Statistical Analysis Plan

Study ID: 212496

Official Title of Study: A phase 3, randomized, open-label, multi-country study to evaluate the immunogenicity, safety, reactogenicity and persistence of a single dose of the RSVPreF3 OA investigational vaccine and different revaccination schedules in adults aged 60 years and above.

NCT number: NCT04732871

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Date of Document: 27-MAR-2023

Statistical Analysis Plan

Title:	A phase 3, randomized, open-label, multi-country study to evaluate the immunogenicity, safety, reactogenicity and persistence of a single dose of the RSVPreF3 OA investigational vaccine and different revaccination schedules in adults aged 60 years and above.
eTrack study number and Abbreviated Title	212496 (RSV OA=ADJ-004)
Scope:	All data pertaining to the above study except the Safety Review Team analyses for the first 50 participants aged 80 years and above
Date of Statistical Analysis Plan	Amendment 3 Final: 27 Mar 2023

APP 9000058193 Statistical Analysis Plan Template V5 (Effective date: 1July2020)

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LIST OF ABBREVIATIONS

AE	Adverse event
AESI	Adverse Events of Special Interest
ANCOVA	Analysis of Covariance
CD	Community Dwelling
CI	Confidence Interval
COVID-19	Coronavirus Disease 2019
CRF	Case Report Form
CTRS	Clinical Trial Registry Summary
Eli Type	Internal database code for type of elimination code
ELISA	Enzyme-linked immunosorbent assay
ES	Exposed Set
EU/mL	ELISA unit per milliliter
GMC	Geometric mean antibody concentration
GMT	Geometric mean antibody titer
GSK	GlaxoSmithKline
IU/mL	International units per milliliter
LL	Lower Limit of the confidence interval
MedDRA	Medical Dictionary for Regulatory Activities
NA	Not Applicable
PD	Protocol Deviation
PPS	Per-Protocol Set
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SBIR	GSK Biologicals Internet Randomization System
SD	Standard Deviation
SDTM	Study Data Tabulation Model
UL	Upper Limit of the confidence interval

1. DOCUMENT HISTORY

Date	Description	Protocol Version
15 February 2021	First version	Final: 28 October 2020
04 March 2022	Amendment 1:	Final: 28 October 2020
20 June 2022	Amendment 2: • The study design (Figure 1) was updated with the addition of a revaccination dose at Month 24 and a new follow-up visit at Month 25 for the RSV_flexible revaccination group. (Section 3.1). • Table 1 was updated with the addition of a revaccination dose at Month 24 for the RSV_flexible revaccination group (Section 3.1). • Table 6 was updated with the addition of RSV_flexible revaccination group at Months 24 and 25 to the list of elimination codes • SAEs/pIMDs within 30 days following each vaccination (Section 5.4.5)	Final Amendment 1: 01 April 2022
27 Mar 2023	Amendment 3: The elimination code 2100 has been split into 2100h and 2100c (Section 4.2.2). Table 6 has been updated to include these 2 codes. This change will impact the participant disposition tables from ES to PPS for humoral immunogenicity and PPS for CMI. Also the table that shows summary of important protocol deviations leading to elimination from any analyses will be impacted	Amendment 1: 01 April 2022

2. OBJECTIVES/ENDPOINTS

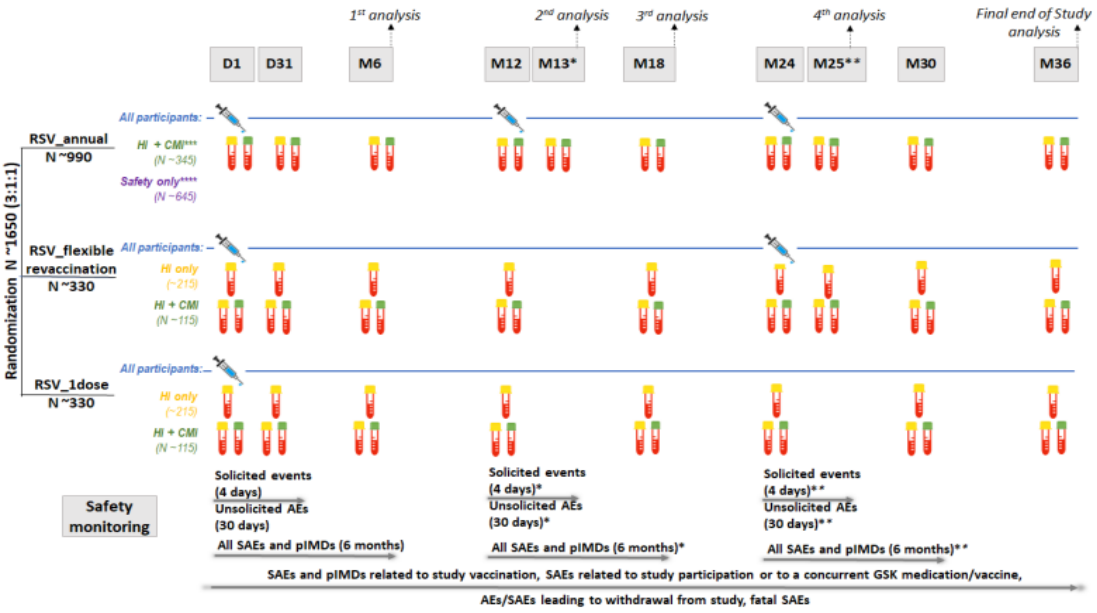
Objectives	Endpoints
Primary	
To evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • Neutralizing antibody titers against RSV-A • Neutralizing antibody titers against RSV-B.
Secondary	
To further evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • RSVPreF3-specific Immunoglobulin G (IgG) antibody concentrations.
To evaluate the humoral immune response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses, up to study end.	Humoral immune response at Months 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Neutralizing antibody titers against RSV-A and RSV-B • RSVPreF3-specific IgG antibody concentrations.
To evaluate the cell-mediated immune (CMI) response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses up to study end.	CMI response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), at Months 6, 12, 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
To evaluate the safety and reactogenicity of each vaccination schedule of the RSVPreF3 OA investigational vaccine in all participants.	• Occurrence of each solicited administration-site and systemic event during a 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) after each vaccination. • Occurrence of any unsolicited AE during a 30-day follow up period (i.e., on the day of vaccination and 29 subsequent days) after each vaccination. • Occurrence of all SAEs and pIMDs up to 6 months after each vaccination. • Occurrence of fatal SAEs, related SAEs and related pIMDs from first vaccination (Day 1) up to study end (Month 36).
Tertiary	
To further characterize immune responses to the RSVPreF3 OA investigational vaccine.	• Any further exploratory immunology in a subset of participants, such as, but not limited to: – Antibodies against specific protein F epitopes. – Potential new immunological markers for protection. – Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing one or any combination of immune marker(s)

CMI = cell-mediated immunity, AE = adverse event, SAE = serious adverse event, pIMD = potential immune-mediated disease.

3. STUDY DESIGN

3.1. Overall design

Figure 1 Study design overview



N = number of participants; D = day; M = month; AE = adverse event; HI = humoral immunity; CMI = cell-mediated immunity; pIMD = potential immune-mediated disease; SAE = serious adverse event.

= blood sample for humoral immune responses (only applicable for ~ 345 participants of the RSV_annual group; and for all participants of the RSV_flexible revaccination and RSV_1dose groups)

= blood sample for CMI (only applicable for ~345 participants of the RSV_annual group, and for ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups).

Note: For group RSV_annual: Revaccination Year 1 = Month 12; Revaccination Year 2 = Month 24; for group RSV_flexible revaccination: Revaccination Year 2 = Month 24.

*Visit and follow-up of solicited events, unsolicited AEs, SAEs and pIMDs are only applicable for RSV_annual group.

**Visit and follow-up of solicited events, unsolicited AEs, SAEs and pIMDs are only applicable for RSV_annual and RSV_flexible revaccination groups.

***For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws and will only be followed up for safety and reactogenicity.

****For those participants in the RSV_annual group without blood sample collections, visits at Months 6, 18, 30 and 36 may be held through a contact. However, contact should be attempted only if site visit is not possible at Months 6, 18, 30 and 36.

- **Type of study**: self-contained.
- **Experimental design**: Phase 3, randomized, open-label, multi-country study with 3 parallel groups (see Figure 1).
- **Duration of the study**: ~ 36 months for each participant.
- **Primary completion date**: Month 12.

- **Control**: none.
- **Blinding**: open-label. Refer to the protocol for details.
- **Data collection**: standardized electronic Case Report Form (eCRF). Solicited events and unsolicited adverse events (AEs) will be collected using a participant Diary card (paper Diary card).
- **Safety monitoring**: the study will be conducted with oversight by the project Safety Review Team (SRT). Please refer to protocol for the description of review of safety data by the SRT.
- **Study groups**: refer to Figure 1 and Table 1 for an overview of the study groups

Table 1 Study groups, intervention and blinding foreseen in the study

Study Groups	Number of participants	Age	Intervention			Blinding
			Primary vaccination	Revaccination Year 1 (Month 12)	Revaccination Year 2 (Month 24)	Open
RSV_annual*	~990	≥60 years	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	X
RSV_flexible revaccination	~330	≥60 years	RSVPreF3 OA investigational vaccine	(none)	RSVPreF3 OA investigational vaccine	X
RSV_1dose	~330	≥60 years	RSVPreF3 OA investigational vaccine	(none)	(none)	X

* RSV_annual group will be split in 3 technical groups in SBIR in order to have a randomization ratio of 1:1:1:1:1: for treatment allocation and avoid predictability.

- **Groups and sub-groups definition for analysis:** Refer to the tables below for a description of the groups and subgroups labels that will be used in the Tables Figures and Listings (TFLs).

  The analyses pertaining to timepoints up to Month 12 will be performed for each group and for the 3 groups pooled. From Month 13 up to study end, the analyses will be performed by group.

  The following group names will be used in the TFLs:

Table 2 Group names and definition for footnote in the TFLs

Group order in tables	Group label in tables	Group definition for footnote
1	RSV_annual	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1, followed by a revaccination dose at 12 months post-Dose 1 and at 24 months post-Dose 1
2	RSV_flexible revaccination	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1 and a revaccination dose at 24 months post-Dose 1
3	RSV_1dose	Participants receiving a single dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1

Table 3 Pooled group names and definition for footnote in the TFLs

Pooled Group label in tables	Groups to be pooled	Pooled definition for footnote	Comment
Total	RSV_annual, RSV_flexible revaccination and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine at Day 1 in all the three groups	Applicable on data reported up to Month 12
Flexible+1dose	RSV_flexible revaccination, and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine at Day 1 in the RSV_flexible revaccination, and RSV_1dose groups	Applicable on data reported at Month 13, Month 18 and Month 24

Table 4 Sub-group names and definitions for footnote in the TFLs

Sub-analysis	Subgroup order in tables	Subgroup label in tables	Subgroup definition for footnote
By age	1	≥65YOA	≥65 years old participants
	2	≥70YOA	≥70 years old participants
	3	≥80YOA	≥80 years old participants
	4	60-69YOA	60-69 years old participants
	5	70-79YOA	70-79 years old participants
By sex	1	Female	Female
	2	Male	Male
By region	1	North America	Participants from North America (United States of America)
	2	Europe	Participants from Europe (Finland, Germany)
	3	Asia	Participants from Asia (Japan, Taiwan)

YOA = Years of age

- **Allocation of participants to assay subsets**

Allocation of participants to assay subsets will be performed using SBIR. The subsets are detailed below.

	RSV_annual	RSV_flexible revaccination	RSV_1dose
HI subset	~345*	All participants (~330)	All participants (~330)
CMI subset	~345*	~115	~115

*For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws, and will only be followed up for safety and reactogenicity.

- **HI subset:** ~345 participants from the RSV_annual group, and all participants from the RSV_flexible revaccination and RSV_1dose groups. These participants will have blood samples collected for testing of humoral immunity at each visit applicable for their study group.
- **CMI subset**: ~345 participants from the RSV_annual group, and ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups. These participants will have additional blood samples collected for CMI testing at each visit applicable for their study group.

4. ANALYSIS SETS

4.1. Definition

Table 5 Populations for analyses

Analysis set	Description
Enrolled set	Participants who agreed to participate in a clinical study after completion of the informed consent process.
Exposed set (ES)	All participants who received at least 1 dose of the study intervention. The allocation in a group is done in function of the administered intervention.
Per Protocol set (PPS)	All participants who received at least 1 dose of the study intervention to which they are randomized and have post-vaccination data, minus participants with protocol deviations that lead to exclusion.

The primary analysis for immunogenicity will be performed on the Per Protocol set (PPS). If the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 5% for at least 1 visit and at least 1 group, a second analysis will be performed on the Exposed Set (ES). The immunogenicity analysis will be performed overall, by sex, by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and by region (North America, Europe, Asia).

4.2. Criteria for eliminating data from Analysis Sets

4.2.1. Elimination from ES

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

4.2.2. Elimination from Per-protocol analysis Set (PPS)

4.2.2.1. Excluded participants

A participant will be excluded from the populations of analysis under the following conditions

- For codes 800, 900, 1030 and 1050: participants will be eliminated for all visits.
- For codes 1040, 1070, 1080, 1090, 2010, 2040, 2050, 2080: participants will be eliminated from a specific visit (at which the condition is met) onwards.
- For codes 2090, 2100h, 2100c, 2120: participants will be eliminated at the specific visit at which the condition is met.

Table 6 List of elimination codes

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
800	Fraudulent data	All	ES and PPS
900	Invalid informed consent	All	ES and PPS
1030	Study intervention not administered at all	All	ES and PPS
1040	Administration of concomitant vaccine(s) forbidden in the protocol • Any investigational or non-registered vaccine other than the study vaccine used during the study period beginning 30 days before the first dose of study vaccine, or planned use during the study period. • Planned or actual administration of a vaccine not foreseen by the study protocol in the period starting 30 days before each dose and ending 30 days after each dose of study vaccine administration, with the exception of inactivated, split virion and subunit influenza vaccines which can be administered up to 14 days before or from 14 days after each study vaccination. • Previous vaccination with an RSV vaccine	All	PPS
1050	Randomization failure	Day 1	PPS
1070	Vaccine administration not according to protocol • Incomplete vaccination course • Participant was vaccinated with the correct vaccine but containing a lower volume • Participant was re-vaccinated while it was not planned	Vaccination visits at Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group and Month 24 applies to the RSV_flexible revaccination group	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Route of the study intervention is not intramuscular • Wrong reconstitution of administered vaccine		
1080	Vaccine administration after a Temperature deviation • Vaccine administered despite a Good Manufacturing Practices (GMP) no-go temperature deviation	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group and Month 24 applies to RSV_flexible revaccination group	PPS
1090	Vaccine administration after expiration	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group and Month 24 belongs to the RSV_flexible revaccination group	PPS
2010	Protocol deviation linked to inclusion/exclusion criteria	All	PPS
2040	Administration of any medication forbidden by the protocol • Any investigational or non-registered medication used during the study period • Administration of long-acting immune-modifying drugs at any time during the study period (e.g. *infliximab*) • Immunoglobulins and/or any blood products administered during the study period • Chronic administration (defined as more than 14 consecutive days in total) of immunosuppressants or other immune-modifying drugs during the period starting	All	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	90 days prior to the study vaccine administration or planned administration during the study period. For corticosteroids, this will mean prednisone ≥20 mg/day, or equivalent.		
2050	Intercurrent medical condition • Participants may be eliminated from the PPS for immunogenicity if, during the study, they incur a condition that has the capability of altering their immune response (intercurrent medical condition) or are confirmed to have an alteration of their initial immune status.	All	PPS
2080	Participants did not comply with dosing schedule • **RSV_annual**: number of days between dose 1 and revaccination dose at Month 12 is outside [350-380 days] • **RSV_annual and RSV_flexible revaccination**: number of days between dose 1 and revaccination dose at Month 24 is outside [715-745 days]	Month 12 and 24	PPS
2090	Participants did not comply with blood sample schedule • Number of days between dose 1 and Day 31 blood sample is outside [30-42 days] • Number of days between dose 1 and Month 6 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 12 blood sample is outside [350-380 days]	Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Number of days between Month 12 and Month 18 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 24 blood sample is outside [715-745 days] • Number of days between Month 24 and Month 30 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 36 blood sample is outside [1080-1110 days] *For RSV_annual group only:* • Number of days between revaccination at Month 12 and Month 13 blood sample is outside [30-42 days] *For RSV_annual and RSV_flexible groups:* • Number of days between revaccination at Month 24 and Month 25 blood sample is outside [30-42 days]		
2600*	Participants not included in the humoral immunogenicity subset	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS
2700*	Participants not included in the cell-mediated immune response subset	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS
2100h	For participants in the humoral immunogenicity subset. • No immunological result at visit x for all the following tests: RSV A/B Neutralizing antibody titer andRSVPreF3-specific IgG antibody concentration	Day 1, Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36 Month 13 and 25 are applicable to the RSV_annual and only Month 25 is applicable to the RSV_flexible revaccination group	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
2100c	For participants in the CMI subset • No immunological result at visit x for all the following tests: Frequency of RSVPreF3-specific CD4+ and CD8+ T cells	Day 1, Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36 Month 13 and 25 are applicable to the RSV_annual and only Month 25 is applicable to the RSV_flexible revaccination group	PPS
2120	Obvious incoherence or abnormality or error in laboratory data • Unreliable released data as a result of confirmed sample mismatch or confirmed inappropriate sample handling at laboratory	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

* codes 2600 and 2700 are not considered as protocol deviations, but those codes will be used to identify participants that are not part of the immunogenicity subsets.

5. STATISTICAL ANALYSES

Standard data derivation rules and stat methods are described in Section 10.1 while the study specific data derivation rules and stat methods are described in Section 9.

For the sake of analysis, data up to Month 12 will be analyzed by group (RSV_annual, RSV_flexible revaccination and RSV_1dose), and also the 3 groups will be pooled. For Month 13, Month 18 and Month 24, the analyses will be tabulated for RSV_annual group versus the pooled RSV_flexible revaccination and RSV_1dose groups. From Month 25 up to study end, the analyses will be performed by group

If more than 5% of participants in the RSV_annual and RSV_flexible revaccination groups receive different number of doses, an analysis by subgroup according to the number of dose(s) administered might be performed. The goal of this analysis is to describe the characteristics of participants in the RSV_annual and RSV_flexible revaccination groups who ultimately receive different numbers of revaccination doses in order to rule out the possibility of systematic differences between these subgroups.

5.1. General considerations

5.1.1. Demography

For a given participant and a given demographic variable, missing measurements will not be replaced.

5.1.2. Immunogenicity

- Any missing or non-evaluable immunogenicity measurement will not be replaced. The descriptive analysis performed for each assay at each timepoint will exclude participants with a missing or non-evaluable measurement. This is applicable to the standard way of computing geometric mean titers/concentrations (GMTs/GMCs).
- During the computation of GMTs/GMCs from the mixed effects model (Section 5.4.3.1). Multiple imputation (MI) technique will not be applied for any missing immunogenicity measurement. This is because the mixed effects model inherently accounts for the missingness, under the assumption that the missing data are missing at random (MAR). Thus considering that missing data are MAR is a reasonable assumption for this descriptive clinical trial, then it is not necessary to perform MI [Rubin et al., 1995]
- The GMTs/GMCs will be computed by taking the anti-logarithm of the arithmetic mean of the $\log_{10}$ transformed titers/concentrations.
- A seronegative participant will be defined as a participant whose antibody titer/concentration is below the technical cut-off value of the assay. A seropositive participant is a participant whose antibody titer/concentration is greater than or equal to the technical cut-off value of the assay.
- Antibody titers/concentrations below the technical assay cut-off will be given an arbitrary value of half the technical assay cut-off for the purpose of GMT/GMC calculation.
- Antibody titers/concentrations above the Upper Limit of Quantification (ULOQ) value will be given the ULOQ value for the purpose of GMT/GMC calculation.
- The mean geometric increase (MGI) is calculated by the geometric mean of ratios of antibody titers/concentrations of each post vaccination time point over pre vaccination (Day 1).

5.1.3. Reactogenicity/Safety

- For a given participant and the analysis of solicited events within 4 days post-vaccination, missing or non-evaluable measurements will not be replaced. Therefore, the analysis of solicited events will include only vaccinated participants with documented solicited safety data (i.e., paper diary completed).
- For analysis of unsolicited AEs, serious adverse events (SAEs), potential immune-mediated diseases (pIMDs) and concomitant medications, all vaccinated participants will be considered. Participants who did not report an event or concomitant medication will be considered as participants without the event or the concomitant medication, respectively.

5.2. Analysis of demography and baseline characteristics

5.2.1. Analysis planned in the protocol

Descriptive summaries will be performed for each group (RSV_annual, RSV_flexible revaccination and RSV_1dose) and overall. The same will be performed in each subset (humoral immune subset and CMI subset).

Demographic/baseline characteristics (age at vaccination in years, sex, race, ethnicity, type of residence (CD/LTCF) and smoking status) will be summarized using descriptive statistics:

- Frequency tables will be generated for categorical variables such as race.
- Mean, median, standard deviation and range will be provided for continuous data such as age.

The distribution of participants will be tabulated as a whole and per group, for each age category, and for each subset.

The number of doses of the study intervention administered will be tabulated by group and by visit.

Withdrawal status will be summarized by group using descriptive statistics:

- The number of participants enrolled into the study as well as the number of participants excluded from Per Protocol (PP) analyses will be tabulated.
- The numbers of participants withdrawn from the study will be tabulated according to the reason for withdrawal.

Participant disposition in the ES and PPS will be reported as a whole and per group

The number and percentage of participants using concomitant medication (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) and during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) will be summarized by group after each vaccine dose and after the revaccination.

5.2.2. Additional considerations

- The analysis of demographic characteristics by group will be performed on the ES and on the PPS.
- Demography and baseline characteristics will also be summarized by sex and region.
- A summary of important protocol deviations leading to elimination from any analyses will be provided by group, based on the Enrolled Set.
- Medical history will be tabulated by System Organ Class (SOC), High Level Term (HLT) and Preferred Term (PT).

5.3. Primary endpoint(s)

5.3.1. Analysis planned in the protocol

The primary analysis for immunogenicity will be performed on the PPS. If the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 5% for at least 1 visit and at least 1 group, a second analysis will be performed on the ES.

5.3.1.1. Humoral immune response – up to Month 12

The analysis of the primary vaccination timepoints up to Month 12 will be performed for each group and for the 3 groups pooled. For each timepoint with blood sample collection for humoral immune response up to Month 12 and for each assay (RSV-A/-B neutralization assays), unless otherwise specified, the following analyses will be performed:

Within groups evaluation:

- Percentage of participants with antibody titers/concentrations above or equal to the technical assay cut-off and their exact 95% confidence interval (CI) will be tabulated.
- GMTs/GMCs and their 95% CI will be tabulated and represented graphically. Furthermore, to account for all the timepoints at which the blood samples are collected, a mixed effects model (see Section 5.4.3.1) will be fitted, from which the GMTs/GMCs will be computed.
- Antibody titers/concentrations will be displayed using reverse cumulative curves.
- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI.

5.4. Secondary endpoint(s)

5.4.1. Humoral immune response

The same analyses as described above for the primary endpoint will be performed for RSVPreF3-specific IgG ELISA up to Month 12 and for each timepoint with blood sample collection for humoral immune response **from study Month 13 up to study end** and for each assay (RSVpreF3-specific IgG **ELISA and RSV-A/-B neutralization assays**). For Month 13, Month 18, and Month 24, the analyses will be tabulated for RSV_annual group versus the pooled RSV_flexible revaccination and RSV_1dose groups. From Month 25 up to study end, the analyses will be performed by group.

