NCT03537508

Immunogenicity and Safety Study of an Investigational Quadrivalent Meningococcal Conjugate Vaccine when Administered Concomitantly with Routine Pediatric Vaccines in Healthy Infants and Toddlers

A Phase III, partially modified double-blind, randomized, parallel-group, active-controlled, multi-center study to compare the immunogenicity and describe the safety of MenACYW conjugate vaccine and MENVEO® when administered concomitantly with routine pediatric vaccines to healthy infants and toddlers in the United States

Statistical Analysis Plan (SAP) - Core Body Part

Trial Code:	MET42
Development Phase:	Phase III
Sponsor:	Sanofi Pasteur Inc. Discovery Drive, Swiftwater, PA 18370-0187, US
Investigational Products:	MenACYW conjugate vaccine: Meningococcal Polysaccharide (Serogroups A, C, Y, and W) Tetanus Toxoid Conjugate Vaccine
Form/Route:	Liquid Solution / Intramuscular
Indication For This Study:	MenACYW conjugate vaccine administered as a 4-dose series to healthy infants and toddlers
Version and Date of the SAP core body part:	Version 3.0, 22Jan2024

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List of Abbreviations

Ab	antibody(ies)
ACIP	Advisory Committee on Immunization Practices
AE	adverse event
AESI	adverse event of special interest
AR	adverse reaction
BL	blood sample
CBER	Center for Biologics and Research
CFU	colony - forming unit
CI	confidence interval
CO2	carbon dioxide
CRB	electronic case report book (a collection of all the case report forms for a subject)
CSR	clinical study report
eCRF	electronic case report form
D	day
DC	diary card
DOD	delta optical density
DTaP	diphtheria, tetanus, pertussis (acellular, component) vaccine
ECL	electrochemiluminescent
EDC	electronic data capture
ELISA	enzyme-linked immunosorbent assay
FAS	Full Analysis Set
FHA	filamentous hemagglutinin
FIM	fimbriae types 2 and 3
FVFS	first visit, first subject
FVLS	first visit, last subject
GBS	Guillain-Barré syndrome
GCI	Global Clinical Immunology
GM	geometric mean
GMC	geometric mean concentrations
GMT	geometric mean titer
GMTR	geometric mean titer ratio
hSBA	serum bactericidal assay using human complement
HB	hepatitis B

HepA	hepatitis A
Hib	*Haemophilus influenzae* type b
H0	null hypothesis
H1	alternative hypothesis
ICF	informed consent form
ICH	International Conference on Harmonisation
Ig	Immunoglobulin
IMD	invasive meningococcal disease
IME	important medical event
IMP	investigational medicinal product
IPV	poliovirus (inactivated) vaccine
IRB	Institutional Review Board
IRT	interactive response technology
ITP	idiopathic thrombocytopenic purpura
LCLS	last contact, last subject
LLOQ	lower limit of quantitation
LLT	lowest level term
MA	memory aid
MAAE	medically-attended adverse event
MedDRA	Medical Dictionary for Regulatory Activities
MCV4	quadrivalent meningococcal conjugate vaccine
MMR	measles, mumps, and rubella
mIU	milli-international units
mL	milliliter(s)
NIMP	non-investigational medicinal product
Ng	nanogram
N.	*Neisseria*
OD	optical density
PCV13	pneumococcal 13-valent conjugate vaccine
PA	Pennsylvania
PPAS	Per-Protocol Analysis Set
PRN	Pertactin
PRP	polyribosyl-ribitol phosphate
PT	pertussis toxoid / toxin
RCDC	reverse cumulative distribution curve
RIA	radioimmunoassay
RV5	pentavalent rotavirus vaccine
SAE	serious adverse event

SafAS	Safety Analysis Set
SAP	statistical analysis plan
SOC	system organ class
TCC	tissue culture control
UAR	unexpected adverse reaction
ULOQ	upper limit of quantitation
V	visit
vs.	versus
VZV	varicella-zoster virus
WHO	World Health Organization
WT	wild-type

1 Introduction

Study (MET42) evaluates the safety and immunogenicity of a 4-dose series of the quadrivalent Meningococcal Polysaccharide (Serogroups A, C, Y, and W) Tetanus Toxoid Conjugate Vaccine (hereafter referred to as MenACYW conjugate vaccine) in healthy infants and toddlers in the US in comparison to MENVEO®, when both are administered concomitantly with routine pediatric vaccines as per Advisory Committee on Immunization Practice (ACIP) vaccine recommendations. Invasive meningococcal disease (IMD) is a serious illness caused by the bacterium *Neisseria meningitidis (N. meningitidis)*, a Gram-negative diplococcus found exclusively in humans. Symptoms may include headache, fever, nausea, vomiting, photophobia, stiff neck, lethargy, myalgia, and a characteristic petechial rash (2). Worldwide, most cases of meningococcal disease are caused by serogroups A, B, C, X, W, and Y (2) (3) (4).The epidemiology of *N. meningitidis* can be described as complex, unpredictable, geographically variable, and changing over time. Meningococcal disease occurs worldwide in both endemic and epidemic forms with seasonal variation. In Europe, the incidence rate of IMD has remained stable over the last 5 to 10 years, with the highest peak occurring in the population less than 4 years of age and a smaller peak in the 15 to 19-year-old age-group. In the US, the incidence rate of IMD in 2013 was 0.14 per 100 000 in all ages, 0.83 per 100 000 in infants less than 1 year of age, 0.62 per 100 000 in toddlers 1 year of age, 0.27 per 100 000 in toddlers and children 2 to 4 years of age, and 0.02 per 100 000 in children and adolescents 5 to 17 years of age. The age specific incidence rate in 2013 per 100 000 was 0.08 in adults 50 to 64 years of age, 0.03 in adults 65 to 74 years of age, 0.14 in adults 75 to 84 years of age, and 0.43 in adults 85 years of age and older (5).

The goal for MenACYW conjugate vaccine is to provide broad protection against IMD caused by serogroups A, C, Y, and W in all age groups including children as young as 6 weeks of age, adolescents, and adults, including those 56 years of age and older.

MenACYW conjugate vaccine has been evaluated in 17 clinical trials: a Phase I trial, MET28, conducted in Canada, a Phase I/II study, MET32, conducted in Australia, 4 Phase II studies, MET39, MET44, MET50, conducted in the USA, and MET54 conducted in Finland, and 6 Phase III studies, MET35, MET43, MET49 and MET56, conducted in the USA, MET51 conducted in EU region (Spain, Germany, Hungary and Finland), and MET57 conducted in Thailand, South Korea, Russia, and Mexico.

MenACYW conjugate vaccine was found to be well tolerated and no unanticipated or new significant safety concerns have been identified in the clinical trials completed to date. The safety profile of MenACYW conjugate vaccine was comparable to that of the meningococcal licensed vaccine used as comparators in the completed studies.
The immunogenicity of MenACYW conjugate vaccine has been demonstrated to be non-inferior to that of the standard of care meningococcal vaccines used as control vaccines in the completed trials with NIMENRIX®, MENVEO®, MENACTRA®, and MENOMUNE®.

Three Phase III studies (including MET42) will generate data to primarily support the licensing of the MenACYW conjugate vaccine in the US with an infant/toddler indication from 6 weeks of

age. MET42 study will generate data which will also significantly contribute towards the overall safety database of the MenACYW conjugate vaccine in US.

2 Study Objectives

2.1 Primary Objectives

1) To demonstrate the non-inferiority of the hSBA (serum bactericidal assay using human complement) vaccine seroresponse[a] to meningococcal serogroups A, C, Y, and W following the administration of a 4-dose series of MenACYW conjugate vaccine compared to a 4-dose series of MENVEO® when given concomitantly with routine pediatric vaccines to infants and toddlers 6 weeks old to 15 months old.
2) To demonstrate the non-inferiority of the hSBA antibody response to meningococcal serogroups A, C, Y, and W following the administration of 3 doses in infancy of MenACYW conjugate vaccine compared to 3 doses in infancy of MENVEO® when given concomitantly with routine pediatric vaccines to infants at 2, 4, and 6 months of age.

2.2 Secondary Objectives

1) To demonstrate the non-inferiority of immune responses of the routine pediatric vaccines administered concomitantly with MenACYW conjugate vaccine as compared with MENVEO® in infants and toddlers 6 weeks old to 18 months old.
2) To assess the antibody responses against meningococcal serogroups A, C, Y, and W after the administration of the 4th dose of MenACYW conjugate vaccine or MENVEO® when both are given concomitantly with routine pediatric vaccines at 12 months of age.
3) To assess the persistence of bactericidal antibodies at 12 months of age in subjects who previously received 3 doses of MenACYW conjugate vaccine or MENVEO® in infancy concomitantly with routine pediatric vaccines at 2, 4, and 6 months of age.
4) To describe the antibody responses against the antigens of the routine pediatric vaccines (Pentacel®, Prevnar 13®, M-M-R® II, VARIVAX®, RotaTeq®, and ENGERIX-B®) when administered concomitantly with either MenACYW conjugate vaccine or MENVEO®
5) To describe the antibody responses against meningococcal serogroups A, C, Y, and W when MenACYW conjugate vaccine versus (vs) MENVEO® is administered concomitantly with routine pediatric vaccines.
6) To describe the antibody responses against meningococcal serogroups A, C, Y, and W when MenACYW conjugate vaccine is administered to children 12 to 15 months of age vs when MenACYW conjugate vaccine is administered to children 15 to 18 months of age, concomitantly with routine pediatric vaccines (Subgroup 1a vs Subgroup 1b), including the bactericidal antibodies persistence and the effect of 4th dose of MenACYW conjugate vaccine at 12 to15 months of age or 15 to 18 months of age.

[a] hSBA vaccine seroresponse for serogroups A, C, Y, and W is defined as:

- For a subject with a pre-vaccination titer < 1:8, the post-vaccination titer must be ≥ 1:16
- For a subject with a pre-vaccination titer ≥ 1:8, the post-vaccination titer must be ≥ 4-fold greater than the pre-vaccination titer

2.3 Observational Objectives

Safety

To describe the safety profile of MenACYW conjugate vaccine and MENVEO® when administered concomitantly with routine pediatric vaccines in healthy infants and toddlers.

3 Description of the Overall Study Design and Plan

3.1 Study Design

This study will be a Phase III, partially modified double-blind, randomized, parallel-group, active-controlled, multi-center study to compare the immunogenicity and safety of MenACYW conjugate vaccine and MENVEO® (Meningococcal [Groups A, C, Y, and W-135] Oligosaccharide Diphtheria CRM_{197} Conjugate Vaccine) when administered concomitantly with routine pediatric vaccines to healthy infants and toddlers in the US.
Approximately 2628 healthy infants aged ≥ 42 to ≤ 89 days will be randomized 2:1 to the following 2 groups:

- **Group 1 (G1):** MenACYW conjugate vaccine and routine vaccines
- **Group 2 (G2):** MENVEO® and routine vaccines

Each group will be further randomized 2:1 in 2 subgroups based on the time of analyses conducted in the 2nd year of life (30 days after the 12-month vaccination or 30 days after the 15-month vaccination, respectively):

- **Group 1:**
  - **Subgroup 1a (G1a)** (12 months): MenACYW conjugate vaccine and routine vaccines at 2, 4, 6, and 12 to 15 months of age
  - **Subgroup 1b (G1b)** (15 months): MenACYW conjugate vaccine at 2, 4, 6, and 15 to 18 months of age and routine vaccines at 2, 4, 6, 12 to 15 months of age, and 15 to 18 months of age
- **Group 2:**
  - **Subgroup 2a (G2a)** (12 months): MENVEO® at 2, 4, 6, and 12 months of age and routine vaccines at 2, 4, 6, 12, and 15 to 18 months of age
  - **Subgroup 2b (G2b)** (15 months): MENVEO® at 2, 4, 6, and 12 months of age and routine vaccines at 2, 4, 6, 12, and 15 to 18 months of age

To be specific, the time of analyses as well as the collection of blood samples conducted in the 2nd year of life for Subgroup 1a and 2a is 30 days after the 12-month vaccination. The time for Subgroup 1b and 2b is 30 days after the 15-month vaccination. The schedule of vaccination and blood sampling is further detailed in Table 3.1.

All subjects will receive the following routine vaccines as per the ACIP recommendations (see Table 3.1).

- Pentacel® (diphtheria, tetanus, pertussis [acellular, component]-poliovirus [inactivated]//*Haemophilus influenzae* type b [DTaP-IPV//Hib]) at 2, 4, 6, and 15 to 18[a] months of age
- Prevnar 13® (pneumococcal 13-valent conjugate vaccine [PCV13]) at 2, 4, 6, and 12 to 15 months of age
- RotaTeq® (pentavalent rotavirus vaccine [RV5]) at 2, 4, and 6 months of age
- ENGERIX-B® (HB vaccine) at 2 and 6 months of age[b]
- M-M-R® II (measles, mumps, rubella [MMR] vaccine) at 12 to 15 months of age
- VARIVAX® (varicella vaccine) at 12 to 15 months of age

In addition, subjects in Subgroup 1b and Group 2 will receive the first dose of hepatitis A (HepA) vaccine (HAVRIX®) at 15 to 18 months of age as part of the study. For Subgroup 1a, there will be no HepA vaccination provided as part of the study. Subjects in Subgroup 1a should be vaccinated, as per standard practice, after the completion of the last study visit.
Blood sample for immunogenicity assessment will be assayed for meningococcal antibodies and for antibodies elicited in response to selected routine pediatric vaccines.
Safety data will be collected as follows: Immediate unsolicited systemic adverse events (AEs) will be collected within 30 minutes after each vaccination. Solicited-AE information will be collected from D0 to D07 after each vaccination; unsolicited-AE information will be collected from D0 to D30 after each vaccination; Serious adverse events (SAE) (including adverse events of special interest [AESIs]) and medically-attended-adverse-event (MAAE) information will be collected throughout the study from Visit 1 until the end of the 6-month follow-up period after the last vaccination.

3.2 Study Plan

Vaccination

A schedule of assessments and study vaccinations is provided in the Tables of Study Procedures and in Table 3.1.

Blood Sampling

All subjects in the study will have 4 blood draws each. Table 3.1 presents the schedule for blood-sampling. The schedule of antigen testing is detailed in Table 3.2.

[a] Subjects in Subgroup 1a will complete the last study visit at 13 to 16 months of age. For subjects in Subgroup 1a, the 4th dose of Pentacel® which is administered at 15 to 18 months of age, will be provided by the Sponsor for completion of the DTaP series with vaccine from the same manufacturer, as per ACIP recommendation. The study personnel / Investigator will be responsible for administering this dose at the recommended age outside of the scope of the study.

[b] First dose of HB vaccine must be given at least 28 days prior to study enrollment.

Table 3.1: Vaccination and blood sampling schedule

Age in months	**2 months**		**4 months**	**6 months**	**7 months**		**12 months**		**13 months**	**15 months**		**16 months**
Visit #	**Visit 1**		**Visit 2**	**Visit 3**	**Visit 4**		**Visit 5***		**Visit 6: 1a† and 2a**	**Visit 6: 1b and 2b Visit 7: 2a**		**Visit 7: 1b and 2b Visit 8: 2a**
Group	**Blood Draw‡**	**Vaccines**	**Vaccines**	**Vaccines**	**Blood Draw**	**Subgroup**	**Blood Draw‡**	**Vaccines**	**Blood Draw**	**Blood Draw‡**	**Vaccines**	**Blood Draw**
1	X	**MenACYW** Pentacel PCV13 rotavirus hepatitis B§	**MenACYW** Pentacel PCV13 rotavirus	**MenACYW** Pentacel PCV13 rotavirus hepatitis B	X	**1a**	X	**MenACYW** MMR varicella PCV13	X	No study visit *Pentacel***		
						1b		MMR varicella PCV13	No study visit	X	**MenACYW** Pentacel hepatitis A	X
2	X	**MENVEO** Pentacel PCV13 rotavirus hepatitis B§	**MENVEO** Pentacel PCV13 rotavirus	**MENVEO** Pentacel PCV13 rotavirus hepatitis B	X	**2a**	X	**MENVEO** MMR varicella PCV13	X		Pentacel hepatitis A	
						2b		**MENVEO** MMR varicella PCV13	No study visit	X	Pentacel hepatitis A	X

*Visit 5 will occur at 12 months of age for Subgroups 2a and 2b, and at 12 to 15 months of age for Subgroups 1a and 1b.
†Last study visit for Subgroup 1a. Routine vaccine can be administered as per standard of care after study procedures are completed. Visit 6 will occur at 13 to16 months of age for Subgroup 1a. For Subgroup 2a Visit 6 occurs at 13 months of age.
‡Blood will be drawn prior to vaccinations.
§ The first dose of HB vaccine must have been received at least 28 days prior to the first study vaccination at Visit 1.
**Subjects in Subgroup 1a will complete the last study visit at 13 to 16 months of age. Pentacel will be provided by the Sponsor to complete the DTaP series with vaccine from the same manufacturer and should be administered as per standard of care. The study personnel / Investigator will be responsible for administering this dose at the recommended age outside of the scope of the study.

Table 3.2: Schedule of antigen testing

Age in months	2 months	7 months		12 months*	13 months*	15 months*	16 months*
Visit #	Visit 1 (pre-vaccination)	Visit 4		Visit 5 (pre-vaccination)	Visit 6: 1a and 2a	Visit 6: 1b and 2b (pre-vaccination)	Visit 7: 1b and 2b
Group			Subgroup				
Group 1 (N=1752)	Meningococcal serogroups A, C, W, Y Pertussis (PT, FHA, PRN, FIM) Rotavirus (IgA)	Meningococcal serogroups A, C, W, Y Hib (PRP) Hep B (HBsAg) Poliovirus (types 1, 2, 3) Pertussis (PT, FHA, PRN, FIM) PCV13† Rotavirus (IgA) Diphtheria Tetanus	**1a** (N=1168)	Meningococcal serogroups A, C, W, Y	Meningococcal serogroups A, C, W, Y Measles Mumps Rubella Varicella PCV13†		
			1b (N=584)			Pertussis (PT, FHA, PRN, FIM) Meningococcal serogroups (A, C, W, Y) Hib (PRP)	Meningococcal serogroups A, C, W, Y Pertussis (PT, FHA, PRN, FIM) Hib (PRP) Polio (types 1,2, 3) Diphtheria Tetanus

Age in months	**2 months**	**7 months**		**12 months***	**13 months***	**15 months***	**16 months***
Visit #	**Visit 1 (pre-vaccination)**	**Visit 4**		**Visit 5 (pre-vaccination)**	**Visit 6: 1a and 2a**	**Visit 6: 1b and 2b (pre-vaccination)**	**Visit 7: 1b and 2b**
Group			**Subgroup**				
Group 2 (N=876)	Meningococcal serogroups A, C, W, Y Pertussis (PT, FHA, PRN, FIM) Rotavirus (IgA)	Meningococcal serogroups A, C, W, Y Hib (PRP) Hep B (HBsAg) Poliovirus (types 1, 2, 3) Pertussis (PT, FHA, PRN, FIM) PCV13† Rotavirus (IgA) Diphtheria Tetanus	**2a** (N=584)	Meningococcal serogroups A, C, W, Y	Meningococcal serogroups A, C, W, Y Measles Mumps Rubella Varicella PCV13†		
			2b (N=292)			Pertussis (PT, FHA, PRN, FIM) Hib (PRP)	Pertussis (PT, FHA, PRN, FIM) Hib (PRP) Polio (types 1,2, 3) Diphtheria Tetanus
Blood volume	3 mL	6 mL		5 mL	5 mL	5 mL	5 mL

Note: For each visit and group or subgroup, antigens are listed in descending order of assay priority (highest to lowest priority).

*During the 2nd year of life study visits, if blood is collected as part of the study, additional blood can be optionally collected via the same venipuncture for screening of iron deficient anemia and/or lead level as per AAP/CDC recommendations. This collection must be done in serum tubes not provided as part of the study and as per local standard of care.

†Anti-pneumococcal serotypes 1, 3, 5, 6A, 7F, 19A, 4, 6B, 9V, 14, 18C, 19F, and 23F.

Collection of safety data

- All subjects will be followed for safety from Visit 1 to 6 months after the last vaccination.
- All subjects will be observed for 30 minutes after each vaccination, and any unsolicited systemic AEs occurring during that time will be recorded as immediate unsolicited systemic AEs in the electronic case report book (CRB).
- The subject's parent/guardian will record information in a diary card about solicited reactions from D0 to D07 after each vaccination and unsolicited AEs from D0 after each vaccination to the next study visit.
- SAEs and MAAEs will be recorded throughout the study. The subject's parent/guardian will record information in a diary card about SAEs and MAAEs from Visit (V) 01 to V02, from V02 to V03, from V03 to V04, from V04 to V05, from V05 to V06, from V06 to V07 (for Subgroups 1b, 2a, and 2b), and from V07 to V08 (for Subgroup 2a only). SAEs and MAAEs will also be recorded in a memory aid (MA) from D31 after the last vaccination visit, until the 6-month follow up phone call. The subject's parent/guardian will be asked to notify the site immediately about any potential SAE at any time during the study.
- Staff will contact the subject's parent/guardian by telephone 8 days (+2 days) after each vaccination visit to identify the occurrence of any SAEs (including AESIs) and/or MAAEs not yet reported and to remind them to complete the diary card and to bring it back at the subsequent visit.
- The completed diary cards will each be collected and reviewed with the subject's parent/guardian at the subsequent visit.
- Staff will contact the subject's parent/guardian by telephone within 14 days before V05 to remind them about the forthcoming study visit. If the subject's participation in the study is discontinued, the information recorded on the diary card will be reviewed at this time and the diary card will be retrieved by the site.
- Staff will contact the subject's parent/ guardian by telephone at 6 months (+30 days) after the last vaccination visit to review the MA and identify the occurrence of any MAAEs and SAEs (including AESIs) that have not been reported.

