Statistical Analysis Plan

A Phase III, observer-blind, randomized, multi-country study to evaluate the lot-to-lot consistency of GSK's investigational RSV maternal vaccine and the immune response, safety and reactogenicity of RSV maternal vaccine when co-administered with GSK's quadrivalent influenza D-QIV vaccine in healthy non-pregnant women 18-49 years of age.

09-JUN-2022

Information Type:	Statistical Analysis Plan (SAP)

TITLE PAGE

Protocol Title: A Phase III, randomized, multi-country study to evaluate the lot-to-lot consistency of GSK's investigational RSV maternal vaccine and the immune response, safety and reactogenicity of RSV maternal vaccine when co-administered with GSK's quadrivalent influenza D-QIV vaccine in healthy non-pregnant women 18-49 years of age.

Study Number: 214709 (RSV MAT-010)

Compound Number: GSK3888550A

Abbreviated Title: A Phase III study to assess the lot-to-lot consistency of GSK's investigational RSV maternal vaccine and the immune response and safety of RSV maternal vaccine when given alone or co-administered with GSK's influenza D-QIV vaccine in healthy non-pregnant women.

Sponsor Name: GlaxoSmithKline Biologicals SA (GSK)

Regulatory Agency Identifier Number(s): IND 018434

Registry	**ID**
Clintrials.gov	NCT05045144

TABLE OF CONTENTS

PAGE

LIST OF TABLES

PAGE

Version history

SAP Version	Approval Date	Protocol Version (Date) on which SAP is Based	Change	Rationale
SAP	29 Jun 2021	Amendment 1 (27 May 2021)	Not Applicable	Original version
Amendment 1	29 Mar 2022	Amendment 2 (17 November 2021)	1. Blinding strategy detail from the study title has been deleted, as they vary between the lot to lot and co-administration groups. 2. Interim analysis has been removed. 3. Components updated with 2021/22 season Flu strains.	1. It was identified that the intervention type amongst the co-administration groups can be determined by the blinded site and sponsor staff using available information in the electronic case report forms (eCRF), therefore the blinding strategy was adapted to single-blind design for the co-administration groups. 2. The decision was made to not go forward with the BLA filing for the RSV Maternal vaccine, and as a result, the interim analysis was removed. 3. New information made available on the Flu strains.
Amendment 2	09 Jun 2022	Amendment 3 (26 May 2022)	1. The primary and secondary objectives were modified to evaluate only 3 influenza strains. The A/Victoria/2570/2019 (H1N1) strain will be evaluated as part of the tertiary objectives. 2. The secondary safety objective and its corresponding endpoint related to assessment of safety of RSV maternal vaccine when given alone and co-administered with Flu D-QIV from vaccination up to study end was recategorized as a primary objective. 3. Further clarified the analysis details for COVID infection.	1. Updated due to issues in validation of A/Victoria/2570/2019 (H1N1) influenza strain. 2. Updated to reflect the change to study's safety assessment plan. 3. A participant is defined as having a suspected, probable or confirmed COVID-19 infection during the first 30 days follow-up period, and after that as having or not having the infection until the study end.

1. INTRODUCTION

The purpose of this SAP is to describe the planned analyses for RSV MAT-010 (214709). Details of the planned analysis are provided.

1.1. Objectives, Estimands and Endpoints

Table 1 Study objectives, endpoints and estimands

Objectives	Endpoints/Estimands
Primary	
Safety • To evaluate the safety and reactogenicity of RSVPreF3 when given alone (pooled lots) or co-administered with Flu D-QIV up to study end.	The number and percentage of participants in each group reporting • Each solicited administration site AE collected during the 7 days follow-up period (Day 1 to Day 7) • Each solicited systemic events in the 7 days follow-up period • Unsolicited AEs collected during the 30 days follow-up period (Day 1 to Day 30) • SAEs in the 30 days follow-up period (Day 1 to Day 30) • SAEs in the 180 days follow-up period (Day 1 to Day 181)
Immunogenicity • To demonstrate the lot-to-lot consistency of 3 lots of the investigational RSVPreF3 vaccine based on GMC of RSVPreF3 IgG ELISA at Day 31	RSVPreF3 IgG ELISA concentration at Day 31 (30 days post administration) Measured by ratio of Geometric mean concentration (GMC) between lots in terms of RSVPreF3 IgG ELISA titers at Day 31 (30 days post administration)
• To demonstrate non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine compared to Flu D-QIV given alone based on Geometric mean titer (GMT) of Flu D-QIV antibody titers against 3 influenza strains at Day 31 post administration	Flu D-QIV antibody titers against 3 influenza strains at Day 31 (30 days post administration) Measured by ratio of Haemagglutinin inhibition (HI) GMT between group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and Flu D-QIV vaccine alone group against 3 strains at Day 31 (30 days post administration)
Secondary	
*Confirmatory Immunogenicity***	
• To demonstrate the non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV compared to given alone based on GMT of RSV A neutralizing antibody at Day 31 post administration	RSV A neutralizing antibody titers at Day 31 (30 days post administration) Measured by ratio of GMT between group of RSVPreF3 vaccine co-administered with Flu D-QIV and RSVPreF3 vaccine alone (pooled lot 1,2,3) group in term of RSV A neutralizing antibody titers at Day 31 (30 days administration)
• To demonstrate the non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 compared to given alone based on Seroconversion rate (SCR*) of Flu D-QIV HI antibody titers against 3 influenza strains at Day 31 post study intervention	• Seroconversion rate to Flu D-QIV HI antibody titers against the 3 influenza strains at Day 31 Measured by the difference of proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with

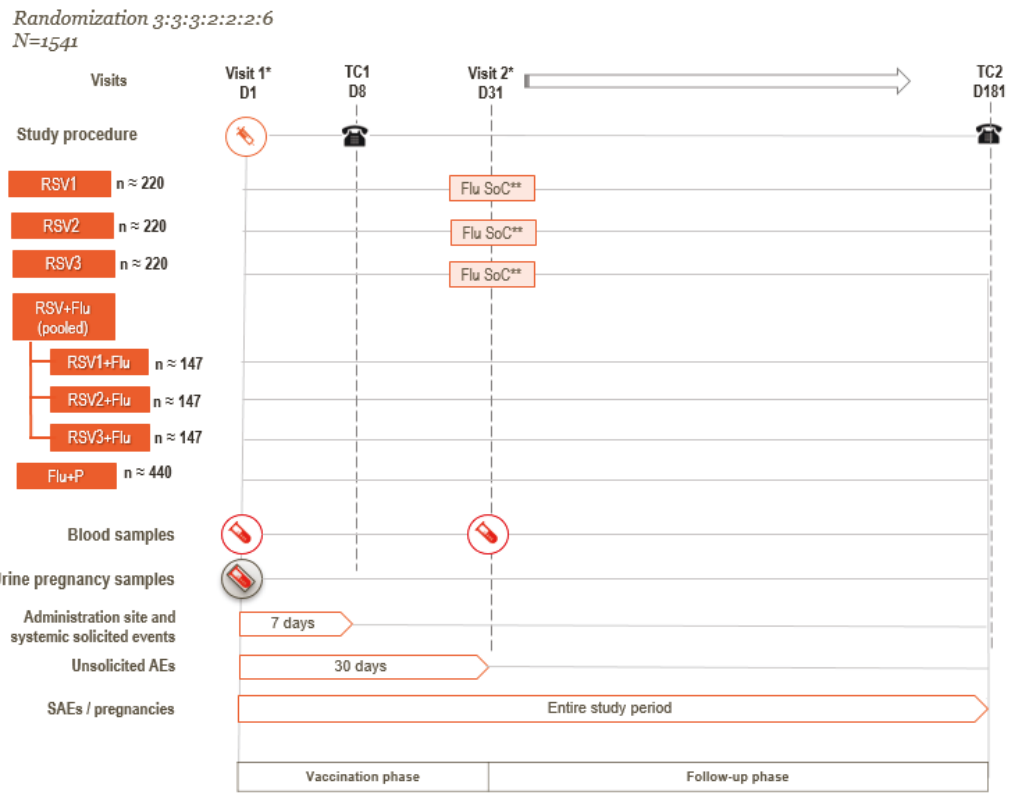
Objectives	Endpoints/Estimands
	RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
Descriptive Immunogenicity	
• To evaluate the humoral immune response of RSVPreF3 vaccine when given alone and co-administered with Flu D-QIV in terms of RSV A, RSV B neutralizing antibody and RSVPreF3 IgG concentration at Day 1 (prior to the vaccination) and Day 31 (post study intervention).	• RSV A, RSV B neutralizing antibody titers and RSV PreF3 IgG concentration at Day 1 and Day 31 GMT of RSV A and RSV B neutralizing antibody, GMC of RSVPreF3 IgG at Day 1 and Day 31, in the group of RSVPreF3 vaccine co-administered with Flu D-QIV and the group of RSVPreF3 vaccine alone
• To evaluate the humoral immune response to the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine in terms of antibody titers against 3 influenza strains at Day 1 and Day 31	Flu D-QIVHI antibody titers against the 3 influenza strains at Day 1 and Day 31 HI GMT in the 3 influenza strains at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
• To evaluate seroprotection rate (SPR) and Seroconversion rate (SCR)* of the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine	Seroprotection rate to Flu D-QIV HI antibody titers against the 3 influenza strains at Day 1 and Day 31 Measured by the proportion of participants achieving an HI antibody titer ≥1:40 at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone • Seroconversion rate to Flu D-QIV HI antibody titers against the 3 influenza strains at Day 31 Measured by the proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
• To evaluate the humoral immune response in 3 individual lots of RSVPreF3	• RSV A, RSV B neutralizing antibody titers and RSVPreF3 IgG antibody concentrations at pre-vaccination and Day 31 Geometric mean titer/concentration (GMT/GMC) in term of RSV A, RSV B neutralizing antibody and RSVPreF3 IgG antibody at Day 1 (pre-vaccination) and Day 31 in each of 3 lots of the investigational PreF3 vaccine.
Tertiary	
To demonstrate non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine compared to Flu D-QIV given alone based on Geometric mean titer (GMT) of Flu D-QIV antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 31 post administration	Flu D-QIV antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 31 (30 days post administration) Measured by ratio of Haemagglutinin inhibition (HI) GMT between group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and Flu D-QIV vaccine alone group against the A/Victoria/2570/2019 (H1N1) strain at Day 31 (30 days post administration)
To demonstrate the non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 compared to given alone based on Seroconversion rate (SCR) of Flu D-QIV HI antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 31 post study intervention	• Seroconversion rate to Flu D-QIV HI antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 31

Objectives	Endpoints/Estimands
	Measured by the difference of proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
To evaluate the humoral immune response to the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine in terms of antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 1 and Day 31	Flu D-QIV HI antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 1 and Day 31 HI GMT in the A/Victoria/2570/2019 (H1N1) influenza strain at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
To evaluate seroprotection rate (SPR) and Seroconversion rate (SCR) of the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine	• Seroprotection rate to Flu D-QIV HI antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 1 and Day 31 Measured by the proportion of participants achieving an HI antibody titer ≥1:40 at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone • Seroconversion rate to Flu D-QIV HI antibody titers against the A/Victoria/2570/2019 (H1N1) influenza strain at Day 31 Measured by the proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
CCI [redacted]	

* The SCR is defined as the proportion of participants with:
A Day 1 (pre-vaccination) serum anti-HI titer <1:10 and a Day 31 (post-vaccination) serum anti-HI titer ≥1:40, or
A Day 1 (pre-vaccination) serum anti-HI titer ≥ 1:10 and a fold increase (post/pre) ≥ 4 at Day 31
** Two confirmatory objectives will be tested sequentially.

1.2. Study Design

Overview of Study Design and Key Features



SOC=Standard of Care
* Screening and vaccination will happen on Visit 1(Day 1). The blood sample collection must be collected before administering the vaccine(s).
** Visit 2 Flu D-QIV vaccinations are not part of experimental design. It serves as an optional vaccination for the participant to decide if they would like to get the vaccination as part of their standard of care. The blood sample collection must be collected before administering the Flu D-QIV vaccine.

Design Features	• Study Phase and Population: Phase III, Non-pregnant women • Design type: Randomized, Parallel, multi-country study • Control type: Placebo controlled, Saline (NaCl) solution • Study duration: The study vaccine will be administered at Visit 1 and the participant will be followed for 6 months until the study conclusion. • Blinding: This study will utilize an observer-blind manner to evaluate the lot-to-lot consistency of GSK RSVPreF3 vaccine and single-blind manner to evaluate the immune response, safety and reactogenicity of RSV maternal vaccine when co-administered with GSK quadrivalent influenza. Detailed blinding design is described in Protocol Section 6.3.5

Overview of Study Design and Key Features	
Study intervention	Study (intervention) groups are described in Table 2
Study intervention Assignment	Approximately 1541 participants will be randomized into 7 study groups (5 main study groups including 1 pooled group [RSV1+Flu, RSV2+Flu, RSV3+Flu]) with randomization ratio of RSV1:RSV2:RSV3: RSV1+Flu: RSV2+Flu: RSV3+Flu: Flu+P as 3:3:3:2:2:2:6. An automated internet based system (SBIR) will be used to randomly allocate a study group and treatment number to each participant. Age (18-32 vs. 33-49) and center will be minimization factors. Minimization factors will have equal weight in the algorithm.

Table 2 Study groups and intervention

Study groups	Approximate number of participants	Age (Min-Max)	Study intervention	Standard of Care Vaccination*
RSV1	220	18 – 49 years	RSV MAT 120 (Lot1)	Flu D-QIV
RSV2	220	18 – 49 years	RSV MAT 120 (Lot2)	Flu D-QIV
RSV3	220	18 – 49 years	RSV MAT 120 (Lot3)	Flu D-QIV
RSV1+Flu	147	18 – 49 years	RSV MAT 120 (Lot 1) Flu D-QIV	
RSV2+Flu	147	18 – 49 years	RSV MAT 120 (Lot 2) Flu D-QIV	
RSV3+Flu	147	18 – 49 years	RSV MAT 120 (Lot 3) Flu D-QIV	
Flu+P	440	18 – 49 years	Flu D-QIV placebo	

RSV MAT 120 =RSVPreF3 vaccine
*Standard of care vaccine administered only to ensure the participants receive the standard of care.

2. STATISTICAL HYPOTHESES

The study includes 2 confirmatory co-primary objectives:

- To demonstrate the lot-to-lot consistency of 3 lots of RSVPreF3 vaccine in terms of geometric mean concentration (GMC) of RSVPreF3 IgG ELISA at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: at least one of the two-sided 95% confidence intervals for the 3 pair-wise GMC ratios between 2 lots are outside the range of 0.67 to 1.5

  H_a: $0.67 < \mu_{lot1}/\mu_{lot2} < 1.5$ and $0.67 < \mu_{lot1}/\mu_{lot3} < 1.5$ and $0.67 < \mu_{lot2}/\mu_{lot3} < 1.5$

  where μ represents the GMC of RSVPreF3 IgG at Day 31 from each lot. The lot-to-lot consistency will be demonstrated if two-sided 95% confidence intervals for the 3 pair-wise GMC ratios at Day 31 falls within 0.67 and 1.5.

- To demonstrate the non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone in terms of geometric mean titer (GMT) of Flu D-QIV HI antibody against the 3 influenza strains (A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019, and B/Phuket/3073/2013) at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $\mu_{\text{co-ad group}}/\ \mu_{\text{alone}} \leq 0.67$ vs H_a: $\mu_{\text{co-ad group}}/\mu_{\text{alone}} > 0.67$

  where μ represents the GMT of Flu D-QIV HI antibody at Day 31. The non-inferiority will be demonstrated if the lower limits of the two-sided 95% CI on the GMT ratio (co-ad group RSV+Flu with 3 lots pooled divided by Flu+P group) is greater than 0.67 at Day 31 post vaccination for all the 3 strains (A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019, and B/Phuket/3073/2013).

In addition, the study also includes 2 confirmatory secondary objectives. These hypothesis tests will be conducted sequentially only if the primary objective of non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone is met.

- To demonstrate the non-inferiority of RSVPreF3 vaccine when co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of GMT of RSV A neutralizing antibody at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $\mu_{\text{co-ad group}}/\ \mu_{\text{alone}} \leq 0.67$ vs H_a: $\mu_{\text{co-ad group}}/\mu_{\text{alone}} > 0.67$

  where μ represents the GMT of RSV A neutralizing antibody at Day 31. The non-inferiority will be demonstrated if the lower limits of the two-sided 95% CI on the GMT ratio (co-ad group RSV+Flu divided by RSV alone group with 3 lots pooled in both groups) is greater than 0.67 at Day 31 post vaccination.

- To demonstrate the non-inferiority for each strain (A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019, and B/Phuket/3073/2013) of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone in terms of seroconversion rate (SCR) at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $p_{\text{alone}} - p_{\text{co-ad group}} \geq 10\%$ vs H_a: $p_{\text{alone}} - p_{\text{co-ad group}} < 10\%$

  where p represents the SCR for each influenza strain at Day 31. The non-inferiority for each strain will be demonstrated if the upper limits of the two-sided 95% CI on the SCR difference (Flu+P group minus co-ad group RSV+Flu with 3 lots pooled) is less than 10% at Day 31 post vaccination.

2.1. Multiplicity Adjustment

To ensure the global type I error for the primary objectives is controlled at 5%, each of the primary objectives will be tested using a nominal one-sided alpha of 2.5%.

If the primary objective of non-inferiority of Flu D-QIV in terms of GMT is met, the secondary confirmatory objective of non-inferiority of RSVPreF3 vaccine in terms of GMC will be tested at alpha of 2.5%, if this objective is met, the non-inferiority of Flu D-QIV in terms of SCR will be tested at alpha of 2.5% for each strain, otherwise, it will not be tested.

3. ANALYSIS SETS

Table 3 Analysis sets

Analysis Set	Definition / Criteria	Analyses Evaluated
Enrolled	Participants who agreed to participate in a clinical study after completion of the informed consent process	Study Population
Exposed	All Participants who received at least 1 dose of the study treatment. The allocation in a group is done as a function of the administered treatment	Safety, Study Population
Full Analysis Set (FAS)	All Participants who received at least 1 dose of the study intervention and have post-vaccination immunogenicity data It will be defined by timepoint, e,g. FAS at Day 31	Immunogenicity
Per-Protocol Set (PPS)	All Participants who received at least 1 dose of the study treatment to which they are randomized and have post-vaccination data minus participants with protocol deviations that lead to exclusion It will be defined by timepoint, e,g. PPS at Day 31	Immunogenicity
Solicited Safety Set	All Participants who received at least 1 dose of the study intervention (Exposed Set) who have solicited safety data	Solicited events
Randomized Set	Randomized set will include all participants who are randomized. The allocation in a group is done as function of the randomized intervention. Please note this set was not included in the protocol, but will be used later in one summary analysis, so it is added here for clarification.	Disposition

3.1. Criteria for eliminating data from Analysis Sets

Elimination codes are used to identify participants to be eliminated from analysis. Detail is provided below for each set.

3.1.1. Elimination from Exposed Set (ES)

Code 1030 (Study vaccine not administered at all), 800 (Fraudulent data) and code 900 (invalid informed consent) will be used for identifying participants eliminated from ES

3.1.2. Elimination from Full Analysis Set (FAS)

A participant will be excluded from the FAS under the following conditions

Table 4 Elimination code and condition

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
800	Fraudulent data	All	All
900	Invalid informed consent	All	All
1030	Study vaccine not administered at all	All	Safety, immunogenicity
2100.Vx	Serological results not available post-vaccination	Visit 2/Day 31	Immunogenicity

Vx indicates participants whose immunogenicity data will be eliminated from a specific visit.