Only the analyses requiring further clarification of the exact timepoints analyzed are summarized below:

Within groups evaluation:

- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI:
  - For each post-primary vaccination time point (at Months 18, 24, 30 and 36 when applicable) over pre-primary vaccination (Day 1),
  - For each post-revaccination time point over corresponding pre-revaccination (Months 12 and 24) in the RSV_annual group,
  - For each post-revaccination timepoint over corresponding pre-revaccination (Month 24) in the RSV_flexible revaccination group,
  - For 1 month post-revaccination at Month 12 (Month 13) over 1 month post-Dose 1 vaccination (Day 31) in RSV_annual group.
  - For 1 month post-revaccination at Month 24 (Month 25) over 1 month post-revaccination at Month 12 (Month 13) in RSV_annual group
  - For 1 month post-revaccination at Month 24 (Month 25) over 1 month post-Dose 1 vaccination (Day 31) in RSV_flexible revaccination group.

In addition, the following evaluation will be performed:

- The kinetics of GMTs/GMCs will be plotted as a function of time for participants with results available at all post vaccination timepoints.

The immunogenicity analysis will be performed overall, by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA), and region (North America, Europe, Asia).

5.4.2. Cell-mediated immune response

Within groups evaluation:

The following parameters will be summarized by group at each timepoint for which blood samples are collected, for the 3 groups pooled for time points up to study Month 12, for Month 13, Month 18 and Month 24, the analyses will be tabulated for RSV_annual group versus the pooled RSV_flexible revaccination and RSV_1dose groups. From Month 25 up to study end, the analyses will be performed by group using descriptive statistics (N, geometric mean, min, Q1, median, Q3, max) in the CMI subset:

- Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17, measured by intracellular cytokine staining (ICS) using peripheral blood mononuclear cells PBMCs.
- The kinetics of RSVPreF3-specific CD4+ T cells frequencies will be plotted as a function of time for participants with results available at all timepoints.
- Fold increase of the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17, measured by ICS at each post-primary vaccination timepoint over pre-vaccination (Day 1) by group, at each post-revaccination over the corresponding pre-revaccination (Month 12/Month 24 in the RSV_annual group and Month 24 in the RSV_flexible revaccination group) ,at one month post-revaccination over 1 month post-previous dose in the RSV_annual group [one month post-revaccination at Month 12 (Month 13) over one month post-Dose 1 vaccination (Day 31) and one month post-revaccination at Month 24 (Month 25) over one month post-revaccination at Month 12 (Month 13)] and in the RSV_flexible revaccination group [one month post-revaccination at Month 24 (Month 25) over one month post-Dose 1 vaccination (Day 31)].

The immunogenicity analysis will be performed overall, by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and region (North America, Europe, Asia).

5.4.3. Additional considerations

5.4.3.1. Mixed effects model

For the GMTs/GMCs calculation based on the mixed effects model mentioned in Section 5.3.1.1, repeated measures analysis of covariance (ANCOVA) model will be fitted including age category (60-69 YOA , 70-79 YOA and ≥80 YOA,), study group (RSV_annual, RSV_flexible revaccination and RSV_1dose), sex (male and female), visit (Day 31, Months 6, 12, 13, 18, 24, 25, 30, 36), as fixed effects, pre-vaccination log10-transformed titers/concentrations as a covariate and the response variable is the post-vaccination log10-transformed titers/concentrations. The PROC MIXED procedure in SAS® will be used to carry out the ANCOVA.

The following SAS codes will be used:

For the analysis by study group, study group and visit will be included as the fixed effects in the model

```
Analysis by group
PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid group visit ;
MODEL post-vac=pre-vacc group visit visit*group/ noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=unsubject=subjid;
LSMEANS visit*group/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by age category, age category and visit will be included as the fixed effects in the model. However, this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, age category and visit.

```
Analysis by age-category

PROC MIXED DATA=SERO method=reml empirical ;
 CLASS subjid visit  agecat ;
MODEL post-vac=pre-vacc agecat visit visit* agecat /noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*agecat/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by sex, sex and visit will be included as the fixed effects in the model. However, this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, sex and visit.

```
Analysis by sex

PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid visit  agecat ;
MODEL post-vac=pre-vacc sex visit visit* sex / noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*sex/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by region, region and visit will be included as the fixed effects in the model. However this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, region and visit.

```
Analysis by region

PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid visitregion ;
MODEL post-vac=pre-vacc region visit visit* region / noint scl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*region/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

The GMTs/GMCs, lower and upper confidence limits are obtained from the following data step:

```
DATA LS;
   SET LS;
   gm=10**estimate;
   ll=10**lower;
   ul=10**upper;
 RUN;
```

The above SAS codes may be adapted in case of convergence issues.

5.4.3.2. Cell-mediated immune response

Within groups evaluation

- The frequency of RSVPreF3 specific-CD4+ T cells will be displayed graphically using boxplots (min, Q1, median, Q3, max), by group and timepoint.
- The RSVPreF3-specific CD4+/ CD8+ T cell frequencies will be obtained by subtracting the background value to the antigen-induced value, and by setting to 1 all values less than or equal to zero for geomean calculation and graphical representation. Frequencies will be expressed as the number of cells per million of CD4+ T cells.
- More specifically, the frequencies of RSVPreF3-specific CD4+/ CD8+ T cells expressing at least 2 activation markers including at least one cytokine [$Freq^{2+}$] will be computed as follows:

$$Freq^{2+}_{Background} = \frac{n^{2+}_{background}}{N^{CD4}_{Background}} \quad \text{and} \quad Freq^{2+}_{Induction} = \frac{n^{2+}_{Induction}}{N^{CD4}_{Induction}},$$

And

$$Freq^{2+}_{Specific} = Freq^{2+}_{Induction} - Freq^{2+}_{Background}$$

where

$n^{2+}_{Background}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with medium only (background),

$n^{2+}_{Induction}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with a pool of peptides covering RSVPreF3 (induction),

$N^{CD4}_{Back/Ind}$ = Total number of CD4 T cells involved in the assay (background or induction).

For the computation of the fold increase (post- over pre-vaccination) of the frequency of RSVPreF3-specific CD4+ T-cells identified as expressing **at least 2 activation marker(s) including at least one cytokine*** among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.the results **below the LLOQ** of the assay will be replaced by the value of the LLOQ.

* cytokines are IL-2, TNF-α, IFN-γ, IL-13, and IL-17

5.4.3.3. Assessment of long-term persistence of the immune response at Months 18, 24 and 36 after dose 1

The exploratory analysis for modelling the long-term persistence of immune response (humoral and cellular) post dose 1 vaccination will constitute of the following;

- A piece-wise linear mixed model, power law model and modified power law model [David et al, 2009] for repeated measurements (all data available from Day 31, Month 6, 12, 18, 24, 30, 36 post-Dose 1 vaccination, will be used to model over time
  - the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
  - the neutralizing antibody titers against RSV-A, RSV-B and RSVPreF3-specific IgG antibody concentrations.

The ***piece-wise linear mixed model*** is expressed as follows

The piece-wise model fits the data on non-overlapping time intervals. This corresponds to the observed decay of humoral and cellular antibodies. Each piece of the model uses a linear function. Break points will be selected on the basis of Akaike's Information Criterion (AIC) [Pan, 2001].

$$(ft) = \beta_0 + \beta_1 t \qquad \text{if } x \leq t < xx \text{ months}$$

$$(ft) = \beta_0 + \beta_1 t + \beta_2 (t - xx) \quad \text{if } xx \leq t < xxx \text{ months.}$$

$$(ft) = \beta_0 + \beta_1 t + \beta_2 (t - xx) + \beta_3 (t - xxx) \quad \text{if } t \geq xxx \text{ months}$$

where $f(t)$ is the log-10 transformed antibody titer/concentration/ RSVPreF3-specific CD4+ T cells at time t post-vaccination, β_0 is the intercept and $\beta_1 (i = 1,2,3\ldots)$ are the

parameters corresponding to the time intervals, respectively. All the model effects will be fitted using the SAS PROC MIXED standard code for linear regression methodology for repeated data.

```
ODS OUTPUT  FitStatistics= stat;
ODS OUTPUT  ConvergenceStatus= conv;
PROC MIXED DATA=kin&i.&j. EMPIRICAL COVTEST NOCLPRINT IC;
CLASS pid;
MODEL log_val = int_m int_m_pw_m&i.*m&i. int_m_pw_m&j.*m&j./s cl
covb outp=pred outpm=predm;
RANDOM intercept/SUBJECT=pid;
MAKE "solutionF" out=est;
RUN;
```

The ***power-law model*** is expressed as follows:

$$f(t) = k - a\log(c + t)$$

where $f(t)$ is the log-10 transformed antibody titer/concentration/ RSVPreF3-specific CD4+ T cells at time t post-vaccination, k is the peak log level, a is the decay rate, and c is an arbitrary small constant (set to zero in this case). The estimated maximum likelihood (ML) values for the model parameters will be obtained using the PROC NLMIXED SAS procedure.

```
PROC NLMIXED DATA=kineticm;
PARMS  se=&se  a0 =&a  k0 =&k  sk=&sk sa=&sa ska=&ska;
q = (1/&time)**a;
lg=&log(q);
mean = k + lg;
MODEL log_val ~ NORMAL(mean,se*se);
RANDOM k a ~ NORMAL([k0,a0], [sk*sk, ska, sa*sa]) SUBJECT=pid out=indiv;
RUN;
```

The ***modified power-law model*** is expressed as follows:

It is an extension of the power law model, and it is expressed as follows.

$$f(t) = k + \log[(1 - \pi)(c + t)^a + \pi]$$

The quantity π (between 0 and 1) represents the proportion of antibodies that are produced in the long term. When the value of π is positive, this implies long-term antibody persistence, as noted by [Fraser et al, 2007]. If π = 0, then the modified power-law model reduces to the standard power-law model. The estimated maximum likelihood (ML) values for the model parameters will be obtained using the PROC NLMIXED SAS procedure.

```
PROC NLMIXED DATA=kineticm ;
PARMS  pi=&pi se=&se  a0 =&a  k0 =&k  sk=&sk sa=&sa ska=&ska;
q = (1-pi)*(1/&time)**a;
lg=&log(q+pi);
mean = k + lg;
MODEL log_val ~ NORMAL(mean,se*se);
RANDOM k a ~ NORMAL([k0,a0], [sk*sk, ska, sa*sa]) SUBJECT=pid out =indiv
RUN;
```

- The three models based on data up to Month 36 will be presented graphically. Models based on data up to Month 18 vs. Month 24, Month 24 vs. Month 30 and Month 30 vs. Month 36 will be presented side by side.
- The above models and SAS codes may be adapted in case of convergence issues.

5.4.3.4. Sensitivity analyses

- In case the COVID-19 pandemic is still ongoing during the study conduct, the following sensitivity analyses might be considered.
  - During the assessment of long-term persistence of the immune response, the models described in Section 5.4.3.3 might be fitted taking into account participants who received the first dose of study vaccine and with full follow-up (attending all study visits).
  - Similarly, a mixed model might be fitted for the evaluation of GMT/GMC based on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

5.4.4. Safety analysis

Primary vaccination and revaccination dose:

The safety analysis will be performed on the ES. A descriptive analysis by group, for the 3 groups pooled up to Month 12, for the RSV_flexible revaccination and RSV_1dose groups pooled vs RSV_annual group for Month 13, Month 18 and Month 24, and by group from Month 25 up to study end, will present a summary of:

- The number and percentage of participants with at least one administration site event (solicited and unsolicited), with at least one systemic event (solicited and unsolicited) and with any AE during the 4-day or 30-day follow-up period will be tabulated with exact 95% CI after each dose. The same computations will be done for Grade 3 AEs, and for Grade 3 non-serious AEs and for AEs resulting in medically attended visit.
- The number and percentage of participants reporting each individual solicited administration site event (any grade, Grade 3 and resulting in medically attended visit) and solicited systemic event (any grade, Grade 3 and resulting in medically attended visit) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- For fever, the number and percentage of participants reporting fever by half degree (°C) cumulative increments during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- The incidence of each solicited administration site event and solicited systemic event (any grade and grade 3) will be represented graphically after each dose.

- The number and percentage of participants with any unsolicited AEs during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) with its exact 95% CI will be tabulated for each dose by group and by Medical Dictionary for Regulatory Activities (MedDRA) Primary System Organ Class (SOC), High Level Term (HLT) and preferred term (PT). Similar tabulation will be done for Grade 3 unsolicited AEs, for any causally related unsolicited AEs, for Grade 3 causally related unsolicited AEs and for unsolicited AEs resulting in a medically attended visit. The verbatim reports of unsolicited AEs will be reviewed by a qualified person and the signs and symptoms will be coded according to the MedDRA Dictionary for Adverse Reaction Terminology. Every verbatim term will be matched with the appropriate Preferred Term.
- The number and percentage of participants with at least one report of SAE classified by the MedDRA Primary SOC, HLT and PTs and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI. The same tabulation will be presented for causally related SAEs and fatal SAEs,.
- The number and percentage of participants with at least one report of causally related SAEs classified by the MedDRA Primary SOC, HLT and PTs will be reported after each dose and from Day 1 up to study end with exact 95% CI. The same tabulation will be presented for fatal SAEs.
- All SAEs will also be described in detail in a tabular listing.
- All AEs/SAEs leading to study/intervention discontinuation from dose 1 up to study end will be tabulated.
- The number and percentage of participants with at least one report of pIMD classified by the MedDRA Primary SOC, HLT and PTs and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI. The same tabulation will be presented for causally related pIMDs
- The number and percentage of participants with at least one causally related pIMD classified by the MedDRA Primary SOC, HLT and PTs will be reported after each dose and from Day 1 up to study end with exact 95% CI.
- All pIMDs will also be described in detail in a tabular listing.
- The analyses of unsolicited AEs will include SAEs (unless otherwise specified).

The analysis of safety will also be performed by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA) and region (North America, Europe, Asia).

5.4.5. Additional considerations

- Compliance in completing solicited events information will be tabulated after each dose .
- The number and percentage of participants with at least one administration site event **(solicited only)**, with at least one systemic event **(solicited only)** and with any **solicited** event during the 4-day follow-up period after each dose will be tabulated with exact 95% CI. The same computations will be done for Grade 3 AEs and for AEs resulting in a medically attended visit.
- The number of days with solicited events reported during the 4-day follow-up period will be tabulated for each solicited event, after each dose using descriptive statistics (mean, min, Q1, median, Q3, maximum).
- The number of each solicited events (any and Grade 3) that are ongoing beyond the 4-day follow-up period will be reported with the duration in days (mean, min, Q1, median, Q3, maximum).
- The list of solicited administration site and systemic events, and definition of intensity are described in Section 10.3.3 and 10.3.9.1 of the protocol.
- The number and percentage of participants with at least one report of SAE classified by the MedDRA Primary SOC, HLT and PTs and reported within 30 days following each vaccination will be tabulated with the exact 95% CI. The same tabulation will be presented for pIMDs.

  The measurement of erythema/swelling (in mm) and fever (in °C/F) will be categorized as follows:

Table 7 Intensity grading scale for solicited events

Grading	Erythema/swelling	Fever
0:	≤ 20 mm	< 38.0°C (100.4°F)
1:	> 20 - ≤ 50 mm	≥ 38.0°C(100.4°F) - ≤ 38.5°C(101.3°F)
2:	> 50 - ≤ 100 mm	> 38.5°C(101.3°F) - ≤ 39.0°C(102.2°F)
3:	> 100 mm	> 39.0°C(102.2°F)

Combined Solicited events and Unsolicited AEs

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

Solicited events will be coded by MedDRA as per the following codes:

Solicited event	Lower level term code	Corresponding Lower level term decode
Pain	10022086	Injection site pain
Erythema	10022061	Injection site erythema
Swelling	10053425	Injection site swelling
Fever	10016558	Fever
Headache	10019211	Headache
Fatigue	10016256	Fatigue
Myalgia	10028411	Myalgia
Arthralgia	10003239	Arthralgia

5.4.5.1. Concomitant Medication

Medications will be coded using the GSKDRUG dictionary.

5.4.6. Sensitivity Analyses

If more than 10% of participants miss the revaccination dose or don't have safety data reported after revaccination, the safety analysis described in section 5.4.4 will also be conducted on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

6. ANALYSIS INTERPRETATION

All analyses are descriptive.

7. INTERIM ANALYSES

7.1. Sequence of analyses

Analyses to evaluate objectives and endpoints will be performed stepwise:

- **Month 6:** A first analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 6 are available for all participants. This analysis will be considered as final for those endpoints. *
- **Month 13**: A second analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 12 are available for all participants, as well as data up to 1 month after the first revaccination (Month 13) in the RSV_annual group. This analysis will be considered as final for those endpoints *.

- **Month 18**: A third analysis will be performed on all safety and/or immunogenicity data available and as clean as possible, when data for secondary endpoints up to Month 18 are available for all participants. This analysis will be considered as final for those endpoints *. Safety and immunogenicity analysis may be performed at different time depending on data availability.
- **Month 25**: A fourth analysis will be performed on all safety, reactogenicity and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 24 are available for all participants, as well as data up to 1 month after the second revaccination (Month 25) in the RSV_annual group and up to 1 month after the first revaccination (Month 25) in the RSV_flexible revaccination group. This analysis will be considered as final for those endpoints *.
- The final **End of Study** analysis will be performed when all reactogenicity, safety and immunogenicity data for at least primary and secondary endpoints up to study conclusion are available for all participants (Month 36).
- If the data for tertiary endpoints become available at a later stage, (an) additional analysis/analyses will be performed.
- *Each analysis can be considered as final, as it is based on data that is as clean as possible. However, the consecutive analysis of the same time point might slightly differ at the next analysis e.g when M6 data are re-analyzed at the time of M13 analysis. If major changes are identified, they will be described in the clinical study report.

Description	Analysis ID	Disclosure Purpose (CTRS=public posting, SR=study report, internal)
End of study analysis	E1_01	CTRS, SR
Analysis up to Month 6	E1_02	SR
Analysis up to Month 13	E1_03	CTRS, SR
Analysis up to Month 18	E1_04	Internal
Analysis up to Month 25	E1_05	Internal

7.2. Statistical considerations for interim analyses

All analyses will be conducted on final data (as clean as possible) and therefore no statistical adjustment for interim analyses is required.

8. CHANGES TO THE PROTOCOL DEFINED STATISTICAL ANALYSIS

This statistical analysis plan complements the analyses described in the protocol.

The changes compared to the planned statistical analysis specified in the Protocol amendment 1 (Dated: 01 April 2022) are described below:

- The number and percentage of participants with at least one administration site event **(solicited only)**, with at least one systemic event **(solicited only)** and with any **solicited** event during the 4-day follow-up period after vaccination

- Related SAEs/pIMDs and Fatal SAEs will be tabulated after each dose.
- SAEs/pIMDs within 30 days following each vaccination.

9. NON-STANDARD DATA DERIVATION RULES AND STATISTICAL METHODS

The following sections describe additional derivation rules and statistical methods which are not presented in Section 10.1 (Business rules for standard data derivations and statistical methods).

9.1. Handling of missing data

9.1.1. Dates

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

- AE start dates with missing day:
  - If the month is not the same as the vaccine dose, then the imputed start date will be the 1st of the month
  - If the event starts in the same month as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the vaccine dose given during that month. If 'before vaccination' is selected, the imputed date will be one day before the vaccine dose given during that month.
- AE start dates with missing day and month:
  - If the year is not the same as the vaccine dose, then the imputed start date will be the 1st of January.
  - If the event starts in the same year as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the first vaccine dose given during that year. If 'before vaccination' is selected, the imputed date will be one day before the first vaccine dose given during that year.
- AE end dates with missing day: the imputed end date will be the last day of the month (30 or 31) or the study conclusion date whichever comes first.
- AE end dates with missing day and month: the imputed end date will be the last day of the year (31st of December) or the study conclusion date whichever comes first.

All incomplete concomitant medication/vaccination start/end date will follow the rules above.

10. ANNEXES

10.1. Business rules for standard data derivations and statistical methods

This section contains standard rules for data display and derivation for clinical and epidemiological studies.

10.1.1. 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 AE 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 vaccination is selected, the relative dose for the event will be the one administered on the start day of the event. If 'before vaccination is selected, the relative dose for the event will be the dose prior to this one.

10.1.2. Handling of missing data

10.1.2.1. 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 15th
- A missing day and month will be replaced by June 30th.

See also exceptions in Section 9.1.

10.1.2.2. Laboratory data

Refer to Section 5.1.2 for more details.

10.1.2.3. Daily recording of solicited events

10.1.2.3.1. Studies with paper diaries

For studies using paper diaries which have questions in the CRF indicating the presence or absence of solicited events, the following rules are applicable:

- Denominators for the summary of administration site (or systemic) solicited events will be calculated using the number of participants who respond "Yes" or "No" to the question concerning the occurrence of administration site (or systemic) events.

- When a specific solicited event is marked as having not occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=N for the specified post-dose period for the event in question), all daily measurements will be imputed as Grade 0.
- When a specific solicited event is marked as having occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=Y for the specified post-dose period for the event in question), any missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.
- When the occurrence of a specific solicited event is not present (i.e. SDTM CE.CEOCCUR is neither Y nor N for the specified post-dose period for the event in question) but the group of solicited events (administration site or systemic) is marked as having occurred (i.e. SDTM CE.CEOCCUR=Y), all missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.

The following table shows how participants contribute to each category for a specific solicited event over the Day X to Day Y post-dose period:

Solicited event category	Participants included in the calculation of the numerator
Any	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y or with the adverse event marked as present and at least one missing daily recording between Day X and Day Y
At least grade 1	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y
At least grade 2	All participants with at least one occurrence of the adverse event at grade 2 or grade 3 between Day X and Day Y
At least grade 3	All participants with at least one occurrence of the adverse event at grade 3 between Day X and Day Y

10.1.2.4. Unsolicited adverse events

Unsolicited AE summaries are including SAEs unless specified otherwise.

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

10.1.3. Data derivation

10.1.3.1. 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

In this study, we will collect only the year of birth. The rules for handling missing day and/or month in the DOB are given in Section 10.1.2.1.

Since only the year of birth will be collected in the eCRF, there might be some discrepancies between the age computed using standard derivation rules and the age category used in SBIR for the minimization.

Therefore, for the analysis by age, the age categories defined in Table 4 will be determined according to the information entered in SBIR (variable AGEGRP in SDTM SUPPDM), except for "≥65 YOA" category which will be obtained using the derived age because this category is not used in SBIR for minimization

10.1.3.2. 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

10.1.3.3. Numerical serology results

Numerical serology results will be derived from the content of IS.ISORRES in the SDTM dataset. For humoral 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 and value is <=ULOQ	value
"value" and value is > ULOQ	ULOQ*
All other cases	Missing

*This rule will be used to compute descriptive statistics (GMTs, fold increase etc).

10.1.3.4. Geometric mean titers (GMTs) and concentrations (GMCs)

GMT or GMC calculations are performed by taking the inverse logarithm of the mean of the log titer or concentration transformations. Non quantifiable antibody titers or concentrations will be converted as described in Section 10.1.3.3 for the purpose of GMT/GMC calculation. Cut-off values are defined by the laboratory before the analysis.

10.1.3.5. 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).

10.1.3.6. 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 3 March 2018 and ends on 12 March 2018 has a duration of 10 days.

The duration of solicited events will be calculated as the sum of the individual days with the event reported at grade 1 or higher or reported as missing ("NOT DONE"), during the solicited event period. The duration of solicited events ongoing beyond the solicited period will be calculated as end date – start date + 1. The goal of this change was to align this text with the standard programs.