Table 3.3: Study procedures for infant vaccination (aged 2 months to 7 months)

Visit / Contact	**Visit 1**	**Telephone Call (TC) 1***	**Visit 2**	**TC 2***	**Visit 3†**	**TC 3***	**Visit 4**
Approximate age of subjects	**2 months (42 – 89 days)**	**--**	**4 months**	**--**	**6 months (164 – 224 days)**	**--**	**7 months**
Trial timelines (days)	**Day 0**	**Visit 1 +8 days**	**Visit 1+ 60 days**	**Visit 2 +8 days**	**Visit 2 + 60 days**	**Visit 3 +8 days**	**Visit 3 + 30 days**
Time windows (days)	**-**	**+ 2 days**	**+ 14 days**	**+ 2 days**	**+14 days**	**+ 2 days**	**+21 days**
Informed consent	X						
Inclusion/exclusion criteria	X						
Collection of demographic data	X						
Medical history (including maternal immunization history)	X						
Physical examination ‡	X						
Temperature measurement §	X		X		X		
Contact IRT system for randomization, subject number, and vaccine allocation.	X						
Contact IRT system for vaccine assignments			X		X		
Review of temporary contraindications for blood sampling**	X						X
Blood sampling (BL)	BL1†† (3 mL)						BL2 (6 mL)
Review of warning and precautions to vaccinations	X		X		X		
Review of contraindications to subsequent vaccinations and conditions for withdrawal‡‡			X		X		
Vaccination with MenACYW conjugate vaccine or MENVEO®	X		X		X		
Vaccination with routine pediatric vaccines§§	X		X		X		
Immediate surveillance (30 minutes)	X		X		X		
Diary card (DC) provided	DC1		DC2		DC3		DC4***
Telephone call		X		X		X	
Diary card reviewed and collected			DC1		DC2		DC3
Recording of solicited injection site and systemic AEs	X		X		X		
Recording of unsolicited AEs	Collected from D0 to D30 after each vaccination						
Reporting of serious adverse events (SAEs, including AESIs) and medically-attended AEs (MAAEs)†††	To be reported throughout the trial						

Collection of reportable concomitant medications	X		X		X		X

Note: IRT: interactive response technology
*This call is made 8 days after the respective vaccinations. If day 8 falls on a weekend or holiday, the telephone call may be made on the following business day. During this telephone call, the staff will find out whether the subject experienced any SAE (including AESIs) and/or MAAE not yet reported and will remind the subject's parent/guardian to continue using the diary card, to bring the diary card to the study center at the next visit, and confirm the date and time of the next visit.
†At Visit (V) 03, subjects must be at least 24 weeks of age (164 days), the minimum age for the final dose of HB in the vaccine series, and no more than 32 weeks of age (the maximum age for the 3rd dose of rotavirus vaccine administration).
‡Physical examination should be performed as per standard of care.
§Temperature needs to be measured before each vaccination and recorded in the source documents.
**Should a subject receive oral or injectable antibiotic therapy within 3 days prior to any blood draw, the investigator will postpone that blood draw until it has been 3 days since the subject last received oral or injectable antibiotic therapy. Postponement must still be within the timeframe for blood draw. If postponement would result in the sample collection falling outside of this timeframe, the blood sample should be collected without postponement, and it should be appropriately documented that the sample was taken less than 3 days after stopping antibiotic treatment.
††Blood sample will be drawn prior to vaccinations.
‡‡Physical examination should be performed on the basis of relevant medical history at the time of the visit according to the investigator's clinical judgment. Temperature needs to be measured before each vaccination and recorded in the source documents
§§Routine pediatric vaccines at V01: Pentacel®, Prevnar 13®, RotaTeq®, ENGERIX-B®; at V02: Pentacel®, Prevnar 13®, and RotaTeq®; at V03: Pentacel®, Prevnar 13®, RotaTeq®, ENGERIX-B®
***The diary card is used only for the recording of SAEs and medically-attended AEs from D31 after completion of the first year's vaccinations (V04) until the vaccination visit in the second year of life (V05). The subject's parent /guardian will bring the completed diary card to the study site at V05.
†††AESIs will be collected throughout the trial as SAEs to ensure that the events are communicated to the Sponsor in an expedited manner and followed up until the end of the follow-up period or resolution, as per the assigned causality.

Table 3.4: Study procedures for Subgroup 1a

Phase III trial, 6 Visits, 4 Vaccination Visits, 6 Telephone Calls, 4 Blood Samples, 16 to 19 Months Duration per Subject

Visit / Contact	**Visit 1**	**Telephone Call (TC) 1***	**Visit 2**	**TC 2***	**Visit 3**	**TC 3***	**Visit 4**	**TC4†**	**Visit 5‡**	**TC5***	**Visit 6**	**TC6§ Follow-up Contact**
Approximate age of subjects	**2 months (42 – 89 days)**	**--**	**4 months**	**--**	**6 months (164 – 224 days)**	**--**	**7 months**		**12 - 15 months**	**--**	**13 - 16 months**	**18 - 21 months**
Trial timelines (days)	**Day 0**	**Visit 1 + 8 days**	**Visit 1+ 60 days**	**Visit 2 +8 days**	**Visit 2 + 60 days**	**Visit 3 +8 days**	**Visit 3 + 30 days**	**Visit 5 – 14 days**		**Visit 5 +8 days**	**Visit 5 + 30 days**	**Visit 5 + 180 days**
Time windows (days)	**-**	**+ 2 days**	**+ 14 days**	**+ 2 days**	**+ 14 days**	**+ 2 days**	**+21 days**			**+ 2 days**	**+21 days**	**+ 30 days**
Informed consent	X											
Inclusion/exclusion criteria	X											
Collection of demographic data	X											
Medical history (including maternal immunization history)	X											
Physical examination**	X								X			
Temperature measurement ††	X		X		X				X			
Contact IRT system for randomization, subject number, and vaccine allocation.	X											
Contact IRT system for vaccine assignments			X		X				X			
Review of temporary contraindications for blood sampling‡‡	X						X		X		X	
Blood sampling (BL)§§	BL1*** (3 mL)						BL2 (6 mL)		BL3*** (5 mL)		BL4*** (5 mL)	
Review of warning and precautions to vaccinations	X		X		X				X			
Review of contraindications to subsequent vaccinations and conditions for withdrawal †††			X		X				X			
Vaccination with MenACYW conjugate vaccine or MENVEO®	**X**		**X**		**X**				**X**			

Visit / Contact	Visit 1	Telephone Call (TC) 1*	Visit 2	TC 2*	Visit 3	TC 3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6	TC6§ Follow-up Contact
Vaccination with routine pediatric vaccines‡‡‡	**X**		**X**		**X**				**X**			
Immediate surveillance (30 minutes)	X		X		X				X			
Diary card (DC) provided	DC1		DC2		DC3		DC4		DC5			
Telephone call		X		X		X		X		X		X
Diary card reviewed and collected			DC1		DC2		DC3		DC4		DC5	
Recording of solicited injection site and systemic AEs	X		X		X				X			
Recording of unsolicited AEs	Collected from D0 to D30 after each vaccination							Collected from D0 to D30 after each vaccination				
Reporting of serious adverse events (SAEs, including AESIs) and medically-attended AEs (MAAEs) §§§	To be reported throughout the Study								To be reported throughout the study			
Collection of reportable concomitant medications	X		X		X		X		X		X	
Memory aid (MA) provided††††											MA	
Trial termination record (termination of active portion of the trial)											X	

IRT: interactive response technology

*This call is made 8 days after the respective vaccinations. If day 8 falls on a weekend or holiday, the telephone call may be made on the following business day. During this telephone call, the staff will find out whether the subject experienced any SAE (including AESI) and/or MAAE not yet reported and will remind the subject's parent/guardian to continue using the diary card, to bring the diary card to the study center at the next visit, and confirm the date and time of the next visit

†Staff will contact the subject's parent/ guardian by telephone within 14 days before V05 to remind them about the forthcoming study visit. If the subject's participation in the study is discontinued, the information recorded on the diary card will be reviewed at this time and the diary card will be retrieved by the site.

‡At V05, subjects must be between 12 and 15 months of age (from the day subjects become 12 months old until the day before becoming16 months old).

§Staff will contact the subject's parent/ guardian by telephone at 6 months (+ 30 days) after the last vaccination visit to identify the occurrence of any SAEs (including any AESIs) and MAAEs not yet reported. The final telephone call will continue until contact is made or 28 days have passed at which time the subject will be considered lost to follow-up.

**Physical examination should be done as per standard of care.

††Temperature needs to be measured before each vaccination and recorded in the source documents.

‡‡Should a subject receive oral or injectable antibiotic therapy within 3 days prior to any blood draw, the investigator will postpone that blood draw until it has been 3 days since the subject last received oral or injectable antibiotic therapy. Postponement must still be within the timeframe for blood draw. If postponement would result in the sample collection falling outside of this timeframe, the blood sample should be collected without postponement, and it should be documented appropriately that the sample was taken less than 3 days after stopping antibiotic treatment.

§§During the 2nd year of life study visits, if blood is collected as part of the study, additional blood can be optionally collected via the same venipuncture for screening of iron deficient anemia and/or lead level as per AAP/CDC recommendations. This collection must be done in serum tubes not provided as part of the study and as per local standard of care.

***Blood sample will be drawn prior to vaccinations.

†††Physical examination should be performed on the basis of relevant medical history at the time of the visit according to the investigator's clinical judgment. Temperature needs to be measured before each vaccination and recorded in the source documents.

‡‡‡ Routine pediatric vaccines at V05: Prevnar 13®, M-M-R® II, and VARIVAX®. At V06, routine vaccines not part of the study could be administered as per standard of care after completion of all study procedures.

§§§ AESIs will be collected throughout the trial as SAEs to ensure that the events are communicated to the Sponsor in an expedited manner and followed up until the end of the follow-up period or resolution, as per the assigned causality.

††††The MA is used only for the recording of SAEs (including AESIs) and MAAEs from Visit 6 to the 6-month follow-up phone call at TC6.

Table 3.5: Study procedures for Subgroup 1b

Phase III trial, 7 Visits, 5 Vaccination Visits, 7 Telephone Calls, 4 Blood Samples, 19 to 22 Months Duration per Subject

Visit / Contact	Visit 1	Telephone Contact (TC) 1*	Visit 2	TC2*	Visit3	TC3 *	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6§	TC6*	Visit 7	TC7** Follow-up Contact
Approximate age of subjects	**2 months (42 - 89 days)**	**--**	**4 months**	**--**	**6 months (164 – 224 days)**	**--**	**~7 mont hs**		**12 – 15 months**	**--**	**15-18 months**		**16-19 months**	**21 - 24 months**
Trial timelines (days)	**Day 0**	**Visit 1 + 8 days**	**Visit 1+ 60 days**	**Visit 2+ 8 days**	**Visit 2 + 60 days**	**Visit 3 + 8 days**	**Visit 3 + 30 days**	**Visit 5 – 14 days**		**Visit 5+ 8 days**		**Visit 6 + 8 days**	**Visit 6 + 30 days**	**Visit 6 + 180 days**
Time windows (days)	**--**	**+2 days**	**+14 days**	**+ 2 days**	**+14 days**	**+ 2 days**	**+21 days**			**+ 2 days**		**+ 2 days**	**+21 days**	**+ 30 days**
Informed consent	X													
Inclusion / exclusion criteria	X													
Collection of demographic data	X													
Medical history (including maternal immunization history)	X													
Physical examination ††	X								X					
Temperature measurement‡‡	X		X		X				X		X			
Contact IRT system for randomization, subject number, and vaccine allocation.	X													
Contact IRT system for vaccine assignments			X		X				X		X			

Visit / Contact	Visit 1	Telephone Contact (TC) 1*	Visit 2	TC2*	Visit3	TC3 *	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6§	TC6*	Visit 7	TC7** Follow-up Contact
Review of temporary contraindications for blood sampling§§	X						X				X		X	
Blood Sampling (BL)†††	BL1*** (3 mL)						BL2 (6 mL)				BL3*** (5 mL)		BL4 (5 mL)	
Review of warning and precautions to vaccinations	X		X		X				X		X			
Review of contraindications to subsequent vaccinations and conditions for withdrawal ‡‡‡			X		X				X		X			
Vaccination with MenACYW conjugate vaccine or MENVEO®	**X**		**X**		**X**						**X**			
Vaccination with routine pediatric vaccines§§§	**X**		**X**		**X**				**X**		**X**			
Immediate surveillance (30 minutes)	X		X		X				X		X			
Diary Card (DC) provided	DC1		DC2		DC3		DC4		DC5		DC6			
Telephone call		X		X		X		X		X		X		X
Diary card reviewed and collected			DC1		DC2		DC3		DC4		DC5		DC6	
Recording of solicited injection site and systemic AEs	X		X		X				X		X			
Recording of unsolicited AEs	Collected from D0 to D30 after each vaccination							Collected from D0 to D30 after each vaccination						

Visit / Contact	Visit 1	Telephone Contact (TC) 1*	Visit 2	TC2*	Visit3	TC3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6§	TC6*	Visit 7	TC7** Follow-up Contact
Reporting of serious adverse events (SAEs, including AESIs) and medically-attended AEs (MAAEs) ****	To be reported throughout the study							To be reported throughout the study						
Collection of reportable concomitant medications	X		X		X		X		X		X		X	
Memory Aid (MA) provided††††													MA	
Trial termination record (termination of active portion of the trial)													X	

IRT: interactive response technology

*This call is made 8 days after the respective vaccinations. If day 8 falls on a weekend or holiday, the telephone call may be made on the following business day. During this telephone call, the staff will find out whether the subject experienced any SAE (including AESI) and/or MAAE not yet reported and will remind the subject's parent/guardian to continue using the diary card, to bring the diary card to the study center at the next visit, and confirm the date and time of the next visit

†Staff will contact the subject's parent/ guardian by telephone within 14 days before V05 to remind them about the forthcoming study visit. If the subject's participation in the study is discontinued, the information recorded on the diary card will be reviewed at this time and the diary card will be retrieved by the site.

‡At V05, subjects must be between 12 and 15 months of age (from the day subjects become 12 months old until the day before becoming 16 months old).

§At V06, Subjects must be between 15 and 18 months of age (from the day subjects become 15 months old until the day before becoming19 months old).

**Staff will contact the subject's parent/ guardian by telephone at 6 months (+ 30 days) after the last vaccination visit to identify the occurrence of any SAEs (including any AESIs) and medically-attended AEs not yet reported. The final telephone call will continue until contact is made or 28 days have passed at which time the subject will be considered lost to follow-up

††Physical examination should be done as per standard of care.

‡‡Temperature needs to be measured before each vaccination and recorded in the source documents.

§§Should a subject receive oral or injectable antibiotic therapy within 3 days prior to any blood draw, the investigator will postpone that blood draw until it has been 3 days since the subject last received oral or injectable antibiotic therapy. Postponement must still be within the timeframe for blood draw. If postponement would result in the sample collection falling outside of this timeframe, the blood sample should be collected without postponement, and it should be documented appropriately that the sample was taken less than 3 days after stopping antibiotic treatment

***Blood sample will be drawn prior to vaccinations.

†††During the 2nd year of life study visits, if blood is collected as part of the study, additional blood can be optionally collected via the same venipuncture for screening of iron deficient anemia and/or lead level as per AAP/CDC recommendations. This collection must be done in serum tubes not provided as part of the study and as per local standard of care.

‡‡‡Physical examination should be performed on the basis of relevant medical history at the time of the visit according to the investigator's clinical judgment. Temperature needs to be measured before each vaccination and recorded in the source documents.

§§§Routine pediatric vaccines at V05: Prevnar 13®, M-M-R® II, and VARIVAX®; at V06: Pentacel®, HAVRIX®.

****AESIs will be collected throughout the trial as SAEs to ensure that the events are communicated to the Sponsor in an expedited manner and followed up until the end of the follow-up period or resolution, as per the assigned causality.

††††The memory aid is used only for the recording of SAEs (including AESIs) and MAAEs from Visit 7 to the 6-month follow up phone call at TC7.

Table 3.6: Study procedures for Subgroup 2a

Phase III trial, 8 Visits, 5 Vaccination Visits, 7 Telephone Calls, 4 Blood Samples, 19 to 22 Months Duration per Subject

Visit / Contact	Visit 1	Telephone contact (TC)1*	Visit 2	TC2*	Visit 3	TC3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6	Visit7§	TC6*	Visit 8	TC7** Follow-up Contact
Approximate age of subjects	**2 months (42 - 89 days)**	**--**	**4 months**	**--**	**6 months (164 – 224 days)**	**--**	**~7 months**		**12 months**	**--**	**13 months**	**15-18 months**	**--**	**16-19 months**	**21 - 24 months**
Trial timelines (days)	**Day 0**	**Visit 1 + 8 days**	**Visit 1+ 60 days**	**Visit 2+ 8 days**	**Visit 2 + 60 days**	**Visit 3 + 8 days**	**Visit 3 + 30 days**	**Visit 5 – 14 days**		**Visit 5+ 8 days**	**Visit 5+ 30 days**		**Visit 7 +8 days**	**Visit 7 + 30 days**	**Visit 7 + 180 days**
Time windows (days)	**--**	**+2 days**	**+14 days**	**+ 2 days**	**+14 days**	**+ 2 days**	**+21 days**			**+ 2 days**	**+21 days**		**+2 days**	**+21 days**	**+ 30 days**
Informed consent	X														
Inclusion / exclusion criteria	X														
Collection of demographic data	X														
Medical history (including maternal immunization history)	X														
Physical examination††	X								X						
Measurement of temperature‡‡	X		X		X				X			X			
Contact IRT system for randomization, subject number, and vaccine allocation.	X														

Visit / Contact	Visit 1	Telephone contact (TC)1*	Visit 2	TC2*	Visit 3	TC3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6	Visit7§	TC6*	Visit 8	TC7** Follow-up Contact
Contact IRT system for vaccine assignments			X		X				X			X			
Review of temporary contraindications for blood sampling§§	X						X		X		X				
Blood sampling*** (BL)	BL1††† (3 mL)						BL2 (6 mL)		BL3††† (5 mL)		BL4 (5 mL)				
Review of warning and precautions to vaccinations	X		X		X				X			X			
Review of contraindications to subsequent vaccinations and conditions for withdrawal ‡‡‡			X		X				X			X			
Vaccination with MenACYW conjugate vaccine or MENVEO®	**X**		**X**		**X**				**X**						
Vaccination with routine pediatric vaccines§§§	**X**		**X**		**X**				**X**			**X**			
Immediate surveillance (30 min)	X		X		X				X			X			
Diary card (DC) provided	DC1		DC2		DC3		DC4		DC5		DC6	DC7			
Telephone call		X		X		X		X		X			X		X

Visit / Contact	Visit 1	Telephone contact (TC)1*	Visit 2	TC2*	Visit 3	TC3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6	Visit7§	TC6*	Visit 8	TC7** Follow-up Contact
Diary card reviewed and collected			DC1		DC2		DC3		DC4		DC5	DC6		DC7	
Recording of solicited injection site and systemic AEs	X		X		X				X			X			
Recording of unsolicited AEs	Collected from D0 to D30 after each vaccination							Collected from D0 to D30 after each vaccination							
Reporting of serious adverse events (SAEs, including AESIs) and medically-attended AEs (MAAEs)****	To be reported throughout the study							To be reported throughout the study							
Collection of reportable concomitant medications	X		X		X		X		X		X	X		X	
Memory aid (MA) provided††††														MA	
Trial termination record (termination of active portion of the trial)														X	

IRT: interactive response technology
*This call is made 8 days after the respective vaccinations. If day 8 falls on a weekend or holiday, the telephone call may be made on the following business day. During this telephone call, the staff will find out whether the subject experienced any SAE (including AESI) and/or MAAE not yet reported and will remind the subject's parent/guardian to continue using the diary card, to bring the diary card to the study center at the next visit, and confirm the date and time of the next visit
†Staff will contact the subject's parent/ guardian by telephone within 14 days before V05 to remind them about the forthcoming study visit. If the subject's participation in the study is discontinued, the information recorded on the diary card will be reviewed at this time and the diary card will be retrieved by the site.
‡At V05, subjects must be 12 months of age (from the day subjects become 12 months old until the day before becoming 13 months old).
§At V07, Subjects must be between 15 and 18 months of age (from the day subjects become 15 months until the day before becoming 19 months of age).
**Staff will contact the subject's parent/ guardian by telephone at 6 months (+ 30 days) after the last vaccination visit to identify the occurrence of any SAEs (including any AESIs) and medically-attended AEs not yet reported. The final telephone call will continue until contact is made or 28 days have passed at which time the subject will be considered lost to follow-up.

