, Canada, and Australia. Both vaccines have a well-defined safety and immunogenicity profile.

In an effort to further improve the available rabies vaccines and optimize the manufacturing process and life cycle management, Sanofi Pasteur is developing a Purified Vero Rabies Vaccine – serum free (SF), henceforth referred to as VRVg.

VRVg is issued from the Wistar Rabies Pitman Moore/WI 38 1503-3M strain. VRVg is highly purified with very low residual deoxyribonucleic acid (DNA) content (< 100 pg per dose) and it is produced without raw material derived from human or animal origin, and without antibiotics. VRVg is compliant with standards set by the European Union Pharmacopoeia, the World health Organization (WHO), and the US Food and Drug Administration (FDA).

VRVg constitutes an improvement of Verorab [redacted]

VRVg development has been conducted through stepwise adjustments based on various regulatory, pharmaceutical, and /or clinical rationales. Two formulations referred to as VRVg-1 and VRVg-2, have been subsequently explored and tested in overall 6 clinical studies. [redacted] Sanofi Pasteur made the decision to modify the initial formulation of VRVg-1 in order to ensure an enhanced vaccine immune response. Following the results with the second formulation, VRVg-2, and the acceptance of the health authorities to go to phase III, next step consists of 2 Phase III non-inferiority studies: VRV13 and the present study VRV12. Both studies compared VRVg-2 with the 2 rabies vaccines marketed by Sanofi Pasteur (Verorab® and Imovax® Rabies vaccines): in a simulated post-exposure (PEP) Essen (5-doses) intramuscular (IM) regimen in adults (VRV13); and in a pre-exposure prophylaxis (PrEP) IM regimen comprising adult and pediatric populations (the present VRV12 study).

2 Trial Objectives

2.1 Primary Objective

Immunogenicity

To demonstrate that VRVg-2 is non-inferior to Verorab and Imovax Rabies vaccines in each age group (pediatric and adult populations, respectively) when administered as a 3-dose PrEP regimen, in terms of proportion of subjects achieving an RVNA titer ≥ 0.5 IU/mL at D42, ie, 14 days after the 3rd injection (for Cohort 1).

2.2 Secondary Objectives

Immunogenicity

1) To demonstrate that the observed proportion of overall subjects (pooled pediatric and adult subjects) in the VRVg-2 group achieving an RVNA titer ≥ 0.5 IU/mL at D42 is at least 99%, with a lower limit of the 95% confidence interval (CI) of at least 97%, only if the primary objective is achieved
2) To demonstrate that VRVg-2 is non-inferior to Verorab and Imovax Rabies vaccines in each age group (pediatric and adult populations, respectively), in terms of proportion of subjects achieving an RVNA titer ≥ 0.5 IU/mL at D28, ie, 21 days after the 2nd injection, only if the 1st secondary immunogenicity objective is achieved
3) To demonstrate that 2-dose VRVg-2 at D28 is non-inferior to 3-dose Imovax Rabies at D42 in each age group (pediatric and adult populations, respectively), in terms of proportion of subjects achieving an RVNA titer ≥ 0.5 IU/mL, only if the 2nd secondary immunogenicity objective is achieved
4) To demonstrate that the observed proportion of overall subjects (pooled pediatric and adult subjects) in the VRVg-2 group achieving an RVNA titer ≥ 0.5 IU/mL at D28 is at least 99%, with a lower limit of the 95% CI of at least 97%, only if the 3rd secondary immunogenicity objective is achieved
5) To demonstrate that 2-dose Imovax Rabies at D28 is non-inferior to 3-dose Imovax Rabies at D42 in the overall subjects (pooled pediatric and adult subjects) in Cohort 1, in terms of proportion of subjects achieving an RVNA titer ≥ 0.5 IU/mL, only if the 4th secondary immunogenicity objective is achieved
6) To describe the immune response induced by VRVg-2 versus Verorab and Imovax Rabies vaccines at D28 (ie, 21 days after the 2nd injection), and at D42 (ie, 14 days after the 3rd injection) in all age groups (pediatric and adult populations, respectively)
7) To describe the immune response induced by VRVg-2 at D14 after a single booster dose of VRVg-2 administered at M12 after the primary series with VRVg-2, Verorab, or Imovax Rabies vaccines in the subset of adult subjects (for Cohort 1)

8) To describe persistence of the immune response at M6, M12, M18, and pre-booster between M24 up to M36 post primary series vaccination in the subset of adults who are randomized to receive a booster and completed the 2-dose vaccination in the primary series (for Cohort 2)

9) To describe the immune response induced by VRVg-2 at D14 after a single booster dose of VRVg-2 administered between M24 up to M36 after the primary series with VRVg-2, Verorab, or Imovax Rabies vaccines in the subset of adult subjects who are randomized to receive a booster and completed the 2-dose vaccination in the primary series (for Cohort 2)

Safety

1) To describe the safety profile of VRVg-2 versus Verorab and Imovax Rabies vaccines, after each vaccine injection, in each age group (for Cohort 1 and Cohort 2).

2) To describe the safety of a single booster dose of VRVg-2 among the subset of adults following primary series of 3-dose in addition to booster dose at M12 (for Cohort 1).

3) To describe the safety of a single booster dose of VRVg-2 among the subset of adults following one week 2-dose primary series in addition to booster dose between M24 up to M36 (for Cohort 2).

3 Description of the Overall Trial Design and Plan

3.1 Trial Design

This will be a multi-center, observer-blind, controlled, randomized, Phase III study in 1700 healthy subjects (505 pediatric subjects [aged 1 year to <18 years] and 1195 adult subject [aged ≥18 years]). Pediatric subjects will be enrolled in Cohort 1 only, and adult subjects will be enrolled in two cohorts (Cohorts 1 and 2). Pediatric subjects (505 subjects) and adult subjects (505 subjects) in Primary Series Cohort 1 will be vaccinated according to a primary series of 3 vaccine injections of VRVg-2 (Group 1), Verorab vaccine (Group 2), or Imovax Rabies vaccine (Group 3), at day [D] 0, D7, and D28. Adult subjects in Primary Series Cohort 2 will be vaccinated according to a one week 2-dose schedule PrEP regimen of either VRVg-2 (Group 4), Verorab vaccine (Group 5), or Imovax Rabies vaccine (Group 6) at D0 and D7. A subset of 170 adult subjects from Primary Series Cohort 1 will receive a booster dose of VRVg-2 at M12 regardless of the vaccine use in the primary series. A subset of 230 adult subjects from Primary Series Cohort 2 will be followed up for evaluation of immunogenicity persistence after primary series (including blood sample collection at M6, M12, M18, and pre-booster between M24 up to M36) and will receive a booster dose of VRVg-2 between M24 up to M36. Vaccination of primary series and booster dose will be administered through the IM route.

Subjects will be randomized in 3:1:1 (Groups 1, 2, and 3 in Cohort 1; and Groups 4, 5, and 6 in Cohort 2, respectively) with the resulting number of subjects assigned to each study group shown in Table 3.1.

Table 3.1: Distribution of subjects according to vaccination group

			Vaccine	Number of adult subjects	Number of pediatric subjects
Primary Series	Cohort 1 3-dose PrEP regimen	Group 1	VRVg-2	303	303
		Group 2	Verorab®	101	101
		Group 3	Imovax® Rabies	101	101
	Cohort 2 one week 2-dose PrEP regimen	Group 4	VRVg-2	414	NA
		Group 5	Verorab®	138	NA
		Group 6	Imovax® Rabies	138	NA
Booster Phase	Cohort 1 (M12)	Group 1	VRVg-2 (primed with VRVg-2)	102	NA
		Group 2	VRVg-2 (primed with Verorab®)	34	NA
		Group 3	VRVg-2 (primed with Imovax® Rabies)	34	NA
	Cohort 2 (M24 up to M36)	Group 4	VRVg-2 (primed with VRVg-2)	138	NA
		Group 5	VRVg-2 (primed with Verorab®)	46	NA
		Group 6	VRVg-2 (primed with Imovax® Rabies)	46	NA

3.2 Trial Plan

The trial plan is summarized in tables of study procedures, given by Table 3.2 (primary series Cohort 1), Table 3.3 (primary series and booster phase Cohort 1), Table 3.4 (primary series Cohort 2) and Table 3.5 (primary series, immunogenicity persistence and booster phase Cohort 2). The randomization and vaccinations during the primary series and booster phase for Cohort 1 will be conducted in a blinded manner. For Cohort 2, randomization and vaccinations from the enrollment up to V04 (D35, 28 days after the last vaccination in the primary series) will be conducted in a blinded manner, and then the study data will be unblinded for the first statistical analysis as defined in Section 5.4.

Table 3.2 Table of study procedures: 3-Dose Primary Series (Cohort 1)

Phase III Trial, 5 Visits, 1 Phone Call, 3 Vaccinations, 3 Blood Samples, 7 Months Period per Subject

Visit	**V01**	**V02**	**V03**	**V04**	**V05**	**Phone Call**
Visit interval	**VAC1**	**VAC2= VAC1+7D**	**VAC3 = VAC1+28D**	**VAC3+14D**	**VAC3+28D**	**VAC3*+6M**
Indicative Days (D)/Months (M)	**D0**	**D7 +1D**	**D28 ±3D**	**D42 ±3D**	**D56 ±3D**	**M7 +14D**
Informed consent signed†	√					
Demographic data	√					
Urine pregnancy test‡	√	√	√			
Physical examination§	√	√	√	√	√	
Past and current significant medical history	√					
Inclusion & exclusion criteria	√					
Randomization / IRT call	√	√	√			
Blood sampling for serology**	**BL01**		**BL02**	**BL03**		
Vaccine injection	√	√	√			
30-minute observation period	√	√	√			
Diary Card (DC) **Memory Aid (MA)**						
Provided	DC1	DC2	DC3		MA1	
Checked		DC1	DC2	DC3	DC3	MA1
Collected		DC1	DC2		DC3	
Injection site reactions and Systemic Event Assessment	√	√	√	√	√	
Temporary contraindications		√	√			
Definitive contraindications		√	√			
Reportable concomitant medication	√	√	√	√	√	
Termination Record					√	
Pregnancy cases	Collected throughout the entire study period					
SAEs and AESIs	Collected throughout the entire study period					

* VAC3 or last vaccine injection in the event of early terminated subject contacted for the 6 months follow-up period through a phone call

†In addition if applicable, one AF has to be signed by subjects as required by local Ethics Committee or country regulations

‡ For subjects of childbearing potential

§ Complete physical examination at V01 and medically-driven physical examination for remaining visits

** Blood sample volume drawn from subjects aged 1 year to <2 years will be 3 mL, for subjects ≥2 years to <18 years will be 5 mL, and for subjects ≥18 years will be 6 mL.

Table 3.3 Table of study procedures: 3-Dose Primary Series + M12 Booster - Adult Subset (Cohort 1)

Phase III Trial, 8 Visits, 2 Phone Calls, 3 Vaccinations, 1 Booster dose, 5 Blood Samples, approximately 18 Months Period per Subject

Visit	**V01**	**V02**	**V03**	**V04**	**V05**	**Phone Call**	**V06**	**V07**	**V08**	**Phone Call**
Visit interval	**VAC1**	**VAC2= VAC1+7D**	**VAC3 = VAC1+28D**	VAC3+14D	VAC3+28D	VAC3*+6M	**Booster VAC4= VAC1+12M**	**D14 post booster dose VAC4+14D**	**D28 post booster dose VAC4+28D**	**6 months FU post booster dose VAC4+6M**
Indicative Days (D)/Months (M)	**D0**	**D7 +1D**	**D28 ±3D**	**D42 ±3D**	**D56 ±3D**	**M7 +14D**	**M12 +14D**	**M12+D14 +1D**	**M12+D28 ±3D**	**M18 +14D**
Informed consent signed	√									
Demographic data	√									
Urine pregnancy test‡	√	√	√				√			
Physical examination§	√	√	√	√	√		√		√	
Past and current significant medical history	√						√			
Inclusion & exclusion criteria	√									
Randomization / IRT call	√	√	√							
Blood sampling for serology**	**BL01**		**BL02**	**BL03**			**BL04**	**BL05**		
Vaccine injection	√	√	√				√			
30-minute observation period	√	√	√				√			
Diary Card (DC) **Memory Aid (MA)** **Provided** **Checked**	DC1	DC2 DC1	DC3 DC2	DC3	MA1 DC3	MA1	DC4 MA1	DC4	MA2 DC4	MA2

Visit	V01	V02	V03	V04	V05	Phone Call	V06	V07	V08	Phone Call
Visit interval	VAC1	VAC2= VAC1+7D	VAC3 = VAC1+28D	VAC3+14D	VAC3+28D	VAC3*+6M	Booster VAC4= VAC1+12M	D14 post booster dose VAC4+14D	D28 post booster dose VAC4+28D	6 months FU post booster dose VAC4+6M
Indicative Days (D)/Months (M)	D0	D7 +1D	D28 ±3D	D42 ±3D	D56 ±3D	M7 +14D	M12 +14D	M12+D14 +1D	M12+D28 ±3D	M18 +14D
Collected		DC1	DC2		DC3		MA1		DC4	
Injection site reactions and Systemic Event Assessment	√	√	√	√	√		√			
Temporary contraindications		√	√				√			
Definitive contraindications		√	√				√			
Reportable concomitant medication	√	√	√	√	√		√	√	√	
Termination Record									√	
Pregnancy cases	Collected throughout the entire study period									
SAEs and AESIs	Collected throughout the entire study period									

* VAC3 or last vaccine injection in the event of early terminated subject contacted for the 6 months follow-up period through a phone call