3.1.3. Elimination from Per-protocol Set (PPS)

A participant will be excluded from the PPS under the following conditions

Table 5 Elimination code and condition

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
800	Fraudulent data	All	All
900	Invalid informed consent	All	All
1030	Study vaccine not administered at all	All	Safety, immunogenicity
1040.Vx*	Administration of concomitant vaccine(s) forbidden in the protocol	Visit 2/Day 31	Immunogenicity
1050	Randomisation failure	All	Immunogenicity
1060	Randomisation code was broken	All	Immunogenicity

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
1070**	Subjects got vaccinated with the correct vaccine but containing an incorrect volume	All	Immunogenicity
1070**	Vaccination not according to protocol (site of injection, route of administration, wrong replacement of study treatment administered)	All	Immunogenicity
1070**	Study treatment not prepared as per protocol (e.g., reconstitution)	All	Immunogenicity
1070**	Other deviations related to wrong study treatment/administration/dose	All	Immunogenicity
1070**	Study treatment administered while contraindication	All	Immunogenicity
1080	Vaccine temperature deviation	All	Immunogenicity
1090	Expired vaccine administered	All	Immunogenicity
2010	Protocol violation (inclusion/exclusion criteria) DOB – VAC – 18-49 years BMI <=40 kg/m^2	All	Immunogenicity
2040.Vx*	Administration of any medication forbidden by the protocol	Visit 2/Day 31	Immunogenicity
2050.Vx*	Intercurrent medical conditions which are exclusionary as per protocol	Visit 2/Day 31	Immunogenicity
2090.Vx	Subjects did not comply with blood sample schedule: • For PPS at Day 31, check the interval from vaccination to day 31 BS = 31 – 38 days;	Visit 2/Day 31	Immunogenicity
2100.Vx	Serological results not available post-vaccination	Visit 2/Day 31	Immunogenicity
2120.Vx	Obvious incoherence or abnormality or error in data	All	Immunogenicity
2130.Vx	Testing performed on samples not aligned with ICF	All	Immunogenicity

*Attribution of these elimination codes to subject need CRDL review of individual listing
** Attribution of code 1070 to a subject requires CRDL confirmation
Vx+ indicates participants whose immunogenicity data will be eliminated from a specific visit onwards; Vx indicates participants whose immunogenicity data will be eliminated from a specific visit.
DOB-Date of Birth, VAC-Vaccination, BS- Blood Sample, BMI-Body Mass Index.

3.1.4. Elimination from solicited safety set

Code 1030 (Study vaccine not administered at all), code 800 (fraudulent data) and code 900 (invalid informed consent) and code 1160 (no post-vaccination solicited safety data) will be used for identifying participants eliminated from the solicited safety set.

4. STATISTICAL ANALYSES

4.1. General Considerations

Unless otherwise specified, reactogenicity analysis and safety analysis will be performed on solicited safety set and Exposed Set respectively. Immunogenicity analysis will be performed on the Per Protocol set and may be performed on the Full Analysis Set if necessary.

4.1.1. General Methodology

Unless otherwise specified, continuous data will be summarized using descriptive statistics: n, mean, standard deviation (std), median, minimum and maximum. Categorical data will be summarized as the number and percentage of participants in each category.

95% CI for proportion will be based on exact Clopper-Pearson confidence interval [Clopper, 1934].

95% CI for group difference in proportion will be based on Miettinen and Nurminen confidence interval [Miettinen, 1985].

95% CI for GMT(C) will be based on a back transformation of student confidence interval for the mean of log-transformation.

95% CI for GMT(C) ratio between groups will be based on a back transformation of confidence interval for the mean difference on log-transformation.

For a given participant and given immunogenicity measurement, missing or non-evaluable measurements will neither be imputed nor be replaced, and therefore will not be included in immunogenicity analysis.

For between group statistical modelling analysis, participants having a result at both the baseline and the considered timepoint will be included in the analysis.

Participants who prematurely withdrew from study will not be replaced.

4.1.2. Definition

Solicited events and their intensity scales are defined as below:

Baseline: For all endpoints the baseline value will be the latest pre-dose assessment with a non-missing value, including those from unscheduled visits. If time is not collected, Day 1 assessments are assumed to be taken prior to vaccination and used as baseline. Unless otherwise stated, if baseline data is missing no derivation will be performed and baseline will be set to missing.

For the RSV and FLU antigens:

- A seronegative participant will be defined as a participant whose antibody titer/concentration is below the cut-off value of the assay.
- A seropositive participant is a participant whose antibody titer/concentration is greater than or equal to the cut-off value of the assay.
- MGI (mean geometric increase): The geometric mean of the within participant ratios of the post-dose titer over the pre-dose titer.

Please refer to Section 6.2.4.6 for cut-off value for each assay.

For the FLU antigens:

- SCR (seroconversion rate): The percentage of participants who have either a HI pre-dose titer < 1:10 and a post-dose titer ≥ 1:40 or a pre-dose titer ≥1:10 and at least a four-fold increase in post-dose titer.
- SPR (seroprotection rate): The percentage of participants with a serum HI titer ≥1:40 that usually is accepted as indicating protection.

4.2. Primary Endpoint(s) Analyses

4.2.1. Safety analysis

The analysis of solicited administration site event (associated with RSV or Flu) and solicited systemic event will be performed on solicited safety set.

- The number and percentage with exact 95% CI of participants reporting each solicited administration site event (any grade, each grade) and solicited systemic event (any, each grade) during the 7-day follow-up period after dosing will be tabulated by maximum intensity per participant for each study group.
- For fever during the 7-day follow-up period after vaccination, the number and percentage of participants reporting any fever (i.e., temperature ≥38 °C) and fever by half degree (°C) cumulative increments, any Grade 3 fevers, will be reported.
- Duration in days of solicited administration site and systemic events within 7 days after vaccination will be tabulated by study group and overall. The derivation rule of duration in days for solicited events is detailed in section 6.2.4.9.

The analysis of unsolicited AEs and SAEs will be performed on ES.

- The number and percentage of participants reporting unsolicited AEs within 30 days after dosing with exact 95% CIs will be tabulated by group and by Medical Dictionary for Regulatory Activities (MedDRA) terms.
- Similar tabulations will be done for Grade 3 unsolicited AEs, for any causally related unsolicited AEs, and for Grade 3 related unsolicited AEs.
- The number and percentage of participants reporting at least one SAE within 30 days after dosing with exact 95% CIs will be tabulated by group and by MedDRA terms.
- The number and percentage of participants reporting at least one SAE up to study end with exact 95% CIs will be tabulated by group and by MedDRA terms.
- By-participant listings of SAEs, and AEs leading to study withdrawal will be prepared.

4.2.2. Immunogenicity analysis

- Lot-to-lot consistency analysis

The GMC ratio and its two-sided 95% CI between 2 lots in terms of RSVPreF3 IgG ELISA concentration at Day 31 will be computed by back transformation of its mean difference and 95% CI on log10 transformed scale estimated from the t-distribution.

Summary table will show the number of observations included in the calculation for each lot, GMC of each lot, and 3 pair-wise GMC ratios with corresponding 95% CIs.

Lot-to-lot consistency will be demonstrated if all 3 two-sided 95% CIs for GMC ratio fall within 0.67 and 1.5.

- Non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone with respect to 3 Flu D-QIV strains at Day 31

  The GMT ratio and its two-sided 95% CIs between the 2 groups (RSV+Flu with 3 lots pooled divided by Flu+P group) for A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019, and B/Phuket/3073/2013 will be computed by back transformation of its mean difference and 95% CI on log10 transformed scale estimated from the t-distribution.

  Summary table will show for each group the number of observations included in the calculation and GMT value, and between group GMT ratio with corresponding 95% CI.

  Non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine will be demonstrated if lower bounds of all 3 two-sided 95% CIs are above 0.67.

4.2.3. Sensitivity analyses

For primary immunogenicity endpoints, between study group comparisons may also be explored through statistical modelling.

- Lot-to-lot consistency analysis

  The ANCOVA model on the log10 transformation of the concentrations may be explored, and include the lot as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination concentrations as covariates. Covariates in the model may be further adjusted when performing the analysis if appropriate and needed, therefore, the following SAS code can serve as a reference and may be adjusted according to the analysis needs.

```
PROC GLM data=sero;
  CLASS group age_cat;
     MODEL log_val = baseline group age_cat group*age_cat;
      LSMEANS group/pdiff cl alpha=0.05;
     RUN;
```

  where log_val represents the log-transformed RSVPreF3 IgG antibody concentration at Day 31, baseline is pre-vaccination RSVPreF3 IgG antibody level on log-transformed scale, group includes each lot, age_cat is the age category at vaccination (18-32 vs. 33-49).

The 3 pair-wise GMC ratios between 2 lots and their two-sided 95% CI will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

Summary table will show the number of observations included in the model for each lot, adjusted GMC of each lot, and 3 pair-wise adjusted GMC ratios with corresponding 95% CIs.

- Non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone with respect to 3 Flu D-QIV strains at Day 31

  The ANCOVA model on the log10 transformation of the titers may be explored, and include the study group (RSV+Flu with 3 lots pooled, Flu+P) as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination titers as covariates, covariates in the model may be further adjusted when performing the analysis if appropriate and needed. Similar SAS code could be referenced and may be further adjusted at time of analysis:

```
PROC GLM data=sero;
  CLASS group age_cat;
     MODEL log_val = baseline group age_cat group*age_cat;
      LSMEANS group/pdiff cl alpha=0.05;
     RUN;
```

  where log_val represents the log-transformed antibody level at Day 31, baseline is pre-vaccination antibody level on log-transformed scale, group includes RSV+Flu with 3 lots pooled and Flu+P, age_cat is the age category at vaccination (18-32 vs. 33-49).

  The GMT ratio and its two-sided 95% CIs between the 2 groups (RSV+Flu with 3 lots pooled divided by Flu+P group) for A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019, and B/Phuket/3073/2013 will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

  Summary tables will show for each group the number of observations included in the model and model adjusted GMT, and between group adjusted GMT ratios with the corresponding 95% CIs.

4.3. Secondary Endpoint(s) Analyses

4.3.1. Immunogenicity analysis

4.3.1.1. Secondary confirmatory analysis

In order to control the global type I error, the objectives will be assessed sequentially.

- Non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of RSV A neutralizing antibody titers at Day 31

  The GMT ratio and its two-sided 95% CI between 2 groups (RSV+Flu group with 3 lots pooled divided by RSV alone group with 3 lots pooled) will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from t-distribution.

  Summary table will show for each group the number of observations included in the calculation and GMT value, and between group GMT ratio with corresponding 95% CI.

  No interference on RSVPreF3 vaccine from quadrivalent seasonal influenza vaccine (Flu D-QIV) will be demonstrated if lower bound of the two-sided 95% CI of GMT ratio is above 0.67.

- Non-inferiority for each of the 3 strains of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone at Day 31 in terms of Seroconversion rate (SCR)

  The SCR difference and its two-sided 95% CI between 2 groups (Flu+P group minus RSV+Flu group with 3 lots pooled) will be calculated for each strain (A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019, and B/Phuket/3073/2013) at Day 31. 95% CI calculation will be based on Miettinen and Nurminen confidence interval [Miettinen, 1985].

  Summary table will show for each group the number of observations included in the calculation, number and percentage of seroconverted participants, and between group SCR difference with corresponding 95% CI.

  For each strain, non-inferiority will be demonstrated if the upper bound of the two-sided 95% CI is less than 10% at Day 31 post vaccination.

4.3.1.2. Descriptive immunogenicity analysis

For RSV A, RSV B neutralizing antibody titers and RSVPreF3 IgG antibody concentrations at pre-vaccination and Day 31 from each lot (RSV1, RSV2, and RSV3), as well as for the group of RSV alone with 3 lots pooled (RSV1, RSV2 and RSV3 pooled) and RSV+Flu group with 3 lots pooled, the following analysis will be performed:

- Number and percentage of participants with antibody titers/concentrations above assay cut-off will be tabulated with its exact 95% CI by group.
- GMT/GMCs will be tabulated with 95% CI at each timepoint and represented graphically by group.
- MGI from Day 31 over pre-vaccination will be tabulated with 95% CI by group.

For Flu D-QIV HI antibody titers against 3 influenza strains at pre-vaccination and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine with 3 lots pooled (RSV+Flu) and the group of Flu D-QIV vaccine administered alone (Flu+P), the following analysis will be performed:

- Number and percentage of participants with antibody titers above assay cut-off and their exact 95% CI will be tabulated by group.
- GMTs will be tabulated with 95% CI at each timepoint and represented graphically by group.
- MGI from Day 31 over pre-vaccination will be tabulated with 95% CI by group.
- Number and percentage of participants achieving seroconversion for HI antibody at Day 31 will be tabulated with its exact 95% CI by group.
- Number and percentage of participants achieving HI antibody titer ≥ 1:40 at pre-vaccination and Day 31 will be tabulated with its exact 95% CI by group.

4.3.2. Sensitivity analyses

ANCOVA modelling analysis may be explored for non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of RSV A neutralizing antibody titers at Day 31.

The ANCOVA model will be based on the log10 transformation of the titers, and include the study group (RSV+Flu with 3 lots pooled, RSV alone group with 3 lots pooled) as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination titers as covariates, covariates in the model may be further adjusted when performing the analysis if appropriate and needed. Similar SAS code will be explored and may be further adjusted at time of analysis:

```
PROC GLM data=sero;
  CLASS group age_cat;
  MODEL log_val = baseline group age_cat group*age_cat;
  LSMEANS group/pdiff cl alpha=0.05;
RUN;
```

where log_val represents the log-transformed RSV A neutralizing antibody titer at Day 31, baseline is pre-vaccination RSV A neutralizing antibody titer on log-transformed scale, group includes Flu+RSV with 3 lots pooled and RSV alone with 3 lots pooled, age_cat is the age category at vaccination (18-32 vs. 33-49).

The GMT ratio and its two-sided 95% CIs between the 2 groups (RSVPreF3+Flu D-QIV with 3 lots pooled divided by RSVPreF3 alone group with 3 lots pooled) will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

Summary tables will show for each group the number of observations included in the model and model adjusted GMT, and between group adjusted GMT ratios with the corresponding 95% CIs.

4.4. Tertiary Endpoint(s) Analyses

The same statistical methods that are used to analyse the 3 strains in the primary and secondary endpoints will be used to analyse the A/Victoria/2570/2019 (H1N1) IVR-215 strain.

4.5. Other Safety Analyses

Other safety analyses will be based on the Exposed Set, unless otherwise specified.

4.5.1. Combined solicited and unsolicited events

For clintrial.gov and EudraCT posting purposes, a summary of combined solicited and unsolicited adverse events will be produced by System Organ Class and preferred terms and according to occurrence of each event.

Solicited administration site events and solicited systemic events will be coded by MedDRA as per the following codes

Solicited symptom	Lower level term code	Corresponding Lower level term decode
Pain	Injection site pain	10022086
Redness	Injection site redness	10022098
Swelling	Injection site swelling	10053425
Fatigue	Fatigue	10016256
Fever	Fever	10016558
Nausea	Nausea	10028813
Vomiting	Vomiting	10047700
Diarrhea	Diarrhea	10012727
Abdominal pain	Abdominal pain	10000081
Headache	Headache	10019211

Please note: the coding will be double checked during the analysis

4.5.2. COVID-19 Assessment and COVID-19 AEs

Numbers and percentage of participants with a COVID-19 infection throughout the study period will be summarized by group based on ES.

COVID-19 infections identified during the 30 days follow-up period (Day 1 to Day 30) will be further assessed as the following:

A participant is defined as having a suspected, probable or confirmed COVID-19 infection during the study if the answer is “Confirmed”, “Probable” or “Suspected” to the case diagnosis question from the COVID-19 coronavirus infection assessment eCRF.

Numbers and percentage of participants with a suspected, probable or confirmed COVID-19 infection will be summarized by group based on ES.

Number and percentage of participants who had a COVID-19 test performed and number and percentage of participants with positive, negative and indeterminate results will be summarized by group on ES.

4.5.3. Additional Safety Assessments (if applicable)

Vital signs will be summarized by group using descriptive statistics at all timepoint(s) the information is collected on ES and PPS. The parameters include but may not be limited to systolic blood pressure, diastolic blood pressure, temperature, heart rate, respiratory rate, height, weight and body mass index.

4.6. Other Analyses

Not applicable.

4.7. Changes to Protocol Defined Analyses

There were no changes or deviations to the planned statistical analysis specified in the protocol amendment 3 (Dated: [26-MAY-2022]).

5. SAMPLE SIZE DETERMINATION

Approximately 1541 participants will be randomized to achieve appropriately 1400 evaluable participants.

Assessments of both immunogenicity and safety data were considered when determining sample size for this study.

Participants who withdraw from the study will not be replaced.

The sections below describe the assumptions, adjustment and methodology used for the sample size calculation.

5.1. Primary objective for Lot-to-Lot consistency

The primary objective of lot-to-lot consistency will be evaluated on the RSVPreF3 immunogenicity as measured by RSVPreF3 IgG ELISA concentration at Day 31.

The lot-to-lot consistency will be demonstrated only if two-sided 95% confidence intervals for the 3 pair-wise geometric mean ratios of RSVPreF3 IgG ELISA concentration at Day 31 (30 days post vaccination) falls within 0.67 and 1.5.

With the assumptions of a SD of log10 transformed RSVPreF3 IgG ELISA concentration of 0.35, true GMC ratio of 1 between 2 lots, type I error of 0.025, and 10% of non-evaluable rate, 220 enrolled participants per lot provides at least 99% global power to conclude lot-to-lot consistency (see details in Table 7).

Table 7 Power to demonstrate Lot-to-Lot consistency on immune responses of RSVPreF3 IgG ELISA concentration at Day 31 post vaccination

Endpoint	Clinically acceptable bounds for consistency	Group description	Number of evaluable participants in each lot	Reference SD**	Power
*RSVPreF3 IgG GMC ratio	(0.67, 1.5)	Lot 1/2	200	0.35	99.8%
*RSVPreF3 IgG GMC ratio	(0.67, 1.5]	Lot 1/3	200	0.35	99.8%
*RSVPreF3 IgG GMC ratio	(0.67, 1.5]	Lot 2/3	200	0.35	99.8%
Global power					99.3%

* Pass 2019, Two-Sample T-test for equivalence allowing unequal variance, alpha=2.5%;
**References used for the sample size calculation: Study *RSV MAT 011* (209141)

5.2. Primary objective: To demonstrate Non-inferiority of immune response of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine or administered alone at Day 31 post dose vaccination

Possible interference of RSVPreF3 on influenza immune responses will be evaluated by using Flu D-QIV HI antibody titers against the 3 influenza strains from RSV+Flu (pooled of RSV1+Flu, RSV2+Flu, RSV3+Flu) and Flu+P group at Day 31. In the description of the subsequent analysis, the RSV+Flu vaccine group are RSV1+Flu, RSV2+Flu, RSV3+Flu groups pooled.

The hypothesis is that co-administration of RSVPreF3+ Flu D-QIV vaccine is non-inferior to Flu D-QIV vaccine with respect to GMT ratio for immune response of influenza antigen in non-pregnant women at Day 31 post vaccination.

The criteria to evaluate non-inferiority for A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019, and B/Phuket/3073/2013 is that the lower limits of the 95% CI on the GMT ratio (RSV+Flu divided by Flu+P) is greater than 0.67 at Day 31 post vaccination.

Table 8 presents the power on the ratio of the GMT of Flu D-QIV 3 strains. The power is at least 95% when the sample size is 400 participants per group with standard deviation (SD) of 0.6 on its log10 transformation and non-inferiority margin of 0.67 between RSVPreF3 co-administrated with Flu D-QIV group and Flu D-QIV vaccine administered alone group.

Table 8 Power to demonstrate non-inferiority of immune response between Flu D-QIV vaccine co-administered with RSVPreF3 or administered alone in term of GMT for Flu D-QIV strains at Day 31 post 1st vaccination with assumed GMT ratio 1

Endpoint	NI criteria	Number of evaluable participants in each group	Reference*	Power
B/Washington/02/2019 GMT ratio	LL of 95% CI for GMC ratio >0.67	400	0.60	98.56%
A/Tasmania/503/2020 (H3N2)** IVR-221 GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
B/Phuket/3073/2013 GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
Global power				95.68%

By Pass 2019, Non-Inferiority Tests for Two Means using Differences, one-sided alpha=2.5%;
*References used for the sample size calculation: Study Flu D-QIV -008 E1_01 Table 34
**A/Tasmania/503/2020 (H3N2) IVR-221 (an A/Cambodia/e0826360/2020 (H3N2)-like virus).

5.3. Secondary objectives

In order to control the global type I error, the objectives will be assessed sequentially.

5.3.1. To demonstrate the non-inferiority of immune response between RSVPreF3 co-administered with Flu D-QIV and RSVPreF3 alone in term of RSV A Neutralizing antibody titers at Day 31 post administration

Hypothesis under consideration is that immune response when the co-administration of RSVPreF3 vaccine and Flu D-QIV vaccine is non-inferior to that when RSVPreF3 vaccine (pooled RSV1, RSV2 and RSV3 group) is administered alone with respect to GMT ratio for RSV A neutralizing antibody titer in non-pregnant women at Day 31 post vaccination.

This hypothesis test is conducted only if the primary objective of non-inferiority of immune response between Flu D-QIV vaccine when co-administered with RSVPreF3 and Flu D-QIV vaccine administered alone is met.

The criteria to evaluate non-inferiority for RSV A Nab is that the lower limits of the 95% CI on the GMT ratio (RSV+Flu group divided by RSV alone group with 3 lots pooled in both groups) is greater than the pre-defined clinical limit of 0.67 at Day 31 post vaccination.

