A Randomized, Blind, Controlled Phase III Clinical Trial of Safety and Immunogenicity of Influenza Vaccine (Split Virion), Inactivated, Quadrivalent in Children Aged 6 to 35 Months

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Statistical Analysis Company：	Beijing Key Tech Statistics Technology Co., Ltd.

Statistical Analysis Plan

Approval Form

Statistical Analysis Company

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Approval Form

Sponsor

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1 Abbreviations

AE	Adverse Event
AESI	Adverse Events of Special Interest
ASaT	All Subjects as Treated
CI	Confidence Interval
FAS	Full Analysis Set
GBS	Guillain-Barré syndrome
GMT	Geometric Mean Titer
GMI	Geometric Mean Increase
HI	Haemagglutination Inhibition
ITT	Intention-to-Treat
J2R	Jump to Reference
LOCF	Last Observation Carried Forward
MedDRA	Medical Dictionary for Regulatory Activities
PPS	Per Protocol Set
PT	Preferred Term
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SOC	System Organ Class
SS	Safety Set
TEAE	Treatment Emergent Adverse Event
TPA	Tipping point analysis
WHODD	World Health Organization Drug Dictionaries

2 Introduction

2.1 Preface

This document is the Statistical Analysis Plan (SAP) for the "A Randomized, Blind, Controlled Phase III Clinical Trial of Safety and Immunogenicity of Influenza Vaccine (Split Virion), Inactivated, Quadrivalent in Children Aged 6 to 35 Months". It mainly expounds the specific statistical analysis methods used for analyzing and reporting the baseline characteristics of the subjects, immunogenicity evaluation, and safety evaluation.

This SAP will be finalized and approved before the database is locked. The corresponding statistical analysis programming work will be gradually improved as the research data accumulates until the database is locked.

The TFL shells of this SAP will be provided separately as an attachment.

2.2 Purpose of Analysis

The purpose of this SAP is to evaluate the immunogenicity and safety of the Quadrivalent Influenza Vaccine developed by Sinovac Biotech Co., Ltd. when administered to infants and young children aged 6 to 35 months. The corresponding statistical analysis results will be presented in the final statistical analysis report, clinical study report, article publication, and other requirements.

Post hoc exploratory analyses can further mine the study data, but since it is unpredictable, it is not described in this SAP. If it occurs subsequently, the corresponding statistical analysis methods for post hoc exploratory analysis will be detailed in the final statistical analysis report and clinical study report.

Additional analyses conducted for other purposes, such as manuscript publication, regulatory authority requirements, or sponsor requests, are likewise not described in this SAP because they could not be anticipated. If they occur, the corresponding statistical methods for these additional analyses may not be detailed in the final clinical study report, but will be detailed in the document presenting the additional results.

2.3 Changes in the Planned Analysis

There are no changes to the analyses planned in the protocol.

3 Study Objective, Estimand, and Endpoint

3.1 Objective

3.1.1 Primary Objective

− To evaluate the non-inferiority of immunogenicity in participants aged 6-35 months after 2 doses of 0.5mL quadrivalent influenza vaccine (0.5mL-dose QIV) compared with two trivalent influenza vaccines (TIVs, including BV or BY) for the shared strains, in terms of seroconversion rate (SCR) and Geometric mean titer (GMT) for HI antibody;

− To evaluate the non-inferiority of immunogenicity in participants aged 6-35 months after 2 doses of 0.25mL quadrivalent influenza vaccine (0.25mL-dose QIV) compared with two trivalent influenza vaccines (including BV or BY) for the shared strains, in terms of SCR and GMT for HI antibody;

− To evaluate the absolute criteria of HI antibody SCR and seroprotection rate (SPR) for each strain specified in the protocol in participants aged 6-35 months after two doses of 0.25mL-dose or 0.5mL-dose QIVs.

3.1.2 Secondary Objective

− To evaluate the non-inferiority of immunogenicity in participants aged 6-35 months after 2 doses of 0.5mL-dose QIV compared with 0.25mL-dose QIV for each strain, in terms of SCR and GMT of HI antibody;

− To evaluate the superiority of immunogenicity in participants aged 6-35 months after 2 doses of 0.25mL-dose or 0.5mL-dose QIVs compared with two TIVs (including BV or BY) for the unique strain, in terms of SCR and GMT of HI antibody;

− To evaluate the immunogenicity in participants aged 6-35 months after vaccination of 2 doses of 0.25mL-dose or 0.5mL-dose QIVs;

- To evaluate the safety of 0.25mL-dose or 0.5mL-dose QIVs in participants aged 6-35 months.

3.2 Estimand

3.2.1 Primary Estimand

Table 3.2.1.1 Primary Estimand

	Primary Estimand
Population	The target population involves all participants aged 6 to 35 months who have completed two doses of vaccination.
Variable	(1) Whether the H1N1 and H3N2 HI antibodies are seroconverted in the test group (0.5mL or 0.25mL) and the pooled TIV group 28 days after the second dose. (2) Whether the BV HI antibody is seroconverted in the test group (0.5mL or 0.25mL) and the TIV-BV groups 28 days after the second dose. (3) Whether the BY HI antibody is seroconverted in the test group (0.5mL or 0.25mL) and the TIV-BY group 28 days after the second dose. (4) H1N1 and H3N2 HI antibody titers of the test group (0.5mL or 0.25mL) and the pooled TIV group 28 days after the second dose. (5) BV HI antibody titers of the test group (0.5mL or 0.25mL) and the TIV-BV groups 28 days after the second dose. (6) BY HI antibody titers of the test group (0.5mL or 0.25mL) and the TIV-BY group 28 days after the second dose. (7) Whether the H1N1, H3N2, BV, and BY HI antibody titer reach the protective antibody level in the test group (0.5mL or 0.25mL) 28 days after the second dose.
Treatments	Subjects received two doses of the test vaccine or the control vaccine according to the 0,28-day schedule.
Intercurrent Events and Treatment Strategies	The definition is shown in Table 3.2.1.2.
Population-level Summary	(1) Difference between the SCRs of H1N1 and H3N2 HI antibodies of the test group (0.5mL or 0.25mL) and the pooled TIV group 28 days after the second dose. (2) Difference between the SCRs of BV serum HI antibodies of the test group (0.5mL or 0.25mL) and the TIV-BV group 28 days after the second dose. (3) Difference between the SCRs of BY HI antibodies of the test group (0.5mL or 0.25mL) and the TIV-BY group 28 days after the second dose vaccination. (4) GMT ratios of H1N1 and H3N2 HI antibodies of the test group (0.5mL or 0.25mL) and the pooled TIV group 28 days after the second dose (5) GMT ratios of BV serum HI antibodies of the test group (0.5mL or 0.25mL) and the trivalent control group (including BV) after 28 days of full-course vaccination. (6) GMT ratios of BY serum HI antibodies of the test group (0.5mL or 0.25mL) and the trivalent control group (including BY) after 28 days of full-course vaccination. (7) Seroprotection rates (SPRs) of H1N1, H3N2, BV, and BY HI antibody in the test group (0.5mL or 0.25mL) after 28 days after the second dose.