‡ For subjects of childbearing potential

§ Complete physical examination at V01 and medically-driven physical examination for remaining visits

** Blood sample volume drawn will be 6 mL.

Table 3.4 Table of study procedures: One week 2-Dose Primary Series - Adults (Cohort 2)

Phase III Study, 4 Visits, 1 Phone Call, 2 Vaccinations, 2 Blood Samples, approximately 7 Months Period per Subject

Visit	**V01**	**V02**	**V03**	**V04**	**Phone Call‡**
Visit interval	**VAC1**	**VAC2= VAC1+7D**	**VAC1+28D**	**VAC2+28D**	**VAC2*+6M**
Indicative Days (D)	**D0**	**D7 +1D**	**D28 ±3D**	**D35 ±3D**	**~M7 +14D**
Informed consent signed	√				
Demographic data	√				
Urine pregnancy test‡	√	√			
Physical examination§	√	√	√	√	
Past and current significant medical history	√				
Inclusion & exclusion criteria	√				
Randomization / IRT call	√	√			
Blood sampling for serology**	**BL01**		**BL02**		
Vaccine injection	√	√			
30-minute observation period	√	√			
Diary Card (DC) **Memory Aid (MA)** **Provided** **Checked** **Collected**	DC1	DC2 DC1 DC1	DC2 DC2	MA1	MA1
Injection site reactions and Systemic Event Assessment	√	√	√	√	
Temporary contraindications		√			
Definitive contraindications		√			
Reportable concomitant medication	√	√	√	√	
Termination Record				√	
Pregnancy cases	Collected throughout the entire study period				
SAEs and AESIs	Collected throughout the entire study period				

* VAC2 or last vaccine injection in the event of early terminated subject contacted for the 6 months follow-up period through a phone call

‡ For subjects of childbearing potential

§ Complete physical examination at V01 and medically-driven physical examination for remaining visits

**Blood sample volume drawn from subjects ≥ 18 years will be 6 mL

‡ Administrative calls: in the interim between 6-month phone call after last vaccination in the primary series and booster dose, the study staff is encouraged (but not obligated) to contact the participants to check on their general well-being and interest to continue with the study

Table 3.5 Table of study procedures: One week 2-Dose Primary Series + Blood Samples for Immunogenicity Persistence at M6, M12, M18, pre-booster M24 up to M36 + M24 up to M36 Booster – Adult Subset (Cohort 2)

Phase III Study, 10 Visits, 1 Phone Call, 2 Vaccinations, 1 Booster Dose, 7 Blood Samples, approximately up to 42 Months Period per Subject

Visit	**V01**	**V02**	**V03**	**V04**	**V05**	**V06**	**V07**	**V08**	**V09**	**V10**	**Phone Call**
Visit interval	**VAC1**	**VAC2= VAC1+7D**	VAC1+28D	VAC2+28D	VAC2+6M	VAC2+12M	VAC2+18M	**Booster VAC3= VAC1+24M up to 36M**	**D14 post-booster dose VAC3+14D**	**D28 post-booster dose VAC3+28D**	**6-month follow-up post-booster dose VAC3+6M**
Indicative days (D)	**D0**	**D7 +1D**	**D28 ±3D**	**D35 ±3D**	**M6 +14D**	**M12 +14D**	**M18 +14D**	**M24 up to M36 +14D**	**[M24 up to M36+14D] +14D +1D**	**[M24 up to M36+14D] +D28 ±3D**	**M30 up to M42+14D**
Informed consent signed	√										
Demographic data	√										
Urine pregnancy test‡	√	√						√			
Physical examination§	√	√	√	√				√		√	
Past and current significant medical history	√							√			
Inclusion & exclusion criteria	√										
Randomization/ IRT call	√	√									
Blood sampling for serology**	**BL01**		**BL02**		**BL03**	**BL04**	**BL05**	**BL06**	**BL07**		
Vaccine injection	√	√						√			

Visit	**V01**	**V02**	**V03**	**V04**	**V05**	**V06**	**V07**	**V08**	**V09**	**V10**	**Phone Call**
Visit interval	**VAC1**	**VAC2= VAC1+7D**	VAC1+28D	VAC2+28D	VAC2+6M	VAC2+12M	VAC2+18M	**Booster VAC3= VAC1+24M up to 36M**	**D14 post-booster dose VAC3+14D**	**D28 post-booster dose VAC3+28D**	**6-month follow-up post-booster dose VAC3+6M**
Indicative days (D)	**D0**	**D7 +1D**	**D28 ±3D**	**D35 ±3D**	**M6 +14D**	**M12 +14D**	**M18 +14D**	**M24 up to M36 +14D**	**[M24 up to M36+14D] +14D +1D**	**[M24 up to M36+14D] +D28 ±3D**	**M30 up to M42+14D**
30-minute observation period	√	√						√			
Diary Card (DC)											
Memory Aid (MA)											
Provided	DC1	DC2						DC3		MA	
Checked		DC1	DC2	DC2					DC3	DC3	MA
Collected		DC1		DC2						DC3	
Diary Card (DC) SAE Follow-Up											
Provided				DC SAE							
Checked					DC SAE	DC SAE	DC SAE	DC SAE			
Collected								DC SAE			
Injection site reactions and Systemic Event Assessment	√	√	√	√				√			
Temporary contraindications		√						√			
Definitive contraindications		√						√			
Reportable concomitant medication	√	√	√	√	√†	√†	√†	√	√	√	
Termination Record										√	

Visit	**V01**	**V02**	**V03**	**V04**	**V05**	**V06**	**V07**	**V08**	**V09**	**V10**	**Phone Call**
Visit interval	**VAC1**	**VAC2= VAC1+7D**	VAC1+28D	VAC2+28D	VAC2+6M	VAC2+12M	VAC2+18M	**Booster VAC3= VAC1+24M up to 36M**	**D14 post-booster dose VAC3+14D**	**D28 post-booster dose VAC3+28D**	**6-month follow-up post-booster dose VAC3+6M**
Indicative days (D)	**D0**	**D7 +1D**	**D28 ±3D**	**D35 ±3D**	**M6 +14D**	**M12 +14D**	**M18 +14D**	**M24 up to M36 +14D**	**[M24 up to M36+14D] +14D +1D**	**[M24 up to M36+14D] +D28 ±3D**	**M30 up to M42+14D**
Pregnancy cases	Collected throughout the entire study period***										
SAEs and AESIs	Collected throughout the entire study period***										

‡ For subjects of childbearing potential
§ Complete physical examination at V01 and medically-driven physical examination for remaining visits
** Blood sample volume drawn will be 6 mL
† Only for concomitant medication belonging to categories 2 or 3 (refer to Section 6.9 of protocol V6.0)
***Information about SAEs, AESIs, and cases of pregnancy will be recorded until 6 months after the primary series and until 6 months after the booster dose for adults in Cohort 2. Between the 6-month follow-up visit after primary series (V05) and booster dose visit (V08), AESIs will not be collected, only fatal SAEs and related SAEs will be collected.

Blood Sampling:

Pediatric and adult subjects in Primary Series Cohort 1 will provide 3 blood samples: at D0 (baseline, prior to the first vaccine injection), at D28 (21 days after the second vaccine injection, and prior to the third vaccination), and at D42 (14 days after the third vaccine injection). Adult subjects in Primary Series Cohort 2 will provide 2 blood samples: at D0 (baseline, prior to the first vaccine injection), and D28 (21 days after the second dose vaccine injection).

The subsets of adult subjects who will be part of the booster phase will have 2 and 5 additional blood samples respectively: at M12 (prior to the booster dose injection), and at M12 +D14 (14 days after the booster dose injection) for Booster Phase Cohort 1; and at M6, M12, M18, pre-booster between M24 up to M36 (prior to the booster vaccine injection), and between M24 up to M36 +D14 (14 days after booster vaccine injection) for Immunogenicity Persistence and Booster Phase Cohort 2.

Collection of Safety Data:

Safety information will be recorded in all subjects participated primary series and/or booster phase, during the vaccination period and up to 28 days after vaccination, in terms of occurrence of adverse events (AEs), serious adverse events (SAEs), and adverse events of special interest (AESIs). In addition, SAEs and AESIs will be collected up to 6 months after the last vaccination in the primary series for the subjects who will not receive the booster vaccination. For the subset of adult subjects who will receive the booster vaccination, the SAEs and AESIs will be collected up to 6 months after the booster vaccination (ie, up to M18 for booster phase of Cohort 1 or M30 up to M42 for booster phase Cohort 2). Only fatal or related SAEs will be collected from the end of the primary series 6 months follow-up until the booster vaccination for the subsets of adult subjects in the Immunogenicity Persistence and Booster Phase Cohort 2 (see Table 3.5).

4 Endpoints and Assessment Methods

4.1 Primary Endpoints and Assessment Methods

See Section 9.1 of the protocol.

4.2 Secondary Endpoints and Assessment Methods

See Section 9.2 of the protocol.

4.3 Derived Endpoints: Calculation Methods

4.3.1 Safety

The following terms are used in the standard safety tables to describe the safety events.

- AE: Adverse event includes solicited reaction and unsolicited event (including non-serious and serious adverse events).
- Adverse Reaction (AR): Corresponds to AE related to the study vaccines (investigational product), unless otherwise specified.
- Immediate AE: Unsolicited systemic AE checked "Yes" in the field of "immediate (within 30 minutes from the vaccination)" by the Investigator in the electronic Case Report Form (eCRF).
- Solicited reaction: Event pre-listed in the eCRF, and which occurred during the solicited period (within 7 days after the day of vaccination).
- Unsolicited AE: AE recorded in the eCRF as Unsolicited Systemic Events or Unsolicited Injection Site Reactions.
    - Unsolicited vaccination injection site reactions are to be considered as related to the vaccine injection and therefore analyzed as ARs.
    - Unsolicited AEs will be analyzed up to 28 days after any and each vaccination (primary series and booster phase). Unsolicited AEs occurring before or after the defined period will not be presented in the summary for unsolicited AEs but will be presented in a separate listing. The exceptions are SAEs and AESIs which are unsolicited AEs collected during the whole study period (from the first vaccination to the end of 6 months safety follow-up period).
- SAE: Unsolicited AE considered serious by the investigator (reconciled with Global Pharmacovigilance database).

- AESI: Is one of scientific and medical concern specific to the Sponsor's product or program, for which ongoing monitoring and rapid communication by the Investigator to the Sponsor can be appropriate (reconciled with Global Pharmacovigilance database). AESIs defined in the protocol are:
  - Anaphylactic reaction
  - Encephalitis
  - Convulsion

4.3.1.1 Solicited Reactions

4.3.1.1.1 Daily Intensity

All daily records for solicited reactions will be derived into daily intensity according to the following classification: None, Grade 1, Grade 2, Grade 3, or Missing (Unknown).

For the derivation of daily intensities, the following sequential steps will be applied:

1) Solicited reactions (except Fever/Pyrexia) with eCRF presence recorded as "No" and with all daily records missing (Unknown) then all daily intensities will be derived as None.
2) For a temperature partially missing after decimal point, the data will be analyzed replacing "MD" (missing data) by zero. For example, a "39.MD" daily temperature will be considered as "39.0°C" at the time of analysis.
3) For non-measurable solicited reactions, daily intensities will correspond to daily records reported in the clinical database. For measurable solicited reactions the daily measurements reported in the clinical database will be converted based upon the intensity scales defined in the protocol; this assumes a reaction that is too large to measure (non-measurable, "NM") is Grade 3. Note the intensity could be considered "None" (not a reaction) in the analysis despite being considered a reaction by the investigator (eg, swelling measurement > 0 mm but < 25 mm in subjects aged ≥12 years).

Note: The maximum intensity on the ongoing period is derived from the record of the maximum intensity/measurement after the end of the solicited period following the rule described above.

4.3.1.1.2 Maximum Overall Intensity

Maximum overall intensity is derived from the daily intensities computed as described in Section 4.3.1.1.1 and is calculated as the maximum of the daily intensities over the period considered.

4.3.1.1.3 Presence

Presence is derived from the maximum overall intensity over the time period considered:

- None: No presence
- Grade 1, Grade 2, or Grade 3: Presence
- Missing or Unknown: Missing presence

Subjects with at least one non-missing presence for a specific endpoint will be included in the analysis. Conversely, those without a non-missing presence will not be included in the analysis of the endpoint.

4.3.1.1.4 Time of Onset

Time of onset is derived from the daily intensities computed as described in Section 4.3.1.1.1. It corresponds to the first day with intensity of Grade 1, Grade 2, or Grade 3.

Note: If a reaction is not continuous (ie, reaction occurs over two separate periods of time intervened by at least one daily intensity Missing or None) then the time of onset is the first day of the first occurrence.

Note: for solicited systemic reactions, if a reaction is ongoing at the time of the next vaccination, it will be treated differently depending on whether or not it has increased in intensity following the later vaccination.

- If it has not increased in intensity after the next vaccination, it is to be attributed to the earlier vaccination, and is counted as just a single occurrence in safety analysis. The time of onset is the first day of the first occurrence in the earlier vaccination.
- If it has increased in intensity after the next vaccination, it will be counted as 2 separate occurrences after each vaccination in safety analysis (the 2 occurrences will be attributed to the earlier vaccination and later vaccination, respectively). The date of the later vaccination will be considered to be the end date for the first occurrence and the start date of the second occurrence, respectively.

4.3.1.1.5 Number of Days of Occurrence During the Solicited Period

Number of days of occurrence over the period considered is derived from the daily intensities computed as described in Section 4.3.1.1.1. It corresponds to the number of days with daily intensities of Grade 1, Grade 2, or Grade 3. Number of days of occurrence on the solicited period with a specified intensity may also be derived.