††Physical examination should be done as per standard of care.
‡‡Temperature needs to be measured before each vaccination and recorded in the source documents.
§§Should a subject receive oral or injectable antibiotic therapy within 3 days prior to any blood draw, the investigator will postpone that blood draw until it has been 3 days since the subject last received oral or injectable antibiotic therapy. Postponement must still be within the timeframe for blood draw. If postponement would result in the sample collection falling outside of this timeframe, the blood sample should be collected without postponement, and it should be documented appropriately that the sample was taken less than 3 days after stopping antibiotic treatment.
***During the 2nd year of life study visits, if blood is collected as part of the study, additional blood can be optionally collected via the same venipuncture for screening of iron deficient anemia and/or lead level as per AAP/CDC recommendations. This collection must be done in serum tubes not provided as part of the study and as per local standard of care.
†††Blood sample will be drawn prior to vaccinations.
‡‡‡Physical examination should be performed on the basis of relevant medical history at the time of the visit according to the investigator's clinical judgment. Temperature needs to be measured before each vaccination and recorded in the source documents.
§§§ Routine pediatric vaccines at V05: Prevnar 13®, M-M-R® II, and VARIVAX®; at V07: Pentacel®, HAVRIX®
**** AESIs will be collected throughout the trial as SAEs to ensure that the events are communicated to the Sponsor in an expedited manner and followed up until the end of the follow-up period or resolution, as per the assigned causality.
††††The memory aid is used only for the recording of SAEs (including AESIs) and MAAEs from V08 to the 6-month follow up phone call at TC7.

Table 3.7: Study procedures for Subgroup 2b

Phase III trial, 7 Visits, 5 Vaccination Visits, 7 Telephone Calls, 4 Blood Samples, 19 to 22 Months Duration per Subject

Visit	Visit 1	Telephone contact (TC)1*	Visit 2	TC2*	Visit 3	TC3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6§	TC6*	Visit 7	TC7** Follow-up Contact
Approximate age of subjects	**2 months (42 - 89 days)**	--	**4 months**	--	**6 months (164 – 224 days)**	--	**~7 months**		**12 months**	--	**15-18 months**	--	**16-19 months**	**21 - 24 months**
Trial timelines (days)	**Day 0**	**Visit 1 + 8 days**	**Visit 1+ 60 days**	**Visit 2+ 8 days**	**Visit 2 + 60 days**	**Visit 3 +8 days**	**Visit 3 +30 days**	**Visit 5 – 14 days**		**Visit 5 +8 days**		**Visit 6 +8 days**	**Visit 6 +30 days**	**Visit 6 + 180 days**
Time windows (days)	**–**	**+2 days**	**+14 days**	**+ 2 days**	**+14 days**	**+ 2 days**	**+21 days**			**+ 2 days**		**+2 days**	**+21 days**	**+ 30 days**
Informed consent	X													
Inclusion / exclusion criteria	X													
Collection of demographic data	X													
Medical history (including maternal immunization history)	X													
Physical examination††	X								X					
Temperature measurement ‡‡	X		X		X				X		X			
Contact IRT system for randomization, subject number, and vaccine allocation.	X													
Contact IRT system for vaccine assignments			X		X				X		X			
Review of temporary contraindications for blood sampling§§	X						X				X		X	

Visit	Visit 1	Telephone contact (TC)1*	Visit 2	TC2*	Visit 3	TC3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6§	TC6*	Visit 7	TC7** Follow-up Contact
Blood sampling (BL)***	BL1††† (3 mL)						BL2 (6 mL)				BL3††† (5 mL)		BL4 (5 mL)	
Review of warning and precautions to vaccinations	X		X		X				X§§§		X			
Review of contraindications to subsequent vaccinations and conditions for withdrawal‡‡‡			X		X				X		X			
Vaccination with MenACYW conjugate vaccine or MENVEO®	**X**		**X**		**X**				**X**					
Vaccination with routine pediatric vaccines§§§	**X**		**X**		**X**				**X**		**X**			
Immediate surveillance (30 minutes)	X		X		X				X		X			
Diary card (DC) provided	DC1		DC2		DC3		DC4		DC5		DC6			
Telephone call		X		X		X		X		X		X		X
Diary card reviewed and collected			DC1		DC2		DC3		DC4		DC5		DC6	
Recording of solicited injection site and systemic AEs	X		X		X				X		X			
Recording of unsolicited AEs	Collected from D0 to D30 after each vaccination							Collected from D0 to D30 after each vaccination						
Reporting of serious adverse events (SAEs, including AESIs) and medically-attended AEs (MAAEs)****	To be reported throughout the study							To be reported throughout the study						

Visit	Visit 1	Telephone contact (TC)1*	Visit 2	TC2*	Visit 3	TC3*	Visit 4	TC4†	Visit 5‡	TC5*	Visit 6§	TC6*	Visit 7	TC7** Follow-up Contact
Collection of reportable concomitant medications	X		X		X		X		X		X		X	
Memory aid (MA) provided††††													MA	
Trial termination record (termination of active portion of the trial)													X	

IRT: interactive response technology

*This call is made 8 days after the respective vaccinations. If day 8 falls on a weekend or holiday, the telephone call may be made on the following business day. During this telephone call, the staff will find out whether the subject experienced any SAE (including AESI) and/or MAAE not yet reported and will remind the subject's parent/guardian to continue using the diary card, to bring the diary card to the study center at the next visit, and confirm the date and time of the next visit

†Staff will contact the subject's parent/ guardian by telephone within 14 days before V05 to remind them about the forthcoming study visit. If the subject's participation in the study is discontinued, the information recorded on the diary card will be reviewed at this time and the diary card will be retrieved by the site.

‡At V05 subjects must be 12 months of age (from the day subjects become 12 months of age until the day before becoming 13 months old).

§At V06 subjects must be between 15 and 18 months of age (from the day subjects become 15 months old until the day before becoming19 months old).

**Staff will contact the subject's parent/ guardian by telephone at 6 months (+ 30 days) after the last vaccination visit to identify the occurrence of any SAEs (including any AESIs) and medically-attended AEs not yet reported. The final telephone call will continue until contact is made or 28 days have passed at which time the subject will be considered lost to follow-up.

††Physical examination should be done as per standard of care.

‡‡Temperature needs to be measured before each vaccination and recorded in the source documents.

§§Should a subject receive oral or injectable antibiotic therapy within 3 days prior to the second blood draw, the investigator will postpone that blood draw until it has been 3 days since the subject last received oral or injectable antibiotic therapy. Postponement must still be within the timeframe for blood draw. If postponement would result in the sample collection falling outside of this timeframe, the blood sample should be collected without postponement, and it should be documented appropriately that the sample was taken less than 3 days after stopping antibiotic treatment

***During the 2nd year of life study visits, if blood is collected as part of the study, additional blood can be optionally collected via the same venipuncture for screening of iron deficient anemia and/or lead level as per AAP/CDC recommendations. This collection must be done in serum tubes not provided as part of the study and as per local standard of care.

†††Blood sample will be drawn prior to vaccinations.

‡‡‡Physical examination should be performed on the basis of relevant medical history at the time of the visit according to the investigator's clinical judgment. Temperature needs to be measured before each vaccination and recorded in the source documents

§§§Routine pediatric vaccines at V05: Prevnar 13®, M-MR® II, and VARIVAX®; at V06: Pentacel®, HAVRIX®

****AESIs will be collected throughout the trial as SAEs to ensure that the events are communicated to the Sponsor in an expedited manner and followed up until the end of the follow-up period or resolution, as per the assigned causality.

††††The MA is used only for the recording of SAEs (including AESIs) and MAAEs from V07 to the 6-month follow up phone call at TC7.

4 Endpoints and Assessment Methods

4.1 Primary Endpoints and Assessment Methods

4.1.1 Immunogenicity

4.1.1.1 Immunogenicity Endpoints

The primary endpoints for the evaluation of immunogenicity are:

1) Meningococcal serogroups A, C, Y, and W antibody titers measured by hSBA before first study vaccination on D0 and 30 days after the 4th meningococcal vaccination (Subgroup 1a vs Subgroup 2a).
2) Antibody titers ≥ 1:8 against meningococcal serogroups A, C, Y, and W measured by hSBA assessed 30 days after vaccination(s) at 6 months of age (Group 1 vs Group 2).

4.1.1.2 Immunogenicity Assessment Methods

All assays will be performed at Global Clinical Immunology (GCI), Swiftwater, Pennsylvania (PA) or at a qualified contract laboratory for GCI.
The assay method to be used is summarized below. Laboratory technicians conducting the immunogenicity assays will be blinded to the group to which each subject was assigned.

Antibodies to meningococcal antigens (hSBA Method)
Functional meningococcal antibody activity against serogroups A, C, Y, and W will be measured in hSBA. Two-fold dilutions of test sera are prepared in sterile 96-well microtiter plates. Serogroup-specific meningococcal bacteria along with human complement are added to the serum dilutions and allowed to incubate. After this incubation period, an agar overlay medium is added to the serum/complement/bacteria mixture, allowed to harden, and then incubated overnight at 37°C with 5% carbon dioxide (CO_2). Bacterial colonies present in the wells are then counted. The endpoint titer is determined by the reciprocal serum dilution yielding ≥ 50% killing as compared to the mean of the complement control wells. The lower limit of quantitation (LLOQ) of the hSBA assay is a titer of 1:4.
This method will be performed on blood sampling 1 (BL1) and BL2 collected from all groups, BL3 collected from Subgroups 1a, 1b and 2a, and BL4 collected from Subgroups 1a, 1b and 2a.

4.2 Secondary Endpoints and Assessment Methods

4.2.1 Immunogenicity

4.2.1.1 Immunogenicity Endpoints

1) The following serological endpoints will be assessed (assessment of routine vaccines):

- D0 (before first vaccination) for Group 1 and Group 2:
  - Anti-rotavirus serum immunoglobulin (Ig) A antibody concentrations
- 30 days after the 6-month vaccination (after the 3rd dose) for Group 1 and Group 2:

  - IgG antibodies against hepatitis B surface antigen (anti-HB) concentrations ≥ 10 milli-international units (mIU)/mL
  - Anti polyribosyl-ribitol phosphate (PRP) antibody concentrations ≥ 0.15 micrograms/milliliter (µg/mL)
  - Anti PRP antibody concentrations ≥ 1.0 µg/mL
  - Anti-poliovirus types (1, 2, and 3) antibody titers ≥ 1:8
  - Anti-rotavirus serum IgA antibody concentrations with ≥ 3-fold rise over baseline
  - Anti-rotavirus serum IgA antibody geometric mean concentrations (GMCs)
  - Anti-pertussis antibody concentrations (PT, FHA, PRN, and FIM) (GMCs)
  - Anti-pneumococcal antibody concentrations (for serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F) (GMCs)
- 30 days after the 12-month vaccinations for Subgroup 1a and Subgroup 2a:
  - Anti-measles antibody concentrations ≥ 255 mIU/mL
  - Anti-mumps antibody concentrations ≥ 10 mumps antibody (Ab) units/mL
  - Anti-rubella antibody concentrations ≥ 10 IU/mL
  - Anti-varicella antibody concentrations ≥ 5 glycoprotein enzyme-linked immunosorbent assay (gpELISA) units/mL
  - Anti-pneumococcal antibody concentrations (for serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F) (GMCs)
- Before the 15-month vaccination for Subgroup 1b and Subgroup 2b:
  - Anti-pertussis antibody concentrations (PT, FHA, PRN, and FIM).
- 30 days after the 15-month vaccinations for Subgroup 1b and Subgroup 2b:
  - Anti-PRP antibody concentrations ≥ 1.0 µg/mL
  - Anti-poliovirus types 1, 2, and 3 antibody titers ≥ 1:8
  - Anti-pertussis antibody concentrations (PT, FHA, PRN, and FIM) (vaccine response)[a]

2) The following serological endpoints will be assessed (effect of 4th dose of MenACYW or Menveo):

- Before the 12-month vaccination (pre-4th dose) for Subgroups 1a and 2a
  - hSBA meningococcal serogroups A, C, Y and W antibody titers
- 30 days after the 12-month vaccination for Subgroups 1a and 2a
  - hSBA meningococcal serogroups A, C, Y and W antibody titers
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers≥ 4-fold rise from pre-4th dose (at 12 months of age) to post-dose 4 vaccination

3) The following serological endpoints will be assessed (persistence of bactericidal antibodies after infant vaccination with MenACYW or Menveo):

[a] Pertussis vaccine response definition:

- Pre-vaccination < LLOQ, then post-vaccination should be ≥ 4x the LLOQ
- Pre- vaccination ≥ LLOQ but < 4x the LLOQ, then post-vaccination should achieve a 4-fold rise (post-vaccination/ Pre-vaccination ≥ 4)
- Pre-vaccination≥ 4x the LLOQ, then post-vaccination should achieve a 2-fold response (post-vaccination/ Pre-vaccination ≥ 2)

- 30 days after the 6-month vaccination and before the 12-month vaccination for Subgroup 1a and Subgroup 2a
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥1:4 and ≥1:8

4) The following serological endpoints will be assessed:

- Day 0 (before first vaccination) for Groups 1 and 2
  - Anti-pertussis antibody concentrations (PT, FHA, PRN, FIM)
- 30 days after the 6-month vaccination (after the 3rd dose) for Group 1 and Group 2:
  - Anti-PRP antibody concentrations
  - Anti-diphtheria antibody concentrations
  - Anti-diphtheria antibody concentrations ≥ 0.01 IU/mL
  - Anti-diphtheria antibody concentrations ≥ 0.1 IU/mL
  - Anti-tetanus antibody concentrations
  - Anti-tetanus antibody concentrations ≥ 0.01 IU/mL
  - Anti-tetanus antibody concentrations ≥ 0.1 IU/mL
  - Anti-HBs antibody concentrations
  - Anti-HBs concentrations ≥ 100 mIU/mL
  - Anti-polio (types 1, 2, and 3) antibody titers ≥1:8
  - Anti-rotavirus serum IgA antibody concentrations
  - Anti-rotavirus serum IgA antibody concentrations with ≥4-fold rise over baseline
  - Anti-pertussis (PT, FHA, PRN, and FIM) antibody concentrations (vaccine response)
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 0.35 µg/mL
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 1 µg/mL
- 30 days after the 12-month vaccinations for Subgroup 1a and Subgroup 2a:
  - Anti-measles antibody concentrations
  - Anti-mumps antibody concentrations
  - Anti-rubella antibody concentrations
  - Anti-varicella antibody concentrations
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 0.35 µg/mL
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 1 µg/mL
- at the 15-month vaccinations for Subgroup 1b and Subgroup 2b to evaluate immune persistence after primary series vaccination with Hib and pertussis vaccines:
  - Anti-PRP antibody concentration ≥ 0.15 µg/mL
  - Anti-PRP antibody concentrations
  - Anti-pertussis (PT, FHA, PRN, and FIM) antibody concentrations
- 30 days after the 15-month vaccinations for Subgroup 1b and Subgroup 2b:
  - Anti-PRP antibody concentrations
  - Anti-diphtheria antibody concentrations
  - Anti-diphtheria antibody concentrations ≥ 0.1 IU/mL
  - Anti-diphtheria antibody concentrations ≥ 1.0 IU/mL
  - Anti-tetanus antibody concentrations
  - Anti-tetanus antibody concentrations ≥ 0.1 IU/mL

- Anti-tetanus antibody concentrations ≥ 1.0 IU/mL
- Anti-polio (types 1, 2, and 3) antibody titers
- Anti-pertussis antibody concentrations (PT, FHA, PRN, and FIM) (GMCs)

5) The following serological endpoints will be assessed:

- D0 (before first vaccination) for Group 1 and Group 2:
  - hSBA meningococcal serogroups A, C, Y and W antibody titers
- 30 days after the 6-month vaccination (after the 3rd dose) for Group 1 and Group 2:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - Titer distribution and reverse cumulative distribution curves (RCDCs)
  - hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 1:4 and ≥ 1:8
  - hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 4-fold rise from pre-vaccination (D0) to post-vaccination
  - hSBA vaccine seroresponse

- Before the 12-month vaccination for Subgroups 1a and 2a and before the 15-month vaccination for Subgroup 1b:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers

- 30 days after the 12-month vaccinations for Subgroup 1a and 2a and 30 days after the 15-month vaccination for Subgroup 1b:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - Titer distribution and RCD
  - hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 1:4 and ≥ 1:8
  - hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 4-fold rise from pre-vaccination (D0) to post-dose 4 vaccination
  - hSBA vaccine seroresponse

6) The following serological endpoints will be assessed:

- D0 (before first vaccination) for Subgroup 1a and Subgroup 1b:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
- 30 days after the 6-month vaccination and before the 12-month vaccination for Subgroup 1a and before the 15-month vaccination for Subgroup 1b to evaluate the immune persistence after infant vaccination with MenACYW conjugate vaccine:
  - hSBA meningococcal serogroups A, C, Y and W antibody titers
  - hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 1:4 and ≥ 1:8
- 30 days after the 12-month vaccinations for Subgroup 1a and 30 days after the 15-month vaccination for Subgroup 1b, including evaluation of the effect of the 4th dose of MenACYW conjugate vaccine:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - hSBA meningococcal serogroups A, C, Y and W antibody titers ratio (Subgroup 1b/Subgroup 1a)
  - hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 1:4 and ≥ 1:8

- hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 1:8 difference (Subgroup 1b – Subgroup 1a)
- hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 4-fold rise from pre-vaccination (D0) to post-4th dose vaccination
- hSBA meningococcal serogroups A, C, Y and W antibody titers ≥ 4-fold rise from pre-4th dose vaccination to post-4th dose vaccination
- hSBA vaccine seroresponse
- hSBA vaccine seroresponse difference (Subgroup 1b –Subgroup 1a)

4.2.1.2 Immunogenicity Assessment Methods

The assay methods to be used are summarized below. Laboratory technicians conducting the immunogenicity assays will be blinded to the group to which each subject was assigned.
The immunogenicity hSBA assessment method for meningococcal serogroups A, C, Y, and W antibody titers for the secondary endpoints is the same as that presented in Section 4.1.1.2.

Anti-Rotavirus IgA Antibodies

Anti-rotavirus IgA antibodies in human serum will be measured by ELISA. Microtiter plates are coated with rabbit anti-rotavirus antibody and then viral lysate (positive wells) or control cell lysate (negative wells) is added. Diluted serum samples (test samples, reference standard, and quality controls) are incubated in the wells. Unbound antibodies are washed from the wells, and enzyme-conjugated anti-human IgA immunoglobulin is added. The enzyme conjugate binds to the antigen-antibody complex. Excess conjugate is washed away, and a specific colorimetric substrate is added. Bound enzyme catalyzes a hydrolytic reaction, which causes color development. A reference standard assayed on each plate is used to calculate the amount of specific anti-rotavirus IgA antibody in the units assigned by the reference standard (U/mL of serum).

This method will be performed on BL1 and BL2 collected from all groups.

Anti-Diphtheria, Tetanus, and Pertussis Antibodies

The DTaP (Diphtheria, Tetanus, and Pertussis) ECL (electrochemiluminescent) is a multiplexed serological assay which allows for the simultaneous quantification of human antibodies to 6 specific antigens including diphtheria toxoid, tetanus toxoid, and 4 pertussis antigens: PT, FHA, FIM and PRN. In this assay, each well of a 96-well microtiter plate is pre-coated in precise positions with the 6 different antigens in a multi-spot fashion. Following incubation with serum samples, antigen-specific antibodies bind to the respective antigens. The captured antibodies are then detected using a sulfotag conjugated anti-human IgG conjugate. Electrical stimulation of the conjugate in the presence of a chemiluminescent substrate results in the generation of a light signal from each specific spot that is captured by a camera in relative light units. The signal generated is directly proportional to the amount of antibodies present in the sample, which is quantified using software and based on an established reference standard sample curve. The LLOQ for Diphtheria is 0.005 IU/mL, the LLOQ for Tetanus is 0.01 IU/mL and the LLOQ for Pertussis antigens is 2.00 EU/mL.

This method will be performed on BL1 and BL2 collected from all groups, and BL3 and BL4 collected from Subgroups 1b and 2b.

Anti-Hepatitis B Antibodies

Anti-HB surface antibodies (anti-HBs) will be measured by the commercially available VITROS ECi/ECiQ Immunodiagnostic System using chemiluminescence detection technology. The VITROS ECi Immunodiagnostic system uses an antibody-mediated antigen sandwich formation to detect the presence of anti-HBs total immunoglobulin in human serum. This involves the reaction of anti-HBs in the sample with plasma-derived HBsAg (ad and ay subtypes) coated onto the wells. A horseradish peroxidase (HRP)-labeled HBsAg conjugate (ad and ay subtypes) then complexes with the bound anti-HBs forming an antigen sandwich. Substrate is then added which catalyzes HRP, producing light. The light signals are read by the VITROS ECi/ECiQ. Immunodiagnostic System and the amount of HRP conjugate bound is directly proportional to the concentration of anti-HBs present in the sample. Results are reported in mIU/mL by comparison to a calibrator provided by the manufacturer that has been calibrated according to the World Health Organization (WHO) First International Reference Preparation for Antibody to HBsAg (1977). The LLOQ is 5 mIU/mL.

This method will be performed on BL2 collected from all groups.