Definitions:

Susceptible population: HI antibody titer <1:10 before immunization

Non-susceptible population: HI antibody titer ≥1:10 before immunization

Seroconversion: HI antibody titer <1:10 before immunization, and the antibody titer ≥1:40 after immunization, or

HI antibody titer ≥1:10 before immunization, and the antibody titer after immunization increased ⩾4 folds.

Protection: HI antibody titer ≥1:40 as the limit of protective antibody

Table 3.2.1.2 Intercurrent Events and Treatment Strategies of Primary Estimand

Intercurrent Event	Treatment Strategy	Remarks
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]

3.2.2 Secondary Estimand 1

Table 3.2.2.1 Secondary Estimand 1

	Secondary Estimand 1
Population	The target population involves all participants aged 6 to 35 months who have completed two doses of vaccination.
Variable	(1) Whether the H1N1, H3N2, BV, and BY HI antibodies are seroconverted in both the test group 2 (0.5mL) and the test group 1 (0.25mL) 28 days after the second dose. (2) H1N1, H3N2, BV, and BY serum HI antibody titers of the test group 2 (0.5mL) and the test group 1 (0.25mL) 28 days after the second dose.
Treatments	As the same as the primary estimand
Intercurrent Events and Treatment Strategies	As the same as the primary estimand
Population-level Summary	(1) Difference between the SCRs of H1N1, H3N2, BV, and BY serum HI antibodies of the test group (0.5mL) and the test group (0.25mL) 28 days after the second dose. (2) GMT ratios of H1N1, H3N2, BV, and BY serum HI antibodies between the test group (0.5mL) and the test group (0.25mL) 28 days after the second dose.

3.2.3 Secondary Estimand 2

Table 3.2.3.1 Secondary Estimand 2

	Secondary Estimand 2
Population	The target population involves all participants aged 6 to 35 months who have completed two doses of vaccination.
Variable	(1) Whether the BV HI antibody is seroconverted in the test group (0.5mL or 0.25mL) and the TIV-BY group 28 days after the second dose. (2) Whether the BY HI antibody is seroconverted in the test group (0.5mL or 0.25mL) and the TIV-BV group 28 days after the second dose.

	Secondary Estimand 2
	(3) BV HI antibody titers of the test group (0.5mL or 0.25mL) and the TIV-BY group 28 days after the second dose. (4) BY HI antibody titers of the test group (0.5mL or 0.25mL) and the TIV-BV group 28 days after the second dose.
Treatments	As the same as the primary estimand
Intercurrent Events and Treatment Strategies	As the same as the primary estimand
Population-level Summary	(1) Difference between the SCRs of BV HI antibody of the test group (0.5mL or 0.25mL) and TIV-BY group 28 days after the second dose. (2) Difference between the SCRs of BY serum HI antibody of the test group (0.5mL or 0.25mL) and TIV-BV 28 days after the second dose. (3) GMT ratios of BV HI antibodies of the test group (0.5mL or 0.25mL) and the TIV-BY group 28 days after the second dose. (4) GMT ratios of BY HI antibodies of the test group (0.5mL or 0.25mL) and the TIV-BV group 28 days after the second dose.

3.2.4 Secondary Estimand 3

Table 3.2.4.1 Secondary Estimand 3

	Secondary Estimand 3
Population	The target population involves the subjects (susceptible population) with the HI antibody titer <1:10 before immunization, aged 6 to 35 months, who have completed the full-course vaccination of two doses.
Variable	As the same as the primary estimand
Treatments	As the same as the primary estimand
Intercurrent Events and Treatment Strategies	As the same as the primary estimand
Population-level Summary	As the same as the primary estimand

3.3 Safety Endpoint

(1) Adverse event (AE):

- **Solicited adverse event:** Including adverse events at inoculated sites (local) and non-inoculated sites (systemic) that occur within 7 days after each dose of vaccination
  - **Solicited local adverse events (inoculated sites):** tenderness/pain, induration, swelling, flush, rash, and itching
  - **Solicited systemic adverse events (including vital signs) (non-inoculated sites):** pyrexia (axillary temperature), acute allergic reaction, skin and mucosal abnormalities, diarrhea, anorexia, vomiting, nausea, cough, fatigue, irritation, and suppression.
- **Unsolicited adverse event:** Any adverse event other than solicited AEs or solicited AEs that occur outside the solicitation period.

(2) SAE: SAE that occurs from the first dose of vaccination up to 6 months after the second dose of vaccination.

(3) Adverse events of special interest (AESI): Adverse events of special interest that occur from the first dose of vaccination up to 6 months after the second dose of vaccination (including Guillain-Barré syndrome (GBS), seizure, encephalitis, myelitis, neuritis, Bell's palsy, vasculitis, and thrombocytopenia).

4 Study Design

4.1 Overall Design

This study is a multi-center, randomized, double-blind, controlled trial design. The control vaccines will be Anflu ®, trivalent influenza virus split vaccines already marketed by Sinovac (including type B Victoria or type B Yamagata). This trial is a multi-center, randomized, double-blinded, and positive-controlled design. The control vaccine is Anflu®, two0.25mL-dose trivalent influenza virus split vaccines (TIVs, containing a B Victoria lineage or B Yamagata lineage） marketed by Sinovac. This trial will be conducted on children aged 6 to 35 months with the informed consent of their guardians. A total of 3,300 participants are planned to be recruited and randomly assigned to 4 groups in a ratio of 2:2:1:1, namely the 0.25mL-dose QIV group, 0.5ml-dose QIV group, TIV-BV group, and TIV-BY group. All participants receive 2 doses of the test vaccine or control vaccine according to the 0, 28-day immunization schedule. Among them, subjects aged 6 to 11 months (before their first birthday) will receive intramuscular injections in the anterolateral thigh, and those aged 12 to 35 months will receive intramuscular injections in the deltoid muscle of the upper arm.

Table 4.1.1 Phase III Study Design

Group	Vaccine	Sample Size	Immunization schedule (days)	Antibody test blood collection point (day)
Test Group 1	Influenza Vaccine (Split Virion), Inactivated, Quadrivalent (0.25mL)	1100	0，28	0，56
Test Group 2	Influenza Vaccine (Split Virion), Inactivated, Quadrivalent (0.5mL)	1100	0，28	0，56
Control Group 1	Influenza Vaccine (Split Virion), Inactivated, Anflu (0.25mL, including BV)	550	0，28	0，56
Control Group 2	Influenza Vaccine (Split Virion), Inactivated, Anflu (0.25mL, including BY)	550	0，28	0，56
Total	\	3300	\	\

Safety evaluation: All participants are observed for immediate reactions for 30 minutes after each dose, solicited systemic and local adverse events within 0-7 days, and unsolicited adverse events within 0-28 days after each dose. Additionally, monitoring of serious adverse events (SAEs) and adverse events of special interest (AESIs) is conducted from the first dose administration to 6 months after the second vaccination to evaluate the safety.