10.1.3.7. Counting rules for combining solicited and unsolicited AEs

For output combining solicited and unsolicited AEs, all SAEs will be considered systemic events since the administration site flag is not included in the Expedited Adverse Event CRF pages. Unsolicited AEs with missing administration site flag will also be considered systemic.

Multiple events with the same PT which start on the same day are counted as only one occurrence.

In case a solicited event is worsening to Grade 3 after the solicited period, it should be reported only in the following table.

Summary of grade 3 AEs (solicited and unsolicited) within 30 days following each dose and overall

10.1.3.8. 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.

10.1.4. Display of decimals

10.1.4.1. Percentages

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

10.1.4.2. Demographic/baseline characteristics statistics

The mean, median, and SD for continuous baseline characteristics 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 maximum and minimum of transformed body temperatures will be displayed with one decimal.

10.1.4.3. Serological summary statistics

For each assay, GMT or GMC and their confidence limits will be presented with one decimal, as well as GMT/GMC fold increase from pre-dose.

The mean, median, SD and quartile values for frequency of RSVPreF3-specific-CD4+ and CD8+ T cells and for the fold increase (post- over pre-vaccination) will be presented with one decimal. The minimum and maximum values will be presented with no decimal.

10.1.5. Statistical methodology

10.1.5.1. Exact confidence intervals around proportions

The exact confidence intervals around within-group proportions are derived using the method of Clopper and Pearson [Clopper, 1934].

11. 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.

12. REFERENCES

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

David M-P, Van Herck K, Hardt K, Tibaldi F, Dubin G, Descamps D, et al. Long-term persistence of anti-HPV-16 and-18 antibodies induced by vaccination with the AS04-adjuvanted cervical cancer vaccine: modeling of sustained antibody responses. GYNECOLOGIC ONCOLOGY. 2009;115(3):S1–6.

Fraser C, Tomassini JE, Xi L, Golm G, Watson M, Giuliano AR, et al. Modeling the long-term antibody response of a human papillomavirus (HPV) virus-like particle (VLP) type 16 prophylactic vaccine. Vaccine 2007 May 22;25(21):4324-33

Pan W Akaike's information criterion in generalized estimating equations Biometrics, 57 (1) (2001), pp. 120-125

Rubin, D. B., Stern, H. S. AND Vehovar, V. Handling “don’t know” survey responses: the case of the Slovenian plebiscite. Journal of the American Statistical Association 90, 1995, 822–828

	**Statistical Analysis Plan**
Title:	A phase 3, randomized, open-label, multi-country study to evaluate the immunogenicity, safety, reactogenicity and persistence of a single dose of the RSVPreF3 OA investigational vaccine and different revaccination schedules in adults aged 60 years and above.
eTrack study number and Abbreviated Title	212496 (RSV OA=ADJ-004)
Scope:	All data pertaining to the above study except the Safety Review Team analyses for the first 50 participants aged 80 years and above
Date of Statistical Analysis Plan	Amendment 2 Final: 20 Jun 2022

APP 9000058193 Statistical Analysis Plan Template V5 (Effective date: 1July2020)

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LIST OF ABBREVIATIONS

AE	Adverse event
AESI	Adverse Events of Special Interest
ANCOVA	Analysis of Covariance
CD	Community Dwelling
CI	Confidence Interval
COVID-19	Coronavirus Disease 2019
CRF	Case Report Form
CTRS	Clinical Trial Registry Summary
Eli Type	Internal database code for type of elimination code
ELISA	Enzyme-linked immunosorbent assay
EU/mL	ELISA unit per milliliter
ES	Exposed Set
GMC	Geometric mean antibody concentration
GMT	Geometric mean antibody titer
GSK	GlaxoSmithKline
IU/mL	International units per milliliter
LL	Lower Limit of the confidence interval
MedDRA	Medical Dictionary for Regulatory Activities
NA	Not Applicable
PD	Protocol Deviation
PPS	Per-Protocol Set
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SBIR	GSK Biologicals Internet Randomization System
SD	Standard Deviation
SDTM	Study Data Tabulation Model
UL	Upper Limit of the confidence interval

1. DOCUMENT HISTORY

Date	Description	Protocol Version
15 February 2021 04 March 2022 20 Jun 2022	First version Amendment 1: Amendment 2: • The study design (Figure 1) was updated with the addition of a revaccination dose at Month 24 and a new follow-up visit at Month 25 for the RSV_flexible revaccination group. (Section 3.1). • Table 1 was updated with the addition of a revaccination dose at Month 24 for the RSV_flexible revaccination group (Section 3.1). • Table 6 was updated with the addition of RSV_flexible revaccination group at Months 24 and 25 to the list of elimination codes • SAEs/pIMDs within 30 days following each vaccination (Section 5.4.5)	Final: 28 October 2020 Final: 28 October 2020Final Amendment 1: 01 April 2022

2. OBJECTIVES/ENDPOINTS

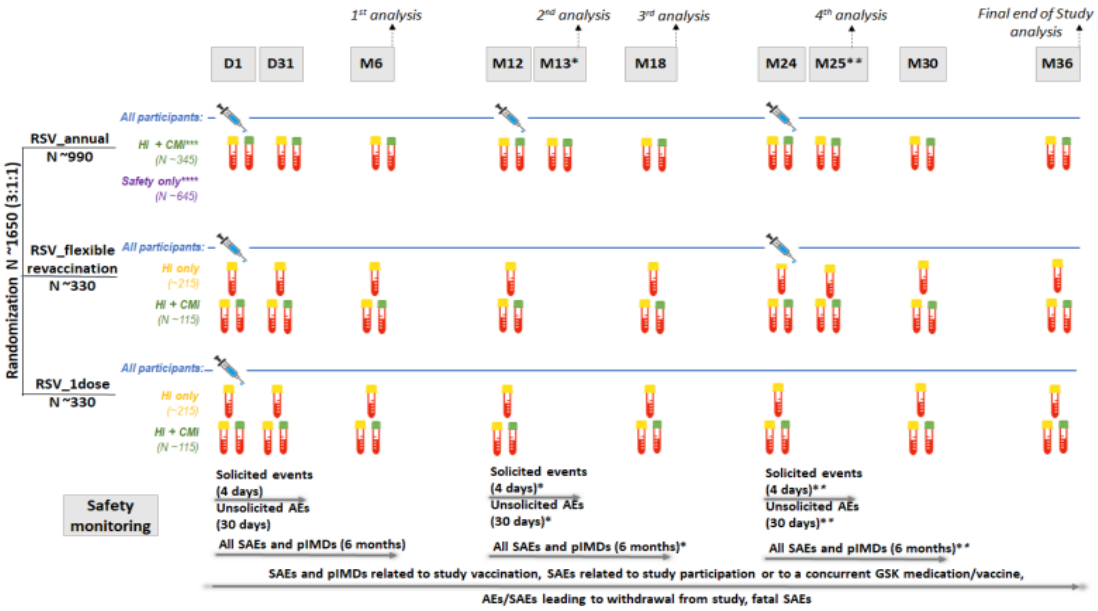
Objectives	Endpoints
Primary	
To evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • Neutralizing antibody titers against RSV-A • Neutralizing antibody titers against RSV-B.
Secondary	
To further evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • RSVPreF3-specific Immunoglobulin G (IgG) antibody concentrations.
To evaluate the humoral immune response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses, up to study end.	Humoral immune response at Months 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Neutralizing antibody titers against RSV-A and RSV-B • RSVPreF3-specific IgG antibody concentrations.
To evaluate the cell-mediated immune (CMI) response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses up to study end.	CMI response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), at Months 6, 12, 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
To evaluate the safety and reactogenicity of each vaccination schedule of the RSVPreF3 OA investigational vaccine in all participants.	• Occurrence of each solicited administration-site and systemic event during a 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) after each vaccination. • Occurrence of any unsolicited AE during a 30-day follow up period (i.e., on the day of vaccination and 29 subsequent days) after each vaccination. • Occurrence of all SAEs and pIMDs up to 6 months after each vaccination. • Occurrence of fatal SAEs, related SAEs and related pIMDs from first vaccination (Day 1) up to study end (Month 36).
Tertiary	
To further characterize immune responses to the RSVPreF3 OA investigational vaccine.	• Any further exploratory immunology in a subset of participants, such as, but not limited to: – Antibodies against specific protein F epitopes. – Potential new immunological markers for protection. – Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing one or any combination of immune marker(s)

CMI = cell-mediated immunity, AE = adverse event, SAE = serious adverse event, pIMD = potential immune-mediated disease.

3. STUDY DESIGN

3.1. Overall design

Figure 1 Study design overview



N = number of participants; D = day; M = month; AE = adverse event; HI = humoral immunity; CMI = cell-mediated immunity; pIMD = potential immune-mediated disease; SAE = serious adverse event.

= blood sample for humoral immune responses (only applicable for ~ 345 participants of the RSV_annual group; and for all participants of the RSV_flexible revaccination and RSV_1dose groups)

= blood sample for CMI (only applicable for ~345 participants of the RSV_annual group, and for ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups).

Note: For group RSV_annual: Revaccination Year 1 = Month 12; Revaccination Year 2 = Month 24; for group RSV_flexible revaccination: Revaccination Year 2 = Month 24.

*Visit and follow-up of solicited events, unsolicited AEs, SAEs and pIMDs are only applicable for RSV_annual group.

**Visit and follow-up of solicited events, unsolicited AEs, SAEs and pIMDs are only applicable for RSV_annual and RSV_flexible revaccination groups.

***For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws and will only be followed up for safety and reactogenicity.

****For those participants in the RSV_annual group without blood sample collections, visits at Months 6, 18, 30 and 36 may be held through a contact. However, contact should be attempted only if site visit is not possible at Months 6, 18, 30 and 36.

- **Type of study**: self-contained.
- **Experimental design**: Phase 3, randomized, open-label, multi-country study with 3 parallel groups (see Figure 1).
- **Duration of the study**: ~ 36 months for each participant.
- **Primary completion date**: Month 12.
- **Control**: none.

- **Blinding**: open-label. Refer to the protocol for details.
- **Data collection**: standardized electronic Case Report Form (eCRF). Solicited events and unsolicited adverse events (AEs) will be collected using a participant Diary card (paper Diary card).
- **Safety monitoring**: the study will be conducted with oversight by the project Safety Review Team (SRT). Please refer to protocol for the description of review of safety data by the SRT.
- **Study groups**: refer to Figure 1 and Table 1 for an overview of the study groups

Table 1 Study groups, intervention and blinding foreseen in the study

Study Groups	Number of participants	Age	Intervention			Blinding
			Primary vaccination	Revaccination Year 1 (Month 12)	Revaccination Year 2 (Month 24)	Open
RSV_annual*	~990	≥60 years	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	X
RSV_flexible revaccination	~330	≥60 years	RSVPreF3 OA investigational vaccine	(none)	RSVPreF3 OA investigational vaccine	X
RSV_1dose	~330	≥60 years	RSVPreF3 OA investigational vaccine	(none)	(none)	X

* RSV_annual group will be split in 3 technical groups in SBIR in order to have a randomization ratio of 1:1:1:1:1: for treatment allocation and avoid predictability.

- **Groups and sub-groups definition for analysis:** Refer to the tables below for a description of the groups and subgroups labels that will be used in the Tables Figures and Listings (TFLs).

  The analyses pertaining to timepoints up to Month 12 will be performed for each group and for the 3 groups pooled. From Month 13 up to study end, the analyses will be performed by group.

  The following group names will be used in the TFLs:

Table 2 Group names and definition for footnote in the TFLs

Group order in tables	Group label in tables	Group definition for footnote
1	RSV_annual	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1, followed by a revaccination dose at 12 months post-Dose 1 and at 24 months post-Dose 1
2	RSV_flexible revaccination	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1 and a revaccination dose at 24 months post-Dose 1
3	RSV_1dose	Participants receiving a single dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1

Table 3 Pooled group names and definition for footnote in the TFLs

Pooled Group label in tables	Groups to be pooled	Pooled definition for footnote	Comment
Total	RSV_annual, RSV_flexible revaccination and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine at Day 1 in all the three groups	Applicable on data reported up to Month 12
Flexible+1dose	RSV_flexible revaccination, and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine at Day 1 in the RSV_flexible revaccination, and RSV_1dose groups	Applicable on data reported at Month 13, Month 18 and Month 24

Table 4 Sub-group names and definitions for footnote in the TFLs

Sub-analysis	Subgroup order in tables	Subgroup label in tables	Subgroup definition for footnote
By age	1	≥65YOA	≥65 years old participants
	2	≥70YOA	≥70 years old participants
	3	≥80YOA	≥80 years old participants
	4	60-69YOA	60-69 years old participants
	5	70-79YOA	70-79 years old participants
By sex	1	Female	Female
	2	Male	Male
By region	1	North America	Participants from North America (United States of America)
	2	Europe	Participants from Europe (Finland, Germany)
	3	Asia	Participants from Asia (Japan, Taiwan)

YOA = Years of age

- **Allocation of participants to assay subsets**

Allocation of participants to assay subsets will be performed using SBIR. The subsets are detailed below.

	RSV_annual	RSV_flexible revaccination	RSV_1dose
HI subset	~345*	All participants (~330)	All participants (~330)
CMI subset	~345*	~115	~115

*For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws, and will only be followed up for safety and reactogenicity.

- **HI subset:** ~345 participants from the RSV_annual group, and all participants from the RSV_flexible revaccination and RSV_1dose groups. These participants will have blood samples collected for testing of humoral immunity at each visit applicable for their study group.
- **CMI subset**: ~345 participants from the RSV_annual group, and ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups. These participants will have additional blood samples collected for CMI testing at each visit applicable for their study group.

4. ANALYSIS SETS

4.1. Definition

Table 5 Populations for analyses

Analysis set	Description
Enrolled set	Participants who agreed to participate in a clinical study after completion of the informed consent process.
Exposed set (ES)	All participants who received at least 1 dose of the study intervention. The allocation in a group is done in function of the administered intervention.
Per Protocol set (PPS)	All participants who received at least 1 dose of the study intervention to which they are randomized and have post-vaccination data, minus participants with protocol deviations that lead to exclusion.

The primary analysis for immunogenicity will be performed on the Per Protocol set (PPS). If the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 5% for at least 1 visit and at least 1 group, a second analysis will be performed on the Exposed Set (ES). The immunogenicity analysis will be performed overall, by sex, by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and by region (North America, Europe, Asia).

4.2. Criteria for eliminating data from Analysis Sets

4.2.1. Elimination from ES

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

4.2.2. Elimination from Per-protocol analysis Set (PPS)

4.2.2.1. Excluded participants

A participant will be excluded from the populations of analysis under the following conditions

- For codes 800, 900, 1030 and 1050: participants will be eliminated for all visits.
- For codes 1040, 1070, 1080, 1090, 2010, 2040, 2050, 2080: participants will be eliminated from a specific visit (at which the condition is met) onwards.
- For codes 2090, 2100, 2120: participants will be eliminated at the specific visit at which the condition is met.

Table 6 List of elimination codes

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
800	Fraudulent data	All	ES and PPS
900	Invalid informed consent	All	ES and PPS
1030	Study intervention not administered at all	All	ES and PPS
1040	Administration of concomitant vaccine(s) forbidden in the protocol • Any investigational or non-registered vaccine other than the study vaccine used during the study period beginning 30 days before the first dose of study vaccine, or planned use during the study period. • Planned or actual administration of a vaccine not foreseen by the study protocol in the period starting 30 days before each dose and ending 30 days after each dose of study vaccine administration, with the exception of inactivated, split virion and subunit influenza vaccines which can be administered up to 14 days before or from 14 days after each study vaccination. • Previous vaccination with an RSV vaccine	All	PPS
1050	Randomization failure	Day 1	PPS
1070	Vaccine administration not according to protocol • Incomplete vaccination course • Participant was vaccinated with the correct vaccine but containing a lower volume	Vaccination visits at Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group and Month 24 applies	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Participant was re-vaccinated while it was not planned • Route of the study intervention is not intramuscular • Wrong reconstitution of administered vaccine	to the RSV_flexible revaccination group	
1080	Vaccine administration after a Temperature deviation • Vaccine administered despite a Good Manufacturing Practices (GMP) no-go temperature deviation	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group and Month 24 applies to RSV_flexible revaccination group	PPS
1090	Vaccine administration after expiration	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group and Month 24 belongs to the RSV_flexible revaccination group	PPS
2010	Protocol deviation linked to inclusion/exclusion criteria	All	PPS
2040	Administration of any medication forbidden by the protocol • Any investigational or non-registered medication used during the study period • Administration of long-acting immune-modifying drugs at any time during the study period (e.g. *infliximab*) • Immunoglobulins and/or any blood products administered during the study period	All	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Chronic administration (defined as more than 14 consecutive days in total) of immunosuppressants or other immune-modifying drugs during the period starting 90 days prior to the study vaccine administration or planned administration during the study period. For corticosteroids, this will mean prednisone ≥20 mg/day, or equivalent.		
2050	Intercurrent medical condition • Participants may be eliminated from the PPS for immunogenicity if, during the study, they incur a condition that has the capability of altering their immune response (intercurrent medical condition) or are confirmed to have an alteration of their initial immune status.	All	PPS
2080	Participants did not comply with dosing schedule • **RSV_annual**: number of days between dose 1 and revaccination dose at Month 12 is outside [350-380 days] • **RSV_annual and RSV_flexible revaccination**: number of days between dose 1 and revaccination dose at Month 24 is outside [715-745 days]	Month 12 and 24	PPS
2090	Participants did not comply with blood sample schedule • Number of days between dose 1 and Day 31 blood sample is outside [30-42 days]	Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Number of days between dose 1 and Month 6 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 12 blood sample is outside [350-380 days] • Number of days between Month 12 and Month 18 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 24 blood sample is outside [715-745 days] • Number of days between Month 24 and Month 30 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 36 blood sample is outside [1080-1110 days] *For RSV_annual group only:* • Number of days between revaccination at Month 12 and Month 13 blood sample is outside [30-42 days] *For RSV_annual and RSV_flexible groups:* • Number of days between revaccination at Month 24 and Month 25 blood sample is outside [30-42 days]		
2600*	Participants not included in the humoral immunogenicity subset	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS
2700*	Participants not included in the cell-mediated immune response subset	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS
2100	Serological results not available • No immunological result at visit x for all the following	Day 1, Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	tests: RSV A/B Neutralizing antibody titer, RSVPreF3-specific IgG antibody concentration and RSVPreF3-specific CD4+/CD8+ T cells frequency	Month 13 and 25 are applicable to the RSV_annual and only Month 25 is applicable to the RSV_flexible revaccination group	
2120	Obvious incoherence or abnormality or error in laboratory data • Unreliable released data as a result of confirmed sample mismatch or confirmed inappropriate sample handling at laboratory	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

* codes 2600 and 2700 are not considered as protocol deviations, but those codes will be used to identify participants that are not part of the immunogenicity subsets.

5. STATISTICAL ANALYSES

Standard data derivation rules and stat methods are described in Section 10.1 while the study specific data derivation rules and stat methods are described in Section 9.

For the sake of analysis, data up to Month 12 will be analyzed by group (RSV_annual, RSV_flexible revaccination and RSV_1dose), and also the 3 groups will be pooled. For Month 13, Month 18 and Month 24, the analyses will be tabulated for RSV_annual group versus the pooled RSV_flexible revaccination and RSV_1dose groups. From Month 25 up to study end, the analyses will be performed by group

If more than 5% of participants in the RSV_annual and RSV_flexible revaccination groups receive different number of doses, an analysis by subgroup according to the number of dose(s) administered might be performed. The goal of this analysis is to describe the characteristics of participants in the RSV_annual and RSV_flexible revaccination groups who ultimately receive different numbers of revaccination doses in order to rule out the possibility of systematic differences between these subgroups.

5.1. General considerations

5.1.1. Demography

For a given participant and a given demographic variable, missing measurements will not be replaced.

5.1.2. Immunogenicity

- Any missing or non-evaluable immunogenicity measurement will not be replaced. The descriptive analysis performed for each assay at each timepoint will exclude participants with a missing or non-evaluable measurement. This is applicable to the standard way of computing geometric mean titers/concentrations (GMTs/GMCs).
- During the computation of GMTs/GMCs from the mixed effects model (Section 5.4.3.1). Multiple imputation (MI) technique will not be applied for any missing immunogenicity measurement. This is because the mixed effects model inherently accounts for the missingness, under the assumption that the missing data are missing at random (MAR). Thus considering that missing data are MAR is a reasonable assumption for this descriptive clinical trial, then it is not necessary to perform MI [Rubin et al., 1995]
- The GMTs/GMCs will be computed by taking the anti-logarithm of the arithmetic mean of the $\log_{10}$ transformed titers/concentrations.
- A seronegative participant will be defined as a participant whose antibody titer/concentration is below the technical cut-off value of the assay. A seropositive participant is a participant whose antibody titer/concentration is greater than or equal to the technical cut-off value of the assay.
- Antibody titers/concentrations below the technical assay cut-off will be given an arbitrary value of half the technical assay cut-off for the purpose of GMT/GMC calculation.
- Antibody titers/concentrations above the Upper Limit of Quantification (ULOQ) value will be given the ULOQ value for the purpose of GMT/GMC calculation.
- The mean geometric increase (MGI) is calculated by the geometric mean of ratios of antibody titers/concentrations of each post vaccination time point over pre vaccination (Day 1).

5.1.3. Reactogenicity/Safety

- For a given participant and the analysis of solicited events within 4 days post-vaccination, missing or non-evaluable measurements will not be replaced. Therefore, the analysis of solicited events will include only vaccinated participants with documented solicited safety data (i.e., paper diary completed).
- For analysis of unsolicited AEs, serious adverse events (SAEs), potential immune-mediated diseases (pIMDs) and concomitant medications, all vaccinated participants will be considered. Participants who did not report an event or concomitant medication will be considered as participants without the event or the concomitant medication, respectively.

5.2. Analysis of demography and baseline characteristics

5.2.1. Analysis planned in the protocol

Descriptive summaries will be performed for each group (RSV_annual, RSV_flexible revaccination and RSV_1dose) and overall. The same will be performed in each subset (humoral immune subset and CMI subset).

Demographic/baseline characteristics (age at vaccination in years, sex, race, ethnicity, type of residence (CD/LTCF) and smoking status) will be summarized using descriptive statistics:

- Frequency tables will be generated for categorical variables such as race.
- Mean, median, standard deviation and range will be provided for continuous data such as age.

The distribution of participants will be tabulated as a whole and per group, for each age category, and for each subset.

The number of doses of the study intervention administered will be tabulated by group and by visit.

Withdrawal status will be summarized by group using descriptive statistics:

- The number of participants enrolled into the study as well as the number of participants excluded from Per Protocol (PP) analyses will be tabulated.
- The numbers of participants withdrawn from the study will be tabulated according to the reason for withdrawal.

Participant disposition in the ES and PPS will be reported as a whole and per group

The number and percentage of participants using concomitant medication (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) and during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) will be summarized by group after each vaccine dose and after the revaccination.

5.2.2. Additional considerations

- The analysis of demographic characteristics by group will be performed on the ES and on the PPS.
- Demography and baseline characteristics will also be summarized by sex and region.
- A summary of important protocol deviations leading to elimination from any analyses will be provided by group, based on the Enrolled Set.
- Medical history will be tabulated by System Organ Class (SOC), High Level Term (HLT) and Preferred Term (PT).

5.3. Primary endpoint(s)

5.3.1. Analysis planned in the protocol

The primary analysis for immunogenicity will be performed on the PPS. If the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 5% for at least 1 visit and at least 1 group, a second analysis will be performed on the ES.