Anti-*Haemophilus influenzae* type b *(Anti-PRP) Antibodies*

Anti-PRP concentrations will be measured using a Farr-type radioimmunoassay (RIA). Serum levels of anti-*Haemophilus influenzae* type b (Hib) capsular PRP antibody are determined by RIA, in which serum samples are incubated with radiolabeled PRP (^{3}H-PRP) in the presence of ^{36}Cl (volume marker). Specific antibodies bind to tritiated capsular PS to form antigen-antibody complexes. These complexes are precipitated with ammonium sulfate and collected by centrifugation. The radioactivity is measured in the precipitated pellet in counts per minute and is proportional to the amount of anti-Hib capsular PS antibody present in the serum sample. The concentration of anti-PRP antibody in the serum sample is determined from the concentration response curve generated by the titration results of dilutions of the reference standard analyzed in the assay. Results are reported in µg/mL by comparison to the Center for Biologics Evaluation and Research (CBER), Lot No. 1983 reference standard. The LLOQ of the anti-PRP RIA is 0.06 µg/mL.

This method will be performed on BL2 collected from all groups, and BL3 and BL4 collected from Subgroups 1b and 2b, only.

Anti-Polio (types 1, 2, and 3) Antibodies

Anti-poliovirus types 1, 2, and 3 will be measured by neutralization assay. Serial dilutions of sera are mixed with challenge poliovirus and incubated with cultured Vero cells that are sensitive to poliovirus. Specific neutralizing antibodies contained in the sera bind to and neutralize the challenge poliovirus. The neutralized poliovirus does not affect cellular viability and these cells

continue to metabolize and release CO_2, reducing the pH of the culture medium. Cell survival correlates with the change in the pH indicator (phenol red to yellow at pH ≤ 7.0) contained in the medium. In the absence of neutralizing antibodies, the challenge poliovirus reduces cellular metabolism and CO_2 production. Therefore, the pH does not decrease and a color change is not detected. The poliovirus mouse inoculation test measures the functional serum antibody response to poliovirus by utilizing Vero cells (African green monkey kidney cells) and wild type poliovirus strains 1, 2, and 3 (Mahoney, MEF-1, and Saukett, respectively) as the challenge virus. The Karber method is used to determine the serum dilution that neutralized 50% of the challenge virus. Results are expressed as titers (1/dilution [dil]). The LLOQ of the anti-poliovirus types 1, 2, and 3 assays is 4 (1/dil). The ULOQ of the anti-poliovirus types 1, 2, and 3 assays is 65536 (1/dil).

This method will be performed on BL2 collected from all groups and BL4 collected from Subgroups 1b and 2b.

Anti-Pneumococcal Antibodies

The pneumococcal capsular PS (PnPS) IgG ECL assay is used to quantitate the amount of anti-*Streptococcus pneumoniae* PS (serotypes 1, 3, 4, 5, 6A, 6B, 7F, 8, 9N, 9V, 10A, 11A, 12F, 14, 15B, 18C, 19A, 19F, 22F, 23F and 33F) antibodies in human serum. In this method, purified antigen of 8 PnPS are coated into defined spots within the wells of a 96-well microtiter plate by MesoScale Discovery using 3 types of plates to cover all 21 PnPS. Diluted serum samples (test samples, reference standard, and quality controls), pre-treated with pneumococcal cell wall absorbents (to reduce the interference of non-specific antibodies in the assay), are incubated in the wells. Specific antibodies in the serum samples bind to the immobilized antigen. Unbound antibodies are washed from the wells, and SULFO-TAG-conjugated anti-human immunoglobulin is added. The antibody conjugate binds to the antigen-antibody complex. Excess conjugate is washed away, and read buffer is added. The plate is read using electrochemiluminescence on an MSD imager. The intensity of the generated light is proportional to the amount of specific antibody bound to the antigen-coated spots. An international reference standard assayed on each plate is used to calculate the amount of anti-pneumococcal IgG antibodies (µg/mL) in human serum. The LLOQ for all PnPS serotypes is 0.15 µg/mL.

This method will be performed on BL2 for all groups and BL4 collected from Subgroups 1a and 2a.

Anti-Measles Antibodies

The purpose of the Bulk Measles IgG EIA (Enzyme Immunoassay) is to detect total IgG antibody to measles virus before and after vaccination with a measles-containing vaccine. Plates are coated in house using inactivated measles antigen that is bound to solid phase microtiter plates. The antigen is derived from Measles Edmonston strain-infected Vero cells. Serum or plasma is added to the coated plates and samples positive for measles antibodies will bind to the measles antigen-coated plates, forming antibody-antigen complexes. The bound antibody-antigen complexes can then be detected using an Alkaline Phosphatase labeled anti-human IgG. Color development occurs as a result of the addition of an enzyme-specific substrate Phenolphthalein

Monophosphate. The color intensity is then measured spectrophotometrically with the highest intensity of color correlating to a high level of measles antibody and lowest color intensity correlating to low levels of measles antibody. Quantitation of the human IgG antibody to measles virus or titer is determined by comparison of the resulting optical density (OD) to a standard curve. The reference standard is a pool of human sera that has been calibrated against the WHO anti-measles reference standard, lot NIBSC 66/202. The concentration of anti-measles antibody in a sample is reported in milli-International Units per milliliter of serum (mIU/mL). The clinical endpoint for the measles assay is 255 mIU/mL. The LLOQ is 60 mIU/mL and the ULOQ is 7680 mIU/mL.

This method will be performed on BL4 collected from Subgroups 1a and 2a.

Anti-Mumps Antibodies

The purpose of the mumps enzyme-linked immunosorbent assay (ELISA) is to detect IgG antibody to mumps virus before and after vaccination with a mumps virus-containing vaccine. The assay uses an earlier passage of the Jeryl Lynn® mumps virus (Jeryl Lynn® 135 [JL135],<12 passages) which is considered to be a wild-type (WT)-like strain. The reactivity of the sera to the mumps antigens prepared from uninfected Vero cells (denoted as tissue culture control [TCC] wells) is subtracted from that of JL135-infected Vero cells. JL135 mumps virus antigen or TCC is bound to solid phase microtiter plates and serum containing mumps antibody is added. The mumps antibody bound to the WT mumps antigen-coated plates forms an antibody-antigen complex. The bound antibody-antigen complex is then detected using an enzyme-labeled anti-human IgG. Color development occurs with the addition of a substrate and color intensity is measured spectrophotometrically. Results are obtained as a difference of the average duplicate of each optical density (OD) of JL135 mumps antigen wells and the average duplicate OD of TCC wells for each serum sample (noted as delta optical density [DOD]). Quantitation of the human IgG antibody to mumps virus, or antibody concentration, is determined by comparison of the resulting test DOD to a standard curve. The reference standard is an individual human serum. Results for the assay are reported as the concentration of antibody in Mumps antibody units/mL. The clinical endpoint and the LLOQ for the mumps assay is 10 Mumps Ab units/mL and the ULOQ for the mumps assay is 640 Mumps Ab units/mL.

This method will be performed on BL4 collected from Subgroups 1a and 2a.

Anti-Rubella Antibodies

The purpose of the Bulk Rubella IgG EIA (Enzyme Immunoassay) is to detect total IgG antibody to rubella virus before and after vaccination with a rubella-containing vaccine. Plates are coated in house using inactivated rubella antigen that is bound to solid phase microtiter plates. The antigen is derived from Rubella HPV-77 infected Vero cells. Serum is added to the coated plates and samples positive for rubella antibodies will bind to the rubella antigen-coated plates, forming antibody-antigen complexes. The bound antibody-antigen complexes can then be detected using an Alkaline Phosphatase labeled anti-human IgG. Color development occurs as a result of the addition of an enzyme-specific substrate, Phenolphthalein Monophosphate. The color intensity is then measured spectrophotometrically with the highest intensity of color correlating to a high level of rubella antibody and lowest color intensity correlating to low levels of rubella antibody.

Quantitation of the human IgG antibody to rubella virus or titer is determined by comparison of the resulting analysis OD to a standard curve. The reference standard is an individual human

serum that has been calibrated against the WHO anti-rubella reference standard. The concentration of anti-rubella antibody in a sample is reported in International Units per milliliter of serum (IU/mL). The clinical endpoint for the rubella assay is 10 IU/mL. The LLOQ is 5 IU/mL and the ULOQ is 320 IU/mL.

This method will be performed on BL4 collected from Subgroups 1a and 2a.

Anti-Varicella Antibodies

The purpose of the glycoprotein enzyme-linked immunosorbent assay (gpELISA) is to detect IgG antibody to varicella-zoster virus (VZV) before and after vaccination with VZV-containing vaccine(s). This method detects antibodies to VZV glycoprotein (gp), which have been purified from MRC-5 cells infected with the KMcC strain of VZV by lectin affinity chromatography. The assay uses the "second antibody" format with varicella glycoprotein (gp) antigen and MRC5 TCC glycoprotein coated on the solid phase microtiter plate. Diluted sera are dispensed into two VZV gp antigen coated wells and two MRC5 gp coated wells for each standard curve point, control, and sample. Antibody to the Varicella virus in a test sample binds to the antigen coated plate. Antibody to varicella glycoprotein in a test sample, bound to the antigen on the solid phase microtiter plate is subsequently detected using goat anti-human IgG alkaline phosphatase conjugate. After substrate addition for color development, quantitation is obtained by comparison of sample DOD to a standard curve. The DOD is determined by subtracting the average OD of the TCC coated wells from its corresponding VZV gp average OD with a standard curve. Assay results are reported as concentration of antibody in gpELISA units/mL. The clinical endpoint for the varicella assay is 5 gpELISA Ab units/mL and the LLOQ is 0.625 gpELISA Ab units/mL. This method will be performed on BL4 collected from Subgroups 1a and 2a.

4.3 Observational Endpoints and Assessment Methods

4.3.1 Immunogenicity

There are no observational objectives for immunogenicity in this study.

4.3.2 Safety

4.3.2.1 Safety Definitions

The following definitions are taken from the ICH E2A Guideline for Clinical Safety Data Management: Definitions and Standards for Expedited Reporting.

Adverse Event (AE):

An AE is any untoward medical occurrence in a patient or in a clinical investigation subject administered a medicinal product and which does not necessarily have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

Therefore, an AE may be:

- A new illness
- The worsening of a pre-existing condition
- An effect of the vaccination, including the comparator
- A combination of the above

All AEs include serious and non-serious AEs.

Surgical procedures are not AEs; they are the actions taken to treat a medical condition. It is the condition leading to the action taken that is the AE (if it occurs during the study period).

Pre-existing medical conditions are not to be reported as AEs. However, if a pre-existing medical condition worsens following study interventions in frequency or intensity, or if according to the Investigator there is a change in its clinical significance, this change should be reported as an AE (exacerbation). This applies equally to recurring episodes of pre-existing conditions (e.g., asthma) if the frequency or intensity increases post-vaccination.

Serious Adverse Event (SAE):

Serious and *severe* are not synonymous. The term *severe* is often used to describe the intensity of a specific event as corresponding to Grade 3. This is not the same as *serious* which is based on subject / event outcome or action criteria usually associated with events that pose a threat to a subject's life or functioning. Seriousness, not severity, serves as a guide for defining regulatory reporting obligations.

An SAE is any untoward medical occurrence that at any dose

- Results in death
- Is life-threatening[a]
- Requires inpatient hospitalization or prolongation of existing hospitalization[b]
- Results in persistent or significant disability / incapacity[c]
- Is a congenital anomaly / birth defect
- Is an important medical event (IME)

Medical and scientific judgment should be exercised in deciding whether expedited reporting is appropriate in other situations, such as IMEs that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the health of the subject or may require intervention to prevent one of the other outcomes listed in the definition above. These IMEs should also usually be considered serious. Examples of such events include allergic bronchospasm

[a] The term "life-threatening" refers to an event in which the subject was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe.

[b] All medical events leading to hospitalizations will be recorded and reported as SAEs, with the exception of: hospitalization planned before inclusion into the study or outpatient treatment with no hospitalization.

[c] "Persistent or significant disability or incapacity" means that there is a substantial disruption of a person's ability to carry out normal life functions.

requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse, new-onset diabetes, or autoimmune disease.

Adverse Reaction:

All noxious and unintended responses to a medicinal product related to any dose should be considered adverse reactions (ARs).

(The phrase "responses to a medicinal product" means that a causal relationship between a medicinal product and an AE is at least a reasonable possibility)

The following additional definitions are used by Sanofi Pasteur:

Immediate Event/Reaction:

Immediate events are recorded to capture medically relevant unsolicited systemic AEs (including those related to the product administered) that occur within the first 30 minutes after vaccination.

Solicited Reaction:

A solicited reaction is an "expected" AR (sign or symptom) observed and reported under the conditions (nature and onset) pre-listed in the protocol and CRB.
Examples of solicited reactions include injection site tenderness or irritability occurring between D0 and D07 after vaccination.

By definition, solicited reactions are to be considered as being related to the product administered. The assessment of these reactions by the Investigator is mandatory. For injectable vaccines, solicited reactions can either be solicited injection site reactions or solicited systemic reactions.

Unsolicited AE / AR:

An unsolicited AE is an observed AE that does not fulfill the conditions prelisted in the CRB in terms of diagnosis and/or onset window post-vaccination. For example, if headache between D0 and D07 is a solicited reaction (i.e., pre-listed in the protocol and CRB), then a headache starting on D07 is a solicited reaction, whereas headache starting on D08 post-vaccination is an unsolicited AE. Unsolicited AEs includes both serious (SAEs) and non-serious unsolicited AEs.

Injection Site Reaction:

An injection site reaction is an AR at and around the injection site. Injection site reactions are commonly inflammatory reactions. They are considered to be related to the product administered.

Systemic AE:

Systemic AEs are all AEs that are not injection or administration site reactions. They therefore include systemic manifestations such as headache, fever, as well as localized or topical manifestations that are not associated with the vaccination or administration site (e.g., erythema that is localized but that is not occurring at the injection site).

Adverse Event of Special Interest (AESI):

AESI is an event for which ongoing monitoring and rapid communication by the Investigator to the Sponsor must be done. Such an event might warrant further investigation in order to characterize and understand it. Depending on the nature of the event, rapid communication by the study Sponsor to other parties (e.g., regulators) might also be warranted.

Medically-Attended Adverse Event (MAAE)

An MAAE is defined, for the purpose of this study, as a new onset of a condition that prompts the subject or subject's parent/guardian to seek unplanned medical advice at a health care provider's office or Emergency Department. This definition excludes pre-planned medical office visits for routine pediatric check-ups or follow-up visits of chronic conditions with an onset prior to entry in the study. Health care provider contact made over the phone or by email will be considered a physician office visit for the purpose of MAAE collection. The outcome of the health care provider contact (whether it results in a prescription or not) will not be considered as a basis for reporting the event as an MAAE and all contacts should be reported. Sufficient data should be collected for the event to allow an assessment of the causality and diagnosis, if possible.

4.3.2.2 Safety Endpoints

The endpoints for the evaluation of safety are:

- Occurrence, nature (Medical Dictionary for Regulatory Activities [MedDRA] preferred term), duration, intensity, relationship to vaccination, and whether the event led to early termination from the study, of any unsolicited systemic AEs reported in the 30 minutes after each vaccination
- Occurrence, time of onset, number of days of occurrence, intensity, action taken, and whether the reaction led to early termination from the study, of solicited (prelisted in the subject's diary card and electronic case report book [CRB]) injection site reactions occurring up to D07 after each vaccination
- Occurrence, time of onset, number of days of occurrence, intensity, action taken, and whether the reaction led to early termination from the study, of solicited (prelisted in the subject's diary card and book CRB) systemic reactions occurring up to D07 after each vaccination
- Occurrence, nature (MedDRA preferred term), time of onset, duration, intensity, action taken, relationship to vaccination (for systemic AEs only), and whether the event led to early termination from the study, of unsolicited AEs up to D30 after each vaccination
- Occurrence, nature (MedDRA preferred term), time of onset, duration, seriousness criteria, relationship to vaccination, outcome, and whether the event led to early termination from the

study, of SAEs (including AESIs) throughout the trial from Visit 1 to the 6-month follow-up contact after the last vaccination
- Occurrence, nature (MedDRA preferred term), time of onset, duration, intensity, action taken, relationship to vaccination (for systemic AEs only), and whether the event led to early termination from the study for MAAEs throughout the trial from Visit 1 to the 6-month follow-up contact after the last vaccination.

4.3.2.3 Safety Assessment Methods

At each vaccination visit, the Investigator or a delegate will perform a physical examination on the basis of relevant medical history according to the Investigator's clinical judgment and will ask the parent / guardian about any solicited reactions and unsolicited AEs recorded in the diary card, as well as about any other AEs that may have occurred since the previous visit. All relevant data will be transcribed into the CRB according to the instructions provided by the Sponsor.

4.3.2.3.1 Immediate Post-vaccination Observation Period

Subjects will be kept under observation for 30 minutes after each vaccination to ensure their safety. The post-vaccination observation should be documented in the source document. Any AE that occurs during this period will be noted on the source document and recorded in the CRB, as follows:

- Unsolicited systemic AEs will be recorded as immediate AEs in the CRB (presence marked as "yes" and details collected).
- Solicited and unsolicited injection site reactions and solicited systemic reactions will be recorded in the CRB in the same way as any reactions starting on the day of vaccination.
- SAEs will be recorded in the CRB and reported to the Sponsor in the same way as any other SAEs, according to the procedures described in Section 10 of the protocol.

4.3.2.3.2 Reactogenicity (Solicited Reactions from Day 0 to Day 7 After Each Vaccination)

After the first vaccination, parents / guardians will be provided with a diary card, a digital thermometer, and a flexible ruler, and will be instructed how to use them. The following items will be recorded by the subject's parent / guardian in the diary card on the day of each vaccination and for the next 7 days (i.e., D0 to D07) until resolution:

- Daily temperature, with the route by which it was taken
- Daily measurement or intensity grade of all other solicited injection site and systemic reactions
- Action taken for each event (e.g., medication)

The action(s) taken by the parent or guardian to treat and/or manage any solicited reactions will be classified in the CRB using the following list (all applicable items should be checked):

- None
- Medication
- Health care provider contact

- Hospitalized
- Discontinuation of study vaccination

Parents / guardians will be contacted by telephone 8 days after each vaccination to remind them to record all safety information in the diary card.

If the timing of the telephone call should fall on a weekend or a holiday, the call should be made on the next business day. If contact is not made on the designated day, study staff will continue calling until contact is made. Every telephone attempt and its outcome will be documented in the source document.

Table 4.1 and Table 4.2 present, respectively, the injection site reactions and systemic reactions that are prelisted in the diary cards and CRB, together with the intensity scales.

Table 4.1: Solicited injection site reactions: terminology, definitions, and intensity scales

CRB term (MedDRA lowest level term [LLT])	Injection site tenderness	Injection site erythema	Injection site swelling
MedDRA preferred term	Injection site pain	Injection site erythema	Injection site swelling
Diary card term	Tenderness	Redness	Swelling
Definition	Pain when the injection site is touched or injected limb mobilized	Presence of a redness including the approximate point of needle entry	Swelling at or near the injection site Swelling or edema is caused by a fluid infiltration in tissue or cavity and, depending on the space available for the fluid to disperse, swelling may be either soft (typically) or firm (less typical) to touch and thus can be best described by looking at the size of the swelling
Intensity scale*	Grade 1: Minor reaction when injection site is touched Grade 2: Cries or protests when injection site is touched Grade 3: Cries when injected limb is mobilized, or the movement of the injected limb is reduced	Grade 1: > 0 to < 25 mm Grade 2: ≥ 25 to < 50 mm Grade 3: ≥ 50 mm	Grade 1: > 0 to < 25 mm Grade 2: ≥ 25 to < 50 mm Grade 3: ≥ 50 mm

* For the subjective reaction of tenderness, parents / guardians will record the intensity level (Grade 1, 2, or 3) in the diary card. For the measurable reactions of redness and swelling, they will record just the size of the reaction, and the classification as Grade 1, 2, or 3 will be assigned at the time of the statistical analysis.

Table 4.2: Solicited systemic reactions: terminology, definitions, and intensity scales

CRB term (MedDRA LLT)	Fever	Vomiting	Crying abnormal	Drowsiness	Appetite lost	Irritability
MedDRA preferred term	Pyrexia	Vomiting	Crying	Somnolence	Decreased appetite	Irritability
Diary card term	Temperature	Vomiting	Abnormal crying	Drowsiness	Loss of appetite	Irritability
Definition	Elevation of temperature to ≥ 38.0°C (≥ 100.4°F)	Vomiting does not include spitting up	Inconsolable crying without a determined reason	Reduced interest in surroundings, or increased sleeping	See intensity scale	An excessive response to stimuli: increased fussiness, whining, and fretfulness despite attempts to comfort the infant and despite caregiver responses that would normally be soothing
Intensity scale*	Grade 1: ≥ 38.0°C to ≤ 38.5°C **or** ≥ 100.4°F to ≤ 101.3°F	Grade 1: 1 episode per 24 hours	Grade 1: < 1 hour	Grade 1: Sleepier than usual or less interested in surroundings	Grade 1: Eating less than normal	Grade 1: Easily consolable
	Grade 2: > 38.5°C to ≤ 39.5°C **or** > 101.3°F to ≤ 103.1°F	Grade 2: 2–5 episodes per 24 hours	Grade 2: 1–3 hours	Grade 2: Not interested in surroundings or did not wake up for a feed / meal	Grade 2: Missed 1 or 2 feeds / meals completely	Grade 2: Requiring increased attention
	Grade 3: > 39.5°C **or** > 103.1°F	Grade 3: ≥ 6 episodes per 24 hours or requiring parenteral hydration	Grade 3: > 3 hours	Grade 3: Sleeping most of the time or difficult to wake up	Grade 3: Refuses ≥ 3 feeds / meals or refuses most feeds / meals	Grade 3: Inconsolable

* For all reactions but fever, parents / guardians will record the intensity level (Grade 1, 2, or 3) in the diary card. For fever, they will record the body temperature, and the classification as Grade 1, 2, or 3 will be assigned at the time of the statistical analysis based on the unit used to measure the temperature and the intensity scale.