Immunogenicity evaluation: About 2 to 3mL of venous blood is collected from each subject before vaccination and 28 days after full-course vaccination, respectively, and then the serum is isolated and

used for influenza hemagglutination inhibition (HI) antibody test to evaluate the immunogenicity of the vaccine.

4.2 Randomization

This clinical trial is a randomized, blinded, positive-controlled design. Then, random blind code will be generated by a third-party randomization statistician independent of the project using the SAS statistical software (version 9.4) according to the preset block lengths, with the subject numbers ranging from 4001 to 7300. Vaccines will be randomly assigned and blinded according to random numbers

4.3 Sample Size

This trial requires the test group: ① the non-inferiority comparison with the control groups is established and the next statistical test is carried out; ② when the absolute criteria of SCR and SPR are achieved, the trial can be considered as successful and a positive conclusion can be drawn for the test group. In this trial, when evaluating between the test group 1 (0.25-mL QIV) and test group 2 (0.5mL-QIV), the strategy of sequential test is adopted. First, test group 2 is evaluated. If the positive conclusion of test group 2 is established, test group 1 is further evaluated. Therefore, class I error correction is no longer carried out. The sample size of phase III clinical trials is estimated first by non-inferiority design, namely the QIV group is non-inferior to the TIV group. The primary endpoints of this study include two indicators: SCR and GMT. Only when the SCR and GMT of four serotypes in test group are non-inferior to the control vaccine, the investigational vaccine is considered to be non-inferior to the control vaccine. The total power is 80%, the type II error is 0.2. Based on the different sample size requirements of the two indicators, the type II error is allocated, with 0.16 for SCR comparison and 0.04 for GMT comparison.

(1) Calculation of sample size with the SCR as the indicator:

According to the results of the previous Anflu phase IV clinical trial and other literature, the SCR of the types contained in the vaccine 28 days after vaccination is over 65%, which is used as the reference proportion to estimate the sample size. The test level is taken as one-sided α=0.025, the power of non-inferiority comparison for the SCR of a serotype is taken as 1-β1/4=1-0.16/4=96%, and the non-inferiority threshold Δ is taken as -10%. When comparing the non-inferiority of BY or BV strain, NT:NC=2:1, and PASS software is used to calculate the sample size, the test group needs 914 samples and the control group needs 457 samples. When the non-inferiority test of H1N1 or H3N2 strain is carried out, two control groups are pooled. The sample size ratio of the test group and the pooled control group is 1:1, and the test group and the pooled control group need 625 participants.

(2) Calculation of sample size with the GMT as the indicator:

Using the antibody GMT 28 days after vaccination as the outcome measure, the non-inferiority margin is ≥2/3 of the 95%CI lower limit of GMT ratio between the test group and the control group, and the non-inferiority margin after logarithmic conversion is Δ=-0.176. The test level is taken as one-sided α=0.025, and the power of a certain serotype GMT for non-inferiority comparison is taken as 1-β2/4=1-0.04/4=99%, and the standard deviation after logarithmic transformation is taken as σ =0.7 with

reference to the previous research results. When comparing the non-inferiority of BY or BV, NT:NC=2:1, and PASS software is used to calculate the sample size, the test group needs 874 samples and the control group needs 437 samples. When the non-inferiority test of H1N1 or H3N2 type is carried out, two control groups are pooled. The sample size ratio of the test group and the pooled control group is 1:1, and the test group and the pooled control group need 582 participants each.

When the SCR and GMT of the test group reach the hypothesis of non-inferiority, the absolute criteria of the SCR and seroprotection rate (SPR) of the test group are further hypothesized for test. The trial requires that the SCR and SPR of the four strains of immune antibodies reach the absolute criteria, and the total power of the trial is 80%, so the power of each test is 1- (0.2/8)=0.975.

(3) Calculation of sample size with the absolute criteria of SCR:

Assuming that the SCR after vaccination is 65%, the test level is one-sided α=0.025, the lower limit of the confidence interval of the SCR should be greater than 30%, and the power of each test is 97.5%. The sample size is calculated by PASS software, and the test group needs 28 samples.

(4) Calculation of sample size with the absolute criteria of SPR:

Assuming that the SCR after vaccination is 75%, the test level is one-sided α=0.025, the lower limit of the confidence interval of the SPR should be greater than 60%, and the power of each test is 97.5%. The sample size is calculated by PASS software, and the test group needs 146 samples.

In consideration of the above the maximum sample size and the dropout rate of about 17%, the sample size of each test group is 1,100, and the sample size of each control group is 550, with a total sample size of 3,300.

5 Analysis Sets

(1) Immunogenicity Analysis Set

Immunogenicity analyses will be performed primarily based on the Per Protocol Set (PPS), with complementary analyses based on the Full Analysis Set (FAS) and the modified Full Analysis Set (mFAS).

- **Modified Full Analysis Set (mFAS)**

The subject population defined according to intention-to-treat (ITT) principles includes all randomized subjects who have completed the full course of vaccinations, completed blood collection prior to vaccination, and had an effective antibody titer value.

- **Full Analysis Set (FAS)**

The subject population defined according to intention-to-treat (ITT) principles includes all randomized subjects who have completed at least one vaccination, completed blood collection prior to vaccination, and had an effective antibody titer value. Subjects who are administered the vaccine erroneously will be included in the immunogenicity analysis as randomized.

- **Per Protocol Set (PPS)**

It will be a subset of FAS and included all subjects who met the inclusion criteria and did not meet

exclusion criteria, randomized and received full-course vaccination, had a blood sample collected within the time window required by the protocol, and obtained blood sample test results before/after immunization. The subjects who met the following conditions will be excluded from PPS:

1) Those who have significant protocol deviations (affecting immunogenicity evaluation).
2) Those who are vaccinated with the wrong vaccine.
3) Subject received a protocol-prohibited medication/vaccine.

- Other investigational or unregistered products (drugs or vaccines) that are not investigational vaccines.
- Use of influenza vaccines other than investigational vaccines.
- Long-term use (for more than 14 consecutive days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed).
- Immunoglobulins or blood products.

4) Other conditions that may affect the evaluation of immunogenicity of 28 days after full-course vaccination.

Define the above mFAS, FAS, and PPS for each type (including H1N1, H3N2, BV, and BY), respectively.

(2) Safety Set (SS)

The safety set includes all randomized subjects who have received at least one vaccination. In the safety evaluation, subjects with vaccination erroneously will be included in the statistical analysis based on the actual vaccine groups they received, according to the principle of ASaT (All Subjects as Treated).

Safety analysis will be conducted for each dose based on the actual number of vaccinations administered. The safety set for dose 1 includes all subjects who received the first dose of the investigational vaccine, which is denoted as SS1. The safety set for dose 2 includes all subjects who have received the second dose of the investigational vaccine, which is denoted as SS2.

Each analysis set will be discussed and decided by the Principal Investigator, Sponsor, statistician, and data management manager in the blind review meeting before the database is locked.