If a SD of log10 transformed RSV A Nab of 0.4 is assumed, with an assumption of GMT ratio of 1.0, there will be at least 99% chance that the lower bound of 95% CI for the ratio of GMT of RSV A Nab titer between RSVPreF3 co-administration with Flu D-QIV and administered alone is above 0.67 (Table 9)

Table 9 Power to demonstrate non-inferiority of immune response of RSVPreF3 vaccine when co-administered with Flu D-QIV vaccine or administered alone at Day 31 post vaccination

Endpoint	NI criteria	N1:N2 (evaluable)	Reference*	Power
RSV A GMT ratio	LL of 95% CI for GMT ratio >0.67	400:600	0.40	99.99%

By Pass 2019 Non-Inferiority Tests for Two Means using Differences, one-sided alpha=2.5%;
*References used for the sample size calculation: Study RSV MAT 011 (209141)
N1: Number of evaluable participants in the co-administration of RSVPreF3 vaccine + Flu D-QIV vaccine.
N2: Number of evaluable participants in the RSVPreF3 vaccine alone group with 3 lots pooled.

5.3.2. To demonstrate the non-inferiority of immune response between RSVPreF3 co-administered with Flu D-QIV and Flu D-QIV alone in term of SCR at Day 31 post 1st dose administration

Potential interference of RSVPreF3 on influenza immune responses will be evaluated by comparing the Day 31 seroconversion rate of Flu D-QIV HI antibody titers against the 3 influenza strains between RSV+Flu group and Flu+P group.

The criteria to evaluate non-inferiority with respect to the SCR difference for Flu D-QIV antibody titers against 3 influenza strains is that the upper limits of the 95% CI on the SCR difference (Flu+P group minus RSV+Flu) is less than the pre-defined clinical limit of 10% at Day 31 post vaccination.

With the assumptions of SCR for the reference group as indicated in Table 10, significance level 0.025, non-inferiority margin 10%, 400 evaluable participants (10% non-evaluable rate) provides at least 80% power to conclude non-inferiority for each strain (Table 10).

Table 10 Powers to demonstrate non-inferiority in terms of HI antibody SCR for Flu D-QIV strains between Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and administered alone at Day 31 post vaccination

Endpoint	NI criteria	Anticipated SCR*	Number of participants per group (evaluable)	Power
B/Washington/02/2019 SCR	10%	65%	400	84.39%
A/Tasmania/503/2020 (H3N2)** IVR-221 SCR	10%	65%	400	84.39%
B/Phuket/3073/2013 SCR	10%	65%	400	84.39%

By Pass 2019, Non-Inferiority Tests for the difference between two proportions, alpha=2.5%
*SCR used in the reference group for power calculation is from Study Tdap-0.3-008, Suppl. Table 36.
**A/Tasmania/503/2020 (H3N2) IVR-221 (an A/Cambodia/e0826360/2020 (H3N2)-like virus).

6. SUPPORTING DOCUMENTATION

6.1. Appendix 1 Study Population Analyses

6.1.1. Participant Disposition

Summary of participant disposition will be performed on ES. The number and percentage of participants who completed the study and who prematurely withdrew from the study including the reasons for study withdrawal will be summarized by each group and overall.

6.1.2. Demographic and Baseline Characteristics

The analysis of demography will be performed on Enrolled Set, ES and PPS using descriptive statistics. The parameters including but may not be limited to age, age category (18-32 vs 33-49), ethnicity, race, country and childbearing potential will be summarized by group.

Summary of past medical history and current medical conditions will be performed on ES by Medical Dictionary for Regulatory Activities (MedDRA) term. Un-coded medical conditions or medical history will be summarized under 'Other' category.

Vaccination history will be coded using GSK Drug dictionaries. Summary of vaccination history will be performed on ES by group.

6.1.3. Protocol Deviations

Important protocol deviations will be summarized based on ES by group.

Protocol deviations will be tracked by the study team throughout the conduct of the study. These protocol deviations will be reviewed to identify those considered as important as follows:

- Data will be reviewed prior to unblinding and freezing the database to ensure all important deviations (where possible without knowing the study intervention details) are captured and categorised in the protocol deviations dataset.
- This dataset will be the basis for the summaries of important protocol deviations.
- The summary will include number and percentage of participants with important protocol deviations by deviation category for each study group.
- An individual listing of protocol deviation will also be provided.

Protocol deviations which result in exclusion from the analysis set will be summarized on ES by group.

- Data will be reviewed prior to unblinding and freezing the database to ensure all deviations leading to analysis population exclusions are captured and categorised in the protocol deviations ADaM dataset (note these exclusions are not captured in the SDTM dataset).
- The summary will include number and percentage of participants with protocol deviations leading to exclusion by deviation category for each study group.

In addition to the above summary, separate summaries may be produced for important protocol deviations related to COVID-19, and important protocol deviations not related to COVID-19 respectively.

6.1.4. Concomitant Medications and Vaccinations

Concomitant medications and vaccinations will be coded and summarized using GSK Drug dictionary.

- The number and percentage of participants taking concomitant medications (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) will be summarized by group. A listing will also be provided.
- The number and percentage of participants taking concomitant vaccinations will be summarized by group. A listing will also be provided.

6.1.5. Additional Analyses Due to the COVID-19 Pandemic

Depending on how the Covid-19 situation evolves, the SAP might be amended to reflect the analysis corresponding to Covid-19.

6.2. Appendix 2 Data Derivations Rule

6.2.1. Study Day and Reference Dates

The safety reference date is the study intervention start date and will be used to calculate study day for safety measures.

The study day is calculated as below:

- Assessment Date = Missing → Study Day = Missing
- Assessment Date < Reference Date → Study Day = Assessment Date – Ref Date
- Assessment Data ≥ Reference Date → Study Day = Assessment Date – Ref Date + 1

6.2.2. Attributing events to vaccine doses

The dose relative to an event is the most recent study dose given to a participant prior to the start of a given event. For example, if the start date of an adverse event is between Dose 1 and Dose 2, the relative dose will be Dose 1.

If an event starts on the same day as a study dose, the relative dose will be derived from the additional information provided in the case report form (CRF) using the contents of the flag indicating if the event occurred before or after study dose. If 'after study dose' is selected, the relative dose for the event will be the one administered on the start day of the event. If 'before study dose' is selected, the relative dose for the event will be the dose prior to this one.

6.2.3. Handling of missing data

6.2.3.1. Dates

When partially completed dates (i.e. dates missing a day and/or month) are used in calculations, the following standard rules will be applied:

- A missing day will be replaced by 15
- A missing day and month will be replaced by June 30th.

The following exceptions apply:

- Adverse event start dates with missing day:
  - If the event starts in the same month as at least one of the study doses, the contents of AE.AESTRTPT (the flag indicating if the event occurred before or after study dose) will be used to complete the date. If 'after study dose' is selected, the imputed start date will match the first (or only) study dose given during that month. If 'before study dose' is selected, the imputed date will be one day before the first (or only) study dose given during that month.
- Adverse event start dates with missing day and month:
  - If the event starts in the same year as at least one of the study doses, the contents of AE.AESTRTPT (the flag indicating if the event occurred before or after study dose) will be used to complete the date. If 'after study dose' is selected, the imputed start date will match the first (or only) study dose given during that year. If 'before study dose' is selected, the imputed date will be one day before the first (or only) study dose given during that year.

All other cases of incomplete AE or concomitant medication/vaccination start date will follow the standard rules above.

6.2.3.2. Daily recording of solicited events

6.2.3.2.1. Studies with electronic diaries

For studies using electronic diaries for the collection of solicited events, a solicited event will be considered present only when a daily recording of grade 1 or more is present.

6.2.3.3. Unsolicited adverse events

Unsolicited adverse event summaries are including serious adverse events unless specified otherwise.

Missing severity, relationship with study vaccine, and outcome of unsolicited adverse events will not be replaced and will appear as 'UNKNOWN' when displayed in a statistical output.

6.2.4. Data derivation

6.2.4.1. Age at first dose in years

When age at first dose is to be displayed in years, it will be calculated as the number of complete calendar years between the date of birth and the date of first dose. For example:

DOB = 10SEP1983, Date of first dose = 09SEP2018 -> Age = 34 years

DOB = 10SEP1983, Date of first dose = 10SEP2018 -> Age = 35 years

6.2.4.2. Weight

Weight will be presented in kilograms. Weights reported in pounds will be converted as follows:

Weight in kilograms = Weight in pounds / 2.2

6.2.4.3. Height

Height will be presented in centimeters. Heights reported in feet and inches will be converted as follows:

Height in centimeters = Height in inches x 2.54

6.2.4.4. Body mass index (BMI)

BMI will be calculated as follows:

BMI = (Weight in kilograms) / (Height in meters)2

6.2.4.5. Temperature

Temperatures will be presented in degrees Celsius (°C). Temperatures reported in degrees Fahrenheit (°F) will be converted as follows:

Temperature (Celsius) = ((Temperature (Fahrenheit) - 32) x 5)/9

6.2.4.6. Numerical serology results

Numerical serology results will be derived from the content of IS.ISORRES in the SDTM dataset. For assays with a specific cut-off, the following derivation rules apply:

IS.ISORRES	Derived value
"NEG", "-", or "(-)"	cut-off/2
"POS", "+", or "(+)"	cut-off
"< value" and value is <= assay cut-off	cut-off/2
"< value" and value is > assay cut-off	value
"> value" and value is < assay cut-off	cut-off/2
"> value" and value is >= assay cut-off	value
"value" and value is < cut-off	cut-off/2
"value" and value is >= cut-off	value
All other cases	missing

The cut-off tests for immunogenicity evaluation will be as per following:

System	Component	Method	Unit	Cut-off *(LLOQ)*	*ULOQ*
Serum	RSV-A Neutralising Antibody	NEUTRALISATION	ED60	18	*123535*
Serum	RSV-A Neutralising Antibody	NEUTRALISATION	IU/mL	56	*217400*
Serum	RSVPreF3 IgG antibody concentrations	ELISA	EU/mL	*25*	*251769*
Serum	RSV-B Neutralising Antibody	NEUTRALISATION	ED60	*30*	*138336*
Serum	RSV-B Neutralising Antibody	NEUTRALISATION	IU/mL	*44*	*171279*
Serum	A/Victoria/2570/2019 (H1N1)	HI assay	*TBD*	*TBD*	*TBD*
Serum	A/Tasmania/503/2020 (H3N2)	HI assay	*TBD*	*TBD*	*TBD*
Serum	B/Washington/02/2019	HI assay	*TBD*	*TBD*	*TBD*
Serum	B/Phuket/3073/2013	HI assay	*TBD*	*TBD*	*TBD*

Note: the assay cut-off (LLOQ), ULOQ and units may be further adjusted at time of analysis when notified by the lab.

6.2.4.7. Geometric mean titres (GMTs) and concentrations (GMCs)

Geometric Mean Titre (GMT) or Concentration (GMC) calculations are performed by taking the inverse logarithm of the mean of the log titre or concentration transformations. Non quantifiable antibody titres or concentrations will be converted as described in section 6.2.4.6 for the purpose of GMT/GMC calculation. The cut-off value is defined by the laboratory before the analysis.

6.2.4.8. Onset day

The onset day for an event (e.g. AE, concomitant medication/vaccination) is the number of days between the last study dose and the start date of the event. This is 1 for an event occurring on the same day as a study dose (and reported as starting after study dose).

6.2.4.9. Duration of events

The duration of an event with a start and end date will be the difference between the start and end date plus one day, i.e. an event that starts on 03MAR2018 and ends on 12MAR2018 has a duration of 10 days.

The duration of solicited events will be calculated as the sum of the individual days with the adverse event reported at grade 1 or higher during the solicited event period.

6.2.4.10. Counting rules for occurrences of solicited events

When the occurrences of solicited events are summarized, each event recorded as having occurred during a specific period will be counted as only one occurrence regardless of the number of days on which it occurs. Also, in the case of co-administered study interventions, an administration site event recorded for a participant following multiple study interventions will be counted as only one occurrence.

6.2.5. Display of decimals

6.2.5.1. Percentages

Percentages and their corresponding confidence limits will be displayed with one decimal except for 100% in which case no decimal will be displayed.

6.2.5.2. Differences in percentages

Differences in percentages and their corresponding confidence limits will be displayed with two decimals.

6.2.5.3. Demographic/baseline characteristics statistics

The mean, median, and standard deviation for continuous baseline characteristics (height, weight, body mass index (BMI), pre-dose body temperature) will be presented with one decimal.

The minimum and maximum values and quartile values (if required) will be presented with the same number of decimals as the observed values.

The maxima and minima of transformed height/weight variables will be displayed without decimals.

The maximum and minimum of transformed body temperatures will be displayed with one decimal.

6.2.5.4. Serological summary statistics

The number of decimals used when displaying geometric mean titers (GMT) or concentrations (GMC) and their confidence limits is assay specific based on the magnitude of the assay result post-dose and the clinically relevant assay threshold. The same number of decimals will be used for a given assay regardless of the timepoint presented.

Lowest clinically relevant threshold	Example	Number of decimals to display
<0.3	Diphtheria, tetanus, anti-PRP	3
>=0.3 and <4	*Streptococcus pneumoniae*, Meningococal bactericide	2
>=4 and <1000	Measles, rubella, varicella, polio	1
>=1000	CMI	0

GMT/GMC fold increase from pre-dose follows the same principle. Namely when the lowest clinically relevant threshold is 2 fold, 2 decimals are displayed while when the lowest clinically relevant threshold is 4 fold, 1 decimal is displayed.

GMT or GMC group ratios and their confidence limits will be displayed with 2 decimals regardless of the actual values.

7. REFERENCES

Clopper CJ, Pearson E. The Use of Confidence or Fiducial Limits Illustrated in the case of the Binomial. *Biometrika.* 1934;26:404-13.

Miettinen, O. S. and Nurminen, M. Comparative analysis of two rates. Statistics in Medicine, 1985;4,213-226.

Information Type:	Statistical Analysis Plan (SAP)

TITLE PAGE

Protocol Title: A Phase III, randomized, multi-country study to evaluate the lot-to-lot consistency of GSK's investigational RSV maternal vaccine and the immune response, safety and reactogenicity of RSV maternal vaccine when co-administered with GSK's quadrivalent influenza D-QIV vaccine in healthy non-pregnant women 18-49 years of age.

Study Number: 214709 (RSV MAT-010)

Compound Number: GSK3888550A

Abbreviated Title: A Phase III study to assess the lot-to-lot consistency of GSK's investigational RSV maternal vaccine and the immune response and safety of RSV maternal vaccine when given alone or co-administered with GSK's influenza D-QIV vaccine in healthy non-pregnant women.

Sponsor Name: GlaxoSmithKline Biologicals SA (GSK)

Regulatory Agency Identifier Number(s): IND 018434

Registry	**ID**
Clintrials.gov	NCT05045144

TABLE OF CONTENTS

PAGE

LIST OF TABLES

PAGE

Version history

SAP Version	Approval Date	Protocol Version (Date) on which SAP is Based	Change	Rationale
SAP	29 Jun 2021	Amendment 1 (27 May 2021)	Not Applicable	Original version
Amendment 1	29 Mar 2022	Amendment 2 (17 November 2021)	1. Blinding strategy detail from the study title has been deleted, as they vary between the lot to lot and co-administration groups. 2. Interim analysis has been removed. 3. Components updated with 2021/22 season Flu strains.	1. It was identified that the intervention type amongst the co-administration groups can be determined by the blinded site and sponsor staff using available information in the electronic case report forms (eCRF), therefore the blinding strategy was adapted to single-blind design for the co-administration groups. 2. The decision was made to not go forward with the BLA filing for the RSV Maternal vaccine, and as a result, the interim analysis was removed. 3. New information made available on the Flu strains.

1. INTRODUCTION

The purpose of this SAP is to describe the planned analyses for RSV MAT-010 (214709). Details of the planned analysis are provided.

1.1. Objectives, Estimands and Endpoints

Table 1 Study objectives, endpoints and estimands

Objectives	Endpoints/Estimands
Primary	
Safety • To evaluate the safety and reactogenicity of RSVPreF3 when given alone (pooled lots) or co-administered with Flu D-QIV up to Day 31 post administration.	The number and percentage of participants in each group reporting • Each solicited administration site AE collected during the 7 days follow-up period (Day 1 to Day 7) • Each solicited systemic events in the 7 days follow-up period • Unsolicited AEs collected during the 30 days follow-up period (Day 1 to Day 30) • SAEs in the 30 days follow-up period (Day 1 to Day 30)
Immunogenicity • To demonstrate the lot-to-lot consistency of 3 lots of the investigational RSVPreF3 vaccine based on GMC of RSVPreF3 IgG ELISA at Day 31	RSVPreF3 IgG ELISA concentration at Day 31 (30 days post administration) Measured by ratio of Geometric mean concentration (GMC) between lots in terms of RSVPreF3 IgG ELISA titers at Day 31 (30 days post administration)
• To demonstrate non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine compared to Flu D-QIV given alone based on Geometric mean titer (GMT) of Flu D-QIV antibody titers against 4 influenza strains at Day 31 post administration	Flu D-QIV antibody titers against 4 influenza strains at Day 31 (30 days post administration) Measured by ratio of Haemagglutinin inhibition (HI) GMT between group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and Flu D-QIV vaccine alone group against 4 strains at Day 31 (30 days post administration)
Secondary	
Secondary Safety	
• To evaluate the safety of RSVPreF3 when given alone and co-administered with Flu D-QIV from vaccination up to study end.	The number and percentage of participants reporting SAEs from first vaccination up to study end. [Day 1 to Day 181]
*Confirmatory Immunogenicity***	
• To demonstrate the non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV compared to given alone based on GMT of RSV A neutralizing antibody at Day 31 post administration	RSV A neutralizing antibody titers at Day 31 (30 days post administration) Measured by ratio of GMT between group of RSVPreF3 vaccine co-administered with Flu D-QIV and RSVPreF3 vaccine alone (pooled lot 1,2,3) group in term of RSV A neutralizing antibody titers at Day 31 (30 days administration)
• To demonstrate the non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 compared to given alone based on Seroconversion rate	• Seroconversion rate to Flu D-QIV HI antibody titers against the 4 influenza strains at Day 31

Objectives	Endpoints/Estimands
(SCR**) of Flu D-QIV HI antibody titers against 4 influenza strains at Day 31 post study intervention	Measured by the difference of proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
Descriptive Immunogenicity	
• To evaluate the humoral immune response of RSVPreF3 vaccine when given alone and co-administered with Flu D-QIV in terms of RSV A, RSV B neutralizing antibody and RSVPreF3 IgG concentration at Day 1 (prior to the vaccination) and Day 31 (post study intervention).	• RSV A, RSV B neutralizing antibody titers and RSV PreF3 IgG concentration at Day 1 and Day 31 GMT of RSV A and RSV B neutralizing antibody, GMC of RSVPreF3 IgG at Day 1 and Day 31, in the group of RSVPreF3 vaccine co-administered with Flu D-QIV and the group of RSVPreF3 vaccine alone
• To evaluate the humoral immune response to the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine in terms of antibody titers against 4 influenza strains at Day 1 and Day 31	Flu D-QIVHI antibody titers against the 4 influenza strains at Day 1 and Day 31 HI GMT in the 4 influenza strains at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
• To evaluate seroprotection rate (SPR)* and Seroconversion rate (SCR)** of the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine	Seroprotection rate to Flu D-QIV HI antibody titers against the 4 influenza strains at Day 1 and Day 31 Measured by the proportion of participants achieving an HI antibody titer ≥1:40 at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone • Seroconversion rate to Flu D-QIV HI antibody titers against the 4 influenza strains at Day 31 Measured by the proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
• To evaluate the humoral immune response in 3 individual lots of RSVPreF3	• RSV A, RSV B neutralizing antibody titers and RSVPreF3 IgG antibody concentrations at pre-vaccination and Day 31 Geometric mean titer/concentration (GMT/GMC) in term of RSV A, RSV B neutralizing antibody and RSVPreF3 IgG antibody at Day 1 (pre-vaccination) and Day 31 in each of 3 lots of the investigational PreF3 vaccine.
Tertiary Objective	
CCI [redacted]	

*SPR is defined as the percentage of vaccines with a serum HI titer >1:40 that usually is accepted as indicating protection.