Important notes for the accurate assessment of temperature:

Parents / guardians are to measure body temperature once per day, preferably always at the same time. The optimal time for measurement is the evening when body temperature is the highest. Temperature is also to be measured at the time of any apparent fever. The observed daily temperature and the route of measurement are to be recorded in the diary card, and the highest temperature will be recorded by the site in the CRB. The preferred route for this study is rectal. Pre-vaccination temperature is also systematically collected by the Investigator on the source document. Tympanic thermometers must not be used.

4.3.2.3.3 Unsolicited Adverse Events

In addition to recording solicited reactions, parents / guardians will be instructed to record any other medical events that may occur during the 30-day period following each vaccination. Space will be provided in the diary card for this purpose.

Information on SAEs will be collected and assessed throughout the study, from the time of vaccination until 6 months after the last vaccination. Any SAE occurring at any time during the study will be reported by the Investigator in the CRB according to the completion instructions provided by the Sponsor; this includes checking the "Serious" box on the AE CRF and completing the appropriate Safety Complementary Information CRF. All information concerning the SAE is to be reported either as part of the initial reporting or during follow-up reporting if relevant information became available later (e.g., outcome, medical history, results of investigations, copy of hospitalization reports). In case a subject experiences febrile convulsion (neurological event associating fever and seizure), the assessment will be performed according to the "Guideline for definition and collection of cases of febrile convulsion", and this event will be considered an SAE. See Section 10 of the protocol for further details on SAE reporting.

For each unsolicited AE (whether serious or non-serious), the following information is to be recorded:

- Start and stop dates[a]
- Intensity of the event:
  - For measurable unsolicited AEs that are part of the list of solicited reactions, the size of the AE as well as the temperature for fever will be collected and analyzed based on the corresponding scale used for solicited reactions (see Table 4.1 and Table 4.2).

  - All other unsolicited AEs will be classified according to the following intensity scale:
    - Grade 1: A type of AE that is usually transient and may require only minimal treatment or therapeutic intervention. The event does not generally interfere with usual activities of daily living.

[a] The stop date of all related AEs will be actively solicited. For other events, the Investigator will provide the stop date when it becomes available. AEs for which no stop date was obtained during the course of the study will be considered as ongoing at the end of the study.

- Grade 2: A type of AE that is usually alleviated with additional therapeutic intervention. The event interferes with usual activities of daily living, causing discomfort but poses no significant or permanent risk of harm to the research participant.
- Grade 3: A type of AE that interrupts usual activities of daily living, or significantly affects clinical status, or may require intensive therapeutic intervention.

- Whether the AE was related to the investigational product (for unsolicited systemic AEs)
  - The Investigator will assess the causal relationship between the AE and the investigational product as either "Not related" or "Related", as described Section 4.3.2.3.6.

Action taken for each AE (e.g., medication)

- The action(s) taken by the parent / guardian to treat and/or manage any unsolicited AEs will be classified in the CRB using the following list (all applicable items should be checked):
  - None
  - Medication
  - Health care provider contact
  - Hospitalized
  - Discontinuation of study vaccination
- Whether the AE was Serious
  For each SAE, the Investigator will complete all seriousness criteria that apply (outcome, elapsed time, and relationship to study procedures)
- Whether the AE caused study discontinuation

4.3.2.3.4 Adverse Events of Special Interest

An AESI is defined as an event for which ongoing monitoring and rapid communication by the Investigator to the Sponsor must be done. The following AEs will be captured as AESIs throughout the study:

- Generalized seizures (febrile and non-febrile) (6) (7)
- Kawasaki disease (8) (9) (10)
- Guillain-Barré syndrome (11)
- Idiopathic thrombocytopenic purpura (ITP) (12) (13)

These events have been listed as AESIs based on the feedback received from the European Union regulators.
No safety concerns relating to these AESIs have been identified with the use of MenACYW conjugate vaccine in the completed clinical trials. Because of their medical importance and to ensure expedited communication to the Sponsor, these AESIs are to be considered and collected as SAEs and reported to the Sponsor according to the procedure described in the protocol. Further

instructions on the data collection for these events and the relevant definitions will be provided in the Operating Guidelines.

4.3.2.3.5 Medically-Attended Adverse Events

MAAE information will be collected throughout the study. MAAEs will be recorded as unsolicited AEs for up to D30 after each vaccination and as MAAEs until the next study visit on the appropriate diary cards. MAAEs that occur from D31 after the last vaccination visit until the 6-month follow up phone call will be recorded as such in the appropriate memory aid. An MAAE that occurs within the study period but meets the definition of an SAE should be reported only on the SAE Reporting Form. The Investigator will assess the causal relationship between the MAAE and the investigational or study product as either "Not related" or "Related", as described in Section 4.3.2.3.6..

4.3.2.3.6 Assessment of Causality

The Investigator will assess the ***causal relationship*** between each unsolicited systemic AE and the product administered as ***not related*** or ***related***, based on the following definitions:

- Not related – The AE is clearly / most probably caused by other etiologies such as an underlying condition, therapeutic intervention, or concomitant therapy; or the delay between vaccination and the onset of the AE is incompatible with a causal relationship; or the AE started before the first vaccination (screening phase, if applicable)
- Related – There is a "reasonable possibility" that the AE was caused by the product administered, meaning that there is evidence or arguments to suggest a causal relationship

Note: By convention, all AEs reported at the injection site (whether solicited or unsolicited) and all solicited systemic AEs are considered to be related to the administered product and therefore are referred to as reactions and do not require the Investigator's opinion on relatedness.

AEs likely to be related to the product, whether serious or not, that persist at the end of the study will be followed up by the Investigator until their complete disappearance or the stabilization of the subject's condition. The Investigator will inform the Sponsor of the date of final disappearance of the event or the date of "chronicity" establishment.

4.4 Derived Endpoints: Calculation Methods

4.4.1 Immunogenicity

4.4.1.1 Computed Values for Analysis

In order to appropriately manage extreme values (undetectable responses < lower limit of quantitation [LLOQ] and ≥ upper limit of quantitation [ULOQ]) for analysis purposes, the following computational rule is applied to the values provided in the clinical database for each BL drawn:

- If a value is < LLOQ, then use the computed value LLOQ/2
- If a value is between ≥ LLOQ and < ULOQ, then use the value
- If a value is ≥ ULOQ, then use the computed value ULOQ

4.4.1.2 Fold-rise

The derived endpoint fold-rise is driven by both baseline and post-baseline computed values and is computed as follows. Generally, for extreme values, this algorithm minimizes the numerator and maximizes the denominator.

- If the baseline computed value is < LLOQ and the post-baseline computed value is < LLOQ, then the fold-rise is 1
- If the baseline computed value is ≥ LLOQ and the post-baseline computed value is ≥ LLOQ, then the fold-rise is post-baseline computed value / baseline computed value
- If the baseline computed value is ≥ LLOQ and the post-baseline computed value is < LLOQ, then the fold-rise is (LLOQ/2) / baseline computed value
- If the baseline computed value is < LLOQ and the post-baseline computed value is ≥ LLOQ, then the fold-rise is post-baseline computed value /LLOQ

Note: If baseline or post-baseline is missing, then fold-rise is missing.

4.4.1.3 hSBA Vaccine Seroresponse

The derived hSBA vaccine seroresponse indicator for serogroups A, C, Y, and W will be taken as "Yes" if:
When evaluating the immunogenicity of 3-dose series of MenACYW conjugate vaccine comparable to MENVEO®:

- For a subject with a pre-1st dose vaccination titer < 1:8, the post-3rd dose vaccination titer must be ≥ 1:16
- For a subject with a pre-1st dose vaccination titer ≥ 1:8, the post-3rd dose vaccination titer must be ≥ 4-fold greater than the pre-1st dose vaccination titer

When evaluating the immunogenicity of 4-dose series of MenACYW conjugate vaccine comparable to MENVEO®:

- For a subject with a pre-1st dose vaccination titer < 1:8, the post-4th dose vaccination titer must be ≥ 1:16
- For a subject with a pre-1st dose vaccination titer ≥ 1:8, the post-4th dose vaccination titer must be ≥ 4-fold greater than the pre-1st dose vaccination titer

When evaluating the effect of 4th dose of MenACYW conjugate vaccine comparable to MENVEO®:

- For a subject with a pre-4th dose vaccination titer < 1:8, the post-4th dose vaccination titer must be ≥ 1:16
- For a subject with a pre-4th dose vaccination titer ≥ 1:8, the post-4th dose vaccination titer must be ≥ 4-fold greater than the pre-4th dose vaccination titer

4.4.1.4 Pertussis Vaccine Seroresponse

The derived Pertussis vaccine seroresponse indicator will be taken as "Yes" if:
When assessing the immunogenicity after 3rd dose of Pentacel® vaccine

- Pre-1st vaccination concentration < LLOQ, then post-3rd vaccination concentration should be ≥ 4x the LLOQ
- Pre-1st vaccination concentration ≥ LLOQ but < 4x the LLOQ, then post-3rd vaccination concentration should achieve a 4-fold rise (post-3rd vaccination/ pre-1st vaccination ≥ 4)
- Pre-1st vaccination concentration ≥ 4x the LLOQ, then post-3rd vaccination concentration should achieve a 2-fold response (post-3rd vaccination/ pre-1st vaccination ≥ 2)

When assessing the immunogenicity after 4th dose of Pentacel® vaccine
- Pre-booster (4th) vaccination concentration < LLOQ, then post- booster (4th) vaccination concentration should be ≥ 4x the LLOQ
- Pre-booster (4th) vaccination concentration ≥ LLOQ but < 4x the LLOQ, then post- booster (4th) vaccination concentration should achieve a 4-fold rise (post-4th vaccination/ pre-4th vaccination ≥ 4)
- Pre- booster (4th) vaccination concentration ≥ 4x the LLOQ, then post- booster (4th) vaccination concentration should achieve a 2-fold response (post-4th vaccination/ pre-4th vaccination ≥ 2)

4.4.2 Safety

4.4.2.1 Solicited Reactions

4.4.2.1.1 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 an investigator presence recorded as “No” and with all daily records missing, then all daily intensities will be derived as None.
2) For non-measurable (NM) 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 (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.

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.

4.4.2.1.2 Maximum Overall Intensity

Maximum overall intensity is derived from the daily intensities as described in Section 4.4.2.1.1 and is calculated as the maximum of the daily intensities over the period considered.

4.4.2.1.3 Presence

Presence is derived from the maximum overall intensity on the period considered:

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

The time period is displayed as D0-D3, D4-D7, D8 and later.

4.4.2.1.4 Time of Onset

Time of onset is derived from the daily intensities as described in Section 4.4.2.1.1. It corresponds to the first day with intensity of Grade 1, Grade 2, or Grade 3.

Note: If a reaction is not continuous (i.e., 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.

Time of onset period is displayed as, D0-D3, D4-D7.

4.4.2.1.5 Number of Days of Occurrence during the solicited period

Number of days of occurrence over the solicited period (D0 to D7) after each vaccination is derived from the daily intensities as described in Section 4.4.2.1.1. It corresponds to the number of days with one of daily intensities of Grade 1, Grade 2, or Grade 3. Number of days of occurrence on the solicited period with a specified intensity (e.g., Grade 3) may also be derived.

4.4.2.1.6 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 occurrence after each vaccination is derived from the daily intensities and the stop date of the reaction after the end of the solicited period. The overall number of days of occurrence is:

> (End date of the solicited event – last vaccination date) + (number of days of occurrence within the solicited period) – length of the solicited period + 1

If the end date of the solicited reaction is missing or is incomplete (contains missing data), then the overall number of days of occurrence will be considered as Missing.

4.4.2.1.7 Ongoing

Ongoing is derived from the last daily intensity of the solicited period as described in Section 4.4.2.1.1 and the maximum intensity on the ongoing period after each vaccination. 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)

4.4.2.2 Unsolicited AEs

4.4.2.2.1 Presence

An observation will be considered an event if it has at least a verbatim term and is not Grade 0 (None) intensity event.

Grade 0 events are not included in safety analysis but are included in separate listings.

4.4.2.2.2 Intensity

Intensity will be derived according to the following classification:

None, Grade 1, Grade 2, Grade 3, or Missing (Unknown).

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 as the intensity scales defined in the Section 4.4.2.1.1 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.

Intensity for the other unsolicited non-serious AEs will correspond to the value reported in the CRF. The maximum intensity corresponds to the highest intensity for a unique term.

4.4.2.2.3 Last Vaccination

Last vaccination before an unsolicited AE is derived from the start date of the unsolicited AE provided in the CRF 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.

4.4.2.2.4 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 4.4.2.2.3:
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.
The unsolicited AEs will be analyzed "Within 30 days" after each vaccination, which corresponds to AEs with a time of onset between day 0 and day 30.
Unsolicited AE that occurred before vaccination (negative time of onset) will not be included in analysis but will be listed separately.
Unsolicited AE which is non-serious, non-AESI and non-MAAE that occurred with a time of onset higher than defined above will not be included in analysis but will be listed separately.

- For unsolicited AE with missing day, month and year, the unsolicited AE will be classified as "Within 30 days"
- For unsolicited AE with partially missing start date, the partial available information will be used to determine if this AE is classified "Within 30 days" or "Not within 30 days". An AE will be categorized as "Not within 30 days" only if there is clear evidence from the partially missing start date that this AE happens before the first vaccination or after the last vaccination + 30 days. In all other situations, this AE is considered as "Within 30 days". Situations may happen as:

    - If the start date of AE has missing Day and non-missing Month and Year
        - If the "Month/Year of AE start date" < "Month/Year of first vaccination date", then it is clear that this unsolicited AE happened before the first vaccination and this unsolicited AE will not be included in the analysis but will be listed separately.
        - Else if the "Month/Year of last vaccination date" <= "Month/Year of AE start date" <= "Month/Year of (last vaccination date + 30 days)", then this unsolicited AE will be categorized as "Within 30 days".
        - Else if the "Month/Year of AE start date" > "Month/Year of (last vaccination date + 30 days)", then this unsolicited AE will be categorized as "not within 30 days". If the AE is non-serious, non-AESI and non-MAAE, then it will not be included in the analysis but will be listed separately.

    - If the start date of AE has missing Day and Month and non-missing Year:
        - If the "Year of AE start date" < "Year of first vaccination date", then it is clear that this unsolicited AE happens before the first vaccination and this

unsolicited AE will not be included in the analysis but will be listed separately.

- Else if the "Year of last vaccination date" <= "Year of AE start date" <= "Year of (last vaccination date + 30 days)", then this unsolicited AE will be categorized as "Within 30 days".
- Else if the "Year of AE start date" > "Year of (last vaccination date + 30 days)", then this unsolicited AE will be categorized as "not within 30 days". If the AE is non-serious, non-AESI and non-MAAE, then it will not be included in the analysis but will be listed separately.

A few examples of missing time of onset with start date of AE partially missing:

First injection date	Last injection date	Start date of the AE	Injection date + 30	Will be analyzed "Within 30 days" ?
16Oct2023	16Oct2023	Missing	N/A	Y
16Oct2023	16Oct2023	Sep2023	N/A	N
16Oct2023	16Oct2023	Oct2023	15Nov2023	Y
16Oct2023	16Oct2023	Nov2023	15Nov2023	Y
16Oct2023	16Oct2023	Dec2023	15Nov2023	N
05Jan2023	05Jan2023	2022	N/A	N
16Oct2023	16Oct2023	2023	15Nov2023	Y
08Dec2023	08Dec2023	2024	07Jan2024	Y
16Oct2023	16Oct2023	2024	15Nov2023	N

Time of onset period is displayed as D0-D3, D4-D7, D8-D14, D15 or later, and Missing.

Note: For project at a later stage, even if it's recommended to use the above rule, it's possible to follow the below rule, for homogeneity need:
The unsolicited AEs will be analyzed "Within 30 days" after each vaccination, which corresponds to Aes with a time of onset between day 0 and day 30 or missing .An AE with missing time of onset will be considered to have occurred just after the last vaccination (computed according to the section 4.4.2.2.3), so will be included in these tables.

4.4.2.2.5 Duration

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

End date of unsolicited AE - start date of unsolicited AE + 1.

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

4.4.2.3 SAEs (including AESIs)

An event will be considered as a serious event if "Yes" is checked for "Serious" in the CRF.
An event will be considered as an AESI if "Yes" is checked for "Is the event an AESI?" in the CRF.
SAEs (including AESIs) will be analyzed throughout the study using the following periods:

- Within 7 days (D0 to D7) after vaccination
- Within 30 days (D0 to D30) after vaccination
- During the study (i.e., all SAEs occurred during the study), including the 6-month follow-up period
- During the 6-month follow-up

4.4.2.4 Medically-Attended Adverse Event (MAAE)

An event will be considered as a MAAE if "Yes" is checked for "Is the event an MAAE?" in the CRF.
MAAE will be analyzed during the following time periods:

- Within 7 days after vaccination
- Within 30 days after vaccination
- During the study (i.e., all MAAEs occurred during the study), including the 6-month follow-up period.

4.4.2.5 Other Safety Endpoints

4.4.2.5.1 Action Taken

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

4.4.2.5.2 Seriousness

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

4.4.2.5.3 Outcome

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

4.4.2.5.4 Causality

This information will be summarized as collected in the field "Relationship to Investigational Product". Missing causal relationship will be handled as described in Section 5.4.1.2 .
Relationship to study procedure is only presented in the listing.

4.4.2.5.5 AEs Leading to Study Discontinuation

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 Discontinuation.

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 participant's status?" has "Adverse Event" checked.
- Safety overview table: A subject who either on the "Completion at End of Study" form, question" What was the participant's 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/Preferred Term (SOC/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.

4.4.3 Derived Other Variables

4.4.3.1 Age for Demographics

The age of a subject in the study was the calendar age in days at the time of inclusion.

4.4.3.2 Race, Preterm and Full-term Birth for Subgroup Analysis

Race will be presented as a demographic characteristic of interest using the following modalities:

- White
- Black
- Asian
- Other

Note: The original race categories collected in the study will still be used for demographics analyses. The American Indian or Alaska Native, Native Hawaiian or Other Pacific Islander, Mixed origin, Not Reported and Unknown will be classified to "Other" stratum for subgroup analysis purpose only.

Preterm will be derived as gestational age less than 37 weeks, and full-term birth will be defined as gestational age having at least 37 weeks between Group 1 and Group 2.

4.4.3.3 Subject Duration

The duration of a subject in the study until last visit is computed as follows:

Maximum (Date of last visit, Date of term form) – (Date of V01) +1.

The duration of a subject in the study including follow-up is computed as follows:

Maximum (Date of last visit, Date of term form, Date of last follow-up contact) − (Date of V01) +1.

4.4.3.4 Duration of the Study

The duration of the study until last visit is computed as follows:

Maximum of all subjects (Date of last visit, Date of termination form) − minimum for all subjects (Date of V01) +1.

The duration of the study including follow-up is computed as follows:

Maximum of all subjects (Date of last visit, Date of termination form, Date of last follow-up contact) − minimum for all subjects (Date of V01) +1

5 Statistical Methods and Determination of Sample Size

The statistical analyses will be performed under the responsibility of the Sponsor's Biostatistics platform using SAS® Version 9.4 software or later. The results of the statistical analysis will be available in the final clinical study report (CSR).

For descriptive purposes, the following statistics in Table 5.1 will be presented. In general, categorical variables will be summarized and presented by frequency counts, proportion percentages and confidence interval CIs. The 95% CIs of point estimates will be calculated using the normal approximation for quantitative data and the exact binomial distribution (Clopper-Pearson method) for proportions (14).

For immunogenicity results, assuming that Log_{10} transformation of the titers / data follows a normal distribution, at first, the mean and the 95% CI will be calculated on Log_{10} (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. For GMTs and GMCs, 95% CIs of point estimates will be calculated using normal approximation assuming they are log-normally distributed (log10 scale).

Reverse cumulative distribution curve (RCDC) figures will be provided for the antibody titers against meningococcal serogroups A, C, W, and Y.

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, 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 (seroresponse, vaccine response, cutoff)**	Number and percentage (95% CIs) of subjects.
	Continuous data (titer / concentration)	Log_{10}: Mean and standard deviation. Anti-Log_{10} (work on Log_{10} distribution, and anti-Log_{10} applied): Geometric mean (GM), 95% CI of the GM Graphical representation by Reverse Cumulative Distribution Curve (RCDC).

5.1 Statistical Methods

5.1.1 Hypotheses and Statistical Methods for Primary Objectives

5.1.1.1 Hypotheses

The primary objectives will be met if the following primary hypotheses are rejected:

- ***Primary Hypothesis 1 (MenACYW vaccine seroresponse rate after 4th dose):***
  Thirty days after the administration of the 4th dose of MenACYW conjugate vaccine at 12 to 15 months of age or MENVEO at 12 months of age, the percentages of subjects who achieve an hSBA seroresponse for meningococcal serogroups A, C, Y, and W in Subgroup 1a are non-inferior to the corresponding percentages in Subgroup 2a.