6 Statistical Considerations

6.1 General Considerations

6.1.1 General Principles

- **Descriptive Statistics**

Unless otherwise specified, the following descriptive statistical summaries will be provided based on variable types:

- ✓ Continuous variables will be statistically described using mean, standard deviation, median, quartile (Q1, Q3), minimum, and maximum values.
- ✓ Categorical or ordinal variables will be summarized using frequencies and percentages, where the number of subjects in the corresponding analysis set will be used as the denominator in the percentage calculations. The 95%CIs for percentages will be calculated using the Clopper-Pearson method.

➢ **Decimal Places**

Unless otherwise specified, the decimal places in the analysis report shall be executed by the following rules:

- ✓ The minimum and maximum values are consistent with the maximum number of decimal places in the original data.
- ✓ The number of decimal places in the mean, median, quartile (Q1, Q3), geometric mean, standard deviation, and 95% confidence interval is one more than the maximum number of decimal places in the original data.
- ✓ Percentages, rates, and rate differences are retained to two decimal places.
- ✓ If P-values≥0.0001 it will be reported to four decimal places. If P-values<0.0001 it will be presented as '< 0.0001'.
- ✓ The test statistics of all statistical tests will be retained to three decimal places.
- ✓ Derived data should be retained to two decimal places.

6.1.2 Derived and Transformed Data

➢ **Baseline**

The last non-missing assessment on or before the first dose of vaccination will be defined as the baseline.

➢ **Date Conversion**

Month=days/30.4375, Year=days/365.25, rounding to 1 decimal place.

➢ **Adverse Event**

Occurrence time of AE (days) = Start date of AE - the corresponding vaccination date

AE duration(days) = End date of AE - Start date of AE + 1

➢ **Treatment Emergent Adverse Event (TEAE)**

TEAEs are defined as adverse events that occur after the first vaccination (including the day of the first vaccination) or worsen after vaccination. Programming is carried out according to the following rules:

- ✧ If the start date of adverse events is after (including simultaneously) the date of the first vaccination, they will be counted as TEAE.
- ✧ If the start date of adverse events is before the date of the first vaccination, they will be counted as non-TEAE.
- ✧ If the start date of adverse event or the date of the first vaccination is missing, making it impossible to determine whether the adverse event occurred after the first vaccination, then such adverse events shall all be counted as TEAE.

➢ **Handling of incorrect vaccination**

- ✧ In the immunogenicity evaluation, subjects will be analyzed according to the vaccine they were randomized to receive, in accordance with the Intention-To-Treat (ITT) principle.
- ✧ In the safety evaluation, subjects will be analyzed according to the vaccine they actually received, in accordance with the All Subjects as Treated (ASaT).

➢ **Relationship of AEs**

- ✧ Related to the study, vaccine means that the relationship between the adverse event and the study vaccine is "possibly related", "probably related", or "definitely related".

- ✧ Unrelated to the study vaccine means that the relationship between the adverse event and the study vaccine is "possibly unrelated" or "definitely unrelated".

➢ **Coding**

AEs, SAEs, and concomitant procedures will be encoded according to version 27.0 of the Medical Dictionary for Regulatory Activities (MedDRA). Concomitant medications/concomitant vaccines will be encoded according to the WHODD_2024-Mar-1 version of the World Health Organization Drug Dictionaries (WHODD).

➢ **Prior and Concomitant Medication**

The determination of prior and concomitant medication uses follows the following rules, respectively:

- ✓ Prior medication, medications with an end date that is before the first vaccination date.
- ✓ Concomitant medication, medications with a start date before the first vaccination date and an end date after any vaccination date, or with a start date after the first vaccination date.
- ✓ When it cannot be determined, the CRF collection shall prevail.

Prior and concomitant procedures determination rule is the same as that of prior and concomitant medications.

6.1.3 Visit Window

For by-visit summaries, data recorded at the nominal visit will be presented. That is, unscheduled measurements will not be included in by visit summaries but might contribute to the baseline timepoint and/or maximum value, where required.

6.1.4 Analysis Software

All analyses will be conducted using SAS version 9.4.

6.1.5 Table and Listing

➢ **Table**

Results will be generally summarized by groups (including test group 1, test group 2, control group 1, and control group 2). Groups are generally presented in the form of columns, and the specific group display will be adjusted according to the actual situation.

➢ **Listing**

Unless otherwise specified, all listings will include group, site, subject number, and the original data will be displayed first. The listing will be generally sorted in sequence by group, site, subject number, visit, or other relevant times (such as the start date of AE).

6.2 Disposition of Subjects

The number of subjects screened, enrolled, and completed the trial for each group, as well as the number of subjects entered into each analysis set, will be summarized separately, and the reasons for subjects who failed to be screened and the reasons for those who withdrew will be analyzed. The list of subjects who failed to be screened, the list of subjects who withdrew, and the list of subjects excluded from each analysis set will be presented separately. A flow chart of the trial will be provided.

6.3 Demographic and Baseline Characteristics

Descriptive statistics will be performed separately for the following demographic data and baseline characteristics, including age(in months), gender, ethnicity, body length/height, weight, and pre-vaccination axillary temperature.

Based on the distributional characteristics of the variables, ANOVA will be used to statistically compare between-group differences in age, body length/height, weight, and pre-vaccination axillary temperature. The Chi-square test or Fisher's exact test will be used to statistically compare between-group differences in gender and ethnicity. Demographic data and baseline characteristics will be listed.

The previous severe medical history and current medical history will be encoded using MedDRA version 26.1 or higher. The number of subjects and incidence rates of previous severe medical history/current medical history will be calculated, respectively, according to the SOC and PT grades, and Fisher's exact test will be used to statistically test the differences in the frequency of previous severe medical history/current medical history between groups. All medical history will be listed.

The above analysis is based on mFAS, FAS, and PPS.

6.4 Compliance

Protocol deviations in each group will be summarized by total and classification. All protocol deviations will be listed.

Descriptive statistics of whether completed each vaccination, whether completed the immunogenicity blood collection will be presented for each group, and the Chi-square test /Fisher's exact test will be used to compare the differences between groups.

Compliance analysis will be based on all randomized subjects.

6.5 Concomitant Medications/Concomitant Vaccines/Concomitant Procedures

Concomitant medications will be encoded using the WHODD_2023-Mar-1 or higher version, and the number of subjects and rate of concomitant medications will be calculated by Anatomic Therapeutic Chemical (ATC) level 2 and Preferred Name (PN). Fisher's exact test will be used to test the differences between groups. Concomitant medications will be listed.

Concomitant vaccines will be encoded using the WHODD_2023-Mar-1 version, and the number of subjects and rate of concomitant vaccines will be summarized by ATC level 4 and preferred names. Fisher's exact test will be used to test the differences between groups. Concomitant vaccines will be listed.

Concomitant procedures will be encoded using the MedDRA 26.1 or higher version, and the number of subjects and rate of concomitant procedures will be calculated according to SOC and PT. Fisher's exact test will be used to test the differences between groups. Concomitant procedures will be listed.

Above concomitant medications/concomitant vaccines/concomitant procedures will be analyzed based on SS, SS1, and SS2.

6.6 Analysis of Intercurrent Events

The intercurrent events of the primary estimand in each group will be summarized, and Fisher's exact

test will be used to compare the between-group differences. A list of intercurrent events will be presented.