** The SCR is defined as the proportion of participants with:

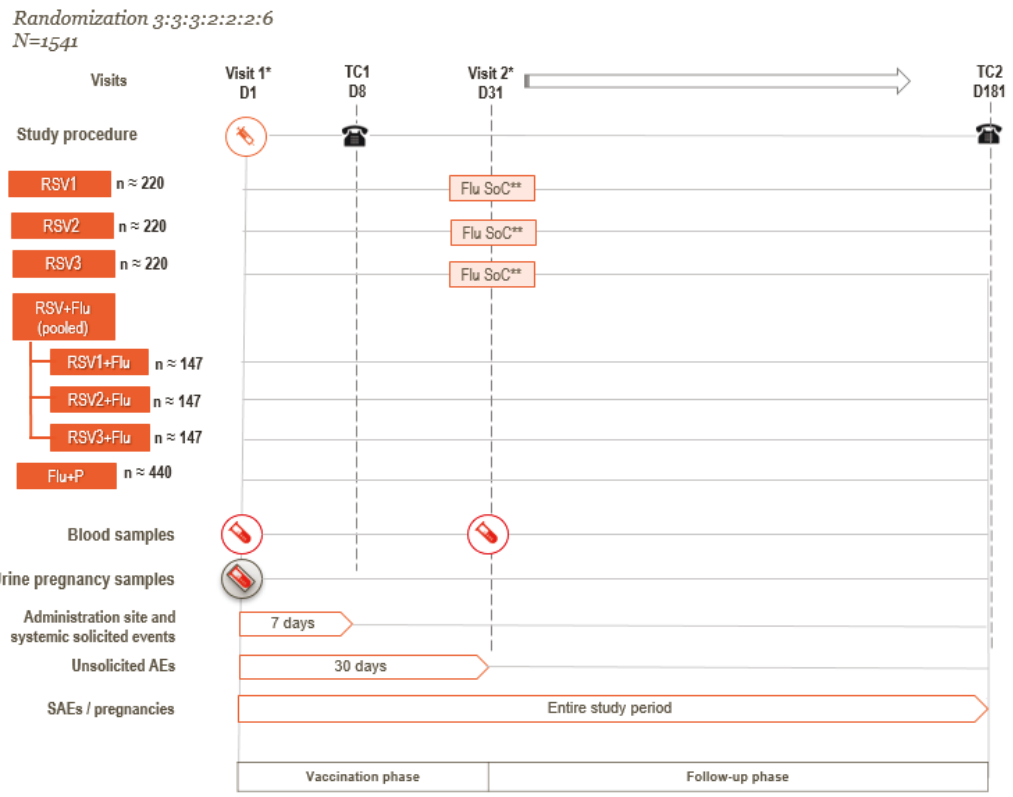
A Day 1 (pre-vaccination) serum anti-HI titer <1:10 and a Day 31 (post-vaccination) serum anti-HI titer ≥1:40, or

A Day 1 (pre-vaccination) serum anti-HI titer ≥ 1:10 and a fold increase (post/pre) ≥ 4 at Day 31

*** Two confirmatory objectives will be tested sequentially.

1.2. Study Design

Overview of Study Design and Key Features



SOC=Standard of Care
* Screening and vaccination will happen on Visit 1(Day 1). The blood sample collection must be collected before administering the vaccine(s).
** Visit 2 Flu D-QIV vaccinations are not part of experimental design. It serves as an optional vaccination for the participant to decide if they would like to get the vaccination as part of their standard of care. The blood sample collection must be collected before administering the Flu D-QIV vaccine.

Design Features	• Study Phase and Population: Phase III, Non-pregnant women • Design type: Randomized, Parallel, multi-country study • Control type: Placebo controlled, Saline (NaCl) solution • Study duration: The study vaccine will be administered at Visit 1 and the participant will be followed for 6 months until the study conclusion. • Blinding: This study will utilize an observer-blind manner to evaluate the lot-to-lot consistency of GSK RSVPreF3 vaccine and single-blind manner to evaluate the immune response, safety and reactogenicity of RSV maternal vaccine when co-administered with GSK quadrivalent influenza. Detailed blinding design is described in Protocol Section 6.3.5

Overview of Study Design and Key Features	
Study intervention	Study (intervention) groups are described in Table 2
Study intervention Assignment	Approximately 1541 participants will be randomized into 7 study groups (5 main study groups including 1 pooled group [RSV1+Flu, RSV2+Flu, RSV3+Flu]) with randomization ratio of RSV1:RSV2:RSV3: RSV1+Flu: RSV2+Flu: RSV3+Flu: Flu+P as 3:3:3:2:2:2:6. An automated internet based system (SBIR) will be used to randomly allocate a study group and treatment number to each participant. Age (18-32 vs. 33-49) and center will be minimization factors. Minimization factors will have equal weight in the algorithm.

Table 2 Study groups and intervention

Study groups	Approximate number of participants	Age (Min-Max)	Study intervention	Standard of Care Vaccination*
RSV1	220	18 – 49 years	RSV MAT 120 (Lot1)	Flu D-QIV
RSV2	220	18 – 49 years	RSV MAT 120 (Lot2)	Flu D-QIV
RSV3	220	18 – 49 years	RSV MAT 120 (Lot3)	Flu D-QIV
RSV1+Flu	147	18 – 49 years	RSV MAT 120 (Lot 1) Flu D-QIV	
RSV2+Flu	147	18 – 49 years	RSV MAT 120 (Lot 2) Flu D-QIV	
RSV3+Flu	147	18 – 49 years	RSV MAT 120 (Lot 3) Flu D-QIV	
Flu+P	440	18 – 49 years	Flu D-QIV placebo	

RSV MAT 120 =RSVPreF3 vaccine
*Standard of care vaccine administered only to ensure the participants receive the standard of care.

2. STATISTICAL HYPOTHESES

The study includes 2 confirmatory co-primary objectives:

- To demonstrate the lot-to-lot consistency of 3 lots of RSVPreF3 vaccine in terms of geometric mean concentration (GMC) of RSVPreF3 IgG ELISA at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: at least one of the two-sided 95% confidence intervals for the 3 pair-wise GMC ratios between 2 lots are outside the range of 0.67 to 1.5

  H_a: $0.67 < \mu_{lot1}/\mu_{lot2} < 1.5$ and $0.67 < \mu_{lot1}/\mu_{lot3} < 1.5$ and $0.67 < \mu_{lot2}/\mu_{lot3} < 1.5$

  where μ represents the GMC of RSVPreF3 IgG at Day 31 from each lot. The lot-to-lot consistency will be demonstrated if two-sided 95% confidence intervals for the 3 pair-wise GMC ratios at Day 31 falls within 0.67 and 1.5.

- To demonstrate the non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone in terms of geometric mean titer (GMT) of Flu D-QIV HI antibody against the 4 influenza strains (A/Victoria/2570/2019 (H1N1) IVR-215, A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019) at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $\mu_{\text{co-ad group}}/\ \mu_{\text{alone}} \leq 0.67$ vs H_a: $\mu_{\text{co-ad group}}/\mu_{\text{alone}} > 0.67$

  where μ represents the GMT of Flu D-QIV HI antibody at Day 31. The non-inferiority will be demonstrated if the lower limits of the two-sided 95% CI on the GMT ratio (co-ad group RSV+Flu with 3 lots pooled divided by Flu+P group) is greater than 0.67 at Day 31 post vaccination for all the 4 strains (A/Victoria/2570/2019 (H1N1) IVR-215, A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019).

In addition, the study also includes 2 confirmatory secondary objectives. These hypothesis tests will be conducted sequentially only if the primary objective of non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone is met.

- To demonstrate the non-inferiority of RSVPreF3 vaccine when co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of GMT of RSV A neutralizing antibody at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $\mu_{\text{co-ad group}}/\ \mu_{\text{alone}} \leq 0.67$ vs H_a: $\mu_{\text{co-ad group}}/\mu_{\text{alone}} > 0.67$

  where μ represents the GMT of RSV A neutralizing antibody at Day 31. The non-inferiority will be demonstrated if the lower limits of the two-sided 95% CI on the GMT ratio (co-ad group RSV+Flu divided by RSV alone group with 3 lots pooled in both groups) is greater than 0.67 at Day 31 post vaccination.

- To demonstrate the non-inferiority for each strain (A/Victoria/2570/2019 (H1N1) IVR-215, A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019) of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone in terms of seroconversion rate (SCR) at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $p_{\text{alone}} - p_{\text{co-ad group}} \geq 10\%$ vs H_a: $p_{\text{alone}} - p_{\text{co-ad group}} < 10\%$

  where p represents the SCR for each influenza strain at Day 31. The non-inferiority for each strain will be demonstrated if the upper limits of the two-sided 95% CI on the SCR difference (Flu+P group minus co-ad group RSV+Flu with 3 lots pooled) is less than 10% at Day 31 post vaccination.

2.1. Multiplicity Adjustment

To ensure the global type I error for the primary objectives is controlled at 5%, each of the primary objectives will be tested using a nominal one-sided alpha of 2.5%.

If the primary objective of non-inferiority of Flu D-QIV in terms of GMT is met, the secondary confirmatory objective of non-inferiority of RSVPreF3 vaccine in terms of GMC will be tested at alpha of 2.5%, if this objective is met, the non-inferiority of Flu D-QIV in terms of SCR will be tested at alpha of 2.5% for each strain, otherwise, it will not be tested.

3. ANALYSIS SETS

Table 3 Analysis sets

Analysis Set	Definition / Criteria	Analyses Evaluated
Enrolled	Participants who agreed to participate in a clinical study after completion of the informed consent process	Study Population
Exposed	All Participants who received at least 1 dose of the study treatment. The allocation in a group is done as a function of the administered treatment	Safety, Study Population
Full Analysis Set (FAS)	All Participants who received at least 1 dose of the study intervention and have post-vaccination immunogenicity data It will be defined by timepoint, e,g. FAS at Day 31	Immunogenicity
Per-Protocol Set (PPS)	All Participants who received at least 1 dose of the study treatment to which they are randomized and have post-vaccination data minus participants with protocol deviations that lead to exclusion It will be defined by timepoint, e,g. PPS at Day 31	Immunogenicity
Solicited Safety Set	All Participants who received at least 1 dose of the study intervention (Exposed Set) who have solicited safety data	Solicited events
Randomized Set	Randomized set will include all participants who are randomized. The allocation in a group is done as function of the randomized intervention. Please note this set was not included in the protocol, but will be used later in one summary analysis, so it is added here for clarification.	Disposition

3.1. Criteria for eliminating data from Analysis Sets

Elimination codes are used to identify participants to be eliminated from analysis. Detail is provided below for each set.

3.1.1. Elimination from Exposed Set (ES)

Code 1030 (Study vaccine not administered at all), 800 (Fraudulent data) and code 900 (invalid informed consent) will be used for identifying participants eliminated from ES

3.1.2. Elimination from Full Analysis Set (FAS)

A participant will be excluded from the FAS under the following conditions

Table 4 Elimination code and condition

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
800	Fraudulent data	All	All
900	Invalid informed consent	All	All
1030	Study vaccine not administered at all	All	Safety, immunogenicity
2100.Vx	Serological results not available post-vaccination	Visit 2/Day 31	Immunogenicity

Vx indicates participants whose immunogenicity data will be eliminated from a specific visit.

3.1.3. Elimination from Per-protocol Set (PPS)

A participant will be excluded from the PPS under the following conditions

Table 5 Elimination code and condition

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
800	Fraudulent data	All	All
900	Invalid informed consent	All	All
1030	Study vaccine not administered at all	All	Safety, immunogenicity
1040.Vx*	Administration of concomitant vaccine(s) forbidden in the protocol	Visit 2/Day 31	Immunogenicity
1050	Randomisation failure	All	Immunogenicity
1060	Randomisation code was broken	All	Immunogenicity

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
1070**	Subjects got vaccinated with the correct vaccine but containing an incorrect volume	All	Immunogenicity
1070**	Vaccination not according to protocol (site of injection, route of administration, wrong replacement of study treatment administered)	All	Immunogenicity
1070**	Study treatment not prepared as per protocol (e.g., reconstitution)	All	Immunogenicity
1070**	Other deviations related to wrong study treatment/administration/dose	All	Immunogenicity
1070**	Study treatment administered while contraindication	All	Immunogenicity
1080	Vaccine temperature deviation	All	Immunogenicity
1090	Expired vaccine administered	All	Immunogenicity
2010	Protocol violation (inclusion/exclusion criteria) DOB – VAC – 18-49 years BMI <=40 kg/m^2	All	Immunogenicity
2040.Vx*	Administration of any medication forbidden by the protocol	Visit 2/Day 31	Immunogenicity
2050.Vx*	Intercurrent medical conditions which are exclusionary as per protocol	Visit 2/Day 31	Immunogenicity
2090.Vx	Subjects did not comply with blood sample schedule: • For PPS at Day 31, check the interval from vaccination to day 31 BS = 31 – 38 days;	Visit 2/Day 31	Immunogenicity
2100.Vx	Serological results not available post-vaccination	Visit 2/Day 31	Immunogenicity
2120.Vx	Obvious incoherence or abnormality or error in data	All	Immunogenicity
2130.Vx	Testing performed on samples not aligned with ICF	All	Immunogenicity

*Attribution of these elimination codes to subject need CRDL review of individual listing
** Attribution of code 1070 to a subject requires CRDL confirmation
Vx+ indicates participants whose immunogenicity data will be eliminated from a specific visit onwards; Vx indicates participants whose immunogenicity data will be eliminated from a specific visit.
DOB-Date of Birth, VAC-Vaccination, BS- Blood Sample, BMI-Body Mass Index.

3.1.4. Elimination from solicited safety set

Code 1030 (Study vaccine not administered at all), code 800 (fraudulent data) and code 900 (invalid informed consent) and code 1160 (no post-vaccination solicited safety data) will be used for identifying participants eliminated from the solicited safety set.

4. STATISTICAL ANALYSES

4.1. General Considerations

Unless otherwise specified, reactogenicity analysis and safety analysis will be performed on solicited safety set and Exposed Set respectively.

In general, Immunogenicity analysis will be performed on the Per Protocol set. If, in any study group and at any timepoint, the percentage of vaccinated participants with immunogenicity results excluded from the Per Protocol set is 5% or more, a second analysis based on the Full Analysis Set will be performed to complement the Per Protocol analysis for some immunogenicity analysis.

4.1.1. General Methodology

Unless otherwise specified, continuous data will be summarized using descriptive statistics: n, mean, standard deviation (std), median, minimum and maximum. Categorical data will be summarized as the number and percentage of participants in each category.

95% CI for proportion will be based on exact Clopper-Pearson confidence interval [Clopper, 1934].

95% CI for group difference in proportion will be based on Miettinen and Nurminen confidence interval [Miettinen, 1985].

95% CI for GMT(C) will be based on a back transformation of student confidence interval for the mean of log-transformation.

95% CI for GMT(C) ratio between groups will be based on a back transformation of confidence interval for the mean difference on log-transformation.

For a given participant and given immunogenicity measurement, missing or non-evaluable measurements will neither be imputed nor be replaced, and therefore will not be included in immunogenicity analysis.

For between group statistical modelling analysis, participants having a result at both the baseline and the considered timepoint will be included in the analysis.

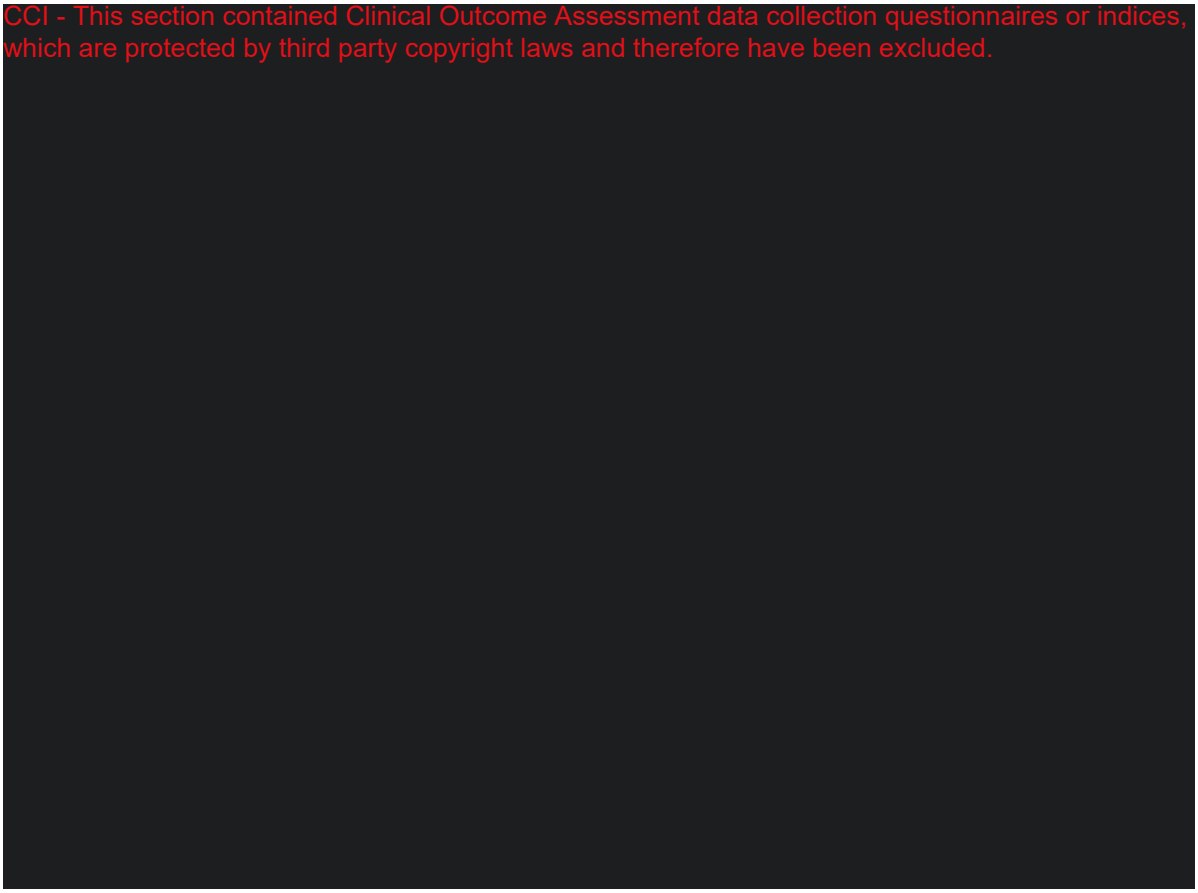
Participants who prematurely withdrew from study will not be replaced.

4.1.2. Definition

Solicited events and their intensity scales are defined as below:

Table 6 Intensity scales for solicited events

CCI - This section contained Clinical Outcome Assessment data collection questionnaires or indices, which are protected by third party copyright laws and therefore have been excluded.



Baseline: For all endpoints the baseline value will be the latest pre-dose assessment with a non-missing value, including those from unscheduled visits. If time is not collected, Day 1 assessments are assumed to be taken prior to vaccination and used as baseline. Unless otherwise stated, if baseline data is missing no derivation will be performed and baseline will be set to missing.

For the RSV and FLU antigens:

- A seronegative participant will be defined as a participant whose antibody titer/concentration is below the cut-off value of the assay.
- A seropositive participant is a participant whose antibody titer/concentration is greater than or equal to the cut-off value of the assay.
- MGI (mean geometric increase): The geometric mean of the within participant ratios of the post-dose titer over the pre-dose titer.

Please refer to Section 6.2.4.6 for cut-off value for each assay.

For the FLU antigens:

- SCR (seroconversion rate): The percentage of participants who have either a HI pre-dose titer < 1:10 and a post-dose titer ≥ 1:40 or a pre-dose titer ≥1:10 and at least a four-fold increase in post-dose titer.
- SPR (seroprotection rate): The percentage of participants with a serum HI titer ≥1:40 that usually is accepted as indicating protection.

4.2. Primary Endpoint(s) Analyses

4.2.1. Safety analysis

The analysis of solicited administration site event (associated with RSV or Flu) and solicited systemic event will be performed on solicited safety set.

- The number and percentage with exact 95% CI of participants reporting each solicited administration site event (any grade, each grade) and solicited systemic event (any, each grade) during the 7-day follow-up period after dosing will be tabulated by maximum intensity per participant for each study group.
- For fever during the 7-day follow-up period after vaccination, the number and percentage of participants reporting any fever (i.e., temperature ≥38 °C) and fever by half degree (°C) cumulative increments, any Grade 3 fevers, will be reported.
- Duration in days of solicited administration site and systemic events within 7 days after vaccination will be tabulated by study group and overall. The derivation rule of duration in days for solicited events is detailed in section 6.2.4.9.

The analysis of unsolicited AEs and SAEs will be performed on ES.

- The number and percentage of participants reporting unsolicited AEs within 30 days after dosing with exact 95% CIs will be tabulated by group and by Medical Dictionary for Regulatory Activities (MedDRA) terms.
- Similar tabulations will be done for Grade 3 unsolicited AEs, for any causally related unsolicited AEs, and for Grade 3 related unsolicited AEs.
- The number and percentage of participants reporting at least one SAE within 30 days after dosing with exact 95% CIs will be tabulated by group and by MedDRA terms.
- By-participant listings of SAEs and AEs leading to study withdrawal will be prepared.

4.2.2. Immunogenicity analysis

- Lot-to-lot consistency analysis

  The GMC ratio and its two-sided 95% CI between 2 lots in terms of RSVPreF3 IgG ELISA concentration at Day 31 will be computed by back transformation of its mean difference and 95% CI on log10 transformed scale estimated from the t-distribution.

Summary table will show the number of observations included in the calculation for each lot, GMC of each lot, and 3 pair-wise GMC ratios with corresponding 95% CIs.