4.3.1.1.6 Overall Number of Days of Occurrence

If a reaction is ongoing at the end of the solicited period, then the overall number of days of presence is derived from the daily intensities and the end date of the reaction after the end of the solicited period. The overall number of days of presence is:

- (End date – last vaccination date) + (number of days of presence within the solicited period) − length of the solicited period + 1

If the end date is missing or incomplete (contains missing data), the overall number of days of presence will be considered as "Missing".

4.3.1.1.7 Ongoing

Ongoing is derived from the last daily intensity of the solicited period computed as described in Section 4.3.1.1.1 and the maximum intensity on the ongoing period. The investigator's ongoing flag is not used because the measurement would determine the ongoing status of the reaction.

- Ongoing: if the last daily intensity of the solicited period is at least Grade 1 and the maximum intensity on the ongoing period is at least Grade 1
- Not ongoing: if the last daily intensity of the solicited period is None or the maximum intensity on the ongoing period is None.
- Missing: all other conditions (in this case, it is not included in the denominator of the ongoing analysis in the safety tables).

4.3.1.2 Unsolicited AEs (Including SAEs and AESIs)

4.3.1.2.1 Presence

An observation will be considered an event if it has at least a verbatim term and is not a Grade 0 intensity event.

Grade 0 events should not be included in safety analysis but are included in the separate listing "Unsolicited adverse events not included in the safety analysis."

4.3.1.2.2 Intensity

Intensity for unsolicited AEs will be derived according to the following classification: Grade 0 (None), Grade 1, Grade 2, Grade 3, or Missing

If the unsolicited AE is measurable and its preferred term is part of the list of solicited reactions, then the measurement is derived based upon and following the same rule of the intensity scales defined in the protocol for that measurable injection site or systemic reaction. Note the intensity could be considered as "None" (not a reaction) in the analysis despite being considered a reaction by the investigator (eg, swelling measurement > 0 mm but < 25 mm in subject aged ≥12 years).

Intensity for the other unsolicited AEs will correspond to the value reported in the eCRF.

The maximum intensity corresponds to the highest intensity for a unique term.

4.3.1.2.3 Last Vaccination

Last vaccination before an unsolicited AE is derived from the start date of the unsolicited AE provided in the eCRF and is calculated as follows:

- If an unsolicited AE has a complete start date and different to any of the vaccination dates, the start date is used to determine the last vaccination before the unsolicited AE
- If the start date is missing or partially missing, or equal to any vaccination date, then the visit number in the "Appeared after Visit" or similar field, is used to determine the last vaccination before the unsolicited AE

4.3.1.2.4 Time of Onset

Time of onset is derived from the start date of the unsolicited AE and the date of last vaccination as described in Section 4.3.1.2.3:

- Time of Onset = start date of the unsolicited AE − date of last vaccination before the unsolicited AE

The time of onset is considered as missing only if one or both dates are missing or partially missing.

An unsolicited AE with missing time of onset will be considered to have occurred just after the last vaccination (computed according to the Section 4.3.1.2.3), so will be included in the safety analysis.

Note: Unsolicited AE that occurred before vaccination (negative time of onset) will not be included in analysis but will be listed separately.

4.3.1.2.5 Duration

Duration is derived from the start and end dates of the unsolicited AE:

- Duration = end date of unsolicited AE − start date of unsolicited AE + 1.

The duration is considered as missing only if one or both of the start and end dates of the unsolicited AE is missing or partially missing.

4.3.1.2.6 Serious Adverse Events

An event will be considered as a serious event if "Yes" is checked for "Serious" in the eCRF.

4.3.1.2.7 Adverse Events of Special Interest

An unsolicited event will be considered as an AESI if "Yes' is checked for "Is the event an AESI?" in the eCRF.

4.3.1.3 Other Safety Endpoints

4.3.1.3.1 Pregnancy

This information will not be included in the analysis but will be listed separately. No derivation or imputation will be done.

4.3.1.3.2 Action Taken

This information will be summarized as collected, including missing observations. No derivation or imputation will be done.

4.3.1.3.3 Seriousness

This information will be summarized as collected. No derivation or imputation will be done.

4.3.1.3.4 Outcome

This information will be summarized as collected. No derivation or imputation will be done.

4.3.1.3.5 Causality

This information will be summarized as collected in the field "Relationship to investigational product". Missing causal relationship to study vaccine (investigational product) will be handled as described in Section 5.3.2.2.

Relationship to study procedure for SAE is only presented in the listing.

4.3.1.3.6 Adverse Events Leading to Study Discontinuation

Adverse Events Leading to Study Discontinuation are defined as AEs leading to discontinuation of the study during each active phase (primary series Cohort 1: from Visit 1 up to Visit 5; booster phase Cohort 1: from Visit 6 up to Visit 8; primary series Cohort 2: from Visit 1 up to Visit 4; booster phase Cohort 2: from Visit 8 up to Visit 10). AEs leading to discontinuation of the study during safety follow-up phase are not to be considered for this category.

This information will be summarized as collected. A flag is available in the clinical database for all AEs in order to identify AEs leading to study discontinuation before the end of each active phase.

In general, the items that are counted are:

- Disposition table: A subject who, on the "Completion at End of Study" form question "What was the subjects' status?" has "Adverse Event" checked.
- Safety overview table: A subject who has either on the "Completion at End of Study" form, question "What was the subjects' status?" has "Adverse Event" checked or lists a solicited AE that has "Caused Study Termination" checked that is at least Grade 1 or an unsolicited AE that has "Caused Study Discontinuation" checked that is at least Grade 1 or missing and is within the time period indicated.
- System Organ Class (SOC)/Preferred Term (PT) table: A solicited AE that has "Caused Study Termination" checked that is at least Grade 1 or an unsolicited AE that has "Caused Study Discontinuation" checked that is at least Grade 1 or missing and is within the time period indicated.

4.3.2 Immunogenicity

4.3.2.1 Computed Values for Analysis

In order to appropriately manage duplicate values for analysis purposes, the individual geometric mean (GM) of both values will be computed for each blood sample.

If a value is < Lower Limit of Quantitation (LLOQ) (ie, 0.2 international unit [IU]/mL), then the computed value LLOQ/2 (ie, 0.1 IU/mL) will be used for the GM calculation.

If a value is > Upper Limit of Quantitation (ULOQ) (ie, 900.0 IU/mL), then 900.0 IU/mL will be used for the GM calculation.

For the qualitative Advisory Committee on Immunization Practices (ACIP) criteria reading (stated as Complete or Incomplete twice for each sample), the individual ACIP status will be derived as follows:

- If the 2 values are Complete, then the sample status will be Complete
- If the 2 values are Incomplete, then the sample status will be Incomplete
- If 1 value is Complete and the other one is Incomplete, the status will be Undefined
- If 1 value is Complete and other one is Not Reported, the status will be Complete
- If 1 value is Incomplete and other one is Not Reported, the status will be Incomplete
- If 2 values are Not Reported, the status will be Not Reported

4.3.3 Efficacy

Not applicable.

4.3.4 Derived Other Variables

4.3.4.1 Duration of a Subject in the Study

For Cohorts 1 and 2, the duration of a subject in the primary series (from Visit 1 up to the end of 6-month follow up after the last vaccination in the primary series) is computed as follows: Maximum (Visit dates in primary series, Termination date of active phase in the primary series, Follow-up date of the primary series) − date of V01 + 1.

For Cohort 1, the duration of an adult in the primary series and the booster phase (from Visit 1 up to the end of 6-month follow up after the booster dose) is computed as follows: Maximum (Visit dates, Termination date of active phase in the primary series, Follow-up date of the primary series, Termination date of active phase in the booster phase, Follow-up date of the booster phase) − date of V01 + 1.

For Cohort 2, the duration of an adult in the primary series and the immunogenicity persistence and booster phase (from Visit 1 up to the end of 6-month follow up after the booster dose) is computed as follows: Maximum (Visit dates, Termination date of active phase in the primary series, Follow-up date of the primary series, Termination date of Immunogenicity persistence phase, Termination date of active phase in the booster phase, Follow-up date of the booster phase) − date of V01 + 1.

4.3.4.2 Duration of the Study

- Primary series (all subjects)

The duration of the active phase in the primary series (from Visit 1 up to Visit 5 for Cohort 1; from Visit 1 up to Visit 4 for Cohort 2) is computed as follows: Maximum of all subjects (Visit dates, Termination date of active phase in the primary series) − Minimum of all subjects (date of V01) + 1.

The duration of the follow-up period in the primary series is computed as: Maximum of all subjects (Follow-up date of the primary series) − Minimum of all subjects (Termination date of active phase in the primary series) + 1.

The duration of the primary series is computed as: Maximum of all subjects (Visit dates within the primary series, Termination date of active phase in the primary series, Follow-up date of the primary series) − Minimum of all subjects (date of V01) + 1.

- Booster phase for the adult subsets

The duration after the active phase of the primary series but before booster phase (from Visit 5 up to Visit 6 for Cohort 1; from Visit 4 up to Visit 8 for Cohort 2) is computed as follows:

  - Cohort 1: Maximum of all adults in the subset (date of V06) − Minimum of all adults in the subset (Termination date of active phase in the primary series) + 1
  - Cohort 2: Maximum of all adults in the subset (date of V08) − Minimum of all adults in the subset (Termination date of active phase in the primary series) + 1

The duration of the active phase of the booster phase (from Visit 6 up to Visit 8 for Cohort 1; from Visit 8 up to Visit 10 for Cohort 2) is computed as follows:

- Cohort 1: Maximum of all adults in the subset (Visit dates, Termination date of active phase in the booster phase) − Minimum of all adults in the subset (date of V06) + 1.
- Cohort 2: Maximum of all adults in the subset (Visit dates, Termination date of active phase in the booster phase) − Minimum of all adults in the subset (date of V08) + 1.

The duration of the follow-up of the booster phase is computed as: Maximum of all adults in the subset (Follow-up date of the booster phase) − Minimum of all adults in the subset (Termination date of active phase in the booster phase) + 1

The duration of the booster phase is computed as:

- Cohort 1: Maximum of all adults in the subset (Visit dates within the booster phase, Termination date of active phase in the booster phase, Follow-up date of the booster phase) − Minimum of all adults in the subset (date of V06) + 1.
- Cohort 2: Maximum of all adults in the subset (Visit dates within the booster phase, Termination date of active phase in the booster phase, Follow-up date of the booster phase) − Minimum of all adults in the subset (date of V08) + 1.

- The whole study (primary series and booster phase) for the adult subsets

The duration of the whole study is computed as below.

- Cohort 1: Maximum of all adults in the subset (Visit dates, Termination date of active phase in the booster phase, Follow-up date of the booster phase) − Minimum of all adults in the subset (date of V01) + 1.
- Cohort 2: Maximum of all adults in the subset (Visit dates, Termination date of immunogenicity persistence phase, Termination date of active phase in the booster phase, Follow-up date of the booster phase) − Minimum of all adults in the subset (date of V01) + 1.

4.3.4.3 Age and Age group

The age of a subject in the study is based on the age collected in eCRF. For subjects 12 to 23 months, age in months will be used. For subjects 2 years and above, age in years will be used.

The following age groups will be derived:

Primary series (all subjects)

- Pediatric (< 18 years), including: 12 to 23 months, 2 to 11 years, 12 to 17 years
- Adults (≥18 years), including: 18 to 40 years, 41 to 64 years, ≥ 65 years

Booster phase (adult subsets)

- 18 to 40 years, 41 to 64 years, and ≥ 65 years

The quantitative descriptive statistics (eg, Mean, SD, Max, Min, Median, Q1 and Q3) of age in demographics summary table(s) is based on the age in months or year collected in eCRF as well.

5 Statistical Methods and Determination of Sample Size

The statistical analysis will be performed under the responsibility of the Sponsor's Biostatistics platform with the SAS software, at least version 9.4 (SAS Institute, Cary, North Carolina, USA).

The results of the statistical analysis will be available in the final clinical study report (CSR).

For descriptive purposes, the following statistics will be presented:

Table 5.1: Descriptive statistics produced

Baseline characteristics and follow-up description	**Categorical data**	Number of subjects. Percentage of subjects.
	Continuous data	Mean, standard deviation, median, quartiles, minimum, and maximum.
Clinical safety results	**Categorical data**	Solicited: Number and percentage (95% CIs) of subjects. Unsolicited: Number and percentage (95% CIs) of subjects, and number of events.
Immunogenicity results	**Categorical data (cutoff)**	Number and percentage (95% CIs) of subjects.
	Continuous data (titer / data)	Log10: Mean and standard deviation (SD). Anti-Log10 (work on Log10 distribution, and anti-Log10 applied): Geometric mean, 95% CI of the geometric mean, median, quartiles, minimum, and maximum. Graphical representation by Reverse Cumulative Distribution Curve (RCDC).

The CI for the single proportion will be calculated using the exact binomial method (Clopper-Pearson method), quoted by Newcombe (1), ie, using the inverse of the beta integral with SAS®.

For immunogenicity results, assuming that Log10 transformation of the titers / data follows a normal distribution, at first, the mean and the 95% CI will be calculated on Log10 (titers / data) using the usual calculation for normal distribution (using Student's t distribution with n-1 degree of freedom), then antilog transformations will be applied to the results of calculations, in order to provide geometric means (GMs) and their 95% CI.