5.3.1.1. Humoral immune response – up to Month 12

The analysis of the primary vaccination timepoints up to Month 12 will be performed for each group and for the 3 groups pooled. For each timepoint with blood sample collection for humoral immune response up to Month 12 and for each assay (RSV-A/-B neutralization assays), unless otherwise specified, the following analyses will be performed:

Within groups evaluation:

- Percentage of participants with antibody titers/concentrations above or equal to the technical assay cut-off and their exact 95% confidence interval (CI) will be tabulated.
- GMTs/GMCs and their 95% CI will be tabulated and represented graphically. Furthermore, to account for all the timepoints at which the blood samples are collected, a mixed effects model (see Section 5.4.3.1) will be fitted, from which the GMTs/GMCs will be computed.
- Antibody titers/concentrations will be displayed using reverse cumulative curves.
- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI.

5.4. Secondary endpoint(s)

5.4.1. Humoral immune response

The same analyses as described above for the primary endpoint will be performed for RSVPreF3-specific IgG ELISA up to Month 12 and for each timepoint with blood sample collection for humoral immune response **from study Month 13 up to study end** and for each assay (RSVpreF3-specific IgG **ELISA and RSV-A/-B neutralization assays**). For Month 13, Month 18, and Month 24, the analyses will be tabulated for RSV_annual group versus the pooled RSV_flexible revaccination and RSV_1dose groups. From Month 25 up to study end, the analyses will be performed by group.

Only the analyses requiring further clarification of the exact timepoints analyzed are summarized below:

Within groups evaluation:

- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI:
  - For each post-primary vaccination time point (at Months 18, 24, 30 and 36 when applicable) over pre-primary vaccination (Day 1),
  - For each post-revaccination time point over corresponding pre-revaccination (Months 12 and 24) in the RSV_annual group,
  - For each post-revaccination timepoint over corresponding pre-revaccination (Month 24) in the RSV_flexible revaccination group,
  - For 1 month post-revaccination at Month 12 (Month 13) over 1 month post-Dose 1 vaccination (Day 31) in RSV_annual group.
  - For 1 month post-revaccination at Month 24 (Month 25) over 1 month post-revaccination at Month 12 (Month 13) in RSV_annual group
  - For 1 month post-revaccination at Month 24 (Month 25) over 1 month post-Dose 1 vaccination (Day 31) in RSV_flexible revaccination group.

In addition, the following evaluation will be performed:

- The kinetics of GMTs/GMCs will be plotted as a function of time for participants with results available at all post vaccination timepoints.

The immunogenicity analysis will be performed overall, by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA), and region (North America, Europe, Asia).

5.4.2. Cell-mediated immune response

Within groups evaluation:

The following parameters will be summarized by group at each timepoint for which blood samples are collected, for the 3 groups pooled for time points up to study Month 12, for Month 13, Month 18 and Month 24, the analyses will be tabulated for RSV_annual group versus the pooled RSV_flexible revaccination and RSV_1dose groups. From Month 25 up to study end, the analyses will be performed by group using descriptive statistics (N, geometric mean, min, Q1, median, Q3, max) in the CMI subset:

- Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17, measured by intracellular cytokine staining (ICS) using peripheral blood mononuclear cells PBMCs.
- The kinetics of RSVPreF3-specific CD4+ T cells frequencies will be plotted as a function of time for participants with results available at all timepoints.
- Fold increase of the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17, measured by ICS at each post-primary vaccination timepoint over pre-vaccination (Day 1) by group, at each post-

revaccination over the corresponding pre-revaccination (Month 12/Month 24 in the RSV_annual group and Month 24 in the RSV_flexible revaccination group) ,at one month post-revaccination over 1 month post-previous dose in the RSV_annual group [one month post-revaccination at Month 12 (Month 13) over one month post-Dose 1 vaccination (Day 31) and one month post-revaccination at Month 24 (Month 25) over one month post-revaccination at Month 12 (Month 13)] and in the RSV_flexible revaccination group [one month post-revaccination at Month 24 (Month 25) over one month post-Dose 1 vaccination (Day 31)].

The immunogenicity analysis will be performed overall, by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and region (North America, Europe, Asia).

5.4.3. Additional considerations

5.4.3.1. Mixed effects model

For the GMTs/GMCs calculation based on the mixed effects model mentioned in Section 5.3.1.1, repeated measures analysis of covariance (ANCOVA) model will be fitted including age category (60-69 YOA , 70-79 YOA and ≥80 YOA,), study group (RSV_annual, RSV_flexible revaccination and RSV_1dose), sex (male and female), visit (Day 31, Months 6, 12, 13, 18, 24, 25, 30, 36), as fixed effects, pre-vaccination log10-transformed titers/concentrations as a covariate and the response variable is the post-vaccination log10-transformed titers/concentrations. The PROC MIXED procedure in SAS® will be used to carry out the ANCOVA.

The following SAS codes will be used:

For the analysis by study group, study group and visit will be included as the fixed effects in the model

```
Analysis by group
PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid group visit ;
MODEL post-vac=pre-vacc group visit visit*group/ noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=unsubject=subjid;
LSMEANS visit*group/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by age category, age category and visit will be included as the fixed effects in the model. However, this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, age category and visit.

Analysis by age-category

```
PROC MIXED DATA=SERO method=reml empirical ;
 CLASS subjid visit  agecat ;
MODEL post-vac=pre-vacc agecat visit visit* agecat /noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*agecat/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by sex, sex and visit will be included as the fixed effects in the model. However, this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, sex and visit.

Analysis by sex

```
PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid visit  agecat ;
MODEL post-vac=pre-vacc sex visit visit* sex / noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*sex/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by region, region and visit will be included as the fixed effects in the model. However this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, region and visit.

```
Analysis by region

PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid visitregion ;
MODEL post-vac=pre-vacc region visit visit* region / noint scl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*region/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

The GMTs/GMCs, lower and upper confidence limits are obtained from the following data step:

```
DATA LS;
   SET LS;
   gm=10**estimate;
   ll=10**lower;
   ul=10**upper;
 RUN;
```

The above SAS codes may be adapted in case of convergence issues.

5.4.3.2. Cell-mediated immune response

Within groups evaluation

- The frequency of RSVPreF3 specific-CD4+ T cells will be displayed graphically using boxplots (min, Q1, median, Q3, max), by group and timepoint.
- The RSVPreF3-specific CD4+/ CD8+ T cell frequencies will be obtained by subtracting the background value to the antigen-induced value, and by setting to 1 all values less than or equal to zero for geomean calculation and graphical representation. Frequencies will be expressed as the number of cells per million of CD4+ T cells.
- More specifically, the frequencies of RSVPreF3-specific CD4+/ CD8+ T cells expressing at least 2 activation markers including at least one cytokine [$Freq^{2+}$] will be computed as follows:

$$Freq^{2+}_{Background} = \frac{n^{2+}_{background}}{N^{CD4}_{Background}} \quad \text{and} \quad Freq^{2+}_{Induction} = \frac{n^{2+}_{Induction}}{N^{CD4}_{Induction}},$$

And

$$Freq^{2+}_{Specific} = Freq^{2+}_{Induction} - Freq^{2+}_{Background}$$

where

$n^{2+}_{Background}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with medium only (background),

$n^{2+}_{Induction}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with a pool of peptides covering RSVPreF3 (induction),

$N^{CD4}_{Back/Ind}$= Total number of CD4 T cells involved in the assay (background or induction).

For the computation of the fold increase (post- over pre-vaccination) of the frequency of RSVPreF3-specific CD4+ T-cells identified as expressing **at least 2 activation marker(s) including at least one cytokine** among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.the results **below the LLOQ** of the assay will be replaced by the value of the LLOQ.

5.4.3.3. Assessment of long-term persistence of the immune response at Months 18, 24 and 36 after dose 1

The exploratory analysis for modelling the long-term persistence of immune response (humoral and cellular) post dose 1 vaccination will constitute of the following;

- A piece-wise linear mixed model, power law model and modified power law model [David et al, 2009] for repeated measurements (all data available from Day 1, 31 and Month 6, 12, 18, 24, 30, 36 post-Dose 1 vaccination, will be used to model over time
  - the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
  - the neutralizing antibody titers against RSV-A, RSV-B and RSVPreF3-specific IgG antibody concentrations.

The ***piece-wise linear mixed model*** is expressed as follows

The piece-wise model fits the data on three non-overlapping time intervals. This corresponds to the observed decay of humoral and cellular antibodies. Each piece of the model uses a linear function. Three break points, Months x, xx and xxx, will be selected on the basis of Akaike's Information Criterion (AIC) [Pan, 2001].

$$(ft) = \beta_0 + \beta_1 t \qquad \text{if x} \leq \text{t} < \text{xx months}$$

$$(ft) = \beta_0 + \beta_1 t + \beta_2 (t - xx) \quad \text{if xx} \leq \text{t} < \text{xxx months.}$$

$$(ft) = \beta_0 + \beta_1 t + \beta_2 (t - xx) + \beta_3 (t - xxx) \quad \text{if t} \geq \text{xxx months}$$

where $f(t)$ is the log-10 transformed antibody titer/concentration/ RSVPreF3-specific CD4+ T cells at time t post-vaccination, β_0 is the intercept and $\beta_1 (i = 1,2,3)$ are the parameters corresponding to the three time intervals, respectively. All three model effects

will be fitted using the SAS PROC MIXED standard code for linear regression methodology

```
ODS OUTPUT  FitStatistics= stat;
ODS OUTPUT  ConvergenceStatus= conv;
PROC MIXED DATA=kin&i.&j. EMPIRICAL COVTEST NOCLPRINT IC;
CLASS pid;
MODEL log_val = int_m int_m_pw_m&i.*m&i. int_m_pw_m&j.*m&j./s cl
covb outp=pred outpm=predm;
RANDOM intercept/SUBJECT=pid;
MAKE "solutionF" out=est;
RUN;
```

The ***power-law model*** is expressed as follows:

$$f(t) = k - a\log(c + t)$$

where $f(t)$ is the log-10 transformed antibody titer/concentration/ RSVPreF3-specific CD4+ T cells at time t post-vaccination, k is the peak log level, a is the decay rate, and c is an arbitrary small constant (set to zero in this case). The estimated maximum likelihood (ML) values for the model parameters will be obtained using the PROC NLMIXED SAS procedure.

```
PROC NLMIXED DATA=kineticm;
PARMS  se=&se  a0 =&a  k0 =&k  sk=&sk sa=&sa ska=&ska;
q = (1/&time)**a;
lg=&log(q);
mean = k + lg;
MODEL log_val ~ NORMAL(mean,se*se);
RANDOM k a ~ NORMAL([k0,a0], [sk*sk, ska, sa*sa]) SUBJECT=pid out=indiv;
RUN;
```

The ***modified power-law model*** is expressed as follows:

It is an extension of the power law model, and it is expressed as follows.

$$f(t) = k + \log[(1 - \pi)(c + t)^a + \pi]$$

The quantity π (between 0 and 1) represents the proportion of antibodies that are produced in the long term. When the value of π is positive, this implies long-term antibody persistence, as noted by [Fraser et al, 2007]. If π = 0, then the modified power-law model reduces to the standard power-law model. The estimated maximum likelihood (ML) values for the model parameters will be obtained using the PROC NLMIXED SAS procedure.

```
PROC NLMIXED DATA=kineticm ;
PARMS  pi=&pi se=&se  a0 =&a  k0 =&k  sk=&sk sa=&sa ska=&ska;
q = (1-pi)*(1/&time)**a;
lg=&log(q+pi);
mean = k + lg;
MODEL log_val ~ NORMAL(mean,se*se);
RANDOM k a ~ NORMAL([k0,a0], [sk*sk, ska, sa*sa]) SUBJECT=pid out =indiv
RUN;
```

- The three models based on data up to Month 36 will be presented graphically. Models based on data up to Month 18 vs. Month 24, Month 24 vs. Month 30 and Month 30 vs. Month 36 will be presented side by side.

- The above models and SAS codes may be adapted in case of convergence issues.

5.4.3.4. Sensitivity analyses

- In case the COVID-19 pandemic is still ongoing during the study conduct, the following sensitivity analyses might be considered.
  - During the assessment of long-term persistence of the immune response, the models described in Section 5.4.3.3 might be fitted taking into account participants who received the first dose of study vaccine and with full follow-up (attending all study visits).
  - Similarly, a mixed model might be fitted for the evaluation of GMT/GMC based on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

5.4.4. Safety analysis

Primary vaccination and revaccination dose:

The safety analysis will be performed on the ES. A descriptive analysis by group, for the 3 groups pooled up to Month 12, for the RSV_flexible revaccination and RSV_1dose groups pooled vs RSV_annual group for Month 13, Month 18 and Month 24, and by group from Month 25 up to study end, will present a summary of:

- The number and percentage of participants with at least one administration site event (solicited and unsolicited), with at least one systemic event (solicited and unsolicited) and with any AE during the 4-day or 30-day follow-up period will be tabulated with exact 95% CI after each dose. The same computations will be done for Grade 3 AEs, and for Grade 3 non-serious AEs and for AEs resulting in medically attended visit.
- The number and percentage of participants reporting each individual solicited administration site event (any grade, Grade 3 and resulting in medically attended visit) and solicited systemic event (any grade, Grade 3 and resulting in medically attended visit) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- For fever, the number and percentage of participants reporting fever by half degree (°C) cumulative increments during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- The incidence of each solicited administration site event and solicited systemic event (any grade and grade 3) will be represented graphically after each dose.

- The number and percentage of participants with any unsolicited AEs during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) with its exact 95% CI will be tabulated for each dose by group and by Medical Dictionary for Regulatory Activities (MedDRA) Primary System Organ Class (SOC), High Level Term (HLT) and preferred term (PT). Similar tabulation will be done for Grade 3 unsolicited AEs, for any causally related unsolicited AEs, for Grade 3 causally related unsolicited AEs and for unsolicited AEs resulting in a medically attended visit. The verbatim reports of unsolicited AEs will be reviewed by a qualified person and the signs and symptoms will be coded according to the MedDRA Dictionary for Adverse Reaction Terminology. Every verbatim term will be matched with the appropriate Preferred Term.
- The number and percentage of participants with at least one report of SAE classified by the MedDRA Primary SOC, HLT and PTs and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI. The same tabulation will be presented for causally related SAEs and fatal SAEs,.
- The number and percentage of participants with at least one report of causally related SAEs classified by the MedDRA Primary SOC, HLT and PTs will be reported after each dose and from Day 1 up to study end with exact 95% CI. The same tabulation will be presented for fatal SAEs.
- All SAEs will also be described in detail in a tabular listing.
- All AEs/SAEs leading to study/intervention discontinuation from dose 1 up to study end will be tabulated.
- The number and percentage of participants with at least one report of pIMD classified by the MedDRA Primary SOC, HLT and PTs and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI. The same tabulation will be presented for causally related pIMDs
- The number and percentage of participants with at least one causally related pIMD classified by the MedDRA Primary SOC, HLT and PTs will be reported after each dose and from Day 1 up to study end with exact 95% CI.
- All pIMDs will also be described in detail in a tabular listing.
- The analyses of unsolicited AEs will include SAEs (unless otherwise specified).

The analysis of safety will also be performed by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA) and region (North America, Europe, Asia).

5.4.5. Additional considerations

- Compliance in completing solicited events information will be tabulated after each dose .
- The number and percentage of participants with at least one administration site event **(solicited only)**, with at least one systemic event **(solicited only)** and with any **solicited** event during the 4-day follow-up period after each dose will be tabulated with exact 95% CI. The same computations will be done for Grade 3 AEs and for AEs resulting in a medically attended visit.
- The number of days with solicited events reported during the 4-day follow-up period will be tabulated for each solicited event, after each dose using descriptive statistics (mean, min, Q1, median, Q3, maximum).
- The number of each solicited events (any and Grade 3) that are ongoing beyond the 4-day follow-up period will be reported with the duration in days (mean, min, Q1, median, Q3, maximum).
- The list of solicited administration site and systemic events, and definition of intensity are described in Section 10.3.3 and 10.3.9.1 of the protocol.
- The number and percentage of participants with at least one report of SAE classified by the MedDRA Primary SOC, HLT and PTs and reported within 30 days following each vaccination will be tabulated with the exact 95% CI. The same tabulation will be presented for pIMDs.

  The measurement of erythema/swelling (in mm) and fever (in °C/F) will be categorized as follows:

Table 7 Intensity grading scale for solicited events

Grading	Erythema/swelling	Fever
0:	≤ 20 mm	< 38.0°C (100.4°F)
1:	> 20 - ≤ 50 mm	≥ 38.0°C(100.4°F) - ≤ 38.5°C(101.3°F)
2:	> 50 - ≤ 100 mm	> 38.5°C(101.3°F) - ≤ 39.0°C(102.2°F)
3:	> 100 mm	> 39.0°C(102.2°F)

Combined Solicited events and Unsolicited AEs

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

Solicited events will be coded by MedDRA as per the following codes:

Solicited event	Lower level term code	Corresponding Lower level term decode
Pain	10022086	Injection site pain
Erythema	10022061	Injection site erythema
Swelling	10053425	Injection site swelling
Fever	10016558	Fever
Headache	10019211	Headache
Fatigue	10016256	Fatigue
Myalgia	10028411	Myalgia
Arthralgia	10003239	Arthralgia

5.4.5.1. Concomitant Medication

Medications will be coded using the GSKDRUG dictionary.

5.4.6. Sensitivity Analyses

If more than 10% of participants miss the revaccination dose or don't have safety data reported after revaccination, the safety analysis described in section 5.4.4 will also be conducted on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

6. ANALYSIS INTERPRETATION

All analyses are descriptive.

7. INTERIM ANALYSES

7.1. Sequence of analyses

Analyses to evaluate objectives and endpoints will be performed stepwise:

- **Month 6:** A first analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 6 are available for all participants. This analysis will be considered as final for those endpoints. *
- **Month 13**: A second analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 12 are available for all participants, as well as data up to 1 month after the first revaccination (Month 13) in the RSV_annual group. This analysis will be considered as final for those endpoints *.
- **Month 18**: A third analysis will be performed on all safety and/or immunogenicity data available and as clean as possible, when data for secondary endpoints up to

Month 18 are available for all participants. This analysis will be considered as final for those endpoints *. Safety and immunogenicity analysis may be performed at different time depending on data availability.

- **Month 25**: A fourth analysis will be performed on all safety, reactogenicity and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 24 are available for all participants, as well as data up to 1 month after the second revaccination (Month 25) in the RSV_annual group and up to 1 month after the first revaccination (Month 25) in the RSV_flexible revaccination group. This analysis will be considered as final for those endpoints *.
- The final **End of Study** analysis will be performed when all reactogenicity, safety and immunogenicity data for at least primary and secondary endpoints up to study conclusion are available for all participants (Month 36).
- If the data for tertiary endpoints become available at a later stage, (an) additional analysis/analyses will be performed.
- *Each analysis can be considered as final, as it is based on data that is as clean as possible. However, the consecutive analysis of the same time point might slightly differ at the next analysis e.g when M6 data are re-analyzed at the time of M13 analysis. If major changes are identified, they will be described in the clinical study report.

Description	Analysis ID	Disclosure Purpose (CTRS=public posting, SR=study report, internal)
End of study analysis	E1_01	CTRS, SR
Analysis up to Month 6	E1_02	SR
Analysis up to Month 13	E1_03	CTRS, SR
Analysis up to Month 18	E1_04	Internal
Analysis up to Month 25	E1_05	Internal

7.2. Statistical considerations for interim analyses

All analyses will be conducted on final data (as clean as possible) and therefore no statistical adjustment for interim analyses is required.

8. CHANGES TO THE PROTOCOL DEFINED STATISTICAL ANALYSIS

This statistical analysis plan complements the analyses described in the protocol.

The changes compared to the planned statistical analysis specified in the Protocol amendment 1 (Dated: 01 April 2022) are described below:

- The number and percentage of participants with at least one administration site event **(solicited only)**, with at least one systemic event **(solicited only)** and with any **solicited** event during the 4-day follow-up period after vaccination
- Related SAEs/pIMDs and Fatal SAEs will be tabulated after each dose.
- SAEs/pIMDs within 30 days following each vaccination.

9. NON-STANDARD DATA DERIVATION RULES AND STATISTICAL METHODS

The following sections describe additional derivation rules and statistical methods which are not presented in Section 10.1 (Business rules for standard data derivations and statistical methods).

9.1. Handling of missing data

9.1.1. Dates

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

- AE start dates with missing day:
  - If the month is not the same as the vaccine dose, then the imputed start date will be the 1st of the month
  - If the event starts in the same month as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the vaccine dose given during that month. If 'before vaccination' is selected, the imputed date will be one day before the vaccine dose given during that month.
- AE start dates with missing day and month:
  - If the year is not the same as the vaccine dose, then the imputed start date will be the 1st of January.
  - If the event starts in the same year as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the first vaccine dose given during that year. If 'before vaccination' is selected, the imputed date will be one day before the first vaccine dose given during that year.
- AE end dates with missing day: the imputed end date will be the last day of the month (30 or 31) or the study conclusion date whichever comes first.
- AE end dates with missing day and month: the imputed end date will be the last day of the year (31st of December) or the study conclusion date whichever comes first.

All incomplete concomitant medication/vaccination start/end date will follow the rules above.

10. ANNEXES

10.1. Business rules for standard data derivations and statistical methods

This section contains standard rules for data display and derivation for clinical and epidemiological studies.

10.1.1. 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 AE 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 vaccination is selected, the relative dose for the event will be the one administered on the start day of the event. If 'before vaccination is selected, the relative dose for the event will be the dose prior to this one.

10.1.2. Handling of missing data

10.1.2.1. 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 15th
- A missing day and month will be replaced by June 30th.

See also exceptions in Section 9.1.

10.1.2.2. Laboratory data

Refer to Section 5.1.2 for more details.

10.1.2.3. Daily recording of solicited events

10.1.2.3.1. Studies with paper diaries

For studies using paper diaries which have questions in the CRF indicating the presence or absence of solicited events, the following rules are applicable:

- Denominators for the summary of administration site (or systemic) solicited events will be calculated using the number of participants who respond "Yes" or "No" to the question concerning the occurrence of administration site (or systemic) events.

- When a specific solicited event is marked as having not occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=N for the specified post-dose period for the event in question), all daily measurements will be imputed as Grade 0.
- When a specific solicited event is marked as having occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=Y for the specified post-dose period for the event in question), any missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.
- When the occurrence of a specific solicited event is not present (i.e. SDTM CE.CEOCCUR is neither Y nor N for the specified post-dose period for the event in question) but the group of solicited events (administration site or systemic) is marked as having occurred (i.e. SDTM CE.CEOCCUR=Y), all missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.

The following table shows how participants contribute to each category for a specific solicited event over the Day X to Day Y post-dose period:

Solicited event category	Participants included in the calculation of the numerator
Any	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y or with the adverse event marked as present and at least one missing daily recording between Day X and Day Y
At least grade 1	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y
At least grade 2	All participants with at least one occurrence of the adverse event at grade 2 or grade 3 between Day X and Day Y
At least grade 3	All participants with at least one occurrence of the adverse event at grade 3 between Day X and Day Y

10.1.2.4. Unsolicited adverse events

Unsolicited AE summaries are including SAEs unless specified otherwise.

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

10.1.3. Data derivation

10.1.3.1. 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

In this study, we will collect only the year of birth. The rules for handling missing day and/or month in the DOB are given in Section 10.1.2.1.

Since only the year of birth will be collected in the eCRF, there might be some discrepancies between the age computed using standard derivation rules and the age category used in SBIR for the minimization.