Lot-to-lot consistency will be demonstrated if all 3 two-sided 95% CIs for GMC ratio fall within 0.67 and 1.5.

- Non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone with respect to 4 Flu D-QIV strains at Day 31

  The GMT ratio and its two-sided 95% CIs between the 2 groups (RSV+Flu with 3 lots pooled divided by Flu+P group) for A/Victoria/2570/2019 (H1N1) IVR-215, A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019 will be computed by back transformation of its mean difference and 95% CI on log10 transformed scale estimated from the t-distribution.

  Summary table will show for each group the number of observations included in the calculation and GMT value, and between group GMT ratio with corresponding 95% CI.

  Non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine will be demonstrated if lower bounds of all 4 two-sided 95% CIs are above 0.67.

4.2.3. Sensitivity analyses

For primary immunogenicity endpoints, between study group comparisons will also be explored through statistical modelling.

- Lot-to-lot consistency analysis

  The ANCOVA model on the log10 transformation of the concentrations will be explored, and will include the lot as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination concentrations as covariates. Covariates in the model may be further adjusted when performing the analysis if appropriate and needed, therefore, the following SAS code can serve as a reference and may be adjusted according to the analysis needs.

```
PROC GLM data=sero;
  CLASS group age_cat;
     MODEL log_val = baseline group age_cat group*age_cat;
      LSMEANS group/pdiff cl alpha=0.05;
     RUN;
```

  where log_val represents the log-transformed RSVPreF3 IgG antibody concentration at Day 31, baseline is pre-vaccination RSVPreF3 IgG antibody level on log-transformed scale, group includes each lot, age_cat is the age category at vaccination (18-32 vs. 33-49).

  The 3 pair-wise GMC ratios between 2 lots and their two-sided 95% CI will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

Summary table will show the number of observations included in the model for each lot, adjusted GMC of each lot, and 3 pair-wise adjusted GMC ratios with corresponding 95% CIs.

- Non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone with respect to 4 Flu D-QIV strains at Day 31

  The ANCOVA model on the log10 transformation of the titers will be explored, and will include the study group (RSV+Flu with 3 lots pooled, Flu+P) as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination titers as covariates, covariates in the model may be further adjusted when performing the analysis if appropriate and needed. Similar SAS code could be referenced and may be further adjusted at time of analysis:

```
PROC GLM data=sero;
  CLASS group age_cat;
     MODEL log_val = baseline group age_cat group*age_cat;
      LSMEANS group/pdiff cl alpha=0.05;
     RUN;
```

  where log_val represents the log-transformed antibody level at Day 31, baseline is pre-vaccination antibody level on log-transformed scale, group includes RSV+Flu with 3 lots pooled and Flu+P, age_cat is the age category at vaccination (18-32 vs. 33-49).

  The GMT ratio and its two-sided 95% CIs between the 2 groups (RSV+Flu with 3 lots pooled divided by Flu+P group) for A/Victoria/2570/2019 (H1N1) IVR-215, A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019 will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

  Summary tables will show for each group the number of observations included in the model and model adjusted GMT, and between group adjusted GMT ratios with the corresponding 95% CIs.

4.3. Secondary Endpoint(s) Analyses

4.3.1. Safety analysis

The analysis of SAEs from the first vaccination up to study end will be performed on ES.

- The number and percentage of participants reporting at least one SAE from vaccination up to study end with exact 95% CIs will be tabulated by group and by MedDRA terms
- By participant listings of SAEs and (S)AEs leading to study withdrawal will be prepared.

4.3.2. Immunogenicity analysis

4.3.2.1. Secondary confirmatory analysis

In order to control the global type I error, the objectives will be assessed sequentially.

- Non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of RSV A neutralizing antibody titers at Day 31

  The GMT ratio and its two-sided 95% CI between 2 groups (RSV+Flu group with 3 lots pooled divided by RSV alone group with 3 lots pooled) will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from t-distribution.

  Summary table will show for each group the number of observations included in the calculation and GMT value, and between group GMT ratio with corresponding 95% CI.

  No interference on RSVPreF3 vaccine from quadrivalent seasonal influenza vaccine (Flu D-QIV) will be demonstrated if lower bound of the two-sided 95% CI of GMT ratio is above 0.67.

- Non-inferiority for each of the 4 strains of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone at Day 31 in terms of Seroconversion rate (SCR)

  The SCR difference and its two-sided 95% CI between 2 groups (Flu+P group minus RSV+Flu group with 3 lots pooled) will be calculated for each strain (A/Victoria/2570/2019 (H1N1) IVR-215, A/Tasmania/503/2020 (H3N2) IVR-221, B/Washington/02/2019) at Day 31. 95% CI calculation will be based on Miettinen and Nurminen confidence interval [Miettinen, 1985].

  Summary table will show for each group the number of observations included in the calculation, number and percentage of seroconverted participants, and between group SCR difference with corresponding 95% CI.

  For each strain, non-inferiority will be demonstrated if the upper bound of the two-sided 95% CI is less than 10% at Day 31 post vaccination.

4.3.2.2. Descriptive immunogenicity analysis

For RSV A, RSV B neutralizing antibody titers and RSVPreF3 IgG antibody concentrations at pre-vaccination and Day 31 from each lot (RSV1, RSV2, and RSV3), as well as for the group of RSV alone with 3 lots pooled (RSV1, RSV2 and RSV3 pooled) and RSV+Flu group with 3 lots pooled, the following analysis will be performed:

- Number and percentage of participants with antibody titers/concentrations above assay cut-off will be tabulated with its exact 95% CI by group.
- GMT/GMCs will be tabulated with 95% CI at each timepoint and represented graphically by group.
- MGI from Day 31 over pre-vaccination will be tabulated with 95% CI by group.

For Flu D-QIV HI antibody titers against 4 influenza strains at pre-vaccination and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine with 3 lots pooled (RSV+Flu) and the group of Flu D-QIV vaccine administered alone (Flu+P), the following analysis will be performed:

- Number and percentage of participants with antibody titers above assay cut-off and their exact 95% CI will be tabulated by group.
- GMTs will be tabulated with 95% CI at each timepoint and represented graphically by group.
- MGI from Day 31 over pre-vaccination will be tabulated with 95% CI by group.
- Number and percentage of participants achieving seroconversion for HI antibody at Day 31 will be tabulated with its exact 95% CI by group.
- Number and percentage of participants achieving HI antibody titer ≥ 1:40 at pre-vaccination and Day 31 will be tabulated with its exact 95% CI by group.

4.3.3. Sensitivity analyses

ANCOVA modelling analysis will be explored for non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of RSV A neutralizing antibody titers at Day 31.

The ANCOVA model will be based on the log10 transformation of the titers, and will include the study group (RSV+Flu with 3 lots pooled, RSV alone group with 3 lots pooled) as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination titers as covariates, covariates in the model may be further adjusted when performing the analysis if appropriate and needed. Similar SAS code will be explored and may be further adjusted at time of analysis:

```
PROC GLM data=sero;
  CLASS group age_cat;
  MODEL log_val = baseline group age_cat group*age_cat;
  LSMEANS group/pdiff cl alpha=0.05;
RUN;
```

where log_val represents the log-transformed RSV A neutralizing antibody titer at Day 31, baseline is pre-vaccination RSV A neutralizing antibody titer on log-transformed scale, group includes Flu+RSV with 3 lots pooled and RSV alone with 3 lots pooled, age_cat is the age category at vaccination (18-32 vs. 33-49).

The GMT ratio and its two-sided 95% CIs between the 2 groups (RSVPreF3+Flu D-QIV with 3 lots pooled divided by RSVPreF3 alone group with 3 lots pooled) will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

Summary tables will show for each group the number of observations included in the model and model adjusted GMT, and between group adjusted GMT ratios with the corresponding 95% CIs.

4.4. Tertiary Endpoint(s) Analyses

Not applicable

4.5. Other Safety Analyses

Other safety analyses will be based on the Exposed Set, unless otherwise specified.

4.5.1. Combined solicited and unsolicited events

For clintrial.gov and EudraCT posting purposes, a summary of combined solicited and unsolicited adverse events will be produced by System Organ Class and preferred terms and according to occurrence of each event.

Solicited administration site events and solicited systemic events will be coded by MedDRA as per the following codes

Solicited symptom	Lower level term code	Corresponding Lower level term decode
Pain	Injection site pain	10022086
Redness	Injection site redness	10022098
Swelling	Injection site swelling	10053425
Fatigue	Fatigue	10016256
Fever	Fever	10016558
Nausea	Nausea	10028813
Vomiting	Vomiting	10047700
Diarrhea	Diarrhea	10012727
Abdominal pain	Abdominal pain	10000081
Headache	Headache	10019211

Please note: the coding will be double checked during the analysis

4.5.2. COVID-19 Assessment and COVID-19 AEs

A participant is defined as having a suspected, probable or confirmed COVID-19 infection during the study if the answer is “Confirmed”, “Probable” or “Suspected” to the case diagnosis question from the COVID-19 coronavirus infection assessment eCRF.

Numbers and percentage of participants with a suspected, probable or confirmed COVID-19 infection will be summarized by group based on ES.

Number and percentage of participants who had a COVID-19 test performed and number and percentage of participants with positive, negative and indeterminate results will be summarized by group on ES.

4.5.3. Additional Safety Assessments (if applicable)

Vital signs will be summarized by group using descriptive statistics at all timepoint(s) the information is collected on ES and PPS. The parameters include but may not be limited to systolic blood pressure, diastolic blood pressure, temperature, heart rate, respiratory rate, height, weight and body mass index.

4.6. Other Analyses

Not applicable.

4.7. Changes to Protocol Defined Analyses

An interim analysis was planned in the protocol (Dated: 17-NOV-2021) but removed in this statistical analysis plan amendment 1. A final and inclusive study analysis will be performed when all data are available. An end of study report containing all available data will be written and made available to the investigators.

5. SAMPLE SIZE DETERMINATION

Approximately 1541 participants will be randomized to achieve appropriately 1400 evaluable participants.

Assessments of both immunogenicity and safety data were considered when determining sample size for this study.

Participants who withdraw from the study will not be replaced.

The sections below describe the assumptions, adjustment and methodology used for the sample size calculation.

5.1. Primary objective for Lot-to-Lot consistency

The primary objective of lot-to-lot consistency will be evaluated on the RSVPreF3 immunogenicity as measured by RSVPreF3 IgG ELISA concentration at Day 31.

The lot-to-lot consistency will be demonstrated only if two-sided 95% confidence intervals for the 3 pair-wise geometric mean ratios of RSVPreF3 IgG ELISA concentration at Day 31 (30 days post vaccination) falls within 0.67 and 1.5.

With the assumptions of a SD of log10 transformed RSVPreF3 IgG ELISA concentration of 0.35, true GMC ratio of 1 between 2 lots, type I error of 0.025, and 10% of non-evaluable rate, 220 enrolled participants per lot provides at least 99% global power to conclude lot-to-lot consistency (see details in Table 7).

Table 7 Power to demonstrate Lot-to-Lot consistency on immune responses of RSVPreF3 IgG ELISA concentration at Day 31 post vaccination

Endpoint	Clinically acceptable bounds for consistency	Group description	Number of evaluable participants in each lot	Reference SD**	Power
*RSVPreF3 IgG GMC ratio	(0.67, 1.5)	Lot 1/2	200	0.35	99.8%
*RSVPreF3 IgG GMC ratio	(0.67, 1.5]	Lot 1/3	200	0.35	99.8%
*RSVPreF3 IgG GMC ratio	(0.67, 1.5]	Lot 2/3	200	0.35	99.8%
Global power					99.3%

* Pass 2019, Two-Sample T-test for equivalence allowing unequal variance, alpha=2.5%;
**References used for the sample size calculation: Study *RSV MAT 011* (209141)

5.2. Primary objective: To demonstrate Non-inferiority of immune response of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine or administered alone at Day 31 post dose vaccination

Possible interference of RSVPreF3 on influenza immune responses will be evaluated by using Flu D-QIV HI antibody titers against the 4 influenza strains from RSV+Flu (pooled of RSV1+Flu, RSV2+Flu, RSV3+Flu) and Flu+P group at Day 31. In the description of the subsequent analysis, the RSV+Flu vaccine group are RSV1+Flu, RSV2+Flu, RSV3+Flu groups pooled.

The hypothesis is that co-administration of RSVPreF3+ Flu D-QIV vaccine is non-inferior to Flu D-QIV vaccine with respect to GMT ratio for immune response of influenza antigen in non-pregnant women at Day 31 post vaccination.

The criteria to evaluate non-inferiority for A/Victoria/2570/2019 (H1N1) IVR-215, A/Tasmania/503/2020 (H3N2) IVR-221 and B/Washington/02/2019 is that the lower limits of the 95% CI on the GMT ratio (RSV+Flu divided by Flu+P) is greater than 0.67 at Day 31 post vaccination.

Table 8 presents the power on the ratio of the GMT of Flu D-QIV 4 strains. The power is at least 94% when the sample size is 400 participants per group with standard deviation (SD) of 0.6 on its log10 transformation and non-inferiority margin of 0.67 between RSVPreF3 co-administrated with Flu D-QIV group and Flu D-QIV vaccine administered alone group.

Table 8 Power to demonstrate non-inferiority of immune response between Flu D-QIV vaccine co-administered with RSVPreF3 or administered alone in term of GMT for Flu D-QIV strains at Day 31 post 1st vaccination with assumed GMT ratio 1

Endpoint	NI criteria	Number of evaluable participants in each group	Reference*	Power
A/Victoria/2570/2019 (H1N1) IVR-215 GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
B/Washington/02/2019 GMT ratio	LL of 95% CI for GMC ratio >0.67	400	0.60	98.56%
A/Tasmania/503/2020 (H3N2)** IVR-221 GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
B/Phuket/3073/2013 GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
Global power				94.24%

By Pass 2019, Non-Inferiority Tests for Two Means using Differences, one-sided alpha=2.5%;
*References used for the sample size calculation: Study Flu D-QIV -008 E1_01 Table 34
**A/Tasmania/503/2020 (H3N2) IVR-221 (an A/Cambodia/e0826360/2020 (H3N2)-like virus).

5.3. Secondary objectives

In order to control the global type I error, the objectives will be assessed sequentially.

5.3.1. To demonstrate the non-inferiority of immune response between RSVPreF3 co-administered with Flu D-QIV and RSVPreF3 alone in term of RSV A Neutralizing antibody titers at Day 31 post administration

Hypothesis under consideration is that immune response when the co-administration of RSVPreF3 vaccine and Flu D-QIV vaccine is non-inferior to that when RSVPreF3 vaccine (pooled RSV1, RSV2 and RSV3 group) is administered alone with respect to GMT ratio for RSV A neutralizing antibody titer in non-pregnant women at Day 31 post vaccination.

This hypothesis test is conducted only if the primary objective of non-inferiority of immune response between Flu D-QIV vaccine when co-administered with RSVPreF3 and Flu D-QIV vaccine administered alone is met.

The criteria to evaluate non-inferiority for RSV A Nab is that the lower limits of the 95% CI on the GMT ratio (RSV+Flu group divided by RSV alone group with 3 lots pooled in both groups) is greater than the pre-defined clinical limit of 0.67 at Day 31 post vaccination.

If a SD of log10 transformed RSV A Nab of 0.4 is assumed, with an assumption of GMT ratio of 1.0, there will be at least 99% chance that the lower bound of 95% CI for the ratio of GMT of RSV A Nab titer between RSVPreF3 co-administration with Flu D-QIV and administered alone is above 0.67 (Table 9)

Table 9 Power to demonstrate non-inferiority of immune response of RSVPreF3 vaccine when co-administered with Flu D-QIV vaccine or administered alone at Day 31 post vaccination

Endpoint	NI criteria	N1:N2 (evaluable)	Reference*	Power
RSV A GMT ratio	LL of 95% CI for GMT ratio >0.67	400:600	0.40	99.99%

By Pass 2019 Non-Inferiority Tests for Two Means using Differences, one-sided alpha=2.5%;
*References used for the sample size calculation: Study RSV MAT 011 (209141)
N1: Number of evaluable participants in the co-administration of RSVPreF3 vaccine + Flu D-QIV vaccine.
N2: Number of evaluable participants in the RSVPreF3 vaccine alone group with 3 lots pooled.

5.3.2. To demonstrate the non-inferiority of immune response between RSVPreF3 co-administered with Flu D-QIV and Flu D-QIV alone in term of SCR at Day 31 post 1st dose administration

Potential interference of RSVPreF3 on influenza immune responses will be evaluated by comparing the Day 31 seroconversion rate of Flu D-QIV HI antibody titers against the 4 influenza strains between RSV+Flu group and Flu+P group.

The criteria to evaluate non-inferiority with respect to the SCR difference for Flu D-QIV antibody titers against 4 influenza strains is that the upper limits of the 95% CI on the SCR difference (Flu+P group minus RSV+Flu) is less than the pre-defined clinical limit of 10% at Day 31 post vaccination.

With the assumptions of SCR for the reference group as indicated in Table 10, significance level 0.025, non-inferiority margin 10%, 400 evaluable participants (10% non-evaluable rate) provides at least 80% power to conclude non-inferiority for each strain (Table 10).

Table 10 Powers to demonstrate non-inferiority in terms of HI antibody SCR for Flu D-QIV strains between Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and administered alone at Day 31 post vaccination

Endpoint	NI criteria	Anticipated SCR*	Number of participants per group (evaluable)	Power
A/Victoria/2570/2019 (H1N1) IVR-215 SCR	10%	60%	400	82.49%
B/Washington/02/2019 SCR	10%	65%	400	84.39%
A/Tasmania/503/2020 (H3N2)** IVR-221 SCR	10%	65%	400	84.39%
B/Phuket/3073/2013 SCR	10%	65%	400	84.39%

By Pass 2019, Non-Inferiority Tests for the difference between two proportions, alpha=2.5%
*SCR used in the reference group for power calculation is from Study Tdap-0.3-008, Suppl. Table 36.
**A/Tasmania/503/2020 (H3N2) IVR-221 (an A/Cambodia/e0826360/2020 (H3N2)-like virus).

6. SUPPORTING DOCUMENTATION

6.1. Appendix 1 Study Population Analyses

6.1.1. Participant Disposition

Summary of participant disposition will be performed on ES. The number and percentage of participants who completed the study and who prematurely withdrew from the study including the reasons for study withdrawal will be summarized by each group and overall.

6.1.2. Demographic and Baseline Characteristics

The analysis of demography will be performed on Enrolled Set, ES and PPS using descriptive statistics. The parameters including but may not be limited to age, age category (18-32 vs 33-49), ethnicity, race, country and childbearing potential will be summarized by group.

Summary of past medical history and current medical conditions will be performed on ES by Medical Dictionary for Regulatory Activities (MedDRA) term. Un-coded medical conditions or medical history will be summarized under 'Other' category.

Vaccination history will be coded using GSK Drug dictionaries. Summary of vaccination history will be performed on ES by group.

6.1.3. Protocol Deviations

Important protocol deviations will be summarized based on ES by group.

Protocol deviations will be tracked by the study team throughout the conduct of the study. These protocol deviations will be reviewed to identify those considered as important as follows:

- Data will be reviewed prior to unblinding and freezing the database to ensure all important deviations (where possible without knowing the study intervention details) are captured and categorised in the protocol deviations dataset.
- This dataset will be the basis for the summaries of important protocol deviations.
- The summary will include number and percentage of participants with important protocol deviations by deviation category for each study group.
- An individual listing of protocol deviation will also be provided.

Protocol deviations which result in exclusion from the analysis set will be summarized on ES by group.

- Data will be reviewed prior to unblinding and freezing the database to ensure all deviations leading to analysis population exclusions are captured and categorised in the protocol deviations ADaM dataset (note these exclusions are not captured in the SDTM dataset).
- The summary will include number and percentage of participants with protocol deviations leading to exclusion by deviation category for each study group.

In addition to the above summary, separate summaries may be produced for important protocol deviations related to COVID-19, and important protocol deviations not related to COVID-19 respectively.

6.1.4. Concomitant Medications and Vaccinations

Concomitant medications and vaccinations will be coded and summarized using GSK Drug dictionary.

- The number and percentage of participants taking concomitant medications (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) will be summarized by group. A listing will also be provided.
- The number and percentage of participants taking concomitant vaccinations will be summarized by group. A listing will also be provided.

6.1.5. Additional Analyses Due to the COVID-19 Pandemic

Depending on how the Covid-19 situation evolves, the SAP might be amended to reflect the analysis corresponding to Covid-19.

6.2. Appendix 2 Data Derivations Rule

6.2.1. Study Day and Reference Dates

The safety reference date is the study intervention start date and will be used to calculate study day for safety measures.