GM is defined as follows:

$$GM = \left(\prod_{i=1}^{n} y_i \right)^{1/n} = 10^{\wedge}\left(\frac{1}{n} \sum_{i=1}^{n} \log_{10}(y_i) \right),$$

where $(y_1, y_2, \ldots, y_n)$ are the observed titers or individual ratios for each subject.

5.1 Statistical Methods

5.1.1 Hypotheses and Statistical Methods for Primary Objective

5.1.1.1 Hypotheses

In each age group (ie, pediatric and adult subjects, respectively) of Cohort 1, the immunogenicity of VRVg-2 will be compared to that of Verorab® and Imovax® Rabies vaccines at D42, ie, 14 days after the third vaccine injection, using a non-inferiority (NI) testing.

For each comparison, the primary parameter will be the difference of the proportions of subjects with an RVNA titer ≥ 0.5 IU/mL at D42 between the 2 compared vaccine groups in Cohort 1.

The hypotheses tested will be the following:

Pediatric subjects:

H_{01}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Verorab\ (Group\ 2)} \leq -5\%$

H_{11}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Verorab\ (Group\ 2)} > -5\%$

H_{02}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Imovax\ Rabies\ (Group\ 3)} \leq -5\%$

H_{12}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Imovax\ Rabies\ (Group\ 3)} > -5\%$

Adult subjects:

H_{03}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Verorab\ (Group\ 2)} \leq -5\%$

H_{13}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Verorab\ (Group\ 2)} > -5\%$

H_{04}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Imovax\ Rabies\ (Group\ 3)} \leq -5\%$

H_{14}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Imovax\ Rabies\ (Group\ 3)} > -5\%$

Where P is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D42.

VRVg-2 will be considered as non-inferior to the control vaccine if the hypothesis H_0 is rejected.

5.1.1.2 Statistical Methods

For the NI hypotheses testing, the statistical methodology will be based on the two-sided 95% CI of the difference of proportions of subjects with an RVNA titer ≥ 0.5 IU/mL at D42 between the VRVg-2 group and each of the comparator vaccines for subjects in Cohort 1. The 95% CI for differences will be calculated using Wilson score method without continuity correction (2).

Let the difference of proportions with an RVNA titer ≥ 0.5 IU/mL between the 2 compared groups $\hat{\theta} = p_1 - p_2$, then $L = \hat{\theta} - \delta$ and $U = \hat{\theta} + \varepsilon$ are respectively the lower and the upper limits of the CI, where:

$$\delta = Z_{0.025}\sqrt{\left\{\frac{l_1(1-l_1)}{n_1} + \frac{u_2(1-u_2)}{n_2}\right\}}$$

$$\varepsilon = Z_{0.025}\sqrt{\left\{\frac{l_2(1-l_2)}{n_2}+\frac{u_1(1-u_1)}{n_1}\right\}}$$

l_1 and u_1 are calculated from the CI of the single proportion in Group 1 given by:

$$\frac{(2n_1p_1 + Z^2_{0.025} \pm Z_{0.025}\sqrt{(Z^2_{0.025} + 4n_1p_1(1-p_1))})}{2(n_1 + Z^2_{0.025})}$$

l_2 and u_2 are calculated from the CI of the single proportion in comparator vaccine group (Group 2 or Group 3) given by:

$$\frac{(2n_2p_2 + Z^2_{0.025} \pm Z_{0.025}\sqrt{(Z^2_{0.025} + 4n_2p_2(1-p_2))})}{2(n_2 + Z^2_{0.025})}$$

where $Z_{0.025}$ is the upper 97.5th percentile of the standard normal distribution.

Each null hypothesis will be rejected if the lower limit of the two-sided 95% CI of the difference of the 2 proportions ($P_{VRVg\text{-}2}$-$P_{control}$) is > -5%.

The primary objective of NI will be demonstrated if all null hypotheses are rejected.

5.1.2 Hypotheses and Statistical Methods for Secondary Objectives

5.1.2.1 Hypotheses

- **Secondary immunogenicity objective 1: Superiority in the 3-dose VRVg-2 group at D42**

Only if the primary objective is achieved, then the 1st secondary objective will be assessed for overall subjects from the VRVg-2 group (pooled pediatric and adult subjects) in Cohort 1.

The hypotheses tested will be the following:

H_{01}: $\hat{P}_{VRVg-2\ (Group\ 1)} < 99\%$ or H_{02}: $P_{VRVg\text{-}2\ (Group\ 1)} < 97\%$

H_{11}: $\hat{P}_{VRVg-2\ (Group\ 1)} \geq 99\%$ and H_{12}: $P_{VRVg\text{-}2\ (Group\ 1)} \geq 97\%$

Where $\hat{P}$ is the observed proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D42, and P is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D42.

VRVg-2 will be considered as sufficient at D42 if the hypothesis H_0 is rejected.

- **Secondary immunogenicity objective 2: NI testing of 2-dose VRVg-2 at D28 versus 2-dose Verorab and Imovax Rabies at D28**

Only if the 1st secondary objective is achieved, the immunogenicity of VRVg-2 will be compared to that of Verorab and Imovax Rabies vaccines at D28, ie, 14 days after the second vaccine injection, using NI testing in each age group (Pediatric subjects: Cohort 1; Adult subjects: pooled Cohort 1 and Cohort 2), respectively.

For each comparison, the primary parameter will be the difference in the proportions of subjects with an RVNA titer ≥ 0.5 IU/mL at D28 between the 2 compared vaccine groups.

The hypotheses tested will be the following:

Pediatric subjects:

H_{01}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Verorab\ (Group\ 2)} \leq -5\%$

H_{11}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Verorab\ (Group\ 2)} > -5\%$

H_{02}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Imovax\ Rabies\ (Group\ 3)} \leq -5\%$

H_{12}: $P_{VRVg\text{-}2\ (Group\ 1)} - P_{Imovax\ Rabies\ (Group\ 3)} > -5\%$

Adult subjects:

H_{03}: $P_{VRVg\text{-}2\ (Groups\ 1+4)} - P_{Verorab\ (Groups\ 2+5)} \leq -5\%$

H_{13}: $P_{VRVg\text{-}2\ (Groups\ 1+4)} - P_{Verorab\ (Groups\ 2+5)} > -5\%$

H_{04}: $P_{VRVg\text{-}2\ (Groups\ 1+4)} - P_{Imovax\ Rabies\ (Groups\ 3+6)} \leq -5\%$

H_{14}: $P_{VRVg\text{-}2\ (Groups\ 1+4)} - P_{Imovax\ Rabies\ (Groups\ 3+6)} > -5\%$

Where P is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D28.

The 2-dose VRVg-2 will be considered as non-inferior to the 2-dose control vaccine if the hypothesis H_0 is rejected.

- **Secondary immunogenicity objective 3: NI testing of 2-dose VRVg-2 at D28 versus 3-dose Imovax Rabies at D42**

Only if the 2nd secondary objective is achieved, the immunogenicity of 2-dose VRVg-2 at D28 will be compared to that of 3-dose Imovax Rabies vaccines at D42, using NI testing in each age group (Pediatric subjects: Cohort 1; Adult subjects: pooled Cohort 1 and Cohort 2), respectively.

For each comparison, the primary parameter will be the difference in the proportions of subjects with an RVNA titer ≥ 0.5 IU/mL between the 2 compared vaccine groups.

The following hypotheses will be tested:

Pediatric subjects (Cohort 1 only):

H_0: $P_{VRVg\text{-}2\ at\ D28\ (Group\ 1)} - P_{Imovax\ Rabies\ at\ D42\ (Group\ 3)} \leq -10\%$

H_1: $P_{VRVg\text{-}2\ at\ D28\ (Group\ 1)} - P_{Imovax\ Rabies\ at\ D42\ (Group\ 3)} > -10\%$

Adult subjects (Cohorts 1 and 2):

H_0: $P_{VRVg\text{-}2\ at\ D28\ (Groups\ 1+4)} - P_{Imovax\ Rabies\ at\ D42\ (Group\ 3)} \leq -10\%$

H_1: $P_{VRVg\text{-}2\ at\ D28\ (Groups\ 1+4)} - P_{Imovax\ Rabies\ at\ D42\ (Group\ 3)} > -10\%$

With $P_{VRVg\text{-}2\ at\ D28}$ is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D28 for VRVg-2, and $P_{Imovax\ Rabies\ at\ D42}$ is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D42 for Imovax Rabies.

The 2-dose VRVg-2 will be considered as non-inferior to 3-dose Imovax Rabies if the hypothesis H_0 is rejected.

In addition, the lower limit of the confidence interval will be assessed against -5% margin as a supplemental NI analysis.

- **Secondary immunogenicity objective 4: Superiority in the 2-dose VRVg-2 group at D28**

Only if the 3rd secondary objective is achieved, then for overall subjects in VRVg-2 group (pooled pediatric and adult subjects) from pooled Cohort 1 and Cohort 2, the 4th secondary objective will be assessed.

The hypotheses tested will be the following:

H_{01}: $\hat{P}_{VRVg-2\ (Groups\ 1+4)} < 99\%$ or H_{02}: $P_{\text{VRVg-2 (Groups 1+4)}} < 97\%$

H_{11}: $\hat{P}_{VRVg-2\ (Groups\ 1+4)} \geq 99\%$ and H_{12}: $P_{\text{VRVg-2 (Groups 1+4)}} \geq 97\%$

With $\hat{P}$ is the observed proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D28, and P is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D28.

VRVg-2 will be considered as sufficient at D28 if the hypothesis H_0 is rejected.

- **Secondary immunogenicity objective 5: NI testing of 2-dose Imovax Rabies at D28 versus 3-dose Imovax Rabies at D42**

Only if the 4th secondary objective is achieved, the immunogenicity of 2-dose Imovax Rabies at D28 will be compared to that of 3-dose Imovax Rabies vaccines at D42, using NI testing in overall subjects (pooled pediatric and adult subjects) in Cohort 1 only.

For each comparison, the primary parameter will be the difference in the proportions of subjects with an RVNA titer ≥ 0.5 IU/mL between the 2 compared timepoints.

The following hypotheses will be tested:

H_0: $P_{\text{Imovax Rabies at D28 (Group 3)}} - P_{\text{Imovax Rabies at D42 (Group 3)}} \leq -10\%$

H_1: $P_{\text{Imovax Rabies at D28 (Group 3)}} - P_{\text{Imovax Rabies at D42 (Group 3)}} > -10\%$

With $P_{\text{Imovax Rabies at D28}}$ is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D28 for Imovax Rabies in Cohort 1, and $P_{\text{Imovax Rabies at D42}}$ is the proportion (%) of subjects with an RVNA titer ≥ 0.5 IU/mL at D42 for Imovax Rabies in Cohort 1.

The 2-dose Imovax Rabies will be considered as non-inferior to 3-dose Imovax Rabies if the hypothesis H_0 is rejected.

In addition, the lower limit of the confidence interval will be assessed against -5% margin as a supplemental NI analysis.

5.1.2.2 Statistical Methods

The hypotheses associated with the secondary immunogenicity objectives will be tested only if the primary objective is demonstrated, and then each of the secondary immunogenicity objectives will be evaluated sequentially following a fixed-sequence method as specified below (5) (6) (7).

This method allows to conclude on any of the hypothesis testing conducted at one-sided 0.025 nominal alpha without inflation of overall Type I error, provided the sequence specified hereafter is respected:

- **Secondary immunogenicity objective 1: Superiority in the 3-dose VRVg-2 group at D42**

Only if the primary objective is achieved at D42, the superiority of VRVg-2 group at D42 will be assessed on overall subjects in VRVg-2 group.

The null hypothesis will be rejected and the 1st secondary immunogenicity objective will be achieved if the observed proportion of subjects with an RVNA titer ≥ 0.5 IU/mL at D42 in VRVg-2 group is at least 99.0%, and the lower limit of the 95% CI of the proportion calculated using the exact binomial distribution (Clopper Pearson method) (1) is at least 97%.

- **Secondary immunogenicity objective 2: NI testing of 2-dose VRVg-2 at D28 versus 2-dose Verorab and Imovax Rabies at D28**

Only if the 1st secondary immunogenicity objective is achieved, based on the same statistical approach for the primary objective, the non-inferiority of VRVg-2 compared to each of the comparator vaccines will be tested at D28, in each age group (ie, in pediatric subjects and adult subjects, respectively), with the two-sided 95% CI of the difference of the 2 proportions at D28, calculated using Wilson score method without continuity correction (2).

The null hypothesis will be rejected if the lower limit of the two-sided 95% CI of the difference of the 2 proportions (P_{VRVg-2}-$P_{control}$) is > -5%.

The 2nd secondary immunogenicity objective of NI at D28 will be demonstrated if all null hypotheses are rejected.

- **Secondary immunogenicity objective 3: NI testing of 2-dose VRVg-2 at D28 versus 3-dose Imovax Rabies at D42**

Only if the 2nd secondary immunogenicity objective is achieved, based on the similar statistical approach for the primary objective, the non-inferiority of 2-doseVRVg-2 compared to 3-dose Imovax Rabies will be tested, in each age group (ie, in pediatric subjects and adult subjects, respectively), with the two-sided 95% CI of the difference of the 2 proportions, calculated using Wilson score method without continuity correction (2).

The null hypothesis will be rejected if the lower limit of the two-sided 95% CI of the difference of the 2 proportions ($P_{VRVg-2\ at\ D28}$ -$P_{Imovax\ Rabies\ at\ D42}$) is > -10%.

The 3rd secondary immunogenicity objective will be demonstrated if all null hypotheses are rejected.

In addition, supplemental analysis will be performed based on NI margin of -5%, using the same statistical method.