Therefore, for the analysis by age, the age categories defined in Table 4 will be determined according to the information entered in SBIR (variable AGEGRP in SDTM SUPPDM), except for "≥65 YOA" category which will be obtained using the derived age because this category is not used in SBIR for minimization

10.1.3.2. 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

10.1.3.3. Numerical serology results

Numerical serology results will be derived from the content of IS.ISORRES in the SDTM dataset. For humoral 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 and value is <=ULOQ	value
"value" and value is > ULOQ	ULOQ*
All other cases	Missing

*This rule will be used to compute descriptive statistics (GMTs, fold increase etc).

10.1.3.4. Geometric mean titers (GMTs) and concentrations (GMCs)

GMT or GMC calculations are performed by taking the inverse logarithm of the mean of the log titer or concentration transformations. Non quantifiable antibody titers or concentrations will be converted as described in Section 10.1.3.3 for the purpose of GMT/GMC calculation. Cut-off values are defined by the laboratory before the analysis.

10.1.3.5. 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).

10.1.3.6. 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 3 March 2018 and ends on 12 March 2018 has a duration of 10 days.

The duration of solicited events will be calculated as the sum of the individual days with the event reported at grade 1 or higher or reported as missing ("NOT DONE"), during the solicited event period. The duration of solicited events ongoing beyond the solicited period will be calculated as end date – start date + 1. The goal of this change was to align this text with the standard programs.

10.1.3.7. Counting rules for combining solicited and unsolicited AEs

For output combining solicited and unsolicited AEs, all SAEs will be considered systemic events since the administration site flag is not included in the Expedited Adverse Event CRF pages. Unsolicited AEs with missing administration site flag will also be considered systemic.

Multiple events with the same PT which start on the same day are counted as only one occurrence.

In case a solicited event is worsening to Grade 3 after the solicited period, it should be reported only in the following table.

Summary of grade 3 AEs (solicited and unsolicited) within 30 days following each dose and overall

10.1.3.8. 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.

10.1.4. Display of decimals

10.1.4.1. Percentages

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

10.1.4.2. Demographic/baseline characteristics statistics

The mean, median, and SD for continuous baseline characteristics 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 maximum and minimum of transformed body temperatures will be displayed with one decimal.

10.1.4.3. Serological summary statistics

For each assay, GMT or GMC and their confidence limits will be presented with one decimal, as well as GMT/GMC fold increase from pre-dose.

The mean, median, SD and quartile values for frequency of RSVPreF3-specific-CD4+ and CD8+ T cells and for the fold increase (post- over pre-vaccination) will be presented with one decimal. The minimum and maximum values will be presented with no decimal.

10.1.5. Statistical methodology

10.1.5.1. Exact confidence intervals around proportions

The exact confidence intervals around within-group proportions are derived using the method of Clopper and Pearson [Clopper, 1934].

11. 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.

12. REFERENCES

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

David M-P, Van Herck K, Hardt K, Tibaldi F, Dubin G, Descamps D, et al. Long-term persistence of anti-HPV-16 and-18 antibodies induced by vaccination with the AS04-adjuvanted cervical cancer vaccine: modeling of sustained antibody responses. GYNECOLOGIC ONCOLOGY. 2009;115(3):S1–6.

Fraser C, Tomassini JE, Xi L, Golm G, Watson M, Giuliano AR, et al. Modeling the long-term antibody response of a human papillomavirus (HPV) virus-like particle (VLP) type 16 prophylactic vaccine. Vaccine 2007 May 22;25(21):4324-33

Pan W Akaike's information criterion in generalized estimating equations Biometrics, 57 (1) (2001), pp. 120-125

Rubin, D. B., Stern, H. S. AND Vehovar, V. Handling “don’t know” survey responses: the case of the Slovenian plebiscite. Journal of the American Statistical Association 90, 1995, 822–828

Statistical Analysis Plan

Title:	A phase 3, randomized, open-label, multi-country study to evaluate the immunogenicity, safety, reactogenicity and persistence of a single dose of the RSVPreF3 OA investigational vaccine and different revaccination schedules in adults aged 60 years and above.
eTrack study number and Abbreviated Title	212496 (RSV OA=ADJ-004)
Scope:	All data pertaining to the above study except the Safety Review Team analyses for the first 50 participants aged 80 years and above
Date of Statistical Analysis Plan	Amendment 1 Final: 04 Mar 2022

APP 9000058193 Statistical Analysis Plan Template V5 (Effective date: 1July2020)

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LIST OF ABBREVIATIONS

AE	Adverse event
AESI	Adverse Events of Special Interest
ANCOVA	Analysis of Covariance
CD	Community Dwelling
CI	Confidence Interval
COVID-19	Coronavirus Disease 2019
CRF	Case Report Form
CTRS	Clinical Trial Registry Summary
Eli Type	Internal database code for type of elimination code
ELISA	Enzyme-linked immunosorbent assay
ELU/mL	ELISA unit per milliliter
ES	Exposed Set
GMC	Geometric mean antibody concentration
GMT	Geometric mean antibody titre
GSK	GlaxoSmithKline
IU/mL	International units per milliliter
LL	Lower Limit of the confidence interval
LTCF	Long Term Care Facility
MedDRA	Medical Dictionary for Regulatory Activities
NA	Not Applicable
PD	Protocol Deviation
PPS	Per-Protocol Set
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SBIR	GSK Biologicals Internet Randomization System
SD	Standard Deviation
SDTM	Study Data Tabulation Model
UL	Upper Limit of the confidence interval

1. DOCUMENT HISTORY

Date	Description	Protocol Version
15 February 2021 04 Mar 2022	First version Amendment 1: • The percentage has been changed to 5% in this statement: If in any study group the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 5%, a second analysis will be performed on the Exposed Set (section 4) • The definition of randomized set and elimination code 1010 were deleted (section 4.2.1) • Changes in the demography and baseline characteristics (section 5.2) – Addition of demography analysis by sex and region – Clarify that study intervention administered will also be tabulated by visit – Deletion of participant disposition analysis in the ES and PPS by age category. • Changes in the immunogenicity analysis section 5.4.1 and section 5.4.2 – Deletion of some analyses • Changes in the safety analysis (section 5.4.1 and section 5.4.2) – Deletion of some analyses – Unsolicited AEs, SAEs, pIMDs to also be presented by High Level Term (HLT) – Solicited events (any and Grade 3) that are ongoing beyond the 4-day follow-up period to be considered – Safety analysis to be presented also by region • Additional details about persistence modelling have been added (section 5.4.3.3)	Final: 28 October 2020 Final: 28 October 2020

2. OBJECTIVES/ENDPOINTS

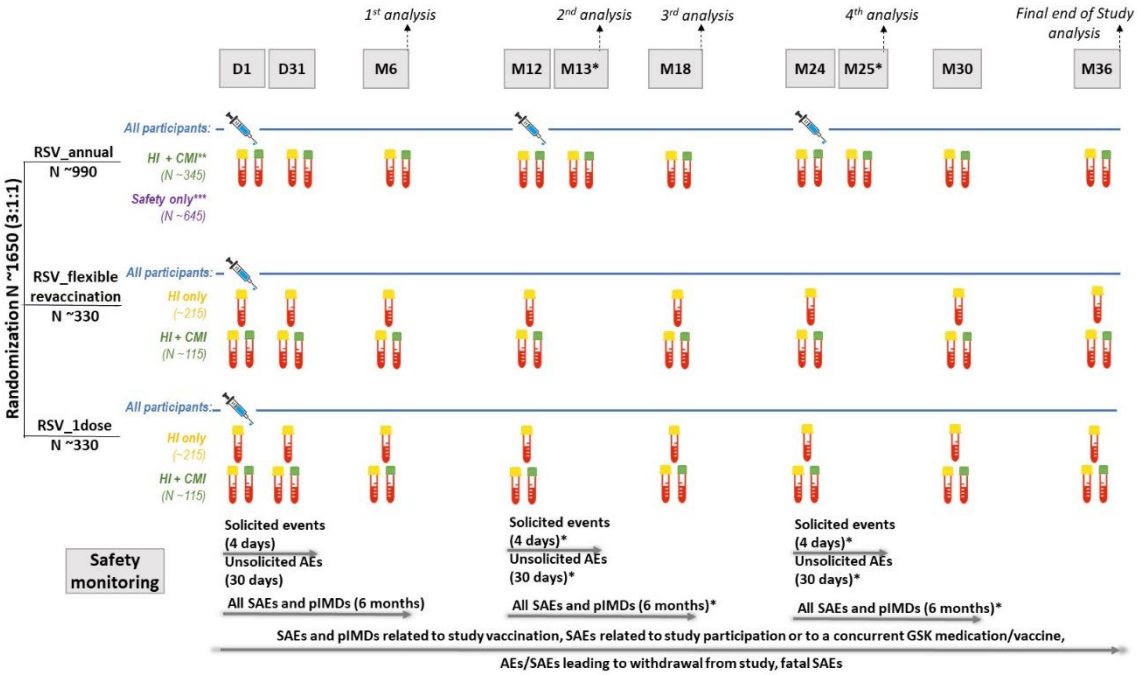
Objectives	Endpoints
Primary	
To evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • Neutralizing antibody titers against RSV-A • Neutralizing antibody titers against RSV-B.
Secondary	
To further evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • RSVPreF3-specific Immunoglobulin G (IgG) antibody concentrations.
To evaluate the humoral immune response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses, up to study end.	Humoral immune response at Months 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Neutralizing antibody titers against RSV-A and RSV-B • RSVPreF3-specific IgG antibody concentrations.
To evaluate the cell-mediated immune (CMI) response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses up to study end.	CMI response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), at Months 6, 12, 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
To evaluate the safety and reactogenicity of each vaccination schedule of the RSVPreF3 OA investigational vaccine in all participants.	• Occurrence of each solicited administration-site and systemic event during a 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) after each vaccination. • Occurrence of any unsolicited AE during a 30-day follow up period (i.e., on the day of vaccination and 29 subsequent days) after each vaccination. • Occurrence of all SAEs and pIMDs up to 6 months after each vaccination. • Occurrence of fatal SAEs, related SAEs and related pIMDs from first vaccination (Day 1) up to study end (Month 36).
Tertiary	
To further characterize immune responses to the RSVPreF3 OA investigational vaccine.	• Any further exploratory immunology in a subset of participants, such as, but not limited to: – Antibodies against specific protein F epitopes. – Potential new immunological markers for protection. – Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing one or any combination of immune marker(s)

CMI = cell-mediated immunity, AE = adverse event, SAE = serious adverse event, pIMD = potential immune-mediated disease.

3. STUDY DESIGN

3.1. Overall design

Figure 1 Study design overview



N = number of participants; D = Day; M = Month; AE = adverse event; HI = humoral immunity; CMI = cell-mediated immunity; pIMD = potential immune-mediated disease; SAE = serious adverse event.

= blood sample for humoral immune responses (only applicable for the 345 participants of the RSV_annual group; and for all participants of the RSV_flexible revaccination and RSV_1dose groups)

= blood sample for CMI (only applicable for the ~345 participants of the RSV_annual group, and ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups).

Note: For group RSV_annual, Revaccination Year 1 = Month 12; Revaccination Year 2 = Month 24.

* Visit and follow-up of solicited events, unsolicited AEs, SAEs and pIMDs only applicable for RSV_annual group

** For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws, and will only be followed up for safety and reactogenicity.

***For those participants in the RSV_annual group without blood sample collections, visits at Months 6, 18, 30 and 36 may be through a phone contact. However, phone contact should be attempted only if site visit is not possible at Months 6, 18, 30 and 36.

- **Type of study**: self-contained.
- **Experimental design**: Phase 3, randomized, open-label, multi-country study with 3 parallel groups (see Figure 1).
- **Duration of the study**: ~ 36 months for each participant.
- **Primary completion date**: Month 12.
- **Control**: none.
- **Blinding**: open-label. Refer to the protocol for details.

- **Data collection**: standardized electronic Case Report Form (eCRF). Solicited events and unsolicited AEs will be collected using a participant Diary card (paper Diary card).
- **Safety monitoring**: the study will be conducted with oversight by the project Safety Review Team (SRT). Please refer to protocol for the description of review of safety data by the SRT.
- **Study groups**: refer to Figure 1 and Table 1 for an overview of the study groups

Table 1 Study groups, intervention and blinding foreseen in the study

Study Groups	Number of participants	Age	Intervention			Blinding
			Primary vaccination	Revaccination Year 1 (Month 12)	Revaccination Year 2 (Month 24)	Open
RSV_annual*	~990	≥60 years	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	X
RSV_flexible revaccination**	~330	≥60 years	RSVPreF3 OA investigational vaccine			X
RSV_1dose	~330	≥60 years	RSVPreF3 OA investigational vaccine	(none)	(none)	X

* RSV_annual group will be split in 3 technical groups in SBIR in order to have a randomization ratio of 1:1:1:1:1: for treatment allocation and avoid predictability.
**Based on immunogenicity data from this study and efficacy results from the Phase 3 study RSV OA=ADJ-006, a revaccination might be decided for this group.

- **Groups and sub-groups definition for analysis:** Refer to the tables below for a description of the groups and subgroups labels that will be used in the Tables Figures and Listings (TFLs).

  For some of the analyses up to 12 months post dose 1, the 3 study groups will be pooled, and from Month 13, the RSV_flexible revaccination and RSV_1dose groups will be pooled, at applicable timepoints. The following group names will be used in the TFLs:

Table 2 Group names and definition for footnote in the TFLs

Group order in tables	Group label in tables	Group definition for footnote
1	RSV_annual	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1, followed by a revaccination dose at 12 months post-Dose 1 and at 24 months post-Dose 1
2	RSV_flexible revaccination	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1 and a revaccination dose will be given whenever a revaccination would be needed
3	RSV_1dose	Participants receiving a single dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1

Table 3 Pooled group names and definition for footnote in the TFLs

Pooled Group label in tables	Groups to be pooled	Pooled definition for footnote	Comment
Total	RSV_annual, RSV_flexible revaccination and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine in all three groups	Applicable on data reported up to Month 12
Flexible+1dose	RSV_flexible revaccination, and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine in the RSV_flexible revaccination, and RSV_1dose groups	Applicable on data reported from Month 13

Table 4 Sub-group names and definitions for footnote in the TFLs

Sub-analysis	Subgroup order in tables	Subgroup label in tables	Subgroup definition for footnote
By age	1	≥65YOA	≥65 years old participants
	2	≥70YOA	≥70 years old participants
	3	≥80YOA	≥80 years old participants
	4	60-69YOA	60-69 years old participants
	5	70-79YOA	70-79 years old participants
By sex	1	Female	Female
	2	Male	Male
By region	1	North America	Participants from North America (United States of America)
	2	Europe	Participants from Europe (Finland, Germany)
	3	Asia	Participants from Asia (Japan, Taiwan)

YOA = Years of age

- **Allocation of participants to assay subsets**

Allocation of participants to assay subsets will be performed using SBIR. The subsets are detailed below.

	RSV_annual	RSV_flexible revaccination	RSV_1dose
HI subset	~345*	All participants (~330)	All participants (~330)
CMI subset	~345*	~115	~115

*For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws, and will only be followed up for safety and reactogenicity.

- **HI subset:** ~345 participants from the RSV_annual group, and all participants from the RSV_flexible revaccination and RSV_1dose groups. These participants will have blood samples collected for testing of humoral immunity at each visit applicable for their study group.
- **CMI subset**: ~345 participants from the RSV_annual group, and ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups. These participants will have additional blood samples collected for CMI testing at each visit applicable for their study group.

4. ANALYSIS SETS

4.1. Definition

Table 5 Populations for analyses

Analysis set	Description
Enrolled set	Participants who agreed to participate in a clinical study after **completion of the informed consent process.**
Exposed set (ES)	All participants who received at least 1 dose of the study intervention. The allocation in a group is done in function of the administered intervention.
Per Protocol set (PPS)	All participants who received at least 1 dose of the study intervention to which they are randomized and have post-vaccination data, minus participants with protocol deviations that lead to exclusion.

The primary analysis for immunogenicity will be performed on the Per Protocol set (PPS). If in any study group the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 5%, a second analysis will be performed on the Exposed Set (ES). The immunogenicity analysis will be performed overall, by sex, by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and by region (North America, Europe, Asia).

4.2. Criteria for eliminating data from Analysis Sets

4.2.1. Elimination from Exposed Set (ES)

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

4.2.2. Elimination from Per-protocol analysis Set (PPS)

4.2.2.1. Excluded participants

A participant will be excluded from the populations of analysis under the following conditions

- For codes 800, 900, 1030 and 1050: participants will be eliminated for all visits
- For codes 1040, 1070, 1080, 1090, 2010, 2040, 2050, 2080: participants will be eliminated from a specific visit (at which the condition is met) onwards.
- For codes 2090, 2100, 2120, 2130: participants will be eliminated at the specific visit at which the condition is met.

Table 6 List of elimination codes

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
800	Fraudulent data	All	ES and PPS
900	Invalid informed consent	All	ES and PPS
1030	Study intervention not administered at all	All	ES and PPS
1040	Administration of concomitant vaccine(s) forbidden in the protocol • Any investigational or non-registered vaccine other than the study vaccine used during the study period beginning 30 days before the first dose of study vaccine, or planned use during the study period. • Planned or actual administration of a vaccine not foreseen by the study protocol in the period starting 30 days before each dose and ending 30 days after each dose of study vaccine administration, with the exception of inactivated, split virion and subunit influenza vaccines which can be administered up to 14 days before or from 14 days after each study vaccination. • Previous vaccination with an RSV vaccine	All	PPS
1050	Randomization failure	Day 1	PPS
1070	Vaccine administration not according to protocol • Incomplete vaccination course • Participant was vaccinated with the correct vaccine but containing a lower volume	Vaccination visits at Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group only	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Participant was re-vaccinated while it was not planned • Route of the study intervention is not intramuscular • Wrong reconstitution of administered vaccine		
1080	Vaccine administration after a Temperature deviation • Vaccine administered despite a Good Manufacturing Practices (GMP) no-go temperature deviation	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group only	PPS
1090	Vaccine administration after expiration	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group only	PPS
2010	Protocol deviation linked to inclusion/exclusion criteria	All	PPS
2040	Administration of any medication forbidden by the protocol • Any investigational or non-registered medication used during the study period • Administration of long-acting immune-modifying drugs at any time during the study period (e.g. *infliximab*) • Immunoglobulins and/or any blood products administered during the study period • Chronic administration (defined as more than 14 consecutive days in total) of immunosuppressants or other immune-modifying drugs during the period starting 90 days prior to the study	All	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	vaccine administration or planned administration during the study period. For corticosteroids, this will mean prednisone ≥20 mg/day, or equivalent.		
2050	Intercurrent medical condition • Participants may be eliminated from the PPS for immunogenicity if, during the study, they incur a condition that has the capability of altering their immune response (intercurrent medical condition) or are confirmed to have an alteration of their initial immune status.	All	PPS
2080	Participants did not comply with dosing schedule • **RSV_annual**: number of days between dose 1 and revaccination dose at Month 12 is outside [350-380 days] • **RSV_annual**: number of days between dose 1 and revaccination dose at Month 24 is outside [715-745 days]	Month 12 and 24	PPS
2090	Participants did not comply with blood sample schedule • Number of days between dose 1 and Day 31 blood sample is outside [30-42 days] • Number of days between dose 1 and Month 6 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 12 blood sample is outside [350-380 days] • Number of days between Month 12 and Month 18 blood	Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	sample is outside [180-210 days] • Number of days between dose 1 and Month 24 blood sample is outside [715-745 days] • Number of days between Month 24 and Month 30 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 36 blood sample is outside [1080-1110 days] *For RSV_annual group only:* • Number of days between revaccination at Month 12 and Month 13 blood sample is outside [30-42 days] • Number of days between revaccination at Month 24 and Month 25 blood sample is outside [30-42 days]		
2600*	Participants not included in the humoral immunogenicity subset	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS
2700*	Participants not included in the cell-mediated immune response subset	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS
2100	Serological results not available post-vaccination • No immunological result at visit x for all the following tests: RSV A/B Neutralizing antibody titer, RSVPreF3-specific IgG antibody concentration and RSVPreF3-specific CD4+/CD8+ T cells frequency	Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36 Month 13 and 25 are applicable to the RSV_annual group only	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
2120	Obvious incoherence or abnormality or error in laboratory data • Unreliable released data as a result of confirmed sample mismatch or confirmed inappropriate sample handling at laboratory	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

* codes 2600 and 2700 are not considered as protocol deviations, but those codes will be used to identify participants that are not part of the immunogenicity subsets.

5. STATISTICAL ANALYSES

Standard data derivation rules and stat methods are described in section 10.1 while the study specific data derivation rules and stat methods are described in section 9.

For the sake of analysis, data up to Month 12 will be analyzed by group (RSV_annual, RSV_flexible revaccination and RSV_1dose), and also the 3 groups will be pooled. From Month 13, the analysis will be tabulated by group, and also for RSV_annual versus the pooled RSV_flexible revaccination and RSV_1dose groups, at applicable time points.

If more than 5% of participants in the RSV_annual group received only 1 dose or only 2 doses, an analysis by subgroup according to the number of dose(s) administered will be performed. The goal of this analysis is to describe the characteristics of participants in the RSV_annual group who ultimately receive different numbers of revaccination doses in order to rule out the possibility of systematic differences between these subgroups.

5.1. General considerations

5.1.1. Demography

For a given participant and a given demographic variable, missing measurements will not be replaced.

5.1.2. Immunogenicity

- Any missing or non-evaluable immunogenicity measurement will not be replaced. The descriptive analysis performed for each assay at each time point will exclude participants with a missing or non-evaluable measurement. This is applicable to the standard way of computing GMTs/GMCs.
- During the computation of GMTs/GMCs from the mixed effects model (section 5.4.3.1). Multiple imputation (MI) technique will not be applied for any missing immunogenicity measurement. This is because the mixed effects model inherently

accounts for the missingness, under the assumption that the missing data are missing at random (MAR). Thus considering that missing data are MAR is a reasonable assumption for this descriptive clinical trial, then it is not necessary to perform MI [Rubin et al., 1995]

- The geometric mean titers/concentrations (GMTs/GMCs) will be computed by taking the anti-logarithm of the arithmetic mean of the $\log_{10}$ transformed titers/concentrations.
- 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.
- Antibody titers/concentrations below the assay cut-off will be given an arbitrary value of half the assay cut-off for the purpose of GMT/GMC calculation.
- Antibody titers/concentrations above the Upper Limit of Quantification (ULOQ) value will be given the ULOQ value for the purpose of GMT/GMC calculation.
- The mean geometric increase (MGI) is calculated by the geometric mean of ratios of antibody titers/concentrations of each post-primary vaccination time point over pre-primary vaccination (Day 1).

5.1.3. Reactogenicity/Safety

- For a given participant and the analysis of solicited events within 4 days post-vaccination, missing or non-evaluable measurements will not be replaced. Therefore, the analysis of solicited events will include only vaccinated participants with documented solicited safety data (i.e., paper diary completed).
- For analysis of unsolicited AEs, SAEs, pIMDs and concomitant medications, all vaccinated participants will be considered. Participants who did not report an event or concomitant medication will be considered as participants without the event or the concomitant medication, respectively.

5.2. Analysis of demography and baseline characteristics

5.2.1. Analysis planned in the protocol

Descriptive summaries will be performed for each group (RSV_annual, RSV_flexible revaccination and RSV_1dose) and overall. The same will be performed in each subset (humoral immune subset and CMI subset).

Demographic/baseline characteristics (age at vaccination in years, sex, race, ethnicity, type of residence (CD/LTCF) and smoking status) will be summarized using descriptive statistics:

- Frequency tables will be generated for categorical variables such as race.
- Mean, median, standard deviation and range will be provided for continuous data such as age.