The above intercurrent events will be analyzed based on all randomized subjects.

6.7 Study Hypothesis

✧ **Hypothesis test I:** For all subjects aged 6-35 months, 28d after completing of full-course vaccination, the SCR of serum HI antibody against influenza virus H1N1 and H3N2 subtypes in the test group is non-inferior to that in the pooled control group. That is:

$$\text{Null hypothesis } H_{10}: \pi_{iT}-\pi_{iC} < \Delta_1$$

$$\text{Alternative hypothesis } H_{1a}: \pi_{iT}-\pi_{iC} \geqslant \Delta_1$$

Where π_{iT}, π_{iC}represent the post-vaccination SCRs for serotype i (i=1, 2 corresponding to H1N1 and H3N2, respectively) in test group and combined control group. The non-inferiority margin is Δ_1=-10%, and the significance level is one-sided α=0.025.

✧ **Hypothesis test II**: For all subjects aged 6-35 months, 28d after completing of full-course vaccination, the GMT of serum HI antibody against influenza virus H1N1 and H3N2 subtypes in the test group is non-inferior to that in the pooled control group. That is:

$$\text{Null hypothesis } H_{20}: \frac{GMT_{iT}}{GMT_{iC}} < \Delta_2,$$

$$\text{Alternative hypothesis } H_{2a}: \frac{GMT_{iT}}{GMT_{iC}} \geqslant \Delta_2,$$

Where GMT_{iT}, GMT_{iC} represent the post-vaccination GMT for serotype i (i=1, 2 corresponding to H1N1 and H3N2, respectively) in test group and pooled control Group. The non-inferiority margin is Δ_2=2/3, and the significance level is one-sided α=0.025.

✧ **Hypothesis test III**: For all subjects aged 6-35 months, 28d after completing of full-course vaccination, the SCR of serum HI antibody against the influenza virus BV subtype in the test group is non-inferior to that in the combined control group. That is:

$$\text{Null hypothesis } H_{10}: \pi_{iT}-\pi_{iC} < \Delta_1,$$

$$\text{Alternative hypothesis } H_{1a}: \pi_{iT}-\pi_{iC} \geqslant \Delta_1,$$

Where π_{iT}, π_{iC} represent the post-vaccination SCRs for serotype i (i=3) in the test group and control group 1. The non-inferiority margin is Δ_1=-10%, and the significance level is one-sided α=0.025.

✧ **Hypothesis test IV**: For all subjects aged 6-35 months, 28d after completing of full-course vaccination, the GMT of serum HI antibody against influenza virus BV and BY subtypes in the test group is non-inferior to that in the control group 1. That is:

$$\text{Null hypothesis } H_{20}: \frac{GMT_{iT}}{GMT_{iC}} < \Delta_2,$$

$$\text{Alternative hypothesis } H_{2a}: \frac{GMT_{iT}}{GMT_{iC}} \geqslant \Delta_2,$$

Where GMT_{iT}, GMT_{iC} represent the post-vaccination GMT for serotype i (i=3)in the test group and control group 1. The non-inferiority margin is Δ_2=2/3, and the significance level is one-sided α=0.025.

✧ **Hypothesis test V**: For all subjects aged 6-35 months, 28d after completing of full-course

vaccination, the SCR of serum HI antibody against the influenza virus BY subtype in the test group is non-inferior to that in the control group 2. That is:

$$\text{Null hypothesis } H_{10}: \pi_{iT}-\pi_{iC} < \Delta_1,$$

$$\text{Alternative hypothesis } H_{1a}: \pi_{iT}-\pi_{iC} \geqslant \Delta_1,$$

Where π_{iT}, π_{iC}represent the post-vaccination SCRs for serotype i (i=4) in the test group and control group 2. The non-inferiority margin is Δ_1=-10%, and the significance level is one-sided α=0.025.

✧ **Hypothesis test VI:** For all subjects aged 6-35 months, 28d after completing of full-course vaccination, the GMT of serum HI antibody against the influenza virus BY subtype in the test group is non-inferior to that in the control group 2. That is**:**

$$\text{Null hypothesis } H_{20}: \frac{GMT_{iT}}{GMT_{iC}} < \Delta_2,$$

$$\text{Alternative hypothesis } H_{2a}: \frac{GMT_{iT}}{GMT_{iC}} \geqslant \Delta_2,$$

Where GMT_{iT}, GMT_{iC} represent the post-vaccination GMT for serotype i (i=4)in the test group and control group 2. The non-inferiority margin is Δ_2=2/3, and the significance level is one-sided α=0.025.

✧ **Hypothesis test VII:** For all subjects aged 6-35 months, 28d after completing of full-course vaccination, the lower limit of the 95% CI for the SCR of serum HI antibody against influenza virus H1N1, H3N2, BV, and BY subtypes in the test group is ≥30%. That is:

$$\text{Null hypothesis } H_{30}: \pi_{iT} < \pi_1,$$

$$\text{Alternative hypothesis } H_{3a}: \pi_{iT} \geq \pi_1$$

Where π_{iT} represents the post-vaccination SCR for serotype i (i=1, 2 , 3, 4 corresponding to H1N1, H3N2, BV, and BY, respectively) in the test group, π_1=30%, and the significance level is one-sided α=0.025.

✧ **Hypothesis test VIII:** For all subjects aged 6-35 months, 28d after completing of full-course vaccination, the lower limit of the 95% CI for the SPR of serum HI antibody against influenza virus H1N1, H3N2, BV, and BY subtypes in the test group is ≥60%. That is:

$$\text{Null hypothesis } H_{40}: \pi_{iT} < \pi_2,$$

$$\text{Alternative hypothesis } H_{4a}: \pi_{iT} \geq \pi_2$$

Where π_{iT} represents the post-vaccination SPR for serotype i(i=5, 6, 7 8 corresponding to H1N1, H3N2, BV, and BY, respectively) in the test group, π_2=60%, and the significance level is one-sided α=0.025.

6.8 Immunogenicity Evaluation

Immunogenicity analysis will be based on mFAS, PPS, and FAS. The analysis based on the FAS will not take into account the intercurrent events "early discontinuation of treatment due to loss of visits, withdrawal of informed consent, etc., and failure to complete the full-course vaccination". In addition, if the antibody test result is "<1:10", it is counted as "5". If the antibody test result is ">1:10240", it is counted as "10241".

(1) Immunogenicity will be compared between the test group and a pooled trivalent control group of two different sizes for H1N1 and H3N2 types, respectively.

✧**Analysis of post-vaccination SCR**

In the PPS analysis, the 28d post-total vaccination serum HI SCR in all populations, the 28d post-total vaccination serum HI SCR in the susceptible population, and the 28d post-total vaccination serum HI SCR in the non-susceptible population will be calculated in the test group 1, test group 2, and pooled control group under the H1N1 and H3N2 types, respectively, and the two-sided 95%CIs will be calculated using the Clopper-Pearson method. Meanwhile, the Miettinen &Nurminen method will be used to calculate the two-sided 95%CI of the inter-group rate difference, and the Chi-square test/Fisher's exact test will be used for the statistical test of inter-group difference.