Null hypothesis (H0): p (men, G1a) - p (men, G2a) ≤ - 10%
Alternative hypothesis (H1): p (men, G1a) - p (men, G2a) > - 10%
where p(men, G1a) and p(men, G2a) are the percentages of subjects who achieve hSBA vaccine seroresponse[a] in Subgroup 1a and Subgroup 2a, respectively. Each of the serogroups A, C, Y, and W will be tested separately. If the lower limit of the 2-sided 95% confidence interval (CI) of the difference between the 2 proportions is > - 10% for each serogroup, the inferiority assumption will be rejected.

[a] hSBA vaccine seroresponse for serogroups A, C, Y and W is defined as:

- For a subject with a pre-vaccination titer < 1:8, the post-vaccination titer must be ≥ 1:16.
- For a subject with a pre-vaccination titer ≥ 1:8, the post-vaccination titer must be at least 4-fold greater than the pre-vaccination titer.

The overall non-inferiority of this primary hypothesis 1 will be demonstrated if all 4 individual null hypotheses are rejected.

- ***Primary Hypothesis 2 (MenACYW hSBA titer ≥ 1:8 after 3rd dose)***
  Thirty days after the administration of the 3rd dose of MenACYW conjugate vaccine or MENVEO at 6 months of age, the percentages of subjects who achieve hSBA ≥ 1:8 for meningococcal serogroups A, C, Y, and W in Group 1 are non-inferior to the corresponding percentages in Group 2.

Null hypothesis (H0): p(men, G1) - p(men, G2) ≤ - 10%
Alternative hypothesis (H1): p(men, G1) - p(men, G2) > - 10%

where p(men, G1) and p(men, G2) are the percentages of subjects who achieve hSBA achieve ≥ 1:8 in Group 1 and Group 2, respectively. Each of the serogroups A, C, Y, and W will be tested separately. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 10% for each serogroup, the inferiority assumption will be rejected.

The overall non-inferiority of this primary hypothesis 2 will be demonstrated if all 4 individual null hypotheses are rejected.

5.1.1.2 Statistical Methods

For these two primary hypotheses, each of the serogroups A, C, Y, and W will be tested separately. If the lower limit of the 2-sided 95% confidence interval (CI) of the difference between the 2 proportions is > -10% for each serogroup, the inferiority assumption will be rejected.
For each of the 4 non-inferiority hypotheses using the percentage rates (eg, the percentage of subjects achieving hSBA vaccine seroresponse rates or hSBA ≥ 1:8), the CI of the difference in proportions will be computed using the Wilson Score method without continuity correction (15).

Let $\hat{\theta} = p_1 - p_2$, 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 I 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 Group II 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.

For primary hypothesis 1, where Group I and Group II above refer to Subgroup 1a and 2a, respectively. For primary hypothesis 2, where Group I and Group II above refer to Group 1 and Group 2, respectively.

5.1.2 Hypotheses and Statistical Methods for Secondary Objectives

Summary of non-inferiority hypotheses for the secondary objectives are described in Table 5.2.

5.1.2.1 Hypotheses

I. First year evaluation at 30 days after the 6-month vaccinations (after the 3rd dose)

- **Secondary Hypothesis 1 (Anti-hepatitis B %** ≥ 10 mIU/mL**)**
  Thirty days after the 6-month HB vaccine administration, the percentage of subjects who achieve ≥ 10 mIU/mL in anti-HB surface antibody concentrations in Group 1 is non-inferior to that in Group 2.

Null hypothesis (H0): p(hep, G1) – p(hep, G2) ≤ - 10%
Alternative hypothesis (H1): p(hep, G1) – p(hep, G2) > - 10%
where p(hep, G1) and p(hep, G2) are the percentages of subjects in Group 1 and Group 2, respectively, who achieve ≥ 10 mIU/mL in anti-HB surface antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 2 (Anti-PRP % ≥ 0.15 µg/mL)**
  Thirty days after the 6-month Pentacel® vaccination, the percentage of subjects who achieve ≥ 0.15 µg/mL in anti-PRP antibody concentrations in Group 1 are non-inferior to that in Group 2.

Null hypothesis (H0): p(prp, G1) – p(prp, G2) ≤ - 5%
Alternative hypothesis (H1): p(prp, G1) – p(prp, G2) > -5%
where p(prp, G1) and p(prp, G2) are the percentages of subjects in Group 1 and Group 2 , respectively, who achieve ≥ 0.15 µg/mL in anti-PRP antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > -5%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 3 (Anti-PRP % ≥ 1.0 µg/mL)**
  Thirty days after the 6-month Pentacel® vaccination, the percentage of subjects who achieve ≥ 1.0 µg/mL in anti-PRP antibody concentrations in Group 1 are non-inferior to that in Group 2.

Null hypothesis (H0): p(prp, G1) – p(prp, G2) ≤ - 10%
Alternative hypothesis (H1): p(prp, G1) – p(prp, G2) > - 10%
where p(prp, G1) and p(prp, G2) are the percentage of subjects in Group 1 and Group 2, respectively, who achieve ≥ 1.0 µg/mL in anti-PRP antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 4 (Anti-polio % titer** ≥ 1:8 **)**
  Thirty days after the 6-month Pentacel® vaccination, the percentages of subjects who achieve ≥ 1:8 in anti-polio antibody titers (type 1, type 2, and type 3) in Group 1 are non-inferior to those in Group 2.

Null hypothesis (H0): p(pol, G1) – p(pol, G2) ≤ - 5%

Alternative hypothesis (H1): p(pol, G1) – p(pol, G2) > - 5%
where p(pol, G1) and p(pol, G2) are the percentages of subjects in Group 1 and Group 2, respectively, who achieve ≥ 1:8 in anti-polio antibody titers (type 1, type 2, and type 3). Each of the antigens of type 1, type 2, and type 3 will be tested separately. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 5% for each type, the inferiority assumption will be rejected.

- **Secondary Hypothesis 5 (Anti- rotavirus % ≥ 3-fold rise)**
  Thirty days after the 6-month rotavirus vaccine administration, the percentages of subjects who achieve ≥ 3-fold rise in serum IgA antibody concentrations against the rotavirus in Group 1 is non-inferior to those in Group 2.

Null hypothesis (H0): p(rota, G1) - p(rota, G2) ≤ - 10%
Alternative hypothesis (H1): p(rota, G1) - p(rota, G2) > -10%
where p(rota, G1) and p(rota, G2) are the percentages of subjects in Group 1 and Group 2, respectively, who achieve ≥ 3-fold rise in serum anti-rotavirus IgA antibody concentrations (serotypes G1, G2, G3, G4, and P1A[8]). If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > -10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 6 (Anti-rotavirus; GMC)**
  Thirty days after the 6-month rotavirus vaccine administration, the GMCs of the serum IgA antibodies against the rotavirus in Group 1 are non-inferior to the GMCs in Group 2.

Null hypothesis (H0): GMC(rota,G1) / GMC(rota, G2) ≤ 2/3
Alternative hypothesis (H1): GMC(rota,G1) / GMC(rota, G2) > 2/3
where GMC(rota, G1) and GMC(rota,G2) are the GMCs of the serum IgA antibodies against the rotavirus in Group 1 and Group 2, respectively. If the lower limit of the 2-sided 95% CI of the ratio of the GMCs from the 2 groups is > 2/3, the inferiority assumption will be rejected.

- **Secondary Hypothesis 7 (Anti-pertussis; GMC)**
  Thirty days after the 6-month Pentacel® vaccination, the GMCs of antibodies against the pertussis antigens (PT, FHA, PRN, and FIM) in Group 1 are non-inferior to the GMCs in Group 2.

Null hypothesis (H0): GMC(pert,G1) / GMC(pert, G2) ≤ 2/3
Alternative hypothesis (H1): GMC(pert,G1) / GMC(pert, G2) > 2/3
where GMC(pert, G1) and GMC(pert,G2) are the GMCs of antibodies against the pertussis antigens (PT, FHA, PRN, and FIM) in Group 1 and Group 2, respectively.
Each of the antigens of PT, FHA, PRN, and FIM will be tested separately. If the lower limit of the 2-sided 95% CI of the ratio of the GMCs from the 2 groups is > 2/3 for each antigen, the inferiority assumption will be rejected.

- **Secondary Hypothesis 8 (Anti-pneumococcal; GMC)**
  Thirty days after the 6-month Prevnar 13® vaccination, the GMCs of antibodies against the pneumococcal antigens (serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F) in Group 1 are non-inferior to the GMCs in Group 2.

Null hypothesis (H0): GMC(pne, G1) / GMC(pne, G2) ≤ 1/2
Alternative hypothesis (H1): GMC(pne, G1) / GMC(pne, G2) >1/2
where GMC(pne, G1) and GMC(pne, G2) are the GMCs of antibodies against the pneumococcal antigens (serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F) in Group 1 and Group 2, respectively.

Each of the serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F will be tested separately. If the lower limit of the 2-sided 95% CI of the ratio of the GMCs from the 2 groups is > 1/2 for each serotype, the inferiority assumption will be rejected.

II. Second year evaluation at 30 days after the 12-month vaccinations

- **Secondary Hypothesis 9 (Anti- measles % ≥ 255 mIU/mL)**
  Thirty days after the 12-month M-M-R® II vaccination, the percentages of subjects who achieve ≥ 255 mIU/mL in anti-measles antibody concentrations in Subgroup 1a are non-inferior to those in Subgroup 2a.

Null hypothesis (H0): p(mea, G1a) – p(mea, G2a) ≤ - 10%
Alternative hypothesis (H1): p(mea, G1a) – p(mea, G2a) > - 10%
where p(mea, G1a) and p(mea, G2a) are the percentages of subjects in Subgroup 1a and Subgroup 2a, respectively, who achieve ≥ 255 mIU/mL in anti-measles antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 10 (Anti-mumps% ≥ 10 mumps Ab units/mL)**
  Thirty days after the 12-month M-M-R® II vaccination, the percentages of subjects who achieve ≥10 Mumps Ab units/mL in anti-mumps antibody concentrations in Subgroup 1a are non-inferior to those in Subgroup 2a.

Null hypothesis (H0): p(mum, G1a) – p(mum, G2a) ≤ - 10%
Alternative hypothesis (H1): p(mum, G1a) – p(mum, G2a) > -10%
where p(mum, G1a) and p(mum, G2a) are the percentages of subjects in Subgroup 1a and Subgroup 2a, respectively, who achieve ≥ 10 Mumps Ab units/mL in anti-mumps antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 11 (Anti-rubella% ≥ 10 IU/mL)**
  Thirty days after the 12-month M-M-R® II vaccination, the percentages of subjects who achieve ≥ 10 IU/mL in anti-rubella antibody concentrations in Subgroup 1a are non-inferior to those in Subgroup 2a.

Null hypothesis (H0): p(rub, G1a) – p(rub, G2a) ≤ - 10%
Alternative hypothesis (H1): p(rub, G1a) – p(rub, G2a) > - 10%
where p(rub, G1a) and p(rub, G2a) are the percentage of subjects in Subgroup 1a and Subgroup 2a, respectively, who achieve ≥ 10 IU/mL in anti-rubella antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 12 (Anti-varicella% ≥ 5 gpELISA Ab units/mL)**
  Thirty days after the 12-month VARIVAX® vaccination, the percentage of subjects who achieve ≥ 5 gpELISA units/mL in anti-varicella antibody concentrations in Subgroup 1a is non-inferior to those in Subgroup 2a.

Null hypothesis (H0): p(var, G1a) – p(var, G2a) ≤ - 10%
Alternative hypothesis (H1): p(var, G1a) – p(var, G2a) > - 10%
where p(var, G1a) and p(var, G2a) are the percentages of subjects in Subgroup 1a and Subgroup 2a, respectively, who achieve ≥ 5 gpELISA units/mL in anti-varicella antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is >- 10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 13 (Anti-pneumococcal; GMC)**
  Thirty days after the 12-month Prevnar13® vaccination, the GMCs of antibodies against the pneumococcal antigens (serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F) in Subgroup 1a are non-inferior to the GMCs in Subgroup 2a.

Null hypothesis (H0): GMC(pne, G1a) / GMC(pne, G2a) ≤ 1/2
Alternative hypothesis (H1): GMC(pne, G1a) / GMC(pne, G2a) > 1/2
where GMC(pne, G1a) and GMC(pne,G2a) are the GMCs of antibodies against the pneumococcal antigens (serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F) in Subgroup 1a and Subgroup 2a, respectively. Each of the serotypes 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F will be tested separately. If the lower limit of the 2-sided 95% CI of the ratio of the GMCs from the 2 groups is > 1/2 for each serotype, the inferiority assumption will be rejected.

III. Second year evaluation at 30 days after the 15-month vaccination

- **Secondary Hypothesis 14 (Anti-PRP% ≥ 1.0 µg/mL)**
  Thirty days after the 15-month Pentacel® vaccination, the percentage of subjects who achieve ≥ 1.0 µg/mL in anti-PRP antibody concentrations in Subgroup 1b is non-inferior to that in Subgroup 2b.

Null hypothesis (H0): p(prp, G2b) – p(prp, G1b) ≤-10%
Alternative hypothesis (H1): p(prp, G2b) – p(prp, G1b) > -10%
where p(prp, G1b) and p(prp, G2b) are the percentages of subjects in Subgroup 1b and Subgroup 2b, respectively, who achieve ≥ 1.0 µg/mL in anti-PRP antibody concentrations. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 10%, the inferiority assumption will be rejected.

- **Secondary Hypothesis 15 (Anti-polio % titer ≥8)**
  Thirty days after the 15-month Pentacel® vaccination, the percentages of subjects who achieve ≥ 1:8 in anti-polio antibody titers (type 1, type 2, and type 3) in Subgroup 1b are non-inferior to those in Subgroup 2b.

Null hypothesis (H0): p(pol, G1b) – p(pol, G2b) ≤ - 5%
Alternative hypothesis (H1): p(pol, G1b) – p(pol, G2b) > - 5%
where p(pol, G1b) and p(pol, G2b) are the percentages of subjects in Subgroup 1b and Subgroup 2b, respectively, who achieve ≥ 1:8 in anti-polio antibody titers (type 1, type 2, and type 3). Each of the antigens of type 1, type 2, and type 3 will be tested separately. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > - 5% for each serotype, the inferiority assumption will be rejected.

- **Secondary Hypothesis 16 (Anti-pertussis; vaccine response rate)**
  Thirty days after the 15-month Pentacel® vaccination, the percentages of subjects with a pertussis vaccine response[a] for the pertussis antigens (PT, FHA, PRN, and FIM) in Subgroup 1b are non-inferior to the percentages in Subgroup 2b.

Null hypothesis (H0): p(pert, G1b) - p(pert, G2b) ≤ - 10%
Alternative hypothesis (H1): p(pert, G1b) - p(pert, G2b) > - 10%

[a] **Pertussis vaccine response is defined as**:
- Pre-booster (4th) vaccination concentration< LLOQ, then post- booster (4th) vaccination concentration should be ≥ 4x the LLOQ
- Pre- booster (4th) vaccination concentration ≥ LLOQ but < 4x the LLOQ, then post- booster (4th) vaccination concentration should achieve a 4-fold rise (post-4th vaccination/ Pre-4th vaccination ≥ 4)
- Pre- booster (4th) vaccination concentration ≥ 4x the LLOQ, then post- booster (4th) vaccination concentration should achieve a 2-fold response (post-4th vaccination/ Pre-4th vaccination ≥ 2)

where p(pert, G1b) and p(pert, G2b) are the percentages of subjects who achieve a pertussis vaccine response[a] in Subgroup 1b and Subgroup 2b, respectively. Each of the antigens of PT, FHA, PRN, and FIM will be tested separately. If the lower limit of the 2-sided 95% CI of the difference between the 2 proportions is > -10% for each antigen, the inferiority assumption will be rejected.

Table 5.2: Summary of non-inferiority hypotheses for the secondary objectives

Evaluation Time	Comparison Groups (G)	Antigen	Endpoint	Non-inferiority margin	Hypothesis #
1st Year, 30 days after the 6-month vaccination	G1 vs G2	Hepatitis B	% ≥ 10 mIU/mL	10%	1
		PRP	% ≥ 0.15 µg/mL	5%	2
		PRP	% ≥ 1.0 µg/mL	10%	3
		Polio†	% ≥ 1:8	5%	4
		Rotavirus	% ≥ 3-fold rise	10%	5
		Rotavirus	GMC	1.5	6
		Pertussis*	GMC	1.5	7
		Pneumococcal‡	GMC	2	8
2nd Year, 30 days after the 12-month vaccination	G1a vs G2a	Measles	% ≥ 255 mIU/mL	10%	9
		Mumps	% ≥ 10 mumps Ab units/mL	10%	10
		Rubella	% ≥ 10 IU/mL	10%	11
		Varicella	% ≥ 5 gpELISA units/mL	10%	12
		Pneumococcal‡	GMC	2	13
2nd Year, 30 days after the 15-month vaccination	G1b vs G2b	PRP	% ≥ 1.0µg/mL	10%	14
		Polio†	% ≥ 1:8	5%	15
		Pertussis*	Response rate	10%	16

* Pertussis: PT, FHA, PRN, and FIM
† Polio: type 1, type 2, type 3
‡ Pneumococcal: 1, 3, 4, 5, 6A, 6B, 7F, 9V, 14, 18C, 19A, 19F, and 23F

5.1.2.2 Statistical Methods

- For each of the non-inferiority hypotheses (hypothesis 1 to 5, 9 to 12 and 14 to 16) using the percentage rates (eg, vaccine response rate), the CI of the difference in proportions will be computed using the Wilson Score method without continuity correction (15).

Let $\hat{\theta} = p_1 - p_2$, 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 I 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 Group II 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.
where Group I and Group II above refer to Group 1a and Group 2a, Subgroup 1 and 2, or Subgroup 1b and 2b respectively for corresponding hypotheses.

- For each of the non-inferiority hypotheses (hypothesis 6 to 8 and 13) using the GMC ratios, the statistical methodology will be based on the use of the 2-sided 95% CI of difference in means of post-vaccination Log10 transformed concentrations between Group1 and Group 2 or between Subgroup 1a and Subgroup 2a with normal approximation. Logarithm transformation of the individual post-vaccination concentrations will be calculated. Assuming that Log_{10} transformation of the data is normally distributed, the 95% CI for the difference in Log_{10}(GMC) between two groups will be in the form:

$$\bar{X}_i - \bar{X}_j \pm t(1-\alpha/2, n_i + n_j - 2) \cdot S\sqrt{1/n_i + 1/n_j}$$

where $\bar{X}_i = \log_{10}(\text{GMC})$ is the mean of $\log_{10}$(concentration) of Group *i*,
$S^2 = [(n_i - 1)\, S_i^2 + (n_j - 1)\, S_j^2] / (n_i + n_j - 2)$ is the pooled sample variance,
n_i and S_i^2 are the sample size and sample variance of Group *i*,
$t(1-\alpha/2, n_i + n_j - 2)$ is the 100(1-α/2) percentile of the *t*-distribution with degrees of freedom $df = n_i + n_j - 2$.

The 95% CI for the GMC ratios between Group 1 and Group 2 or between Subgroup 1a and Subgroup 2a will be formed by taking the antilogarithms of the lower and upper limits of the 95% CI for the difference in $\log_{10}$(GMC) between both vaccine groups.

5.1.3 Statistical Methods for Descriptive Secondary Objectives

Descriptive analyses on the antibody titers against meningococcal serogroups A, C, W, and Y for all subjects in each group providing a blood sample at corresponding time points (Visit 1and Visit 4 for Group 1 and 2; Visit 5 and Visit 6 for Subgroup 1a and 2a only; Visit 7 and Visit 8 for Subgroup 1b and 2b only) using hSBA will include but not be limited to:

- hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8
- hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise
- hSBA vaccine seroresponse[a]
- Titer distribution and RCDC

Descriptive analyses on the antibody responses against the antigens for the routine pediatric vaccines (Pentacel®, Prevnar 13®, M-M-R® II, VARIVAX®, RotaTeq®, and ENGERIX-B®)

- Geometric mean concentrations (GMCs)
- Proportion of subjects with vaccine response
- Proportion of subjects with concentrations meeting the specific criteria

More details related to descriptive analyses in immunogenicity responses are described in Table **5.3** and Table 5.4.