The distribution of participants will be tabulated as a whole and per group, for each age category, and for each subset.

The number of doses of the study intervention administered will be tabulated by group and by visit.

Withdrawal status will be summarized by group using descriptive statistics:

- The number of participants enrolled into the study as well as the number of participants excluded from Per Protocol (PP) analyses will be tabulated.
- The numbers of participants withdrawn from the study will be tabulated according to the reason for withdrawal.

Participant disposition in the ES and PPS will be reported as a whole and per group

The number and percentage of participants using concomitant medication (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) and during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) will be summarized by group after each vaccine dose and overall, and after the revaccination.

5.2.2. Additional considerations

- The analysis of demographic characteristics by group will be performed on the ES and on the PPS.
- Demography and baseline characteristics will also be summarized by sex and region

5.3. Primary endpoint(s)

5.3.1. Analysis planned in the protocol

The primary analysis for immunogenicity will be performed on the Per Protocol set (PPS). If in any study group the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 5%, a second analysis will be performed on the Exposed Set (ES).

5.3.1.1. Humoral immune response – up to Month 12

The analysis of the primary vaccination time points up to Month 12 will be performed for each group and for the 3 groups pooled. For each time point with blood sample collection for humoral immune response up to Month 12 and for each assay (RSV-A/-B neutralization assays), unless otherwise specified, the following analyses will be performed:

Within groups evaluation:

- Percentage of participants with antibody titers/concentrations above a positivity cut-off and their exact 95% confidence interval (CI) will be tabulated.
- GMTs/GMCs and their 95% CI will be tabulated and represented graphically. Furthermore, to account for all the time points at which the blood samples are collected, a mixed effects model (see section 5.4.3.1) will be fitted, from which the GMTs/GMCs will be computed.
- Antibody titers/concentrations will be displayed using reverse cumulative curves.
- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI.

5.4. Secondary endpoint(s)

5.4.1. Humoral immune response

The same analyses as described above for the primary endpoint will be performed for PreF3 IgG ELISA up to Month 12 and for each time point with blood sample collection for humoral immune response **from study Month 13 up to study end** and for each assay (preF3 IgG **ELISA and RSV-A/-B neutralization assays**). The analysis of the post-primary vaccination time points from study Month 13 up to study end will be performed for each group and for the pooled RSV_flexible revaccination and RSV_1dose groups.

Only the analyses requiring further clarification of the exact time points analysed are summarized below:

Within groups evaluation:

- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI:
  - For each post-primary vaccination time point (at Months 18, 24, 30 and 36 when applicable) over pre-primary vaccination (Day 1),
  - For each post-revaccination time point over corresponding pre-revaccination (Months 12/24) in the RSV_annual group,
  - For 1 month post-revaccination at Month 12 (Month 13) over 1 month post-Dose 1 vaccination (Day 31) in RSV_annual group.
  - For 1 month post-revaccination at Month 24 (Month 25) over 1 month post-revaccination at Month 12 (Month 13) in RSV_annual group

In addition, the following evaluation will be performed:

- The kinetics of GMTs/GMCs will be plotted as a function of time for participants with results available at all post-primary vaccination time points.

The immunogenicity analysis will be performed overall, by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA), and region (North America, Europe, Asia).

5.4.2. Cell-mediated immune response

Within groups evaluation:

The following parameters will be summarized by group at each time point for which blood samples are collected, for the 3 groups pooled for time points up to study Month 12, for the pooled RSV_flexible revaccination and RSV_1dose up to study end using descriptive statistics (N, geometric mean, min, Q1, median, Q3, max) in the CMI subset:

- Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17, measured by intracellular cytokine staining (ICS) using peripheral blood mononuclear cells PBMCs.
- The kinetics of RSVPreF3-specific CD4+ T cells frequencies will be plotted as a function of time for participants with results available at all primary vaccination time points.
- Fold increase of the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17, measured by ICS at each post-primary vaccination time point over pre-vaccination (Day 1) by group, at each post-revaccination over the corresponding pre-revaccination and at one month post-revaccination over 1 month post-previous dose in the RSV_annual group.

The immunogenicity analysis will be performed overall, by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and region (North America, Europe, Asia).

5.4.3. Additional considerations

5.4.3.1. Mixed effects model

For the GMTs/GMCs calculation based on the mixed effects model mentioned in section 5.3.1.1, repeated measures analysis of covariance (ANCOVA) model will be fitted including age category (60-69 YOA , 70-79 YOA and ≥80 YOA,), study group (RSV_annual, RSV_flexible revaccination and RSV_1dose), sex (male and female), visit (Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36) , as fixed effects, pre-vaccination log10-transformed titers/concentrations as a covariate and the response variable is the post-vaccination log10-transformed titers/concentrations. The PROC MIXED procedure in SAS® will be used to carry out the ANCOVA.

The following SAS codes will be used:

For the analysis by study group, study group and visit will be included as the fixed effects in the model

```
Analysis by group
PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid group visit ;
MODEL post-vac=pre-vacc group visit visit*group/ noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=unsubject=subjid;
LSMEANS visit*group/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by age category, age category and visit will be included as the fixed effects in the model. However, this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, age category and visit.

```
Analysis by age-category

PROC MIXED DATA=SERO method=reml empirical ;
 CLASS subjid visit  agecat ;
MODEL post-vac=pre-vacc agecat visit visit* agecat /noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
 REPEATED visit/type=un subject=subjid;
 LSMEANS visit*agecat/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by sex, sex and visit will be included as the fixed effects in the model. However, this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, sex and visit.

```
Analysis by sex

PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid visit  agecat ;
MODEL post-vac=pre-vacc sex visit visit* sex / noint s cl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*sex/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

For the analysis by region, region and visit will be included as the fixed effects in the model. However this is only applicable at timepoints where the 3 study groups will be pooled. Otherwise for the timepoints where pooling of the 3 groups is not possible, the fixed effects will include study group, region and visit.

```
Analysis by region

PROC MIXED DATA=SERO method=reml empirical;
CLASS subjid visitregion ;
MODEL post-vac=pre-vacc region visit visit* region / noint scl;
RANDOM intercept/ subject=usubjid g v vcorr type = un;
REPEATED visit/type=un subject=subjid;
LSMEANS visit*region/ E cl;
ODS OUTPUT LSMEANS=LS;
RUN;
```

The Geometric mean antibody titers/concentrations, lower and upper confidence limits are obtained from the following data step:

```
DATA LS;
   SET LS;
   gm=10**estimate;
   ll=10**lower;
   ul=10**upper;
 RUN;
```

The above SAS codes may be adapted in case of convergence issues.

5.4.3.2. Cell-mediated immune response

Within groups evaluation

- The frequency of RSVPreF3 specific-CD4+ T cells will be displayed graphically using boxplots (min, Q1, median, Q3, max), by group and timepoint.
- The RSVPreF3-specific CD4+/ CD8+ T cell frequencies will be obtained by subtracting the background value to the antigen-induced value, and by setting to 1 all values less than or equal to zero for geomean calculation and graphical representation. Frequencies will be expressed as the number of cells per million of CD4+ T cells.
- More specifically, the frequencies of RSVPreF3-specific CD4+/ CD8+ T cells expressing at least 2 activation markers including at least one cytokine [$Freq^{2+}$] will be computed as follows:

$$Freq^{2+}_{Background} = \frac{n^{2+}_{background}}{N^{CD4}_{Background}} \quad \text{and} \quad Freq^{2+}_{Induction} = \frac{n^{2+}_{Induction}}{N^{CD4}_{Induction}},$$

And

$$Freq^{2+}_{Specific} = Freq^{2+}_{Induction} - Freq^{2+}_{Background}$$

where

$n^{2+}_{Background}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with medium only (background)

$n^{2+}_{Induction}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with a pool of peptides covering RSVPreF3 (induction)

$N^{CD4}_{Back/Ind}$= Total number of CD4 T cells involved in the assay (background or induction)

For the computation of the fold increase (post over pre-vaccination) of the frequency of RSVPreF3-specific CD4+ T-cells identified as expressing **at least 2 marker(s)** among IL-2, CD40L, TNF-α, IFN-γ, the results **below the LLOQ** of the assay will be replaced by the value of the LLOQ.

5.4.3.3. Assessment of long-term persistence of the immune response at Months 18, 24 and 36 after dose 1.

The exploratory analysis for modelling the long-term persistence of immune response (humoral and cellular) post dose 1 vaccination will constitute of the following;

- A piece-wise linear mixed model, power law model and modified power law model [David et al, 2009] for repeated measurements (all data available from Day 1, 31 and Month 6, 12, 18, 24, 30, 36 post dose 1 vaccination, will be used to model over time
  - the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
  - the neutralizing antibody titers against RSV-A, RSV-B and RSVPreF3-specific IgG antibody concentrations.

The ***piece-wise linear mixed model*** is expressed as follows

The piece-wise model fits the data on three non-overlapping time intervals. This corresponds to the observed decay of humoral and cellular antibodies. Each piece of the model uses a linear function. Three break points, Months x, xx and xxx, will be selected on the basis of Akaike's Information Criterion (AIC) [Pan, 2001].

$$(ft) = \beta_0 + \beta_1 t \qquad \text{if x} \leq \text{t} < \text{xx months}$$

$$(ft) = \beta_0 + \beta_1 t + \beta_2 (t - xx) \quad \text{if xx} \leq \text{t} < \text{xxx months.}$$

$$(ft) = \beta_0 + \beta_1 t + \beta_2 (t - xx) + \beta_3 (t - xxx) \quad \text{if t} \geq \text{xxx months}$$

where $f(t)$ is the log antibody titer/concentration/ RSVPreF3-specific CD4+ T cells at time t post-vaccination, β_0 is the intercept and $\beta_1 (i = 1,2,3)$ are the parameters corresponding to the three time intervals, respectively. All three model effects will be fitted using the SAS PROC MIXED standard code for linear regression methodology

```
ODS OUTPUT  FitStatistics= stat;
ODS OUTPUT  ConvergenceStatus= conv;
PROC MIXED DATA=kin&i.&j. EMPIRICAL COVTEST NOCLPRINT IC;
CLASS pid;
MODEL log_val = int_m int_m_pw_m&i.*m&i. int_m_pw_m&j.*m&j./s cl
covb outp=pred outpm=predm;
RANDOM intercept/SUBJECT=pid;
MAKE "solutionF" out=est;
RUN;
```

The ***power-law model*** is expressed as follows:

$$f(t) = k - a\log(c + t)$$

where $f(t)$ is the log antibody titer/concentration/ RSVPreF3-specific CD4+ T cells at time t post-vaccination, k is the peak log level, a is the decay rate, and c is an arbitrary small constant (set to zero in this case). The estimated maximum likelihood (ML) values for the model parameters will be obtained using the PROC NLMIXED SAS procedure.

```
PROC NLMIXED DATA=kineticm;
PARMS  se=&se  a0 =&a  k0 =&k  sk=&sk sa=&sa ska=&ska;
q = (1/&time)**a;
lg=&log(q);
mean = k + lg;
MODEL log_val ~ NORMAL(mean,se*se);
RANDOM k a ~ NORMAL([k0,a0], [sk*sk, ska, sa*sa]) SUBJECT=pid out=indiv;
RUN;
```

The ***modified power-law model*** is expressed as follows:

It is an extension of the power law model, and it is expressed as follows.

$$f(t) = k + \log[(1 - \pi)(c + t)^a + \pi]$$

The quantity π (between 0 and 1) represents the proportion of antibodies that are produced in the long term. When the value of π is positive, this implies long-term antibody persistence, as noted by[Fraser et al, 2007]. If π = 0, then the modified power-law model reduces to the standard power-law model. The estimated maximum likelihood (ML) values for the model parameters will be obtained using the PROC NLMIXED SAS procedure.