In the mFAS and FAS analyses, multiple imputation methods will be used to impute in the missing data in the post-vaccination antibodies, and the 28d post-total vaccination serum HI SCR in all populations, the 28d post-total vaccination serum HI SCR in the susceptible population, and the 28d post-total vaccination serum HI SCR in the non-susceptible population will be calculated for all populations in the test group 1, test group 2, and combined control group under the H1N1 and H3N2 types, respectively based on the imputed datasets. The analysis results of pooled post-imputed datasets will be pooled using Rubin's method to obtain the pooled SCR, the difference in rates between the two groups, and their 95%CIs for each group. If the CI for pooled rate or rate difference is not estimable, the rate or rate difference and its 95%CI will be estimated using the Bootstrap method, which will be sampled 1,000 times with putbacks and seeded = 111.

- ✓ If the lower limit of the two-sided 95%CI for the 28d post-total vaccination serum HI SCR of the test group for this type is ≥30%, this type has reached the absolute standard for SCR.
- ✓ If the lower limit of the two-sided 95%CI of SCR difference for 28d post-total vaccination serum HI antibody between the two groups (test group 1-pooled control group) and (test group 2-pooled control group) for this type is ≥-10%, the post-vaccination SCR of the test group for this type can be considered to be non-inferior to that of the pooled control group. Otherwise, the non-inferiority test is considered to have failed.

✧ **Analysis of post-vaccination antibody SPR**

The post-vaccination antibody SPR in all populations, 28d post-total vaccination serum HI SPR in the susceptible population, and the 28d post-total vaccination serum HI SPR in the non-susceptible population, and the 28d post-total vaccination serum HI SPR in the population of <40 before vaccination, and the 28d post-total vaccination serum HI SPR in the population of ≥40 before vaccination under the H1N1 and H3N2 types in test group 1, test group 2, and pooled control group will be analyzed using the same statistical methods as those used for post-vaccination SPR.

If the lower limit of the two-sided 95%CI for the 28d post-total vaccination serum HI SPR of the test group for this type is ≥60%, this type has reached the absolute standard for SPR.

✧ **GMT and GMI analysis**

In PPS analysis, the covariance analysis model will be employed to analyze the antibodies under H1N1 and H3N2 types, respectively. After logarithmic transformation, the serum HI GMT 28d post-vaccination will be fitted to the covariance analysis model for analysis, with the serum HI GMT 28d post-vaccination after logarithmic transformation as a dependent variable of the model, the logarithmic transformation value of pre-vaccination serum HI antibody GMT as a covariate of the model, and groups will be the fixed effect in the model. The post-vaccination logarithmic transformation value of each group and the least square mean of the difference between groups will be calculated based on the

model. After inverse logarithmic transformation, the post-vaccination antibody corrected geometric mean based on the least square mean of each group, the corrected ratio between groups (test group 1/pooled control group), (test group 2/pooled control group), (test group 1/test group 2), and their 95%CIs will be calculated.

In the mFAS and FAS analyses, the missing data in 28d post-total vaccination serum HI antibody will be imputed using the multiple imputation method, and each imputed dataset will be statistically analyzed using the same covariance analysis model as in the PPS analyses. The analytical results of the imputed datasets will be pooled using Rubin's method and inverse-logarithmically transformed to obtain the corrected GMT for each group, corrected GMT ratio between each pair of groups, and corresponding 95%CIs.

If the lower limit of the two-sided 95%CI for (test group 1/pooled control group) and (test group 2/pooled control group) is ≥2/3, the post-vaccination antibody GMT of the test group for this type is considered to be non-inferior to that of the pooled control group. Otherwise, the non-inferiority test is considered to have failed.

The same method as mentioned above will be adopted to perform a logarithmic transformation on the GMT of serum HI antibody in the susceptible population 28d post-total vaccination, and then fit the covariance analysis model for analysis.

Meanwhile, in the PPS analysis, all population, susceptible population, non-susceptible population, population of <40 before vaccination and population of ≥40 before vaccination under the H1N1 and H3N2 types in the test group 1, test group 2, and pooled control group will be statistically described in terms of HI antibody GMT and GMI, respectively, and the group differences will be statistically examined by using the ANOVA with logarithmic transformation. Meanwhile, the 28d post-total vaccination antibody`s GMT/GMI ratio and corresponding 95%CIs will be calculated for the two groups, and the inter-group difference will be statistically tested by t-test after logarithmic transformation.

In the mFAS and FAS analyses, the missing data in the post-vaccination antibodies will be imputed using the multiple imputation method, and each imputed dataset will be statistically analyzed using the same method as in the PPS analysis. The analytical results of the imputed datasets will be pooled using Rubin's method and inverse-logarithmically transformed to obtain the GMT for each group, the GMI for each group, the GMT ratio between each pair of groups, the GMI ratio between each pair of groups, as well as their corresponding 95%CIs.

The reverse cumulative distribution plots of antibody titers for H1N1 and H3N2 will be generated separately for test group 1, test group 2, and the pooled control group.

(2) The non-inferiority comparison will be performed for the BV serotype between the test group and the control group

✧ **Analysis of post-vaccination SCR**

In the PPS analysis, the 28d post-total vaccination serum HI SCR in all populations, the 28d post-total vaccination serum HI SCR in the susceptible population, and the 28d post-total vaccination serum HI SCR in the non-susceptible population will be calculated in test group 1, test group 2, control group 1, and control group 2 under the BV type, respectively, and the two-sided 95%CIs will be calculated using the Clopper-Pearson method. Meanwhile, the Miettinen &Nurminen method will

be used to calculate the two-sided 95%CI of the inter-group rate difference, and the Chi-square test/Fisher's exact test will be used for the statistical test of inter-group difference.

In the mFAS and FAS analyses, multiple imputation methods will be used to impute the missing data in the post-vaccination antibodies, and the 28d post-total vaccination serum HI SCR in all populations, the 28d post-total vaccination serum HI SCR in the susceptible population, and the 28d post-total vaccination serum HI SCR in the non-susceptible population will be calculated for all populations in test group 1, test group 2, control group 1, and control group 2 under the BV type, respectively, based on the imputed datasets. The analysis results of pooled post-imputed datasets will be pooled using Rubin's method to obtain the pooled SCR, the difference in rates between the two groups, and their 95%CIs for each group. If the CI for pooled rate or rate difference is not estimable, the rate or rate difference and its 95%CI will be estimated using the Bootstrap method, which will be sampled 1,000 times with putbacks and seeded = 111.

- ✓ If the lower limit of the two-sided 95%CI for the 28d post-total vaccination serum HI SCR of the test group for this type is ≥30%, this type has reached the absolute standard for SCR.
- ✓ If the lower limit of the two-sided 95%CI of SCR difference for 28d post-total vaccination serum HI antibody between the two groups (test group 1-control group 1) and (test group 2-control group 1) for this type is ≥-10%, the post-vaccination SCR of the test group for this type can be considered to be non-inferior to that of the control group 1. Otherwise, the non-inferiority test is considered to have failed.