The study day is calculated as below:

- Assessment Date = Missing → Study Day = Missing
- Assessment Date < Reference Date → Study Day = Assessment Date – Ref Date
- Assessment Data ≥ Reference Date → Study Day = Assessment Date – Ref Date + 1

6.2.2. Attributing events to vaccine doses

The dose relative to an event is the most recent study dose given to a participant prior to the start of a given event. For example, if the start date of an adverse event is between Dose 1 and Dose 2, the relative dose will be Dose 1.

If an event starts on the same day as a study dose, the relative dose will be derived from the additional information provided in the case report form (CRF) using the contents of the flag indicating if the event occurred before or after study dose. If 'after study dose' is selected, the relative dose for the event will be the one administered on the start day of the event. If 'before study dose' is selected, the relative dose for the event will be the dose prior to this one.

6.2.3. Handling of missing data

6.2.3.1. Dates

When partially completed dates (i.e. dates missing a day and/or month) are used in calculations, the following standard rules will be applied:

- A missing day will be replaced by 15
- A missing day and month will be replaced by June 30th.

The following exceptions apply:

- Adverse event start dates with missing day:
  - If the event starts in the same month as at least one of the study doses, the contents of AE.AESTRTPT (the flag indicating if the event occurred before or after study dose) will be used to complete the date. If 'after study dose' is selected, the imputed start date will match the first (or only) study dose given during that month. If 'before study dose' is selected, the imputed date will be one day before the first (or only) study dose given during that month.
- Adverse event start dates with missing day and month:
  - If the event starts in the same year as at least one of the study doses, the contents of AE.AESTRTPT (the flag indicating if the event occurred before or after study dose) will be used to complete the date. If 'after study dose' is selected, the imputed start date will match the first (or only) study dose given during that year. If 'before study dose' is selected, the imputed date will be one day before the first (or only) study dose given during that year.

All other cases of incomplete AE or concomitant medication/vaccination start date will follow the standard rules above.

6.2.3.2. Daily recording of solicited events

6.2.3.2.1. Studies with electronic diaries

For studies using electronic diaries for the collection of solicited events, a solicited event will be considered present only when a daily recording of grade 1 or more is present.

6.2.3.3. Unsolicited adverse events

Unsolicited adverse event summaries are including serious adverse events unless specified otherwise.

Missing severity, relationship with study vaccine, and outcome of unsolicited adverse events will not be replaced and will appear as 'UNKNOWN' when displayed in a statistical output.

6.2.4. Data derivation

6.2.4.1. Age at first dose in years

When age at first dose is to be displayed in years, it will be calculated as the number of complete calendar years between the date of birth and the date of first dose. For example:

DOB = 10SEP1983, Date of first dose = 09SEP2018 -> Age = 34 years

DOB = 10SEP1983, Date of first dose = 10SEP2018 -> Age = 35 years

6.2.4.2. Weight

Weight will be presented in kilograms. Weights reported in pounds will be converted as follows:

Weight in kilograms = Weight in pounds / 2.2

6.2.4.3. Height

Height will be presented in centimeters. Heights reported in feet and inches will be converted as follows:

Height in centimeters = Height in inches x 2.54

6.2.4.4. Body mass index (BMI)

BMI will be calculated as follows:

BMI = (Weight in kilograms) / (Height in meters)2

6.2.4.5. Temperature

Temperatures will be presented in degrees Celsius (°C). Temperatures reported in degrees Fahrenheit (°F) will be converted as follows:

Temperature (Celsius) = ((Temperature (Fahrenheit) - 32) x 5)/9

6.2.4.6. Numerical serology results

Numerical serology results will be derived from the content of IS.ISORRES in the SDTM dataset. For assays with a specific cut-off, the following derivation rules apply:

IS.ISORRES	Derived value
"NEG", "-", or "(-)"	cut-off/2
"POS", "+", or "(+)"	cut-off
"< value" and value is <= assay cut-off	cut-off/2
"< value" and value is > assay cut-off	value
"> value" and value is < assay cut-off	cut-off/2
"> value" and value is >= assay cut-off	value
"value" and value is < cut-off	cut-off/2
"value" and value is >= cut-off	value
All other cases	missing

The cut-off tests for immunogenicity evaluation will be as per following:

System	Component	Method	Unit	Cut-off *(LLOQ)*	*ULOQ*
Serum	RSV-A Neutralising Antibody	NEUTRALISATION	ED60	18	*123535*
Serum	RSV-A Neutralising Antibody	NEUTRALISATION	IU/mL	56	*217400*
Serum	RSVPreF3 IgG antibody concentrations	ELISA	EU/mL	*25*	*251769*
Serum	RSV-B Neutralising Antibody	NEUTRALISATION	ED60	*30*	*138336*
Serum	RSV-B Neutralising Antibody	NEUTRALISATION	IU/mL	*44*	*171279*
Serum	A/Victoria/2570/2019 (H1N1)	HI assay	*TBD*	*TBD*	*TBD*
Serum	A/Tasmania/503/2020 (H3N2)	HI assay	*TBD*	*TBD*	*TBD*
Serum	B/Washington/02/2019	HI assay	*TBD*	*TBD*	*TBD*
Serum	B/Phuket/3073/2013	HI assay	*TBD*	*TBD*	*TBD*

Note: the assay cut-off (LLOQ), ULOQ and units may be further adjusted at time of analysis when notified by the lab.

6.2.4.7. Geometric mean titres (GMTs) and concentrations (GMCs)

Geometric Mean Titre (GMT) or Concentration (GMC) calculations are performed by taking the inverse logarithm of the mean of the log titre or concentration transformations. Non quantifiable antibody titres or concentrations will be converted as described in section 6.2.4.6 for the purpose of GMT/GMC calculation. The cut-off value is defined by the laboratory before the analysis.

6.2.4.8. Onset day

The onset day for an event (e.g. AE, concomitant medication/vaccination) is the number of days between the last study dose and the start date of the event. This is 1 for an event occurring on the same day as a study dose (and reported as starting after study dose).

6.2.4.9. Duration of events

The duration of an event with a start and end date will be the difference between the start and end date plus one day, i.e. an event that starts on 03MAR2018 and ends on 12MAR2018 has a duration of 10 days.

The duration of solicited events will be calculated as the sum of the individual days with the adverse event reported at grade 1 or higher during the solicited event period.

6.2.4.10. Counting rules for occurrences of solicited events

When the occurrences of solicited events are summarized, each event recorded as having occurred during a specific period will be counted as only one occurrence regardless of the number of days on which it occurs. Also, in the case of co-administered study interventions, an administration site event recorded for a participant following multiple study interventions will be counted as only one occurrence.

6.2.5. Display of decimals

6.2.5.1. Percentages

Percentages and their corresponding confidence limits will be displayed with one decimal except for 100% in which case no decimal will be displayed.

6.2.5.2. Differences in percentages

Differences in percentages and their corresponding confidence limits will be displayed with two decimals.

6.2.5.3. Demographic/baseline characteristics statistics

The mean, median, and standard deviation for continuous baseline characteristics (height, weight, body mass index (BMI), pre-dose body temperature) will be presented with one decimal.

The minimum and maximum values and quartile values (if required) will be presented with the same number of decimals as the observed values.

The maxima and minima of transformed height/weight variables will be displayed without decimals.

The maximum and minimum of transformed body temperatures will be displayed with one decimal.

6.2.5.4. Serological summary statistics

The number of decimals used when displaying geometric mean titers (GMT) or concentrations (GMC) and their confidence limits is assay specific based on the magnitude of the assay result post-dose and the clinically relevant assay threshold. The same number of decimals will be used for a given assay regardless of the timepoint presented.

Lowest clinically relevant threshold	Example	Number of decimals to display
<0.3	Diphtheria, tetanus, anti-PRP	3
>=0.3 and <4	*Streptococcus pneumoniae*, Meningococal bactericide	2
>=4 and <1000	Measles, rubella, varicella, polio	1
>=1000	CMI	0

GMT/GMC fold increase from pre-dose follows the same principle. Namely when the lowest clinically relevant threshold is 2 fold, 2 decimals are displayed while when the lowest clinically relevant threshold is 4 fold, 1 decimal is displayed.

GMT or GMC group ratios and their confidence limits will be displayed with 2 decimals regardless of the actual values.

7. REFERENCES

Clopper CJ, Pearson E. The Use of Confidence or Fiducial Limits Illustrated in the case of the Binomial. *Biometrika.* 1934;26:404-13.

Miettinen, O. S. and Nurminen, M. Comparative analysis of two rates. Statistics in Medicine, 1985;4,213-226.

Information Type:	Statistical Analysis Plan (SAP)

TITLE PAGE

Protocol Title: A Phase III, observer-blind, randomized, multi-country study to evaluate the lot-to-lot consistency of GSK's investigational RSV maternal vaccine and the immune response, safety and reactogenicity of RSV maternal vaccine when co-administered with GSK's quadrivalent influenza D-QIV vaccine in healthy non-pregnant women 18-49 years of age.

Study Number: 214709 (RSV MAT-010)

Compound Number: GSK3888550A

Abbreviated Title: A Phase III study to assess the lot-to-lot consistency of GSK's investigational RSV maternal vaccine and the immune response and safety of RSV maternal vaccine when given alone or co-administered with GSK's influenza D-QIV vaccine in healthy non-pregnant women.

Sponsor Name: GlaxoSmithKline Biologicals SA (GSK)

Regulatory Agency Identifier Number(s): IND 018434

Registry	**ID**
Clintrials.gov	Not available yet

TABLE OF CONTENTS

PAGE

LIST OF TABLES

PAGE

Version history

SAP Version	Approval Date	Protocol Version (Date) on which SAP is Based	Change	Rationale
SAP	12 Aug 2021	Amendment 1 (27 May 2021)	Not Applicable	Original version

1. INTRODUCTION

The purpose of this SAP is to describe the planned analyses for RSV MAT-010 (214709). Details of the planned analysis including the final analyses, are provided.

1.1. Objectives, Estimands and Endpoints

Table 1 Study objectives, endpoints and estimands

Objectives	Endpoints/Estimands
Primary	
Safety • To evaluate the safety and reactogenicity of RSVPreF3 when given alone (pooled lots) or co-administered with Flu D-QIV up to Day 31 post administration.	The number and percentage of participants in each group reporting ● Each solicited administration site AE collected during the 7 days follow-up period (Day 1 to Day 7) ● Each solicited systemic events in the 7 days follow-up period ● Unsolicited AEs collected during the 30 days follow-up period (Day 1 to Day 30) ● SAEs in the 30 days follow-up period (Day 1 to Day 30)
Immunogenicity • To demonstrate the lot-to-lot consistency of 3 lots of the investigational RSVPreF3 vaccine based on GMC of RSVPreF3 IgG ELISA at Day 31	RSVPreF3 IgG ELISA concentration at Day 31 (30 days post administration) Measured by ratio of Geometric mean concentration (GMC) between lots in terms of RSVPreF3 IgG ELISA titers at Day 31 (30 days post administration)
• To demonstrate non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine compared to Flu D-QIV given alone based on Geometric mean titer (GMT) of Flu D-QIV antibody titers against 4 influenza strains at Day 31 post administration	Flu D-QIV antibody titers against 4 influenza strains at Day 31 (30 days post administration) Measured by ratio of Haemagglutinin inhibition (HI) GMT between group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and Flu D-QIV vaccine alone group against 4 strains at Day 31 (30 days post administration)
Secondary	
Secondary Safety	
• To evaluate the safety of RSVPreF3 when given alone and co-administered with Flu D-QIV from vaccination up to study end.	The number and percentage of participants reporting SAEs from first vaccination up to study end. [Day 1 to Day 181]
*Confirmatory Immunogenicity***	
• To demonstrate the non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV compared to given alone based on GMT of RSV A neutralizing antibody at Day 31 post administration	RSV A neutralizing antibody titers at Day 31 (30 days post administration) Measured by ratio of GMT between group of RSVPreF3 vaccine co-administered with Flu D-QIV and RSVPreF3 vaccine alone (pooled lot 1,2,3) group in term of RSV A neutralizing antibody titers at Day 31 (30 days administration)

Objectives	Endpoints/Estimands
• To demonstrate the non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 compared to given alone based on Seroconversion rate (SCR**) of Flu D-QIV HI antibody titers against 4 influenza strains at Day 31 post study intervention	• Seroconversion rate to Flu D-QIV HI antibody titers against the 4 influenza strains at Day 31 Measured by the difference of proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
Descriptive Immunogenicity	
• To evaluate the humoral immune response of RSVPreF3 vaccine when given alone and co-administered with Flu D-QIV in terms of RSV A, RSV B neutralizing antibody and RSVPreF3 IgG concentration at Day 1 (prior to the vaccination) and Day 31 (post study intervention).	• RSV A, RSV B neutralizing antibody titers and RSV PreF3 IgG concentration at Day 1 and Day 31 GMT of RSV A and RSV B neutralizing antibody, GMC of RSVPreF3 IgG at Day 1 and Day 31, in the group of RSVPreF3 vaccine co-administered with Flu D-QIV and the group of RSVPreF3 vaccine alone
• To evaluate the humoral immune response to the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine in terms of antibody titers against 4 influenza strains at Day 1 and Day 31	Flu D-QIVHI antibody titers against the 4 influenza strains at Day 1 and Day 31 HI GMT in the 4 influenza strains at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
• To evaluate seroprotection rate (SPR)* and Seroconversion rate (SCR)** of the Flu D-QIV vaccine when given alone and co-administered with RSVPreF3 vaccine	Seroprotection rate to Flu D-QIV HI antibody titers against the 4 influenza strains at Day 1 and Day 31 Measured by the proportion of participants achieving an HI antibody titer ≥1:40 at Day 1 and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone • Seroconversion rate to Flu D-QIV HI antibody titers against the 4 influenza strains at Day 31 Measured by the proportion of participants achieving seroconversion for HI antibody at Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and the group of Flu D-QIV vaccine alone
• To evaluate the humoral immune response in 3 individual lots of RSVPreF3	• RSV A, RSV B neutralizing antibody titers and RSVPreF3 IgG antibody concentrations at pre-vaccination and Day 31 Geometric mean titer/concentration (GMT/GMC) in term of RSV A, RSV B neutralizing antibody and RSVPreF3 IgG antibody at Day 1 (pre-vaccination) and Day 31 in each of 3 lots of the investigational PreF3 vaccine.
Tertiary Objective	
CCI	

*SPR is defined as the percentage of vaccines with a serum HI titer >1:40 that usually is accepted as indicating protection.

** The SCR is defined as the proportion of participants with:

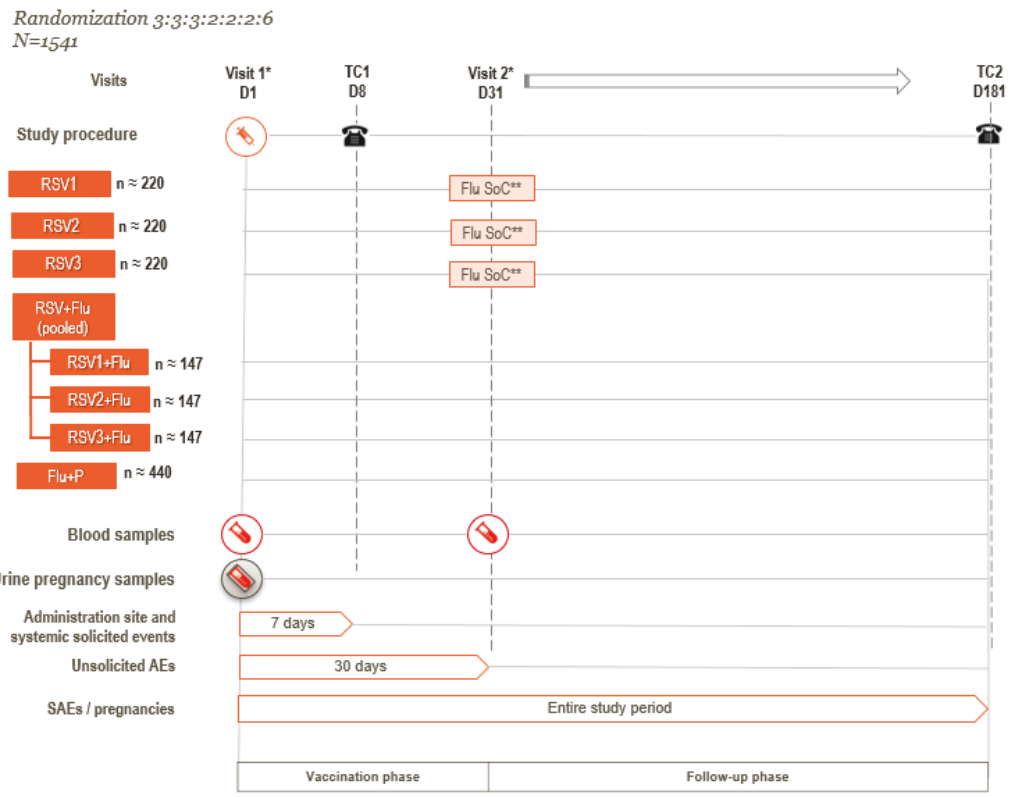
A Day 1 (pre-vaccination) serum anti-HI titer <1:10 and a Day 31 (post-vaccination) serum anti-HI titer ≥1:40, or

A Day 1 (pre-vaccination) serum anti-HI titer ≥ 1:10 and a fold increase (post/pre) ≥ 4 at Day 31

*** Two confirmatory objectives will be tested sequentially.

1.2. Study Design

Overview of Study Design and Key Features



SOC=Standard of Care
* Screening and vaccination will happen on Visit 1(Day 1). The blood sample collection must be collected before administering the vaccine(s).
** Visit 2 Flu D-QIV vaccinations are not part of experimental design. It serves as an optional vaccination for the participant to decide if they would like to get the vaccination as part of their standard of care. The blood sample collection must be collected before administering the Flu D-QIV vaccine.

Design Features	• Study Phase and Population: Phase III, Non-pregnant women • Design type: Randomized, Parallel, multi-country study • Control type: Placebo controlled, Saline (NaCl) solution • Study duration: The study vaccine will be administered at Visit 1 and the participant will be followed for 6 months until the study conclusion. • Blinding: This study will utilize an observer blinding design that is described in Protocol Section 6.3.5
Study intervention	Study (intervention) groups are described in Table 2
Study intervention Assignment	Approximately 1541 participants will be randomized into 7 study groups (5 main study groups including 1 pooled group [RSV1+Flu, RSV2+Flu, RSV3+Flu]) with randomization ratio of

Overview of Study Design and Key Features	
	RSV1:RSV2:RSV3: RSV1+Flu: RSV2+Flu: RSV3+Flu: Flu+P as 3:3:3:2:2:2:6. An automated internet based system (SBIR) will be used to randomly allocate a study group and treatment number to each participant. Age (18-32 vs. 33-49) and center will be minimization factors. Minimization factors will have equal weight in the algorithm.
Interim Analysis	The first analysis will evaluate safety, reactogenicity, and immunogenicity data and will be performed when all participants have completed visits up to (and including) Visit 2 (Day 31) and the data is available.

Table 2 Study groups and intervention

Study groups	Approximate number of participants	Age (Min-Max)	Study intervention	Standard of Care Vaccination*
RSV1	220	18 – 49 years	RSV MAT 120 (Lot1)	Flu D-QIV
RSV2	220	18 – 49 years	RSV MAT 120 (Lot2)	Flu D-QIV
RSV3	220	18 – 49 years	RSV MAT 120 (Lot3)	Flu D-QIV
RSV1+Flu	147	18 – 49 years	RSV MAT 120 (Lot 1) Flu D-QIV	
RSV2+Flu	147	18 – 49 years	RSV MAT 120 (Lot 2) Flu D-QIV	
RSV3+Flu	147	18 – 49 years	RSV MAT 120 (Lot 3) Flu D-QIV	
Flu+P	440	18 – 49 years	Flu D-QIV placebo	

RSV MAT 120 =RSVPreF3 vaccine

*Standard of care vaccine administered only to ensure the participants receive the standard of care.

2. STATISTICAL HYPOTHESES

The study includes 2 confirmatory co-primary objectives:

- To demonstrate the lot-to-lot consistency of 3 lots of RSVPreF3 vaccine in terms of geometric mean concentration (GMC) of RSVPreF3 IgG ELISA at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: at least one of the two-sided 95% confidence intervals for the 3 pair-wise GMC ratios between 2 lots are outside the range of 0.67 to 1.5

  H_a: $0.67 < \mu_{lot1}/\mu_{lot2} < 1.5$ and $0.67 < \mu_{lot1}/\mu_{lot3} < 1.5$ and $0.67 < \mu_{lot2}/\mu_{lot3} < 1.5$

  where μ represents the GMC of RSVPreF3 IgG at Day 31 from each lot. The lot-to-lot consistency will be demonstrated if two-sided 95% confidence intervals for the 3 pair-wise GMC ratios at Day 31 falls within 0.67 and 1.5.