- **Secondary immunogenicity objective 4: Superiority in the 2-dose VRVg-2 group at D28**

Only if the 3rd secondary immunogenicity objective is achieved, based on the same statistical approach for the 1st secondary immunogenicity objective, the superiority of VRVg-2 group at D28 will be assessed on overall subjects in VRVg-2 group.

The null hypothesis will be rejected and the 4th secondary immunogenicity objective will be achieved if the observed proportion of subjects with an RVNA titer ≥ 0.5 IU/mL at D28 in VRVg-2 group is at least 99%, and the lower limit of the 95% CI of the proportion calculated using the exact binomial distribution (Clopper Pearson method) (1) is at least 97%.

- **Secondary immunogenicity objective 5: NI testing of 2-dose Imovax Rabies at D28 versus 3-dose Imovax Rabies at D42**

Only if the 4th secondary objective is achieved, the 5th secondary immunogenicity objective will be assessed on overall subjects in Imovax Rabies group in Cohort 1 only, using the statistical methods mentioned below, based on NI margin of -10%.

In addition, supplemental analysis will be performed based on NI margin of -5%, using the same statistical method.

Generalized Linear Model

The null hypothesis will be rejected and 5th secondary immunogenicity objective will be achieved if the lower limit of the two-sided 95% CI of the difference of the 2 proportions ($P_{\text{Imovax Rabies at D28}}$ -$P_{\text{Imovax Rabies at D42}}$) is > -10%, which will be calculated using a generalized linear model (GLM) for repeated measured data with categorical response under binomial distribution (link function=identity) (8).

All subjects in Imovax Rabies group in Cohort 1 (Group 3) with at least one available immunogenicity assessment at D28 or D42 will be included in the GLM. An example of SAS code is presented as below.

```
proc genmod data=immuno;
    class ID time_point;
    model response(event="1") = timepoint / dist=binomial link=identity;
    repeated subject=ID;
    lsmeans time_point / diff cl;
run;
```

where event="1" refers to an occurrence of RVNA titer ≥ 0.5 IU/mL at D28 or D42 in Imovax Rabies Group in Cohort 1 (Group 3), timepoints is the time point for repeated immunogenicity measures including D28 and D42, and ID is the unique subject identifier for each subject.

MOVER Wilson score method

In case GLM did not converge (e.g, if the seroconversion rate is 0% or 100% at D28 or D42 in Group 3 in Cohort 1), MOVER Wilson score method for paired binomial proportion (9) will be used to calculate the two-sided 95% CI of the difference between D28 and D42.

Only subjects with available immunogenicity data at both D28 and D42 will be included in the analysis using Mover Wilson score method as below.

The observed subjects with available immunogenicity data at both D28 and D42 can be summarized in a 2 × 2 contingency table, as shown in Table 5.2.

Table 5.2: The observed subjects with available RVNA titer assessments at both D28 and D42

Time points		D42		
		RVNA titer ≥ 0.5 IU/mL	**RVNA titer < 0.5 IU/mL**	**Total**
D28	**RVNA titer ≥ 0.5 IU/mL**	n_{11}	n_{12}	n_{1+}
	RVNA titer < 0.5 IU/mL	n_{21}	n_{22}	n_{2+}
	Total	n_{+1}	n_{+2}	N

where n_{11} corresponds to the number of subjects with an RVNA titer ≥ 0.5 IU/mL at both D28 and D42. n_{12} is the number of subjects with an RVNA titer ≥ 0.5 IU/mL at D28 and an RVNA titer < 0.5 IU/mL at D42. n_{21} is the number of subjects with an RVNA titer < 0.5 IU/mL at D28 and an RVNA titer ≥ 0.5 IU/mL at D42. n_{22} is the number of subjects with an RVNA titer < 0.5 IU/mL at both D28 and D42. n_{1+} and n_{+1} are the numbers of subjects with an RVNA titer ≥ 0.5 IU/mL at D28 and D42, respectively. n_{2+} and n_{+2} are the numbers of subjects with an RVNA titer < 0.5 IU/mL at D28 and D42, respectively. N is the total number of subjects with available immunogenicity data at both D28 and D42.

For paired data, let the difference of proportions of subjects with an RVNA titer ≥ 0.5 IU/mL between the 2 compared timepoints $\hat{\theta} = p_1 - p_2$, then $L^* = \hat{\theta} - v$ and $U^* = \hat{\theta} + \lambda$ are the lower and the upper limits of the CI, respectively, where:

$$v = \sqrt{(p_1 - l_1)^2 + (u_2 - p_2)^2 - 2\hat{\psi}(p_1 - l_1)(u_2 - p_2)}$$

$$\lambda = \sqrt{(u_1 - p_1)^2 + (p_2 - l_2)^2 - 2\hat{\psi}(u_1 - p_1)(p_2 - l_2)}$$

l_1 and u_1 are calculated from the CI of the single proportion of subjects with an RVNA titer ≥ 0.5 IU/mL at D28 (ie, $P_{\text{Imovax Rabies at D28}}$) given by:

$$(l_1, u_1) = \frac{2n_{1+} + Z_{0.025}^2 \pm Z_{0.025}\sqrt{Z_{0.025}^2 + 4n_{1+}(1 - \frac{n_{1+}}{N})}}{2(N + Z_{0.025}^2)}$$

l_2 and u_2 are calculated from the CI of the single proportion of subjects with an RVNA titer ≥ 0.5 IU/mL at D42 (ie, $P_{\text{Imovax Rabies at D42}}$) given by:

$$(l_2, u_2) = \frac{2n_{+1} + Z_{0.025}^2 \pm Z_{0.025}\sqrt{Z_{0.025}^2 + 4n_{+1}(1 - \frac{n_{+1}}{N})}}{2(N + Z_{0.025}^2)}$$

where $Z_{0.025}$ is the upper 97.5th percentile of the standard normal distribution.

$\hat{\psi}$ is an estimate of the correlation coefficient between $\hat{p}_1$ and $\hat{p}_2$. $\hat{\psi}$ will be 0 if any of the marginal sums $(n_{1+}, n_{2+}, n_{+1}, n_{+2})$ is 0. Otherwise, let $A = n_{11}n_{22} - n_{12}n_{21}$ and compute $\hat{\psi}$ as

$$\hat{\psi} = \begin{cases} (A - N/2)/\sqrt{n_{1+}n_{2+}n_{+1}n_{+2}} & \text{if } A > N/2, \\ 0 & \text{if } 0 \leq A \leq N/2, \\ A/\sqrt{n_{1+}n_{2+}n_{+1}n_{+2}} & \text{if } A < 0. \end{cases}$$

- **Other secondary immunogenicity and safety objectives**

All other secondary endpoints will be described by age group and vaccine group using descriptive statistical methods without hypothesis testing, as follows.

Immunogenicity endpoints

A descriptive analysis of RVNA titers (IU/mL) will be performed by vaccine group and age group for the following statistics at each timepoint (D0 and D28 [for subjects in Cohort 1 and Cohort 2], D42 [for subjects in Cohort 1]; M12 and M12+D14 [for subjects in Booster Phase of Cohort 1]; M6, M12, M18, M24 up to M36, and M24 up to M36+D14 [for subjects in Immunogenicity Persistence and Booster Phase of Cohort 2]):

- Number of subjects with RVNA titer available
- Number and percentage of subjects with RVNA titer ≥ 0.5 IU/mL, and 95% CI of the proportion
- Number and percentage of subjects with RVNA titer ≥ 0.2 (LLOQ) IU/mL, and 95% CI of the proportion
- Number and percentage of subjects with complete or incomplete results at the starting dilution (1/5) of the RFFIT assay, and 95% CI of the proportion
  - Among the analysis population
  - Among the subset of subjects with a determined result (identical duplicate values, either Complete or Incomplete)
- GMTs and 95% CI of RVNA titers
- The GM of individual titer ratio (GMTR) between the post-vaccination time points and the baseline (D0) within each group, and 95% CI:
  - D28/D0
  - D42/D0
  - M6/D0
  - M12/D0
  - (M12+D14)/D0
  - M18/D0

- (M24 up to M36)/D0
- (M24 up to M36+D14)/D0

- The GMTR between the post-booster vaccination time point and the corresponding pre-booster vaccination time point within each group, and 95% CI:
  - (M12+D14)/M12
  - (M24 up to M36+D14)/ M24 up to M36
- Distribution of titers (minimum, Q1, median, Q3, maximum): based on Log10 transformation first and then antilog transformation.
- Log10 (Titer): mean and SD
- RCDC of RVNA titers

Safety endpoint

The analysis will be descriptive using the Safety Analysis Set (SafAS) in the primary series and the Safety Analysis Set for booster (SafASB), for each vaccination group. The statistics presented on Table 5.1 will be produced.

The safety analysis will report the occurrence of solicited reactions and the incidence of unsolicited AEs, including SAEs and AESIs, over the safety observation period by age group and vaccine group according to the vaccine received.

- Immediate unsolicited systemic AEs reported in the 30 minutes after each vaccination
- Solicited injection site reactions occurring within 7 days after each vaccination
- Solicited systemic reactions occurring within 7 days after each vaccination
- Unsolicited injection site AEs occurring within 28 days after each vaccination
- Unsolicited systemic AEs occurring between each vaccination and up to 28 days after the last vaccination
- SAEs and AESIs throughout the study (until 6 months after last vaccination for the primary vaccination series and throughout the entire study for the subset of subjects included in the booster phase)

In order to avoid any under-estimation of the incidences, the number of subjects with documented safety will be used as denominator of the frequencies.

- For solicited reactions, the denominator is the total number of subjects who have non-missing data for the particular category of reaction during the time period concerned.

  Note: for solicited systemic reactions, if a reaction is ongoing at the time of the next vaccination, it will be treated differently depending on whether or not it has increased in intensity following the later vaccination.

  - If it has not increased in intensity after the next vaccination, it is to be attributed to the earlier vaccination, and is counted as just a single occurrence in safety analysis. The time of onset is the first day of the first occurrence in the earlier vaccination.

- If it has increased in intensity after the next vaccination, it will be counted as 2 separate occurrences after each vaccination in safety analysis (the 2 occurrences will be attributed to the earlier vaccination and later vaccination, respectively). The date of the later vaccination will be considered as the end date for the first occurrence and the start date of the second occurrence, respectively.

- For unsolicited events, the denominator is the total number of subjects who were vaccinated at the dose analyzed (for the analyses after each dose) or the total number of subjects who were vaccinated at least one dose (for the analyses after any dose).

For safety parameters, 95% Cis of point estimates of proportion will be calculated using the exact binomial distribution (Clopper-Pearson method) for proportions (1).

30-minutes Post-Vaccination Observation Period

Unsolicited systemic AEs occurring within 30 minutes after each vaccination will be presented in summary safety tables, analyzed according to their causality (relationship to the investigational product) and Grade 3 intensity.

Solicited Reactions

The solicited injection site reactions and the solicited systemic reactions will be presented according to the term listed in the eCRF, separately.

The solicited injection site and systemic reactions will be analyzed within 7 days after each vaccination.

Each type of solicited reactions will be presented after each injection according to:

- *Maximum intensity during the solicited period:*

- Grade 1
- Grade 2
- Grade 3

- *Time of onset categories are in* Table 5.3

Table 5.3: Time of onset categories for solicited reactions

	Injection Site Reactions	Systemic Reactions
Post each dose in the primary series, booster dose	D0-D3 D4-D7	D0-D3 D4-D7

- *Range of number of days of occurrence categories are in* Table 5.4

Table 5.4: Range of number of days of occurrence categories for solicited reactions during the solicited period

	Injection Site Reactions	Systemic Reactions
Post each dose in the primary series, booster dose	1 - 3 days 4 - 7 days 8 days	1 - 3 days 4 - 7 days 8 days

Note: Cohort 1 planned to receive 3 doses and Cohort 2 planned to receive 2 doses, respectively, in the primary series.

- *Range of overall number of days of occurrence categories are in* Table 5.5

Table 5.5: Range of overall number of days of occurrence categories for solicited reactions

	Injection Site Reactions	Systemic Reactions
Post each dose in the primary series, booster dose	1 - 3 days* 4 - 7 days 8 days or more Missing end date	1 - 3 days* 4 - 7 days 8 days or more Missing end date

* For solicited reactions still ongoing at D08 after each dose, the range of overall number of days of occurrence category will be 2-3 days, 4-7 day, 8 days or more, and missing end date.

Note: Cohort 1 planned to receive 3 doses and Cohort 2 planned to receive 2 doses, respectively, in the primary series.

- *Action Taken:*

- Missing
- None
- Medication
- Health care provider contact
- Hospitalization
- Discontinuation of study vaccination

Solicited reaction after any injection in the primary series

"After any injection" is for primary series only, means after all scheduled vaccine injections of the primary vaccination series, and at least after the first vaccine injection (D0).