✧ **Analysis of post-vaccination antibody SPR**

The post-vaccination antibody SPR in all populations, 28d post-total vaccination serum HI SPR in the susceptible population, and the 28d post-total vaccination serum HI SPR in the non-susceptible population, and the 28d post-total vaccination serum HI SPR in the population of <40 before vaccination, and the 28d post-total vaccination serum HI SPR in the population of ≥40 before vaccination under the H1N1 and H3N2 types in test group 1, test group 2, control group 1 and control group 2 will be analyzed using the same statistical methods as those used for post-vaccination SPR.

If the lower limit of the two-sided 95%CI for the 28d post-total vaccination serum HI SPR of the test group for this type is ≥60%, this type has reached the absolute standard for SPR.

✧ **GMT and GMI analysis**

In PPS analysis, the covariance analysis model will be employed to analyze the antibodies under the BV type in the test group 1, test group 2, control group 1, and control group 2, respectively. After logarithmic transformation, the serum HI GMT 28d post-vaccination will be fitted to the covariance analysis model for analysis, with the serum HI GMT 28d post-vaccination after logarithmic transformation as a dependent variable of the model, the logarithmic transformation value of pre-vaccination serum HI antibody GMT as a covariate of the model, and groups will be the fixed effect in the model. The post-vaccination logarithmic transformation value of each group and the least square mean of the difference between groups will be calculated based on the model. After inverse logarithmic transformation, the post-vaccination antibody corrected geometric mean based on the least square mean of each group, the corrected ratio between groups (test group 1/control group 1), (test group 2/control group 1), (test group 1/control group 2), (test group 2/control group 2) and (test group 1/test group 2) and their 95%CIs will be calculated.

In the mFAS and FAS analyses, the missing data in 28d post-total vaccination serum HI antibody will

be imputed using the multiple imputation method, and each imputed dataset will be statistically analyzed using the same covariance analysis model as in the PPS analyses. The analytical results of the imputed datasets will be pooled using Rubin's method and inverse-logarithmically transformed to obtain the corrected GMT for each group, the corrected GMT ratios between each pair of groups, and corresponding 95%CIs.

If the lower limit of the two-sided 95%CI for (test group 1/control group 1) and (test group 2/control group 1) is ≥2/3, the post-vaccination antibody GMT of the test group for this type is considered to be non-inferior to that of the pooled control group. Otherwise, the non-inferiority test is considered to have failed.

Meanwhile, in the PPS analysis, all population, susceptible population, non-susceptible population, population of <40 before vaccination and population of ≥40 before vaccination under the BV type in the test group 1, test group 2, control group 1 and control group 2 will be statistically described in terms of HI antibody GMT and GMI, respectively, and the group differences will be statistically examined by using the ANOVA with logarithmic transformation. Meanwhile, the 28d post-total vaccination antibody`s GMT/GMI ratio and corresponding 95%CIs will be calculated for the two groups, and the inter-group difference will be statistically tested by t-test after logarithmic transformation.

In the mFAS and FAS analyses, the missing data in the post-vaccination antibodies will be imputed using the multiple imputation method, and each imputed dataset will be statistically analyzed using the same method as in the PPS analysis. The analytical results of the imputed datasets will be pooled using Rubin's method and inverse-logarithmically transformed to obtain the GMT for each group, the GMI for each group, the GMT ratio between each pair of groups, the GMI ratio between each pair of groups, as well as their corresponding 95%CIs.

The reverse cumulative distribution of antibody titers for BV will be generated separately for test group 1, test group 2, control group 1, and control group 2.

(3) An intergroup comparison of non-inferiority for BY is made between test groups and control groups.

Immunogenicity will be compared between the test group (test group 1 or test group 2) and control group 2 for BY types, respectively, using the same method as for BV types.

Forest plots of the SCR, SPR, and GMT of HI antibody of each serotype in all populations 28d post-total vaccination will be plotted.

Sensitivity Analysis: In mFAS analysis, the missing data in the serum HI antibody and SCR of HI antibody 28d post-total vaccination will be analyzed for sensitivity analysis by using the Tipping Point Analysis (TPA) and the Jump to Reference (J2R) method based on δ adjustment. Evaluate the robustness impact of missing data on the test results. For details, please refer to Section 6.10 on the handling of Missing Data.

6.9 Safety Evaluation

The following safety evaluation will be performed based on SS.

6.9.1 Adverse Event

Adverse events (AEs) and SAEs will be medically coded with MedDRA version 26.1 or higher, and classified and statistically analyzed according to SOC and PT levels. In addition, solicited adverse events (inoculated sites (local) adverse events and non-inoculated sites (systemic) adverse events) will be classified and statistically analyzed per the provisions of the protocol. This trial will mainly be to make a statistical analysis of the TEAE, and list the AEs before vaccination in the form of a list.

This trial mainly conducts statistical analysis on adverse events that occur during vaccination, including those that occur within 28 days after each dose of vaccination. Unless otherwise specified, all adverse events mentioned below are those that occurred during the vaccination period.

The number of event, number of subjects, and incidence of the following AEs will be calculated by each group and the total. Fisher's exact test will be used to statistically test the differences between groups:

- All AEs.
  - AEs related to the investigational vaccine.
  - AEs unrelated to the investigational vaccine.
- AEs with an incidence rate ≥10%.
  - AEs related to the investigational vaccine with an incidence rate ≥10%.
- AEs with an incidence rate ≥1%.
  - AEs related to the investigational vaccine with an incidence rate ≥1%.
- AEs of different severity.
  - AEs related to the investigational vaccine of different severities.
  - AEs unrelated to the investigational vaccine of different severities.
- AEs of different doses.
  - AEs related to the investigational vaccine of different doses.
  - AEs unrelated to the investigational vaccine of different doses.
- AEs that occur within 30 minutes, 0-7 days, and 0-28 days.
  - AEs related to the investigational vaccine that occur within 30 minutes, 0-7 days, and 0-28 days.
- AEs unrelated to the investigational vaccine that occur within 30 minutes, 0-7 days, and 0-28 days.
- AEs leading to dropout.
  - AEs related to the investigational vaccine leading to dropout.

Meanwhile, the Kaplan-Meier method will be used to calculate the median duration of AEs and their 95% confidence intervals in each group, respectively (using the Brookmeyer & Crowley method of log-log transformation), and the log-rank test will be used to statistically test the differences between groups. The AEs after each dose of vaccination will be statistically analyzed respectively. The analysis of AEs for each dose will be based on the safety datasets of each dose.

Present the forest plots to compare the differences in the incidence rates of AEs between the two groups (test group 1 - control group 1), (test group 1 - control group 2), (test group 2 - control group 1), and (test group 2 - control group 1) according to the top ten preferred terms for incidence rates in the vaccine group.