```
PROC NLMIXED DATA=kineticm ;
PARMS  pi=&pi se=&se  a0 =&a  k0 =&k  sk=&sk sa=&sa ska=&ska;
q = (1-pi)*(1/&time)**a;
lg=&log(q+pi);
mean = k + lg;
MODEL log_val ~ NORMAL(mean,se*se);
RANDOM k a ~ NORMAL([k0,a0], [sk*sk, ska, sa*sa]) SUBJECT=pid out =indiv
RUN;
```

- The three models based on data up to M36 will be presented graphically. Models based on data up to M18 vs M24, M24 vs M30 and M30 vs M36 will be presented side by side.

- The above models and SAS codes may be adapted in case of convergence issues.

5.4.3.4. Sensitivity analyses

- In case the COVID-19 pandemic is still ongoing during the study conduct, the following sensitivity analyses might be considered.
  - During the assessment of long-term persistence of the immune response, the models described in section 5.4.3.3 might be fitted taking into account; Participants who received the first dose of study vaccine and with full follow-up (attending all study visits)
  - Similarly, a mixed model will be fitted for the evaluation of GMT/GMC based on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

5.4.4. Safety analysis

Primary vaccination and revaccination dose:

The safety analysis will be performed on the ES. A descriptive analysis by group, for the 3 groups pooled up to Month 12, for the RSV_flexible revaccination and RSV_1dose groups pooled vs RSV_annual group from study Month 13 up to study end will present a summary of:

- The number and percentage of participants with at least one administration site event (solicited and unsolicited), with at least one systemic event (solicited and unsolicited) and with any AE during the 4-day or 30-day follow-up period will be tabulated with exact 95% CI after each dose. The same computations will be done for Grade 3 AEs, and for Grade 3 non-serious AEs and for AEs resulting in medically attended visit.
- The number and percentage of participants reporting each individual solicited administration site event (any grade, Grade 3 and resulting in medically attended visit) and solicited systemic event (any grade, Grade 3 and resulting in medically attended visit) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- For fever, the number and percentage of participants reporting fever by half degree (°C) cumulative increments during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- The incidence of each solicited administration site event and solicited systemic event (any grade and grade 3) will be represented graphically after each dose.
- The number and percentage of participants with any unsolicited AEs during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) with its exact 95% CI will be tabulated for each dose by group and by Medical Dictionary for Regulatory Activities (MedDRA) Primary System Organ Class (SOC), ***High Level Term (HLT) and*** preferred term (PT). Similar tabulation will be done for Grade 3 unsolicited AEs, for any causally related unsolicited AEs, for Grade 3 causally related unsolicited AEs and for unsolicited AEs resulting in a medically attended

visit. The verbatim reports of unsolicited AEs will be reviewed by a qualified person and the signs and symptoms will be coded according to the MedDRA Dictionary for Adverse Reaction Terminology. Every verbatim term will be matched with the appropriate Preferred Term.

- The number and percentage of participants with at least one report of SAE classified by the MedDRA Primary SOC, HLT and PTs and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI.
- The number and percentage of participants with at least one report of causally related SAE classified by the MedDRA Primary SOC, HLT and PTs and reported during the entire study period will be tabulated with exact 95% CI.
- The same tabulation will be presented for fatal SAEs.
- All SAEs will also be described in detail in a tabular listing.
- All AEs/SAEs leading to study/intervention discontinuation from dose 1 up to study end will be tabulated.
- The number and percentage of participants with at least one report of pIMD classified by the MedDRA Primary SOC, HLT and PTs and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI.
- The number and percentage of participants with at least one causally related pIMD classified by the MedDRA Primary SOC, HLT and PTs and reported during the entire study period will be tabulated with exact 95% CI.
- All pIMDs will also be described in detail in a tabular listing.
- The analyses of unsolicited AEs will include SAEs (unless otherwise specified).

The analysis of safety will also be performed by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA) and region (North America, Europe, Asia).

5.4.5. Additional considerations

- Compliance in completing solicited events information will be tabulated after each dose and overall.
- The number of days with solicited events reported during the 4-day follow-up period will be tabulated for each solicited event, after each dose using descriptive statistics (mean, min, Q1, median, Q3, maximum).
- The number of each solicited events (any and Grade 3) that are ongoing beyond the 4-day follow-up period will be reported with the duration in days (mean, min, Q1, median, Q3, maximum).
- The list of solicited administration site and systemic events, and definition of intensity are described in Section 10.3.3 and 10.3.9.1 of the protocol.

The measurement of erythema/swelling (in mm) and fever (in °C/F) will be categorized as follows:

Table 7 Intensity grading scale for solicited events

Grading	Erythema/swelling	Fever
0:	≤ 20 mm	< 38.0°C (100.4°F)
1:	> 20 - ≤ 50 mm	≥ 38.0°C(100.4°F) - ≤ 38.5°C(101.3°F)
2:	> 50 - ≤ 100 mm	> 38.5°C(101.3°F) - ≤ 39.0°C(102.2°F)
3:	> 100 mm	> 39.0°C(102.2°F)

Combined Solicited events and Unsolicited AEs

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

Solicited events will be coded by MedDRA as per the following codes:

Solicited event	Lower level term code	Corresponding Lower level term decode
Pain	10022086	Injection site pain
Erythema	10022061	Injection site erythema
Swelling	10053425	Injection site swelling
Fever	10016558	Fever
Headache	10019211	Headache
Fatigue	10016256	Fatigue
Myalgia	10028411	Myalgia
Arthralgia	10003239	Arthralgia

5.4.5.1. Concomitant Medication

Medications will be coded using the GSKDRUG dictionary.

5.4.6. Sensitivity Analyses

If more than 10% of participants miss the revaccination dose or don't have safety data reported after revaccination, the safety analysis described in section 5.4.4 will also be conducted on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

6. ANALYSIS INTERPRETATION

All analyses are descriptive.

7. INTERIM ANALYSES

7.1. Sequence of analyses

Analyses to evaluate objectives and endpoints will be performed stepwise:

- **Month 6:** A first analysis* will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 6 are available for all participants. This analysis will be considered as final for those endpoints.

  *Note: the analysis at Month 6 may be merged with the analysis at the subsequent timepoint.

- **Month 13**: A second analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 12 are available for all participants, as well as data up to 1 month after the first revaccination (Month 13) in the RSV_annual group. This analysis will be considered as final for those endpoints.
- **Month 18**: A third analysis will be performed on all safety and/or immunogenicity data available and as clean as possible, when data for secondary endpoints up to Month 18 are available for all participants. This analysis will be considered as final for those endpoints. Safety and immunogenicity analysis may be performed at different time depending on data availability.
- **Month 25**: A fourth analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 24 are available for all participants, as well as data up to 1 month after the second revaccination (Month 25) in the RSV_annual group. This analysis will be considered as final for those endpoints.
- The final **End of Study** analysis will be performed when all reactogenicity, safety and immunogenicity data for at least primary and secondary endpoints up to study conclusion are available for all participants (Month 36).
- If the data for tertiary endpoints become available at a later stage, (an) additional analysis/analyses will be performed.

Description	Analysis ID	Disclosure Purpose (CTRS=public posting, SR=study report, internal)
End of study analysis	E1_01	CTRS, SR
Analysis up to Month 6	E1_02	SR
Analysis up to Month 13	E1_03	CTRS, SR
Analysis up to Month 18	E1_04	Internal
Analysis up to Month 25	E1_05	Internal

7.2. Statistical considerations for interim analyses

All analyses will be conducted on final data (as clean as possible) and therefore no statistical adjustment for interim analyses is required.

8. CHANGES TO THE PROTOCOL DEFINED STATISTICAL ANALYSIS

This statistical analysis plan complements the analyses described in the protocol.

Analysis sets: The percentage has been changed from 10% to 5%, the rationale for changing is to align with other phase 3 RSV OA studies (see section 4).

Randomized set: This cohort has been deleted from the SAP, simply because it is not needed when there is no screening visit and elimination code 1010 has been deleted as well (see section 4.2.1).

Statistical analyses

The following changes have been made for the demography analyses

Demography (see section 5.2.1 and section 5.2.2**)**

Participant disposition in the ES and PPS will not be conducted by age category. The analysis by age category was considered not relevant for a phase 3 study.

Demography and baseline characteristics will also be summarized by age category, region and sex. Since the immunogenicity analyses will be conducted by these variables, it was considered relevant by the study team that the demography analyses should also be summarized by these variables.

The number of doses of the study intervention administered will be tabulated by group and by visit. This analysis will also be done by visit, since we have different vaccination schedules for the 3 study groups.

Regarding immunogenicity(see section 5.4.1 and section 5.4.2) and safety (see section 5.4.4**)**, the following analyses are not considered relevant for a phase 3 study hence have been deleted. In addition, we aim at reducing the number of tables produced thus limiting analyses to only important tables.

Immunogenicity (see section 5.4.1and section 5.4.2)

Humoral Immunogenicity

Individual post-primary vaccination results (at Months 18, 24, 30 and 36 when applicable) versus pre-vaccination results (Day 1) by group and individual post-revaccination results versus the corresponding pre-revaccination results (Months 12/24) in the RSV_annual group will be provided using scatter plots.

Distribution of the fold increase of the antibody titers/concentrations (post- over pre-primary vaccination dose, post- over the corresponding pre-revaccination and 1 month post-revaccination over 1 month post-previous dose) will be tabulated when applicable.

The ratio of fold increase (pre to post-vaccination) of anti-RSVPreF3-specific antibody concentrations over the fold increase (pre to post-vaccination) of anti-RSV-A/-B neutralizing antibody titers will be tabulated using descriptive statistics and displayed graphically using scatter plots.

Cell-mediated immune response

The kinetics of RSVPreF3-specific CD8+ T cells frequencies will be plotted as a function of time for participants with results available at all primary vaccination timepoints

Fold increase of the frequency of RSVPreF3-specific CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17 at each post-primary vaccination timepoint over pre-vaccination (Day 1) , at each post-revaccination over the corresponding pre-revaccination and at one month post-revaccination over 1 month post-previous dose in the RSV_annual group.

Safety (see section 5.4.4**)**

The prevalence of each solicited administration site event and solicited systemic event (any grade and grade 3) will be represented graphically over time for each group after each dose.

The following safety analyses have been added.

Summary tables of unsolicited adverse events and SAEs/pIMDs will be generated by SOC, **HLT** and PTs.

Safety analyses to be conducted by region in order to align with the immunogenicity analyses.

The number of each solicited events (any and Grade 3) that are ongoing beyond the 4-day follow-up period will be reported with the duration in days (mean, min, Q1, median, Q3, maximum) (see section 5.4.5).

The persistence modelling has been updated (see section 5.4.3.3).

8.1. Other changes in the SAP

Solicited events will be coded by MedDRA as per the following codes:

Solicited event	Lower level term code	Corresponding Lower level term decode
Erythema	10022061	Injection site erythema
Fever	10016558	Fever

The aim is to align the lower level term in the SAP with the coding done in CE domain in SDTM.

Clarification on how age at first dose in years will be computed (see section 10.1.3.1)

Clarification on how duration of events will be calculated (see section 10.1.3.6).

Clarification on counting rules for combining grade 3 solicited and unsolicited adverse events (see section 10.1.3.7)

9. NON-STANDARD DATA DERIVATION RULES AND STATISTICAL METHODS

The following sections describe additional derivation rules and statistical methods which are not presented in section 10.1 (Business rules for standard data derivations and statistical methods).

9.1. Handling of missing data

9.1.1. Dates

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

- Adverse event start dates with missing day:
  - If the month is not the same as the vaccine dose, then the imputed start date will be the 1st of the month
  - If the event starts in the same month as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the vaccine dose given during that month. If 'before vaccination' is selected, the imputed date will be one day before the vaccine dose given during that month.
- Adverse event start dates with missing day and month:
  - If the year is not the same as the vaccine dose, then the imputed start date will be the 1st of January.
  - If the event starts in the same year as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the first vaccine dose given during that year. If 'before vaccination' is selected, the imputed date will be one day before the first vaccine dose given during that year.
- Adverse event end dates with missing day: the imputed end date will be the last day of the month (30 or 31) or the study conclusion date whichever comes first.
- Adverse event end dates with missing day and month: the imputed end date will be the last day of the year (31st of December) or the study conclusion date whichever comes first.

All incomplete concomitant medication/vaccination start/end date will follow the rules above.

10. ANNEXES

10.1. Business rules for standard data derivations and statistical methods

This section contains standard rules for data display and derivation for clinical and epidemiological studies.

10.1.1. 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 vaccination is selected, the relative dose for the event will be the one administered on the start day of the event. If 'before vaccination is selected, the relative dose for the event will be the dose prior to this one.

10.1.2. Handling of missing data

10.1.2.1. 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.

See also exceptions in section 9.1.

10.1.2.2. Laboratory data

Refer to section 5.1.2 for more details

10.1.2.3. Daily recording of solicited events

10.1.2.3.1. Studies with paper diaries

For studies using paper diaries which have questions in the CRF indicating the presence or absence of solicited events, the following rules are applicable:

- Denominators for the summary of administration site (or systemic) solicited events will be calculated using the number of participants who respond "Yes" or "No" to the question concerning the occurrence of administration site (or systemic) events.

- When a specific solicited event is marked as having not occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=N for the specified post-dose period for the event in question), all daily measurements will be imputed as Grade 0.
- When a specific solicited event is marked as having occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=Y for the specified post-dose period for the event in question), any missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.
- When the occurrence of a specific solicited event is not present (i.e. SDTM CE.CEOCCUR is neither Y nor N for the specified post-dose period for the event in question) but the group of solicited events (administration site or systemic) is marked as having occurred (i.e. SDTM CE.CEOCCUR=Y), all missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.

The following table shows how participants contribute to each category for a specific solicited event over the Day X to Day Y post-dose period:

Solicited event category	Participants included in the calculation of the numerator
Any	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y or with the adverse event marked as present and at least one missing daily recording between Day X and Day Y
At least grade 1	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y
At least grade 2	All participants with at least one occurrence of the adverse event at grade 2 or grade 3 between Day X and Day Y
At least grade 3	All participants with at least one occurrence of the adverse event at grade 3 between Day X and Day Y

10.1.2.4. 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.

10.1.3. Data derivation

10.1.3.1. 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

In this study, we will collect only the year of birth. The rules for handling missing day and/or month in the DOB are given in section 10.1.2.1.

Since only the year of birth will be collected in the eCRF, there might be some discrepancies between the age computed using standard derivation rules and the age category used in SBIR for the minimization.

Therefore, for the analysis by age, the age categories defined in Table 4 will be determined according to the information entered in SBIR (variable AGEGRP in SDTM SUPPDM), except for "≥65 YOA" category which will be obtained using the derived age because this category is not used in SBIR for minimization

10.1.3.2. 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

10.1.3.3. Numerical serology results

Numerical serology results will be derived from the content of IS.ISORRES in the SDTM dataset. For humoral 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 and value is <=ULOQ	value
"value" and value is > ULOQ	ULOQ*
All other cases	Missing

*This rule will be used to compute descriptive statistics (GMTs, fold increase etc).

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

GMT or 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 10.1.3.3 for the purpose of GMT/GMC calculation. Cut-off values are defined by the laboratory before the analysis.

10.1.3.5. 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).

10.1.3.6. 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 3 March 2018 and ends on 12 March 2018 has a duration of 10 days.

The duration of solicited events will be calculated as the sum of the individual days with the event reported at grade 1 or higher or reported as missing ("NOT DONE"), during the solicited event period. The duration of solicited events ongoing beyond the solicited period will be calculated as end date – start date + 1. The goal of this change was to align this text with the standard programs.

10.1.3.7. Counting rules for combining solicited and unsolicited adverse events

For output combining solicited and unsolicited adverse events, all serious adverse events will be considered systemic events since the administration site flag is not included in the expedited adverse event CRF pages. Unsolicited adverse events with missing administration site flag will also be considered systemic.

Multiple events with the same preferred term which start on the same day are counted as only one occurrence.

In case a solicited event is worsening to Grade 3 after the solicited period, it should be reported only in the following table.

Summary of grade 3 adverse events (solicited and unsolicited) within 30 days following each dose and overall

10.1.3.8. 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.

10.1.4. Display of decimals

10.1.4.1. Percentages

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

10.1.4.2. Demographic/baseline characteristics statistics

The mean, median, and standard deviation for continuous baseline characteristics 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 maximum and minimum of transformed body temperatures will be displayed with one decimal.

10.1.4.3. Serological summary statistics

For each assay, geometric mean titers (GMT) or concentrations (GMC) and their confidence limits will be presented with one decimal, as well as GMT/GMC fold increase from pre-dose.

The mean, median, standard deviation and quartile values for frequency of RSVPreF3 specific-CD4+ and CD8+ T cells and for the fold increase (post over pre-vaccination) will be presented with one decimal. The minimum and maximum values will be presented with no decimal.

10.1.5. Statistical methodology

10.1.5.1. Exact confidence intervals around proportions

The exact confidence intervals around within-group proportions are derived using the method of Clopper and Pearson [Clopper, 1934].

11. 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.

12. REFERENCES

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

David M-P, Van Herck K, Hardt K, Tibaldi F, Dubin G, Descamps D, et al. Long-term persistence of anti-HPV-16 and-18 antibodies induced by vaccination with the AS04-adjuvanted cervical cancer vaccine: modeling of sustained antibody responses. GYNECOLOGIC ONCOLOGY. 2009;115(3):S1–6.

Fraser C, Tomassini JE, Xi L, Golm G, Watson M, Giuliano AR, et al. Modeling the long-term antibody response of a human papillomavirus (HPV) virus-like particle (VLP) type 16 prophylactic vaccine. Vaccine 2007 May 22;25(21):4324-33

Pan W Akaike's information criterion in generalized estimating equations Biometrics, 57 (1) (2001), pp. 120-125

Rubin, D. B., Stern, H. S. AND Vehovar, V. Handling “don’t know” survey responses: the case of the Slovenian plebiscite. Journal of the American Statistical Association 90, 1995, 822–828

Statistical Analysis Plan

Title:	A phase 3, randomized, open-label, multi-country study to evaluate the immunogenicity, safety, reactogenicity and persistence of a single dose of the RSVPreF3 OA investigational vaccine and different revaccination schedules in adults aged 60 years and above.
eTrack study number and Abbreviated Title	212496 (RSV OA=ADJ-004)
Scope:	All data pertaining to the above study except the Safety Review Team analyses for the first 50 participants aged 80 years and above
Date of Statistical Analysis Plan	Final: 9 February 2021

APP 9000058193 Statistical Analysis Plan Template V5 (Effective date: 1July2020)

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LIST OF ABBREVIATIONS

AE	Adverse event
AESI	Adverse Events of Special Interest
CD	Community Dwelling
CI	Confidence Interval
COVID-19	Coronavirus Disease 2019
CRF	Case Report Form
CTRS	Clinical Trial Registry Summary
Eli Type	Internal database code for type of elimination code
ELISA	Enzyme-linked immunosorbent assay
ELU/mL	ELISA unit per milliliter
ES	Exposed Set
GMC	Geometric mean antibody concentration
GMT	Geometric mean antibody titre
GSK	GlaxoSmithKline
IU/mL	International units per milliliter
LL	Lower Limit of the confidence interval
LTCF	Long Term Care Facility
MedDRA	Medical Dictionary for Regulatory Activities
NA	Not Applicable
PD	Protocol Deviation
PPS	Per-Protocol Set
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SBIR	GSK Biologicals Internet Randomization System
SD	Standard Deviation
SDTM	Study Data Tabulation Model
UL	Upper Limit of the confidence interval

1. DOCUMENT HISTORY

Date	Description	Protocol Version
15 February 2021	First version	Final: 28 October 2020

2. OBJECTIVES/ENDPOINTS

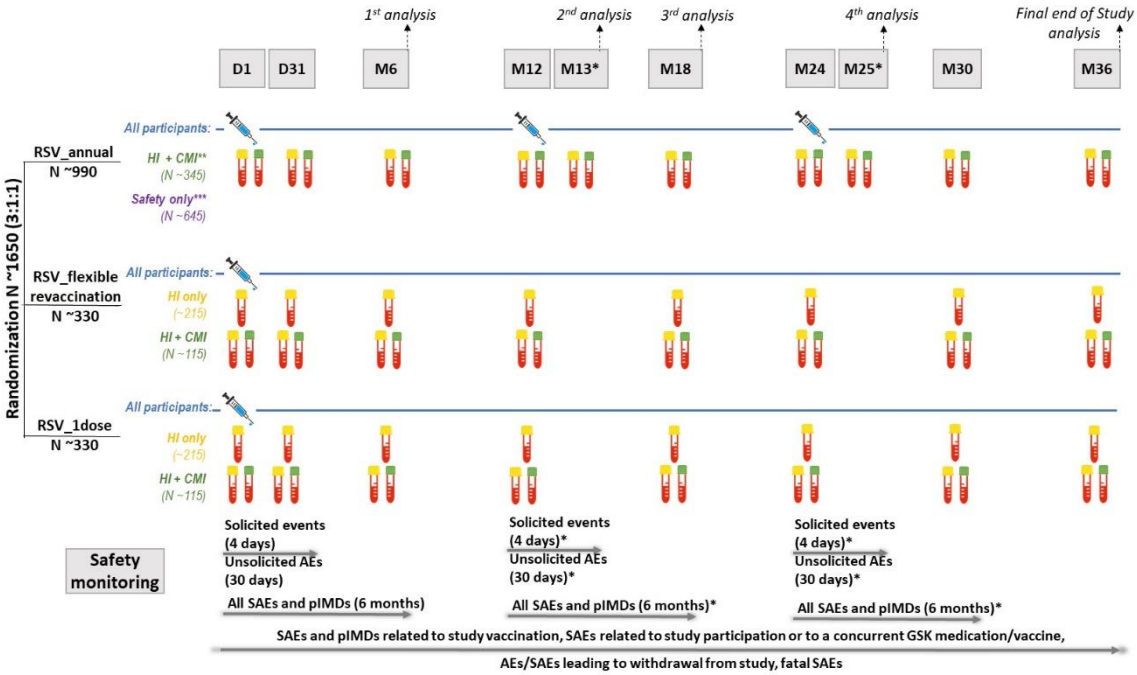
Objectives	Endpoints
Primary	
To evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • Neutralizing antibody titers against RSV-A • Neutralizing antibody titers against RSV-B.
Secondary	
To further evaluate the humoral immune response following a 1-dose primary schedule of RSVPreF3 OA investigational vaccine up to 12 months post-Dose 1.	Humoral immune response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), and at 6 and 12 months post-Dose 1 (Months 6 and 12), in a subset of participants: • RSVPreF3-specific Immunoglobulin G (IgG) antibody concentrations.
To evaluate the humoral immune response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses, up to study end.	Humoral immune response at Months 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Neutralizing antibody titers against RSV-A and RSV-B • RSVPreF3-specific IgG antibody concentrations.
To evaluate the cell-mediated immune (CMI) response following 1 dose of the RSVPreF3 OA investigational vaccine and following revaccination doses up to study end.	CMI response at pre-vaccination (Day 1), 30 days post-Dose 1 (Day 31), at Months 6, 12, 18, 24, 30 and 36 post-Dose 1, and at 1 month after each revaccination dose (Months 13 and 25), in a subset of participants: • Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
To evaluate the safety and reactogenicity of each vaccination schedule of the RSVPreF3 OA investigational vaccine in all participants.	• Occurrence of each solicited administration-site and systemic event during a 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) after each vaccination. • Occurrence of any unsolicited AE during a 30-day follow up period (i.e., on the day of vaccination and 29 subsequent days) after each vaccination. • Occurrence of all SAEs and pIMDs up to 6 months after each vaccination. • Occurrence of fatal SAEs, related SAEs and related pIMDs from first vaccination (Day 1) up to study end (Month 36).
Tertiary	
To further characterize immune responses to the RSVPreF3 OA investigational vaccine.	• Any further exploratory immunology in a subset of participants, such as, but not limited to: – Antibodies against specific protein F epitopes. – Potential new immunological markers for protection. – Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing one or any combination of immune marker(s)

CMI = cell-mediated immunity, AE = adverse event, SAE = serious adverse event, pIMD = potential immune-mediated disease.

3. STUDY DESIGN

3.1. Overall design

Figure 1 Study design overview



N = number of participants; D = Day; M = Month; AE = adverse event; HI = humoral immunity; CMI = cell-mediated immunity; pIMD = potential immune-mediated disease; SAE = serious adverse event.

= blood sample for humoral immune responses (only applicable for the 345 participants of the RSV_annual group; and for all participants of the RSV_flexible revaccination and RSV_1dose groups)

= blood sample for CMI (only applicable for the ~345 participants of the RSV_annual group, and ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups).

Note: For group RSV_annual, Revaccination Year 1 = Month 12; Revaccination Year 2 = Month 24.

* Visit and follow-up of solicited events, unsolicited AEs, SAEs and pIMDs only applicable for RSV_annual group

** For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws, and will only be followed up for safety and reactogenicity.

***For those participants in the RSV_annual group without blood sample collections, visits at Months 6, 18, 30 and 36 may be through a phone contact. However, phone contact should be attempted only if site visit is not possible at Months 6, 18, 30 and 36.

- **Type of study**: self-contained.
- **Experimental design**: Phase 3, randomized, open-label, multi-country study with 3 parallel groups (see Figure 1).
- **Duration of the study**: ~ 36 months for each participant.
- **Primary completion date**: Month 12.
- **Control**: none.
- **Blinding**: open-label. Refer to the protocol for details.

- **Data collection**: standardized electronic Case Report Form (eCRF). Solicited events and unsolicited AEs will be collected using a participant Diary card (paper Diary card).
- **Safety monitoring**: the study will be conducted with oversight by the project Safety Review Team (SRT). Please refer to protocol for the description of review of safety data by the SRT.
- **Study groups**: refer to Figure 1 and Table 1 for an overview of the study groups

Table 1 Study groups, intervention and blinding foreseen in the study

Study Groups	Number of participants	Age	Intervention			Blinding
			Primary vaccination	Revaccination Year 1 (Month 12)	Revaccination Year 2 (Month 24)	Open
RSV_annual*	~990	≥60 years	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	RSVPreF3 OA investigational vaccine	X
RSV_flexible revaccination**	~330	≥60 years	RSVPreF3 OA investigational vaccine			X
RSV_1dose	~330	≥60 years	RSVPreF3 OA investigational vaccine	(none)	(none)	X

* RSV_annual group will be split in 3 technical groups in SBIR in order to have a randomization ratio of 1:1:1:1:1: for treatment allocation and avoid predictability.
**Based on immunogenicity data from this study and efficacy results from the Phase 3 study RSV OA=ADJ-006, a revaccination might be decided for this group.

- **Groups and sub-groups definition for analysis:** Refer to the tables below for a description of the groups and subgroups labels that will be used in the Tables Figures and Listings (TFLs).

  For some of the analyses up to 12 months post dose 1, the 3 study groups will be pooled, and from Month 13, the RSV_flexible revaccination and RSV_1dose groups will be pooled, at applicable timepoints. The following group names will be used in the TFLs:

Table 2 Group names and definition for footnote in the TFLs

Group order in tables	Group label in tables	Group definition for footnote
1	RSV_annual	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1, followed by a revaccination dose at 12 months post-Dose 1 and at 24 months post-Dose 1
2	RSV_flexible revaccination	Participants receiving the first dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1 and a revaccination dose will be given whenever a revaccination would be needed
3	RSV_1dose	Participants receiving a single dose (Dose 1) of RSVPreF3 OA investigational vaccine at Day 1

Table 3 Pooled group names and definition for footnote in the TFLs

Pooled Group label in tables	Groups to be pooled	Pooled definition for footnote	Comment
All	RSV_annual, RSV_flexible revaccination and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine in all three groups	Applicable on data reported up to Month 12
Flexible+1dose	RSV_flexible revaccination, and RSV_1dose	Participants receiving one dose of RSVPreF3 OA investigational vaccine in the RSV_flexible revaccination, and RSV_1dose groups	Applicable on data reported from Month 13

Table 4 Sub-group names and definitions for footnote in the TFLs

Sub-analysis	Subgroup order in tables	Subgroup label in tables	Subgroup definition for footnote
By age	1	≥65YOA	≥65 years old participants
	2	≥70YOA	≥70 years old participants
	3	≥80YOA	≥80 years old participants
	4	60-69YOA	60-69 years old participants
	5	70-79YOA	70-79 years old participants
By sex	1	Female	Female
	2	Male	Male
By region	1	North America	Participants from North America (United States of America)
	2	Europe	Participants from Europe (Finland, Germany)
	3	Asia	Participants from Asia (Japan, Taiwan)

YOA = Years of age

- **Allocation of participants to assay subsets**

Allocation of participants to assay subsets will be performed using SBIR. The subsets are detailed below.

	RSV_annual	RSV_flexible revaccination	RSV_1dose
HI subset	~345*	All participants (~330)	All participants (~330)
CMI subset	~345*	~115	~115

*For the RSV_annual group, the same ~345 participants will be part of both the HI and CMI subsets. The remaining ~645 participants will have no blood draws, and will only be followed up for safety and reactogenicity.

- **HI subset:** ~345 participants from the RSV_annual group, and all participants from the RSV_flexible revaccination and RSV_1dose groups. These participants will have blood samples collected for testing of humoral immunity at each visit applicable for their study group.
- **CMI subset**: ~345 participants from the RSV_annual group, and ~115 participants each from the RSV_flexible revaccination and RSV_1dose groups. These participants will have additional blood samples collected for CMI testing at each visit applicable for their study group.

4. ANALYSIS SETS

4.1. Definition

Table 5 Populations for analyses

Analysis set	Description
Enrolled set	Participants who agreed to participate in a clinical study after **completion of the informed consent process.**
Exposed set (ES)	All participants who received at least 1 dose of the study intervention. The allocation in a group is done in function of the administered intervention.
Per Protocol set (PPS)	All participants who received at least 1 dose of the study intervention to which they are randomized and have post-vaccination data, minus participants with protocol deviations that lead to exclusion.

The primary analysis for immunogenicity will be performed on the Per Protocol set (PPS). If in any study group the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 10%, a second analysis will be performed on the Exposed Set (ES). The immunogenicity analysis will be performed overall, by sex, by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and by region (North America, Europe, Asia).

4.2. Criteria for eliminating data from Analysis Sets

4.2.1. Randomized Set

All participants who will be randomized to 1 of the 3 study groups at Day 1 will be included in the Randomized Set. Code 1010 (Vaccine number not allocated) will be used for identifying participants eliminated from the Randomized Set (Table 6)

4.2.2. Elimination from Exposed Set (ES)

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

4.2.3. Elimination from Per-protocol analysis Set (PPS)

4.2.3.1. Excluded participants

A participant will be excluded from the populations of analysis under the following conditions

- For codes 800, 900, 1010, 1030 and 1050: participants will be eliminated for all visits
- For codes 1040, 1070, 1080, 1090, 2010, 2040, 2050, 2080: participants will be eliminated from a specific visit (at which the condition is met) onwards.
- For codes 2090, 2100, 2120, 2130: participants will be eliminated at the specific visit at which the condition is met.