The solicited reaction will be presented after any injections in the primary series and presented after booster vaccination according to:

- Occurrence during the solicited period for solicited reactions, and overall occurrence for solicited reactions still ongoing at D08

- Time of onset period

- Intensity grade: the maximum intensity grade will be computed during the solicited period for all solicited reactions

Unsolicited Adverse Events

The unsolicited systemic AEs will be analyzed per cohort:

- Cohort 1
  - "Between the first and the second vaccination", which corresponds to AEs reported at V02 and occurring after the first dose *
  - "Between the second and the third vaccination", which corresponds to AEs reported at V03 and occurring after the second dose †
  - "Up to 28 days after the third vaccination", which corresponds to AEs reported at V05 and occurring after the third vaccination
  - "Up to 28 days after any vaccine injection in the primary series", which corresponds to AEs belonging to any of the above three categories
  - "Up to 28 days after booster dose", which corresponds to AEs reported at V07 or V08 and occurring after booster dose, for an adult subset
- Cohort 2
  - "Between the first and the second vaccination", which corresponds to AEs reported at V02 and occurring after the first dose *
  - "Up to 28 days after the second vaccination", which corresponds to AEs reported at V03 or V04 and occurring after the second vaccination
  - "Up to 28 days after any vaccine injection in the primary series", which corresponds to AEs belonging to any of the above two categories
  - "Up to 28 days after booster dose", which corresponds to AEs reported at V09 or V10 and occurring after booster dose, for an adult subset

The unsolicited injection site reactions will be analyzed within 28 days (from D0 to D28) after each vaccination, and within 28 days after any vaccination. An unsolicited injection site reaction

* For subjects interrupting their vaccination schedule after the first injection, unsolicited AEs until 28 days after the first dose will be collected

† For subjects interrupting their vaccination schedule after the second injection, unsolicited AEs until 28 days after the second dose will be collected

(**excluding SAE and AESI**) reported with an onset > 28 days after a vaccination will not be included in the safety analysis, but will be listed separately.

The unsolicited systemic AEs will be analyzed between each vaccination if vaccinations are separated less than 28 days, or up to 28 days after each vaccination, if the next vaccination is interrupted, and up to 28 days after any vaccination. An unsolicited systemic AE (excluding SAE and AESI) reported with an onset > 28 days after vaccination will not be included in the safety analysis, but will be listed separately.

Note: An unsolicited AE with missing time of onset will be considered to have **occurred within or up to 28 days after the last vaccination** (computed according to Section 4.3.1.2.3), so will be included in safety analysis.

The unsolicited AEs will be summarized in the safety overview and analyzed according to their nature (SOC and PT of the MedDRA classification), causality (relationship to the vaccine injection as assessed by the investigator), maximum intensity, time of onset and duration.

The occurrence of any unsolicited AE will be presented after any and each injection according to:

- *Maximum intensity:*

- Missing
- Grade 1
- Grade 2
- Grade 3

- *Time of onset categories*

- Missing
- D0-D3
- D4-D7
- D8-D14
- >=D15

- *Maximum duration:*

- Missing
- 1 - 3 days
- 4 - 7 days
- 8 - 14 days
- 15 days or more

SAEs and AESIs

The SAEs and AESIs, which are included in the unsolicited AEs, occurred during the study will be analyzed on the following periods for each cohort:

- Primary series
    - During the active phase of the primary series: from date of first vaccination (V01) to 28 days after the date of last exposure to study vaccine in the primary series (date of last exposure in the primary series + 28 days)
    - During the 6-month follow-up period of the primary series: from 29 days after the last exposure to study vaccine in the primary series (date of last exposure in the primary series + 29 days) to end of 6-month follow-up in the primary series (last visit date or contact date collected in eCRF in the primary series)
    - During the primary series: from date of first vaccination (V01) to end of 6-month follow-up in the primary series (last visit date or contact date collected in eCRF in the primary series)
- Between the end of 6-month follow-up of the primary series and before booster dose (V06 for Groups 1 to 3 [Cohort 1] or V08 for Groups 4 to 6 [Cohort 2]) of the adult subset (Note: for Cohort 2, only fatal or related SAEs are collected and AESI will not be collected during this period)
- Booster phase of the adult subset
    - During the active phase of the booster phase: from date of booster dose (V06 for Groups 1 to 3 [Cohort 1] or V08 for Groups 4 to 6 [Cohort 2]) to 28 days after the booster dose (V08 for Groups 1 to 3 [Cohort 1] or V10 for Groups 4 to 6 [Cohort 2])
    - During the 6-month follow-up period of the booster phase: from 29 days after the booster dose (V08 for Groups 1 to 3 [Cohort 1] or V10 for Groups 4 to 6 [Cohort 2]) to end of 6-month follow-up post booster dose (last visit date or contact date for booster phase collected in eCRF)
    - During the booster phase: from date of booster dose (V06 for Groups 1 to 3 [Cohort 1] or V08 for Groups 4 to 6 [Cohort 2]) to end of 6-month follow-up post the booster dose
    - During the whole study period: from date of first vaccination (V01) to end of 6-month follow-up in the booster phase (last visit date or contact date for booster phase collected in eCRF)

SAEs will be analyzed according to their nature (SOC and PT of the MedDRA classification), causality (relationship to the study vaccine), seriousness criterion and outcome.

AESIs will be analyzed according to their nature (SOC and PT of the MedDRA classification) and causality.

5.1.3 Exploratory Analyses

5.1.3.1 Subgroups Analysis: Impact of Demographic Factors and Center

With the aim to provide the same standards and granularity in terms of investigational results across VRVg studies, and comply with health authorities requirements (3), the possible influence of following factors on the immunogenicity and safety results will be analyzed using descriptive statistics. Thus, the main immunogenicity and safety parameters will be described according to gender, age group, ethnicity (only if more than 5% of subjects had different ethnicity), race (only if more than 5% of subjects had different race), and center.

Race and ethnicity will be defined according to current guidelines (3).

Age groups are the following ones:

- Pediatric (<18 years): including 12 to 23 months, 2 to 11 years, and 12 to 17 years;
- Adult (≥18 years): including 18 to 40 years, 41 to 64 years, and ≥65 years.

These exploratory analyses will be primarily performed on the FASI, FASI for Immunogenicity Persistence and FASI for Booster (for immunogenicity), or the SafAS and SafASB (for safety).

In case of any safety signal detected in the VRVg-2 vaccines, a statistical comparison will be performed between VRVg-2 group and each of the control groups (Verorab® and Imovax® Rabies) with a Fisher exact test in order to check for statistical significance (alpha=0.05, two-sided).

5.1.3.2 Impact of the COVID-19 pandemic

To evaluate the possible impact of the COVID-19 pandemic on the study conduction, main immunogenicity and safety results, the following sensitive analyses will be performed but not limited to:

- Impact of COVID-19 pandemic on subject's disposition, based on enrolled subjects

Note: Subjects impacted by COVID-19 refers to subjects with at least one major/critical protocol deviation due to COVID-19, or who did not complete the study due to COVID-19, or who reported "VISIT NOT DONE" on safety follow up in pandemic form.

- Major or critical deviations due to COVID-19 by randomized group, based on randomized subjects.
- Immunogenicity analysis primarily based on FASI, FASI for Immunogenicity Persistency and FASI for Booster
    - Impact of COVID-19 pandemic on immunogenicity analysis.
    - Impact of medical history of suspected/confirmed COVID-19 pandemic on immunogenicity analysis.
    - Impact of the use of concomitant medications that are used during the study/or were used within 2 months before enrollment for treating/preventing COVID-19 (ie, Hydroxychloroquine and chloroquine) on immunogenicity.

- If any subjects received at least one dose of vaccination but took any concomitant medications for treating/preventing COVID-19 (ie, Hydroxychloroquine and chloroquine) within 2 months before enrollment, then the following immunogenicity analysis will be conducted based on PPAS
  - Immunogenicity of subjects who did not take any concomitant medications for treating/preventing COVID-19 (ie, Hydroxychloroquine and chloroquine) within 2 months before enrollment.
- Safety analysis based on SafAS and SafASB
  - Impact of COVID-19 pandemic on safety overview up to 28 days after any vaccination.
  - Impact of medical history of suspected/confirmed COVID-19 pandemic on safety overview up to 28 days after any vaccination.
  - Impact of the use of concomitant medications that are used during the study/or were used within 2 months before enrollment for treating/preventing COVID-19 (ie, Hydroxychloroquine and chloroquine) on safety overview up to 28 days after any vaccination.

Note: if only few subjects were impacted by COVID-19 (<5% in randomized subjects in the primary series or in the booster phase), only the corresponding listings will be provided instead of tables. Exception is the immunogenicity evaluation for the subjects who did not take any concomitant medications for treating/preventing COVID-19 (ie, Hydroxychloroquine and chloroquine) within 2 months before enrollment base on PPAS.

5.1.3.3 Supplementary Analysis: Impact of Cohort and Age Group

To assess the possible impact of Cohort and age group on the key secondary immunogenicity objectives, the supplementary analysis will be performed by Cohort (Cohort 1, Cohort 2) for adult subjects and by age group (pediatric, adult) for subjects in Cohort 1, respectively, with descriptive statistics only using the same methodology defined in Section 5.1.2.2 to calculate the 95% CIs for single proportions and 95% CIs for the difference between 2 proportions, but without hypothesis testing.

5.2 Analysis Sets

5.2.1 Full Analysis Set

The Full Analysis Set (FAS) is defined as the subset of randomized subjects who received at least 1 dose of the study vaccine in the primary series.

The Full Analysis Set for Immunogenicity (FASI) is defined as a subset of the FAS, including all subjects from FAS who have a baseline RVNA titer lower than 0.5 IU/mL.

The FAS for Immunogenicity Persistence (FASP) is defined as the subset of randomized subjects from Cohort 2 who received a complete primary rabies vaccination series (ie, one week 2-dose

PrEP regimen), and have at least one valid post-vaccination serology result at M6, M12, M18 or M24 up to M36.

The FASI for Immunogenicity Persistence (FASIP) is defined as the subset of FASP, including all subjects from the FASP who have a baseline titer lower than 0.5 IU/mL.

The FAS for Booster at M12 (FASB1) is defined as the subset of subjects who received the booster dose at M12, after a complete primary rabies vaccination series (ie, 3-dose PrEP regimen) in Cohort 1.

The FAS for Booster at M24 up to M36 (FASB2) is defined as the subset of subjects who received the booster dose between M24 up to M36, after a complete primary rabies vaccination series (ie, one week 2-dose PrEP regimen) in Cohort 2.

The FASI for Booster at M12 (FASIB1) is defined as the subset of FASB1, including all subjects from FASB1 with a baseline titer lower than 0.5 IU/mL.

The FASI for Booster at M24 up to M36 (FASIB2) is defined as the subset of FASB2, including all subjects from FASB2 with a baseline titer lower than 0.5 IU/mL.

The analysis of immunogenicity addresses endpoints involving pre- and post-injection titers. The analysis will include all available data for each time point.

5.2.2 Per-Protocol Analysis Set

Two Per-protocol Analysis Sets (PPASs) are defined for key immunogenicity objectives:

- PPAS for D42
- PPAS for D28

Two additional PPASs are defined for descriptive analyses of booster dose:

- PPAS for Booster at M12
- PPAS for Booster at M24 up to M36

Adherence to the definition of the PPAS may also be decided during the blinded data review, ie, before breaking the code and locking the data base.

5.2.2.1 Per-Protocol Analysis Set for D42

The PPAS for D42 is a subset of the FAS which will be used for primary objective assessment at D42 (V04) for subjects in Cohort 1. The subjects presenting with at least one of the following relevant protocol deviations before D42 (ie, 14 days after the third vaccine injection) will be excluded from the PPAS for D42:

- Subject did not meet all protocol-specified inclusion criteria or met at least one of the protocol-specified exclusion criteria
- Subject did not complete the 3-dose vaccination schedule
- Subject received a vaccine other than the one that he / she was randomized to receive
- Preparation and / or administration of vaccine was not done as per-protocol

- Subject did not receive vaccine in the proper time window
  - Vaccination dose 2 in [D07-D09]
  - Vaccination dose 3 in [D25-D31]
- Subject did not provide post-dose 3 serology sample at V04 in the proper time window [D11, D17] post the third vaccination (ie, Vac3 +14D ±3D), or a post-dose 3 serology sample was not drawn at V04
- Subject received a protocol-prohibited therapy/medication/vaccine (belonging to categories 2 and 3) before V04 (D42)
- Subject's serology sample is missing or did not produce valid test results at D0 or D42
- Seropositive subject at D0, ie, RVNA titer ≥ LLOQ
- Subject developed a protocol-specified withdrawal criterion during the study but was not withdrawn before V04 (D42)

5.2.2.2 Per-Protocol Analysis Set for D28

The PPAS for D28 is also subset of the FAS, which will be primarily used for secondary non-inferiority objective evaluation at D28 (V03) for subjects in Primary Series Cohort 1 and Cohort 2. The subjects presenting with at least one of the following relevant protocol deviations before D28 (ie, 21 days after the second vaccine injection) will be excluded from the PPAS for D28:

- Subject did not meet all protocol-specified inclusion criteria or met at least one of the protocol-specified exclusion criteria
- Subject did not complete the 2-dose vaccination schedule at D0 (V01) and D7 (V02)
- Subject received a vaccine other than the one that he / she was randomized to receive at the first 2 doses
- Preparation and / or administration of vaccine was not done as per-protocol at D0 (V01) and D7 (V02)
- Subject did not receive vaccine in the proper time window
  - Vaccination dose 2 in [D07-D09]
- Subject did not provide post-dose 2 serology sample at V03 in the proper time window [D25-D31] or a post-dose 2 serology sample was not drawn at V03
- Subject received a protocol-prohibited therapy/medication/vaccine (belonging to categories 2 and 3) before V03 (D28)
- Subject's serology sample is missing or did not produce valid test results at D0 or D28
- Seropositive subject at D0, ie, RVNA titer ≥ LLOQ
- Subject developed a protocol-specified withdrawal criterion during the study but was not withdrawn before V03 (D28)