[a] hSBA vaccine seroresponse for serogroups A, C, Y, and W is defined as:
When evaluating the immunogenicity of 3 dose series:
For a subject with a pre-1st vaccination titer < 1:8, the post-3rd vaccination titer must be ≥ 1:16
For a subject with a pre-1st vaccination titer ≥ 1:8, the post-3rd vaccination titer must be ≥ 4-fold greater than the pre-1st vaccination titer
When evaluating the immunogenicity of 4 dose series:
For a subject with a pre-1st vaccination titer < 1:8, the post-4th dose vaccination titer must be ≥ 1:16
For a subject with a pre-1st vaccination titer ≥ 1:8, the post-4th dose vaccination titer must be ≥ 4-fold greater than the pre-1st vaccination titer
When evaluating the effect of the 4th dose of MenACYW:
For a subject with a pre-4th dose vaccination titer < 1:8, the post-4th dose vaccination titer must be ≥ 1:16
For a subject with a pre-4th dose vaccination titer ≥ 1:8, the post-4th dose vaccination titer must be ≥ 4-fold greater than the pre-4th dose vaccination titer

Table 5.3: Immunogenicity response to antigens in meningococcal vaccine (A, C, Y, and W-135)

Antigens, Time Points and Groups	Endpoints
hSBA meningococcal serogroups A, C, Y, and W antibody titers for Group 1 and Group 2: 1. 30 days after the 3rd dose (after the 6-month vaccination)	• RCDCs • hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8 • hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre- vaccination (D0) to post-vaccination • hSBA vaccine seroresponse
hSBA meningococcal serogroups A, C, Y, and W antibody titers: 1. 30 days after the 4th dose (after the 12-month vaccination) for Subgroup 1a and Subgroup 2a 2. 30 days after the 4th dose (after the 15-month vaccination) for Subgroup 1b	• RCDCs • hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8 • hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre-vaccination (D0) to post-dose 4 vaccination • hSBA vaccine seroresponse
hSBA meningococcal serogroups A, C, Y, and W antibody titers to evaluate the immune persistence of MenACYW: For Subgroup 1a and Subgroup 2a (persistence at 12 month), Subgroup 1a and Subgroup 1b (persistence at 12 month vs 15 month): 1. 30 days after the 3rd dose (after the 6-month vaccination) 2. D0 before the 4th dose (before the 12-month vaccination for Subgroup 1a and before the 15-month vaccination for Subgroup 1b)	• hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8

Antigens, Time Points and Groups	Endpoints
hSBA meningococcal serogroups A, C, Y, and W antibody titers to evaluate the effect of 4th dose of MenACYW: For Subgroup 1a and Subgroup 1b (12 mo vs 15 mo): 1. D0 before the 4th dose (before the 12-month vaccination for Subgroup 1a and before the 15-month vaccination for Subgroup 1b) 2. 30 days after the 4th dose (after the 12-month vaccination for Subgroup 1a and after the 15-month vaccination for Subgroup 1b)	• hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre-4th dose vaccination to post-4th vaccination
hSBA meningococcal serogroups A, C, Y, and W antibody titers for Subgroup 1a and Subgroup 1b (12 mo vs 15 mo): 1. D0 before the 4th dose (before the 12-month vaccination for Subgroup 1a and before the 15-month vaccination for Subgroup 1b) 2. 30 days after the 4th dose (after the 12-month vaccination for Subgroup 1a and after the 15-month vaccination for Subgroup 1b)	• hSBA meningococcal serogroups A, C, Y, and W antibody titers ratio (Subgroup 1b/1a) • hSBA meningococcal serogroups A, C, Y, and W antibody titers titer ≥ 1:4 and ≥ 1:8 • hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:8 difference (Subgroup 1b-1a) • hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre-vaccination (D0) to post-4th vaccination • hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre-4th dose vaccination to post-4th vaccination • hSBA vaccine seroresponse difference (Subgroup 1b-1a)

Table 5.4: Immunogenicity response to antigens in routine pediatric vaccines

Antigens, Time Points and Groups	Endpoints
PRP (Hib) • D30 after the 6-month vaccination for Group 1 and Group 2 • D30 after the 15-month vaccination for Subgroup 1b and Subgroup 2b	• GMC
To evaluate immune persistence for PRP (Hib): • D30 after the 6-month vaccination for Subgroup 1b and Subgroup 2b • D0 before the 15-month vaccination for Subgroup 1b and Subgroup 2b	• GMC • Proportion of subjects with concentrations ≥ 0.15µg/mL
Pertussis: PT, FHA, PRN, FIM • D0 before 2-month (first) vaccination for Group 1 and Group 2 • D30 after the 6-month vaccination for Group 1 and Group 2 • D30 after the 15-month vaccination for Subgroup 1b and Subgroup 2b	• GMC For the second time point only: • Proportion of subjects with vaccine response after the 3rd dose (using post-3rd Dose and pre-1st Dose)
To evaluate immune persistence for Pertussis: • D30 after the 6-month vaccination for Subgroup 1b and Subgroup 2b • D0 before the 15-month vaccination for Subgroup 1b and Subgroup 2b	• GMC
Poliovirus : type 1, type 2, type 3 • D30 after the 6-month vaccination for Group 1 and Group 2 • D30 after the 15-month vaccination for Subgroup 1b and Subgroup 2b	• GMT
Hepatitis B • D30 after the 6-month vaccination for Group 1 and Group 2	• GMC • Proportion of subjects with concentrations ≥ 10 mIU/mL
Rotavirus: IgA • D0 before first vaccination for Group 1 and Group 2 (baseline) • D30 after the 6-month vaccination for Group 1 and Group 2	• GMC • Proportion of subjects with titer≥ 4-fold rise over baseline
Tetanus, Diphtheria • D30 after the 6-month vaccination for Group 1 and Group 2 And • D30 after the 15-month vaccination for Subgroup 1b and Subgroup 2b	• GMC • Proportion of subjects with concentrations ≥ 0.01 IU/mL • Proportion of subjects with concentrations ≥ 0.1 IU/mL

Antigens, Time Points and Groups	Endpoints
Pneumococcal (PCV13): • D30 after the 6-month vaccination for Group 1 and Group 2 • D30 after the 12-month vaccination for Subgroup 1a and Subgroup 2a	• GMC
Measles, Mumps, Rubella, Varicella • D30 after the 12-month vaccination for Subgroup 1a and Subgroup 2a.	• GMC

5.1.4 Statistical Methods for Observational Objectives

No hypotheses will be tested.

5.1.4.1 Statistical Methods

Safety

Safety results will be described for subjects in all study groups. The main parameters for the safety endpoints will be described by 95% Cis using the exact binomial method (Clopper-Pearson method) (15).
Safety analyses will contain at least the descriptions listed in Table 5.5:

Table 5.5: Statistical analyses for safety observational objectives

Safety Events	Time and Group	Description
Immediate unsolicited non-serious systemic AE	Within 30 minutes after each vaccination in Group 1 and Group 2	Proportion of subjects that have the event, MedDRA terms, intensity, relationship to vaccine, study discontinuation, duration
Solicited injection site reactions	D0 to D7 after each vaccination in Group 1 and Group 2	Proportion of subjects that have the event, time of onset, duration, intensity, action taken, study discontinuation, number of days of occurrence, temperature collection routes
Solicited systemic reactions		
Unsolicited non-serious AE/AR	D0 to D30 after each vaccination in Group 1 and Group 2	Proportion of subjects that have the event, MedDRA terms, time of onset, duration, intensity, relationship, action taken, study discontinuation
AEs leading to study discontinuation	D0 to D30 after each vaccination in Group 1 and Group 2; D31 to next visit after each vaccination in Group1 and Group2;	Proportion of subjects that have the event, MedDRA terms, time of onset, duration, intensity, relationship,

Safety Events	Time and Group	Description
	During the whole study in Group 1 and Group 2	action taken, study discontinuation
MAAE	D0 to D30 after each vaccination in Group 1 and Group 2; D31 to next visit after each vaccination in Group1 and Group2; During the whole study in Group 1 and Group 2	Proportion of subjects that have the event, MedDRA terms, onset, duration, intensity, relationship, action taken, study discontinuation
SAE (including AESI)	D0 to D7 after each vaccination in Group1 and Group 2; D0 to D30 after each vaccination in Group1 and Group 2; D31 to next visit after each vaccination in Group1 and Group2; During the whole study in Group1 and Group 2	Proportion of subjects that have the event, MedDRA terms, onset, duration, relationship, seriousness criteria, outcome, study discontinuation

Note: "Group1 and Group2" mentioned in the above table 5.5 for safety analyses also includes the Subgroup 1a, Subgroup 1b, Subgroup 2a and Subgroup 2b, when it is applicable.

5.1.5 Complementary Output

5.1.5.1 Sensitivity Analysis due to Blood Sample Issues

If applicable and necessary, additional immunogenicity sensitivity analyses will be performed for subjects with blood samples handled incorrectly during collection, processing, storage or shipment, and may have potential impact on the analysis results (eg, blood samples stored out of temperature after a power outage) based on the PPASs. The outputs will be provided in Appendix 15 of the CSR.
The endpoint for the sensitivity analyses will be the GMT.

- hSBA GMTs and 95% CI at each time point for each group by blood sample status – Per-Protocol Analysis Set 1
- hSBA GMTs and 95% CI at each time point for each group by blood sample status – Per-Protocol Analysis Set 2
- hSBA GMTs and 95% CI at each time point for each group by blood sample status – Per-Protocol Analysis Set 3

5.1.5.2 Subgroup Analysis by Race and Gender

Subgroup analyses by gender and race for will be provided in Appendix 15 of the CSR.
The gender subgroup analyses will have two categories (Female and Male), and the race subgroup analyses will have four categories (White, Black, Asian, and Other).

For primary and secondary immune responses after meningococcal vaccinations, these subgroup analyses will be conducted between Group 1 and Group 2 as well as between Subgroup 1a and 2a by gender and race.
For safety responses, these subgroup analyses will be mostly performed between Group 1 and Group 2 by gender and race.

Immunogenicity analyses

The followings are the serological endpoints will be assessed by gender and race:

1) hSBA vaccine seroresponse to meningococcal serogroups A, C, Y, and W assessed before first study vaccination on D0 and 30 days after the 4th meningococcal vaccination (Subgroup 1a vs Subgroup 2a).
2) Antibody titers ≥ 1:8 against meningococcal serogroups A, C, Y, and W measured by hSBA assessed 30 days after vaccinations at 6 months of age (Group 1 vs Group 2).
3) The following serological endpoints will be assessed (including effect of 4th dose of MenACYW or Menveo):

- Before the 12-month vaccination (pre-4th dose) for Subgroups 1a and 2a
  - hSBA meningococcal serogroups A, C, Y and W antibody titers
- 30 days after the 12-month vaccination for Subgroups 1a and 2a
  - hSBA meningococcal serogroups A, C, Y and W antibody titers
  - Percentage of subjects with titer ≥ 4-fold rise form pre-dose 4 vaccination to post-dose 4 vaccination and 95%CI

4) The following serological endpoints will be assessed (persistence of bactericidal antibodies after infant vaccination with MenACYW or Menveo):

- 30 days after the 6-month vaccination and before the 12-month vaccination for Subgroup 1a and Subgroup 2a
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - GMTs with 95% CI
  - Percentage of subjects with titer ≥1:4 and ≥1:8 and 95% CI

Safety analyses

Safety overview after any vaccine injection by gender– Overall Safety Analysis Set
Safety overview after vaccine injections at 2 months of age by gender – Safety Analysis Set 1
Safety overview after vaccine injections at 4 months of age by gender – Safety Analysis Set 2
Safety overview after vaccine injections at 6 months of age by gender – Safety Analysis Set 3
Safety overview after vaccine injections at 12 months of age by gender – Safety Analysis Set 4
Safety overview after vaccine injections at 15 months of age by gender – Safety Analysis Set 5
Safety overview after vaccine injections by gender –Safety Analysis Set for All 4-Dose Vaccination
Safety overview after any vaccine injection by race – Overall Safety Analysis Set
Safety overview after vaccine injections at 2 months of age by race – Safety Analysis Set 1
Safety overview after vaccine injections at 4 months of age by race – Safety Analysis Set 2
Safety overview after vaccine injections at 6 months of age by race – Safety Analysis Set 3
Safety overview after vaccine injections at 12 months of age by race – Safety Analysis Set 4
Safety overview after vaccine injections at 15 months of age by race – Safety Analysis Set 5
Safety overview after vaccine injections by race – Safety Analysis Set for All 4-Dose Vaccination

5.1.5.3 Subgroup Analyses by Preterm and Full-term Birth

Subgroup analyses by preterm (gestational age < 37 weeks) and full-term birth (gestational age ≥ 37 weeks) between Group 1 and Group 2 including Subgroup 1a, Subgroup 1b and Subgroup 2a will be provided in Appendix 15 of the CSR.

Immunogenicity analyses

The followings serological endpoints will be assessed:

- D0 (before first vaccination) for Group 1 and Group 2 by preterm and full-term birth:
  - hSBA meningococcal serogroups A, C, Y and W antibody titers
- 30 days after the 6-month vaccination (after the 3rd dose) for Group 1 and Group 2 by preterm and full-term birth:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - Titer distribution and reverse cumulative distribution curves (RCDCs)
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre-vaccination (D0) to post-3rd vaccination
  - hSBA vaccine seroresponse
- D0 before first vaccination for Subgroup 1a, Subgroup 1b and Subgroup 2a (G1a vs G2a and G1a vs G1b) by preterm and full-term birth:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
- 30 days after the 12-month vaccinations for Subgroup 1a and Subgroup 2a, after the 15-month vaccinations for Subgroup 1b (G1a vs G2a and G1a vs G1b) by preterm and full-term birth:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - Titer distribution and RCDCs
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre-vaccination (D0) to post-dose 4 vaccination
  - hSBA vaccine seroresponse
- 30 days after the 3rd dose (after 6-month vaccination) and before the 4th dose of MenACYW conjugate vaccine or MENVEO® (before 12-month vaccination for Subgroup 2a and Subgroup 1a, before 15-month vaccination for Subgroup 1b) to evaluate the immune persistence (G1a vs G2a and G1a vs G1b) by preterm and full-term birth:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8
- 30 days after the 4th dose (after 12-month vaccination for Subgroup 1a and Subgroup 2a, after 15-month vaccination for Subgroup 1b) of MenACYW conjugate vaccine or MENVEO® during 2nd year of life vaccination, including evaluating the effect of the 4th dose (G1a vs G2a and G1a vs G1b) by pre-term and full-term birth:
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 1:4 and ≥ 1:8
  - hSBA meningococcal serogroups A, C, Y, and W antibody titers ≥ 4-fold rise from pre-4th dose vaccination to post-4th dose vaccination
  - hSBA vaccine seroresponse

Safety analyses

Safety overview after any vaccine injection by the preterm and full-term birth – Overall Safety Analysis Set
Safety overview after vaccine injections at 2 months of age by the preterm and full-term birth – Safety Analysis Set 1
Safety overview after vaccine injections at 4 months of age by the preterm and full-term birth – Safety Analysis Set 2
Safety overview after vaccine injections at 6 months of age by the preterm and full-term birth – Safety Analysis Set 3
Safety overview after vaccine injections at 12 months of age by the preterm and full-term birth – Safety Analysis Set 4
Safety overview after vaccine injections at 15 months of age by the preterm and full-term birth – Safety Analysis Set 5
Safety overview after vaccine injections by the preterm and full-term birth –Safety Analysis Set for All 4-Dose Vaccination
Summary of unsolicited AEs within 30 days after vaccine injections by preterm and full-term birth - Overall Safety Analysis Set for Any Dose
Sensitivity Analysis due to COVID-19 Pandemic

The impact of COVID-19 pandemic situation on study conduction will be summarized through impact on visit procedures, study completion and major/critical protocol deviations due to COVID-19.

The subjects impacted by COVID-19 pandemic situation will be defined as the 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 a 6-month follow up not completed in pandemic form. If more than 10% of subjects are impacted as per this definition, baseline and demographics characteristics, and the main immunogenicity and safety endpoints will also be summarized in the subsets of subjects impacted/ non-impacted subjects to assess the potential impact of COVID-19 situation on study outcome.

The assessment of the impact COVID-19 pandemic will be based on but not limited to the following analysis:

- To summarize the impact of COVID-19 on the overall study conduct
  - Early termination due to COVID-19
  - Impact on visit conduct (visit not done, partially done, data collection method/procedure change)
  - Major and critical protocol deviations due to COVID-19

- To summarize disposition across study visits for subjects impacted/not impacted by COVID-19
- To summarize baseline demographics by randomized group for subjects impacted /not impacted by COVID-19
- To provide an individual listing of subjects impacted by COVID-19 and how they were impacted

- To provide a listing of visits impacted by COVID-19 and how they were impacted
- To assess the potential impact of COVID-19 on the main immunogenicity and safety endpoints in the subsets of impacted/non-impacted subjects

5.2 Analysis Sets

Three analysis sets will be used: the Full Analysis Set (FAS), the Per-Protocol Analysis set (PPAS), and the SafAS.

5.2.1 Full Analysis Set

There will be 3 FASs for this study. Immunogenicity analyses will be performed on the FAS for exploratory purposes.

5.2.1.1 Full analysis set 1 (FAS1) for infant vaccination (< 12 months of age):

The FAS1 is defined as the subset of all randomized subjects who received at least 1 dose of the study vaccine[a] in infancy (at Visit 1 to Visit 3, < 12 months of age) and have a valid post-vaccination serology result in infancy. All subjects will be analyzed according to the treatment group to which they were randomized.

5.2.1.2 Full analysis set 2 (FAS2) for immunogenicity persistence evaluation:

The FAS2 is defined as the subset of all randomized subjects who received at least 1 dose of the study vaccine in infancy (at Visit 1 to Visit 3, < 12 months of age) and have a valid pre-vaccination serology result at Visit 5 before the 12-month vaccination for Subgroups 1a and 2a or at Visit 6 before the 15-month vaccination for Subgroups 1b and 2b. All subjects will be analyzed according to the treatment group to which they were randomized.

5.2.1.3 Full analysis set 3 (FAS3) for 2nd year of life vaccination (≥ 12 months of age):

The FAS3 is defined as the subset of all randomized subjects who received at least 1 dose of the study vaccine in the 2nd year of life (≥ 12 months of age) and have a valid post-vaccination serology result in the 2nd year of life. All subjects will be analyzed according to the treatment group to which they were randomized.

5.2.2 Per-Protocol Analysis Set

Immunogenicity analyses will primarily be performed on PPAS. The PPAS is a subset of the FAS. There will be 3 per-protocol analysis sets (PPAS) corresponding to the 3 FASs:

- PPAS for infant vaccination (PPAS1)
- PPAS for immunogenicity persistence evaluation (PPAS2)
- PPAS for 2nd year of life vaccination (PPAS3)

[a] A study vaccine is any vaccine that is administered as part of the study, including the investigational product (MenACYW conjugate vaccine), the control vaccine (Menveo®) and the study-specific routine vaccines.

5.2.2.1 Per-Protocol Analysis Set 1 (PPAS1)

Serology obtained 30 days after the last vaccination visit at 6 months of age for all antigens will be used for immunogenicity analyses of infant stage of the study.
The subjects presenting with at least one of the following relevant protocol deviations during infancy will be excluded from the PPAS1:

- 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 vaccination schedule for infant year of the study
- 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:
  - Visit 1: 42 to 89 days of age
  - Visit 2: Visit 1 + 60 days (+14 days)
  - Visit 3: Visit 2 + 60 days (+14 days)
- Subject did not provide a post-dose serology sample in the proper time window or a post-dose serology sample was not drawn:
  - Blood sampling 2: Visit 3 + 30 days (+21 days)
- Subject received a protocol-prohibited therapy, medication or vaccine.
- Subject had other protocol violations that affected the subject's immune response, as determined by the clinical team before locking the database.

In addition to the reasons listed above, subjects will also be excluded from the PPAS1 if their serology sample did not produce a valid test result.

Vaccine correctness required by the PPAS1 includes not only the 3 doses of MenACYW conjugate vaccine or MENVEO®, but also the concomitant vaccines as scheduled (Pentacel®, PCV13, rotavirus, and hepatitis B) during infant stage.

In the event of a local or national immunization program with a pandemic influenza vaccine or coronavirus vaccine or any other vaccine as needed, subjects who receive one or more doses of the pandemic influenza or corona virus vaccine at any time during the study will not be withdrawn from the study.

5.2.2.2 Per-Protocol Analysis Set 2 (PPAS2)

Pre-vaccination serology obtained at Visit 5 before 12-month vaccinations for Subgroups 1a and 2a or Visit 6 before 15-month vaccinations for Subgroups 1b and 2b will be used for immunogenicity persistence analyses.

The subjects presenting with at least one of the following relevant protocol deviations during infancy will be excluded from the PPAS2:

- 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 vaccination schedule for infant year of the study
- 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:

  - Visit 1: 42 to 89 days of age
  - Visit 2: Visit 1 + 60 days (+14 days)
  - Visit 3: Visit 2 + 60 days (+14 days)
- A pre-dose serology sample at Visit 5 for Subgroups 1a or 2a before 12-month vaccinations or Visit 6 for Subgroups 1b and 2b before 15-month vaccinations was not drawn.
- Subject received a protocol-prohibited therapy, medication or vaccine.
- Subject had other protocol violations that affected the subject's immune response, as determined by the clinical team before locking the database.

In addition to the reasons listed above, subjects will also be excluded from the PPAS2 if their serology sample did not produce a valid test result.

Vaccine correctness required by the PPAS2 includes not only the 3 doses of MenACYW conjugate vaccine or MENVEO®, but also the concomitant vaccines as scheduled (Pentacel®, PCV13, rotavirus, and hepatitis B) during infant stage.

In the event of a local or national immunization program with a pandemic influenza vaccine or coronavirus vaccine or any other vaccine as needed, subjects who receive one or more doses of the pandemic influenza or corona virus vaccine at any time during the study will not be withdrawn from the study.