When calculating the incidence of AEs, if the same AE occurs multiple times in the same subject, it is recorded as one occurrence. When calculating the number of events of AEs, if the same subject experiences the same AE multiple times, it is recorded multiple times. When analyzing the severity and correlation to the investigational vaccine of AEs, if the same AE occurs multiple times in the same subject, the most severe or the most relevant AE to the investigational vaccine should be selected for analysis.
Display the list of AEs related to the investigational vaccine, the list of AEs unrelated to the investigational vaccine, the list of AEs leading to drop out, and the list of Grade 3 or above AEs.

6.9.2 Serious Adverse Event

This trial mainly conducted statistical analysis on SAEs that occurred after the first dose of vaccination, including those that occurred within 6 months from the first dose of vaccination to the full course of vaccination.
The number of events, number of subjects, and incidence rates of the following types of serious adverse events will be calculated, respectively, by total and for each group. Fisher's exact test will be used to test the differences between groups.
The number, number of cases and incidence of the following SAEs will be calculated by each group and the total. Fisher's exact test will be used to statistically test the differences between groups:

- All SAEs.
- SAEs related to the investigational vaccine.
- SAEs unrelated to the investigational vaccine.

List all SAEs.

6.9.3 Adverse Events of Special Interest

This trial mainly conducted statistical analysis on AESIs that occurred after the first dose of vaccination, including events observed from the first dose until 6 months after the full vaccination. (including GBS, seizure, encephalitis, myelitis, neuritis, Bell's palsy, vasculitis, and thrombocytopenia).
The number of event, number of subjects, and incidence of the following AESIs will be calculated by each group and the total. Fisher's exact test will be used to statistically test the differences between groups:

- All AESIs.
- AESIs related to the investigational vaccine.
- AESIs unrelated to the investigational vaccine.

List all AESIs.

6.10 Handling of Missing Data

In mFAS and FAS set analyses, for missing data in the primary estimand under the assumption of Missing at Random, the Multiple Imputation method will be applied to handle missing data before non-inferiority assessment. The imputation will be performed 10 times, with the random seed set to seed=111222333.

For each imputed complete dataset, the following endpoints will be calculated at 28d post-total vaccination for each serotype:

- SCR
- SPR
- GMT (Geometric Mean Titer)
- GMI (Geometric Mean Increase)

The results from all imputed datasets will then be pooled using Rubin's rules to obtain pooled estimates for SCRs, SPRs, GMTs, and GMIs.

Additionally, for the primary estimand in the mFAS analysis set, sensitivity analyses will be conducted under the assumption of Missing Not at Random to evaluate the robustness of the results. The following methods will be applied:

- TPA (Tipping Point Analysis) method
- δ-adjusted J2R (Jump to Reference) method

Sensitivity Analysis 1: To evaluate the impact of missing values on the primary estimand, TPA will be used for the sensitivity analysis.

- ✓ For the continuous variable, HI antibody, the TPA method will gradually adjust the possible values of the missing data of HI antibody titers of each serotype after logarithmic conversion, 28d post-total vaccination in each group, to find the critical point that reverses the conclusion of GMT analysis. Among them, the smaller the absolute value of the critical point is, the less impact the missing data has on the trial conclusion, and the better the robustness of the trial conclusion is. The Rubin method will be adopted to combine the analysis results of the datasets after each imputation, and the GMT and its 95% confidence interval of each group, as well as the corrected GMT ratio and its 95% confidence interval between each pair of groups (test group 1 / pooled control group, test group 2 / pooled control group) will be obtained through inverse logarithmic transformation. Among them, the number of seeds for multiple imputation will be taken as seed=111222333, and impute 10 times each time.
- ✓ For the categorical veriable, SCR, let the number of subjects with missing HI antibody titers after 28d post-total vaccination in test group 1 that seroconverted in all analysis sets after multiple imputation in main analysis be denoted as n1, and the number of subjects with missing HI antibody titers 28d post-total vaccination in the pooled control group that did not seroconvert in all analysis sets after multiple imputation in main analysis be denoted as n2. The critical point analysis method will sequentially reverse the seroconverted subjects in test group 1 from 0, 1, 2,..., ni (i = 1, 2) to non-seroconverted, and reverse the non-seroconverted subjects in the pooled control group to seroconverted, to find the critical point that reverses the conclusion of the SCR analysis. The Rubin method will be used to combine the analysis results of each imputed dataset to obtain the pooled SCR, the rate difference between each pair of groups, and their 95% confidence intervals. The seed number for multiple imputations is set as seed = 111222333, and each imputation is performed 10 times. If the confidence interval of the pooled rate or rate difference cannot be estimated, the Bootstrap method is used to estimate the rate or rate difference and their 95% confidence intervals. Bootstrap will perform 1000 times of sampling with replacement, with the seed number set as seed = 111.

In addition, a critical point analysis plot will be drawn to provide a visual basis for result determination.

Using the same method to conduct a sensitivity analysis for test group 2 and the pooled control group.

Sensitivity Analysis 2: To evaluate the impact of missing values on the primary estimand, the δ-adjusted J2R method will be used for sensitivity analysis.

- ✓ For the continuous variable, HI antibody, multiple imputation based on the distribution $N(\bar{X} - 0.176, {S_1}^2)$ will be performed for missing data of HI antibody titers of each serotype after logarithmic conversion, 28d post-total vaccination in test group 1. Multiple imputation based on the distribution $N(\bar{X}, {S_2}^2)$ will be performed for missing data of HI antibody titers of each serotype after logarithmic conversion, 28d post-total vaccination in the pooled control group. Here, $\bar{X}$ represents the mean of non-missing log-transformed 28d post-total vaccination HI antibody titers calculated from the pooled control group, S_1 is the standard deviation is calculated from non-missing data in test group 1, and S_2 is the standard deviation is calculated from non-missing data in the pooled control group. The Rubin method will be adopted to combine the analysis results of the datasets after each imputation, and the GMT and its 95% confidence interval of each group, as well as the corrected GMT ratio and its 95% confidence interval between each pair of groups (test group 1 / pooled control group) will be obtained through inverse logarithmic transformation. Among them, the number of seeds for multiple imputation will be taken as seed=111222444, and impute 10 times each time.
- ✓ For the categorical variable, SCR, multiple imputation will be performed based on the Bernoulli(p_1-0.1) distribution, where p_1 is the SCR calculated from non-missing data in the pooled control group. The Rubin method will be used to combine the analysis results of each imputed dataset to obtain the pooled SCR, the rate difference between each pair of groups, and their 95% confidence intervals. The seed number for multiple imputations is set as seed = 111222555, and each imputation is performed 10 times. If the confidence interval of the pooled rate or rate difference cannot be estimated, the Bootstrap method is used to estimate the rate or rate difference and their 95% confidence intervals. Bootstrap will perform 1000 times of sampling with replacement, with the seed number set as seed = 222.

Using the same method to conduct a sensitivity analysis for test group 2 and the pooled control group.

The same method as mentioned above will be used to conduct a sensitivity analysis for the non-inferiority comparison between the BV type and BY type test groups and the control group.

This trial will no longer process the missing data in other immunogenicity endpoints and safety endpoints.

6.11 Multiplicity

[redacted]

[redacted]

VERSION HISTORY

Version	Date	Author	Notes
V1.0	2024-06-18	[redacted]	Original Version