Table 6 List of elimination codes

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
800	Fraudulent data	All	ES and PPS
900	Invalid informed consent	All	ES and PPS
1010	Vaccine number not allocated	Day 1	Randomized Set, ES, PPS
1030	Study intervention not administered at all	All	ES and PPS
1040	Administration of concomitant vaccine(s) forbidden in the protocol • Any investigational or non-registered vaccine other than the study vaccine used during the study period beginning 30 days before the first dose of study vaccine, or planned use during the study period. • Planned or actual administration of a vaccine not foreseen by the study protocol in the period starting 30 days before each dose and ending 30 days after each dose of study vaccine administration, with the exception of inactivated, split virion and subunit influenza vaccines which can be administered up to 14 days before or from 14 days after each study vaccination. • Previous vaccination with an RSV vaccine	All	PPS
1050	Randomization failure	Day 1	PPS
1070	Vaccine administration not according to protocol • Incomplete vaccination course	Vaccination visits at Day 1, Month 12, and 24 Months 12 and 24 are applicable to the	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Participant was vaccinated with the correct vaccine but containing a lower volume • Participant was re-vaccinated while it was not planned • Route of the study intervention is not intramuscular • Wrong reconstitution of administered vaccine	RSV_annual group only	
1080	Vaccine administration after a Temperature deviation • Vaccine administered despite a Good Manufacturing Practices (GMP) no-go temperature deviation	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group only	PPS
1090	Vaccine administration after expiration	Day 1, Month 12, and 24 Months 12 and 24 are applicable to the RSV_annual group only	PPS
2010	Protocol deviation linked to inclusion/exclusion criteria	All	PPS
2040	Administration of any medication forbidden by the protocol • Any investigational or non-registered medication used during the study period • Administration of long-acting immune-modifying drugs at any time during the study period (e.g. *infliximab*) • Immunoglobulins and/or any blood products administered during the study period • Chronic administration (defined as more than 14 consecutive days in total) of immunosuppressants or other	All	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	immune-modifying drugs during the period starting 90 days prior to the study vaccine administration or planned administration during the study period. For corticosteroids, this will mean prednisone ≥20 mg/day, or equivalent.		
2050	Intercurrent medical condition • Participants may be eliminated from the PPS for immunogenicity if, during the study, they incur a condition that has the capability of altering their immune response (intercurrent medical condition) or are confirmed to have an alteration of their initial immune status.	All	PPS
2080	Participants did not comply with dosing schedule • **RSV_annual**: number of days between dose 1 and revaccination dose at Month 12 is outside [350-380 days] • **RSV_annual**: number of days between dose 1 and revaccination dose at Month 24 is outside [715-745 days]	Month 12 and 24	PPS
2090	Participants did not comply with blood sample schedule • Number of days between dose 1 and Day 31 blood sample is outside [30-42 days] • Number of days between dose 1 and Month 6 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 12 blood sample is outside [350-380 days]	Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

Code	Condition under which the code is used	Visit (timepoints) where the code is checked	Applicable for analysis set
	• Number of days between Month 12 and Month 18 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 24 blood sample is outside [715-745 days] • Number of days between Month 24 and Month 30 blood sample is outside [180-210 days] • Number of days between dose 1 and Month 36 blood sample is outside [1080-1110 days] *For RSV_annual group only:* • Number of days between revaccination at Month 12 and Month 13 blood sample is outside [30-42 days] • Number of days between revaccination at Month 24 and Month 25 blood sample is outside [30-42 days]		
2100	Serological results not available post-vaccination • No immunological result at visit x for all the following tests: RSV A/B Neutralizing antibody titer, RSVPreF3-specific IgG antibody concentration and RSVPreF3-specific CD4+/CD8+ T cells frequency	Day 31, Month 6, 12, 13, 18, 24, 25, 30, 36 Month 13 and 25 are applicable to the RSV_annual group only	PPS
2120	Obvious incoherence or abnormality or error in laboratory data • Unreliable released data as a result of confirmed sample mismatch or confirmed inappropriate sample handling at laboratory	Day 1, 31, Month 6, 12, 13, 18, 24, 25, 30, 36	PPS

5. STATISTICAL ANALYSES

Standard data derivation rules and stat methods are described in section 10.1 while the study specific data derivation rules and stat methods are described in section 9.

For the sake of analysis, data up to Month 12 will be analyzed by group (RSV_annual, RSV_flexible revaccination and RSV_1dose), and also the 3 groups will be pooled. From Month 13, the analysis will be tabulated by group, and also for RSV_annual versus the pooled RSV_flexible revaccination and RSV_1dose groups, at applicable time points.

If more than 5% of participants in the RSV_annual group received only 1 dose or only 2 doses, an analysis by subgroup according to the number of dose(s) administered will be performed. The goal of this analysis is to describe the characteristics of participants in the RSV_annual group who ultimately receive different numbers of revaccination doses in order to rule out the possibility of systematic differences between these subgroups.

5.1. General considerations

5.1.1. Demography

For a given participant and a given demographic variable, missing measurements will not be replaced.

5.1.2. Immunogenicity

- Any missing or non-evaluable immunogenicity measurement will not be replaced. The descriptive analysis performed for each assay at each time point will exclude participants with a missing or non-evaluable measurement.
- The geometric mean titers/concentrations (GMTs/GMCs) will be computed by taking the anti-logarithm of the arithmetic mean of the $\log_{10}$ transformed titers/concentrations.
- 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.
- Antibody titers/concentrations below the assay cut-off will be given an arbitrary value of half the assay cut-off for the purpose of GMT/GMC calculation.
- Antibody titers/concentrations above the Upper Limit of Quantification (ULOQ) value will be given the ULOQ value for the purpose of GMT/GMC calculation.
- The mean geometric increase (MGI) is calculated by the geometric mean of ratios of antibody titer/concentrations of each post-primary vaccination time point over pre-primary vaccination (Day 1).

5.1.3. Reactogenicity/Safety

- For a given participant and the analysis of solicited events within 4 days post-vaccination, missing or non-evaluable measurements will not be replaced. Therefore, the analysis of solicited events will include only vaccinated participants with documented solicited safety data (i.e., paper diary completed).
- For analysis of unsolicited AEs, SAEs, pIMDs and concomitant medications, all vaccinated participants will be considered. Participants who did not report an event or concomitant medication will be considered as participants without the event or the concomitant medication, respectively.

5.2. Analysis of demography and baseline characteristics

5.2.1. Analysis planned in the protocol

Descriptive summaries will be performed for each group (RSV_annual, RSV_flexible revaccination and RSV_1dose) and overall. The same will be performed in each subset (humoral immune subset and CMI subset).

Demographic/baseline characteristics (age at vaccination in years, sex, race, ethnicity, type of residence (CD/LTCF) and smoking status) will be summarized using descriptive statistics:

- Frequency tables will be generated for categorical variables such as race.
- Mean, median, standard deviation and range will be provided for continuous data such as age.
- The distribution of participants will be tabulated as a whole and per group, for each age category and for each subset.

The number of doses of the study intervention administered will be tabulated by group.

Withdrawal status will be summarized by group using descriptive statistics:

- The number of participants enrolled into the study as well as the number of participants excluded from Per Protocol (PP) analyses will be tabulated.
- The numbers of withdrawn participants will be tabulated according to the reason for withdrawal.

Participant disposition in the ES and PPS will be reported as a whole and per group, and for each age category.

- The number and percentage of participants using concomitant medication (any medication, any antipyretic and any antipyretic taken prophylactically, respectively) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) and during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) will be summarized by group after each vaccine dose and overall, and after the revaccination.

5.2.2. Additional considerations

- The analysis of demographic characteristics by group will be performed on the ES and on the PPS.
- Demography and baseline characteristics will also be summarized by country.
- The smoking status and exposure history of participants will be tabulated according to the
  - number of years of smoking exposure for both current and former smokers.
  - number of years of smoking exposure, summarized separately for tobacco and electronic smoking devices

5.3. Primary endpoint(s)

5.3.1. Analysis planned in the protocol

The primary analysis for immunogenicity will be performed on the Per Protocol set (PPS). If in any study group the percentage of vaccinated participants with serological results excluded from the PPS for immunogenicity is at least 10%, a second analysis will be performed on the Exposed Set (ES).

5.3.1.1. Humoral immune response – up to Month 12

The analysis of the primary vaccination time points up to Month 12 will be performed for each group and for the 3 groups pooled. For each time point with blood sample collection for humoral immune response up to Month 12 and for each assay (RSV-A/-B neutralization assays), unless otherwise specified, the following analyses will be performed:

Within groups evaluation:

- Percentage of participants with antibody titers/concentrations above a positivity cut-off and their exact 95% confidence interval (CI) will be tabulated.
- GMTs/GMCs and their 95% CI will be tabulated and represented graphically. Furthermore, to account for all the time points at which the blood samples are collected, a mixed model will be fitted, from which the GMTs/GMCs will be computed.
- Antibody titers/concentrations will be displayed using reverse cumulative curves.
- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI.
- Individual post-vaccination results (at Days 31 and at Months 6 and 12) versus pre-vaccination results (Day 1) will be plotted using scatter plots.
- Distribution of the fold increase of the antibody titers/concentrations (post- over pre-primary vaccination titers) will be tabulated.

5.4. Secondary endpoint(s)

5.4.1. Humoral immune response

The same analyses as described above for the primary endpoint will be performed for PreF3 IgG ELISA up to Month 12 and for each time point with blood sample collection for humoral immune response **from study Month 13 up to study end** and for each assay (preF3 IgG **ELISA and RSV-A/-B neutralization assays**). The analysis of the post-primary vaccination time points from study Month 13 up to study end will be performed for each group and for the pooled RSV_flexible revaccination and RSV_1dose groups.

Only the analyses requiring further clarification of the exact time points analyzed are summarized below:

Within groups evaluation:

- The MGI i.e. geometric mean of ratios of antibody titer/concentrations will be tabulated with 95% CI:
  - For each post-primary vaccination time point (at Months 18, 24, 30 and 36 when applicable) over pre-primary vaccination (Day 1),
  - For each post-revaccination time point over corresponding pre-revaccination (Months 12/24) in the RSV_annual group,
  - For 1 month post-revaccination at Month 12 (Month 13) over 1 month post-Dose 1 vaccination (Day 31) in RSV_annual group.
  - For 1 month post-revaccination at Month 24 (Month 25) over 1 month post-revaccination at Month 12 (Month 13) in RSV_annual group
- Individual post-primary vaccination results (at Months 18, 24, 30 and 36 when applicable) versus pre-vaccination results (Day 1) by group and individual post-revaccination results versus the corresponding pre-revaccination results (Months 12/24) in the RSV_annual group will be provided using scatter plots.
- Distribution of the fold increase of the antibody titers/concentrations (post- over pre-primary vaccination dose, post- over the corresponding pre-revaccination and 1 month post-revaccination over 1 month post-previous dose) will be tabulated when applicable.
- The ratio of fold increase (pre to post-vaccination) of anti-RSVPreF3-specific antibody concentrations over the fold increase (pre to post-vaccination) of anti-RSV-A/-B neutralizing antibody titers will be tabulated using descriptive statistics and displayed graphically using scatter plots.

In addition, the following evaluations will be performed:

- The kinetics of GMTs/GMCs will be plotted as a function of time for participants with results available at all post-primary vaccination time points.

The immunogenicity analysis will be performed overall, by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and region (North America, Europe, Asia).

5.4.2. Cell-mediated immune response

Within groups evaluation:

The following parameters will be summarized by group at each time point for which blood samples are collected, for the 3 groups pooled for time points up to study Month 12, for the pooled RSV_flexible revaccination and RSV_1dose up to study end using descriptive statistics (N, geometric mean, min, Q1, median, Q3, max) in the CMI subset:

- Frequency of RSVPreF3-specific CD4+ and/or CD8+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
- The kinetics of RSVPreF3-specific CD4+ T cells frequencies will be plotted as a function of time for participants with results available at all primary vaccination time points.
- Fold increase of the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17 at each post-primary vaccination time point over pre-vaccination (Day 1) by group, at each post-revaccination over the corresponding pre-revaccination and at one month post-revaccination over 1 month post-previous dose in the RSV_annual group.

The immunogenicity analysis will be performed by sex, age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA, and region (North America, Europe, Asia).

5.4.3. Additional considerations

- **Descriptive statistics** of the cell-mediated immune response will be tabulated and displayed graphically using boxplots (min, Q1, median, Q3, max), by group and timepoint.

  The RSVPreF3-specific CD4+ T cell frequencies will be obtained by subtracting the background value to the antigen-induced value, and by setting to 1 all values less than or equal to zero for geomean calculation and graphical representation. Frequencies will be expressed as the number of cells per million of CD4+ T cells.

  More specifically, the frequencies of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine [$Freq^{2+}$] will be computed as follows:

  $$Freq^{2+}_{Background} = \frac{n^{2+}_{background}}{N^{CD4}_{Background}} \quad \text{and} \quad Freq^{2+}_{Induction} = \frac{n^{2+}_{Induction}}{N^{CD4}_{Induction}},$$

  And

  $$Freq^{2+}_{Specific} = Freq^{2+}_{Induction} - Freq^{2+}_{Background}$$

where

$n^{2+}_{Background}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with medium only (background)

$n^{2+}_{Induction}$= number of CD4+ T cells expressing at least 2 activation markers including at least one cytokine after stimulation with a pool of peptides covering RSVPreF3 (induction)

$N^{CD4}_{Back/Ind}$= Total number of CD4 T cells involved in the assay (background or induction)

Same computations will be done for all CMI responses that will be analysed, i.e.:

- Frequency of RSVPreF3-specific CD4+ and/or CD8+T-cells expressing **at least 2 activation markers including at least 1 cytokine*** among CD40L, 41BB, IL-2, TNF-α, IFN-γ, IL-13, and IL-17, as measured by ICS using PBMCs.

  * cytokines are IL-2, TNF-α, IFN-γ, IL-13, and IL-17

5.4.3.1. Assessment of long-term persistence of the immune response at Months 12, 24 and 36 after dose 1.

The exploratory analysis for modelling the long-term persistence of immune response (humoral and cellular) post dose 1 vaccination will constitute of the following;

- A piece-wise linear mixed model, power law model and modified power law model [David, 2009] for repeated measurements (all data available from Day 1, 31 and Month 6, 12, 18, 24, 30, 36 post dose 1 vaccination, for the 3 groups pooled for time points up to study Month 12, for the pooled RSV_flexible revaccination and RSV_1dose from Month 13 up to study end) will be used to model over time
  - the frequency of RSVPreF3-specific CD4+ T cells expressing at least 2 activation markers including at least one cytokine among CD40L, 4-1BB, IL-2, TNF-α, IFN-γ, IL-13, IL-17.
  - the neutralizing antibody titers against RSV-A, RSV-B and RSVPreF3-specific IgG antibody concentrations.
- The three models based on data up to M36 will be presented graphically. Models based on data up to M12 and on data up to M36 will be presented side by side.

5.4.3.2. Sensitivity analyses

- In case the COVID-19 pandemic is still ongoing during the study conduct, the following sensitivity analyses might be considered.
  - During the assessment of long-term persistence of the immune response, the models described in section 5.4.3.1 might be fitted taking into account; Participants who received the first dose of study vaccine and with full follow-up (attending all study visits)

- Similarly, a mixed model will be fitted for the evaluation of GMT/GMC based on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

5.4.4. Safety analysis

Primary vaccination and revaccination dose:

The safety analysis will be performed on the ES. A descriptive analysis by group, for the 3 groups pooled up to Month 12, for the RSV_flexible revaccination and RSV_1dose groups pooled vs RSV_annual group from study Month 13 up to study end will present a summary of:

- The number and percentage of participants with at least one administration site event (solicited and unsolicited), with at least one systemic event (solicited and unsolicited) and with any AE during the 4-day or 30-day follow-up period will be tabulated with exact 95% CI after each dose. The same computations will be done for Grade 3 AEs, and for Grade 3 non-serious AEs and for AEs resulting in medically attended visit.
- The number and percentage of participants reporting each individual solicited administration site event (any grade, Grade 3 and resulting in medically attended visit) and solicited systemic event (any grade, Grade 3 and resulting in medically attended visit) during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- For fever, the number and percentage of participants reporting fever by half degree (°C) cumulative increments during the 4-day follow-up period (i.e., on the day of vaccination and 3 subsequent days) will be tabulated for each group after each dose.
- In addition, the prevalence of each solicited administration site event and solicited systemic event (any grade and grade 3) will be represented graphically over time for each group after each dose. The incidence of each solicited administration site event and solicited systemic event (any grade and grade 3) will be represented graphically for each group after each dose.
- The number and percentage of participants with any unsolicited AEs during the 30-day follow-up period (i.e., on the day of vaccination and 29 subsequent days) with its exact 95% CI will be tabulated for each dose by group and by Medical Dictionary for Regulatory Activities (MedDRA) preferred term. Similar tabulation will be done for Grade 3 unsolicited AEs, for any causally related unsolicited AEs, for Grade 3 causally related unsolicited AEs and for unsolicited AEs resulting in a medically attended visit. The verbatim reports of unsolicited AEs will be reviewed by a qualified person and the signs and symptoms will be coded according to the MedDRA Dictionary for Adverse Reaction Terminology. Every verbatim term will be matched with the appropriate Preferred Term.

- The number and percentage of participants with at least one report of SAE classified by the MedDRA Preferred Terms and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI.
- The number and percentage of participants with at least one report of causally related SAE classified by the MedDRA Preferred Terms and reported during the entire study period will be tabulated with exact 95% CI.
- The same tabulation will be presented for fatal SAEs.
- All SAEs will also be described in detail.
- All AEs/SAEs leading to study/intervention discontinuation from dose 1 up to study end will be tabulated.
- The number and percentage of participants with at least one report of pIMD classified by the MedDRA Preferred Terms and reported from Dose 1 up to 6 months post-Dose 1 and from revaccination dose up to 6 months post-revaccination will be tabulated with exact 95% CI.
- The number and percentage of participants with at least one causally related pIMD classified by the MedDRA Preferred Terms and reported during the entire study period will be tabulated with exact 95% CI.
- All pIMDs will also be described in detail.
- The analyses of unsolicited AEs will include SAEs (unless otherwise specified).

The analysis of safety will also be performed by age category (age at Dose 1: ≥ 65 YOA, ≥70 YOA, ≥80 YOA, 60-69 YOA and 70-79 YOA).

5.4.5. Additional considerations

- Compliance in completing solicited events information will be tabulated after each dose and overall.
- The number of days with solicited events reported during the 4-day follow-up period will be tabulated for each solicited event, after each dose using descriptive statistics (mean, min, Q1, median, Q3, maximum).
- The list of solicited administration site and systemic events, and definition of intensity are described in Section 10.3.3 and 10.3.9.1 of the protocol.

  The measurement of erythema/swelling (in mm) and fever (in °C/F) will be categorized as follows:

Table 7 Intensity grading scale for solicited events

Grading	Erythema/swelling	Fever
0:	≤ 20 mm	< 38.0°C (100.4°F)
1:	> 20 - ≤ 50 mm	≥ 38.0°C(100.4°F) - ≤ 38.5°C(101.3°F)
2:	> 50 - ≤ 100 mm	> 38.5°C(101.3°F) - ≤ 39.0°C(102.2°F)
3:	> 100 mm	> 39.0°C(102.2°F)

Combined Solicited events and Unsolicited AEs

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

Solicited events will be coded by MedDRA as per the following codes:

Solicited event	Lower level term code	Corresponding Lower level term decode
Pain	10022086	Injection site pain
Erythema	10015150	Erythema
Swelling	10053425	Injection site swelling
Fever	10016558	Pyrexia
Headache	10019211	Headache
Fatigue	10016256	Fatigue
Myalgia	10028411	Myalgia
Arthralgia	10003239	Arthralgia

5.4.5.1. Concomitant Medication

Medications will be coded using the GSKDRUG dictionary.

5.4.6. Sensitivity Analyses

If more than 10% of participants miss the revaccination dose or don't have safety data reported after revaccination, the safety analysis described in section 5.4.4 will also be conducted on participants with full follow-up (attending all study visits).

The goal of this sensitivity analysis is to determine the extent to which results might be impacted by considering the different scenarios.

6. ANALYSIS INTERPRETATION

All analyses are descriptive.

7. INTERIM ANALYSES

7.1. Sequence of analyses

Analyses to evaluate objectives and endpoints will be performed stepwise:

- **Month 6:** A first analysis* will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 6 are available for all participants. This analysis will be considered as final for those endpoints.

  *Note: the analysis at Month 6 may be merged with the analysis at the subsequent timepoint.

- **Month 13**: A second analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 12 are available for all participants, as well as data up to 1 month after the first revaccination (Month 13) in the RSV_annual group. This analysis will be considered as final for those endpoints.
- **Month 18**: A third analysis will be performed on all safety and/or immunogenicity data available and as clean as possible, when data for secondary endpoints up to Month 18 are available for all participants. This analysis will be considered as final for those endpoints. Safety and immunogenicity analysis may be performed at different time depending on data availability.
- **Month 25**: A fourth analysis will be performed on all reactogenicity, safety and immunogenicity data available and as clean as possible, when data for at least primary and secondary endpoints up to Month 24 are available for all participants, as well as data up to 1 month after the second revaccination (Month 25) in the RSV_annual group. This analysis will be considered as final for those endpoints.
- The final **End of Study** analysis will be performed when all reactogenicity, safety and immunogenicity data for at least primary and secondary endpoints up to study conclusion are available for all participants (Month 36).
- If the data for tertiary endpoints become available at a later stage, (an) additional analysis/analyses will be performed.

Description	Analysis ID	Disclosure Purpose (CTRS=public posting, SR=study report, internal)	Reference for TFL TOC
End of study analysis	E1_01	CTRS, SR	See column 5 in TFL TOC
Analysis up to Month 6	E1_02	CTRS, SR	See column 5 in TFL TOC
Analysis up to Month 13	E1_03	CTRS, SR	See column 5 in TFL TOC
Analysis up to Month 18	E1_04	Internal	See column 5 in TFL TOC
Analysis up to Month 25	E1_05	Internal	See column 5 in TFL TOC

7.2. Statistical considerations for interim analyses

All analyses will be conducted on final data (as clean as possible) and therefore no statistical adjustment for interim analyses is required.

8. CHANGES TO THE PROTOCOL DEFINED STATISTICAL ANALYSIS

Not Applicable

9. NON-STANDARD DATA DERIVATION RULES AND STATISTICAL METHODS

The following sections describe additional derivation rules and statistical methods which are not presented in section 10.1 (Business rules for standard data derivations and statistical methods).

9.1. Handling of missing data

9.1.1. Dates

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

- Adverse event start dates with missing day:
  - If the month is not the same as the vaccine dose, then the imputed start date will be the 1st of the month
  - If the event starts in the same month as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the vaccine dose given during that month. If 'before vaccination' is selected, the imputed date will be one day before the vaccine dose given during that month.
- Adverse event start dates with missing day and month:
  - If the year is not the same as the vaccine dose, then the imputed start date will be the 1st of January.
  - If the event starts in the same year as the vaccine dose, the flag indicating if the event occurred before or after vaccination (AE.AESTRTPT) will be used to complete the date. If 'after vaccination' is selected, the imputed start date will match the first vaccine dose given during that year. If 'before vaccination' is selected, the imputed date will be one day before the first vaccine dose given during that year.
- Adverse event end dates with missing day: the imputed end date will be the last day of the month (30 or 31) or the study conclusion date whichever comes first.
- Adverse event end dates with missing day and month: the imputed end date will be the last day of the year (31st of December) or the study conclusion date whichever comes first.

All incomplete concomitant medication/vaccination start/end date will follow the rules above.

10. ANNEXES

10.1. Business rules for standard data derivations and statistical methods

This section contains standard rules for data display and derivation for clinical and epidemiological studies.

10.1.1. 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 vaccination is selected, the relative dose for the event will be the one administered on the start day of the event. If 'before vaccination is selected, the relative dose for the event will be the dose prior to this one.

10.1.2. Handling of missing data

10.1.2.1. 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.

See also exceptions in section 9.1.

10.1.2.2. Laboratory data

Missing laboratory results (including immunological data) will not be replaced.

10.1.2.3. Daily recording of solicited events

10.1.2.3.1. Studies with paper diaries

For studies using paper diaries which have questions in the CRF indicating the presence or absence of solicited events, the following rules are applicable:

- Denominators for the summary of administration site (or systemic) solicited events will be calculated using the number of participants who respond "Yes" or "No" to the question concerning the occurrence of administration site (or systemic) events.
- When a specific solicited event is marked as having not occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=N for the specified post-dose period for the event in question), all daily measurements will be imputed as Grade 0.
- When a specific solicited event is marked as having occurred following a specific study dose (i.e. SDTM CE.CEOCCUR=Y for the specified post-dose period for the event in question), any missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.

- When the occurrence of a specific solicited event is not present (i.e. SDTM CE.CEOCCUR is neither Y nor N for the specified post-dose period for the event in question) but the group of solicited events (administration site or systemic) is marked as having occurred (i.e. SDTM CE.CEOCCUR=Y), all missing daily recordings will be given imputed values to allow them to contribute to the 'Any' rows but not to specific grade rows of the solicited event summary tables.

The following table shows how participants contribute to each category for a specific solicited event over the Day X to Day Y post-dose period:

Solicited event category	Participants included in the calculation of the numerator
Any	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y or with the adverse event marked as present and at least one missing daily recording between Day X and Day Y
At least grade 1	All participants with at least one occurrence of the adverse event at grade 1, grade 2, or grade 3 between Day X and Day Y
At least grade 2	All participants with at least one occurrence of the adverse event at grade 2 or grade 3 between Day X and Day Y
At least grade 3	All participants with at least one occurrence of the adverse event at grade 3 between Day X and Day Y

10.1.2.4. 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.

10.1.3. Data derivation

10.1.3.1. 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

In this study, we will collect only the year of birth. The rules for handling missing day and/or month in the DOB are given in section 10.1.2.1.

10.1.3.2. 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

10.1.3.3. 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 and value is <=ULOQ	value
"value" and value is > ULOQ	ULOQ*
All other cases	Missing

*This rule will be used to compute descriptive statistics (GMTs, fold increase,etc). All values might be displayed in scatter plots with individual results.

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

GMT or 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 10.1.3.3 for the purpose of GMT/GMC calculation. Cut-off values are defined by the laboratory before the analysis.

10.1.3.5. 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).

10.1.3.6. 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 3 March 2018 and ends on 12 March 2018 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.

10.1.3.7. Counting rules for combining solicited and unsolicited adverse events

For output combining solicited and unsolicited adverse events, all serious adverse events will be considered systemic events since the administration site flag is not included in the expedited adverse event CRF pages. Unsolicited adverse events with missing administration site flag will also be considered systemic.

Multiple events with the same preferred term which start on the same day are counted as only one occurrence.

10.1.3.8. 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.

10.1.4. Display of decimals

10.1.4.1. Percentages

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

10.1.4.2. Demographic/baseline characteristics statistics

The mean, median, and standard deviation for continuous baseline characteristics 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 maximum and minimum of transformed body temperatures will be displayed with one decimal.

10.1.4.3. Serological summary statistics

For each assay, geometric mean titers (GMT) or concentrations (GMC) and their confidence limits will be presented with one decimal, as well as GMT/GMC fold increase from pre-dose.

The mean, median, standard deviation and quartile values for frequency of RSVPreF3 specific-CD4+ and CD8+ T cells and for the fold increase (post over pre-vaccination) will be presented with one decimal. The minimum and maximum values will be presented with no decimal.

10.1.5. Statistical methodology

10.1.5.1. Exact confidence intervals around proportions

The exact confidence intervals around within-group proportions are derived using the method of Clopper and Pearson [Clopper, 1934].

11. 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.

12. REFERENCES

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

David MP, Van Herck K, Hardt K, Tibaldi F, Dubin G, Descamps D et al. Long-term persistence of anti-HPV-16 and -18 antibodies induced by vaccination with the AS04-adjuvanted cervical cancer vaccine: modeling of sustained antibody responses. Gynecol Oncol 2009; 115 (Suppl 3):S1-6.