- To demonstrate the non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone in terms of geometric mean titer (GMT) of Flu D-QIV HI antibody against the 4 influenza strains (A/H1N1, A/H3N2, B/Victoria lineage, and B/Yamagata lineage) at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $\mu_{\text{co-ad group}}/\ \mu_{\text{alone}} \leq 0.67$ vs H_a: $\mu_{\text{co-ad group}}/\mu_{\text{alone}} > 0.67$

  where μ represents the GMT of Flu D-QIV HI antibody at Day 31. The non-inferiority will be demonstrated if the lower limits of the two-sided 95% CI on the GMT ratio (co-ad group RSV+Flu with 3 lots pooled divided by Flu+P group) is greater than 0.67 at Day 31 post vaccination for all the 4 strains (A/H1N1, A/H3N2, B/Victoria lineage, and B/Yamagata lineage).

In addition, the study also includes 2 confirmatory secondary objectives. These hypothesis tests will be conducted sequentially only if the primary objective of non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone is met.

- To demonstrate the non-inferiority of RSVPreF3 vaccine when co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of GMT of RSV A neutralizing antibody at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $\mu_{\text{co-ad group}}/\ \mu_{\text{alone}} \leq 0.67$ vs H_a: $\mu_{\text{co-ad group}}/\mu_{\text{alone}} > 0.67$

  where μ represents the GMT of RSV A neutralizing antibody at Day 31. The non-inferiority will be demonstrated if the lower limits of the two-sided 95% CI on the GMT ratio (co-ad group RSV+Flu divided by RSV alone group with 3 lots pooled in both groups) is greater than 0.67 at Day 31 post vaccination.

- To demonstrate the non-inferiority for each strain (A/H1N1, A/H3N2, B/Victoria lineage, and B/Yamagata lineage) of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone in terms of seroconversion rate (SCR) at Day 31.

  Null hypothesis vs. Alternative hypothesis:

  H_0: $p_{\text{alone}} - p_{\text{co-ad group}} \geq 10\%$ vs H_a: $p_{\text{alone}} - p_{\text{co-ad group}} < 10\%$

  where p represents the SCR for each influenza strain at Day 31. The non-inferiority for each strain will be demonstrated if the upper limits of the two-sided 95% CI on the SCR difference (Flu+P group minus co-ad group RSV+Flu with 3 lots pooled) is less than 10% at Day 31 post vaccination.

2.1. Multiplicity Adjustment

To ensure the global type I error for the primary objectives is controlled at 5%, each of the primary objectives will be tested using a nominal one-sided alpha of 2.5%.

If the primary objective of non-inferiority of Flu D-QIV in terms of GMT is met, the secondary confirmatory objective of non-inferiority of RSVPreF3 vaccine in terms of

GMC will be tested at alpha of 2.5%, if this objective is met, the non-inferiority of Flu D-QIV in terms of SCR will be tested at alpha of 2.5% for each strain, otherwise, it will not be tested.

3. ANALYSIS SETS

Table 3 Analysis sets

Analysis Set	Definition / Criteria	Analyses Evaluated
Enrolled	Participants who agreed to participate in a clinical study after completion of the informed consent process	Study Population
Exposed	All Participants who received at least 1 dose of the study treatment. The allocation in a group is done as a function of the administered treatment	Safety, Study Population
Full Analysis Set (FAS)	All Participants who received at least 1 dose of the study intervention and have post-vaccination immunogenicity data It will be defined by timepoint, e,g. FAS at Day 31	Immunogenicity
Per-Protocol Set (PPS)	All Participants who received at least 1 dose of the study treatment to which they are randomized and have post-vaccination data minus participants with protocol deviations that lead to exclusion It will be defined by timepoint, e,g. PPS at Day 31	Immunogenicity
Solicited Safety Set	All Participants who received at least 1 dose of the study intervention (Exposed Set) who have solicited safety data	Solicited events
Randomized Set	Randomized set will include all participants who are randomized. The allocation in a group is done as function of the randomized intervention. Please note this set was not included in the protocol, but will be used later in one summary analysis, so it is added here for clarification.	Disposition

3.1. Criteria for eliminating data from Analysis Sets

Elimination codes are used to identify participants to be eliminated from analysis. Detail is provided below for each set.

3.1.1. Elimination from Exposed Set (ES)

Code 1030 (Study vaccine not administered at all), 800 (Fraudulent data) and code 900 (invalid informed consent) will be used for identifying participants eliminated from ES

3.1.2. Elimination from Full Analysis Set (FAS)

A participant will be excluded from the FAS under the following conditions

Table 4 Elimination code and condition

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
800	Fraudulent data	All	All
900	Invalid informed consent	All	All
1030	Study vaccine not administered at all	All	Safety, immunogenicity
2100.Vx	Serological results not available post-vaccination	Visit 2/Day 31	Immunogenicity

Vx indicates participants whose immunogenicity data will be eliminated from a specific visit.

3.1.3. Elimination from Per-protocol Set (PPS)

A participant will be excluded from the PPS under the following conditions

Table 5 Elimination code and condition

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
800	Fraudulent data	All	All
900	Invalid informed consent	All	All
1030	Study vaccine not administered at all	All	Safety, immunogenicity

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
1040.Vx*	Administration of concomitant vaccine(s) forbidden in the protocol	Visit 2/Day 31	Immunogenicity
1050	Randomisation failure	All	Immunogenicity
1060	Randomisation code was broken	All	Immunogenicity
1070**	Subjects got vaccinated with the correct vaccine but containing an incorrect volume	All	Immunogenicity
1070**	Vaccination not according to protocol (site of injection, route of administration, wrong replacement of study treatment administered)	All	Immunogenicity
1070**	Study treatment not prepared as per protocol (e.g. reconstitution)	All	Immunogenicity
1070**	Other deviations related to wrong study treatment/administration/dose	All	Immunogenicity
1070**	Study treatment administered while contraindication	All	Immunogenicity
1080	Vaccine temperature deviation	All	Immunogenicity
1090	Expired vaccine administered	All	Immunogenicity
2010	Protocol violation (inclusion/exclusion criteria) DOB – VAC – 18-49 years BMI <=40 kg/m^2	All	Immunogenicity
2040.Vx*	Administration of any medication forbidden by the protocol	Visit 2/Day 31	Immunogenicity
2050.Vx*	Intercurrent medical conditions which are exclusionary as per protocol	Visit 2/Day 31	Immunogenicity
2090.Vx	Subjects did not comply with blood sample schedule: • For PPS at Day 31, check the interval from vaccination to day 31 BS = 31 – 38 days;	Visit 2/Day 31	Immunogenicity
2100.Vx	Serological results not available post-vaccination	Visit 2/Day 31	Immunogenicity

Code	Condition under which the code is used	Visit (timepoints) where the code is applicable	Applicable for analysis set/endpoint
2120.Vx	Obvious incoherence or abnormality or error in data	All	Immunogenicity
2130.Vx	Testing performed on samples not aligned with ICF	All	Immunogenicity

*Attribution of these elimination codes to subject need CRDL review of individual listing
** Attribution of code 1070 to a subject requires CRDL confirmation
Vx+ indicates participants whose immunogenicity data will be eliminated from a specific visit onwards; Vx indicates participants whose immunogenicity data will be eliminated from a specific visit.
DOB-Date of Birth, VAC-Vaccination, BS- Blood Sample, BMI-Body Mass Index.

3.1.4. Elimination from solicited safety set

Code 1030 (Study vaccine not administered at all), code 800 (fraudulent data) and code 900 (invalid informed consent) and code 1160 (no post-vaccination solicited safety data) will be used for identifying participants eliminated from the solicited safety set.

4. STATISTICAL ANALYSES

4.1. General Considerations

Unless otherwise specified, reactogenicity analysis and safety analysis will be performed on solicited safety set and Exposed Set respectively.

In general, Immunogenicity analysis will be performed on the Per Protocol set. If, in any study group and at any timepoint, the percentage of vaccinated participants with immunogenicity results excluded from the Per Protocol set is 5% or more, a second analysis based on the Full Analysis Set will be performed to complement the Per Protocol analysis for some immunogenicity analysis.

4.1.1. General Methodology

Unless otherwise specified, continuous data will be summarized using descriptive statistics: n, mean, standard deviation (std), median, minimum and maximum. Categorical data will be summarized as the number and percentage of participants in each category.

95% CI for proportion will be based on exact Clopper-Pearson confidence interval (Clopper, 1934).

95% CI for group difference in proportion will be based on Miettinen and Nurminen confidence interval (Miettinen, 1985).

95% CI for GMT(C) will be based on a back transformation of student confidence interval for the mean of log-transformation.

95% CI for GMT(C) ratio between groups will be based on a back transformation of confidence interval for the mean difference on log-transformation.

For a given participant and given immunogenicity measurement, missing or non-evaluable measurements will neither be imputed nor be replaced, and therefore will not be included in immunogenicity analysis.

For between group statistical modelling analysis, participants having a result at both the baseline and the considered timepoint will be included in the analysis.

Participants who prematurely withdrew from study will not be replaced.

4.1.2. Definition

Solicited events and their intensity scales are defined as below:

Table 6 Intensity scales for solicited events

CCI - This section contained Clinical Outcome Assessment data collection questionnaires or indices, which are protected by third party copyright laws and therefore have been excluded.

Baseline: For all endpoints the baseline value will be the latest pre-dose assessment with a non-missing value, including those from unscheduled visits. If time is not collected, Day 1 assessments are assumed to be taken prior to vaccination and used as baseline. Unless otherwise stated, if baseline data is missing no derivation will be performed and baseline will be set to missing.

For the RSV and FLU antigens:

- A seronegative participant will be defined as a participant whose antibody titer/concentration is below the cut-off value of the assay.
- A seropositive participant is a participant whose antibody titer/concentration is greater than or equal to the cut-off value of the assay.
- MGI (mean geometric increase): The geometric mean of the within participant ratios of the post-dose titer over the pre-dose titer.

Please refer to Section 6.2.4.6 for cut-off value for each assay.

For the FLU antigens:

- SCR (seroconversion rate): The percentage of participants who have either a HI pre-dose titer < 1:10 and a post-dose titer ≥ 1:40 or a pre-dose titer ≥1:10 and at least a four-fold increase in post-dose titer.
- SPR (seroprotection rate): The percentage of participants with a serum HI titer ≥1:40 that usually is accepted as indicating protection.

4.2. Primary Endpoint(s) Analyses

4.2.1. Safety analysis

The analysis of solicited administration site event (associated with RSV or Flu) and solicited systemic event will be performed on solicited safety set.

- The number and percentage with exact 95% CI of participants reporting each solicited administration site event (any grade, each grade) and solicited systemic event (any, each grade) during the 7-day follow-up period after dosing will be tabulated by maximum intensity per participant for each study group.
- For fever during the 7-day follow-up period after vaccination, the number and percentage of participants reporting any fever (i.e., temperature ≥38 °C) and fever by half degree (°C) cumulative increments, any Grade 3 fevers, will be reported.
- Duration in days of solicited administration site and systemic events within 7 days after vaccination will be tabulated by study group and overall. The derivation rule of duration in days for solicited events is detailed in section 6.2.4.9.

The analysis of unsolicited AEs and SAEs will be performed on ES.

- The number and percentage of participants reporting unsolicited AEs within 30 days after dosing with exact 95% CIs will be tabulated by group and by Medical Dictionary for Regulatory Activities (MedDRA) terms.
- Similar tabulations will be done for Grade 3 unsolicited AEs, for any causally related unsolicited AEs, and for Grade 3 related unsolicited AEs.
- The number and percentage of participants reporting at least one SAE within 30 days after dosing with exact 95% CIs will be tabulated by group and by MedDRA terms.
- By-participant listings of SAEs and AEs leading to study withdrawal will be prepared.

4.2.2. Immunogenicity analysis

- Lot-to-lot consistency analysis

  The GMC ratio and its two-sided 95% CI between 2 lots in terms of RSVPreF3 IgG ELISA concentration at Day 31 will be computed by back transformation of its mean difference and 95% CI on log10 transformed scale estimated from the t-distribution.

  Summary table will show the number of observations included in the calculation for each lot, GMC of each lot, and 3 pair-wise GMC ratios with corresponding 95% CIs.

  Lot-to-lot consistency will be demonstrated if all 3 two-sided 95% CIs for GMC ratio fall within 0.67 and 1.5.

- Non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone with respect to 4 Flu D-QIV strains at Day 31

  The GMT ratio and its two-sided 95% CIs between the 2 groups (RSV+Flu with 3 lots pooled divided by Flu+P group) for A/H1N1 strain, A/H3N2 strain, B/Victoria lineage and B/Yamagata lineage will be computed by back transformation of its mean difference and 95% CI on log10 transformed scale estimated from the t-distribution.

  Summary table will show for each group the number of observations included in the calculation and GMT value, and between group GMT ratio with corresponding 95% CI.

  Non-inferiority of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine will be demonstrated if lower bounds of all 4 two-sided 95% CIs are above 0.67.

4.2.3. Sensitivity analyses

For primary immunogenicity endpoints, between study group comparisons will also be explored through statistical modelling.

- Lot-to-lot consistency analysis

  The ANCOVA model on the log10 transformation of the concentrations will be explored, and will include the lot as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination concentrations as covariates. Covariates in the model may be further adjusted when performing the analysis if appropriate and needed, therefore, the following SAS code can serve as a reference and may be adjusted according to the analysis needs.

```
PROC GLM data=sero;
  CLASS group age_cat;
     MODEL log_val = baseline group age_cat group*age_cat;
      LSMEANS group/pdiff cl alpha=0.05;
     RUN;
```

  where log_val represents the log-transformed RSVPreF3 IgG antibody concentration at Day 31, baseline is pre-vaccination RSVPreF3 IgG antibody level on log-transformed scale, group includes each lot, age_cat is the age category at vaccination (18-32 vs. 33-49).

  The 3 pair-wise GMC ratios between 2 lots and their two-sided 95% CI will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

  Summary table will show the number of observations included in the model for each lot, adjusted GMC of each lot, and 3 pair-wise adjusted GMC ratios with corresponding 95% CIs.

- Non-inferiority of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone with respect to 4 Flu D-QIV strains at Day 31

  The ANCOVA model on the log10 transformation of the titers will be explored, and will include the study group (RSV+Flu with 3 lots pooled, Flu+P) as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination titers as covariates, covariates in the model may be further adjusted when performing the analysis if appropriate and needed. Similar SAS code could be referenced and may be further adjusted at time of analysis:

```
PROC GLM data=sero;
  CLASS group age_cat;
     MODEL log_val = baseline group age_cat group*age_cat;
      LSMEANS group/pdiff cl alpha=0.05;
     RUN;
```

  where log_val represents the log-transformed antibody level at Day 31, baseline is pre-vaccination antibody level on log-transformed scale, group includes RSV+Flu

with 3 lots pooled and Flu+P, age_cat is the age category at vaccination (18-32 vs. 33-49).

The GMT ratio and its two-sided 95% CIs between the 2 groups (RSV+Flu with 3 lots pooled divided by Flu+P group) for A/H1N1, A/H3N2, B/Victoria lineage and B/Yamagata lineage will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

Summary tables will show for each group the number of observations included in the model and model adjusted GMT, and between group adjusted GMT ratios with the corresponding 95% CIs.

4.3. Secondary Endpoint(s) Analyses

4.3.1. Safety analysis

The analysis of SAEs from the first vaccination up to study end will be performed on ES.

- The number and percentage of participants reporting at least one SAE from vaccination up to study end with exact 95% CIs will be tabulated by group and by MedDRA terms
- By participant listings of SAEs and (S)AEs leading to study withdrawal will be prepared.

4.3.2. Immunogenicity analysis

4.3.2.1. Secondary confirmatory analysis

In order to control the global type I error, the objectives will be assessed sequentially.

- Non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of RSV A neutralizing antibody titers at Day 31

  The GMT ratio and its two-sided 95% CI between 2 groups (RSV+Flu group with 3 lots pooled divided by RSV alone group with 3 lots pooled) will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from t-distribution.

  Summary table will show for each group the number of observations included in the calculation and GMT value, and between group GMT ratio with corresponding 95% CI.

  No interference on RSVPreF3 vaccine from quadrivalent seasonal influenza vaccine (Flu D-QIV) will be demonstrated if lower bound of the two-sided 95% CI of GMT ratio is above 0.67.

- Non-inferiority for each of the 4 strains of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine as compared to Flu D-QIV vaccine administered alone at Day 31 in terms of Seroconversion rate (SCR)

  The SCR difference and its two-sided 95% CI between 2 groups (Flu+P group minus RSV+Flu group with 3 lots pooled) will be calculated for each strain (A/H1N1, A/H3N2, B/Victoria lineage and B/Yamagata lineage) at Day 31. 95% CI calculation will be based on Miettinen and Nurminen confidence interval (Miettinen, 1985).

  Summary table will show for each group the number of observations included in the calculation, number and percentage of seroconverted participants, and between group SCR difference with corresponding 95% CI.

  For each strain, non-inferiority will be demonstrated if the upper bound of the two-sided 95% CI is less than 10% at Day 31 post vaccination.

4.3.2.2. Descriptive immunogenicity analysis

For RSV A, RSV B neutralizing antibody titers and RSVPreF3 IgG antibody concentrations at pre-vaccination and Day 31 from each lot (RSV1, RSV2, and RSV3), as well as for the group of RSV alone with 3 lots pooled (RSV1, RSV2 and RSV3 pooled) and RSV+Flu group with 3 lots pooled, the following analysis will be performed:

- Number and percentage of participants with antibody titers/concentrations above assay cut-off will be tabulated with its exact 95% CI by group.
- GMT/GMCs will be tabulated with 95% CI at each timepoint and represented graphically by group.
- MGI from Day 31 over pre-vaccination will be tabulated with 95% CI by group.

For Flu D-QIV HI antibody titers against 4 influenza strains at pre-vaccination and Day 31 in the group of Flu D-QIV vaccine co-administered with RSVPreF3 vaccine with 3 lots pooled (RSV+Flu) and the group of Flu D-QIV vaccine administered alone (Flu+P), the following analysis will be performed:

- Number and percentage of participants with antibody titers above assay cut-off and their exact 95% CI will be tabulated by group.
- GMTs will be tabulated with 95% CI at each timepoint and represented graphically by group.
- MGI from Day 31 over pre-vaccination will be tabulated with 95% CI by group.
- Number and percentage of participants achieving seroconversion for HI antibody at Day 31 will be tabulated with its exact 95% CI by group.
- Number and percentage of participants achieving HI antibody titer $\geq$ 1:40 at pre-vaccination and Day 31 will be tabulated with its exact 95% CI by group.

4.3.3. Sensitivity analyses

ANCOVA modelling analysis will be explored for non-inferiority of RSVPreF3 vaccine co-administered with Flu D-QIV vaccine as compared to RSVPreF3 vaccine administered alone in terms of RSV A neutralizing antibody titers at Day 31.

The ANCOVA model will be based on the log10 transformation of the titers, and will include the study group (RSV+Flu with 3 lots pooled, RSV alone group with 3 lots pooled) as fixed effect, age category at the time of vaccination (18-32 vs. 33-49) and pre-vaccination titers as covariates, covariates in the model may be further adjusted when performing the analysis if appropriate and needed. Similar SAS code will be explored and may be further adjusted at time of analysis:

```
PROC GLM data=sero;
  CLASS group age_cat;
  MODEL log_val = baseline group age_cat group*age_cat;
  LSMEANS group/pdiff cl alpha=0.05;
RUN;
```

where log_val represents the log-transformed RSV A neutralizing antibody titer at Day 31, baseline is pre-vaccination RSV A neutralizing antibody titer on log-transformed scale, group includes Flu+RSV with 3 lots pooled and RSV alone with 3 lots pooled, age_cat is the age category at vaccination (18-32 vs. 33-49).

The GMT ratio and its two-sided 95% CIs between the 2 groups (RSVPreF3+Flu D-QIV with 3 lots pooled divided by RSVPreF3 alone group with 3 lots pooled) will be computed by exponentiating the mean difference and its 95% CI on log10 transformed scale estimated from ANCOVA model.

Summary tables will show for each group the number of observations included in the model and model adjusted GMT, and between group adjusted GMT ratios with the corresponding 95% CIs.

4.4. Tertiary Endpoint(s) Analyses

Not applicable

4.5. Other Safety Analyses

Other safety analyses will be based on the Exposed Set, unless otherwise specified.

4.5.1. Combined solicited and unsolicited events

For clintrial.gov and EudraCT posting purposes, a summary of combined solicited and unsolicited adverse events will be produced by System Organ Class and preferred terms and according to occurrence of each event.