5.2.2.3 Per-Protocol Analysis Set 3 (PPAS3)

During the second year of life, the immunogenicity analyses for various antigens are performed on serology obtained 30 days after the given antigen administration if blood collection is planned at that time.
The subjects presenting with at least one of the following relevant protocol deviations will be excluded from the PPAS3:

- 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 vaccination schedule including the infant and the second year of the study
  - Subgroups 1a and 2a: Up to 12-month vaccinations, including the infant schedule
  - Subgroups 1b and 2b: Up to 15-month vaccinations, including the infant schedule and the 12 month vaccinations
- Subject received a vaccine other than the one that he / she was randomized to receive during both the infant and second year of the study
- Preparation and / or administration of vaccine was not done as per-protocol during both the infant and second year of the study
- Subject did not receive vaccine in the proper time window during the second year of the study:
  - Subgroup 1a
    - Visit 5: 12 to 15 months of age
  - Subgroup 1b
    - Visit 5: 12 to 15 months of age
    - Visit 6: 15 to 18 months of age

  - Subgroup 2a
    - Visit 5: 12 months of age
  - Subgroup 2b
    - Visit 5: 12 months of age
    - Visit 6: 15 to 18 months of age
- Subject did not provide a post-dose serology sample in the proper time window or a post-dose serology sample was not drawn for a given antigen specific analysis:
  - Blood sampling 4:
    - Subgroups 1a and 2a: Visit 5 + 30 days (+21 days)
    - Subgroups 1b and 2b: Visit 6 +30 days (+21 days)
- Subject received a protocol-prohibited therapy, medication or vaccine.
- Subject had other protocol violations that affected the subject's immune response, as determined by the clinical team before locking the database.

In addition to the reasons listed above, subjects will also be excluded from the PPAS3 if their serology sample did not produce a valid test result.
Vaccine correctness required by the PPAS3 includes not only the dose of MenACYW conjugate vaccine or MENVEO®, but also the concomitant vaccines as scheduled in 2nd year of study at least.
In the event of a local or national immunization program with a pandemic influenza vaccine or coronavirus vaccine or any other vaccine as needed, subjects who receive one or more doses of the pandemic influenza or coronavirus vaccine at any time during the study will not be withdrawn from the study.

5.2.3 Safety Analysis Set

The SafAS is defined as those subjects who have received at least one dose of the study vaccines[a] and have any safety data available. Specific SafAS will be defined and used after each vaccination. Safety analysis after all 4-dose vaccinations will be conducted as well.
All subjects will have their safety analyzed after each dose according to the vaccine they actually received, after any dose, and after all 4 doses according to the vaccine received at the first dose.
Safety data recorded for a vaccine received out of the protocol design will be excluded from the analysis (and listed separately).

5.2.3.1 Overall Safety Analysis Set for Any Dose

The overall SafAS is defined as those subjects who have received at least one dose of the study vaccines and have any safety data available. All subjects will have their safety analyzed after any dose according to the vaccine received at the first dose.
Safety data recorded for a vaccine received out of the protocol design will be excluded from the analysis (and listed separately).

5.2.3.2 Safety Analysis Set for Vaccination at 2 Months of Age

The SafAS1 for vaccination at around 2 months of age is defined as those subjects who have received the study vaccine at Visit 1 around 2 months of age and have any safety data available.

[a] for which safety data are scheduled to be collected

All subjects will have their safety analyzed after the Visit 1 dose according to the vaccines they actually received at Visit 1.
Safety data recorded for a vaccine received out of the protocol design at Visit 1 will be excluded from the analysis (and listed separately).

5.2.3.3 Safety Analysis Set for Vaccination at 4 Months of Age

The SafAS2 is defined as those subjects who have received the study vaccine at Visit 2 around 4 months of age and have any safety data available. All subjects will have their safety analyzed after this dose according to the vaccines they actually received at that visit.
Safety data recorded for a vaccine received out of the protocol design at that Visit 2 will be excluded from the analysis (and listed separately).

5.2.3.4 Safety Analysis Set for Vaccination at 6 Months of Age

The SafAS3 is defined as those subjects who have received the study vaccine at Visit 3 at around 6 months of age and have any safety data available. All subjects will have their safety analyzed after this dose according to the vaccines they actually received at that visit.
Safety data recorded for a vaccine received out of the protocol design at that Visit 3 will be excluded from the analysis (and listed separately).

5.2.3.5 Safety Analysis Set for Vaccination at 12 Months of Age

The safety analysis set 4 (SafAS4) is defined as those subjects who have received the study vaccine at Visit 5 around 12-15 months of age and have any safety data available. All subjects will have their safety analyzed after this dose according to the vaccine they actually received at that visit.
Safety data recorded for a vaccine received out of the protocol design at that Visit 5 will be excluded from the analysis (and listed separately).

5.2.3.6 Safety Analysis Set for Vaccination at 15 Months of Age

The SafAS5 is defined as those subjects who have received the study vaccine around 15-18 months of age and have any safety data available. All subjects will have their safety analyzed after this dose according to the vaccine they actually received at that visit.
Safety data recorded for a vaccine received out of the protocol design around 15-18 months of age will be excluded from the analysis (and listed separately).

5.2.3.7 Safety Analysis Set for all 4-Dose Vaccination

The SafAS6 is defined as those subjects who have received all 4 doses of the study vaccine (3 doses in infancy and one dose in the 2nd year of life at 12 or 15 month of age) and have any safety data available. All 4-dose vaccinations received in a series should be either all MenACYW or all Menveo®.
Safety data recorded for subjects not receiving all 4 doses of MenACYW or Menveo® will be excluded from the analysis (and listed separately).

5.3 Population used in Analyses

Immunogenicity analyses will primarily be performed on the Per-Protocol Analysis Set (PPAS) including PPAS1, PPAS2 and PPAS3. If the difference between PPAS and FAS is greater than 10%, a supplemental analysis based on FAS will be performed to evaluate consistency of the results.
The safety analysis will be performed on the Safety Analysis Sets (SafAS, and SafAS1 through SafAS6).

5.4 Handling of Missing Data and Outliers

5.4.1 Safety

No replacement will be done. In all subject listings, partial and missing data will be clearly indicated as missing.

5.4.1.1 Immediate

For unsolicited non-serious 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.

5.4.1.2 Causality

By convention, all events reported at the injection site (either solicited or unsolicited) will be considered as related to administered product and then referred to as reactions. In a same way, all solicited systemic events pre-listed in the CRF are also considered as related to vaccination and will be considered as reactions.

- For unsolicited systemic AE, missing relationship will be considered as related to study vaccine at the time of analysis. Missing relationship to non-IMP with a start date on or after 22APR2020[a] will be considered related to non-IMP vaccine.
- The missing relationship to study procedures for SAEs will not be imputed. The relationship with study procedures (SAEs only) will not be included in analysis but will be listed separately.

The original relationship information will be presented as collected in the AE listings with relationship collected.

5.4.1.3 Intensity

For solicited reactions, missing intensities will be handled as described in Section 4.4.2.1.1. For unsolicited non-serious AEs, missing intensities will remain missing and will not be imputed.

[a] 22APR2020 corresponds to the start date of collection in database of the relationship for non-IMP unsolicited systemic AEs.

5.4.1.4 Start Date and End Date

Missing or partially missing start dates or end dates for unsolicited Aes (including SAEs) 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 within or not within the defined time window (according to the section 4.4.2.2.4), according to the last vaccination (computed according to the section 4.4.2.2.3). If either the start date or end date is missing or partially missing, the duration will be considered missing.
Missing or partially missing end dates for ongoing solicited AEs will remain missing and not be imputed.

5.4.2 Immunogenicity

Missing data will not be imputed. No test or search for outliers will be performed.
The computational rule for undetectable responses < LLOQ and ≥ ULOQ is demonstrated in Section 4.4.1.1.
The derived endpoint of fold-rise is computed for extreme values, to minimize the numerator and maximizes the denominator as in Section 4.4.1.2.

5.5 Interim / Preliminary Analysis

No interim or preliminary analyses are planned.

5.6 Determination of Sample Size and Power Calculation

Calculation of Sample Size:

Approximately 2628 subjects will be enrolled. An estimated maximum of up to 34.1% non-evaluable subjects will result in approximately 1732 subjects in the PPAS available for immunogenicity analyses. Group 1 will have 1155 evaluable subjects and Group 2 will have 577 evaluable subjects.

In case of unexpected situation or any study hold resulting in an unexpected number of unevaluable subjects, total sample size may be increased to replace withdrawn, or unevaluable subjects.

For the Primary Objective 1 (after the 4th dose):
With 770 evaluable subjects in Subgroup 1a and 385 evaluable subjects in Subgroup 2a, the study will have around 98.4% power by using the Farrington and Manning's method to declare the non-inferiority of Subgroup 1a vs Subgroup 2a.

For the Primary Objective 2 (after the 3rd dose):
With 1155 evaluable subjects in Group 1 and 577 evaluable subjects in Group 2, the study will have around 98.8% power by using the Farrington and Manning's method to declare the non-inferiority of Group 1 vs Group 2.

Co-primary Objectives:
The study will have around 97.2% overall power by using the Farrington and Manning's method to declare the non-inferiority for co-primary objectives.

Table 5.6: Power estimates to reject the primary hypotheses

Primary #	Antigen	Endpoint	Non-inferiority margin	Estimated response*	Power (%)
Primary 1 (after 4th dose)	**A**	Seroresponse Rate	10%	80%	98.4
	C	Seroresponse Rate	10%	90%	99.98
	Y	Seroresponse Rate	10%	90%	99.98
	W	Seroresponse Rate	10%	90%	99.98
Primary 2 (after 3rd dose)	**A**	% ≥1:8	10%	70%	99.2
	C	% ≥1:8	10%	80%	99.9
	Y	% ≥1:8	10%	80%	99.9
	W	% ≥1:8	10%	80%	99.9
	Overall				97.2

Note: Evaluable subjects for Primary 1: Subgroup 1a: n = 770 subjects; Subgroup 2a: n = 385 subjects

Evaluable subjects for Primary 2: Group 1: n = 1155 subjects; Group2: n = 577 subjects

*Estimated responses are based on results observed in MENVEO® V59_33 (NCT01000311) and in study MET39 (Group 1 TetraMen-T 2, 4, 6, 12 months).

For the Secondary Objectives:

I. First year evaluation at 30 days after the 6-month vaccinations (after the 3rd dose)

With 1155 evaluable subjects in Group 1 and 577 evaluable subjects in Group 2, the study will have around 94.2% power by using the Farrington and Manning's method to declare the non-inferiority of Group 1 vs Group 2.

Table 5.7: Power estimates to reject the secondary hypotheses 1, 2, 3, 4, 5, 6, 7, and 8

Hypothesis #	Antigen	Endpoint	Non-inferiority margin	Estimated response*	Power (%)
1	Hepatitis B	% ≥ 10 mIU/mL	10%	98%	>99.9
2	PRP	% ≥ 0.15 µg/mL	5%	91%	94.3
3	PRP	% ≥ 1 µg/mL	10%	81%	99.9
4	Polio type 1	% ≥ 1:8	5%	≥ 99%	> 99.9
	Polio type 2	% ≥ 1:8	5%	≥ 99%	> 99.9
	Polio type 3	% ≥ 1:8	5%	≥ 99%	> 99.9
5	Rotavirus	% 3-fold rise	10%	95.2%	>99.9
6	Rotavirus	GMC	1.5	274.46 (323.52, 232.83) SD=2.00	>99.9
7	PT	GMC	1.5	65.0 (48.1, 87.7) SD=1.23	> 99.9
	FHA	GMC	1.5	91.4 (73.1, 114.3)	> 99.9

Hypothesis #	Antigen	Endpoint	Non-inferiority margin	Estimated response*	Power (%)
				SD=0.92	
	PRN	GMC	1.5	31.0 (19.9, 48.4) SD=1.83	>99.9
	FIM	GMC	1.5	173.5 (111.2, 269.1) SD=1.80	>99.9
8	Pneumococcal 1	GMC	2	3.5 (2.2 , 5.5) SD=1.10	> 99.9
	Pneumococcal 3	GMC	2	3.3 (1.9 , 6.0) SD=1.46	> 99.9
	Pneumococcal 4	GMC	2	2.0 (1.2 , 3.1) SD=1.07	> 99.9
	Pneumococcal 5	GMC	2	1.7 (1.2, 2.3) SD=0.74	> 99.9
	Pneumococcal 6A	GMC	2	4.8 (3.2, 7.3) SD=1.02	> 99.9
	Pneumococcal 6B	GMC	2	2.4 (1.1 ,5.3) SD=1.93	> 99.9
	Pneumococcal 7F	GMC	2	4.2 (2.5 , 7.0) SD=1.25	> 99.9
	Pneumococcal 9V	GMC	2	1.8 (1.3 , 2.6) SD=0.90	> 99.9
	Pneumococcal 14	GMC	2	8.3 (5.3 ,13.1) SD=1.11	> 99.9
	Pneumococcal 18C	GMC	2	2.3 (1.6 , 3.2) SD=0.81	> 99.9
	Pneumococcal 19A	GMC	2	1.6 (0.9 , 2.8) SD=1.37	> 99.9
	Pneumococcal 19F	GMC	2	3.8 (2.1, 6.7) SD=1.38	> 99.9
	Pneumococcal 23F	GMC	2	2.7 (1.6 ,4.7) SD=1.35	> 99.9

Hypothesis #	Antigen	Endpoint	Non-inferiority margin	Estimated response*	Power (%)
	Overall				94.2

Evaluable subjects: Group 1: n=1155; Group 2: n=577

*All estimates are from MET39, Group 2, 7-month or Group 1, 7-month data with the exception of rotavirus estimate from study V260-006 (NCT00090233) and study V419-005 (NCT01337167).

II. Second year evaluation at 30 days after the 12-month vaccinations

With 770 evaluable subjects in Subgroup 1a and 385 evaluable subjects in Subgroup 2a, the study will have around >99.9% power by using the Farrington and Manning's method to declare the non-inferiority of Subgroup 1a vs Subgroup 2a.

Table 5.8: Power estimates to reject the secondary hypotheses 9, 10, 11, 12 and 13

Hypothesis #	Antigen	Endpoint	Non-inferiority margin	Estimated response*	Power (%)
9	Measles	% ≥ 255 mIU/mL	10%	95%	> 99.9
10	Mumps	% ≥ 10 Mumps AbU/mL	10%	95%	> 99.9
11	Rubella	% ≥ 10 IU/mL	10%	95%	> 99.9
12	Varicella	% ≥ 5 gpELISA Ab units/mL	10%	95%	> 99.9
13	Pneumococcal 1	GMC	2	4.0 (2.2,7.2) SD= 1.37	> 99.9
	Pneumococcal 3	GMC	2	2.1 (1.5, 3.1) SD=0.86	> 99.9
	Pneumococcal 4	GMC	2	2.2 (1.5,3.2) SD=0.87	> 99.9
	Pneumococcal 5	GMC	2	1.3 (0.9, 2.0) SD=1.00	> 99.9
	Pneumococcal 6A	GMC	2	10.4 (6.3,17.3) SD=1.18	> 99.9
	Pneumococcal 6B	GMC	2	7.7 (4.5,13.4) SD=1.29	> 99.9
	Pneumococcal 7F	GMC	2	7.0 (4.4, 11.0) SD=1.05	> 99.9
	Pneumococcal 9V	GMC	2	1.9 (1.5, 2.5) SD=0.64	> 99.9
	Pneumococcal 14	GMC	2	10.0 (4.9, 20.2) SD=1.64	> 99.9
	Pneumococcal 18C	GMC	2	2.9 (2.2, 3.9) SD=0.69	> 99.9
	Pneumococcal 19A	GMC	2	9.1 (5.4, 15.1)	> 99.9

Hypothesis #	Antigen	Endpoint	Non-inferiority margin	Estimated response*	Power (%)
				SD=1.12	
	Pneumococcal 19F	GMC	2	14.0 (9.3, 21.0) SD=0.94	> 99.9
	Pneumococcal 23F	GMC	2	8.6 (5.7, 13.2) SD=1.00	> 99.9
	Overall				> 99.9

Evaluable subjects: of Subgroup 1a : n=770 subjects; Subgroup 2a: n = 385 subjects

*All estimates are from MET39, Group 1, 13-month data, except for the measles, mumps, rubella, and varicella estimates from study V59P21 (NCT00626327) and studyV419-007 (NCT01341639).

III. Second year evaluation at 30 days after the 15-month vaccinations

With 385 evaluable subjects in Subgroup 1b and 192 evaluable subjects in Subgroup 2b, the study will have around 98.6% power by using the Farrington and Manning's method to declare the non-inferiority of Subgroup 1b vs. Subgroup 2b.

Table 5.9: Power estimates to reject the secondary hypotheses 14, 15, and 16

Hypothesis #	Antigen	Endpoints	Non-inferiority margin	Estimated response*	Power (%)
14	PRP	% ≥ 1.0 µg/mL	10%	95%	99.9
15	Polio type 1	% ≥ 1:8	5%	≥ 99%	99.8
	Polio type 2	% ≥ 1:8	5%	≥ 99%	99.9
	Polio type 3	% ≥ 1:8	5%	≥ 99%	99.9
16	PT	Response Rate	10%	94%	99.8
	FHA	Response Rate	10%	95%	99.9
	PRN	Response Rate	10%	93%	99.5
	FIM	Response Rate	10%	95%	99.9
	Overall				98.6

Evaluable subjects: Subgroup 1b: n=385 subjects; Subgroup 2b: n = 192 subjects
*All estimates are from MET39, Group 2, 16-month data

5.7 Data Review for Statistical Purposes

Reviews of the data are anticipated through the data review process led by Data Management before database lock. This review of the data will include a statistical review.
Besides, an internal safety management team (SMT) will review the data being generated from all the ongoing studies with MenACYW conjugate vaccine at regular intervals for any new safety signals or safety concerns.

5.8 Changes in the Conduct of the Trial or Planned Analyses

Additional subgroup analysis based on preterm and full-term baseline status were added following communication with CBER.

Moreover, concomitant medications category fields were inactivated, and concomitant medications were coded with WHODrug dictionary by the coding specialists.

For the secondary objective 4, additional endpoints were added for PCV13 vaccine as follow:

- 30 days after the 6-month vaccination (after the 3rd dose) for Group 1 and Group 2:
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 0.35 µg/mL
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 1 µg/mL
- 30 days after the 12-month vaccinations for Subgroup 1a and Subgroup 2a:
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 0.35 µg/mL
  - Anti-pneumococcal antibody concentrations (PCV13)≥ 1 µg/mL.

Sections 4.4.2.2.3, 4.4.2.2.4 and 5.4.1.4 were updated according to new version of the Standard Safety Tables.

6 References List

1. Schillie S, Vellozzi C, Reingold A, et al. Prevention of hepatitis B virus infection in the United States: Recommendations of the Advisory Committee on Immunization Practices. MMWR. 2018; 67(1);1-31.
2. Harrison LH, Granoff DM, Pollard AJ. Meningococcal capsular group A, C, W, and Y conjugate vaccines. In: Plotkin SA, Orenstein WA, Offit PA, Edwards KM, editors. Vaccines. 7th ed. Philadelphia (PA): Elsevier;2018:619-43
3. Borrow R, Alarcón P, Carlos J, et al. The Global Meningococcal Initiative: global epidemiology, the impact of vaccines on meningococcal disease and the importance of herd protection. Expert Review of Vaccines. 2016;16(4):313-328.
4. Harrison OB, Claus H, Jiang Y, et al. Description and nomenclature of Neisseria meningitidis capsule locus. Emerg Infect Dis. 2013;19(4):566-73
5. Pollard AJ. Global epidemiology of meningococcal disease and vaccine efficacy. Pediatr Infect Dis J. 2004;23(12 Supp):S274-9.
6. Marcy SM, Kohl KS, Dagan R, et al. Fever as an adverse event following immunization: case definition and guidelines of data collection, analysis and presentation. Vaccine. 2004; 22:551-556.
7. Bonhoeffer J, Menkes J, Gold M, et al. Generalized convulsive seizure as an adverse event following immunization: case definition and guidelines for data collection, analysis, and presentation. Vaccine. 2004;22:557-62.
8. Newburger JW, Takahashi M, Gerber MA, et al. Diagnosis, treatment, and long-term management of Kawasaki disease: a statement for health professionals from the Committee on Rheumatic Fever, Endocarditis, and Kawasaki Disease, Council on Cardiovascular Disease in the Young, American Heart Association. Pediatrics. 2004;114(6):1708-33.
9. Centers for Disease Control and Prevention. Kawasaki Syndrome Case Report 2003. Available online: http://www.cdc.gov/kawasaki/pdf/ks_case_report-fillable.pdf. Accessed 17 November 2017.
10. Phuong LK, Bonetto C, Buttery J, et al and The Brighton Collaboration Kawasaki Disease Working Group. Kawasaki disease and immunisation: standardised case definition & guidelines for data collection, analysis. Vaccine. 2016;34(51):6582-96.
11. Sejvar JJ, Kohl KS, Gidudu J, et al. Guillain-Barré syndrome and Fisher syndrome: case definitions and guidelines for collection, analysis, and presentation of immunization safety data. Vaccine. 2011;29(3):599-612.
12. Chu YW, Korb J, Sakamoto M. Idiopathic thrombocytopenic purpura. Pediatr Rev. 2000;21(3):95-104.
13. Wise RP, Bonhoeffer J, Beeler J, et al. Thrombocytopenia: case definition and guidelines for collection, analysis, and presentation of immunization safety data. Vaccine.2007;25(31):5717-24
14. Newcombe R.G., Two-sided confidence intervals for the single proportion: comparison of seven methods, Statistics in Medicine, (1998) 17, 857-872
15. Newcombe R. Interval estimation for the difference between independent proportions: comparison of eleven methods. Stat Med. 1998;17:873-90.