5.2.2.3 Per-Protocol Analysis Set for Booster at M12

The PPAS for booster at M12 (V07) is a subset of the FASB1, which will be used for secondary objective evaluation in the booster phase of Cohort 1, for a subset of adult who received a 3-dose regimen and a single booster dose of VRVg-2 one year after the first vaccination of the primary series (ie, at M12), regardless of the type of vaccination received in the primary series. The adult subjects in Booster Phase of Cohort 1, presenting with at least one of the following relevant protocol deviations before M12+D14 (ie, 14 days after the booster dose injection) will be excluded from the PPAS for booster at M12:

- Subject did not meet all protocol-specified inclusion criteria or met at least one of the protocol-specified exclusion criteria
- Subject received a vaccine other than the one that he / she was randomized to receive in either primary series or booster phase
- Preparation and / or administration of vaccine was not done as per-protocol in either primary series or booster phase
- Subject did not receive booster vaccine in the proper time window at V06
  - Vaccination 4 in [M12, M12 +14D] after Vaccination dose 1
- Subject did not provide post-booster dose serology sample at V07 in the proper time window [D14, D15] post the booster vaccination (VAC 4) or a post-booster dose serology sample was not drawn at V07
- Subject received a protocol-prohibited therapy/medication/vaccine (belonging to categories 2 and 3) before V07 (M12 + D14)
- Subject's serology sample is missing or did not produce valid test results at V06 (before booster vaccination) or V07 (D14 after booster vaccination)
- Seropositive subject at D0 in the primary series, ie, RVNA titer ≥ LLOQ
- Subject developed a protocol-specified withdrawal criterion during the study but was not withdrawn before V07 (M12 + D14)

5.2.2.4 Per-Protocol Analysis Set for Booster at M24 up to M36

The PPAS for booster at M24 up to M36 (V09) is a subset of the FASB2, which will be used for secondary objective evaluation in the booster phase of Cohort 2, for a subset of adult who received a one week 2 doses regimen in the primary series and a single booster dose of VRVg-2 between M24 and up to M36 after the first vaccination of the primary series, regardless of the type of vaccination received in the primary series. The adult subjects in Booster Phase of Cohort 2 presenting with at least one of the following relevant protocol deviations before M24 up to M36+D14 (ie, 14 days after the booster dose injection) will be excluded from the PPAS for booster at M24 up to M36:

- Subject did not meet all protocol-specified inclusion criteria or met at least one of the protocol-specified exclusion criteria

- Subject received a vaccine other than the one that he/ she was randomized to receive in either primary series or booster phase
- Preparation and/ or administration of vaccine was not done as per-protocol in either primary series or booster phase
- Subject did not receive booster vaccine in the proper time window at V08
  - Vaccination 3 in [M24 up to M36, M24 up to M36 +14D] after Vaccination dose 1
- Subject did not provide post-booster dose serology sample at V09 in the proper time window [D14, D15] post the booster vaccination (VAC 3), or a post-booster dose serology sample was not drawn at V09 (D14 after booster vaccination)
- Subject received a protocol-prohibited therapy/medication/vaccine (belonging to categories 2 and 3) before V09 (D14 after booster vaccination)
- Subject's serology sample is missing or did not produce valid test results at V08 (before booster vaccination) or V09 (D14 after booster vaccination)
- Seropositive subject at D0 in the primary series, ie, RVNA titer ≥ LLOQ
- Subject developed a protocol-specified withdrawal criterion during the study but was not withdrawn before V09 (D14 after booster vaccination)

5.2.3 Safety Analysis Set

In the primary series, the Safety Analysis Set (SafAS) is defined for each dose as the subset of subjects having received this dose. All subjects will have their safety analyzed after each dose according to the study vaccine they actually received in each cohort and after any dose according to the study vaccines received at the first dose in each cohort.

Safety data recorded for a vaccine received out of the protocol design will be excluded from the analysis (and listed separately). Thus, if a subject does not receive any study vaccine at a given visit or if the study vaccine received does not correspond to any protocol group at a specific dose, the subject is excluded from the SafAS at this dose in the primary series; however, the subject will be included in the analysis for all doses combined (referred to as analysis "after any dose") in the primary series according to the first dose received that corresponds to a protocol group.

The Safety Analysis Set for Booster (SafASB) is defined for the subset of subjects who received the booster dose (VRVg-2 vaccine). The corresponding vaccination group will be determined by their actual vaccination group in the primary series in each Cohort (Groups 1 to 6).

5.2.4 Other Analysis Set(s)

Enrolled subjects

Enrolled subjects are subjects for whom an eCRF has been created.

Randomized subjects

A randomized subject is a subject for whom a vaccine group has been allocated.

Randomized subjects for Booster

A pre-identified adult subset for whom a booster dose (VRVg-2) has been allocated at the time of randomization in each Cohort.

5.2.5 Populations Used in Analyses

Due to randomization error of IRT in Cohort 2 noticed once the randomizations were completed on 02 October 2022, there were an excess of 101 adults accidently randomized into the subset of Immunogenicity Persistence and Booster Phase. Those 101 adults in Cohort 2 were assigned only to participate in one week 2-dose primary series; they will not have immunogenicity persistency assessments and booster vaccination. The 101 adults will be excluded from the subset of Immunogenicity Persistence and Booster Phase Cohort 2 in all statistical analysis.

For primary and secondary objectives, the following population will be used in analyses.

The immunogenicity data for subjects at D0, D28 and D42 will be analyzed on the per-protocol analysis set (PPAS) for D42 and the PPAS for D28 primarily, and also on Full Analysis Set for Immunogenicity (FASI) and the Full Analysis Set (FAS) as supplementary analysis, if applicable (i.e, the difference between number of subjects in the PPAS and number of subjects in the FAS is not less than 10%).

The immunogenicity persistence data for adult subset subjects at M6, M12, M18, and M24 up to M36 (Immunogenicity Persistence and Booster Phase of Cohort 2) will be analyzed on the FASI for Immunogenicity Persistence (FASIP) and FAS for Immunogenicity Persistence (FASP), and also on the PPAS for booster at M24 up to M36.

The immunogenicity data for the booster subset subjects at M12 and M12+D14 (Cohort 1) will be analyzed on the FAS for Booster at M12 (FASB1), and also on the FASI for Booster at M12 (FASIB1) and the PPAS for booster at M12, by overall and according to the vaccine randomized in the primary series.

The immunogenicity data for the booster subset subjects at M24 up to M36 and 14 days after the booster (Cohort 2) will be analyzed on the FAS for Booster at M24 up to M36 (FASB2), and also on the FASI for Booster at M24 up to M36 (FASIB2) and the PPAS for booster at M24 up to M36, by overall and according to the vaccine randomized in the primary series.

The safety analysis will be based on the Safety Analysis Set (SafAS) in the primary series and the Safety Analysis Set for booster (SafASB), according to the actual vaccine received in the primary series.

Table 5.6 presents the detail populations used in the statistical analysis.

Table 5.6: Populations used in the analyses

		Analysis sets	Analysis by	Analysis group
Primary Objective	**D42 NI testing**	PPAS for D42 (main analysis) FASI, FAS	Randomized vaccine group in the primary series	Cohort 1: • Groups 1, 2, 3
Key Secondary Objectives	**Objective 1: D42 Superiority testing in VRVg-2**	PPAS for D42 (main analysis) FASI, FAS	Randomized vaccine group in the primary series	Cohort 1: • Group 1
	Objective 2: 2-dose VRVg-2 at D28 versus 2-dose Verorab and Imovax Rabies at D28	PPAS for D28 (main analysis) FASI, FAS	Randomized vaccine group in the primary series	Pooled Cohort 1 and Cohort 2: • Pooled Group 1 + Group 4 • Pooled Group 2 + Group 5 • Pooled Group 3 + Group 6
	Objective 3: 2-dose VRVg-2 at D28 versus 3-dose Imovax Rabies at D42	2-dose VRVg-2: PPAS for D28 (main analysis) 3-dose Verorab/Imovax Rabie: PPAS for D42 (main analysis) FASI, FAS	Randomized vaccine group in the primary series	For Pediatrics, Cohort 1: • Group 1, Group 3 For Adults, pooled Cohort 1 and Cohort 2: • Pooled Group 1+4, Group 3
	Objective 4: D28 Superiority testing in VRVg-2	PPAS for D28 (main analysis) FASI, FAS	Randomized vaccine group in the primary series	Pooled Cohort 1 and Cohort 2: • Pooled Group 1 + Group 4
	Objective 5: 2-dose Imovax Rabies at D28 versus 3-dose Imovax Rabies at D42	2-dose Imovax Rabies: PPAS for D28 (main analysis) 3-dose Imovax Rabies: PPAS for D42 (main analysis) FASI, FAS	Randomized vaccine group in the primary series	Cohort 1: • Group 3
Other Secondary Objectives	**Immunogenicity description for primary series**	PPAS for D42, PPAS for D28 FASI, FAS	Randomized vaccine group in the primary series	<u>For D0 and D28</u>: Cohort 1 and Cohort 2, respectively; and pooled Cohort 1 and Cohort 2 • Groups 1, 2, 3, 4, 5, 6 • Pooled Group 1 + Group 4 • Pooled Group 2 + Group 5 • Pooled Group 3 + Group 6 <u>For D42</u>: Cohort 1 • Groups 1, 2, 3
	Immunogenicity description for booster phase	FASIB1, FASIB2 FASB1, FASB2 PPAS for booster at M12, PPAS for booster at M24 up to M36	Randomized vaccine group in the primary series	

	Immunogenicity Persistence description	FASP FASIP PPAS for booster at M24 up to M36	Randomized vaccine group in the primary series	<u>For M6, M12, M18, and M24 up to M36:</u> Cohort 2 • Groups 4, 5, 6 <u>For booster phase:</u> Cohort 1 and Cohort 2, respectively • Groups 1, 2, 3 • Groups 4, 5, 6
	Safety description	SafAS	Received vaccine group in the primary series	
		SafASB	Received vaccine group in the first dose of primary series	
Exploratory analyses	**Supplemental by-cohort and by-age group descriptive analyses for key secondary immunogenicity objectives**	PPAS FASI, FAS	Randomized vaccine group in the primary series	Similar as above for "Key Secondary Objectives"
	Other Main immunogenicity endpoints description	FASI FASIP FASIB	Randomized vaccine group in the primary series	Similar as above for "Other Secondary Objectives"
		PPAS*	Randomized vaccine group in the primary series	
	Main safety endpoints description	SafAS	Received vaccine group in the primary series	
		SafASB	Received vaccine group in the first dose of primary series	

* Only for the immunogenicity analysis of the subjects without COVID-19 concomitant medications that are used during the study or were used within 2 months before enrollment for treating/preventing (ie, Hydroxychloroquine and chloroquine).
Note: Cohort 1 includes pediatric subjects and adult subjects. Cohort 2 includes adult subjects only. Booster dose assessment is for adult subsets in Cohort 1 (M12) and Cohort 2 (M24 up to M35), respectively. Immunogenicity persistence assessment is for adult subject in Cohort 2 only.

5.3 Handling of Missing Data and Outliers

5.3.1 Immunogenicity

No imputation of missing values and no test or search for outliers will be performed. Only the available (non-missing) data will be included in the analysis, unless otherwise specified.

5.3.2 Safety

5.3.2.1 Immediate

For unsolicited systemic AEs, a missing response to the "Immediate" field is assumed to have occurred after the 30-minute surveillance period and will not be imputed.

5.3.2.2 Causality

By convention, all events reported at the investigational or control vaccines injection site (either solicited or unsolicited) will be considered as related to the study vaccine (investigational product, including VRVg-2, Verorab® and Imovax® Rabies) and then referred to as reactions.In a same way, all solicited systemic events pre-listed in the eCRF are also considered as related to study vaccination and will be considered as reactions.

For unsolicited systemic AE, missing relationship to the study vaccine (investigational product, including VRVg-2, Verorab® and Imovax® Rabies) in eCRF will be considered as related to study vaccine at the time of analysis.

The missing relationship to study procedures for SAEs will not be imputed.

5.3.2.3 Measurements

Partially missing temperatures will be handled as described in Section 4.3.1.1.1.

5.3.2.4 Intensity

For solicited reactions, missing intensities will be handled as described in Section 4.3.1.1.1. For unsolicited AEs, missing intensities will remain missing and will not be imputed.

5.3.2.5 Start Date and Stop Date

Missing or partially missing start dates or end dates for unsolicited AEs will remain missing and not be imputed. If the start date is missing or partially missing, the time of onset will be considered to be missing. Nevertheless, unsolicited AEs with missing time of onset will be included in analyses according to the visit collected.

Missing or partially missing end dates for AEs (solicited reactions and unsolicited AEs) will remain missing and not be imputed.

5.3.2.6 Action Taken

Missing actions taken will remain missing and not be imputed.

5.3.3 Efficacy

Not applicable.

5.4 Interim / Preliminary Analysis

The statistical analysis will be performed in 4 steps:

- The first statistical analysis will be done once all immunogenicity data collected up to D28 (V03, 21 days after the second vaccination) in the primary series of Cohort 2 and all safety data up to D35 (V04, 28 days after the second vaccination) in the primary series of Cohort 2 are available (ie, at the end of 6-month safety follow-up of the primary series and booster phase in Cohort 1; and up to D35 [V04] in the primary series of Cohort 2). The randomization and vaccination exposure information for all enrolled subjects will be unblinded at the time of first statistical analysis.
- The second statistical analysis will be carried out once the 12-month immunogenicity persistence data and safety data in Cohort 2 (V06) are collected.
- The third statistical analysis will be carried out once all the immunogenicity and safety data up to 28 days after the booster dose in Cohort 2 (V10) are collected.
- The fourth statistical analysis will be carried out once the 6-month safety follow-up data post the booster vaccination in Cohort 2 (M30 up to M42) are collected.