Solicited administration site events and solicited systemic events will be coded by MedDRA as per the following codes

Solicited symptom	Lower level term code	Corresponding Lower level term decode
Pain	Injection site pain	10022086
Redness	Injection site redness	10022098
Swelling	Injection site swelling	10053425
Fatigue	Fatigue	10016256
Fever	Fever	10016558
Nausea	Nausea	10028813
Vomiting	Vomiting	10047700
Diarrhea	Diarrhea	10012727
Abdominal pain	Abdominal pain	10000081
Headache	Headache	10019211

Please note: the coding will be double checked during the analysis

4.5.2. COVID-19 Assessment and COVID-19 AEs

A participant is defined as having a suspected, probable or confirmed COVID-19 infection during the study if the answer is “Confirmed”, “Probable” or “Suspected” to the case diagnosis question from the COVID-19 coronavirus infection assessment eCRF.

Numbers and percentage of participants with a suspected, probable or confirmed COVID-19 infection will be summarized by group based on ES.

Number and percentage of participants who had a COVID-19 test performed and number and percentage of participants with positive, negative and indeterminate results will be summarized by group on ES.

4.5.3. Additional Safety Assessments (if applicable)

Vital signs will be summarized by group using descriptive statistics at all timepoint(s) the information is collected on ES and PPS. The parameters include but may not be limited to systolic blood pressure, diastolic blood pressure, temperature, heart rate, respiratory rate, height, weight and body mass index.

4.6. Other Analyses

Not applicable.**Interim Analyses**

The first analysis will evaluate safety, reactogenicity, and immunogenicity data and will be performed when all participants have completed visits up to (and including) Visit 2 (Day 31) and the data is available. At this point, the statistician will be unblinded (i.e. will have access to the individual participant treatment assignments), but no individual listings will be provided to investigators until the end of study and an end of study report containing all available data is developed.

The final, end of study analysis will be performed when all data for at least primary and secondary endpoints up to study conclusion are available. Individual listings will only be

provided at this stage. An integrated clinical study report (CSR) containing all available data will be written and made available to the investigators.

4.8. Changes to Protocol Defined Analyses

There were no changes or deviations to the originally planned statistical analysis specified in the protocol (Dated: [23-APR-2021]).

5. SAMPLE SIZE DETERMINATION

Approximately 1541 participants will be randomized to achieve appropriately 1400 evaluable participants.

Assessments of both immunogenicity and safety data were considered when determining sample size for this study.

Participants who withdraw from the study will not be replaced.

The sections below describe the assumptions, adjustment and methodology used for the sample size calculation.

5.1. Primary objective for Lot-to-Lot consistency

The primary objective of lot-to-lot consistency will be evaluated on the RSVPreF3 immunogenicity as measured by RSVPreF3 IgG ELISA concentration at Day 31.

The lot-to-lot consistency will be demonstrated only if two-sided 95% confidence intervals for the 3 pair-wise geometric mean ratios of RSVPreF3 IgG ELISA concentration at Day 31 (30 days post vaccination) falls within 0.67 and 1.5.

With the assumptions of a SD of log10 transformed RSVPreF3 IgG ELISA concentration of 0.35, true GMC ratio of 1 between 2 lots, type I error of 0.025, and 10% of non-evaluable rate, 220 enrolled participants per lot provides at least 99% global power to conclude lot-to-lot consistency (see details in Table 7).

Table 7 Power to demonstrate Lot-to-Lot consistency on immune responses of RSVPreF3 IgG ELISA concentration at Day 31 post vaccination

Endpoint	Clinically acceptable bounds for consistency	Group description	Number of evaluable participants in each lot	Reference SD**	Power
*RSVPreF3 IgG GMC ratio	(0.67, 1.5)	Lot 1/2	200	0.35	99.8%
*RSVPreF3 IgG GMC ratio	(0.67, 1.5]	Lot 1/3	200	0.35	99.8%
*RSVPreF3 IgG GMC ratio	(0.67, 1.5]	Lot 2/3	200	0.35	99.8%
Global power					99.3%

* Pass 2019, Two-Sample T-test for equivalence allowing unequal variance, alpha=2.5%;
**References used for the sample size calculation: Study *RSV MAT 011* (209141)

5.2. Primary objective: To demonstrate Non-inferiority of immune response of Flu D-QIV vaccine when co-administered with RSVPreF3 vaccine or administered alone at Day 31 post dose vaccination

Possible interference of RSVPreF3 on influenza immune responses will be evaluated by using Flu D-QIV HI antibody titers against the 4 influenza strains from RSV+Flu (pooled of RSV1+Flu, RSV2+Flu, RSV3+Flu) and Flu+P group at Day 31. In the description of the subsequent analysis, the RSV+Flu vaccine group are RSV1+Flu, RSV2+Flu, RSV3+Flu groups pooled.

The hypothesis is that co-administration of RSVPreF3+ Flu D-QIV vaccine is non-inferior to Flu D-QIV vaccine with respect to GMT ratio for immune response of influenza antigen in non-pregnant women at Day 31 post vaccination.

The criteria to evaluate non-inferiority for A/H1N1, A/H3N2, B/Victoria lineage, and B/Yamagata lineage is that the lower limits of the 95% CI on the GMT ratio (RSV+Flu divided by Flu+P) is greater than 0.67 at Day 31 post vaccination.

Table 8 presents the power on the ratio of the GMT of Flu D-QIV 4 strains. The power is at least 94% when the sample size is 400 participants per group with standard deviation (SD) of 0.6 on its log10 transformation and non-inferiority margin of 0.67 between RSVPreF3 co-administrated with Flu D-QIV group and Flu D-QIV vaccine administered alone group.

Table 8 Power to demonstrate non-inferiority of immune response between Flu D-QIV vaccine co-administered with RSVPreF3 or administered alone in term of GMT for Flu D-QIV strains at Day 31 post 1st vaccination with assumed GMT ratio 1

Endpoint	NI criteria	Number of evaluable participants in each group	Reference*	Power
A/H1N1 GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
B/Victoria lineage GMT ratio	LL of 95% CI for GMC ratio >0.67	400	0.60	98.56%
A/H3N2 GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
B/Yamagata lineage GMT ratio	LL of 95% CI for GMT ratio >0.67	400	0.60	98.56%
Global power				94.24%

By Pass 2019, Non-Inferiority Tests for Two Means using Differences, one-sided alpha=2.5%;
*References used for the sample size calculation: Study Flu D-QIV -008 E1_01 Table 34

5.3. Secondary objectives

In order to control the global type I error, the objectives will be assessed sequentially.

5.3.1. To demonstrate the non-inferiority of immune response between RSVPreF3 co-administered with Flu D-QIV and RSVPreF3 alone in term of RSV A Neutralizing antibody titers at Day 31 post administration

Hypothesis under consideration is that immune response when the co-administration of RSVPreF3 vaccine and Flu D-QIV vaccine is non-inferior to that when RSVPreF3 vaccine (pooled RSV1, RSV2 and RSV3 group) is administered alone with respect to GMT ratio for RSV A neutralizing antibody titer in non-pregnant women at Day 31 post vaccination.

This hypothesis test is conducted only if the primary objective of non-inferiority of immune response between Flu D-QIV vaccine when co-administered with RSVPreF3 and Flu D-QIV vaccine administered alone is met.

The criteria to evaluate non-inferiority for RSV A Nab is that the lower limits of the 95% CI on the GMT ratio (RSV+Flu group divided by RSV alone group with 3 lots pooled in both groups) is greater than the pre-defined clinical limit of 0.67 at Day 31 post vaccination.

If a SD of log10 transformed RSV A Nab of 0.4 is assumed, with an assumption of GMT ratio of 1.0, there will be at least 99% chance that the lower bound of 95% CI for the ratio of GMT of RSV A Nab titer between RSVPreF3 co-administration with Flu D-QIV and administered alone is above 0.67 (Table 9)

Table 9 Power to demonstrate non-inferiority of immune response of RSVPreF3 vaccine when co-administered with Flu D-QIV vaccine or administered alone at Day 31 post vaccination

Endpoint	NI criteria	N1:N2 (evaluable)	Reference*	Power
RSV A GMT ratio	LL of 95% CI for GMT ratio >0.67	400:600	0.40	99.99%

By Pass 2019 Non-Inferiority Tests for Two Means using Differences, one-sided alpha=2.5%;
*References used for the sample size calculation: Study RSV MAT 011 (209141)
N1: Number of evaluable participants in the co-administration of RSVPreF3 vaccine + Flu D-QIV vaccine.
N2: Number of evaluable participants in the RSVPreF3 vaccine alone group with 3 lots pooled.

5.3.2. To demonstrate the non-inferiority of immune response between RSVPreF3 co-administered with Flu D-QIV and Flu D-QIV alone in term of SCR at Day 31 post 1st dose administration

Potential interference of RSVPreF3 on influenza immune responses will be evaluated by comparing the Day 31 seroconversion rate of Flu D-QIV HI antibody titers against the 4 influenza strains between RSV+Flu group and Flu+P group.

The criteria to evaluate non-inferiority with respect to the SCR difference for Flu D-QIV antibody titers against 4 influenza strains is that the upper limits of the 95% CI on the SCR difference (Flu+P group minus RSV+Flu) is less than the pre-defined clinical limit of 10% at Day 31 post vaccination.

With the assumptions of SCR for the reference group as indicated in Table 10, significance level 0.025, non-inferiority margin 10%, 400 evaluable participants (10% non-evaluable rate) provides at least 80% power to conclude non-inferiority for each strain (Table 10).

Table 10 Powers to demonstrate non-inferiority in terms of HI antibody SCR for Flu D-QIV strains between Flu D-QIV vaccine co-administered with RSVPreF3 vaccine and administered alone at Day 31 post vaccination

Endpoint	NI criteria	Anticipated SCR*	Number of participants per group (evaluable)	Power
A/H1N1 SCR	10%	60%	400	82.49%
B/Victoria lineage SCR	10%	65%	400	84.39%
A/H3N2 SCR	10%	65%	400	84.39%
B/Yamagata lineage SCR	10%	65%	400	84.39%

By Pass 2019, Non-Inferiority Tests for the difference between two proportions, alpha=2.5%
*SCR used in the reference group for power calculation is from Study Tdap-0.3-008, Suppl. Table 36.

6. SUPPORTING DOCUMENTATION

6.1. Appendix 1 Study Population Analyses

6.1.1. Participant Disposition

Summary of participant disposition will be performed on ES. The number and percentage of participants who completed the study and who prematurely withdrew from the study including the reasons for study withdrawal will be summarized by each group and overall.

6.1.2. Demographic and Baseline Characteristics

The analysis of demography will be performed on Enrolled Set, ES and PPS using descriptive statistics. The parameters including but may not be limited to age, age category (18-32 vs 33-49), ethnicity, race, country and childbearing potential will be summarized by group.

Summary of past medical history and current medical conditions will be performed on ES by Medical Dictionary for Regulatory Activities (MedDRA) term. Un-coded medical conditions or medical history will be summarized under 'Other' category.

Vaccination history will be coded using GSK Drug dictionaries. Summary of vaccination history will be performed on ES by group.

6.1.3. Protocol Deviations

Important protocol deviations will be summarized based on ES by group.

Protocol deviations will be tracked by the study team throughout the conduct of the study. These protocol deviations will be reviewed to identify those considered as important as follows:

- Data will be reviewed prior to unblinding and freezing the database to ensure all important deviations (where possible without knowing the study intervention details) are captured and categorised in the protocol deviations dataset.
- This dataset will be the basis for the summaries of important protocol deviations.
- The summary will include number and percentage of participants with important protocol deviations by deviation category for each study group.
- An individual listing of protocol deviation will also be provided.

Protocol deviations which result in exclusion from the analysis set will be summarized on ES by group.

- Data will be reviewed prior to unblinding and freezing the database to ensure all deviations leading to analysis population exclusions are captured and categorised in the protocol deviations ADaM dataset (note these exclusions are not captured in the SDTM dataset).
- The summary will include number and percentage of participants with protocol deviations leading to exclusion by deviation category for each study group.

In addition to the above summary, separate summaries may be produced for important protocol deviations related to COVID-19, and important protocol deviations not related to COVID-19 respectively.

6.1.4. Concomitant Medications and Vaccinations

Concomitant medications and vaccinations will be coded and summarized using GSK Drug dictionary.

- The number and percentage of participants taking concomitant medications (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) will be summarized by group. A listing will also be provided.
- The number and percentage of participants taking concomitant vaccinations will be summarized by group. A listing will also be provided.

6.1.5. Additional Analyses Due to the COVID-19 Pandemic

Depending on how the Covid-19 situation evolves, the SAP might be amended to reflect the analysis corresponding to Covid-19.

6.2. Appendix 2 Data Derivations Rule

6.2.1. Study Day and Reference Dates

The safety reference date is the study intervention start date and will be used to calculate study day for safety measures.

The study day is calculated as below:

- Assessment Date = Missing → Study Day = Missing
- Assessment Date < Reference Date → Study Day = Assessment Date – Ref Date
- Assessment Data ≥ Reference Date → Study Day = Assessment Date – Ref Date + 1

6.2.2. Attributing events to vaccine doses

The dose relative to an event is the most recent study dose given to a participant prior to the start of a given event. For example, if the start date of an adverse event is between Dose 1 and Dose 2, the relative dose will be Dose 1.

If an event starts on the same day as a study dose, the relative dose will be derived from the additional information provided in the case report form (CRF) using the contents of the flag indicating if the event occurred before or after study dose. If 'after study dose' is selected, the relative dose for the event will be the one administered on the start day of the event. If 'before study dose' is selected, the relative dose for the event will be the dose prior to this one.

6.2.3. Handling of missing data

6.2.3.1. Dates

When partially completed dates (i.e. dates missing a day and/or month) are used in calculations, the following standard rules will be applied:

- A missing day will be replaced by 15
- A missing day and month will be replaced by June 30th.

The following exceptions apply:

- Adverse event start dates with missing day:
  - If the event starts in the same month as at least one of the study doses, the contents of AE.AESTRTPT (the flag indicating if the event occurred before or after study dose) will be used to complete the date. If 'after study dose' is selected, the imputed start date will match the first (or only) study dose given during that month. If 'before study dose' is selected, the imputed date will be one day before the first (or only) study dose given during that month.
- Adverse event start dates with missing day and month:
  - If the event starts in the same year as at least one of the study doses, the contents of AE.AESTRTPT (the flag indicating if the event occurred before or after study dose) will be used to complete the date. If 'after study dose' is selected, the imputed start date will match the first (or only) study dose given during that year. If 'before study dose' is selected, the imputed date will be one day before the first (or only) study dose given during that year.

All other cases of incomplete AE or concomitant medication/vaccination start date will follow the standard rules above.

6.2.3.2. Daily recording of solicited events

6.2.3.2.1. Studies with electronic diaries

For studies using electronic diaries for the collection of solicited events, a solicited event will be considered present only when a daily recording of grade 1 or more is present.

6.2.3.3. Unsolicited adverse events

Unsolicited adverse event summaries are including serious adverse events unless specified otherwise.

Missing severity, relationship with study vaccine, and outcome of unsolicited adverse events will not be replaced and will appear as 'UNKNOWN' when displayed in a statistical output.

6.2.4. Data derivation

6.2.4.1. Age at first dose in years

When age at first dose is to be displayed in years, it will be calculated as the number of complete calendar years between the date of birth and the date of first dose. For example:

DOB = 10SEP1983, Date of first dose = 09SEP2018 -> Age = 34 years

DOB = 10SEP1983, Date of first dose = 10SEP2018 -> Age = 35 years

6.2.4.2. Weight

Weight will be presented in kilograms. Weights reported in pounds will be converted as follows:

Weight in kilograms = Weight in pounds / 2.2

6.2.4.3. Height

Height will be presented in centimeters. Heights reported in feet and inches will be converted as follows:

Height in centimeters = Height in inches x 2.54

6.2.4.4. Body mass index (BMI)

BMI will be calculated as follows:

BMI = (Weight in kilograms) / (Height in meters)^2

6.2.4.5. Temperature

Temperatures will be presented in degrees Celsius (°C). Temperatures reported in degrees Fahrenheit (°F) will be converted as follows:

Temperature (Celsius) = ((Temperature (Fahrenheit) - 32) x 5)/9

6.2.4.6. Numerical serology results

Numerical serology results will be derived from the content of IS.ISORRES in the SDTM dataset. For assays with a specific cut-off, the following derivation rules apply:

IS.ISORRES	Derived value
"NEG", "-", or "(-)"	cut-off/2
"POS", "+", or "(+)"	cut-off
"< value" and value is <= assay cut-off	cut-off/2
"< value" and value is > assay cut-off	value
"> value" and value is < assay cut-off	cut-off/2
"> value" and value is >= assay cut-off	value
"value" and value is < cut-off	cut-off/2
"value" and value is >= cut-off	value
All other cases	missing

The cut-off tests for immunogenicity evaluation will be as per following:

System	Component	Method	Unit	Cut-off *(LLOQ)*	*ULOQ*
Serum	RSV-A Neutralising Antibody	NEUTRALISATION	ED60	18	*123535*
Serum	RSV-A Neutralising Antibody	NEUTRALISATION	IU/mL	56	*217400*
Serum	RSVPreF3 IgG antibody concentrations	ELISA	EU/mL	*25*	*251769*
Serum	RSV-B Neutralising Antibody	NEUTRALISATION	ED60	*30*	*138336*
Serum	RSV-B Neutralising Antibody	NEUTRALISATION	IU/mL	*44*	*171279*
Serum	A/Victoria/2570/2019 (H1N1)	HI assay	*TBD*	*TBD*	*TBD*
Serum	A/Tasmania/503/2020 (H3N2)	HI assay	*TBD*	*TBD*	*TBD*
Serum	B/Washington/02/2019	HI assay	*TBD*	*TBD*	*TBD*
Serum	B/Phuket/3073/2013	HI assay	*TBD*	*TBD*	*TBD*

Note: the assay cut-off (LLOQ), ULOQ and units may be further adjusted at time of analysis when notified by the lab.

6.2.4.7. Geometric mean titres (GMTs) and concentrations (GMCs)

Geometric Mean Titre (GMT) or Concentration (GMC) calculations are performed by taking the inverse logarithm of the mean of the log titre or concentration transformations. Non quantifiable antibody titres or concentrations will be converted as described in section 6.2.4.6 for the purpose of GMT/GMC calculation. The cut-off value is defined by the laboratory before the analysis.

6.2.4.8. Onset day

The onset day for an event (e.g. AE, concomitant medication/vaccination) is the number of days between the last study dose and the start date of the event. This is 1 for an event occurring on the same day as a study dose (and reported as starting after study dose).

6.2.4.9. Duration of events

The duration of an event with a start and end date will be the difference between the start and end date plus one day, i.e. an event that starts on 03MAR2018 and ends on 12MAR2018 has a duration of 10 days.

The duration of solicited events will be calculated as the sum of the individual days with the adverse event reported at grade 1 or higher during the solicited event period.

6.2.4.10. Counting rules for occurrences of solicited events

When the occurrences of solicited events are summarized, each event recorded as having occurred during a specific period will be counted as only one occurrence regardless of the number of days on which it occurs. Also, in the case of co-administered study interventions, an administration site event recorded for a participant following multiple study interventions will be counted as only one occurrence.

6.2.5. Display of decimals

6.2.5.1. Percentages

Percentages and their corresponding confidence limits will be displayed with one decimal except for 100% in which case no decimal will be displayed.

6.2.5.2. Differences in percentages

Differences in percentages and their corresponding confidence limits will be displayed with two decimals.

6.2.5.3. Demographic/baseline characteristics statistics

The mean, median, and standard deviation for continuous baseline characteristics (height, weight, body mass index (BMI), pre-dose body temperature) will be presented with one decimal.

The minimum and maximum values and quartile values (if required) will be presented with the same number of decimals as the observed values.

The maxima and minima of transformed height/weight variables will be displayed without decimals.

The maximum and minimum of transformed body temperatures will be displayed with one decimal.

6.2.5.4. Serological summary statistics

The number of decimals used when displaying geometric mean titers (GMT) or concentrations (GMC) and their confidence limits is assay specific based on the magnitude of the assay result post-dose and the clinically relevant assay threshold. The same number of decimals will be used for a given assay regardless of the timepoint presented.

Lowest clinically relevant threshold	Example	Number of decimals to display
<0.3	Diphtheria, tetanus, anti-PRP	3
>=0.3 and <4	*Streptococcus pneumoniae*, Meningococal bactericide	2
>=4 and <1000	Measles, rubella, varicella, polio	1
>=1000	CMI	0

GMT/GMC fold increase from pre-dose follows the same principle. Namely when the lowest clinically relevant threshold is 2 fold, 2 decimals are displayed while when the lowest clinically relevant threshold is 4 fold, 1 decimal is displayed.

GMT or GMC group ratios and their confidence limits will be displayed with 2 decimals regardless of the actual values.

7. REFERENCES

Clopper CJ, Pearson E. The Use of Confidence or Fiducial Limits Illustrated in the case of the Binomial. *Biometrika.* 1934;26:404-13.

Miettinen, O. S. and Nurminen, M. Comparative analysis of two rates. Statistics in Medicine, 1985;4,213-226.