No statistical adjustment is necessary because there are no repeat analyses of the same parameter.

5.5 Determination of Sample Size and Power Calculation

5.5.1 Primary Series

Original Plan:

An alpha level of 2.5% (one-sided hypothesis) has been chosen to calculate the sample size.

Originally, assuming a proportion of subjects with an RVNA titer ≥ 0.5 IU/mL of 99% for both VRVg-2 and control vaccines in each age group at D42, a clinical acceptable difference of -5% for the difference of proportions at D42 betweenVRVg-2 and the control vaccines, and a power of at least 95% for each of the NI testing in the primary objective, with an unbalanced randomization ratio of 3:1:1 (VRVg-2: Verorab vaccine: Imovax Rabies vaccine), 258 evaluable subjects in the VRVg-2 group, and 86 evaluable subjects in each of the Verorab and Imovax Rabies vaccine groups who are planned to receive 3 vaccinations (at D0, D7 and D28) in primary series in each age group, will be necessary to provide a global power of 81.8% for the primary NI objective, using the Farrington and Manning (FM) method.

If the primary objective is met at D42, a total of 516 subjects evaluable in the VRVg-2 group will ensure 85.0% of power for the 1st secondary immunogenicity objective of superiority at D42. If the 1st secondary immunogenicity objective is met, overall power for the 2nd secondary immunogenicity NI objective of 2-dose VRVg-2 at D28 versus 2-dose comparator vaccines at D28 will be higher than 80%.

Under the assumption that 15% of subjects will not be evaluable in the PPAS, 303 subjects in the VRVg-2 group and 101 subjects in each of the Verorab and Imovax Rabies vaccine groups must be enrolled in each age group of the study per the original plan, as Primary Series Cohort 1.

Updated Plan for Cohort 1 and Cohort 2 (Protocol Amendments 2, 3 and 4):

Then, according to the latest results from the VAJ00001 study and to be conservative, the estimation of proportion of adult subjects with an RVNA titer ≥ 0.5 IU/mL at D28 was adjusted from 99% to 96.5% for both VRVg-2 and control vaccines. With the aim to secure the study power for the 2nd secondary immunogenicity NI objective at D28 and maintain the randomization ratio, 609 evaluable adults in the VRVg-2 group, and 203 evaluable adults in each of the control vaccine group (Verorab and Imovax Rabies) will be necessary to provide a power of 81.7% to demonstrate the 2nd secondary immunogenicity objective at D28, using the FM method.

If the 2nd secondary immunogenicity objective is met at D28, a total of 867 subjects evaluable in the VRVg-2 group will ensure 99.2% of power for the secondary objective of superiority for VRVg-2 at D28 in the overall subjects (pooled pediatric and adult subjects).

Therefore, under the attrition rate of 15%, 690 additional adults (414 in VRVg-2; 138 in Verorab; 138 in Imovax Rabies) were planned to be enrolled in the study as Cohort 2 to receive 2 vaccinations at D0 and D7 in the primary series. Those 690 additional adults in Cohort 2 were planned to be pooled with the 505 adults enrolled in Cohort 1, for testing the 2nd secondary NI objective at D28. The 414 additional adults in VRVg-2 group in Cohort 2 were planned to be pooled with the 303 pediatric subjects and 303 adult subjects in VRVg-2 group in Cohort 1, for testing the secondary superiority objective for VRVg-2 at D28.

Therefore, a total of 1700 subjects (505 pediatric subjects and 1195 adult subjects) were planned to be enrolled in total by 2 Cohorts, including 505 pediatric subjects and 505 adult subjects in Cohort 1 who are planned to receive 3 doses of vaccinations in the primary series, and 690 adult subjects in Cohort 2 who were planned to receive 2 doses of vaccinations in the primary series.

As a consequence, a total of 1708 subjects (505 pediatric subjects and 1203 adult subjects) were actually enrolled, including 505 pediatric subjects and 505 adult subjects in Primary Series Cohort 1 and 698 adult subjects (including 8 replacements) in Primary Series Cohort 2.

The 3:1:1 design is chosen to optimize the NI testing for immunogenicity and to increase the size of the safety database.

Updated power estimations with additional objectives (Protocol Amendment 5):

Based on live blinded data observed from Cohort 1, the estimated attrition rate of Cohort 1 was adjusted from 15% to about 13.5% at both D28 and D42 for pediatric subjects, and adjusted to about 21.0% at D28 and 19.8% at D42 for adult subjects, respectively. Assuming the actual attrition rate of adult subjects in Cohort 2 is similar to Cohort 1, the adjusted estimations for evaluable number of subjects in the PPAS in each age group are as below:

- Pediatric subjects (Cohort 1 only): at both D28 and D42, there will be about 261 and 88 evaluable subjects in VRVg-2 group and each of the comparator vaccine groups, respectively.
- Adult subjects: at D28, there will be about 570 and 190 evaluable subjects in VRVg-2 group and each of the comparator vaccine groups, respectively (pooled Cohort 1 and Cohort 2); at D42, there will be about 243 and 81 evaluable subjects in VRVg-2 group and each of the comparator vaccine groups, respectively (Cohort 1 only).

Moreover, 2 new secondary immunogenicity objectives were added as below, both with a NI margin of -10%:

- Secondary immunogenicity objective 3: To demonstrate the NI of a 2-dose VRVg-2 PrEP at D28 versus 3-dose Imovax Rabies PrEP at D42 in each age group
- Secondary immunogenicity objective 5: To demonstrate the NI of a 2-dose Imovax Rabies PrEP at D28 versus 3-dose Imovax Rabies PrEP at D42 in the overall subjects (pooled pediatric and adult subjects) in Cohort 1

Finally, the primary objectives and all 5 key secondary immunogenicity objectives will be evaluated sequentially. Each of the key immunogenicity objective will be tested only if the previous objective is achieved.

Based on the updated study objectives and the adjusted number of evaluable subjects, the power to demonstrate each of the key immunogenicity objectives is presented as below:

Table 5.7: Sample size and power estimation for primary and key secondary immunogenicity objectives

Key Immunogenicity Objective	Age group	Evaluable N in the PPAS	Estimation/Margin	Power (%)
Primary Objective	Pediatric Subjects (Cohort[C] 1)	VRVg-2: 261 Verorab: 88	Seroconversion (SC) rate at D42: 99.0% NI margin: -5%	95.4[a]
		VRVg-2: 261 Imovax Rabies: 88	Same as above	95.4 [a]
	Adult Subjects (C1)	VRVg-2: 243 Verorab: 81	Same as above	93.8 [a]
		VRVg-2: 243 Imovax Rabies: 81	Same as above	93.8 [a]

				Overall: 80.0 [a]
Key Secondary Objectives				
#1	Overall (Pooled Pediatric and Adult Subjects in C1)	VRVg-2: 504	Sufficiency threshold: SC rate at D42 ≥99.0%, with lower limit (LL) of 95%CI ≥97.0%	**86.3** [b]
#2	Pediatric Subjects (C1)	VRVg-2: 261 Verorab: 88	SC rate at D28: 99.0% NI margin: -5%	95.4 [a]
		VRVg-2: 261 Imovax Rabies: 88	Same as above	95.4 [a]
	Adult Subjects (Pooled C1+C2)	VRVg-2: 570 Verorab: 190	SC rate at D28: 96.5% NI margin: -5%	93.8 [a]
		VRVg-2: 570 Imovax Rabies: 190	Same as above	93.8 [a]
				Overall: 80.1 [a]
#3	Pediatric Subjects (C1)	VRVg-2 at D28: 261 Imovax Rabies at D42: 88	SC rate at D28: 99.0% SC rate at D42: 99.0% NI margin: -10%	>99.9 [a]
	Adult Subjects (D28 from Pooled C1+C2, D42 from C1)	VRVg-2 at D28 (Pooled Cohort 1+ Cohort 2): 570 Imovax Rabies at D42: 81	SC rate at D28: 96.5% SC rate at D42: 99.0%NI margin: -10%	>99.9 [a]
				Overall: >99.9 [a]
#4	Overall (Pooled Pediatric and Adult Subjects in Pooled C1 and C2)	VRVg-2: 831	Sufficiency threshold: SC rate at D42 ≥99.0%, with LL of 95%CI ≥97.0%	**98.9** [b]
#5	Overall (Pooled Pediatric and Adult Subjects in C1 only)	Imovax Rabies at D28: 168 Imovax Rabies at D42: 168	SC rate at D28: 96.5% SC rate at D42: 99.0% NI margin: -10%	**90.0** [c]

a Power calculated using Farrington and Manning (FM) method.

b Power calculated using Binomial Exact method.

c Power calculated using simulation (10000 times, assuming correction coefficient [Rho]=0.5), based on general linear model (GLM) for repeated measured data with categorical response under binomial distribution (link function=identify).

5.5.2 Booster Phase

A subset of 170 adults in Primary Series Cohort 1 who received the 3-dose PrEP regimen in the primary series will be included in the booster phase of the study at M12; having 102 subjects receiving the complete 3-dose regimen schedule with VRVg-2 (primary series + booster) and 170 subjects receiving VRVg-2 as a booster (regardless the vaccine received in the primary series) in the FASB1 will provide the following level of precision around the expected percentage of subjects with an RVNA titer ≥ 0.5 IU/mL at D14 after the booster vaccination at M12:

Number of subjects with RVNA titer ≥ 0.5 IU/mL on M12+D14	**FASB1 – VRVg-2 group (N=102)**		**Number of subjects with RVNA titer ≥ 0.5 IU/mL on M12+D14**	**FASB1 (N=170)**	
	% observed	**95% CI**		**% observed**	**95% CI**
102	100.0	(96.45 – 100)	**170**	100.0	(97.85 – 100)
101	99.0	(94.66 – 99.98)	**169**	99.4	(96.77 – 99.99)
100	98.0	(93.10 – 99.76)	**168**	98.8	(95.81 – 99.86)
99	97.1	(91.64 – 99.39)	**167**	98.2	(94.93 – 99.63)
98	96.1	(90.26 – 98.92)	**166**	97.6	(94.09 – 99.36)

Note: The 95% CI for the single proportion is calculated using the exact binomial method (Clopper-Pearson method), quoted by Newcombe (1).

A subset of 230 adults who received the one week 2-dose PrEP regimen will be included in the Immunogenicity Persistence and Booster Phase Cohort 2. Assuming approximate 15% of subjects will not be evaluable in the FASB2 for booster evaluation at M24 up to M36, having 117 subjects receiving the complete 2 doses regimen schedule with VRVg-2 (primary series + booster) and 195 evaluable subjects receiving VRVg-2 as a booster (regardless the vaccine received in the primary series) will provide the following level of precision around the expected percentage of subjects with an RVNA titer ≥ 0.5 IU/mL at D14 after the booster vaccination at M24 up to M36:

Number of subjects with RVNA titer ≥ 0.5 IU/mL between M24 up to M36+D14	**FASB2 – VRVg-2 group (N=117)**		**Number of subjects with RVNA titer ≥ 0.5 IU/mL between M24 up to M36+D14**	**FASB2 (N=195)**	
	% observed	**95% CI**		**% observed**	**95% CI**
117	100.00	(96.90; 100)	**195**	100.00	(98.13; 100)
116	99.15	(95.33; 99.98)	**194**	99.49	(97.18; 99.99)
115	98.29	(93.96; 99.79)	**193**	98.97	(96.34; 99.88)

114	97.44	(92.69; 99.47)	**192**	98.46	(95.57; 99.68)
113	96.58	(91.48; 99.06)	**191**	97.95	(94.83; 99.44)

Note: The 95% CI for the single proportion is calculated using the exact binomial method (Clopper-Pearson method), quoted by Newcombe (1).

5.6 Data Review for Statistical Purposes

A treatment blind review of the data has been anticipated through the data review process led by data management before database lock. This review of the data included a statistical review.

5.7 Changes in the Conduct of the Trial or Planned Analyses

Not applicable.

6 References List

1 Newcombe R.G., Two-sided confidence intervals for the single proportion: comparison of seven methods. Statistics in Medicine, 1998;17, 857-872.

2 Newcombe RG. Interval estimation for the difference between independent proportions: comparison of eleven methods. Stat Med. 1998;17(8):873-90.

3 Collection of Race and Ethnicity Data in Clinical Trials – Guidance for Industry and Food and Drug Administration Staff [October 2016]. Available from: www.fda.gov/ucm/groups/fdagov-public/@fdagov-afda-gen/documents/document/ucm126396.pdf.

4 Farrington CP, Manning G. Test statistics and sample size formulae for comparative binomial trials with null hypothesis of non-zero risk difference or non-unity relative risk. Stat Med. 1990;9(12):1447-54.

5. Food and Drug Administration (FDA). Multiple Endpoints In Clinical Trials Guidance for Industry. [Available from: https://www.fda.gov/media/102657/download

6. European Medicines Agency (EMA). Guideline on multiplicity issues in clinical trials. EMA/CHMP/44762/2017 2016 [Available from: https://www.ema.europa.eu/en/documents/scientific-guideline/draft-guideline-multiplicity-issues-clinical-trials_en.pdf.

7. Alosh, M., Bretz, F. and Huque, M. (2014), Advanced multiplicity adjustment methods in clinical trials. Statist. Med., 33: 693-713. https://doi.org/10.1002/sim.5974

8. SAS. Usage Note 46997: Estimating the risk (proportion) difference for matched pairs data with binary response. Available from: https://support.sas.com/kb/46/997.html.

9. Fagerland MW, Lydersen S, Laake P. Recommended tests and confidence intervals for paired binomial proportions. Stat Med. 2014;33(16):2850-2875.